Diss. ETH No. 18402

Total Syntheses of Anguinomycins C & D, Synthetic Studies on Sporolides and Preparation of Eudistomin Derivatives: Biological Evaluation Against Cancer and Malaria

A dissertation submitted to the

SWISS FEDERAL INSTITUTE OF TECHNOLOGY

ETH ZURICH

for the degree of

Doctor of Sciences

Presented by

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Zürich 2009

Acknowledgements

I am grateful to Prof. Dr. *Karl Gademann* for the possibility to work under his supervision during my PhD. I would like to thank him for his confidence and the freedom let to me during my research as well as for his enthusiasm, help, advice and efforts whenever I need him. I really appreciated the interesting discussions during theses years and I have to thank him also for teaching me that in chemistry patience and perseverance are fundamentals, especially when the chemistry failed. I am thankful for the interesting discussion during the group meeting and for the learning experience during his lectures.

I am thankful to Prof. Dr. *Erick M. Carreira* for having given me the possibility to join his group and benefit of the very enriching discussions during the group seminars. I would like to thank him for his help and advice whenever I need him.

I would like to thank Prof. Dr. *Karl-Heinz Altmann* for accepting the co-examination of my thesis.

Prof. Dr. *Karl Gademann*, Prof. Dr. *Erick M. Carreira*, Prof. Dr. *Karl-Heinz Altmann*, Dr. *Tom Woods* and *Jean-Yves Wach* are acknowledged for critically proofreading and thereby substantially improving this manuscript.

A special thanks to Dr. *Corey Stephenson, Christian Defieber*, Dr. *Thomas Fessard, Damien Barbaras, Florian Kleinbeck*, Dr. *Jose Carlos Gonzalez-Gomez, Stefan Reber* and Dr. *Isabelle Lyothier* at the ETHZ as well as to *Jean-Yves Wach, Massimo Binaghi, Adrien Lawrence, Cyril Portmann*, Dr. *Tom Woods*, Dr. *Henning Jessen*, Dr. *Joanna Kobylinska*, *Cindy Fellay*, *Filippo De Simone*, Prof. Dr. *Jerôme Waser*, Prof. Dr. *Pierre Vogel* at the EPFL for the useful discussions on chemistry problems.

I would like to thanks *Jean-Yves Wach*, *Massimo Binaghi* and *Cindy Fellay* as team co-workers for their contribution to the progress of the sporolides project.

Special thanks for nice moments at the ETHZ to Dr. *Corey Stephenson*, *Christian Defieber*, Dr. *Thomas Fessard*, *Damien Barbaras*, Dr. *Jose-Carlos Gonzalez-Gomez*, Dr. *Andrew Waltman*, Dr. *Shinji Fujimori*, *Florian Kleinbeck*, Dr. *Yann Bethuel* and *Stefan Reber* and a big thanks to all Carreira's group members for the friendship.

Special thanks for nice moments at the EPFL to *Massimo Binaghi*, *Filippo De Simone*, *Damien Barbaras*, *Jean-Yves Wach*, *Adrien Lawrence*, *Cyril Portmann*, Dr. *Tom Woods*, Dr. *Henning Jessen*, Dr. *Joanna Kobylinska*, *Cindy Fellay*, Dr. *Chandan Kumar Jana*, Prof. Dr. *Jerôme Waser*, *Davinia Fernandez-Gonzalez*, *Riccardo Torosantucci* and a big thanks to all Gademann's and Waser's group members for the friendship.

I am thankful to Prof. Dr. *Ulrike Kutay*, Dr. *Stephan Güttinger* and *Ivo Zemp* for biological evaluation of anguinomycins and derivatives and to Prof. Dr. *Reto Brun*, Dr. *Marcel Kaiser* for biological evaluation of nostocarboline and derivatives.

For the administrative help as well as other helpful advices I would like to thank *Franziska Peyer*, *Brown Vida* and *Monique Borcard*.

Many thanks go also to the collaborators of the technical staff at the ETHZ and the EPFL for their services and precious contribution in the analyses.

Ringrazio parenti, amici e tutti coloro che mi hanno aiutato durante questi anni. Un ringraziamento speciale a *Vanessa* per il sostegno, l'incoraggiamento e per i momenti di felicità trascorsi assieme.

Infine, ringrazio soprattutto i miei genitori e le mie sorelle, *Katia* e *Diana*, per aver condiviso con me bei momenti, come per avermi sostenuto e incoraggiato durante quelli più difficili. Vi ringrazio per il vostro aiuto e per aver sempre creduto in me.

Alla mia famiglia
con affetto e gratitudine

Publications & Presentations

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1) Z. Grote, S. Bonazzi, R. Scopelliti and K. Severin
Bridged Dihydroxypyridine Ligands for the Synthesis of Expanded Helicates
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2) K. Gademann and S. Bonazzi
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3) S. Bonazzi, S. Güttinger, I. Zemp, U. Kutay and K. Gademann
Total Synthesis, Configuration, and Biological Evaluation of Anguinomycin C
Angew. Chem. Int. Ed. **2007**, *46,* 8707-8710; *Angew. Chem.* **2007**, *119,* 8862-8865.

4) J.-Y. Wach, S. Bonazzi and K. Gademann
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6) K. Gademann, D. Barbaras, S. Bonazzi, L. Patiny, R. Scopelliti, P. Schneider, S. T. Cole, M. Kaiser and R. Brun
Antimalarial and Antitubercular Nostocarboline Derivatives: Synthesis, *in vitro* and *in vivo* Biological Evaluation
Chem. Med. Chem. **2009**, *submitted.*

7) S. Bonazzi, R. Scopelliti, M. Kaiser, R. Brun and K. Gademann
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Anguinomycins C & D and Derivatives: Total Syntheses and Biological Evaluation on CRM1-Mediated Nucleocytoplasmic Transport
J. Am. Chem. Soc. **2009**, *in preparation.*

9) S. Bonazzi, M. Binaghi, J.-Y. Wach, C. Fellay and K. Gademann
Synthetic Studies on the Sporolides: Exploration of the Enediyne Route
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J.-Y. Wach, S. Bonazzi and K. Gademann
Combining Devices and Drugs by Synthetic Natural Product Hybrids
European Cells & Materials Journal **2007**, *14*, 127.

Oral Presentations

First Total Synthesis of Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi and K. Gademann.
CUSO Summer school - Target Synthesis: Challenges, Strategies and Methods, September 2nd – 6th 2007, Villars.

Total Synthesis, Configuration and Biological Evaluation of Anguinomycin C
S. Bonazzi, S. Güttinger, U. Kutay, I. Zemp and K. Gademann.
SCS Fall Meeting, September 11th 2008, Zurich.

Poster Presentations

Synthetic Studies on Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi, S. Güttinger, U. Kutay, I. Zemp and K. Gademann
SCS Fall Meeting 2006, October 13th, Zurich.

First Total Synthesis of Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi and K. Gademann
1st Japanese-Swiss symposium on chemical biology, June 25th- 26th 2007, Lausanne.

First Total Synthesis of Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi and K. Gademann
8th Tetrahedron symposium - Challenges in Organic Chemistry (50th anniversary of Tetrahedron), June 26th – 29th 2007, Berlin.

Combining Devices and Drugs by Synthetic Natural Product Hybrids
J.-Y. Wach, S. Bonazzi and K. Gademann
BIOSURF VII - Functional Interfaces for Directing Biological Response, August 28th – 31th 2007, Zurich.

First Total Synthesis of Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi and K. Gademann
CUSO Summer school - Target Synthesis: Challenges, Strategies and Methods, September 2nd – 6th 2007, Villars.

First Total Synthesis of Anguinomycin C: a Tumor-Selective Polyketide
S. Bonazzi, S. Güttinger, U. Kutay, I. Zemp and K. Gademann
SCS Fall Meeting 2007, September 12th, Lausanne.

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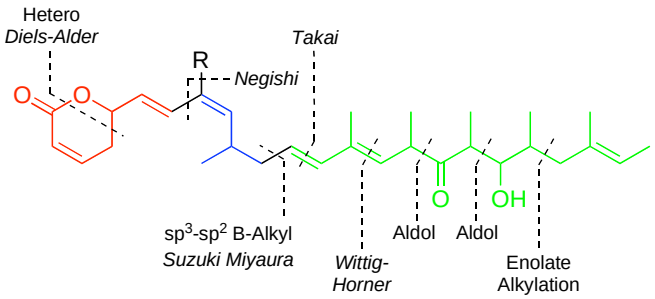
Abstract

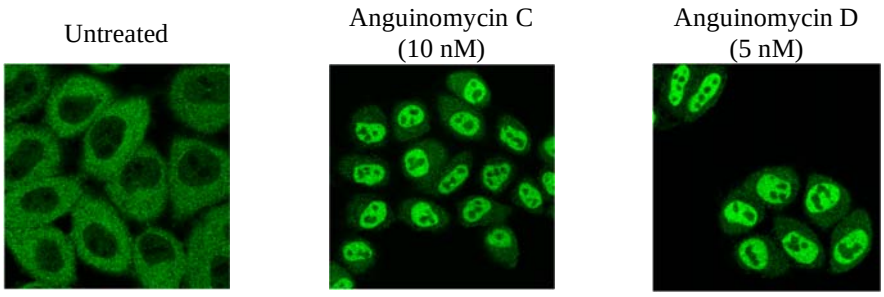
Natural products continue to play a central role in drug discovery and synthetic organic chemists inspire themselves from nature for the development of new strategies and for the preparation of highly complex biologically active structures for treating human diseases. Anguinomycins C and D are antitumor antibiotics belonging to the leptomycin family that were isolated from *Streptomyces* microorganisms in 1995. These compounds selectively target retinoblastoma tumor suppressor protein (pRb) inactivated cancer cells and induce only growth arrest on normal cells. The absolute configuration of these compounds was previously unknown. In this thesis the first total syntheses of anguinomycins C and D is presented as well as the preparation of their derivatives and their biological evaluation. The preparation of the lactone moiety was characterized by Cr-catalyzed hetero-*Diels-Alde*r reaction. The central part deriving from the Roche ester was coupled to the lactone fragment *via* a tandem hydrozirconation-Negishi cross-coupling reaction and the residue (R) installed using the *Negishi* cross-coupling reactions with stereoinversion. The polyketide chain was characterized by alkylation and aldol reactions using the *Seebach* modification of the *Evans* auxiliary. *Wittig* reaction and *Takai* olefination afforded the polyketide fragment, which was coupled *via* sp^3-sp^2 *Suzuki* cross-coupling to give the skeleton of the two targets. Final modification furnished anguinomycins C and D in 29 steps (longest linear sequence 19 steps) with overall yields of 6.7 and 6.0% respectively.



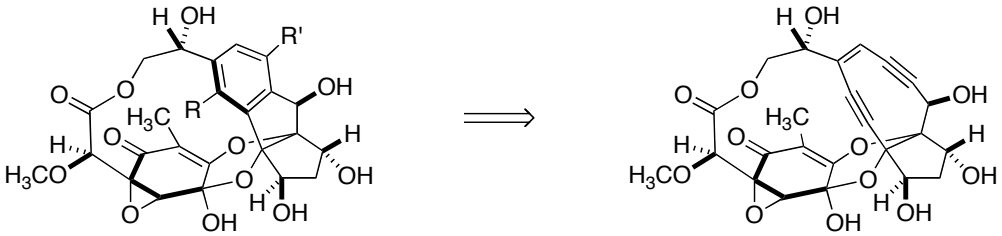
Anguinomycins C (R = CH_3) & D (R = C_2H_5)

Anguinomycins C and D were submitted to biological evaluation as CRM1-mediated nucleocytoplasmic transport inhibitor and both compounds confirmed their high activity inducing inhibition at 10 and 5 nM respectively. In addition, derivatives were prepared in order to investigate the mode of action and the structure-activity

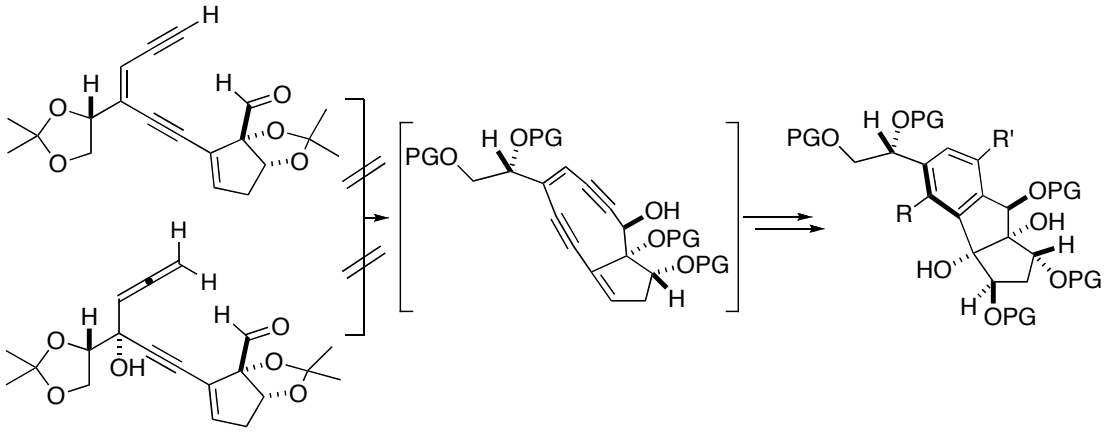
relationship. New analogs, which displayed high activity, were identified forming the basis for the development of more powerful and selective nucleocytoplasmic transport inhibitors for cancer treatment.



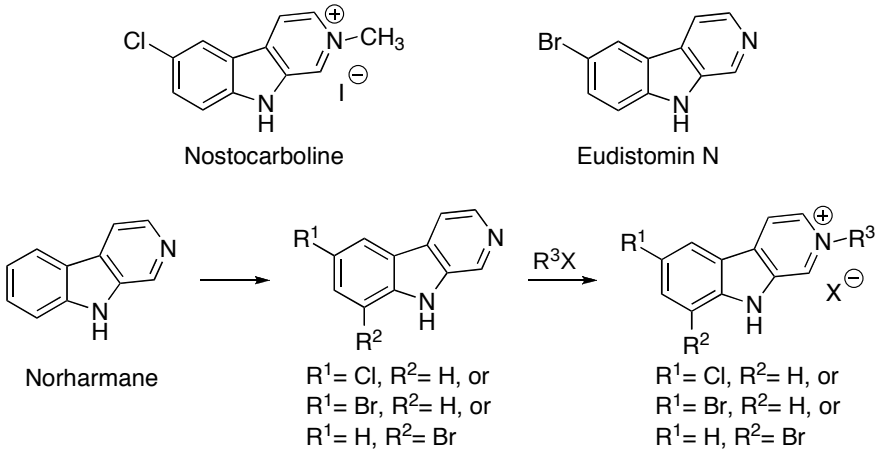
Sporolides A and B, which are proposed to derive from the *Bergmann* cyclization of an endiyne precursor, are complex marine natural products isolated from *Salinospora tropica* in 2005. These compounds did not show interesting biological activity, probably because the active substrate is the enediyne precursor prior to cyclization. Their unusual architecture displaying 22 out of 24 carbons sp^2 hybridized or oxygenated, 7 rings and 10 stereogenic centers makes them challenging targets for total synthesis.



Synthetic studies for the development of a biomimetic approach to the chlorinated cyclopenta[*a*]indene ring are presented. Preparation of the 9-membered enediyne ring from both an enediyne and a diyne precursor was investigated. The chemistry was characterized by *Morita-Baylis-Hillman* reaction, *Sharpless* asymmetric dihydroxylation, enediyne formation *via Wittig* reaction and $CeCl_3$•2LiCl-mediated acetylide addition. Although preliminary attempts to form the 9-membered enediyne core structure were unsuccessful, investigation are ongoing.



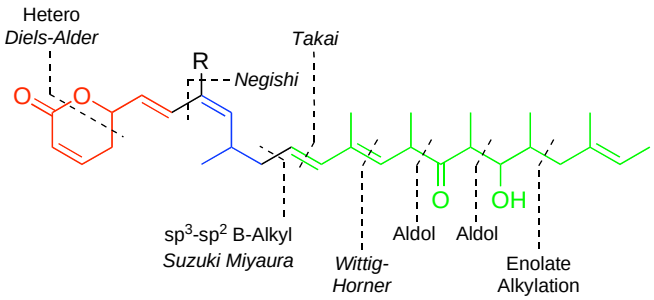
Malaria remains a huge problem in developing countries and its parasite affects 300-500 million people causing 1-3 million of deaths each year. In 2005 *Gademann* and co-workers isolated nostocarboline from freshwater cyanobacterium *Nostoc* 78-12A. This β-carbolinium ion displayed antimalarial activity against *Plasmodium falciparum* with an IC_{50} of 194 nM and good selectivity being more than 600 times less toxic against L6 rat myoblast cell line. It was decided to prepare beta-carbolinium ion derivatives of nostocarboline and eudistomin N for biological evaluation against malaria. The compounds were prepared following a straightforward procedure based on halogenation and *N*-alkylation of the common starting material norharmane.



In vitro biological evaluation against *Plasmodium falciparum* identified five compounds with interesting activity and selectivity. Between them two new 6-bromo-9H-carbolinium ion displaying IC_{50} of 18 and 32 nM with a selectivity against L6 rat myoblast cell line of 4783 and 2443 respectively. The five products were selected for biological evaluation *in vivo* in a *P. berghei* mouse model and biological assays are currently ongoing.

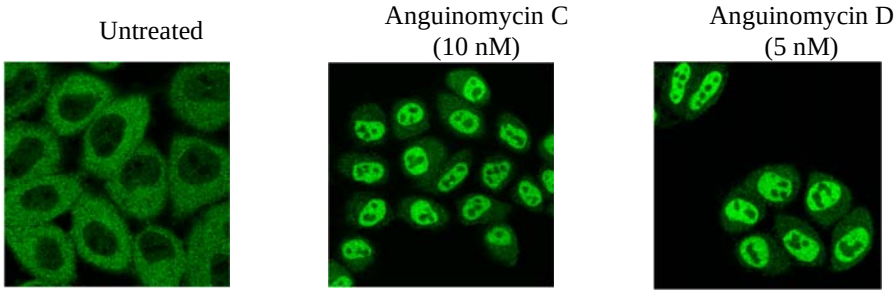
Riassunto

I prodotti naturali continuano a ricoprire un ruolo fondamentale nella scoperta di nuovi farmaci. Il chimico organico s'ispira costantemente alla natura per progettare e sintetizzare complesse strutture d'interesse biologico. Le anguinomicine C e D, la cui configurazione assoluta era finora sconosciuta, sono antibiotici antitumorali appartenenti alla famiglia delle leptomicine e sono state isolate nel 1995 da microorganismi del genere *Streptomyces*. Queste molecole hanno dimostrato un'attività selettiva verso le cellule neoplastiche nelle quali la proteina del retinoblastoma (pRb), un soppressore tumorale, è disattivata. Le cellule sane subiscono al contempo solamente un arresto temporaneo della crescita. In questa tesi saranno presentate le prime sintesi totali delle anguinomicine C e D, la sintesi di derivati e la loro valutazione biologica. Il lattone è stato sintetizzato utilizzando una reazione etero-*Diels-Alder* catalizzata dal cromo. Il frammento centrale, derivante dall'estere di Roche, è stato condensato con l'intermediario lattonico tramite una sequenza d'idrozirconazione seguita da *Negishi* cross-coupling. Il sostituente (R) è stato introdotto attraverso un *Negishi* cross-coupling con stereoinversione della configurazione. La sintesi della catena polichetidica è caratterizzata da un'alchilazione seguita da due condesazioni aldoliche utilizzando la variante di *Seebach* dell'ausiliare chirale di *Evans*. Una successiva reazione di *Wittig* seguita da un olefinazione di *Takai* ha fornito la catena polichetidica, la qual è stata condensata mediante sp^3-sp^2 *Suzuki cross-coupling* formando lo scheletro delle due molecole target. Quattro ulteriori passaggi hanno permesso di ottenere le angiunomicine C e D in un numero complessivo di 29 tappe (sequenza lineare più lunga 19 tappe) e una resa globale rispettivamente di 6.7 e 6.0%.

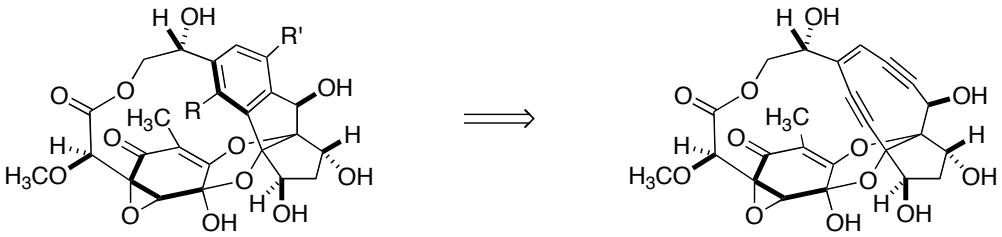


Anguinomycins C (R = CH_3) & D (R = C_2H_5)

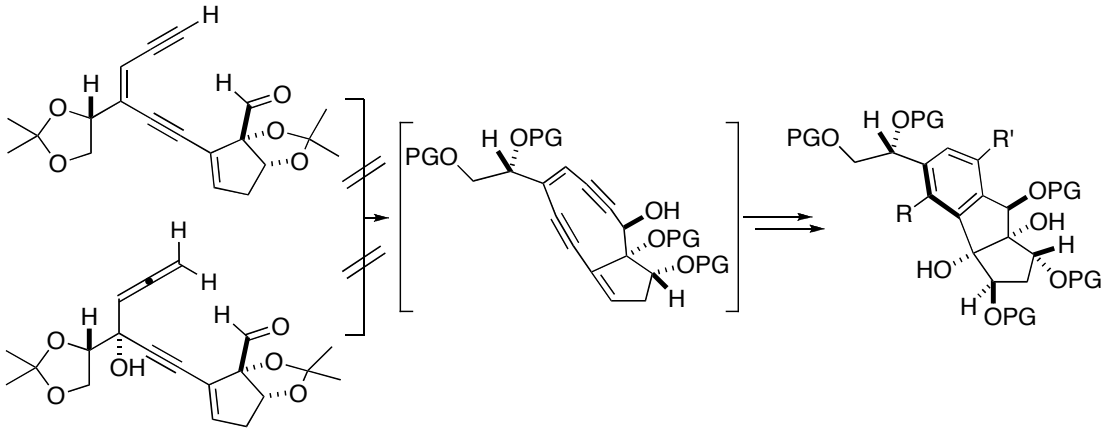
I risultati biologici hanno dimostrato che le anguinomicine C e D agiscono come inibitori del trasporto nucleocitoplasmatico mediato dalla proteina CRM1 a concentrazioni rispettivamente di 10 e 5 nM. Sono stati inoltre sintetizzati alcuni derivati al fine di indagare sul meccanismo d'azione e sulla correlazione struttura-attività. Alcuni derivati hanno mostrato un'elevata attività biologica, fornendo le basi per lo sviluppo di più potenti e selettivi inibitori del trasporto nucleocitplasmatico per la cura del cancro.



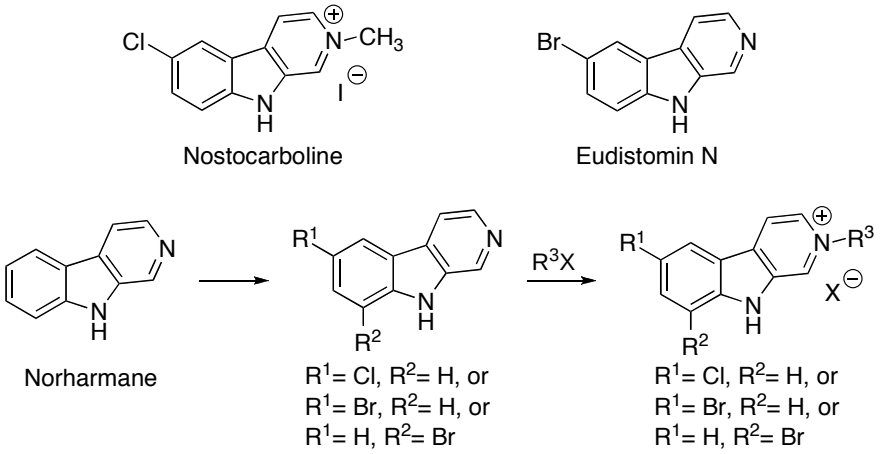
Gli sporolidi A e B sono dei complessi prodotti naturali di origine marina isolati nel 2005 dalla *Salinospora tropica* e sembrerebbero derivare dalla ciclizzazione di *Bergman* di un precursore enediinico. Entrambe le strutture non hanno mostrato alcun'attività biologica, probabilmente perchè la molecola attiva era il loro precursore enediinico. L'inusuale architettura di questi due prodotti che mostrano 22 dei 24 atomi di carbonio ibridati sp^2 o ossigenati, 7 cicli e 10 centri stereogenici li rendono degli obbiettivi stimolanti per la sintesi totale.



In questo lavoro sono presentati degli studi sintetici mirati allo sviluppo di un approccio biomimetico dell'anello clorato ciclopenta[*a*]indene. È stata investigata la preparazione dell'anello enediinico a 9 termini sia a partire da un precursore enediinico che da uno diinico. La strategia è caratterizzata dalla reazione di *Morita-Baylis-Hillman*, da una diidrossilazione di *Sharpless*, dalla formazione dell'endiino tramite reazione di *Wittig* e da un'addizione d'actilide mediata da $CeCl_3$•2LiCl. Sebbene i primi tentativi non abbiano portato alla formazione del ciclo a 9 termini, ulteriori studi sono tutt'ora in corso.



La malaria rappresenta tutt'oggi una piaga per i paesi in via di sviluppo e il suo parassita infetta 300-500 milioni di persone causandone la morte di 1-3 milioni ogni anno. Nel 2005, *Gademann* e collaboratori hanno isolato la nostocarbolina dal cianobatterio d'acqua dolce *Nostoc* 78-12A. La nostocarbolina ha mostrato attività antimalariche contro il *Plasmodium falciparum* con un IC_{50} di 194 nM e un'elevata selettività, risultando 600 volte meno tossica nei confronti delle cellule mioblastiche di ratto L6. In questo progetto si è deciso di preparare ioni β-carbolinici derivati dalla nostocarbolina e dall'eudistomina N per sottoporli a test biologici. I prodotti sono stati preparati seguendo una rapida sequenza basata sull'alogenazione e la *N*-alchilazione del norharmane.



I test biologici hanno identificato cinque composti con un'interessante attività e selettività contro il *Plasmodium falciparum*. Due di questi ioni 6-bromo-9H-carbolinio hanno mostrato una IC_{50} di 18 e 32 nM risultando rispettivamente 4783 e 2443 volte meno tossici nei confronti delle cellule mioblastiche di ratto L6. I cinque prodotti sono stati selezionati per test biologici *in vivo* sui topi infetti da *P. berghei* e gli esami sono tuttora in corso.

List of Abbreviations, Acronyms and Symbols

$[\alpha]^{T}_{D}$	specific rotation at temperature T at the sodium D line
Ac	acetyl
aq	aqueous
br	broad
Bu	butyl
°C	degrees centigrade
c	concentration
calcd	calculated
cat.	catalytic
CAM	ceric ammonium molybdate
Cp	cyclopentadienyl
CRM1	chromosome maintenance region 1 or exportin 1
CSA	camphorsulfonic acid
δ	NMR chemical shift in ppm downfield from standard TMS
d	doublet
DBU	1,8-diazabicyclo[5.4.0]undec-7-ene
DCC	Dicyclohexylcarbodiimide
DDQ	2,3-dichloro-5,6-dicyano-1,4-benzoquinone
DIBAL-H	diisobutylaluminium hydride
DMAP	4-*N*,*N*-dimethylaminopyridine
DMF	*N*,*N*-dimethylformamide
DMP	*Dess-Martin* periodinane
DMSO	dimethyl sulfoxide
d.r.	diastereomeric ratio
e.e.	enantiomeric excess
EI	electron impact ionization
eq.	equation
equiv	equivalent(s)
Et	ethyl
EtOAc	ethyl acetate
FC	flash chromatography

g	gram
GC	gas chromatography
h	hour(s)
Hz	hertz (s^{-1})
i	iso
J	coupling constant
KHMDS	potassium bis(trimetylsilyl)amide
L	liter
LDA	lithium diisopropylamide
LHMDS	lithium bis(trimetylsilyl)amide
LMB	leptomycin B
LR	low resolution
m	multiplet
M	molarity ($mol.L^{-1}$)
Me	methyl
MeOH	methanol
mg	milligram
min	minute(s)
mL	milliliter
μL	microliter
mmol	millimol
MS	mass spectroscopy
ν	frequency (cm^{-1})
n.d.	not determined
NES	nuclear export signal
NMR	nuclear magnetic resonance
p	para
PCC	pyridinium chlorochromate
PDC	pyridinium dichromate
Ph	phenyl
PIFA	phenyliodine(III) bis(trifluoroacetate)
PMB	4-methoxybenzyl
ppm	parts per million
PPTS	pyridinium 4-toluenesulfonate

Pr	propyl
pRb	protein retinoblastoma
q	quartet
quint.	quintet
quant.	quantitative
R_f	retention factor
RT	room temperature
s	singlet
sext.	sextet
sept.	septet
t	triplet
T	temperature
TBAF	tetra-*n*-butylammonium fluoride
TBDPS	*tert*-butyldiphenylsilyl
TBS	*tert*-buthyldimethylsilyl
TES	triethylsilyl
THF	tetrahydrofuran
TIPS	triisopropylsilyl
TLC	thin layer chromatography
TMS	trimethylsilyl

1. Introduction: Natural Products and Drug Discovery

The adaptation of life to different environments over time and the biodiversity among ecosystems has resulted in the generation of a vast array of natural compounds and consequently, an almost endless source of inspiration for synthetic organic chemists. Nature has followed an evolutionary selection process for millions of years and the structure of natural products is well selected in such a way to give a desired biological activity.[1] The investigation of natural sources forms the basis for the discovery of new biologically active compounds and recent reports have shown that pharmaceuticals of natural origin or derivatives represent more than 50% of the drugs on the market.[2] Natural products are found everywhere and their research is not only limited to terrestrial organisms; seventy percent of the earth's surface is covered in water and the marine ecosystem also represent an important and interesting resource of new chemical structures.[3]

Natural products and their sources have for a long time been recognized and employed by man to treat human diseases. The earliest records of the use of plants and herbs in medicine originated in Egypt and Mesopotamia and dated from 2900 BC and from 2600 BC respectively.[4] Today, natural products remain an important source for the discovery and development of biologically active compounds with therapeutic effects. Although, in recent years the investigation of natural products as a source of chemotherapeutic agents has declined in favor of new drug discovery approaches such as combinatorial chemistry or computer based molecular modeling.[5] However, 25 years of drug research using combinatorial chemistry resulted in only one new chemical entity being discovered and approved for drug use using this method.[2b] The failure of this modern approach shows how difficult it is to randomly generate potent and selective compounds and this method has been commented on by *Danishefsky*: *"a small collection of smart compounds may be more valuable than a much larger*

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[2] a) D. J. Newman, G. M. Cragg, K. M. Snader, *J. Nat. Prod.* **2003**, *66*, 1022-1037; b) D. J. Newman, G. M. Cragg, *J. Nat. Prod.* **2007**, *70*, 461-477.

[3] W. Fenical, P. R. Jensen, *Nat. Chem. Biol.* **2006**, *2*, 666-673.

[4] D. J. Newman, G. M. Cragg, K. M. Snader, *Nat. Prod. Rep.* **2000**, *17*, 215-234.

[5] G. M. Cragg, D. J. Newman, K. M. Snader, *J. Nat. Prod.* **1997**, *60*, 52-60.

hodgepodge collection mindlessly assembled".[6] Although natural products remain fundamental to drug development, there are also some limitations due to the amount of compounds that can be isolated. Often, the amount of isolated material is extremely low and not sufficient for complete characterization or for biological studies. Organic synthesis is a powerful method allowing the exploration of underinvestigated compounds and synthetic organic chemists, inspired by nature, have developed methods and strategies in order to recreate the target molecules in laboratory. The synthetic preparation of compounds allows the elucidation of structures and further biological investigations in order to understand targets, metabolism and mode of action of the selected compounds. Once the target and the active part of the molecule are identified, it is possible to prepare derivatives or simplify the structure and study their activity. This forms the basis of what will be presented in this thesis, with the synthesis and biological studies of bioactive compounds against cancer and malaria.

[6] S. Borman, *Chem. Eng. News* **2002**, *80*, 23-24.

2. Total Syntheses and Biological Evaluation of Anguinomycins C & D

2.1. Natural Products for Cancer Treatment

Cancer remains a major disease worldwide and for many of its forms there is no definitive treatment available. The development of effective new drugs to treat cancers is currently a challenging goal in drug discovery and clinical therapy and the search for new, potent and selective pharmaceuticals has proven to be particularly difficult.[7] The development of new anticancer agents relies heavily on natural products; in fact 60% of the antitumoral compounds on the market today have natural origin.[2b] Among the anticancer agents currently in use there are the paclitaxels (Taxol® (**I**) and Taxotere®), the vinca alkaloids (**II**) and camptothecin (**III**) (Figure 1).[8] Recently, other classes of compounds such as the epothilones[9] have shown promise in the battle against cancer and one of its derivatives (Ixempra® (**IV**) or ixabepilone) was approved in 2007 by the FDA for the treatment of breast cancer.[10] Anther compound, the hybrid antibody-calicheamicin conjugate (Mylotarg®) (**V**) has been also approved for the treatment of acute myeloid leukemia (Figure 1).[11] In addition to natural product research, organic synthesis has also made important contribution to the discovery of new anticancer agents by allowing the preparation of small molecule natural products in the laboratory.[12]

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7 A. Kamb, S. Wee, C. Lengauer, *Nat. Rev. Drug Discovery* **2007**, *6*, 115-120.

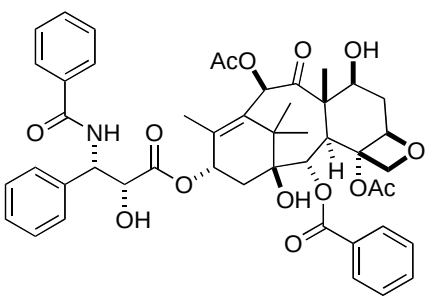
8 F. Guéritte, J. Fahy, in *Anticancer Agents from Natural Products* (Eds.: G. M. Cragg, D. G. I. Kingston, D. J. Newman), Eds. Taylor & Francis, Boca Raton London New York Singapore, **2005**, pp. 123-135.

9 K. H. Altmann, J. Gertsch, *Nat. Prod. Rep.* **2007**, *24*, 327-357.

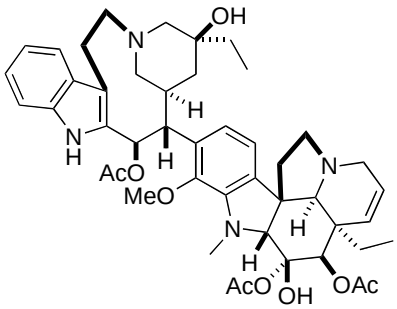
10 http://www.medicalnewstoday.com/articles/85726.php (retrieved Feb., 3th, 2009).

11 a) http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm?fuseaction=Search.DrugDetails (retrieved Feb., 3th, 2009); b) R. V. J. Chari, *Acc. Chem. Res.* **2008**, *41*, 98-107.

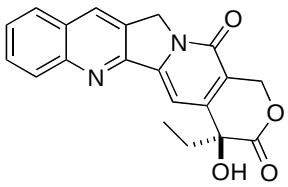
12 R. M. Wilson, S. J. Danishefsky, *Chem. Soc. Rev.* **2007**, *36*, 1207-1226.



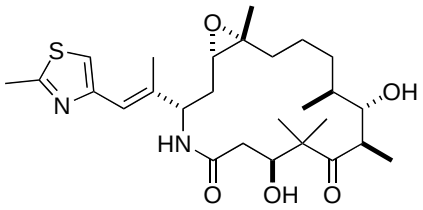
Paclitaxel (Taxol®) (**I**)



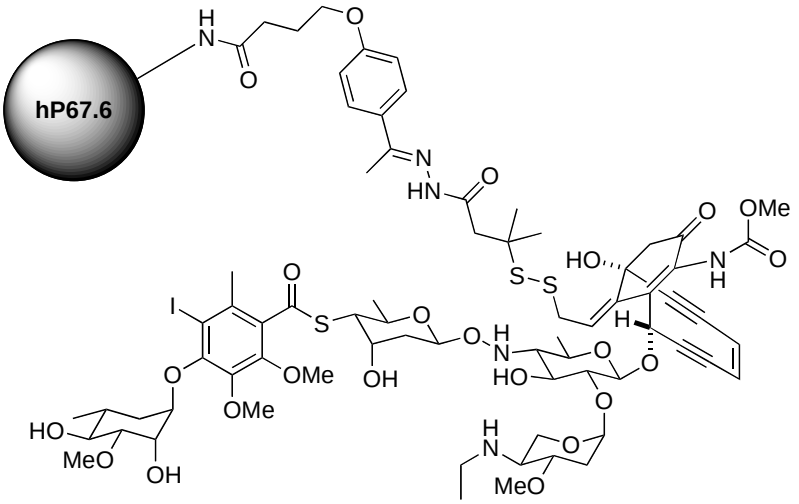
Vinca alkaloid (**II**)



Camptothecin (**III**)



Ixabepilone (Ixempra®) (**IV**)



hP67.6 Antibody-calicheamicin conjugate (Mylotarg®) (**V**)

Figure 1: Antitumoral compounds from natural origin on the market.

Interesting molecule targets for cancer research that are based on natural compounds are small molecules that can interfere in the cell cycle. The cell cycle regulates cellular proliferation, and malfunctioning during the cell cycle is the basis for cancer development.[13] Cells possess a control system that repairs mistakes that occur during cell replication and in the case of irreparable problems cell apoptosis is induced.[14] Studies have confirmed that a major problem common to several cancers is

[13] D. T. Hung, T. F. Jamison, S. L. Schreiber, *Chem. Biol.* **1996**, *3*, 623-639.

[14] A. J. Levine, *Annu. Rev. Biochem.* **1993**, *62*, 623-651.

that during cancer cell proliferation the control systems are often deactivated or modified and uncontrolled replication starts.[15] At this point, bioactive agents able to interact in the cell cycle to stop this uncontrolled replication are required. The cell cycle is divided in four phases and many anticancer agents can selectively act in one of them following specific modes of action.[13] Examples of some different compounds, which display anticancer properties, will be treated in more detail in the following sections.

2.2. The Leptomycin Family

2.2.1. Overview

The leptomycin family is a class of polyketides, which display potent anticancer activity.[16] The first members discovered were leptomycin A and B (LMB) (**VI**) (Figure 2), which were isolated from a *Streptomyces* strain in 1983.[17] After a first classification as antifungal compounds,[18] their potent antitumoral activity was elucidated.[19] Over the following years other compounds belonging to the leptomycin family were isolated and classified, including callystatin[20] (**VII**), leptolstatin[21] (**VIIIa**), ratjadone[22] (**IX**), kazusamycins[23] (**X**), leptofuranins[24] (**XI-XIV**) and anguinomycins[25] (**XV-XVIII**) (Figure 2). A biosynthetic pathway for the leptomycins

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[15] M. S. Greenblatt, W. P. Bennett, M. Hollstein, C. C. Harris, *Cancer Res.* **1994**, *54*, 4855-4878.

[16] Review: M. Kalesse, M. Christmann, *Synthesis* **2002**, 981-1003.

[17] a) T. Hamamoto, S. Gunji, H. Tsuji, T. Beppu, *J. Antibiot.* **1983**, *36*, 639-645; b) T. Hamamoto, H. Seto, T. Beppu, *J. Antibiot.* **1983**, *36*, 646-650.

[18] T. Hamamoto, T. Uozumi, T. Beppu, *J. Antibiot.* **1985**, *38*, 1573-1580.

[19] K. Komiyama, K. Okada, S. Tomisaka, *J. Antibiot.* **1985**, *38*, 427-429.

[20] M. Kobayashi, K. Higuchi, N. Murakami, H. Tajima, S. Aoki, *Tetrahedron Lett.* **1997**, *38*, 2859-2862.

[21] a) K. Abe, M. Yoshida, S. Horinouchi, T. Beppu, *J. Antibiot.* **1993**, *46*, 728-734; b) K. Abe, M. Yoshida, H. Naoki, S. Horinouchi, T. Beppu, *J. Antibiot.* **1993**, *46*, 735-740.

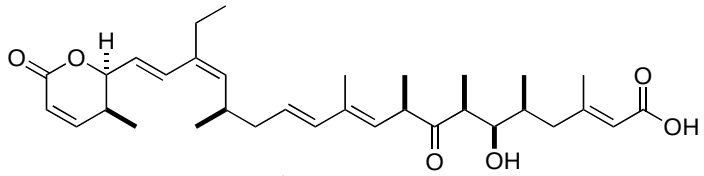
[22] Isolation: K. Gerth, D. Schummer, G. Hofle, H. Irschik, H. Reichenbach, *J. Antibiot.* **1995**, *48*, 973-976. Biological evaluation: a) M. Kalesse, M. Christmann, U. Bhatt, M. Quitschalle, E. Claus, A. Saeed, A. Burzlaff, C. Kasper, L. O. Haustedt, E. Hofer, T. Scheper, W. Beil, *ChemBioChem* **2001**, *2*, 709-714; b) A. Burzlaff, M. Kalesse, C. Kasper, T. Scheper, *Appl. Microbiol. Biotechnol.* **2003**, *62*, 174-179.

[23] Isolation: I. Umezawa, K. Komiyama, H. Oka, *J. Antibiot.* **1984**, *37*, 706-711; K. Funaishi, K. Kawamura, Y. Sugiura, *J. Antibiot.* **1987**, *40*, 778-785. Biological evaluation: a) K. Komiyama, K. Okada, Y. Hirokawa, *J. Antibiot.* **1985**, *38*, 224-229; b) E. Yoshida, Y. Nishimuta, K. Naito, *J. Antibiot.* **1987**, *40*, 391-393; c) E. Yoshida, K. Komiyama, K. Naito, Y. Watanabe, K. Takamiya, A. Okura, K. Funaishi, K. Kawamura, S. Funayama, I. Umezawa, *J. Antibiot.* **1987**, *40*, 1596-1604; d) K. Takamiya, E. Yoshida, T. Takahashi, A. Okura, M. Okanishi, K. Komiyama, I. Umezawa, *J. Antibiot.* **1988**, *41*, 1854-1861.

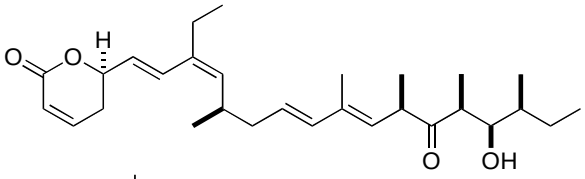
[24] a) Y. Hayakawa, K. Y. Sohda, K. Furihata, T. Kuzuyama, K. Shin-ya, H. Seto, *J. Antibiot.* **1996**, *49*, 974-979; b) Y. Hayakawa, K. Y. Sohda, H. Seto, *J. Antibiot.* **1996**, *49*, 980-984.

[25] a) Y. Hayakawa, K. Adachi, N. Komeshima, *J. Antibiot.* **1987**, *40*, 1349-1352; b) Y. Hayakawa, K. Y. Sohda, K. Shin-Ya, T. Hidaka, H. Seto, *J. Antibiot.* **1995**, *48*, 954-961.

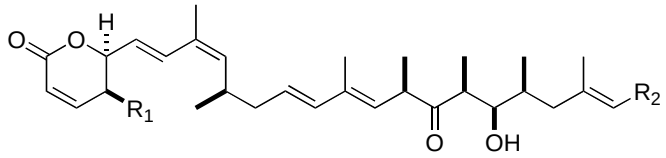
has also been reported.[26] All compounds belonging to the leptomycin family display an α,β-unsaturated lactone and two diene systems separated by two sp^3-hybridized carbons, suggesting that these structural motifs are important for biological target recognition and activity.



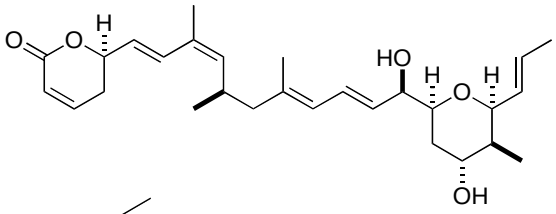
Leptomycin B (**VI**)



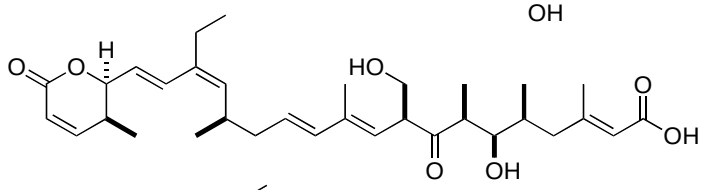
(–)-Callystatin A (**VII**)



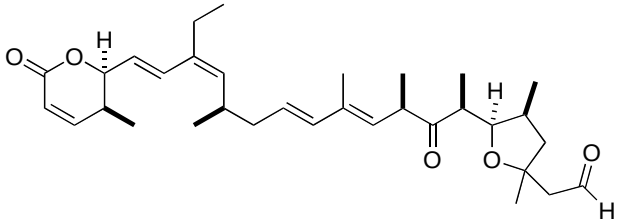
Leptolstatin (**VIIIa**)
R_1 = H; R_2 = CH_2OH
Leptostatin (**VIIIb**)
R_1 = CH_3; R_2 = CH_3



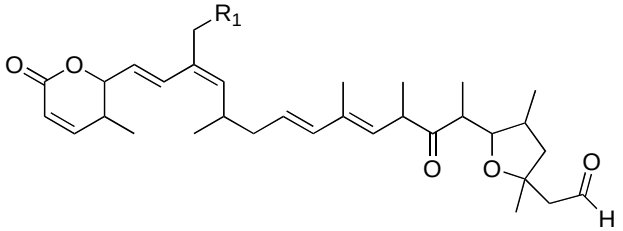
(+)-Ratjadone (**IX**)



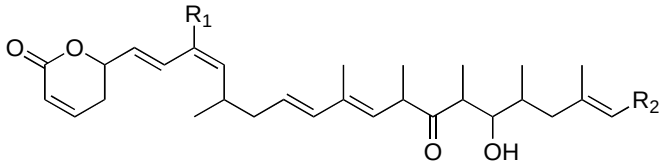
(–)-Kazusamycin A (**X**)



Leptofuranin D (**XI**)



Leptofuranins
A: R_1 = CH_2OH; R_2 = H (**XII**)
B: R_1 = CH_2OH; R_2 = CH_3 (**XIII**)
C: R_1 = CHO; R_2 = H (**XIV**)



Anguinomycins
A: R_1 = CH_3; R_2 = COOH (**XV**)
B: R_1 = C_2H_5; R_2 = COOH (**XVI**)
C: R_1 = CH_3; R_2 = CH_3 (**XVII**)
D: R_1 = C_2H_5; R_2 = CH_3 (**XVIII**)

Figure 2: The leptomycin family.

[26] T. Hamamoto, T. Uozumi, T. Beppu, *J. Antibiot.* **1985**, *38*, 533-535.

Following their discovery, several biological investigations on these compounds were carried out and LMB (**VI**) (Figure 2) itself was found to be a strong inhibitor of the nucleocytoplasmic transport of proteins.[27] The mode of action is unknown, but results suggest that a covalent addition between a protein involved in the nucleocytoplasmic transport and LMB takes place.[28] Although LMB has been evaluated in clinical trials and finally abandoned due to its toxicity,[29] its use is frequently reported as a tool compound in cell biology. Recent publications reported the possibility of synergic therapy by administration of different anticancer agents combined with LMB.[30] Earlier this year, *Mutka* and co-workers reported astonishing results, during their search for new nuclear export inhibitors, their LMB derivatives were shown to have the same potency than LMB, but up to 16-fold better tolerated *in vivo*.[31] Moreover, new compounds were found to be selective between normal and cancer cells. These promising results encourage chemists to find less toxic nucleocytoplasmic transport inhibitors in order to develop a new therapy against cancer. We decided to develop a synthesis for the anguinomycins C (**XVII**) and D (**XVII**) (Figure 2), which, as reported in literature,[25b] display selectivity between normal and tumoral cells. In this project a series of analogs will also be prepared and submitted for biological evaluation in order to understand the mode of action, the target and the selectivity of these compounds.

2.2.2. Biological Activity and Mode of Action

The most investigated member of the leptomycin family has been LMB (**VI**) (Figure 2). After the discovery of its potent cytotoxicity (10 ng/mL on rat 3Y1 fibroblasts),[18] further biological investigations on this compound were performed. LMB causes cell-cycle arrest in the G1 and G2 phases in eukaryotic cells[32] and

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[27] N. Kudo, B. Wolff, T. Sekimoto, E. P. Schreiner, Y. Yoneda, M. Yanagida, S. Horinouchi, M. Yoshida, *Exp. Cell Res.* **1998**, *242*, 540-547.

[28] N. Kudo, N. Matsumori, H. Taoka, D. Fujiwara, E. P. Schreiner, B. Wolff, M. Yoshida, S. Horinouchi, *Proc. Natl. Acad. Sci. U. S. A.* **1999**, *96*, 9112-9117.

[29] E. S. Newlands, G. J. S. Rustin, M. H. Brampton, *Br. J. Cancer* **1996**, *74*, 648-649.

[30] a) A. Aloisi, S. Di Gregorio, F. Stagno, P. Guglielmo, F. Mannino, M. P. Sormani, P. Bruzzi, C. Gambacorti-Passerini, G. Saglio, S. Venuta, R. Giustolisi, A. Messina, P. Vigneri, *Blood* **2006**, *107*, 1591-1598; b) R. K. Kancha, N. Von Bubnoff, C. Miething, C. Peschel, K. S. Götze, J. Duyster, *Haematologica* **2008**, *93*, 1718-1722.

[31] S. C. Mutka, W. Q. Yang, S. D. Dong, S. L. Ward, D. A. Craig, P. B. M. W. M. Timmermans, S. Murli, *Cancer Res.* **2009**, *69*, 510-517.

[32] M. Yoshida, M. Nishikawa, K. Nishi, K. Abe, S. Horinouchi, T. Beppu, *Exp. Cell Res.* **1990**, *187*, 150-156.

selectively targets the chromosome maintenance region 1 (CRM1 or exportin 1),[33] a protein involved in nucleocytoplasmic transport.[34] The evolutionary conserved nature of CRM1 highlights its importance as a receptor for leucine-rich nuclear export signal (NES) and its central role for NES-dependent nuclear export of protein complexes in eukaryotic cells.[35] In order to perform transport from the nucleus to the cytoplasm, CRM1 has to recognize the nuclear export signal (NES) present on the *cargo* and form the complex CRM1/NES-*cargo*/RanGTP. The complex is consequently shuttled out of the nucleus, the *cargo* and the RanGDP, are then released and CRM1 transported back to the nucleus.

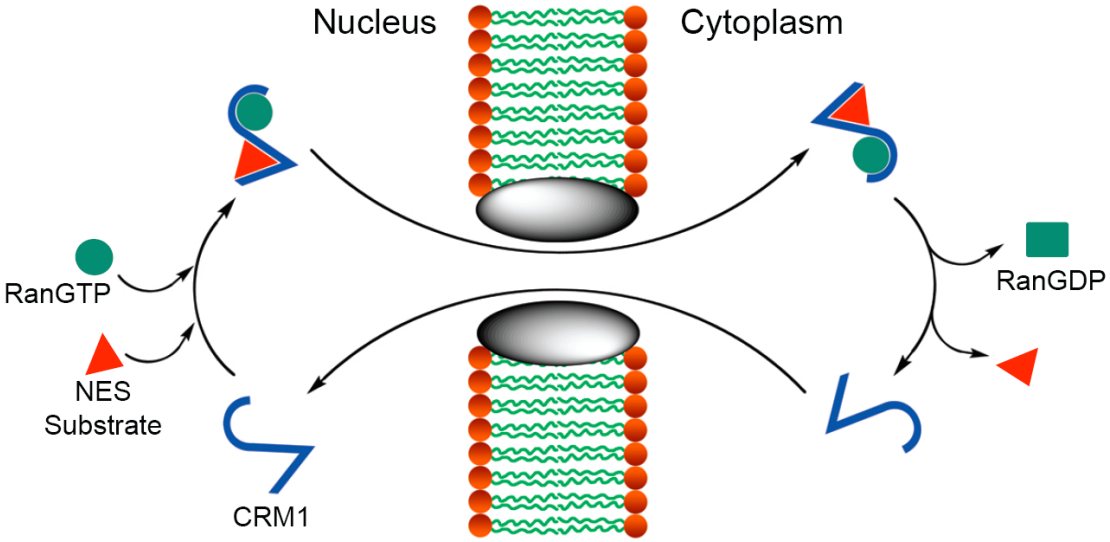

Figure 3: CRM-1 mediated NES protein export.

In the human body, there are so-called tumor-suppressor proteins,[14,36] such as the retinoblastoma protein (pRb)[37] and the p53 protein,[38] that prevent the proliferation of cancerous cells. During cell replication, there are checkpoints controlling the process that allows the transition to the next phase if no problem is detected. Conversely, if damage to the genetic material has occurred, tumor suppressor proteins are charged to

[33] a) K. Nishi, M. Yoshida, D. Fujiwara, M. Nishikawa, S. Horinouchi, T. Beppu, *J. Biol. Chem.* **1994**, *269*, 6320-6324; b) K. Stade, C. S. Ford, C. Guthrie, K. Weis, *Cell* **1997**, *90*, 1041-1050; c) M. Fornerod, M. Ohno, M. Yoshida, I. W. Mattaj, *Cell* **1997**, *90*, 1051-1060.

[34] E. A. Nigg, *Nature* **1997**, *386*, 779-787.

[35] a) U. Kutay, S. Güttinger, *Trends in Cell Biology* **2005**, *15*, 121-124; b) C. Drahl, B. F. Cravatt, E. J. Sorensen, *Angew. Chem., Int. Ed.* **2005**, *44*, 5788-5809.

[36] a) R. A. Weinberg, *Science* **1991**, *254*, 1138-1146; b) R. Weinberg, *Neuron* **1993**, *11*, 191-196.

[37] a) J. Bartek, J. Bartkova, J. Lukas, *Exp. Cell Res.* **1997**, *237*, 1-6; b) S. Herwig, M. Strauss, *Eur. J. Biochem.* **1997**, *246*, 581-601.

[38] a) M. E. Perry, A. J. Levine, *Current Opinion in Genetics and Development* **1993**, *3*, 50-54; b) T. Jacks, R. A. Weinberg, *Nature* **1996**, *381*, 643-644.

stop the cell cycle and repair the damages or induce apoptosis. This is the case for pRb that can stop the cell cycle at the phase G1 if problems are detected.[39] Directly related to pRb there is p53, also an oncosuppressive protein that leads to apoptosis when accumulated in the nucleus.[40] However, the way in which these two proteins really function and interact is highly complex and not yet clear. Investigations show that they can regulate each other through a complex network of interactions and the fate of the cell is dependent upon them (Figure 4).[41] Tumor-suppressors play a central role in mammalian cell cycles and their deactivation leads to uncontrolled proliferation. This process is common during the development of a wide variety of human cancers and is a key factor in tumorigenesis.[42] When the anti-apoptotic function of pRb is interrupted, the cell is subjected to a p53-mediated apoptosis allowing the elimination of cells in which the pRb pathway is deregulated.[43] If p53 is also mutated or deactivated, the cell loses the ability to defend itself and uncontrolled tumoral cell proliferation starts. The localization of wild-type p53 in the nucleus is fundamental if it is to perform its tumor-suppressor function,[44] whereas p53 mutants are translocated into the cytoplasm.[45] Some tumors have a common mechanism to deactivate p53 and therefore abrogating its functionality. For reasons that remain unclear, the wild-type p53 is sequestered in the cytoplasm compromising its tumor-suppressor ability.[46]

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[39] R. A. Weinberg, *Cell* **1995**, *81*, 323-330.

[40] S. Laín, D. Xirodimas, D. P. Lane, *Exp. Cell Res.* **1999**, *253*, 315-324.

[41] N. Godefroy, C. Lemaire, B. Mignotte, J. L. Vayssière, *Apoptosis* **2006**, *11*, 659-661.

[42] a) B. Vogelstein, *Nature* **1990**, *348*, 681-682; b) M. B. Kastan, O. Onyekwere, D. Sidransky, B. Vogelstein, R. W. Craig, *Cancer Res.* **1991**, *51*, 6304-6311; c) C. C. Harris, M. Hollstein, *N. Engl. J. Med.* **1993**, *329*, 1318-1327.

[43] S. D. Morgenbesser, B. O. Williams, T. Jacks, R. A. DePinho, *Nature* **1994**, *371*, 72-74.

[44] G. Shaulsky, N. Goldfinger, A. Peled, V. Rotter, *Cell Growth Differ.* **1991**, *2*, 661-667.

[45] J. Martinez, I. Georgoff, A. J. Levine, *Genes Dev.* **1991**, *5*, 151-159.

[46] a) U. M. Moll, M. Laquaglia, J. Benard, G. Riou, *Proc. Natl. Acad. Sci. U. S. A.* **1995**, *92*, 4407-4411; b) U. M. Moll, A. G. Ostermeyer, R. Haladay, B. Winkfield, M. Frazier, G. Zambetti, *Mol. Cell. Biol.* **1996**, *16*, 1126-1137; c) A. G. Ostermeyer, E. Runko, B. Winkfield, B. Ahn, U. M. Moll, *Proc. Natl. Acad. Sci. U. S. A.* **1996**, *93*, 15190-15194.

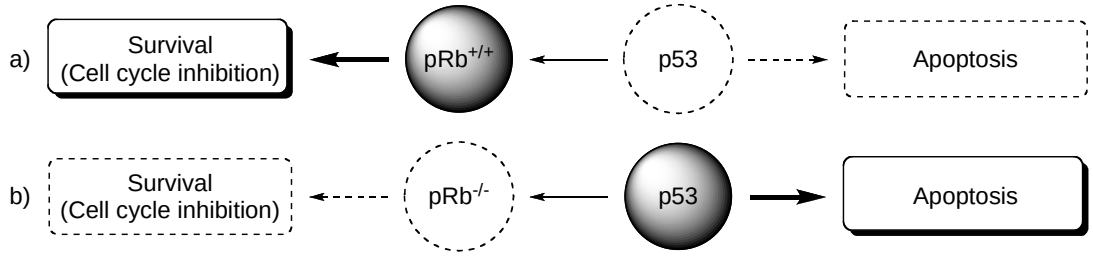

Figure 4: pRb and p53 interaction. a) When $pRb^{+/+}$ is functional, cell cycle arrest and survival of the cell is induced. b) When $pRb^{-/-}$ is inactive, p53 will induce apoptosis.

In normal cells, the concentration of p53 is regulated by CRM1. The NES on p53 is recognized by CRM1, which then transports the cargo out of the nucleus. If the interaction between CRM1 and p53 is blocked, the tumor suppressor is accumulated leading to apoptosis.[47] Biological studies show that LMB inhibits this transport by a probable *Michael*-type addition of the thiol group of the cysteine residue in position 529 in CRM1 to the LMB (Figure 5).[35b]

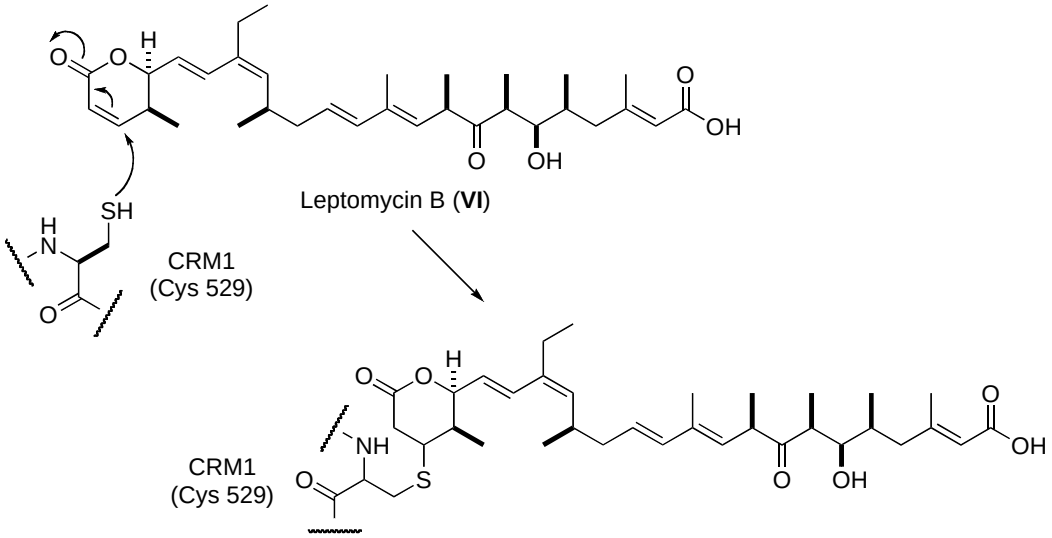

Figure 5: Postulated mechanism of action of LMB (**VI**) against CRM-1.

These results are supported by the fact that functionalization at the β position of the α,β-unsaturated lactone are not tolerated and a complete loss of the CRM1 inhibition is observed.[27] The same results were also reported for ratjadone (**IX**).[48] Further investigations have been performed with different nucleocytoplasmic

[47] M. Kanai, K. Hanashiro, S. H. Kim, S. Hanai, A. H. Boulares, M. Miwa, K. Fukasawa, *Nat. Cell Biol.* **2007**, *9*, 1175-1183.
[48] T. Meissner, E. Krause, U. Vinkemeier, *FEBS Lett.* **2004**, *576*, 27-30.

inhibitors, confirming the addition of Cys-529 of CRM1. The inhibitory effects are comparable to those observed for LMB and suggest that the same binding site on CRM1 is shared.[49] The binding of LMB to CRM1 inhibits the recognition of leucine rich NES since they share the same binding site.[50] However, it is still unclear whether the addition of Cys-529 to LMB induces a conformational change in exportin1 or just sterically blocks the approach of the NES.[50] In addition, it appears that Cys-529 is not fundamental for CRM1 functionality as replacing the cysteine by another amino acid did not affect the nucleocytoplasmic transport.[51] However, the presence of a *Michael* acceptor on the inhibiting agent has been demonstrated to play a fundamental role for activity, but there was not sufficient information to fully elucidate the mode of action. A more complex mechanism than the 1,4-addition on the α,β-unsaturation is probably involved.[52] The results reported by *Mutka* and co-workers[31] also revealed the importance of the linear chain for tuning the selectivity of the inhibitor. Their new derivative was able to selectively kill cancer cells whilst only inducing cell cycle arrest in normal lung fibroblast cells. The treatment of cancer cells with the derivative leads to a rapid and continuous block of the nucleocytoplasmic transport, with an increment of the apoptosis due to the overexpression of p53. Normal cells were not subjected to apoptosis and the cell cycle was just arrested with consequent decreasing of cells proliferation; p53 was not overexpressed. Even with a persistent halt of the cell cycle, the normal lung fibroblast cells remained viable and at the end of the treatment they could regain normal proliferation.[31] The aimed relocalization of p53 protein is a promising technique to regulate cell proliferation and selective apoptosis.

Another protein directly related to CRM1 is the human immunodeficiency virus type 1 (HIV-1) regulatory protein Rev; the NES of Rev is recognized by the exportin 1 and translocated out of the nucleus. HIV-1 Rev protein plays a fundamental role in the regulation of the HIV-1 mRNA which promotes the export of unspliced and partially spliced mRNA. Rev export is a necessary condition for Rev function.[53]

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[51] N. Kudo, N. Matsumori, H. Taoka, D. Fujiwara, E. P. Schreiner, B. Wolff, M. Yoshida, S. Horinouchi, *Proc. Natl. Acad. Sci. U. S. A.* **1999**, *96*, 9112-9117.

[52] T. Van Neck, C. Pannecouque, E. Vanstreels, M. Stevens, W. Dehaen, D. Daelemans, *Bioorg. Med. Chem.* **2008**, *16*, 9487-9497.

[53] B. Wolff, G. Cohen, J. Hauber, D. Meshcheryakova, C. Rabeck, *Exp. Cell Res.* **1995**, *217*, 31-41.

Biological results show that inhibition of CRM1 results in the arrest of Rev translocation[54] and can be considered a potential approach for anti-HIV therapy.[55]

2.2.3. Total Syntheses and Synthetic Studies

2.2.3.1. The Syntheses of Callystatin

The leptomycin family has been widely investigated and several total syntheses of its members have been reported. Callystatin is the most commonly synthesized compound with ten total syntheses published.[56] The retrosynthetic approaches adopted by the different groups are shown in figure 6.

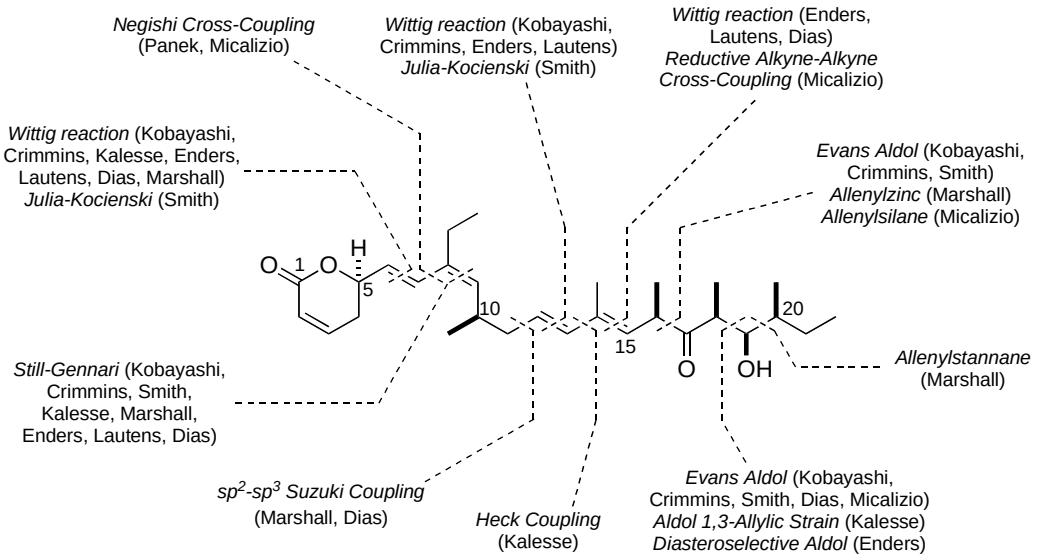

Figure 6: Retrosynthetic approach for the syntheses of (–)-callystatin A (**VII**).

Although the strategies outlined above are all different, careful analysis reveals several common intermediates. This is the case for the synthesis of *Kobayashi*, *Crimmins*, *Kalesse*, *Enders*, *Lautens* and *Dias* and co-workers where a common aldehyde intermediate **XXIII** was obtained. *Kobayashi* and co-workers started the

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54 B. Wolff, J. J. Sanglier, Y. Wang, *Chem. Biol.* **1997**, *4*, 139-147.

55 Y. Wang, M. Ponelle, J. J. Sanglier, B. Wolff, *Helv. Chim. Acta* **1997**, *80*, 2157-2167.

56 a) N. Murakami, W. Wang, M. Aoki, Y. Tsutsui, M. Sugimoto, M. Kobayashi, *Tetrahedron Lett.* **1998**, *39*, 2349-2352; b) M. T. Crimmins, B. W. King, *J. Am. Chem. Soc.* **1998**, *120*, 9084-9085; c) A. B. Smith III, B. M. Brandt, *Org. Lett.* **2001**, *3*, 1685-1688; d) M. Kalesse, M. Quitschalle, C. P. Khandavalli, A. Saeed, *Org. Lett.* **2001**, *3*, 3107-3109; e) J. L. Vicario, A. Job, M. Wolberg, M. Müller, D. Enders, *Org. Lett.* **2002**, *4*, 1023-1026; f) J. A. Marshall, M. P. Bourbeau, *J. Org. Chem.* **2002**, *67*, 2751-2754; g) M. Lautens, T. A. Stammers, *Synthesis* **2002**, 1993-2012; h) N. F. Langille, J. S. Panek, *Org. Lett.* **2004**, *6*, 3203-3206; i) L. C. Dias, P. R. R. Meira, *J. Org. Chem.* **2005**, *70*, 4762-4773; j) H. A. Reichard, J. C. Rieger, G. C. Micalizio, *Angew. Chem., Int. Ed.* **2008**, *47*, 7837-7840.

synthesis with the TBDPS protected (*S*)-glycidol (**XIX**), which undergoes epoxide ring opening by the attack of deprotonated 3-phenylsulphonylorthopropionate (**XX**) to afford the orthoester **XXI**. Treatment with DBU induced lactonization and elimination of the phenylsulfinic acid gave the α,β-unsaturated lactone **XXII**. Additional transformations afforded aldehyde **XXIII** in nine steps from commercially available (*S*)-glycidol (**XIX**) (Scheme 1, eq. 1). *Crimmins* and co-workers also started the synthesis from the TBDPS protected (*S*)-glycidol **XIX** and obtained the common aldehyde **XXIII** in six steps using ring closing metathesis to form the unsaturated lactone (Scheme 1, eq. 2). A different approach was adopted by *Kalesse* and co-workers who prepared the six-membered ring *via* a hetero *Diels-Alder* (HDA) reaction. Commercially available ethyl glyoxylate (**XXIV**) and 1-methoxy-1,3-butadiene (**XXV**) were reacted in solvent free conditions catalyzed by BINOL/Ti(O*i*Pr)$_4$. The product was obtained in 65% yield, 98% *e.e*. at C(5) and 1:10 *d.r.* (C(1):C(5) *anti*:*syn*).[57] A further three transformations afforded aldehyde **XXIII** (Scheme 1, eq. 3). *Enders* and co-workers adopted an enzymatic reduction approach initially developed by *Müller* and co-workers.[58] Reduction of 3,5-dioxocarboxylate[59] **XXVI** by baker's yeast gave the hydroxyketoester **XXVII** in 50% yield and 94% *e.e.*. The product **XXVII** was subsequently converted to aldehyde **XXIII** using standard chemistry (Scheme 1, eq. 4). This procedure required eight steps from 3,5-dioxocarboxylate **XXVI**, which was prepared in one step from commercially available *tert*-butyl acetoacetate and methyl chloroacetate. *Lautens* and co-workers started their synthesis by treatment of the THP protected propargylic alcohol **XXVIII** with *n*BuLi, the generated anion then attacks the epoxide ring of the TBDPS protected (*S*)-glycidol (**XIX**) affording homo propargylic alcohol **XXIX**. The product was transformed to aldehyde **XXIII** using standard reactions in six steps from (*S*)-glycidol (**XIX**) (Scheme 1, eq. 5). *Dias* and co-workers began with diol **XXX**[60] derived from the selective reduction of the diethyl (*S*)-malate and in a nine step sequence also achieved the aldehyde intermediate **XXIII** (Scheme 1, eq. 6).

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[57] M. Quitschalle, M. Christmann, U. Bhatt, M. Kalesse, *Tetrahedron Lett.* **2001**, *42*, 1263-1265.

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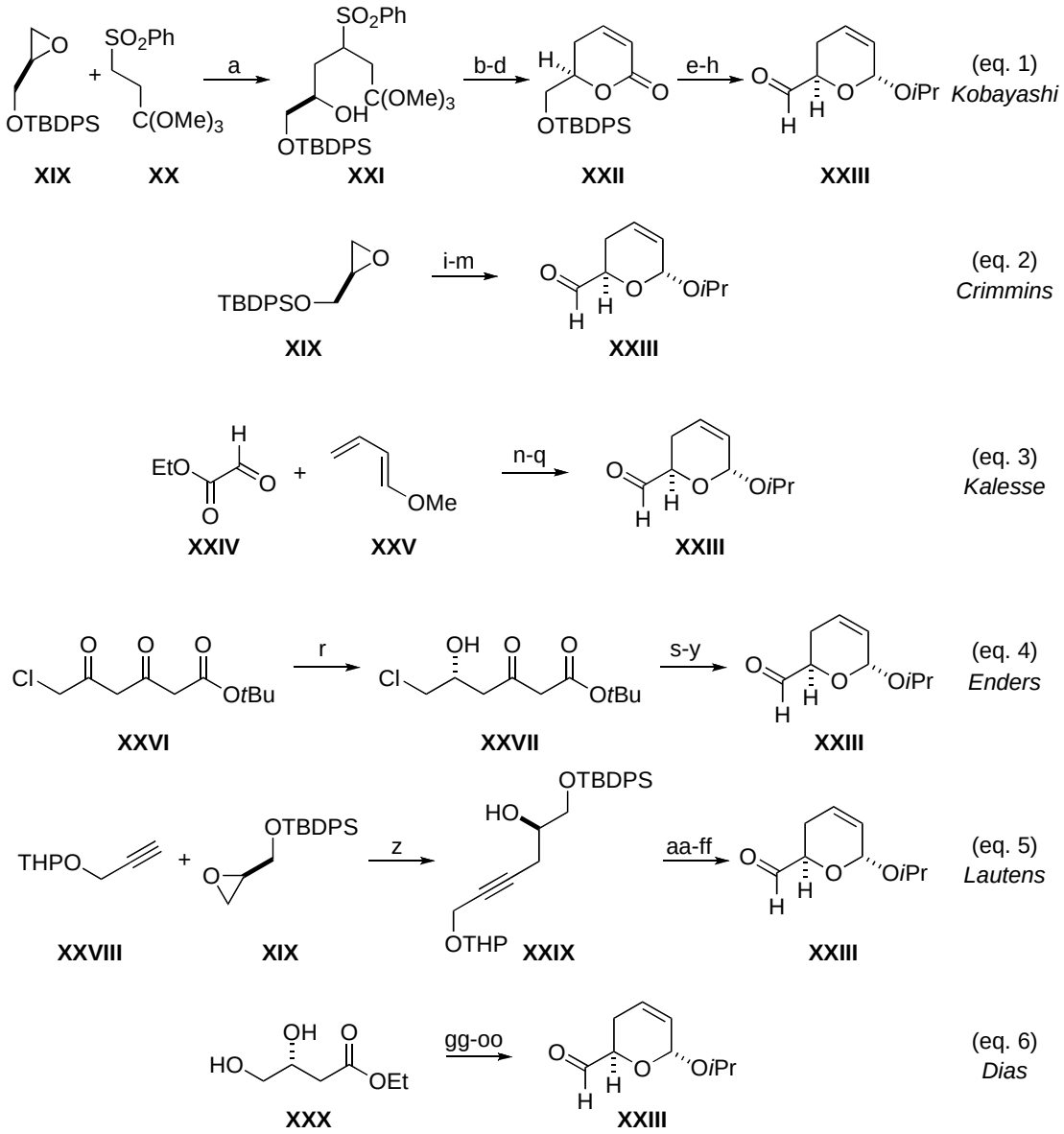
[60] S. Saito, T. Ishikawa, A. Kuroda, K. Koga, T. Moriwake, *Tetrahedron* **1992**, *48*, 4067-4086.



Scheme 1: a) **XX**, *n*BuLi, DMPU, THF, –20 ºC → –5 ºC; b) H_2SO_4 (3 M)-THF (3:1); c) *p*TsOH, 4 Å MS, $ClCH_2CH_2Cl$, 70 ºC; d) Et_3N, DBU, $ClCH_2CH_2Cl$, –10 ºC, 82% (4 steps); e) DIBAL-H, CH_2Cl_2, –78 ºC; f) *i*PrOH, PPTS, benzene, 82% (2 steps); g) TBAF, THF; h) Swern oxidation, 99% (2 steps); i) vinyl magnesium bromide, CuI, 85%; j) acroleine diisopropyl acetal, PPTS; k) $Cl_2(Cy_3P)_2Ru$=CHPh, 71% (2 steps); l) TBAF; m) Swern oxidation, 90% (2 steps); n) Ti(*i*PrO)$_4$, (+)-BINOL, 4 Å MS, CH_2Cl_2, 65%, 98% *e.e.*, 1:10 *d.r.*; o) $LiAlH_4$, Et_2O, 0 ºC; p) *i*PrOH, PPTS; q) Swern oxidation, 77% (3 steps); r) baker's yeast, 50%, 94% *e.e.*; s) $NaBH_4$, EtOH, 0 ºC; t) *p*TsOH (cat.), toluene, reflux, 78% (2 steps); u) DIBAL-H, CH_2Cl_2, –78 ºC; v) *i*PrOH, PPTS, benzene, 60 ºC, 79%(2 steps); w) TBAA, NMP, 85 ºC; x) K_2CO_3, MeOH, RT, 86% (2 steps); y) Swern oxidation, 95%; z) *n*BuLi, THF, –78 °C; then BF_3•Et_2O; then **XIX**, 78%; aa) PPTS, EtOH, 50 °C, 3 h, 99%; bb) Lindlar cat., toluene, H_2 (1 atm), 20 °C, 3 h, 99%; cc) MnO_2, CH_2Cl_2, 20 °C, 13 h, 75% (based on conversion); dd) *i*PrOH, PPTS, 20 °C, 45 min, 93%; ee) TBAF; ff) Swern oxidation; gg) TBSCl, imidazole, DMF, RT, 2 h, 95%; hh) DIBAL-H, CH_2Cl_2, –78 °C; ii) ethyl 2-((bis(*o*-tolyloxy))phosphoryl)acetate, NaH, THF, –78 °C, 75% (2 steps); jj) Dowex, MeOH, RT, 72 h, 95%; kk) TBSOTf, 2,6-lutidine, CH_2Cl_2, RT, 91%; ll) DIBAL-H, CH_2Cl_2, –23 °C, 90%; mm) *i*PrOH, PPTS, RT, 1 h, 87%; nn) TBAF, THF, RT, 15 h, 99%; oo) Swern oxidation, –78 °C, 95%.

A different intermediate was achieved by *Smith* and co-workers who prepared sulfone **XXXII**, for a *Julia-Kocienski* olefination,[61] *via* a [4+2] cycloaddition between 1-methoxy-1,3-butadiene (**XXV**) and *Oppolzer* sultam **XXXI** in five steps (Scheme 2, eq. 1).[62] The sultam **XXXI** was prepared in two steps from commercially available (2*S*)-bornane-10,2-sultam.[63] *Marshall* and co-workers opened the PMB protected (*S*)-glycidol **XXXIV** by addition of the lithium acetylide **XXXIII** affording alcohol **XXXV** which, was transformed in a three steps sequence to aldehyde **XXXVI** (Scheme 2, eq. 2). *Panek* and co-workers started their synthesis with the addition of allylmagnesium bromide to aldehyde **XXXVII**.[64] Enantioselective kinetic resolution of the racemic alcohol **XXXVIII** with lipase *Pseudomonas* AK produced the desired (*R*)-enantiomer **XXXIX** in 46% yield and more than 95% *e.e.*. The unreacted (*S*)-enantiomer (–)-**XXXVIII** was separated and converted to the (*R*)-enantiomer **XXXIX** *via* a *Mitsunobu* reaction.[65] Ring closing metathesis and protecting group manipulations gave terminal alkyne **XL** in six steps (seven to convert the unreacted (*S*)-enantiomer (–)-**XXXVIII**) from aldehyde **XXXVII** (Scheme 2, eq. 3). The most recent synthesis was reported by *Micalizio* and co-workers who referenced our paper for the formation of the terminal alkyne **XL** (Scheme 2, eq. 4).[66] Subsequently hydrozirconation using *Schwartz* reagent[67] furnished the vinyl iodide compound **XLII** in 86% yield. The details of our approach will be presented in chapter 2.4 which are concerned with the total synthesis of the anguinomycins C and D. All the syntheses so far have required a large number of steps to obtain the six-member ring of callystatin (**VII**). The most straightforward way was the *Diels-Alder* approach adopted by *Kalesse* and co-workers allowing the formation of aldehyde **XXIII** in four steps (Scheme 1, eq. 3).[57]

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[61] P. R. Blakemore, W. J. Cole, P. J. Kocienski, A. Morley, *Synlett* **1998**, *1998*, 26-28.
[62] T. Bauer, C. Chapuis, A. Jezewski, J. Kozak, J. Jurczak, *Tetrahedron: Asymmetry* **1996**, *7*, 1391-1404.
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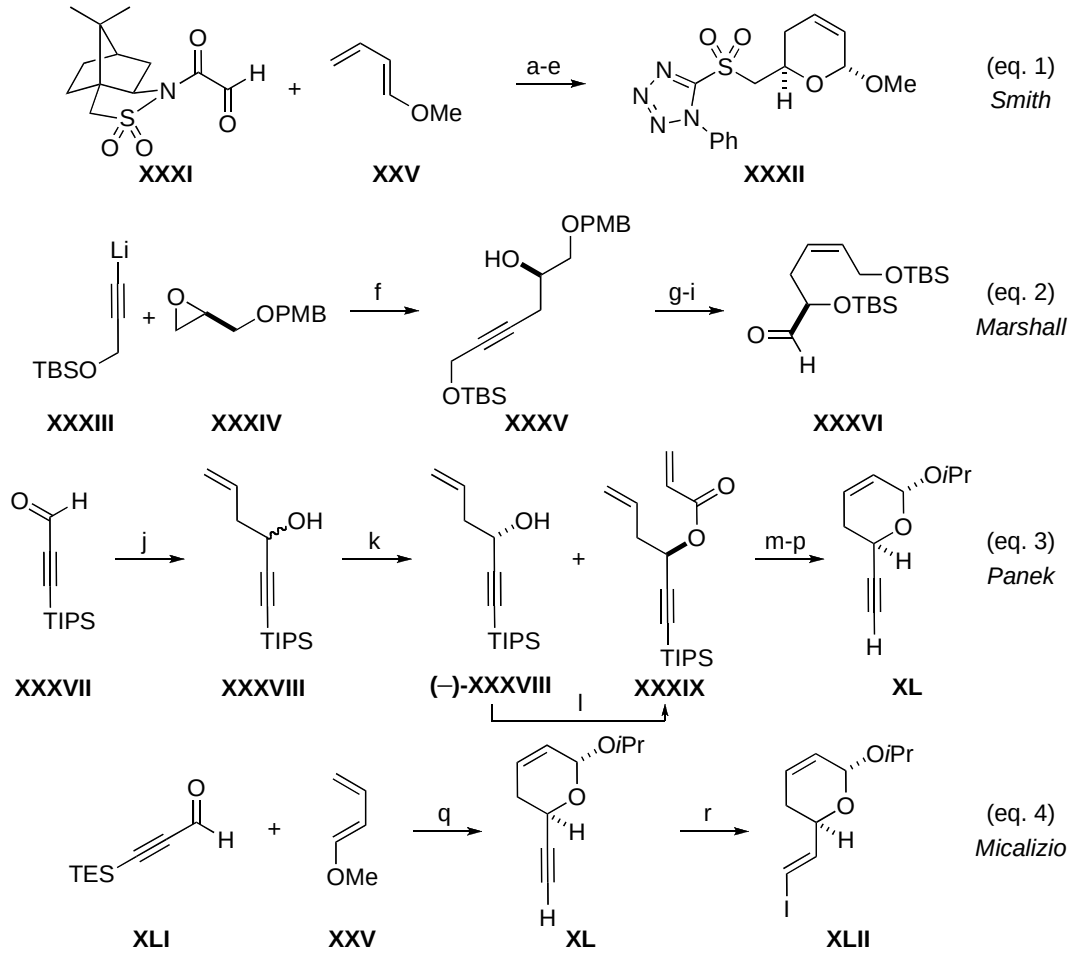
[65] O. Mitsunobu, M. Yamada, *Bull. Chem. Soc. Jpn.* **1967**, *40*, 2380-2382.
[66] S. Bonazzi, S. Güttinger, I. Zemp, U. Kutay, K. Gademann, *Angew. Chem., Int. Ed.* **2007**, *46*, 8707-8710.
[67] D. W. Hart, J. Schwartz, *J. Am. Chem. Soc.* **1974**, *96*, 8115;



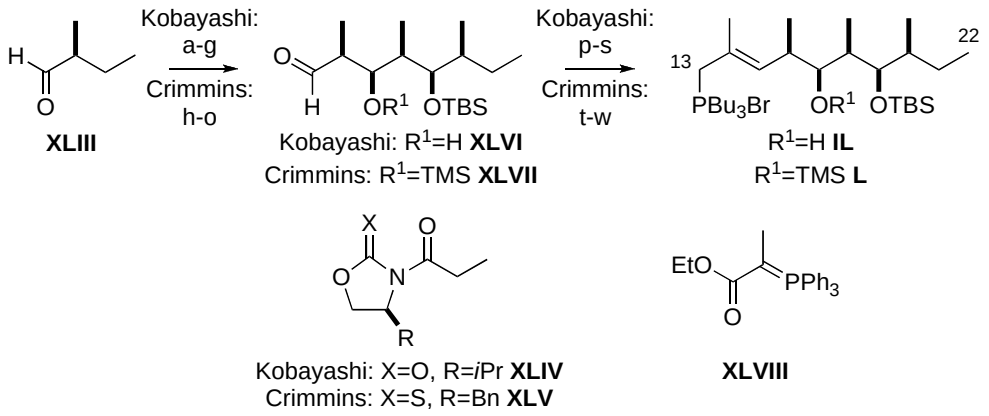
Scheme 2: a) $Eu(fod)_3$ (2 mol %) or without catalyst, CH_2Cl_2, 1 atm, 20 °C, 20 h; b) PPTS, MeOH, RT, 15 h, c) $LiAlH_4$, THF, 90%; d) 1-phenyl-1*H*-tetrazole-5-thiol, DEAD/Ph_3P, THF, 0 ºC → RT, 99%; e) H_2O_2/EtOH/H_2O, $(NH_4)_6Mo_7O_{24}$•$4H_2O$, 69%; f) BF_3•OEt_2, 91%; g) H_2/Pd-$BaSO_4$, quinoline, benzene, 99%; h) DDQ, 88%; i) Swern oxidation, 82%; j) allylmagnesium bromide, THF, –20 °C, 99%; k) vinyl acrylate, lipase AK, hexanes, 7 days, RT, 44% (–)-**XXXVIII**, 46% **XXXIX**, *e.e.* > 95%; l) DIAD, acrylic acid, PPh_3, THF, 0 °C → RT, 86%; m) Grubbs I, CH_2Cl_2, 40 °C, 83%; n) DIBAL-H, –78 °C, CH_2Cl_2; o) *i*PrOH, PPTS, benzene, 80 °C, 82% (2 steps); p) 1.3:1 AcOH/TBAF, THF, RT, 91%; q) [ref. 66]; q) Cp_2ZrHCl, THF, then I_2, 86%.

The strategies adopted for the preparation of the polyketide chain and the two diene systems were principally based on aldol reactions, metal catalyzed cross-couplings, *Wittig* reactions and *Still-Gennari* olefinations.[68] For the synthesis of the C(13)-C(22) fragment, *Kobayashi* and co-workers and *Crimmins* and co-workers adopted basically the same strategy. The synthesis was characterized by two *syn*-aldol reactions starting from (*S*)-2-methylbutanal **XLIII**.[69] *Kobayashi* and co-workers

[68] W. C. Still, C. Gennari, *Tetrahedron Lett.* **1983**, *24*, 4405-4408.

[69] J. D. White, G. L. Bolton, A. P. Dantanarayana, C. M. J. Fox, R. N. Hiner, R. W. Jackson, K. Sakuma, U. S. Warrier, *J. Am. Chem. Soc.* **1995**, *117*, 1908-1939.

employed the acylated *Evans* auxiliary[70] **XLIV** for both aldol reactions. The first aldol adduct was obtained in 98% yield and a 9:1 diastereomeric ratio, while the second one as a single isomer in 85% yield. *Crimmins* and co-workers opted for the propyonyloxazolidinethione auxiliary **XLV** furnishing the two aldol products in 83% and 81% yield respectively in a diastereomeric ratio greater than 98:2. In the first aldol reaction the use of the propyonyloxazolidinethione auxiliary **XLV** gave better stereocontrol when compared to the *Evans* auxiliary, which furnished the product in a 9:1 diastereomeric ratio. Aldehyde **XLVI** was obtained by *Kobayashi* and co-workers in seven steps, while *Crimmins* and co-workers generated aldehyde **XLVII** in eight steps. Both groups reacted their aldehydes with commercially available (carbethoxyethylidene)triphenylphosphorane (**XLVIII**) *via* a *Wittig* reaction. The products were converted to the phosphonium salt **IL** for *Kobayashi* and **L** for *Crimmins* in three further steps (Scheme 3). Interestingly, *Kobayashi* noticed problems during the synthesis when trying to protect the hydroxy group on C(17), probably due to steric reasons and decided to keep it as the free hydroxy. In contrast, *Crimmins* protected it with a TMS group in high yield.

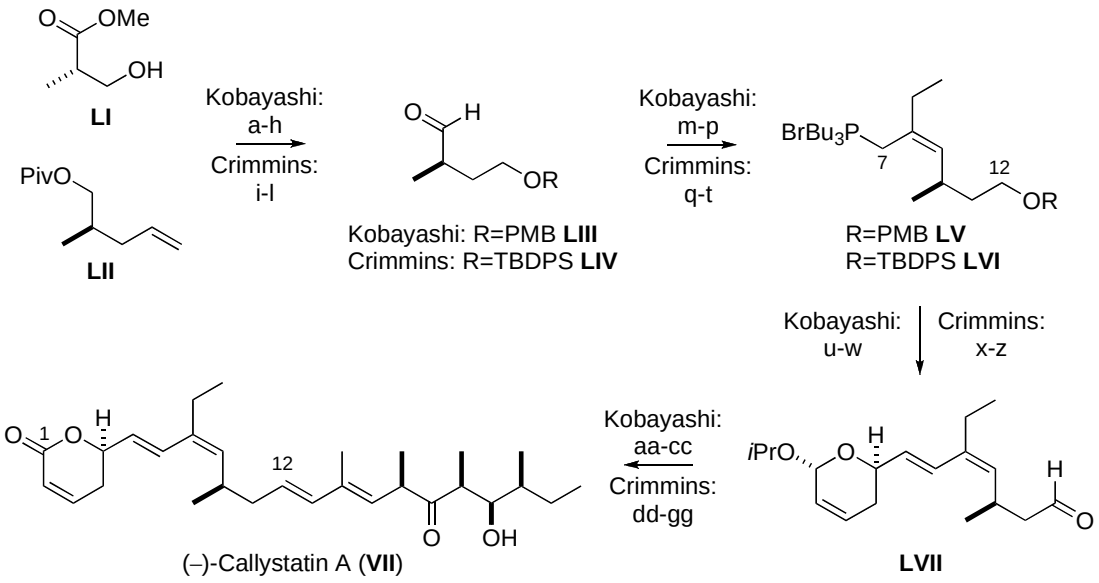


Scheme 3: a) **XLIV**, nBu$_2$BOTf, Et$_3$N, –78 ºC → 0 ºC, 98%, *d.r.* = 9:1; b) AlMe$_3$, MeONHMe•HCl, CH$_2$Cl$_2$, –20 ºC → 0 ºC, 95%; c) TBSOTf, 2,6-lutidine, CH$_2$Cl$_2$, –20 ºC, 100%; d) DIBAL-H, THF, –78 ºC, 76%; e) **XLIV**, nBu$_2$BOTf, Et$_3$N, –78 ºC → 0 ºC, 85%; f) AlMe$_3$, MeONHMe•HCl, CH$_2$Cl$_2$, –78 ºC → 0 ºC, 92%; g) LiAlH$_4$, Et$_2$O, 0 ºC, 96%; h) **XLV**, TiCl$_4$, (–)-sparteine, 83%, *d.r.* > 98:2; i) TBSOTf; j) LiBH$_4$; k) Swern oxidation, 83% (3 steps); l) **XLV**, TiCl$_4$, (–)-sparteine, 81%, *d.r.* > 98:2; m) TMSOTf; n) LiBH$_4$; o) Swern oxidation, 71% (3 steps); p) **XLVIII**, toluene, 94%; q) DIBAL-H, CH$_2$Cl$_2$, –78 ºC, 100%; r) CBr$_4$, Ph$_3$P, 2,6-lutidine, CH$_3$CN, 99%; s) Bu$_3$P, CH$_3$CN, 100%; t) **XLVIII**, CH$_2$Cl$_2$, 40 ºC, 93%; u) DIBAL-H; v) CBr$_4$, Ph$_3$P; w) Bu$_3$P, 82% (3 steps).

The remaining C(7)-C(12) fragment was prepared by *Kobayashi* and co-workers from commercially available methyl (*S*)-(+)-3-hydroxyisobutyrate (**LI**) and readily

[70] D. A. Evans, J. Bartroli, T. L. Shih, *J. Am. Chem. Soc.* **1981**, *103*, 2127-2129.

transformed to aldehyde **LIII** in an eight step sequence. The same aldehyde **LIV**, but with a different protecting group was obtained by *Crimmins* and co-workers in four steps, starting from olefin **LII**, prepared in three steps from allyl iodide.[71] Both groups reacted the aldehyde itself in a *Still-Gennari* olefination; for *Crimmins* a *Z*/*E* ratio of 8:1 was observed, *Kobayashi* did not reported a selectivity. Formation of the phosphonium salt **LV** (resp. **LVI**) was then achieved by both groups following the same three step procedure. Subsequent *Wittig* reaction with aldehyde **XXIII** gave selectively only the (*E*)-coupled C(1)-C(12) fragment which, after deprotection and oxidation afforded the common intermediate **LVII**. Aldehyde **LVII** was reacted with phosphonium salt **IL** (resp. **L**) *via* a *Wittig* reaction with exclusive formation of the *E* product and final modifications converted the coupled product to (–)-callystatin A (**VII**) (Scheme 4). Starting from commercially available materials, the synthesis of *Kobayashi* and co-workers required 39 steps (longest linear sequence 18 steps) and that reported by *Crimmins* and co-workers 37 steps (longest linear sequence 18 steps).

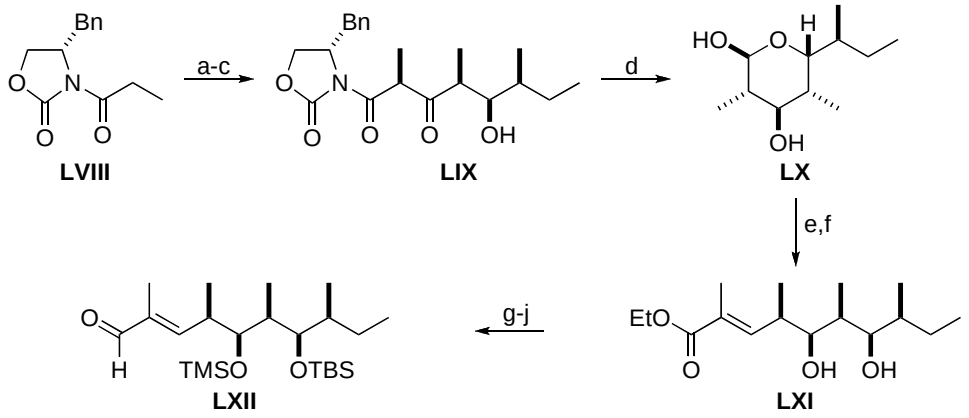


Scheme 4: a) TBDPSCl, imidazole, CH_2Cl_2; b) $LiBH_4$, THF, reflux, 95% (2 steps); c) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, –78 °C; d) $BrPh_3PCH_3$, *n*BuLi, THF, 0 °C; e) BH_3•OEt_2, H_2O_2, 90% (3 steps); f) PMBBr, NaH, THF, 96%; g) TBAF, THF, 97%; h) Swern oxidation; i) O_3, $NaBH_4$; j) TBDPSCl, imidazole, 80% (2 steps); k) DIBAL-H, CH_2Cl_2, –78 °C; l) Swern oxidation, 88% (2 steps); m) $EtO_2CCH(Et)PO(OCH_2CF_3)_2$, KHMDS, 18-crown-6, THF, –78 °C → 0 °C, 92% (2 steps); n) DIBAL-H, CH_2Cl_2, –78 °C; o) CBr_4, Ph_3P, 2,6-lutidine, CH_3CN; p) Bu_3P, CH_3CN, 92% (3 steps); q) $EtO_2CCH(Et)PO(OCH_2CF_3)_2$, KHMDS, 18-crown-6, THF, –78 °C → 0 °C, *Z*:*E* = 8:1 ; r) DIBAL-H, CH_2Cl_2, –78 °C, 88%, (2 steps); s) CBr_4, Ph_3P, 2,6-lutidine, CH_3CN; t) Bu_3P, CH_3CN, 94% (2 steps); u) $LiCH_2S(O)CH_3$, toluene, then **XXIII**, –78 °C → 0 °C; v) DDQ, CH_2Cl_2-$NaHCO_3$ (0.5%) (9:1); w) Swern oxidation, 82% (3 steps); x) *t*BuOK, then **XXIII**, 80%; y) TBAF; z) Swern

[71] a) L. E. Overman, L. A. Robinson, J. Zablocki, *J. Am. Chem. Soc*. **1992**, *114*, 368-369; b) D. A. Evans, S. L. Bender, J. Morris, *J. Am. Chem. Soc.* **1988**, *110*, 2506-2526.

oxidation, 91% (2steps); aa) **IL**, $LiCH_2S(O)CH_3$, then **LVII**, toluene, –78 °C → RT, 72%; bb) PCC, CH_2Cl_2, 80%; cc) HF•pyridine, 74%; dd) **L**, *t*BuOK, then **LVII**, 90%; ee) H_2O, PPTS, acetone; ff) TPAP, CH_2Cl_2; gg) HF•pyridine, THF, 43% (3 steps).

In *Smith*'s approach, the major disconnections are maintained, but the partner functionalities are reversed. The phosphonium salt **L** (Scheme 3) obtained by *Crimmins* and co-workers for the *Wittig* reaction was functionalized as an aldehyde in *Smith*'s synthesis. The β-ketoamide **LIX** was prepared in three steps from acylated oxazolidinone **LVIII** using *Evans* strategy (Scheme 5).[72] The first aldol reaction occurred with complete diastereoselectivity in 88% yield, while for the second one a mixture *syn*/*anti* 4:1 in 65% yield was obtained due to mismatch problems. DIBAL-H reduction generated the cyclic hemiacetal **LX** which undergoes a *Wittig* reaction with (carbetoxyethylidene)triphenylphosporane (**XLVIII**) in a *E*/*Z* selectivity of 10:1. Selective TBS protection of diol **LXI** gave a poor yield (45%) and regioselectivity (10:1). A three step sequence furnished aldehyde **LXII**.

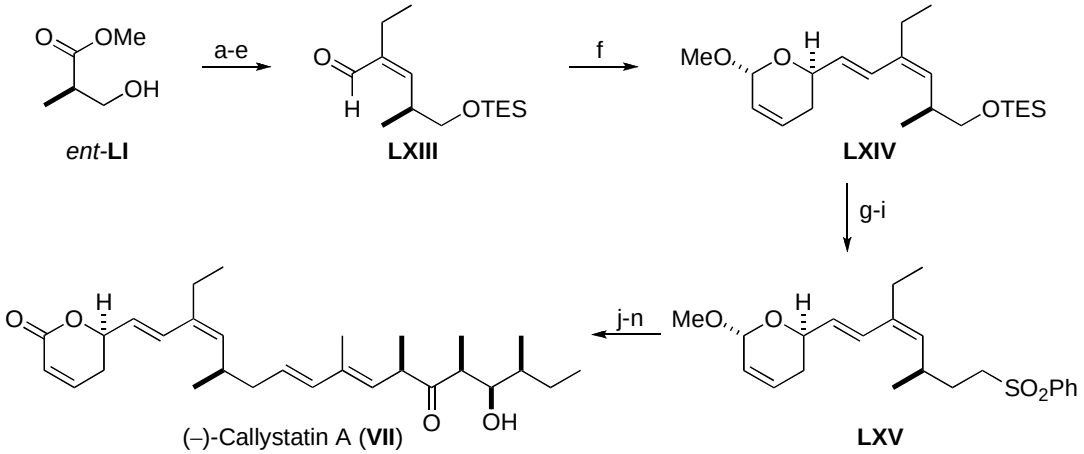


Scheme 5: a) Bu_2BOTf, Et_3N, propionaldehyde, 88%; b) SO_3•pyridine, Et_3N, DMSO, 85%; c) **XLIII**, *i*Pr_2EtN, $TiCl_4$, 65%; d) DIBAL–H, –78 ºC → –40 ºC, 65%; e) **XLVIII**, 96%, *E*/*Z* 10:1; f) CSA, $CHCl_3$, 92%; g) TBSOTf, 2,6-lutidine, 45%, 10:1; h) TMSOTf, CH_2Cl_2, –78 ºC, 90%; i) DIBAL-H, 87%; j) MnO_2, 79%.

Aldehyde **LXIII** was prepared in five steps from commercially available (*R*)-3-hydroxy isobutyric (*ent*-**LI**) acid *via* a *Still-Gennari* olefination affording a mixture of *Z*/*E* (8:1). *Julia-Kocienski* olefination between aldehyde **LXIII** and sulfone **XXXII** gave exclusively the *E* coupled product **LXIV**, but in poor yield (35%). Intermediate **LXIV** was deprotected and transformed into a phenyl sulfone derivative **LXV**, which was deprotonated with *n*BuLi and added to the aldehyde **LXII** (Scheme 6). The resulting alkoxy anion was trapped by the addition of Ac_2O to prepare the

[72] a) D. A. Evans, E. Vogel, J. V. Nelson, *J. Am. Chem. Soc.* **1979**, *101*, 6120-6123; b) D. A. Evans, J. V. Nelson, E. Vogel, T. R. Taber, *J. Am. Chem. Soc.* **1981**, *103*, 3099-3111.

intermediate for elimination. The mixture of acetates was treated with a sodium amalgam to furnish the product as a *E*/*Z* 3.5:1 mixture of olefins. Final modifications afforded (–)-callystatin A (**VII**) (Scheme 6). The reported strategy required 32 steps (longest linear sequence 15 steps). Some problems were encountered during the synthesis, like the poor yield in the Julia-*Kocienski* olefination as well as the scarce selectivity in the final coupling between the phenyl sulfone derivative **LXV** and the aldehyde **LXII**. Once again, the *Still-Gennari* coupling was found not to be completely selective and a *Z*/*E* 8:1 mixture was obtained.



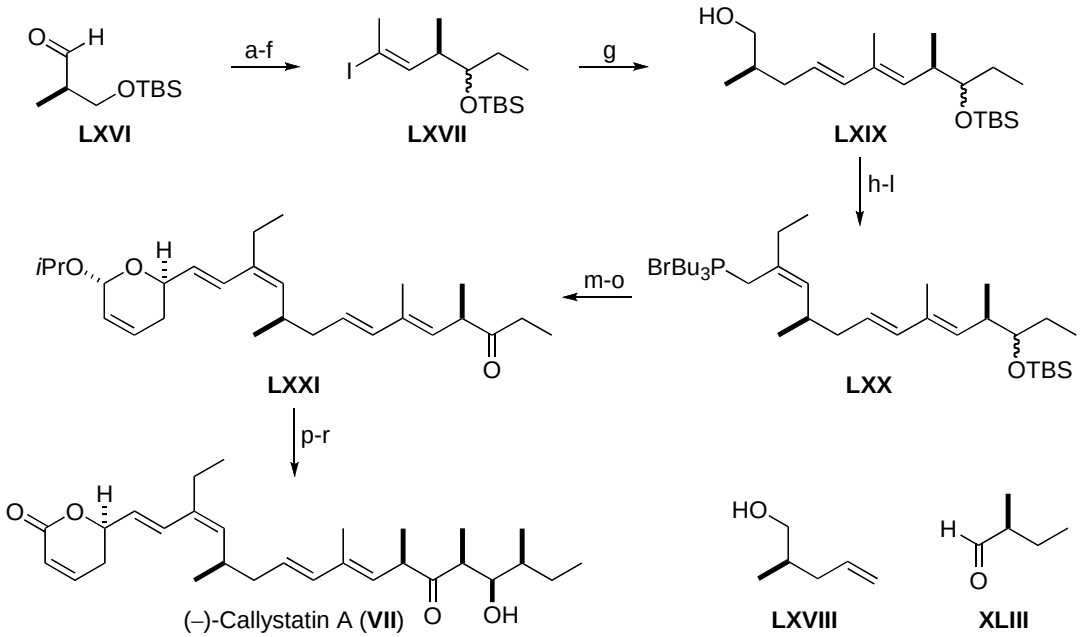
ent-**LI** **LXIII** **LXIV** **LXV**

Scheme 6: a) TESCl, imidazole, DMAP, 99%; b) DIBAL-H, 86%; c) $MeO_2CCH(Et)PO(OCH_2CF_3)_2$, 18-crown-6, KHMDS, THF, 0 ºC, 79%, *Z*/*E* 8:1; d) DIBAL-H, 88%; e) MnO_2, 88%; f) **XXXII**, NaHMDS, HMPA, DME, –78 ºC, 35%; g) PPTS, MeOH, 99%; h) Ph_3P, DEAD, MeI, 92%; i) $PhSO_2Me$, *n*BuLi, THF, toluene, HMPA, 65%; j) **LXV**, *n*BuLi, –78 ºC, **LXII**, then Ac_2O, 76%, *E*/*Z* 3.5:1; k) Na(Hg), K_2HPO_4, THF-MeOH, 92%, *E*/*Z* 3.5:1; l) HOAc, THF, H_2O, 72%; m) PCC, HOAc, 72%; n) HF•pyridine, 79%.

The *Kalesse* synthesis was set up in a way to avoid protecting group problems in the final steps. The synthesis started from aldehyde **LXVI**, which is readily available from (*R*)-(–)-3-hydroxyisobutyrate (*ent*-**LI**).[73] After five steps, the formation of the (*E*)-vinyl iodide compound **LXVII** *via* hydrozirconation showed problems of selectivity and a 3:1 mixture of regioisomers was obtained. Isolation of the (*E*)-vinyl iodide **LXVII** product and *Heck* coupling with alcohol **LXVIII**, itself prepared in two steps using Evans procedure,[71b] afforded the coupled product **LXIX** in 65% yield. The alcohol **LXIX** was subsequently transformed into the *Wittig* reagent **LXX** in five steps, once more *via* a *Still-Gennari* olefination affording the *Z*/*E* 8:1 mixture. The phosphonium salt **LXX** was reacted with the aldehyde **XXIII** *via* a *Wittig* reaction

[73] S. D. Burke, J. E. Cobb, K. Takeuchi, *J. Org. Chem.* **1990**, *55*, 2138-2151.

and the product deprotected and oxidized to afford the ethyl ketone **LXXI**. Aldol reaction between ethyl ketone **LXXI** and (*S*)-2-methylbutanal **XLIII** gave a mixture of *syn*/*anti* in a 2:1 ratio. Finally, formation of the lactol and oxidation with MnO_2 afforded (–)-callystatin A (**VII**) (Scheme 7). A synthesis of 28 steps was reported (longest linear sequence 21 steps). Some problems of selectivity were encountered during the synthesis, especially in the last aldol reaction, which gave a *syn*/*anti* 2:1 mixture of diastereoisomers and for the preparation of the (*E*)-vinyl iodide compound **LXVII**. Finally, the *Still-Gennari* olefination afforded a *Z*/*E* 8:1 mixture.

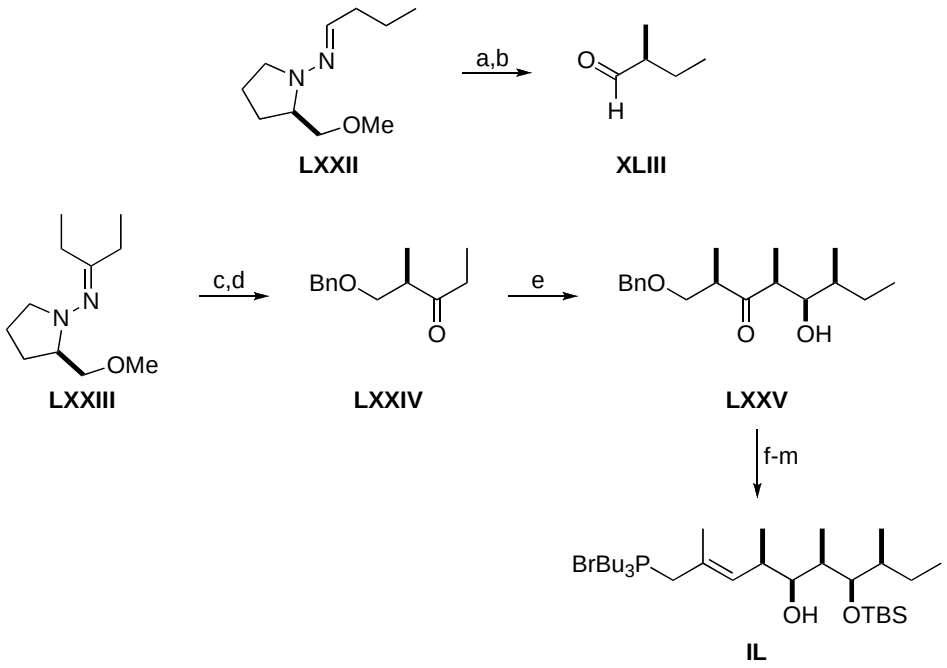


Scheme 7: a) EtMgBr, Et_2O; b) TBSOTf, 2,6-lutidine; c) CSA, acetone/H_2O, 72% (3 steps); d) Dess-Martin periodinane; e) CBr_4, Ph_3P then *n*BuLi, MeI, 80% (2 steps); f) $Cp_2ZrCl(H)$, I_2, 60%, 3:1 *d.r.*; g) **LXVIII**, $Pd(OAc)_2$, Ag(OAc), DMF, 65%; h) Dess-Martin periodinane; i) $EtO_2CCH(Et)PO(OCH_2CF_3)_2$, 18-crown-6, KHMDS, THF, 0 ºC, 65% (2 steps), *Z*/*E* 8:1; j) DIBAL-H, CH_2Cl_2, –78 ºC, 96%; k) CBr_4, Ph_3P, CH_3CN; l) Bu_3P, CH_3CN; m) *t*BuOK, **XXIII**, toluene, 0 ºC, 72% (3 steps); n) TBAF, THF; o) Swern oxidation, 73% (2 steps); p) LiHMDS, THF, –78 ºC then aldehyde **XLIII**, 63%, 2:1 *d.r.*; q) PPTS, acetone/H_2O 3:1; r) MnO_2, CH_2Cl_2, pyridine, 81% (2 steps).

The *Enders* synthesis is characterized by the SAMP/RAMP methodology.[74] The synthesis of the C(13)-C(22) fragment started with the methylation of the RAMP hydrazone of butanal **LXXII** and following ozonolysis afforded (*S*)-2-methylbutanal **XLIII**. In a similar way, the RAMP hydrazone of 3-pentanone **LXXIII** was alkylated with benzyloxymethyl chloride (BOMCl), followed by ozonolysis giving the chiral ketone **LXXIV** in 85% yield and 96% *e.e.*. Subsequent $Sn(OTf)_2$ mediated aldol

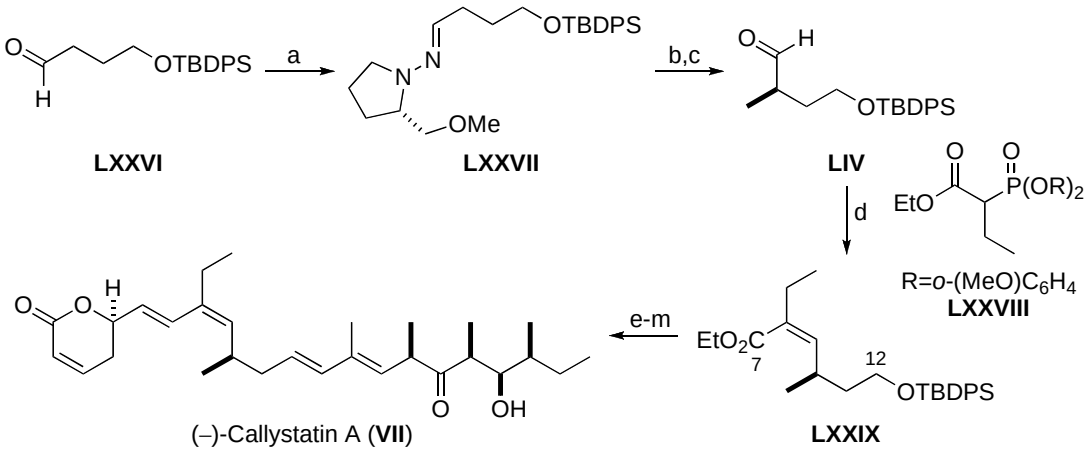
[74] D. Enders, *Asymmetric Synthesis, Vol. 3*, Eds J. D. Morrison, Academic Press, Orlando, **1984**.

reaction between aldehyde **XLIII** and chiral ketone **LXXIV** afforded the hydroxyketone **LXXV** in 87% yield and 94% *e.e.*. A further eight step sequence, including a diastereoselective reduction with DIBAL-H in 81% yield and 91:9 *d.r.*, gave the phosphonium salt **IL** (Scheme 8) already encountered in the Kobayashi synthesis (Scheme 3).



Scheme 8: a) LDA, THF, 0 °C, MeI, THF, –100 °C; b) O_3, CH_2Cl_2, –78 °C, 72% (2 steps); c) LDA, Et_2O, 0 °C, BOMCl, THF, –100 °C; d) O_3, CH_2Cl_2, –78 °C, 85% (2 steps), 96% *e.e.*; e) $Sn(OTf)_2$, Et_3N, CH_2Cl_2 then **XLIII**, –78 °C, 87%, 94% *e.e.*; f) TBSOTf, 2,6-lutidine; g) H_2, Pd/C; h) DIBAL-H, CH_2Cl_2, –78 °C, 81% (3 steps), 91:9 *d.r.*; i) Swern oxidation; j) **XLVIII**, 77% (2 steps); k) DIBAL-H, CH_2Cl_2, –78 °C; l) CBr_4, Ph_3P, 2,6-lutidine, CH_3CN, RT, 77% (2 steps); m) Bu_3P, CH_3CN.

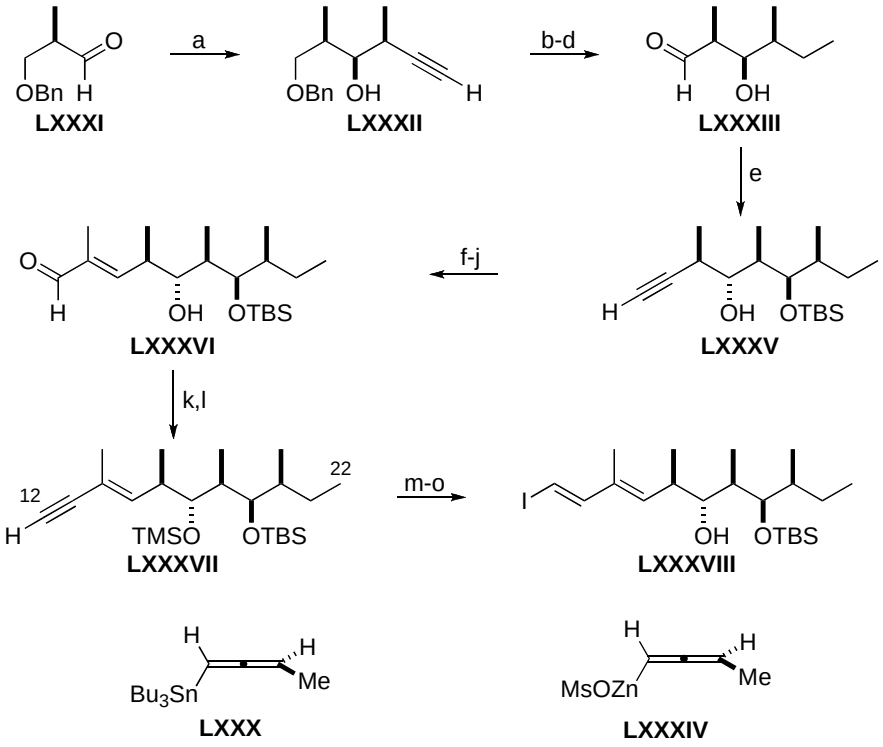
The synthesis of the C(7)-C(12) subunit started with the conversion of the aldehyde **LXXVI** to the corresponding SAMP hydrazone **LXXVII**. Deprotonation and methylation gave the alkylated product in excellent yield and in greater than 95:5 *d.r.*; which was converted to the corresponding aldehyde **LIV** *via* ozonolysis. *Horner-Wadsworth-Emmons* olefination with phosphonate **LXXVIII** afforded the product **LXXIX** in 91% yield and a *Z*/*E* ratio of 34:1. The remaining nine steps furnishing (–)-callystatin A (**VII**) (Scheme 9) followed a similar procedure to the *Crimmins* strategy (Scheme 4). This synthesis is quite long requiring 40 steps (longest linear sequence 16 steps), but no problems of selectivity were encountered.



Scheme 9: a) SAMP, 95%, *d.r.* > 95:5; b) LDA, THF, 0 °C, MeI, THF, –100 °C; c) O_3, CH_2Cl_2, –78 °C, 73% (2 steps); d) **LXXVIII**, NaH, THF, RT, 91%, *Z*/*E* 34:1; e) DIBAL-H, CH_2Cl_2; f) CBr_4, Ph_3P, CH_3CN, 92% (2 steps); g) Bu_3P, CH_3CN; h) *t*BuOK, toluene, 0 °C, then **XXIII**, 86%; i) TBAF, THF, j) Swern oxidation, 91% (2 steps); k) **IL**, $LiCH_2S(O)CH_3$, toluene, –78 °C, 71% (2 steps), l) PCC, HOAc, benzene; m) HF•pyridine, THF, 72% (2 steps).

The *Marshall* synthesis was characterized by the chiral allenylstannane methodology developed in the same group.[75] The synthesis of the C(12)-C(22) fragment started by the addition of allenylstannane **LXXX**[76] to the aldehyde **LXXXI**, itself readily available from (*R*)-(–)-3-hydroxyisobutyrate (*ent*-**LI**), affording alcohol **LXXXII** in a *syn* fashion. A three step sequence using standard transformations afforded the aldehyde **LXXXIII**, which was reacted with allenylzinc reagent **LXXXIV** to give alcohol **LXXXV**,[76] this time in *anti* orientation and as a single diasteroisomer. Subsequently, with a standard six step procedure, aldehyde **LXXXVI** was obtained and after protection of the secondary alcohol, *Seyferth-Gilbert* homologation[77] afforded terminal alkyne **LXXXVII** in 91% yield. The TMS group was removed and the vinyl iodide product **LXXXVIII** obtained *via* vinyl tin intermediate (Scheme 10).

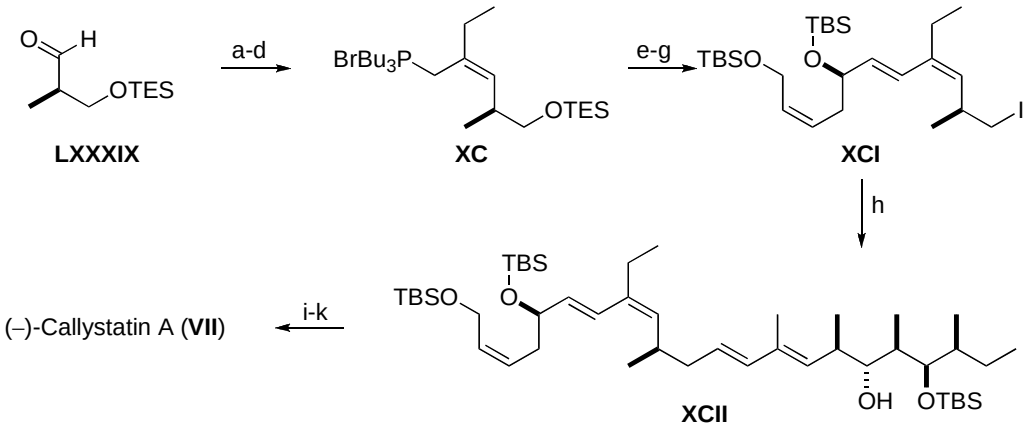
[75] a) J. A. Marshall, Z. H. Lu, B. A. Johns, *J. Org. Chem.* **1998**, *63*, 817-823; b) J. A. Marshall, N. D. Adams, *J. Org. Chem.* **1998**, *63*, 3812-3813.
[76] J. A. Marshall, *Chem. Rev.* **1996**, *96*, 31-47.
[77] J. C. Gilbert, U. Weorasooriya, *J. Org. Chem.* **1979**, *44*, 4997-4998.



Scheme 10: a) **LXXX**, BF_3•OEt_2, 82%; b) TBSOTf, 2,6-lutidine, 88%; c) H_2/Pd-C, EtOH, 80%; d) Swern oxidation, 99%; e) **LXXXIV**, 72%; f) H_2/Pd-$BaSO_4$, quinoline, toluene, 99%; g) O_3, CH_2Cl_2, Ph_3P, 82%; h) Ph_3P=C(Me)CO_2Et, 99%; i) DIBAL-H, 87%; j) MnO_2, CH_2Cl_2; k) TMSCl, Et_3N, DMAP, 82% (2 steps); l) $(MeO)_2P(O)CHN_2$, *t*BuOK, THF, –78 ºC → –30 ºC, 91%; m) PPTS, MeOH, 88%; n) Bu_3SnH, $PdCl_2$•$(Ph_3P)_2$, THF; o) I_2, CH_2Cl_2, 83% (2 steps).

The phosphonium salt **XC** was obtained following similar procedure as reported by *Kobayashi*,[56a] but starting from aldehyde **LXXXIX** derived from (*R*)-(–)-3-hydroxyisobutyrate (*ent*-**LI**). *Wittig* reaction between phosphonium salt **XC** and aldehyde **XXXVI** afforded the condensed product in good yield. Selective deprotection of the TES group and transformation of the primary alcohol into the iodide gave the alkyl iodide compound **XCI**. Palladium-catalyzed sp^3-sp^2 *Suzuki* coupling between the vinyl iodide compound **LXXXVIII** and the alkyl iodide **XCI** using *Johnson*'s protocol[78] afforded the coupled product **XCII** in 73% yield. Final modifications concluded the synthesis of (–)-callystatin A (**VII**) (Scheme 11). This synthesis required 39 steps (longest linear sequence 18 steps) and highlighted the efficiency of the allenyl-metal additions and of the sp^3-sp^2 *Suzuki* coupling.

[78] C. R. Johnson, M. P. Braun, *J. Am. Chem. Soc.* **1993**, *115*, 11014-11015.



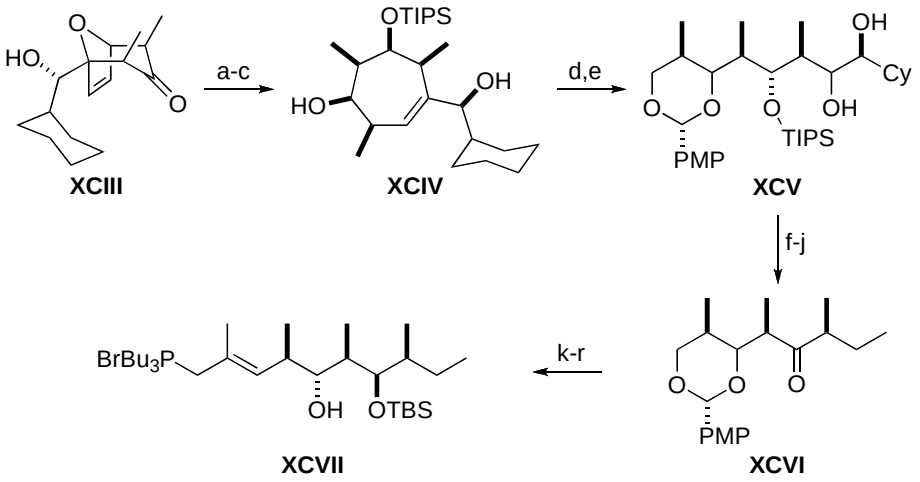
Scheme 11: a) $(CF_3CH_2O)_2P(O)CH(Et)CO_2Et$, *n*BuLi, 15-crown-5, 84%, 6-8:1 *d.r.*; b) DIBAL-H, 92%; c) CBr_4, Ph_3P, $NEt(iPr)_2$, 93%; d) Bu_3P, MeCN; e) *t*BuOK, 88% (2 steps); f) PPTS, MeOH-THF 9:1, 0 °C, 81%; g) I_2, Ph_3P, imidazole, benzene-Et_2O, 89%; h) **XCI**, *t*BuLi, 9-MeO-9-BBN then **LXXXVIII**, $Pd(dppf)Cl_2$, $AsPh_3$, Cs_2CO_3, DMF-H_2O, 73%; i) Dess-Martin periodinane, 70%; j) HF•Et_3N, 70%; (k) MnO_2, 66%.

The *Lautens* strategy was characterized by the use of [3.2.1]oxabicycle for the formation of polypropionates. The synthesis of the polyketidic chain started from the enantiomerically pure [3.2.1]oxabicycle **XCIII** obtained by a [4+3] cycloaddition[79] between 2,4-dibromo-3-pentanone and the 2-furylmethanol derivative.[80] Subsequent selective reduction of the ketone, TIPS protection of the alcohol and treatment with MeLi in presence of $CeCl_3$ afforded the cycloheptene **XCIV**. The ring was oxidatively opened *via* ozonolysis and the resulting aldehyde reduced with an oxidative work-up. The selective protection of the bis-diol was problematic and the protected diol **XCV** was formed in poor yield (52%). Diol cleavage under *Criegee* conditions[81] and further standard transformations afforded compound **XCVI**. Selective reduction of the ketone was achieved after screening several sets of conditions in 68% yield and more than 10:1 *d.r.*. The phosphonium salt **XCVII**, similar to **IL** in *Kobayashi*'s synthesis, was obtained in seven additional steps using standard reactions (Scheme 12).

[79] M. Lautens, R. Aspiotis, J. Colucci, *J. Am. Chem. Soc.* **1996**, *118*, 10930-10931.

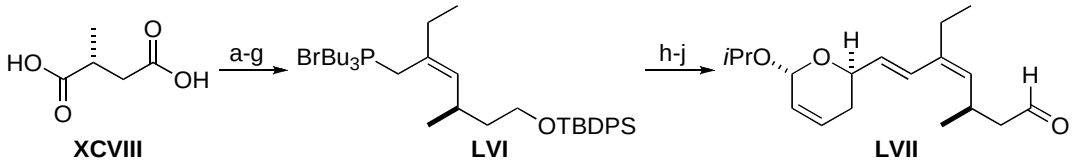
[80] T. Kametani, M. Tsubuki, Y. Tatsuzaki, T. Honda, *J. Chem. Soc., Perkin Trans. 1* **1990**, 639-646.

[81] R. Criegee, *Ber.* **1931**, *64*, 260-266.



Scheme 12: a) $LiBH_4$, THF, 0 °C → RT, 4 h, 87%; b) TIPSOTf, CH_2Cl_2, 2,6-lutidine, 0 °C, 90%; c) MeLi, $CeCl_3$, THF-Et_2O, –78 → –15 °C, 85%; d) O_3, CH_2Cl_2-MeOH, –78 °C, then $NaBH_4$, 69 h, 20 °C, 91%; e) 4-methoxybenzylidene, CSA (3 mol%), CH_2Cl_2 (0.7 M), 20 °C; f) $Pb(OAc)_4$, benzene-MeOH, 0 °C, 0.5 h, 97%; g) $Ph_3P{=}CH_2$, THF, –15 °C → RT, 18 h, 95%; h) 10% Pd/C, EtOAc, H_2, 0.5 h, RT, 99%; i) TBAF, THF, RT, 66 h, 99%; j) TPAP, NMO, CH_2Cl_2, 2.5 h, 97%; k) L-Selectride, toluene, 20 °C, 68%, *d.r.* > 10:1; l) TBSOTf, CH_2Cl_2, 2,6-lutidine, –15 °C, 2 h, 88%; m) $Pd(OH)_2$/C, H_2 (1 atm), *i*PrOH, 20 °C, 1 h, 97%; n) $(COCl)_2$, DMSO, CH_2Cl_2, –78 °C, Et_3N, 95%; o) $(Me)(Ph_3P{=})CCO_2Et$, CH_2Cl_2, reflux, 13.5 h, 78%; p) DIBAL-H, CH_2Cl_2, –78 °C, 10 min, 96%; q) Ph_3P, CBr_4, MeCN, 2,6-lutidine, RT, 5 min, 79%; r) Bu_3P, MeCN, 24 °C, 1 h.

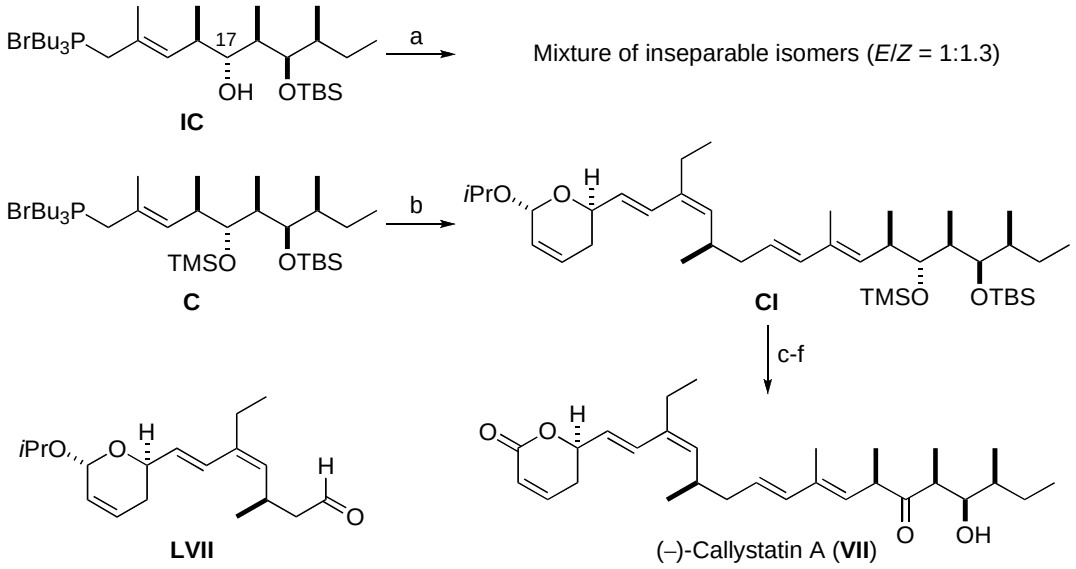
The C(7)-C(12) fragment in *Lautens* strategy was identical to the one previously synthesized by *Crimmins* and co-workers (Scheme 4). *Lautens* and co-workers started from commercially available (*R*)-α-methylsuccinic acid **XCVIII** and obtained phosphonium salt **LVI** in seven steps. Aldehyde **LVII** was formed in an identical fashion to *Crimmins via* a *Wittig* reaction between phosphonium salt **LVI** and the aldehyde **XXIII** (Scheme 13).



Scheme 13: a) $LiAlH_4$, THF, 0 °C → 24 °C, 17.5 h, 87%; b) TBDPSCl, DBU, DMF, –50 °C, 0.5 h, 61%; c) DMSO, CH_2Cl_2, $(COCl)_2$, –78 °C, Et_3N, 99%; d) $EtO_2CCH(Et)PO(OCH_2CF_3)_2$, KHMDS, 16-crown-6; e) DIBAL-H; f) PPh_3, CBr_4; g) PBu_3; h) **LVI**, *t*BuOK, then **XXIII**, 82% (5 steps); i) TBAF; j) Swern oxidation.

The final *Wittig* reaction between aldehyde **LVII** and phosphonium salt **IC** proved problematic with no *E*/*Z* selectivity and formation of an inseparable mixture of isomers. In their analogous reaction *Crimmins* and co-workers[56b] reported a complete *E* selectivity and the problem was explained by *Lautens* in the stereochemistry of

C(17) of the employed phosphonium salt **IC** (Scheme 14). The problem was solved by protecting the free hydroxy group and using the modified phosphonium salt **C**, which afforded product **CI** in high yield (94%) and a *E*/*Z* selectivity greater than 19:1. Hydrolysis of the *i*PrO-lactol proved to be challenging and in initial trials lactone ring opening affording the corresponding α,β-unsaturated aldehyde was observed. After optimization of the hydrolysis conditions to furnish the lactol, (–)-callystatin A (**VII**) was obtained in three further steps (Scheme 14). The target was achieved, but the synthesis was long and needed 45 steps (longest linear sequence 27 steps). Another weakness was the poor selectivity during the protection of the *bis*-diol where half of an advanced intermediate was lost. However, the [3.2.1]oxabycycle methodology enabled the 1,3,5-*syn*,*syn*-trimethyl arrangement required in the polyketidic chain to be obtained.

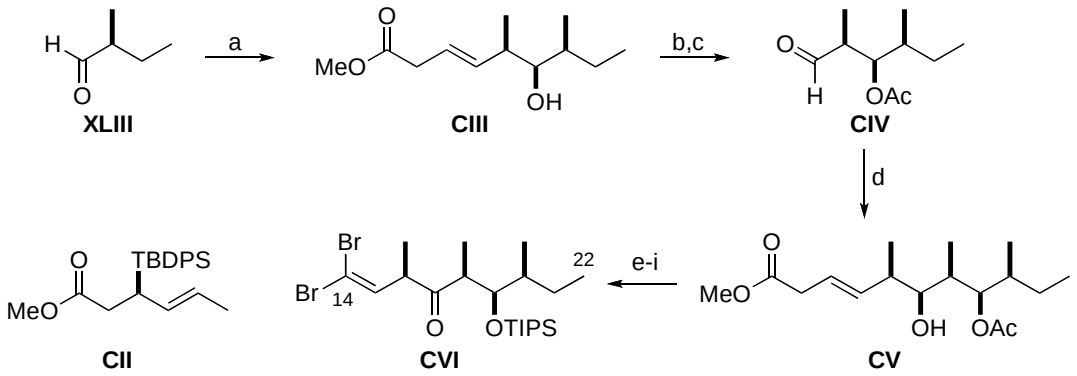


Scheme 14: a) **IC**, *t*BuOK, toluene-THF, 0 °C, then **LVII**, 92%, *E*/*Z* = 1:1.3; b) **C**, *t*BuOK, toluene-THF, 0 °C, then **LVII**, 94%, *E*/*Z* > 19:1; c) HOAc, THF-H_2O, 20 °C; d) MnO_2, CH_2Cl_2, 20 °C; e) Dess-Martin periodinane, CH_2Cl_2, 20 °C; f) HF•pyridine, pyridine, THF, 0 °C → 20 °C.

The *Panek* approach to the C(14)-C(22) fragment started from (*S*)-2-methylbutanal **XLIII** which, was treated with allylsilane **CII**[82] in presence of $TiCl_4$ to afford homoallylic alcohol **CIII** in 84% yield and 10:1 *d.r.*. Protection of the hydroxy group and subsequent ozonolysis gave aldehyde **CIV** which, was reacted with allylsilane **CII** in presence of $TiCl_4$ to give homoallylic alcohol **CV** in 20:1 *d.r.*.

[82] R. A. Ward, G. Procter, *Tetrahedron* **1995**, *51*, 12821-12836.

Subsequent standard transformations furnished the dibromo olefin **CVI** in five steps (Scheme 15).

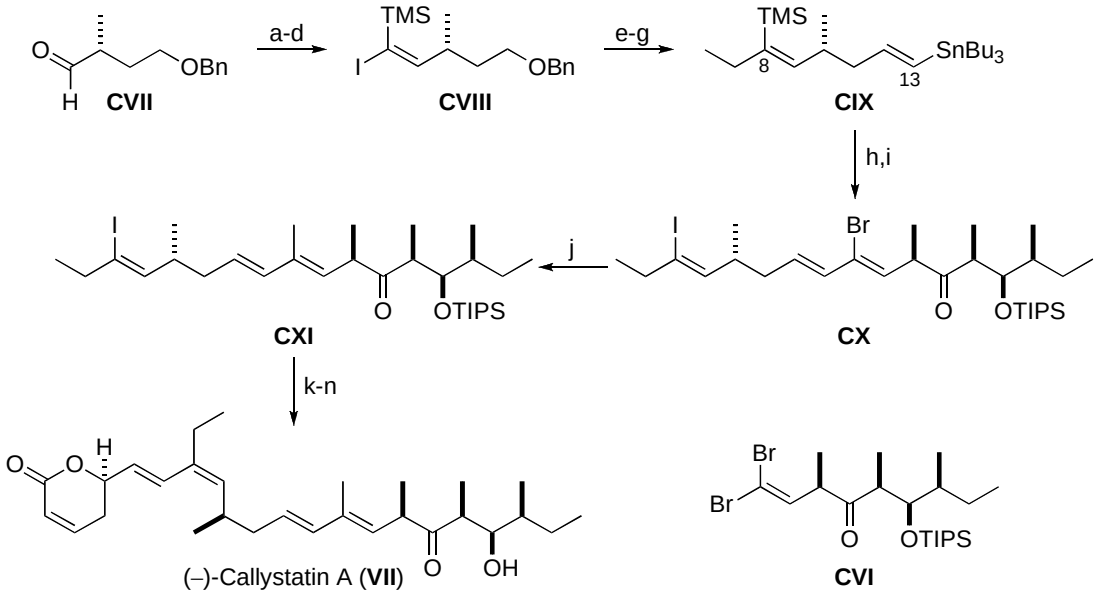


Scheme 15: a) **CII**, $TiCl_4$, CH_2Cl_2, –30 °C, 84%, 10:1 *d.r.* (crude); b) Ac_2O, pyridine, DMAP, CH_2Cl_2, RT, 99%; c) O_3, pyridine, MeOH/ CH_2Cl_2, –78 °C, then Me_2S; d) **CII**, $TiCl_4$, CH_2Cl_2, –30 °C, 68% (two steps), 20:1 *d.r.* (crude); e) O_3, pyr, MeOH/ CH_2Cl_2, –78 °C, then Me_2S; f) CBr_4, PPh_3, 2,6-lutidine, CH_2Cl_2, 0 °C → RT, 82% (two steps); g) K_2CO_3, MeOH, RT, 99%; h) TIPSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C, 90%; i) PCC, CH_2Cl_2, RT, 85%.

The synthesis of the C(8)-C(13) subunit began with aldehyde **CVII**, prepared following *Myers* procedure.[83] *Corey-Fuchs* reaction, *Fritsch-Buttenberg-Wiechell* rearrangement and TMS protection afforded the protected alkyne, which was treated with *Schwartz* reagent. The organozirconocene intermediate was then trapped with iodine to afford the vinyl iodide product **CVIII** in more than 20:1 *d.r.*. Palladium-catalyzed *Negishi* cross coupling installed the ethyl group at the C(8) position. Further transformations and final vinylstannylation afforded the *E*-vinyl stannane **CIX** in 68% yield and in *E*/*Z* 20:1 selectivity. Palladium-catalyzed *Stille* coupling between vinyl stannane **CIX** and dibromo olefin **CVI** afforded the *trans* coupled product in 92% yield and as a single isomer. Treatment of the coupled product with *N*-iodosuccinimide added the iodide at the C(8) position with retention of the stereochemistry to afford product **CX**. Subsequently, *Negishi* cross coupling in the presence of Me_2Zn occurred in modest yield (51%) and with a surprising selectivity for the bromide over the iodide. The vinyl iodide compound **CXI**, was coupled *via Negishi* cross coupling to the organozinc partner formed *in situ* by treatment of the terminal alkyne **XL** with *Schwartz* reagent and transmetallation to zinc. Final modifications gave (–)-callystatin A (**VII**) (Scheme 16). The target was achieved in

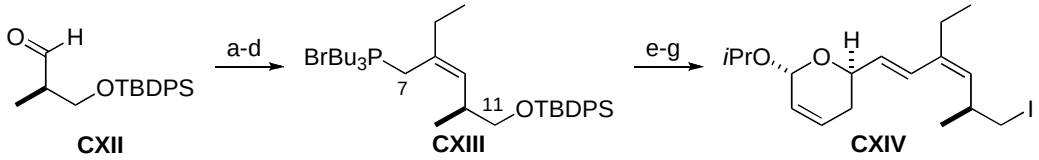
[83] A. G. Myers, B. H. Yang, H. Chen, L. McKinstry, D. J. Kopecky, J. L. Gleason, *J. Am. Chem. Soc.* **1997**, *119*, 6496-6511.

37 steps overall (longest linear sequence 18 steps), highlighting the use of chiral allylsilanes in total synthesis.



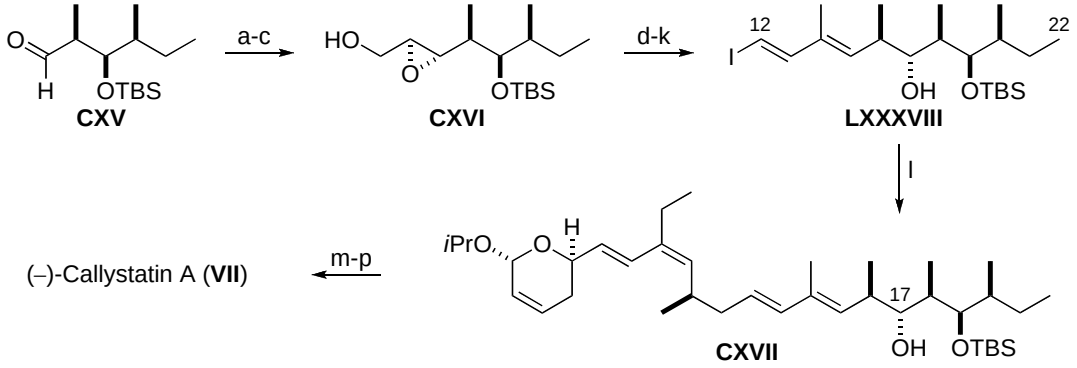
Scheme 16: a) CBr_4, PPh_3, 2,6-lutidine, CH_2Cl_2, 0 °C → RT, 96%; b) *n*BuLi, THF, –78 °C, then TMSCl, –78 °C → RT, 98%; c) Cp_2ZrHCl, THF, 50 °C, 1 h, then I_2, THF, RT, 89%, *d.r.* > 20:1 (crude); d) $ZnCl_2$, EtZnBr, $Pd(PPh_3)_4$, THF, RT, 96%; e) DDQ, CH_2Cl_2/H_2O, RT, 83%; f) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, –78 °C; 92%; g) $CrCl_2$, Bu_3SnCHI_2, DMF, 0 °C → RT, 68%, *E*/*Z* > 20:1; h) **CVI**, Pd_2dba_3, P(2-furyl)$_3$, toluene, 100 °C, 92%; i) NIS, EtCN, 84%; j) Me_2Zn, $Pd(tBu_3P)_2$, THF, 0 °C, 93%; k) i. **XL**, Cp_2ZrHCl, THF, RT, ii. $ZnCl_2$, THF, iii. **CXI**, $Pd(PPh_3)_4$, RT, 51%; l) AcOH, wet THF, RT; m) PDC, CH_2Cl_2, RT, 74% (two steps); n) HF• pyridine, THF, RT, 88%.

The *Dias* approach to the C(7)-C(11) fragment was identical to the protocol adopted by *Marshall*,[56f] but with a TBDPS protection instead of the TES group on the aldehyde **CXII**. In this case the *Still-Gennari* olefination gave better selectivity than in the previously discussed synthesis with a diastereomeric ratio greater than 93:7. The phosphonium salt **CXIII** was reacted with aldehyde **XXIII** *via* a *Wittig* reaction in good yield and more than 95:5 *d.r.*. The product was deprotected with TBAF and the hydroxy group replaced with iodide to afford the alkyl iodide product **CXIV** (Scheme 17).



Scheme 17: a) $EtO_2CCH(Et)PO(OCH_2CF_3)_2$, NaH, THF, 0 °C, 90%, *Z*/*E* > 93:7; b) DIBAL-H, CH_2Cl_2, –23 °C, 90%; c) CBr_4, PPh_3, CH_3CN, RT, 1 h, 95%; d) PBu_3, CH_3CN, RT, 96%; e) $LiCH_2S(O)CH_3$, toluene, then **XXIII**, –78 °C, 1 h, 82%, *E*/*Z* > 95:5; f) TBAF, THF, RT, 16 h; g) I_2, PPh_3, CH_2Cl_2, imidazole, RT, 1 h, 90% (2 steps).

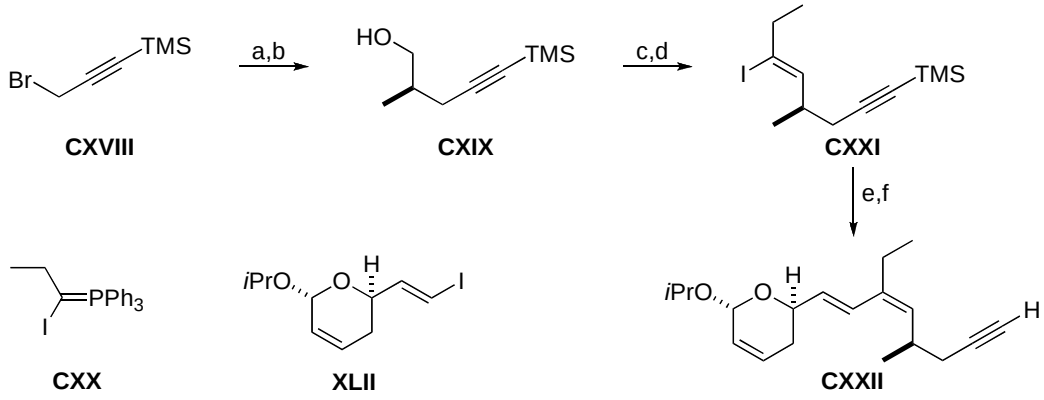
Concerning the C(12)-C(22) fragment, the aldehyde **CXV** was obtained following the strategy already adopted by *Kobayashi* (Scheme 3), based on *Evans* aldol reactions. The aldehyde **CXV** subsequently subjected to *Horner-Wadsworth-Emmons* olefination, DIBAL-H reduction and *m*CPBA-mediated epoxidation afforded alcohol **CXVI** in more than 95:5 *d.r.*. The product was then modified with an eight step sequence to the (*E*)-vinyl iodide compound **LXXXVIII**, including *Wittig* and *Takai* reaction in a *E*/*Z* ratio greater than 95:5. The vinyl iodide compound **LXXXVIII**, also an intermediate in the *Marshall* synthesis, undergoes palladium-catalyzed sp^3-sp^2 *Suzuki* cross coupling with the alkyl iodide **CXIV** to afford the coupled product **CXVII**. In addition to vinyl iodide **LXXXVIII**, the analogous fragment with the alcohol at C(17) protected with a TMS group was also prepared. However, cross-coupling with this bis-protected fragment resulted in a 34:66 mixture of protected/deprotected products due to loss of the TMS group under the reaction conditions. As reported by *Lautens* and co-workers,[56g] hydrolysis of the *i*PrO-lactol gave problems and the same α,β-unsaturated aldehyde was observed. After lactol formation an additional three steps gave (–)-callystatin A (**VII**) (Scheme 18). The target was obtained in 39 steps (longest linear sequence 20 steps). This synthesis did not present a new strategy, but highlights the utility of the sp^3-sp^2 *Suzuki* cross coupling. A second point to note is that the (*R)* configuration of C(17) gave problems during the *i*PrO-lactol hydrolysis affording the formation of side products. It appears that the distance between the hydroxy group at C(17) and the *i*PrO group at C(1) is too large to justify a possible influence between them. However, no groups have so far given an explanation for this result and it remains an experimental observation.



Scheme 18: a) $(EtO)_2POCH_2CO_2Me$, NaH, THF, RT, 12 h, 90%; b) DIBAL-H, CH_2Cl_2, –23 °C, 2 h, 90%; c) *m*CPBA, CH_2Cl_2, 0 °C, 1 h, 96%, *d.r.* > 95:5; d) Me_2CNLi_2, THF, –20 °C, 20 h, 90%; e) Swern oxidation; f) (carbethoxyethylidine)triphenylphosphorane, $ClCH_2CH_2Cl$, 15 h, 89% (2 steps); g) DIBAL-H, CH_2Cl_2, –23 °C, 2 h; h) MnO_2, CH_2Cl_2, RT, 2 h; i) TMSCl, Et_3N, DMAP, CH_2Cl_2, 0 °C, 2 h; j) CHI_3, $CrCl_2$, THF, RT, 1 h, 45% (4

steps), *E*/*Z* > 95:5; k) CSA (cat.), EtOH, RT, 12 h, 96%; l) **CXIV**, *t*BuLi, 9-MeO-9-BBN then **LXXXVIII**, Pd(dppf)Cl_2, $AsPh_3$, Cs_2CO_3, DMF-H_2O, RT, 15 h, 67%; m) AcOH, THF, H_2O, RT; n) MnO_2, CH_2Cl_2, RT, 20 h, 72%; o) Dess-Martin periodinane, CH_2Cl_2, RT, 1 h, 81%; p) HF•pyridine, pyridine, 0 °C → RT, 72 h, 77%.

The *Micalizio* synthesis of the C(8)-C(13) fragment started from TMS protected propargyl bromide **CXVIII** that was converted to alcohol **CXIX** *via* alkylation of the *Evans* auxiliary and reduction.[84] Oxidation of **CXIX** and subsequent reaction with the phosphor ylide **CXX**,[85] *in situ* prepared from commercially available *n*-propyltriphenyl phosphonium bromide, afforded the (*Z*)-vinyl iodide **CXXI** in poor yield (45%) and *E*/*Z* selectivity (1.7/1.0 in favor of the undesired isomer). The isomeric vinyl iodides mixture required a separation by HPLC methods to isolate the *Z* isomer. The (*Z*)-vinyl iodide **CXXI** was subjected to *Negishi* cross-coupling with vinyl iodide **XLII** and the coupled product deprotected furnishing terminal alkyne **CXXII** (Scheme 19).



Scheme 19: a) (4*S*)-(+)-4-benzyl-3-propionyl-2-oxazolidinone, NaHMDS, –78 °C; then **CXVIII**, 75%; b) $LiBH_4$, Et_2O, H_2O, 91%; c) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2; d) i. $Ph_3P(Pr)Br$, *n*BuLi, THF, ii. I_2, THF, iii. NaHMDS, iv. aldehyde addition, 45%, *E*/*Z* 1.7:1; e) **CXXI**, $ZnCl_2$, *t*BuLi, Et_2O; then **XLII**, $Pd(PPh_3)_4$, 72%; f) TBAF, THF, 83%.

Preparation of the C(14)-C(22) fragment began from the commercially available (*S*)-(–)-2-methyl-1-butanol (**CXXIII**) which was converted in three steps following the *Kobayashi* strategy[56a] to the *Weinreb* amide **CXXIV**. Further transformations and reaction with allenylsilane **CXXV**[86] afforded alcohol **CXXVI** in 7:1 *d.r.*. Titanium-mediated reductive cross-coupling between terminal alkyne **CXXII** and alcohol

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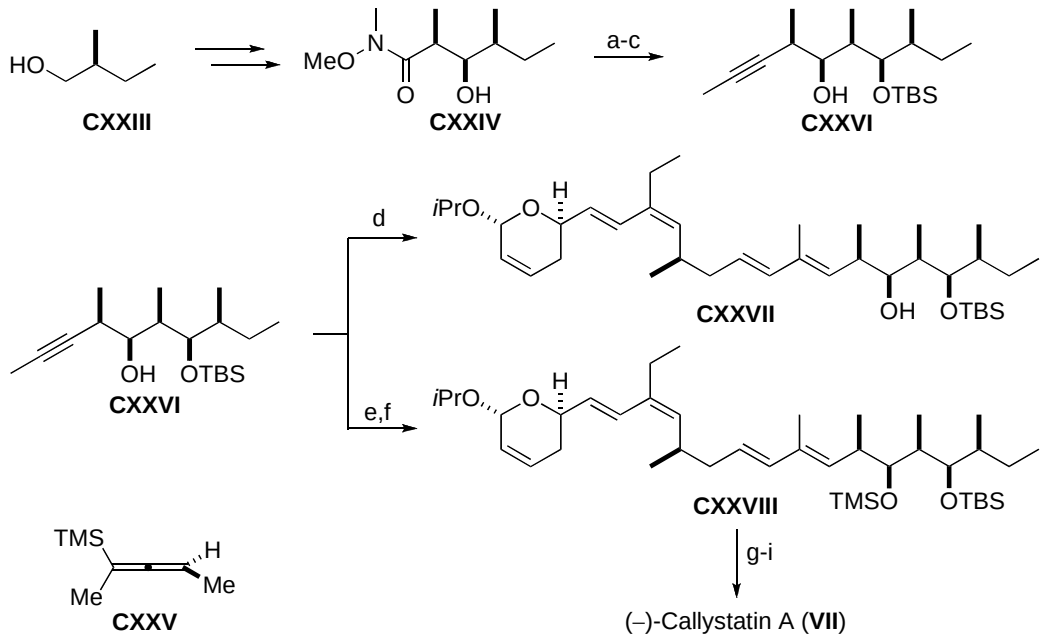
84 C. J. Forsyth, J. Xu, S. T. Nguyen, I. A. Samdal, L. R. Briggs, T. Rundberget, M. Sandvik, C. O. Miles, *J. Am. Chem. Soc.* **2006**, *128*, 15114-15116.

85 H. Arimoto, M. D. Kaufman, K. Kobayashi, Y. Qiu, A. B. Smith III, *Synlett* **1998**, 765-767.

86 a) M. J. C. Buckle, I. Fleming, *Tetrahedron Lett.* **1993**, *34*, 2383-2386, b) A. B. Bahadoor, A. Flyer, G. C. Micalizio, *J. Am. Chem. Soc.* **2005**, *127*, 3694-3695.

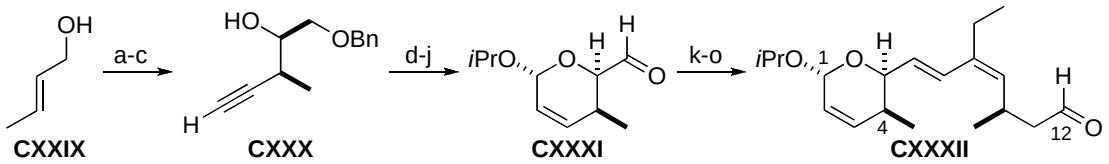
CXXVI furnished the coupled product **CXXVII** in poor yield (45%) and selectivity (3:1 *d.r.*). Better results were obtained by protecting the hydroxy group of **CXXVI**; the cross-coupling gave the coupled product **CXXVIII** in 75% yield and 5:1 *d.r.*. Final transformations furnished synthetic (–)-callystatin A (**VII**) (Scheme 20). The reported synthesis required 25 steps (longest linear sequence 11 steps) and highlighted the titanium-mediated reductive alkyne-alkyne cross-coupling. This strategy was the most straightforward, but suffered from poor selectivity in some steps. The preparation of the (*Z*)-vinyl iodide was achieved in 45% yield and a 1.7/1.0 *E*/*Z* ratio in favor of the undesired isomer and required HPLC separation. Moreover, the key step afforded in the best case a 5:1 mixture of isomers at an advanced point of the synthesis that also had to be separated by HPLC.



Scheme 20: a) TBSOTf, 2,6-lutidine, CH_2Cl_2, 82%; b) DIBAL-H, THF, –78 °C; c) **CXXV**, $TiCl_4$, CH_2Cl_2, –78 °C, 65% (2 steps), 7:1 *d.r.*; d) **CXXVI**, *n*BuLi, $ClTi(OiPr)_3$, *c*-C_5H_9MgCl, toluene, –78 °C → –30 °C, then **CXXII,** –78 °C → –30 °C, then NH_4Cl(aq.), 43%, 3:1 *d.r.*; e) TMSOTf, 2,6-lutidine, CH_2Cl_2, 89%; f) alkyne, $ClTi(OiPr)_3$, *c*-C_5H_9MgCl, toluene, –78 °C → –30 °C); then **CXXII,** –78 °C → –30 °C, then NH_4Cl(aq.), 75%, 5:1 *d.r.*; g) PPTS, H_2O, acetone, 71%; h) PCC, AcOH, 3 Å MS, benzene, 83%; i) HF•pyridine, pyridine, THF, 41%.

2.2.3.2. The Synthesis of Leptomycin B

Despite the popularity of leptomycin B as a tool in chemical biology, only a single total synthesis has been reported.[87] In this, *Kobayashi* and co-workers adopted the same strategy they used for the synthesis of callystatin (Scheme 1, 3 and 4).[56a] The same disconnections are maintained and the fragments adapted to the leptomycin structure. Leptomycin B differs from callystatin by the methyl group at C(4) and the α,β-unsaturated carboxylic acid at the end of the polyketide chain. The synthesis of the lactone fragment started from commercially available (*E*)-crotyl alcohol (**CXXIX**). Epoxidation using *Sharpless* methodology,[88] protection of the hydroxy group and selective epoxide ring opening afforded alcohol **CXXX**. The product was converted to the aldehyde **CXXXI** using standard chemistry. *Wittig* reaction between aldehyde **CXXXII** and phosphonim salt **LV**, already used in the synthesis of callystatin (Scheme 4), afforded the C(1)-C(12) fragment in modest yield, which was transformed to aldehyde **CXXXII** in five further steps (Scheme 21).



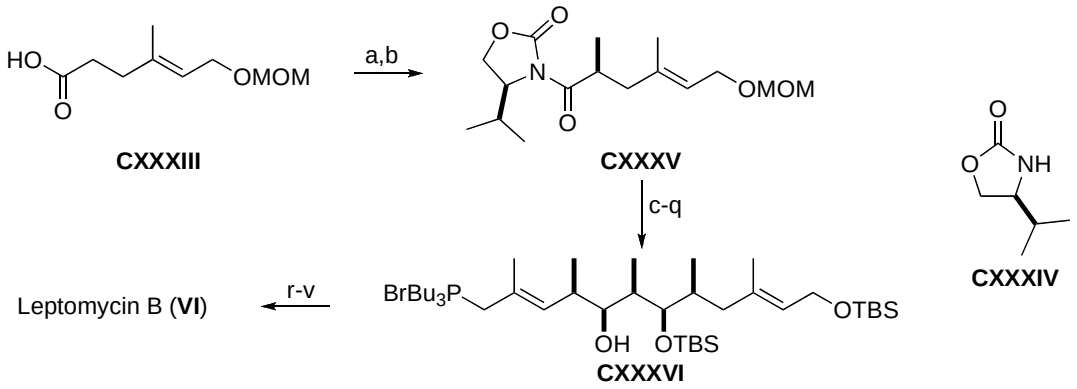
Scheme 21: a) (+)-DIPT, Ti(*i*PrO)$_4$, TBHP, CH_2Cl_2, –20 ºC, 75%, 96% *e.e.*; b) BnBr, NaH, TBAI, THF, 97%; c) Li-acetylide•EDA complex, HMPA, 66%; d) LDA, CO_2, THF, –78 ºC → –65 ºC; e) H_2, Pd/$BaSO_4$, quinoline, EtOH; f) benzene, reflux, 70% (3 steps); g) DIBAL-H, CH_2Cl_2, –78 ºC; h) *i*PrOH, PPTS, benzene, 55% (2 steps); i) Lithium di-*tert*-butylbiphenyl, THF, –78 ºC, 89%; j) Swern oxidation, 99%; k) **LV**, $LiCH_2S(O)CH_3$, toluene, –78 °C → 5 ºC, 59%; l) Dowex HCR-W2, acetone-H_2O, 40 ºC; m) Ag_2CO_3-Celite, benzene, 50 ºC, 94% (2 steps); n) DDQ, CH_2Cl_2-*t*BuOH-buffer 90:1:9, 89%; o) Dess-Martin periodinane, CH_2Cl_2, 99%.

The synthesis of the polyketidic chain started with carboxylic acid **CXXXIII**, which was obtained by ozonolysis of geraniol. Condensation of compound **CXXXIII** with lithium (*S*)-(–)-4-isopropyl-2-oxazolidinone (**CXXXIV**) and subsequent methylation gave product **CXXXV** in 11:1 *d.r.*. The synthesis of the phosphonium salt **CXXXVI** was carried out following a similar procedure to that employed by the same group in their synthesis of callystatin (Scheme 3). *Wittig* reaction between aldehyde **CXXXII** (Scheme 21) and phosphonium salt **CXXXVI** afforded the condensed (*E*)-product as a sole isomer, which after final transformations afforded

87 M. Kobayashi, W. Wang, Y. Tsutsui, M. Sugimoto, N. Murakami, *Tetrahedron Lett.* **1998**, *39*, 8291-8294.

88 T. Katsuki, K. B. Sharpless, *J. Am. Chem. Soc.* **1980**, *102*, 5974-5976.

leptomycin B (**VI**) (Scheme 22). LMB was synthesized in 40 steps (longest linear sequence 25 steps) and is comparable to the (–)-callystatin synthesis in terms of both length and strategy.



Scheme 22: a) PivCl, Et_3N, 76%; b) **CXXXIV**, LiHMDS, THF, –78 ºC, then MeI, –78 ºC → 0 ºC, 80%, 11:1 *d.r.*; c) Me_3Al, MeONHMe•HCl, CH_2Cl_2, –20 ºC → 0 ºC, 98%; d) DIBAL-H, CH_2Cl_2, –78 ºC, 98%; e) *n*BuBOTf, Et_3N, THF, –78 ºC → 0 ºC, 82%; f) Me_3Al, MeONHMe•HCl, CH_2Cl_2, –20 ºC → 0 ºC, 95%; g) TBSOTf, 2,6-lutidine, CH_2Cl_2, –20 ºC, 85%; h) DIBAL-H, THF, –78 ºC, 85%; i) *n*BuBOTf, Et_3N, THF, –78 ºC → 0 ºC, 94%; j) Me_3Al, MeONHMe•HCl, CH_2Cl_2, –20 ºC → RT, 99%; k) $LiAlH_4$, Et_2O, 0 ºC, 90%; l) $Ph_3P{=}CHCO_2Et$, toluene, 83%; m) DIBAL-H, CH_2Cl_2, –78 ºC, quant.; n) CBr_4, Ph_3P, 2,6-lutidine, MeCN, quant.; o) Me_2BBr, CH_2Cl_2, –78 ºC, 98%; p) TBSCl, imidazole, CH_2Cl_2, quant.; Bu_3P, MeCN, 93%; q) $LiCH_2S(O)CH_3$, –78 ºC → 5 ºC, 90%; r) Dess-Martin periodinane, 71%; s) HF•pyridine, pyridine; t) MnO_2, benzene; u) $NaClO_2$, NaH_2PO_4, H_2O_2, MeCN, 73% (2 steps).

2.2.3.3. The Syntheses of (+)- and (–)-Ratjadone

Two total syntheses of ratjadone have been reported. In their synthesis of (+)-ratjadone,[89] *Kalesse* and co-workers maintained the same principal disconnection as in the synthesis of callystatin. The C(7)-C(13) fragment started from alcohol **CXXXVII**, which was prepared in six steps from (*R*)-(–)-3-hydroxyisobutyrate (*ent*-**LI**).[90] Formation of the vinyl iodide *via* a *Negishi* carbometalation procedure,[91] followed by standard transformations using the same procedure adopted in the callystatin synthesis afforded phosphonium salt **CXXXVIII**. *Wittig* reaction between phosphonium salt **CXXXVIII** and the aldehyde **XXIII** already used in the callystatin synthesis (Scheme 7) afforded the vinyl iodide fragment **CXXIX** as a single isomer (Scheme 23).

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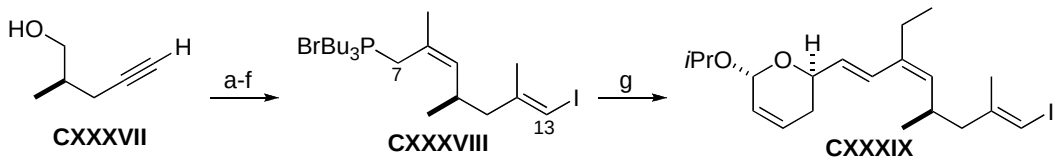
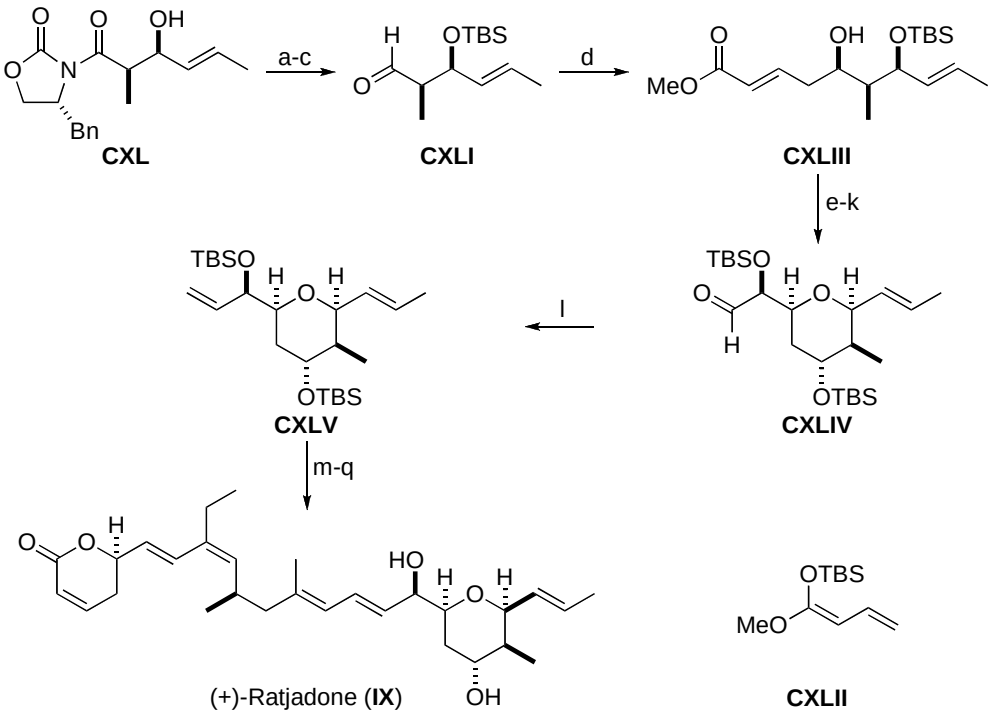
[89] M. Christmann, U. Bhatt, M. Quitschalle, E. Claus, M. Kalesse, *Angew. Chem., Int. Ed.* **2000**, *39*, 4364-4366.
[90] R. Baker, M. A. Brimble, *Tetrahedron Lett.* **1986**, *27*, 3311-3314.
[91] D. E. Van Horn, E. I. Negishi, *J. Am. Chem. Soc.* **1978**, *100*, 2252-2254.



Scheme 23: a) Cp_2ZrCl_2, $AlMe_3$, I_2, CH_2Cl_2, THF –15 ºC → 25 ºC, 83%; b) Dess-Martin periodinane, 81%; c) $(CF_3CH_2O)_2P(O)CHMeCO_2Et$, KHMDS, 18-crown-6, THF, –78 ºC, 85%; d) DIBAL-H, CH_2Cl_2, –78 ºC, 77%; e) CBr_4, Ph_3P, MeCN; f) Bu_3P, MeCN, 87% (2 steps); v) **XXIII**, *t*BuOK, toluene, 0 ºC, 89%.

The tetrahydropyranyl moiety was synthesized from the known aldol product **CXL**.[92] Conversion to the aldehyde **CXLI** and reaction with the ketene acetal **CXLII**[93] *via* vinylogous *Mukaiyama* aldol reaction[94] afforded alcohol **CXLIII** in 80% yield and 19:1 *d.r.*. Compound **CXLIII** was cyclized to give aldehyde **CXLIV**, which was converted by *Tebbe* olefination to the tetrahydropyrane moiety **CXLV**. Palladium-catalyzed *Heck* coupling between vinyl iodide **CXXXIX** and tetrahydropyrane moiety **CXLV** followed by standard transformations afforded (+)-ratjadone (**IX**) (Scheme 24). The target was achieved in 36 steps (longest linear sequence 19 steps) without any remarkable problems during the synthesis.



Scheme 24: a) MeONHMe•HCl, Me_3Al, CH_2Cl_2, –20 ºC → RT; b) TBSOTf, 2,6-lutidine, CH_2Cl_2, –78 ºC; c) DIBAL-H, CH_2Cl_2, –78 ºC, 83% (3 steps); d) **CXLII**, $B(C_6F_5)_3$, CH_2Cl_2-

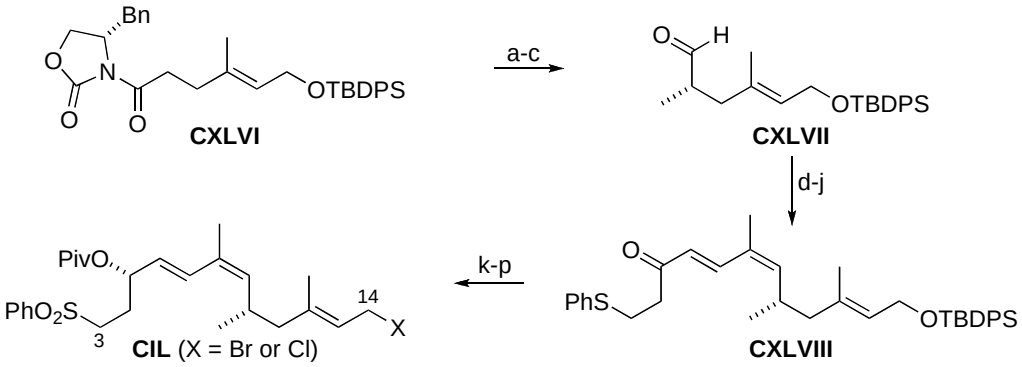
[92] D. A. Evans, D. L. Rieger, T. K. Jones, S. W. Kaldor, *J. Org. Chem.* **1990**, *55*, 6260-6268.

[93] R. V. Hoffman, H. O. Kim, *J. Org. Chem*. **1991**, *56*, 1014-1019.

[94] D. A. Evans, W. Cameron Black, *J. Am. Chem. Soc*. **1993**, *115*, 4497-4513.

Et_2O 9:1, –78 ºC, *d.r.* > 19:1, 80%; e) DIBAL-H, THF, –78 ºC; f) *m*CPBA, $NaHCO_3$, CH_2Cl_2, 0 ºC, 85% (2 steps); g) TBAF, THF, 88%; h) amberlyst-15, THF, 93%; i) TBSOTf, 2,6-lutidine, CH_2Cl_2, –78 ºC, 87%; j) $CHCl_3$•HCl, 97%; k) Dess-Martin periodinane, 92%; l) Tebbe olefination, THF, 0 ºC, 95%; m) **CXXXIX**, $Pd(OAc)_2$, Bu_4NBr, Cs_2CO_3, Et_3N, DMF, 80%; n) PPTS, H_2O, acetone, 83%; o) TBSOTf, 2,6-lutidine, CH_2Cl_2, 0 ºC, 83%; p) MnO_2, CH_2Cl_2, pyridine, 77%; q) HF•pyridine, pyridine, THF, 76%.

In the *Williams* and co-workers synthesis of (–)-ratjadone[95] the formation of the lactone was left until the end. The preparation of the C(3)-C(14) fragment started from the imide **CXLVI**.[87,96] Alkylation *via Evans* protocol[97] and standard transformations afforded aldehyde **CXLVII**, which was subjected to *Still-Gennari* olefination and transformed to ketone **CXLVIII** in six further steps. The ketone **CXLVIII** was reduced under *Terashima* conditions[98] employing (–)-*N*-methylephedrine, resulting in a 5:1 mixture of diastereoisomeric alcohols. The major product was then converted to the bromide **CIL** in five steps (Scheme 25). The bromide **CIL** (X = Br) was obtained as a 2.5:1 mixture with the allylic chloride (X = Cl) due to the presence of chlorine from the previous step, in which the compound was not purified.



Scheme 25: a) [ref. 87]; b) $LiBH_4$, MeOH, Et_2O, 87%; c) Swern oxidation, 94%; d) Still–Gennari olefination, 99%; e) DIBAL-H, 98%; f) Swern oxidation; g) CBr_4, Ph_3P, CH_2Cl_2; h) *n*BuLi, THF, –78 ºC, 85% (3 steps); i) $Cp_2Zr(H)Cl$, CH_2Cl_2, 23 ºC, then Me_2Zn, –65 ºC, then 3-(phenylthio)propanal, –65 ºC → 0 ºC, 92%; j) Dess-Martin periodinane, 64%; k) $LiAlH_4$, (–)-*N*-methylephedrine, EtNHPh, Et_2O, –78 ºC, 98%, 5:1 *d.r.*; l) $(NH_4)_6Mo_7O_{24}$, H_2O_2, EtOH (aq.), 0 ºC, 90%; m) PivCl, pyridine, DMAP, CH_2Cl_2, 100%; n) TBAF, THF, 100%; o) collidine, methanesulfonyl chloride, CH_2Cl_2, 0 °C, 3 h; p) LiBr, THF, RT, 15 min, 82%, mixture 2.5:1 bromide/chloride.

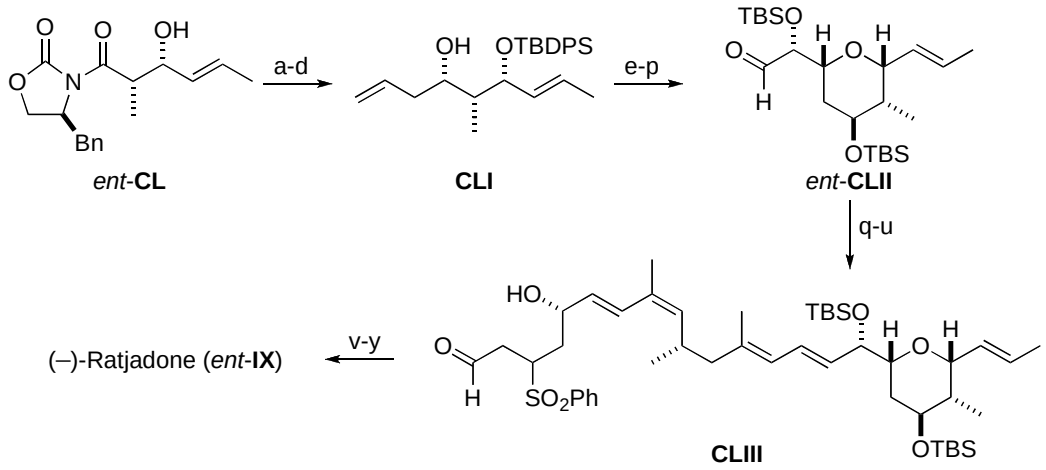
[95] D. R. Williams, D. C. Ihle, S. V. Plummer, *Org. Lett.* **2001**, *3*, 1383-1386.

[96] a) P. A. Plé, A. Hamon, G. Jones, *Tetrahedron* **1997**, *53*, 3395-3400.

[97] D. A. Evans, M. D. Ennis, D. J. Mathre, *J. Am. Chem. Soc.* **1982**, *104*, 1737-1739.

[98] a) S. Terashima, N. Tanno, K. Koga, *J. Chem. Soc., Chem. Commun.* **1980**, 1026-1027; b) S. Terashima, N. Tanno, K. Koga, *Chem. Lett.* **1980**, 981-984.

The synthesis of the tetrahydropyranyl fragment by *Williams* and co-workers followed a similar procedure to that of *Kalesse* and co-workers and also started from the known aldol product *ent*-**CL**. The major difference was the use of allyl Ipc_2B(allyl)borane instead of the vinylougous *Mukayama* aldol reaction to afford compound **CLI** in good yield and 94:6 *d.r.*. Aldehyde *ent*-**CLII** was obtained in further twelve steps using standard transformations. The bromide **CIL** was transformed *in situ* into the phosphonium salt and reacted with aldehyde *ent*-**CLII** to afford the coupled product in 72% yield and in *E*/*Z* 16:1 ratio. Further transformations and *Ley* oxidation[99] of alcohol **CLIII** gave the saturated lactone, which after elimination and removal of the protecting group afforded (–)-ratjadone (*ent*-**IX**) (Scheme 26). The target was achieved in 48 steps (longest linear sequence 30 steps), a longer sequence when compared with the *Kalesse* synthesis of (+)-ratjadone.[89] The large difference in the number of steps is principally due to the long protocol adopted for the formation of the lactone cycle that in the *Kalesse* synthesis was obtained in only four steps *via* a *Diels-Alder* reaction.

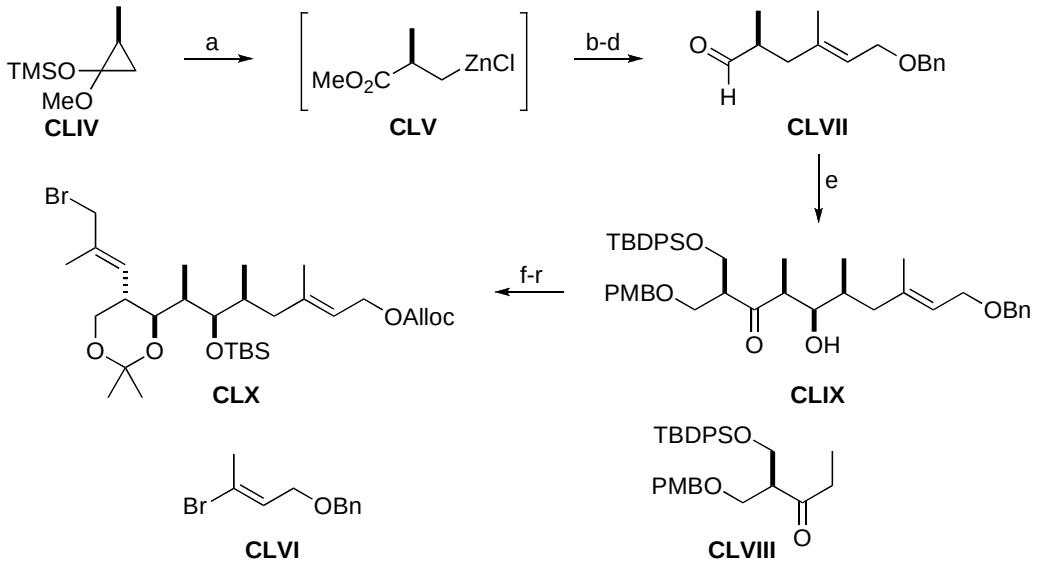


Scheme 26: a) TBDPSCl, imidazole, CH_2Cl_2; b) $LiBH_4$, Et_2O, H_2O, –20 °C → 5 °C, 87% (2 steps); c) Swern oxidation, 98%; d) *B*-allyldiisocamphenylborane, Et_2O, –78 °C, 91%, 94:6 *d.r.*; e) PMB trichloroacetimidate, CSA, CH_2Cl_2, 67%; f) AD-mix-α, *t*BuOH, H_2O, then $NaIO_4$, THF (aq.), quant.; g) $Ph_3P{=}CHCO_2Me$, CH_2Cl_2, 88%; h) DIBAL-H, CH_2Cl_2, –78 °C, 96%; i) (+)-DET, Ti(*i*PrO)$_4$, 4 Å MS, TBHP, CH_2Cl_2, 98%; j) PivCl, pyridine, CH_2Cl_2, 95%; k) TBAF, THF, 40 °C, 82%; l) CSA, CH_2Cl_2, 90%; m) CAN, MeCN, H_2O, quant.; n) TBSCl, imidazole, DMAP, DMF, 91%; o) DIBAL-H, CH_2Cl_2, –78 °C, 99%; p) Dess-Martin periodinane, 93%; q) **CIL**, PBu_3, 48 h, then *ent*-**CLII**, toluene, 0 °C, then *t*BuOK, THF, 72%, *E*/*Z* 16:1; r) DIBAL-H, CH_2Cl_2, –78 °C, 89%; s) TESCl, pyridine, CH_2Cl_2, 94%; t) *n*BuLi, THF, HMPA, then ethylene oxide, 78%; u) Dess-Martin periodinane; v) PPTS, EtOH, 0 °C; w) TPAP, NMO, 4 Å MS, 86% (3 steps); x) DBU, toluene, 87%; y) HF•pyridine, pyridine, THF, 76%.

[99] W. P. Griffith, S. V. Ley, G. P. Whitcombe, A. D. White, *J. Chem. Soc., Chem. Commun.* **1987**, 1625-1627.

2.2.3.4. The Synthesis of (–)-Kazusamycin A

The only synthesis of (–)-kazusamycin A was reported by *Kuwajima* and co-workers.[100] The synthesis of the polyketide chain started with the palladium catalyzed cross coupling between zinc homoenolate **CLV**, derived from cyclopropane **CLIV**,[101] and vinyl bromide **CLVI**.[102] The coupled product was obtained in more than 99% *e.e.* and converted into aldehyde **CLVII**, which underwent Sn-mediated aldol reaction with ketone **CLVIII**[103] to afford alcohol **CLIX** in 93:7 *d.r.*. The product was converted into bromide **CLX** in an additional thirteen steps employing standard chemistry (Scheme 27).



Scheme 27: a) $ZnCl_2$, Et_2O; b) $PdCl_2[P(o\text{-}Tol)_3]_2$ (2 mol %), **CLVI**, THF, 68%, *e.e.* > 99%; c) DIBAL-H, CH_2Cl_2, –78 → 45 °C, 96%; d) Dess-Martin periodinane, CH_2Cl_2, quant.; e) **CLVII**, $Sn(OTf)_2$, Et_2N, CH_2Cl_2, –78 °C, then **CLVII**, 77%, 93% *d.r.*; f) Et_2BOMe, $NaBH_4$, THF-MeOH, –78 °C, 78%; g) TBAF, THF, 92%; h) $Me_2C(OMe)_2$, PPTS, acetone, 86%; i) TBSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C, 94%; j) DDQ, CH_2Cl_2, 0 °C, 68%; k) Dess-Martin periodinane, CH_2Cl_2; l) $Ph_3P{=}CMeCO_2Et$, 69% (two steps); m) DIBAL-H, CH_2Cl_2, –78 °C, 89%; n) TIPSCl, imidazole, DMF, 96%; o) Na, liq. NH_3-THF, –78 °C, quant.; p) AllocCl, pyridine, THF, 96%; q) TBAF, THF, 98%; r) Ph_3P, CBr_4, CH_2Cl_2, 94%.

The preparation of the lactone moiety started from the acylated *Evans* auxiliary *ent*-**LVIII**. The unsaturated ester **CLXI** was obtained in seven steps *via* standard transformations and then deprotected with acidic resin, cyclized to gave α,β-unsaturated lactone and converted into aldehyde **CLXII**. This aldehyde was also an

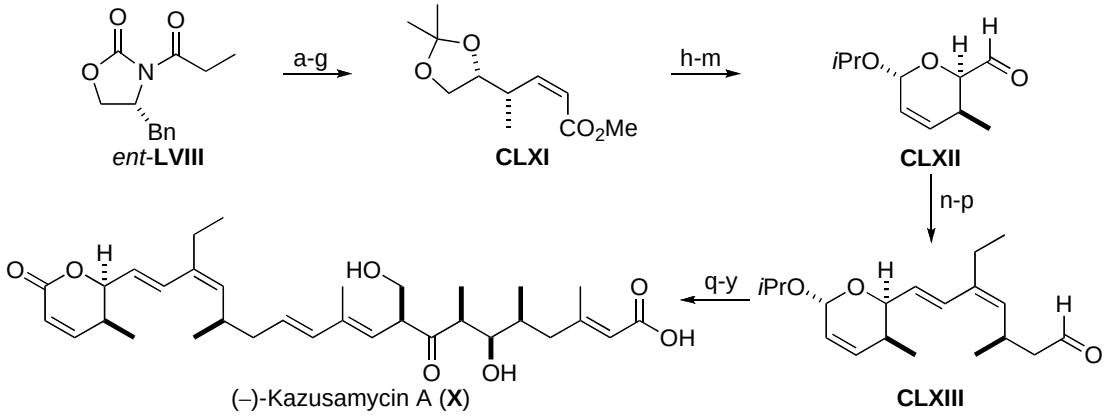
100 N. Arai, N. Chikaraishi, S. Omura, I. Kuwajima, *Org. Lett.* **2004**, *6*, 2845-2848.

101 E. Nakamura, K. Sekiya, I. Kuwajima, *Tetrahedron Lett.* **1987**, *28*, 337-340.

102 E. J. Corey, M. G. Bock, A. P. Kozikowski, *Tetrahedron Lett.* **1978**, *No. 12*, 1051-1054.

103 a) G. Egri, E. Fogassy, L. Novák, L. Poppe, *Tetrahedron: Asymmetry* **1997**, *8*, 547-557, b) S. Zhou, H. Chen, W. Liao, S. H. Chen, G. Li, R. Ando, I. Kuwajima, *Tetrahedron Lett.* **2005**, *46*, 6341-6344.

intermediate in the LMB synthesis of *Kobayashi* and co-workers (Scheme 21).[87] *Wittig* reaction between aldehyde **CLXII** and phosphonium salt **LVI**[104] afforded the coupled product in *E*/*Z* 7:1 ratio, the protected alcohol present in this intermediate was converted to the aldehyde **CLXIII**. A second *Wittig* reaction, between aldehyde **CLXIII** and phosphonium salt **CLX**, and final modifications afforded (–)-kazusamycin A (**X**) (Scheme 28). The target was achieved in 56 steps (longest linear sequence 33 steps). The synthesis wanted to showcase the efficiency of the *Paterson*'s aldol methodology,[105] unfortunately preparation of ketone **CLVIII** required ten steps increasing the synthesis length. Moreover, preparation of the *i*PrO-lactol fragment **CLXIII** was also long requiring a thirteen step sequence.



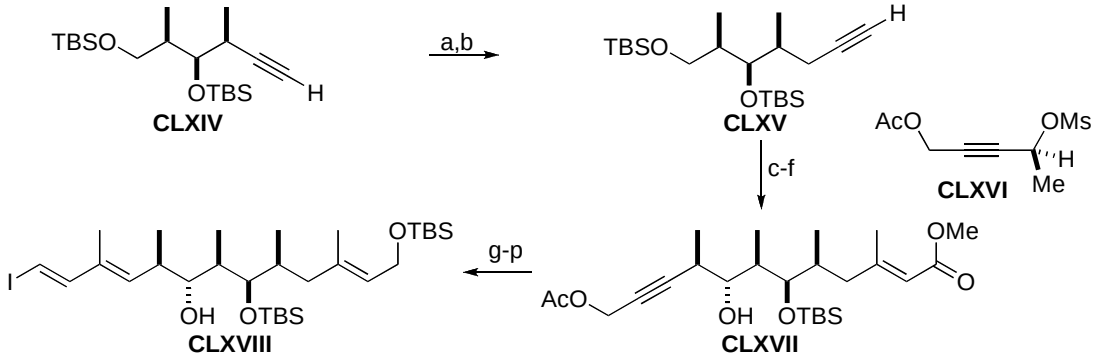
Scheme 28: a) Bu_2BOTf, Et_3N, $BnOCH_2CHO$, CH_2Cl_2, –78 °C → 0 °C, 96%, *d.r.* > 99:1; b) H_2, Pd/C, PPTS, $Me_2C(OMe)_2$, acetone, 90%; c) $LiBH_4$, MeOH, 0 °C, 95%; d) Dess-Martin periodinane, CH_2Cl_2; e) Ph_3P, CBr_4, Zn, CH_2Cl_2, 60% (2 steps); f) *n*BuLi, $ClCO_2Me$, THF, –78 °C → RT, 93%; g) H_2, Lindlar catalyst, MeOH, 96%; h) Dowex 50WX8, MeOH, then Amberlyst 15, CH_2Cl_2; i) TBDPSCl, imidazole, DMF, 57% (2 steps); j) DIBAL-H, CH_2Cl_2, –78 °C, 82%; k) PPTS, *i*PrOH, benzene, 85%; l) TBAF, THF, 85%; m) $(COCl)_2$, DMSO, CH_2Cl_2, –78 °C, then Et_3N, –78 °C → RT, 98%; n) **LVI**, Bu_3P, CH_3CN, then **CLXII**, *t*BuOK, toluene-THF, 0 °C, 91%, *E*/*Z* 7:1; o) TBAF, THF, 99%; p) $(COCl)_2$, DMSO, CH_2Cl_2, –78 °C, then Et_3N, –78 °C → RT, 92%; q) **CLX**, Bu_3P, CH_3CN, then **CLXIII**, *t*BuOK, 83%; r) PPTS, MeOH, 84% (3 cycles); s) TIPSCl, imidazole, DMF, 95%; t) Dess-Martin periodinane, CH_2Cl_2, 95%; u) PPTS, acetone (aq.), 91% (3 cycles); v) $Pd(PPh_3)_4$, dimedone, THF, 96%; w) MnO_2, CH_2Cl_2, 49%; x) $NaClO_2$, 2-methyl-2-butene, *t*BuOH (aq.), 80%; y) HF•pyridine, pyridine, THF, 74%.

[104] Prepared following the Kobayashi procedure in the synthesis of callystatin.

[105] I. Paterson, *Pure Appl. Chem.* **1992**, *64*, 1821-1830.

2.2.3.5. The Synthesis of Leptofuranin D

Marshall and co-workers published the only synthesis of leptofuranin D.[106] Having already synthesized callystatin, their synthesis contains a similar sequence of reactions and intermediates. The synthesis of the polyketide chain began from alkyne **CLXIV**[107] that was hydroborated and treated with the *Ohira* reagent[108] to afford alkyne **CLXV**. Water-accelerated carboalumination of the alkyne using *Wipf* conditions,[109] followed by standard transformations and a palladium catalyzed coupling of the allenylzinc reagent (generated from (*S*)-propargylic mesylate **CLXVI**[76]) gave the *anti* adduct **CLXVII** as a 9:1 mixture of diastereoisomers. A subsequent ten step procedure furnished the vinyl iodide compound **CLXVIII** (Scheme 29).



Scheme 29: a) Cy_2BH, then NaOH, H_2O_2, 95%; b) $MeCOC(N_2)PO(OMe)_2$, K_2CO_3, 75% (2 steps); c) Cp_2ZrCl_2, $AlMe_3$, H_2O, then $ClCO_2Me$, 64%; d) PPTS, MeOH, 91%; e) Swern oxidation, 97%; f) **CLXVI**, $Pd(OAc)_2$, PPh_3, Et_2Zn, 72%, 9:1 *d.r.*; g) Bu_3SnH, THF, $Pd(PPh_3)_2Cl_2$; h) I_2, CH_2Cl_2, 82% (2 steps); i) TMSC≡CH, $Pd(PPh_3)_2Cl_2$, CuI, Et_3N, 87%; j) NaOH, MeOH-THF, 90%; k) MsCl, Et_3N, CH_2Cl_2; l) LiBr, 2-butanone; m) $LiBEt_3H$, THF, 78% (3 steps); n) TBSCl, imidazole, CH_2Cl_2, 82%; o) Bu_3SnH, $Pd(PPh_3)_2Cl_2$, THF; p) I_2, CH_2Cl_2, 82% (2 steps).

The synthesis of the C(1)-C(11) fragment started from monoprotected ethylene glycol **CLXIX**. Oxidation and addition of a chiral allenylstannane **LXXX**[56f] gave alcohol **CLXX** as a *syn*/*anti* 83:17 mixture of diastereoisomers. This intermediate was transformed to alkyl iodide **CLXXI** and coupled with vinyl iodide compound **CLXVIII** following the same protocol adopted during the synthesis of callystatin to afford leptofuranin D as a mixture 1:1 of inseparable isomers at C(22) (Scheme 30).

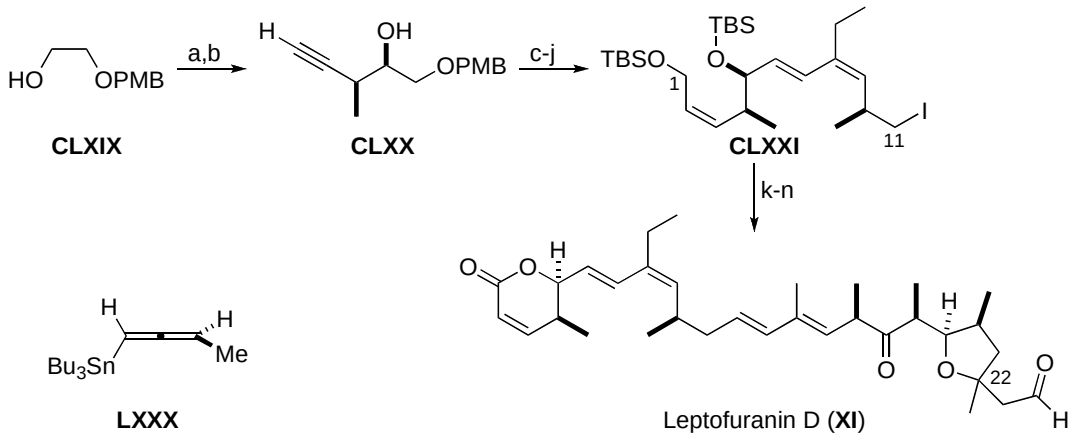
106 J. A. Marshall, G. M. Schaaf, *J. Org. Chem.* **2003**, *68*, 7428-7432.

107 Prepared following the Marshall procedure in the synthesis of Callystatin.

108 S. Ohira, *Synth. Commun.* **1989**, *19*, 561-564.

109 P. Wipf, S. Lim, *Angew. Chem., Int. Ed.* **1993**, *32*, 1068-1071.

The target was achieved in 39 steps (longest linear sequence 25 steps) and similarly to their synthesis of callystatin, the use of chiral allenyl-metal reagents was highlighted. The absolute stereochemistry of all the stereogenic centers were elucidated except that at C(22), which remains unknown.



Scheme 30: a) Swern oxidation, 70%; b) **LXXX**, $MgBr_2$, 85%, 83:17 *d.r.*; c) *n*BuLi, $(CH_2O)_n$, THF; d) TBSOTf, 2,6-lutidine, CH_2Cl_2, 83% (2 steps); e) H_2/Pd-$BaSO_4$ (5%), quinoline, toluene, 90%; f) DDQ, CH_2Cl_2, pH = 7, 91%; g) Swern oxidation, 98%; h) **XC**, *t*BuOK, toluene, 85%; i) PPTS, MeOH-THF, 74%; j) I_2, PPh_3, imidazole, 94%; k) **CLXXI**, *t*BuLi, Et_2O, 9-MeO-9-BBN, then **CLXVIII**, Pd(dppf)Cl_2, $AsPh_3$, Cs_2CO_3, DMF-H_2O, 82%; l) Dess-Martin periodinane, 71%; m) HF•Et_3N, 74%; n) MnO_2, CH_2Cl_2, 43%, 1:1 *d.r.*.

2.2.3.6. The Synthesis of Leptostatin

Marshall and co-workers reported the synthesis of leptostatin (**VIIIb**) together with three diastereoisomers at C(4) and C(5).[110] The adopted strategy required no new chemistry and simply combined a fragment from the synthesis of callystatin (**LXXXVIII**, Scheme 10) with a fragment from the synthesis of leptostatin (**CLXXI**, Scheme 30). The only major change in the procedure was the use of the pivaloyl protected chiral allenylstannane **CLXXIII** instead of the chiral allenylstannane **LXXX** (Scheme 10).[76] The alcohol **CLXXIV** was obtained as a 3:1 mixture of *syn*/*anti* diastereoisomers. After standard modifications, the polyketide chain **LXXXVIII** of callystatin and the alkyl iodide fragment **CLXXI** of leptofuranin were combined *via* sp^3-sp^2 *Suzuki* cross coupling affording leptostatin (**VIIIb**) (Scheme 31). This synthesis required 43 steps (longest linear sequence 22 steps) and is similar in length and strategy to the synthesis of callystatin by the same group.

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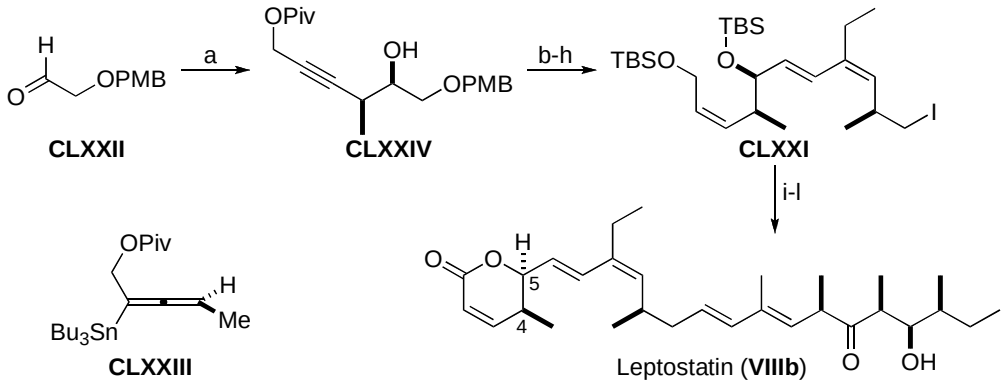
[110] J. A. Marshall, A. M. Mikowski, M. P. Bourbeau, G. M. Schaaf, F. Valeriote, *Bioorg. Med. Chem. Lett.* **2006**, *16*, 320-323.



Scheme 31: a) **CLXXIII**, $MgBr_2$, 89%, 3:1 *d.r.*; b) DIBAL-H; c) TBSOTf, 2,6-lutidine, CH_2Cl_2, 85-87%; H_2/Lindlar cat., 93-99%; d) DDQ, CH_2Cl_2, 82-85%; e) Swern oxidation, 93-96%; f) **XC**, *t*BuOK, toluene, 72-84%; g) PPTS, MeOH, 71-75%; h) I_2, PPh_3, imidazole, 87-94%; i) **CLXXI**, *t*BuLi, Et_2O, 9-MeO-9-BBN, then **LXXXVIII**, $Pd(dppf)Cl_2$, K_3PO_4, DMF, 86-98%; j) Dess-Martin periodinane, 71-86%; k) HF•Et_3N, 72-80%; l) Ag_2CO_3/ Celite, benzene, 75%.

2.2.4. Conclusion

The first part of the introduction regarding the leptomycin family discussed the biological studies, the use as biological tools and the most recent promising results as potential therapeutic agents of these compounds. The different synthetic strategies were then presented comparing the methods adopted by the different groups. Several times, the syntheses started from non-commercially available or advanced intermediates meaning that more steps have to be taken in account to evaluate the adopted strategies. Here, the total number of steps for each synthesis with the longest linear sequence beginning from commercially available starting materials is reported (Table 1).

Table 1: Resume of the synthesized members of the leptomycin family.

Compound	Group	Year	Steps (total)	Steps longest linear sequence (starting material)
Callystatin	*Kobayashi*	1998	39	18 (Roche ester **LI**)
	Crimmins	1998	37	18 (allyl iodide)
	Smith	2001	32	15 (oxazolidinone **LVIII)**
	Kalesse	2001	28	21 (Roche ester *ent*-**LI)**
	Enders	2002	40	15 (RAMP)
	Marshall	2002	39	18 (Roche ester *ent*-**LI**)
	Lautens	2002	45	27 (cyclohexanal)
	Panek	2004	37	18 (pseudoephedrine)
	Dias	2005	39	20 ((*S*)-2-methyl-1-butanal)
	Micalizio	2008	25	11 ((*S*)-2-methyl-1-butanal)
Leptomycin B	*Kobayashi*	1998	40	25 (geraniol)
(+)-Ratjadone	*Kalesse*	2000	36	19 (Roche ester *ent*-**LI**)
(–)-Ratjadone	*Williams*	2001	48	30 (geraniol)
(–)-Kazusamycin A	*Kuwajima*	2004	56	33 (diethylethoxymetylenemalonate)
Leptofuranin D	*Marshall*	2003	39	25 (Roche ester *ent*-**LI**)
Leptostatin	*Marshall*	2006	43	25 (Roche ester *ent*-**LI**)

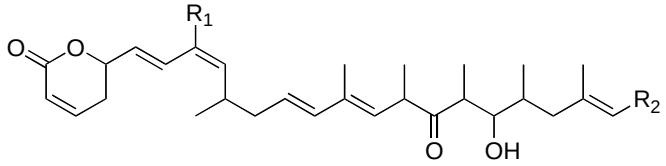
Following analysis of all the strategies we realized that some problems were common to several syntheses. Before planning our synthesis, some considerations had to be taken into account in order to avoid problematic steps. Firstly, it is evident that the most efficient method to form the lactone fragment is *via* a *Diels-Alder* reaction, as demonstrated by *Kalesse* and co-workers in their synthesis of callystatin.[56d] Another potential problem is related to the stereochemistry of the hydroxy group at C(17). As observed in the synthesis of *Lautens* and co-workers and *Dias* and co-workers, the *anti* configuration of C(17) gave unexpected problems. In the case of *Lautens*, the *Wittig* reaction for the formation of the C(12)-C(13) bond in the presence of the free hydroxy group afforded a *E*/*Z* 1:1.3 mixture of inseparable isomers (Scheme 14). Protection with TMS solved the problem to afford the coupled products

as a *E*/*Z* 19:1 mixture. The *anti* configuration in the polyketide chain also caused problems to *Dias* and co-worker. The formation of the C(11)-C(12) bond *via* a *Suzuki* cross-coupling with the TMS protected alcohol at C(17) afforded a protected/deprotected 34:66 mixture of products.[56i] In this case the problem was overcome by leaving the hydroxy group at C(17) unprotected (Scheme 18). Another problem encountered by both groups was the formation of α,β-unsaturated aldehyde derived from the lactol ring opening during the hydrolysis step. Among all the syntheses this problem was observed only when the *anti* configuration in the polyketide chain was present. Until now, for the all-*syn* configuration nobody has reported any problems related to the hydrolysis step. Therefore, it will be our configuration of choice during the synthesis. Concerning the final oxidation of the lactol to the lactone and the formation of the ketone at C(17), almost all the groups opted for PCC or the two step MnO_2/DMP sequence. More interesting was the final deprotection, where several groups encountered degradation problems of the starting material, especially in the synthesis of callystatin. Although *Kobayashi*, *Crimmins*, *Smith*, *Enders* and *Panek* and co-workers could obtain the target product using commercially available HF•pyridine solution (Schemes 4, 6, 9, 16), but other groups *i.e. Marshall* and *Lautens* and co-workers could not reproduce these results and obtained predominantly decomposition of the starting material (Schemes 11, 14). The acid-induced decomposition of the substrate using commercially available HF•pyridine was also observed by *Kalesse* and *Boger* and co-workers in their syntheses of ratjadone[89] (Scheme 24) and fostriecin[111] respectively. Both groups solved this problem by buffering the HF•pyridine solution with an additional portion of pyridine. The same solution was chosen by *Lautens*, *Dias* and *Micalizio* and co-workers in their synthesis of callystatin (Schemes 14, 18, 20), whereas *Marshall* and co-worker opted for the HF•Et_3N solution (Scheme 11).

[111] D. L. Boger, S. Ichikawa, W. Zhong, *J. Am. Chem. Soc.* **2001**, *123*, 4161-4167.

2.3. Anguinomycins A-D: Isolation and First Biological Evaluation

Anguinomycins A and B were isolated in 1985 and anguinomycins C and D in 1995 (Figure 7) from a strain belonging to *Streptomyces* microorganisms, during the research of new antitumor antibiotics using pRb-inactivated cells.[25] To date, no total syntheses of these compounds have been reported and their absolute and relative configuration remain unknown. In common with the other members of the leptomycin family, anguinomycins contains the electrophilic α,β-unsaturated lactone thought to be responsible for biological activity.



Anguinomycins
A: $R_1 = CH_3$; R_2 = COOH (**XV**)
B: $R_1 = C_2H_5$; R_2 = COOH (**XVI**)
C: $R_1 = CH_3$; $R_2 = CH_3$ (**XVII**)
D: $R_1 = C_2H_5$; $R_2 = CH_3$ (**XVIII**)

Figure 7: Anguinomycins A-D

The first biological studies highlighted the remarkable biological profile of these polyketides, which display very potent activity and high cytotoxicity to murine P388 leukemia cells (IC_{50} = 0.1-0.2 ng/mL) and potent antitumor activity in mice. A more significant discovery is that the anguinomycins display selectivity between normal and transformed tumoral cells, inducing cell-cycle arrest in normal cells but apoptosis in pRb-inactivated cells in picomolar concentrations. The cytotoxicity and selectivity test were performed on rat glia cells and transformed glia cells where pRb was inactivated by viral oncoproteins (Figure 8 and 9).[25]

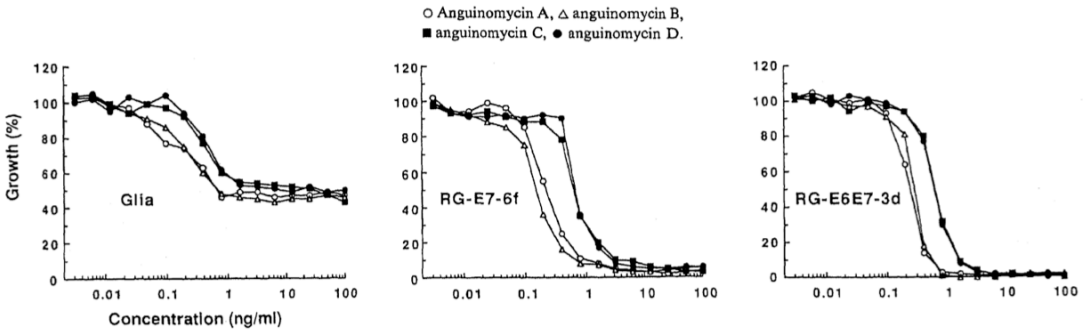


Figure 8: Anguinomycins A-D growth effects on normal and transformed rat glia cells.[25b]

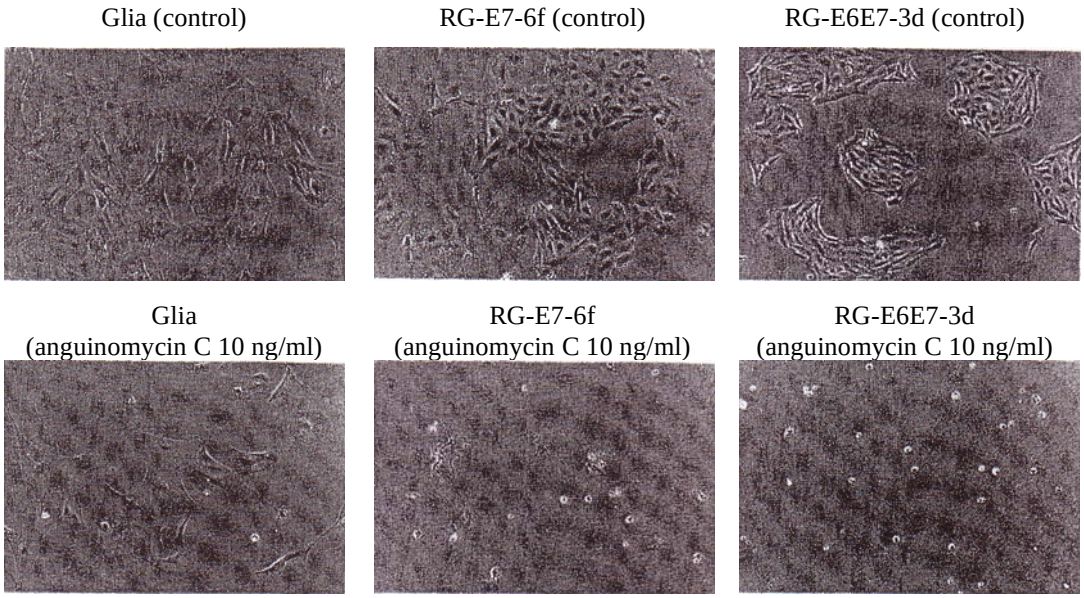


Figure 9: Anguinomycins C morphology effects on normal and transformed rat glia cells after 72 hours.[25b]

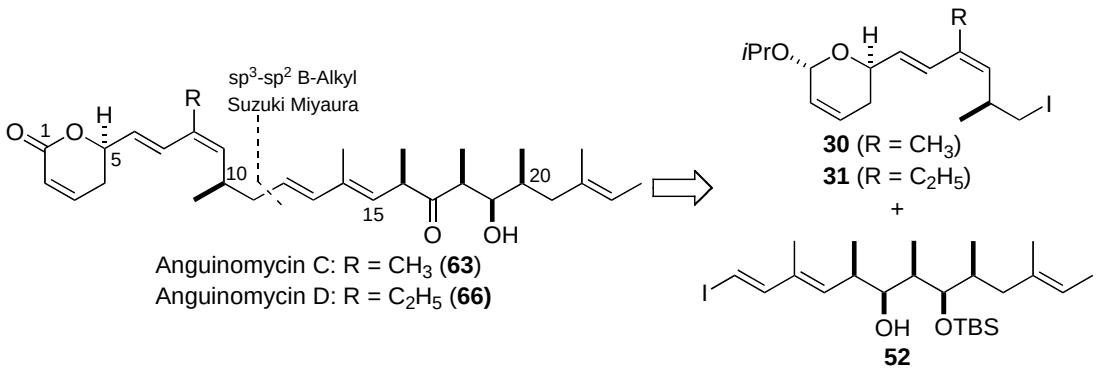
Flow cytometry analysis revealed that anguinomycins C and D induce cell growth arrest in the G1 phase,[25b] known to be a consequence of the p53 accumulation in the nuclei.[44] Surprisingly, apoptosis is also observed in p53-inactivated cells, leaving open the possibility that anguinomycins activate different signal pathways. These promising results encouraged us to prepare anguinomycins C and D in synthetic form and investigate their biological activity profile in the context of new antitumoral compounds.

2.4. Total Synthesis of Angunomycin C & D

2.4.1. Retrosynthetic Analysis and Strategy Considerations

To date, no total syntheses of the anguinomycins have been reported. Since anguinomycins C and D belong to the leptomycin family, we decided to plan our strategy in such a way to end the synthesis with the same configuration as found in LMB (**VI**, Figure 2). The strategic plan is characterized by metal-catalyzed reactions and aldol chemistry. We decided to disconnect the molecule between C(11) and C(12) giving two main fragments **30** (resp. **31**) and **52** that can be coupled *via* sp^3-sp^2 boron alkyl *Suzuki-Miyaura* cross-coupling (Scheme 32).[56f] This approach will circumvent possible problems related to the standard *Wittig* reaction between C(12) and C(13),

which, as observed previously (Chapter 2.2.3), sometimes gives rise to poor *E*/*Z* selectivity. The lactone will be protected as *i*PrO-lactol ether until the end of the synthesis to avoid problems due to the presence of the *Michael* acceptor and the ketone at C(17) that will be formed in the final steps, again to avoid unwanted side reactions.



Scheme 32: Retrosynthetic analysis. C(11)-C(12) disconnection.

The alkyl iodide fragments **30** and **31** will be constructed from the alkyne **8** and the dibromoolefin **20** *via* a tandem hydrozirconation/*Negishi* cross coupling and *Negishi* cross coupling with stereoinversion (Scheme 33).[112] The terminal alkyne **8** will be subjected to hydrozirconation[56h,67,113] and transmetallation to the vinyl Zn species. The organozinc intermediate will be used directly in the first *Negishi* cross-coupling[56h,114] that has to occur with *trans* selectivity. The second cross-coupling requires an inversion of the configuration in order to obtain the *cis* configuration at C(8)-C(9) and we propose to use the protocol developed by *Negishi*.[115] The dibromoolefin **20** is readily prepared from (*R*)-(–)-3-hydroxyisobutyrate (**9**) using standard chemistry. The hetero-*Diels-Alder* reaction using Cr(III)-catalyst developed by *Jacobsen*[116] is our method of choice for the facile preparation of the *i*PrO-lactol **8** from the aldehyde **2** and the commercially available 1-methoxy-1,3-butadiene (**3**) (Scheme 33). As discussed before, other approaches would be more time consuming and offer no real advantage in terms of selectivity and yield.

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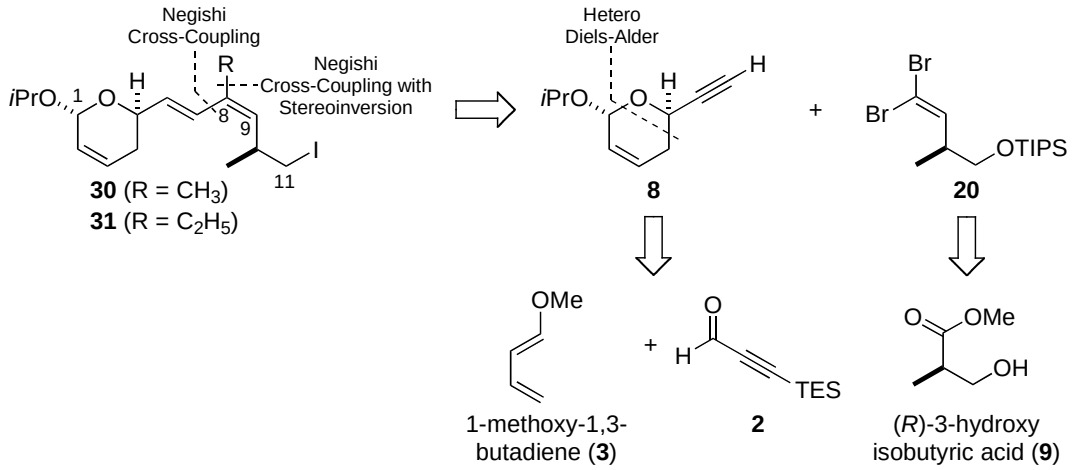
112 Preliminary investigations towards the preparation of compound **30** were undertaken during my diploma work.

113 a) F. Zeng, E.-I. Negishi, *Org. Lett.* **2002**, *4*, 703-706; b) P. Wipf, J. Heike, *Tetrahedron* **1996**, *52*, 12853-12910.

114 E. I. Negishi, A. O. King, N. Okukado, *J. Org. Chem.* **1977**, *42*, 1821-1823.

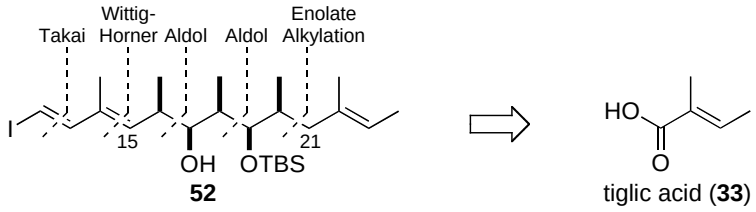
115 a) X. Zeng, Q. Hu, M. Qian, E.-I. Negishi, *J. Am. Chem. Soc.* **2003**, *125*, 13636-13637; b) X. Zeng, Q. Hu, M. Qian, E.-I. Negishi, *Angew. Chem., Int. Ed.* **2004**, *43*, 2259-2263.

116 a) A. G. Dossetter, T. F. Jamison, E. N. Jacobsen, *Angew. Chem., Int. Ed.* **1999**, *38*, 2398-2400; b) D. E. Chavez, E. N. Jacobsen, *Angew. Chem., Int. Ed.* **2001**, *40*, 3667-3670; c) K. Gademann, D. E. Chavez, E. N. Jacobsen, *Angew. Chem., Int. Ed.* **2002**, *41*, 3059-3061.



Scheme 33: Retrosynthetic analysis of the C(1)-C(11) fragment.

The polyketide chain synthesis is characterized by enolate alkylation and aldol reactions using the *Seebach* modification[117] of the *Evans* auxiliary[70] and starting from commercially available tiglic acid (**33**) (Scheme 34). The presence of two phenyl groups on the 4-isopropyl-2-oxazolidinone increases the stability of the auxiliary against nucleophilic attack, increases the selectivity of the reactions and the tendency of the obtained products to crystallize. As previously discussed, we opted for an all-*syn* configuration keeping the hydroxy group at C(17) unprotected in order to avoid problems during the sp^3-sp^2 boron alkyl *Suzuki-Miyaura* cross-coupling and the hydrolysis of the *i*PrO-lactol ether. The *trans* double bond between C(15) and C(16) will be installed *via* a *Wittig-Horner* reaction. *Takai* reaction for the formation of the (*E*)-vinyl iodide fragment **52** was expected to be problematic because of the bad *E*/*Z* selectivity, usually observed with α,β-unsaturated aldehydes.[118]



Scheme 34: Retrosynthetic analysis of the C(12)-C(24) fragment.

117 T. Hintermann, D. Seebach, *Helv. Chim. Acta* **1998**, *81*, 2093-2126.

118 a) K. Takai, K. Nitta, K. Utimoto, *J. Am. Chem. Soc.* **1986**, *108*, 7408-7410; b) T. Okazoe, K. Takai, K. Utimoto, *J. Am. Chem. Soc.* **1987**, *109*, 951-953; c) B. H. Lipshutz, B. Amorelli, *J. Am. Chem. Soc.* **2009**, *131*, 1396-1397.

2.4.2. Synthesis of the Dihydropyran Fragment

The synthesis of anguinomycins C and D started with the preparation of 3-triethylsilylpropynal (**2**)[119] by treatment of triethylsilylacetylene (**1**) with ethylmagnesium bromide followed by quenching with DMF.[120] The dihydropyran **5** was obtained *via* a hetero *Diels-Alder* reaction between aldehyde **2** and commercially available 1-methoxy-1,3-butadiene (**3**) (Scheme 35). The reaction was catalyzed by the Cr(III) catalyst (**4**) developed by *Jacobsen* and co-workers[121] under solvent-free conditions and in presence of 4 Å molecular sieves.[116] The work-up used in the preparation of the Cr(III) catalyst was demonstrated to affect its properties as well as its reactivity during the reaction.[122] We opted for a neutral aqueous work-up in order to obtain the dimeric species of the catalyst. The product **5** of the hetero *Diels-Alder* reaction was obtained in high yield (86%) and enantioselectivity (96% *e.e.*) and as a 5:1 diastereomeric mixture due to epimerization at the anomeric center under the reaction conditions. Attempts to use directly the MeO-protected lactol **5** for the continuation of the synthesis proved to problematic. The diastereoisomers could be easily separated by chromatography, but after deprotection of the silyl group with TBAF, the resulting terminal alkyne **6** was volatile and difficult to handle. We opted to treat the diastereomeric mixture obtained in the *Diels-Alder* reaction under acidic conditions in *i*PrOH to afford the *i*PrO-protected lactol **7** as a single diastereoisomer in the more thermodynamically stable configuration. Final deprotection with TBAF and purification on silicagel afforded the deprotected alkyne **8** as a colorless oil, which even if less volatile than alkyne **6**, was carefully concentrated (Scheme 35).

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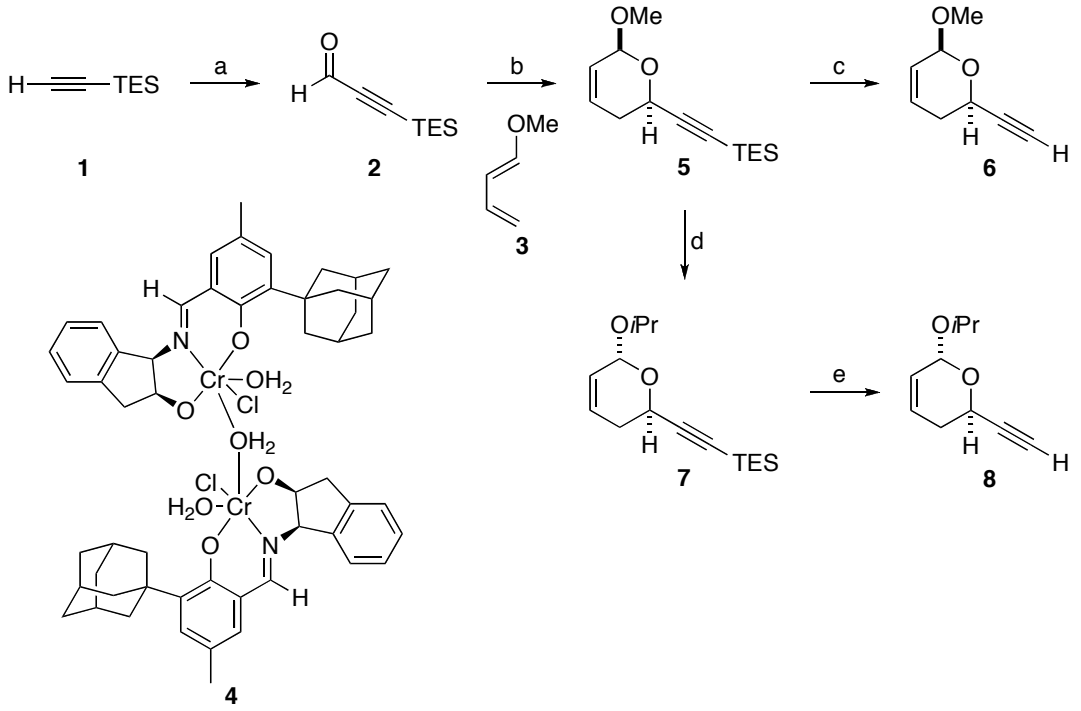
119 3-triethylsilylpropynal (**2**) is also commercially available from Fluorochem.

120 M. J. Plater, S. Aiken, G. Bourhill, *Tetrahedron* **2002**, *58*, 2415-2422.

121 D. E. Chavez, E. N. Jacobsen, *Org. Synth.* **2005**, *82*, 34-42.

122 E. R. Jarvo, B. M. Lawrence, E. N. Jacobsen, *Angew. Chem., Int. Ed.* **2005**, *44*, 6043-6046.



Scheme 35: a) EtMgBr, then DMF, 67%; b) **3**, **4** (2.3 mol %), 4 Å MS, RT, 86%; *d.r.* = 5:1, 96% *e.e.*; c) TBAF, THF, 0 °C → RT; d) PTSA, *i*PrOH, RT, 86%; e) TBAF (1 M in THF), THF, 0 °C → RT, 95%.

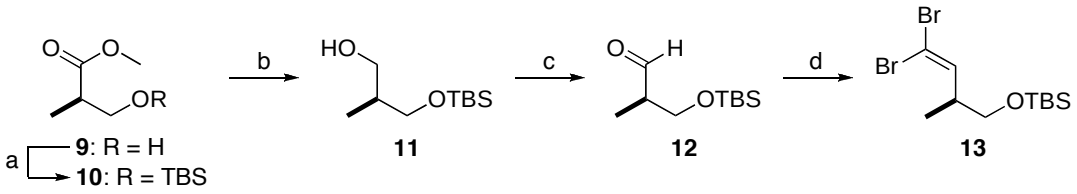
The adopted *Diels-Alder* approach allowed the preparation of the dihydropyran fragment **8** in only three steps from commercially available 3-triethylsilylpropynal (**2**). The Cr(III) catalyst (**4**) developed by *Jacobsen* and co-workers proved to be a good choice for the hetero-*Diels-Alder* reaction furnishing the product in high yield and enantioselectivity. Compared with other approaches reported in literature (Scheme 1 and 2), this route was the most straightforward, giving quick access to the target intermediate **8** and being amenable to scale up.

2.4.3. The Tandem Hydrozirconation-*Negishi* Cross Coupling

The coupling partner for the *Negishi* cross coupling was prepared from (*R*)-(–)-3-hydroxyisobutyrate (**9**), which was protected by treatment with TBSCl and imidazole. The resulting ester **10** was reduced in quantitative yield to the alcohol **11**, which was oxidized to afford **12** *via* a *Parikh-Doering* oxidation.[123] The aldehyde **12** was

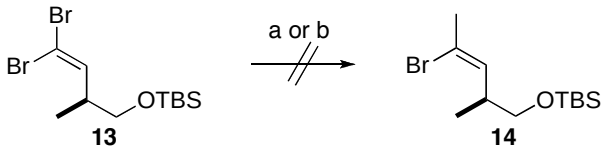
[123] a) J. R. Parikh, W. E. Von Doering, *J. Am. Chem. Soc.* **1967**, *89*, 5505-5507; b) Y. Guindon, L. Murtagh, V. Caron, S. R. Landry, G. Jung, M. Bencheqroun, A. M. Faucher, B. Guérin, *J. Org. Chem.* **2001**, *66*, 5427-5437.

converted to the dibromoolefin **13**[113a] in 85% yield using the *Corey-Fuchs* reaction (Scheme 36).[124]



Scheme 36: a) TBSCl, imidazole, CH_2Cl_2, 2 h; b) DIBAL-H (1.0 M in hexane), CH_2Cl_2, –78 °C → RT, 1 h 15 min; c) pyridine•SO_3, Et_3N, DMSO, RT, 3 h; d) PPh_3, CBr_4, Zn powder, CH_2Cl_2, 2 days, then **12**, 1 day, 85% (4 steps).

Before to attempting the planned hydrozirconation-*Negishi* cross coupling reaction with the previously synthesized fragments, it was decided to attempt to insert the methyl group in a *trans*-selective fashion on the dibromolefin **13**. Both, *Negishi*[114] and *Suzuki*[125] cross coupling failed to give the desired methyl-substituted product **14** and it was decided to revert to the planned procedure without spending more time in further investigations (Scheme 37).



Scheme 37: a) Me_2Zn, $Pd(PPh_3)_4$, 45 °C; b) $MeB(OH)_2$, $Pd(PPh_3)_4$, then TlOEt, RT.

The tandem hydrozirconation-*Negishi* cross coupling reaction started with the preparation of the *Schwartz* reagent (Cp_2ZrHCl) by treatment of commercially available Cp_2ZrCl_2 with $LiAlH_4$.[126] The terminal alkyne **8** was treated with *Schwartz* reagent at 0 °C and then allowed to return to RT to afford the *E*-alkenyl zirconocene intermediate **15** *via* stereospecific *syn* hydrometallation. *In situ* transmetallation to the organozinc intermediate **16** was achieved by addition of a solution of dried $ZnCl_2$ in THF.[127] A solution of dibromoolefin **13** in the presence of $Pd(PPh_3)_4$ (10 mol %) in THF was added and the resulting solution transferred into the vinylzinc solution. After 16 hours at 45 °C the reaction was quenched (Scheme 38). ^{1}H-NMR indicated

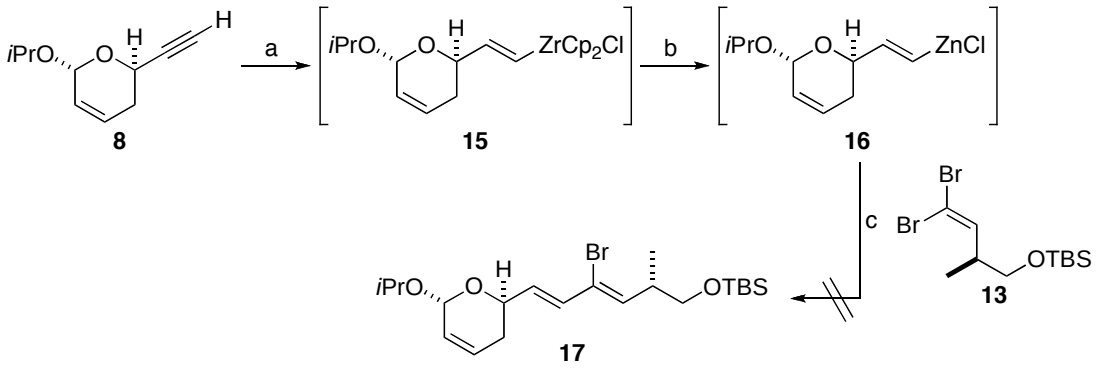
[124] E. J. Corey, P. L. Fuchs, *Tetrahedron Lett.* **1972**, *13*, 3769-3772.

[125] a) N. Miyaura, K. Yamada, A. Suzuki, *Tetrahedron Lett.* **1979**, *20*, 3437-3440; b) M. F. Jacobsen, J. E. Moses, R. M. Adlington, J. E. Baldwin, *Org. Lett.* **2005**, *7*, 2473-2476.

[126] S. L. Buchwald, S. J. LaMaire, R. B. Nielsen, B. T. Watson, S. M. King, *Tetrahedron Lett.* **1987**, *28*, 3895-3898.

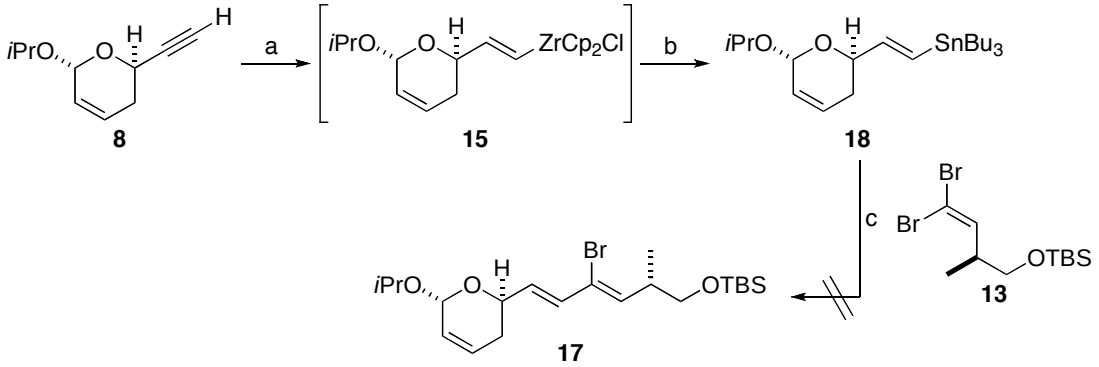
[127] The $ZnCl_2$ was flamed-dried under high vacuum.

partial formation of the coupled product **17**, but the TBS protecting group did not survive the reaction condition. Moreover non-identifiable side products were formed.



Scheme 38: a) Cp_2ZrHCl, THF; b) $ZnCl_2$, THF; c) **13**, $Pd(PPh_3)_4$ (5 mol %), 45 °C.

In order to overcome this problem it was decided to use the *Stille* cross coupling reaction. After formation of the organozirconocene intermediate **15**, transmetallation to the organotin species **18** was achieved by addition of Bu_3SnOMe.[128] After 22 hours at 40 °C, the solution of dibromoolefin **13** in presence of $Pd(PPh_3)_4$ (10 mol %) in THF was added and the resulting solution transferred into the vinyltin solution. The reaction was stirred for 2.5 days at 45 °C, but again the formation of a mixture of side products accompanied by the loss of the TBS protecting group was observed (Scheme 39).

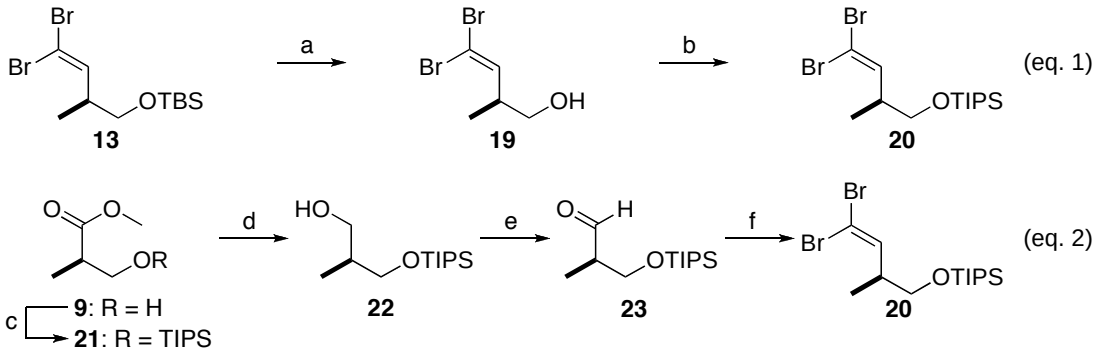


Scheme 39: a) Cp_2ZrHCl, THF; b) Bu_3SnMeO, THF; c) **13**, $Pd(PPh_3)_4$ (5 mol %), 45 °C.

At this point it was decided that changing of the TBS group with a TIPS group could make the substrate more stable toward the reaction conditions. Dibromoolefin **13** was deprotected with TBAF to give alcohol **19**, which was protected again by

[128] P. Wipf, H. Jahn, *Tetrahedron* **1996**, *52*, 12853-12910.

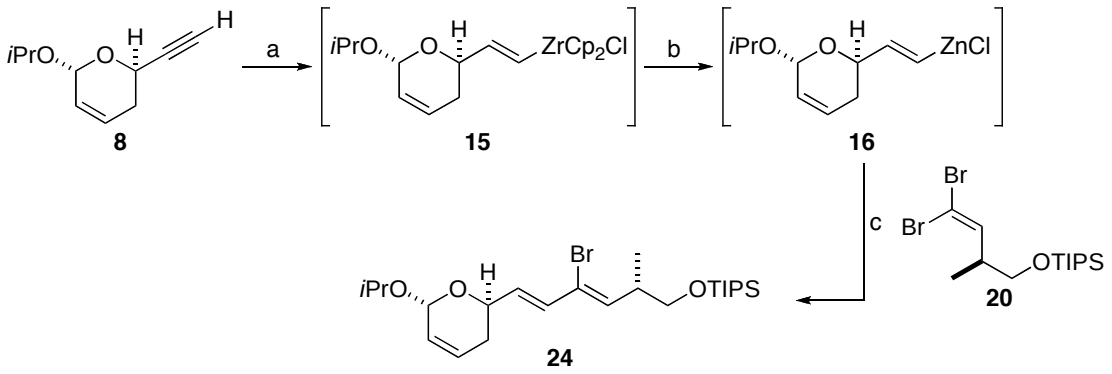
treatment with TIPSCl to afford the dibromoolefin **20**[129] in good yield (Scheme 40, eq. 1). The same dibromoolefin could also be obtained in four steps from (*R*)-(–)-3-hydroxyisobutyrate (**9**) following the same protocol adopted for the preparation of dibromoolefin **13**. Protection of the (*R*)-(–)-3-hydroxyisobutyrate (**9**) in the first step with TIPSCl afforded ester **21**, which was reduced to alcohol **22** by treatment with DIBAL-H. *Parikh-Doering* oxidation gave the aldehyde **23**, which was transformed to the dibromoolefin **20** *via* a *Corey-Fuchs* reaction (Scheme 40, eq. 2).



Scheme 40: a) TBAF (1.0 M in THF), THF, 0 °C → RT, 3 h, 47%; b) TIPSCl, imidazole, DMAP (cat.), CH_2Cl_2, overnight, 93%; c) TIPSCl, imidazole, DMAP (cat.), CH_2Cl_2, overnight, quant.; d) DIBAL-H (1.0 M in hexane), CH_2Cl_2, –78 °C → –15 °C, 1.5 h; e) pyridine•SO_3, Et_3N, DMSO, RT, 1.5 h, quant. (2 steps); f) PPh_3, CBr_4, CH_2Cl_2, 0 °C, 2.5 h, 64%.

At this point the tandem hydrozirconation-*Negishi* cross coupling reaction was tried again. A yellow solution of $Pd(PPh_3)_4$ (5 mol %) in THF was treated with a solution of DIBAL-H (10 mol %) to give a dark red solution. After 30 minutes, the new dibromoolefin **20** in THF was added and the resulting mixture transferred into the separately prepared vinylzinc solution **16**, *in situ* prepared from terminal alkyne **8**. The mixture was stirred 10 hours at 40 °C and after work up and purification, the coupled (*E*)-product **24** was obtained in 81% yield as a single diastereoisomer (Scheme 41). Even if Pd(0) was employed in this reaction and a reduction of the metal not required, the use of a small amount of DIBAL-H resulted in an increase of the yield.

[129] K. Komatsu, K. Tanino, M. Miyashita, *Angew. Chem., Int. Ed.* **2004**, *43*, 4341-4345.



Scheme 41: a) Cp_2ZrHCl, 0 °C → RT, THF, 1 h; b) $ZnCl_2$, THF, RT, 30 min; c) **20**, $Pd(PPh_3)_4$ (5 mol %), DIBAL-H (1.0 M in hexane), RT → 40 °C, 13 h, 81%.

^{1}H-NMR spectroscopic analysis of product **24** confirmed that the reaction had occurred in a completely selective fashion giving the (6*E*,8*Z*) isomer. Moreover NOE measurement with irradiation of H-C(7), H-C(9) and H-C(6) clearly demonstrated the spatial interaction between these two protons possible only for the *trans* configuration[130] (Figure 10-14).

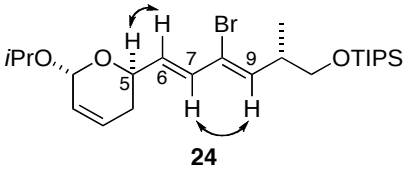


Figure 10: NOE effects for compound **24.**

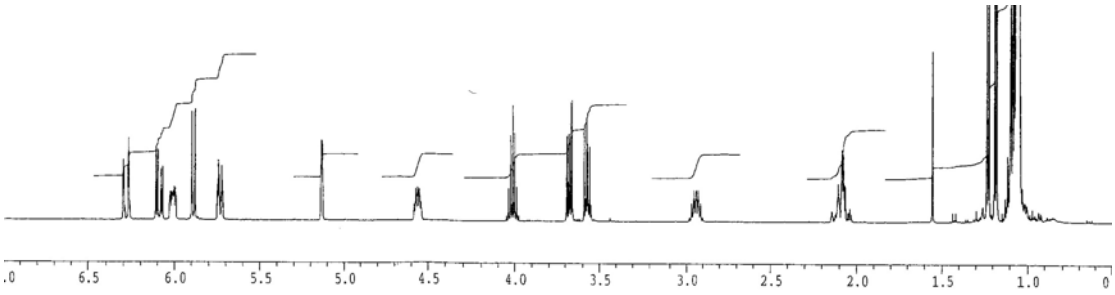


Figure 11: ^{1}H-NMR spectrum for compound **24.**

[130] In compound **24** the double bond at C(8) has a *cis* configuration as the bromine takes priority however, when considering the *Negishi* cross-coupling with stereoinversion the configuration of compound **24** must be considered relative to the *i*Pr-lactol moiety as this is conserved and the bromine is replaced with a lower priority substituent.

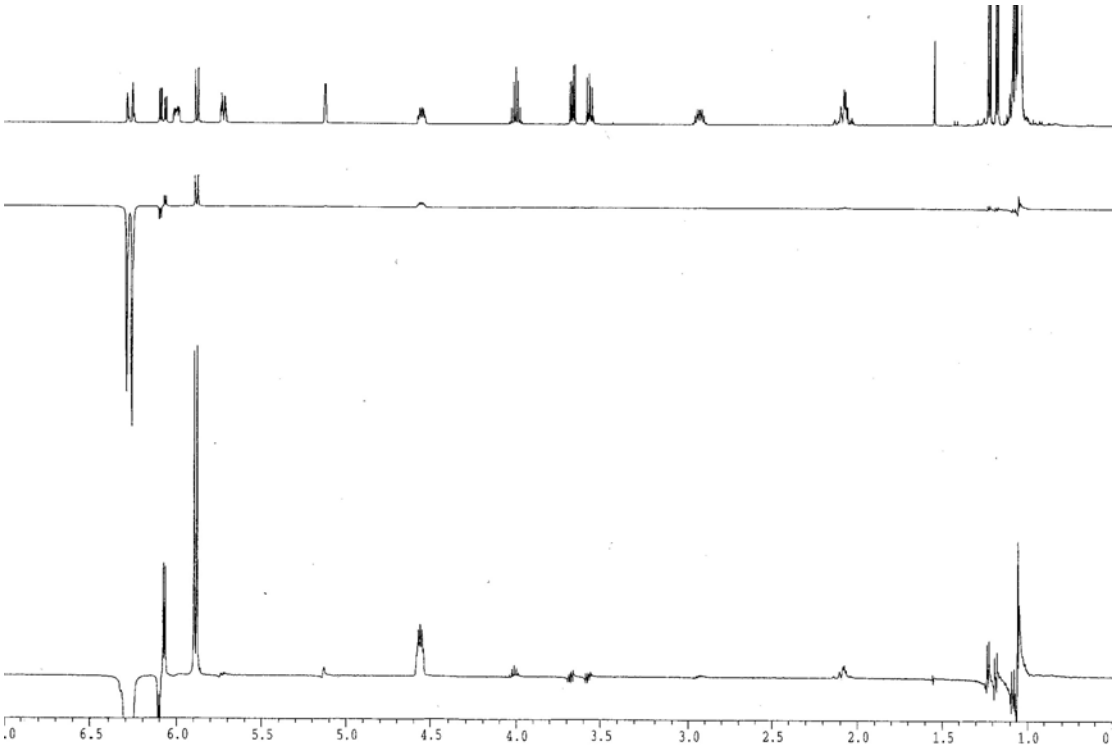


Figure 12: NOE spectrum for compound **24**: H-C(7) irradiation**.**

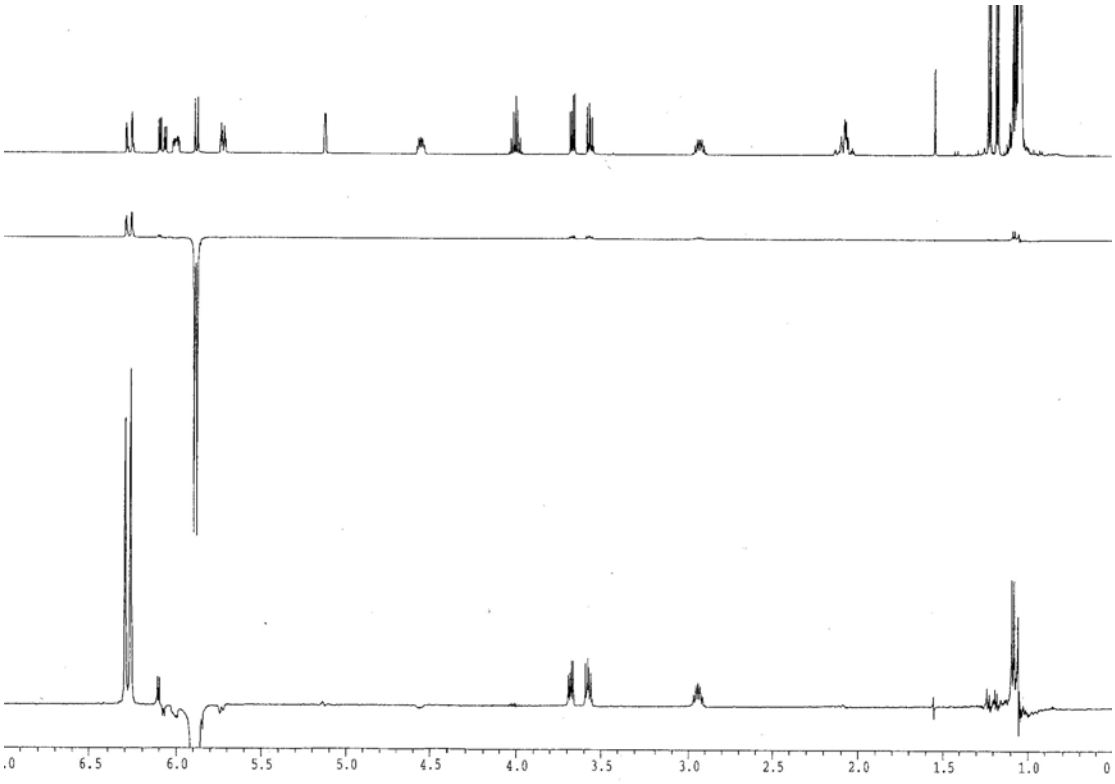


Figure 13: NOE spectrum for compound **24**: H-C(8) irradiation**.**

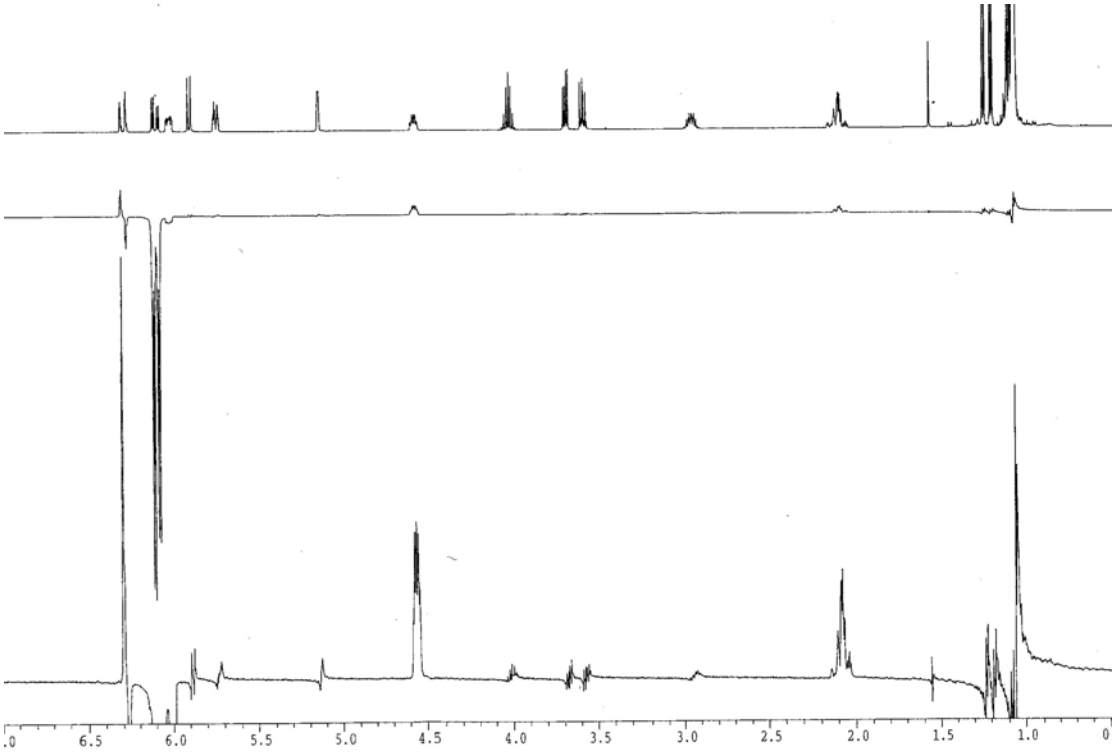
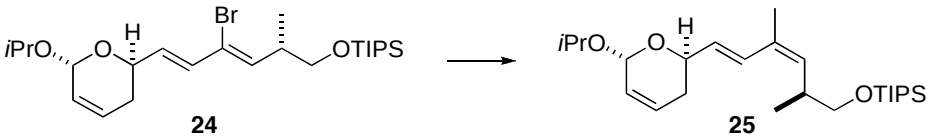


Figure 14: NOE spectrum for compound **24**: H-C(6) irradiation**.**

2.4.4. The Pd-Catalyzed *Negishi* Cross-Coupling with Stereoinversion

The first *Negishi* cross-coupling between alkyne **8** and dibromoolefin **20** afforded the coupled *trans* product **24**, but for the preparation of anguinomycins C and D a *cis* configuration at C(8) was required. To achieve this it was decided to apply the procedure developed by *Negishi* and co-workers[115] allowing the installation of the missing residue at C(8) with inversion of the double bond configuration at the same center. The nature of the catalyst employed in the Pd-catalyzed alkenylation of alkenyl halide influences the selectivity of the resulting product.[115] This reaction can occur with retention or inversion of the configuration depending on the ligands on the palladium. Since in the first *Negishi* cross coupling we obtained the *trans* product **24**, we now needed the alkylation to occur with inversion of the configuration affording the *cis* product (6*E*,8*Z*) present in the anguinomycins structure. With this reaction it would be possible to prepare both the intermediate for the synthesis of anguinomycin C, with a methyl group at C(8) and the intermediate for anguinomycin D, with an ethyl group at C(8). We started with the preparation of the intermediate for the synthesis of anguinomycin C adding Me_2Zn (2 M in toluene) to a solution of

alkenylhalide **24** and $Pd(PPh_3)_4$ (10 mol %) in THF under argon atmosphere. The resulting orange-colored solution was stirred for 24 hours at 45 °C, then a second addition of Me_2Zn was added and complete conversion of the starting material was achieved after an additional 14 hours. As expected, the use of $Pd(PPh_3)_4$ afforded to the *cis* product **25** as a single isomer in 68% yield.



Scheme 42: $Pd(PPh_3)_4$ (10 mol %), Me_2Zn (2.0 M in toluene), THF, 45 °C, 38 h, 68%, *cis*/*trans* > 97:3.

^{1}H-NMR, HSQC and COSY spectroscopic analysis of product **25** confirmed that the reaction had occurred in a completely selective fashion furnishing only the *cis* product as a single isomer. Moreover, NOE measurement confirmed the desired *cis* configuration. Irradiation of H-C(9) showed a NOE effect on the protons of the inserted Me-C(8), the same effect was also observed for H-C(6) and H-C(9) when protons of Me-C(8) were irradiated and for H-C(10) when irradiation on H-C(7) was performed. These interactions confirmed the spatial proximity of the irradiated protons possible only for the *cis* isomer (Figure 15-19).

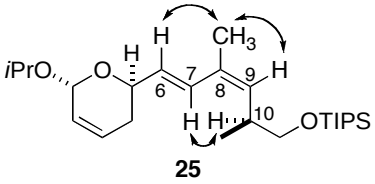


Figure 15: NOE effects for compound **25.**

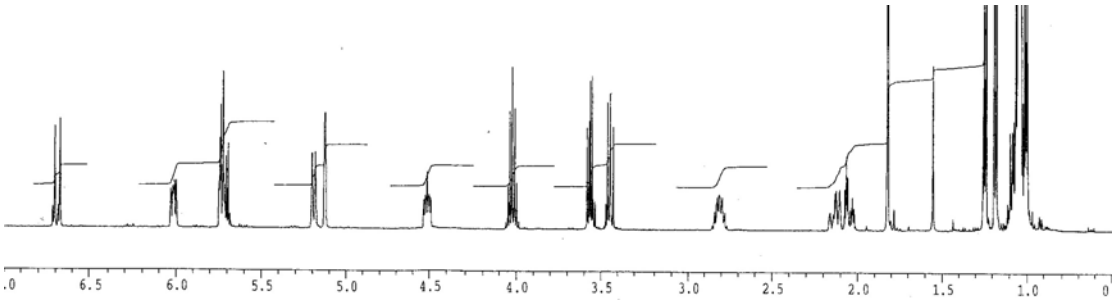


Figure 16: ^{1}H-NMR spectrum for compound **25.**

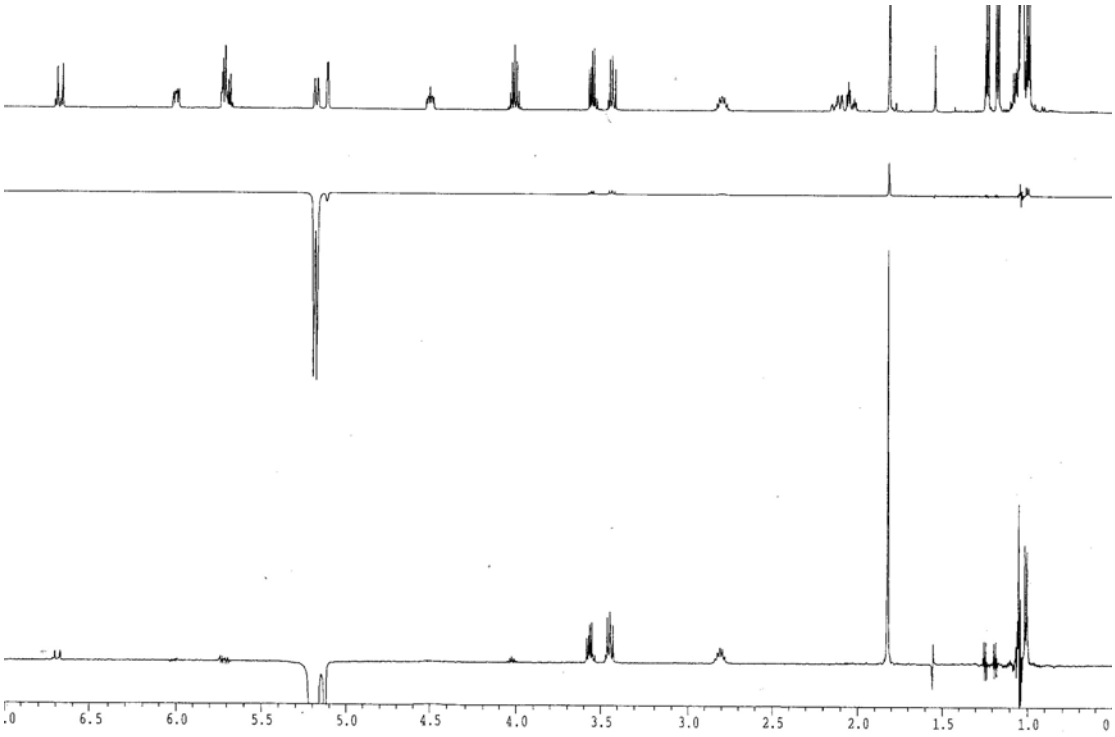


Figure 17: NOE spectrum for compound **25**: H-C(9) irradiation**.**

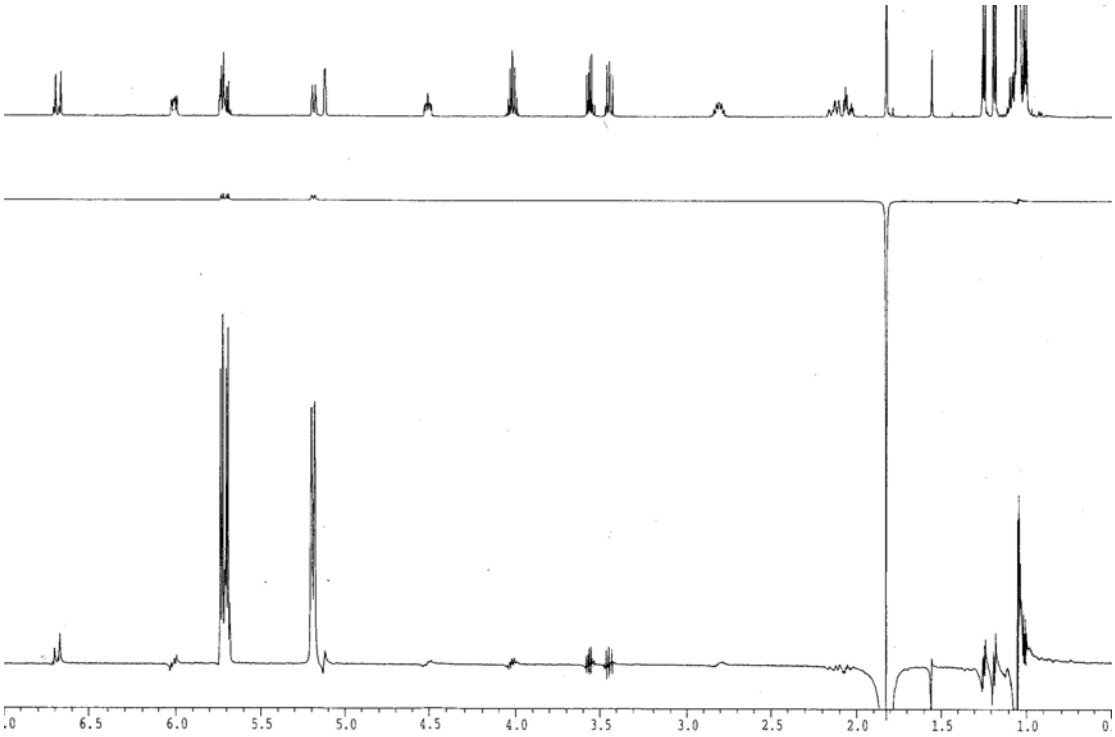


Figure 18: NOE spectrum for compound **25**: H-C(8) irradiation**.**

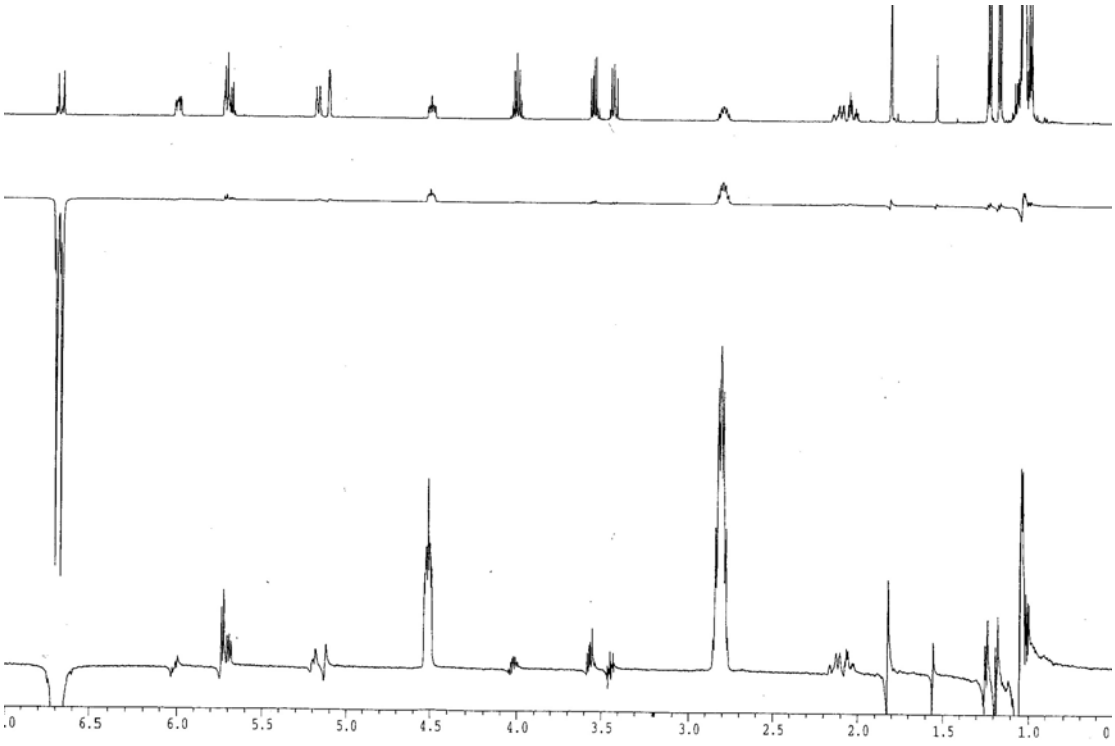


Figure 19: NOE spectrum for compound **25**: H-C(7) irradiation.

The same *Negishi* cross coupling with stereoinversion reaction was adopted for the synthesis of the fragment for anguinomycin D. In this case we used Et_2Zn (1.5 M in toluene), which was added to a solution of alkenylhalide **24** and $Pd(PPh_3)_4$ (10 mol %) in THF. However, following work-up we obtained a mixture of unreacted starting material, and the *cis* and *trans* products. Separation of the two isomers by chromatography on SiO_2 was unsuccessful; the similar polarity of the *cis* and *trans* isomer did not allow the isolation of the pure desired isomer. Attempts to force the reaction by adding more Et_2Zn were unsuccessful and promoted formation of the undesired *trans* isomer. Optimizations of the reaction conditions were attempted, but without success. Moreover the results revealed to be irreproducible when using the same conditions a different ratio of *cis* and *trans* products was obtained. In order to understand the problem we tried to repeat the reaction with Me_2Zn to achieve the previously obtained *cis* product **25**, but this time unsuccessfully. The reproducibility issues led us to think that the problem may be due to the presence of oxygen in the solvent. To investigate this problem it was decided to degas the solvent prior to use using three freeze/pump/thaw cycles. Better results were obtained with the *cis* product as the major isomer, but conversion of the starting material was not complete and formation of the *trans* compound was still present. At this point we chose to screen

several different palladium catalysts for the reaction (Figure 20 & Table 2). The reactions were run adding Et_2Zn (2.0 equiv) to a solution of alkenylhalide **24** and the catalyst (5-10 mol %) in THF under an argon atmosphere. After addition the tube was sealed and the reaction mixture stirred at 50 °C. The screened catalysts were $Pd(PPh_3)_4$, $PdCl_2$(dppf), $Pd(P\textit{t}Bu_3)_2$, $PdCl_2$(DPEphos), *trans*-di(μ-acetato)bis[o-(tolyl-phosphino)benzyl] dipalladium (II) and allyl[1,3-bis(mesityl)imidazol-2-ylidene] palladium chloride.

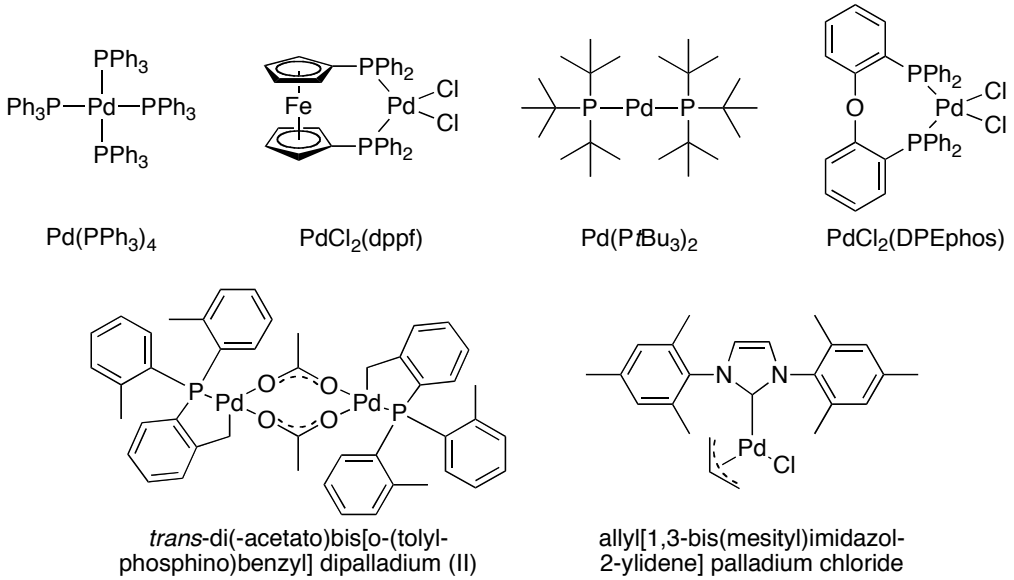


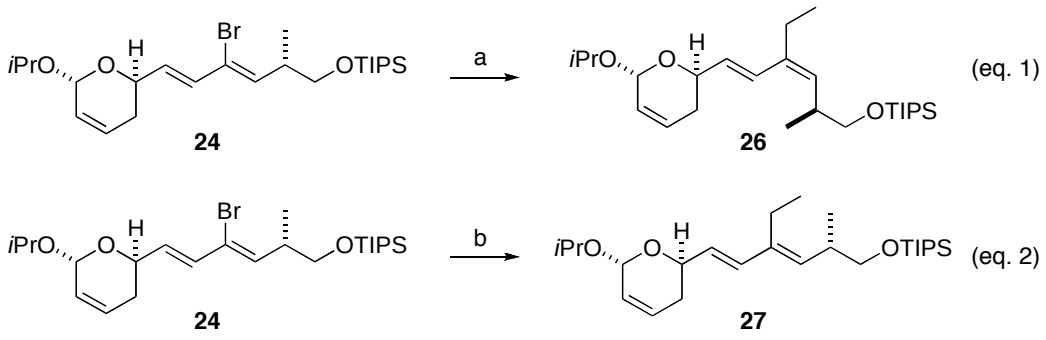
Figure 20: Evaluated Pd catalysts for the *Negishi* cross coupling with inversion/retention of the configuration.

Table 2: Screening different catalysts for the transformation of **24** to **26** (Scheme 43).

Catalyst	Equivalents (mol %)	Concentration (M *vs* **24**)	Reaction time (h)	Ratio[e]	Yield (%)
$Pd(PPh_3)_4$	5	0.06	24	0.14/1.00/0.38[a]	n.d.
$Pd(PPh_3)_4$	10	0.1	28	0.16/1.00/0.17 [a]	n.d.
$PdCl_2$(dppf)	10	0.05	20	1.00/1.08/0.66	n.d.
$Pd(P\textit{t}Bu_3)_2$[b]	10	0.05	3.5	0/0/1.00	75%
$PdCl_2$(DPEphos)[c]	5	0.08	14	0/1.00/0	84%
$PdCl_2$(DPEphos)[c]	10	0.08	14	0/1.00/0	84%
trans-di(μ-acetato)bis[o-(tolyl-phosphino)benzyl] dipalladium (II)[d]	10	0.05	20	0/0/1.00	65%
allyl[1,3-bis(mesityl) imidazol-2-ylidene] palladium chloride [b]	10	0.05	20	0/0/1.00	77%

[a] The results for $Pd(PPh_3)_4$ are not reproducible, using same conditions a different ratio starting material/*cis*/*trans* could be obtained. [b] During the addition of Et_2Zn the solution turns immediately to dark-brown colored. [c] During all the reaction the solution maintained an orange-red color. [d] At ca. 35 °C, the solution turn to dark-brown colored. [c] The ratio is reported as **24**/**26**/**27**.

In agreement with data reported by *Negishi* and co-workers,[115a] $PdCl_2$(DPEphos) revealed to be the best catalyst to achieve alkenylation with inversion of configuration. The *cis* adduct **26**, the intermediate for the synthesis of anguinomycin D, was obtained in 84% yield as a single isomer (Scheme 43, eq. 1). The reaction proved to be extremely clean, without traces of remaining starting material or the undesired *trans* isomer. It is probably the higher thermal stability of $PdCl_2$(DPEphos) compared to $Pd(PPh_3)_4$ that made the catalyst efficient over a longer time without any decomposition. The percentage of the employed catalyst (5 mol % or 10 mol %) did not influence the yield and selectivity of the reaction, affording in both cases only the *cis* isomer. Concerning $Pd(PPh_3)_4$ and $PdCl_2$(dppf) the results were not the same. As mentioned earlier, $Pd(PPh_3)_4$ gave a mixture of products and the same was observed for $PdCl_2$(dppf), opposite to that reported by *Negishi* and co-workers.[115a] In addition to the previously reported $Pd(PtBu_3)_2$, the *trans*-di(μ-acetato)bis[o-(tolyl-phosphino)benzyl] dipalladium (II) and the allyl[1,3-bis(mesityl)imidazol-2-ylidene] palladium chloride also afforded exclusively the *trans* adduct **27**, *via* retention of configuration. The yields were 75% (Scheme 43, eq. 2), 65% and 77% respectively.



Scheme 43: a) $PdCl_2$(DPEphos) (5 mol %), Et_2Zn (1.5 M in toluene), THF, 50 °C, 14 h, 84%, *cis*/*trans* > 97:3; b) $Pd(PtBu_3)_2$ (10 mol %), Et_2Zn (1.5 M in toluene), THF, 50 °C, 3.5 h, 75%, *trans*/*cis* > 97:3.

The mechanism of the stereoinversion remains unclear and the inversion was observed only for dienylpalladium intermediates generated *via* oxidative addition. Initial explanations from *Negishi* were based on thermodynamic stabilities of the involved π-allylpalladium intermediates involved and on sterics between the substituents (Figure 21, eq. 1).[115] The widely accepted π-σ-π rearrangement for the stereoisomerization of allylpalladium species can not be effective in this case as a

double inversion of configuration would be observed.[131] To date the only proposed mechanism for the observed stereoinversion was reported by *Negishi* himself in 2005,[132] but not demonstrated (Figure 21, eq. 2).

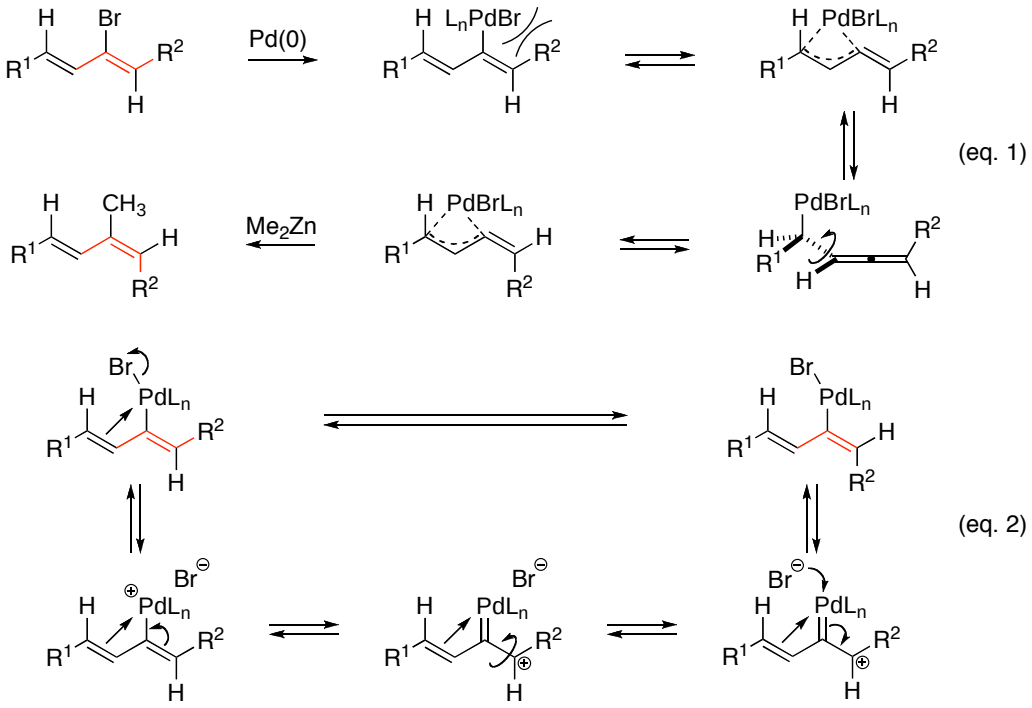
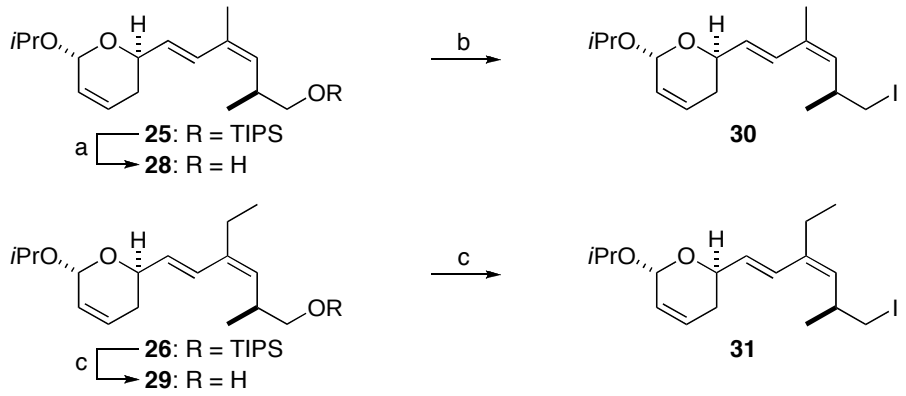


Figure 21: Postulated mechanisms for the *Negishi* cross coupling with inversion of the configuration.

At this point for both products **25** and **26** the silyl group was removed by treatment with TBAF in THF affording the deprotected products **28** and **29** respectively in 99% and 98% yield. Subsequently, the alcohols **28** and **29** were treated with I_2, PPh_3 and imidazole in toluene to get the alkyl iodide products **30** and **31** in high yields (Scheme 44).

[131] L. Acemoglu, J. M. J. Williams, in *Handbook of Organopalladium Chemistry for Organic Synthesis* (Ed.: E. I. Negishi), Wiley-Interscience, New York, **2002**, pp. 1689-1705.
[132] E. I. Negishi, Q. Hu, Z. Huang, M. Qian, G. Wang, *Aldrichimica Acta* **2005**, *38*, 71-88.



Scheme 44: a) TBAF (1.0 M in THF), THF, 0 °C → RT, 2 h, 99%; b) PPh_3, imidazole, I_2, toluene/Et_2O, 0 °C → RT, 2 h, 75%; c) TBAF (1.0 M in THF), THF, 0 °C → RT, 1.5 h, 98%; d) PPh_3, imidazole, I_2, toluene/Et_2O, 0 °C, 45 min, 89%.

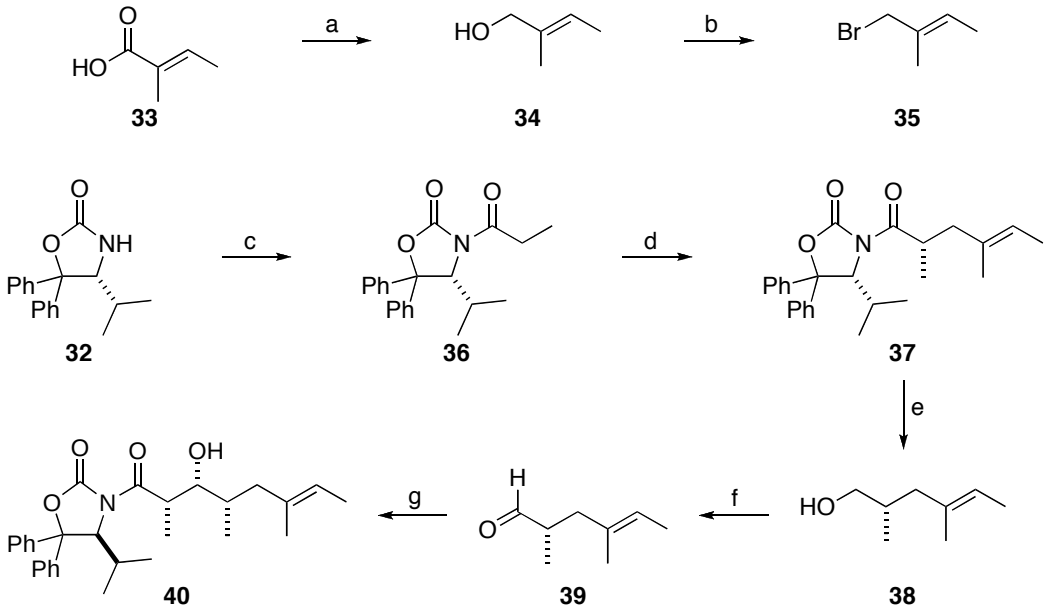
The alkyl iodide fragments **30** and **31** for the synthesis of anguinomycins C and D were obtained in 7 steps from commercially available 3-triethylsilylpropynal (**2**) in an overall yield of 29% and 42% respectively. In synthesis reported in this chapter, the *Negishi* cross coupling with stereoinversion was found to be of great use. After optimization of the conditions, high yield and selectivity as well as reproducibility of the results could be achieved. This reaction also allowed target fragments **30** and **31** to be obtained from the common intermediate **24**, minimizing the number of steps where the same chemistry would have to be done twice for the synthesis of similar fragments.

2.4.5. Synthesis of the Polyketidic Chain

As previously discussed, the synthesis of the polyketide chain would be based on *Evans* aldol strategy,[70] but using the DIOZ auxiliary (4-isopropyl-5,5-diphenyloxazolidin-2-one) (**32** and *ent*-**32**) developed by *Seebach* and co-workers (Scheme 45).[117] The additional phenyl groups on the auxiliary increases its stability against nucleophilic attack allowing the formation of the lithiated oxazolidinone using *n*BuLi at 0 °C, instead of –78 °C. Moreover, the presence of the two Ph groups increases the selectivity of the reactions as well as the crystallinity of the obtained intermediates. This auxiliary has already demonstrated its utility in total synthesis when being used by chemists at *Novartis* for the synthesis of discodermolide.[133] As

[133] a) O. Loiseleur, G. Koch, T. Wagner, *Org. Process Res. Dev.* **2004**, *8*, 597-602; b) O. Loiseleur, G. Koch, J. Cercus, F. Schürch, *Org. Process Res. Dev.* **2005**, *9*, 259-271.

previously discussed we opted for an all-*syn* configuration of the polyketide chain and the hydroxy group at C(17) would be kept free for the entire synthesis. Moreover, anguinomycin C and D displayed the same side chain allowing a unique synthesis of the polyketide chain for both targets. The synthesis of the polyketide chain started from commercially available tiglic acid (**33**), which was sequentially reduced by $LiAlH_4$ to alcohol **34** and then transformed to allylic bromide **35** in good yield. The acylated auxiliary **36** was prepared in 95% yield by deprotonation of the (*R*)-4-isopropyl-5,5-diphenyloxazolidin-2-one (**32**) with *n*BuLi at 0 °C and then addition of propionyl chloride. Treatment of the acylated chiral auxiliary **36** with LDA generated the lithium enolate, which reacted with allylic bromide **35** *via* enantioselective alkylation to give the adduct **37** in high yield (92%) and excellent selectivity (*d.r.* > 97:3) as a crystalline white solid. The auxiliary was removed with $LiAlH_4$ furnishing alcohol **38** in quantitative yield and allowing the recycling of the cleaved (*R*)-4-isopropyl-5,5-diphenyloxazolidin-2-one (**32**). *Swern* oxidation afforded aldehyde **39**, which was reacted with the boron enolate generated by treatment of *ent*-**36** with Bu_2BOTf to give the *syn*-aldol **40**. The product was obtained in 77% yield and a diastereomeric ratio of 87:13 (Scheme 45). Although not optimal, this selectivity is comparable to the best selectivities achieved by other groups using similar substrates *e.g.*, *Kobayashi* and co-workers (Scheme 3).[56a]



Scheme 45: a) $LiAlH_4$, Et_2O, 0 °C → RT, 4 h, 86%; b) PBr_3, Et_2O, 0 °C → RT, 3.5 h, 73%; c) *n*BuLi (1.6 M in hexane), 0 °C, then propionyl chloride, RT, overnight, 95%; d) **36**, LDA, –78 °C, 30 min, then **35**, THF, –78 °C → –10 °C, 26 h, 92%, *d.r.* > 97:3; e) $LiAlH_4$, Et_2O, 0 °C → RT, 3.5 h, quant.; f) oxalyl chloride, DMSO, CH_2Cl_2, –78 °C, then **38**, 15 min, then

NEt_3, –78 °C → 0 °C, 50 min, 99%; g) *ent*-**36**, Bu_2BOTf (1.0 M in CH_2Cl_2), NEt_3, –5 °C, 1 h, then –78 ° C, **39**, CH_2Cl_2, –78 °C → 0 °C, 2 h, 77%, *d.r.* > 87:13.

For this aldol reaction we had a match case with the *Re* face of the enolate attacking the *Si* face of the aldehyde. The transition state **TS1** afforded the *syn*-aldol **40** (Figure 22). The undesired diastereoisomer was not characterized, but based on literature precedent,[117] it is thought that a π-π interaction between a phenyl group on the auxiliary and the exocyclic double bond could stabilize **TS2** affording the *anti*-aldol **41**.[134] In this case the *Si* face of the enolate attacks the *Si* face of the aldehyde. The two diastereoisomers displayed similar R_f values, but separation by repeated flash chromatography on SiO_2 was possible affording pure *syn*-aldol **40**.

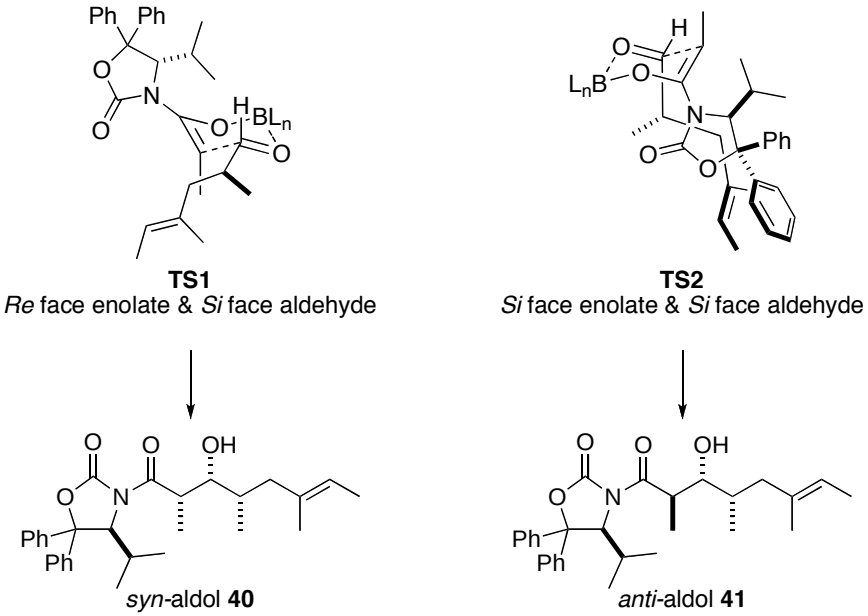
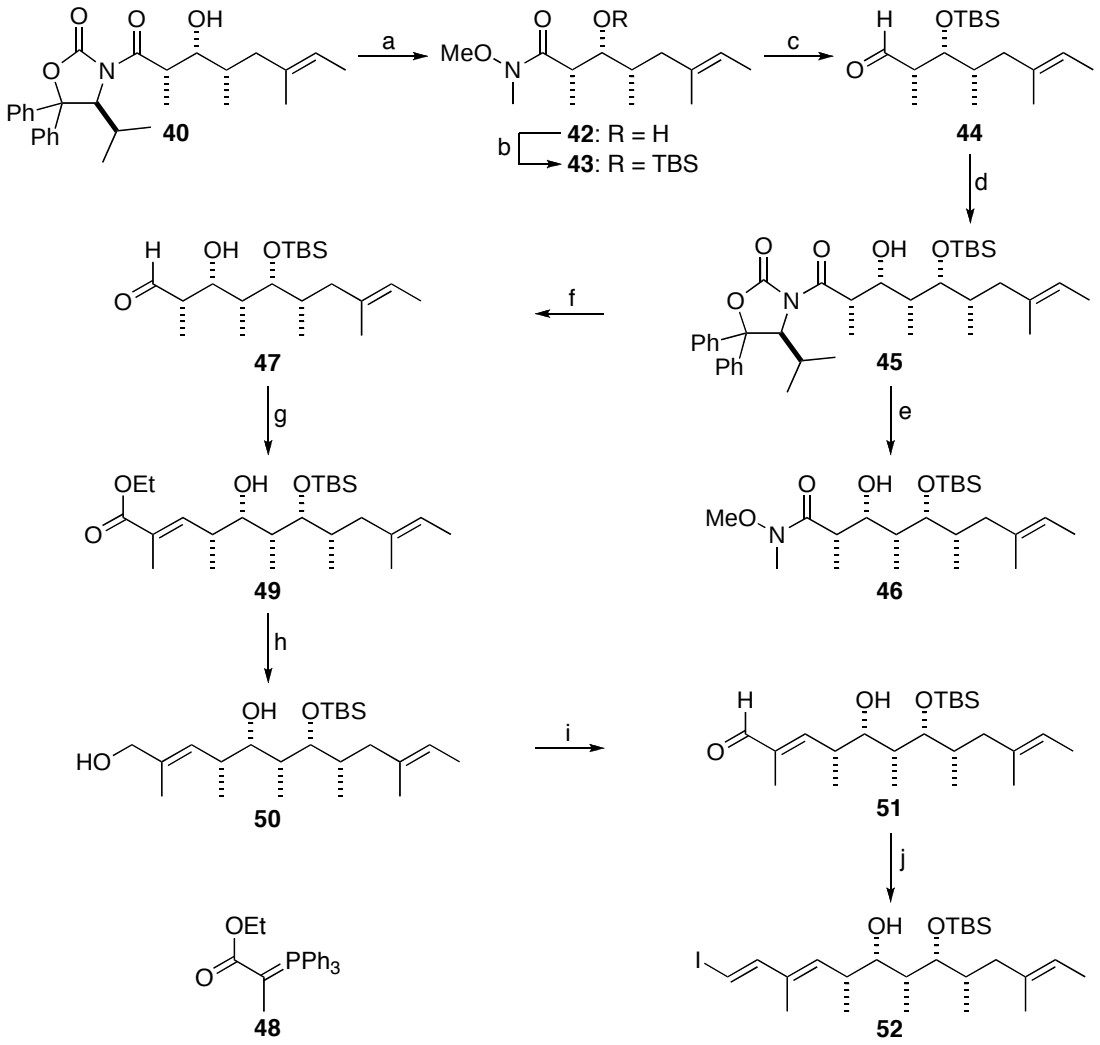


Figure 22: Transition states of the B-mediated aldol reaction. *Si* face attack of the aldehyde by the *Re* face of the enolate afforded to the *syn*-aldol **40**. *Si* face attack of the aldehyde by the *Si* face of the enolate afforded to the *syn*-aldol **40**.

The *syn*-aldol **40** was transformed to the *Weinreb* amide **42** in good yield (86%) using N,O-dimethylhydroxylamine hydrochloride and $AlMe_3$ in CH_2Cl_2. Amide **42** could be crystallized in hexane and the all-*syn* configuration confirmed by X-ray crystallographic analysis (Figure 23, a). Protection of the alcohol with TBS afforded product **43**, which was transformed to the aldehyde **44** by treatment with DIBAL-H in excellent yield. A second B-mediated aldol reaction using the same auxiliary, *ent*-**36** and aldehyde **44** afforded the *syn*-aldol adduct **45** in excellent diastereoselectivity (*d.r.*

134 The same interaction was observed by *Seebach* and co-workers using the same chiral auxiliary and phenylaldehyde as electrophile. T. Hintermann, D. Seebach, *Helv. Chim. Acta* **1998**, *81*, 2093-2126.

> 97:3) and 61% yield. As for the first boron-mediated aldol reaction, we had a matched case with the *Re* face of the enolate attacking the *Si* face of the aldehyde. The transition state **TS1** (Figure 22) is the same as before and afforded the *syn*-aldol **45**. In this case, no formation of the *anti*-aldol was observed, maybe because the longer chain on aldehyde **44** does not allow the interaction between its terminal double bond and the Ph group of the auxiliary *ent*-**36** as was observed in the formation of *anti*-aldol **41** (Figure **22**). Attempts to convert *syn*-aldol **45** to the *Weinreb* amide **46** proved to be difficult and even under forcing reaction conditions, the product **46** was obtained in poor 41% yield. Analyzing other approaches in the literature, we ascertained that *Kobayashi* and co-workers using the same conditions could convert a similar aldol, but with the *Evans* auxiliary, in good yield.[56a] We thought that the constraint came from the hindrance of the phenyl groups on the auxiliary and it was decided to remove it using $LiAlH_4$. Surprisingly, after optimization of the conditions ($LiAlH_4$ in toluene), the *syn*-aldol **45** could be converted directly to the aldehyde **47**, probably due to steric reasons. *Wittig* reaction between aldehyde **47** and (carbethoxyethylidene) triphenylphosphorane (**48**) afforded the α,β-unsaturated ester **49** in excellent yield (99%) and as a single isomer. Reduction with DIBAL-H afforded alcohol **50** and following MnO_2 oxidation gave the α,β-unsaturated aldehyde **51** in 80% yield over two steps. Aldehyde **51** (M.p. = 75-77 °C) could be crystallized in hexane and X-ray crystallographic analysis guaranteed the unambiguous determination of the all-*syn* relative configuration of the polyketide chain (Figure 23, b). The transformation of the aldehyde **51** to the vinyl iodide **52** *via* a *Takai* reaction was expected to be problematic due to selectivity issues of this reaction when using α,β-unsaturated aldehyde.[118] In our case, no problems were encountered and the vinyl iodide **52** was achieved in excellent yield and selectivity (*d.r.* > 97:3) (Scheme 46).



Scheme 46: a) MeONHMe·HCl, $AlMe_3$, CH_2Cl_2, 0 °C → RT, then **40**, CH_2Cl_2, 0 °C → RT, 15 h, 86%; b) TBSOTf, 2,6-lutidine, –20 °C → 0 °C, 1 h, 99%; c) DIBAL-H (1 M in hexane), THF, –78 °C, 1 h, quant.; d) *ent*-**36**, Bu_2BOTf (1 M in CH_2Cl_2), NEt_3, –5 °C, 45 min, then –78 ° C, **39,** CH_2Cl_2, –78 °C → 0 °C, 3 h, 61%, *d.r.* > 97:3; e) MeONHMe·HCl, $AlMe_3$, CH_2Cl_2, 0 °C → RT, then **45**, CH_2Cl_2, 0 °C → RT, 68 h, 41%; f) $LiAlH_4$ (1 M in Et_2O), toluene, –17 °C, 20 min, 83%; g) **47**, toluene, then **48**, RT → 35 °C, 5 h, 99%, *d.r.* > 97:3; h) DIBAL-H (1.0 M in hexane), THF, –78 °C → –10 °C, 1.5 h, 93%; i) MnO_2, CH_2Cl_2, RT, 2.5 h, 86%; j) $CrCl_2$, CHI_3, THF, 0 °C, 2 h, quant., *d.r.* > 97:3.

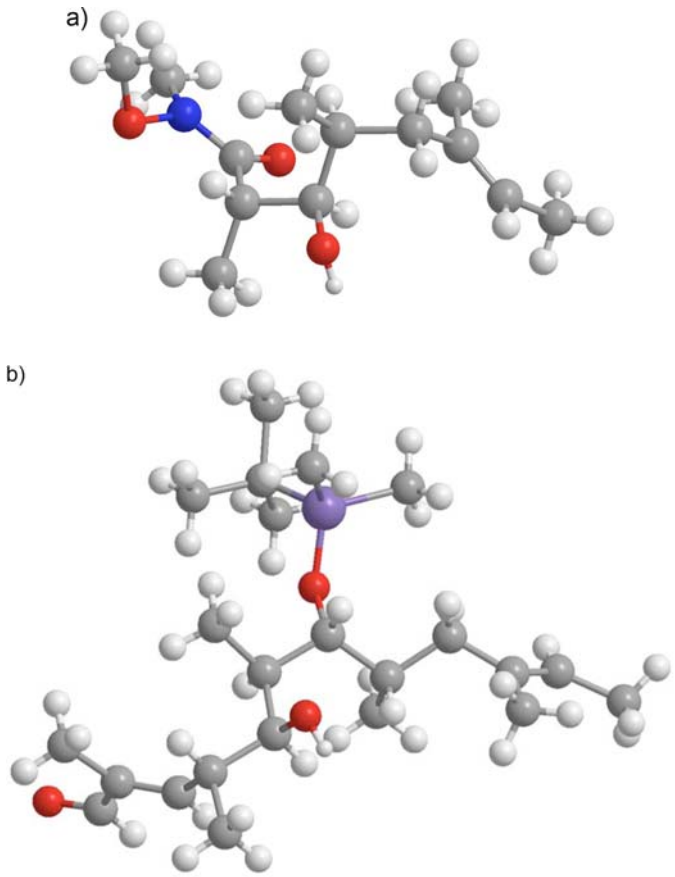
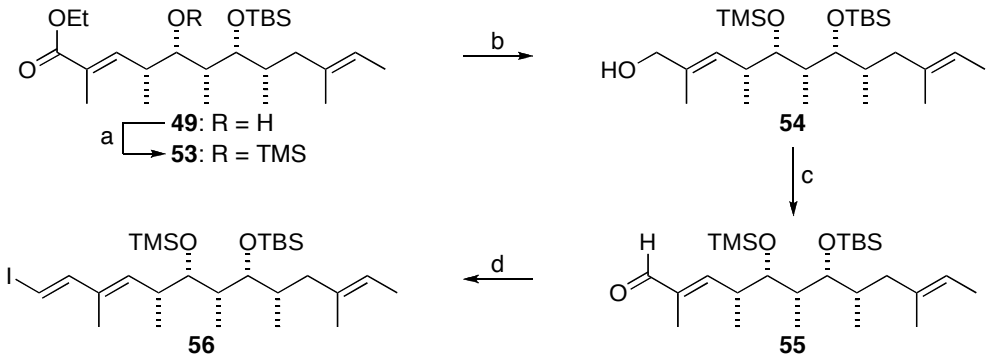


Figure 23: X-ray crystallographic analysis: a) amide **42**; b) aldehyde **51**.

At the same time we also prepared a small fraction of polyketide chain with the hydroxy group at C(17) TMS protected in order to evaluate the difference in the sp^3-sp^2 *Suzuki* cross coupling. The α,β-unsaturated ester **49** was protected with TMSCl in 77% yield affording product **53**. Following reduction of this substrate with DIBAL-H gave alcohol **54**, which was oxidized using *Swern* conditions to afford aldehyde **55** in quantitative yield. *Takai* reaction afforded the all-protected vinyl iodide **56** in 88% yield and in a *E*/*Z* ratio of 95:5 (Scheme 47).



Scheme 47: a) TMSCl, DMAP, NEt_3, CH_2Cl_2, 0 °C, 1 h, 77%; b) DIBAL-H (1.0 M in hexane), CH_2Cl_2, –78 °C, 1 h, quant.; c) MnO_2, CH_2Cl_2, RT, 3.5 h, quant.; d) $CrCl_2$, CHI_3, THF, 0 °C, 2.5 h, 88%, *d.r.* > 95:5.

The vinyl iodide fragment **52** was prepared in 15 steps from commercially available tiglic acid (**33**) in an overall yield of 15%. In this part we highlighted the strength of the *Evans* aldol chemistry[70] and in this case using the *Seebach* modification[117] of the *Evans* auxiliary for the synthesis of polyketides. Moreover, we also saw the usefulness of the *Takai* reaction for the synthesis of vinyl iodides.

2.4.6. The *Suzuki* sp^3-sp^2 Cross Coupling and Completion of the Synthesis

Having all the fragments in hand we attempted the sp^3-sp^2 *Suzuki* cross coupling following *Johnson*'s conditions[78] previously used by *Marshall* and co-workers in the synthesis of callystatin.[56f] The alkyl iodide **30** was reacted with 9-MeO-9-BBN and *t*BuLi at –78 °C forming the boronate intermediate **57**, to which a solution containing vinyl iodide **52**, Cs_2CO_3, $AsPh_3$ and $PdCl_2$(dppf) in a mixture of DMF/water was added. The reaction proceeded smoothly and the coupled product **58a** featuring the complete skeleton of anguinomycin C was isolated in 80% yield (Scheme 48).[135] The same procedure was applied to the fully protected vinyl iodide **56** and boronate intermediate **57**. However, this time the reaction did not give a good result and the desired coupled product, **58b**, was obtained in poor yield (Scheme 48). Having confirmed the superiority, in terms of yield, of the vinyl iodide **52** with the free hydroxy group at C(17) in the sp^3-sp^2 *Suzuki* cross coupling we performed the reaction with compound **31** for the preparation of the anguinomycin D skeleton. Alkyl iodide **31** was reacted with 9-MeO-9-BBN and *t*BuLi at –78 °C forming the boronate intermediate **59**, to which a solution containing vinyl iodide **52**, Cs_2CO_3, $AsPh_3$ and $PdCl_2$(dppf) in a mixture of DMF/water was added. As expected the reaction proceeded smoothly, but during purification on SiO_2 the appearance of a side product was observed. Two fractions were collected, one containing the pure coupled product **60** in 48% yield and the second in the same amount containing a mixture of product **60** and a side compound that we did not characterize, but supposed to be the product due to epimerization at C(17) (Scheme 48). It was decided to use the mixed fraction in the next steps without further purification.

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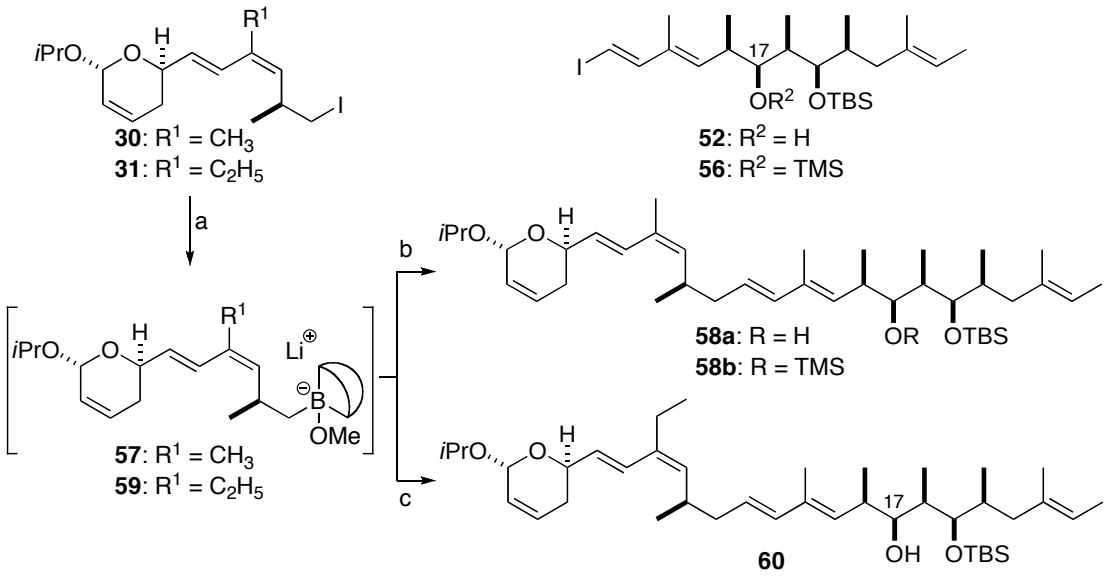
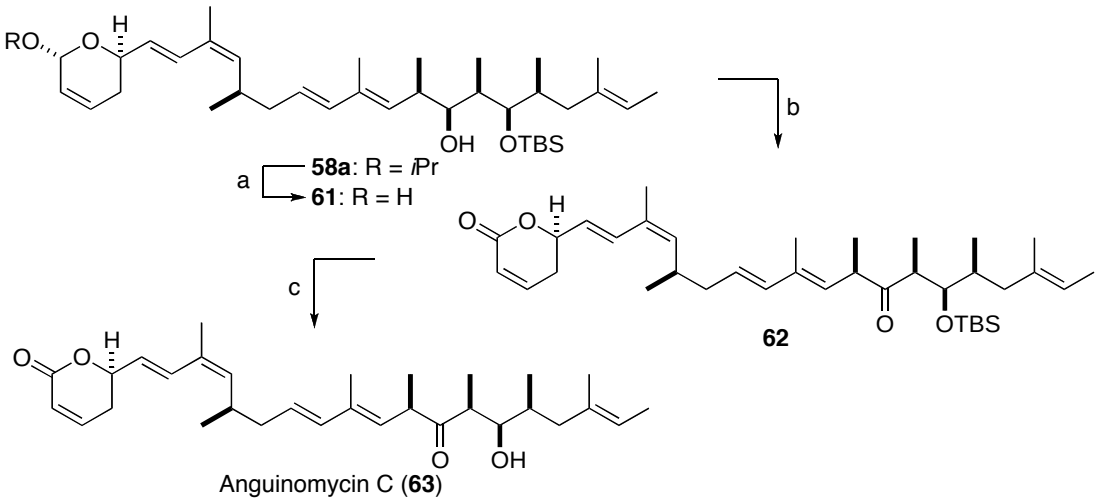
[135] Unsuccessful results for the cross-coupling of similar substrates using K_3PO_3 instead of Cs_2CO_3 and $AsPh_3$ were reported by Chakraborty and co-workers. T. K. Chakraborty, R. K. Goswami, M. Sreekanth, *Tetrahedron Lett.* **2007**, *48*, 4075-4078.



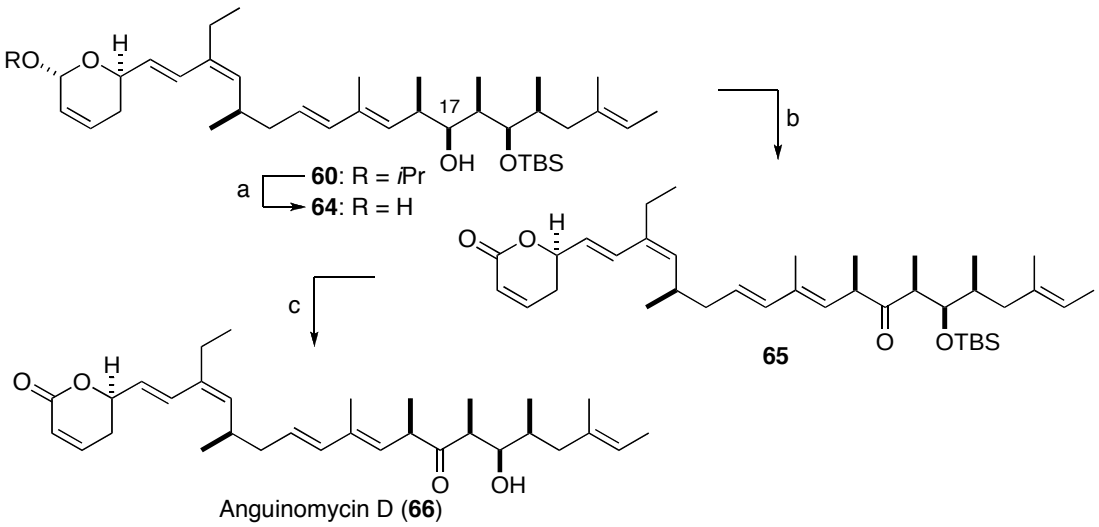
Scheme 48: a) **30** (resp. **31**), 9-MeO-9-BBN (1.0 M in hexane), *t*BuLi (1.5 M in pentane), Et_2O, then THF, –78 °C → RT, 1 h; b) **52** (resp. **56**), $Pd(dppf)Cl_2•CH_2Cl_2$ (5 mol %), $AsPh_3$ (15 mol %), Cs_2CO_3, H_2O, DMF, then **57**, RT, overnight, 80% (resp. 34%); c) **52**, $Pd(dppf)Cl_2•CH_2Cl_2$ (5 mol %), $AsPh_3$ (15 mol %), Cs_2CO_3, H_2O, DMF, then **59**, RT, 20 h, 48%.

Final steps for completion of the total syntheses were then performed. Thus, treatment of product **58a** under acidic conditions (PPTS) in a mixture of acetone/water cleaved the acetal in 95% yield to give lactol **61**. Surprisingly, attempted *Dess-Martin* oxidation of the lactol only oxidized the alcohol on the polyketide chain and did not form the lactone from the starting lactol. A further oxidation step using MnO_2 was then required to furnish lactone **62** in modest yield (47% over two steps). Finally, the TBS was removed using HF•pyridine buffered with pyridine. Quenching of the excess HF by buffering with pyridine proved to be crucial in avoiding degradation of the product. After work-up the crude material was directly purified by semipreparative HPLC to afford anguinomycin C (**63**) in 82% yield (Scheme 49).



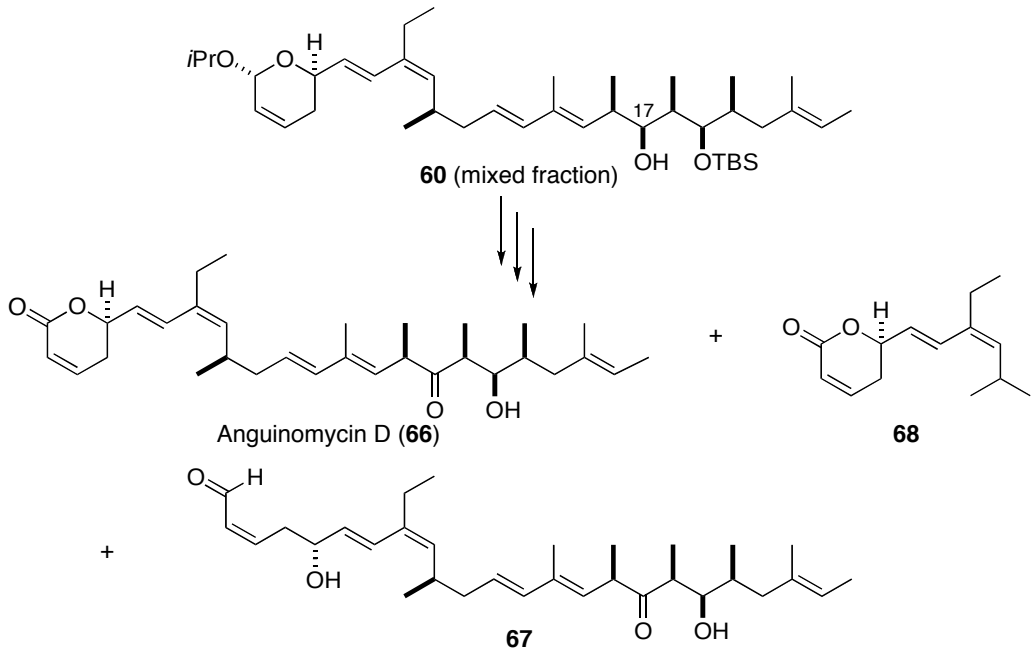
Scheme 49: a) PPTS, acetone/water (3:1), RT, 22 h, 95%; b) i. DMP, CH_2Cl_2, RT, 4 h; ii. MnO_2, CH_2Cl_2, RT, 14 h, 47%; c) HF•pyridine, pyridine, RT, 4.5 days, 82%.

A similar procedure was adopted for the final transformation of intermediate **60** to anguinomycin D. Acid-catalyzed cleavage of the acetal in product **60** afforded lactol **64** in good yield. Due to the poor yield obtained during the two step oxidation of compound **61** (DMP and MnO_2) in the synthesis of anguinomycin C (**63**) we chose to use PCC for the oxidation of both the alcohol at C(17) and the lactol in compound **64**. The oxidation was successful and afforded lactone **65**, which was directly treated with HF•pyridine solution buffered with pyridine for the final deprotection. This time, in order to avoid the aqueous work-up, we cooled the reaction to 0 °C and added some SiO_2 to the reaction to quench the excess of HF•pyridine. The resulting mixture was then loaded directly on to a column of SiO_2 and chromatographed affording anguinomycin D (**66**) in 60% yield (over two steps) (Scheme 50).



Scheme 50: a) PPTS, acetone/water (5/1), RT, 22 h, 91%; b) PCC, 4 Å MS, AcOH, CH_2Cl_2, RT, 1.5 h; c) HF•pyridine, pyridine, RT, 4.5 days, 60% (2 steps).

A second reaction batch containing a mixture of product **60** and the by product thought to be due to the epimerization at C(17) was subjected to the same sequence. Thus, acid-promoted cleavage of the acetal, oxidation using PCC and final removal of the silyl protecting group afforded a mixture of products that were purified by chromatography on SiO_2. Three compounds were isolated, anguinomycin D (**66**), the α,β-unsaturated aldehyde **67** and trace amounts of compound **68**. Aldehyde **67** results from opening of the lactol ring, a problem also encountered by *Lautens* and co-workers in their synthesis of callystatin (Scheme 14).[56g] Whilst compound **68** probably derives from degradation of the boronate intermediate in the *Suzuki* reaction (Scheme 51).



Scheme 51: Products generated when using a mixture of diastereoisomers at C(17).

In this section we highlighted the strength of the sp^3-sp^2 *Suzuki* cross coupling. The reaction was performed using advanced fragments furnishing the skeleton of the target molecules. After the cross-coupling only minor modifications were required to achieve synthetic samples of the anguinomycins C (**63**) and D (**66**), minimizing the risk of working with complex compounds that could be easily degraded. The total syntheses of anguinomycins C and D were obtained in a total of 29 steps (longest linear sequence 18 steps from diphenyloxazolidinone (**32**)). In addition two other products, the α,β-unsaturated aldehyde **67** and compound **68**, were isolated and submitted with anguinomycin C (**63**) and D (**66**) for biological evaluation (See chapter 2.4.9).

2.4.7. Physical Data of Anguinomycin C & D

Both anguinomycin C (**63**) and D (**66**) appeared as a colorless oil. Optical rotation values, the UV traces and IR spectra of the synthetic products matches with those reported in the literature and the high resolution ESI confirmed the correct masses (Table 3). The spectroscopic data of synthetic anguinomycins C and D were identical to those reported in literature (Figure 24a & b).[25] For anguinomycin C, the hydroxy group at C(17) which is not present in the natural compound spectrum is visible on ^{1}H-NMR spectrum of the synthetic sample at 2.40 ppm. Anguinomycins C (**63**) and D (**66**) have reduced stability because epimerization can take place in the polyketide chain. Moreover, they are good *Micheal* acceptors, which increases their reactivity towards nucleophilic attack. Anguinomycin C could be stored at –20 °C for more than one year without decomposition. Moreover, anguinomycins C and D are soluble in EtOH and could be stored as a solution at –20 °C for several days without degradation.[136]

Table 3: Physico-chemical properties of anguinomycins C and D.

	Anguinomycin C	Anguinomycin D
Formula	$C_{31}H_{46}O_4$	$C_{32}H_{48}O_4$
$[\alpha]^{22.5}_{D}$	–101.2° (*c* 0.0064, MeOH)	–112.0° (*c* 0.014, MeOH)
HRMS-ESI (calcd.)	505.3281 $[M + Na]^+$ (505.3294)	519.3429 $[M + Na]^+$ (519.3450)
UV λ_{max}	241 nm in MeOH	242 nm in MeOH
IR ν_{max}	3440, 2927, 1709	n.d.

[136] Anguinomycin A is commercially available from Alexis-biochemicals or Bioaustralis and long-term storage at –20 °C is recommend. This compound is soluble in EtOH (recommended) or MeOH but is unstable in DMSO.

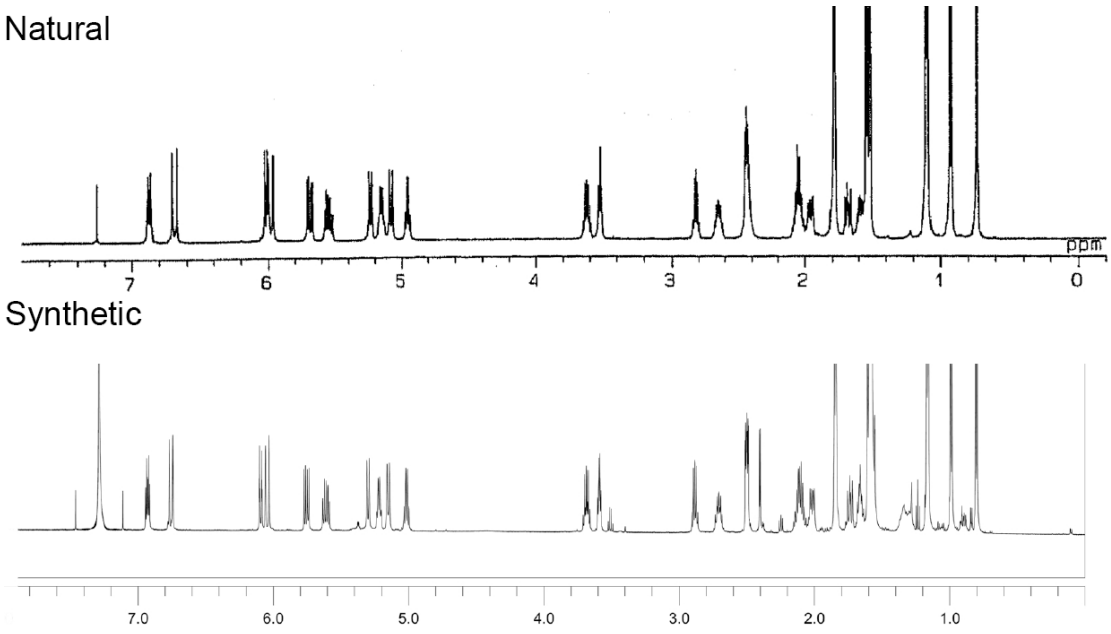


Figure 24a: Comparison of ^{1}H-NMR spectrum of natural and synthetic anguinomycin C.

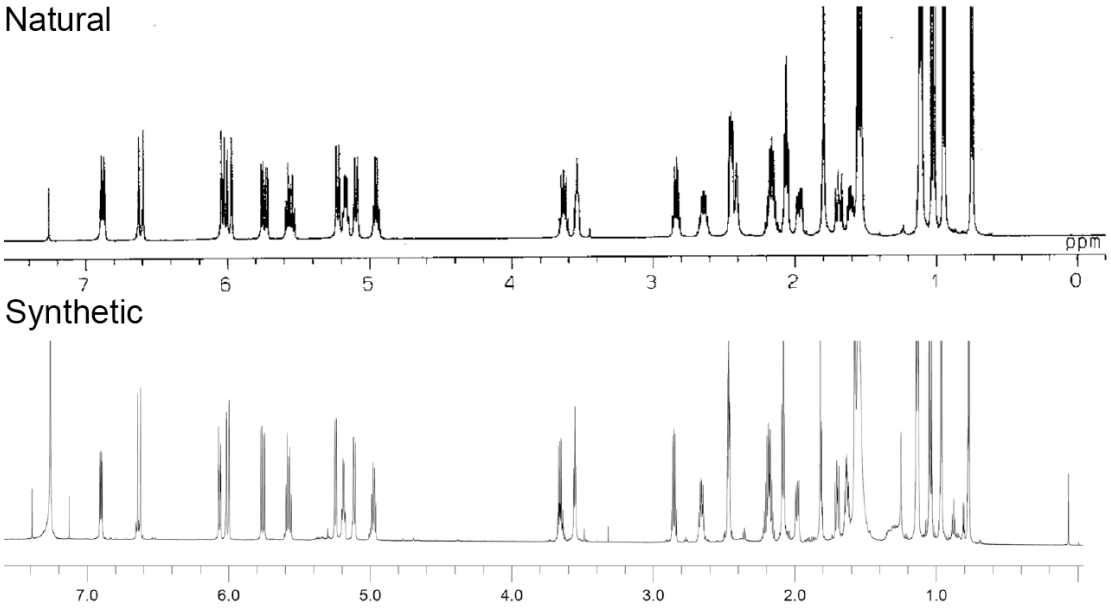
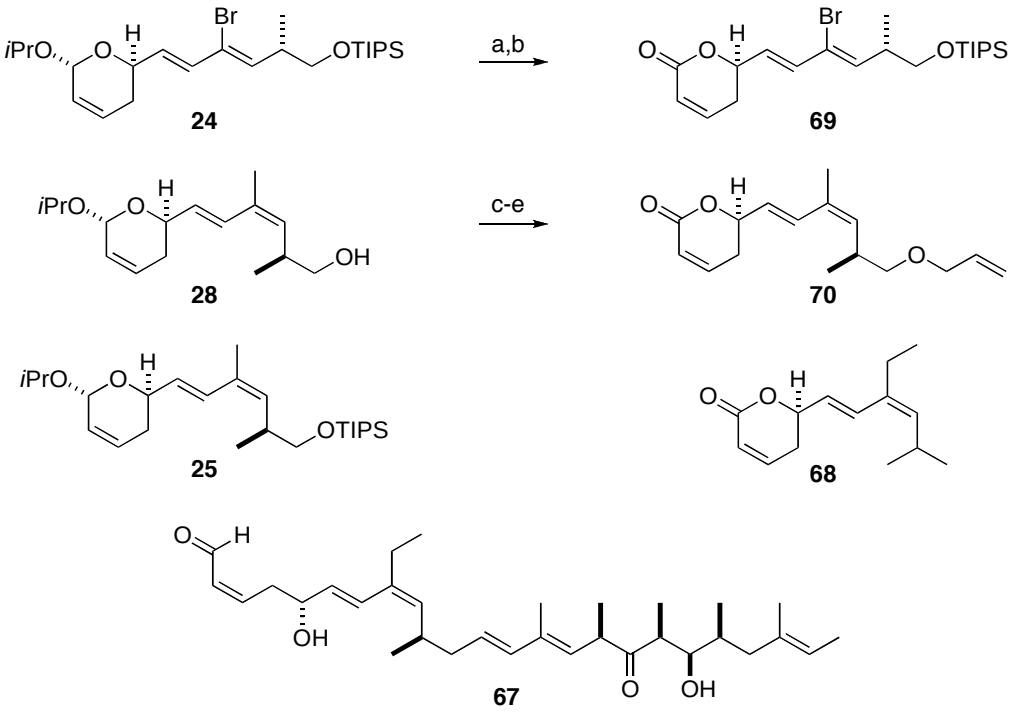


Figure 24b: Comparison of ^{1}H-NMR spectrum of natural and synthetic anguinomycin D.

2.4.8. Synthesis of Anguinomycin Derivatives

In order to investigate the mode of action and structure-activity relationships (SAR) for anguinomycins, we prepared several derivatives that were submitted for biological evaluation. The first derivatives synthesized displayed modifications at the lactone moiety and in the side chain to gain an understanding of which part of the molecule was important for the activity. The first results indicated that the lactone functionality was fundamental for the activity. Consequently, a second batch of derivatives were prepared in order to check if a modulation of the potency could be possible by modifying the side chain. However, these results will be not presented in this thesis because part of the PhD work of *Jean-Yves Wach* at the EPFL.

Derivative **69** was prepared from alkenylhalide **24** by removal of the acetal and then oxidation with MnO_2 (Scheme 52). This derivative conserved the lactone moiety. The chain was removed and in addition to the presence of a bromine at C(8), the diene system displayed *E,E* configuration and not the *E,Z* of the natural product. Compound **70** was prepared from alcohol **28** by reaction with allyl bromide and then following the same procedure as for derivative **69** (Scheme 52). The chain was substituted by a short residue, but the lactone moiety was maintained as well as the diene system in the *E,Z* configuration with the methyl group at C(8) as in anguinomycin C. In addition to these derivatives, intermediates **24** and **25**, both with the lactone masked as *i*PrO-lactol and for **24** displaying the wrong *E,E* configuration, were submitted for biological evaluation. In addition, we also submitted the α,β-unsaturated aldehyde **67** and compound **68** (Scheme 52), which were isolated during the synthesis of anguinomycin D. Interestingly, compound **67** displays the same side chain as the natural compound but the α,β-unsaturated lactone is replaced by an α,β-unsaturated aldehyde. Product **68**, which lacks the polyketide chain, is a truncated version of the natural compound.



Scheme 52: a) PPTS, acetone/water (3/1), RT, 2 h; b) MnO_2, CH_2Cl_2/pyridine (1:0.025), 1.5 h, 31% (2 steps); c) NaH, DMF, –20 °C, 25 min, then allyl bromide, –20 °C → RT, 8 h, 60%; d) PPTS, acetone/water (3/1), RT, 2 h; e) MnO_2, CH_2Cl_2/pyridine (1:0.025), 3 h, 29% (2 steps).

2.4.9. Biological Evaluation

Compounds such LMB selectively inhibit the CRM1-mediated nucleocytoplasmic transport by blocking the interaction between CRM1 and the NES signal of the cargo.[27,28] In order to test if anguinomycins C and D and the prepared derivatives could also inhibit this process, we analyzed how treatment of cells with these products influenced the intramolecular localization of the human protein Rio2. This protein is a cytoplasmic protein kinase this is exported from the nucleus to the cytoplasm by CRM1.[137] Inhibition of CRM1-mediated transport would result in an accumulation of the Rio2 protein in the nucleus. HeLa cells were incubated with different concentrations of anguinomycin C, anguinomycin D and derivatives for 90 minutes and then fixed with paraformaldehyde. LMB was used as a standard reference. After treatment, the localization of the Rio2 protein was determined by indirect immunofluorescence using specific antibodies, which target human Rio2.

[137] J. Rouquette, V. Choesmel, P. E. Gleizes, *EMBO J.* **2005**, *24*, 2862-2872.

Both anguinomycins C and D caused a strong accumulation of the Rio2 protein in the nucleus and displayed similar activity to the standard reference LMB, whereas in untreated control cells the Rio2 protein was localized in the cytoplasm. The results confirmed that both anguinomycins C and D are potent inhibitors of CRM1-mediated nucleocytoplasmic transport. Anguinomycin D displayed full inhibition at 5 nM, while anguinomycin C showed a weak inhibition at the same concentration and reaching full inhibition at 10 nM. These values can be compared with that of LMB, which fully inhibits nucleocytoplasmic transport at 1 nM. (Figure 25a & b).

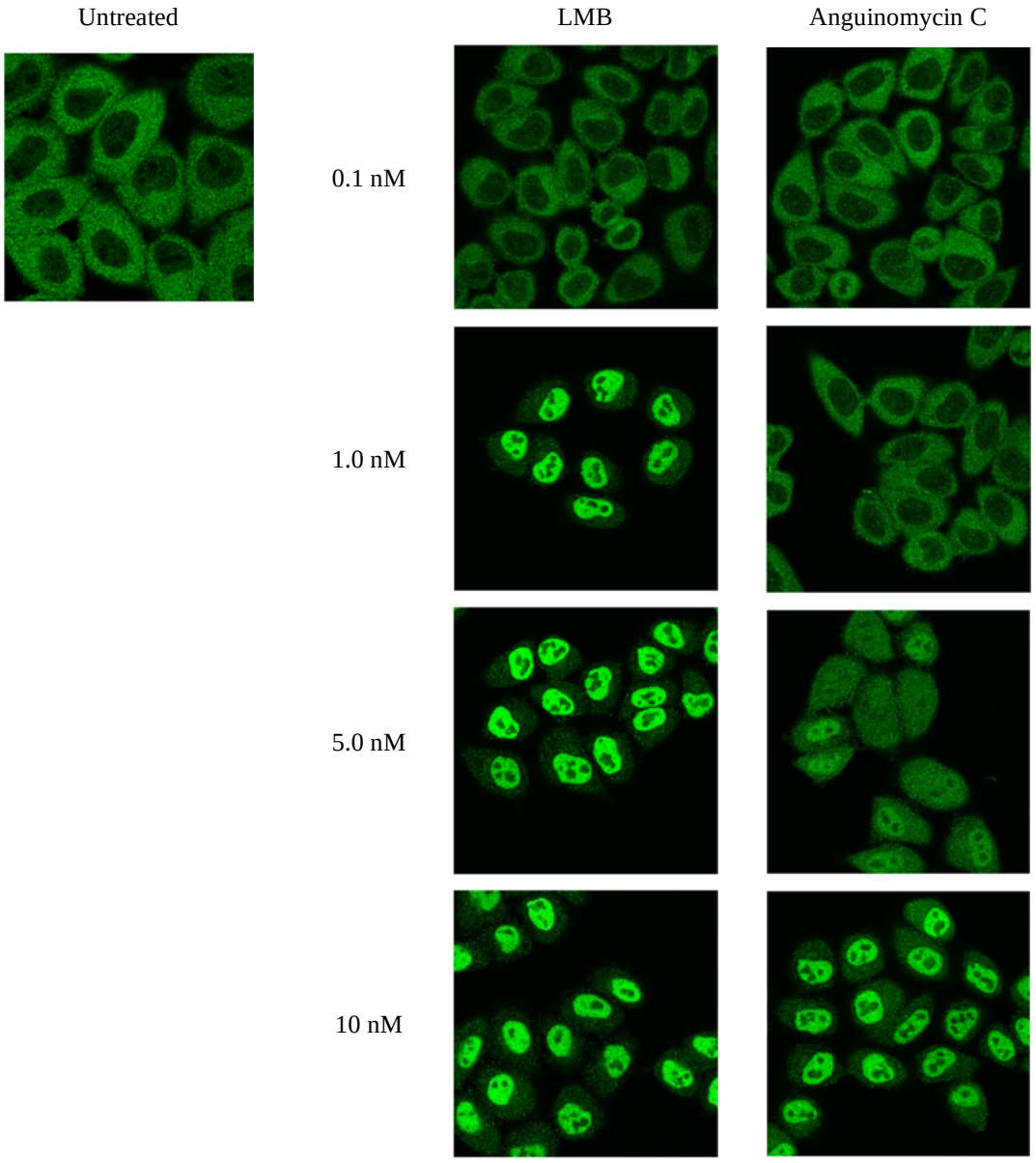


Figure 25a: Anguinomycin C inhibition of CRM1-dependent nuclear export of Rio2 in HeLa cells.

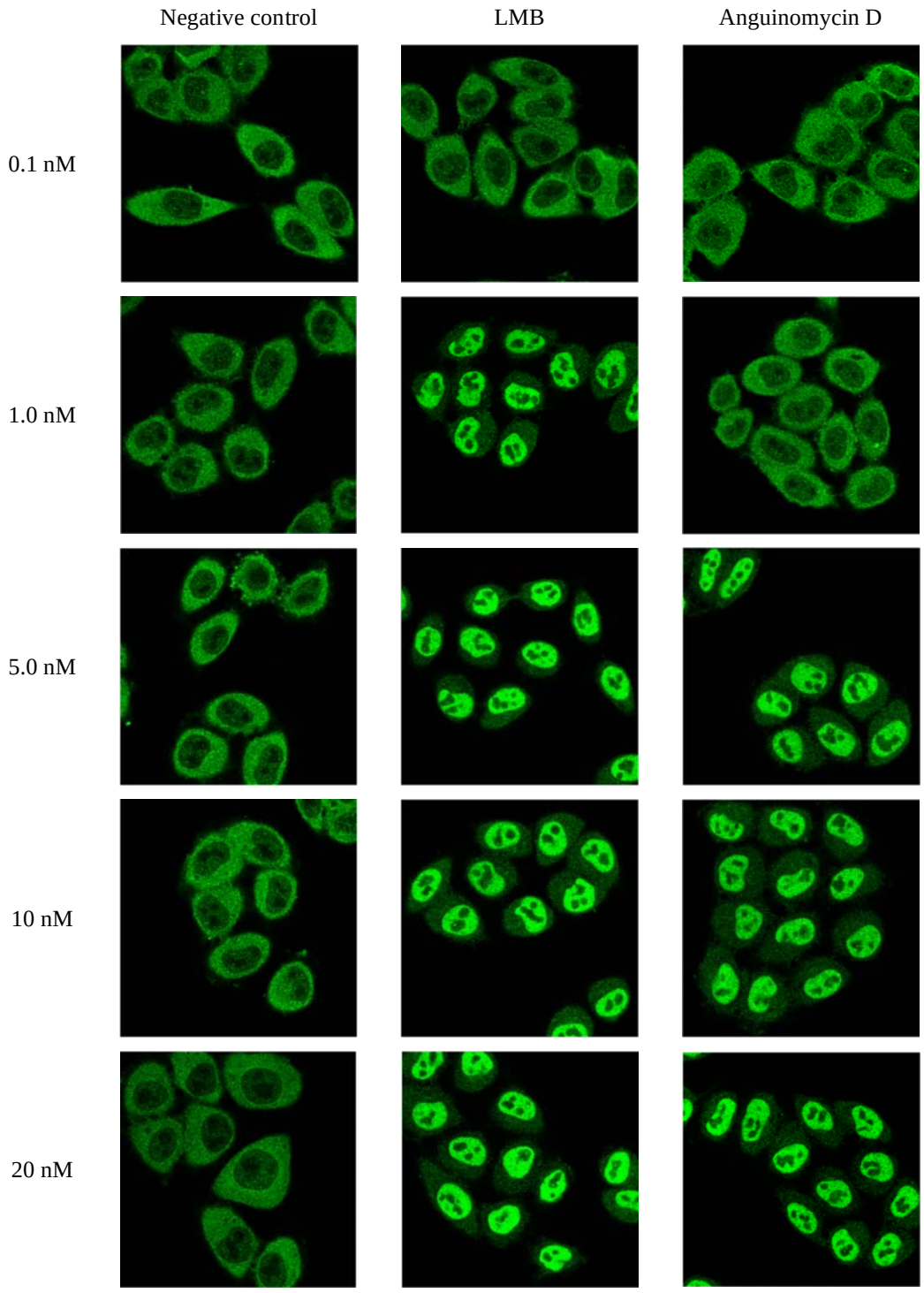


Figure 25b: Anguinomycin D inhibition of CRM1-dependent nuclear export of Rio2 in HeLa cells. Compound **60** was used as negative control.

Derivatives **24** and **25** (Scheme 52), which are structurally very different to anguinomycin, were then tested against the same target and did not show activity at concentrations below 100 μM. For derivative **69** it was not clear from the image if even weak inhibition was achieved at 100 μM. However, activity at 10 μM was observed for derivative **70** which contains the same C(1)-C(11) fragment as formed in

natural anguinomycin C (Figure 26). These initial results indicate that the activity is derived mainly from the lactone moiety, which therefore must play a crucial role in the mode of action while the side chain was probably involved in the molecular recognition.

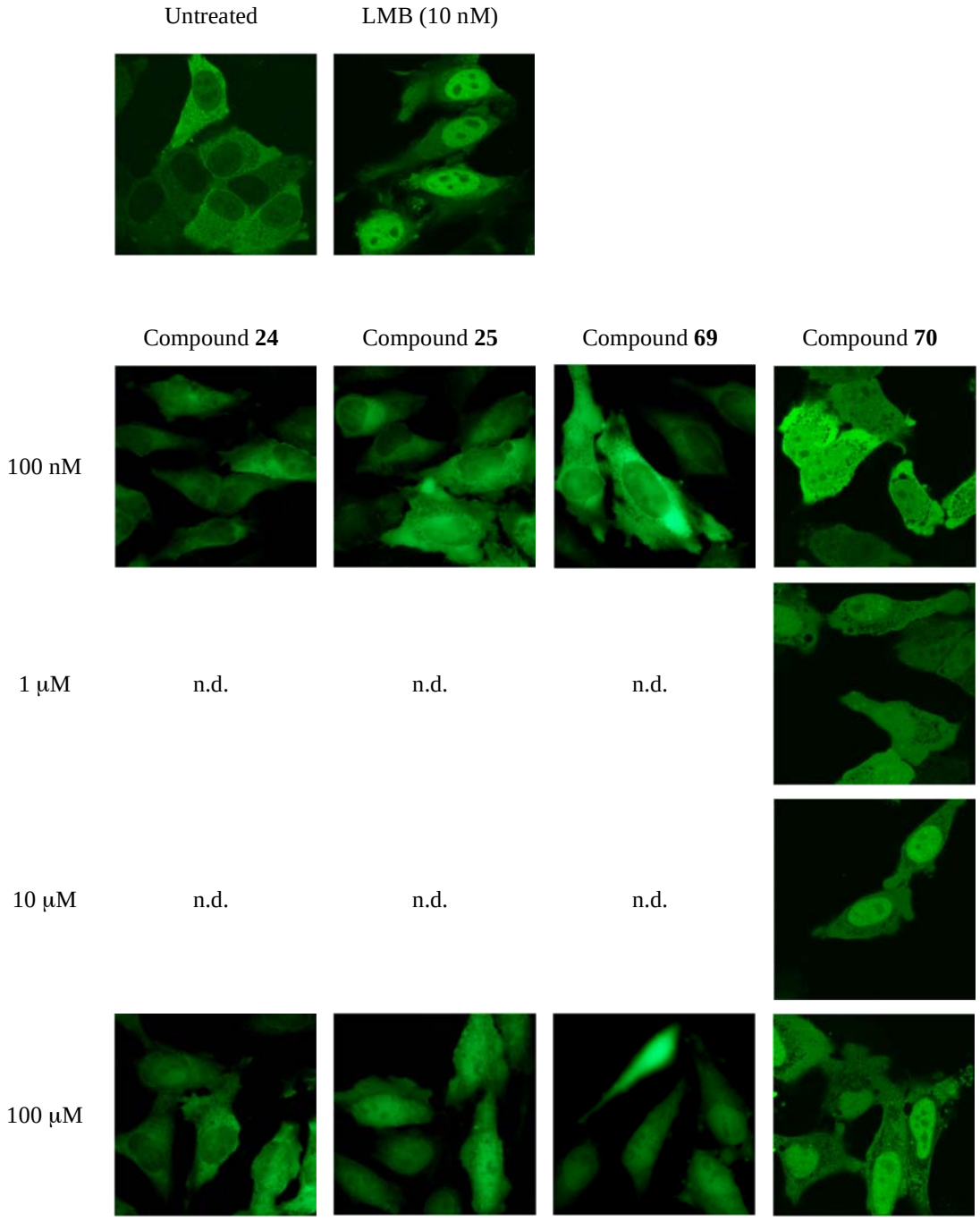


Figure 26: Compounds **24** and **25, 69**, **70** inhibition of CRM1-dependent nuclear export of Rio2 in HeLa cells.

Very interesting results were obtained for the α,β-unsaturated aldehyde **67** and lactone **68**. The results show that at 50 nM, aldehyde **67** shows weak inhibition and at 100 nM full inhibition is observed (Figure 27). This compound displays the same side chain as in natural anguinomycins C and D, but the *Micheal* acceptor has been replaced by the α,β-unsaturated aldehyde resulting in a 10-fold loss in activity. Although ten times less active than the parent compound, at 50 nM it can be considered highly active. More surprising was the high activity displayed by product **68**, which lacks the polyketide chain. The compound caused a small accumulation of the Rio2 protein in the nucleus at 10 nM and full inhibition was observed at 50 nM (Figure 27). This result highlights the fundamental role of the α,β-unsaturated lactone in the inhibition of CRM1-mediated nucleocytoplasmic transport, supporting the thesis that the side chain plays a role in the molecular recognition and modulation of the activity. Even though compound **68** was a drastic simplification of the natural anguinomycins C and D, the activity decreased by less than one order of magnitude. From the synthetic point of view, lactone **68** would be much easier to prepare than the natural compounds resulting in a gain of time and resources.

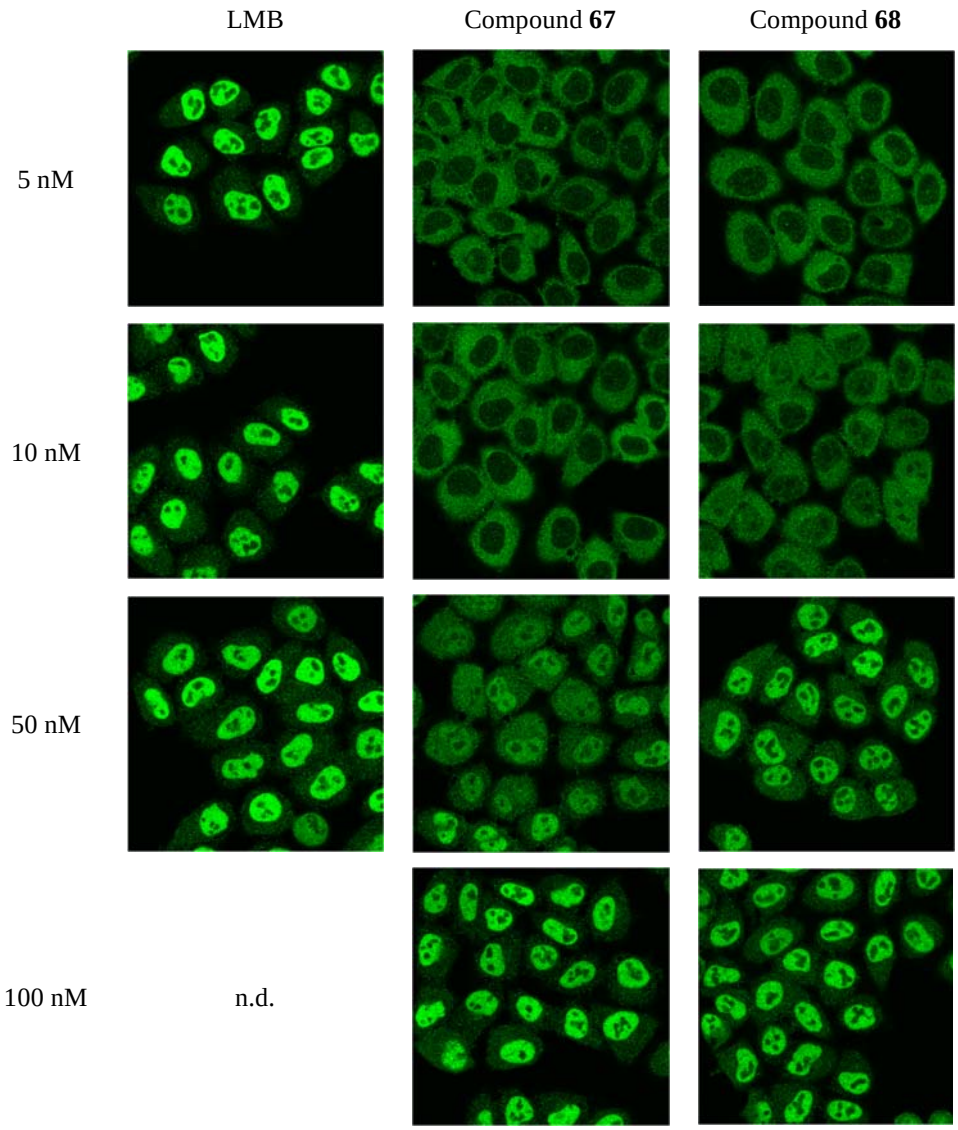


Figure 27: Compounds **67** and **68** inhibition of CRM1-dependent nuclear export of Rio2 in HeLa cells.

The biological results obtained in this work have to be compared with the SAR investigation of other groups on related compounds. *Kobayashi* and co-workers reported biological investigations on callystatin (Figure 2) and its derivatives,[138] proving the fundamental importance of the lactone fragment for the activity. The same studies showed that modifications of the polyketide chain cause a loss in activity of about one order of magnitude compared with the natural callystatin. In addition, inversion of configuration at C(5) causes a 350-fold loss in activity. *Kalesse* and co-workers performed similar investigations on ratjadone (Figure 2),[139] showing again

[138] N. Murakami, M. Sugimoto, M. Kobayashi, *Bioorg. Med. Chem.* **2001**, *9*, 57-67.

[139] M. Kalesse, M. Christmann, U. Bhatt, M. Quitschalle, E. Claus, A. Saeed, A. Burzlaff, C. Kasper, L. O. Haustedt, E. Hofer, T. Scheper, W. Beil, *ChemBioChem* **2001**, *2*, 709-714.

that the lactone was crucial for activity. However, for this compound inversion of configuration at C(10) resulted in a complete loss of activity. Recently, *Mutka* and co-workers reported new derivatives of LMB displaying the same potency as the parent compound, but with a higher selectivity towards normal and cancer cells.[31] Structure-activity relationship studies performed on anguinomycins C and D and their derivatives were in agreement with the literature. Derivatives **24** and **25** clearly proved that the absence of the lactone resulted in a complete loss on the activity. Compound **70** displayed a loss in activity of three orders of magnitude compared to the parent compound, which is in agreement with results obtained by *Kobayashi* and co-workers for callystatin derivatives. The high activity of compound **68** (IC_{50} = 50 nM) can be explained by the fact that this derivative is a shortened version of the parent compound, which lacks the polyketide chain but still contains the important lactone moiety. It is possible that modifications in the polyketide chain cause the molecule to change conformation, inducing steric constraints, which reduce its binding affinity for CRM1. These promising results prompt further investigation on derivatives of anguinomycins C and D, which are currently ongoing.

2.5. Conclusion

This chapter was dedicated to the leptomycin family, a class of compounds giving promising results in the domain of cancer research targeting the nucleocytoplasmic transport. We started with a discussion of the biology of these natural products that presently is not fully understood and requires broader investigations. In the second part we analyzed the reported chemistry and especially the efforts concerning callystatin. The need to find more selective inhibitors of nucleocytoplasmic transport and the unveiled structure of anguinomycins C and D prompted us to develop a synthesis for these natural products. With its six unknown stereogenic centers, the lactone ring, the two diene systems and the polyketide chain, anguinomycins present a good target for total synthesis. During the planning of our synthesis we tried to avoid all steps that, as reported for similar compounds, could present problems and compromise the whole synthesis. The chosen disconnections allowed us to get fragments of similar complexity resulting in a highly convergent synthesis and noteworthy steps include the Cr(III) catalyzed hetero *Diels-Alder* gave straightforward access to the dihydropyrane ring in high yield and selectivity. The *Negishi* cross-coupling under stereoinversion furnished the *cis* product for both anguinomycins C and D from a common starting material. Moreover, to date the use of this reaction in total synthesis has not been reported and we have demonstrated its applicability in the domain. Once more, the *Evans* aldol reaction and in this case using the *Seebach* modification of the auxiliary (DIOZ) proved to be of great use for the synthesis of the polyketide chain. The total synthesis also definitively establishes the absolute configuration of anguinomycins C and D as 5*R*,10*R*,16*R*,18*S*,19*R*,20*S*, which as proposed earlier, matches that of LMB. The total syntheses of anguinomycins C and D were achieved in 29 steps with a longest linear sequence of 18 steps from (*R*)-4-isopropyl-5,5-diphenyloxazolidin-2-one (**32**) and with an overall yield of 6.7% and 6.0% respectively. To date no other total syntheses of anguinomycins C and D have been reported in literature and we can only compare our work with the routes proposed by other groups for the preparation of related compounds. Almost all the reported syntheses of compounds belonging to the leptomycin family required a major number of steps. This is valid also for callystatin, even though it displays a shorter polyketide chain compared to the anguinomycins. Unfortunately, for several syntheses the overall yield of the longest linear sequence

could not be calculated because advanced starting materials were employed. Here we report a resume of the number of steps required for all the reported syntheses of members of the leptomycin family (Table 4).

Table 4: Resume of the synthesized members of the leptomycin family

Compound	Group	Year	Steps (total)	Steps longest linear sequence (starting material)
Callystatin	*Kobayashi*	1998	39	18 (Roche ester **LI**)
	Crimmins	1998	37	18 (allyl iodide)
	Smith	2001	32	15 (oxazolidinone **LVIII)**
	Kalesse	2001	28	21 (Roche ester *ent*-**LI)**
	Enders	2002	40	15 (RAMP)
	Marshall	2002	39	18 (Roche ester *ent*-**LI**)
	Lautens	2002	45	27 (cyclohexanal)
	Panek	2004	37	18 (pseudoephedrine)
	Dias	2005	39	20 ((*S*)-2-methyl-1-butanal)
	Micalizio	2008	25	11 ((*S*)-2-methyl-1-butanal)
Leptomycin B	*Kobayashi*	1998	40	25 (geraniol)
(+)-Ratjadone	*Kalesse*	2000	36	19 (Roche ester *ent*-**LI**)
(–)-Ratjadone	*Williams*	2001	48	30 (geraniol)
(–)-Kazusamycin A	*Kuwajima*	2004	56	33 (diethylethoxymetylenemalonate)
Leptofuranin D	*Marshall*	2003	39	25 (Roche ester *ent*-**LI**)
Leptostatin	*Marshall*	2006	43	25 (Roche ester *ent*-**LI**)
Anguinomycin C	***This work***	**2007**	**29**	**18 (diphenyloxazolidinone 32)**
Anguinomycin D	***This work***	**2008**	**29**	**18 (diphenyloxazolidinone 32)**

Following the total syntheses of the two natural compounds we investigated the biology of these products and more precisely the mode of action and the structure-activity relationships. Several derivatives were prepared and submitted for biological evaluation. The results confirmed the crucial importance of the lactone ring for the activity and also that the activity can be modulated by changing the side chain, which

mainly plays a role in the molecular recognition. Both anguinomycins C and D displayed a strong inhibition of the CRM1-mediated nucleocytoplasmic transport at 5 nm, confirming their powerful activity. Moreover, a new compound **68** that caused accumulation of the Rio2 protein in the nucleus at less than 50 nm was identified. This compound was a simplification of the parent natural products and it maintained strong activity. In terms of time and economy, the synthesis of this compound would be a gain compared to the preparation of the natural anguinomycins C and D and its application as a tool in chemical biology or eventually as a drug candidate could be envisaged. These results prompt further research of new strong nucleocytoplasmic transport inhibitors and evaluation of derivatives of anguinomycin C and D are currently under investigation. It is hoped that the work outlined in this chapter will help to better understand the relationship between CRM1 and the leptomycin family and maybe contribute to the search for more powerful and selective nucleocytoplasmic transport inhibitors for cancer treatment.

3. Synthetic Studies on Sporolides

3.1. Isolation, Structure Elucidation and Biological Activity

Sporolides A (**CLXXV**) and B (**CLXXVI**) are complex marine macrolides isolated from the culture broth CNB-392, then assigned as *Salinospora tropica* (Figure 28).[140] The strain was isolated in 1989 from marine sediments (–1 m) near to Chaub Cay, Bahamas and cultivation of this group of actinomycetes required seawater for growth. The actinomycetes genus *Salinospora* is an impressive source of compounds and culture extracts have shown that more than 80% of the produced structures inhibited in vitro growth of human colon carcinoma HCT-116 and 35% displayed antibacterial properties.[141] Among the molecules produced by the genus *Salinospora*, which displayed interesting biological properties were salinosporamide A (**CLXXVII**) (Figure 28), rifamycin, staurosporine, saliniketal and cyclomarin A.[142] Salinosporamide A with its unusual fused γ-lactam-β-lactone ring structure was the first compound isolated from the strain *Salinospora tropica*.[143] The potent biological activity as a proteasome inhibitor of this compound led it, in 2005, to enter into clinical trials for cancer treatment.[144] Further investigation of this strain resulted in the isolation of the two new metabolites, sporolides A and B. These compounds display an interesting architectural structure, featuring 22 out of 24 carbons that are either sp^2 hybridized or oxygenated, 7 rings and 10 stereogenic centers. This molecular complexity makes these structures challenging targets for total synthesis. These molecules are basically formed from two main fragments; a chlorinated cyclopenta[*a*]indene ring and a cyclohexanone fragment. In the first biological assays sporolides A and B did not show activity against human colon carcinoma HCT-116, methicillin-resistant *Staphylococcus aureus* or vancomycin-resistant *Enterococcus faecium*.[140]

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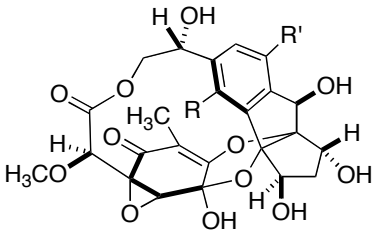
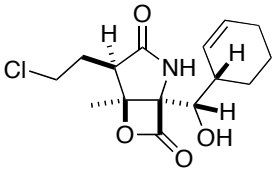
[140] G. O. Buchanan, P. G. Williams, R. H. Feling, C. A. Kauffman, P. R. Jensen, W. Fenical, *Org. Lett.* **2005**, *7*, 2731-2734.
[141] W. Fenical, P. R. Jensen, *Nat. Chem. Biol.* **2006**, *2*, 666-673.
[142] P. R. Jensen, P. G. Williams, D. C. Oh, L. Zeigler, W. Fenical, *Appl. Environ. Microbiol.* **2007**, *73*, 1146-1152.
[143] R. H. Feling, G. O. Buchanan, T. J. Mincer, C. A. Kauffman, P. R. Jensen, W. Fenical, *Angew. Chem., Int. Ed.* **2003**, *42*, 355-357.
[144] W. Fenical, P. R. Jensen, M. A. Palladino, K. S. Lam, G. K. Lloyd, B. C. Potts, *Bioorg. Med. Chem.* **2009**, *17*, 2175-2180.



Sporolide A: R = Cl, R' = H (**CLXXV**)
Sporolide B: R = H, R' = Cl (**CLXXVI**)



Salinosporamide A (**CLXXVII**)

Figure 28: Sporolides A (**CLXXV**) & B (**CLXXVI**) and salinosporamide A (**CLXXVII**).

3.2. Biosynthetic Considerations

The unusual structure of sporolides A (**CLXXV**) and B (**CLXXVI**) encouraged chemists to investigate its biosynthesis. The aromatic moiety of sporolides was hypothesized by *Fenical* and co-workers to derive from an unstable nine-membered ring enediyne precursor, which can undergo a *Bergmann* cyclization[145] with trapping of the biradical by a chlorine source.[146] Sequencing of the *Salinospora tropica* genome by *Moore* and co-workers demonstrated the validity of the hypothesis.[147] In this strain, a very high percentage (9.9%) of the genome devoted to natural product assembly was observed. In the genome there were clusters recognized involved in the biosynthesis of enediyne polyketides. In particular, genes encoding the postulated biosynthesis *via* a nine-membered ring enediyne as well as those encoding the coupling with the cyclohexanone subunit derived from tyrosine were identified (Scheme 53).[148] An interesting point to note was that between 15 type I polyketide synthase-associated modules (PKS) recognized, none contained the whole sequence which would lead to the complete reduction of the carbonyl groups to saturated methylene carbons, in agreement with the highly oxygenated structure of sporolides.[147]

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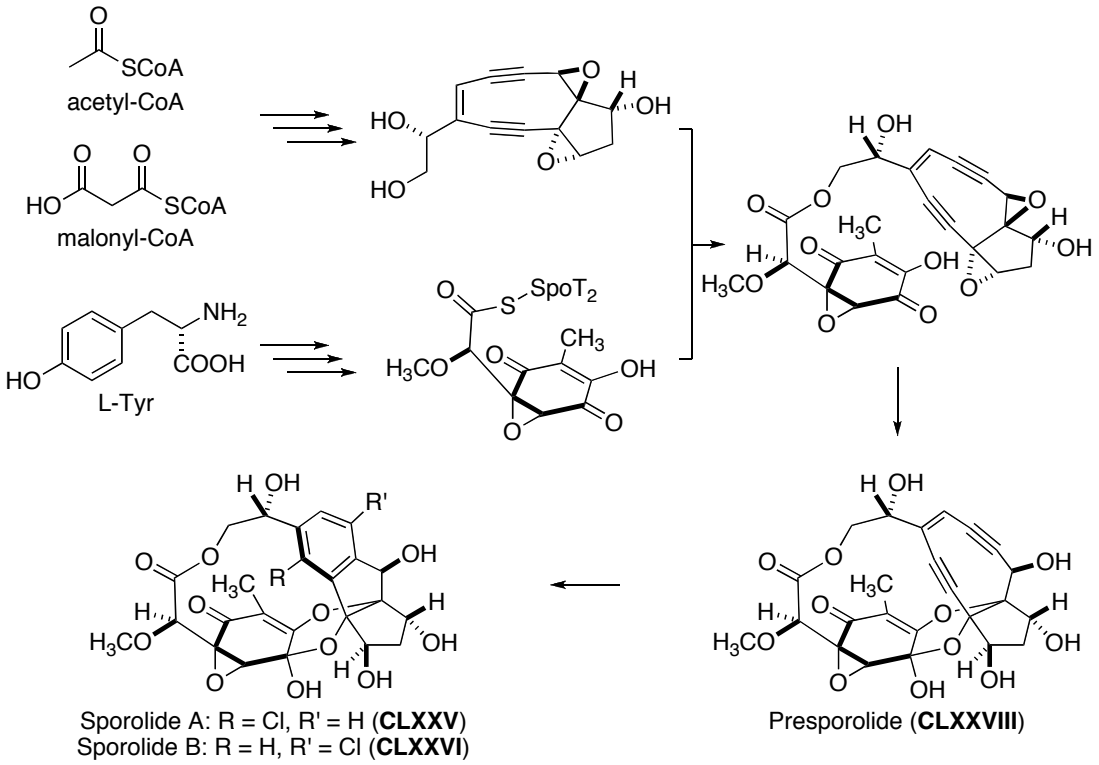
145 R. R. Jones, R. G. Bergman, *J. Am. Chem. Soc.* **1972**, *94*, 660-661.

146 W. Fenical, P. R. Jensen, *Nat. Chem. Biol.* **2006**, *2*, 666-673.

147 D. W. Udwary, L. Zeigler, R. N. Asolkar, V. Singan, A. Lapidus, W. Fenical, P. R. Jensen, B. S. Moore, *Proc. Natl. Acad. Sci. U. S. A.* **2007**, *104*, 10376-10381.

148 R. P. McGlinchey, M. Nett, B. S. Moore, *J. Am. Chem. Soc.* **2008**, *130*, 2406-2407.



Scheme 53: Biosynthetic pathway for sporolides A (**CLXXV**) & B (**CLXXVI**) proposed by Moore and co-workers based on *Salinospora tropica* genome sequencing.[148]

3.3. Enediyne Natural Products as Antitumor Agents

3.3.1. History, Mode of Action, Activity and Stability of Enediynes

The enediyne antitumor antibiotics are a class of compounds discovered in the mid 1980s with the isolation of neocarzinostatin (**CLXXIX**),[149] calicheamicin (**CLXXX**),[150] esperamicins (**CLXXXI**)[151] and dynemycin (**CLXXXII**)[152] (Figure 29). The unprecedented molecular structure of these compounds as well as their exceptional biological activity engendered great interest in the chemistry community. The popularity of the enediyne increased quickly, resulting in hundreds of studies and

[149] a) K. Edo, M. Mizugaki, Y. Koide, *Tetrahedron Lett.* **1985**, *26*, 331-334; b) A. G. Myers, P. J. Proteau, T. M. Handel, *J. Am. Chem. Soc.* **1988**, *110*, 7212-7214.

[150] a) M. D. Lee, T. S. Dunne, M. M. Siegel, C. C. Chang, G. O. Morton, D. B. Borders, *J. Am. Chem. Soc.* **1987**, *109*, 3464-3466; b) M. D. Lee, T. S. Dunne, C. C. Chang, G. A. Ellestad, M. M. Siegel, G. O. Morton, W. J. McGahren, D. B. Borders, *J. Am. Chem. Soc.* **1987**, *109*, 3466-3468.

[151] a) J. Golik, J. Clardy, G. Dubay, G. Groenewold, H. Kawaguchi, M. Konishi, B. Krishnan, H. Ohkuma, K. I. Saitoh, T. W. Doyle, *J. Am. Chem. Soc.* **1987**, *109*, 3461-3462; b) J. Golik, G. Dubay, G. Groenewold, H. Kawaguchi, M. Konishi, B. Krishnan, H. Ohkuma, K. I. Saitoh, T. W. Doyle, *J. Am. Chem. Soc.* **1987**, *109*, 3462-3464.

[152] a) M. Konishi, H. Ohkuma, K. Matsumoto, T. Tsuno, H. Kamei, T. Miyaki, T. Oki, H. Kawaguchi, G. D. VanDuyne, J. Clardy, *J. Antibiot.* **1989**, *42*, 1449-1452; b) M. Konishi, H. Ohkuma, T. Tsuno, T. Oki, G. D. VanDuyne, J. Clardy, *J. Am. Chem. Soc.* **1990**, *112*, 3715-3716.

synthesis papers. Today, compounds presenting an enediyne core are considered extremely potent antitumor antibiotics and activity in the femtomolar range has been reported.[153] Encouraging results from this class of compounds have been reported, *e.g.* the antibody-calicheamicin conjugate (Mylotarg®) (**V**, Figure 1), which is used to treat acute myelogenous leukemia.[154]

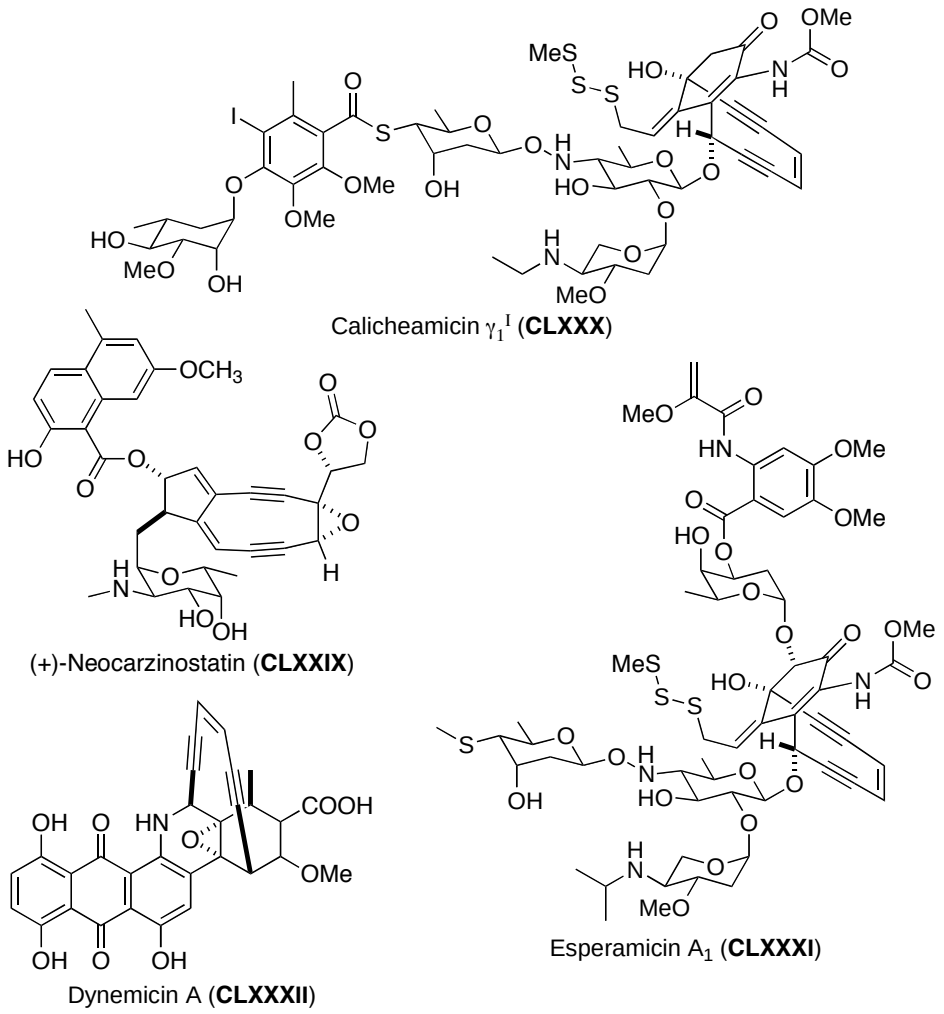


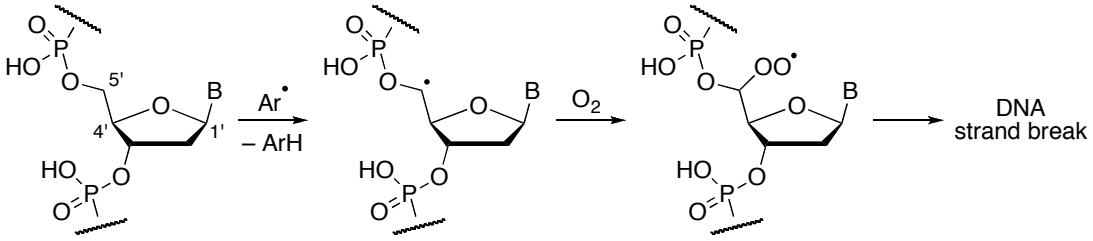
Figure 29: First enediynes antitumor antibiotics discovered: (+)-neocarzinostatin (**CLXXIX**), calicheamicin γ^I (**CLXXX**), esperamicin (**CLXXXI**) and dynemicin A (**CLXXXII**).

The impressive biological activity of the enediyne antitumor antibiotics is directly derived from their ability to generate a diradical species *via Bergman* cyclization[145] and induce DNA strand cleavage. *Bergman* reported in 1972 the thermal cyclization

[153] P. R. Hamann, L. M. Hinman, I. Hollander, C. F. Beyer, D. Lindh, R. Holcomb, W. Hallett, H. R. Tsou, J. Upeslacis, D. Shochat, A. Mountain, D. A. Flowers, I. Bernstein, *Bioconjugate Chem.* **2002**, *13*, 47-58.

[154] P. R. Hamann, J. I. Upeslacis, D. B. Borders, in *Anticancer Agents from Natural Products* (Eds.: G. M. Cragg, D. G. I. Kingston, D. J. Newman), Eds. Taylor & Francis, Boca Raton London New York Singapore, **2005**, pp. 451-473.

of (*Z*)-3-ene-1,5-diyne species *via* the benzene-1,4-diyl radical, but elevated temperature were required. Several investigations were reported understanding the factors that influence the cycloaromatization process in natural enediynes. There are two important factors, proximity of the carbons atoms forming the new C-C bond, which has to be between 3.4 Å and 2.9 Å for a spontaneous cyclization at room temperature and the ring strain.[155] In natural enediynes the cycloaromatization occurs spontaneously at physiological temperature generating the diradical species, which induces DNA breaking (Scheme 54).[156] The DNA double-helix cleavage can be summarized in four steps: a) recognition of specific structural feature attached to the enediyne and binding to DNA; b) activation of the enediyne; c) diradical formation *via Bergman* cyclization; d) abstraction of a proton of deoxyribose in DNA inducing strain cleavage.[157] Depending on the endiyne natural product, proton abstraction can be preferentially initiated at different positions of the deoxyribose, generally 5', 4' or 1'.[158]



Scheme 54: DNA strand clevage initiated at C5' by proton abstraction. The same mechanism applies for initiation at C4' and C1'. B = nucleobase, Ar• = radical generated by *Bergmann* cycloaromatization.

Nicolaou and co-workers recognized at least three important functional domains characterizing these classes of natural products: a) the "warhead" which is responsible for the activity by generating the DNA damaging fragment; b) the "delivery system" that carries the warhead to the target; and c) the "triggering device" that when activated initiates the cascade reaction forming the active diradical.[158] Natural enediynes are labile molecules and they can be divided into two classes, the 9- and the 10-membered ring unit. The latter are more stable than the related 9-membered rings,

[155] M. Kar, A. Basak, *Chem. Rev.* **2007**, *107*, 2861-2890.
[156] D. M. Lopez-Larraza, K. Moore Jr, P. C. Dedon, *Chem. Res. Toxicol.* **2001**, *14*, 528-535.
[157] J. W. Grissom, G. U. Gunawardena, D. Klingberg, D. Huang, *Tetrahedron* **1996**, *52*, 6453-6518.
[158] K. C. Nicolaou, W. M. Dai, *Angew. Chem., Int. Ed.* **1991**, *30*, 1387-1416.

which need a stabilizing protein to avoid undergoing cycloaromatization.[159] The 9-membered ring enediynes are usually isolated as a non-covalently bound complex with their respective apoprotein, which prevents the cycloaromatization of the chromophore.[160] However, the protein cannot fully stabilize the highly reactive chromophore, which decomposes upon aging. A second goal of the chromoprotein is also to act as a shuttle to deliver the active enediyne to the target, the DNA.[159] The challenge for organic chemists is to prepare enediyne system having a "decent half-life" (10-36 hours at biological temperature, 37 °C) or stable precursors that can be activated to induce cycloaromatization.[155] Today, the enediyne antitumor antibiotics and their derivatives remain lead candidates in the battle against cancer.[161] The research in this field continues and efforts to find new potent and selective compounds have given encouraging results, *e.g.* the hybrid antibody-calicheamicin conjugate (Mylotarg®) (**V**, Figure 1) and neocarzinostatin (**CLXXIX**, Figure 29) which are used in cancer therapy,[154] or dynemicin prodrugs which can selectively be activated in the tumor cells.[162]

3.3.2. Nine-Membered Ring Natural Endiynes

3.3.2.1. The Neocarzinostatin Chromophore

Neocarzinostatin (NCS) was isolated in 1965 from *Streptomyces carzinostaticus* Var. F41,[163] but its structure was not elucidated until twenty years later.[149] The chromophore (**CLXXIX**) was isolated as a 1:1 complex with its apoprotein composed of a 113 amino acid polypeptide chain.[164] Several synthetic studies have been reported on this compound, but only two total syntheses of the NCS aglycon[165] and one of the

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159 a) N. Zein, R. Reiss, M. Bernatowicz, M. Bolgar, *Chem. Biol.* **1995**, *2*, 451-455; b) J. Kandaswamy, P. Hariharan, T. K. S. Kumar, C. Yu, T. J. Lu, D. H. Chin, *Anal. Biochem.* **2008**, *381*, 18-26.

160 T. Usuki, M. Inoue, M. Hirama, T. Tanaka, *J. Am. Chem. Soc.* **2004**, *126*, 3022-3023.

161 M. Gredicak, I. Jeric, *Acta Pharmaceutica* **2007**, *57*, 133-150.

162 S. C. Sinha, L. S. Li, G. P. Miller, S. Dutta, C. Rader, R. A. Lerner, *Proc. Natl. Acad. Sci. U. S. A.* **2004**, *101*, 3095-3099.

163 N. Ishida, K. Miyazaki, K. Kumagai, M. Rikimaru, *J. Antibiot.* **1965**, *18*, 68-76.

164 A. Teplyakov, G. Obmolova, K. Wilson, K. Kuromizu, *Eur. J. Biochem.* **1993**, *213*, 737-741.

165 a) A. G. Myers, M. Hammond, Y. Wu, J. N. Xiang, P. M. Harrington, E. Y. Kuo, *J. Am. Chem. Soc.* **1996**, *118*, 10006-10007; b) S. Kobayashi, M. Hori, G. X. Wang, M. Hirama, *J. Org. Chem.* **2006**, *71*, 636-644.

entire chromophore have been published.[166] The NCS chromophore displays potent antitumor and antibacterial activity *via* oxygen-mediated DNA cleavage.[167] In 1987 *Myers* proposed the mode of action of the chromophore; in which the nucleophilic attack of a thiol group at C(12) induced the *Bergmann* cycloaromatization *via* a cumulene intermediate (Scheme 55).[168] The naphtoate residue of the NCS chromophore intercalates into the DNA positioning the enediyne for the DNA cleavage.[169] Moreover, several studies to elucidate the details of the DNA cleavage were reported, supporting other mechanisms than the proton abstraction at the 4'-position of deoxyribose in DNA.[170] The clinical applications of NCS were initially limited due to its extreme toxicity. The problem was later overcome by conjugation of the chromophore with a biocompatible polymer. The poly(styrene-co-maleic acid)-NCS conjugate (SMANCS) displays high biological activity and high tumor-targeting efficiency and has been approved in Japan for the treatment of liver cancer.[171]

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166 a) A. G. Myers, J. Liang, M. Hammond, P. M. Harrington, Y. Wu, E. Y. Kuo, *J. Am. Chem. Soc.* **1998**, *120*, 5319-5320; b) A. G. Myers, R. Glatthar, M. Hammond, P. M. Harrington, E. Y. Kuo, J. Liang, S. E. Schaus, Y. Wu, J.-N. Xiang, *J. Am. Chem. Soc.* **2002**, *124*, 5380-5401.

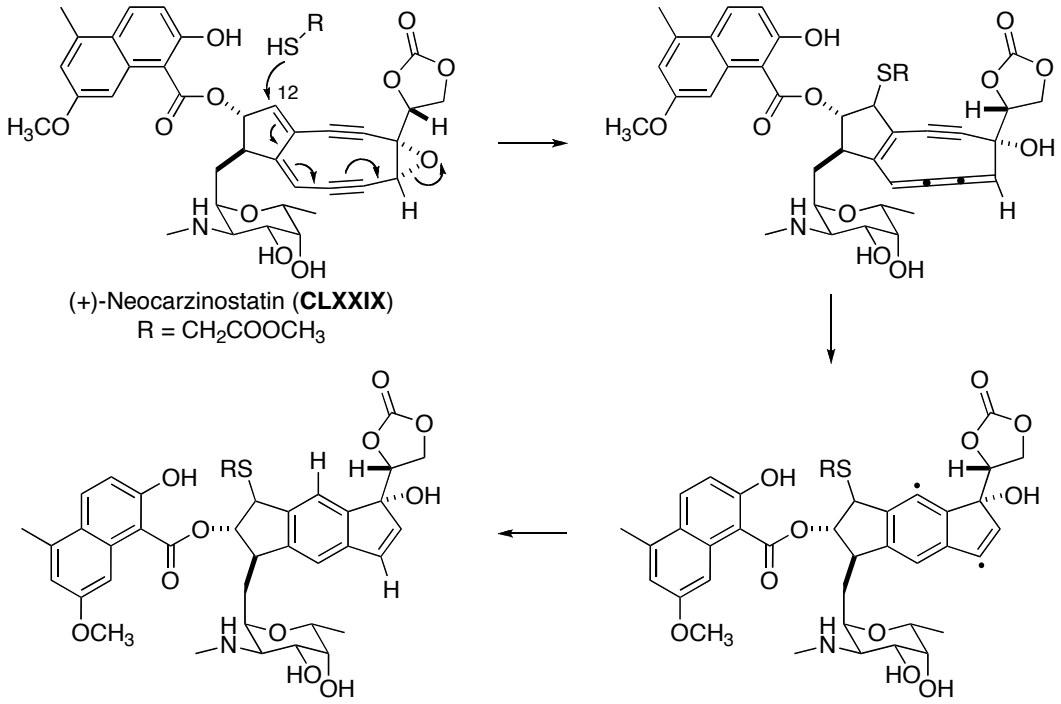
167 a) L. S. Kappen, M. A. Napier, I. H. Goldberg, *Proc. Natl. Acad. Sci. U. S. A.* **1980**, *77*, 1970-1974; b) L. F. Povirk, I. H. Goldberg, *Biochemistry* **1984**, *23*, 6304-6311.

168 A. G. Myers, *Tetrahedron Lett.* **1987**, *28*, 4493-4496.

169 a) L. F. Povirk, N. Dattagupta, B. C. Warf, I. H. Goldberg, *Biochemistry* **1981**, *20*, 4007-4014; b) S. H. Lee, I. H. Goldberg, *Biochemistry* **1989**, *28*, 1019-1026.

170 a) L. S. Kappen, I. H. Goldberg, *Biochemistry* **1983**, *22*, 4872-4878; b) D. H. Chin, L. S. Kappen, I. H. Goldberg, *Proc. Natl. Acad. Sci. U. S. A.* **1987**, *84*, 7070-7074; c) I. Saito, H. Kawabata, T. Fujiwara, H. Sugiyama, T. Matsuura, *J. Am. Chem. Soc.* **1989**, *111*, 8302-8303; d) B. L. Frank, L. Worth Jr, D. F. Christner, J. W. Kozarich, J. Stubbe, L. S. Kappen, I. H. Goldberg, *J. Am. Chem. Soc.* **1991**, *113*, 2271-2275; e) D. H. Chin, I. H. Goldberg, *Biochemistry* **1993**, *32*, 3611-3616; f) L. S. Kappen, I. H. Goldberg, *Science* **1993**, *261*, 1319-1321.

171 H. Maeda, in *Enediyne Antibiotics as Antitumor Agents* (Eds.: D. B. Borders, T. W. Doyle), Marcel Dekker, New York, **2005**, pp. 363-382.



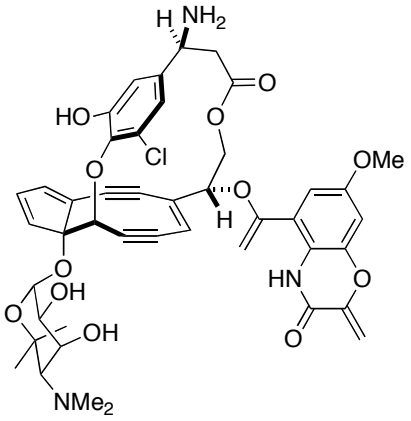
Scheme 55: Mode of action of the NCS chromophore (**CLXXIX**) proposed by *Myers* and co-workers. Thiol attack at C(12) induces *Bergmann* cycloaromatization *via* a cumulene intermediate.

3.3.2.2. The C-1027 Chromophore

The chromoprotein enediyne natural product C-1027 was isolated in 1988 from *Streptomyces globisporus* C-1027[172] as a 1:1 complex of the chromophore (**CLXXXIII**) (Figure 30) with its apoprotein composed of 110 amino acid.[173] The free chromophore is highly labile and cycloaromatization spontaneously occurrs in ethanol at 25 °C with a half-life of 50 minutes. Even though the synthesis of advanced intermediates of the C-1027 chromophore have been published,[174] no total synthesis has been reported. This antibiotic displays an extremely potent anticancer activity towards several tumors. Its cytotoxicity is higher than that of the previously discussed NCS chromophore and the cycloaromatization *via p*-benzene diradical spontaneously

[172] J. Hu, Y. C. Xue, M. Y. Xie, R. Zhang, T. Otani, Y. Minami, Y. Yamada, T. Marunaka, *J. Antibiot.* **1988**, *41*, 1575-1579.
[173] a) T. Otani, Y. Minami, T. Marunaka, R. Zhang, M. Y. Xie, *J. Antibiot.* **1988**, *41*, 1580-1585; b) T. Otani, Y. Minami, K. Sakawa, K. Yoshida, *J. Antibiot.* **1991**, *44*, 564-568; c) T. Matsumoto, Y. Okuno, Y. Sugiura, *Biochem. Biophys. Res. Commun.* **1993**, *195*, 659-666.
[174] a) M. Inoue, T. Sasaki, S. Hatano, M. Hirama, *Angew. Chem., Int. Ed.* **2004**, *43*, 6500-6505; b) M. Inoue, I. Ohashi, T. Kawaguchi, M. Hirama, *Angew. Chem., Int. Ed.* **2008**, *47*, 1777-1779.

occurs without presence of the thiol group.[175] To date no clinical use of C-1027 has been reported, but investigations are under way.[176]



C-1027 chromophore (**CLXXXIII**)

Figure 30: The C-1027 chromophore (**CLXXXIII**).

3.3.2.3. The Maduropeptin Chromophore

The maduropeptin chromophore (**CLXXXIV**) was isolated in 1991 from *Nomadura madurae* as a 1:1 complex with its acidic, water-soluble apoprotein of 32 kDa.[177] Several synthetic studies have been reported, but only a single total synthesis of the aglycon chromophore has been published.[178] This 9-membered ring enedyine displayed potent antitumoral and antibacterial properties[177] resulting in a mixture of single- and double-strand cleavage of DNA.[179] The labile chromophore can be dissociated from the carrier protein by treatment with methanol, forming the corresponding methanol adduct (**CLXXXV**) (Scheme 56). The methanol adduct represent a stable prodrug of the labile enedyine chromophore and a mechanism of action starting from this stabilized adduct has been postulated.[180] To date no clinical application of this compound has been reported.

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[175] Y. J. Xu, Y. S. Zhen, I. H. Goldberg, *Biochemistry* **1994**, *33*, 5947-5954.

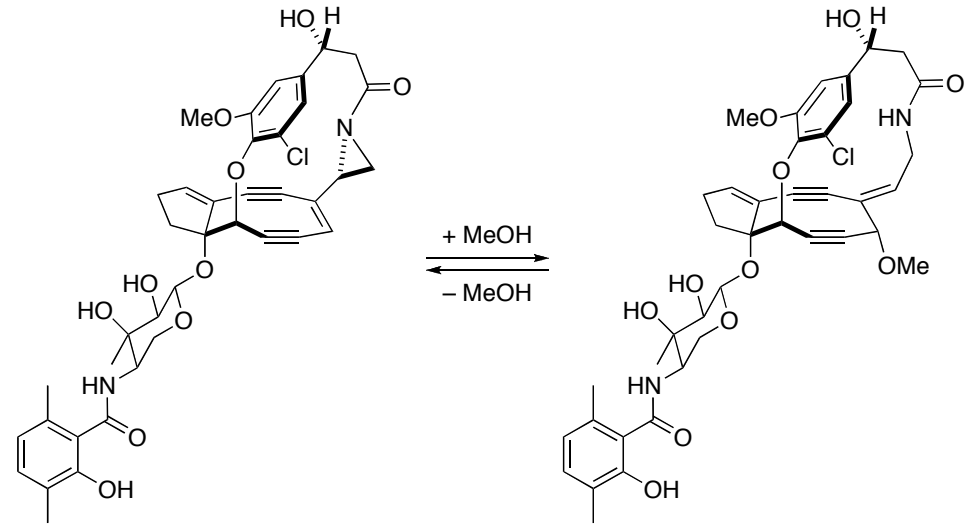
[176] M. Inoue, *Bull. Chem. Soc. Jpn.* **2006**, *79*, 501-510.

[177] M. Hanada, H. Ohkuma, T. Yonemoto, K. Tomita, M. Ohbayashi, H. Kamei, T. Miyaki, M. Konishi, H. Kawaguchi, S. Forenza, *J. Antibiot.* **1991**, *44*, 403-414.

[178] K. Komano, S. Shimamura, M. Inoue, M. Hirama, *J. Am. Chem. Soc.* **2007**, *129*, 14184-14186.

[179] N. Zein, W. Solomon, K. L. Colson, D. R. Schroeder, *Biochemistry* **1995**, *34*, 11591-11597.

[180] D. R. Schroeder, K. L. Colson, S. E. Klohr, N. Zein, D. R. Langley, M. S. Lee, J. A. Matson, T. W. Doyle, *J. Am. Chem. Soc.* **1994**, *116*, 9351-9352.



Maduropeptin chromophore (**CLXXXIV**) Methanol adduct (**CLXXXV**)

Scheme 56: The maduropeptin chromophore (**CLXXXIV**) and its corresponding methanol adduct (**CLXXXV**).

3.3.2.4. The Kedarcidin Chromophore

The chromoprotein antitumor antibiotic kedarcidin was isolated in 1991 from a culture of actinomycete strain L585-6.[181] The structure of the chromophore (**CLXXXVI**) was elucidated one year later (Figure 31).[182] Due to the high instability of the chromophore, its structure has been characterized through a series of revisions of the configuration. The first one in 1997 by *Hirama* and co-workers[183] and the second one in 2007 by *Myers* and co-workers.[184] In between, *Myers* and co-workers reported the first synthesis of the originally proposed aglycon chromophore,[185] which was then corrected with the right configuration in the second synthesis in 2007.[184] A second synthesis of the kedarcidin aglycon chromophore was recently reported by *Hirama* and co-workers.[186] Similarly to the previously presented 9-membered ring enediynes, the kedarcidin chromophore also displays strong activity against several

[181] a) K. S. Lam, G. A. Hesler, D. R. Gustavson, A. R. Crosswell, J. M. Veitch, S. Forenza, K. Tomita, *J. Antibiot.* **1991**, *44*, 472-478; b) S. J. Hofstead, J. A. Matson, A. R. Malacko, H. Marquardt, *J. Antibiot.* **1992**, *45*, 1250-1254.

[182] a) J. E. Leet, D. R. Schroeder, S. J. Hofstead, J. Golik, K. L. Colson, S. Huang, S. E. Klohr, T. W. Doyle, J. A. Matson, *J. Am. Chem. Soc.* **1992**, *114*, 7946-7948; b) J. E. Leet, D. R. Schroeder, D. R. Langley, K. L. Colson, S. Huang, S. E. Klohr, M. S. Lee, J. Golik, S. J. Hofstead, T. W. Doyle, J. A. Matson, *J. Am. Chem. Soc.* **1993**, *115*, 8432-8443.

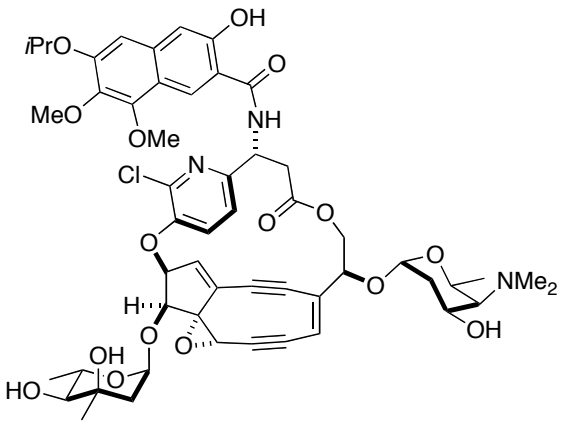
[183] S. Kawata, S. Ashizawa, M. Hirama, *J. Am. Chem. Soc.* **1997**, *119*, 12012-12013.

[184] F. Ren, P. C. Hogan, A. J. Anderson, A. G. Myers, *J. Am. Chem. Soc.* **2007**, *129*, 5381-5383.

[185] A. G. Myers, P. C. Hogan, A. R. Hurd, S. D. Goldberg, *Angew. Chem., Int. Ed.* **2002**, *41*, 1062-1067.

[186] K. Ogawa, Y. Koyama, I. Ohashi, I. Sato, M. Hirama, *Angew. Chem., Int. Ed.* **2009**, *48*, 1110-1113.

tumors with an IC_{50} of 1 nM for HCT116 colon carcinoma cell lines. This enediyne induces single strand cleavage of DNA in a selective way recognizing the TCCT sequence.[187]



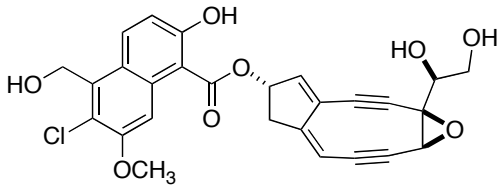
Kedarcidin chromophore (**CLXXXVI**)

Figure 31: The kedarcidin chromophore (**CLXXXVI**).

3.3.2.5. The N1999A2 Enediyne Antibiotic

The N1999A2 antibiotic (**CLXXXVII**) was isolated in 1998 from *Streptomyces* sp. AJ9493 (Figure 32).[188] This 9-membered enediyne ring differs from the previously discussed compounds, as it can be isolated and is stable in the absence of a carrier protein. Structurally, the compound is similar to the neocarzinostatin chromophore, but lacks the presence of the amino sugar. In 2006 *Myers* and co-workers reported the enantioselective synthesis of N1999A2.[189] The N1999A2 antitumor antibiotic displays potent inhibition of various tumor cell lines growth, with *in vivo* IC_{50} values from pico- to nano-molar range. Similarly to the NCS chromophore, N1999A2 also has the naphtoate residue, which can intercalate into the DNA base pairs and the strain cleavage occurs by preferential attack on the thymine base.[190]

[187] N. Zein, K. L. Colson, J. E. Leet, D. R. Schroeder, W. Solomon, T. W. Doyle, A. M. Casazza, *Proc. Natl. Acad. Sci. U. S. A.* **1993**, *90*, 2822-2826.
[188] T. Ando, M. Ishii, T. Kajiura, T. Kameyama, K. Miwa, Y. Sugiura, *Tetrahedron Lett.* **1998**, *39*, 6495-6498.
[189] N. Ji, H. O'Dowd, B. M. Rosen, A. G. Myers, *J. Am. Chem. Soc.* **2006**, *128*, 14825-14827.
[190] N. Miyagawa, D. Sasaki, M. Matsuoka, M. Imanishi, T. Ando, Y. Sugiura, *Biochem. Biophys. Res. Commun.* **2003**, *306*, 87-92.

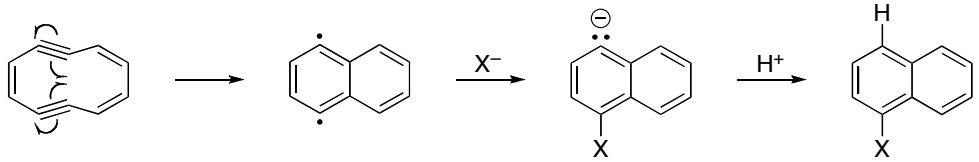


N1999A2 chromophore (**CLXXXVII**)

Figure 32: The N1999A2 chromophore (**CLXXXVII**).

3.4. Total Synthesis and Synthetic Studies on Sporolides

The first synthetic study on sporolides was published by O'Connor and co-workers in 2007. The study demonstrated the possibility of trapping the *p*-benzyne diradical formed *via* Bergman cyclization with a nucleophilic addition of a chlorine anion in the presence of a weak acid (Scheme 57).[191] The result provided an explanation to how the halogen could be incorporated and also why sporolides A and B were isolated as 1:1 mixture (Figure 28).

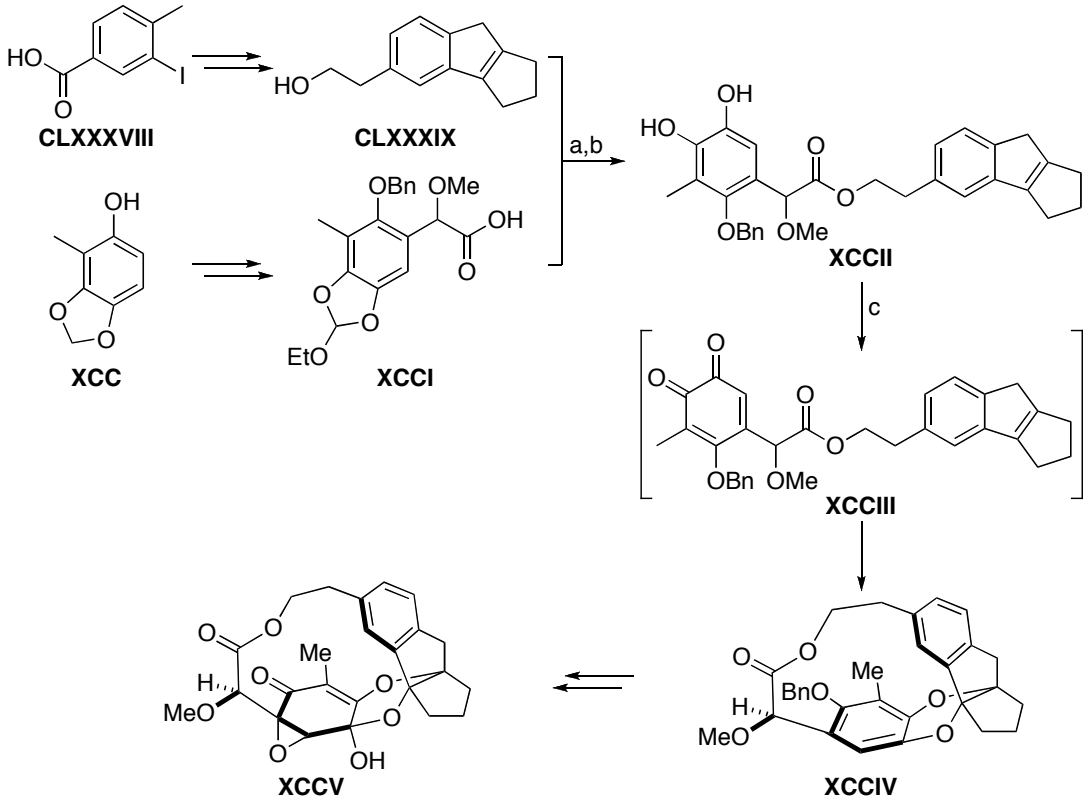


Scheme 57: Proposed mechanism for the generation of haloaromatic compounds *via* halide addition to a *p*-benzene diradical derived from an enediyne.

In 2008, *Nicolaou* and co-workers reported the first synthesis of a model system of sporolide B with an intramolecular [4+2] cycloaddition as a key step (Scheme 58).[192] For this study the chlorinated cyclopenta[*a*]indene ring was simplified removing all the substituents and the fragment (**CLXXXIX**) prepared in ten steps from commercially available 3-iodo-4-methylbenzoic acid (**CLXXXVIII**).[193] The building block (**XCCI**) was synthesized in seven steps as a racemic mixture from the phenol derivative (**XCC**), which was itself prepared in three steps following a known procedure.[194] The two fragments were coupled and the catechol deprotected to give compound (**XCCII**). Treatment with AgO_2 allowed the *in situ* generation of the *o*-

191 C. L. Perrin, B. L. Rodgers, J. M. O'Connor, *J. Am. Chem. Soc.* **2007**, *129*, 4795-4799.
192 K. C. Nicolaou, H. Wang, Y. Tang, *Angew. Chem., Int. Ed.* **2008**, *47*, 1432-1435.
193 S. Shankar, G. Vaidyanathan, D. Affleck, P. C. Welsh, M. R. Zalutsky, *Bioconjugate Chem.* **2003**, *14*, 331-341.
194 X. C. Chen, J. C. Chen, M. De Paolis, J. P. Zhu, *J. Org. Chem.* **2005**, *70*, 4397-4408.

quinone intermediate (**XCCIII**), which directly undergoes a *Diels-Alder* reaction to afford the macrocycle (**XCCIV**) in 60% yield. No details on the selectivity were reported; further modifications afforded the model system (**XCCV**).

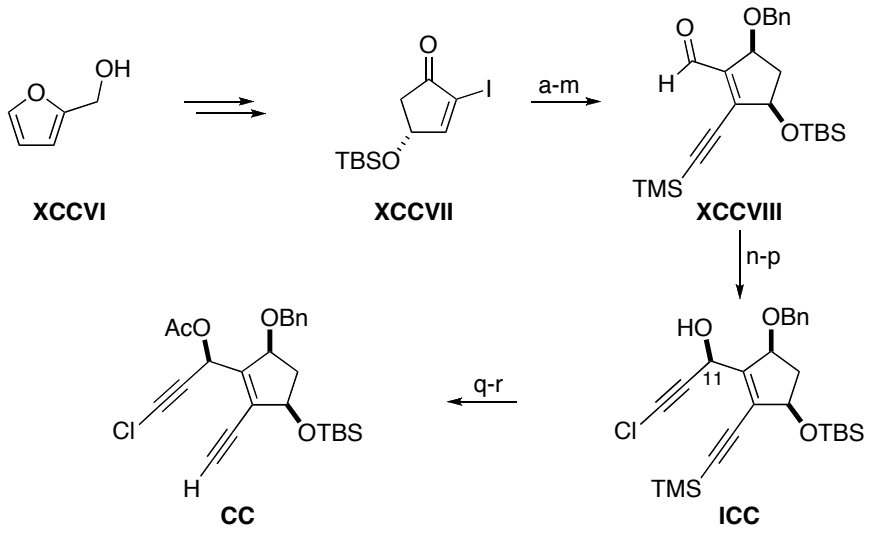


Scheme 58: a) **XCCI** (1.25 equiv), **CLXXXIX** (1.0 equiv), DCC (1.3 equiv), DMAP (0.2 equiv), CH_2Cl_2, 25 °C, 3 h, 84%; b) $TsOH•H_2O$, MeOH, 25 °C, 24 h, 98%; c) Ag_2O (2.0 equiv), toluene (0.005 M), 120 °C, 1 h, 60%.

Recently, *Nicolaou* and co-workers reported the first total synthesis of sporolide B (**CLXXVI**).[195] For the synthesis they did not opt for a biosynthetic approach based one enediyne intermediate, but on intramolecular cycloadditions. The chlorobenzenoid indane motif was synthesized *via* a ruthenium-catalyzed intermolecular [2+2+2] cycloaddition and the macrocycle furnishing the sporolide B skeleton *via* the previously presented *Diels-Alder* reaction. The synthesis of the chlorinated cyclopenta[*a*]indene ring fragment started from iodoenone (**XCCVII**), which was synthesized in nine steps from furfuryl alcohol (**XCCVI**).**78**[,196] Iodoenone (**XCCVII**) was transformed to aldehyde (**XCCVIII**) in thirteen steps using standard chemistry. The product was subsequently treated with a solution of

[195] K. C. Nicolaou, Y. Tang, J. Wang, *Angew. Chem., Int. Ed.* **2009**, *48*, 3449-3453.
[196] T. T. Curran, D. A. Hay, C. P. Koegel, J. C. Evans, *Tetrahedron* **1997**, *53*, 1983-2004.

lithiochloroacetylene in situ prepared from *cis*-1,2-dichloroethylene to afford the alcohol product with the undesired stereochemistry at C(11). The problem was solved using an oxidation and reduction sequence to afford alcohol (**ICC**). Two additional steps of protecting group manipulation gave the acetoxy chloroacetylene fragment (**CC**) (Scheme 59).

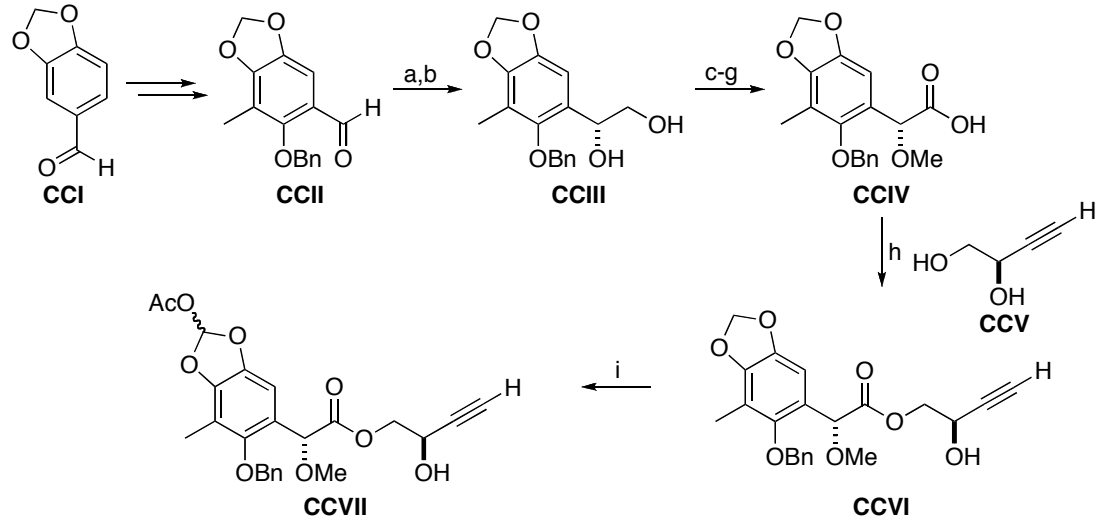


Scheme 59: a) $NaBH_4$, $CeCl_3$•$7H_2O$, MeOH, –78 °C, 1 h; b) NaH, THF, 0 °C, 30 min; then BnBr, TBAI, THF, 0 → 25 °C, 16 h, 95% (2 steps); c) $PdCl_2(PPh_3)_2$ (0.05 equiv), Et_3N, CO (balloon pressure), MeOH, 70 °C, 3 h, 95%; d) DIBAL-H (1.0 M toluene), toluene, –78 → 10 °C, 1 h, 95%; e) DHP (1.5 equiv), TsOH•H_2O, CH_2Cl_2, 0 °C, 30 min; f) TBAF (1.0 M THF), THF, 25 °C, 3 h; g) DMP, $NaHCO_3$, CH_2Cl_2, 25 °C, 30 min, 83% (3 steps); h) I_2, CH_2Cl_2/pyridine (1:1), 25 °C, 15 h, 80%; i) $NaBH_4$, $CeCl_3$•$7H_2O$, MeOH, –78 °C, 1 h; j) TBSCl, imidazole, DMAP, CH_2Cl_2, 25 °C, 3 h, 94% (2 steps); k) TMS-acetylene, $PdCl_2(PPh_3)_2$ (0.02 equiv), CuI (0.04 equiv), Et_2NH, 25 °C, 16 h, 98%; l) Et_2AlCl (1.8 M toluene), CH_2Cl_2, –25 → 25 °C, 2 h, 99%; m) DMP, $NaHCO_3$, CH_2Cl_2, 25 °C, 1 h, 79%; n) cis-1,2-dichloroethylene, MeLi (1.6 M Et_2O), Et_2O, 0 °C, 30 min, then **XCCVIII**, Et_2O, 0 °C, 10 min; o) DMP, $NaHCO_3$, CH_2Cl_2, 25 °C, 1 h, 93% (2 steps); p) DIBAL-H (1.0 M toluene), toluene, –78 °C, 30 min, 81%; q) K_2CO_3, MeOH, 25 °C, 1 h, 99%; r) Ac_2O, Et_3N, DMAP, CH_2Cl_2, 0 °C, 30 min, 98%.

The second building block, featuring the cyclohexanone subunit, started from aldehyde (**CCII**), which was prepared in four steps from commercially available piperonal (**CCI**) following a known procedure.[197] Following *Wittig* reaction and *Sharpless* asymmetric dihydroxylation using AD-mix-β afforded diol (**CCIII**) in 98% *ee*. The product was converted to the carboxylic acid (**CCIV**) in five additional steps and then coupled with the acetylinic alcohol (**CCV**) to afford hydroxy ester (**CCVI**) in 73% yield. The acetylinic alcohol (**CCV**) was prepared in four steps from

[197] N. Saito, K. Tashiro, Y. Maru, K. Yamaguchi, A. Kubo, *J. Chem. Soc., Perkin Trans. 1* **1997**, 53-69.

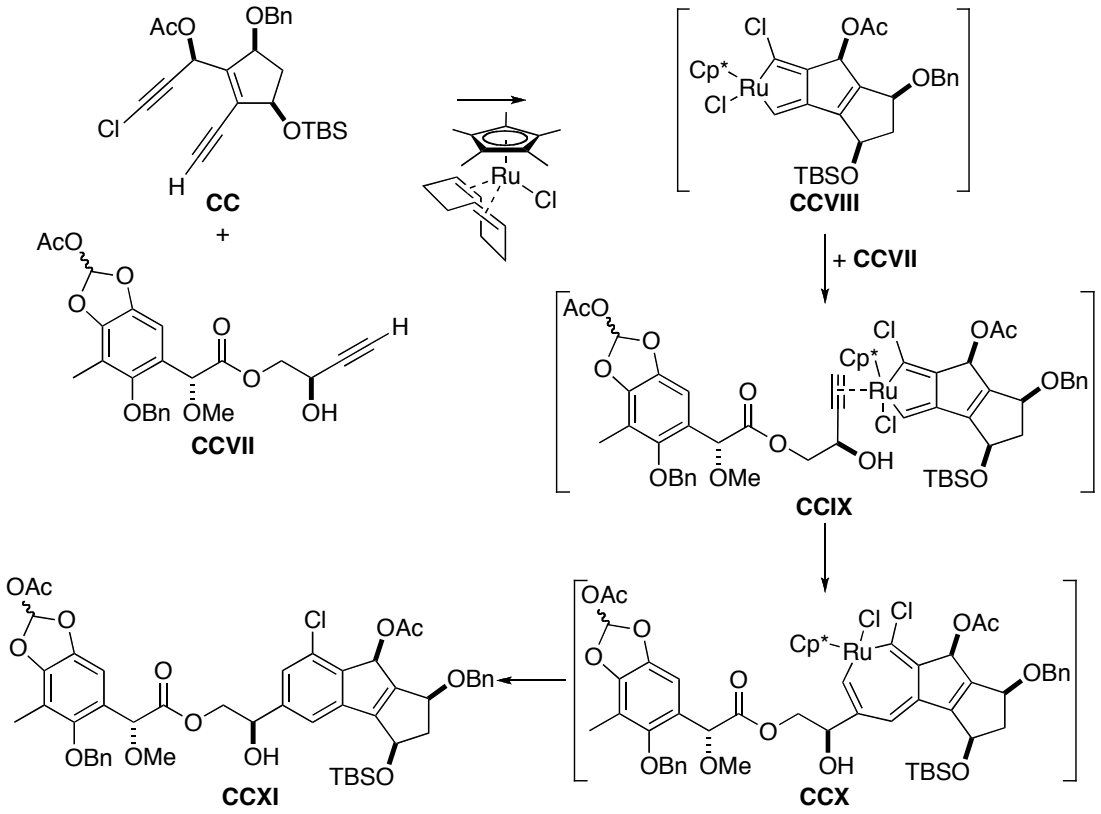
commercially available (+)-2,3-*O*-isopropylidene-L-threitol[198] or in two more additional steps from commercially available L-diethyl tartrate.[199] Treatment of hydroxy ester (**CCVI**) with $Pb(OAc)_4$ afforded the target compound (**CCVII**) in 89% yield as a 8:1 mixture of diastereoisomers (Scheme 60).



Scheme 60: a) $MePPh_3Br$, KHMDS (1.0 M toluene), THF, 0 °C, 30 min, then **CCII**, THF, –78 °C → 0 °C, 30 min, 98%; b) AD-mix-*β*, *t*BuOH/H_2O (1:1), 25 °C, 8 h, 96%; c) TBSCl, Et_3N, DMAP, CH_2Cl_2, 25 °C, 8 h, 99%; d) *t*BuOK, MeI, MeCN, 0 → 25 °C, 16 h, 95%; e) TBAF (1.0 M THF), THF, 25 °C, 16 h, 99%; f) DMP, $NaHCO_3$, CH_2Cl_2, 25 °C, 1 h, 78%; g) $NaClO_2$, NaH_2PO_4•$2H_2O$, 2-methyl-2-butene, *t*BuOH/H_2O (1:1), 25 °C, 30 min, 96%; h) 20, EDCI, DMAP, CH_2Cl_2, 25 °C, 3 h, 73%; i) $Pb(OAc)_4$, benzene, 75 °C, 1 h, 89%.

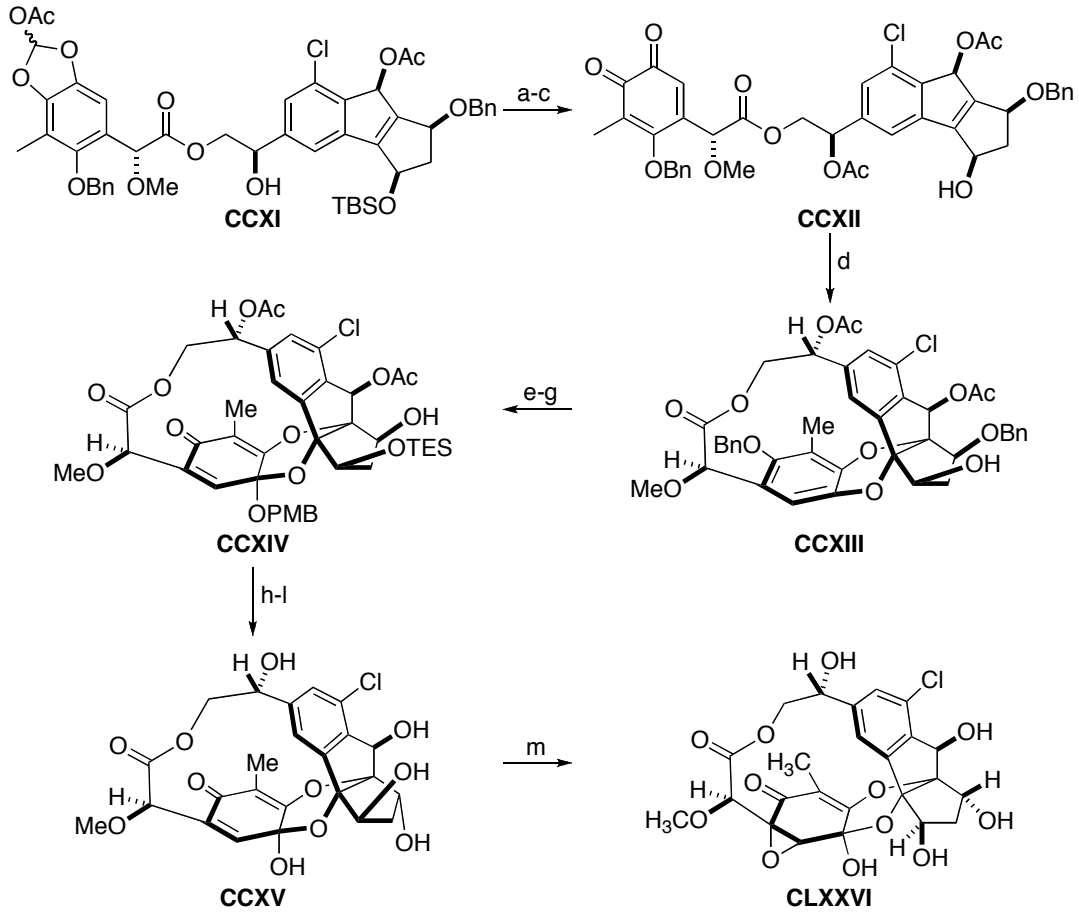
The [2+2+2] cycloaddition between acetylene fragment (**CC**) and the terminal alkyne (**CCVII**), catalyzed by [Cp*RuCl(cod)] proceeded smoothly to afford compound (**CCXI**) in 87% yield. The product was obtained as a single regioisomer and a proposition of the mechanism is proposed in scheme 61.

[198] J. S. Yadav, M. C. Chander, B. V. Joshi, *Tetrahedron Lett.* **1988**, *29*, 2737-2740.
[199] P. W. Feit, *J. Med. Chem.* **1964**, *7*, 14-17.



Scheme 61: CC (1.0 equiv), **CCVII** (1.1 eq.), Cp*RuCl(cod) (0.07 equiv), DCE, 25 °C, 30 min, 87%.

Two additional protecting group manipulations and an oxidation of the catechol to the *o*-quinone gave compound (**CCXII**). The key *Diels-Alder* reaction, which was previously explored in the model system (Scheme 58),[192] was attempted by heating (**CCXII**) in toluene and afforded the desired product (**CCXIII**) in 21% yield (50% recovered starting material). The reaction took place with remarkable diasteroselectivity, probably due to sterics reasons induced by the substituents on the dienophile. Two more steps of protecting group modification followed by treatment with $PhI(OCOCF_3)_2$ in presence of PMBOH gave the *p*-ketal quinone (**CCXIV**) in 75% yield. Five additional steps furnished deoxysporolide (**CCXV**), which was subjected to the final epoxidation with *t*BuOOH and DBU to afford sporolide B (**CLXXVI**) in 63% yield (Scheme 62). To date this remains the only reported total synthesis of sporolide B and it required 58 steps from commercially available starting materials (longest linear sequence of 41 steps from furfuryl alcohol (**XCCVI**)).



Scheme 62: a) Ac_2O, Et_3N, DMAP, CH_2Cl_2, 0 °C, 30 min, 92%; b) HF (48% aqueous solution), MeCN, 25 °C, 30 min, then MeOH, 25 °C, 3 h, 74%; c) Ag_2O, CH_2Cl_2, 25 °C, 30 min, 94%; d) toluene, 110 °C, 1.5 h, 21%; e) TESOTf, Et_3N, CH_2Cl_2, 0 °C, 30 min, 95%; f) H_2, $Pd(OH)_2$ (10% on carbon), EtOAc, 25 °C, 4 h, 92%; g) PIFA, PMBOH, K_2CO_3, MeCN, 0 °C, 30 min, 75%; h) DMP, CH_2Cl_2, 25 °C, 1 h, 90%; i) HF (48% aqueous solution), MeCN, 25 °C, 2 h, 85%; j) $Me_4NBH(OAc)_3$, MeCN/AcOH (10:1), 25 °C, 2 h, 85%; k) DDQ, CH_2Cl_2/H_2O (10:1), 25 °C, 5 h, 70%; l) DBU, CH_2Cl_2/MeOH (3:1), 40 °C, 4 h, 78%; m) *t*BuOOH, DBU, CH_2Cl_2, 40 °C, 3 h, 63%.

3.5. Towards the Total Synthesis of the Sporolides

3.5.1. Strategy 1 - Synthesis of the 9-Membered Core *via* the Enediyne

3.5.1.1. Retrosynthetic Analysis and Strategy Considerations

Our synthesis of sporolides A and B had been planned in order to follow a biomimetic approach for the formation of the chlorinated cyclopenta[*a*]indene ring. We proposed to pass through an enediyne system and a *Bergmann* cyclization, trapping the resulting diradical by a chloride source. This approach would allow the preparation of both fragments for the synthesis of sporolides A and B. Moreover, this pathway would give the possibility of investigating possible precursors for both

sporolides and potentially discovering new potent anticancer drugs. As already discussed, these compounds did not display interesting activity, this because the isolated sporolides are already the results of the *Bergmann* cycloaromatization. Probably, the reactive enediyne precursor was the biosynthetic main product of the *Salinospora tropica*. This is also supported by the fact that sporolides were isolated without an apoprotein that in most cases stabilize the 9-membered enediyne system. To date there are no reports concerning the existence of a carrier protein stabilizing a potential precursor of sporolides A and B, even though biosynthetic studies support the enediyne pathway for their formation.[147]

Sporolides A (**125**) and B (**126**) can be split into two fragments, **123** and **124**, by disconnecting at the ester and the acetal/vinylogous ester (Scheme 63). This thesis will concentrate on the preparation of fragment **124**, because subunit **123** is part of the PhD work of *Jean-Yves Wach* at the EPFL. As discussed above, fragment **124** will derive from a *Bergmann* cycloaromatization of enediyne **122a** with subsequent trapping of the newly formed diradical by a chloride source. The stability of the 9-membered ring enediyne **122a** will be evaluated during the synthesis. There is the possibility that intermediate **122a** cannot be isolated as it might spontaneously undergo cycloaromatization when the 9-membered ring is formed. We planned to prepare the enediyne by intramolecular acetylide addition to the aldehyde in compound **97**, as reported in literature for similar substrates.[165a] Compound **97** will result from the *Sonogashira* coupling[200] of the enediyne subunit **94**[201] and the vinyl triflate **77**. Fragment **94** will be prepared following similar procedure to that reported by *Myers* and co-workers in the synthesis of the neocarzinostatin chromophore starting from L-(*S*)-glyceraldehyde acetonide (**85**).[165a] The aldehyde will be itself synthesized according to the procedure developed at Hoffmann-La Roche starting from L-ascorbic acid (**82**).[202] Fragment **77** derives from cyclopentenone (**71**) and is characterized by a *Sharpless* asymmetric dihydroxylation.[203] A second approach

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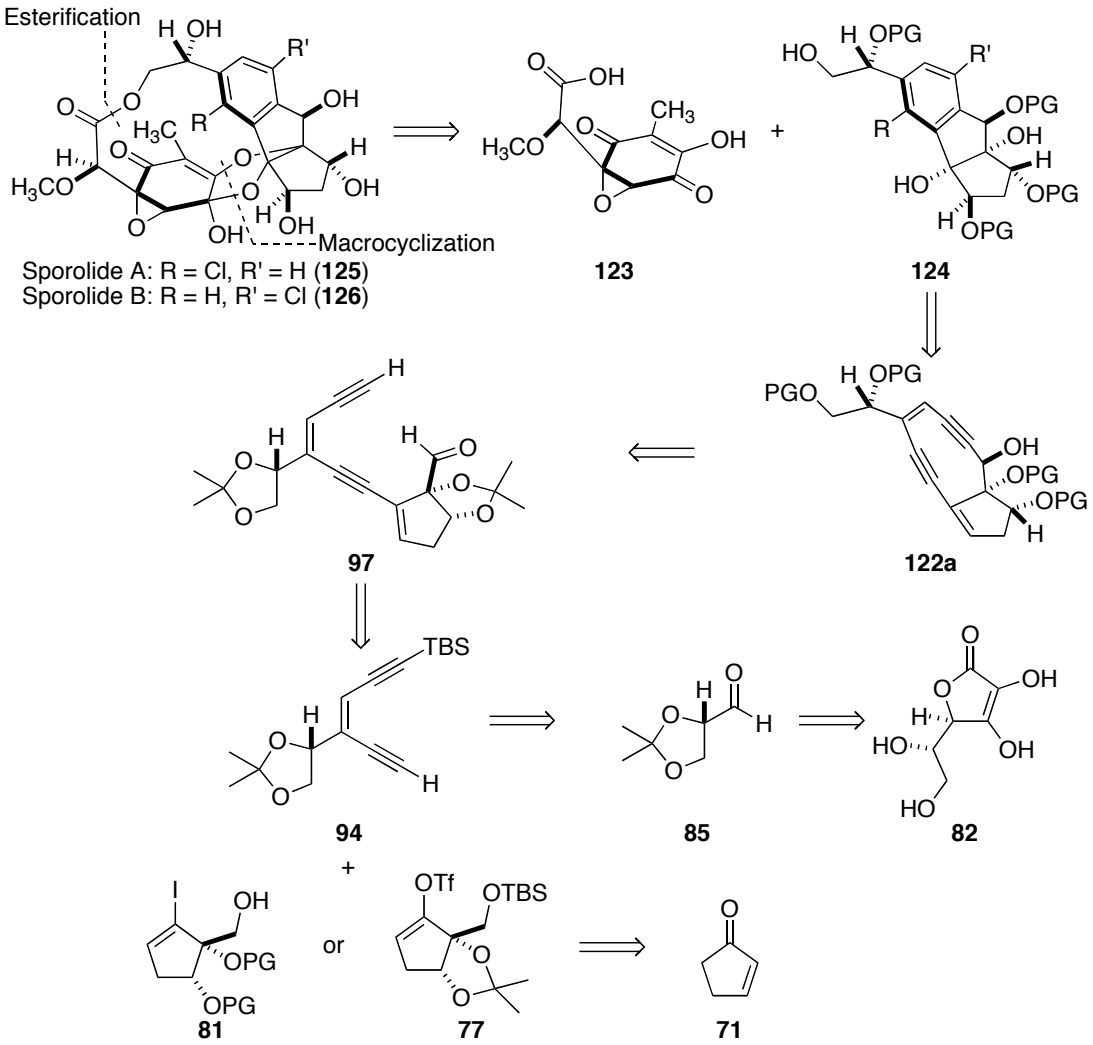
200 K. Sonogashira, Y. Tohda, N. Hagihara, *Tetrahedron Lett.* **1975**, *16*, 4467-4470.

201 Fragment **94** was prepared following the procedure reported by *Myers* and co-workers for the synthesis of the neocarzinostatin chromophore. A. G. Myers, R. Glatthar, M. Hammond, P. M. Harrington, E. Y. Kuo, J. Liang, S. E. Schaus, Y. Wu, J.-N. Xiang, *J. Am. Chem. Soc.* **2002**, *124*, 5380-5401.

202 C. Hubschwerlen, *Synthesis* **1986**, *1986*, 962-964.

203 a) E. N. Jacobsen, I. Marko, W. S. Mungall, G. Schroeder, K. B. Sharpless, *J. Am. Chem. Soc.* **1988**, *110*, 1968-1970; b) K. B. Sharpless, W. Amberg, Y. L. Bennani, G. A. Crispino, J. Hartung, K. S. Jeong, H. L. Kwong, K. Morikawa, Z. M. Wang, *J. Org. Chem.* **1992**, *57*, 2768-2771; c) H. C. Kolb, M. S. VanNieuwenhze, K. B. Sharpless, *Chem. Rev.* **1994**, *94*, 2483-2547.

towards enediyne subunit **97** was also envisaged starting from enediyne subunit **94** and compound **81**, which was obtained from cyclopentenone **71** *via* enantioselective kinetic resolution using Pig Liver Esterase (PLE).[204]

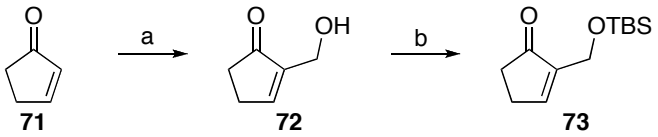


Scheme 63: Retrosynthetic analysis of sporolides A (**125**) and B (**126**).

[204] a) C. Tanyeli, E. Turkut, I. Mecidoglu Akhmedov, *Tetrahedron: Asymmetry* **2004**, *15*, 1729-1733; b) F. D. Özdemirhan, M. Celik, S. AtlI, C. Tanyeli, *Tetrahedron: Asymmetry* **2006**, *17*, 287-291.

3.5.1.2. The Vinyl Triflate and the Vinyl Iodide Fragments

The synthesis of sporolides started from commercially available cyclopentenone (**71**).[205] *Morita-Baylis-Hillman* reaction[206] using aqueous formaldehyde in a mixture methanol/chloroform and in the presence of $PPhMe_2$ as catalyst gave rapid access to the alcohol **72** in excellent yield (97%) (Scheme 64). The reaction time had not to exceed one hour or degradation of the product was observed. Moreover, isolation of the product proved to be challenging because removal of the solvent resulted in degradation of the product. To overcome the problem, after one hour stirring the reaction mixture was dried by addition of $MgSO_4$ and directly loaded on to a column of silica to afford alcohol **72**. The product was subsequently protected with a TBS group to afford enone **73** in 99% yield.



Scheme 64: a) HCHO (37% in H_2O), $PPhMe_2$ (6 mol %), $CHCl_3$, MeOH, RT, 1 h, 97%; b) TBSCl, imidazole, CH_2Cl_2, RT, overnight, 99%.

Sharpless asymmetric dihydroxylation88 of compound **73** proved extremely difficult due to the electron deficient alkene and several sets of conditions were screened. From the literature it is known that electron poor olefins are less reactive than electron neutral or rich olefins towards osmium tetroxide and usually the problem is overcome by increasing the catalyst concentration.[203c,207] Moreover, $MeSO_2NH_2$ (1-2 equiv) can be added to the reaction in order to accelerate the hydrolysis of the Os(IV) glycolate intermediate in the catalytic cycle.[203b] This faster hydrolysis is usually required for hindered olefins or for reaction at low temperatures.[208] First attempts to afford the dihydroxylated product **74** were carried out using standard AD-mix-β enriched with $K_2OsO_2(OH)_2$ (2 mol %) in several solvents. The reactions did not proceed smoothly and even when the starting material

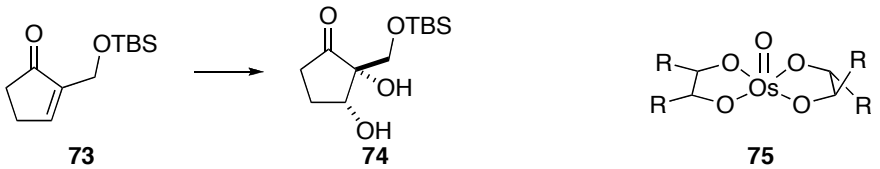
[205] The synthesis of the compounds **71-76** was performed by Massimo Binaghi in the context of the diploma work under my supervision at the EPFL.

[206] a) K. Morita, Z. Suzuki, H. Hirose, *Bull. Chem. Soc. Jpn.* **1968**, *41*, 2815; b) A. B. Baylis, M. E. D. Hillman, Offenlegungsschrift 2155113, US Patent 3,743,669, **1972** [Chem. Abstr. 1972, 1977, 34174q]; c) H. Ito, Y. Takenaka, S. Fukunishi, K. Iguchi, *Synthesis* **2005**, *2005*, 3035-3038.

[207] Y. L. Bennani, K. B. Sharpless, *Tetrahedron Lett.* **1993**, *34*, 2079-2082.

[208] T. Göbel, K. B. Sharpless, *Angew. Chem., Int. Ed.* **1993**, *32*, 1329-1331.

was completely consumed, the product **74** was isolated only in poor yields (20-28%) (Table 5, entries 1-4). It was suspected that the diol product **74** was unstable toward the basic conditions and therefore, it was decided to change the reaction conditions and use *N*-methylmorpholine *N*-oxide (NMO) as oxidant. The reaction was performed using $K_2OsO_2(OH)_2$ (2 mol %), $(DHQD)_2$-PHAL (3 mol %), NMO and $MeSO_2NH_2$ (1 equiv) in a monophasic mixture water/acetone (1:3) at 0 °C. After 6 hours the product **74** was obtained in 89% yield, but the optical rotation ($[\alpha]^{23.5}_D$ = –21.8° (*c* 0.78, $CHCl_3$)) (Table 5, entry 5) was slightly lower than the previously obtained (Table 5, entry 3). Faster conversion was obtained adopting the same conditions at RT to afford diol **74** in 95% yield in 45 minutes (Table 5, entry 6). In literature is reported that using NMO as cooxidant a second catalytic cycle affording bis-glycolate complex **75** is acting.[203c] The formation of a bis-glycolate complex **75** would result in a decrease of the enantiomeric excess, but this can avoid by slow addition of the olefin. We performed the reaction using $K_2OsO_2(OH)_2$ (1.5 mol %), $(DHQD)_2$-PHAL (3 mol %), NMO and $MeSO_2NH_2$ (1.5 equiv) in a monophasic mixture water/acetone (1:3) at RT and adding a solution of the olefin at a rate of 0.06 mmol/min (over 2.5 hours). After addition the reaction was stirred for additional 30 minutes and the product **74** was isolated in 92% yield (Scheme 65). An increase in the optical rotation was observed ($[\alpha]^{22.9}_D$ = –28.9° (*c* 0.99, $CHCl_3$)) (Table 5, entry 7).[209] Attempts to assess the enantioselectivity *via* chiral stationary phases on HPLC revealed to be unsuccessful and also by injecting a racemic mixture of **74**, only a single peak was observed. We decided to continue the synthesis without measuring the enantiomeric excess and check if diastereoisomers would appear after the coupling with the enediyne subunit.



73 **74** **75**

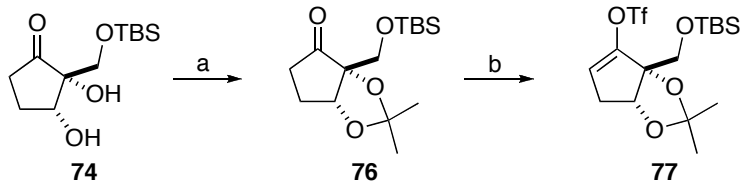
Scheme 65: a) $K_2OsO_2(OH)_2$ (1.5 mol %), $(DHQD)_2$-PHAL (3.0 mol %), NMO, $MeSO_2NH_2$, water/acetone (1:3), then **73** (0.06 mmol/min), RT, 3 h, 92%.

[209] C. R. Johnson, M. R. Barbachyn, *J. Am. Chem. Soc.* **1984**, *106*, 2459-2461. Literature value: $[\alpha]^{25.0}_D$ = –34.7° (*c* 1.00, $CHCl_3$).

Table 5: Screened conditions for the preparation of the dihydroxylated compound (**74**).

Entry	Conditions	Reaction time	Yield	Optical rotation	Comments
1	AD-mix-β, $K_2OsO_2(OH)_4$ (2 mol %), $MeSO_2NH_2$, H_2O/*t*BuOH (1:1), 0 °C → RT	48 h	20%	n.d.	Side product formed and no starting material recovered
2	Entry 1 + $NaHCO_3$ buffering	48 h	20%	n.d.	Side product formed and no starting material recovered
3	AD-mix-β, $K_2OsO_2(OH)_4$ (2 mol %), $MeSO_2NH_2$, H_2O/AcOEt (1:1), RT	40 h	24%	–24.5°	Starting material recovered
4	AD-mix-β, $K_2OsO_2(OH)_4$ (2 mol %), $MeSO_2NH_2$, H_2O/ *t*BuOH/AcOEt (8:9:9), RT	18 h	28%	n.d.	Side product formed and no starting material recovered
5	$K_2OsO_2(OH)_4$ (2 mol %), $(DHQD)_2$-PHAL (3 mol %) NMO, $MeSO_2NH_2$, H_2O/ acetone (1:3), 0 °C	6 h	89%	–21.8°	–
6	$K_2OsO_2(OH)_4$ (2 mol %), $(DHQD)_2$-PHAL (3 mol %) NMO, $MeSO_2NH_2$, H_2O/ acetone (1:3), RT	45 min	95%	n.d.	–
7	$K_2OsO_2(OH)_4$ (1.5 mol %), $(DHQD)_2$-PHAL (3 mol %) NMO, $MeSO_2NH_2$, H_2O/ acetone (1:3), RT	2.5 h	92%	–28.9°	Slow addition of the olefin (0.06 mmol/min)

Dihydroxylated compound **74** was protected using 2,2-dimethoxypropane under acidic catalysis in a mixture DMF/acetone to afford the acetonide **76** in 83% yield. The product was then treated with lithium diisopropyl amide (LDA) and the enolate trapped with $N(PhTf_2)$ to give the target vinyl triflate **77** in 85% yield (Scheme 66).



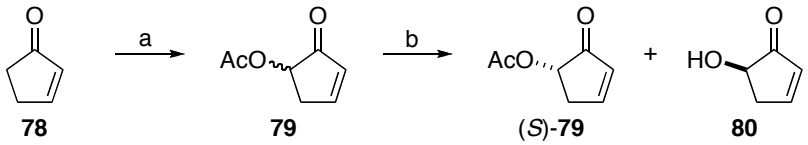
74 **76** **77**

Scheme 66: a) 2,2-dimethoxypropane, PPTS, DMF/acetone (3:1), 0 °C → RT, 84%; b) LDA, **76**, THF, –10 °C, 20 min, then $PhNTf_2$, –10 °C → RT, 15 h, 85%.

In addition to the vinyl triflate **77**, the synthesis of the analogous vinyl iodide **81** was investigated (Scheme 67). As reported in literature,[210] treatment of freshly distilled cyclopentenone with $Mn(OAc)_3$ resulted in the formation of the α-

[210] a) C. Tanyeli, E. Turkut, I. Mecidoglu Akhmedov, *Tetrahedron: Asymmetry* **2004**, *15*, 1729-1733; b) F. D. Özdemirhan, M. Celik, S. Atli, C. Tanyeli, *Tetrahedron: Asymmetry* **2006**, *17*, 287-291.

acetoxylated cyclic ketone **79**. In contrast to that reported in the literature, the reaction gave a poor yield (16%) with considerable formation of unwanted by products. Despite the poor yield product **79** was submitted to an enantioselective kinetic resolution using Pig Liver Esterase (PLE). Unfortunately, we could not reproduce the reported results and only partial resolution was achieved. Therefore, it was decided to abandon this way and concentrate on the vinyl triflate compound **77**.



Scheme 67: a) $Mn(OAc)_3$, benzene, 16%; b) PLE, buffer phosphate (pH 7), 20 °C, 16 h.

3.5.1.3. The Enediyne Fragment

The synthesis of the enediyne fragment started from commercially available L-ascorbic acid (**82**) (Scheme 68).[211] Hydrogenation of the starting material **82** using a thick-membrane 2L-ballon filled with hydrogen and 10% Pd/C as catalyst in water[212] at 65 °C afforded L-gulono-1,4-lactone (**83**) in 77% yield after 72 hours. Selective protection of one of the diols was performed using 2-methoxypropane in DMF to give 5,6-*O*-isopropylidene-L-gulono-1,4-lactone (**84**) in 99% yield. Treatment of **84** with sodium periodate afforded L-(*S*)-glyceraldehyde acetonide (**85**), which as reported in literature cannot be completely extracted from the aqueous phase resulting in the loss of up to 30% of the product.[213] Due to its reduced stability the product **85** was maintained as a solution in THF and directly used in the next step. Thus, ethynyltrimethylsilane was deprotonated at –78 °C using LHMDS and added to a solution of L-(*S*)-glyceraldehyde acetonide (**85**) in THF to afford a diastereomeric mixture (1.3:1) of alcohols **86** in 28% yield. The poor yield was probably due to the extraction and stability problems of the starting material **85**. The diastereoisomeric mixture of alcohol **86** was oxidized to the ketone **87** using PDC and after work-up the instable product **87** was kept in solution and directly used in the next step.

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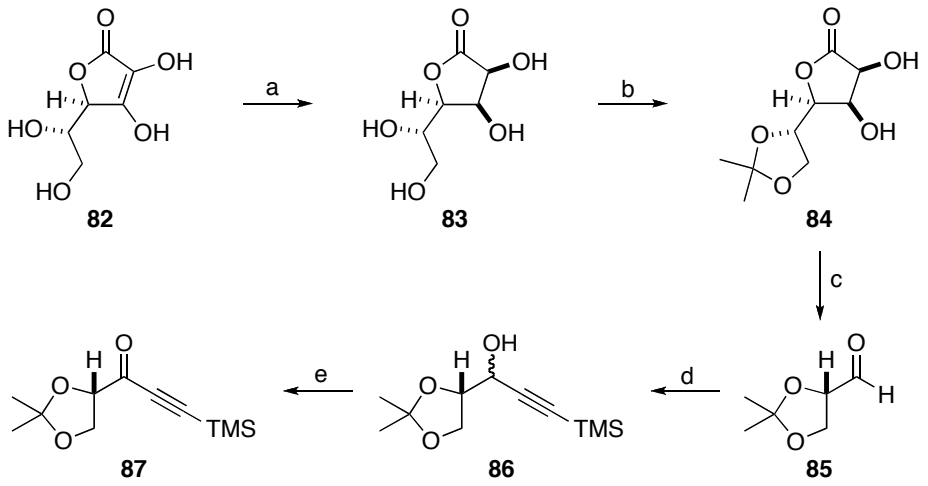
[211] The synthesis of the compounds **82-94** was performed by Cindy Fellay in the context of the diploma work under my supervision at the EPFL.

[212] Before addition of Pd/C and H_2 the solution was degassed bubbling Ar for 30 minutes.

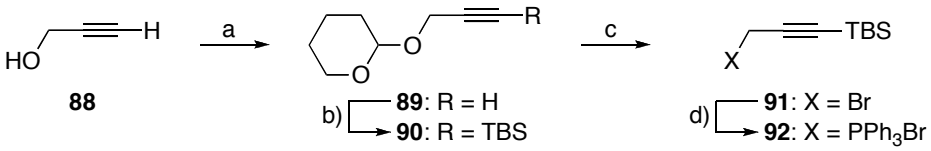
[213] C. Hubschwerlen, J. L. Specklin, J. Higelin, *Org. Synth.* **1995**, *72*, 1-5.



82 **83** **84** **85** **86** **87**

Scheme 68: a) H_2, Pd/C 10%, H_2O, 60 °C, 72h, 77%; b) PTSA•H_2O, 2-methoxypropene, DMF, RT, 2.5 days, 99%; c) $NaIO_4$, NaOH (3 M), H_2O, < 7 °C, pH 4-6, then **84**, 25 °C, 1.5 h; d) ethynyltrimethylsilane, LHMDS, THF, –78 °C, 30 min, then **85**, THF, –78 °C, 1 h, 28% (2 steps); e) 3 Å MS, PDC, AcOH, CH_2Cl_2, RT, 1 h.

The propargylic phosphonium salt **92** for the *Wittig* reaction was prepared starting from commercially available propargyl alcohol (**88**). Protection of the starting material with dihydropyran (DHP) under acidic conditions afforded the protected compound **89** in 98% yield.[214] Subsequent protection of the terminal alkyne with a TBS protecting group gave compound **90** in 99% yield. Direct transformation of **90** to the propargylic bromide compound **91** was achieved using a mixture of triphenylphosphine and bromine at –15 °C. The product **91** was directly used without further purification and treated with triphenylphosphine in toluene to afford the phosphonium salt **92** in 77% yield over two steps (Scheme 69).



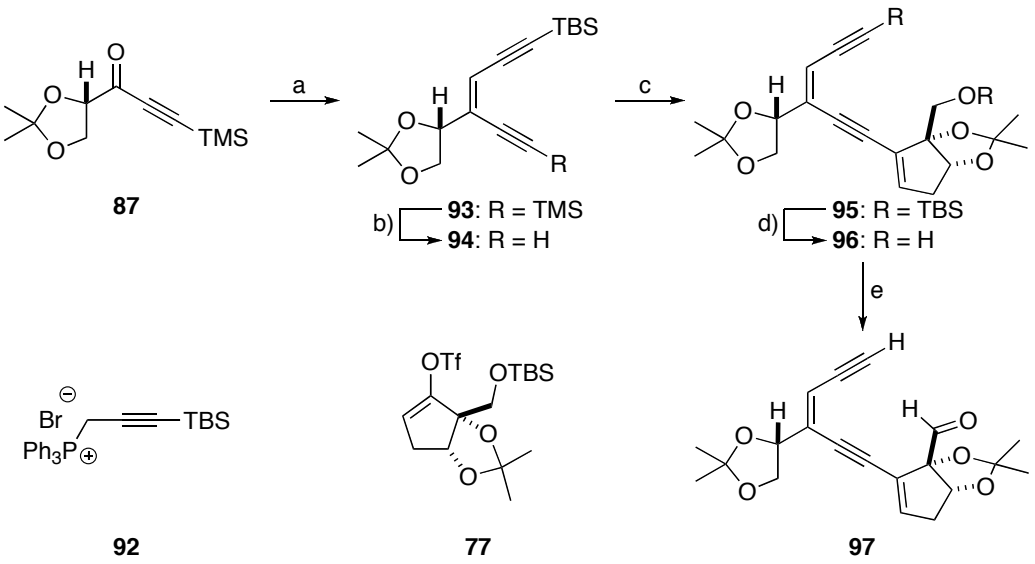
88

Scheme 69: a) PTSA•H_2O, DHP, 65 °C, 3h, 98%; b) *n*BuLi (1.6 M in hexane), TBSCl, THF, –18 °C, 45 min, 99%; c) PPh_3, Br_2, CH_2Cl_2, –15 °C, 30 min, then **90**, RT, 8 h; d) PPh_3, toluene, RT, 42 h, 77% (2 steps).

The phosphonium salt **92** was dissolved in THF, cooled to –78 °C and treated with KHMDS to generate the corresponding ylide. Addition of ketone **87** afforded the enediyne **93** as a 3:1 mixture of *E*/*Z* products (Scheme 70). Purification by chromatography on SiO_2 allowed isolation of the *E* product in 39% yield, together

[214] R. A. Earl, L. B. Townsend, *Org. Synth.* **1981**, *60*, 81-87.

with other fractions containing a mixture of the two isomers. Treatment of the TMS-protected alkyne with K_2CO_3 in MeOH at 0 °C gave the deprotected alkyne **94** in 97% yield. The terminal alkyne **94** was coupled with the vinyl triflate **77** *via Sonogashira* coupling in the presence of CuI and catalyzed by $Pd(PPh_3)_4$ to afford compound **95** in 65% yield.[200] Subsequent deprotection of both TBS protecting groups using TBAF gave alcohol **96**, which was oxidized to the aldeyde **97** *via Swern* oxidation.[215]



Scheme 70: a) **92**, KHMDS (0.5 M in toluene), THF, –78 °C → –40 °C, then **87**, –15 °C, 1.5 h, 34%, *d.r.* = 2.7:1; b) K_2CO_3, MeOH, 0 °C, 45 min, 97%; c) DIPEA, 2,6-lutidine, CuI (30 mol %), $Pd(PPh_3)_4$ (5 mol %), DMF, RT, 1 h 15 min, 65%; d) TBAF (1.0 M in THF), THF, –20 °C → 0 °C, 1 h 45 min, 86%; e) oxalyl chloride, DMSO, CH_2Cl_2, –78 °C, 20 min, then **96**, 30 min, then DIPEA, –78 °C → 0 °C, 50 min, 86%.

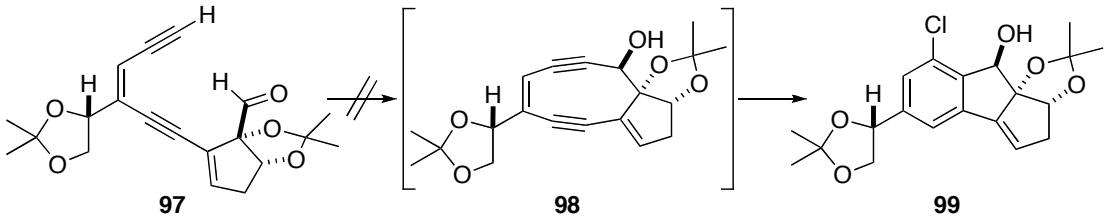
3.5.1.4. The Nine-Membered Ring Enediyne Formation - The Dead End

With aldehyde **97** in hand we tried the cyclization for the formation of the 9-memberd ring. It was decided to adopt the conditions already used by *Myers* and coworkers in the synthesis of the NCS chromophore (**CLXXIX**, Scheme 55).[166b] Aldehyde **97** was added to a mixture of $CeCl_3$ and LiCl in THF, followed by addition of lithium bis(dimethylphenylsilyl)azide[216] at –78 °C (Scheme 71). At this temperature no formation of new products was observed and the reaction was allowed to warm to 0 °C. The reaction was monitored by TLC at intervals between –78 °C and

[215] K. Omura, D. Swern, *Tetrahedron* **1978**, *34*, 1651-1660.

[216] S. Masamune, J. W. Ellingboe, W. Choy, *J. Am. Chem. Soc.* **1982**, *104*, 5526-5528.

0 °C but no formation of product was observed. All attempts to form the 9-membered ring enediyne or directly the cycloaromatized compound failed and under more forcing reaction conditions only formation of uncharacterized side products was observed. These results are in agreement with those reported by *Hirama* and co-workers during their study on 9-membered ring enediyne.[217] An explanation could be in the rigidity of the enediyne structure, in which the two sp^2 hybridized centers block the flexibility of the molecule and as a consequence the attack of the acetylide. It is possible that the formation of the 9-membered ring requires more energy and could take place at higher temperatures, but in our case side reactions take place first. At this point we encountered a dead end and we had to revise our synthetic approach to the chlorinated cyclopenta[*a*]indene ring.



Scheme 71: Failed attempts to form the 9-membered ring **98** or directly the cycloaromatized product **99** using $CeCl_3$, LiCl, lithium bis(dimethylphenylsilyl)azide, THF, –78 °C → 0 °C.

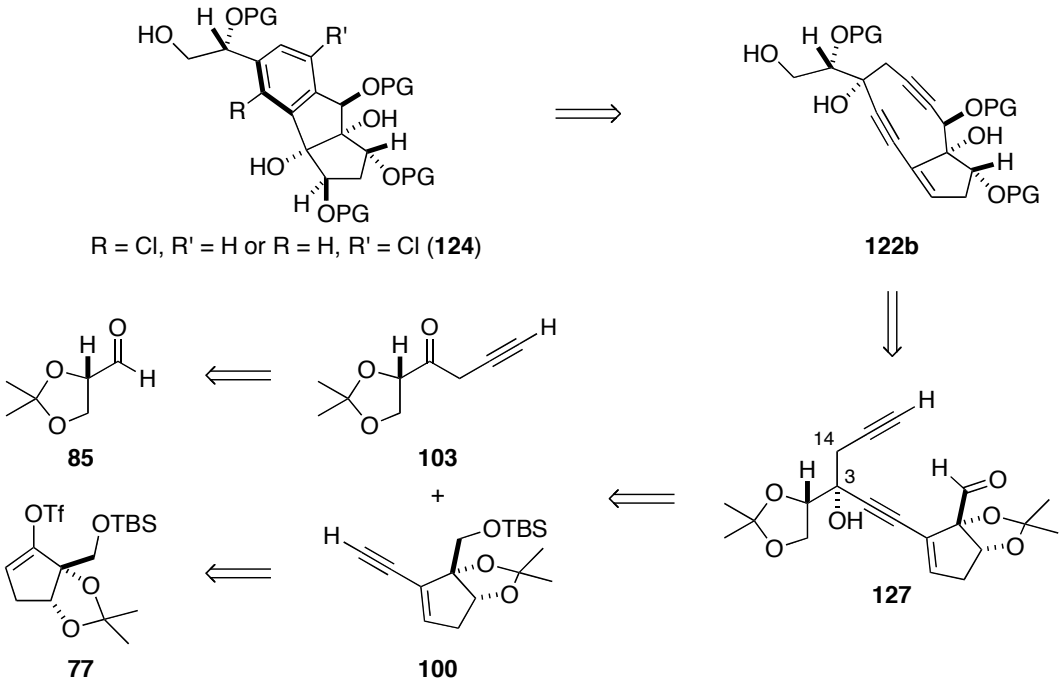
3.5.2. Strategy 2 - Synthesis of the Nine-Membered Core *via* a Diyne

3.5.2.1. Revision of the Retrosynthetic Analysis

Due to the problems encountered in the formation of the 9-membered enediyne core we proposed an approach allowing the formation of a 9-membered ring diyne core without the two sp^2 centers of the enediyne at C(3) and C(14) (Scheme 72). We planned to obtain the chlorinated cyclopenta[*a*]indene ring **124** from the 9-membered diyne ring core **122b**. The absence of the two sp^2 centers would give the molecule more flexibility and hopefully a better possibility of forming the 9-membered core. Moreover, the presence of the hydroxy group at C(3) would not only increase the stability of the core by preventing the cycloaromatization, but also act as a switch for the *Bergmann* cyclization upon its elimination. As in the previous approach, the cyclic core would be formed by intramolecular addition of the acetylide to the

[217] T. Mita, S. Kawata, M. Hirama, *Chem. Lett.* **1998**, *27*, 959-960.

aldehyde in compound **127**. Compound **127** will derive from the addition of alkyne **100** to ketone **103**. The alkyne fragment can be prepared by a *Sonogashira* coupling between commercially available trimethylsilylacetylene and the vinyl triflate **77** used in the first approach. The ketone **103** will be prepared by addition of propargyl magnesium bromide to the L-(*S*)-glyceraldehyde acetonide **85** followed by an oxidation. With this new approach we will also avoid problems related to the *Wittig* reaction for the formation of the enediyne system that as previously discussed gave a 3:1 mixture of *E*/*Z* isomers.

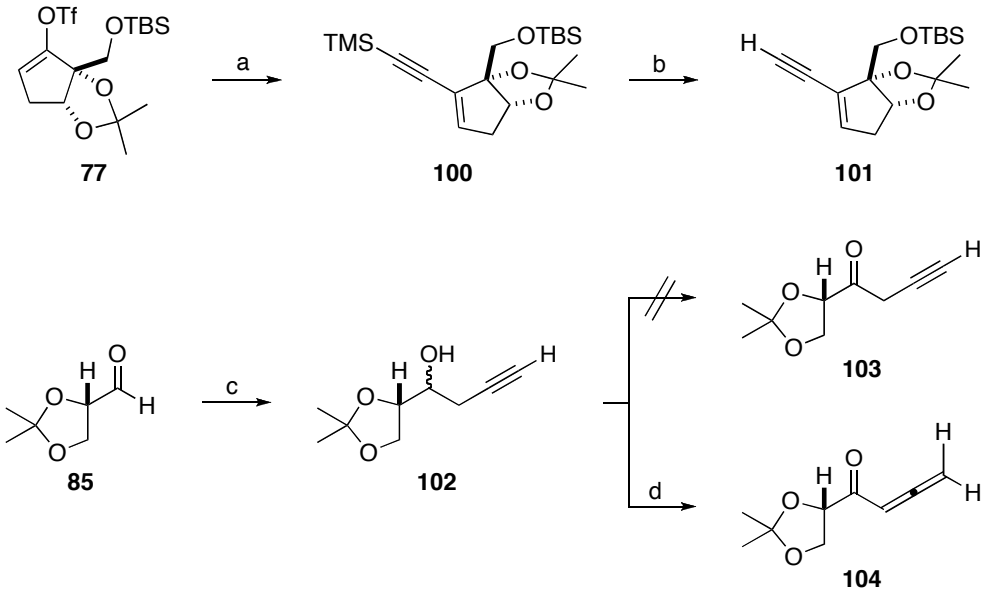


Scheme 72: Revised retrosynthetic analysis of sporolides A and B based on the diyne intermediate **127**.

3.5.2.2. Toward the 9-Membered Diyne Core

Vinyl triflate **77** was reacted with commercially available trimethylsilylacetylene *via* a palladium catalyzed *Sonogashira* coupling to afford adduct **100** in quantitative yield (Scheme 73). The product was then deprotected using K_2CO_3 in MeOH to give the target terminal alkyne **101**. The preparation of the ketone **103** was started with the preparation of a solution of L-(*S*)-glyceraldehyde acetonide **85** in THF using the same method adopted previously (Scheme 68). Separately, propargyl magnesium bromide was prepared by dropwise addition of a solution of propargyl bromide to a mixture of

Mg(turning) and $HgCl_2$ (cat.) in Et_2O. The cooled *Grignard* reagent was subsequently added to the precooled (–20 °C) solution of L-(*S*)-glyceraldehyde acetonide **85** and the resulting solution allowed to return to RT. The alcohol adduct **102** was obtained in 43% yield over two steps as a mixture 1.0:0.6 of diastereoisomers. Problems were encountered when we tried to oxidize the diastereoisomeric mixture to the ketone **103**. All the conditions employed, comprising PDC and PCC, TPAP/NMO[218] and *Oppenauer* oxidation[219] failed to generate the desired product **103** and only starting material was recovered. We then tried *Dess-Martin* periodinane oxidation and with relief the formation of a new product on TLC was observed. The reaction proceeded smoothly, but surprisingly the product was not the expected alkyne **103**, but the corresponding allene **104**. Allenes are known to undergo isomerization to alkynes when treated with strong bases, as in the *Zipper* reaction.[220] With this in mind the synthesis was continued with allene **104**.



Scheme 73: a) trimethylsilylacetylene, DIPEA, 2,6-lutidine, CuI (30 mol %), $Pd(PPh_3)_4$ (5 mol %), DMF, RT, 1.5 h, quant.; b) K_2CO_3, MeOH, 0 °C, 3.5 h, quant.; c) Mg(turning), $HgCl_2$ (cat.), I_2 (cat.), propargyl bromide (80% in toluene), Et_2O, reflux, 1 h, then **85**, –20 °C → RT, 2 h, 43%, *d.r.* = 1.00:0.06 (2 steps); d) DMP, CH_2Cl_2, RT, 4 h, 87%.

With both fragments in hand we investigated the alkyne **101** addition to the keto allene **104** (Scheme 74). The terminal alkyne **101** was deprotonated with *n*BuLi and

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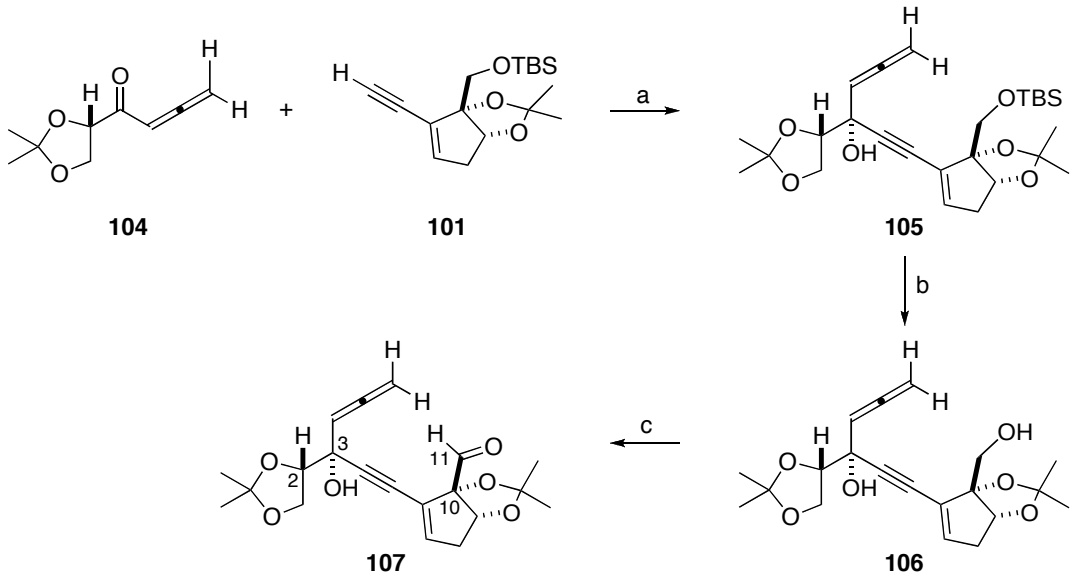
[218] W. P. Griffith, S. V. Ley, G. P. Whitcombe, A. D. White, *J. Chem. Soc., Chem. Commun.* **1987**, 1625-1627.

[219] R. V. Oppenauer, *Recl. Trav. Chim. Pays-Bas* **1937**, *56*, 137-144.

[220] a) C. A. Brown, A. Yamashita, *J. Am. Chem. Soc.* **1975**, *97*, 891-892; b) C. A. Brown, A. Yamashita, *J. Chem. Soc., Chem. Commun.* **1976**, 959-960; c) S. R. Macaulay, *J. Org. Chem.* **1980**, *45*, 734-735.

transferred into a cooled (–78 °C) solution of keto allene **104** in THF. The reaction was allowed to warm up slowly and followed at regular intervals by TLC, but no formation of the desired adduct **105** was observed even at RT. After work-up only the starting terminal alkyne **101** was recovered, while the keto allene **104** had degraded. We tried to run the reaction using the same conditions, but this time adding HMPA in order to avoid possible aggregates interfering with the reaction, but this was also unsuccessful. At this point we decided to activate the acceptor using a *Lewis* acid and opted for a solution of $CeCl_3$•2LiCl[221] in THF. As in the previous protocol, the acetylide, formed by treatment of the terminal alkyne **101** with *n*BuLi, was transferred into a cooled (–78 °C) solution of keto allene **104** activated by $CeCl_3$•2LiCl. The resulting mixture was heated to –40 °C and allowed to return to 0 °C over two hours. After optimization of the reaction conditions the alcohol **105** was obtained in 75% yield in a diastereoisomeric ratio of 94:6. Based on literature precedent on similar systems,[222] we expected to obtain diastereoisomer **105** with the stereochemistry as reported in scheme 74. As reported by *Hirama* and co-workers, the configuration of centers C(2), C(3) and C(10) influence the outcome of the configuration at C(11) in the 9-membered ring forming step.[222] Based on this precedent, the configuration at C(3) shown in compound **107** would lead exclusively to the desired diastereoisomer. It was decided not to spend any more time confirming the configuration, as at this point we were more interested in exploring whether the formation of the 9-membered ring would be possible. Moreover, the hydroxy group at C(11) would be removed later in the synthesis to allow cycloaromatization. The synthesis continued by removal of the TBS protecting group to afford the deprotected product **106** in quantitative yield. The primary alcohol was subsequently oxidized using DMP furnishing the corresponding aldehyde **107** in 88% yield.

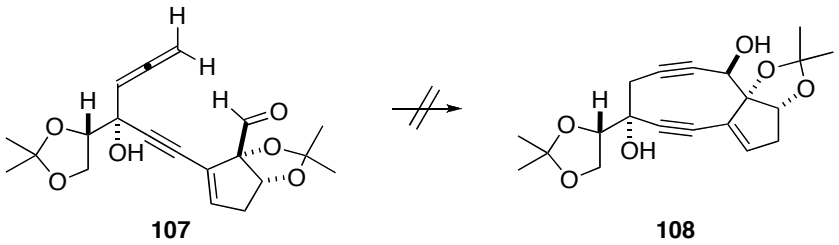
[221] a) A. Krasovskiy, F. Kopp, P. Knochel, *Angew. Chem., Int. Ed.* **2006**, *45*, 497-500; b) B. M. Trost, J. Waser, A. Meyer, *J. Am. Chem. Soc.* **2007**, *129*, 14556-14557.
[222] I. Sato, Y. Akahori, K. I. Iida, M. Hirama, *Tetrahedron Lett.* **1996**, *37*, 5135-5138.



Scheme 74: a) **101**, *n*BuLi (1.6 M in hexane), –78 °C, 30 min, then **104**, $CeCl_3$•2LiCl (0.2 M in THF), –40 °C → 0 °C, 2 h, 75%, *d.r.* = 94:6; b) TBAF (1.0 M in THF), THF, 0 °C → RT, 3.5 h, quant.; c) DMP, CH_2Cl_2, 88%.

3.5.2.3. The Nine-Membered Ring from the Allene - The Dead End

Before trying the isomerization of the allene using standard methods,[220a,b] we wanted to investigate if it was possible to form the 9-membered ring directly from the allene **107** (Scheme 75). Strong bases are known to induce the isomerization of allenes to terminal alkynes.[223] Moreover, following isomerization, the acetylide would be generated *in situ* and add directly to the aldehyde to afford the 9-membered ring **108**. We screened several reaction conditions trying different *Lewis* acids, bases, reaction temperatures and times (Table 6). All reactions failed without providing the desired product **108**. In general the starting material **107** was stable until ca. –30 °C when treated with $LiN(Me_2Ph)_2$ or LHMDS, but more forcing reaction conditions only caused degradation of the starting material **107**.



Scheme 75: Failed 9-membered ring **108** formation from compound **107**.

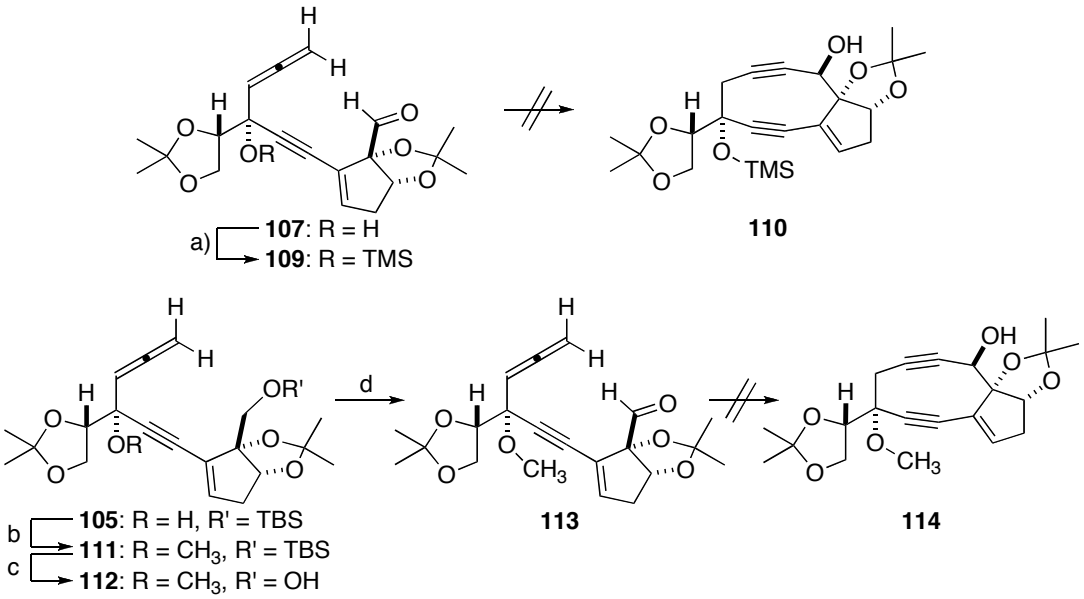
[223] a) D. R. Taylor, *Chem. Rev.* **1967**, *67*, 317-359; b) H. W. Thompson, *J. Org. Chem.* **1967**, *32*, 3712-3713; c) L. Crombie, M. A. Horsham, R. J. Blade, *Tetrahedron Lett.* **1987**, *28*, 4879-4882.

Table 6: Screened condition for the 9-membered ring **108** formation from aldehyde **107**.

Lewis acid (equiv)	Base (equiv)	Reaction temperature	Reaction time
$CeCl_3$•2LiCl (3)	$LiN(Me_2Ph)_2$ (5)	–78 °C → –40 °C	3 h
$CeCl_3$•2LiCl (4)	$LiN(Me_2Ph)_2$ (6)	–78 °C → –10 °C	3 h
$CeCl_3$•2LiCl (8)	$LiN(Me_2Ph)_2$ (12)	–78 °C → RT	3 h 30 min
$CeCl_3$•2LiCl (1)	$LiN(Me_2Ph)_2$ (5)	–45 °C → –20 °C	4 h
$CeCl_3$•2LiCl (10)	$LiN(Me_2Ph)_2$ (9)	–78 °C → –15 °C	17 h
$CeCl_3$•2LiCl (3)	KHMDS (3)	–78 °C → –30 °C	3 h 10 min
$CeCl_3$•2LiCl (3)	*n*BuLi (5)	–78 °C → 0 °C	3 h 20 min
$CeCl_3$ (3)	$LiN(Me_2Ph)_2$ (5)	–78 °C → –40 °C	3 h
$CeCl_3$ (10)	$LiN(Me_2Ph)_2$ (9)	–45 °C → –20 °C	1 h 30 min
$CeCl_3$ (3)	LHMDS (15)	–78 °C → –15 °C	2 h 15 min
$CeCl_3$ (20)	LHMDS (15)	–40 °C → –15 °C	22 h
LiCl (50)	LHMDS (15)	–78 °C → –15 °C	2 h 15 min
$Yb(OTf)_3$ (3)	$LiN(Me_2Ph)_2$ (6)	–45 °C → –10 °C	4 h 15 min

We decided to repeat the reaction, but this time with the hydroxy group at C(3) protected in order to avoid the formation of a the negative charge on the alkoxy group that could interfere with the reaction. We protected the hydroxy group in two different ways; in compound **109** with a TMS group and in product **111** with a methyl group (Scheme 76). This approach was risky because protecting the hydroxy group at C(3) makes its elimination easier with respect to the unprotected hydroxy group, which was stabilized by *in situ* formation of the alkoxy group. The TMS protected product **109** was prepared in 67% yield by treatment of compound **107** with TMSOTf at –78 °C. Compound **113** was synthesized from the tertiary alcohol **105** in three steps. Treatment of **105** with *Meerwein* salt[224] in presence of 4 Å MS and proton sponges furnished the methylated product **111** in 83% yield. Subsequent removal of the TBS protecting group using TBAF gave the primary alcohol **112** in excellent yield. Final *Swern* oxidation furnished the aldehyde **113**, which was directly used in the next step without further purification. As for compound **107**, we screened different conditions using compounds **109** and **113** (Scheme 76 and Table 7). Also in this case the results were unsuccessful and forcing reaction conditions caused only degradation of the starting material.

[224] a) H. Meerwein, G. Hinz, P. Hofmann, E. Kroning, E. Pfeil, *J. Prakt. Chem.* **1937**, *147*, 257-285; b) H. Meerwein, E. Battenberg, H. Gold, E. Pfeil, G. Willfang, *J. Prakt. Chem.* **1939**, *154*, 83-156.

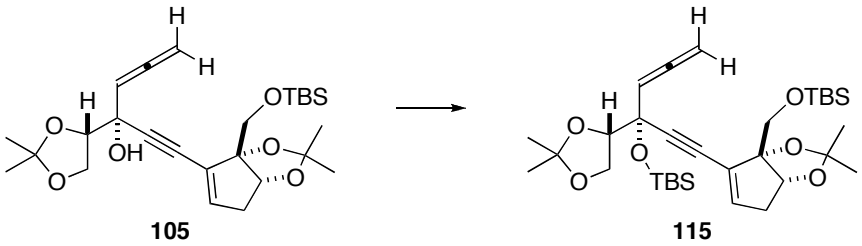


Scheme 76: a) TMSOTf, 2,6-lutidine, CH_2Cl_2, –78 °C, 1.5 h, 67%; b) Me_3OBF_4, 4Å MS, proton sponge, CH_2Cl_2, 0 °C → RT, 20 h, 83%; c) TBAF (1.0 M in THF), THF, 0 °C → RT, 45 min, 98%; d) oxalyl chloride, DMSO, CH_2Cl_2, –78 °C, 20 min, then **112**, 30 min, then DIPEA, –78 °C → 0 °C, 2 h 10 min.

Table 7: Screened condition for the 9-membered ring formation from aldehydes **109** and **113**.

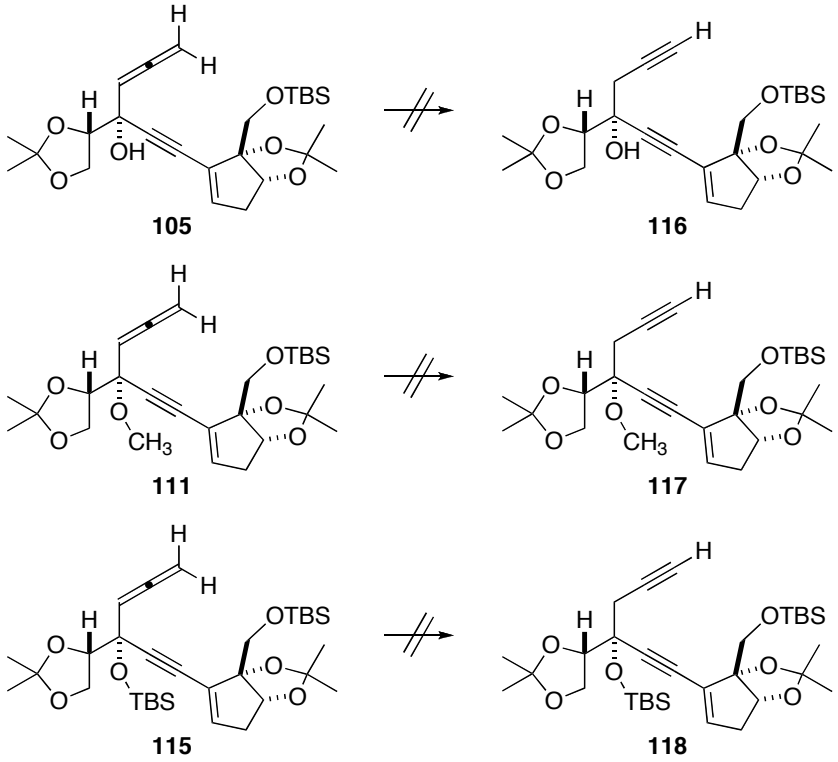
Compound	Lewis acid (equiv)	Base (equiv)	Reaction temperature	Reaction time
109	$CeCl_3$•2LiCl (4)	$LiN(Me_2Ph)_2$ (9)	–78 °C → RT	2 h
109	$CeCl_3$•2LiCl (3)	LHMDS (8)	–78 °C → RT	3 h
109	$CeCl_3$•2LiCl (3)	LDA (8)	–78 °C → RT	3 h
109	$CeCl_3$ (3)	$LiN(Me_2Ph)_2$ (9)	–78 °C → RT	2 h
109	$CeCl_3$ (3)	KHMDS (4)	–78 °C → 0 °C	3 h
109	LiCl (50)	$LiN(Me_2Ph)_2$ (9)	–78 °C → RT	2 h
113	$CeCl_3$•2LiCl (13)	$LiN(Me_2Ph)_2$ (9)	–40 °C → –20 °C	3 h
113	$CeCl_3$•2LiCl (3)	LHMDS (9)	–40 °C → –25 °C	42 h
113	$CeCl_3$ (10)	$LiN(Me_2Ph)_2$ (24)	–40 °C → –25 °C	39 h

After the unsuccessful attempts of *in situ* isomerization of the allene to the terminal alkyne, we tried to do it by standard method. Potassium 3-aminopropylamide (KAPA) is the most commonly used base to induce the zip reaction, which forms the terminal alkyne very fast at 0 °C.[220a,b] We tried the isomerization on three different substrates; compounds **105** and **111** which have already encountered and product **115** which was prepared by TBS protection of tertiary alcohol **105** (Scheme 77).



Scheme 77: TMSOTf, 2,6-lutidine, CH_2Cl_2, –40 °C → RT, 4 h.

Compounds **105**, **111** and **115** were treated at 0 °C or RT at different concentration of KAPA[225] and reaction times (Scheme 78 and Table 8). None of the reactions furnished the desired products (**116**-**118**) and usually only degradation of the starting material was observed, except for entry 1 and 2 in which all the starting material was recovered. All attempts to form the 9-membered ring from the allene failed and once again it was achieved a dead-end where a new revision of the strategy was required.



Scheme 78: Failed attemps of allene-terminal alkyne isomerization for compounds **105, 111** and **115**.

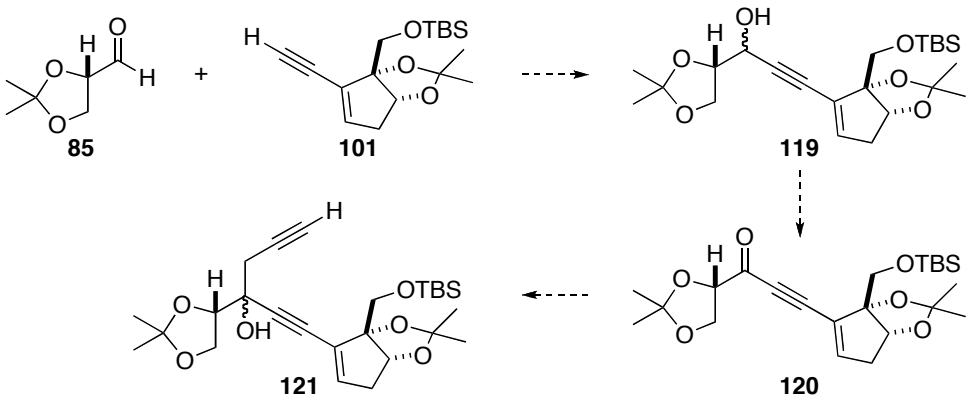
[225] Preparation of KAPA (0.25 M in 1,3-diaminopropane (DPA)): DPA (2.1 mL) was added to KH (0.52 mmol) at RT. The resulting mixture was stirred until complete consuming of KH to afford a clear-brown solution.

Table 8: Screened condition using KAPA for the isomerization of compounds **105, 111** and **115**.

Entry	Compound	KAPA (equiv)	Reaction conditions (T, time)
1	**105**	15	0 °C, 45 min, then RT, 15 min
2	**111**	1.3	RT, 15 min
3	**111**	3.2	0 °C, 2 h
4	**111**	7.4	0 °C, 3.5 h, then RT, 14 h
5	**115**	5	0 °C, 2 h
6	**115**	5	0 °C, 4 h, then RT, 17 h

3.5.3. Comments and Perspectives

In our investigation on sporolides two synthetically approaches were unsuccessfully evaluated. The first time we expected to obtain the 9-membered enediyne ring or directly the cycloaromatized product starting from the enediyne core **97**, but the cyclization failed. The second time we initially planned to obtain the 9-membered ring passing through a diyne system, but problems during the oxidation of alcohol **102** were encountered leading to the formation of the keto-allene **104** and not the desired alkyne **103**. It was decided to continue the synthesis with the allene, as it was hoped that isomerization to the terminal alkyne would be possible later in the synthesis. However, this attempt also led to a dead-end. The isomerization required reaction conditions that were not tolerated by our substrates and degradation of the starting material occurred first. Although several conditions for the isomerization were tried, further investigation of this reaction is required. Moreover, there are also other possible routes to test, for example, reverse the steps for the alkyne **101** addition on L-(*S*)-glyceraldehyde acetonide **85**. If the terminal alkyne **101** were added first, the obtained mixture of alcohols **119** could be oxidized avoiding problems of allene formation. Subsequent reaction of ketone **120** with a propargyl *Grignard* reagent would generate the desired product **121** (Scheme **79**).



Scheme 79: Proposed route for the continuation of the synthesis.

3.6. Conclusion

This chapter was dedicated to the synthetic studies on sporolides A and B and allowed us to take a journey into the world of the enediyne natural products. Due to the phenomenal biological activity of the enediynes, these compounds were initially considered as powerful antitumor agents sparking great interest in chemistry, biology and medicine. Today, the enediyne antitumor antibiotics are lead candidates in the battle against cancer and several member of this family are in clinical phase trials or even used as therapeutic treatments. The research on these compounds continues and more potent and more selective compounds are in development. This drove us to start our investigation on sporolides A and B, two macrolides supposed to derive from the cycloaromatization of an enediyne precursor. The complex architectural structure of the sporolides, displaying 22 of 24 carbons sp^2 hybridized or oxygenated, 7 rings and 10 stereogenic centers makes these really challenging targets for total synthesis. When we started this project, no total syntheses of sporolides had been reported. We planned our synthesis in such a way as to follow a biomimetic pathway allowing the possible biological evaluation of the sporolide enediyne precursor. Unfortunately, for the two pathways were unsuccessfully investigated. Among the interesting reactions encountered in this synthetic study we found the *Morita-Baylis-Hillman* reaction, *Sharpless* asymmetric dihydroxylation, enediyne formation *via Wittig* reaction and $CeCl_3$•2LiCl-mediated acetylide addition. I think that for this project not all is lost and the synthesis can be continued using the previously prepared intermediates. As previously discussed, there are pathways still open and maybe they will prove to be a good choice to achieve the target.

4. Nostocarboline and Eudistomin N Derivatives as Potential Antimalarial Agents

4.1. Introduction

Malaria remains a huge problem in developing countries with 40% of the worldwide population living in high-risk infection areas.[226] This disease affects 300-500 million people and causes over 1 million deaths each year, especially among infants and children.[227] The protozoal parasites of the genus *Plasmodium* are the origin of this disease and mosquitoes of the genus *Anopheles* are their vectors. Humans can be infected by four species of *Plasmodium*: *Plasmodium malariae*, *P. Ovale*, *P. vivax* and *P. falciparum*. The latter is the most dangerous species affording to the highest number of deaths. Controlling and preventing the spread of malaria by targeting the vector would prove difficult. Several antimalarial drugs are currently on the market including quinine and its derivatives, *e.g.* chloroquine.[226] More recently, combination therapies also proved to be useful in the battle against this disease. However, even though therapies are available, malaria infections continue to increase and resistance of malaria parasites to the currently used drugs is becoming more and more common, making the situation alarming.[226] Moreover, no vaccine is available, making parasite chemotherapy the only way to fight this disease.

4.2. Nostocarboline and Malaria

In 2005, *Gademann* and co-workers isolated an acetyl- and butyryl-cholinesterase and trypsin inhibitor nostocarboline (**CCXVI**) from the freshwater cyanobacterium *Nostoc* 78-12A (Figure 33).[228] In addition, this new quaternary *β*-carboline alkaloid **CCXVI** displayed potent algicides activity inhibiting the growth of phytoplanktal organisms.[229] The malaria parasite *Plasmodium falciparum* contains an organelle of

[226] J. Wiesner, R. Ortmann, H. Jomaa, M. Schlitzer, *Angew. Chem., Int. Ed.* **2003**, *42*, 5274-5293.
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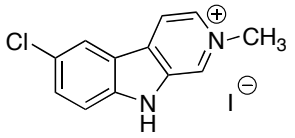
[228] P. G. Becher, J. Beuchat, K. Gademann, F. Jüttner, *J. Nat. Prod.* **2005**, *68*, 1793-1795.
[229] J. F. Blom, T. Brütsch, D. Barbaras, Y. Bethuel, H. H. Locher, C. Hubschwerlen, K. Gademann, *Org. Lett.* **2006**, *8*, 737-740.

cyanobacterial origin, the apicoplast,[230] which has been suggested to be a target for antiplasmodial agents.[231]



Nostocarboline (**CCXVI**)

Figure 33: Nostocarboline (**CCXVI**).

Several natural products from cyanobacteria have been shown to possess antiplasmodial activity. Calothrixins A and B inhibited *Plasmodium falciparum* with an IC_{50} value of 60 nM and 180 nM respectively, but without selectivity for HeLa human cancer cell lines.[232] Activity at 8.2 μM against the same tropical parasite was observed for venturamide B with a lower cytotoxicity (86 μM) to green monkey Vero kidney cells.[233] Symplocamide A displayed IC_{50} value of 0.95 μM against *P. falciparum*, but also a strong cytotoxicity.[234] In 2008, *Gademann* and co-workers isolated aerucyclamides A-D from the cyanobacterium *Microcystis aeruginosa* PCC 7806.[235] Aerucyclamide B displayed submicromolar IC_{50} value against chloroquine-resistant strain K1 of *P. falciparum* with large selectivity (IC_{50} = 120 μM) with respect to the L6 rat myoblast cell line. Low micromolar activities were measured for aerucyclamides C and D with almost no toxicity to the L6 cell line. Studies on *β*-carbolinium cation derivatives have also been reported and results have shown that these compounds exhibit strong activity against malaria.[236] Moreover, the presence of the positive charge on these π-delocalized lipophilic cationic (DLC) structures results

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231 a) S. A. Ralph, M. C. D'Ombrain, G. I. McFadden, *Drug Resistance Updates* **2001**, *4*, 145-151; b) S. Sato, R. J. M. Wilson, *Curr. Top. Microbiol. Immunol.* **2005**, *295*, 251-273.

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233 R. G. Linington, J. González, L. D. Urena, L. I. Romero, E. Ortega-Barría, W. H. Gerwick, *J. Nat. Prod.* **2007**, *70*, 397-401.

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235 a) C. Portmann, J. F. Blom, K. Gademann, F. Jüttner, *J. Nat. Prod.* **2008**, *71*, 1193-1196; b) C. Portmann, J. F. Blom, M. Kaiser, R. Brun, F. Jüttner, K. Gademann, *J. Nat. Prod.* **2008**, *71*, 1891-1896.

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in an increase of the activity compared to their corresponding neutral carbolines.[237] Several natural *β*-carbolinium cations displaying activity against malaria are known (Scheme 34): normelinoline F (**CCXVII**) displayed an IC_{50} value of 13.6 μM and no cytotoxicity.[238] Fascaplysin[239] (**CCXVIII**), cryptolepine[240] (**CCXIX**) and isoneocryptolepine[241] (**CCXX**) display activity at the submicromolar scale against resistant K1 strain of *P. falciparum* (IC_{50} = 0.184 μM, 0.12 μM and 0.23 μM respectively) but with a reduced selectivity against rat myoblast L6 (IC_{50} = 9.2 μM, 1.12 μM and 4.23 μM respectively).

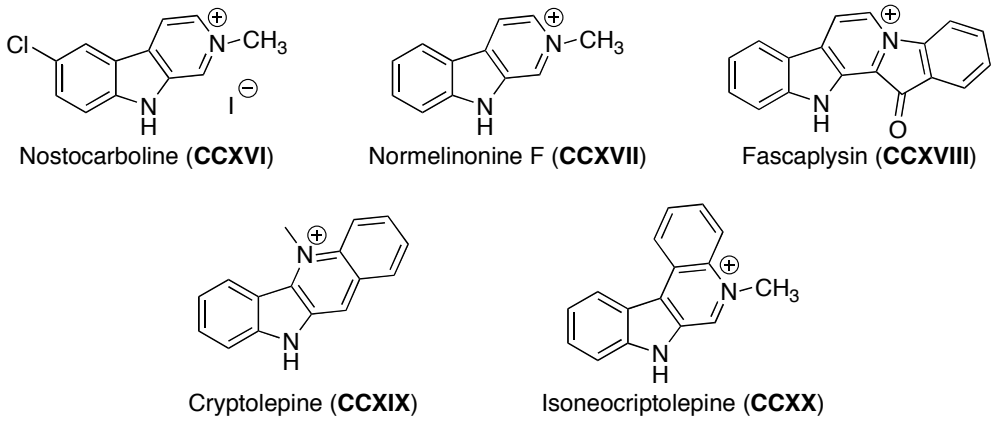

Figure 34: natural *β*-carbolinium cation: nostocarboline (**CCXVI**), normelinoline F (**CCXVII**), fascaplysin (**CCXVIII**), cryptolepine (**CCXIX**) and isoneocryptolepine (**CCXX**).

Nostocarboline (**CCXVI**) was prepared by *Gademann* and co-workers in a straightforward way from norharmane (**CCXXI**)[229] (Figure 35) and then tested against *Plasmodium falciparum*. The compound **CCXVI** displayed strong antiplasmodial activity with an IC_{50} value of 194 nM, but also a large selectivity being more than 600

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[239] a) D. M. Roll, C. M. Ireland, H. S. M. Lu, J. Clardy, *J. Org. Chem.* **1988**, *53*, 3276-3278; b) G. Kirsch, G. M. Köng, A. D. Wright, R. Kaminsky, *J. Nat. Prod.* **2000**, *63*, 825-829.

[240] a) C. W. Wright, *J. Pharm. Pharmacol.* **2007**, *59*, 899-904; b) L. Dassonneville, K. Bonjean, M. C. De Pauw-Gillet, P. Colson, C. Houssier, J. Quetin-Leclercq, L. Angenot, C. Bailly, *Biochemistry* **1999**, *38*, 7719-7726; c) C. W. Wright, J. Addae-Kyereme, A. G. Breen, J. E. Brown, M. F. Cox, S. L. Croft, Y. Gokcek, H. Kendrick, R. M. Phillips, P. L. Pollet, *J. Med. Chem.* **2001**, *44*, 3187-3194; d) S. Van Miert, T. Jonckers, K. Cimanga, L. Maes, B. Maes, G. Lemière, R. Dommisse, A. Vlietinck, L. Pieters, *Experimental Parasitology* **2004**, *108*, 163-168; e) J. Lavrado, A. Paulo, J. Gut, P. J. Rosenthal, R. Moreira, *Bioorg. Med. Chem. Lett.* **2008**, *18*, 1378-1381.

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times less toxic against L6 rat myoblast cell line.[242] Dimers of nostocarboline were prepared in one additional step (Figure 35) and tested against malaria, displaying activity that reached 18 nM with a large selectivity >2600-fold against the L6 cell line.[242] Four dimers **CCXXII-CCXXV** and nostocarboline (**CCXVI**) were selected for *in vivo* evaluation in a *P. berghei* mouse model. All the dimers displayed low activity and did not influence the survival time of mice, while nostocarboline (**CCXVI**) displayed almost a 50% reduction in parasitaemia and increased the survival time at a dose of 50 mg/kg.[243] After these results a search for more active and selective compound based on quaternary β-carboline alkaloids was required and it was decided to prepare derivatives of nostocarboline (**CCXVI**) and eudistomin N[244] (**CCXXVI**) (Figure 35) for biological evaluation against malaria.

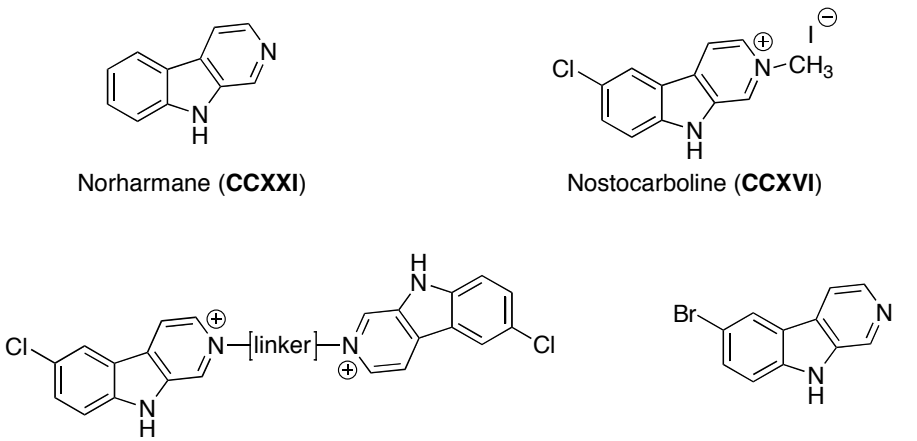

Figure 35: norharmane (**CCXXI**), nostocarboline (**CCXVI**), dimer derivatives (**CCXXII-CCXXV**), eudistomin N (**CCXXVI**).

242 D. Barbaras, M. Kaiser, R. Brun, K. Gademann, *Bioorg. Med. Chem. Lett.* **2008**, *18*, 4413-4415.

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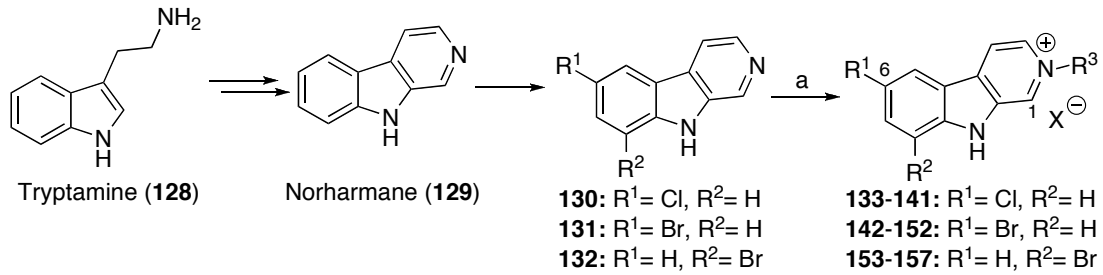
4.3. Preparation of Nostocarboline and Eudistomin N Derivatives

In this project it was decided to synthesize quaternary *β*-carboline alkaloids and to evaluate these compounds against the malaria parasite *P. falciparum*. Derivatives of nostocarboline as well as *N*-alkylated eudistomin N analogs were chosen as targets. The precursors, 6-chloro norharmane (**130**), 6-bromo norharmane (**131**) and 8-bromo norharmane (**132**), were prepared by chlorination and bromination of norharmane (**129**) respectively, which itself was readily accessible from tryptamine in accordance to a literature procedure.[229,245] The three precursors **130**-**132** were then alkylated with a series of electrophiles to directly afford the desired derivatives **133**-**157** with different residues on the pyridine nitrogen. A general procedure was adopted for their preparation. A mixture of starting material **130** or **131** or **132** and the selected electrophile in CH_3CN or *i*PrOH was stirred at 85 °C in a sealed tube for between 1 to 22 hours. The reaction was concentrated and triturated or precipitated in CH_3CN or in a mixture of CH_3CN/Et_2O. The precipitate was then dissolved in MeOH and all insoluble residues removed by filtration. The filtrate was finally concentrated and dried under high vacuum to afford the desired derivatives **133**-**157** as crystalline compounds in yields between 23% to quantitative (Scheme 80 and Table 9). The derivatives **133**-**157** were submitted to the Swiss Tropical Institute to biological evaluation against malaria.



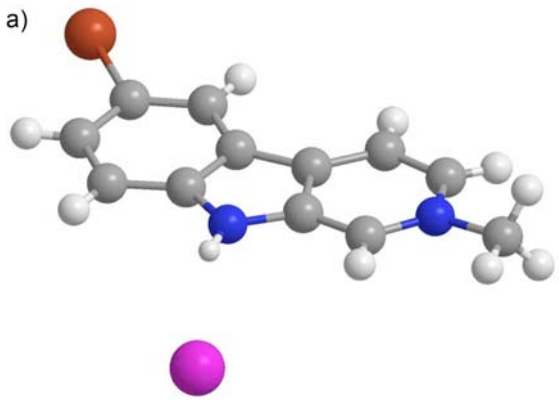
Scheme 80: a) **130** or **131** or **132**, R^3X, CH_3CN or *i*PrOH, 85 °C, 1-21 h, 23-100%. For R^3 see Table 9.

245 a) B. T. Ho, K. E. Walker, *Org. Synth.* **1971**, *51*, 136-138; b) C. Portmann, C. Prestinari, T. Myers, J. Scharte, K. Gademann, *ChemBioChem* **2009**, *10*, 889-895.

Table 9: Derivatives prepared by alkylation of compounds **130**, **131**, **132**.

Compound	Time [h]	R^1	R^2	R^3	X	Yield [%]
133	4	Cl	H	$-CH_3$	I	62
134	18	Cl	H	$-C_2H_5$	I	43
135	21	Cl	H	allyl	Br	45
136	overnight	Cl	H	–*n*Bu	I	37
137	overnight	Cl	H	$-(CH_2)_3COOCH_3$	Br	24
138	15	Cl	H	benzyl	Br	58
139	overnight	Cl	H	*p*-fluoro benzyl	Br	96
140	overnight	Cl	H	*p*-nitro benzyl	Br	55
141	overnight	Cl	H	3-phenyl propyl	Br	31
142	18	Br	H	$-CH_3$	I	71
143	15	Br	H	$-C_2H_5$	I	83
144	15	Br	H	allyl	Br	66
145	15	Br	H	–*n*Bu	I	43
146	15	Br	H	$-(CH_2)_3COOCH_3$	Br	79
147	15	Br	H	benzyl	Br	quant.
148	1	Br	H	*p*-fluoro benzyl	Br	96
149	22	Br	H	*m*-fluoro benzyl	Br	44
150	5	Br	H	*p*-nitro benzyl	Br	99
151	15	Br	H	3-phenyl propyl	Br	94
152	5	Br	H	2-naphtyl	Br	82
153	overnight	H	Br	$-C_2H_5$	I	23
154	overnight	H	Br	allyl	Br	24
155	overnight	H	Br	benzyl	Br	44
156	overnight	H	Br	*p*-fluoro benzyl	Br	25
157	overnight	H	Br	2-naphtyl	Br	43

Compounds **142** and **144** were recrystallized from MeOH and a mixture Et_2O/hexane respectively and submitted to X-ray analysis (Figure 36).



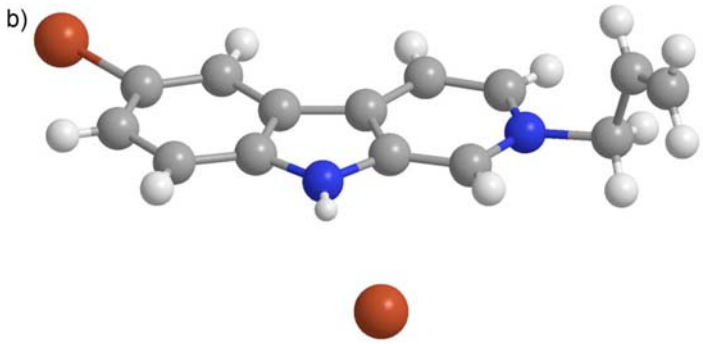
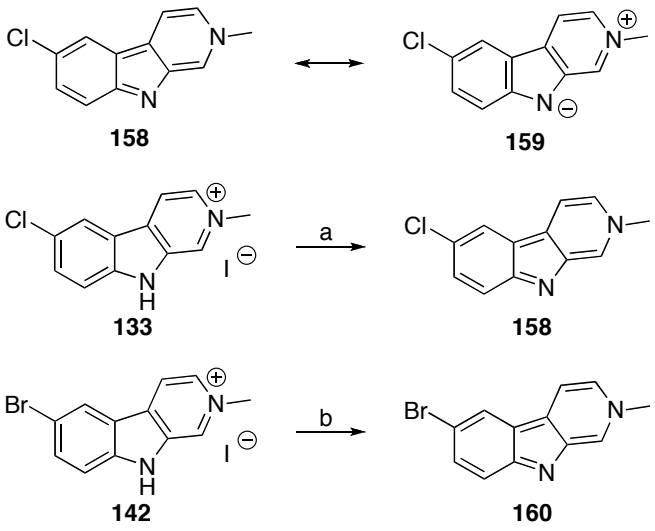


Figure 36: X-ray crystallographic analysis: a) compound **142**; b) compound **144**. Red = Br, fuchsia = I, blue = N, grey = C, white = H.

Nostocarboline (**133**) was isolated as the hydroiodide salt, but under basic conditions, it is present as an anhydronium base represented by two different resonance structures **158** and **159** (Scheme 81). The anhydronium bases **158** and **160** were prepared by treatment of a mixture of nostocarboline (**133**) or 6-bromo nostocarboline (**142**) in EtOAc with a solution of NaOH that immediately generated a bright yellow solution. The products **158** and **160** were carefully isolated avoiding any contact with acidic sources. The two anhydronium bases **158** and **160** could be recrystallized from a mixture MeOH/Et_2O/hexane and were submitted to X-ray analysis (Scheme 81 and Figure 37), confirming their stability. It is thought that in biological medium, the pH-dependent equilibrium between the anhydronium base and the corresponding salt play a crucial role for antimalarial activity.



Scheme 81: a) NaOH (1 M), EtOAc, RT, 10 min, 88%; b) NaOH (3 M), EtOAc, RT, 15 min, quant.

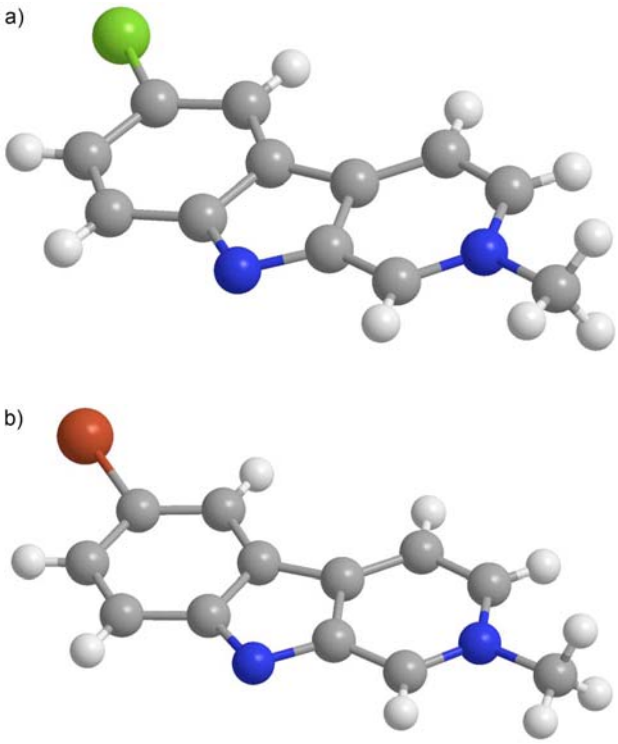


Figure 37: X-ray crystallographic analysis: a) nostocarboline anhydronium base **158**; b) bromo nostocarboline anhydronium base **160.** Red = Br, green = Cl, blue = N, grey = C, white = H.

The effect of pH on nostocarboline (**133**) was investigated by dissolving it in solutions between pH 8 to 12, the emission spectra were observed and a change in color and fluorescence became visible. Nostocarboline (**133**) is brown and when irradiated at 366 nm it emits a blue-green fluorescence, whereas the corresponding anhydronium base **158** is yellow and emits a strong yellow fluorescence. The equivalent point was observed between pH 10 and 11 (Figure 38).

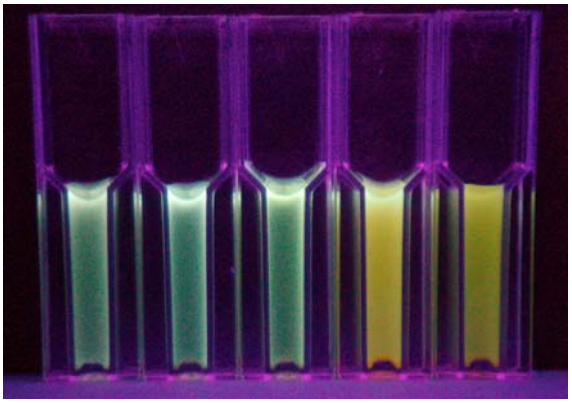


Figure 38: Nostocarboline (**133**) solutions at (from left) pH 8, 9, 10, 11, 12 and irradiated at 366 nm.

The same experiment was carried out with 6-bromo nostocarboline (**142**), which was dissolved in solutions between pH 1 to 14. Both 6-bromo nostocarboline (**142**) and its corresponding anhydronium base **160** are yellow and when irradiated at 366 nm, **142** emits green fluorescence, whereas **160** emits a yellow fluorescence. The equivalent point was observed between pH 10 and 11 (Figure 39). In addition, formation of a gel was observed when compound **142** was stored at pH 1.

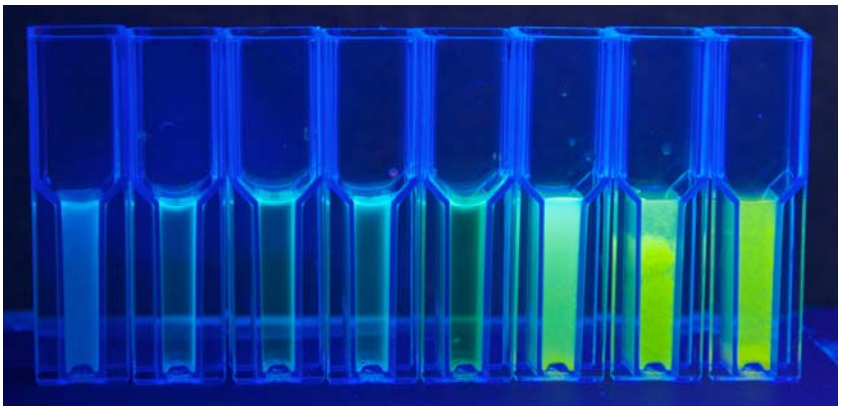


Figure 39: six-bromo nostocarboline (**142**) solutions at (from left) pH 1, 6, 8, 9, 10, 11, 12, 14 and irradiated at 366 nm.

4.4. Biological Evaluation

The prepared nostocarboline and eudistomin derivatives **133-157** (Scheme 80 and Table 9) were submitted for *in vitro* biological evaluation against four parasites: *Leishmania donovani* MHOM-ET-67/L82 axenic amastigotes, *Trypanosoma brucei rhodesiense* STIB 900, *Trypanosoma cruzi* Tulahuen C2C4 and *Plasmodium falciparum* K1 strain.[246] The cytotoxicity against rat myoblast L6 cells and the selectivity index (SI) were also reported. Moreover, for nostocarboline derivatives **133-141** two additional parameters, of which total surface area (S)[247] of the molecule and the calculated logP (clogP)[248] were reported. These parameters allow the investigation of the influence of the residue (R) on the antiplasmodial activity.

[246] Biological evaluation was performed at the Parasite Chemotherapy section of the Swiss Tropical Institute (STI) in collaboration with Prof. Dr. R. Brun and Dr. M. Kaiser.

[247] The total surface area (S) in $Å^2$ was calculated using moloc (http://www.moloc.ch) by Dr. L. Patiny, Institute of Chemistry and Chemical Engineering (ISIC) at the Swiss Federal institute of Technology of Lausanne (EPFL).

[248] clogP was calculated using Osiris Property Explorer developed by Thomas Sander from Actelion (Basel). The partition coefficient logP mesure the distribution of the compound between octanol and water.

Biological evaluation revealed only weak activity of nostocarboline (**133**) and its derivatives **134**-**141** against *Leishmania donovani*, *Trypanosoma brucei rhodesiense* and *Trypanosoma cruzi* (Table 10). The strongest activity was displayed by the 3-phenylpropyl substituted compound **141** with an activity of 6.2 µM against *T. brucei* and roughly three times less activity against *L. donovani* and *T. cruzi*. In contrast, the same compounds **133**-**141** exhibit stronger activity against *Plasmodium falciparum* with activity between single digit micromolar and submicromolar scale (Table 10). The most active compound was nostocarboline (**133**) with an IC_{50} of 194 nM and a low cytotoxicity of 120.9 µM resulting in a SI value of 634. The correlation study between the total surface area of the molecule and the antiplasmodial activity reveals interesting results (Figure 40). Increasing the total area by replacing the residue (R) with a larger group resulted in a loss of the antiplasmodial activity and an augmentation of the cytotoxicity of the compound. The same behaviour was reflected in the selectivity index, which dropped from 634 to 9 when increasing the size of the residue from a methyl to a benzyl group.

Table 10: Antiparasitic *in vitro* activities of the nostocarboline derivatives **133**-**141**. All results are reported as IC_{50} values in µM.

Comp.	R	*L.d.*[a]	*T.b.*[b]	*T.c.*[c]	*P.f.*[d]	Cytotox.[e]	SI[f]	SA[g]	clogP[h]
133	$-CH_3$	34.3	70.5	>87.1	0.194	120.9	634	204.97	2.83
134	$-C_2H_5$	251.0	36.8	100.2	0.452	113.0	250	222.48	3.16
135	allyl	196.1	33.5	57.8	0.831	126.5	152	235.23	3.33
136	–*n*Bu	112.1	11.6	103.8	1.616	74.0	46	254.52	4.09
137	$-(CH_2)_3COOCH_3$	116.6	105.6	114.6	3.143	207.1	66	300.72	3.65
138	benzyl	112.6	6.2	24.6	2.997	27.0	9	276.2	4.38
139	*p*-fluoro benzyl	110.5	18.1	87.7	4.672	111.3	24	285.7	4.44
140	*p*-nitro benzyl	145.6	21.3	32.8	2.209	42.2	19	307.03	4.25
141	3-phenyl propyl	18.6	6.2	20.9	1.608	71.3	44	n.d.	n.d.

[a] *Leishmania donovani* MHOM-ET-67/L82. Standard reference: miltefosine: IC_{50} = 0.54 µM. [b] *Trypanosoma brucei* rhodesiense STIB 900. Standard reference: melarsoprol: IC_{50} = 10 nM. [c] *Trypanosoma cruzi* Tulahuen C2C4. Standard reference: benznidazole: IC_{50} = 1.637 µM. [d] *Plasmodium falciparum* K1. Standard reference: chloroquine: IC_{50} = 181 nM. [e] Cytotoxicity against rat myoblast L6 cells. [f] The selectivity index is calculated by IC_{50}(L6)/IC_{50}(*P.f.*). [g] Total surface area occupied by the molecule [$Å^2$]. [h] cLogP was calculated using Osiris Property Explorer.

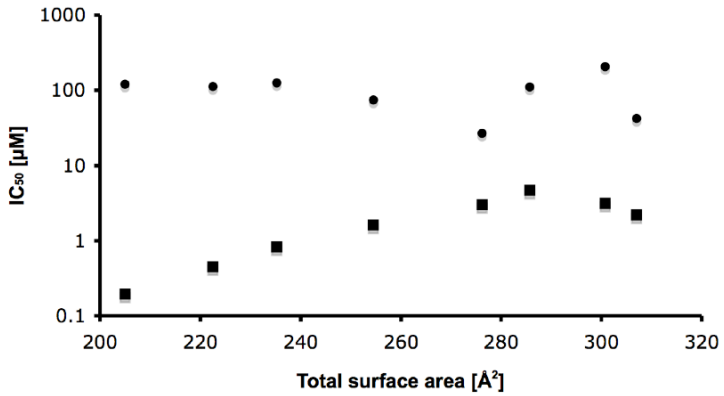


Figure 40: Antiplasmodial activity (■) and cytotoxicity (●) of nostocarboline (**133**) and derivatives **134-141** reported as IC_{50} values plotted against the total surface area. Increasing in the total area results in decreased activity and increased cytotoxicity of the compounds.

For the eudistomin derivatives **142-157** (Scheme 80 and Table 9) biological evaluation also revealed only weak activity against *Leishmania donovani*, *Trypanosoma brucei rhodesiense* and *Trypanosoma cruzi* (Table 11). From the 6-bromo derivatives **142-152**, the strongest activity was displayed by the 2-naphtyl substituted compound **152** with activities of 17.7 µM, 4.1 µM and 7.5 µM against *L. donovani*, *T. brucei* and *T. cruzi* respectively. Similar results were obtained for the 8-bromo derivatives **153-157**, with the 2-naphtyl substituted compound **157** displaying the strongest activities of 46.4 µM, 4.4 µM and 7.9 µM against the same three parasites. In contrast, stronger activities against *Plasmodium falciparum* were observed with IC_{50} values reaching 18 nM and 32 nM for the 6-bromo derivatives **142** and **143** respectively with the methyl and the ethyl groups as substituents. In addition, these two compounds exhibit a low cytotoxicity of 86.1 µM and 78.8 µM, resulting in a very high selectivity index of 4783 and 2443 respectively (Table 11). Moreover, for both the 6-bromo derivatives **142-152** and 8-bromo derivatives **153-157** it was evident that increasing the size of the residue on the pyridine nitrogen caused a loss of activity and an increase in cytotoxicity. This trend directly influenced also the SI value that for the 6-bromo derivatives **142-152** drastically passed from 4783 for the methyl derivative **142** to 11 for the 2-naphtyl derivative **152**. Interesting was also to compare the compound **143** and **153**; both compounds had the ethyl group as *N*-substituent, but differed in the position of the Br on the carboline ring. Compound **143** with the bromine at C(6) displayed an IC_{50} value of 32 nM, while compound **153** with the bromine at C(8) displayed an IC_{50} value of 6.6 µM, resulting in a loss of activity of

more than 200-fold. This difference in activity disappeared when a large substituent was present in the 6- and 8-bromo series.

Table 11: Antiparasitic *in vitro* activities of the eudistomin derivatives **142**-**157**. All results are reported as IC_{50} values in µM.

Comp.	R	*L.d.*[a]	*T.b.*[b]	*T.c.*[c]	*P.f.*[d]	Cytotox.[e]	SI[f]
142	$-CH_3$	>231.3	47.2	131.9	0.018	86.1	4783
143	$-C_2H_5$	>223.2	26.4	90.5	0.032	78.8	2443
144	allyl	20.1	17.4	86.6	0.492	130.9	266
145	–*n*Bu	35.1	11.7	35.3	1.151	68.7	60
146	$-(CH_2)_3COOCH_3$	>203.6	111.0	64.6	2.330	162.3	70
147	benzyl	21.2	5.2	16.3	2.368	61.6	26
148	*p*-fluoro benzyl	16.4	4.7	13.3	1.761	22.6	13
149	*m*-fluoro benzyl	11.0	5.6	25.9	2.153	41.5	19
150	*p*-nitro benzyl	38.2	10.8	14.1	1.330	18.1	14
151	3-phenyl propyl	20.4	4.5	12.6	0.459	27.5	60
152	2-naphtyl	17.7	4.1	7.5	0.481	5.1	11
153	$-C_2H_5$	>223.2	25.0	169.6	6.624	153.4	23
154	allyl	123.1	26.2	153.6	6.738	112.2	17
155	benzyl	66.5	7.0	42.9	2.583	28.7	11
156	*p*-fluoro benzyl	53.4	3.8	28.7	3.279	17.5	5
157	2-naphtyl	46.4	4.4	7.9	0.502	3.7	7

[a] *Leishmania donovani* MHOM-ET-67/L82. Standard reference: miltefosine: IC_{50} = 0.54 µM. [b] *Trypanosoma brucei* rhodesiense STIB 900. Standard reference: melarsoprol: IC_{50} = 10 nM. [c] *Trypanosoma cruzi* Tulahuen C2C4. Standard reference: benznidazole: IC_{50} = 1.637 µM. [d] *Plasmodium falciparum* K1. Standard reference: chloroquine: IC_{50} = 181 nM. [e] Cytotoxicity against rat myoblast L6 cells. [f] The selectivity index is calculated by IC_{50}(L6)/IC_{50}(*P.f.*).

In general high cytotoxicity was observed for large substituents and in particular for phenyl or naphtyl groups, *i.e.* compound **138** (IC_{50} = 27.0 µM) (Table 10), **152** (IC_{50} = 5.1 µM) or **157** (IC_{50} = 3.7 µM) (Table 11). A possible explanation can be attributed to these lipophilic substituents making the molecule a large lipophilic cation that can act as DNA intercalator. This is observed for cryptolepine (**CCXIX**, Figure 34), a potent topoisomerase II inhibitor, of which the strong cytotoxicity was thought to be derived from its DNA intercalation properties.[240b] Compounds **133**, **134** and **142**-**144**, displaying potent and selective antiplasmodial activity *in vitro* were selected for *in vivo* evaluation in a *P. berghei* mouse model and biological assays are currently ongoing.

4.5. Conclusion

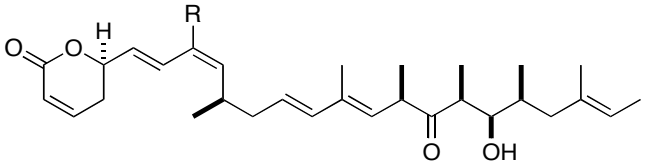
In this chapter, the preparation and biological evaluation of new quaternary *β*-carboline alkaloids derived from nostocarboline and eudistomin N against four parasites has been presented. All the derivatives did not show submicromolar activity against *Leishmania donovani*, *Trypanosoma brucei*, *Trypanosoma cruzi*, but a pronounced activity against *Plasmodium falciparum* was found. Among the derivatives synthesized from 6-chloro norharmane (**130**), the parent natural product nostocarboline (**133**) was demonstrated to be the most active and selective. For this natural product, *in vitro* evaluation gave an IC_{50} value of 194 nM comparable with the activity of currently used chloroquine (IC_{50} = 181 nM) against the *P. falciparum* parasite. In addition, nostocarboline (**133**) displayed also low cytotoxicity against rat myoblast L6 cells, resulting in a selectivity index of 634. Based on nostocarboline derivatives **133**-**141**, a correlation between the activity and the size of the quaternary *β*-carboline was proposed. Increasing of the substituent size on the pyridine nitrogen of the carboline resulted in a decrease of the activity and an increase of the cytotoxicity. The trend was supposed to be derived from the aptitude of larger lipophilic cation to act as DNA intercalators.

Stronger *in vitro* activities were obtained for 6-bromo norharmane derivatives **142**-**152** and in particular for the *N*-methylated compound **142**, which displayed an excellent IC_{50} value against the *P. falciparum* parasite of 18 nM. Moreover this compound displayed a low cytotoxicity against rat myoblast L6 cells (IC_{50} = 86.1 μM), resulting in a selectivity index of 4783. In contrast, a reduced activity was displayed by 8-bromo-norharmane **153**-**157** derivatives. A comparison between compounds **143** and **153**, both with an ethyl group on the pyridine nitrogen, but the Br groups at different position, demonstrated a loss of activity of more than 200-fold of these isomeric structures. The *in vitro* most active compounds **133**, **134** and **142**-**144** are currently in *in vivo* evaluation in a *P. berghei* mouse model, but unfortunately results are not yet available.

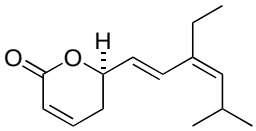
The first *in vitro* results for these derivatives are encouraging, displaying high activity and selectivity against *P. falciparum* K1. Moreover, from a synthetic and economic point of view, nostocarboline and eudistomin derivatives boast a simple, cheap and straightforward preparation making them good candidates as potential antimalarial agents.

5. Conclusion

This thesis was dedicated to the synthesis and the biological evaluation of natural products and their derivatives in order to contribute to the development of new therapeutics against cancer and malaria. In the first chapter the total syntheses of anguinomycins C and D were presented. These syntheses allowed the elucidation of the configuration of both compounds and they were achieved in 29 steps with a longest linear sequence of 18 steps and an overall yield of 6.7% and 6.0% respectively. In addition, derivatives were prepared in order to study the mode of action and the structure-activity relationship.

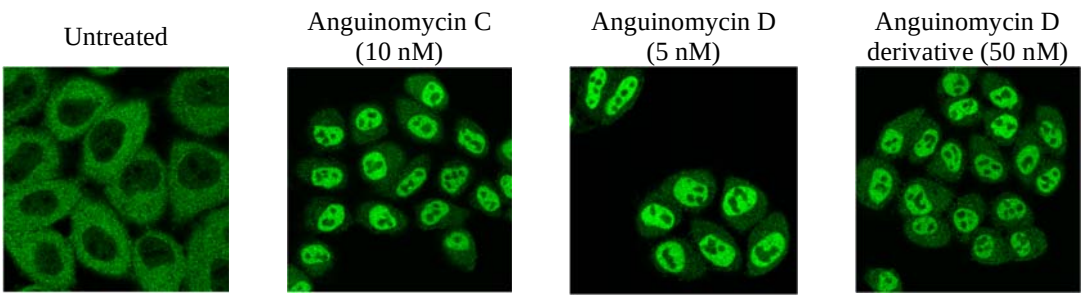


Anguinomycins C (R = CH_3) & D (R = C_2H_5)

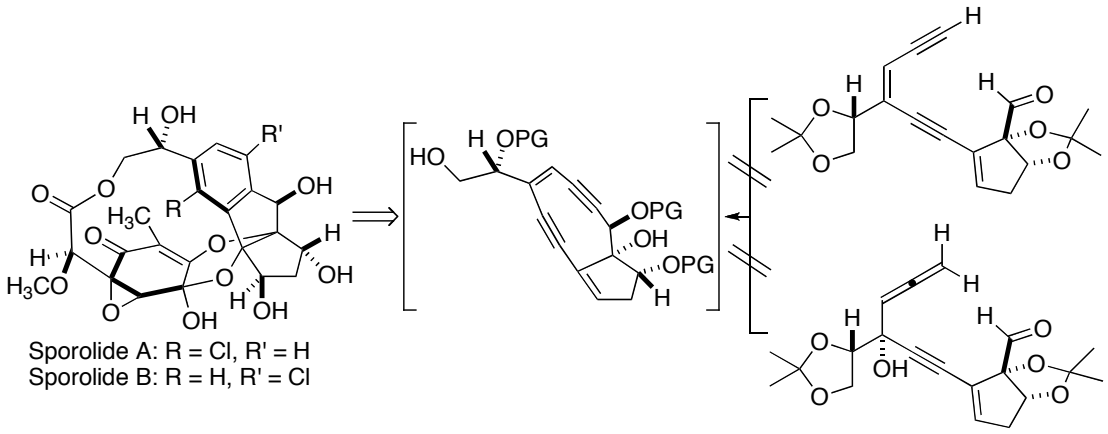


Anguinomycin D derivative

Biological evaluation of the synthesized compounds on their ability to inhibit the CRM1-mediated nucleocytoplasmic transport confirmed the high activity of anguinomycins C (IC_{50} = 10 nM) and D (IC_{50} = 5 nM). The most interesting derivative that was prepared, a shorter version of anguinomycin D, caused accumulation of the Rio2 protein in the nucleus at less than 50 nM. Structure-activity relationships characterized the lactone moiety as the key part of the molecule responsible for activity. These results prompt further investigation towards this class of compounds and biological evaluation of other derivatives is currently ongoing. This work demonstrates that novel potent inhibitors of the nucleocytoplasmic transport based on naturally occurring molecules could contribute to the development of a therapy for cancer treatment.

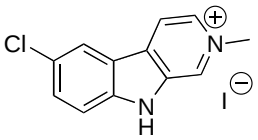


The second part of this work was dedicated to synthetic studies on sporolides A and B, two complex marine natural compounds proposed to derive from the *Bergmann* cyclization of an enediyne precursor. Enediynes are known to be highly cytotoxic compounds that induce DNA strand breaks. For the synthesis of sporolides A and B a biomimetic approach through an enediyne intermediate was investigated. Among the key reactions were a *Morita-Baylis-Hillman*, a *Sharpless* asymmetric dihydroxylation, an enediyne formation *via Wittig* reaction and a $CeCl_3$•2LiCl-mediated acetylide addition. Although preliminary attempts to form the 9-membered enediyne core structure were unsuccessful, further investigations are ongoing.

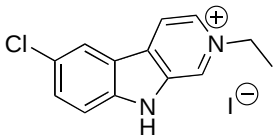


The last chapter was dedicated to malaria, a disease that kills over 1 million humans annually mostly in developing countries. The development of new quaternary *β*-carboline alkaloids derived from nostocarboline and eudistomin N against malaria parasites was reported. A straightforward synthesis allowed the preparation of these derivatives, which were submitted to biological evaluation against four parasites. The derivatives display pronounced activity against *Plasmodium falciparum.* Among the 6-chloro norharmane derivatives, the parent compound nostocarboline proved to be the most active and selective with an IC_{50} value of 194 nM and low cytotoxicity against rat myoblast L6 cells, resulting in a selectivity index of 634. Stronger activities were measured for 6-bromo norharmane derivatives, where the *N*-methylated analog of eudistomin N displayed an IC_{50} value of 18 nM and an elevated selectivity being 4783 times less toxic against L6 rat myoblast cell line. Five derivatives were selected and are currently under *in vivo* biological evaluation in a *P. berghei* mouse model. Structure-activity relationship analysis of the derivatives clearly shows a trend between the surface area of the compounds and its activity,

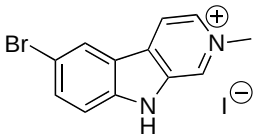
furnishing some leads for the development of new potent compounds. The simple, cheap and straightforward synthesis of these derivatives makes them interesting candidates as antiplasmodial drugs.



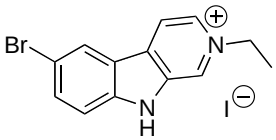
Nostocarboline
0.194 μM (*P. falciparum* K1)
120.9 μM (L6 rat cell line)



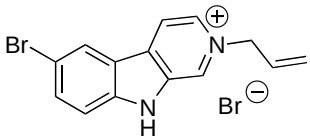
0.452 μM (*P. falciparum* K1)
113.0 μM (L6 rat cell line)



0.018 μM (*P. falciparum* K1)
86.1 μM (L6 rat cell line)



0.032 μM (*P. falciparum* K1)
78.8 μM (L6 rat cell line)



0.492 μM (*P. falciparum* K1)
130.9 μM (L6 rat cell line)

When considering therapeutic small molecule treatments they should be able to be constructed in an economic and efficient manner. Although natural compounds are often too complex to be considered as useful candidates in this sense, they are often a source of inspiration and motivation in the discovery process. In this work we have demonstrated the importance of nature as a source of biologically active compounds and how synthetic chemistry can contribute in the development and the discovery of therapeutics for treating human diseases. The synergy between synthetic chemistry and natural products remains fundamental and continues to play an important role in science.

6. Experimental Section

6.1. General Methods and Materials

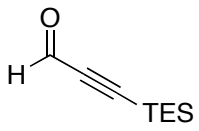
Unless otherwise stated, chemicals were purchased from Sigma-Aldrich, ABCR, Acros or Lancaster and used without further purification. Solvents for work-up and chromatography were distilled from technical quality. Solvents used for chemical transformations were either puriss quality or dried by filtration through activated aluminium oxide under argon or nitrogen (H_2O content < 30 ppm, *Karl-Fisher* titration). All non-aqueous reactions were run in oven-dried or flame-dried glassware under a positive pressure of argon or nitrogen. Concentration under reduced pressure was performed by rotary evaporation at 40 °C (unless otherwise specified). Yields refer to purified, dried and spectroscopically pure compound. Analytical thin layer chromatography (TLC) was performed on Merck silica gel 60 F_{254} plates (0.25 mm thickness) precoated with fluorescent indicator. The developed plates were examined under UV light and stained with ceric ammonium molybdate followed by heating. Flash chromatography was performed using silica gel 60 (230-240 mesh) from Fluka using a forced flow eluant at 0.3-0.5 bar pressure. Kugelrohr distillations were performed with a Büchi Glass Oven B-585. All ^1H and ^{13}C NMR spectra were recorded using either Varian Gemini 300 MHz (^1H) or 75 MHz (^{13}C), Varian Mercury 300 MHz (^1H) or 75 MHz (^{13}C), Bruker DRX 500 MHz (^1H) or 125 MHz (^{13}C), Bruker DPX 400 MHz (^1H) or 100 MHz (^{13}C), Bruker DRX 600 MHz (^1H) or 150 MHz (^{13}C), Bruker Advance 800 MHz (^1H) or 200 MHz (^{13}C) FT spectrometers at room temperature. Chemical shifts δ are reported in ppm, multiplicity is reported as follows: s = singlet, d = doublet, t = triplet, q = quartet, quint. = quintet, sext. = sextet, sept. = septet, m = multiplet or unresolved and coupling constant *J* in Hz. Analytical gas chromatography (GC) was performed on *Hewelett Packard, HP6810. Column*: supelco β dex 120, 30 m x 0.25 mm x 0.25 μm. *Carrier gas*: H_2. *Temperature*: 120 °C isothermal. *Flow*: 2 mL/min. *Split ratio*: 40:1. *Detector*: FID. Analytical high-performance liquid chromatography (HPLC) was performed on a *Dionex Chromatography System* (Interface Chromeleon, ASI-100 automated sample injector, UV detector 170U or PDA-100 photodiode array detector, pump P680, TCC thermostated column compartment, degaser, MSQ-ESI mass spectrometric detector).

The *flow rate* was 1 mL / min. *Column*: Phenomenex Gemini (5 µm) (C18 (150 x 4.6 mm)), solvent A: H_2O, solvent B: MeOH). Semi-preparative reversed-phase high-performance liquid chromatography (SP-HPLC) was performed on a *Dionex Chromatography System* (Interface Chromeleon, UV detector 170U or PDA-100 photodiode array detector, pump P680, TCC thermostated column compartment, degaser). The *flow rate* was 5 mL / min. *Column*: Phenomenex Gemini (5 µm) (C18 110A (150 x 10 mm)), solvent A: H_2O, solvent B: MeOH). All separations were performed at ambient temperature. IR spectra were recorded using a *Varian 2000 FT-IR ATR Spectrometer* or *Varian 800 FT-IR ATR Spectrometer*. The absorptions are reported in cm^{-1} and the IR bands were assigned as *s* (strong), *m* (medium) or *w* (weak). Optical rotations $[\alpha]^T_D$ were measured at the sodium D line using a 1 mL cell with a 1 dm path length on a Jasco DIP 1000 digital polarimeter, Jasco P-1020 digital polarimeter, Jasco P-2000 digital polarimeter and the concentration *c* is given in g/100mL and the used solvent is $CHCl_3$, MeOH or H_2O. Elemental analyses were performed by Mikroanalyse Labor of the Laboratorium für Organische Chemie der ETH Zürich or by Dr. *Euro Solari* in the Laboratory of Supramolecular Chemistry at the EPF Lausanne. All masses spectra were recorded by the Mass spectroscopy Service of Laboratorium für Organische Chemie der ETH Zürich on VG-TRIBRID (EI-MS) spectrometer and spectra measured at 70 eV, on TSQ 7000 ESI or by the Mass spectroscopy Service of EPF Lausanne on MICROMASS (ESI) Q-TOF Ultima API. Fragment ions are given in *m/z* with relative intensities (%) in parentheses. X-ray analyses were performed by Dr. *B. Schweizer* at the ETH Zürich or Dr. *R. Scopelliti* at the EPF Lausanne. UV spectra were measured on a *Varian Cary 1 Bio* UV-Visible spectrophotometer in a *Starna* quartz cell (10 mm path length). Lyophilisations were performed using a *Christ Freeze Dryer Alpha 1-2 LD plus.* Melting points (M.p.) were determined using a Büchi B-545 apparatus in open capillaries and are uncorrected.

6.2. Total Syntheses of Anguinomycins C & D

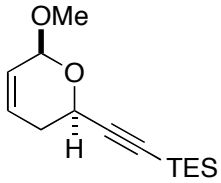
6.2.1. Synthesis of the C(1)-C(7) Fragment

3-(triethylsilyl)propionaldehyde (2)



To a suspension of Mg (0.50 g, 20.0 mmol, 1.00 equiv) in dry THF (80 mL) was added EtBr (1.50 mL, 20.0 mmol, 1.00 equiv) and the mixture was stirred at RT until all Mg was consumed. The resultant solution was added dropwise to a solution of trimethylsilylacetylene (**1**) (3.58 mL, 20.0 mmol, 1.00 equiv). The mixture was heated at reflux for 5 minutes and slowly added *via* canula to a solution of DMF (9.52 mL, 122 mmol, 6.10 equiv) in THF (80 mL) forming a white precipitate. The reaction was heated at reflux for 5 minutes, acidified to pH≈7 with dilute HCl solution, diluted with water (200 mL) and extracted with Et_2O (3 x 100 mL). The organic layer was dried ($MgSO_4$), filtered, and concentrated. The residue was diluted in Et_2O and washed with dilute $CuSO_4$ solution (pH ≈ 5) and saturated $NaHCO_3$. The organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was purified by flash chromatography on SiO_2 (cyclohexane/AcOEt 100:0 → 98:2) to give aldehyde **2** (2.98 g, 13.5 mmol, 67%) as a pale yellow oil. R_f = 0.46 (cyclohexane/AcOEt 9.5:0.5). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 9.18 (s, 1 H), 1.01 (t, *J* = 7.8 Hz, 9 H), 0.68 (q, *J* = 7.8 Hz, 6 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 176.4, 103.4, 101.3, 7.3, 3.8. FTIR ν 2959*m*, 2915*w*, 2879*m*, 2175*w*, 1689*s*, 1670*s*, 1461*w*, 1405*w*, 1262*m*, 1236*m*, 1002*m*, 913*w* cm^{-1}.

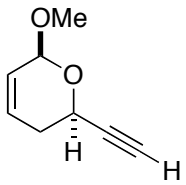
Triethyl(((2*R*,6*S*)-6-methoxy-3,6-dihydro-2*H*-pyran-2-yl)ethynyl)silane (5)



In a 10 mL flask under Ar was added 4 Å molecular sieves (1.26 g), **4** (0.30 g, 0.29 mmol, 0.02 equiv, 2.3 mol%), aldehyde **2** (2.12 g, 12.6 mmol, 1.00 equiv) and 1-methoxy-1,3-butadiene (1.28 mL, 12.6 mmol, 1.00 equiv) and the mixture was stirred at RT for 18 hours. The reaction was diluted with pentane, filtered through Celite and concentrated. The residue was purified by chromatography on SiO_2 (pentane/Et_2O 100:0 → 98:2) to afford **5** (2.73 g, 10.8 mmol, 86%, *e.e.* = 96.2) as a colorless oil. R_f = 0.37 (pentane/Et_2O 9.5:0.5). Optical rotation $[\alpha]^{27.9}_D$ (*c* 0.92, $CHCl_3$) = +105.8°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.96-5.90 (m, 1 H), 5.66 (dq, J_1 = 10.3 Hz, J_2 = 1.9 Hz, 1

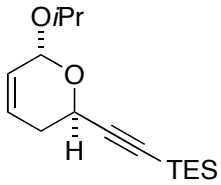
H), 5.01-4.98 (m, 1 H), 4.54 (dd, J_1 = 7.3 Hz, J_2 = 4.9 Hz, 1 H), 3.46 (s, 3 H), 2.42-2.20 (m, 2 H), 0,96 (t, J = 7.9 Hz, 9 H), 0.56 (q, J = 7.9 Hz, 6 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 127.5, 126.6, 105.5, 97.2, 86.1, 61.5, 55.2, 31.3, 7.5, 4.3. GC (β-dex chiral column) (T = 120°C): $t_{R1(minor)}$ = 42.08 minutes, $t_{R2\ (major)}$ = 43.00 minutes and *e.e.* = 96.2. Elemental analysis calcd for $C_{14}H_{24}O_2Si$: [C] 66.61 %, [H] 9.58 %, [O] 12.68 %, [Si] 11.13 %; found [C] 66.61 %, [H] 9.67 %. LRMS-ESI 275.3 (100, $[M+Na]^+$). FTIR ν 2956*m*, 2879*m*, 1982*w*, 1735*w*, 1336*w*, 1036*m*, 763*s*, 740*s* cm^{-1}.

(2*R*,6*S*)-2-ethynyl-6-methoxy-3,6-dihydro-2*H*-pyran (6)



To a solution of **5** (200 mg, 0.79 mmol, 1.00 equiv) in THF (6.30 mL) at 0°C was dropwise added TBAF (1 M in THF) (3.16 mL, 3.16 mmol, 4.00 equiv). The reaction was stirred for 15 min, warmed to RT, stirred for 1 h and quenched with water (20 mL). The mixture was extracted with Et_2O (3 x 30 mL) and the combined organic layers were washed with brine (1 x 30 mL), dried ($MgSO_4$), filtered and carefully concentrated *in vacuo* at 0 °C. The deprotected alkyne **6** was dried over molecular sieves and used directly in the next step without further purification. R_f = 0.27 (pentane/Et_2O 9:1). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.00 (dtd, J_1 = 10.3 Hz, J_2 = 4.0 Hz, J_3 = 1.5 Hz, 1 H), 5.74 (qd, J_1 = 10.3 Hz, J_2 = 2.0 Hz, 1 H), 5.01-4.99 (m, 1 H), 4.62 (dt, J_1 = 5.7 Hz, J_2 = 2.3 Hz, 1 H), 3.50 (s, 3 H), 2.40 (d, J = 2.3 Hz, 1 H), 2.37 (ddd, J_1 = 7.8 Hz, J_2 = 4.1 Hz, J_3 = 2.1 Hz, 2 H).

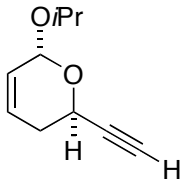
Triethyl(((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)ethynyl)silane (7)



To a solution of *p*TsOH (76.0 mg, 0.40 mmol, 1.00 equiv) in *i*PrOH (0.4 M) (1.00 mL) was added **5** (100 mg, 0.40 mmol, 1.00 equiv) and the solution was stirred at RT for 2 hours. The reaction was quenched with dilute $NaHCO_3$ and extracted with Et_2O (3 x 20 mL). The organic layer was dried ($MgSO_4$), filtered and concentrated to afford **7** (96.0 mg, 0.34 mmol, 86%) as a colorless oil, which was used without further purification. Optical rotation $[\alpha]^{28.7}_D$ (*c* 0.795, $CHCl_3$) = +33.7°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.96 (dd, J_1 = 10.0 Hz, J_2 = 5.4 Hz, 1 H), 5.71 (dd, J_1 = 10.1 Hz, J_2 = 1.1 Hz,

1 H), 5.14 (br. s, 1 H), 4.71 (dd, J_1 = 11.1 Hz, J_2 = 3.7 Hz, 1 H), 4.07 (sept., J = 6.2 Hz, 1 H), 2.41 (dd, J_1 = 17.7 Hz, J_2 = 11.2 Hz, 1 H), 2.23 (dd, J_1 = 17.7 Hz, J_2 = 4.1 Hz, 1 H), 1.29 (d, J = 6.2 Hz, 3 H), 1.19 (d, J = 6.1 Hz, 3 H), 1.00 (t, J = 7.8 Hz, 9 H), 0.63 (q, J = 7.8 Hz, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 128.2, 126.3, 106.1, 93.4, 87.0, 70.3, 58.0, 32.1, 24.2, 24.4, 7.8, 4.7. LRMS-ESI 303.2 (100, $[M+Na]^+$). FTIR ν 2957*m*, 2012*m*, 2877*m*, 2186*w*, 1697*w*, 1461*w*, 1380*w*, 1317*w*, 1182*w*, 1098*w*, 1059*w*, 1024*s*, 1000*s*, 799*w*, 726*s* cm^{-1}.

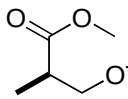
(2*R*,6*R*)-2-ethynyl-6-isopropoxy-3,6-dihydro-2*H*-pyran (8)



To a cooled (0 °C) solution of **7** (2.97 g, 10.6 mmol, 1.00 equiv) in THF (26.0 mL) was added TBAF (1 M in THF) (10.6 mL, 10.6 mmol, 1.0 equiv). The reaction was stirred for 15 minutes, warmed to RT, stirred for 1 hour and quenched with water (50 mL). The mixture was extracted with Et_2O (3 x 40 mL) and the combined organic layers were washed with brine (1 x 60 mL), dried ($MgSO_4$), filtered and carefully concentrated *in vacuo* at 0 °C. The residue was purified by chromatography on SiO_2 (pentane/Et_2O 100:0 → 95:5) to give the deprotected alkyne **8** (1.68 g, 10.1 mmol, 95%) as a colorless volatile oil. R_f = 0.45 (cyclohexane/AcOEt 9:1). Optical rotation $[\alpha]^{26.9}_D$ (*c* 0.58, $CHCl_3$) = +80.6°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.93 (dd, J_1 = 10.1 Hz, J_2 = 5.7 Hz, 1 H), 5.68 (ddd, J_1 = 10.2 Hz, J_2 = 2.9 Hz, J_3 = 1.3 Hz, 1H), 5.10 (br. s, 1 H), 4.67 (dddd, J_1 = 11.2 Hz, J_2 = 3.7 Hz, J_3 = 2.2 Hz, J_4 = 0.6 Hz, 1 H), 4.03 (sept., J = 6.3 Hz, 1 H), 2.44 (d, J = 2.2 Hz, 1 H), 2.37 (dddd, J_1 = 11.2 Hz, J_2 = 4.3 Hz, J_3 = 2.1 Hz, J_4 = 0.6 Hz, 1H), 2.19 (dddd, J_1 = 17.8 Hz, J_2 = 5.2 Hz, J_3 = 3.8 Hz, J_4 = 1.3 Hz, 1 H), 1.25 (d, J = 6.2 Hz, 3 H), 1.16 (d, J = 6.2 Hz, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 128.0, 126.4, 93.3, 83.2, 73.1, 70.3, 57.3, 32.4, 24.2, 22.4. FTIR ν 3306*m*, 2971*m*, 2928*m*, 2053*w*, 1736*w*, 1380*w*, 1184*w*, 1023*m*, 1002*w*, 784*s* cm^{-1}.

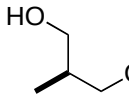
6.2.2. Synthesis of the C(8)-C(11) Fragment

(*R*)-methyl 3-(*tert*-butyldimethylsilyloxy)-2-methylpropanoate (10)



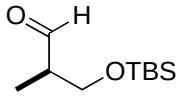
To a cooled (0 °C) solution of (*R*)-methyl-3-hydroxy-2-methyl propionate (**9**) (5.00 mL, 39.7 mmol, 1.00 equiv) in CH_2Cl_2 (40 mL) was added TBSCl (8.38 g, 55.6 mmol, 1.40 equiv) and imidazole (5.95 g, 87.3 mmol, 2.20 equiv) and the reaction was warmed to RT and stirred for 2 hours. The mixture was filtered through Celite, washed with HCl (0.1 M) (100 mL), H_2O (3 x 100 mL) and brine (1 x 100 mL), dried ($MgSO_4$), filtered and concentrated to afford **10** (9.23 g, 39.7 mmol, quant.) which was used without further purification. R_f = 0.60 (cyclohexane/AcOEt 8:2). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 3.76 (dd, J_1 = 9.7 Hz, J_2 = 6.9 Hz, 1 H), 3.66 (s, 3 H), 3.63 (dd, J_1 = 9.7 Hz, J_2 = 6.0 Hz, 1 H), 2.64 (sext., J = 7.0 Hz, 1 H), 1.12 (d, J = 7.0 Hz, 1 H), 0.85 (s, 9 H), 0.02 (d, J = 0.9 Hz, 6 H).

(*S*)-3-(*tert*-butyldimethylsilyloxy)-2-methylpropan-1-ol (11)



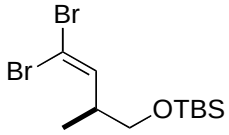
To a cooled (–78 °C) solution of (*R*)-methyl 3-(*tert*-butyldimethylsilyloxy)-2-methylpropanoate (**10**) (9.06 g, 39.0 mmol, 1.00 equiv) in CH_2Cl_2 (190 mL) was dropwise added DIBAL-H (1 M in hexanes) (78.0 mL, 78.0 mmol, 2.00 equiv). The mixture was stirred for 15 minutes at –78 °C, warmed to RT and stirred for 1 hour. The mixture was cooled at –78 °C, quenched with NaH_2PO_4/Na_2HPO_4 buffer solution (pH = 7.2) (94.0 mL) and allowed to return at RT over 1.5 hours. The solution was filtered through Celite and the organic layer was washed with water (2 x 150 mL) and brine (1 x 150 mL). The filter cake was washed with EtOAc (2 x 100 mL) and the aqueous layers were extracted a second time with EtOAc (2 x 100 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated to afford the alcohol **11** (7.97 g, 39.0 mmol, quant.), which was used without further purification. R_f = 0.28 (cyclohexane/AcOEt 8:2). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 3.72 (dd, J_1 = 9.8 Hz, J_2 = 4.5 Hz, 1 H), 3.61 (s, 1 H), 3.55 (dd, J_1 = 16.6 Hz, J_2 = 8.7 Hz, 1 H), 3.46 (dd, J_1 = 14.1 Hz, J_2 = 7.1 Hz, 1 H), 2.90 (s, 1 H), 1.98-1.85 (m, 1H), 0.88 (s, 9 H), 0.82 (d, J = 7.0 Hz, 3 H), 0.06 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 69.0, 68.6, 37.2, 26.0, 18.4, 13.2, –5.4, –5.4.

(*R*)-3-(*tert*-butyldimethylsilyloxy)-2-methylpropanal (12)



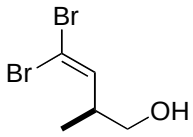
To a cooled (10 °C) solution of alcohol **11** (5.00 g, 24.5 mmol, 1.00 equiv) in DMSO (126 mL) was added Et_3N (8.20 mL, 58.8 mmol, 2.40 equiv) and pyridine sulfur trioxide (7.80 g, 49.0 mmol, 2.00 equiv). The solution was stirred at RT for 3 hours, cooled to 10 °C, quenched with H_2O (60 mL) and extracted with cyclohexane (3 x 80 mL). The organic layer was dried ($MgSO_4$), filtered and concentrated to obtain aldehyde **12** (4.95 g, 24.5 mmol, quant.) as a pale yellow oil which was used without further purification. R_f = 0.60 (cyclohexane/AcOEt 8:2). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 9.73 (d, J = 1.6 Hz, 1 H), 3.74 (ddd, J_1 = 15.6 Hz, J_2 = 9.8 Hz, J_3 = 6.4 Hz, 2 H), 2.71-2.60 (m, 1 H), 1.17 (d, J = 7.1 Hz, 3 H), 0.88 (s, 9 H), 0.06 (s, 6 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 180.0, 64.8, 42.0, 25.7, 18.1, 13.0, –5.6.

(*S*)-*tert*-butyl(4,4-dibromo-2-methylbut-3-enyloxy)dimethylsilane (13)



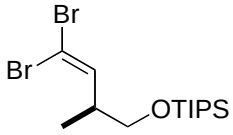
To a suspension of Zn (2.41 g, 36.8 mmol, 3.00 equiv) in CH_2Cl_2 (210 mL) were added PPh_3 (9.65 g, 36.8 mmol, 3.00 equiv) and CBr_4 (12.2 g, 36.8 mmol, 3.00 equiv). The mixture was stirred at RT for 2 days, treated with aldehyde **12** (2.48 g, 12.3 mmol, 1.00 equiv) and stirred for 1 day. The mixture was diluted with cyclohexane and the CH_2Cl_2 was removed *in vacuo*. The residue was triturated with cyclohexane, filtered, washed with cyclohexane and concentrated to obtain dibromo olefin **13** (3.74 g, 10.4 mmol, 85%) as a pale yellow oil. An analytical sample was purified by flash chromatography on SiO_2 (cyclohexane 100%). R_f = 0.36 (cyclohexane 100%). Optical rotation $[\alpha]^{29.0}_D$ (*c* 0.89, $CHCl_3$) = +4.6°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.27 (d, J = 9.3 Hz, 1 H), 3.51 (dd, J_1 = 5.8 Hz, J_2 = 0.8 Hz, 2 H), 2.70-2.56 (m, 1 H), 1.02 (d, J = 6.8 Hz, 3 H), 0.90 (s, 9 H), 0.05 (s, 6 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 141.3, 88.4, 66.0, 41.1, 27.0, 18.4, 15.6, –5.2. HRMS-EI calcd for $C_7H_{13}Br_2OSi$: $[M–C_4H_9]^+$ 298.9097; found 298.9097.

(*S*)-4,4-dibromo-2-methylbut-3-en-1-ol (19)



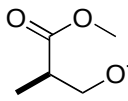
To a cooled (0 °C) solution of dibromo olefin **18** (3.00 g, 8.37 mmol, 1.00 equiv) in THF (21.0 mL) was added TBAF (1 M in THF) (0.87 mL, 8.37 mmol, 1.00 equiv). The solution was stirred 5 minutes at 0 °C, warmed to RT, stirred for 3 hours and quenched with water. The mixture was extracted with Et_2O (4 x 40 mL) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (cyclohexane/EtOAc 99:1 → 98:2) to afford alcohol **19** (0.95 g, 3.90 mmol, 47%) as a colorless oil. R_f = 0.41 (cyclohexane/AcOEt 7:3). Optical rotation $[\alpha]^{27.3}_D$ (*c* 0.68, $CHCl_3$) = +0.6°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.28 (d, *J* = 9.3 Hz, 1 H), 3.53 (d, *J* = 5.2 Hz, 2 H), 2.75-2.61 (m, 1 H), 1.84 (m, 1 H), 1.04 (d, *J* = 6.8 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 140.6, 89.5, 66.1, 41.1, 15.4. HRMS-EI calcd for $C_5H_8Br_2O$: $[M]^+$ 241.8937; found 241.8936. FTIR ν 3332*s*, 2965*m*, 2930*m*, 2872*m*, 1616*w*, 1454*w*, 1380*w*, 1261*w*, 1252*w*, 1036*m*, 989*w*, 784*s* cm^{-1}.

(*S*)-(4,4-dibromo-2-methylbut-3-enyloxy)triisopropylsilane (20)



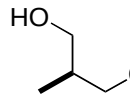
To a cooled (0 °C) solution of alcohol **19** (915 mg, 3.75 mmol, 1.00 equiv) in CH_2Cl_2 (3.75 mL) were added imidazole (511 mg, 7.50 mmol 2.00 equiv), TIPSCl (1.21 mL, 5.63 mmol, 1.50 equiv) and DMAP (cat.). After addition the solution was allowed to return to RT and stirred overnight. The reaction was diluted with CH_2Cl_2, washed with dilute HCl, water (3 x 30 mL) and brine (1 x 30 mL), dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (cyclohexane/EtOAc 100:0 → 99:1) to give product **20** (1.40 g, 3.49 mmol, 93%) as a colorless oil. R_f = 0.47 (hexane 100%). Optical rotation $[\alpha]^{27.6}_D$ (*c* 0.90, $CHCl_3$) = +12.9°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.31 (d, *J* = 9.2 Hz, 1 H), 3.62 (d, *J* = 2.0 Hz, 1 H), 3.60 (d, *J* = 2.1 Hz, 1 H), 2.73-2.58 (m, 1 H), 1.08-1.05 (m, 24 H). Elemental analysis calcd for $C_{14}H_{28}Br_2OSi$: [C] 42.01 %, [H] 7.05 %, [O] 4.00 %, [Si] 7.02 %, [Br] 39.93 %; found [C] 42.06 %, [H] 7.07 %, [Br] 40.02 %. HRMS-EI calcd for $C_{11}H_{21}Br_2OSi$: $[M–C_3H_7]^+$ 354.9723; found 354.9720. FTIR ν 2944*m*, 2866*m*, 1463*w*, 1215*m*, 1109*w*, 755*s*, 670*s* cm^{-1}.

(*R*)-methyl 2-methyl-3-(triisopropylsilyloxy)propanoate (21)



To a cooled (0 °C) solution of (*R*)-methyl-3-hydroxy-2-methyl propionate (**9**) (1.50 mL, 13.6 mmol, 1.00 equiv) in CH_2Cl_2 (13.6 mL) were added imidazole (2.04 g, 30.0 mmol, 2.20 equiv), TIPSCl (4.10 mL, 19.0 mmol, 1.40 equiv), DMAP (cat.). The reaction was allowed to return to RT and stirred overnight. The reaction was diluted with CH_2Cl_2, washed with diluted HCl (pH = 3) (3x), H_2O (2x), dried ($MgSO_4$). Purification by flash chromatography on SiO_2 (cyclohexane/EtOAc 9.5:0.5) afforded product **21** (3.73 g, 13.6 mmol, quant.) as a colorless oil. R_f = 0.53 (cyclohexane/AcOEt 9:1). Optical rotation $[\alpha]^{24.1}_D$ (*c* 1.00, $CHCl_3$) = –19.6°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 3.85 (dd, J_1 = 9.4 Hz, J_2 = 6.7 Hz, 1 H), 3.75 (dd, J_1 = 9.4 Hz, J_2 = 6.0 Hz, 1 H), 3.66 (s, 3 H), 2.72-2.60 (m, 1 H), 1.15 (d, J = 7.0 Hz, 3 H), 1.05-1.00 (m, 21 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 175.7, 65.8, 51.6, 42.8, 18.1, 13.6, 12.1. Elemental analysis calcd for $C_{14}H_{30}O_3Si$: [C] 61.26 %, [H] 11.02 %, [O] 17.49 %, [Si] 10.23 %; found [C] 61.53 %, [H] 10.78 %. HRMS-EI calcd for $C_{11}H_{23}O_3Si$: $[M–C_3H_7]^+$ 231.1411; found 231.1410. FTIR ν 2943*s*, 2867*s*, 1743*s*, 1463*m*, 1435*w*, 1389*w*, 1250*m*, 1198*m*, 1176*m*, 1105*s*, 1068*m*, 1027*w*, 882*m*, 797*w*, 682*m* cm^{-1}.

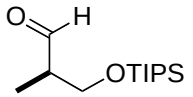
(*S*)-2-methyl-3-(triisopropylsilyloxy)propan-1-ol (22)



To a cooled (–78 °C) solution of **21** (12.4 g, 45.0 mmol, 1.00 equiv) in CH_2Cl_2 (230 mL), DIBAL-H (1 M in hexane) (78.0 mL, 78.0 mmol, 2.00 equiv) was added dropwise. The mixture was stirred for 1 hour at –78 °C, then between –20 °C and –15 °C for 30 minutes. The reaction was quenched by addition of MeOH and saturated Rochelle's salt. The mixture was vigorously stirred at RT for 1 hour. The aqueous phase was extracted with CH_2Cl_2 (3x) and the combined organic layers washed with brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (cyclohexane/EtOAc 9.5:0.5 → 7:3) afforded alcohol **22** (9.30 g, 37.7 mmol, 84%) and aldehyde **23** (1.78 g, 7.3 mmol, 16%) as a colorless oil. R_f = 0.56 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.25, $CHCl_3$) = –6.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 3.87 (dd, J_1 = 9.7 Hz, J_2 = 4.3 Hz, 1 H), 3.69-3.62 (m, 3 H), 3.03 (br. s, 1 H), 2.07-1.96 (m, 1 H), 1.17-1.03 (m, 21 H), 0.86 (d, J = 7.0 Hz, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 69.9, 69.0, 37.5, 18.3, 13.4, 12.1.

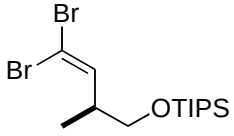
Elemental analysis calcd for $C_{13}H_{30}O_2Si$: [C] 63.35 %, [H] 12.27 %, [O] 12.98 %, [Si] 11.40 %; found [C] 63.55 %, [H] 12.13 %. HRMS-EI calcd for $C_{10}H_{23}O_2Si$: $[M–C_3H_7]^+$ 203.1462; found 203.1464. FTIR ν 3368*m*, 2943*s*, 2866*s*, 1463*m*, 1384*w*, 1247*w*, 1096*s*, 1035*s*, 995*m*, 881*s*, 791*m*, 680*s*, 668*s*, 659*m* cm^{-1}.

(*R*)-2-methyl-3-(triisopropylsilyloxy)propanal (23)



To a cooled (15 °C) solution of alcohol **22** (10.1 g, 40.9 mmol, 1.00 equiv) in DMSO (225 mL) was sequentially added Et_3N (13.7 mL, 98.2 mmol, 2.40 equiv) and pyridine sulfur trioxide (13.0 g, 81.8 mmol, 2.00 equiv). The solution was stirred for 5 minutes at 15 °C, then allowed to return to RT and stirred for 1.5 hours. The solution was cooled with an ice bath, quenched by addition of water (300 mL), diluted with hexane (750 mL) and stirred for 2 hours at RT. The aqueous layer was extracted with hexane (3x) and the combined organic layer washed with water (1x) and brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (cyclohexane/EtOAc 97:3) afforded aldehyde **23** (10.0 g, 40.9 mmol, quant.) as a colorless oil. R_f = 0.83 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.45, $CHCl_3$) = –35.6°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.80 (d, *J* = 1.5 Hz, 1 H), 4.00 (dd, J_1 = 10.0 Hz, J_2 = 5.1 Hz, , 1 H), 3.92 (dd, J_1 = 9.9 Hz, J_2 = 6.4 Hz, 1 H), 2.61-2.53 (m, 1 H), 1.13 (d, *J* = 7.0 Hz, 3 H), 1.10-1.05 (m, 21 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 205.2, 64.4, 49.5, 18.4, 12.3, 10.7. HRMS-EI calcd for $C_{13}H_{28}O_2NaSi$: $[M–Na]^+$ 267.1756; found 267.1762. FTIR ν 2961*m*, 2930*m*, 2858*m*, 1782*m*, 1696*m*, 1461*w*, 1384*m*, 1251*w*, 1204*m*, 1100*w*, 1054*w*, 835*w*, 773*w* cm^{-1}.

(*S*)-(4,4-dibromo-2-methylbut-3-enyloxy)triisopropylsilane (20)

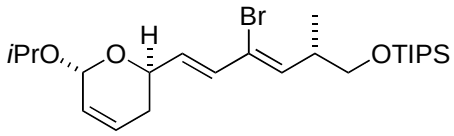


To a cooled (0 °C) solution of CBr_4 (16.0 g, 48.2 mmol, 2.20 equiv) in CH_2Cl_2 (87 mL), PPh_3 (25.3 g, 96.3 mmol, 4.40 equiv) was added in portion over 2 minutes. The solution turned from clear to brown and after 15 minutes at 0 °C, a solution of aldehyde **23** (5.35 g, 21.9 mmol, 1.00 equiv) and 2,6-lutidine (5.61 mL, 48.2 mmol, 2.20 equiv) in CH_2Cl_2 (87 mL) was added by canula over 20 minutes. The resulting dark-brown mixture was stirred at 0 °C for 2.5 hours. The reaction was quenched by addition of saturated

NH_4Cl and stirred for 30 minutes at RT. The aqueous layer was extracted with CH_2Cl_2 (2x) and the combined organic layer washed with saturated $NaHCO_3$ (1x) and brine (1x), dried ($MgSO_4$) and concentrated. The residue was triturated in hexane and the filtered concentrated. Purification by chromatography on SiO_2 (hexane 100%) afforded dibromo olefin **20** (5.58 g, 14.0 mmol, 64%) as a colorless oil. Analytical data matched those previously reported for the preparation of the same compound **20**.

6.2.3. Synthesis of the Alkyl Iodides Fragments

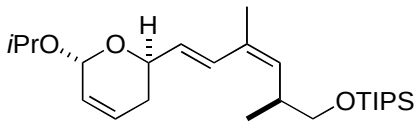
((*S*,3*Z*,5*E*)-4-bromo-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2-methylhexa-3,5-dienyloxy)triisopropylsilane (24)



To a cooled (0 °C) solution of alkyne **8** (312 mg, 1.87 mmol, 1.00 equiv) in THF (9.40 mL, 0.2 M *vs* **8**) was added Cp_2ZrHCl (374 mg, 1.44 mmol, 1.20 equiv). The flask was covered with an aluminium foil, stirred for 5 min at 0 °C and 1 hour at RT. In a separate flask $ZnCl_2$ (357 mg, 2.62 mmol, 1.40 equiv) was fused and dissolved in THF (11.2 mL). The solution was added to the solution of alkenylzirconocene at RT and the reaction stirred at RT for 30 minutes. In a separate flask, to a mixture of $Pd(PPh_3)_4$ (109 mg, 0.09 mmol, 0.05 equiv, 5 mol %) in THF (9.40 mL, 0.2 M *vs* **20**) was added DIBAL-H (10% in hexane) (187 µL, 0.19 mmol, 0.10 equiv, 10 %) and the mixture was stirred 20 minutes at RT and then dibromo olefin **20** (750 mg , 1.87 mmol, 1.00 equiv) was added. The dibromoolefin solution was stirred for 5 minutes at RT and then was added to the organozinc solution. The mixture was stirred 5 minutes at RT and then 13 hours at 40 °C. The reaction was quenched with water (30 mL) and extracted with Et_2O (3 x 40 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (CH_2Cl_2/cyclohexane 7:3) to give the coupled product **24** (756 mg, 1.55 mmol, 83%) as a pale yellow oil. R_f = 0.39 (CH_2Cl_2/cyclohexane 7:3). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.97, $CHCl_3$) = +50.0°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.28 (dd, J_1 = 14.8 Hz, J_2 = 1.2 Hz, 1 H), 6.07 (dd, J_1 = 14.8 Hz, J_2 = 5.3 Hz, 1 H), 6.02-5.97 (m, 1 H), 5.88 (d, J = 8.9 Hz, 1 H), 5.72 (ddd, J_1 = 10.0 Hz, J_2 = 4.3 Hz, J_3 = 2.6 Hz, 1 H), 5.12 (d, J = 2.8 Hz, 1 H), 4.58-4.51 (m, 1 H), 4.00 (sept., J = 6.2 Hz, 1 H), 3.61 (ddd, J_1 = 15.8 Hz, J_2 = 9.4 Hz, J_3 = 5.8 Hz, 2

H), 2.99-2.86 (m, 1 H), 2.10-2.05 (m, 2 H), 1.22 (d, *J* = 6.2 Hz, 3 H), 1.17 (d, *J* = 6.1 Hz, 3 H), 1.05 (s, 24 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 137.3, 133.4, 129.3, 128.2, 126.0, 124.0, 93.1, 69.6, 66.8, 65.7, 39.5, 30.9, 24.0, 22.1, 18.1, 16.2, 12.1. HRMS-EI calcd for $C_{44}H_{43}BrO_3Si$: $[M–C_3H_7]^+$ 443.1612; found 443.1610. FTIR ν 2942*m*, 2893*m*, 2866*m*, 1463*w*, 1383*w*, 1180*w*, 1102*m*, 1028*s*, 1000*m*, 952*w*, 883*w*, 787*m*, 684*m* cm^{-1}.

((*S*,3*Z*,5*E*)-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2,4-dimethyl-hexa-3,5-dienyloxy)triisopropylsilane (25)

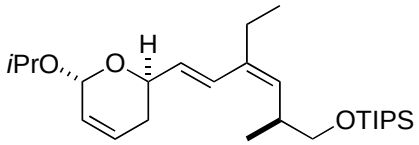


To a solution of **24** (100 mg , 0.23 mmol, 1.00 equiv) in THF (1.00 mL, 0.23 M *vs* **24**) was added $Pd(PPh_3)_4$ (24.0 mg, 0.02 mmol, 0.10 equiv). The solution was stirred for 10 minutes at RT, treated with Me_2Zn (2.0 M in toluene) (0.21 mL, 0.42 mmol, 2.00 equiv) and the reaction was stirred at 45 °C for 24 hours. An additional portion of Me_2Zn (0.10 mL, 0.21 mmol, 1.00 equiv) was added and the solution was stirred at 45 °C for 14 hours. The reaction was quenched with dilute NH_4Cl and extracted with Et_2O (3 x 15 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (CH_2Cl_2/cyclohexane 7:3) to afford product **25** (66.3 mg, 0.16 mmol, 68%, *d.r.* > 97:3) as a colorless oil. R_f = 0.21 (CH_2Cl_2/cyclohexane 7:3). Optical rotation $[\alpha]^{28.2}_D$ (*c* 0.62, $CHCl_3$) = +37.9°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.69 (d, *J* = 15.7 Hz, 1 H), 6.01 (dddd, J_1 = 7.7 Hz, J_2 = 5.3 Hz, J_3 = 1.9 Hz, J_4 = 0.9 Hz, 1 H), 5.77-5.67 (m, 2 H), 5.19 (d, *J* = 9.6 Hz, 1 H), 5.13-5.12 (m, 1 H), 4.54-5.47 (m, 1 H), 4.02 (sept., *J* = 6.2 Hz, 1 H), 3.50 (ddd, J_1 = 16.9 Hz, J_2 = 9.4 Hz, J_3 = 6.5 Hz, 2 H), 2.87-2.74 (m, 1 H), 2.20-2.00 (m, 2 H), 1.82 (d, *J* = 1.2 Hz, 3 H), 1.24 (d, *J* = 6.2 Hz, 3 H), 1.18 (d, *J* = 6.1 Hz, 3 H), 1.05-1.04 (m, 24 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 134.2, 131.3, 129.3, 128.4, 128.1, 126.0, 93.0, 69.4, 68.0, 66.9, 34.9, 30.7, 23.8, 21.9, 20.4, 17.9, 17.5, 11.9. Elemental analysis calcd for $C_{25}H_{46}O_3Si$: [C] 71.03, [H] 10.97, [O] 11.35, [Si] 6.64; found [C] 71.11, [H] 10.99. HRMS-EI calcd for $C_{25}H_{46}O_3Si$: $[M]^+$ 422,3211; found 422.3219. FTIR ν 2942*m*, 2867*m*, 1462*w*, 1382*w*, 1182*w*, 1122*w*, 1101*w*, 1029*m*, 1000*w*, 780*s*, 683*m* cm^{-1}.

Preparation of Cl_2Pd(DPEphos)

A mixture of $PdCl_2$ (200 mg, 1.12 mmol, 1.00 equiv) and LiCl (94.0 mg, 2.24 mmol, 2.00 equiv) in MeOH (2 mL) was heated to 50 °C for 10 minutes. DPE(phos) (638 mg, 1.18 mmol, 1.05 equiv) was added and the resulting mixture stirred at 50 °C for 8.5 hours, then cooled to RT, filtered, washed with MeOH and dried under high vacuum overnight affording Cl_2Pd(DPEphos) (755 mg, 1.05 mmol, 94%) as a yellow powder.

((*S*,3*Z*,5*E*)-4-ethyl-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2-methylhexa-3,5-dienyloxy)triisopropylsilane (26)

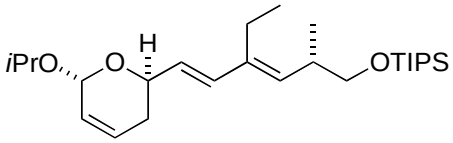


In a 5 mL flask containing Cl_2Pd(DPEphos) (2.20 mg, 0.003 mmol, 0.05 equiv) was added a solution of **24** (30.0 mg , 0.06 mmol, 1.00 equiv) in degassed[249] THF (0.75 mL). To the yellow mixture was slowly added Et_2Zn (1.5 M in toluene) (80 µL, 0.12 mmol, 2.00 equiv) and a pale yellow solution was obtained. The tube was sealed and stirred at 50 °C for 14 hours. The red-brown colored solution was quenched by slow addition of saturated NH_4Cl solution and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (hexane/acetone 99:1) to give product **26** (22.8 mg, 0.05 mmol, 84%, *d.r.* > 97:3) as a colorless oil. R_f = 0.65 (hexane/acetone 99.5:0.5). Optical rotation $[\alpha]^{26.4}_D$ (*c* 0.28, $CHCl_3$) = +38.5°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.61 (d, J = 15.9 Hz, 1 H), 6.04-5.99 (m, 1 H), 5.74 (dd, J_1 = 15.8 Hz, J_2 = 6.1 Hz, 1 H), 5.75-5.69 (m, 2 H), 5.19 (d, J = 9.5 Hz, 1 H), 5.13-5.12 (m, 1 H), 4.54-4.47 (m, 1 H), 4.02 (sept., J = 6.2 Hz, 1 H), 3.51 (ddd, J_1 = 16.6 Hz, J_2 = 9.4 Hz, J_3 = 6.5 Hz, 2 H), 2.79 (dq, J_1 = 9.3 Hz, J_2 = 6.6 Hz, 1 H), 2.20 (qd, J_1 = 7.4 Hz, J_2 = 0.9 Hz, 2 H), 2.14-2.00 (m, 2 H), 1.24 (d, J = 6.3 Hz, 3 H), 1.18 (d, J = 6.2 Hz, 3 H), 1.05-1.04 (m, 27 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 136.9, 132.1, 128.6, 128.4, 127.2, 126.0, 93.1, 69.5, 68.2, 67.1, 34.9, 30.9, 26.4, 23.9, 22.2, 18.1, 17.8, 17.7, 17.6, 13.3, 12.1, 12.0. Elemental analysis calcd for $C_{25}H_{46}O_3Si$: [C] 71.50, [H] 11.08, [O] 10.99, [Si] 6.43; found [C] 71.73, [H] 10.93. HRMS-EI calcd for

[249] The solvent was degassed using three freeze/pump/thaw cycles.

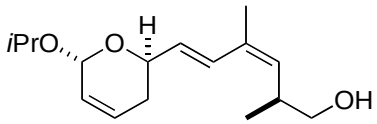
$C_{22}H_{39}O_3Si$: $[M–C_3H_7]^+$ 393.2820; found 393.2830. FTIR ν 2961*m*, 2867*m*, 1463*w*, 1381*w*, 1181*w*, 1100*m*, 1029*m*, 1002*m*, 882*w*, 785*s*, 683*m* cm^{-1}.

((*S*,3*E*,5*E*)-4-ethyl-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2-methylhexa-3,5-dienyloxy)triisopropylsilane (27)



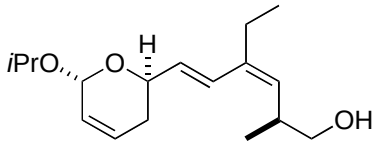
In a 5 mL flask containing $Pd(^tBu_3P)_2$ (0.60 mg, 0.001 mmol, 0.10 equiv) was added a solution of **24** (5.00 mg , 0.01 mmol, 1.00 equiv) in degassed THF (0.2 mL). To the mixture was slowly added Et_2Zn (1.5 M in toluene) (13 µL, 0.02 mmol, 2.00 equiv) and a pale yellow solution was obtained. The tube was sealed and stirred at 50 °C for 3.5 hours. The dark brown solution was quenched by addition of saturated NH_4Cl solution and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (hexane/acetone 99:1 → 99:5) to give product **27** (3.25 mg, 0.008 mmol, 75%, *d.r.* > 97:3) as a colorless oil. R_f = 0.65 (hexane/acetone 99.5:0.5). Optical rotation $[\alpha]^{22.4}_D$ (*c* 0.47, $CHCl_3$) = +21.0°. ^{1}H-NMR (500 MHz, $CDCl_3$) δ 6.17 (d, *J* = 15.9 Hz, 1 H), 6.05-6.02 (m, 1 H), 5.77-5.74 (m, 1 H), 5.67 (dd, J_1 = 15.9 Hz, J_2 = 6.4 Hz, 1 H), 5.27 (d, *J* = 9.5 Hz, 1 H), 5.15-5.14 (m, 1 H), 4.52-4.48 (m, 1 H), 4.06 (sept., *J* = 6.0 Hz, 1 H), 3.61 (dd, J_1 = 9.5 Hz, J_2 = 5.6 Hz, 1 H), 3.48 (dd, J_1 = 9.5 Hz, J_2 = 7.5 Hz, 1 H), 2.73-2.67 (m, 1 H), 2.34-2.25 (m, 2 H), 2.19-2.03 (m, 2 H), 1.28 (d, *J* = 6.4 Hz, 3 H), 1.21 (d, *J* = 6.0 Hz, 3 H), 1.10-1.07 (m, 24 H), 1.05 (d, *J* = 6.8 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 139.3, 135.6, 134.4, 128.6, 126.4, 126.1, 93.1, 69.4, 68.2, 67.0, 35.9, 31.0, 23.9, 22.1, 20.2, 18.1, 17.5, 14.2, 12.0. HRMS-ESI calcd for $C_{26}H_{48}O_3SiNa$: $[M+Na]^+$ 459.3271; found 459.3282. FTIR ν 2963*m*, 2943*m*, 2916*m*, 2866*m*, 1462*w*, 1381*w*, 1180*w*, 1099*w*, 1030*s*, 999*m*, 883*w*, 779*s*, 683*m* cm^{-1}.

(*S*,3*Z*,5*E*)-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2,4-dimethyl-hexa-3,5-dien-1-ol (28)



To a cooled (0 °C) solution of **25** (13.8 mg, 0.03 mmol, 1.00 equiv) in THF (160 μL) was added TBAF (1 M in THF) (64 μL, 0.06 mmol, 2.00 equiv). The reaction was stirred 1 hour at 0 °C and then 1 hour at RT. The reaction was quenched with water and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (hexane/AcOEt 8:2) to give alcohol **28** (8.4 mg, 0.03 mmol, 99%) as a colorless oil. R_f = 0.19 (CH_2Cl_2/AcOEt 9:1). Optical rotation $[\alpha]^{28.9}_D$ (*c* 0.49, $CHCl_3$) = +29.2°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.69 (d, *J* = 15.7 Hz, 1 H), 6.01 (ddd, J_1 = 10.0 Hz, J_2 = 4.7 Hz, J_3 = 2.1 Hz, 1 H), 5.77 (dd, J_1 = 15.8 Hz, J_2 = 6.0 Hz, 1 H), 5.76-5.70 (m, 1 H), 5.17-5.12 (m, 2 H), 4.52 (dt, J_1 = 10.3 Hz, J_2 = 5.3 Hz, 1 H), 4.01 (sept., *J* = 6.2 Hz, 1 H), 3.54-3.35 (m, 2 H), 2.94-2.79 (m, 1 H), 2.19-2.00 (m, 2 H), 1.86 (s, 3 H), 1.24 (d, *J* = 6.2 Hz, 3 H), 1.18 (d, *J* = 6.2 Hz, 3 H), 0.97 (d, *J* = 6.7 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 133.6, 130.5, 128.5, 127.9, 127.8, 126.3, 93.3, 69.8, 67.9, 66.9, 34.9, 30.9, 24.0, 22.2, 20.8, 17.3. HRMS-EI calcd for $C_{16}H_{26}O_3$: $[M]^+$ 266.1877; found 266.1869. FTIR ν 3416*m*, 2970*m*, 2925*m*, 1455*w*, 1379*w*, 1317*w*, 1126*w*, 1100*m*, 1027*s*, 999*s*, 774*m*, 670*m* cm^{-1}.

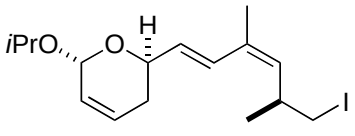
(*S*,3*Z*,5*E*)-4-ethyl-6-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-2-methyl-hexa-3,5-dien-1-ol (29)



To a cooled (0 °C) solution of **26** (200 mg , 0.46 mmol, 1.00 equiv) in THF (3.0 mL) was added TBAF (1 M in THF) (970 μL, 0.97 mmol, 2.10 equiv). The reaction was stirred 5 minutes at 0 °C and then 1.5 hour at RT. The reaction was cooled to 0°C, quenched with water and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (hexane/AcOEt 8:2 → 7:3) to give alcohol **29** (126 mg, 0.45 mmol, 98%) as a colorless oil. R_f = 0.25 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{22.7}_D$ (*c* 0.19, $CHCl_3$) = +21.2°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.62 (d, *J* =

16.0 Hz, 1 H), 6.03-5.99 (m, 1 H), 5.80 (dd, J_1 = 16.0 Hz, J_2 = 6.1 Hz, 1 H), 5.75-5.71 (m, 1 H), 5.14-5.12 (m, 2 H), 4.52 (m, 1 H), 4.02 (sept., J = 6.1 Hz, 1 H), 3.53-3.47 (m, 1 H), 3.41-3.36 (m, 1 H), 2.91-2.80 (m, 1 H), 2.25 (q, J = 7.4 Hz, 2 H), 2.17-2.02 (m, 2 H), 1.35 (dd, J_1 = 8.0, J_2 = 4.2 Hz, 1 H), 1.25 (d, J = 6.1 Hz, 3 H), 1.18 (d, J = 6.1 Hz, 3 H), 1.08 (t, J = 7.4 Hz, 3 H), 0.98 (d, J = 6.4 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 139.7, 131.7, 130.2, 128.9, 127.2, 126.6, 93.7, 70.1, 68.4, 67.4, 35.1, 31.2, 26.9, 24.3, 22.6, 17.7, 13.8. HRMS-ESI calcd for $C_{17}H_{27}O_3Na$: $[M + Na]^+$ 303.1931; found 303.1934. FTIR ν 3426*m*, 2967*m*, 2924*m*, 2874*m*, 1462*w*, 1381*w*, 1315*w*, 1261*w*, 1180*w*, 1099*m*, 1030*s*, 1003*m*, 799*w*, 718*w* cm^{-1}.

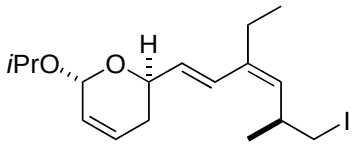
(2*R*,6*R*)-2-((*S*,1*E*,3*Z*)-6-iodo-3,5-dimethylhexa-1,3-dienyl)-6-isopropoxy-3,6-dihydro-2*H*-pyran (30)



To a cooled (0 °C) solution of alcohol **28** (4.00 mg, 0.015 mmol, 1.00 equiv) in a mixture toluene/Et_2O (375 μL/100μL) were added imidazole (14.4 mg, 0.21 mmol, 14.1 equiv) and PPh_3 (21.2 mg, 0.08 mmol, 5.4 equiv) and the resulting mixture stirred at 0 °C for 15 minutes. A solution of I_2 (19.8 mg, 0.078 mmol, 5.2 equiv) in Et_2O (375 μL) was added dropwise and the resulting mixture covered by an aluminium foil, stirred for 10 minutes at 0 °C and then 2 hours at RT. The mixture was directly filtered over cotton and concentrated. The residue was diluted in pentane, the precipitate filtered and the filtrated concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 100:0 → 99:1) afforded alkyl iodide **30** (4.2 mg, 0.011 mmol, 75%) as a colorless oil. R_f = 0.48 (hexane/AcOEt 8.5:1.5). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.11, $CHCl_3$) = +6.4°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.64 (d, J = 15.7 Hz, 1 H), 6.05-6.02 (m, 1 H), 5.80 (dd, J_1 = 15.7 Hz, J_2 = 5.8 Hz, 1 H), 5.77-5.75 (m, 1 H), 5.17 (d, J = 9.5 Hz, 1 H), 5.16 (s, 1 H), 4.58-4.53 (m, 1 H), 4.05 (sept., J = 6.2 Hz, 1 H), 3.17 (dd, J_1 = 9.4 Hz, J_2 = 5.7 Hz, 1 H), 3.09 (dd, J_1 = 9.4 Hz, J_2 = 7.3 Hz, 1 H), 2.92-2.82 (m, 1 H), 2.20-2.03 (m, 2 H), 1.87 (s, 3 H), 1.29 (d, J = 6.1 Hz, 3 H), 1.22 (d, J = 6.1 Hz, 3 H), 1.13 (d, J = 6.6 Hz, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 134.3, 132.7, 131.1, 128.6, 128.0, 126.7, 93.7, 70.1, 67.2, 34.4, 31.2, 24.3, 22.5, 21.9, 20.7, 15.2. HRMS-EI calcd for $C_{16}H_{25}O_2NaI$: $[M + Na]^+$ 399.0797; found 399.0801. FTIR ν

3322*w*, 2968*w*, 2924*w*, 1659*w*, 1377*w*, 1454*w*, 1377*w*, 1180*w*, 1099*w*, 1028*m*, 1000*m*, 785*s* cm^{-1}.

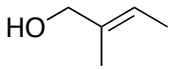
(2*R*,6*R*)-2-((*S*,1*E*,3*Z*)-3-ethyl-6-iodo-5-methylhexa-1,3-dienyl)-6-isopropoxy-3,6-dihydro-2*H*-pyran (31)



To a cooled (0 °C) solution of alcohol **29** (125 mg, 0.45 mmol, 1.00 equiv) in a mixture toluene/Et_2O (2:1) (20 mL), imidazole (425 mg, 6.24 mmol, 14. equiv) and PPh_3 (643 mg, 2.45 mmol, 5.5 equiv) were added and the resulting mixture was stirred at 0 °C for 10 minutes. A solution of I_2 (599 mg, 2.36 mmol, 5.3 equiv) in Et_2O (6 mL) was added dropwise over a period of 15 minutes. The resulting mixture was covered by an aluminium foil and stirred 0 °C for 45 minutes. The mixture was filtered and the precipitate washed with Et_2O. The precipitate was triturated in EtOAc and filtered. The combined organic phase was concentrated and the residue diluted in a mixture hexane/EtOAc 7:3 and filtered over a pad of silica and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 99.5:0.5 → 98:2) afforded alkyl iodide **31** (156 mg, 0.40 mmol, 89%) as a colorless oil. R_f = 0.52 (hexane/AcOEt 9.5:0.5). Optical rotation $[\alpha]^{22.7}_D$ (*c* 1.00, $CHCl_3$) = –2.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.53 (d, *J* = 16.0 Hz, 1 H), 6.03-6.00 (m, 1 H), 5.80 (dd, J_1 = 15.7 Hz, J_2 = 6.1 Hz, 1 H), 5.75-5.72 (m, 1 H), 5.14-5.12 (m, 2 H), 4.54-4.49 (m, 1 H), 4.03 (sept., *J* = 6.4 Hz, 1 H), 3.14 (dd, J_1 = 9.3 Hz, J_2 = 5.4 Hz, 1 H), 3.07 (dd, J_1 = 9.3 Hz, J_2 = 7.4 Hz, 1 H), 2.88-2.79 (m, 1 H), 2.22 (q, *J* = 7.4 Hz, 2 H), 2.14-2.02 (m, 2 H), 1.27 (d, *J* = 6.4 Hz, 3 H), 1.19 (d, *J* = 6.1 Hz, 3 H), 1.11 (d, *J* = 6.7 Hz, 3 H), 1.07 (t, *J* = 7.4 Hz, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 138.5, 132.5, 130.3, 128.9, 127.2, 126.6, 93.7, 70.2, 67.4, 34.4, 31.2, 26.7, 24.4, 22.6, 22.0, 15.7, 13.7. HRMS-ESI calcd for $C_{17}H_{27}O_2NaI$: $[M + Na]^+$ 413.0953; found 413.0941. FTIR ν 2967*m*, 2928*m*, 2878*w*, 1454*w*, 1377*w*, 1315*w*, 1180*w*, 1126*w*, 1099*w*, 1030*s*, 1003*m*, 964*w*, 718*w* cm^{-1}.

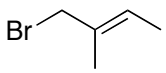
6.2.4. Synthesis of the Polyketidic Chain

(*E*)-2-methylbut-2-en-1-ol (34)



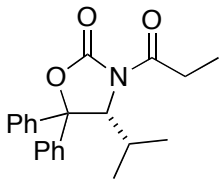
In a 1L three-necked round bottom flask equipped with a condenser, a suspension of $LiAlH_4$ (19.2 g, 510 mmol, 2.05 equiv) in Et_2O (100 mL) was cooled to 0 °C and a solution of tiglic acid (**33**) (24.7 g, 246 mmol, 1.00 equiv) was slowly added over a period of 1 hours. The resulting solution was stirred for 15 minutes at 0 °C and then 3 hours at RT. The reaction was cooled to 0 °C and quenched by careful addition of H_2O (18 mL), NaOH (15 %) (18 mL) and H_2O (54 mL). The white granular aluminium salts were filtered over Celite and washed with Et_2O (3x). The combined organic layers were washed with HCl (1 N) (1x), saturated $NaHCO_3$ solution (1x) and brine (1x), dried ($MgSO_4$) and concentrated to afford alcohol **34** (18.2 g, 211 mmol, 86%) as a colorless oil. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.51-5.44 (m, 1 H), 3.98 (m, 2 H), 1.65 (s, 3 H), 1.62-1.59 (m, 3 H). FTIR ν 3335*s*, 2919*m*, 2863*m*, 1674*w*, 1447*w*, 1381*w*, 1003*s*, 829*w*, 774*w*, 668*m* cm^{-1}.

(*E*)-1-bromo-2-methylbut-2-ene (35)



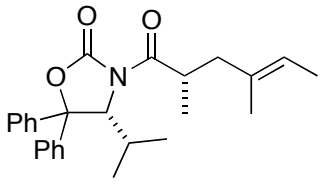
A solution of alcohol **34** (1.00 g, 11.6 mmol, 1.00 equiv) in Et_2O (23.0 mL, 0.5 M) was cooled to 0 °C and PBr_3 (0.55 mL, 5.80 mmol, 0.50 equiv) was added dropwise. The resulting solution was stirred at 0 °C for 30 minutes and then at RT for 3 hours. The reaction was quenched and washed with an aqueous K_2CO_3 solution (1x) and brine (1x), dried ($MgSO_4$) and carefully concentrated under reduced pressure to afford (*E*)-1-bromo-2-methylbut-2-ene (**35**) (1.25 g, 8.41 mmol, 73%) as a colorless oil. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.73-5.65 (m, 1 H), 3.98 (m, 2 H), 1.76-1.75 (m, 3 H), 1.63 (ddd, J_1 = 6.8 Hz, J_2 = 1.6 Hz, J_3 = 0.8 Hz, 3 H).

(*R*)-4-isopropyl-5,5-diphenyl-3-propionyloxazolidin-2-one (36)



To a cooled (0 °C) suspension of (*R*)-4-isopropyl-5,5-diphenyloxazolidin-2-one (**32**) (41.2 g, 0.15 mmol, 1.00 equiv) in THF (580 mL), *n*BuLi (1.6 M in hexane) (96.0 mL, 0.15 mmol, 1.05 equiv) was added dropwise. Propionyl chloride (15.2 mL, 0.18 mmol, 1.20 equiv) was added and the resulting solution stirred 5 minutes at 0 °C and then at RT overnight. The reaction was poured in saturated NH_4Cl solution and extracted with Et_2O (3x). The combined organic layers were washed with HCl (1 M) solution (2x), NaOH (1 M) solution (2x) and brine (1x), dried ($MgSO_4$) and concentrated. The crude was recrystallized in a mixture Et_2O/pentane to afford (*R*)-4-isopropyl-5,5-diphenyl-3-propionyloxazolidin-2-one (**36**) (46.7 g, 0.14 mmol, 95%) as a white crystalline solid. Optical rotation $[\alpha]^{27.8}_D$ (*c* 1.00, $CHCl_3$) = +224.2°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.50-7.46 (m, 2 H), 7.42-7.25 (m, 8 H), 5.38 (d, *J* = 3.3 Hz, 1 H), 2.99-2.89 (m, 1 H), 2.80-2.68 (m, 1 H), 2.02-1.92 (m, 1 H), 1.09 (t, *J* = 7.4 Hz, 1 H), 0.88 (d, *J* = 7.0 Hz, 1 H), 0.76 (d, *J* = 6.8 Hz, 1 H). The same procedure was adopted for the preparation of *ent*-**36** obtained in 94% yield. Optical rotation $[\alpha]^{27.8}_D$ (*c* 1.00, $CHCl_3$) = –228.8°.

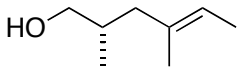
(*R*)-3-((*S*,*E*)-2,4-dimethylhex-4-enoyl)-4-isopropyl-5,5-diphenyloxazolidin-2-one (37)



In a 1L double-necked round bottom flask, a solution of DIPA (11.7 mL, 89.0 mmol, 1.25 equiv) in THF (200 mL) was cooled to 0 °C and *n*BuLi (1.6 M in hexane) (55.7 mL, 89.0 mmol, 1.25 equiv) was slowly added. The resulting solution was stirred at 0 °C for 30 minutes and then cooled to –78 °C. A precooled solution of **36** (24.0 g, 71.0 mmol, 1.00 equiv) in THF (130 mL) was slowly added and the resulting mixture stirred at –78 °C for 30 minutes followed by the slow addition of a precooled solution of (*E*)-1-bromo-2-methylbut-2-ene (**35**) (22.2 g, 149 mmol, 2.10 equiv) in THF (60 mL). The reaction was stirred at –78 °C for 5 minutes and then allowed to warm up to –10 °C while stirring was continued for 26 hours. The reaction was quenched by addition of saturated NH_4Cl solution and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$) and concentrated. The

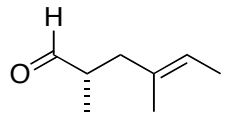
crude pale yellow solid was washed with a small amount of ice-cold pentane to afford product **37** (26.4 g, 65.0 mmol, 92%, *d.r.* > 97:3) as a white crystalline solid. R_f = 0.50 (cyclohexane/EtOAc 9:1). M.p. = 101-103 °C. Optical rotation $[\alpha]^{28.3}_D$ (*c* 1.00, $CHCl_3$) = +177.0°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.48-7.44 (m, 2 H), 7.42-7.26 (m, 8 H), 5.40 (d, *J* = 3.2 Hz, 1H), 5.30-5.22 (m, 1 H), 3.90 (sext., *J* = 7.2 Hz, 1 H), 2.54 (dd, J_1 = 13.4 Hz, J_1 = 7.2 Hz, 1 H), 2.01-1.89 (m, 2 H), 1.65-1.64 (m, 3 H), 1.55 (dd, J_1 = 6.7 Hz, J_2 = 1.0 Hz, 3 H), 0.85 (d, *J* = 7.0 Hz, 3 H), 0.79 (d, *J* = 6.8 Hz, 3 H), 0.74 (d, *J* = 6.7 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 176.7, 152.7, 142.2, 138.0, 132.8, 128.6, 128.4, 128.2, 127.7, 125.7, 125.5, 120.9, 89.0, 64.2, 43.6, 35.3, 29.6, 21.5, 16.1, 16.0, 15.3, 13.2. Elemental analysis calcd for $C_{26}H_{31}NO_3$: [C] 77.01 %, [H] 7.70 %, [N] 3.45 %; found [C] 76.79 %, [H] 7.67 %, [N] 3.52 %. HRMS-EI calcd for $C_{26}H_{31}NO_3$: $[M]^+$ 405.2299; found 405.2301. FTIR ν 2968*w*, 2934*w*, 2888*w*, 1776*s*, 1698*s*, 1495*w*, 1450*m*, 1385*m*, 1371*m*, 1348*m*, 1312*m*, 1246*m*, 1207*s*, 1174*s*, 1149*m*, 1123*m*, 1094*m*, 1056*m*, 1035*w*, 986*s*, 949*m*, 764*s*, 750*s*, 703*s*, 694*s*, 668*s*, 636*m* cm^{-1}.

(*S*,*E*)-2,4-dimethylhex-4-en-1-ol (38)



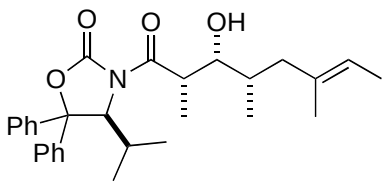
To a cooled (0 °C) suspension of $LiAlH_4$ (1.56 g, 41.2 mmol, 8.00 equiv) in Et_2O (20 mL) was slowly added a solution of **37** (2.09 g, 5.15 mmol, 1.00 equiv) in Et_2O (48 mL). The resulting solution was stirred for 30 minutes at 0 °C and then 3 hours at RT. The reaction was cooled to 0 °C and quenched by addition of H_2O (3 mL), NaOH (15 %) (3 mL) and H_2O (9 mL). The white granular aluminium salts were filtered over Celite and washed with Et_2O (3x). The combined organic layers were dried ($MgSO_4$) and concentrated to afford alcohol **38** (0.66 g, 5.15 mmol, 100%) as a colorless oil. R_f = 0.19 (cyclohexane/EtOAc 8.5:1.5). Optical rotation $[\alpha]^{24.6}_D$ (*c* 0.55, $CHCl_3$) = –4.7°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.24 (qd, J_1 = 6.6 Hz, J_2 = 1.2 Hz, 1 H), 3.52-3.39 (m, 2 H), 2.11-2.02 (m, 1 H), 1.89-1.77 (m, 2 H), 1.61-1.57 (m, 6 H), 0.86 (d, *J* = 6.5 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 134.3, 120.1, 68.5, 44.3, 33.7, 16.8, 15.7, 13.5. FTIR ν 3320*m*, 2917*m*, 1456*w*, 1037*m*, 786*s*, 668*w* cm^{-1}.

(*S*,*E*)-2,4-dimethylhex-4-enal (39)



To a cooled (–78 °C) solution of oxalyl chloride (867 µL, 9.94 mmol, 2.00 equiv) in CH_2Cl_2 (10.5 mL) was added dropwise a solution of DMSO (1.41 mL, 20.0 mmol, 4.00 equiv) in CH_2Cl_2 (10.5 mL). After 5 minutes a solution of alcohol **38** (637 mg, 4.97 mmol, 1.00 equiv) in CH_2Cl_2 (10.0 mL) was slowly added. Stirring at –78 °C was continued for 15 minutes, followed by addition of a solution of NEt_3 (4.16 mL, 29.8 mmol, 6 equiv) in CH_2Cl_2 (10.5 mL). The resulting solution was stirred at –78 °C for 20 minutes and then at 0 °C for 30 minutes. The reaction was quenched by addition of buffer phosphate (pH = 7) (32 mL) and the solution stirred at RT for 15 minutes. The organic phase was separated and the aqueous phase extracted with CH_2Cl_2 (3x). The combined organic layers were washed with water (2x) and brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (CH_2Cl_2/cyclohexane 7:3) afforded aldehyde **39** (619 mg, 4.91 mmol, 99%) as a colorless oil. R_f = 0.42 (pentane/Et_2O 9.5:0.5). Optical rotation $[\alpha]^{22.0}_D$ (*c* 0.93, $CHCl_3$) = +9.9°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 9.61 (d, *J* = 2.1 Hz, 1 H), 5.29-5.23 (m, 1 H), 2.57-2.45 (m, 1 H), 2.41 (dd, J_1 = 13.4 Hz, J_2 = 6.6, Hz, 1 H), 1.98 (dd, J_1 = 13.7 Hz, J_2 = 7.7 Hz, 1 H), 1.59 (s, 3 H), 1.58 (d, *J* = 7.0 Hz, 3 H), 1.03 (d, *J* = 6.8 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 205.5, 132.2, 121.6, 44.5, 40.9, 15.7, 13.5, 13.3. FTIR ν 2922*m*, 1708*w*, 1442*w*, 1378*w*, 777*s* cm^{-1}.

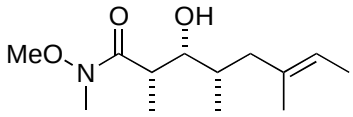
(*S*)-3-((2*S*,3*R*,4*S*,*E*)-3-hydroxy-2,4,6-trimethyloct-6-enoyl)-4-isopropyl-5,5-diphenyloxazolidin-2-one (40)



To a cooled (–5°C) solution of *ent*-**36** (84.4 mg, 0.25 mmol, 1.00 equiv) in CH_2Cl_2 (0.30 mL), Bu_2BOTf (1 M in CH_2Cl_2) (263 µL, 0.26 mmol, 1.05 equiv) was slowly added and the solution turns from colorless to pale green. NEt_3 (42 µL, 0.30 mmol, 1.20 equiv) was slowly added over a period of 5 minutes and the solution turned to pale yellow. Stirring at 0 °C was continued for 1 hour. The resulting solution was cooled to –78 °C and aldehyde **39** (63 mg, 0.50 mmol, 2.00 equiv) in CH_2Cl_2 (0.20 mL) was slowly added and the mixture stirred for 1 hour at –78 °C and finally for 1 hour at 0 °C. The reaction was quenched at 0 °C by

sequentially addition of buffer phosphate (pH = 7) (0.3 mL), MeOH (0.9 mL) and MeOH/H_2O_2 (2:1) (0.9 mL). The mixture was stirred for 1.5 hours at RT before dilution with Et_2O, washed with HCl (0.5 M) (1x), saturated $NaHCO_3$ solution (1x) and brine (1x), dried ($MgSO_4$) and concentrated. The residue was purified by chromatography on SiO_2 (Et_2O/pentane 8:2) to afford product **40** (89.2 mg, 0.19 mmol, 77%, *d.r.* > 87:13) as a white crystalline solid. R_f = 0.33 (pentane/Et_2O 7:3). M.p. = 98-99 °C. Optical rotation $[\alpha]^{24.5}_D$ (*c* 1.00, $CHCl_3$) = –103.6°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.53-7.50 (m, 2 H), 7.43-7.28 (m, 8 H), 5.37 (d, *J* = 3.6 Hz, 1 H), 5.18-5.11 (m, 1 H), 3.83-3.74 (m, 1 H), 3.43 (td, J_1 = 6.7 Hz, J_2 = 4.9 Hz, 1 H), 2.06-1.90 (m, 2 H), 1.86 (d, *J* = 5.1 Hz, 1 H), 1.66-1.57 (m, 2 H), 1.56 (d, *J* = 6.6 Hz, 3 H), 1.51 (s, 3 H), 1.31 (d, *J* = 6.9 Hz, 3 H), 0.86 (d, *J* = 6.9 Hz, 3 H), 0.78 (d, *J* = 6.8 Hz, 3 H), 0.41 (d, *J* = 6.7 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 176.1, 152.4, 142.2, 137.6, 133.6, 128.7, 128.4, 128.3, 127.8, 125.6, 125.2, 120.3, 89.4, 64.6, 44.0, 40.4, 33.0, 29.8, 21.7, 16.5, 15.4, 13.9, 13.5, 13.4. Elemental analysis calcd for $C_{29}H_{37}NO_4$: [C] 74.57 %, [H] 8.19 %, [N] 2.91 %; found [C] 74.68 %, [H] 8.03 %, [N] 2.91 %. HRMS-EI calcd for $C_{29}H_{35}NO_3$: $[M–H_2O]^+$ 445.2611; found 445.2611. FTIR ν 3475*m*, 2965*m*, 2931*m*, 1781*s*, 1697*m*, 1494*w*, 1450*m*, 1374*m*, 1316*w*, 1254*w*, 1208*s*, 1176*s*, 1050*m*, 987*m*, 954*w*, 760*m*, 704*m*, 668*m* cm^{-1}.

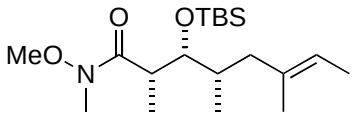
(2*S*,3*R*,4*S*,*E*)-3-hydroxy-*N*-methoxy-*N*,2,4,6-tetramethyloct-6-enamide (42)



To a cooled (0 °C) suspension of MeONHMe•HCl (503 mg, 5.16 mmol, 6.00 equiv) in CH_2Cl_2 (5.2 mL) was added $AlMe_3$ (2 M in toluene) (2.10 mL, 5.16 mmol, 6.00 equiv). The resulting solution was stirred at 0 °C for 5 minutes, then at RT for 1 hour. The clear solution was cooled to 0 °C and **40** (400 mg, 0.86 mmol, 1.00 equiv) in CH_2Cl_2 (1.0 mL) was added by canula. Stirring at 0 °C was continued for 5 minutes, then at RT for 15 hours. The reaction mixture was slowly transferred in a diluted HCl solution (0.5 M) (27.0 mL), diluted with more CH_2Cl_2 and stirred at RT for 1hour. The aqueous layer was separated and extracted with CH_2Cl_2 (3x). The combined organic phases were washed with saturated $NaHCO_3$ (1x) and brine (1x), dried ($MgSO_4$) and concentrated. The residue was diluted in ice-cold Et_2O, the precipitated cleaved auxiliary was filtered and the filtrate was concentrated.

Purification by chromatography on SiO_2 (pentane/Et_2O 4:6) afforded product **42** (179 mg, 0.74 mmol, 86%) as white crystalline solid. An analytical sample was recrystallized (hexane) for X-ray analysis (crystallographic data are given at the end of the experimental part). R_f = 0.21 (pentane/Et_2O 4:6). M.p. = 54-55 °C. Optical rotation $[\alpha]^{22.4}_D$ (*c* 0.50, $CHCl_3$) = +6.7°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.23 (q, *J* = 6.2 Hz, 1 H), 3.70 (s, 3 H), 3.57-3.53 (m, 1 H), 3.33 (d, *J* = 2.5 Hz, 1 H), 3.19 (s, 3 H), 3.12 (br. s, 1 H), 2.08 (d, *J* = 8.5 Hz, 1 H), 1.82-1.68 (m, 2 H), 1.60-1.58 (m, 6 H), 1.19 (d, *J* = 7.0 Hz, 3 H), 0.90 (d, *J* = 6.3 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 178.0, 133.8, 120.4, 75.3, 61.4, 43.7, 36.3, 33.0, 31.9, 15.2, 14.7, 13.2, 11.2. Elemental analysis calcd for $C_{13}H_{25}NO_3$: [C] 64.17 %, [H] 10.35 %, [N] 5.76 %, [O] 19.72 %; found: [C] 64.23 %, [H] 10.46 %, [N] 5.67 %. LRMS-ESI 266.2 (100, $[M + Na]^+$). FTIR ν 3452*m*, 2965*s*, 2934*s*, 1640*s*, 1513*w*, 1457*s*, 1382*s*, 1300*m*, 1249*m*, 1176*m*, 1122*m*, 993*s*, 826*w* cm^{-1}.

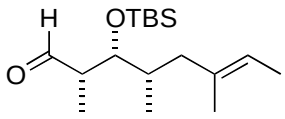
(2*S*,3*R*,4*S*,*E*)-3-(*tert*-butyldimethylsilyloxy)-*N*-methoxy-*N*,2,4,6-tetramethyloct-6-enamide (43)



To a cooled (–20 °C) solution of **42** (467 mg, 1.92 mmol, 1.00 equiv) in CH_2Cl_2 (4.0 mL) were sequentially added 2,6-lutidine (257 µL, 2.21 mmol, 1.15 equiv) and TBSOTf (354 µL, 2.02 mmol, 1.05 equiv). The resulting solution was stirred for 15 min at –20 °C; then at 0 °C for 45 min. The reaction mixture was diluted in more CH_2Cl_2 and washed with diluted citric acid (pH = 4) (1x), saturated $NaHCO_3$ (1x), brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 9:1) afforded **43** (680 mg, 1.90 mmol, 99%) as a clear oil. R_f = 0.38 (hexane/EtOAc 9:1). Optical rotation $[\alpha]^{24.3}_D$ (*c* 1.00 , $CHCl_3$) = +6.8°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.17 (q, *J* = 6.6 Hz, 1 H), 3.85 (dd, J_1 = 8.5 Hz, J_2 = 2.3 Hz, 1 H), 3.69 (s, 3 H), 3.16 (s, 3 H), 3.06 (br. s, 1 H), 2.14 (d, *J* = 12.4 Hz, 1 H), 1.86-1.78 (m, 1 H), 1.71-1.61 (m, 1 H), 1.56 (d, *J* = 6.6 Hz, 3 H), 1.52 (s, 3 H), 1.14 (d, *J* = 7.0 Hz, 3 H), 0.92 (s, 9 H), 0.73 (d, *J* = 6.8 Hz, 3 H), 0.08 (s, 6 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 176.9, 134.3, 119.7, 77.3, 61.4, 44.2, 39.0, 35.9, 32.4, 26.3, 18.6, 15.9, 15.4, 13.4, 13.3, –3.4, –3.5. Elemental analysis calcd for $C_{19}H_{39}NO_3Si$: [C] 63.82 %, [H] 10.99 %, [N] 3.92 %, [O] 13.42 %, [Si] 7.85 %; found [C] 63.79 %, [H] 11.00 %, [N]

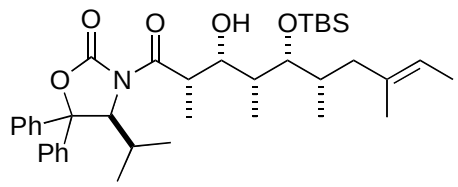
4.10 %. LRMS-ESI 380.2 (100, $[M + Na]^+$). FTIR ν 3369*s*, 2959*m*, 2931*m*, 2857*m*, 1662*s*, 1461*m*, 1382*m*, 1252*m*, 1176*w*, 1108*m*, 1049*s*, 997*s*, 869*m*, 833*s*, 773*s* cm^{-1}.

(2*S*,3*R*,4*S*,*E*)-3-(*tert*-butyldimethylsilyloxy)-2,4,6-trimethyloct-6-enal (44)



To a cooled (–78 °C) solution of **43** (663 mg, 1.85 mmol, 1.00 equiv) in THF (13.2 mL) was added DIBAL-H (1 M in hexane) (3.60 mL, 3.60 mmol, 2.00 equiv). The resulting solution was stirred at –78 °C for 1 hour; then quenched by addition of saturated Rochelle's salt, diluted in Et_2O and vigorously stirred at RT for 1 hour. The aqueous layer was extracted with Et_2O (3x) and the combined organic phase dried ($MgSO_4$) and concentrated (bath T < 20 °C). Purification by chromatography on SiO_2 (hexane/EtOAc 9.5:0.5) afforded aldehyde **44** (551 mg, 1.85 mmol, 100%) as a colorless oil. R_f = 0.70 (cyclohexane/EtOAc 9:1). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.20, $CHCl_3$) = +53.5°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.85 (s, 1 H), 5.22 (q, *J* = 6.5 Hz, 1 H), 4.00-3.98 (m, 1 H), 2.59-2.53 (m, 1 H), 2.16-2.09 (m, 1 H), 1.85-1.78 (m, 2 H), 1.60 (d, *J* = 6.7 Hz, 3 H), 1.57 (s, 3 H), 1.10 (d, *J* = 7.0 Hz, 3 H), 0.92 (s, 9 H), 0.78 (d, *J* = 6.1 Hz, 3 H), 0.11 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 205.7, 134.2, 120.9, 75.9, 51.4, 44.7, 35.1, 26.3, 18.7, 15.8, 14.7, 13.7, 9.7, –3.5, –3.7. HRMS-ESI calcd for $C_{17}H_{35}O_2Si$: $[M + H]^+$ 299.2406, found 299.2419.

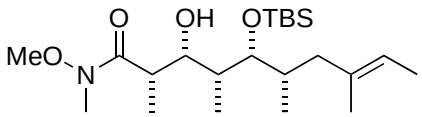
(*S*)-3-((2*S*,3*R*,4*R*,5*R*,6*S*,*E*)-5-(*tert*-butyldimethylsilyloxy)-3-hydroxy-2,4,6,8-tetramethyldec-8-enoyl)-4-isopropyl-5,5-diphenyloxazolidin-2-one (45)



To a cooled (–5°C) solution of *ent*-**36** (81.0 mg, 0.24 mmol, 1.20 equiv) in CH_2Cl_2 (0.48 mL) were sequentially added Bu_2BOTf (1 M in CH_2Cl_2) (240 µL, 0.24 mmol, 1.20 equiv) and NEt_3 (39 µL, 0.28 mmol, 1.40 equiv). Stirring at 0 °C was continued for 45 minutes; then the resulting solution was cooled to –78 °C and aldehyde **44** (59 mg, 0.20 mmol, 1.00 equiv) in CH_2Cl_2 (0.45 mL) was slowly added by canula. The reaction was stirred for 45 minutes at –78 °C, then allowed to return to 0 °C over 3 hours. The reaction was quenched at 0 °C by sequentially addition of buffer phosphate (pH = 7)

(0.24 mL), MeOH (0.72 mL) and MeOH/H_2O_2 (2:1) (0.72 mL). The mixture was stirred at RT for 30 minutes before dilution with Et_2O, washed with HCl (0.5 M) (1x), saturated $NaHCO_3$ (1x) and brine (1x), dried ($MgSO_4$) and concentrated. The residue was purified by chromatography on SiO_2 (hexane/EtOAc 9.5:0.5) to afford **45** (77.0 mg, 0.12 mmol, 61%, *d.r.* > 97:3) as a white crystalline solid. R_f = 0.60 (pentane/Et_2O 7:3). M.p. = 105-107 °C. Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.29, $CHCl_3$) = –118.6°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.52-7.51 (m, 2 H), 7.43-7.41 (m, 2 H), 7.37-7.26 (m, 6 H), 5.44 (d, *J* = 3.5 Hz, 1 H), 5.24 (q, *J* = 6.3 Hz, 1 H), 3.79-3.78 (m, 2 H), 3.50 (t, *J* = 3.8 Hz, 1 H), 2.49 (br. s, 1 H), 2.12 (d, *J* = 12.3 Hz, 1 H), 2.05-1.98 (m, 1 H), 1.82-1.76 (m, 1 H), 1.73-1.68 (m, 1 H), 1.62 (d, *J* = 6.6 Hz, 3 H), 1.58 (s, 3 H), 1.53-1.49 (m, 1 H), 1.36 (d, *J* = 6.4 Hz, 3 H), 0.89 (d, *J* = 7.1 Hz, 3 H), 0.87 (s, 9 H), 0.80 (d, *J* = 6.8 Hz, 3 H), 0.76 (d, *J* = 6.6 Hz, 3 H), 0.67 (d, *J* = 6.9 Hz, 3 H), 0.01 (s, 3 H), –0.24 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 177.3, 152.7, 142.6, 138.3, 134.8, 129.3, 129.0, 128.8, 128.4, 126.2, 125.8, 120.4, 89.7, 77.1, 74.0, 64.3, 44.2, 40.9, 38.4, 35.9, 30.3, 26.5, 22.1, 18.8, 16.7, 15.9, 15.3, 13.9, 13.8, 9.4, –3.0, –3.9. HRMS-ESI calcd for $C_{38}H_{57}NO_5NaSi$: $[M + Na]^+$ 658.3904, found 658.3911. FTIR ν 3360*w*, 2928*m*, 2857*m*, 1786*m*, 1693*w*, 1458*w*, 1374*w*, 1253*w*, 1210*w*, 1044*w*, 892*w*, 766*w*, 689*w* cm^{-1}.

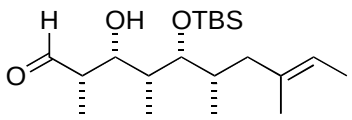
(2*S*,3*R*,4*R*,5*R*,6*S*,*E*)-5-(*tert*-butyldimethylsilyloxy)-3-hydroxy-*N*-methoxy-*N*,2,4,-6,8-pentamethyldec-8-enamide (46)



To a cooled (0 °C) suspension of MeONHMe·HCl (28.0 mg, 0.28 mmol, 6.00 equiv) in CH_2Cl_2 (140 μL) was added $AlMe_3$ (2 M in toluene) (142 μL, 0.28 mmol, 6.00 equiv). The resulting solution was stirred at 0 °C for 5 minutes, then at RT for 45 minutes. The clear solution was cooled to 0 °C and **45** (30.0 mg, 0.05 mmol, 1.00 equiv) in CH_2Cl_2 (100 μL) was added. Stirring at 0 °C was continued for 5 minutes, then at RT for 68 hours. The reaction was quenched by slow addition of diluted HCl solution (0.5 M) and stirred at RT for 1hour. The aqueous layer was separated and extracted with CH_2Cl_2 (3x). The combined organic phases were washed with saturated $NaHCO_3$ (1x) and brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 6:4) afforded product **46** (8.1

mg, 0.02 mmol, 41%). R_f = 0.19 (pentane/Et_2O 1:1). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.12, $CHCl_3$) = –7.5°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.19 (q, *J* = 6.21 Hz, 1 H), 3.83-3.79 (m, 1 H), 3.71 (s, 3 H), 3.54 (t, *J* = 3.5 Hz, 1 H), 3.21 (br s, 1 H), 3.19 (s, 3 H), 3.14 (br s, 1 H), 2.15 (d, *J* = 12.5 Hz, 1 H), 1.85-1.70 (m, 3 H), 1.57 (d, *J* = 7.3 Hz, 3 H), 1.55 (s, 3 H), 1.19 (d, *J* = 7.0 Hz, 3 H), 0.97 (d, *J* = 7.0 Hz, 3 H), 0.91 (s, 9 H), 0.78 (d, *J* = 6.6 Hz, 3 H), 0.07 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 178.4, 134.8, 120.5, 78.6, 74.5, 62.0, 44.6, 39.2, 38.1, 35.9, 26.6, 18.9, 15.9, 15.6, 13.8, 12.6, 10.7, –3.1, –3.5. HRMS-ESI calcd for $C_{22}H_{45}NO_4SiNa$: $[M + Na]^+$ 438.3016, found 338.3010. FTIR ν 3456*w*, 2959*m*, 2931*m*, 2858*w*, 1642*w*, 1462*w*, 1384*w*, 1254*w*, 1095*w*, 1041*m*, 1001*m*, 834*m*, 776*s*, 677*m*, 630*m* cm^{-1}.

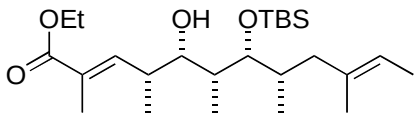
(2*S*,3*R*,4*R*,5*R*,6*S*,*E*)-5-(*tert*-butyldimethylsilyloxy)-3-hydroxy-2,4,6,8-tetramethyl-dec-8-enal (47)



To a cooled (–17 °C) solution of **45** (320 mg, 0.50 mmol, 1.00 equiv) in toluene (10 mL) was slowly added a solution of $LiAlH_4$ (1 M in Et_2O) (1.00 mL, 1.00 mmol, 2.00 equiv). The resulting solution was stirred for 20 minutes, then quenched at –17 °C by dropwise addition of saturated Rochelle's salt and diluted in Et_2O. The mixture was vigorously stirred at RT for 2 hours, then extracted with Et_2O (3x) and the combined organic phase dried ($MgSO_4$) and concentrated (bath T < 20 °C). The residue was diluted in Et_2O and the precipitated cleaved auxiliary recovered. The filtered was concentrated and the residue purified by chromatography on SiO_2 (pentane/Et_2O 9:1) to afford aldehyde **47** (149 mg, 0.42 mmol, 83%) as a colorless oil. R_f = 0.28 (pentane/Et_2O 7:3). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.08, $CHCl_3$) = –23.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.73 (d, *J* = 1.2 Hz, 1 H), 5.21 (q, *J* = 6.4 Hz, 1 H), 4.03 (q, *J* = 5.2 Hz, 1 H), 3.58 (dd, J_1 = 4.2 Hz, J_2 = 2.9 Hz, 1 H), 2.68-2.62 (m, 1 H), 2.20 (d, *J* = 12.3 Hz, 1 H), 1.97 (d, *J* = 4.4 Hz, 1 H), 1.89-1.77 (m, 2 H), 1.76-1.67 (m, 1 H), 1.60 (d, *J* = 6.8 Hz, 3 H), 1.57 (s, 3 H), 1.17 (d, *J* = 7.1 Hz, 3 H), 1.00 (d, *J* = 6.9 Hz, 3 H), 0.94 (s, 9 H), 0.81 (d, *J* = 6.7 Hz, 3 H), 0.11 (s, 3 H), 0.09 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 204.8, 134.5, 120.7, 78.2, 73.2, 50.1, 44.3, 39.3, 36.1, 26.5, 18.8, 15.8, 13.7, 10.0, 8.8, –2.8, –3.5. HRMS-ESI calcd for $C_{20}H_{40}O_3SiNa$: [M +

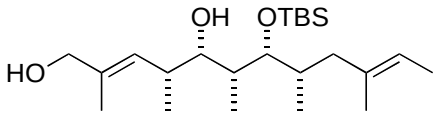
Na]$^+$ 379.2644, found 379.2639. FTIR ν 2957*m*, 2931*m*, 2859*m*, 1727*w*, 1462*w*, 1384*w*, 1255*w*, 1096*w*, 1032*w*, 837*w*, 775*w* cm^{-1}.

(2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-ethyl 7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,-10-pentamethyldodeca-2,10-dienoate (49)



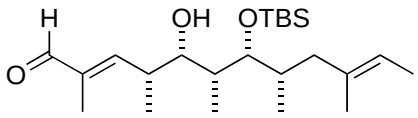
To a solution of aldehyde **47** (61.1 mg, 0.17 mmol, 1.00 equiv) in toluene (1.7 mL) was added 1-carbethoxyethylidentriphenylphosphorane (123.2 mg, 0.34 mmol, 2.00 equiv) and the mixture was stirred at 35 °C for 5 hours. The reaction was diluted in pentane, filtered over cotton and concentrated. The residue was purified by chromatography on SiO_2 (pentane/Et_2O 9:1) to afford **49** (73.8 mg, 0.17 mmol, 99%, *d.r.* > 97:3). R_f = 0.39 (pentane/Et_2O 8:2). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.09, $CHCl_3$) = +24.7°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.53 (dd, J_1 = 10.5 Hz, J_2 = 1.2 Hz, 1 H), 5.21 (q, *J* = 6.2 Hz, 1 H), 4.27-4.15 (m, 2 H), 3.65 (t, *J* = 3.9 Hz, 1 H), 3.53-3.49 (m, 1 H), 2.70-2.60 (m, 1 H), 2.17 (d, *J* = 12.6 Hz, 1 H), 1.93 (d, *J* = 4.3 Hz, 1 H), 1.89 (d, *J* = 1.1 Hz, 3 H), 1.87-1.82 (m, 1 H), 1.80-1.74 (m, 1 H), 1.72-1.66 (m, 1 H), 1.59 (d, *J* = 6.6 Hz, 3 H), 1.57 (s, 3 H), 1.31 (t, *J* = 7.1 Hz, 3 H), 1.10 (d, *J* = 6.6 Hz, 3 H), 0.94 (s, 9 H), 0.87 (d, *J* = 7.0 Hz, 3 H), 0.77 (d, *J* = 6.7 Hz, 3 H), 0.12 (s, 3 H), 0.11 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 168.5, 144.0, 134.7, 127.7, 120.5, 79.9, 78.6, 60.9, 44.1, 39.0, 38.0, 35.7, 26.5, 18.7, 16.9, 15.9, 15.0, 14.6, 13.7, 13.0, 8.8, –2.8, –3.7. HRMS-ESI calcd for $C_{25}H_{49}O_4Si$: [M + H]$^+$ 441.3400, found 441.3404. FTIR ν 3519*w*, 2959*m*, 2923*m*, 2858*m*, 1712*m*, 1650*w*, 1462*w*, 1369*w*, 1252*m*, 1094*m*, 1038*m*, 835*m*, 773*m*, 675*m* cm^{-1}.

(2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-2,4,6,8,10-pentamethyl-dodeca-2,10-diene-1,5-diol (50)



To a cooled (–78 °C) solution of **49** (67.0 mg, 0.15 mmol, 1.00 equiv) in THF (1.6 mL) was slowly added DIBAL-H (1M in hexane) (800 µL, 0.80 mmol, 5.30 equiv). The resulting solution was allowed to return to–15 °C and stirred from –15 °C to –5 °C over 1.5 hours. The reaction was quenched by addition of MeOH, diluted in saturated Rochelle's salt and Et_2O and vigorously stirred at RT for 1 hour. The aqueous layer was extracted with Et_2O (3x) and the combined organic phase dried ($MgSO_4$) and concentrated (bath T < 25 °C). Purification by chromatography on SiO_2 (pentane/Et_2O 9:1 → 7:3) afforded diol **50** (56.3 mg, 0.14 mmol, 93%) as a colorless oil. R_f = 0.15 (pentane/Et_2O 7:3). Optical rotation $[\alpha]^{22.5}_D$ (*c* 0.41, $CHCl_3$) = –1.0°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.23-5.17 (m, 2 H), 4.01 (s, 2 H), 3.63-3.60 (m, 1 H), 3.39 (d, *J* = 8.8 Hz, 1 H), 2.59-2.49 (m, 1 H), 2.16 (d, *J* = 12.2 Hz, 1 H), 1.91-1.75 (m, 4 H), 1.71 (d, *J* = 0.5 Hz, 3 H), 1.59 (d, *J* = 7.0 Hz, 3 H), 1.57 (s, 3 H), 1.04 (d, *J* = 6.6 Hz, 3 H), 0.93 (s, 9 H), 0.88 (d, *J* = 7.0 Hz, 3 H), 0.76 (d, *J* = 6.6 Hz, 3 H), 0.11 (s, 3 H), 0.10 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 135.0, 134.8, 129.1, 120.4, 79.9, 78.9, 69.2, 44.3, 38.5, 36.8, 35.6, 26.5, 18.8, 17.8, 15.9, 14.9, 14.3, 13.8, 9.0, –2.8, –3.6. HRMS-ESI calcd for $C_{23}H_{46}O_3NaSi$: $[M + Na]^+$ 421.3114, found 421.3116. FTIR ν 3349*m*, 2956*m*, 2930*m*, 2860*m*, 1459*w*, 1383*w*, 1253*m*, 1070*m*, 1035*m*, 1011*m*, 836*m*, 775*m*, 676*m* cm^{-1}.

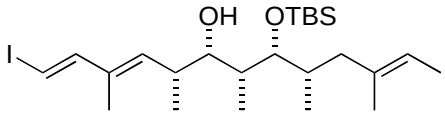
(2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (51)



To a solution of diol **50** (121 mg, 0.30 mmol, 1.00 equiv) in CH_2Cl_2 (3.0 mL), MnO_2 (396 mg, 4.50 mmol, 15.0 equiv) was added. The mixture was stirred at RT for 2.5 hours, then filtered over Celite, rinsed with CH_2Cl_2 and concentrated (bath T < 25 °C). The α,β-unsaturated aldehyde **51** (103 mg, 0.26 mmol, 86%) crystallized under high vacuum. An analytical sample was recrystallized (hexane) for X-ray analysis and the rest directly used in the next step without further

purification (crystallographic data are given at the end of the experimental part). R_f = 0.37 (pentane/Et_2O 7:3). M.p. = 75-77 °C. Optical rotation $[\alpha]^{22.5}_D$ (*c* 0.82, $CHCl_3$) = –10.9°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.42 (s, 1 H), 6.27 (dd, J_1 = 10.3 Hz, J_2 = 1.0 Hz, 1 H), 5.21 (q, *J* = 6.3 Hz, 1 H), 3.65 (t, *J* = 3.8 Hz, 1 H), 3.59-3.56 (m, 1 H), 2.92-2.82 (m, 1 H), 2.18 (d, *J* = 12.8 Hz, 1 H), 2.00 (d, *J* = 4.2 Hz, 1 H), 1.92-1.83 (m, 1 H), 1.81 (d, *J* = 0.9 Hz, 3 H), 1.79-1.73 (m, 1 H), 1.66-1.63 (m, 1 H), 1.60 (d, *J* = 7.1 Hz, 3 H), 1.57 (s, 3 H), 1.16 (d, *J* = 6.6 Hz, 3 H), 0.94 (s, 9 H), 0.90 (d, *J* = 7.0 Hz, 3 H), 0.77 (d, *J* = 6.8 Hz, 3 H), 0.13 (s, 3 H), 0.11 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 195.6, 156.4, 139.1, 134.5, 120.6, 79.8, 78.1, 44.1, 39.3, 38.3, 35.7, 26.5, 18.7, 16.7, 15.9, 15.2, 13.8, 9.9, 8.9, –2.8, –3.7. HRMS-ESI calcd for $C_{23}H_{44}O_3SiNa$: $[M + Na]^+$ 419.2957, found 419.2960. FTIR ν 3520*w*, 2961*m*, 2928*m*, 2889*m*, 2885*m*, 1667*m*, 1635*w*, 1459*w*, 1378*w*, 1251*w*, 1096*w*, 1073*w*, 1040*w*, 1011*m*, 974*w*, 883*m*, 772*m*, 681*m* cm^{-1}.

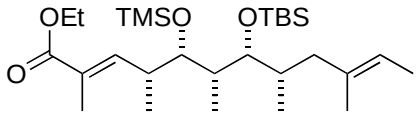
(1*E*,3*E*,5*R*,6*S*,7*R*,8*R*,9*S*,11*E*)-8-(*tert*-butyldimethylsilyloxy)-1-iodo-3,5,7,9,11-pentamethyltrideca-1,3,11-trien-6-ol (52)



To a cooled (–5 °C) suspension of $CrCl_2$ (446 mg, 3.63 mmol, 24.00 equiv) in dry THF (4.4 mL) was slowly added a solution of *α,β*-unsaturated aldehyde **51** (60.0 mg, 0.15 mmol, 1.00 equiv) and CHI_3 (358 mg, 0.91 mmol, 6.00 equiv) in THF (4.4 mL). The dark brown mixture was covered with an aluminium foil and stirred between –5 and 0 °C for 2.5 hours. The mixture was quenched by addition of water and extracted with Et_2O (3x). The combined organic layers were washed with saturated sodium thiosulfate (1x), water (1x), dried ($MgSO_4$) and concentrated (bath T < 20 °C). Purification by chromatography on SiO_2 (pentane/Et_2O 9:1) afforded vinyl iodide **52** (78.4 mg, 0.15 mmol, quant., *d.r.* > 97:3) as a colorless oil. R_f = 0.68 (pentane/Et_2O 7:3). Optical rotation $[\alpha]^{22.4}_D$ (*c* 0.60, $CHCl_3$) = +25.4°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.04 (d, *J* = 14.6 Hz, 1 H), 6.20 (d, *J* = 14.6 Hz, 1 H), 5.24-5.19 (m, 2 H), 3.63 (t, *J* = 3.9 Hz, 1 H), 3.43-3.40 (m, 1 H), 2.65-2.56 (m, 1 H), 2.17 (d, *J* = 12.3 Hz, 1 H), 1.87 (d, *J* = 4.3 Hz, 1 H), 1.86-1.82 (m, 1 H), 1.77 (d, *J* = 0.7 Hz, 3 H), 1.76-1.70 (m, 2 H), 1.60 (d, *J* = 6.9 Hz, 3 H), 1.58 (s, 3 H), 1.06 (d, *J* = 6.6 Hz, 3 H), 0.94 (s, 9 H), 0.86 (d, *J* = 7.0 Hz, 3 H), 0.77 (d, *J* =

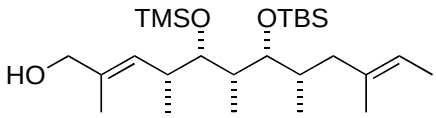
6.6 Hz, 3 H), 0.12 (s, 3 H), 0.11 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 150.0, 137.0, 134.7, 134.3, 120.5, 80.0, 78.8, 74.1, 44.2, 38.8, 37.4, 35.7, 26.5, 18.8, 17.6, 15.9, 15.0, 13.8, 12.6, 8.8, –2.7, –3.6. HRMS-ESI calcd for $C_{24}H_{45}O_2SiINa$: $[M + Na]^+$ 543.2131, found 543.2133. FTIR ν 3482*w*, 2958*m*, 2929*m*, 2858*m*, 1461*w*, 1387*w*, 1254*w*, 1091*w*, 1039*w*, 980*w*, 950*w*, 836*w*, 774*w*, 678*w* cm^{-1}.

(2*E*,4*R*,5*S*,6*S*,7*R*,8*S*,10*E*)-ethyl 7-(*tert*-butyldimethylsilyloxy)-2,4,6,8,10-penta-methyl-5-(trimethylsilyloxy)dodeca-2,10-dienoate (53)



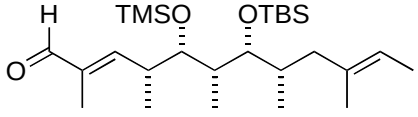
To a cooled (–5 °C) solution of **49** (7.6 mg, 0.017 mmol, 1.00 equiv) in CH_2Cl_2 (170 µL) were sequentially added DMAP (2.0 mg, 0.017 mmol, 1.00 equiv), NEt_3 (14 µL, 0.102 mmol, 6.00 equiv) and TMSCl (6.6 µL, 0.052 mmol, 3.00 equiv). The resulting solution was stirred at 0 °C for 1 hour; then quenched by addition of saturated NH_4Cl and extracted with CH_2Cl_2 (3x). The combined organic layers were dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 9.75:0.25) afforded the α,β-unsaturated ester **53** (6.7 mg, 0.13 mmol, 77%). R_f = 0.76 (pentane/Et_2O 9:1). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.285, $CHCl_3$) = +14.4°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.59 (dd, J_1 = 10.4 Hz, J_2 = 1.0 Hz, 1 H), 5.20-5.17 (m, 1 H), 4.26-4.10 (m, 2 H), 3.49 (dd, J_1 = 6.0 Hz, J_2 = 4.3 Hz, 1 H), 3.42 (dd, J_1 = 7.0 Hz, J_2 = 2.1 Hz, 1 H), 2.73-2.64 (m, 1 H), 2.03 (d, *J* = 12.7 Hz, 1 H), 1.91 (d, *J* = 10.9 Hz, 1 H), 1.85 (d, *J* = 1.0 Hz, 3 H), 1.82-1.76 (s, 1 H), 1.72-1.64 (s, 1 H), 1.59-1.56 (m, 6 H), 1.28 (t, *J* = 7.1 Hz, 3 H), 0.97 (d, *J* = 6.7 Hz, 3 H), 0.91 (s, 9 H), 0.85 (d, *J* = 6.9 Hz, 3 H), 0.70 (d, *J* = 6.5 Hz, 3 H), 0.15 (s, 9 H), 0.05 (s, 3 H), 0.02 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 168.6, 145.2, 134.6, 127.0, 120.6, 77.5, 77.4, 60.8, 45.8, 40.6, 37.8, 35.0, 26.7, 19.0, 16.0, 15.9, 14.6, 13.8, 13.0, 12.6, 12.0, 1.3, –2.6, –2.9. HRMS-ESI calcd for $C_{28}H_{56}O_4Si_2Na$: $[M + Na]^+$ 535.3615, found 535.3610. FTIR ν 2958*m*, 2932*m*, 2859*w*, 1714*m*, 1460*w*, 1384*w*, 1252*m*, 1096*m*, 1032*m*, 836*m*, 772*w*, 750*w*, 676*w*, 631*s* cm^{-1}.

(2*E*,4*R*,5*S*,6*S*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-2,4,6,8,10-pentamethyl-5-(trimethylsilyloxy)dodeca-2,10-dien-1-ol (54)



To a cooled (–78 °C) solution of α,β-unsaturated ester **53** (5.5 mg, 0.01 mmol, 1.00 equiv) in CH_2Cl_2 (100 µL) was slowly added DIBAL-H (1 M in hexane) (20 µL, 0.02 mmol, 2.00 equiv). The resulting solution was stirred at –78 °C for 1 hour, then quenched by addition of MeOH (0.1 mL), saturated Rochelle's salt (2 mL), diluted in more CH_2Cl_2 (2 mL) and vigorously stirred at RT for 1 hour. The aqueous layer was extracted with CH_2Cl_2 (3x) and the combined organic layers dried ($MgSO_4$) and concentrated (bath T < 20 °C). Purification by chromatography on SiO_2 (pentane/Et_2O 9:1) afforded alcohol **54** (4.7 mg, 0.01 mmol, 100%) as a colorless oil. R_f = 0.19 (cyclohexane/EtOAc 9:1). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.29, $CHCl_3$) = +1.7°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.23-5.21 (m, 2 H), 4.02 (d, *J* = 5.7 Hz, 2 H), 3.45-3.41 (m, 2 H), 2.64-2.54 (s, 1 H), 2.03 (d, *J* = 12.0 Hz, 1 H), 1.94 (t, *J* = 11.8 Hz, 1 H), 1.86-1.78 (m, 1 H), 1.78-1.72 (m, 1 H), 1.70 (s, 3 H),1.61-1.59 (m, 6 H), 0.95 (d, *J* = 7.0 Hz, 3 H), 0.93 (s, 9 H), 0.87 (d, *J* = 6.9 Hz, 3 H), 0.71 (d, *J* = 6.6 Hz, 3 H), 0.16 (s, 9 H), 0.08 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 134.7, 134.3, 130.1, 120.6, 78.1, 77.8, 69.3, 46.0, 40.2, 36.8, 34.7, 26.7, 19.0, 17.5, 16.0, 14.4, 13.8, 12.3, 11.9, 1.4, –2.5, –2.9. FTIR ν 3314*w*, 2958*m*, 2929*m*, 2858*w*, 1462*w*, 1381*w*, 1251*m*, 1127*w*, 1105*w*, 1061*w*, 1032*m*, 866*w*, 836*m*, 772*w* cm^{-1}.

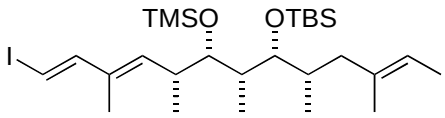
(2*E*,4*R*,5*S*,6*S*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-2,4,6,8,10-pentamethyl-5-(trimethylsilyloxy)dodeca-2,10-dienal (55)



To a solution of alcohol **54** (5.2 mg, 0.011 mmol, 1.00 equiv) in CH_2Cl_2 (110 µL) was added MnO_2 (14.7 mg, 0.165 mmol, 15.0 equiv). The mixture was stirred at RT for 3.5 hours, then filtered over Celite, rinsed with CH_2Cl_2 and concentrated (bath T < 20 °C). The α,β-unsaturated aldehyde **55** was obtained in quantitative yield and directly used in the next step without further purification. R_f = 0.60 (pentane/Et_2O 9:1). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.27, $CHCl_3$) = –1.9°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.41 (s, 1 H), 6.32 (d, *J* = 10.3 Hz, 1 H), 5.23-5.22 (m, 1 H), 3.58 (dd, J_1 = 6.5 Hz, J_2 = 4.0 Hz, 1 H), 3.45 (dd, J_1 = 7.1 Hz, J_2 = 2.1 Hz, , 1 H),

2.97-2.89 (m, 1 H), 2.05 (d, *J* = 12.5 Hz, 1 H), 1.93 (d, *J* = 11.0 Hz, 1 H), 1.84-1.82 (m, 1 H), 1.80 (d, *J* = 0.7 Hz, 3 H), 1.70-1.65 (m, 1 H), 1.62-1.60 (m, 6 H), 1.06 (d, *J* = 6.7 Hz, 3 H), 0.93 (s, 9 H), 0.89 (d, *J* = 6.9 Hz, 3 H), 0.72 (d, *J* = 6.6 Hz, 3 H), 0.18 (s, 9 H), 0.09 (s, 3 H), 0.08 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 195.7, 157.5, 138.5, 134.5, 120.8, 77.5, 77.0, 45.8, 40.9, 38.3, 34.9, 26.7, 19.0, 16.1, 16.0, 13.8, 12.7, 12.0, 9.9, 1.4, –2.6, –2.9. HRMS-ESI calcd for $C_{26}H_{53}O_3Si_2$: $[M]^+$ 469.3533, found 469.3534. FTIR ν 2959*m*, 2930*m*, 2858*w*, 1694*m*, 1471*w*, 1462*w*, 1381*w*, 1252*m*, 1123*w*, 1107*w*, 1031*m*, 837*m*, 772*w*, 631*s* cm^{-1}.

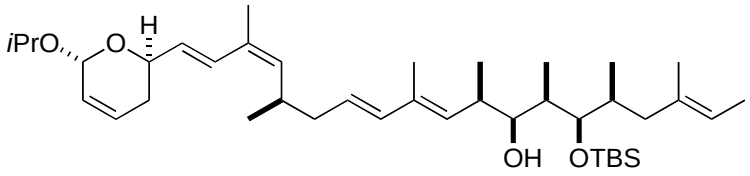
(4*S*,5*S*,6*R*)-4-((*R*,3*E*,5*E*)-6-iodo-4-methylhexa-3,5-dien-2-yl)-2,2,5,8,8,9,9-hepta-methyl-6-((*S*,*E*)-4-methylhex-4-en-2-yl)-3,7-dioxa-2,8-disiladecane (56)



To a cooled (–5 °C) suspension of $CrCl_2$ (11.0 mg, 0.088 mmol, 8.00 equiv) in dry THF (300 µL) was added a solution of α,β-unsaturated aldehyde **55** (5.1 mg, 0.011 mmol, 1.00 equiv) and CHI_3 (9.0 mg, 0.022 mmol, 2.00 equiv) in THF (200 µL). The dark brown mixture was covered with an aluminium foil and stirred at 0 °C for 2.5 hours. The mixture was diluted with Et_2O (2 mL) and water (1.5 mL) and the aqueous phase extracted with Et_2O (3x). The combined organic layers were washed with water (2x), saturated sodium thiosulfate solution (1x), dried ($MgSO_4$) and concentrated (bath T < 20 °C). Purification by chromatography on SiO_2 (pentane 100%) afforded vinyl iodide **56** (5.7 mg, 0.010 mmol, 88%, *d.r.* > 95:5) as a colorless oil. R_f = 0.16 (pentane 100%). Optical rotation $[\alpha]^{25.0}_D$ (*c* 0.105, $CHCl_3$) = +24.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.02 (d, *J* = 14.6 Hz, 1 H), 6.19 (d, *J* = 14.6 Hz, 1 H), 5.26 (d, *J* = 10.2 Hz, 1 H), 5.23 (dd, J_1 = 12.8 Hz, J_2 = 6.7 Hz, 1 H), 3.46-3.43 (m, 2 H), 2.70-2.61 (m, 1 H), 2.03 (d, *J* = 11.9 Hz, 1 H), 1.94 (t, *J* = 11.8 Hz, 1 H), 1.83-1.78 (m, 1 H), 1.76 (s, 3 H), 1.73-1.69 (m, 1 H), 1.62-1.61 (m, 6 H), 0.96 (d, *J* = 6.7 Hz, 3 H), 0.93 (s, 9 H), 0.85 (d, *J* = 6.8 Hz, 3 H), 0.71 (d, *J* = 6.6 Hz, 3 H), 0.16 (s, 9 H), 0.07 (s, 3 H), 0.04 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 150.0, 138.1, 134.6, 133.8, 120.7, 77.8, 73.9, 46.0, 40.5, 37.4, 34.7, 30.1, 26.7, 19.0, 17.3, 16.0, 13.8, 12.6, 12.4, 11.9, 1.4, –2.6, –2.8. HRMS-ESI calcd for $C_{27}H_{53}O_2ISi_2Na$: $[M + Na]^+$ 615.2527, found 615.2536. FTIR ν 2958*m*, 2928*m*, 2857*w*, 1461*w*, 1381*w*, 1253*w*, 1105*w*, 1032*w*, 890*w*, 836*w*, 772*w*, 631*s* cm^{-1}.

6.2.5. The Suzuki sp^3-sp^2 Cross Coupling and Synthesis Completion

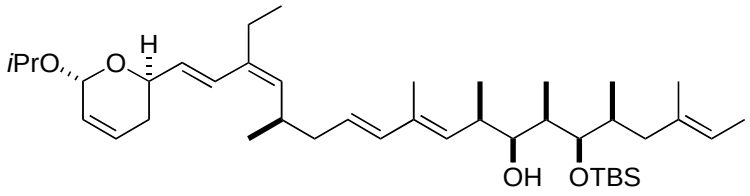
(2*E*,5*S*,6*R*,7*R*,8*S*,9*R*,10*E*,12*E*,15*R*,16*Z*,18*E*)-6-(*tert*-butyldimethylsilyloxy)-19-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-3,5,7,9,11,15,17-heptamethyl-nonadeca-2,10,12,16,18-pentaen-8-ol (58a)



To a solution of alkyl iodide **30** (29.0 mg, 0.077 mmol, 1.30 equiv) in Et_2O (850 µL) was added 9-MeO-9-BBN (1 M in hexane) (202 µL, 0.202 mmol, 3.42 equiv). The resulting solution was cooled to –78 °C and treated with *t*BuLi (1.5 M in pentane) (118 µL, 0.177 mmol, 3.00 equiv). After 5 minutes THF (850 µL) was added and the solution allowed to return to RT; stirring was continued for 1 hour. Separately in another flask vinyl iodide **52** (30.7 mg, 0.059 mmol, 1.00 equiv) was taken up in DMF (850 µL) to which Pd(dppf)Cl_2•CH_2Cl_2 (2.2 mg, 0.003 mmol, 0.05 equiv), $AsPh_3$ (2.8 mg, 0.009, 0.15 equiv), Cs_2CO_3 (77.0 mg, 0.236 mmol, 4.0 equiv) and H_2O (26 µL, 1.416 mmol, 24 equiv) were sequentially added. The alkyl boronate solution was transferred in the DMF solution and the resulting red-brown mixture stirred at RT overnight. The reaction was diluted with water and extracted with Et_2O (3x). The combined organic layers were washed with water (1x) and brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 92.5:7.5) afforded **58a** (30.2 mg, 0.047 mmol, 80%) as a pale yellow oil. R_f = 0.13 (pentane/Et_2O 9:1). Optical rotation $[\alpha]^{22.0}_D$ (*c* 0.34, $CHCl_3$) = +52.1°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.70 (d, *J* = 15.7 Hz, 1 H), 6.06-6.02 (m, 1 H), 6.02 (d, *J* = 15.5 Hz, 1 H), 5.77-5.70 (m, 2 H), 5.53 (dt, J_1 = 15.5 Hz, J_2 = 7.2 Hz, 1 H), 5.24-5.18 (m, 2 H), 5.15 (s, 1 H), 5.09 (d, *J* = 9.9 Hz, 1 H), 4.57-4.51 (m, 1 H), 4.05 (sept., *J* = 6.2 Hz, 1 H), 3.63-3.61 (m, 1 H), 3.39 (dd, J_1 = 8.86 Hz, J_2 = 2.57 Hz, 1 H), 2.75-2.68 (m, 1 H), 2.65-2.55 (m, 1 H), 2.20-2.02 (m, 5 H), 1.90-1.86 (m, 1 H), 1.84 (s, 3 H), 1.81-1.78 (m, 3 H), 1.75 (s, 3 H), 1.59 (d, *J* = 5.5 Hz, 3 H), 1.57 (s, 3 H), 1.27 (d, *J* = 6.2 Hz, 3 H), 1.21 (d, *J* = 6.1 Hz, 3 H), 1.05 (d, *J* = 6.5 Hz, 3 H), 0.99 (d, *J* = 6.6 Hz, 3 H), 0.94 (s, 9 H), 0.86 (d, *J* = 7.0 Hz, 3 H), 0.76 (d, *J* = 6.6 Hz, 3 H), 0.11 (s, 3 H), 0.10 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 137.9, 136.5, 134.8, 133.5, 133.4, 130.4, 129.6, 128.9, 128.6, 126.5, 126.3, 120.3, 93.7, 79.9, 79.1, 70.0, 67.4, 44.3, 41.2, 38.6, 37.3, 35.6, 32.5, 31.2, 26.6, 24.3,

22.5, 20.9, 20.8, 18.8, 18.0, 15.9, 14.9, 13.8, 13.2, 8.9, –2.8, –3.6. HRMS-ESI calcd for $C_{40}H_{70}O_4SiNa$: $[M + Na]^+$ 665.4941, found 665.4946. FTIR ν 3503*w*, 2962*m*, 2928*m*, 2859*m*, 1459*w*, 1381*w*, 1317*w*, 1253*w*, 1181*w*, 1099*m*, 1029*m*, 1001*m*, 964*m*, 836*w*, 774*w*, 718*w*, 678*w* cm^{-1}.

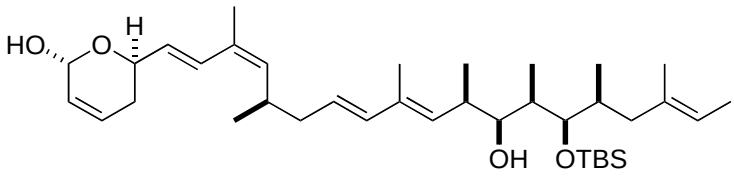
(2*E*,5*S*,6*R*,7*R*,8*S*,9*R*,10*E*,12*E*,15*R*,16*Z*,18*E*)-6-(*tert*-butyldimethylsilyloxy)-17-ethyl-19-((2*R*,6*R*)-6-isopropoxy-3,6-dihydro-2*H*-pyran-2-yl)-3,5,7,9,11,15-hexamethylnonadeca-2,10,12,16,18-pentaen-8-ol (60)



To a solution of alkyl iodide **31** (49.0 mg, 0.12 mmol, 1.30 equiv) in Et_2O (1.3 mL) was added 9-MeO-9-BBN (1M in hexane) (330 µL, 0.33 mmol, 3.42 equiv). The resulting solution was cooled to –78 °C and treated with *t*BuLi (1.5 M in pentane) (192 µL, 0.29 mmol, 3.00 equiv). After 5 minutes THF (1.3 mL) was added and the solution allowed to return to RT; stirring was continued for 1 hour. Separately in another flask vinyl iodide **52** (50.0 mg, 0.096 mmol, 1.00 equiv) was taken up in DMF (1.3 mL) to which $Pd(dppf)Cl_2•CH_2Cl_2$ (4.0 mg, 0.005 mmol, 0.05 equiv), $AsPh_3$ (4.4 mg, 0.014, 0.15 equiv), Cs_2CO_3 (125 mg, 0.384 mmol, 4.00 equiv) and H_2O (41 µL, 2.30 mmol, 24 equiv) were sequentially added. The alkyl boronate solution was transferred in the DMF solution and the resulting red-brown mixture stirred at RT for 20 hours. The reaction was diluted with water and extracted with Et_2O (3x). The combined organic layers were washed with water (1x) and brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 98:2) afforded **60** (30.0 mg, 0.046 mmol, 48%) as a pale yellow oil and a second fraction (41.4 mg) containing a mixture of product and a side compound that was directly used in the next step without further purifications. R_f = 0.71 (hexane/EtOAc 8:2). Optical rotation $[\alpha]^{22.2}_D$ (*c* 0.30, $CHCl_3$) = +50.5°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.59 (d, *J* = 15.7 Hz, 1 H), 6.03-6.00 (m, 1 H), 5.99 (d, *J* = 15.7 Hz, 1 H), 5.76-5.70 (m, 2 H), 5.53-5.46 (m, 1 H), 5.19-5.16 (m, 2 H), 5.13 (s, 1 H), 5.06 (d, *J* = 9.9 Hz, 1 H), 4.53-4.48 (m, 1 H), 4.02 (sept., *J* = 6.1 Hz, 1 H), 3.61-3.59 (m, 1 H), 3.37-3.33 (m, 1 H), 2.71-2.63 (m, 1 H), 2.61-2.53 (m, 1 H), 2.22-2.01 (m, 7 H), 1.89-1.76 (m, 4 H), 1.72 (s, 3 H), 1.59-1.55

(m, 6 H), 1.25 (d, *J* = 6.1 Hz, 3 H), 1.19 (d, *J* = 6.1 Hz, 3 H), 1.04 (m, 6 H), 0.97 (d, *J* = 6.4 Hz, 3 H), 0.91 (m, 9 H), 0.84 (d, *J* = 7.0 Hz, 3 H), 0.74 (d, *J* = 6.7 Hz, 3 H), 0.09 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 136.6, 136.2, 135.9, 134.9, 133.6, 133.4, 129.0, 128.9, 127.7, 126.6, 126.4, 120.4, 93.7, 80.0, 79.2, 70.1, 67.6, 44.4, 41.4, 38.7, 37.4, 35.7, 32.4, 31.3, 26.8, 26.6, 24.3, 22.6, 21.0, 18.8, 18.1, 16.0, 14.9, 13.9, 13.8, 13.3, 9.00, –2.9, –3.6. HRMS-ESI calcd for $C_{41}H_{72}O_4SiNa$: $[M + Na]^+$ 679.5098, found 679.5063. FTIR ν 3503*w*, 2963*m*, 2928*m*, 2858*w*, 1458*w*, 1381*w*, 1323*w*, 1254*w*, 1099*w*, 1030*m*, 1003*m*, 964*w*, 833*w*, 775*s* cm^{-1}.

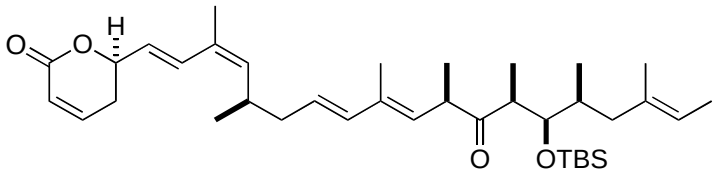
(2*R*,6*R*)-6-((1*E*,3*Z*,5*R*,7*E*,9*E*,11*R*,12*S*,13*R*,14*R*,15*S*,17*E*)-14-(*tert*-butyldimethylsilyloxy)-12-hydroxy-3,5,9,11,13,15,17-heptamethylnonadeca-1,3,7,9,17-pentaenyl)-5,6-dihydro-2*H*-pyran-2-ol (61)



To a solution of **58a** (6.8 mg, 0.011 mmol, 1.00 equiv) in a mixture acetone/water (3/1) (220 µL), PPTS (1.3 mg, 0.005 mmol, 0.5 equiv) was added and the resulting solution stirred at RT for 22 hours. The reaction was diluted with water, extracted with Et_2O (3x) and the combined organic layer dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 9:1 → 7:3) afforded alcohol **61** (6.3 mg, 0.010 mmol, 95%) as a pale yellow oil. R_f = 0.20 (pentane/Et_2O 7:3). Optical rotation $[\alpha]^{22.8}_D$ (*c* 0.10, $CHCl_3$) = +53.1°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.73 (d, *J* = 15.7 Hz, 1 H), 6.11-6.07 (m, 1 H), 6.02 (d, *J* = 15.5 Hz, 1 H), 5.85 (dd, J_1 = 10.1 Hz, J_2 = 0.8 Hz, 1 H), 5.73 (dd, J_1 = 15.7 Hz, J_2 = 6.5 Hz, 1 H), 5.53 (dt, J_1 = 15.5 Hz, J_2 = 7.3 Hz, 1 H), 5.48 (br. s, 1 H), 5.24 (d, *J* = 9.7 Hz, 1 H), 5.23-5.18 (m, 1 H), 5.10 (d, *J* = 9.9 Hz, 1 H), 4.61-4.56 (m, 1 H), 3.63-3.61 (m, 1 H), 3.42-3.39 (m, 1 H), 2.79-2.69 (m, 2 H), 2.66-2.56 (m, 1 H), 2.21-2.01 (m, 5 H), 1.91-1.86 (m, 1 H), 1.84 (s, 3 H), 1.82-1.78 (m, 3 H), 1.75 (s, 3 H), 1.59-1.57 (m, 6 H), 1.05 (d, *J* = 6.5 Hz, 3 H), 0.98 (d, *J* = 6.6 Hz, 3 H), 0.94 (s, 9 H), 0.87 (d, *J* = 7.0 Hz, 3 H), 0.76 (d, *J* = 6.6 Hz, 3 H), 0.11 (s, 3 H), 0.10 (s, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 138.2, 136.5, 134.9, 133.6, 133.4, 130.2, 129.2, 129.1, 126.4, 126.3, 120.3, 89.7, 79.9, 79.1, 67.8, 44.3, 41.2, 38.6, 37.3, 35.6, 32.5, 31.2, 26.6, 21.0, 20.8, 18.8, 17.9, 15.9, 14.9, 13.8, 13.3, 9.0, –2.8, –3.6.

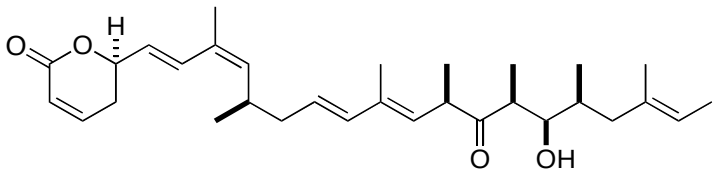
HRMS-ESI calcd for $C_{37}H_{64}O_4SiNa$: $[M + Na]^+$ 623.4472, found 623.4475. FTIR ν 3396*w*, 2959*m*, 2928*m*, 2859*w*, 1684*w*, 1457*w*, 1382*w*, 1253*w*, 1094*w*, 1033*w*, 964*w*, 835*w*, 772*w*, 680*m* cm^{-1}.

(*R*)-6-((1*E*,3*Z*,5*R*,7*E*,9*E*,11*R*,13*S*,14*R*,15*S*,17*E*)-14-(*tert*-butyldimethylsilyloxy)-3,5,9,11,13,15,17-heptamethyl-12-oxononadeca-1,3,7,9,17-pentaenyl)-5,6-dihydro-2*H*-pyran-2-one (62)



To a solution of alcohol **61** (3.2 mg, 0.005 mmol, 1.00 equiv) in CH_2Cl_2 (100 µL) was added DMP (5.6 mg, 0.013 mmol, 1.00 equiv) and the resulting mixture stirred at RT for 4 hours. The mixture was directly loaded over a pipette column of silica and eluted with pentane/Et_2O 9.5/0.5 → 7/3. The mixture of lactol-ketone and lactone-ketone was concentrated and directly treated with MnO_2 (7.0 mg, 0.080 mmol, 15.0 equiv) in CH_2Cl_2 (300 µL) at RT for 14 hours. The mixture was filtered over Celite, washed with CH_2Cl_2 and concentrated to afford α,β-unsaturated lactone **62** (1.5 mg, 0.003 mmol, 47%) as a pale yellow oil, which was directly used in the next step without further purification. R_f = 0.19 (pentane/Et_2O 7:3).

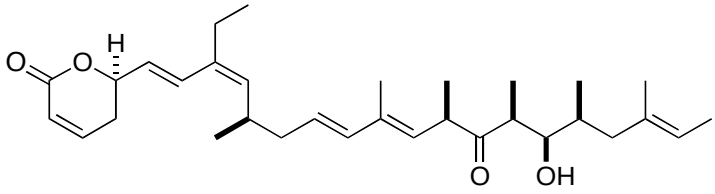
(*R*)-6-((1*E*,3*Z*,5*R*,7*E*,9*E*,11*R*,13*S*,14*R*,15*S*,17*E*)-14-hydroxy-3,5,9,11,13,15,17-heptamethyl-12-oxononadeca-1,3,7,9,17-pentaenyl)-5,6-dihydro-2*H*-pyran-2-one (anguinomycin C) (63)



In a 10 ml plastic vial under Ar, a solution of α,β-unsaturated lactone **62** (1.4 mg, 0.002 mmol, 1.00 equiv) in THF (300 µL) was cooled to 0 °C and treated dropwise with a solution of HF·pyridine (120 µL) and pyridine (60 µL) in THF (200 µL). After addition the

resulting pale yellow solution was allowed to return to RT and stirred for 4.5 days. The reaction mixture was diluted in Et_2O and transferred by canula in a saturated $NaHCO_3$ solution and extracted with Et_2O (3x). The combined organic layers were washed with saturated NH_4Cl (1x), dried ($MgSO_4$) and concentrated. The crude mixture was directly purified by HPLC to afford anguinomycin C (**63**) (0.9 mg, 0.0019 mmol, 82%) as a colorless oil. Optical rotation $[\alpha]^{23.1}_D$ (*c* 0.012, $CHCl_3$) = –116.7°. Optical rotation $[\alpha]^{22.5}_D$ (*c* 0.0064, MeOH) = –101.2°. ^{1}H-NMR (600 MHz, $CDCl_3$) δ 6.93 (dt, J_1 = 9.8 Hz, J_2 = 4.3 Hz, 1 H), 6.76 (d, J = 15.6 Hz, 1 H), 6.09 (td, J_1 = 9.7 Hz, J_2 = 1.8 Hz, 1 H), 6.04 (d, J = 15.6 Hz, 1 H), 5.75 (dd, J_1 = 15.6 Hz, J_2 = 6.9 Hz, 1 H), 5.61 (dt, J_1 = 15.5 Hz, J_2 = 7.4 Hz, 1 H), 5.30 (d, J = 9.8 Hz, 1 H), 5.22 (qd, J_1 = 6.6 Hz, J_2 = 1.1 Hz, 1 H), 5.15 (d, J = 10.1 Hz, 1 H), 5.01 (dt, J_1 = 7.3 Hz, J_2 = 7.1 Hz, 1 H), 3.69 (dq, J_1 = 10.1 Hz, J_2 = 6.7 Hz, 1 H), 3.59 (ddd, J_1 = 5.5 Hz, J_2 = 5.5 Hz, J_3 = 4.0 Hz, 1 H), 2.88 (qd, J_1 = 7.1 Hz, J_2 = 5.7 Hz, 1 H), 2.74-2.67 (m, 1 H), 2.51-2.49 (m, 2 H), 2.40 (d, J = 4.0 Hz, 1 H), 2.15-2.06 (m, 2 H), 2.02 (dd, J_1 = 13.0 Hz, J_2 = 6.1 Hz, 1 H), 1.85 (d, J = 1.1 Hz, 3 H), 1.84 (d, J = 1.1 Hz, 3 H), 1.74 (dd, J_1 = 13.0 Hz, J_2 = 8.8 Hz, 1 H), 1.69-1.64 (m, 1 H), 1.60 (dd, J_1 = 6.8 Hz, J_2 = 0.5 Hz, 3 H), 1.58 (s, 3 H), 1.17 (d, J = 7.1 Hz, 3 H), 1.16 (d, J = 6.6 Hz, 3 H), 0.99 (d, J = 6.7 Hz, 3 H), 0.80 (d, J = 6.6 Hz, 3 H). ^{13}C-NMR (150 MHz, $CDCl_3$) δ 215.4, 163.7, 144.3, 138.7, 135.8, 135.1, 133.6, 130.4, 129.1, 128.1, 127.3, 125.0, 121.3, 120.1, 78.3, 74.0, 46.1, 45.3, 43.7, 40.4, 32.8, 31.9, 29.7, 20.3, 20.0, 15.8, 14.9, 13.8, 13.0, 12.7, 11.8. HRMS-ESI calcd for $C_{31}H_{46}O_4Na$: $[M + Na]^+$ 505.3294, found 505.3281. FTIR ν 3440*m*, 2963*m*, 2927*m*, 2856*w*, 1709*m*, 1454*w*, 1381*w*, 1248*w*, 891*m* cm^{-1}. UV spectrum λ_{max} = 241 nm in MeOH. Analytical HPLC R_t = 32.35 minutes (C_{18}, 60%-100% MeOH in 50 minutes). Semi-preparative HPLC R_t = 38.82 minutes (C_{18}, 60%-80% MeOH in 50 minutes).

(*R*)-6-((1*E*,3*Z*,5*R*,7*E*,9*E*,11*R*,13*S*,14*R*,15*S*,17*E*)-3-ethyl-14-hydroxy-5,9,11,13,15,-17- hexamethyl-12-oxononadeca-1,3,7,9,17-pentaenyl)-5,6-dihydro-2*H*-pyran-2-one (anguinomycin D) (66)



To a solution of **60** (27.0 mg, 0.041 mmol, 1.00 equiv) in a mixture acetone/water (5/1) (830 µL) was added PPTS (6.0 mg, 0.024 mmol, 0.4 equiv) and the resulting solution stirred at RT for 43 hours. The reaction was transferred in a saturated $NaHCO_3$ solution, extracted with Et_2O (3x) and the combined organic layer washed with brine (1x), dried ($MgSO_4$) and concentrated. Purification by chromatography on SiO_2 (pentane/Et_2O 9:1 → 1:1) afforded the lactol (23.0 mg, 0.037 mmol, 91%).

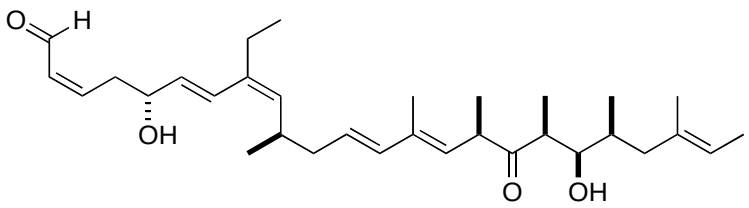
To a solution of lactol (1.30 mg, 0.002 mmol, 1.00 equiv) in CH_2Cl_2 (0.5 mL), 4Å MS (50 mg), PCC (3.00 mg, 0.013 mmol, 6.00 equiv) and glacial acetic acid (12 µL, 0.21 mmol, 100 equiv) were sequentially added and the resulting mixture stirred at RT for 1.5 hours. The mixture was directly loaded over a column of silicagel and eluted with hexane/AcOEt 8.5/1.5 → 1/1 to afford the ketolactone intermediate which was directly used in the last step.

In a 10 ml plastic tube a solution of the previous obtained ketolactone in THF (0.5 mL) was cooled to 0 °C. Pyridine (100 µL) and HF·pyridine (100 µL) were sequentially added and the tube sealed. After 5 minutes the solution was allowed to return to RT and stirred for 4.5 days. The solution was cooled to 0 °C and silicagel (100 mg) was added. After 5 minutes, the mixture was loaded on a pipette column of silicagel and eluted with hexane/EtOAc 8:2 → 1:1 affording anguinomycin D (**66**) (0.62 mg, 0.0013 mmol, 60%) as a colorless oil. An analytical sample of anguinomycin D was purified by HPLC. R_f = 0.17 (hexane/EtOAc 6:4). Optical rotation $[\alpha]^{22.7}_D$ (*c* 0.014, MeOH) = –112.0°. ^{1}H-NMR (800 MHz, $CDCl_3$) δ 6.90 (dddd, J_1 = 9.7 Hz, J_2 = 4.9 Hz, J_3 = 3.6 Hz, J_4 = 0.8 Hz, 1 H), 6.63 (d, J = 15.8 Hz, 1 H), 6.06 (dt, J_1 = 9.8 Hz, J_2 = 1.8 Hz, 1 H), 6.01 (d, J = 15.6 Hz, 1 H), 5.76 (dd, J_1 = 15.7 Hz, J_2 = 6.9 Hz, 1 H), 5.58 (dt, J_1 = 15.5 Hz, J_2 = 7.3 Hz, 1 H), 5.25 (d, J = 9.8 Hz, 1 H), 5.19 (qd, J_1 = 6.8 Hz, J_2 = 1.2 Hz, 1 H), 5.11 (d, J = 10.1 Hz, 1 H), 4.99-4.96 (m, 1 H) or 4.98 (dtd, J_1 = 6.9 Hz, J_2 = 7.6 Hz, J_3 = 0.8 Hz, 1 H), 3.66 (dq, J_1 = 10.1 Hz, J_2 = 6.7 Hz, 1 H), 3.55 (t, J = 5.6 Hz, 1 H), 2.87 (dt, J_1 = 5.7 Hz, J_2 = 7.1 Hz,

1 H), 2.68-2.64 (m, 1 H), 2.48-2.46 (m, 2 H), 2.22-2.15 (m, 2 H), 2.08 (t, J = 7.0 Hz, 2 H), 1.98 (dd, J_1 = 13.1 Hz, J_2 = 6.2 Hz, 1 H), 1.82 (d, J = 1.2 Hz, 3 H), 1.70 (dd, J_1 = 13.1 Hz, J_2 = 8.6 Hz, 1 H), 1.65-1.61 (m, 1 H), 1.57 (dd, J_1 = 6.7 Hz, J_2 = 0.8 Hz, 3 H), 1.55 (s, 3 H), 1.14 (d, J = 7.3 Hz, 3 H), 1.13 (d, J = 7.1 Hz, 3 H), 1.04 (t, J = 7.5 Hz, 3 H), 0.96 (d, J = 6.6 Hz, 3 H), 0.77 (d, J = 6.6 Hz, 3 H). ^{13}C-NMR (150 MHz, $CDCl_3$) δ 215.8, 164.1, 144.7, 137.3, 136.2, 135.4, 135.3, 134.0, 130.0, 128.4, 127.7, 124.8, 121.7, 120.5, 79.9, 74.4, 46.5, 45.6, 44.1, 40.8, 33.2, 32.2, 30.1, 26.4, 20.8, 16.3, 15.3, 14.2, 13.5, 13.4, 13.1, 12.2. HRMS-ESI calcd for $C_{32}H_{48}O_4Na$: $[M + Na]^+$ 519.3450; found 519.3429. UV spectrum λ_{max} = 242 nm in MeOH. Analytical HPLC R_t = 32.87 minutes (C_{18}, 60% → 100% MeOH in 50 minutes).

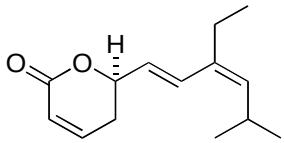
Following the same three last steps procedure using the mixed fraction of product **60**, in addition to anguinomycin D, the following compounds were isolated:

2*Z*,5*R*,6*E*,8*Z*,10*R*,12*E*,14*E*,16*R*,18*S*,19*R*,20*S*,22*E*)-8-ethyl-5,19-dihydroxy-10,14,-16,18,20,22-hexamethyl-17-oxotetracosa-2,6,8,12,14,22-hexaenal (67)



R_f = 0.11 (hexane/EtOAc 7:3). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.56 (d, J = 7.5 Hz, 1 H), 6.91 (dt, J_1 = 15.8 Hz, J_2 = 7.5 Hz, 1 H), 6.56 (d, J = 15.8 Hz, 1 H), 6.23 (ddt, J_1 = 15.8 Hz, J_2 = 7.9 Hz, J_3 = 1.3 Hz, 1 H), 6.04 (d, J = 15.3 Hz, 1 H), 5.73 (dd, J_1 = 15.3 Hz, J_2 = 6.6 Hz, 1 H), 5.64-5.59 (m, 1 H), 5.24-5.20 (m, 2 H), 5.16 (d, J = 10.1 Hz, 1 H), 4.41 (q, J = 6.1 Hz, 1 H), 3.70-3.65 (m, 1 H), 3.60 (t, J = 5.3 Hz, 1 H), 2.91-2.86 (m, 1 H), 2.70-2.66 (m, 1 H), 2.65-2.63 (m, 1 H), 2.22-2.18 (m, 1 H), 2.11 (t, J = 6.6 Hz, 1 H), 2.01 (dd, J_1 = 13.2 Hz, J_2 = 6.1 Hz, 1 H), 1.84 (d, J = 1.3 Hz, 3 H), 1.76-1.73 (m, 1 H), 1.68-1.64 (m, 1 H), 1.61 (d, J = 6.6 Hz, 3 H), 1.58-1.57 (m, 3 H), 1.17 (d, J = 7.0 Hz, 3 H), 1.16 (d, J = 6.6 Hz, 3 H), 1.07 (t, J = 7.5 Hz, 3 H), 1.00 (d, J = 7.0 Hz, 3 H), 0.91 (t, J = 7.0 Hz, 3 H), 0.80 (d, J = 6.6 Hz, 3 H). HRMS-ESI calcd for $C_{32}H_{50}O_4Na$: $[M + Na]^+$ 521.3607; found 521.3607. Analytical HPLC R_t = 32.37 minutes (C_{18}, 60% → 100% MeOH in 50 minutes). λ_{max} = 239 nm.

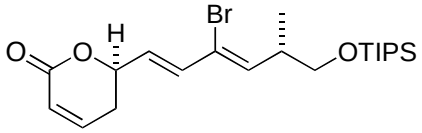
(*R*)-6-((1*E*,3*Z*)-3-ethyl-5-methylhexa-1,3-dienyl)-5,6-dihydro-2*H*-pyran-2-one (68)



R_f = 0.44 (hexane/EtOAc 7:3). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.93 (dt, J_1 = 9.6 Hz, J_2 = 4.1 Hz, 1 H), 6.70 (d, J = 15.8 Hz, 1 H), 6.09 (dt, J_1 = 9.8 Hz, J_2 = 1.9 Hz, 1 H), 5.78 (dd, J_1 = 15.8 Hz, J_2 = 6.9 Hz, 1 H), 5.29 (d, J = 9.6 Hz, 1 H), 5.05-4.99 (m, 1 H), 2.81-2.72 (m, 1 H), 2.52-2.48 (m, 2 H), 2.19 (q, J = 7.4 Hz, 2 H), 1.07 (t, J = 7.4 Hz, 3 H), 1.00 (d, J = 1.5 Hz, 3 H), 0.98 (d, J = 1.4 Hz, 3 H). HRMS-ESI calcd for $C_{14}H_{20}O_2$: $[M]^+$ 221.1542; found 221.1548. Analytical HPLC R_t = 38.55 minutes (C_{18}, 30% → 80% MeOH in 50 minutes), λ_{max} = 239 nm.

6.2.6. Synthesis of Anguinomycin Derivatives

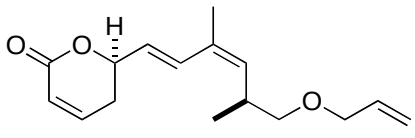
(*R*)-6-((*S*,1*E*,3*Z*)-3-bromo-5-methyl-6-(triisopropylsilyloxy)hexa-1,3-dienyl)-5,6-dihydropyran-2-one (69)



To a solution of **24** (15 mg, 0.03 mmol, 1.00 equiv) in a mixture of acetone/water (3:1) (0.62 mL) was added PPTS (7.7 mg, 0.03 mmol, 1.00 equiv). The solution was stirred for 2 hours, quenched with water and extracted with Et_2O (3 x 15 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated to afford the crude lactol. To a suspension of MnO_2 (161 mg, 1.86 mmol, 60.0 equiv) in a mixture of CH_2Cl_2/pyridine (1:0.025) (0.61 mL) was added the crude lactol. The reaction was stirred for 1.5 hours, diluted with CH_2Cl_2, filtered through Celite and washed with water (1x). The aqueous layer was extracted with CH_2Cl_2 (3x) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (CH_2Cl_2/cyclohexane 8:2) affording α,β-unsaturated lactone **69** (4.30 mg, 0.01 mmol, 31%) as a colorless oil. R_f = 0.11 (cyclohexane/AcOEt 9:1). Optical rotation $[\alpha]^{28.6}_D$ (*c* 0.355, $CHCl_3$) = +46.1°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ, 6.90 (ddd, J_1 = 9.8 Hz, J_2 = 5.2 Hz, J_3 = 3.3 Hz, 1 H), 6.39 (ddd, J_1 = 14.8 Hz, J_2 = 1.3 Hz, J_3 = 0.6 Hz, 1 H), 6.10 (d, J = 5.6 Hz, 1 H), 6.09-6.04 (m, 1 H), 5.97 (d, J = 8.9 Hz, 1 H), 5.07 (dtd, J_1 = 9.9 Hz, J_2 = 5.5 Hz, J_3 = 1.2 Hz, 1 H), 3.63 (ddd, J_1 = 21.5 Hz, J_2 = 9.5 Hz, J_3 = 5.7 Hz, 2 H), 3.00-2.87 (m, 1 H), 2.51-

2.45 (m, 2 H), 1.09-1.05 (m, 24 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 163.5, 144.4, 139.5, 132.0, 129.3, 123.0, 121.6, 76.7, 66.7, 39.6, 30.0, 18.1, 16.2, 12.1. HRMS-EI calcd for $C_{21}H_{35}BrO_3Si$: $[M–C_3H_7]^+$ 399.0986; found 399.0984. FTIR ν 3020*w*, 2944*w*, 1721*w*, 1215*m*, 1112*w*, 751*s* cm^{-1}.

(*R*)-6-((*S*,1*E*,3*Z*)-6-(allyloxy)-3,5-dimethylhexa-1,3-dienyl)-5,6-dihydropyran-2-one (70)



To a cooled (–20 °C) suspension of NaH (1.00 mg, 0.04 mmol, 1.00 equiv) in DMF (100 µL) was added a solution of alcohol **28** (10.6 mg , 0.04 mmol, 1.00 equiv) in DMF (100 µL). The mixture was stirred at –20 °C for 25 minutes, treated with allyl bromide (3.80 µL, 0.04 mmol, 1.10 equiv), warmed to RT and stirred for 8 hours. The reaction was quenched with water and extracted with Et_2O (3 x 30 mL) and the combined organic layers were washed with brine (3 x 50 mL), dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (CH_2Cl_2 100%) to give allylated product (7.40 mg, 0.02 mmol, 60%) as a colorless oil. R_f = 0.31 (cyclohexane/AcOEt 9:1). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.68 (d, J = 15.7 Hz, 1 H), 6.04-5.98 (m, 1 H), 5.90 (ddt, J_3 = 17.1 Hz, J_2 = 10.4 Hz, J_1 = 5.5 Hz, 1 H), 5.76-5.69 (m, 2 H), 5.26 (dq, J_1 = 1.7 Hz, J_2 = 17.3 Hz, 1 H), 5.21-5.12 (m, 3 H), 4.55-4.48 (m, 1 H), 4.02 (sept., J = 6.2 Hz, 1 H), 3.96 (ddd, J_1 = 1.3 Hz, J_2 = 2.6 Hz, J_3 = 5.5 Hz, 2 H), 3.27 (ddd, J_1 = 6.8 Hz, J_2 = 9.2 Hz, J_3 = 16.6 Hz, 2 H), 3.00-2.86 (m, 1 H), 2.20-2.00 (m, 2 H), 1.83 (d, J = 1.2 Hz, 3 H), 1.25 (d, J = 6.2 Hz, 3 H), 1.18 (d, J = 6.2 Hz, 3 H), 1.01 (d, J = 6.7 Hz, 3 H). HRMS-EI calcd for $C_{19}H_{30}O_3$: $[M]^+$ 306.2190; found 306.2191.

To a solution of allylated product (7.40 mg , 0.02 mmol, 1.00 equiv) in a mixture of acetone/water (3/1) (0.48 mL) was added PPTS (6.10 mg, 0.02 mmol, 1.00 equiv). The reaction was stirred at RT for 2 hours, quenched with water and extracted with Et_2O (3 x 15 mL). The combined organic layers were dried ($MgSO_4$) filtered and concentrated to afford the crude lactol. The residue was dissolved in a mixture of CH_2Cl_2/pyridine (1/0.025) (0.48 mL), treated with MnO_2 (125 mg, 1.44 mmol, 60.0 equiv) and the suspension was stirred for 3 hours. The mixture was diluted with CH_2Cl_2 and filtered through Celite, washed with water and extracted with CH_2Cl_2.

The organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was purified by chromatography on SiO_2 (CH_2Cl_2/AcOEt 97:3) to give α,β-unsaturated lactone **70** (1.70 mg, 0.01 mmol, 29%) as a colorless oil. R_f = 0.34 (CH_2Cl_2/AcOEt 97:3). Optical rotation $[\alpha]^{28.3}_D$ (*c* 0.160, $CHCl_3$) = +53.4°. ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.90 (dt, J_1 = 9.7 Hz, J_2 = 4.2 Hz, 1 H), 6.75 (d, J = 15.8 Hz, 1 H), 6.06 (dt, J_1 = 9.7 Hz, J_2 = 1.8 Hz, 1 H), 5.96-5.83 (m, 1 H), 5.76 (dd, J_1 = 15.7 Hz, J_2 = 6.7 Hz, 1 H), 5.30-5.22 (m, 2 H), 5.19-5.14 (m, 1 H), 5.01 (q, J = 7.2 Hz, 1 H), 3.96 (dd, J_1 = 5.5 Hz, J_2 = 0.9 Hz, 2 H), 3.32-3.21 (m, 2 H), 2.91 (ddd, J_1 = 9.5 Hz, J_2 = 6.7 Hz, J_3 = 13.3 Hz, 1 H), 2.48 (ddd, J_1 = 6.3 Hz, J_2 = 4.7 Hz, J_3 = 2.0 Hz, 2 H), 1.83 (s, 3 H), 1.00 (d, J = 6.7 Hz, 3 H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 163.7, 144.4, 135.8, 134.7, 130.8, 130.3, 125.7, 121.5, 116.6, 78.5, 74.9, 71.9, 32.5, 30.0, 20.5, 18.0. HRMS-EI calcd for $C_{16}H_{22}O_3$: $[M]^+$ 262.1564; found 262.1564.

6.2.7. Biological Evaluations

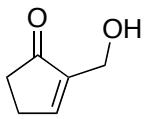
Cell culture techniques, antibodies and indirect immunofluorescence.

HeLa cells were cultured at 37 °C in Dulbecco's modified eagle's medium (DMEM), supplemented with 10% fetal calf serum, 100 units/ml penicillin and 100 μg/ml streptomycin. For studying the inhibition of CRM1-mediated nuclear export, HeLa cells were grown on coverslips for 24 h to about 75% confluency. Cells were then incubated with different concentrations of LMB (LC laboratories, USA) or anguinomycins C or D for 90 min at 37 °C. For detection of Rio2, cells were fixed in 4% paraformaldehyde for 15 min and permeabilized for 5 minutes in 1 x detergent (0.1% Triton-X, 0.02% SDS in 1xPBS). Incubation with α-Rio2 antibody (polyclonal antibody, raised against recombinant full-length human Rio2 in rabbit, affinity-purified) and fluorescently labeled secondary antibody (a-rabbit, Alexa 488-labeled, Invitrogen). Pictures were acquired using a Leica TCS NT1 laser-scanning confocal microscope.

6.3. Synthetic Studies on Sporolides

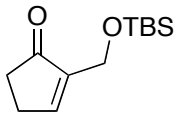
6.3.1. Synthesis of the Vinyl Triflate Fragment

2-(hydroxymethyl)cyclopent-2-enone (72)



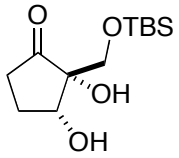
To a solution of 2-cyclopenten-1-one (**71**) (1.02 mL, 12.2 mmol, 1.00 equiv) in $CHCl_3$ (15 mL) and MeOH (10 mL) was added formaldehyde (37% solution in H_2O) (1.5 mL, 20.2 mmol, 1.5 equiv) at RT. A solution of dimethylphenylphosphine (100 μL, 0.72 mmol, 0.06 equiv, 6 mol%) in $CHCl_3$ (10 mL) was added to the reaction and the resulting mixture was stirred at RT for 1 h. The mixture was directly dried by addition of $MgSO_4$ and directly purified by chromatography on SiO_2 (hexane/EtOAc 1:3) afforded alcohol **72** (1.32 g, 11.8 mmol, 97%) as a white crystalline solid. R_f = 0.13 (EtOAc/hexane 3:1). M.p. = 71 °C. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.57-7.53 (m, 1 H), 4.40 (d, J = 5.6 Hz, 2 H), 2.67 (dt, J_1 = 4.3 Hz, J_2 = 2.2 Hz, 2 H), 2.49-2.47 (m, 2 H), 2.39 (t, J = 5.6 Hz, 1 H).

2-((*tert*-butyldimethylsilyloxy)methyl)cyclopent-2-enone (73)



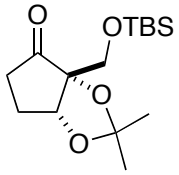
To a cooled (–10°C) solution of alcohol **72** (7.50 g, 66.9 mmol, 1.00 equiv) in CH_2Cl_2 (65 mL) were sequentially added imidazole (10.0 g, 147 mmol, 2.20 equiv) and TBSCl (14.1 g, 93.6 mmol, 1.40 equiv). After 5 minutes, the solution was allowed to return to RT and stirred overnight. The solution washed with a diluted citric acid solution (pH ≈ 4) (50 mL) and brine (70 mL). The combined organic layer was dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 9:1) afforded enone **73** (15.1 g, 66.5 mmol, 99%) as white solid. R_f = 0.27 (EtOAc/hexane 1:9). M.p. = 32 °C. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.57-7.54 (m, 1 H), 4.40-4.38 (m, 2 H), 2.63 (dt, J_1 = 4.4 Hz, J_2 = 2.6 Hz, 2 H), 2.48-2.45 (m, 2 H), 0.94 (s, 9 H), 0.11 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 209.0, 158.4, 146.8, 58.7, 35.8, 27.2, 26.3, 18.7, –5.0. FTIR ν 2955*w*, 2930*w*, 2887*w*, 2857*w*, 1702*m*, 1643*w*, 1463*w*, 1396*w*, 1254*m*, 1194*w*, 1116*m*, 996*m*, 835*s*, 776*s*, 665*w* cm^{-1}.

(2*R*,3*R*)-2-((*tert*-butyldimethylsilyloxy)methyl)-2,3-dihydroxycyclopentanone (74)



A solution of $(DHQD)_2PHAL$ (203 mg, 0.26 mmol, 0.03 equiv, 3.0 mol%), NMO (1.55 g, 13.3 mmol, 1.50 equiv), methanesulfonamide (1.22 g, 13.3 mmol, 1.50 equiv) and $K_2OsO_2(OH)_4$ (48 mg, 0.13 mmol, 0.015 equiv, 1.5 mol%) in a mixture of water/acetone (1:3) (100 mL) was stirred at RT for 15 minutes. A solution of enone **73** (2.00 g, 8.83 mmol, 1.00 equiv) in a mixture water/acetone (1:3) (24 mL) was slowly added at RT during a period of 2.5 hours (~ 0.16 mL/min) under vigorous stirring. After addition, the reaction was stirred for additional 30 minutes before the addition of $NaHSO_3$ (4.0 g). The resulting mixture was stirred for 1 hour; then a solution of saturated $NaHCO_3$ (60 mL) was added and the mixture stirred for 5 minutes. The precipitate was dissolved by addition of water and the aqueous phase was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic layer was washed with a diluted citric acid solution (0.5 M) (250 mL) and brine (300 mL), dried (Na_2SO_4), filtered and concentrated. Purification by chromatography on SiO_2 (CH_2Cl_2/EtOAc 1:0 → 10:3) afforded the dihydroxylated compound **74** (2.12 g, 8.14 mmol, 92%) as colorless viscous liquid. R_f = 0.58 (CH_2Cl_2/EtOAc 10:3). Optical rotation $[\alpha]^{22.9}_D$ (*c* 0.99, $CHCl_3$) = –28.9°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 4.23 (dd, J_1 = 4.0 Hz, J_2 = 2.4 Hz, 1 H), 3.68 (s, 2 H), 3.28 (s, 1 H), 2.74 (s, 1 H), 2.53-2.44 (m, 1 H), 2.35-2.18 (m, 2 H), 2.12-2.05 (m, 1 H), 0.91 (s, 9 H), 0.06 (d, *J* = 4.0 Hz, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 217.3, 81.4, 72.7, 65.5, 33.0, 26.2, 26.0, 18.6, –5.2. FTIR ν 3430*m*, 2954*m*, 2931*m*, 2896*w*, 2858*m*, 1749*s*, 1466*w*, 1403*w*, 1362*w*, 1256*s*, 1109*s*, 1081*s*, 961*w*, 838*s*, 779*s*, 670*s* cm^{-1}.

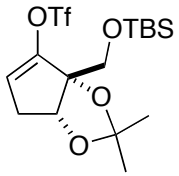
(3a*R*,6a*R*)-3a-((*tert*-butyldimethylsilyloxy)methyl)-2,2-dimethyldihydro-3a*H*-cyclopenta[*d*][1,3]dioxol-4(5*H*)-one (76)



To a cooled (0 °C) solution of dihydroxylated compound **74** (2.20 mg, 8.50 mmol, 1.00 equiv) in a mixture of anhydrous DMF/acetone (3:1) (53 mL) were added 2,2-dimetoxypropane (15.7 mL, 126 mmol, 15.0 equiv) and PPTS (151 mg, 0.60 mmol, 0.07 equiv, 7 mol%) and the resulting solution was stirred at RT for 16 hours. Brine (50 mL) and water (50 mL) were added to the reaction mixture and the aqueous phase was extracted with Et_2O (3 x 100 mL). The combined organic layers were washed with brine (1x), dried

($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (CH_2Cl_2 100%) afforded **76** (2.12 g, 7.06 mmol, 84%) as a colorless liquid. R_f = 0.42 (CH_2Cl_2). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 4.74 (d, *J* = 4.0 Hz, 1 H), 3.97 (d, *J* = 9.6 Hz, 1 H), 3.70 (d, *J* = 9.6 Hz, 1 H), 2.63-2.53 (m, 1 H), 2.26-2.16 (m, 2 H), 2.08-1.98 (m, 1 H), 1.37 (s, 3 H), 1.33 (s, 3 H), 0.83 (s, 9 H), 0.03 (s, 3 H), 0.00 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 217.7, 111.6, 85.7, 82.1, 63.6, 34.6, 27.5, 26.1, 24.1, 18.6, –5.2. HRMS-ESI calcd for $C_{15}H_{28}O_4SiNa$: $[M + Na]^+$ 323.1655, found 323.1665. FTIR ν 2987*w*, 2932*m*, 2859*m*, 1757*s*, 1466*w*, 1374*m*, 1252*m*, 1213*m*, 1183*w*, 1090*s*, 1002*m*, 838*s*, 779*s*, 743*w*, 668*s* cm^{-1}.

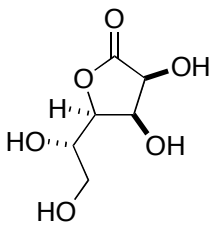
(3a*R*,6a*R*)-3a-((*tert*-butyldimethylsilyloxy)methyl)-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta[*d*][1,3]dioxol-4-yl trifluoromethanesulfonate (77)



A cooled (0 °C) solution of DIPA (0.82 mL, 5.80 mmol, 1.20 equiv) in anhydrous THF (10 mL) was treated with *n*BuLi (1.6 M in hexane) (3.32 mL, 5.31 mmol, 1.10 equiv). The resulting solution was stirred at 0°C for 30 min, then cooled to –10°C and a solution of **76** (1.45 g, 4.83 mmol, 1.00 equiv) in anhydrous THF (12 mL) was slowly added. The yellow solution was stirred at –10°C for 20 minutes before the addition of a solution of $PhNTf_2$ (2.41 g, 6.76 mmol, 1.40 equiv) in anhydrous THF (15 mL). The reaction was stirred for 5 min at –10°C, then allowed to return to RT and stirred for 15 hours. The mixture was quenched by addition of water (50 mL) and extracted with Et_2O (3 x 50 mL). The combined organic layers were washed with brine (150 mL), dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/CH_2Cl_2 8:2) afforded vinyl triflate **77** (1.78 g, 4.12 mmol, 85%) as a colorless liquid. R_f = 0.13 (CH_2Cl_2/ hexane 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.76 (s, 1 H), 4.64 (d, *J* = 5.4 Hz, 1 H), 3.87 (d, *J* = 10.2 Hz, 1 H), 3.69 (d, *J* = 10.2 Hz, 1 H), 2.59 (ddd, J_1 = 17.3 Hz, J_2 = 5.1 Hz, J_3 = 1.9 Hz, 1 H), 2.48 (dd, J_1 = 17.3 Hz, J_2 = 2.9 Hz, 1 H), 1.46 (s, 3 H), 1.40 (s, 3 H), 0.88 (s, 9 H), 0.08 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 147.9, 117.3, 112.2, 91.1, 80.2, 63.4, 33.8, 27.7, 26.1, 18.5, –5.3. HRMS-ESI calcd for $C_{16}H_{27}O_6F_3SSiNa$: $[M + Na]^+$ 455.1147, found 455.1154. FTIR ν 2989*w*, 2954*w*, 2933*m*, 2861*w*, 1659*w*, 1470*w*, 1425*s*, 1374*w*, 1211*s*, 1142*s*, 1093*s*, 1060*m*, 990*w*,930*w*, 840*s*, 779*m*, 663*m* cm^{-1}.

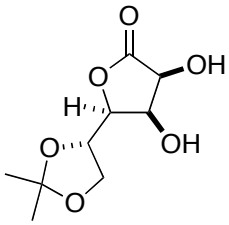
6.3.2. Synthesis of the Enediyne Fragment

L-gulono-1,4-lactone (83)



Argon was bubbled through a solution of L-ascorbic acid (**82**) (10.0 g, 56.8 mmol, 1.00 equiv) in water (80 mL) for 30 minutes. Pd/C (10%) was added to the solution and the flask put under H_2 atmosphere. The reaction was heated to 65 °C and vigorously stirred for 72 hours. The reaction was allowed to return to RT and filtered over cotton. The aqueous solution was freezed and lyophilized. The crude was triturated in a mixture EtOAc/MeOH (1:1), filtered and dried under high vacuum affording L-gulono-1,4-lactone (**83**) (36.8 g, 0.21 mol, 77 %) as white crystals. R_f = 0.61 (CH_2Cl_2/MeOH 7:4). Optical rotation $[\alpha]^{24.5}_D$ (*c* 0.494, H_2O) = +57.8°. ^{1}H-NMR (400 MHz, DMSO-d_6) δ 5.80 (d, *J* = 7.5 Hz, 1 H), 5.34 (d, *J* = 3.8 Hz, 1 H), 4.98 (d, *J* = 5.3 Hz, 1 H), 4.66 (t, *J* = 5.7 Hz, 1 H), 4.43 (dd, J_1 = 7.4 Hz, J_2 = 4.5 Hz, 1 H), 4.22 (dd, J_1 = 8.1 Hz, J_2 = 2.4 Hz, 1 H), 4.17 (dd, J_1 = 7.0 Hz, J_2 = 4.0 Hz, 1 H), 3.73 (dq, J_1 = 8.4 Hz, J_2 = 4.7 Hz, 1 H), 3.54-3.43 (m, 2 H). ^{13}C-NMR (100 MHz, DMSO-d_6) δ 176.3, 80.9, 70.8, 70.1, 69.5, 62.0. FTIR ν 3367*s*, 3228*m*, 2913*w*, 1778*s*, 1426*m*, 1349*w*, 1254*m*, 1181*m*, 1132*s*, 1080*m*, 1048*s*, 993*s*, 958*s*, 907*m*, 860*m*, 791*s* cm^{-1}.

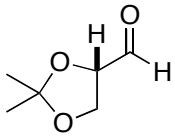
5,6-*O*-isopropylidene-L-gulono-1,4-lactone (84)



To a cooled (0 °C) solution of L-gulono-1,4-lactone (**83**) (7.00 g, 39.3 mmol, 1.00 equiv) in DMF (72 mL) was added PTSA (67.7 mg, 0.39 mmol, 0.01 equiv). After 10 minutes, 2-methoxypropene (4.72 mL, 51.1 mmol, 1.30 equiv) was added dropwise to the colorless solution. The reaction was allowed to return to RT and stirred for 24 hours. A second portion of 2-methoxypropene (2.18 mL, 23.6 mmol, 0.60 equiv) was added and the solution stirred overnight. The reaction was quenched by addition of $NaHCO_3$ (0.5 g), filtered over cotton, washed with a small amount of DMF and concentrated. Addition of toluene to the orange oil and evaporation was repeated twice to give 5,6-*O*-isopropylidene-L-gulono-1,4-lactone (**84**) (8.50 g, 38.9 mmol, 99%) as a pale orange solid, which was used without further purification in the next step. R_f = 0.43 (CH_2Cl_2/MeOH 9:1). Optical rotation

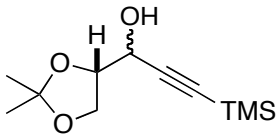
$[\alpha]^{24.2}_{D}$ (*c* 0.46, MeOH) = +38.3°. ^{1}H-NMR (400 MHz, DMSO-d_6) δ 5.89 (d, *J* = 7.4 Hz, 1 H), 5.45 (d, *J* = 4.1 Hz, 1 H), 4.42 (dd, J_1 = 7.3 Hz, J_2 = 4.7 Hz), 4.30-4.24 (m, 2 H), 4.21 (dd, J_1 = 7.2 Hz, J_2 = 4.4 Hz, 1 H), 4.07 (dd, J_1 = 8.6 Hz, J_2 = 6.2 Hz, 1 H), 3.76 (dd, J_1 = 8.6 Hz, J_2 = 6.1 Hz, 1 H), 1.35 (s, 3 H), 1.29 (s, 3 H). FTIR ν 3515*w*, 3455*m*, 2997*w*, 2932*w*, 2871*w*, 1758*m*, 1408*w*, 1375*m*, 1313*w*, 1297*m*, 1197*s*, 11473*s*, 1074*s*, 1042*m*, 982*s*, 933*m*, 892*w*, 841*m*, 779*m*, 684*m*, 628*m* cm^{-1}.

L-(*S*)-glyceraldehyde acetonide (85)



A suspension of sodium metaperiodate (8.83 g, 41.2 mmol, 2.00 equiv) in water (21 mL) was cooled to 4 °C with an ice bath. A solution of NaOH (3 M) (13.8 mL, 41.2 mmol, 2.00 equiv) was added dropwise at such a rate that the temperature did not exceed 7 °C, in order to set the pH between 4 and 6. The pH was finally adjusted to 6-7. The cooling bath was removed and 5,6-*O*-isopropylidene-L-gulono-1,4-lactone (**84**) (4.50 g, 20.6 mmol, 1.00 equiv) was added in one portion. Temperature was maintained between 20 °C and 30 °C, occasional use of an ice bath was therefore required and the pH was adjusted to 5 using HCl (1 M). After 90 minutes the suspension was saturated with NaCl (2 g), filtered through a Büchner funnel and the white solid was washed with brine. The pH of the combined aqueous layer was adjusted to 6-7 with a solution of Na_2CO_3 (15 %) and extracted with EtOAc (8x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. The crude aldehyde **85** was directly used in the next step without further purification. R_f = 0.49 (CH_2Cl_2/MeOH 9:1). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.60 (d, *J* = 1.3 Hz, 1 H), 4.52 (ddd, J_1 = 7.1 Hz, J_2 = 4.6 Hz, J_3 = 1.2 Hz, 1 H), 4.08-4.02 (m, 2 H), 1.37 (s, 3 H), 1.33 (s, 3 H).

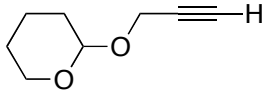
(*S*)-1-(2,2-dimethyl-1,3-dioxolan-4-yl)-3-(trimethylsilyl)prop-2-yn-1-ol (86)



To a cooled (0 °C) solution of HMDS (5.3 mL, 25.0 mmol, 1.21 equiv) in anhydrous THF (23 mL) was added dropwise *n*BuLi (1.6 M in hexane) (14.2 mL, 22.7 mmol, 1.10 equiv) and the resulting colorless solution was stirred for 30 minutes at 0 °C. The LHMDS solution was transferred to a cooled (–78 °C) solution of ethynyltrimethylsilane (3.5

mL, 24.7 mmol, 1.20 equiv) in THF (100 mL). After 30 minutes, a solution of aldehyde **85**, obtained by sodium metaperiodate cleavage of 5,6-*O*-isopropylidene-L-gulono-1,4-lactone (**84**) (8.83 g, 41.2 mmol, 2.00 equiv) and directly diluted in THF (31 mL), was added dropwise to the reaction mixture. After 1 hour at –78 °C, the reaction was quenched by addition of saturated NH_4Cl (15 mL) and the mixture volume reduced to a volume of ca. 40 mL. The mixture was extracted with EtOAc (3x) and the combined organic layers washed with water (1x) and brine (1x), dried (Na_2SO_4), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/CH_2Cl_2 8:2) afforded alcohol **86** (1.32 g, 5.76 mmol, 28 % (over 2 steps)) as a mixture of diastereomers (*d.r.* = 1.3:1). R_f = 0.40 (hexane/EtOAc 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$). *Major diastereomer* δ 4.25 (dt, J_1 = 6.5 Hz, J_2 = 3.7 Hz, 1 H), 4.17 (dd, J_1 = 12.2 Hz, J_2 = 6.7 Hz, 1 H), 4.05 (dd, J_1 = 14.8 Hz, J_2 = 7.9 Hz, 1 H), 2.34 (d, J = 4.2 Hz, 1 H), 1.45 (s, 3 H), 1.38 (s, 3 H), 0.17 (s, 9 H). *Minor diastereomer* δ 4.50 (t, J = 4.2 Hz, 1 H), 4.32 (dd, J_1 = 7.2 Hz, J_2 = 4.2 Hz, 1 H), 3.90 (dd, J_1 = 8.6 Hz, J_2 = 5.3 Hz, 1 H), 2.23 (d, J = 4.7 Hz, 1 H), 1.56 (s, 3 H), 1.47 (s, 3H), 0.17 (s, 9 H). ^{13}C-NMR (100 MHz, $CDCl_3$). *Major diastereomer* δ 110.5, 102.3, 91.5, 78.8, 66.2, 65.1, 26.8, 25.3, –0.3. *Minor diastereomer* δ 110.1, 102.2, 91.5, 77.8, 64.8, 62.8, 26.3, 25.3, –0.3. HRMS-ESI calcd for $C_{11}H_{20}O_3NaSi$: $[M+Na]^+$ 251.1079; found 251.1085. FTIR ν 3428*m*, 2987*m*, 2961*m*, 2900*m*, 2175*w*, 1374*m*, 1251*s*, 1215*m*, 1156*s*, 1068*s*, 843*s*, 761*w* cm^{-1}.

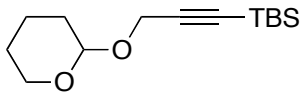
2-(prop-2-ynyloxy)tetrahydro-2*H*-pyran (89)



In a three-necked flask equipped with a thermometer, a dropping funnel and a reflux condenser, dihydropyran (35.4 mL, 0.39 mol, 1.07 equiv) was heated to 60 °C. PTSA (cat.) was added, followed by dropwise addition of propargyl alcohol (**88**) (20.8 mL, 0.36 mol, 1.00 equiv) over a period of 45 minutes. During the addition the solution turned from dark to light yellow and the temperature was maintained at 60 °C. After addition the resulting mixture was stirred at 60-65 °C for 3 hours. The reaction was quenched by addition of $NaHCO_3$ (0.5 g) and the mixture stirred for an additional hour. The mixture was filtered and purified by distillation under reduced pressure (4.2 mbar, 47 °C) using a Vigreux column affording alkyne **89** (49.4 g, 0.35 mol, 98%) as a colorless oil. R_f =

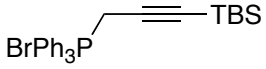
0.71 (hexane/EtOAc 7:3). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 4.82 (t, J = 3.3 Hz, 1 H), 4.30 (dd, J_1 = 15.7 Hz, J_2 = 2.4 Hz, 1 H), 4.23 (dd, J_1 = 15.7 Hz, J_2 = 2.4 Hz, 1 H), 3.87-3.81 (m, 1 H), 3.55-3.52 (m, 1 H), 2.41 (t, J = 2.4 Hz, 1 H), 1.88-1.54 (m, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 96.7, 79.7, 73.9, 61.9, 53.9, 30.1, 25.2, 18.9.

tert-butyldimethyl(3-(tetrahydro-2*H*-pyran-2-yloxy)prop-1-ynyl)silane (90)



A cooled (–18 °C) solution of alkyne **89** (5.00 g, 35.7 mmol, 1.00 equiv) in THF (70 mL) was treated with dropwise addition of *n*BuLi (1.6 M in hexane) (23.4 mL, 37.5 mmol, 1.05 equiv); the solution turned to orange. After addition, the solution was stirred for 15 minutes, before addition of a TBSCl (5.64 g, 37.5 mmol, 1.05 equiv) solution in THF (10 mL). The resulting mixture was stirred for 45 minutes, then quenched by addition of water (5 mL). A citric acid solution (pH = 4) (10 mL) was added and the mixture extracted with EtOAc (3x). The combined organic layers were washed with saturated $NaHCO_3$ (1x), brine (1x), dried ($MgSO_4$), filtered and concentrated affording **90** (8.99 g, 35.3 mmol, 99 %) as an orange liquid, which was used in the next step without further purification. R_f = 0.60 (hexane/EtOAc 9:1). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 4.85 (t, J = 3.3 Hz, 1 H), 4.28 (br s, 2 H), 3.84 (ddd, J_1 = 11.2 Hz, J_2 = 8.9 Hz, J_3 = 2.7 Hz), 3.55-3.51 (m, 1 H), 1.85-1.54 (m, 6 H), 0.93 (s, 9 H), 0.11 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 102.1, 96.5, 89.0, 61.9, 54.6, 30.2, 26.0, 25.3, 19.0, 16.4, –4.7.

(3-*tert*-butyldimethylsilyl)propargyltriphenylphosphonium bromide (92)

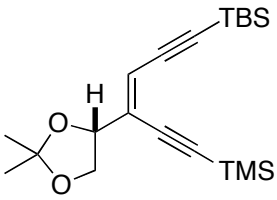


To a cooled (–15 °C) solution of triphenylphosphine (4.64 g, 17.7 mmol, 1.25 equiv) in CH_2Cl_2 (36 mL) was added dropwise bromine (0.91 mL, 17.7 mmol, 1.25 equiv). After 30 minutes at –15 °C, a solution of **90** (3.61 g, 14.2 mmol, 1.00 equiv) in CH_2Cl_2 (6 mL) was slowly added and the resulting mixture allowed to return to RT and stirred for 7 hours. The reaction mixture was diluted with water and the aqueous layer extracted with CH_2Cl_2 (2x). The combined organic layers were washed with brine (1x), dried ($MgSO_4$), filtered and concentrated. The crude was triturated and the precipitate abundantly washed with pentane. The filtrate was concentrated to afford protected propargyl bromide **91**,

which was directly used in the next step without further purification. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 3.92 (s, 2 H), 0.94 (s, 9 H), 0.11 (s, 6 H).

The crude **91** was diluted in toluene (150 mL) and triphenylphosphine (31.6 g, 0.12 mol, 1.30 equiv) was added. The mixture was covered with an aluminium foil and stirred at RT for 42 hours. The precipitate was filtered and thoroughly washed with toluene affording the phosphonium bromide salt **92** (35.4 g, 71.4 mmol, 77%) as a beige solid. M.p. = 210.6 °C. ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.96-7.93 (m, 3 H), 7.85-7.79 (m, 12 H), 5.13 (d, *J* = 16.6 Hz, 2 H), 0.68 (s, 9 H,), –0.04 (s, 6 H).

(*R*,*E*)-*tert*-butyl(4-(2,2-dimethyl-1,3-dioxolan-4-yl)-6-(trimethylsilyl)hexa-3-en-1,5-diynyl)dimethylsilane (93)

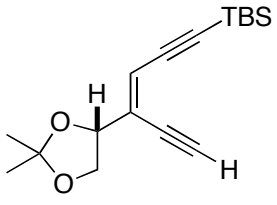


To a solution of alcohol **86** (1.32 g, 5.76 mmol, 1.00 equiv) in dry CH_2Cl_2 (30 mL) were sequentially added 3 Å molecular sieves (3.9 g), PDC (3.25 g, 8.65 mmol, 1.50 equiv) and glacial acetic acid (0.56 mL, 9.80 mmol, 1.70 equiv). The resulting dark mixture was stirred at RT for 1 hour, then Celite (2.7 g) was added and the mixture stirred for additional 30 minutes. The suspension was filtered through a plug of Celite and the cake was washed with CH_2Cl_2 (30 mL). Heptane (100 mL) was added and the solution reduced to ca. 25 mL. A mixture pentane/Et_2O (2/1) (100 mL) was added and the mixture filtered through a plug of $MgSO_4$. The colorless filtrate was washed with water (2x) and saturated $NaHCO_3$ (2x). The organic layer was dried ($MgSO_4$) and filtered. The filtered was reduced to a volume of ca. 20 mL, anhydrous THF (30 mL) was added and the solution reduced again to a volume of ca. 20 mL. The operation was repeated two times and the final yellow concentrate diluted in THF (30 mL). The solution was stored under argon atmosphere at –20 °C and directly used in the next step without further purification.

To a cooled (–78 °C) suspension of phosphonium bromide salt **92** (3.29 g, 6.63 mmol, 1.15 equiv) in THF (60 mL), KHMDS (0.5 M in toluene) (12.7 mL, 6.34 mmol, 1.10 equiv) was slowly added over a period of 15 minutes. The resulting orange suspension was kept for 15 minutes at –78 °C, then warmed up to –40 °C and stirred for 2 hours. The solution was heated to –15 °C and after 5 minutes, the crude propargylic ketone

87 solution was added *via* canula over a period of 30 minutes. After 1 hour, the reaction was quenched by addition of saturated NH_4Cl and extracted with pentane (3x). The combined organic layers were washed with water (2x) and brine (2x), dried (Na_2SO_4), filtered and concentrated. A careful purification by chromatography on SiO_2 (hexane/EtOAc 99:1) allowed the isolation of a pure fraction of (*E*)-bis-silyl enedyine **93** (814 mg, 2.24 mmol, 39%), (*Z*)-bis-silyl enedyine (*Z*)-**93** (225 mg, 0.62 mmol, 11%) and a mixed (*E*)/(*Z*) fraction (708 mg, 1.95 mmol, 34%) from an initial mixture (*E*)/(*Z*) (2.7:1). R_f (propargylic ketone **87**) = 0.58; R_f ((*E*)-bis-silyl enedyine **93**) = 0.76; R_f ((*Z*)-bis-silyl enedyine (*Z*)-**93**) = 0.83 (hexane/EtOAc 8:2). Optical rotation $[\alpha]^{22.2}_D$ (*c* 0.507, $CHCl_3$) = –36.0°. ^{1}H-NMR (400 MHz, $CDCl_3$) (*E*)-bis-silyl enedyine δ 6.06 (d, J = 1.1 Hz, 1 H), 4.53 (dt, J_1 = 6.7 Hz, J_2 = 0.9 Hz, 1 H), 4.17 (dd, J_1 = 8.3 Hz, J_2 = 6.6 Hz, 1 H), 3.91 (dd, J_1 = 8.3 Hz, J_2 = 7.2 Hz, 1 H), 1.45 (s, 3 H), 1.40 (s, 3 H), 0.97 (s, 9 H), 0.20 (s, 9 H), 0.14 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 133.8, 116.2, 110.3, 104.4, 102.4, 101.3, 100.3, 77.4, 68.9, 26.3, 26.2, 25.9, 16.6, –0.2, –4.6. DEPT-135 NMR (100 MHz, $CDCl_3$) *CH_3 & CH* δ 116.7, 77.8, 26.7, 26.6, 26.3, 0.2, –4.2; *CH_2* δ 69.4. FTIR ν 2956*m*, 2929*m*, 2858*w*, 2147*w*, 1468*w*, 1373*w*, 1251*m*, 1218*w*, 1069*m*, 841*m*, 775*w* cm^{-1}. ^{1}H-NMR (400 MHz, $CDCl_3$) (*Z*)-bis-silyl enedyine δ 6.00 (s, 1 H), 5.16 (dd, J_1 = 7.7 Hz, J_2 = 6.6 Hz, 1 H), 4.16 (dd, J_1 = 8.2 Hz, J_2 = 6.4 Hz, 1 H), 3.85 (t, J = 8.1 Hz, 1 H), 1.48 (s, 3 H), 1.42 (s, 3 H), 0.94 (s, 9 H), 0.19 (s, 9 H), 0.13 (s, 6 H).

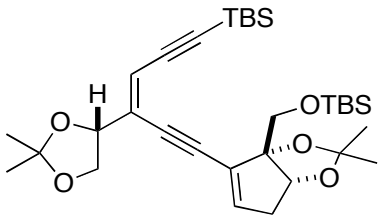
(*R*,*E*)-*tert*-butyl(4-(2,2-dimethyl-1,3-dioxolan-4-yl)hexa-3-en-1,5-diynyl)dimethylsilane (94)



To a cooled (0 °C) solution of (*E*)-bis-silyl enedyine **93** (52.6 mg, 0.15 mmol, 1.00 equiv) in MeOH (0.7 mL) was added in one portion anhydrous K_2CO_3 (20.1 mg, 0.15 mmol, 1.00 equiv). After 45 minutes, the reaction was quenched with water and the aqueous layer extracted with pentane (4x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated affording **94** (40.8 mg, 0.09 mmol, 97%), which was directly used in next reaction without further purification. R_f = 0.46 (hexane/EtOAc 92:8). Optical rotation $[\alpha]^{22.2}_D$ (*c* 0.524, $CHCl_3$) = –41.2°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.14 (s, 1 H), 4.57 (dt, J_1 = 6.7 Hz, J_2 = 1.1 Hz, 1 H), 4.19 (dd,

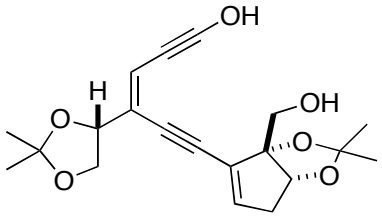
J_1 = 8.4 Hz, J_2 = 6.7 Hz, 1 H), 3.90 (dd, J_1 = 8.4 Hz, J_2 = 6.8 Hz, 1 H), 3.37 (s, 1 H), 1.46 (s, 3 H), 1.40 (s, 3 H), 0.97 (s, 9 H), 0.14 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 133.1, 117.3, 110.5, 102.1, 101.7, 86.0, 79.5, 77.1, 68.9, 26.3, 26.1, 25.8, 16.6, –4.7. FTIR ν 3294*w*, 2988*w*, 2955*m*, 2930*m*, 2887*w*, 2858*m*, 2361*w*, 2139*w*, 1468*w*, 1374*w*, 1252*m*, 1219*w*, 1154*w*, 1097*w*, 1068*m*, 940*w*, 840*w*, 630*s* cm^{-1}.

tert-butyl(((3a*R*,6a*R*)-4-((*E*)-6-(_tert_-butyldimethylsilyl)-3-((*R*)-2,2-dimethyl-1,3-dioxolan-4-yl)hexa-3-en-1,5-diynyl)-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta-[*d*][1,3]dioxol-3a-yl)methoxy)dimethylsilane (95)



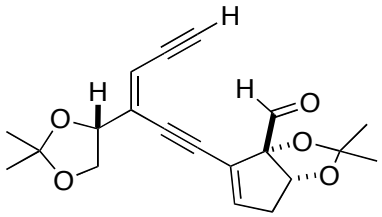
To a solution of **94** (70 mg, 0.24 mmol, 1.00 equiv) and vinyl triflate **77** (105 mg, 0.24 mmol, 1.00 equiv) in DMF (1.2 mL) were sequentially added DIPEA (160 μL, 0.97 mmol, 4.00 equiv), 2,6-lutidine (57 μL, 0.49 mmol, 2.00 equiv), CuI (14 mg, 0.07 mmol, 0.30 equiv, 30 mol %) and $Pd(PPh_3)_4$ (14 mg, 0.01 mmol, 0.05 equiv, 5 mol %). The resulting dark red suspension was stirred at RT for 75 minutes, then quenched by addition of saturated NH_4Cl, extracted with Et_2O (3x). The combined organic layers were washed with water (1x) and brine (2x), dried (Na_2SO_4), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/CH_2Cl_2 1:1 → 0:1) afforded **95** (90.4 mg, 0.16 mmol, 65%) as a brown oil. R_f = 0.17 (hexane/CH_2Cl_2 2:8). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.13 (br s, 1 H), 6.08 (s, 1 H), 4.64 (d, J = 4.6 Hz, 1 H), 4.57 (dt, J_1 = 6.7 Hz, J_2 = 0.8 Hz, 1 H), 4.20 (dd, J_1 = 8.3 Hz, J_2 = 6.6 Hz, 1 H), 3.91 (dd, J_1 = 8.3 Hz, J_2 = 7.1 Hz, 1 H), 3.77 (q, J = 10.2 Hz, 2 H), 2.62 (ddd, J_1 = 18.9 Hz, J_2 = 4.6 Hz, J_3 = 2.3 Hz, 1 H), 2.54 (dd, J_1 = 18.9 Hz, J_2 = 2.8 Hz, 1 H), 1.46 (s, 3 H), 1.40 (s, 3 H), 1.38 (s, 3 H), 1.37 (s, 3 H), 0.96 (s, 9 H), 0.86 (s, 9 H), 0.14 (s, 6 H), 0.05 (s, 3 H), 0.03 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 139.1, 133.9, 127.2, 114.9, 110.6, 110.4, 102.7, 101.2, 95.9, 93.3, 88.0, 80.8, 77.4, 69.1, 64.2, 38.4, 27.8, 27.2, 26.3, 26.2, 25.9, 25.8, 18.2, 16.7, –4.6, –5.4, –5.5. DEPT-135 NMR (100 MHz, $CDCl_3$) *CH_3 & CH* δ 139.1, 114.9, 80.8, 77.4, 27.8, 27.2, 26.3, 26.2, 25.9, 25.8, –4.6, –5.4, –5.5; *CH_2* δ 69.1, 64.2, 38.4. FTIR ν 2987*w*, 2953*m*, 2931*m*, 2958*m*, 1468*w*, 1371*w*, 1251*m*, 1217*m*, 1155*w*, 1093*m*, 1007*w*, 940*w*, 838*m*, 777*m*, 683*m*, 631*w* cm^{-1}.

((3a*R*,6a*R*)-4-((*E*)-3-((*R*)-2,2-dimethyl-1,3-dioxolan-4-yl)hexa-3-en-1,5-diynyl)-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta[*d*][1,3]dioxol-3a-yl)methanol (96)



To a cooled (–20 °C) solution of enedyine **95** (45.3 mg, 0.08 mmol, 1.00 equiv) in THF (1.5 mL) was added TBAF (1 M in THF) (174 μL, 0.17 mmol, 2.20 equiv). After 5 minutes at –20 °C, the brown solution was allowed to reach 0 °C and stirred for 1 hour and 45 minutes. The reaction was quenched by addition of water and extracted with Et_2O (3x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 7:3) afforded alcohol **96** (23.4 mg, 0.07 mmol, 86%) as a clear brown oil. R_f = 0.61 (CH_2Cl_2/MeOH 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.22 (br t, *J* = 1.9 Hz, 1 H), 6.06 (d, *J* = 1.2 Hz, 1 H), 4.67 (d, *J* = 4.6 Hz, 1 H), 4.60 (t, *J* = 6.6 Hz, 1 H), 4.22 (dd, J_1 = 8.3 Hz, J_2 = 6.7 Hz, 1 H), 3.95-3.90 (m, 1 H), 3.91 (dd, J_1 = 8.4 Hz, J_2 = 6.8 Hz, 1 H), 3.55 (br d, *J* = 11.3 Hz, 1 H), 3.34 (d, *J* = 2.3 Hz, 1 H), 2.69 (ddd, J_1 = 19.4 Hz, J_2 = 4.6 Hz, J_3 = 2.4 Hz, 1 H), 2.61 (dd, J_1 = 19.3 Hz, J_2 = 2.8 Hz, 1 H), 1.47 (s, 3 H), 1.42 (s, 9 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 140.3, 135.4, 126.8, 114.9, 111.4, 111.0, 96.6, 92.8, 88.5, 85.2, 81.2, 80.0, 77.5, 69.5, 62.8, 38.4, 28.3, 27.8, 26.7, 26.3. FTIR ν 3473*w*, 3283*w*, 2987*w*, 2932*w*, 2877*w*, 1457*w*, 1374*m*, 1245*m*, 1216*m*, 1155*m*, 1063*s*, 990*w*, 961*w*, 923*w*, 898*w*, 856*m*, 794*w*, 763*w*, 631*s* cm^{-1}.

(3a*S*,6a*R*)-4-((*E*)-3-((*R*)-2,2-dimethyl-1,3-dioxolan-4-yl)hexa-3-en-1,5-diynyl)-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta[*d*][1,3]dioxole-3a-carbaldehyde (97)

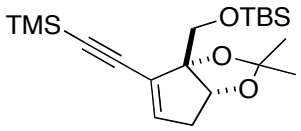


To a cooled (–78 °C) solution of oxalyl chloride (11.5 μL, 0.14 mmol, 2.00 equiv) in CH_2Cl_2 (0.2 mL) was added dropwise a solution of DMSO (24 μL, 0.34 mmol, 5.00 equiv) in CH_2Cl_2 (0.2 mL). The resulting clear solution was stirred for 20 minutes at –78 °C, then a solution of alcohol **96** (23.4 mg, 0.07 mmol, 1.00 equiv) in CH_2Cl_2 (0.1 mL) was added dropwise. After 30 minutes at –78 °C a solution of DIPEA (46.5 μL, 0.27 mmol, 4.00 equiv) in CH_2Cl_2 (0.1 mL) was slowly added and the resulting solution stirred at –78 °C for 20 minutes,

then allowed to return to 0 °C and stirred for 30 minutes. The reaction was quenched by addition of water and extracted with CH_2Cl_2 (3x). The combined organic layers were washed with saturated NH_4Cl, dried (Na_2SO_4), filtered and concentrated to afford aldehyde **97** (20.0 mg, 0.06 mmol, 86%) as a brown oil. The crude was directly used in the next reaction without further purification. R_f = 0.25 (hexane/EtOAc 7:3). Optical rotation $[\alpha]^{23.3}_D$ (*c* 0.425, $CHCl_3$) = +21.5°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.85 (s, 1 H), 6.30 (t, *J* = 2.3 Hz, 1 H), 6.04 (br s, 1 H), 4.74 (d, *J* = 5.5 Hz, 1 H), 4.56 (t, *J* = 6.4 Hz, 1 H), 4.18 (dd, J_1 = 8.4 Hz, J_2 = 6.7 Hz, 1 H), 3.86 (dd, J_1 = 8.3 Hz, J_2 = 6.9 Hz, 1 H), 3.32 (d, *J* = 2.3 Hz, 1 H), 2.78 (ddd, J_1 = 19.5 Hz, J_2 = 5.5 Hz, J_3 = 2.4 Hz, 1 H), 2.64 (dd, J_1 = 19.5 Hz, J_2 = 2.8 Hz, 1 H), 1.48 (s, 3 H), 1.44 (s, 3 H), 1.40 (s, 3 H), 1.39 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 199.5, 141.5, 134.7, 124.0, 114.9, 113.0, 110.5, 99.1, 91.7, 87.9, 85.0, 80.5, 80.2, 77.0, 68.9, 38.4, 27.3, 26.4, 26.2, 25.8. DEPT-135 NMR (100 MHz, $CDCl_3$) *CH_3 & CH* δ 200.0, 142.0, 115.4, 85.5, 80.6, 77.4, 27.7, 26.8, 26.7, 26.2; *CH_2* δ 69.4, 38.8. FTIR ν 3278*w*, 2987*w*, 2924*m*, 2854*w*, 1732*m*, 1459*w*, 1375*m*, 1248*m*, 1214*s*, 1154*m*, 1066*s*, 984*w*, 926*w*, 860*m*, 736*w*, 647*w* cm^{-1}.

6.3.3. Toward the 9-Membered Ring from the Allene

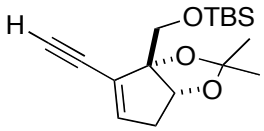
tert-butyl(((3a*R*,6a*R*)-2,2-dimethyl-4-((trimethylsilyl)ethynyl)-6,6a-dihydro-3a*H*-cyclopenta[*d*][1,3]dioxol-3a-yl)methoxy)dimethylsilane (100)



To a solution of vinyl triflate **77** (1.15 g, 2.66 mmol, 1.00 equiv) in DMF (14 mL) were sequentially added trimethylsilylacetylene (416 µL, 2.92 mmol, 1.10 equiv), 2,6-lutidine (620 µL, 5.32 mmol, 2.00 equiv), DIPEA (1.74 mL, 10.6 mmol, 4.00 equiv), CuI (152 mg, 0.80 mmol, 0.30 equiv, 30 mol%) and $Pd(PPh_3)_4$ (154 mg, 0.13 mmol, 0.05 equiv, 5 mol%). The resulting dark-brown solution was stirred at RT for 1.5 hours, then quenched by addition of saturated NH_4Cl and extracted with Et_2O (3x). The combined organic layers were washed with water (2x) and brine (1x), dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 98:2 → 95:5) afforded **100** (1.01 g, 2.65 mmol, quant.) as a colorless oil. R_f = 0.54 (hexane/CH_2Cl_2 2:8). Optical rotation $[\alpha]^{21.8}_D$ (*c* 0.20, $CHCl_3$) = +39.2°.

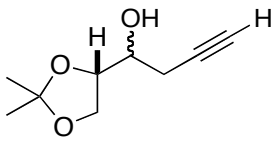
^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.13 (s, 1 H), 4.65 (d, *J* = 4.8 Hz, 1 H), 3.87 (d, *J* = 9.9 Hz, 1 H), 3.76 (d, *J* = 9.9 Hz, 1 H), 2.59 (ddd, J_1 = 18.9 Hz, J_2 = 4.8 Hz, J_3 = 2.2 Hz, 1 H), 2.51 (dd, J_1 = 8.9 Hz, J_2 = 2.6 Hz, 1 H), 1.44 (s, 3 H), 1.41 (s, 3 H), 0.88 (s, 9 H), 0.21 (s, 9 H), 0.07 (d, *J* = 2.2 Hz, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 139.2, 127.9, 111.1, 99.9, 97.6, 96.1, 81.4, 64.6, 38.7, 28.2, 27.5, 26.2, 18.6, 0.42, –5.01, –5.20. HRMS-ESI calcd for $C_{20}H_{36}O_3Si_2Na$: $[M+Na]^+$ 403.2101; found 403.2100. FTIR ν 2943*w*, 2932*w*, 2859*w*, 2149*w*, 1468*w*, 1370*w*, 1250*m*, 1212*w*, 1084*m*, 992*w*, 839*s*, 778*m*, 665*m* cm^{-1}.

tert-butyl(((3a*R*,6a*R*)-4-ethynyl-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta[*d*]-[1,3]dioxol-3a-yl)methoxy)dimethylsilane (101)



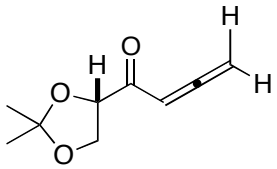
To a cooled (0 °C) solution of **100** (1.00 g, 2.63 mmol, 1.00 equiv) in MeOH (24 mL) was added K_2CO_3 (654 mg, 4.73 mmol, 1.80 equiv) and the resulting mixture stirred at 0 °C for 3.5 hours. The reaction was quenched by addition of water and extracted with Et_2O (3x). The combined organic layers were washed with brine (1x), dried ($MgSO_4$), filtered and concentrated to afford alkyne **101** (810 mg, 2.63 mmol, quant.) as a colorless oil. R_f = 0.41 (hexane/CH_2Cl_2 2:8). Optical rotation $[\alpha]^{21.5}_D$ (*c* 0.845, $CHCl_3$) = +39.6°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.19 (s, 1 H), 4.67 (d, *J* = 4.8 Hz, 1 H), 3.86 (d, *J* = 10.2 Hz, 1 H), 3.79 (d, *J* = 10.2 Hz, 1 H), 3.03 (s, 1 H), 2.62 (dd, J_1 = 18.9 Hz, J_2 = 3.8 Hz, 1 H), 2.54 (dd, J_1 = 18.6 Hz, J_2 = 2.9 Hz, 1 H), 1.45 (s, 3 H), 1.41 (s, 3 H), 0.89 (s, 9 H), 0.08 (s, 3 H), 0.07 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 139.6, 126.9, 111.2, 96.1, 81.3, 80.4, 78.6, 64.4, 38.6, 28.2, 27.4, 26.2, 18.6, –5.1, –5.2. HRMS-ESI calcd for $C_{17}H_{28}O_3SiNa$: $[M+Na]^+$ 331.1705; found 331.1713. FTIR ν 3314*w*, 2931*m*, 2859*w*, 1468*w*, 1370*m*, 1250*m*, 1214*m*, 1087*s*, 991*w*, 838*s*, 778*s*, 664*s*, 629*s* cm^{-1}.

(*S*)-1-(2,2-dimethyl-1,3-dioxolan-4-yl)but-3-yn-1-ol (102)



To a mixture of Mg(turning) (1.65 g, 68 mmol, 2.00 equiv) in Et_2O (55 mL) were sequentially added $HgCl_2$ (cat.) and I_2 (cat.) and the mixture refluxed for 5 minutes, before the slow addition of propargyl bromide (80 % solution in toluene) (10.1 g, 68.0 mmol, 2.00 equiv). The resulting mixture was refluxed for 1 hour, then cooled to 0 °C and transferred by canula over a period of 30 minutes on a cooled (–20 °C) solution of aldehyde **85**, obtained by sodium metaperiodate cleavage of 5,6-*O*-isopropylidene-L-gulono-1,4-lactone (**84**) (7.42 g, 34.0 mmol, 1.00 equiv) and directly diluted in Et_2O (35 mL). During the transfer, additional Et_2O (20 mL) was added to help stirring. After addition the mixture was allowed to return to RT and stirred for 2 hours. The reaction was quenched by addition of saturated NH_4Cl and extracted with Et_2O (4x); the combined organic layers were dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 9:1 → 7:3) afforded alcohol **102** (2.47 g, 14.5 mmol, 43% over 2 steps) as a mixture of diastereoisomers (*d.r.* = 1.00:0.60), which was directly used in the next step. R_f = 0.57 (hexane/EtOAc 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$). *Mixture of diastereoisomers* δ 4.20 (dd, J_1 = 6.4 Hz, J_2 = 5.1 Hz, 0.6 H), 4.10-4.02 (m, 1.6 H), 3.99-3.94 (m, 1 H), 3.85 (dd, J_1 = 8.3 Hz, J_2 = 6.7 Hz, 0.6 H), 3.77-3.75 (m, 1 H), 3.70 (t, J = 5.8 Hz, 0.6 H), 2.57-2.39 (m, 3.2 H), 2.09 (t, J = 2.6 Hz, 1 H), 2.07 (t, J = 2.6 Hz, 0.6 H), 1.46 (s, 1.8 H), 1.43 (s, 3 H), 1.39 (s, 1.8 H), 1.37 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$). *Major diastereomer* δ 109.8, 80.3, 77.7, 71.7, 70.5, 66.3, 27.1, 25.6, 24.0. *Minor diastereomer* δ 110.0, 80.4, 77.8, 71.2, 70.7, 66.4, 26.9, 25.6, 24.3.

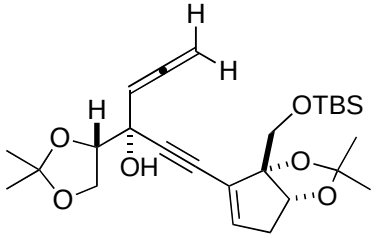
1-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-buta-2,3-dien-1-one (104)



To a cooled (0 °C) solution of alcohol **102** (177 mg, 1.10 mmol, 1.00 equiv) in CH_2Cl_2 (8 mL) was added DMP (583 mg, 1.40 mmol, 1.30 equiv). After 5 minutes the resulting mixture was allowed to return to RT and stirred for 4 hours; then diluted in a mixture hexane/EtOAc (9.5:0.5), directly loaded on a column of silicagel. Purification by chromatography on SiO_2 (hexane/EtOAc 9.5:0.5 → 9:1) afforded allene **104** (160 mg, 0.95 mmol, 87%) as a pale yellow oil. R_f = 0.52

(hexane/EtOAc 7:3). Optical rotation $[\alpha]^{21.3}_D$ (*c* 1.08, $CHCl_3$) = –71.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.12 (t, J = 6.7 Hz, 1 H), 5.34 (dd, J_1 = 15.4 Hz, J_2 = 6.7 Hz, 1 H), 5.29 (dd, J_1 = 13.4 Hz, J_2 = 4.5, 1 H), 4.86 (dd, J_1 = 7.7 Hz, J_2 = 6.1 Hz, 1 H), 4.27 (dd, J_1 = 8.3 Hz, J_2 = 7.7 Hz, 1 H), 4.03 (dd, J_1 = 8.6 Hz, J_2 = 6.1 Hz, 1 H), 1.51 (s, 3 H), 1.44 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 217.0, 197.7, 111.5, 93.2, 80.2, 79.0, 67.3, 26.3, 25.9. FTIR ν 2889*m*, 2937*w*, 1958*m*, 1932*m*, 1763*w*, 1691*s*, 1457*w*, 1374*m*, 1260*m*, 1214*s*, 1152*m*, 1066*s*, 964*w*, 844*s* cm^{-1}.

(*R*)-1-[(*R*)-3a-((*R*)-*tert*-butyl-dimethyl-silanyloxymethyl)-2,2-dimethyl-6,6a-dihydro-3aH-cyclopenta[1,3]dioxol-4-yl]-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-hexa-4,5-dien-1-yn-3-ol (105)

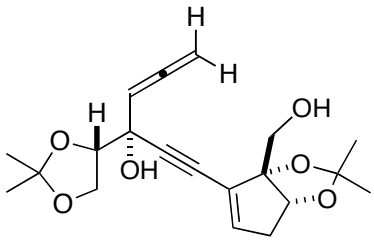


To a cooled (–78 °C) solution of alkyne **101** (810 mg, 2.63 mmol, 1.10 equiv) in THF (12 mL) was added *n*BuLi (1.6 M in hexane) (1.64 mL, 2.63 mmol, 1.10 equiv) and the resulting solution stirred at –78 °C for 30 minutes. Separately in another flask, to a cooled (–78 °C) solution of allene **104** (407 mg, 2.42 mmol, 1.00 equiv) in THF (9 mL) was added a solution of $CeCl_3$•2LiCl (0.2 M in THF) (13.2 mL, 2.63 mmol, 1.10 equiv). After 5 minutes, the deprotonated alkyne solution was transferred by canula and the resulting solution heated to –40 °C and leaved to return to 0 °C over 2 hours. The reaction was quenched by addition of saturated NH_4Cl and extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 10:0 → 8:2) afforded **105** (860 mg, 1.80 mmol, 75%, *d.r.* = 94:6) as a pale yellow oil. A fraction of unreacted alkyne **101** (259 mg, 0.54 mmol) was recovered. R_f = 0.33 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{20.9}_D$ (*c* 0.955, $CHCl_3$) = +19.8°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.12 (s, 1 H), 5.40 (t, J = 6.7 Hz, 1 H), 5.02 (d, J = 6.7 Hz, 2 H), 4.63 (d, J = 4.5 Hz, 1 H), 4.27 (t, J = 6.7 Hz, 1 H), 4.13 (dd, J_1 = 8.6 Hz, J_2 = 6.7 Hz, 1 H), 4.07 (dd, J_1 = 8.6 Hz, J_2 = 6.7 Hz, 1 H), 3.78 (s, 2 H), 2.81 (s, 1 H), 2.59 (ddd, J_1 = 18.6 Hz, J_2 = 4.5 Hz, J_3 = 2.2 Hz, 1 H), 2.52 (dd, J_1 = 18.6 Hz, J_2 = 2.9 Hz, 1 H), 1.51 (s, 3 H), 1.40 (s, 3 H), 1.39 (s, 3 H), 1.39 (s, 3 H), 0.88 (s, 9 H), 0.07 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR

(100 MHz, $CDCl_3$) δ 207.5, 139.2, 126.9, 111.0, 111.0, 96.2, 94.9, 90.3, 81.5, 81.2, 81.2, 80.0, 70.8, 66.6, 64.5, 38.6, 28.2, 27.6, 26.7, 26.2, 25.9, 18.6, –5.0, –5.1. HRMS-ESI calcd for $C_{26}H_{40}O_6SiNa$: $[M+Na]^+$ 499.2492; found 499.2478. FTIR ν 3429*w*, 2987*w*, 2932*m*, 2859*w*, 1960*w*, 1463*w*, 1372*m*, 1252*s*, 1215*s*, 1158*m*, 1077*s*, 1007*w*, 930*w*, 840*s*, 779*m*, 664*m* cm^{-1}.

$CeCl_3$•2LiCl solution (0.2 M in THF): $CeCl_3$•$7H_2O$ (1.12 g, 3.00 mmol, 1.00 equiv) and LiCl (254 mg, 6.00 mmol, 2.00 equiv) were dried in a Schlenk tube under HV (< 0.1 mbar) with gradually increase of the temperature from 25 °C to 150 °C over 3 hours and then additional 2 hours at 150 °C. During the process a fluent constant stirring was required to maintain the mixture as a white homogeneous fine powder. The mixture was put under Ar, cooled to RT and THF (15 mL) was added. The resulting white suspension was vigorously stirred overnight to obtain a clear solution.[250] The $CeCl_3$•2LiCl solution can be stored under Ar in the fridge for more than one week without degradation.

(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-1-[(*R*)-3a-((*R*)-hydroxymethyl)-2,2-dimethyl-6,6a-dihydro-3a*H*-cyclopenta[1,3]dioxol-4-yl]-hexa-4,5-dien-1-yn-3-ol (106)

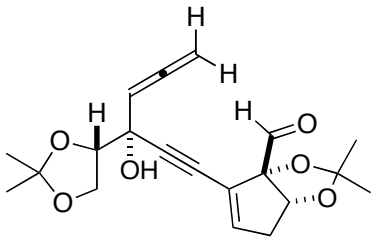


To a cooled (0 °C) solution of allene **105** (22.0 mg, 0.05 mmol, 1.00 equiv) in THF (2 mL) was added TBAF (1.0 M in THF) (104 µL, 0.10 mmol, 2.25 equiv). The resulting solution was stirred at 0 °C for 5 minutes, then allowed to return to RT and stirred for 3.5 hours. The reaction was transferred in a mixture water/Et_2O, the organic phase separated and washed with water (1x) and brine (1x), dried (Na_2SO_4), filtered and concentrated. Purification by chromatography on SiO_2 (CH_2Cl_2/EtOAc 8:2 → 4:6) afforded **106** (16.7 mg, 0.05 mmol, quant.) as a pale yellow oil. R_f = 0.15 (CH_2Cl_2/AcOEt 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.18 (s, 1 H), 5.40 (t, *J* = 6.7 Hz, 1 H), 5.04 (d, *J* = 6.7 Hz, 2 H), 4.65 (d, *J* = 4.5 Hz, 1 H), 4.26 (t, *J* = 6.4 Hz, 1 H), 4.13 (dd, J_1 = 8.6 Hz, J_2 = 7.0 Hz, 1 H), 4.05 (dd, J_1 = 8.6 Hz, J_2 = 6.4 Hz, 1 H), 3.90

[250] The complete solubilization of the $CeCl_3$•2LiCl salt can sometimes require more time.

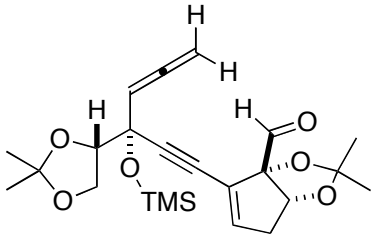
(dd, J_1 = 11.8 Hz, J_2 = 4.2 Hz, 1 H), 3.52 (dd, J_1 = 11.8 Hz, J_2 = 9.3 Hz, 1 H), 2.87 (s, 1 H), 2.66 (ddd, J_1 = 18.9 Hz, J_2 = 4.6 Hz, J_3 = 2.2 Hz, 1 H), 2.59 (dd, J_1 = 18.6 Hz, J_2 = 2.2 Hz, 1 H), 1.94 (dd, J_1 = 9.6 Hz, J_2 = 4.2 Hz, 1 H), 1.52 (s, 3 H), 1.43 (s, 3 H), 1.42 (s, 3 H), 1.41 (s, 3 H).

(*R*)-4-[(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-3-hydroxy-hexa-4,5-dien-1-yn-yl]-2,2-dimethyl-6,6a-dihydro-cyclopenta[1,3]dioxole-3a-carbaldehyde (107)



To a solution of alcohol **106** (16.0 mg, 0.044 mmol, 1.00 equiv) in CH_2Cl_2 (0.7 mL) was added DMP (24 mg, 0.057 mmol, 1.30 equiv) and the resulting mixture stirred at RT for 2.5 hours. The reaction was diluted in a mixture hexane/EtOAc (9.5:0.5) and directly loaded on a column of silica. Purification by chromatography on SiO_2 (hexane/EtOAc 9.5:0.5 → 4:6) afforded aldehyde **107** (14.0 mg, 0.039 mmol, 88%) as a pale yellow oil. R_f = 0.31 (hexane/AcOEt 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.84 (s, 1 H), 6.28 (t, J = 2.6 Hz, 1 H), 5.36 (t, J = 6.7 Hz, 1 H), 5.02 (d, J = 6.7 Hz, 2 H), 4.73 (d, J = 5.4 Hz, 1 H), 4.24 (t, J = 6.4 Hz, 1 H), 4.10 (dd, J_1 = 8.3 Hz, J_2 = 6.7 Hz, 1 H), 4.00 (dd, J_1 = 8.3 Hz, J_2 = 6.4 Hz, 1 H), 2.84 (br s, 1 H), 2.77 (ddd, J_1 = 19.5 Hz, J_2 = 5.8 Hz, J_3 = 2.6 Hz, 1 H), 2.63 (dd, J_1 = 19.2 Hz, J_2 = 2.6 Hz, 1 H), 1.51 (s, 3 H), 1.50 (s, 3 H), 1.43 (s, 3 H), 1.40 (s, 3 H).

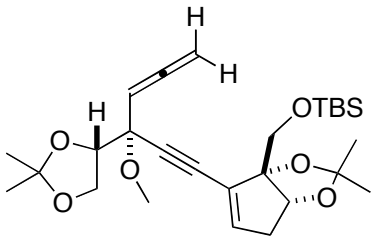
(*R*)-4-[(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-3-trimethylsilanyloxy-hexa-4,5-dien-1-ynyl]-2,2-dimethyl-6,6a-dihydro-cyclopenta[1,3]dioxole-3a-carbaldehyde (109)



To a cooled (–78 °C) solution of **107** (15.0 mg, 0.04 mmol, 1.00 equiv) in CH_2Cl_2 (2.0 mL) were sequentially added 2,6-lutidine (18 µL, 0.15 mmol, 3.75 equiv) and TMSOTf (17 µL, 0.09 mmol, 2.25 equiv). The resulting solution was stirred at –78 °C for 1.5 hours, then quenched by co-addition of a saturated $NaHCO_3$ solution and MeOH

and extracted with CH_2Cl_2 (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 9:1 → 1:1) afforded **109** (11.6 mg, 0.03 mmol, 67%) as a pale yellow oil. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.85 (s, 1 H), 6.25 (t, *J* = 2.2 Hz, 1 H), 5.36 (t, *J* = 6.7 Hz, 1 H), 4.92 (d, *J* = 6.4 Hz, 2 H), 4.72 (d, *J* = 5.4 Hz, 1 H), 4.16 (t, *J* = 6.4 Hz, 1 H), 4.07-3.97 (m, 2 H), 2.76 (ddd, J_1 = 19.2 Hz, J_2 = 5.4 Hz, J_3 = 2.2 Hz, 1 H), 2.63 (dd, J_1 = 19.2 Hz, J_2 = 2.9 Hz, 1 H), 1.49 (s, 3 H), 1.46 (s, 3 H), 1.44 (s, 3 H), 1.39 (s, 3 H), 0.21 (s, 9 H).

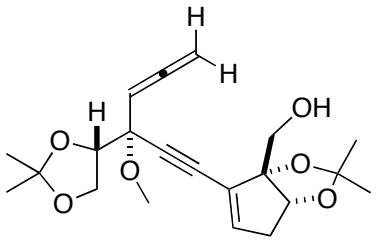
tert-butyl-{(*R*)-(*R*)-4-[(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-3-methoxy-hexa-4,5-dien-1-ynyl]-2,2-dimethyl-6,6a-dihydro-cyclopenta[1,3]dioxol-3a-ylmethoxy}-dimethyl-silane (111)



To a cooled (0 °C) solution of allene **105** (100 mg, 0.21 mmol, 1.00 equiv), 4Å MS (400 mg) and proton sponge (135 mg, 0.63 mmol, 3.00 equiv) in CH_2Cl_2 (1.0 mL) was added Me_3OBF_4 (63 mg, 0.42 mmol, 2.00 equiv). The resulting mixture was stirred 30 minutes at 0 °C, then allowed to return to RT and stirred for 2 hours. The mixture was cooled to 0 °C and a second portion of 4Å MS (200 mg), proton sponge (70 mg, 0.32 mmol, 1.50 equiv) and Me_3OBF_4 (32 mg, 0.21 mmol, 1.00 equiv) were sequentially added. After 5 minutes at 0 °C the mixture was allowed to return to RT and stirred for 1.5 hours. The mixture was cooled once more to 0 °C and a third portion 4Å MS (400 mg), proton sponge (135 mg, 0.63 mmol, 3.00 equiv) and Me_3OBF_4 (63 mg, 0.42 mmol, 2.00 equiv) were sequentially added. After 5 minutes at 0 °C the mixture was allowed to return to RT and stirred for 16 hours. The reaction was diluted in a mixture hexane/EtOAc 2:1, filtered over Celite and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 9:1 → 8:2) afforded **111** (85.0 mg, 0.17 mmol, 83%) as a pale yellow oil. R_f = 0.69 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{21.8}_D$ (*c* 0.83, $CHCl_3$) = –9.6°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.11 (s, 1 H), 5.13 (t, *J* = 6.7 Hz, 1 H), 4.96 (d, *J* = 6.7 Hz, 2 H), 4.62 (d, *J* = 4.5 Hz, 1 H), 4.27 (t, *J* = 6.7 Hz, 1 H), 4.09-4.06 (m, 1 H), 4.02-3.98 (m, 1 H), 3.79 (s, 2 H), 3.42 (s, 3 H), 2.59 (ddd, J_1 = 18.6 Hz, J_2 = 4.5 Hz, J_3 = 2.2 Hz, 1 H), 2.51 (dd, J_1 = 18.9 Hz, J_2 = 2.9 Hz,

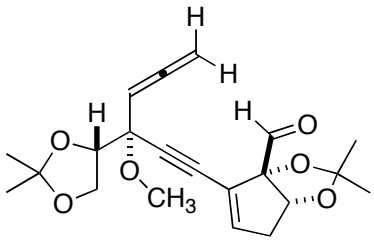
1 H), 1.50 (s, 3 H), 1.40 (s, 3 H), 1.39 (s, 6 H), 0.88 (s, 9 H), 0.07 (s, 3 H), 0.05 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 209.4, 138.8, 127.1, 111.2, 111.0, 96.2, 91.6, 87.4, 83.7, 81.9, 81.2, 78.8, 78.6, 66.7, 64.4, 52.9, 38.5, 28.3, 27.6, 26.8, 26.3, 26.2, 18.6, –5.0, –5.1. DEPT-135 NMR (100 MHz, $CDCl_3$) *CH_3 & CH* δ 138.8, 91.6, 81.9, 81.2, 52.9, 28.3, 27.6, 26.8, 26.3, 26.2, –5.0, –5.1; *CH_2* δ 78.6, 66.7, 64.4, 38.5. HRMS-ESI calcd for $C_{27}H_{42}O_6SiNa$: $[M+Na]^+$ 513.2648; found 513.2662. FTIR ν 2990*w*, 2932*m*, 2862*w*, 1960*w*, 1466*w*, 1369*m*, 1250*m*, 1211*m*, 1157*m*, 1080*s*, 953*w*, 837*s*, 775*m*, 667*s* cm^{-1}.

{(*R*)-4-[(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-3-methoxy-hexa-4,5-dien-1-ynyl]-2,2-dimethyl-6,6a-dihydro-cyclopenta[1,3]dioxol-3a-yl}-methanol (112)



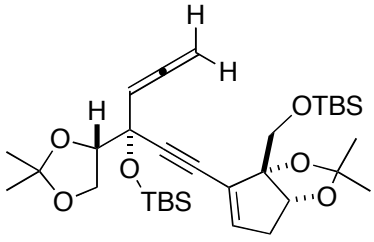
To a cooled (0 °C) solution of allene **111** (82.0 mg, 0.17 mmol, 1.00 equiv) in THF (2 mL) was added TBAF (1.0 M in THF) (335 μL, 0.34 mmol, 2.00 equiv). The resulting solution stirred at 0 °C for 5 minutes, then allowed to return to RT and stirred for 45 minutes. The reaction was diluted with water, extracted with Et_2O (3x) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 8:2 → 6:4) afforded alcohol **112** (61.5 mg, 0.16 mmol, 98%) as a pale yellow oil. R_f = 0.11 (hexane/AcOEt 8:2). Optical rotation $[\alpha]^{21.1}_D$ (*c* 0.58, $CHCl_3$) = –9.9°. ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.17 (m, 1 H), 5.12 (t, *J* = 6.7 Hz, 1 H), 4.98 (d, *J* = 6.4 Hz, 2 H), 4.65 (d, *J* = 4.5 Hz, 1 H), 4.27 (t, *J* = 6.7 Hz, 1 H), 4.08 (dd, J_1 = 8.6 Hz, J_2 = 6.7 Hz, 1 H), 3.98 (d, *J* = 7.4 Hz, 1 H), 3.92 (d, *J* = 11.8 Hz, 1 H), 3.53-3.48 (m, 1 H), 3.42 (s, 3 H), 2.66 (ddd, J_1 = 19.2 Hz, J_2 = 4.8 Hz, J_3 = 2.6 Hz, 1 H), 2.59 (dd, J_1 = 18.2 Hz, J_2 = 2.2 Hz, 1 H), 1.93 (br s, 1 H), 1.50 (s, 3 H), 1.43 (s, 3 H), 1.41 (s, 6 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 209.5, 139.4, 126.5, 111.3, 111.2, 96.5, 91.5, 88.2, 83.2, 81.7, 79.9, 79.0, 78.6, 66.7, 62.6, 53.0, 38.1, 28.3, 27.8, 26.8, 26.2. HRMS-ESI calcd for $C_{21}H_{28}O_6$: $[M]^+$ 377.1964; found 377.1959. FTIR ν 3487*m*, 2982*m*, 2932*m*, 2824*w*, 1960*w*, 1454*w*, 1377*m*, 1258*m*, 1219*s*, 1153*w*, 1080*s*, 1053*s*, 995*w*, 856*m* cm^{-1}.

(*R*)-4-[(*R*)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-3-methoxy-hexa-4,5-dien-1-yn-yl]-2,2-dimethyl-6,6a-dihydro-cyclopenta[1,3]dioxole-3a-carbaldehyde (113)



To a cooled (–78 °C) solution of freshly distilled oxalyl chloride (18 µL, 0.21 mmol, 8.00 equiv) in CH_2Cl_2 (0.5 mL) was slowly added a solution of DMSO (38 µL, 0.54 mmol, 20.0 equiv) in CH_2Cl_2 (0.5 mL). After 20 minutes a solution of alcohol **112** (10.0 mg, 0.03 mmol, 1.00 equiv) in CH_2Cl_2 (0.5 mL) was added and the resulting mixture stirred at –78 °C for 30 minutes. A solution of DIPEA (71 µL, 0.43 mmol, 16.0 equiv) in CH_2Cl_2 (0.5 mL) was slowly added and the clear solution stirred at –78 °C for 10 minutes, then allowed to return to 0 °C and stirred for 2 hours. The reaction was quenched by addition of a buffer phosphate solution (pH = 7), extracted with CH_2Cl_2 (3x) and the combined organic layers washed with brine (1x), dried ($MgSO_4$), filtered and concentrated. The aldehyde **113** was directly used in the next step without further purifications. R_f = 0.73 (hexane/AcOEt 4:6). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.85 (s, 1 H), 6.27 (t, *J* = 2.2 Hz, 1 H), 5.09 (t, *J* = 6.7 Hz, 1 H), 4.96 (d, *J* = 6.4 Hz, 2 H), 4.73 (d, *J* = 5.4 Hz, 1 H), 4.25 (t, *J* = 6.7 Hz, 1 H), 4.07 (dd, J_1 = 8.3 Hz, J_2 = 6.7 Hz, 1 H), 3.94 (t, *J* = 7.7 Hz, 1 H), 3.40 (s, 3 H), 2.77 (ddd, J_1 = 19.5 Hz, J_2 = 5.8 Hz, J_3 = 2.6 Hz, 1 H), 2.64 (dd, J_1 = 19.2 Hz, J_2 = 2.9 Hz, 1 H), 1.50 (s, 3 H), 1.48 (s, 3 H), 1.44 (s, 3 H), 1.41 (s, 3 H).

(1*R*,6a*R*)-6-[(*R*)-3-(*tert*-butyl-dimethyl-silanyloxy)-3-((*S*)-2,2-dimethyl-[1,3]dioxolan-4-yl)-hexa-4,5-dien-1-ynyl]-6a-(*tert*-butyl-dimethyl-silanyloxymethyl)-2,2-dimethyl-4,6a-dihydro-3a*H*-cyclopenta[1,3]dioxole (115)



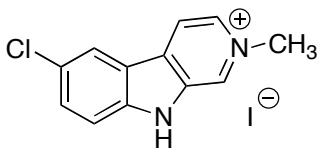
To a cooled (–40 °C) solution of allene **105** (5.0 mg, 0.01 mmol, 1.00 equiv) in CH_2Cl_2 (2.0 mL) were sequentially added 2,6-lutidine (28 µL, 0.24 mmol, 24.0 equiv) and TBSOTf (41 µL, 0.18 mmol, 18.0 equiv). The resulting solution was allowed to return to RT and stirred for 4 hours. The reaction was quenched by addition of water and extracted with CH_2Cl_2 (3x). The combined organic layers were washed with a

saturated NH_4Cl solution (1x), brine (1x), dried ($MgSO_4$), filtered and concentrated. Purification by chromatography on SiO_2 (hexane/EtOAc 99:1 → 95:5) afforded **115,** which was directly used in the next step. R_f = 0.33 (hexane/AcOEt 9.5:0.5). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.09 (s, 1 H), 5.43 (t, *J* = 6.4 Hz, 1 H), 4.91 (d, *J* = 6.4 Hz, 2 H), 4.63 (d, *J* = 4.8 Hz, 1 H), 4.21 (dd, J_1 = 7.0 Hz, J_2 = 6.1 Hz, 1 H), 4.11-4.04 (m, 2 H), 3.77 (s, 2 H), 2.60 (ddd, J_1 = 18.6 Hz, J_2 = 4.5 Hz, J_1 = 2.2 Hz, 1 H), 2.52 (dd, J_1 = 18.6 Hz, J_2 = 2.9 Hz, 1 H), 1.59 (s, 3 H), 1.47 (s, 3 H), 1.39 (s, 3 H), 1.37 (s, 3 H), 0.91 (s, 9 H), 0.88 (s, 9 H), 0.23 (s, 3 H), 0.19 (s, 3 H), 0.07 (s, 3 H), 0.06 (s, 3 H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 207.7, 138.7, 127.3, 111.0, 110.7, 96.2, 95.2, 91.6, 83.2, 82.1, 81.2, 78.7, 72.0, 66.2, 64.5, 38.5, 28.3, 27.6, 26.7, 26.3, 26.2, 25.9, 18.7, 18.6, –2.6, –2.8, –5.0, –5.1.

6.4. Preparation of Nostocarboline and Eudistomin Derivatives

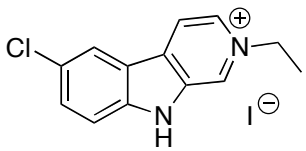
6.4.1. Six-Chloronorharmane Derivatives

6-chloro-2-methyl-9*H*-beta-carbolin-2-ium iodide (133)



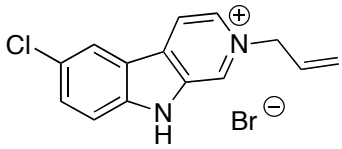
To a solution of 6-chloronorharmane (**130**) (100 mg, 0.49 mmol, 1.00 equiv) in *i*PrOH (5.0 mL) was added methyl iodide (154 μL, 2.47 mmol, 5.00 equiv). The flask was sealed and heated at 85 °C for 4 hours. The reaction was concentrated then the residue triturated in a mixture Et_2O/CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **133** (67.0 mg, 0.31 mmol, 62%) as a crystalline solid. M.p. = 271.0-272.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.29 (s, 1 H), 8.70 (d, *J* = 6.8 Hz, 1 H), 8.56 (d, *J* = 6.4 Hz, 1 H), 8.49 (s, 1 H), 7.80-7.79 (m, 2 H), 4.90 (s, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 142.9, 136.0, 133.2, 132.2, 132.1, 130.5, 127.3, 122.4, 120.5, 117.9, 114.1, 48.5. HRMS-ESI calcd for $C_{12}H_{10}ClN_2$: $[M]^+$ 217.0533; found 217.0540. FTIR ν 3495*w*, 3055*m*, 3001*m*, 2307*w*, 1647*m*, 1574*w*, 1516*m*, 1493*s*, 1450*m*, 1385*w*, 1323*m*, 1285*s*, 1250*m*, 1219*m*, 1153*s*, 1069*s*, 934*w*, 880*m*, 833*m*, 806*s*, 745*m* cm^{-1}.

6-chloro-2-ethyl-9*H*-beta-carbolin-2-ium iodide (134)



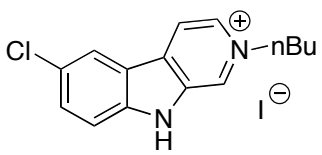
To a mixture of 6-chloronorharmane (**130**) (500 mg, 2.47 mmol, 1.00 equiv) in CH_3CN (15 mL) was added ethyl iodide (490 μL, 6.18 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 18 hours. The reaction was concentrated then the residue was dissolved in a minimum amount of CH_3CN, the product precipitated by addition on Et_2O, collected by filtration and washed with a mixture CH_3CN/Et_2O. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **134** (384 mg, 1.07 mmol, 43%) as a crystalline brown solid. M.p. = 215.0-216.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.38 (s, 1 H), 8.74 (d, J = 6.8 Hz, 1 H), 8.66 (dd, J_1 = 6.8 Hz, J_2 = 1.2 Hz, 1 H), 8.51-8.50 (m, 1 H), 7.81-7.80 (m, 2 H), 4.85 (q, J = 7.2 Hz, 2 H), 1.77 (t, J = 7.2 Hz, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 142.9, 136.1, 132.3, 132.3, 132.1, 129.3, 127.3, 122.4, 120.6, 118.2, 113.8, 56.9, 16.0. HRMS-ESI calcd for $C_{13}H_{12}ClN_2$: $[M]^+$ 231.0689; found 231.0692. FTIR ν 3418*w*, 3098*m*, 3017*m*, 2288*w*, 1647*m*, 1570*w*, 1489*s*, 1447*s*, 1319*m*, 1281*s*, 1250*m*, 1169*m*, 1142*s*, 1065*s*, 937*w*, 876*m*, 806*s*, 725*m*, 706*m* cm^{-1}.

2-allyl-6-chloro-9*H*-beta-carbolin-2-ium bromide (135)



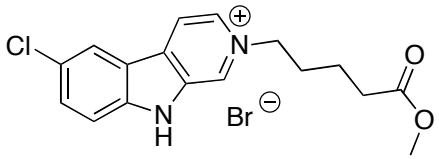
To a solution of 6-chloronorharmane (**130**) (50.0 mg, 0.25 mmol, 1.00 equiv) in *i*PrOH (4.0 mL) was added allyl bromide (43 μL, 0.50 mmol, 2.00 equiv). The flask was sealed and heated at 85 °C for 21 hours. The reaction was concentrated then the residue was dissolved in a minimum amount of CH_3CN, the product precipitated by addition on Et_2O, collected by filtration and washed with a mixture CH_3CN/Et_2O. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **135** (36.0 mg, 0.11 mmol, 45%) as a crystalline solid. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.33 (s, 1 H), 8.75 (d, J = 6.4 Hz, 1 H), 8.60 (d, J = 6.4 Hz, 1 H), 8.51 (d, J = 0.7 Hz, 1 H), 7.84-7.77 (m, 2 H), 6.29 (m, 1 H), 5.56 (d, J = 9.8 Hz, 1 H), 5.55 (d, J = 18.0 Hz, 1 H), 5.41 (d, J = 6.1 Hz, 2 H). The analytical data matched those reported in literature.[229]

2-butyl-6-chloro-9*H*-beta-carbolin-2-ium iodide (136)



To a solution of 6-chloronorharmane (**130**) (30.0 mg, 0.15 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added iodobutane (42 μL, 0.37 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was concentrated then the residue triturated in a mixture Et_2O/ CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **136** (21.2 mg, 0.055 mmol, 37%) as a crystalline solid. M.p. = 213.0-214.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.37 (s, 1 H), 8.73 (d, *J* = 6.8 Hz, 1 H), 8.64 (d, *J* = 6.4 Hz, 1 H), 8.51 (s, 1 H), 7.81-7.80 (m, 2 H), 4.80 (t, *J* = 7.5 Hz, 2 H), 2.12 (quint., *J* = 7.5 Hz, 2 H), 1.49 (sext., *J* = 7.5 Hz, 2 H), 1.06 (t, *J* = 7.2 Hz, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.0, 136.2, 132.3, 132.3, 132.2, 129.5, 127.3, 122.4, 120.6, 118.1, 114.1, 61.3, 33.6, 19.2, 12.5. HRMS-ESI calcd for $C_{15}H_{16}ClN_2$: $[M]^+$ 259.1002; found 259.0999. FTIR ν 3445*w*, 3032*s*, 2997*s*, 2959*s*, 2858*m*, 1651*m*, 1570*w*, 1516*m*, 1489*s*, 1450*s*, 1323*m*, 1281*s*, 1165*m*, 1142*s*, 1065*s*, 903*w*, 872*m*, 806*s*, 756*m*, 725*s* cm^{-1}.

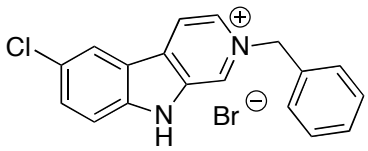
6-chloro-2-(4-methoxycarbonyl-butyl)-9*H*-beta-carbolin-2-ium bromide (137)



To a solution of 6-chloronorharmane (**130**) (30.0 mg, 0.15 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added methyl bromovalerate (53 μL, 0.37 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was concentrated then the residue triturated in a mixture Et_2O/ CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **137** (14.5 mg, 0.036 mmol, 24%) as a crystalline solid. M.p. = 159.0-160.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.36 (s, 1 H), 8.74 (d, *J* = 6.4 Hz, 1 H), 8.64 (d, *J* = 6.0 Hz, 1 H), 8.53 (s, 1 H), 7.83 (s, 2 H), 4.81-4.78 (m, 2 H), 3.68 (s, 3 H), 2.48 (t, *J* = 7.2 Hz, 2 H), 2.19-2.13 (m, 2 H), 1.76-1.72 (m, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 173.8, 143.1, 136.2, 132.4, 132.3, 129.6, 127.4, 122.4, 120.6, 118.1, 114.1, 61.0, 50.7, 32.4, 30.8, 21.1. HRMS-ESI calcd for $C_{17}H_{18}ClN_2O_2$: $[M]^+$ 317.1057; found 317.1062. FTIR ν 3426*w*, 3036*w*, 2994*w*, 2951*m*, 2905*w*, 1728*s*,

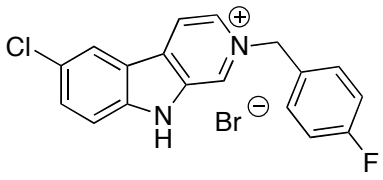
1647*m*, 1570*w*, 1520*m*, 1493*m*, 1439*m*, 1350*m*, 1281*s*, 1227*m*, 1157*s*, 1126*s*, 1069*s*, 984*s*, 891*m*, 810*s*, 752*s* cm^{-1}.

2-benzyl-6-chloro-9*H*-beta-carbolin-2-ium bromide (138)



To a solution of 6-chloronorharmane (**130**) (50.0 mg, 0.25 mmol, 1.00 equiv) in *i*PrOH (8.0 mL) was added benzyl bromide (60 μL, 0.50 mmol, 2.00 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was concentrated then the residue was dissolved in a minimum amount of CH_3CN, the product precipitated by addition on Et_2O, collected by filtration and washed with a mixture CH_3CN/Et_2O. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **138** (54.0 mg, 0.14 mmol, 58%) as a crystalline solid. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.44 (s, 1 H), 8.73-8.68 (m, 2 H), 8.48 (s, 1 H), 7.79 (s, 2 H), 7.56 (dd, J_1 = 7.8 Hz, J_2 = 1.7 Hz, 2 H), 7.51-7.46 (m, 3 H), 5.99 (s, 2 H). The analytical data matched those reported in literature.[229]

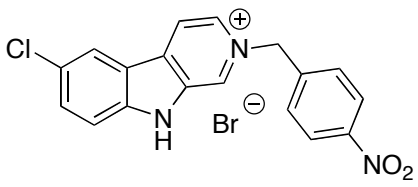
6-chloro-2-(4-fluoro-benzyl)-9*H*-beta-carbolin-2-ium bromide (139)



To a solution of 6-chloronorharmane (**130**) (30.0 mg, 0.15 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 4-fluorobenzyl bromide (69 μL, 0.37 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was concentrated then the residue triturated in a mixture Et_2O/ CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **139** (56.3 mg, 0.14 mmol, 96%) as a crystalline solid. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.50 (s, 1 H), 8.75 (d, J = 6.8 Hz, 1 H), 8.71 (dd, J_1 = 6.8 Hz, J_2 = 1.6 Hz, 1 H), 8.51 (t, J = 1.2 Hz, 1 H), 7.82 (d, J = 1.2 Hz, 2 H), 7.67 (d, J = 5.2 Hz, 1 H), 7.65 (d, J = 5.2 Hz, 1 H), 7.23 (t, J = 8.7 Hz, 2 H), 6.00 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 163.4 (d, J = 248.4 Hz), 143.1, 136.1, 132.6, 132.5, 132.4, 130.9 (d, J = 9.7 Hz), 130.4 (d, J = 3.7 Hz), 129.6, 127.4, 122.5, 120.5,

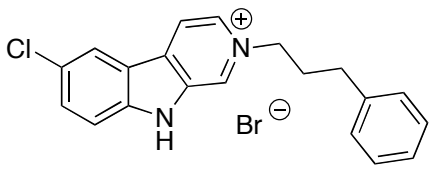
118.3, 116.0 (d, *J* = 22.0 Hz), 114.2, 63.2. HRMS-ESI calcd for $C_{18}H_{13}ClFN_2$: $[M]^+$ 311.0751; found 311.0738. FTIR ν 3460*w*, 3414*w*, 3044*m*, 2982*m*, 2893*w*, 1647*m*, 1605*m*, 1570*w*, 1512*m*, 1489*m*, 1454*m*, 1350*w*, 1281*m*, 1223*m*, 1161*s*, 1119*s*, 1069*s*, 883*w*, 826*s*, 779*m*, 760*m* cm^{-1}.

6-chloro-2-(4-nitro-benzyl)-9*H*-beta-carbolin-2-ium bromide (140)



To a solution of 6-chloronorharmane (**130**) (30.0 mg, 0.15 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 4-nitrobenzyl bromide (80 mg, 0.37 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was concentrated then the residue triturated in a mixture Et_2O/ CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **140** (34.6 mg, 0.083 mmol, 55%) as a crystalline solid. M.p. = 210.0-211.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.55 (s, 1 H), 8.80 (d, *J* = 6.8 Hz, 1 H), 8.74 (d, *J* = 6.8 Hz, 1 H), 8.54 (s, 1 H), 8.34 (d, *J* = 8.7 Hz, 2 H), 7.84 (s, 2 H), 7.76 (d, *J* = 8.7 Hz, 2 H), 6.17 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 144.6, 143.2, 141.2, 136.2, 132.8, 132.7, 132.7, 130.1, 129.3, 127.6, 124.0, 122.6, 120.5, 118.5, 114.2, 62.8. HRMS-ESI calcd for $C_{18}H_{13}ClN_3O_2$: $[M]^+$ 338.0696; found 338.0686. FTIR ν 3387*w*, 3059*m*, 3009*m*, 1647*m*, 1609*m*, 1574*w*, 1520*s*, 1493*s*, 1454*m*, 1342*s*, 1285*s*, 1161*m*, 1130*m*, 1069*m*, 1018*w*, 856*m*, 806*s*, 733*s*, 710*m* cm^{-1}.

6-chloro-2-(3-phenyl-propyl)-9*H*-beta-carbolin-2-ium bromide (141)

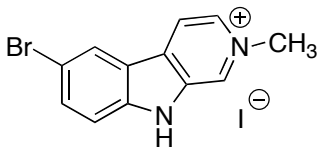


To a solution of 6-chloronorharmane (**130**) (30.0 mg, 0.15 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 1-bromo-3-phenylpropane (56 μL, 0.37 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was concentrated then the residue triturated in a mixture Et_2O/ CH_3CN and the precipitate collected by filtration. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was

concentrated and dried under high vacuum affording **141** (18.7 mg, 0.047 mmol, 31%) as a crystalline solid. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.31 (s, 1 H), 8.68 (d, *J* = 6.4 Hz, 1 H), 8.60 (d, *J* = 6.4 Hz, 1 H), 8.49 (t, *J* = 1.2 Hz, 1 H), 7.81 (d, *J* = 1.2 Hz, 2 H), 7.25 (s, 2 H), 7.24 (s, 2 H), 7.13-7.09 (m, 1 H), 4.83 (t, *J* = 7.2 Hz, 2 H), 2.83 (t, *J* = 7.2 Hz, 2 H), 2.51-2.45 (m, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.2, 140.1, 136.2, 132.3, 132.2, 129.7, 128.2, 128.0, 127.2, 125.9, 122.3, 120.6, 118.0, 114.2, 61.2, 32.6, 32.1, 22.7. HRMS-ESI calcd for $C_{20}H_{18}ClN_2$: $[M]^+$ 321.1158; found 321.1146. FTIR ν 3418*w*, 3024*m*, 2990*m*, 2943*m*, 2905*m*, 2843*w*, 1643*m*, 1574*s*, 1520*m*, 1493*s*, 1450*s*, 1412*s*, 1319*m*, 1285*s*, 1157*s*, 1123*s*, 1069*s*, 922*m*, 876*m*, 826*s*, 741*m* cm^{-1}.

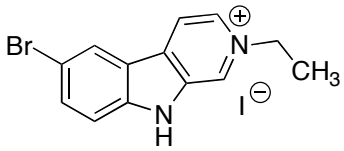
6.4.2. Six-Bromonorharmane derivatives

6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (142)



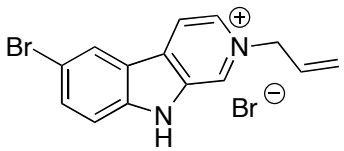
To a solution of 6-bromonorharmane (**131**) (500 mg, 2.47 mmol, 1.00 equiv) in CH_3CN (15 mL) was added methyl iodide (380 μL, 6.18 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 18 hours. The reaction was cooled with an ice-bath, the precipitate filtered, washed with CH_3CN and Et_2O. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **142** (685 mg, 1.76 mmol, 71%) as a yellow solid. An analytical sample was recrystallized (MeOH) for X-ray analysis (crystallographic data are given at the end of the experimental part). M.p. = 292.0-293.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.28 (s, 1 H), 8.71 (d, *J* = 6.4 Hz, 1 H), 8.67 (d, *J* = 2.0 Hz, 1 H), 8.57 (d, *J* = 6.4 Hz, 1 H), 7.94 (dd, J_1 = 8.7 Hz, J_2 = 1.6 Hz, 1 H), 7.75 (d, *J* = 9.1 Hz, 1 H), 4.58 (s, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.1, 135.8, 134.8, 133.3, 131.9, 130.4, 125.6, 121.1, 117.9, 114.4, 114.4, 48.5. HRMS-ESI calcd for $C_{12}H_{10}N_2Br$: $[M]^+$ 261.0027; found 261.0029. FTIR ν 3040*m*, 1643*m*, 1566*w*, 1520*w*, 1485*s*, 1447*s*, 1323*m*, 1277*s*, 1254*s*, 1146*m*, 1123*m*, 1053*m*, 872*m*, 810*s*, 729*m*, 694*m* cm^{-1}.

6-bromo-2-ethyl-9*H*-beta-carbolin-2-ium iodide (143)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added ethyl iodide (12 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **143** (20.0 mg, 0.05 mmol, 83%) as a crystalline solid. M.p. = 226.5-227.5 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.37 (s, 1 H), 8.74 (d, J = 6.4 Hz, 1 H), 8.68 (d, J = 1.6 Hz, 1 H), 8.65 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 7.94 (dd, J_1 = 8.7 Hz, J_2 = 2.0 Hz, 1 H), 7.76 (d, J = 8.7 Hz, 1 H), 4.84 (q, J = 7.2 Hz, 2 H), 1.76 (t, J = 7.2 Hz, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.3, 136.0, 134.8, 132.2, 132.1, 129.3, 125.6, 121.2, 118.2, 114.4, 114.4, 56.9, 16.0. HRMS-ESI calcd for $C_{13}H_{12}N_2Br$: $[M]^+$ 275.0184; found 275.0192. FTIR ν 3514*w*, 3055*s*, 2955*m*, 1647*s*, 1612*w*, 1516*m*, 1493*s*, 1450*s*, 1319*m*, 1281*s*, 1250*s*, 1165*m*, 1142*s*, 1053*s*, 937*m*, 868*s*, 814*s*, 802*s*, 725*s* cm^{-1}.

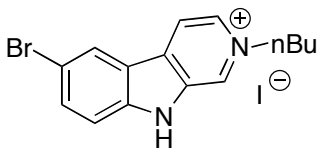
2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (144)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added allyl bromide (13 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **144** (14.6 mg, 0.04 mmol, 66%) as a crystalline solid. An analytical sample was recrystallized (Et_2O/hexane) for X-ray analysis (crystallographic data are given at the end of the experimental part). M.p. = 195.0-196.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.35 (s, 1 H), 8.75 (d, J = 6.4 Hz, 1 H), 8.68 (d, J = 1.6 Hz, 1 H), 8.62 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 7.95 (dd, J_1 = 8.7 Hz, J_2 = 2.0 Hz, 1 H), 7.76 (d, J = 8.7 Hz, 1 H), 6.30 (m, 1 H), 5.57 (dd, J_1 = 9.9, J_2 = 1.2 Hz), 5.56 (dd, J_1 = 15.9 Hz, J_2 = 1.2 Hz), 5.43 (d, J = 6.0 Hz, 3 H). ^{13}C-NMR (125

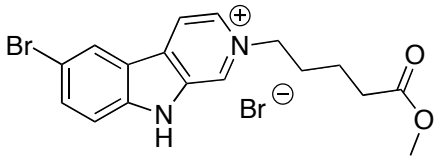
MHz, CD_3OD) δ 143.3, 135.9, 135.0, 132.5, 132.4, 131.4, 129.6, 125.7, 121.3, 121.1, 118.2, 114.5, 114.4, 63.0. HRMS-ESI calcd for $C_{14}H_{12}N_2Br$: $[M]^+$ 287.0184; found 287.0179. FTIR ν 3024*m*, 2997*m*, 1643*s*, 1570*w*, 1512*m*, 1489*s*, 1450*s*, 1358*w*, 1315*m*, 1281*s*, 1254*s*, 1123*s*, 1053*s*, 991*m*, 937*s*, 833*s*, 814*s*, 768*s*, 725*m* cm^{-1}.

2-butyl-6-bromo-9*H*-beta-carbolin-2-ium iodide (145)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added iodobutane (17 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **145** (11.0 mg, 0.026 mmol, 43%) as a crystalline solid. M.p. = 231.5-232.5 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.38 (s, 1 H), 8.73 (d, *J* = 6.4 Hz, 1 H), 8.66 (d, *J* = 1.2 Hz, 1 H), 8.65 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 7.93 (dd, J_1 = 9.1 Hz, J_2 = 2.0 Hz, 1 H), 7.75 (d, *J* = 8.3 Hz, 1 H), 4.80 (t, *J* = 7.5 Hz, 2 H), 2.12 (quint., *J* = 7.5 Hz, 2 H), 1.50 (sext., *J* = 7.5 Hz, 2 H), 1.06 (t, *J* = 7.5 Hz, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.2, 135.9, 134.9, 132.5, 132.2, 129.5, 125.6, 121.1, 118.1, 114.4, 114.4, 61.3, 33.6, 19.2, 12.5. HRMS-ESI calcd for $C_{15}H_{16}N_2Br$: $[M]^+$ 303.0497; found 303.0508. FTIR ν 3028*s*, 2994*s*, 2955*s*, 2855*m*, 1647*m*, 1570*w*, 1516*m*, 1489*s*, 1447*s*, 1319*m*, 1281*s*, 1254*m*, 1165*m*, 1138*s*, 1049*s*, 1022*w*, 903*w*, 864*s*, 802*s*, 725*s* cm^{-1}.

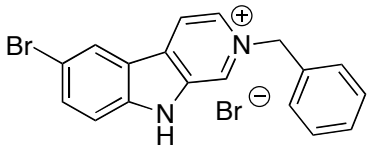
6-bromo-2-(4-methoxycarbonyl-butyl)-9*H*-beta-carbolin-2-ium bromide (146)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added methyl bromovalerate (22 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **146** (20.9 mg,

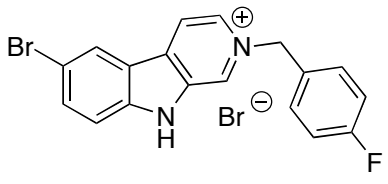
0.047 mmol, 79%) as a crystalline solid. M.p. = 203.5-204.5 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.39 (s, 1 H), 8.66-8.65 (m, 2 H), 8.66 (dd, J_1 = 6.8 Hz, J_2 = 1.2 Hz, 1 H), 7.94 (dd, J_1 = 8.7 Hz, J_2 = 1.6 Hz, 1 H), 7.75 (d, J = 8.7 Hz, 1 H), 4.82 (t, J = 7.5 Hz, 2 H), 3.68 (s, 3 H), 2.48 (t, J = 7.2 Hz, 2 H), 2.17 (quint., J = 7.5 Hz, 2 H), 1.74 (quint., J = 7.2 Hz, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 173.8, 143.2, 135.9, 134.9, 132.5, 132.2, 129.6, 125.7, 121.1, 118.1, 114.4, 114.4, 61.0, 50.8, 32.4, 30.8, 21.1. HRMS-ESI calcd for $C_{17}H_{18}N_2O_2Br$: $[M]^+$ 361.0552; found 361.0555. FTIR ν 3024*s*, 2986*s*, 2947*s*, 2913*s*, 2843*m*, 1736*s*, 1643*s*, 1612*w*, 1574*w*, 1520*m*, 1489*s*, 1447*s*, 1366*m*, 1285*s*, 1242*s*, 1200*s*, 1153*s*, 1126*s*, 1092*m*, 972*m*, 922*m*, 872*s*, 826*s*, 741*s* cm^{-1}.

2-benzyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (147)



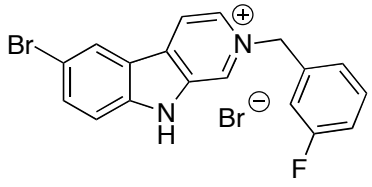
To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added benzyl bromide (18 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **147** (25.0 mg, 0.06 mmol, quant.) as a crystalline solid. M.p. = 235.5-236.5 °C. ^{1}H–NMR (500 MHz, CD_3OD) δ 9.45 (s, 1 H), 8.73 (d, J = 6.0 Hz, 1 H), 8.71 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 8.66 (dd, J_1 = 2.0 Hz, J_2 = 0.8 Hz, 1 H), 7.93 (dd, J_1 = 8.7 Hz, J_2 = 2.0 Hz, 1 H), 7.74 (d, J = 8.3 Hz, 1 H), 7.57 (dd, J_1 = 7.9 Hz, J_2 = 2.0 Hz, 2 H), 7.51-7.47 (m, 3 H), 5.99 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.3, 135.9, 135.0, 134.3, 132.5, 132.4, 129.5, 129.4, 129.2, 128.4, 125.7, 121.1, 118.3, 114.5, 114.4, 64.1. HRMS-ESI calcd for $C_{18}H_{14}N_2Br$: $[M]^+$ 337.0340; found 337.0336. FTIR ν 3021*m*, 2986*m*, 2943*m*, 2889*m*, 2843*m*, 1647*m*, 1566*w*, 1520*w*, 1489*s*, 1454*s*, 1319*m*, 1281*s*, 1254*m*, 1200*w*, 1161*m*, 1126*m*, 1053*m*, 1026*w*, 868*m*, 814*s*, 733*s*, 706*s* cm^{-1}.

6-bromo-2-(4-fluoro-benzyl)-9*H*-beta-carbolin-2-ium bromide (148)



To a solution of 6-bromonorharmane (**131**) (25.0 mg, 0.10 mmol, 1.00 equiv) in CH_3CN (1.5 mL) was added 4-fluorobenzylbromide (31 μL, 0.25 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 1 hour. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **148** (42.2 mg, 0.096 mmol, 96%) as a crystalline solid. M.p. = 279.5-280.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.43 (s, 1 H), 8.73-8.66 (m, 3 H), 7.94 (dd, J_1 = 8.8 Hz, J_2 = 1.7 Hz, 1 H), 7.75 (d, J = 8.8 Hz, 1 H), 7.62 (dd, J_1 = 8.5 Hz, J_2 = 5.4 Hz, 2 H), 7.23 (t, J = 8.5 Hz, 2 H), 5.97 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 163.8 (d, J = 248.0 Hz), 143.7, 136.3, 135.5, 132.9, 132.8, 131.2 (d, J = 8.8 Hz), 130.8 (d, J = 3.2 Hz), 129.9, 126.1, 121.5, 118.7, 116.4 (d, J = 22.1 Hz), 114.9, 114.8, 63.6. HRMS-ESI calcd for $C_{18}H_{13}FN_2Br$: $[M]^+$ 355.0246; found 355.0232. FTIR ν 3040*m*, 2982*m*, 2943*m*, 2886*m*, 1647*m*, 1605*w*, 1508*s*, 1489*s*, 1454*s*, 1350*m*, 1277*s*, 1250*m*, 1223*s*, 1165*s*, 1119*s*, 1053*m*, 826*s*, 779*s*, 698*s* cm^{-1}.

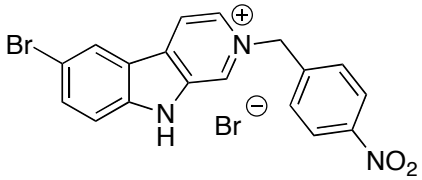
6-bromo-2-(3-fluoro-benzyl)-9*H*-beta-carbolin-2-ium bromide (149)



To a solution of 6-bromonorharmane (**131**) (25.0 mg, 0.10 mmol, 1.00 equiv) in CH_3CN (1.5 mL) was added 3-fluorobenzylbromide (31 μL, 0.25 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 22 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **149** (19.3 mg, 0.044 mmol, 44%) as a crystalline solid. M.p. = 237.0-238.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.48 (s, 1 H), 8.74 (d, J = 6.8 Hz, 1 H), 8.72 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 8.64 (d, J = 2.0 Hz, 1 H), 7.91 (dd, J_1 = 9.1 Hz, J_2 = 2.0 Hz, 1 H), 7.74 (d, J = 9.1 Hz, 1 H), 7.54-7.50 (m, 1 H), 7.40-7.36 (m, 2 H), 7.22 (ddd, J_1 = 9.1 Hz, J_2 = 8.3 Hz, J_3 = 2.4 Hz, 1 H), 6.02 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 165.4 (d, J = 247.4 Hz), 145.6, 138.9 (d, J = 8.2 Hz), 138.1, 137.3, 134.8, 134.7, 133.4 (d, J = 8.2

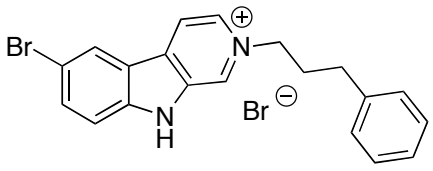
Hz), 131.9, 128.0, 126.5 (d, *J* = 2.7 Hz), 123.3, 120.6, 118.4 (d, *J* = 21.1 Hz), 117.6 (d, *J* = 23.9 Hz), 116.8, 116.7, 65.5. HRMS-ESI calcd for $C_{18}H_{13}FN_2Br$: $[M]^+$ 355.0246; found 355.0232. FTIR ν 3453*w*, 3040*m*, 2947*m*, 2893*m*, 2839*m*, 2696*w*, 1643*m*, 1593*m*, 1516*m*, 1485*s*, 1450*s*, 1319*m*, 1281*s*, 1254*s*, 1150*m*, 1123*s*, 1053*m*, 876*m*, 806*s*, 752*s* cm^{-1}.

6-bromo-2-(4-nitro-benzyl)-9*H*-beta-carbolin-2-ium bromide (150)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 4-nitrobenzyl bromide (32.4 mg, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 5 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **150** (27.6 mg, 0.060 mmol, 99%) as a crystalline solid. M.p. = 261.0-262.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.52 (s, 1 H), 8.79 (d, *J* = 6.4 Hz, 1 H), 8.74 (d, *J* = 6.0 Hz, 1 H), 8.70 (s, 1 H), 8.34 (d, *J* = 8.3 Hz, 2 H), 7.96 (d, *J* = 8.3 Hz, 1 H), 7.78-7.74 (m, 3 H), 6.16 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 148.5, 143.5, 141.2, 136.0, 135.3, 132.8, 132.7, 130.1, 129.3, 125.8, 124.0, 121.1, 118.5, 114.7, 114.5, 62.8. HRMS-ESI calcd for $C_{18}H_{13}N_3O_2Br$: $[M]^+$ 382.0191; found 382.0183. FTIR ν 3140*w*, 3048*m*, 3009*m*, 2955*w*, 2855*w*, 1643*m*, 1605*w*, 1516*s*, 1489*s*, 1450*m*, 1339*s*, 1285*s*, 1258*m*, 1223*m*, 1161*m*, 1057*w*, 945*m*, 856*m*, 818*s*, 729*s* cm^{-1}.

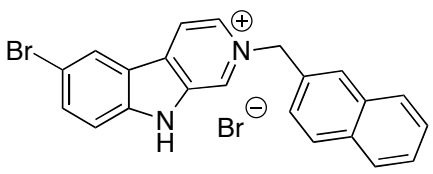
6-bromo-2-(3-phenyl-propyl)-9*H*-beta-carbolin-2-ium bromide (151)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 1-bromo-3-phenylpropane (23 μL, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 15 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any

precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **151** (25.2 mg, 0.056 mmol, 94%) as a crystalline solid. M.p. = 257.5-258.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.30 (s, 1 H), 8.68 (d, *J* = 6.8 Hz, 1 H), 8.65 (d, *J* = 1.6 Hz, 1 H), 8.62 (dd, J_1 = 6.4 Hz, J_2 = 0.8 Hz, 1 H), 7.93 (dd, J_1 = 9.1 Hz, J_2 = 2.0 Hz, 1 H), 7.74 (d, *J* = 8.7 Hz, 1 H), 7.25 (s, 2 H), 7.24 (s, 2 H), 7.12 (m, 1 H), 4.83 (t, *J* = 7.5 Hz, 2 H), 2.83 (t, *J* = 7.2 Hz, 2 H), 2.49 (quint, *J* = 7.5 Hz, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.1, 140.0, 135.8, 134.9, 132.4, 132.2, 129.6, 128.2, 128.0, 125.9, 125.6, 121.1, 118.1, 114.4, 114.3, 61.2, 32.6, 32.1. HRMS-ESI calcd for $C_{20}H_{18}N_2Br$: $[M]^+$ 365.0653; found 365.0653. FTIR ν 3410*w*, 3024*s*, 2986*s*, 2943*s*, 2839*m*, 1639*s*, 1609*m*, 1570*w*, 1516*w*, 1489*s*, 1450*s*, 1315*m*, 1281*s*, 1254*s*, 1157*s*, 1126*s*, 1049*m*, 972*w*, 907*w*, 876*s*, 826*s*, 822*s*, 733*s*, 694*s* cm^{-1}.

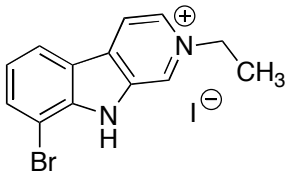
6-bromo-2-naphthalen-2-ylmethyl-9*H*-beta-carbolin-2-ium bromide (152)



To a solution of 6-bromonorharmane (**131**) (15.0 mg, 0.06 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 2-bromomethyl naphtalene (33.2 mg, 0.15 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C for 5 hours. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **152** (22.9 mg, 0.049 mmol, 82%) as a crystalline solid. M.p. = 223.5-224.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.49 (s, 1 H), 8.78-8.73 (m, 2 H), 8.67 (d, *J* = 1.6 Hz, 1 H), 8.11 (s, 1 H), 7.99-7.91 (m, 4 H), 7.75 (d, *J* = 9.1 Hz, 1 H), 7.60-7.58 (m, 3 H), 6.15 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.3, 136.0, 135.1, 133.6, 133.4, 132.6, 132.5, 131.5, 129.6, 129.3, 128.3, 127.9, 127.5, 127.0, 126.7, 125.7, 124.9, 121.1, 118.3, 114.5, 114.4, 64.3. HRMS-ESI calcd for $C_{22}H_{16}N_2Br$: $[M]^+$ 387.0497; found 387.0499. FTIR ν 3615*w*, 3537*w*, 3368*w*, 3040*m*, 2986*m*, 2936*m*, 2882*m*, 2839*m*, 2797*m*, 2646*w*, 1643*m*, 1609*w*, 1520*m*, 1489*s*, 1450*m*, 1319*m*, 1281*s*, 1157*m*, 1126*s*, 1053*m*, 968*w*, 872*m*, 818*s*, 775*s*, 733*s*, 706*m* cm^{-1}.

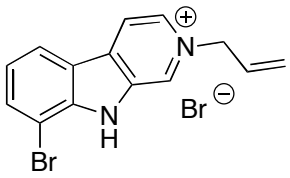
6.4.3. Eight-Bromonorharmane derivatives

8-bromo-2-ethyl-9*H*-beta-carbolin-2-ium iodide (153)



To a solution of 8-bromonorharmane (**132**) (10.0 mg, 0.05 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added ethyl iodide (10 μL, 0.13 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **153** (4.60 mg, 0.011 mmol, 23%) as a crystalline solid. M.p. = 292.0-293.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.28 (s, 1 H), 8.77 (d, *J* = 6.4 Hz, 1 H), 8.70 (d, *J* = 6.4 Hz, 1 H), 8.48 (d, *J* = 7.9 Hz, 1 H), 8.06 (d, *J* = 7.5 Hz, 1 H), 7.45 (t, *J* = 7.9 Hz, 1 H), 4.86 (q, *J* = 7.9 Hz, 2 H), 1.77 (t, *J* = 7.2 Hz, 3 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.1, 135.9, 134.4, 133.7, 132.6, 129.3, 123.0, 122.4, 121.1, 118.5, 105.1, 57.0, 16.0. HRMS-ESI calcd for $C_{13}H_{12}N_2Br$: $[M]^+$ 275.0184; found 275.0182. FTIR ν 3356*w*, 3048*m*, 3009*m*, 2326*w*, 1643*m*, 1555*m*, 1497*m*, 1470*s*, 1327*s*, 1246*m*, 1215*m*, 1130*s*, 1034*m*, 837*s*, 791*s*, 748*s* cm^{-1}.

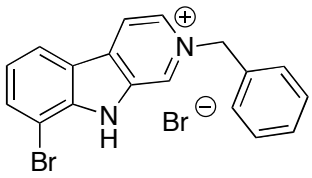
2-allyl-8-bromo-9*H*-beta-carbolin-2-ium bromide (154)



To a solution of 8-bromonorharmane (**132**) (10.0 mg, 0.05 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added allyl bromide (11 μL, 0.13 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **154** (4.50 mg, 0.012 mmol, 24%) as a crystalline solid. M.p. = 220.0-221.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.25 (s, 1 H), 8.79 (d, *J* = 6.4 Hz, 1 H), 8.66 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 8.49 (dd, J_1 = 7.9 Hz, J_2 = 0.8 Hz, 1 H), 8.07 (dd, J_1 = 7.5 Hz, J_2 = 0.8 Hz, 1 H), 7.46 (t, *J* = 7.9 Hz, 1 H), 6.34-6.26 (m, 1 H), 5.58 (dd, J_1 = 10.3 Hz, J_2 = 1.2 Hz, 1 H), 5.57 (dd, J_1 = 16.7 Hz, J_2 = 1.2 Hz, 1 H), 5.45 (d, *J* = 6.4 Hz, 2 H). ^{13}C-NMR (125 MHz,

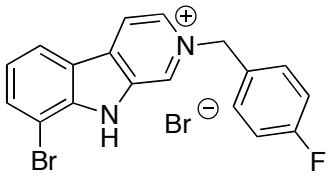
CD_3OD) δ 143.2, 135.8, 134.5, 134.0, 133.0, 131.4, 129.5, 123.1, 122.4, 121.4, 121.0, 118.5, 105.1, 63.1. HRMS-ESI calcd for $C_{14}H_{12}N_2Br$: $[M]^+$ 287.0184; found 287.0178. FTIR ν 3352*w*, 3051*m*, 3017*m*, 2974*m*, 2905*m*, 2858*m*, 1647*m*, 1616*w*, 1558*m*, 1501*m*, 1470*s*, 1327*s*, 1300*m*, 1219*m*, 1138*m*, 1115*m*, 1034*m*, 1011*m*, 953*m*, 810*m*, 783*s*, 745*s*, 683*m* cm^{-1}.

2-benzyl-8-bromo-9*H*-beta-carbolin-2-ium bromide (155)



To a solution of 8-bromonorharmane (**132**) (10.0 mg, 0.05 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added benzyl bromide (15 μL, 0.13 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **155** (9.10 mg, 0.022 mmol, 44%) as a crystalline solid. M.p. = 235.0-236.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.34 (s, 1 H), 8.77 (d, *J* = 6.4 Hz, 1 H), 8.75 (d, *J* = 6.4 Hz, 1 H), 8.46 (d, *J* = 7.9 Hz, 1 H), 8.04 (d, *J* = 7.5 Hz, 1 H), 7.58-7.56 (m, 2 H), 7.53-7.48 (m, 3 H), 7.43 (t, *J* = 7.5 Hz, 1 H), 6.02 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.2, 135.8, 134.5, 134.2, 133.9, 133.1, 129.5, 129.4, 129.3, 128.5, 123.1, 122.4, 121.0, 118.6, 105.1, 64.1. HRMS-ESI calcd for $C_{18}H_{14}N_2Br$: $[M]^+$ 337.0340; found 337.0350. FTIR ν 3399*w*, 3055*m*, 3017*m*, 1643*m*, 1562*w*, 1520*m*, 1497*m*, 1470*s*, 1454*m*, 1327*s*, 1254*m*, 1119*m*, 1034*w*, 818*m*, 787*m*, 748*s*, 706*s* cm^{-1}.

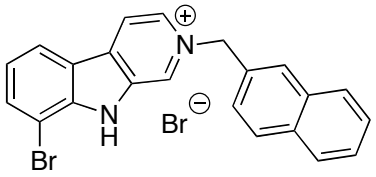
8-bromo-2-(4-fluoro-benzyl)-9*H*-beta-carbolin-2-ium bromide (156)



To a solution of 8-bromonorharmane (**132**) (10.0 mg, 0.05 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 4-fluorobenzyl bromide (16 μL, 0.13 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **156** (5.50 mg,

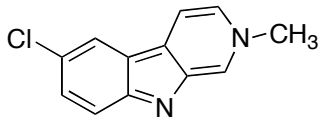
0.013 mmol, 25%) as a crystalline solid. M.p. = 259.5-260.5 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.34 (s, 1 H), 8.78 (d, *J* = 6.8 Hz, 1 H), 8.74 (dd, J_1 = 6.4 Hz, J_2 = 0.8 Hz, 1 H), 8.47 (d, *J* = 8.3 Hz, 1 H), 8.05 (d, *J* = 7.9 Hz, 1 H), 7.64 (dd, J_1 = 8.7 Hz, J_2 = 5.2 Hz, 2 H), 7.44 (t, *J* = 7.9 Hz, 1 H), 7.25 (t, *J* = 8.7 Hz, 2 H), 6.01 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 163.5 (d, *J* = 248.4 Hz), 143.2, 135.8, 134.6, 134.0, 133.0, 130.9 (d, *J* = 8.2 Hz), 130.23 (d, *J* = 2.7 Hz), 129.4, 123.1, 122.5, 121.0, 118.7, 116.1 (d, *J* = 22.9 Hz), 105.1, 63.2. HRMS-ESI calcd for $C_{18}H_{13}N_2BrF$: $[M]^+$ 355.0246; found 355.0257. FTIR ν 3364*w*, 3044*m*, 3001*m*, 2978*m*, 1647*m*, 1562*m*, 1512*m*, 1497*m*, 1470*s*, 1327*s*, 1250*m*, 1138*m*, 1115*m*, 860*m*, 810*m*, 783*s*, 748*s* cm^{-1}.

8-bromo-2-naphthalen-2-ylmethyl-9*H*-beta-carbolin-2-ium bromide (157)



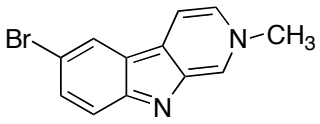
To a solution of 8-bromonorharmane (**132**) (10.0 mg, 0.05 mmol, 1.00 equiv) in CH_3CN (0.5 mL) was added 2-bromomethyl naphtalene (28 mg, 0.13 mmol, 2.50 equiv). The flask was sealed and heated at 85 °C overnight. The reaction was cooled to RT, the precipitate filtered, washed with CH_3CN and pentane. The product was dissolved in MeOH and any precipitate removed by filtration. The filtrated was concentrated and dried under high vacuum affording **157** (10.0 mg, 0.021 mmol, 43%) as a crystalline solid. M.p. = 244.5-245.0 °C. ^{1}H-NMR (500 MHz, CD_3OD) δ 9.40 (s, 1 H), 8.81 (dd, J_1 = 6.4 Hz, J_2 = 1.2 Hz, 1 H), 8.77 (d, *J* = 6.4 Hz, 1 H), 8.46 (d, *J* = 7.9 Hz, 1 H), 8.12 (s, 1 H), 8.03 (d, *J* = 7.5 Hz, 1 H), 7.99 (d, *J* = 8.7 Hz, 1 H), 7.97 (dd, J_1 = 6.0 Hz, J_2 = 3.6 Hz, 1 H), 7.93 (dd, J_1 = 6.0 Hz, J_2 = 3.6 Hz, 1 H), 7.61-7.58 (m, 3 H), 7.43 (t, *J* = 7.9 Hz, 1 H), 6.19 (s, 2 H). ^{13}C-NMR (125 MHz, CD_3OD) δ 143.2, 135.8, 134.5, 134.0, 133.7, 133.4, 133.2, 131.4, 129.5, 129.3, 128.4, 127.9, 127.5, 127.0, 126.8, 125.0, 123.1, 122.4, 121.0, 118.6, 105.1, 64.3. HRMS-ESI calcd for $C_{22}H_{16}N_2Br$: $[M]^+$ 387.0497; found 387.0512. FTIR ν 3372*m*, 3051*m*, 3013*m*, 2928*m*, 1643*m*, 1562*w*, 1516*m*, 1474*m*, 1331*s*, 1258*m*, 1126*s*, 1034*w*, 864*m*, 806*s*, 783*s*, 752*s* cm^{-1}.

6-chloro-2-methyl-2*H*-beta-carboline (158)



To a mixture of 6-chloro-2-methyl-9*H*-beta-carbolin-2-ium iodide (**133**) (33.0 mg, 0.096 mmol, 1.00 equiv) in EtOAc (15.0 mL) a solution of NaOH (1 M) (7.5 mL) was added dropwise. The starting material immediately dissolves to generate a strong yellow mixture that was stirred at RT for 10 minutes. The mixture was extracted with EtOAc (3x) recovering carefully only the organic phases. The combined organic phases could not be dried using standard salts (Na_2SO_4 or $MgSO_4$) without reprotonation of the generated base and they were directly concentrated and dried under high vacuum to afford the anhydronium base **158** (18.1 mg, 0.084 mmol, 88%) as a yellow solid. An analytical sample was recrystallized (MeOH/Et_2O/hexane) for X-ray analysis (crystallographic data are given at the end of the experimental part). ^{1}H-NMR (400 MHz, CD_3OD) δ 8.79 (s, 1 H), 8.25 (d, *J* = 6.2 Hz, 1 H), 8.17 (d, *J* = 2.1 Hz, 1 H), 7.92 (dd, J_1 = 6.5 Hz, J_2 = 1.2 Hz, 1 H), 7.69 (d, *J* = 8.8 Hz, 1 H), 7.49 (dd, J_1 = 8.8 Hz, J_2 = 2.1 Hz, 1 H), 4.38 (s, 3 H). ^{13}C-NMR (100 MHz, CD_3OD) δ 179.0, 153.6, 144.4, 131.6, 129.2, 126.3, 123.3, 121.4, 121.1, 118.2, 115.9, 46.2. HRMS-ESI calcd for $C_{12}H_{10}ClN_2$: $[M+H]^+$ 217.0533; found 217.0525. FTIR ν 3005*w*, 2932*w*, 2855*w*, 1570*s*, 1408*s*, 1335*m*, 1285*m*, 1246*m*, 1157*m*, 1092*w*, 1053*m*, 1015*m*, 922*m*, 872*w*, 806*m*, 783*m*, 752*m*, 702*m* cm^{-1}.

6-bromo-2-methyl-2*H*-beta-carboline (160)



To a mixture of 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**) (47.0 mg, 0.12 mmol, 1.00 equiv) in EtOAc (15.0 mL) a solution of NaOH (3 M) (7.5 mL) was added dropwise. The starting material immediately dissolves to generate a strong yellow mixture that was stirred at RT for 15 minutes. The mixture was extracted with EtOAc (3x) recovering carefully only the organic phases. The combined organic phases could not be dried using standard salts (Na_2SO_4 or $MgSO_4$) without reprotonation of the generated base and they were directly concentrated and dried under high vacuum to afford the anhydronium base **160** (31.5 mg, 0.12 mmol, quant) as a yellow solid. An analytical sample was recrystallized (MeOH/Et_2O/hexane) for X-ray analysis (crystallographic data are given at the end of the experimental part).

^{1}H-NMR (400 MHz, CD_3OD) δ 8.72 (s, 1 H), 8.28 (dd, J_1 = 2.1 Hz, J_2 = 0.6 Hz, 1 H), 8.16 (d, J = 6.2 Hz, 1 H), 7.81 (dd, J_1 = 6.2 Hz, J_2 = 1.2 Hz, 1 H), 7.63 (dd, J_1 = 9.1 Hz, J_2 = 0.6 Hz, 1 H), 7.57 (dd, J_1 = 9.1 Hz, J_2 = 2.1 Hz, 1 H), 4.32 (s, 3 H). ^{13}C-NMR (100 MHz, CD_3OD) δ 179.0, 154.7, 145.0, 131.7, 131.3, 125.8, 124.3, 122.3, 119.0, 115.7, 110.2, 46.1. HRMS-ESI calcd for $C_{12}H_{10}N_2Br$: $[M+H]^+$ 261.0027; found 261.0031. FTIR ν 3063*w*, 3009*w*, 2936*w*, 1624*m*, 1570*s*, 1423*s*, 1335*m*, 1285*s*, 1246*m*, 1153*m*, 1123*m*, 1038*m*, 922*m*, 880*m*, 806*s*, 787*m*, 752*m*, 687*m* cm^{-1}.

6.4.4. Biological Evaluation

Determination of antiprotozoal and cytotoxic activity. *In vitro* assays with *T. b. rhodesiense* STIB 900 bloodstream forms, *P. falciparum* K1 erythocytic stages, *T. cruzi* Tulahen Lac Z C4 amastigotes in L6 cells (rat skeletal myoblasts) and *L. donovani* MHOM-ET/67/L82 axenic amastigotes as well as for cytotoxicity using L6 cells were carried out as previously reported.[251]

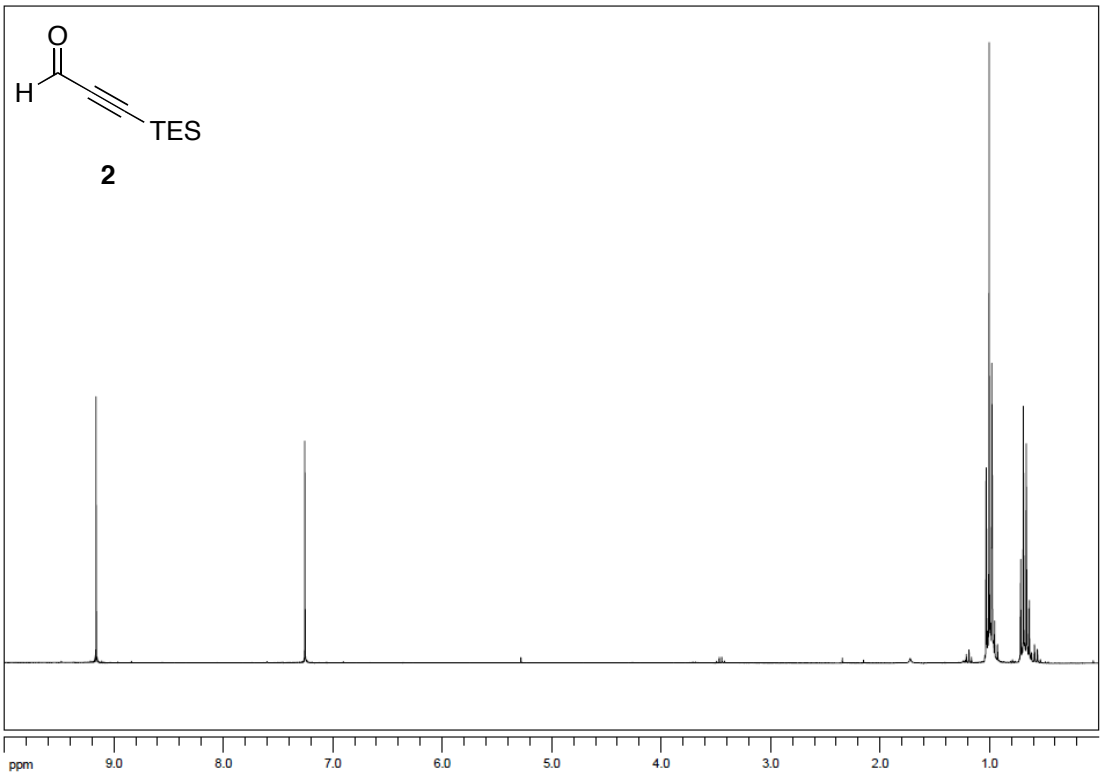
Bacteria and MIC determination. Actinobacterial species used in this study were *Corynebacterium glutamicum* ATCC13032, *Mycobacterium smegmatis* mc^2155 and *Mycobacterium tuberculosis* H37Rv. These were grown in 7H9 medium and tested for susceptibility to nostocarboline derivatives using the resazurin-reduction method.[252] The minimal inhibitory concentration (MIC_{99}) was defined as the lowest drug concentration that prevented growth of 99% of the cells.

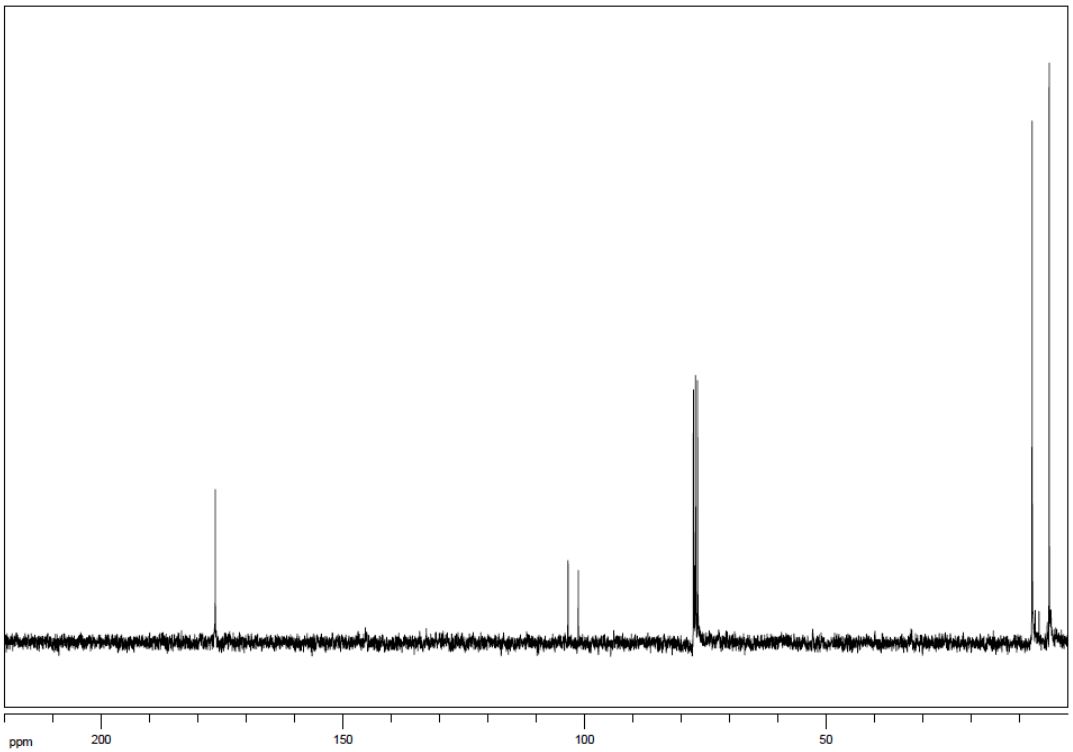
[251] S. Ganapaty, P. S. Thomas, G. Karagianis, P. G. Waterman, R. Brun, *Phytochemistry* **2006**, *67*, 1950-1956.

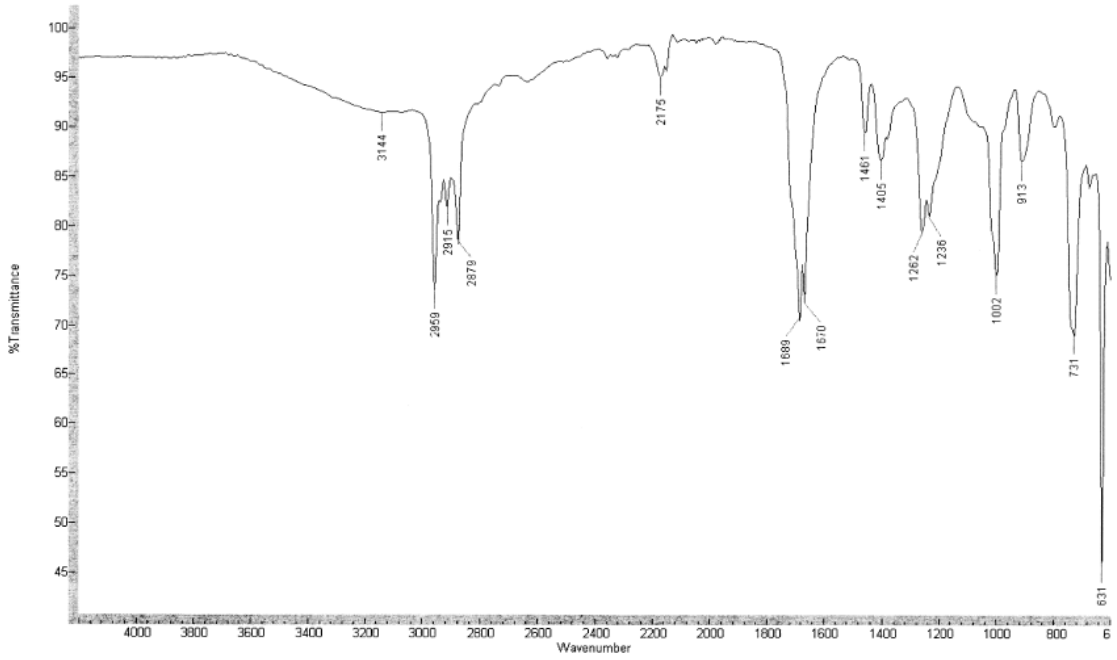
[252] J. C. Palomino, A. Martin, M. Camacho, H. Guerra, J. Swings, F. Portaels, *Antimicrob. Agents Chemother.* **2002**, *46*, 2720-2722.

6.5. Spectra

6.5.1. Spectra from the Anguinomycins C & D Project



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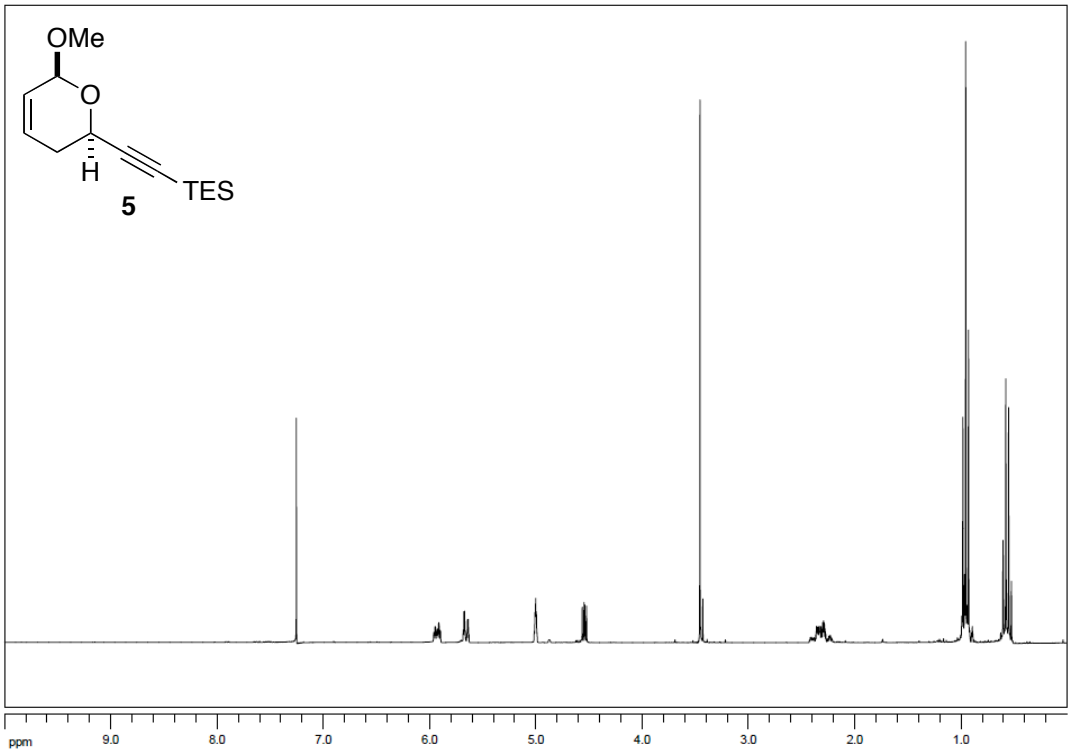
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O
H
TES
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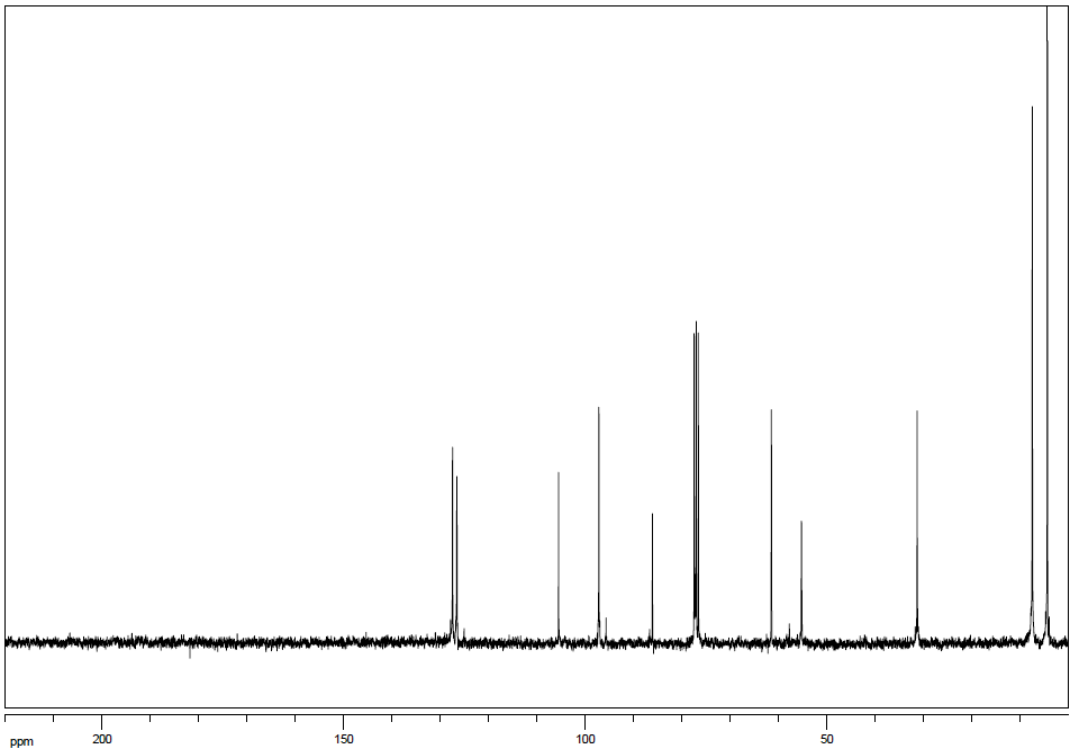
ppm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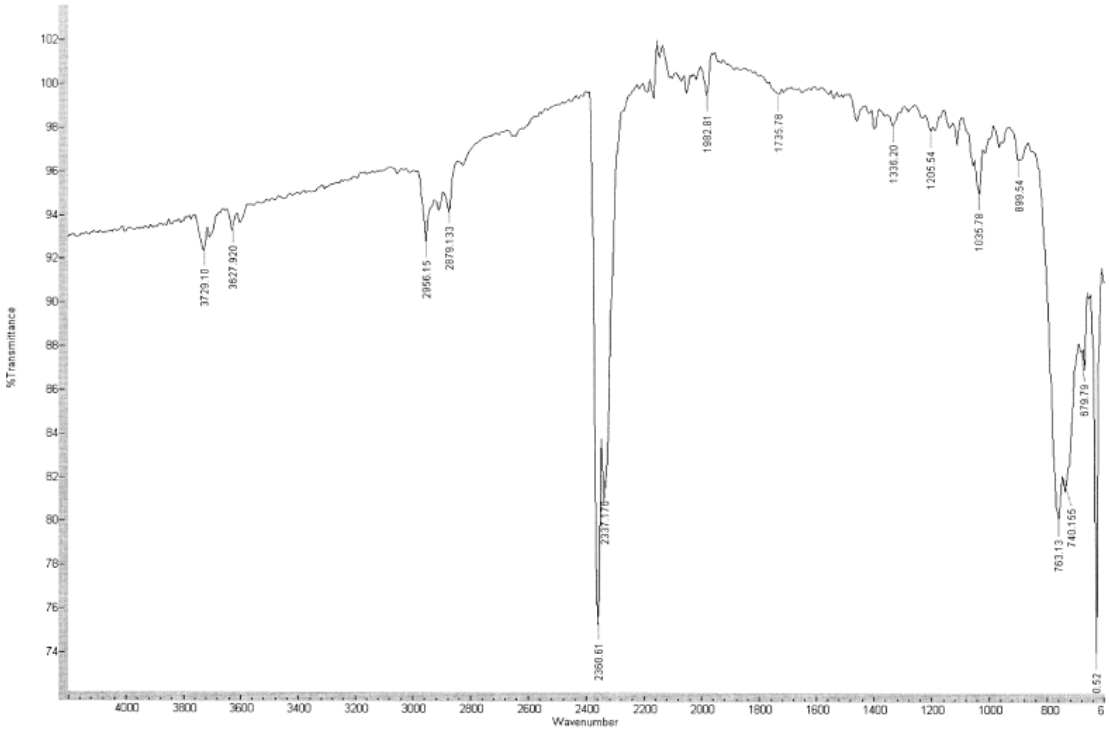
150
100
50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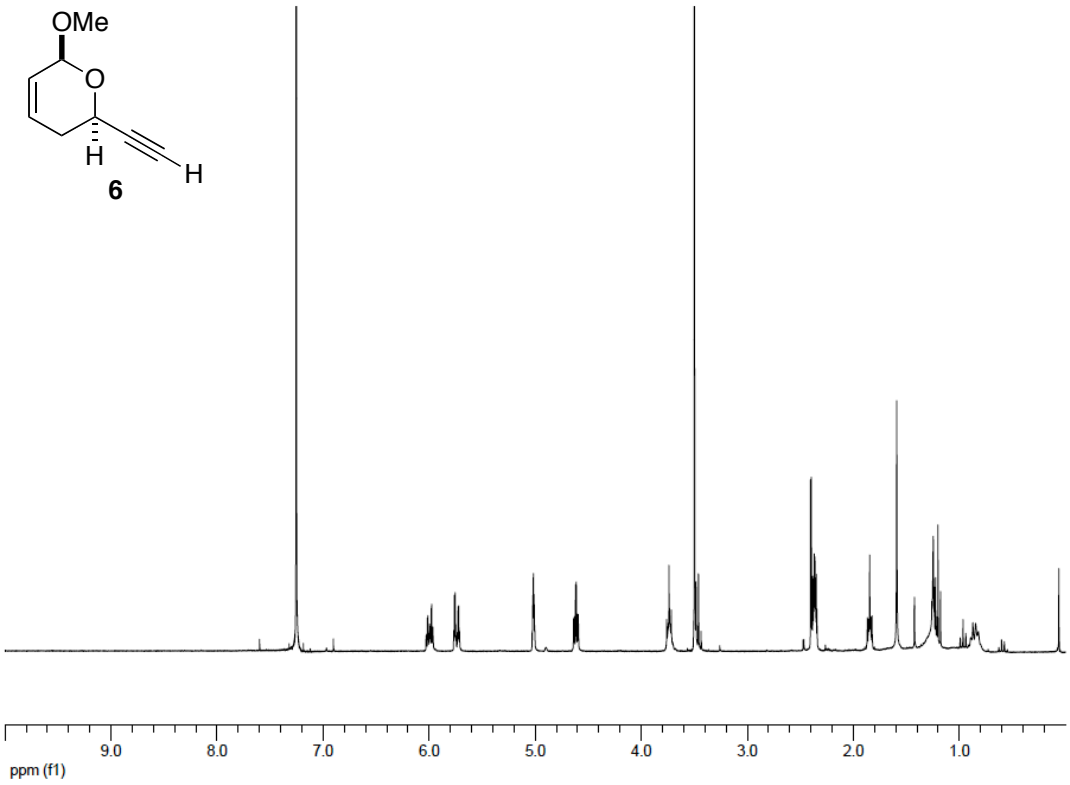
0.52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H
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3.0
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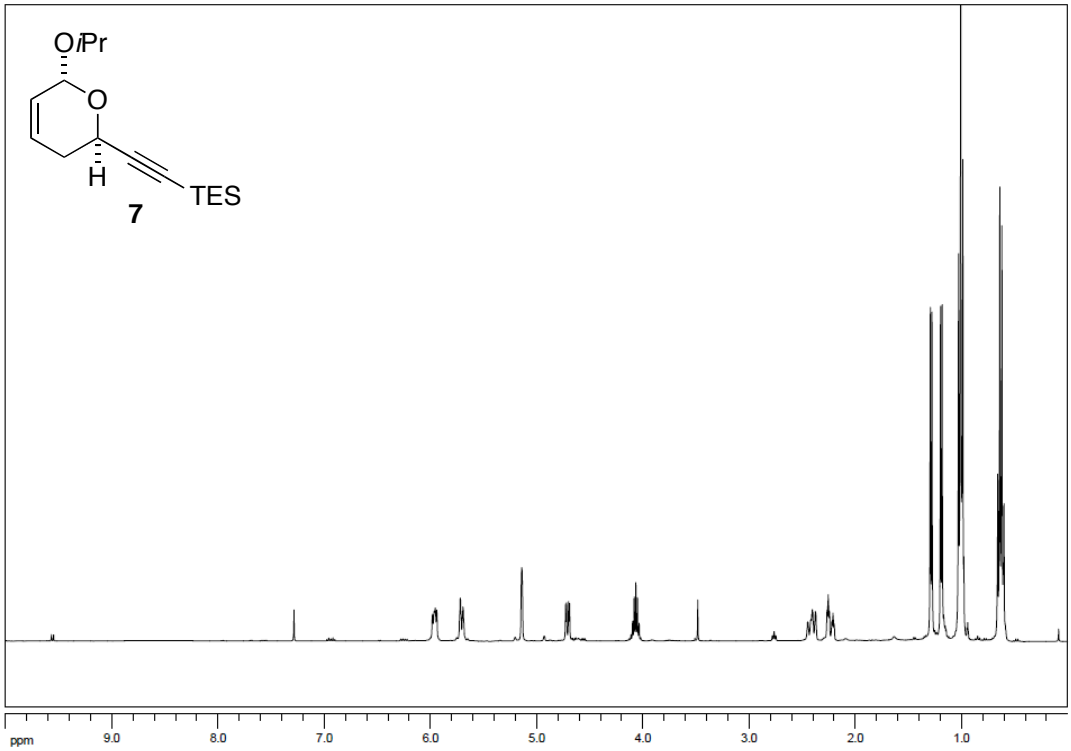
1.0
ppm (f1)


O*i*Pr
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ppm
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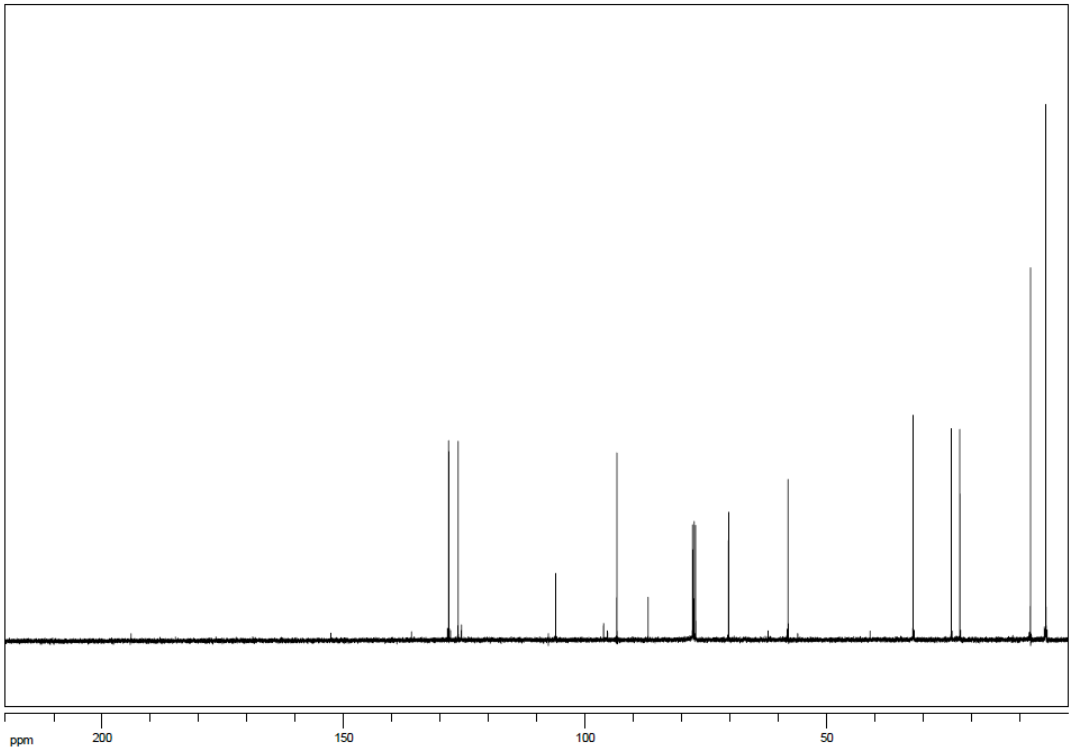
1.0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150
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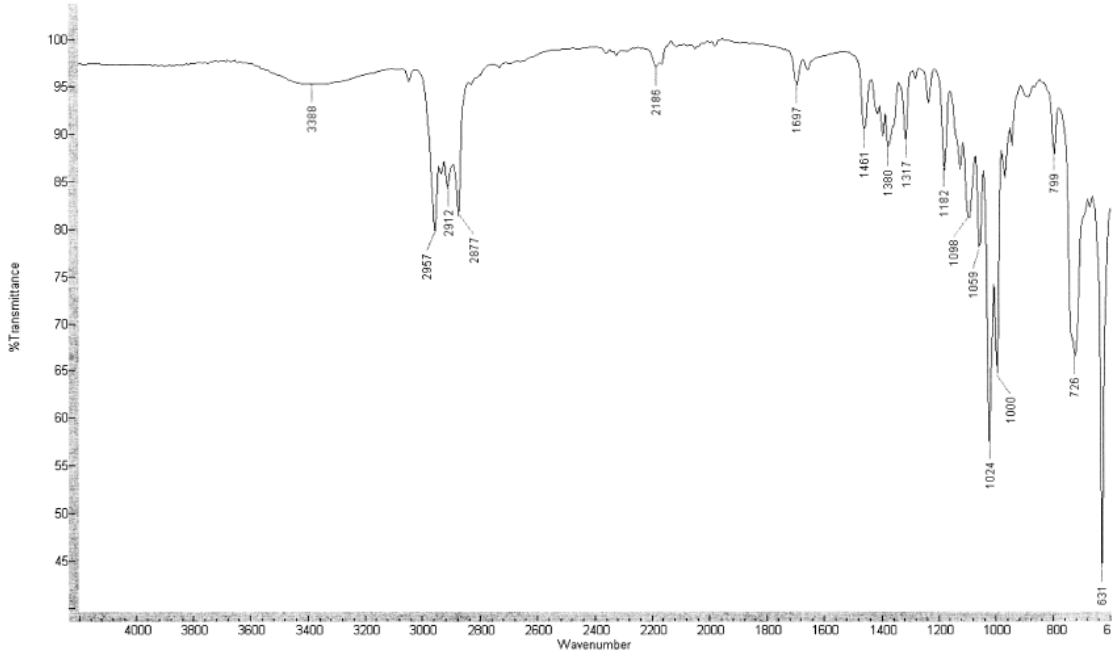
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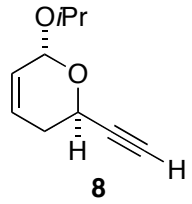
631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H
H

8

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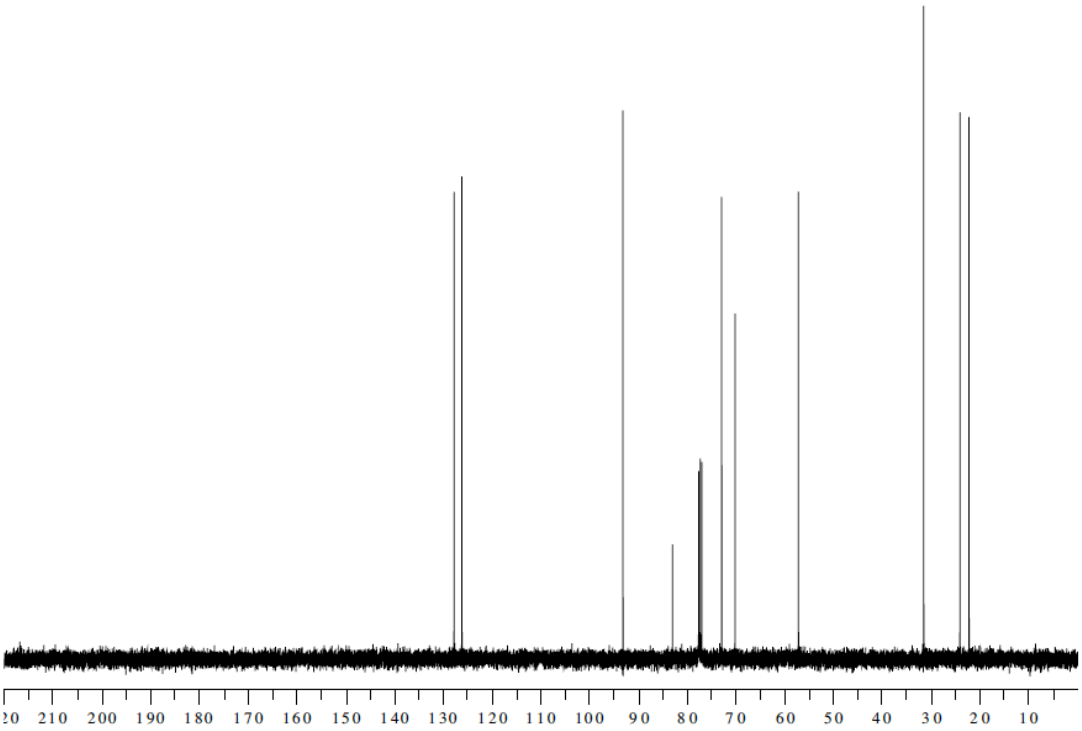
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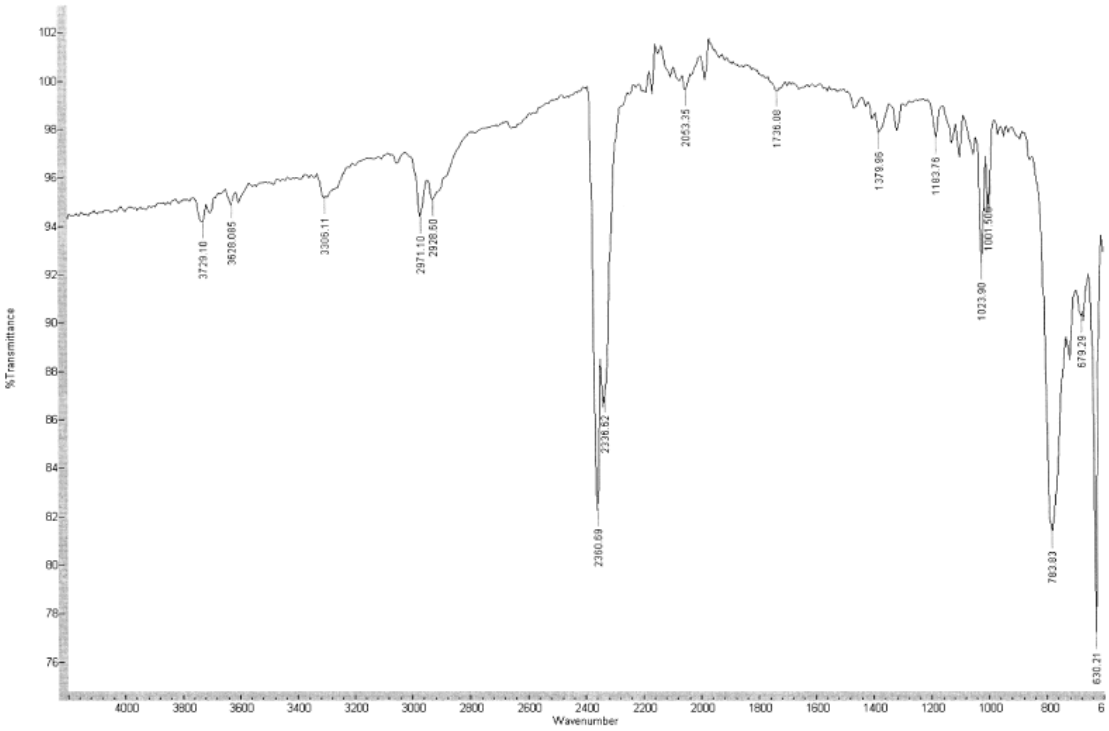
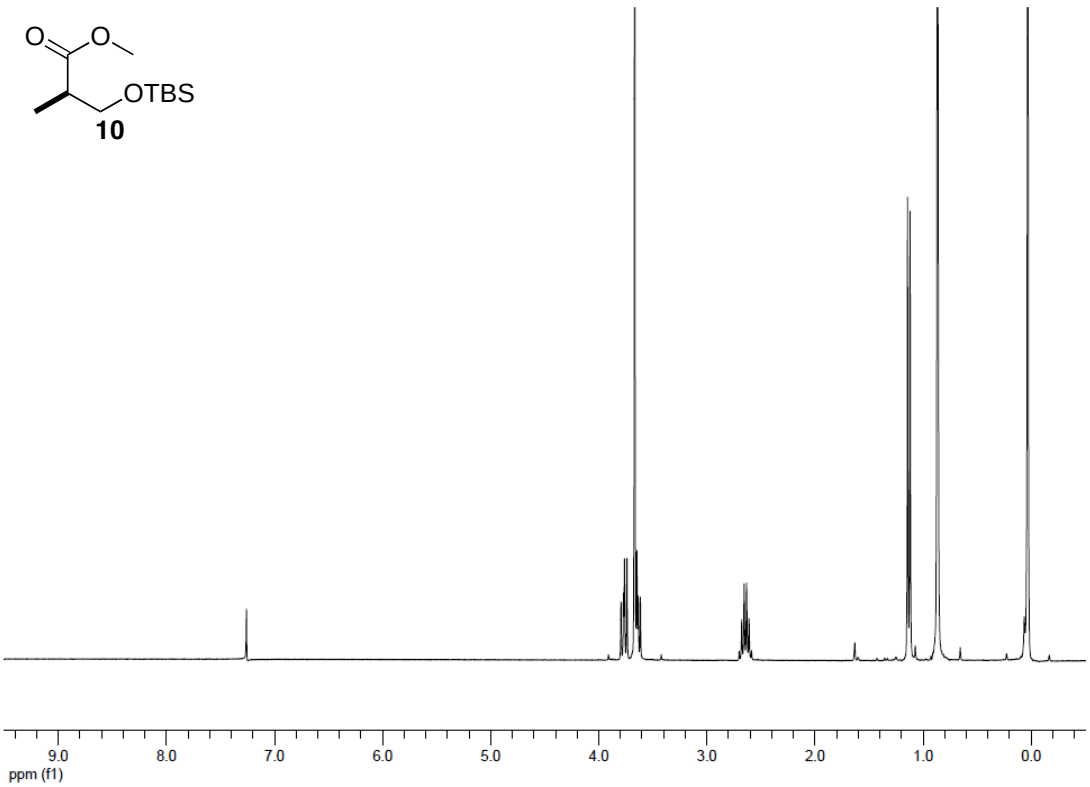

%Transmittance
Wavenumber
3729.10
3638.085
3306.11
2971.10
2928.60
2360.69
2336.62
2053.35
1736.08
1378.86
1183.78
1023.90
1001.50
879.29
783.83
630.21


O
O
OTBS
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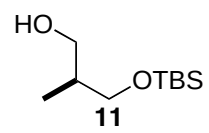
ppm (f1)


HO
OTBS

11

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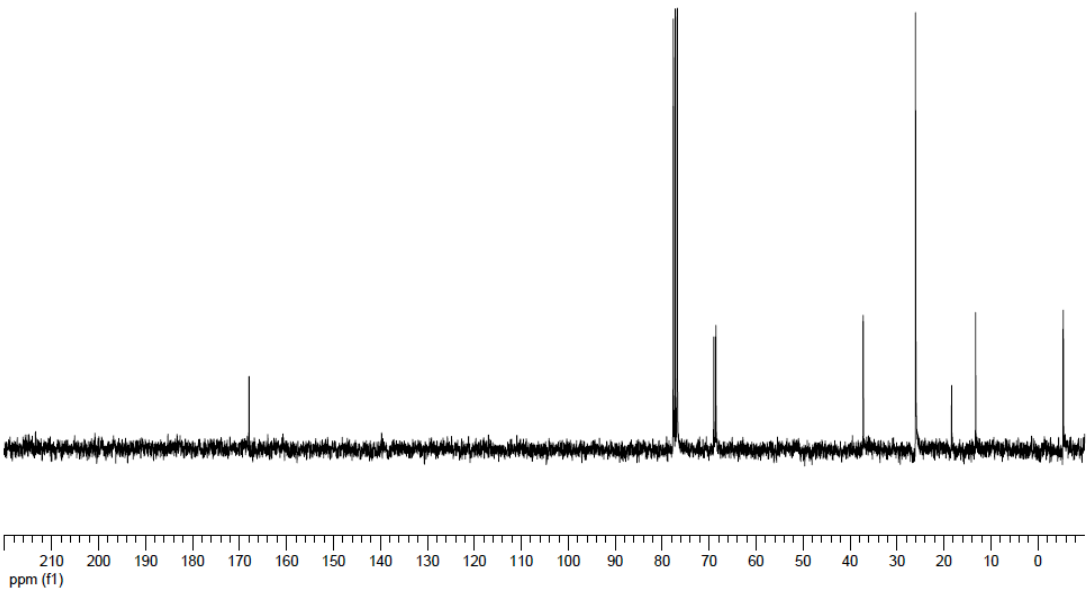
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)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200
190
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150
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110
100
90
80
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50
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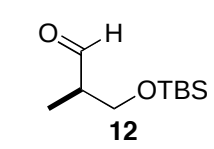
30
20
10
0
ppm (f1)


O
H
OTBS

12

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7.0
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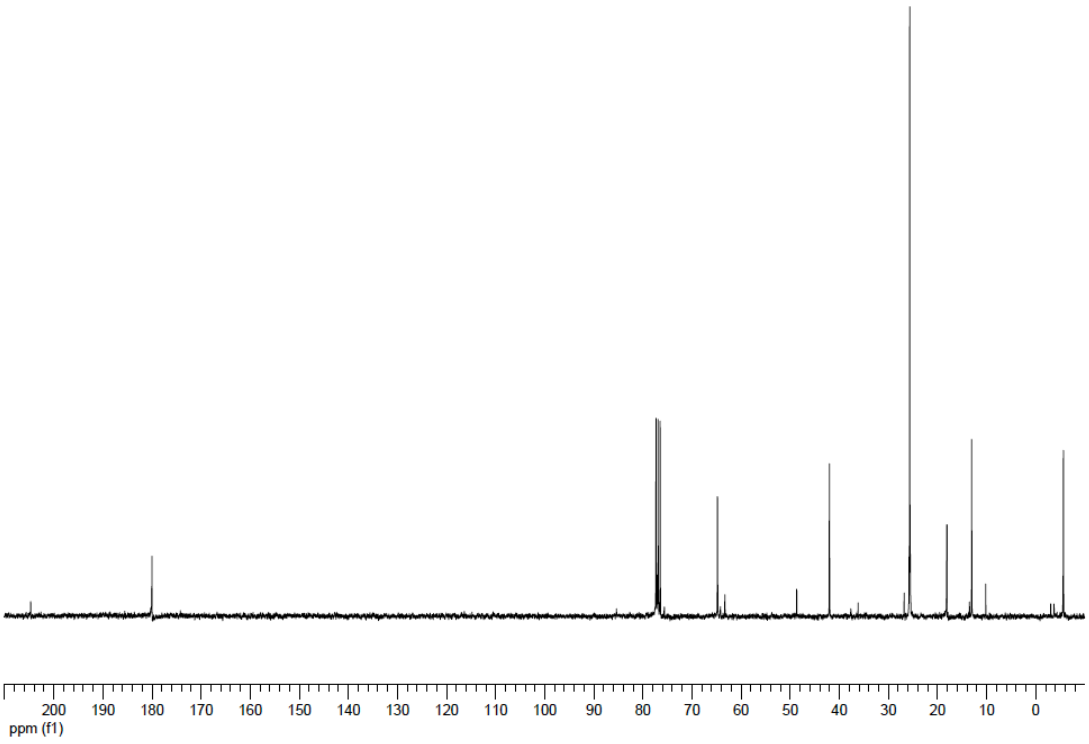
4.0
3.0
2.0
1.0
0.0
ppm (f1)


200
190
180
170
160
150
140
130
120
110
100
90
80
70
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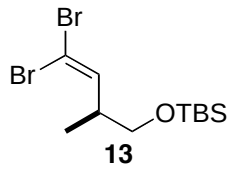
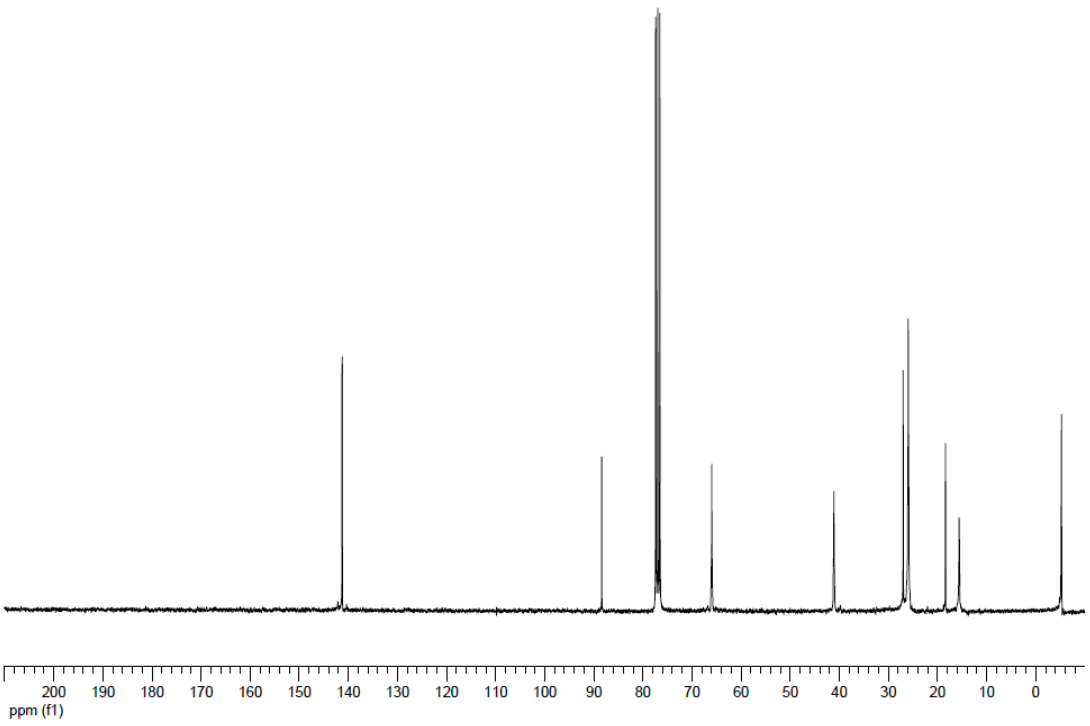
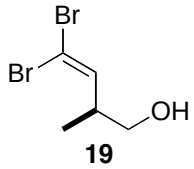
50
40
30
20
10
0
ppm (f1)


Br
Br
OTBS

13

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)


200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)


Br
Br
OH

19

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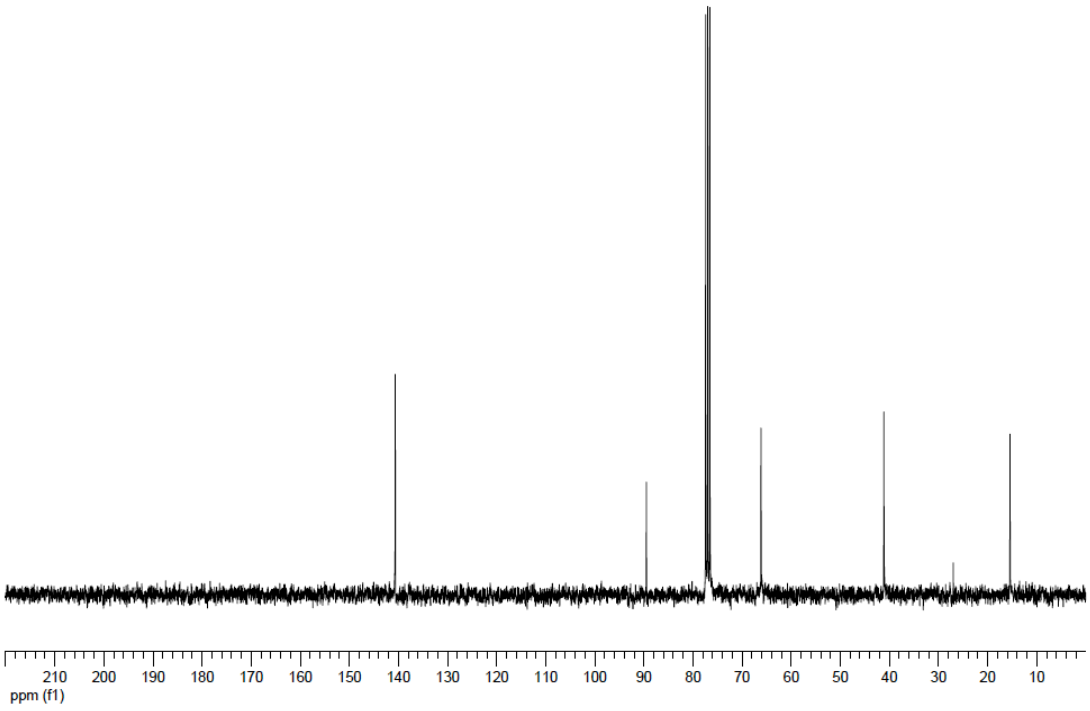
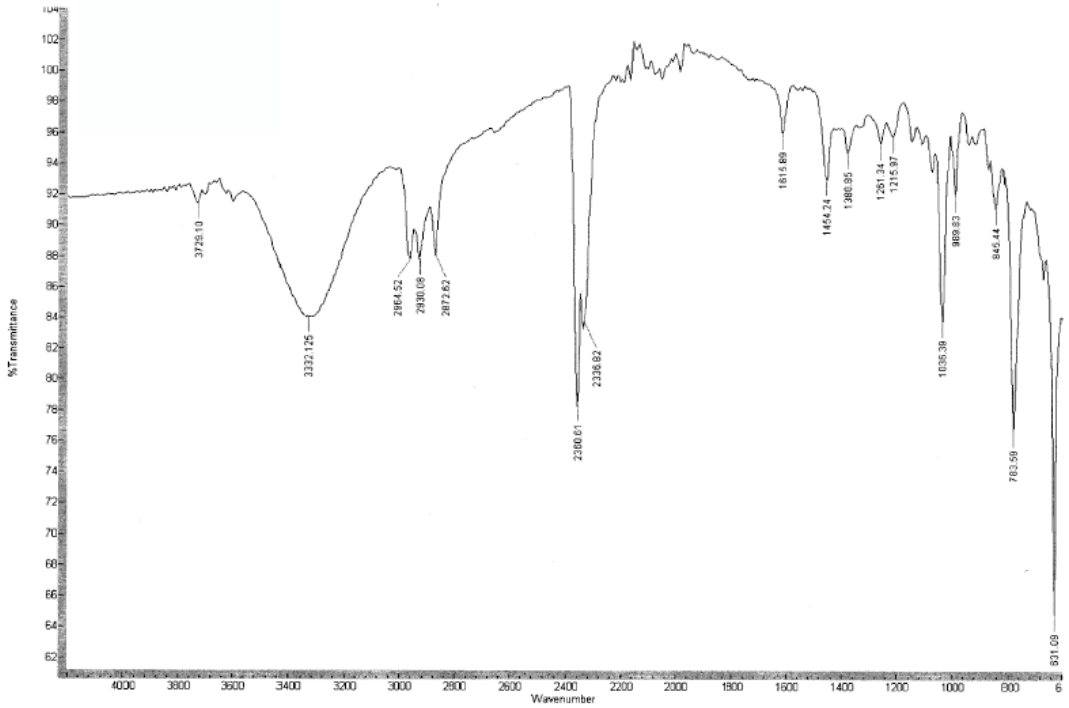
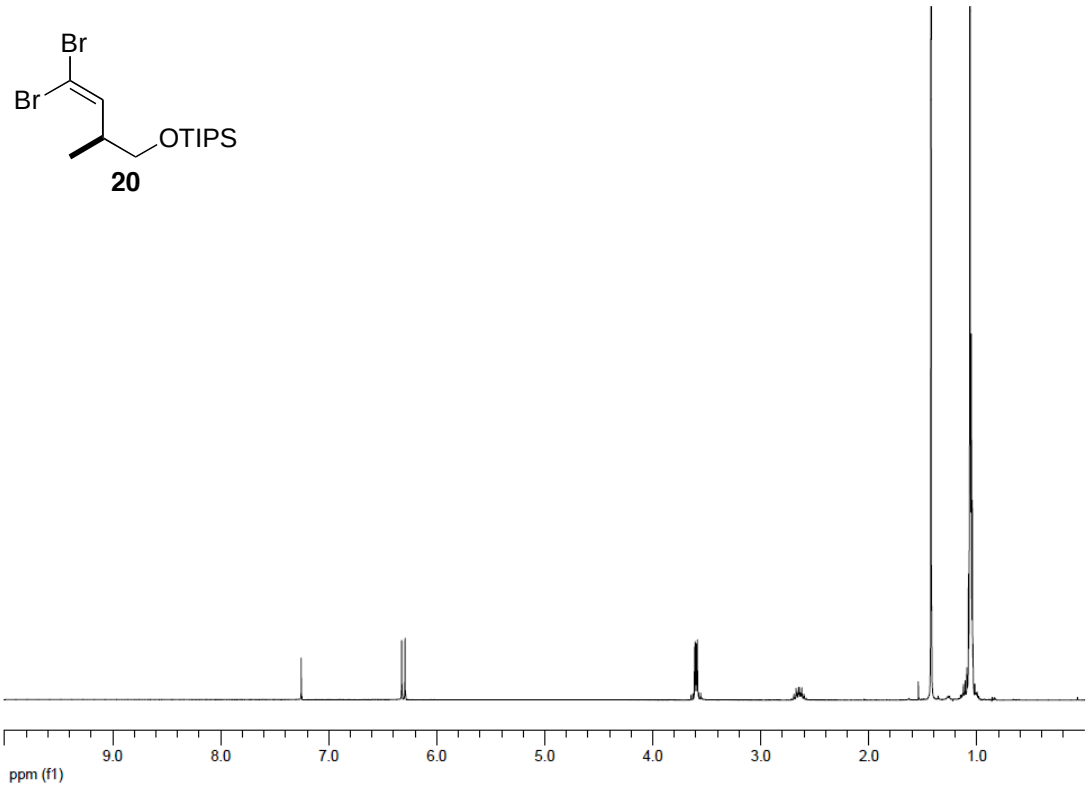
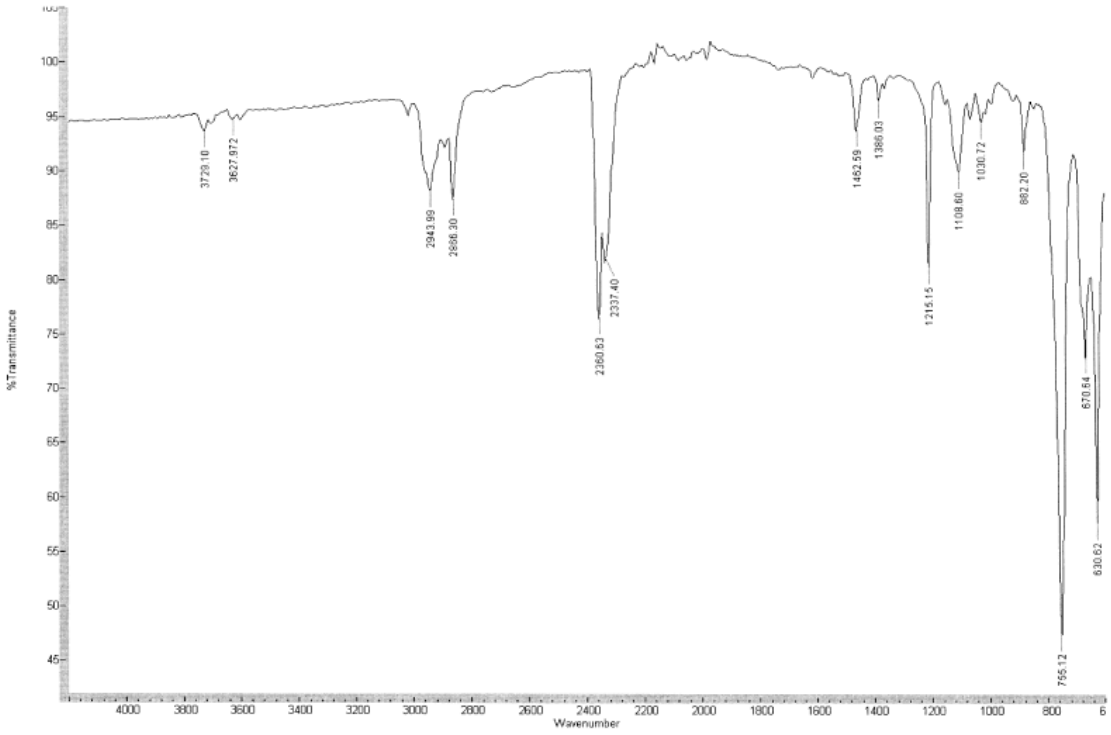
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)


210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm (f1)


%Transmittance
Wavenumber
3729.10
3332.125
2964.52
2930.08
2872.62
2360.61
2336.92
1615.89
1454.24
1380.95
1261.34
1215.97
1038.38
969.83
845.44
783.59
631.09


Br
Br
OTIPS
20
ppm (f1)


%Transmittance
Wavenumber
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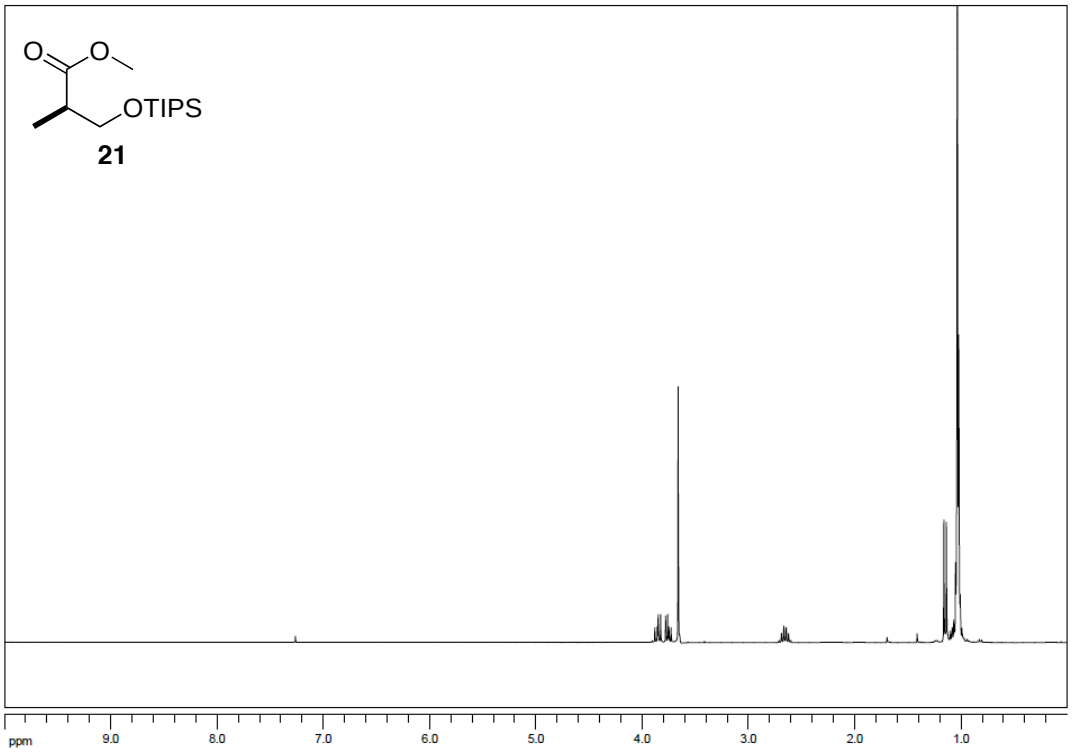
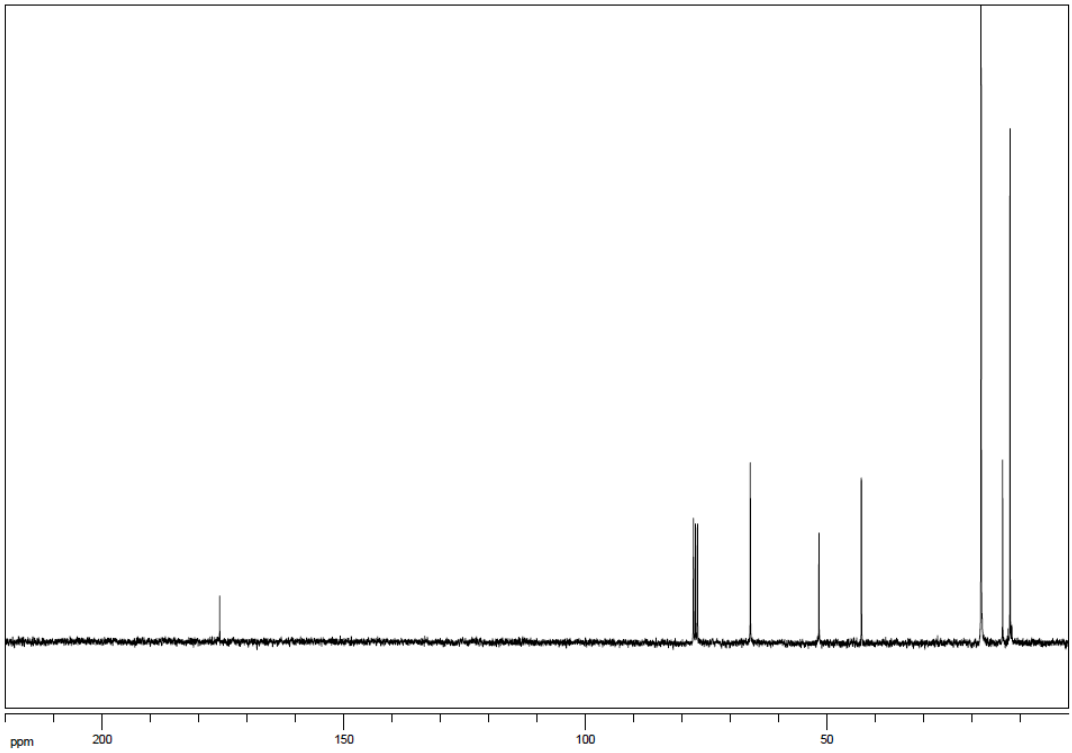
3627.972
2943.99
2866.30
2360.63
2337.40
1462.59
1386.03
1215.15
1108.60
1036.72
882.20
755.12
670.64
630.62


OTIPS
21
ppm


ppm
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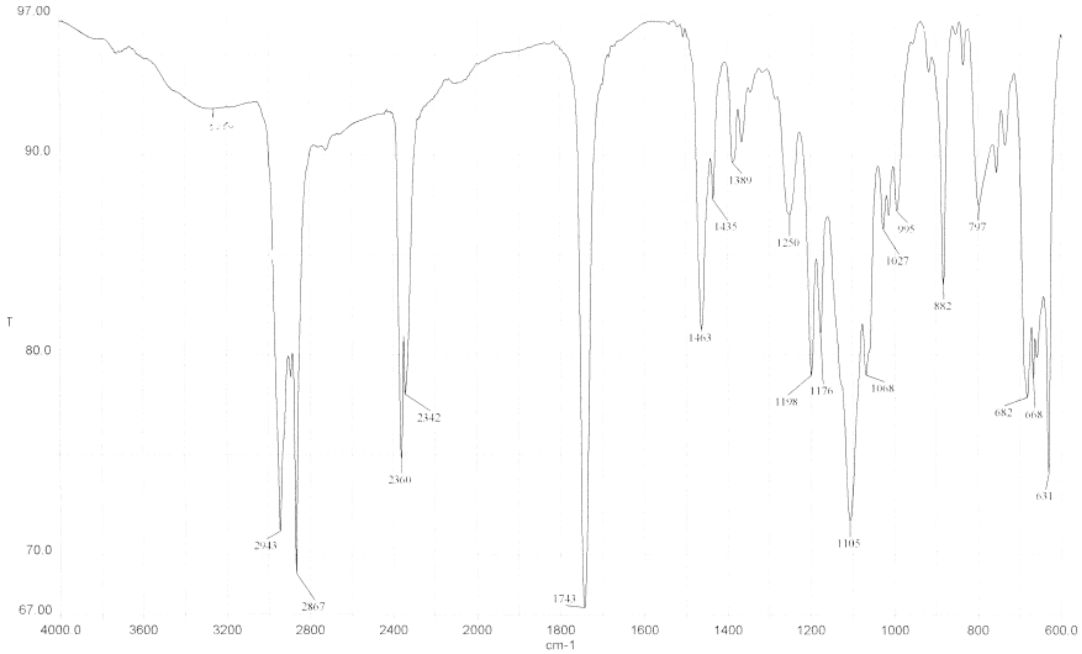
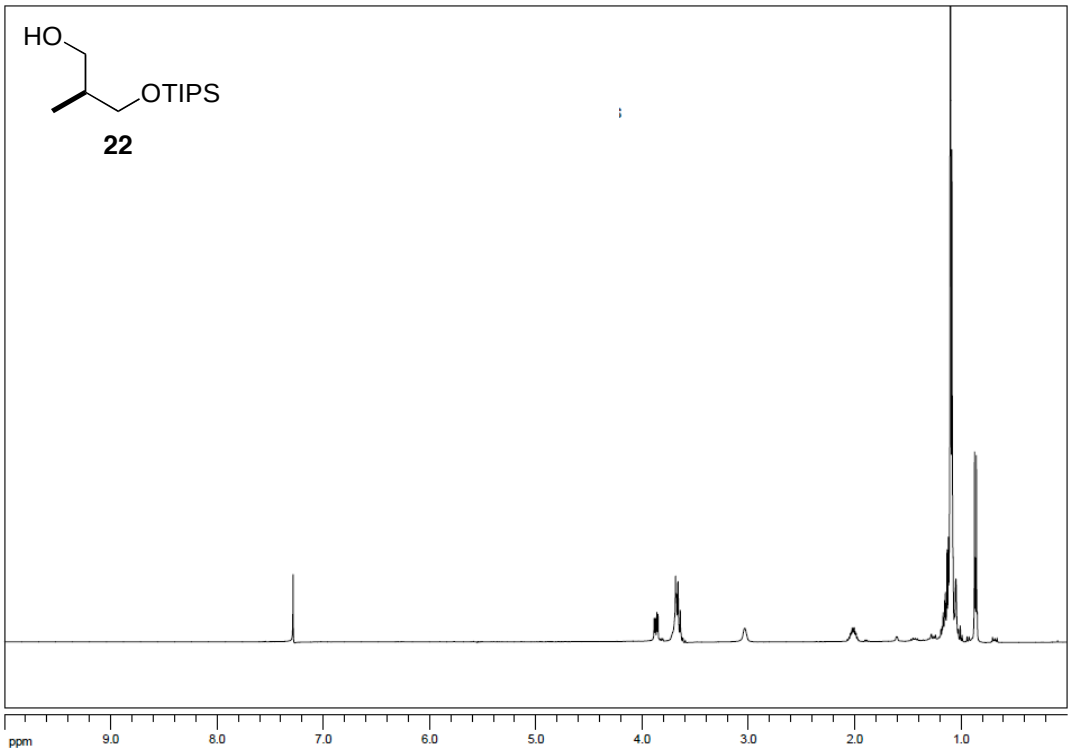
150
100
50


97.00
90.0
80.0
70.0
67.00
T
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1
2943
2867
2342
2360
1743
1463
1435
1389
1250
1198
1176
1105
1068
1027
995
882
797
682
668
631


HO
OTIPS
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ppm
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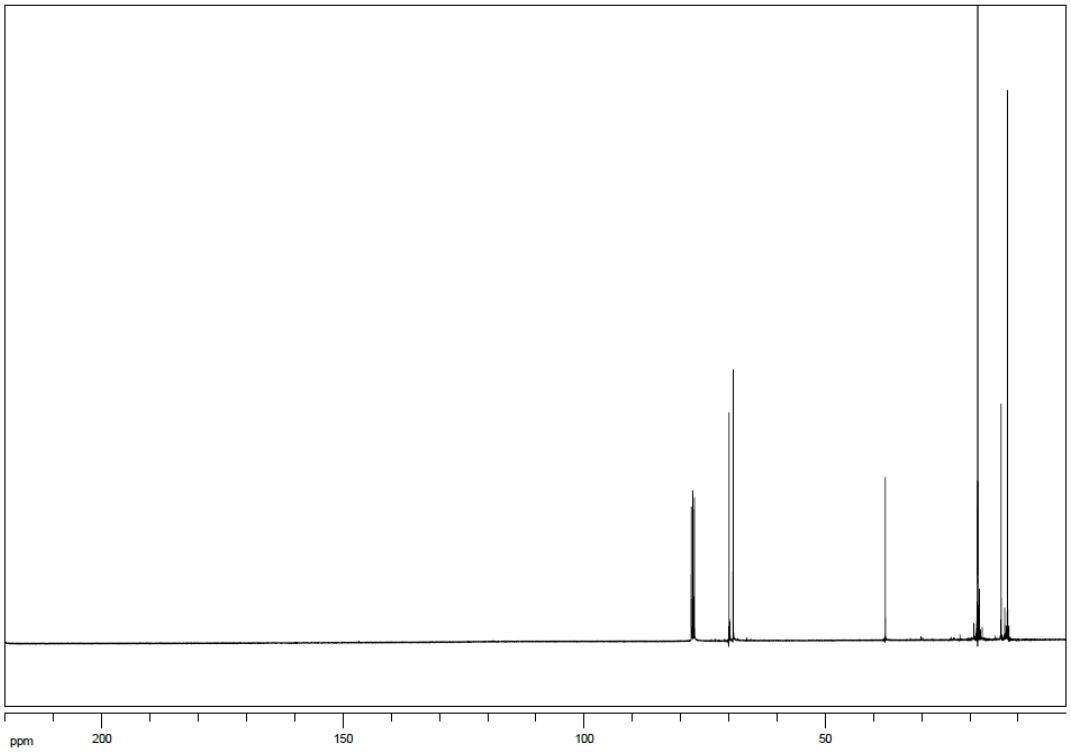
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
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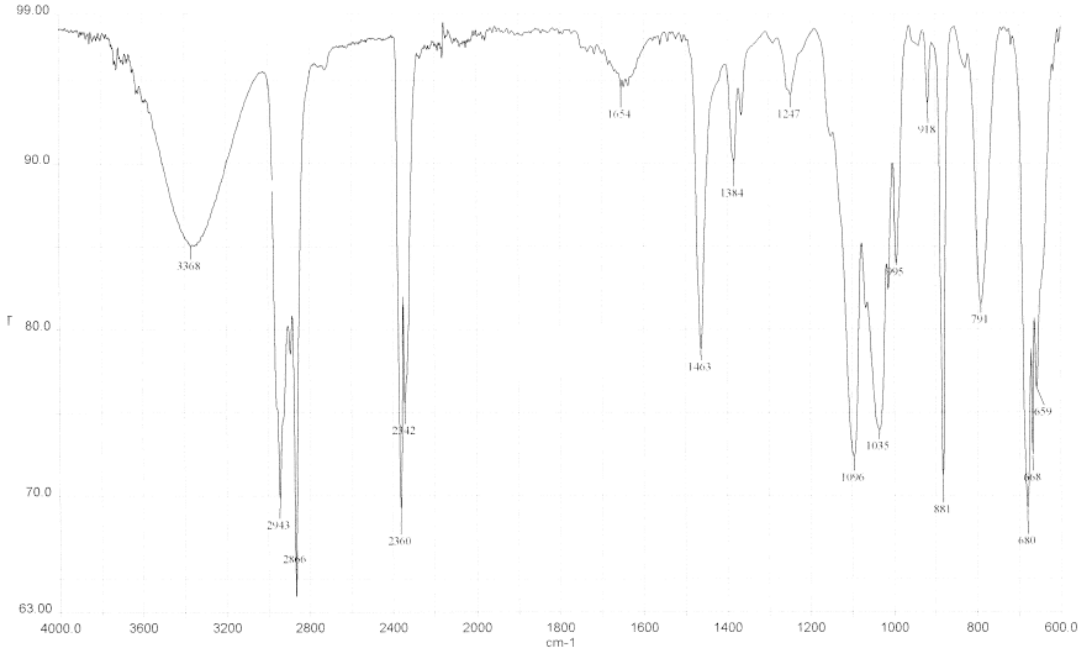
150
100
50


99.00
90.0
80.0
70.0
63.00
T
3368
2943
2866
2342
2360
1654
1463
1384
1247
1096
1035
995
918
881
791
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1400
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800
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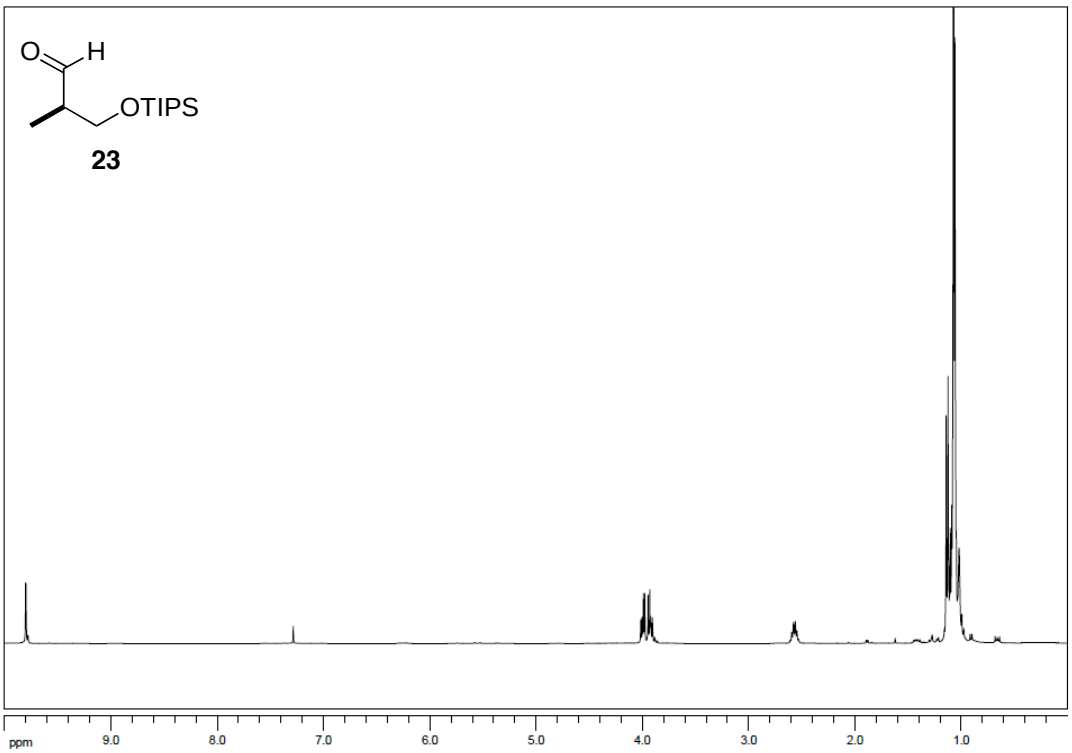
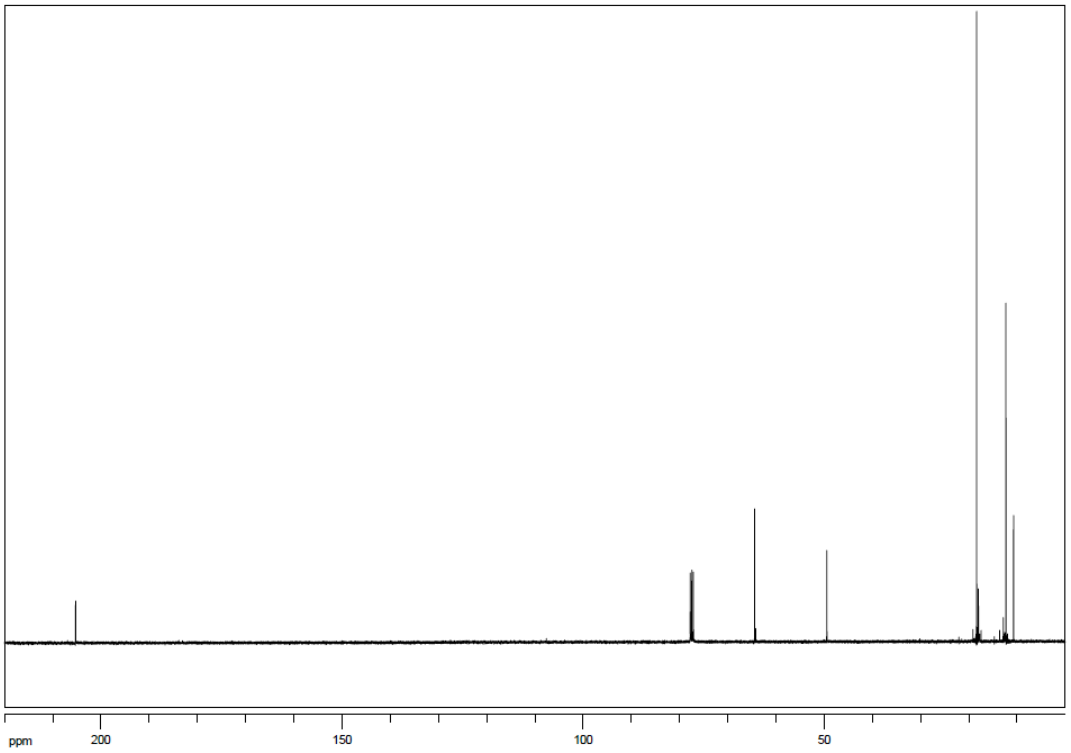
cm-1


O
H
OTIPS
23
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
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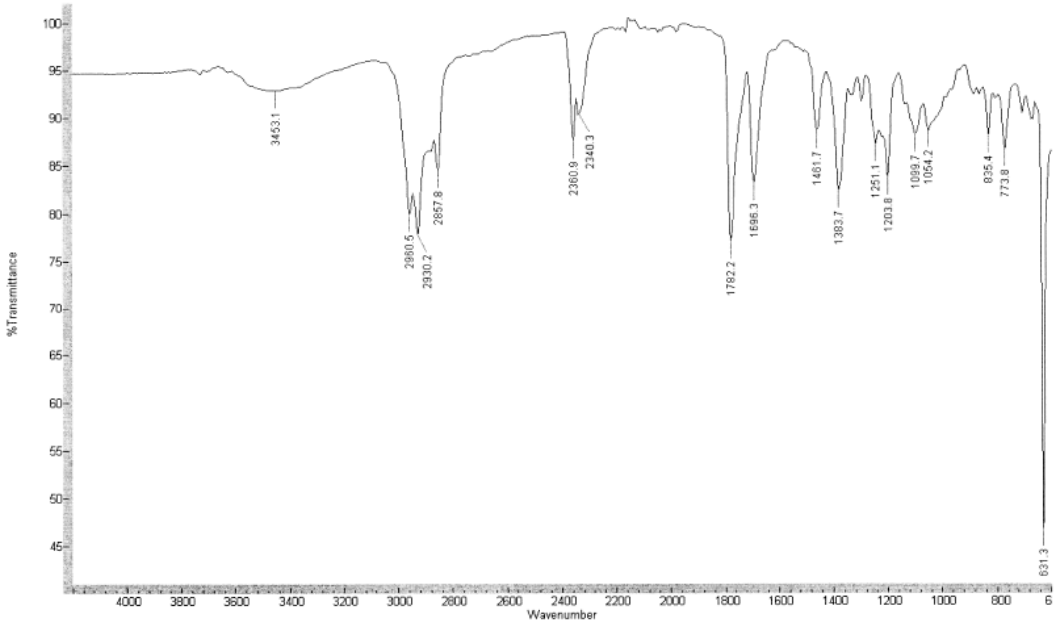
150
100
50


%Transmittance
Wavenumber
3453.1
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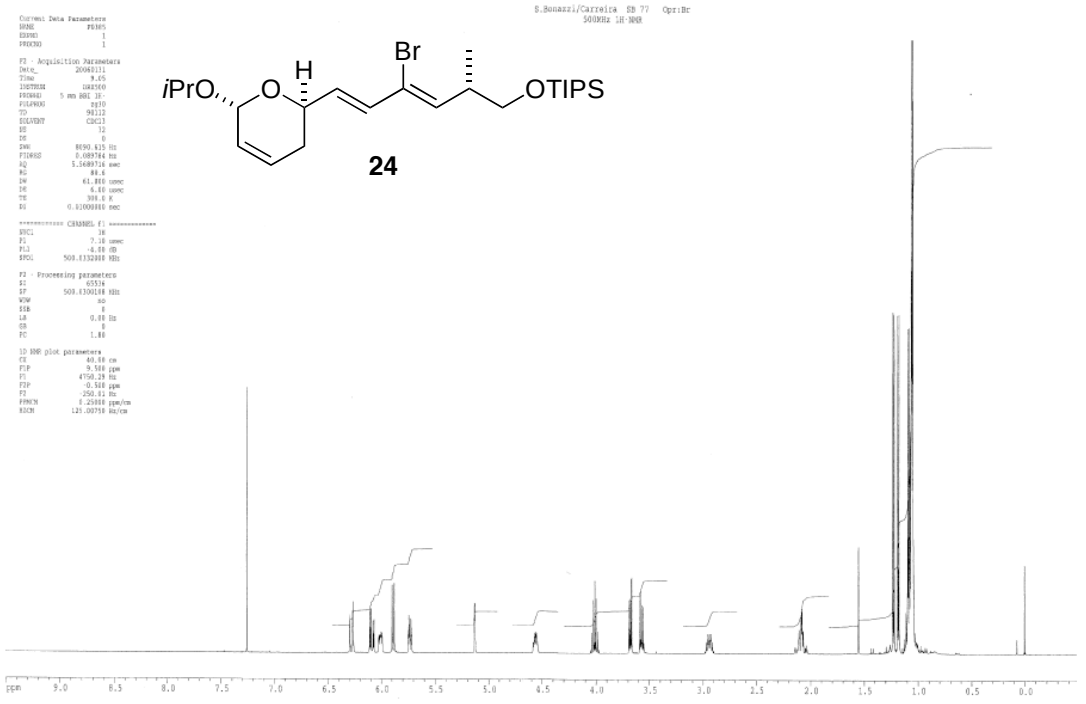
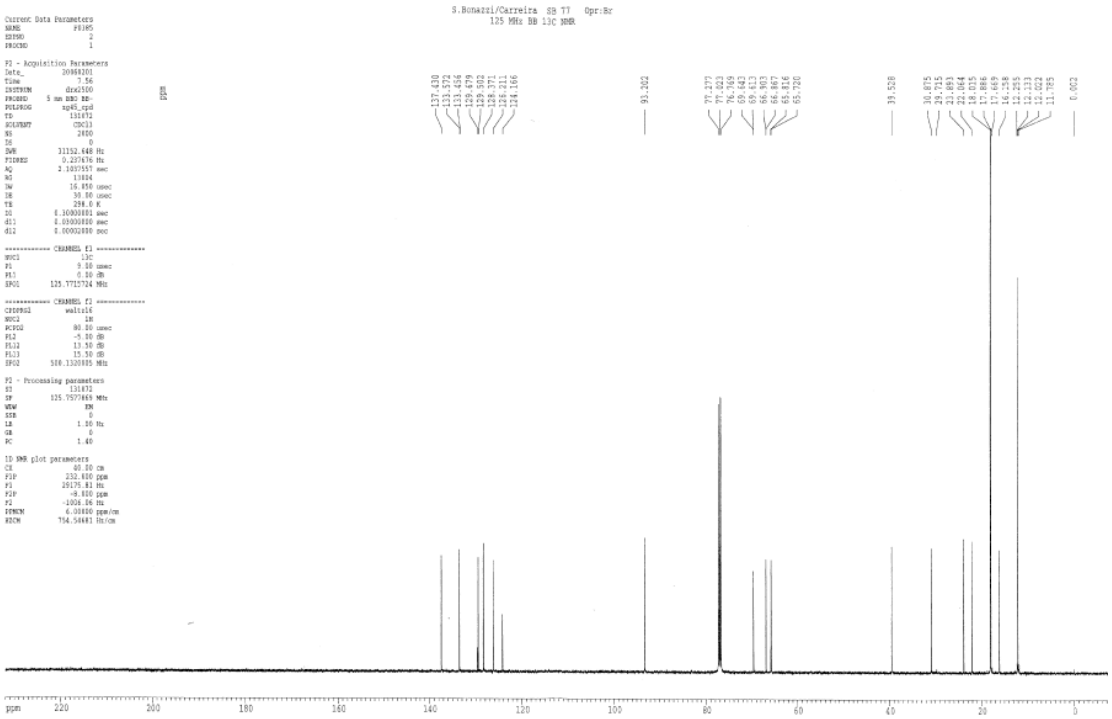
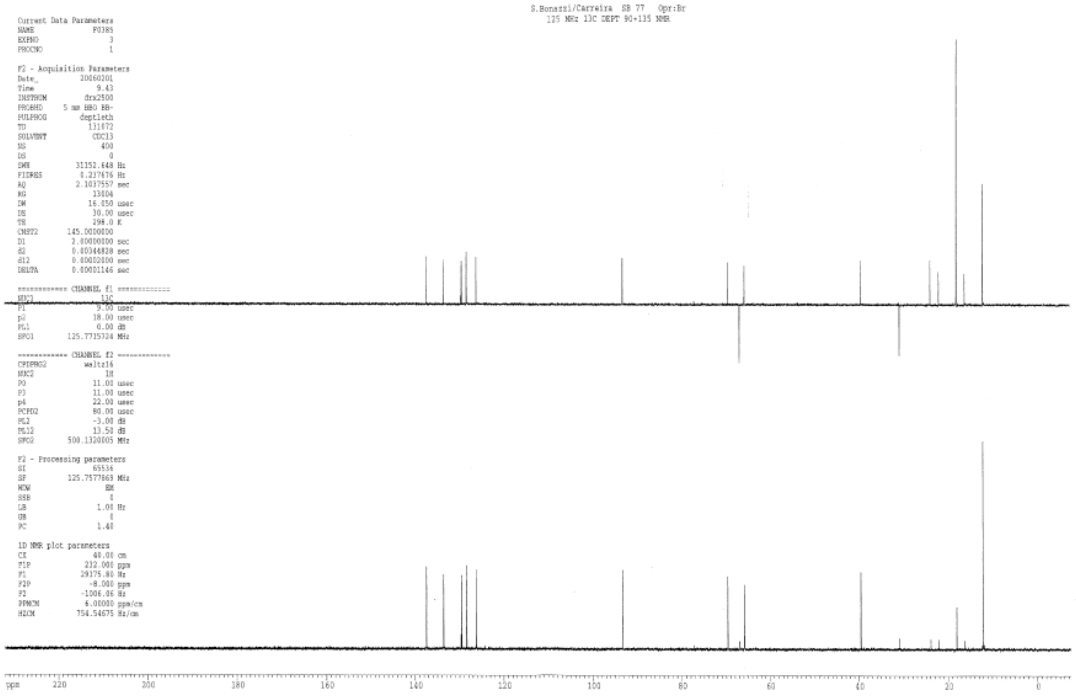
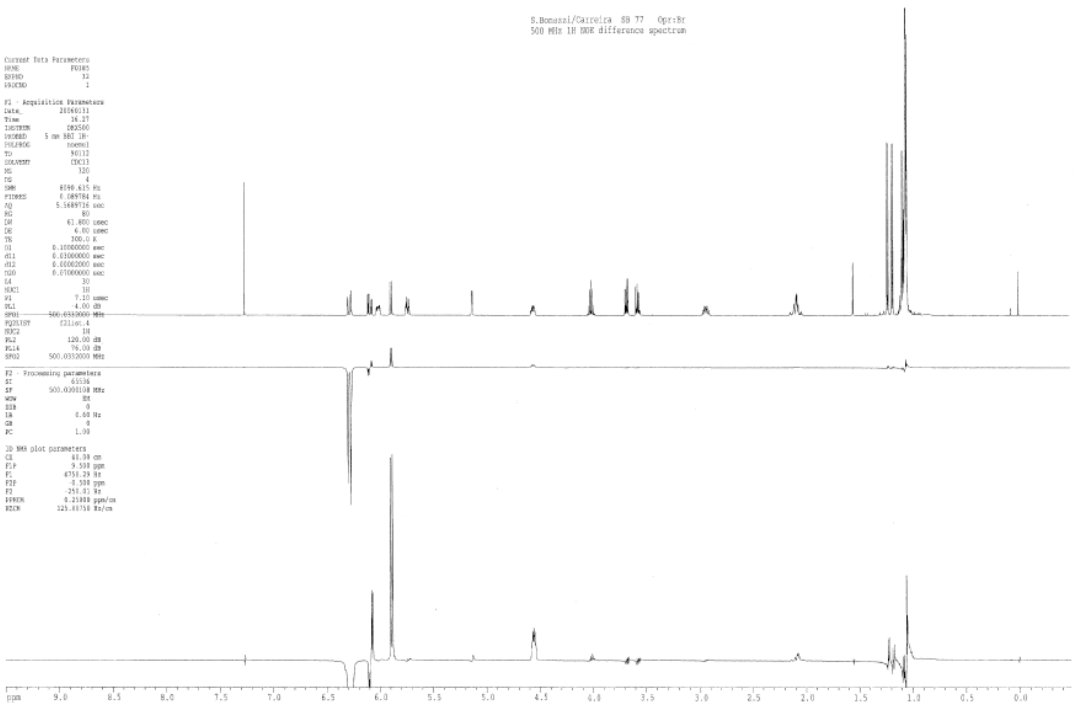
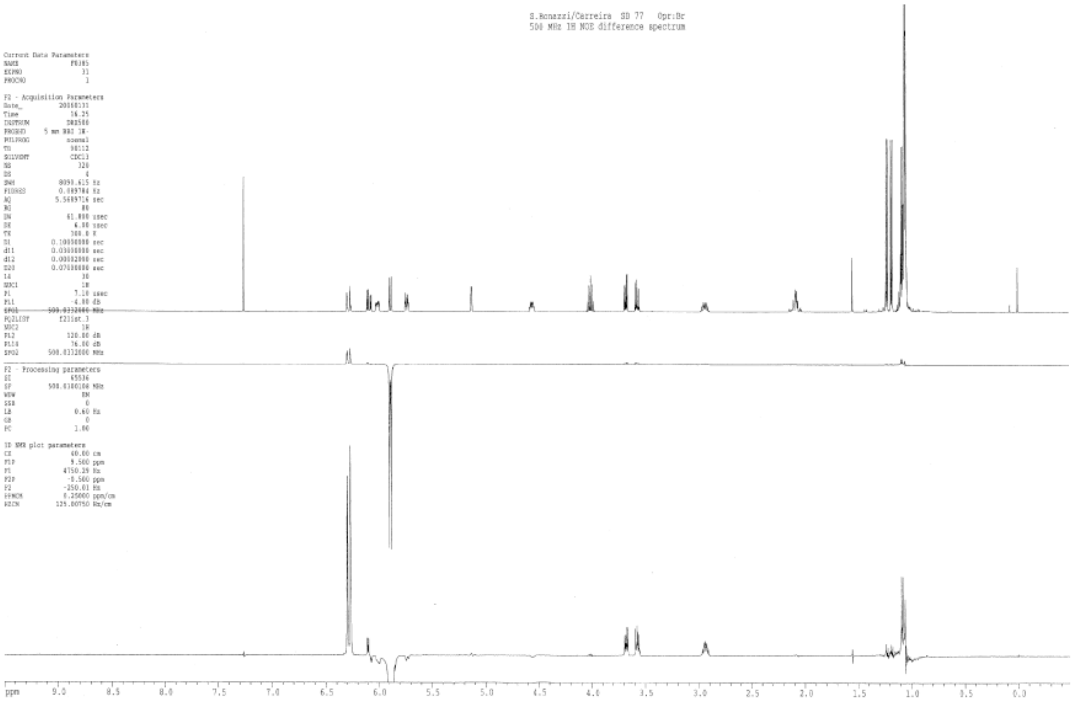
2930.2
2857.8
2360.9
2340.3
1782.2
1696.3
1461.7
1383.7
1251.1
1203.8
1099.7
1054.2
835.4
773.8
631.3

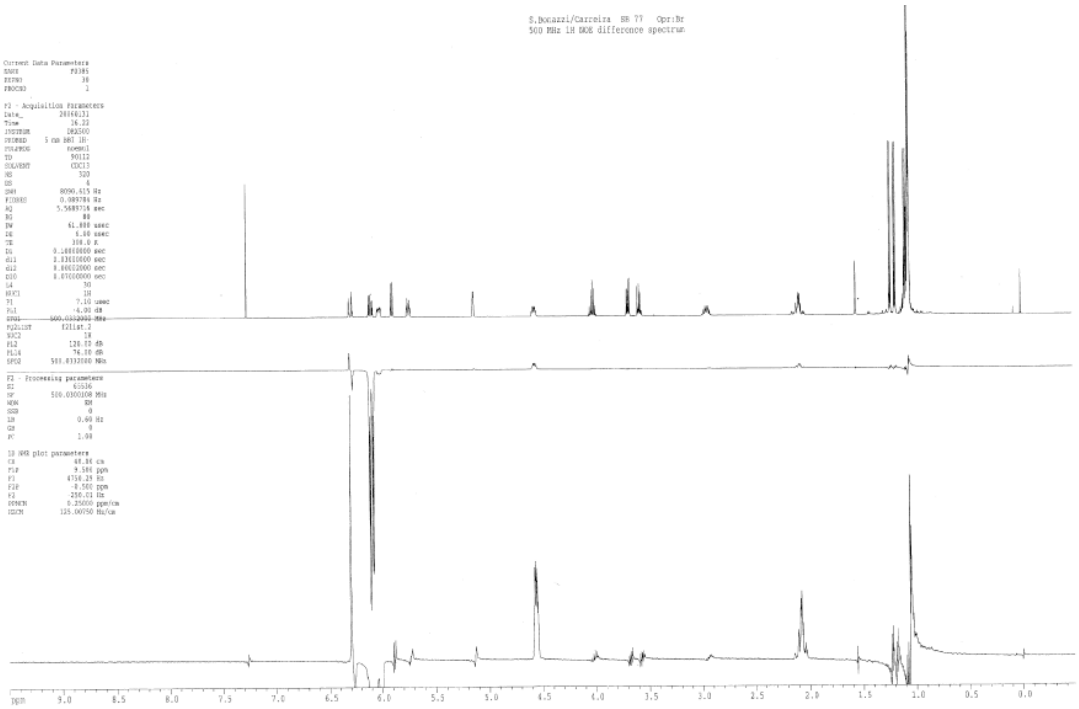

S.Bonazzi/Carreira SB 77 Opr:Br
500MHz 1H-NMR
Br
H
iPrO
O
OTIPS
24
ppm

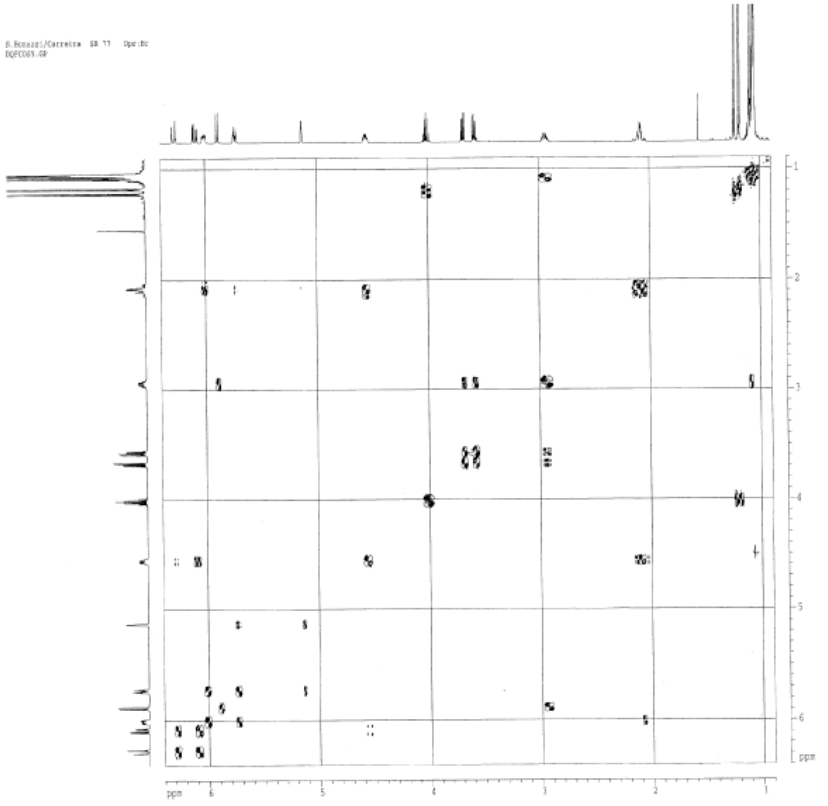

S.Bonazzi/Carreira SB 77 Opr:Br
125 MHz BB 13C NMR
ppm

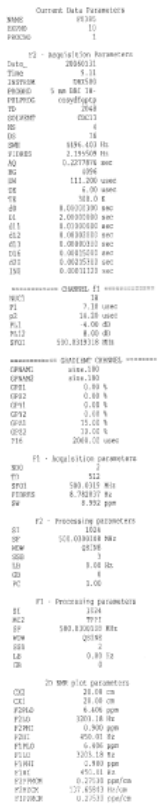

S.Bonazzi/Carreira SB 77 Opr:Br
125 MHz 13C DEPT 90+135 MHz
ppm

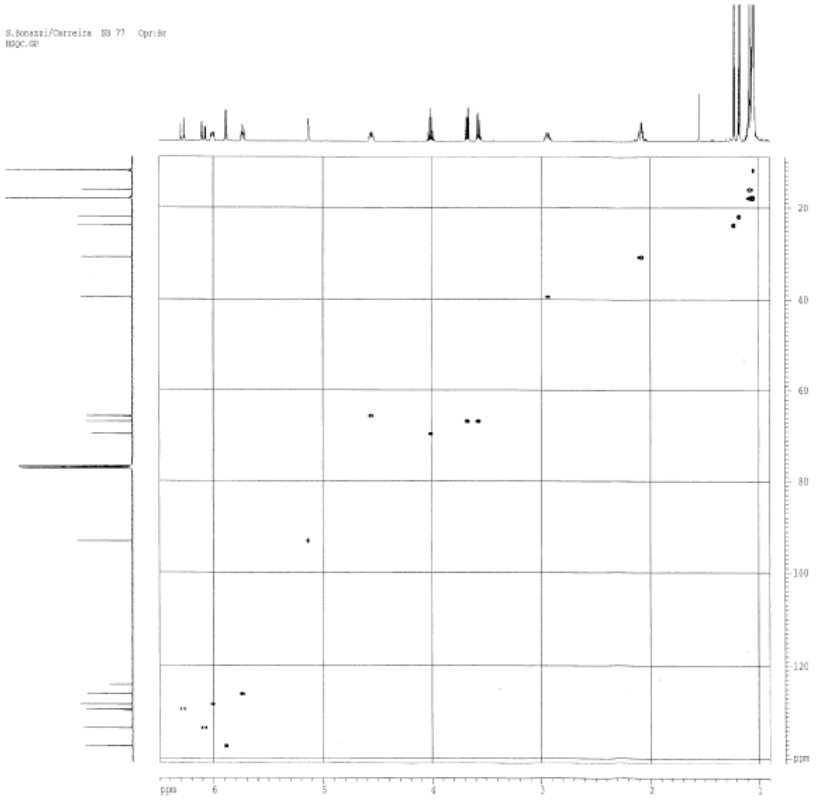

S.Bonazzi/Carreira SB 77 Opr:Br
500 MHz 1H NOE difference spectrum
ppm

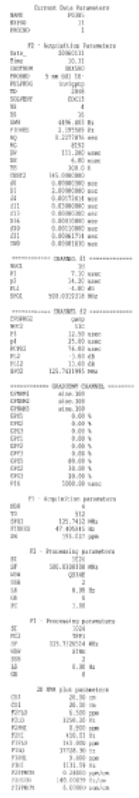

S.Bonazzi/Carreira SB 77 Opr:Br
500 MHz 1H NOE difference spectrum
ppm

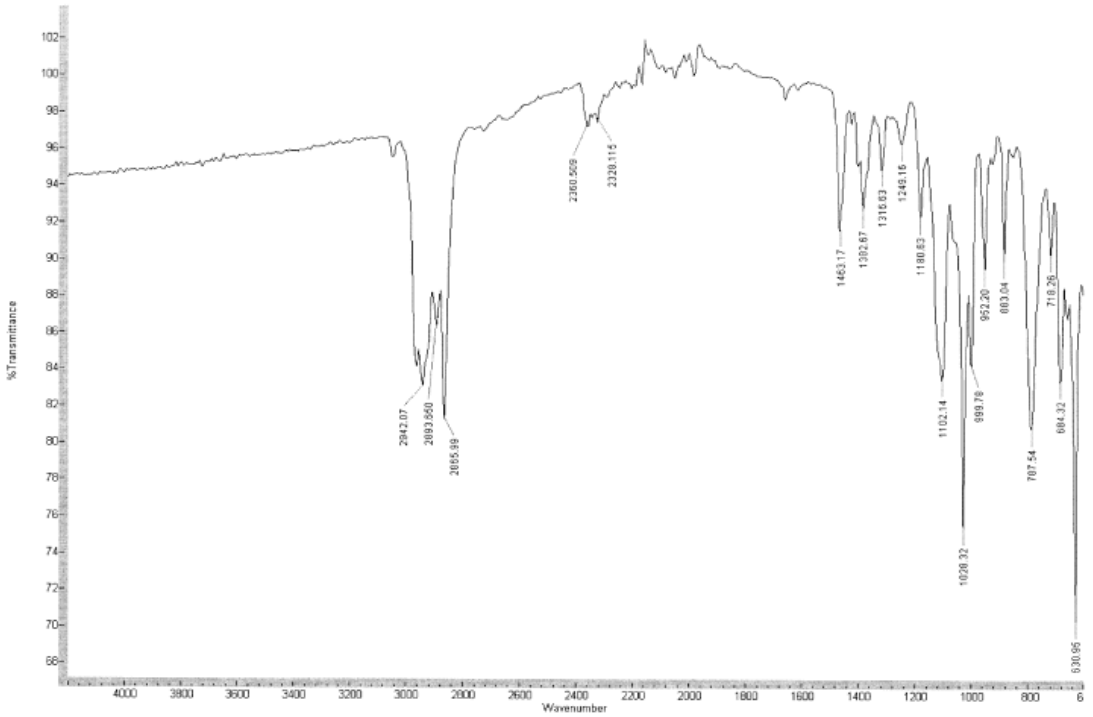

S.Bonazzi/Carreira SB 77 Opr:Br
500 MHz 1H NOE difference spectrum
ppm










%Transmittance
Wavenumber
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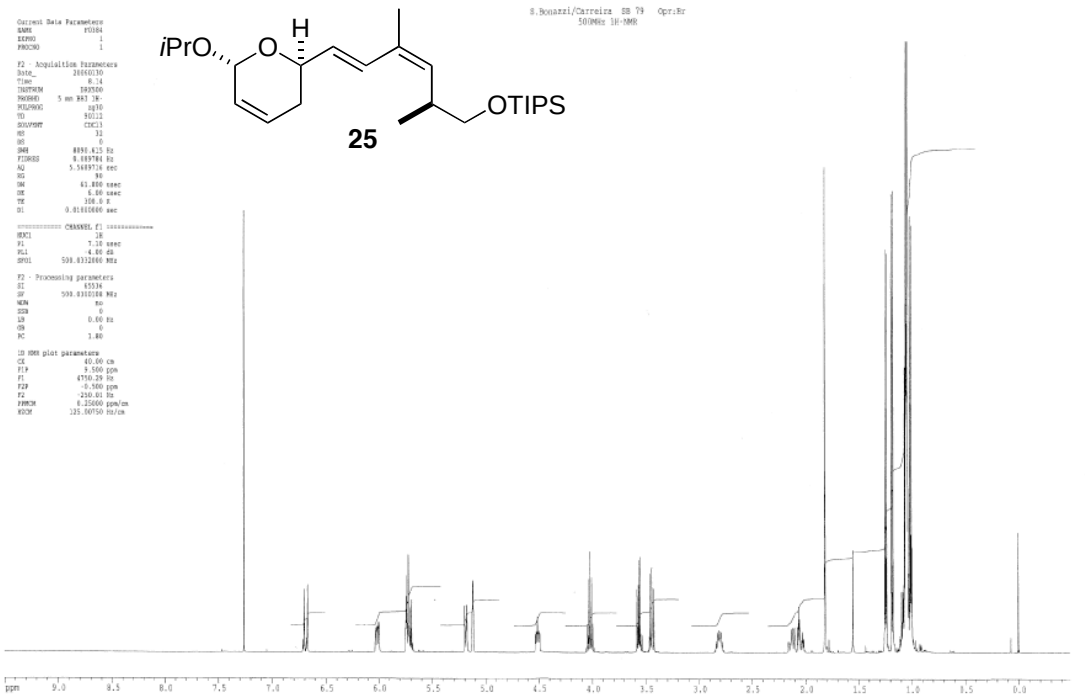
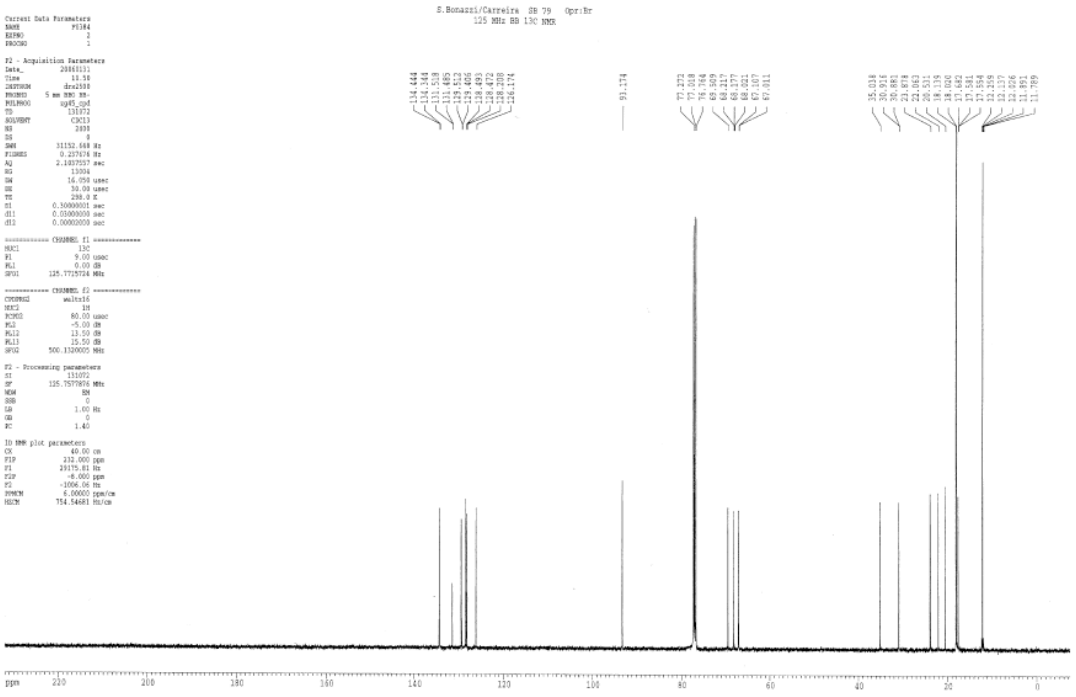
2665.99
2360.509
2328.115
1463.17
1382.67
1316.63
1249.16
1180.63
1102.14
1028.32
999.78
952.20
883.04
818.26
707.54
684.32
630.95


iPrO
O
H
OTIPS
25


S.Bonazzi/Carreira SB 79 Opr:Br
125 MHz BB 13C NMR
Current Data Parameters
NAME P1184
EXPNO 2
PROCNO 1
F2 - Acquisition Parameters
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Time 11.16
INSTRUM drx2500
PROBHD 5 mm BBO BB-
PULPROG zg45_cpd
TD 131072
SOLVENT CDCl3
NS 2400
DS 0
SWH 31152.648 Hz
FIDRES 0.237676 Hz
AQ 2.1037557 sec
RG 13004
DW 16.050 usec
DE 30.00 usec
TE 298.0 K
D1 0.50000001 sec
d11 0.03000000 sec
d12 0.00002000 sec
========== CHANNEL f1 ==========
NUC1 13C
P1 9.00 usec
PL1 0.00 dB
SFO1 125.7715724 MHz
========== CHANNEL f2 ==========
CPDPRG2 waltz16
NUC2 1H
PCPD2 80.00 usec
PL2 -5.00 dB
PL12 13.50 dB
PL13 15.50 dB
SFO2 500.1320005 MHz
F2 - Processing parameters
SI 131072
SF 125.7577876 MHz
WDW EM
SSB 0
LB 1.00 Hz
GB 0
PC 1.40
1D NMR plot parameters
CX 40.00 cm
F1P 232.000 ppm
F1 29175.81 Hz
F2P -8.000 ppm
F2 -1006.06 Hz
PPMCM 6.00000 ppm/cm
HZCM 754.54681 Hz/cm
14.444
14.344
131.528
128.455
126.923
126.406
126.493
126.472
126.288
126.174
93.174
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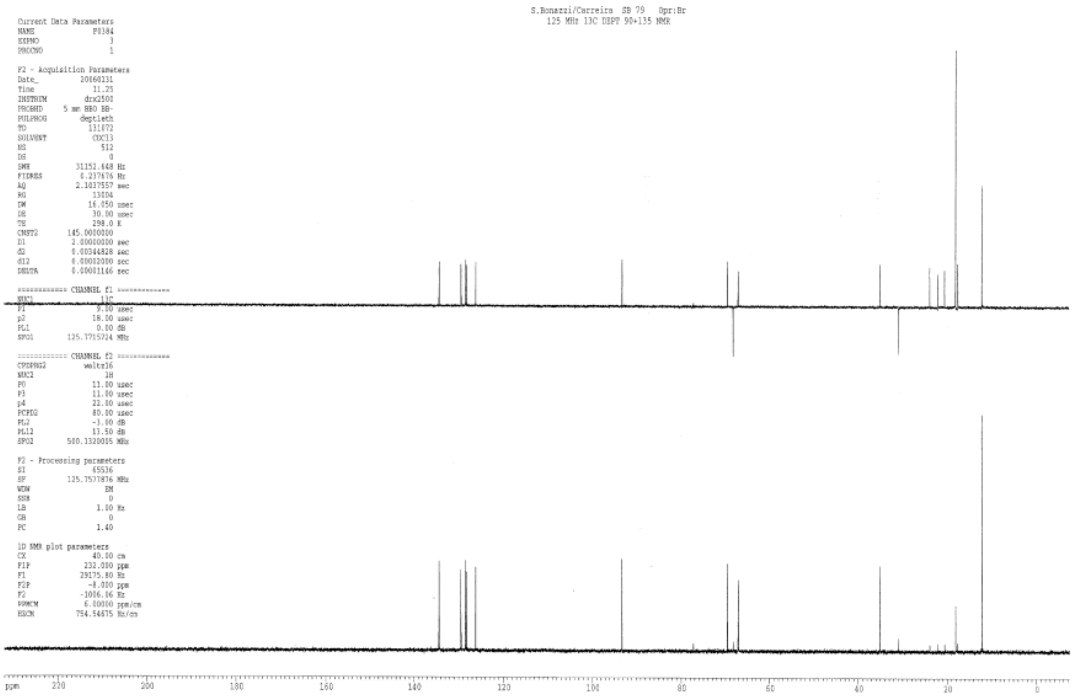
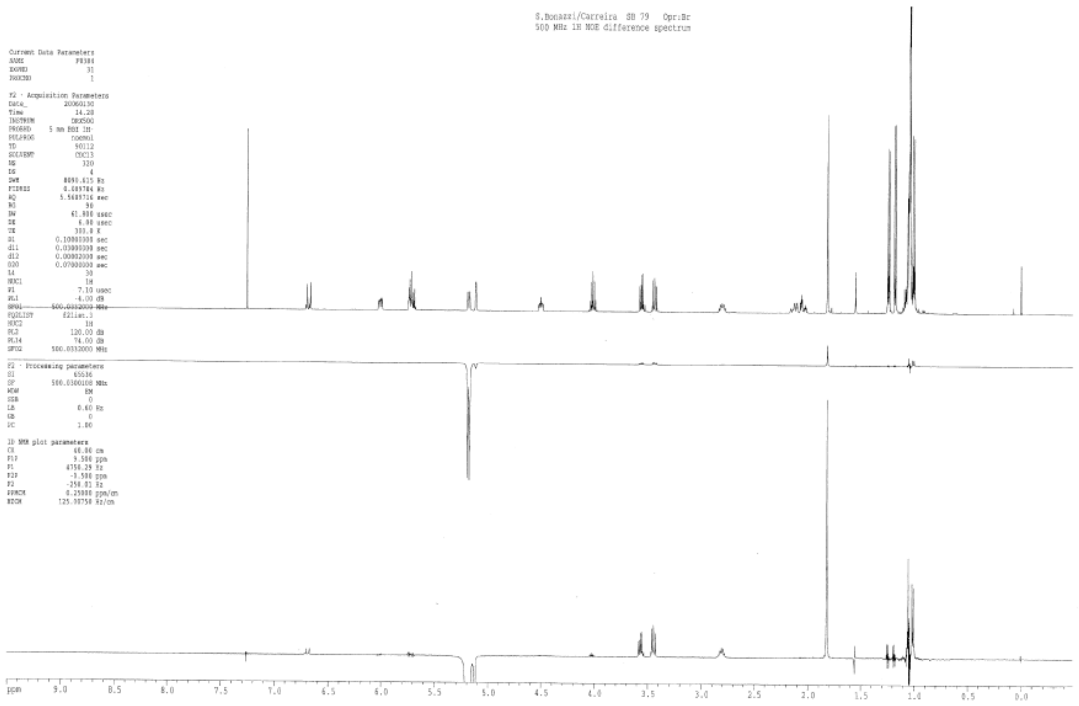
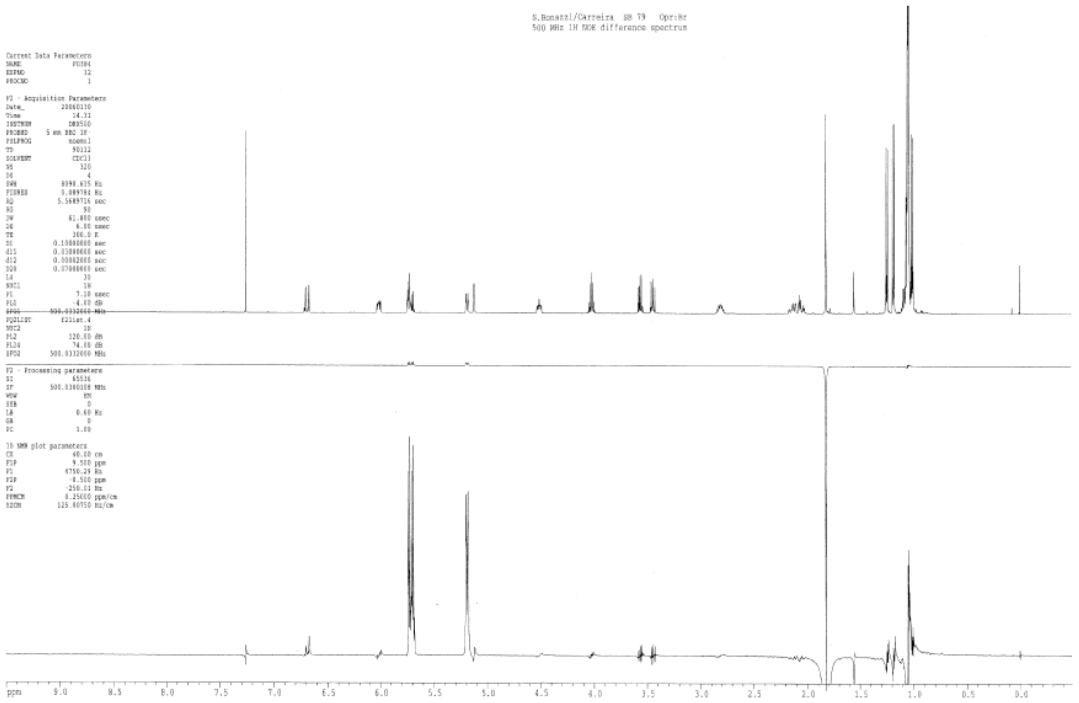
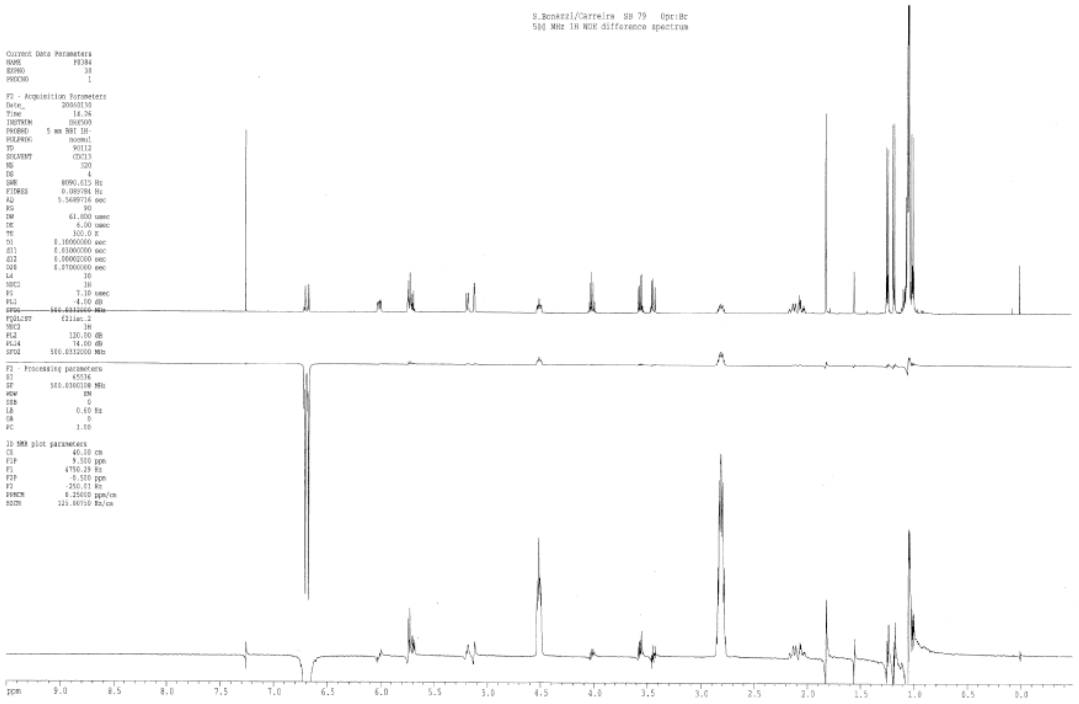
76.784
69.569
68.311
68.177
68.021
67.107
67.011
35.038
30.916
29.893
26.788
24.063
20.531
18.136
18.000
17.642
17.561
17.504
13.596
13.177
12.026
11.891
11.789
ppm
220
200
180
160
140
120
100
80
60
40
20
0


S.Bonazzi/Carreira SB 79 Opr:Br
125 MHz 13C DEPT 90+135 NMR
Current Data Parameters
NAME P1184
EXPNO 3
PROCNO 1
F2 - Acquisition Parameters
Date_ 20660131
Time 11.25
INSTRUM drx2500
PROBHD 5 mm BBO BB-
PULPROG deptletb
TD 131072
SOLVENT CDCl3
NS 512
DS 0
SWH 31152.648 Hz
FIDRES 0.237676 Hz
AQ 2.1037557 sec
RG 13004
DW 16.050 usec
DE 30.00 usec
TE 298.0 K
CNST2 145.0000000
D1 1.00000000 sec
d2 0.00344828 sec
d12 0.00002000 sec
DELTA 0.00031146 sec
========== CHANNEL f1 ==========
NUC1 13C
P1 9.00 usec
p2 18.00 usec
PL1 0.00 dB
SFO1 125.7715724 MHz
========== CHANNEL f2 ==========
CPDPRG2 waltz16
NUC2 1H
P0 11.00 usec
P3 11.00 usec
p4 22.00 usec
PCPD2 80.00 usec
PL2 -1.00 dB
PL12 13.50 dB
SFO2 500.1320005 MHz
F2 - Processing parameters
SI 65536
SF 125.7577876 MHz
WDW EM
SSB 0
LB 1.10 Hz
GB 0
PC 1.40
1D NMR plot parameters
CX 40.00 cm
F1P 232.000 ppm
F1 29175.80 Hz
F2P -8.000 ppm
F2 -1006.06 Hz
PPMCM 6.00000 ppm/cm
HZCM 754.54675 Hz/cm
ppm
220
200
180
160
140
120
100
80
60
40
20
0


S.Bonazzi/Carreira SB 79 Opr:Rr
500 MHz 1H NOE difference spectrum
ppm


S.Bonazzi/Carreira SB 79 Opr:Rr
500 MHz 1H NOE difference spectrum
ppm


S.Bonazzi/Carreira SB 79 Opr:Hr
500 MHz 1H NOE difference spectrum
Current Data Parameters
NAME PE384
EXPNO 38
PROCNO 1
F2 - Acquisition Parameters
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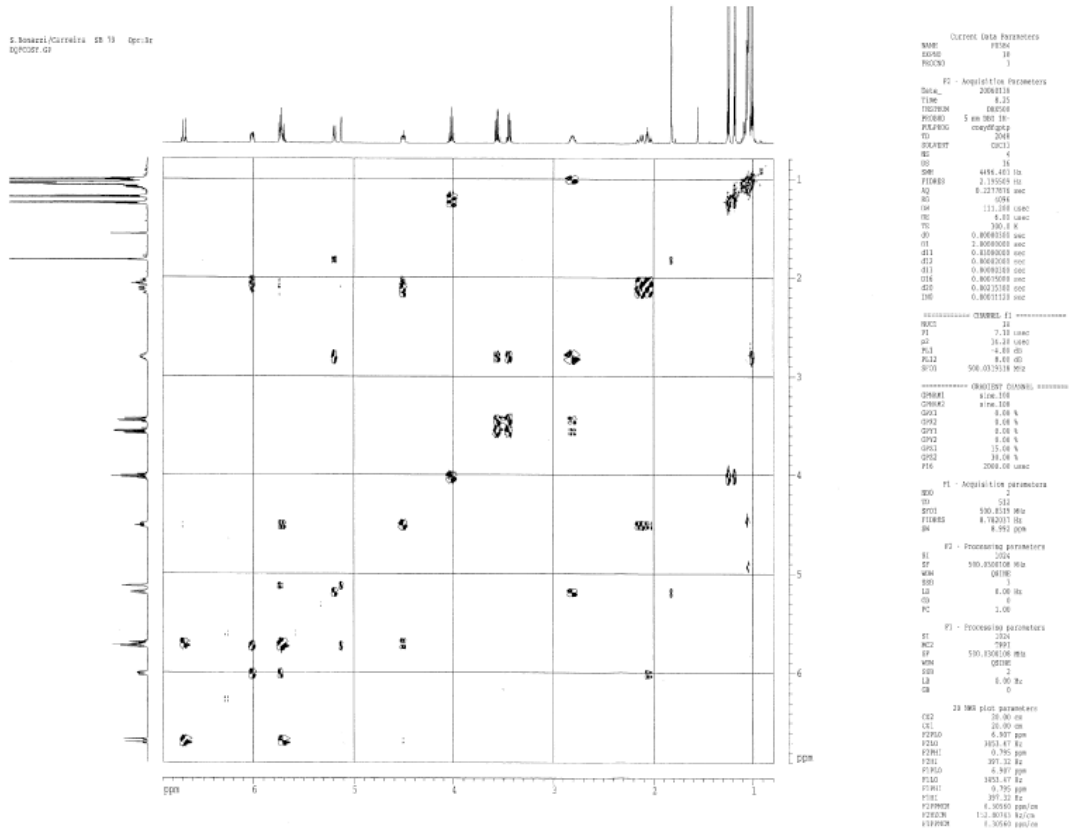
Time 14.24
INSTRUM DRX500
PROBHD 5 mm BBI 1H-
PULPROG noesydfgp
TD 90112
SOLVENT CDCl3
NS 320
DS 4
SWH 8090.615 Hz
FIDRES 0.089784 Hz
AQ 5.5689716 sec
RG 90
DW 61.800 usec
DE 6.00 usec
TE 300.0 K
D1 8.16000000 sec
d11 0.03000000 sec
d12 0.00002000 sec
D20 0.07000000 sec
L4 10
NUC1 1H
P1 7.10 usec
PL1 4.00 dB
SFO1 500.0332000 MHz
F2 - Processing parameters
SI 65536
SF 500.0300108 MHz
WDW EM
SSB 0
LB 0.60 Hz
GB 0
PC 1.00
1D NMR plot parameters
CX 40.00 cm
F1P 9.500 ppm
F1 4750.29 Hz
F2P -0.500 ppm
F2 -250.01 Hz
PPMCM 0.25600 ppm/cm
HZCM 128.00710 Hz/cm
ppm


S.Bonazzi/Carreira SB 79 Opr:Hr
DQFCOSY.G3
ppm
Current Data Parameters
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EXPNO 38
PROCNO 1
F2 - Acquisition Parameters
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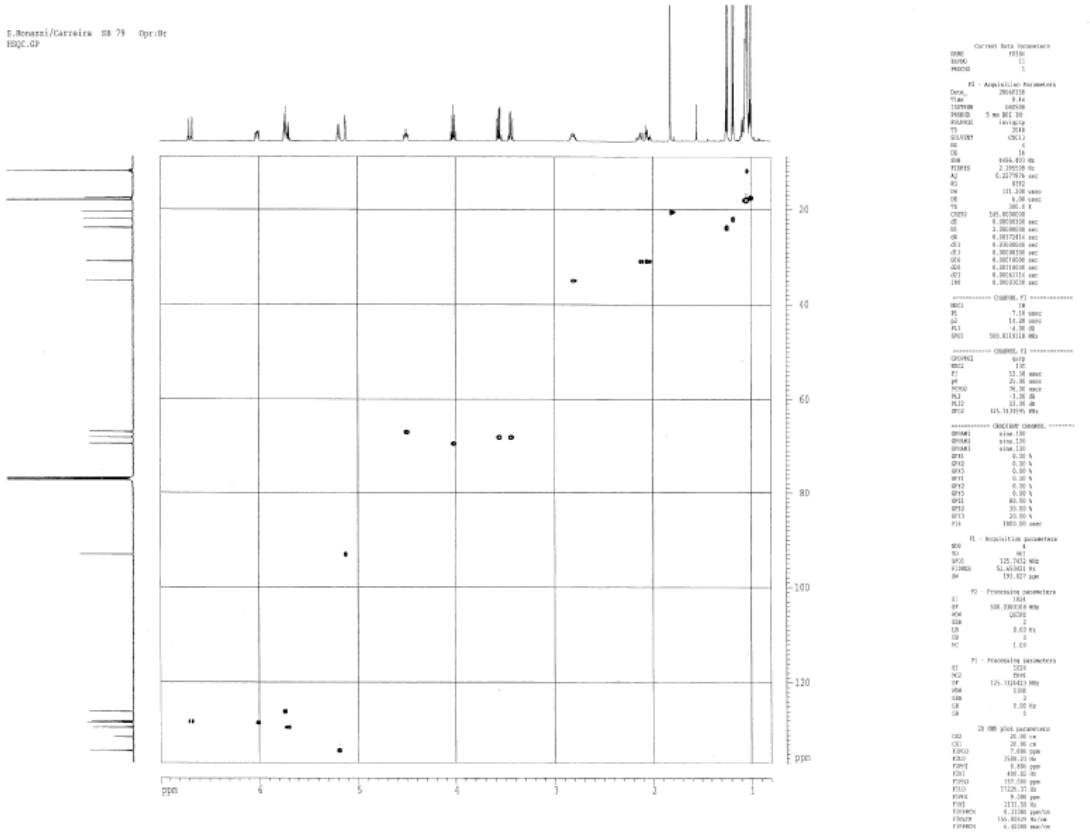
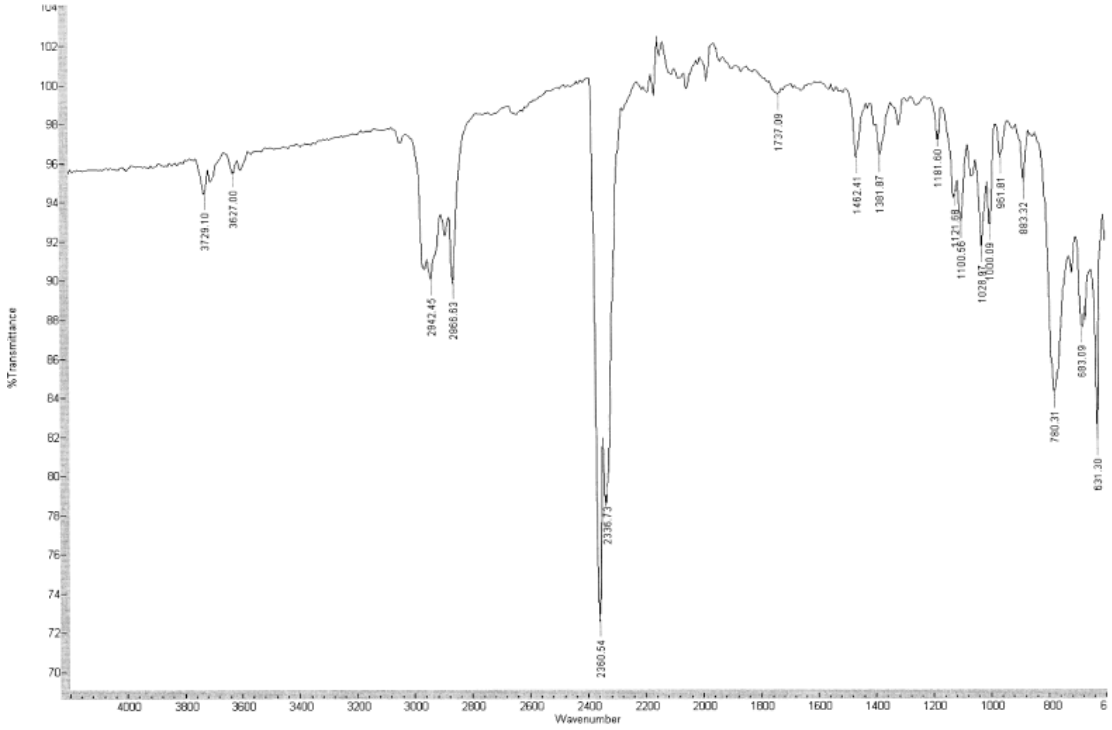
Time 8.25
INSTRUM DRX500
PROBHD 5 mm BBI 1H-
PULPROG cosydfgsp
TD 2048
SOLVENT CDCl3
NS 4
DS 16
SWH 4496.403 Hz
FIDRES 2.195569 Hz
AQ 0.2277876 sec
RG 4096
DW 111.200 usec
DE 6.00 usec
TE 300.0 K
d0 0.00003088 sec
D1 2.00000000 sec
d11 0.01000000 sec
d12 0.00003088 sec
d13 0.00003088 sec
D16 0.00075000 sec
d20 0.00235388 sec
IN0 0.00011128 sec
CHANNEL f1
NUC1 1H
P1 7.10 usec
p2 14.20 usec
PL1 -4.00 dB
PL12 8.00 dB
SFO1 500.0335318 MHz
GRADIENT CHANNEL
GPNAM1 sine.100
GPNAM2 sine.100
GPX1 0.00 %
GPX2 0.00 %
GPY1 0.00 %
GPY2 0.00 %
GPZ1 15.00 %
GPZ2 30.00 %
P16 2000.00 usec
F1 - Acquisition parameters
NDO 2
TD 512
SFO1 500.8535 MHz
FIDRES 0.782037 Hz
SW 8.992 ppm
F2 - Processing parameters
SI 2024
SF 500.8300108 MHz
WDW QSINE
SSB 0
LB 0.00 Hz
GB 0
PC 1.00
F1 - Processing parameters
SI 2024
MC2 TPPI
SF 500.8300106 MHz
WDW QSINE
SSB 2
LB 0.00 Hz
GB 0
2D NMR plot parameters
CX2 20.00 cm
CX1 20.00 cm
F2PLO 6.907 ppm
F2HZ 3453.47 Hz
F2PH1 0.795 ppm
F2H1 397.32 Hz
F1PLO 6.907 ppm
F1HZ 3453.47 Hz
F1PH1 0.795 ppm
F1H1 397.32 Hz
F2PPMCM 0.30560 ppm/cm
F2HZCM 152.80741 Hz/cm
F1PPMCM 0.30560 ppm/cm

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Wavenumber
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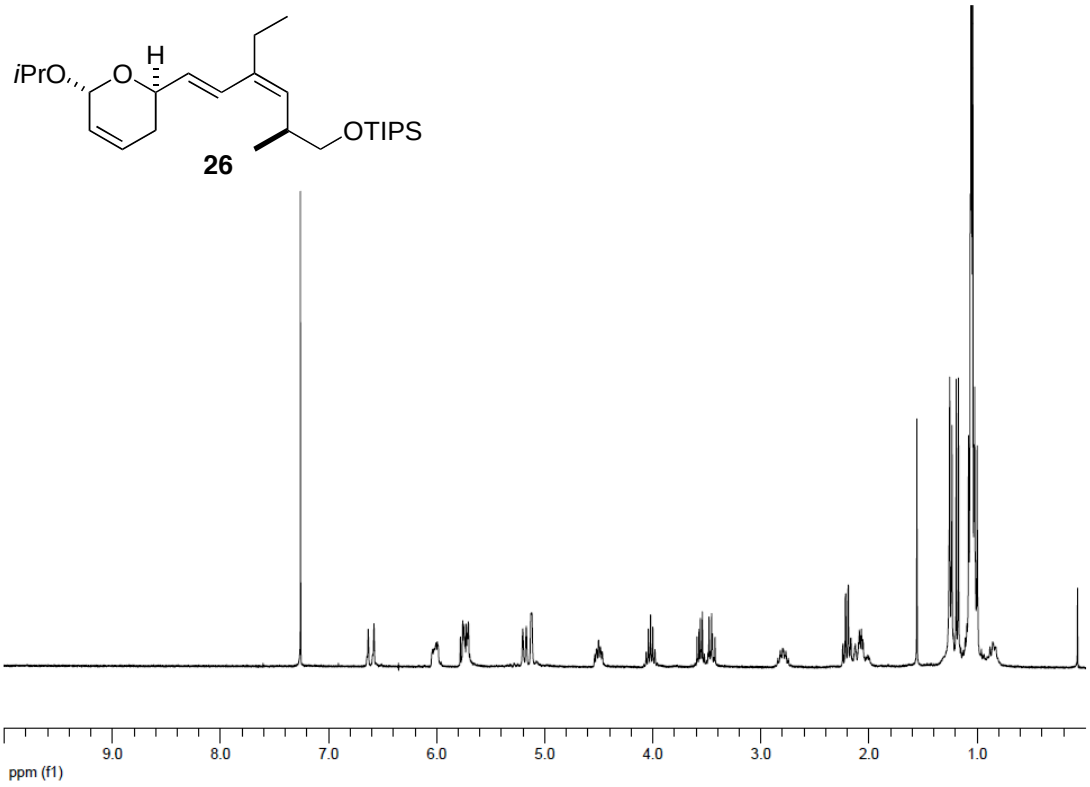
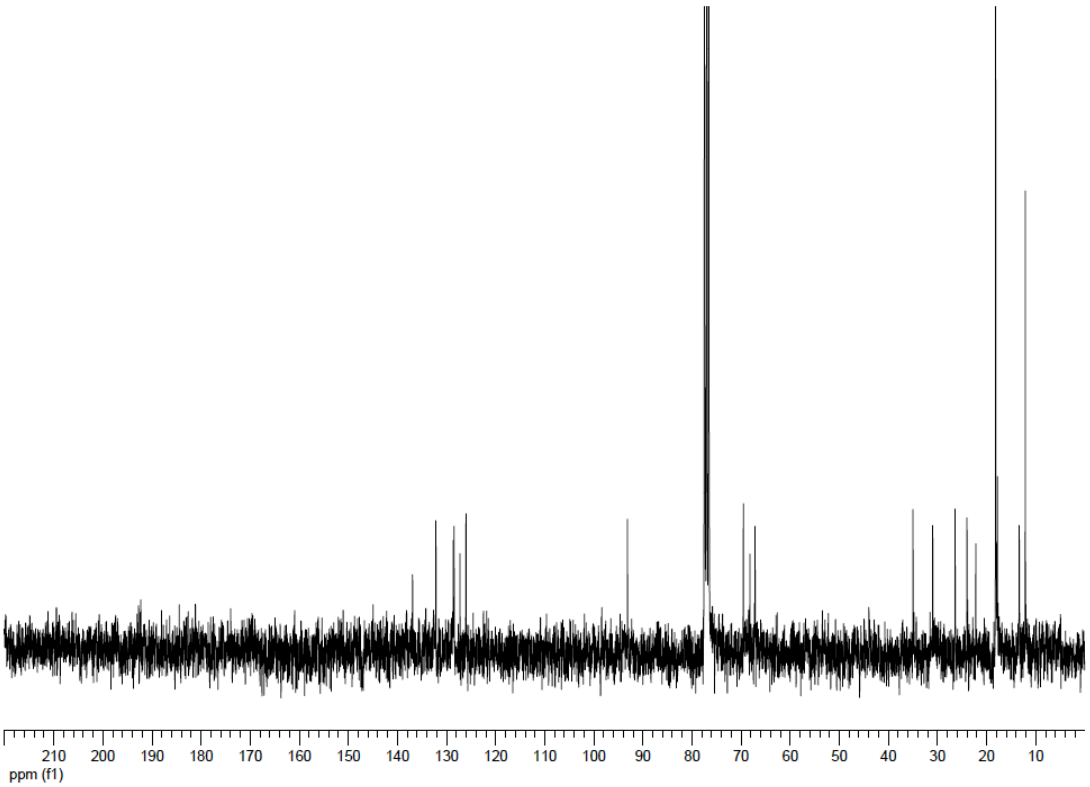
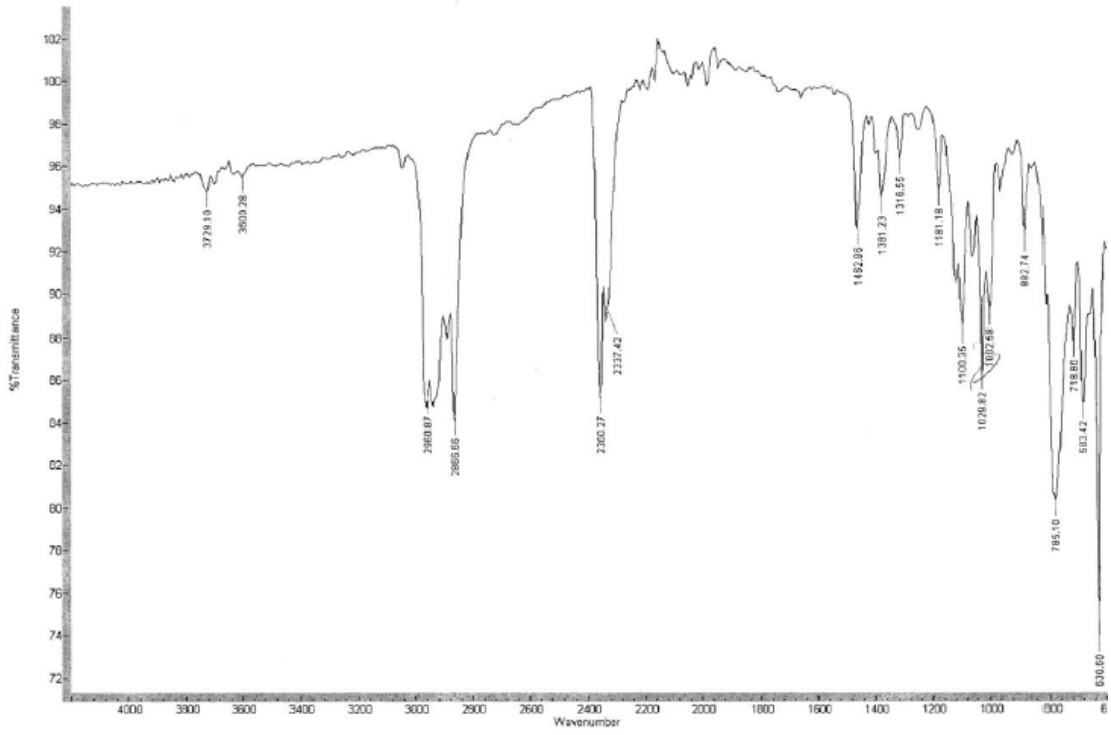
2942.45
2868.63
2360.54
2336.73
1737.09
1462.41
1381.87
1181.60
1171.68
1100.56
1028.87
1000.09
961.81
883.32
780.31
693.09
631.30


iPrO
O
H
OTIPS
26
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)


210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm (f1)


%Transmittance
Wavenumber
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3800
3600
3400
3200
3000
2800
2600
2400
2200
2000
1800
1600
1400
1200
1000
800
3729.10
3603.28
2960.87
2865.65
2360.27
2337.42
1492.06
1381.23
1316.55
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1100.35
1070.82
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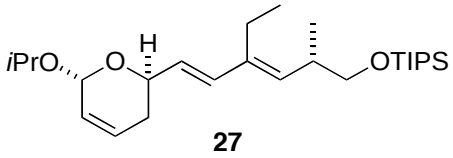
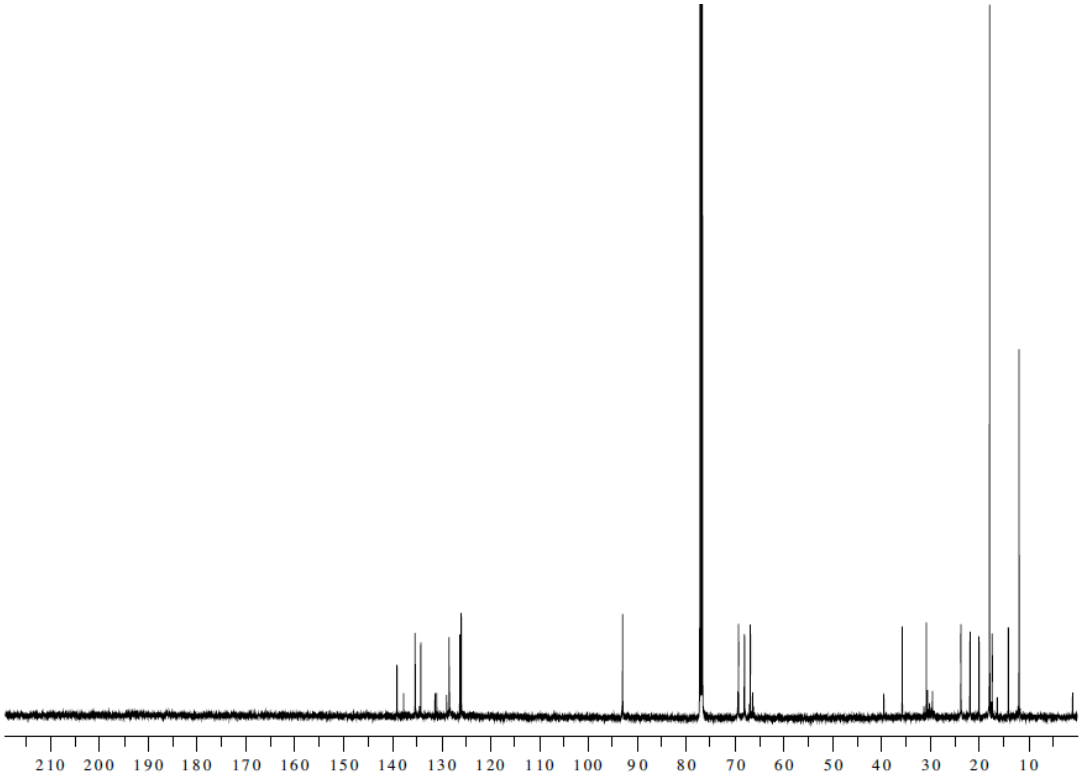
982.74
785.10
718.80
583.42
630.60


*i*PrO
O
H
OTIPS

27

0
9
8
7
6
5
4
3
2
1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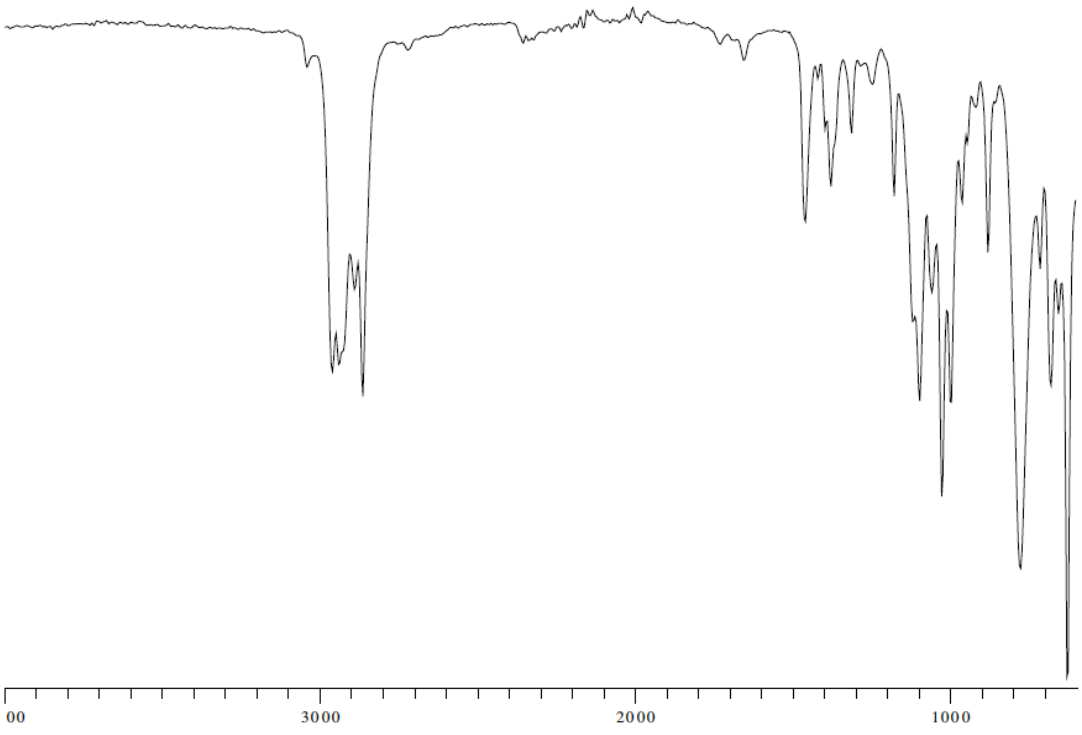
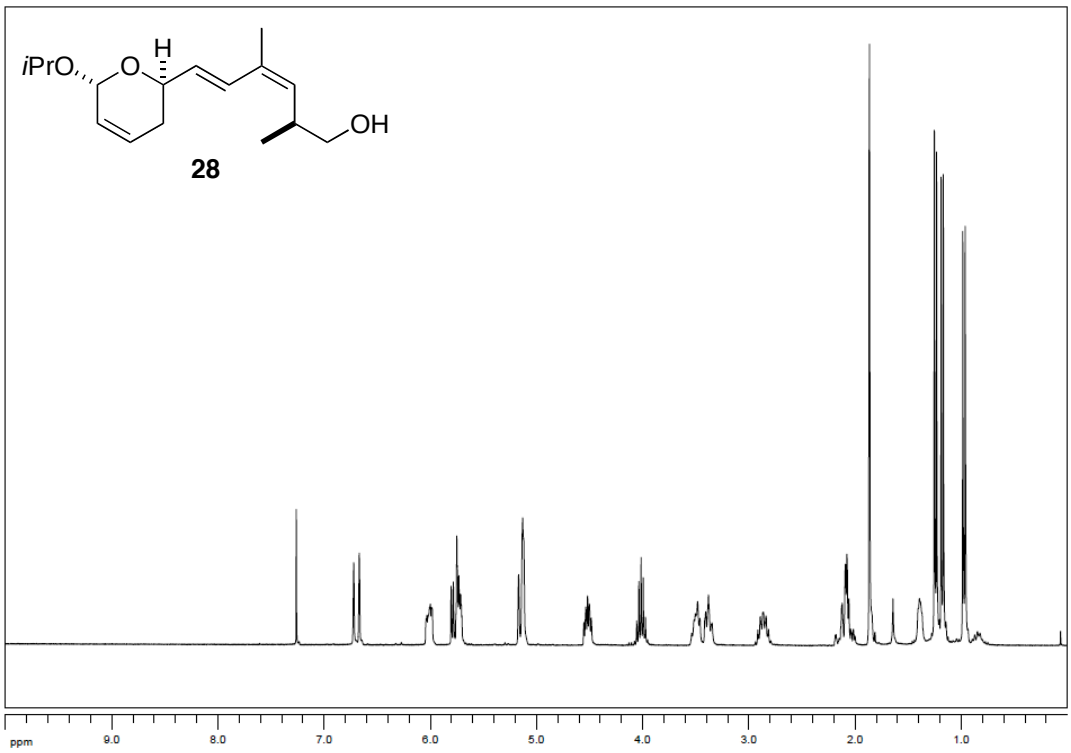

00 3000 2000 1000


iPrO
O
H
OH
28
ppm
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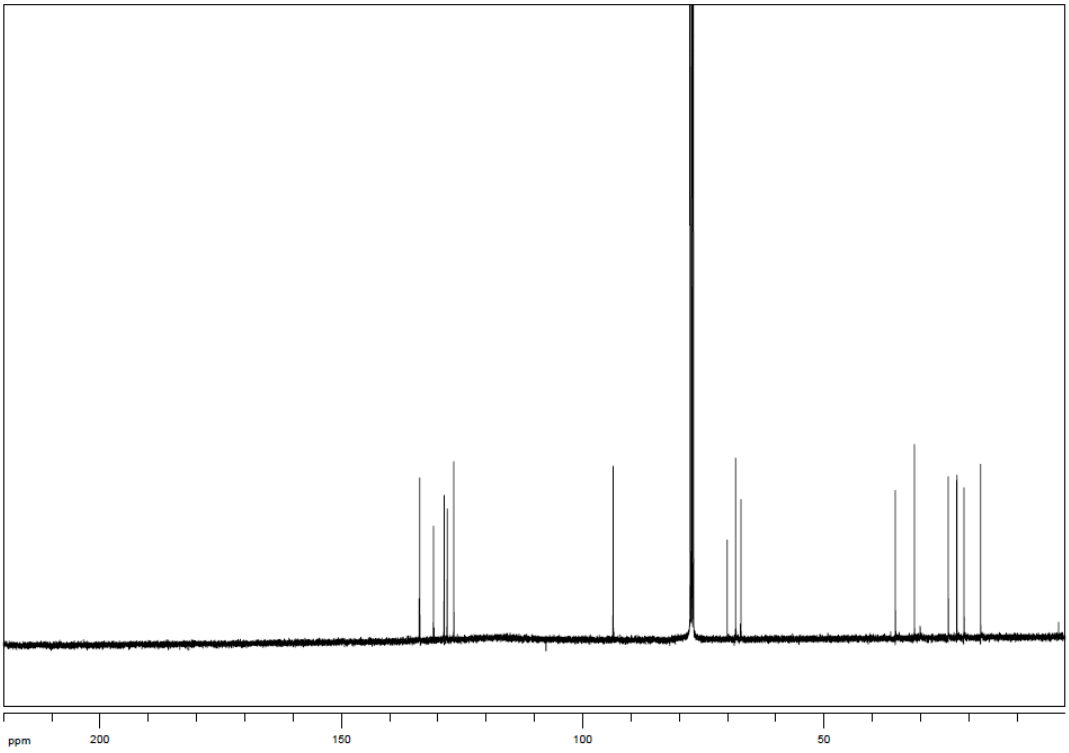
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
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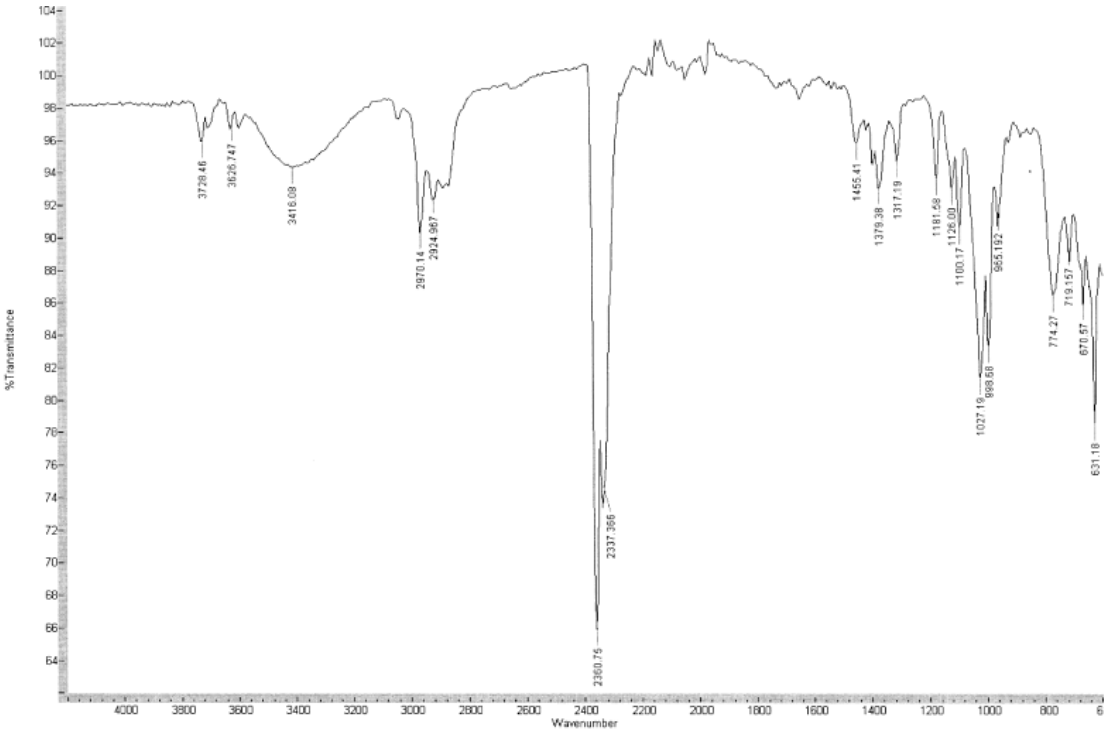
150
100
50


%Transmittance
Wavenumber
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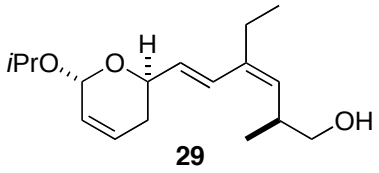
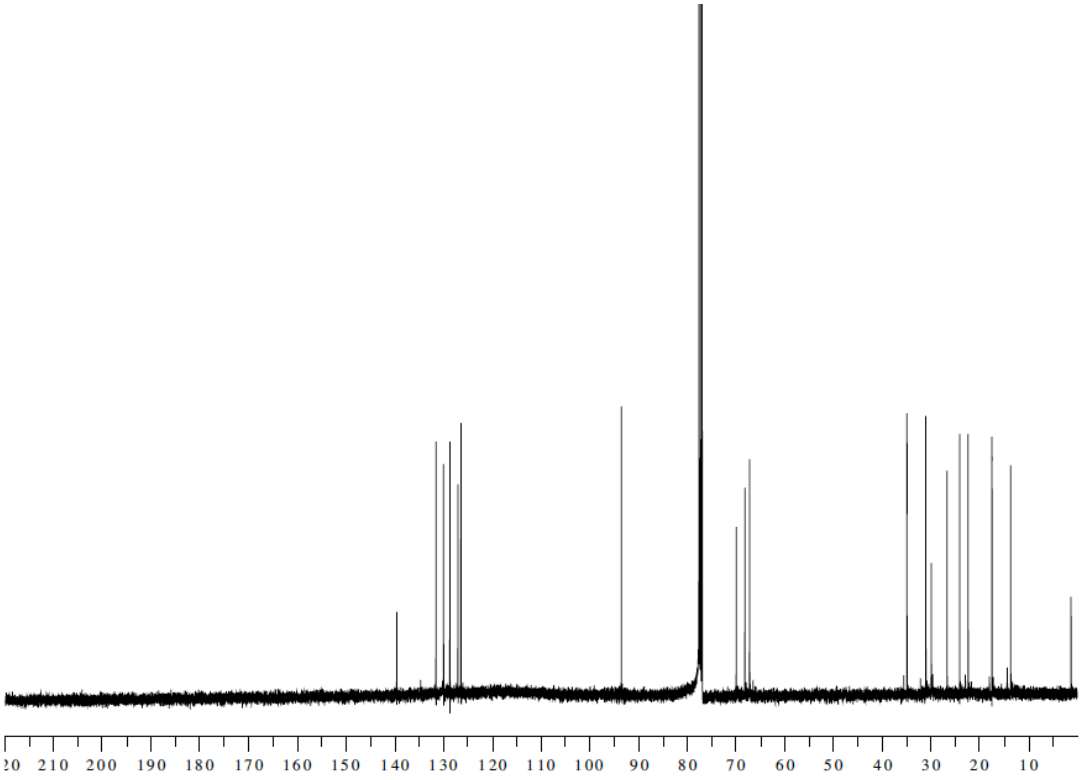
3625.747
3416.08
2970.14
2924.967
2360.75
2337.366
1455.41
1379.38
1317.19
1191.58
1126.03
1100.17
1027.19
988.58
965.192
774.27
719.157
670.57
631.18


*i*PrO
O
H
OH

29

0
9
8
7
6
5
4
3
2
1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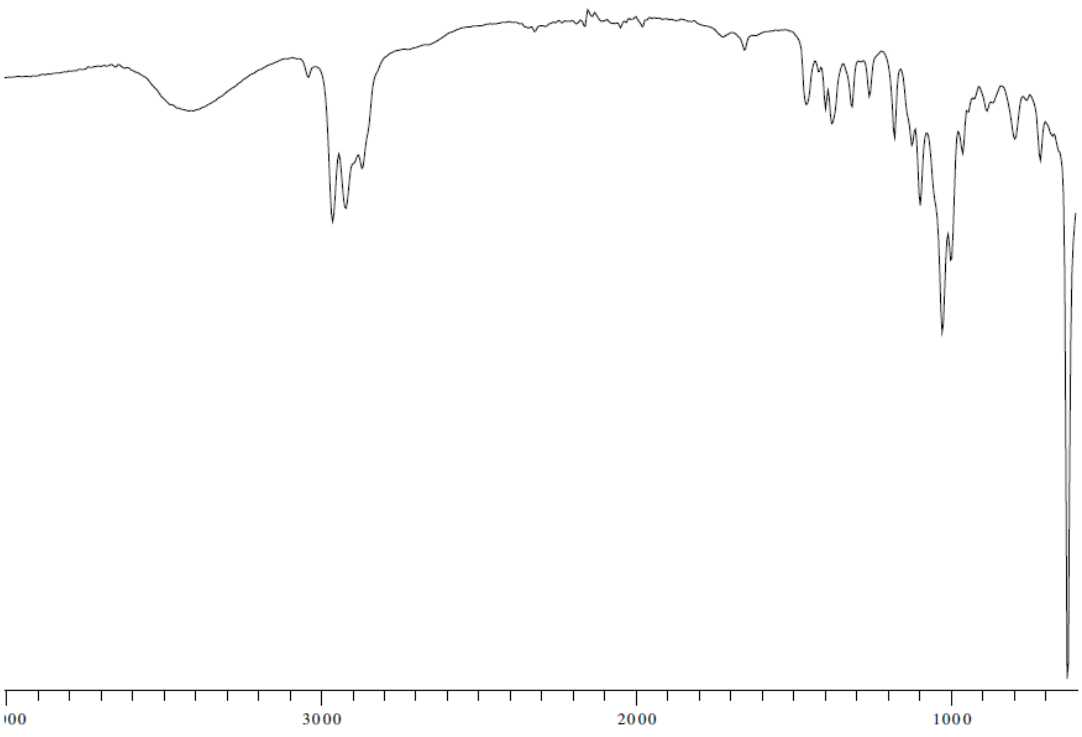
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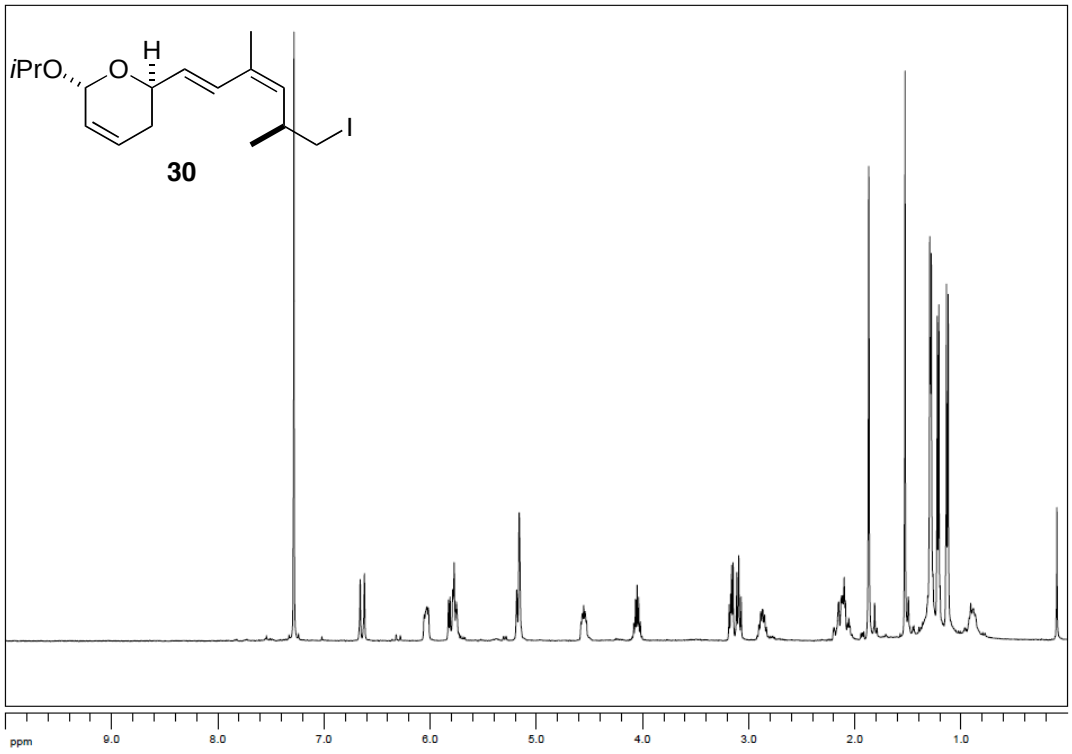
2000
1000


iPrO
O
H
I
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ppm
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6.0
5.0
4.0
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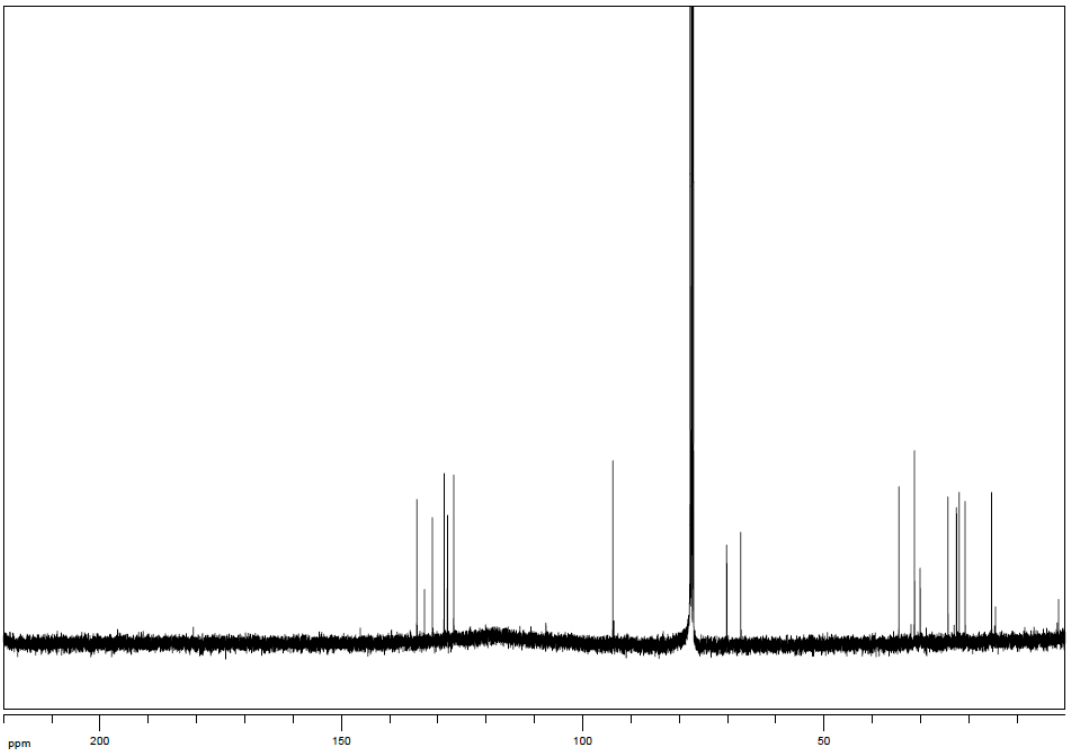
2.0
1.0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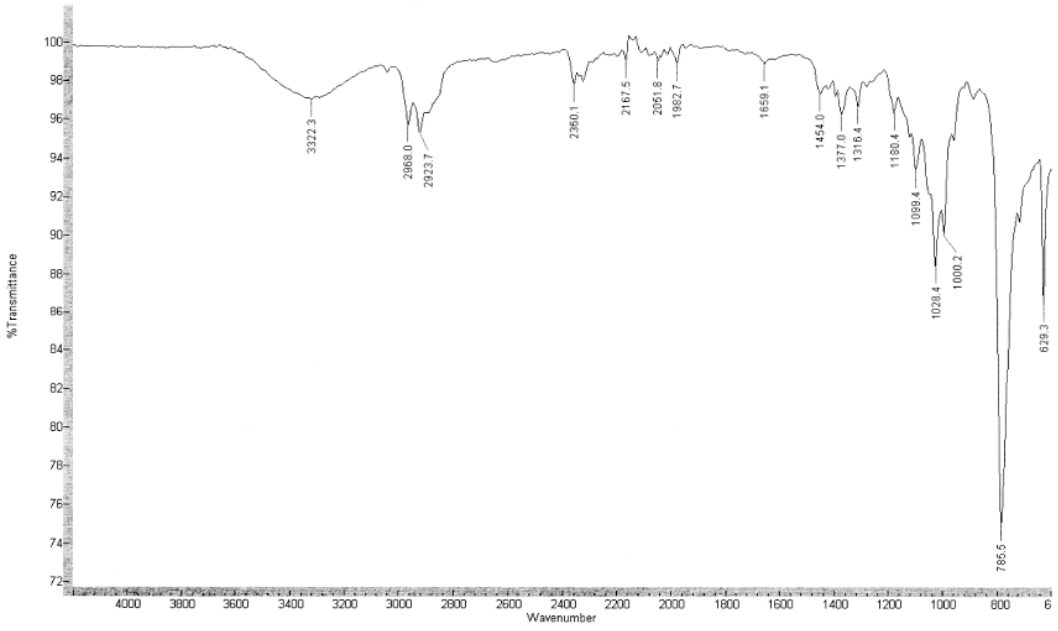
150
100
50


%Transmittance
Wavenumber
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2923.7
2360.1
2167.5
2051.8
1982.7
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1377.0
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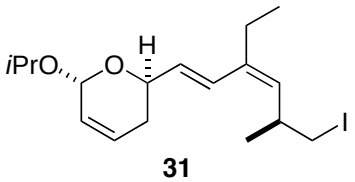
1099.4
1028.4
1000.2
785.5
629.3


*i*PrO
O
H
I

31

0
9
8
7
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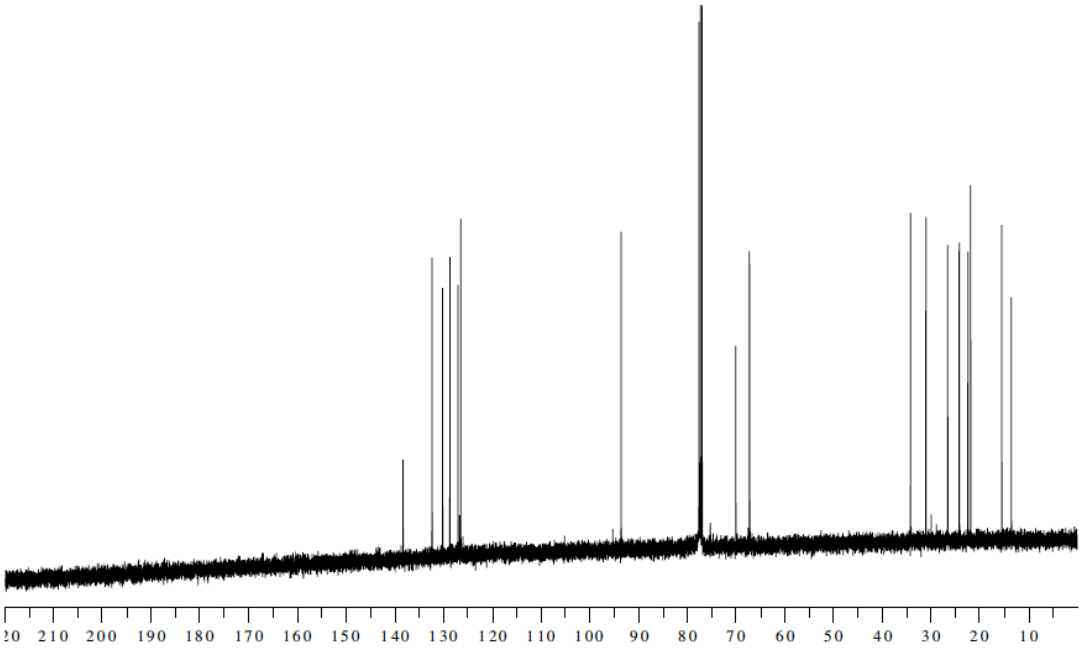
5
4
3
2
1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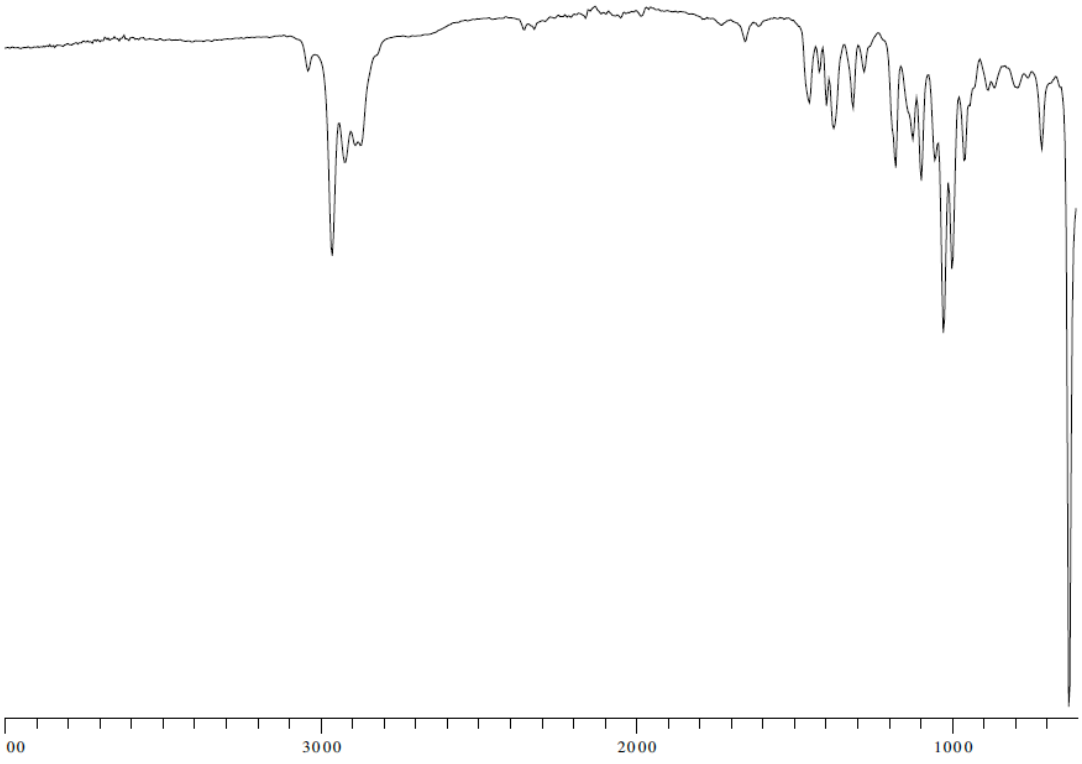
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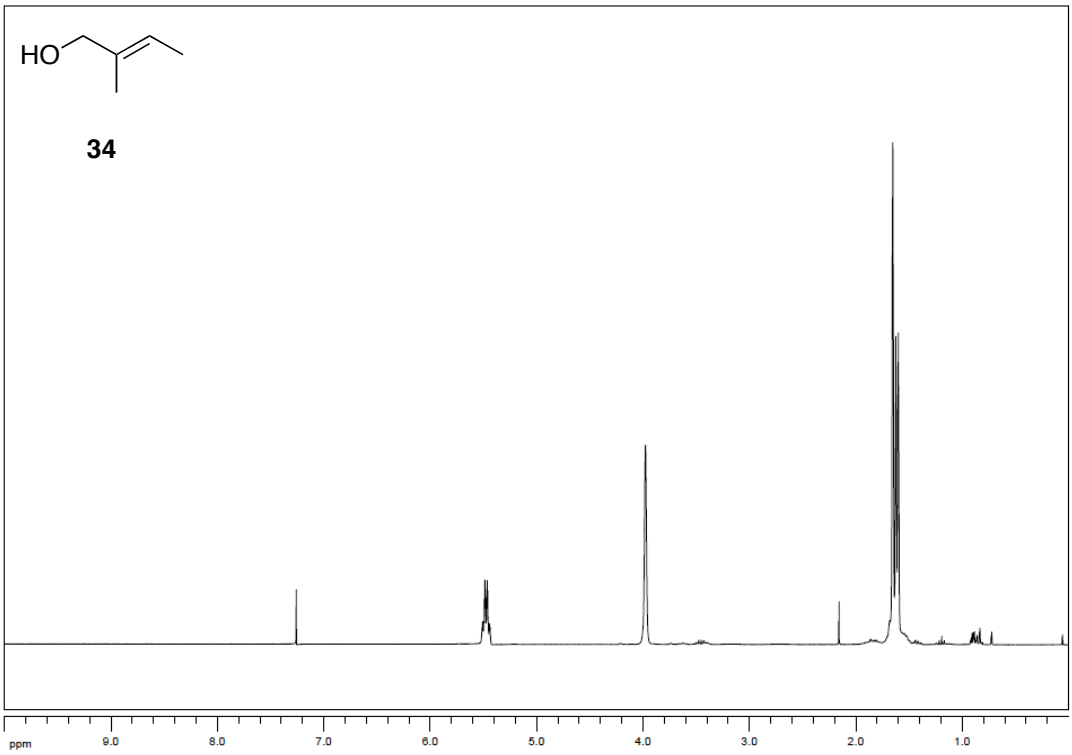
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ppm
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6.0
5.0
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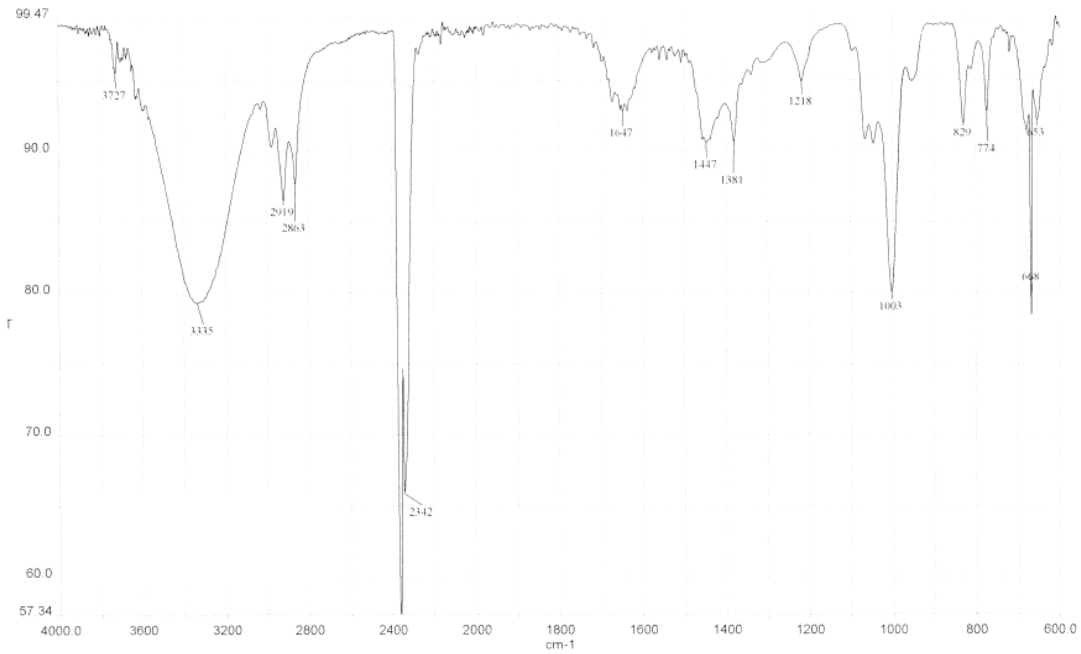
2.0
1.0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90.0
80.0
70.0
60.0
57.34
T
3727
3335
2919
2863
2342
1647
1447
1381
1218
1003
829
774
653
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3200
2800
2400
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1600
1400
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800
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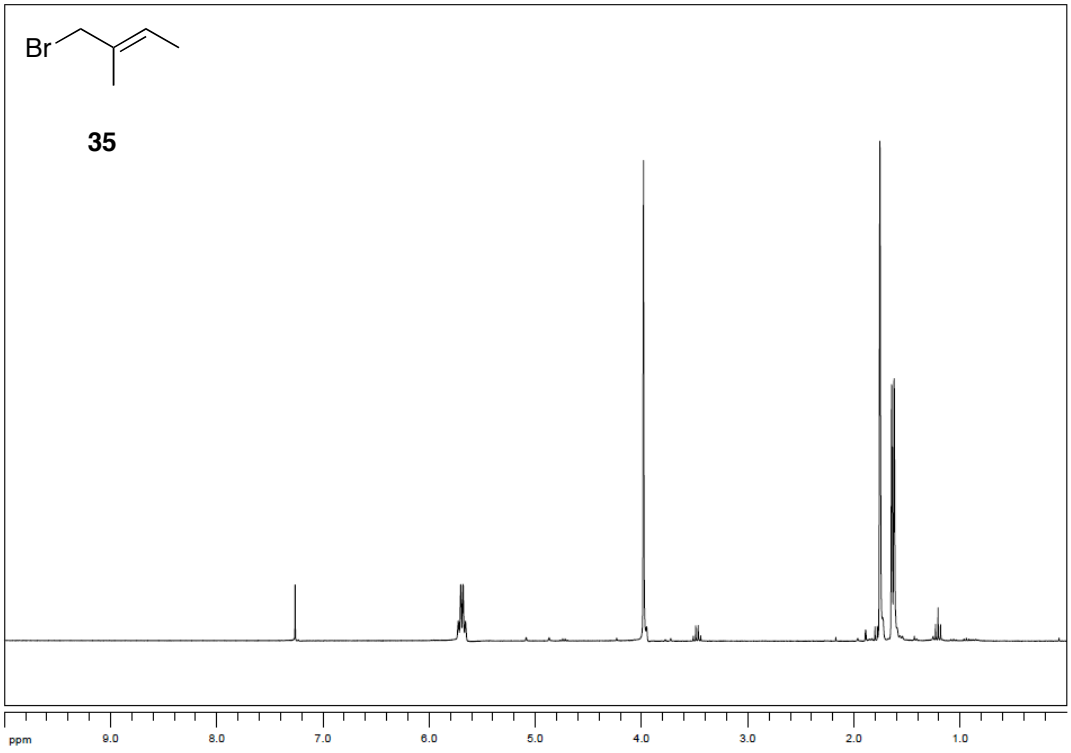
cm-1


Br
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6.0
5.0
4.0
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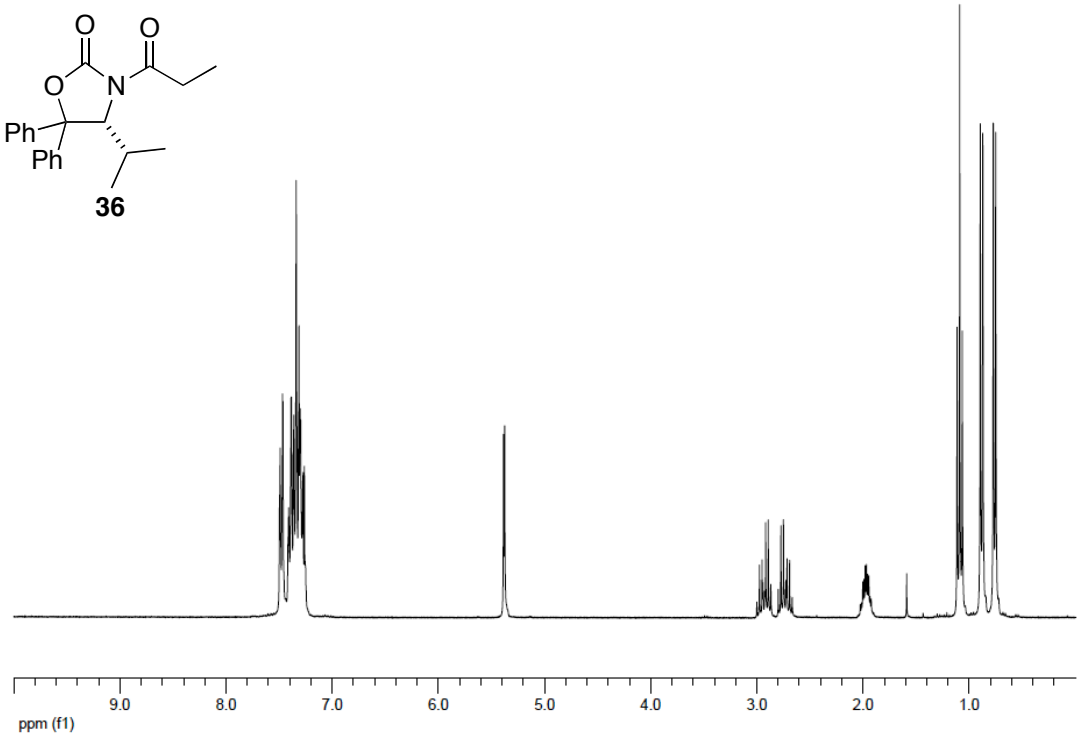
2.0
1.0


O
O
O
N
Ph
Ph
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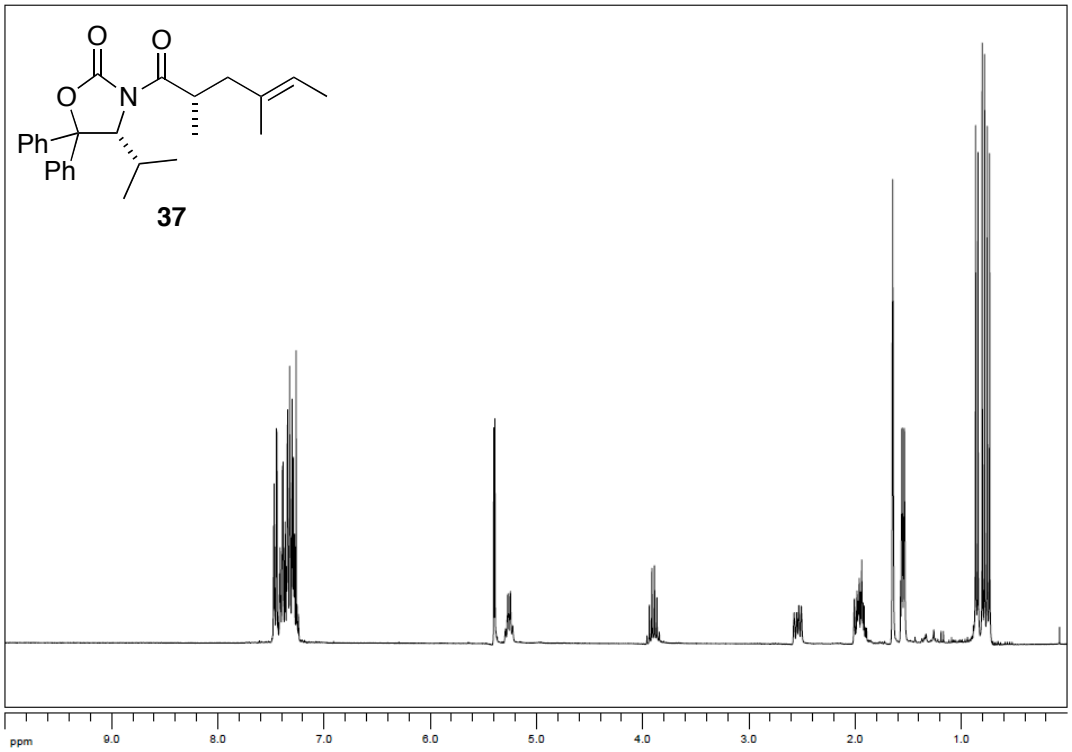
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)


O
O
O
N
Ph
Ph
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ppm
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8.0
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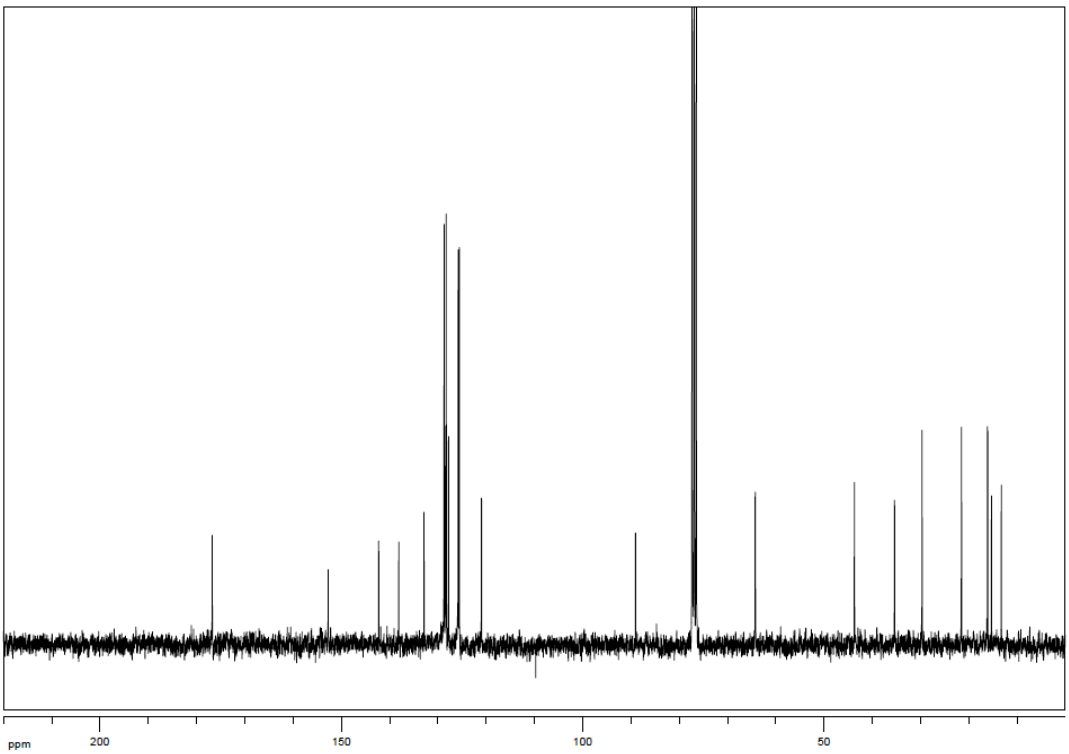
6.0
5.0
4.0
3.0
2.0
1.0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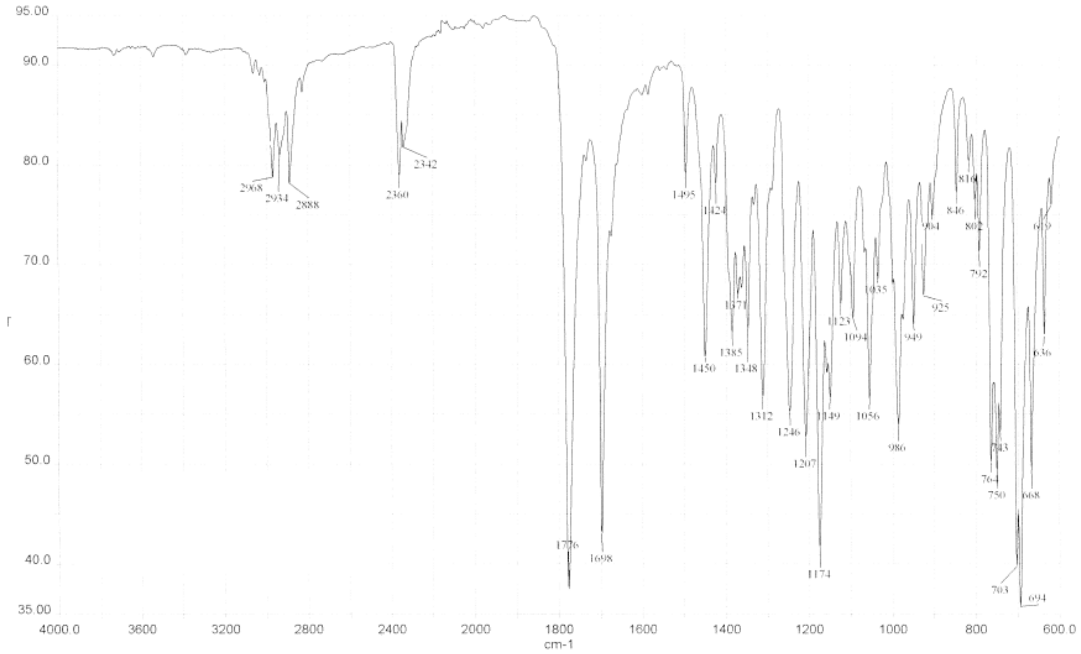
150
100
50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80.0
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60.0
50.0
40.0
35.00
T
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1
2968
2934
2888
2360
2342
1706
1698
1495
1424
1450
1385
1348
1312
1246
1207
1174
1149
1123
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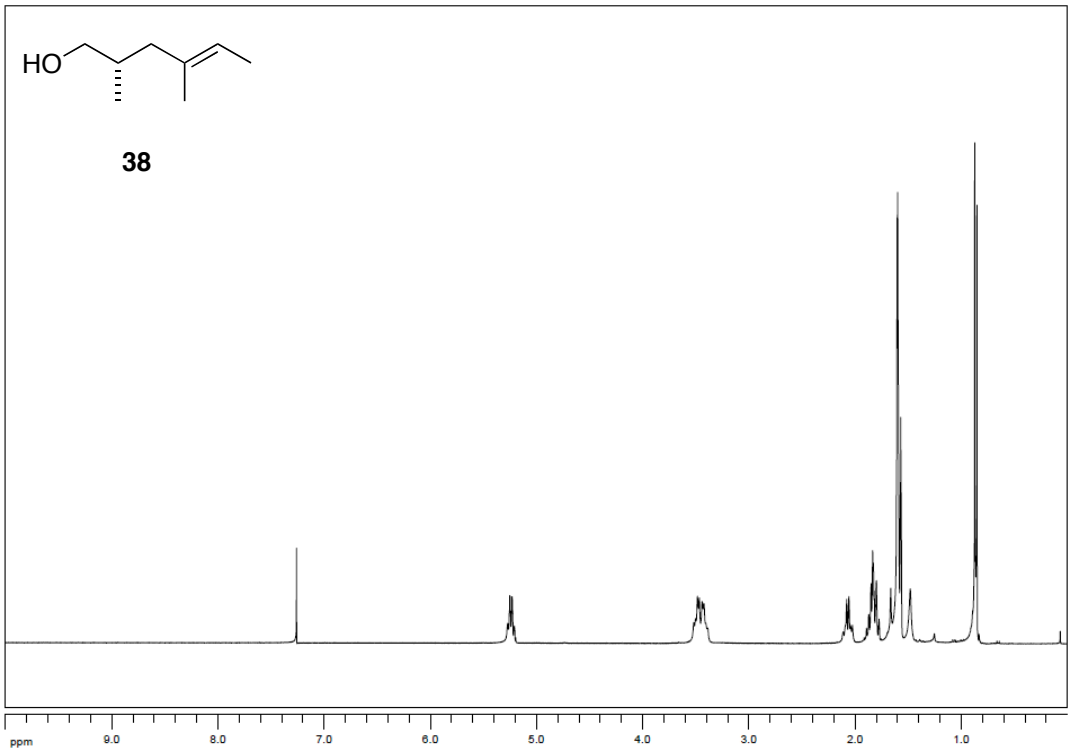
816
802
792
764
750
743
703
694
668
636


HO
38
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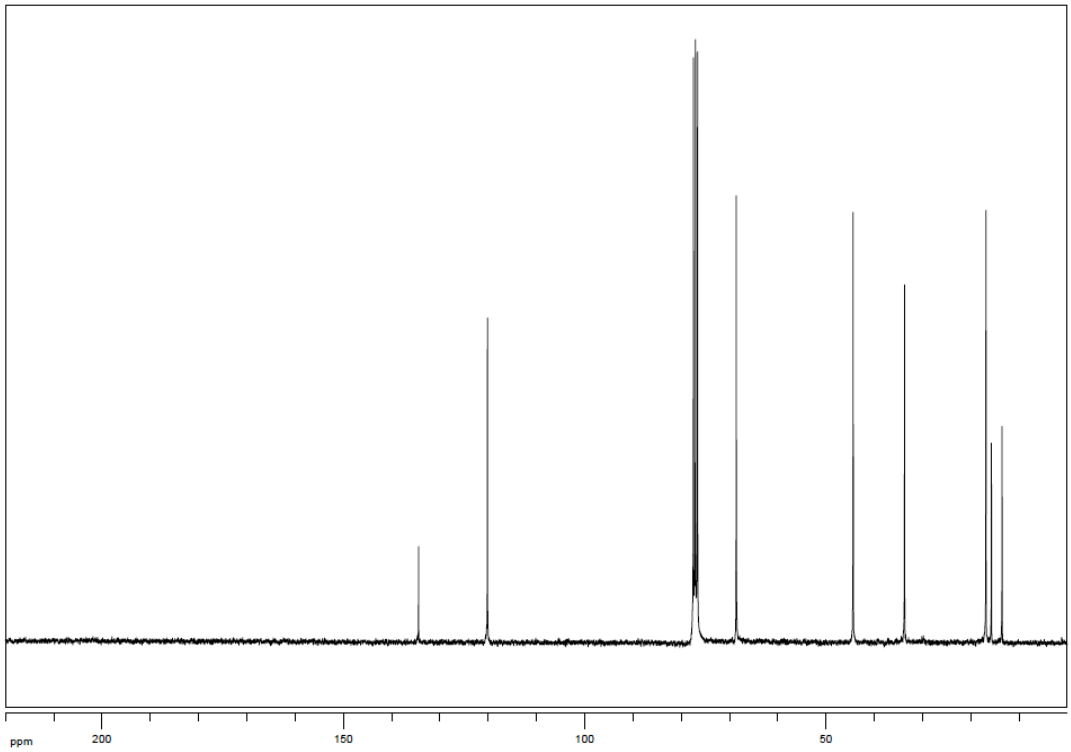
1.0


ppm
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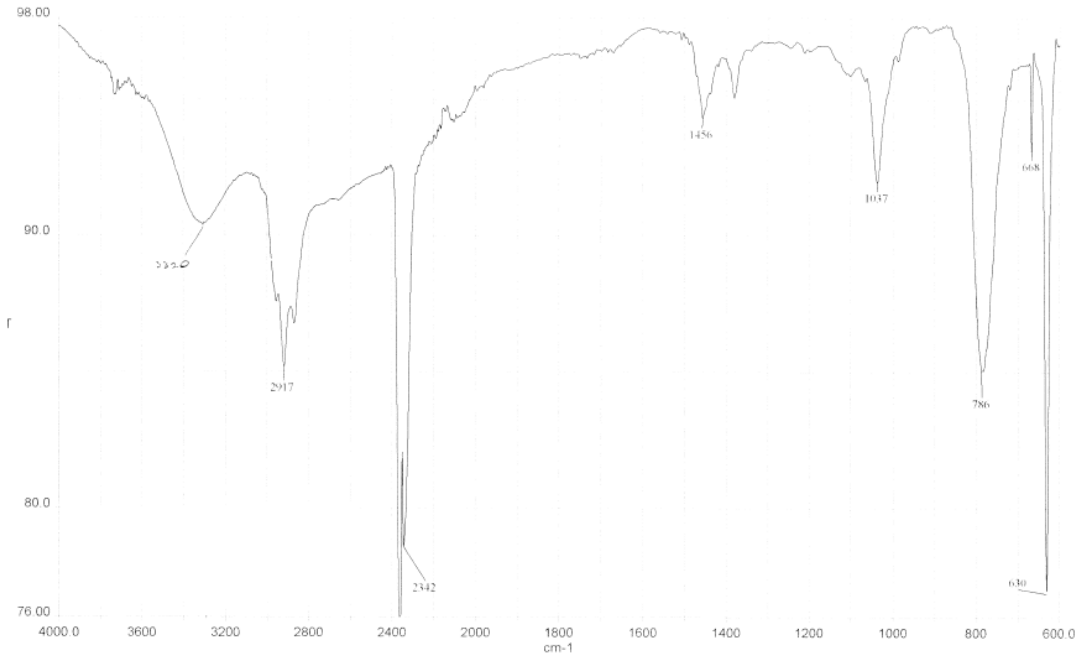
150
100
50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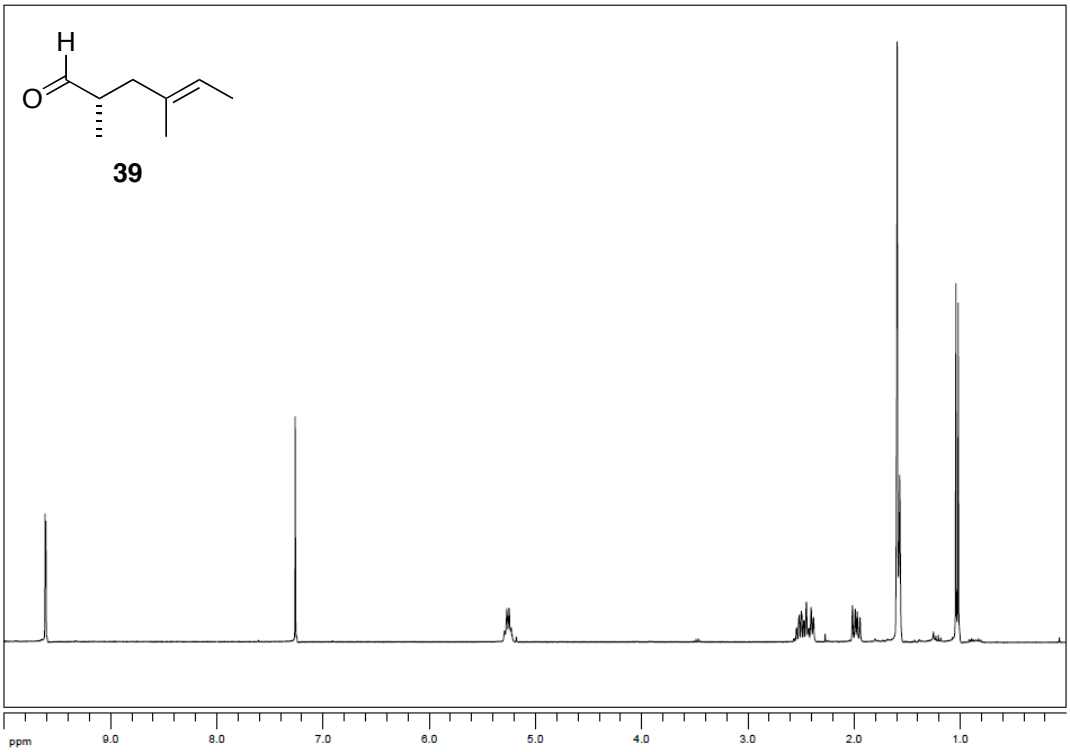
90.0
80.0
76.00
T
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1
2917
2342
1456
1047
786
668
630


H
O
39
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4.0
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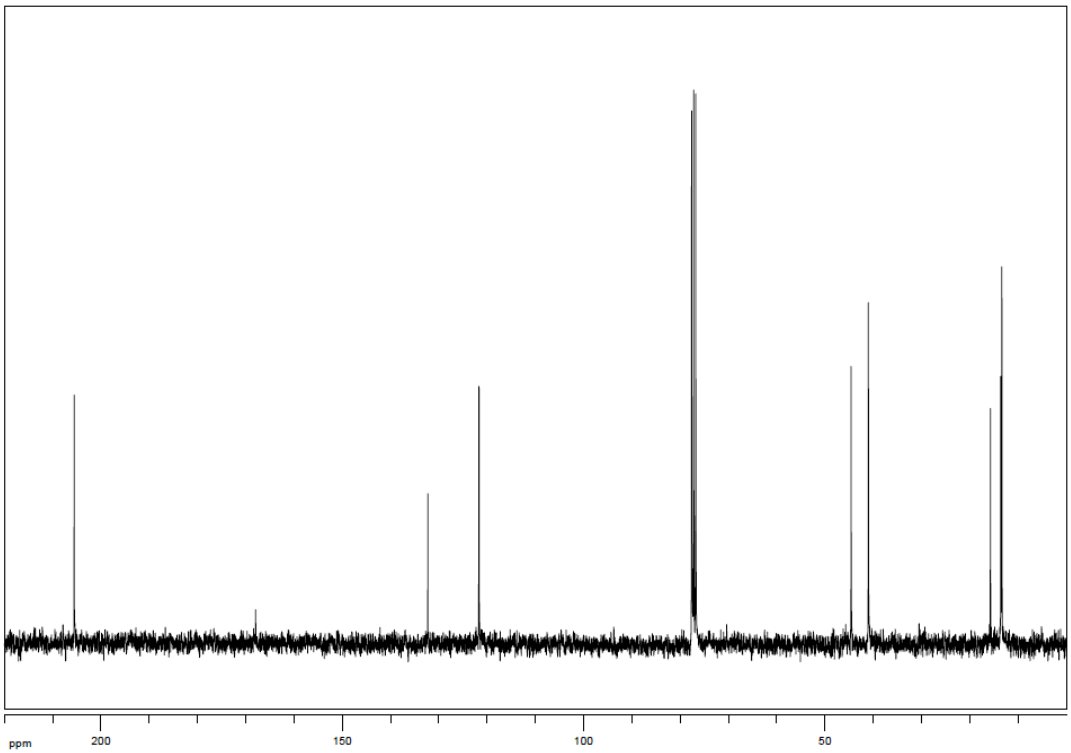
2.0
1.0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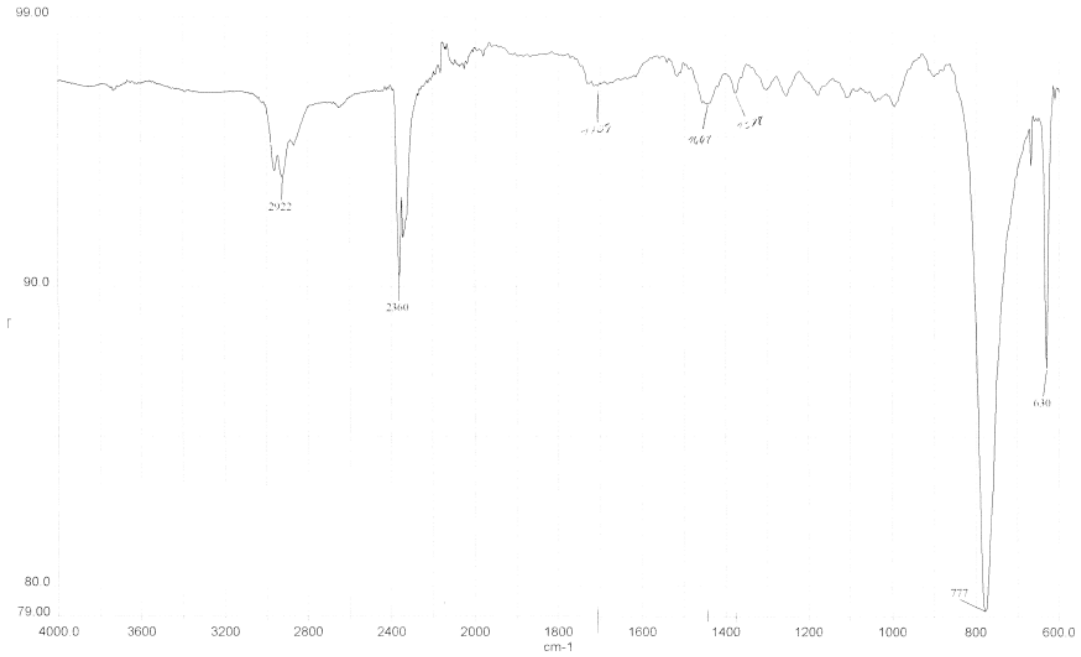
150
100
50


99.00
90.0
80.0
79.00
2922
2360
777
630
4000.0
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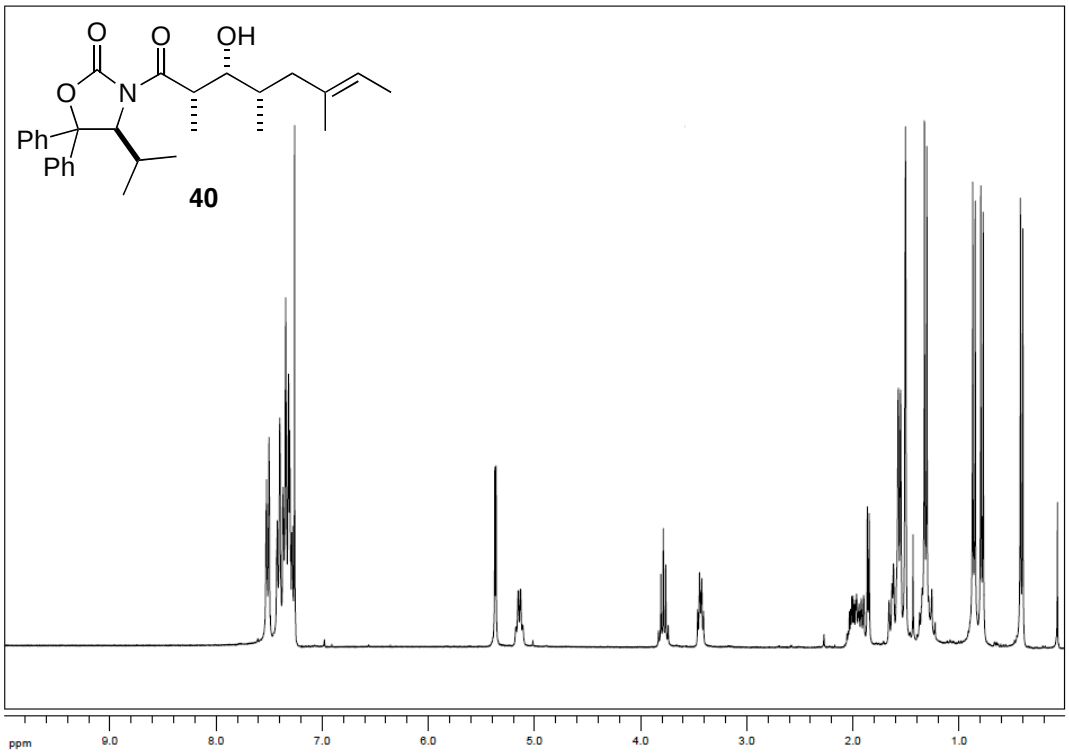
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1


O
O
OH
O
N
Ph
Ph
40
ppm
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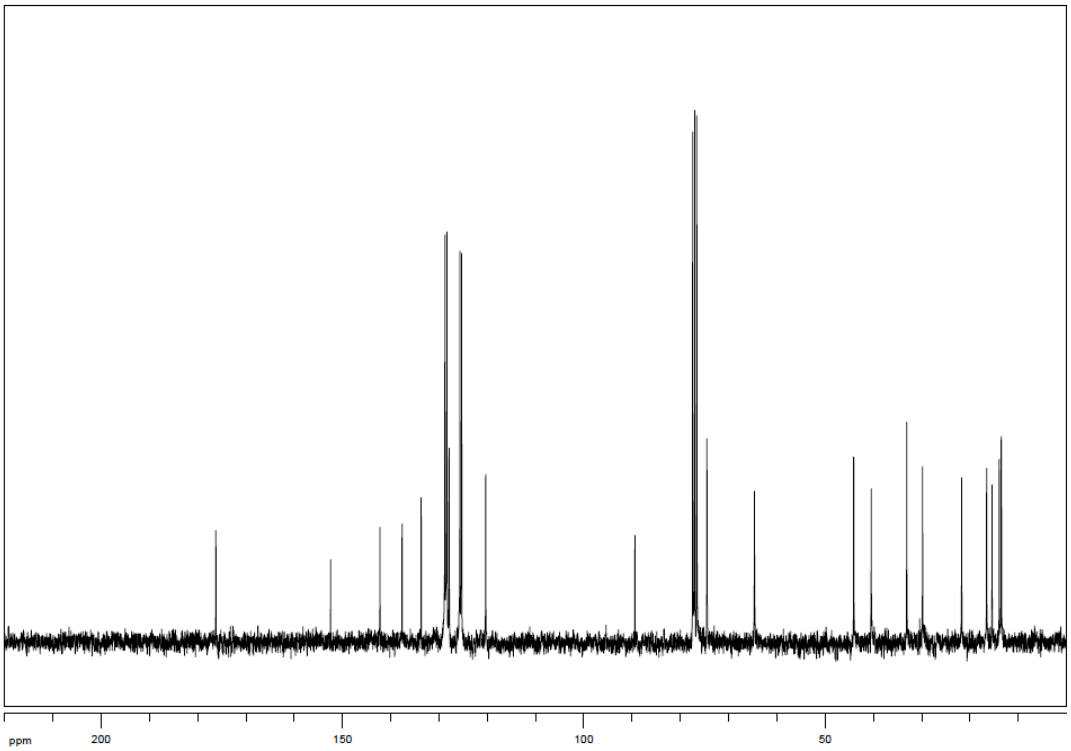
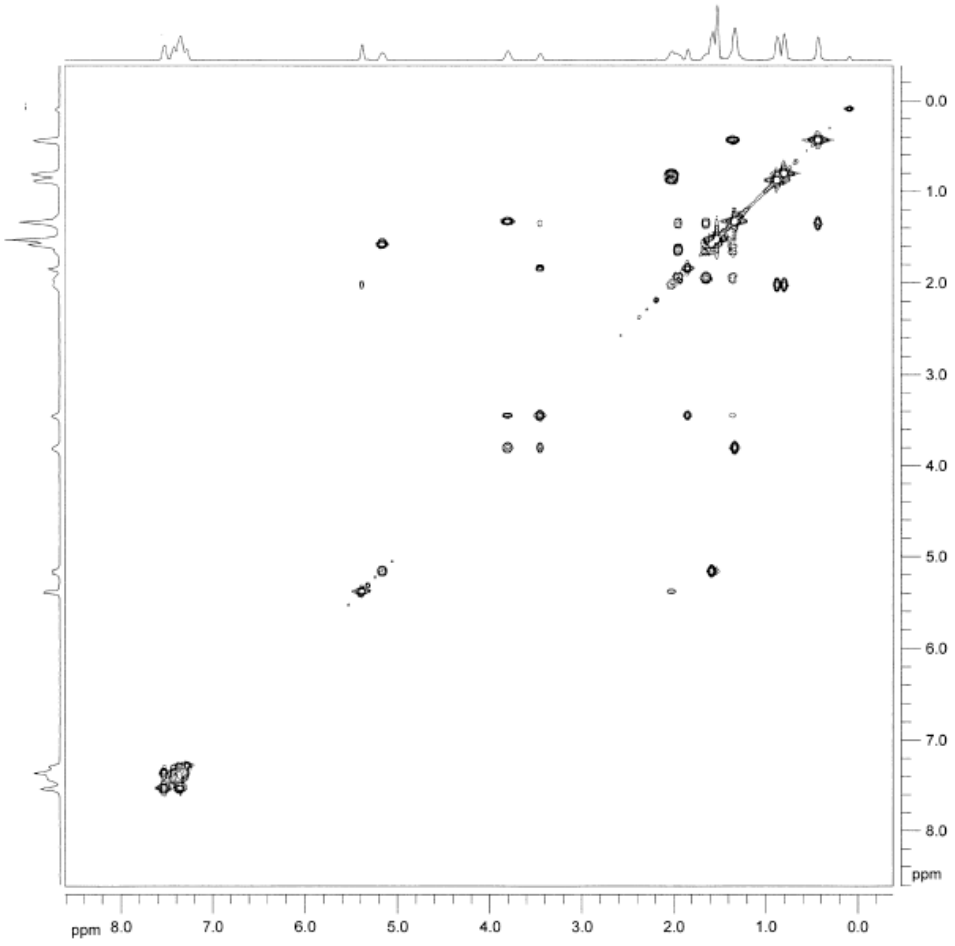
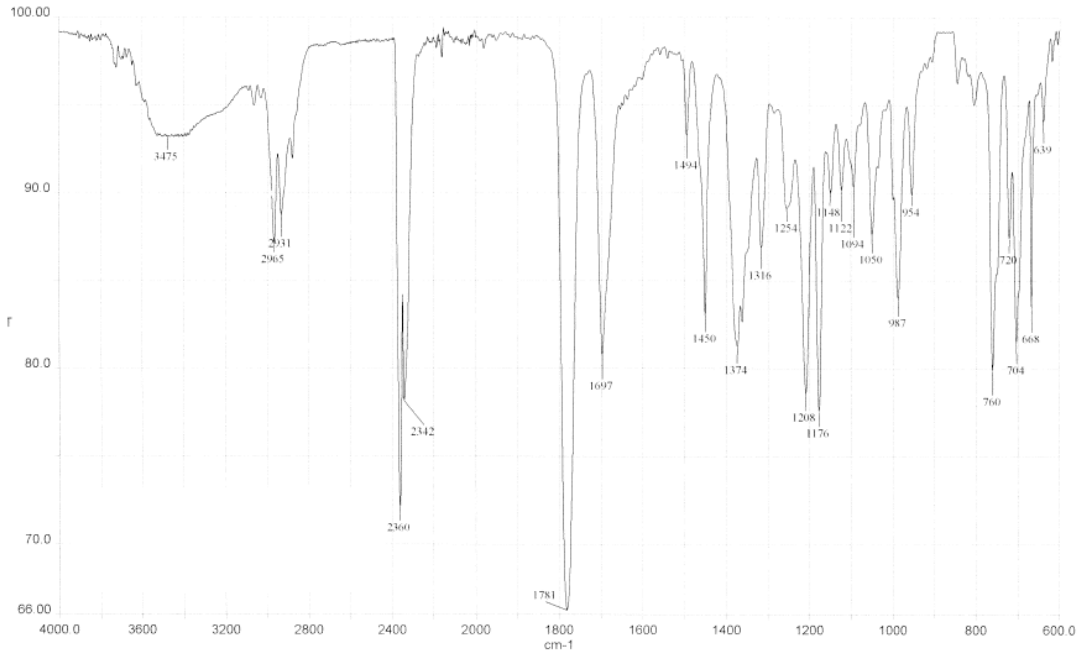
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
200
150
100
50


0.0
1.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
ppm
ppm 8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0


100.00
90.0
80.0
70.0
66.00
T
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1
3475
2931
2965
2342
2360
1781
1697
1494
1450
1374
1316
1254
1208
1176
1148
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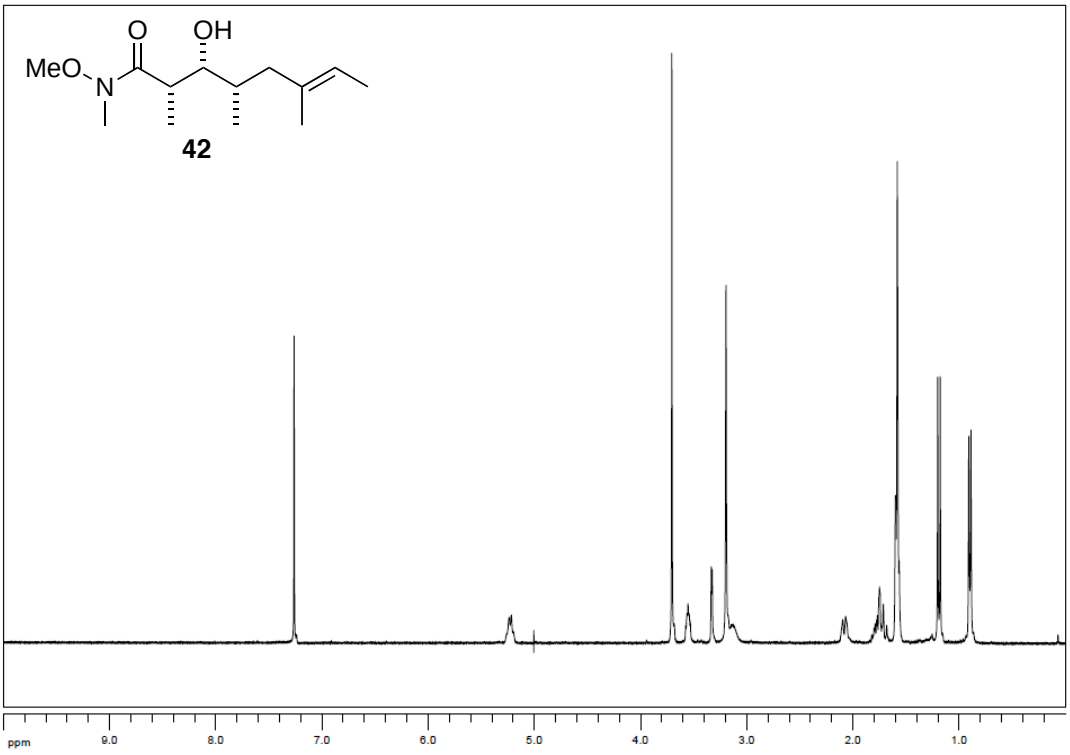
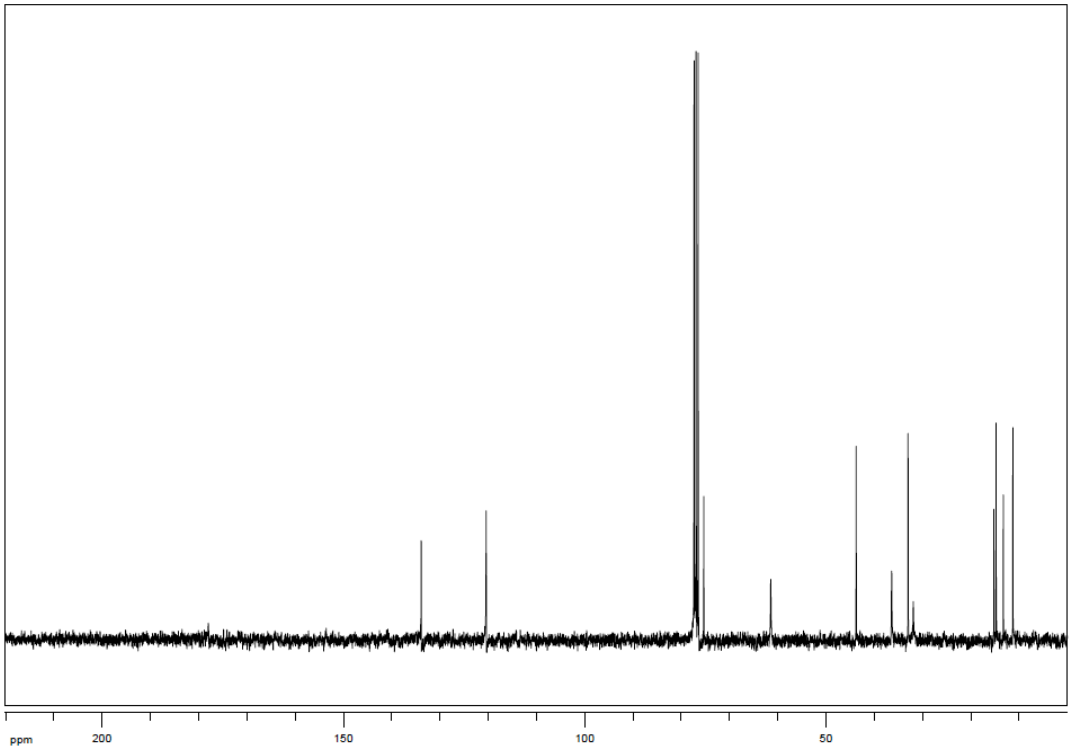
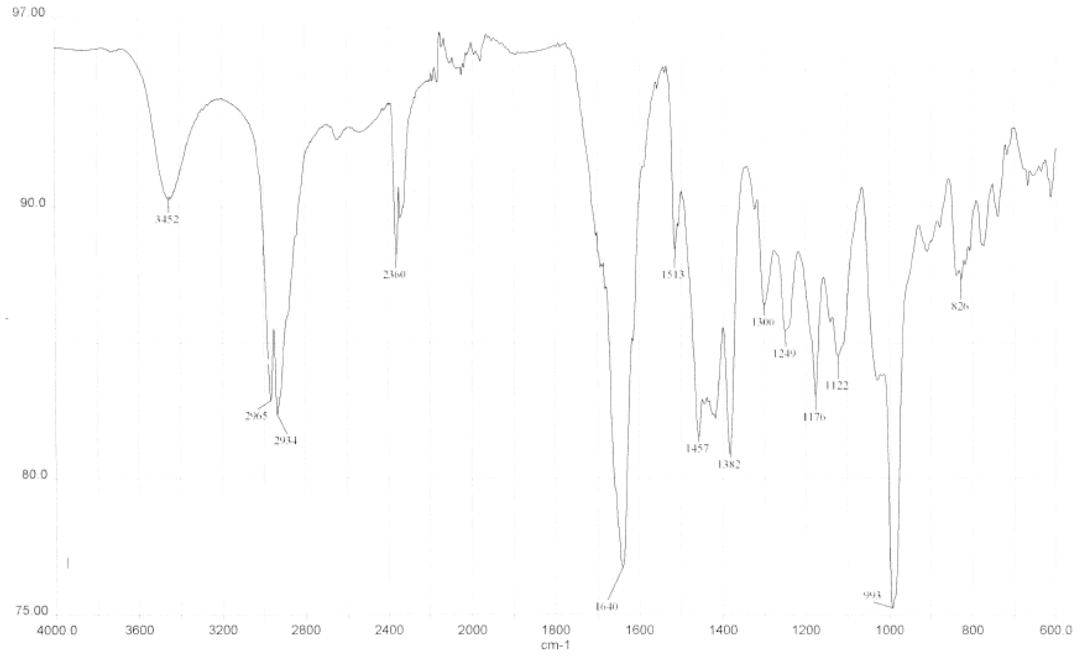
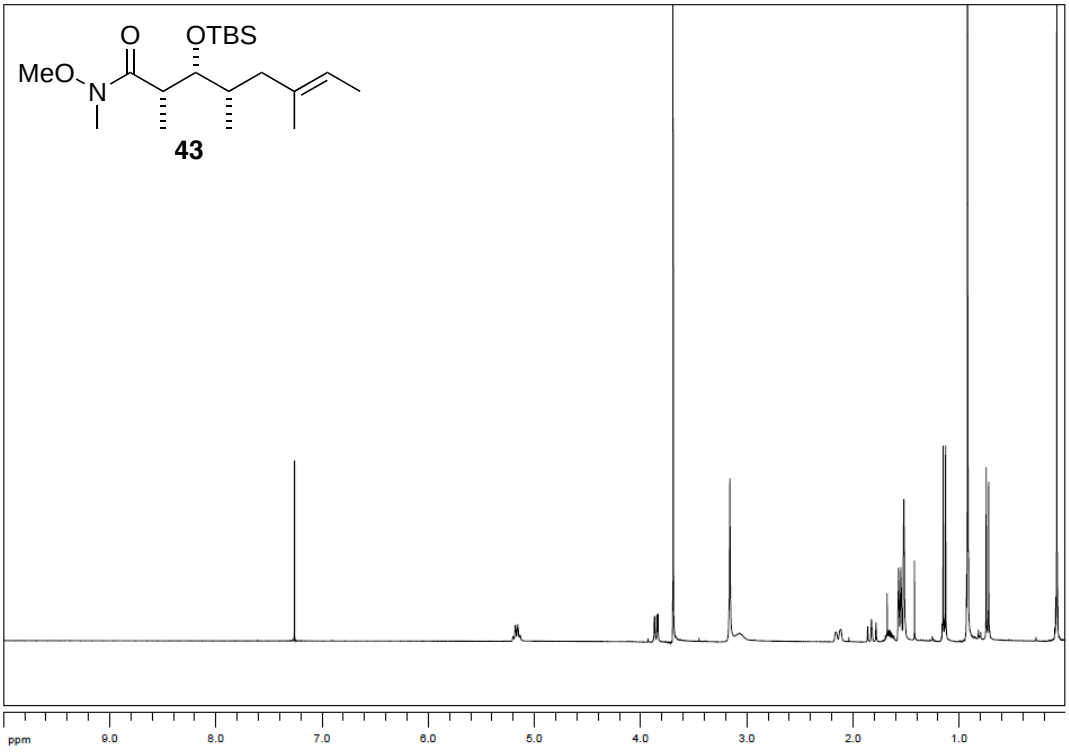
1094
1050
987
954
760
720
704
668
639


MeO
N
O
OH
42
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
200
150
100
50


97.00
90.0
80.0
75.00
3452
2965
2934
2360
1640
1513
1457
1382
1300
1249
1176
1122
993
826
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1



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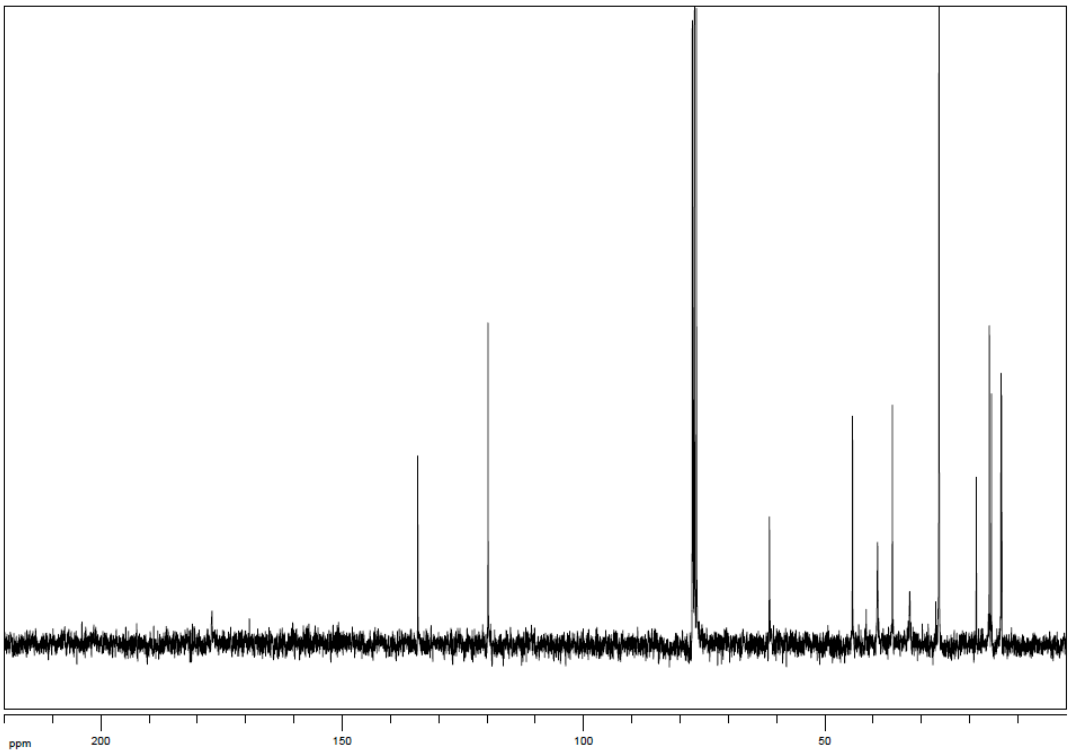
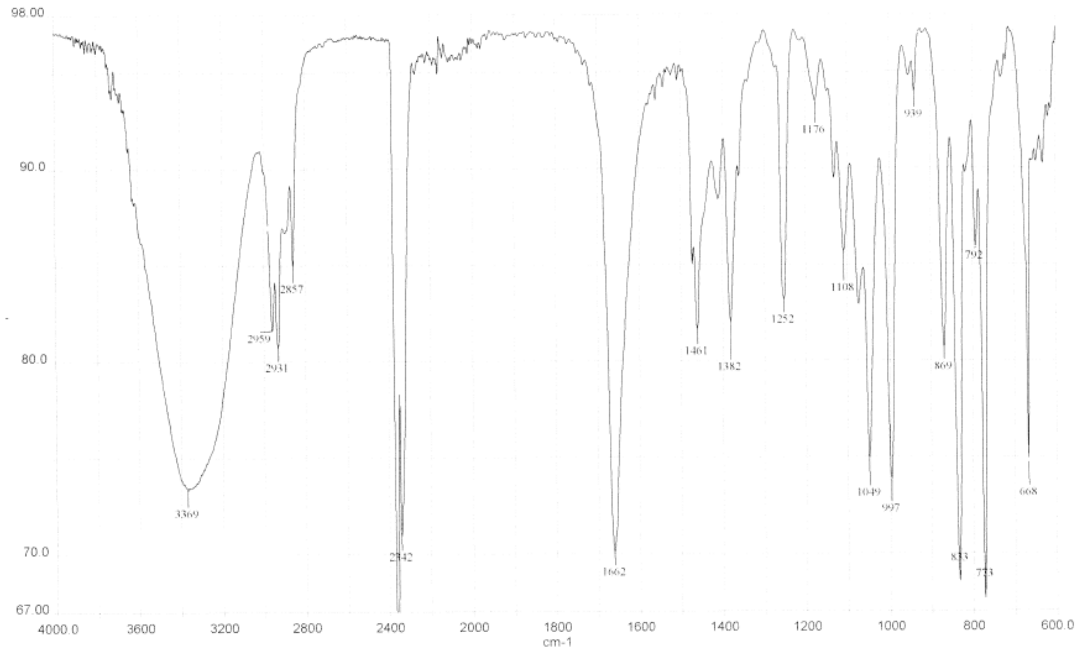
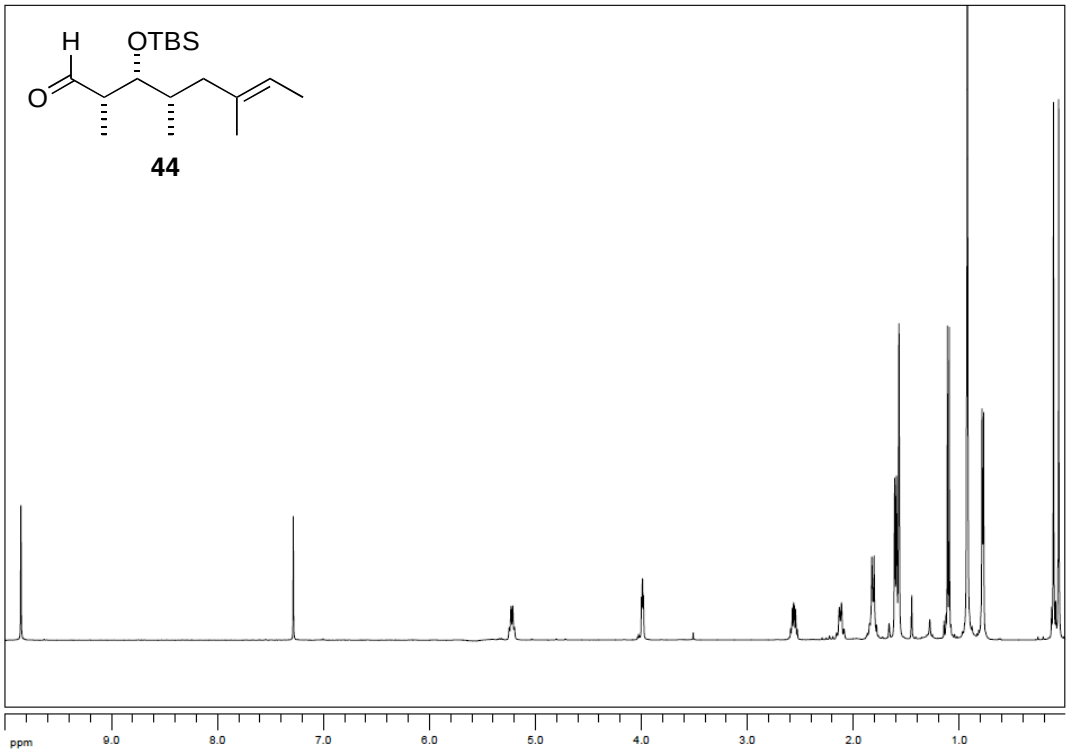



98.00
90.0
80.0
70.0
67.00
4000.0
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600.0
cm-1
3369
2959
2931
2857
2842
1662
1461
1382
1252
1176
1108
1049
997
939
869
792
668
833
773


H
OTBS
O
44
ppm
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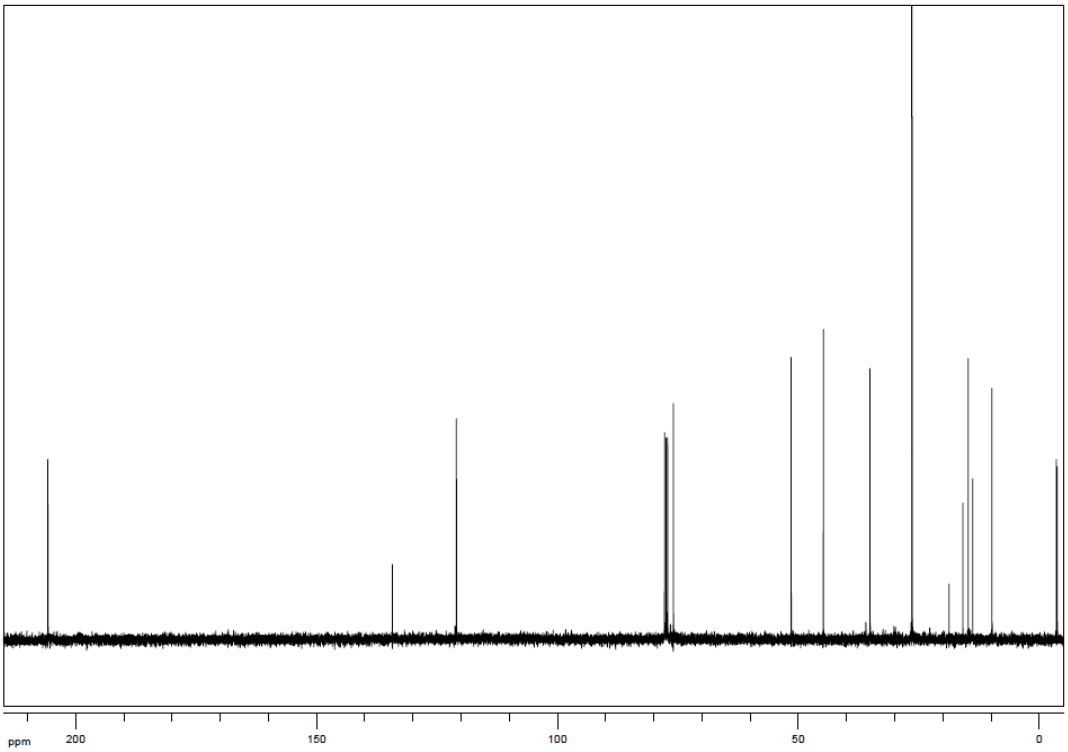
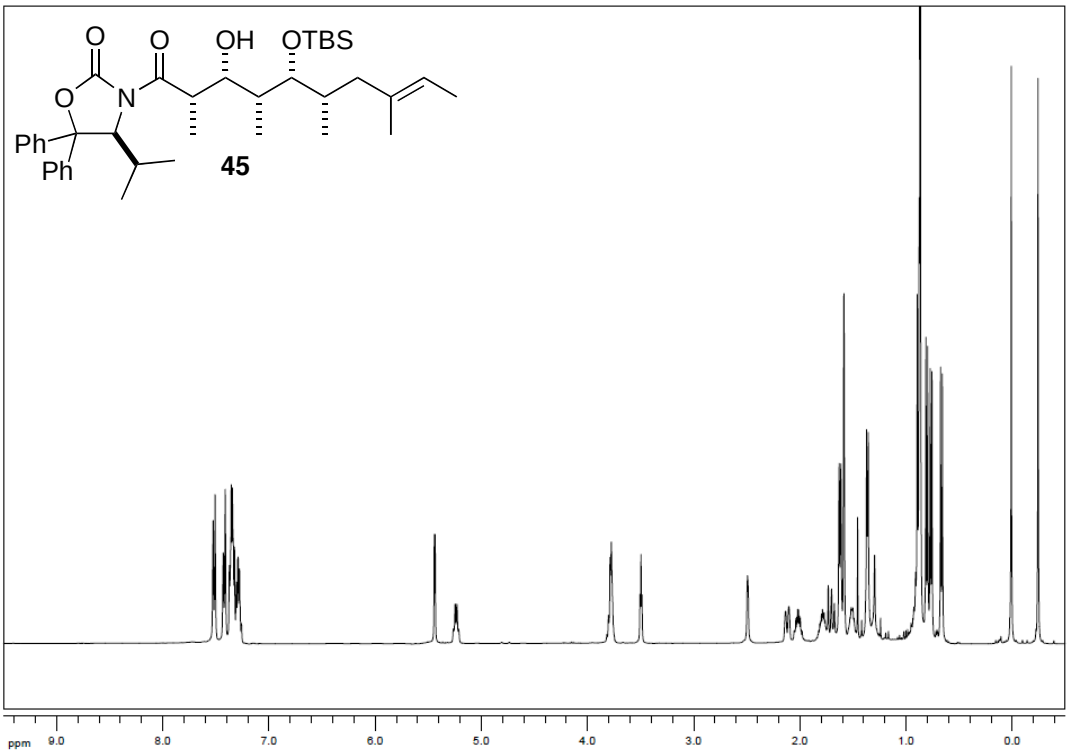
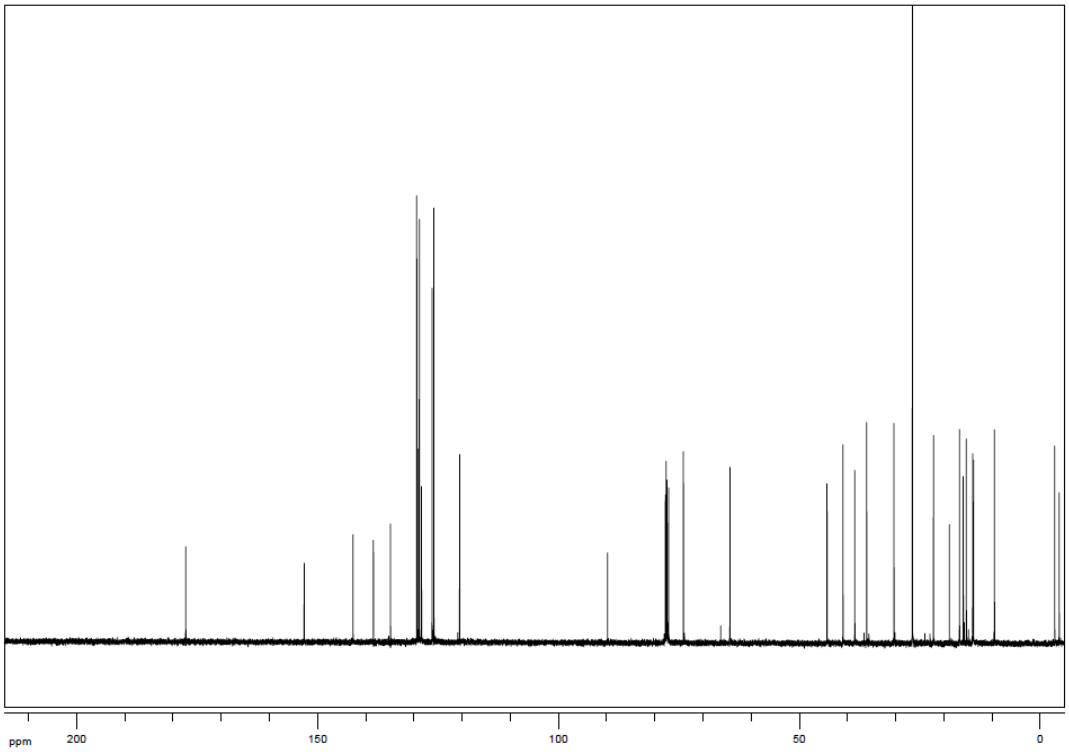
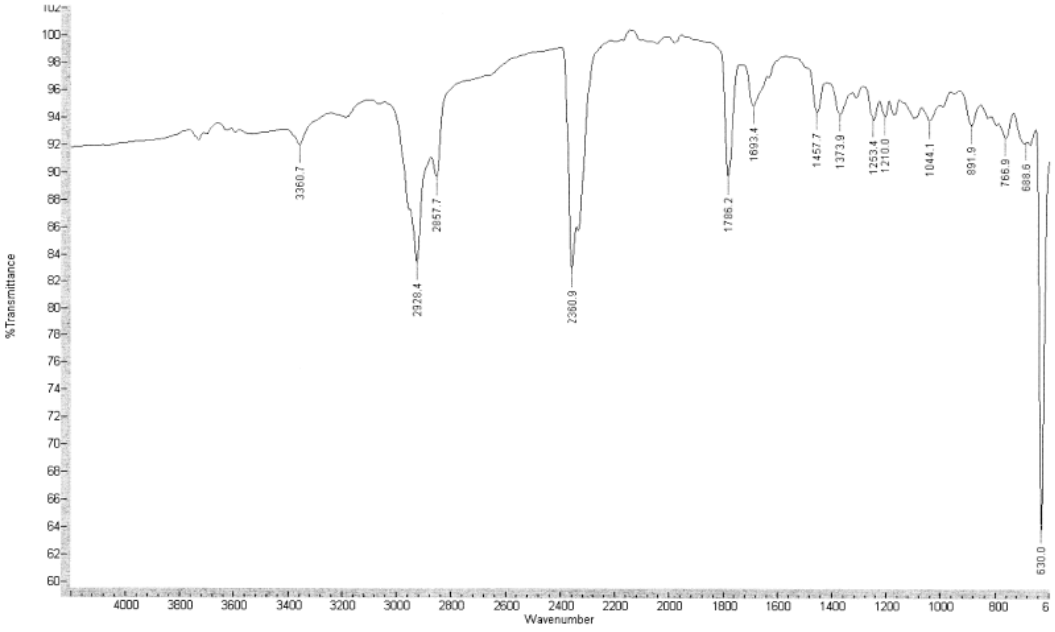
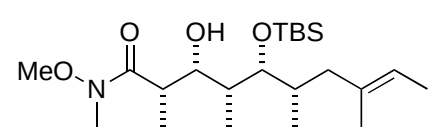
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
200
150
100
50
0


O
O
OH
OTBS
O
N
Ph
Ph
45
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0

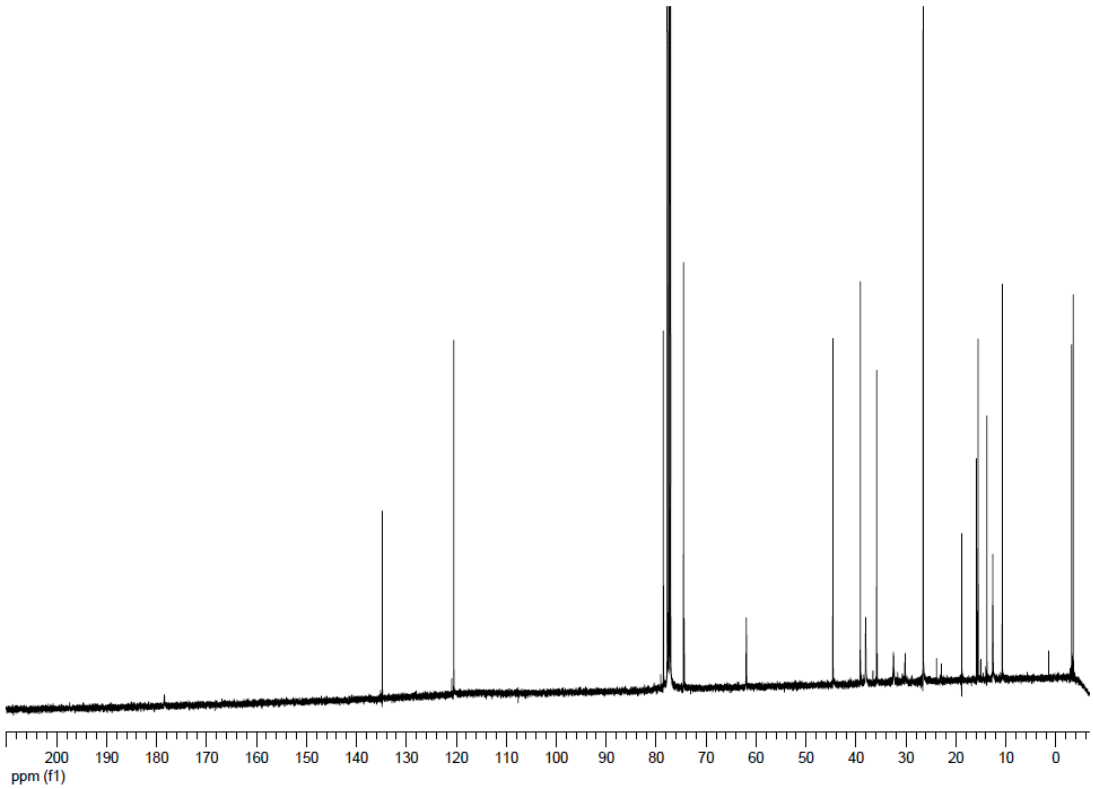

ppm
200
150
100
50
0

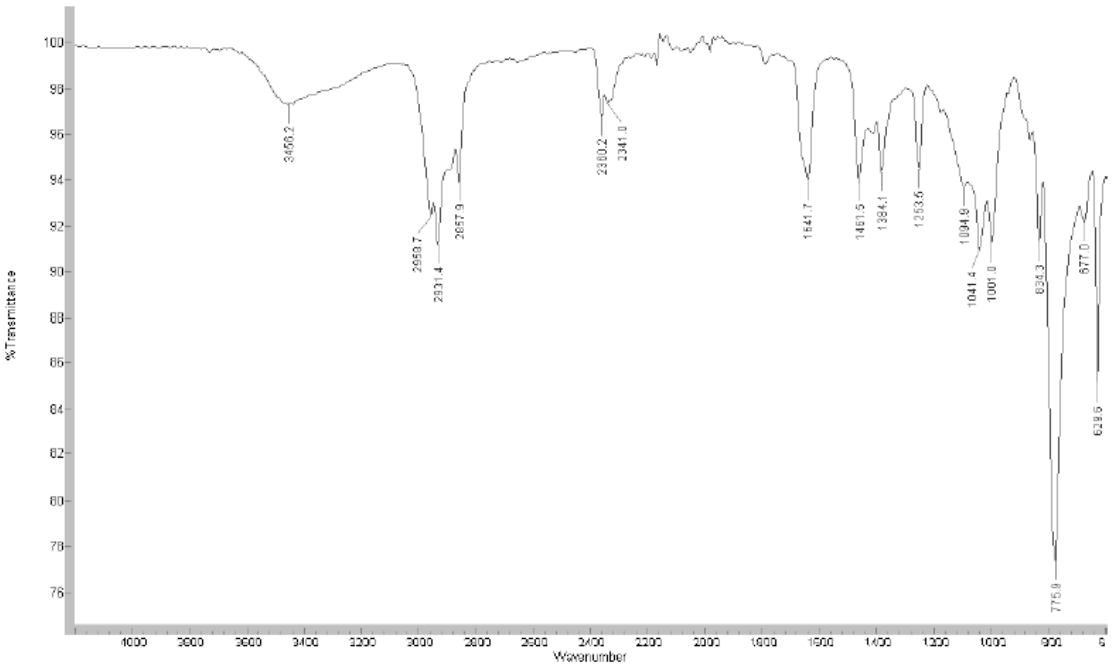

%Transmittance
Wavenumber
3360.7
2928.4
2857.7
2360.9
1786.2
1693.4
1457.7
1373.9
1253.4
1210.0
1044.1
891.9
766.9
688.6
630.0

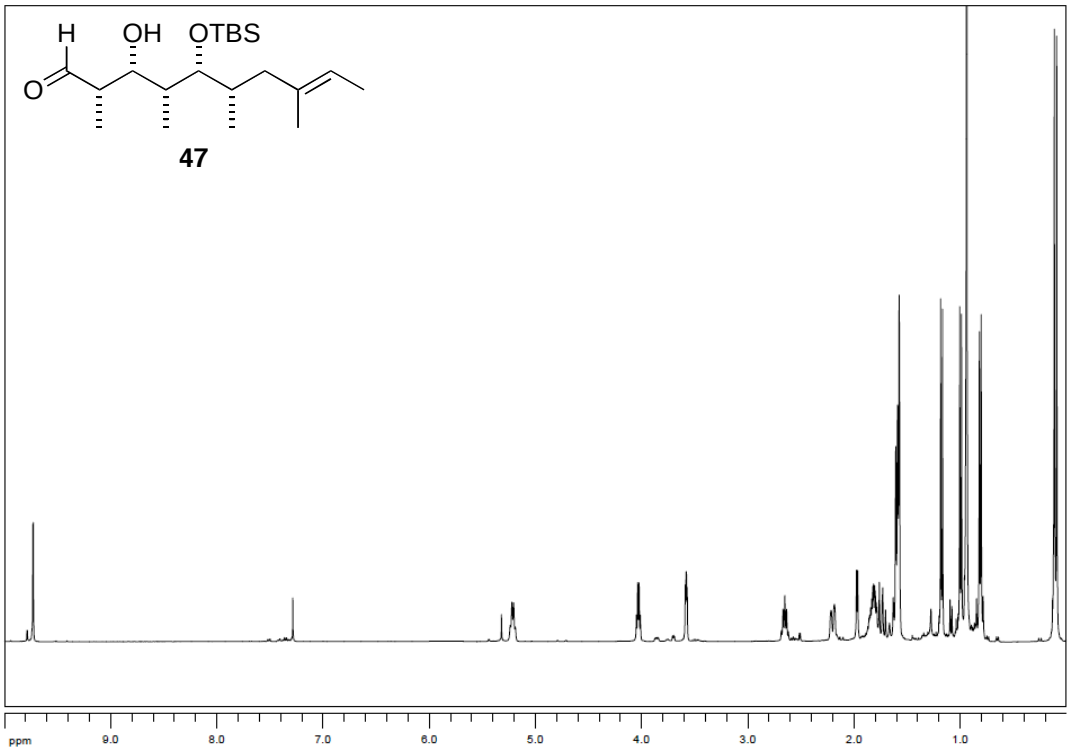

O
OH
OTBS
MeO
N

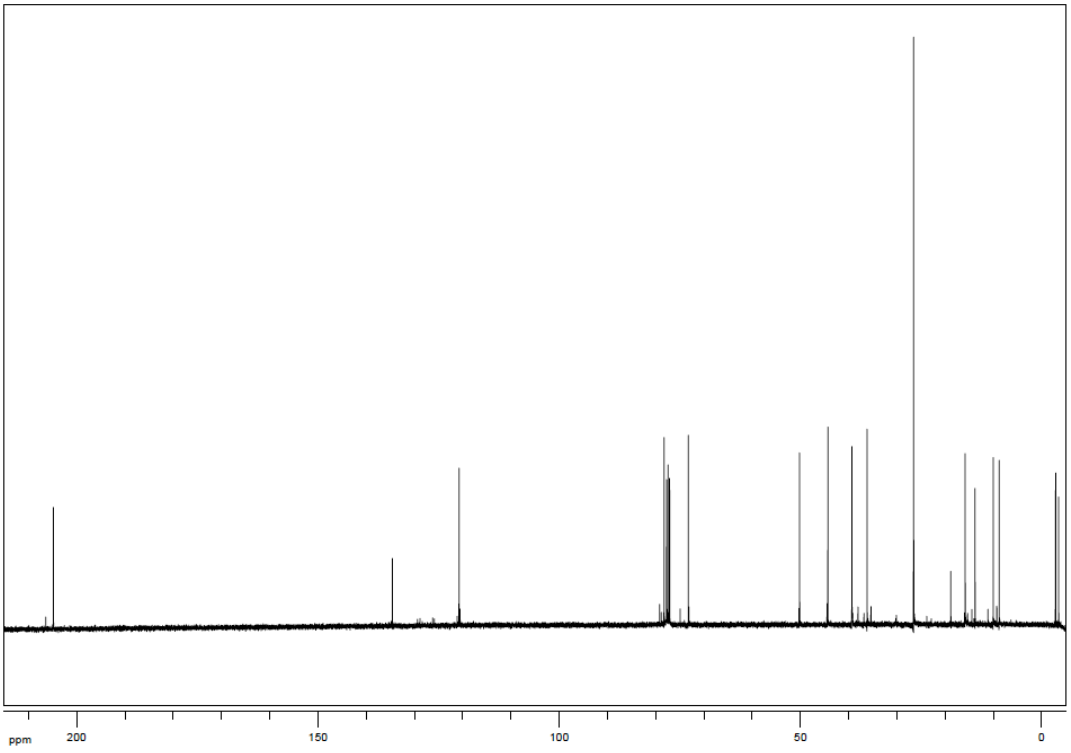
46

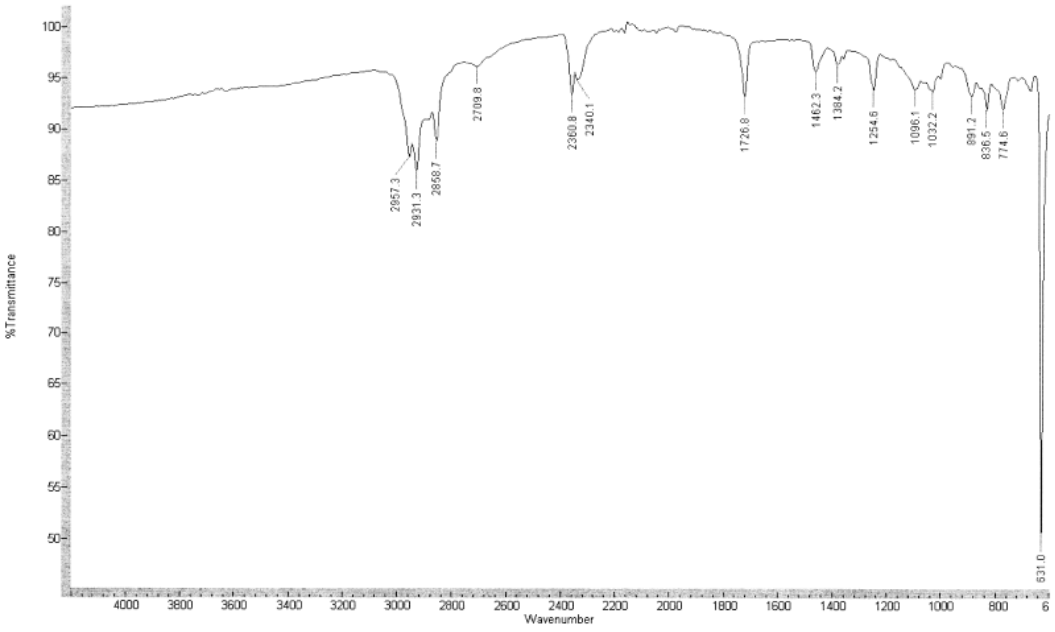
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

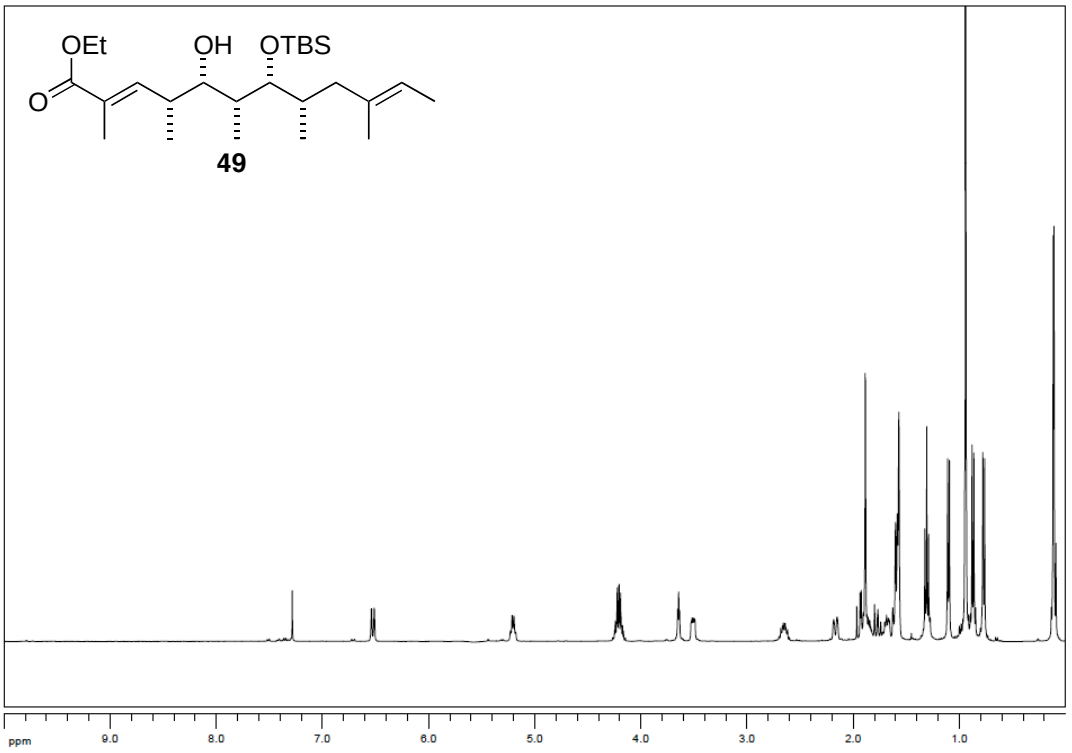

200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)


%Transmittance
Wavenumber
3456.2
2959.7
2931.4
2857.9
2360.2
2341.0
1641.7
1461.5
1384.1
1253.6
1094.9
1041.4
1001.0
838.3
775.9
677.0
629.5

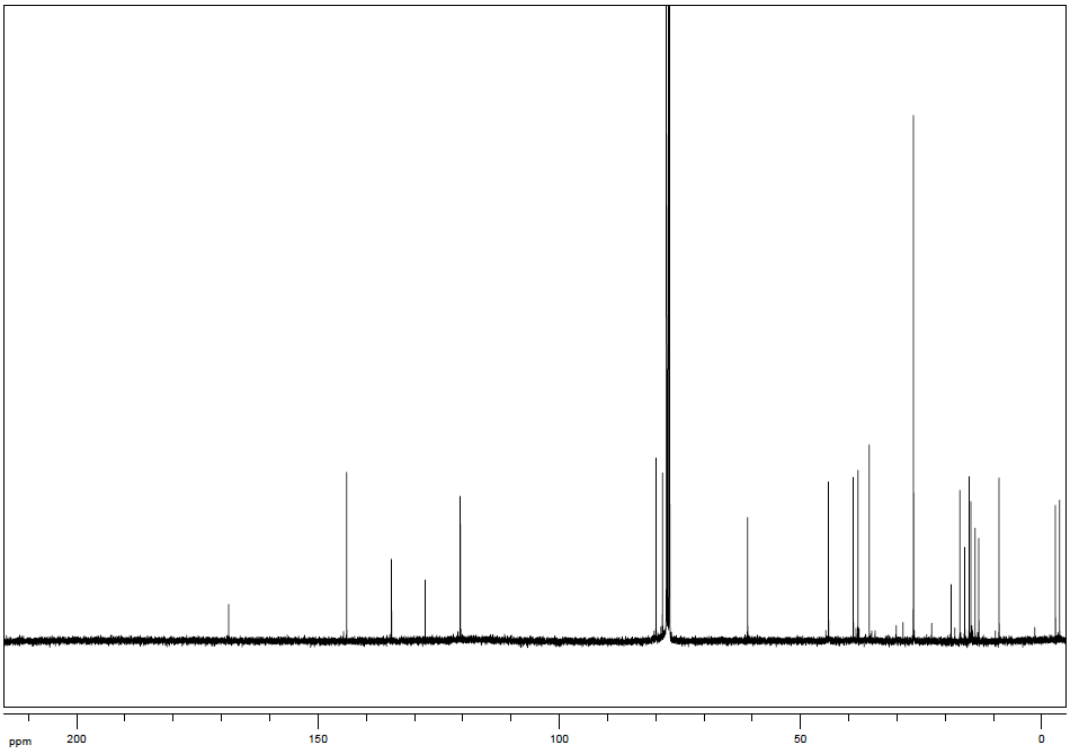

H
OH
OTBS
O
47
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0

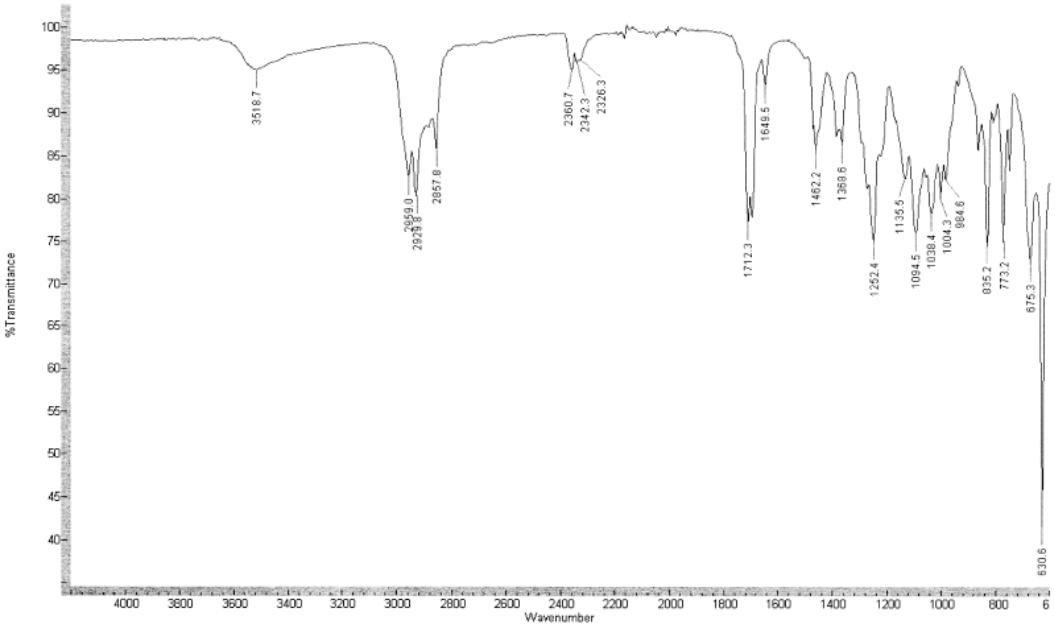

ppm
200
150
100
50
0


%Transmittance
Wavenumber
2957.3
2931.3
2858.7
2709.8
2360.8
2340.1
1726.8
1462.3
1384.2
1254.6
1096.1
1032.2
891.2
836.5
774.6
631.0



49




%Transmittance
Wavenumber
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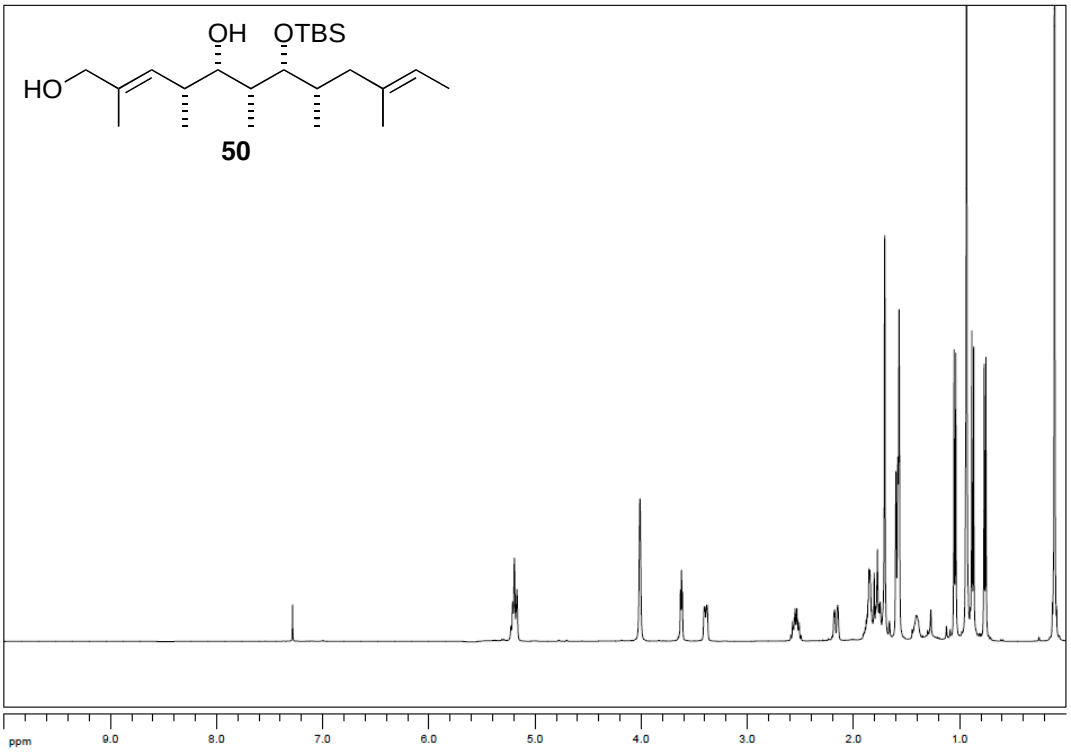
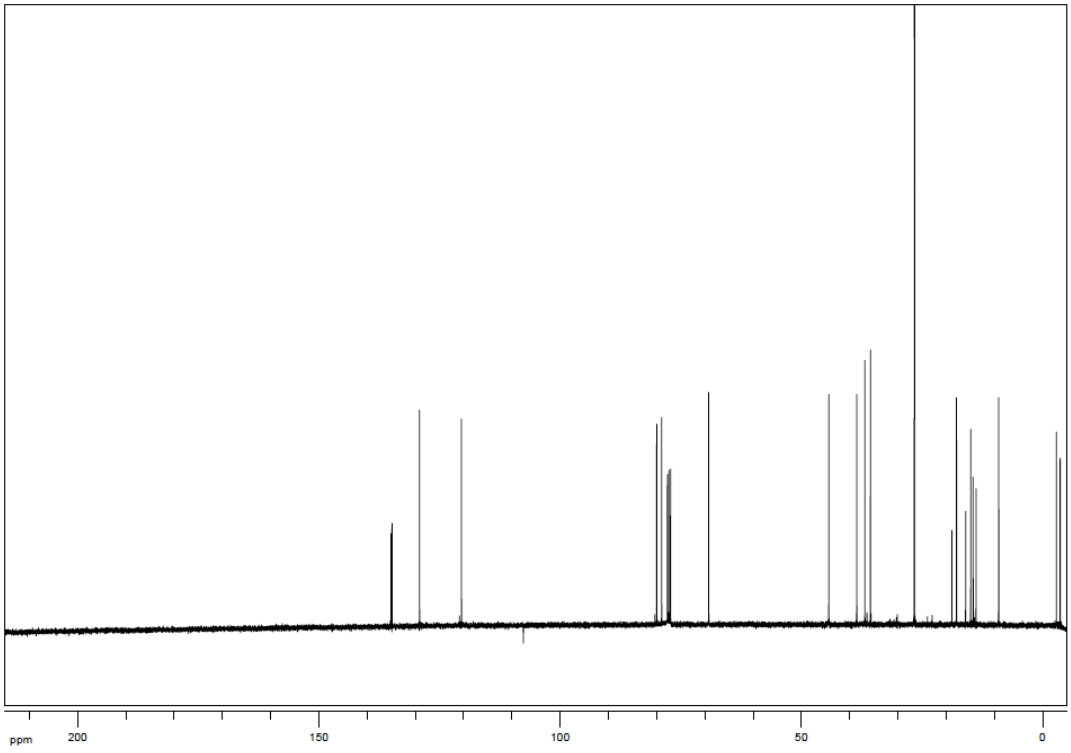
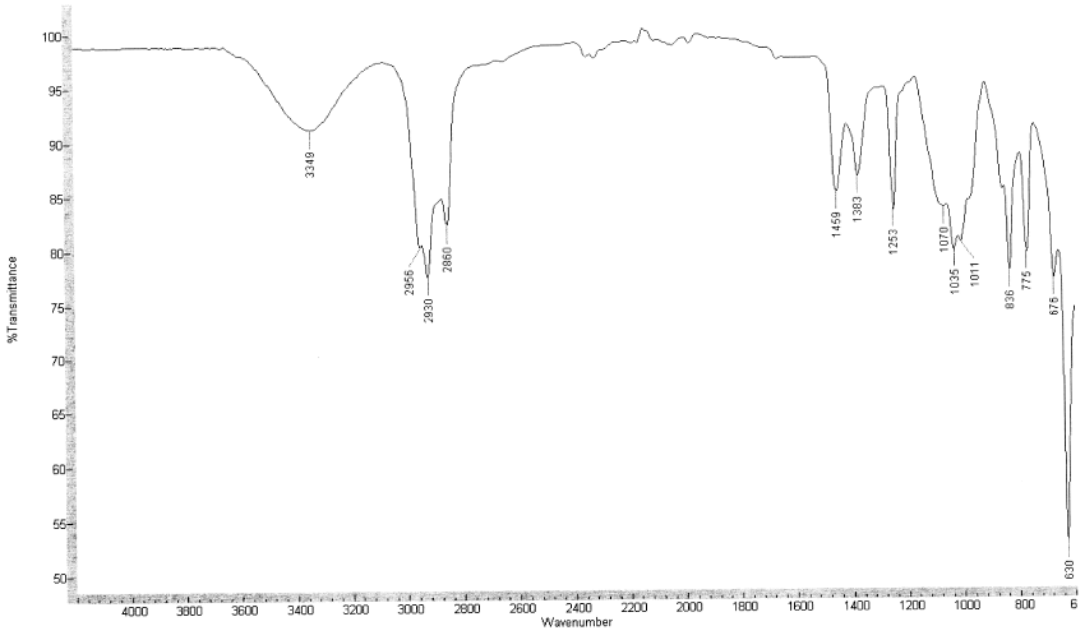
2923.0
2857.8
2360.7
2342.3
2326.3
1712.3
1649.5
1462.2
1368.6
1252.4
1135.5
1094.5
1038.4
1004.3
984.6
835.2
773.2
675.3
630.6


OH
OTBS
HO
50
ppm


ppm
200
150
100
50
0


%Transmittance
Wavenumber
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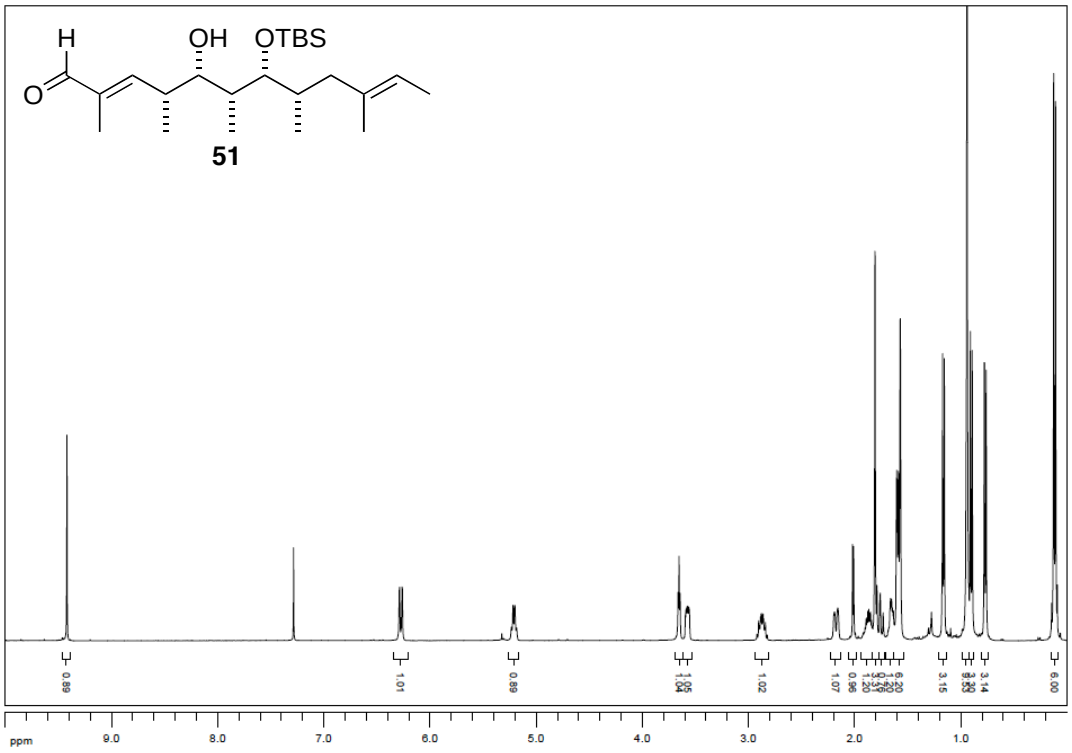
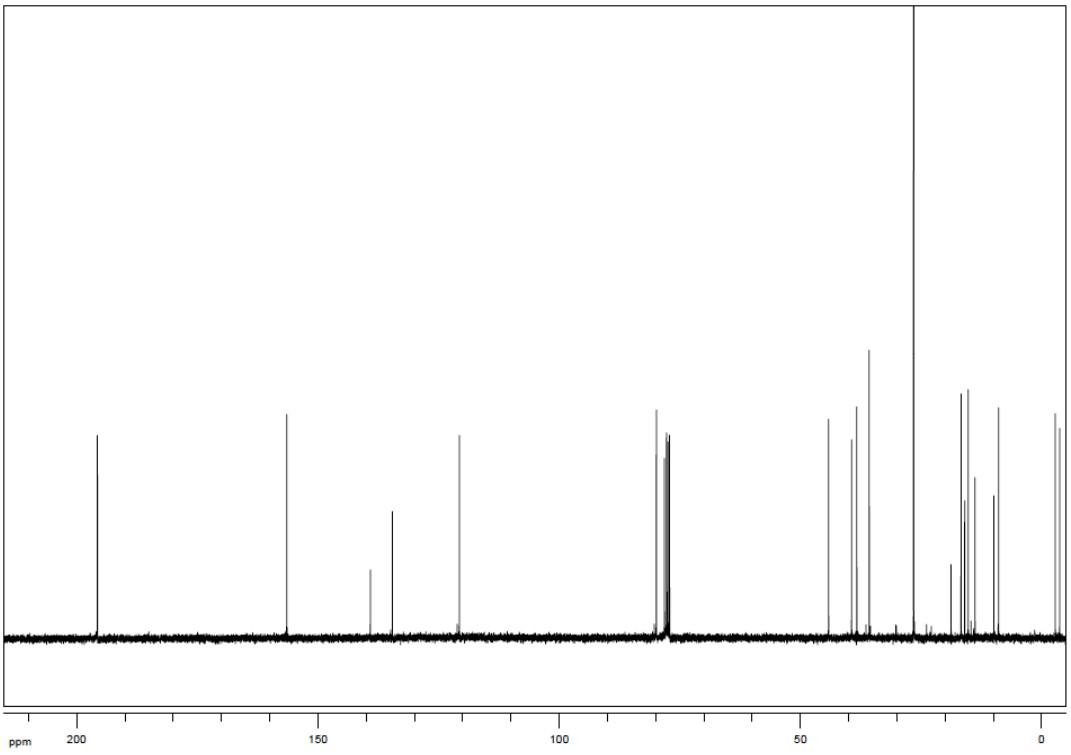
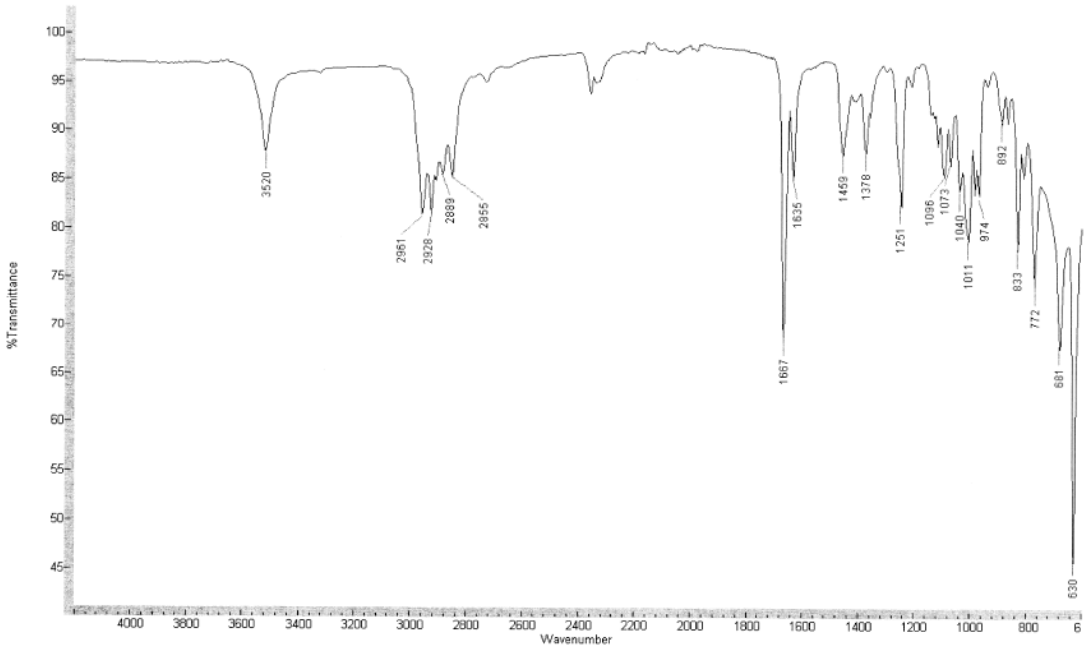
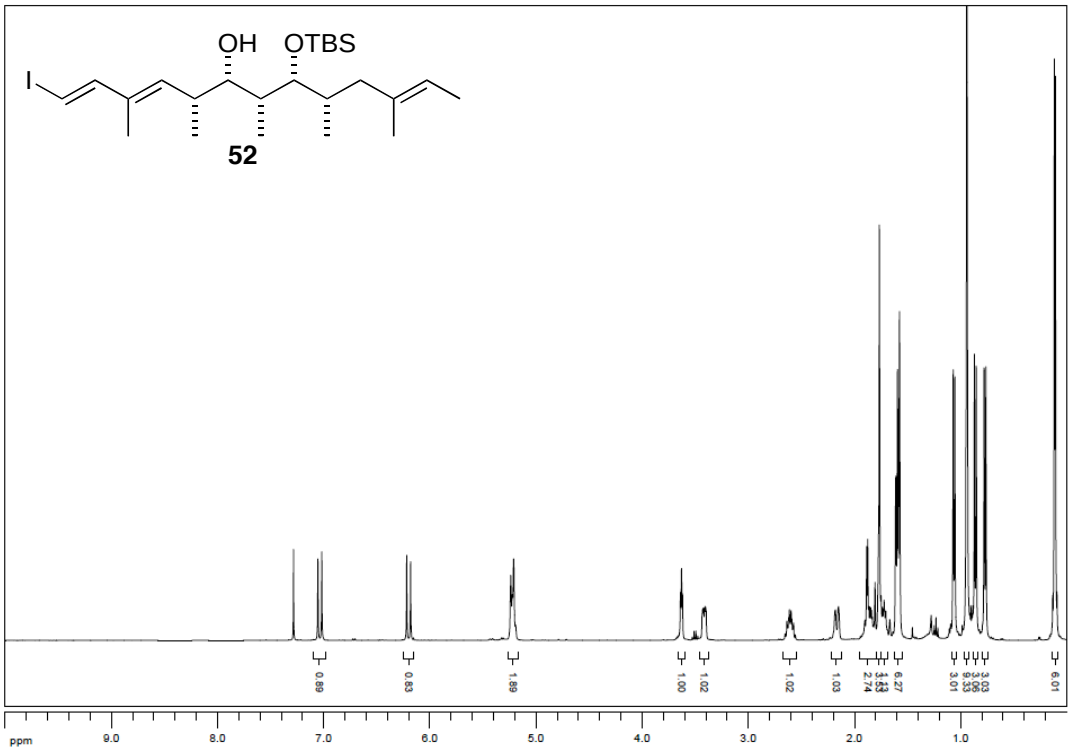
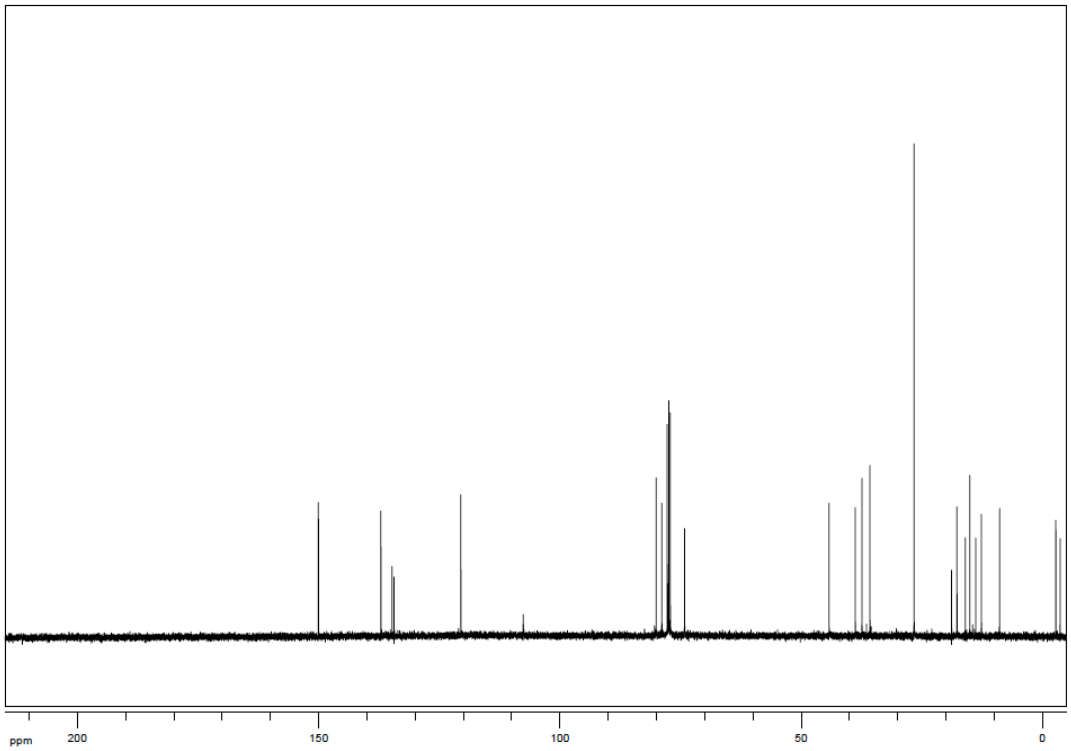
2956
2930
2860
1459
1383
1253
1070
1035
1011
836
775
675
630

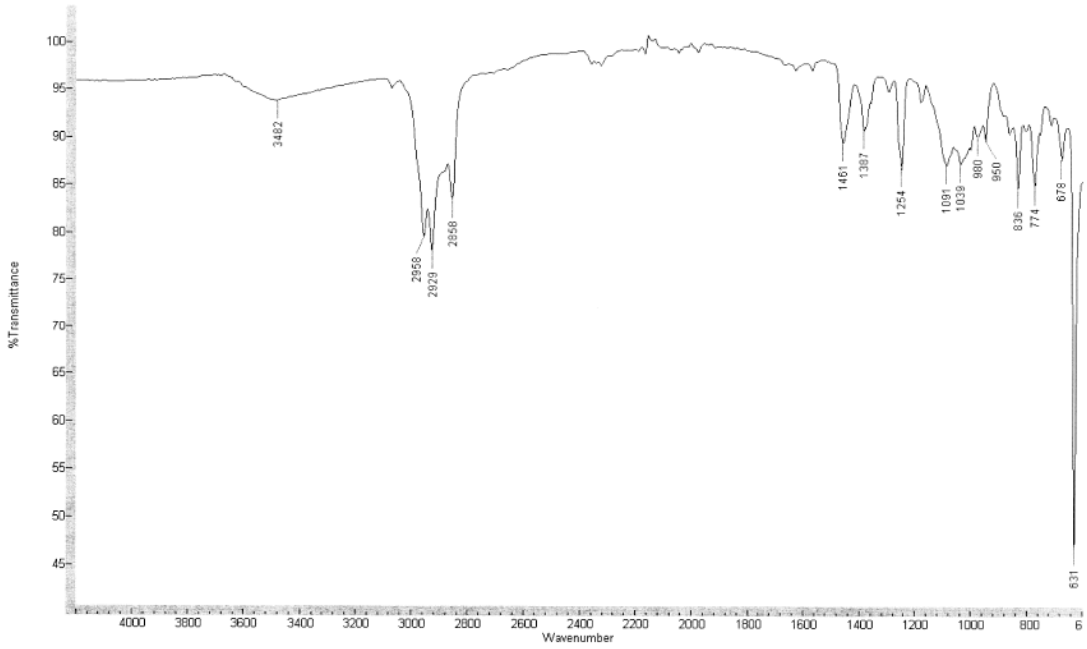

H
OH
OTBS
O
51
0.89
1.01
0.89
1.05
1.04
1.02
1.07
0.96
1.20
6.20
3.15
3.14
6.00
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0

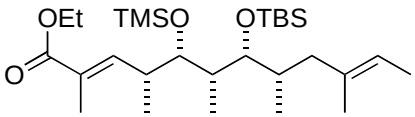

ppm
200
150
100
50
0


%Transmittance
Wavenumber
3520
2961
2928
2889
2955
1667
1635
1459
1378
1251
1096
1073
1040
1011
974
892
833
772
681
630


OH
OTBS
I
52
ppm

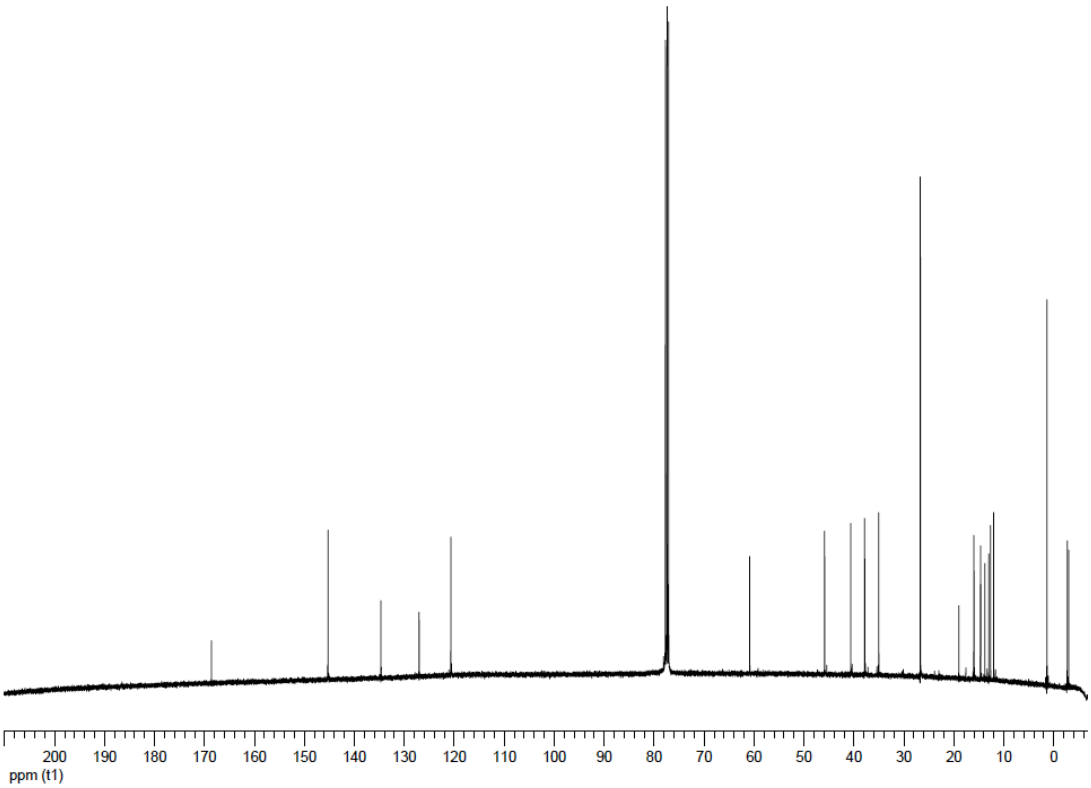

ppm
200
150
100
50
0

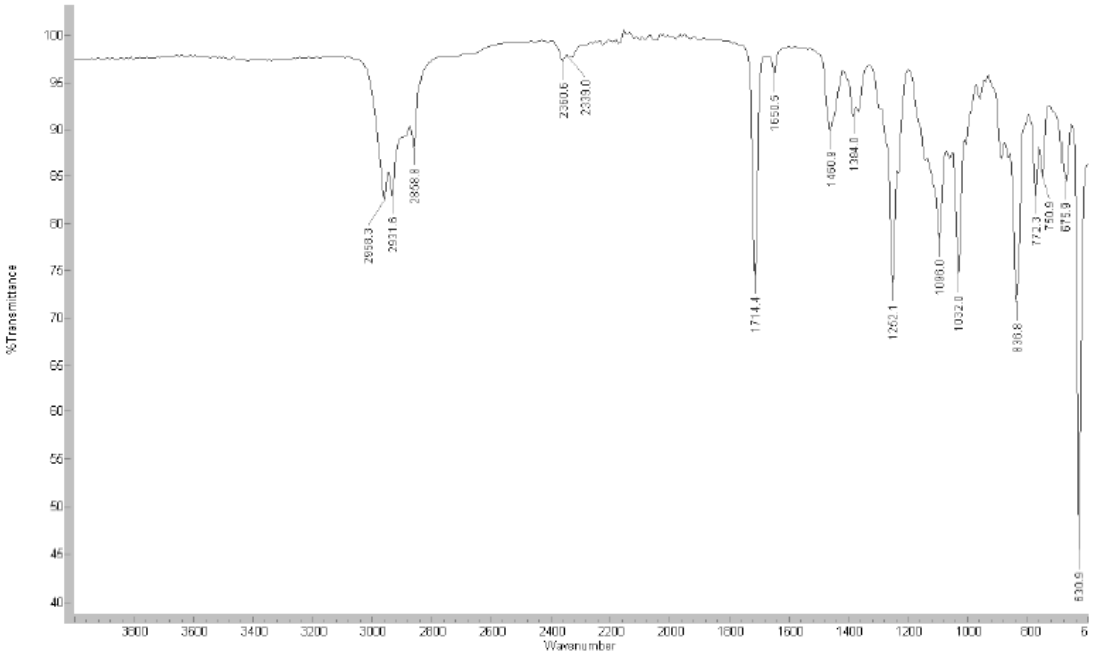

%Transmittance
100
95
90
85
80
75
70
65
60
55
50
45
3482
2958
2929
2856
1461
1387
1254
1091
1039
980
950
836
774
678
631
4000
3800
3600
3400
3200
3000
2800
2600
2400
2200
2000
1800
1600
1400
1200
1000
800
6
Wavenumber

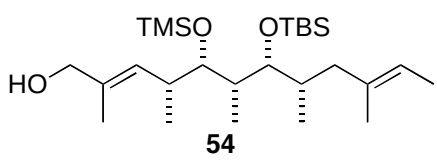

OEt
TMSO
OTBS
O

53

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (t1)

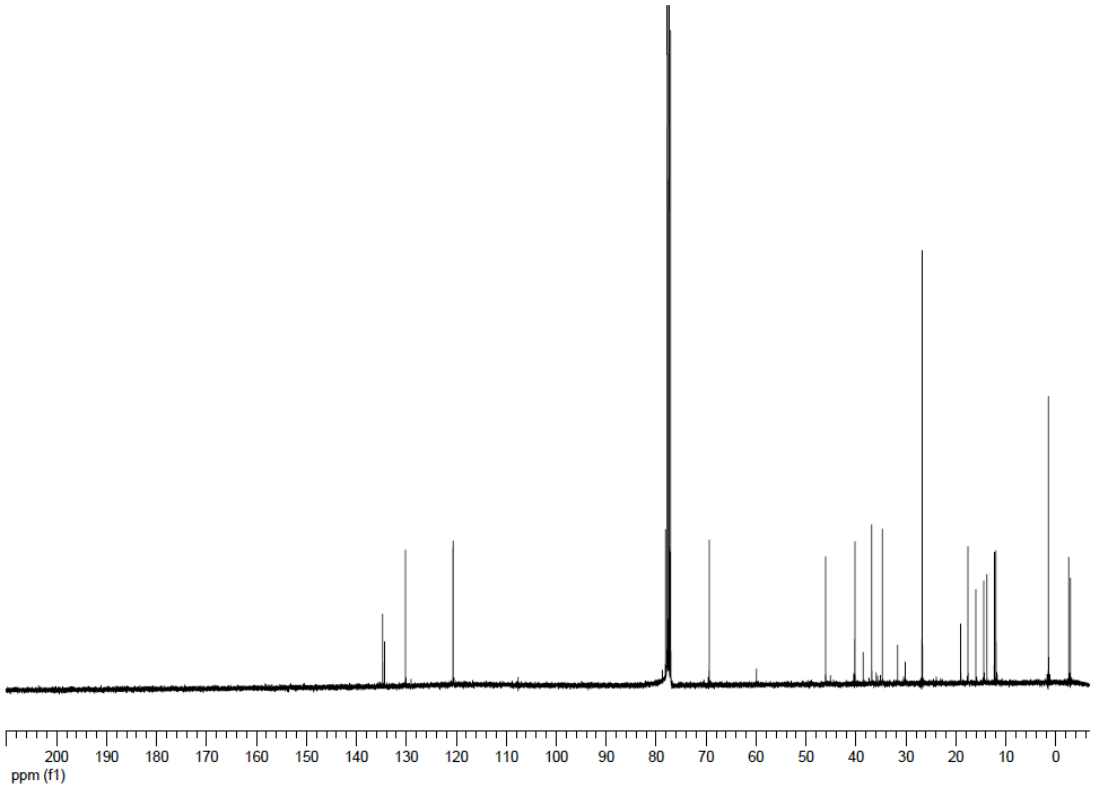

200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (t1)

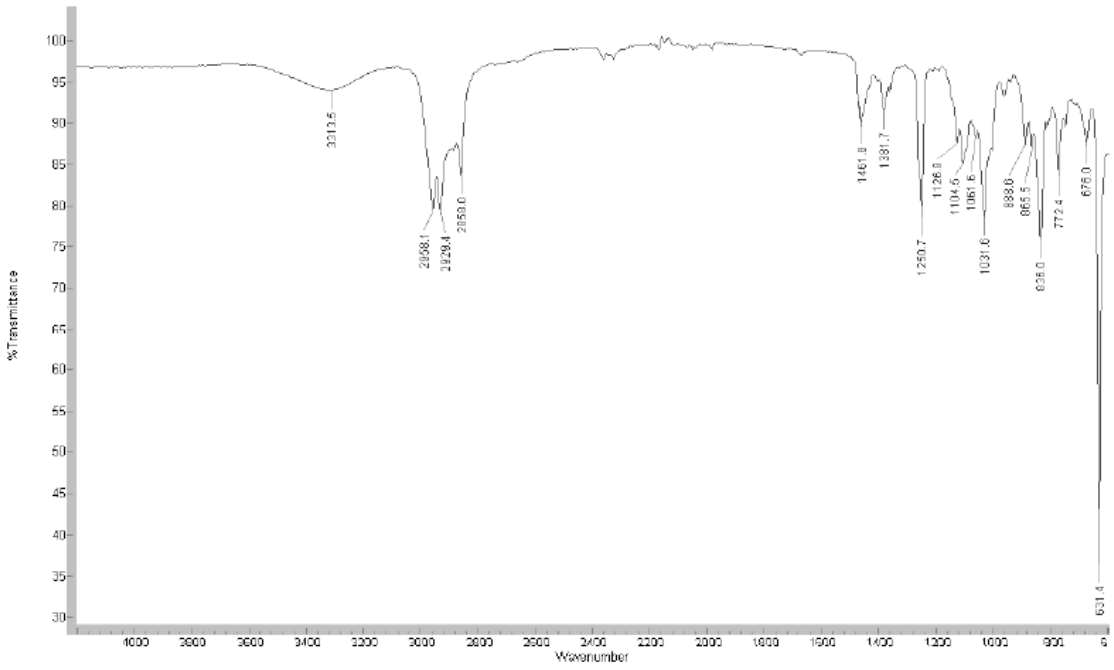

%Transmittance
Wavenumber
2958.3
2931.6
2856.8
2360.6
2339.0
1714.4
1650.5
1460.9
1364.0
1252.1
1086.0
1032.0
838.8
772.3
750.9
675.9
630.9

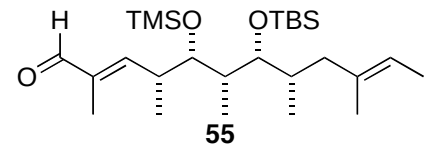

TMSO
OTBS
HO

54

ppm (f1)

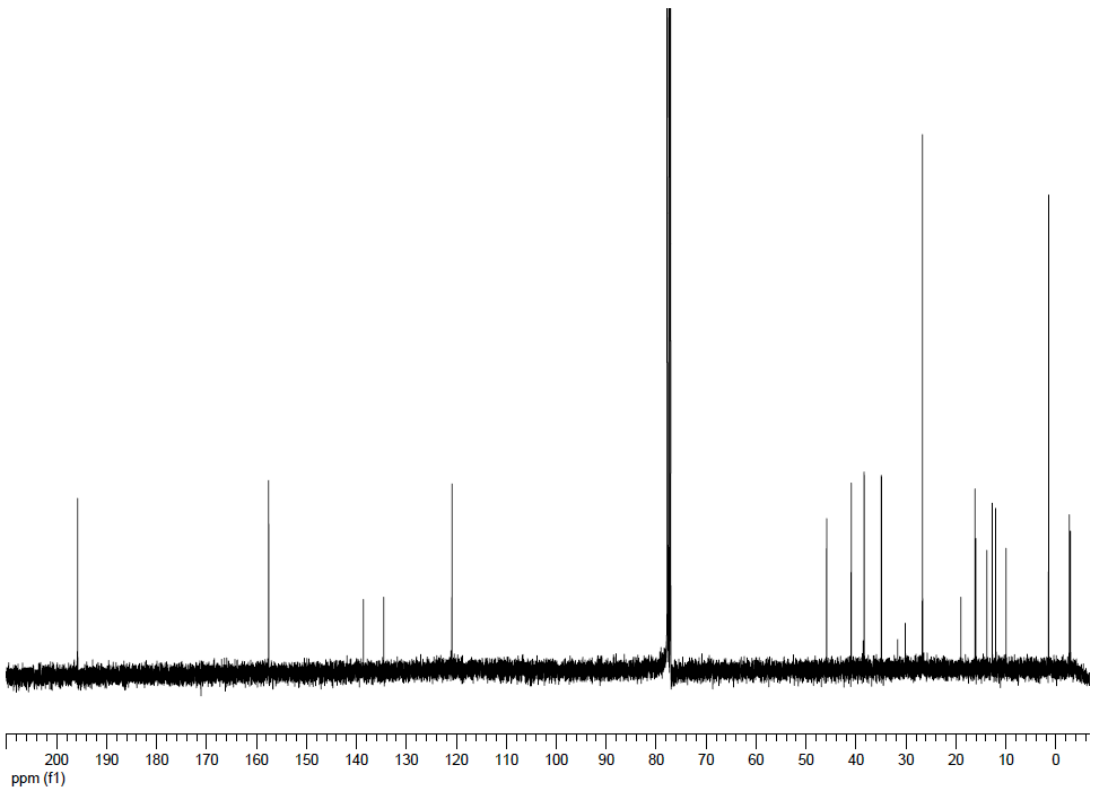

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

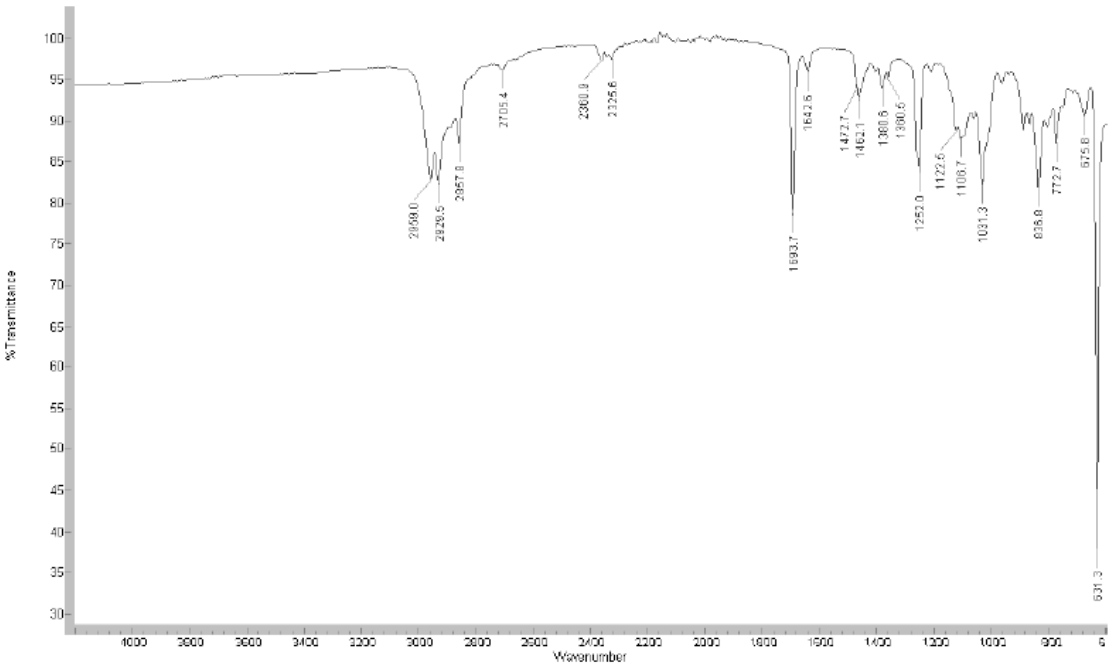

%Transmittance
100 95 90 85 80 75 70 65 60 55 50 45 40 35 30
3313.5
2958.1
2929.4
2859.0
1461.8
1381.7
1250.7
1126.0
1104.6
1061.6
1031.0
888.8
865.5
836.0
772.4
676.0
631.4
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800 6
Wavenumber

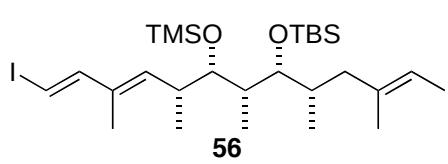

H
TMSO
OTBS
O

55

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

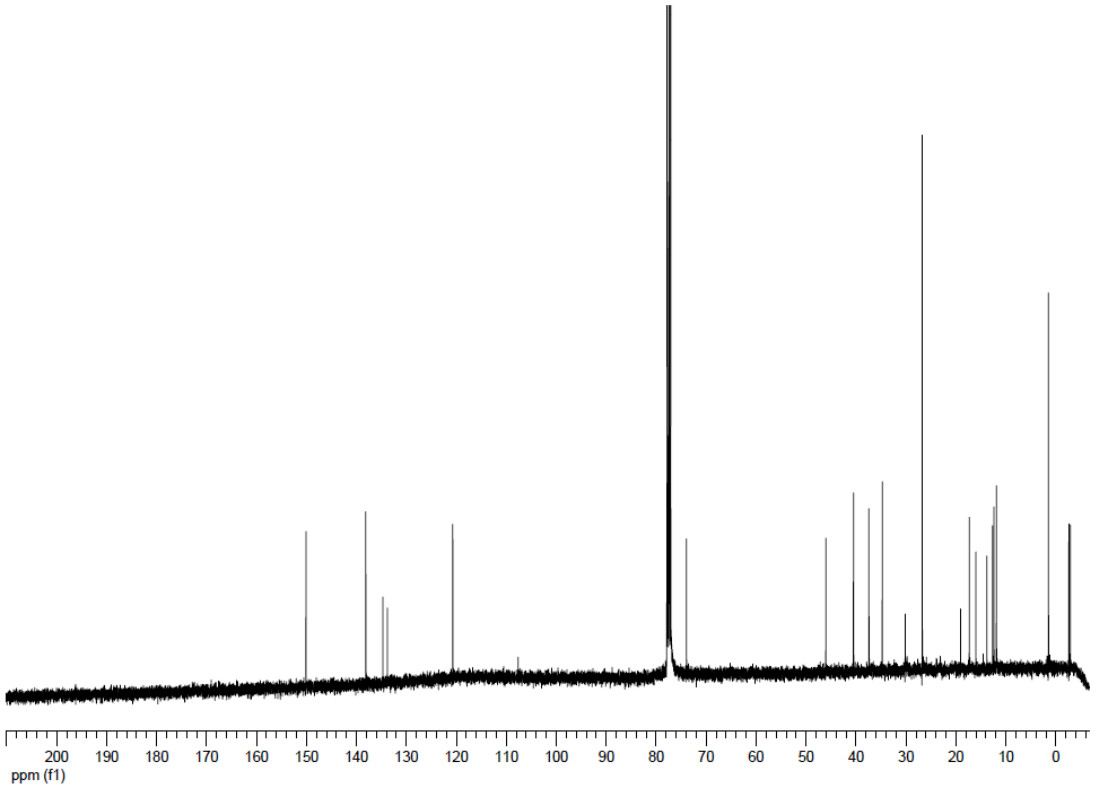

200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)

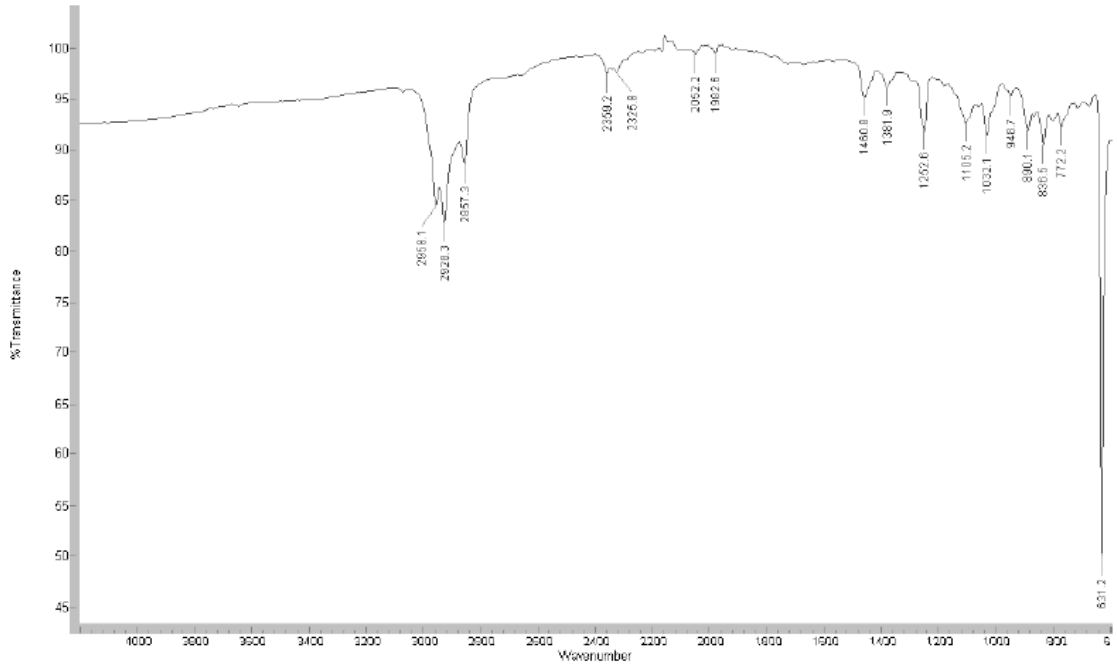

%Transmittance
Wavenumber
2959.0
2926.5
2857.8
2705.4
2360.9
2325.6
1693.7
1642.6
1472.7
1462.1
1388.6
1380.6
1252.0
1122.6
1108.7
1031.3
836.8
772.7
675.8
631.3

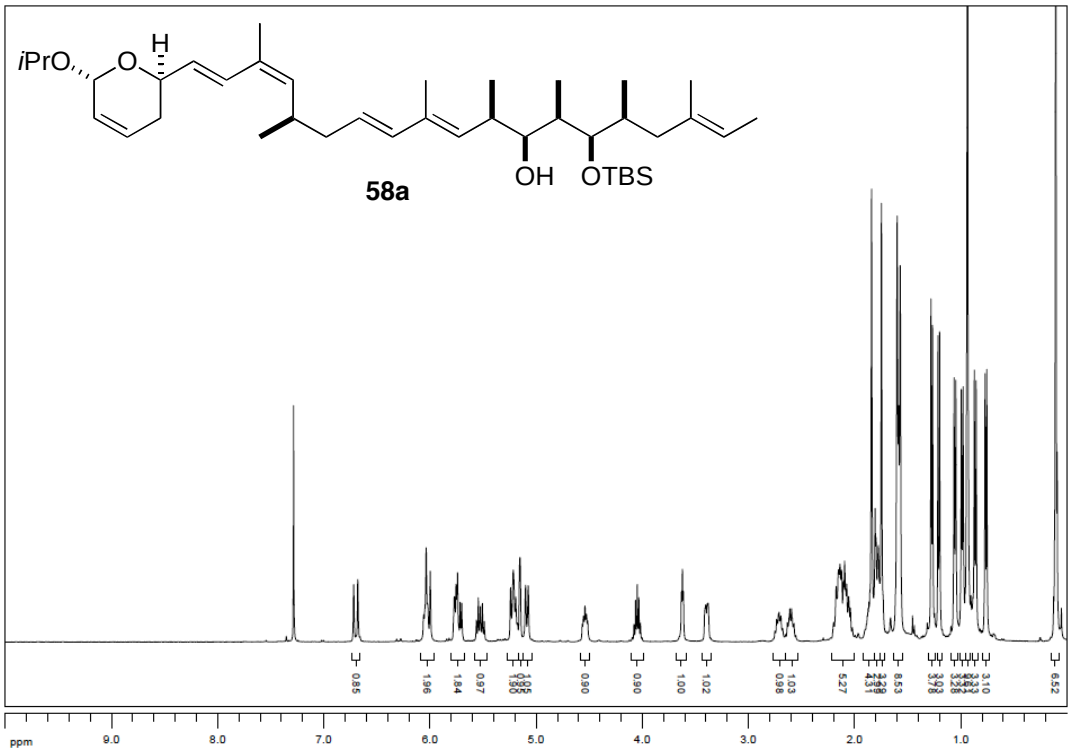

TMSO
OTBS
I

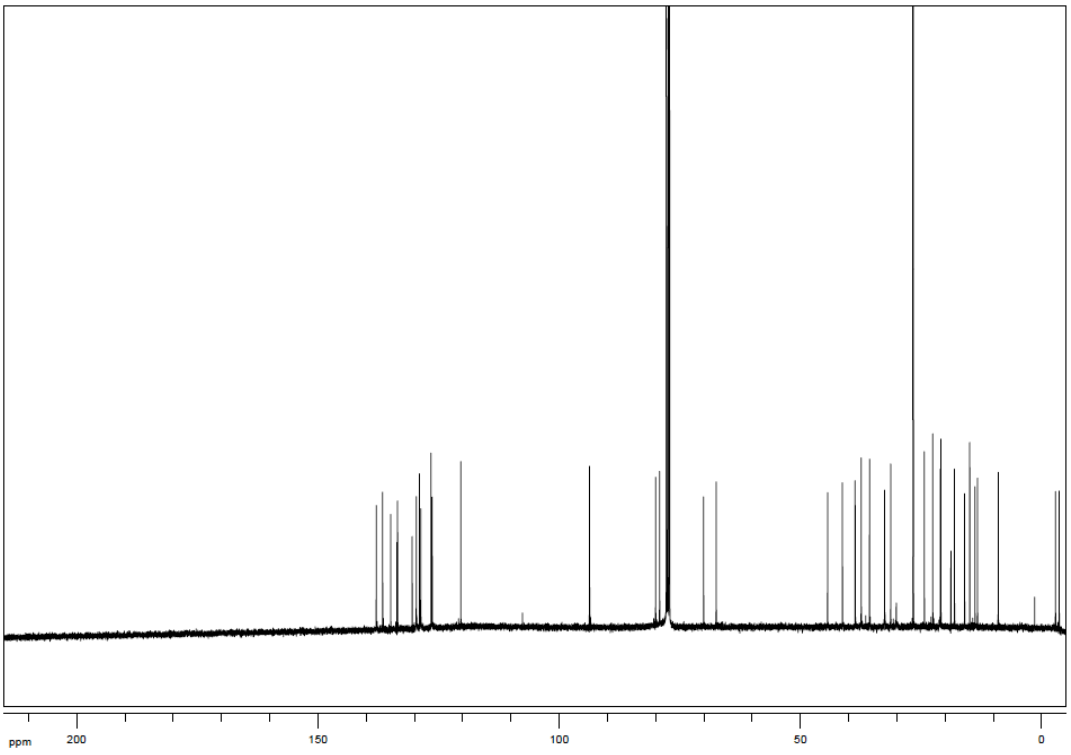
56

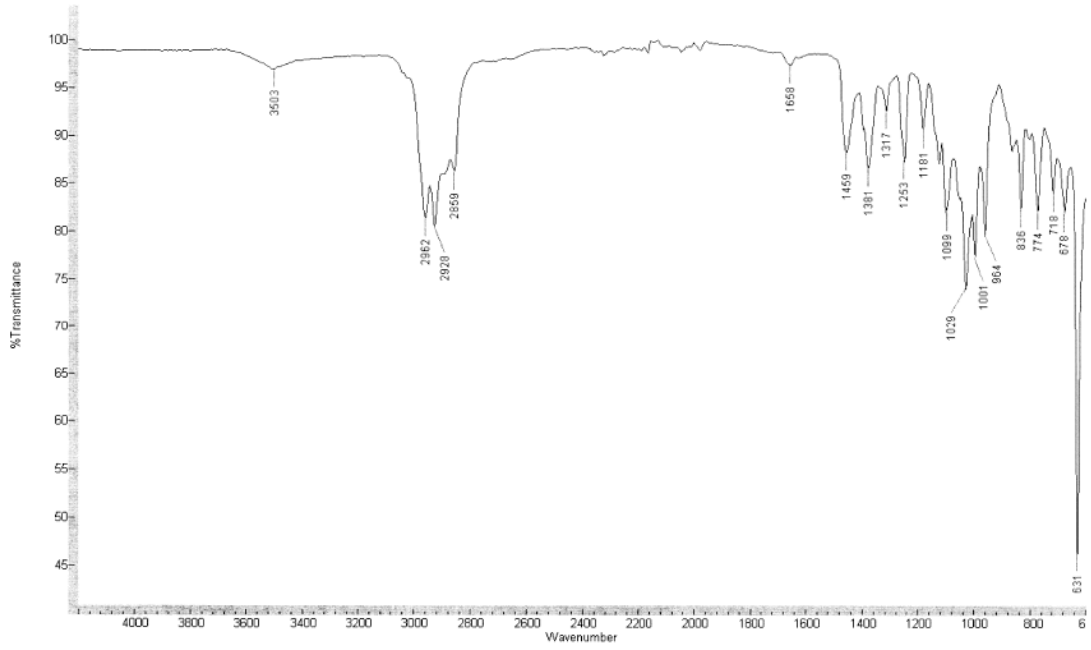
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

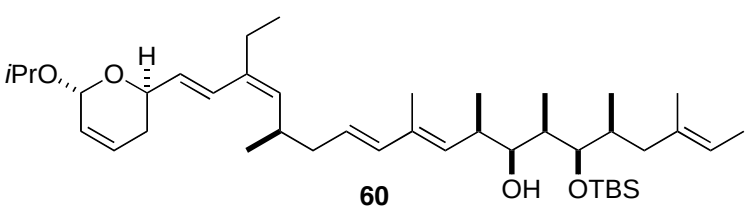

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)


%Transmittance
Wavenumber
2958.1
2928.3
2857.3
2359.2
2326.8
2052.2
1962.6
1456.8
1361.9
1262.6
1105.2
1032.1
946.7
890.1
836.5
772.2
631.2


iPrO
O
H
OH
OTBS
58a
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0

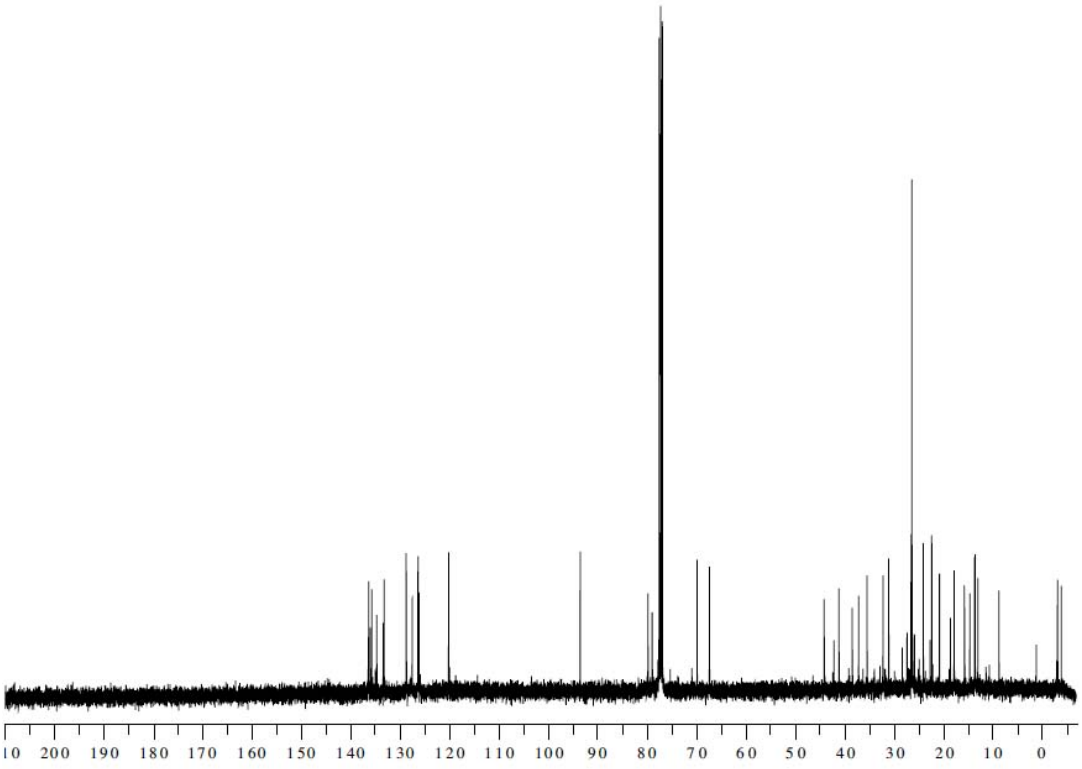

ppm
200
150
100
50
0

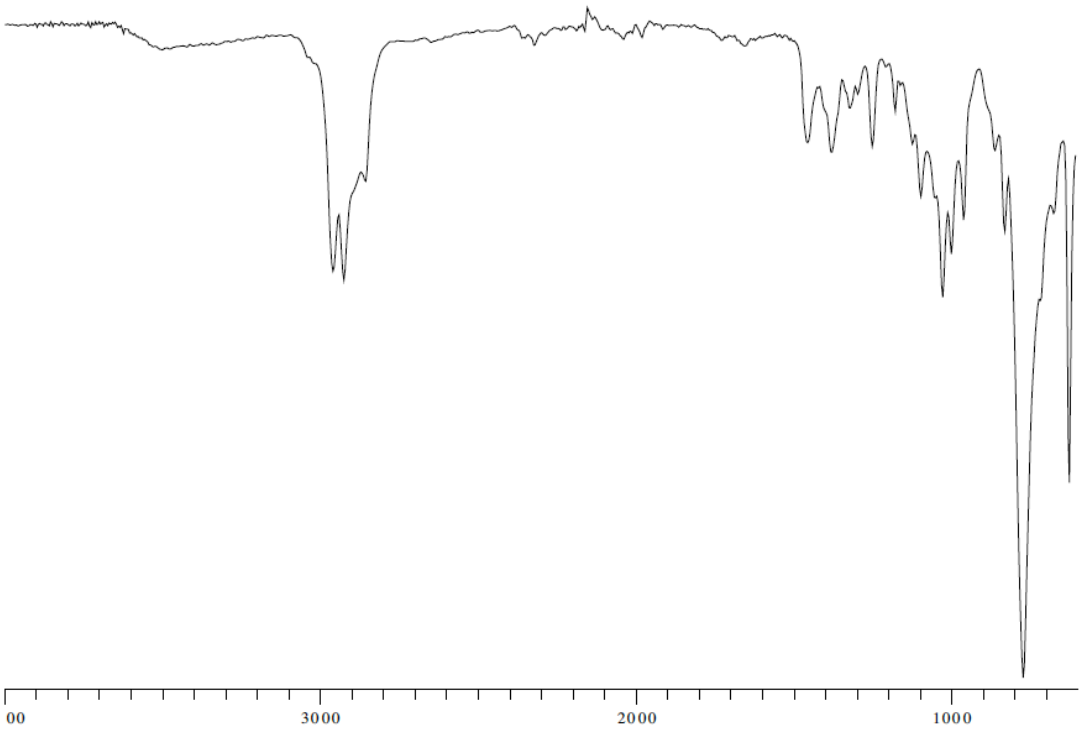

%Transmittance
Wavenumber
3503
2962
2928
2859
1658
1459
1381
1317
1253
1181
1099
1029
1001
964
836
774
718
678
631

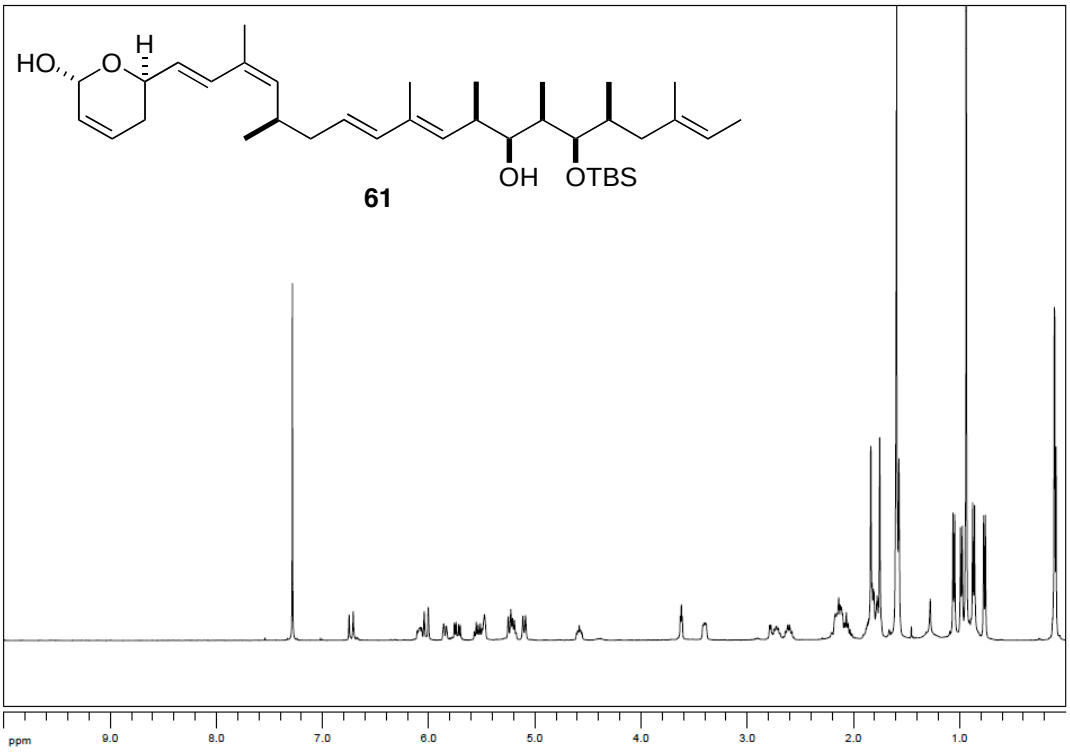

iPrO
O
H
OH
OTBS

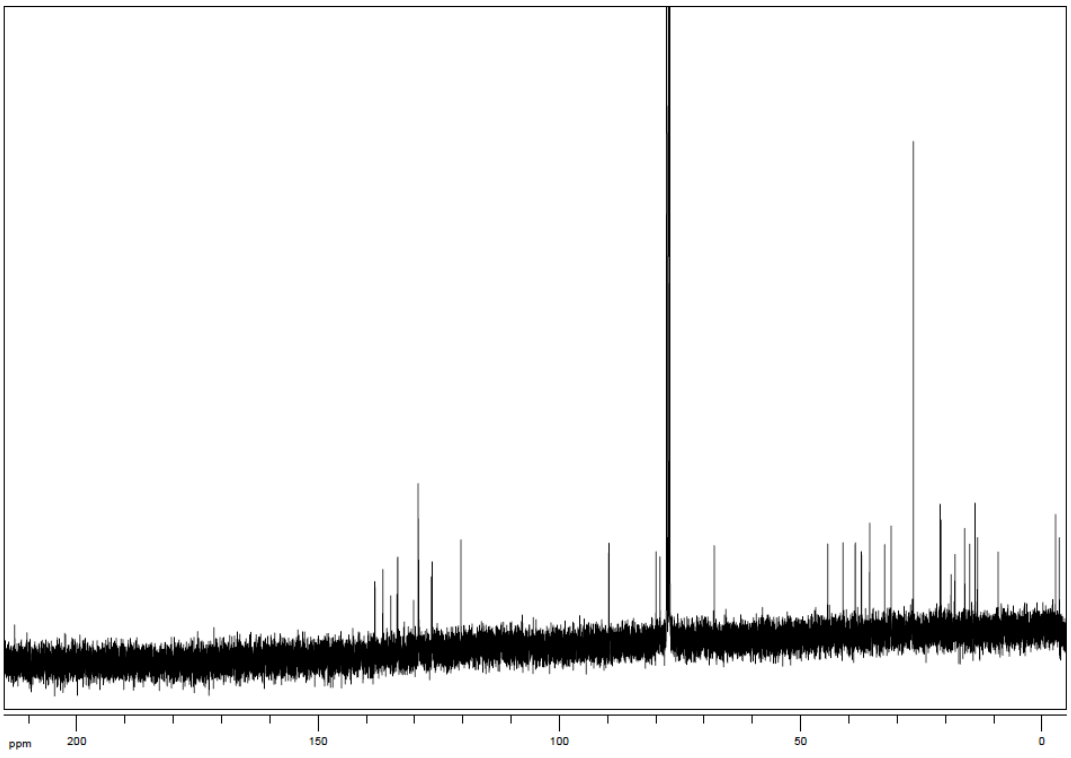
60

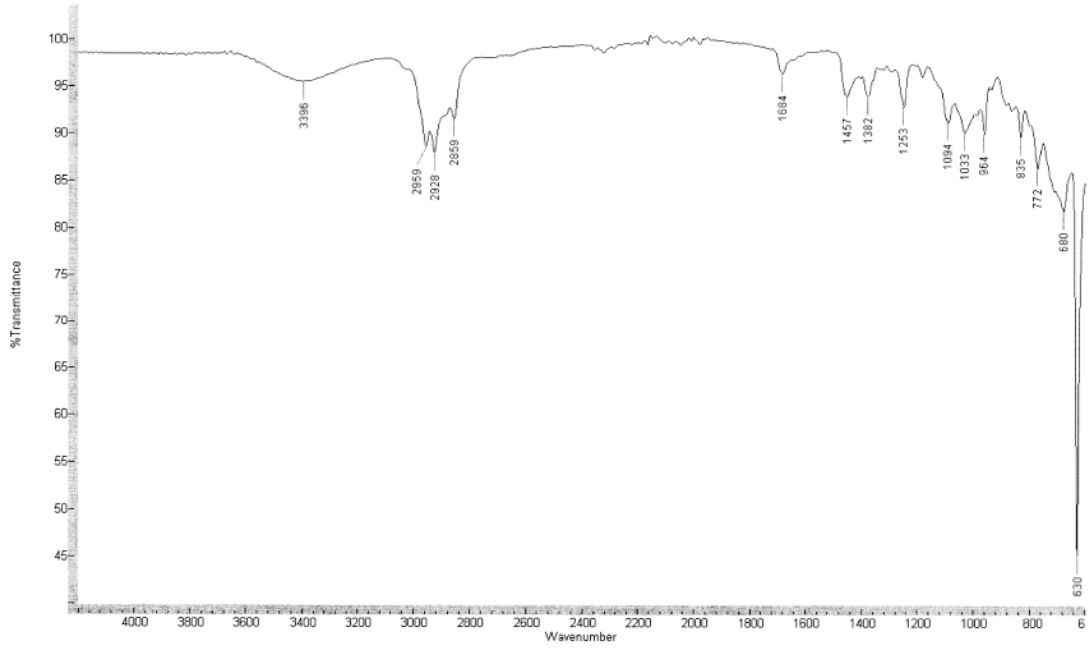
0
9
8
7
6
5
4
3
2
1

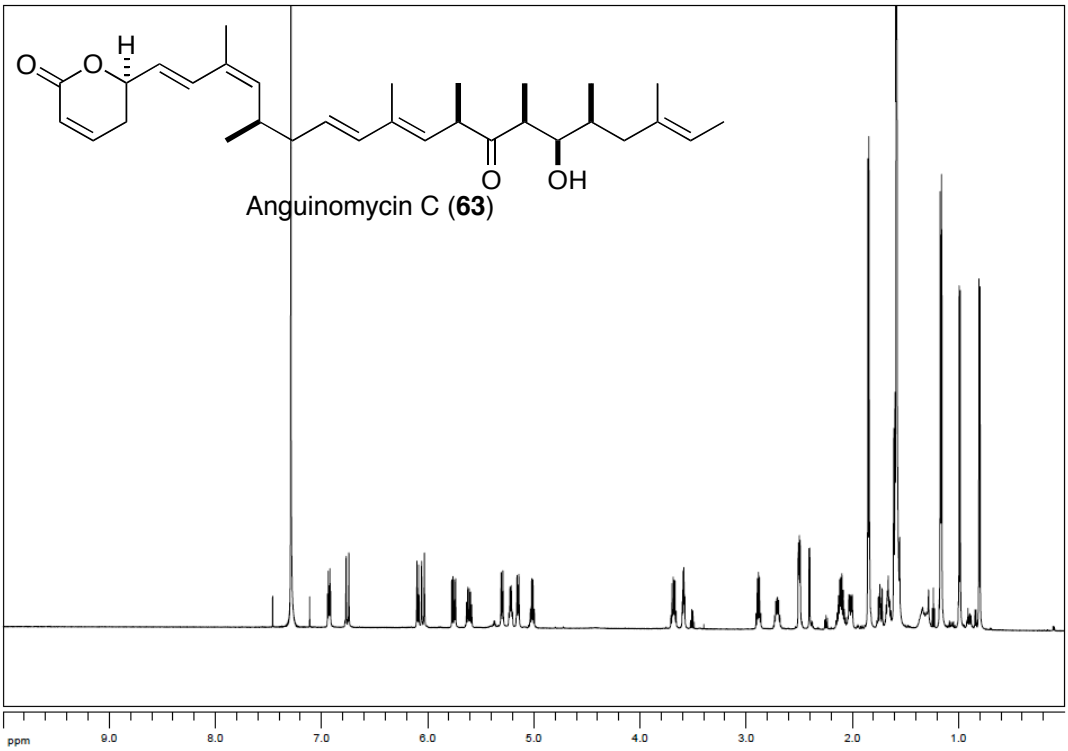

10 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0


00 3000 2000 1000

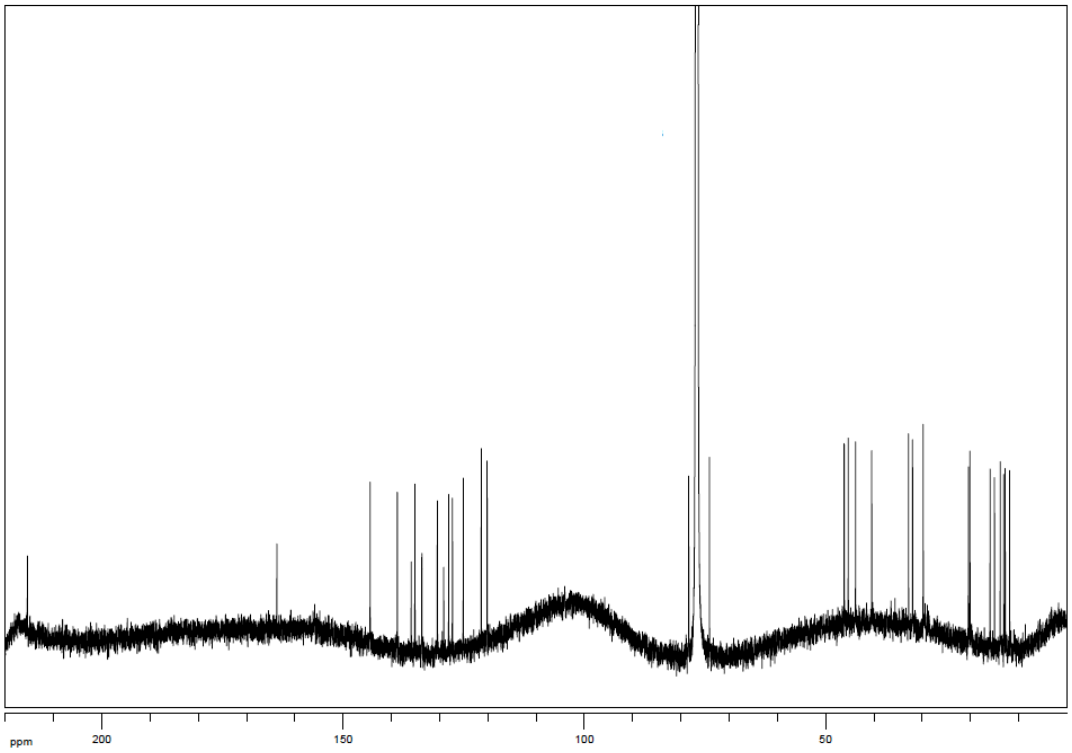

HO
O
H
OH
OTBS
61
ppm
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0


ppm
200
150
100
50
0

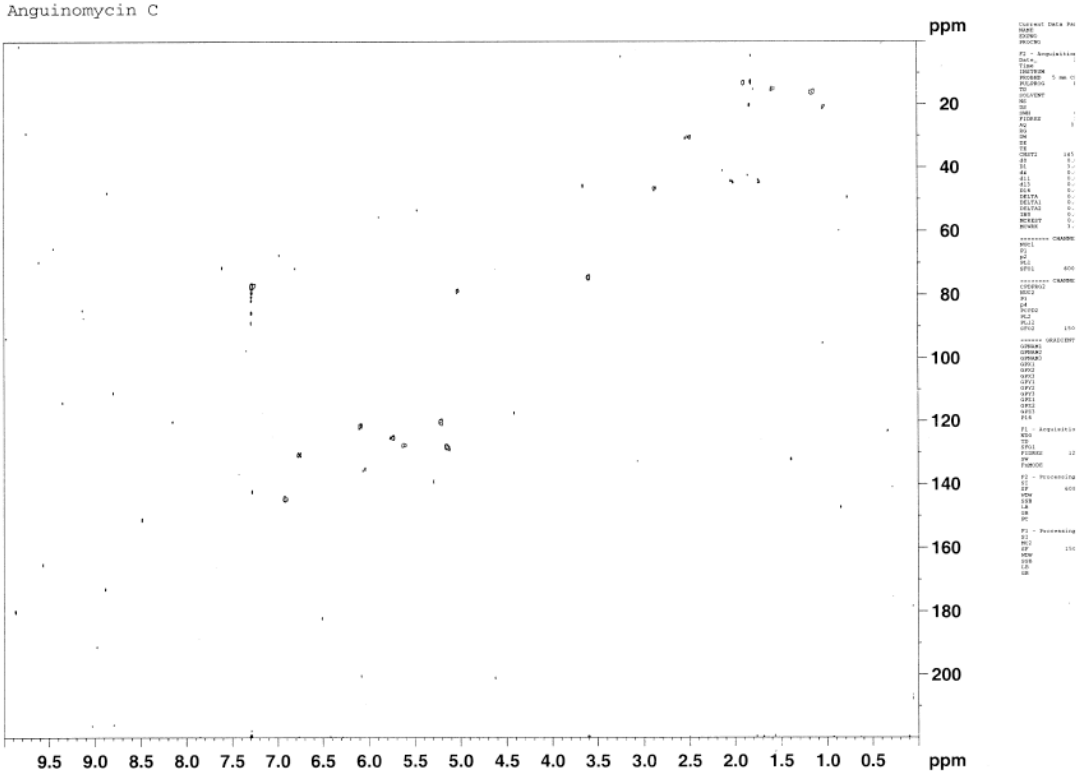


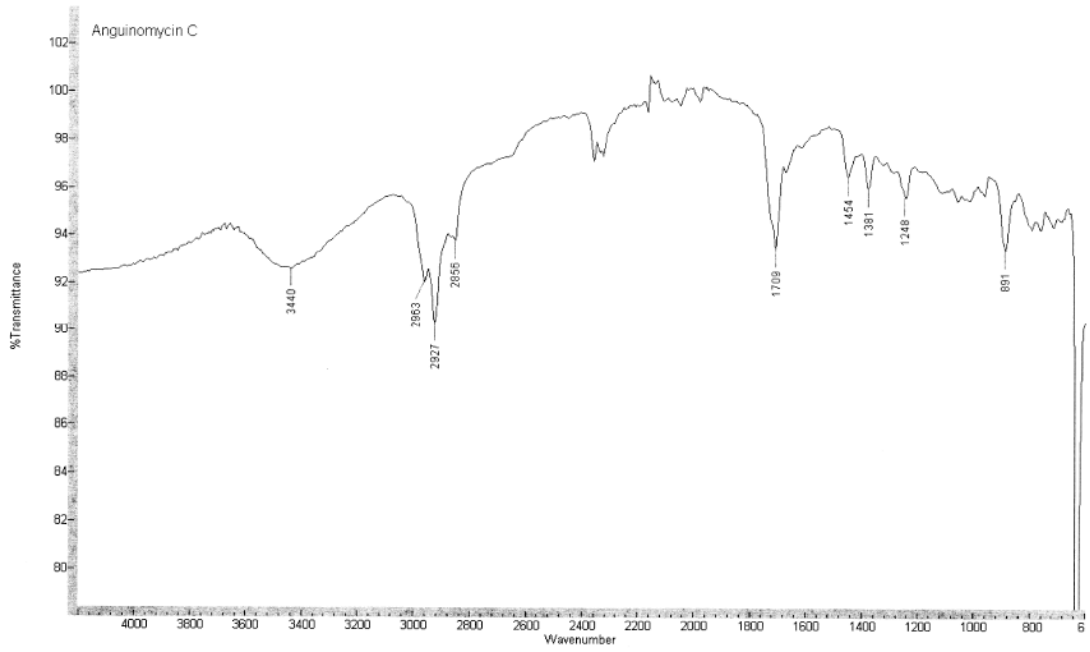


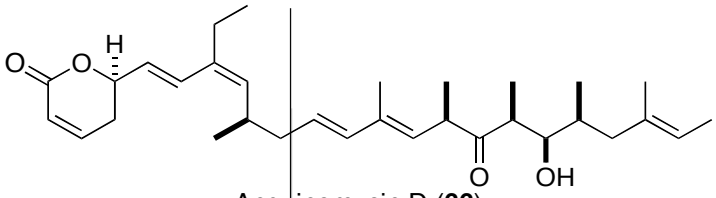
Anguinomycin C (**63**)



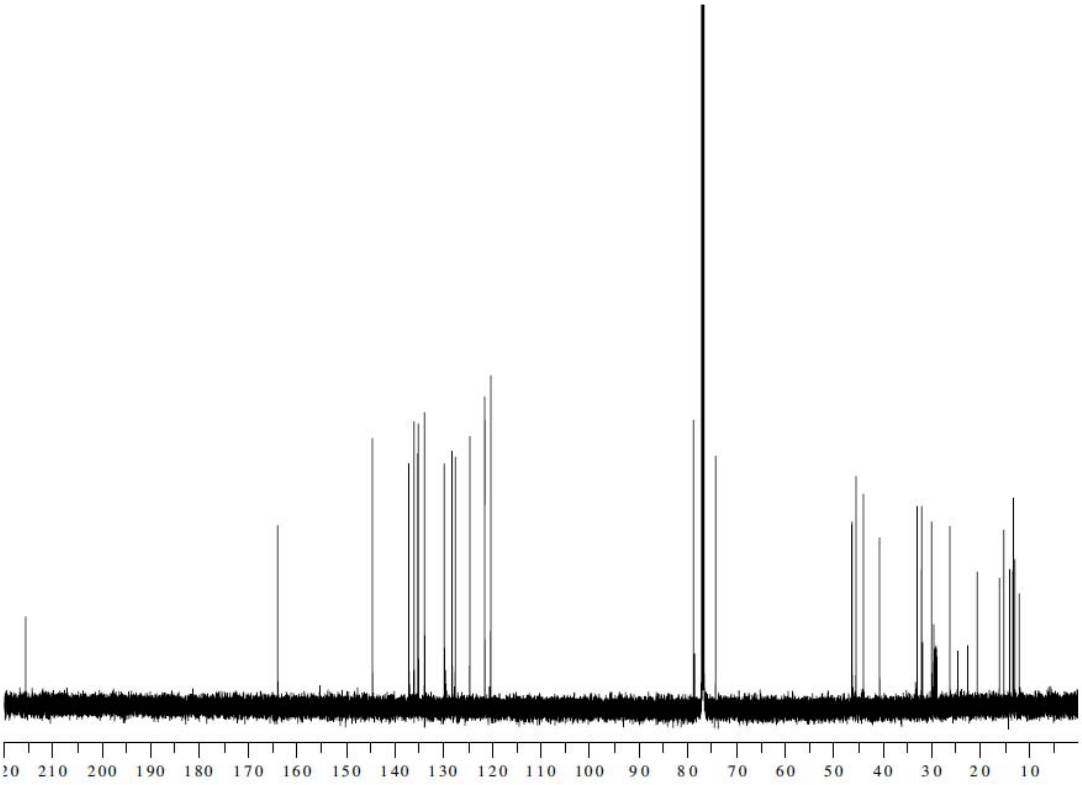
Anguinomycin C

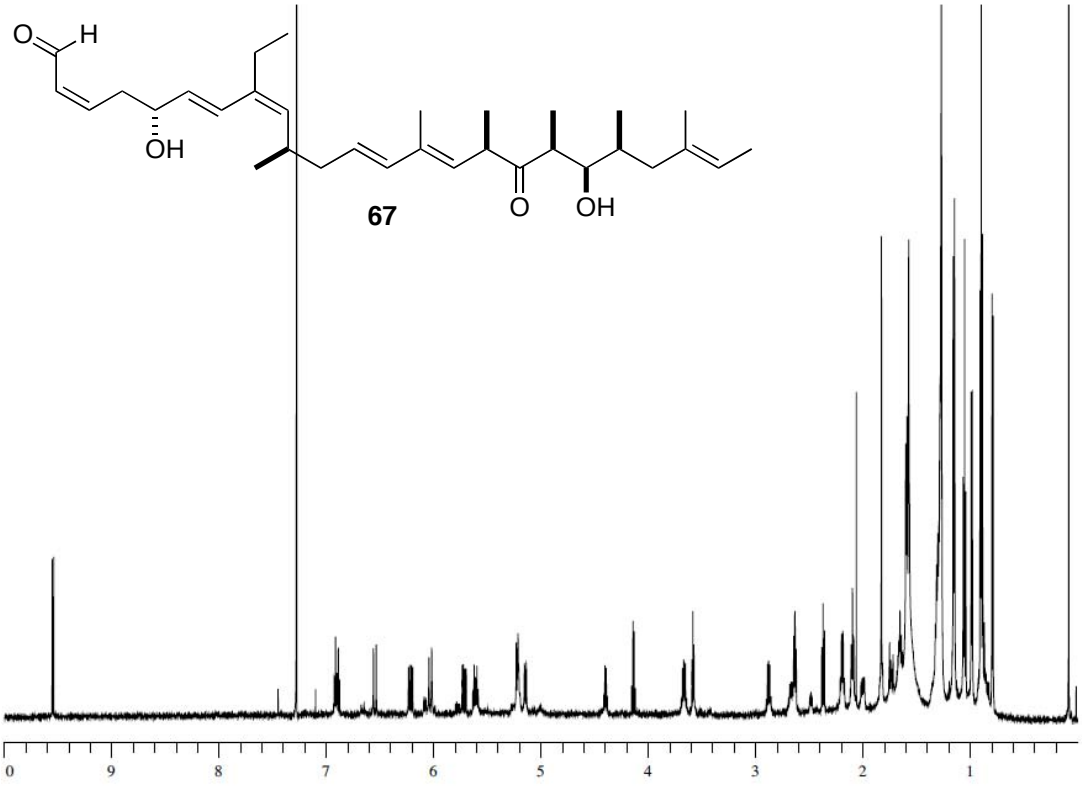


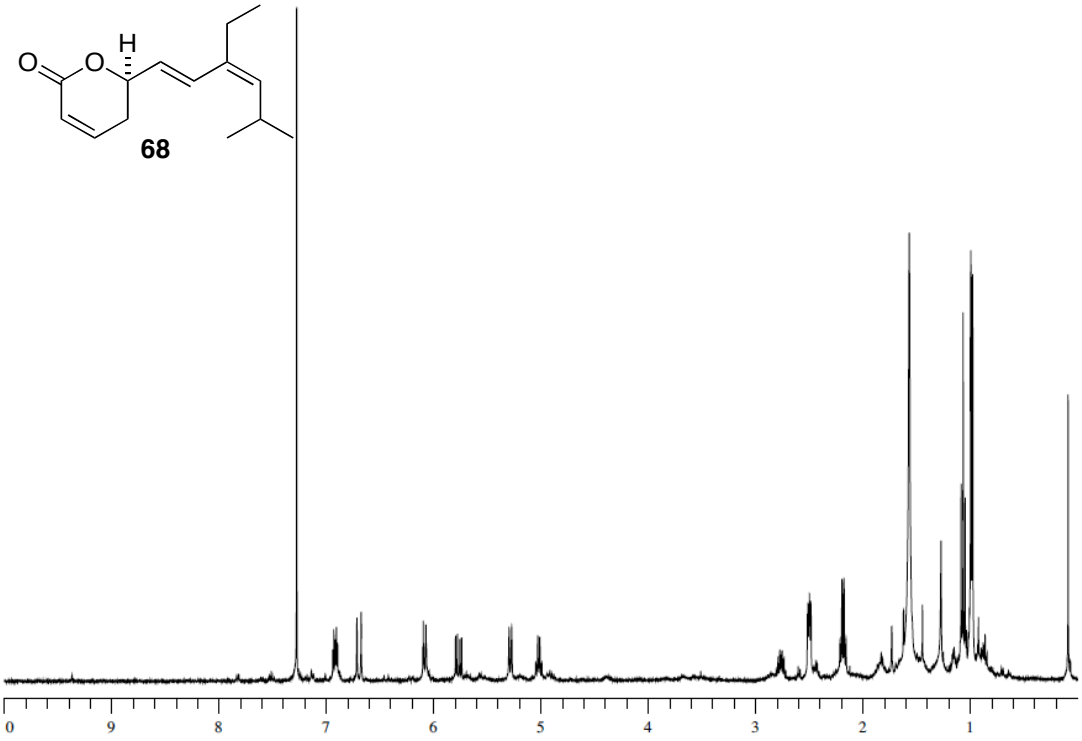


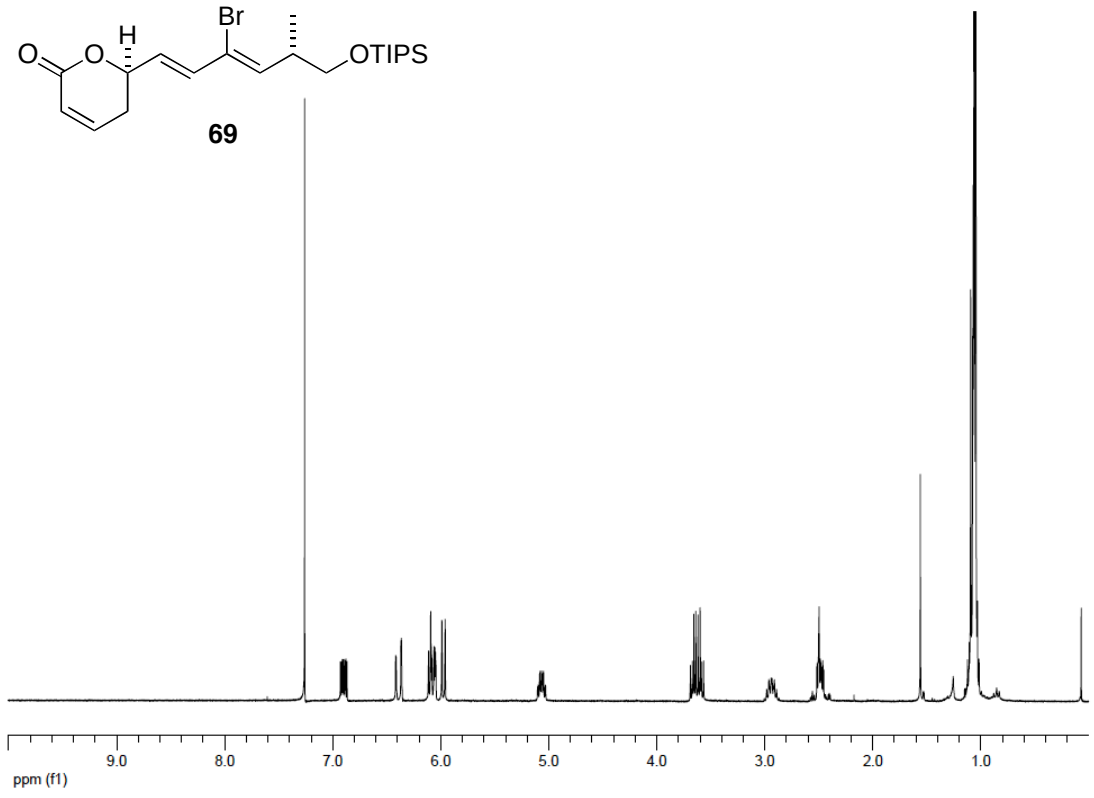


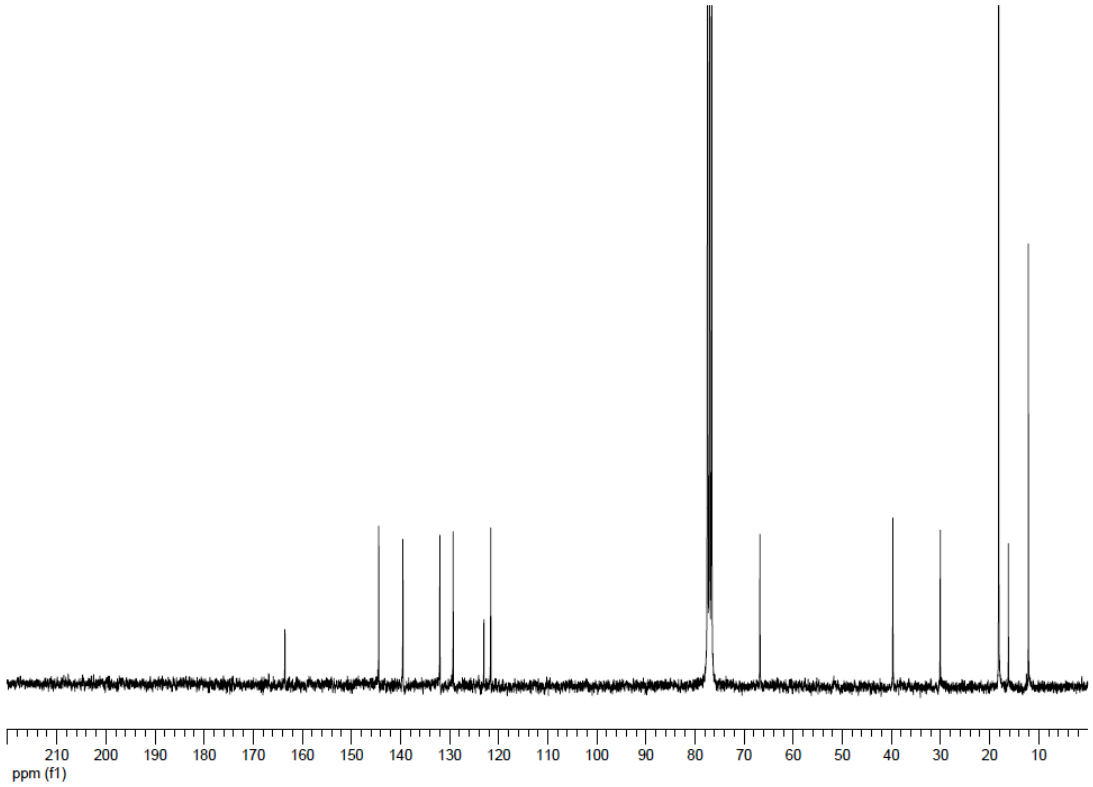
Anguinomycin D (**66**)

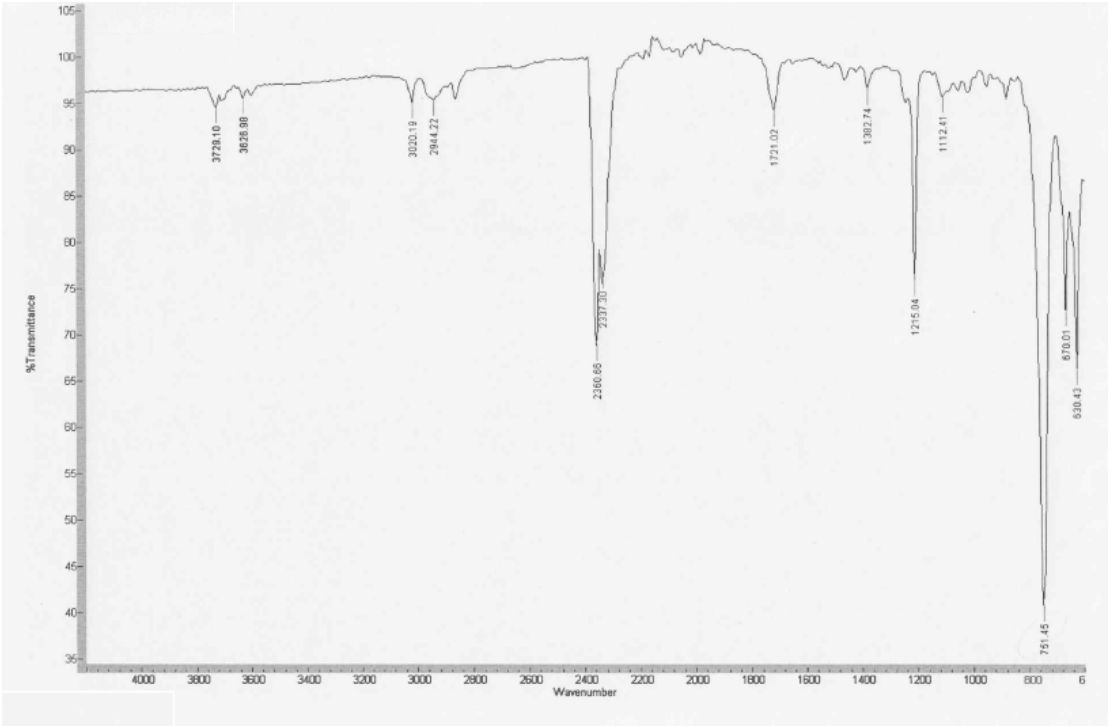

220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10

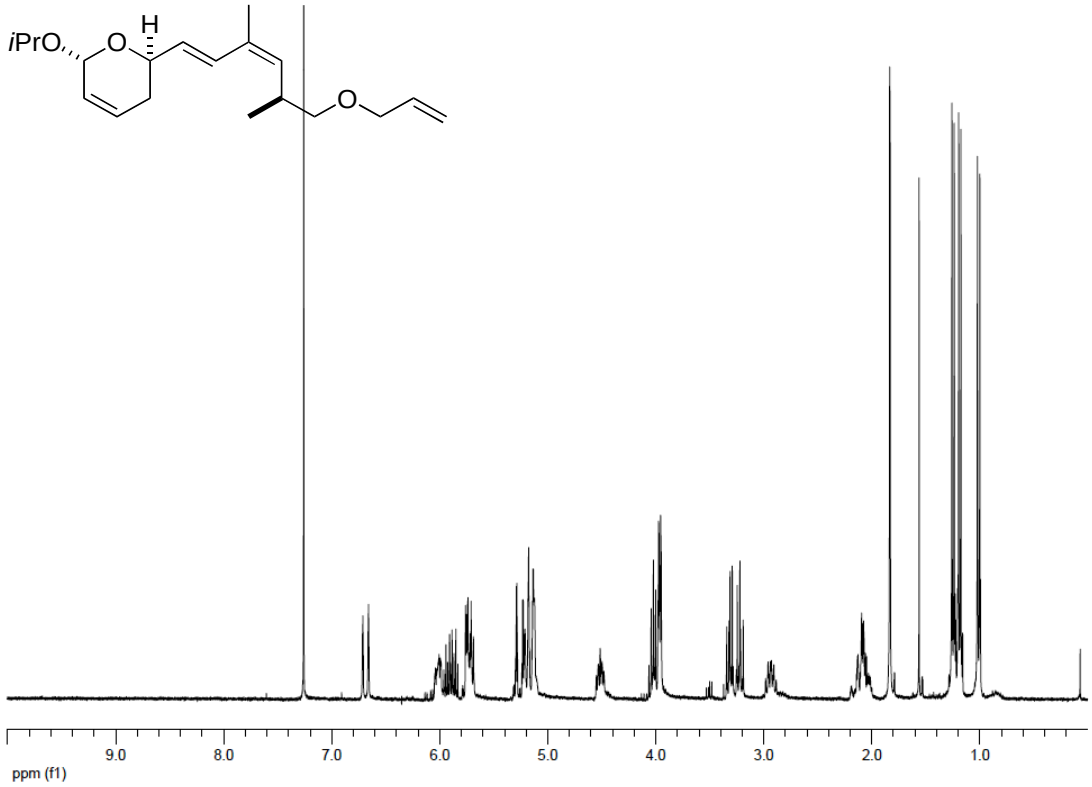

O
H
OH
O
OH
67
0 9 8 7 6 5 4 3 2 1

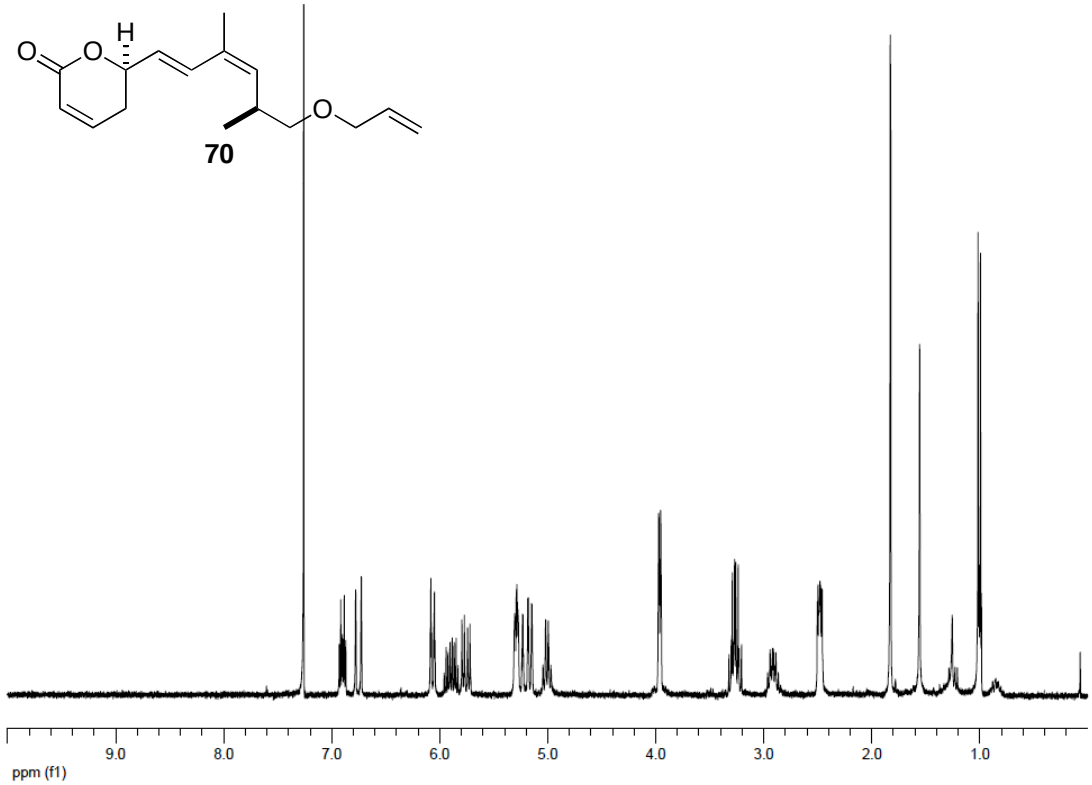

O
O
H
68
0
9
8
7
6
5
4
3
2
1


Br
O
O
H
OTIPS
69
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

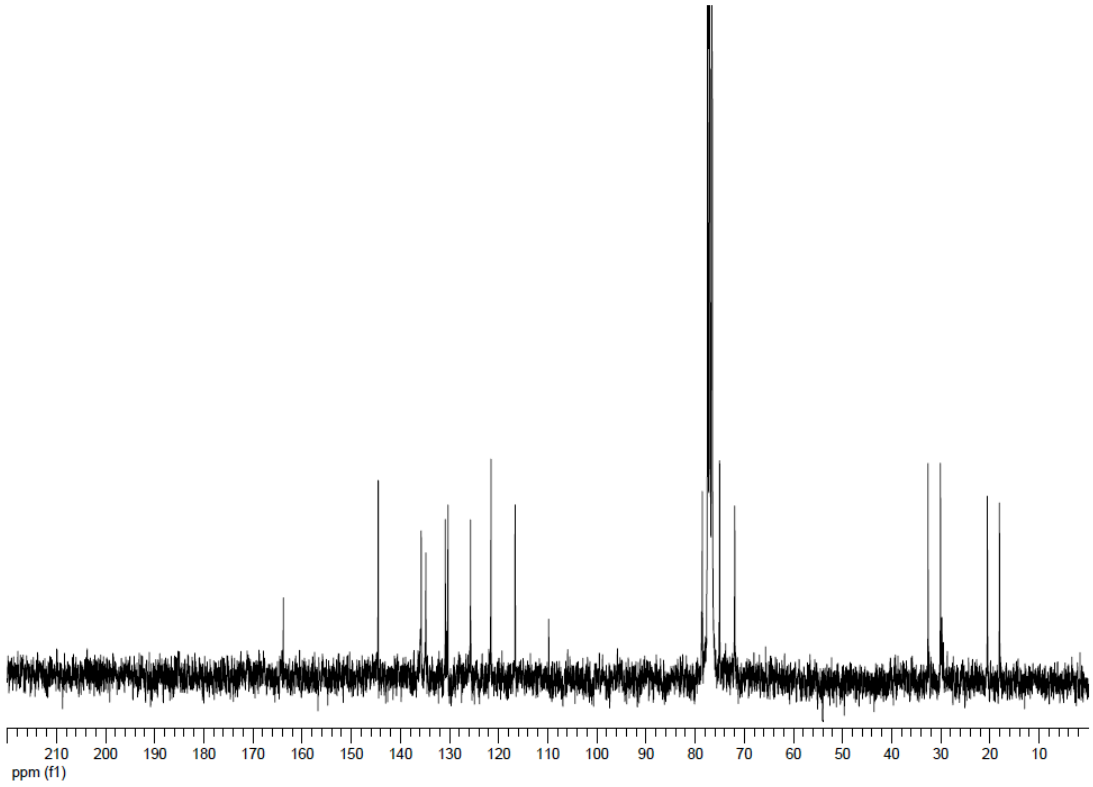

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
ppm (f1)


%Transmittance
Wavenumber
3729.10
3626.98
3020.19
2944.22
2360.66
2337.30
1721.02
1380.74
1215.04
1112.41
751.45
670.01
630.43

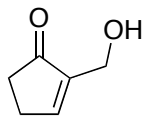

iPrO
O
H
O
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)


O
O
H
O
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

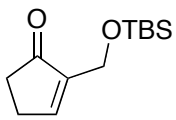
70


210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm (f1)

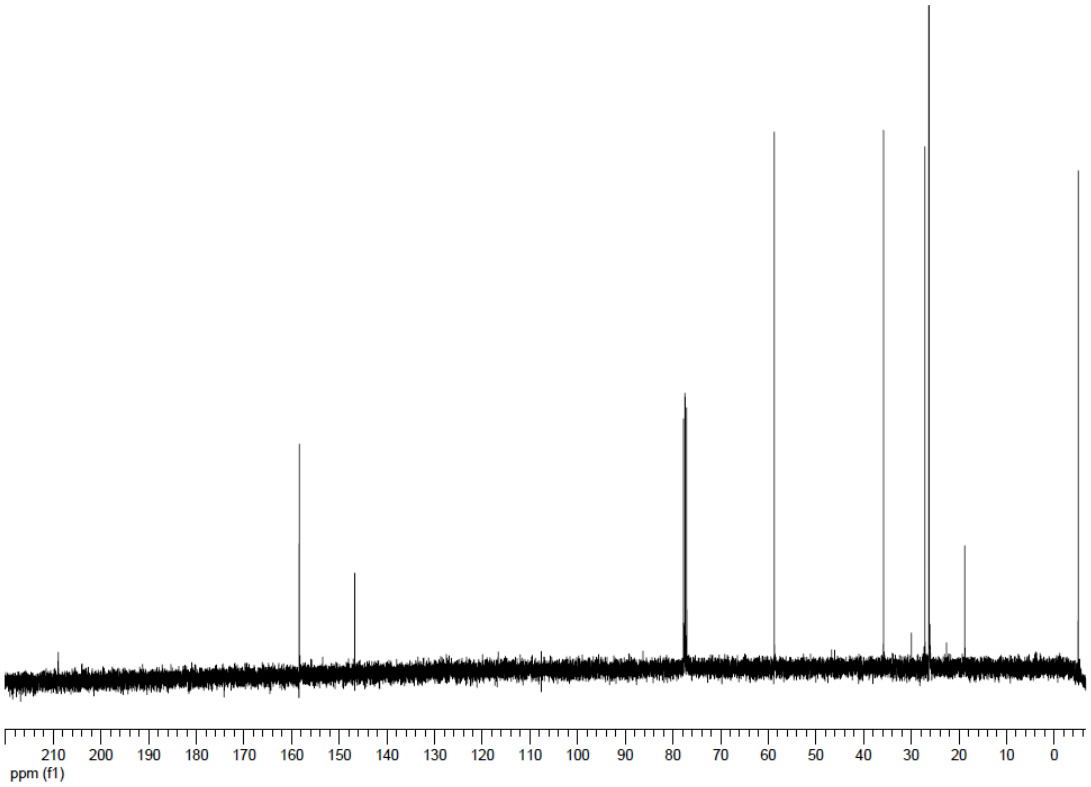
6.5.2. Spectra from the Sporolides Project

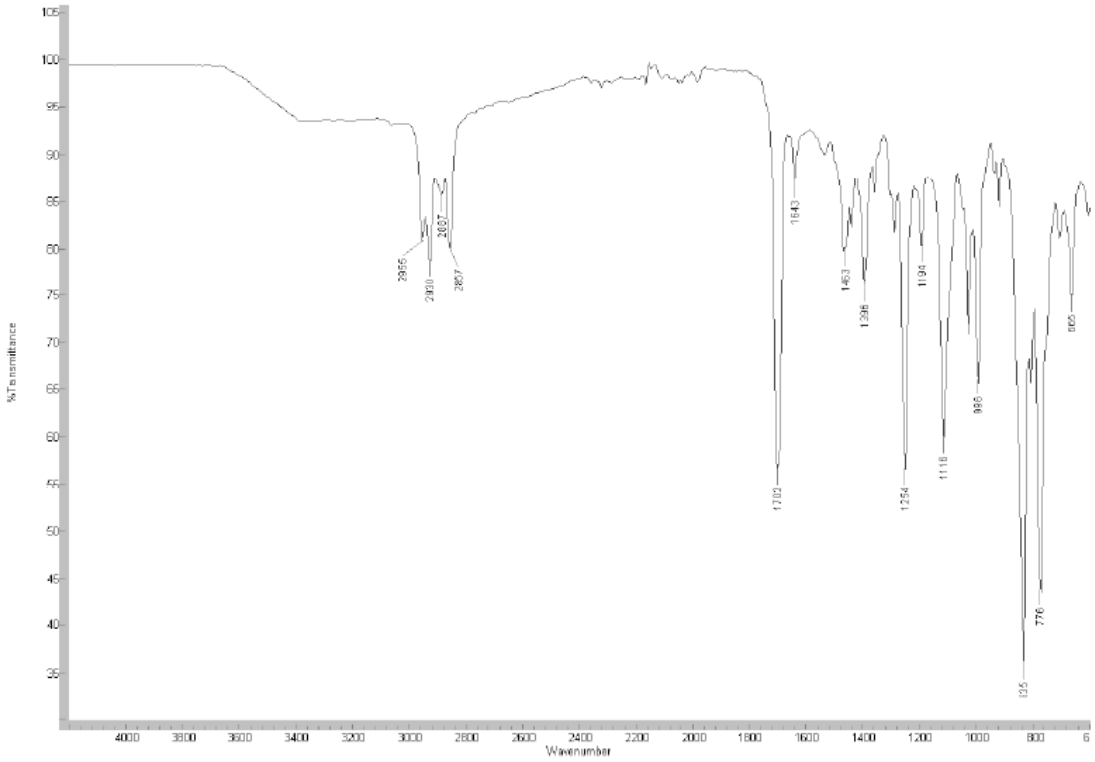


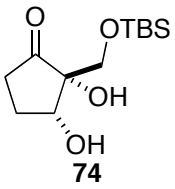
72



73

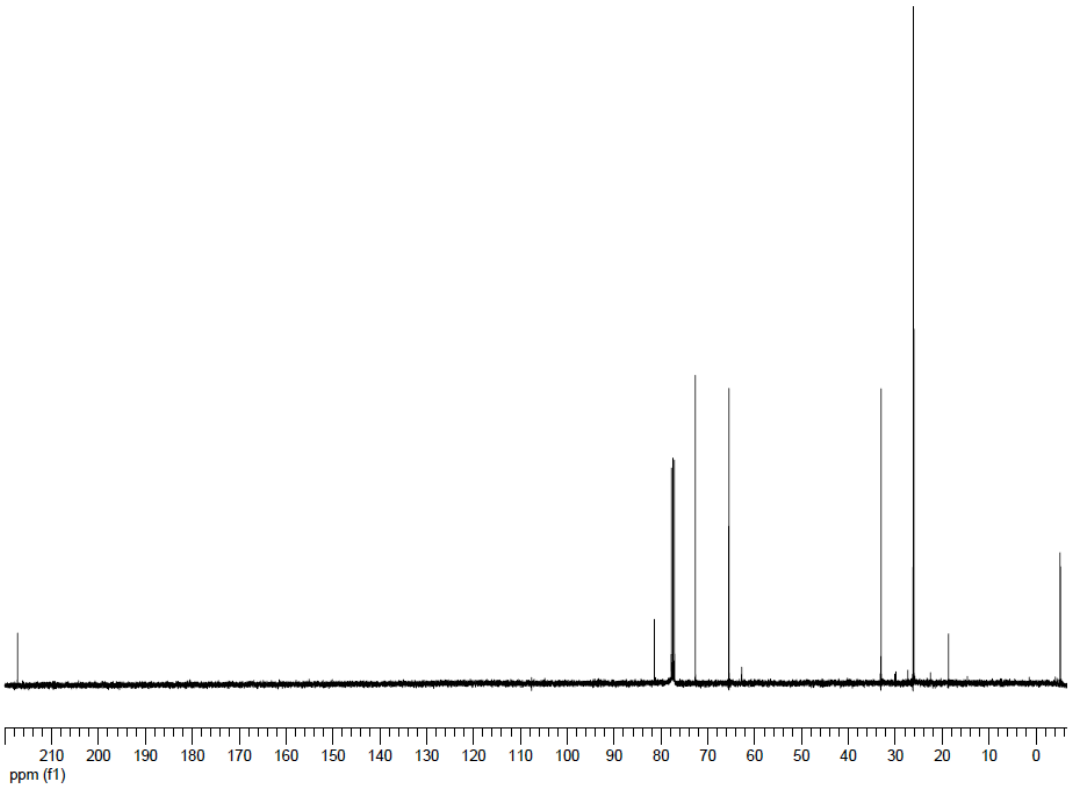

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

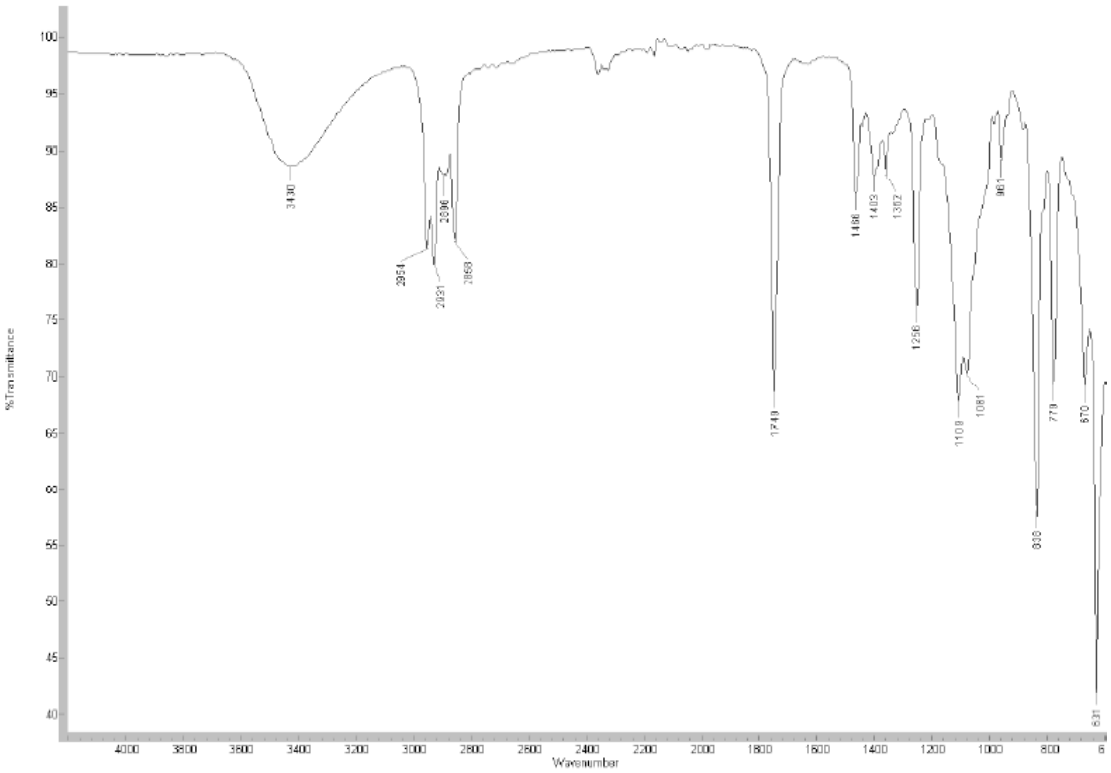

%Transmittance
2955
2930
2857
1702
1643
1463
1396
1254
1194
1118
996
865
776
635
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800
Wavenumber

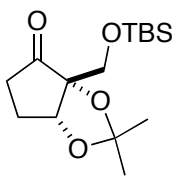

O
OTBS
OH
OH

74

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

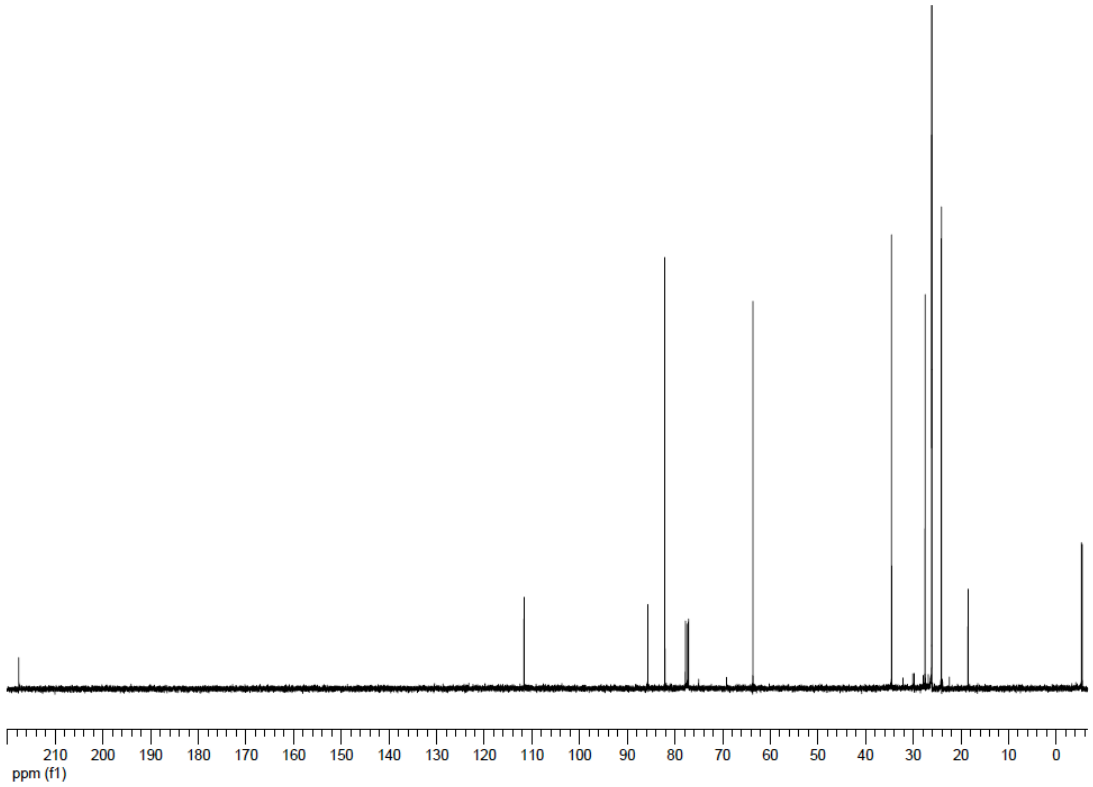

210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)

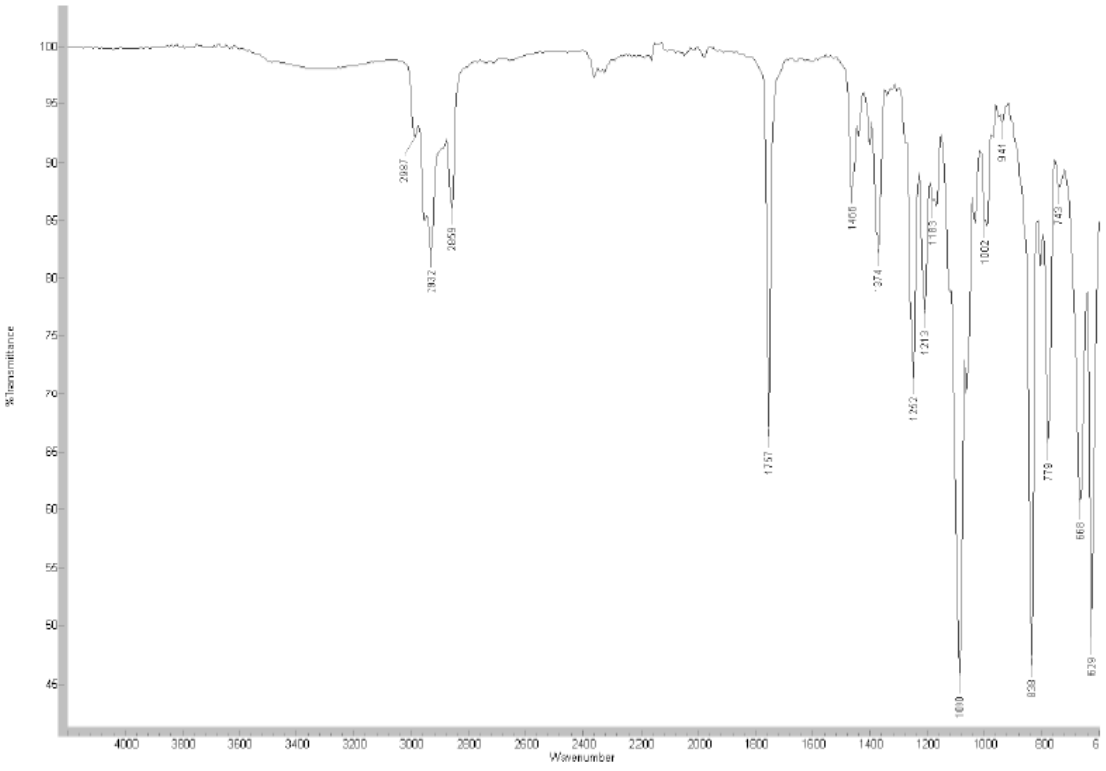

%Transmittance
Wavenumber
3430
2954
2931
2896
2859
1749
1466
1403
1382
1256
1110
1081
961
838
779
670
631

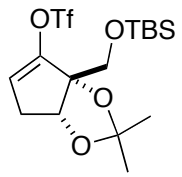

O
OTBS
O
O

76

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

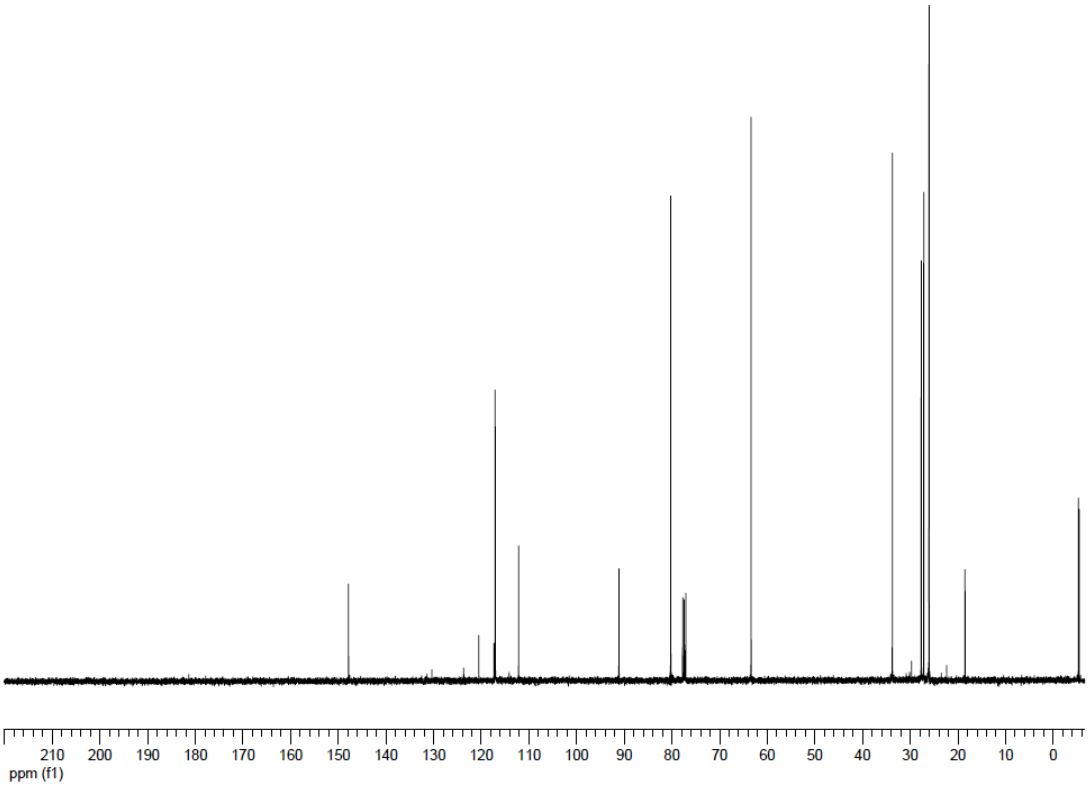

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

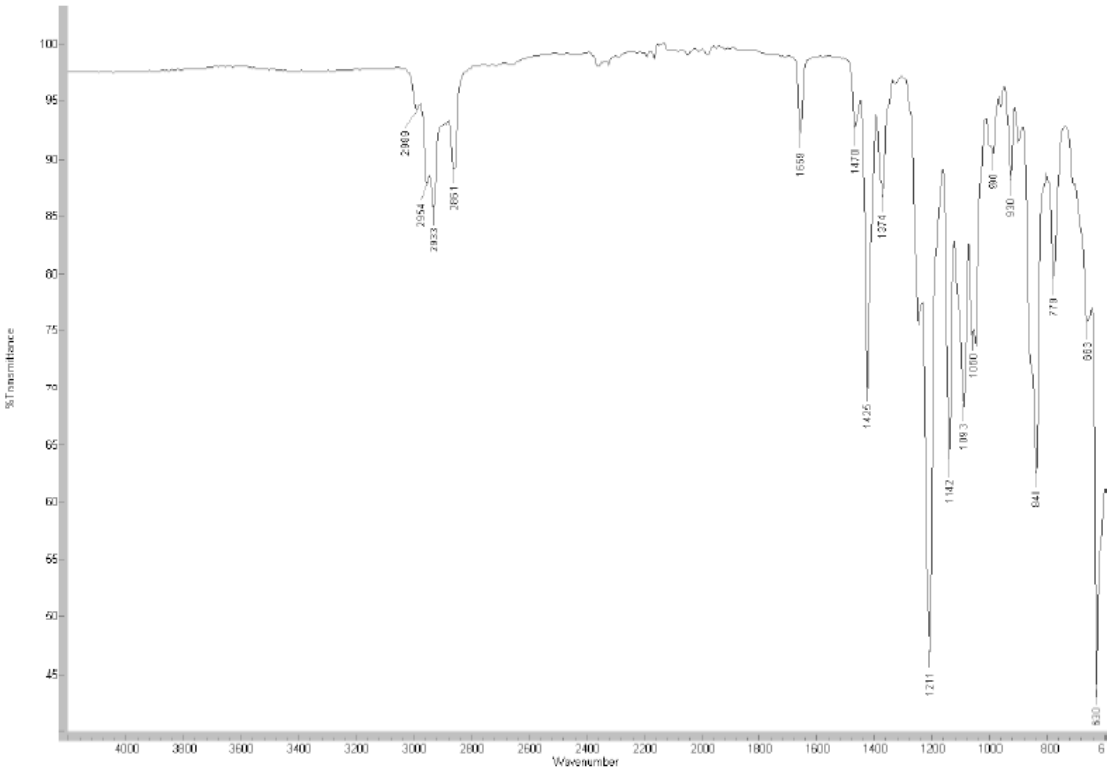

%Transmittance
100 95 90 85 80 75 70 65 60 55 50 45
2987
2932
2859
1757
1466
1374
1252
1213
1183
1002
941
779
743
668
639
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800 6
Wavenumber

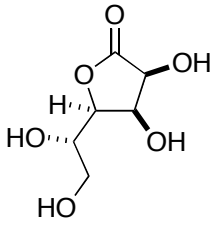

OTf
OTBS
O
O

77

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

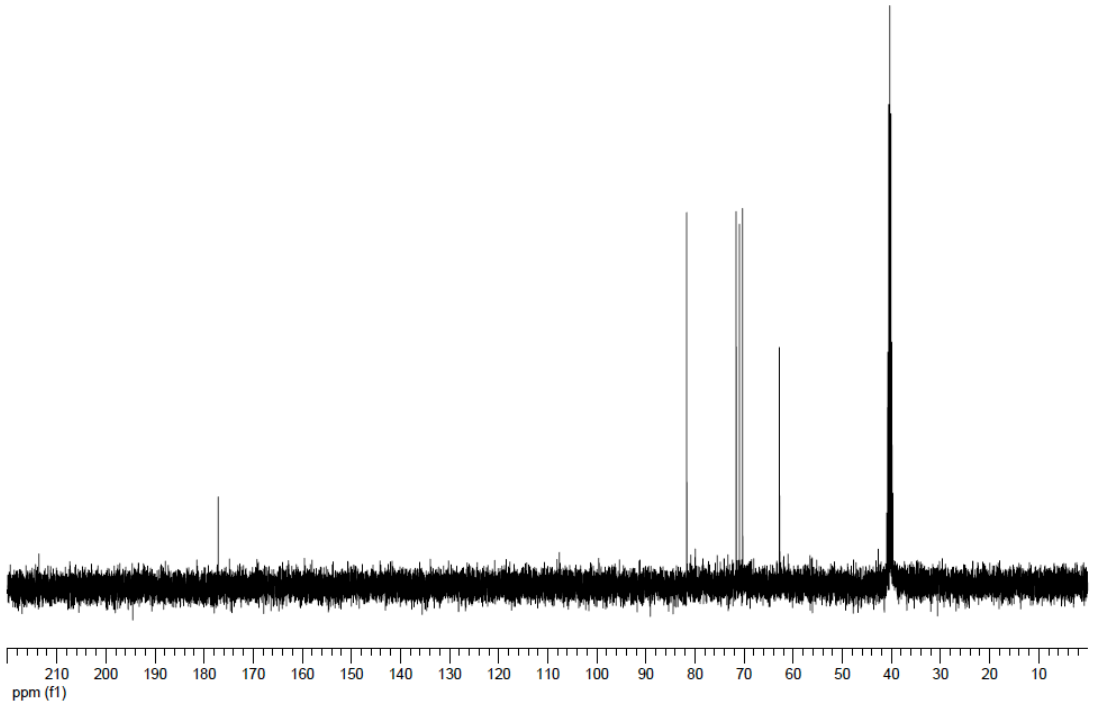

210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)

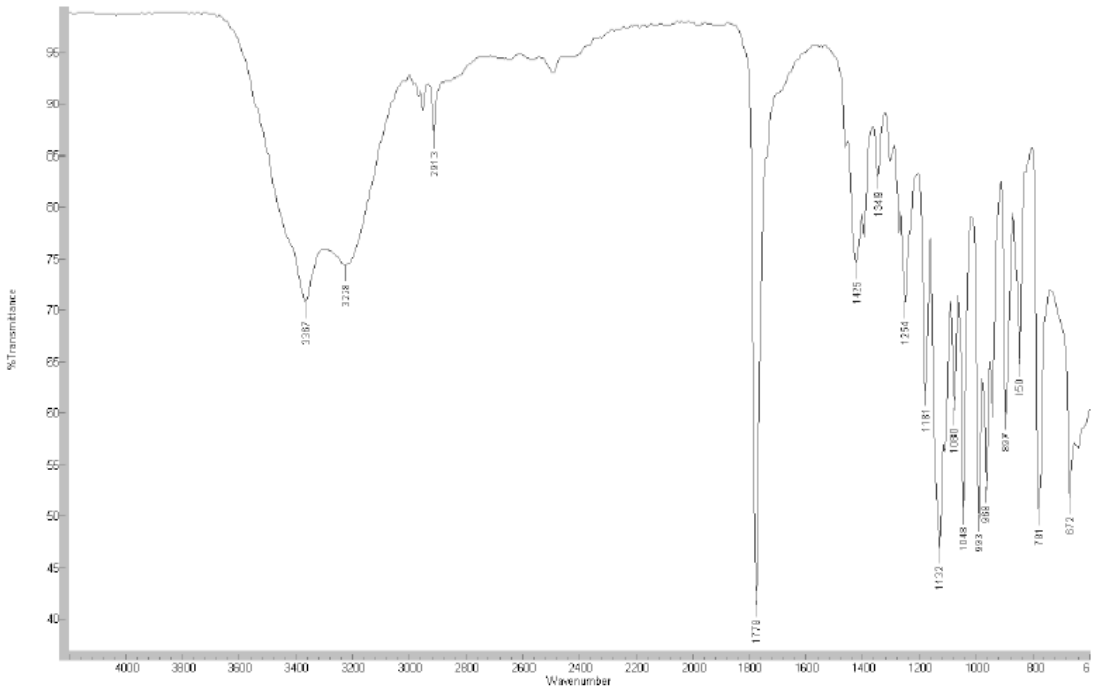

%Transmittance
Wavenumber
2969
2954
2933
2851
1698
1478
1374
1425
1211
1142
1083
1080
996
930
774
644
563
530

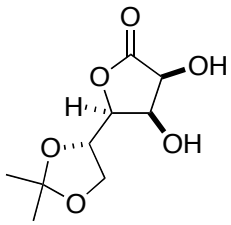

O
O
OH
H
HO
OH
HO

83

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

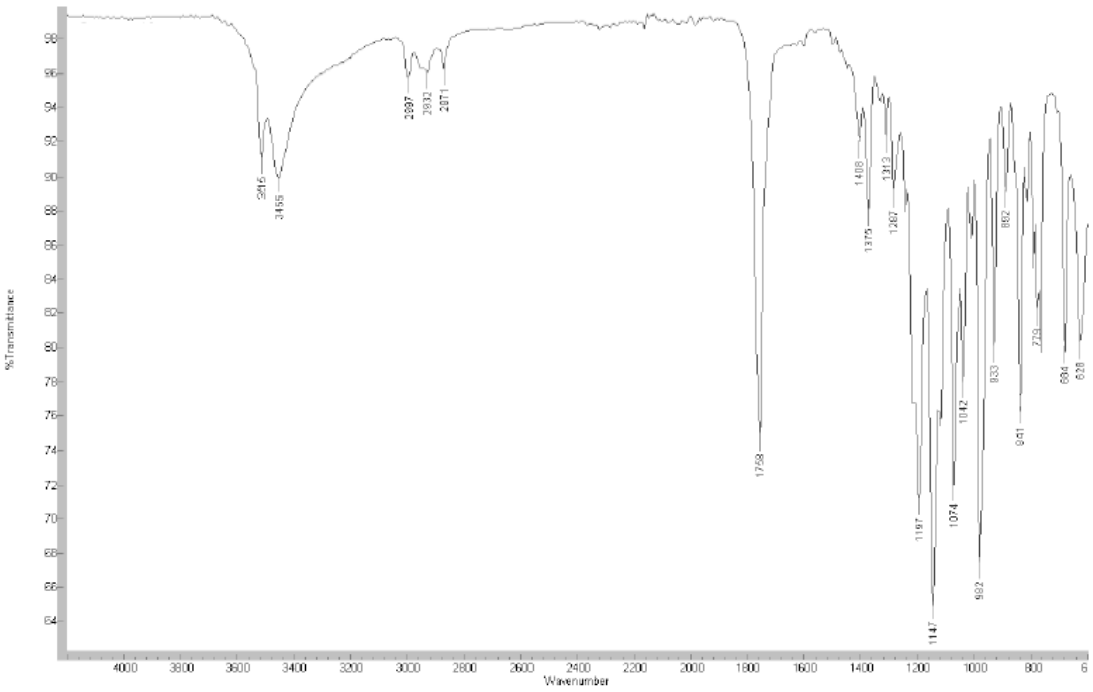

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
ppm (f1)

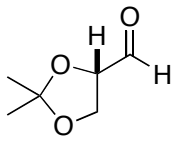

%Transmittance
3387
3228
2913
1778
1425
1348
1254
1181
1132
1080
1048
988
893
781
672
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800
Wavenumber


O
O
OH
H
O
OH
O

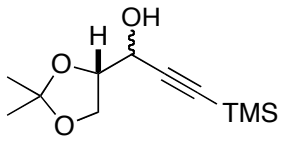
84

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (t1)

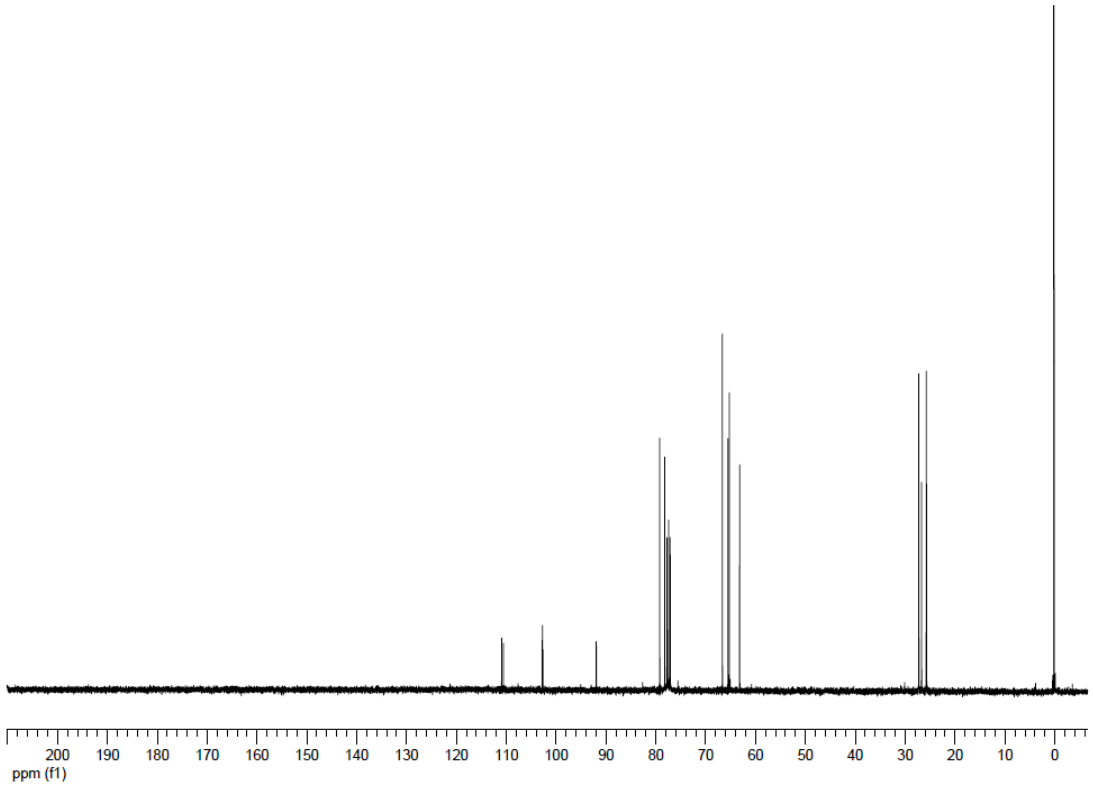

%Transmittance
Wavenumber
3455
2997
2932
2871
1759
1408
1375
1312
1267
1197
1147
1074
1042
982
941
892
833
729
684
638

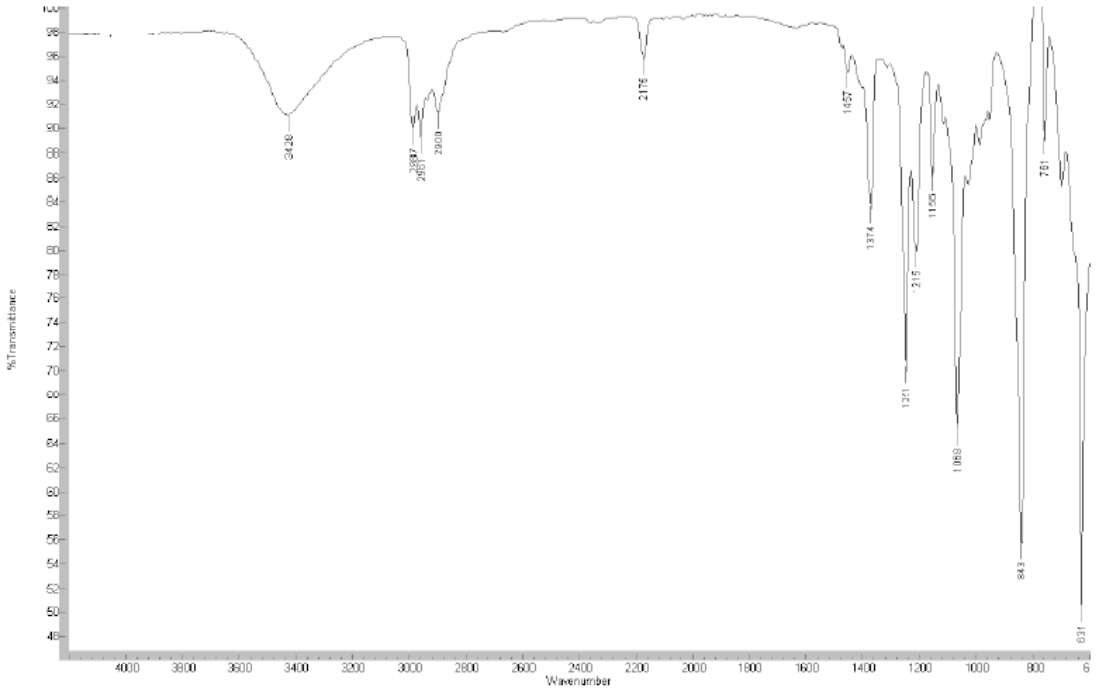


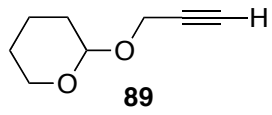
85



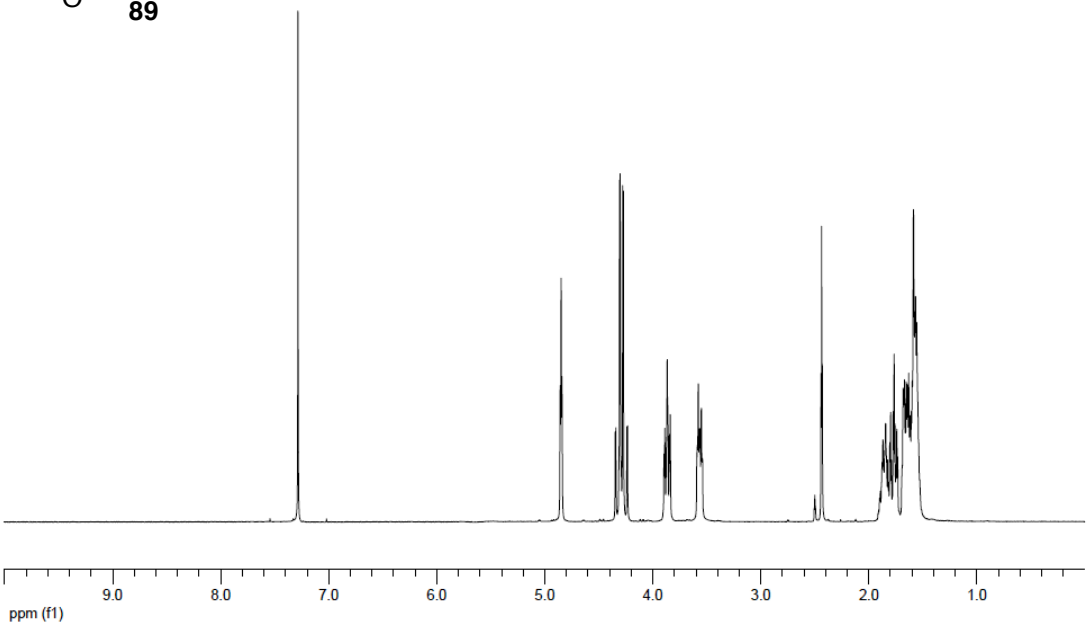
86

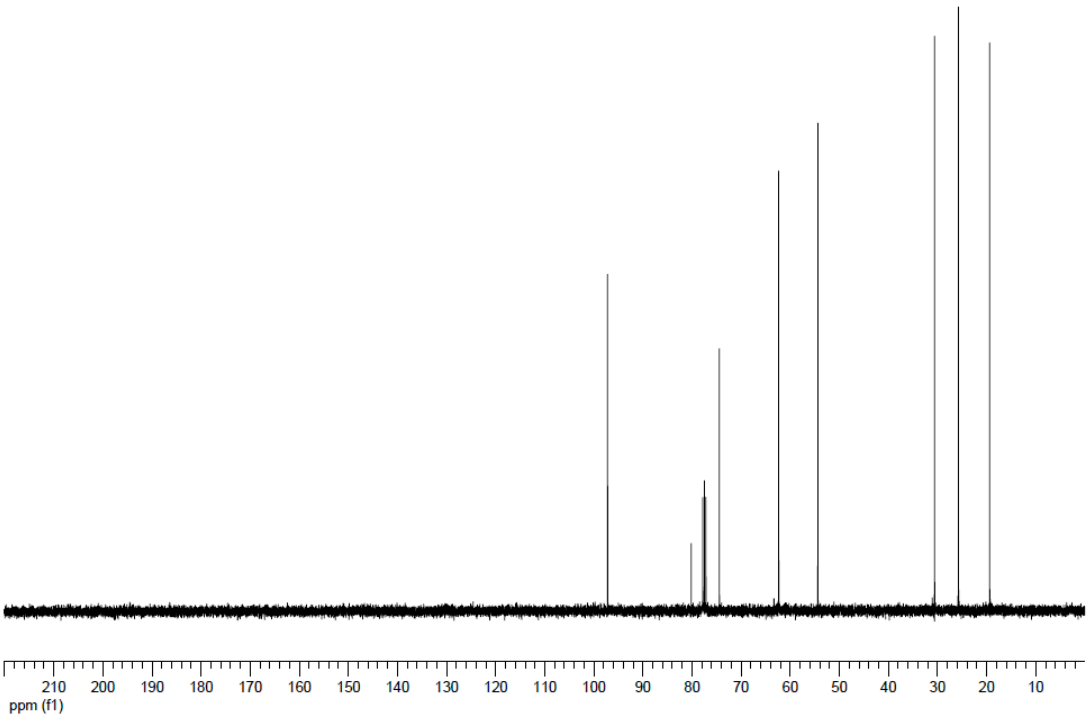

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

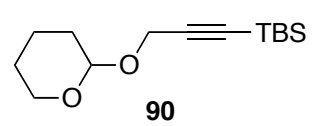

%Transmittance
3429
2887
2951
2909
2176
1457
1374
1321
1215
1155
1060
843
781
631
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800
Wavenumber


H
O
O

89

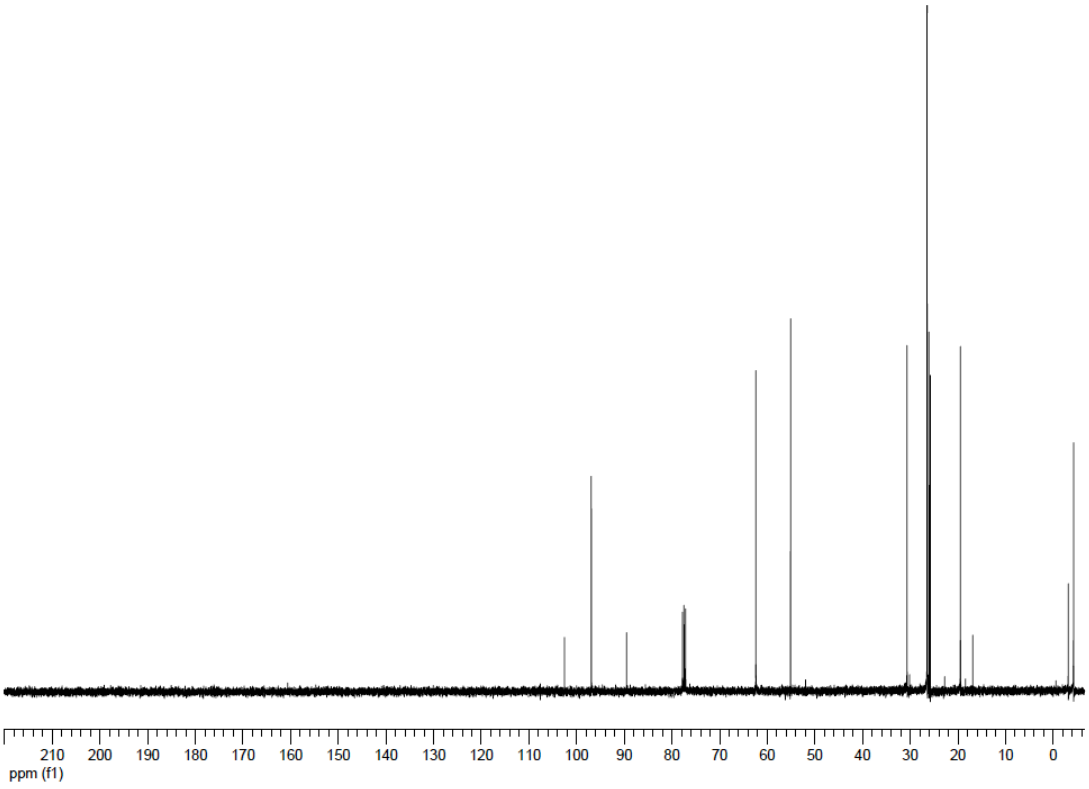

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

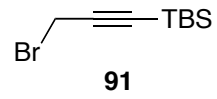

210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm (f1)


TBS
O
O

90

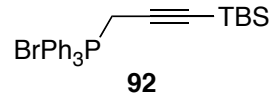
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)


210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)


TBS
Br

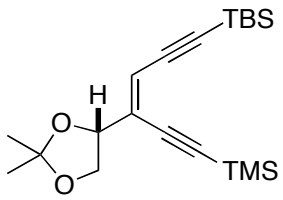
91

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)


TBS
BrPh3P

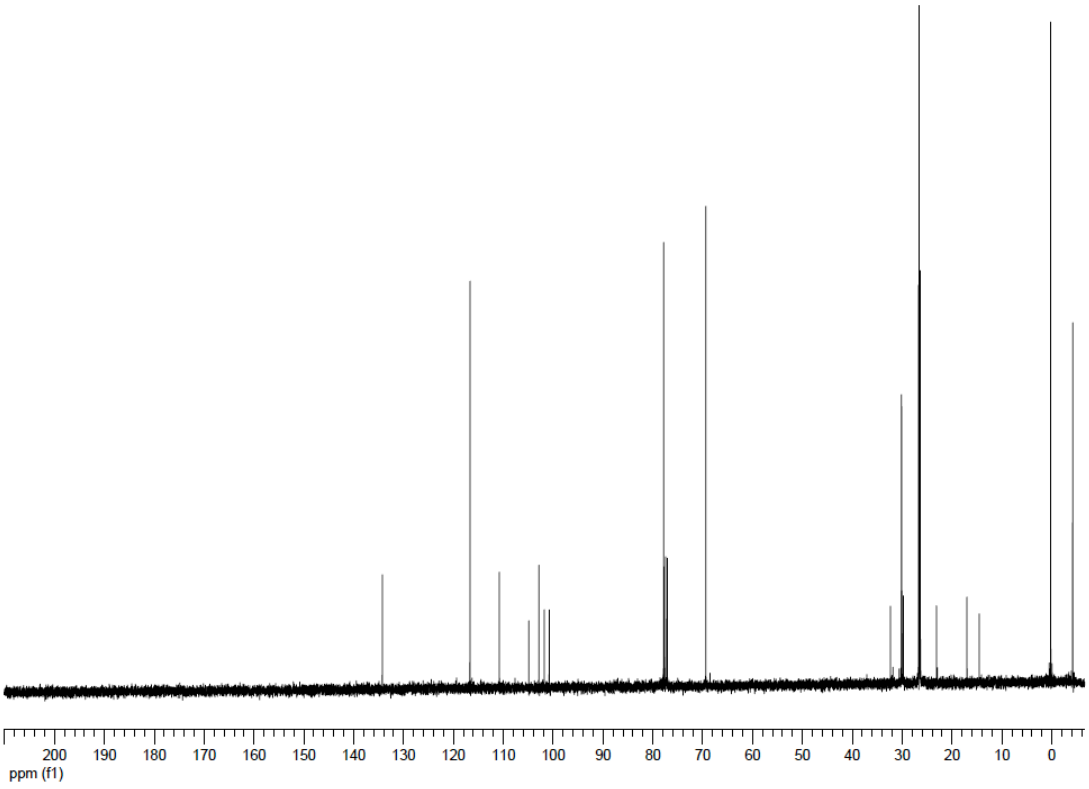
92

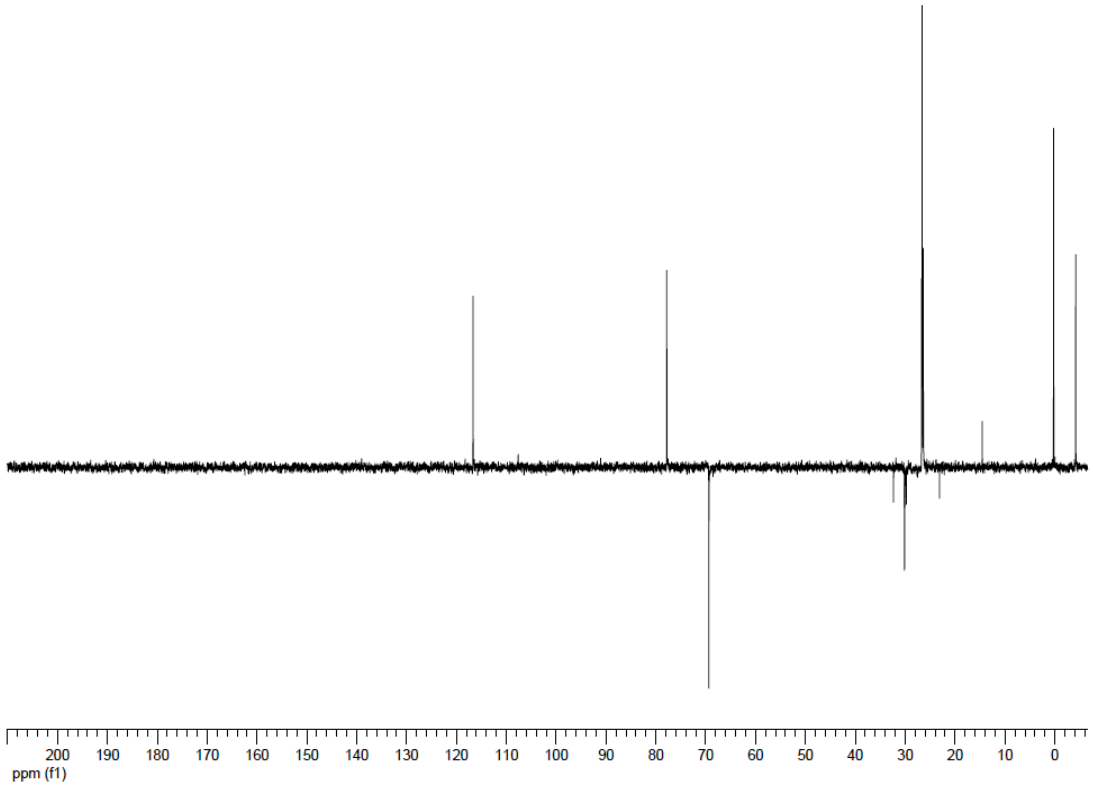
12.0
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

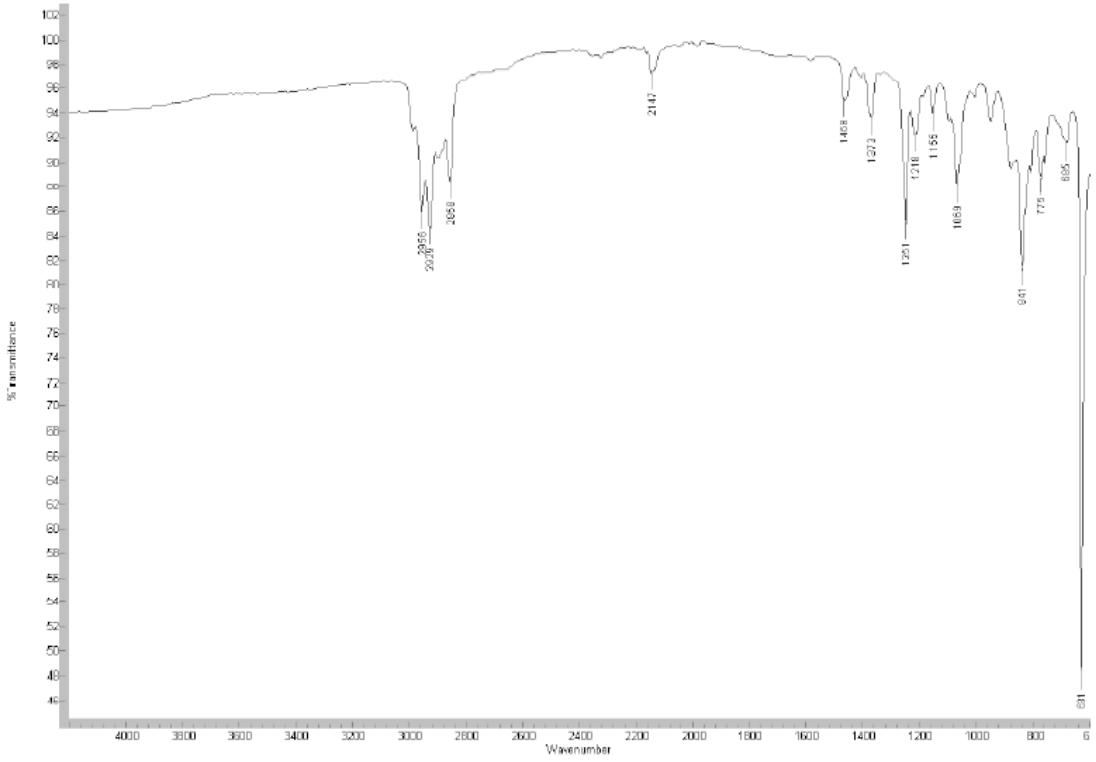

TBS
H
O
O
TMS

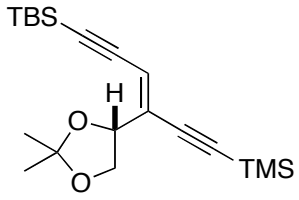
93

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)


200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm (f1)

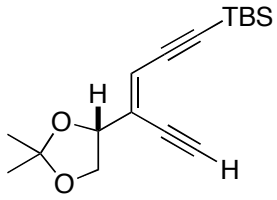

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)


% transmittance
2956
2926
2858
2147
1468
1373
1251
1216
1155
1059
941
775
695
631
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800 6
Wavenumber


TBS
H
O
O
TMS

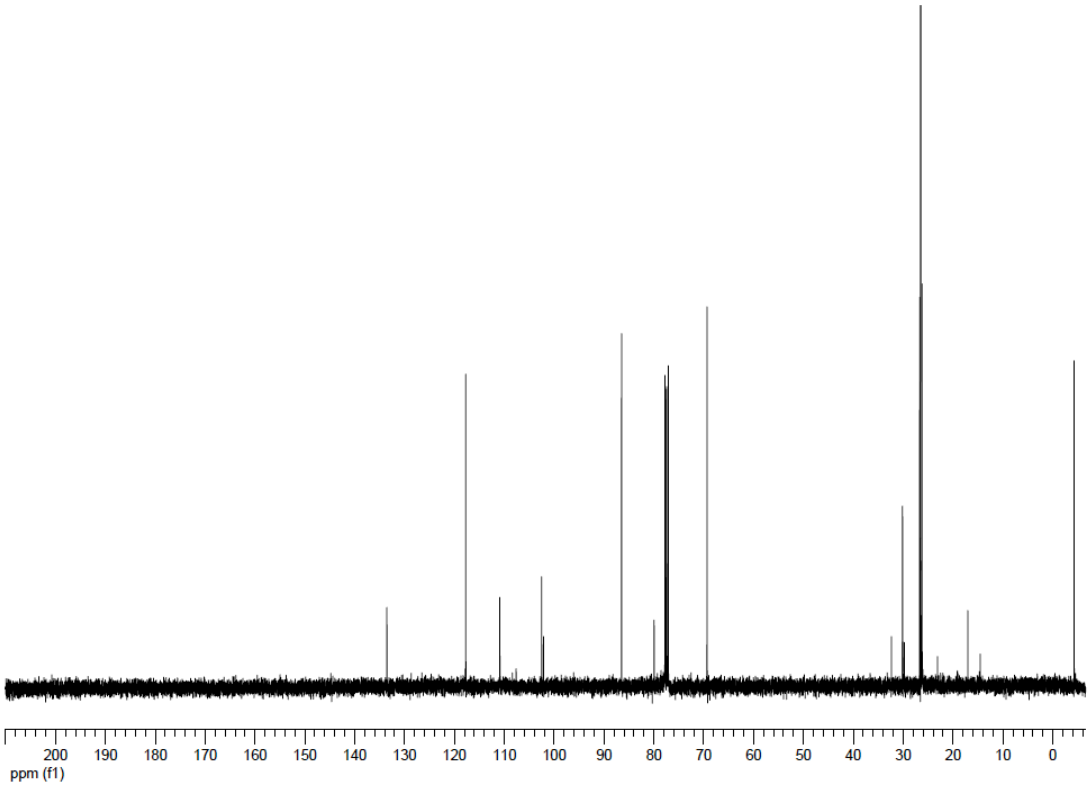
(*Z*)-**93**

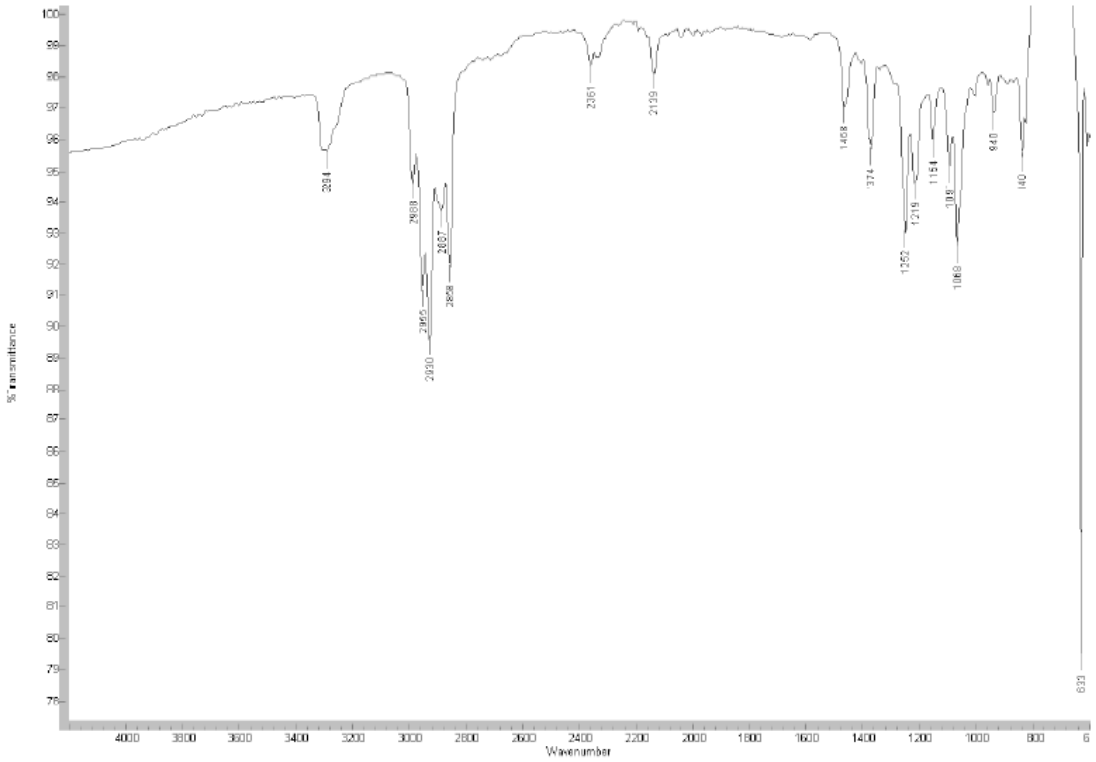
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

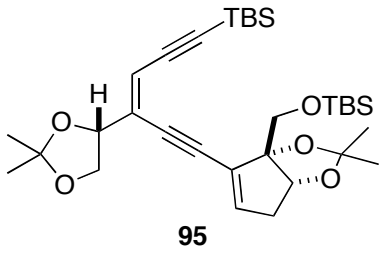

TBS
H
O
O
H

94

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (f1)

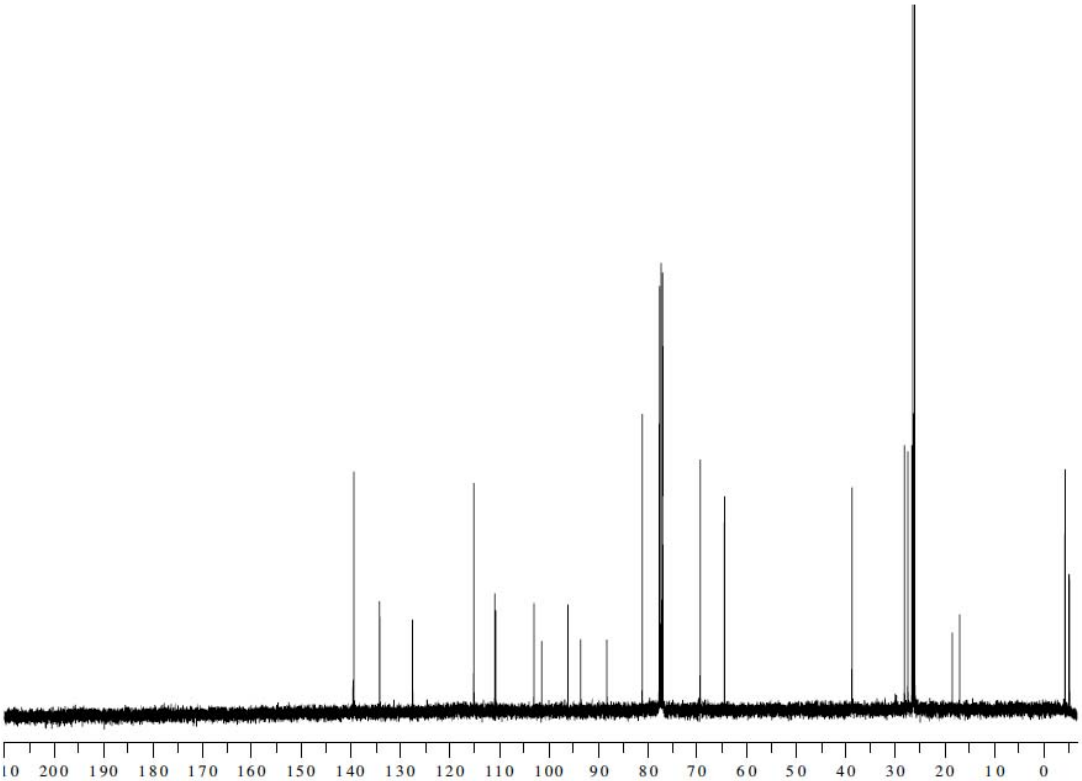

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

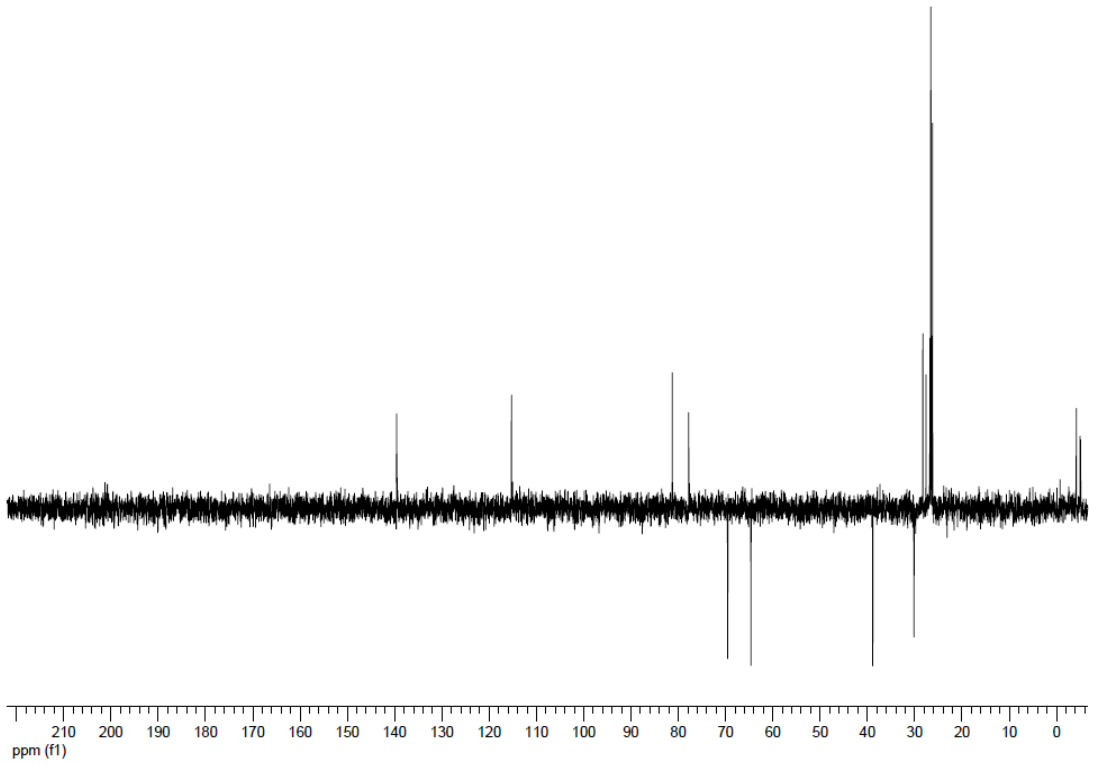

% Transmittance
Wavenumber
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800 6
2361
2199
1468
1374
1252
1219
1154
1068
940
140
633

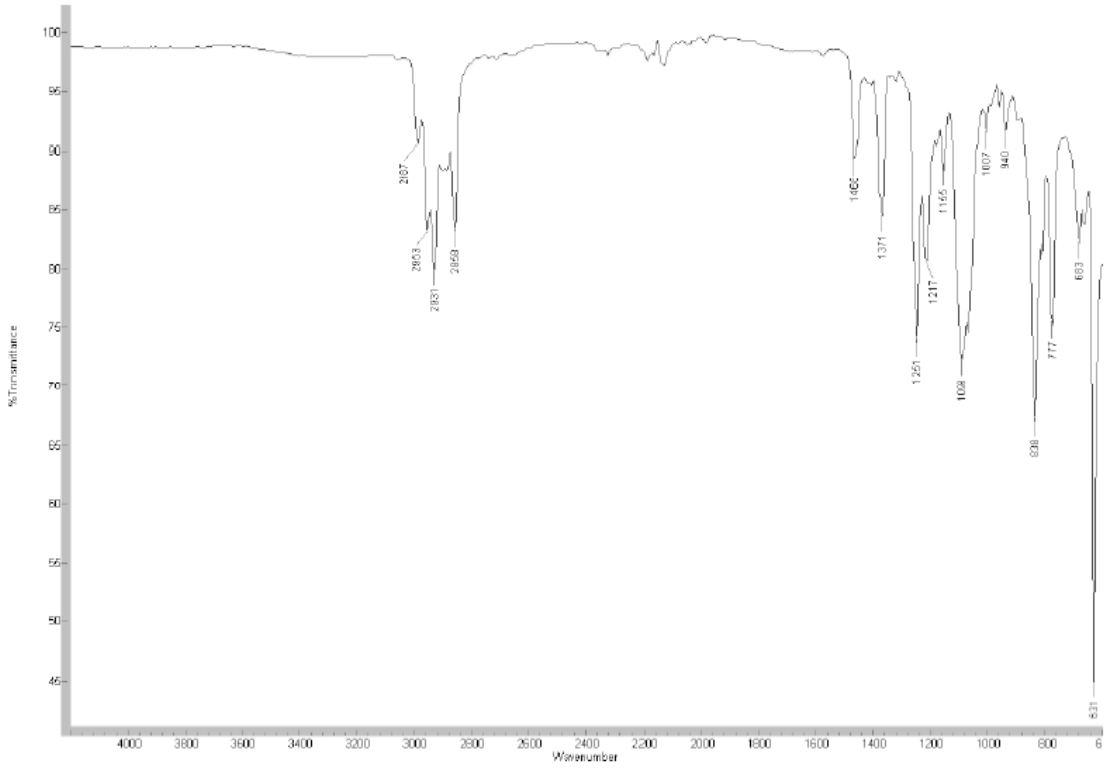

TBS
H
O
O
OTBS
O
O

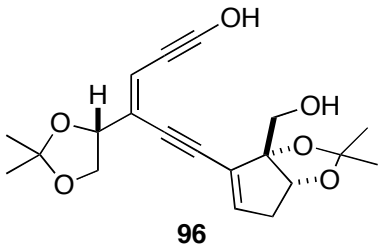
95

9
8
7
6
5
4
3
2
1
0


210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0

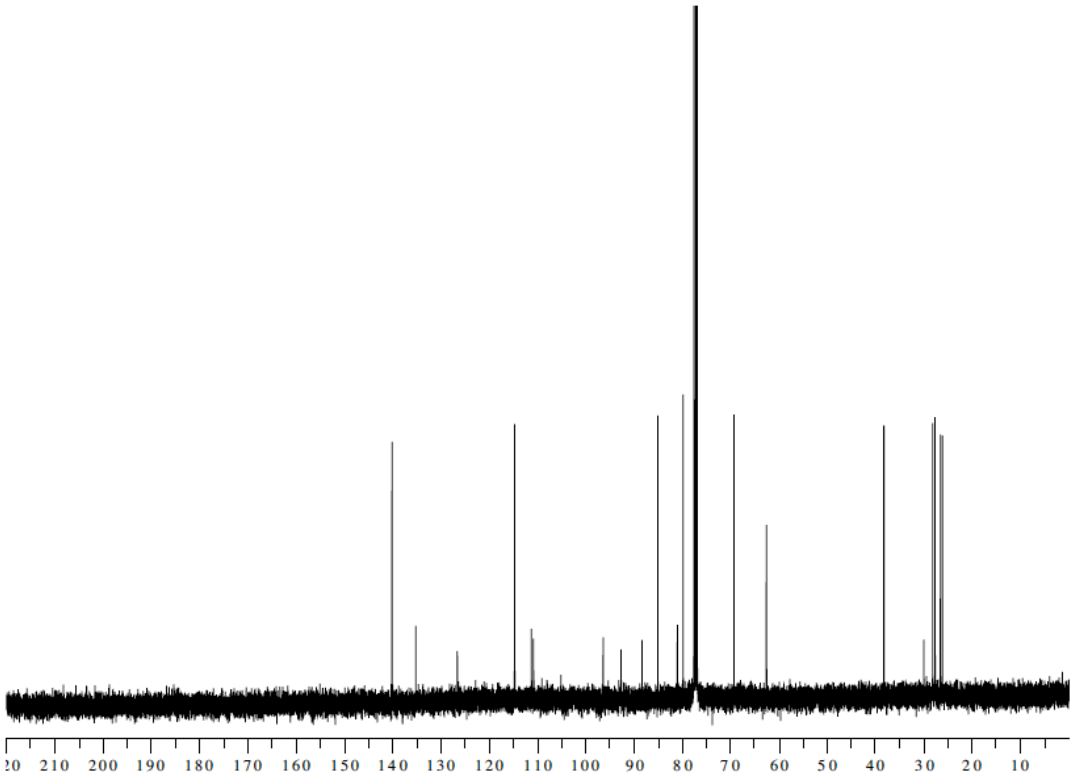

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
ppm (f1)

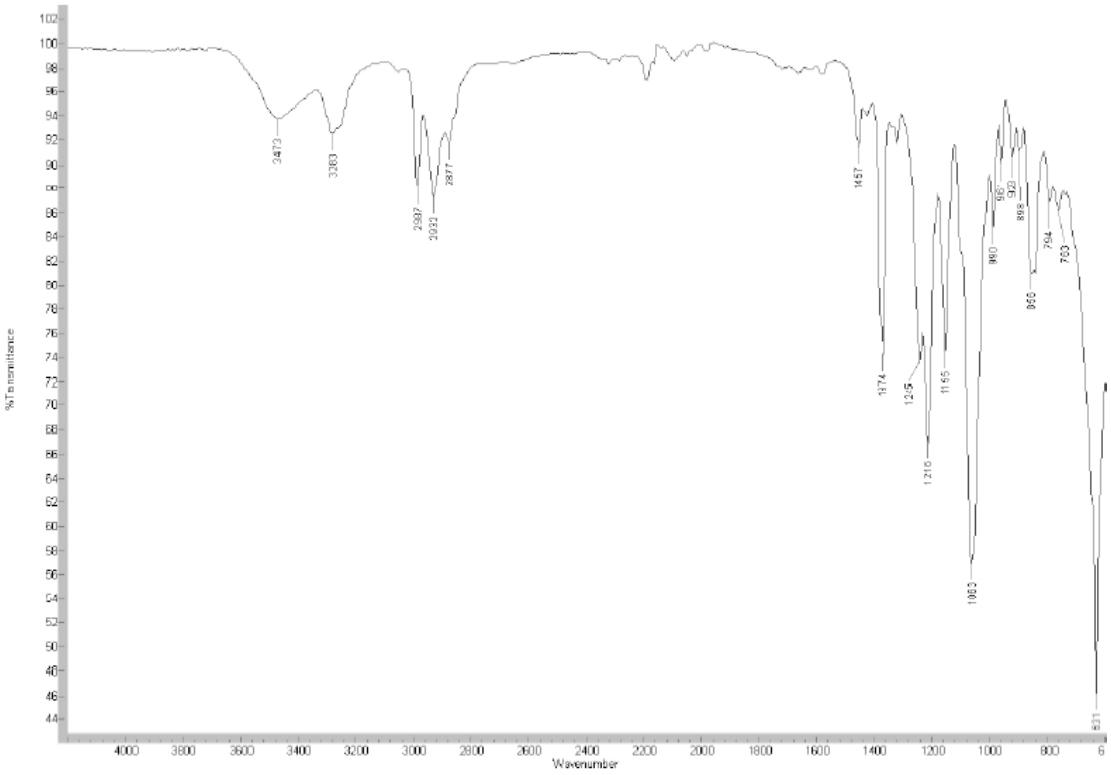

%Transmittance
Wavenumber
2957
2853
2831
2869
1466
1371
1251
1217
1165
1028
1007
940
838
777
683
631

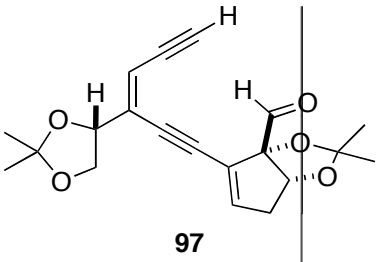

OH
H
O
O
OH
O
O

96

0
9
8
7
6
5
4
3
2
1

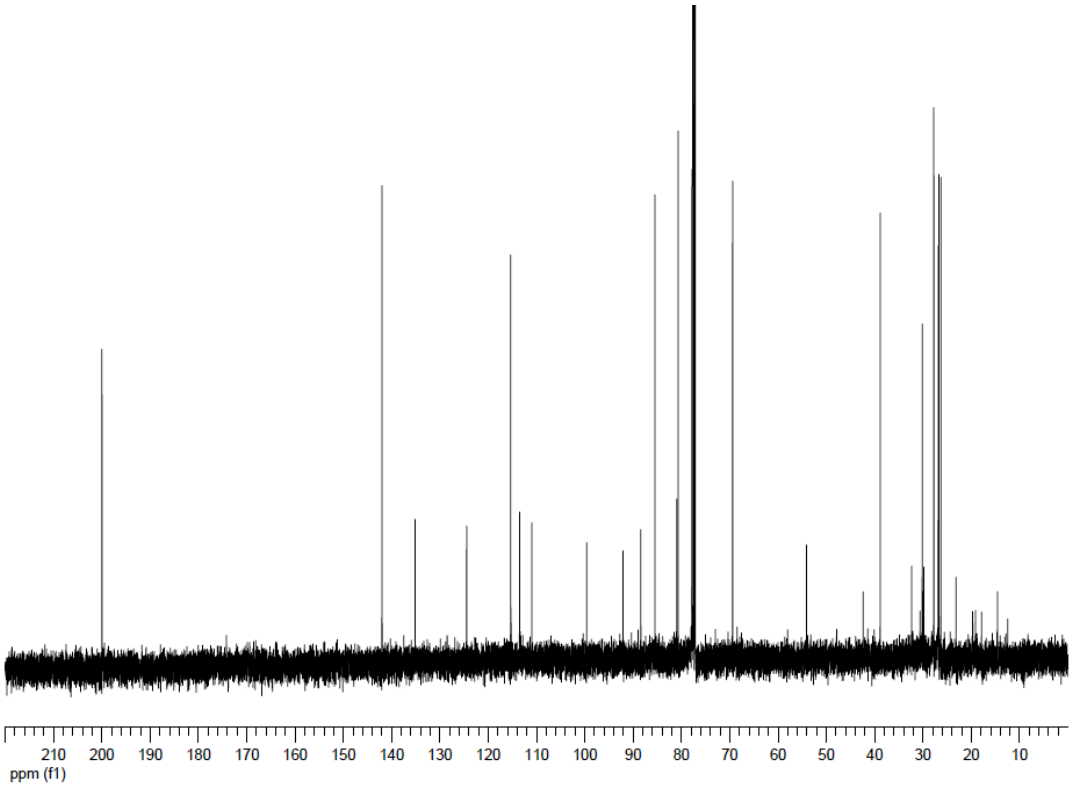

210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10

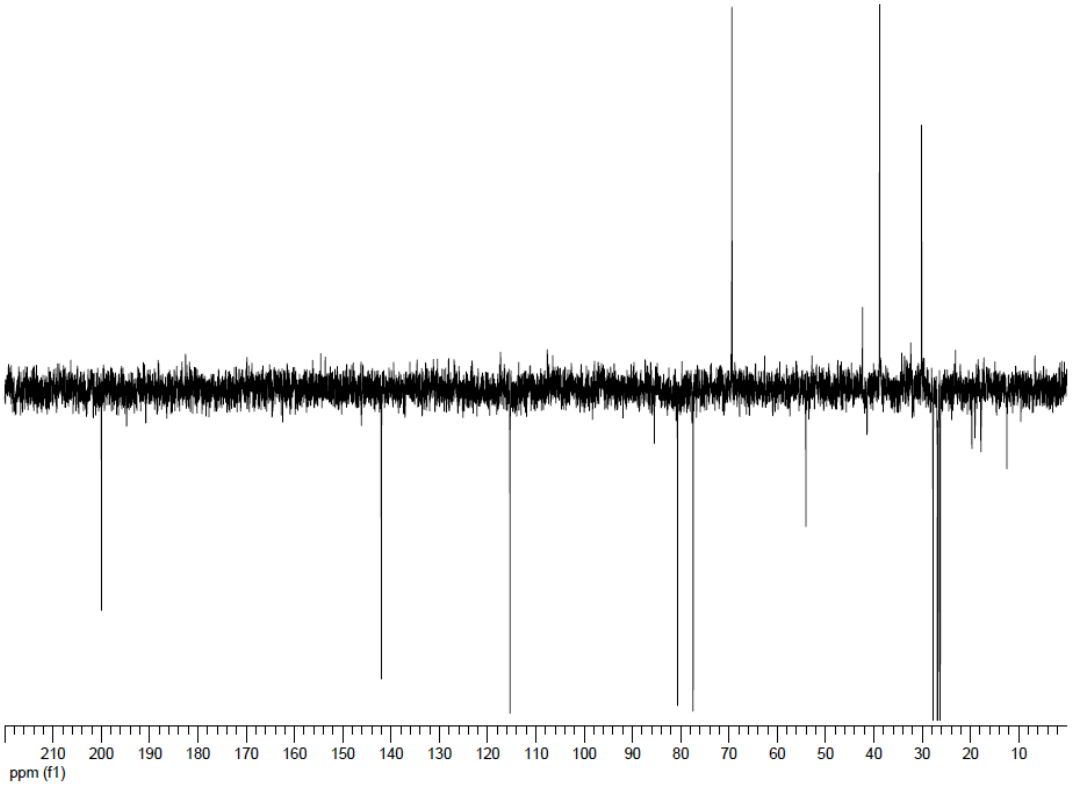

%Transmittance
Wavenumber
3473
3363
2997
2933
2877
1457
1374
1295
1210
1165
1063
980
960
928
905
856
794
753
631

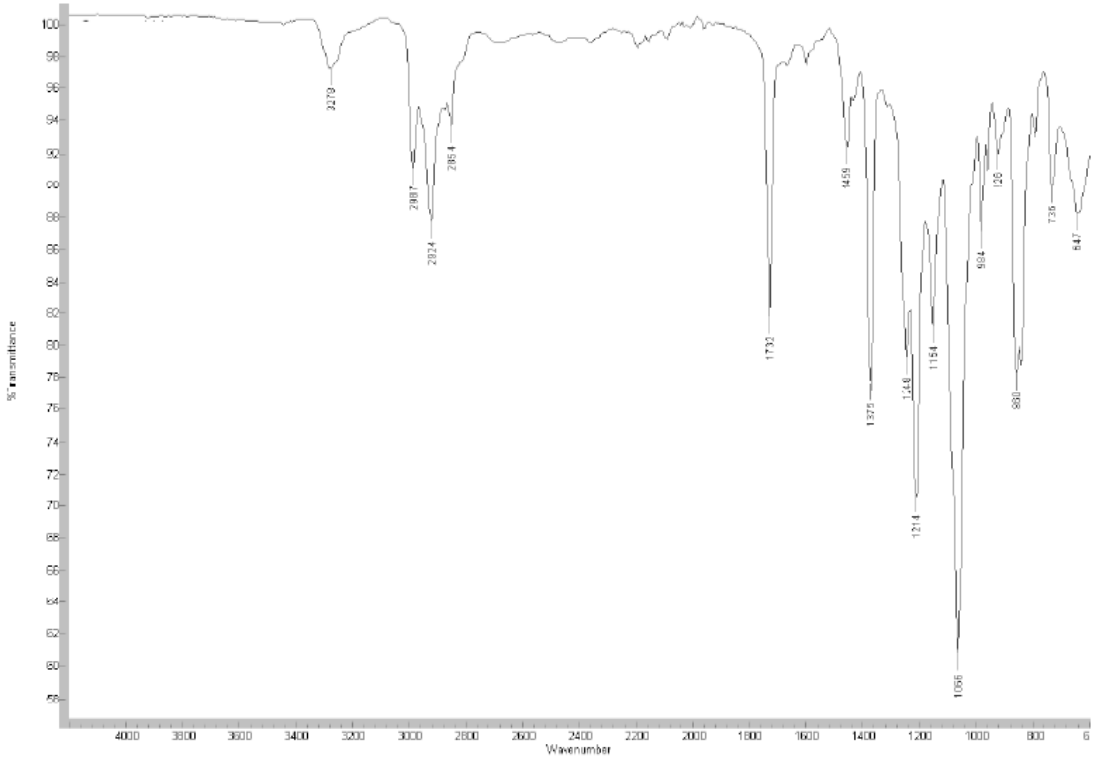

H
H
H
O
O
O
O
O

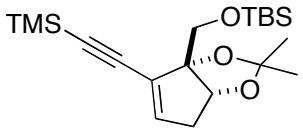
97

9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)


210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
ppm (f1)

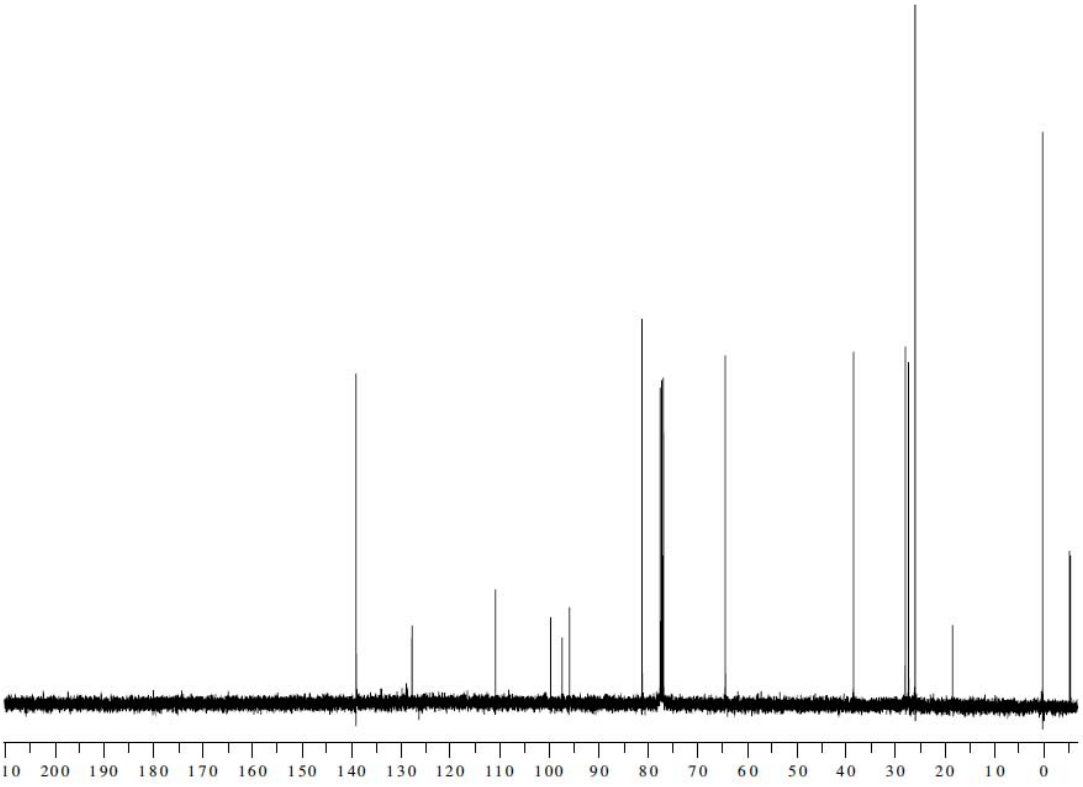

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
ppm (f1)


% Transmittance
Wavenumber
3279
2987
2924
2864
1732
1459
1375
1248
1214
1154
1066
994
905
860
735
647


TMS
OTBS
O
O

100

9
8
7
6
5
4
3
2
1
0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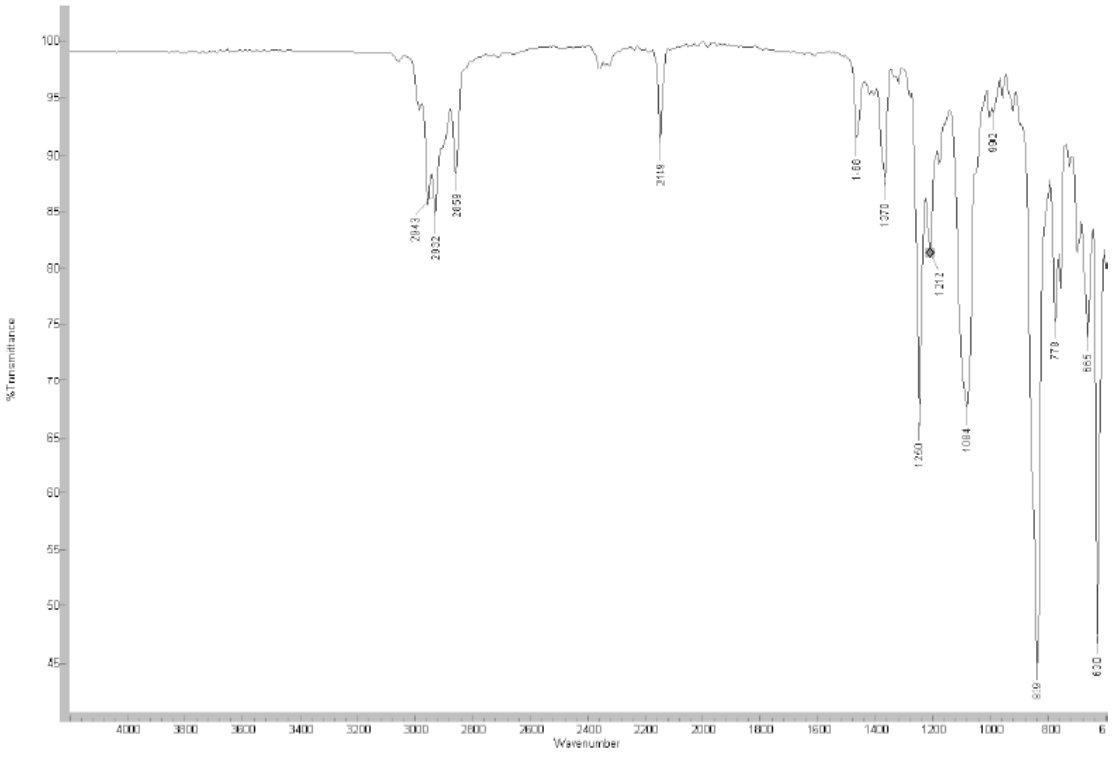
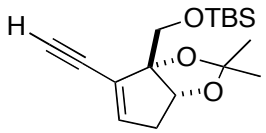
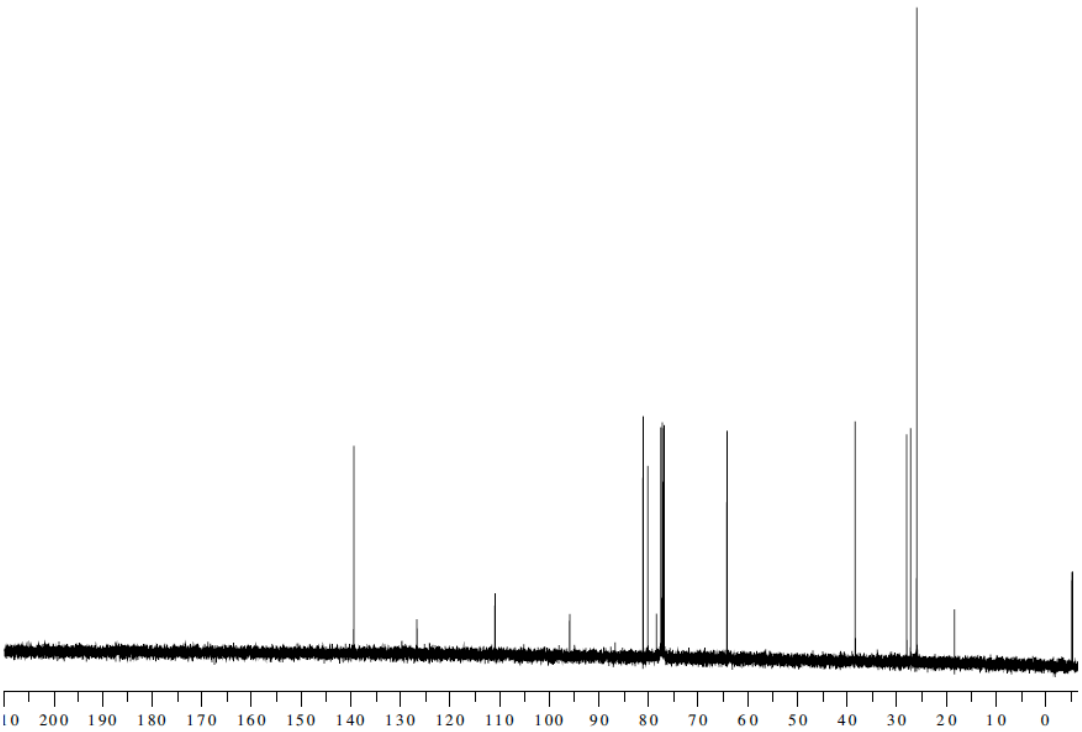

%Transmittance
Wavenumber
2943
2932
2859
2119
1466
1370
1212
1260
1084
992
778
665
630
4000 3800 3600 3400 3200 3000 2800 2600 2400 2200 2000 1800 1600 1400 1200 1000 800 6


H
OTBS
O
O

101

9
8
7
6
5
4
3
2
1
0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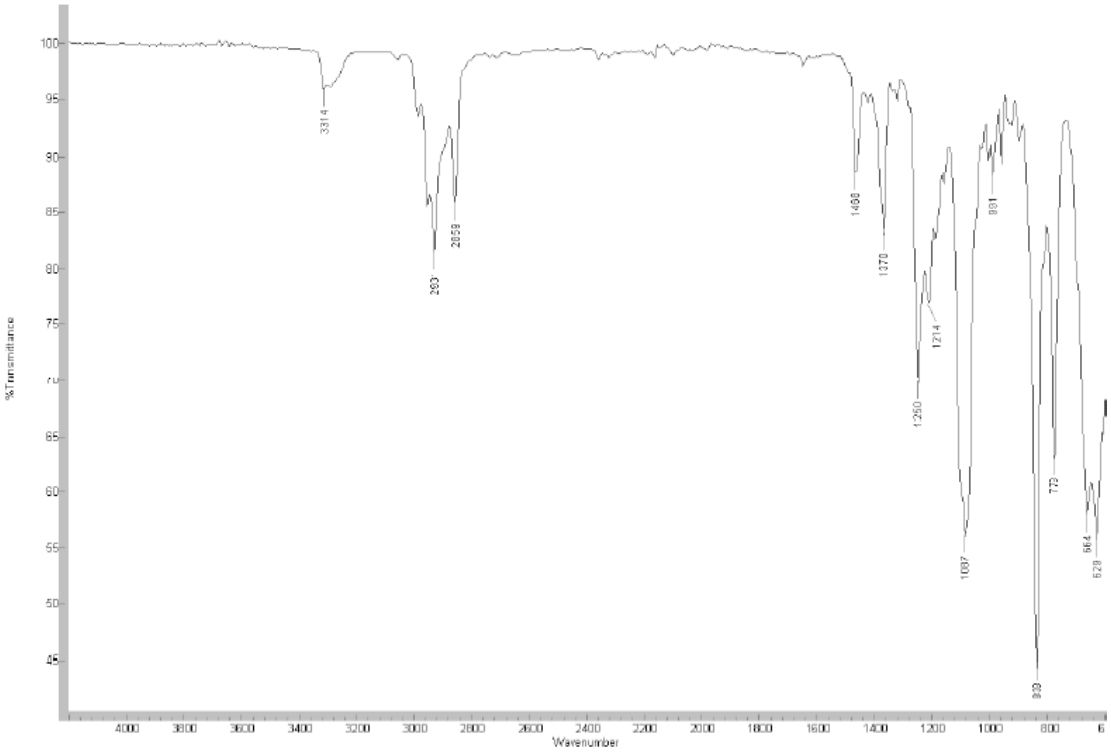
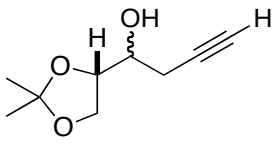
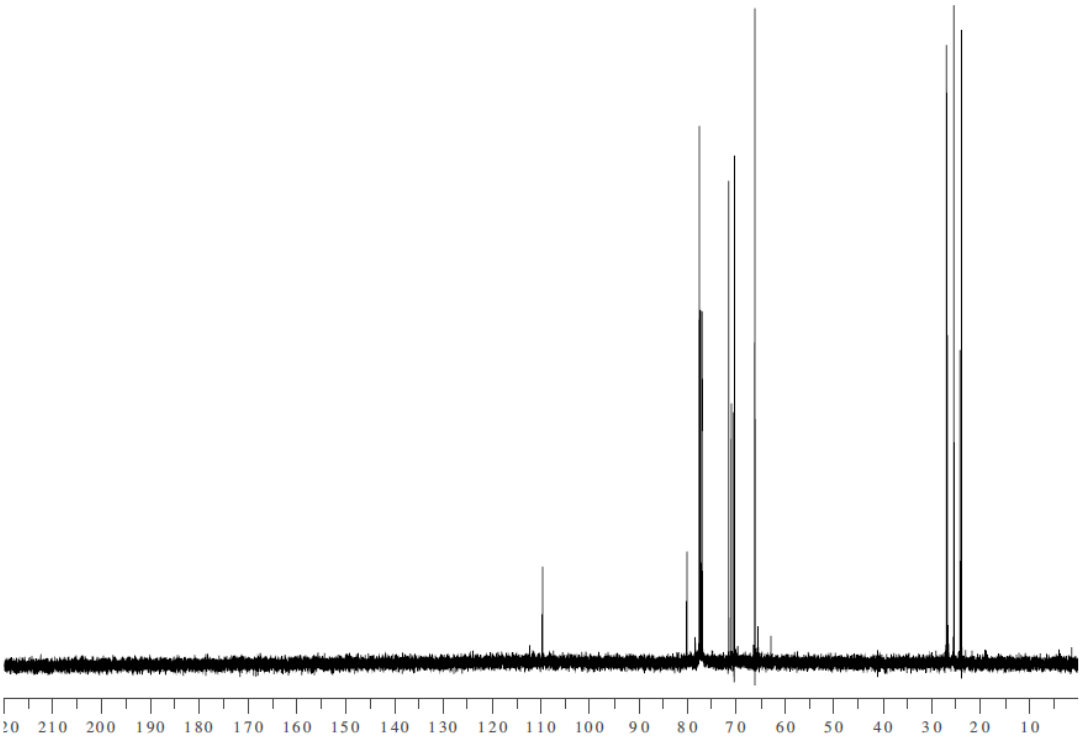
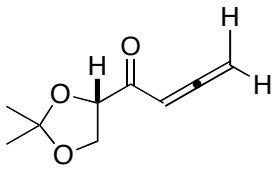

%Transmittance
Wavenumber
3314
2983
2859
1498
1370
1260
1214
1087
981
919
773
664
628


OH
H
H
O
O

102

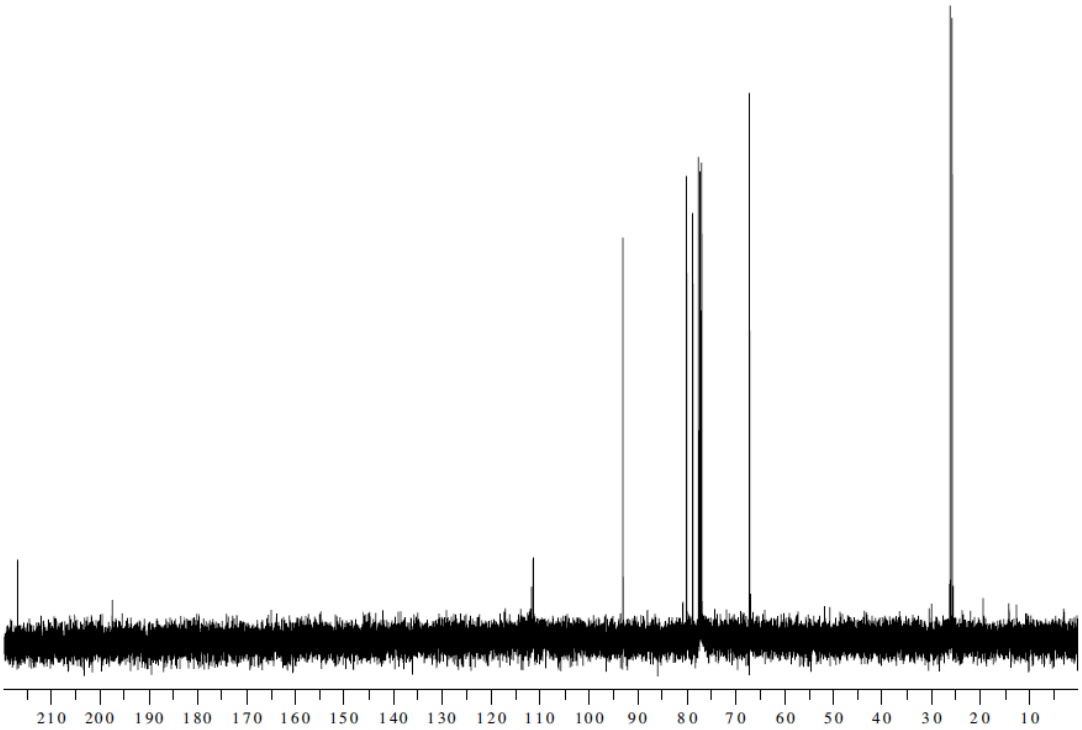
0
9
8
7
6
5
4
3
2
1

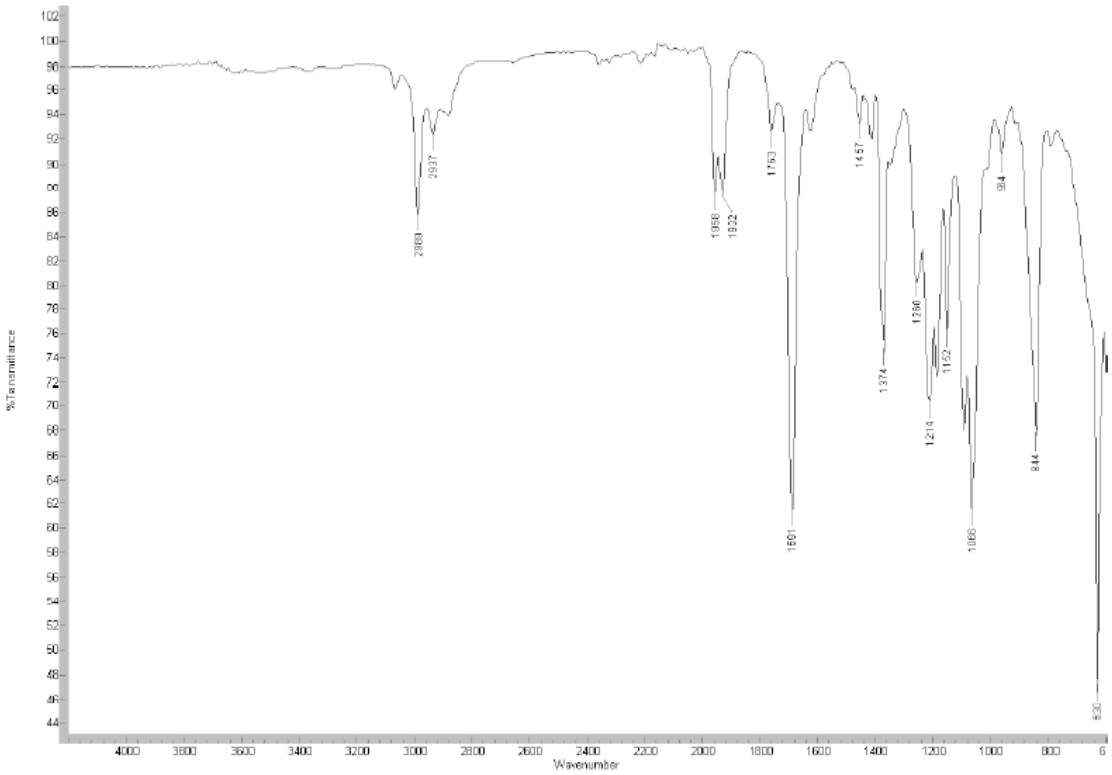

220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10

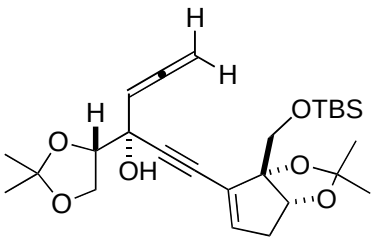

H
O
H
O
H
O

104

0 9 8 7 6 5 4 3 2 1

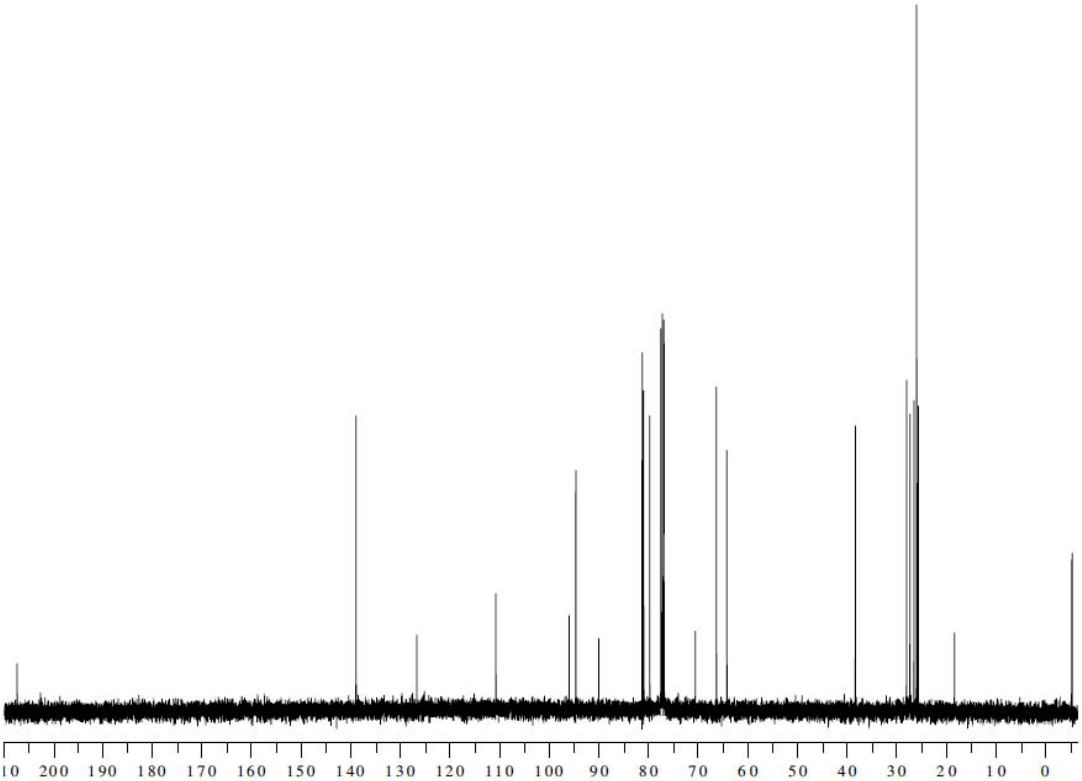

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10


%Transmittance
Wavenumber
2869
2937
1958
1932
1763
1691
1457
1374
1290
1214
1152
1068
984
844
530


H
H
H
OTBS
O
O
OH
O
O

105

9
8
7
6
5
4
3
2
1
0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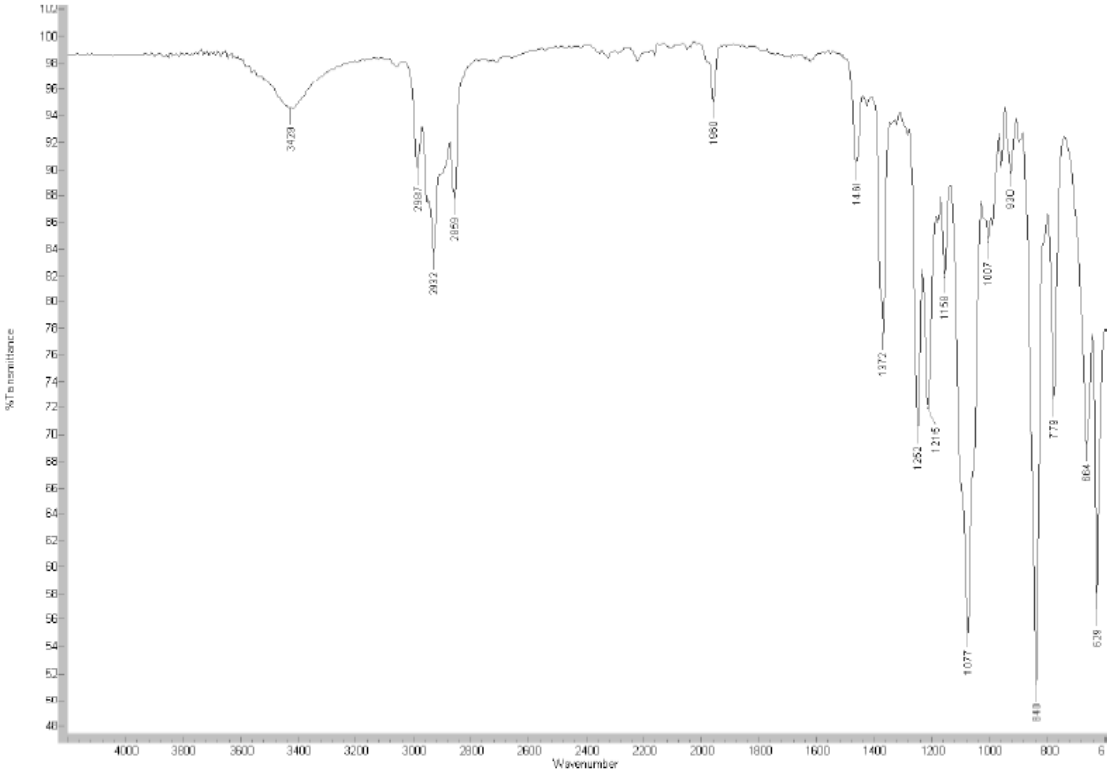
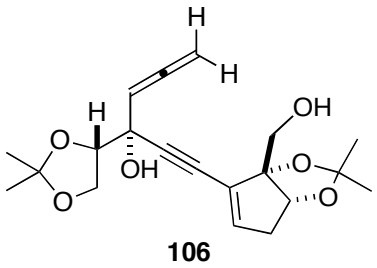
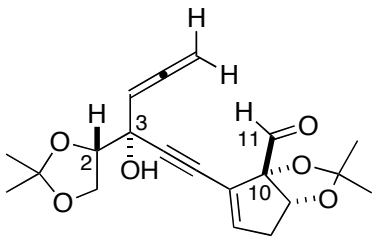

%Transmittance
Wavenumber
3429
2987
2932
2869
1960
1491
1372
1252
1215
1158
1077
1007
930
848
778
664
629

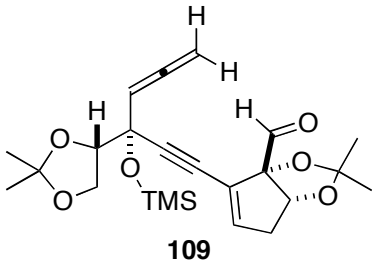

H
H
H
OH
OH
O
O
O
O

106

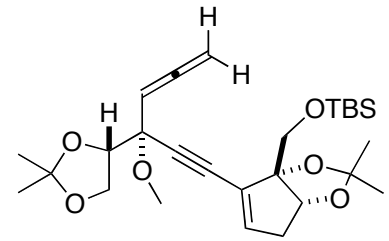
0
9
8
7
6
5
4
3
2
1



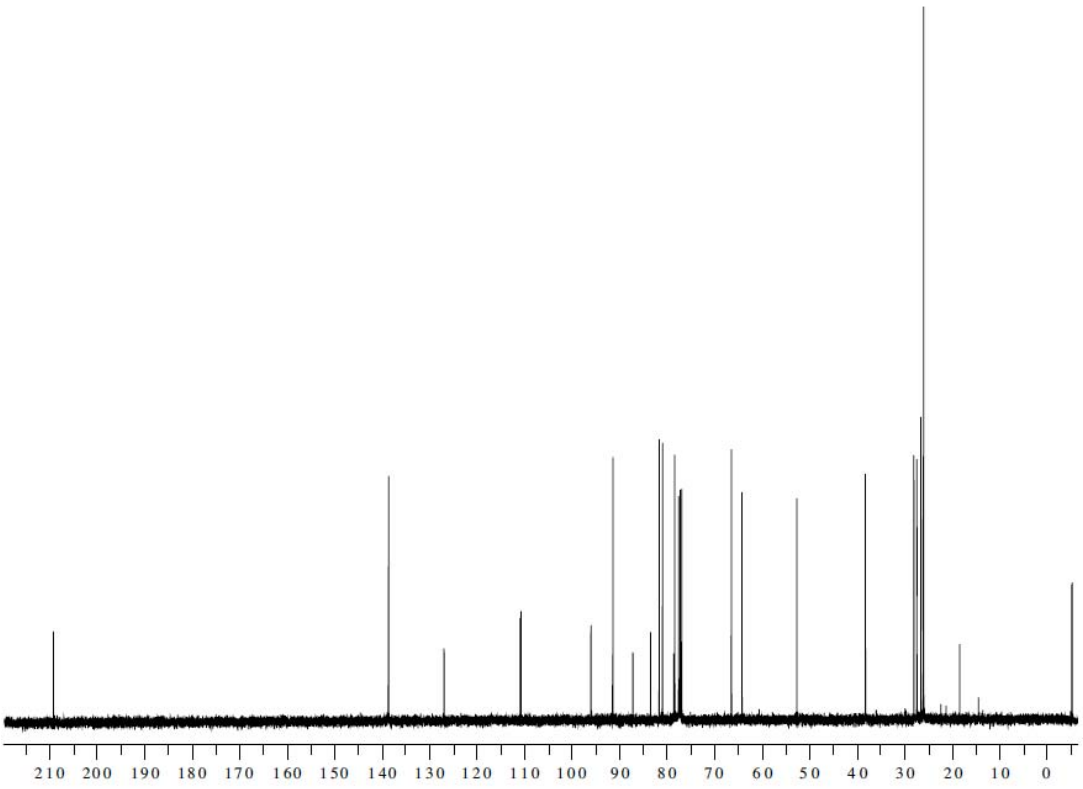
107

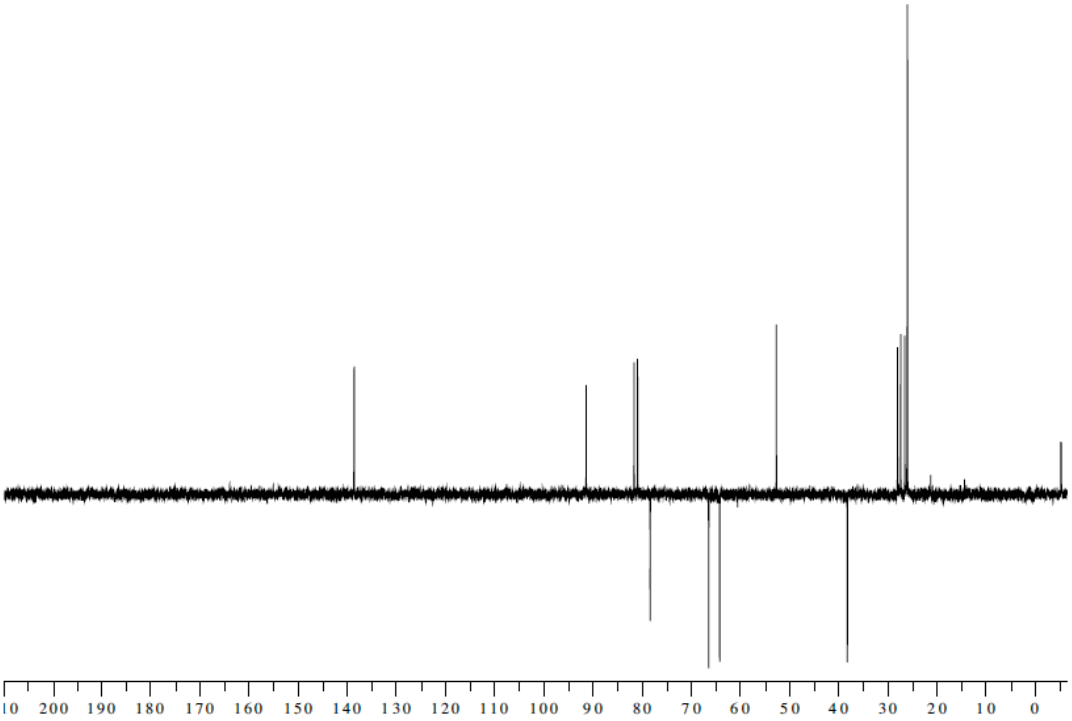


109


H
H
H
OTBS
O
O
O
O
O

9
8
7
6
5
4
3
2
1
0


210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0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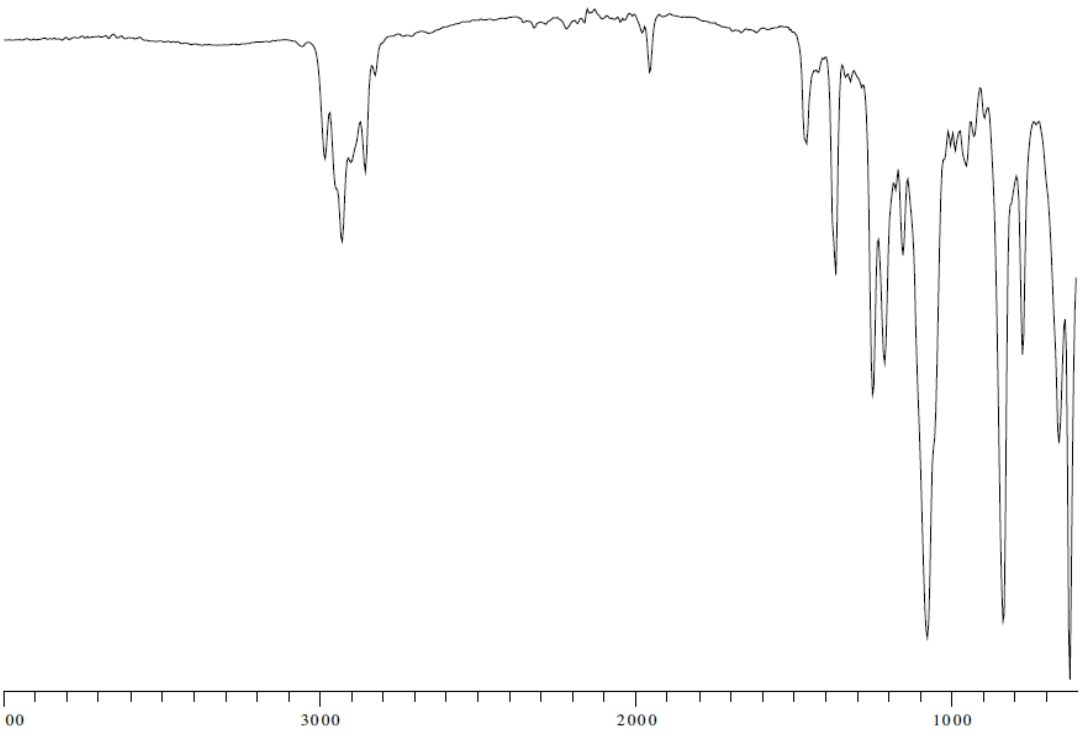
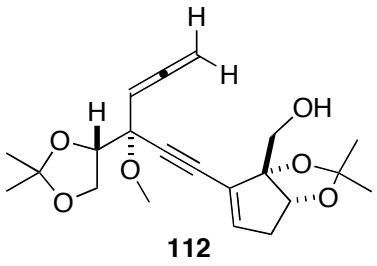
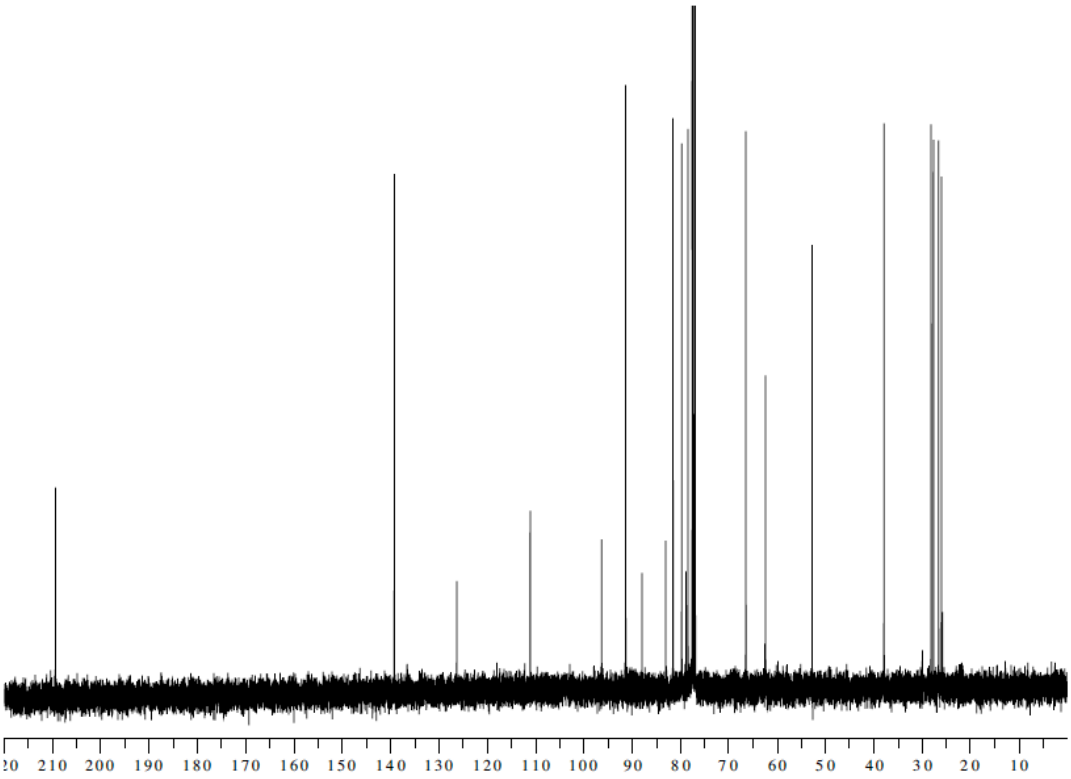
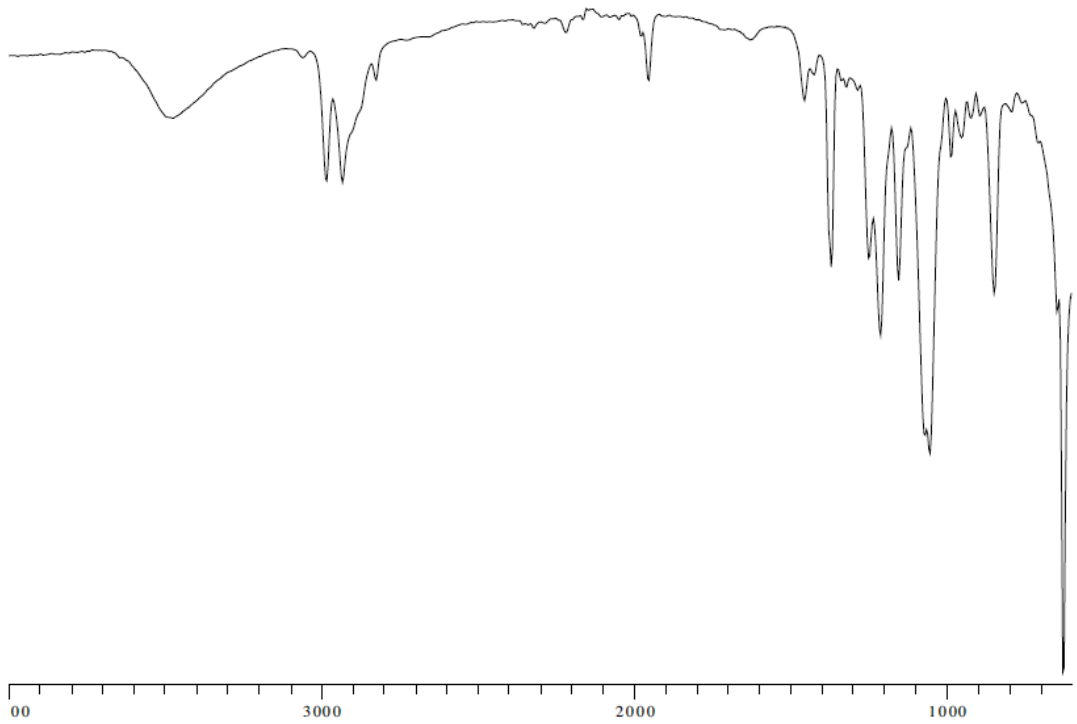

00 3000 2000 1000

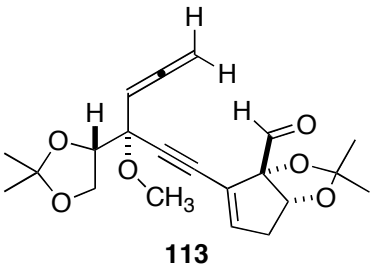

H
H
H
OH
O
O
O
O
O

112

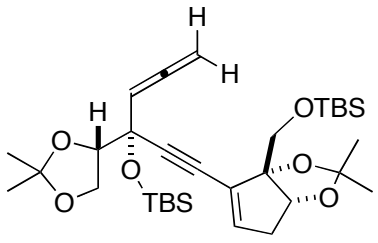
0
9
8
7
6
5
4
3
2
1


220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10



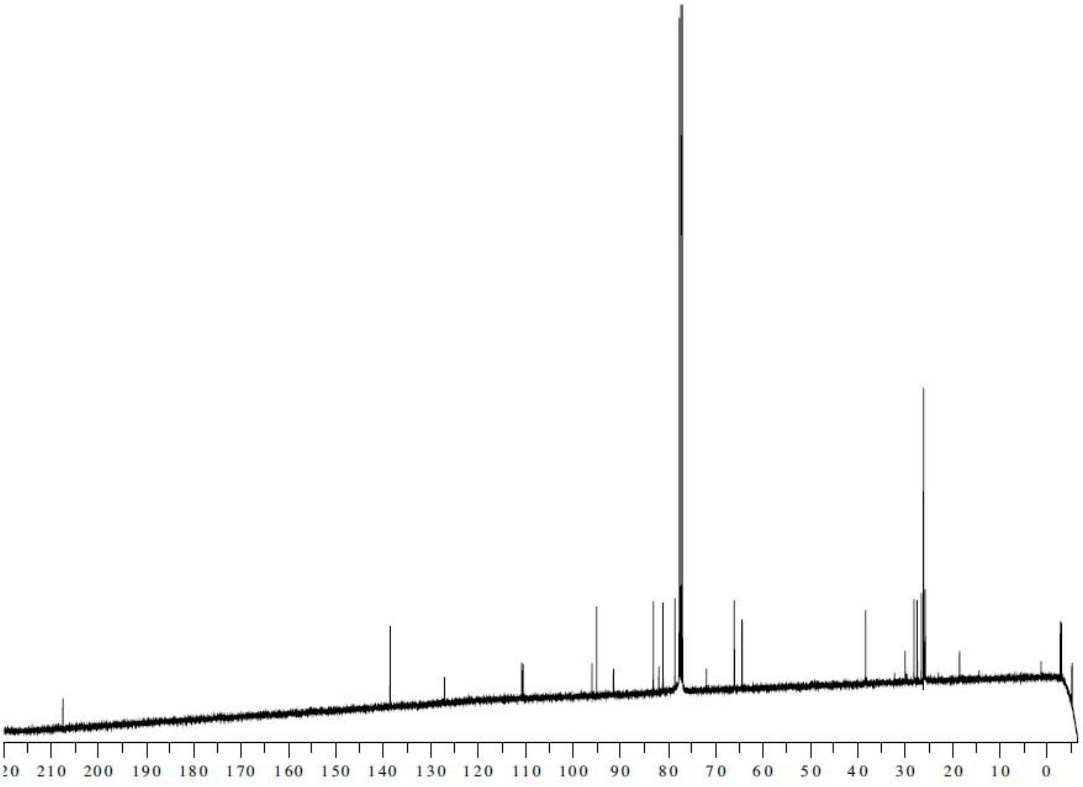


113

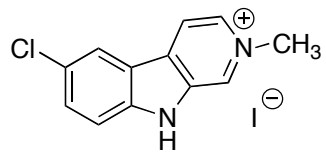

H
H
H
OTBS
O
O
O
TBS
O
O

115

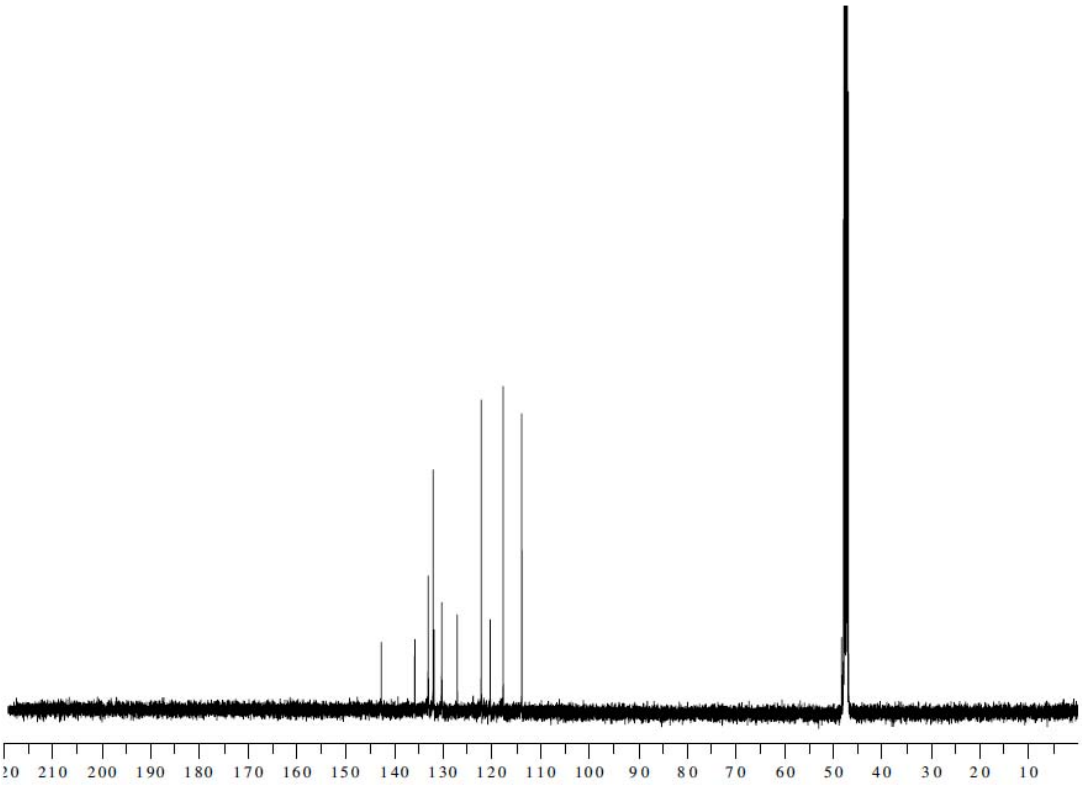
9
8
7
6
5
4
3
2
1
0

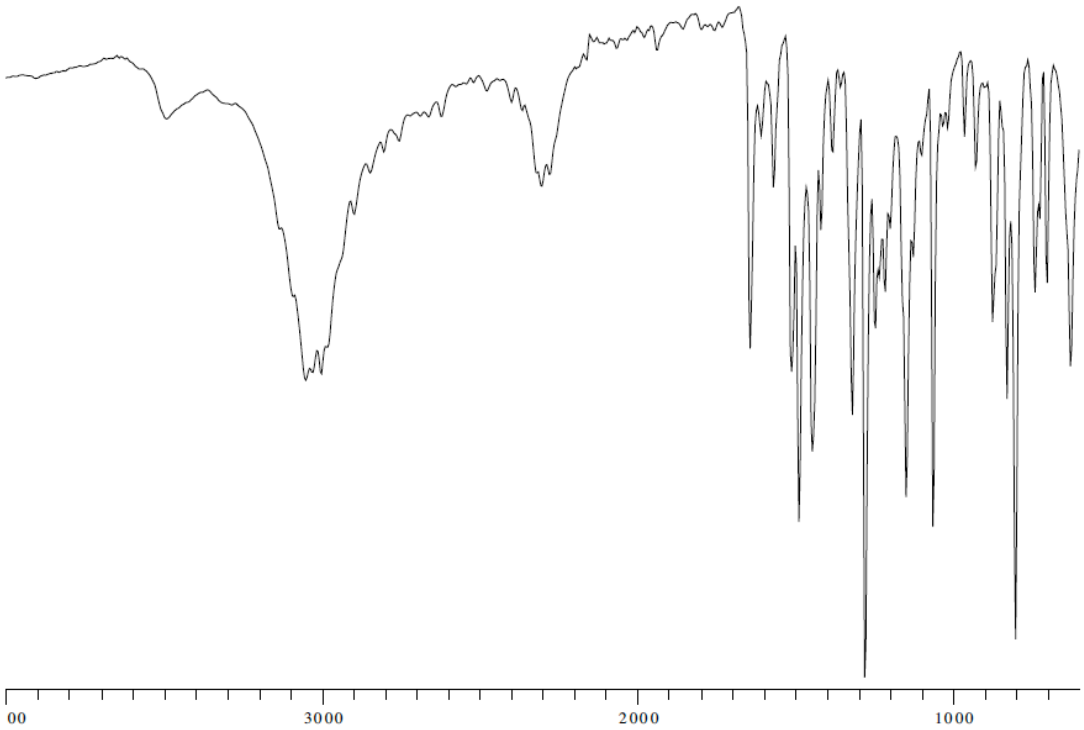

210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

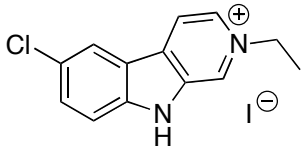
6.5.3. Spectra from the Nostocarboline and Eudistomin Derivatives Project



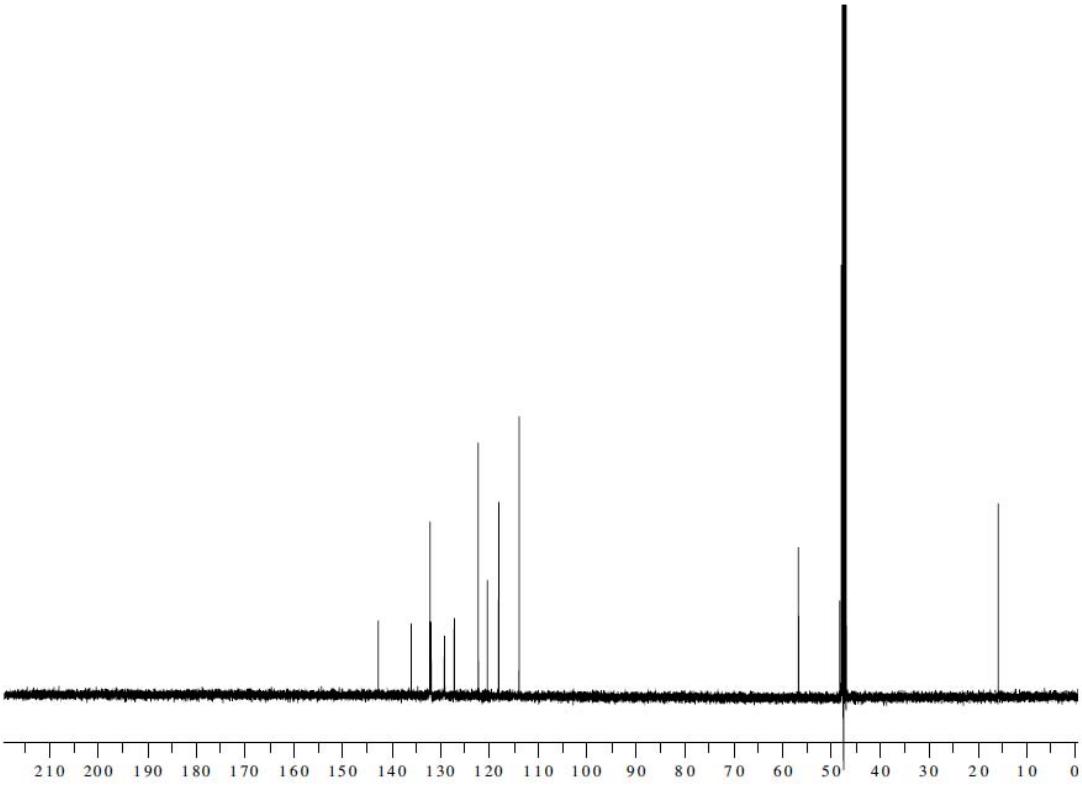
133







134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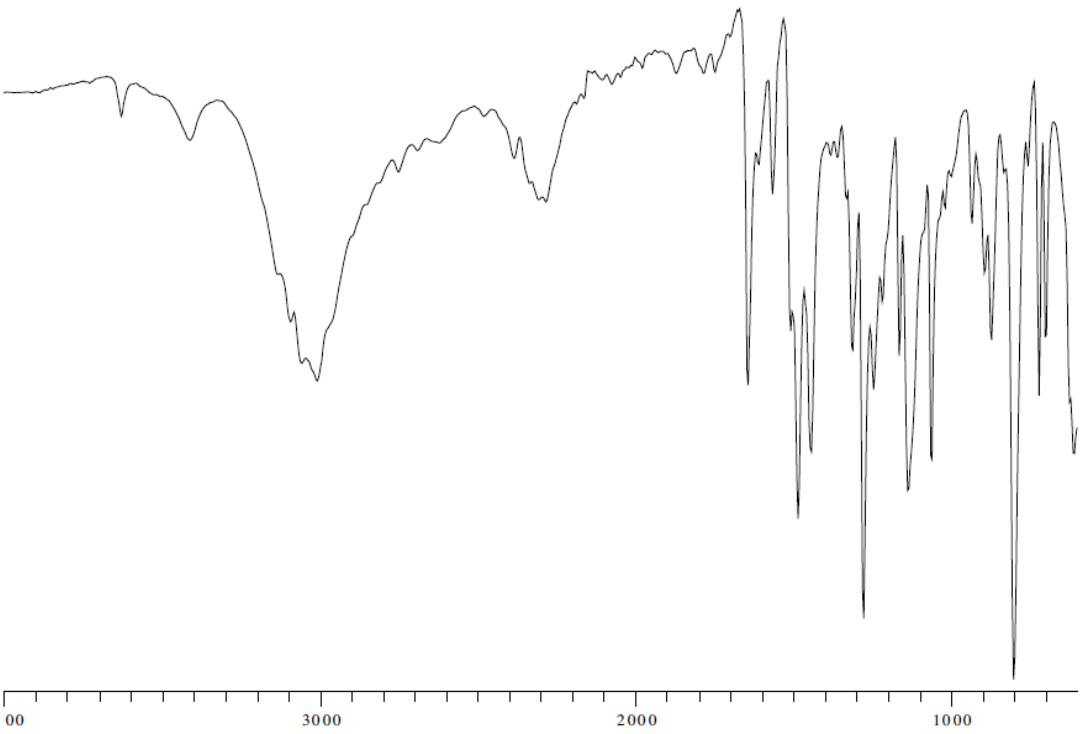
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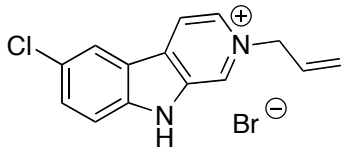

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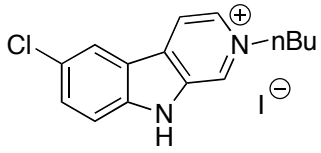

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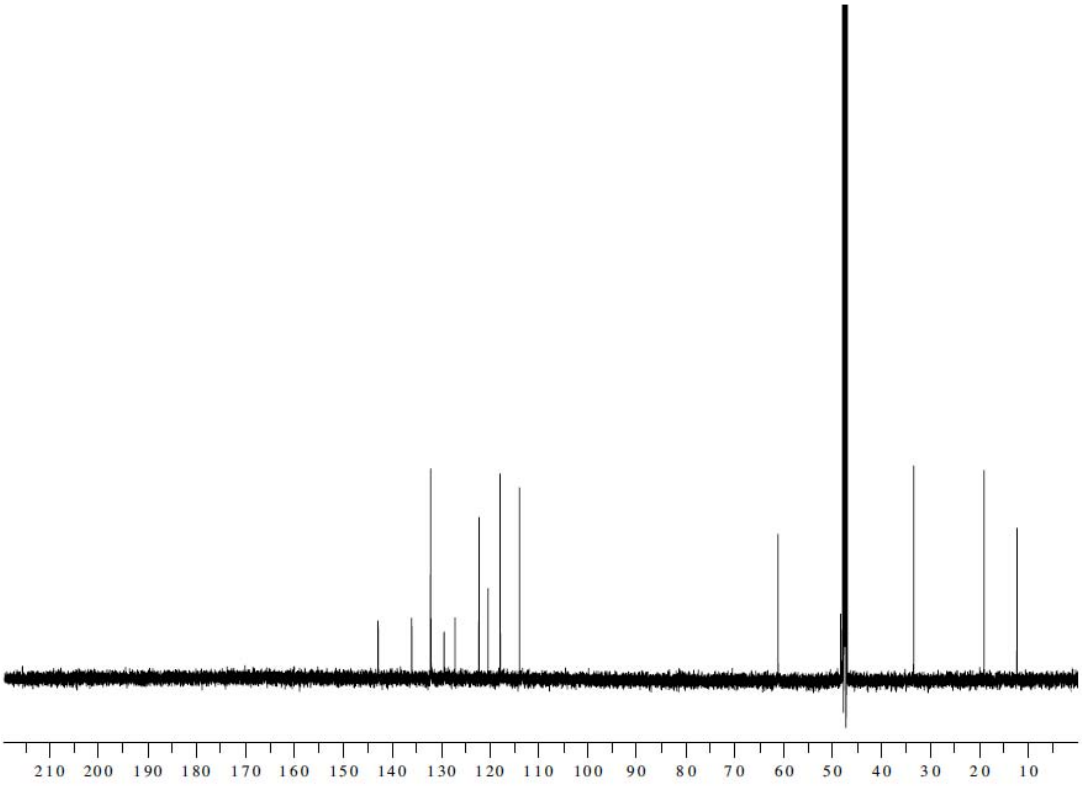
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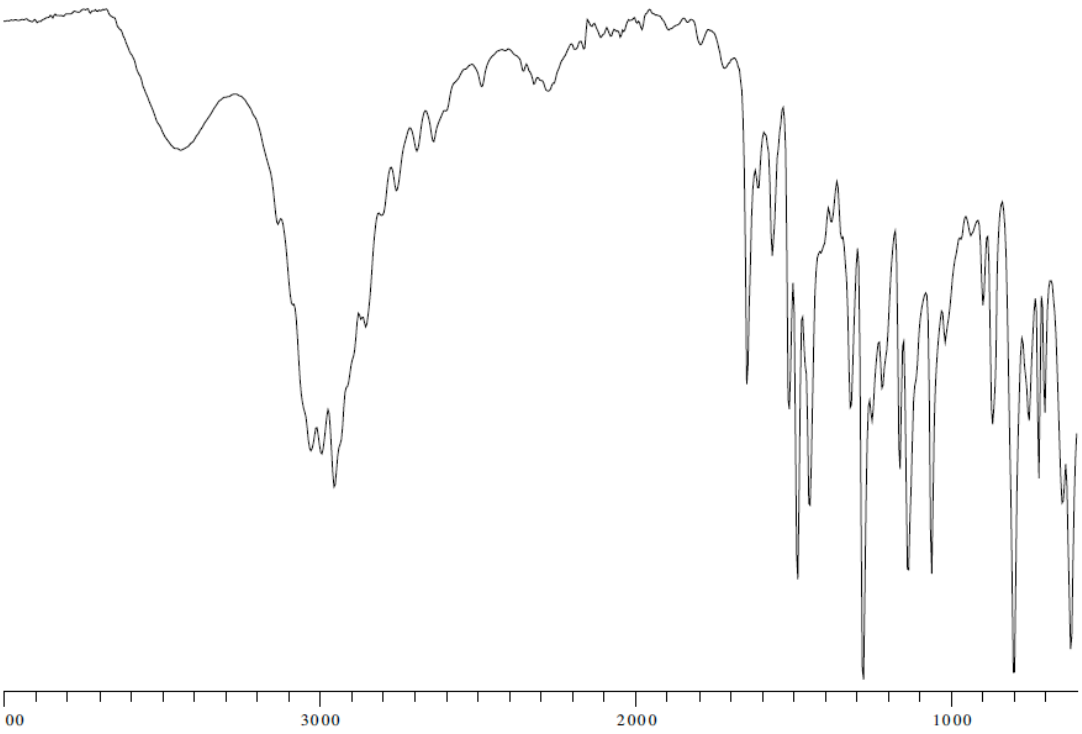
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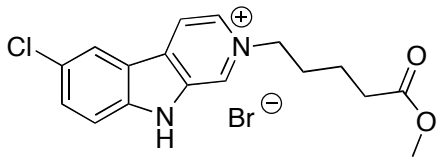
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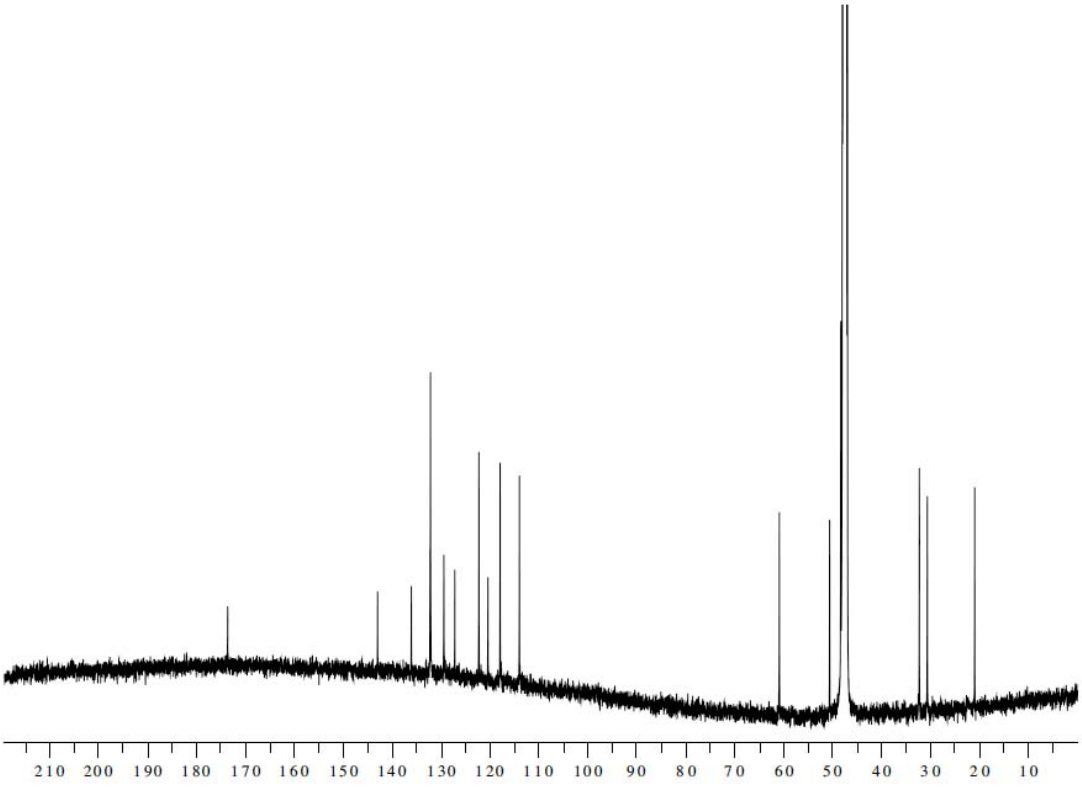
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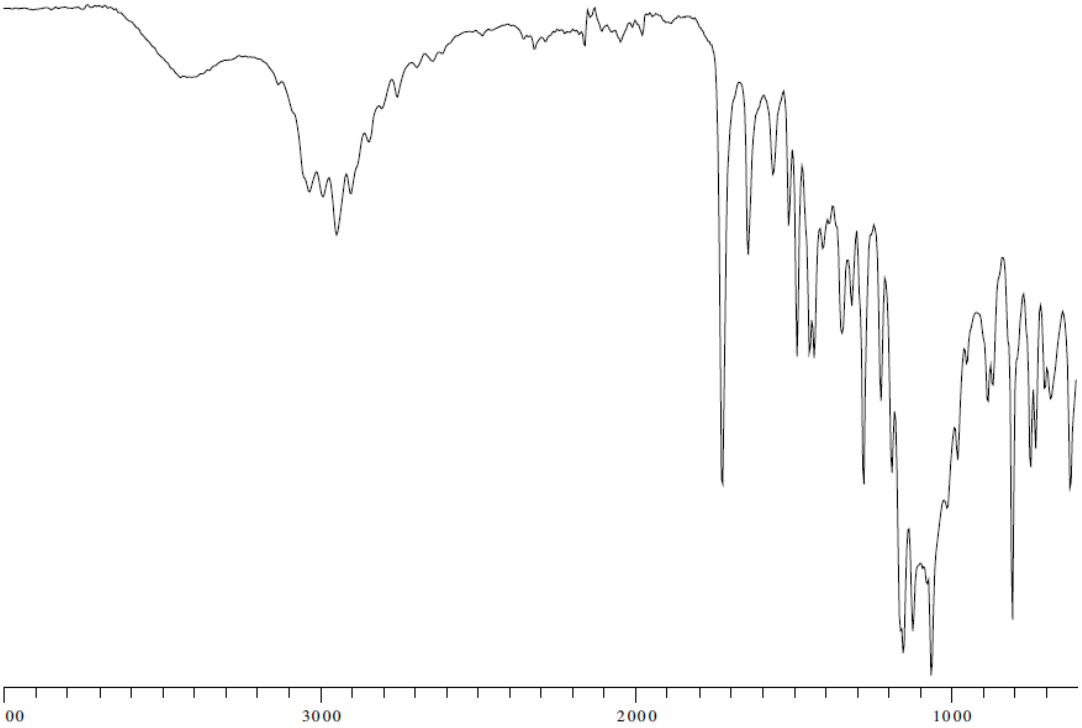

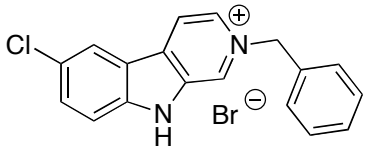

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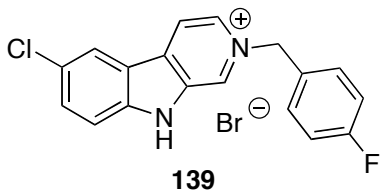
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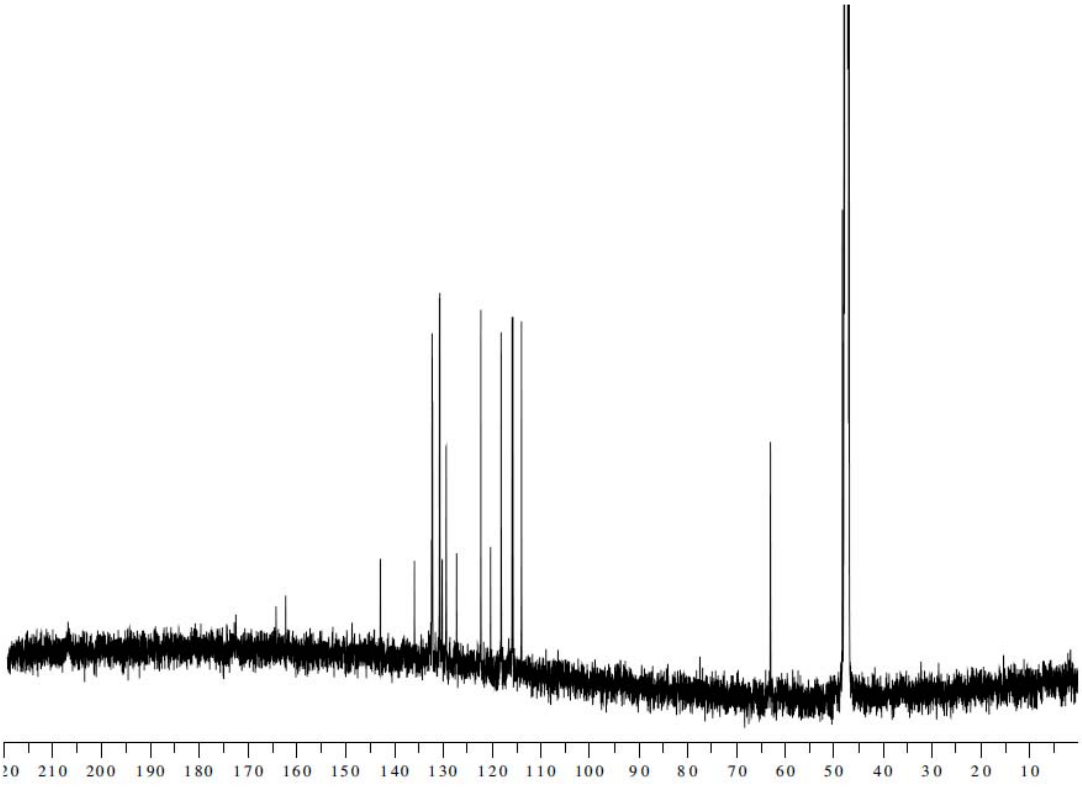
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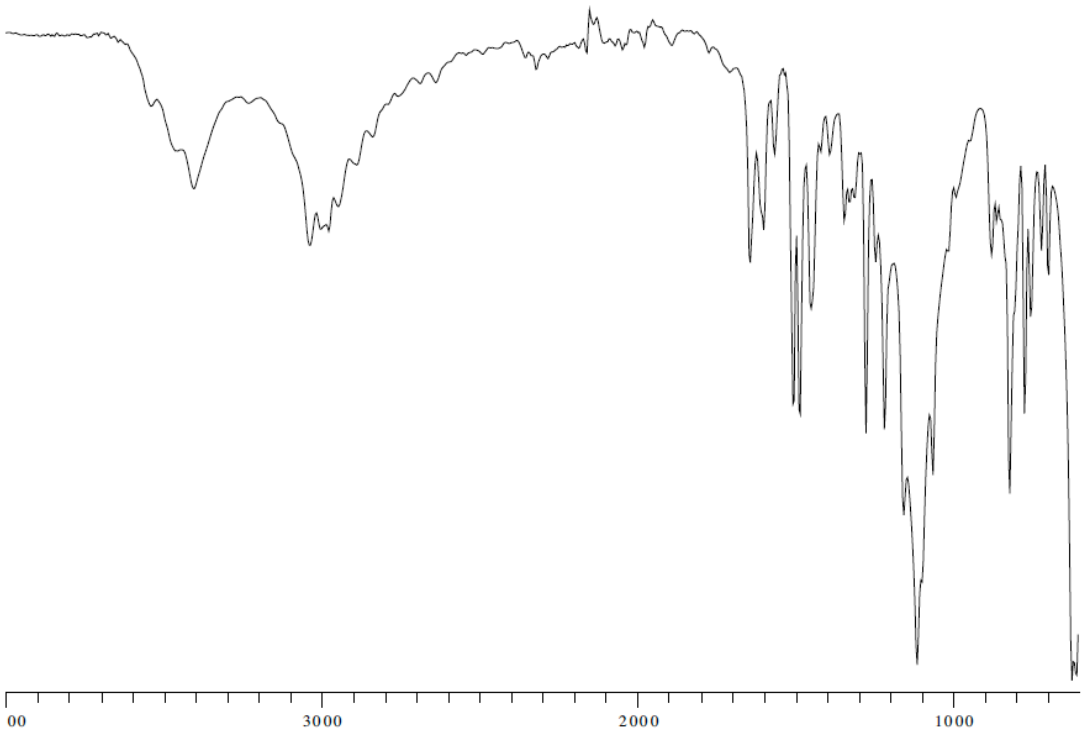

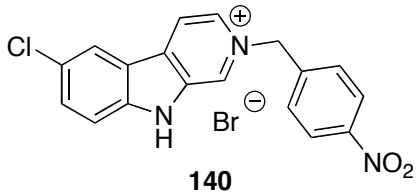

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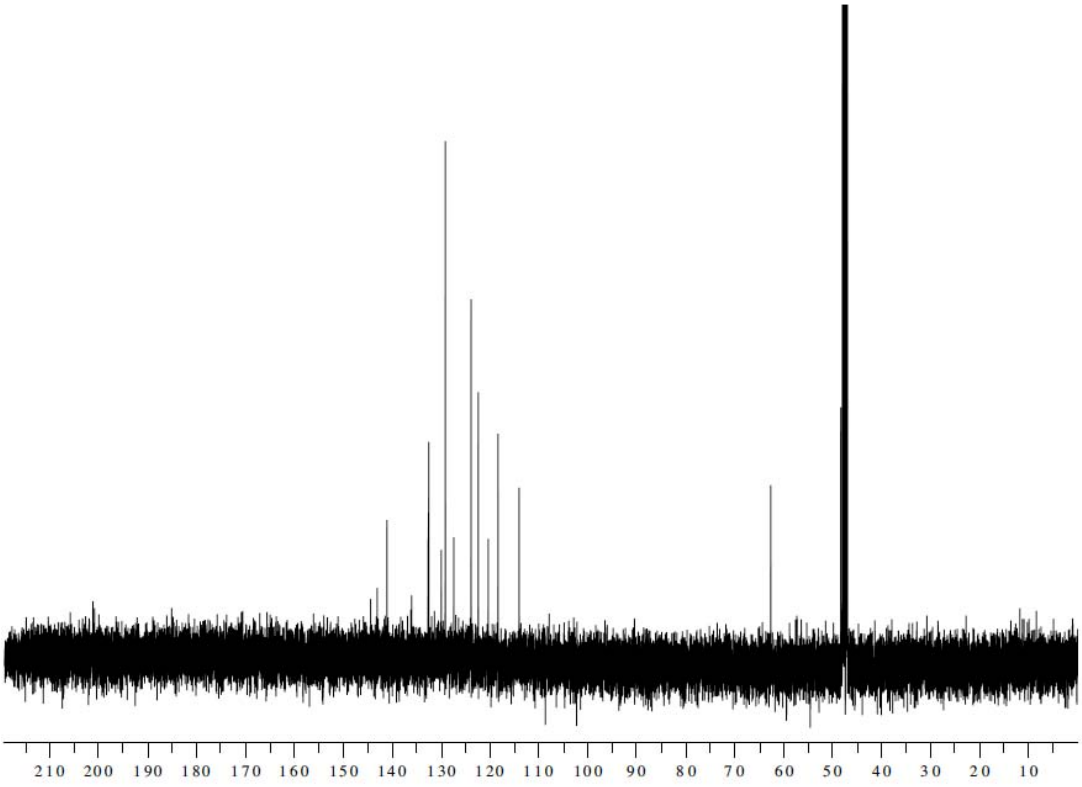
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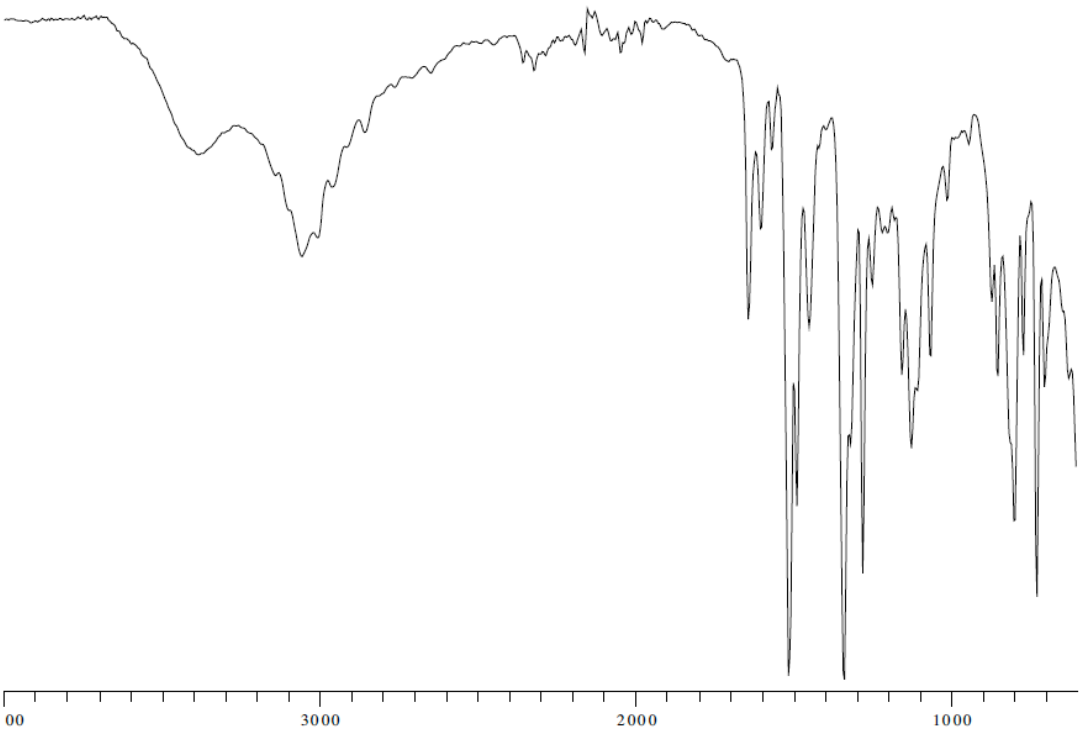
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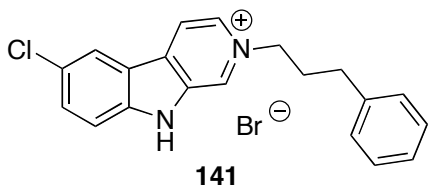
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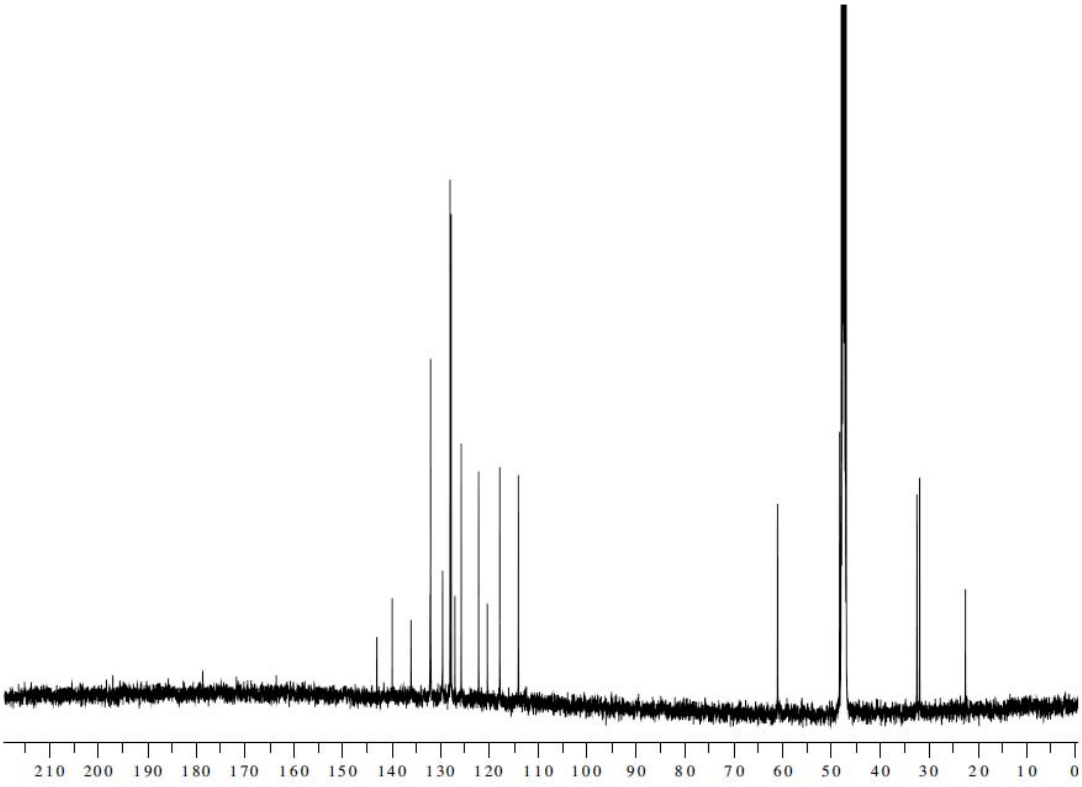
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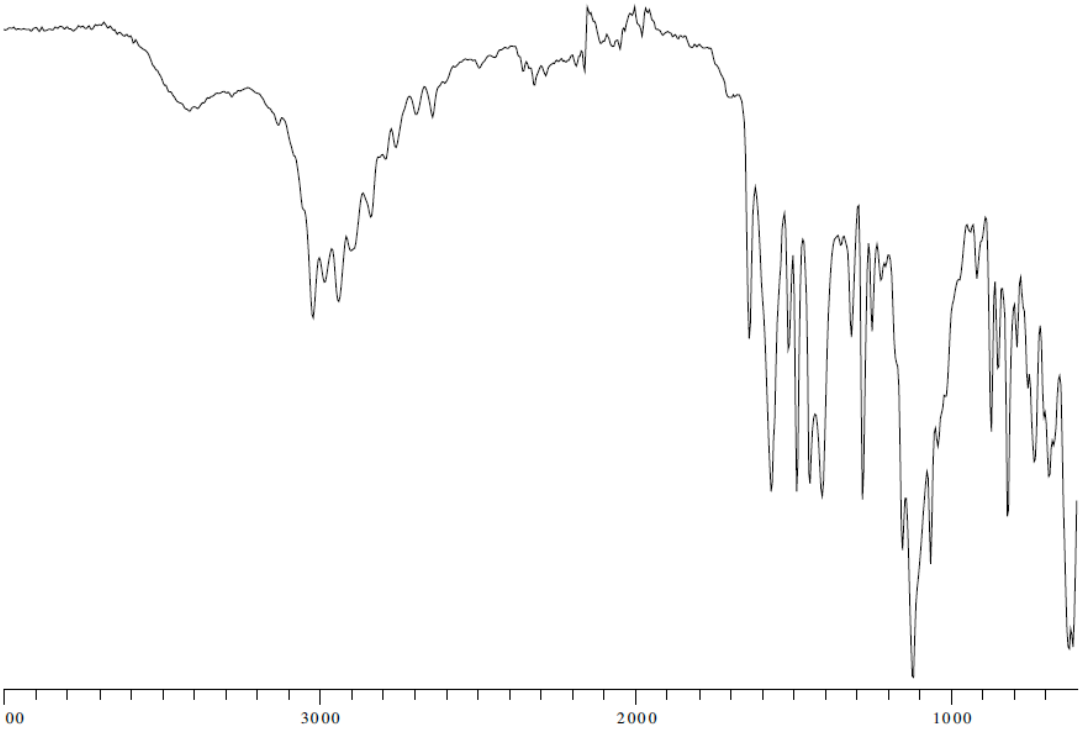

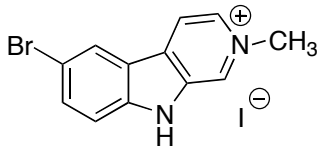

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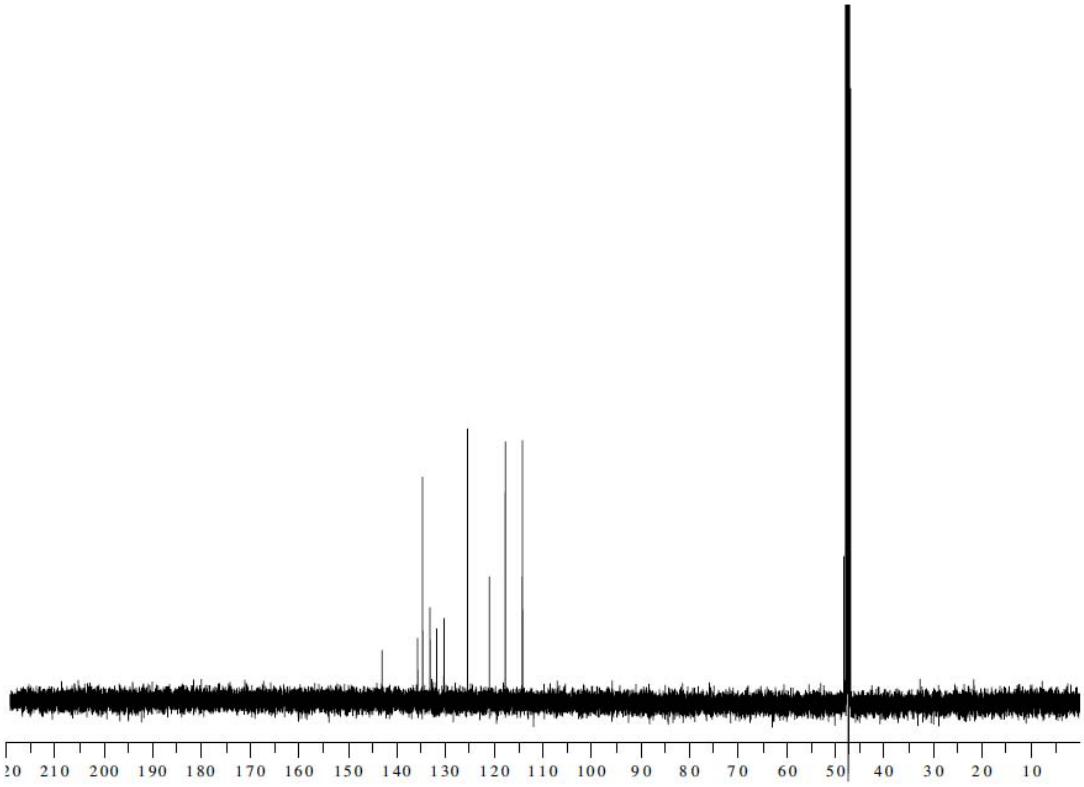
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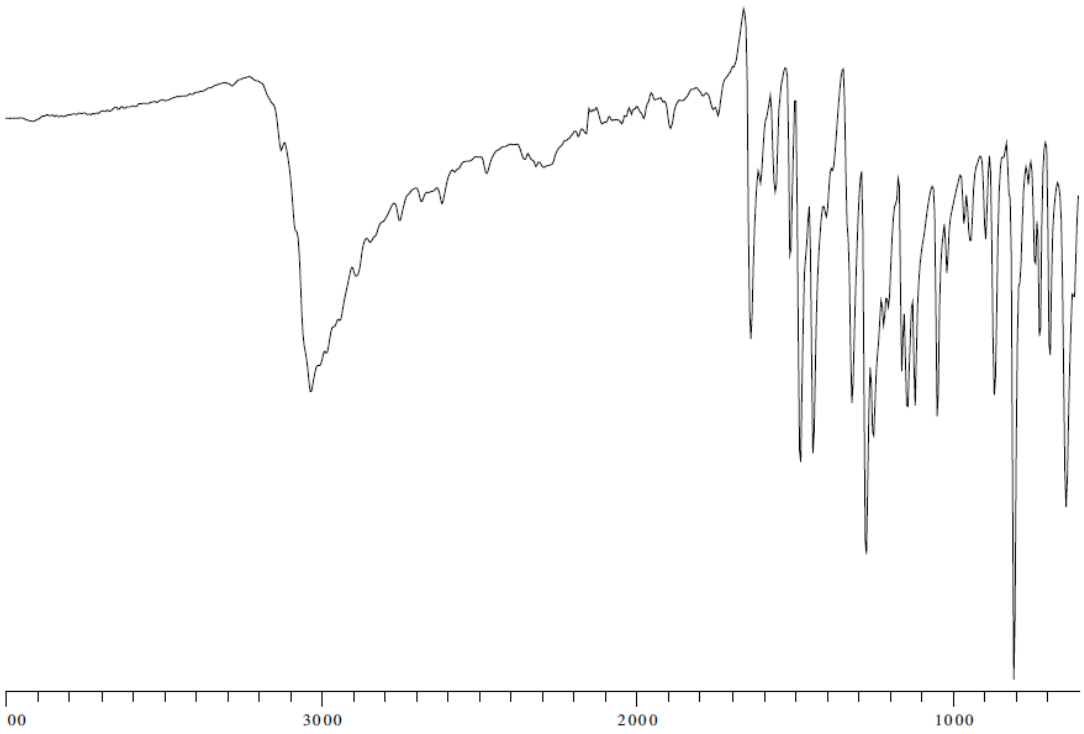
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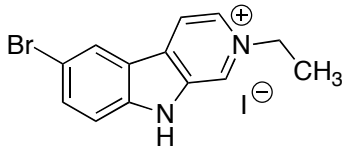
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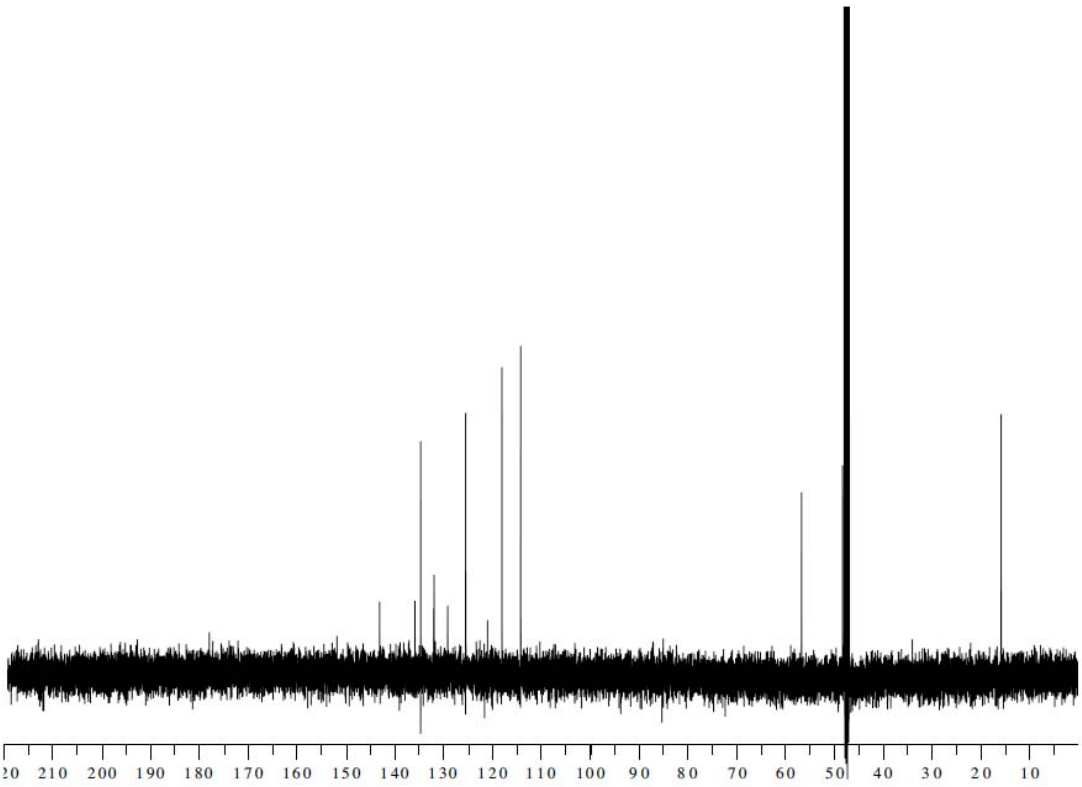
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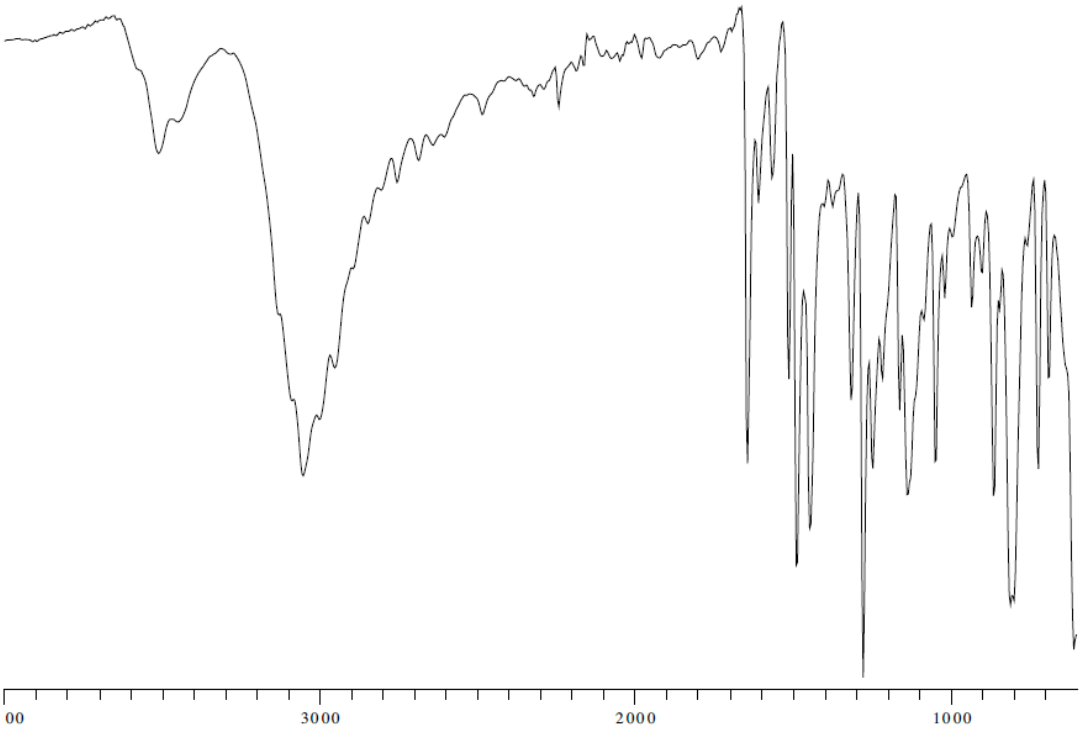

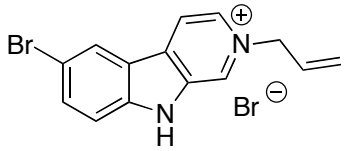

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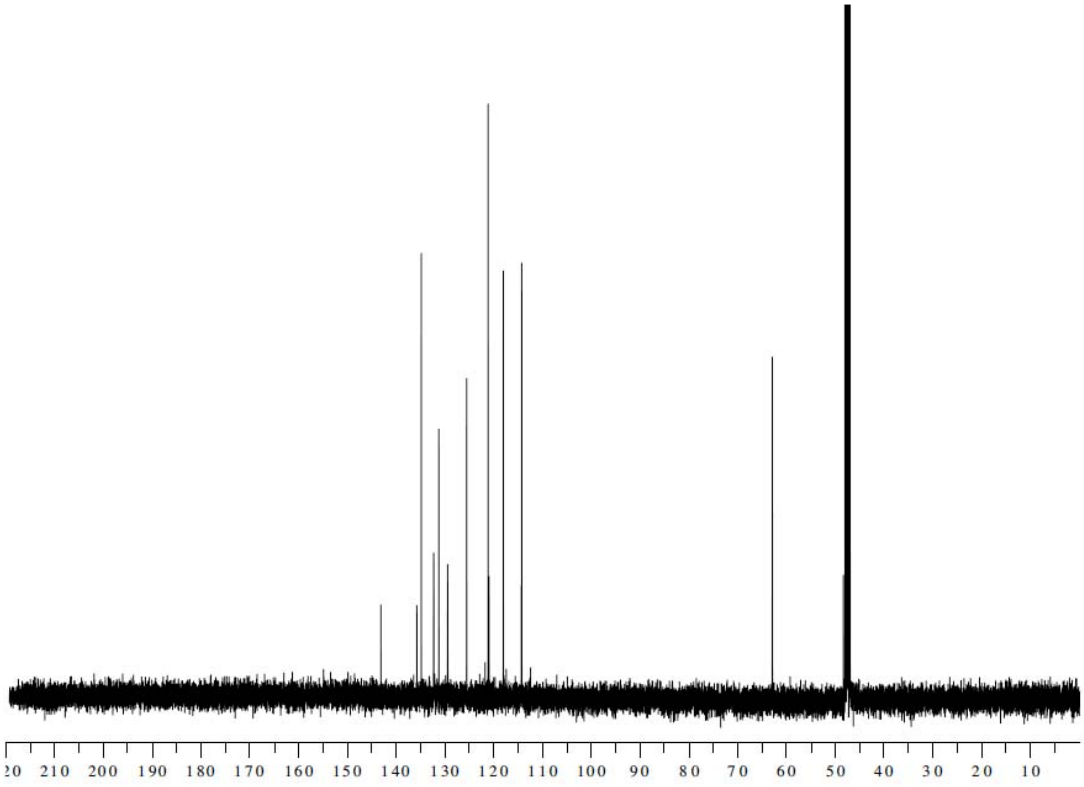
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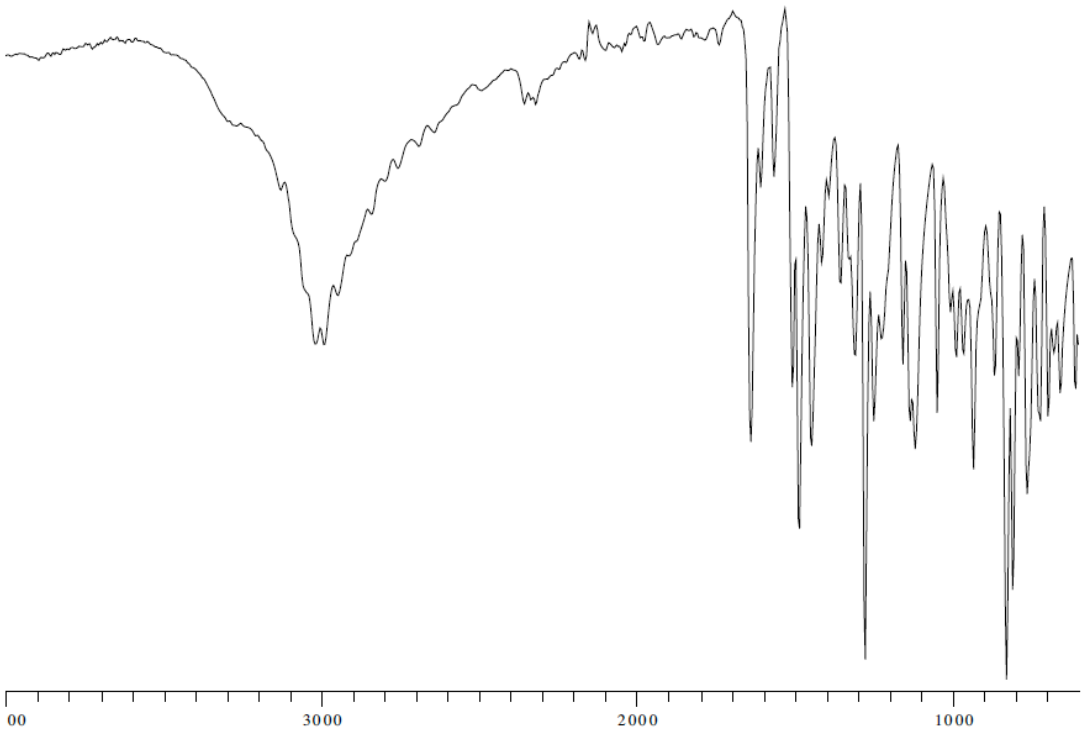
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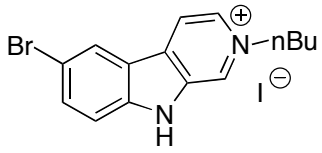
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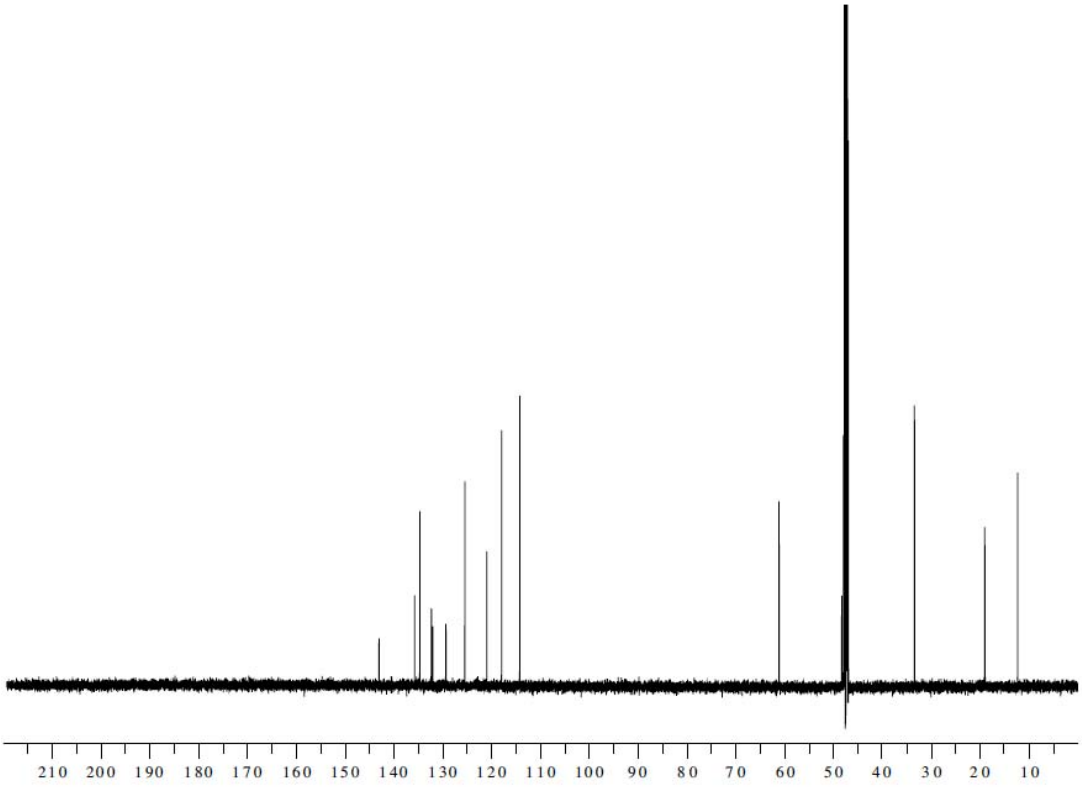
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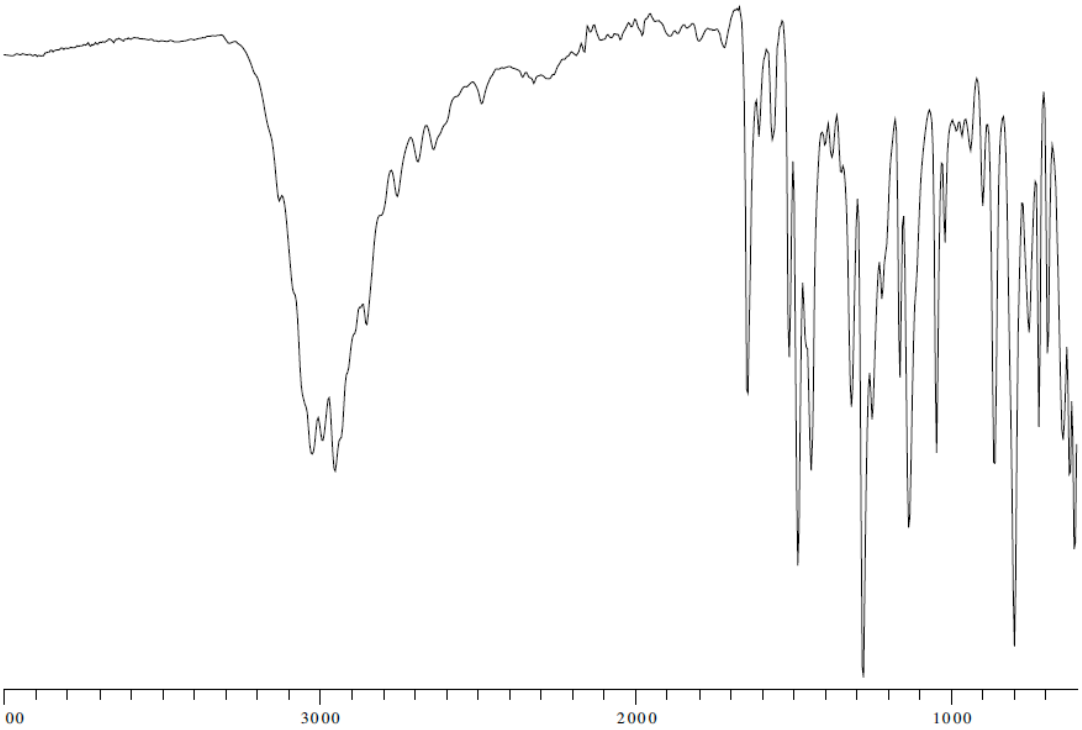

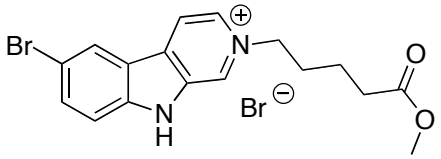

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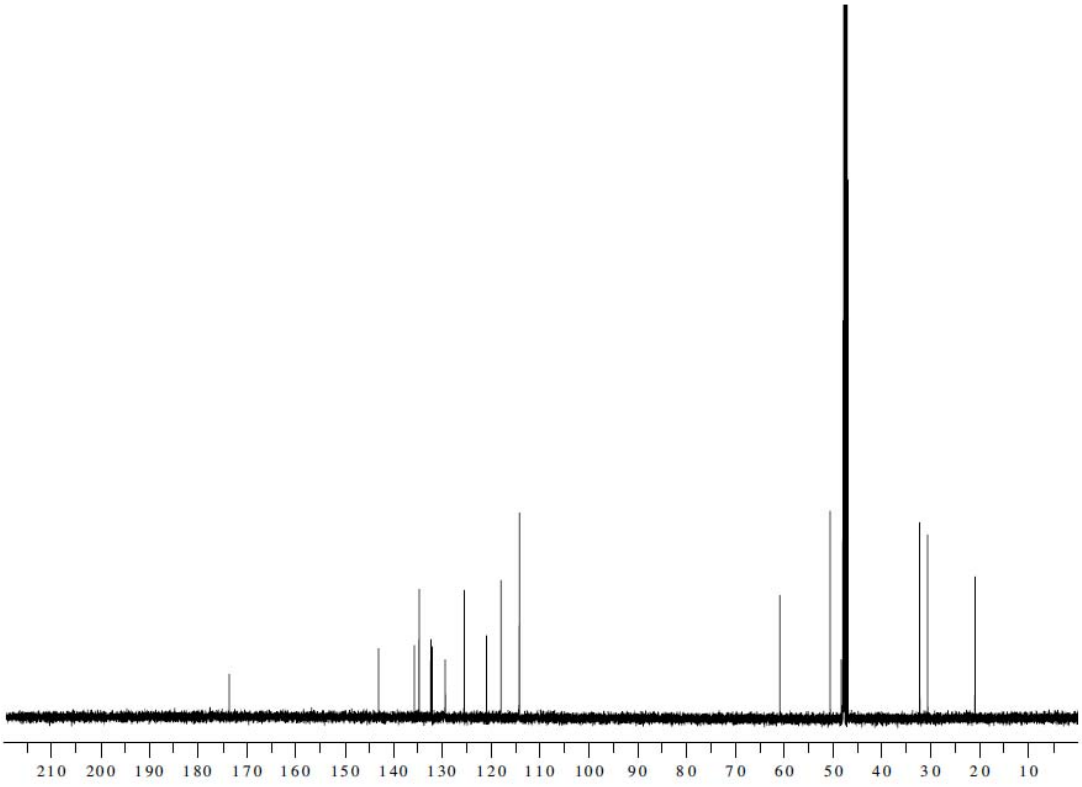
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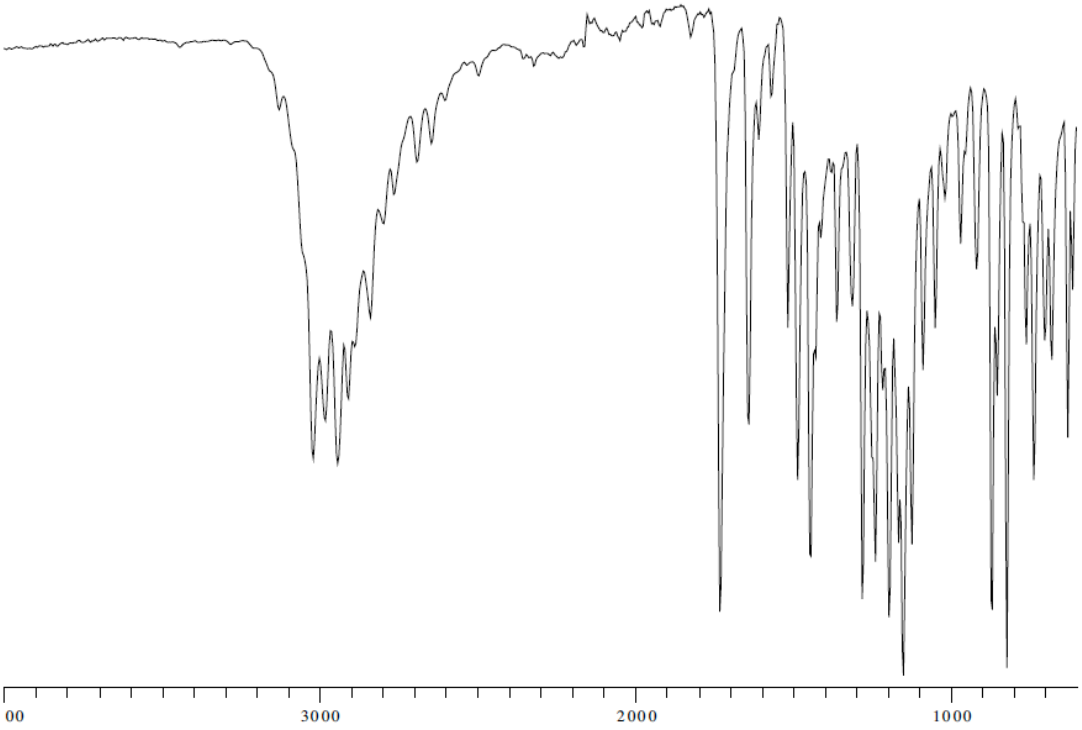
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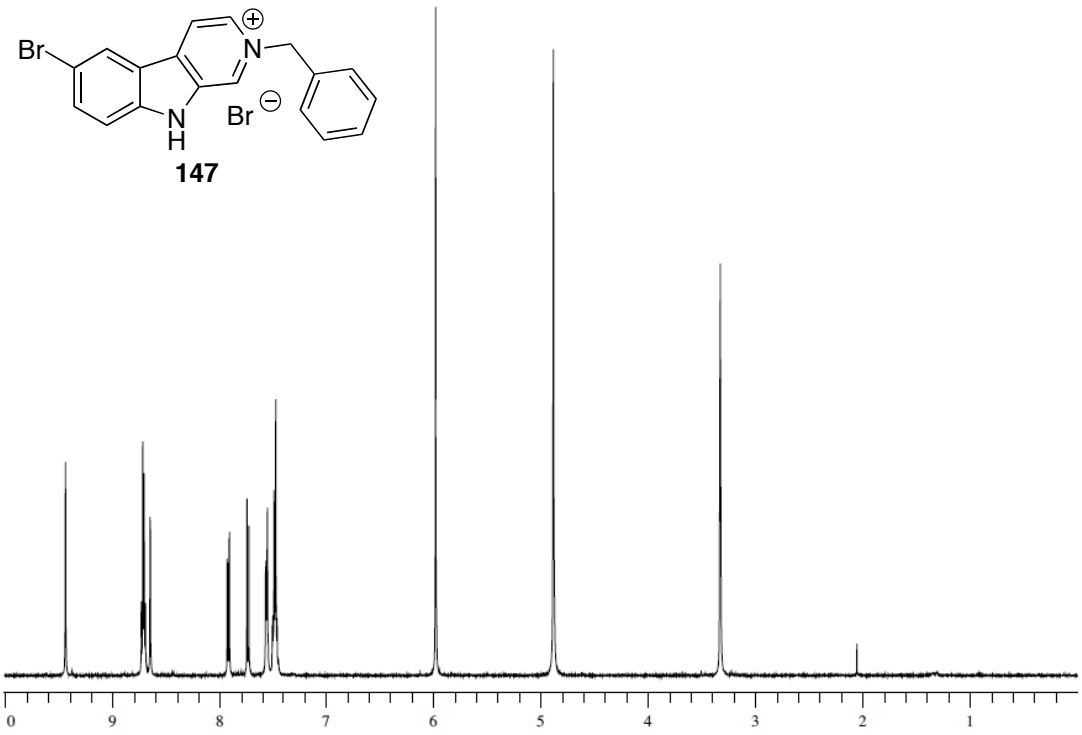
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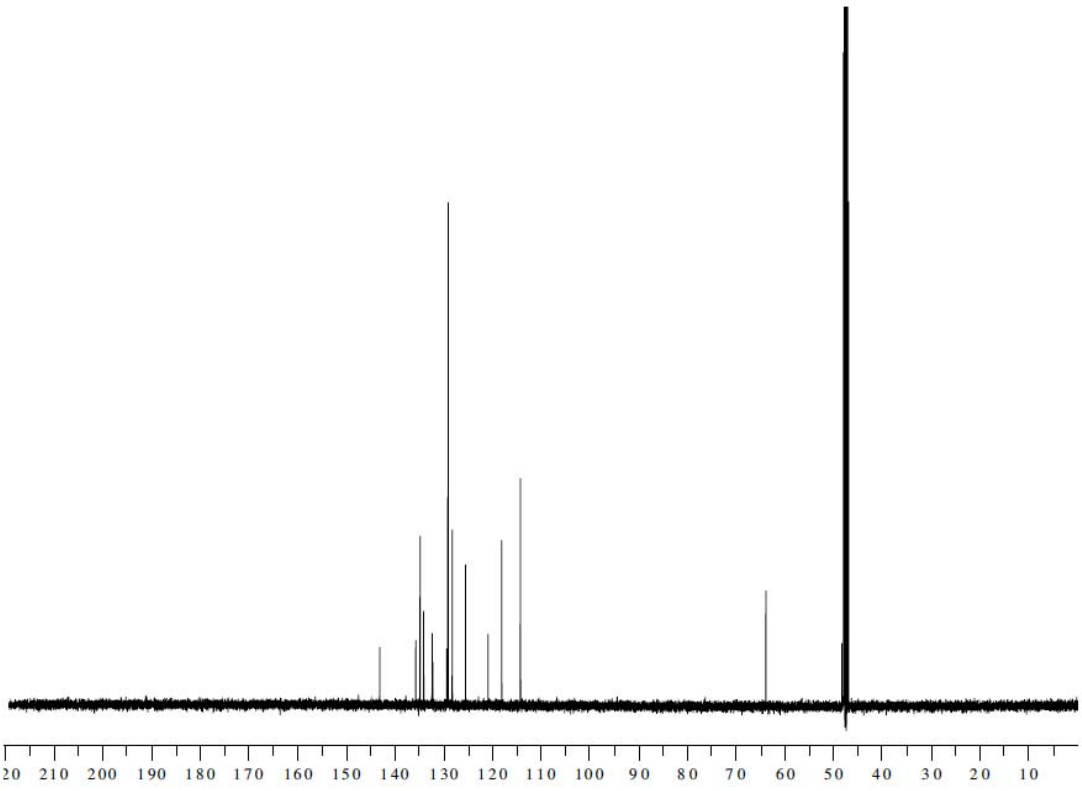
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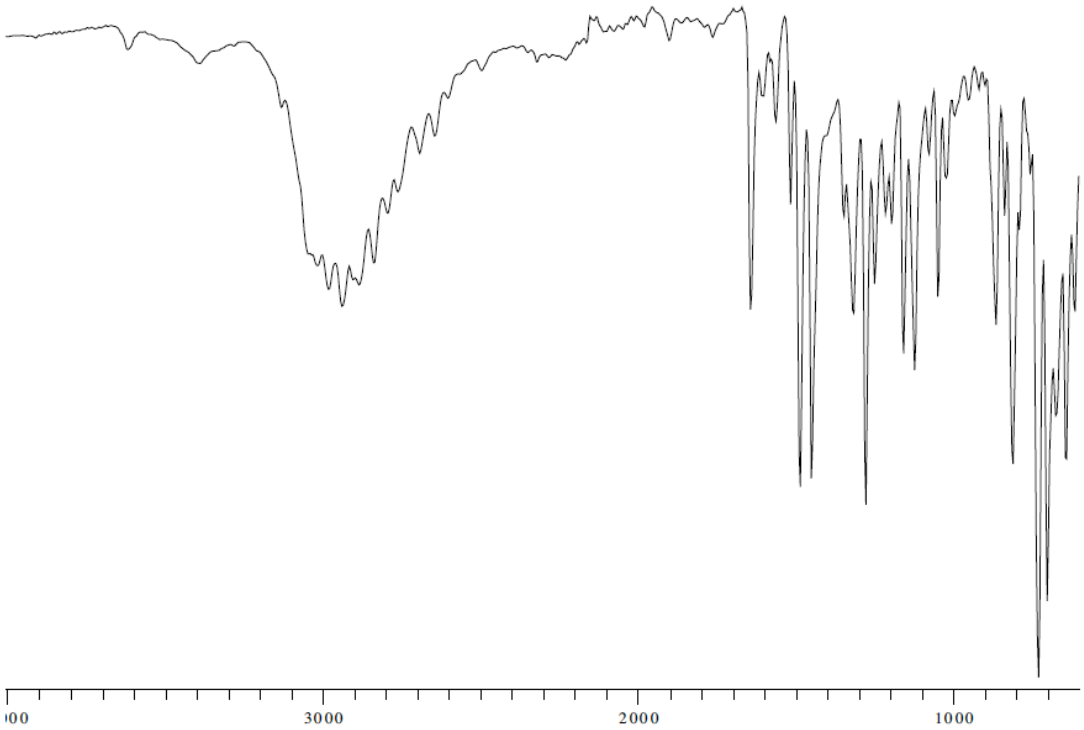

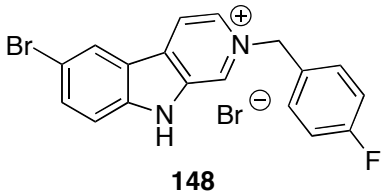

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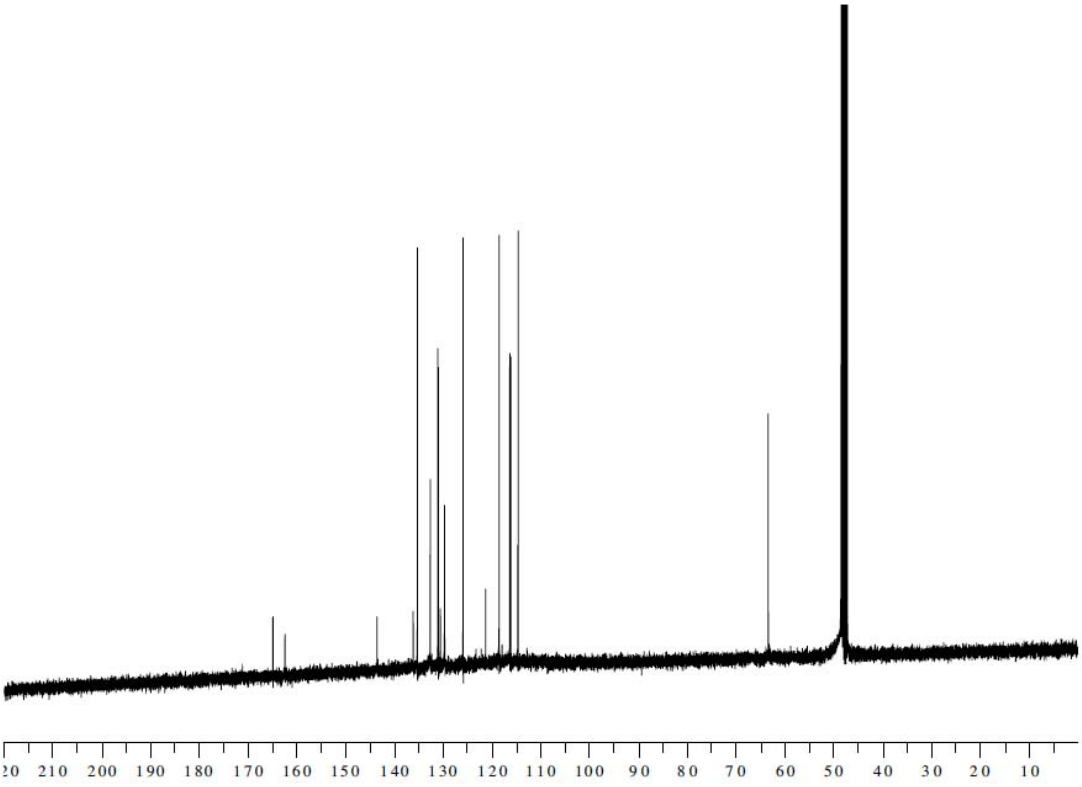
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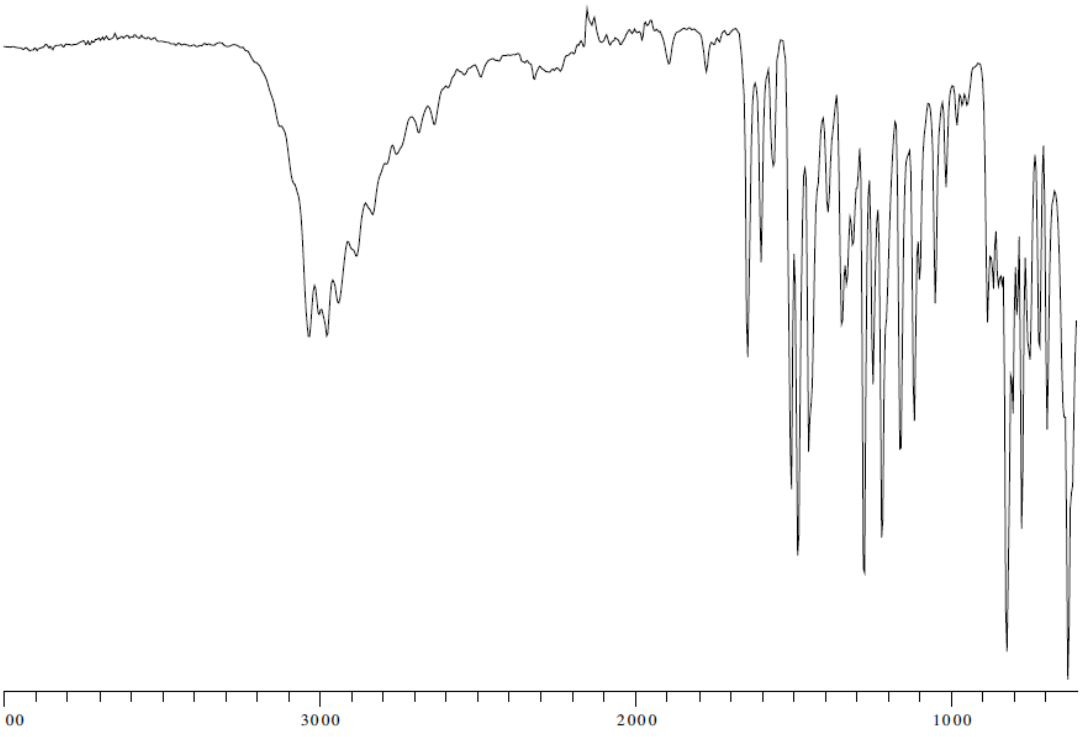
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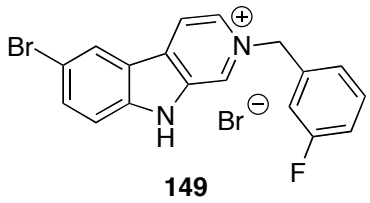
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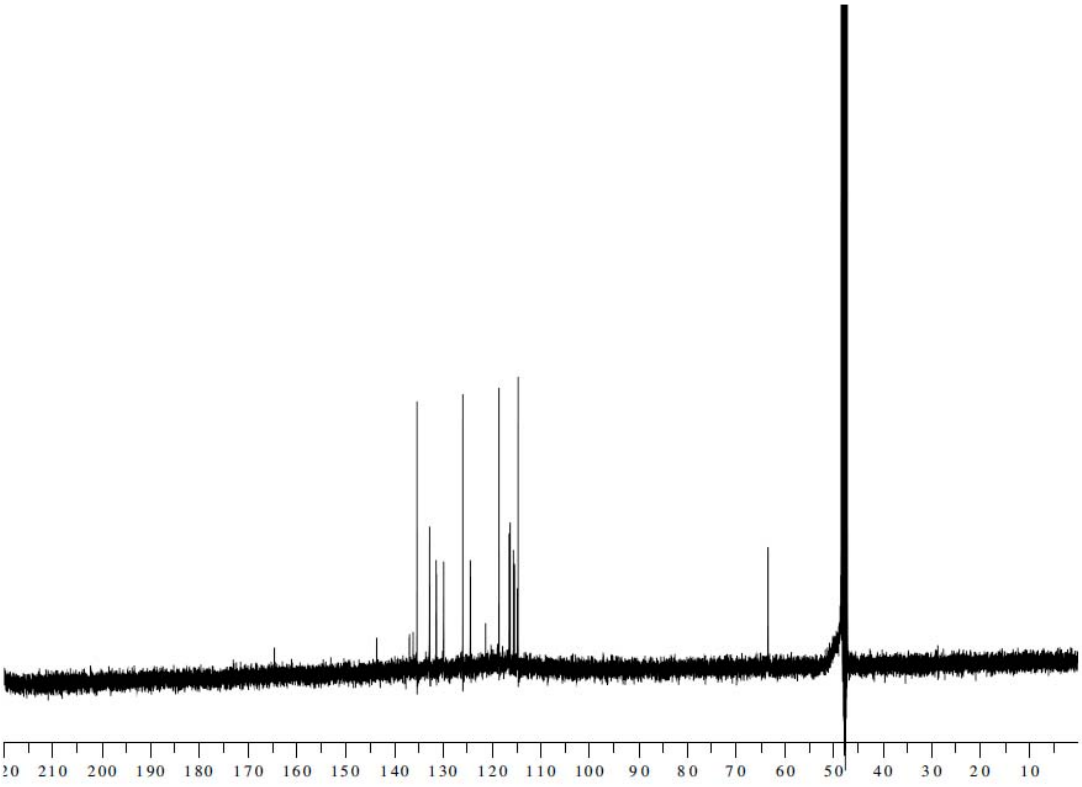
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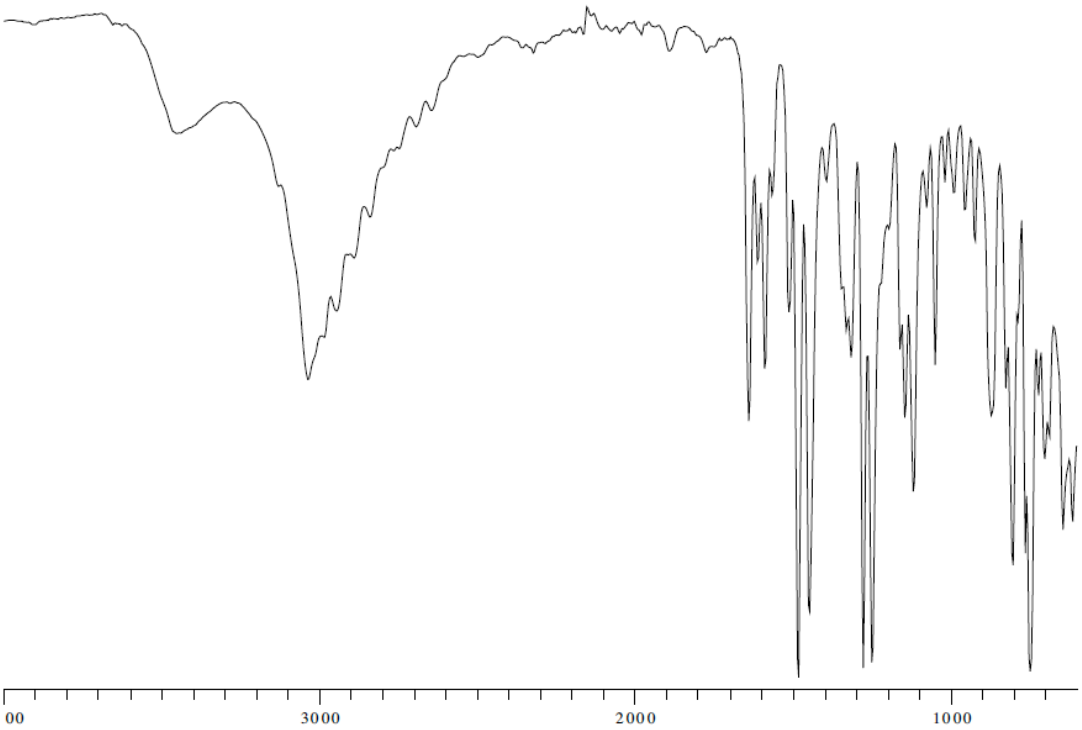

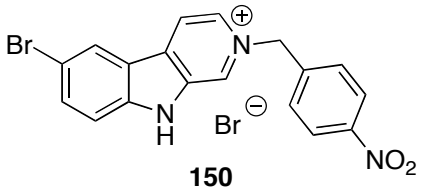

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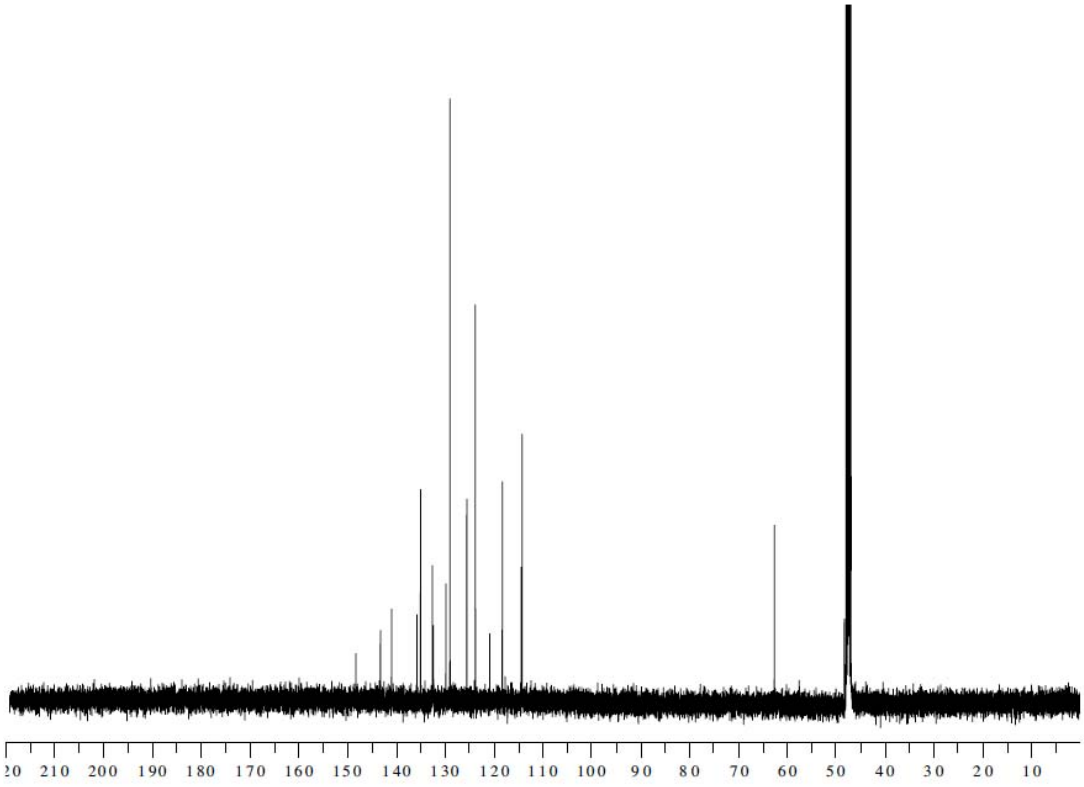
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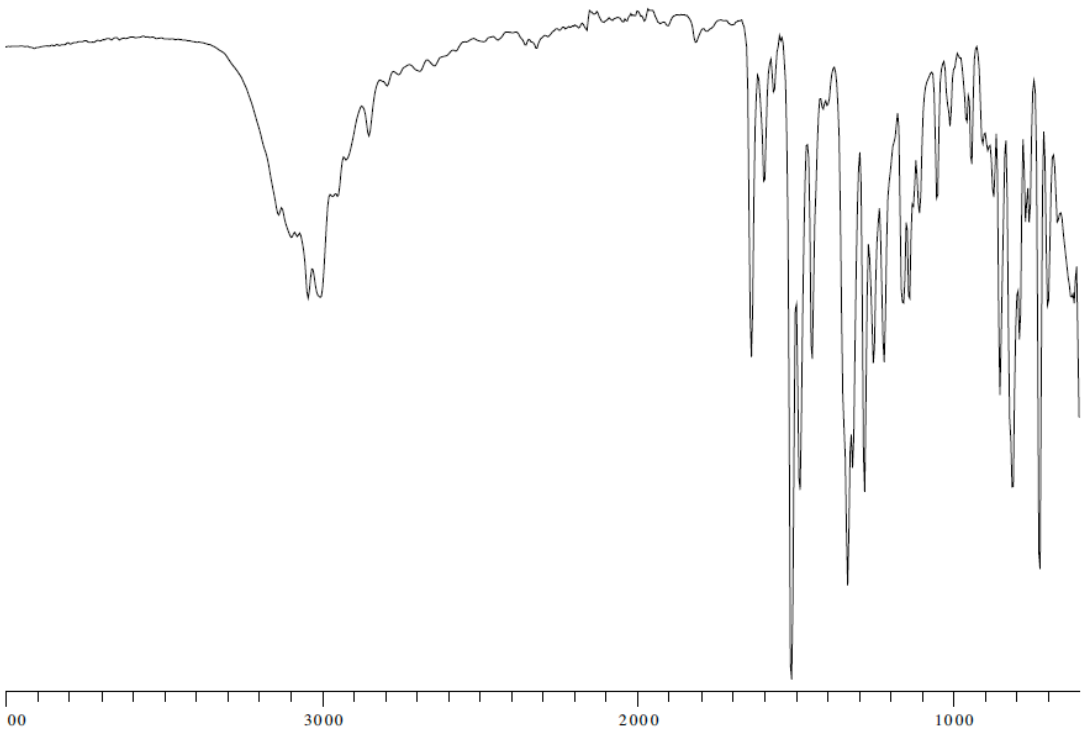
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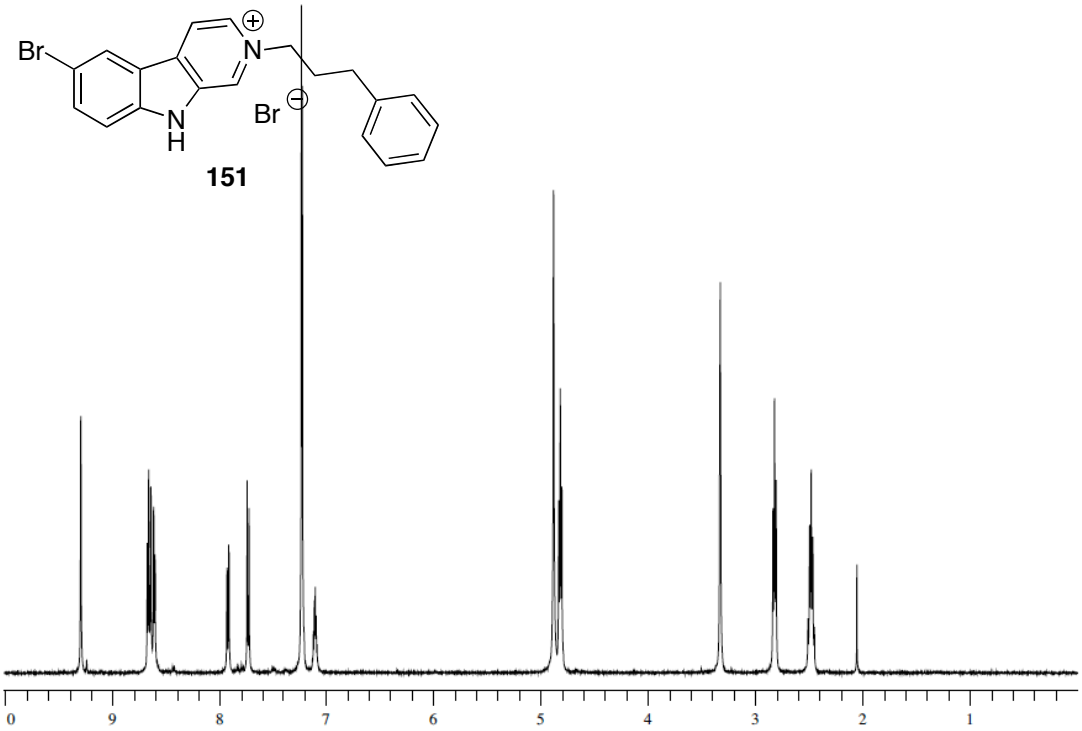
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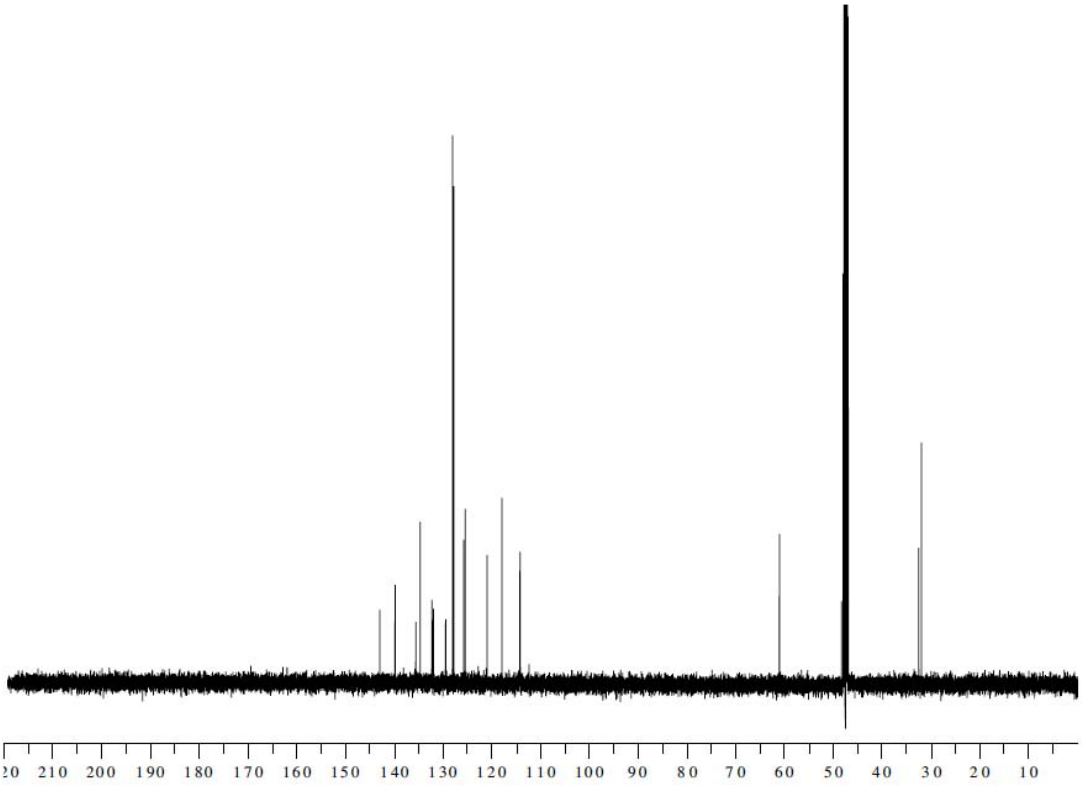
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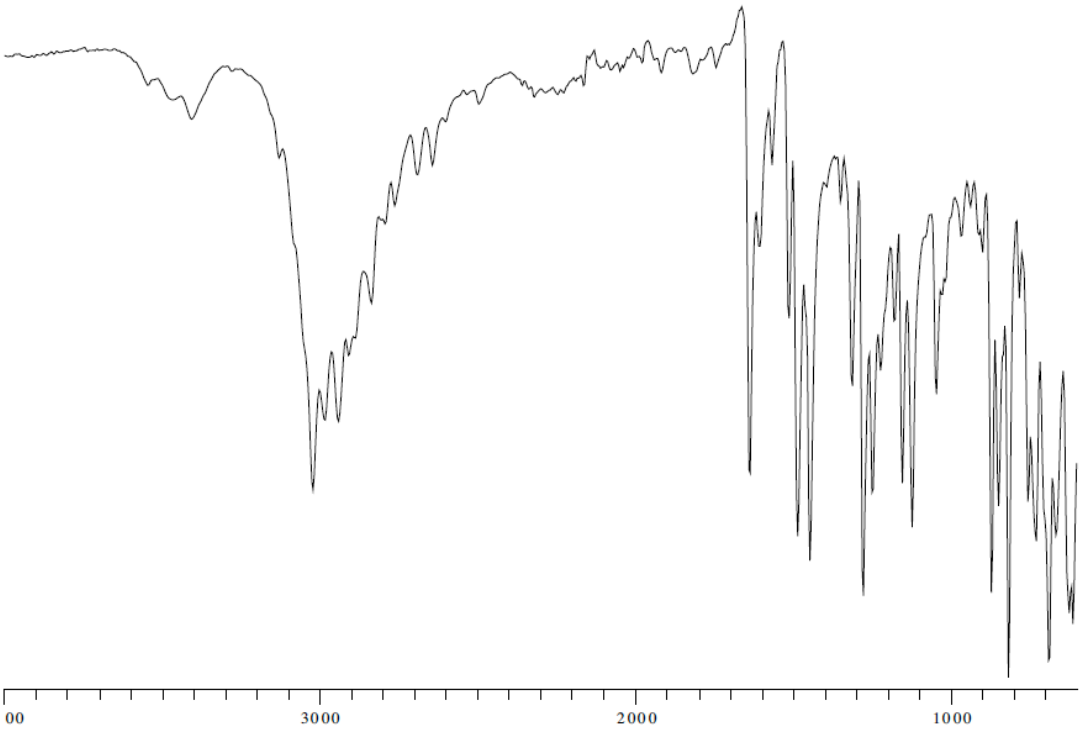

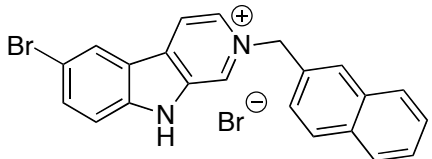

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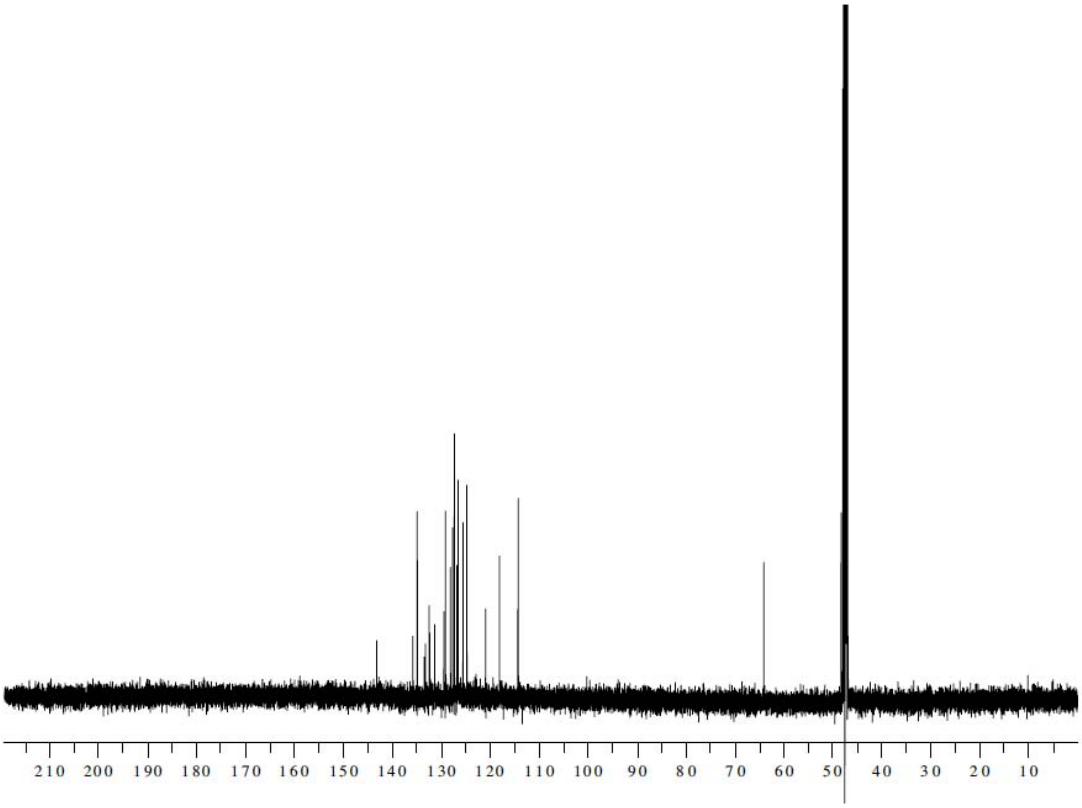
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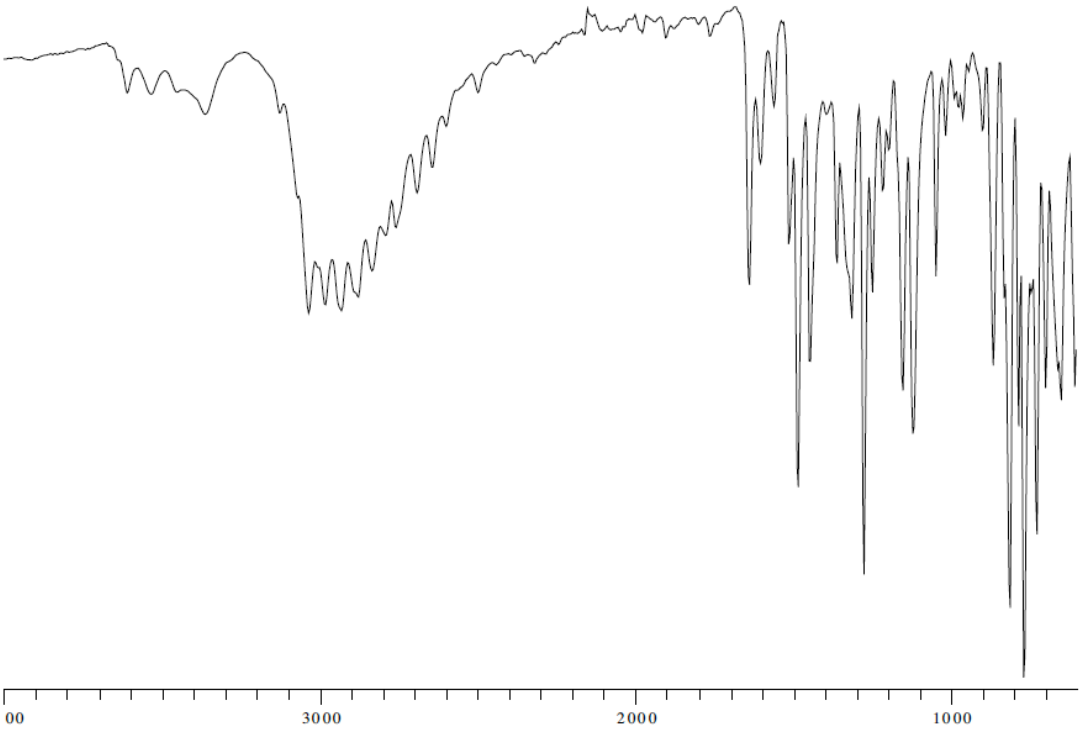
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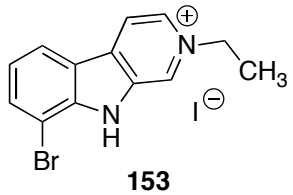
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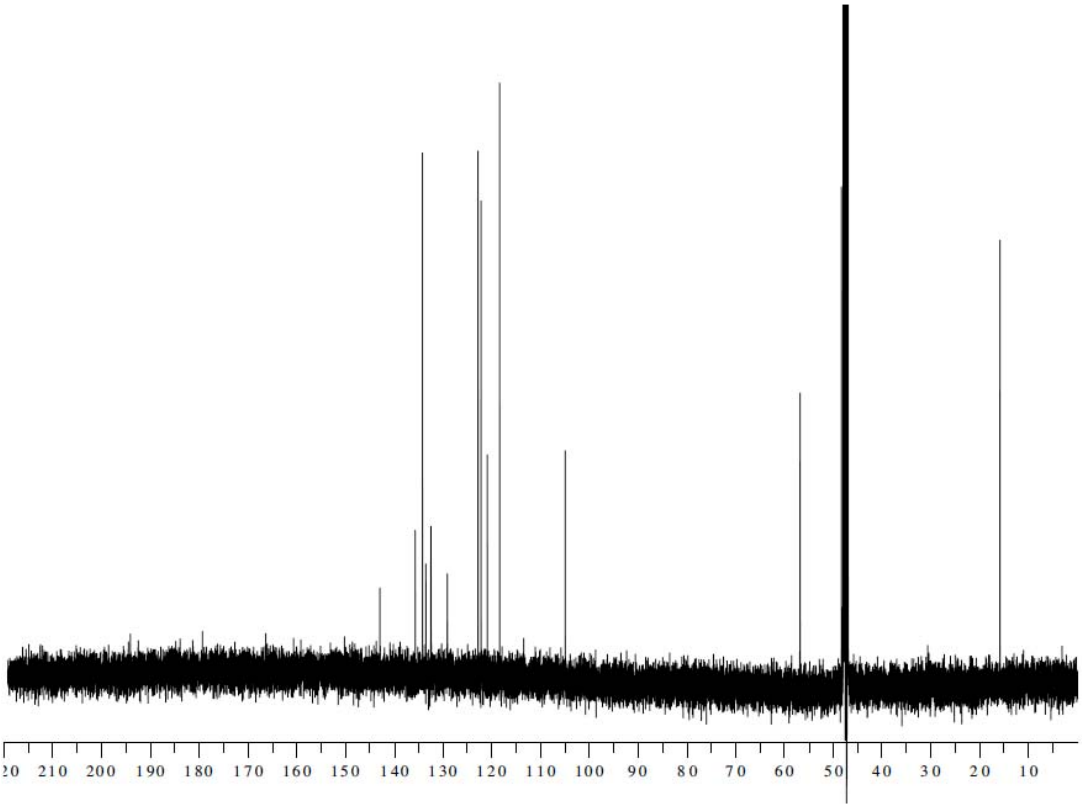
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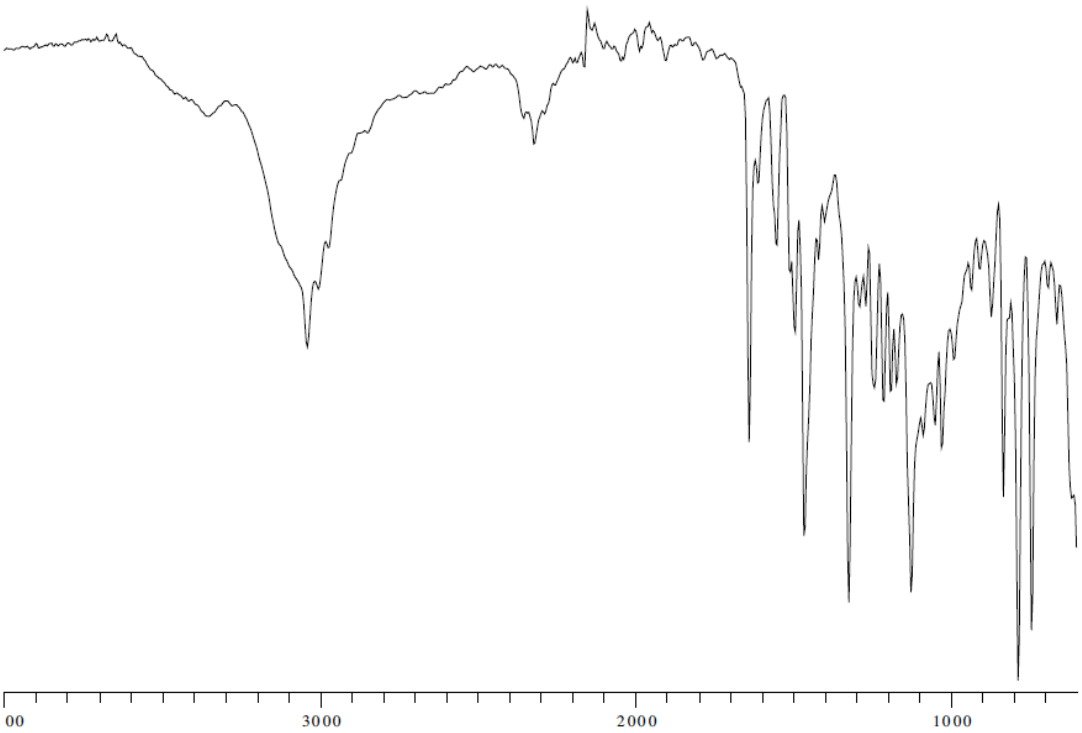

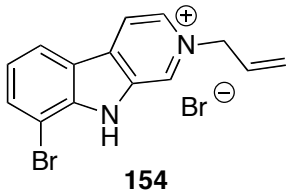

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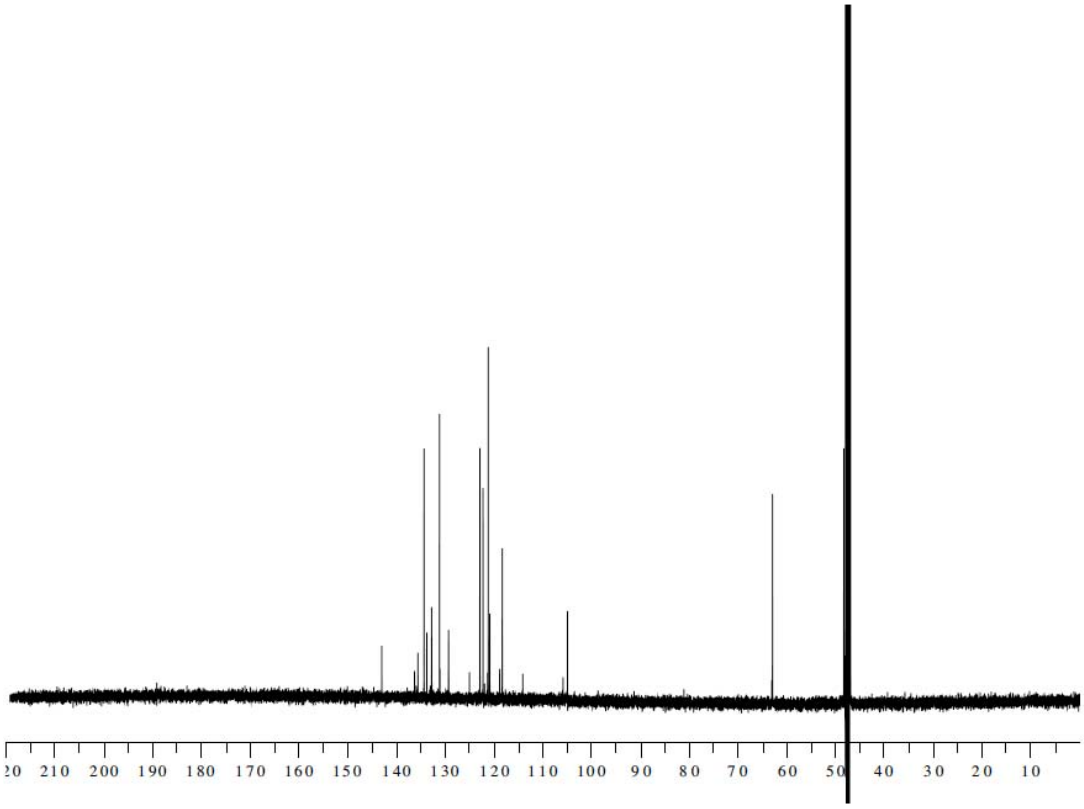
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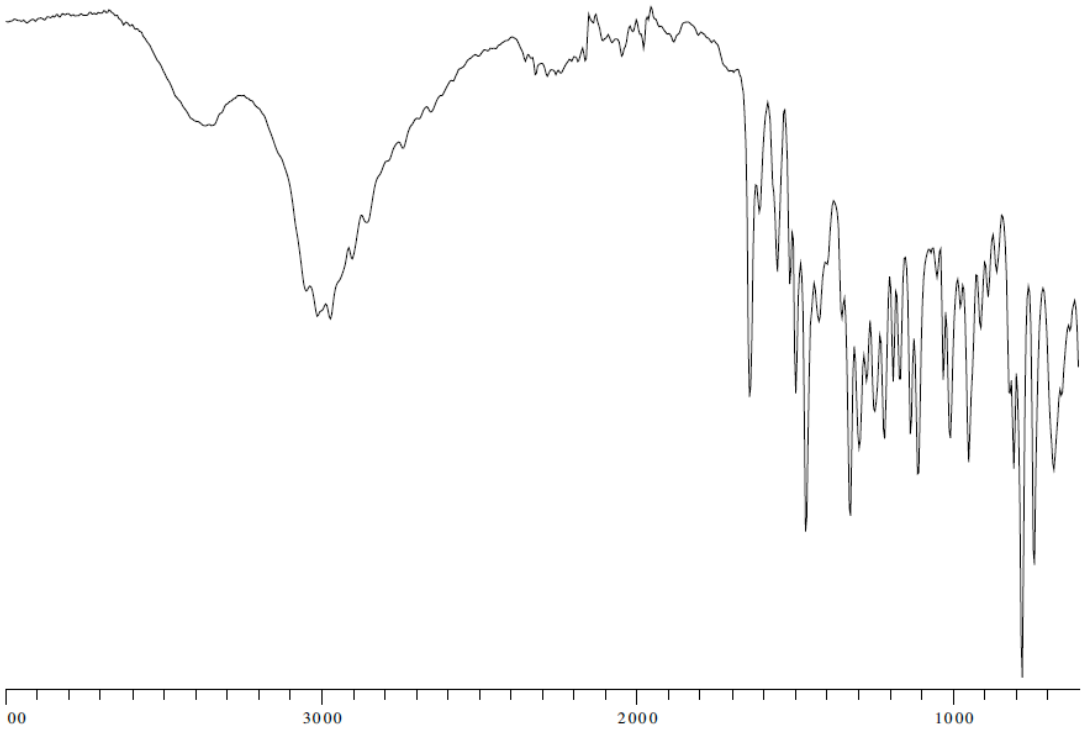
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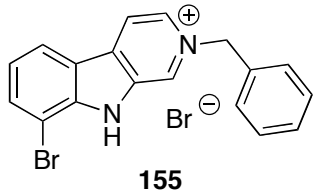
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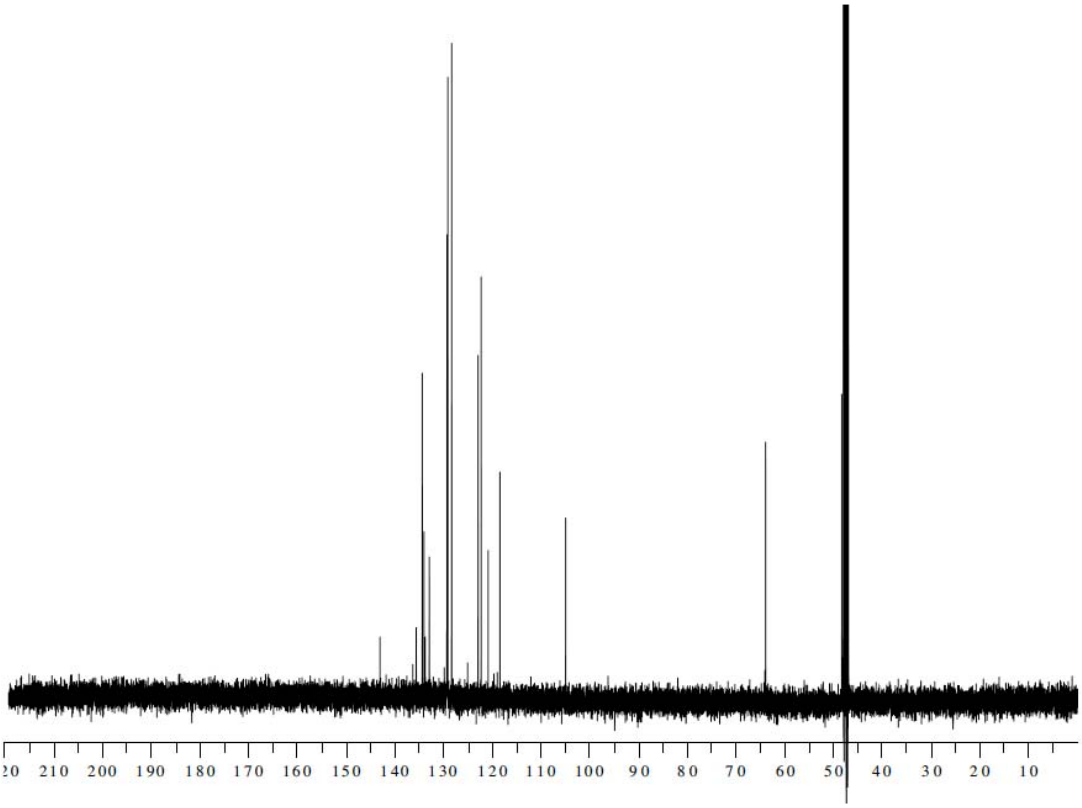
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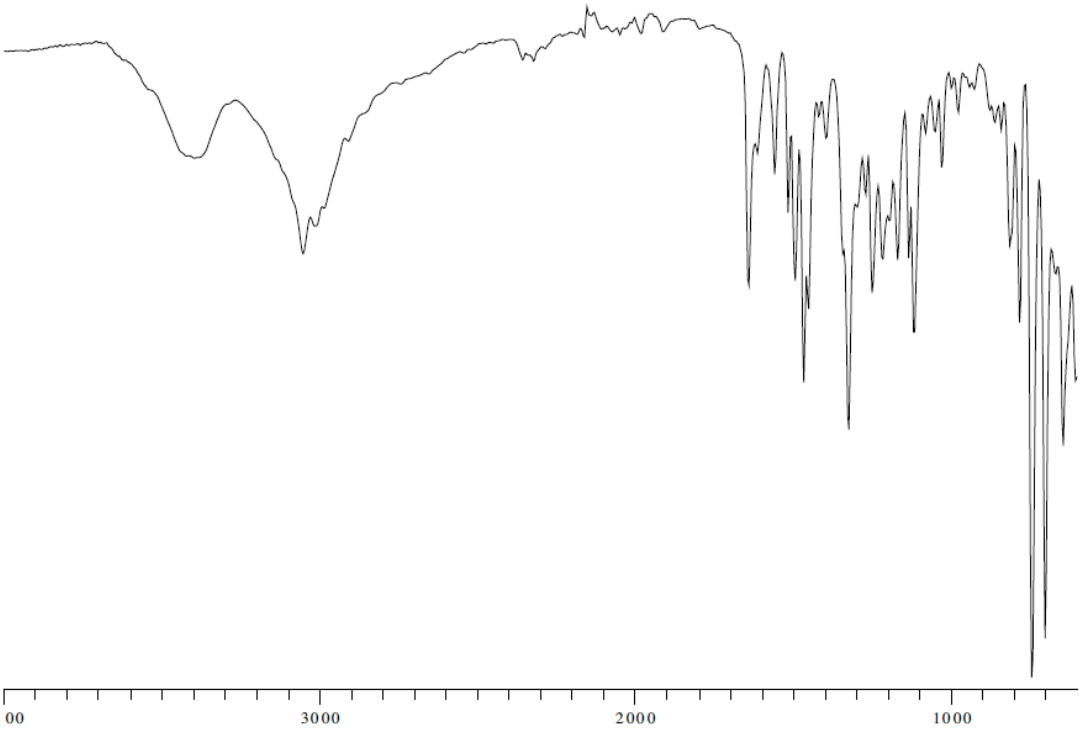

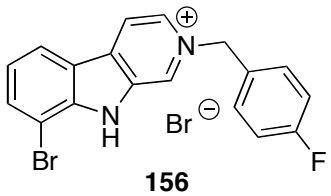

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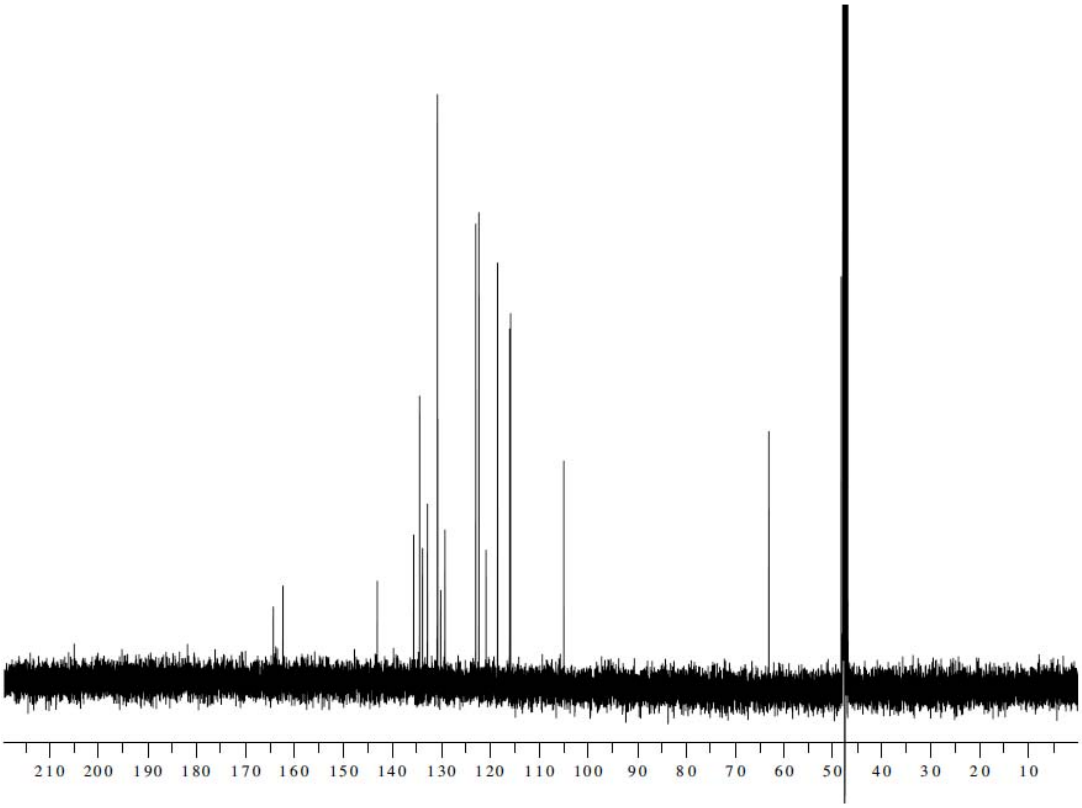
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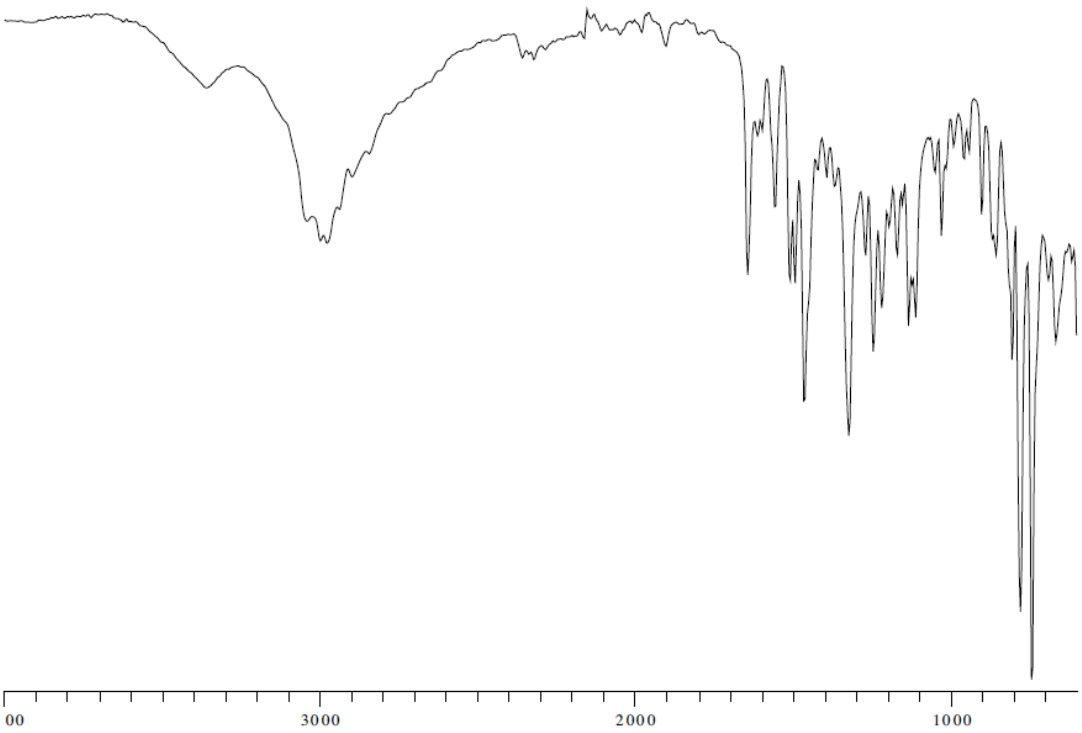
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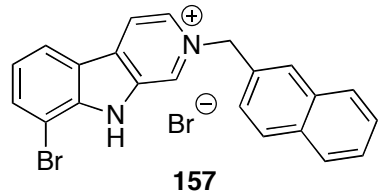
N
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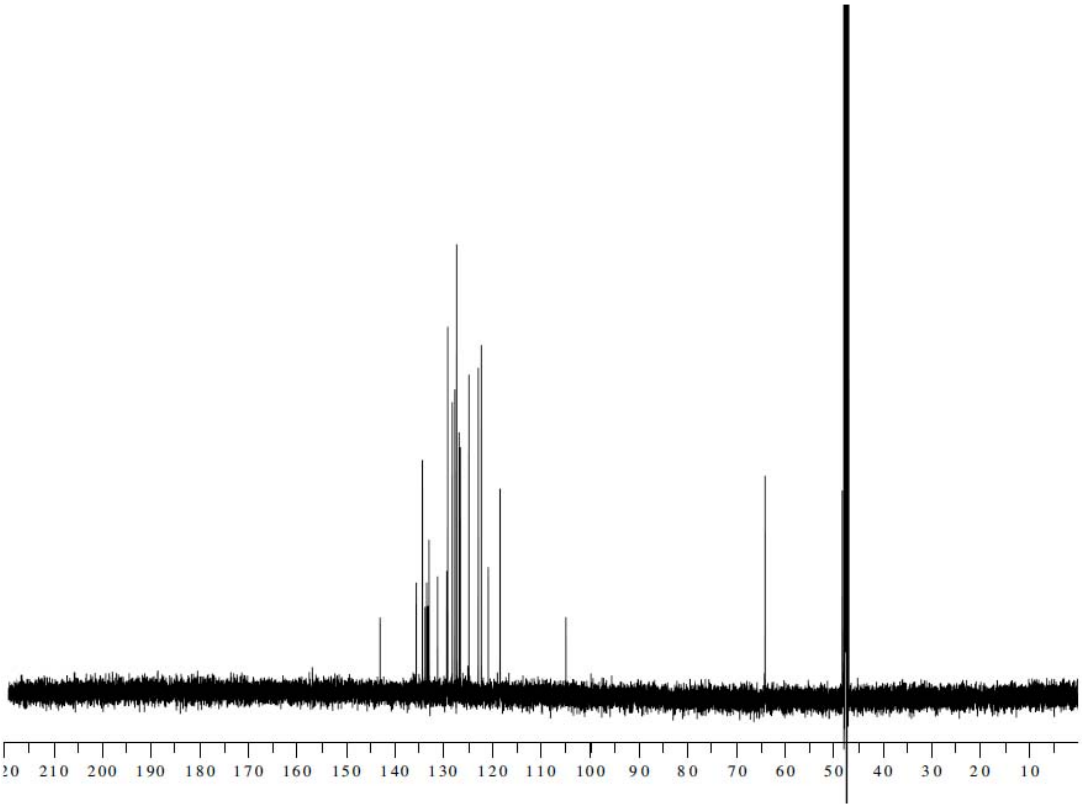
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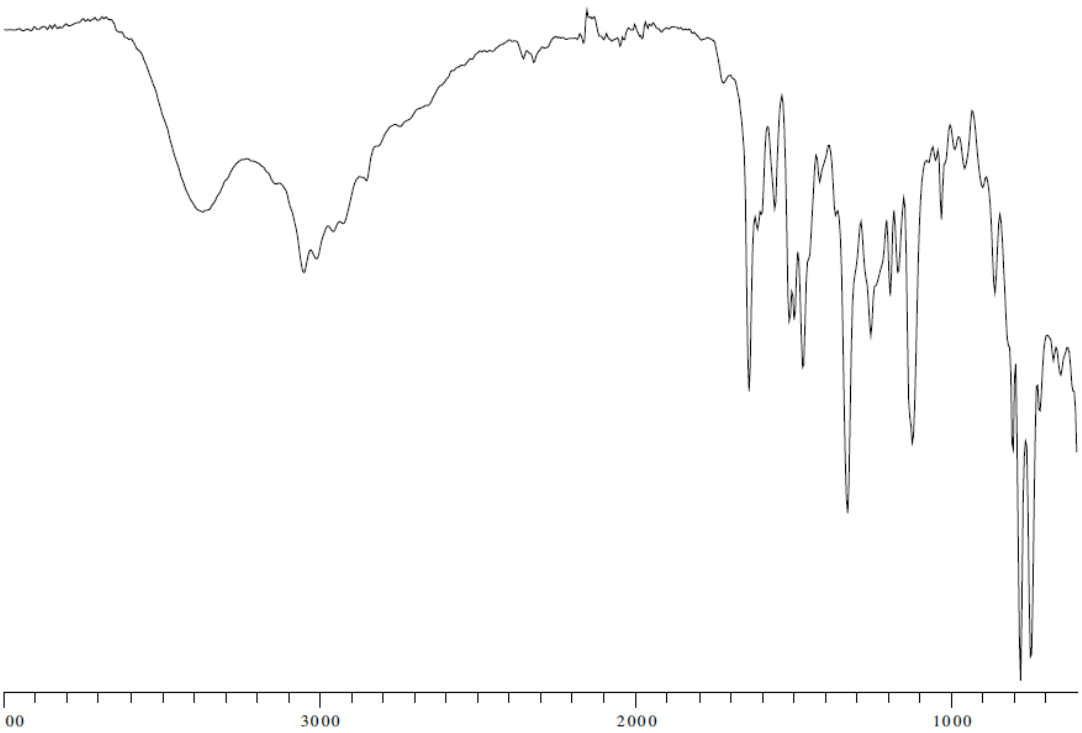

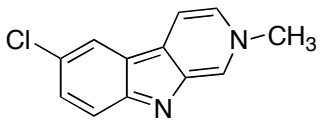

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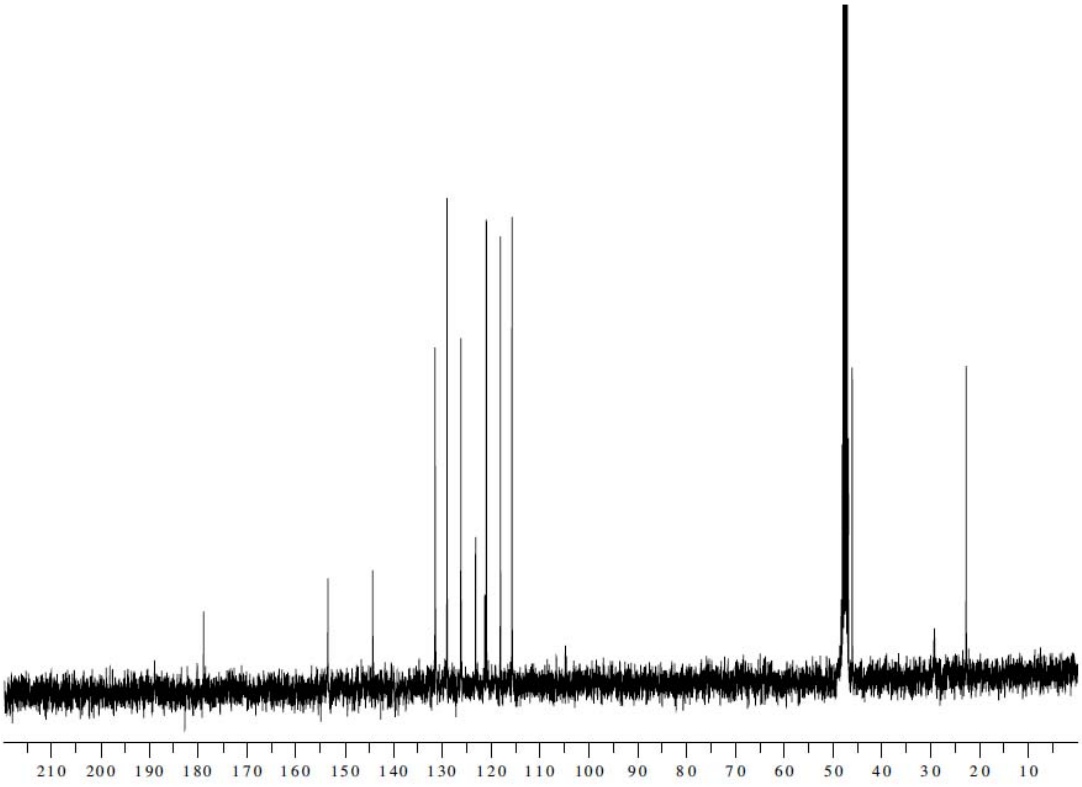
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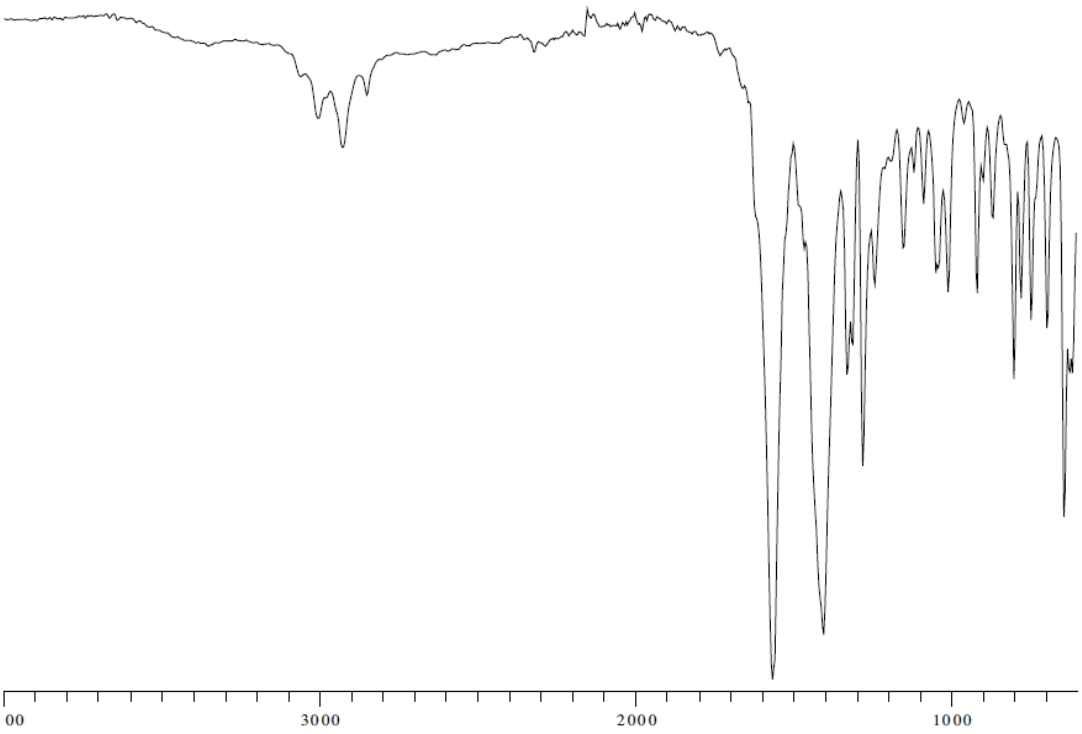
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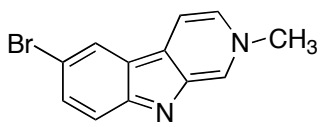
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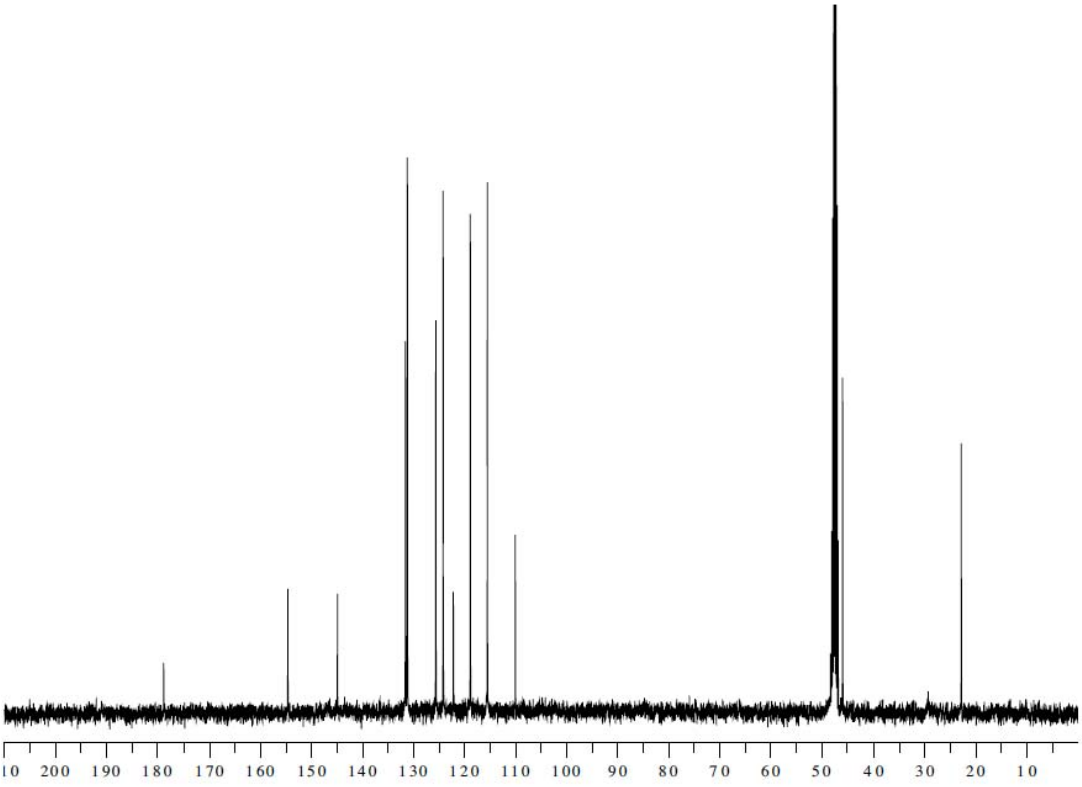
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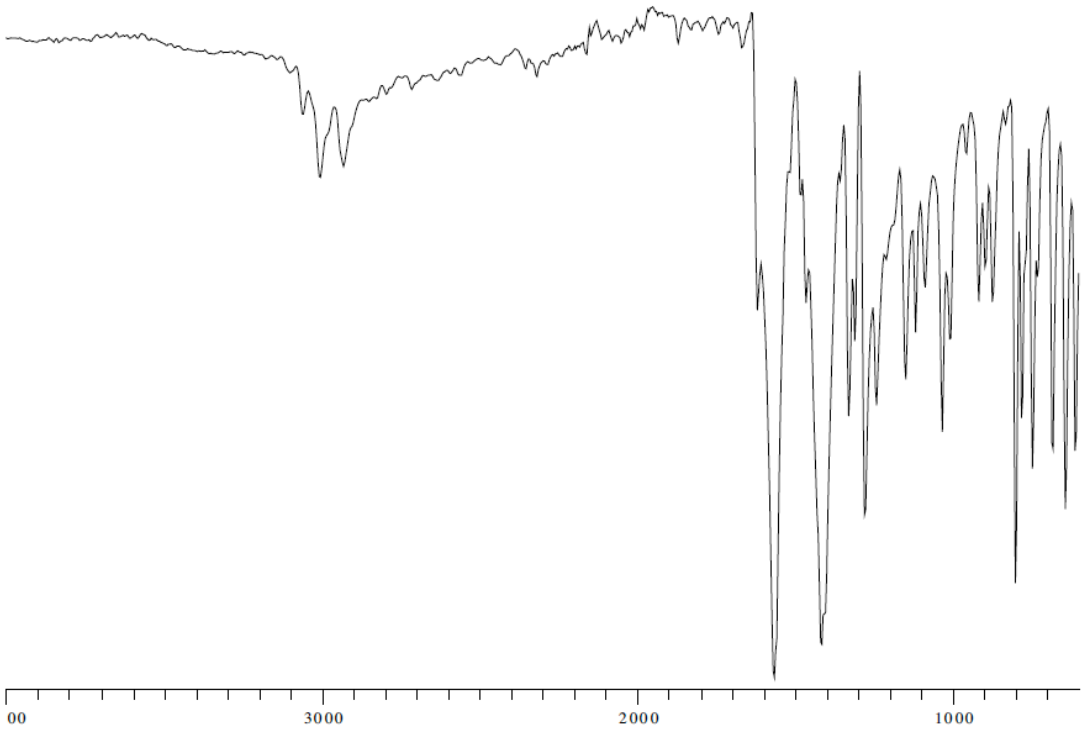
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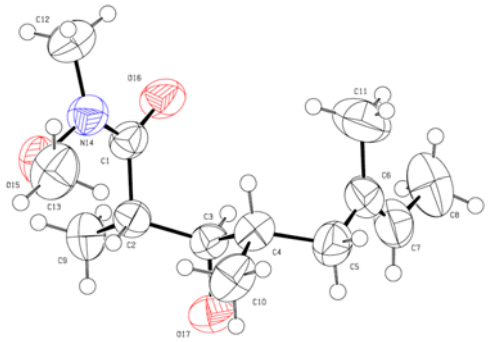
6.6. Crystallographic Data

Crystallographic Data for (2*S*,3*R*,4*S*,*E*)-3-hydroxy-*N*-methoxy-*N*,2,4,6-tetramethyloct-6-enamide (42) (CCDC674800)



Abstract

We present the crystal and molecular structure of (2*S*,3*R*,4*S*,*E*)-3-hydroxy-*N*-methoxy-*N*,2,4,6-tetramethyloct-6-enamide (**42**)

Comment[253]

The study of the titled structure was undertaken to establish its three dimensional structure. Geometries are tabulated below. All diagrams and calculations were performed using maXus (Bruker Nonius, Delft & MacScience, Japan).

Experimental

Crystal data

[253] S. Mackay, C. J. Gilmore, C. Edwards, N. Stewart, K. Shankland, *maXus Computer Program for the Solution and Refinement of Crystal Structures* **1999**. Bruker Nonius, The Netherlands, MacScience, Japan & The University of Glasgow; C. K. Johnson, *ORTEP*--II. *A Fortran Thermal--Ellipsoid Plot Program* **1976**. Report ORNL-5138. Oak Ridge National Laboratory, Oak Ridge, Tennessee, USA; Z. Otwinowski, W. Minor, *In Methods in Enzymology* **1997**, *276*, edited by C. W. Carter, Jr. & R. M. Sweet pp. 307--326, New York:Academic Press; A. Altomare, M. C. Burla, M. Camalli, G. L. Cascarano, C. Giacovazzo, A. Guagliardi, A. G. G. Moliterni, R. Spagna, *J. Appl. Cryst.* **1999,** *32*, 115--119; G. M. Sheldrick, *SHELXL*97. *Program for the Refinement of Crystal Structures* **1997**, University of Göttingen, Germany.

$C_{13}H_{25}NO_3$
$C_{13}H_{25}NO_3$
M_r = 243.347
Monoclinic $P2_1$
a = 8.5218 (3)Å
b = 9.5856 (4)Å
c = 9.8281 (4)Å
α = 90.00°
β = 111.648 (2)°
γ = 90.00°

V = 746.20 (5)Å^3
Z = 2

D_x = 1.083 Mg m^{-3}
Density measured by: not measured
fine-focus sealed tube
Mo *K*α radiation λ = 0.71073
Cell parameters from 4731 refl.
θ = 0.998—27.485 °
μ = 0.076 mm^{-1}
T = 298 K
Cube
0.7 x 0.5 x 0.24 mm
Colourless
Crystal source: Seeberger laboratory

Data collection

KappaCCD CCD diffractometer
Absorption correction: none
3267 measured reflections
3253 independent reflections
2636 observed reflections
Criterion: >2sigma(I)

R_{int} = 0.031
θ_{max} = 27.50 °
h = -11 →11
k = -12 →12
l = -12 →12

Refinement

Refinement on F^2
fullmatrix least squares refinement
R(all) = 0.0658
R(gt) = 0.0513
wR(ref) = 0.1627
wR(gt)= 0.1464
S(ref) = 1.089
3253 reflections
154 parameters
1 restraints
H positions constr

Calculated weights $1/[\sigma^2(I_o)+(I_o+I_c)^2/900]$
Δ/σ_{max} = 0.001
$\Delta\rho_{max}$ = 0.118eÅ^3
$\Delta\rho_{min}$ = -0.133eÅ^3
Extinction correction: none
Atomic scattering factors from International Tables Vol C Tables 4.2.6.8 and 6.1.1.4
Flack parameter = 0.8 (12)
Flack H D (1983), *Acta Cryst.* A39, 876-881

Data collection: KappaCCD
Cell refinement: HKL Scalepack (Otwinowski & Minor 1997)
Data reduction: Denzo and Scalepak (Otwinowski & Minor, 1997)
Program(s) used to solve structure: *SIR*97(Cascarano al.,*Acta Cryst.*,1996,A52,C-79)
Program(s) used to refine structure: *SHELXL*-97 (Sheldrick, 1997)

Table 1. *Fractional atomic coordinates and equivalent isotropic thermal parameters (Å^2)*
$U_{eq} = 1/3\Sigma_i\Sigma_j U_{ij} a_i^* a_j^* \boldsymbol{a}_i.\boldsymbol{a}_j$.

	x	y	z	U_{eq}	Occ
O15	0.4624 (2)	0.13136 (19)	0.09136 (15)	0.0734 (5)	1
O16	0.5725 (2)	0.16592 (17)	0.46659 (15)	0.0756 (5)	1
O17	0.6126 (2)	0.58537 (14)	0.35834 (16)	0.0644 (4)	1
N14	0.5317 (3)	0.0979 (2)	0.23996 (19)	0.0644 (5)	1
C1	0.5406 (3)	0.1991 (2)	0.3379 (2)	0.0555 (4)	1
C2	0.5139 (2)	0.3488 (2)	0.28732 (18)	0.0532 (4)	1
C3	0.6496 (2)	0.44252 (19)	0.39736 (19)	0.0504 (4)	1
C4	0.8276 (3)	0.4111 (2)	0.4034 (2)	0.0580 (5)	1
C5	0.9593 (3)	0.4908 (3)	0.5312 (2)	0.0719 (6)	1
C6	0.9636 (3)	0.4592 (3)	0.6812 (2)	0.0690 (5)	1
C7	0.9432 (3)	0.5587 (3)	0.7665 (3)	0.0785 (7)	1
C8	0.9499 (4)	0.5465 (5)	0.9203 (3)	0.1116 (12)	1
C9	0.3350 (3)	0.3917 (3)	0.2720 (3)	0.0742 (6)	1

C10	0.8514 (3)	0.4454 (3)	0.2610 (3)	0.0749 (6)	1
C11	0.9960 (6)	0.3108 (4)	0.7273 (4)	0.1168 (14)	1
C12	0.5262 (4)	-0.0497 (3)	0.2723 (3)	0.0842 (7)	1
C13	0.5851 (4)	0.1171 (4)	0.0259 (3)	0.0938 (8)	1
H17	0.5523	0.6165	0.3997	0.097	1
H2	0.5227	0.3564	0.1930	0.064	1
H3	0.6461	0.4257	0.4925	0.060	1
H4	0.8451	0.3127	0.4198	0.070	1
H5A	1.0703	0.4715	0.5294	0.086	1
H5B	0.9388	0.5900	0.5137	0.086	1
H7	0.9207	0.6524	0.7302	0.094	1
H8A	0.8393	0.5616	0.9219	0.167	1
H8B	1.0263	0.6150	0.9804	0.167	1
H8C	0.9885	0.4549	0.9572	0.167	1
H9A	0.3160	0.4867	0.2392	0.089	1
H9B	0.3230	0.3836	0.3651	0.089	1
H9C	0.2540	0.3324	0.2021	0.089	1
H10A	0.9653	0.4244	0.2711	0.090	1
H10B	0.8294	0.5427	0.2391	0.090	1
H10C	0.7746	0.3905	0.1831	0.090	1
H11A	0.9955	0.3000	0.8242	0.140	1
H11B	1.1035	0.2823	0.7263	0.140	1
H11C	0.9086	0.2540	0.6603	0.140	1
H12A	0.5777	-0.0647	0.3759	0.101	1
H12B	0.5852	-0.1028	0.2233	0.101	1
H12C	0.4103	-0.0790	0.2384	0.101	1
H13A	0.5358	0.1397	-0.0763	0.113	1
H13B	0.6241	0.0222	0.0371	0.113	1
H13C	0.6785	0.1783	0.0733	0.113	1

Table 2. *Anisotropic displacement parameters (Å^2)*

	U_{11}	U_{12}	U_{13}	U_{22}	U_{23}	U_{33}
O15	0.0763 (10)	-0.0070 (8)	0.0195 (6)	0.0857 (11)	-0.0122 (7)	0.0543 (7)
O16	0.1066 (13)	-0.0215 (9)	0.0340 (8)	0.0687 (9)	0.0038 (6)	0.0551 (7)
O17	0.0802 (9)	0.0025 (7)	0.0364 (7)	0.0513 (8)	-0.0005 (6)	0.0693 (8)
N14	0.0763 (11)	-0.0042 (8)	0.0233 (8)	0.0579 (10)	-0.0064 (7)	0.0579 (8)
C1	0.0603 (10)	-0.0103 (8)	0.0258 (8)	0.0561 (10)	-0.0013 (8)	0.0542 (9)
C2	0.0586 (10)	-0.0052 (9)	0.0223 (8)	0.0574 (10)	0.0007 (8)	0.0461 (8)
C3	0.0572 (9)	-0.0021 (8)	0.0245 (7)	0.0495 (9)	-0.0006 (7)	0.0489 (8)
C4	0.0596 (11)	-0.0029 (8)	0.0296 (9)	0.0542 (10)	-0.0047 (8)	0.0656 (10)
C5	0.0577 (12)	-0.0091 (10)	0.0209 (10)	0.0746 (15)	-0.0037 (11)	0.0797 (14)
C6	0.0612 (11)	0.0057 (11)	0.0102 (9)	0.0665 (12)	0.0032 (10)	0.0679 (11)
C7	0.0649 (13)	0.0066 (12)	0.0091 (10)	0.0846 (17)	-0.0080 (12)	0.0721 (13)
C8	0.090 (2)	0.015 (2)	0.0152 (14)	0.155 (4)	-0.0147 (18)	0.0768 (16)
C9	0.0579 (12)	-0.0028 (11)	0.0171 (10)	0.0852 (16)	-0.0042 (11)	0.0734 (13)
C10	0.0778 (14)	-0.0166 (13)	0.0496 (11)	0.0814 (15)	-0.0135 (12)	0.0826 (14)
C11	0.166 (4)	0.032 (2)	0.023 (2)	0.079 (2)	0.0135 (15)	0.0860 (19)
C12	0.0972 (18)	-0.0073 (13)	0.0340 (15)	0.0575 (12)	-0.0065 (13)	0.0966 (17)
C13	0.110 (2)	-0.0175 (17)	0.0480 (14)	0.103 (2)	-0.0279 (15)	0.0788 (15)

Table 3 . *Geometric parameters (Å, °)*

O15—N14	1.396 (2)	N14—C12	1.455 (3)
O15—C13	1.422 (3)	C1—C2	1.508 (3)
O16—C1	1.233 (2)	C2—C9	1.532 (3)
O17—C3	1.426 (2)	C2—C3	1.546 (2)
N14—C1	1.349 (3)	C3—C4	1.526 (3)

C4—C10	1.522 (3)	C9—H9A	0.9600
C4—C5	1.544 (3)	C9—H9B	0.9599
C5—C6	1.492 (3)	C9—H9C	0.9600
C6—C7	1.323 (4)	C10—H10A	0.9600
C6—C11	1.488 (4)	C10—H10B	0.9600
C7—C8	1.495 (4)	C10—H10C	0.9601
O17—H17	0.8200	C11—H11A	0.9600
C2—H2	0.9600	C11—H11B	0.9600
C3—H3	0.9600	C11—H11C	0.9600
C4—H4	0.9600	C12—H12A	0.9600
C5—H5A	0.9700	C12—H12B	0.9600
C5—H5B	0.9700	C12—H12C	0.9600
C7—H7	0.9601	C13—H13A	0.9600
C8—H8A	0.9600	C13—H13B	0.9600
C8—H8B	0.9600	C13—H13C	0.9600
C8—H8C	0.9600		

N14—O15—C13	110.6 (2)	C8—C7—H7	112.2
C1—N14—O15	118.18 (18)	C7—C8—H8A	109.5
C1—N14—C12	122.8 (2)	C7—C8—H8B	109.5
O15—N14—C12	114.60 (18)	H8A—C8—H8B	109.5
O16—C1—N14	118.6 (2)	C7—C8—H8C	109.5
O16—C1—C2	122.23 (18)	H8A—C8—H8C	109.5
N14—C1—C2	119.13 (16)	H8B—C8—H8C	109.5
C1—C2—C9	108.13 (18)	C2—C9—H9A	109.0
C1—C2—C3	109.85 (15)	C2—C9—H9B	109.7
C9—C2—C3	111.87 (17)	H9A—C9—H9B	109.5
O17—C3—C4	108.52 (16)	C2—C9—H9C	109.7
O17—C3—C2	109.65 (15)	H9A—C9—H9C	109.5
C4—C3—C2	112.85 (15)	H9B—C9—H9C	109.5
C10—C4—C3	112.94 (18)	C4—C10—H10A	109.4
C10—C4—C5	109.76 (18)	C4—C10—H10B	109.6
C3—C4—C5	110.36 (17)	H10A—C10—H10B	109.5
C6—C5—C4	116.7 (2)	C4—C10—H10C	109.4
C7—C6—C11	123.3 (3)	H10A—C10—H10C	109.5
C7—C6—C5	121.3 (2)	H10B—C10—H10C	109.5
C11—C6—C5	115.4 (2)	C6—C11—H11A	109.7
C6—C7—C8	128.3 (3)	C6—C11—H11B	109.8
C3—O17—H17	109.5	H11A—C11—H11B	109.5
C1—C2—H2	109.6	C6—C11—H11C	108.9
C9—C2—H2	108.3	H11A—C11—H11C	109.5
C3—C2—H2	109.1	H11B—C11—H11C	109.5
O17—C3—H3	109.9	N14—C12—H12A	109.8
C4—C3—H3	108.5	N14—C12—H12B	109.9
C2—C3—H3	107.5	H12A—C12—H12B	109.5
C10—C4—H4	107.6	N14—C12—H12C	108.7
C3—C4—H4	106.9	H12A—C12—H12C	109.5
C5—C4—H4	109.2	H12B—C12—H12C	109.5
C6—C5—H5A	108.1	O15—C13—H13A	109.9
C4—C5—H5A	108.1	O15—C13—H13B	108.7
C6—C5—H5B	108.1	H13A—C13—H13B	109.5
C4—C5—H5B	108.1	O15—C13—H13C	109.8
H5A—C5—H5B	107.3	H13A—C13—H13C	109.5
C6—C7—H7	119.5	H13B—C13—H13C	109.5

C13—O15—N14—C1	116.5 (2)	C12—N14—C1—C2	-168.9 (2)
C13—O15—N14—C12	-86.4 (3)	O16—C1—C2—C9	-78.4 (2)
O15—N14—C1—O16	167.07 (19)	N14—C1—C2—C9	102.5 (2)
C12—N14—C1—O16	12.0 (4)	O16—C1—C2—C3	43.9 (3)
O15—N14—C1—C2	-13.9 (3)	N14—C1—C2—C3	-135.14 (19)

C1—C2—C3—O17	-173.36 (15)	C2—C3—C4—C5	-171.09 (16)
C9—C2—C3—O17	-53.3 (2)	C10—C4—C5—C6	-174.7 (2)
C1—C2—C3—C4	65.5 (2)	C3—C4—C5—C6	60.2 (3)
C9—C2—C3—C4	-174.36 (18)	C4—C5—C6—C7	-122.8 (3)
O17—C3—C4—C10	-56.1 (2)	C4—C5—C6—C11	58.6 (4)
C2—C3—C4—C10	65.6 (2)	C11—C6—C7—C8	0.9 (5)
O17—C3—C4—C5	67.2 (2)	C5—C6—C7—C8	-177.6 (2)

Crystallographic Data for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyl oxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (51) (CCDC674799)

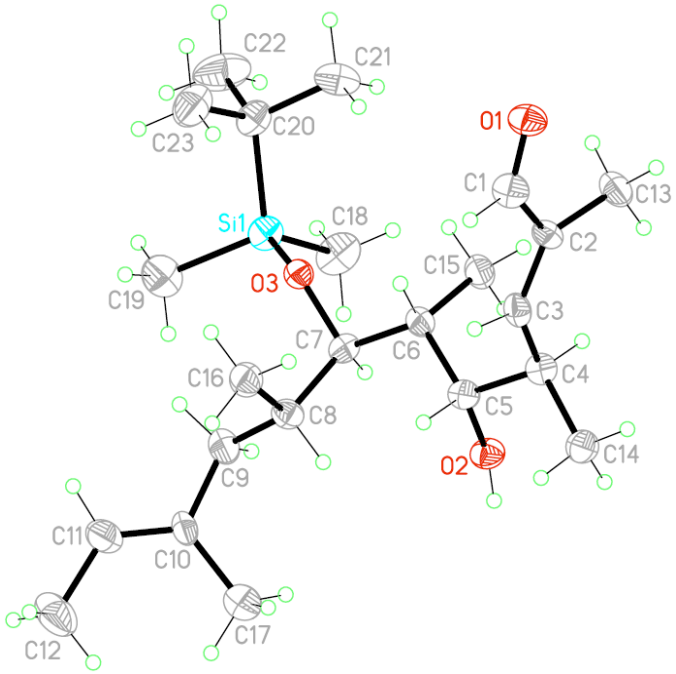


Table 1. Crystal data and structure refinement for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**).

Identification code	**51**	
Empirical formula	C23 H44 O3 Si	
Formula weight	396.67	
Temperature	100(2) K	
Wavelength	0.71073 Å	
Crystal system	Orthorhombic	
Space group	P2(1)2(1)2(1)	
Unit cell dimensions	a = 8.404(5) Å	α= 90°.
	b = 14.93(3) Å	β= 90°.
	c = 20.26(5) Å	γ = 90°.
Volume	2541(8) $Å^3$	
Z	4	
Density (calculated)	1.037 Mg/m^3	
Absorption coefficient	0.110 mm^{-1}	
F(000)	880	
Crystal size	0.42 x 0.17 x 0.06 mm^3	

Theta range for data collection	3.31 to 23.20°.
Index ranges	-9<=h<=9, -16<=k<=16, -22<=l<=22
Reflections collected	27234
Independent reflections	3591 [R(int) = 0.1531]
Completeness to theta = 23.20°	98.7 %
Absorption correction	Semi-empirical from equivalents
Max. and min. transmission	1.0000 and 0.1855
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	3591 / 0 / 246
Goodness-of-fit on F^2	1.079
Final R indices [I>2sigma(I)]	R1 = 0.0670, wR2 = 0.1138
R indices (all data)	R1 = 0.1170, wR2 = 0.1332
Absolute structure parameter	0.1(3)
Extinction coefficient	0.0030(11)
Largest diff. peak and hole	0.229 and -0.216 e.$Å^{-3}$

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3) for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**). U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

x	y	z	U(eq)
Si(1)	5210(2)	7809(1)	8474(1)	38(1)
O(1)	11173(4)	8462(2)	5235(2)	42(1)
O(2)	3118(4)	9390(2)	6184(2)	36(1)
O(3)	5240(3)	7747(2)	7669(1)	30(1)
C(1)	9727(6)	8398(3)	5199(2)	40(1)
C(2)	8573(5)	9082(3)	5414(2)	30(1)
C(3)	7030(6)	8868(3)	5370(2)	32(1)
C(4)	5600(5)	9403(3)	5546(2)	32(1)
C(5)	4518(5)	8875(3)	6035(2)	30(1)
C(6)	5334(5)	8629(3)	6682(2)	30(1)
C(7)	4244(5)	8111(3)	7159(2)	31(1)
C(8)	3206(5)	7374(3)	6845(2)	32(1)
C(9)	1974(5)	7029(3)	7341(3)	40(1)
C(10)	565(5)	6557(3)	7056(2)	34(1)
C(11)	238(6)	5712(4)	7195(3)	51(2)

C(12)	-1180(6)	5174(4)	6987(3)	69(2)
C(13)	9261(5)	9957(3)	5659(3)	43(2)
C(14)	4672(6)	9637(3)	4913(2)	41(1)
C(15)	6001(6)	9461(3)	7034(2)	40(1)
C(16)	4230(6)	6615(3)	6567(3)	41(1)
C(17)	-523(6)	7125(4)	6634(3)	56(2)
C(18)	4465(7)	8915(3)	8762(3)	55(2)
C(19)	3902(6)	6921(4)	8836(3)	54(2)
C(20)	7329(6)	7586(4)	8741(2)	40(1)
C(21)	8463(6)	8300(4)	8460(3)	57(2)
C(22)	7460(7)	7618(5)	9500(3)	77(2)
C(23)	7857(7)	6664(4)	8498(3)	61(2)

Table 3. Bond lengths [Å] and angles [°] for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**).

Si(1)-O(3)	1.633(5)
Si(1)-C(18)	1.859(6)
Si(1)-C(19)	1.873(6)
Si(1)-C(20)	1.890(5)
O(1)-C(1)	1.221(5)
O(2)-C(5)	1.438(5)
O(2)-H(2)	0.8400
O(3)-C(7)	1.437(5)
C(1)-C(2)	1.474(7)
C(1)-H(1)	0.9500
C(2)-C(3)	1.338(6)
C(2)-C(13)	1.513(7)
C(3)-C(4)	1.486(6)
C(3)-H(3)	0.9500
C(4)-C(14)	1.541(7)
C(4)-C(5)	1.559(6)
C(4)-H(4)	1.0000
C(5)-C(6)	1.524(7)
C(5)-H(5)	1.0000
C(6)-C(15)	1.537(6)
C(6)-C(7)	1.540(6)

C(6)-H(6)	1.0000
C(7)-C(8)	1.541(6)
C(7)-H(7)	1.0000
C(8)-C(16)	1.531(6)
C(8)-C(9)	1.532(6)
C(8)-H(8)	1.0000
C(9)-C(10)	1.495(6)
C(9)-H(9A)	0.9900
C(9)-H(9B)	0.9900
C(10)-C(11)	1.321(7)
C(10)-C(17)	1.511(7)
C(11)-C(12)	1.497(7)
C(11)-H(11)	0.9500
C(12)-H(12A)	0.9800
C(12)-H(12B)	0.9800
C(12)-H(12C)	0.9800
C(13)-H(13A)	0.9800
C(13)-H(13B)	0.9800
C(13)-H(13C)	0.9800
C(14)-H(14A)	0.9800
C(14)-H(14B)	0.9800
C(14)-H(14C)	0.9800
C(15)-H(15A)	0.9800
C(15)-H(15B)	0.9800
C(15)-H(15C)	0.9800
C(16)-H(16A)	0.9800
C(16)-H(16B)	0.9800
C(16)-H(16C)	0.9800
C(17)-H(17A)	0.9800
C(17)-H(17B)	0.9800
C(17)-H(17C)	0.9800
C(18)-H(18A)	0.9800
C(18)-H(18B)	0.9800
C(18)-H(18C)	0.9800
C(19)-H(19A)	0.9800
C(19)-H(19B)	0.9800
C(19)-H(19C)	0.9800
C(20)-C(23)	1.527(7)

C(20)-C(21)	1.539(7)
C(20)-C(22)	1.543(8)
C(21)-H(21A)	0.9800
C(21)-H(21B)	0.9800
C(21)-H(21C)	0.9800
C(22)-H(22A)	0.9800
C(22)-H(22B)	0.9800
C(22)-H(22C)	0.9800
C(23)-H(23A)	0.9800
C(23)-H(23B)	0.9800
C(23)-H(23C)	0.9800

O(3)-Si(1)-C(18)	111.7(2)
O(3)-Si(1)-C(19)	111.0(2)
C(18)-Si(1)-C(19)	108.0(3)
O(3)-Si(1)-C(20)	105.1(2)
C(18)-Si(1)-C(20)	112.6(3)
C(19)-Si(1)-C(20)	108.4(3)
C(5)-O(2)-H(2)	109.5
C(7)-O(3)-Si(1)	133.4(3)
O(1)-C(1)-C(2)	125.7(5)
O(1)-C(1)-H(1)	117.2
C(2)-C(1)-H(1)	117.2
C(3)-C(2)-C(1)	116.9(5)
C(3)-C(2)-C(13)	126.7(5)
C(1)-C(2)-C(13)	116.4(4)
C(2)-C(3)-C(4)	129.7(5)
C(2)-C(3)-H(3)	115.1
C(4)-C(3)-H(3)	115.1
C(3)-C(4)-C(14)	109.4(4)
C(3)-C(4)-C(5)	110.6(4)
C(14)-C(4)-C(5)	110.4(4)
C(3)-C(4)-H(4)	108.8
C(14)-C(4)-H(4)	108.8
C(5)-C(4)-H(4)	108.8
O(2)-C(5)-C(6)	108.5(4)
O(2)-C(5)-C(4)	109.9(4)
C(6)-C(5)-C(4)	113.9(4)

O(2)-C(5)-H(5)	108.1
C(6)-C(5)-H(5)	108.1
C(4)-C(5)-H(5)	108.1
C(5)-C(6)-C(15)	111.7(4)
C(5)-C(6)-C(7)	113.1(4)
C(15)-C(6)-C(7)	109.4(4)
C(5)-C(6)-H(6)	107.5
C(15)-C(6)-H(6)	107.5
C(7)-C(6)-H(6)	107.5
O(3)-C(7)-C(6)	107.1(4)
O(3)-C(7)-C(8)	110.9(4)
C(6)-C(7)-C(8)	115.9(4)
O(3)-C(7)-H(7)	107.5
C(6)-C(7)-H(7)	107.5
C(8)-C(7)-H(7)	107.5
C(16)-C(8)-C(9)	111.8(4)
C(16)-C(8)-C(7)	111.2(4)
C(9)-C(8)-C(7)	110.6(4)
C(16)-C(8)-H(8)	107.7
C(9)-C(8)-H(8)	107.7
C(7)-C(8)-H(8)	107.7
C(10)-C(9)-C(8)	116.1(4)
C(10)-C(9)-H(9A)	108.3
C(8)-C(9)-H(9A)	108.3
C(10)-C(9)-H(9B)	108.3
C(8)-C(9)-H(9B)	108.3
H(9A)-C(9)-H(9B)	107.4
C(11)-C(10)-C(9)	122.2(5)
C(11)-C(10)-C(17)	122.1(5)
C(9)-C(10)-C(17)	115.7(4)
C(10)-C(11)-C(12)	128.1(5)
C(10)-C(11)-H(11)	116.0
C(12)-C(11)-H(11)	116.0
C(11)-C(12)-H(12A)	109.5
C(11)-C(12)-H(12B)	109.5
H(12A)-C(12)-H(12B)	109.5
C(11)-C(12)-H(12C)	109.5
H(12A)-C(12)-H(12C)	109.5

H(12B)-C(12)-H(12C)	109.5
C(2)-C(13)-H(13A)	109.5
C(2)-C(13)-H(13B)	109.5
H(13A)-C(13)-H(13B)	109.5
C(2)-C(13)-H(13C)	109.5
H(13A)-C(13)-H(13C)	109.5
H(13B)-C(13)-H(13C)	109.5
C(4)-C(14)-H(14A)	109.5
C(4)-C(14)-H(14B)	109.5
H(14A)-C(14)-H(14B)	109.5
C(4)-C(14)-H(14C)	109.5
H(14A)-C(14)-H(14C)	109.5
H(14B)-C(14)-H(14C)	109.5
C(6)-C(15)-H(15A)	109.5
C(6)-C(15)-H(15B)	109.5
H(15A)-C(15)-H(15B)	109.5
C(6)-C(15)-H(15C)	109.5
H(15A)-C(15)-H(15C)	109.5
H(15B)-C(15)-H(15C)	109.5
C(8)-C(16)-H(16A)	109.5
C(8)-C(16)-H(16B)	109.5
H(16A)-C(16)-H(16B)	109.5
C(8)-C(16)-H(16C)	109.5
H(16A)-C(16)-H(16C)	109.5
H(16B)-C(16)-H(16C)	109.5
C(10)-C(17)-H(17A)	109.5
C(10)-C(17)-H(17B)	109.5
H(17A)-C(17)-H(17B)	109.5
C(10)-C(17)-H(17C)	109.5
H(17A)-C(17)-H(17C)	109.5
H(17B)-C(17)-H(17C)	109.5
Si(1)-C(18)-H(18A)	109.5
Si(1)-C(18)-H(18B)	109.5
H(18A)-C(18)-H(18B)	109.5
Si(1)-C(18)-H(18C)	109.5
H(18A)-C(18)-H(18C)	109.5
H(18B)-C(18)-H(18C)	109.5
Si(1)-C(19)-H(19A)	109.5

Si(1)-C(19)-H(19B)	109.5
H(19A)-C(19)-H(19B)	109.5
Si(1)-C(19)-H(19C)	109.5
H(19A)-C(19)-H(19C)	109.5
H(19B)-C(19)-H(19C)	109.5
C(23)-C(20)-C(21)	109.0(5)
C(23)-C(20)-C(22)	109.1(5)
C(21)-C(20)-C(22)	107.7(5)
C(23)-C(20)-Si(1)	109.9(4)
C(21)-C(20)-Si(1)	110.8(4)
C(22)-C(20)-Si(1)	110.3(4)
C(20)-C(21)-H(21A)	109.5
C(20)-C(21)-H(21B)	109.5
H(21A)-C(21)-H(21B)	109.5
C(20)-C(21)-H(21C)	109.5
H(21A)-C(21)-H(21C)	109.5
H(21B)-C(21)-H(21C)	109.5
C(20)-C(22)-H(22A)	109.5
C(20)-C(22)-H(22B)	109.5
H(22A)-C(22)-H(22B)	109.5
C(20)-C(22)-H(22C)	109.5
H(22A)-C(22)-H(22C)	109.5
H(22B)-C(22)-H(22C)	109.5
C(20)-C(23)-H(23A)	109.5
C(20)-C(23)-H(23B)	109.5
H(23A)-C(23)-H(23B)	109.5
C(20)-C(23)-H(23C)	109.5
H(23A)-C(23)-H(23C)	109.5
H(23B)-C(23)-H(23C)	109.5

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**). The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2\ a^{*2}U^{11} + ...\ + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}

Si(1)	43(1)	41(1)	30(1)	-3(1)	0(1)	2(1)
O(1)	28(2)	49(2)	48(3)	-6(2)	2(2)	4(2)
O(2)	28(2)	38(2)	42(2)	-3(2)	2(2)	7(2)
O(3)	28(2)	32(2)	31(2)	1(2)	1(2)	-1(2)
C(1)	42(3)	42(3)	36(3)	-8(3)	0(3)	0(3)
C(2)	29(3)	34(3)	28(3)	-4(3)	-1(2)	-6(2)
C(3)	45(3)	27(3)	23(3)	-3(2)	-2(2)	-9(3)
C(4)	34(3)	33(3)	28(3)	-4(2)	2(2)	1(2)
C(5)	29(3)	27(3)	33(3)	-9(2)	0(2)	4(2)
C(6)	30(2)	28(3)	32(3)	-2(2)	0(2)	-3(2)
C(7)	31(3)	34(3)	28(3)	1(2)	-1(2)	4(2)
C(8)	34(3)	36(3)	28(3)	-4(3)	-2(2)	-3(2)
C(9)	40(3)	35(4)	45(3)	1(3)	3(3)	-6(3)
C(10)	35(3)	27(3)	40(3)	4(3)	5(3)	-7(2)
C(11)	35(3)	45(4)	71(4)	0(3)	-5(3)	-6(3)
C(12)	40(3)	51(4)	115(6)	-9(4)	16(4)	-5(3)
C(13)	31(3)	30(3)	67(4)	-6(3)	6(3)	-3(2)
C(14)	40(3)	49(3)	33(3)	10(3)	-2(3)	-6(3)
C(15)	46(3)	38(3)	36(3)	5(3)	-5(3)	-12(3)
C(16)	45(3)	37(3)	41(3)	-2(3)	-4(3)	-3(3)
C(17)	43(3)	58(4)	66(4)	10(3)	-4(3)	-8(3)
C(18)	69(4)	57(4)	39(3)	-12(3)	-3(3)	16(3)
C(19)	60(4)	63(5)	39(4)	-3(3)	6(3)	-9(3)
C(20)	47(3)	42(4)	30(3)	0(3)	0(3)	5(3)
C(21)	42(3)	71(4)	58(4)	-7(4)	-13(3)	-6(3)
C(22)	64(4)	124(7)	43(4)	-2(4)	-15(3)	11(4)
C(23)	59(4)	60(4)	62(4)	14(4)	3(4)	13(3)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters ($Å^2$x 10^{3}) for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**).

x	y	z	U(eq)
H(2)	2479	9360	5865	54
H(1)	9309	7860	5018	48
H(3)	6817	8286	5202	38

H(4)	5952	9972	5762	38
H(5)	4176	8308	5814	36
H(6)	6255	8232	6573	36
H(7)	3512	8555	7370	37
H(8)	2615	7650	6467	39
H(9A)	2516	6615	7649	48
H(9B)	1586	7545	7603	48
H(11)	997	5408	7461	61
H(12A)	-1905	5554	6732	103
H(12B)	-1731	4947	7380	103
H(12C)	-830	4670	6714	103
H(13A)	8409	10402	5698	64
H(13B)	10063	10172	5345	64
H(13C)	9756	9865	6091	64
H(14A)	5394	9915	4592	61
H(14B)	3812	10056	5020	61
H(14C)	4220	9090	4723	61
H(15A)	5139	9891	7111	60
H(15B)	6822	9738	6758	60
H(15C)	6466	9283	7458	60
H(16A)	4828	6333	6927	61
H(16B)	4974	6857	6240	61
H(16C)	3543	6167	6357	61
H(17A)	-129	7131	6179	83
H(17B)	-544	7738	6807	83
H(17C)	-1600	6873	6643	83
H(18A)	5153	9391	8590	82
H(18B)	4471	8931	9245	82
H(18C)	3376	9006	8602	82
H(19A)	2785	7068	8749	81
H(19B)	4079	6887	9313	81
H(19C)	4156	6341	8634	81
H(21A)	9551	8175	8607	85
H(21B)	8138	8893	8617	85
H(21C)	8421	8287	7976	85
H(22A)	6812	7138	9692	115
H(22B)	7077	8199	9660	115
H(22C)	8573	7537	9631	115

H(23A)	8956	6553	8638	91
H(23B)	7795	6644	8015	91
H(23C)	7159	6204	8686	91

Table 6. Torsion angles [°] for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**).

C(18)-Si(1)-O(3)-C(7)	-33.9(5)
C(19)-Si(1)-O(3)-C(7)	86.7(4)
C(20)-Si(1)-O(3)-C(7)	-156.3(4)
O(1)-C(1)-C(2)-C(3)	-176.3(5)
O(1)-C(1)-C(2)-C(13)	4.0(8)
C(1)-C(2)-C(3)-C(4)	179.9(5)
C(13)-C(2)-C(3)-C(4)	-0.4(9)
C(2)-C(3)-C(4)-C(14)	112.6(6)
C(2)-C(3)-C(4)-C(5)	-125.6(6)
C(3)-C(4)-C(5)-O(2)	-178.2(4)
C(14)-C(4)-C(5)-O(2)	-57.0(5)
C(3)-C(4)-C(5)-C(6)	59.8(5)
C(14)-C(4)-C(5)-C(6)	-179.0(4)
O(2)-C(5)-C(6)-C(15)	-66.8(5)
C(4)-C(5)-C(6)-C(15)	55.9(5)
O(2)-C(5)-C(6)-C(7)	57.1(5)
C(4)-C(5)-C(6)-C(7)	179.8(4)
Si(1)-O(3)-C(7)-C(6)	125.6(4)
Si(1)-O(3)-C(7)-C(8)	-107.1(4)
C(5)-C(6)-C(7)-O(3)	166.9(4)
C(15)-C(6)-C(7)-O(3)	-67.9(5)
C(5)-C(6)-C(7)-C(8)	42.5(5)
C(15)-C(6)-C(7)-C(8)	167.7(4)
O(3)-C(7)-C(8)-C(16)	-57.1(5)
C(6)-C(7)-C(8)-C(16)	65.3(5)
O(3)-C(7)-C(8)-C(9)	67.8(5)
C(6)-C(7)-C(8)-C(9)	-169.8(4)
C(16)-C(8)-C(9)-C(10)	-75.4(5)
C(7)-C(8)-C(9)-C(10)	160.1(4)
C(8)-C(9)-C(10)-C(11)	118.9(6)

C(8)-C(9)-C(10)-C(17)	-65.0(6)
C(9)-C(10)-C(11)-C(12)	175.3(5)
C(17)-C(10)-C(11)-C(12)	-0.5(9)
O(3)-Si(1)-C(20)-C(23)	-59.1(4)
C(18)-Si(1)-C(20)-C(23)	179.1(4)
C(19)-Si(1)-C(20)-C(23)	59.7(4)
O(3)-Si(1)-C(20)-C(21)	61.4(4)
C(18)-Si(1)-C(20)-C(21)	-60.3(5)
C(19)-Si(1)-C(20)-C(21)	-179.7(4)
O(3)-Si(1)-C(20)-C(22)	-179.5(4)
C(18)-Si(1)-C(20)-C(22)	58.8(5)
C(19)-Si(1)-C(20)-C(22)	-60.6(5)

Symmetry transformations used to generate equivalent atoms:

Table 7. Hydrogen bonds for (2*E*,4*R*,5*S*,6*R*,7*R*,8*S*,10*E*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxy-2,4,6,8,10-pentamethyldodeca-2,10-dienal (**51**). [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
O(2)-H(2)...O(1)#1	0.84	2.15	2.879(6)	144.6

Symmetry transformations used to generate equivalent atoms:

#1 x-1,y,z

Crystallographic Data for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (142)

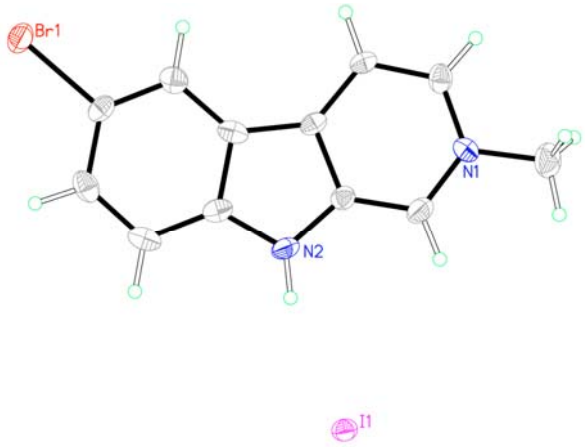


Table 1. Crystal data and structure refinement for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**).

Identification code	**142**	
Empirical formula	C12 H10 Br I N2	
Formula weight	389.03	
Temperature	140(2) K	
Wavelength	0.71073 Å	
Crystal system	Monoclinic	
Space group	P2(1)/c	
Unit cell dimensions	a = 10.7180(9) Å	α= 90°.
	b = 15.5515(16) Å	β= 93.188(8)°.
	c = 7.4595(6) Å	γ = 90°.
Volume	1241.42(19) $Å^3$	
Z	4	
Density (calculated)	2.081 Mg/m^3	
Absorption coefficient	5.772 mm^{-1}	
F(000)	736	
Crystal size	0.31 x 0.21 x 0.14 mm^3	
Theta range for data collection	3.03 to 26.37°.	
Index ranges	-13<=h<=13, -19<=k<=19, -6<=l<=9	
Reflections collected	2517	
Independent reflections	2517 [R(int) = 0.0000]	
Completeness to theta = 26.37°	99.3 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.446 and 0.162	
Refinement method	Full-matrix least-squares on F^2	

Data / restraints / parameters	2517 / 0 / 146
Goodness-of-fit on F^2	1.129
Final R indices [I>2sigma(I)]	R1 = 0.0561, wR2 = 0.1504
R indices (all data)	R1 = 0.0629, wR2 = 0.1530
Largest diff. peak and hole	4.346 and -1.602 e.Å^{-3}

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters (Å^{2}x 10^3) for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**). U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

x	y	z	U(eq)
Cl(1)	695(2)	2789(1)	3951(2)	34(1)
N(1)	5808(5)	5840(1)	8639(4)	19(1)
N(2)	7973(5)	4397(1)	7848(4)	20(1)
C(1)	3537(6)	5758(2)	7848(5)	20(1)
C(2)	2719(6)	5232(2)	7073(5)	18(1)
C(3)	4255(6)	4782(1)	7068(5)	17(1)
C(4)	6642(6)	4875(2)	7895(5)	18(1)
C(5)	7361(6)	5415(2)	8697(5)	20(1)
C(6)	4125(6)	4190(1)	6447(5)	17(1)
C(7)	2318(6)	3824(2)	5537(5)	19(1)
C(8)	2859(6)	3260(2)	5163(5)	22(1)
C(9)	5108(6)	3049(2)	5671(5)	22(1)
C(10)	6885(7)	3400(2)	6565(5)	25(1)
C(11)	6430(6)	3985(2)	6974(5)	17(1)
C(12)	6594(7)	6419(2)	9452(5)	26(1)

Table 3. Bond lengths [Å] and angles [°] for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**).

Cl(1)-C(8)	1.752(4)
N(1)-C(5)	1.344(4)
N(1)-C(1)	1.360(5)
N(1)-C(12)	1.481(4)
N(2)-C(4)	1.364(4)
N(2)-C(11)	1.365(4)
C(1)-C(2)	1.367(5)

C(1)-H(1)	0.9500
C(2)-C(3)	1.387(5)
C(2)-H(2)	0.9500
C(3)-C(6)	1.424(5)
C(3)-C(4)	1.431(5)
C(4)-C(5)	1.390(5)
C(5)-H(5)	0.9500
C(6)-C(7)	1.403(5)
C(6)-C(11)	1.427(5)
C(7)-C(8)	1.377(5)
C(7)-H(7)	0.9500
C(8)-C(9)	1.401(5)
C(9)-C(10)	1.368(5)
C(9)-H(9)	0.9500
C(10)-C(11)	1.414(5)
C(10)-H(10)	0.9500
C(12)-H(12A)	0.9800
C(12)-H(12B)	0.9800
C(12)-H(12C)	0.9800

C(5)-N(1)-C(1)	122.4(3)
C(5)-N(1)-C(12)	118.9(3)
C(1)-N(1)-C(12)	118.7(3)
C(4)-N(2)-C(11)	103.3(3)
N(1)-C(1)-C(2)	121.1(3)
N(1)-C(1)-H(1)	119.5
C(2)-C(1)-H(1)	119.5
C(1)-C(2)-C(3)	118.9(3)
C(1)-C(2)-H(2)	120.6
C(3)-C(2)-H(2)	120.6
C(2)-C(3)-C(6)	136.4(3)
C(2)-C(3)-C(4)	119.5(3)
C(6)-C(3)-C(4)	104.1(3)
N(2)-C(4)-C(5)	127.0(3)
N(2)-C(4)-C(3)	114.0(3)
C(5)-C(4)-C(3)	118.9(3)
N(1)-C(5)-C(4)	119.3(3)
N(1)-C(5)-H(5)	120.4

C(4)-C(5)-H(5)	120.4
C(7)-C(6)-C(3)	133.8(3)
C(7)-C(6)-C(11)	121.3(3)
C(3)-C(6)-C(11)	104.9(3)
C(8)-C(7)-C(6)	117.3(3)
C(8)-C(7)-H(7)	121.3
C(6)-C(7)-H(7)	121.3
C(7)-C(8)-C(9)	122.4(3)
C(7)-C(8)-Cl(1)	119.6(3)
C(9)-C(8)-Cl(1)	118.0(3)
C(10)-C(9)-C(8)	120.9(3)
C(10)-C(9)-H(9)	119.5
C(8)-C(9)-H(9)	119.5
C(9)-C(10)-C(11)	119.1(3)
C(9)-C(10)-H(10)	120.4
C(11)-C(10)-H(10)	120.4
N(2)-C(11)-C(10)	127.3(3)
N(2)-C(11)-C(6)	113.7(3)
C(10)-C(11)-C(6)	119.0(3)
N(1)-C(12)-H(12A)	109.5
N(1)-C(12)-H(12B)	109.5
H(12A)-C(12)-H(12B)	109.5
N(1)-C(12)-H(12C)	109.5
H(12A)-C(12)-H(12C)	109.5
H(12B)-C(12)-H(12C)	109.5

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**). The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2\ a^{*2}U^{11} + ... + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
Cl(1)	42(1)	23(1)	36(1)	-8(1)	8(1)	-9(1)
N(1)	31(2)	12(2)	15(2)	-1(1)	5(1)	-5(1)
N(2)	24(2)	15(2)	21(2)	-1(1)	3(1)	3(1)
C(1)	26(2)	14(2)	21(2)	4(2)	8(2)	6(2)

C(2)	18(2)	18(2)	16(2)	0(2)	1(2)	-1(2)
C(3)	25(2)	14(2)	10(2)	0(2)	4(2)	-2(2)
C(4)	24(2)	17(2)	13(2)	3(2)	4(2)	-2(2)
C(5)	22(2)	18(2)	18(2)	2(2)	2(2)	-2(2)
C(6)	27(2)	13(2)	13(2)	4(2)	7(2)	0(2)
C(7)	22(2)	18(2)	17(2)	4(2)	6(2)	0(2)
C(8)	35(2)	17(2)	15(2)	-2(2)	8(2)	-10(2)
C(9)	36(2)	11(2)	24(2)	1(2)	13(2)	1(2)
C(10)	26(2)	18(2)	31(2)	4(2)	9(2)	3(2)
C(11)	25(2)	13(2)	14(2)	4(2)	6(2)	1(2)
C(12)	41(3)	14(2)	24(2)	-3(2)	9(2)	-6(2)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters (Å^{2}x 10^{3}) for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**).

x	y	z	U(eq)
H(1)	2501	6071	7833	24
H(2)	1127	5176	6548	22
H(5)	8933	5484	9281	24
H(7)	783	3960	5193	23
H(9)	5402	2655	5391	27
H(10)	8405	3253	6907	30
H(12A)	8182	6390	10208	40
H(12B)	5622	6549	10267	40
H(12C)	6497	6700	8420	40

Table 6. Torsion angles [°] for 6-bromo-2-methyl-9*H*-beta-carbolin-2-ium iodide (**142**).

C(5)-N(1)-C(1)-C(2)	0.3(5)
C(12)-N(1)-C(1)-C(2)	179.9(3)
N(1)-C(1)-C(2)-C(3)	-1.3(5)
C(1)-C(2)-C(3)-C(6)	179.3(4)
C(1)-C(2)-C(3)-C(4)	0.9(5)
C(11)-N(2)-C(4)-C(5)	177.8(4)
C(11)-N(2)-C(4)-C(3)	0.1(4)

C(2)-C(3)-C(4)-N(2)	178.4(3)
C(6)-C(3)-C(4)-N(2)	-0.4(4)
C(2)-C(3)-C(4)-C(5)	0.5(5)
C(6)-C(3)-C(4)-C(5)	-178.4(3)
C(1)-N(1)-C(5)-C(4)	1.1(5)
C(12)-N(1)-C(5)-C(4)	-178.4(3)
N(2)-C(4)-C(5)-N(1)	-179.1(3)
C(3)-C(4)-C(5)-N(1)	-1.5(5)
C(2)-C(3)-C(6)-C(7)	1.2(7)
C(4)-C(3)-C(6)-C(7)	179.7(4)
C(2)-C(3)-C(6)-C(11)	-178.0(4)
C(4)-C(3)-C(6)-C(11)	0.6(3)
C(3)-C(6)-C(7)-C(8)	-179.5(4)
C(11)-C(6)-C(7)-C(8)	-0.4(5)
C(6)-C(7)-C(8)-C(9)	0.6(5)
C(6)-C(7)-C(8)-Cl(1)	-177.6(2)
C(7)-C(8)-C(9)-C(10)	-0.3(6)
Cl(1)-C(8)-C(9)-C(10)	177.9(3)
C(8)-C(9)-C(10)-C(11)	-0.1(5)
C(4)-N(2)-C(11)-C(10)	-179.6(3)
C(4)-N(2)-C(11)-C(6)	0.3(4)
C(9)-C(10)-C(11)-N(2)	-179.9(3)
C(9)-C(10)-C(11)-C(6)	0.2(5)
C(7)-C(6)-C(11)-N(2)	-179.9(3)
C(3)-C(6)-C(11)-N(2)	-0.6(4)
C(7)-C(6)-C(11)-C(10)	0.0(5)
C(3)-C(6)-C(11)-C(10)	179.3(3)

Symmetry transformations used to generate equivalent atoms:

Crystallographic Data for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (144)

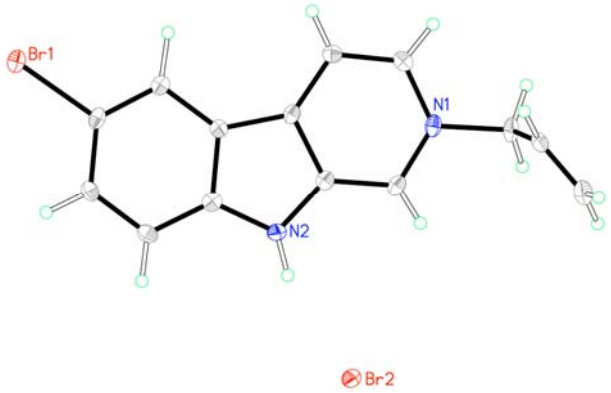


Table 1. Crystal data and structure refinement for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**).

Identification code	**144**	
Empirical formula	C14 H12 Br2 N2	
Formula weight	368.08	
Temperature	100(2) K	
Wavelength	0.71073 Å	
Crystal system	Monoclinic	
Space group	P2(1)/c	
Unit cell dimensions	a = 8.1536(7) Å	α= 90°.
	b = 22.969(2) Å	β= 107.832(8)°.
	c = 7.4050(6) Å	γ = 90°.
Volume	1320.2(2) $Å^3$	
Z	4	
Density (calculated)	1.852 Mg/m^3	
Absorption coefficient	6.123 mm^{-1}	
F(000)	720	
Crystal size	0.79 x 0.27 x 0.07 mm^3	
Theta range for data collection	3.37 to 27.50°.	
Index ranges	-10<=h<=10, -29<=k<=29, -9<=l<=9	
Reflections collected	51084	
Independent reflections	3031 [R(int) = 0.0461]	
Completeness to theta = 27.50°	99.8 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.651 and 0.289	
Refinement method	Full-matrix least-squares on F^2	

Data / restraints / parameters	3031 / 0 / 163
Goodness-of-fit on F^2	1.153
Final R indices [I>2sigma(I)]	R1 = 0.0260, wR2 = 0.0602
R indices (all data)	R1 = 0.0306, wR2 = 0.0625
Largest diff. peak and hole	0.604 and -0.660 e.Å^{-3}

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters (Å^{2}x 10^3) for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**). U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

x	y	z	U(eq)
Br(1)	3950(1)	5037(1)	2152(1)	18(1)
N(1)	1943(3)	8521(1)	395(3)	13(1)
N(2)	5420(3)	7611(1)	3358(3)	14(1)
C(1)	871(3)	8091(1)	-574(4)	15(1)
C(2)	1296(3)	7511(1)	-225(4)	14(1)
C(3)	2865(3)	7374(1)	1124(3)	12(1)
C(4)	3954(3)	7836(1)	2087(3)	13(1)
C(5)	3468(3)	8415(1)	1715(4)	14(1)
C(6)	3746(3)	6841(1)	1888(3)	12(1)
C(7)	3319(3)	6249(1)	1531(4)	13(1)
C(8)	4486(3)	5847(1)	2583(4)	14(1)
C(9)	6022(3)	6011(1)	3983(4)	14(1)
C(10)	6451(3)	6592(1)	4346(4)	14(1)
C(11)	5313(3)	7010(1)	3267(4)	13(1)
C(12)	1332(4)	9139(1)	72(4)	17(1)
C(13)	568(3)	9321(1)	1590(4)	14(1)
C(14)	1036(4)	9794(1)	2630(4)	19(1)
Br(2)	8149(1)	8675(1)	4836(1)	15(1)

Table 3. Bond lengths [Å] and angles [°] for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**).

Br(1)-C(8)	1.915(3)
N(1)-C(5)	1.348(3)
N(1)-C(1)	1.368(3)
N(1)-C(12)	1.499(3)
N(2)-C(4)	1.377(3)

N(2)-C(11)	1.384(3)
N(2)-H(2)	0.8800
C(1)-C(2)	1.380(4)
C(1)-H(1)	0.9500
C(2)-C(3)	1.397(4)
C(2)-H(2A)	0.9500
C(3)-C(4)	1.428(3)
C(3)-C(6)	1.443(3)
C(4)-C(5)	1.390(4)
C(5)-H(5)	0.9500
C(6)-C(7)	1.409(3)
C(6)-C(11)	1.423(3)
C(7)-C(8)	1.382(4)
C(7)-H(7)	0.9500
C(8)-C(9)	1.411(4)
C(9)-C(10)	1.385(4)
C(9)-H(9)	0.9500
C(10)-C(11)	1.402(4)
C(10)-H(10)	0.9500
C(12)-C(13)	1.501(4)
C(12)-H(12A)	0.9900
C(12)-H(12B)	0.9900
C(13)-C(14)	1.319(4)
C(13)-H(13)	0.9500
C(14)-H(14A)	0.9500
C(14)-H(14B)	0.9500

C(5)-N(1)-C(1)	123.3(2)
C(5)-N(1)-C(12)	118.4(2)
C(1)-N(1)-C(12)	118.2(2)
C(4)-N(2)-C(11)	108.3(2)
C(4)-N(2)-H(2)	125.9
C(11)-N(2)-H(2)	125.9
N(1)-C(1)-C(2)	121.0(2)
N(1)-C(1)-H(1)	119.5
C(2)-C(1)-H(1)	119.5
C(1)-C(2)-C(3)	118.3(2)
C(1)-C(2)-H(2A)	120.8

C(3)-C(2)-H(2A)	120.8
C(2)-C(3)-C(4)	118.8(2)
C(2)-C(3)-C(6)	135.2(2)
C(4)-C(3)-C(6)	106.0(2)
N(2)-C(4)-C(5)	129.1(2)
N(2)-C(4)-C(3)	109.8(2)
C(5)-C(4)-C(3)	121.1(2)
N(1)-C(5)-C(4)	117.4(2)
N(1)-C(5)-H(5)	121.3
C(4)-C(5)-H(5)	121.3
C(7)-C(6)-C(11)	120.9(2)
C(7)-C(6)-C(3)	132.8(2)
C(11)-C(6)-C(3)	106.3(2)
C(8)-C(7)-C(6)	116.8(2)
C(8)-C(7)-H(7)	121.6
C(6)-C(7)-H(7)	121.6
C(7)-C(8)-C(9)	122.6(2)
C(7)-C(8)-Br(1)	118.18(19)
C(9)-C(8)-Br(1)	119.18(19)
C(10)-C(9)-C(8)	121.0(2)
C(10)-C(9)-H(9)	119.5
C(8)-C(9)-H(9)	119.5
C(9)-C(10)-C(11)	117.7(2)
C(9)-C(10)-H(10)	121.2
C(11)-C(10)-H(10)	121.2
N(2)-C(11)-C(10)	129.3(2)
N(2)-C(11)-C(6)	109.6(2)
C(10)-C(11)-C(6)	121.0(2)
N(1)-C(12)-C(13)	109.7(2)
N(1)-C(12)-H(12A)	109.7
C(13)-C(12)-H(12A)	109.7
N(1)-C(12)-H(12B)	109.7
C(13)-C(12)-H(12B)	109.7
H(12A)-C(12)-H(12B)	108.2
C(14)-C(13)-C(12)	123.6(2)
C(14)-C(13)-H(13)	118.2
C(12)-C(13)-H(13)	118.2
C(13)-C(14)-H(14A)	120.0

C(13)-C(14)-H(14B)	120.0
H(14A)-C(14)-H(14B)	120.0

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**). The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2\ a^{*2}U^{11} + ... + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
Br(1)	20(1)	12(1)	21(1)	1(1)	6(1)	1(1)
N(1)	18(1)	10(1)	14(1)	2(1)	8(1)	2(1)
N(2)	12(1)	13(1)	15(1)	0(1)	1(1)	-2(1)
C(1)	13(1)	17(1)	14(1)	1(1)	4(1)	2(1)
C(2)	14(1)	15(1)	12(1)	0(1)	4(1)	-1(1)
C(3)	14(1)	10(1)	12(1)	1(1)	6(1)	-1(1)
C(4)	14(1)	13(1)	13(1)	1(1)	6(1)	-1(1)
C(5)	17(1)	12(1)	14(1)	-1(1)	7(1)	-1(1)
C(6)	11(1)	13(1)	12(1)	1(1)	5(1)	0(1)
C(7)	14(1)	13(1)	13(1)	0(1)	6(1)	-1(1)
C(8)	17(1)	10(1)	16(1)	0(1)	8(1)	-1(1)
C(9)	13(1)	15(1)	15(1)	2(1)	5(1)	3(1)
C(10)	11(1)	18(1)	14(1)	1(1)	4(1)	1(1)
C(11)	14(1)	15(1)	13(1)	0(1)	7(1)	-2(1)
C(12)	23(1)	10(1)	18(1)	4(1)	8(1)	5(1)
C(13)	12(1)	15(1)	16(1)	4(1)	4(1)	3(1)
C(14)	22(1)	17(1)	17(1)	3(1)	7(1)	6(1)
Br(2)	15(1)	13(1)	16(1)	0(1)	3(1)	-2(1)

Table 5. Hydrogen coordinates ($x\ 10^4$) and isotropic displacement parameters ($Å^2 x\ 10^{\ 3}$) for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**).

	x	y	z	U(eq)
H(2)	6279	7814	4101	17
H(1)	-183	8192	-1501	18

H(2A)	541	7213	-885	16
H(5)	4184	8724	2366	17
H(7)	2279	6132	611	15
H(9)	6773	5719	4688	17
H(10)	7481	6704	5293	17
H(12A)	455	9175	-1189	20
H(12B)	2310	9397	95	20
H(13)	-300	9082	1814	17
H(14A)	1901	10041	2437	22
H(14B)	507	9887	3572	22

Table 6. Torsion angles [°] for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**).

C(5)-N(1)-C(1)-C(2)	0.8(4)
C(12)-N(1)-C(1)-C(2)	-174.6(2)
N(1)-C(1)-C(2)-C(3)	-1.0(4)
C(1)-C(2)-C(3)-C(4)	0.3(4)
C(1)-C(2)-C(3)-C(6)	179.2(3)
C(11)-N(2)-C(4)-C(5)	178.2(2)
C(11)-N(2)-C(4)-C(3)	-0.6(3)
C(2)-C(3)-C(4)-N(2)	179.6(2)
C(6)-C(3)-C(4)-N(2)	0.3(3)
C(2)-C(3)-C(4)-C(5)	0.7(4)
C(6)-C(3)-C(4)-C(5)	-178.6(2)
C(1)-N(1)-C(5)-C(4)	0.1(4)
C(12)-N(1)-C(5)-C(4)	175.6(2)
N(2)-C(4)-C(5)-N(1)	-179.5(2)
C(3)-C(4)-C(5)-N(1)	-0.9(4)
C(2)-C(3)-C(6)-C(7)	0.0(5)
C(4)-C(3)-C(6)-C(7)	179.0(3)
C(2)-C(3)-C(6)-C(11)	-179.0(3)
C(4)-C(3)-C(6)-C(11)	0.1(3)
C(11)-C(6)-C(7)-C(8)	0.2(4)
C(3)-C(6)-C(7)-C(8)	-178.6(2)
C(6)-C(7)-C(8)-C(9)	1.3(4)
C(6)-C(7)-C(8)-Br(1)	179.66(17)
C(7)-C(8)-C(9)-C(10)	-1.3(4)

Br(1)-C(8)-C(9)-C(10)	-179.61(19)
C(8)-C(9)-C(10)-C(11)	-0.4(4)
C(4)-N(2)-C(11)-C(10)	-176.8(2)
C(4)-N(2)-C(11)-C(6)	0.6(3)
C(9)-C(10)-C(11)-N(2)	179.1(2)
C(9)-C(10)-C(11)-C(6)	1.9(4)
C(7)-C(6)-C(11)-N(2)	-179.5(2)
C(3)-C(6)-C(11)-N(2)	-0.4(3)
C(7)-C(6)-C(11)-C(10)	-1.8(4)
C(3)-C(6)-C(11)-C(10)	177.3(2)
C(5)-N(1)-C(12)-C(13)	-79.5(3)
C(1)-N(1)-C(12)-C(13)	96.2(3)
N(1)-C(12)-C(13)-C(14)	127.6(3)

Symmetry transformations used to generate equivalent atoms:

Table 7. Hydrogen bonds for 2-allyl-6-bromo-9*H*-beta-carbolin-2-ium bromide (**144**). [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
N(2)-H(2)...Br(2)	0.88	2.45	3.260(2)	153

Symmetry transformations used to generate equivalent atoms:

Crystallographic Data for 6-chloro-2-methyl-2*H*-beta-carboline (158) (CCDC 728844)

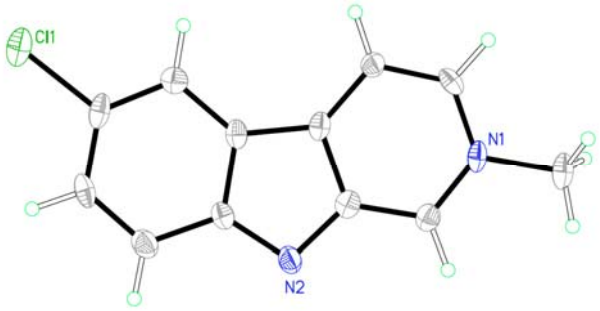


Table 1. Crystal data and structure refinement for 6-chloro-2-methyl-2*H*-beta-carboline (**158**).

Identification code	**158**	
Empirical formula	C12 H9 Cl N2	
Formula weight	216.66	
Temperature	140(2) K	
Wavelength	0.71073 Å	
Crystal system	Monoclinic	
Space group	P2(1)/c	
Unit cell dimensions	a = 6.0420(8) Å	α = 90°.
	b = 22.907(3) Å	β = 104.655(14)°.
	c = 7.2662(11) Å	γ = 90°.
Volume	973.0(2) $Å^3$	
Z	4	
Density (calculated)	1.479 Mg/m^3	
Absorption coefficient	0.354 mm^{-1}	
F(000)	448	
Crystal size	0.23 x 0.12 x 0.10 mm^3	
Theta range for data collection	3.03 to 26.36°.	
Index ranges	-7<=h<=7, -28<=k<=27, -8<=l<=9	
Reflections collected	8596	
Independent reflections	1978 [R(int) = 0.0795]	
Completeness to theta = 26.36°	99.3 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	1.00000 and 0.85917	
Refinement method	Full-matrix least-squares on F^2	
Data / restraints / parameters	1978 / 0 / 136	
Goodness-of-fit on F^2	1.000	

Final R indices [I>2sigma(I)]	R1 = 0.0644, wR2 = 0.1287
R indices (all data)	R1 = 0.1227, wR2 = 0.1456
Largest diff. peak and hole	0.400 and -0.411 e.Å^{-3}

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters (Å^{2}x 10^3) for 6-chloro-2-methyl-2*H*-beta-carboline (**158**). U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

x		y	z	U(eq)
Cl(1)	695(2)	2789(1)	3951(2)	34(1)
N(1)	5808(5)	5840(1)	8639(4)	19(1)
N(2)	7973(5)	4397(1)	7848(4)	20(1)
C(1)	3537(6)	5758(2)	7848(5)	20(1)
C(2)	2719(6)	5232(2)	7073(5)	18(1)
C(3)	4255(6)	4782(1)	7068(5)	17(1)
C(4)	6642(6)	4875(2)	7895(5)	18(1)
C(5)	7361(6)	5415(2)	8697(5)	20(1)
C(6)	4125(6)	4190(1)	6447(5)	17(1)
C(7)	2318(6)	3824(2)	5537(5)	19(1)
C(8)	2859(6)	3260(2)	5163(5)	22(1)
C(9)	5108(6)	3049(2)	5671(5)	22(1)
C(10)	6885(7)	3400(2)	6565(5)	25(1)
C(11)	6430(6)	3985(2)	6974(5)	17(1)
C(12)	6594(7)	6419(2)	9452(5)	26(1)

Table 3. Bond lengths [Å] and angles [°] for 6-chloro-2-methyl-2*H*-beta-carboline (**158**).

Cl(1)-C(8)	1.752(4)
N(1)-C(5)	1.344(4)
N(1)-C(1)	1.360(5)
N(1)-C(12)	1.481(4)
N(2)-C(4)	1.364(4)
N(2)-C(11)	1.365(4)
C(1)-C(2)	1.367(5)
C(1)-H(1)	0.9500
C(2)-C(3)	1.387(5)

C(2)-H(2)	0.9500
C(3)-C(6)	1.424(5)
C(3)-C(4)	1.431(5)
C(4)-C(5)	1.390(5)
C(5)-H(5)	0.9500
C(6)-C(7)	1.403(5)
C(6)-C(11)	1.427(5)
C(7)-C(8)	1.377(5)
C(7)-H(7)	0.9500
C(8)-C(9)	1.401(5)
C(9)-C(10)	1.368(5)
C(9)-H(9)	0.9500
C(10)-C(11)	1.414(5)
C(10)-H(10)	0.9500
C(12)-H(12A)	0.9800
C(12)-H(12B)	0.9800
C(12)-H(12C)	0.9800

C(5)-N(1)-C(1)	122.4(3)
C(5)-N(1)-C(12)	118.9(3)
C(1)-N(1)-C(12)	118.7(3)
C(4)-N(2)-C(11)	103.3(3)
N(1)-C(1)-C(2)	121.1(3)
N(1)-C(1)-H(1)	119.5
C(2)-C(1)-H(1)	119.5
C(1)-C(2)-C(3)	118.9(3)
C(1)-C(2)-H(2)	120.6
C(3)-C(2)-H(2)	120.6
C(2)-C(3)-C(6)	136.4(3)
C(2)-C(3)-C(4)	119.5(3)
C(6)-C(3)-C(4)	104.1(3)
N(2)-C(4)-C(5)	127.0(3)
N(2)-C(4)-C(3)	114.0(3)
C(5)-C(4)-C(3)	118.9(3)
N(1)-C(5)-C(4)	119.3(3)
N(1)-C(5)-H(5)	120.4
C(4)-C(5)-H(5)	120.4
C(7)-C(6)-C(3)	133.8(3)

C(7)-C(6)-C(11)	121.3(3)
C(3)-C(6)-C(11)	104.9(3)
C(8)-C(7)-C(6)	117.3(3)
C(8)-C(7)-H(7)	121.3
C(6)-C(7)-H(7)	121.3
C(7)-C(8)-C(9)	122.4(3)
C(7)-C(8)-Cl(1)	119.6(3)
C(9)-C(8)-Cl(1)	118.0(3)
C(10)-C(9)-C(8)	120.9(3)
C(10)-C(9)-H(9)	119.5
C(8)-C(9)-H(9)	119.5
C(9)-C(10)-C(11)	119.1(3)
C(9)-C(10)-H(10)	120.4
C(11)-C(10)-H(10)	120.4
N(2)-C(11)-C(10)	127.3(3)
N(2)-C(11)-C(6)	113.7(3)
C(10)-C(11)-C(6)	119.0(3)
N(1)-C(12)-H(12A)	109.5
N(1)-C(12)-H(12B)	109.5
H(12A)-C(12)-H(12B)	109.5
N(1)-C(12)-H(12C)	109.5
H(12A)-C(12)-H(12C)	109.5
H(12B)-C(12)-H(12C)	109.5

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for 6-chloro-2-methyl-2*H*-beta-carboline (**158**). The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2\ a^{*2}U^{11} + ... + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
Cl(1)	42(1)	23(1)	36(1)	-8(1)	8(1)	-9(1)
N(1)	31(2)	12(2)	15(2)	-1(1)	5(1)	-5(1)
N(2)	24(2)	15(2)	21(2)	-1(1)	3(1)	3(1)
C(1)	26(2)	14(2)	21(2)	4(2)	8(2)	6(2)
C(2)	18(2)	18(2)	16(2)	0(2)	1(2)	-1(2)
C(3)	25(2)	14(2)	10(2)	0(2)	4(2)	-2(2)

C(4)	24(2)	17(2)	13(2)	3(2)	4(2)	-2(2)
C(5)	22(2)	18(2)	18(2)	2(2)	2(2)	-2(2)
C(6)	27(2)	13(2)	13(2)	4(2)	7(2)	0(2)
C(7)	22(2)	18(2)	17(2)	4(2)	6(2)	0(2)
C(8)	35(2)	17(2)	15(2)	-2(2)	8(2)	-10(2)
C(9)	36(2)	11(2)	24(2)	1(2)	13(2)	1(2)
C(10)	26(2)	18(2)	31(2)	4(2)	9(2)	3(2)
C(11)	25(2)	13(2)	14(2)	4(2)	6(2)	1(2)
C(12)	41(3)	14(2)	24(2)	-3(2)	9(2)	-6(2)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters ($Å^2$x 10^{3}) for 6-chloro-2-methyl-2*H*-beta-carboline (**158**).

x	y	z	U(eq)
H(1)	2501	6071	7833	24
H(2)	1127	5176	6548	22
H(5)	8933	5484	9281	24
H(7)	783	3960	5193	23
H(9)	5402	2655	5391	27
H(10)	8405	3253	6907	30
H(12A)	8182	6390	10208	40
H(12B)	5622	6549	10267	40
H(12C)	6497	6700	8420	40

Table 6. Torsion angles [°] for 6-chloro-2-methyl-2*H*-beta-carboline (**158**).

C(5)-N(1)-C(1)-C(2)	0.3(5)
C(12)-N(1)-C(1)-C(2)	179.9(3)
N(1)-C(1)-C(2)-C(3)	-1.3(5)
C(1)-C(2)-C(3)-C(6)	179.3(4)
C(1)-C(2)-C(3)-C(4)	0.9(5)
C(11)-N(2)-C(4)-C(5)	177.8(4)
C(11)-N(2)-C(4)-C(3)	0.1(4)
C(2)-C(3)-C(4)-N(2)	178.4(3)
C(6)-C(3)-C(4)-N(2)	-0.4(4)

C(2)-C(3)-C(4)-C(5)	0.5(5)
C(6)-C(3)-C(4)-C(5)	-178.4(3)
C(1)-N(1)-C(5)-C(4)	1.1(5)
C(12)-N(1)-C(5)-C(4)	-178.4(3)
N(2)-C(4)-C(5)-N(1)	-179.1(3)
C(3)-C(4)-C(5)-N(1)	-1.5(5)
C(2)-C(3)-C(6)-C(7)	1.2(7)
C(4)-C(3)-C(6)-C(7)	179.7(4)
C(2)-C(3)-C(6)-C(11)	-178.0(4)
C(4)-C(3)-C(6)-C(11)	0.6(3)
C(3)-C(6)-C(7)-C(8)	-179.5(4)
C(11)-C(6)-C(7)-C(8)	-0.4(5)
C(6)-C(7)-C(8)-C(9)	0.6(5)
C(6)-C(7)-C(8)-Cl(1)	-177.6(2)
C(7)-C(8)-C(9)-C(10)	-0.3(6)
Cl(1)-C(8)-C(9)-C(10)	177.9(3)
C(8)-C(9)-C(10)-C(11)	-0.1(5)
C(4)-N(2)-C(11)-C(10)	-179.6(3)
C(4)-N(2)-C(11)-C(6)	0.3(4)
C(9)-C(10)-C(11)-N(2)	-179.9(3)
C(9)-C(10)-C(11)-C(6)	0.2(5)
C(7)-C(6)-C(11)-N(2)	-179.9(3)
C(3)-C(6)-C(11)-N(2)	-0.6(4)
C(7)-C(6)-C(11)-C(10)	0.0(5)
C(3)-C(6)-C(11)-C(10)	179.3(3)

Symmetry transformations used to generate equivalent atoms:

Crystallographic Data for 6-bromo-2-methyl-2*H*-beta-carboline (160)

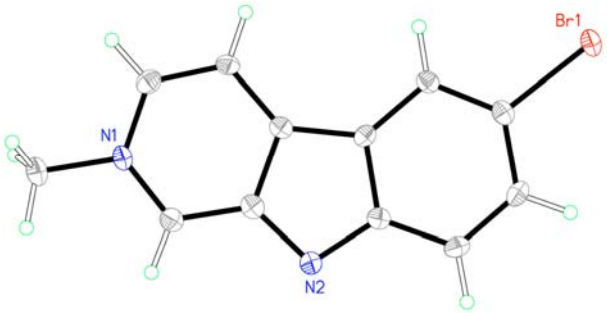


Table 1. Crystal data and structure refinement for 6-bromo-2-methyl-2*H*-beta-carboline (**160**).

Identification code	**160**	
Empirical formula	C12 H9 Br N2	
Formula weight	261.12	
Temperature	100(2) K	
Wavelength	0.71073 Å	
Crystal system	Monoclinic	
Space group	P2(1)/c	
Unit cell dimensions	a = 6.0749(8) Å	α = 90°.
	b = 23.242(3) Å	β = 105.542(11)°.
	c = 7.3601(8) Å	γ = 90°.
Volume	1001.2(2) $Å^3$	
Z	4	
Density (calculated)	1.732 Mg/m^3	
Absorption coefficient	4.068 mm^{-1}	
F(000)	520	
Crystal size	0.39 x 0.19 x 0.15 mm^3	
Theta range for data collection	3.37 to 27.51°.	
Index ranges	-7<=h<=7, -30<=k<=30, -9<=l<=9	
Reflections collected	20538	
Independent reflections	2291 [R(int) = 0.0817]	
Completeness to theta = 27.51°	99.8 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.543 and 0.335	
Refinement method	Full-matrix least-squares on F^2	
Data / restraints / parameters	2291 / 0 / 136	
Goodness-of-fit on F^2	1.189	
Final R indices [I>2sigma(I)]	R1 = 0.0420, wR2 = 0.0698	
R indices (all data)	R1 = 0.0599, wR2 = 0.0755	

Largest diff. peak and hole 0.584 and -0.507 e.Å^{-3}

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters (Å^{2}x 10^3) for 6-bromo-2-methyl-2*H*-beta-carboline (**160**). U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
Br(1)	500(1)	2786(1)	3736(1)	18(1)
N(1)	5742(5)	5843(1)	8675(4)	15(1)
N(2)	7956(5)	4427(1)	7832(4)	16(1)
C(1)	3447(6)	5756(2)	7896(5)	17(1)
C(2)	2635(6)	5235(1)	7105(4)	14(1)
C(3)	4203(6)	4797(2)	7076(4)	13(1)
C(4)	6608(6)	4897(2)	7900(4)	15(1)
C(5)	7312(6)	5431(2)	8714(5)	17(1)
C(6)	4092(6)	4212(1)	6438(4)	13(1)
C(7)	2287(6)	3846(1)	5506(4)	14(1)
C(8)	2847(6)	3294(2)	5102(4)	15(1)
C(9)	5126(6)	3090(2)	5603(5)	17(1)
C(10)	6886(6)	3442(2)	6517(5)	17(1)
C(11)	6414(6)	4014(2)	6939(5)	14(1)
C(12)	6498(6)	6414(2)	9500(5)	20(1)

Table 3. Bond lengths [Å] and angles [°] for 6-bromo-2-methyl-2*H*-beta-carboline (**160**).

Br(1)-C(8)	1.915(3)
N(1)-C(5)	1.346(4)
N(1)-C(1)	1.373(4)
N(1)-C(12)	1.481(4)
N(2)-C(4)	1.374(4)
N(2)-C(11)	1.378(4)
C(1)-C(2)	1.376(5)
C(1)-H(1)	0.9500
C(2)-C(3)	1.399(5)
C(2)-H(2)	0.9500
C(3)-C(6)	1.435(5)

C(3)-C(4)	1.443(5)
C(4)-C(5)	1.395(5)
C(5)-H(5)	0.9500
C(6)-C(7)	1.412(5)
C(6)-C(11)	1.434(4)
C(7)-C(8)	1.381(5)
C(7)-H(7)	0.9500
C(8)-C(9)	1.415(5)
C(9)-C(10)	1.371(5)
C(9)-H(9)	0.9500
C(10)-C(11)	1.413(5)
C(10)-H(10)	0.9500
C(12)-H(12A)	0.9800
C(12)-H(12B)	0.9800
C(12)-H(12C)	0.9800

C(5)-N(1)-C(1)	122.6(3)
C(5)-N(1)-C(12)	119.2(3)
C(1)-N(1)-C(12)	118.1(3)
C(4)-N(2)-C(11)	103.6(3)
N(1)-C(1)-C(2)	121.0(3)
N(1)-C(1)-H(1)	119.5
C(2)-C(1)-H(1)	119.5
C(1)-C(2)-C(3)	118.6(3)
C(1)-C(2)-H(2)	120.7
C(3)-C(2)-H(2)	120.7
C(2)-C(3)-C(6)	136.3(3)
C(2)-C(3)-C(4)	119.6(3)
C(6)-C(3)-C(4)	104.1(3)
N(2)-C(4)-C(5)	127.3(3)
N(2)-C(4)-C(3)	113.8(3)
C(5)-C(4)-C(3)	118.9(3)
N(1)-C(5)-C(4)	119.3(3)
N(1)-C(5)-H(5)	120.3
C(4)-C(5)-H(5)	120.3
C(7)-C(6)-C(11)	121.0(3)
C(7)-C(6)-C(3)	133.8(3)
C(11)-C(6)-C(3)	105.2(3)

C(8)-C(7)-C(6)	117.4(3)
C(8)-C(7)-H(7)	121.3
C(6)-C(7)-H(7)	121.3
C(7)-C(8)-C(9)	122.3(3)
C(7)-C(8)-Br(1)	119.6(3)
C(9)-C(8)-Br(1)	118.0(3)
C(10)-C(9)-C(8)	120.5(3)
C(10)-C(9)-H(9)	119.7
C(8)-C(9)-H(9)	119.7
C(9)-C(10)-C(11)	119.5(3)
C(9)-C(10)-H(10)	120.2
C(11)-C(10)-H(10)	120.2
N(2)-C(11)-C(10)	127.4(3)
N(2)-C(11)-C(6)	113.3(3)
C(10)-C(11)-C(6)	119.2(3)
N(1)-C(12)-H(12A)	109.5
N(1)-C(12)-H(12B)	109.5
H(12A)-C(12)-H(12B)	109.5
N(1)-C(12)-H(12C)	109.5
H(12A)-C(12)-H(12C)	109.5
H(12B)-C(12)-H(12C)	109.5

Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ($Å^2 x\ 10^3$) for 6-bromo-2-methyl-2*H*-beta-carboline (**160**). The anisotropic displacement factor exponent takes the form: $-2\pi^2[\ h^2\ a^{*2}U^{11} + ... + 2\ h\ k\ a^*\ b^*\ U^{12}\ ]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
Br(1)	21(1)	15(1)	18(1)	-3(1)	3(1)	-4(1)
N(1)	18(2)	13(1)	12(1)	-1(1)	3(1)	-3(1)
N(2)	15(2)	15(2)	18(2)	-2(1)	3(1)	0(1)
C(1)	18(2)	17(2)	15(2)	2(1)	5(1)	4(1)
C(2)	12(2)	17(2)	13(2)	2(1)	1(1)	0(1)
C(3)	15(2)	16(2)	9(2)	2(1)	2(1)	-1(1)
C(4)	18(2)	17(2)	9(2)	1(1)	3(1)	-1(1)
C(5)	17(2)	16(2)	16(2)	2(1)	3(1)	-1(1)

C(6)	16(2)	13(2)	11(2)	3(1)	4(1)	2(1)
C(7)	14(2)	15(2)	12(2)	2(1)	2(1)	-1(1)
C(8)	17(2)	15(2)	13(2)	1(1)	4(1)	-3(1)
C(9)	20(2)	13(2)	20(2)	1(1)	8(2)	3(1)
C(10)	14(2)	19(2)	20(2)	3(1)	5(1)	4(1)
C(11)	14(2)	16(2)	14(2)	3(1)	4(1)	0(1)
C(12)	24(2)	17(2)	17(2)	-4(1)	4(2)	-4(2)

Table 5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters (Å^{2}x 10^{3}) for 6-bromo-2-methyl-2*H*-beta-carboline (**160**).

x	y	z	U(eq)
H(1)	2404	6060	7904	20
H(2)	1043	5175	6589	17
H(5)	8884	5504	9290	20
H(7)	746	3975	5170	17
H(9)	5439	2706	5304	21
H(10)	8411	3302	6863	21
H(12A)	8126	6399	10168	30
H(12B)	5616	6520	10386	30
H(12C)	6252	6702	8492	30

Table 6. Torsion angles [°] for 6-bromo-2-methyl-2*H*-beta-carboline (**160**).

C(5)-N(1)-C(1)-C(2)	-0.1(5)
C(12)-N(1)-C(1)-C(2)	179.9(3)
N(1)-C(1)-C(2)-C(3)	-1.2(5)
C(1)-C(2)-C(3)-C(6)	179.0(4)
C(1)-C(2)-C(3)-C(4)	1.1(5)
C(11)-N(2)-C(4)-C(5)	178.1(3)
C(11)-N(2)-C(4)-C(3)	0.4(4)
C(2)-C(3)-C(4)-N(2)	178.1(3)
C(6)-C(3)-C(4)-N(2)	-0.4(4)
C(2)-C(3)-C(4)-C(5)	0.1(5)
C(6)-C(3)-C(4)-C(5)	-178.3(3)

C(1)-N(1)-C(5)-C(4)	1.4(5)
C(12)-N(1)-C(5)-C(4)	-178.6(3)
N(2)-C(4)-C(5)-N(1)	-179.0(3)
C(3)-C(4)-C(5)-N(1)	-1.4(5)
C(2)-C(3)-C(6)-C(7)	2.4(7)
C(4)-C(3)-C(6)-C(7)	-179.5(3)
C(2)-C(3)-C(6)-C(11)	-177.8(4)
C(4)-C(3)-C(6)-C(11)	0.2(3)
C(11)-C(6)-C(7)-C(8)	0.1(5)
C(3)-C(6)-C(7)-C(8)	179.8(3)
C(6)-C(7)-C(8)-C(9)	0.6(5)
C(6)-C(7)-C(8)-Br(1)	-177.1(2)
C(7)-C(8)-C(9)-C(10)	-0.3(5)
Br(1)-C(8)-C(9)-C(10)	177.4(3)
C(8)-C(9)-C(10)-C(11)	-0.7(5)
C(4)-N(2)-C(11)-C(10)	-179.4(3)
C(4)-N(2)-C(11)-C(6)	-0.2(4)
C(9)-C(10)-C(11)-N(2)	-179.5(3)
C(9)-C(10)-C(11)-C(6)	1.3(5)
C(7)-C(6)-C(11)-N(2)	179.7(3)
C(3)-C(6)-C(11)-N(2)	0.0(4)
C(7)-C(6)-C(11)-C(10)	-1.0(5)
C(3)-C(6)-C(11)-C(10)	179.2(3)

Symmetry transformations used to generate equivalent atoms:

Curriculum Vitae

Simone Bonazzi, born July 1st 1982 in Locarno, Switzerland

Education

09.1997 – 06.2001	Maturità federale tipo Biologia–Chimica (BIC), Locarno High school (Liceo Cantonale Locarno), Switzerland
10.2001 – 06.2005	M.Sc. in molecular and biological chemistry, Swiss Federal Institute of Technology of Lausanne (EPFL), Switzerland
07.2004 – 09.2004	Internship: Inpharzam Ricerche SA (Zambon Group), Laboratory of Medicinal Chemistry, Taverne, Switzerland
10.2004 – 02.2005	Semester project in the group of Prof. Dr. *S. Pitsch*, Laboratory of Nucleic Acid Chemistry, Swiss Federal Institute of Technology of Lausanne (EPFL), Switzerland: *Synthesis of RNA Phosphoramidites with 2'O–Amino Linker Modification*
03.2005 – 06.2005	Semester project in the group of Prof. Dr. *K. Severin*, Laboratory of Supramolecular Chemistry, Swiss Federal Institute of Technology of Lausanne (EPFL), Switzerland: *Self Assembly of Metallomacrocyclic π-Ligand/M (M =Rh, Ir, Ru) Complexes Using a Hexadentate Ligand*
08.2005 – 09.2005	Research project in the group of Prof. Dr. *J. K. M. Sanders* and Dr. *S. Otto*, Department of Chemistry, University of Cambridge, England: *Synthesis of Squaramide Based Building Blocks for Dynamic Combinatorial Libraries*
10.2005 – 03.2006	Diploma Thesis under the supervision of Prof. Dr. *E. M. Carreira* and Dr. *K. Gademann*, Laboratorium für Organische Chemie, Swiss Federal Institute of Technology of Zürich (ETHZ), Switzerland: *Synthesis of C(1)–C(11) Fragment of Anguinomycin and Derivatives as Inhibitors for Nucleocytoplasmic Transport*
04.2006 – 05.2009	Ph.D. studies in organic chemistry under the supervision of Prof. Dr. *K. Gademann* and Prof. Dr. *E. M. Carreira*, Laboratorium für Organische Chemie, Swiss Federal Institute of Technology of Zürich (ETHZ), Switzerland and Chemical Synthesis Laboratory, Swiss Federal Institute of Technology of Lausanne (EPFL), Switzerland: *Total Syntheses of Anguinomycins C & D, Synthetic Studies on Sporolides and Preparation of Eudistomin Derivatives: Biological Evaluation Against Cancer and Malaria*

Teaching Experience

During my Ph.D. thesis, I was responsible for the supervision and training of three diploma students, two semester students, one apprentice and one internship student. I was teaching assistant for three years in master courses (*Structure and reactivity* and *Target synthesis*) and in bachelor courses (*Fonctions et réactions organiques II*).

Zürich, May 2009 Simone Bonazzi