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Total Synthesis of Cyclopropyl-Epothilone B Analogs

and

Studies Towards the Total Synthesis of Michaolide E

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Presented by

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„Wenn man in der Mitte des Pferderennens merkt,

dass man auf einem Esel sitzt, da muss man weitermachen!“

Bernd Stromberg, Capitol, Abteilungsleiter Schadensregulierung L-Z

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Curriculum Vitae

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10.2003-06.2008	MSc in Interdisciplinary Sciences, ETH Zürich, Switzerland
08.2000-08.2003	Matura, TSME Romanshorn, Switzerland
08.1998-08.2000	Commercial apprenticeship in school of administration, Kreuzlingen, Switzerland

Publications

- Metri, P., Schiess, R., Prasad, K. "Total Synthesis of (-)-Bengamide E" *Chemistry An Asian Journal* **2012**,
- Pfeiffer, B., Gaugaz, F. Z., Schiess, R., Altmann, K.-H. "Epothilones as Lead Structures for New Anticancer Drugs" *Drug Discovery from Natural Products* **2012**, 339.
- Schiess, R., Gertsch, J., Schweizer, B. W., Altmann, K.-H. "Stereoselective Synthesis of 12,13-Cyclopropyl-Epothilone B and Side-Chain-Modified Variants" *Org. Lett.* **2011**, *13*, 1436.
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Abstract

Epothilones (Figure 1) are naturally occuring microtubule-stabilizers that inhibit the growth of human cancer cells at nM or even sub-nM concentrations. Based on their attractive preclinical profile the epothilones have served as important lead structures in the search for improved anticancer drugs. These efforts have led to the identification of nine epothilones that were investigated in clinical trials, with the approval of the epothilone B lactam ixabepilone (Ixempra®) for the treatment of breast cancer as the most tangible result.

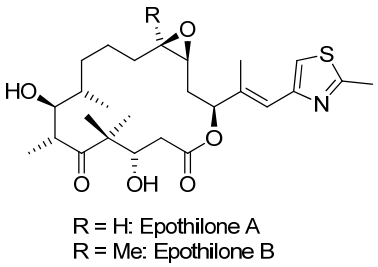


Figure 1: Naturally occurring epothilones; Epo A and Epo B.

As for other cytotoxic anticancer agents epothilones do not discriminate between cancer and normal cells, thus, increasing their tumor cell selectivity would significantly improve their therapeutic potential. Before this background it was one of the objectives of this PhD thesis to provide new functionalized epothilone analogs for the construction of epothilone-based antibody-drug conjugates (ADC) with enhanced tumor selectivity. In order to ensure maximum potency and metabolic stability, these analogs were derived from 12,13-cyclopropyl Epo B as the parent structure, with modifications easily addressable for antibody attachment located in the heterocyclic part of the C15 side chain (Figure 2).

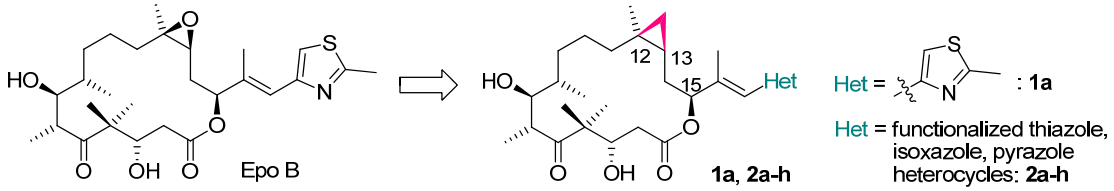
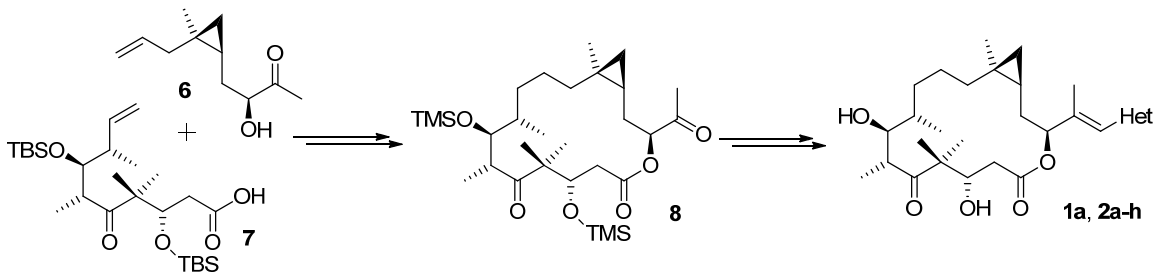


Figure 2: Target structures **1a**, **2a-h** as the active drug cargo for the construction of ACD's.

As a first step towards the development of cyclopropyl-epothilone-based ADC's an efficient synthetic route to cyclopropyl-Epo B (**1a**) and side chain-modified analogs **2a-h** was established. Target structures **1a** and **2a-h** were accessed through a convergent approach, which comprised the assembly of the key building blocks **6** and **7** *via* an esterification/ring-closing metathesis sequence to obtain the macrocyclic core with a truncated side that was amenable to elaboration into cyclopropyl-epothilone B (**1a**) (thus completing the first total

synthesis of this analog) and side chain-modified variants (**2a-h**) by *Wittig*-type chemistry (Scheme 1). All target structures could thus be accessed from a single advanced intermediate.



Scheme 1: Convergent synthesis of target structures **1a** and **2a-h**.

Cyclopropyl-epothilone B (**1a**) showed similar antiproliferative activity against human cancer cell lines as natural Epo B. Likewise, several functionalized derivatives **2a-h** showed IC_{50} values for the *in vitro* inhibition of human cancer cell growth in the low nM range, which makes them potential candidates for the construction of epothilone-based ADC.

In addition to the synthesis of **1a** and **2a-h**, an optimized synthesis was elaborated for the potent hypermodified Epo A analog **3a**, which had been previously identified in the group and which could potentially be conjugated to tumor-targeting antibodies through the selective acylation of the C7 hydroxy group. Key steps of the optimized route towards **3a** are a *syn*-aldol addition to install the stereocenters at C6 and C7, *Julia-Kocienski* olefination to establish the C10/C11 bond, ring-closure by *Shiina* macrocyclization, and a late stage introduction of the heterocycles by means of *Wittig* olefination (Figure 3).

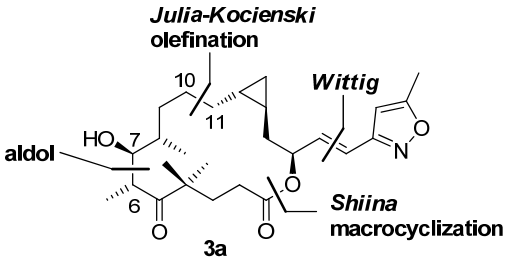
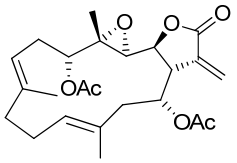


Figure 3: Structure of hypermodified Epo A (**3a**) and key retrosynthetic disconnections.

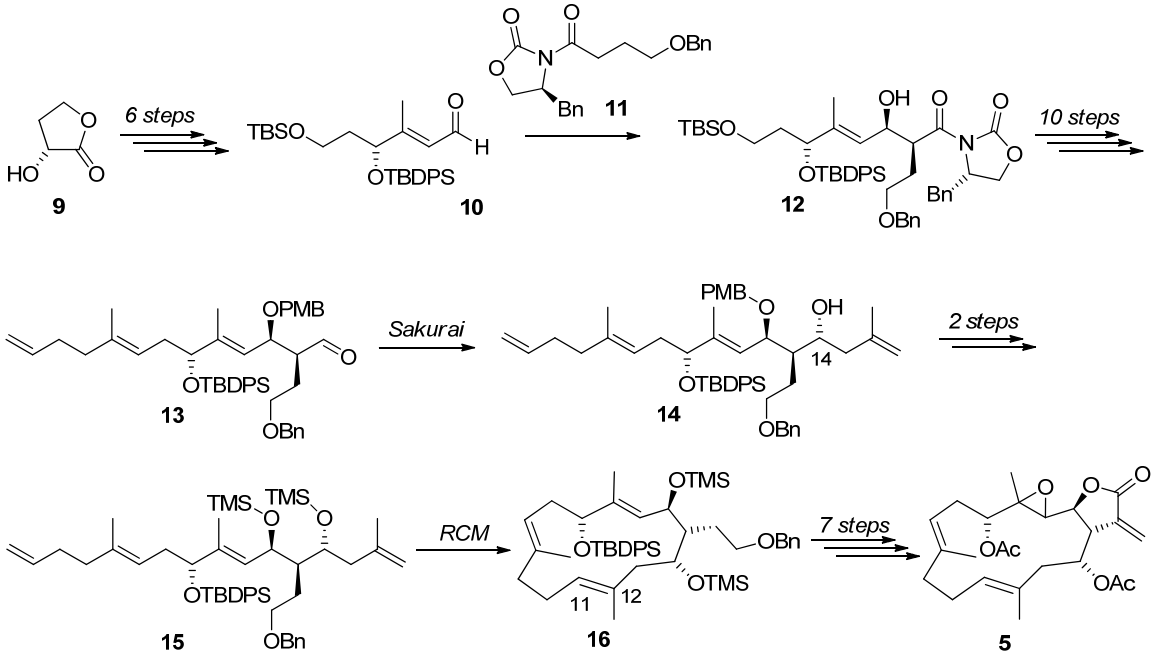
The second part of this thesis describes studies towards the total synthesis of michaolide E (**4**) (Figure 4), a new highly cytotoxic member of the cembranolide family of natural products. **4** has been isolated from the soft coral *Lobophytum michaelae* and demonstrated to inhibit the growth of the human colon adenocarcinoma cell line HT-29 with an IC_{50} value of 115 nM and the mouse lymphocytic leukemia line P-388 with an IC_{50} of 16 nM.



Michaolide E (**4**)

Figure 4: Structure of michaolide E (**4**).

To date, no total synthesis of michaolide E (**4**) has been reported and no structure-activity studies been described. It was an objective of this thesis to establish an efficient and stable synthesis of this interesting natural product in order to provide a starting point for the exploration of michaolide E (**4**) as a potential lead structure for anticancer drug discovery. The synthesis discussed in this thesis was based on the following key transformations (Scheme 2): (1) An *Evans syn*-aldol reaction between aldehyde **10** and imide **11** to provide the desired aldol product **12** as a single isomer; (2) a highly selective *Sakurai* addition with aldehyde **13** to establish the C14 stereocenter; (3) ring-closing metathesis (RCM) to construct the 14-membered macrocycle **16**, and regioselective lactonization (Scheme 2).



Scheme 2: Synthesis towards michaolide E (**4**).

Unfortunately, the spectral data of the product **5** obtained after directed epoxidation and final methenylation α to the lactone carbonyl did not correspond with those reported for natural michaolide E (**4**). At this point we assume that this is a consequence of the formation of the wrong epoxide isomer; however, based on the currently available data it cannot be excluded that the structure of the natural product may have been misassigned.

Zusammenfassung

Die Epothilone (Abbildung 1) sind Naturstoffe mit ausgeprägten Effekten auf das zelluläre Mikrotubuligerüst und einer daraus resultierenen potenten Hemmwirkung auf das Wachstum humaner Krebszellen und Tumore. Aufgrund ihrer interessanten biologischen Aktivität wurden die Epothilone zu vielversprechenden Leistrukturen auf dem Gebiet der Krebsmittelforschung und bis zum heutigen Tag wurden neun Epothilone in klinischen Studien geprüft. Eine dieser Verbindungen, das Epothilon B Analogon Ixabepilone (Ixempra®), wurde 2007 für die Behandlung von Brustkrebs zugelassen.

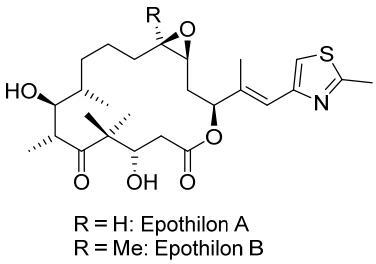


Abbildung 1: Natürlich vorkommende Epothilone; Epo A und Epo B.

Grundsätzlich führt jedoch die fehlende Selektivität der Epothilone gegenüber Krebszellen und die damit verbundenen unerwünschten Nebenwirkungen zu Limitierungen bei deren therapeutischer Anwendung. Eine Möglichkeit dieses Problem zu lösen besteht im Versuch einer zielgerichteten Wirkstoffabgabe im Tumor, wie sie z. B. mittels der Anwendung von Antikörper-Wirkstoff Konjugaten erreicht werden kann. In diesem Kontext ist es ein Ziel dieser Doktorarbeit neue funktionalisierte Epothilon-Analoga herzustellen, welche die Herstellung von Antikörper-Wirkstoff Konjugaten erlauben sollten. Um eine hohe Wirksamkeit und gute metabolische Stabilität dieser Analoga zu gewährleisten, wurden diese als Derivate des 12,13-Cyclopropyl Epo B konzipiert, wobei eine leichte Verknüpfbarkeit mit einem Antikörper durch eine Funktionalisierung der Heterozyklen in der Seitenkette erreicht werden sollte (Abbildung 2).

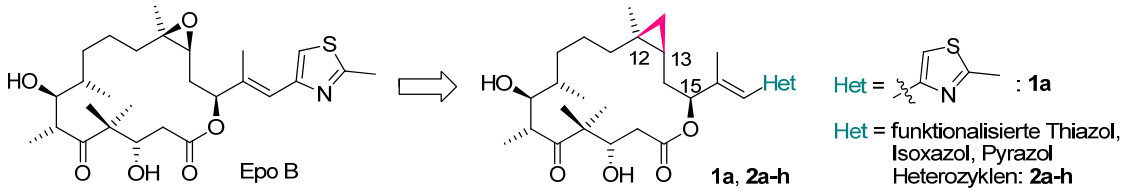
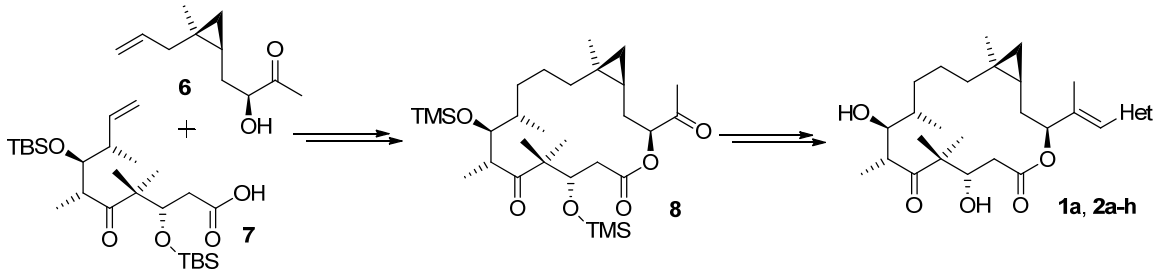


Abbildung 2: Zielstrukturen **1a** und **2a-h**, welche als Wirkstoff für die Bildung von Antikörper-Wirkstoff Konjugaten dienen.

Um eine praktische Grundlage für die Herstellung der entsprechenen Antikörper-Wirkstoff Konjugate zu schaffen, wurde zuerst eine effiziente Synthese des Cyclopropyl-Epo B (**1a**) und entsprechender seitenkettenmodifizierter Analoga **2a-h** ausgearbeitet. Diese Zielstrukturen wurden über eine konvergente Synthese hergestellt, welche auf der Veresterung des sekundären Alkohols (**6**) und der Säure (**7**) basiert (Schema 1) und in einer nachfolgenden Macrozyklisierung über eine Ringschluss-Metathese zur Bildung des makrozyklischen Grundgerüst beruht. Das so erhaltene makrozyklische Methylketon wurde dann mittels einer *Wittig* Reaktion in Cyclopropyl-Epo B oder durch *Wittig*- wie *HWE* Reaktionen in die entsprechenden seitenkettenmodifizierten Derivate überführt. Diese Strategie führte zur erstmaligen Totalsynthese des Cyclopropyl-Epo B (**1a**).



Schema 1: Konvergente Synthese von Zielstrukturen **1a** und **2a-h**.

Cyclopropyl-Epo B (**1a**) zeigt eine ähnlich potente antiproliferative Aktivität gegenüber Krebszellen wie das natürliche Epo B. Die verschiedenen funktionalisierten Derivate **2a-h** hemmen das Wachstum humaner Krebszellen ebenfalls mit IC_{50} Werten im tiefen nanomolaren Bereich und kommen somit als mögliche Wirkstoffkomponenten für Antikörper-Wirkstoff Konjugate in Betracht.

Zusätzlich zur Synthese der obigen Cyclopropyl-Epo B Analoga wurde eine optimierte Synthese des hochpotenten, hypermodifizierten Epo A Analogons **3a** (Abbildung 3) ausgearbeitet, welches bereits früher in der Forschungsgruppe untersucht worden war. Analogon **3a** kann über die sekundäre Alkoholgruppierung an C7 mit einem Linker modifizert und dann mit Antikörpern verbunden werden. Die Schlüsselschritte der optimierten Synthese von **3a** sind eine *syn*-Aldol Addition zum Aufbau der Stereozentren an C6 und C7, eine *Julia-Kocienski* Olefinierung zur Bildung der C10/C11 Bindung, ein Ringschluss über eine *Shiina* Makrolaktonisierung und die Einführung der Heterozyklen über eine *Wittig* Reaktion am Schluss der Synthese.

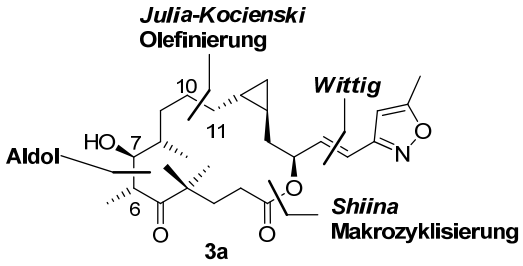


Abbildung 3: Struktur von hypermodifiziertem Epo A (**3a**) und Schlüsselschritte der Retrosynthese.

Der zweite Teil dieser Doktorarbeit beschreibt erste Studien zur Totalsynthese von Michaolid E (**4**) (Abbildung 4), einem neuen cytotoxischen Naturstoff aus der Familie der Cembranolide. **4** wurde aus der Weichkoralle *Lobophytum michaelae* isoliert und hemmt das Wachstum der humanen Kolonkarzinom Krebszelllinie HT-29 und der murinen Lymphknoten Krebszelllinie P-388 IC_{50} Werten von 115 nM bzw. 16 nM.

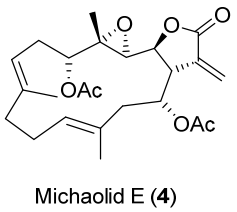
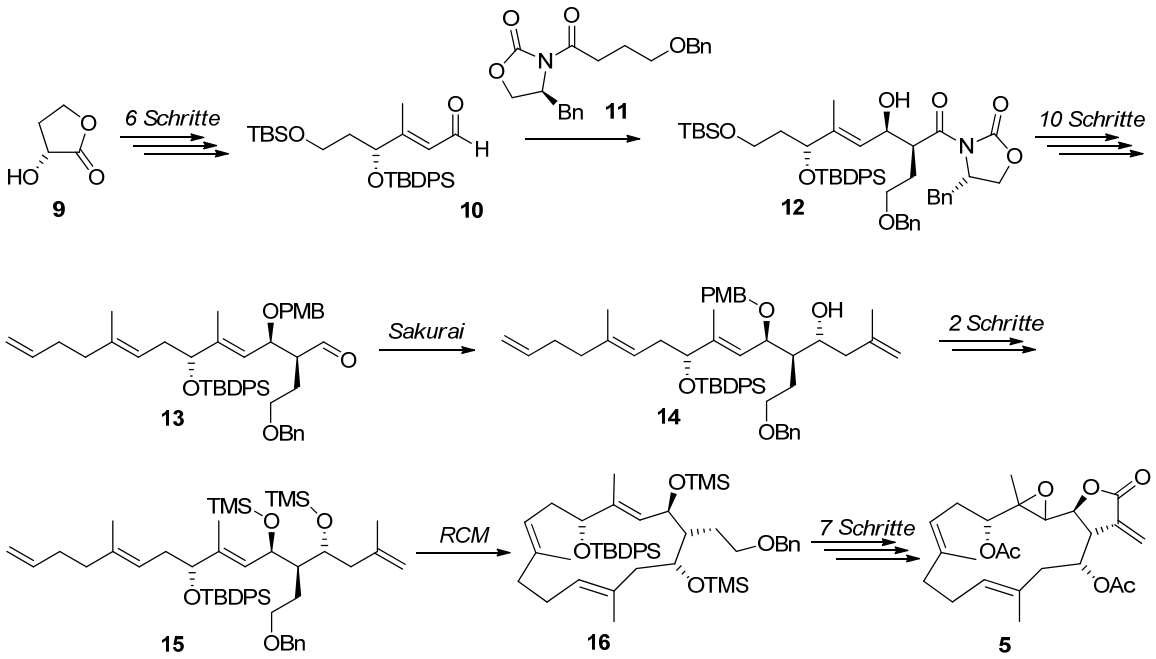


Abbildung 4: Struktur von Michaolid E (**4**).

Bis heute wurde keine Totalsynthese von Michaolid E (**4**) veröffentlicht und es wurden keine SAR Studien beschrieben. Es war ein weiteres Ziel dieser Doktorarbeit eine effiziente und stabile Synthese dieses interessanten Naturstoffs auszuarbeiten, welche als Basis für die Erforschung von Michaolid E (**4**) als Leitstruktur für die Wirkstoffforschung gegen Krebs dienen soll. Die in dieser Arbeit beschriebene Synthese umfasst folgende wichtige Transformationen (Schema 2): (1) Eine *Evans syn*-Aldol Addition zwischen dem α-chiralen Aldehyd **10** und dem Imid **11** lieferte das gewünschte Aldolprodukt **12** als einziges Isomer; (2) eine selektive *Sakurai* Addition an Aldehyd **13** ermöglichte den Aufbau des Stereozentrums an C14; (3) eine Ringschluss-Metathese zur Bildung des Makrozyklus **16**; und (4) eine regioselektive Lactonisierung (Schema 2).



Schema 2: Synthese von Michaolide E (**4**).

Leider stimmten die spektroskopischen Daten des Produkts **5**, welches nach der gerichteten Epoxidierung und abschliessenden Methenylierung α zur Carbonylgruppe des Lactonrings erhalten wurde, nicht mit den Daten für das natürliche Michaolid E (**4**) überein. Wir nehmen zum jetzigen Zeitpunkt an, dass die Ursache für diese Abweichung auf die Bildung des unerwünschten Epoxidisomers zurückzuführen ist. Jedoch kann aufgrund der vorliegenden Daten ein Fehler in der Strukturzuordnung des Naturstoffs nicht mit Sicherheit ausgeschlossen werden.

Abbreviations

ADC	Antibody-drug conjugate
Ar	Aryl
BAIB	Bis(acetoxy)iodobenzene
Bn	Benzyl
CBS	*Corey-Bakshi-Shibata* catalyst
CPS	Cerium(IV)sulfate-phosphomolybdic acid
CSA	Camphorsulfonic acid
DCC	Dicyclohexylcarbodiimide
dr	Diastereomeric ratio
DEAD	Diethyl azodicarboxylate
DIBAL	Diisobutylaluminum hydride
DIPEA	*N*,*N*-Diisopropyl ethyl amine
DMAP	4-(Dimethylamino)pyridine
DMDO	Dimethyl dioxirane
DMF	Dimethylformamide
DMP	Dess-Martinperiodinane
DMSO	Dimethyl sulfoxide
DPPA	Diphenylphosphoryl azide
EDCI	1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide
Epo	Epothilone
EtOAc	Ethyl acetate
FDA	Food and Drug Administration
HF	Hydrofluoric acid
HMPA	Hexamethylphosphoramide
HOBt	Hydroxybenzotriazole
IC_{50}	Half maximal inhibitory concentration
KHMDS	Potassium hexamethyldisilazide
LRMS	Low resolution mass spectrometry
LDA	Lithium diisopropylamide
LiHMDS	Lithium hexamethyldisilazide
m-CPBA	*m*-Chloroperoxybenzoic acid
MCDI	1-Cyclohexl-3-(2-morpholinmethyl)carbodiimide metho-*p*-toluenesulfonate

MeCN	Acetonitrile
MeOH	Methanol
MNBA	2-Methyl-6-nitrobenzoic anhydride
MSA	Microtubule-stabilizing agent
MsCl	Methanesulfonyl chloride
MTBE	Methyl *tert*-butyl ether
n-BuLi	*n*-Butyllithium
NaH	Sodium hydride
NaHMDS	Sodium hexamethyldisilazide
NMO	*N*-methyl morpholine
PDC	Pyridinium dichromate
PMB	*p*-Methoxybenzyl ether
p-TsOH	*p*-Toluenesulfonic acid
py	Pyridine
RCM	Ring-closing metathesis
RCAM	Ring-closing alkyne metathesis
rt	Room temperature
SAR	Structure-activity relationship
SM	Starting material
TBAI	Tetrabutylammonium iodide
TBS	*tert*-Butyldimethylsilyl
t-BuOH	*tert*-Butanol
t-BuOK	Potassium *tert*-butoxide
TEMPO	2,2,6,6-Tetramethylpiperidinyloxyl
Tf	Trifluoromethanesulfonyl (triflyl)
THF	Tetrahydrofuran
TLC	Thin layer chromatography
TIPS	Triisopropylsilane
TPAP	Tetrapropylammonium perruthenate
TrisNHNH$_2$	2,4,6-Triisopropylbenzenesulfonohydrazide

1

1 Total Synthesis of CP-Epo B Analogs and SAR Studies

1.1 Introduction

1.1.1 Microtubule-Stabilizing Agents as Anticancer Drugs

Microtubules are an integral part of the cytoskeleton and as such they play a central role in the process of separating duplicated chromosomes before cell division. As a consequence, the interference with microtubule function leads to inhibition of cell division and proliferation, which has made the tubulin/microtubule system an important and also successful target for anticancer drugs.[1] Depending on the molecular mechanism by which such compounds interfere with the tubulin/microtubule system, they can be grouped into two distinct classes. Either they inhibit the assembly of tubulin heterodimers into microtubule polymers (tubulin polymerization inhibitors) or they stabilize pre-existing microtubules under otherwise non-stabilizing conditions (microtubule stabilizers).[2]

Tubulin polymerization inhibitors such as the vinca alkaloids[3],[4] have been employed in cancer therapy for half a century while the clinical history of agents that promote tubulin polymerization and stabilize microtubules dates back only to 1993, when the natural product taxol (**17**) (paclitaxel; Taxol®) (Figure 5) was introduced for the treatment of breast, ovarian, and lung cancer.[5],[6],[7] Three years later followed the FDA approval of the semisynthetic taxol analog docetaxel (Taxotere®) .

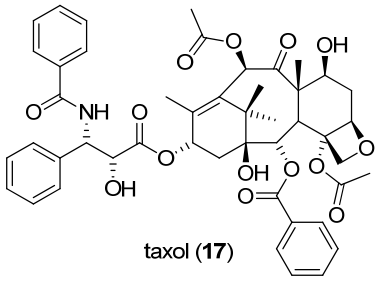


Figure 5: Structure of taxol (**17**).

Over the last two decades a variety of substances, both in the field of depolymerizing and polymerizing compounds have been identified. New structures for tubulin depolymerization such as combretastatin (**18**) or the cryptophycins (**19**)[2] (Figure 6) and a number of microtubule-stabilizing agents as dictyostatin (**20**),[9] discodermolide (**21**),[8] eleutherobin (**22**),[9] laulimalide (**23**),[10] peloruside A (**24**),[11] zampanolide (**25**)[12] (Figure 7) and the epothilones have been discovered.[13]

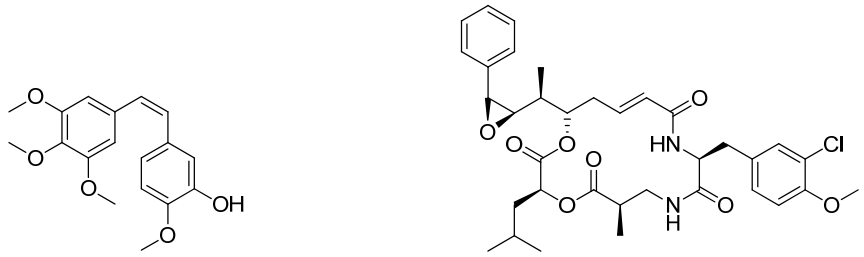


Figure 6: Examples of tubulin polymerization inhibitors.

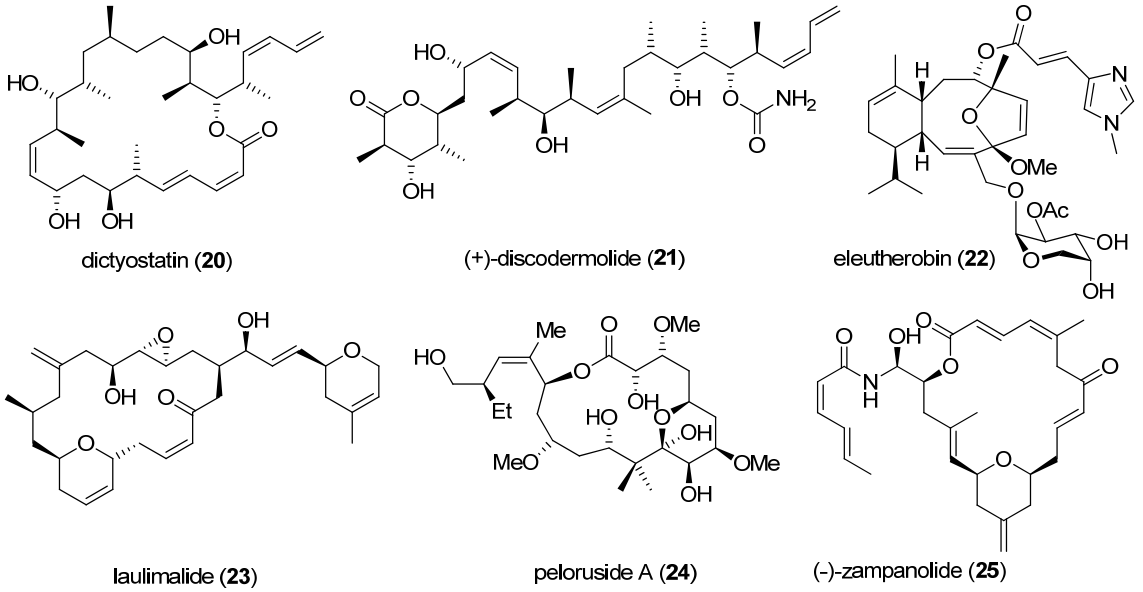


Figure 7: Examples of microtubule-stabilizing agents.

1.1.2 Microtubule Structure and Function

As indicated above, the biological effects of epothilones are based on their ability to bind to microtubules. Thereby they alter the intrinsic stability and the dynamic properties of the microtubules.[13] Microtubules are one of the major components of the cytoskeleton and as such they play an important role in the development and maintenance of cell shape, in the intracellular transport of vehicles, mitochondria and other components.[14] In the context of cell division and proliferation, they are of critical importance for the segregation of the sister chromatids, which has to precede cyctokinesis, i. e. the final division of the cell into two new daughter cells.[15]

Microtubules are polymers of α/β tubulin heterodimers, which are arranged as hollow cylinders composed of 13 protofilaments, i.e. linear strings of head-to-tail arranged α- and β-tubulin subunits (Figure 8). A characteristic feature of microtubules is their ability to lengthen and shorten by addition or loss of α/β tubulin from the microtubule ends. This process is

referred to as "dynamic instability".[1],[16],[17] The second dynamic process affecting microtubule dynamics is called "treadmilling".[18],[19],[20] It constitutes the migration of tubulin subunits from the plus end to the minus end of the polymer. *In vitro*, the growth and shrinkage can take place at both ends of the microtubule cylinder by the addition or loss of α/β-tubulin heterodimers. The plus-end, where β-tubulin is exposed to solvent, is more dynamic than the minus-end terminating with α-tubulin.[1],[16],[21]

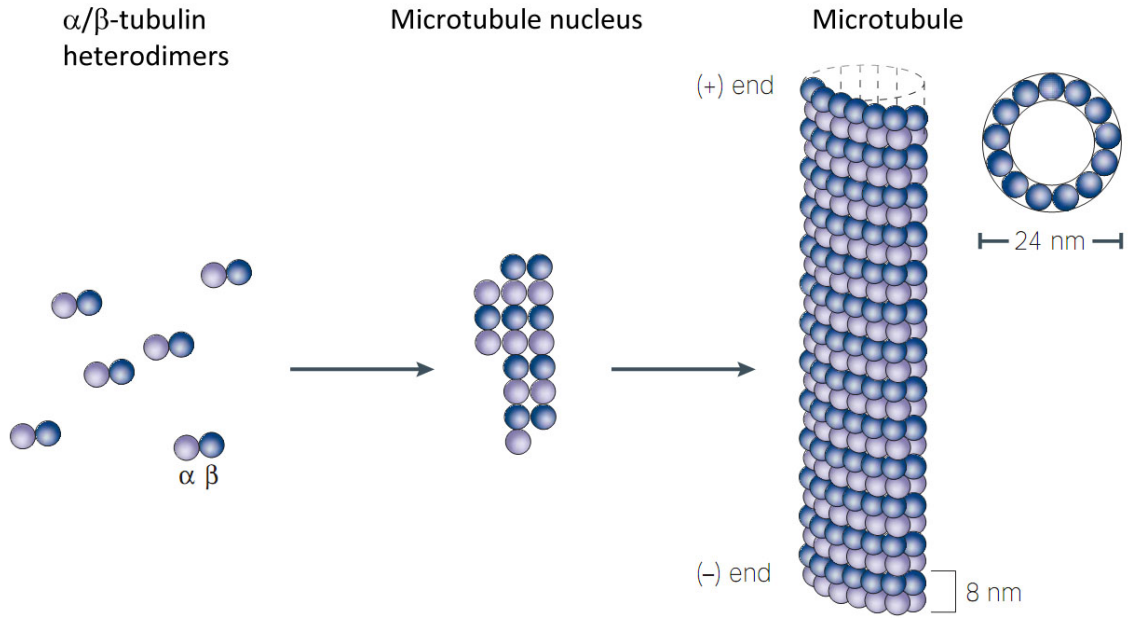


Figure 8: Polymerization of α/β-tubulin subunits into microtubules and the structure of microtubules.

In contrast, in cells, the minus-end is anchored to the microtubule-organizing center, from which the plus-end is able to grow and shrink.[16] In cell division, the separation of the chromatids proceeds with microtubules, which are emanating from the two spindle poles (Figure 9, prometaphase). The microtubules are attached to the kinetochors of the chromosomes and after alignment of the chromosomes in the center of the cell (Figure 9, metaphase), the chromatids separate in anaphase and migrate towards the opposite poles of the cell guided by microtubules.[15] Thus, microtubule dynamics are fundamental for the proper assembly of the mitotic spindle and for the movement of the sister chromatids to the spindle poles (Figure 9, anaphase). It is therefore not of a surprise that agents interfering with microtubule dynamics have a profound effect on cell division. By suppressing microtubule dynamics the spindle is no longer capable of forming properly and the cell cycle cannot progress from metaphase to anaphase.[22] As a consequence, the cell cycle is blocked and apoptosis is induced.[23]

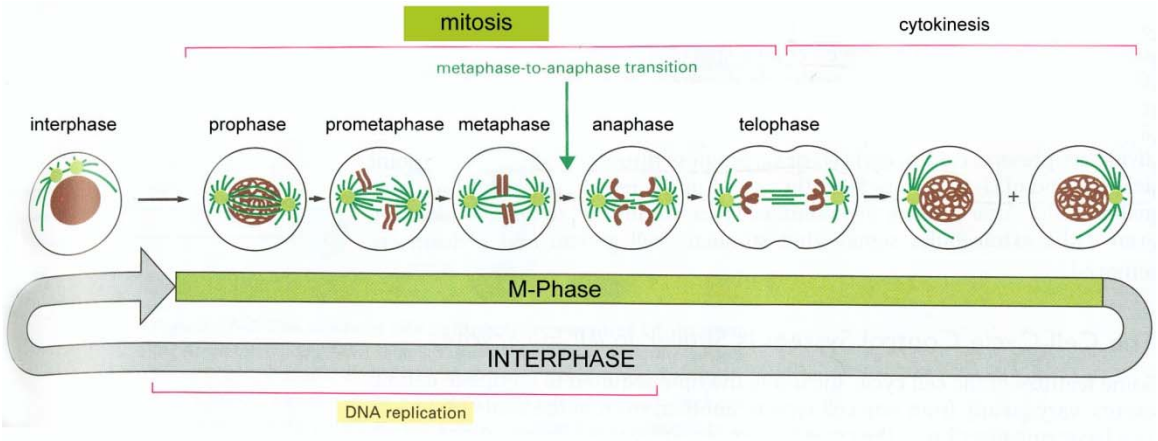


Figure 9: Cell cycle.[24]

1.2 Epothilones

1.2.1 Discovery and *in vitro* Activities

Epothilones are natural products first isolated in 1987 from the myxobacterium *Sorangium cellulosum Sc 90* (collected at the banks of the river Zambesi in Southern Africa) by *Reichenbach* and *Höfle* at the "Gesellschaft für Biotechnologische Forschung (GBF)" in Braunschweig, Germany (now called the *Helmholtz* Centre for Infection Research).[25] Based on an activity-guided fractionation process, epothilone A (Epo A) and B (Epo B) (Figure 10) were isolated in the context of an antifungal screening program. Their name reflects their basic structural features, as they contain an epoxide moiety, a thiazole ring and a ketone functionality.[26]

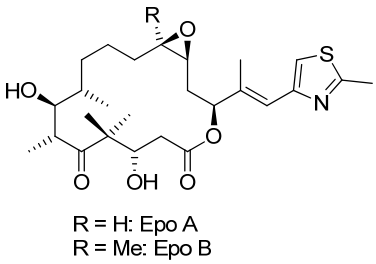


Figure 10: Structures of Epo A and B.

The mode of action of the epothilones was unraveled only eight years after the compounds' original discovery by *Bollag et al.*, who demonstrated that they were microtubule-stabilizing agents (MSA). Intriguingly at the time, Epo B was found to be an even more active tubulin-polymerizing agent than the prototypical MSA taxol and it was able to displace taxol from microtubules, thus indicating that both compounds bind to the same site on β-tubulin.[13]

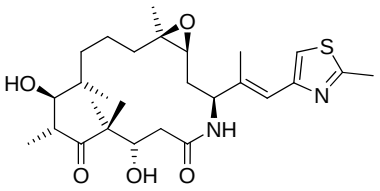
Moreover, in contrast to taxol (**17**), Epo A and B showed low susceptibility to P-glycoprotein-mediated drug efflux and, as a consequence, they also inhibited the growth of multidrug-resistant cancer cell lines with near to full activity.[13],[27],[28],[29] Subsequently, the compounds were also shown to remain unaffected by tubulin mutations that render taxol inactive.[30] IC_{50} values for the *in vitro* cancer cell growth inhibition are in the single-digit nanomolar range for Epo A, a potency comparable to that of taxol, whereas IC_{50} values for Epo B can be in the subnanomolar range. In general, the activity of Epo B is ca. ten times higher than that of Epo A (Table 1).[31]

Table 1: Inhibition of the growth of human cancer cell lines.[31]

Cell line	**IC_{50} [nM]**		
	Epo A	**Epo B**	**Taxol**
HCT-116 (colon)	2.51	0.32	2.79
A549 (lung)	2.67	0.23	3.19
MCF-7 (breast)	1.49	0.18	1.80
NCI/ADR[a,b]	27.5	2.92	9105
KB-31 (cervix)	2.10	0.19	2.31
KB-8511[a,c]	1.90	0.19	533

[a] Multidrug-resistant cell line. [b] Multiple resistance mechanisms/MDR.
[c] P-gp overexpression/MDR

The epothilones possess better water-solubility than taxol (**17**),[32] which should in principle allow the use of less problematic clinical formulation vehicles than those required for taxol;[7] taxol is formulated with the surfactant Cremophor EL which is responsible for anaphylactic reactions.[33] Over the last 14 years several epothilone-type agents have entered clinical trials in humans. Epo B itself was developed by Novartis, but the compound was abandoned in 2010 after a Phase III trial in ovarian cancer had not demonstrated superiority over standard of care.[34] In contrast, the epothilone B lactam ixabepilone (**26**) (BMS-247550; Ixempra®) was approved by the FDA for breast cancer treatment in 2007.[35]

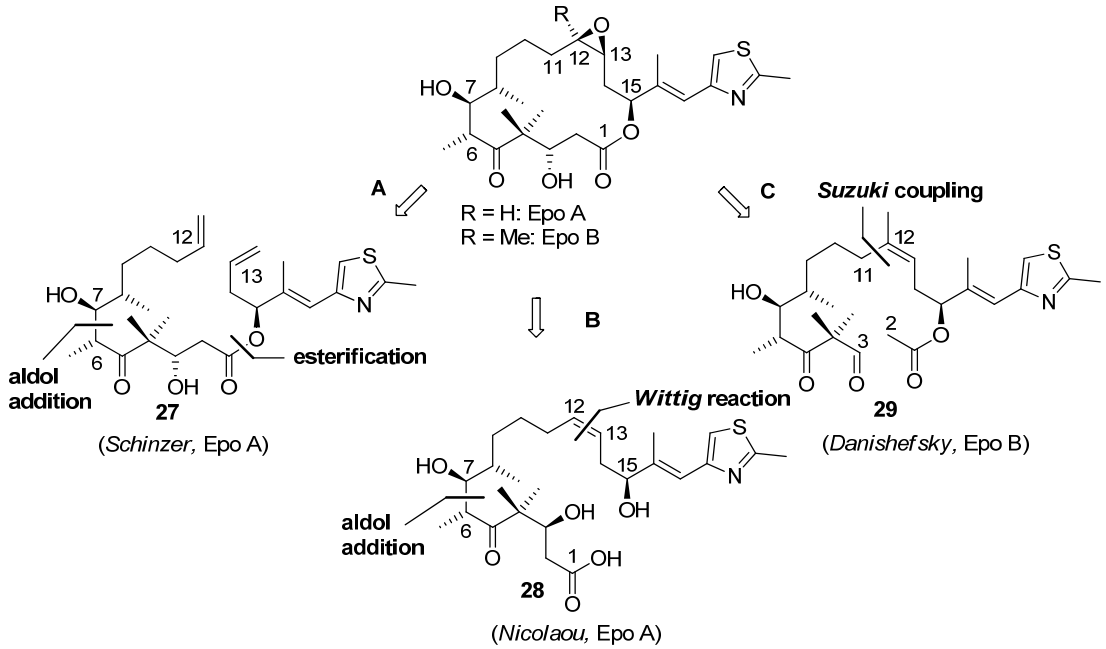


Ixabepilone (**26**)

Figure 11: Structure of ixabepilone (**26**); Ixempra®.

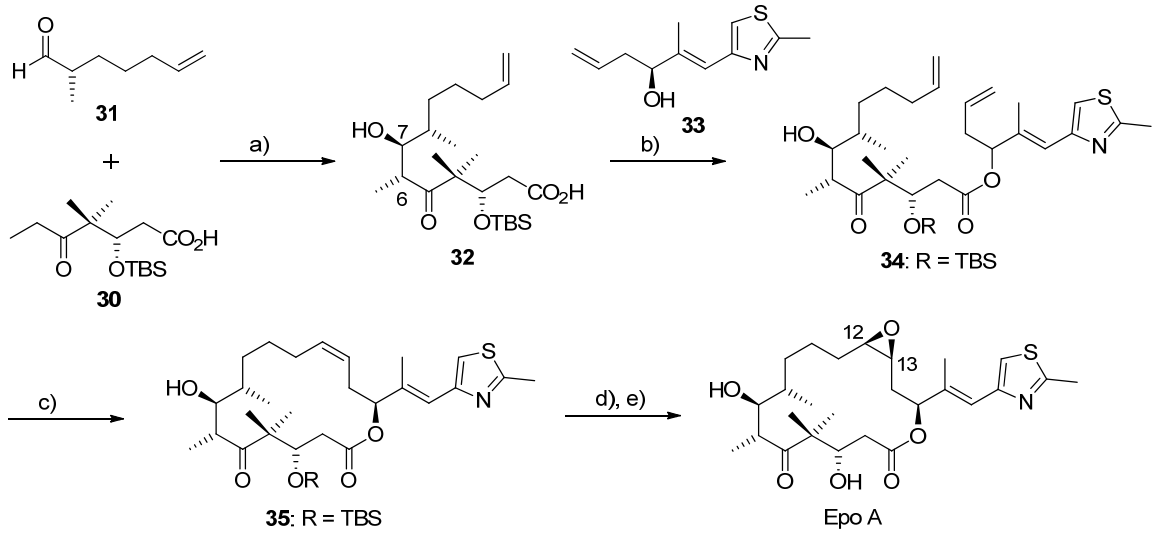
1.2.2 Syntheses of Natural Epothilones

Only a year after the absolute configuration of Epo A and B had been disclosed by *Höfle* and *co*-workers[32] the first total syntheses of Epo A and B were published at the end of 1996 and early 1997 by the groups of *Danishefsky*,[36] *Nicolaou* [37],[38],[39],[40] and *Schinzer*,[41] respectively. Scheme 3 shows the three different approaches to ring closure and other key steps that are characteristic for these total syntheses. The approaches of *Schinzer* (A) and *Nicolaou* (B) are similar in so far as they both use an aldol addition between an α-chiral aldehyde and an ethyl ketone to create stereocenters C6 and C7. *Schinzer* chose RCM (ring closing metathesis) for C12/C13 double bond formation, while *Nicolaou* made use of a *Wittig* reaction to create the (*Z*)-olefin and employed macrolactonization of seco acid **28** to close the macrocycle. *Danishefky* (C), however, used a *Suzuki* coupling to create the C11-C12 carbon-carbon bond and a surprisingly selective macroaldolization for ring-closure. Ring-closure was followed by final epoxidation of the C12/C13 *cis*-double bond to afford the final products Epo A and B.



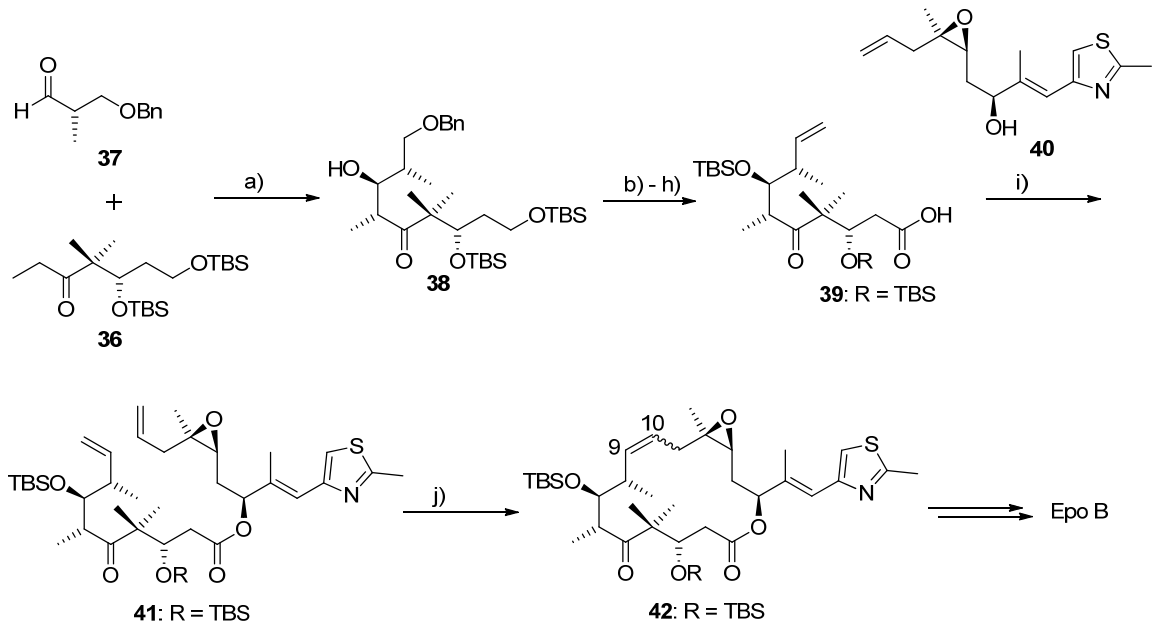
Scheme 3: Early Retrosyntheses of Epo A by *Schinzer* and *Nicolaou*, and Epo B by *Danishefsky*.

In addition to the macrolactonization approach, *Nicolaou et al*. published a synthesis of Epo A where the macrocycle was closed at C12/C13 by means of RCM (Scheme 4).[42] The stereocenters at C6 and C7 were installed *via* aldol addition of the dianion of carboxylic acid **30** and α-chiral aldehyde **31** giving acid **32** as the major isomer (dr 2:1). Subsequent esterification with alcohol **33** under *Steglich*[43] conditions yielded diene **34** and the stage was set for the crucial ring-closing reaction. The macrocycle **35** formed in satisfying 50% yield together with 35% of the undesired *E*-isomer. TBS-deprotection was carried out under acidic conditions and final epoxidation with *m*-CPBA afforded Epo A in 55% yield together with 20% of the 12α,13α-epoxide.



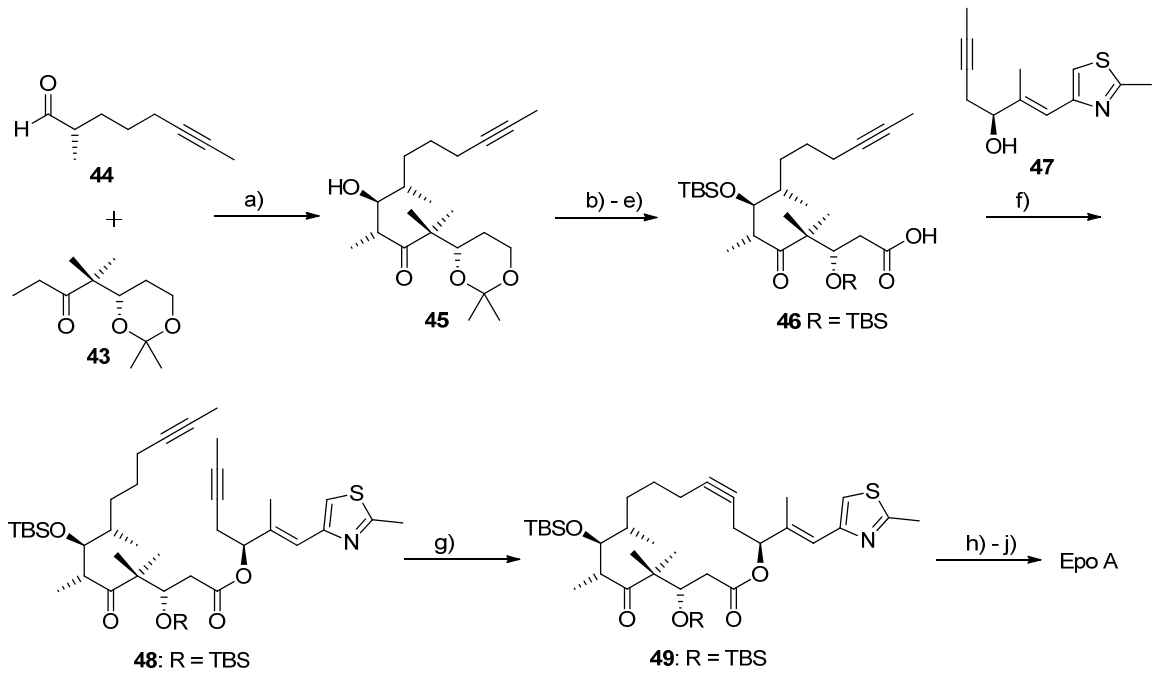
Scheme 4: a) LDA, THF, -78 °C to -40 °C, then **31**, -78 °C to -40 °C, dr 2:1; b) DCC, DMAP, toluene, **33**, 45% over two steps; c) *Grubbs* I (15 mol%), CH_2Cl_2, 50%; d) TFA, CH_2Cl_2, 0 °C, 98%; e) *m*-CPBA, benzene, 0 °C, 55%.

In the following years a number of other approaches to form the macrocycle were reported and only a few of them will be discussed here. *Sun* and *Sinha* demonstrated that diene **41** undergoes macrocyclization at C9/C10 in the presence of *Grubbs* 2nd generation catalyst very efficiently, albeit in an unselective manner (Scheme 5).[44] Note here that prior attempts of the *Danishefsky* group to close the macrocycle at C9/C10 by ring-closing metathesis, for reasons that remain unexplained, had failed.[45] The preparation of diene **41** by *Sun* and *Sinha* is conceptually similar to *Nicolaou*'s approach described above (Scheme 4).



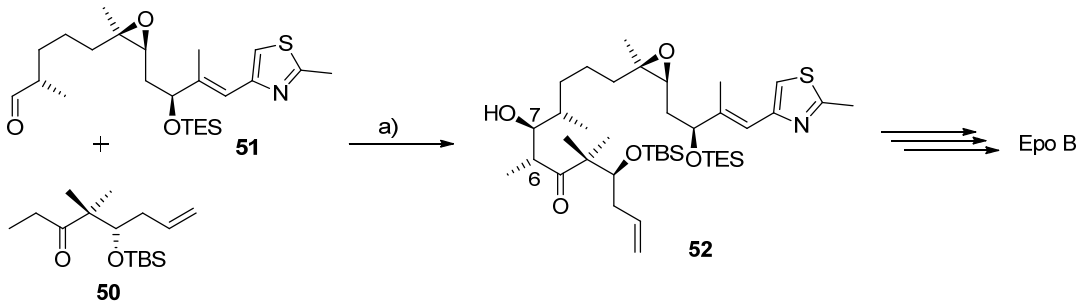
Scheme 5: a) LDA, THF, temperature, yield and selectivity not given; b) TBSOTf, 2,6-lutidine, CH_2Cl_2, 95%; c) H_2, Pd/C, EtOH; d) DMP, CH_2Cl_2; e) $MePPh_3I$, *n*-BuLi, THF, 66% over three steps; f) *p*-TsOH, MeOH; g) DMP, CH_2Cl_2; h) $NaClO_2$, 73% over three steps; i) EDCl, DMAP, CH_2Cl_2, 0 °C, 82%; j) *Grubbs* II (20 mol%), CH_2Cl_2, reflux, 89%, *E*/*Z* 1:1.

The early syntheses of epothilone that involved RCM-based ring-closure at C12/C13 by *Danishefsky*,[46] *Nicolaou*[47] (Scheme 4) and *Schinzer*[41] (Scheme 3) were characterized by a lack of *E*/*Z* selectivity. (This is also true for *Sinha*'s approach, although in this case the configuration of the double bond formed in the RCM step was inconsequential). *Fürstner et al*. provided a solution to overcome this limitation by applying a ring-closing alkyne metathesis (RCAM) approach to afford macrolide **49**, which upon *Lindlar* hydrogenation was converted exclusively into the *Z*-isomer (Scheme 6). The synthesis of the dialkyne **48** precursor made use of an aldol addition and esterification as the key steps[48],[49]



Scheme 6: a) LDA, THF, -78 °C, then **44**, 70%, dr 7:1; b) PPTS, MeOH, 85%, c) TBSOTf, 2,6-lutidine, 92%; d) CSA, CH_2Cl_2/MeOH, 78%; e) PDC, DMF, 83%; f) DCC, DMAP, CH_2Cl_2, 81%; g) Mo(*t*-BuN-3,5-sylyl)$_3$ (10 mol%), toluene/CH_2Cl_2, 80 °C, 80%; h) *Lindlar*, quinoline, H_2 (1 atm), CH_2Cl_2, quant; i) aq. HF, Et_2O/MeCN, 79%; j) DMDO, CH_2Cl_2, -78 °C, 70%.

Another epothilone synthesis that deserves special mention was reported by *Mulzer* and *co*-workers.[50],[51] In all syntheses before *Mulzer*'s work, the C12/C13 *Z*-olefin was epoxidized at the final stage of the synthesis. Obviously, this strategy was chosen in order to avoid for the presumably labile epoxide to be carried through a multistep synthesis. Contrary to this concern, *Mulzer* and *co*-workers could demonstrate that the epoxide in **51** could be installed prior to the formation of the C6/C7 bond *via* aldol addition and then carried through the synthesis without problems that would be related to the presence of the epoxide moiety (Scheme 7).[50],[51]



Scheme 7: a) LDA, THF, -78 °, then **51**, 92%, dr 20:1.

1.2.3 Analog Syntheses and SAR of Epothilones

The chemistry of epothilones has been extensively explored and several different total syntheses of natural epothilones have been established. At the same time this chemistry was employed for the construction of structurally modified analogs in order to investigate whether structural modifications could be identified that would result in improved activity against human cancer cells and/or would produce compounds with more favorable pharmacological properties *in vivo*. For structure-activity relationship (SAR) studies a large number of synthetic and semisynthetic analogs with modifications in essentially all sections of the epothilone skeleton have been investigated. These studies have indeed led to more potent derivatives, compounds with better pharmacological properties and analogs that are simpler to synthesize. The corresponding studies have been summarized in several reviews;[52],[53],[54],[55],[56] and in the following only those results and conclusions that are of relevance for this project will be discussed.

1.2.3.1 C12/C13 Modifications

Early SAR investigations were focused on the C12-C13 *cis*-epoxide moiety. The first structures to be investigated in this context were those that were intermediates in the total synthesis of Epo A or B. An important result that arose from these studies is the fact that deoxyepothilones (Epo C and Epo D, Figure 12: left) showed only slightly reduced biological activity compared to their epoxide containing parent compounds Epo A and B, respectively.[36],[37],[46],[57] In particular, the substrate for the final epoxidation step in the synthesis of Epo B, *i.e*. the natural product Epo D, emerged as an important analog from early SAR studies.

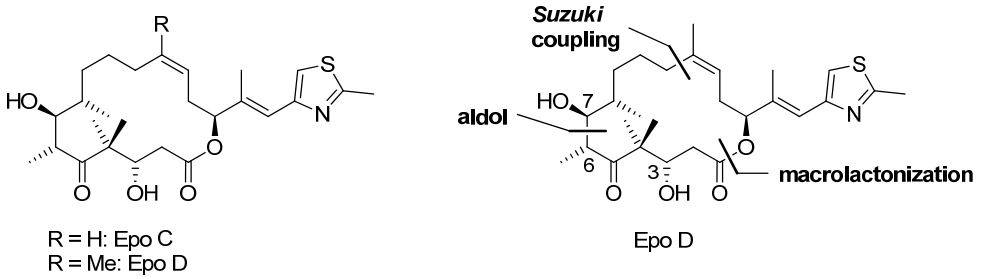
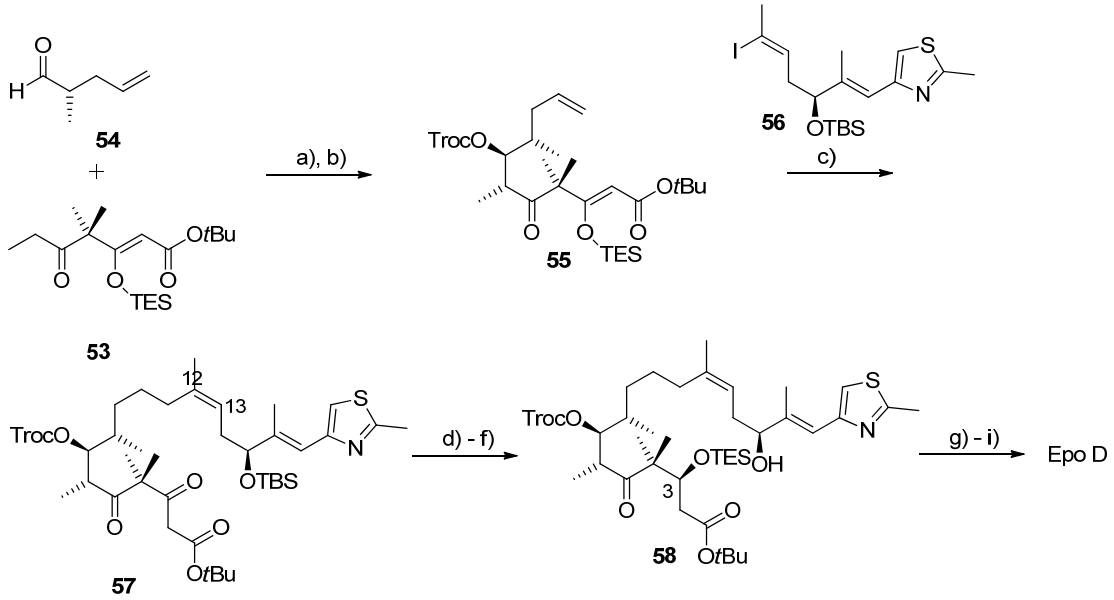


Figure 12: Structure of deoxyepothilones Epo C and Epo D and key retrosynthetic disconnections .

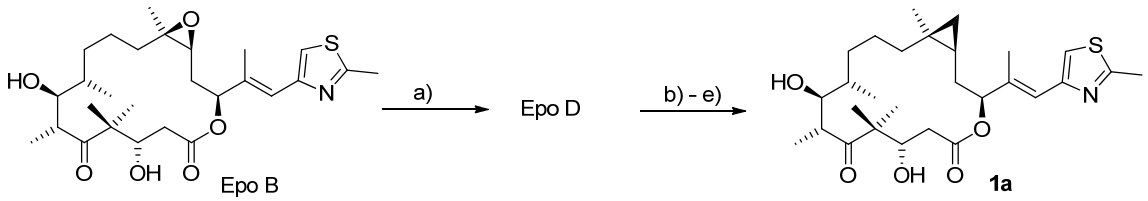
Scheme 8 summarizes *Danishefsky*'s second generation synthesis of Epo D that was developed in the context of extensive preclinical profiling of the compound.[58],[59] In this improved synthesis of Epo D the critical C6/C7 stereodiad was established by an aldol addition of ethyl ketone **53** to aldehyde **54**, which gave the desired aldol product with a selectivity of 5.5:1. *Suzuki* coupling of terminal olefin **55** with vinyl iodide **56** served to construct the C12-C13 *Z* double bond and a highly selective *Noyori* reduction[60] of the C3-keto group produced the stereocenter at C3. Obviously, this approach also provided improved access to Epo B.



Scheme 8: a) LDA, THF, -30 °C to -120 °C, then **54**, 60%, dr 5.5:1; b) TrocCl, pyridine, CH_2Cl_2, 0 °C to rt, then 0.5 N HCl in MeOH, 0 °C, 87 %; c) 9-BBN, THF, then **56**, [Pd(dppf)Cl_2], $AsPh_3$, Cs_2CO_3, H_2O, DMF; d) 0.4 N HCl in MeOH, 50% (over two steps); e) [(*R*)-(binap)$RuCl_2$], H_2 (83 bar), MeOH, HCl, 88%, dr 95:5; f) TESOTf, 2,6-lutidine, CH_2Cl_2, -78 °C to rt, then HCl/MeOH, 77%; g) 2,4,6-trichlorobenzoyl chloride, NEt_3, DMAP, toluene, 78%; h) SmI_2, cat. NiI_2, THF, -78 °C, 95%; i) HF·pyridine, THF, 98%.

The biological activity of Epo C and D showed that the potent inhibition of the growth of human cancer cells by epothilones did not critically depend on the presence of an epoxide moiety. This conclusion was then confirmed by the investigation of the cyclopropane analogs of Epo A and B, which demonstrated that the substitution of the epoxide ring by a cyclopropane moiety is well tolerated, with the corresponding cyclopropyl analogs of Epo A and B being essentially equipotent with the natural products.[61],[62] Cyclopropyl-epothilones were first prepared by the BMS group *via* semisynthesis from fermentatively produced Epo A

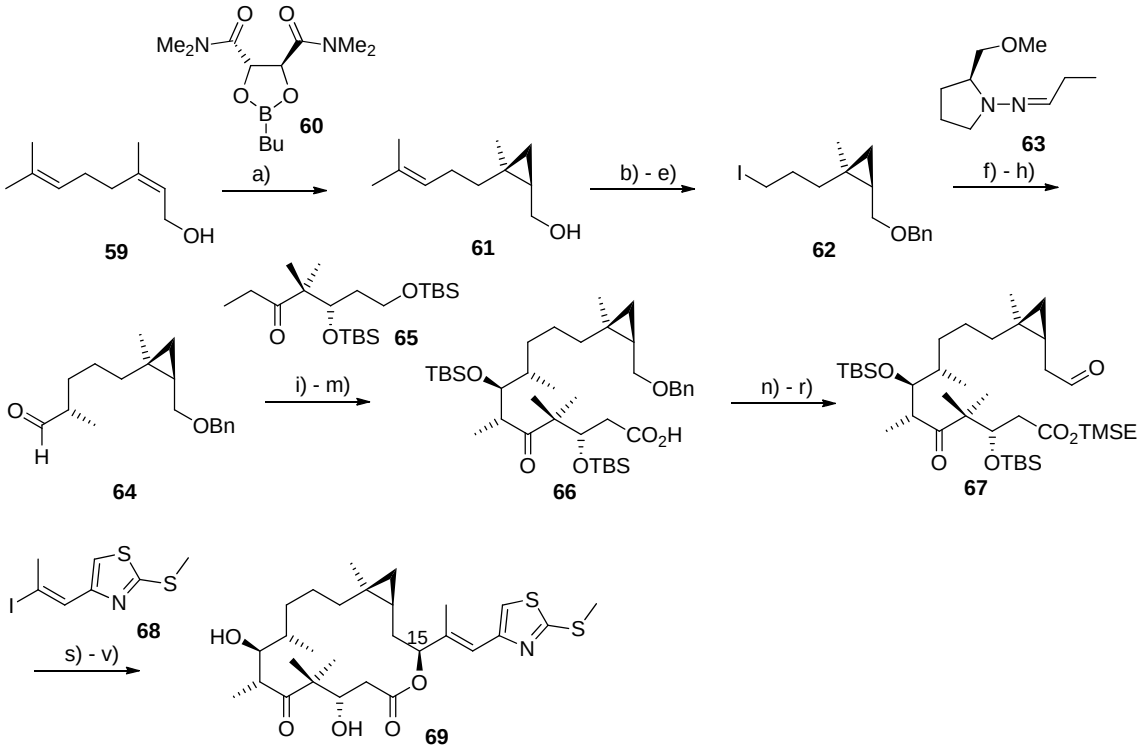
or B. As shown in Scheme 9 for cyclopropyl-Epo B (**1a**), the natural products Epo A or B are first deoxygenated to Epo C or D and subsequently the cyclopropane ring is installed.[61]



Scheme 9: a) WCl_6, *n*-BuLi, 78%; b) TBSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C; c) benzyltriethylammonium chloride, 50% aq. NaOH, $CHBr_3$, 45 °C, 30%; d) Bu_3SnH, AIBN, 70 °C; e) 20% TFA/CH_2Cl_2, -15 °C.

Subsequent to the work at BMS, *Nicolaou* and *co*-workers also developed stereoselective synthetic routes to cyclopropyl-epothilones.[62],[63],[64],[65] In addition to the preparation of cyclopropyl-Epo A and B, the approach developed by *Nicolaou* allowed the late stage attachment of the heterocycle-bearing side chain at C15. As an example, *Nicolaou*'s synthesis of *cis*-cyclopropyl Epo B analog **69** is shown in Scheme 10.[1] The cyclopropane moiety was introduced *via Charette* cyclopropanation[66] of *cis*-geraniol **59**. Cyclopropane **61** was then transformed into iodide **62**, which underwent *Enders*[67] alkylation with (-)-SAMP hydrazone **63**. The resulting aldehyde **64** was then connected to ethyl ketone **65** *via* aldol addition. The aldol product was elaborated into aldehyde **67** through an oxidation and homologation sequence. Aldehyde **67** was subsequently subjected to (non-selective) *Nozaki-Hiyama-Kishi*[68],[69] coupling with vinyl iodide **68**. The resulting mixture of isomers was cyclized under *Yamaguchi* conditions. At this stage the isomers could be separated by silica gel chromatography and final deprotection gave targeted cyclopropyl Epo B analog **69**.[63] This analog with the methylsulfanyl thiazole ring was chosen as it demonstrated to be a very potent inhibitor of the growth of human cancer cells. This compound is more active than its parent natural product Epo B exhibiting IC_{50} values in the low subnanomolar range as against the human ovarian cancer cell line 1A9 (IC_{50}: 0.1 nM for **69**, 0.6 nM for Epo B).

[1] *Trans*-cyclopropane analogs have been synthesized the same way by starting from *trans*-geraniol.



Scheme 10: a) $ZnEt_2$, CH_2I_2, **59**, DME, 80%, 95% ee; b) NaH, BnBr, DMF, 0 °C to rt, 100%; c) O_3, CH_2Cl_2/MeOH, -78 °C, then $NaBH_4$, -78 °C to rt, 83%; d) MsCl, NEt_3, CH_2Cl_2; e) NaI, acetone, 91% over two steps; f) LDA, THF, 0 °C, then **62**, - 78 °C to -10 °C, 87%; g) MeI, reflux; h) 3 M HCl/pentane, 91% over two steps; i) LDA, THF/Et_2O, -78 °C to -40 °C, then **64**, -78 °C, 80%, dr 14:1; j) TBSOTf, 2,6-lutidine, CH_2Cl_2, -20 °C; k) HF·py, py/THF, 0 °C, 86% over two steps; l) $(COCl)_2$, DMSO, CH_2Cl_2, NEt_3, -78 °C to 0 °C; m) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH/THF/H_2O; n) TMSE-OH, EDCI, DMAP, DMF, 73% over three steps; o) $Pd(OH)_2$/C, H_2, EtOH/EtOAc, 89%; p) $(COCl)_2$, DMSO, CH_2Cl_2, NEt_3, -78 °C to 0 °C, 99%; q) $MeOCH_2PPh_3Cl$, *n*-BuLi, THF, 0 °C, -78 °C to 0 °C, 79%; r) PPTS, dioxane/H_2O, 70 °C, 81%; s) $CrCl_2$, $NiCl_2$, 4-*t*-Bu-py, then **67**, DMSO; t) TBAF, THF, 0 °C to rt, 45% over two steps; u) NEt_3, 2,4,6-trichlorobenzoyl chloride, THF, 0 °C, then DMAP, toluene, 75 °C, 32%; v) aq. TFA, CH_2Cl_2, 71%.

1.2.3.2 C9/C10 Modifications

As first demonstrated by the *Danishefsky* group, the incorporation of an *E*-configured double bond between C9 and C10 in Epo D can result in an increase in antiproliferative activity (KOS-1584 (**70**); Figure 13),[70],[71] KOS-1584 (**70**) exhibits an IC_{50} value of 0.9 nM against the T-cell acute lymphoblastic leukemia cell line CCRF-CEM while Epo D was about four times less active (IC_{50}: 3.6 nM). The corresponding *Z*-isomer was markedly less active.[72] This observation is in agreement with spectroscopic studies, which indicated that the bioactive conformation of epothilones is characterized by *anti*-periplanar conformations about

the C9/C10 and C10/C11 bonds, respectively.[73] Based on its overall profile KOS-1584 (**70**) emerged as a very promising candidate for drug development and entered clinical trials.[74]

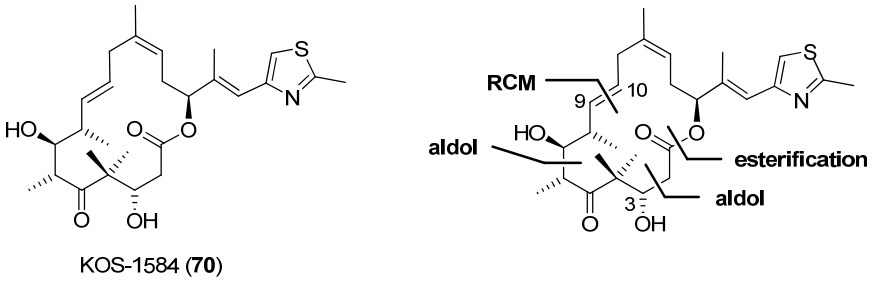
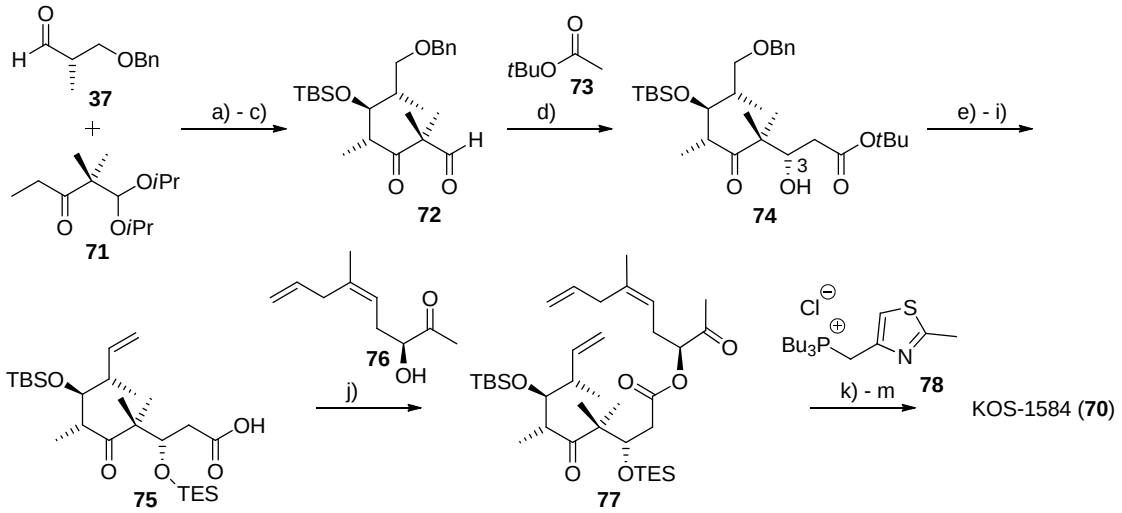


Figure 13: Structure of 9,10-dehydro Epo D (**70**) and key retrosynthetic disconnections.

The *Danishefsky* synthesis of KOS-1584 (**70**) made use of an aldol addition of ethyl ketone **71** to aldehyde **37**, giving the desired diastereomer in a 5.6:1 ratio, a second aldol addition to establish the chiral center at C3, an esterification to couple acid **75** and alcohol **76** and a RCM to form the macrocycle; the latter yielding the *E*-isomer exclusively (Scheme 11). Finally the heterocycle **78** was introduced *via HWE* olefination to afford KOS-1584 (**70**).[70],[71] It should be noted that this approach allows the facile attachment also of non-natural heterocycles at the final stage of the synthesis, a feature that was exploited by the *Danishefsky* group later in the synthesis of iso-fludelone (Section 1.2.3.4, Figure 15).



Scheme 11: a) LDA, THF, -90 °C, then **37**, 78%, dr 85:15; b) TBSOTf, 2,6-lutidine, CH_2Cl_2, -40 °C to -20 °C, 97%; c) *p*-TsOH·H_2O (cat.), THF/H_2O, 64 °C, 98%; d) LDA, cpTiCl(OR)$_2$, Et_2O, -78 °C, then **72**, 86%, dr 20:1; e) TESCl, imidazole, DMF, 0 °C to rt, 98%; f) H_2, Pd/C, EtOH, 83%; g) TPAP, NMO, CH_2Cl_2, 95%; h) $MePPh_3I$, *n*-BuLi, THF, -78 °C to -5 °C, 78%; i) TESOTf, 2,6-lutidine, CH_2Cl_2, 0 °C to rt; j) EDCI, DMAP, CH_2Cl_2, **76**, 0 °C to rt, 81% (over two steps); k) *Grubbs* II, toluene, 110 °C, 78%; l) KHMDS, **78**, THF, -78 °C to -20 °C, 76%; m) HF·pyridine, THF, 97%.

The combination of potent cytotoxicity and improved plasma stability of KOS-1584 (**70**) over Epo D provided the impetus for *Danishefky* and *co*-workers to synthesize substantial amounts of KOS-1584 (**70**) *via* the approach outlined in Scheme 11, in order to evaluate its *in vivo* efficacy.[75] The compound was advanced into clinical development and its safety, tolerability, and activity were evaluated in a Phase I dose escalation study.[74]

1.2.3.3 C3 Modifications

Modifications of epothilones at C3 were first investigated jointly by the BMS group and the *Höfle* research group at the GBF. This included the semisynthesis of 3-deoxy-2,3-didehydro derivates **79** and **80** (Figure 14), which were readily obtained from natural epothilones *via* bis-formylation at O3 and O7 and subsequent treatment of the bis-formyl ester with ammonia.[76] Remarkably, analogs **79** and **80** retain almost full activity of the parent products. Before this background *Altmann* and *co*-workers synthesized 3-deoxy Epo B **82** to address the question whether the saturated 3-deoxy derivatives (Figure 14) would be significantly less active.

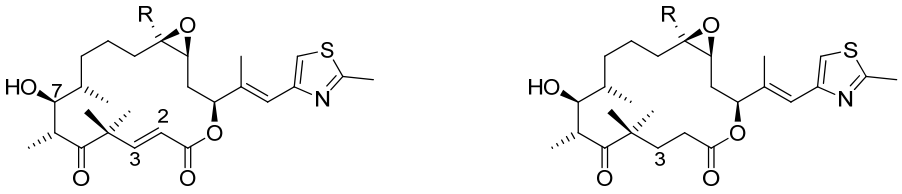
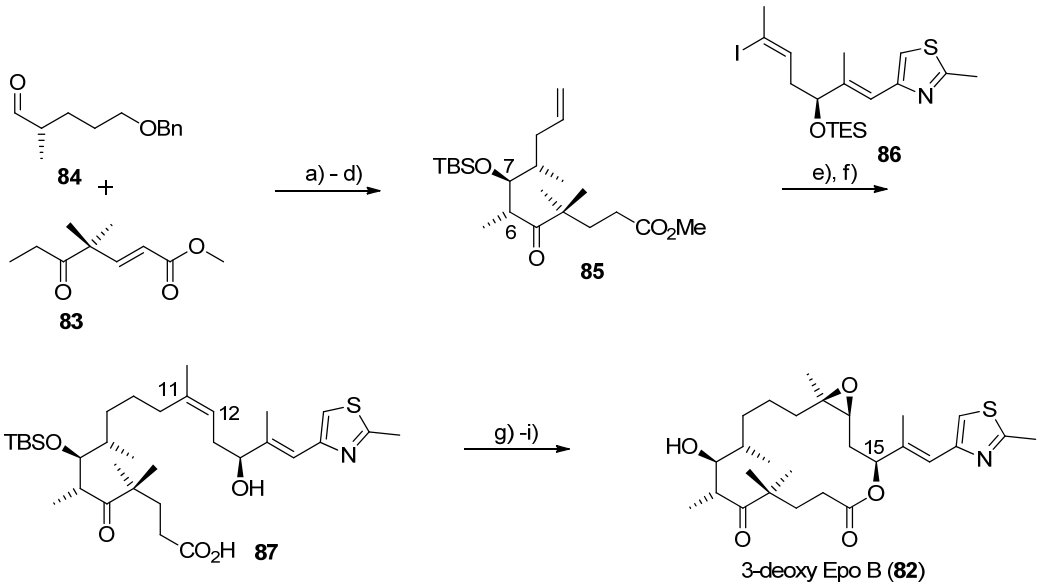


Figure 14: Structures of 3-deoxy-2,3-didehydro (left) and 3-deoxy derivatives (right).

Aldol addition between ethyl ketone **83** and α-chiral aldehyde **84** served to install the stereocenters at C6 and C7 (Scheme 12). The formation of the C11/C12 bond was achieved via *Suzuki-Miyaura* coupling[77] of olefin **85** and vinyl iodide **86**. Saponification and subsequent *Yamaguchi* macrolactonization,[78] followed by TBS-deprotection gave 3-deoxy Epo D. Final epoxidation was carried out using $MeReO_3/H_2O_2$ in a pyridine/water solvent mixture to give 3-deoxy Epo B (**82**) with remarkable selectivity (9:1).[79]



Scheme 12: a) LDA, THF, -78 °C, 58 %; b) TBSOTf, 2,6-lutidine, -10 °C, 82 %; c) H_2, Pd/C, MeOH, 97%; d) *o*-NO_2-(C_6H_4)SeCN, PBu_3, $NaHCO_3$, H_2O_2, 69 %; e) 9-BBN, THF, then **86**, Cs_2CO_3, [$PdCl_2(dppf)_2$], $AsPh_3$, DMF, -10 °C to rt, 55 %; f) LiOH, *i*-PrOH/water, 60 °C, 98 %; g) 2,4,6-trichlorobenzoyl chloride, NEt_3, THF, 0 °C; h) HF·py, THF, 90 % (2:1 mixture of isomers at C15); i) $MeReO_3$, H_2O_2/py/water, 72 % (9:1 mixture of epoxide isomers).

Intriguingly, analog **82** exhibits potent tubulin-polymerizing activity; it demonstrated to be almost as potent as Epo B and superior to Epo A or taxol (**17**). In addition, the compound retained high antiproliferative activity with IC_{50} values in the low nanomolar range as against the human epidermoid cancer cell lines KB-31 and KB-8511 (IC_{50} 7.4 nM and 4.0 nM, respectively, versus 0.29 nM and 0.22 nM for Epo B). Thus, its cellular potency is only 25 times lower than that of Epo B but comparable with that of Epo A or taxol (**17**). This result shows that the presence of the 3-hydroxy group is not crucial for potent biological activity of epothilone-type structures.[79]

1.2.3.4 Side Chain Modifications

Modifications of the heterocyclic side chain of epothilones have been extensively investigated. Based on the results of these studies the natural thiazole ring can be substituted by a variety of other heterocycles without loss or even with an improvement in activity.[57],[80] In addition, it was demonstrated that the removal of the allylic methyl group at C16 results only in a minor loss of activity.[31] Different strategies for the introduction of the side chain to the epothilone framework have been reported. Two of them, which rely on a late stage attachment of the heterocycle, have already been discussed here (Scheme 10 and 11). Among

side chain-modified analogs, sagopilone (ZK-Epo (**88**),[81] C21-amino-Epo B (BMS-310705 (**89**)),[53],[82] 20-desmethyl-20-methylsulfanyl-Epo B (ABJ879 (**90**))[65],[83] and iso-fludelone (**91**)[84] have entered clinical trials (Figure 15).

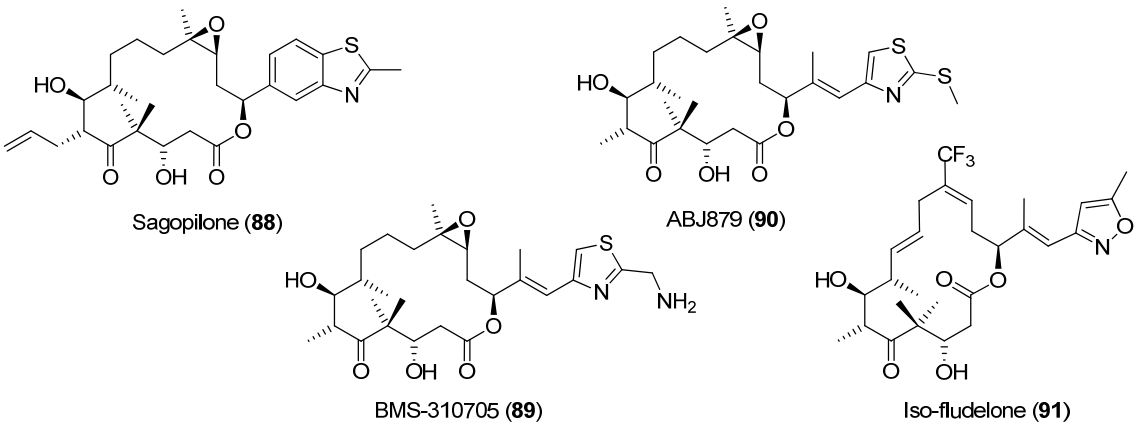


Figure 15: Structures of sagopilone (**88**), BMS-310705 (**89**), ABJ879 (**90**), and iso-fludelone (**91**).

The development of the isoxazole based analog iso-fludelone (**91**) nicely demonstrates how the cellular activity can be improved by the substitution of the natural thiazole ring with an appropriate heterocycle.[84] In order to improve the therapeutic index of KOS-1584 (**70**) (Figure 13), *Danishefsky* and *co*-workers explored replacing the three hydrogen atoms of the 26-methyl group with fluorine atoms. The synthesis discussed above for KOS-1584 (**70**) (Scheme 11) could be directly applied to the desired fluorinated analog, which has been termed fludelone (**92**) (Figure 16). This compound indeed proved to be less toxic than KOS-1584 (**70**) but also lost some of the antiproliferative activity in comparison to KOS-1584 (**70**). The IC_{50} values against the human T-cell lymphoblastic leukemia cancer cell line CCRF-CEM are 0.27 nM for **70** and 0.71 nM for fludelone (**92**); against the human lung carcinoma cell line A549 0.09 nM for **70** and 0.31 nM for fludelone (**92**). In an attempt to restore some of the potency that had been lost in the transition from KOS-1584 (**70**) to fludelone (**92**), the *Danishefsky* group designed an isoxazole-based analog of fludelone, which they named iso-fludelone (**91**) (Figure 16).[84] In fact, iso-fludelone (**91**) was found to be more potent in *vitro* than fludelone (**92**) and it is also metabolically more stable. The compound exhibits an IC_{50} value of 0.27 nM against CCRF-CEM and 0.05 nM against A549, respectively.[84]

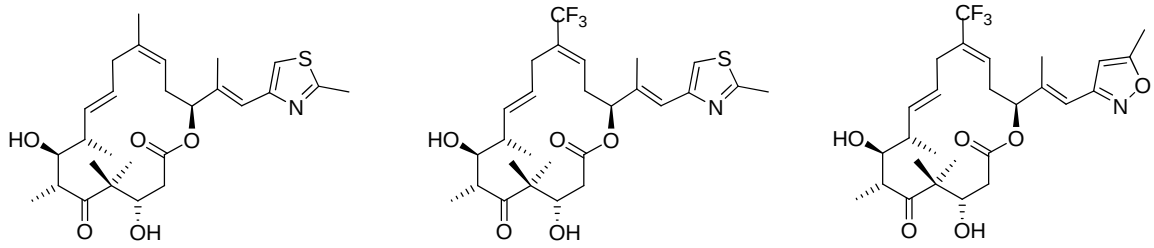


Figure 16: Structure of KOS-1584 (**70**), Fludelone (**92**) and Iso-fludelone (**91**).

As previously discussed in section 1.2.3.1, *Nicolaou* and *co*-workers developed a synthesis of epothilone analogs that relied on the late stage attachment of the heterocycle side chain (Scheme 10). In the course of an intensive evaluation of the antiproliferative activity of side chain modified epothilone analogs pyrazole-based compounds **93** and **94** exhibited cellular activity superior to Epo B. Against the human ovarian cancer cell line 1A9 **93** exhibited an IC_{50} of 0.50 nM and **94** of 0.06 nM versus 0.99 nM for Epo B. The growth of cancer cell line KB-8511 was inhibited with an IC_{50} of 0.19 nM for **93** and with 0.09 nM for **94** while Epo B exhibited an IC_{50} of 0.42 nM.

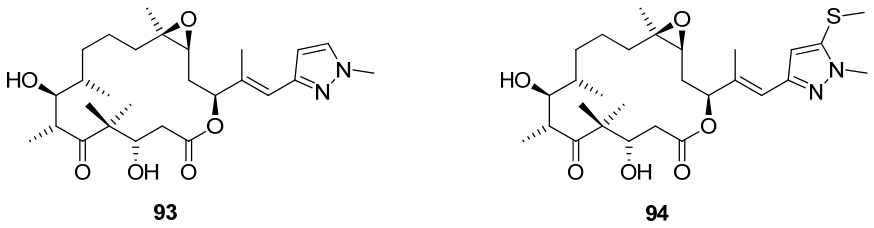


Figure 17: Structure of pyrazole-based Epo B analogs **93** and **94**.

These results indicate that the incorporation of pyrazole heterocycles might result in an increased potency.

1.2.4 Epothilones in Clinical Trials

Nine epothilone-type compounds (Figure 18) have entered clinical trials in humans, including the natural product Epo B (developed by Novartis as EPO906 or patupilone). Patupilone was developed up to Phase III, where it did not show a significant overall survival advantage of patients with advanced ovarian cancer, refractory or resistant to platinum-based therapy, compared to standard therapy. Based on these data Novartis refrained from filing the compound for registration.[34] Clinical trials with KOS-862 (Epo D), KOS-1584 (**70**), sagopilone (**88**) (ZK-EPO), BMS-310705 (**89**) and ABJ879 (**90**) appear to have been terminated or at least put on hold. Iso-fludelone (**91**) entered Phase I clinical trials in 2011, but no data on this trial have been reported. Detailed up-to-date information on epothilones in clinical trials can be obtained from the Prous Integrity database.[85]

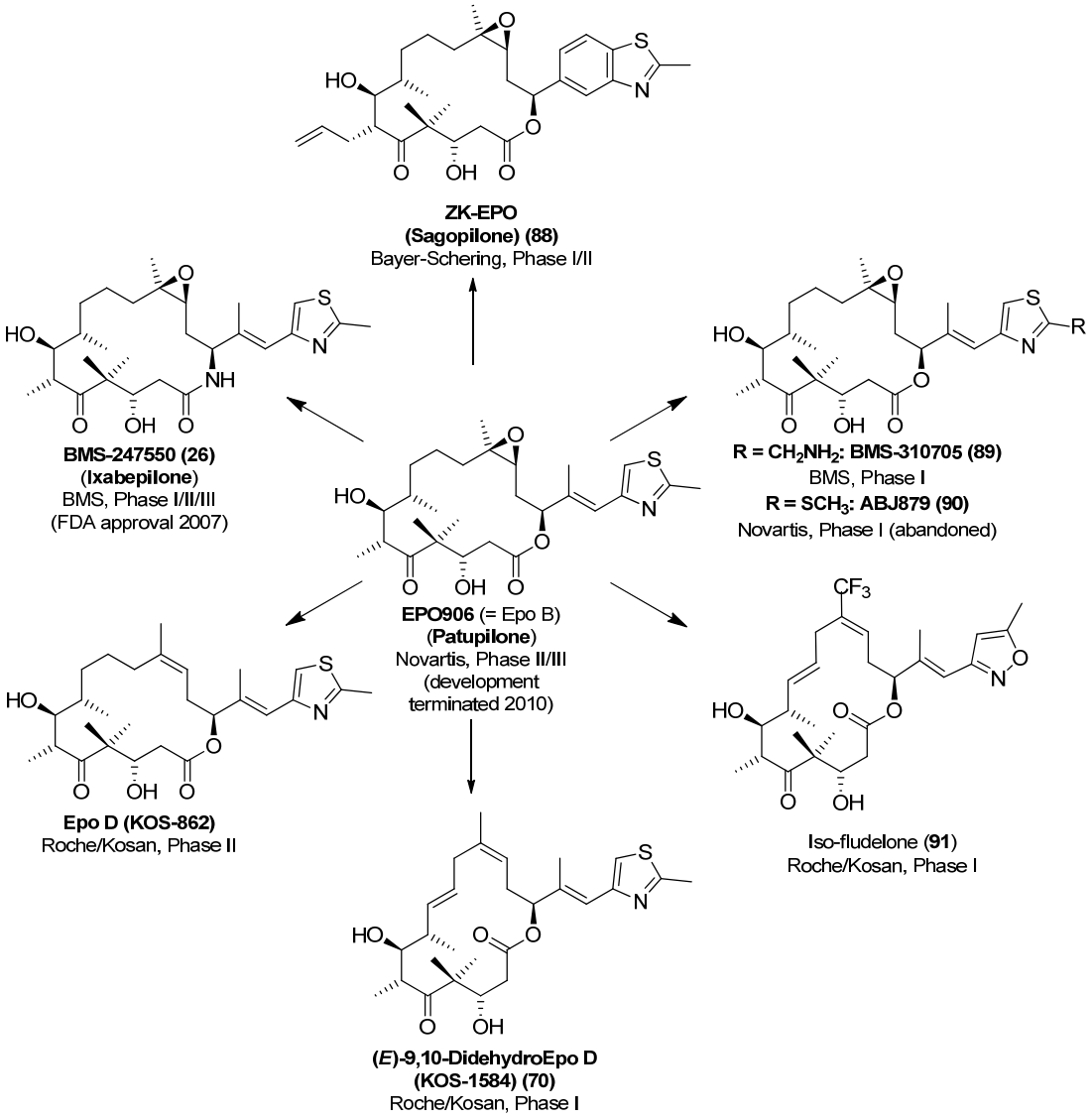
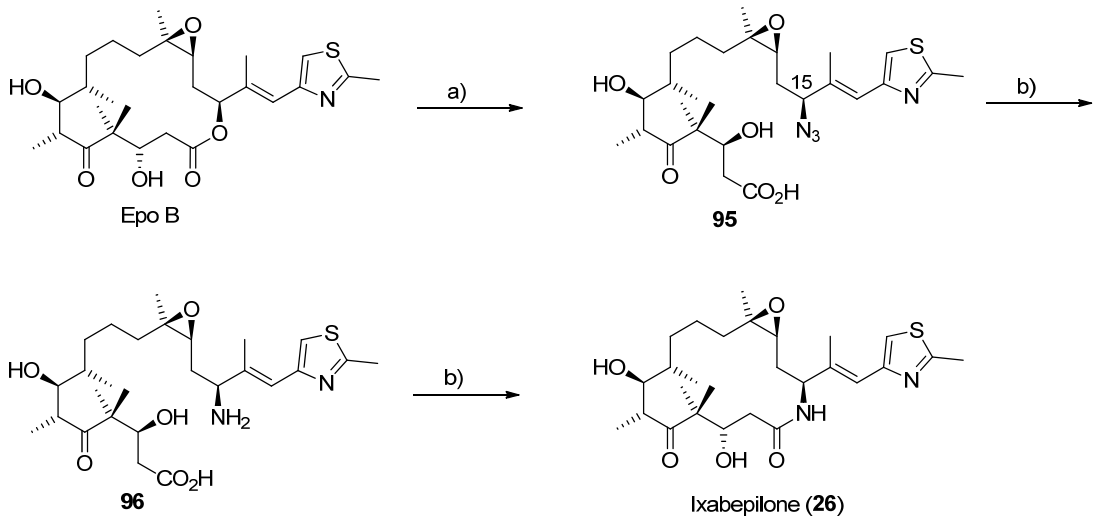


Figure 18: Epothilones that have been advanced to clinical trials.

Early pharmacokinetic studies with epothilones in rodents pointed to a distinct vulnerability of the ester bond in the macrocycle to hydrolysis by (rodent) plasma esterases. Although esterase activity in rodent plasma is known to be substantially higher than in humans, these early findings raised concerns about the metabolic stability of natural, macrolactone-based epothilones also in humans. In response to these concerns the group at BMS conceived the metabolically more stable lactam analog of Epo B, i. e. **26**, as an alternative for therapeutic applications in humans.[86] This work has resulted in an efficient process for the preparation of the Epo B lactam **26**, a compound which was developed into a clinical anticancer drug and is marketed under the trade name Ixempra® (generic name ixabepilone (**26**), Scheme 13).[35, 87] Ixabepilone (**26**) is produced by semisynthesis from Epo B by exploiting the allylic nature of the epothilone lactone moiety. The latter can opened by treatment with $Pd(PPh_3)_4$ in the presence of sodium azide, which leads to the formation of azido acid **95** with complete retention of configuration at C15 (Scheme 13).[86] Subsequent reduction of the azide moiety under *Staudinger* conditions[88] followed by macrolactamization gives ixabepilone **26**. This semisynthetic route was optimized into a one pot procedure, which yields **26** in a single day in 23% yield.[86]



Scheme 13: a) $Pd(PPh_3)_4$ (10 mol%), NaN_3, THF/H_2O, 45 °C, 70%; b) PMe_3, THF/H_2O, 71%; c) DPPA, $NaHCO_3$, DMF, 4 °C, 43% or EDCI, HOBt, MeCN/DMF, rt, 65%.

Danishefsky and *co*-workers have also reported a fully synthetic route to ixabepilone (**26**).[89]

In addition to the analogs alluded to above, a tumor-targeting Epo A analog-folic acid conjugate **97** (BMS-753493) has entered clinical trials.[90] Receptor-specific targeting is an

approach that enables selective delivery of cytotoxic drugs to cancer cells, thereby avoiding the collateral damage that accompanies their uptake by normal cells. The folate receptor is a cell surface glycoprotein that is expressed in relatively high levels in human epithelial cancers, but has limited expression in normal tissue.[91] It binds folic acid and conjugates tightly (dissociation constant for folic acid K_d 10^{-9} M).[92] Upon binding the folic acid is internalized by endocytosis: therefore conjugation of an antiproliferative agent to folic acid is a promising strategy to target drugs to tumors.[92],[93] BMS-753493 (**97**) was advanced to Phase I clinical trials, but seems no longer to be under active development.

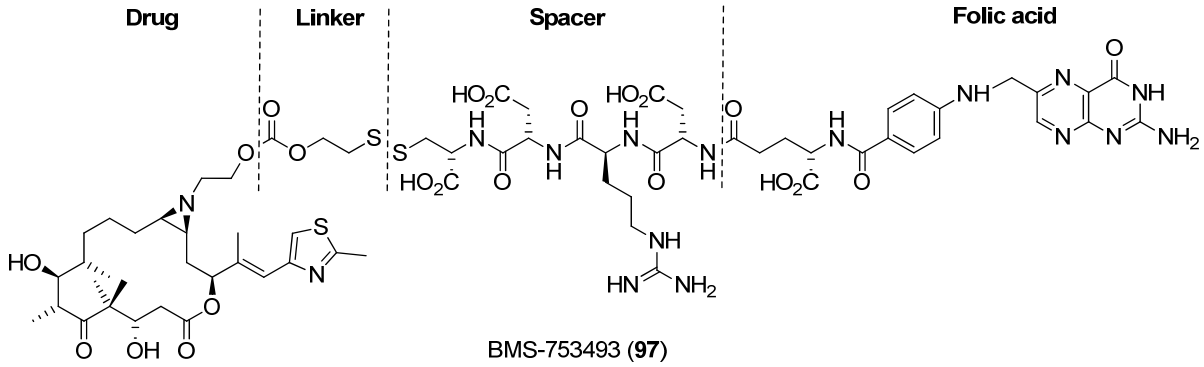


Figure 19: Structure of the epothilone-folate conjugate BMS-753493 (**97**).

1.3 Immunoconjugates

Due to the lack of real selectivity of tubulin modulators against tumor cells and the resulting side effects, a sophisticated antibody-drug conjugate (ADC) that releases the active agent near the tumor cells could be used for directed tumor targeting (for recent reviews see ref [94],[95],[96]). As a consequence, the numerous side effects of drugs could be diminished. These immunoconjugates generally consist of three elements: a monoclonal antibody, the active agent and a chemical linker between the two (Figure 20).[96],[97]

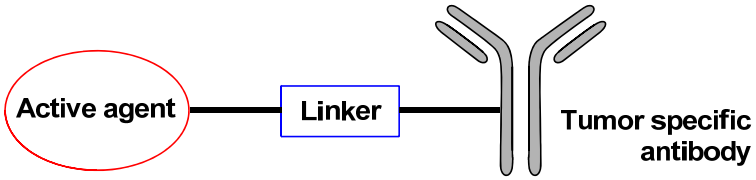


Figure 20: Schematic representation of an antibody-drug conjugate (ADC).

1.3.1 Mechanism of Action

The antibody part of an ADC either binds to tumor specific antigens that are exclusively expressed on tumor cells or to overexpressed tumor-associated antigens.[98] Conceptually, the accumulation of immunoconjugates at the surface of tumor cells leads to internalization and the intracellular release of the active agent by cleavage of the chemical linker between the drug and the antibody. Immunoconjugates that are not internalized can also release their drug cargo outside of the cell followed by passive diffusion of the small molecule across the tumor cell membrane. For effective tumor targeting of ADC's, certain requirements need to be met,[99] including (1) the availability of a highly specific antibody; (2) IC_{50}-values of the active agent in the subnanomolar range; (3) the chemical linker should be stable in the systemic circulation and allow release of the active agent inside the tumor cell or at the cell surface; and (4) the immunoconjugate itself should not cause an immune response.

1.3.2 Clinically Approved Immunoconjugates

Three examples of clinically tested and approved antibody-drug conjugates are Mylotarg® (Manufacturer: Pfizer), Adcetris® (Manufacturer: Seattle Genetics) and Kadcycla® (Manufacturer: Roche). Mylotarg® (Gemtuzumab ozogamicin) is an antibody-drug conjugate

consisting of the cytotoxic drug calicheamicin and a monoclonal antibody against the transmembrane receptor CD33.[100] It was approved in 2001 and used to treat acute myelogenous leukemia (AML) in elderly patients until it was withdrawn from the market in 2010 due to safety concerns and a lack of increased benefit over conventional therapies,[100] but the drug is still available to patients through access programs. Adcetris® (Brentuximab vedotin) is composed of the tubulin polymerization inhibitor monomethyl auristatin E (MMAE, Vedotin) as the active drug moiety and the chimeric monoclonal antibody brentuximab which is directed against the membrane protein CD30. Brentuximab vedotin was approved by the FDA in 2011 for the treatment of anaplastic large cell lymphoma (ALCL) and Hodgkin lymphoma.[101],[102] Kadcycla® (Trastuzumab emtansine) consists of the monoclonal antibody trastuzumab recognizing the HER2 receptor linked by a thioether to the cytotoxic agent maytansine, which binds to tubulin dimers and thereby inhibits tubulin polymerization.[103] Trastuzumab emtansine was approved by the FDA in 2013 for breast cancer treatment.[104]

1.4 Aims and Scope

As discussed above, a large number of epothilone analogs have been investigated in SAR studies since the original discovery of Epo A and B. However, one of the fundamental problems associated with cytotoxic anticancer agents, namely the narrow selectivity window between cancer and normal cells, is not fully addressed by any of these analogs, including those that have been advanced to clinical trials. Before this background the construction of epothilone-based ADC's represents one of the long term goals of the work initiated in this PhD thesis. In an initial phase this required the identification of highly potent functionalized analogs that would be suitable for conjugation to tumor-specific antibodies. Thereby, the lack of selectivity of epothilones for cancer cells over normal cells should be addressed. Specifically, this work has focused on functionalized cyclopropyl analogs of Epo B, the most potent natural epothilone, as the active drug cargo of ADC's. As discussed in section 1.2.3.1, 12,13-cyclopropyl (CP) analogs of Epo A and B have been found to be equally potent, or even somewhat more potent than the respective epoxide-based parent compounds.[61] At the same time the replacement of the natural 12,13-epoxide moiety by a cyclopropane ring should lead to enhanced chemical and, in particular, metabolic stability, given the susceptibility of the oxirane ring to undesired chemical transformations[105] and metabolic attack.[106] An *E*-configured double bond might be incorporated at C9/C10. Such an incorporation resulted in an increase antiproliferative activity as first shown for Epo D.[70],[71] As mentioned in section 1.2.3.4, heterocycles other than the natural thiazole moiety can result in increased activity.[57],[80] In particular, analogs containing isoxazole[84] or pyrazole[107] rings were found to be highly potent.

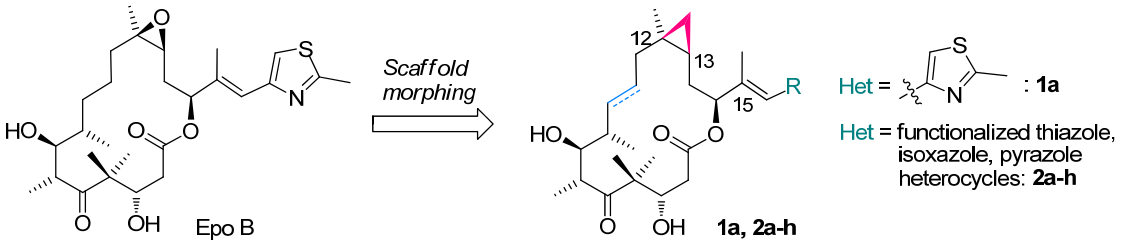
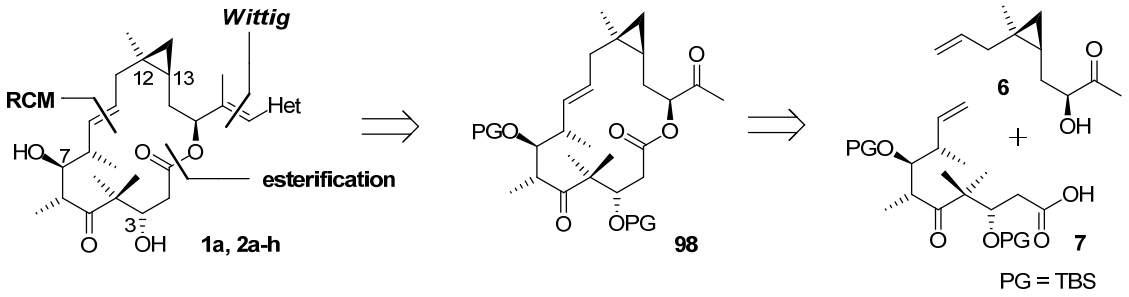


Figure 21: Target structures **1a, 2a-h** of novel epothilone derivatives.

As a first step in the development of cycloproyl-epothilone-based ADC's one of the objectives of this PhD thesis was to establish efficient synthetic access to side chain-functionalized cyclopropyl-based analogs of Epo B **2a-h** (Figure 21). As part of this objective

a new total synthesis of cyclopropyl-Epo B (**1a**) was to be developed that would form the basis for the synthesis of side chain modified analogs. Conceptually, target structures **1a** and **2a-h** were to be accessed through a convergent strategy, which comprises the coupling of the two building blocks **6** and **7** *via* an esterification/ring-closing metathesis (RCM) sequence to obtain the macrocyclic core (Scheme 14). The introduction of different heterocycles was to be performed at a late stage of the synthesis, in order to access a variety of side chain-modified analogs from a single advanced intermediate.



Scheme 14: Retrosynthetic strategy.

The questions to be answered in this PhD thesis was whether (1) the previous SAR data, which were all derived from 12,13-epoxide-based analogs, could be extrapolated to cyclopropyl-epothilones, and (2) whether functional groups could be attached to these heterocycles without loss in potency that would allow the selective attachment of tumor-targeted antibodies. Such a functional group was thought to be required as the natural hydroxy groups at C3 and C7 were considered not to be reactive enough.

In summary, epothilone analogs of the general structure **1a** and **2a-h** (Figure 21) were to be targeted for synthesis as potential drug cargo for epothilone-based ADCs. In order to ensure maximum potency these analogs were to be based on the Epo B scaffold, although this would lead to somewhat enhanced synthetic complexity. In order to ensure a high level of flexibility with regard to the specific structure of the heterocycle, the introduction of the side chain heterocycle was to be performed only during the final stages of the synthesis.

1.5 Results and Discussion

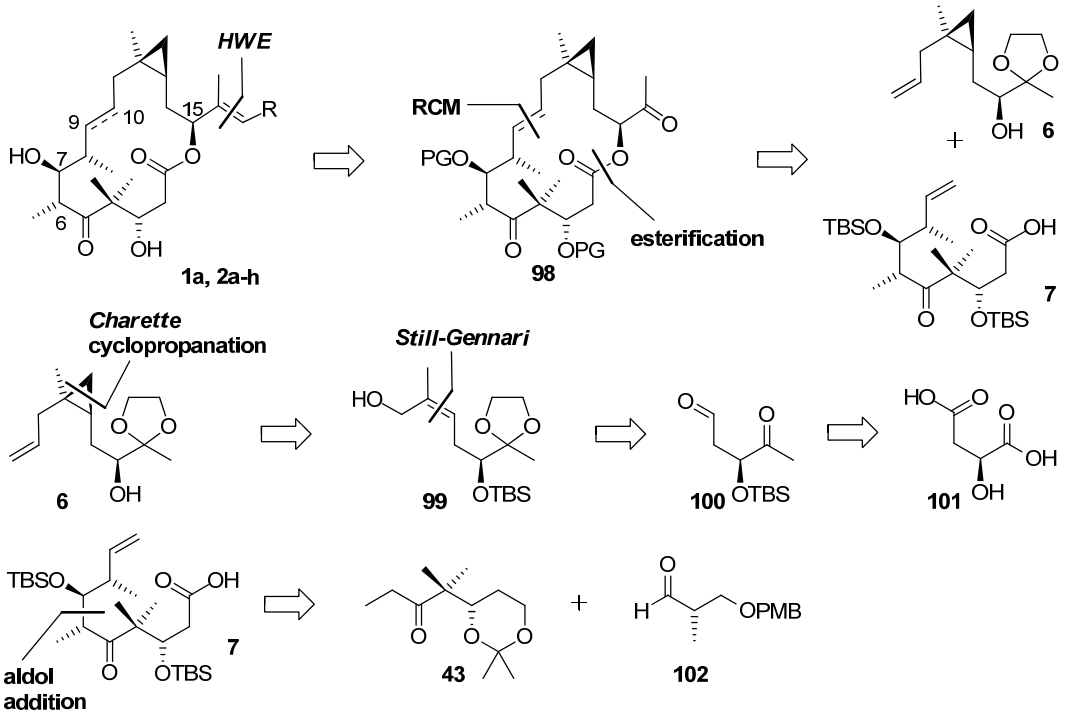
1.5.1 Cyclopropyl-Epo B and Side Chain-modified Analogs

1.5.1.1 Synthetic Planning

As discussed in section 1.2.3.1, *Nicolaou*'s synthesis of cyclopropyl-epothilones (CP-epothilones) was based on the late stage addition of a side chain vinyl iodide to a C15 aldehyde by means of a *Nozaki-Hiyama-Kishi* coupling[68] as a key step (Section 1.2.3.1, Scheme 10).[62],[63],[64],[65] Unfortunately, the coupling was essentially nonselective, thus leading to a substantial loss of material at a late stage of the synthesis. In an attempt to overcome this limitation, an alternative strategy to CP-Epo B (**1a**) and its analogs was planned, where the heteroaryl-vinyl side chain was to be established through *HWE* olefination chemistry with methyl ketone **98** (Scheme 15). An analogous approach had been employed by *Danishefsky* and *co*-workers in the synthesis of 9,10-dehydro Epo D (**70**) (section 1.2.3.2, Scheme 11)[70],[71] and its 26-trifluoromethyl variant (fludelone) (**92**)[75] as well as by *Avery* and *co*-workers in their synthesis of Epo A to generate the epoxidation precursor Epo C.[108] Based on these previous reports, the elaboration of ketone **98** into the CP-Epo B (analogs) appeared to be a feasible approach. It should also be noted, however, that *Höfle* and *co*-workers had been unable to re-establish Epo A from the corresponding (epoxide-containing) side chain ketone, in spite of significant optimization attempts,[109] thus indicating that the chemistry employed by *Avery* and *Danishefsky* for the synthesis of Epo C and Epo D (derivatives), respectively, might not necessarily be extendable to cyclopropane-containing ketone **98**.

As illustrated in Scheme 15, methyl ketone **98** was to be obtained through RCM between C9 and C10 of the desired macrocycle followed by the selective reduction of C9/C10 double bond. The requisite diene precursor for the macrocyclization reaction would be assembled from alcohol **6** and acid **7**; the bis-TBS protected version of the latter had been previously synthesized in the group.[110] Very similar to *Schinzer*'s and *Fürstner*'s synthesis of Epo A (section 1.2.2, Scheme 6), the synthesis makes use of an aldol addition of *Schinzer* ketone **43** and α-chiral aldehyde **102** to install the stereocenters at C6 and C7. The stereoselective establishment of the cyclopropane moiety in **6** was to be achieved through *Charette* cyclopropanation of allylic alcohol **99**, which would be derived from keto aldehyde **100** by means of *Still-Gennari* olefination[111]. Based on literature precedence it was felt that *Still-*

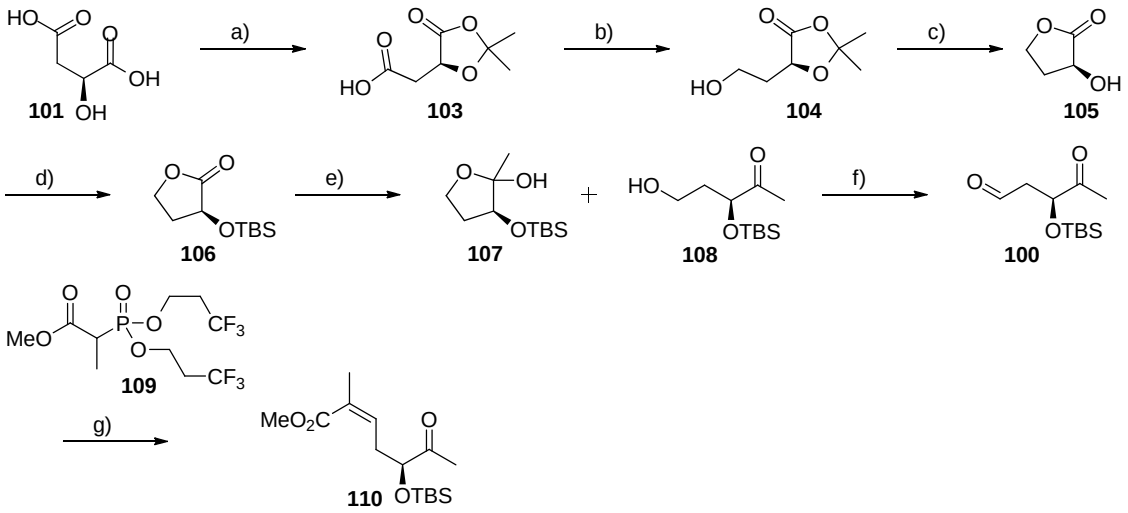
Gennari olefination of the aldehyde group would be feasible selectively in the presence of the keto group at C16 in **100**,[112] while the latter would have to be protected in subsequent steps. Lastly, aldehyde **100** was planned to be prepared from *S*-malic acid (**101**) as a defined source of chirality at C15.[113]



Scheme 15: Retrosynthesis of target structures **1a**, **2a-h**.

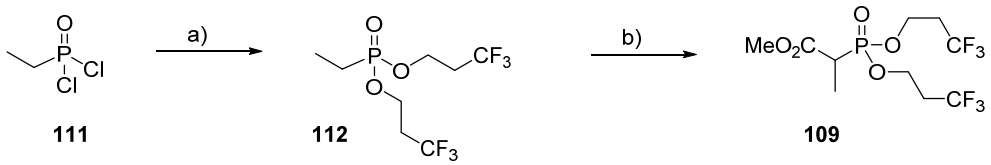
1.5.1.2 Total Synthesis of CP-Epothilone B (1a)

Starting from commercially available *S*-malic acid (**101**), α-hydroxy lactone **105** was prepared in a 3-step literature sequence (Scheme 16).[114],[115],[116] First, **101** was treated with 2,2-dimethoxypropane in the presence of *p*-TSA to yield acetal **103**. Subsequently, the acid in **103** was reduced to the alcohol **104** by the use of BH_3·THF complex. Upon addition of a catalytic amount of *p*-TSA the formation of α-hydroxy lactone **105** was triggered, which was TBS protected in the next step. Treatment of **106** with MeLi gave a mixture of cyclic hemiacetal **107** and the corresponding open chain hydroxy ketone **108**. Synthetically useful conversion of this mixture into aldehyde **100** could only be achieved by DMP[117] oxidation giving a yield of 67%, while all other oxidation methods investigated did not provide any of the aldehyde (PDC, *Swern*) or gave only low yields (py·SO_3). Subsequent *Still-Gennari* olefination[111] with phosphonate **109** furnished the desired *Z*-isomer **110** exclusively.



Scheme 16: a) 2,2-dimethoxypropane, *p*-TSA, 77%; b) BH_3·THF, THF; c) *p*-TSA, benzene, 56% over two steps; d) TBSCl, imidazole, DMF, 97%; e) MeLi, THF, -78 °C, 89%; f) DMP, CH_2Cl_2, 67%; g) KHMDS, 18-C-6, THF, -78 °C, then **100**, -78 °C, 77%.

The requisite the *Still-Gennari* reagent **109** was synthesized according to literature procedures from commercially available ethylphosphonic dichloride **111** which was first converted into phosphonate **112** (Scheme 17). Subsequent acylation gave the reagent **109**.[118]



Scheme 17: a) Trifluoroethanol, NEt_3, THF, 98%; b) LiHMDS, $ClCO_2Me$, THF, -78 °C, 73%.

The geometry of the double bond in **110** was firmly established by means of NOESY experiments. The presence of the strong NOE between the olefinic proton and the protons at the methyl group attached at the double bond confirmed the formation of the *Z*-isomer (Figure 22).

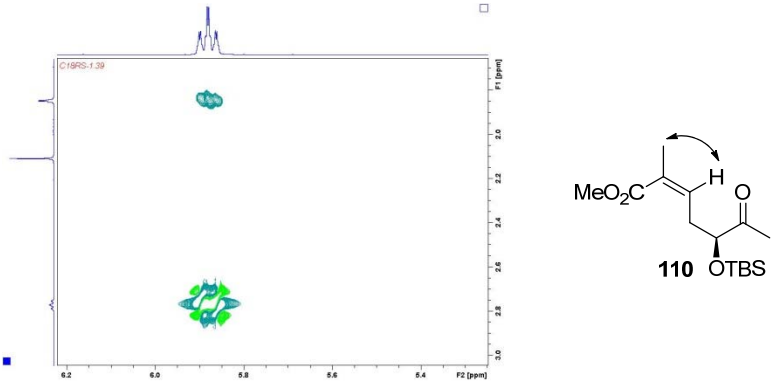
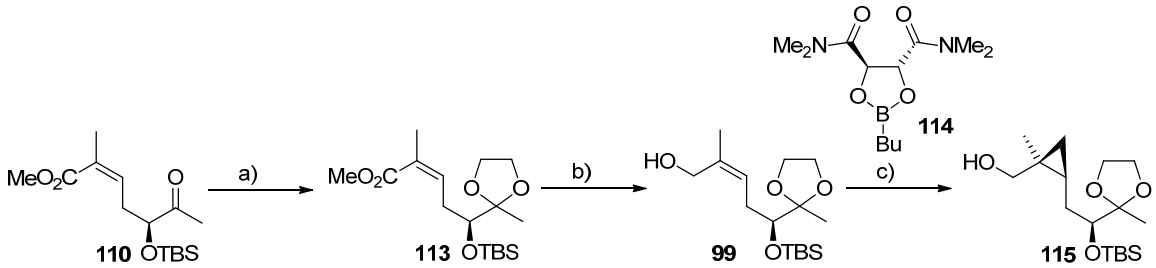


Figure 22: NOESY spectrum shows that the olefinic proton is in closed vicinity to the methyl group.

Acetal protection of the ketone functionality in **110** by treatment with ethylene glycol in the presence of triethyl orthoformate and *p*-TSA followed by reduction of the ester moiety with DIBAL-H led to allylic alcohol **99** in excellent overall yield (94% for two steps) (Scheme 18). The latter underwent highly stereoselective *Charette* cyclopropanation (dr 18:1) to afford alcohol **115** in high yield (88% for single isomer).[66] It should be noted here that violent explosions have been reported for *Charette* cyclopropanations carried out on scales of 8 mmol or higher, due to the exothermicity associated with the formation of $Zn(CH_2I)_2$. However, careful temperature control during the addition of CH_2I_2 to the $Zn(Et)_2$ solution allowed the cylopropanation of **99** to be carried out safely also on a larger scale.[113]



Scheme 18: a) Ethyleneglycol, $CH(OEt)_3$, *p*-TSA, 40 °C, 97%; b) DIBAL-H, CH_2Cl_2, -78 °C, 97%; c) $Zn(Et)_2$, CH_2I_2, **99**, CH_2Cl_2, 0 °C to rt, 98%, dr 18:1.

The product ratio for the cyclopropanation reaction could be readily determined by ^{1}H-NMR spectroscopy (Figure 23). Separation of the two diastereomers by silica gel chromatography provided pure **115**.

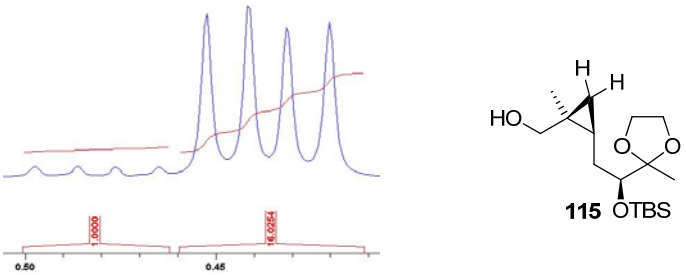
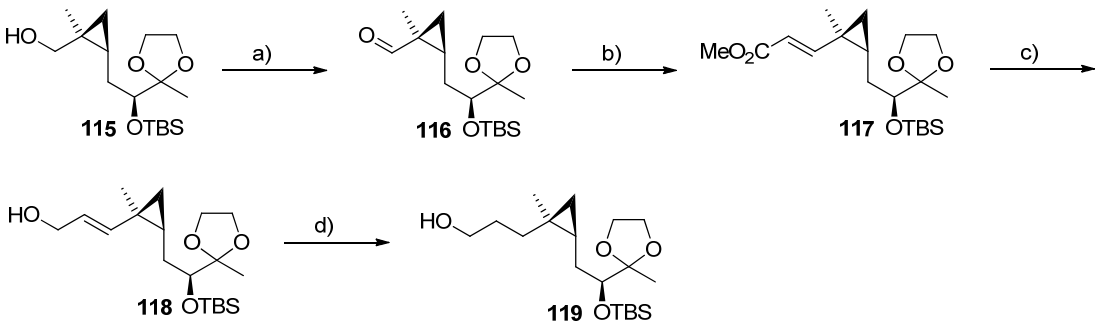


Figure 23: ^{1}H-NMR signal of one of the two CH_2 protons of the two cyclopropane diastereomers.

With this key reaction successfully implemented, our efforts were then directed toward installing the terminal double bond required for RCM-based macrocyclization. After *Swern* oxidation of **115**, the resulting aldehyde **116** was subjected to *Wittig*-olefination with Ph_3PCHCO_2Et to furnish α,β-unsaturated ester **117** as a single isomer (Scheme 19). Subsequent reduction of the ester moiety with DIBAL-H followed by reduction of the double bond with $NaBH_4/CoCl_2{\cdot}6H_2O$[119] provided saturated alcohol **119**.



Scheme 19: a) $(COCl)_2$, DMSO, NEt_3, -78 °C to rt, 98%; b) Ph_3PCHCO_2Et, CH_2Cl_2, reflux, 99%; c) DIBAL-H, CH_2Cl_2, -78 °C, 98%; d) $NaBH_4$, $CoCl_2 \cdot 6H_2O$, MeOH/DMF, 95%.

Dissolution of **118** in MeOH and slow evaporation of the solvent resulted in the formation of needles which were suitable for X-ray crystallographic analysis. The crystal structure confirmed the predicted stereochemical outcome of the *Charette* cyclopropanation (Figure 24).[2]

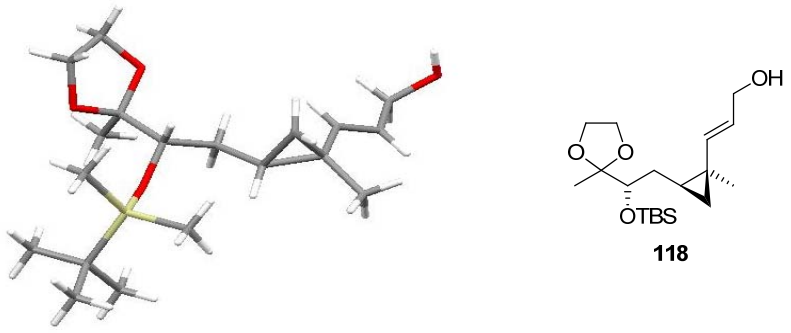
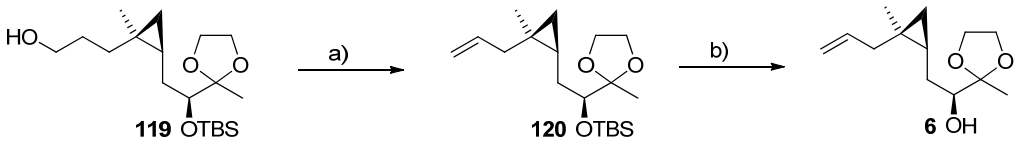


Figure 24: Crystal structure of allylic alcohol **118**.

Finally, the terminal double bond in **120** was installed by means of seleniumoxide eliminiation[120] and subsequent TBS deprotection provided alcohol **6** in a total of 12 steps and excellent overall yield (25%) from commercially available α-hydroxy lactone **105**.[113]

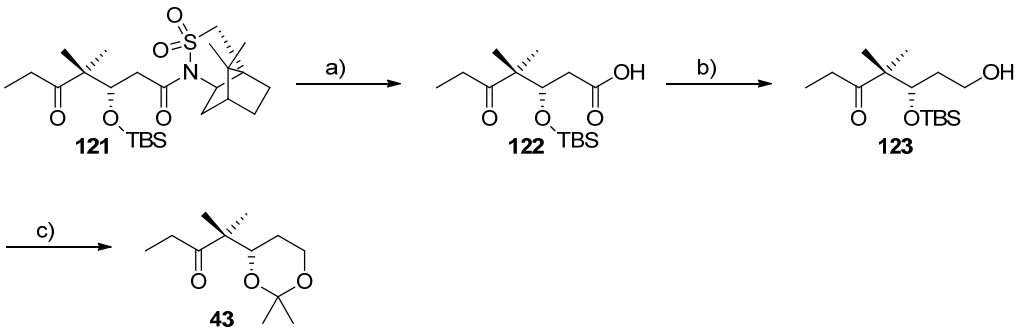


Scheme 20: a) i. *o*-$NO_2PhSeCN$, PBu_3, THF. ii. H_2O_2, $NaHCO_3$, 30 °C to 50 °C, 84%; b) $TBAF \cdot 3H_2O$, THF, 50 °C, 93%.

Building block acid **7** was synthesized from the protected β-hydroxy ketone (**121**)[121] following a protocol that been previously developed in the group (Scheme 21). Cleavage of

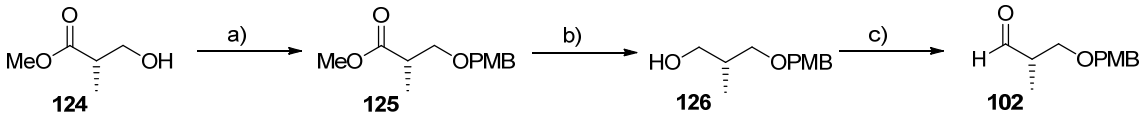
[2] The structure has been deposited in the Cambridge Crystallographic Data Base (deposition number CCDC 808039).

the *Oppholzer* sultam in **121** with $LiOH/H_2O_2$ afforded acid **122**, which was reduced to alcohol **123** with $BH_3{\cdot}SMe_2$ in the presence of stoichiometric amounts of $B(OMe)_3$. Note that if the $B(OMe)_3$ was not distilled freshly before use, the yield of this reaction would drop dramatically. Treatment of **123** with TFA in acetone then provided the desired ketone **43** in 38% yield for the three-step sequence from β-hydroxy imide **121**.[121]



Scheme 21: a) LiOH, H_2O_2, THF/H_2O, 70%; b) $BH_3{\cdot}SMe_2$, $B(OMe)_3$, THF, 15 °C, 56%; c) TFA, acetone, 96%.

α-Chiral aldehyde **102** was prepared from Roche ester **124** by PMB protection, reduction of the ester moiety with LAH and reoxidation of the resulting primary alcohol under *Swern* conditions (Scheme 22). Attempts at the direct conversion of the ester **125** into aldehyde **102** was accompanied by overreduction to alcohol **126** (5%).[3]

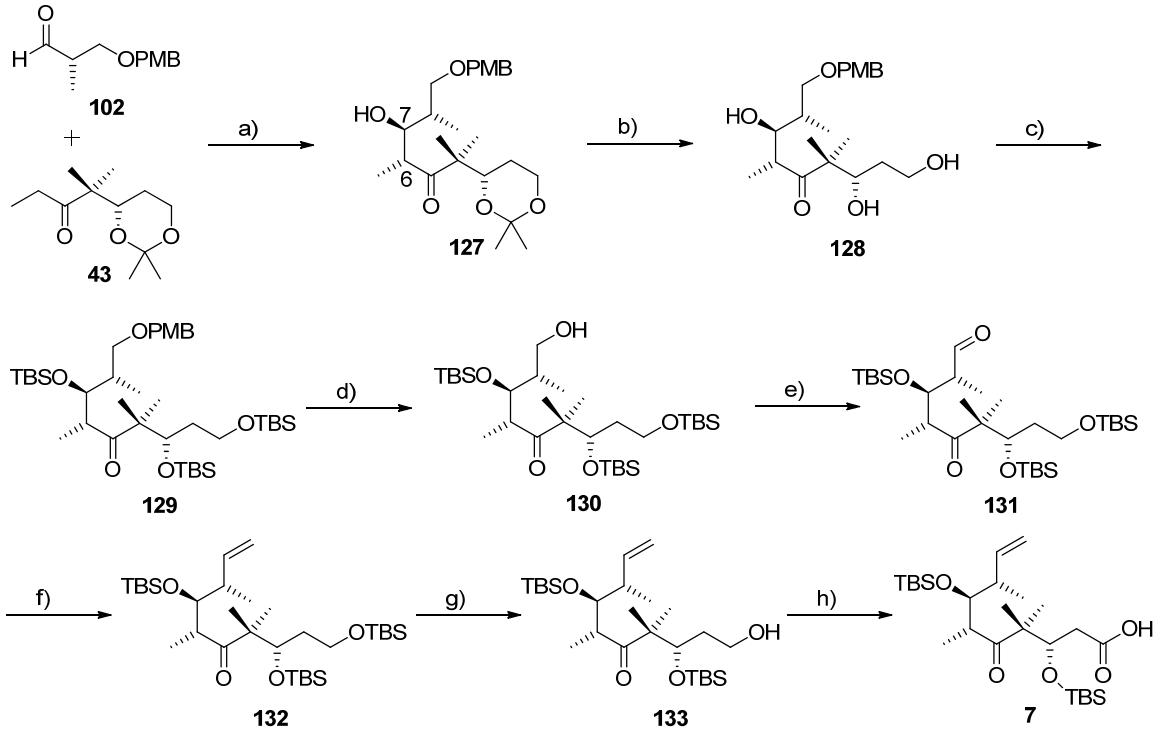


Scheme 22: a) *p*-Methoxybenzyl trichloroacetimidate, PPTS, CH_2Cl_2, 95%; b) LAH, Et_2O, 0 °C, 91%; c) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to 0 °C, used without purification.

With ketone **43** and aldehyde **102** in hand, the stage was set for the crucial aldol addition. Treatment of **43** with LDA at -78 °C followed by addition of **102** gave aldol product **127** in 76% yield and a diastereomeric ratio of 8:1 (Scheme 23). The undesired *syn*-isomer could be removed from the desired diastereomer **127** by silica gel chromatography. Cleavage of the acetal moiety under acidic conditions to give diol **128** followed by (re)protection of all free hydroxy functionalities as TBS-ethers gave **129** in 94% yield overall two steps. Quite surprisingly, ^1H- and ^{13}C-NMR analysis of the tris-TBS ether **129** indicated this material to be an inseparable 6:1 mixture of isomers, which was assumed to be the result of racemization of

[3] Aldehyde **102** racemizes on silica gel and therefore was not purified by means of columns chromatography.

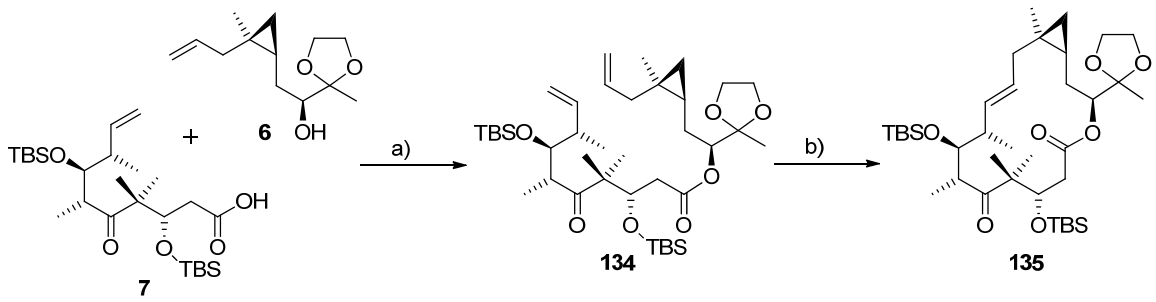
aldehyde **102** prior to aldol addition.[4] This tentative assignment of the minor component of this mixture as the 6*S*,7*R*,8*R* isomer of **129** was based on the fact that split signals were observed only for a few resonances, thus indicating the presence of stereoisomers. In principle, racemization of **102** prior to aldol addition should also have led to the formation of small quantities of a fourth (6*R*,7*S*,8*R*) diastereomer, which was, however, not detectable. Presumably this isomer is present only in quantities below the detection limit of the ^{1}H-NMR analysis; alternatively, it may have been removed by chromatography. It was anticipated that the minor diastereomer would be separable from the desired product at a later stage of the synthesis. Catalytic hydrogenation of **129** gave primary alcohol **130**, which was oxidized to aldehyde **131** with TPAP/NMO (Scheme 23).[122] **131** underwent *Wittig* olefination, thus establishing the terminal double bond required for the RCM reaction. Selective deprotection of **132** followed by oxidation of the resulting free primary hydroxy group to the carboxylic acid stage produced **7**, which was thus obtained in 57% overall yield from aldol product **127**. Unfortunately, the undesired diastereomer formed in the aldol step could not be removed in any of the steps leading from **127** to acid **7**, and the latter was still present as an 8.5:1 diastereomeric mixture.



Scheme 23: a) LDA, THF, -78 °C, 76%, dr 8:1; b) PPTS, MeOH, 96%; c) TBSOTf, 2,6-ludidine, CH_2Cl_2, -78 °, 98%; d) H_2, Pd/C, EtOH, 92%; e) TPAP, NMO, CH_2Cl_2, 4Å mol. sieves, used without purification; f) $MePPh_3Br$, LiHMDS, **131**, THF, 0 °C, 89%; g) CSA, CH_2Cl_2/MeOH, 89%; h) PDC, DMF, 82%.

[4] A ^{13}C (1k scans) experiment of the aldol product **127** confirmed the presence of the other substance.

Alcohol **6** and acid **7** were subjected to esterification under *Yamaguchi* conditions[78] giving ester **134** in 94% yield (Scheme 24); the use of EDCI only afforded 60% of **134**. Initial attempts at RCM of **134** using the *Grubbs* 2nd generation catalyst in toluene at 85 °C suffered from long reaction times (until full consumption of diene **134)** and from very low yields (<10%). The same was true for the *Hoveyda-Grubbs* 2nd generation catalyst, while *Grubbs* 1st generation catalyst did not trigger any cyclization. Delightfully, a change of the solvent from toluene to dichloromethane provided the solution to both problems, the long reaction times as well as the low yield. Diene **134** underwent cyclization in the presence of *Grubbs* 2nd generation catalyst (12 mol%) in refluxing dichloromethane in 80% yield and with excellent selectivity (*E*/*Z* 12:1) within 8 hours (Scheme 24).[113]



Scheme 24: a) 2,4,6-trichlorobenzoyl chloride, NEt_3, DMAP, benzene, 94%; b) *Grubbs* II (12 mol%), CH_2Cl_2, reflux, 80%, *E*/*Z* 12:1.

Fortunately, the undesired 6*S*,7*R*,8*R* isomer originating from the aldol addition could finally be removed at the stage of the macrocycle **135**. In the next step the C9/C10 double bond formed in the macrocyclization step was to be reduced, a transformation that proved to be highly challenging. In a first series of experiments the use of triisopropylbenzylsulfonylhydrazide ($TrisNHNH_2$) was investigated in different solvents and the base was varied (Table 2, Entries 1 to 3). The conversion for the reaction was very slow and little product was formed that could not be purified. With $KCO_2N{=}NCO_2K$ (PADA) (24 to 50 eq.) as a hydrogen source only little conversion was observed after 24 hours and the reaction did not proceed any further. At the same time hydrogenations suffered from simultaneous cyclopropane ring-opening. Thus, catalytic hydrogenation over palladium on activated charcoal gave the desired product in 45% yield together with substantial amounts of the cyclopropane opened side product.[5] Similar observations were made with *Crabtree* catalyst;[123] the reaction was somewhat slower, but more selective for the reduction of the

[5] ^{1}H-NMR spectrum showed the presence of an additional methyl group and the resultion mass spectrometry confirmed the presence of a mass of +2 relative to the desired product **136**. No effort was undertaken to characterize the side procuct any further.

double bond, leading to a yield of 56%. For both methods, variations in pressure and/or catalyst loading did not result in better yields, they only influenced the rate of the reaction. No reaction was observed with the *Wilkinson* catalyst.[124] After extensive optimization, hydrogenation over *Lindlar* catalyst at a hydrogen pressure of 7.5 bar was found to provide the saturated macrocycle most efficiently (80% yield). While cyclopropane ring-opening could not be avoided completely even under these conditions, it occurred at a much slower rate than double bond reduction (Table 2, Entry 9).

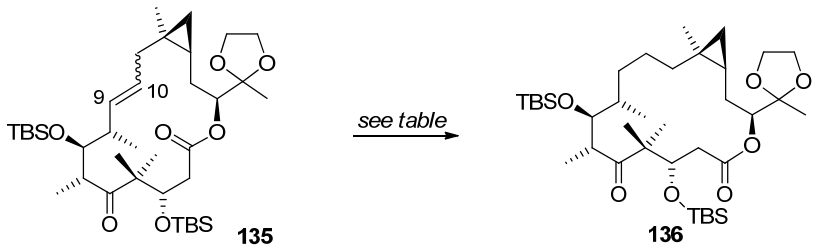
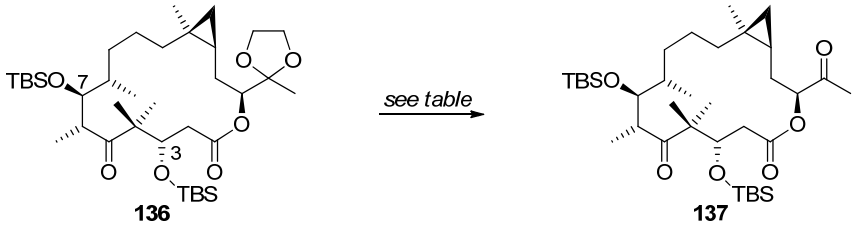


Table 2: Reduction of the C9/C10 double bond.

Entry	Reagent		Solvent	Reaction
1	$TrisNHNH_2$ (10-30 eq.)	NEt_3	DCE	little product formation
2	$TrisNHNH_2$ (30 eq.)	K_3PO_4	THF	little product formation
3	$TrisNHNH_2$ (30 eq.)	K_3PO_4	MeCN	no product formed
4	PADA (24 eq.)	AcOH	CH_2Cl_2	8% conversion after 12h, reisolated SM
5	PADA (50 eq.)	AcOH	MeOH	10% conversion after 12h, reisolated SM
6	Pd/C (0.05-0.4 eq.)	7.5 bar H_2	EtOH	45% product for 0.4 eq. catalyst used (12 h)
7	*Crabtree* cat (0.05-0.4 eq.)	7.5 bar H_2	CH_2Cl_2	56% product for 0.4 eq. catalyst used (24 h)
8	*Wilkinson* cat. (1.2 eq.)	7.5 bar H_2	THF	no product formed
9	*Lindlar* cat (0.5 eq.)	7.5 bar H_2	EtOH	80% product (14 h)

With this obstacle removed the next challenge was the hydrolysis of the acetal moiety in **136** in the presence of the two TBS-ethers at C3 and C7. Unfortunately, the selective hydrolysis/removal of the cyclic acetal could not be accomplished under a variety of conditions investigated (Table 3).[113]

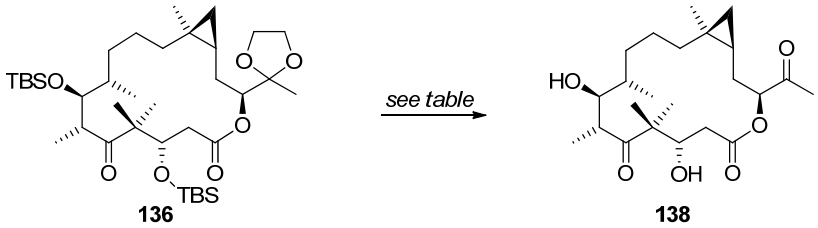


136 **137**

Table 3: Attempts to selectively hydrolyze the acetal in **136**.

Entry	Reagent	Solvent	Reaction
1	PPTS (0.1-2 eq.)	acetone	product misses one TBS-group
2	AcOH	THF/H_2O	4 different products (unkown)
3	PPh_3, CBr_4	THF	one product (unkown)
4	$CuCl{\cdot}2H_2O$	MeCN	4 different products (unknown)
5	$FeCl_3{\cdot}6H_2O$ (1.2-3.5 eq.)	CH_2Cl_2	TBS groups lost, some acetal hydrolyzed
6	*p*-TSA (1.3 eq.)	acetone/H_2O	TBS groups lost, acetal remained
7	HCl (2M)	THF	one TBS group lost, acetal uncleaved
8	NaI, $CeCl_3{\cdot}7H_2O$	MeCN/H_2O	one TBS group lost, little acetal hydrolyzed
9	Montmorrilonite K10	benzene	one TBS group lost, little acetal hydrolyzed
10	Thiourea	EtOH/H_2O	no reaction
11	$PdCl_2(MeCN)_2$	acetone	one TBS group lost, little acetal hydrolyzed

In response to these difficulties the reduction product **136** was submitted to global deprotection (which was to be followed by reprotection of the hydroxy groups). While selectivity was no longer an issue, most attempted conditions did not yield the deprotected product **138** in good yield (Table 4). It was best achieved with $FeCl_3{\cdot}6H_2O$.

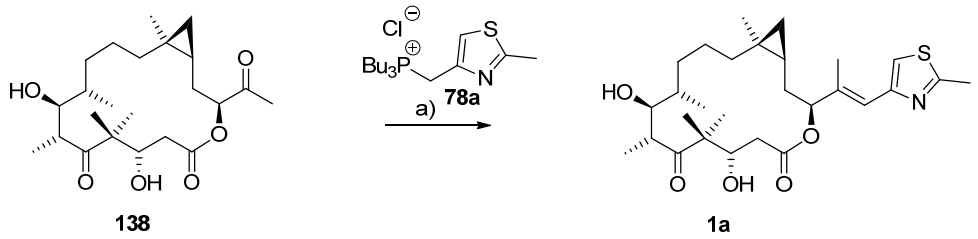


136 **138**

Table 4: Global deprotection of **136**.

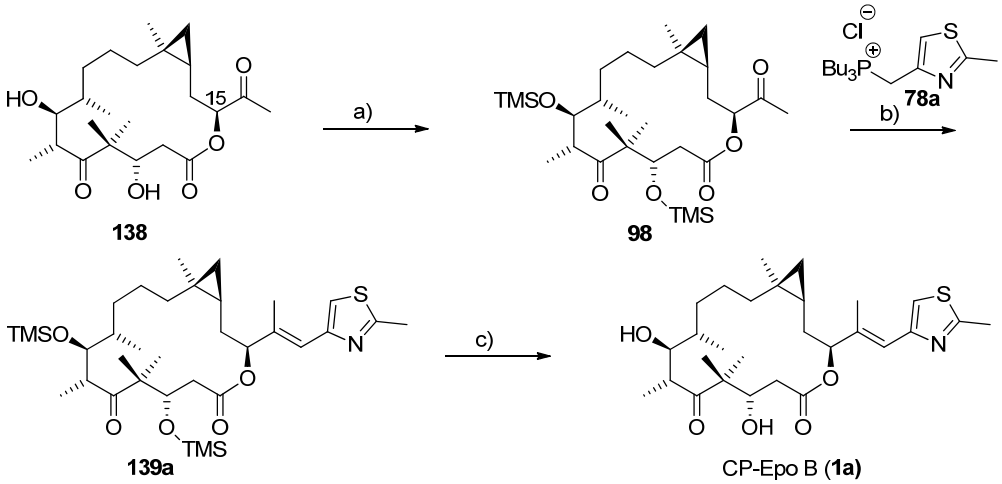
Entry	Reagent	Solvent	Reaction
1	Montmorrilonite K10	benzene	product formed in low yield
2	Amberlyst 15	dioxane/H_2O	only TBS groups got deprotected
3	CSA (3.5 – 10 eq.)	CH_2Cl_2/MeOH	product formed in low yield
4	TMSI, NaI	MeCN	product formed in low yield
5	$FeCl_3{\cdot}6H_2O$ (5 eq.)	CH_2Cl_2	85% yield

As there are a few literature examples of *Wittig* reactions in the presence of free secondary hydroxy groups,[125],[126] it was attempted to attach the thiazole heterocycle to methyl ketone **138** (Scheme 25). First, NaHMDS was used as a base, but this did not result in any product formation.[6] Next, KHMDS was used as a base together with 6 equivalents of the ylide; these conditions afforded CP-Epo B **1a** in 26% yield. Attempts to improve the yield by varying the number of equivalents of the ylid (4 and 9 eq.) failed and only traces of **1a** could be isolated. In light of the moderate yield achieved with ketone **138**, subsequent reactions were conducted with protected hydroxy groups.[113]



Scheme 25: a) KHMDS, **138**, THF, -78 °C to 0 °C, 0-26%.

While TMSCl turned out not to be reactive enough to protect both alcohol moieties, the use of TMSOTf gave bis silylether **98** in good yield without causing any racemization at C15 (Scheme 26). **98** underwent *Wittig* olefination in reasonable yield and selectivity to give thiazole derivate **139** in 67% yield and an *E*/*Z* ratio of 6:1. Final deprotection under acidic conditions afforded CP-Epo B (**1a**). In summary, the first total synthesis of CP-Epo B (**1a**) was developed in 8% overall yield and 19 steps in the longest linear sequence. CP-Epo B (**1a**) has been previously prepared by semisynthesis from Epo D, albeit in low yield.[61]

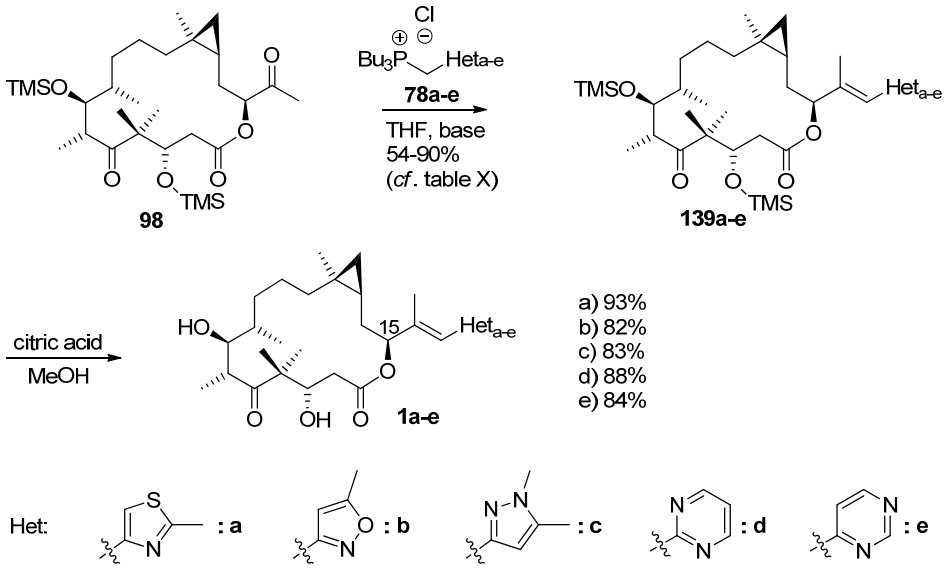


Scheme 26: a) TMSOTf, 2,6-lutidine, CH_2Cl_2, -78 °C, 100%; b) KHMDS, THF, -78 °C, then **98**, -78 °C to -20 °C, 67%, *E*/*Z* 6:1; c) Citric acid, MeOH, 93%.

[6] The *Wittig* salt **43** was prepared from the corresponding commercially available chloride in a single step.

1.5.1.3 Synthesis of Side Chain-modified Analogs of CP-Epo B

As illustrated in Scheme 27 and Table 5, methyl ketone **98** proved to be a highly suitable substrate for heterocycle attachment *via Wittig* olefination not only for the preparation of CP-Epo B (**1a**), but also for a series of different analogs. In all cases investigated, the reactions proceeded with good selectivity in favor of the desired *E* isomers which could be isolated in acceptable to good yields (Table 5). However, distinct differences were observed between different phosphoranes with regard to selectivity and also reactivity (Table 5). Thus, while **139a-c** were already formed upon warming of the reaction mixture to -20 °C (after deprotonation of the phosphonium salts at -78 °C and addition of **98** at the same temperature), the reaction of **98** with the phosphoranes derived from **78d** and **78e** required temperatures of 25 °C and, quite remarkably, 75 °C, respectively. Attempts to accelerate the reaction of **98** with the phosphorane derived from **139d** by raising the temperature to 75 °C resulted in decomposition.[113]



Scheme 27: Synthesis of target structures **1a-e**.

Table 5: *Wittig* reactions with ketone **98**.

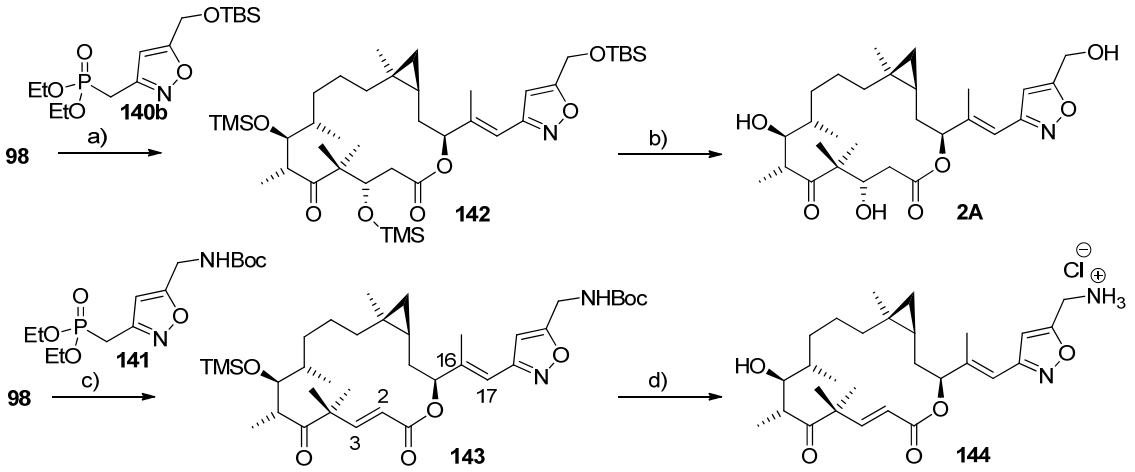
***Wittig* salt**	**Base**	**Temperature**	**Product**	***E*/*Z***	**Yield**
78a	KHMDS	-78 to -20 °C	**1a**	6:1	67%[a]
78b	KHMDS	-78 to -20 °C	**1b**	17:1	74%[a]
78c	*n*-BuLi	-78 to -20 °C	**1c**	13:1	85%[a]
78d	*n*-BuLi	-78 to 25 °C	**1d**	10:1	90%[b]
78e	*n*-BuLi	-78 to 75 °C	**1e**	7:1	54%[b,c]

[a] Mixture of *E* and *Z* isomers. [b] Mixture of *E* isomer and presumed C15 epimer (see text). [c] 77% based on recovered starting material.

Noteworthy, after chromatographic separation of **1d** and **1e** from the corresponding *Z* isomers, their ^{1}H- and ^{13}C-NMR spectra still indicated the presence of an isomeric impurity that could not be separated (ca. 12%). The identity of this impurity was not established, but it is well conceivable that the stereocenter at C15 partially epimerizes under the more forcing conditions of the *Wittig* reaction required for **1d** and **1e**.[113]

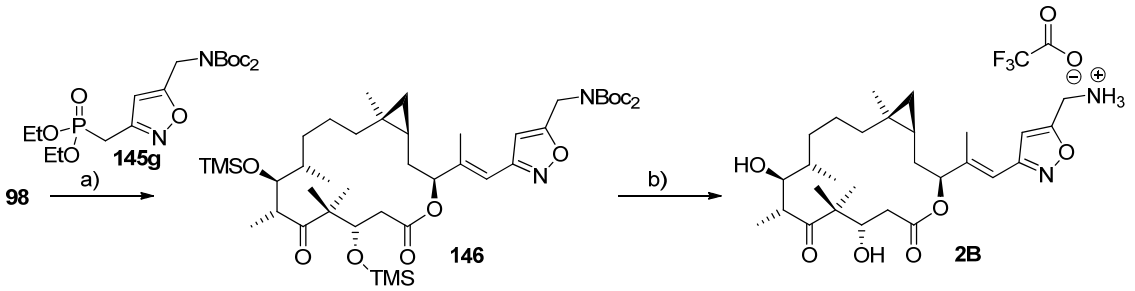
As for CP-Epo B (**1a**) deprotection of **1b-e** was achieved with citric acid to provide CP-Epo B analogs **1b-e** in excellent yields (Scheme 27). For **1d**, the minor isomer formed in the *Wittig* reaction could be removed by preparative HPLC, whereas **1e** could only be obtained as a 10:1 mixture of the desired structure and its presumed C15 epimer (and was evaluated as such in cellular experiments).

In a next phase, the *HWE* reaction of methyl ketone **98** with functionalized isoxazole heterocycles **140b** and **141** was investigated, in order to provide derivates that would be suitable for antibody conjugation (Scheme 28, for the synthesis of **140b** and **141** *vide infra*). Phosphonate **140b** was coupled to methyl ketone **98** to provide **142** as a separable 3:1 *E*/*Z* mixture in 97% yield; the *E*-isomer was isolated in 68% yield and the *Z*-isomer in 28% yield. The *E* and *Z* isomer were individually deprotected with CSA to give functionalized CP-Epo B analogs **2a** and **2Za** (only shown for **2a** in Scheme 28). The reaction of mono-Boc protected phosphonate **141** with methyl ketone **98** surprisingly did not yield the expected coupling product, instead the C2/C3 elimination product **143** was formed in 28% yield (inseparable mixture of an C16/C17 *E*/*Z* 4:1 mixture). The mixture was deprotected with TFA to yield 3-deoxy-2,3-dehydro CP-Epo B analog **144** in 94% yield; silica gel chromatography gave 69% of the desired C16/C17 *E* isomer.



Scheme 28: a) LiHMDS, THF, -78 °C, **98**, -78 °C to rt, 97%, *E*/*Z* 3:1; b) CSA, CH_2Cl_2/MeOH, 0 °C, 90%; c) *n*-BuLi, THF, -78 °C, **98**, -78 °C to rt, 28%, *E*/*Z* 4:1; d) TFA, CH_2Cl_2, 94%.

Varying the base used did not allow suppression of the elimination. As it was felt that the NH proton in **141** could be responsible for the observed elimination, the amino group in **141** was double-Boc protected; *HWE* olefination with the doubly protected phosphonate **145g** resulted only in the formation of the desired coupling product **146**, which was obtained in 52% yield as a single isomer (Scheme 29). No elimination products were observed in this reaction. Final deprotection using TFA gave CP-Epo B analog **2B**.



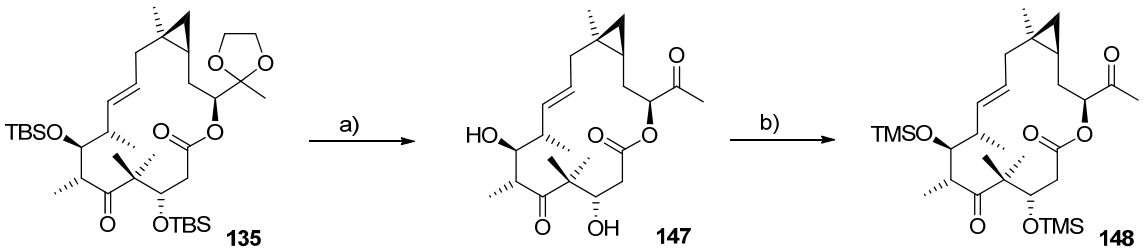
Scheme 29: a) LiHMDS, THF, -78 °C, **98**, -78 °C to rt, 52%, single isomer; b) TFA, CH_2Cl_2, 94%.

1.5.1.4 Analogs of 9,10-dehydro Epo B

As discussed in section 1.2.3.2, the incorporation of an *E* double bond at C9/C10 in Epo B or D leads to enhanced antiproliferative activity or the corresponding dehydro compounds are at least equipotent with the corresponding saturated variants. As shown above, the ring-closing metathesis reaction of diene **134** not only gave the macrocyclic product in good yield, but also with high selectivity in favor of an *E*-configured isomer that was separable from the minor *Z* isomer. This led us to prepare a series of side chain modified analogs of

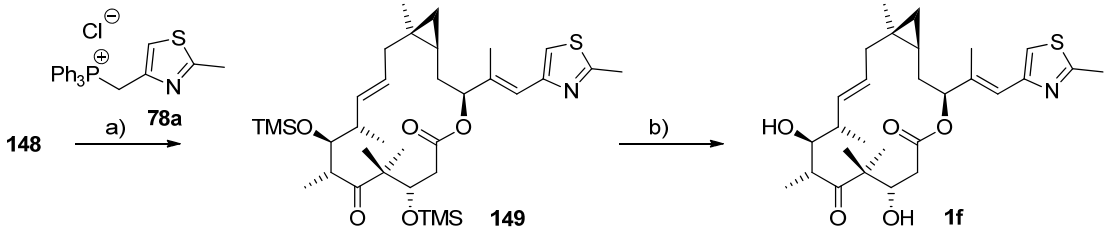
C9/C10-dehydro CP-Epo B and to evaluate their biological activity and potential suitability for antibody conjugation.

As illustrated in Scheme 30, the global deprotection of macrolactone **135** was again carried out with $FeCl_3 \cdot 6H_2O$. The resulting diol **147** was double TMS-protected to give methyl ketone **148** in 83% yield over two steps.



Scheme 30: a) $FeCl_3 \cdot 6H_2O$, CH_2Cl_2, 85%; b) TMSOTf, 2,6-lutidine, CH_2Cl_2, -78 °C, 98%.

In order to establish a reference point for the activity of 9,10-dehydro CP-Epo B analogs thiazole-derived analog **1f** was synthesized first (Scheme 31). The *Wittig* reaction of methylketone **148** with the phosphorane generated from **78a** gave **149** in 79% yield as an inseparable 7:1 *E*/*Z* mixture; deprotection with citric acid then delivered 9,10-dehydro CP-Epo B analog **1f** together with its C16/C17 *Z* isomer in 92% yield. The *Z* isomer resulting from the *Wittig* reaction could be separated by preparative HPLC. Compound **1f** demonstrated to be about 2-fold more active than its saturated analog **1a**.



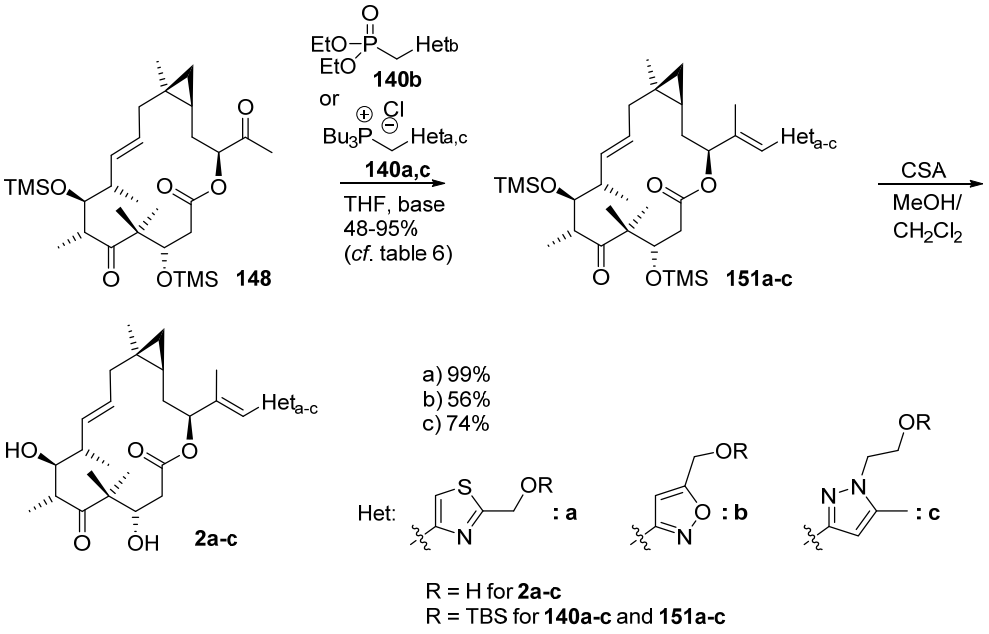
Scheme 31: a) KHMDS, THF, -78 °C, **148**, -78 °C to -20 °C, 79%, *E*/*Z* 7:1; b) citric acid, MeOH, 92%.

The coupling of the functionalized thiazole heterocycle to methyl ketone **148** was first attempted using the phosphonate **150**, which had already been synthesized in sufficient amounts (Section 1.5.1.9, Scheme 41). However, independent of the base used only decomposition was observed in the reaction and no product was formed. Hence, the *Wittig* salt **140a** was synthesized and subjected to reaction with methyl ketone **148**. This approach produced the desired product in 75% yield as a separable 7:1 mixture of *E*/*Z* isomers

(Scheme 32, Table 6). Deprotection of **151a** with CSA gave thiazole-based 9,10-dehydro CP-Epo B analog **2a** in quantitative yield.

A remarkable observation was made for the coupling of isoxazole-based phosphonate **140b** to the ketone **148**. If KHMDS was used as a base, none of the desired product was formed, but the C2/C3 elimination product was isolated in 78% yield as a 1:1 mixture of C16/C17 *E*/*Z* isomers. As the incorporation of an *E* double bond at C2/C3 in epothilones is known to be well tolerated in terms of biological activity (Section 1.2.3.3),[76] the inseparable mixture was deprotected under acidic conditions and the resulting 2,3-dehydro analogs **2d** and **2e** were separated by preparative HPLC. Quite intriguingly, a simple change of the counter ion of the base employed in the *HWE* reaction reversed the outcome. Thus, if LiHMDS was used as a base only the *HWE* product **151b** was formed in excellent yield of 95%, albeit in an unselective manner (*E*/*Z* 1:1, separable mixture). Both isomers were individually deprotected in the presence of CSA to give isoxazole-based analogs **2b** and its C16/C17 *Z* isomer **2Zb** in yields of 56% (*E* isomer) and 67% (*Z* isomer).

The pyrazole-based *Wittig* salt **140c** underwent olefination with methyl ketone **148** in 48% yield to deliver a separable 2:1 mixture of *E*/*Z* isomers. The *E* isomer was deprotected with CSA to give pyrazole-based 9,10-dehydro CP-Epo B analog **2c** in 74% yield.



Scheme 32: Synthesis of target structures **2a-c**.

Table 6: *Wittig* and *HWE* reactions with ketone **148**.

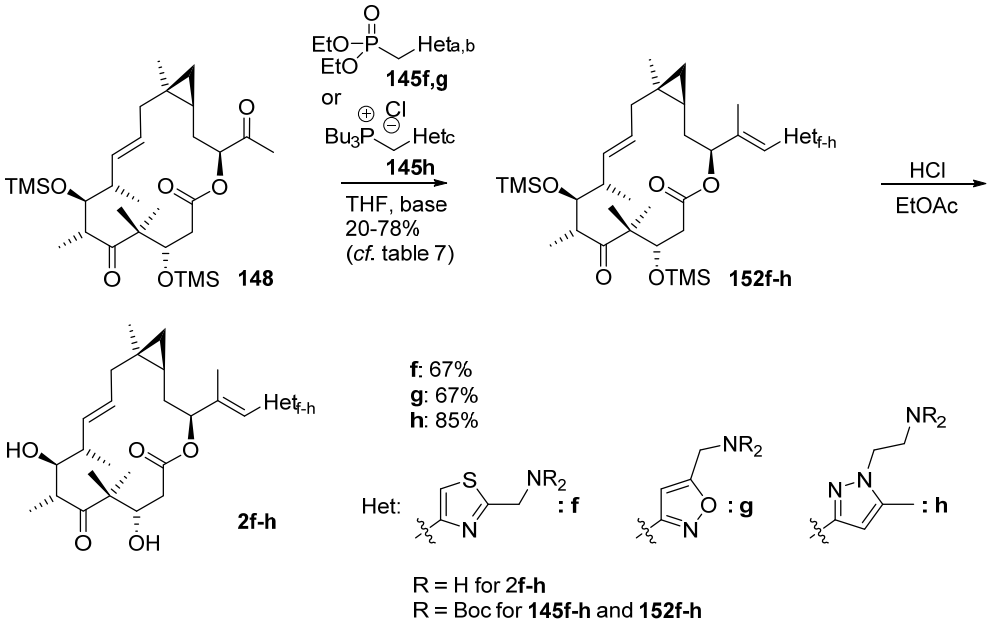
	base	**temperature**	**product**	***E*/*Z***	**yield**
***Wittig* salt 140a**	*n*-BuLi	-78 °C to 0 °C	**2a**	7:1	75%[a]
Phosphonate 140b	LiHMDS	-78 °C to -20 °C	**2b**	1:1	95%[a]
***Wittig* salt 140c**	*n*-BuLi	-78 °C to rt	**2c**	2:1	48%[a]

[a] Mixture of *E* and *Z* isomers.

The synthesis of analogs bearing free amino groups employed phosphonates derived from the corresponding double-Boc protected aminoalkyl heterocycles. In analogy to previous observations in the synthesis of **2B** the coupling of mono-protected amines only led to elimination of water across the C2/C3-bond or/and gave only very low yields of the desired coupling product and the elimination product. Thus, double-Boc protected thiazole phosphonate **145f** underwent *HWE* olefination with methyl ketone **148** to give the desired product in 42% yield as a single isomer (Scheme 33, Table 7). This contrasts with the behaviour of the corresponding TBS protected phosphonate **150** (Section 1.5.1.9, Scheme 41), where only decomposition was observed in the attempted reaction with **148**. Removal of the Boc groups by the use of hydrochloric acid in EtOAc yielded **2f** in 67% yield.

In contrast to thiazole-derived phosphonate **145f**, coupling of the isoxazole derivate **145g** to the macrocycle yielded 78% of a 3.5:1 mixture of separable *E*/*Z* isomers. Both isomers were individually deprotected with hydrochloric acid to give isoxazole-based analog **2g** in 67% yield and its C16/C17 *Z* isomer **2Zg** in 93% yield.

Pyrazole-derived *Wittig* salt **145h** only underwent olefination in 20% yield with a 10:1 ratio of inseparable *E*/*Z* isomers. It is well conceivable that the phosphonate corresponding to **145h** would have been the better choice for the elaboration of the aminoethyl pyrazole side chain in **2h**. However, as sufficient material was obtained with the phosphorane this possible way to improve the yield of the coupling reaction was not evaluated. Deprotection using hydrochloric acid in EtOAc gave the corresponding pyrazole-based analog **2h** in 85% yield as a single isomer. All 9,10-dehydro CP-Epo B analogs **2f-h** were obtained as hydrochlorides.



Scheme 33: Synthesis of target structures **2f-h**.

Table 7: *Wittig* and *HWE* reactions with ketone **148**.

X	Base	Temperature	Product	*E*/*Z*	Yield
Phosphonate 145f	LiHMDS	-78 °C to rt	**2f**	single isomer	42%
Phosphonate 145g	LiHMDS	-78 °C to -20 °C	**2g**	3.5:1	78%[a]
***Wittig* salt 145h**	*n*-BuLi	-78 °C to rt	**2h**	10:1	20%[a]

[a] Mixture of *E* and *Z* isomers.

1.5.1.5 Biological Evaluation

All compounds were evaluated for their antiproliferative activity against the human cancer cell lines A549 (lung), MCF-7 (breast) and HCT116 (colon). These experiments were carried out in collaboration with Prof. Jürg Gertsch, University of Berne. Cells were exposed to the compounds for 72 h.

As for CP-Epo B (**1a**) and its analogs with unfunctionalized heterocycles in their side chains (**1b-e**), all compounds are potent inhibitors of cancer cell proliferation, with IC_{50} values in the single digit nM or even sub-nM range (Table 8).

Table 8: Antiproliferative activity of Cp-Epo B analogs **1a-e** (IC_{50} values [nM]).

Cell line	1a	1b	1c	1d	1e
A549	0.70±0.2	0.30±0.07	4.7±0.8	0.90±0.13	1.7±0.15
MCF-7	0.80±0.3	0.40±0.06	8.5±1.5	0.80±0.19	1.9±0.23
HCT116	1.30±0.2	0.30±0.03	3.8±0.7	0.40±0.08	0.9±0.07

[a] IC_{50} values of 0.33, 0.34, and 0.16 nM have been reported for Epo B against the A549, MCF-7, and HCT116 cell lines respectively.[127]

Compared to Epo B, CP-Epo B (**1a**) appears to be 2-8-fold less active; IC_{50} values similar to the corresponding epoxide-based analogs[7] were also observed for **1d**/**1e** (< 3-fold difference in all cases), although **1d** and **1e** showed a tendency for slightly enhanced activity. The most potent compound investigated was isoxazole derivative **1b**, which is in line with previous findings by the *Danishefsky* group on the activity of isoxazole-containing variants of 9,10-dehydro-12,13-deoxy-epothilones.[84] In light of its sub-nM potency **1b** is an attractive candidate for the construction of ADC's.

With regard to CP-Epo B analogs **2A**, **2ZA**, **2B**, which bear a hydroxy- or aminomethyl group attached to the heterocycle, compound **2A**, quite intriguingly was found to be about as active as Epo B and its parent unfunctionalized isoxazole CP-Epo B (**1b**). Unfortunately, the corresponding amine **2B** is about 10-fold less active than **2A**. A further significant decrease in activity (> 100-fold) is then observed for the C16/C17 *Z* isomer **2ZA** (Table 9).

[7] These compounds were kindly provided by the Novartis Institute for Biomedical Research in Basel, Switzerland.

Table 9: Antiproliferative activity of functionalized Cp-Epo B analogs **2A**, **2ZA**, **2B** ($IC_{50's}$ [nM]).

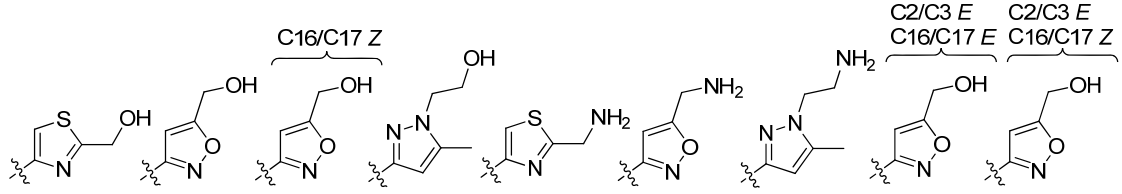
cell line	2A	2ZA	2B
A549	0.19±0.09	148±14	1.8±0.31
MCF-7	0.43±0.17	203±31	2.1±0.70
HCT116	0.20±0.10	170±12	1.5±0.92

IC_{50} values of 0.33, 0.34, and 0.16 nM have been reported for Epo B against the A549, MCF-7, and HCT116 cell lines respectively.

Based on its sub-nM potency **2A** was selected as an attractive candidate for the construction of ADC's (see below).

As for the effect of desaturation of CP-Epo B analogs at the C9/C10 position, the 9,10-deyhdro analogs of CP-Epo B **1f** and hydroxymethyl CP-Epo B **2b** appear to be as potent as their saturated, unfunctionalized parent compounds, which demonstrates that the incorporation of the hydroxy moiety is well tolerated and leads to equipotent analogs (Table 10). The 2,3-deyhdro isoxazole analog **2d** retains most of the activity of **2b**, which confirms previous observations by *Vite* and *co*-workers with Epo A/B and the corresponding C2/C3-dehydro analogs (Section 1.2.3.3, Figure 14).[76]

Table 10: Antiproliferative activity of Cp-Epo B analogs **2a-h** (IC_{50} values [nM])



cell line	2a	2b	2Zb	2c	2f	2g	2h	2d	2e
A549	0.75±0.10	0.25±0.08	29.4±1.5	5.1±0.73	0.76±0.06	1.73±0.53	364±12	2.4±0.53	13.6±0.83
MCF-7	0.82±0.09	0.36±0.09	23.7±2.0	7.0±0.59	0.69±0.10	2.32±0.29	530±27	1.8±0.66	16.3±1.30
HCT116	0.90±0.11	0.32±0.03	24.2±2.3	8.2±0.19	1.10±0.09	2.38±0.23	482±11	3.5±0.52	17.0±2.50

IC_{50} values of 0.33, 0.34, and 0.16 nM have been reported for Epo B against the A549, MCF-7, and HCT116 cell lines respectively.

For the three amino group-containing analogs **2f**, **2g**, and **2h** the picture looks different. While thiazole-derivative **2f** was as active as alcohol **2a**, the aminomethyl isoxazole CP-Epo B (**2g)** was about 10-fold less active than alcohol **2b**, which was in line with previous observations for the corresponding 9,10-saturated compounds (see Table 9). The substitution of the

pyrazole alcohol **2c** by an amine leads to almost inactive compound amine **2h**. Thus, the substitution of the hydroxy group in **2c** by an amino group leads to a > 100-fold loss in potency.

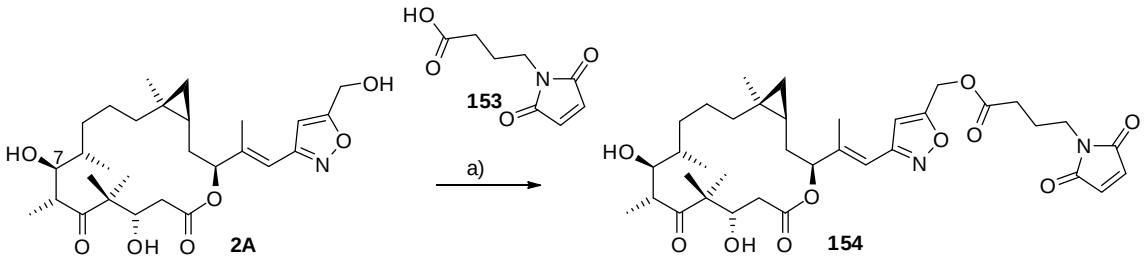
The two C16/C17 *Z* isomers investigated, **2Zb** and **2e,** are both significantly less active than the corresponding *E* isomers, but they still exhibit IC_{50} values in the low nM range.

The conclusions derived from the cellular experiments described above can be summarized as follows:

- In accordance with literature data for Epo, the substitution of the epoxide ring in Epo B by a cyclopropane moiety does not lead to any substantial change in antiproliferative activity.[61]
- Likewise, the incorporation of an *E* double bond at C9/C10 in side chain-modified CP-Epo B analogs maintains full activity or even leads to slightly more active analogs.
- The replacement of the thiazole heterocycle in CP-Epo B by an isoxazole moiety results in enhanced antiproliferative activity (similar to the effect of the same modification in fludelone (**92**)).[84]
- 3-Deoxy-2,3-didehydro CP-Epo B analogs retain almost the full activity of their parent compounds. This effect is again similar to what is observed for epoxide-based epothilones.[76]
- The introduction of hydroxyalkyl-modified heterocycles such as **2a** and **2b** is very well tolerated and produces analogs with potencies comparable to those of analogs with non-functionalized heterocycles for all attempted.
- In contrast, heterocycles bearing primary amino groups are clearly less potent than the corresponding primary alcohols. The reasons for this effect are unknown at this point, but similar observations have been reported for benzimidazole-based Epo B analogs with pendant aminoethyl or hydroxyethyl substituents on one of the benzimidazole nitrogens.
- The C16/17 *Z* isomers do not lose all activity but are still potent inhibitors of cancer cell proliferation with IC_{50} values in the low nM range.

1.5.1.6 Antibody Drug Conjugates

Based on its potent antiproliferative activity hydroxymethyl isoxazole CP-Epo B **2A** (Scheme 34) was selected for orientating experiments on the evaluation of antibody-drug conjugate. The conjugation work and the preliminary biological evaluation of the resulting ADC were carried out by Elena Perrino in the group of Prof. *Neri* at the ETH Zurich and shall be briefly summarized here. The antibody selected for this work was the SIP-F8 fragment, a small immunoprotein (SIP) consisting of two scFv fragments linked by five amino acids that recognizes the alternatively spliced extra-domain A (EDA) of fibronectin with high specificity.[128] This domain is overexpressed on neo-vasculature structures of solid tumors and is a well characterized marker of tumor angiogenesis.[129] As angiogenesis is a feature of the most aggressive solid cancers and the structures are accessible through the bloodstream, SIP-F8 is a promising candidate for directed tumor targeting.[128],[129] SIP-F8 contains one cystine disulfide bridge which can be reduced to the corresponding cysteines. The thiol groups of the cysteine side chains are sufficiently nucleophilic to undergo 1,4-addition reactions with suitable acceptor molecules. Therefore, a linker moiety bearing such an acceptor group was attached to CP-Epo B analog through the primary alcohol functionality in **2A**, which was esterified with acid **153** (Scheme 34). Conceptually, it was expected that the drug would be released from the ADC construct by slow hydrolysis in the tumor vasculature.[130] The esterification was carried out under *Yamaguchi* conditions giving ester **154** in 44% yield. Surprisingly, it turned out that ester **154** is not stable under the reaction conditions and yields of the reaction decreased with longer reaction times. If the reaction was quenched after 0.5 h the desired product **154** was isolated in 23% yield. A shorter reaction time of 20 min gave a yield of 44%. Further reduced reaction times might have resulted better yields, but this was not investigated. With an excess of the reagents, very surprisingly, esterification of the C7-hydroxy group was also observed. This observation indicated that the secondary alcohol moiety at C7 *per se* is nucleophilic enough to undergo esterification and, as a consequence, functionalized heterocycles may not be a requirement for the construction of epothilone-based ADCs.



Scheme 34: 2,4,6-Trichlorobenzoyl chloride, NEt_3, DMAP, benzene, 44%.

Ester **154** could be successfully attached to the SIP-F8 antibody as demonstrated by mass spectrometry (ESI) (Figure 25). (Work carried out by Elena Perrino).

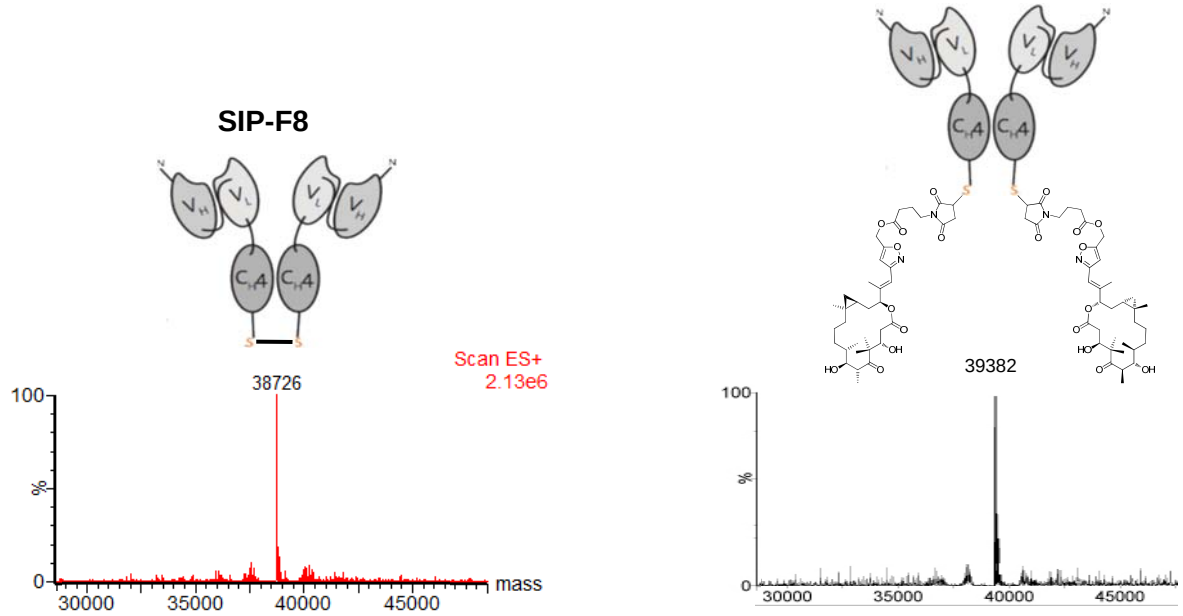
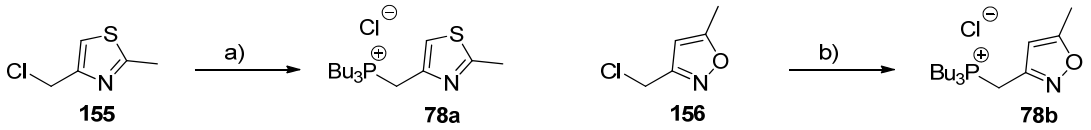


Figure 25: MS (ESI) of the SIP-F8 antibody; MS of the ADC (right).

Initial *in vivo* experiments with the ADC indicated a measurable antitumor effect of the conjugate (murine teratocarcinoma, 1.9 nmol/mouse a day). As the tumors were still growing, however, the dose of the ADC was increased (5.7 nmol/mouse a day) in a second experiment, but this resulted in body weight loss of the mice and, hence, the experiments had to be abandoned. No further evaluation of this particular ADC was undertaken.

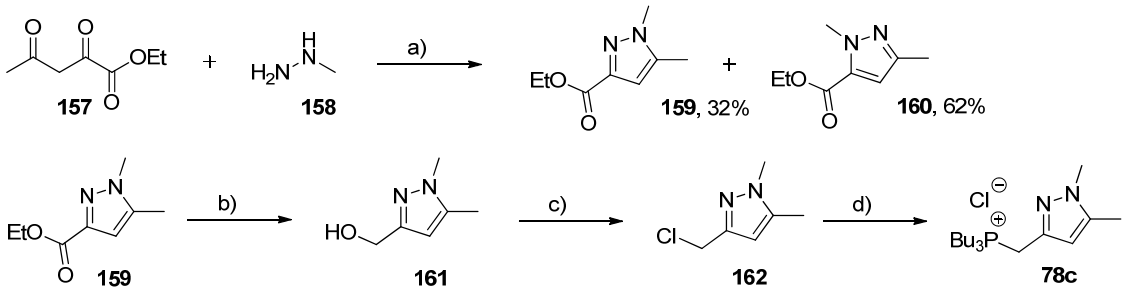
1.5.1.7 Synthesis of Unfunctionalized Heterocycles 78a-e

Phosphonium salts **78a** and **78b** were prepared from the corresponding commercially available chlorides **155** and **156** in high yield by heating the latter with PBu_3 in toluene or DMF, respectively (Scheme 35).



Scheme 35: a) PBu_3, 40 °C, toluene, 93%; b) PBu_3, 40 °C, DMF, 84%.

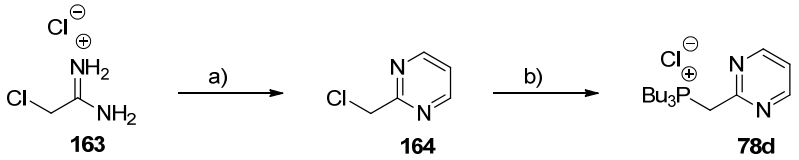
At the time the pyrazole-derived phosphonium salt **78c** was synthesized, a synthesis for the functionalized pyrazole-based phosphonium salt **140c** (Section 1.5.1.10, Scheme 50) had already been established. Thus, the same synthetic strategy could be followed. The four step sequence started from ethyl acetopyruvate **157** (Scheme 36). The latter was treated with methylhydrazine **158** to produce pyrazole **159** in 32% yield. Unfortunately, the undesired regioisomer **160** was the favoured product of the reaction and could be isolated in 62% yield. Variation of the temperature within the range from -78 °C up to 60° did not change the isomer ratio significantly. The presence or the absence of a NOE between the protons at the two methyl groups in **159** and **160** helped to assign the regioisomers. Although **159** was obtained only in moderate yield, in light of the very cheap starting materials this was considered acceptable. DIBAL-H reduction of ester **159** gave alcohol **161**, which was converted into chloride **162** with $SOCl_2$. Noteworthy, chloride **162** was found to be volatile. Finally, reaction of **162** with tributylphophine gave phosphonium salt **78c**.



Scheme 36: a) EtOH, 0 °C, 94%; b) DIBAL-H, CH_2Cl_2, -78 °C, 72%; c) $SOCl_2$, CH_2Cl_2, 99%; d) PBu_3, DMF, 82%.

Pyrimidine-derived phosphonium salt **78d** was synthesized from chloroacetimidate hydrochloride (**163**) and 1,1,3,3-tetramethoxypropane which reacted to give 2-(chloromethyl)pyrimidine **164** in 35% yield (Scheme 37). It should be noted that chloride

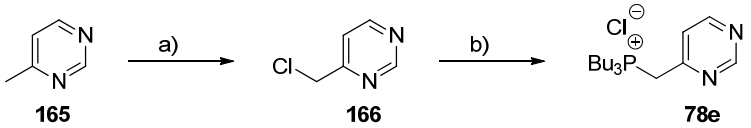
164 is volatile, which was unexpected and may have contributed to the moderate isolated yield; in contrast, 4-(chloromethyl)pyrimidine (**166**) (Scheme 38) is non-volatile. In spite of this unanticipated problem, a sufficient amount of chloride **164** was obtained, which was then transformed into phosphonium salt **78d** by treatment with tributylphosphine.



163 **164** **78d**

Scheme 37: a) 1,1,3,3-tetramethoxypropane, 35%; b) PBu_3, DMF, 91%.

The synthesis of 4-pyrimidine derivative **78e** in the first step involved the addition of trichloroisocyanuric acid to 4-methyl pyrimidine **165**, which furnished chloride **166** in 48% yield. 15% of the dichloro product was also isolated, while unconverted starting material accounted for the remainder of the mass balance. The yield of 48%, although seemingly moderate, is probably close to the optimum for the reaction, since chloride **166** is more reactive than the starting material. Treatment of **166** with tributylphophine gave *Wittig* salt **78e**.

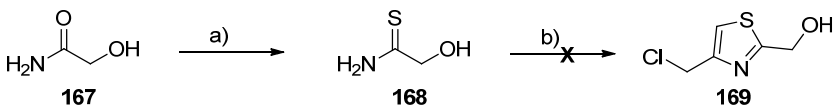


165 **166** **78e**

Scheme 38: a) Trichloroisocyanuric acid, $CHCl_3$, 48%; b) PBu_3, DMF, 98%.

1.5.1.8 Functionalized Thiazole Heterocycles

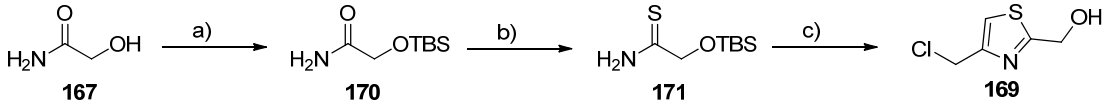
Our initial approach to hydroxymethyl thiazole-derived phosphonium salt **140a** was to proceed through 2-hydroxymethyl-4-chloromethyl-thiazole (**169**) (Scheme 39). The latter was initially thought to be obtained by reaction of 2-hydroxythioacetamide **168** with 1,3-dichloroacetone. However, while **168** was accessible from commercially available 2-hydroxyacetamide **167** by sulfurization with *Lawesson*'s reagent[131] in moderate yield (28%), the subsequent reaction of 1,3-dichloroacetone with the thioamide **168** did not lead the formation of the desired substituted heterocycle **169**.



167 **168** **169**

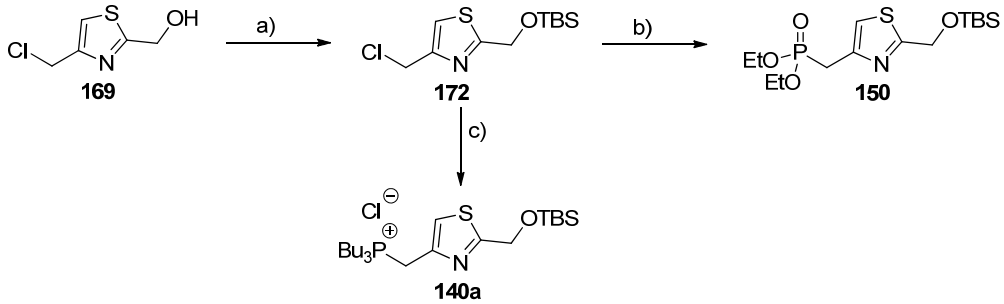
Scheme 39: a) *Lawesson*'s reagent, dioxane, reflux, 28%; b) 1,3-dichloroacetone, EtOH, no product formed.

It was suspected that protection of the alcohol functionality in **167** would enable the formation of the thiazole ring. Hence, the hydroxy group in amide **167** was protected as a TBS-ether in 96% yield; (Scheme 40); a TBS-protecting group was chosen as this would allow global deprotection of the ultimate target molecule in a single step. As for the introduction of the protecting group before the thiation step, it was hoped that would also lead to improved yields of the corresponding thioamide **171** over the unprotected thioamide **168**. Formation of the 2-TBSoxy thioacetamide **171** was most efficient in refluxing dioxane (67%; Scheme 40); toluene at 65 °C gave 49% of **171** and only 14% under reflux conditions. Subsequently, addition of 1,3-dichloroacetone to **171** gave exclusively the deprotected thiazole derivate **169** in 50% yield. In order to neutralize the hydrochloric acid formed during the reaction, pyridine (1.2 eq.) was added to the reaction mixture, which allowed the isolation of 10% of the TBS-protected thiazole derivate together with 20% of **169**. The addition of an excess of pyridine (10 eq.) resulted in complete decomposition of both products.



Scheme 40: a) TBSCl, imidazole, DMF, 96%; b) *Lawesson*'s reagent, dioxane, reflux, 67%; c) 1,3-dichloroacetone, EtOH, 50%.

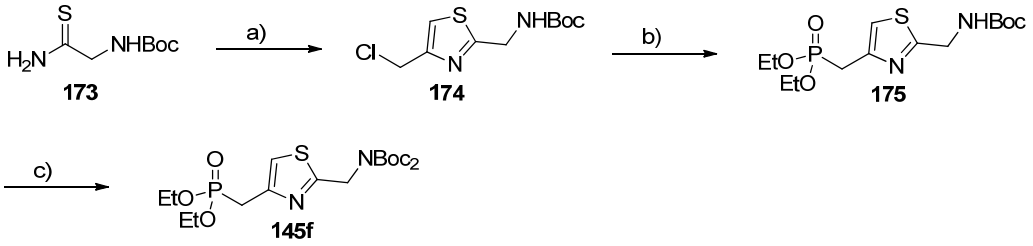
No further efforts were made to optimize the cyclization reaction. Rather, **169** was reprotected (Scheme 41). Finally, *Arbuzov* reaction of **172** with $P(OEt)_3$ afforded phosphonate **150**, albeit in low yield, while the formation of the *Wittig* salt **140a** proceeded in higher yield.



Scheme 41: a) TBSCl, imidazole, DMF, 91%; b) $P(OEt)_3$, 160 °C, 19%; c) PBu_3, DMF, 94%.

The synthesis of protected aminomethyl thiazole-based phosphonate **145f** followed the same approach as for **140a**. Treatment of commercially available thioacetamide **173** with 1,3-dichloroacetone gave chloride **174** in moderate yield, which was then transformed into phosphonate **175** under *Arbuzov* conditions in 87% yield (Scheme 42). As the *Wittig* reaction

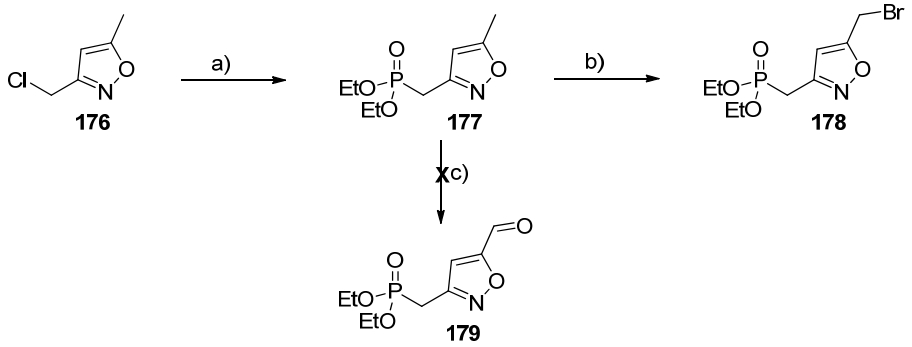
of mono-Boc protected heterocycles caused severe problems (*vide supra*) the amine was converted into the bis-Boc derivative **145f** by treatment with Boc_2O in 97% yield.



Scheme 42: a) 1,3-dichloroacetone, EtOH, 35%; b) $P(OEt)_3$, 160 °C, 87%; c) Boc_2O, DMAP, MeCN, 97%.

1.5.1.9 Functionalized Isoxazole Heterocycles

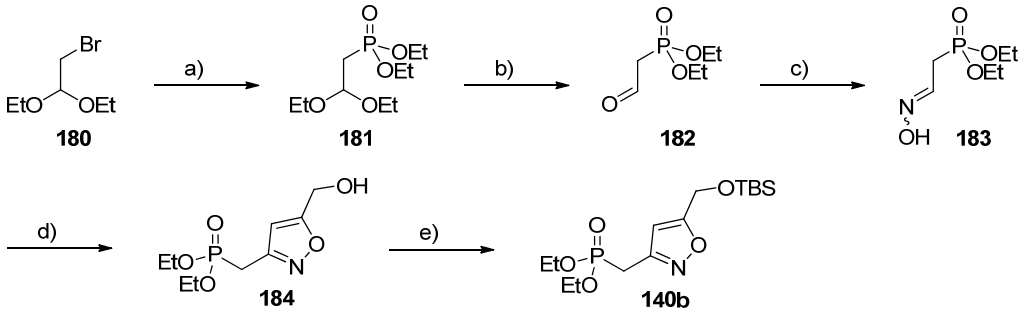
Initial attempts at the synthesis of the isoxazole derivative **140b** were based on the idea to establish the hydroxymethyl group by functionalization of an existing heterocycle. Thus, isoxazole derivate **176** was brominated with NBS to give bromide **177**, but the reaction suffered from low reproducibility, with yields varying between 20% and 60%. After the starting material had been consumed by about two thirds, dibromination would usually set in. As the yield tended to be lower on larger scale, the installation of an aldehyde functionality was investigated as an alternative to bromination, to provide a precursor for the hydroxymethyl group (Scheme 43). Unfortunately, selenoxide did not effect the desired transformation under a variety of conditions investigated.



Scheme 43: a) $P(OEt)_3$, 90%; b) NBS, AIBN, CCl_4, 20% to 60%; c) SeO_2, dioxane, reflux, no product.

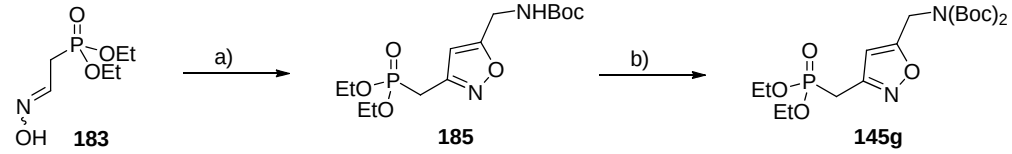
In light of these difficulties, an alternative strategy was elaborated, which relied on a 1,3-dipolar cycloaddition to construct the isoxazole heterocycle as reported by *Lee* and *co*-workers (Scheme 44).[132] The phosphonate group was installed at an early stage of the synthesis *via Arbuzov* reaction between bromide **180** and $P(OEt)_3$ which provided **181** in 70% yield. Hydrolysis of the acetal group in **181** gave aldehyde **182**, which was transformed into

the oxime **183** by addition of hydroxyamine in 75% yield. The stage was now set for ring construction by a cycloaddition reaction. Treatment of **183** with NCS in the presence of NEt_3 served to form the nitrile oxide *in situ*, which underwent 1,3-dipolar cycloaddition with propargylic alcohol to give isoxazole derivate **184** as the only regioisomer in 45% yield. Finally, the free hydroxyl group was TBS-protected to afforded the desired phosphonate **140b** in 21% overall yield from bromide **180**.



Scheme 44: a) $P(OEt)_3$, 70%; b) aq. HCl (2%), 94%; c) NH_2OH, $EtOH/H_2O$, 75%; d) NCS, NEt_3, $CHCl_3$, then propargylic alcohol, 45%; e) TBSCl, imidazole, DMF, 94%.

In order to access aminomethyl isoxazole **145g**, the nitrile oxide derived *in situ* from oxime **183** (Scheme 45) was submitted to 1,3-cycloaddition with *N*-Boc-propargyl amine, to give isoxazole **185** selectively in 55% yield. Reaction with Boc_2O then gave the desired bis-BOC derivative **145g** in 85% yield.

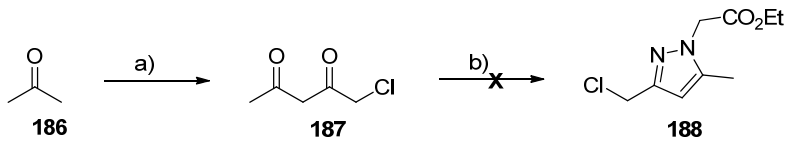


Scheme 45: a) NCS, NEt_3, $CHCl_3$, then *N*-Boc-propargyl amine, 55%; b) Boc_2O, DMAP, MeCN, 85%.

1.5.1.10 Functionalized Pyrazole Heterocycles

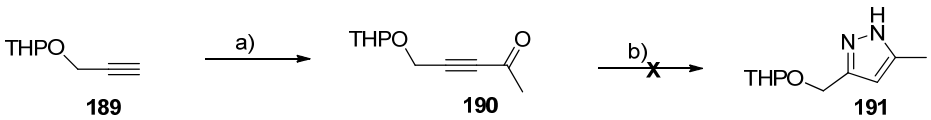
The initial approach towards the synthesis of pyrazole-derived phosphonates **140c** and **145g** was based on the formation of the chloromethyl pyrazole precursor **188** by the reaction of ethyl 2-hydrazinylacetate with 1,3-diketone **187** (Scheme 46). The latter was obtained by quenching the lithium enolate of acetone (**186**) with ethyl chloroacetate in 25% yield. The early introduction of the phosphonate group *via Arbuzov* reaction with the chloride failed, most probably due to the limited thermal stability of chloride **187**. Regrettably, addition of

ethyl 2-hydrazinylacetate to **187** did not result in the formation of the pyrazole heterocycle **188**.



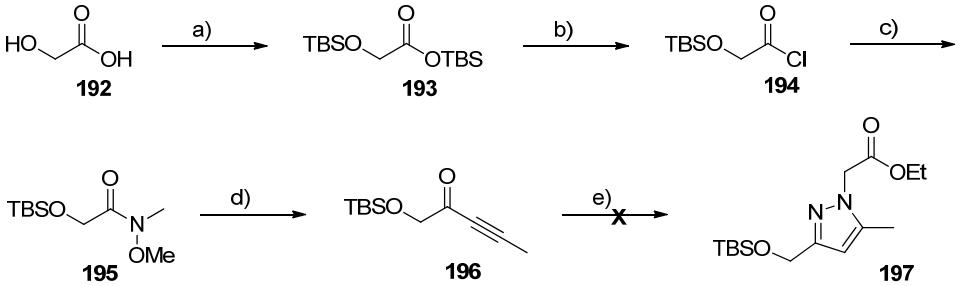
Scheme 46: a) LDA, ethyl chloroacetate, THF -78 °C, 25%; b) Ethyl 2-hydrazinylacetate, EtOH, reflux, no product formed.

Likewise, attempts to construct pyrazole derivative **191** by reaction of hydrazine with ynone **190** were unsuccessful (Scheme 47).



Scheme 47: a) *n*-BuLi, $ZnCl_2$, acetyl chloride, THF, -78 °C, 60%; b) hydrazine, MeOH, no product formed.

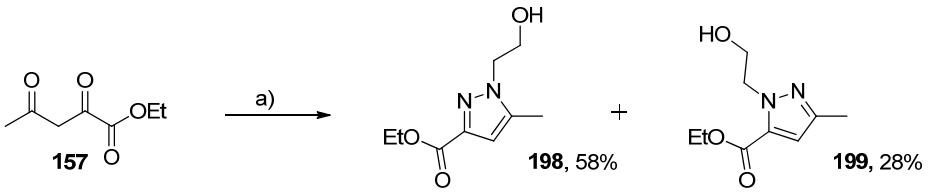
A very similar addition would have been the pivotal step in the following reaction sequence (Scheme 48). Glycolic acid (**192**) was doubly TBS-protected to give **193**, which was transformed into the corresponding acid chloride **194** by treatment with oxalyl chloride in DMF. After formation of *Weinreb* amide **195**, the latter was converted into ynone **196** with MeCCMgBr. Unfortunately, reaction of **196** with ethyl 2-hydrazinylacetate failed to give any of the desired pyrazole derivate **197**.



Scheme 48: a) TBSCl, imidazole, DMF, 86%; b) $(COCl)_2$, CH_2Cl_2/DMF, 0 °C; c) (OMe)MeNH·HCl, pyridine, CH_2Cl_2, 50% over two steps; d) MeCCMgBr, THF, 0 °C, 51%; e) Ethyl 2-hydrazinylacetate, EtOH, reflux, no product formed.

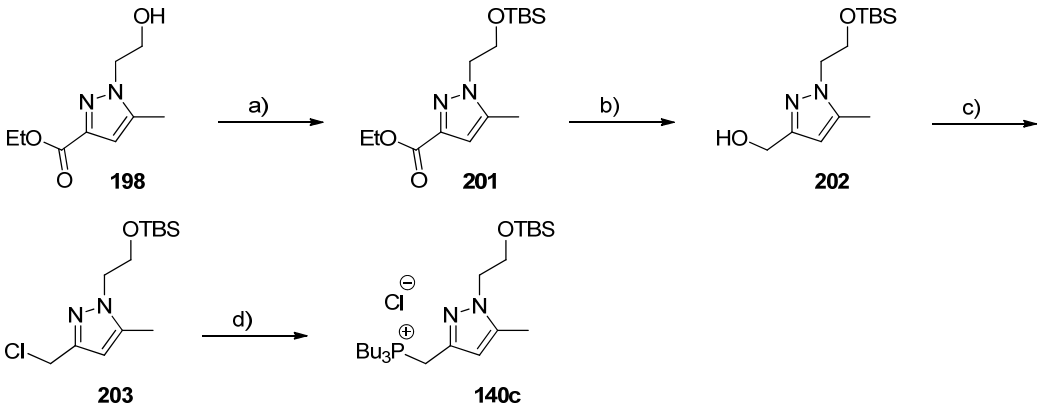
A successful approach to the required pyrazole phosphonium salt **140c** could finally be developed based on the reaction of ethyl acetopyruvate **157** with 2-hydroxyethylhydrazine, to afford pyrazole derivate **198** in 58% yield, together with its separable regioisomer **199** (28%) (Scheme 49). A strong NOE between the methyl group and the CH_2 protons attached to the

nitrogen of the side chain was observed for **198**, which established the desired regiochemistry. In contrast, no NOE between these protons was observed for the minor regioisomer **199.** This assignment was finally confirmed by an X-ray crystallographic analysis of the minor regiosiomer **199.** Remarkably, the addition of methyl hydrazine to ethyl acetopyruvate **157**, which was carried out later, led to the preferential formation of the regioisomer corresponding to **199** (Section 1.5.1.7, Scheme 36).



Scheme 49: a) 2-Hydrazinylethanol, EtOH, 60 °C, 58%.

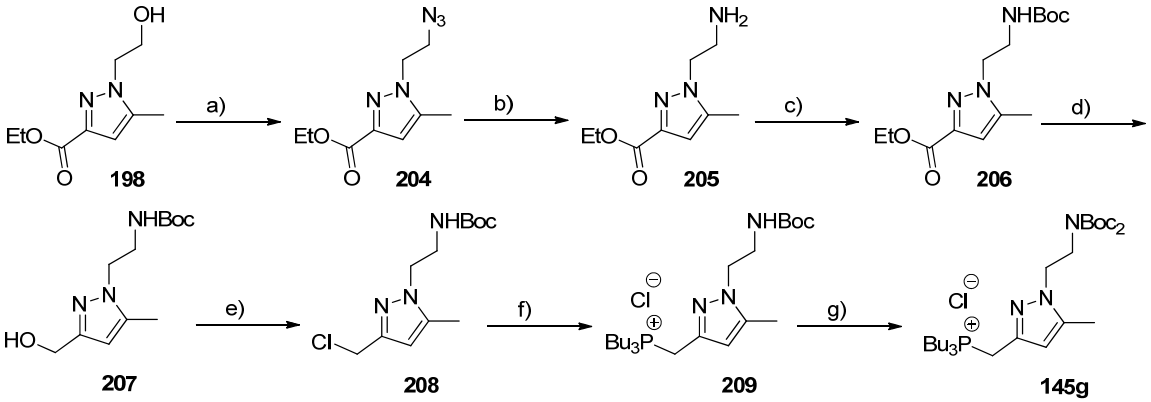
TBS-protection of the primary hydroxy group in **198** followed by reduction of the ester moiety with DIBAL-H gave primary alcohol **202** in 98% yield (Scheme 50). Mesylation and *in situ* displacement of the mesyloxy group by chloride anion then gave **203** (73%). Note that chloride formation from **202** with thionyl chloride led to concurrent deprotection of the primary hydroxy group. Finally, the phosphonium salt **140c** was obtained by treatment of chloride **203** with tributylphosphine. Attempts to furnish the corresponding phosphonate suffered from low yields.



Scheme 50: a) TBSCl, imidazole, DMF, 94%; b) DIBAL-H, CH_2Cl_2, -78 °C, 98%; c) MsCl, 2,6-lutidine, LiCl, DMF, 73%; d) PBu_3, DMF, 98%.

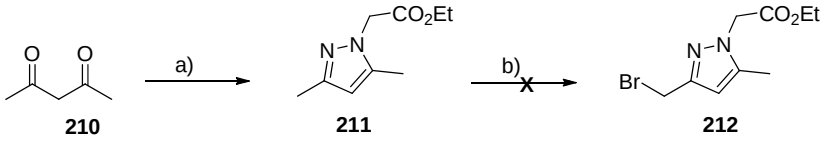
Primary alcohol **198** was also converted into azide **204** by mesylation and subsequent reaction of the mesylate with sodium azide in 96% yield (Scheme 51). Introduction of the azide group in a single step by means of DPPA was less efficient, as **204** was very difficult to purify from the reaction mixture. Reduction of **204** by catalytic hydrogenation yielded amine **205** in 88%

yield,[8] which was mono-Boc protected. Subsequent DIBAL-H reduction of the ester moiety gave primary alcohol **207**, which was transformed into chloride **208** by reaction with thionyl chloride. Treatment of **208** with tributylphosphine gave phosphonium salt **209**, which was finally converted into its bis-Boc derivative **145g** in 97% yield.



Scheme 51: a) i. MsCl, NEt_3, CH_2Cl_2; ii. NaN_3, DMF, 96%; b) H_2, Pd/C, EtOH, 88%; c) Boc_2O, NEt_3, CH_2Cl_2, 89%; d) DIBAL-H, CH_2Cl_2, -78 °C, 89%; e) $SOCl_2$, CH_2Cl_2, 75%; f) PBu_3, DMF, 40 °C, 93%; g) Boc_2O, DMAP, MeCN, 97%.

It should be noted here that attempts to functionalize preformed pyrazole derivative **211** were unsuccessful. As for isoxazole **177**, treatment of **211** with NBS under radical conditions did not produce any of the desired product **212** (Scheme 52).



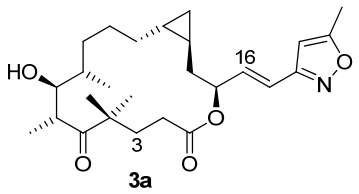
Scheme 52: a) Ethyl 2-hydrazinylacetate, EtOH, reflux, 27%; b) NBS, benzoyl peroxide, CCl_4, reflux, no product formed.

[8] Initially, amine **205** was purified by silica gel chromatography using EtOAc (85%), MeOH (10%), NEt_3 (5%) as eluents, which led to some transesterifcation. This was avoided in the following using EtOH instead of MeOH.

1.5.2 Synthesis of Hypermodified Epothilone A Analogs

Given the complex synthesis of the CP-Epo B analogs discussed in the previous sections, these compounds may be suboptimal drug cargos for ADC development. Clearly, compounds that would be synthetically more readily accessible would be more desirable for this purpose. In this context hypermodified epothilone analogs as they have been previously developed in our group may be of significant relevance, as they combine high potency with simplified structural features.[133]

An example of a hypermodified Epo A analog is isoxazole derivative **3a** (C. N. Kuzniewski, O. Horlacher, K.-H. Altmann, unpublished results) (Figure 26).



3a

Figure 26: Hypermodified Epo A analog **3a**.

Compared to natural epothilones, and in particular to Epo B as the most active natural epothilone, analog **3a** is characterized by a number of structural modifications. These include:

- The removal of the hydroxy group at position C3, which eliminates one of seven chiral centers from the core structure and thus simplifies the synthesis.
- Removal of the side chain methyl group at position C16, which results in synthetic simplification.
- The presence of a *trans*-substituted cyclopropane ring in place of the natural *cis*-epoxide moiety. The epoxide to cyclopropane exchange is expected to lead to enhanced metabolic stability. Compared to the CP-Epo B analogs discussed above, which incorporate a trisubstituted cyclopropane moiety, *trans*-disubstituted cyclopropanes are more readily accessible.

3a showed profound growth inhibition against three different tumor cell lines A549 (lung), MCF-7 (breast), and HCT116 (colon) with IC_{50} values in the sub-nM range (Table 11, collaboration with Prof. Juerg Gertsch, University of Berne, unpublished data).

Table 11: Antiproliferative activity of hypermodified CP-Epo A analog **3a** (IC_{50} values [nM]).

	A549	**MCF-7**	**HCT116**
3a	0.94±0.08	0.89±0.14	0.72±0.09

IC_{50} values of 0.33, 0.34, and 0.16 nM have been reported for Epo B against the A549, MCF-7, and HCT116 cell lines respectively.

In light of its structural and biological properties hypermodified **3a** could be a promising candidate for the construction of ADCs. The specific objective of the particular sub-project pursued in this thesis was the elaboration of an efficient and economic strategy for the synthesis of **3a** or **3b**. This work was carried out as part of the Master theses of Stefan Vetterli and Rahel Bregy.

Conceptually, the synthesis should be based on the approach previously developed in the group by C. Kuzniewski as part of his PhD thesis[134] (Section 1.5.2.1, Figure 28), but it should avoid the use of expensive catalysts (e.g. *Grubbs*' catalyst) and allow to synthesize either structure **3a** or **3b** (Figure 27) on a multigram scale. Analog **3b** is a (additionally) functionalized variant of **3a** and while the latter would have to be esterified *via* the hydroxy group at C7 for the construction of ADC's (*vide supra*), **3b** incorporates an extra hydroxy functionality, which could be used as coupling site for the attachment of an antibody.

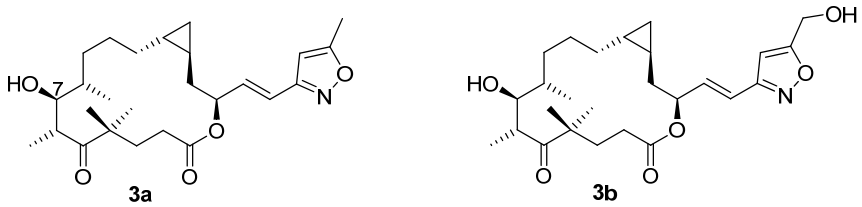


Figure 27: Target structures **3a** and **3b**.

1.5.2.1 Synthesis of Hypermodified Epothilones by C. Kuzniewski

The Kuzniewski synthesis of hypermodified Epo A analog **213**, which is closely related to the target structure addressed in this thesis, made use of a *syn*-aldol addition to install the stereocenters at C6 and C7, an esterification/RCM sequence to construct the macrocycle and a late stage introduction of the heterocycle *via Wittig* olefination (Figure 28).

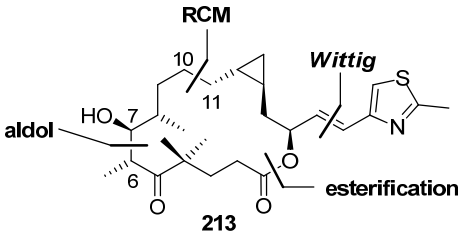
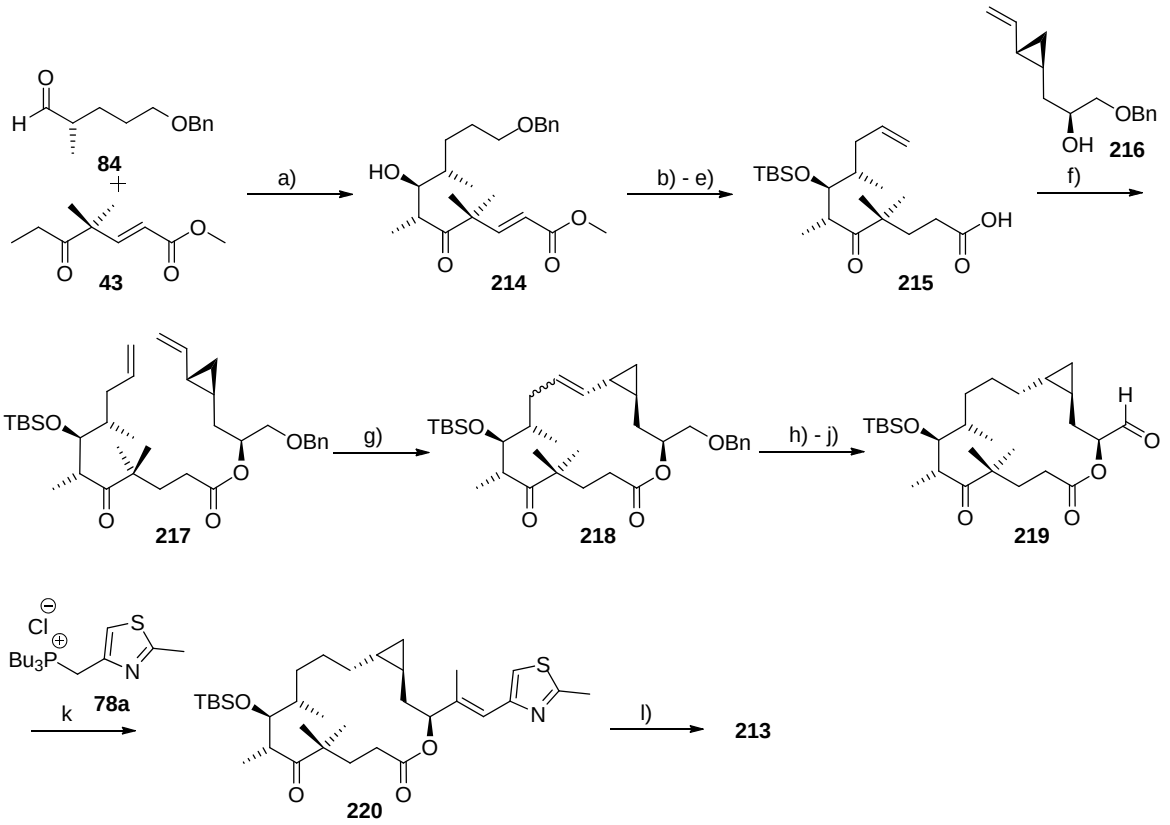


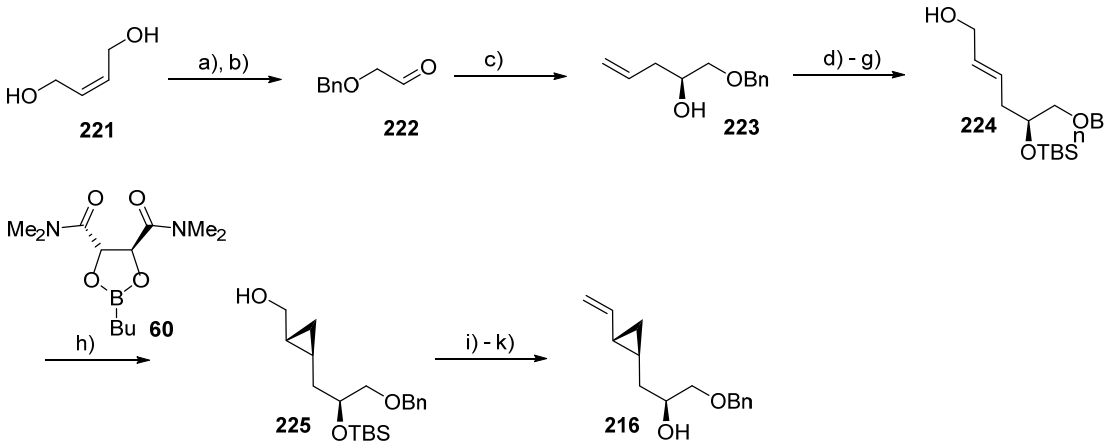
Figure 28: Structure of thiazole-based hypermodified Epo A analog **213** and key retrosynthetic disconnections.

In the actual synthesis the critical aldol reaction was performed between α-chiral aldehyde **84** (obtained in 5 steps and 15% overall yield from δ-valerolactone)[121] and the titanium enolate of keto acid **43** which gave the desired product **214** in 55% yield and a diastereomeric ratio of 5:1 (Scheme 53). TBS protection of the newly formed secondary hydroxy group, simultaneous benzyl ether cleavage and double bond reduction by catalytic hydrogenation, *Grieco-Sharpless* elimination,[120] and final ester hydrolysis provided acid **215** in 27% overall yield from aldehyde **84**. *Yamaguchi* esterification of acid **215** with alcohol **216** gave diene **217** in quantitative yield, which cyclized in the presence of *Hoveyda-Grubbs* 1st generation catalyst to form macrocycle **218** as a mixture of isomers in 68% yield (*E*/*Z* 1.3:1) (Scheme 53). Subsequent benzyl ether cleavage with $BCl_3 \cdot SMe_2$ (65% yield), reduction of the double bond with $TrisNHNH_2$, and oxidation of the primary hydroxy group gave aldehyde **219**. The latter underwent *Wittig* olefination to afford the desired coupling product **220** in 53% yield as a single isomer. Final deprotection was achieved with HF·pyridine to yield hypermodified Epo A analog **213** in 73% yield.



Scheme 53: a) $TiCl_4$, *i*-Pr_2NEt, CH_2Cl_2, -78 °C to 0 °C, 55%, dr 5:1; b) TBSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C to rt, 92%; c) H_2, Pd/C, MeOH, 97%; d) *o*-$NO_2PhSeCN$, PBu_3, $NaHCO_3$, H_2O_2, THF, 30 °C to 40 °C, 73%; e) LiOH, *i*-PrOH/H_2O, 60 °C, quant.; f) 2,4,6-Trichlorobenzoyl chloride, NEt_3, benzene, **216**, DMAP, quant; g) *Hoveyda-Grubbs* I (5 mol %), toluene, 60 °C, 68%, *E*/*Z* 1.3:1); h) $BCl_3 \cdot SMe_2$, CH_2Cl_2, 65%; i) $TrisNHNH_2$, NEt_3, DCE, 50 °C, 76%; j) TPAP, NMO, CH_2Cl_2; k) K-*t*OBu, THF, 0 °C; then **219**, -78 °C to 0 °C, 53%; l) $HF \cdot Py$, 77%.

As shown in Scheme 54 the key steps in the synthesis of alcohol **216** were a stereoselective *Brown* allylation[135] of aldehyde **222**, to form allylic alcohol **223** with 80% yield and good enantioselectivity, and the *Charette* cyclopropanation of allylic alcohol **224**. The latter transformation gave the desired cyclopropane derivate **225** in 61% yield and a diastereomeric ratio of 10:3. Building block **216** was obtained in 11 steps and an overall yield of 6% based on 1,4-butendiol (**221**).



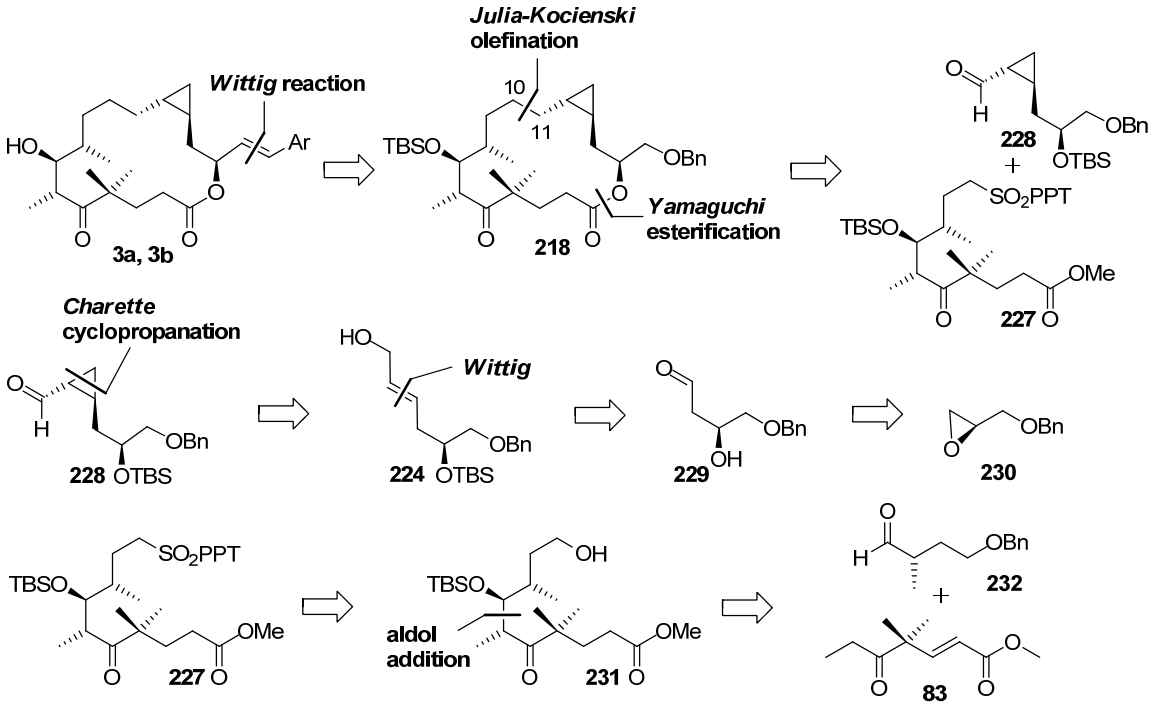
Scheme 54: a) NaH, BnBr, DMF, 0 °C to rt, 82%; b) O_3, MeOH/CH_2Cl_2, -78 °C, then SMe_2, -78 °C to rt, 77%; c) (-)-$(Ipc)_2$B-allyl, Et_2O, -100 °C, then ethanolamine, rt, 80%; d) TBSCl, imidazole, DMF, 0 °C to rt, 91%; e) O_3, MeOH, -78 °C,; then SMe_2, -78 °C to rt, 73%; f) $Ph_3PCHCOOEt$, CH_2Cl_2, 0 °C to rt, 94%; g) DIBAL-H, CH_2Cl_2, -78 °C, 84%; h) $ZnEt_2$, CH_2I_2, DME, CH_2Cl_2, -10 °C to rt, 61%; i) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2 -78 °C, to 0 °C, 57%; j) $MePPh_3Br$, *n*-BuLi, THF, -10 °C to rt, 95%; k) CSA, MeOH/CH_2Cl_2, 0 °C to rt, 91%.

The described synthesis is certainly stable and not inefficient, nevertheless, it suffers from a few drawbacks. Several reaction yields are only moderate as for example the aldol addition (55%), the macrocyclization by means of RCM (68%) or the cleavage of the benzyl ether with $BCl_3{\cdot}SMe_2$ (65%). In addition, in the synthesis of the alcohol **216** (Scheme 54) the *Charette* cyclopropanation (61%, dr 10:1) as well as the subsequent *Swern* oxidation (57%) of the primary hydroxy group in cyclopropane derivate **225** are rather low yielding. Furthermore, the synthesis makes use of toxic reagents like the *o*-$NO_2PhSeCN$ used for the elimination or rather expensive catalysts as the *Hoveyda-Grubbs* 1st generation catalyst necessary to form the macrocycle **218**.

In an attempt to optimize the Kuzniewski synthesis the named drawbacks should be avoided and an efficient synthesis, which ignores the use of toxic or expensive reagents was to be established.

1.5.2.2 New Retrosynthesis of 3a and 3b

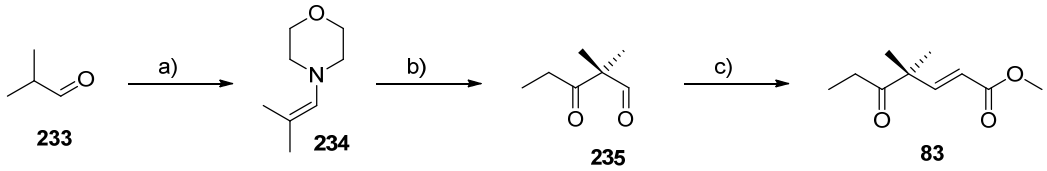
In contrast to the synthesis developed by C. Kuzniewski for analogs of type **3a** and **3b**, the (alternative) strategy envisioned for the Master theses of Stefan Vetterli and Rahel Bregy (Scheme 55) would be based on ring-closure by macrolactonization rather than by RCM to construct the 16-membered macrocycle. A second key step in our approach was a *Julia-Kocienski* olefination,[136] which would combine sulfone **227** with aldehyde **228** to establish the C10/C11 bond and thus the entire carbon framework of the macrocyclic core structure. Sulfone **227** would be accessible *via* a *Mitsunobu* reaction[137] of **231** with 1-phenyl-1*H*-tetrazole-5-thiol and subsequent oxidation of the sulfide to the sulfone. As for the Kuzniewski synthesis, the C6-C7 bond would be formed by a *syn* aldol reaction between ethyl ketone **83** and α-chiral aldehyde **232** and the cyclopropane ring would be obtained by a stereoselective *Charette* cyclopropanation of allylic alcohol **224**. Finally, aldehyde **229** was to be obtained *via* hydrolysis of the corresponding thioacetal, which was readily accessible from commercially available (*R*)-(+)-glycidol (**230**).



Scheme 55: Retrosynthesis of target structures **3a**, **3b**.

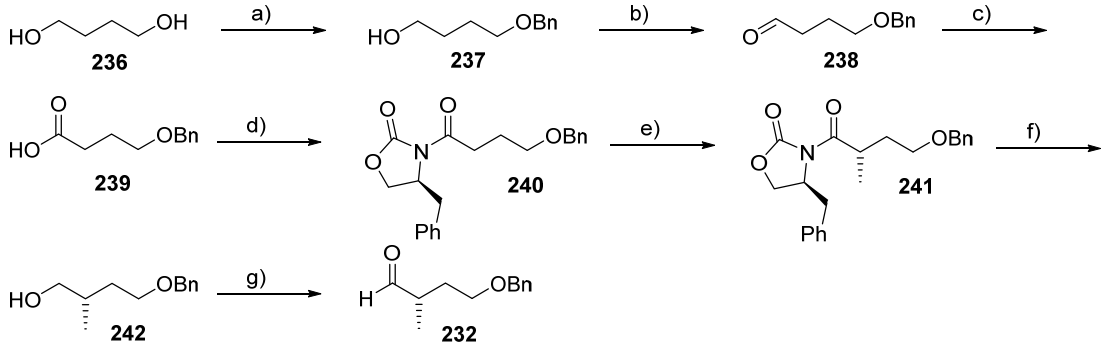
1.5.2.3 Forward Synthesis

The known ethyl ketone **83** was synthesized from isobutyraldehyde (**233**) in a three-step sequence in 28% overall yield (Scheme 56).[138]



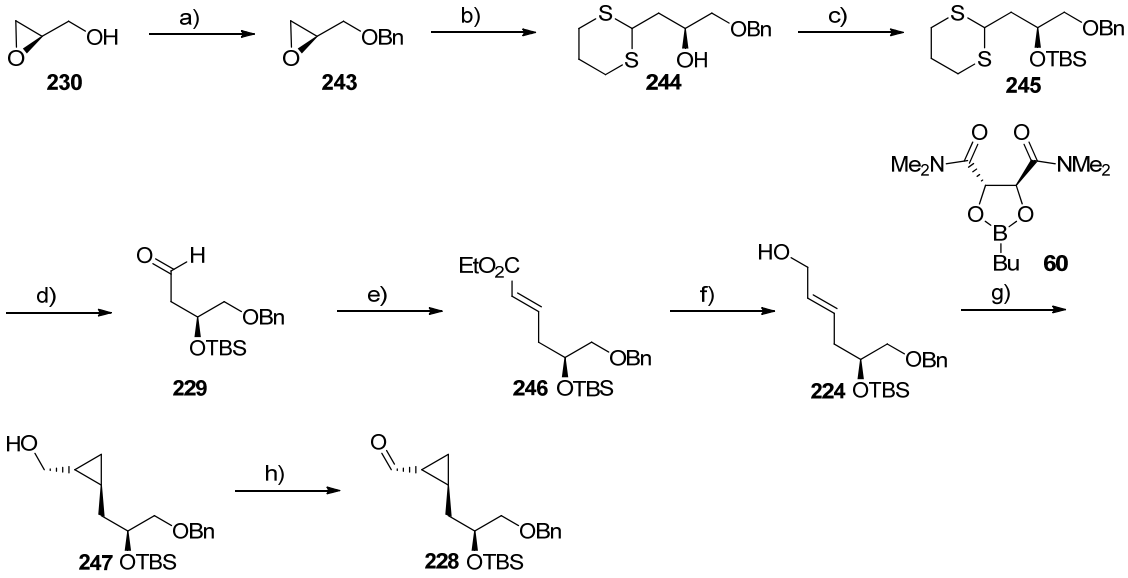
Scheme 56: a) Morpholine, *p*-TsOH, CH_2Cl_2, 0 °C to reflux, 89%; b) Propionyl chloride, MTBE, rt to reflux, 35%; c) Trimethyl phosphonoacetate, *n*-BuLi, THF, -78 °C to rt, 90%. *E*/*Z* 20:1.

α-Chiral aldehyde **232** was synthesized starting from 1,4-butanediol (**236**), which was mono-benzylated with benzyl bromide (Scheme 57). The free hydroxy group in **237** was then oxidized sequentially to acid **239** by *Swern* oxidation and subsequent *Pinnick* oxidation[139] of the ensuing aldehyde.[140] Alternatively, **239** was accessed by ring-opening of γ-butyrolactone with KOH and protection of the resulting hydroxy group by treatment with benzyl bromide in 98% yield. This route is two steps shorter than the synthesis from 1,4-butanediol (**236**), but it suffers from the drawback of a five day synthesis procedure.[141] Acid **239** was then converted into imide **240** by activation as a mixed anhydride and reaction with the (*S*)-4-Benzyl-2-oxazolidinone in the presence of *n*-BuLi.[142] Deprotonation of imide **240** with NaHMDS at -78 °C and reaction with MeI gave **241** with a dr of 14:1. Reductive cleavage of the *Evans*-auxiliary with $LiBH_4$ followed by *Swern* oxidation then yielded the desired α-chiral aldehyde **232** (Scheme 57). Aldehyde **232** was found to be highly unstable and to decompose within a few hours; it was thus used rapidly and without purification for the following aldol reaction.



Scheme 57: a) NaH, BnBr, TBAI, THF, 98%; b) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to rt, 94%; c) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH/H_2O, 78%; d) i. EtOCOCl, NEt_3, Et_2O, 0 °C. ii. *n*-BuLi, (*S*)-4-Benzyl-2-oxazolidinone, THF, -78 °C, 93%; e) NaHMDS, MeI, THF, -78 °C to 0 °C, dr 14:1, 88%; f) $LiBH_4$, MeOH, THF, 0 °C, 80%; g) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to rt, 90%.

The synthesis of cyclopropane-containing aldehyde **228** started form commercially available (*R*)-(+)-glycidol (**230**), which was benzylated with benzyl bromide to afford **243** in 88% yield (Scheme 58). Regioselective opening of the epoxide with lithiated 1,3-dithiane followed by TBS protection of the resulting secondary alcohol gave TBS-ether **245** in 84% yield over two steps. Cleavage of the thioacetal moiety was achieved by treatment of **245** with $Hg(ClO_4)_2 \cdot 3H_2O$ in 92% yield, while NBS and $HgCl_2$ remained ineffective.[143] The resulting aldehyde **229** underwent *Wittig* olefination with Ph_3PCHCO_2Et to afford *E* olefin **246** with 19:1 selectivity over the corresponding *Z* isomer. In contrast, *HWE* olefination with trimethylphosphono acetate gave about the same yield, but was less selective (*E*/*Z* 5:1). Subsequent DIBAL-H reduction of the ester moiety in **246** gave allylic alcohol **224** in 90% yield, which was subjected to *Charette* cyclopropanation. The desired cyclopropane derivate **247** formed with an excellent yield of 98% and selectivity (dr 20:1) (Scheme 58).

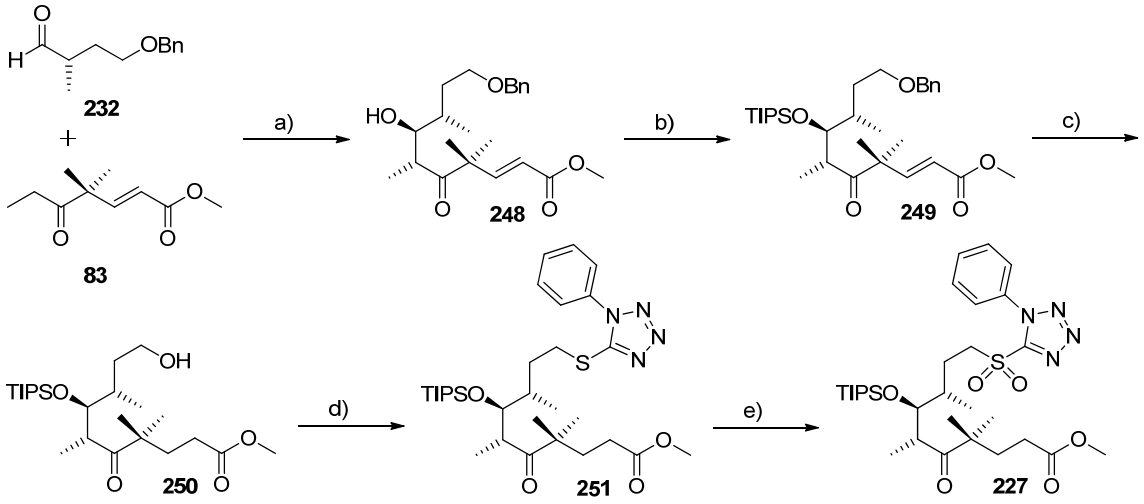


Scheme 58: a) NaH, BnBr, DMF, 0 °C to rt, 88%; b) *n*-BuLi, 1,3-Dithiane, THF, -78 °C to 0 °C, 87%; c) TBSCl, imidazole, DMF, 96%; d) $Hg(ClO_4)_2$, $CaCO_3$, THF/H_2O, 92%; e) Ph_3PCHCO_2Et, CH_2Cl_2, 0 °C to rt, 94%, *E*/*Z* 19:1; f) DIBAL-H, CH_2Cl_2, -78 °C, 90%; g) $ZnEt_2$, CH_2I_2, **224**, 0 °C, 98%, dr 20:1; h) DMP, CH_2Cl_2, 97%.

The final oxidation to aldehyde **228** was best achieved with DMP, while other oxidation procedures, such as *Swern* oxidation, or the use of PDC[144] or Tempo[145] gave **228** in lower yield (50-65%). This is in line with the fact that Kuzniewski reported the oxidation of **247** under *Swern* conditions to deliver aldehyde **228** in a yield of 56%. Important improvements could also be realized in the *Charette*-cyclopropanation step. By running the reaction at a constant temperature of 0 °C, the yield could be improved from 61% to 98% and the diastereoselectivity was enhanced from 10:3 to 20:1.[133] In conclusion, a highly efficient

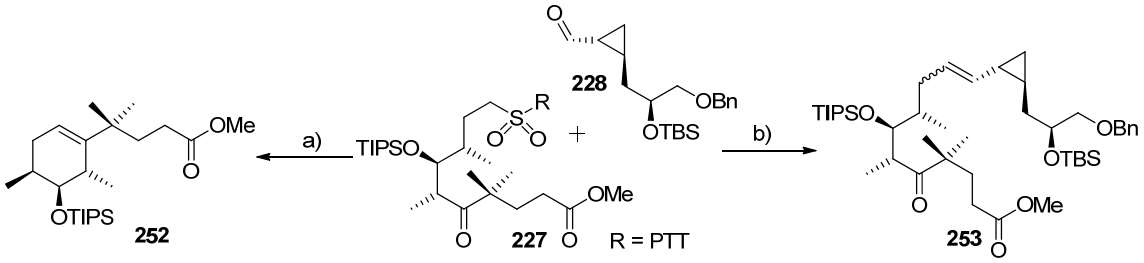
route to cyclopropane-containing aldehyde **228** was established, which gave the desired building block with an overall yield of 54% over 8 steps from (*R*)-(+)-glycidol (**230**). Several grams of **228** were synthesized.

With key building blocks **83**, **228** and **232** in hand the focus was put on the assembly of these fragments. Thus, ethyl ketone **83** was treated with $TiCl_4$ and DIPEA to form preferentially the *Z*-enolate, which was then reacted with the α-chiral aldehyde **232** to give the *syn* aldol product **248** in a diastereomeric ratio of 3.7:1 and a yield of 56% (Scheme 59). Other conditions explored for enolate formation, such as LDA, $Sn(OTf)_2/NEt_3$ and *n*-Bu_2BOTf/DIPEA proved to be inferior to the $TiCl_4$/DIPEA approach, both with regard to yield and selectivity. As mentioned earlier, aldehyde **232** is unstable as well as volatile and, hence, is difficult to purify. This might be part of the reason for the moderate yield obtained for the aldol addition. **248** was then protected as TIPS-ether **249** by reaction with TIPSOTf in 94% yield (TIPSCl proved to be unreactive). A side product was isolated in this reaction in small amounts (< 5%), originating from the intramolecular addition of the C7-hydroxy group to the α,β-unsaturated ester moiety in **248** to form a 6-membered ring. Palladium catalyzed hydrogenation served to remove the benzyl group and to reduce the C2/C3 double bond in the same step. The resulting primary alcohol **250** was then transformed into the sulfide **251** by means of a *Mitsunobu* reaction[146] with 1-phenyl-1*H*-tetrazole-5-thiol in 96% yield. Finally, *m*-CPBA oxidation of the sulfide resulted in the precursor **227** for the critical *Julia-Kocienski* olefination (94%). Apart from the aldol reaction, which only worked in a moderate yield of 56%, all other steps leading to sulfone **227** were high-yielding. Consequently, this fragment could also be synthesized on a multigram scale.



Scheme 59: a) $TiCl_4$, DIPEA, CH_2Cl_2, -78 °C, 56%, dr 3.7:1; b) TIPSOTf, 2,6-lutidine, CH_2Cl_2, -78 °C to rt, 94%; c) Pd/C, H_2, MeOH, 88%; d) 1-Phenyl-1*H*-tetrazole-5-thiol, PPh_3, DEAD, THF, 0 °C, 96%; e) *m*-CPBA, CH_2Cl_2, 94%.

The stage was thus set to explore the pivotal *Julia-Kocienski* olefination between sulfone **227** and aldehyde **228**. It was observed that the sulfone **227**, if deprotonated before the addition of aldehyde **228**, underwent an intramolecular cyclization reaction to form **252** (Scheme 60). Therefore, sulfone **227** was deprotonated in the presence of aldehyde **228** (*Barbier* conditions)[136] which completely surpressed the undesired cyclization reaction and afforded olefin **253** in an excellent 90% yield. The moderate selectivity of the reaction with an *E*/*Z* ratio of 3:1 was inconsequential, as the double bond was to be reduced in the next step.



Scheme 60: a) LiHMDS, THF, -78 °C, then **228**, 46%; b) LiHMDS, THF, -78°C, 90%, *E*/*Z* 3:1.

The reduction of the double bond in **253** proved to be troublesome (Table 12). *Wilkinson*'s catalyst and hydrazine/H_2O_2/Cu(II) gave no conversion of starting material, while catalytic hydrogenations over Pd/C or *Lindlar* catalyst suffered from concomitant reductive opening of the cyclopropane moiety.[9] To our delight, the double bond could be reduced in a fully selective way by making use of the diimide source $TrisNHNH_2$ in an excellent yield of 95%.

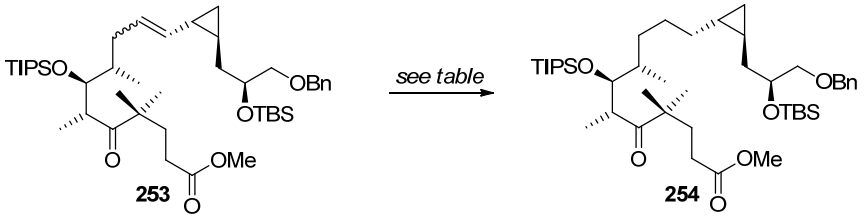
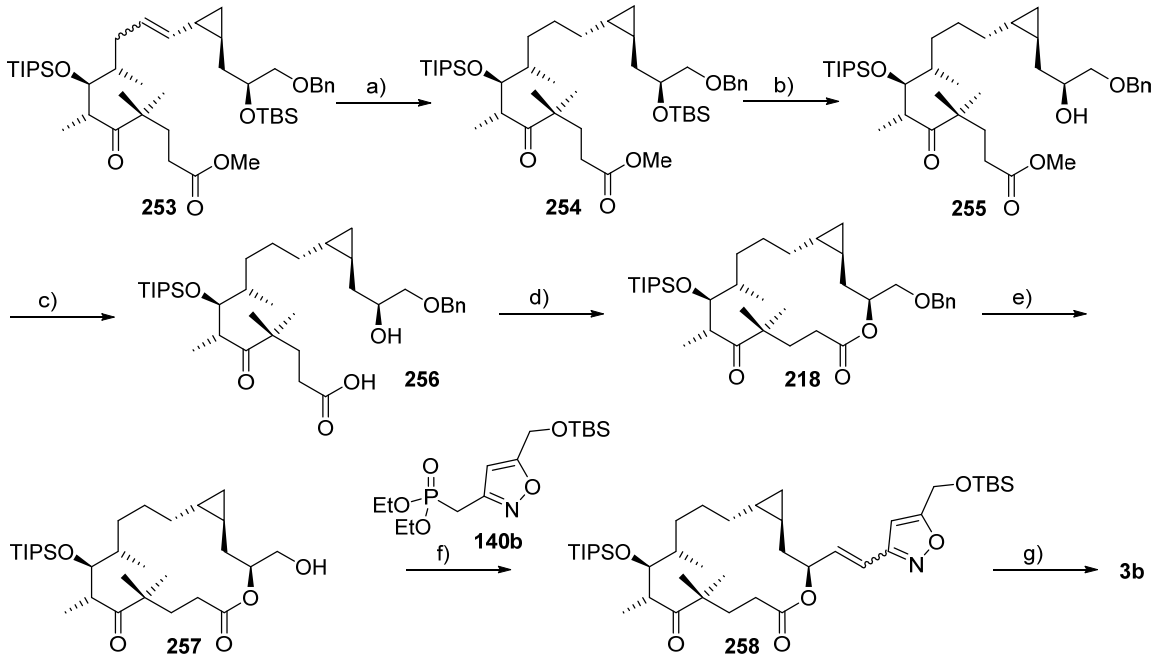


Table 12: Reduction of double bond in **253**.

Entry	Reagent		Solvent	Reaction
1	*Wilkinson* cat. (1.2 eq.)	6.5 bar H_2	THF	no conversion
2	Hydrazine/H_2O_2, Cu(II)		MeOH	no conversion
3	Pd/C	6.5 bar H_2	MeOH	15% product, 35% cyclopropane opened based on LRMS
4	*Lindlar* cat. (0.5 eq.)	6.5 bar H_2	EtOH	28% product, 18% cyclopropane opened based on LRMS
5	$TrisNHNH_2$	NEt_3	DME	95% yield

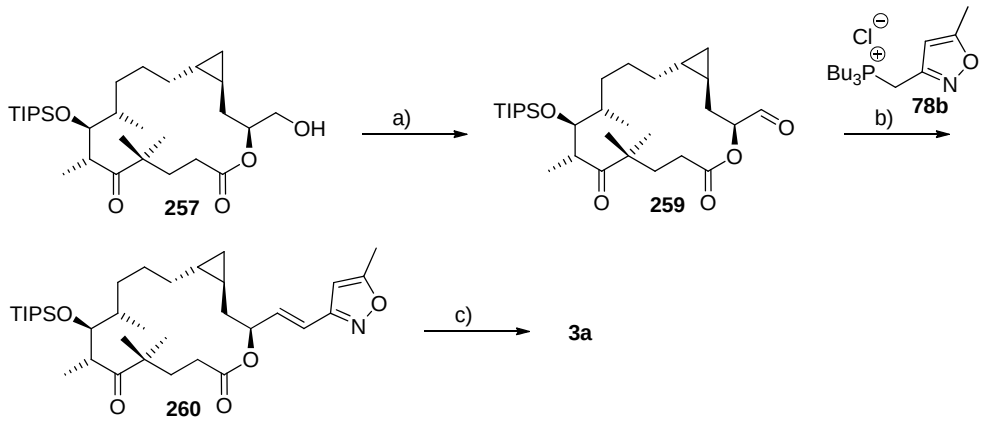
[9] Analysis of the reaction mixture by low resolution mass spectrometry showed the presence of a mass +2 relative to the mass of the desired product **254**. Based on the experiences gained in the synthesis of CP-Epo B (**1a**) the side product is believed to be formed *via* opening of the cyclopropane.

The TBS group in **254** was then selectively removed with CSA to yield secondary alcohol **255** in 95% yield (Scheme 61). Initial attempts to saponify the ester moiety in **255** with $LiOH^{\bullet}H_2O$ in THF/H_2O gave no conversion, while the use of isopropanol/H_2O as the solvent mixture led to the formation of a transesterification side product. It was reasoned that the use of a sterically more hindered alcohol like *t*-BuOH would not induce any transesterification. This assumption proved to be valid and saponification of **255** with $LiOH{\cdot}H_2O$ in *t*-BuOH/H_2O produced carboxylic acid **256** in quantitative yield. Macrolactonization was then achieved by *Yamaguchi* esterification,[78] which gave macrolactone **218** in 75% yield. Subsequent removal of the benzyl protecting group with $BCl_3{\bullet}SMe_2$ (the conditions that had worked best in Kuzniewski's synthesis) turned out to be problematic, due to translactonization of the desired product **257**, which formed the undesired macrolactone in up to 30% yield. As this side reaction could not be avoided, different conditions for benzyl ether cleavage were explored. While DDQ provided the desired product only in very moderate yield (30-35%), $FeCl_3{\bullet}6H_2O$ only led to cleavage of the TIPS-ether and $AlCl_3$/*N*,*N*-dimethylaniline remained ineffective. Ultimately, cleavage of the benzyl ether was still best performed with $BCl_3{\bullet}SMe_2$, which under optimized conditions delivered the free alcohol **257** in 48% yield. Oxidation of **257** with TPAP/NMO[122] gave the corresponding aldehyde, which was immediately reacted with phosphonate **140b** to give the protected hypermodified epothilone **258**, albeit only in a moderate yield of 20% over two steps and as an inseparable 1:1 mixture of *E*/*Z* isomers (Scheme 61). The yield could be increased to 40% if the primary alcohol **257** was oxidized by means of DMP.[117] The use of LiHMDS to deprotonate phosphonate **140b** gave none of the desired coupling product (no conversion). Final deprotection was carried out with HF·pyridine to yield target structure **3b** in 77% yield as 1:1 mixture of *E*/*Z* isomers that was inseparable by silica gel chromatography. Separation of the isomers was finally achieved by means of preparative HPLC.



Scheme 61: a) 2,4,6-Trisisopropylbenzenesulfonohydrazide, NEt_3, DME, 50 °C, 95%; b) CSA, CH_2Cl_2/MeOH, 95%; c) $LiOH \cdot H_2O$, *t*-BuOH/H_2O, quant.; d) 2,4,6-Trichlorobenzoyl chloride, NEt_3, DMAP, THF, 75%; e) $BCl_3 \cdot SMe_2$, CH_2Cl_2, 48%; f) i. DMP, CH_2Cl_2; ii. *t*-BuOK, **140b**, , THF, 40% over two steps, *E*/*Z* 1:1; g) HF·pyridine, THF, 77%, *E*/*Z* 1:1.

Target structure **3a** was synthesized in the same way. Gratifyingly, the *Wittig* reaction between aldehyde **259** and the phosphorane derived from phosphonium salt **78b** gave the desired hypermodified epothilone analog **3a** in 91% yield over two steps as a single isomer (Scheme 62). Final removal of the TIPS group was achieved with $FeCl_3 \cdot 6H_2O$ to afford **3a** in 74% yield or with HF·pyridine, which gave **3a** in 68% yield.



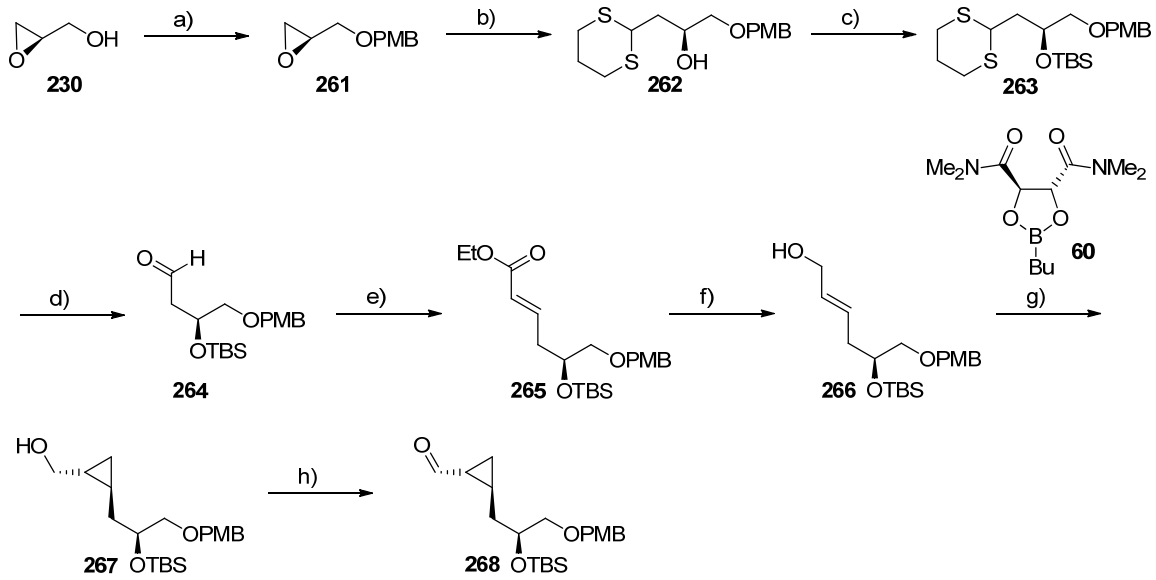
Scheme 62: a) DMP, CH_2Cl_2; b) *t*-BuOK, **259**, THF, 91% over two steps, single isomer; c) $FeCl_3 \cdot 6H_2O \cdot CH_2Cl_2$, 74%.

While the syntheses described above provide reasonably efficient access to the targeted hypermodified Epo A analogs and are clearly superior to Kuzniewski's approach, they still

suffer from the serious shortcoming that half of the valuable macrocycle **218** is lost in the course of the debenzylation reaction. As the efficiency of the deprotection was not amenable to any improvement, the use of a PMB group was investigated as an alternative to a plain benzyl group; the former should be susceptible to oxidative cleavage with DDQ. This idea was encouraged by the fact that benzyl ether cleavage in **218** using a large excess of DDQ (10 eq.), although only moderately efficient, only produced very small amounts of the translactonization side product.

1.5.2.4 Attempts at Further Synthetic Improvements

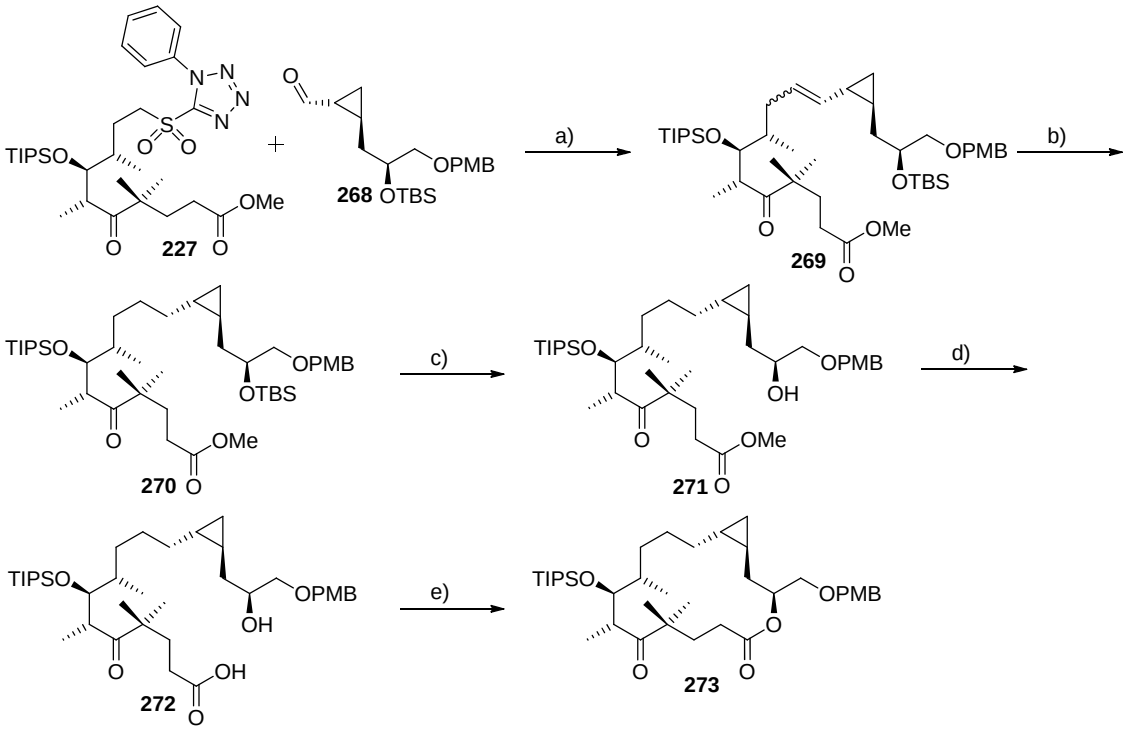
In order to explore the usefulness of a PMB group for the protection of the hydroxymethyl group on C15 of the macrocycle, aldehyde **268** was required. The latter could be prepared from PMB-protected *R*-glycidol in analogy to the synthesis of aldehyde **228** in 52% overall yield (8 steps; Scheme 63).



Scheme 63: a) NaH, PMBCl, DMF, 89%; b) *n*-BuLi, 1,3-Dithiane, THF, -78 °C to 0 °C, 84%; c) TBSCl, imidazole, DMF, quant.; d) $Hg(ClO_4)_2$, $CaCO_3$, THF/H_2O, 90%; e) Ph_3PCHCO_2Et, CH_2Cl_2, 0 °C to rt, 86%, sinlge isomer; f) DIBAL-H, CH_2Cl_2, -78 °C, 94%; g) $ZnEt_2$, CH_2I_2, **266**, 0 °C, 99%, dr 15:1; h) DMP, CH_2Cl_2, 94%.

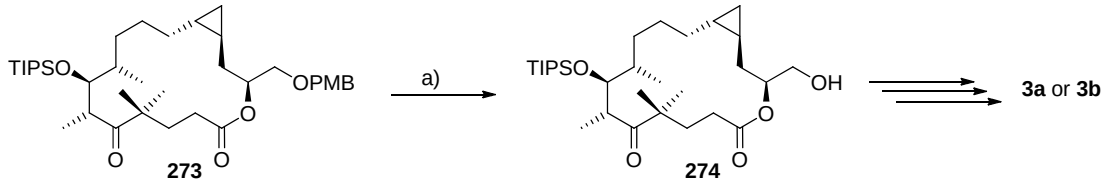
Pleasingly, the subsequent crucial coupling of the aldehyde **268** and sulfone **227** provided olefin **269** in an excellent yield of 92% (Scheme 64). All subsequent steps could be performed with similar efficiencies as for the corresponding benzyl-protected intermediates up to the stage of seco acid **272**. Surprisingly, macrolactonization of **272** under *Yamaguchi* conditions

gave only moderate yields of 40-45% in comparison to 75% for the benzyl protected seco acid **256**. In the context of optimizing the established prior synthesis of **3a** and **3b**, this yield was not deemed acceptable and alternatives were evaluated. Very much to our delight, the use of the *Shiina* macrolactonization protocol[147] yielded the macrocycle **273** in 97% yield.



Scheme 64: a) LiHMDS, THF, -78°C, 92%, *E*/*Z* 4.5:1; b) TrisNHNH$_2$, NEt$_3$, DME, 50 °C, 95%; c) CSA, CH_2Cl_2/MeOH, 90%; d) LiOH·H_2O, *t*-BuOH/H_2O, quant.; e) MNBA, DMAP, CH_2Cl_2, 97%.

The final question to be answered was whether the removal of the PMB group from **273** would indeed be more efficient, i. e. higher-yielding than the debenzylation of **218**. Very much to our satisfaction, treatment of PMB-ether **273** with DDQ allowed the isolation of the desired primary alcohol **274** in 84% yield (Scheme 65).



Scheme 65: a) DDQ, CH_2Cl_2/phosphate buffer pH 7, 84%.

In conclusion, a highly efficient synthesis of hypermodified epothilone analogs such as **3a** and **3b** has been established. This approach is clearly superior to the approach previously elaborated for these types of analogs by Kuzniewski and it has allowed the synthesis of several grams of the macrocycle **273**.

1.5.2.5 Antibody-Drug Conjugates

As discussed in section 1.5.1.6, there was some indication that the C7-hydroxy group was sufficiently reactive to serve as an attachment point for a linker to a tumor-targeting antibody (see text to Scheme 34). For the construction of **3a**-derived ADCs a self-immolative disulfide linker was selected that has been extensively studied by the *Ojima* group, who found this linker to be stable in the bloodstream and to be efficiently cleaved by glutathione (Figure 29).[148]

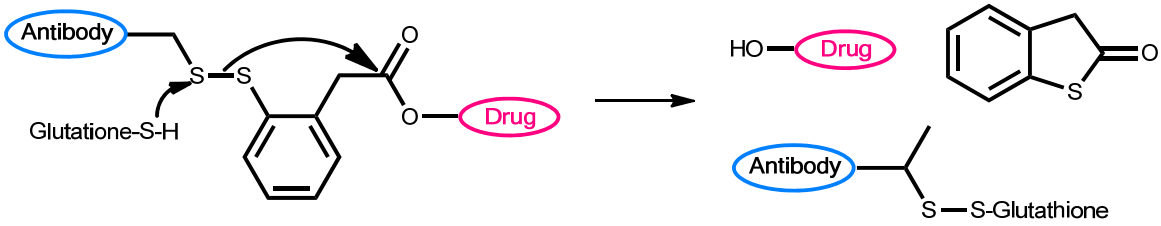
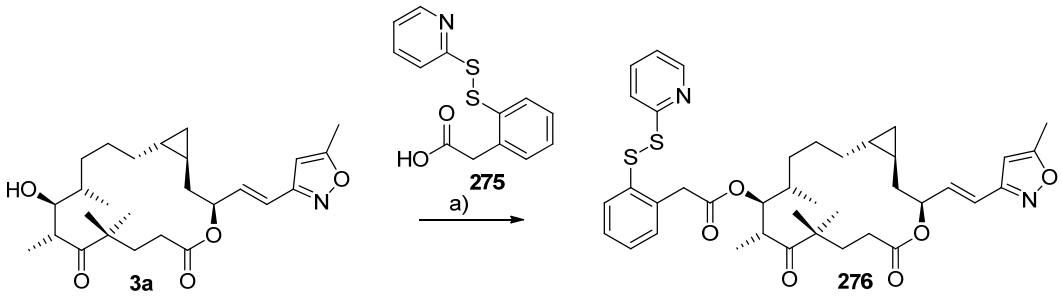


Figure 29: Self-immolative disulfide linker according to Ojima *et al.*[148]

The esterification of **3a** with acid **275** was first attempted using the DCC/DMAP protocol,[43] which gave ester **276** in a moderate yield of 16% due to low conversion. The use of EDCI as a coupling agent turned out to be even less efficient, providing ester **276** in only 10% yield, while the *Yamaguchi* protocol did not lead to any product formation. In all cases the unconsumed starting material **3a** could be reisolated.



Scheme 66: a) DCC, DMAP, CH_2Cl_2, 16%.

Despite the low yield the esterification reaction, sufficient amounts of ester **276** could be obtained for initial coupling experiments with an antibody. These experiments were carried out by Martina Steiner in the group of Prof. *Neri*. Conjugation of **276** to the SIP-F8 antibody was attempted *via* disulfide exchange,[130] but, unfortunately ester **276** showed such low water solubility that no conjugation was observed in DMF/H_2O or DMSO/H_2O 9:1 solvent mixtures.

As an alternative to disulfide exchange, *Neri* and *co*-workers have also developed a novel conjugation protocol that involves the coupling of aldehydes to antibodies without the need of a chemical linker (Figure 30).[149] The aldehyde drug undergoes thiazolidine formation by reacting with the 1,2-aminothiol functionality introduced in the antibody by engineering of the N terminus.[150] Upon thiazolidine cleavage the free drug and native antibody are restored, which limits immunogenic reactions, which may be associated with the presence of residual linker moieties on the antibody.[151]

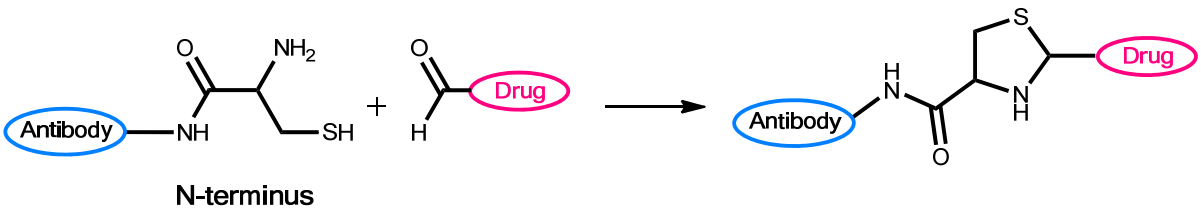
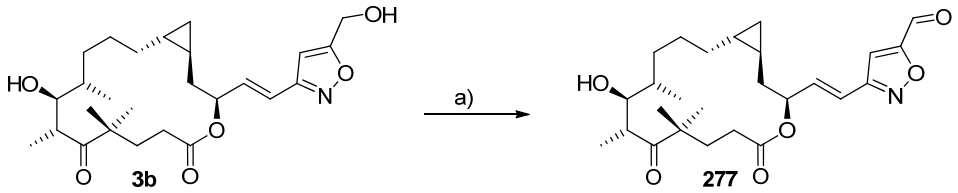


Figure 30: Linkage of the drug to the antibody *via* thiazolidine formation.

It was thus attempted to couple aldehyde **277** through thiazolidine formation; **277** was accessible through oxidation of alcohol **3b** and was anticipated to be more water soluble than **276** as the compound features two free hydroxy groups (as opposed to only one in **276**). (Scheme 67).



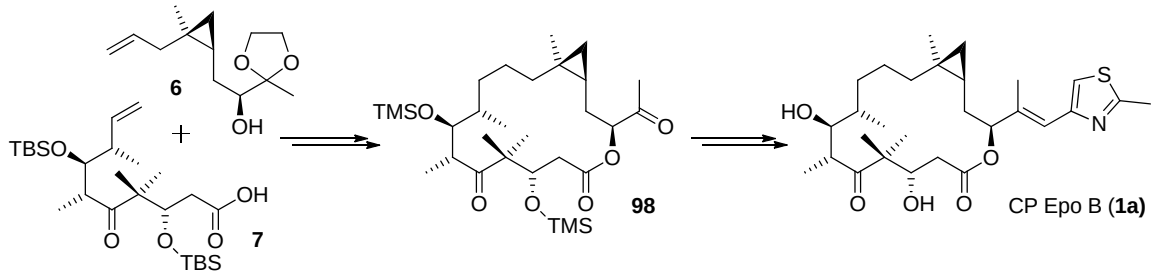
Scheme 67: a) Tempo, BAIB, CH_2Cl_2, 0 °C to rt, 71%.

Unfortunately, even aldehyde **277** was not sufficiently soluble in the required solvent mixtures for a conjugation reaction to proceed.

This is the status of this project at the time of writing of this thesis. Possible ways to overcome the solubility problem will be briefly discussed in the *Conclusion and Outlook* section.

1.6 Conclusion and Outlook

As a prelude for the construction of epothilone-based ADC's the first total synthesis of CP-Epo B (**1a**) has been developed, based on efficient RCM-mediated macrocyclization and the late stage elaboration of the thiazole-bearing side chain. While the acid building block **7** could be obtained according to literature procedures, alcohol **6** was synthesized from commercially available (*S*)-(-)-α-hydroxy-γ-butyrolactone in 12 steps and 25% overall yield (Scheme 68). **1a** was ultimately obtained in 19 steps for the longest linear sequence in 8% overall yield.



Scheme 68: Synthesis of CP-Epo B (**1a**).

Methyl ketone **98** and its 9,10-dehydro analog **148** served as common advanced precursors for the synthesis of a variety of side chain-modified CP-Epo B analogs, including (additionally) functionalized derivatives **2a-h**.

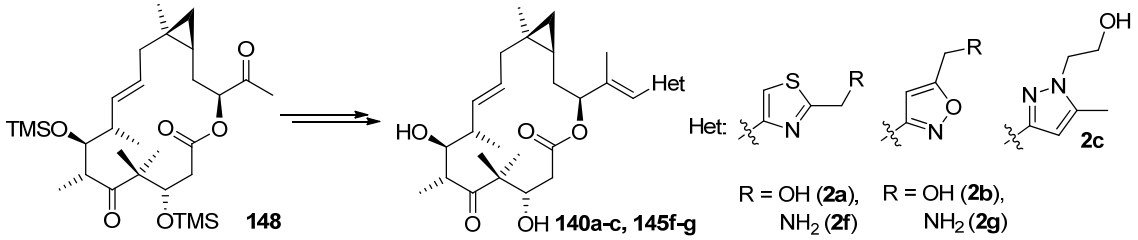


Figure 31: Selection of CP side chain-modified Epo B analogs.

All compounds shown in Figure 31 are nM or sub-nM inhibitors of cancer cell proliferation, which makes them interesting candidates for the construction of epothilone-based ADCs. In particular, hydroxymethyl isoxazole CP-Epo B (**2A**) (Figure 32) was found to be more potent than natural Epo B, exhibiting IC_{50} values of 0.25 nM and 0.36 nM against the human lung cancer cell line A549 and the human breast cancer cell line MCF-7, respectively. Thus, **2A** was chosen for the construction of an ADC. A linker bearing a maleimido functionality was attached to **2A** *via* esterification of the primary hydroxy group and the thiols of the cysteine side chains of the SIP-F8 antibody underwent 1,4-addition to form the ADC (Figure 32).

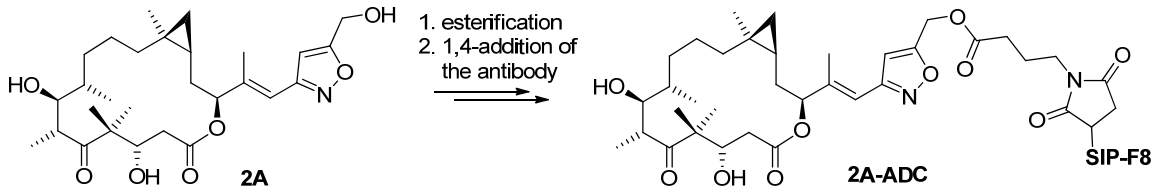


Figure 32: Antibody-drug conjugate with hydroxymethyl isoxazole CP-Epo B **2A** as the drug cargo.

Initial *in vivo* experiments with **2A-ADC** indicated a measurable antitumor effect. As the tumors were still growing, however, the dose of the **2A-ADC** was increased in a second experiment, which resulted in body weight loss of the mice. As a consequence, the experiments had to be abandoned and no further evaluation was undertaken.

In light of the rather complex synthesis of the above CP-Epo B analogs, structurally simplified epothilone analogs were investigated. Hypermodified isoxazole Epo A analog **3a** (Figure 33) had previously been identified in our group as an attractive lead structure, as it combines high potency with simplified structural features.

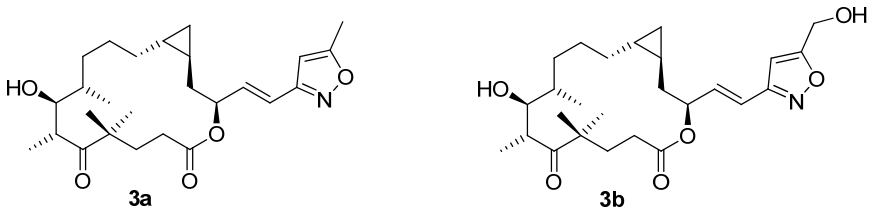
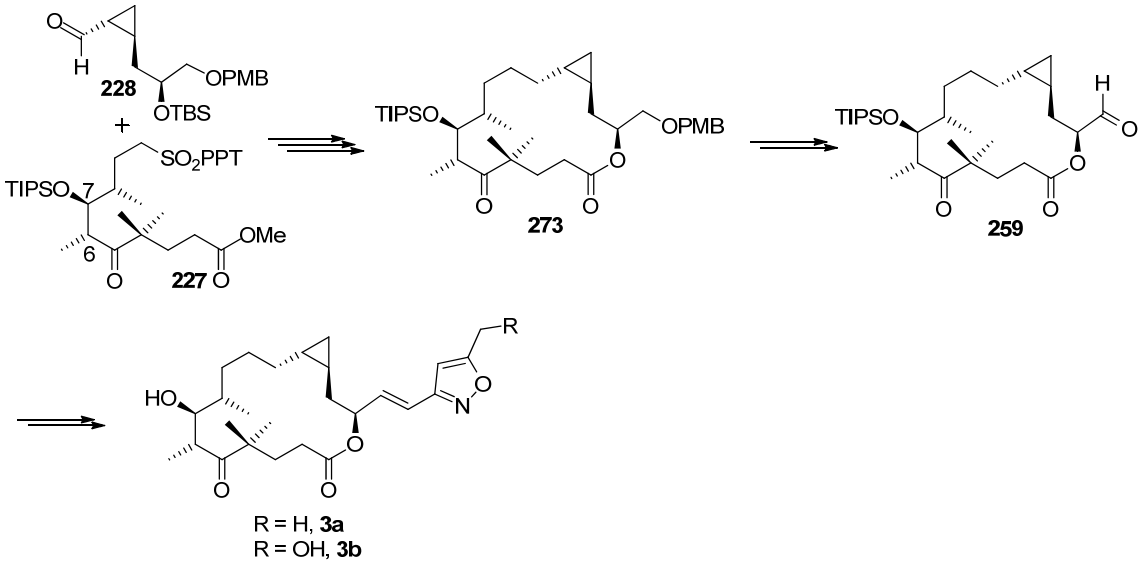


Figure 33: Hypermodified Epo A analog **3a** and **3b**.

In this thesis an efficient synthesis of **3a** and it hydroxymethyl derivative **3b** was elaborated that provides access to these compounds on a multigram scale. Key steps in the assembly of these hypermodified epothilones were (1) a *Julia-Kocienski* olefination between sulfone **227** and aldehyde **228**, (2) ring-closure by *Shiina* macrolactonization to afford **273** in almost quantitative yield, and (3) *Wittig* olefination of aldehyde **259** with the isoxazole-based phosphoranes to elaborate the C15 side chain (Scheme 69).



Scheme 69: Synthesis of hypermodified Epo A analogs **3a** and **3b**.

Hypermodified Epo A analog **3a** could be esterified with acid **275** to give activated disulfide **276**. Conjugation of **276** to the SIP-F8 antibody was attempted *via* disulfide exchange, but failed because of the low water solubility of **276**. Likewise, conjugation by thiazolidine formation between engineered N-terminal 1,2-aminothiol functionality in the SIP-F8 antibody and aldehyde **277** was unsuccessful because of insufficient solubility of **277**.

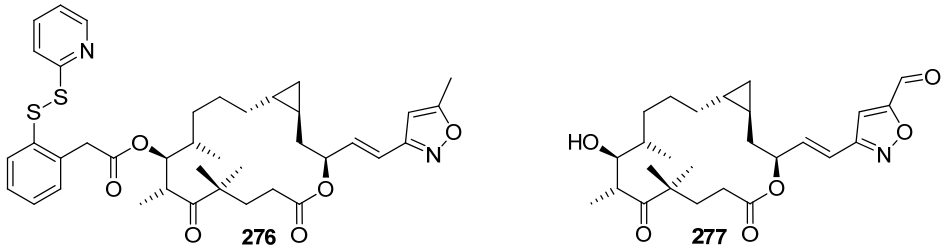


Figure 34: Structure of activated disulfide **276** and aldehyde **277**.

Future work on the construction of epothilone-derived ADC will have to address this solubility issue. A possible approach to overcome this problem would be the attachment of one or several polar, solubilizing moieties to the phenyl ring of the self-immolative chemical linker in **276** (Figure 35). The investigation of this strategy was, however, outside of the scope of this PhD project.

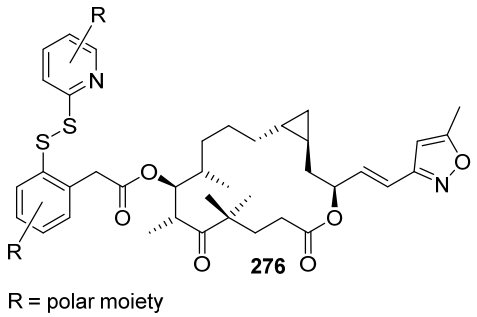


Figure 35: Attachment of a polar moiety at the self-immolative chemical linker in **276**.

2

2 Studies Towards the Total Synthesis of Michaolide E

2.1 Introduction

2.1.1 Isolation and Structural Variants of Cembranes and Cembranolides

Cembranes and cembranolides are cyclic diterpenes. The first member of this group of natural products was described in 1951, when *Haagen-Smit* and *co*-workers reported the isolation of cembrene (**277**) (Figure 36), a crystalline diterpene, from the oleoresin of *Pinus Albicaulis*.[152] About a decade later, studies by *Dauben et al*.,[153] as well as by *Kobayashi* and *Akiyoshi*[154] established the structure of this compound as the first member of a new family of diterpenes, which were named cembranes. Since then, the number of cembranes has grown continuously and to date hundreds of cembranes and cembranolides have been described in literature.

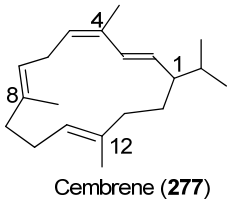
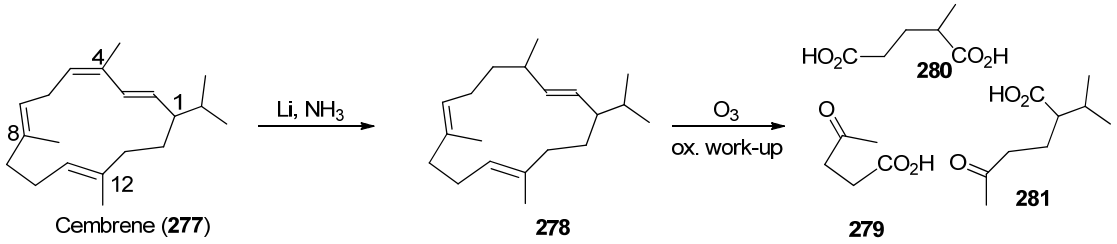


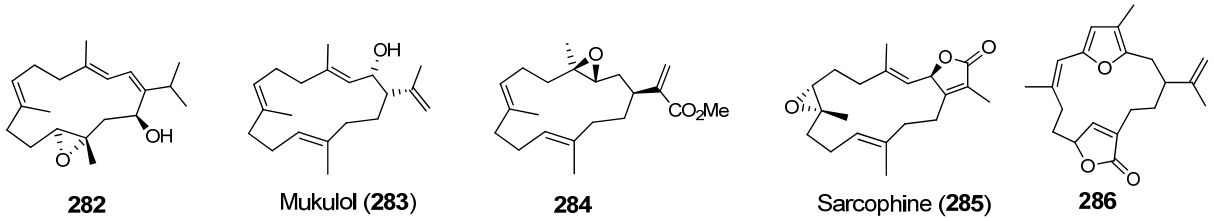
Figure 36: Cembrene (**277**): the first isolated member of the cembrane family of natural products.

Dauben et al. showed that the first isolated cembrane **1**, $C_{20}H_{32}$, contained four double bonds and one ring.[155] Chemical reduction of **277** with lithium in liquid ammonia mainly gave compound **278**, which upon ozonolysis followed by oxidative work-up resulted in the formation of three different acids (Scheme 70). They were characterized as levulinic (**279**), 2-isopropyl-5-oxohexanoic (**281**) and 2-methylglutaric acid (**280**), respectively. The fact that each of these acids was difunctional and all three together accounted for twenty carbon atoms, led to the conclusion that cembrene **1** consisted of an isoprenoid-type fourteen-membered carbocyclic ring.[155]



Scheme 70: Chemical modification of cembrene (**277**) to elucidate its structure.[155]

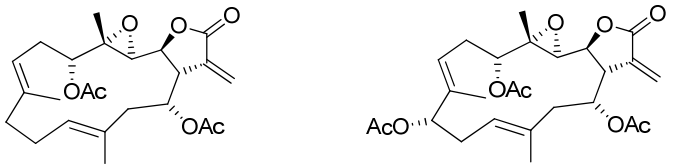
A few natural cembranes have been found in the animal kingdom but many more have been isolated from plants.[156] Those originating from terrestrial plants have been mainly found in pines and tobacco. While the oleoresins of conifers are common sources of hydrocarbons and simple alcohols in the cembrane series, the tobacco plant allows the isolation of chemically more complex natural products like di- and tri-oxygenated derivatives. A much larger number of cembranoids, and typically of a much greater chemical diversity, has been encountered in marine invertebrates, namely the Caribbean gorgonians and the Pacific soft corals. Cembrane diterpenoids from these marine sources mostly bear alcohol functionalities at several positions, they may contain one or more epoxide rings, and often include a lactone moiety.[157],[158] These latter cembrane diterpenoids are named cembranolides. Since the cembrane-type diterpenoids are a large and structurally varied family of natural products, they have been classified. All cembranes and cembranolides share a 14-membered carbon macrocycle as a common structural feature. Different types of cembranes are depicted in Figure 37 to illustrate their structural diversity. The simplest group of cembranes includes isopropyl and isopropenyl cembranes such as **282** and **283** and the somewhat more complex isopropyl or isopropenyl acid cembranes such as **284**.[157] The structurally more complex cembranolides contain of a five-, six-, or seven-membered lactone ring fused to the macrocycle (as exemplified by **285**). Another group of cembranes are the furanocembranoids (**286**). They consist of the 14-membered carbon macrocycle as well as a furan heterocycle.[157]



282 Mukulol (**283**) **284** Sarcophine (**285**) **286**

Figure 37: Structural variants of the cembrane family of diterpenes.

New highly cytotoxic members of the cembranolide family were isolated from the soft coral *Lobophytum michaelae* by *Wang* and *co*-workers in 2001.[159] The dichloromethane extract of *Lobophytum michaelae* showed significant cytotoxic activity against the human colon adenocarcinoma cell line HT-29 and against the mouse lymphocytic leukemia line P-388. Fractionation of the dichloromethane extract led to the isolation of eleven new cytotoxic cembranolides, namely michaolides A-K and the known crassolide **287**.[156] The most potent of these cembranolides, michaolide E (**4**) and crassolide (Figure 38) exhibit IC_{50} values against HT-29 and P-388 cancer cell lines in the low nanomolar range.[159]



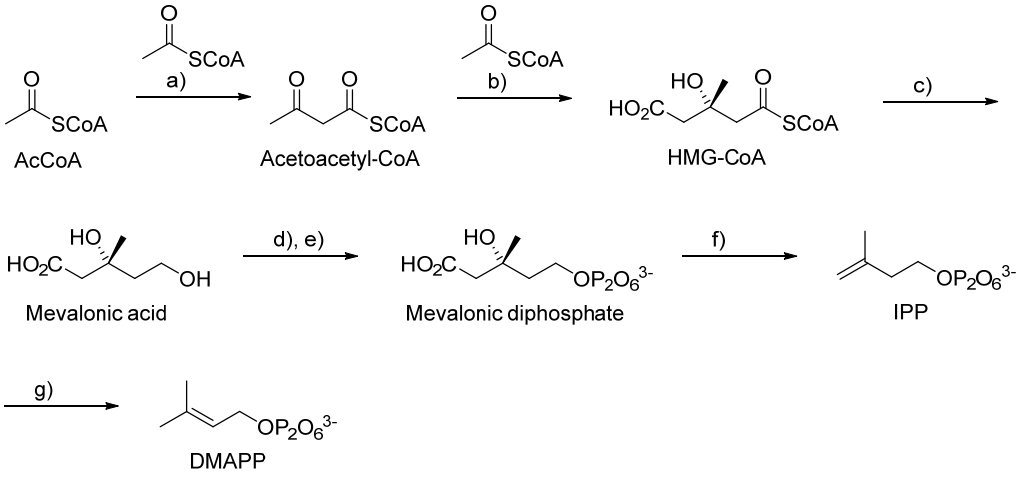
Michaolide E (**4**) Crassolide (**287**)

Figure 38: Highly cytotoxic members of the cembranolide family.

Thus, michaolide E (**4**) was demonstrated to inhibit the growth of HT-29 and P-388 cells with IC_{50}'s of 115 nM and 16 nM, respectively, while crassolide (**287**) was equipotent with michaolide E (**4**) against HT-29 cells (IC_{50} 102 nM) and slightly less potent against P-388 cells (82 nM). Due to its interesting biological activity, its dense arrangement of stereogenic centers and its bicyclic overall structure that features a 5-membered lactone *trans*-fused to a 14-membered macrocycle, michaolide E (**4**) is an attractive and challenging target for total synthesis.

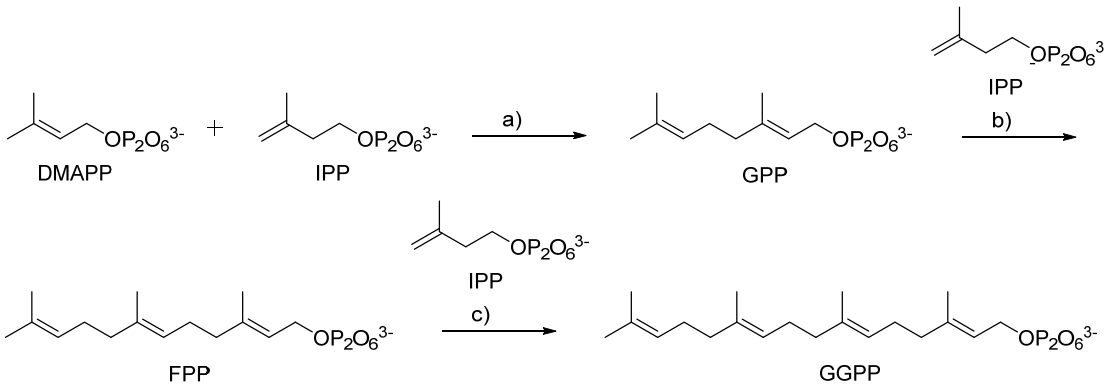
2.1.2 Biogenesis of Cembranes

Cembrene A (**288**) is produced *via* the classical mevalonate pathway for terpene biosynthesis.[160] The extensive family of isoprenoid compounds is derived from two basic building blocks, isopentenyl diphosphate (IPP) and dimethylallyl diphosphate (DMAPP) (Scheme 71). These two precursors are produced from acetyl-coenzyme A (AcCoA) *via* mevalonic acid as the key intermediate (Scheme 71).[161]



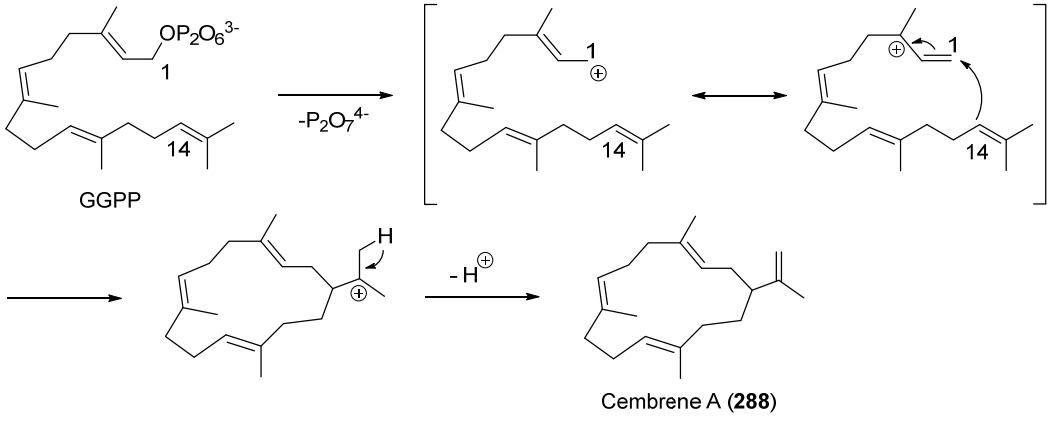
Scheme 71: The mevalonate pathway of the biosynthesis of terpenoids. Enzymes involved in the individual steps are as follows: a) Acetoacetyl-CoA-thiolase; b) 3-hydroxy-3-methyl-glutaryl (HMG)-CoA-synthase; c) HMG-CoA-reductase; d) mevalonate-5-phospho transferase; e) phosphomevalonate kinase; f) pyrophosphate mevalonate decarboxylase; g) IPP-isomerase.

All carbon skeletons of linear isoprenoids are assembled by repetitive head-to-tail connections of IPP with DMAPP. The formation of diterpenes requires the linear assembly of four C5 building blocks (Scheme 72). Coupling of IPP to DMAPP gives geranyl diphosphate (GPP, C_{10}). Addition of IPP to GPP affords farnesyl diphosphate (FPP), which provides is the carbon skeleton of the sesquiterpenes (C_{15}). Upon further coupling of IPP to FPP, the geranylgeranyl diphosphate is formed (GGPP), which is the parent compound of all diterpenes (C_{20}).[161]



Scheme 72: Linear precursor in diterpene biosynthesis. Enzymes involved in the individual steps are as follows: a) Geranyl diphosphate synthase; b) farnesyl diphosphate synthase; c) geranylgeranyl diphosphate synthase.

The biogenesis of cyclic and polycyclic terpenes is usually assumed to proceed *via* carbocation intermediates. In the following the biosynthesis of cembrane A (**288**) – which serves as a pheromone of various termites[162] – is used to exemplify how the 14-membered ring is constructed.[160] Upon dissociation of the pyrophosphate anion from GGPP a resonance-stabilized allylic carbocation is formed, which triggers the cyclization to the fourteen-membered ring skeleton of cembrene A (**288**). The resulting tertiary carbokation is transformed into the cembrene (**288**) *via* abstraction of a proton from one of the two methyl groups attached to the cationic center (Scheme 73).[163]



Scheme 73: Formation of the cembrene A (**288**) *via* cyclization of GGPP.

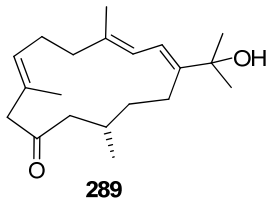
2.1.3 Biological Activity of Cembranes and Cembranolides

Not only are there many structural variants of the cembranes and cembranolides known, they also display a wide range of biological activities.[164] Thus, cembranes can serve a physiological role as insect trail pheremones, the can display antiinflammatory activity, or they can potently inhibit the proliferation of cancer cells.[158],[159],[164],[165] It would be beyond the scope of this section to discuss every cembranolide for which biological data appeared in literature. Rather, the following discussion will focus on compounds exhibiting superior biological activity and members of each class of cembranes and cembranolides will be considered.

2.1.3.1 Antiinflammatory activity

Inflammation is the physiological response to the injury of tissues.[166] Macrophages constitute a main component of the immune system and exhibit a pivotal role in inflammatory responses. They can secrete a variety of inflammatory mediators, such as nitric oxide (NO), prostaglandin E_2 (PGE_2) and cytokines.[167],[168] The production of NO is controlled by nitric oxid synthases (NOS), which include endothelial NOS, neuronal NOS and inducible NOS (iNOS). During inflammation iNOS induces high levels of NO in macrophages.[169] PGE_2, which functions as a mediator of the inflammatory response and induces pain is produced from arachidonic acid through catalysis of cyclooxygenase-2 (COX-2).[170],[171] Hence, the reduction of NO and PGE_2 in macrophages by suppressing the expression of iNOS and COX-2 or by inhibiting their enzymatic activities represents a promising (and also validated) approach for the development of antiinflammatory drugs.[172] In the following the ability of cembranes and cembranolides to reduce the level of iNOS and COX-2 in murine macrophages (RAW 264.7 cells) will be discussed.

In the group of isopropyl cembranes a few members demonstrated antiinflammatory activity such as **289**, which has been isolated from the Formosan soft corals of the genus *Sinularia* in 2008 by *Ahmed* and *co*-workers (Figure 39).[173]

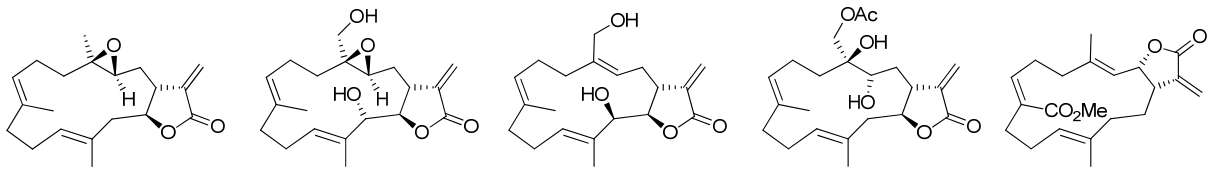


289

Figure 39: Isopropyl cembrane **289** exhibiting antiinflammatory activity.

This compound was found to inhibit the accumulation of the pro-inflammatory iNOS protein in LPS-stimulated RAW 264.7 cells by about 30% at 50 μM concentration, while it was inactive toward the expression of COX-2 protein.

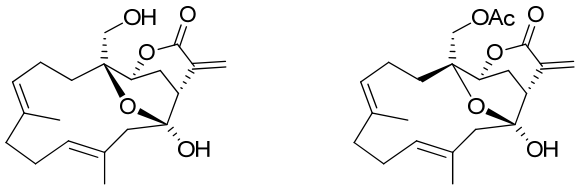
Among cembranolides a number of compounds show antiinflammatory activity that is superior to that of cembranes.[165] For example, Cembranolide **290**, which was isolated from the Formosan soft coral *Sarcophyton crassocaule* in 2010 by *Lin* and *co*-workers (Figure 40), reduced the level of iNOS protein to 4.6% at a concentration of 10 μM; in addition, the expression of COX-2 was also significantly reduced (to 3.9%).[174] The group of *Chen* isolated cembranolides **291** and **292** in 2008 (Figure 40). At a concentration of 10 μM, both compounds suppressed iNOS expression almost completely (to 1.4% and 0.0%, respectively) and to some extent were also able to reduce COX-2 levels (to 42.9% and 42.5%, respectively).[175] Extraction of the soft coral *Lobophytum durum* by the same group resulted in the isolation of cembranolide **293** (Figure 40), which exhibited antiinflammatory activity in a similar range as **291**.[176] In the same year *cis*-cembranolide **294** was isolated from the Formosan soft coral *Sarcophyton crassocaule* by *Chao* and *co*-workers (Figure 40). This compound reduced the level of iNOS to 2.3% and of COX-2 to 34.5% at a concentration of 10 μM.[177]



290 **291** **292** **293** **294**

Figure 40: Cembranolides **290-294** exhibiting significant antiinflammatory activity.

In 2009 Cheng and *co*-workers reported the isolation of 6-membered lactone cembranolides from the soft coral *Lobophytum durum*. This included compounds **295** and **296** (Figure 41), which reduced the levels of iNOS to 11.0% and 0.0% and of COX-2 to 66.7% and 34.7%, respectively at a concentration of 10 μM.[178]



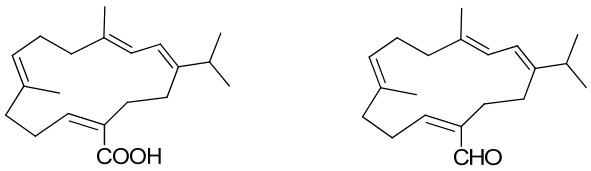
295 **296**

Figure 41: 6-membered lactone cembranolides **295** and **296** exhibiting antiinflammatory activity.

Regarding the structural features associated with cembranolides with potent antiinflammatory activity, it appears that the presence of the α-methylene-γ-lactone functionality is required to significantly reduce the expression of iNOS and COX-2 in LPS-stimulated RAW 264.7 cells. Based on this observation it may be speculated that a covalent interaction of the compounds with the proteins *via* the *Michael* acceptor system occurs.

2.1.3.2 Cytotoxic activity

Among the isopropyl cembranes a few members are found that exhibit cytotoxic activity as for example neocrotocembraneic acid (**297**) and neocrotocembraneic aldehyde (**298**), which were isolated from the stem bark of *Croton oblongifolius* by *Roengsumran* in 1999 (Figure 42).[179] Their cytotoxic activity against the mouse leukemia line P-388 was evaluated, with acid **297** exhibiting an IC_{50} value of 137 μM, while aldehyde **298** was about 6 times more potent (23 μM).[180]

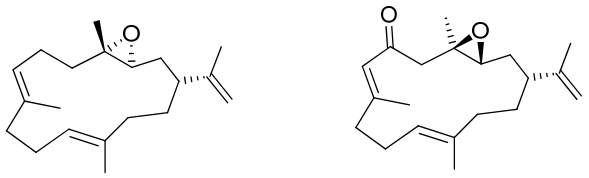


297 **298**

Figure 42: Isopropyl cembrane **297** and **298** exhibiting moderate cytotoxic activity.

In the same year isopropenyl cembrane **299** was isolated from the soft coral *Nephthea brassica* by *Duh* and *co*-workers (Figure 43). This compound showed antiproliferative activity against human cancer cell lines in the low micromolar range, exhibiting IC_{50}'s of 11.9 μM against human lung carcinoma cell lines A549, 9.0 μM against the human colon carcinoma cell lines HT-29 and 1.3 μM against the mouse leukemia cell line P-388. [181] The

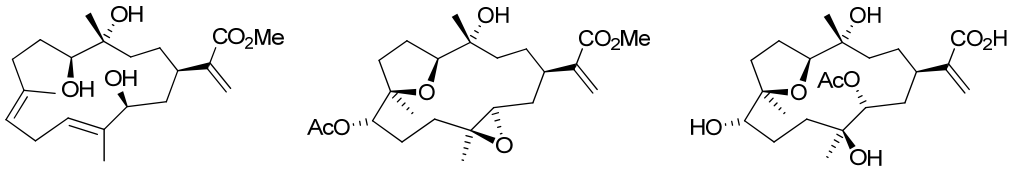
group of *Ortega* isolated isopropenyl cembrane **300** from the gorgonian *Leptogorgia laxa* in 2008 (Figure 43). The compound was demonstrated to be about as active as **299** (IC_{50} values of 5.6 µM and 10.9 µM against A549 and HT-29 cells, respectively).[182]



299 **300**

Figure 43: Epoxy-isopropyl cembranes **299** and **300**. These compounds exhibit moderate cytotoxic activity.

In the course of investigations on the chemical diversity of soft corals in the South China Sea, *Ma* and *co*-workers collected an unidentified species of the genus *Dendronephtya*. This led to the isolation of three cytotoxic isopropenyl acid cembranes **301-303** (Figure 44). All compounds showed selective and significant inhibition of the growth of the human cancer cell line BGC-823 with IC_{50} values of 136 nM (**301**), 47 nM (**302**), and 466 nM (**303**), respectively), while no activity was observed against other cancer cell lines (HCT8, Bel-7402, A549, and A2780).[183]



301 **302** **303**

Figure 44: Isopropenyl acid cembrane **301-303** exhibiting cytotoxic activity.

The most active compounds of these natural products are found among the cembranolides. *Iwashima* and *co*-workers isolated α-methylene γ-lactone cembranolide **304** from *Clavularia koellikeri* in 2000 (Figure 45). The antiproliferative activity of this compound was examined against the human colorectal adenocarcinoma cell line (DLD-1) and human T-lymphocyte leukemia cells (MOLT-4). **304** showed IC_{50} values of 8.9 µM (DLD-1) and 1.9 µM (MOLT-4).[184] The first chemical investigation of the gorgonian octocoral *Eunicea pinta* by the group of *Shi* resulted in the isolation of α-methylene γ-lactone cembranolides **305** and **306** (Figure 45). While compound **305** was demonstrated to exhibit significant cytotoxic activity against the non-small cell lung cancer cell line NCI 60 (IC_{50} 2.7 µM) and TK-10 renal cancer cells (IC_{50} 0.4 µM), compound **306** displayed strong growth inhibition of human T lymphocytic leukemia cells (MOLT-4, IC_{50} 26 nM).[185]

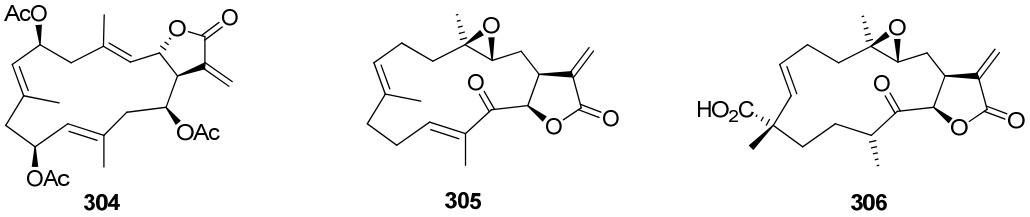


Figure 45: Cembranolides **304-306** exhibiting significant cytotoxic activity.

307 and **308** belong to the most active compounds among the furanocembranoids (Figure 46). They were isolated by *Pudhom* and *co*-workers in 2007 as part of an effort to isolate biologically active compounds from *Croton oblongifolius*. Both compounds showed broad cytotoxic activity against all cell lines tested (BT474, CHAGO, HEP-G2, KATO-3, SW-620) with IC_{50}'s around 20 µM.[186]

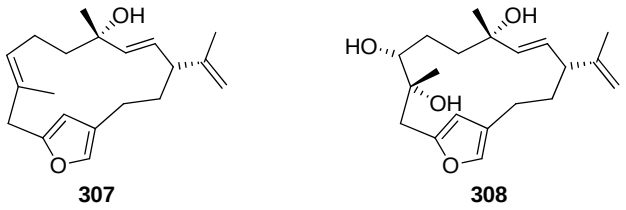


Figure 46: Furanocembranoids **307** and **308** exhibiting cytotoxic activity.

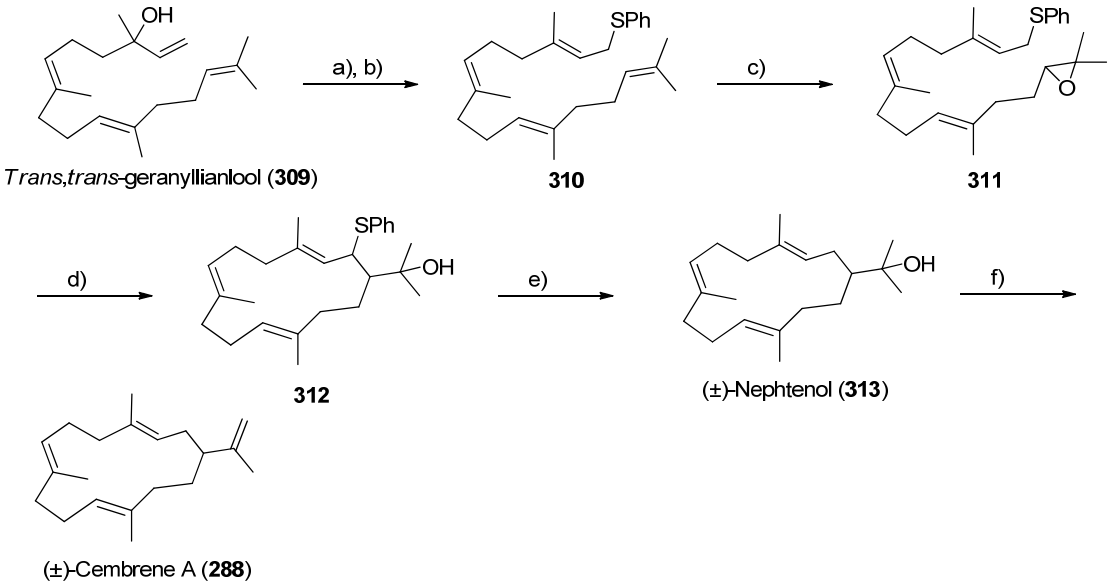
Based on the available structure-activity data, the presence of an α-methylene γ-lactone ring is not absolutely required for antiproliferative activity against human cancer cells by cembranolides. However, as for cembranolides with antiinflammatory activity, the most active compounds do include this structural element.

2.1.4 Total Synthesis of Cembranes and Cembranolides

In this section, previous total syntheses of cembranes and cembranolides will be briefly reviewed. Again, it is not the intention of this chapter to provide a comprehensive account of all syntheses that have been developed. Rather, the different concepts and approaches that have been pursued to access natural products of the cembrane and cembranolide families will be highlighted for specific examples. In particular, the last section of this chapter will provide an overview of the different strategies that have been employed to close the 14-membered carbon macrocycle.

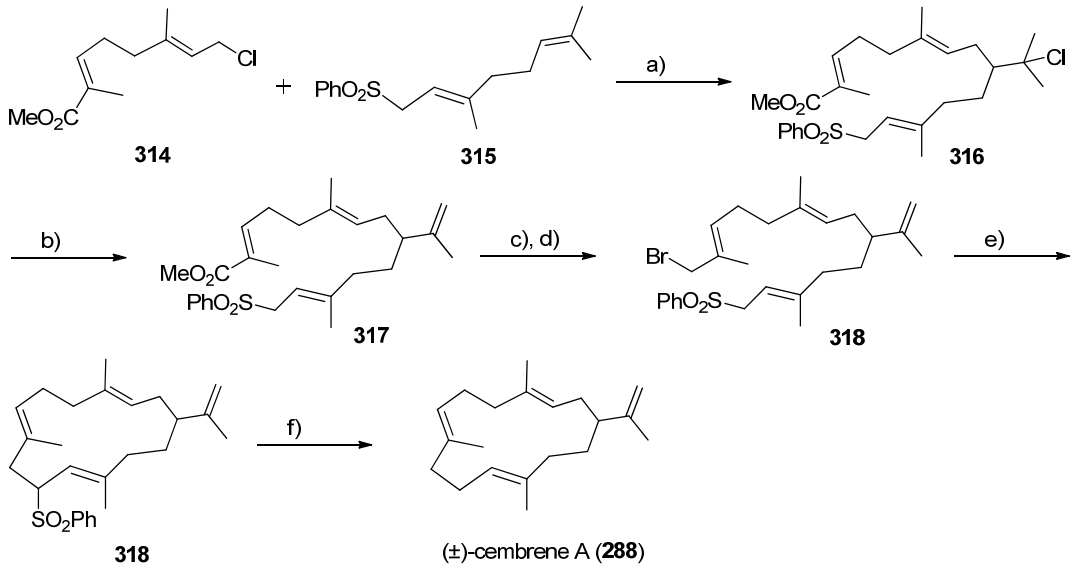
2.1.4.1 Total Syntheses of Cembrene A

One of the first syntheses of a cembrane-type natural product was reported in 1975 by *Kodama* and *co*-workers, who prepared cembrene A (**288**) by making use of an intramolecular nucleophilic addition of a sulfur-stabilized carbanion to an epoxide (Scheme 4).[187] *Trans*,*trans*-geranyllinalool **309** was converted into the thioether **310** *via* an *Appel*[188] reaction and subsequent nucleophilic displacement of the bromide substituent. *Van Tamelen*'s procedure[189] allowed the selective epoxidation of the terminal double bond in moderate yield (42%). Upon treatment of thioether **311** with *n*-BuLi the cyclization was triggered to give macrocycle **312** in 62% yield. Reductive cleavage of the thioether gave nepthenol (**313**). Finally, dehydration of the tertiary alcohol afforded the *Hofmann* product (±)-cembrane A (**288**).



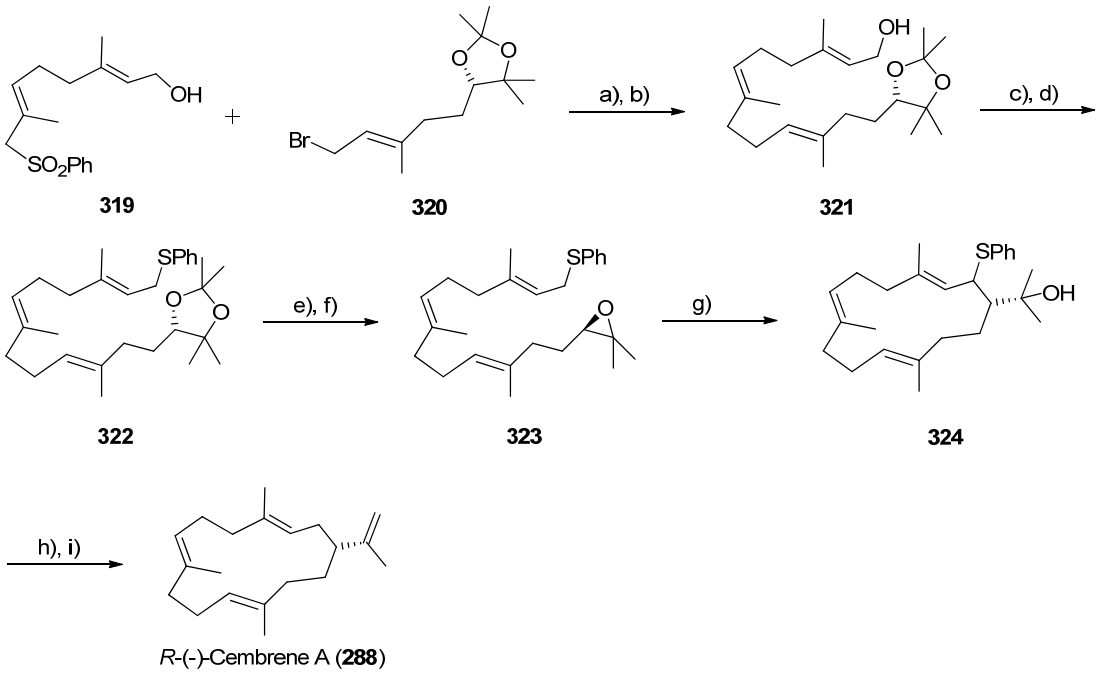
Scheme 74: a) PBr_3; b) NaSPh, 73% over two steps; c) i) NBS, aq. THF; ii) K_2CO_3, MeOH, 42%; d) *n*-BuLi, DBU, THF, -78 °C, 62%; e) Li, $EtNH_2$, -78 °C, 30%; f) $SOCl_2$, py, 95%.

Racemic cembrene A (**288**) was also synthesized by *Takayanagi* and *co*-workers in 1978, using a convergent route with a regiospecific coupling of two functionalized geranyl units as the key step (Scheme 75). The advantage of this approach is that functional groups can be introduced regioselectively prior to cyclization.[190] Treatment of chloride **314** in the presence of sulfone **315** and $SnCl_4$ as a *Lewis* acid produced tertiary chloride **316**. The subsequent elimination was carried out by spraying the tertiary chloride onto a silica plate that was kept at room temperature for four days. If the dehydrochlorination was induced by addition of LiBr and Li_2CO_3, the more stable *Zaitsev* product was formed in 80% yield. Subsequent LAH reduction of the ester moiety gave the corresponding allylic alcohol, which was converted into the bromide **318** by making use of the *Appel* reaction in 14% overall yield from **317**. Macrocycle formation was then induced by addition of LDA to afford **319**. Final reductive cleavage of the sulfonyl group gave (±)-cembrene A (**288**).



Scheme 75: a) $SnCl_4$, CH_2Cl_2, -94 °C, 44%; b) silica gel, 80%; c) LAH, THF, -20 °C, quant.; d) PPh_3, CBr_4, MeCN; e) LDA, THF, -78 °C, 56% over two steps; f) Li, $EtNH_2$, -78 °C, 69%.

The first enantioselective total synthesis of *R*-(-)-cembrene A (**288**) was reported by *Schwabe* and *co*-workers in 1988.[191] As for the *Kodama* synthesis of racemic cembrene A (**288**) (Scheme 75), their convergent synthesis employed an intramolecular nucleophilic addition of a sulfur-stabilized anion to an epoxide, but in this case the epoxide moiety was chiral (Scheme 76). Coupling of the sulfonyl fragment **319** with the allylic bromide **320**, which was synthesized from *L*-serine in 12 steps and 17% overall yield,[192] gave the linear precursor. Subsequently, the sulfonyl group was removed reductively to give the allylic alcohol **321**, which was transformed into thioether **322**. After cleavage of the acetonide moiety the formation of the required epoxide was triggered by treatment of the resulting diol with MsCl in the presence of K_2CO_3. Subsequent addition of *n*-BuLi to **323** then induced cyclization. Finally, the thioether was removed and the tertiary alcohol was eliminated using $SOCl_2$ in pyridine to produce *R*-(-)-cembrane A (**288**).

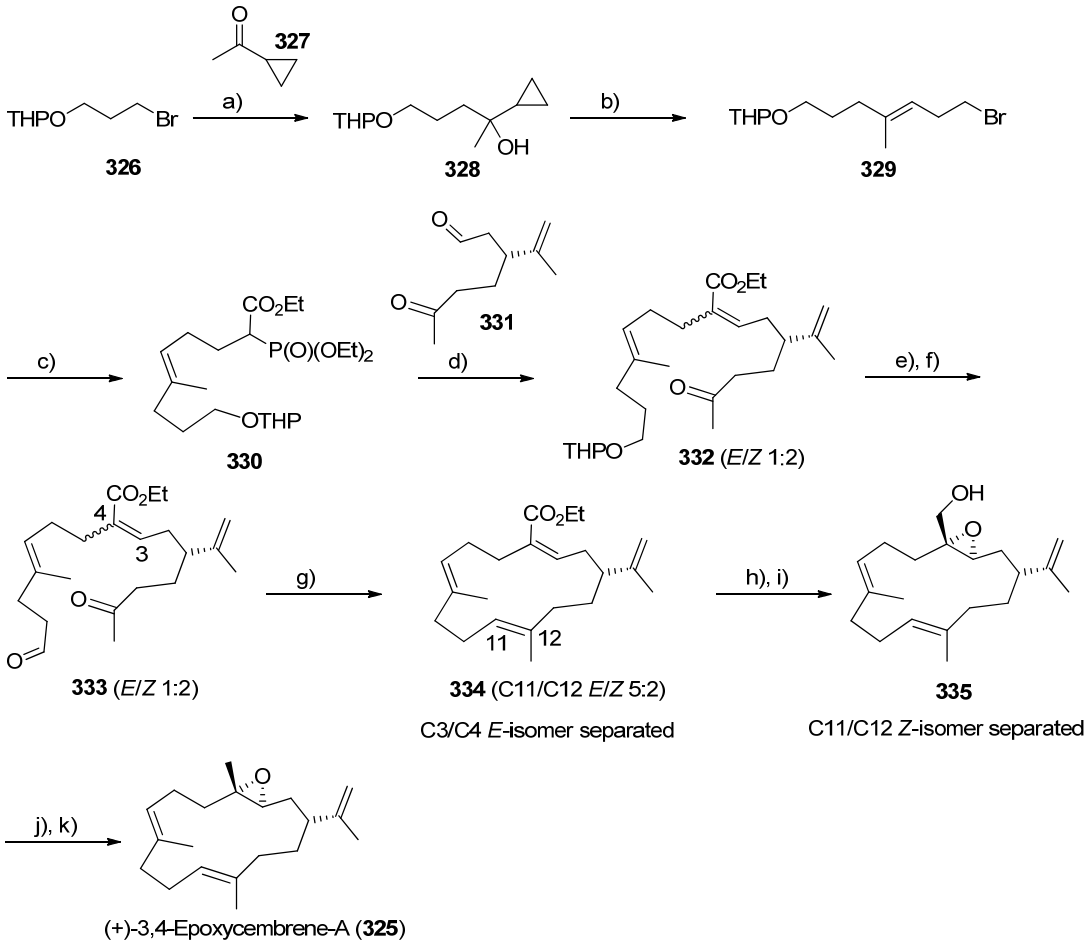


Scheme 76: a) *n*-BuLi, THF, -78 °C, 81%; b) Na/Hg, EtOH, reflux, 90%; c) PBr_3, py, Et_2O, -10 °C; d) NaSPH, MeOH, 81% over two steps; e) Amberlite IR-120, ethylenglykol/DME, 60 °C, 91%; f) i) MsCl, py; ii) K_2CO_3, MeOH, 32%; g) *n*-BuLi, THF, -78 °C, 27%; h) Na, *t*-BuOH, 90 °C, 55%; i) $SOCl_2$, py, 45%.

2.1.4.2 Total Syntheses of Natural Epoxy Cembrenoids

Out of the group of epoxy cembrenoids (+)-3,4-epoxycembrene A (**325**) shall serve as an example. It was first isolated in 1981 by *Bowden et al.*[193] from the Australian soft coral *Sinularia facile*. The absolute configuration of this epoxy derivative of cembrane *R*-(-)-cembrene A (**288**) (*vide supra*, Scheme 76) was determined by means chemical degradation and spectroscopic techniques. **325** demonstrated to inhibit the growth of human cancer cell lines with moderate potency in the low micromolar range.[181] In 2001 *Liu et al.* reported the first total synthesis of (+)-3,4-epoxycembrene A (**325**) making use of a chiral pool protocol and a *Sharpless* asymmetric epoxidation[194] to install the chiral centers (Scheme 77).[195] The assembly of the carbon skeleton of **325** commenced with the addition of the lithium derivative of bromide **326** to cyclopropyl methyl ketone **327** which gave cyclopropane **328** in 84% yield.[196] Treatment of **328** with LiBr in the presence of TMSCl then provided the rearranged homoallylic bromide **329** in 79% overall yield.[197] Subsequent alkylation of **329** gave phosphonate **330**, which underwent *Horner-Wadsworth-Emmons* (HWE) olefination with aldehyde **331** (derived from *R*-(+)-limonene by ozonolysis[198]) to form a mixture of *E*/*Z* isomers in a 1:2 ratio. Deprotection of the primary hydroxy group with

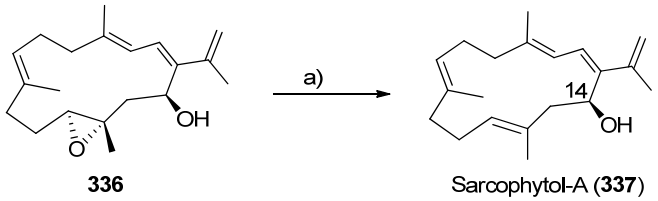
followed by PCC oxidation gave keto aldehyde **333**, which was subjected to *McMurry* olefination[199] to achieve cyclization. The reaction proceeded in 41% yield and gave a mixture of C11/C12 *E*/*Z* isomers in a 5:2 ratio. At this stage the undesired C3/C4 *E*-isomer could be separated. DIBAL-H reduction of the ester moiety in **334** gave the corresponding allylic alcohol as a separable 5:2 mixture of C11/C12 isomers. Subsequent *Sharpless* asymmetric epoxidation of the desired isomer afforded epoxide **335** (90%, dr 20:1). Finally, the alcohol **335** was transformed into the corresponding iodide by means of an *Appel* reaction; the latter was then reductively dehalogenated to yield (+)-3,4-epoxycembrene-A (**325**).



Scheme 77: a) Li, THF, then **327**, 84%; b) LiBr, TMSCl, CH_2Cl_2, 79%; c) NaH, $(EtO)_2P(O)CH_2CO_2Et$, DMF, 60 °C 79%; d) *n*-BuLi, THF, -78 °C to -20 °C, then **331**, -20 °C, 70%; e) *p*-TsOH, MeOH; f) PCC, NaOAc, CH_2Cl_2, 85% over two steps; g) $TiCl_3$, Zn-Cu, DME, reflux, 41%; h) DIBAL-H, CH_2Cl_2, -78 °C, 92%; i) $Ti(OiPr)_4$, *D*-(-)-DET, *t*-BuOOH, CH_2Cl_2, -20 °C, 90%; j) PPh_3, imidazole, py, I_2, Et_2O/MeCN, 0 °C; k) $NaBH_3(CN)$, HMPA/THF, 60 °C, 77% over two steps.

Another member of the epoxy cembrenoids is (+)-11,12-epoxysarcophytol A (**336**). Its synthesis will be briefly discussed as it demonstrates that the epoxide moiety is stable enough to be carried through a multistep synthesis. **336** had been isolated by *Bowden* and *co*-workers

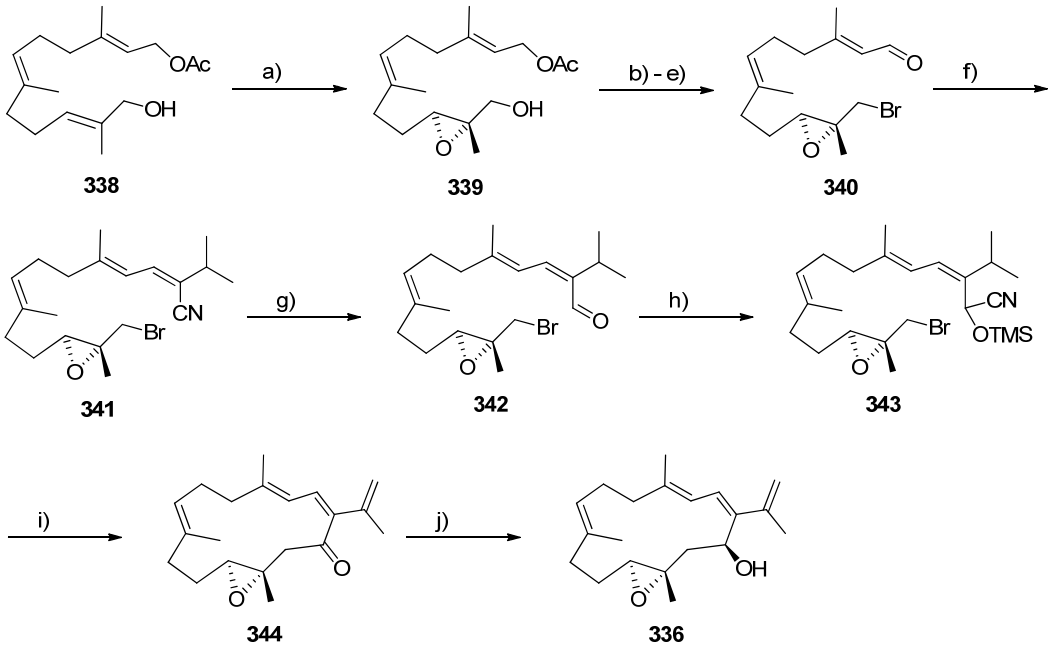
from an Australian marine soft coral *Lobophytum* in 1983.[200] The configuration of the hydroxy group at C14 was assigned as *S* by a zinc-copper-mediated reductive elimination of the epoxide moiety, which results in the formation of the known cembrane diterpenoid sarcophytol A (**337**) (Scheme 78).[201]



336 Sarcophytol-A (**337**)

Scheme 78: a) zinc dust, $CuSO_4$, EtOH, reflux.

Li and *co*-workers reported the first total synthesis of **336**, which includes a *Sharpless* asymmetric epoxidation, an intramolecular cyanohydrin alkylation and CBS reduction as the key transformations (Scheme 79).[202] The synthesis started from readily available *trans*,*trans*-farnesol derivative **338**, which underwent a *Sharpless* asymmetric epoxidation to give epoxide **339** (95%, 98% ee) The resulting alcohol **339** was then converted into the corresponding bromide, followed by a deprotection/oxidation sequence which resulted in the formation of enal **340** in 75% overall yield from epoxy alcohol **339**. The aldehyde was then condensed with 3-methyl-2-(diethylphosphono) butanenitrile under *HWE* conditions to yield the desired *Z* olefin **341**.[203],[204],[205] Reduction of the cyano group gave an imine that was hydrolyzed under aqueous acidic conditions to provide unsaturated aldehyde **342**.[206] Treatment of **342** with TMSCN gave the cyanohydrin trimethylsilyl ether **343**, which upon addition of LiHMDS formed the desired macrocycle. Subsequent treatment of the crude cyclization product with a catalytic amount of TBAF gave epoxy ketone **344**. Selective CBS reduction finally led to the natural product **336**.

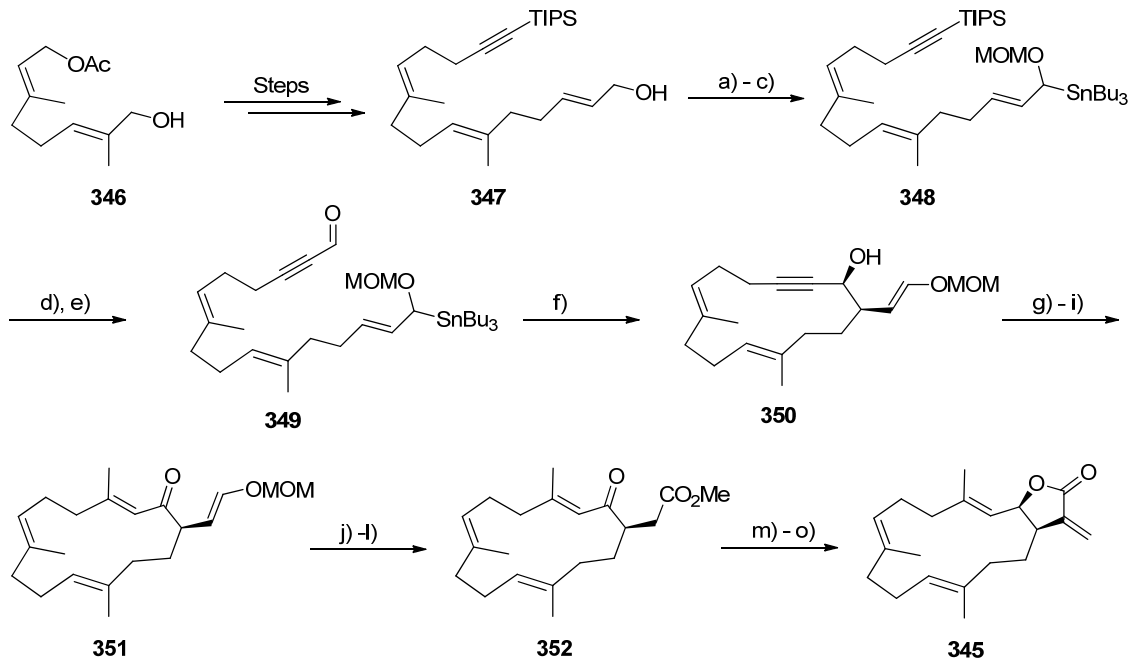


Scheme 79: a) $Ti(OiPr)_4$, *L*-(+)-DET, *t*-BuOOH, CH_2Cl_2, -40 °C, 95%, 98% ee; b) MsCl, NEt_3, CH_2Cl_2, -10 °C to 0 °C; c) LiBr, acetone, 50 °C, 82% over two steps; d) K_2CO_3, MeOH; e) MnO_2, hexane, 91% over two steps; f) LiHMDS, $(Me)_2CHCH(CN)P(O)(OEt)_2$, toluene, -78 °C, then **340**, 90%; g) DIBAL-H, hexane, -78 °C, then 10% aq. oxalic acid, 0 °C, 88%; h) TMSCN, cat. KCN/18-C-6, THF; i) LiHMDS, reflux, then TBAF, 10% aq. THF, 85% over two steps; j) $BH_3{\cdot}SMe_2$, CBS (10 mol%), toluene, 0 °C, 88%.

2.1.4.3 Total Synthesis of Cembranolides

Marshall and *Crooks* total synthesis of (±)-*cis*-cembranolide **345**, published in 1987, deserves special mentioning as it features the addition of lithium dimethyl cuprate to an ynone and an α-alkoxy allylstannane macrocyclization (Scheme 80).[207],[208],[209] The initial steps of the synthesis involved the elaboration of oxidized geranyl acetate **346** into allylic alcohol **347**,[207] which was then oxidized to the corresponding enal. Subsequently, $LiSnBu_3$ was added to the α,β-unsaturated aldehyde and the resulting alcohol was MOM-protected to give **348**. After TIPS-removal, deprotonation of the terminal alkyne moiety with LDA and quenching of the alkyne anion with paraformaldehyde led to the formation of a propargylic alcohol, which was oxidized to the ynone **349**. Treatment of **349** with the *Lewis* acid $BF_3{\cdot}OEt_2$ afforded the macrocycle **350** in good yield as a 7:1 diastereomeric mixture in favour of the desired 1*R*,2*S* isomer. Propargylic alcohol **350** was then oxidized to the ynone, which underwent a non-selective cuprate addition with $LiCuMe_2$ to give a 1:1 mixture of the *E* and *Z* isomeric addition products. The *Z* isomer could be isomerized by addition of isopropyl thiolate to give

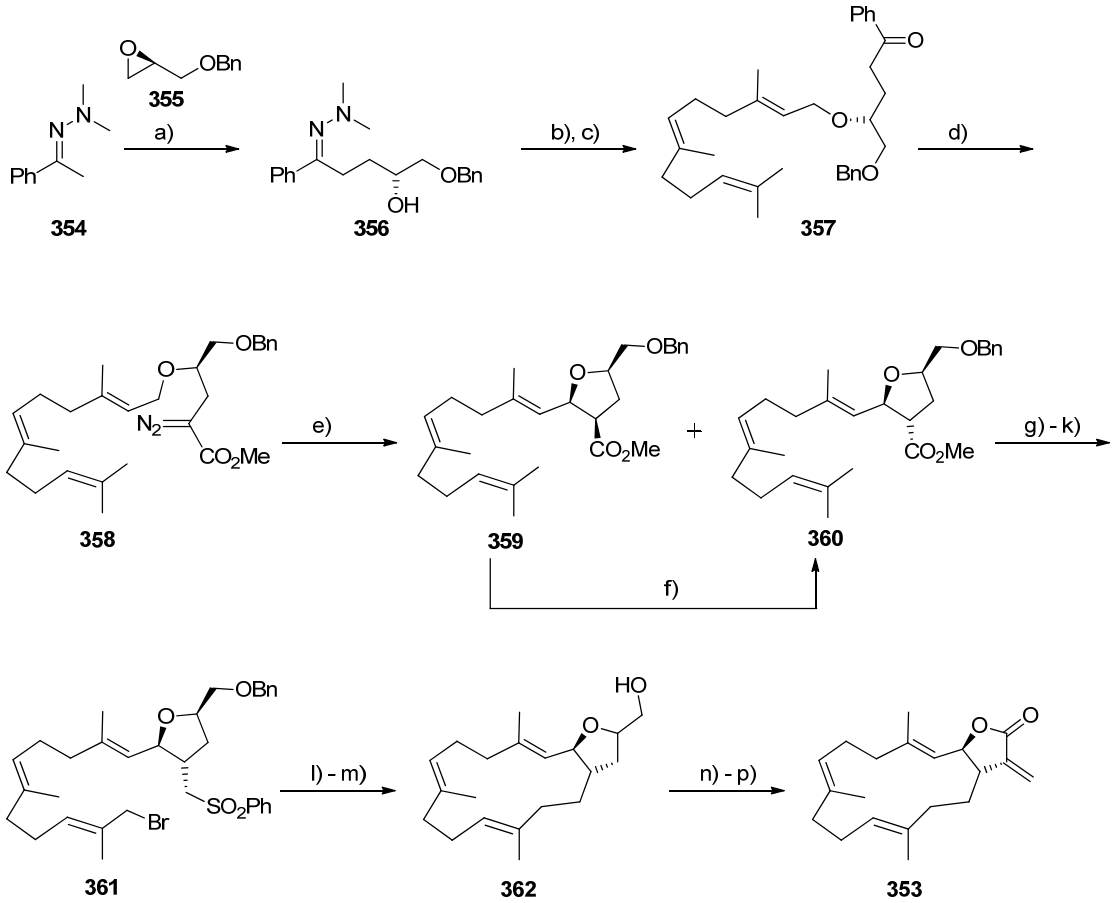
the *E* isomer **351** as the sole product. Acidic removal of the MOM-protecting group was followed by oxidation of the ensuing aldehyde to the acid; the latter was esterified by treatment with diazomethane to give ester **352**. Finally, the keto group was reduced with $NaBH_4$ to provide a 10:1 mixture of lactones in favour of the desired *cis*-fused system. Installation of the methylene group by hydroxymethylation with LDA and paraformaldehyde and subsequent dehydration then gave the natural product **345**.



Scheme 80: a) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to RT; b) $LiSnBu_3$, THF, -78 °C; c) MOMCl, iPr_2NEt, CH_2Cl_2, 0 °C; d) LDA, THF, -78 °C, then $(CH_2O)_n$, 50% over four steps; e) 1,1'-azodicarbonyldipiperidine, *t*-BuOMgBr, THF, 0 °C, 85%; f) $BF_3{\cdot}OEt_2$, CH_2Cl_2, -78 °C, 88%, dr 7:1; g) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to RT, 87%; h) $LiCuMe_2$, THF, 0 °C, 97%, dr 1:1; i) LiS*i*Pr, THF, 90%; j) HCl, H_2O, 83%; k) PDC, DMF, 78% ; l) CH_2N_2, Et_2O, 0 °C, 95% ; m) $NaBH_4$, EtOH, 70%, dr 10:1; n) LDA, THF, -78 °C to -20 °C, then $(CH_2O)_n$, 66%; o) MCDI, $CuCl_2$, MeCN, 60 °C, 69%.

With regard to the above discussed synthesis of (±)-*cis*-cembranolide **345** the asymmetric total synthesis of its *trans* analogue (-)-*trans*-cembranolide **353** by *Taber* and *Song* shall be shown. The key transformation of the synthesis is a diastereoselective Rh-mediated cyclization of diazo ester **358** to form a tetrahydrofuran ring (Scheme 81).[210] The latter was obtained from epoxide **355**, which was opened with the lithium anion derived from hydrazone **354**. The resulting γ-hydroxy hydrazone **356** was then alkylated with *trans*,*trans*-farnesyl bromide. Subsequent treatment with aqueous acid afforded ketone **357**, which upon treatment with dimethyl carbonate and NaH, followed by exposure to DBU and 4-nitrobenzenesulfonlyl azide gave α-diazo ester **358**.[211] Treatment of **358** with catalytic

amounts of rhodium octanoate triggered cyclization to tetrahydrofuran derivates **359** and **360** in a 2:1 ratio. The lack of stereocontrol in the cyclization was not an issue, since the minor diastereomer **359** could be epimerized to **360** with NaOMe. After the introduction of the sulfone moiety *via* nucleophilic substitution, one of the terminal methyl groups was oxidized with SeO_2 and the resulting allylic alcohol was transformed into bromide **361**. Sulfone **361** cyclized on exposure to LDA[190] and subsequent treatment of the macrocycle with Na in liquid ammonia removed both the phenylsulfonyl group and the benzyl group to give primary alcohol **362**. Oxidative cleavage of **362** with PDC led to the lactone.[212],[213] Final methenylation α to the carbonyl group of the lactone gave (-)-*trans*-cembranolide **353** in 1.2% overall yield from epoxide **355**.[214]

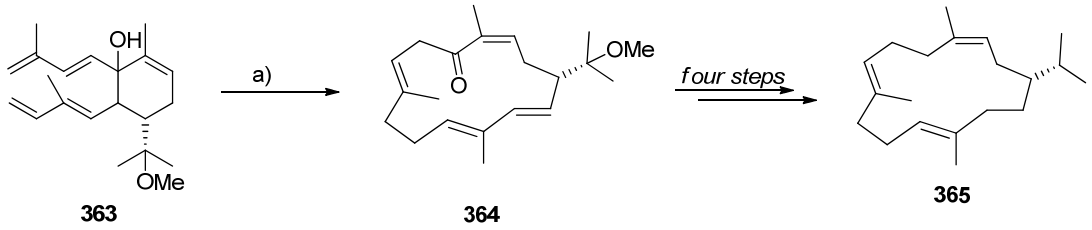


Scheme 81: a) LiHMDS, toluene, 0 °C, then **355**, $Y(OTf)_3$, 80%; b) NaH, THF, 0 °C, then farnesyl bromide, TBAI; c) aq. HCl, THF, 59% over two steps; d) NaH, dimethylcarbonate, DME, then DBU, *p*-NBSA, CH_2Cl_2, 0 °C to RT, 63%; e) $Rh(Oct)_2$, CH_2Cl_2, 58%; f) NaOMe, MeOH, 88%; g) $LiAlH_4$, THF, 0 °C, 84%; h) TosCl, NEt_3, DMAP, CH_2Cl_2, 0 °C; i) TBAI, $NaSO_2Ph$, THF, reflux, 84% over two steps; j) SeO_2, salicylic acid, *t*BuOOH, CH_2Cl_2; k) PPh_3, CBr_4, CH_2Cl_2, 0 °C, 50% over two steps; l) LDA, THF, - 78°C; m) Na, NH_3, THF/EtOH, -78 °C, 61% over two steps; n) PDC, Ac_2O, CH_2Cl_2/DMF, 58%; o) $(OMe)CH(NMe_2)_2$, 90 °C; p) DIBAL-H, THF, -78 °C, 64% over two steps.

2.1.4.4 Different cyclization in Synthesis of Cembranoids

Since the formation of the 14-membered macrocycle is a crucial step in the synthesis of all cembranoids, the different strategies that have been pursued to achieve macrocyclization shall be briefly discussed in this section. The examples discussed in the preceding section have already illustrated the utility of sulfide/sulfo-stabilized carbanion alkylations, the Ti(0)-induced *McMurry* olefination, cyanohydrin-stabilized carbanion alkylations and α-alkoxy allylstannane-based macrocyclizations for the construction of the 14-membered ring.

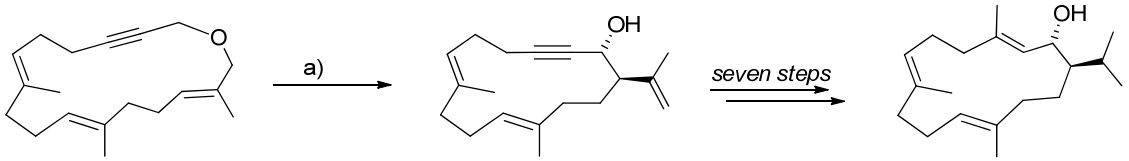
Yet another approach to macrocycle formation has been employed by *Wender* and *co*-workers in their synthesis of (-)-3*Z*-cembrene A (**365**), where they made use of a ring expansion reaction[215],[216] to construct the macrocyclic ring (Scheme 82).[217]



363 **364** **365**

Scheme 82: a) KH, 18-C-6, THF, 55%.

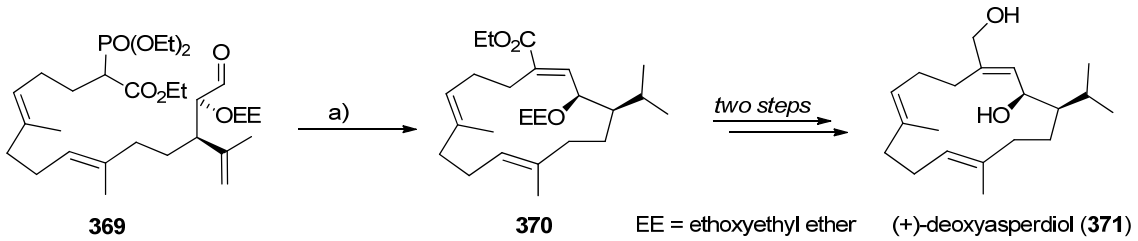
In contrast, *Marshall* employed a rather ingenious a *ring contraction* strategy to establish the macrocycle in the synthesis of *epi*-mukulol (**368**) (Scheme 83).[218] In this case, deprotonation of ether **366** triggers a stereoselective *Wittig* rearrangement.[219]



366 **367** *Epi*-mukulol (**368**)

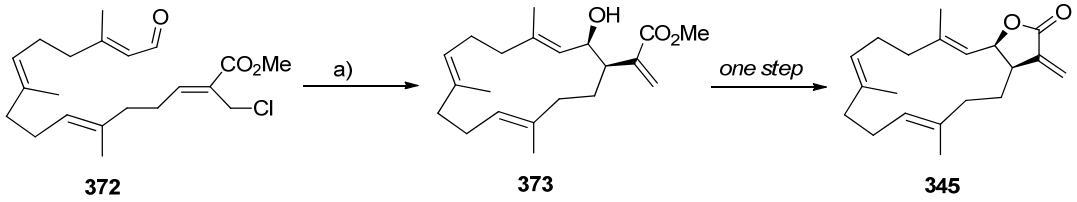
Scheme 83: a) *n*-BuLi, THF/HMPA 3:1, -78 °C, 85%, dr 4.5:1.

The effectiveness of the *HWE* reaction to construct the macrocycle in cembranoids has been demonstrated in the total synthesis of (+)-deoxyasperdiol (**371**) by *Tius* and *Fauq* in 1986 (Scheme 84).[220] The conditions developed by *Masamune*, *Roush* and *Rathke*,[221],[222] which entail the use DBU and lithium chloride in acetonitrile, produced trisubstituted olefin **370** as a 2:1 mixture of *trans*/*cis*-isomers from phosphonate **369**.



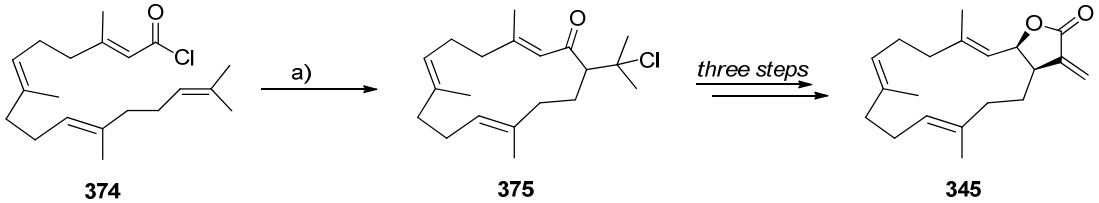
Scheme 84: a) DBU, LiCl, MeCN, 30%, *E*/*Z* 2:1.

A similar approach as that followed by *Marshall* (Section 2.1.4.3, Scheme 80) was employed by *Nishitani* and *co*-workers in their synthesis of (±)-*cis*-cembranolide **345**. However, instead of using an α-alkoxy allylstannane-based macrocyclization, *Nishitani* and *co*-workers made use of a Cr(II)-mediated intramolecular coupling reaction between an allylic chloride and an aldehyde moiety in **372**, which gave the desired *anti*-substituted macrocycle **373** in good yield and selectivity (Scheme 85).[223],[224]



Scheme 85: a) $CrCl_2$, DMF, 81%.

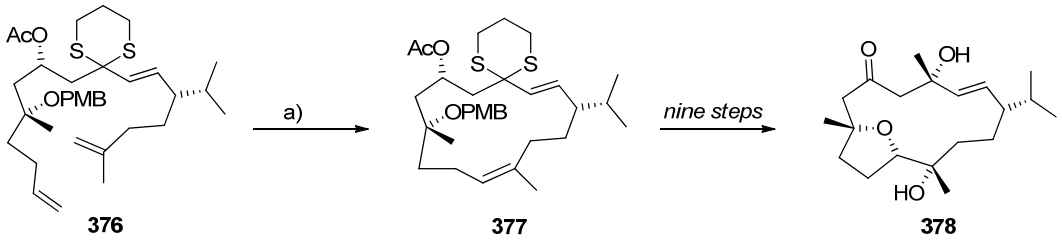
(±)-*cis*-cembranolide **345** has also been prepared *via Friedel-Crafts*-type acylation reaction by *Kato* and *co*-workers (Scheme 86).[225] Remarkably, the reaction proceeded in high yield, giving chloride **375** as the exclusive cyclization product, although the formation of a 10-membered ring would also have been conceivable.



Scheme 86: a) $SnCl_4$, CH_2Cl_2, -78 °C, 71%.

Of particular relevance for the work described in this PhD thesis, *Tietze et al*. in 2008 reported a total synthesis of the polyoxygenated cembrene (**378**) that was based on ring-closure by means of ring-closing metathesis (RCM) (Scheme 87). Remarkably, the macrocycle was formed in high yield and exclusively as the *Z*-isomer (**377**) in the presence of *Grubbs* 2nd generation catalyst. Some general aspects of RCM-based macrocyclizations that involve the

formation of trisubstituted double bonds in the cyclization step will be discussed in greater detail in next section.

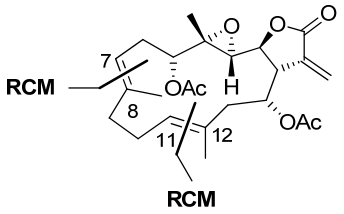


376 **377** **378**

Scheme 87: a) *Grubbs* II (7 mol%), CH_2Cl_2, reflux, 89%.

2.2 Aims and Scope

As indicated in the introduction, michaolide E (**4**) has recently been identified as a new member of the cembranolide family of natural products, which displays highly potent antiproliferative activity against human cancer cells *in vitro*. So far, no total synthesis of michaolide E (**4**) has been reported in the literature and neither has any structure-activity work been described around this interesting natural product. In order to provide a basis for the exploration of michaolide E (**4**) as a potential lead structure for anticancer drug discovery, it was an objective of this thesis to establish an efficient and stable synthesis of this natural product. The chemistry developed in the course of the total synthesis work should then be exploited in a subsequent phase for the synthesis of analogs for structure-activity relationship (SAR) studies (although this would be outside of the scope of this PhD thesis). At the level of synthesis strategy, the work was also to explore the possibility of constructing the 14-membered macrocycle in michaolide E (**4**) by means of RCM between C7/C8 or C11/C12, as this was felt to provide a particular efficient approach to the target structure (Scheme 88).



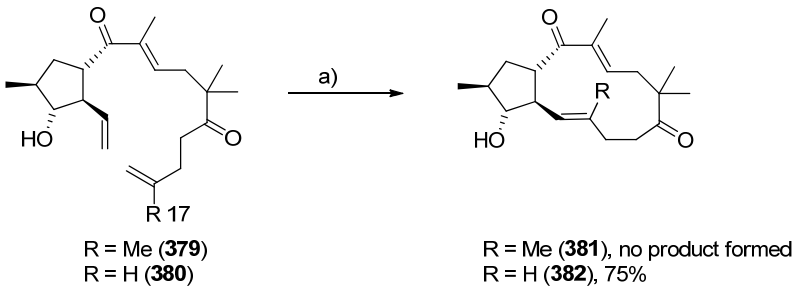
Michaolide E (**4**)

Scheme 88: Intended RCM at C7/C8 or at C11/C12.

However, it was far from clear whether this strategy could be implemented successfully. Apart from the general issue if efficient ring-closure could be achieved, the stereochemical outcome of the cyclization reaction was open to question. As discussed above, only one RCM-based cembranolide synthesis has been reported so far, which afforded exclusively the *Z*-isomer (i. e. the undesired isomer in our approach). In order to put these questions into perspective, the next section will provide a brief overview of the examples of RCM-based macrocyclizations leading to 13-membered or larger rings by the formation of trisubstituted double bonds.

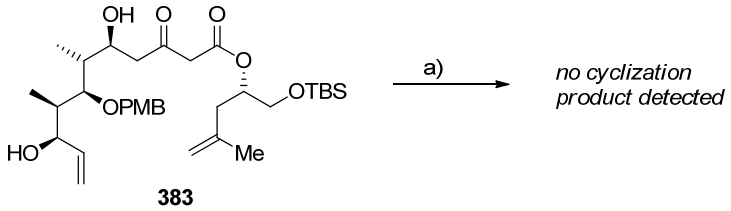
2.2.1 RCM-based Macrocylizations

Unsuccessful attempts at the cyclization of diene **379** have been reported by *Hiersemann* and *co*-workers in the presence of both 1st and 2nd generation *Grubbs* and *Hoveyda-Grubbs* catalysts (in the context of a projected synthesis of 15-acetyl-3-propionyl-characiol) (Scheme 89). In contrast, diene **380**, which lacks the C17 methyl group, yielded the macrocycle **382** in 75% yield using *Grubbs* 2nd generation catalyst.[226] This outcome indicates that steric reasons may be responsible for the failure of diene **379** to cyclize, which would have involved the formation of a trisubstituted double bond.



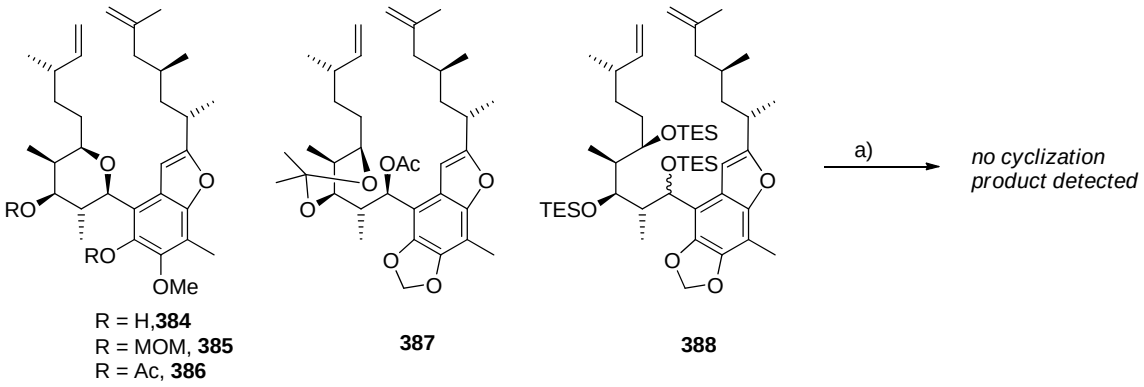
Scheme 89: a) Grubbs II, DCE, 60 °C.

As for the example above, *Hoye* and *Zhao* as part of their studies towards the synthesis of callipeltoside A revealed the unsuccessful construction of a 14-membered macrocycle from diene **383** by means of RCM (Scheme 90).[227]



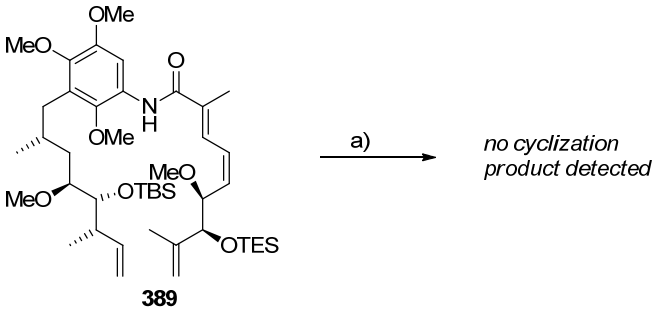
Scheme 90: a) *Grubbs* I and II.

Unsuccessful cyclization attempts have also been reported for 16-membered macrocycles as target structures. Thus, *Mulzer* and *co*-workers attempted to form a 16-membered macrocycle in the course of their studies towards the total synthesis of (-)-kendomycin.[228] None of the depicted dienes **384-388** underwent cyclization using *Grubbs* 2nd generation or *Schrock* catalyst (Scheme 91).



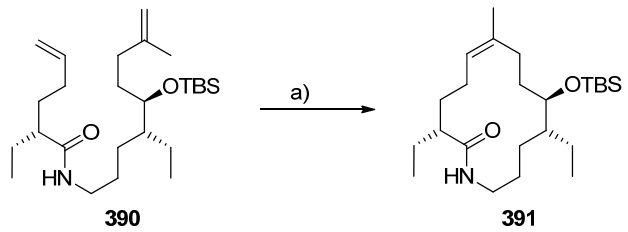
Scheme 91: a) *Grubbs* II or *Schrock* catalysts.

In the context of their total synthesis of (+)-geldanamycin *Ma* and *co*-workers attempted to close a 19-membered ring by means of RCM using *Grubbs* 2nd generation catalyst under a variety of conditions to no avail (Scheme 92).[229]



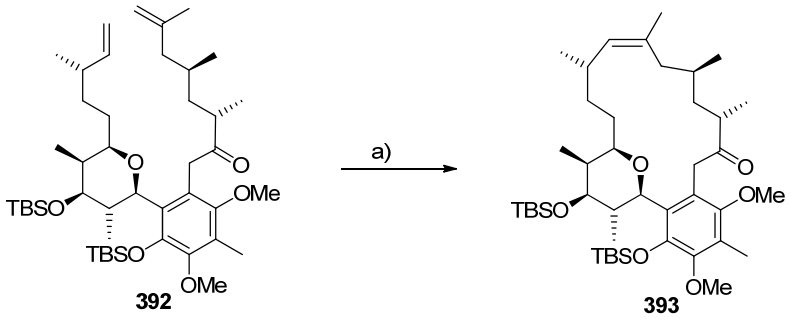
Scheme 92: a) *Grubbs* II.

However, in addition of the examples discussed above, a number of cases have been reported in the literature where RCM-based macrocyclization with the formation of a trisubstituted double bond in a >13-membered ring has been executed successfully. Four of these reports describe the exclusive formation of the *Z*-isomer including *Tietze*'s work on polyoxygenated cembrene (**378**) (Section 2.1.4.4, Scheme 87), where the *Z* olefin was the desired product.[230] Another successful example where a *Z* olefin was intended to be installed by RCM was described by *Hoveyda* and *co*-workers (Scheme 93).[231]



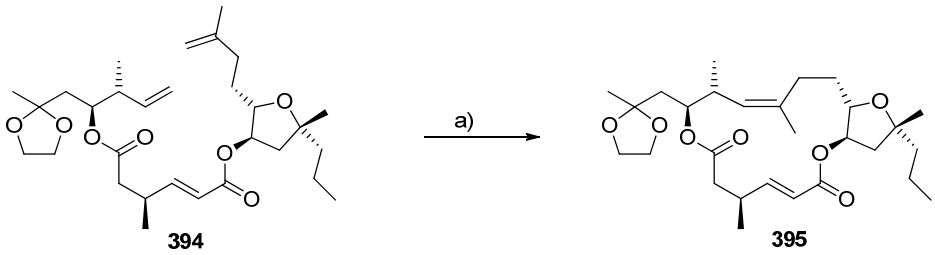
Scheme 93: a) *Schrock* catalyst, benzene, 90%.

In the two other cases of selective *Z* isomer formation the desired product had in fact been the *E*-isomer, as in the context of the total synthesis work on (-)-kendomycin by *Smith et al*. (Scheme 94).[232] However, it is remarkable that the RCM furnished the macrocycle at all, since *Mulzer et al*. failed to close the ring with a very similar substrate (Scheme 91).



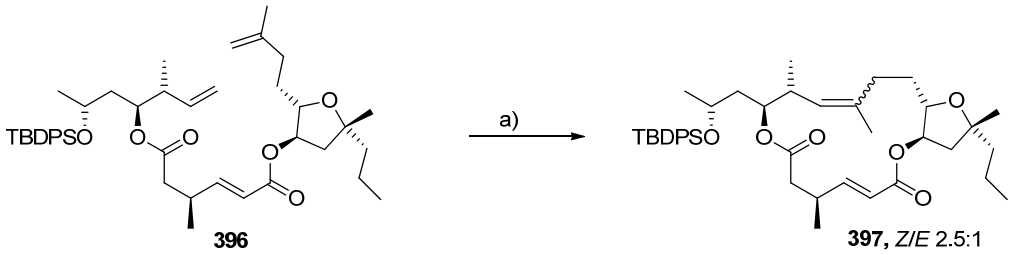
Scheme 94: a) *Grubbs* II, CH_2Cl_2, reflux, 57%.

Undesired *Z* isomer formation was also observed by *Vilarrasa* and *co*-workers in the course of their work on the total synthesis of amphidinolide X. The attempt to cyclize diene **394** using different amounts of *Grubbs* 2nd and *Hoveyda-Grubbs* 2nd generation catalyst only gave the undesired *Z*-isomer in moderate yield (Scheme 95).[233]



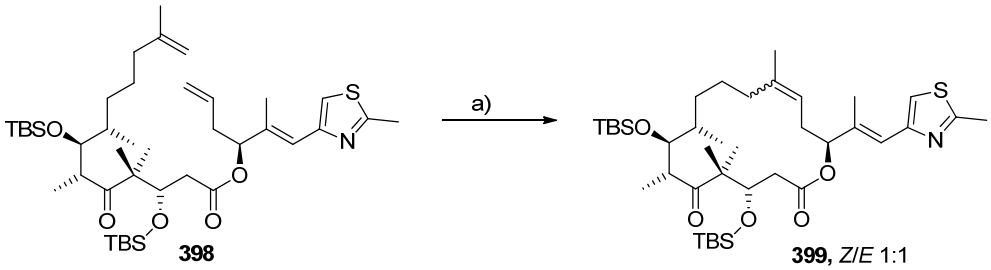
Scheme 95: a) *Grubbs* II or *Hoveyda-Grubbs* II, 30-40%.

In contrast to *Villarasa*'s finding's, *He* and *co*-workers reported the formation of detectable amounts of the *E* isomer upon RCM with diene **396** – structurally very similar to **394** – although the *Z* isomer was still the predominant product (Scheme 96).[234]



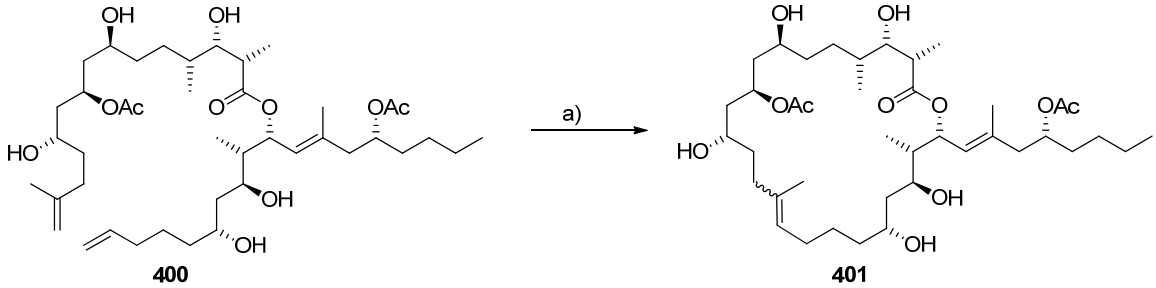
Scheme 96: a) *Grubbs* II, CH_2Cl_2, reflux, 50%, *Z*/*E* 2.5:1.

In other cases macrocyclizations mediated by means of RCM proceeded in an essentially unselective manner. For example, in their total synthesis of epothilone B *Grieco* and *May* isolated a 1:1 mixture of *E*- and *Z*-isomers using *Schrock* catalyst (Scheme 97).[235]



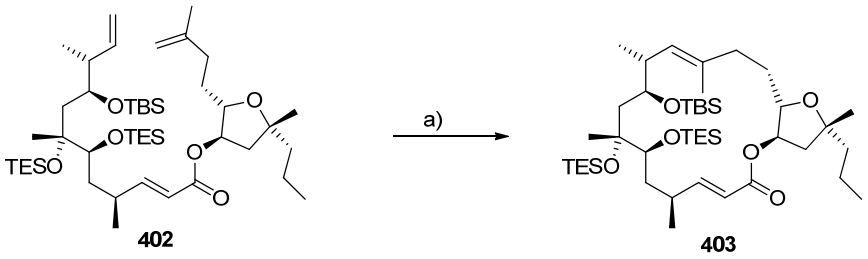
Scheme 97: a) *Schrock* catalyst, benzene, 55 °C, 55%, *Z*/*E* 1:1.

In *Leighton's* synthesis of Dolabelide D a 24-membered ring was to be closed. The macrocycle formed with low stereoselectivity (*E*/*Z* 1.3:1), but in reasonable yield (Scheme 98).[236]



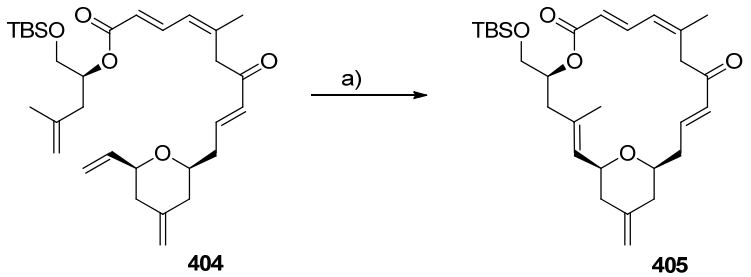
Scheme 98: a) *Grubbs* II, CH_2Cl_2, reflux, 55%, *E*/*Z* 1.3:1.

Surprisingly, *Dai* and *co*-workers have reported an *E*-selective RCM with diene **402** (Scheme 99),[237] which is structurally related to the dienes **394** and **396** that were investigated by *Villarasa* and *He*, respectively (Scheme 95 and 96), and which were favouring formation of the *Z* isomer. This observation highlights the fact that even small changes in substrate structure can have a major impact on the stereochemical outcome of RCM reactions.



Scheme 99: a) *Grubbs* II, CH_2Cl_2, reflux, 40%.

Finally, *Lee* and *co*-workers reported the cyclization of diene **404** to macrocycle **405** as the only isomer in acceptable yield as part of their synthesis of (-)-dactylolide (Scheme 100).[238]



404 **405**

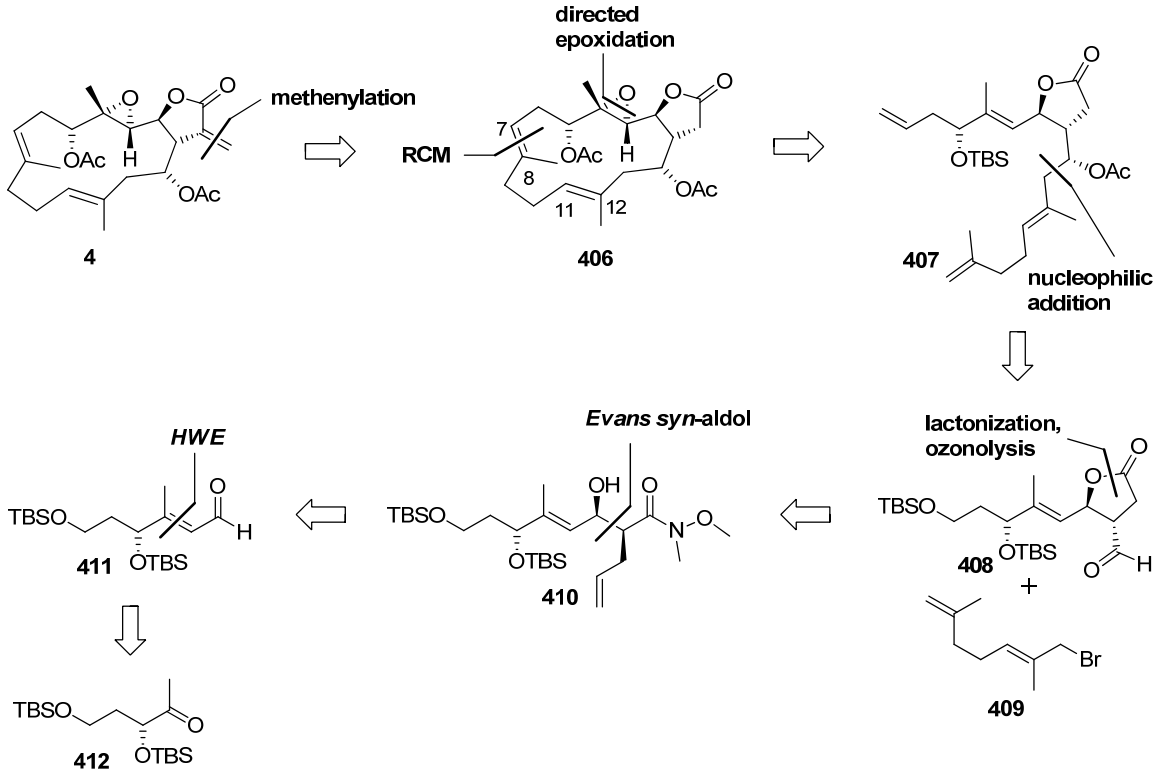
Scheme 100: a) *Grubbs* II, CH_2Cl_2, reflux, 45%.

The described reports clearly demonstrate that the outcome of a macrocyclization mediated by means of RCM resulting in a trisubstituted double bond is not predictable in terms of yield as well as *E* to *Z* stereoselectivity. Nevertheless, the existence of successful cyclizations by RCM favouring the *E* isomer and the fact that there are two trisubstituted double bonds in the target molecule, which means that the macrocyclization could be attempted on different substrates, was hope enough to follow that risky strategy.

2.3 Results and Discussion

2.3.1 RCM at C7/C8; Early Introduction of Lactone

From a retrosynthetic perspective it was planned to introduce the methylene group on the γ-lactone ring at the very end of the synthesis, following the guiding principle that a reactive moiety should be introduced into a target structure as late as possible. As indicated in section 2.2 the macrocyclic ring was to be closed by a ring-closing metathesis (RCM) between C7/C8. The epoxide moiety in **406** was to be introduced by a directed epoxidation subsequent to macrocycle formation. Lactone **407** was envisioned to be obtained by nucleophilic addition of metalated allylic bromide **409** to aldehyde **408**; which should be accessible from *Weinreb* amide **410** *via* an ozonolysis/lactonization/reduction sequence. *Evans syn*-aldol addition would serve to install the two adjacent stereocenters of **410** with a *syn* relationship and the α,β-unsaturated aldehyde **411** required for this aldol reaction would be derived from methyl ketone **412** by means of *HWE* olefination, which is accessible from *D*-malic acid exploiting a reaction sequence discussed earlier in this thesis in the context of the synthesis of side chain-modified cyclopropyl-Epo B analogs (Section 1.5.1.2, Scheme 16).[113]



Scheme 101: Retrosynthetis of michaolide E (**4**). First generation approach.

As for the addition of metalated **409** to aldehyde **408**, a similar transformation had been reported in the literature with a stereochemical outcome that was also required for the synthesis of michaolide E (**4**). While the authors did not discuss the possible reasons for the observed selectivity, a plausible explanation would be that the aldehyde group orientates in such a way as to minimize the dipole moment of the lactone **408** and with the nucleophile preferentially attacking to enter from the *exo* side of the 5-membered ring, which constitutes to a *Re*-face attack (Figure 47).

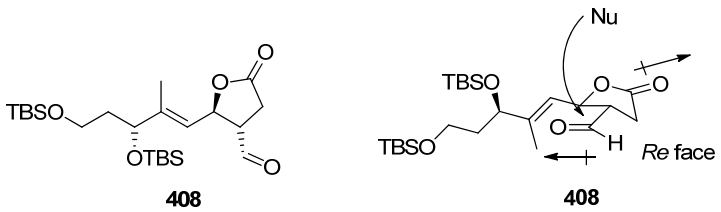
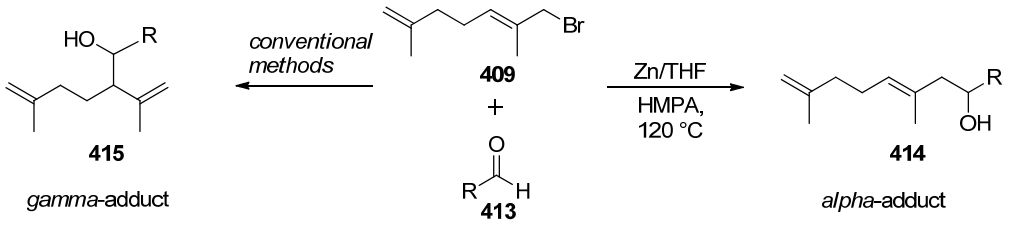


Figure 47: *Re*-face attack of a nucleophile on aldehyde **408**.

A second issue that needed to be addressed for the nucleophilic addition of metalated allylic bromide **409** to aldehyde **408** was the regioselectivity of the reaction. According to a method developed very recently by *Zhang* and *co*-workers the α-adduct **414** is favoured in zinc-mediated additions in the presence of HMPA, albeit at very high temperatures (Scheme 102). However, higher temperatures were expected to compromise the stereoselectivity of the addition and the question was whether proper conditions would be found that would satisfy both, the stereo- as well as the regiochemical requirements for this transformation.



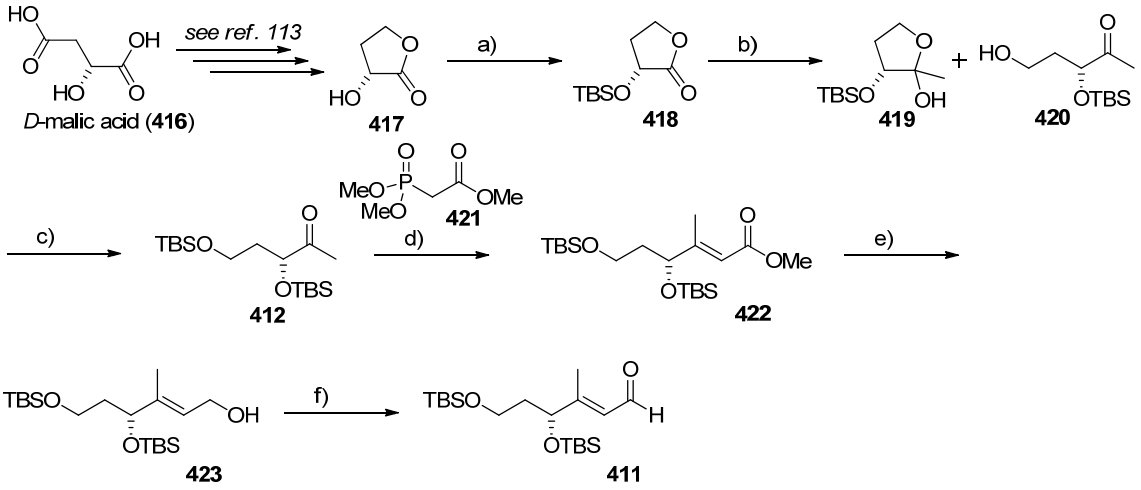
Scheme 102: γ-addition *versus* α-addition of metalated allylic systems.

2.3.1.1 Forward Synthesis

The transformation of *L*-malic acid into α-hydroxy lactone **105** (Section 1.5.1.2, Scheme 16) is known in the literature.[114],[115],[239],[116] For the synthesis of methyl ketone **412,** the sequence was started with *D*-malic acid (**416**), which was transformed into α-hydroxy lactone **417**.[10] TBS-protection of **417** was followed by treatment with methyllithium to give a

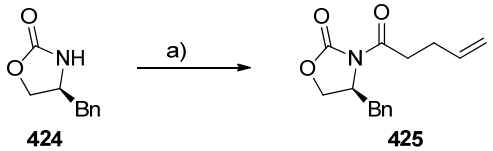
[10] α-Hydroxylactone **417** is commercially available, albeit at a high price.

mixture of TBS-protected hemiacetal **419** and hydroxy ketone **420** (Scheme 103). Treatment of the mixture with TBSCl led to the formation of *bis*-silyl-protected methyl ketone **412**, which underwent *HWE* olefination in high yield and good selectivity. At 65 °C an *E*/*Z* selectivity of 10:1 was observed. This ratio could be improved by lowering the temperature at the expense of longer reaction times. If the reaction was carried out at 45 °C, a selectivity of 12:1 was achieved and conversion was complete within 16 hours. The resulting α,β-unsaturated ester **422** was subsequently reduced to the allylic alcohol **423**; MnO_2 oxidation of **423** yielded α,β-unsaturated aldehyde **411** in 62% overall yield from α-hydroxy lactone **417**.



Scheme 103: a) TBSCl, imidazole, DMF, 96%; b) MeLi, THF, -78 °C, 89%; c) TBSCl, imidazole, CH_2Cl_2, 98%; d) NaH, **421**, THF, then **412**, 45 °C, 91% *E*/*Z* 12:1; e) DIBAL-H, CH_2Cl_2, -78 °C, 89%; f) MnO_2, Et_2O, 91%.

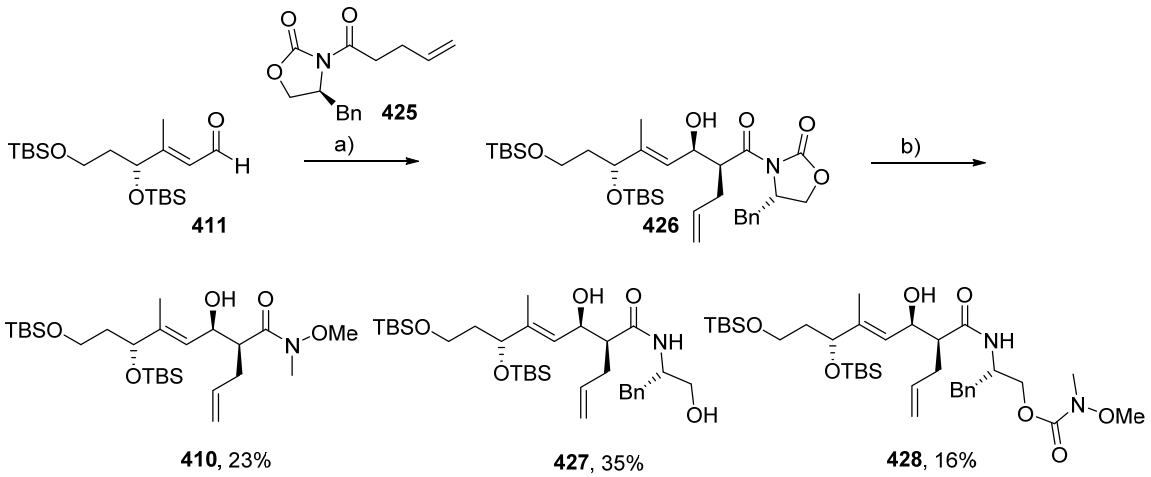
The stage was now set to carry out the *Evans syn*-aldol addition. The *Evans*-auxiliary derivate **425** was synthesized in one step according to the literature (Scheme 104).[240]



Scheme 104: a) *n*-BuLi, pentenoyl chloride, THF, - 78° C, 86%.

Boron-mediated aldol addition of **425** with aldehyde **411** gave the *syn*-aldol product in good yield and with excellent selectivity (Scheme 105). Quite surprisingly, however, the subsequent cleavage of the auxiliary with simultaneous formation of the *Weinreb* amide turned out to be extremely difficult.[11]

[11] No example of a successful installation of a *Weinreb*-amide from an acyl oxazolidinone was found in literature for a substrate with a methyl group attached to the double bond next to the secondary OH group as in **426**.



Scheme 105: a) **425**, Bu_2BOTf, DIPEA, CH_2Cl_2, -78 °C, 80%, dr 30:1; b) $AlMe_3$, $Me(OMe)NH \cdot HCl$, THF, -40 °C, 23%.

The highest achievable yield was 23%. Up to 6 eq. of $AlMe_3$ – the use of Me_2AlCl did not trigger any transamidation – were required to get full conversion. It was reasoned that the secondary hydroxy group might be sterically hindered due to its close proximity to the methyl group attached to the double bond. As a consequence, the amide carbonyl group is not activated by complexation as depicted in **A** (Figure 48, **A**). Rather, complexation and, thus, activation towards nucleophilic attack might involve the carbonyl group of the oxazolidinone ring, resulting in the formation of **428**, which is partially hydrolyzed to **427** during work-up. Alternatively, complexation of the exocyclic imide carbonyl group may have occurred, but the addition of the nucleophile to the amide was sterically hindered (Figure 48, **B**).

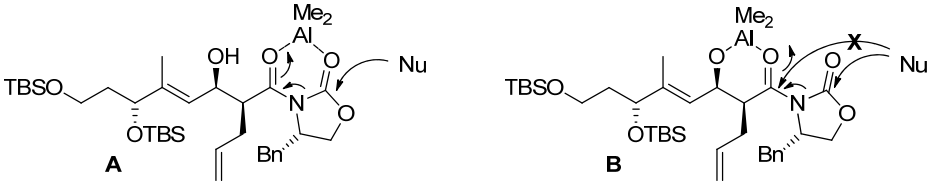
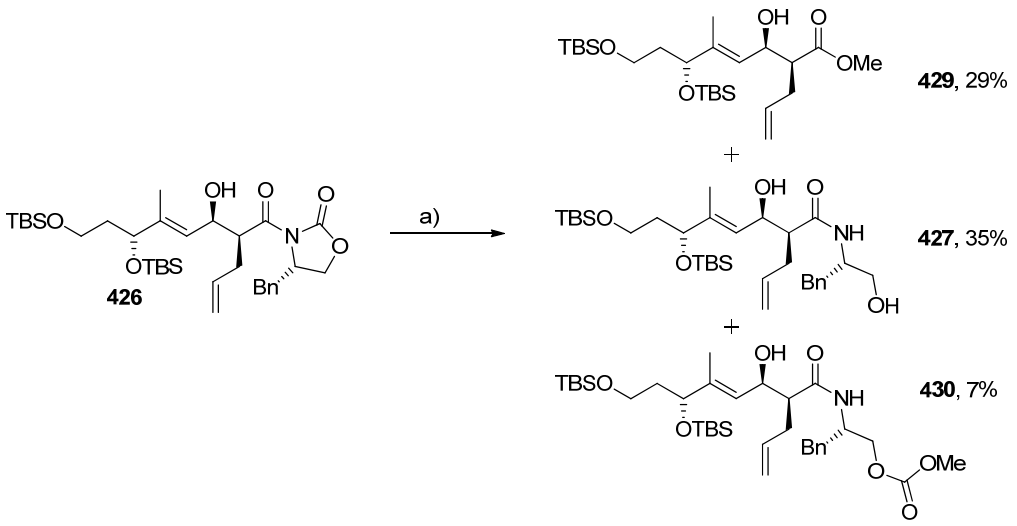


Figure 48: Explanations for the outcome of the removal of the auxiliary.

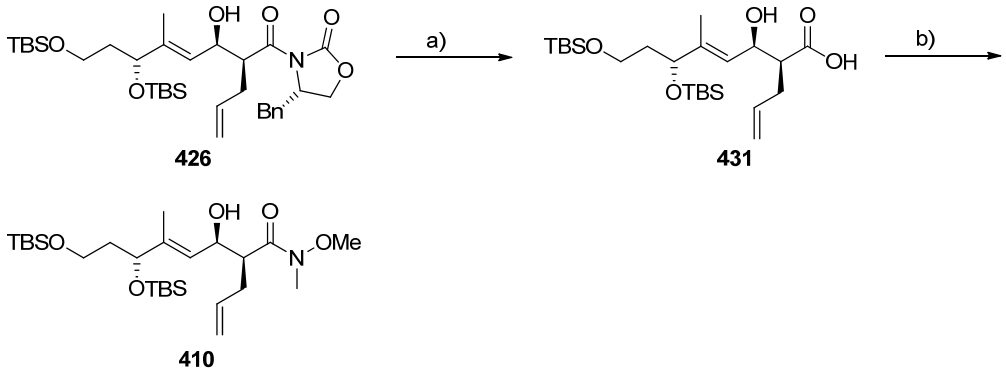
Instead of installing the *Weinreb* amide it was attempted to produce the ester by treating the aldol product **426** with sodium methoxide (Scheme 106).



Scheme 106: NaOMe, MeOH, 0 °C, 29%.

However, as observed for the transamidation reaction the desired product was formed in low yields only and two side products were isolated that originated from the addition of the methoxide ion to the carbonyl group of the oxazolidinone ring.

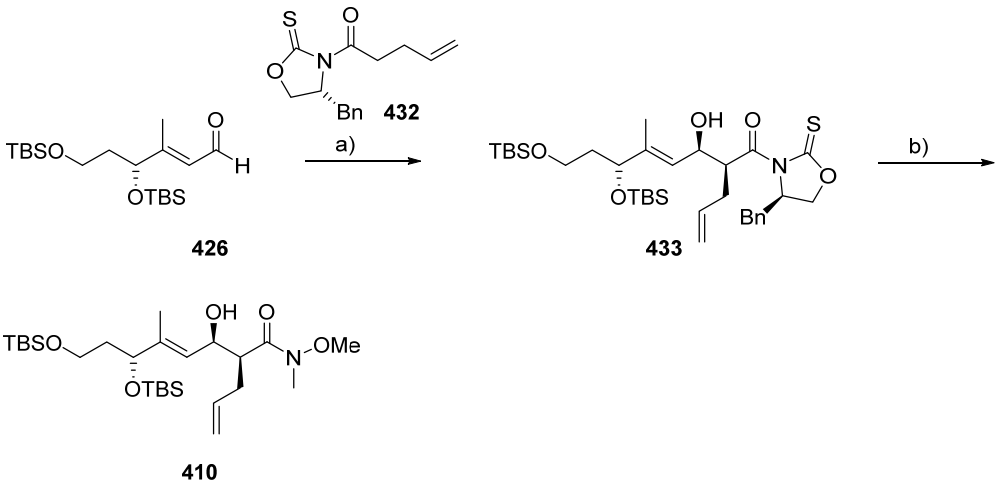
Another possibility to attain the *Weinreb* amide **426** was *via* the corresponding acid **431** (Scheme 107). Albeit not very elegant, this route was hoped to provide enough material so that the subsequent steps could be elaborated.



Scheme 107: a) LiOH, H_2O_2, THF/H_2O, 47%; b) Isobutyl chloroformate, DIPEA, CH_2Cl_2, 0 °C, then Me(OMe)NH·HCl, 16%.

Treatment of **426** with LiOH in the presence of H_2O_2 resulted in the formation of acid **431** in moderate yield, but the subsequent condensation with the *Weinreb* amine gave the *Weinreb* amide **410** only in a very low yield. With the EDCI protocol **410** yields were even lower.

As **426** could not be converted into the desired *Weinreb* amide in a satisfactory manner, the *Evans* auxiliary was exchanged for the *Crimmins*-auxiliary,[12] which has been reported to be cleaved more readily.[241] While the corresponding aldol reaction gave the desired product **433** in good yield, the selectivity of the reaction, unfortunately, was poor and the conversion of **433** into *Weinreb* amide **410** did not work any better than for **426** (Scheme 108).



Scheme 108: a) **432**, $TiCl_4$ (2 eq.), DIPEA (1 eq.), CH_2Cl_2, -78 °C, 80%, dr 2.5:1; b) Me(OMe)NH·HCl, imidazole, CH_2Cl_2, 30%.

The *Evans* auxiliary derivate **425** assumes a conformation that minimizes its dipole moment when exposed to Bu_2BOTf and DIPEA (Figure 49, **A**).[242] Considerations of dipole moment are less relevant for the *Crimmins* auxiliary derivate **432,** which bears a poorly polarizable thiocarbonyl group on the ring.[241] Nevertheless, the deprotonated acyl residue of **432** will orientate in the same way as in **A**, as long as an excess (2.5 eq.) of the amine base is used relative to the *Lewis* acid $TiCl_4$. The second equivalent of the base will coordinate to the metal center, thus preventing coordination of the thiocarbonyl to the metal. The facial selectivity is reversed if the *Lewis* acid is used in excess (2 eq.) relative to the amine base.[241] The enolate now prefers to chelate with the metal *via* the thiocarbonyl group (Figure 49, **B**) which leads to the formation of the non-*Evans syn* aldol adduct. [241]

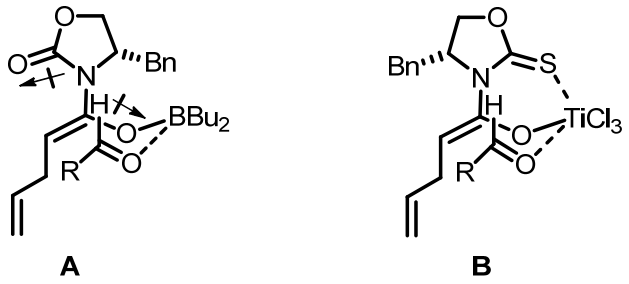
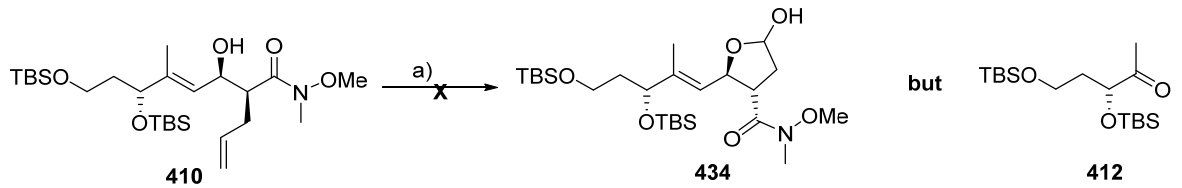


Figure 49: *Evans* enolate (**A**) and *Crimmins* enolate (**B**).

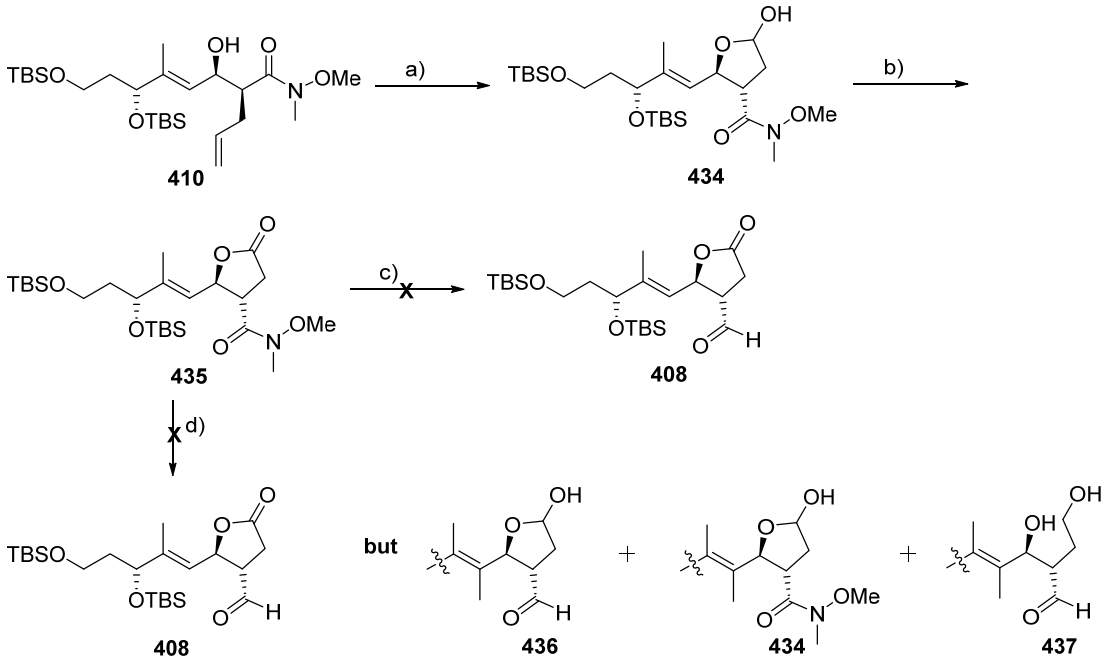
[12] **432** was prepared analogous to the *Evans*-auxiliary derivate **425**.

Regardless of not having found a reaction sequence that would allow access to the *Weinreb* amide **410** in reasonable yield, sufficient amounts of this intermediate were obtained to carry out the subsequent steps (Scheme 109). At first, the terminal double bond was to be transformed into the corresponding aldehyde, which was expected to trigger the formation of lactol **434**. Not unexpected, attempts to achieve this transformation by ozonolysis failed; instead, methyl ketone **412** was produced by preferential cleavage of the internal double bond (Scheme 109).



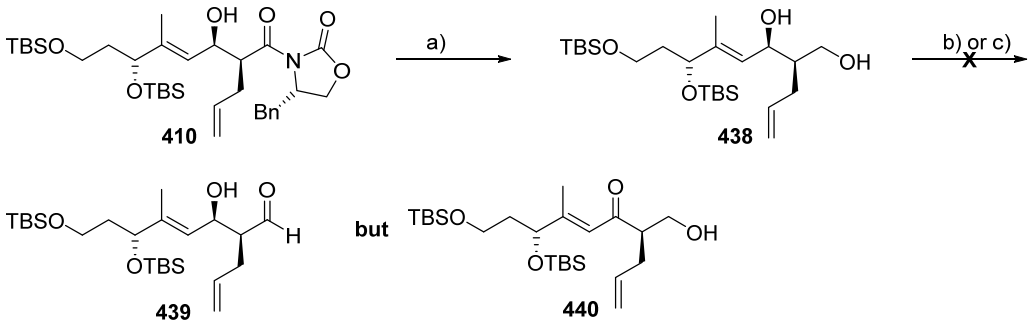
Scheme 109: a) O_3, Me_2S, CH_2Cl_2, MeOH, no product formed.

Fortunately, the use of OsO_4 and $NaIO_4$ yielded the desired lactol **434** in 84% yield (Scheme 110). Subsequent oxidation of **434** with PDC furnished lactone **435** very efficiently. Reduction of the *Weinreb* amide to the aldehyde **408** in the presence of the lactone moiety was first attempted by treatment of **408** with LAH, but this only led to decomposition.[243] DIBAL-H reduction resulted in the formation of three products, lactol aldehyde **436**, lactol *Weinreb* amide **434**, and diol aldehyde **437** (Scheme 110), thus, indicating that *Weinreb* amide **435** could not be reduced selectively.



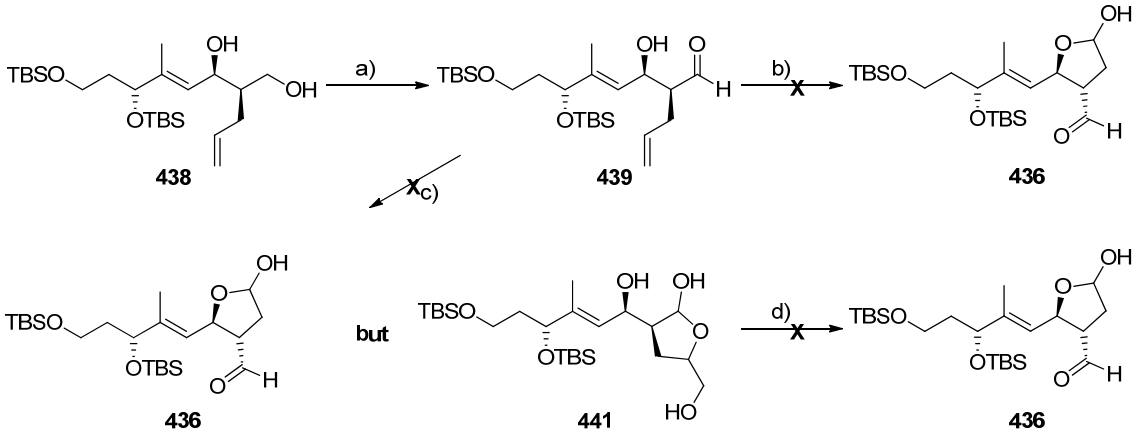
Scheme 110: a) OsO_4, 2,6-lutidine, 1,4-dioxane, H_2O, then $NaIO_4$, 84%; b) PDC, CH_2Cl_2, 92%; c) LAH, Et_2O, -45 °C, decomposition; d) DIBAL-H, THF, -78 °C, side products formed only.

Based on this finding the strategy had to be adjusted slightly. The idea was to install the aldehyde prior to the formation of the lactone ring, in order to circumvent the selectivity issue in the reduction step. Therefore, the auxiliary was reductively cleaved to provide primary alcohol **438** in 73% yield. However, attempted oxidation of **438** either with PDC or DMP[117] afforded exclusively the α,β-unsaturated ketone **440** (Scheme 111).



Scheme 111: a) $LiBH_4$, MeOH, THF, 0 °C, 73%; b) PDC, CH_2Cl_2, only enone formed; c) DMP, CH_2Cl_2, only enone formed.

The allylic secondary hydroxy group thus proved to be more reactive than the (non-allylic) primary one. It was reasoned that the use of a sterically hindered oxidizing agent like TEMPO[145],[244] would shift the preference for oxidation to the primary hydroxy site. This was true to a large extent and allowed the preparation of aldehyde **439** in 73% yield (Scheme 112). In order to transform **439** into lactol **436**, dichloromethane saturated with ozone was slowly added to **439** at low temperature, but as for **410** the internal double bond was cleaved more readily than the terminal one. Unfortunately, the route *via* the diol, obtained with $OsO_4/NaIO_4$, led to the exclusive formation of the undesired lactol **441**, which could not be reacted further to the targeted lactol **436**.



Scheme 112: a) BAIB, TEMPO, CH_2Cl_2, 73%; b) O_3, SMe_2, CH_2Cl_2, -78 °C, cleavage of internal double bond; c) OsO_4, 2,6-lutidine, $NaIO_4$, 1,4-dioxane/H_2O, lactol **441** formed; d) $Pb(OAc)_4$, toluene, no product formed.

To overcome the problem that the diol formed from the terminal double bond reacts instantly with the aldehyde by forming the undesired lactol **441**, the primary hydroxy group in **438** was protected and oxidization to the aldehyde was postponed to a later stage. A TMS or a TES protection group were chosen, since these would allow the deprotection and oxidation to the aldehyde in one pot. TMS protection was found to be difficult because of the enhanced reactivity of the allylic secondary hydroxy group. Treatment of the diol **438** with TMSOTf gave the desired mono-protected isomer **442** in only 25% yield, with substantial amounts of the doubly protected product being formed. Neither the use of the less reactive TMSCl nor the addition of a sterically hindered base like 2,6-lutidine gave a reasonable yield of **442**. In contrast, TES protection to **443** proceeded in acceptable yield (Table 13).

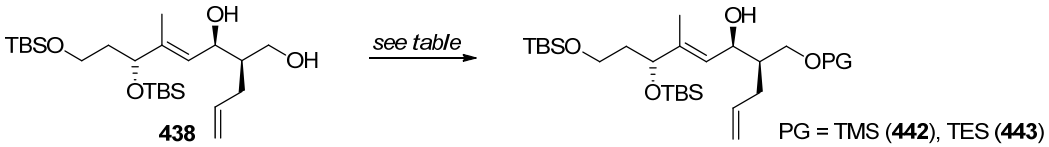
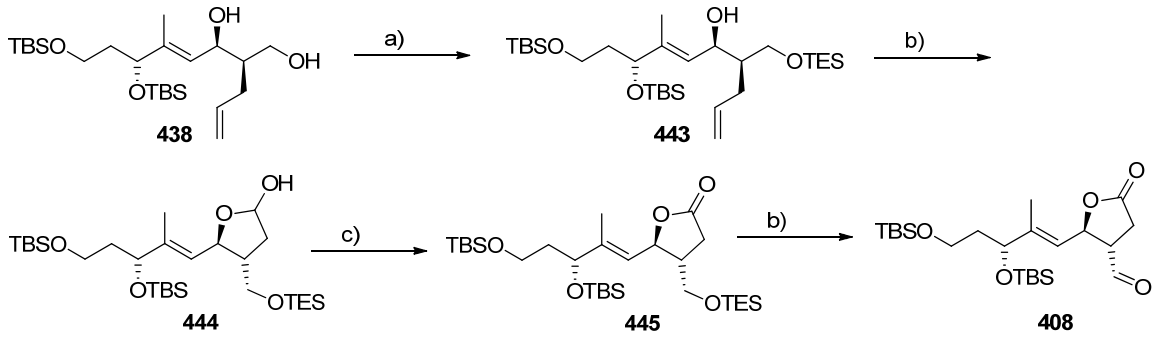


Table 13: TMS or TES protection of primary alcohol **438**.

Entry	Reagent	Base	Temperature	Yield
1	TMSOTf	NEt_3	-78 °C	25%
2	TMSOTf	2,6-lutidine	-78°C	30%
3	TMSCl	imidazole	rt	35%
4	TESCl	imidazole	-25 °C	56%

The terminal double bond was then cleaved with $OsO_4/NaIO_4$ which provided lactol **444** in a moderate yield of 54%, due to limited stability of the TES group under the dihydroxylation/diol cleavage conditions. Oxidation of the lactol **444** to the lactone **445** worked only in poor yield, in spite of the fact that the reaction looked very clean on TLC (Scheme 113). Probably some of the lactone **445** was lost during purification on silica gel. Likewise the subsequent one-pot deprotection/oxidation sequence gave aldehyde **408** only in 33% yield.

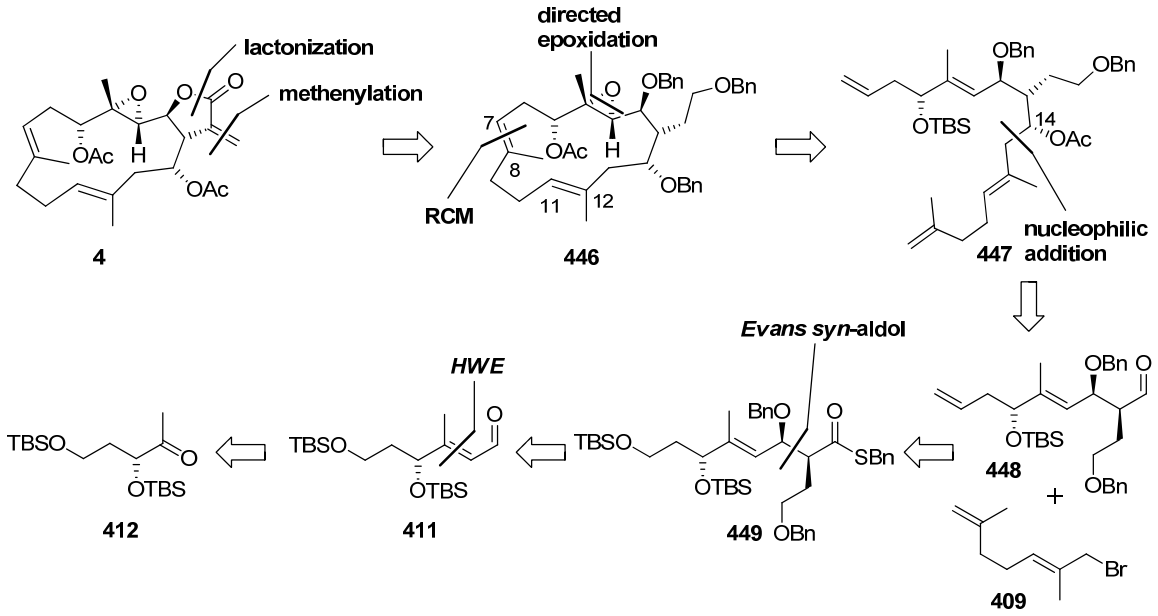


Scheme 113: a) TESCl, imidazole, CH_2Cl_2, 25 °C, 56%; b) OsO_4, $NaIO_4$, 1,4-dioxane/H_2O, 54%; c) PDC, CH_2Cl_2, 33%; d) $(COCl)_2$, DMSO, NEt_3, CH_2Cl_2, -78 °C to rt, 33%.

In principle, attempts could have been made to optimize this first generation route. However, given the fact that the sequence at this point had included several low yielding steps and in light of indications for the limited stability of lactone **445** on silica gel, a change in the overall strategy appeared to be more sensible and promising.

2.3.2 RCM at C7/C8; Late Stage Introduction of Lactone

The revised retrosynthesis of michaolide E (**4**) (Scheme 114) was based on the same disconnections as the original approach except that the lactone ring would be introduced at the very end of the synthesis. The ring was also planned to be closed by a RCM between C7/C8 and the epoxide was to be introduced by a directed epoxidation. The diene **447** was envisioned to be obtained by nucleophilic addition of metalated allylic bromide **409** to aldehyde **448**, which should be accessible from thioester **449**. Thioester **449** would be derived from methyl aldehyde **411** via *Evans syn*-aldol addition, while aldehyde **411** would be obtained from methyl ketone **412** by means of *HWE* olefination. It should be noted that if the nucleophilic addition of the transmetalated allylic bromide **409** would not work reasonably, a methallyl addition to the corresponding aldehyde **448** was planned, which would entail that the ring is closed between C11/C12 rather than C7/C8.



Scheme 114: Revised retrosynthetic analysis of Michaolide E (**4**).

As a consequence, the nucleophilic addition that would establish the chiral center at the future C14 of michaolide E (**4**) would not be carried out in the presence of the lactone ring (Figure 50, left: **A**), but rather on an acyclic substrate **448**. To achieve this, the secondary hydroxy group in β-position to the aldehyde functionality was to be protected with an ether protecting group that would allow β-chelation, in order to direct the nucleophile attack (Figure 50, right: **B**). The group of choice was the benzyl group, which was assumed to be cleavable in the presence of the internal double bonds at a later stage of the synthesis either by catalytic hydrogenation[245] or by means of a *Lewis* acid like BCl_3.

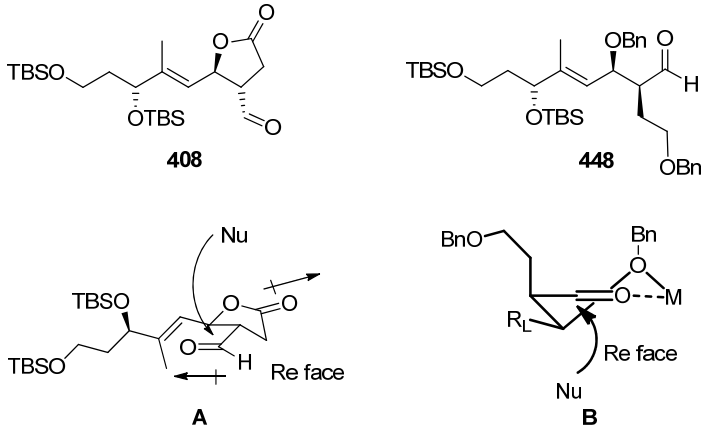
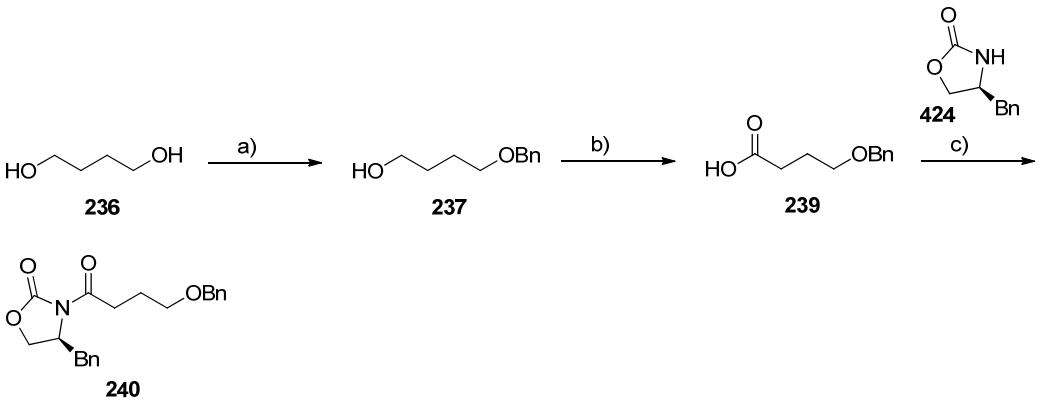


Figure 50: Proposed preferred conformations of **408** and **448** and trajectoris for nucleophilic attack on the aldehyde group. **A**: The aldehyde group orientates in a way as to minimize the overall dipole moment of **A**. **B**: β-Chelation model.

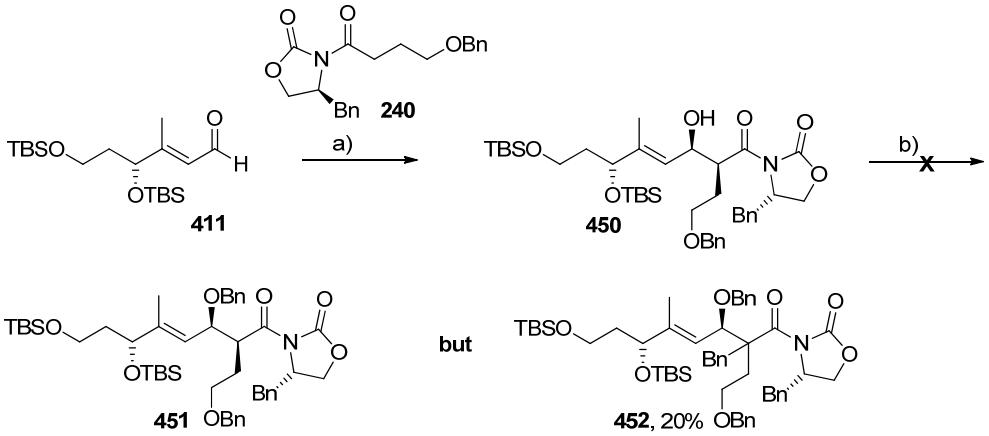
2.3.2.1 Forward Synthesis

In order to implement the small changes in synthetic strategy the acyl oxazolidinone **240** was synthesized starting from 1,4-butanediol (**236**) as described in section 1.5.2.3.[141],[246]



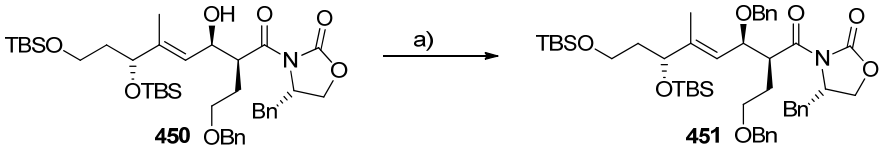
Scheme 115: a) BnBr, NaH, TBAI, THF, 96%; b) CrO_3, H_2SO_4, acetone, 93%; c) EtOCOCl, NEt_3, Et_2O, 0 °C, then **424**, *n*-BuLi, THF, -78 °C to 0 °C, 85%.

Addition of aldehyde **411** to **240** in the presence of Bu_2BOTf and DIPEA gave the *syn*-aldol product **450** in good yield as the only isomer (Scheme 116). Attempts to protect the newly formed secondary hydroxy group as a benzyl ether under basic conditions only gave the α-benzylated amide **452** in low yield (20%). In addition, TLC showed about a 1:1 mixture of starting material and a new product of the same mass shortly after the addition of sodium hydride. Most likely, this finding reflects epimerization of the starting material, due to preferential enolate formation rather than deprotonation of the secondary hydroxy group.



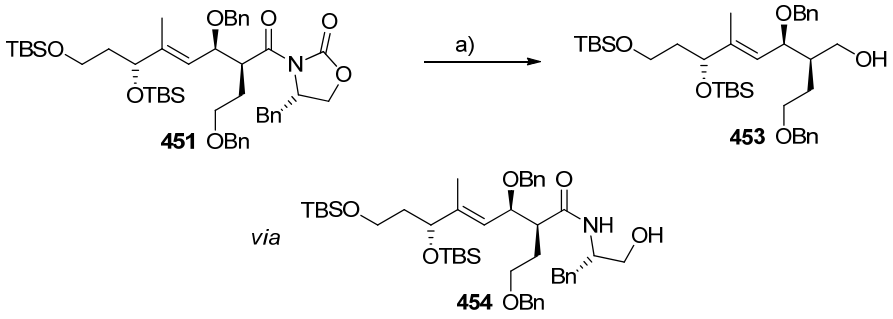
Scheme 116: a) **240**, Bu_2BOTf, DIPEA, CH_2Cl_2, -78 °C, 76%, only isomer; b) NaH, TBAI, BnBr, THF, no product formed.

Thus, benzyl protection was attempted under acidic conditions with benzyl trichloroacetimidate (Bn imidate) (Scheme 117),[247] which gave **451** in 59% yield. In order for the reaction of be successful, the triflic acid had to be added slowly over a period of hours to avoid elimination.



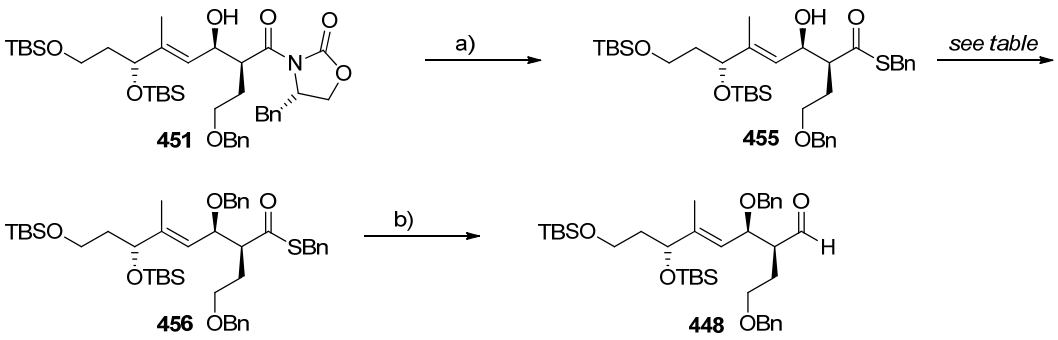
Scheme 117: a) Benzyl 2,2,2-trichloroacetimidate, CF_3SO_3H, cyclohexane/CH_2Cl_2, 59%.

Surprisingly, and in contrast to imide **410** (Section 2.3.1.1, Scheme 111), the subsequent reductive removal of the auxiliary in **452** was very sluggish, requiring up to 50 eq. of $LiBH_4$ to get full conversion and affording alcohol **453** only in moderate yield (40%). One possible reason for the different behavior of **452** compared to similar substrates investigated in this thesis (Scheme 118) could have been that the carbonyl group of the oxazolidinone ring was reduced first and that the reduction of the resulting amide to the desired alcohol required a large excess of $LiBH_4$. If so, lithium aluminium hydride was considered as the better reducing agent for the conversion of **451** into **453**, but the reaction only led to decomposition.



Scheme 118: a) $LiBH_4$, MeOH, THF, 0 °C, 40%.

Due to the fact that the auxiliary could only be removed in low yields, there was a need for an alternative route to produce aldehyde **448** and a plausible approach was the use of thioester **455** as an intermediate (Scheme 119). After some optimization the installation of the thioester moiety could be accomplished in reasonable yield (70%), but the subsequent benzylation of the secondary hydroxy group suffered from low yields (Table 14).

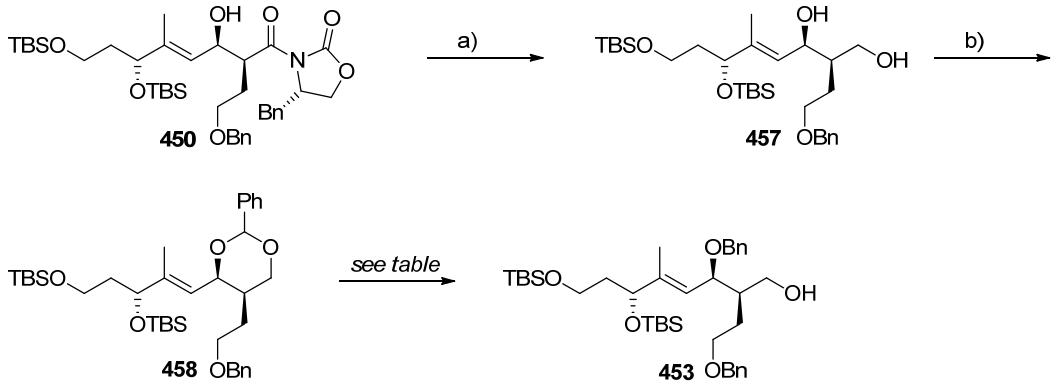


Scheme 119: a) *n*-BuLi, BnSH, $AlMe_3$, CH_2Cl_2/THF, 70%; b) DIBAL-H, CH_2Cl_2, -78 °C, 75%.

Table 14: Benzyl protection of secondary alcohol **455**.

Entry	Reagent	Activation	Solvent	Temperature	Yield
1	Bn trichloroacetimidate	CF_3SO_3H (slowly)	cyclohexane	rt	30%
2	BnBr	Ag_2O	CH_2Cl_2	50 °C	no conversion
3	Bn trichloroacetimidate	$Sc(OTf)_3$	toluene	rt to reflux	no conversion

The best results were obtained by using benzyl 2,2,2-trichloroacetimidate and CF_3SO_3H, but even if the addition of the acid was carried out over a very long time period, the elimination could not be surpressed completely. The reduction of thioester **456** to the aldehyde **448** proceeded in good yield (75%) and without any detectable overreduction to the alcohol.
However, because of the low yield in the benzylation step an alternative route to aldehyde **448** was investigated that involved direct reduction of amide **450** to diol **457** and its subsequent protection as a benzylidene acetal **458**. The latter was obtained in 53% overall yield from aldol product **450** (Scheme 120).



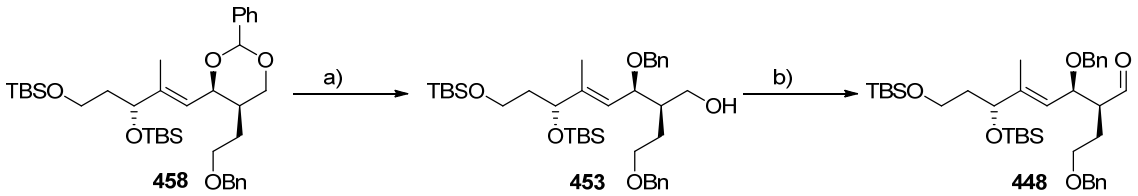
Scheme 120: a) $LiBH_4$, MeOH, THF, 0 °C, 75%; b) Benzaldehyde dimethyl acetal, CSA, CH_2Cl_2, 70%.

The subsequent conversion of benzylidine acetal **458** into the desired free primary alcohol **453** required substantial optimization. Only the use of DIBAL-H allowed the isolation of the desired primary alcohol **453,** while all other conditions applied either led to decomposition or did not yield any conversion (Table 15). Without addition of a *Lewis* acid, DIBAL-H did not give full conversion, even if a large excess was used and the reaction was carried out at higher temperatures. When DIBAL-H was added to a solution of $AlMe_3$ and **458,** full conversion was observed, but the reaction suffered from low yield, as the *Lewis* acid caused partial cleavage of the primary TBS-ether. If the order of addition was reversed and $AlMe_3$ was slowly added to a solution of DIBAL-H and the benzylidene acetal **458** at -20 °C, the yield could be improved to 60% (Table 15, Entry 9). Varying the temperature in either direction resulted in lower yields. In light of these findings, a weaker *Lewis* acid was sought that would not result in loss of the TBS-group, but was still capable of activating the benzylidene acetal towards reductive opening. Fortunately, Me_2AlCl met these criteria and its use resulted in an increased yield of 76% when added at -20 °C. To the best of your knowledge, these conditions have not been reported in literature to date.

Table 15: Reductive opening of benzylidene acetal **458**.

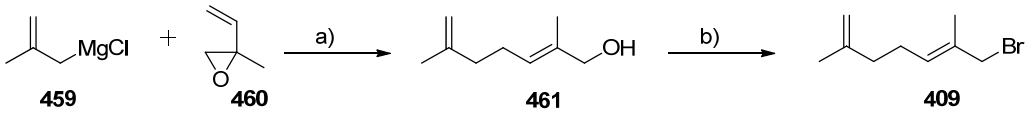
Entry	Reagent	*Lewis* acid	Solvent	Temperature	Yield
1	DIBAL-H		CH_2Cl_2	0 °C to rt	20%
2	Et_3SiH	$PhBCl_2$	DCE	-78 °C	decomposition
3	BH_3	Bu_2BOTf	CH_2Cl_2	-15 to 0 °C	decomposition
4	BH_3	$ZnEt_2$	CH_2Cl_2	0 °C	decomposition
5	LAH	$Sc(OTf)_3$	Et_2O	0 °C	decomposition
6	Red-Al		Et_2O	0 °C to reflux	no reaction
7	$NaBH_3CN$	$TiCl_4$	MeCN	0 °C	primary TBS group lost
8	DIBAL-H	$AlMe_3$	CH_2Cl_2	0 °C	30%
9	DIBAL-H	$AlMe_3$	CH_2Cl_2	-20 to 0 °C	60%
10	DIBAL-H	Me_2AlCl	CH_2Cl_2	-20 °C	76%

Having successfully implemented the reductive opening of the benzylidene acetal **458**, the resulting primary alcohol was oxidized to the aldehyde **448** using TEMPO (Scheme 121). *Swern* conditions only led to decomposition. With aldehyde **448** in hand, the stage was then set for the nucleophilic addition reaction.



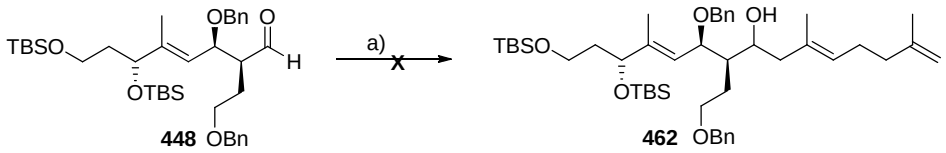
Scheme 121: a) DIBAL-H, Me_2AlCl, CH_2Cl_2, -20 °C, 76%; b) BAIB, TEMPO, CH_2Cl_2, 0 °C, 85%.

The allylic bromide **462**, which was to be transformed into a nucleophile by halogen-metal exchange, was synthesized according to the literature (Scheme 122).[248]



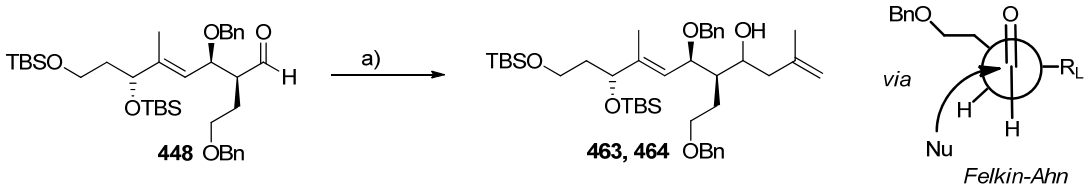
Scheme 122: a) CuI, THF, -30 °C, 81%; b) MsCl, LiBr, THF, 85%.

Initial attempts at the addition of metalated **409** to aldehyde **448** were carried out in the non-coordinating solvent dichloromethane and with Me_2AlCl as the *Lewis* acid, in order to enable β-chelation. Unfortunately, the desired product was not observed under these conditions (Scheme 123).



Scheme 123: a) Me_2AlCl, **409**, Zn, CH_2Cl_2, then **448**, no product formed.

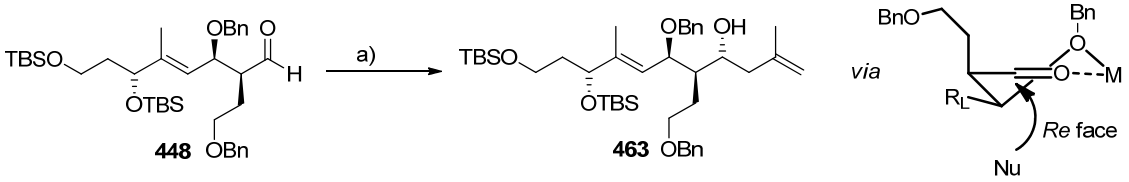
In the following, the reactivity of aldehyde **448** towards nucleophiles was elaborated by the addition of methallyl magnesium chloride. This reaction produced two diastereomers **463** and **464** in a 2:1 ratio in 88% total yield; it could be assumed at the time that the *Felkin-Ahn* product **464** was favoured (Scheme 124).[13]



Scheme 124: a) methallyl magnesium bromide, THF, -78 °C, 88%, dr 1:2.

[13] The anticipated result was later proven by the comparison with the outcome of the *Sakuari* addition, which was favouring the other diastereomer as was confirmed by *Mosher* ester analysis.

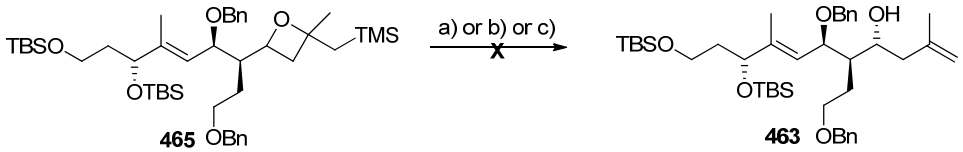
In order to promote a β-chelation path over *Felkin-Ahn* control, the reaction had to be run in a non-coordinating solvent and a suitable combination of a methallyl nucleophile as well as a *Lewis* acid had to be found. The use of methallyl trimethyl silane as a nucleophile was investigated. In the presence of $MeAlCl_2$ the desired secondary alcohol **463** was obtained in 40% yield as a 9:1 diastereomeric mixture in favour of the diastereomer that had been the minor product in the addition of methallylmagnesium bromide, thus, indicating that the pathway *via* the β-chelation was operative dominantly (Scheme 125).



Scheme 125: a) $MeAlCl_2$, methallyl trimethyl silane, CH_2Cl_2, -78 °C, 40%, dr 9:1.

Although the yield of the reaction was moderate, this outcome was very encouraging and this approach was considered to be worth further investigation. A successful implementation of the methallyl addition would mean that the ring would be closed between C11/C12 rather than at C7/C8.

In the above reaction, according to MS analysis and NMR data oxetane **465** was formed as a major side product in 50% yield, which surprisingly could not be reacted further to the product (Scheme 126).



Scheme 126: a) HCl, EtOAc; b) citric acid, MeOH; c) TBAF, THF.

In the absence of a *Lewis* acid the side product **465** was formed exclusively; hence, it was reasoned that a stronger *Le*wis acid might favour the formation of the desired product **463** over the side product **465** (Table 4). With $TiCl_4$ the secondary alcohol **463** was obtained in 37% yield and with 15:1 selectivity. The low yield was a consequence of $TiCl_4$-mediated TBS ether cleavage in the starting material **448** as well as the product **463.** When $SnCl_4$ was used as the *Lewis* acid, **463** was produced with excellent selectivity of 20:1, but only in 37% yield, if the methallyl trimethyl silane was added 10 min after the *Lewis* acid. When the time difference between the additions was shortened to 2 min, the yield could be improved to 76% (Table 16). No such improvement was observed with $TiCl_4$. For $SnCl_4$ a further reduction of the time delay or even a reversal of the order of addition resulted in lower yield, because

increasing amounts of the side product **465** were formed. The diastereomeric ratio could be increased up to 25:1 by lowering the temperature to -90 °C.

Table 16: Methallyl addition to aldehyde **448**.

Entry	Reagent	*Lewis* acid	Solvent	Temperature	Yield, dr
1	methallyl magnesium chloride	Me_2AlCl	CH_2Cl_2	-78 °C	88%, 1:2
2	methallyl trimethyl silane	Me_2AlCl	CH_2Cl_2	-78 °C	40%, 9:1
3	methallyl trimethyl silane	$TiCl_4$	CH_2Cl_2	0 °C	45%, 15:1
4	methallyl trimethyl silane	$SnCl_4$ (10 min.)	CH_2Cl_2	-78 °C	37%, 20:1
5	methallyl trimethyl silane	$SnCl_4$ (5 min.)	CH_2Cl_2	-90 °C	70%, 25:1
6	methallyl trimethyl silane	$SnCl_4$ (2 min.)	CH_2Cl_2	-90 °C	76%, 25:1

In order to prove that the addition had indeed delivered the desired isomer **463**, *Mosher* ester analysis was carried out on the addition product (Figure 51).[249] The analysis gave a coherent picture with all values of d_{SR} being larger than 0 for R_1 and being smaller than 0 for R_2. These data clearly confirm the desired stereochemical outcome of the methallyl addition.

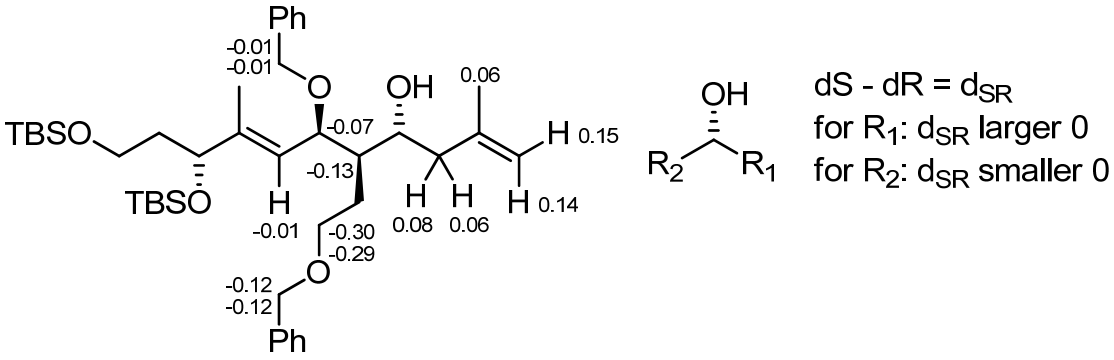
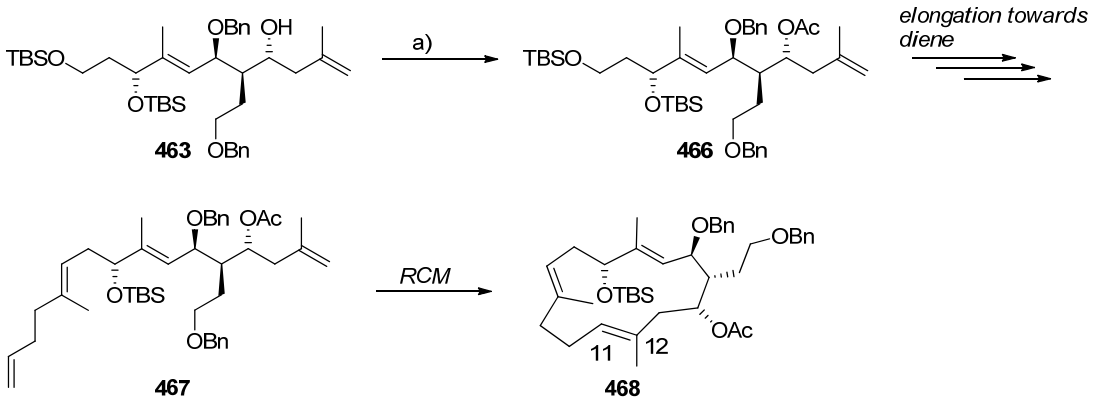


Figure 51: *Mosher* ester analysis of the secondary alcohol **463**.

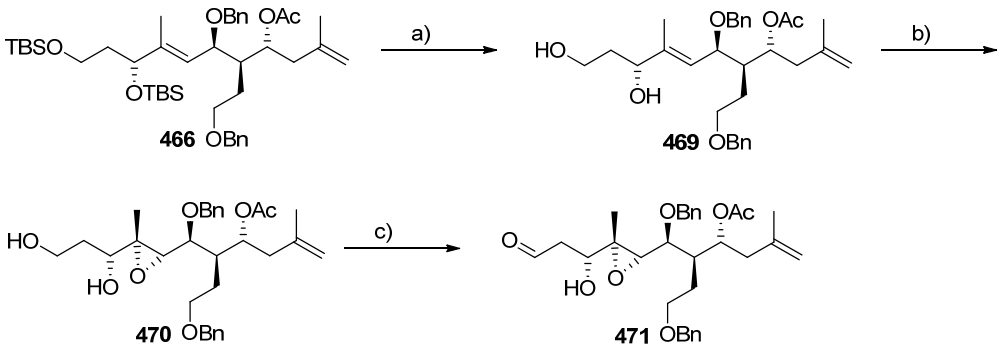
With the successful implementation of the selective methallyl addition to aldehyde **448** the synthetic strategy was modified slightly, such that the projected site of ring closure was shifted from C7/C8 to C11/C12. As a consequence, the nucleophilic addition of metalated allylic bromide **409** to aldehyde **448**, a reaction which was considered to be difficult to establish in any case, was not further pursued.

The modified strategy is depicted in Scheme 127 and following the successful assembly of alcohol **463** was to involve the protection of the newly formed hydroxy group as an acetate, a reaction that proceeded in good yield. It was then planned to cleave the primary TBS-ether and then elaborate the molecule into the required diene **467** for the ring closing metathesis (Scheme 127).



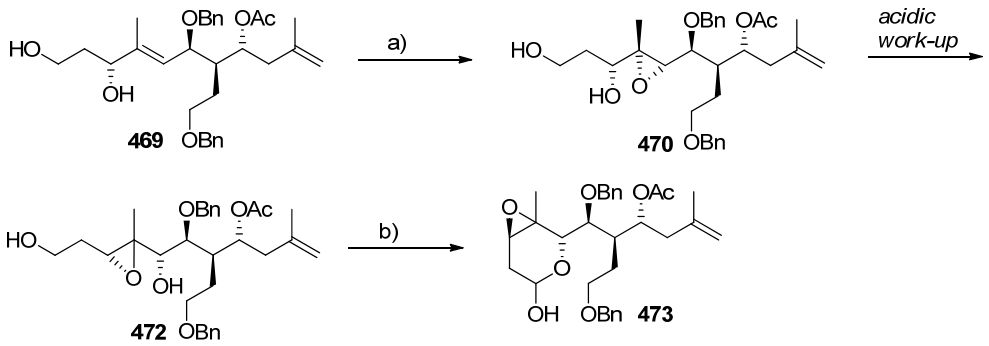
Scheme 127: a) NEt_3, DMAP, Ac_2O, MeCN, 94%.

Somewhat unexpectedly, treatment of **466** either with CSA or TBAF led to the double deprotected product **469** (Scheme 128). However, instead of exploring further conditions, it was felt that the diol **469** could be processed further by directed epoxidation, with the introduction of the epoxide moiety then simply preceding ring closure. Thus, asymmetric *Sharpless* epoxidation of **469** yielded a single isomer of an epoxide in good yield and assuming this material to be epoxide **470** it was reacted with PDC to produce the desired aldehyde **471**. Very surprisingly, however, this oxidation did not give aldehyde **471** (Scheme 128) but yielded exclusively hemiacetal **473** (Scheme 129).



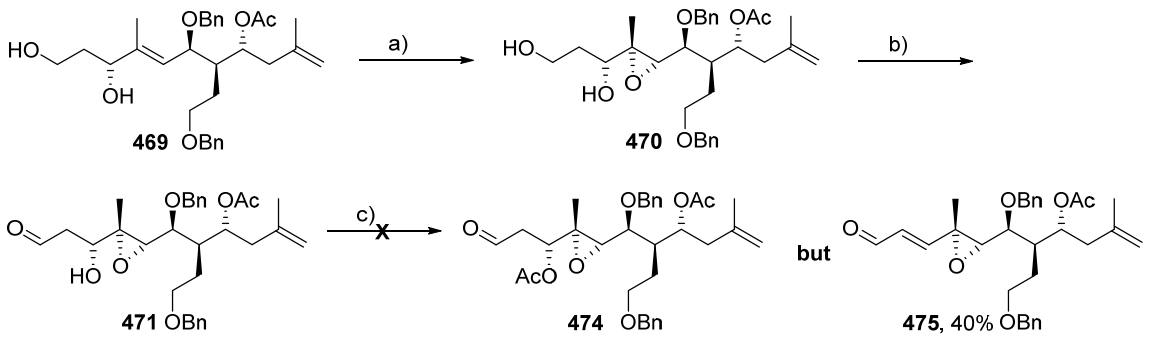
Scheme 128: a) TBAF, THF, 0 °C, 86%; b) (+)-DET, Ti(*i*-OPr)$_4$, *t*-BuOOH, 4A-mol. sieves, CH_2Cl_2, -20 °C; c) PDC, CH_2Cl_2.

This surprising outcome can be explained by a *Payne* rearrangement,[250] which must have occurred during the *Sharpless* epoxidation. The acidic work-up carried out after the actual epoxidation reaction triggered the *Payne* rearrangement, thus resulting in the formation of epoxide **472;** the same product was obtained upon oxidation under unbuffered *m*-CPBA conditions (Scheme 129). PDC oxidation of the primary alcohol then led to the formation of the hemiacetal **473**.



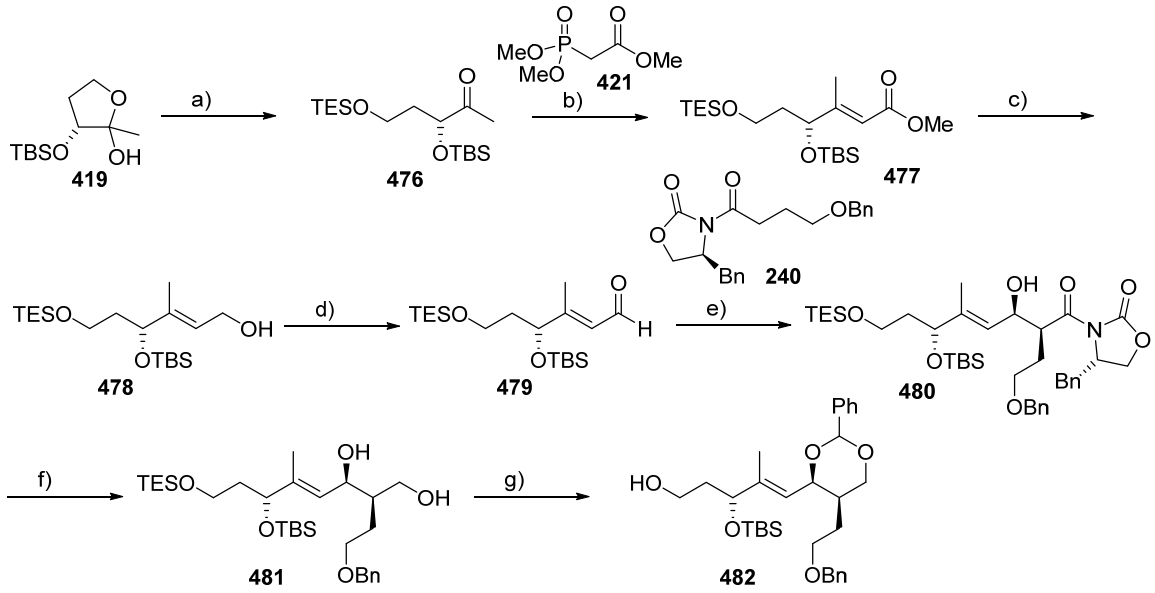
Scheme 129: a) (+)-DET, Ti(*i*-OPr)$_4$, *t*-BuOOH, 4A-mol. sieves, CH_2Cl_2, -20 °C, 73%; c) PDC, CH_2Cl_2, 65%.

Fortunately, the two epoxides **470** and **472** exhibit different R_f values so that the rearrangement could be followed on TLC. When a neutral work-up was applied the rearrangement did not take place and the desired epoxide **470** could be isolated in 65% yield as a single isomer. DMP oxidation[117] of the primary alcohol afforded aldehyde **471** in 60% yield. Using the TEMPO protocol[145] resulted in the same yield. If the DMP oxidation was not buffered with $NaHCO_3$, the major product observed was again hemiacetal **473**. Attempts to protect the secondary hydroxy in aldehyde **471** as an acetate only resulted in the formation of α,β-unsaturated aldehyde **475,** which was obtained in 40% yield (Scheme 130). The acetate was chosen as a protecting group as it is present in the natural product.



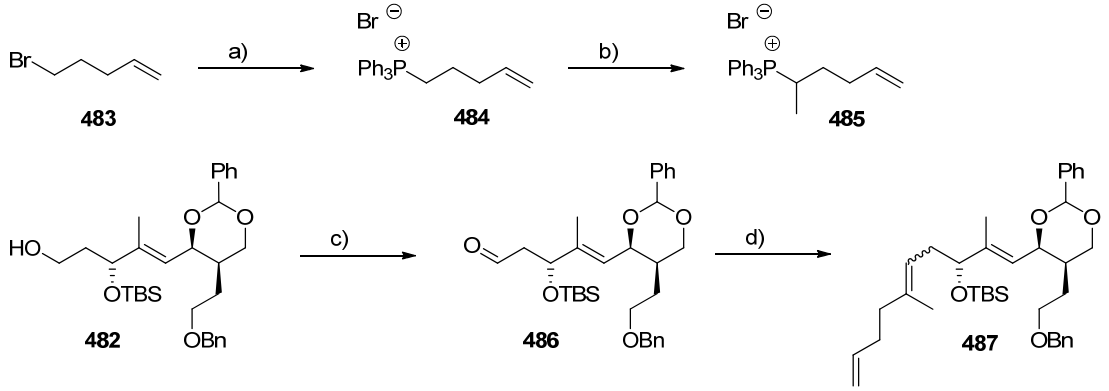
Scheme 130: a) (+)-DET, $Ti(i\text{-}OPr)_4$, t-BuOOH, 4A-mol. sieves, CH_2Cl_2, -20 °C b) DMP, $NaHCO_3$, CH_2Cl_2, 0 °C to rt, 60%; or BAIB, TEMPO, CH_2Cl_2, 0 °C, 60%; c) NEt_3, DMAP, Ac_2O, MeCN, no product isolated.

In light of the problem faced with the acetate protection the original silyl-based protecting group strategy was revised, but with the primary hydroxy group protected as a TES ether, which was expected to be cleavable selectively in the presence of the TBS protected allylic secondary alcohol (Scheme 131).[251] This would also allow the installation of the epoxide moiety at a later stage of the synthesis, as it had been planned originally. The synthesis of the corresponding intermediate **481** paralleled that of the bis-TBS ether **457** (Section 2.3.2.1), but installation of the benzylidene acetal moiety **482** in the presence of 5 mol% CSA was then accompanied by loss of the TES group and the free primary alcohol **482** was isolated in 70% yield (Scheme 131).



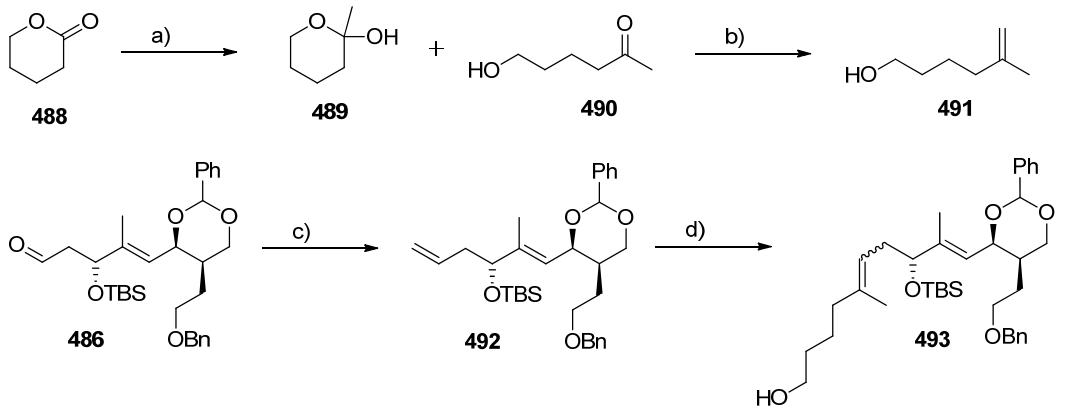
Scheme 131: a) TESCl, imidazole, CH_2Cl_2, 94%; b) NaH, **421**, THF, then **476**, 45 °C, 92%, *E*/*Z* 25:1; c) DIBAL-H, CH_2Cl_2, -78 °C, 92%; d) MnO_2, Et_2O, 90%; e) **240**, Bu_2BOTf, DIPEA, CH_2Cl_2, -78 °C, 76%, dr 10:1; f) $LiBH_4$, MeOH, THF, 0 °C, 70%; g) benzaldehyde dimethyl acetal, CSA, CH_2Cl_2, 70%.

In response to this unexpected development, elaboration of the left hand side of the molecule was pursued before reduction of the acetal moiety. Thus, the primary hydroxy group in **482** was oxidized to the aldehyde **486**. Subsequent reaction with *Wittig* salt **485**, which was synthesized from bromide **483** in a two step sequence, gave trisubstituted olefin **487** in 35% yield as a 1.5:1 mixture of separable double bond isomers (Scheme 132). No effort was made to assign the individual isomers.



Scheme 132: a) PPh_3, MeCN, 81%; b) *n*-BuLi, MeI, THF, 0 °C, 96%; c) DMP, $NaHCO_3$, CH_2Cl_2, 0 °C to rt, 84%; d) **485**, *n*-BuLi, THF, -78 °C, then **486**, 35%, 68% brsm, 1.5:1 mixture of double bond isomers.

Since some starting material was reisolated from the *Wittig* reaction, the yield could likely be improved by using an excess of the ylid derived from **485**, but the low *E*/*Z* selectivity, which was not entirely unexpected,[252] was deemed to be rather difficult to be altered. As an alternative to *Wittig* chemistry, the trisubstituted *trans* double bond was then attempted to be installed by means of cross metathesis (Scheme 133). The disubstituted olefin **491** was synthesized starting from δ-valerolactone **488** by treatment with methyllithium and subsequent *Wittig* olefination.[253] The monosubstituted olefin **492** derived from aldehyde **491** *via* another *Wittig* olefination is thought to be more reactive and the homodimer will be formed initially.[254] By addition of an excess (5 eq.) of the less reactive and synthetically undemanding disubstituted olefin **491** the homodimer was partially cleaved and the formation of the desired heterodimer **493** was observed.[254]



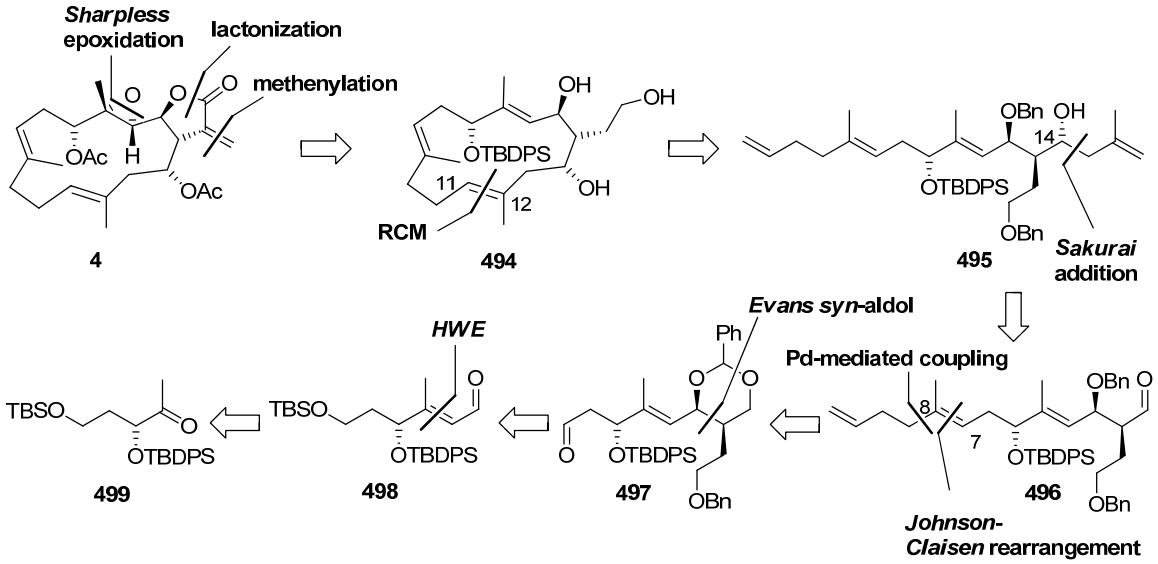
Scheme 133: a) MeLi, Et_2O, 30%; b) methyl triphenyl phosphonium bromide, *n*-BuLi, THF, 82%; c) methyl triphenyl phosphonium bromide, LiHMDS, THF, 88%; d) **491**, *Grubbs* II, CH_2Cl_2, 40 °C, 40%, 1.4:1 mixture of double bond isomers.

The product **493** was isolated in a yield of 40% together with 35% of the homodimer derived form **492**. This implies that the yield could be improved, but unfortunately the reaction suffered from a low selectivity (1.4:1 mixture of double bond isomers), and it was not clear how this problem could possibly be overcome. Again, no effort was made to assign the individual isomers.

At this stage all material was consumed and the question arose whether yet another protecting group strategy should be explored as part of the resynthesis of the intermediates. After carefully weighting different options, a strategy that would continue to rely on silyl ether protecting groups was deemed as most promising, although more stable silyl ether variant than those previously employed were considered necessary. More important perhaps was the need to find an efficient way to install the trisubstituted *trans* double bond at C7/C8 in order to get access to the diene, which would allow the exploration of the crucial macrocyclization by RCM.

2.3.3 RCM at C11/C12, *Johnson-Claisen* Rearrangement

Based on the experimental findings discussed in the preceding sections, the previous retrosyntheses (Section 2.3.1, Scheme 101 and Section 2.3.2, Scheme 114) were modified such that most of the main disconnections would remain the same, except for one major change. In light of the fact that the *Sakurai* addition of trimethyl methallyl silane to aldehyde **448** (Section 2.3.2.1, Table 15) had proven to allow the installation of the C14 chiral center (michaolide numbering) with excellent selectivity, the projected site for RCM-based macrocyclization was shifted to C11/C12 (Scheme 134). In this modified retrosynthesis, the epoxide was to be introduced at the very end by directed epoxidation and the lactone would be also closed in a late stage of the synthesis. As mentioned above, the ring was planned to be closed between C11/C12 by means of RCM and the chiral center was to be installed by addition of trimethyl methallyl silane to aldehyde **496**. The establishment of the trisubstituted *trans* double bond at C7/C8 in **496** was envisioned to be accomplished either by a Pd-mediated cross coupling reaction or by a *Johnson-Claisen* rearrangement of an appropriate precursor. *Evans syn*-aldol addition to aldehyde **498**, which in turn is derived from methyl ketone **499** *via HWE* olefination, would serve to install the two adjacent stereocenters in **497**.

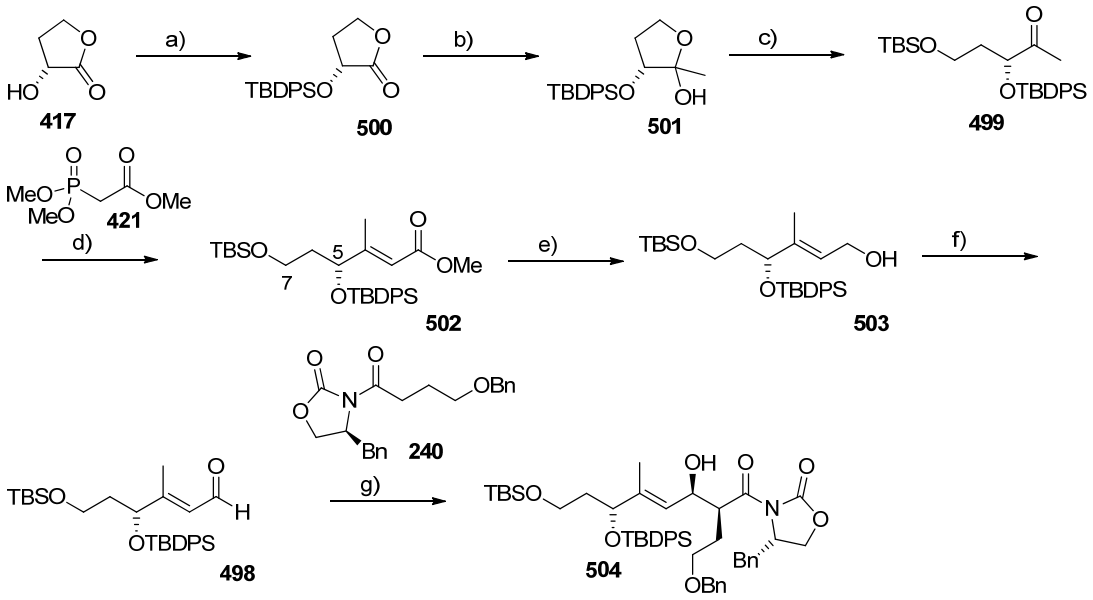


Scheme 134: Retrosynthesis.

2.3.3.1 Forward Synthesis

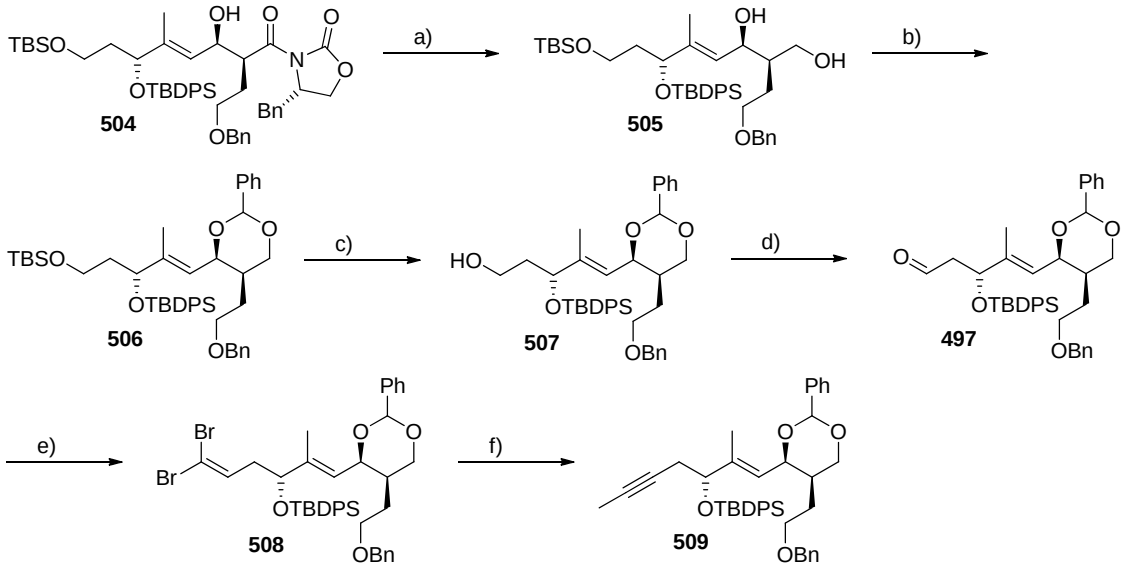
In order to increase protecting group stability, this third generation approach relied on the use of a TBDPS-group for protection of the secondary hydroxy group on C5 and a TBS-group for

the primary one on C7. This approach was expected to allow uncomplicated selective deprotection of the primary OH-group without loss of the protecting group from the C5 alcohol.[251] In addition, both protecting groups should be stable enough to be carried through the synthesis to the point of deliberate cleavage. Starting from α-hydroxy lactone **417** (Section 2.3.1.1, Scheme 103) imide **504** was prepared by the same sequence of reaction as for **450** (Scheme 135). For every single step yields were at least equal to or even higher than those obtained for the corresponding transformations in the synthesis of **450**. Noteworthy, the reaction of aldehyde **498** with imide **240** gave the desired *syn*-aldol product **504** in 90% yield as the only isomer, which was superior to all of the analogous aldol reactions previously described.



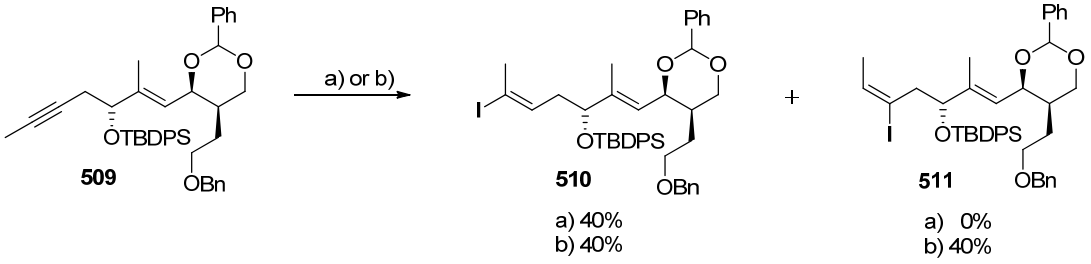
Scheme 135: a) TBDPSCl, imidazole, DMF, 96%; b) MeLi, THF, -78 °C, 94%; c) TBSCl, imidazole, DMF, 96%; d) **421**, NaH, THF, 0 °C to 45 °C, 94%, only isomer; e) DIBAL-H, CH_2Cl_2, -78 °C, 96%; f) MnO_2, Et_2O, 91%; g) **240**, Bu_2BOTf, DIPEA, CH_2Cl_2, -78 °C, 90%, single isomer.

After successful installation of the benzylidene acetal moiety in **506** (Scheme 136), the focus was shifted towards the construction of the trisubstituted *trans* double bond at C7/C8 prior to reduction of the acetal and further elaboration of the right-hand side of this fragment. Acidic removal of the TBS-group gave primary alcohol **507**, which was oxidized with DMP; the resulting aldehyde **497** then underwent *Corey-Fuchs* homologation[255] to afford alkyne **509** in good yield (88 %).



Scheme 136: a) $LiBH_4$, MeOH, THF, 76%; b) benzaldehyde dimethyl acetal, CSA, CH_2Cl_2, 80%; c) PPTS, EtOH, 70%; d) DMP, $NaHCO_3$, CH_2Cl_2, 90%; e) CBr_4, PPh_3, CH_2Cl_2, 79%; f) *n*-BuLi, MeI, THF, -78 °C, 88%.

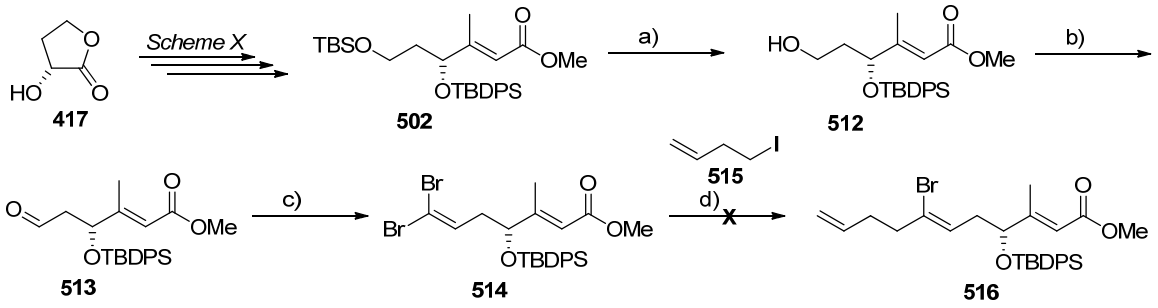
Attempted hydrozirconation-iodination of **509** using the *Schwartz* reagent[256] did not yield the desired vinyl iodide **510** in a satisfactory way (Scheme 137). When carried out in THF, the vinyl iodide **510** could be isolated in 40% along with an unkown inseparable impurity[14] (no formation of the regioisomer **511** was observed), while the use of benzene as the solvent gave an inseparable 1/1 mixture of regioisomers **510** and **511** in 80% yield. It has been reported that isomerization to the more stable, less sterically crowded alkenylzirconium species may be effected in the presence of an excess of *Schwartz* reagent at elevated temperatures.[257],[258] However, neither varying the equivalents of the reagents employed nor conducting the reaction at different temperatures led to any improvement. The significantly different outcome of the reaction depending on the solvent employed is remarkable, although the reason for this remains unclear. On the other hand, the formation of the undesired vinyl iodide **511** in benzene is not a surprise, since α-unbranched alkynes often suffer from low regioselectivity.[259]



Scheme 137: a) Cp_2ZrHCl, then I_2, THF, 50 °C, 40%; b) Cp_2ZrHCl, then I_2, benzene, 50 °C, 40%.

[14] The use of Cp_2ZrCl_2 and DIBAL-H to generate the *Schwartz* reagent *in situ* led to the same result.[108]

Because of the deficiencies of the hydrozirconation-iodination reaction, it was tried to alkylate the dibromide **514** sequentially by two subsequent Pd-mediated couplings.[260],[261] This viability of this strategy was explored with the simpler model substrate **514**, as none of the aldehyde **497** was available anymore at this point (Scheme 138). The synthesis of **514** made use of the previously described intermediate **502**. Selective deprotection of the primary alcohol afforded **512**, which was subsequently oxidized under DMP conditions. The resulting aldehyde **513** then underwent *Corey-Fuchs* homologation in 95% yield to give dibromide **514**.

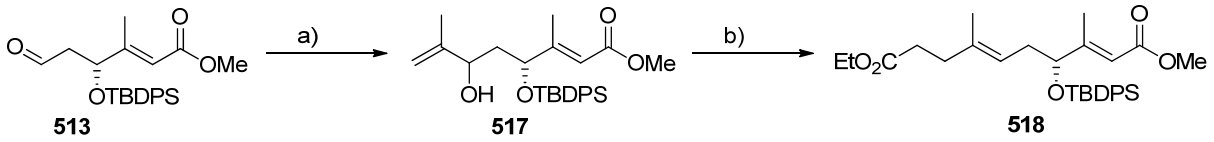


Scheme 138: a) AcOH, THF, H_2O, 96%; b) DMP, $NaHCO_3$, CH_2Cl_2, 92%; c) CBr_4, PPh_3, CH_2Cl_2, 95%; d) **514**, $Pd(PPh_3)_4$, ethanedibromide, THF, **515**, $ZnCl_2$, Mg, I_2, THF, no product formed.

The attempted *Negishi* type couplings[262] of **514** with **515** did not afford any off the desired product **516** as most of the starting material remained unaffected.

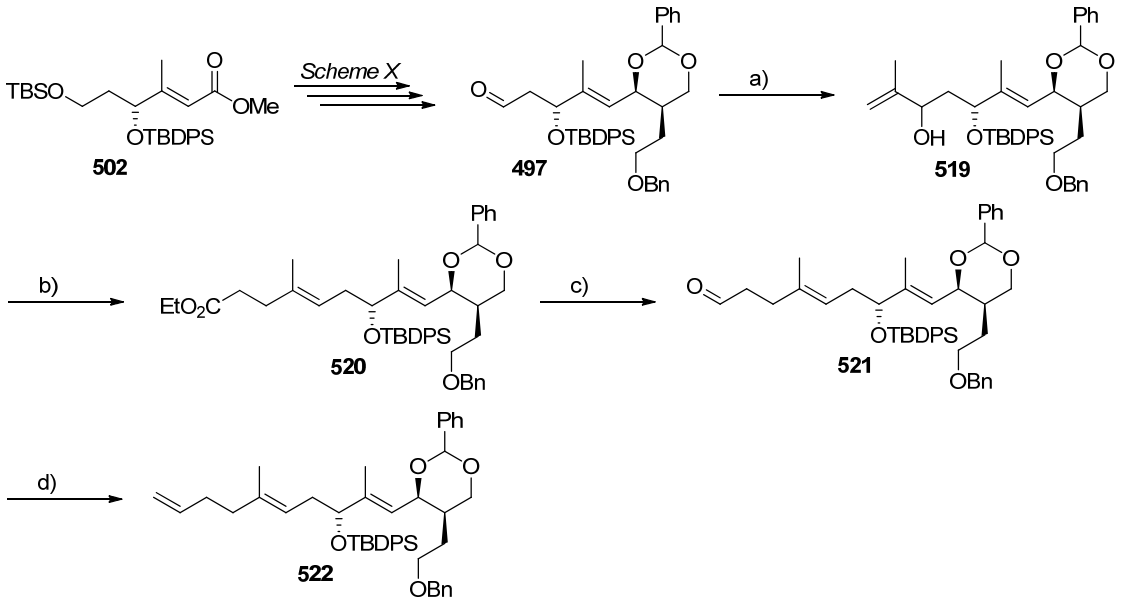
In light of the difficulties met in the iodination of alkyne **509** as well as in the model reaction for a *Negishi* type coupling, the cross-coupling approach for the installation of the C7/C8 double bond was abandoned (at least temporarily at that time). Instead, the *Johnson-Claisen* rearrangement[263] was explored as an alternative, an approach that has already included in the synthetic planning for the third generation approach towards michaolide E (**4**) (Section 2.3.3, Scheme 134)

Because workable amounts of aldehyde **513** were still available when the decision was made to explore this approach also experimentally, the *Johnson-Claisen* rearrangement was attempted on a model substrate derived from this aldehyde **513**. Thus, **513** was converted into secondary alcohol **517** (Scheme 139), which upon heating with triethyl orthoacetate in the presence of catalytic amounts of propionic acid underwent smooth *Johnson-Claisen* rearrangement to deliver ester **518** in 92% yield



Scheme 139: a) Isopropenyl magnesium bromide, THF, -78 °C, 91%; $MeC(OEt)_3$, propionic acid, toluene, reflux, 92%.

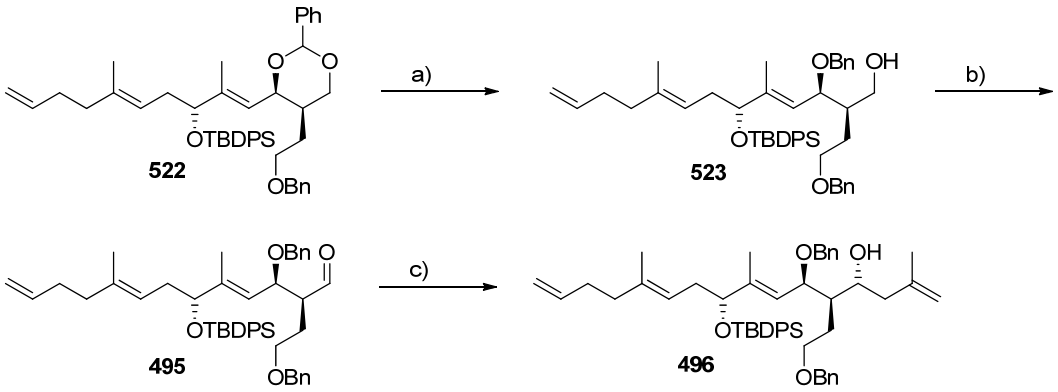
The successful rearrangement of the model substrate led us follow this strategy for the incorporation of the C7/C8 double bond in michaolide E (**4**). Thus, ester **502** (Scheme 140) was elaborated into aldehyde **497** as described in Scheme 135; the latter then underwent *Grignard* addition with isopropenyl magnesium bromide to give a mixture of secondary alcohols **519** in an acceptable yield of 76%. Treatment of **519** with triethyl orthoacetate in the presence of catalytic amounts of propionic acid triggered the *Johnson-Claisen* rearrangement to give ester **520** in 70% yield. Subsequent reduction of the ester moiety with one equivalent of DIBAL-H afforded aldehyde **521** in 81% yield. Little overreduction to the alcohol was observed, which could be isolated and oxidized to the aldehyde **521** under *Swern* conditions in 95% yield. Aldehyde **521** then underwent *Wittig* homologation to furnish terminal olefin **522**.



Scheme 140: a) Isopropenyl magnesium bromide, THF, 76%; b) $MeC(OEt)_3$, propionic acid, toluene, reflux, 70%; c) DIBAL-H, CH_2Cl_2, -78 °C, 81%; d) Methyl triphenylphosphonium bromide, LiHMDS, THF, 75%.

With one of the double bonds required for RCM-based ring-closure installed, what needed to be done next was the proper elaboration of the other end of the molecule. The reductive opening of the benzylidene acetal **522** under the conditions that had been established earlier

yielded primary alcohol **523** in an excellent yield of 92%, which reflects the enhanced stability of the silyl ether protecting groups over those employed in the earlier approaches. Subsequent TEMPO oxidation gave aldehyde **495** (Scheme 141).



Scheme 141: a) DIBAL-H, $AlMe_2Cl$, CH_2Cl_2, 92%; b) BAIB, TEMPO, CH_2Cl_2, 0 °C, 85%; c) methallyl trimethyl silane, $SnCl_4$, CH_2Cl_2, -90 °C, 92%, dr 25:1.

Sakurai addition of methallyl trimethyl silane to aldehyde **495** yielded secondary alcohol **496** in very good yield (92%) if the silane was added 4 minutes after the *Lewis* acid $SnCl_4$. If the silane was added 2 or 6 minutes after the $SnCl_4$, the yield dropped to 50-60%. This narrow time range could be an issue if the reaction was to be carried out on larger scale, but did not pose a problem in the context of this PhD work. Having the diene **496** in hand, the stage was now set for the ring-closing metathesis. Several conditions were tested (Table 17).

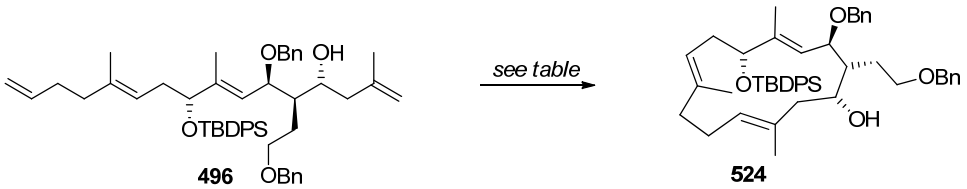


Table 17: Different conditions tested for the RCM of diene **496**.

Entry	Catalyst	Solvent	Temperature	Yield
1	*Grubbs* II	CH_2Cl_2	reflux	little product formation
2	*Grubbs* II	benzene	65 °C	52%
3	*Hoveyda-Grubbs* II	CH_2Cl_2	reflux	little product formation
4	*Hoveyda-Grubbs* II	benzene	65 °C	60%
5	*Piers* (**525**)	CH_2Cl_2	reflux	little product formation
6	Bromo-pyridine (**526**)	CH_2Cl_2	reflux	little product formation

In Figure 52 the not very often used RCM catalysts *Piers* and bromo-pyridine are shown.

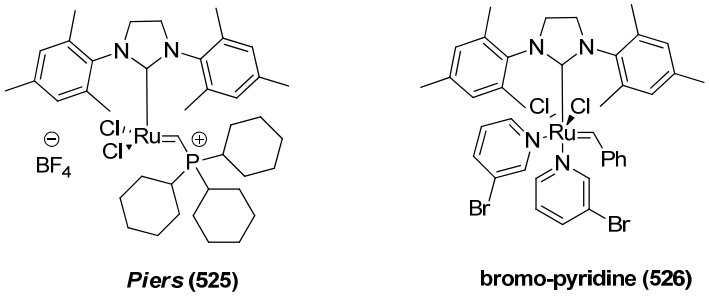


Figure 52: *Piers* catalyst (left) and bromo-pyridine catalyst (right).

Very much to our delight, the ring-closed product **524** was isolated in 52% yield using *Grubbs* 2nd or in 60% yield using *Hoveyda-Grubbs* 2nd generation catalyst, both in benzene at 65 °C. All four conditions investigated led to product formation (Table 16), although with clearly different efficiencies. In refluxing dichloromethane the conversion was very slow in comparison to benzene at 65 °C. Only one isomer was observed and according to NOESY and ROESY experiments the desired *trans* double bond was formed selectively (Figure 53). The absence of an NOE between the olefinic proton attached to C11 at the newly formed double bond and the three protons of the methyl group on C12 together with the detection of an NOE between the methyl protons protons and the two protons at C10 strongly suggest that the *E*-isomer was formed.

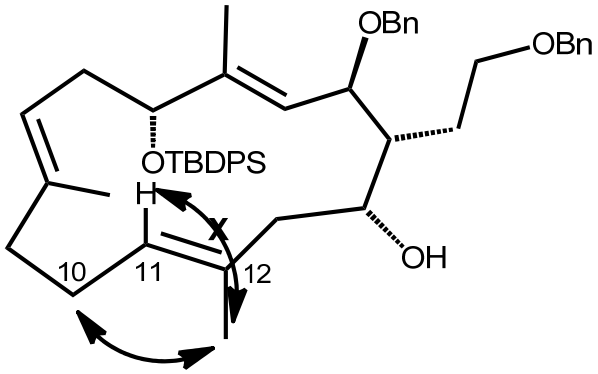
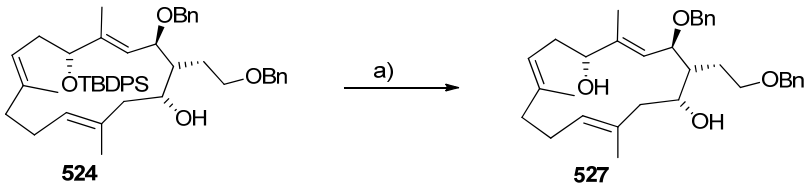


Figure 53: NOE signals of ring-closed product **524**.

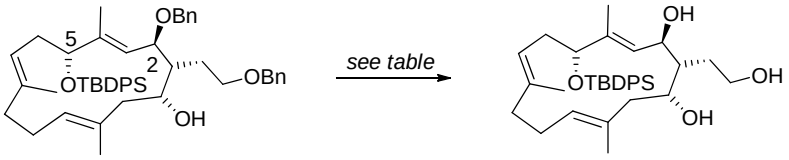
With the feasibility of the RCM-mediated ring-closure demonstrated, we then turned our attention to the removal of the various protecting groups; in particular, the cleavage of the two benzyl ethers in the presence of the three internal trisubstituted double bonds was felt to pose a major challenge. In a first step, the TBDPS group in **524** could be removed with an excess of TAS-F in MeCN in good yield (70%), while other fluoride sources did not yield any (TBAF) or only traces of product (HF-pyridine) (Scheme 142).



524 **527**

Scheme 142: a) TAS-F, MeCN, 80 °C, 70%.

The more difficult task to selectively remove the two benzyl groups was initially approached by treatment of **524** with DDQ, which only led to decomposition (Table 18). The use of BCl_3 did allow the isolation of a mono-deprotected product in yields of 30-40% but the addition of further equivalents of the *Lewis* acid in order to remove the benzyl group on the secondary hydroxy group triggered elimination across the C2-C3 bond. Other *Lewis* acids investigated were TMSI, which only led to decomposition, and $TiCl_4$, which only induced elimination at both, C2 and C5. Catalytic hydrogenation over palladium on activated charcoal, in addition to cleavage of the benzyl ethers, reduced at least one of the double bonds, if carried out in EtOAc or MeOH. In THF the reduction of the double bond was slower so that at least the primary benzyl ether could be cleaved selectively in yields varying between 40-55%.[251] But any attempt to cleave the secondary benzyl ether by addition of more catalyst or by applying higher hydrogen pressure predominately resulted in the reduction of the double bonds.



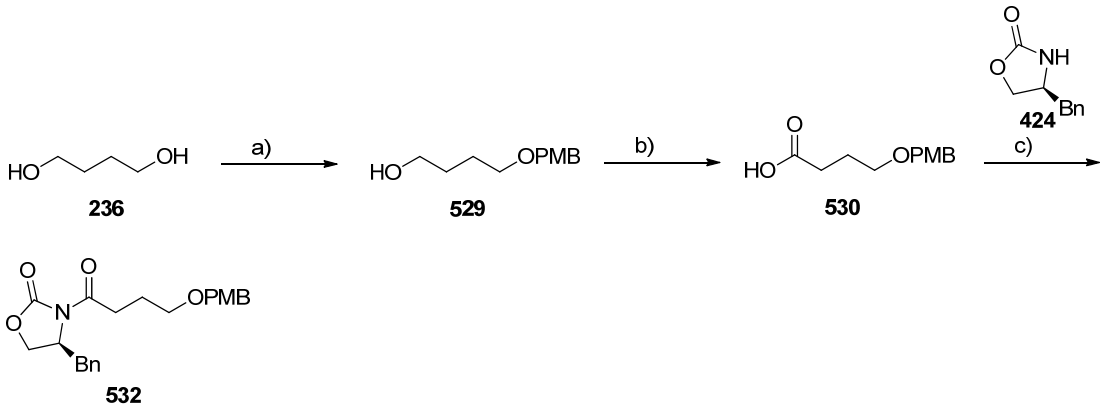
524 **528**

Table 18: Different conditions investigated for the removal of benzyl groups from **524**.

Entry	Reagent	Solvent	Temperature	Reaction
1	DDQ	CH_2Cl_2	rt	decomposition
2	BCl_3	CH_2Cl_2	rt	mono-deprotection, elimination at C2
3	TMSI	CH_2Cl_2	0 °C	decomposition
4	$TiCl_4$	CH_2Cl_2	-78 °C	double elimination at C2 and C5
5	Raney-Ni	EtOH	70 °C	no conversion
6	H_2, Pd/C	EtOAc	rt	mono-deprotection, double bond gets reduced
7	H_2, Pd/C	MeOH	rt	mono-deprotection, double bond gets reduced
8	H_2, Pd/C	THF	rt	mono-deprotection selective, then one double bond gets reduced

This outcome strongly suggested that a mode of protection other than a benzyl ether would be necessary, at least for the secondary allylic hydroxy group at C2. Since the *Sakurai* addition requires a coordinating protecting group, the best choice at this point seemed to be the PMB group. In order for both ether protecting groups to be removable in one step, both the secondary and the primary hydroxy group were to be protected as PMB ethers and deprotection was expected to be feasible with DDQ.

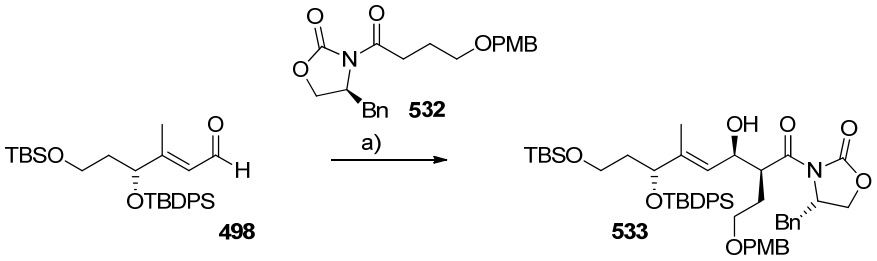
In order to implement this modified strategy, acylated *Evans*-auxiliary **532** bearing a PMB group on the primary hydroxy function was required. This compound was synthesized from 1,4-butandiol (**236**) as described in the literature (Scheme 143).[264]



236 **529** **530** **532**

Scheme 143: a) PMBCl, NaH, TBAI, THF, 96%; b) CrO_3, H_2SO_4, acetone, 93%; c) EtOCOCl, NEt_3, Et_2O, 0 °C, 42%.

Rather surprisingly, **532** behaved very differently from its benzyl-protected variant **240** in the aldol addition with aldehyde **498**. The formation of the enolate had to be carried out at -78 °C rather than at room temperature as it was the case for **240**, at higher temperatures the enolate derived from **532** was not stable as indicated by TLC analysis and no addition took place. If the aldehyde was added at -78 °C, the aldol product **533** was isolated in 40% yield with a low dr of 2:1. Lowering the temperature to -100 °C improved the selectivity up to 4:1, but left the yield unchanged at 40% (Scheme 144).

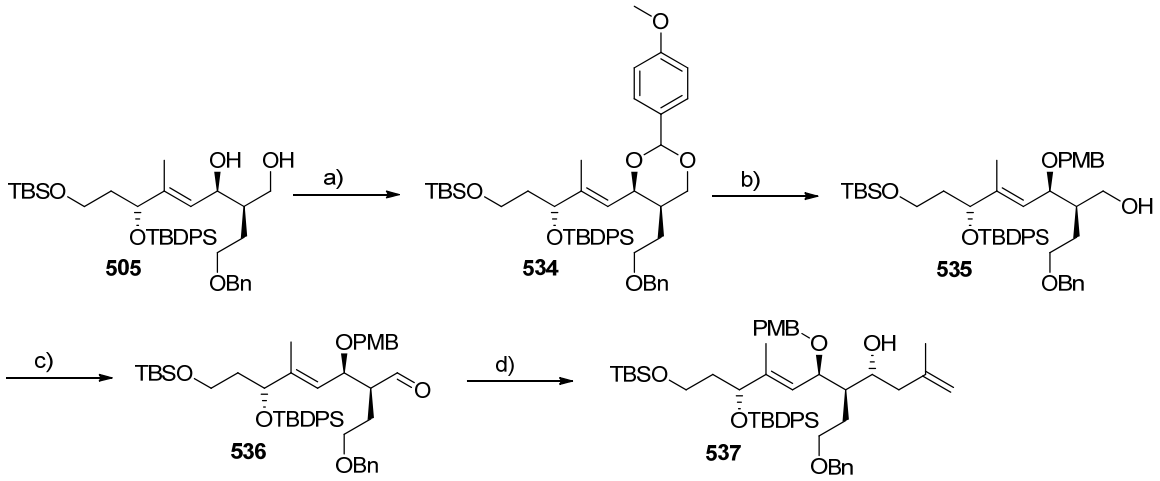


Scheme 144: a) **532**, Bu_2BOTf, DIPEA, CH_2Cl_2, -78 °C, then **532**, -100 °C, 40%, dr 4:1.

As a consequence of the inacceptable yield of the aldol addition, it was decided to maintain the benzyl protection of the primary hydroxy group and use the PMB group only for the protection of the secondary alcohol function. As discussed above (Table 19), there was some indication that the primary benzyl ether could be cleaved in the presence of the double bonds either with BCl_3 or by catalytic hydrogenation over Pd on activated charcoal in THF.

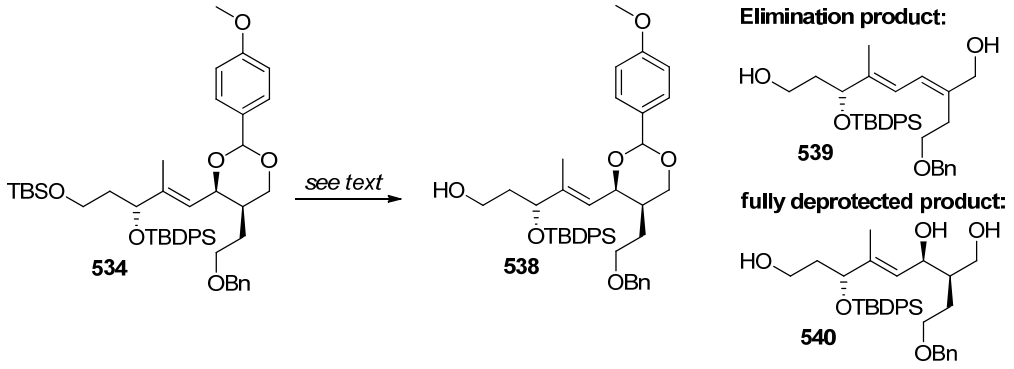
The issue to clarify then was whether the *Sakurai* addition with a PMB ether as coordinating group would still give reasonable selectivity. In the synthesis the addition was planned to be carried out with the aldehyde **495** having the terminal olefin already in place (Section 2.3.3, Scheme 134) but it was felt that the feasibility of the addition could also be investigated on the synthetically easier accessible intermediate **536** that was synthesized in three steps from diol **505** (Scheme 145).

The latter was first converted into its PMB acetal; subsequent reductive opening of the acetal moiety gave primary alcohol **535**, which was oxidized to the aldehyde **536** using TEMPO. Aldehyde **536** then underwent *Sakurai* addition in a reasonable yield of 60%, but more importantly, with a dr of 12:1 the addition was still selective.



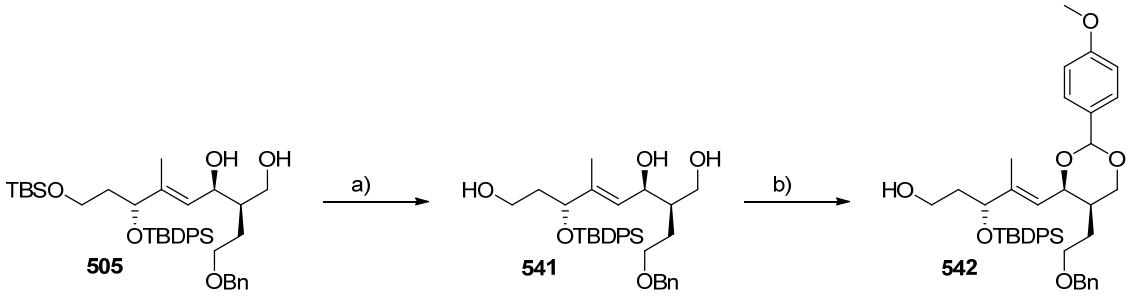
Scheme 145: a) PMB aldehyde dimethyl acetal, CSA, CH_2Cl_2, 85%; b) DIBAL-H, Me_2AlCl, CH_2Cl_2, -20 °C, 90%; c) BAIB, TEMPO, CH_2Cl_2, 0 °C, 89%; d) methallyl trimethyl silane, $SnCl_4$, -90 °C, 60%, dr 12:1.

In light of this success, the synthetic strategy involving combined Bn/PMB protection was further pursued. Attempts to remove the TBS-group in **534**, on route to the intended installation of the terminal double turned out to be troublesome (Scheme 146). While TBAF, buffered TBAF, TAS-F and HF-pyridine only led to decomposition, treatment of **534** with $FeCl_3$ only formed the elimination product **539**. *Bronsted* acids like CSA, PTSA, PPTS and AcOH exclusively afforded the fully deprotected product **540** independent of the amounts of acid used.



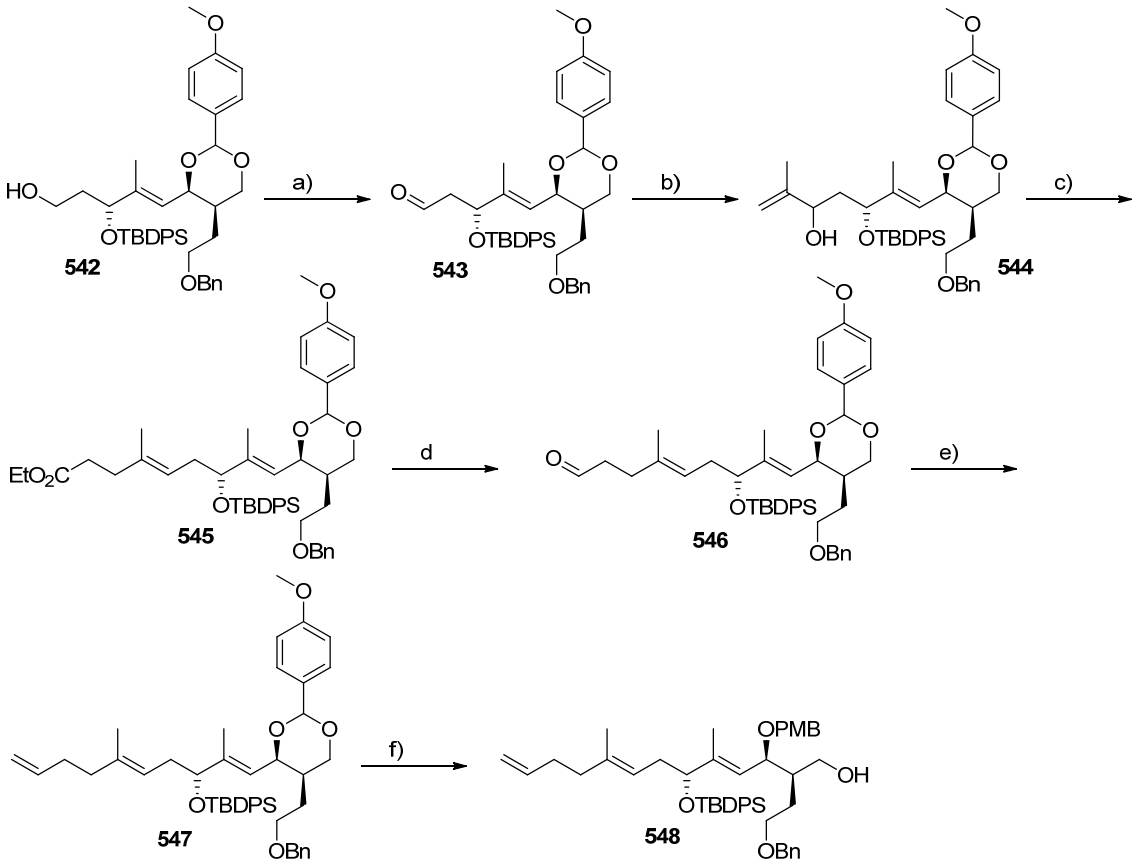
Scheme 146: Deprotection of TBS-ether **534**.

This problem could be overcome, however, by changing the order of steps, i. e. by removing the TBS-group from the diol **505** and converting the resulting free triol **541** into acetal **542**, which leaves the number of steps unchanged (Scheme 147).



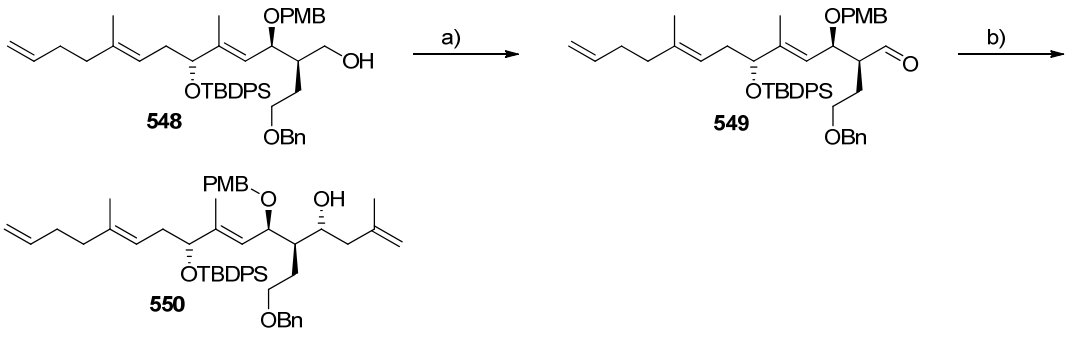
Scheme 147: a) AcOH, THF, H_2O, 81%; b) PMB aldehyde dimethyl acetal, CSA, CH_2Cl_2, 93%.

The free primary alcohol **542** was then oxidized using TEMPO in 91% yield – DMP gave a slightly lower yield (71%) – and the resulting aldehyde **543** was reacted with isopropenylmagnesium bromide to give a mixture of secondary alcohols **544** (Scheme 148). Treatment of this mixture with triethyl orthoacetate then induced the *Johnson-Claisen* rearrangement to afford ester **545**. Reduction of the ester moiety with one equivalent of DIBAL-H gave aldehyde **546** in 83% yield. Again, there was some overreduction to the alcohol; this compound could be isolated and reoxidized to the aldehyde **546** in a subsequent step. Installation of the terminal double bond under *Wittig* conditions proceeded in an excellent yield of 87% using *n*-BuLi as a base. The use of LiHMDS only afforded 70% of the olefin **547**. Opening of the PMB acetal moiety in **547** under the conditions that had been established for the corresponding benzyl acetal **458** (Section 2.3.2.1, Scheme 121) gave primary alcohol **548** almost quantitavely (Scheme 148).



Scheme 148: a) BAIB, TEMPO, CH_2Cl_2, 0 °C, 91%; b) Isopropenyl magnesium bromide, THF -78 °C, 93%; c) $MeC(OEt)_3$, propionic acid, toluene, reflux, 85%; d) DIBAL-H, CH_2Cl_2, -78 °C, 83%; e) Methyl triphenylphosphonium iodide, *n*-BuLi, THF, 87%; f) DIBAL-H, Me_2AlCl, CH_2Cl_2, -20 °C, 99%.

Subsequent DMP oxidation of the primary hydroxy group gave aldehyde **549** in good yield (82%; TEMPO turned out to be less efficient). Gratifyingly, the following *Sakurai* addition showed the same efficiency as had been observed with aldehyde **536** (Scheme 145) and gave secondary alcohol **550** in very good yield (84%) and with a reasonable dr of 10:1 (Scheme 149). Note that the benzyl protected aldehyde **495** (Scheme 141) had given a similar yield but an enhanced dr of 25:1.



Scheme 149: a) DMP, CH_2Cl_2, 82%; b) Methallyl trimethyl silane, $SnCl_4$, CH_2Cl_2, -90 °C, 84%, 96% brsm, dr 10:1.

Secondary alcohol **550** was esterified with both enantiomers of the *Mosher* acid. The analysis of the resulting esters gave a very coherent picture and confirmed the desired stereochemical outcome of the addition (Figure 54).

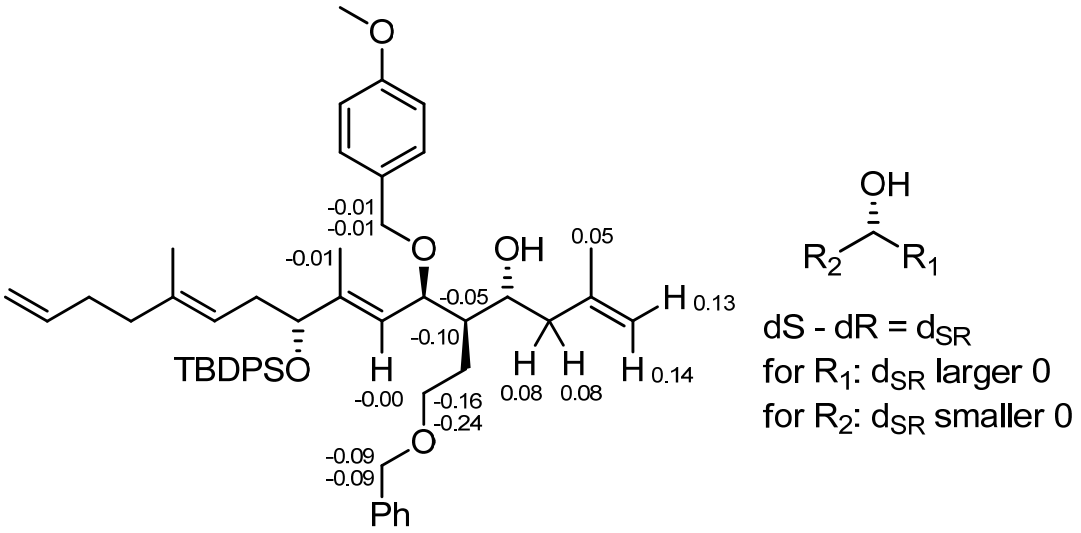
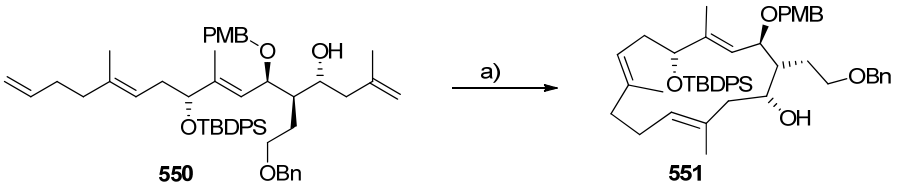


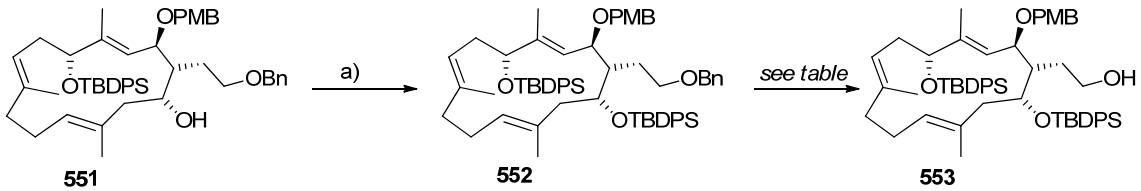
Figure 54: Mosher ester analysis of secondary alcohol **550**.

At this point the stage was again set for the critical RCM reaction. Very gratifyingly, the treatment of **550** with the 2nd generation *Hoveyda-Grubbs* catalyst in benzene at 65 °C gave the macrocycle in 68% yield as a single isomer (Scheme 150).



Scheme 150: a) *Hoveyda-Grubbs* II, benzene, 65 °C, 68%, 78% brsm, single isomer.

After TBDPS-protection of the secondary hydroxy group in **551** with TBDPSOTf (TBDPSCl was inefficient), the removal of the benzyl group was attempted under different conditions (Scheme 151, Table 19). Unfortunately, BCl_3 only resulted in the formation of the elimination product **554**, while catalytic hydrogenation under different conditions gave no conversion.

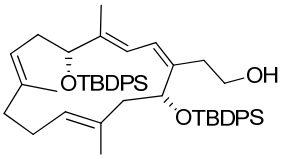


Scheme 151: a) TBDPSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C, 94% .

Table 19: Different conditions exploited to remove the benzyl group on **552**.

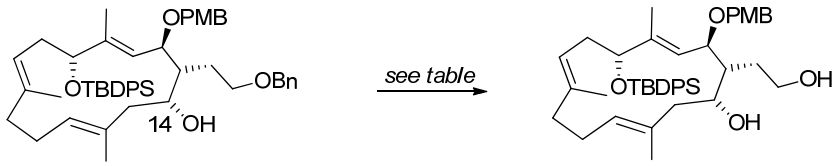
Entry	Reagent	Solvent	Temperature/ pressure	Reaction
1	BCl_3	CH_2Cl_2	0 °C	Elimination
2	H_2, Pd/C	THF	rt, balloon	no reaction
3	H_2, Pd/C	THF	rt, 4 bar	One double bond gets reduced, little formation of product
4	H_2, Pd/C	EtOAc	65 °C, 4 bar	no reaction

Elimination product:



554

Based on the hypothesis that the bulky silyl ether group on C14 could sterically hinder the palladium (or other agents for that matter) to approach the benzyl group, the deprotection was attempted on the unprotected secondary alcohol **551** (Table 20).



551 **555**

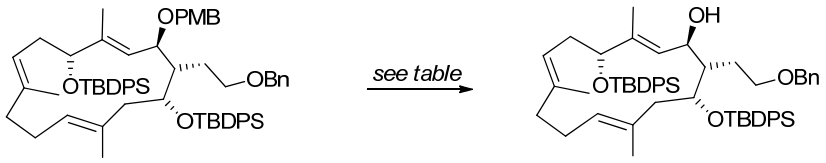
Table 20: Different conditions exploited to remove the benzyl group on **551**.

Entry	Reagent	Solvent	Temperature/ pressure	Reaction
1	BCl_3	CH_2Cl_2	rt	elimination
2	BF_3, EtSH	CH_2Cl_2	rt	no reaction
3	H_2, Pd/C	THF	rt, balloon	52%

With this substrate the removal of the benzyl group from **551** was possible by catalytic hydrogenation in THF, which gave the desired product **555** in 52% yield (Table 19), thus indicating that the sterically less hindered substrate **551** was indeed more susceptible towards hydrogenation. However, on a larger scale (20-30 mg) this result was not reproducible and the yield dropped to 35% at the expense of the reduction of a double bond. This observation may be explained by the lower uptake of hydrogen on a larger scale, which results in prolonged reaction times and thus more extensive exposure of the three double bonds present in both the starting material as well as the product to the reducing conditions.

In light of the troublesome removal of the benzyl group, the decision was taken to investigate cleavage of the PMB-ether prior to benzyl removal, hoping that the benzyl ether would then become sterically more accessible (Table 20). However, treatment of **552** with DDQ resulted

in the isolation of the desired secondary alcohol **556** in no more than 10% yield. Varying the solvent composition or addition of di-*tert*-butyl pyridine did not improve the yield. *Lewis* acids like $MgBr_2 \bullet OEt_2$ or $SnCl_2$ remained ineffective. The best yield was obtained by treatment of **552** with CAN, which gave **556** in 27% yield (Table 21, Entry 9), but this was still far from acceptable.



552 **556**

Table 21: Different conditions exploited to remove the PMB group on **552**.

Entry	Reagent	Solvent	Temperature	Yield
1	DDQ	CH_2Cl_2/H_2O	0 °C	10%
2	DDQ	CH_2Cl_2/phosphate buffer 9:1	rt	8%
3	DDQ	CH_2Cl_2/phosphate buffer 4:1	rt	decomposition
4	DDQ, ditertbutyl py	CH_2Cl_2	rt	decomposition
5	DDQ, diterbutyl py	CH_2Cl_2/H_2O 8:1	rt	10%
6	$MgBr_2 \cdot OEt_2$, SMe_2	CH_2Cl_2	rt	decomposition
7	$SnCl_2$, $PhOCh_2COCl$	$MeCN/H_2O$	rt	elimination
8	Trisparabromophenyl ammonium radical	MeCN	rt	decomposition
9	CAN	$MeCN/H_2O$	rt	27%

At this stage the synthetic strategy was at a critical juncture. Neither of the two ether protecting group could be removed in a satisfactory way and there was significant doubt whether the synthesis could be finished in a reasonable way. In order to explore all conceivable options, it was decided to attempt removal of the PMB-group on the linear substrate **550**, i. e. prior to the RCM (Table 21). While treatment of **550** with $SnCl_4$ resulted in decomposition and DDQ under standard conditions delivered the diol **557** only in 10% yield, the use of 2 eq. CAN in a mixture of MeCN and water (9:1) increased the yield up to 52% (Table 22, Entry 7). Changing the composition of the solvent mixture did not lead to any improvement, but if the deprotection was carried out with 2.7 eq. of CAN and the reaction was stopped prior to full conversion, **557** was obtained in 56% yield. Exposure of the reisolated starting material to the same conditions then provided **557** in an overall yield of 74%. If the reaction was quenched at the point of full conversion, the yield was 56%.

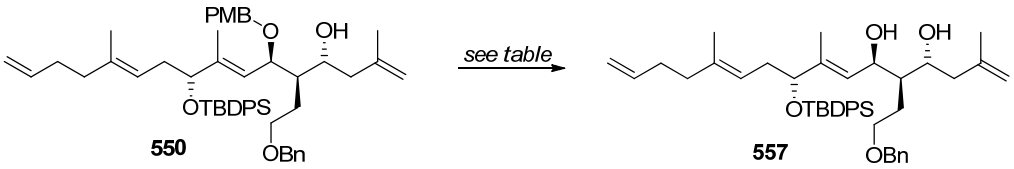
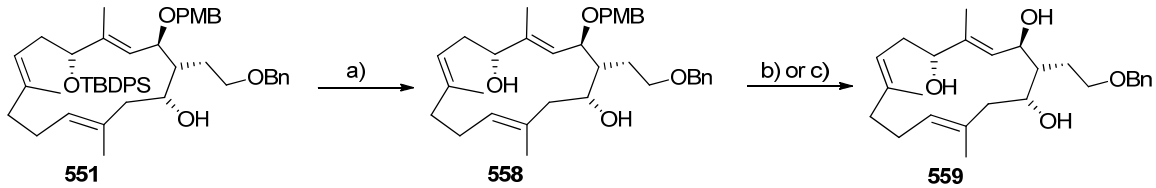


Table 22: Different conditions exploited to remove the PMB group on the linear substrate **550**.

Entry	Catalyst	Solvent	Temperature	Yield
1	$SnCl_4$	CH_2Cl_2	0 °C	decomposition
2	DDQ/$FeCl_3$	CH_2Cl_2/H_2O 9:1	rt	decomposition
3	DDQ/$NaHCO_3$	CH_2Cl_2/H_2O 9:1	rt	10%
4	DDQ	CH_2Cl_2/phosphate buffer 4:1	rt	elimination
5	DDQ	CH_2Cl_2/phosphate buffer 6:1	rt	elimination
6	DDQ	CH_2Cl_2/phosphate buffer 9:1	rt	10%
7	CAN (2.0 eq)	$MeCN/H_2O$ 9:1	rt	52%
8	CAN (2.0 eq)	$MeNO_2$/H2O 9:1	rt	no reaction
9	CAN (2.0 eq)	acetone/H_2O 9:1	rt	37%
10	CAN (2.7 eq)	$MeCN/H_2O$ 9:1	rt	56%, 74% (overall)

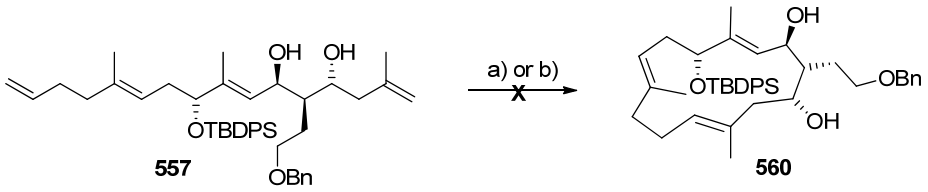
With this result in hand, it was investigated whether these conditions might allow cleavage of the PMB-group at the stage of the macrocycle in the absence of the bulky TBDPS-group (Scheme 152). The silyl ether protecting group was removed from **551** with TAS-F to give allylic alcohol **558**. Subsequent oxidative cleavage of the PMB-group with CAN, however, was far less efficient than on the linear substrate **550**. In addition, DDQ led to exclusive formation of the enone, a side reaction which is known to occur with allylic alcohols.[265],[266],[267] This outcome definitely made it clear that the PMB-group had to be removed prior to ring-closure.



Scheme 152: a) TAS-F, MeCN, 80 °C, 94%; b) CAN, $MeCN/H_2O$ 9:1, 10%; c) DDQ, CH_2Cl_2/buffer pH 7, no product, only enone formed.

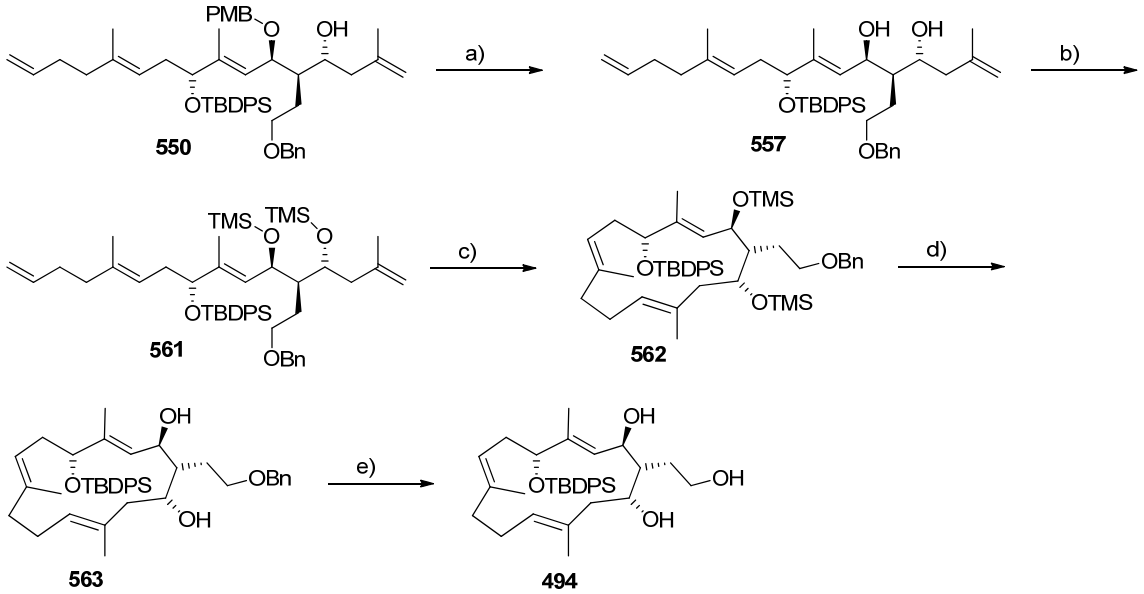
In contrast to diene **550**, with a single free hydroxy group, diene **557,** quite surprisingly**,** did not undergo cyclization in the presence of *Grubbs* 2nd or *Hoveyda-Grubbs* 2nd generation

catalysts, either in benzene or in dichloromethane (Scheme 153). The free hydroxy groups were thus TMS protected and the fully protected diene was subjected to RCM conditions (Scheme 154).



Scheme 153: a) *Grubbs* II, benzene, 65 °C or CH_2Cl_2, reflux; b) *Hoveyda-Grubbs* II, benzene, 65 °C, CH_2Cl_2, reflux.

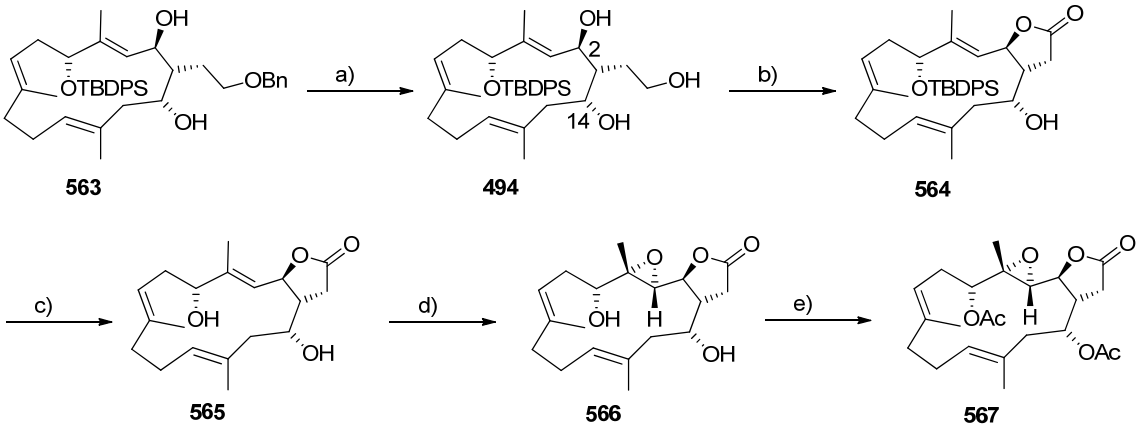
To our delight, bis-silyl ether **561** could be converted into macrocycle **562** as a single isomer in a remarkable yield of 94% with the 2nd generation *Hoveyda-Grubbs* catalyst (Scheme 154). To the best of our knowledge, there is no report in the literature of a similarly efficient RCM-mediated ring-closure to a trisubstituted olefin product. Only a very weak NOE between the methyl group and the olefinic proton at newly formed double bond was observed, which strongly indicated that the ring had closed with a *trans* geometry of this double bond. Subsequent treatment of macrocycle **562** with citric acid yielded diol **563** in 92% yield.



Scheme 154: a) CAN, MeCN/H_2O 9:1, 74% overall; b) TMSOTf, 2,6-lutidine, CH_2Cl_2, -78 °, 94%; c) *Hoveyda-Grubbs* II, benzene, 65 °C, 94%, single isomer; d) Citric acid, MeOH, 92%; e) H_2, Pd/C, hexanol, 76%.

At this stage the benzyl group had to be removed so that the lactonization could be induced by oxidation of the resulting primary alcohol **494**. Out of the many different conditions that had

been evaluated on similar substrates previously (Table 20) catalytic hydrogenation in THF afforded triol **494** in 53% yield on small scale (5 mg), but as previously observed for other cases, the reaction suffered from poor reproducibility even on a slightly larger scale (20 mg). It was found that hexanol was the solvent of choice for the hydrogenation, yielding triol **494** in 76%, but even with this solvent the reaction could not be scaled up. As a consequence, and even though the chances of success were considered to be low, *Birch* conditions were also investigated for the removal of the benzyl group. Very much to our surprise, the reduction afforded the triol **494** in 83% yield (Scheme 155). Experimentally, liquid ammonia was condensed in a flask and a solution of benzyl ether **563** in THF was added at -78 °C. Very small pieces of sodium were then added cautiously to the solution and the reaction was quenched immediately after the mixture had turned blue. If the reaction mixture was stirred longer, one double bond was also reduced. With this obstacle left behind, the next question to be answered was whether the triol **494** would form the desired lactone **564** upon oxidation of the primary hydroxy group. It was assumed that the allylic hydroxy group on C2 would be more nucleophilic than the one on C14, thereby leading to the formation of the targeted lactol, which would then be further oxidized to the lactone **564**, but this outcome was by no means certain. Initial attempts at oxidation of **494** involved treatment with TPAP, which led to the formation of enone as the only observable product. To avoid this reaction path, a sterically more demanding reagent was explored and TEMPO oxidation did indeed induce the formation lactone **564** in an excellent yield of 75% (*via* the corresponding lactol) (Scheme 155).



Scheme 155: a) Na, NH_3, THF, -78 °C, 83%; b) BAIB, TEMPO, CH_2Cl_2, 0 °C, 75%; c) TAS-F, MeCN, 80 °C, 77%; d) $VO(acac)_2$, *t*-BuOOH, benzene, 0 °C to rt, 52%, single isomer; e) NEt_3, DMAP, Ac_2O, MeCN, 84%.

The chemical shift of the allylic proton at C2 strongly indicated that lactonization had taken place through the allylic hydroxy group. While a shift of 5.30 ppm is in fact expected for such a proton, it would be too far downfield for an allylic alcohol (Figure 55).

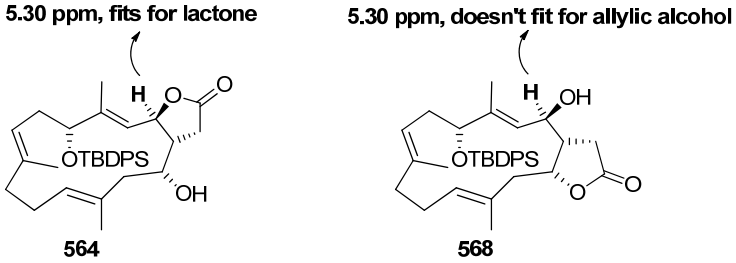
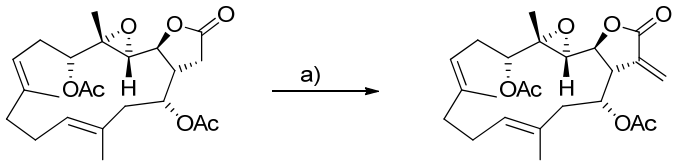


Figure 55: Comparison of the two possible lactone variants.

Removal of the TBDPS-group was achieved by treatment of **564** with TAS-F in acceptable yield. This was followed by directed epoxidation of **565** with $VO(acac)_2$, which gave epoxide **566** in 52% as a single isomer (see, however, below). A substantial amount (30%) of a di-epoxide was also isolated, which arises from additional epoxidation of the double bond of one of two homo-allylic alcohol system.

Attempts to surpress the formation of the di-epoxide side product by changing the solvent (CH_2Cl_2, toluene) or lowering the temperature (to between -78 °C and 0 °C) were not successful. As only limited amounts of **565** were left at this stage, it was decided to convert all of this material into epoxide **566** following the above procedure, rather than to evaluate alternative epoxidation conditions, such as the use of *m*-CPBA or *Sharpless* asymmetric epoxidation. Acetylation of mono-epoxide **566** with Ac_2O gave diacetate **567** in high yield (84%) (Scheme 155). The (seemingly) final question to be addressed in the synthesis of michaolide E (**4**) was the selective methenylation of **567** α to the lactone carbonyl in the presence of two acetate esters. When **567** was exposed to LiHMDS at -78 °C followed by warming to 0 °C and quenching with D_2O, deuterium was exclusively incorporated at the desired position in the lactone ring. This demonstrated that the two acyl groups would not interfere with the methenylation. Indeed, when the lithium enolate of **567** was reacted with *Eschenmoser*'s salt at -78 °C and the reaction mixture was then allowed to reach 0 °C (Scheme 156), the presumably final product **4** could already be observed by TLC without addition of MeI (which is commonly employed to affect the elimination of trimethylamine after addition of the reagent to an enolate). However, based on MS analysis the reaction mixture still contained quantities of the initially formed tertiary amine; thus, after extractive

work-up, the residue was taken up in MeOH and treated with MeI. The seemingly final product **4** could be isolated in 40% yield.

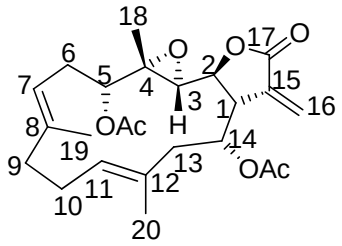


567 **5**

Scheme 156: a) LiHMDS, THF, -78 °C to 0 °C, -78°C, then *Eschenmoser's* salt, -78 °C to 0 °C, MeI, MeOH, 40%.

Unfortunately, comparison of the NMR-data of **5** with the data published for michaolide E (**4**) in the context of the original isolation work[159] revealed significant differences for a number of chemical shift values but also for some coupling constants (Table 23). Based on the spectroscopic data it is clear that synthetic **5** and natural michaolide E (**4**) have different structures. Whether this is due to the fact that the structure of the natural product was misassigned or whether **5** has not the proposed and expected structure remains to be determined. In order to convey a sense for the solidity of the stereochemical assignments for the various stereocenters in **5**, all chiral transformations from the reaction sequence shall be analyzed at the end of this section.

Table 23: Comparison of ^{1}H-NMR data (chemical shifts) of **5** and michaolide E (**4**). Recorded in $CDCl_3$ (assigned by COSY, HSQC, and HMBC experiments). *J* values (in Hz) in parentheses.



Michaolide E (**4**)

Protons	**5**	**Michaolide E (4)**
1	3.23 ddd (1.6, 1.6, 1.5)	3.30 m
2	4.56 dd (8.2, 1.5)	4.99 m
3	3.05 d (8.2)	2.87 d (3.6)
5	5.20 dd (5.0, 4.0)	4.46 dd(3.9, 11.4)
6	2.42 m	2.18 m
	2.42 m	2.45 m
7	5.08 m	4.97 m
9	2.01 m	2.00 m
	2.29 m	2.27 m
10	2.19 m	2.12 m
	2.28 m	2.30 m
11	5.11 m	5.25 m
13	2.21 m	2.26 m
	2.43 m	2.43 m
14	5.10 m	5.26 m
16	5.80 d (1.6)	5.73 d (2.7)
	6.42 d (1.6)	6.40 d (2.1)
18	1.42 s	1.45 s
19	1.60 s	1.67 s
20	1.69 s	1.76 s
5-OAc	2.03 s	2.11 s
14-OAc	1.99 s	2.01 s

Another question to be addressed is the presence of a similar set of signals in the ^{1}H-NMR of about 17% in **5** (Figure 56). It could not be removed by HPLC purification, which led to the idea that it might be a conformer. However, this set of signals remains unchanged upon heating indicating that is not originating from a conformer but maybe rather from an isomer.

What kind of isomer it is and how it could appear in the last reaction remains unclear up to now.

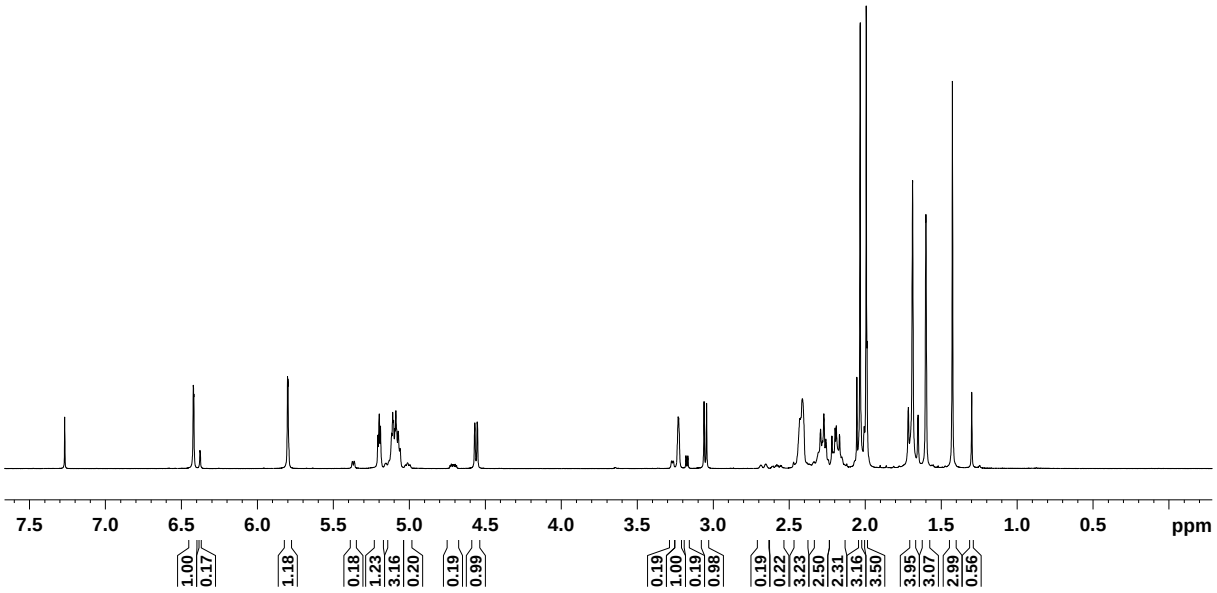
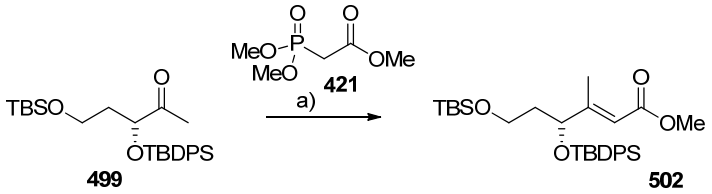


Figure 56: ^{1}H-NMR spectrum of **5**.

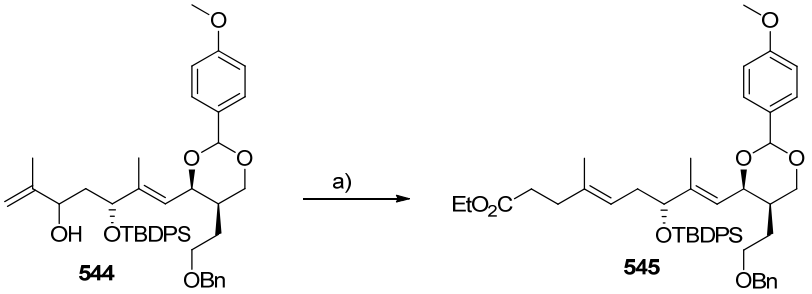
2.3.4 Assessment of Asymmetric Transformations

In the course of the synthesis of **4** three different methods were explored to install the trisubstituted *trans* double bonds between C3/C4, C7/C8, and C11/12, respectively. First, α,β-unsaturated ester **502** was synthesized by means of an *HWE* olefination reaction (Scheme 157). The ester **502** formed as a single isomer and although *HWE* olefinations with ketones exhibit poor to modest *E*-selectivity only, the situation with methyl ketones looks different. Methyl ketones undergo *HWE* olefinations with reasonable to excellent *E*-selectivity.[268],[269],[270],[63] In addition, no NOE between the olefinic proton and the protons at the methyl group, which is attached to the double bond, was observed, so that the exclusive formation of the *Z* isomer can safely be excluded.



Scheme 157: a) **421**, NaH, THF, 0 °C to rt, 94%.

The second trisubstituted *trans* double bond was installed making use of a *Johnson-Claisen* rearrangement giving olefin **545** as a single isomer (Scheme 158).



Scheme 158: $MeC(OEt)_3$, propionic acid, toluene, reflux, 85%.

As depicted in Figure 57 the rearrangement proceeds *via* two possible six-membered chair-like transition states, both of which give the same *trans* olefin**,** because of the fact that R_L prefers to occupy an equatorial position.[263] Many examples of this kind of rearrangement are reported in literature to be *E*-selective.

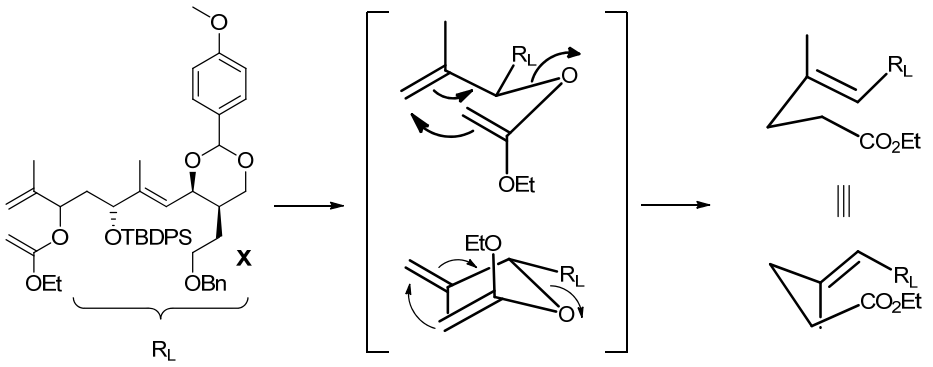
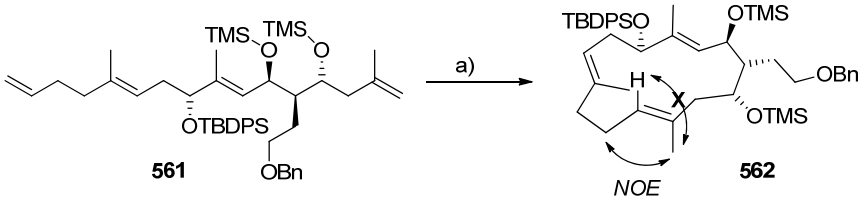


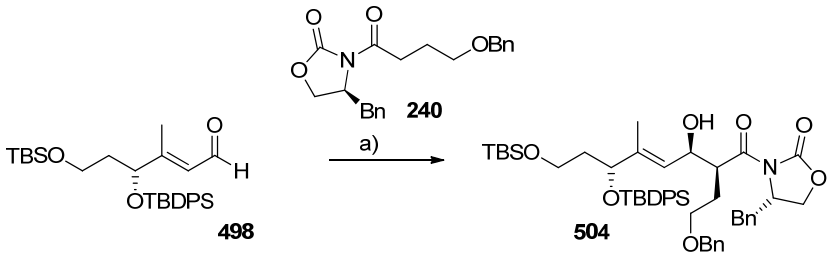
Figure 57: *Johnson-Claisen* rearrangement giving *trans* olefin.

The third trisubstitued *trans* olefin was formed by RCM and this reaction has been extensively discussed in previous sections. The NOE's observed for the different cyclization products strongly indicated the formation of the *trans*-isomer in all cases (Scheme 159), although a *cis* configuration of the double bond cannot be rigorously excluded based on the absence of an NOE.



Scheme 159: a) *Hoveyda-Grubbs* II, benzene, 65 °C, 94%, only isomer.

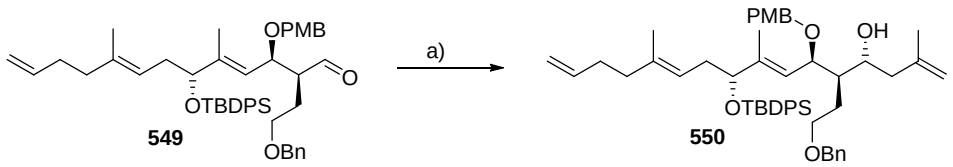
To install the two adjacent stereocenters at C6 and C7 which exhibit a *syn* relationsship, an asymmetric aldol addition was carried between aldehyde **498** and imide **240**. The aldol product **504** was formed as a single isomer (Scheme 160). This type of reaction is very well established and exclusive formation of the undesired *syn* or even an *anti* diastereomer is more than unlikely.[242]



Scheme 160: a) **240**, Bu_2OTf, DIPEA, CH_2Cl_2, -78 °C, 90%, only isomer.

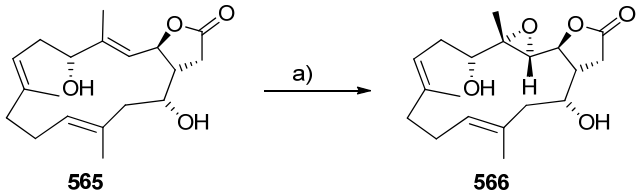
A selective *Sakurai* addition to aldehyde **549** was exploited to arrive at the secondary alcohol **550** (Scheme 161). The very coherent picture provided by the *Mosher* ester analysis

of the addition product (Section 2.3.3.1, Figure 54) confirmed the desired stereochemical outcome of this reaction.



Scheme 161: a) methallyl trimethyl silane, $SnCl_4$, CH_2Cl_2, -90 °C, 84%, 96% brsm, dr 10:1.

The last asymmetric transformation in the synthesis was a directed epoxidation of allylic alcohol **565** mediated by $VO(acac)_2$ to provide a single isomer of an epoxide **566** (Scheme 162).[271]



Scheme 162: a) $VO(acac)_2$, *t*-BuOOH, benzene, 0 °C to rt, 52%, single isomer.

According to *Sharpless* the predicted dihedral angle O-C-C=C for the V-coordinated intermediate in the *t*-BuOOH/$VO(acac)_2$-catalyzed epoxidation of allylic alcohols is approximately 50 °.[272] In Figure 58 the two transition states leading to the *threo* (desired) and to the *erythro* (undesired) product are depicted.

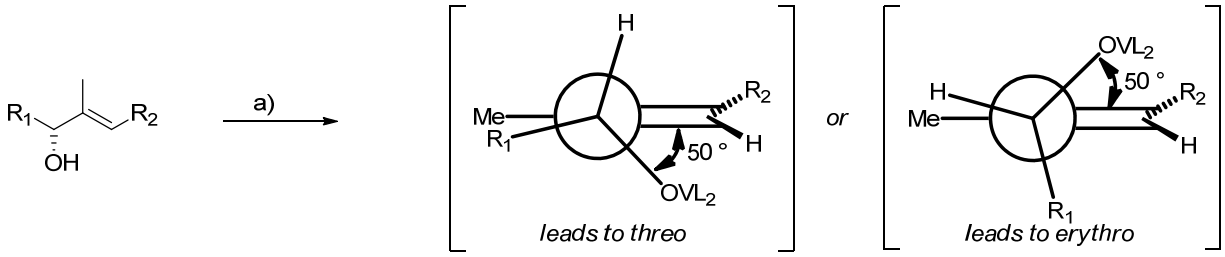
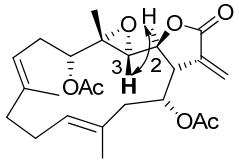


Figure 58: a) $VO(acac)_2$, *t*-BuOOH; Transition states leading to *threo* and *erythro* product, respectively.

For cyclic allylic alcohols bearing a methyl group attached to a double bond α to the hydroxy moiety (Figure 58, motif present in **565**) the stereochemical outcome of the epoxidation is hard to predict. For linear substrates, depending on the most stable conformation either the *threo* (Figure 58, oxygen is positioned beneath the double bond) or the *erythro* (Figure 58, oxygen positioned above the double bond) product is favoured. Due to the fact that in cyclic substrates as the allylic alcohol **565** the rotational freedom around the C-C bond considered in

the *Newman* projection is likely to be restricted, dihydral angles other than 50 °C might have to be taken into account. Both stereochemical outcomes have been reported for the few related (cyclic) systems found in literature.[273],[274]

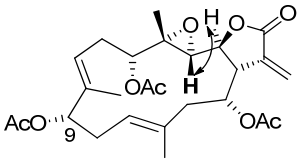
Comparison of the NMR data summarized in Table 22 (Section 2.3.3.1) clearly shows that the coupling constant with largest deviation between **5** and michaolide E (**4**) is associated with the coupling between the protons at C2 and C3 (Figure 59). While synthetic **5** shows a coupling constant of 8.2 Hz, the coupling constant of natural michaolide E is only 3.6 Hz.



4

Figure 59: The epoxide proton in **5** exhibits the largest difference in coupling constant compared to natural michaolide E (**4**).

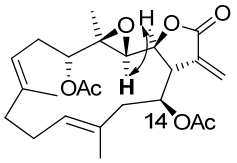
In light of this observation the literature was searched thoroughly for structurally related epoxides and their NMR properties. Crassolide (**287**), a natural product that was isolated together with michaolide E (**4**) from the soft coral *Lobophytum michaelae* exhibits the very same structure as **4** apart from an additional acetylated hydroxy group at C9 (Figure 60). Its structure was confirmed by X-ray crystallography and the proton at the epoxide moiety exhibits a coupling constant of 3.6 Hz.



Crassolide (**287**)

Figure 60: Crassolide (**287**), the epoxide proton exhibits a coupling constant of 3.6 Hz.

The structure of *trans* fused cembranolide **569** from *Lobophytum cristigalli* was determined by X-ray crystallography.[275] In comparison to michaolide E (**4**) and crassolide (**287**) the epoxide has the opposite configuration; likewise, the configuration of C14, bearing one of the acetylated hydroxy groups is inverted. The relevant coupling constant is reported as 9.0 Hz, which makes it almost identical with the one observed for synthetic **5** (Figure 61).



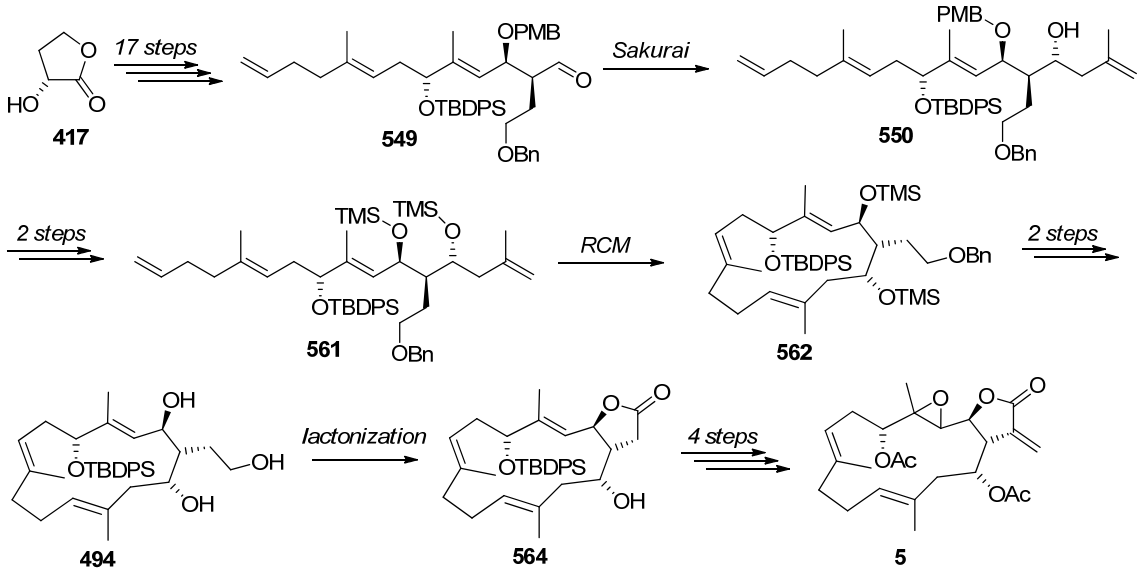
569

Figure 61: In *trans* fused cembranolide (**569**) the epoxide proton exhibits a coupling constant of 9.0 Hz.

The above analysis in combination with the uncertainties associated with the predicted stereochemical outcome of the directed epoxidation of the cyclic allylic alcohol **567** suggests that the latter transformation delivered the undesired *R*/*S* isomer.

2.4 Conclusion and Outlook

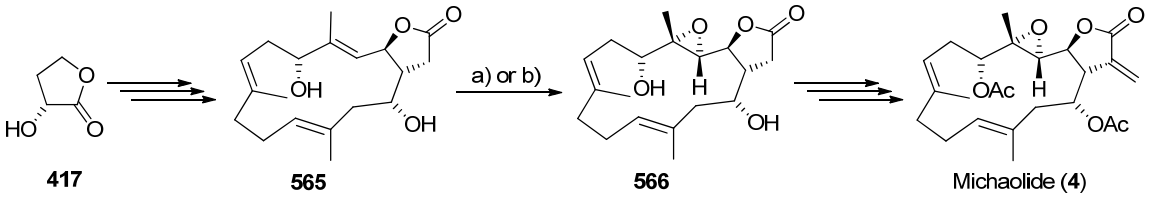
In summary, studies on the enantioselective total synthesis of the cembranolide michaolide E (**4**) have been undertaken. In the initial part of the synthesis commercially available α-hydroxy lactone **417** was elaborated into aldehyde **549,** which underwent selective methallyl addition under β-chelation control (Scheme 162). After a deprotection-protection sequence the pivotal RCM gave all-carbon macrocycle **562** in excellent yield as a single isomer. To the best of our knowledge, this represents the most efficient cyclization to a trisubstituted macrocyclic olefin by means of RCM reported to date. Deprotection of **562** gave triol **494** which upon oxidation formed the lactone **564** regioselectively. Directed epoxidation and final methenylation gave final product **5**.



Scheme 163: Studies towards the synthesis of michaolide E (**4**).

The NMR-data of the final product **5** of our synthesis did not match with the reported data of the isolated natural product. The question whether the structure was misassigned or whether a chiral transformation in the synthetic sequence produced an undesired outcome will be addressed in the near future. Based on the evaluation of relevant literature data for related systems there is reason to believe that the directed epoxidation of the cyclic allylic alcohol **565** produced the undesired *R*/*S* epoxide isomer. Thus, in a next step the established reaction sequence from *D*-malic acid (**416**) to the cyclic allylic alcohol **565** should be repeated and the epoxidation should be carried with, e. g., *m*-CPBA or under *Sharpless* asymmetric epoxidation conditions (Scheme 163). This should provide the epoxide exhibiting the

configuration opposite to that obtained with $VO(acac)_2$/*t*-BuOOH. Completion of the synthesis as established would then be expected to provide michaolide E (Scheme 163).



Scheme 163: a) *m*-CPBA; b) *Sharpless* epoxidation.

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4 Experimental Part

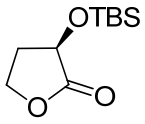
4.1 General Procedures and Analytics

All **solvents** used for reactions were purchased as anhydrous grade from Sigma-Aldrich (puriss., dried over molecular sieves. H_2O < 0.005%). Solvents for extractions, flash column chromatography and thin layer chromatography (TLC) were purchased as commercial grade and distilled prior to use. All non-aqueous reactions were performed under an argon atmosphere using flame-dried glassware and standard syringe/septa techniques. Commercially available reagents were used without further purification, unless otherwise noted. In general, reactions were magnetically stirred and monitored by TLC performed on Merck TLC aluminum sheets (silica gel 60 F254). Spots were visualized with UV light ($\lambda = 254$ nm), through staining with $Ce_2(SO_4)_3$/phosphomolybdic acid/H_2SO_4 (CPS) or $KMnO_4/K_2CO_3$. Chromatographic purification of products was performed using Fluka silica gel 60 for preparative column chromatography (particle size 40-63 μm).

^{1}H- and **^{13}C-NMR** spectra were recorded in $CDCl_3$ on Bruker AV-400 400 MHz and AV-500 500 MHz instruments at room temperature. Chemical shifts (δ) are reported in ppm and are referenced to the solvent signal as an internal standard ($CDCl_3$ δ 7.26 ppm for ^{1}H, δ 77.16 ppm for ^{13}C). All ^{13}C-NMR spectra were measured with complete proton decoupling. Data for NMR spectra are reported as follows: s = singlet, d = doublet, t = triplet, q = quartet, quint = quintet, m = multiplet, br = broad signal, *J* = coupling constant in Hz. **Infrared spectra (IR)** were recorded on a Jasco FT/IR-6200 instrument as thin film. Resonance frequencies are given as wavenumbers in cm^{-1}. **Optical rotations** were measured on a Jasco P-1020 polarimeter operating at the sodium D line with a 10 mm or 100 mm path length cell at 20 °C and are reported as follows: $[\alpha]^T_D$, concentration (g/100 mL), and solvent. **Mass spectra** were recorded by the ETH Zürich MS service. HRMS (ESI) spectra were obtained on a Bruker Daltonics maxis (UHR-TOF) and HRMS (EI) on a Waters Micromass AutoSpec Ultima intstrument.

4.2 Cyclopropyl-Epo B and Side Chain-modified Analogs

4.2.1 Total Synthesis of CP-Epothilone B



106

(*R*)-3-(*tert*-butyldimethylsilyloxy)dihydrofuran-2(3*H*)-one (106). To a solution of (*S*)-α-hydroxybutyrolactone (2.00 g, 19.59 mmol) and imidazole (2.94 g, 43.19 mmol) in DMF (15 mL) was added TBSCl (3.25 g, 21.56 mmol) at room temperature and the mixture was stirred for 22 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (15 mL). The aqueous phase was extracted with Et_2O (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 10:1) afforded 4.11 g (97%) of silyl ether **106** as a colorless oil which turned into a white solid upon storage at -20 °C.

TLC: R_f 0.36 (hexane/EtOAc 10:1, CPS).

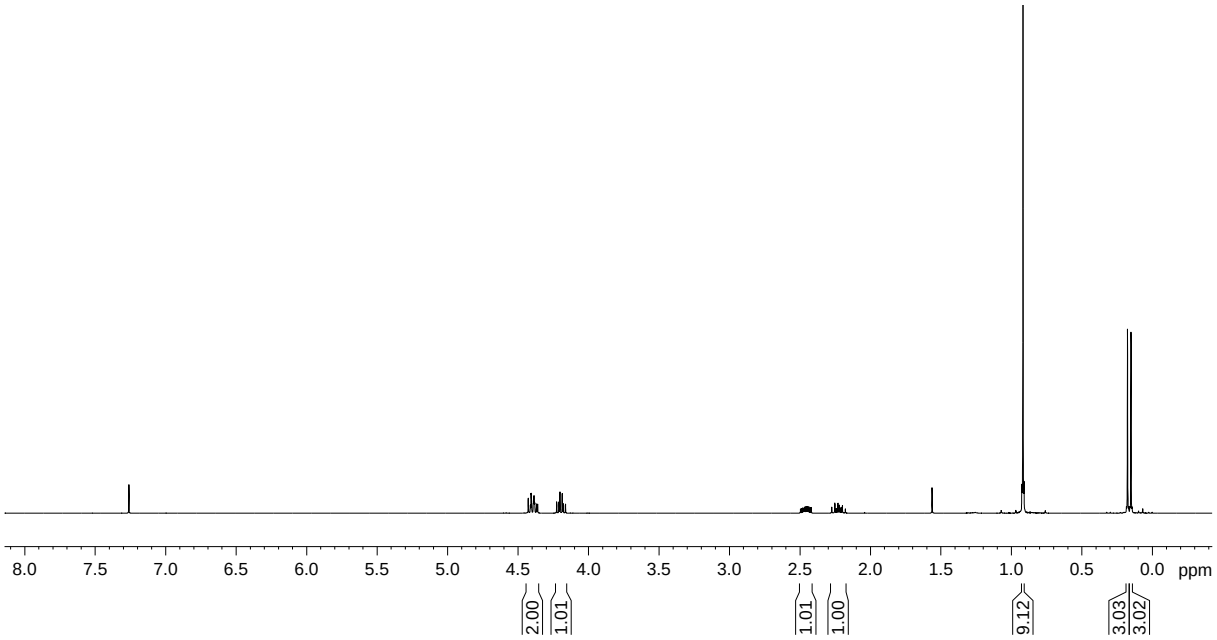
$[\alpha]^{20}_D$: = -32.6° (*c* = 0.33, $CHCl_3$).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.41 (dd, *J* = 8.4, 7.7 Hz, 1H), 4.39 (ddd, *J* = 9.1, 8.4, 3.4 Hz), 4.19 (td, *J* = 9.1, 6.4 Hz, 1H), 2.46 (dddd, *J* = 12.7, 7.7, 6.4, 3.4 Hz, 1H), 2.23 (dddd, *J* = 12.7, 9.1, 8.4, 8.4 Hz, 1H), 0.92 (s, 9H), 0.18 (s, 3H), 0.16 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 176.0, 68.4, 64.9, 32.5, 25.8, 18.4, -4.5, -5.1.

IR (film): ν □2954, 2931, 2858, 1786, 1469, 1359, 1255, 1220, 1152, 1109, 1021, 1000, 945, 840, 781, 698, 671.

HRMS (ESI): calculated for $C_{10}H_{20}O_3Si$ $[M+Na]^+$ 239.1074, found 239.1075.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7.0
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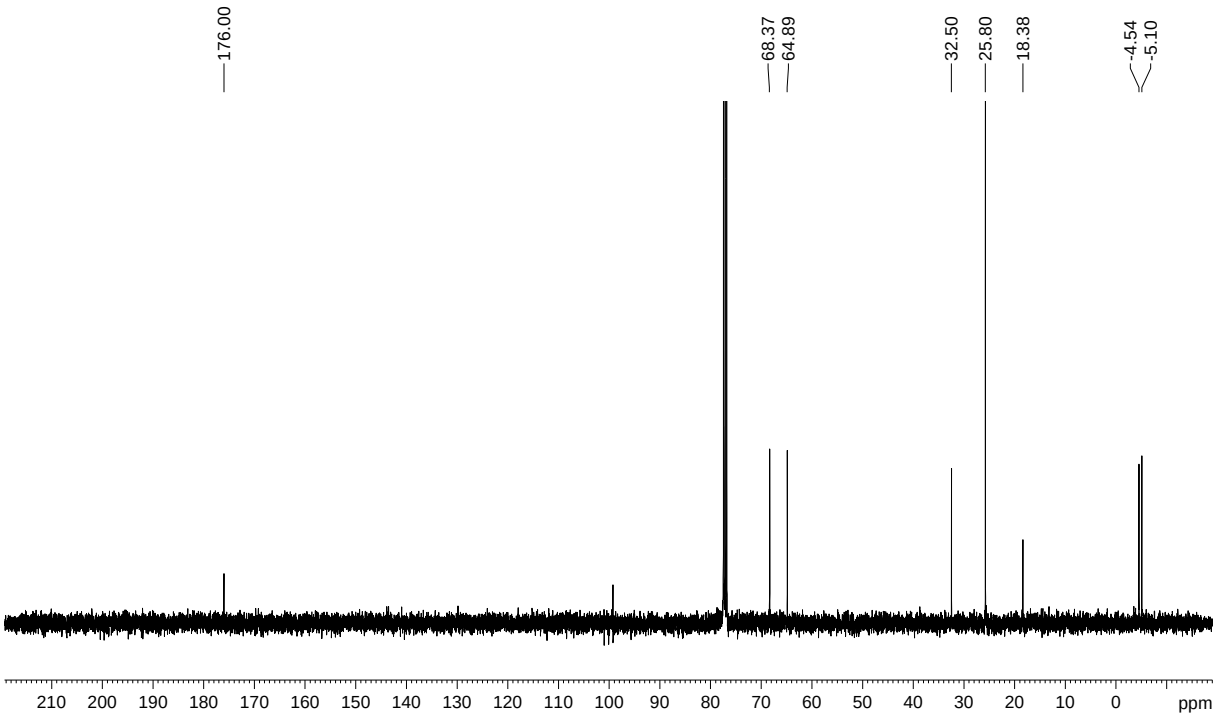
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
ppm
2.00
1.01
1.01
1.00
9.12
3.03
3.02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18.38
-4.54
-5.10
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200
190
180
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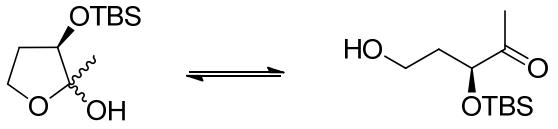
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm



107 **108**

(2*R*,3*R*)-3-(*tert*-butyldimethylsilyloxy)-2-methyltetrahydrofuran-2-ol (107).

(2*S*,3*S*)-3-(*tert*-butyldimethylsilyloxy)-2-methyltetrahydrofuran-2-ol (107).

(*S*)-3-(*tert*-butyldimethylsilyloxy)-5-hydroxypentan-2-one (108). To a solution of silyl ether **106** (2.94 g, 13.59 mmol) in THF (55 mL) was added MeLi (1.6 M in Et_2O, 10.19 mL, 16.31 mmol) dropwise at -78 °C and the reaction mixture was stirred for 3 h at this temperature. The cooling bath was removed and the reaction was quenched by addition of saturated aqueous NH_4Cl (30 mL). The mixture was diluted with saturated aqueous Rochelle salt (20 mL) and Et_2O (50 mL). The phases were separated and the aqueous phase was extracted with Et_2O (2 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 2.81 g (89%) of a mixture of cyclic acetal **107** and linear alcohol **108** as white crystals.

Note: No investigations have been carried out to assign the ^1H and ^{13}C-NMR signals to the single alcohols.

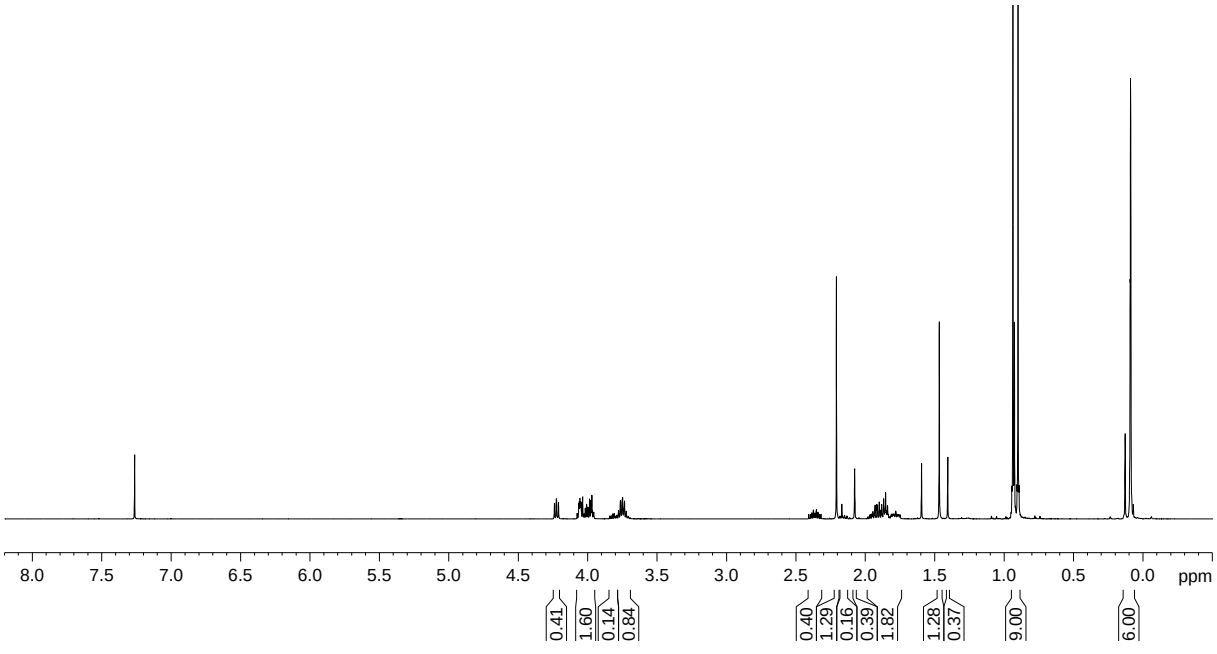
TLC: R_f 0.22 (hexane/EtOAc 5:1, CPS).

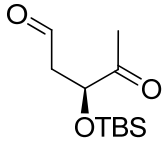
$[\alpha]^{20}_D$: +24.2° (*c* = 0.41, $CHCl_3$).

mp: 52-54 °C.

IR (film): ν 3418 br, 2955, 2930, 2858, 1717, 1472, 1464, 1376, 1254, 1109, 837, 776.

HRMS (ESI): calculated for $C_{11}H_{24}O_3Si$ $[M+Na]^+$ 255.1387, found 255.1397.





100

(*S*)-3-(*tert*-butyldimethylsilyloxy)-4-oxopentanal (100). To a solution of alcohols **107** and **108** (4.04 g, 17.38 mmol) in CH_2Cl_2 (50 mL) was added DMP (15% in CH_2Cl_2, 72.1 mL, 34.77 mmol) at room temperature over a period of 15 min whereupon the reaction mixture turned milky. The reaction mixture was stirred for 1.5 h at room temperature and the reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (30 mL) and saturated aqueous $Na_2S_2O_3$ (30 mL). Stirring was continued for 30 min, when two almost clear phases had formed. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were washed with brine (100 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 7:1) yielded 2.69 g (67%) of aldehyde **100** as a colorless oil.

Note: Because of its behaviour on silica, aldehyde **100** was difficult to purify to homogeneit. The remaining impurities were removed in the next step.

TLC: R_f 0.40 (hexane/EtOAc 4:1, CPS).

$[\alpha]^{20}_D$: -1.5° (*c* = 0.56, $CHCl_3$).

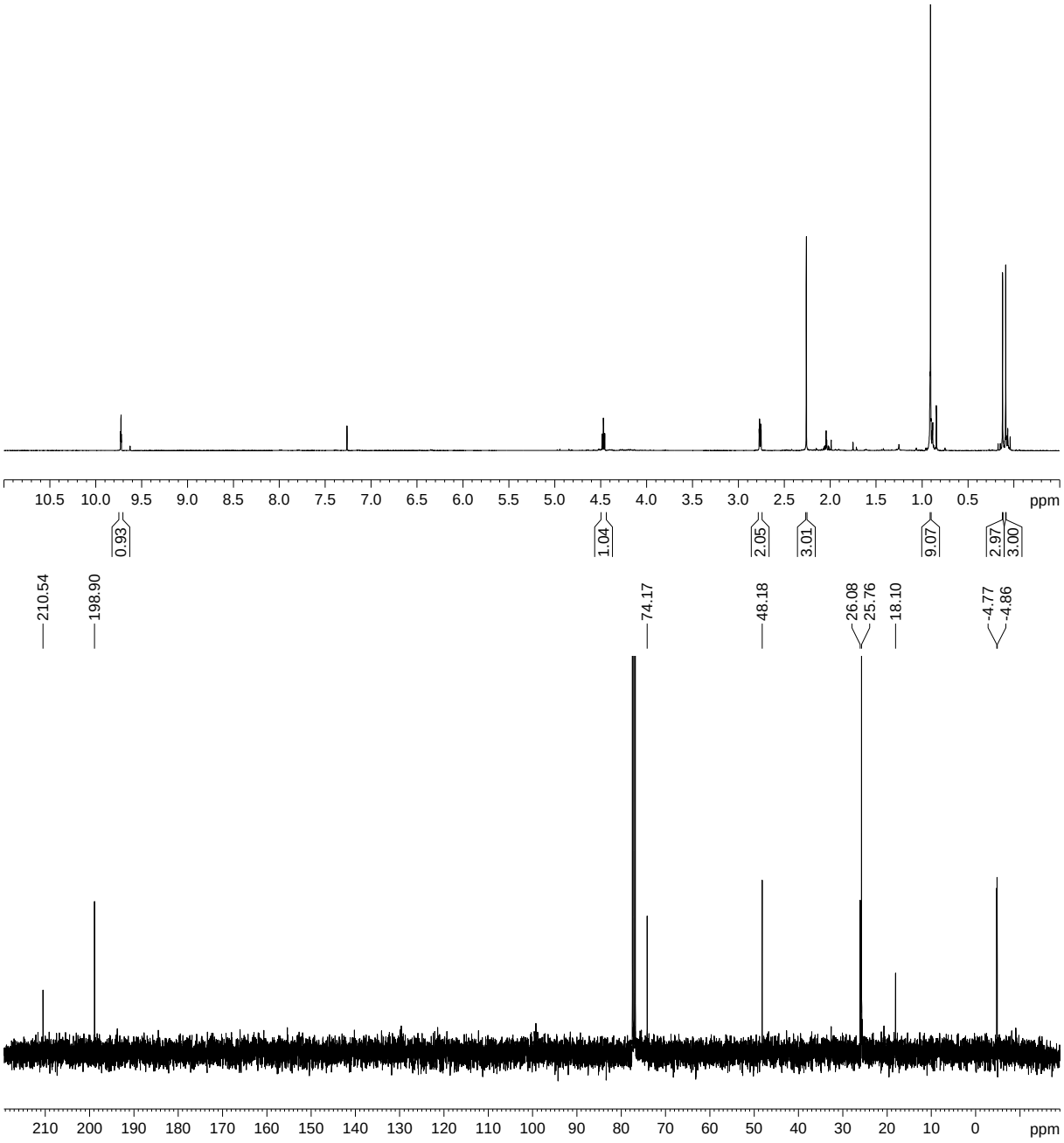
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.73 (t, *J* = 1.6 Hz, 1H), 4.47 (t, *J* = 5.6 Hz, 1H), 2.76 (ddd, *J* = 5.6, 1.6, 1.3 Hz, 1H), 2.75 (m, 1H), 2.26 (s, 3H), 0.91 (s, 9H), 0.12 (s, 3H), 0.09 (s, 3H).

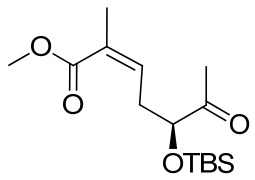
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 210.5, 198.9, 74.2, 48.2, 26.1, 25.8, 18.1, -4.8, -4.9.

IR (film): ν 2954, 2931, 2859, 1719, 1473, 1362, 1255, 1118, 1006, 938, 838, 779.

HRMS (ESI): calculated for $C_{11}H_{22}O_3Si$ $[M+]^+$ 230.1338, found 230.1562.

MS (ESI): calculated for $C_{11}H_{22}O_3Si$ $[M+Na]^+$ 253.1230, found 253.06.





110

(*S*,*Z*)-methyl 5-(*tert*-butyldimethylsilyloxy)-2-methyl-6-oxohept-2-enoate (110). To a solution of phosphonate **109** (3.58 g, 10.79 mmol) and 18-crown-6 (7.78 g, 29.43 mmol) in THF (120 mL) was added KHMDS 2.15 g, 10.79 mmol) at –78 °C in five portions over a period of 15 min. The solution was stirred for 30 min at this temperature and was then

transferred dropwise at -78 °C to a solution of aldehyde **100** (2.26 g, 9.81 mmol) in THF (30 mL) using a cannula. The reaction mixture was stirred for 1 h at this temperature and the reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (100 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to yield 2.09 g (72%) of unsaturated ester **110** as a colorless oil.

Note: The *Z*-configuration of the double bond was firmly established by NOESY-experiment.

TLC: R_f 0.57 (hexane/EtOAc 4:1, UV, CPS).

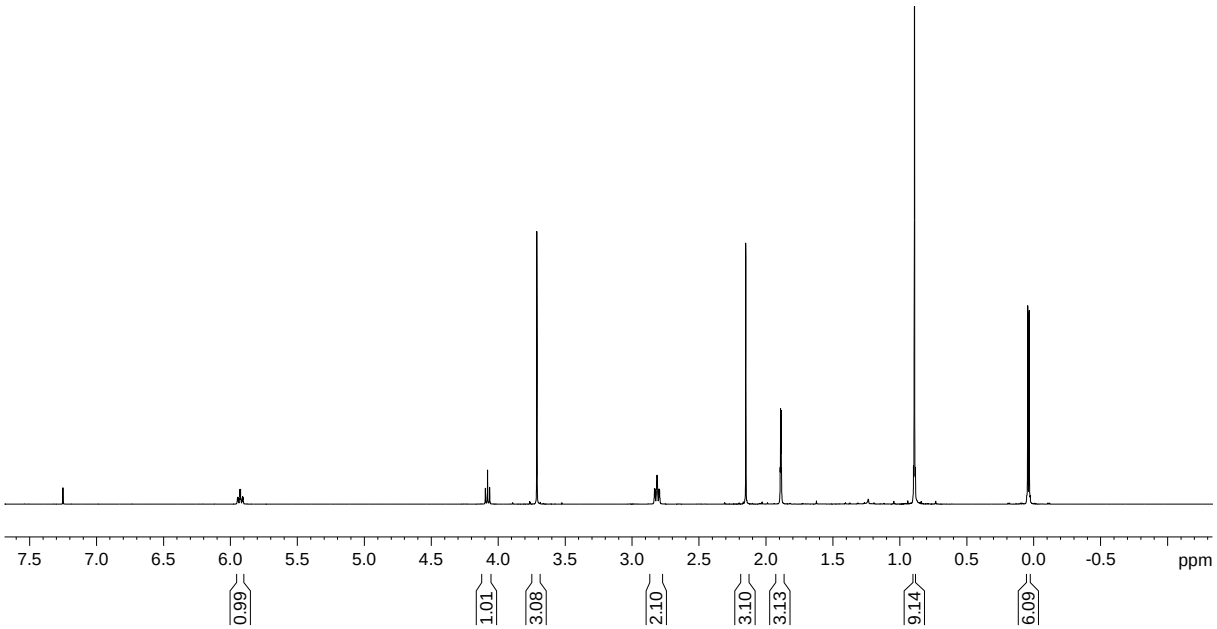
$[\alpha]^{20}_D$: = -11.8° (*c* = 0.75, $CHCl_3$).

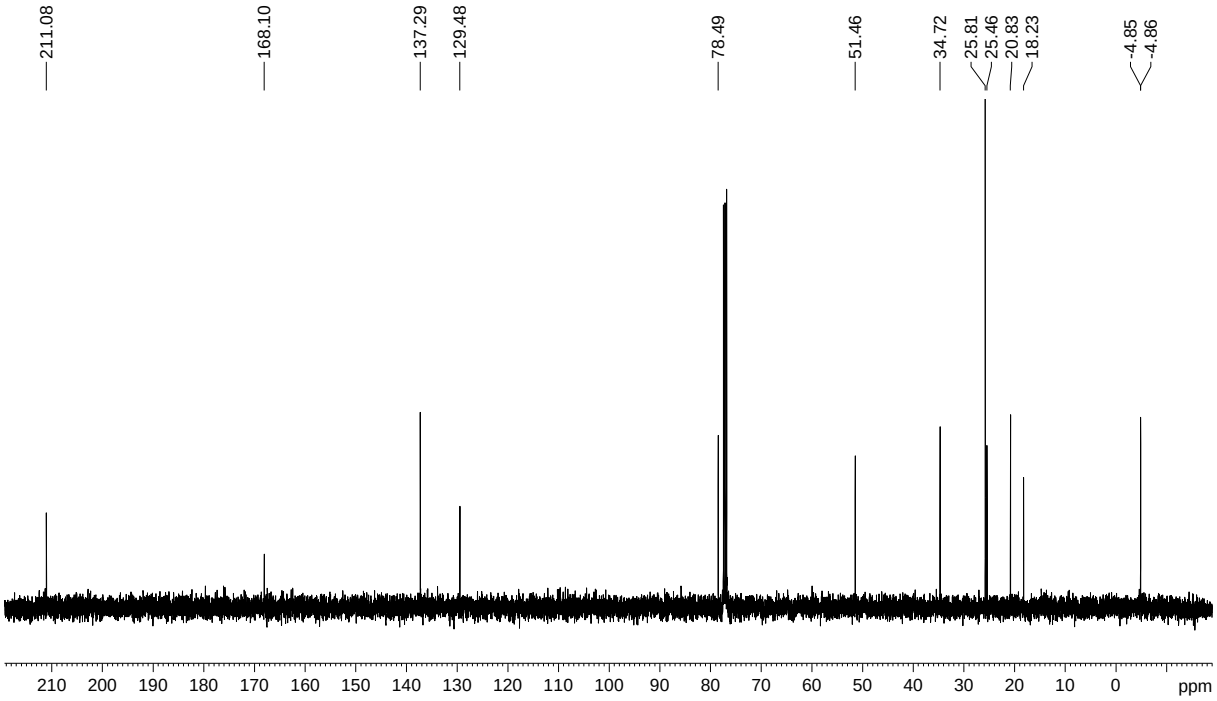
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.95 (tq, *J* = 7.4, 1.4 Hz, 1H), 4.10 (t, *J* = 6.3 Hz, 1H), 3.73 (s, 3H), 2.84 (ddq, *J* = 7.4, 6.3, 1.4 Hz, 2H), 2.17 (s, 3H), 1.91 (dt, *J* = 1.4, 1.4 Hz, 3H), 0.91 (s, 9H), 0.07 (s, 3H), 0.06 (s, 3H).

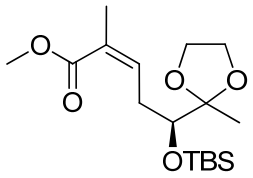
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 211.3, 168.1, 137.3, 129.5, 78.5, 51.5, 34.7, 25.8, 25.5, 20.8, 18.2, -4.8, -4.8.

IR (film): ν 2954, 2931, 2858, 1718, 1457, 1362, 1253, 1219, 1108, 839, 776, 669.

HRMS (ESI): calculated for $C_{15}H_{28}O_4Si$ $[M+Na]^+$ 323.1649, found 323.1645.







113

(*S*,*Z*)-methyl 5-(*tert*-butyldimethylsilyloxy)-2-methyl-5-(2-methyl-1,3-dioxolan-2-yl)pent-2-enoate (113). To a solution of ketone **110** (4.15 g, 13.81 mmol) in ethylene glycol (21 mL) and triethyl orthoformate (21 mL) was added *p*-TSA (53 mg, 0.28 mmol) at room temperature. The reaction mixture was then heated to 40 °C and stirred for 2 h at this temperature. The reaction mixture was then allowed to reach room temperature and the reaction was cautiously quenched by addition of saturated aqueous $NaHCO_3$ (20 mL). The reaction mixture was diluted with Et_2O (30 mL), the phases were separated and the aqueous phase was extracted with Et_2O (2 x 30 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 10:1) gave 4.62 g (97%) of acetal **113** as a colorless oil.

Note: Since ketone **113** and acetal **110** show the same R_f-value, the reaction was followed by MS analysis.

TLC: R_f 0.57 (hexane/EtOAc 4:1, UV, CPS).

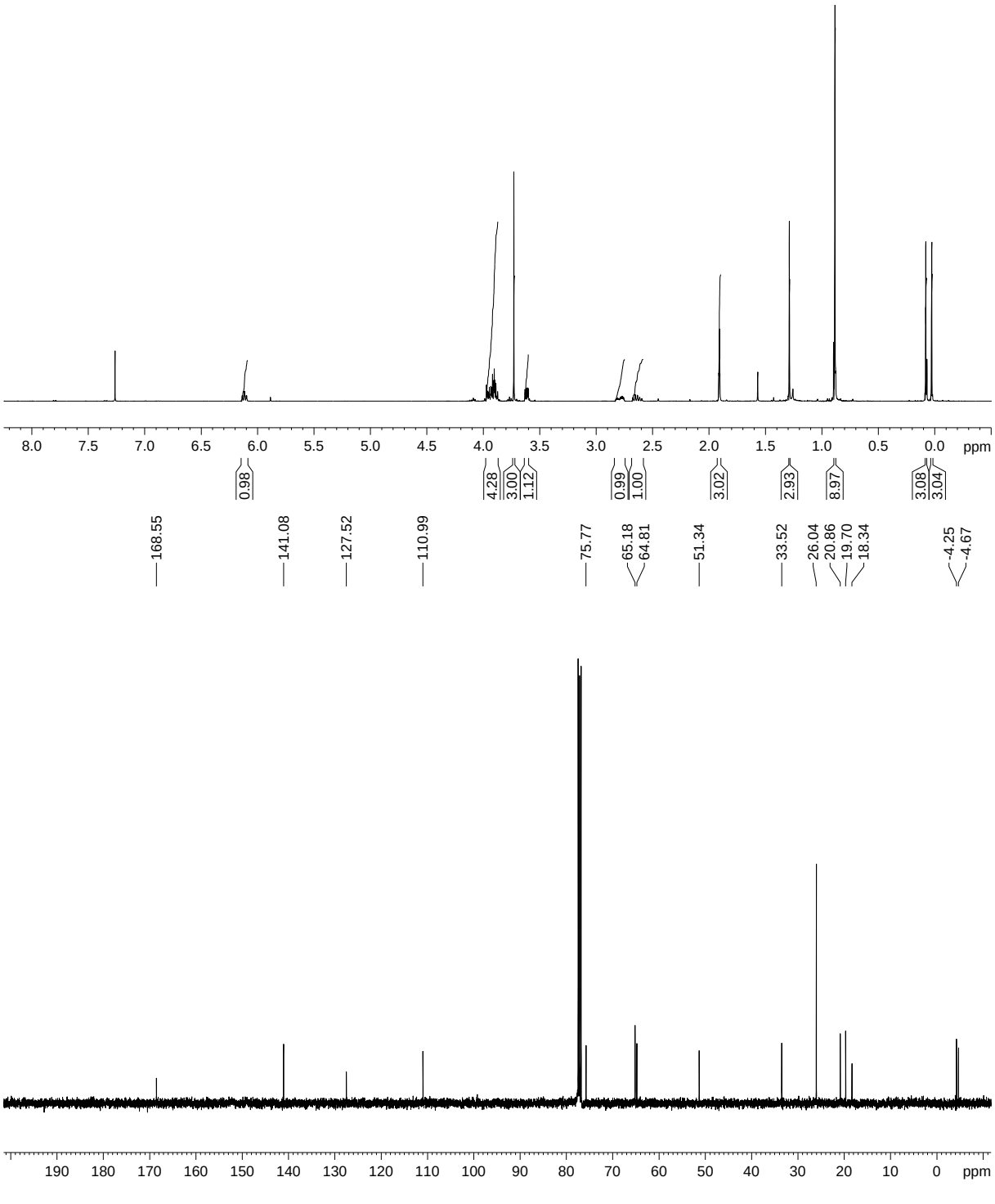
$[\alpha]^{20}_D$: = -27.9° (*c* = 0.65, $CHCl_3$).

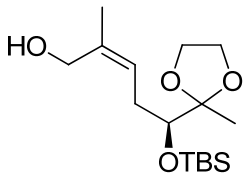
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.12 (tq, *J* = 7.3, 1.4 Hz, 1H), 3.98-3.85 (m, 4H), 3.73 (s, 3H), 3.62 (dd, *J* = 8.1, 3.8 Hz, 1H), 2.79 (dddq, *J* = 15.6, 7.3, 3.8, 1.4 Hz, 1H), 2.63 (dddq, *J* = 15.6, 8.1, 7.3, 1.4 Hz, 1H), 1.91 (dt, *J* = 1.4, 1.4 Hz, 3H), 1.29 (s, 3H), 0.88 (s, 9H), 0.08 (s, 3H), 0.03 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 168.5, 141.1, 127.5, 111.0, 75.8, 65.2, 64.8, 51.3, 33.5, 26.0, 20.9, 19.7, 18.3, -4.3, -4.7.

IR (film): ν 2954, 2931, 2857, 1717, 1457, 1362, 1253, 1219, 1105, 835, 776, 669.

HRMS (ESI): calculated for $C_{17}H_{32}O_5Si$ $[M+Na]^+$ 367.1911, found 367.1925.





99

(*S*,*Z*)-5-(*tert*-butyldimethylsilyloxy)-2-methyl-5-(2-methyl-1,3-dioxolan-2-yl)pent-2-en-1-ol (99). To a solution of ester **113** (4.45 g, 12.92 mmol) in CH_2Cl_2 (200 mL) was added DIBAL-H (1 M in CH_2Cl_2, 28.4 mL, 28.4 mmol) dropwise at -78 °C and the reaction mixture was stirred for 30 min at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (250 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 150 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 3.95 g (97%) of allylic alcohol **99** as a colorless oil.

TLC: R_f 0.29 (hexane/EtOAc 4:1, CPS).

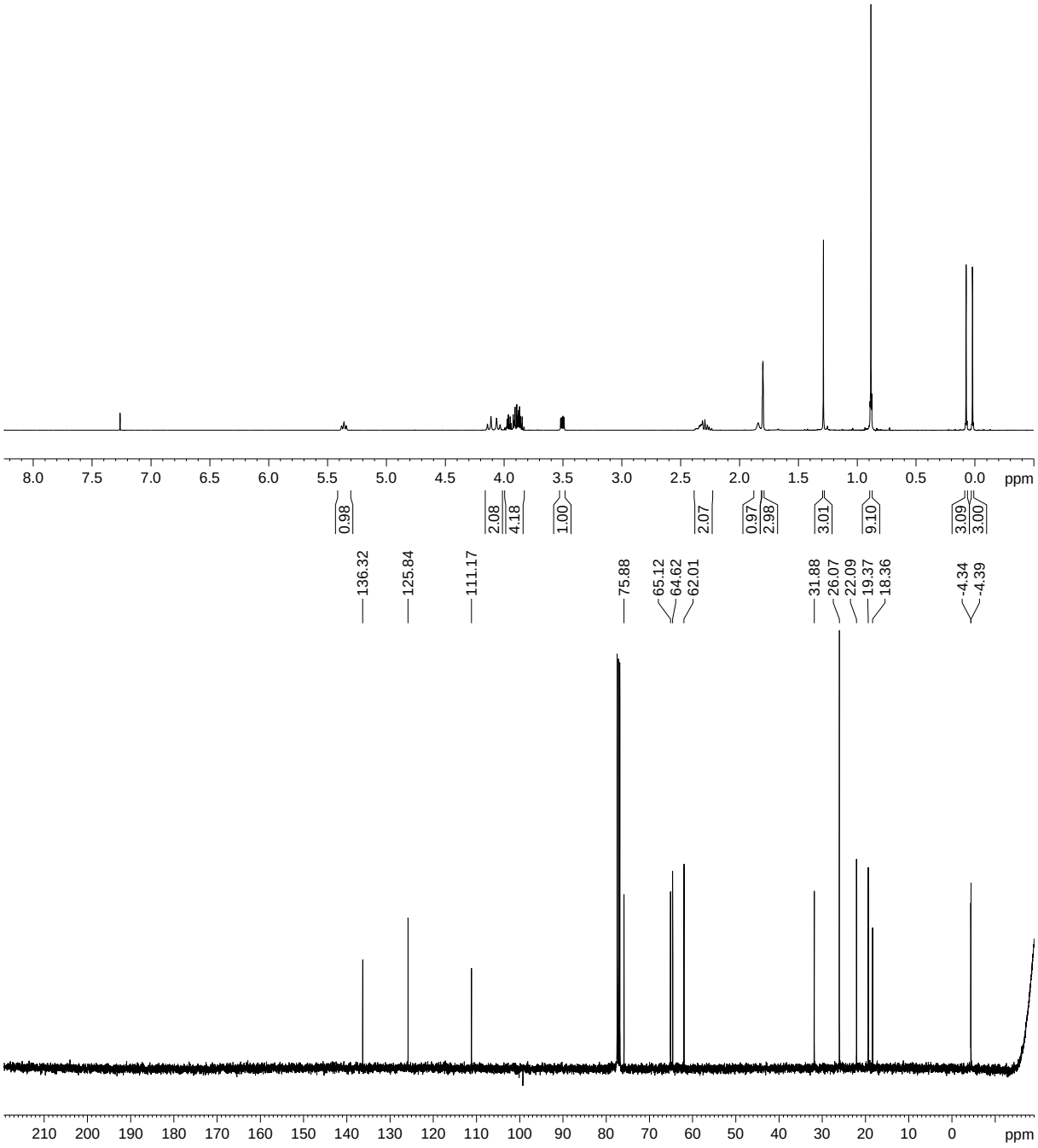
$[\alpha]^{20}_D$: = -2.1° (*c* = 0.69, $CHCl_3$).

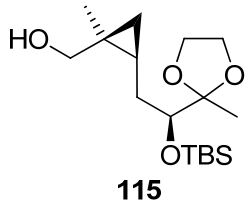
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.36 (tq, *J* = 7.8, 1.2 Hz, 1H), 4.13 (d, *J* = 11.9 Hz, 1H), 4.05 (d, *J* = 11.9 Hz, 1H), 4.00-3.83 (m, 4H), 3.50 (dd, *J* = 8.0, 3.8 Hz, 1H), 2.39-2.23 (m, 2H), 1.84 (br s, 1H), 1.80 (dt, *J* = 1.2, 1.2 Hz, 3H), 1.29 (s, 3H), 0.88 (s, 9H), 0.07 (s, 3H), 0.02 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 136.3, 125.8, 111.2, 75.8, 65.1, 64.6, 62.0, 31.9, 26.1, 22.1, 19.4, 18.4, -4.3, -4.4.

IR (film): ν 3420 br, 2955, 2930, 2856, 1473, 1253, 1219, 1106, 1057, 1006, 949, 835, 776.

HRMS (ESI): calculated for $C_{16}H_{32}O_4Si$ $[M+Na]^+$ 339.1962, found 339.1964.





115

((1*S*,2*S*)-2-((*S*)-2-(*tert*-butyldimethylsilyloxy)-2-(2-methyl-1,3-dioxolan-2-yl)ethyl)-1-methylcyclopropyl)methanol (115). To a solution of Et_2Zn (1 M in hexane, 36.97 mL, 36.97 mmol) in CH_2Cl_2 (80 ml) at 0 °C was added CH_2I_2 (5.96 mL, 73.93 mmol) over a period of 15 min (the interior temperature rose from 0 °C to 2.5 °C). The milky suspension

was stirred for 10 min at this temperature and a preformed solution of (+)-(*R,R*)-2-butyl-*N,N,N',N'*-tetramethyl-1,3,2-dioxaborolane-4,5-dicarboxamide (Charette ligand) (1.86 mL, 7.39 mmol) and allylic alcohol **99** (1.95 g, 6.16 mmol) in CH_2Cl_2 (100 ml) was rapidly added via syringe, whereupon the reaction mixture turned clear. The solution was allowed to reach room temperature and stirred for 1.5 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (150 ml), the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 150 ml). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1 → 5:1) to afford 1.79 g (88%, dr 18:1) of cyclopropyl alcohol **115** as a single isomer as a colorless oil.

Note: Since allylic alcohol **99** and cyclopropyl alcohol **115** show the same R_f-value, the reaction was followed by MS analyis.

According to ref. the exothermicity of the formation of $Zn(CH_2I)_2$ the above procedure sometimes led to violent explosions on a larger scale (8 mmol). In our experience, if the interior temperature is carefully monitored during addition of CH_2I_2 to the Et_2Zn solution, the reaction can be conducted on a larger scale (>> 8 mmol) safely.

TLC: R_f 0.29 (hexane/EtOAc 4:1, CPS).

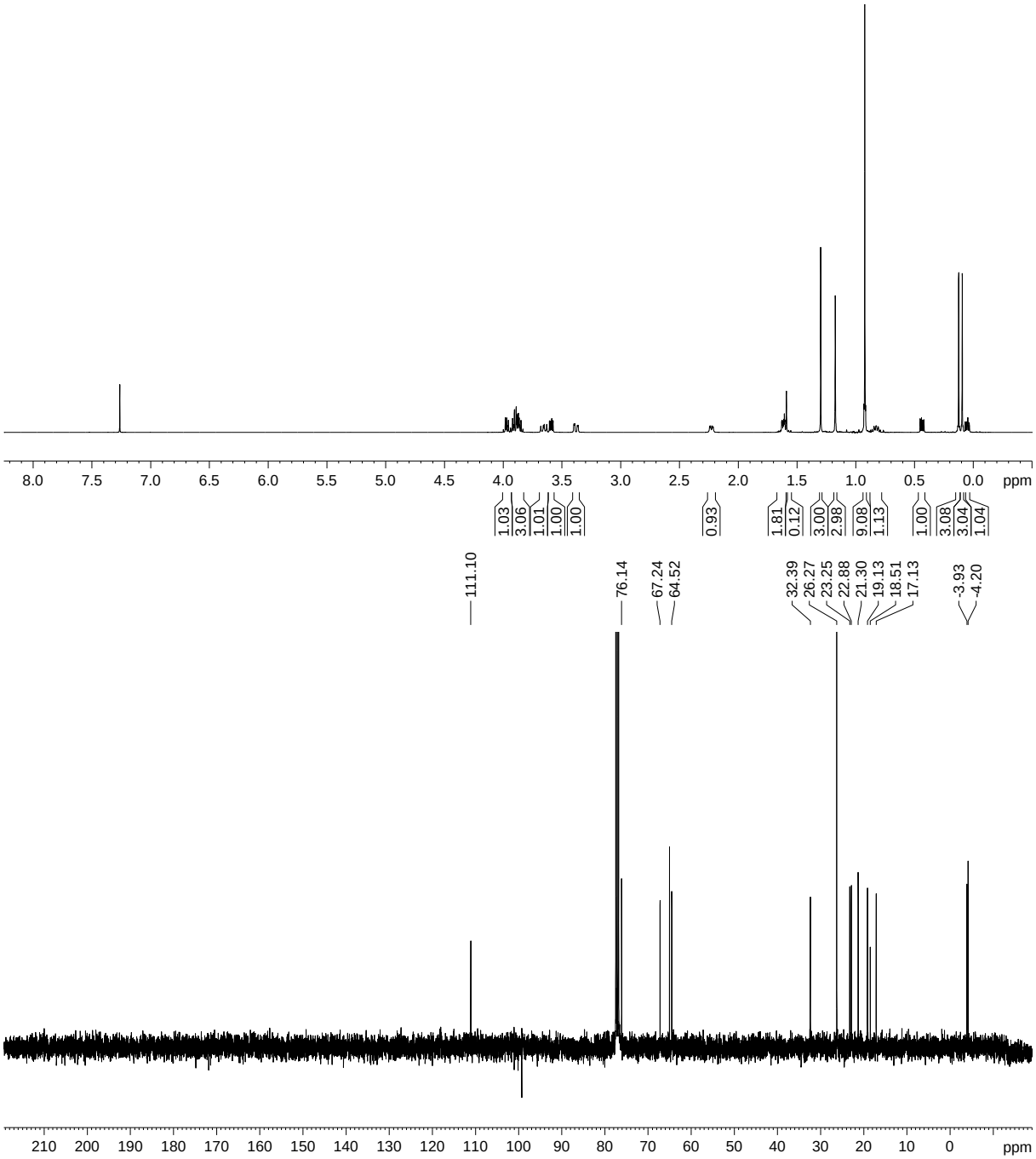
$[\alpha]^{20}_D$: = -45.3° (*c* = 0.57, $CHCl_3$).

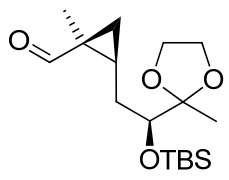
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.00-3.93 (m, 1H), 3.93-3.83 (m, 3H), 3.66 (dd, *J* = 11.7, 8.7 Hz, 1H), 3.59 (dd, *J* = 7.6, 4.4 Hz, 1H), 3.38 (dd, *J* = 11.7, 2.7 Hz, 1H), 2.23 (dd, *J* = 8.7, 3.2 Hz, 1H), 1.67-1.55 (m, 2H), 1.30 (s, 3H), 1.17 (s, 3H), 0.92 (s, 9H), 0.88-0.77 (m, 1H), 0.44 (dd, *J* = 8.5, 4.5 Hz, 1H), 0.13 (s, 3H), 0.10 (s, 3H), 0.05 (dd, *J* = 5.5, 4.5 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 111.0, 76.1, 67.2, 65.2, 64.5, 32.4, 26.3, 23.2, 22.9, 21.3, 19.1, 18.5, 17.1, -3.9, -4.2.

IR (film): ν □3433 br, 2954, 2931, 2858, 1469, 1379, 1252, 1107, 1033, 834, 776.

HRMS (ESI): calculated for $C_{17}H_{34}O_4Si$ $[M+Na]^+$ 353.2119, found 353.2114.





116

(1*S*,2*S*)-2-((*S*)-2-(*tert*-butyldimethylsilyloxy)-2-(2-methyl-1,3-dioxolan-2-yl)ethyl)-1-methylcyclopropanecarbaldehyde (116). To a solution of oxalyl chloride (36 µL, 0.42 mmol) in CH_2Cl_2 (0.5 mL) was added DMSO (80 µL, 1.13 mmol) dropwise at -78 °C. The

reaction mixture was stirred for 5 min at this temperature and alcohol **115** (93 mg, 0.28 mmol) in CH_2Cl_2 (0.5 mL) was added. Stirring was continued for 15 min at -78 °C and Et_3N (136 µL, 0.98 mmol) was added. The cooling bath was removed and the reaction mixture was stirred for 2.5 h at room temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (1 mL), the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 2mL). The combined organic phases were washed with brine (5 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 10:1) afforded 91 mg (98%) of aldehyde **116** as a colorless oil.

TLC: R_f 0.34 (hexane/EtOAc 8:1, CPS).

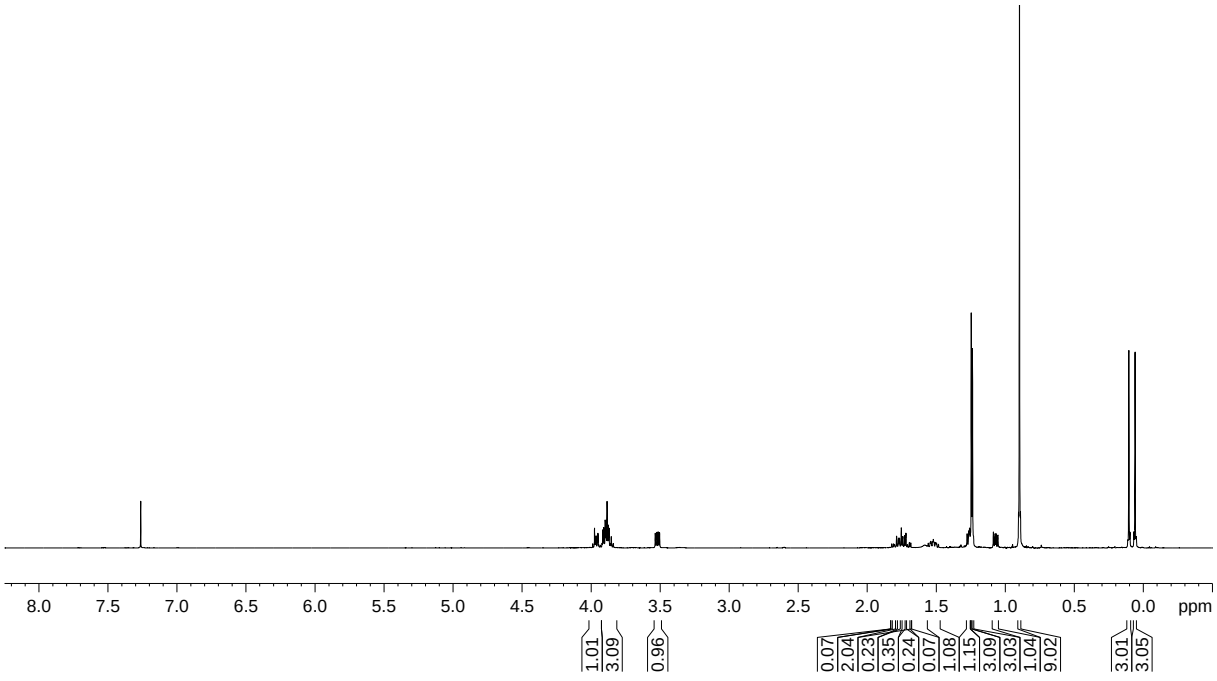
$[\alpha]^{20}_D$: = -75.2° (*c* = 0.71, $CHCl_3$).

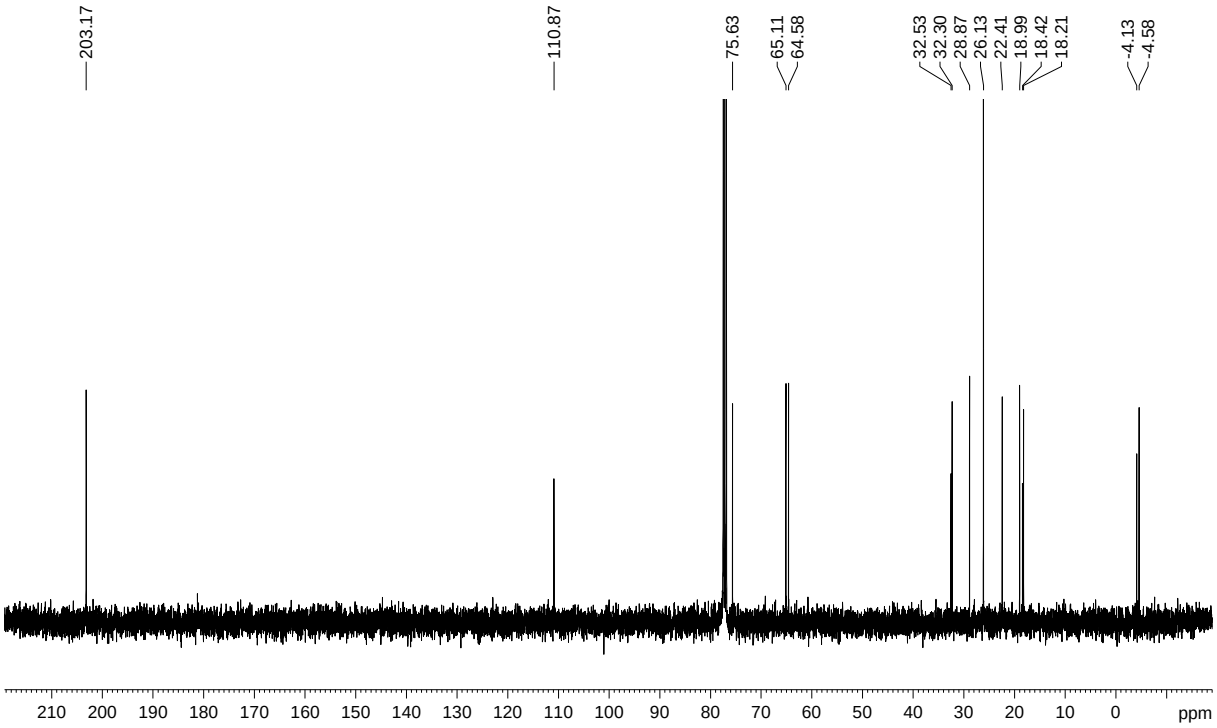
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.17 (s, 1H), 3.99-3.93 (m, 1H), 3.92-3.84 (m, 3H), 3.52 (dd, *J* = 8.2, 4.1 Hz, 1H), 1.79 (ddd, *J* = 13.6, 8.2, 5.4 1H), 1.72 (ddd, *J* = 13.6, 9.2, 4.1 Hz, 1H), 1.52 (dddd, *J* = 9.2, 8.0, 7.0, 5.4 Hz, 1H), 1.26 (dd, *J* = 7.0, 4.9 Hz, 1H), 1.25 (s, 3H), 1.24 (s, 3H), 1.07 (dd, *J* = 8.0, 4.9 Hz, 1H), 0.90 (s, 9H), 0.11 (s, 3H), 0.06 (s, 3H).

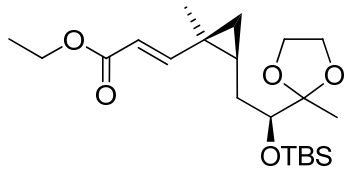
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 203.2, 110.9, 75.6, 65.1, 64.6, 32.5, 32.3, 28.9, 26.1, 22.4, 19.0, 18.4, 18.2, -4.1, -4.6.

IR (film): ν□2955, 2931, 2886, 2858, 2735, 1706, 1469, 1381, 1252, 1107, 1053, 940, 833, 777.

HRMS (ESI): calculated for $C_{17}H_{32}O_4Si$ $[M+Na]^+$ 351.1962, found 351.1956.







117

(*E*)-ethyl 3-((1*S*,2*S*)-2-((*S*)-2-(*tert*-butyldimethylsilyloxy)-2-(2-methyl-1,3-dioxolan-2-yl)ethyl)-1-methylcyclopropyl)acrylate (117). To a solution of aldehyde **116** (3.11 g, 9.47 mmol) in CH_2Cl_2 (72 mL) was added Ph_3PCHCO_2Et (6.60 g, 18.93 mmol) in one portion at room temperature and the reaction mixture was heated to reflux for 72 h. Additional Ph_3PCHCO_2Et (2.48 g, 7.10 mmol) was then added in one portion and the reaction mixture was heated to reflux for 24 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash chromatography (hexane/EtOAc 15:1) to furnish 3.72 g (99%) of unsaturated ester **117** as a white solid.

TLC: R_f 0.42 (hexane/EtOAc 8:1, UV, CPS).

$[\alpha]^{20}_D$: = -66.4° (*c* = 0.53, $CHCl_3$).

mp: 61-63 °C.

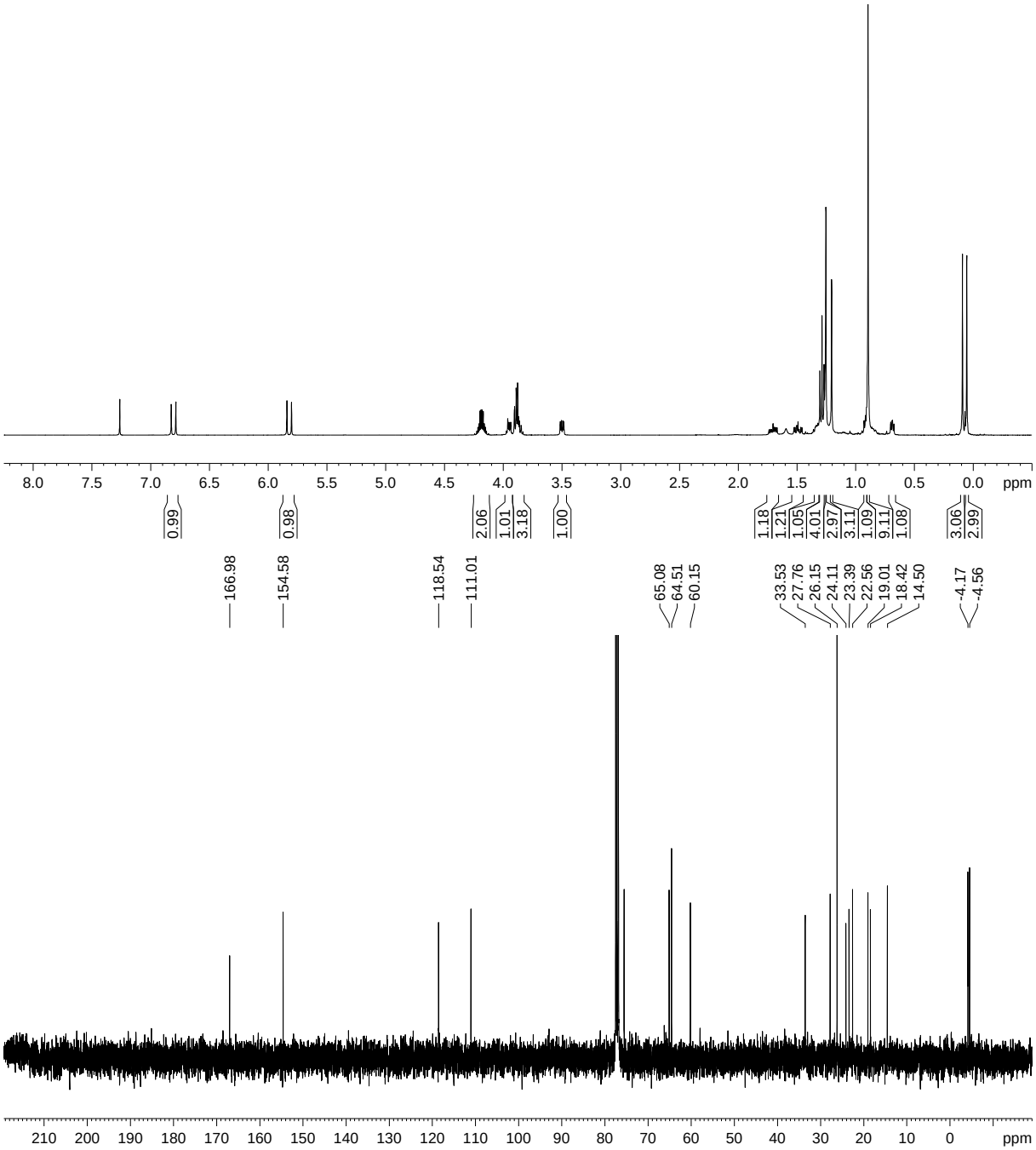
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.81 (d, *J* = 15.6 Hz, 1H), 5.82 (d, *J* = 15.6 Hz, 1H), 4.24-4.13 (m, 2H), 3.98-3.93 (m, 1H), 3.91-3.83 (m, 3H), 3.50 (dd, *J* = 8.7, 3.8 Hz, 1H), 1.70 (ddd, *J* = 13.7, 8.7, 4.8 Hz, 1H), 1.50 (ddd, *J* = 13.7, 9.7, 3.8, 1H), 1.32 (dddd, *J* = 9.7, 8.4, 6.3,

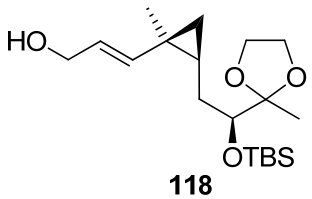
4.8 Hz, 1H), 1.29 (t, *J* = 7.1 Hz, 3H), 1.25 (s, 3H), 1.21 (s, 3H), 0.91 (dd, *J* = 8.4, 4.7 Hz, 1H), 0.90 (s, 9H), 0.69 (dd, *J* = 6.3, 4.7 Hz, 1H), 0.09 (s, 3H), 0.06 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 167.0, 154.6, 118.5, 111.0, 75.5, 65.1, 64.5, 60.2, 33.5, 27.8, 26.2, 24.1, 23.4, 22.6, 19.1, 18.4, 14.5, -4.2, -4.6.

IR (film): ν 2954, 2932, 2886, 2858, 1715, 1637, 1468, 1380, 1251, 1169, 1106, 1043, 947, 834, 776.

HRMS (ESI): calculated for $C_{21}H_{38}O_5Si$ $[M+Na]^+$ 421.2381, found 421.2370.





118

(*E*)-3-((1*S*,2*S*)-2-((*S*)-2-(*tert*-butyldimethylsilyloxy)-2-(2-methyl-1,3-dioxolan-2-yl)ethyl)-1-methylcyclopropyl)prop-2-en-1-ol (118). To a solution of ester **117** (158 mg, 0.46 mmol) in CH_2Cl_2 (7 mL) was added DIBAL-H (1 M in CH_2Cl_2, 1.01 mL, 1.01 mmol) dropwise at -78 °C. The reaction mixture was stirred for 30 min at this temperature and the reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (10 mL). The solution was allowed to warm to room temperature and was left stirring vigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 10 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to give 142 mg (98%) of allylic alcohol **118** as a white solid.

TLC: R_f 0.41 (hexane/EtOAc 2:1, CPS).

$[\alpha]^{20}_D$: = -65.8° (*c* = 0.60, $CHCl_3$).

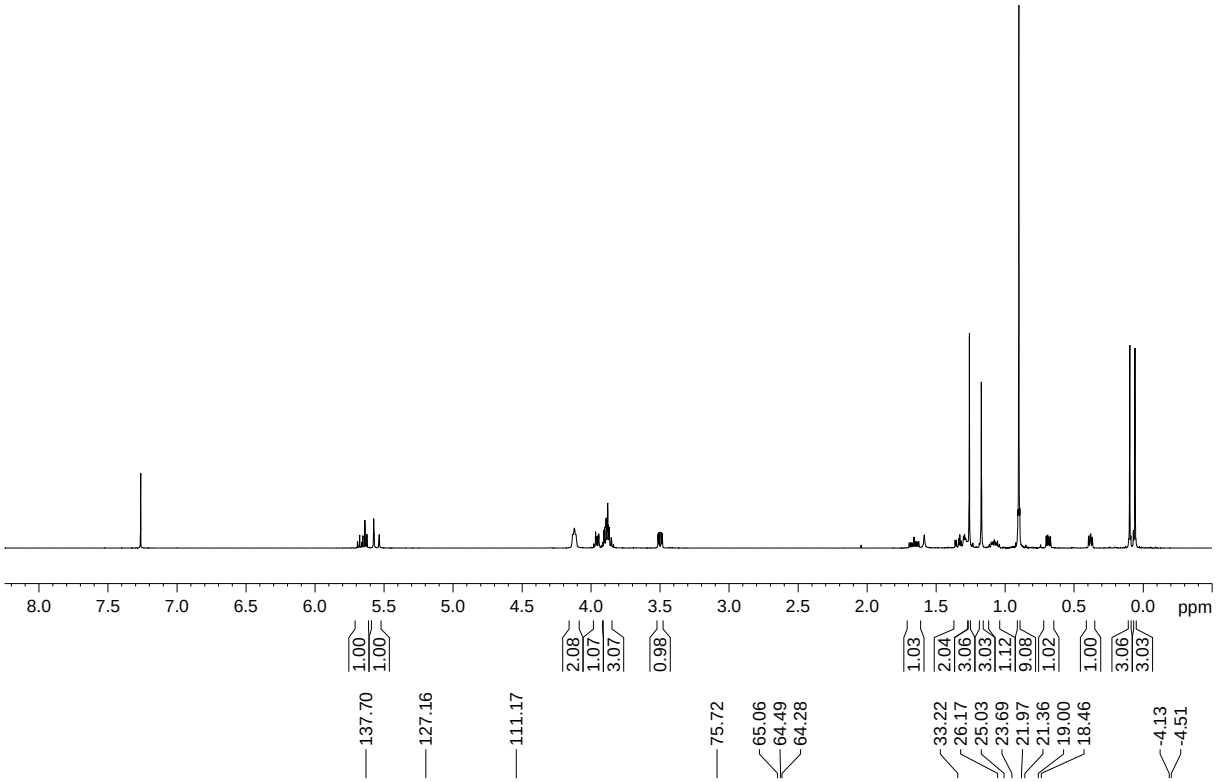
mp: 92-93 °C.

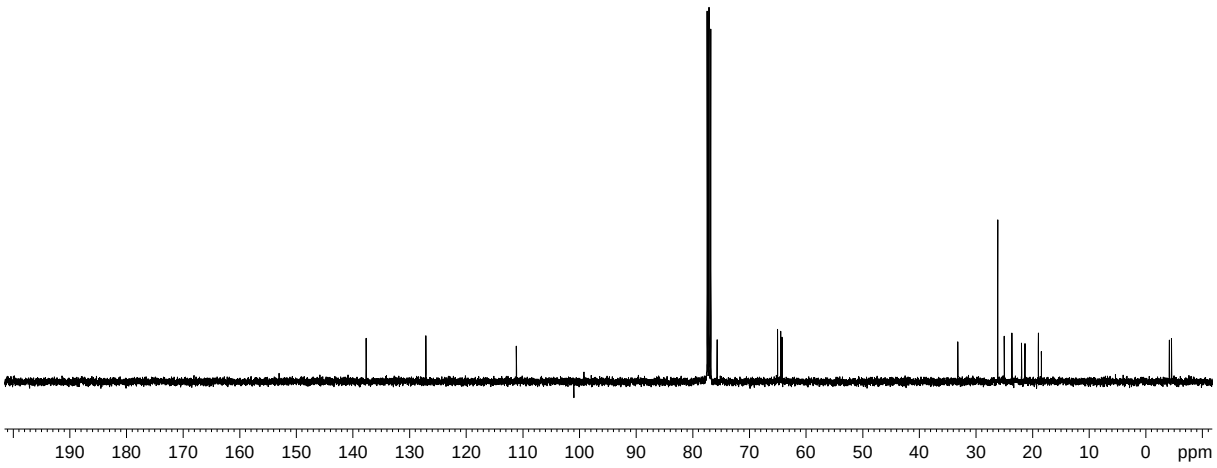
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.66 (dt, *J* = 15.4, 6.0 Hz, 1H), 5.55 (dt, *J* = 15.4, 1.0 Hz, 1H), 4.12 (t, *J* = 4.5 Hz, 2H), 3.99-3.92 (m, 1H), 3.92-3.84 (m, 3H), 3.50 (dd, *J* = 8.9, 3.3 Hz, 1H), 1.66 (ddd, *J* = 13.7, 8.9, 4.6 Hz, 1H), 1.33 (ddd, *J* = 13.7, 9.9, 3.3 Hz, 1H), 1.29 (br s, 1H), 1.26 (s, 3H), 1.17 (s, 3H), 1.08 (dddd, *J* = 9.9, 8.2, 5.9, 4.6 Hz, 1H), 0.90 (s, 9H), 0.69 (dd, *J* = 8.2, 4.4 Hz, 1H), 0.38 (dd, *J* = 5.9, 4.4 Hz, 1H), 0.10 (s, 3H), 0.06 (s, 3H).

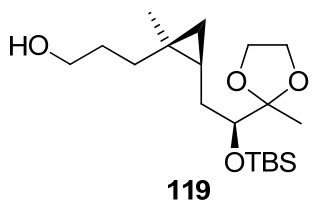
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 137.7, 127.2, 111.2, 75.7, 65.1, 64.5, 64.3, 33.2, 26.2, 25.0, 23.7, 22.0, 21.4, 19.0, 18.5, -4.1, -4.5.

IR (film): ν 3483, 2952, 2929, 2888, 2858, 1653, 1461, 1377, 1252, 1159, 1114, 1064, 971, 837, 776.

HRMS (ESI): calculated for $C_{19}H_{36}O_4Si$ $[M+Na]^+$ 379.2275, found 379.2285.







119

3-((1*S*,2*S*)-2-((*S*)-2-(*tert*-butyldimethylsilyloxy)-2-(2-methyl-1,3-dioxolan-2-yl)ethyl)-1-methylcyclopropyl)propan-1-ol (119). To a solution of allylic alcohol **118** (2.46 g, 6.90 mmol) in MeOH (125 ml) was added $CoCl_2 \bullet 6H_2O$ (1.31 g, 5.51 mmol) in one portion at room temperature and the red solution was left stirring for 25 min at this temperature. $NaBH_4$

(3.13 g, 82.79 mmol) in DMF (50 mL) was then added dropwise whereupon the solution turned black. Stirring was continued for 3 h at room temperature and the reaction was then quenched by addition of water (100 ml). The aqueous phase was extracted with CH_2Cl_2 (3 x 150 ml), the combined organic phases were washed with brine (250 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 5:1) yielded 2.34 g (95%) of alcohol **119** as a white solid.

TLC: R_f 0.41 (hexane/EtOAc 2:1, CPS).

$[\alpha]^{20}_D$: = -54.7° (*c* = 0.35, $CHCl_3$).

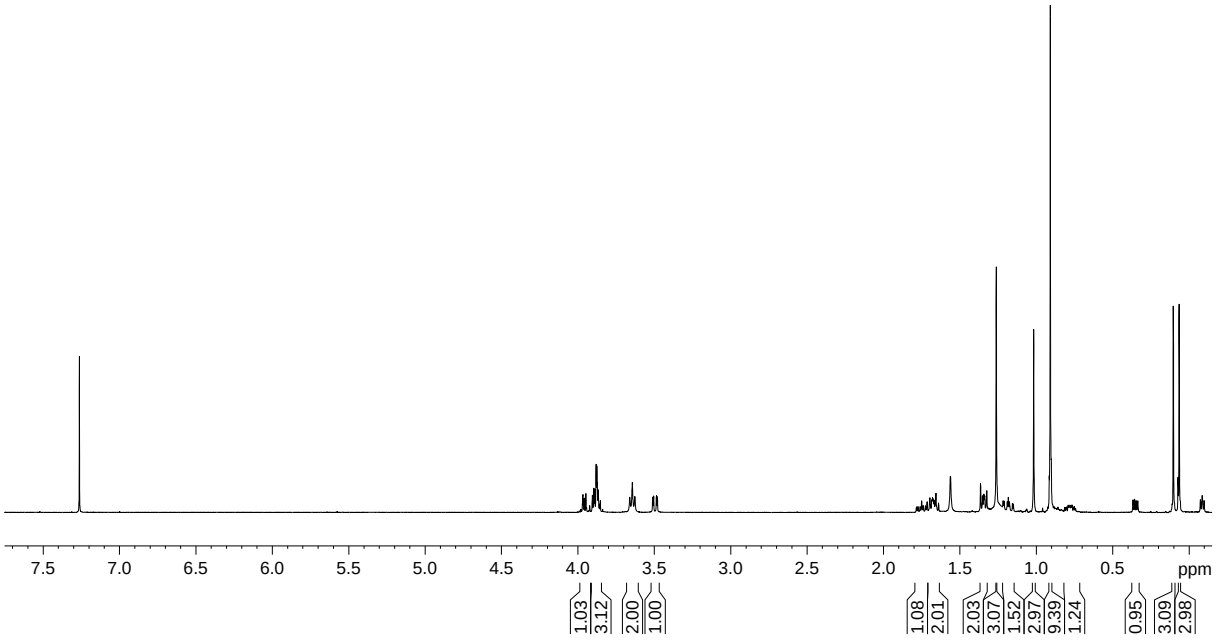
mp: 60-62 °C.

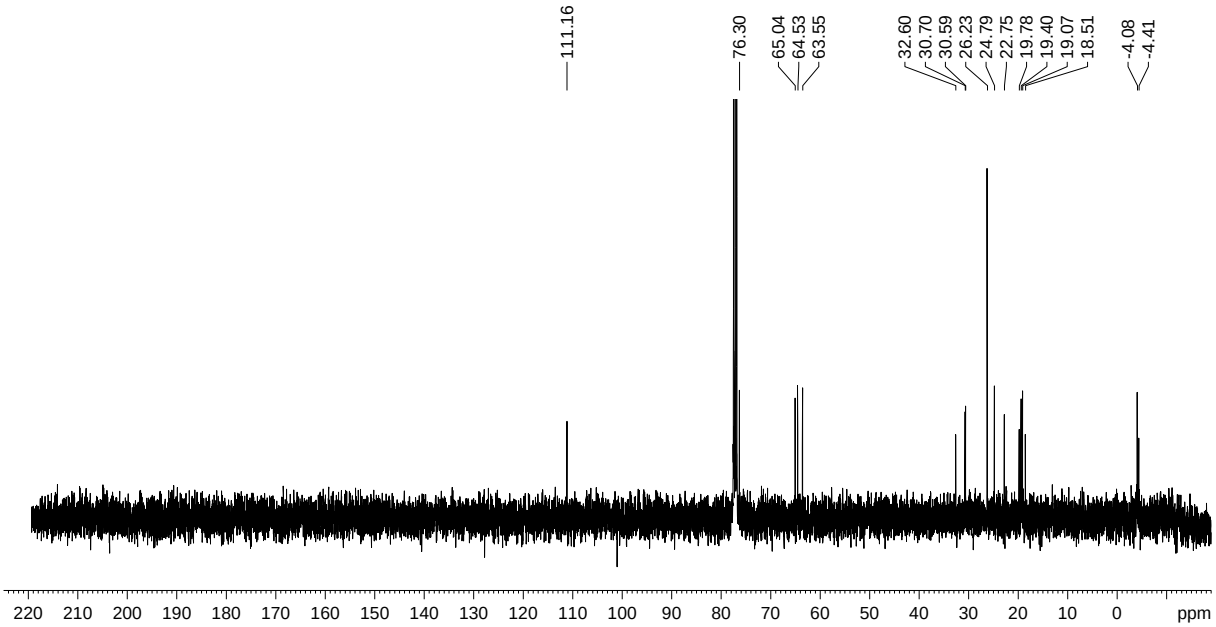
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 3.99-3.92 (m, 1H), 3.92-3.85 (m, 3H), 3.64 (t, *J* = 6.7 Hz, 2H), 3.50 (dd, *J* = 9.5, 2.5 Hz, 1H), 1.75 (ddd, *J* = 13.6, 9.6, 3.9 Hz, 1H), 1.70-1.63 (m, 2H), 1.37-1.32 (m, 2H), 1.26 (s, 3H), 1.18 (ddd, *J* = 13.6, 10.5, 2.5 Hz, 1H), 1.02 (s, 3H), 0.91 (s, 9H), 0.78 (dddd, *J* = 10.5, 8.4, 5.4, 3.9 Hz, 1H), 0.35 (dd, *J* = 8.4, 4.1 Hz, 1H), 0.10 (s, 3H), 0.07 (s, 3H), -0.09 (dd, *J* = 5.4, 4.1 Hz, 1H).

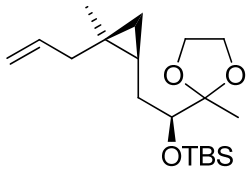
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 111.2 76.3, 65.0, 64.5, 63.6, 32.6, 30.7, 30.5, 26.2, 24.8, 22.8, 19.8, 19.4, 19.1, 18.5, -4.1, -4.4.

IR (film): ν 3389, 2952, 2929, 2857, 1468, 1380, 1251, 1163, 1106, 1054, 834, 775.

HRMS (ESI): calculated for $C_{19}H_{38}O_4Si$ $[M+Na]^+$ 381.2432, found 381.2435.







120

((*S*)-2-((1*S*,2*S*)-2-allyl-2-methylcyclopropyl)-1-(2-methyl-1,3-dioxolan-2-yl)ethoxy)(*tert*-butyl)dimethylsilane (120). To a solution of alcohol **119** (2.48 g, 6.91 mmol) and 2-nitrophenyl selenocyanate (7.06 g, 31.07 mmol) in THF (100 mL) was added Bu_3P (7.67 mL, 31.07 mmol) dropwise at 30 °C and the reaction mixture was stirred at this temperature for 90 min. $NaHCO_3$ (17.40 g, 207.15 mmol) was then added in one portion and a solution of H_2O_2 in water (30%, 24.8 mL) was added dropwise. The reaction mixture was further heated to 45 - 50 °C and stirring was continued for 45 min at this temperature. The reaction mixture was then poured into a mixture of water (20 mL), saturated aqueous NH_4Cl (20 mL) and CH_2Cl_2 (120 mL). The whole mixture was stirred vigorously for 15 min, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to yield 1.97 g (84%) of olefin **120** as a slightly yellow oil.

TLC: R_f 0.38 (hexane/EtOAc 20:1, CPS).

$[\alpha]^{20}_D$: = -50.0° (*c* = 0.51, $CHCl_3$).

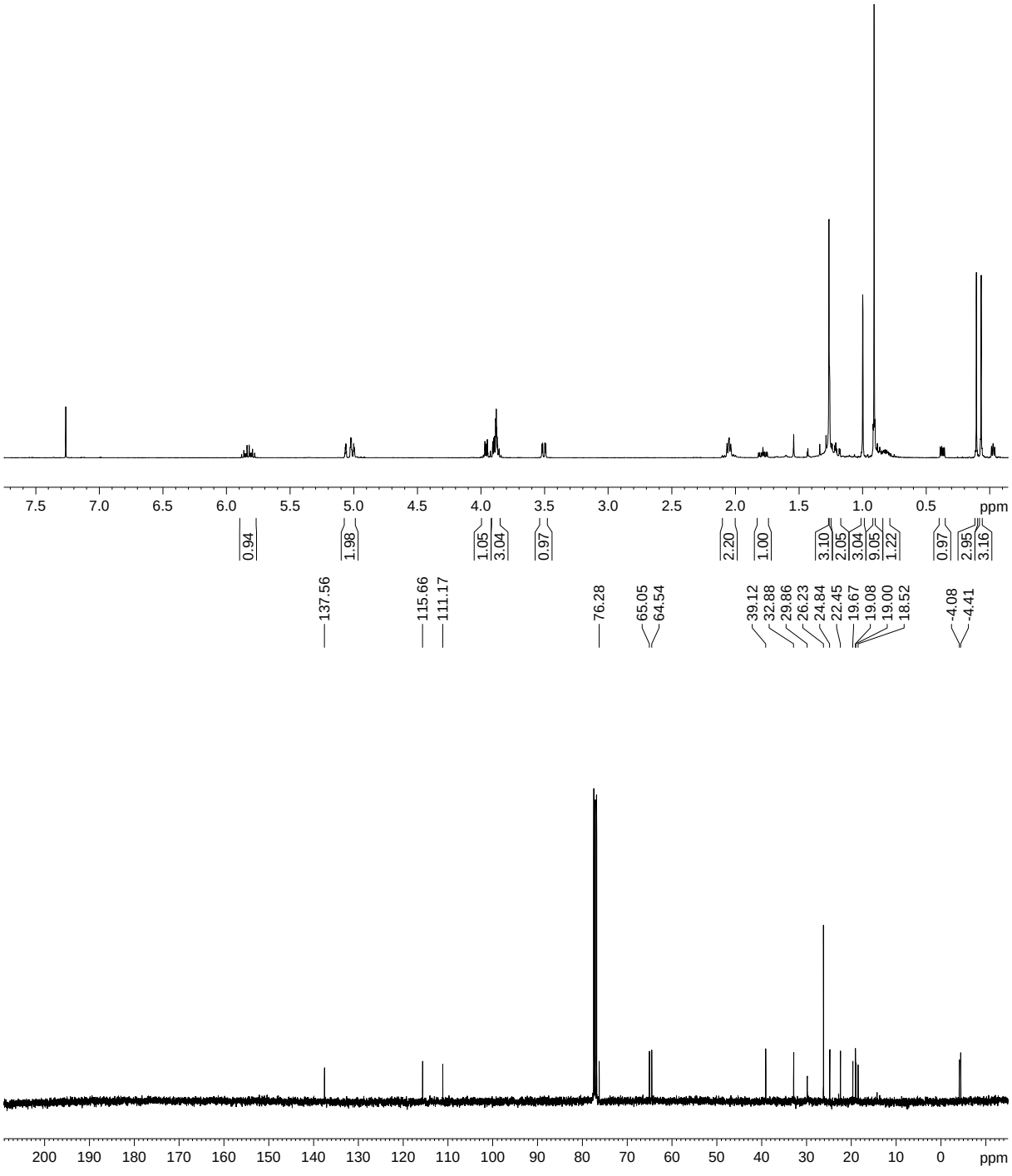
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.83 (ddt, *J* = 17.1, 10.2, 6.9 Hz, 1H), 5.04 (dm, *J* = 17.1 Hz, 1H), 5.01 (dm, *J* = 10.2 Hz, 1H), 4.00-3.92 (m, 1H), 3.92-3.85 (m, 3H), 3.51 (dd, *J* = 9.5,

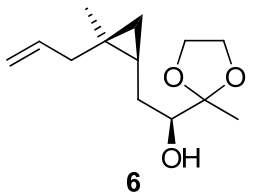
2.5 Hz, 1H), 2.11-2.00 (m, 2H), 1.78 (ddd, *J* = 13.6, 9.5, 3.8 Hz, 1H), 1.26 (s, 3H), 1.21 (ddd, *J* = 13.6, 10.5, 2.5 Hz, 1H), 1.00 (s, 3H), 0.91 (s, 9H), 0.82 (dddd, *J* = 10.5, 8.4, 5.4, 3.8 Hz, 1H), 0.37 (dd, *J* = 8.4, 4.2 Hz, 1H), 0.11 (s, 3H), 0.07 (s, 3H), -0.03 (dd, *J* = 5.4, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 137.6, 115.7, 111.2, 76.3, 65.0, 64.5, 39.1, 32.9, 26.2, 24.8, 22.4, 19.7, 19.1, 19.0, 18.5, -4.1, -4.4.

IR (film): ν 3076, 2955, 2928, 2856, 1641, 1467, 1379, 1251, 1105, 1057, 834, 776.

HRMS (ESI): calculated for $C_{19}H_{36}O_3Si$ $[M+Na]^+$ 363.2326, found 363.2326.





6

(*S*)-2-((1*S*,2*S*)-2-allyl-2-methylcyclopropyl)-1-(2-methyl-1,3-dioxolan-2-yl)ethanol (6). To a solution of silyl ether **120** (1.45 g, 4.26 mmol) in THF (20 mL) was added TBAF•3H_2O (2.28 g, 7.24 mmol) in one portion at 0 °C and the reaction mixture was heated to 50 °C. After 11 h at this temperature the solution was allowed to reach room temperature and the reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (20 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 20 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 5:1) to yield 0.90 g (93%) of alcohol **6** as a colorless oil.

TLC: R_f 0.28 (hexane/EtOAc 4:1, CPS).

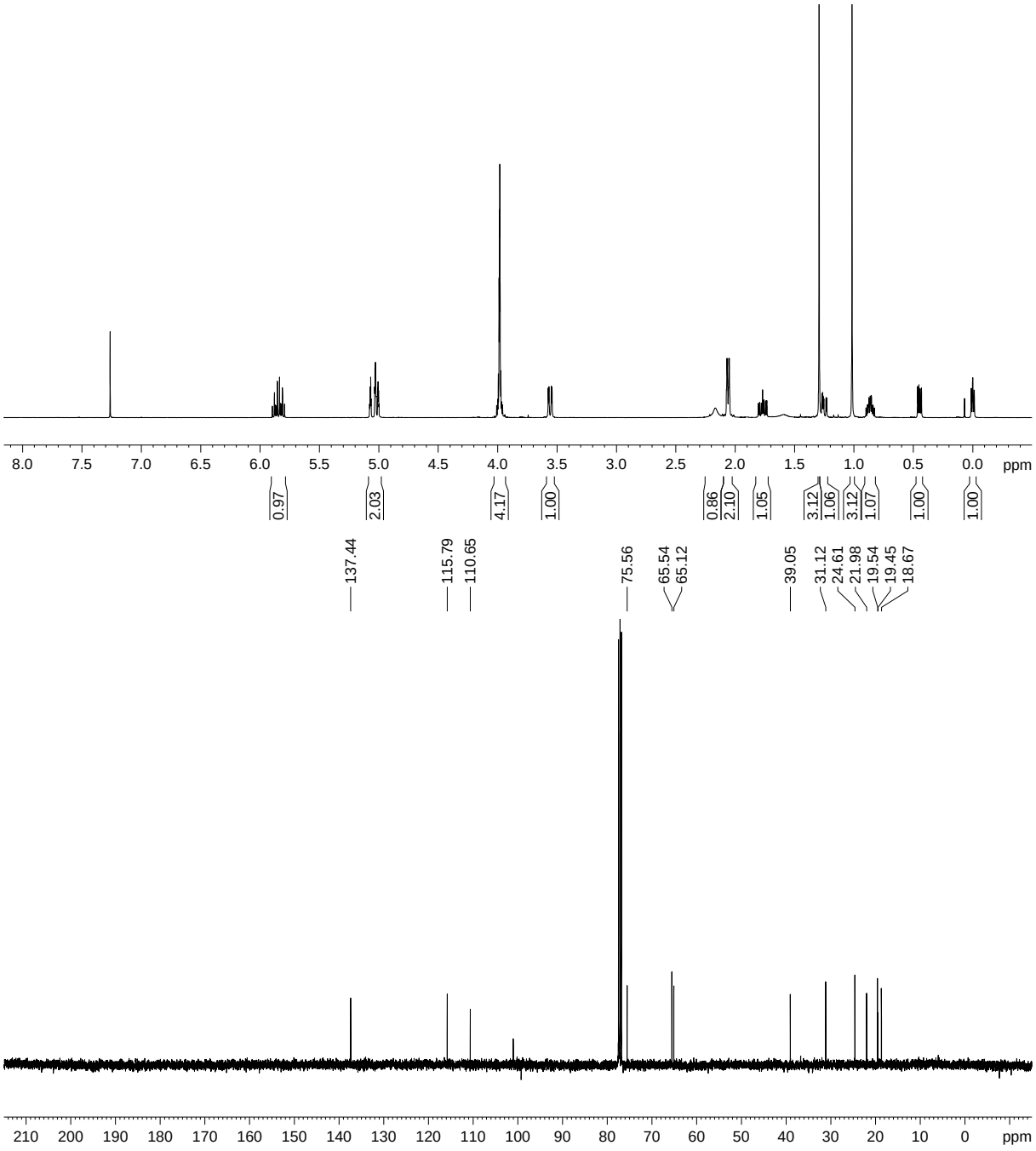
$[\alpha]^{20}_D$: = -82.3° (*c* = 0.45, $CHCl_3$).

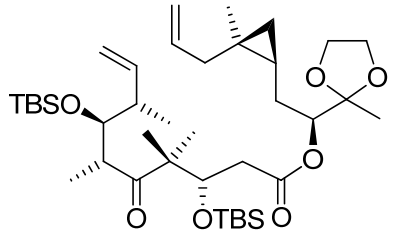
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.85 (ddt, *J* = 17.1, 10.2, 6.9 Hz, 1H), 5.05 (dm, *J* = 17.1 Hz, 1H), 5.02 (dm, *J* = 10.2 Hz, 1H), 4.01-3.95 (m, 4H), 3.56 (dd, *J* = 10.1, 2.5 Hz, 1H), 2.17 (br s, 1H), 2.06 (dq, *J* = 6.9, 1.3 Hz, 2H), 1.77 (ddd, *J* = 14.2, 10.1, 4.5 Hz, 1H), 1.29 (s, 3H), 1.26 (ddd, *J* = 14.2, 9.9, 2.5 Hz, 1H), 1.02 (s, 3H), 0.87 (dddd, *J* = 9.9, 8.4, 5.5, 4.5 Hz, 1H), 0.45 (dd, *J* = 8.4, 4.3 Hz, 1H), 0.00 (dd, *J* = 5.5, 4.3 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 137.4, 115.8, 110.6, 75.6, 65.5, 65.1, 39.0, 31.1, 24.6, 22.0, 19.5, 19.4, 18.7.

IR (film): ν 3482, 3057, 2984, 2955, 2923, 2886, 1638, 1446, 1378, 1296, 1220, 1165, 1057, 995, 949, 911, 877.

HRMS (ESI): calculated for $C_{13}H_{22}O_3Si$ $[M+Na]^+$ 249.1461, found 249.1457.





134

(3*S*,6*R*,7*S*,8*S*)-((*S*)-2-((1*S*,2*S*)-2-allyl-2-methylcyclopropyl)-1-(2-methyl-1,3-dioxolan-2-yl)ethyl) 3,7-bis(*tert*-butyldimethylsilyloxy)-4,4,6,8-tetramethyl-5-oxodec-9-enoate (134).

To a solution of acid **7** (1.73 g, 3.45 mmol) in benzene (34 mL) was added Et_3N (0.96 mL, 6.91 mmol) and 2,4,6-trichlorobenzoyl chloride (0.56 mL, 3.80 mmol) at room temperature and the reaction mixture was stirred for 1 h. Alcohol **6** (0.82 g, 3.63 mmol) in benzene (21 mL) and DMAP (0.55 g, 4.49 mmol) in benzene (13 mL) were then added and the reaction mixture was stirred for 2 h at room temperature. The reaction was then quenched by addition of saturated aqueous $NaHCO_3$ (50 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 2.31 g (94%) of diene **134** as a colorless oil.

Note: Acid **7** was an 8:1 mixture of diastereomers most likely caused by racemization of the aldehyde used in the aldol reaction in the synthesis of the acid **7**. As a result of that, diene **134** is also an 8:1 mixture of diastereomers. The undesired diastereomer was removed in the subsequent RCM step. Racemization can be avoided when the aldehyde is formed directly by reduction of the ester using DIBAL-H rather than by oxidation of the corresponding alcohol under Swern conditions.

TLC: R_f 0.34 (hexane/EtOAc 20:1, CPS).

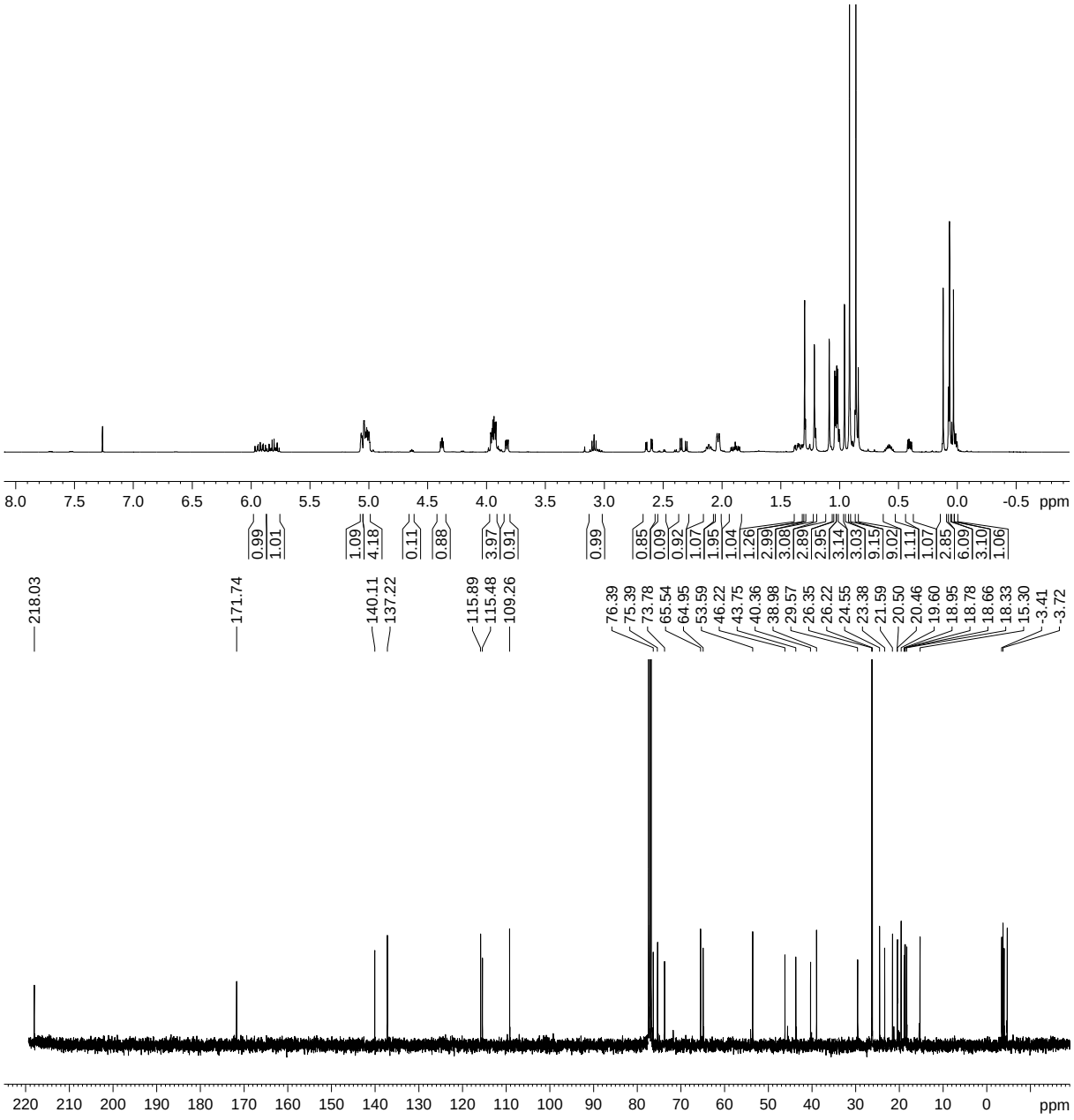
$[\alpha]^{20}_D$: = -32.3° (*c* = 1.08, $CHCl_3$).

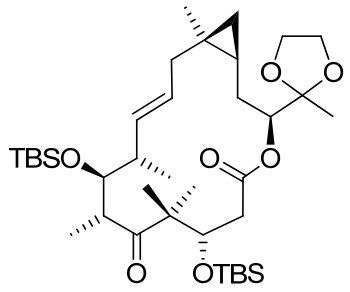
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.92 (ddd, *J* = 17.6, 10.6, 7.6 Hz, 1H), 5.81 (ddt, *J* = 17.0, 10.2, 6.9 Hz, 1H), 5.07-4.99 (m, 5H), 4.38 (dd, *J* = 5.5, 3.8 Hz, 1H), 3.97-3.91 (m, 4H), 3.83 (dd, *J* = 6.9, 2.2 Hz, 1H), 3.07 (dq, *J* = 6.9, 6.9 Hz, 1H), 2.62 (dd, *J* = 17.8, 3.8 Hz, 1H), 2.32 (dd, *J* = 17.8, 5.5 Hz, 1H), 2.11 (qdd, *J* = 7.0, 6.9, 2.2 Hz, 1H), 2.03 (dq, *J* = 6.9, 1.3 Hz, 2H), 1.77 (ddd, *J* = 14.3, 10.2, 4.6 Hz, 1H), 1.35 (ddd, *J* = 14.3, 9.8, 2.8 Hz, 1H), 1.30 (s, 3H), 1.21 (s, 3H), 1.09 (s, 3H), 1.03 (d, *J* = 6.9 Hz, 3H), 1.02 (d, *J* = 7.0 Hz, 3H), 0.96 (s, 3H), 0.92 (s, 9H), 0.86 (s, 9H), 0.58 (dddd, *J* = 9.8, 8.4, 5.2, 4.6 Hz, 1H), 0.41 (dd, *J* = 8.4, 4.4 Hz, 1H), 0.12 (s, 3H), 0.07 (s, 6H), 0.03 (s, 3H), 0.01 (dd, *J* = 5.2, 4.4 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 171.7, 140.1, 137.2, 115.9, 115.5, 109.3, 76.4, 75.4, 73.8, 65.5, 64.9, 53.6, 46.2, 43.7, 40.4, 39.0, 29.6, 26.4, 26.2, 24.5, 23.4, 21.6, 20.5, 20.5, 19.6, 18.9, 18.8, 18.7, 18.3, 15.3, -3.4, -3.7, -4.0, -4.7.

IR (film): ν 2956, 2888, 2858, 1742, 1696, 1471, 1382, 1294, 1254, 1175, 1081, 1046, 988, 913, 874, 836, 776.

HRMS (ESI): calculated for $C_{39}H_{72}O_7Si_2$ $[M+Na]^+$ 731.4709, found 731.4726.





135

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-bis(*tert*-butyldimethylsilyloxy)-8,8,10,12,16-pentamethyl-3-(2-methyl-1,3-dioxolan-2-yl)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-

dione (135). To a solution of diene **134** (256 mg, 0.36 mmol) in CH_2Cl_2 (250 mL) was added 2nd generation Grubbs catalyst (61 mg, 0.072) in CH_2Cl_2 (1 mL) and the reaction mixture was heated to reflux for 14 h. The solution was then cooled to room temperature, filtered through a small plug of silica and the precipitate was rinsed with CH_2Cl_2 (50 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 50:1 → 30:1) to yield 190 mg of olefin **135** (77%) as a single isomer as a colorless foam.

Note: Column chromatography yielded 190 mg of the *E* isomer **135**, 10 mg of a mixture of *E*- and *Z*-isomers and 10 mg of *Z*-isomer **135-*Z***, which was contaminated with impurities derived from the metathesis catalyst. Based on these yields, the RCM reaction produced a *ca*. 12:1 ratio of *E*- and *Z*-isomers.

TLC: R_f 0.40 (hexane/EtOAc 20:1, CPS).

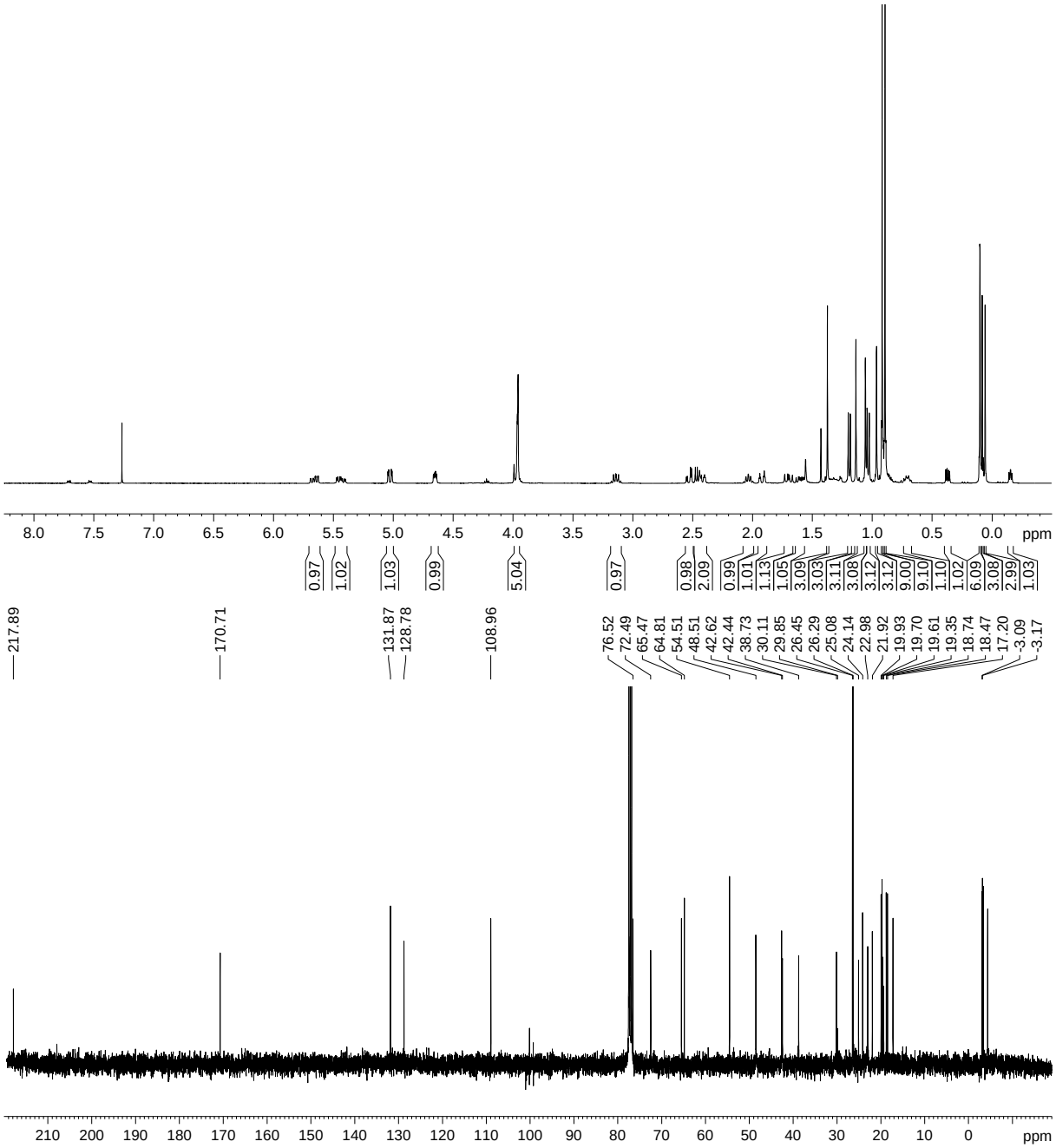
$[\alpha]^{20}_D$: = +14.2° (*c* = 1.01, $CHCl_3$).

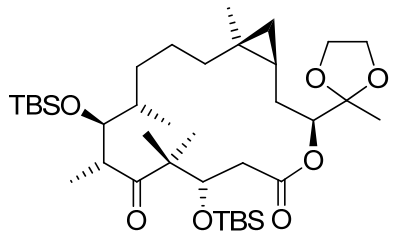
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.66 (ddd, *J* = 15.6, 8.7, 2.0 Hz, 1H), 5.43 (ddd, *J* =15.6, 10.3, 3.4 Hz, 1H), 5.02 (dd, *J* = 11.2, 2.4, 1H), 4.65 (dd, *J* = 6.7, 3.6 Hz, 1H), 3.99-3.94 (m, 5H), 3.14 (dq, *J* = 9.5, 7.2 Hz, 1H), 2.53 (dd, *J* = 13.9, 3.6 Hz, 1H), 2.45 (dd, *J* = 13.9, 6.7 Hz, 1H), 2.42 (ddd, *J* = 15.3, 3.4, 2.0 Hz, 1H), 2.03 (dq, *J* = 8.7, 7.1 Hz, 1H), 1.92 (ddd, *J* = 14.6, 2.4, 2.4 Hz, 1H), 1.70 (dd, *J* = 15.3, 10.3 Hz, 1H), 1.59 (ddd, *J* = 14.6, 11.2, 8.0 Hz, 1H), 1.37 (s, 3H), 1.19 (d, *J* = 7.2 Hz, 3H), 1.14 (s, 3H), 1.06 (s, 3H), 1.03 (d, *J* = 7.1 Hz, 3H), 0.96 (s, 3H), 0.92 (s, 9H), 0.89 (s, 9H), 0.71 (dddd, *J* = 9.1, 8.0, 5.6, 2.4 Hz, 1H), 0.37 (dd, *J* = 9.1, 4.2 Hz, 1H), 0.10 (s, 9H), 0.08 (s, 3H), 0.06 (s, 3H), -0.16 (dd, *J* = 5.6, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 217.9, 170.7, 131.9, 128.8, 109.0, 76.5, 72.5, 65.5, 64.8, 54.5, 48.5, 42.6, 42.4, 38.7, 30.1, 29.4, 26.5, 26.3, 25.1, 24.1, 23.0, 21.9, 19.9, 19.7, 19.6, 19.4, 18.7, 18.5, 17.2, -3.1, -3.2, -3.4, -4.4.

IR (film): ν □2955, 2930, 2889, 2858, 1741, 1694, 1471, 1383, 1297, 1255, 1178, 1088, 1045, 990, 949, 875, 836, 776.

HRMS (ESI): calculated for $C_{37}H_{68}O_7Si_2$ $[M+Na]^+$ 703.4396, found 703.4408.





136

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-bis(*tert*-butyldimethylsilyloxy)-8,8,10,12,16-pentamethyl-3-(2-methyl-1,3-dioxolan-2-yl)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (136). To a solution of olefin **135** (203 mg, 0.30 mmol) in EtOH (8 mL) was added Lindlar

catalyst (380 mg, 0.18 mmol, ~5% Pd on calcium carbonate) in one portion at room temperature and the reaction mixture was stirred under an atmosphere of H_2 (7.5 bar) for 10 h. Additional catalyst (127 mg, 0.06 mmol) was then added in one portion and the solution was stirred under the same conditions for 4 h. The solution was then filtered through a small plug of celite and the precipitate was rinsed with EtOH (5 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 50:1 → 30:1) to afford 162 mg (80%) of saturated macrolactone **136** as a white solid.

Note: With other batches of Lindlar catalyst, the reaction rate was considerably slower and the reaction did not go to completion. In these cases the catalyst was removed by filtration, the filtrate was concentrated under reduced pressure, and the residue was resubmitted to the above reaction conditions to achieve full conversion to the saturated macrolactone **136**. Noteworthy, however, the yield of the reaction remained unchanged for the less active catalyst batches.

TLC: R_f 0.42 (hexane/EtOAc 20:1, CPS).

$[\alpha]^{20}_D$: = +2.9° (*c* = 0.27, $CHCl_3$).

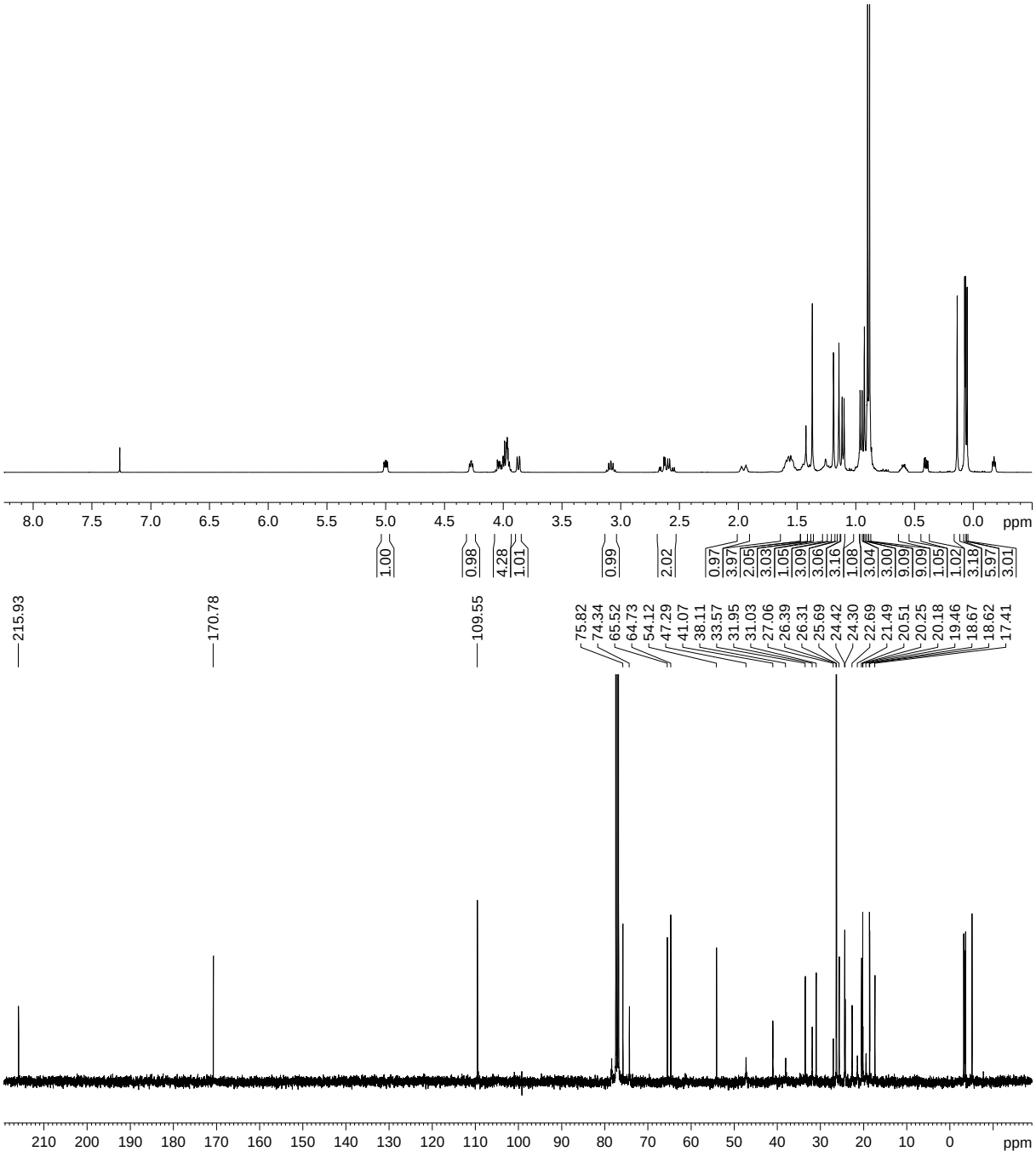
mp: 64-67 °C.

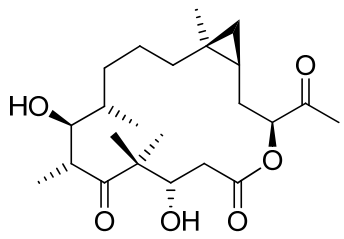
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.00 (dd, *J* = 8.2, 4.0 Hz, 1H), 4.27 (dd, *J* = 6.5, 4.4 Hz, 1H), 4.07-3.94 (m, 4H), 3.87 (dd, *J* = 8.2, 1.0 Hz, 1H), 3.08 (dq, *J* = 8.2, 7.0 Hz, 1H), 2.65 (dd, *J* = 15.9, 4.4 Hz, 1H), 2.57 (dd, *J* = 15.9, 6.5 Hz, 1H), 1.95 (ddd, *J* = 15.1, 4.0, 3.4 Hz, 1H), 1.61-1.47 (m, 4H), 1.46-1.37 (m, 2H), 1.37 (s, 3H), 1.32-1.21 (m, 1H), 1.19 (s, 3H), 1.14 (s, 3H), 1.11 (d, *J* = 6.9 Hz, 3H), 1.01-0.97 (m, 1H), 0.96 (d, *J* = 7.0 Hz, 3H), 0.93 (s, 3H), 0.90 (s, 9H), 0.88 (s, 9H), 0.59 (dddd, *J* = 12.1, 8.7, 5.4, 3.4, 1H), 0.40 (dd, *J* = 8.7, 4.0 Hz, 1H), 0.14 (s, 3H), 0.07 (s, 3H), 0.07 (s, 3H), 0.05 (s, 3H), -0.18 (dd, *J* = 5.4, 4.0 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.9, 170.7, 109.5, 75.8, 74.3, 65.5, 64.7, 54.1, 47.3, 41.1, 38.1, 33.6, 32.0, 31.0, 27.1, 26.4, 26.3, 25.7, 24.4, 24.3, 22.7, 21.5, 20.5, 20.3, 20.2, 19.5, 18.7, 18.6, 17.4, -3.2, -3.5, -3.6, -5.1.

IR (film): ν 2952, 2930, 2886, 2857, 1746, 1696, 1472, 1463, 1382, 1362, 1253, 1178, 1158, 1106, 1085, 1045, 984, 937, 874, 834, 774, 667.

HRMS (ESI): calculated for $C_{37}H_{70}O_7Si_2$ $[M+Na]^+$ 705.4552, found 705.4554.





138

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-acetyl-7,11-dihydroxy-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (138). To a solution of acetal **136** (0.46 g, 0.67 mmol) in CH_2Cl_2 (36 mL) was added $FeCl_3 \cdot 6H_2O$ (0.91 g, 3.37 mmol) in one portion at room

temperature and the reaction mixture was stirred for 4 h. The reaction was then quenched by addition of water (25 mL), the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 0.26 g (94%) of diol **138** as a colorless oil.

TLC: R_f 0.16 (hexane/EtOAc 3:1, CPS).

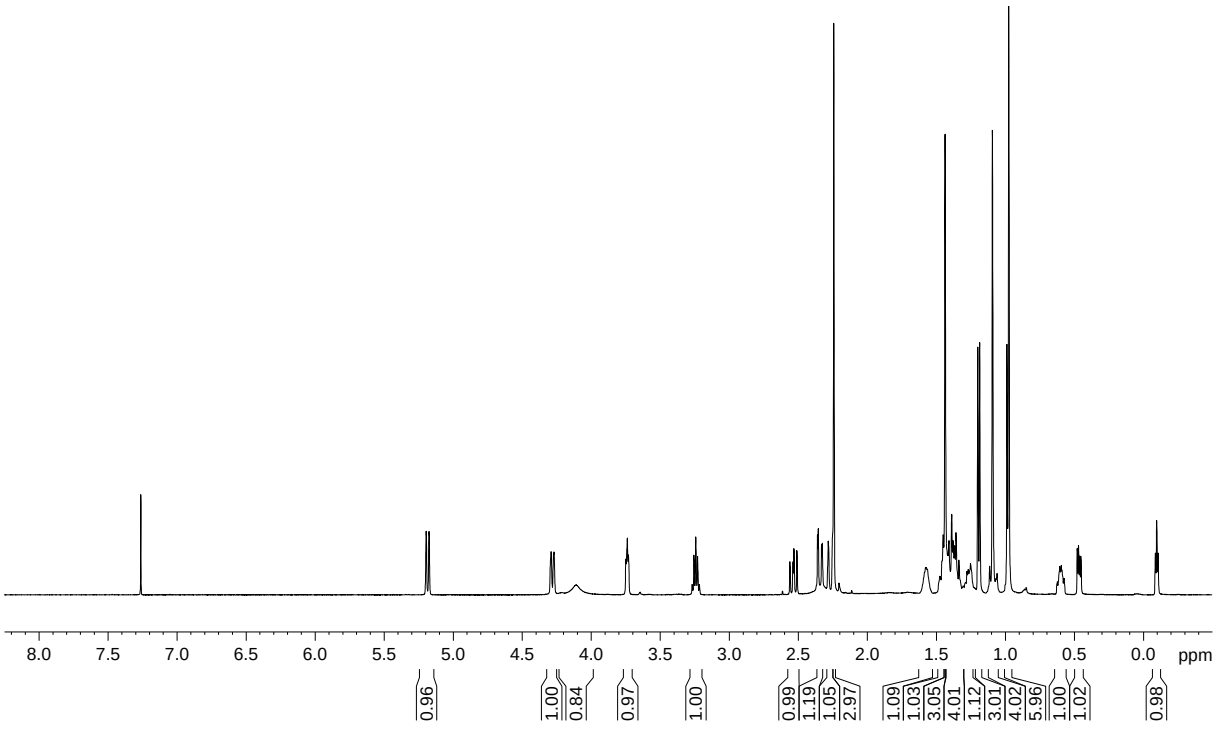
$[\alpha]^{20}_D$: = +22.6° (*c* = 0.67, $CHCl_3$).

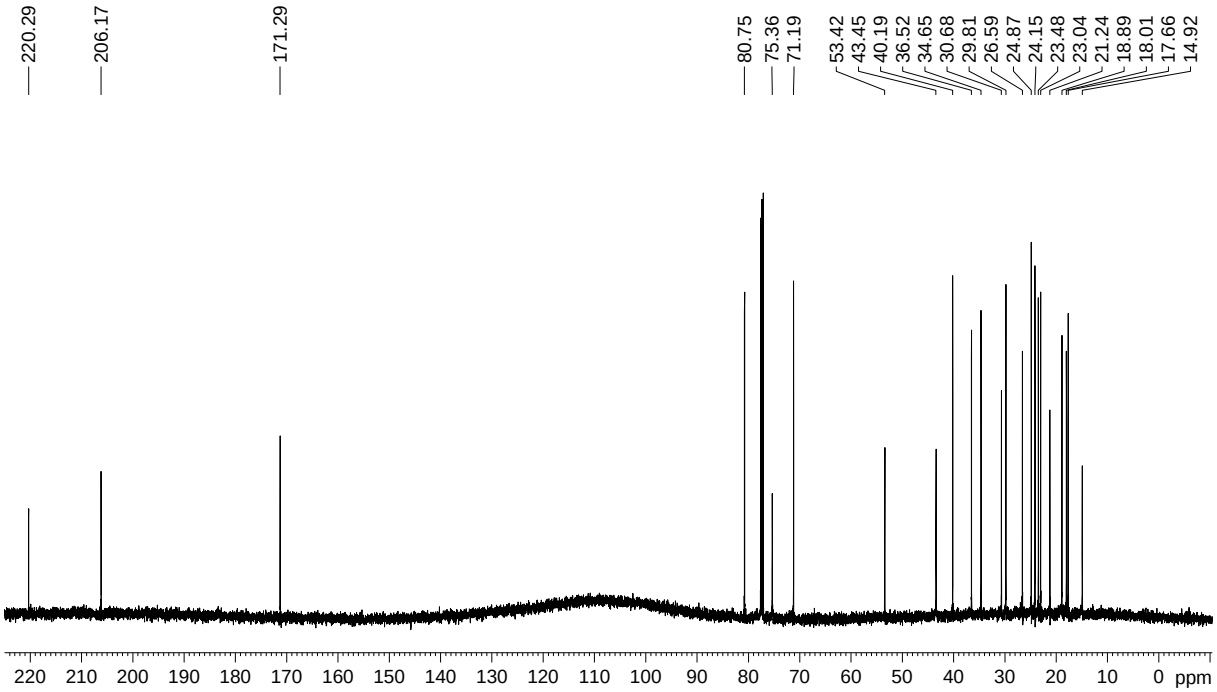
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.18 (dd, *J* = 10.5, 1.7 Hz, 1H), 4.28 (dd, *J* = 10.7, 2.6 Hz, 1H), 4.11 (br s, 1H), 3.74 (dd, *J* = 6.5, 3.4, 1H), 3.24 (qd, *J* = 6.7, 6.5 Hz, 1H), 2.54 (dd, *J* = 14.6, 10.7 Hz, 1H), 2.34 (dd, *J* = 14.6, 2.6 Hz, 1H), 2.27 (ddd, *J* = 15.6, 2.0, 2.0 Hz, 1H), 2.24 (s, 3H), 1.62-1.53 (m, 1H), 1.48-1.45 (m, 1H), 1.44 (s, 3H), 1.44-1.30 (m, 4H), 1.29-1.24 (m, 1H), 1.19 (d, *J* = 6.7 Hz, 3H), 1.12-1.05 (m, 1H), 1.09 (s, 3H), 0.98 (d, *J* = 6.7 Hz, 3H), 0.97 (s, 3H), 0.60 (dddd, *J* = 10.7, 8.7, 5.6, 1.6, 1H), 0.47 (dd, *J* = 8.7, 4.3 Hz, 1H), -0.10 (dd, *J* = 5.6, 4.3 Hz, 1H).

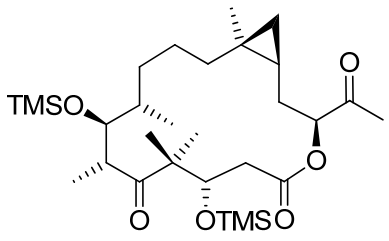
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 220.1, 206.0, 171.1, 80.6, 75.1, 71.0, 53.2, 43.2, 40.0, 36.3, 34.5, 30.5, 29.6, 26.4, 24.7, 23.9, 23.3, 22.8, 21.0, 18.7, 17.8, 17.5, 14.7.

IR (film): ν 3464, 2946, 1742, 1718, 1688, 1457, 1366, 1250, 1180, 1146, 1072, 1009, 981, 958, 736, 671.

HRMS (ESI): calculated for $C_{23}H_{38}O_6$ $[M+Na]^+$ 433.2561, found 433.2580.







98

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-acetyl-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (98). To a solution of diol **138** (96 mg, 0.23 mmol) in CH_2Cl_2 (6 mL) was added 2,6-lutidine (163 µL, 1.40 mmol) at 0 °C. The reaction mixture was cooled to -78 °C and TMSOTf (127 µL, 0.70 mmol) was added dropwise. The reaction mixture was stirred for 1.5 h at this temperature and was then allowed to reach room temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (10 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to yield 129 mg (99%) of protected ketone **98** as a colorless oil.

Note: Upon extensive scratching and storage at -20 °C silyl ether **98** turned into a white solid.

TLC: R_f 0.21 (hexane/EtOAc 12:1, CPS).

$[\alpha]^{20}_D$: = +4.4° (*c* = 0.55, $CHCl_3$).

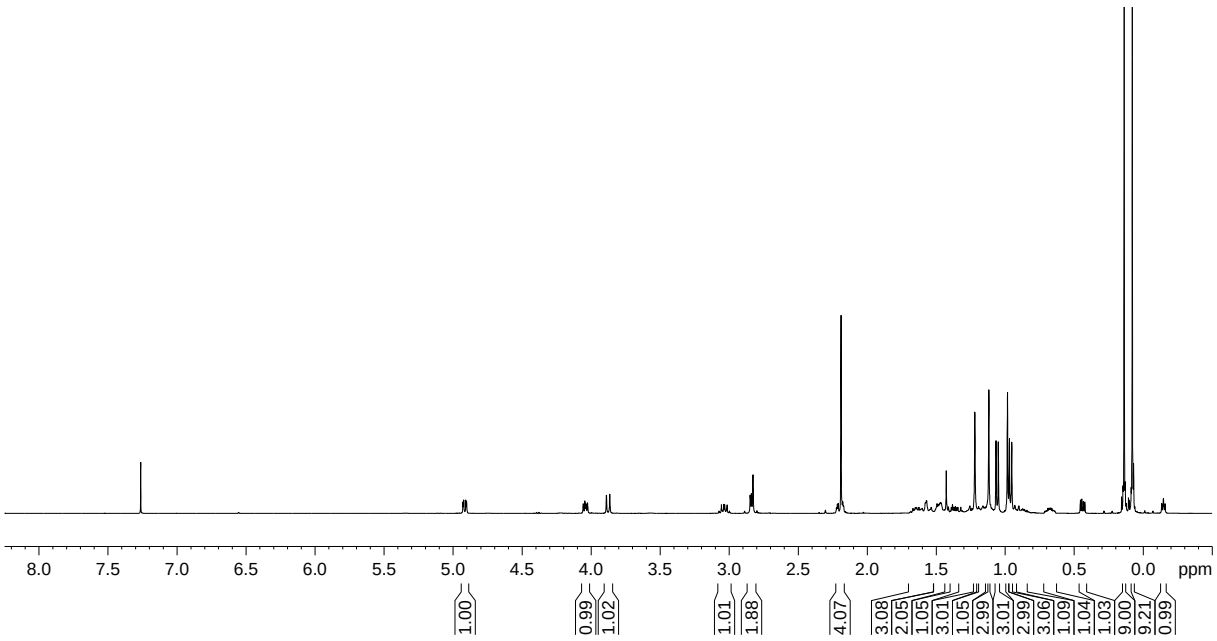
mp: 118-121 °C

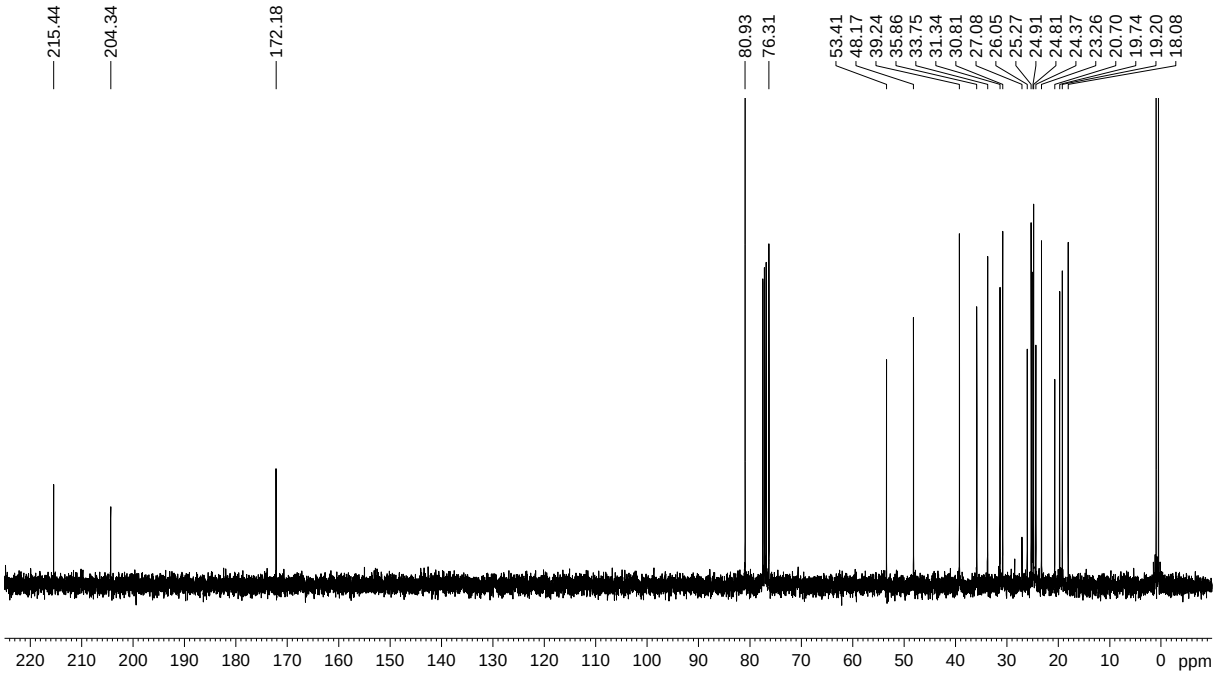
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.91 (dd, *J* = 8.3, 2.4 Hz, 1H), 4.04 (dd, *J* = 8.0, 4.8 Hz, 1H), 3.88 (d, *J* = 9.6 Hz, 1H), 3.04 (dq, *J* = 9.6, 6.7, 1H), 2.84 (d, *J* = 8.0 Hz, 1H), 2.83 (d, *J* = 4.8 Hz, 1H), 2.20 (ddd, *J* = 15.7, 3.1, 2.4, 1H), 2.19 (s, 3H), 1.64 (dq, *J* = 10.5, 6.8 Hz, 1H), 1.57 (td, *J* = 12.7, 3.5 Hz, 2H), 1.52-1.45 (m, 2H), 1.37 (ddd, *J* = 15.7, 11.7, 8.3, 1H), 1.22 (s, 3H), 1.19-1.14 (m, 1H), 1.12 (s, 3H), 1.06 (d, *J* = 6.7, 3H), 0.98 (s, 3H), 0.96 (d, *J* = 6.8 Hz, 3H), 0.92-0.85 (m, 1H), 0.68 (dddd, *J* = 11.7, 8.7, 5.4, 3.1, 1H), 0.44 (dd, *J* = 8.7, 4.2 Hz), 0.14 (s, 9H), 0.08 (s, 9H), -0.15 (dd, *J* = 5.4, 4.2 Hz, 1H).

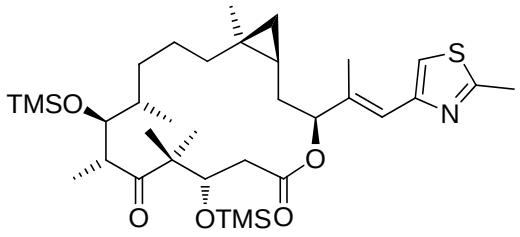
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.4, 204.3, 172.2, 80.9, 76.3, 53.4, 48.2, 39.2, 35.9, 33.8, 31.3, 30.8, 27.1, 26.0, 25.3, 24.9, 24.8, 24.4, 23.3, 20.7, 19.7, 19.2, 18.1, 1.0, 0.5.

IR (film): ν □2954, 1735, 1697, 1458, 1384, 1310, 1251, 1159, 1114, 1081, 1055, 1021, 888, 842, 768.

HRMS (ESI): calculated for $C_{29}H_{55}O_6Si_2$ $[M+H]^+$ 555.3532, found 555.3542.







16a

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-8,8,10,12,16-pentamethyl-3-((*E*)-1-(2-methylthiazol-4-yl)prop-1-en-2-yl)-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (139a). To a solution of phosphonium salt **78a** (42 mg, 0.118 mmol) in THF (1 mL) was added KHMDS (24 mg, 0.118 mmol) in one portion at 0 °C. The reaction mixture was stirred for 0.5 h at this temperature and was then cooled to -78 °C. Methyl ketone **98** (13.0 mg, 0.024 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to afford 10.3 mg (67%) of an inseparable 6:1 mixture of **139a** and its C16-C17 *Z* isomer as a semi solid.

TLC: R_f 0.36 (hexane/EtOAc 9:1, UV, CPS).

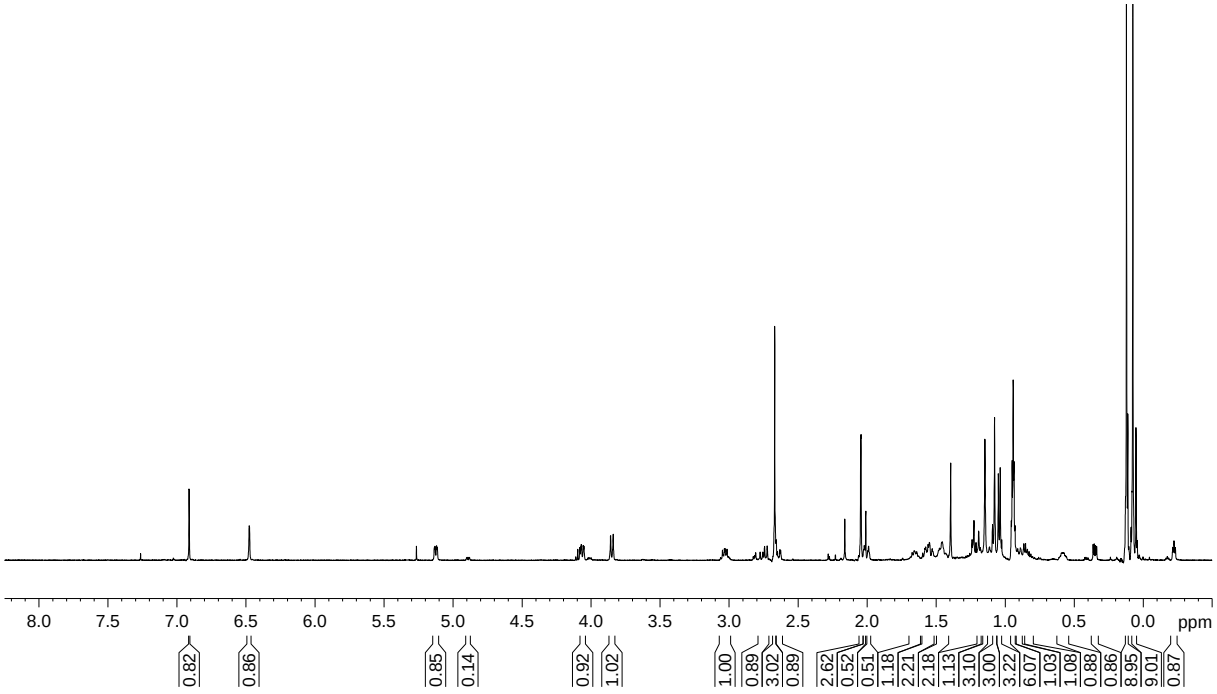
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.91 (s, 1H), 6.48 (s, 1H), 5.13 (dd, *J* = 8.0, 2.4 Hz, 1H), 4.06 (dd, *J* = 9.7, 2.6 Hz, 1H), 3.85 (d, *J* = 9.4 Hz, 1H), 3.03 (dq, *J* = 9.4, 6.8 Hz, 1H), 2.75

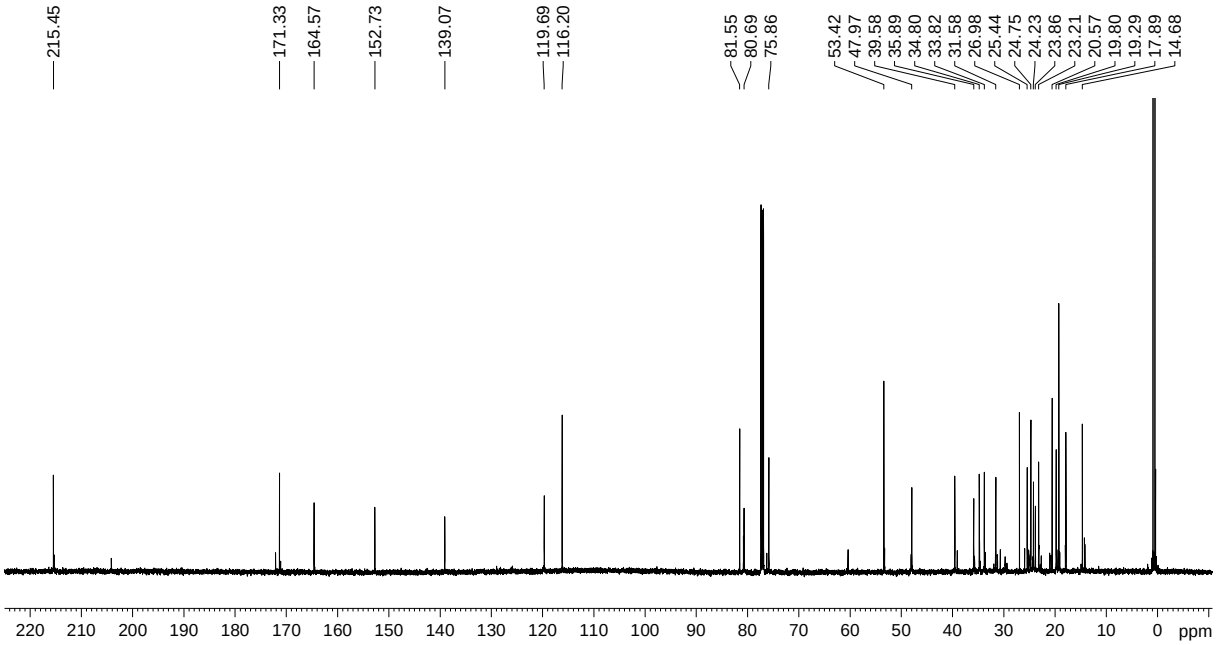
(dd, *J* = 16.1, 9.7 Hz, 1H), 2.67 (s, 3H), 2.64 (dd, *J* = 16.1, 2.6 Hz, 1H), 2.04 (d, *J* = 1.0 Hz, 3H), 2.00 (ddd, *J* = 15.6, 3.1, 2.4 Hz, 1H), 1.65 (dq, *J* = 9.1, 6.9 Hz, 1H), 1.58 (dd, *J* = 11.1, 8.4 Hz, 1H), 1.57-1.52 (m, 1H), 1.49-1.41 (m, 2H), 1.20-1.18 (m, 1H), 1.15 (s, 3H), 1.08 (s, 3H), 1.04 (d, *J* = 6.8 Hz, 3H), 0.94 (d, *J* = 6.8 Hz, 3H), 0.94 (s, 3H), 0.90 (dt, *J* = 9.7, 2.4 Hz, 1H), 0.87-0.79 (m, 1H), 0.58 (dddd, *J* = 10.8, 8.7, 5.4, 3.1 Hz, 1H), 0.35 (dd, *J* = 8.7, 4.1 Hz, 1H), 0.12 (s, 9H), 0.08 (s, 9H), -0.22 (dd, *J* = 5.4, 4.1 Hz, 1H).

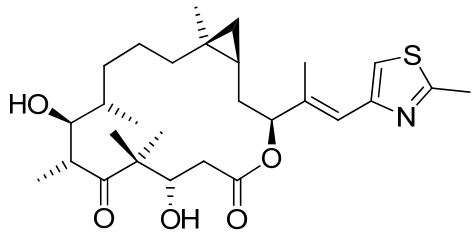
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.4, 171.3, 164.6, 152.7, 139.1, 119.7, 116.2, 81.6, 80.7, 75.9, 53.4, 48.0, 39.6, 35.9, 34.8, 33.8, 31.6, 27.0, 25.4, 24.8, 24.2, 23.9, 23.2, 20.6, 19.8, 19.3, 17.9, 14.7, 0.9, 0.5.

IR (film): ν 2955, 2876, 1741, 1698, 1456, 1381, 1250, 1159, 1115, 1021, 889, 841, 756.

HRMS (ESI): calculated for $C_{34}H_{60}NO_5SSi_2$ $[M+H]^+$ 650.3725, found 650.3724.







1a

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-((*E*)-1-(2-methylthiazol-4-yl)prop-1-en-2-yl)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (1a). To a 6:1 mixture of protected cyclopropyl-Epo B **139a** and its C16-C17 *Z* isomer (8.4 mg, 0.013 mmol) in MeOH (0.5 mL) was added citric acid (7.0 mg, 0.033 mmol) at room temperature. The reaction mixture was stirred for 12 h, a second portion of citric acid (3.0 mg, 0.014 mmol) was then added and stirring was continued for further 10 h. The reaction mixture was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 5:2) to yield 5.2 mg of **139a** (80%) as a single isomer as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 2:1, UV, CPS).

$[\alpha]^{20}_D$: = -67.8° (*c* = 0.33, $CHCl_3$).

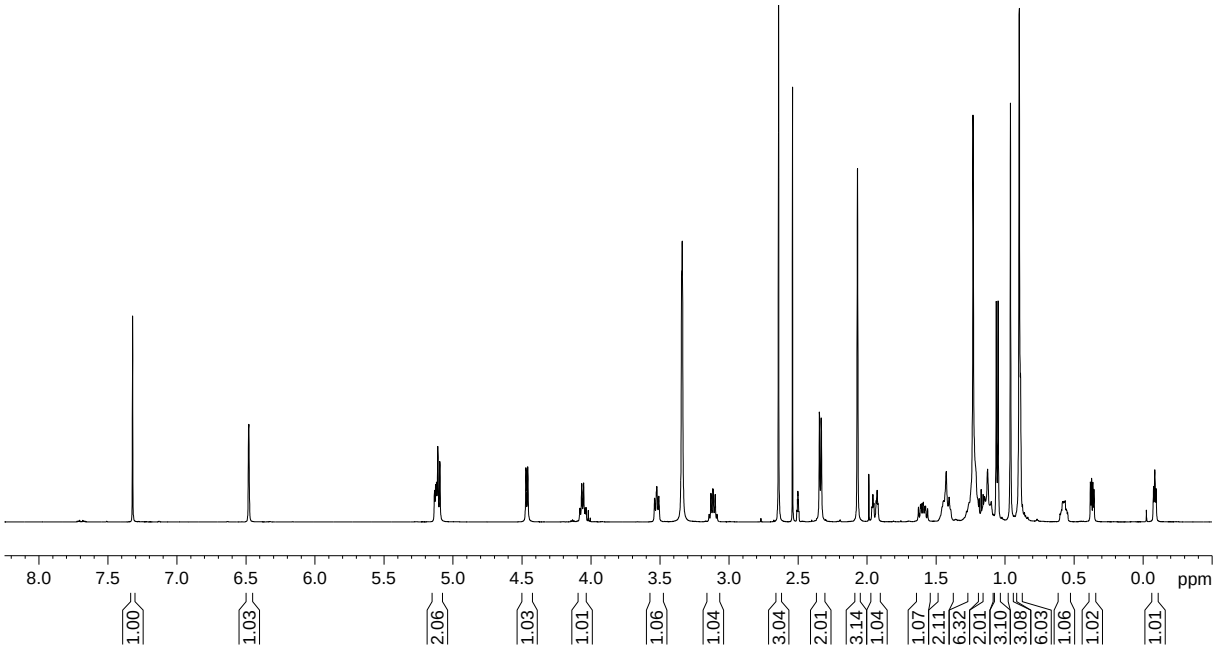
^{1}H-NMR (500 MHz, DMSO-d6): δ = 7.32 (s, 1H), 6.48 (s, 1H), 5.12 (dd, *J* = 7.2, 3.6 Hz, 1H), 5.10 (d, *J* = 6.8 Hz, 1H), 4.46 (d, *J* = 6.6 Hz, 1H), 4.06 (ddd, *J* = 6.8, 6.7, 6.6 Hz, 1H), 3.52 (dd, *J* = 8.4, 6.6 Hz, 1H), 3.11 (dq, *J* = 8.4, 6.7 Hz, 1H), 2.64 (s, 3H), 2.34 (d, *J* = 6.6 Hz, 2H), 2.07 (d, *J* = 0.7 Hz, 3H), 1.94 (ddd, *J* = 15.4, 3.6, 3.2 Hz, 1H), 1.59 (ddd, *J* = 15.4, 10.4, 7.2 Hz, 1H), 1.48-1.38 (m, 2H), 1.25-1.20 (m, 3H), 1.23 (s, 3H), 1.16-1.09 (m, 2H), 1.06 (d, *J* = 6.7 Hz, 3H), 0.96 (s, 3H), 0.89 (s, 3H), 0.89 (d, *J* = 6.7 Hz, 3H), 0.57 (dddd, *J* = 10.4, 8.7, 5.4, 3.2 Hz, 1H), 0.37 (dd, *J* = 8.7, 4.0 Hz, 1H), -0.09 (dd, *J* = 5.4, 4.0 Hz, 1H).

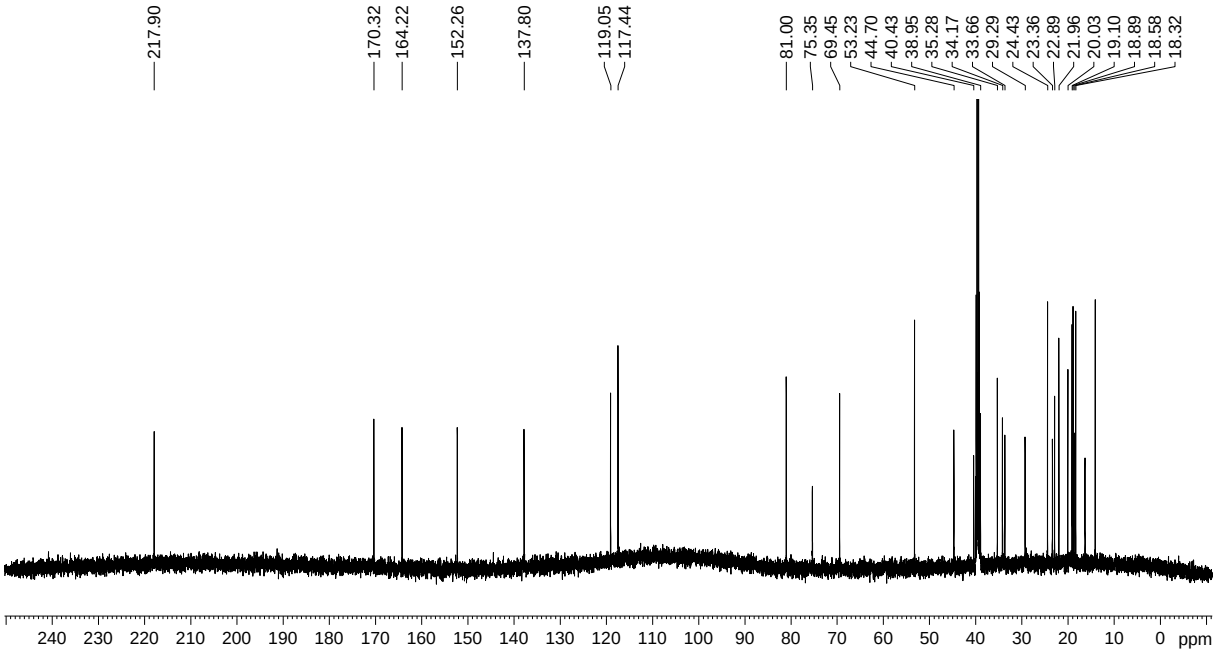
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.9, 170.3, 164.2, 152.3, 137.8, 119.0, 117.4, 81.0, 75.3, 69.4, 53.2, 44.7, 40.4, 38.9, 35.3, 34.2, 33.7, 24.4, 23.4, 22.9, 22.0, 20.0, 19.1, 18.9, 18.6, 18.3, 16.3, 14.1.

IR (film): ν 3423, 2941, 1730, 1687, 1509, 1458, 1377, 1254, 1183, 1148, 1009, 982, 876, 759, 668.

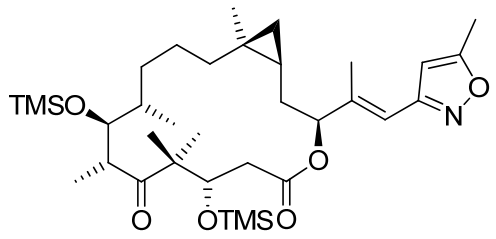
HRMS (ESI): calculated for $C_{28}H_{44}NO_5S$ $[M+H]^+$ 506.2935, found 506.2948.

Analytical HPLC: Method: eluent B 50-60% (linear gradient from 0-12 min), retention time 10.98 min.





4.2.2 Synthesis of Side Chain-modified Analogs of CP-Epo B



139b

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-8,8,10,12,16-pentamethyl-3-((*E*)-1-(5-methylisoxazol-3-yl)prop-1-en-2-yl)-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (139b). To a solution of phosphonium salt **78b** (36 mg, 0.108 mmol) in THF (1 mL) was added KHMDS (22 mg, 0.118 mmol) in one portion at 0 °C. The reaction mixture was stirred for 0.5 h at this temperature and was then cooled to -78 °C. Methyl ketone **98** (12.0 mg, 0.022 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography

(hexane/EtOAc 10:1) to afford 10.2 mg (74%) of an inseparable 17:1 mixture of **139b** and its C16-C17 *Z* isomer as a colorless oil.

TLC: R_f: 0.29 (hexane/EtOAc 9:1, UV, CPS).

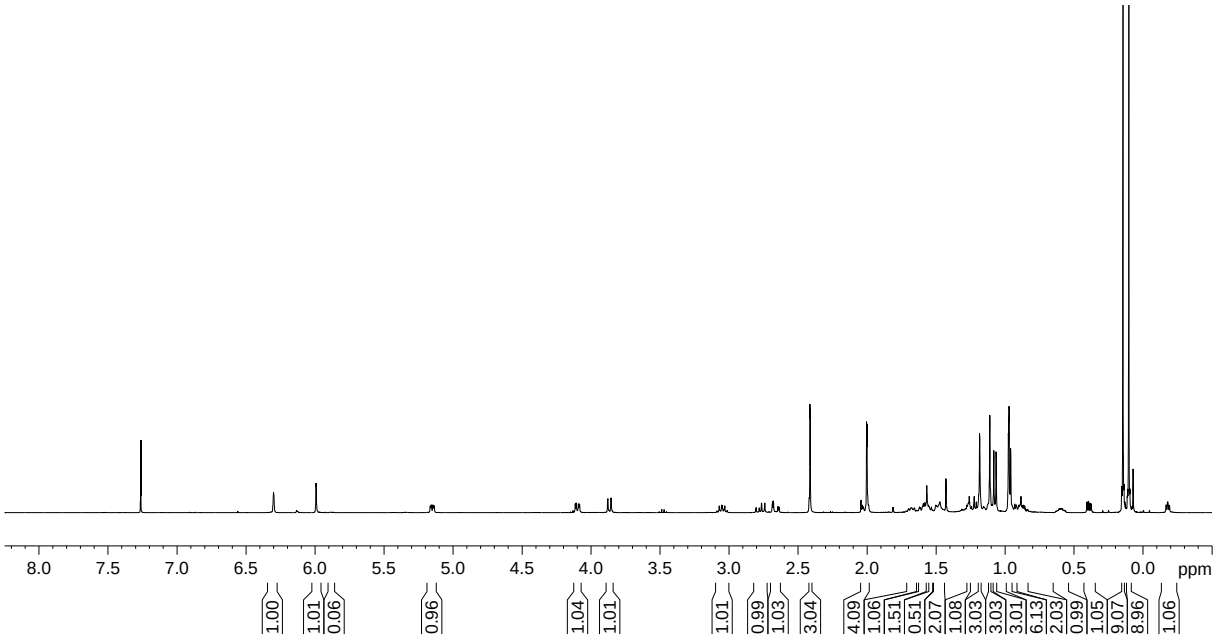
$[\alpha]^{20}_D$: = -5.8° (*c* = 0.50, $CHCl_3$).

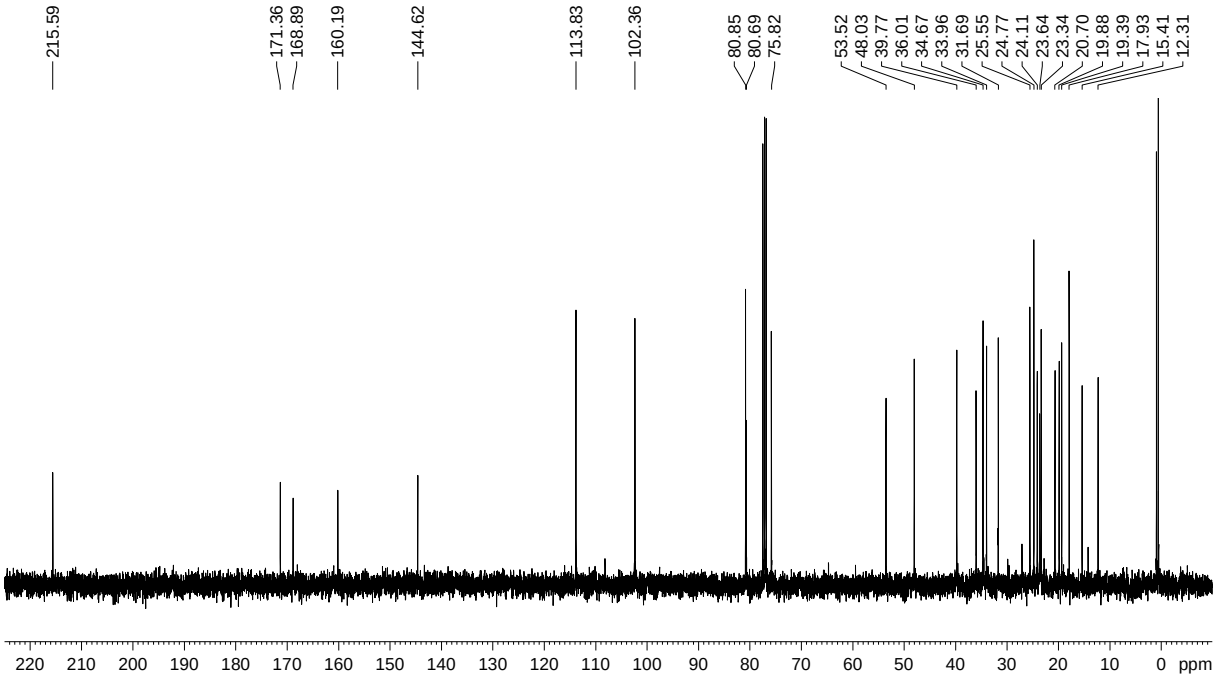
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.30 (dq, *J* = 0.8, 0.8 Hz, 1H), 5.99 (d, *J* = 0.8 Hz, 1H), 5.15 (dd, *J* = 8.0, 2.9 Hz, 1H), 4.10 (dd, *J* = 9.2, 3.0 Hz, 1H), 3.87 (d, *J* = 9.3 Hz, 1H), 3.05 (dq, *J* = 9.3, 6.7 Hz, 1H), 2.77 (dd, *J* = 16.0, 9.2 Hz, 1H), 2.66 (dd, *J* = 16.0, 3.0 Hz, 1H), 2.41 (d, *J* = 0.8 Hz, 3H), 2.04-2.00 (m, 1H), 2.00 (d, *J* = 1.3 Hz, 3H), 1.68 (dq, *J* = 9.3, 6.7 Hz, 1H), 1.63-1.52 (m, 2H), 1.51-1.46 (m, 2H), 1.28-1.25 (m, 1H), 1.18 (s, 3H), 1.11 (s, 3H), 1.07 (d, *J* = 6.7 Hz, 3H), 0.97 (s, 3H), 0.97 (d, *J* = 6.7 Hz, 3H), 0.93-0.85 (m, 2H), 0.60 (dddd, *J* = 11.6, 8.7, 5.3, 3.1 Hz, 1H), 0.39 (dd, *J* = 8.7, 4.1 Hz, 1H), 0.15 (s, 9H), 0.10 (s, 9H), -0.18 (dd, *J* = 5.3, 4.1 Hz, 1H).

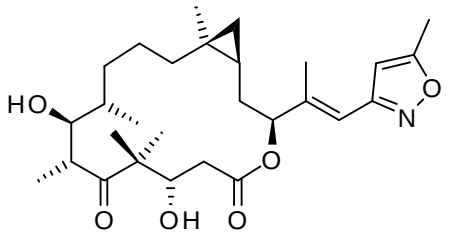
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.6, 171.4, 168.9, 160.2, 144.6, 113.8, 102.4, 80.8, 80.7, 75.8, 53.5, 48.0, 39.8, 36.0, 34.7, 34.0, 31.7, 25.5, 24.8, 24.1, 23.6, 23.3, 20.7, 19.9, 19.4, 17.9, 15.4, 12.3, 0.9, 0.6.

IR (film): ν □2954, 1742, 1697, 1604, 1541, 1519, 1457, 1380, 1252, 1158, 1116, 1053, 1021, 986, 889, 842, 772.

HRMS (ESI): calculated for $C_{34}H_{60}NO_6Si_2$ $[M+H]^+$ 634.3954, found 634.3941.







1b

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-((*E*)-1-(5-methylisoxazol-3-yl)prop-1-en-2-yl)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (1b). To a 17:1 mixture of protected cyclopropyl-Epo B **139b** and its C16-C17 *Z* isomer (10.0 mg, 0.016 mmol) in methanol (0.5 mL) was added citric acid (9.4 mg, 0.045 mmol) at room temperature. The reaction mixture was stirred for 13 h, was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 5:2) to yield 6.0 mg (78%) of **1b** as a single isomer as a colorless oil.

TLC: R_f 0.21 (hexane/EtOAc 2:1, UV, CPS).

$[\alpha]^{20}_D$: = -45.6° (*c* = 0.33, $CHCl_3$).

^{1}H-NMR (500 MHz, DMSO-d_6): δ = 6.38 (s, 1H), 6.28 (s, 1H), 5.14 (d, *J* = 7.1 Hz, 1H), 5.13 (dd, *J* = 7.5, 3.4 Hz, 1H), 4.47 (d, *J* = 6.6 Hz, 1H), 4.07 (ddd, *J* = 7.3, 7.1, 6.2 Hz, 1H), 3.53

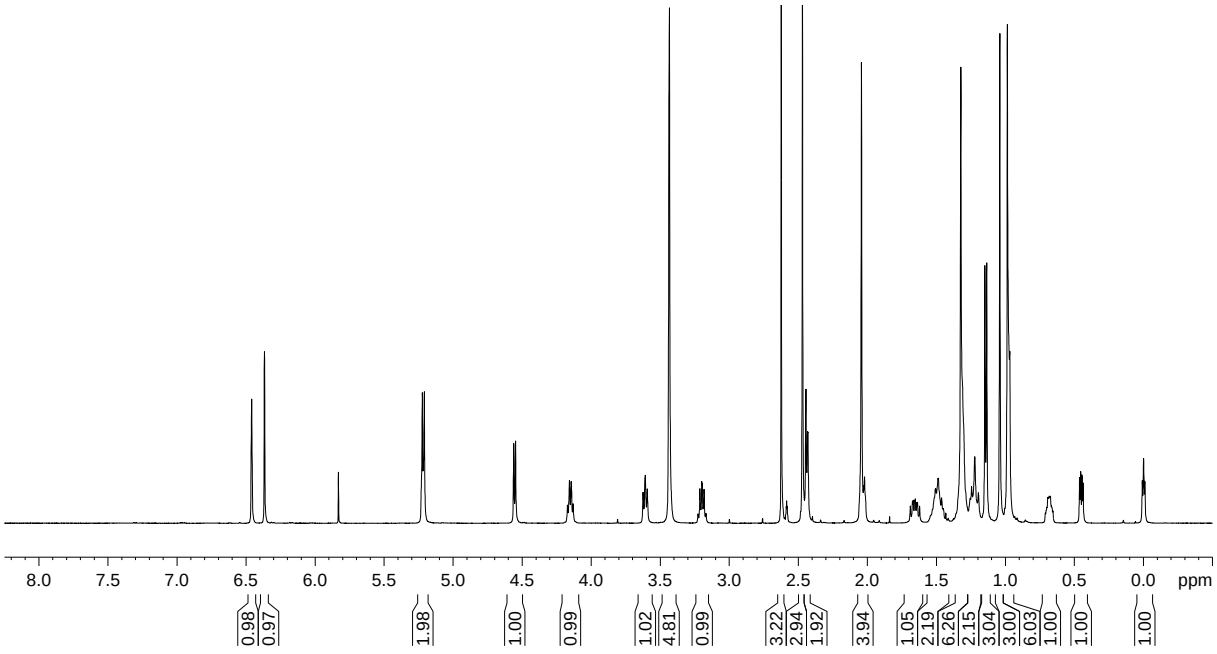
(dd, *J* = 8.6, 6.6 Hz, 1H), 3.11 (dq, *J* = 8.6, 6.7 Hz, 1H), 2.39 (s, 3H), 2.36 (d, *J* = 7.3 Hz, 1H), 2.35 (d, *J* = 6.2 Hz, 1H), 1.96 (s, 3H), 1.95 (ddd, *J* = 15.5, 3.4, 3.4 Hz, 1H), 1.57 (ddd, *J* = 15.5, 10.5, 7.5 Hz, 1H), 1.47-1.34 (m, 2H), 1.23 (s, 3H), 1.24-1.20 (m, 3H), 1.17-1.10 (m, 2H), 1.06 (d, *J* = 6.7, 3H), 0.96 (s, 3H), 0.90 (s, 3H), 0.89 (d, *J* = 6.7 Hz, 3H), 0.60 (dddd, *J* =10.5, 8.7, 5.4, 3.4 Hz, 1H), 0.37 (dd, *J* = 8.7, 4.0 Hz, 1H), -0.08 (dd, *J* = 5.4, 4.0 Hz, 1H).

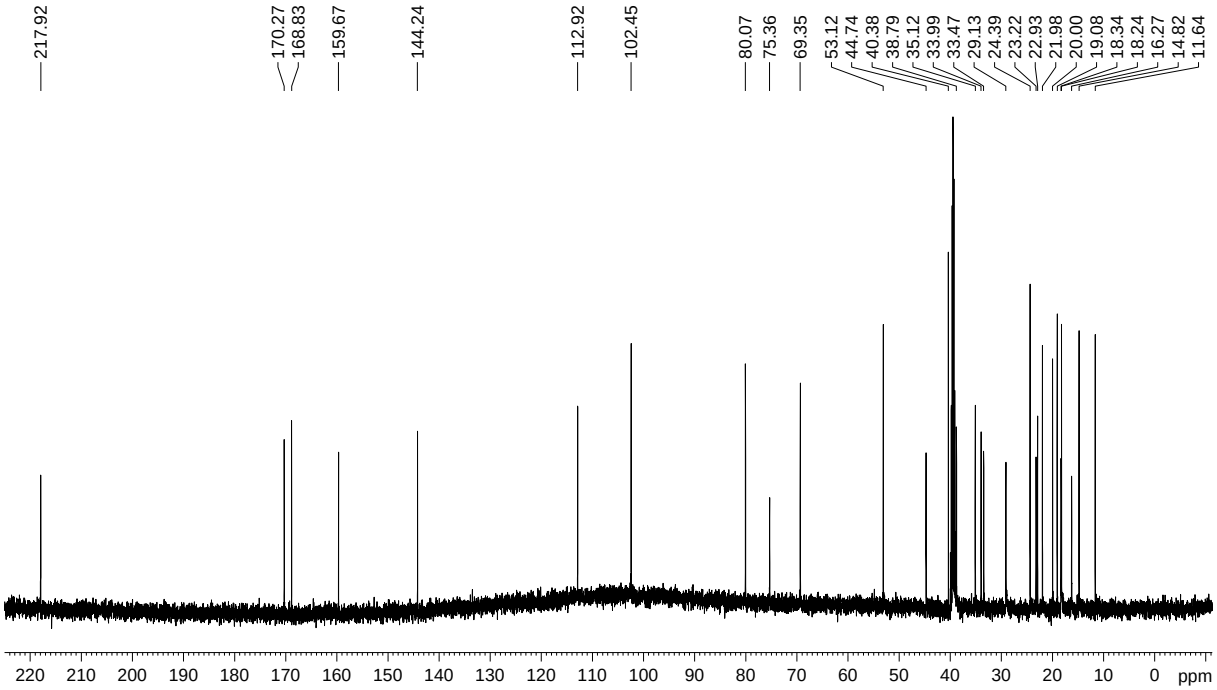
^{13}C-NMR (125 MHz, DMSO-d6): δ = 218.0, 170.3, 168.9, 159.7, 144.3, 112.9, 102.5, 80.1, 75.4, 69.4, 53.2, 44.8, 40.4, 38.8, 35.2, 34.0, 33.5, 29.2, 24.4, 23.3, 23.0, 22.0, 20.1, 19.1, 18.4, 18.3, 16.3, 14.9.

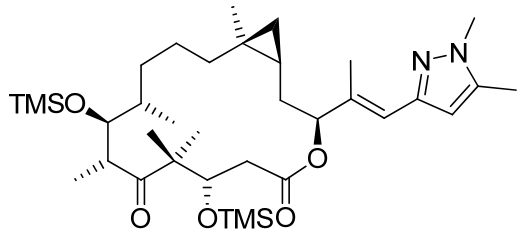
IR (film): ν 3446, 2943, 1732, 1687, 1603, 1456, 1378, 1252, 1147, 1079, 1038, 1009, 983, 770, 670.

HRMS (ESI): calculated for $C_{28}H_{44}NO_6$ $[M+H]^+$ 490.3163, found 490.3156.

Analytical HPLC: Method: eluent B 50-60% (linear gradient from 0-12 min), retention time 9.82 min.







139c

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-((*E*)-1-(1,5-dimethyl-1*H*-pyrazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (139c). To a solution of phosphonium salt **78c** (41.3 mg, 0.119 mmol) in THF (1 mL) was added *n*-BuLi (74 µL, 0.119 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **98** (11.0 mg, 0.020 mmol) in THF (0.5 mL) was added dropwise. The reaction mixture was stirred for 0.5 h at -78 °C, was then allowed to warm to -20 °C and stirred for 1.5 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to afford 10.9 mg (85%) of an inseparable 13:1 mixture of **139c** and its C16-C17 *Z* isomer as a semi-solid.

TLC: R_f 0.35 (hexane/EtOAc 4:1, UV, CPS).

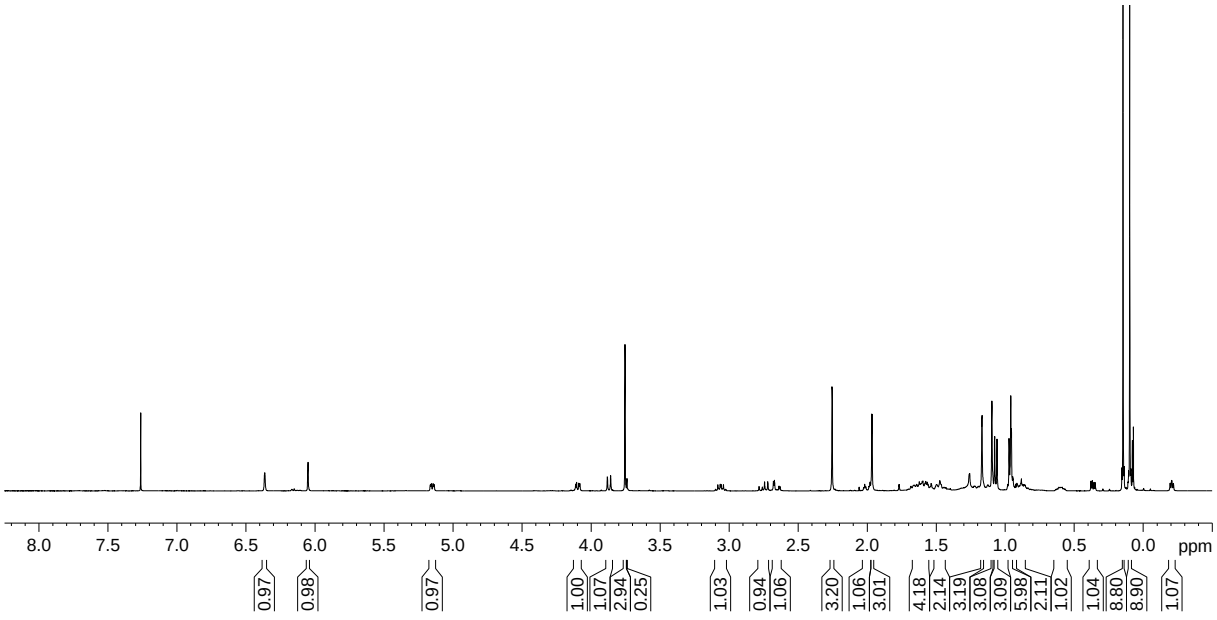
$[\alpha]^{20}_D$: = -7.6° (*c* = 0.55, $CHCl_3$).

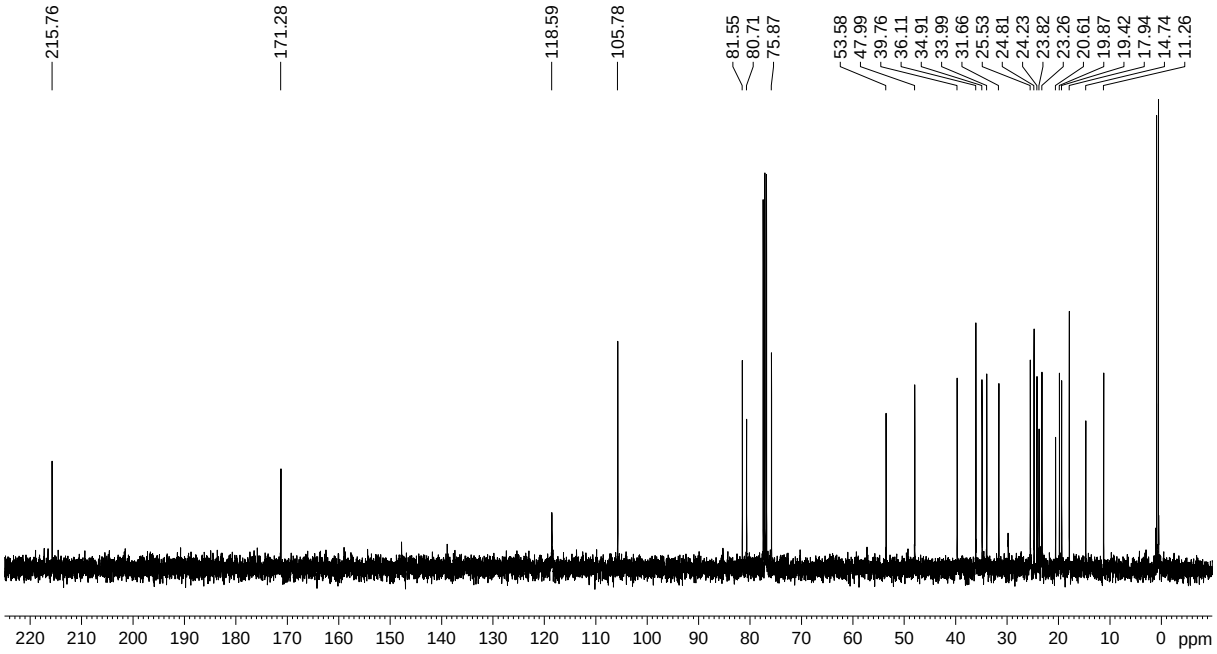
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.37 (s, 1H), 6.05 (s, 1H), 5.15 (dd, *J* = 8.1, 2.8 Hz, 1H), 4.10 (dd, *J* = 9.3, 3.1 Hz, 1H), 3.87 (d, *J* = 9.3 Hz, 1H), 3.75 (s, 3H), 3.06 (dq, *J* = 9.3, 6.8 Hz, 1H), 2.75 (dd, *J* = 16.0, 9.3 Hz, 1H), 2.65 (dd, *J* = 16.0, 3.1 Hz, 1H), 2.25 (d, *J* = 0.6 Hz, 3H), 2.00 (ddd, *J* = 15.4, 3.1, 2.8 Hz, 1H), 1.97 (d, *J* = 1.2 Hz, 3H), 1.69-1.52 (m, 4H), 1.51-1.44 (m, 2H), 1.17 (s, 3H), 1.10 (s, 3H), 1.07 (d, *J* = 6.8 Hz, 3H), 0.96 (d, *J* = 6.8 Hz, 3H), 0.96 (s, 3H), 0.92-0.85 (m, 2H), 0.60 (dddd, *J* = 10.8, 8.7, 5.4, 3.1 Hz, 1H), 0.36 (dd, *J* = 8.7, 4.0 Hz, 1H), 0.15 (s, 9H), 0.10 (s, 9H), -0.20 (dd, *J* = 5.4, 4.0 Hz, 1H).

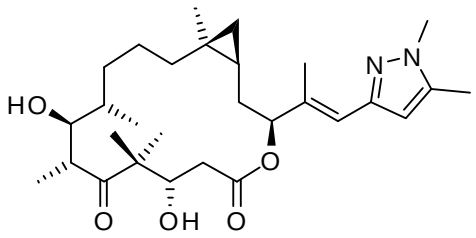
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.8, 171.3, 118.6, 147.8, 139.0, 137.5, 118.5, 105.8, 81.5, 80.7, 75.9, 53.6, 48.0, 39.7, 36.1, 34.9, 34.0, 31.7, 25.5, 24.8, 24.2, 23.8, 23.3, 20.6, 19.9, 19.4, 17.9, 14.8, 11.3, 1.0, 0.6.

IR (film): ν 2951, 1739, 1696, 1550, 1456, 1381, 1251, 1201, 1158, 1114, 1054, 1019, 985, 946, 888, 839, 753.

HRMS (ESI): calculated for $C_{35}H_{63}N_2O_5Si_2$ $[M+H]^+$ 647.4270, found 647.4278.







1c

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-((*E*)-1-(1,5-dimethyl-1*H*-pyrazol-3-yl)prop-1-en-2-yl)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (1c). To a 13:1 mixture of protected cyclopropyl-Epo B **139c** and its C16-C17 *Z* isomer (11.8 mg, 0.018 mmol) in methanol (0.5 mL) was added citric acid (10.7 mg, 0.051 mmol) at room temperature. The reaction mixture was stirred for 8 h, was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 7.1 mg (78%) of **1c** as single isomer as a colorless oil.

TLC: R_f 0.29 (hexane/EtOAc 1:2, UV, CPS).

$[\alpha]^{20}_D$: = -89.4° (*c* = 0.14, $CHCl_3$).

^{1}H-NMR (500 MHz, $CDCl_3$): δ = 6.39 (s, 1H), 6.04 (s, 1H), 5.25 (dd, *J* = 7.3, 5.3 Hz, 1H), 4.04 (dd, *J* = 8.6, 3.0 Hz, 1H), 3.88 (dd, *J* = 4.3, 3.9 Hz, 1H), 3.72 (s, 3H), 3.22 (qd, *J* = 6.8, 4.3 Hz, 1H), 2.77 (br s, 1H), 2.59 (br s, 1H), 2.50 (dd, *J* = 15.3, 8.6 Hz, 1H), 2.42 (dd,

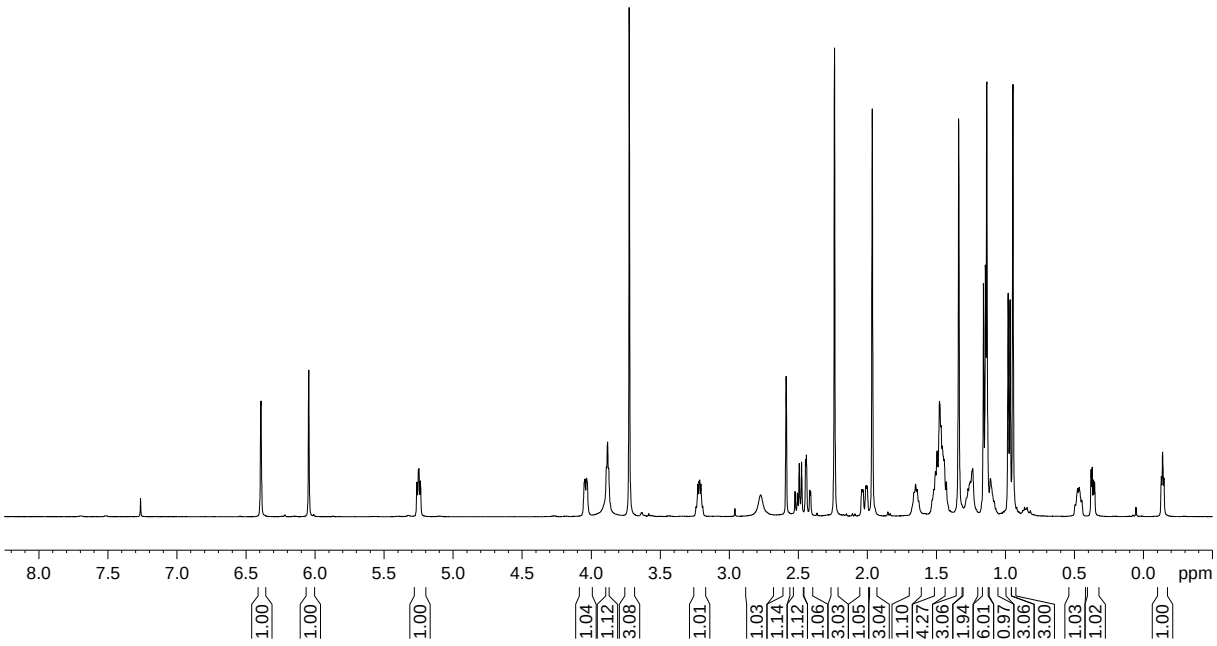
J = 15.3, 3.0 Hz, 1H), 2.24 (s, 3H), 2.02 (ddd, *J* = 14.8, 5.3, 1.7 Hz, 1H), 1.96 (s, 3H), 1.65 (qd, *J* = 6.9, 3.9 Hz, 1H), 1.52-1.43 (m, 5H), 1.34 (s, 3H), 1.28-1.22 (m, 1H), 1.15 (d, *J* = 6.8 Hz, 3H), 1.14 (s, 3H), 1.12-1.05 (m, 1H), 0.97 (d, *J* = 6.9 Hz, 3H), 0.95 (s, 3H), 0.47 (dddd, *J* = 10.8, 8.8, 5.3, 1.7 Hz, 1H), 0.37 (dd, *J* = 8.8, 4.1 Hz, 1H), 0.10 (s, 9H), -0.14 (dd, *J* = 5.3, 4.1 Hz, 1H).

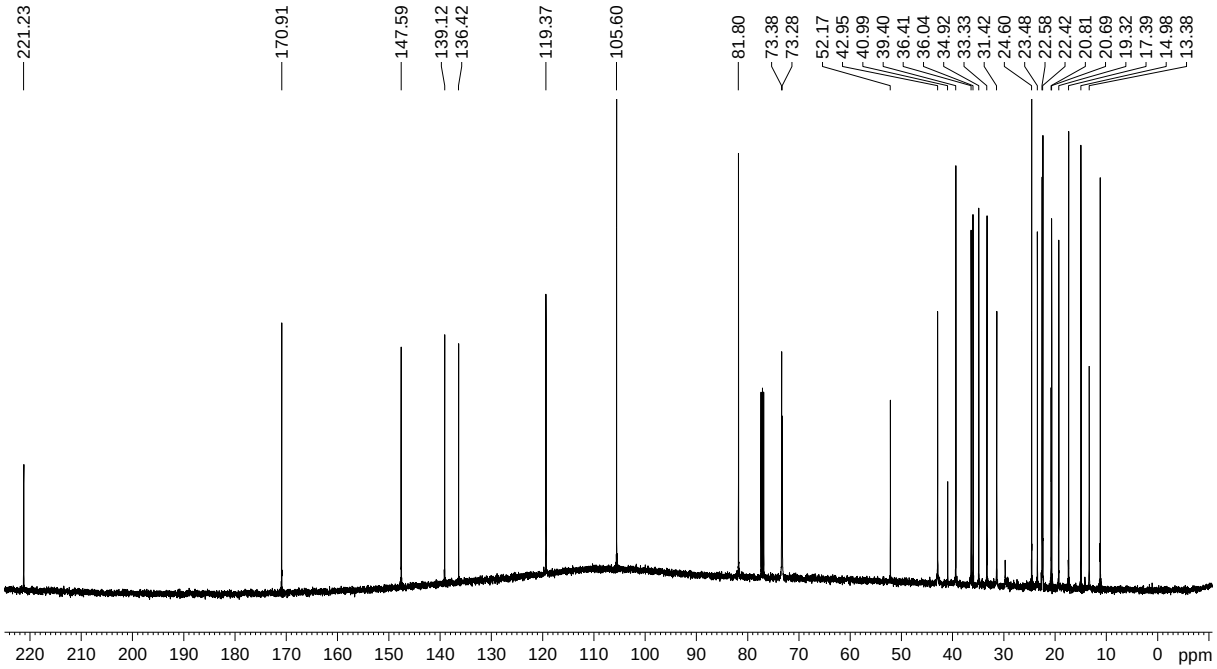
^{13}C-NMR (125 MHz, $CDCl_3$): δ = 221.2, 170.9, 147.6, 139.1, 136.4, 119.4, 105.6, 81.8, 73.4, 73.3, 52.2, 42.9, 41.0, 39.4, 36.4, 36.0, 34.9, 33.3, 31.4, 24.6, 23.5, 22.6, 22.4, 20.8, 20.7, 19.3, 17.4, 15.0, 13.4, 11.2.

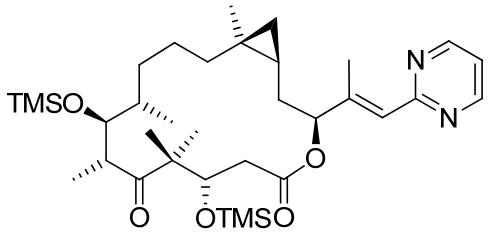
IR (film): ν 3447, 2938, 1727, 1687, 1549, 1458, 1375, 1256, 1016, 870, 803, 754.

HRMS (ESI): calculated for $C_{29}H_{47}N_2O_5$ $[M+H]^+$ 503.3479, found 503.3498.

Analytical HPLC: Method: eluent B 50-60% (linear gradient from 0-12 min), retention time 9.51 min.







139d

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-8,8,10,12,16-pentamethyl-3-((*E*)-1(pyrimidin-2-yl)prop-1-en-2-yl)-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (139d). To a solution of phosphonium salt **78d** (35.8 mg, 0.108 mmol) in toluene (0.5 mL) and THF (0.5 mL) was added *n*-BuLi (62 µL, 0.099 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **98** (5.0 mg, 0.009 mmol) in THF (0.5 mL) was added dropwise. The reaction mixture was stirred for 0.5 h at -78 °C, allowed to reach room temperature and stirred for 12 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 6:1) to afford 5.1 mg (90%) of an inseparable 8:1 mixture of **139d** and its presumed C15 isomer as a colorless oil.

Note: The *Z* isomer was separated but still an isomer is present in the ^{1}H and ^{13}C-NMR. We assume that the center at the C15 isomerised under this relatively harsh conditions (at lower temperature the reaction did not proceed).

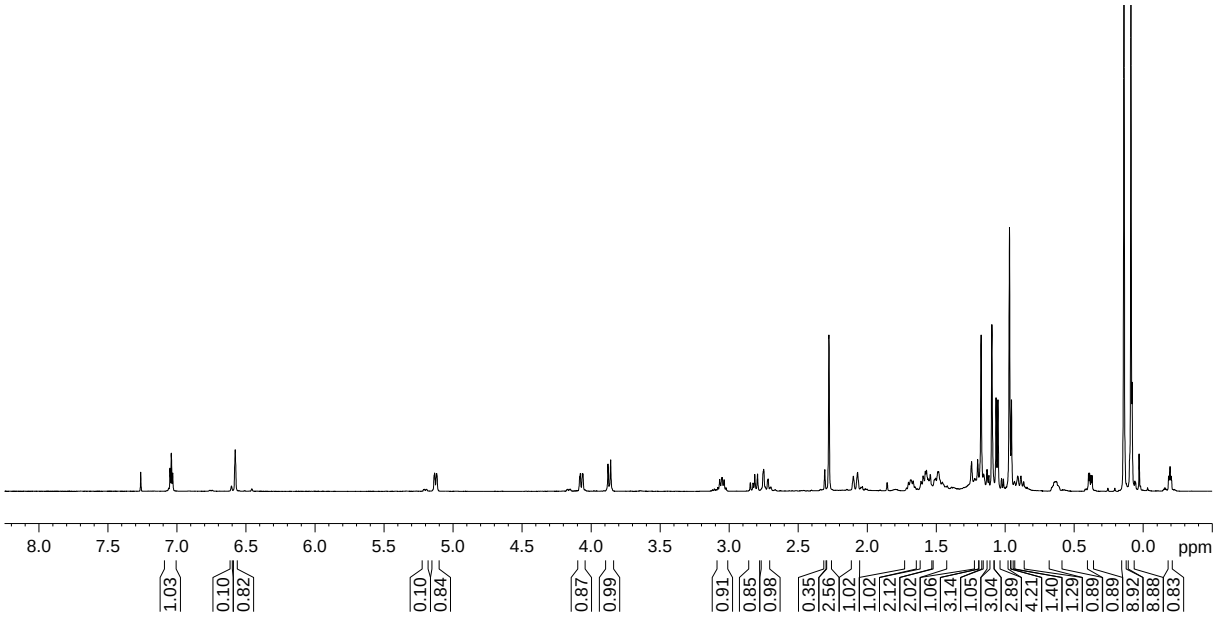
TLC: R_f 0.37 (hexane/EtOAc 4:1, UV, CPS).

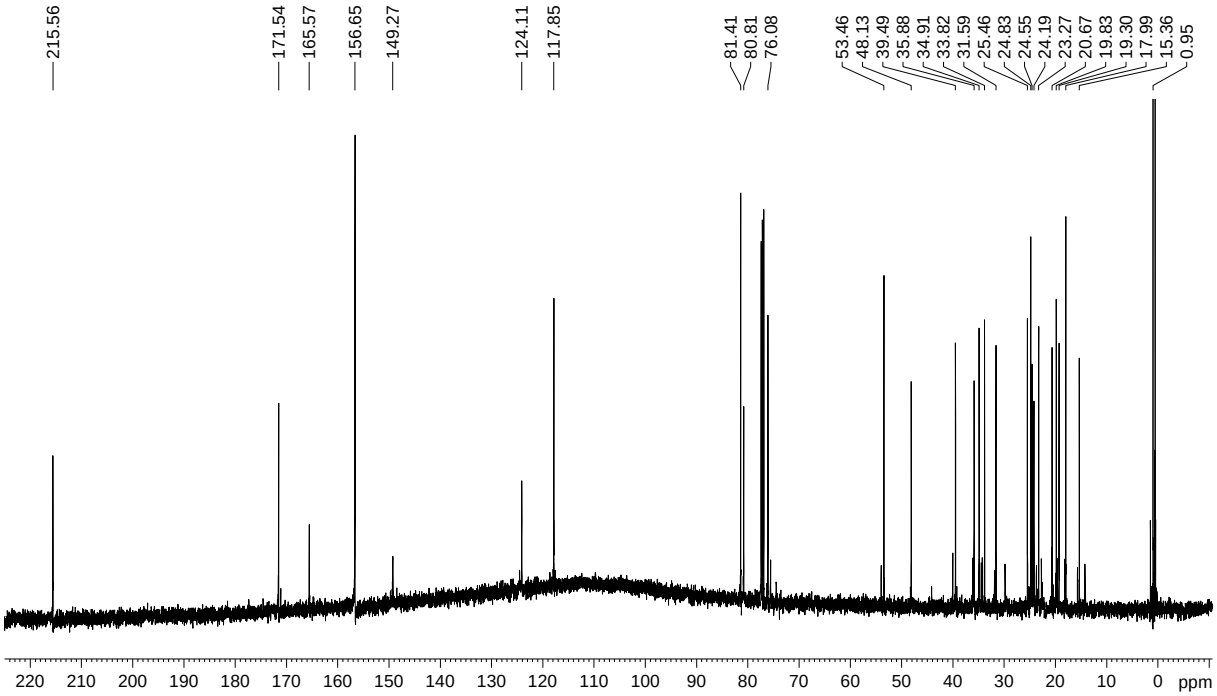
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.69 (d, *J* = 4.9 Hz, 2H), 7.04 (t, *J* = 4.9 Hz, 1H), 6.58 (s, 1H), 5.13 (dd, *J* = 7.8, 2.1 Hz, 1H), 4.07 (dd, *J* = 9.8, 2.1 Hz, 1H), 3.87 (d, *J* = 9.5 Hz, 1H), 3.05 (dq, *J* = 9.5, 6.9 Hz, 1H), 2.82 (dd, *J* = 16.2, 9.8 Hz, 1H), 2.73 (dd, *J* = 16.2, 2.1 Hz, 1H), 2.28 (s, 3H), 2.08 (ddd, *J* = 15.4, 3.2, 2.1 Hz, 1H), 1.68 (dq, *J* = 9.9, 6.9 Hz, 1H), 1.62-1.53 (m, 2H), 1.52-1.41 (m, 2H), 1.23-1.18 (m, 1H), 1.17 (s, 3H), 1.17-1.13 (m, 1H), 1.10 (s, 3H), 1.06 (d, *J* = 6.7 Hz, 1H), 0.97 (s, 3H), 0.96 (d, *J* = 6.9 Hz, 3H), 0.92-0.85 (m, 1H), 0.64 (dddd, *J* = 11.2, 8.7, 5.4, 3.2 Hz, 1H), 0.38 (dd, *J* = 8.7, 4.1 Hz, 1H), 0.14 (s, 9H), 0.11 (s, 9H), -0.19 (dd, *J* = 5.3, 4.1 Hz, 1H).

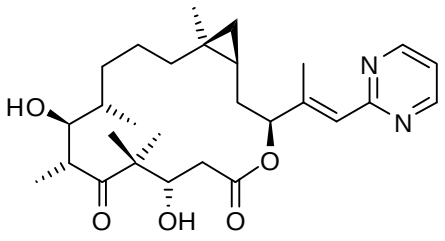
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.6, 171.5, 165.6, 156.6, 149.3, 124.1, 117.8, 81.4, 80.8, 76.1, 53.5, 48.1, 39.5, 35.9, 34.9, 33.8, 31.6, 25.5, 24.8, 24.6, 24.2, 23.3, 20.7, 19.8, 19.3, 18.0, 15.4, 0.9, 0.6.

IR (film): ν □2952, 1738, 1699, 1562, 1461, 1422, 1379, 1255, 1197, 1157, 1116, 1063, 1020, 984, 888, 843, 749.

HRMS (ESI): calculated for $C_{34}H_{59}N_2O_5Si_2$ $[M+H]^+$ 631.3957, found 631.3968.







1d

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-((*E*)-1-(pyrimidin-2-yl)prop-1-en-2-yl)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (1d). To an 8:1 mixture of protected cyclopropyl-Epo B **139d** and its presumed C15 isomer (5.0 mg, 0.008 mmol) in methanol (0.5 mL) was added citric acid (5.0 mg, 0.024 mmol) at room temperature. The reaction mixture was stirred for 12 h, a second portion of citric acid (3.0 mg, 0.014 mmol) was then added and stirring was continued for further 12 h. The reaction mixture was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 3.5 mg (91%) of an 8:1 mixture of **1d** and its presumed C15 isomer as a colorless oil.

Note: The presumed C15 isomer resulting from the Wittig reaction could be separated by preparative HPLC to afford 2.5 mg of **16d**.

TLC: R_f 0.27 (hexane/EtOAc 1:3, UV, CPS).

$[\alpha]^{20}_D$: = -32.3° (*c* = 0.12, $CHCl_3$).

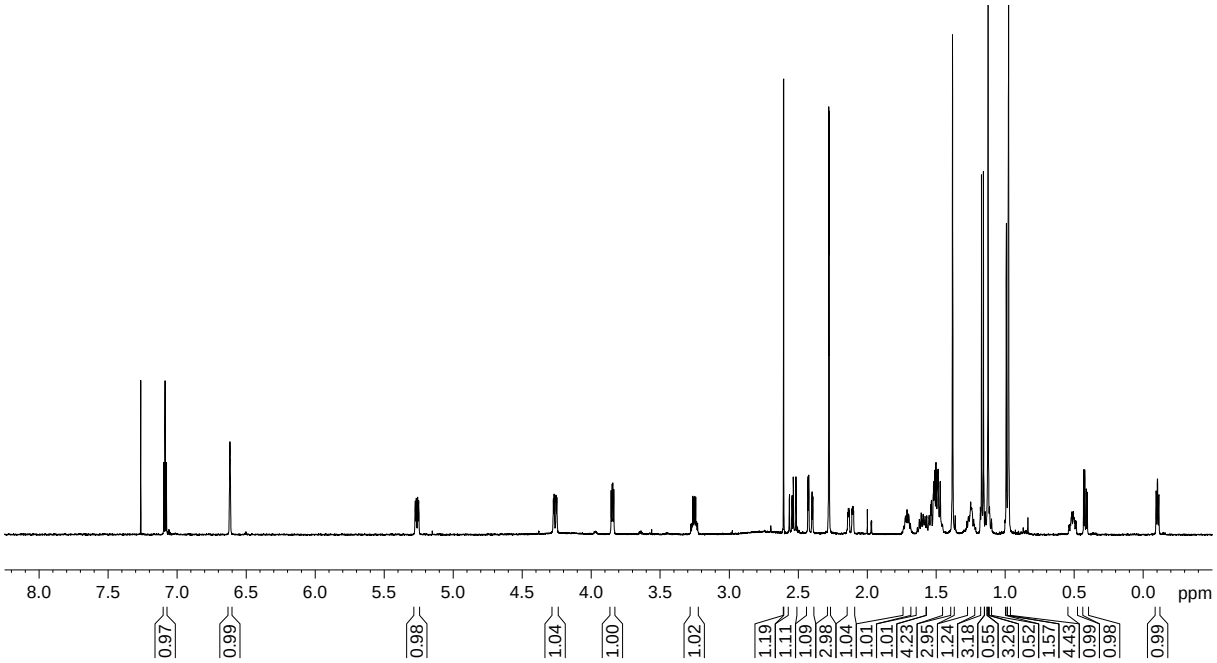
^{1}H-NMR (500 MHz, $CDCl_3$): δ = 8.70 (d, *J* = 4.9 Hz, 2H), 7.09 (t, *J* = 4.9 Hz, 1H), 6.61 (qd, *J* = 1.3, 1.1 Hz, 1H), 5.27 (dd, *J* = 8.9, 3.6 Hz, 1H), 4.26 (dd, *J* = 9.7, 3.0 Hz, 1H), 3.85 (dd, *J* = 5.6, 3.8 Hz, 1H), 3.25 (qd, *J* = 6.9, 3.8 Hz, 1H), 2.61 (s, 1H), 2.54 (dd, *J* = 14.5, 9.7 Hz, 1H), 2.41 (dd, *J* = 14.5, 3.0 Hz, 1H), 2.28 (d, *J* = 1.3 Hz, 3H), 2.12 (ddd, *J* = 15.2, 3.6, 2.0 Hz, 1H), 1.71 (qd, *J* = 6.9, 6.0 Hz, 1H), 1.64-1.57 (m, 1H), 1.56-1.45 (m, 4H), 1.38 (s, 3H), 1.28-1.21 (m, 1H), 1.16 (d, *J* = 6.9 Hz, 3H), 1.16-1.11 (m, 1H), 1.12 (s, 3H), 0.99 (d, *J* = 6.9 Hz, 3H), 0.98 (s, 3H), 0.51 (dddd, *J* = 10.5, 8.9, 5.5, 2.0, 1H), 0.47 (m, 1H), 0.42 (dd, *J* = 8.9, 4.1 Hz, 1H), -0.10 (dd, *J* = 5.5, 4.1 Hz, 1H).

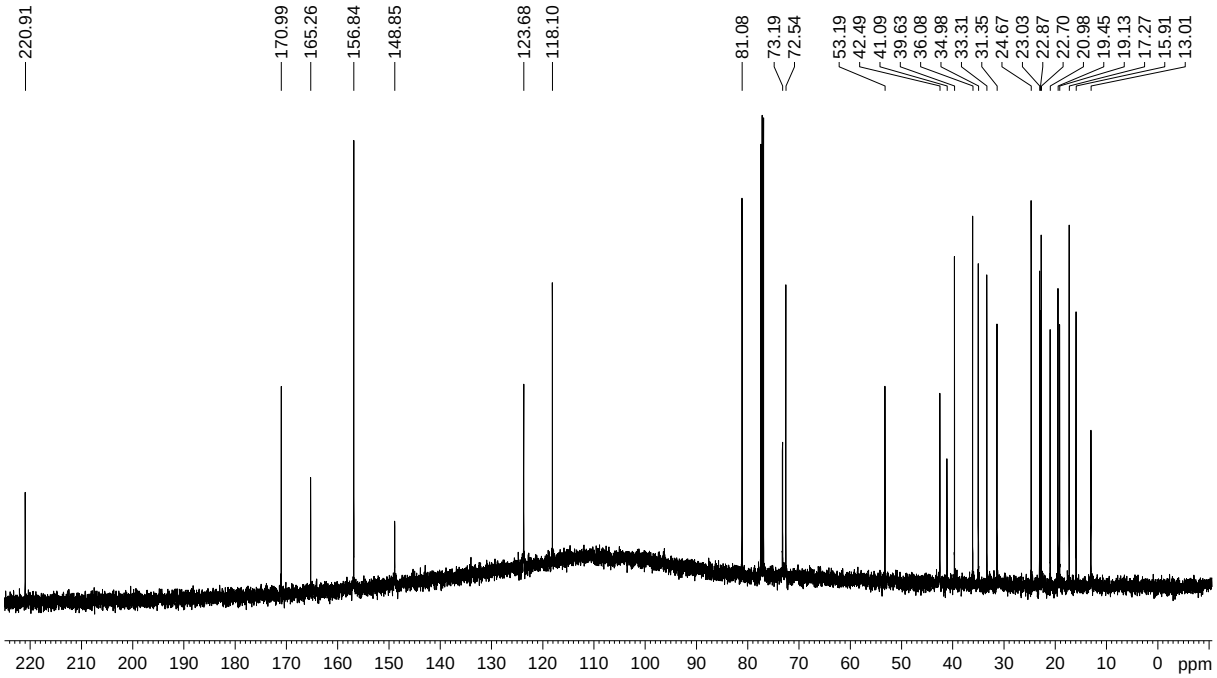
^{13}C-NMR (125 MHz, $CDCl_3$): δ = 220.9, 171.0, 165.3, 156.8, 148.8, 123.7, 118.1, 81.1, 73.2, 72.5, 53.2, 42.5, 41.1, 39.6, 36.1, 35.0, 33.3, 31.3, 24.7, 23.0, 22.9, 22.7, 21.0, 19.4, 19.1, 17.3, 15.9, 13.0.

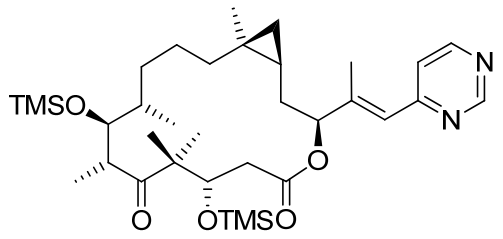
IR (film): ν 3395, 2955, 2927, 2860, 1727, 1688, 1557, 1460, 1423, 1379, 1259, 1074, 1017, 875, 801, 754, 640.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_5$ $[M+H]^+$ 487.3166, found 487.3161.

Analytical HPLC: method: eluent B 45-60% (linear gradient from 0-12 min), retention time 9.33 min.







139e

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-8,8,10,12,16-pentamethyl-3-((*E*)-1-(pyrimidin-4-yl)prop-1-en-2-yl)-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (139e). To a solution of phosphonium salt **78e** (35.8 mg, 0.108 mmol) in toluene (0.5 mL) and THF (0.5 mL) was added *n*-BuLi (68 µL, 0.108 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **98** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise. The reaction mixture was stirred for 0.5 h at -78 °C, allowed to reach room temperature, heated to 75 °C and stirred for 52 h at this temperature. After cooling to room temperature saturated aqueous NH_4Cl (2 mL) was added, the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 6:1) to afford 6.1 mg (54%, 77% brsm) of an inseparable 8:1 mixture of **139e** and its presumed C15 isomer as a colorless oil.

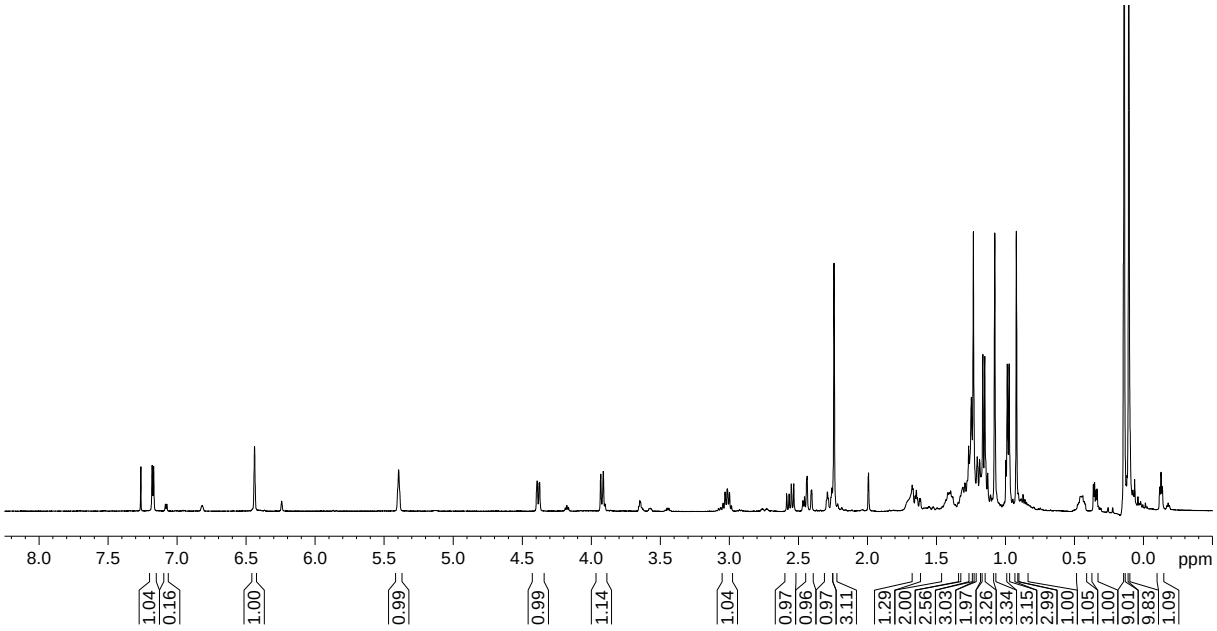
TLC: R_f 0.24 (hexane/EtOAc 4:1, UV, CPS).

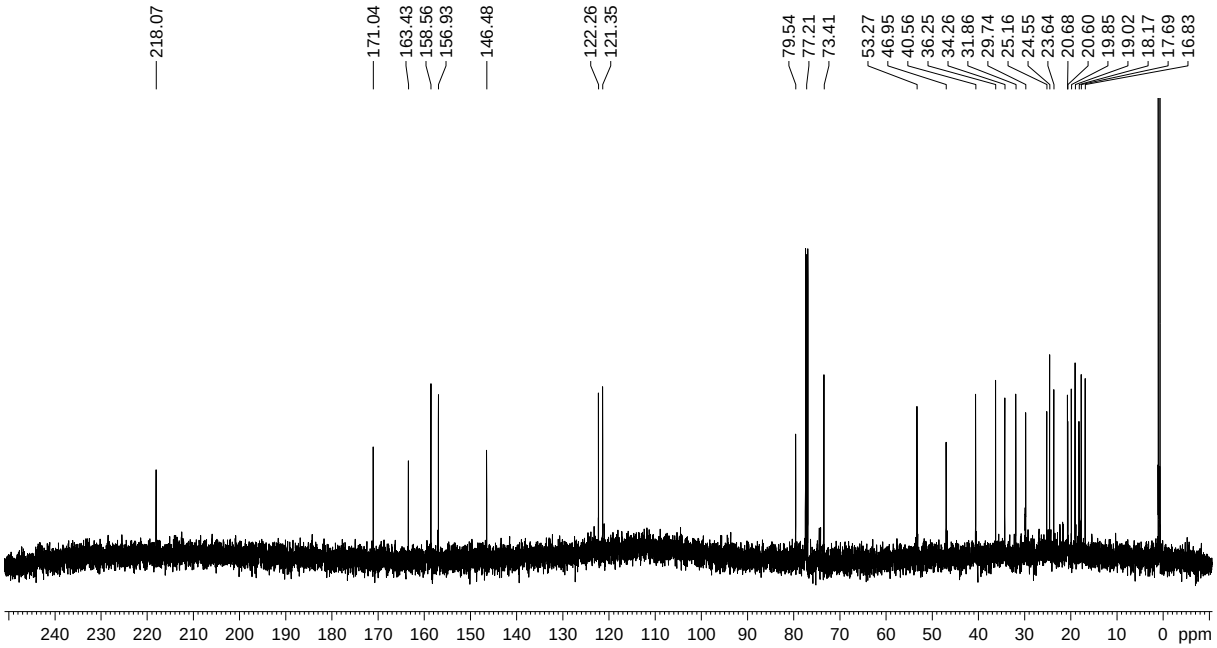
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.17 (s, 1H), 8.65 (d, *J* = 5.1 Hz, 1H), 7.18 (d, *J* = 5.1 Hz, 1H), 6.44 (s, 1H), 5.40 (dd, *J* = 3.5, 3.2 Hz, 1H), 4.38 (dd, *J* = 8.7, 2.1 Hz, 1H), 3.92 (d, *J* = 9.1 Hz, 1H), 3.01 (dq, *J* = 9.1, 6.8 Hz, 1H), 2.56 (dd, *J* = 16.7, 8.7 Hz, 1H), 2.42 (dd, *J* = 16.7, 2.1 Hz, 1H), 2.27 (ddd, *J* = 16.0, 3.2, 3.2 Hz, 1H), 2.24 (s, 3H), 1.65 (ddd, *J* = 16.0, 10.8, 3.5 Hz, 1H), 1.47-1.36 (m, 2H), 1.32-1.26 (m, 2H), 1.23 (s, 3H), 1.21-1.18 (m, 2H), 1.16 (d, *J* = 6.9 Hz, 3H), 1.08 (s, 3H), 0.98 (d, *J* = 6.7 Hz, 3H), 0.92 (s, 3H), 0.90-0.82 (m, 1H), 0.45 (dddd, *J* = 10.8, 8.7, 5.4, 3.2 Hz, 1H), 0.35 (dd, *J* = 8.7, 4.1 Hz, 1H), 0.14 (s, 9H), 0.11 (s, 9H), -0.13 (dd, *J* = 5.4, 4.1 Hz, 1H).

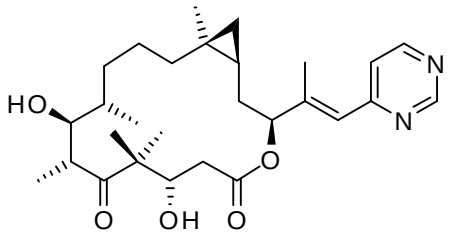
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 171.0, 163.4, 158.6, 156.9, 146.5, 122.3, 121.3, 79.5, 77.2, 73.4, 53.3, 46.9, 40.6, 36.3, 34.3, 31.9, 29.7, 25.2, 24.6, 23.6, 20.7, 20.6, 19.9, 19.0, 18.2, 17.7, 16.8, 1.0, 0.6.

IR (film): ν 2951, 1741, 1695, 1577, 1463, 1383, 1254, 1158, 1114, 1020, 987, 884, 843, 761.

HRMS (ESI): calculated for $C_{34}H_{59}N_2O_5Si_2$ $[M+H]^+$ 631.3957, found 631.3952.







1e

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-((*E*)-1-(pyrimidin-4-yl)prop-1-en-2-yl)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (1e). To an 8:1 mixture of protected cyclopropyl-Epo B **139e** and its presumed C15 isomer (8.0 mg, 0.013 mmol) in methanol (0.5 mL) was added citric acid (8.0 mg, 0.038 mmol) at room temperature. The reaction mixture was stirred for 24 h, was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 5.2 mg (84%) of an 8:1 mixture of **1e** and its presumed C15 isomer as a colorless oil.

Note: The presumed C15 isomer resulting from the Wittig reaction could only be partially separated by preparative HPLC.

TLC: R_f: 0.33 (hexane/EtOAc 1:2, UV, CPS).

$[\alpha]^{20}_D$: = -34.6° (*c* = 0.16, $CHCl_3$).

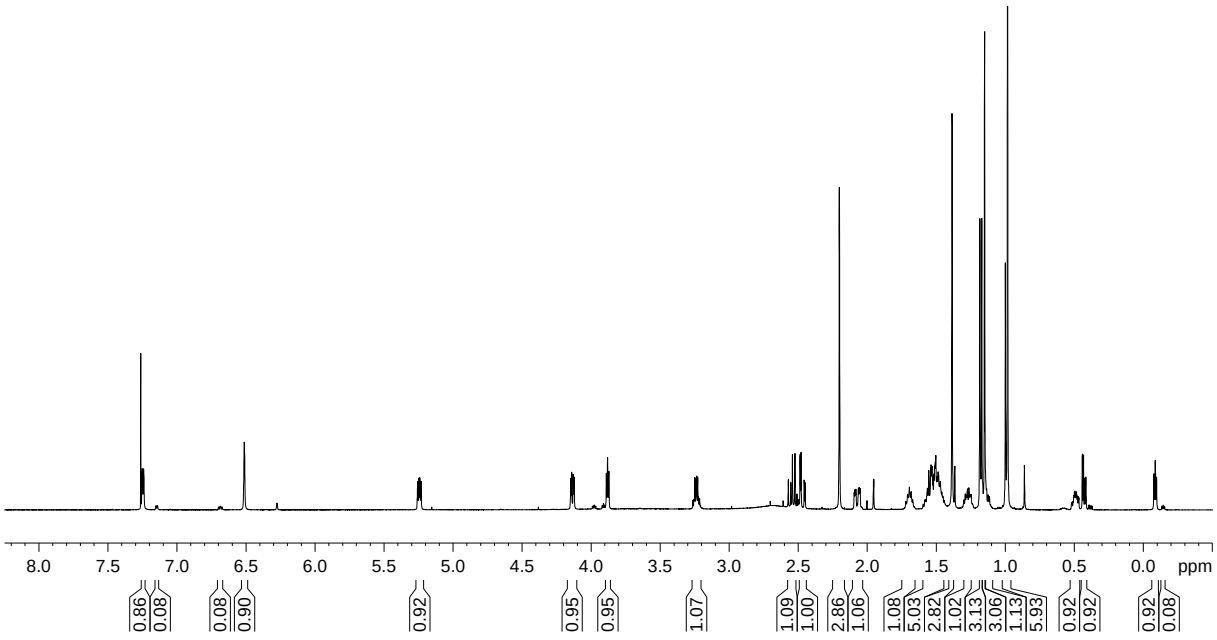
^{1}H-NMR (500 MHz, $CDCl_3$): δ = 9.15 (s, 1H), 8.69 (d, *J* = 3.7 Hz, 1H), 7.25 (d, *J* = 5.1 Hz, 1H), 6.51 (s, 1H), 5.24 (dd, *J* = 8.1, 4.4 Hz, 1H), 4.13 (dd, *J* = 9.0, 3.3 Hz, 1H), 3.88 (dd, *J* = 5.1, 4.1 Hz, 1H), 3.24 (qd, *J* = 6.9, 4.1 Hz, 1H), 2.55 (dd, *J* = 15.0, 9.0 Hz, 1H), 2.46 (dd, *J* = 15.0, 3.3 Hz, 1H), 2.20 (d, *J* = 1.2 Hz, 3H), 2.07 (ddd, *J* = 15.0, 4.4, 2.1 Hz, 1H), 1.69 (qd, *J* = 6.9, 5.1 Hz, 1H), 1.60-1.43 (m, 5H), 1.39 (s, 3H), 1.30-1.24 (m, 1H), 1.18 (d, *J* = 6.9 Hz, 3H), 1.14 (s, 3H), 1.14-1.10 (m, 1H), 0.99 (d, *J* = 6.9 Hz, 3H), 0.98 (s, 3H), 0.49 (dddd, *J* = 10.6, 8.9, 5.4, 2.1 Hz, 1H), 0.43 (dd, *J* = 8.9, 4.1 Hz, 1H), -0.08 (dd, *J* = 5.4, 4.1 Hz, 1H).

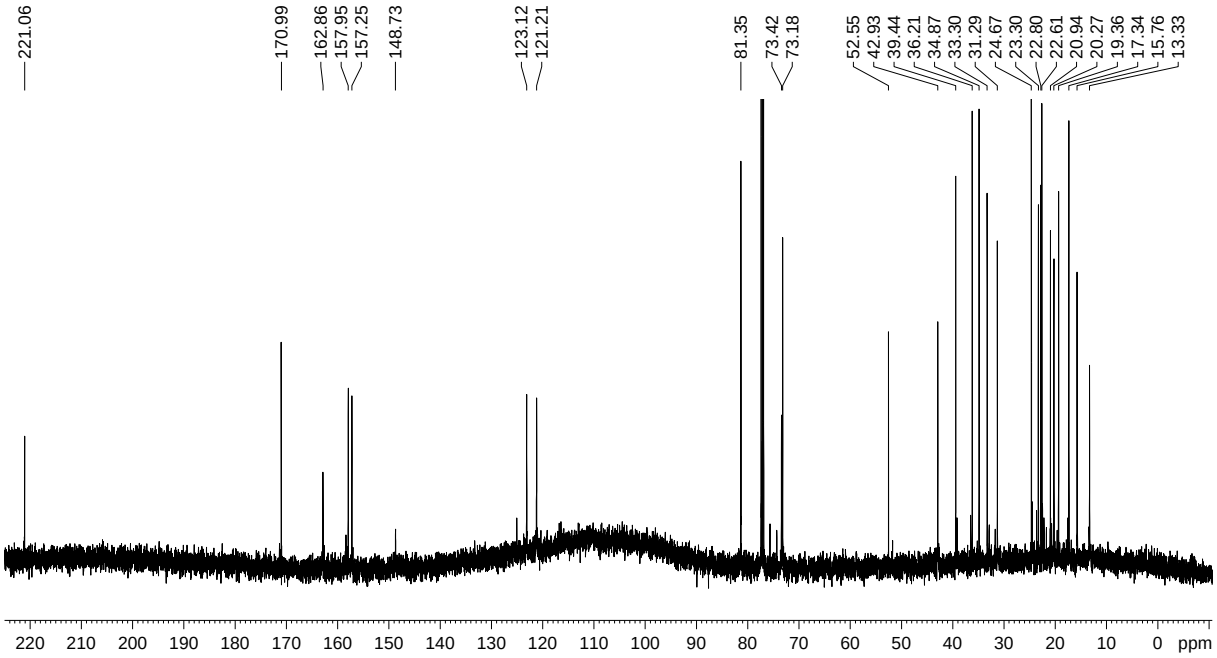
^{13}C-NMR (125 MHz, $CDCl_3$): δ = 221.1, 171.0, 162.9, 157.9, 157.3, 148.7, 123.1, 121.2, 81.4, 73.4, 73.2, 52.5, 42.9, 39.4, 36.2, 34.9, 33.3, 31.3, 24.7, 23.3, 22.8, 22.6, 20.9, 20.3, 19.4, 17.3, 15.8, 13.3.

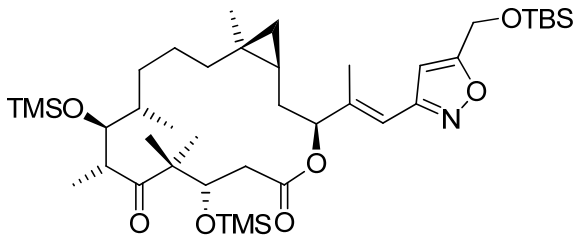
IR (film): ν 3435, 3356, 2928, 2863, 1729, 1685, 1580, 1532, 1462, 1383, 1256, 1149, 1082, 1016, 879, 804, 754, 667.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_5$ $[M+H]^+$ 487.3166, found 487.3174.

Analytical HPLC: Method: eluent B 45% (isokratic), retention time 9.45 min.







142

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-((*E*)-1-(5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (142). To a solution of phosphonate **140b** (85.2 mg, 0.234 mmol) in THF (1.5 mL) was added LiHMDS (1 M in THF, 0.23 mL, 0.234 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Methyl ketone **98** (26.0 mg, 0.047 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to room temperature over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (3 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 27.0 mg (68%) of **142** as a single isomer as a colorless oil.

TLC: R_f: 0.28 (hexane/EtOAc 20:1, UV, CPS).

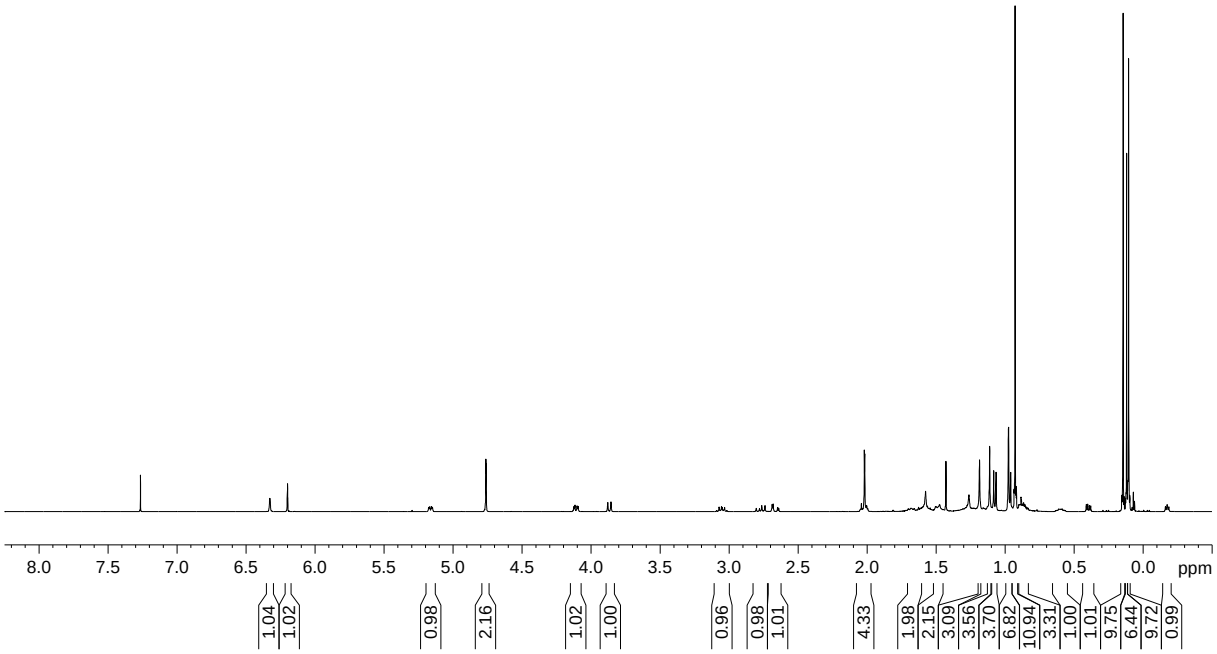
$[\alpha]^{20}_D$: = -5.7° (*c* = 1.23, $CHCl_3$).

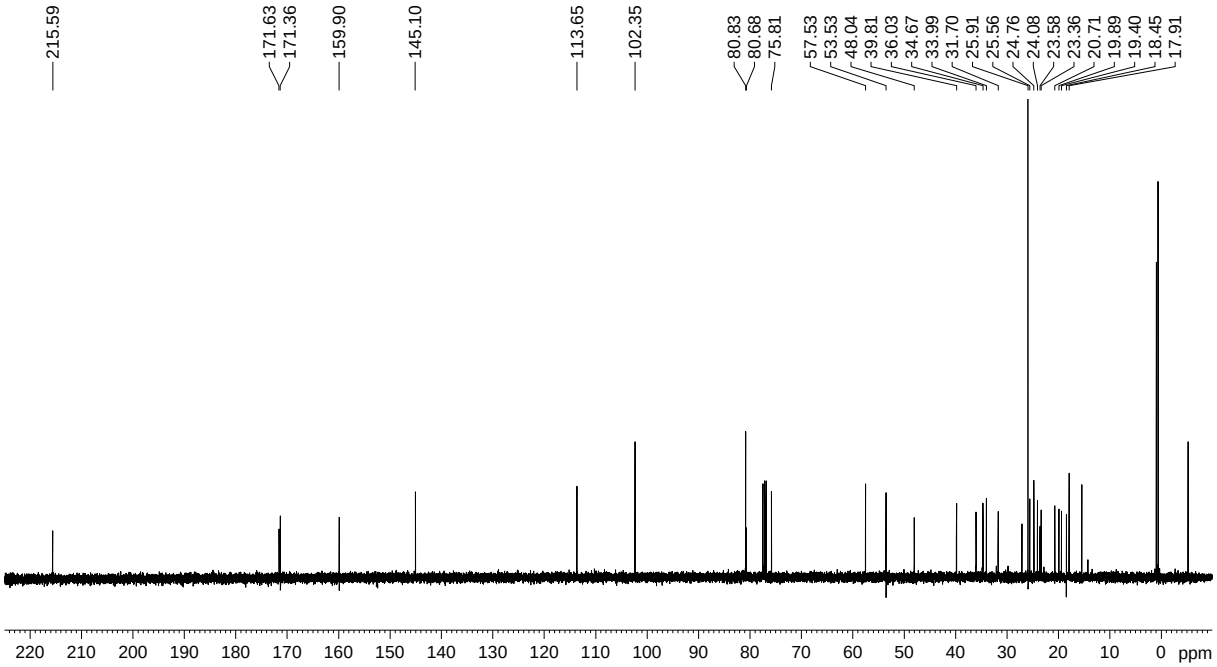
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.33 (q, *J* = 1.2 Hz, 1H), 6.20 (t, *J* = 0.8 Hz, 1H), 5.16 (dd, *J* = 7.9, 2.9 Hz, 1H), 4.76 (d, *J* = 0.8 Hz, 2H), 4.11 (dd, *J* = 9.2, 3.1 Hz, 1H), 3.87 (d, *J* = 9.2 Hz, 1H), 3.05 (dq, *J* = 9.2, 6.8 Hz, 1H), 2.77 (dd, *J* = 16.0, 9.2 Hz, 1H), 2.66 (dd, *J* = 16.0, 3.1 Hz, 1H), 2.02 (d, *J* = 1.2 Hz, 3H), 2.02 (ddd, *J* = 15.3, 3.2, 3.0 Hz, 1H), 1.68 (dq, *J* = 9.2, 6.8 Hz, 1H), 1.61-1.57 (m, 1H), 1.56-1.52 (m, 1H), 1.51-1.47 (m, 1H), 1.251.15 (m, 1H), 1.19 (s, 3H), 1.11 (s, 3H), 1.07 (d, *J* = 6.8 Hz, 3H), 0.98 (s, 3H), 0.97 (d, *J* = 6.8 Hz, 3H), 0.93 (s, 9H), 0.90-0.83 (m, 3H), 0.60 (dddd, *J* = 10.5, 8.7, 5.5, 3.1 Hz, 1H), 0.39 (dd, *J* = 8.7, 4.0 Hz, 1H), 0.15 (s, 9H), 0.12 (s, 6H), 0.11 (s, 9H), -0.17 (dd, *J* = 5.5, 4.0 Hz, 1H).

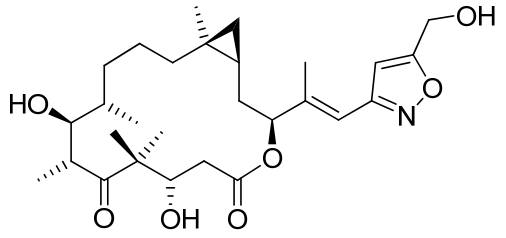
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.6, 171.6, 171.4, 159.9, 145.1, 113.6, 102.3, 80.8, 80.7, 75.8, 57.5, 53.5, 48.0, 39.8, 36.0, 34.7, 34.0, 31.7, 25.9, 25.6, 24.8, 24.1, 23.6, 23.4, 20.7, 19.9, 19.4, 18.4, 17.9, 15.4, 0.9, 0.6, -5.2.

IR (film): ν 2955, 2359, 1742, 1698, 1457, 1380, 1251, 1160, 1115, 1021, 985, 889, 839, 774.

HRMS (ESI): calculated for $C_{40}H_{74}NO_7Si_3$ $[M+H]^+$ 764.4768, found 764.4759.







2A

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-3-((*E*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (2A). To protected cyclopropyl-Epo B **142** (23.0 mg, 0.030 mmol) in CH_2Cl_2 (2 mL) and MeOH (1 mL) was added CSA (7.0 mg, 0.030 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 8 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (5 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 16.0 mg (90%) of **2A** as a white foam.

TLC: R_f 0.13 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -24.5° (*c* = 0.40, $CHCl_3$).

^{1}H-NMR (500 MHz, DMSO-d_6): δ = 6.42 (s, 1H), 6.41 (s, 1H), 5.63 (br s, 1H), 5.13 (dd, *J* = 7.6, 2.7 Hz, 1H), 5.13 (d, *J* = 6.6 Hz, 1H), 4.55 (s, 2H), 4.47 (d, *J* = 6.6 Hz, 1H), 4.07

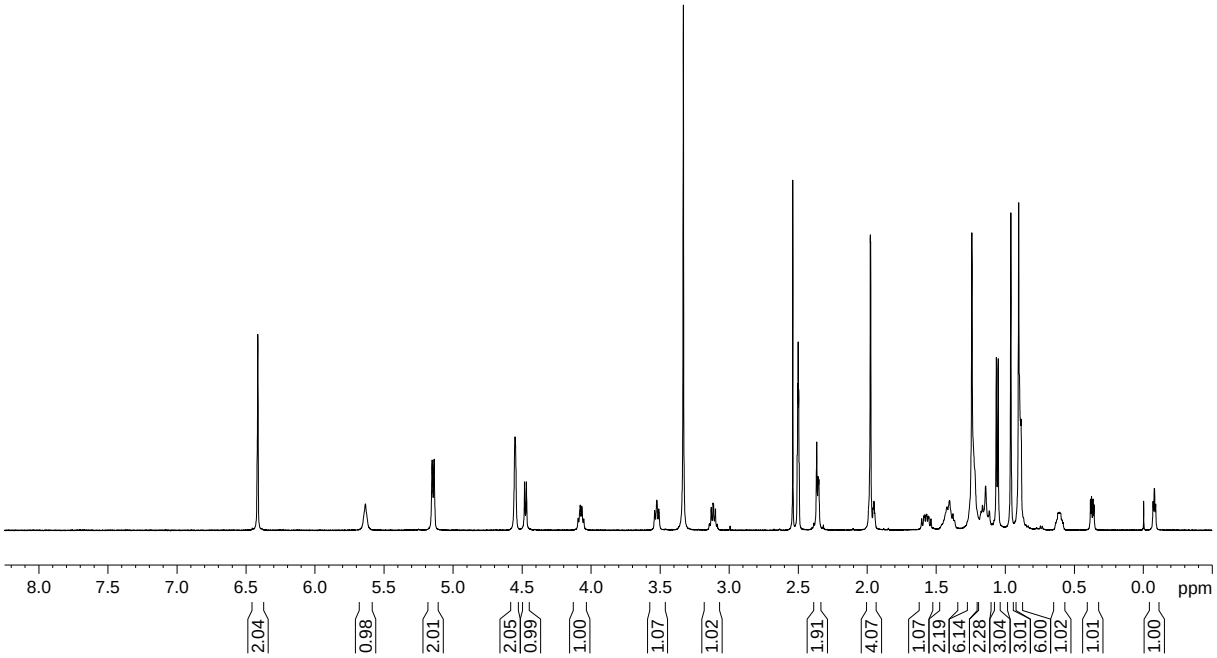
(ddd, *J* = 8.0, 7.2, 5.5 Hz, 1H), 3.52 (dd, *J* = 8.6, 6.6 Hz, 1H), 3.12 (dq, *J* = 8.6, 6.7 Hz, 1H), 2.36 (d, *J* = 8.0 Hz, 1H), 2.36 (d, *J* = 5.5 Hz, 1H), 1.98 (d, *J* = 1.2 Hz, 3H), 1.97 (ddd, *J* = 15.6, 3.4, 2.7 Hz, 1H), 1.57 (ddd, *J* = 15.6, 10.6, 7.6 Hz, 1H), 1.47-1.34 (m, 2H), 1.24 (s, 3H), 1.24-1.20 (m, 3H), 1.17-1.10 (m, 2H), 1.06 (d, *J* = 6.7, 3H), 0.96 (s, 3H), 0.90 (s, 3H), 0.89 (d, *J* = 6.7 Hz, 3H), 0.61 (dddd, *J* =10.6, 8.7, 5.5, 2.7 Hz, 1H), 0.37 (dd, *J* = 8.7, 4.0 Hz, 1H), -0.08 (dd, *J* = 5.5, 4.0 Hz, 1H).

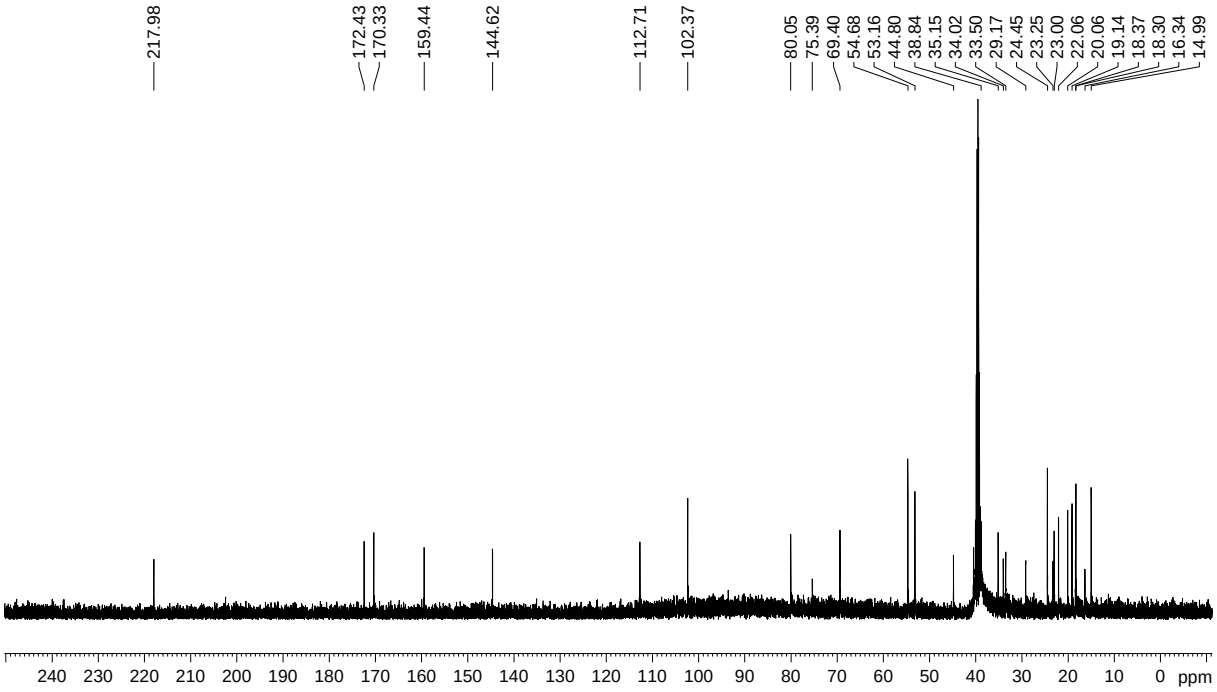
^{13}C-NMR (125 MHz, DMSO-d6): δ = 218.0, 172.4, 170.3, 159.4, 144.6, 112.7, 102.4, 80.0, 75.4, 69.4, 54.7, 53.2, 44.8, 38.8, 35.1, 34.0, 33.5, 29.2, 24.5, 23.3, 23.0, 22.1, 20.1, 19.1, 18.4, 18.3, 16.3, 15.0.

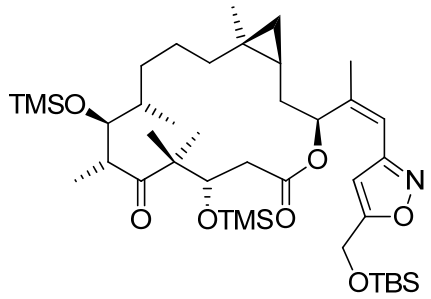
IR (film): ν 3413, 2944, 1729, 1688, 1603, 1456, 1379, 1335, 1253, 1147, 1038, 1008, 983, 958, 937, 755, 668.

HRMS (ESI): calculated for $C_{28}H_{44}NNaO_7$ $[M+Na]^+$ 528.2932, found 528.2948.

Analytical HPLC: Method: eluent B 40-50% (linear gradient from 0-12 min), retention time 8.63 min.







142Z

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-3-((*Z*)-1-(5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (142Z). To a solution of phosphonate **140b** (85.2 mg, 0.234 mmol) in THF (1.5 mL) was added LiHMDS (1 M in THF, 0.23 mL, 0.234 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Methyl ketone **98** (26.0 mg, 0.047 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to room temperature over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (3 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 9.0 mg (25%) of **142Z** as a single isomer as a colorless oil.

TLC: R_f: 0.20 (hexane/EtOAc 20:1, UV, CPS).

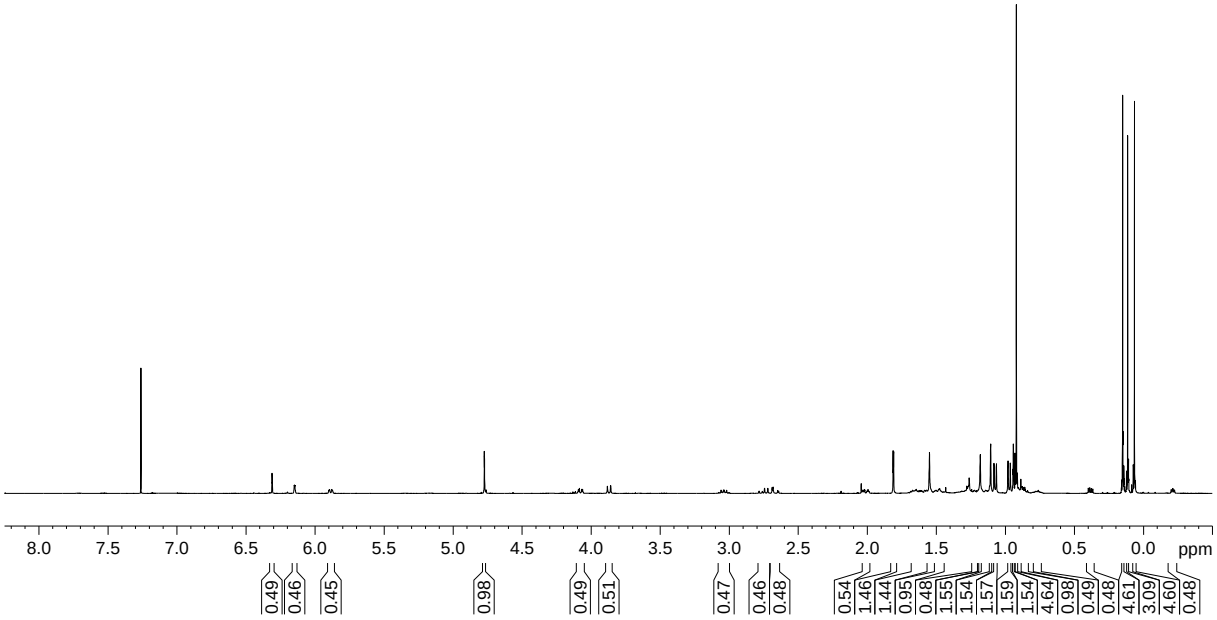
$[\alpha]^{20}_D$: = +15.6° (*c* = 0.50, $CHCl_3$).

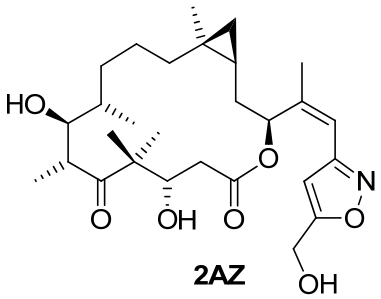
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.31 (t, *J* = 0.8 Hz, 1H), 6.15(q, *J* = 1.5 Hz, 1H), 5.89 (dd, *J* = 8.4, 2.2 Hz, 1H), 4.77 (d, *J* = 0.8 Hz, 2H), 4.08 (dd, *J* = 9.5, 2.9 Hz, 1H), 3.87 (d, *J* = 9.5 Hz, 1H), 3.04 (dq, *J* = 9.5, 6.8 Hz, 1H), 2.75 (dd, *J* = 15.9, 9.5 Hz, 1H), 2.67 (dd, *J* = 15.9, 2.9 Hz, 1H), 2.02 (ddd, *J* = 15.3, 3.1, 2.2 Hz, 1H), 1.81 (d, *J* = 1.5 Hz, 3H), 1.701.56 (m, 3H), 1.52-1.44 (m, 2H), 1.251.20 (m, 1H), 1.18 (s, 3H), 1.11 (s, 3H), 1.07 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.8 Hz, 3H), 0.94 (s, 3H), 0.93 (s, 9H), 0.88-0.83 (m, 2H), 0.77 (dddd, *J* = 10.8, 8.7, 5.6, 3.1 Hz, 1H), 0.38 (dd, *J* = 8.7, 4.1 Hz, 1H), 0.15 (s, 9H), 0.11 (s, 6H), 0.06 (s, 9H), -0.21 (dd, *J* = 5.6, 4.0 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ =

IR (film): ν 2955, 2859, 1742, 1698, 1473, 1457, 1380, 1251, 1160, 1115, 1021, 985, 839, 774.

HRMS (ESI): calculated for $C_{40}H_{74}NO_7Si_3$ $[M+H]^+$ 764.4768, found 764.4764.





2AZ

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-3-((*Z*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (2AZ). To protected cyclopropyl-Epo B C16-C17 *Z* isomer **142Z** (8.0 mg, 0.010 mmol) in

CH_2Cl_2 (0.67 mL) and MeOH (0.33 mL) was added CSA (2.4 mg, 0.010 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 15 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (3 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 3.9 mg (75%) of **2AZ** as a colorless oil.

TLC: R_f 0.13 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = +48.6° (*c* = 0.17, $CHCl_3$).

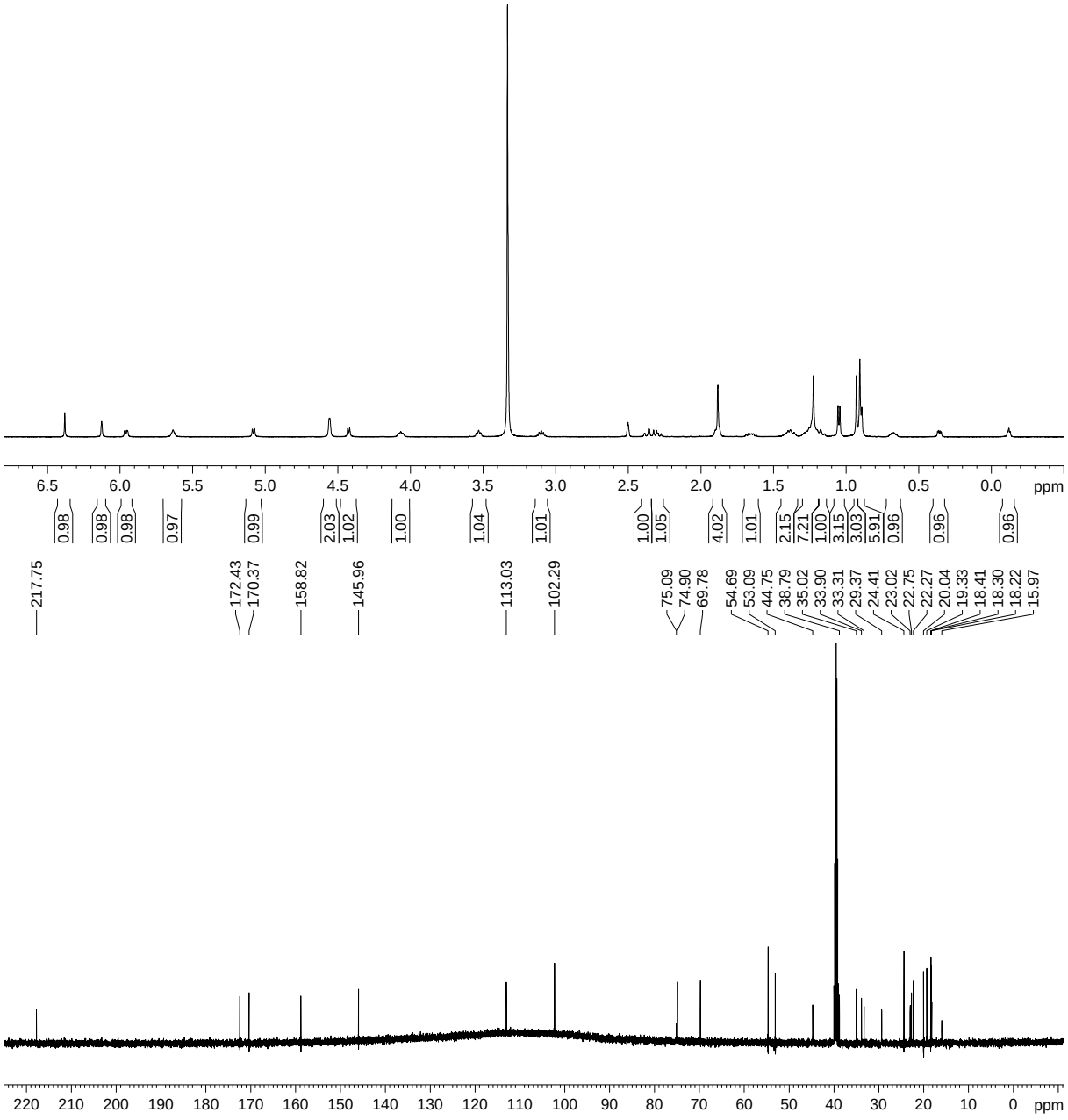
^{1}H-NMR (500 MHz, DMSO-d_6): δ = 6.38 (s, 1H), 6.13 (d, *J* = 1.2 Hz, 1H), 5.96 (dd, *J* = 8.4, 2.3 Hz, 1H), 5.63 (t, *J* = 5.1 Hz, 1H), 5.08 (d, *J* = 7.0 Hz, 1H), 4.56 (s, 2H), 4.43 (d, *J* = 6.6 Hz, 1H), 4.06 (ddd, *J* = 8.4, 6.6, 5.5 Hz, 1H), 3.53 (ddd, *J* = 7.1, 7.0, 7.0 Hz, 1H), 3.10 (qd, *J* = 7.0, 7.0 Hz, 1H), 2.37 (dd, *J* = 15.5, 2.6 Hz, 1H), 2.30 (dd, *J* = 15.5, 10.6 Hz, 1H), 1.89 (ddd, *J* = 14.8, 5.5, 2.3 Hz, 1H), 1.88 (d, *J* = 1.0 Hz, 3H), 1.65 (ddd, *J* = 14.8, 10.3, 8.4 Hz, 1H), 1.44-1.33 (m, 2H), 1.30-1.19 (m, 4H), 1.22 (s, 3H), 1.18-1.14 (m, 1H), 1.05 (d, *J* = 6.6 Hz, 3H), 0.93 (s, 3H), 0.91 (s, 3H), 0.90 (d, *J* = 7.0 Hz, 3H), 0.68 (dddd, *J* = 10.3, 8.8, 5.2, 3.2 Hz, 1H), 0.36 (dd, *J* = 8.8, 4.0 Hz, 1H), -0.12 (dd, *J* = 5.2, 4.0 Hz, 1H);

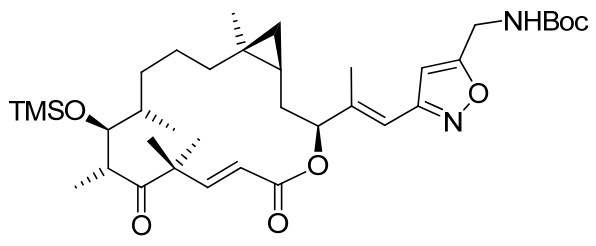
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.7, 172.4, 170.4, 158.8, 146.0, 113.0, 102.3, 75.1, 74.9, 69.8, 54.7, 53.1, 44.7, 38.8, 35.0, 33.9, 33.3, 29.4, 24.4, 23.0, 22.7, 22.3, 20.0, 19.3, 18.4, 18.3, 18.2, 16.0;

IR (film): ν 3429, 3405, 2942, 2875, 1719, 1687, 1604, 1456, 1380, 1289, 1258, 1175, 1148, 1073, 1052, 1032, 1009, 984, 960, 757.

HRMS (ESI): calculated for $C_{28}H_{44}NO_7$ $[M+H]^+$ 506.3112, found 506.3124.

Analytical HPLC: Method: eluent B 40-50% (linear gradient from 0-12 min), retention time 9.26 min.





143

***tert*-butyl ((3-((*E*)-2-((1*S*,3*S*,10*R*,11*S*,12*S*,16*S*,*E*)-8,8,10,12,16-pentamethyl-5,9-dioxo-11-(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-6-en-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methyl)carbamate (143).** To a solution of phosphonate **141** (56.7 mg, 0.163 mmol) in

THF (1.5 mL) was added n-BuLi (1.6 M in hexane, 102 µL, 0.163 mmol) dropwise at -78 °C and the reaction mixture was stirred for 0.5 h at this temperature. Methyl ketone **98** (15.0 mg, 0.033 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to room temperature over a period of 2 h and was then stirred for 10 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 8:1) to afford 5.0 mg (28%) of an inseparable 4:1 mixture of **143** and its C16-C17 *Z* isomer as a colorless oil.

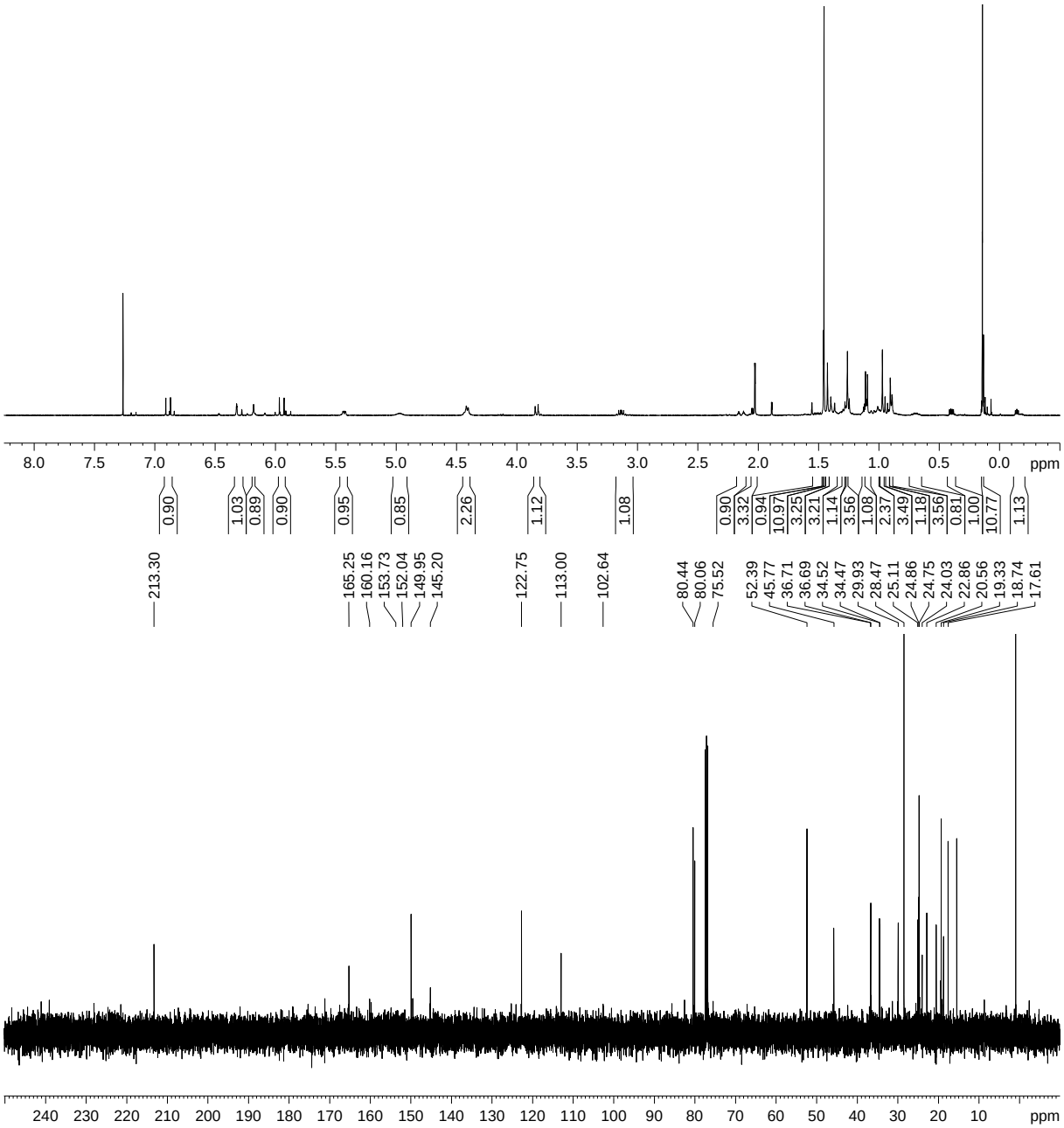
TLC: R_f 0.33 (hexane/EtOAc 4:1, UV, CPS).

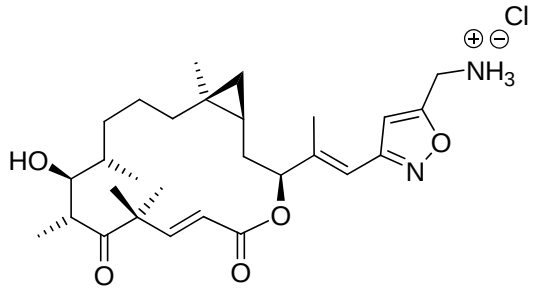
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.89 (d, *J* = 15.9 Hz, 1H), 6.31 (q, *J* = 1.2 Hz, 1H), 6.18 (s, 1H), 5.95 (d, *J* = 15.9 Hz, 1H), 5.43 (dd, *J* = 6.9, 2.4 Hz, 1H), 4.97 (br s, 1H), 4.42 (dd, *J* = 6.2, 1.1 Hz, 2H), 3.83 (d, *J* = 9.6 Hz, 1H), 3.13 (dq, *J* = 9.6, 6.8 Hz, 1H), 2.14 (ddd, *J* = 15.6, 2.6, 2.4 Hz, 1H), 2.03 (d, *J* = 1.2 Hz, 3H), 1.51 (ddd, *J* = 15.6, 11.5, 6.9 Hz, 1H), 1.46 (s, 9H), 1.43 (s, 3H), 1.43-1.37 (m, 2H), 1.31-1.25 (m, 1H), 1.26 (s, 3H), 1.14-1.07 (m, 1H), 1.10 (d, *J* = 6.8 Hz, 3H), 1.08-0.98 (m, 2H), 0.97 (s, 3H), 0.95-0.92 (m, 1H), 0.90 (d, *J* = 6.7 Hz, 3H), 0.70 (dddd, *J* = 11.5, 8.9, 5.5, 2.6 Hz, 1H), 0.40 (dd, *J* = 8.9, 4.3 Hz, 1H), 0.14 (s, 9H), -0.14 (dd, *J* = 5.5, 4.3 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ 213.3, 165.2, 160.2, 153.7, 152.0, 150.0, 145.2, 122.8, 113.0, 102.6, 80.4, 80.1, 75.5, 52.4, 45.8, 36.7, 36.7, 34.5 34.5, 29.9, 28.5 ,25.1, 24.9, 24.8, 24.0, 22.9, 20.6, 19.3, 18.7, 17.6, 15.5, 1.0.

IR (film): ν 3367, 2953, 2875, 1703, 1644, 1602, 1509, 1455, 1367, 1293, 1250, 1171, 1157, 1118, 1084, 1048, 1024, 988, 910, 887, 838, 791, 728.

HRMS (ESI): calculated for $C_{36}H_{58}N_2NaO_7Si$ $[M+Na]^+$ 681.3905, found 681.3914.





144

(3-((*E*)-2-((1*S*,3*S*,10*R*,11*S*,12*S*,16*S*,*E*)-11-hydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadec-6-en-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methanaminium chloride (144). To protected cyclopropyl-Epo B **143** (19.0 mg, 0.025 mmol) was added HCl

(2.25 mL, 2.253 mmol) at room temperature and the reaction mixture was stirred for 14 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5) to yield 9.0 mg (69%) of **144** as a semi solid.

TLC: R_f 0.22 ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5, UV, CPS).

$[\alpha]^{20}_D$: = +78.6° (*c* = 0.29, $CHCl_3$).

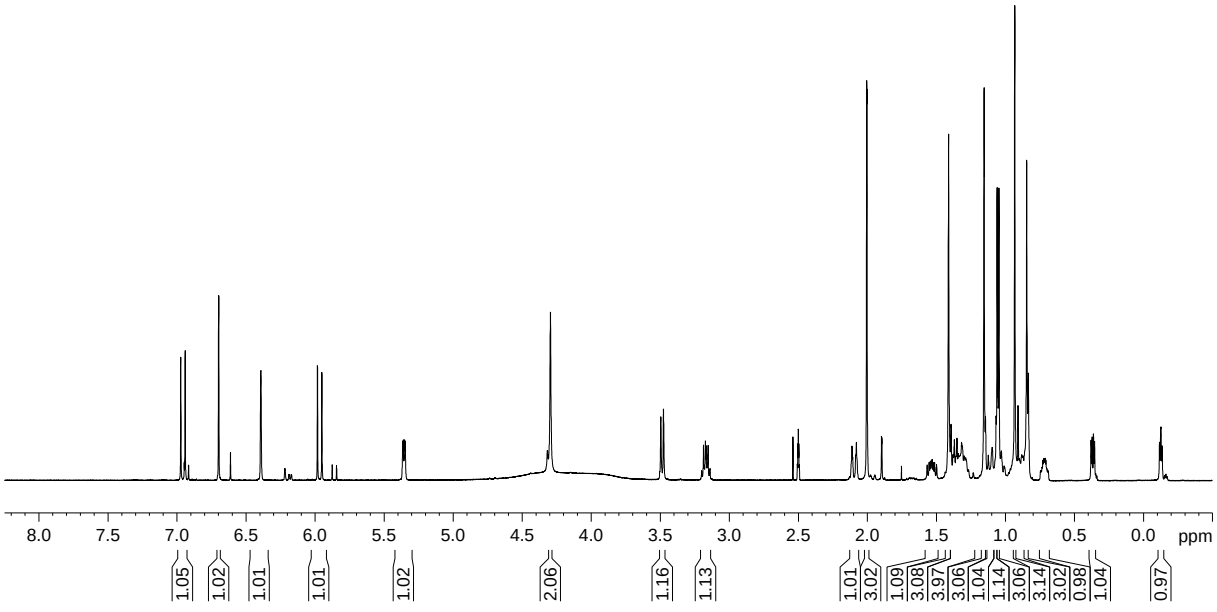
^{1}H-NMR (500 MHz, DMSO-d6): δ = 8.62 (br s, 3H), 6.96 (d, *J* = 15.9 Hz, 1H), 6.70 (s, 1H), 6.39 (q, *J* = 1.1 Hz, 1H), 5.97 (d, *J* = 15.9 Hz, 1H), 5.36 (dd, *J* = 6.9, 2.1 Hz, 1H), 4.30 (s, 2H), 3.49 (d, *J* = 9.7 Hz, 1H), 3.17 (dq, *J* = 9.7, 6.7 Hz, 1H), 2.10 (ddd, *J* = 15.6, 2.7, 2.1 Hz, 1H), 2.00 (d, *J* = 1.1 Hz, 3H), 1.53 (ddd, *J* = 15.6, 11.5, 6.9 Hz, 1H), 1.41 (s, 3H), 1.40-1.26 (m, 4H), 1.15 (s, 3H), 1.12-1.07 (m, 1H), 1.07-1.01 (m, 1H), 1.05 (d, *J* = 6.7 Hz, 3H), 1.04-1.00 (m, 1H), 0.93 (s, 3H), 0.85 (s, 3H), 0.72 (dddd, *J* = 11.5, 8.6, 5.5, 2.7 Hz, 1H), 0.37 (dd, *J* = 8.6, 4.0 Hz, 1H), -0.13 (dd, *J* = 5.5, 4.0 Hz, 1H).

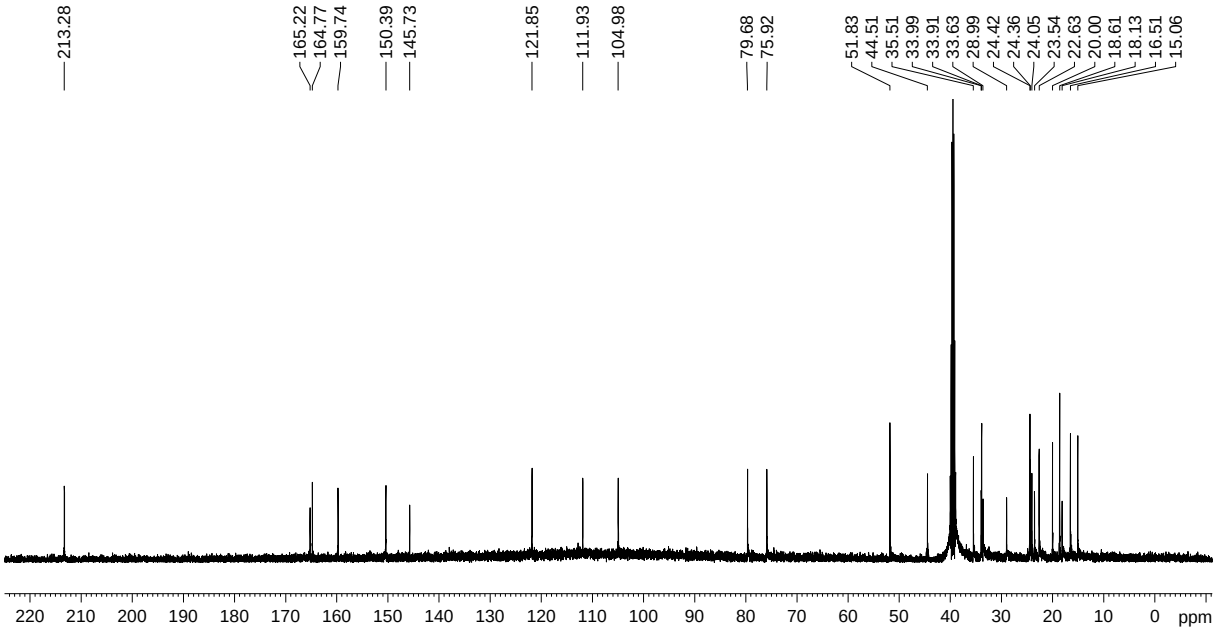
^{13}C-NMR (125 MHz, DMSO-d6): δ = 213.3, 165.2, 164.8, 159.7, 150.4, 145.7, 121.9, 111.9, 105.0, 79.7, 75.9, 51.8, 44.5, 35.5, 34.0, 33.9, 33.6, 29.0, 24.4, 24.4, 24.0, 23.5, 22.6, 20.0, 18.6, 18.1, 16.5, 15.1.

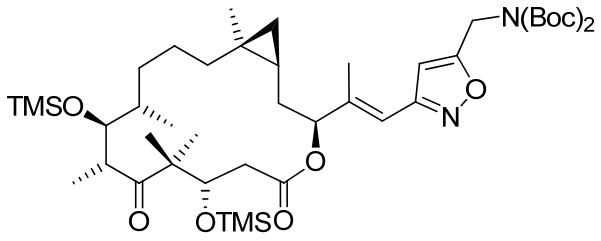
IR (film): ν 3455, 2940, 1700, 1679, 1453, 1377, 1337, 1262, 1201, 1179, 1136, 1050, 1024, 984, 876, 835, 799, 756, 722.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_5S$ $[M+H]^+$ 487.3166, found 487.3162.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 30-50% (linear gradient from 0-12 min), retention time 7.81 min.







146

Di-*tert*-butyl ((3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-8,8,10,12,16-pentamethyl-5,9-dioxo-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadecan-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methyl)dicarbamate (146). To a solution of phosphonate **145g** (48.5 mg, 0.108 mmol) in THF (1.5 mL) was added LiHMDS (1.0 M in THF, 108 µL, 0.108 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Methyl ketone **98** (20.0 mg, 0.036 mmol) in THF (1.0 mL) was added dropwise and the reaction mixture was allowed to warm to room temperature over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (4 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 15.0 mg (52%) of **146** as a single isomer as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 9:1, UV, CPS).

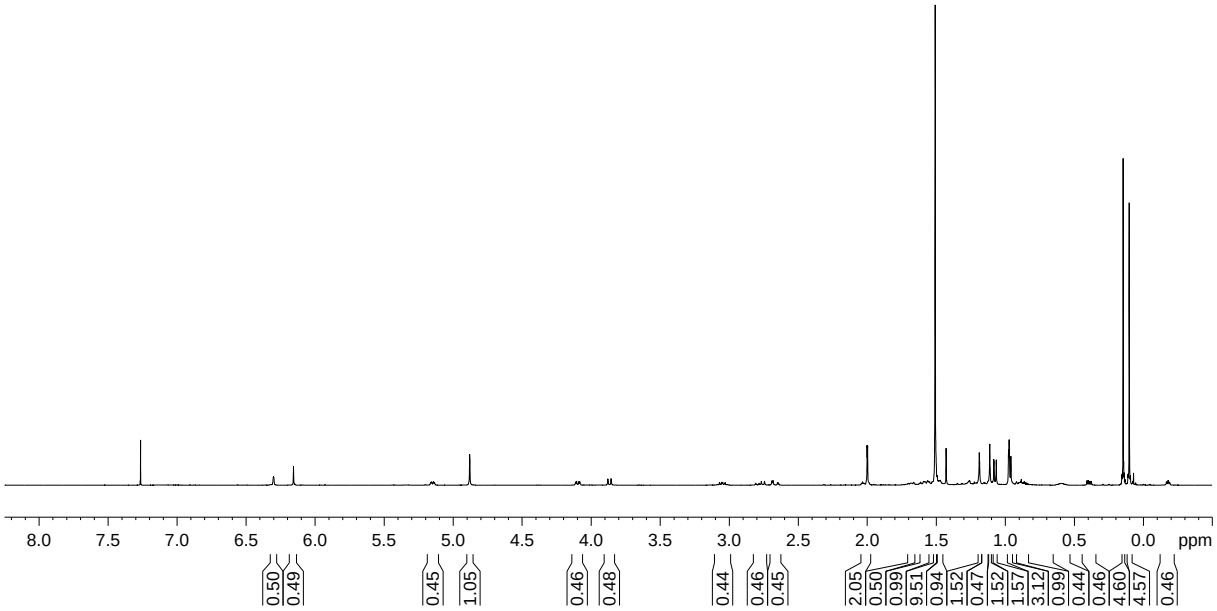
$[\alpha]^{20}_D$: = -4.6° (*c* = 0.75, $CHCl_3$).

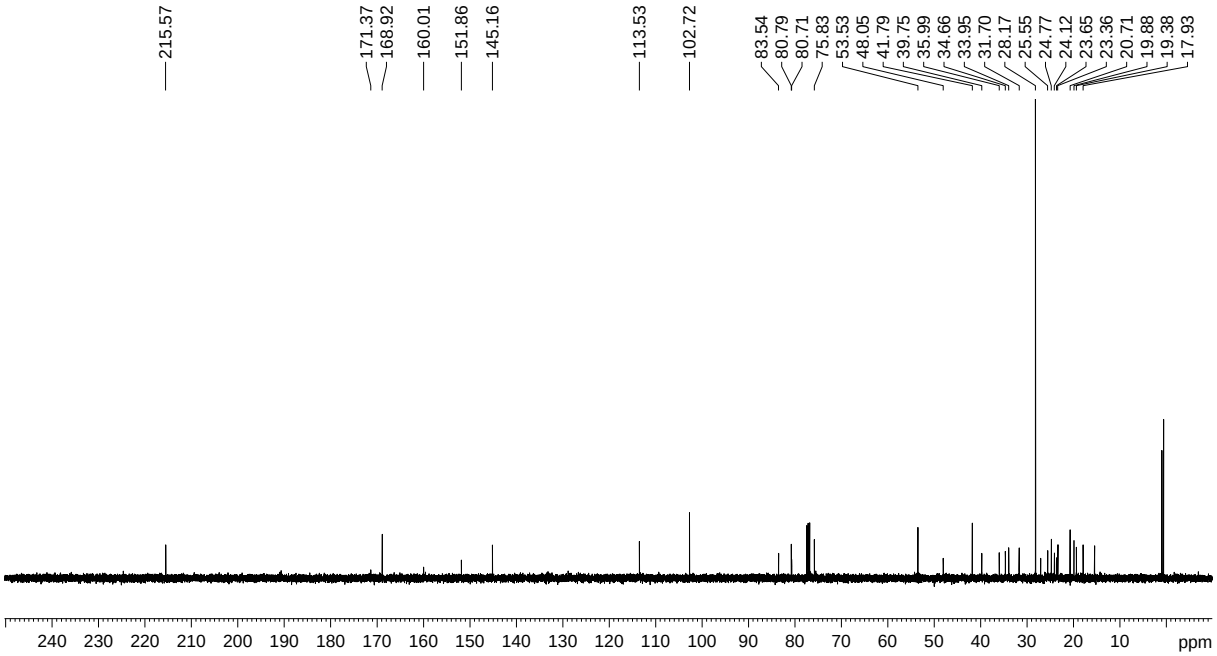
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.30 (q, *J* = 1.2 Hz, 1H), 6.16 (s, 1H), 5.15 (dd, *J* = 7.9, 2.8 Hz, 1H), 4.88 (s, 2H), 4.10 (dd, *J* = 9.3, 3.0 Hz, 1H), 3.87 (d, *J* = 9.3 Hz, 1H), 3.05 (dq, *J* = 9.3, 6.8 Hz, 1H), 2.78 (dd, *J* = 16.0, 9.3 Hz, 1H), 2.66 (dd, *J* = 16.0, 3.0 Hz, 1H), 2.00 (d, *J* = 1.2 Hz, 3H), 2.01 (ddd, *J* = 15.6, 3.6, 2.8 Hz, 1H), 1.71-1.65 (m, 1H), 1.62-1.54 (m, 2H), 1.51 (s, 18H), 1.49-1.45 (m, 2H), 1.19 (s, 3H), 1.17-1.12 (m, 1H), 1.11 (s, 3H), 1.07 (d, *J* = 6.8 Hz, 3H), 0.97 (s, 3H), 0.97 (d, *J* = 6.7 Hz, 3H), 0.92-0.83 (m, 2H), 0.59 (dddd, *J* = 8.7, 5.5 Hz, 1H), 0.39 (dd, *J* = 8.7, 4.0 Hz, 1H), 0.15 (s, 9H), 0.10 (s, 9H), -0.18 (dd, *J* = 5.5, 4.0 Hz, 1H).

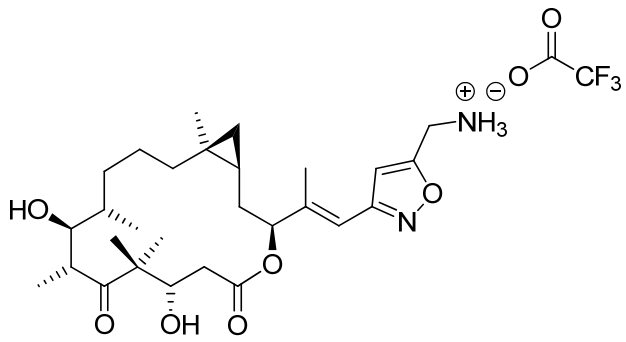
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.6, 171.4, 168.9, 160.0, 151.9, 145.2, 113.5, 102.7, 83.5, 80.8, 80.7, 75.8, 53.5, 48.0, 41.8, 39.8, 36.0, 34.7, 34.0, 31.7, 28.2, 25.6, 24.8, 24.1, 23.6, 23.4, 20.7, 19.9, 19.4, 17.9, 15.5, 0.9, 0.6.

IR (film): ν 2956, 1795, 1473, 1700, 1457, 1386, 1367, 1344, 1306, 1251, 1141, 1115, 1020, 986, 948, 889, 842, 767, 749.

HRMS (ESI): calculated for $C_{44}H_{76}N_2NaO_{10}Si_2$ $[M+H]^+$ 871.4931, found 871.4947.







2B

(3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadecan-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methanaminium 2,2,2-trifluoroacetate (2B). To protected cyclopropyl-Epo B **146** (15.0 mg, 0.018 mmol) was added TFA (0.67 µL, 0.875 mmol) at room temperature and the reaction mixture was stirred for 17 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc/MeOH/NEt_3 90:10:0.5) to yield 9.5 mg (94%) of **2B** as a colorless oil.

TLC: R_f 0.17 (EtOAc/MeOH/NEt_3 90:10:0.5, UV, CPS).

$[\alpha]^{20}_D$: = -20.8° (*c* = 0.53, $CHCl_3$).

^{1}H-NMR (500 MHz, DMSO-d6): δ = 8.77 (br s, 3H), 6.67 (s, 1H), 6.46 (s, 1H), 5.23 (s, 1H), 5.14 (dd, *J* = 7.2, 3.1 Hz, 1H), 4.83 (s, 2H), 4.08 (dd, *J* = 7.2, 6.3 Hz, 1H), 3.53 (d, *J* = 8.7 Hz, 1H), 3.46 (s, 1H), 3.12 (dq, *J* = 8.7, 6.7 Hz, 1H), 2.36 (d, *J* = 6.3 Hz, 1H), 2.36 (d, *J* = 7.2 Hz, 1H), 1.98 (s, 3H), 1.97 (ddd, *J* = 15.6, 3.2, 3.1 Hz, 1H), 1.59 (ddd, *J* = 15.6, 10.5, 7.2 Hz,

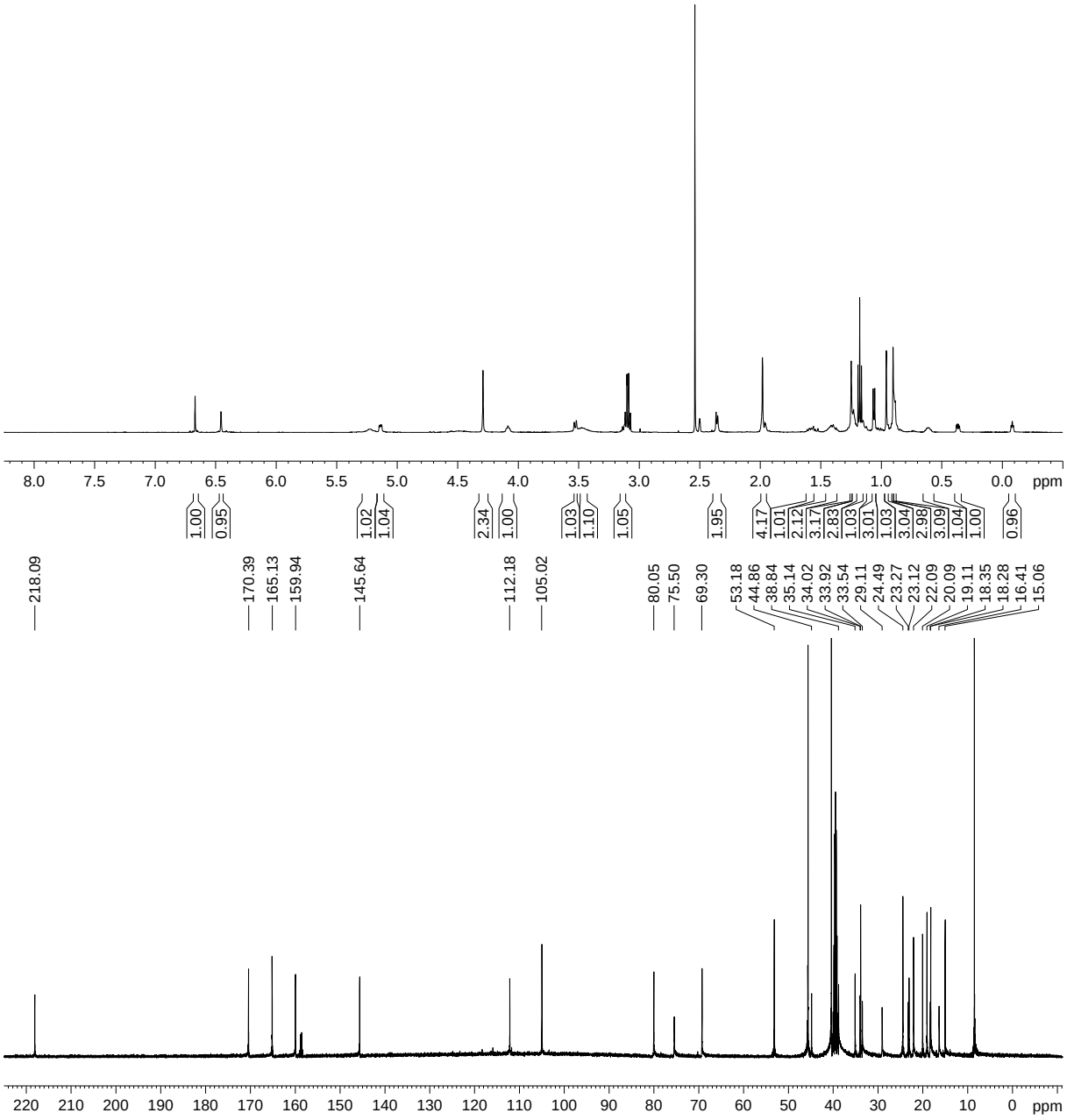
1H), 1.46-1.36 (m, 2H), 1.25 (s, 3H), 1.24-1.20 (m, 3H), 1.15-1.12 (m, 1H), 1.06 (d, J = 6.7 Hz, 3H), 1.04-0.98 (m, 1H), 0.96 (s, 3H), 0.90 (s, 3H), 0.89 (d, J = 6.8 Hz, 3H), 0.61 (dddd, J = 10.5, 8.6, 5.4, 3.2 Hz, 1H), 0.37 (dd, J = 8.6, 4.0 Hz, 1H), -0.08 (dd, J = 5.4, 4.0 Hz, 1H).

^{13}C-NMR (125 MHz, DMSO-d6): δ = 218.1, 170.4, 165.1, 159.9, 145.6, 112.2, 105.0, 80.0, 75.5, 69.3, 53.2, 44.9, 38.8, 35.1, 34.0, 33.9, 33.5, 29.1, 24.5, 23.3, 23.1, 22.1, 20.1, 19.1, 18.3, 18.3, 16.4, 15.1.

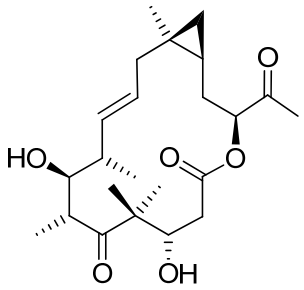
IR (film): ν 3374, 2931, 1730, 1686, 1599, 1455, 1379, 1334, 1260, 1148, 1085, 1025, 1009, 936, 871, 800, 757, 663.

HRMS (ESI): calculated for $C_{28}H_{45}N_2O_6$ $[M+H]^+$, found.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 10-60% (linear gradient from 0-12 min), retention time 10.26 min.



4.2.3 Analogs of 9,10-dehydro Epo B



147

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-acetyl-7,11-dihydroxy-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (147). To a solution of acetal **135** (49 mg, 0.072 mmol) in CH_2Cl_2 (4 mL) was added $FeCl_3 \cdot 6H_2O$ (97 mg, 0.36 mmol) in one portion at room temperature and the reaction mixture was stirred for 7 h. The reaction was then quenched by addition of water (4 mL), the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 4 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 25 mg (85%) of diol **147** as a white foam.

TLC: R_f 0.16 (hexane/EtOAc 3:1, CPS).

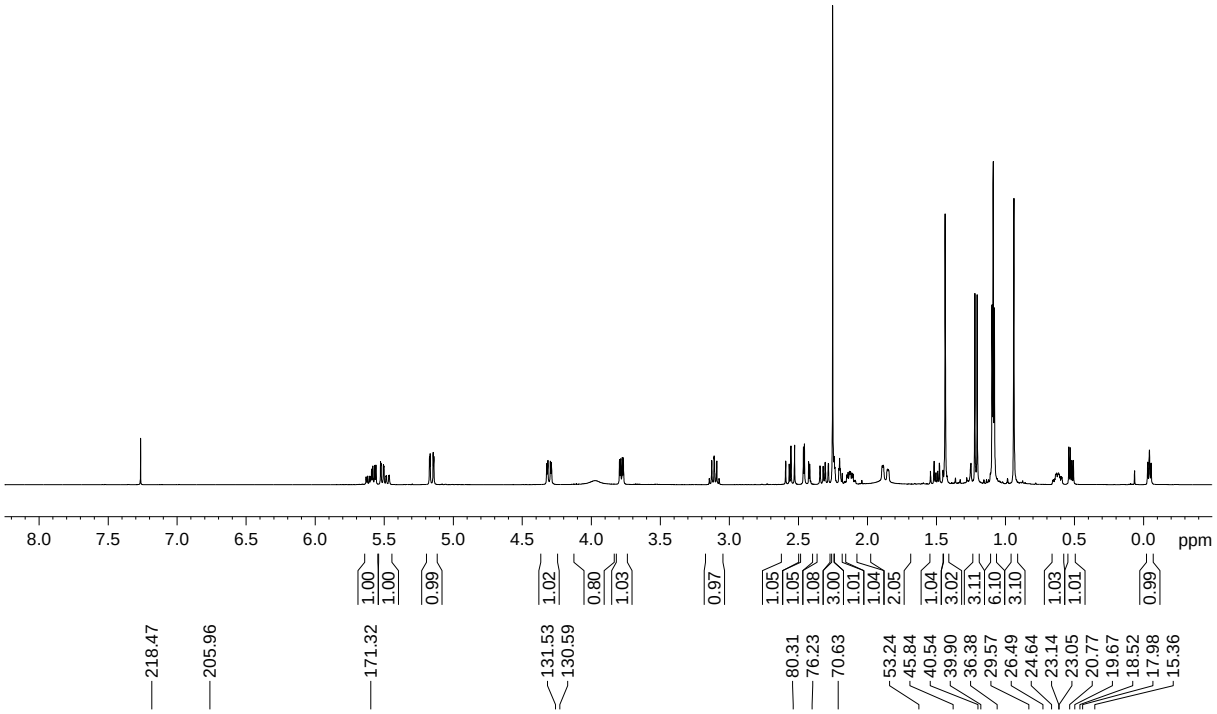
$[\alpha]^{20}_D$: = -16.7° (*c* = 1.12, $CHCl_3$).

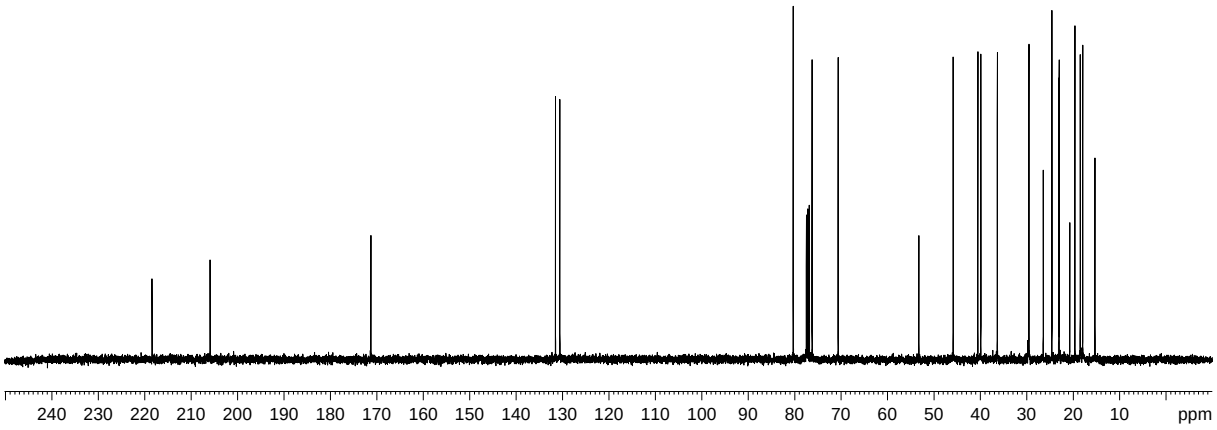
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.59 (ddd, *J* = 15.7, 9.1, 3.8 Hz, 1H), 5.49 (ddd, *J* = 15.7, 8.2, 1.3 Hz, 1H), 5.16 (dd, *J* = 10.6, 1.9 Hz, 1H), 4.31 (dd, *J* = 10.4, 3.0 Hz, 1H), 3.98 (br s, 1H), 3.78 (dd, *J* = 7.8, 3.3 Hz, 1H), 3.11 (dq, *J* = 7.8, 6.8 Hz, 1H), 2.56 (dd, *J* = 15.3, 10.4 Hz, 1H), 2.44 (dd, *J* = 15.3, 3.0 Hz, 1H), 2.31 (dd, *J* = 14.9, 9.1 Hz, 1H), 2.25 (s, 3H), 2.22 (ddd, *J* = 15.6, 2.2, 1.9 Hz, 1H), 2.13 (dqd, *J* = 8.2, 7.0, 3.3 Hz, 1H), 1.87 (ddd, *J* = 14.9, 3.8, 1.3 Hz, 1H), 1.87 (br s, 1H), 1.50 (ddd, *J* = 15.6, 10.6, 10.4 Hz, 1H), 1.44 (s, 3H), 1.21 (d, *J* = 6.8 Hz, 3H), 1.09 (s, 3H), 1.09 (d, *J* = 7.0 Hz, 3H), 0.94 (s, 3H), 0.62 (dddd, *J* = 10.4, 8.9, 5.4, 2.2 Hz, 1H), 0.52 (dd, *J* = 8.9, 4.4 Hz, 1H), -0.04 (dd, *J* = 5.4, 4.4 Hz, 1H).

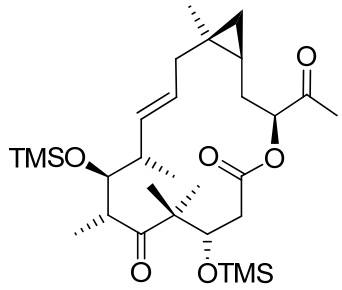
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.5, 206.0, 171.3, 131.5, 130.6, 80.3, 76.2, 70.6, 53.2, 45.8, 40.5, 39.9, 36.4, 29.6, 26.5, 24.6, 23.1, 23.1, 20.8, 19.7, 18.5, 18.0,15.4.

IR (film): ν 3478, 2980, 1743, 1720, 1689, 1452, 1367, 1252, 1182, 1149, 1077, 1011, 980, 942, 877, 754, 672.

HRMS (ESI): calculated for $C_{23}H_{36}O_6$ $[M+Na]^+$ 431.2404, found 431.2410.







148

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-acetyl-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (148). To a solution of diol **147** (240 mg, 0.59 mmol) in CH_2Cl_2 (15 mL) was added 2,6-lutidine (0.41 mL, 3.51

mmol) at 0 °C. The reaction mixture was cooled to -78 °C and TMSOTf (0.32 mL, 1.75 mmol) was added dropwise. The reaction mixture was stirred for 1.5 h at this temperature and was then allowed to reach room temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (20 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 20 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to yield 318 mg (98%) of silyl ether **148** as a colorless oil.

Note: Upon extensive scratching and storage at -20 °C silyl ether **148** turned into a white solid.

TLC: R_f 0.18 (hexane/EtOAc 12:1, CPS).

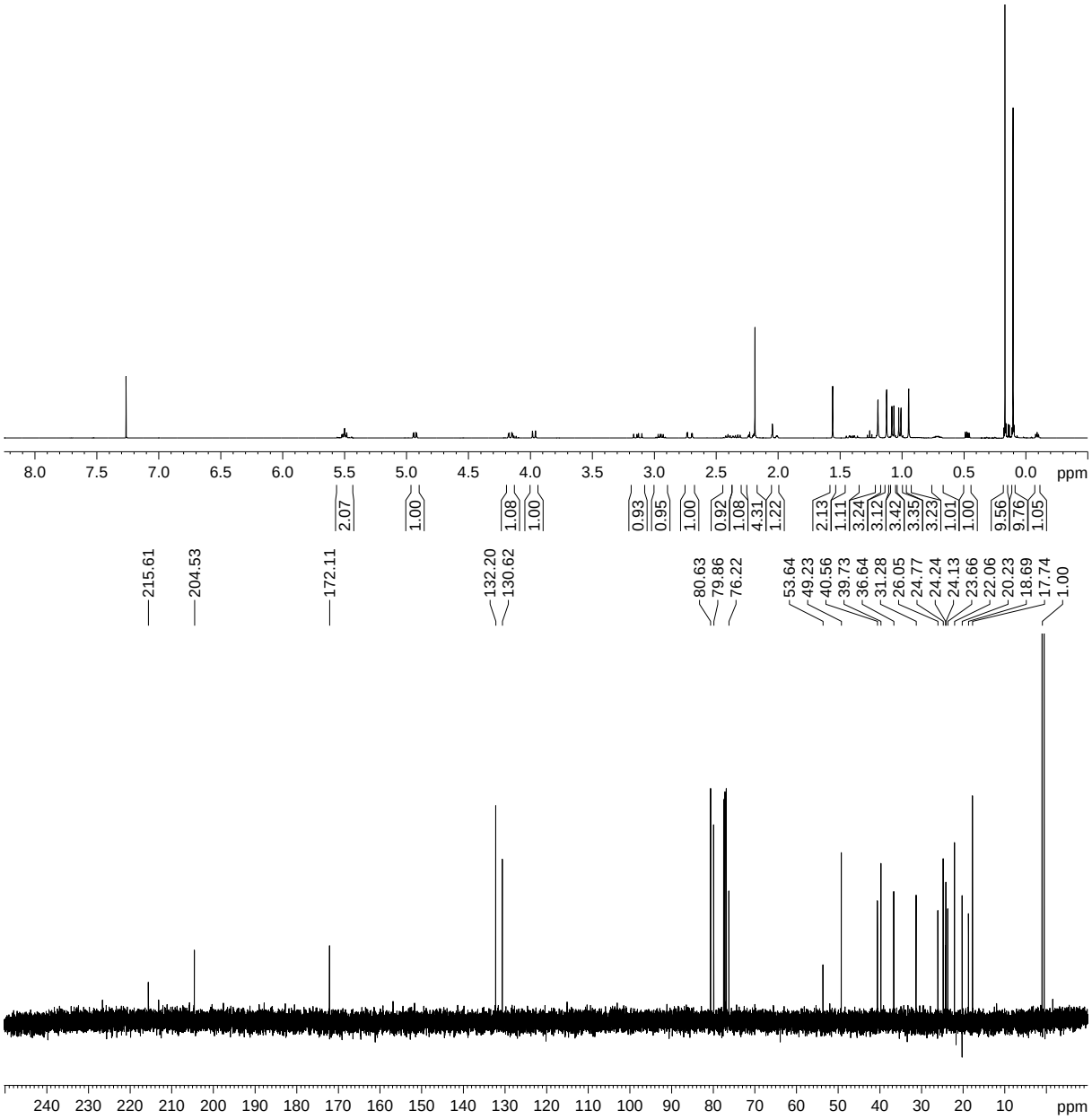
$[\alpha]^{20}_D$: = -10.3° (*c* = 0.35, $CHCl_3$).

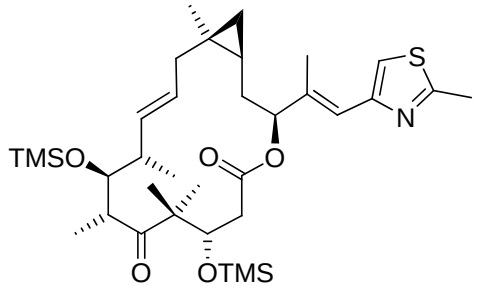
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.53 (ddd, *J* = 15.6, 7.8, 3.2 Hz, 1H), 5.47 (ddd, *J* = 15.6, 7.6, 1.0 Hz, 1H), 4.93 (dd, *J* = 9.2, 2.5 Hz, 1H), 4.16 (dd, *J* = 10.7, 2.1 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.13 (dd, *J* = 15.8, 10.7 Hz, 1H), 2.95 (dq, *J* = 9.7, 6.8 Hz, 1H), 2.71 (dd, *J* = 15.8, 2.1 Hz, 1H), 2.40 (dq, *J* = 7.6, 7.0 Hz, 1H), 2.34 (dd, *J* = 15.3, 7.8 Hz, 1H), 2.21 (dd, *J* = 15.5, 3.2 Hz, 1H), 2.19 (s, 3H), 2.03 (dd, *J* = 15.5, 2.5 Hz, 1H), 1.56 (s, 3H), 1.40 (ddd, *J* = 15.5, 11.4, 9.2 Hz, 1H), 1.19 (s, 3H), 1.12 (s, 3H), 1.07 (d, *J* = 6.8 Hz, 3H), 1.02 (d, *J* = 7.0 Hz, 3H), 0.94 (s, 3H), 0.71 (dddd, *J* = 11.4, 8.6, 5.4, 3.2 Hz, 1H), 0.47 (dd, *J* = 8.7, 4.3 Hz, 1H),0.17 (s, 9H), 0.10 (s, 9H), -0.09 (dd, *J* = 5.4, 4.3 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.6, 204.5, 172.1, 132.2, 130.6, 80.6, 79.9, 76.2, 53.6, 49.2, 40.6, 39.7, 36.6, 31.3, 26.1, 24.8, 24.2, 24.1, 23.7, 22.1, 20.2, 18.7, 17.7, 1.0, 0.5.

IR (film): ν 2957, 2904, 1734, 1695, 1454, 1385, 1365, 1311, 1251, 1200, 1161, 1112, 1086, 1036, 1021, 987, 889, 841, 753.

HRMS (ESI): calculated for $C_{29}H_{55}O_6Si_2$ $[M+H]^+$ 575.3195, found 575.3196.





149

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-8,8,10,12,16-pentamethyl-3-((*E*)-1-(2-methylthiazol-4-yl)prop-1-en-2-yl)-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (149). To a solution of phosphonium salt **78a** (41.1 mg, 0.118 mmol) in THF (1 mL)

was added KHMDS (23.5 mg, 0.118 mmol) in one portion at 0 °C. The reaction mixture was stirred for 0.5 h at this temperature and was then cooled to -78 °C. Methyl ketone **148** (12.0 mg, 0.022 mmol) in THF (0.5 mL) was added dropwise and the solution was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to afford 12.0 mg (79%) of an inseparable 7:1 mixture of **149** and its C16-C17 *Z* isomer as a semi solid.

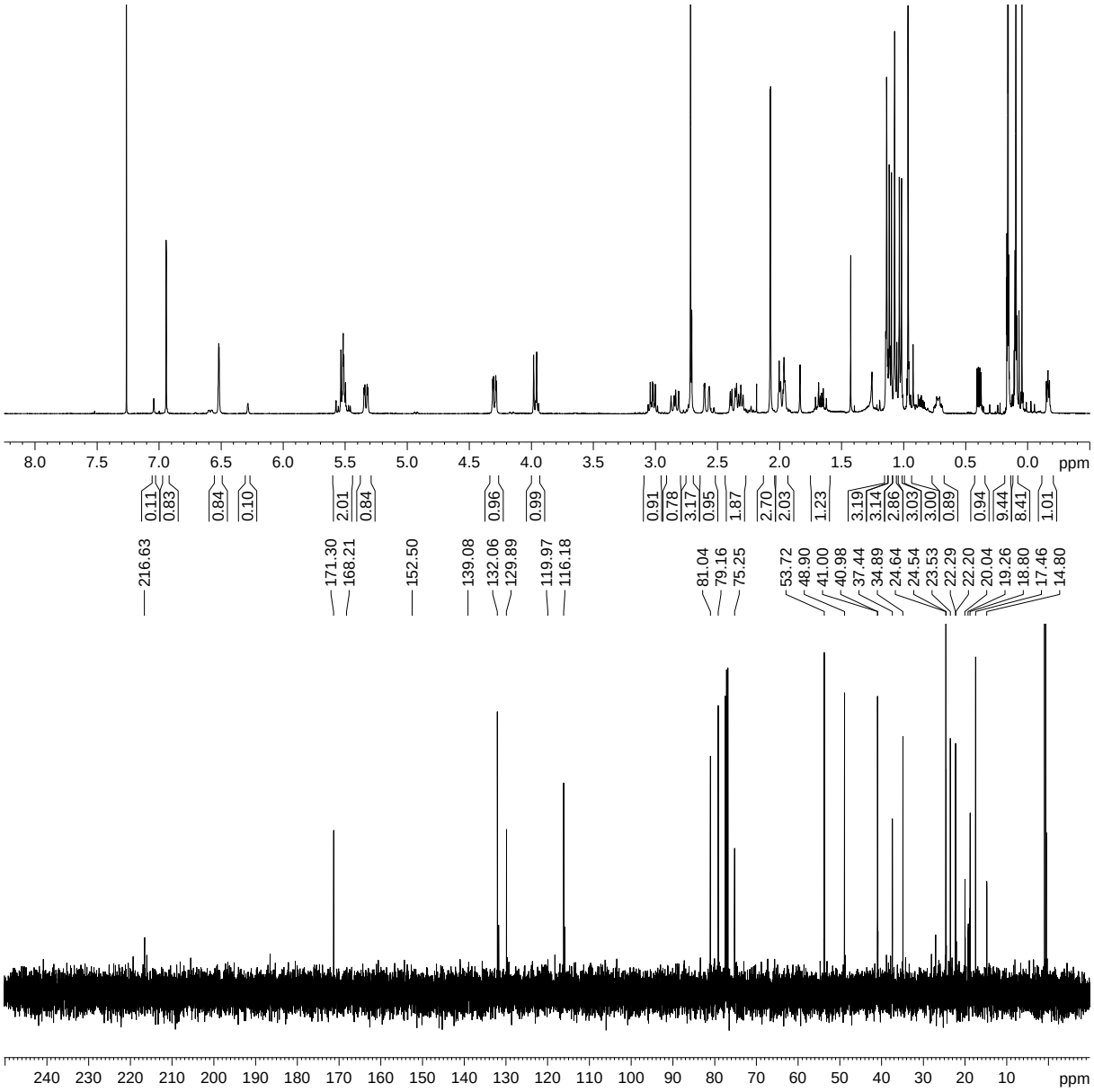
TLC: R_f 0.54 (hexane/EtOAc 9:1, UV, CPS).

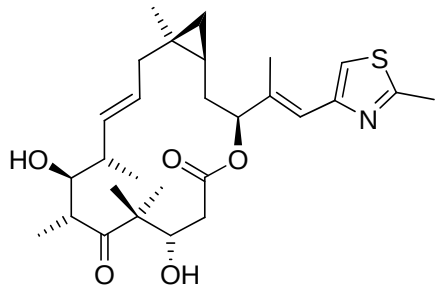
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.94 (s, 1H), 6.52 (q, *J* = 1.2 Hz, 1H), 5.55 (dd, *J* = 15.6, 7.4 Hz, 1H), 5.49 (ddd, *J* = 15.6, 5.6, 5.5 Hz, 1H), 5.33 (dd, *J* = 9.7, 2.6 Hz, 1H), 4.30 (dd, *J* = 9.7, 2.3 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.02 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.85 (dd, *J* = 14.8, 9.7 Hz, 1H), 2.72 (s, 3H), 2.59 (dd, *J* = 14.8, 2.3 Hz, 1H), 2.37 (dd, *J* = 14.8, 5.5 Hz, 1H), 2.31 (dq, *J* = 7.4, 7.0 Hz, 1H), 2.07 (d, *J* = 1.2 Hz, 3H), 1.98 (ddd, *J* = 15.1, 2.6, 2.6 Hz, 1H), 1.98 (dd, 14.8, 5.6 Hz, 1H), 1.67 (ddd, *J* = 15.1, 10.1, 9.8 Hz, 1H), 1.14 (s, 3H), 1.11 (d, *J* = 7.0 Hz, 3H), 1.07 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.72 (dddd, *J* = 10.1, 8.8, 5.6, 2.5 Hz, 1H), 0.39 (dd, *J* = 8.8, 4.1 Hz, 1H), 0.16 (s, 9H), 0.10 (s, 9H), -0.19 (dd, *J* = 5.7, 4.1 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.6, 171.3, 168.2, 152.5, 139.1, 132.1, 129.9, 120.0, 116.2, 81.0, 79.2, 75.2, 53.7, 48.9, 41.0, 41.0, 37.4, 34.9, 24.6, 24.5, 23.5, 22.3, 22.2, 20.0, 19.3, 18.8, 17.5, 14.8 ,1.0, 0.7.

IR (film): ν 2957, 1738, 1694, 1451, 1381, 1304, 1251, 1202, 1181, 1160, 1115, 1091, 1038, 1020, 987, 950, 889, 840, 753, 734.

HRMS (ESI): calculated for $C_{34}H_{58}NO_5SSi_2$ $[M+H]^+$ 648.3569, found 348.3562.





1f

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-3-((*E*)-1-(2-methylthiazol-4-yl)prop-1-en-2-yl)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione(1f) To a 7:1 mixture of protected cyclopropyl-Epo B **149** and its C16-C17 *Z* isomer (12.0 mg, 0.019 mmol) in MeOH (0.5 mL) was added citric acid (11.7 mg, 0.056 mmol) at room

temperature. The reaction mixture was stirred for 4 h, a second portion of citric acid (3.0 mg, 0.014 mmol) was then added and stirring was continued for further 4 h. The reaction mixture was then diluted with water (1 mL) and treated with saturated aqueous $NaHCO_3$ (1 mL) followed by saturated aqueous NH_4Cl (1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 5:2) to yield 8.6 mg (92%) of an inseparable 7:1 mixture of **1f** and its C16-C17 *Z* isomer as a colorless oil.

Note: The C16-C17 *Z* isomer resulting from the Wittig reaction could be separated by preparative HPLC.

TLC: R_f 0.33 (hexane/EtOAc 2:1, UV, CPS).

$[\alpha]^{20}_D$: = -92.3° (*c* = 0.4, $CHCl_3$).

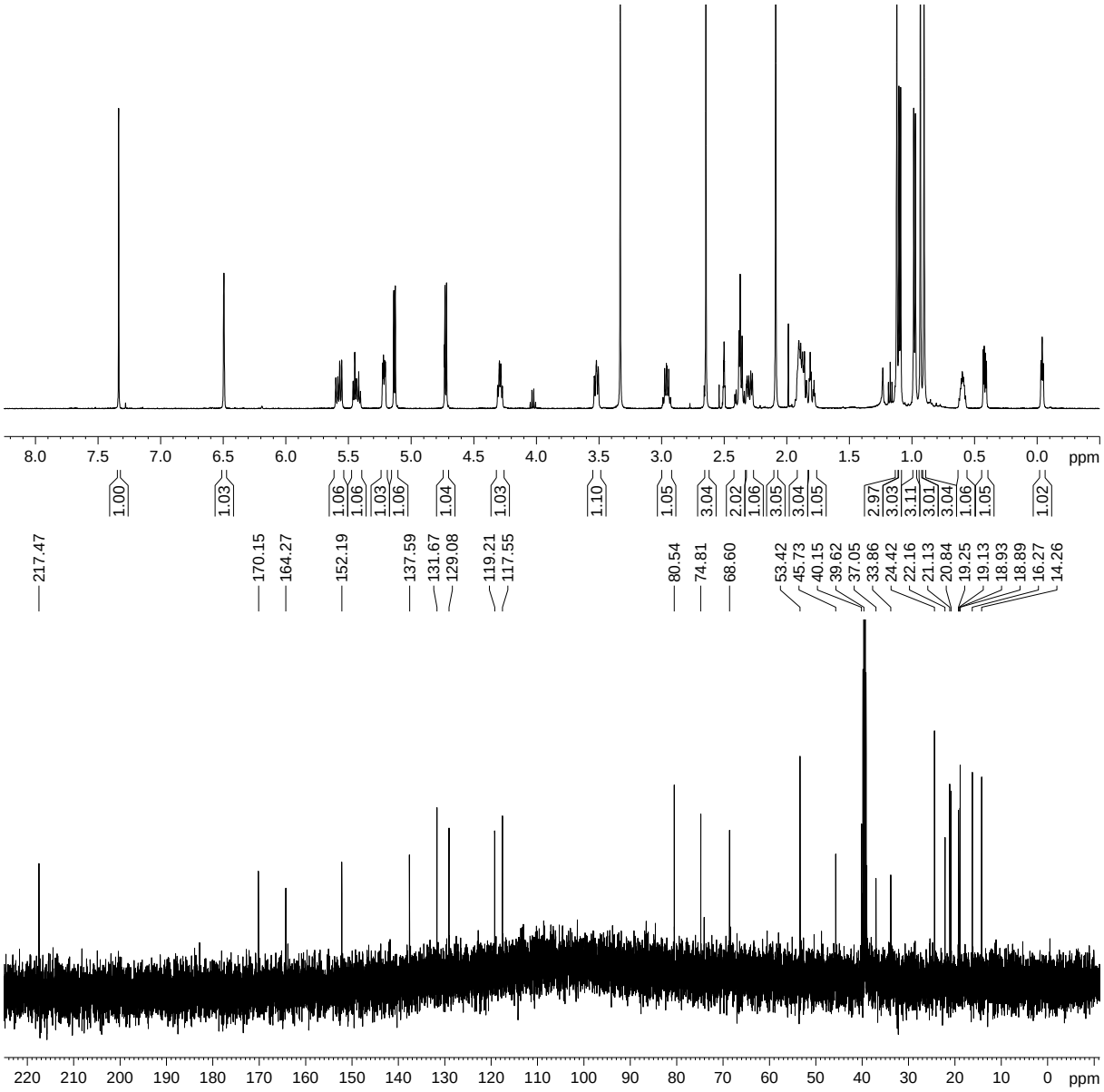
^{1}H-NMR (500 MHz, DMSO-d6): δ = 7.33 (s, 1H), 6.49 (q, *J* = 1.2 Hz, 1H), 5.58 (dd, *J* = 15.7, 8.2 Hz, 1H), 5.43 (ddd, *J* = 15.7, 7.0, 6.6 Hz, 1H), 5.21 (dd, *J* = 7.9, 3.3 Hz, 1H), 5.13 (d, *J* = 6.4 Hz, 1H), 4.72 (d, *J* = 6.4 Hz, 1H), 4.29 (ddd, *J* = 7.5, 6.4, 4.9 Hz, 1H), 3.52 (ddd, *J* = 9.3, 6.4, 1.5 Hz, 1H), 2.96 (dq, *J* = 9.3, 6.8 Hz, 1H), 2.64 (s, 3H), 2.39 (dd, *J* = 16.2, 4.9 Hz, 1H), 2.35 (dd, *J* = 16.2, 7.5 Hz, 1H), 2.29 (dd, *J* = 14.7, 6.6 Hz, 1H), 2.09 (d, *J* = 1.2 Hz, 3H), 1.90 (dqd, *J* = 8.3, 6.9, 1.5 Hz, 1H), 1.88 (dd, *J* = 14.7, 7.0 Hz, 1H), 1.86 (ddd, *J* = 15.3, 8.3, 7.9 Hz, 1H), 1.79 (ddd, *J* = 15.3, 3.6, 3.4 Hz, 1H), 1.12 (s, 3H), 1.10 (d, *J* = 6.8 Hz, 3H), 0.98 (d, *J* = 6.9 Hz, 3H), 0.93 (s, 3H), 0.90 (s, 3H), 0.60 (dddd, *J* = 8.7, 8.3, 5.5, 3.6 Hz, 1H), 0.42 (dd, *J* = 8.7, 3.9 Hz, 1H), -0.04 (dd, *J* = 5.5, 3.9 Hz, 1H).

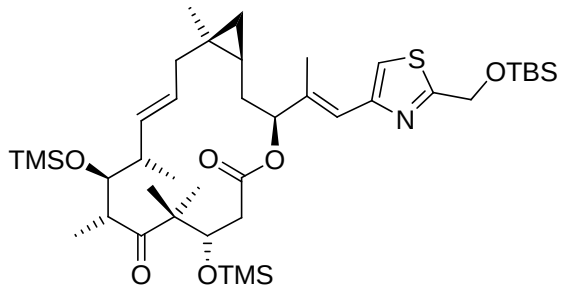
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.5, 170.1, 164.3, 152.2, 137.6, 131.7, 129.1, 119.2, 117.5, 80.5, 74.8, 68.6, 53.4, 45.7, 40.1, 39.6, 37.1, 33.9, 24.4, 22.2, 21.1, 20.8, 19.3, 19.1, 18.9, 18.9, 16.3, 14.3.

IR (film): ν 3417, 2953, 2927, 2859, 1729, 1689, 1606, 1508, 1452, 1421, 1365, 1255, 1186, 1153, 1090, 1009, 980, 958, 840, 774, 671.

HRMS (ESI): calculated for $C_{28}H_{42}NO_5S$ $[M+H]^+$ 504.2778, found 504.2768.

Analytical HPLC: Method: eluent B 50-60% (linear gradient from 0-12 min), retention time 9.41 min.





151a

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-((*E*)-1-(2-((*tert*-butyldimethylsilyloxy)methyl)thiazol-4-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (151a). To a solution of phosphonium salt **140a** (34.7 mg, 0.072 mmol) in THF (1 mL) was added *n*-BuLi (1.6 M in hexane, 42 µL,

0.067 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to 0 °C over a period of 3.5 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 30:1) to afford 9.3 mg (67%) of **151a** as a single isomer as a colorless oil

TLC: R_f 0.30 (hexane/EtOAc 20:1, UV, CPS).

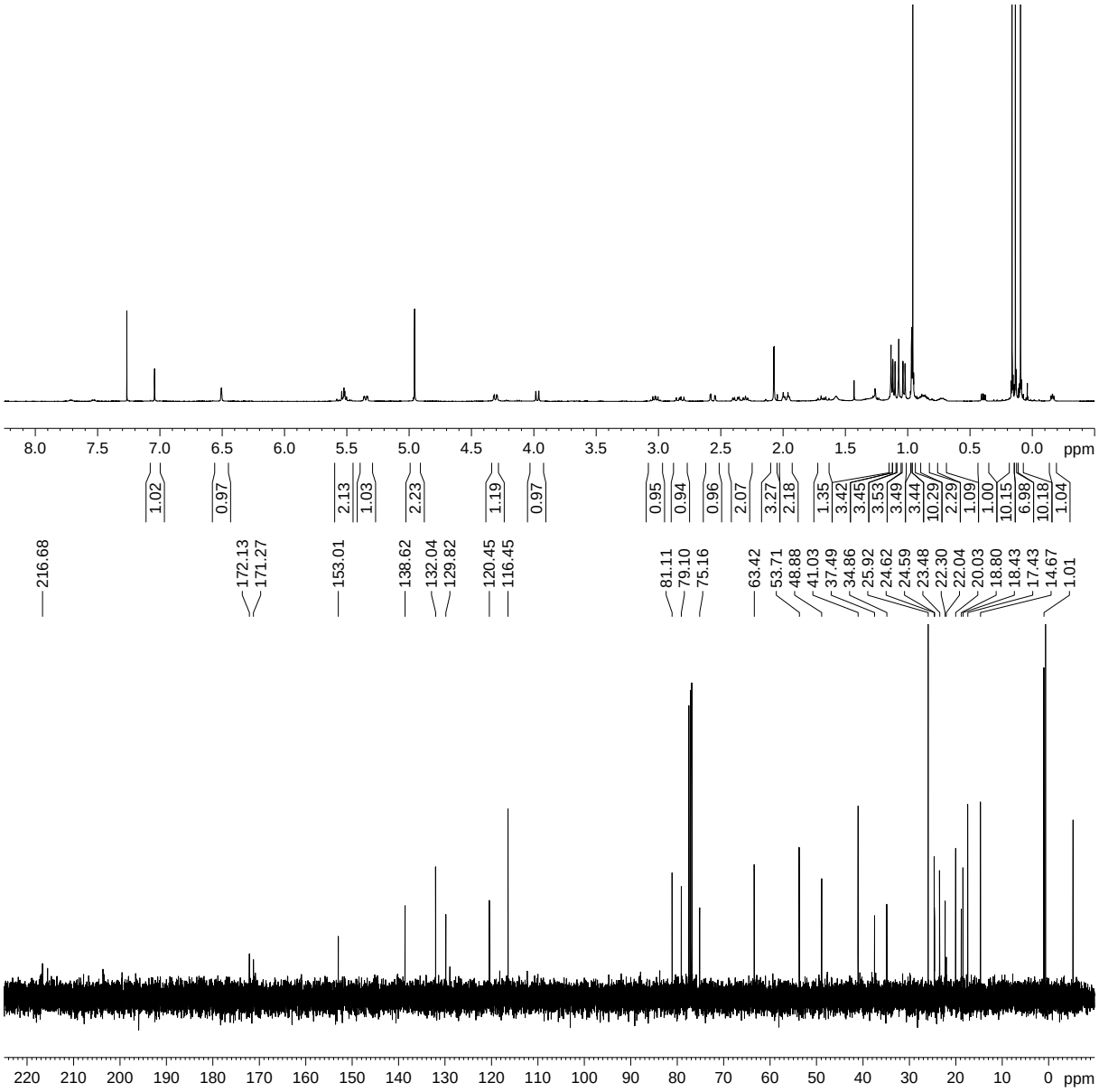
$[\alpha]^{20}_D$: = -11.3° (*c* = 0.47, $CHCl_3$).

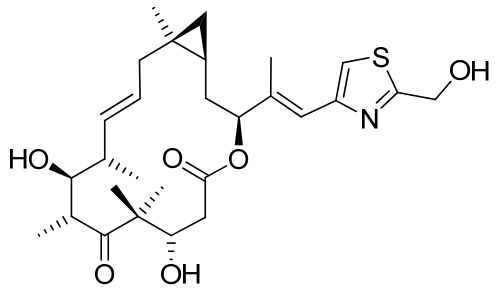
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.04 (s, 1H), 6.50 (q, *J* = 1.2 Hz, 1H), 5.55 (dd, *J* = 15.6, 7.4 Hz, 1H), 5.49 (dd, *J* = 15.6, 5.6 Hz, 1H), 5.35 (dd, *J* = 9.8, 2.5 Hz, 1H), 4.95 (s, 2H), 4.31 (dd, *J* = 9.6, 2.1 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.02 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.83 (dd, *J* = 14.8, 9.6 Hz, 1H), 2.56 (dd, *J* = 14.8, 2.1 Hz, 1H), 2.38 (dd, *J* = 15.6, 5.6 Hz, 1H), 2.30 (dq, *J* = 7.4, 7.0 Hz, 1H), 2.07 (d, *J* = 1.2 Hz, 3H), 1.98 (d, *J* = 15.6 Hz, 1H), 1.97 (ddd, *J* = 15.1, 5.1, 2.5, 1H), 1.68 (ddd, *J* = 15.1, 10.2, 9.8 Hz, 1H), 1.13 (s, 3H), 1.11 (d, *J* = 7.0 Hz, 3H), 1.07 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.96 (s, 9H), 0.72 (dddd, *J* = 10.2, 8.9, 5.6, 2.5 Hz, 1H), 0.39 (dd, *J* = 8.9, 4.1 Hz, 1H), 0.16 (s, 9H), 0.14 (s, 6H), 0.09 (s, 9H), -0.19 (dd, *J* = 5.6, 4.1 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.7, 172.1, 171.3, 153.0, 138.6, 132.0, 129.8, 120.4, 116.4, 81.1, 79.1, 75.2, 63.4, 53.7, 48.9, 41.0, 41.0, 37.5, 34.9, 25.9, 24.6, 24.6, 23.5, 22.3, 22.0, 20.0, 18.8, 18.4, 17.4, 14.7, 1.0, 0.7, -5.3.

IR (film): ν 2955, 2931, 2861, 1738, 1694, 1464, 1382, 1362, 1253, 1039, 987, 890, 840, 780, 754.

HRMS (ESI): calculated for $C_{40}H_{72}NO_6SSi_3$ $[M+H]^+$ 778.4383, found 778.4391.





2a

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-3-((*E*)-1-(2-(hydroxymethyl)thiazol-4-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione(2a) To protected cyclopropyl-Epo B **151a** (9.3 mg, 0.012 mmol) in CH_2Cl_2 (0.5 mL) and MeOH (0.25 mL) was added CSA (3.6 mg, 0.015 mmol) at 0 °C. The reaction mixture

was then allowed to reach room temperature and stirred for 24 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 6.3 mg (99%) of **2a** as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -50.5° (*c* = 0.33, acetone).

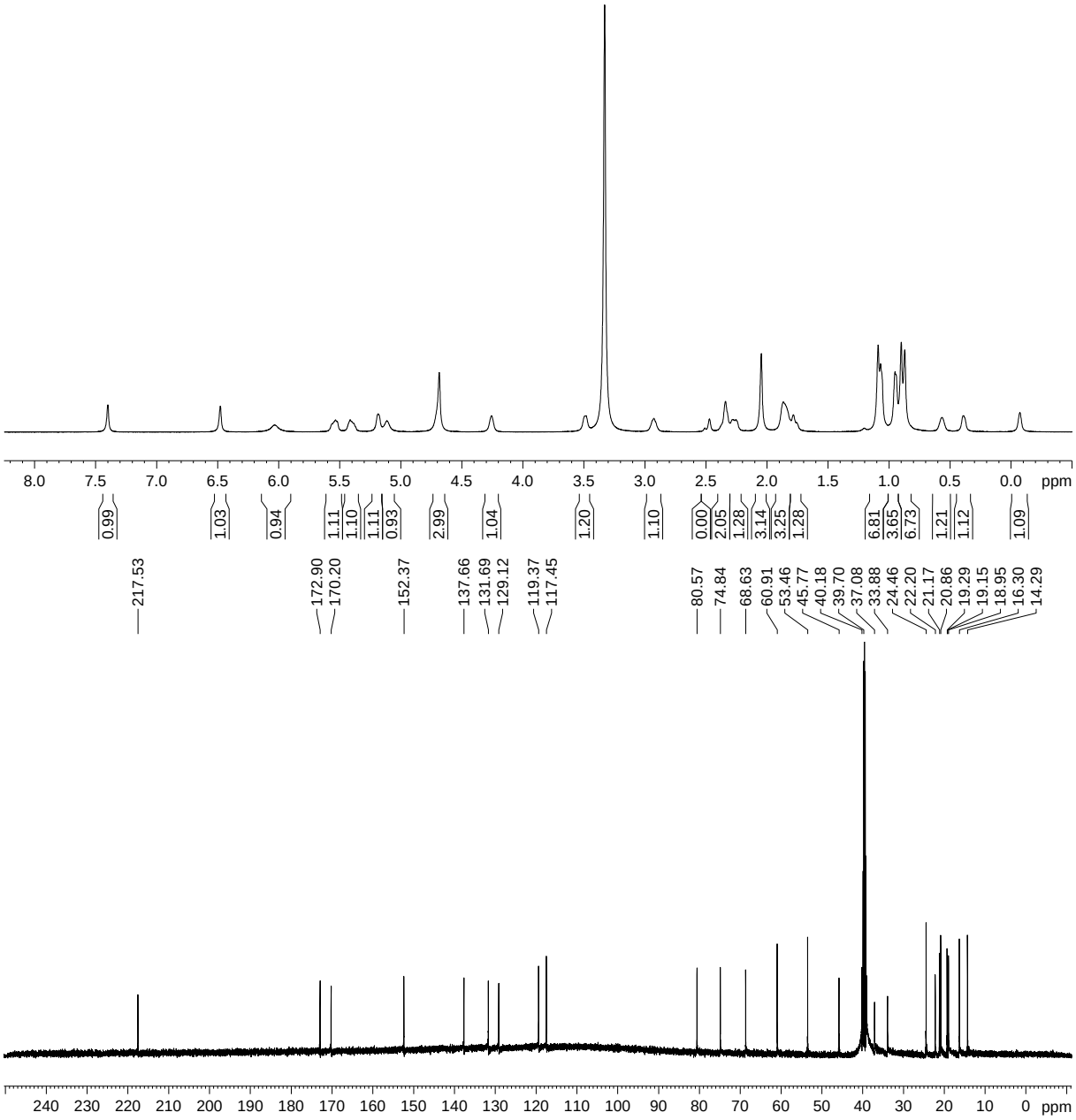
^{1}H-NMR (500 MHz, DMSO-d6): δ = 7.43 (s, 1H), 6.51 (q, *J* = 1.2 Hz, 1H), 6.05 (t, *J* = 5.5 Hz, 1H), 5.57 (dd, *J* = 15.7, 8.3 Hz, 1H), 5.43 (ddd, *J* = 15.7, 6.9, 6.5 Hz, 1H), 5.21 (dd, *J* = 7.8, 3.4 Hz, 1H), 5.14 (d, *J* = 6.4 Hz, 1H), 4.73 (d, *J* = 6.5 Hz, 1H), 4.71 (d, *J* = 5.5 Hz, 2H), 4.29 (ddd, *J* = 7.6, 6.5, 5.0 Hz, 1H), 3.52 (ddd, *J* = 9.3, 6.4, 1.5 Hz, 1H), 2.96 (dq, *J* = 9.3, 6.8 Hz, 1H), 2.38 (d, *J* = 16.3, 5.0 Hz, 1H), 2.34 (dd, *J* = 16.3, 7.6 Hz, 1H), 2.29 (dd, *J* = 14.7, 6.5 Hz, 1H), 2.08 (d, *J* = 1.2 Hz, 3H), 1.90 (dqd, *J* = 8.3, 6.9, 1.5 Hz, 1H), 1.89 (dd, *J* = 14.7, 6.9 Hz, 1H), 1.87 (ddd, *J* = 15.3, 8.5, 7.9 Hz, 1H), 1.80 (ddd, *J* = 15.3, 3.6, 3.4 Hz, 1H), 1.12 (s, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.98 (d, *J* = 6.9 Hz, 3H), 0.93 (s, 3H), 0.90 (s, 3H), 0.60 (dddd, *J* = 8.7, 8.5, 5.4, 3.6 Hz, 1H), 0.42 (dd, *J* = 8.7, 4.0 Hz, 1H), -0.04 (dd, *J* = 5.4, 4.0 Hz, 1H).

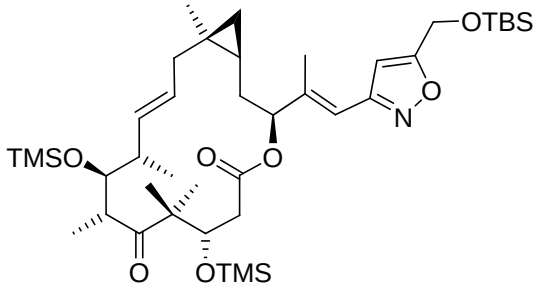
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.5, 172.9, 170.2, 152.4, 137.7, 131.7, 129.1, 119.4, 117.4, 80.6, 74.8, 68.6, 60.9, 53.5, 45.8, 40.2, 39.7, 37.1, 33.9, 24.5, 22.2, 21.2, 20.9, 19.3, 19.2, 18.9, 16.3, 14.3.

IR (film): ν 3397, 2925, 2869, 2856, 1729, 1688, 1455, 1256, 1153, 1052, 1040, 1010, 980, 941.

HRMS (ESI): calculated for $C_{28}H_{42}NO_6S$ $[M+H]^+$ 520.2727, found 520.2726.

Analytical HPLC: Method: eluent B 40-50% (linear gradient from 0-12 min), retention time 9.40 min.





151b

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-((*E*)-1-(5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-

oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (151b). To a solution of phosphonate **140b** (39.4 mg, 0.109 mmol) in THF (1.0 mL) was added LiHMDS (1.0 M in hexane, 99 μL, 0.099 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 6.2 mg (45%) of **151b** as a single isomer as a colorless oil

TLC: R_f 0.16 (hexane/EtOAc 20:1, UV, CPS).

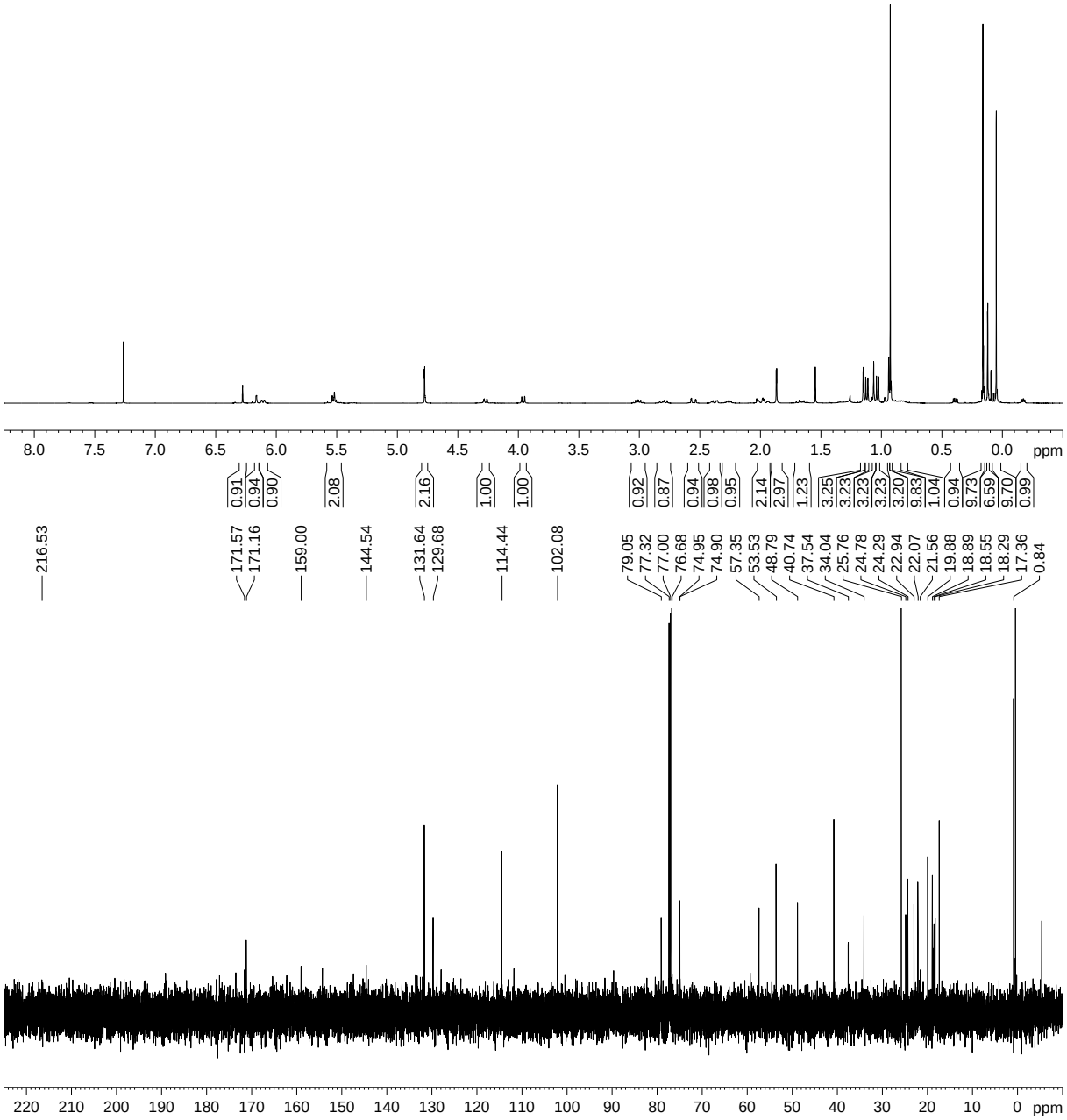
$[\alpha]^{20}_D$: = +33.2° (*c* = 0.31, $CHCl_3$).

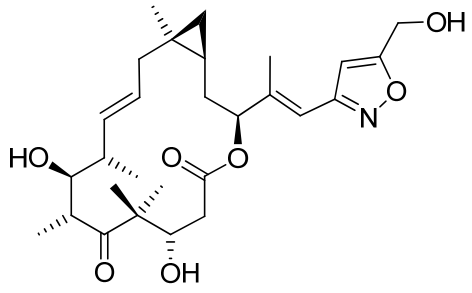
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.28 (t, *J* = 0.7 Hz, 1H), 6.16 (q, *J* = 1.5 Hz, 1H), 6.11 (dd, *J* = 9.6, 2.8 Hz, 1H), 5.55 (dd, *J* = 15.7, 7.1 Hz, 1H), 5.50 (ddd, *J* = 15.7, 5.0, 4.9 Hz, 1H), 4.77 (d, *J* = 0.7 Hz, 2H), 4.27 (dd, *J* = 9.8, 1.7 Hz, 1H), 3.96 (d, *J* = 9.8 Hz, 1H), 3.01 (dq, *J* = 9.8, 7.0 Hz, 1H), 2.80 (dd, *J* = 14.7, 9.8 Hz, 1H), 2.55 (dd, *J* = 14.7, 1.7 Hz, 1H), 2.38 (dd, *J* = 15.3, 4.9 Hz, 1H), 2.26 (dq, *J* = 7.1, 7.0 Hz, 1H), 1.99 (ddd, *J* = 15.0, 2.8, 2.4 Hz, 1H), 1.96 (dd, *J* = 15.3, 5.0, 1H), 1.87 (d, *J* = 1.5 Hz, 3H), 1.66 (ddd, *J* = 15.0, 10.7, 9.6 Hz, 1H), 1.15 (s, 3H), 1.12 (d, *J* = 7.0 Hz, 3H), 1.06 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.94 (s, 3H), 0.93 (s, 9H), 0.82 (dddd, *J* = 10.7, 8.9, 5.6, 2.4 Hz, 1H), 0.39 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.12 (s, 6H), 0.05 (s, 9H), -0.17 (dd, *J* = 5.6, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.5, 171.6, 171.2, 159.0, 144.5, 131.6, 129.7, 114.4, 102.1, 79.1, 74.9, 74.9, 57.4, 53.5, 48.8, 40.7, 37.5, 34.0, 25.8, 24.8, 24.3, 22.9, 22.1, 21.6, 19.9, 18.9, 18.6, 18.3, 17.4, 0.8, 0.4, -5.3.

IR (film): ν 2956, 2930, 2860, 1739, 1694, 1460, 1382, 1302, 1283, 1252, 1202, 1180, 1160, 1113, 1092, 1038, 989, 948, 890, 839, 780, 754.

HRMS (ESI): calculated for $C_{40}H_{72}NO_7Si_3$ $[M+H]^+$ 762.4611, found 762.4608.





2b

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-3-((*E*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-

dione(2b) To protected cyclopropyl-Epo B **151b** (6.2 mg, 0.008 mmol) in CH_2Cl_2 (0.3 mL) and MeOH (0.2 mL) was added CSA (1.9 mg, 0.008 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 15 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 2.1 mg (56%) of **2b** as a colorless oil.

TLC: R_f 0.20 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -7.6° (*c* = 0.38, acetone).

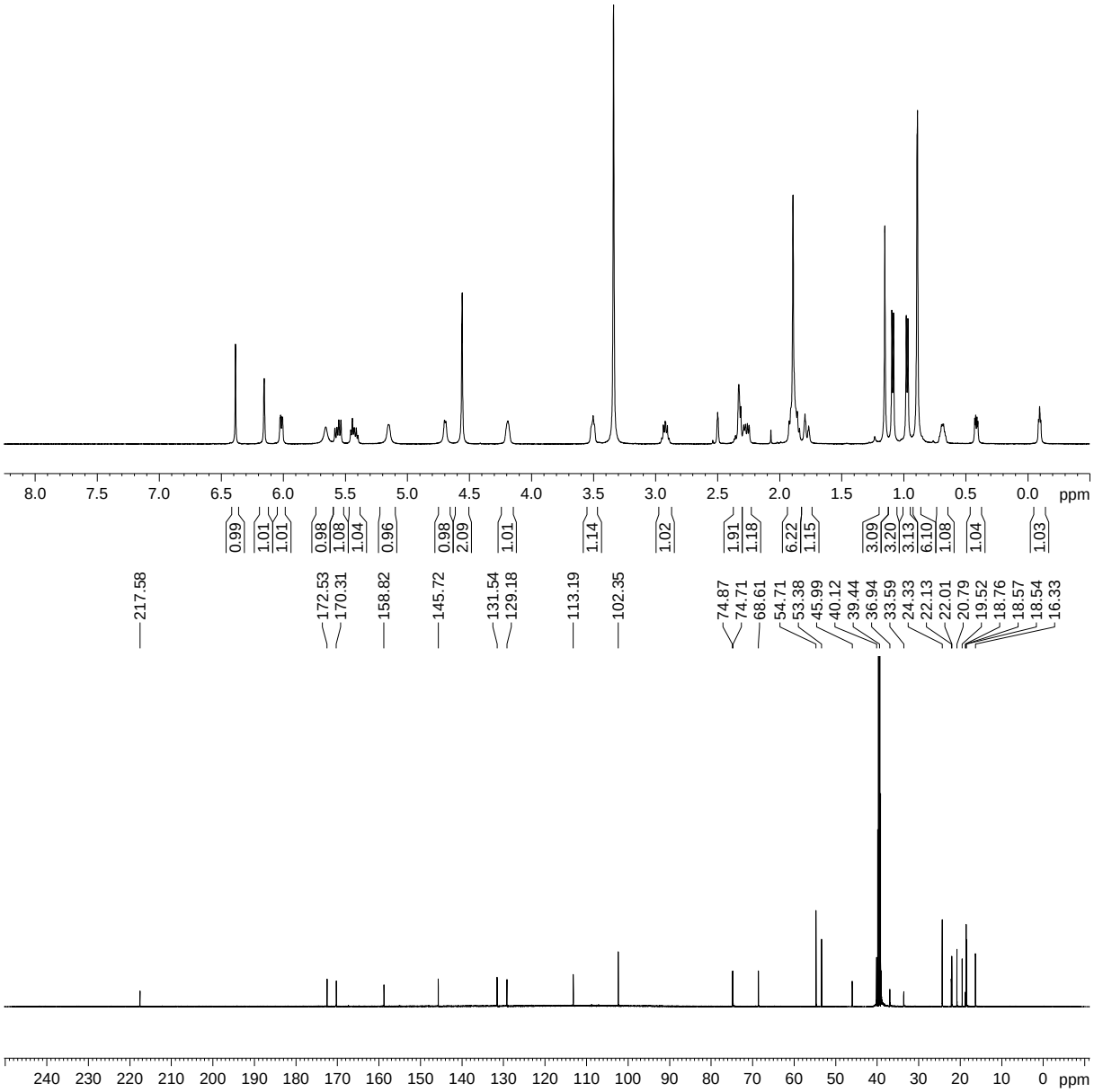
^{1}H-NMR (500 MHz, DMSO-d6): δ = 6.39 (s, 1H), 6.15 (s, 1H), 6.01 (dd, *J* = 8.2, 2.6 Hz, 1H), 5.67 (br s, 1H), 5.56 (dd, *J* = 15.6, 8.2 Hz, 1H), 5.43 (ddd, *J* = 15.6, 7.0, 6.5 Hz, 1H), 5.15 (br s, 1H), 4.69 (d, *J* = 5.4 Hz, 1H), 4.56 (s, 2H), 4.19 (br s, 1H), 3.51 (dd, *J* = 9.0, 5.4 Hz, 1H), 2.92 (dq, *J* = 9.0, 6.8 Hz, 1H), 2.37-2.31 (m, 2H), 2.27 (d, *J* = 14.8, 7.0 Hz, 1H), 1.93-1.88 (m, 2H), 1.89 (s, 3H), 1.86 (dd, *J* = 15.1, 8.8 Hz, 1H), 1.78 (ddd, *J* = 15.1, 2.6, 2.6 Hz, 1H), 1.15 (s, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.8 Hz, 3H), 0.89 (s, 3H), 0.89 (s, 3H), 0.69 (dddd, *J* = 8.8, 8.7, 5.4, 2.6 Hz, 1H), 0.41 (dd, *J* = 8.7, 3.8 Hz, 1H), 0.10 (dd, *J* = 5.8, 3.8 Hz, 1H).

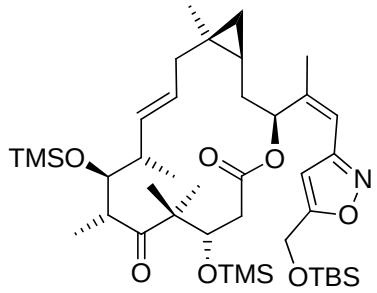
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.6, 172.5, 170.3, 158.8, 145.7, 131.5, 129.2, 113.2, 102.4, 74.9, 74.7, 68.6, 54.7, 53.4, 46.0, 40.1, 39.4, 36.9, 33.6, 24.3, 22.1, 22.0, 20.8, 19.5, 18.8, 18.6, 18.5, 16.3.

IR (film): ν 3391, 2958, 2925, 1730, 1716, 1688, 1456, 1261, 1080, 1038, 1012, 986,978, 938, 808.

HRMS (ESI): calculated for $C_{28}H_{42}NO_7$ $[M+H]^+$ 504.2956, found 504.2957.

Analytical HPLC: Method: eluent B 40-50% (linear gradient from 0-12 min), retention time 7.25 min.





151Zb

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-((*Z*)-1-(5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (151Zb). To a solution of phosphonate **140b**

(39.4 mg, 0.109 mmol) in THF (1.0 mL) was added LiHMDS (1.0 M in THF, 99 µL, 0.099 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 6.8 mg (49%) of **151Zb** as a single isomer as a colorless oil

TLC: R_f 0.21 (hexane/EtOAc 20:1, UV, CPS).

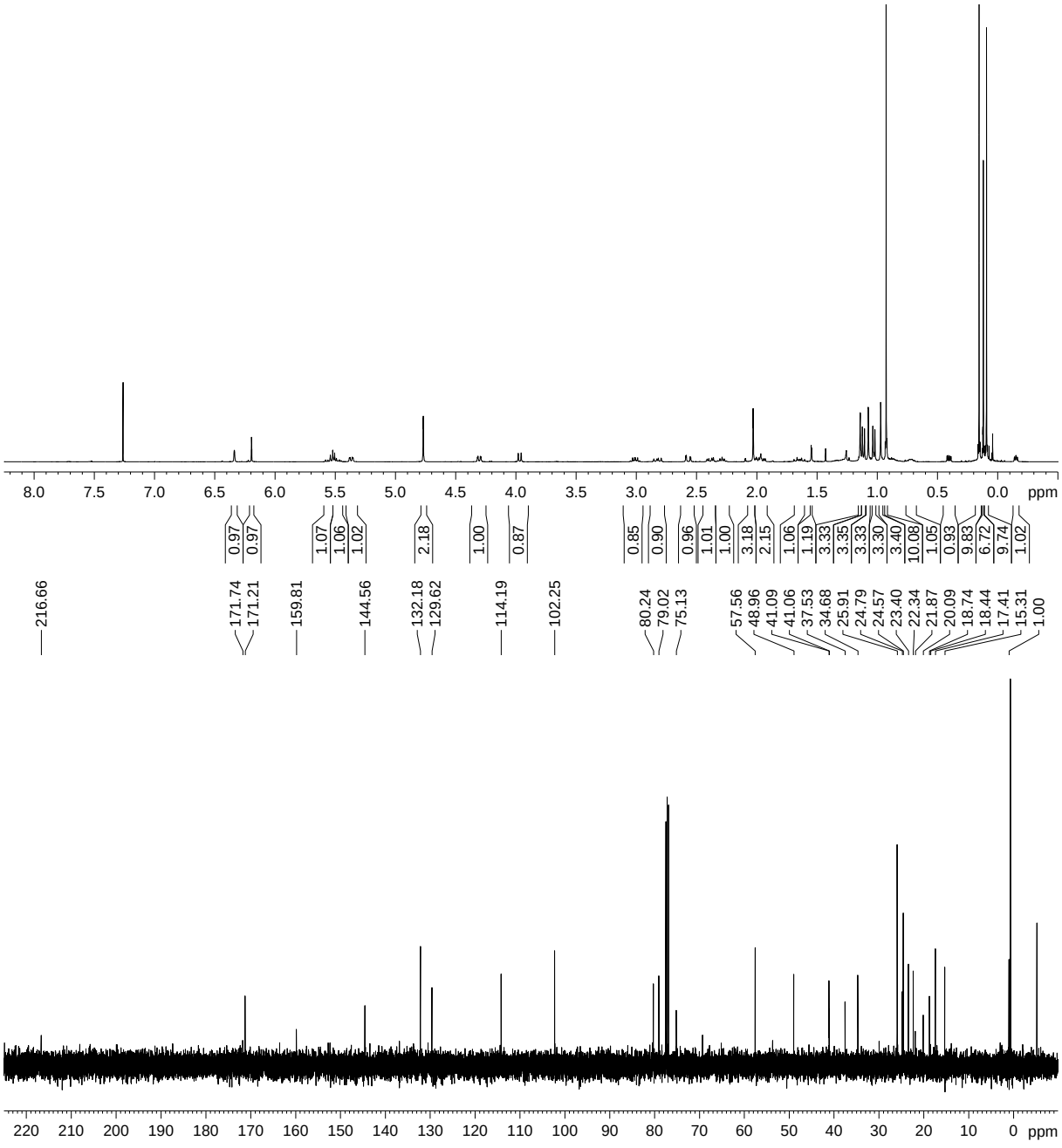
$[\alpha]^{20}_D$: = -1.9° (*c* = 0.34, $CHCl_3$).

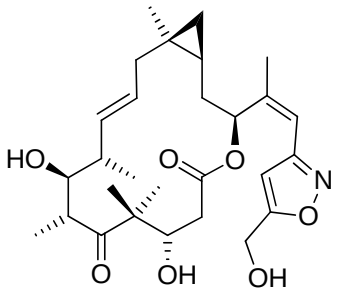
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.34 (q, *J* = 1.2 Hz, 1H), 6.20 (t, *J* = 0.7 Hz, 1H), 5.55 (dd, *J* = 15.7, 7.8 Hz, 1H), 5.48 (ddd, *J* = 15.7, 5.9, 5.4 Hz, 1H), 5.37 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.77 (d, *J* = 0.7 Hz, 2H), 4.31 (dd, *J* = 9.7, 2.0 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.01 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.83 (dd, *J* = 14.7, 9.7 Hz, 1H), 2.57 (dd, *J* = 14.7, 2.0 Hz, 1H), 2.39 (dd, *J* = 15.7, 5.9 Hz, 1H), 2.29 (dq, *J* = 7.8, 7.0 Hz, 1H), 2.03 (d, *J* = 1.2 Hz, 3H), 1.99 (ddd, *J* = 15.1, 2.6, 2.4 Hz, 1H), 1.96 (dd, *J* = 15.7, 5.4, 1H), 1.65 (ddd, *J* = 15.1, 10.2, 9.7 Hz, 1H), 1.14 (s, 3H), 1.12 (d, *J* = 7.0 Hz, 3H), 1.07 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.93 (s, 9H), 0.72 (dddd, *J* = 10.2, 8.7, 5.6, 2.6 Hz, 1H), 0.41 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.12 (s, 6H), 0.09 (s, 9H), -0.19 (dd, *J* = 5.6, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.7, 171.7, 171.2, 159.8, 144.6, 132.2, 129.6, 114.2, 102.3, 80.2, 79.0, 75.1, 57.6, 53.7, 49.0, 41.1, 41.1, 37.5, 34.7, 25.9, 24.8, 24.6, 23.4, 22.3, 21.9, 20.1, 18.7, 18.4, 17.4, 15.3, 1.0, 0.7, -5.2.

IR (film): ν 2955, 2930, 2860, 1740, 1694, 1460, 1380, 1302, 1283, 1252, 1180, 1159, 1112, 1092, 1039, 988, 947, 890, 838, 781, 754.

HRMS (ESI): calculated for $C_{40}H_{72}NO_7Si_3$ $[M+H]^+$ 762.4611, found 762.4602.





2Zb

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-3-((*Z*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-

dione(2Zb) To protected cyclopropyl-Epo B C16-C17 *Z* isomer **151Zb** (6.8 mg, 0.009 mmol) in CH_2Cl_2 (0.3 mL) and MeOH (0.2 mL) was added CSA (2.1 mg, 0.008 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 15 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 2.7 mg (67%) of **2bZ** as a colorless oil.

TLC: R_f 0.17 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -58.4° (*c* = 0.37, acetone).

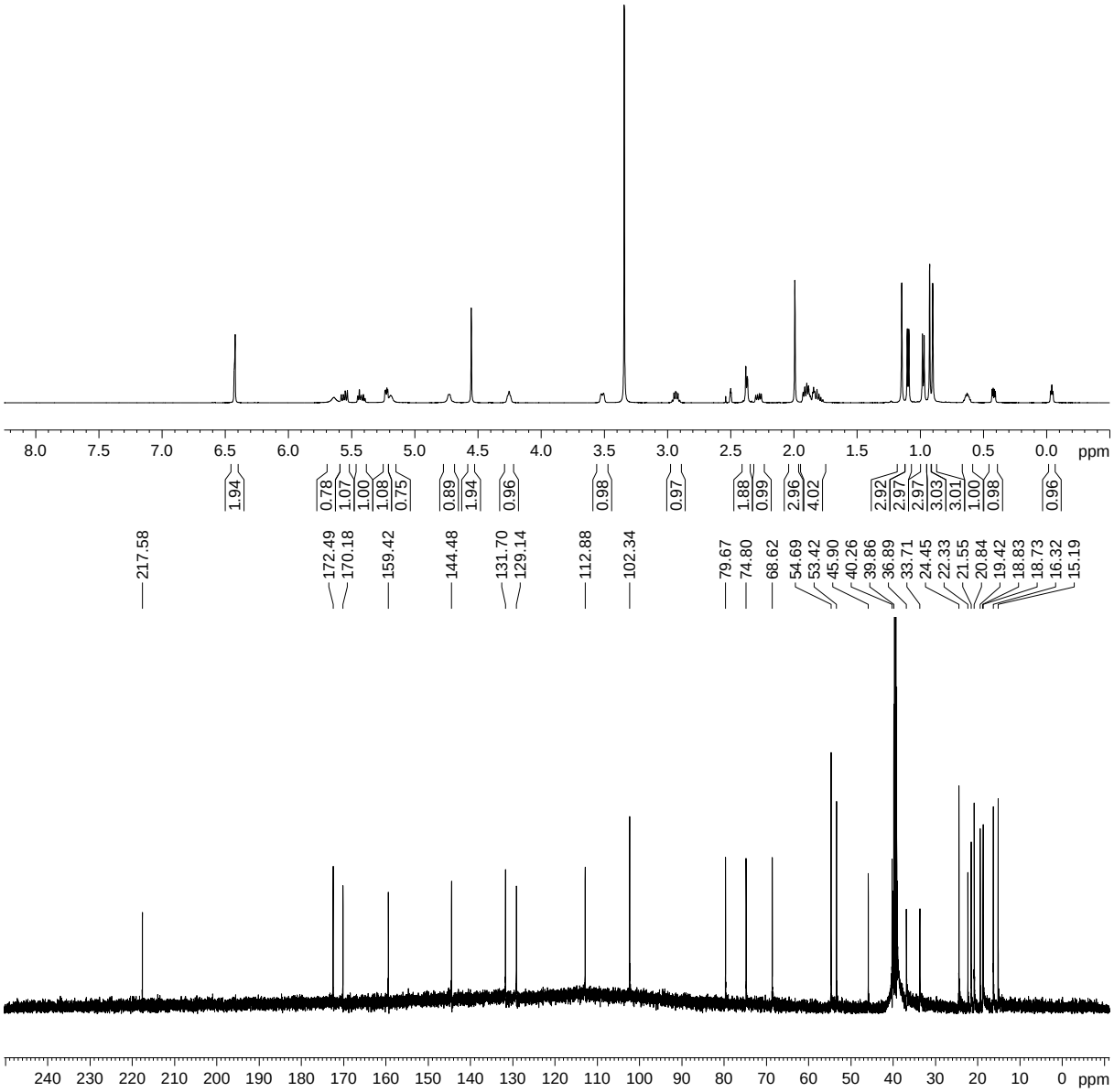
^{1}H-NMR (500 MHz, DMSO-d6): δ = 6.43 (s, 1H), 6.42 (s, 1H), 5.64 (br s, 1H), 5.56 (dd, *J* = 15.7, 8.3 Hz, 1H), 5.42 (ddd, *J* = 15.7, 7.3, 6.8 Hz, 1H), 5.22 (dd, *J* = 7.8, 3.2 Hz, 1H), 5.19 (br s, 1H), 4.73 (br s1H), 4.55 (s, 2H), 4.25 (dd, *J* = 6.4, 5.5 Hz, 1H), 3.52 (dd, *J* = 9.2 Hz, 1H), 2.93 (dq, *J* = 9.2, 6.8 Hz, 1H), 2.37 (d, *J* = 5.5 Hz, 1H), 2.37 (d, *J* = 6.4 Hz, 1H), 2.28 (dd, *J* = 14.9, 7.3 Hz, 1H), 1.99 (s, 3H), 1.91 (dq, *J* = 8.2, 6.8 Hz, 1H), 1.91 (dd, *J* = 14.9, 6.8 Hz, 1H), 1.86 (ddd, *J* = 15.4, 3.6, 3.2 Hz, 1H), 1.79 (dd, *J* = 15.4, 7.8 Hz, 1H), 1.15 (s, 3H), 1.10 (d, *J* = 6.8 Hz, 3H), 0.98 (d, *J* = 6.8 Hz, 3H), 0.93 (s, 3H), 0.89 (s, 3H), 0.63 (dddd, *J* = 8.8, 7.8, 5.4, 3.6 Hz, 1H), 0.42 (dd, *J* = 8.8, 4.0 Hz, 1H), 0.04 (dd, *J* = 5.4, 4.0 Hz, 1H).

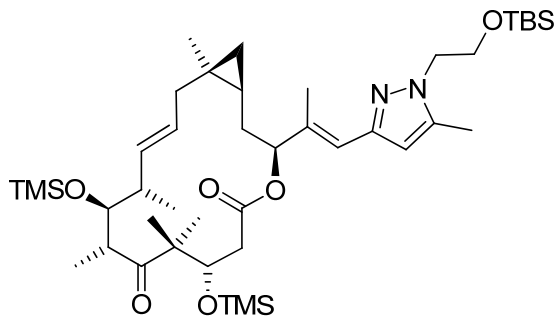
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.6, 172.5, 170.2, 159.4, 144.5, 131.7, 129.1, 112.9, 102.3, 79.7, 74.8, 68.6, 54.7, 53.4, 45.9, 40.3, 39.9, 36.9, 33.7, 24.4, 22.3, 21.5, 20.8, 19.4, 18.8, 18.7, 16.3, 15.2.

IR (film): ν 3445, 3412, 2925, 2870, 1730, 1712, 1689, 1603, 1453, 1368, 1253, 1154, 1072, 1053, 1040, 1011, 979, 958, 938.

HRMS (ESI): calculated for $C_{28}H_{42}NO_7$ $[M+H]^+$ 504.2956, found 504.2959.

Analytical HPLC: Method: eluent B 40-50% (linear gradient from 0-12 min), retention time 8.22 min.





151c

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-3-((*E*)-1-(1-(2-(*tert*-butyldimethylsilyloxy)ethyl)-5-methyl-1*H*-pyrazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-ene-5,9-dione (151c). To a solution of phosphonium salt

140c (49.3 mg, 0.108 mmol) in THF (1 mL) was added *n*-BuLi (1.6 M in hexane, 62 µL, 0.099 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise, the reaction mixture was allowed to warm to room temperature over a period of 3 h and was then stirred for 3 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 15:1) to afford 4.5 mg (32%) of **151c** as a single isomer as a colorless oil.

TLC: R_f 0.26 (hexane/EtOAc 9:1, UV, CPS).

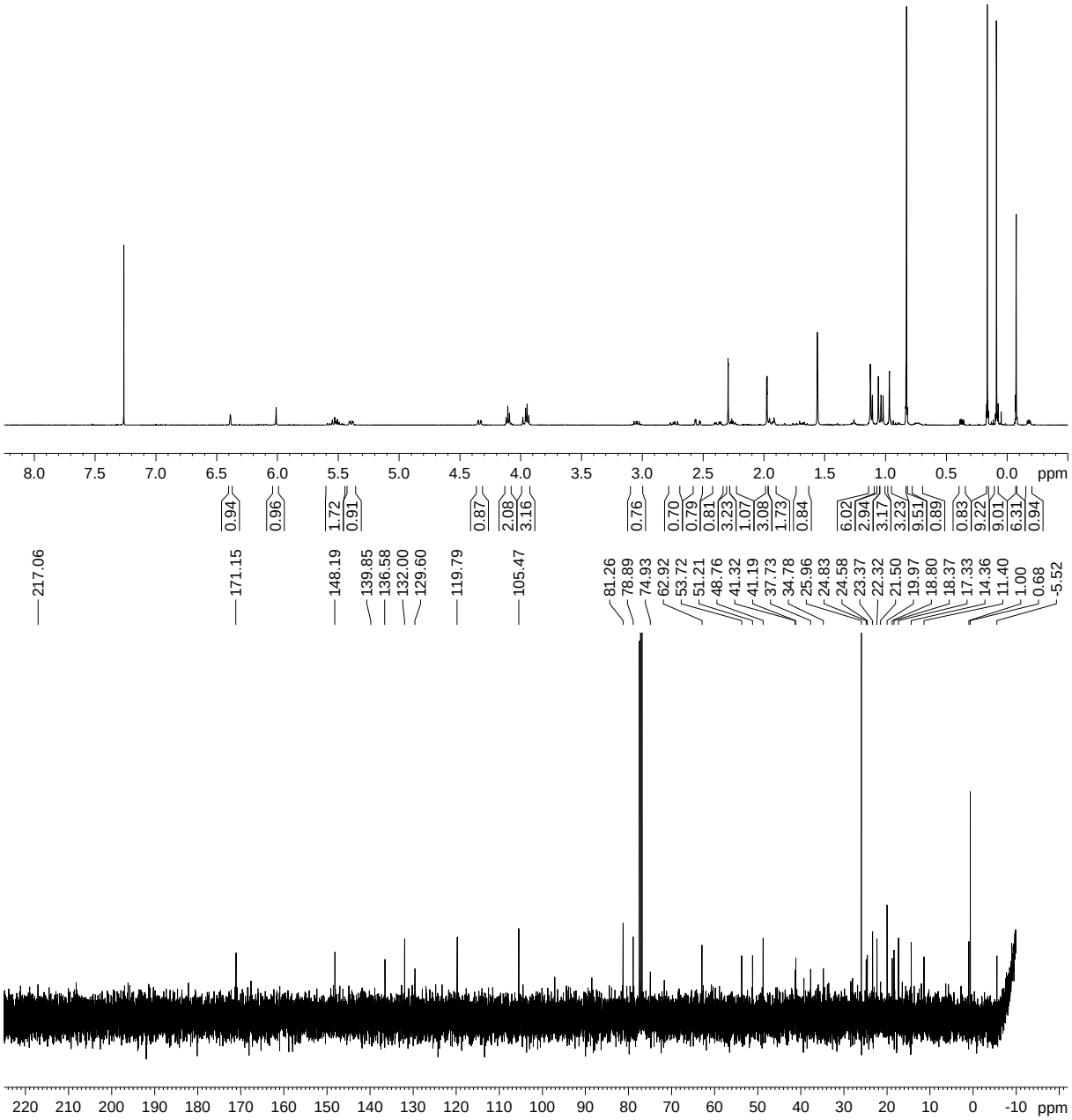
$[\alpha]^{20}_D$: = -1.2° (*c* = 0.21, $CHCl_3$).

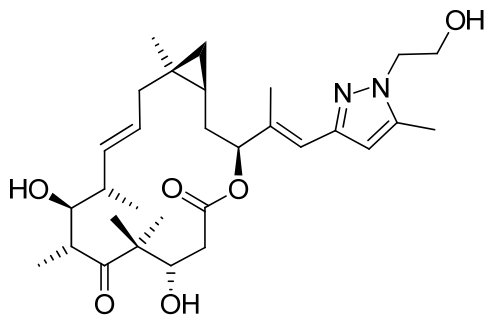
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.39 (q, *J* = 1.2 Hz, 1H), 6.01 (s, 1H), 5.56 (dd, *J* = 15.8, 8.0 Hz, 1H), 5.49 (ddd, *J* = 15.8, 6.0, 5.4 Hz, 1H), 5.39 (dd, *J* = 9.8, 2.7 Hz, 1H), 4.34 (dd, *J* = 9.6, 2.2 Hz, 1H), 4.11 (t, *J* = 5.4 Hz, 2H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.94 (t, *J* = 5.4 Hz, 2H), 3.05 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.74 (dd, *J* = 14.5, 9.6 Hz, 1H), 2.54 (dd, *J* = 14.5, 2.2 Hz, 1H), 2.38 (dd, *J* = 15.2, 5.4 Hz, 1H), 2.29 (s, 3H), 2.25 (dq, *J* = 8.0, 7.0 Hz, 1H), 1.97 (d, *J* = 1.2 Hz, 3H), 1.94 (dd, *J* = 15.2, 6.0 Hz, 1H), 1.93 (ddd, *J* = 15.1, 2.7, 2.6 Hz, 1H), 1.68 (ddd, *J* = 15.1, 9.9, 9.8, 1H), 1.12 (s, 3H), 1.12 (d, *J* = 6.8 Hz, 3H), 1.06 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.83 (s, 9H), 0.74 (dddd, *J* = 9.9, 8.9, 5.7, 2.6 Hz, 1H), 0.37 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.09 (s, 9H), -0.08 (s, 6H), -0.18 (dd, *J* = 5.7, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 217.1, 171.2, 148.2, 139.8, 136.6, 132.0, 129.6, 119.8, 105.5, 81.3, 78.9, 74.9, 62.9, 53.7, 51.2, 48.8, 41.3, 41.2, 37.7, 34.8, 26.0, 24.8, 24.6, 23.4, 22.3, 21.5, 20.0, 18.8, 18.4, 17.3, 14.4, 11.4, 1.0, 0.7, -5.5.

IR (film): ν 2955, 2930, 2860, 1738, 1694, 1468, 1459, 1382, 1302, 1252, 1180, 1160, 1115, 1038, 890, 837, 778, 751.

HRMS (ESI): calculated for $C_{42}H_{77}NO_6Si_3$ $[M+H]^+$ 789.5084, found 789.5091.





2c

(1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-3-((*E*)-1-(1-(2-hydroxyethyl)-5-methyl-1*H*-pyrazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadec-

13-ene-5,9-dione (2c). To protected cyclopropyl-Epo B **151c** (4.2 mg, 0.005 mmol) in CH_2Cl_2 (0.3 mL) and MeOH (0.2 mL) was added CSA (2.4mg, 0.010 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 20 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:3) to yield 2.1 mg (74%) of **2c** as a semi solid.

TLC: R_f 0.11 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -49.2° (*c* = 0.10, $CHCl_3$).

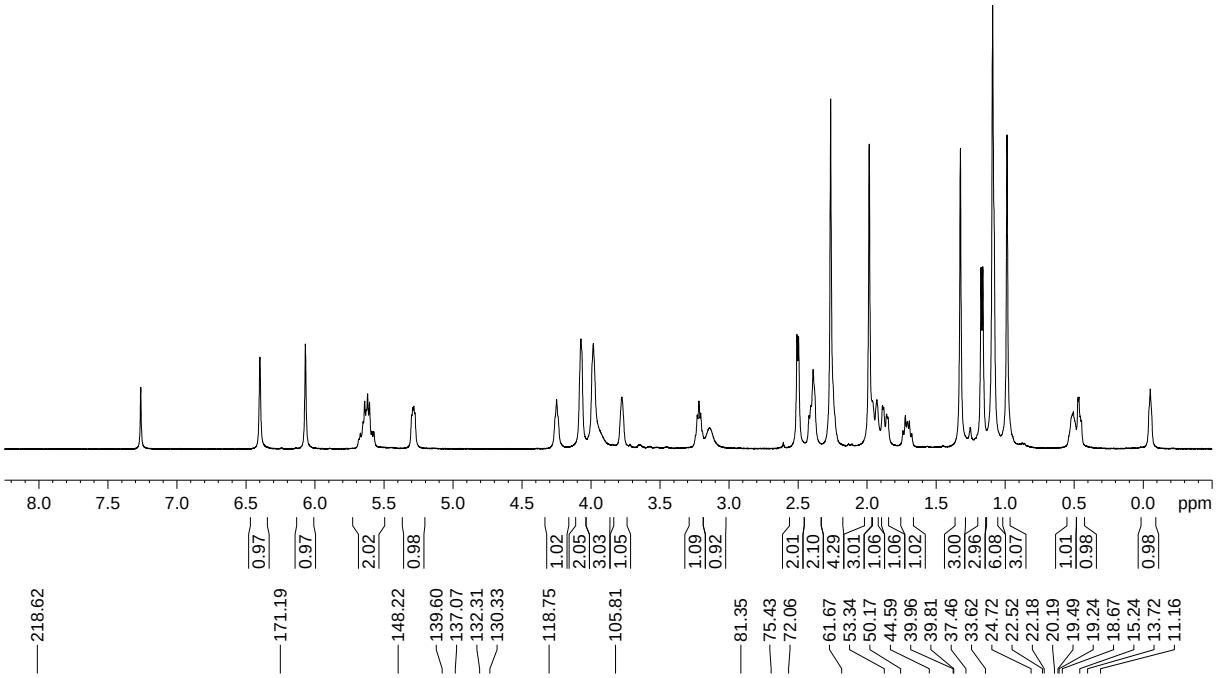
^{1}H-NMR (500 MHz, $CDCl_3$): δ = 6.40 (s, 1H), 6.07 (s, 1H), 5.66 (dd, *J* = 15.6, 6.0 Hz, 1H), 5.60 (ddd, *J* = 15.6, 6.7, 4.8 Hz, 1H), 5.29 (dd, *J* = 8.1, 4.0 Hz, 1H), 4.25 (t, *J* = 5.4 Hz, 1H), 4.07 (t, *J* = 4.1 Hz, 2H), 3.98 (t, *J* = 4.1 Hz, 2H), 3.94 (br s, 1H), 3.78 (d, *J* = 9.2 Hz, 1H), 3.22 (dq, *J* = 9.2, 6.8 Hz, 1H), 3.14 (br s, 1H), 2.50 (d, *J* = 5.4 Hz, 2H), 2.40 (dd, *J* = 15.2, 6.7 Hz, 1H), 2.40 (br s, 1H), 2.26 (s, 3H), 2.25(qd, *J* = 6.8, 6.0 Hz, 1H), 1.99 (s, 3H), 1.94 (dd, *J* = 15.1, 4.0 Hz, 1H), 1.87 (dd, *J* = 15.2, 4.8 Hz, 1H), 1.71 (ddd, *J* = 15.1, 8.6, 8.1 Hz, 1H), 1.32 (s, 3H), 1.17 (d, *J* = 6.8 Hz, 3H), 1.09 (s, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.99 (s, 3H), 0.51 (dddd, *J* = 8.8, 8.7, 5.4, 2.6 Hz, 1H), 0.46 (dd, *J* = 8.4, 3.6 Hz, 1H), 0.05 (dd, *J* = 4.7, 3.6 Hz, 1H).

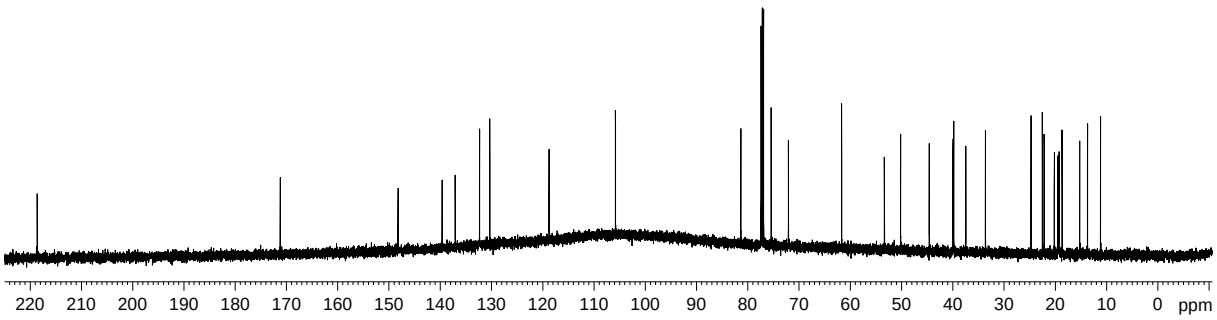
^{13}C-NMR (125 MHz, $CDCl_3$): δ = 218.6, 171.2, 148.2, 139.6, 137.1, 132.3, 130.3, 118.8, 105.8, 81.3, 75.4, 72.1, 61.7, 53.3, 50.2, 44.6, 40.0, 39.8, 37.5, 33.6, 24.7, 22.5, 22.2, 20.2, 19.5, 19.2, 18.7, 15.2, 13.7, 11.2.

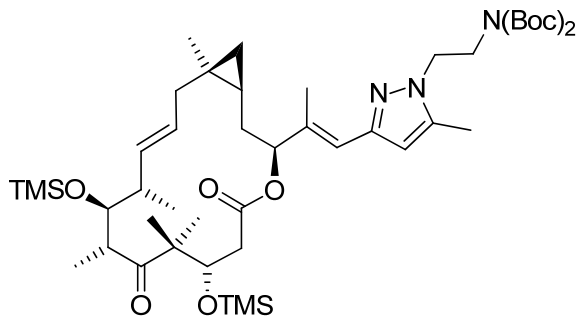
IR (film): ν 3421, 2927, 2873, 1729, 1690, 1545, 1453, 1372, 1290, 1256, 1071, 981, 874, 779, 758, 669, 626, 541.

HRMS (ESI): calculated for $C_{30}H_{47}N_2O_6$ $[M+H]^+$ 531.3429, found 531.3429.

Analytical HPLC: Method: eluent B 40-65% (linear gradient from 0-12 min), retention time 8.37 min.







152h

Di-*tert*-butyl (2-(5-methyl-3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-8,8,10,12,16-pentamethyl-5,9-dioxo-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)-1*H*-pyrazol-1-yl)ethyl)dicarbamate (152h). To a solution of phosphonium salt **145h** (36.0 mg, 0.062 mmol) in THF (1 mL) was added *n*-BuLi (1.6 M in

hexane, 37 µL, 0.059 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cooled to -78 °C. Methyl ketone **148** (9.5 mg, 0.017 mmol) in THF (0.5 mL) was added dropwise, the reaction mixture was allowed to warm to room temperature over a period of 2 h and was then stirred for 5 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 9:1) to afford 3.0 mg (20%) of an inseparable 10:1 mixture of **152h** and its C16-C17 *Z* isomer as a semi-solid.

TLC: R_f 0.13 (hexane/EtOAc 9:1, UV, CPS).

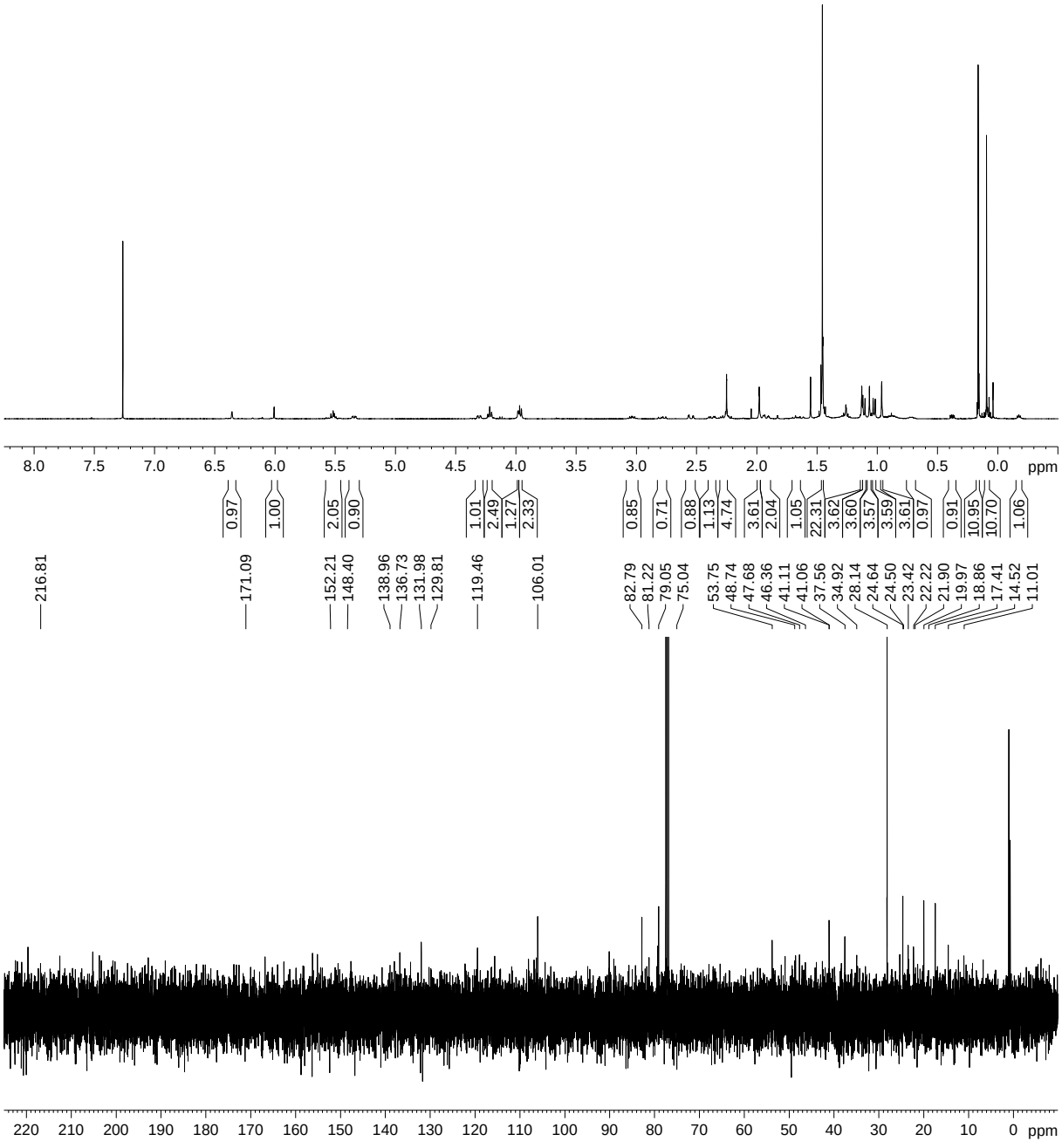
$[\alpha]^{20}_D$: = -1.8° (*c* = 0.15, $CHCl_3$).

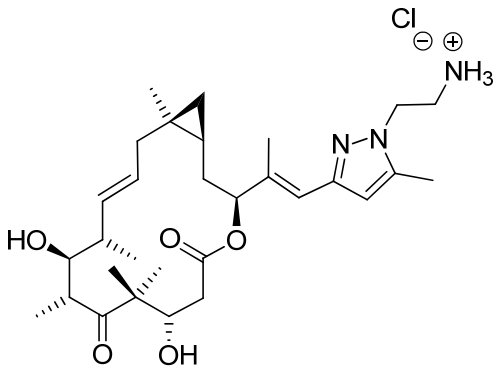
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.36 (q, *J* = 1.2 Hz, 1H), 6.01 (s, 1H), 5.55 (dd, *J* = 15.7, 7.3 Hz, 1H), 5.48 (ddd, *J* = 15.7, 5.6, 5.2 Hz, 1H), 5.34 (dd, *J* = 9.5, 2.6 Hz, 1H), 4.31 (dd, *J* = 9.3, 2.2 Hz, 1H), 4.22 (t, *J* = 6.2 Hz, 2H), 3.97 (t, *J* = 6.2 Hz, 2H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.04 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.79 (dd, *J* = 14.7, 9.3 Hz, 1H), 2.55 (dd, *J* = 14.7, 2.2 Hz, 1H), 2.37 (dd, *J* = 15.3, 5.2 Hz, 1H), 2.25 (s, 3H), 2.25 (dq, *J* = 7.3, 7.0 Hz, 1H), 1.98 (d, *J* = 1.2 Hz, 3H), 1.96 (dd, *J* = 15.3, 5.6, 1H), 1.92 (ddd, *J* = 15.2, 2.7, 2.6 Hz, 1H), 1.66 (ddd, *J* = 15.2, 10.4, 9.5, 1H), 1.46 (s, 18H), 1.13 (s, 3H), 1.11 (d, *J* = 7.0 Hz, 3H), 1.07 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.96 (s, 3H), 0.72 (dddd, *J* = 10.3, 8.9, 5.6, 2.7 Hz, 1H), 0.38 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.09 (s, 9H), -0.17 (dd, *J* = 5.6, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.8, 171.1, 152.2, 148.4, 139.0, 136.7, 132.0, 129.8, 119.5, 106.0, 82.8, 81.2, 79.1, 75.0, 53.7, 48.7, 47.7, 46.4, 41.1, 41.1, 37.6, 34.9, 28.1, 24.6, 24.5, 23.4, 22.2, 21.9, 20.0, 18.9, 17.4, 14.5, 11.0, 1.0, 0.7.

IR (film): ν 2959, 2932, 2872, 1792, 1739, 1696, 1455, 1391, 1366, 1345, 1305, 1177, 1129, 1092, 1039, 987, 949, 890, 843, 754.

HRMS (ESI): calculated for $C_{46}H_{80}N_3O_9Si_2$ $[M+H]^+$ 874.5428, found 874.5428.


8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 ppm
0.97 1.00 2.05 0.90 1.01 2.49 1.27 2.33 0.85 0.71 0.88 1.13 4.74 3.61 2.04 1.05 22.31 3.62 3.60 3.57 3.59 3.61 0.97 0.91 10.95 10.70 1.06
216.81 171.09 152.21 148.40 138.96 136.73 131.98 129.81 119.46 106.01 82.79 81.22 79.05 75.04 53.75 48.74 47.68 46.36 41.11 41.06 37.56 34.92 28.14 24.64 24.50 23.42 22.22 21.90 19.97 18.86 17.41 14.52 11.01
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm


Cl
NH3
N
N
HO
O
O
O
OH

2h

2-(3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)-5-methyl-1*H*-pyrazol-1-yl)ethanaminium chloride (2h). To a 10:1 mixture of protected cyclopropyl-Epo B **152h** and its C16-C17 *Z* isomer (3.0 mg, 0.003 mmol) was added HCl (1.0 M in EtOAc, 0.52 mL, 0.521 mmol) at room temperature. The reaction mixture was stirred for 15 h, a second portion of HCl (0.26 mL, 0.265 mmol) was then added and stirring was continued for further 25 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5) to yield 1.5 mg (85%) of **2h** as a single isomer as a colorless oil.

TLC: R_f 0.05 ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5, UV, CPS).

$[\alpha]^{20}_D$: = -27.8° (*c* = 0.29, MeOH).

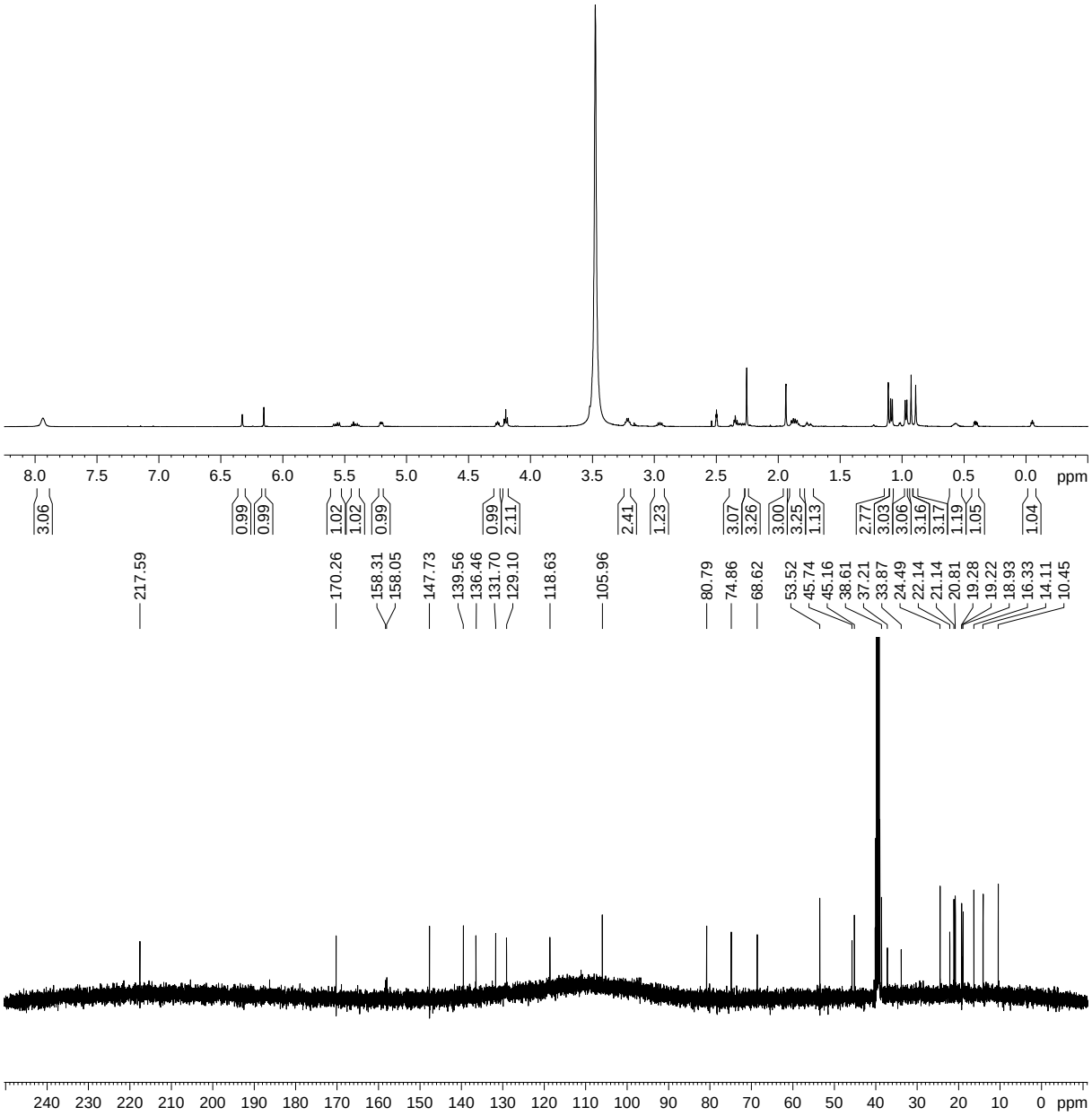
^{1}H-NMR (500 MHz, DMSO-d6): δ = 7.94 (t, *J* = 5.9 Hz, 3H), 6.33 (q, *J* = 0.9 Hz, 1H), 6.15 (s, 1H), 5.57 (dd, *J* = 15.6, 8.1 Hz, 1H), 5.41 (ddd, *J* = 15.6, 6.8, 6.7 Hz, 1H), 5.21 (dd, *J* = 7.7, 3.5 Hz, 1H), 4.27 (dd, *J* = 7.3, 5.0 Hz, 1H), 4.20 (t, *J* = 6.3 Hz, 2H), 3.51 (d, *J* = 9.3 Hz, 1H), 3.22 (tq, *J* = 6.3, 5.9 Hz, 2H), 2.96 (qd, *J* = 9.3, 6.9 Hz, 1H), 2.37 (dd, *J* = 16.1, 5.0 Hz, 1H), 2.34 (dd, *J* = 16.1, 7.3 Hz, 1H), 2.30 (dd, *J* = 14.1, 6.7 Hz, 1H), 2.26 (s, 3H), 1.94 (d, *J* = 0.9 Hz, 3H), 1.87 (dd, *J* = 14.1, 6.8 Hz, 1H), 1.87 (d, *J* = 8.1 Hz, 1H), 1.86 (ddd, *J* = 15.1, 7.8, 7.7 Hz, 1H), 1.75 (ddd, *J* = 15.1, 3.9, 3.5 Hz, 1H), 1.11 (s, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.9 Hz, 3H), 0.93 (s, 3H), 0.89 (s, 3H), 0.51 (dddd, *J* = 8.8, 7.8, 5.4, 4.0 Hz, 1H), 0.41 (dd, *J* = 8.8, 4.0 Hz, 1H), 0.05 (dd, *J* = 5.4, 4.0 Hz, 1H).

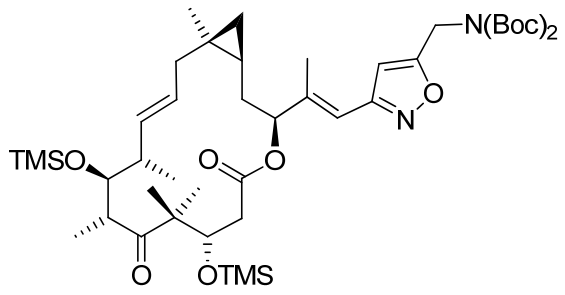
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.6, 170.3, 158.3, 158.0, 147.7, 139.6, 136.5, 131.7, 129.1, 118.6, 106.0, 80.8, 74.9, 68.6, 53.5, 45.7, 45.2, 38.6, 37.2, 33.9, 24.5, 22.1, 21.1, 20.8, 19.3, 19.2, 18.9, 16.3, 14.1, 10.5.

IR (film): ν 3437, 2924, 2871, 1680, 1548, 1452, 1429, 1372, 1254, 1202, 1180, 1135, 1026, 1010, 981, 836, 799, 722.

HRMS (ESI): calculated for $C_{30}H_{48}N_3O_5$ $[M+H]^+$ 530.3588, found 530.3581.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 30-55% (linear gradient from 0-12 min), retention time 8.01 min.





152g

Di-*tert*-butyl ((3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-8,8,10,12,16-pentamethyl-5,9-dioxo-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methyl)dicarbamate (152g). To a solution of phosphonium salt **145g** (48.7

mg, 0.109 mmol) in THF (1 mL) was added LiHMDS (1.0 M in THF, 99 µL, 0.109 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 9.3 mg (61%) of **152g** as a single isomer as a colorless oil.

TLC: R_f 0.35 (hexane/EtOAc 9:1, UV, CPS).

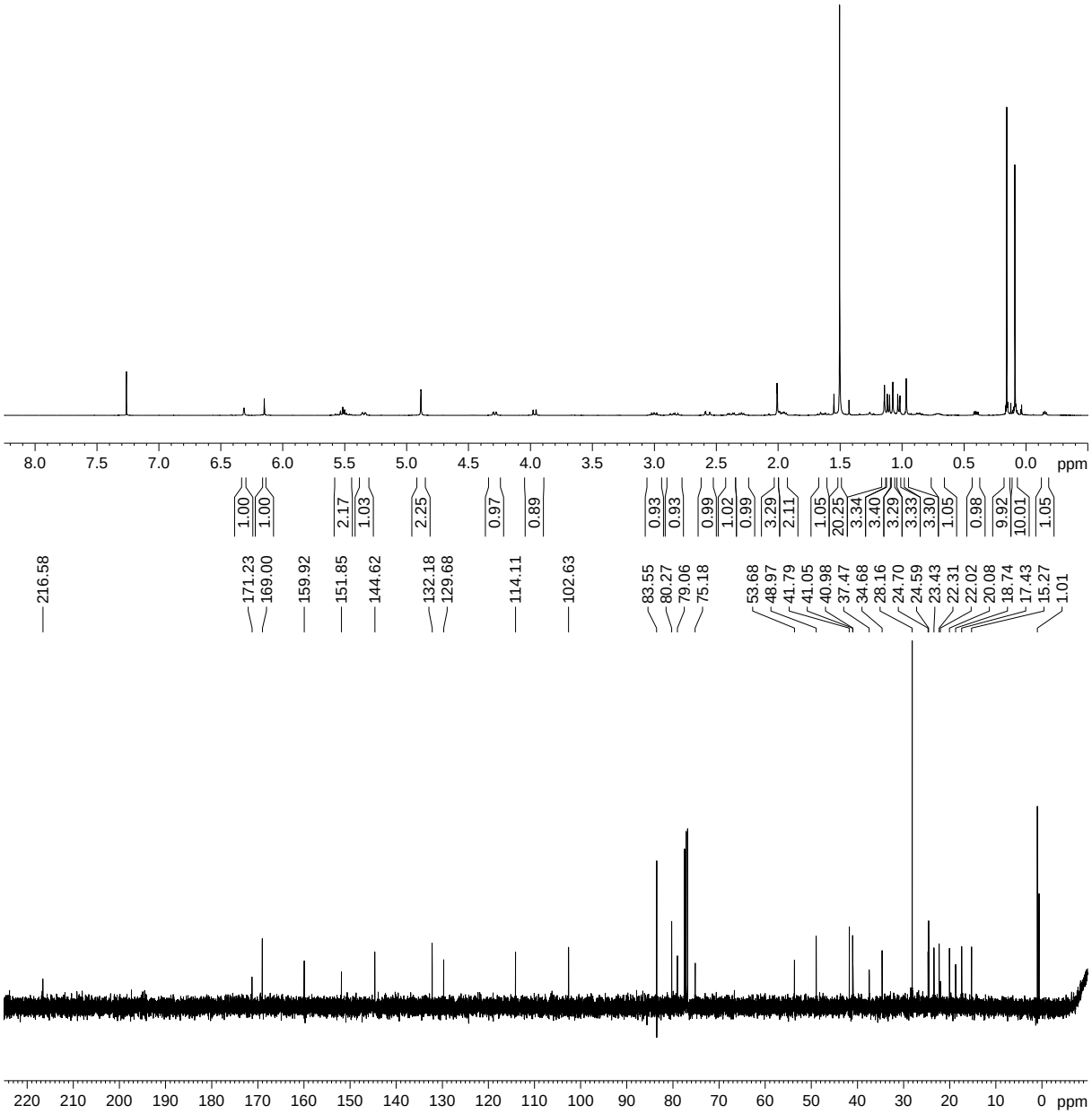
$[\alpha]^{20}_D$: = -2.4° (*c* = 0.42, $CHCl_3$).

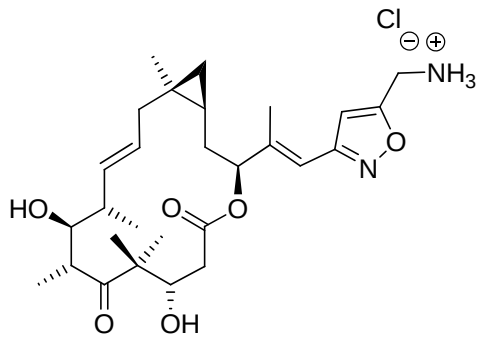
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.31 (q, *J* = 1.2 Hz, 1H), 6.15 (s, 1H), 5.54 (dd, *J* = 15.7, 7.8 Hz, 1H), 5.48 (dddd, *J* = 15.7, 6.0, 5.7 Hz, 1H), 5.35 (dd, *J* = 9.6, 2.4 Hz, 1H), 4.89 (s, 2H), 4.29 (dd, *J* = 9.7, 2.0 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.00 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.84 (dd, *J* = 14.7, 9.7 Hz, 1H), 2.57 (dd, *J* = 14.7, 2.0 Hz, 1H), 2.38 (dd, *J* = 15.0, 5.7 Hz, 1H), 2.30 (dq, *J* = 7.8, 7.0 Hz, 1H), 2.00 (d, *J* = 1.2 Hz, 3H), 1.98 (ddd, *J* = 15.3, 2.6, 2.4 Hz, 1H), 1.96 (dd, *J* = 15.0, 6.0 Hz, 1H), 1.64 (ddd, *J* = 15.3, 9.8, 9.6, 1H), 1.50 (s, 18H), 1.14 (s, 3H), 1.11 (d, *J* = 7.0 Hz, 3H), 1.08 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.71 (dddd, *J* = 9.8, 8.9, 5.7, 2.6 Hz, 1H), 0.40 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.09 (s, 9H), -0.17 (dd, *J* = 5.7, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.6, 171.2, 169.0, 159.9, 151.8, 144.6, 132.2, 129.7, 114.1, 102.6, 83.5, 80.3, 79.1, 75.2, 53.7, 49.0, 41.8, 41.0, 41.0, 37.5, 34.7, 28.2, 24.7, 24.6, 23.4, 22.3, 22.0, 20.1, 18.7, 17.4, 15.3, 1.0, 0.6.

IR (film): ν 2979, 2958, 1795, 1740, 1697, 1602, 1474, 1455, 1423, 1383, 1368, 1305, 1251, 1175, 1160, 1141, 1115, 1037, 949, 890, 840, 761, 754.

HRMS (ESI): calculated for $C_{44}H_{74}N_2O_{10}Si_2$ $[M+Na]^+$ 869.4774, found 869.4793.





2g

(3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)isoxazol-5-

yl)methanaminium chloride (2g). To protected cyclopropyl-Epo B **152g** (9.3 mg, 0.011 mmol) was added HCl (1.0 M in EtOAc, 0.99 mL, 0.988 mmol) at room temperature. The reaction mixture was stirred for 15 h, a second portion of HCl (0.99 mL, 0.988 mmol) was then added and stirring was continued for further 25 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5) to yield 3.7 mg (67%) of **2g** as a colorless oil.

TLC: R_f 0.07 ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5, UV, CPS).

$[\alpha]^{20}_D$: = -74.3° (*c* = 0.38, MeOH).

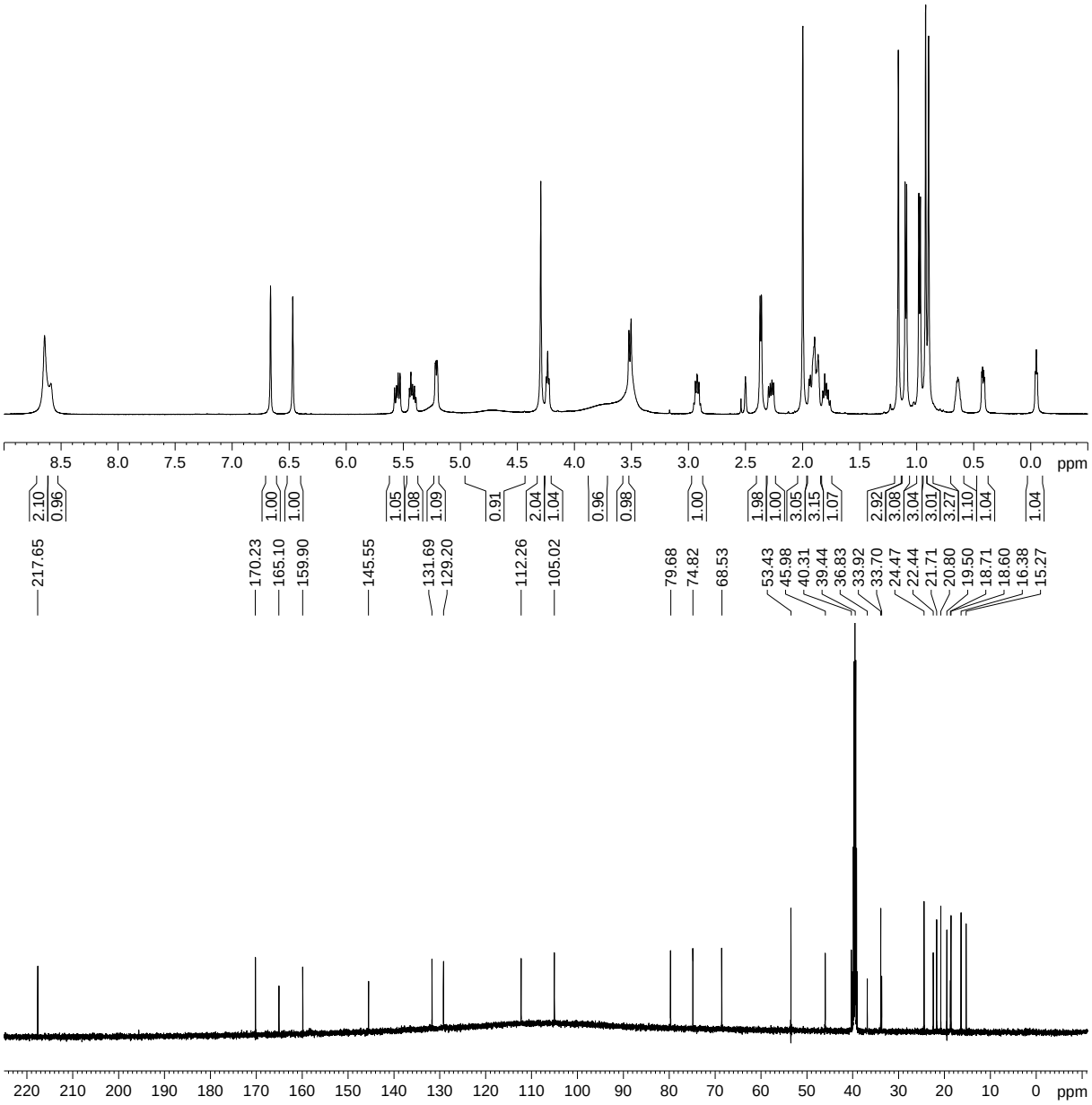
^{1}H-NMR (500 MHz, DMSO-d6): δ = 8.65 (br s, 2H), 8.59 (s, 1H), 6.67 (s, 1H), 6.47 (s, 1H), 5.55 (dd, *J* = 15.7, 8.3 Hz, 1H), 5.42 (ddd, *J* = 15.6, 7.4, 5.9 Hz, 1H), 5.21 (dd, *J* = 7.9, 2.2 Hz, 1H), 4.73 (br s, 1H), 4.29 (s, 2H), 4.23 (t, *J* = 6.3 Hz, 1H), 3.79 (br s, 1H), 3.51 (d, *J* = 9.1 Hz, 1H), 2.92 (dq, *J* = 9.1, 6.8 Hz, 1H), 2.37 (dd, *J* = 16.1, 5.0 Hz, 1H), 2.37 (d, *J* = 6.3 Hz, 2H), 2.28 (dd, *J* = 14.7, 7.4 Hz, 1H), 2.00 (s, 3H), 1.92 (dd, *J* = 14.7, 5.9, 1H), 1.90 (dq, *J* = 8.3, 6.8 Hz, 1H), 1.88 (ddd, *J* = 15.4, 3.9, 2.2 Hz, 1H), 1.78 (dd, *J* = 15.4, 8.7, 7.9 Hz, 1H), 1.16 (s, 3H), 1.10 (d, *J* = 6.8 Hz, 3H), 0.98 (d, *J* = 6.8 Hz, 3H), 0.92 (s, 3H), 0.90 (s, 3H), 0.64 (dddd, *J* = 8.7, 8.7, 4.9, 3.9 Hz, 1H), 0.42 (dd, *J* = 8.7, 3.8 Hz, 1H), 0.05 (dd, *J* = 4.9, 3.8 Hz, 1H).

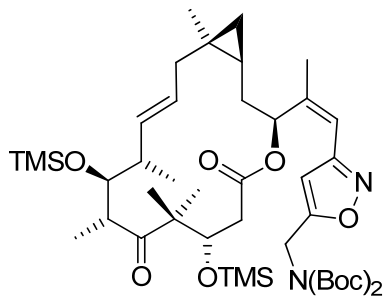
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.7, 170.2, 165.1, 159.9, 145.6, 131.7, 129.2, 112.3, 105.0, 79.7, 74.8, 68.5, 53.4, 46.0, 40.3, 39.4, 36.8, 33.9, 33.7, 24.5, 22.4, 21.7, 20.8, 19.5, 18.7, 18.6, 16.4, 15.3.

IR (film): ν 3418, 2925, 2876, 1729, 1680, 1638, 1451, 1431, 1378, 1253, 1201, 1179, 1134, 1049, 1025, 1007, 980, 937, 834, 780, 722.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_6$ $[M+H]^+$ 503.3116, found 503.3122.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 30-40% (linear gradient from 0-12 min), retention time 7.61 min.



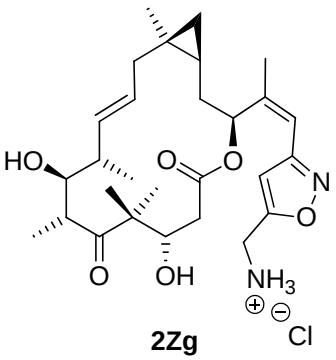


152Zg

Di-*tert*-butyl ((3-((*Z*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-8,8,10,12,16-pentamethyl-5,9-dioxo-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methyl)dicarbamate (152Zg). To a solution of phosphonium salt **145g**

(48.7 mg, 0.109 mmol) in THF (1 mL) was added LiHMDS (1.0 M in THF, 99 µL, 0.109 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 2.7 mg (18%) of **152Zg** as a single isomer as a colorless oil.

TLC: R_f 0.30 (hexane/EtOAc 9:1, UV, CPS).



2Zg

(3-((*Z*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methanaminium chloride (2Zg). To protected cyclopropyl-Epo B **152Zg** (3.5 mg, 0.004 mmol) was added HCl (1.0 M in EtOAc, 0.41 mL, 0.41 mmol) at room temperature. The reaction mixture was stirred for 24 h, a second portion of HCl (0.41 mL, 0.41 mmol) was then added and stirring was continued for further 24 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5) to yield 2.0 mg (93%) of **2Zg** as a colorless oil.

TLC: R_f 0.08 ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5, UV, CPS).

$[\alpha]^{20}_D$: = -6.7° (*c* = 0.047, MeOH).

^{1}H-NMR (500 MHz, DMSO-d6): δ = 8.40 (br s, 3H), 6.60 (s, 1H), 6.19 (q, *J* = 1.2 Hz, 1H), 6.01 (dd, *J* = 8.7, 1.8 Hz, 1H), 5.55 (dd, *J* = 15.6, 8.6 Hz, 1H), 5.41 (dd, *J* = 15.6, 7.6, 6.4 Hz, 1H), 5.20 (d, *J* = 6.6 Hz, 1H), 4.74 (d, *J* = 6.6 Hz, 1H), 4.29 (s, 2H), 4.18 (ddd, *J* = 8.7, 6.6, 5.3 Hz, 1H), 3.51 (dd, *J* = 9.0, 6.6 Hz, 1H), 2.90 (dq, *J* = 9.0, 6.8 Hz, 1H), 2.32 (d, *J* = 8.7 Hz, 1H), 2.31 (d, *J* = 5.3 Hz, 1H),), 2.26 (dd, *J* = 14.6, 7.6 Hz, 1H), 1.92 (dd, J = 14.7, 6.4 Hz, 1H), 1.92-1.88 (m, 1H), 1.91 (d, *J* = 1.2 Hz, 3H), 1.89 (dq, *J* = 8.6, 6.8, Hz, 1H), 1.78 (ddd, *J*

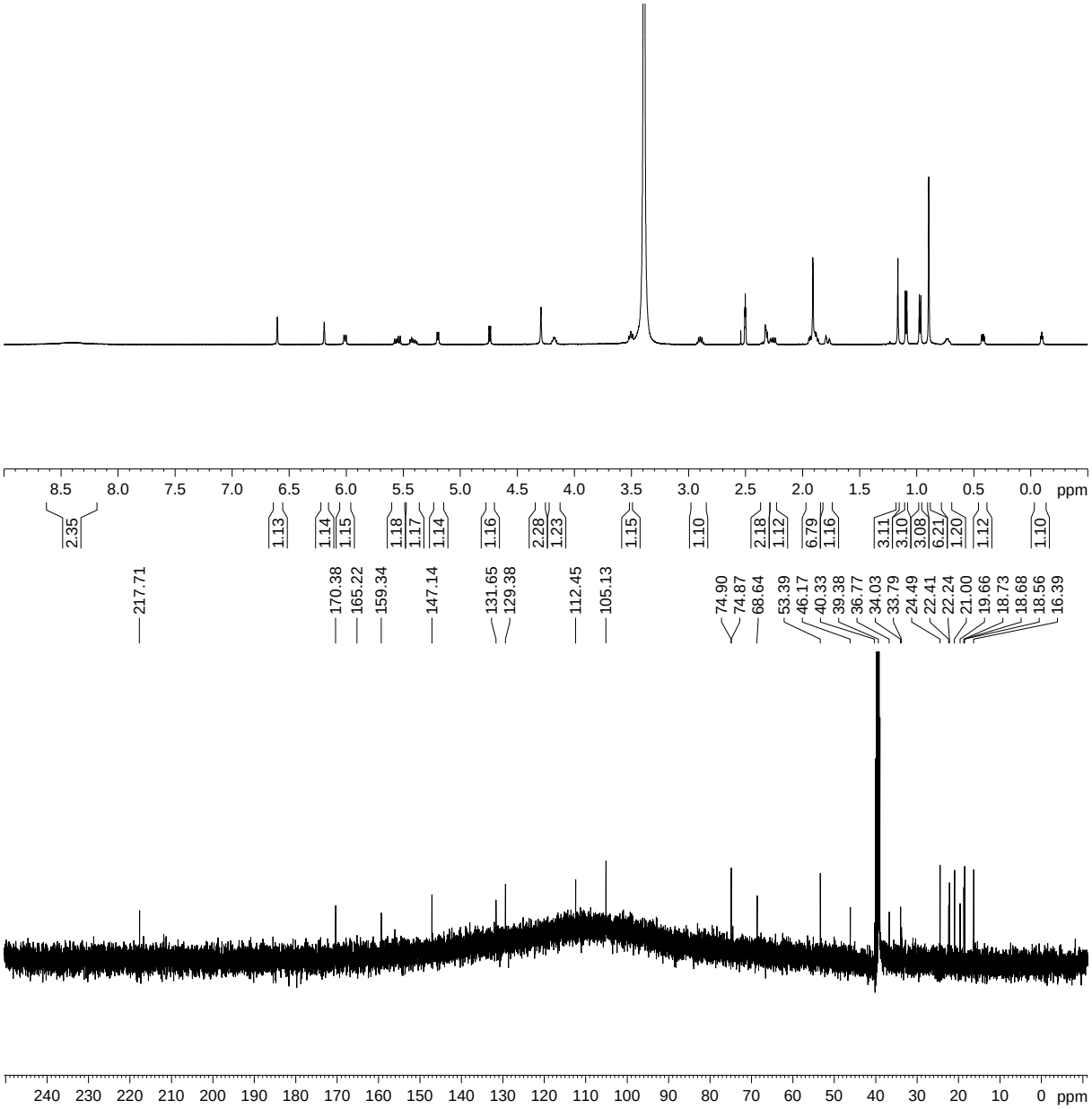
= 15.4, 2.6, 2.6 Hz, 1H), 1.16 (s, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.8 Hz, 3H), 0.89 (s, 3H), 0.89 (s, 3H), 0.73 (dddd, *J* = 8.7, 8.8, 5.8, 3.9 Hz, 1H), 0.42 (dd, *J* = 8.8, 4.0 Hz, 1H), 0.10 (dd, *J* = 5.3, 4.0 Hz, 1H).

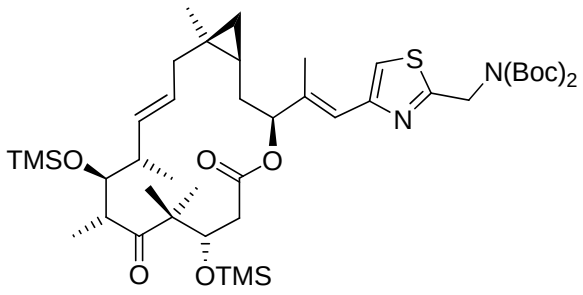
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.7, 170.4, 165.2, 159.3, 147.1, 131.7, 129.4, 112.4, 105.1, 74.9, 74.9, 68.6, 53.4, 46.2, 40.3, 39.4, 36.8, 34.0, 33.8, 24.5, 22.4, 22.2, 21.0, 19.7, 18.7, 18.7, 18.6, 16.4.

IR (film): ν 3450, 2940, 2876, 1711, 1643, 1611, 1568, 1541, 1449, 1415, 1378, 1337, 1293, 1264, 1176, 1149, 1054, 1019, 984, 963, 876, 755, 711, 664, 617.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_6$ $[M+H]^+$ 503.3116, found 503.3120.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 10-40% (linear gradient from 0-12 min), retention time 9.51 min.





152f

Di-*tert*-butyl ((4-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-8,8,10,12,16-pentamethyl-5,9-dioxo-7,11-bis(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)thiazol-2-yl)methyl)dicarbamate (152f). To a solution of phosphonate **145f** (46.3 mg, 0.100 mmol) in THF (1 mL) was added LiHMDS (1.0 M in THF, 91 µL, 0.91 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was then added dropwise and the reaction mixture was allowed to warm to room temperature over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 20:1) to afford 6.7 mg (42%) of **152f** as a single isomer as a colorless oil.

TLC: R_f 0.36 (hexane/EtOAc 9:1, UV, CPS).

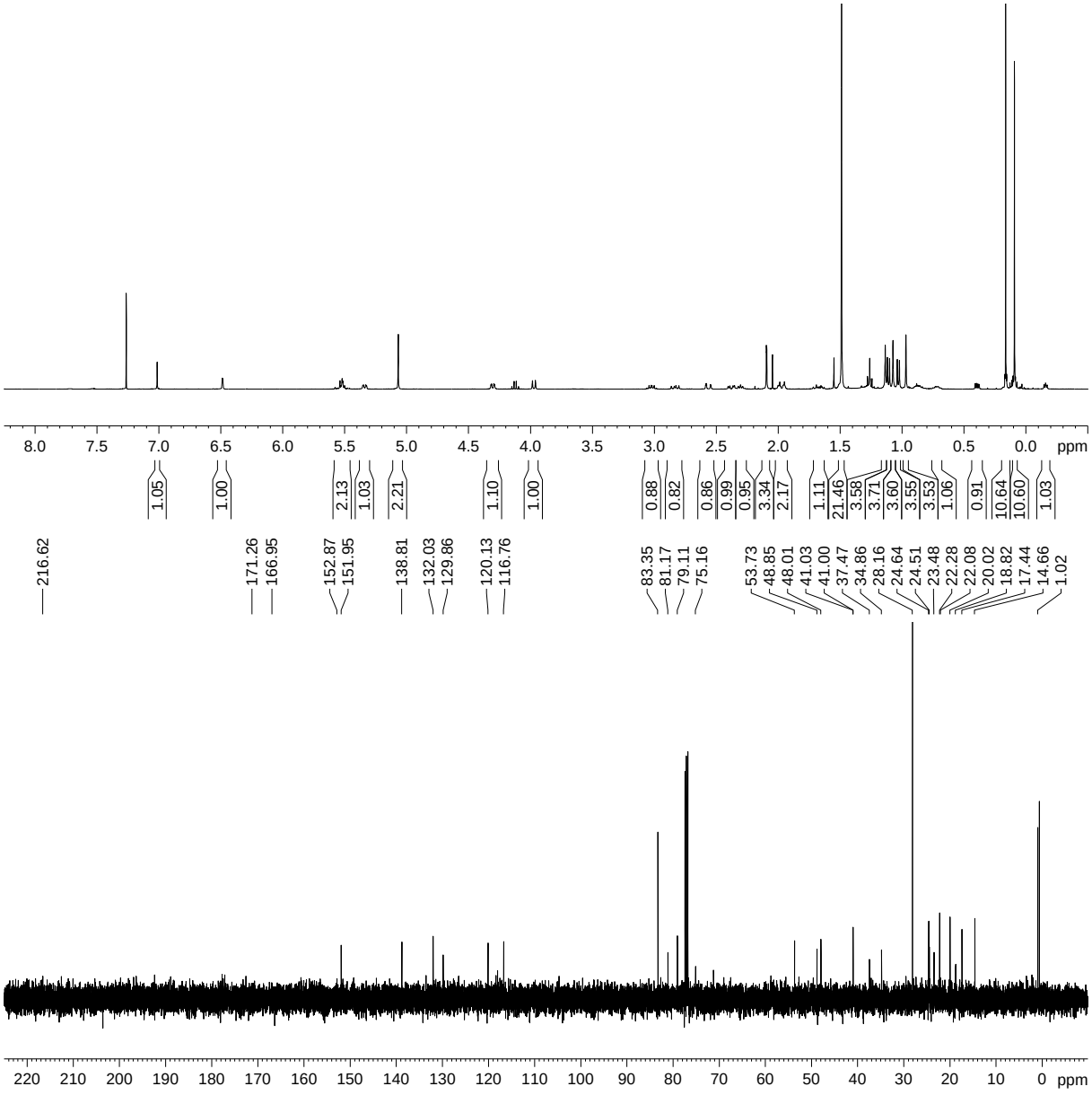
$[\alpha]^{20}_D$: = -10.7° (*c* = 0.29, $CHCl_3$).

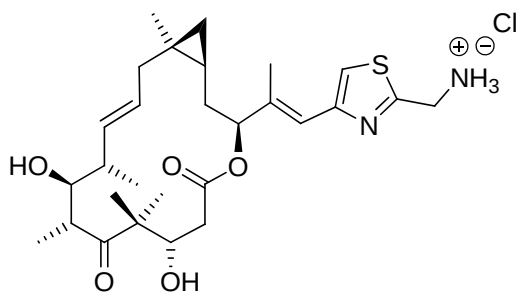
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.01 (s, 1H), 6.49 (q, *J* = 1.2 Hz, 1H), 5.55 (dd, *J* = 15.5, 7.3 Hz, 1H), 5.50 (dddd, *J* = 15.5, 5.6, 5.4 Hz, 1H), 5.34 (dd, *J* = 9.6, 2.5 Hz, 1H), 5.07 (s, 2H), 4.30 (dd, *J* = 9.6, 2.1 Hz, 1H), 3.97 (d, *J* = 9.7 Hz, 1H), 3.02 (dq, *J* = 9.7, 7.0 Hz, 1H), 2.83 (dd, *J* = 14.7, 9.6 Hz, 1H), 2.57 (dd, *J* = 14.7, 2.1 Hz, 1H), 2.38 (dd, *J* = 15.0, 5.4 Hz, 1H), 2.30 (dq, *J* = 7.3, 7.0 Hz, 1H), 2.09 (d, *J* = 1.2 Hz, 3H), 1.98 (ddd, *J* = 15.0, 5.4 Hz, 1H), 1.97 (dd, *J* = 15.1, 2.6, 2.5 Hz, 1H), 1.67 (ddd, *J* = 15.1, 10.1, 9.6, 1H), 1.49 (s, 18H), 1.14 (s, 3H), 1.11 (d, *J* = 7.0 Hz, 3H), 1.07 (s, 3H), 1.03 (d, *J* = 7.0 Hz, 3H), 0.97 (s, 3H), 0.72 (dddd, *J* = 10.1, 8.9, 5.6, 2.6 Hz, 1H), 0.39 (dd, *J* = 8.9, 4.2 Hz, 1H), 0.16 (s, 9H), 0.09 (s, 9H), -0.17 (dd, *J* = 5.6, 4.2 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.6, 171.3, 166.9, 152.9, 152.0, 138.8, 132.0, 129.9, 120.1, 116.8, 83.3, 81.2, 79.1, 75.2, 53.7, 48.9, 48.0, 41.0, 41.0 ,37.5, 34.9, 28.2, 24.6, 24.5, 23.5, 22.3, 22.1, 20.0, 18.8, 17.4, 14.7.

IR (film): ν 2958, 2930, 2873, 1795, 1739, 1696, 1456, 1386, 1367, 1342, 1303, 1253, 1228, 1122, 1092, 1038, 987, 978, 890, 842, 750.

HRMS (ESI): calculated for $C_{44}H_{74}N_2O_9SSi_2$ $[M+Na]^+$ 885.4546, found 885.4557.





2f

(4-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*R*,*E*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadec-13-en-3-yl)prop-1-en-1-yl)thiazol-2-

yl)methanaminium chloride(2f). To protected cyclopropyl-Epo B **152f** (3.0 mg, 0.03 mmol) was added HCl (1.0 M in EtOAc, 0.52 mL, 0.521 mmol) at room temperature. The reaction mixture was stirred for 15 h, a second portion of HCl (0.26 mL, 0.265 mmol) was then added and stirring was continued for further 25 h. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5) to yield 1.5 mg (85%) of **2f** as a colorless oil.

TLC: R_f 0.05 ($CHCl_3$/MeOH/H_2O/AcOH 90:10:1:0.5, UV, CPS).

$[\alpha]^{20}_D$: = -42.2° (*c* = 0.27, MeOH).

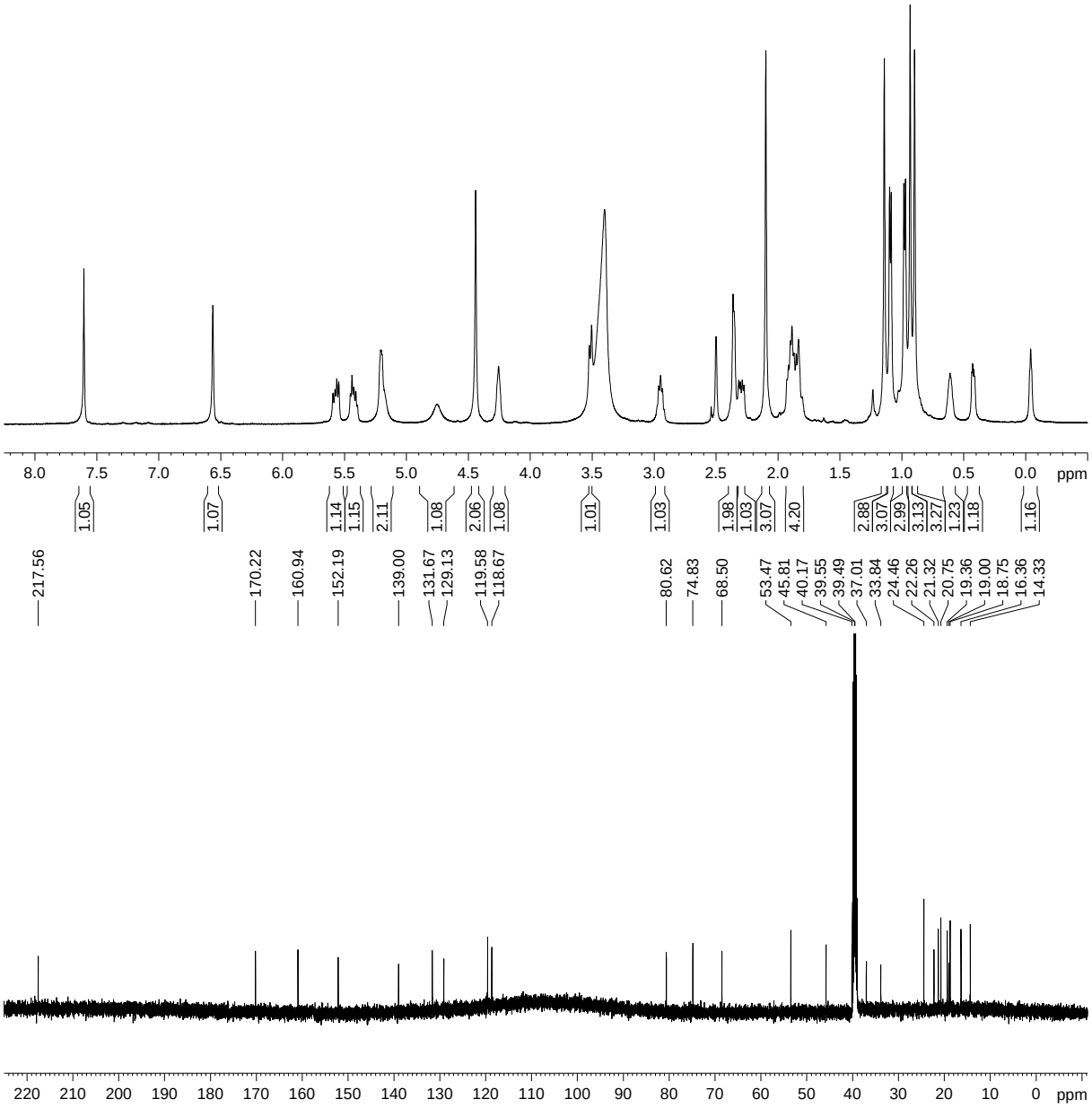
^{1}H-NMR (500 MHz, DMSO-d6): δ = 8.55 (s, 3H), 7.60 (s, 1H), 6.56 (s, 1H), 5.57 (dd, *J* = 15.6, 8.2 Hz, 1H), 5.42 (ddd, *J* = 15.6, 7.4, 6.6 Hz, 1H), 5.20 (dd, *J* = 7.2, 3.8 Hz, 1H), 5.17 (br s, 1H), 4.75 (br s, 1H), 4.44 (s, 2H), 4.26 (t, *J* = 5.8, 5.8 Hz, 1H), 3.51 (d, *J* = 9.6 Hz, 1H), 2.95 (dq, *J* = 9.6, 6.7 Hz, 1H), 2.37 (d, *J* = 5.8 Hz, 2H), 2.30 (dd, *J* = 14.6, 6.6 Hz, 1H), 2.10 (s, 3H), 1.91 (dd, *J* = 14.6, 6.6 Hz, 1H), 1.89 (dq, *J* = 8.2, 6.7 Hz. 1H), 1.84 (ddd, *J* = 15.1, 9.3, 7.9 Hz, 1H), 1.82 (ddd, *J* = 15.1, 3.8, 3.8 Hz, 1H), 1.14 (s, 3H), 1.09 (d, *J* = 6.7 Hz, 3H), 0.98 (d, *J* = 6.7 Hz, 3H), 0.93 (s, 3H), 0.90 (s, 3H), 0.61 (dddd, *J* = 8.7, 8.2, 5.1, 3.9 Hz, 1H), 0.42 (dd, *J* = 8.2, 3.5 Hz, 1H), 0.04 (dd, *J* = 5.1, 3.5 Hz, 1H).

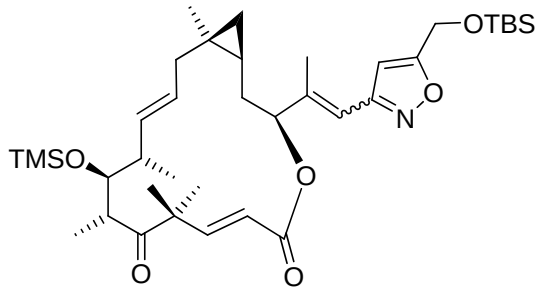
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.6, 170.2, 160.9, 152.2, 139.0, 131.7, 129.1, 119.6, 118.7, 80.6, 74.8, 68.5, 53.5, 45.8, 40.2, 39.5, 39.5, 37.0, 33.8, 24.5, 22.3, 21.3, 20.8, 19.4, 19.0, 18.7, 16.4 ,14.3.

IR (film): ν 3397, 2925, 1731, 1717, 1684, 1255, 1202, 1180, 1137, 1046, 1025, 1008, 984, 836, 800, 723.

HRMS (ESI): calculated for $C_{28}H_{43}N_2O_5S$ $[M+H]^+$ 519.2887, found 519.2882.

Analytical HPLC: Method: A and B with 0.1% TFA, eluent B 30-40% (linear gradient from 0-12 min), retention time 7.94 min.





151 deoxy b

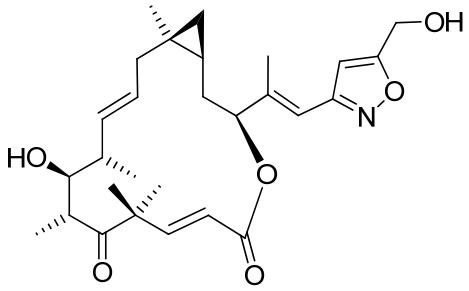
(1*S*,3*S*,6*E*,10*R*,11*S*,12*S*,13*E*,16*R*)-3-(1-(5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-11-(trimethylsilyloxy)-4-oxabicyclo[14.1.0]heptadeca-6,13-diene-5,9-dione (151 3deoxy b). To a solution of

phosphonate **140b** (39.3 mg, 0.108 mmol) in THF (1.0 mL) was added KHMDS (19.8 mg, 0.099 mmol) in one portion at 0 °C. The reaction mixture was stirred for 0.5 h at this temperature and was then cooled to -78 °C. Methyl ketone **148** (10.0 mg, 0.018 mmol) in THF (0.5 mL) was added dropwise and the reaction mixture was allowed to warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 25:1) to afford 9.5 mg (78%) of an inseparable 1:1 mixture of **151 deoxy b** and its C16-C17 *Z* isomer as a colorless oil.

TLC: R_f 0.40, 0.42 (hexane/EtOAc 9:1, UV, CPS).

IR (film): ν 2956, 2930, 2902, 2859, 1719, 1648, 1468, 1459, 1375, 1254, 1135, 1115, 1088, 1032, 984, 889, 837, 780.

HRMS (ESI): calculated for $C_{37}H_{62}NO_6Si_2$ $[M+H]^+$ 672.4110, found 672.4105.



2d

(1*S*,3*S*,6*E*,10*R*,11*S*,12*S*,13*E*,16*R*)-11-hydroxy-3-((*E*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadeca-6,13-diene-5,9-dione (2d). To a 1:1 mixture of protected cyclopropyl-Epo B **151 3deoxy b** and its C16-C17 *Z* isomer (9.5 mg, 0.012 mmol) in CH_2Cl_2 (0.67 mL) and MeOH (0.33 mL) was added CSA (2.9 mg, 0.012 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 18 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 2:1) to yield 3.3 mg (48%) of **2d** as a semisolid.

TLC: R_f 0.42 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_{D}$: = +78.9° (*c* = 1.03, $CHCl_3$).

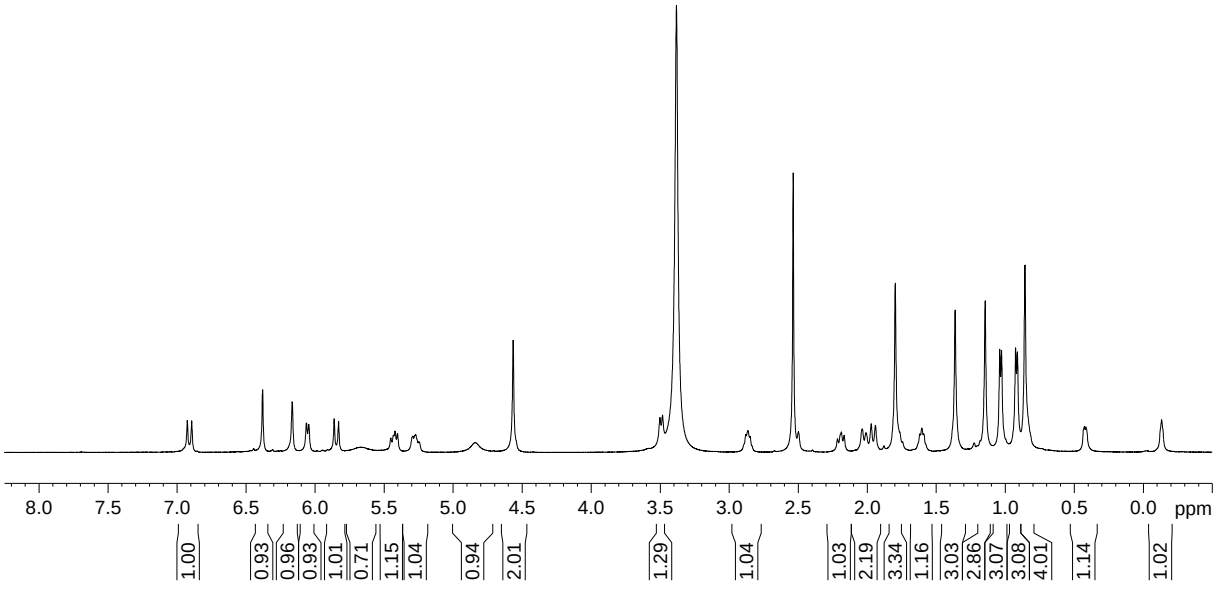
^{1}H-NMR (500 MHz, DMSO-d6): δ = 6.91 (d, *J* = 15.7 Hz, 1H), 6.38 (s, 1H), 6.17 (s, 1H), 6.06 (dd, *J* = 8.7, 2.2 Hz, 1H), 5.85 (d, *J* = 15.7 Hz, 1H), 5.66 (br s, 1H), 5.43 (dd, *J* = 15.5, 9.1 Hz, 1H), 5.27 (ddd, *J* = 15.5, 9.7, 3.4 Hz, 1H), 4.84 (br s, 1H), 4.57 (s, 2H), 3.49 (d, *J* = 9.4Hz, 1H), 2.87 (dq, *J* = 9.4, 6.8 Hz, 1H), 2.20 (dd, *J* = 14.6, 9.7 Hz, 1H), 2.03 (dd, *J* = 14.6, 3.4 Hz, 1H), 1.96 (ddd, *J* = 15.4, 2.4, 2.2 Hz, 1H), 1.80 (s, 3H), 1.78 (ddd, *J* = 15.4, 10.8, 8.7 Hz, 1H), 1.61 (dq, *J* = 9.1, 6.8 Hz, 1H), 1.37 (s, 3H), 1.15 (s, 3H), 1.04 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.8 Hz, 3H), 0.88-0.82 (m, 1H), 0.86 (s, 3H), 0.43 (dd, *J* = 8.8, 3.7 Hz, 1H), -0.13 (dd, *J* = 5.5, 3.7 Hz, 1H).

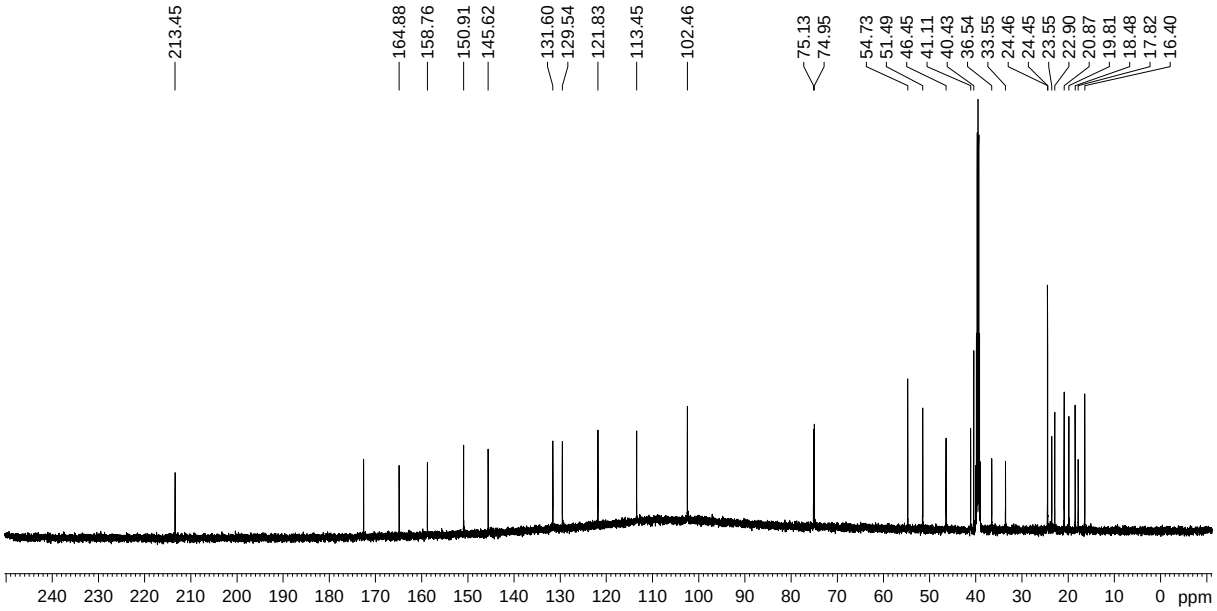
^{13}C-NMR (125 MHz, DMSO-d6): δ = 217.6, 172.5, 170.3, 158.8, 145.7, 131.5, 129.2, 113.2, 102.4, 74.9, 74.7, 68.6, 54.7, 53.4, 46.0, 40.1, 39.4, 36.9, 33.6, 24.3, 22.1, 22.0, 20.8, 19.5, 18.8, 18.6, 18.5, 16.3.

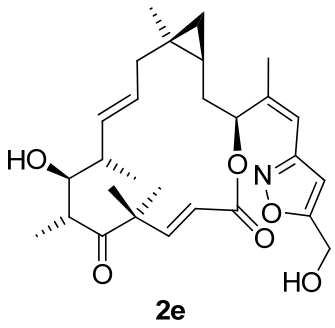
IR (film): ν 3394, 2961, 2926, 2871, 1702, 1646, 1604, 1451, 1368, 1291, 1263, 1179, 1151, 1052, 1026, 980, 962.

HRMS (ESI): calculated for $C_{28}H_{40}NO_6$ $[M+H]^+$ 486.2850, found 486.2851.

Analytical HPLC: Method: eluent B 48% (isokratisch), retention time 11.49 min.







2e

(1*S*,3*S*,6*E*,10*R*,11*S*,12*S*,13*E*,16*R*)-11-hydroxy-3-((*Z*)-1-(5-(hydroxymethyl)isoxazol-3-yl)prop-1-en-2-yl)-8,8,10,12,16-pentamethyl-4-oxabicyclo[14.1.0]heptadeca-6,13-diene-5,9-dione (2e). To a 1:1 mixture of protected cyclopropyl-Epo B **151 3deoxy b** and its C16-C17 *Z* isomer (9.5 mg, 0.012 mmol) in CH_2Cl_2 (0.67 mL) and MeOH (0.33 mL) was added CSA (2.9 mg, 0.012 mmol) at 0 °C. The reaction mixture was then allowed to reach room temperature and stirred for 18 h. The reaction mixture was then diluted with CH_2Cl_2 (2 mL) and the reaction was quenched by addition of saturated aqueous $NaHCO_3$ (2 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 2:1) to yield 3.4 mg (50%) of **2e** as a semisolid.

TLC: R_f 0.32 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = +46.5° (*c* = 0.33, $CHCl_3$).

^{1}H-NMR (500 MHz, DMSO-d6): δ = 6.94 (d, *J* = 15.8 Hz, 1H), 6.45 (s, 1H), 6.31 (q, *J* = 1.2 Hz, 1H), 5.93 (d, *J* = 15.8 Hz, 1H), 5.64 (br s, 1H), 5.44 (ddd, *J* = 15.5, 9.1, 1.6 Hz,

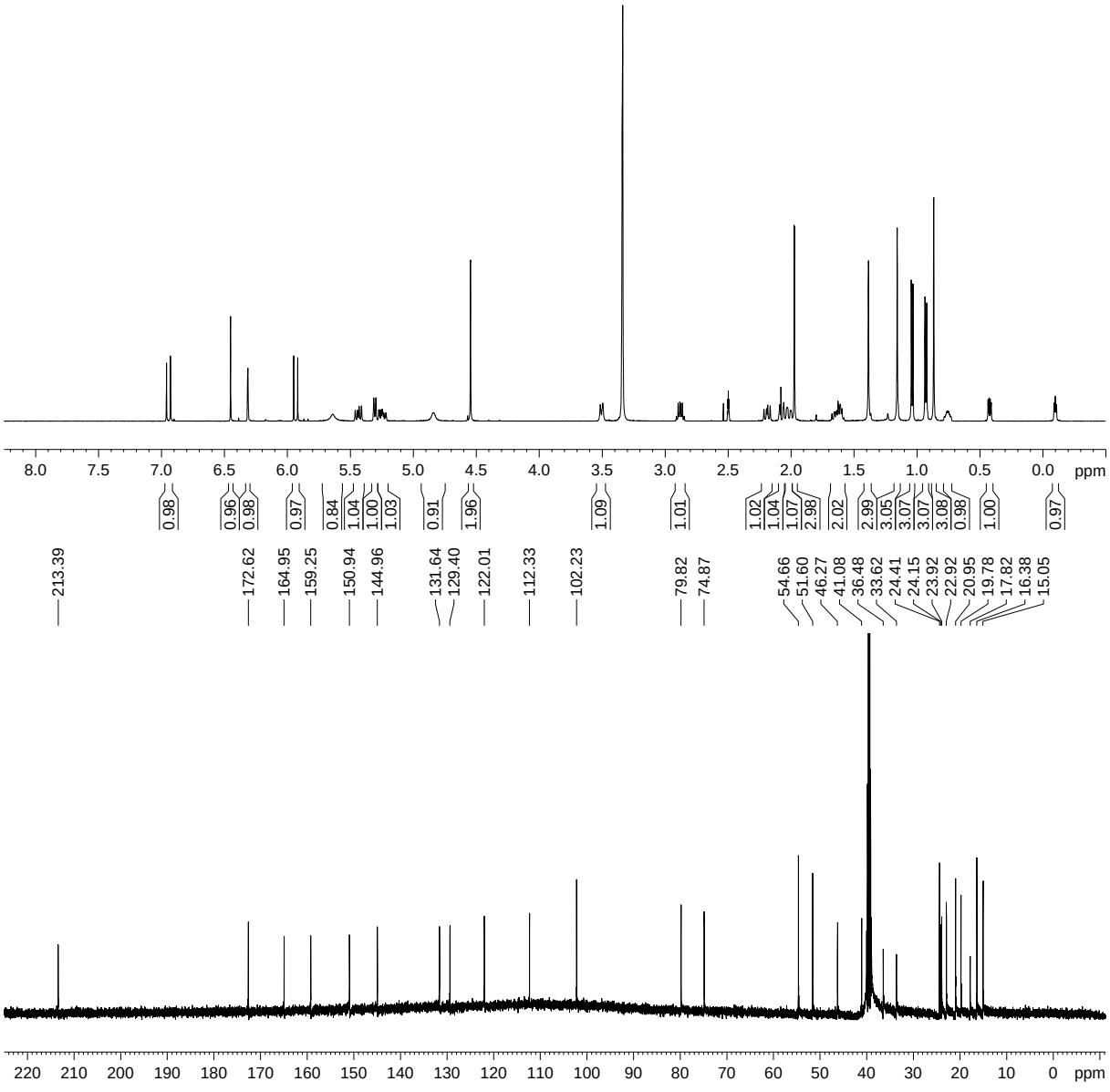
1H), 5.30 (dd, *J* = 8.5, 2.0 Hz, 1H), 5.24 (ddd, *J* = 15.5, 9.8, 3.8 Hz, 1H), 4.84 (br s, 1H), 4.55 (s, 2H), 3.51 (d, *J* = 9.9Hz, 1H), 2.88 (dq, *J* = 9.9, 6.9 Hz, 1H), 2.19 (dd, *J* = 14.7, 9.8 Hz, 1H), 2.08 (ddd, *J* = 15.7, 2.4, 2.0 Hz, 1H), 2.02 (ddd, *J* = 14.7, 3.8, 1.6 Hz, 1H), 1.98 (s, 3H), 1.65 (ddd, *J* = 15.7, 10.9, 8.5 Hz, 1H), 1.61 (dq, *J* = 9.1, 6.8 Hz, 1H), 1.39 (s, 3H), 1.16 (s, 3H), 1.04 (d, *J* = 6.9 Hz, 3H), 0.93 (d, *J* = 6.9 Hz, 3H), 0.87 (s, 3H), 0.76 (dddd, *J* = 10.9, 8.8, 5.6, 2.4 Hz, 1H), 0.42 (dd, *J* = 8.8, 3.9 Hz, 1H), 0.13 (dd, *J* = 5.6, 3.9 Hz, 1H).

^{13}C-NMR (125 MHz, DMSO-d6): δ = 213.4, 172.6, 164.9, 159.3, 150.9, 145.0, 131.6, 129.4, 122.0, 112.3, 102.2, 79.8, 74.9, 54.7, 51.6, 46.3, 41.1, 36.5, 33.6, 24.4, 24.2, 23.9, 22.9, 20.9, 19.8, 17.8, 16.4, 15.0.

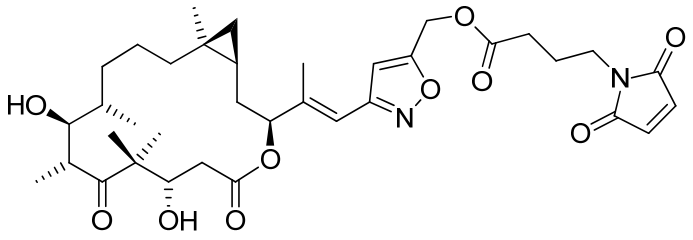
IR (film): ν 3425, 2967, 2926, 2873, 1705, 1643, 1602, 1450, 1373, 1277, 1178, 1151, 1057, 980, 756, 752.

HRMS (ESI): calculated for $C_{28}H_{39}NNaO_6$ $[M+Na]^+$ 508.2670, found 508.2662.

Analytical HPLC: Method: eluent B 48% (isokratisch), retention time 10.06 min.



4.2.4 Antibody Drug Conjugates



154

(3-((*E*)-2-((1*S*,3*S*,7*S*,10*R*,11*S*,12*S*,16*S*)-7,11-dihydroxy-8,8,10,12,16-pentamethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadecan-3-yl)prop-1-en-1-yl)isoxazol-5-yl)methyl 4-(2,5-dioxo-2,5-dihydro-1*H*-pyrrol-1-yl)butanoate (154). To a solution of 4-maleimidobutyric acid (22 mg, 0.123 mmol) in benzene (1.5 mL) was added Et_3N (34 µL, 2.45 mmol) and 2,4,6-trichlorobenzoyl chloride (9.5 µL, 0.064 mmol) at room temperature and the reaction mixture was stirred for 1 h. Alcohol **2A** (31 mg, 0.061 mmol) in THF (1.5 mL) and DMAP (30 mg, 0.245 mmol) in benzene (1 mL) were then added and the reaction mixture was stirred for 0.5 h at room temperature. The reaction mixture was directly purified by flash column chromatography (hexane/EtOAc 1:1) to afford 18 mg (44%) of ester **154** as a colorless oil.

Note: The ester **154** is not stable when an aqueous work-up is carried out. We in addition assume that the ester **154** is not stable to the reaction conditions and that the yield could therefore be improved by shorten the reaction time.

TLC: R_f 0.33 (hexane/EtOAc 1:1, UV, CPS).

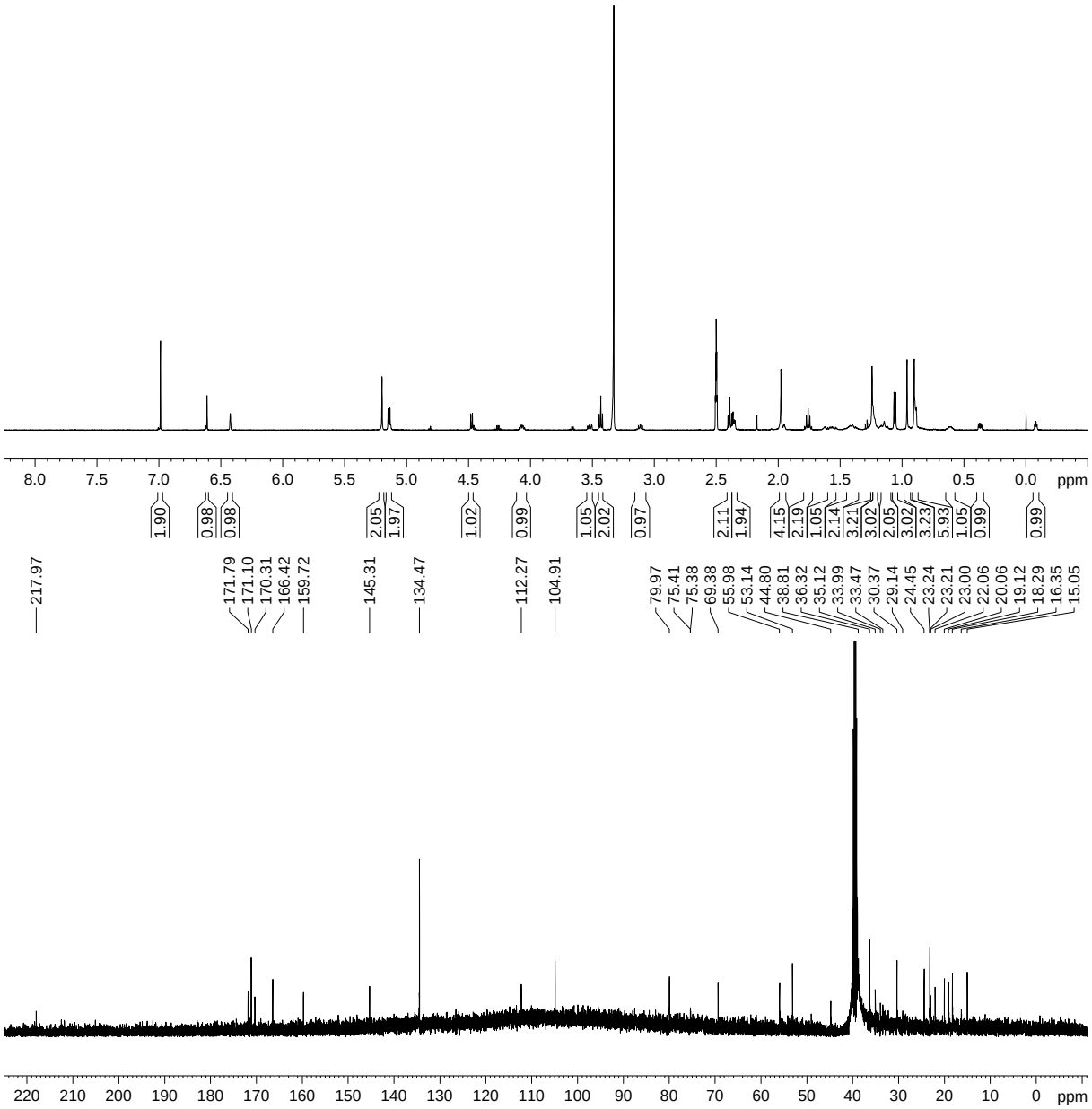
$[\alpha]^{20}_D$: = -8.4° (*c* = 0.40, $CHCl_3$).

^{1}H-NMR (500 MHz, DMSO-d_6): δ = 6.99 (s, 2H), 6.61 (s, 1H), 6.42 (q, *J* = 1.2 Hz, 1H), 5.20 (s, 2H), 5.14 (dd, *J* = 7.3, 2.8 Hz, 1H), 5.14 (d, *J* = 6.8 Hz, 1H), 4.47 (d, *J* = 6.6 Hz, 1H), 4.07 (ddd, *J* = 8.2, 7.2, 5.5 Hz, 1H), 3.52 (dd, *J* = 8.6, 6.8 Hz, 1H), 3.43 (t, *J* = 6.8 Hz, 2H), 3.11 (dq, *J* = 8.6, 6.8 Hz, 1H), 2.39 (t, *J* = 6.8 Hz, 2H), 2.36 (d, *J* = 5.5 Hz, 1H), 2.36 (d, *J* = 8.2 Hz, 1H), 1.98 (d, *J* = 1.2 Hz, 3H), 1.97 (ddd, *J* = 15.5, 3.1, 2.8 Hz, 1H), 1.76 (p, *J* = 6.8 Hz, 2H), 1.57 (ddd, *J* = 15.5, 10.5, 7.6 Hz, 1H), 1.45-1.35 (m, 2H), 1.24 (s, 3H), 1.24-1.20 (m, 3H), 1.17-1.10 (m, 2H), 1.06 (d, *J* = 6.7, 3H), 0.96 (s, 3H), 0.90 (s, 3H), 0.89 (d, *J* = 6.8 Hz, 3H), 0.61 (dddd, *J* =10.7, 8.6, 5.5, 3.1 Hz, 1H), 0.37 (dd, *J* = 8.6, 3.9 Hz, 1H), -0.08 (dd, *J* = 5.5, 3.9 Hz, 1H).

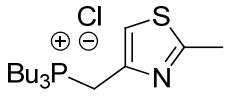
^{13}C-NMR (125 MHz, DMSO-d6): δ = 218.0, 171.8, 171.1, 170.3, 166.4, 159.7, 145.3, 134.5, 112.3, 104.9, 80.0, 75.4, 75.4, 69.4, 56.0, 53.1, 44.8, 38.8, 36.3, 35.1, 34.0, 33.5, 30.4, 29.1, 24.4, 23.2, 23.2, 23.0, 22.1, 20.1, 19.1, 18.3, 16.3, 15.0.

IR (film): ν 3442, 2928, 1736, 1708, 1453, 1410, 1377, 1259, 1171, 1143, 1022, 822, 808, 798, 788, 782, 740, 695, 674, 608.

HRMS (ESI): calculated for $C_{36}H_{50}N_2NaO_{10}$ $[M+Na]^+$ 633.3358, found 633.3360.



4.2.5 Synthesis of Unfunctionalized Heterocycles 78a-e



78a

Tributyl((2-methylthiazol-4-yl)methyl)phosphonium chloride (78a). 78a could be accessed as previously described in the literature.

TLC: R_f 0.25 (CH_2Cl_2 /MeOH 10:1, CPS).

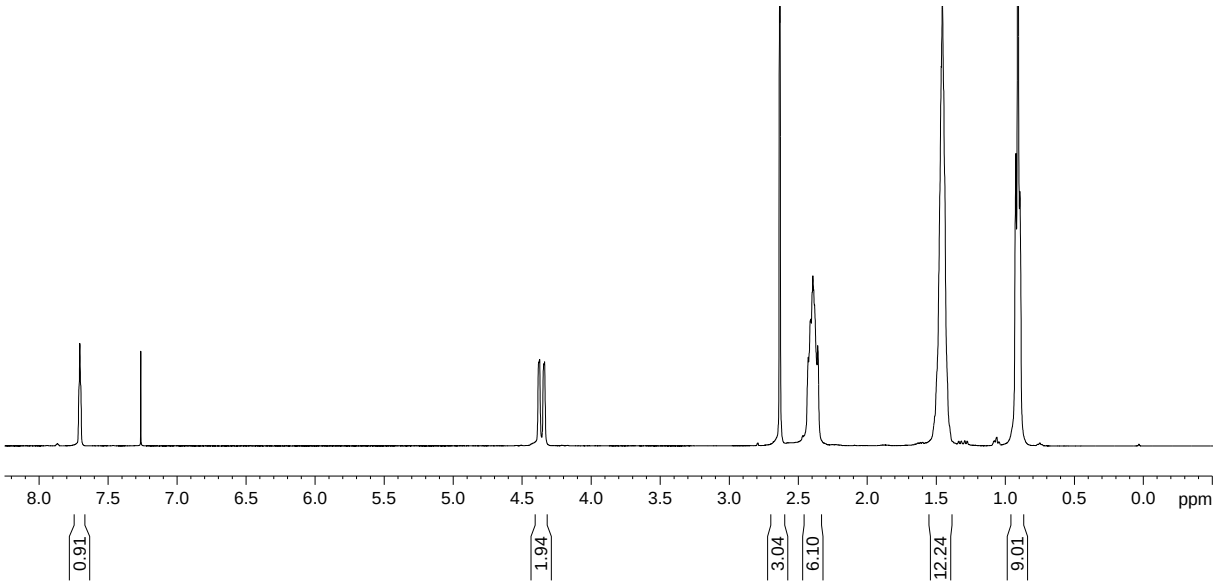
mp: 90-92 °C.

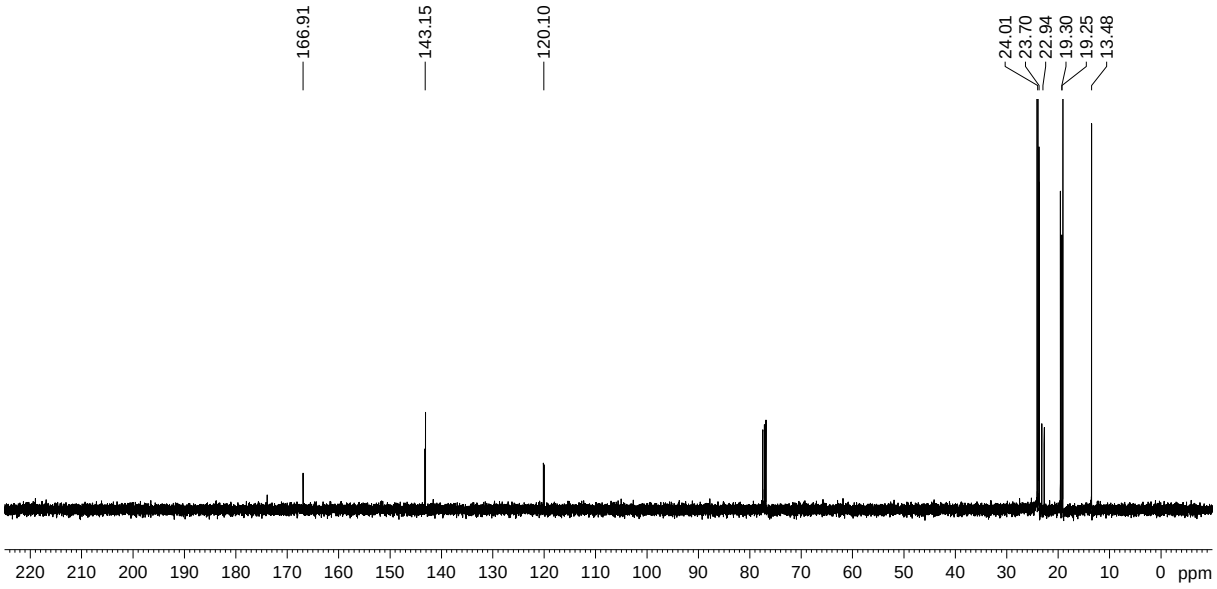
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.70 (t, *J* = 2.8 Hz, 1H), 4.36 (d, *J* = 14.5 Hz, 2H), 2.63 (d, *J* = 1.6 Hz, 3H), 2.44-2.33 (m, 6H), 1.53-1.39 (m, 12H), 0.96-0.87 (m, 9H).

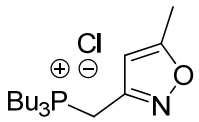
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 166.9 (d, *J* = 1.5 Hz), 143.2 (d, *J* = 9.7 Hz), 120.1 (d, *J* = 9.1 Hz), 24.0 (d, *J* = 15.2 Hz), 23.7 (d, *J* = 4.8 Hz), 22.9 (d, *J* = 48.3 Hz), 19.3 (d, *J* = 46.7 Hz), 19.2, 13.5.

IR (film): ν 2958, 2930, 2871, 1631, 1518, 1462, 1402, 1381, 1319, 1234, 1186, 1234, 1186, 1096, 1006, 955, 914, 805, 718.

HRMS (ESI): calculated for $C_{17}H_{33}NPS$ $[M+]^+$ 314.2066, found 314.2077.







78b

Tributyl((5-methylisoxazol-3-yl)methyl)phosphonium chloride (78b). To 3-(chloromethyl)-5-methylisoxazole (**156**)(100 mg, 0.76 mmol) in DMF (4 mL) was added Bu_3P (0.32 mL, 1.29 mmol) at room temperature. The mixture was left stirring for 24 h, the solvent was concentrated under reduced pressure and the residue was purified by column chromatography (CH_2Cl_2 /MeOH 20:1 → 10:1) to give 212 mg (84%) of phosphonium salt **78b** as a white solid.

TLC: R_f 0.48 (CH_2Cl_2 /MeOH 10:1, CPS).

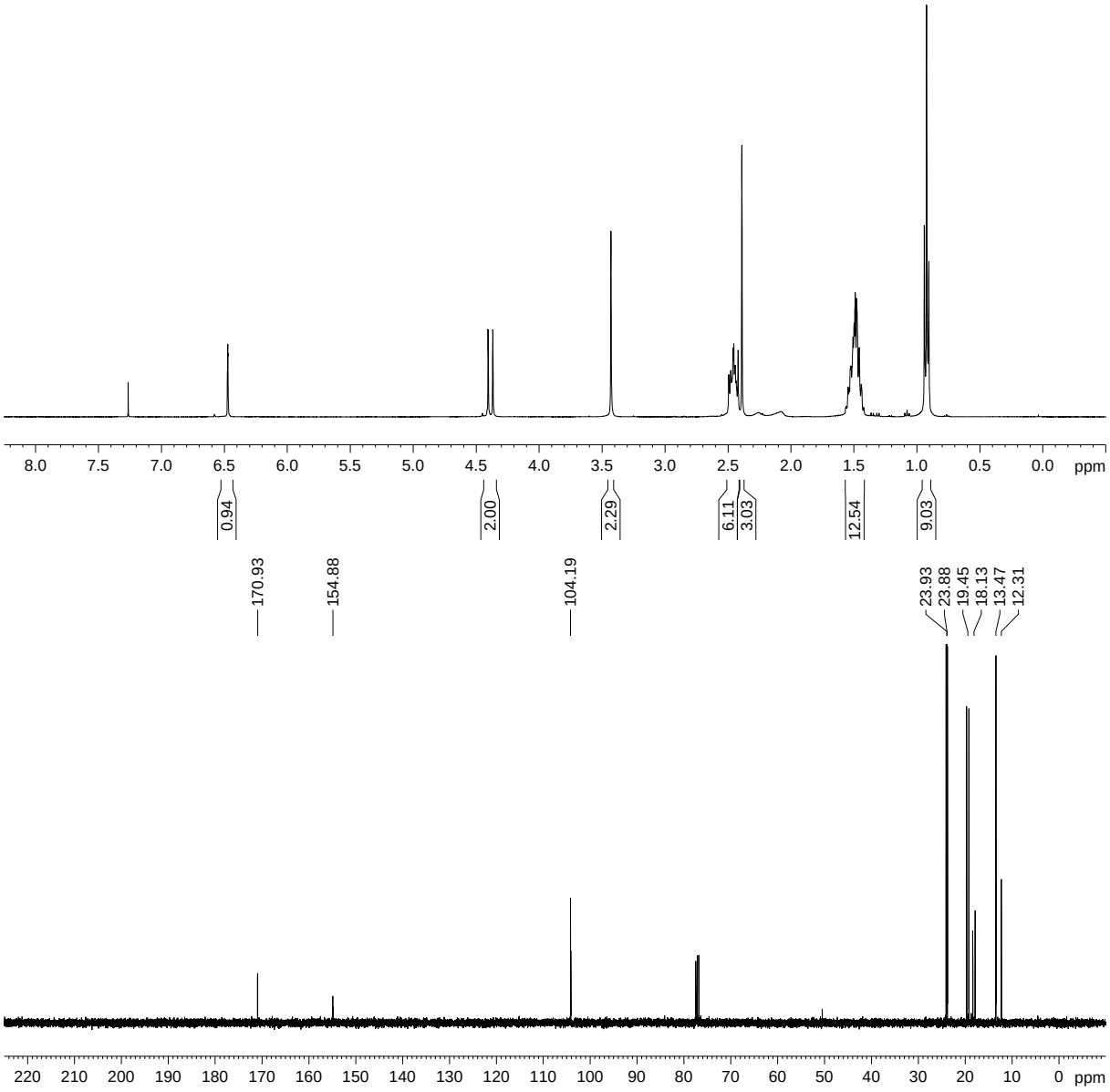
mp: 78-82 °C.

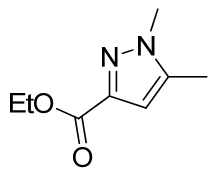
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.48 (s, 1H), 4.42 (d, *J* = 15.0 Hz, 2H), 3.44 (s, 2H), 2.51-2.42 (m, 6H), 2.39 (s, 3), 1.57-1.42 (m, 12H), 0.93 (t, *J* = 7.0 Hz, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.9, 154.9 (d, *J* = 8.1 Hz), 104.2 (d, *J* = 5.0), 23.9 (d, *J* = 28.8 Hz), 23.9 (d, *J* = 8.4 Hz), 19.5 (d, *J* = 46.9 Hz), 18.1 (d, *J* = 49.3 Hz), 13.5, 12.3.

IR (film): ν 2959, 2931, 2871, 1604, 1457, 1425, 1236, 1096, 1004, 913, 833, 723, 680.

HRMS (ESI): calculated for $C_{17}H_{33}NPS$ $[M+]^+$ 298.2294, found 298.2302.





159

Ethyl 1,5-dimethyl-1*H*-pyrazole-3-carboxylate (159). To a solution of ethyl acetopyruvate (1.78 mL, 12.65 mmol) in EtOH (100 mL) was added methyl hydrazine (1.33 mL, 25.29 mmol) dropwise at 0 °C and the reaction mixture was stirred for 1.5 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (50 mL), the phases were separated and the aqueous phase was extracted with EtOAc (3 x 60 mL). The combined organic phases were dried over $MgSO_4$, concentrated under

reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1 → 1:1) to yield 0.65 g (32%) of pyrazole **159** as a colorless oil.

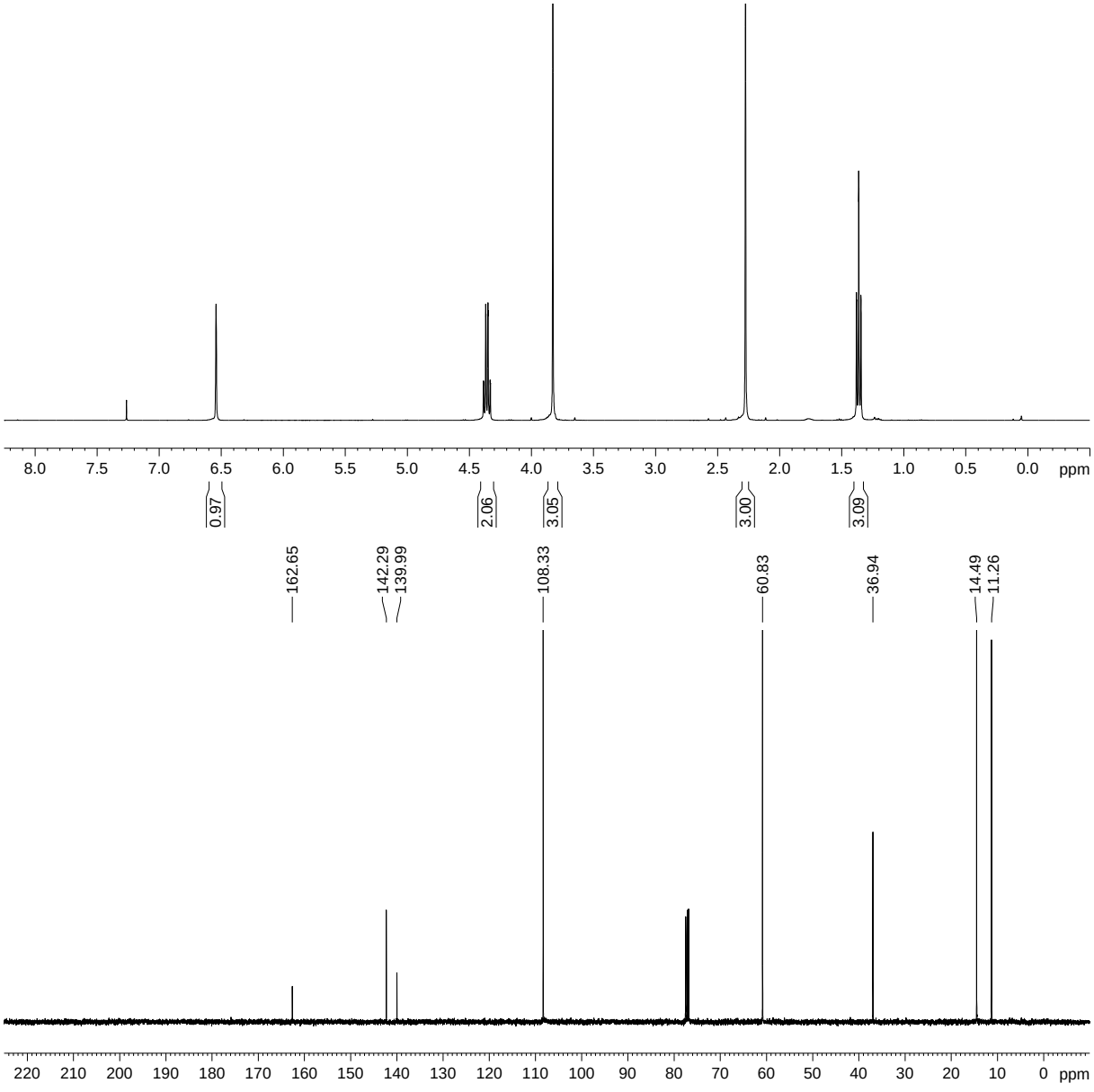
TLC: R_f 0.23 (hexane/EtOAc 1:1, UV, $KMnO_4$).

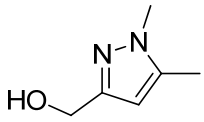
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.54 (q, *J* = 0.9 Hz, 1H), 4.36 (qd, *J* = 7.1, 0.9 Hz, 2H), 3.83 (d, *J* = 0.9 Hz, 3H), 2.27 (d, *J* = 0.9 Hz, 3H), 1.36 (td, *J* = 7.1, 0.9 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.7, 142.3, 140.0, 108.3, 60.8, 36.9, 14.5, 11.3;

IR (film): ν □2982, 2941, 1713, 1650, 1555, 1453, 1385, 1295, 1216, 1180, 1105, 1046, 1027, 779, 642.

HRMS (ESI): calculated for $C_8H_{12}N_2O_2$ $[M+H]^+$ 169.0972, found 169.0976.





161

(1,5-dimethyl-1*H*-pyrazol-3-yl)methanol (161). To a solution of ester **159** (0.72 g, 4.28 mmol) in CH_2Cl_2 (30 mL) was added DIBAL-H (1 M in CH_2Cl_2, 9.42 mL, 9.42 mmol) dropwise at -78 °C. The reaction mixture was stirred for 2 h at this temperature, was then allowed to warm to 0 °C slowly and stirred for 2 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (50 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases got transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc/MeOH 20:1) to give 0.39 g (72%) of alcohol **161** as a white solid.

TLC: R_f 0.24 (EtOAc/MeOH 20:1, $KMnO_4$).

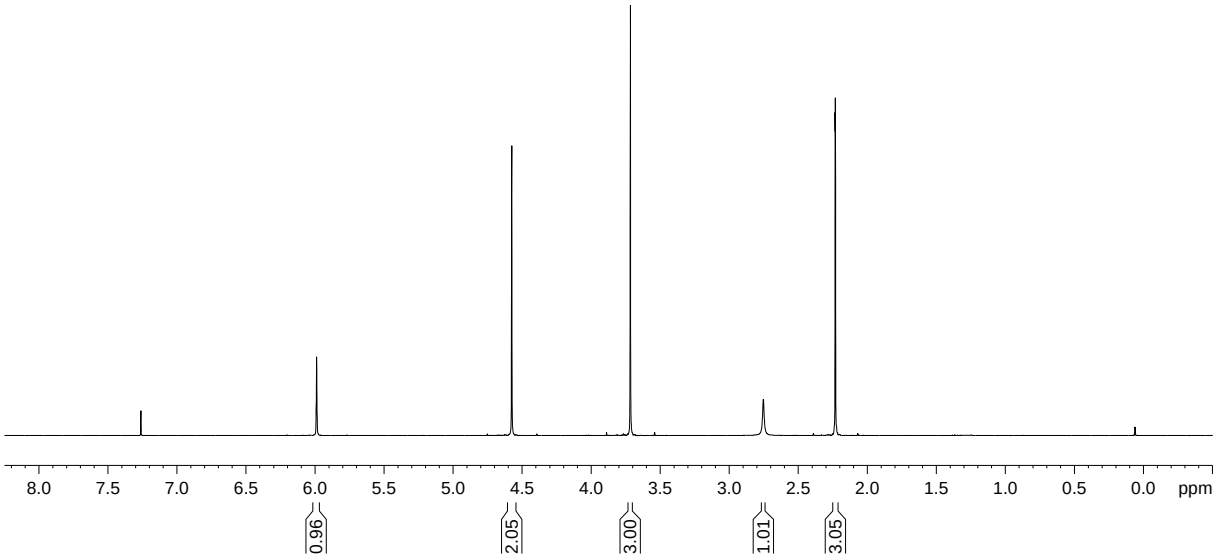
mp: 56-58 °C.

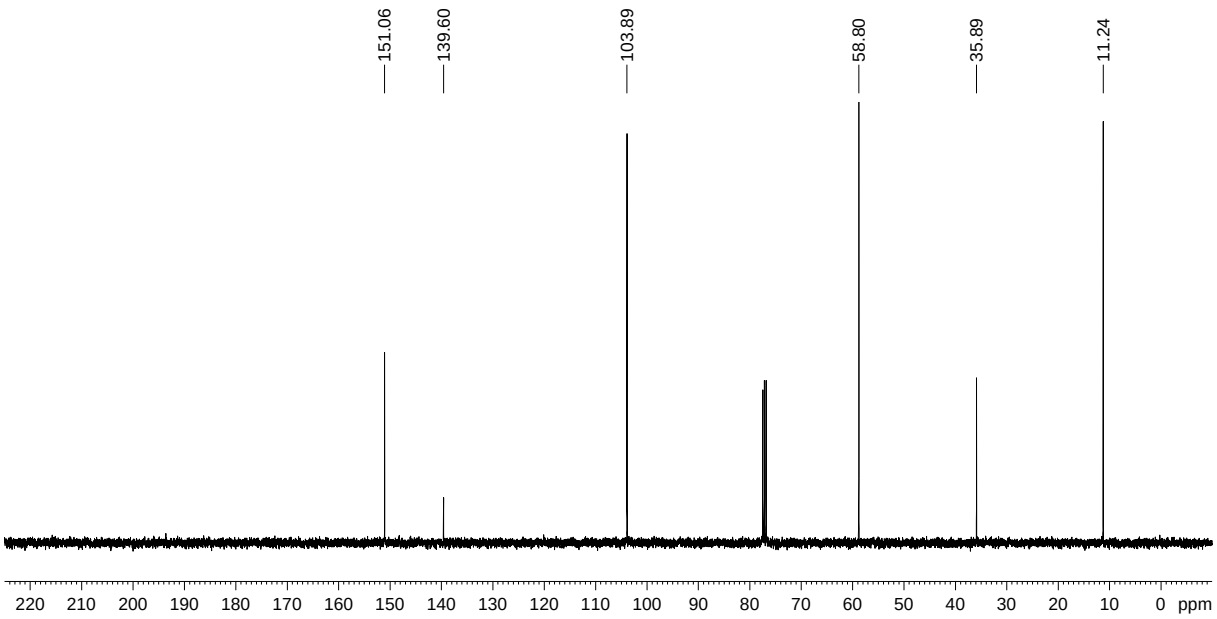
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.99 (d, *J* = 0.6 Hz, 1H), 4.58 (s, 2H), 3.72 (s, 3H), 2.75 (br s, 1H), 2.23 (d, *J* = 0.6 Hz, 3H).

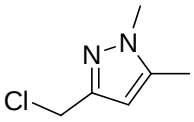
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 151.1, 139.6, 103.9, 58.8, 35.9, 11.2.

IR (film): ν □3301, 2934, 2869, 1551, 1490, 1431, 1389, 1282, 1220, 1141, 1006, 790, 650.

HRMS (EI): calculated for $C_6H_{10}N_2O$ $[M]^+$ 126.0788, found 126.0788.







162

3-(chloromethyl)-1,5-dimethyl-1*H*-pyrazole (162). To a solution of alcohol **161** (3.2 g, 2.50 mmol) in CH_2Cl_2 (25 mL) was added thionyl chloride (0.23 mL, 3.12 mmol) dropwise at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 2 h at this temperature. The reaction mixture was then poured into an ice-cold solution of saturated aqueous $NaHCO_3$ (20 mL) and CH_2Cl_2 (20 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 15 mL). The combined organic phases were washed with water (1 x 50 mL), dried over $MgSO_4$ and concentrated under reduced pressure to give 0.36 g (99%) of chloride **162** as a colorless liquid.

Note: Chloride **162** is volatile. The solvent was evaporated at a pressure of 240 mbar and a water bath temperature of 36 °C.

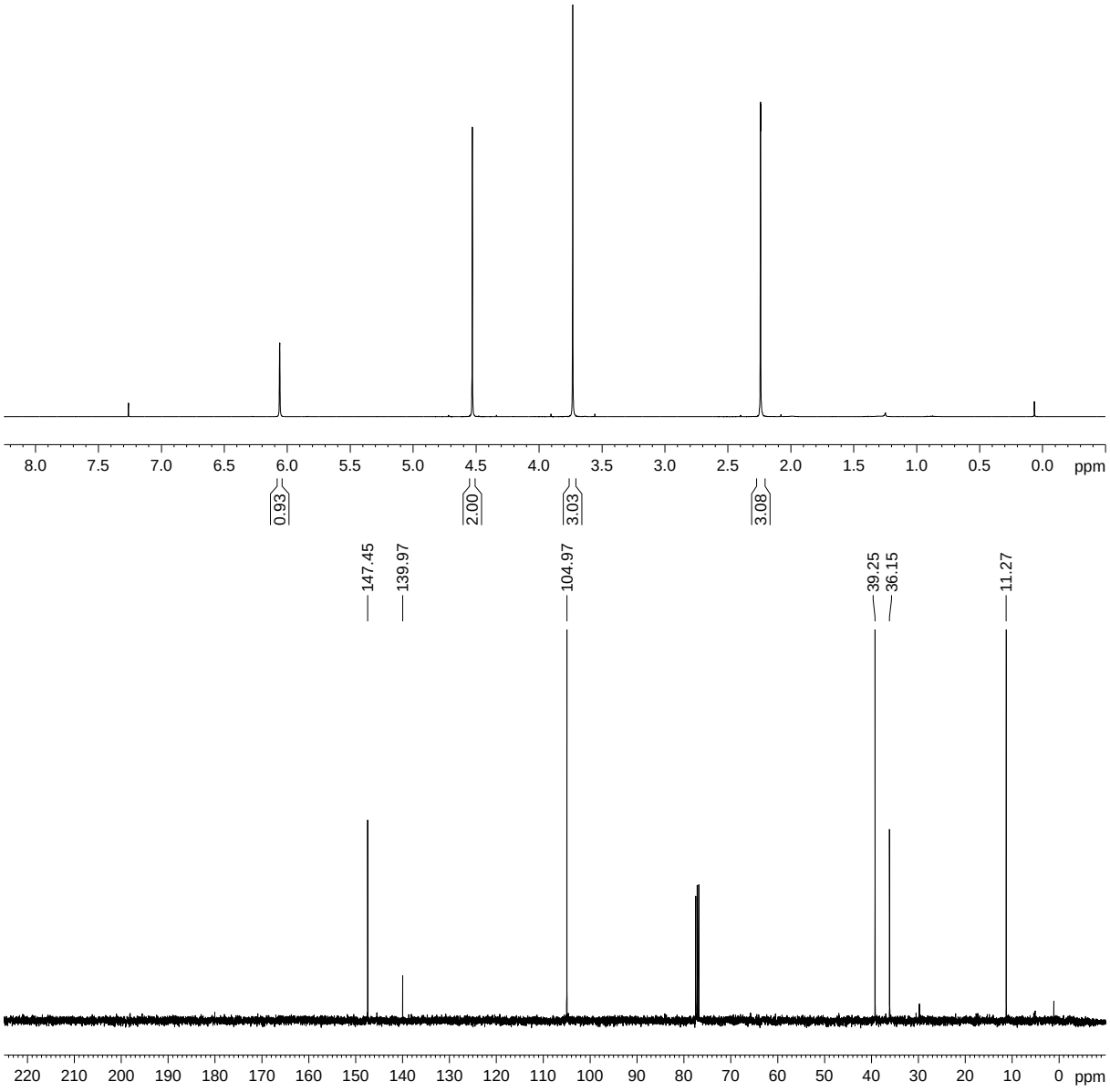
TLC: R_f 0.34 (hexane/EtOAc 2:1, UV, $KMnO_4$).

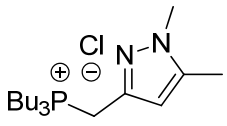
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.06 (d, *J* = 0.5, 1H), 4.53 (s, 2H), 3.73 (s, 3H), 2.24 (d, *J* = 0.5 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 147.4, 140.0, 105.0, 39.3, 36.1, 11.3.

IR (film): ν □2928, 2856, 1553, 1486, 1453, 1432, 1289, 1258, 1154, 1132, 982, 798, 733, 672, 640.

HRMS (EI): calculated for $C_6H_9ClN_2$ $[M]^+$ 144.0449, found 144.0452.





78c

Tributyl((1,5-dimethyl-1*H*-pyrazol-3-yl)methyl)phosphonium chloride (78c). To a solution of chloride **162** (0.23 g, 1.59 mmol) in DMF (12 mL) was added Bu_3P (0.68 mL, 2.70 mmol) at room temperature and the reaction mixture was left stirring for 13 h at this temperature. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1 → 10:1) to give 0.45 g (82%) of Wittig salt **78c** as a colorless oil which turns into a white solid upon storage at -20 °C.

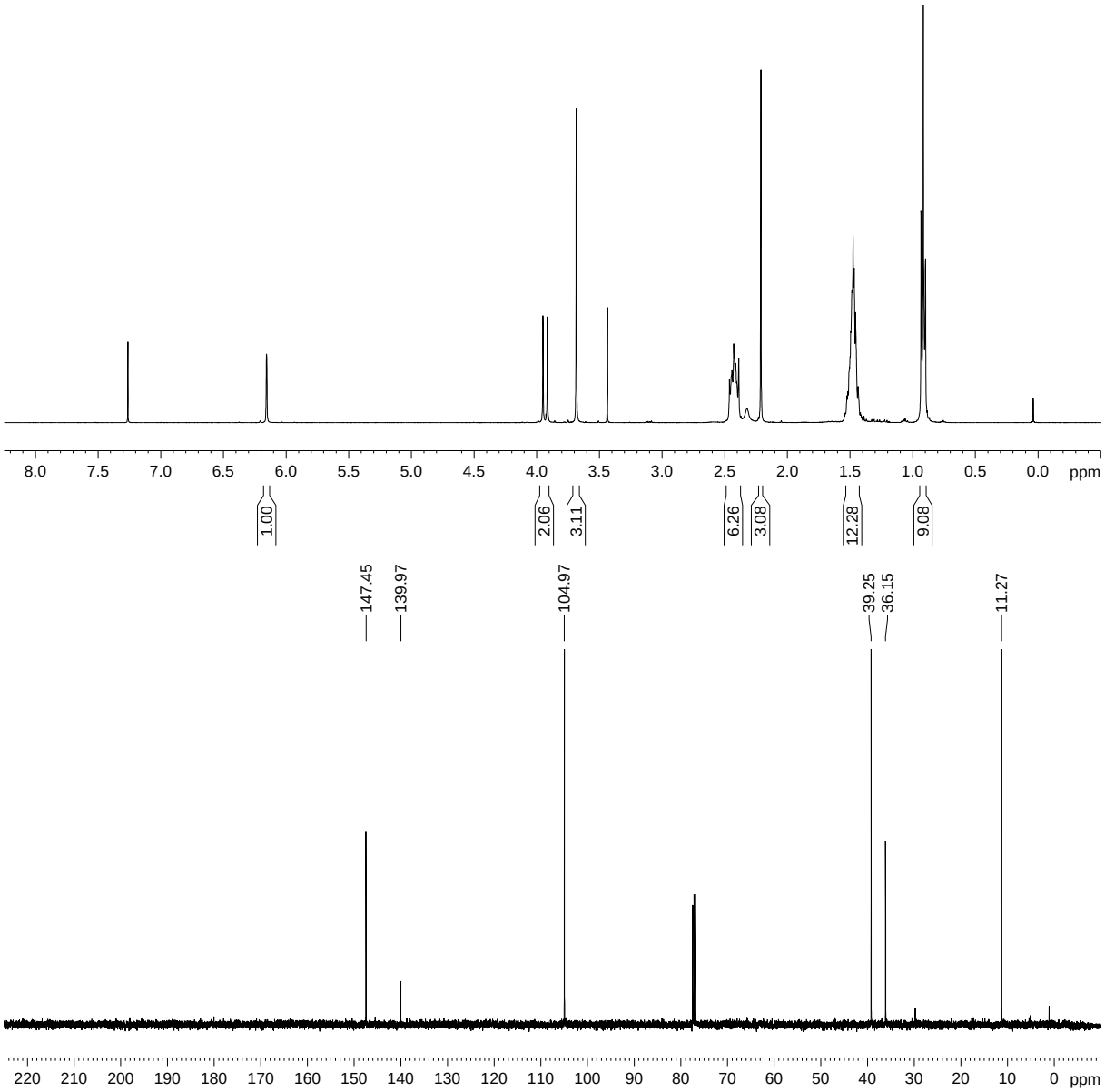
TLC: R_f 0.25 (CH_2Cl_2 /MeOH 10:1, CPS).

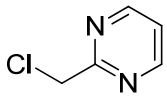
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.16 (s, 1H), 3.93 (d, *J* = 14.1 Hz, 2H), 3.69 (d, *J* = 0.8 Hz, 3H), 2.47-2.39 (m, 6H), 2.21 (s, 3H), 1.54-1.42 (m, 12H), 0.92 (t, *J* = 7.1 Hz, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 140.3, 138.8 (d, *J* = 8.2 Hz), 107.2 (d, *J* = 5.4 Hz), 36.3, 24.0 (d, *J* = 26.3 Hz), 23.9 (d, *J* = 16.0 Hz), 19.8 (d, *J* = 48.4 Hz), 19.2 (d, *J* = 47.1 Hz), 13.5, 11.2.

IR (film): ν 2957, 2931, 2871, 1548, 1456, 1384, 1284, 1235, 1097, 1015, 913, 822, 794, 721, 642.

HRMS (ESI): calculated for $C_{18}H_{36}ClN_2P$ $[M+H]^+$ 311.2611, found 311.2635.





164

2-(chloromethyl)pyrimidine (164). Chloroacetamidine (**163**) (2.13 g, 16.47 mmol) and tetramethoxypropane (5.42 mL, 32.95 mmol) were heated up to 100 °C and stirred for 16 h at this temperature. The reaction mixture was then allowed to reach room temperature and water (20 mL) was added. The phases were separated and the aqueous phase was then extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (pentane/Et_2O 3:2) to yield 0.73 g (34%) of chloride **164** as a colorless liquid.

Note: Chloride **K** is volatile. The solvent was evaporated at a pressure of 820 mbar and a water bath temperature of 36 °C.

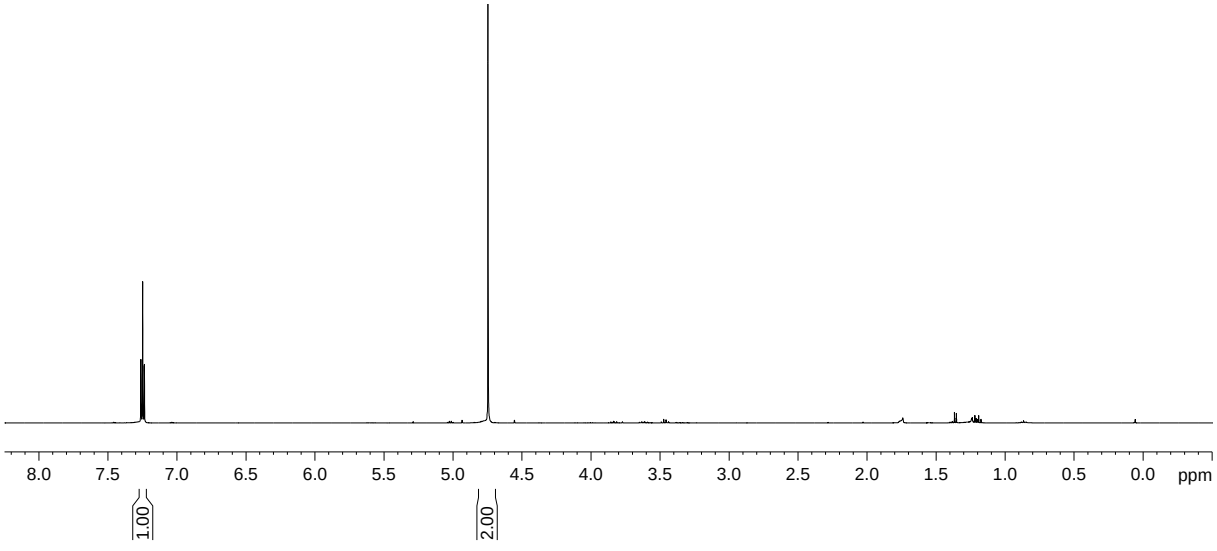
TLC: R_f 0.23 (hexane/EtOAc 2:1, UV, CPS).

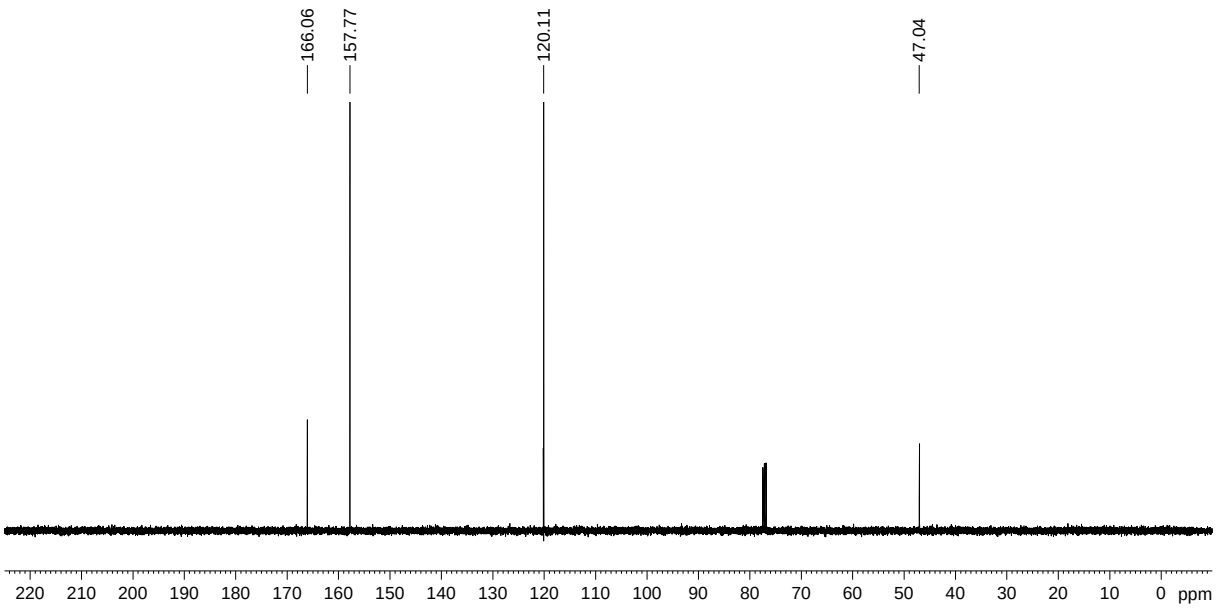
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.77 (d, *J* = 4.9 Hz, 2H), 7.25 (t, *J* = 4.9 Hz, 1H), 4.75 (s, 2H).

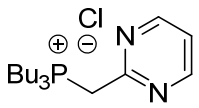
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 166.1, 157.8, 120.1, 47.0.

IR (film): ν □3043, 2975, 1565, 14224, 1305, 1241, 998, 851, 812, 702, 630, 607.

HRMS (EI): calculated for $C_5H_5ClN_2$ $[M]^+$ 128.0136, found 128.0136.







78d

Tributyl(pyrimidin-2-ylmethyl)phosphonium chloride (78d). To a solution of chloride **164** (0.14 g, 1.10 mmol) in DMF (7 mL) was added Bu_3P (0.47 mL, 1.86 mmol) at room temperature and the reaction mixture was left stirring for 24 h at this temperature. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1 → 10:1) to afford 0.33 g (91%) of Wittig salt **78d** as a slightly yellow solid.

TLC: R_f 0.39 (CH_2Cl_2 /MeOH 10:1, UV, $KMnO_4$).

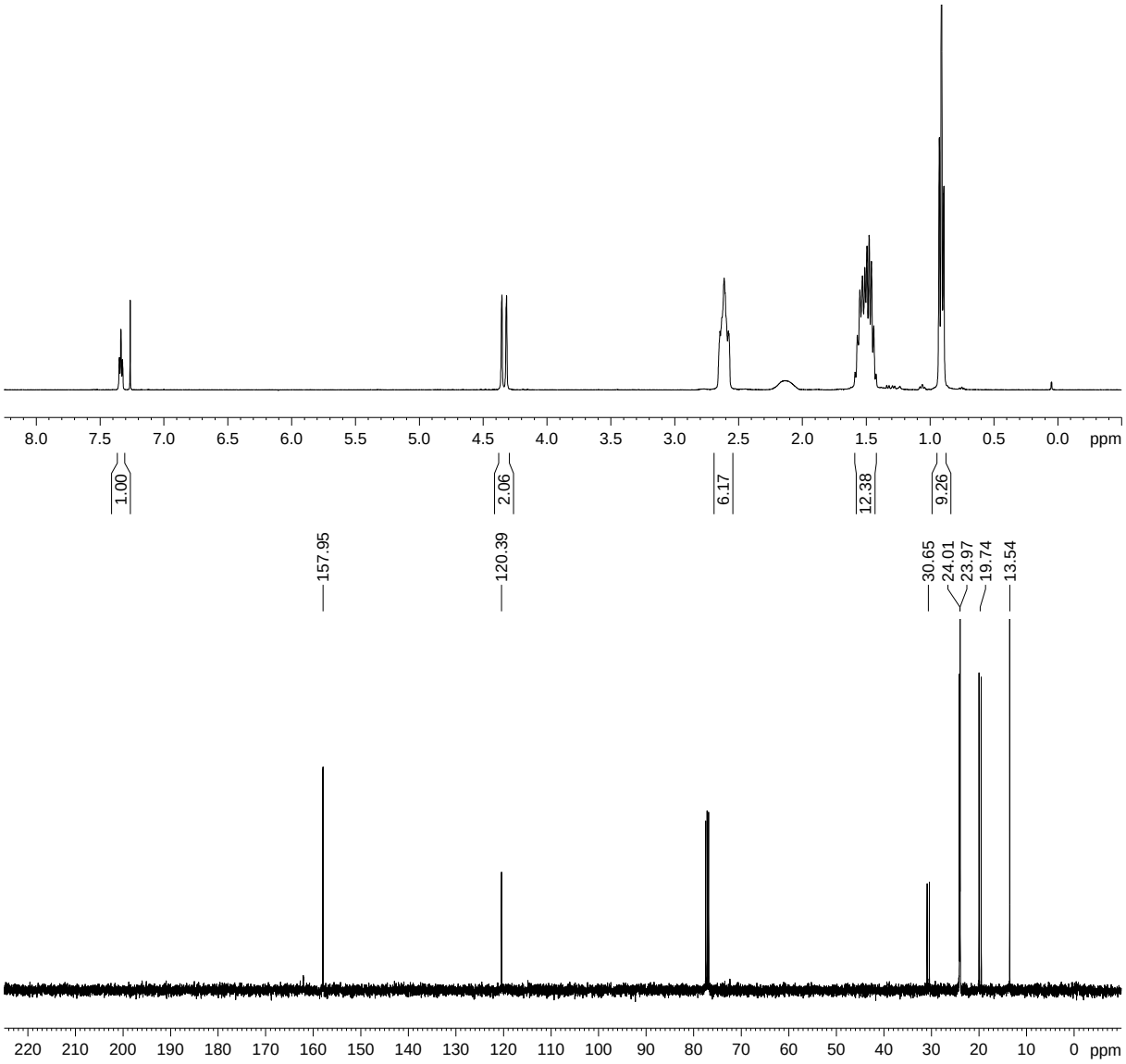
mp: 107-110 °C.

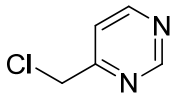
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.71 (d, *J* = 4.9 Hz, 2H), 7.34 (t, *J* = 4.9 Hz, 1H), 4.33 (d, *J* = 14.6 Hz, 2H), 2.65-2.56 (m, 6H), 1.59-1.41 (m, 12H), 0.90 (t, *J* = 7.1 Hz, 9H).

^{13}C-NMR (125 MHz, $CDCl_3$): δ = 158.0, 120.4, 30.6 (d, *J* = 48.0 Hz), 24.0 (d, *J* = 15.6 Hz), 24.0 (d, *J* = 4.8 Hz), 19.7 (d, *J* = 47.2 Hz), 13.5.

IR (film): ν □2959, 2931, 2871, 1564, 1460, 1416, 1235, 1096, 915, 849, 719, 635.

HRMS (ESI): calculated for $C_{17}H_{32}ClN_2P$ $[M+H]^+$ 295.2298, found 295.2307.





166

4-(chloromethyl)pyrimidine (166). To a solution of 4-methylpyrimidine (**165**) (2.00 g, 21.25 mmol) in $CHCl_3$ (30 mL) was added trichloroisocyanuric acid (1.98 g, 8.50 mmol) in one portion at room temperature and the reaction mixture was heated to reflux for 9 h. The reaction mixture was then allowed to reach room temperature and filtered through a small plug of celite. The precipitate was rinsed with CH_2Cl_2 (30 mL) and the filtrate was washed with aqueous 1 M NaOH (1 x 50 mL) and brine (1 x 50 mL). The organic phase was dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash

column chromatography (hexane/EtOAc 2:1) to yield 1.30 g (48%) of chloride **166** as a redish liquid.

Note: 13% of 4-(dichloromethyl)pyrimidine and 10% starting material were isolated.

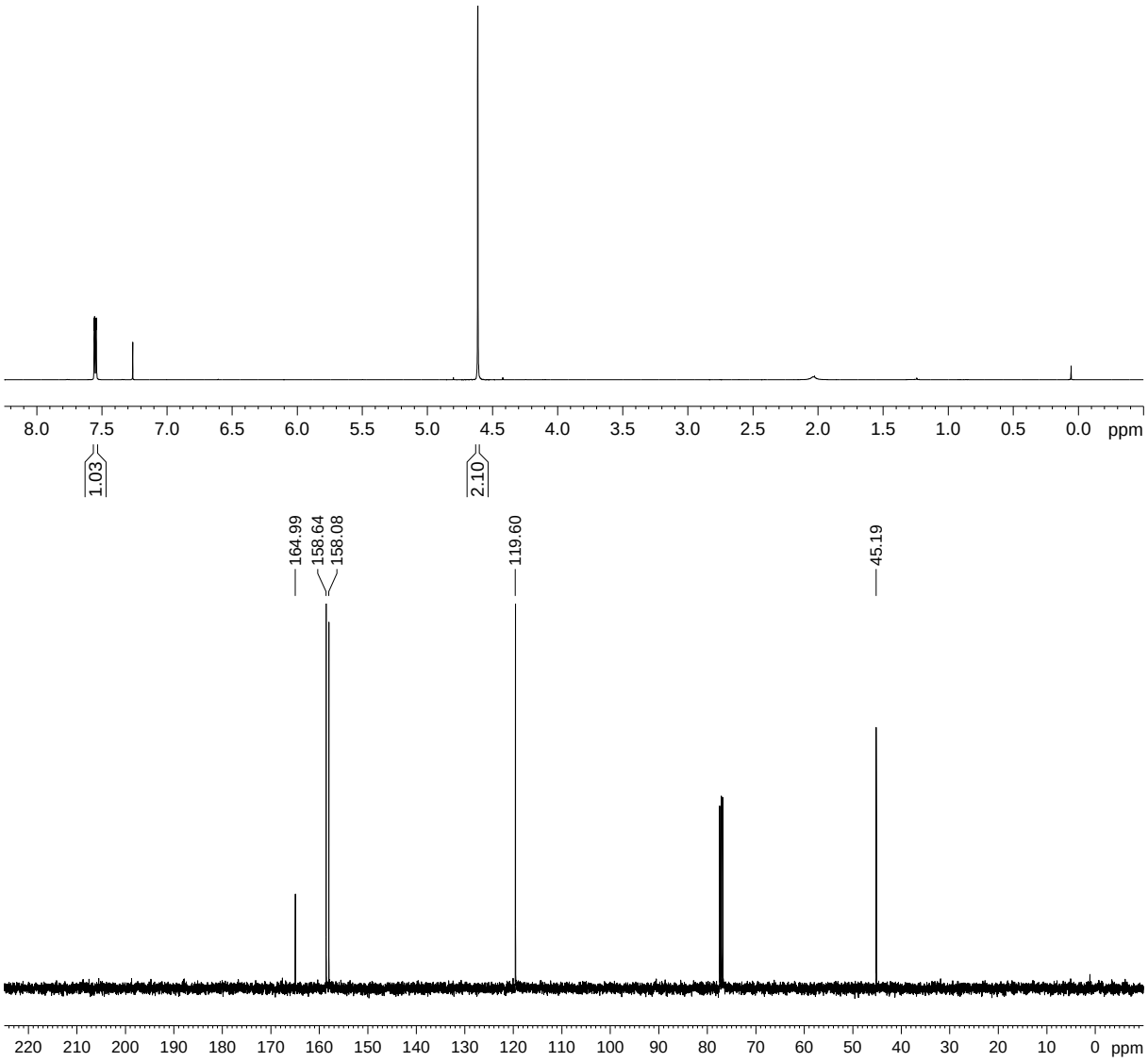
TLC: R_f 0.20 (hexane/EtOAc 2:1, UV, CPS).

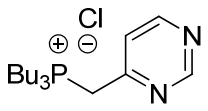
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.17 (d, *J* = 1.3 Hz, 1H), 8.78 (d, *J* = 5.2 Hz, 1H), 7.55 (dm, *J* = 5.2, 1H), 4.61 (s, 2H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 165.0, 158.6, 158.1, 119.6, 45.2.

IR (film): ν □3046, 2952, 1577, 1471, 1387, 1309, 1239, 1156, 993, 925, 840, 707, 681, 579, 486.

HRMS (EI): calculated for $C_5H_5ClN_2$ $[M]^+$ 128.0136, found 128.0139.





78e

Tributyl(pyrimidin-4-ylmethyl)phosphonium chloride (78e). To a solution of chloride **166** (87mg, 0.68 mmol) in DMF (5 mL) was added Bu_3P (0.29 mL, 1.15 mmol) at room temperature and the reaction mixture was left stirring for 23 h at this temperature. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1 →10:1) to afford 220 mg (98%) of Wittig salt **78e** as a slightly brownish solid.

TLC: R_f 0.40 (CH_2Cl_2 /MeOH 10:1, UV, $KMnO_4$).

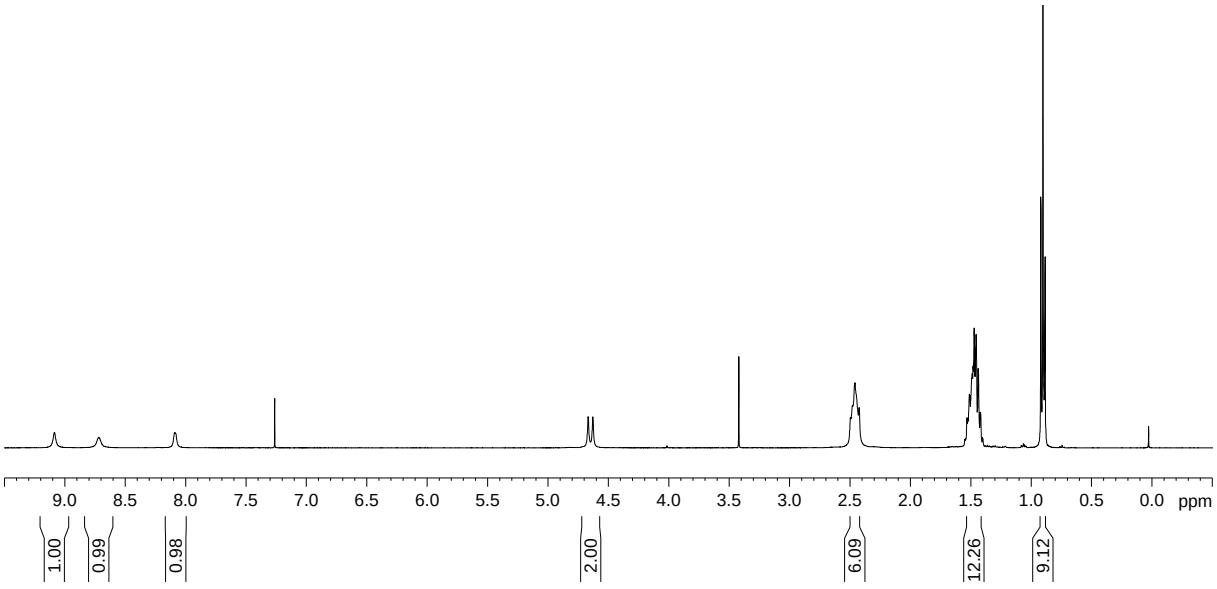
mp: 108-110 °C.

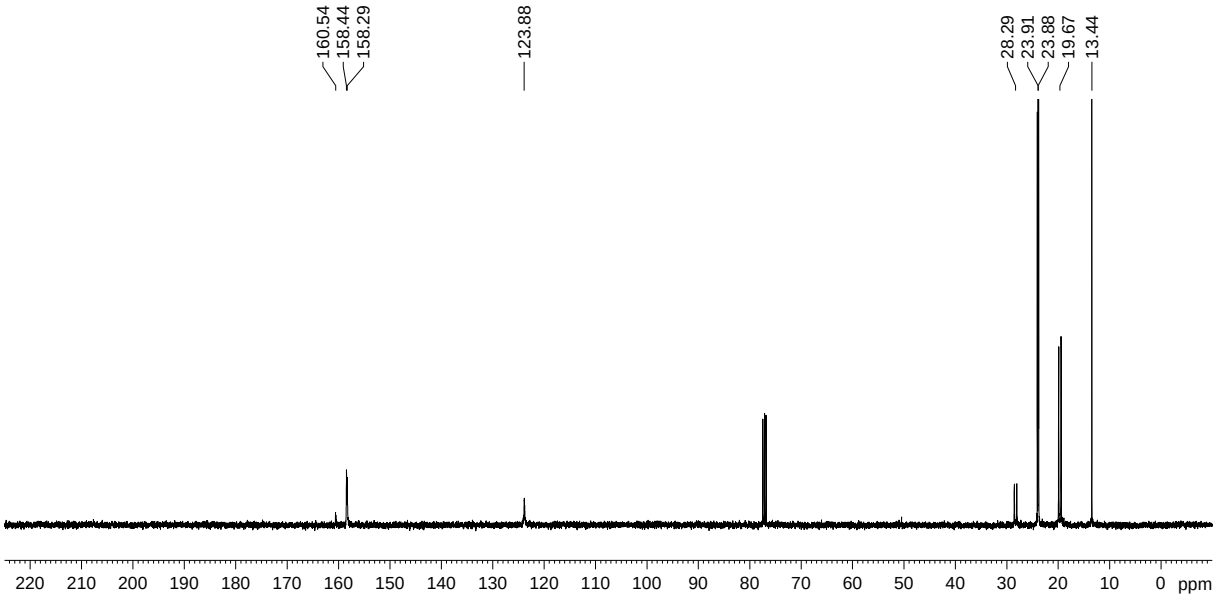
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.08 (d, *J* = 1.1 Hz, 1H), 8.70 (d, *J* = 5.0 Hz, 1H), 8.08 (dd, *J* = 5.0, 1.1, 1H), 4.68 (d, *J* = 15.6, 2H), 2.52-2.42 (m, 6H), 1.54-1.39 (m, 12H), 0.90 (td, *J* = 7.1, 1.0, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 160.5 (d, *J* = 8.0 Hz), 158.4, 158.3, 123.9, 28.3 (d, *J* = 48.0 Hz), 23.9 (d, *J* = 24.8 Hz), 23.9 (d, *J* = 4.0 Hz), 19.7 (d, *J* = 46.8 Hz), 13.4.

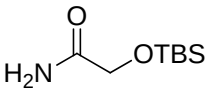
IR (film): ν 2959, 2931, 2871, 1576, 1550, 1464, 1386, 1320, 1233, 1095, 994, 914, 855, 719.

HRMS (ESI): calculated for $C_{17}H_{32}N_2P$ $[M]^+$ 295.2298, found 295.2298.





4.2.6 Functionalized Thiazole Heterocycles



170

2-(*tert*-butyldimethylsilyloxy)acetamide (170). To a solution of alcohol **167** (1.02 g, 13.61 mmol) and imidazole (2.32 g, 34.04 mmol) in DMF (10 mL) was added TBSCl (2.46 g, 16.34 mmol) in one portion at room temperature and the mixture was stirred for 26 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (10 mL). The aqueous phase was extracted with EtOAc (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 1:1) afforded 2.42 g (93%) of silyl ether **170** as a white solid.

TLC: R_f 0.21 (hexane/EtOAc 1:1, CPS).

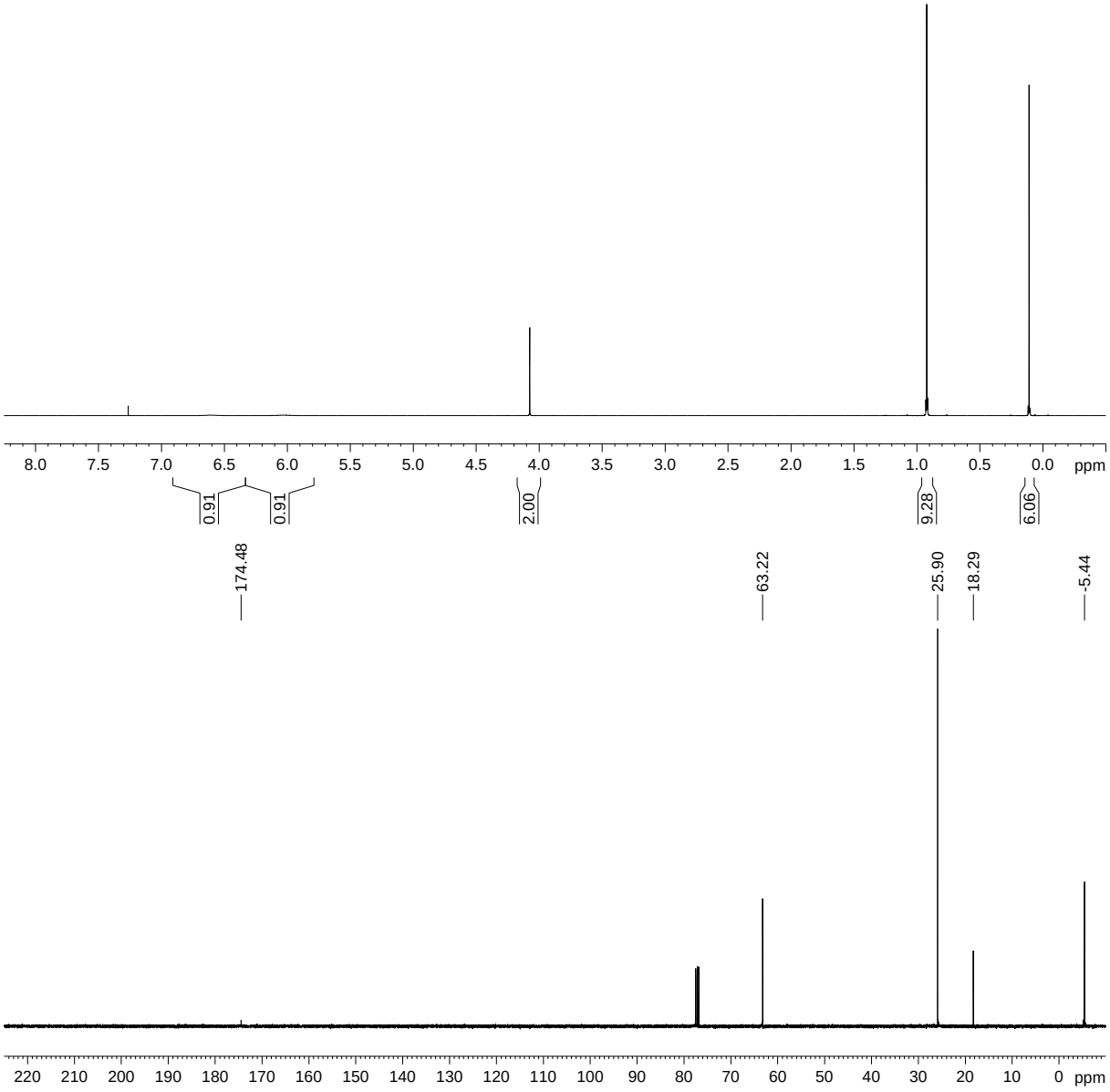
mp: 52-53 °C

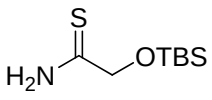
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.61 (br s, 1H), 5.94 (br s, 1H), 4.08 (s, 2H), 0.92 (s, 9H), 0.11 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 174.5, 63.2, 25.9, 18.3, -5.4.

IR (film): ν 3469, 3142, 2952, 2928, 2894, 2858, 1700, 1686, 1596, 1470, 1462, 1402, 1350, 1255, 1110, 1005, 852, 837, 780, 673.

HRMS (ESI): calculated for $C_8H_{20}NO_2Si$ $[M+H]^+$ 190.1258, found 190.1251.





171

2-(*tert*-butyldimethylsilyloxy)ethanethioamide (171). To a solution of amide **170** (1.20 g, 6.34 mmol) in dioxane (25 mL) was added Lawesson's reagent (2.56 mg, 6.34 mmol) in one portion at room temperature and the reaction mixture was then heated up to reflux for 2.5 h. The solution was then cooled and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 20:1) gave 0.82 g (63%) of thioacetamide **171** as a white solid.

TLC: R_f 0.24 (hexane/EtOAc 9:1, UV, CPS).

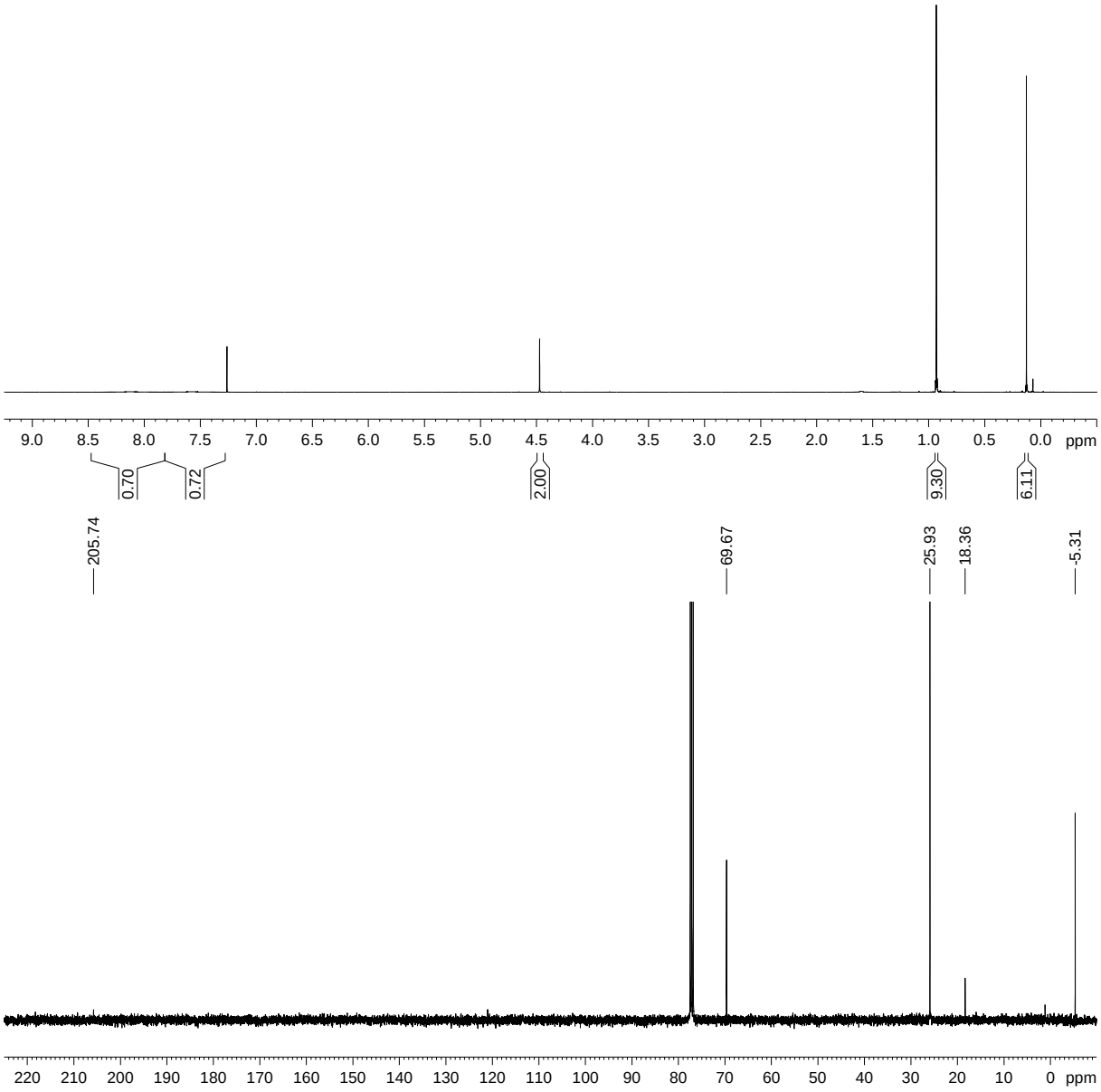
mp: 82-83 °C

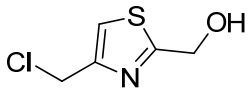
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.17 (br s, 1H), 7.59 (br s, 1H), 4.47 (s, 2H), 0.93 (s, 9H), 0.13 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 205.7, 69.7, 25.9, 18.4, -5.3.

IR (film): ν 3396, 3243, 3119, 2950, 2928, 2895, 2884, 2856, 1619, 1451, 1415, 1360, 1289, 1256, 1099, 969, 863, 836, 814, 779, 746,618.

HRMS (ESI): calculated for $C_8H_{20}NOSSi$ $[M+H]^+$ 206.1029, found 206.1028.





169

(4-(chloromethyl)thiazol-2-yl)methanol (169). A solution of 1,3-dichloropropan-2-one (0.40 g, 3.15 mmol) and thioacetamide **171** (0.54 g, 2.63 mmol) in EtOH (8 mL) was heated to 55 °C for 17 h. The reaction mixture was allowed to reach room temperature, water (15 mL) was added and a pH 8 was adjusted by addition of solid $NaHCO_3$ (0.23 g). The aqueous phase was extracted with EtOAc (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 2:1) yielded 0.21 g (50%) of chloride **169** as a slightly yellow oil.

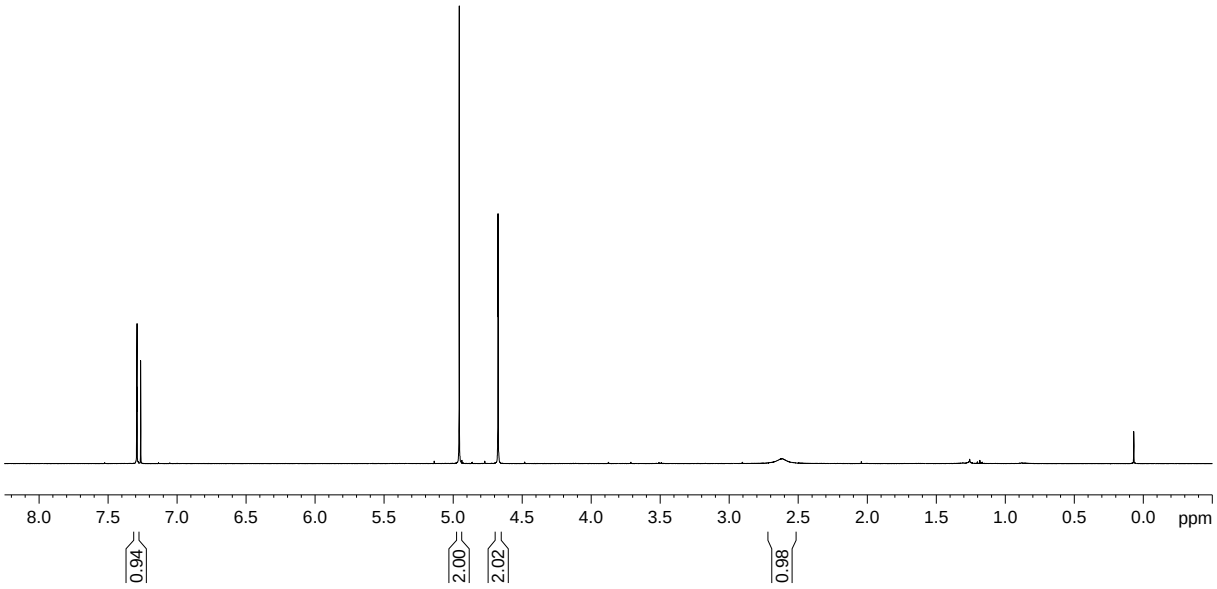
TLC: R_f 0.18 (hexane/EtOAc 2:1, UV, CPS).

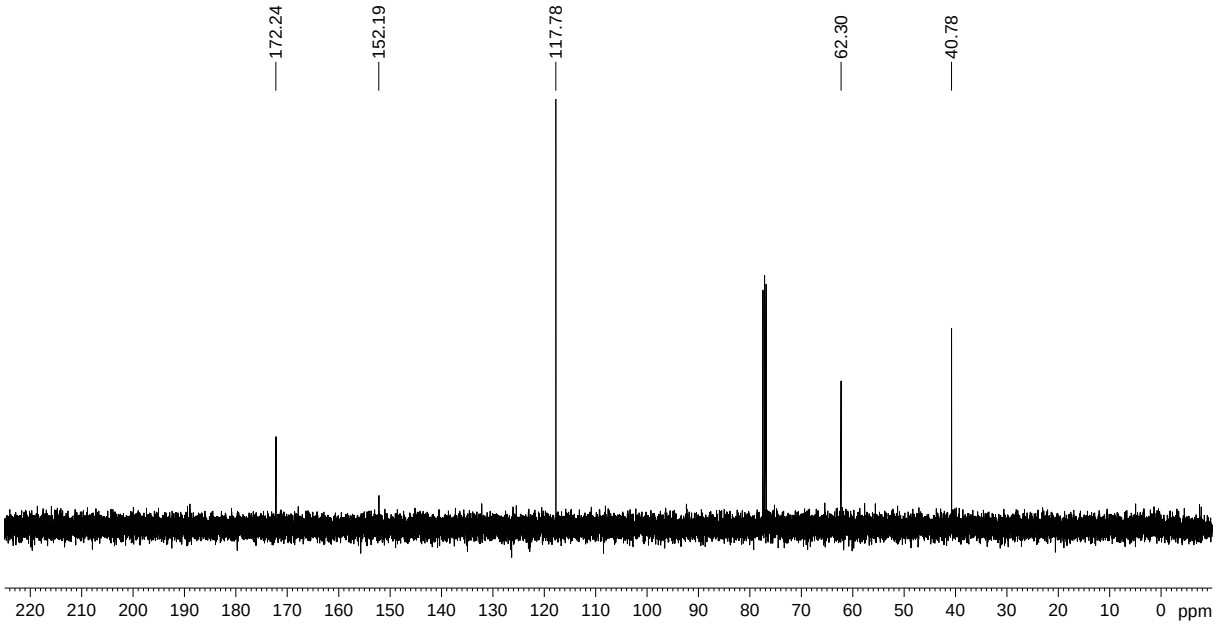
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.29 (t, *J* = 0.6 Hz, 1H), 4.96 (s, 2H), 4.67 (d, *J* = 0.6Hz, 2H), 2.62 (br s, 1H).

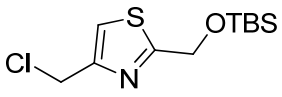
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 172.2, 152.2, 117.8, 62.3, 40.8.

IR (film): ν 3284, 2922, 2857, 1525, 1486, 1432, 1348, 1264, 1186, 1156, 1129, 1065, 967, 772, 714, 653.

HRMS (ESI): calculated for C_5H_7ClNOS $[M+H]^+$ 163.9931, found 163.9932.







172

2-((*tert*-butyldimethylsilyloxy)methyl)-4-(chloromethyl)thiazole (172). To a solution of alcohol **169** (135 mg, 0.83 mmol) and imidazole (124 mg, 1.82 mmol) in DMF (2 mL) was added TBSCl (137 mg, 0.91 mmol) in one portion at room temperature and the mixture was stirred for 22 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (2 mL). The aqueous phase was extracted with EtOAc (3 x 5 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 20:1) afforded 187 mg (91%) of silyl ether **172** as a colorless oil.

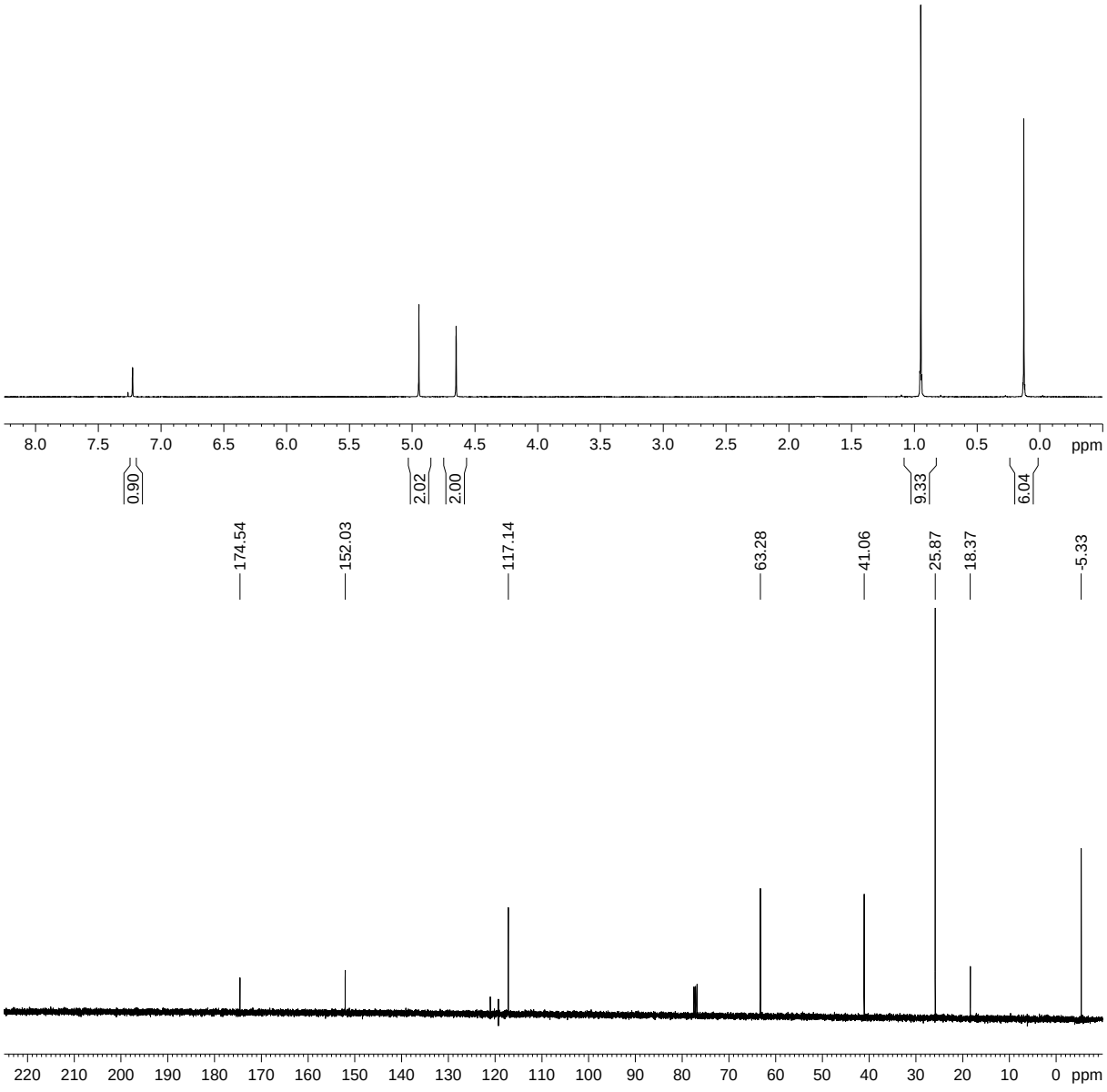
TLC: R_f 0.17 (hexane/EtOAc 50:1, UV, CPS).

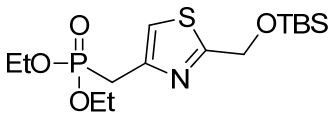
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.23 (t, *J* = 0.6 Hz, 1H), 4.95 (s, 2H), 4.65 (d, *J* = 0.6Hz, 2H), 0.95 (s, 9H), 0.13 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 174.5, 152.0, 117.1, 63.3, 41.0, 25.9, 18.4, -5.3.

IR (film): ν 2954, 2929, 2885, 2858, 1471, 1463, 1355, 1257, 1198, 1131, 1105, 1006, 8366, 778, 697, 667.

HRMS (ESI): calculated for $C_{11}H_{21}ClNOSSi$ $[M+H]^+$ 278.0796, found 278.0797.





150

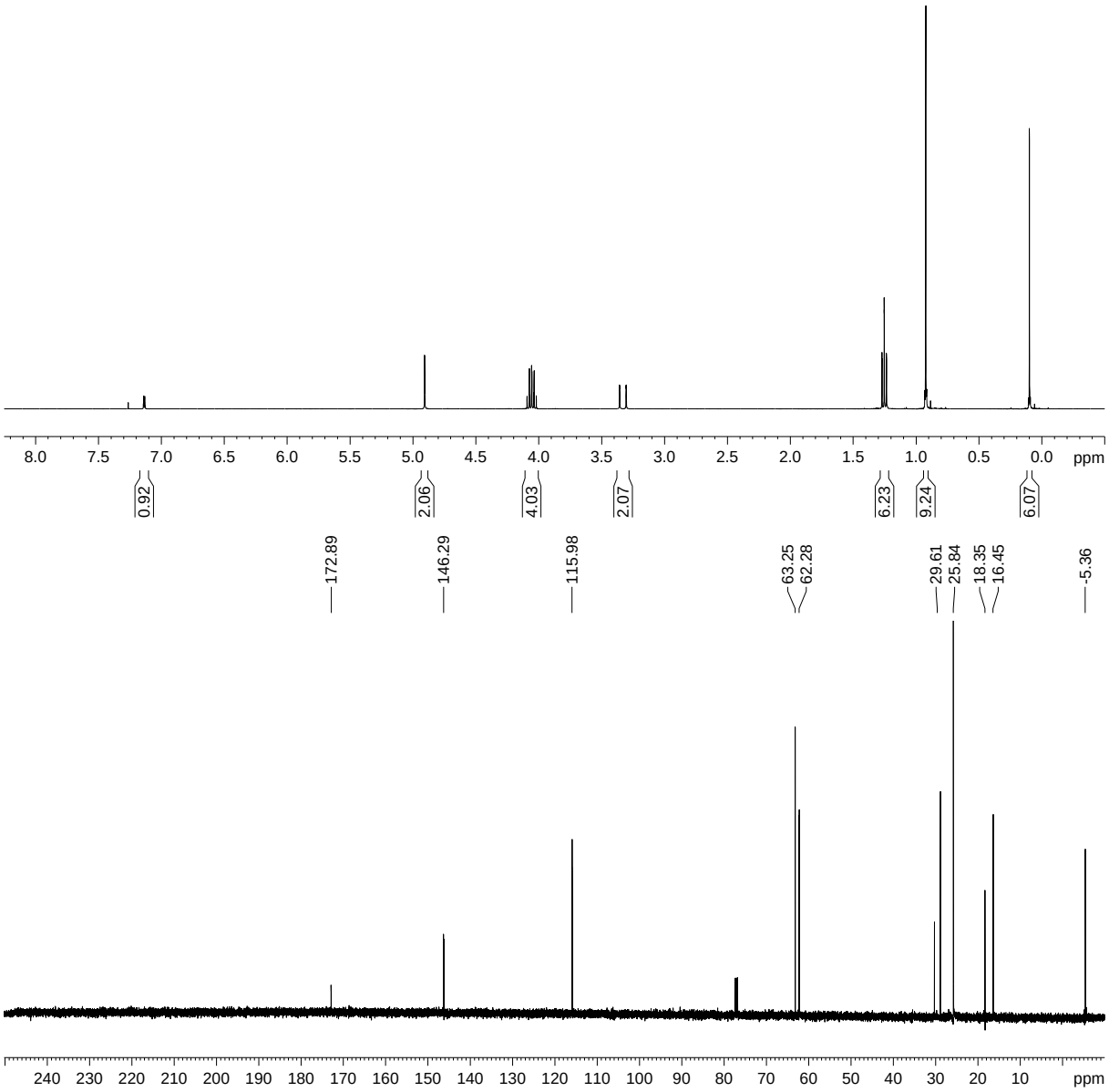
Diethyl ((2-((*tert*-butyldimethylsilyloxy)methyl)thiazol-4-yl)methyl)phosphonate (150). A mixture of chloride **172** (170 mg, 0.61 mmol) and triethyl phosphate (107 mg, 0.64 mmol) was heated up to 160 °C for 6h. The mixture was then cooled and the excess of triethyl phosphate was removed under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 1:3) gave 37 mg (19%) of phsphonate **150** as a colorless oil.

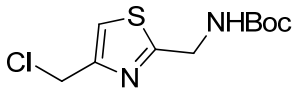
TLC: R_f 0.18 (hexane/EtOAc 1:2, UV, CPS).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.14 (dt, *J* = 3.6, 0.7 Hz, 1H), 4.91 (s, 1H), 4.06 (dq, *J* = 8.1. 7.0 Hz, 4H), 3.33 (dd, *J* = 21.0, 0.7 Hz, 2H), 1.25 (t, *J* = 7.0 Hz, 6H), 0.92 (s, 9H), 0.10 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 172.9, 146.3 (d, *J* = 8.0 Hz), 116.0 (d, *J* = 7.3 Hz), 63.2, 62.3 (d, *J* = 6.4 Hz), 29.6 (d, *J* = 141.0), 25.8, 18.3, 16.5 (d, *J* = 5.9 Hz), -5.4.

IR (film): ν 2984, 2911, 1521, 1394, 1232, 1164, 1052, 1030, 969, 958.





174

***tert*-Butyl ((4-(chloromethyl)thiazol-2-yl)methyl)carbamate (174).** A solution of 1,3-dichloropropan-2-one (100 mg, 0.79 mmol) and thioacetamide (**173**) (150 mg, 0.79 mmol) in EtOH (1.5 mL) over molecular sieves (80 mg) was heated to 45 °C for 36 h. The reaction mixture was then filtered through a small plug of celite and the precipitat was rinsed with EtOH (3 mL). Water (5 mL) was added to the dark filtrate and a pH 8 was adjusted by addition of solid $NaHCO_3$ (70 mg). The aqueous phase was extracted with EtOAc (3 x 5 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 3:1) afforded 67 mg (34%) of chloride **174** as a slightly yellow oil.

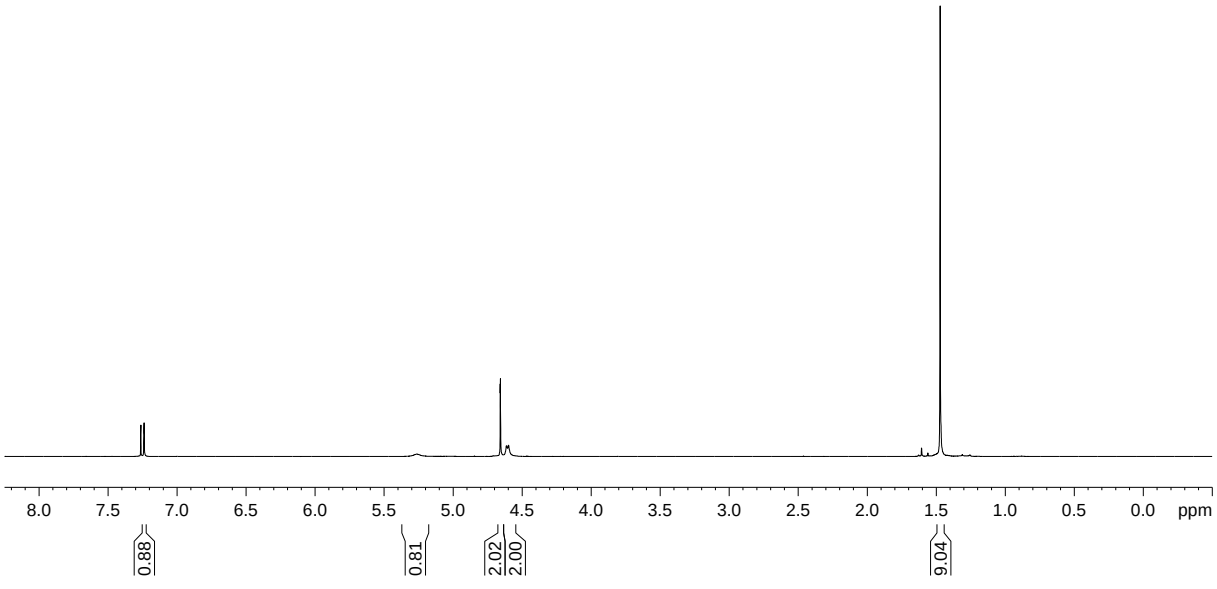
TLC: R_f 0.25 (hexane/EtOAc 3:1, UV, CPS).

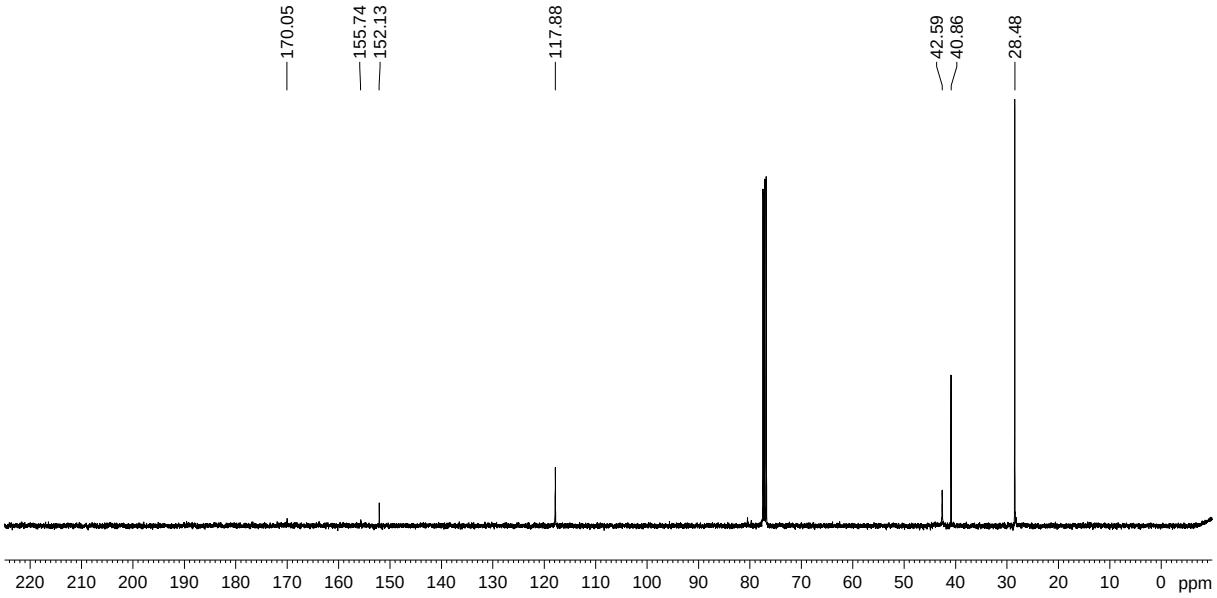
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.24 (t, *J* = 0.6 Hz, 1H), 5.26 (br s, 1H), 4.65 (d, *J* = 0.6 Hz, 2H), 4.60 (d, *J* = 6.0 Hz, 2H), 1.47 (s, 9H).

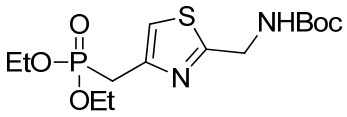
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.0, 155.7, 152.1, 117.9, 80.5, 42.6, 40.9, 28.5.

IR (film): ν 3335, 2978, 1698, 1523, 1367, 1251, 1165, 1165, 934, 715.

HRMS (ESI): calculated for $C_{10}H_{16}ClN_2O_8S$ $[M+H]^+$ 263.0616, found 263.0612.







175

***tert*-Butyl ((4-((diethoxyphosphoryl)methyl)thiazol-2-yl)methyl)carbamate (175).** A mixture of chloride **174** (0.81 g, 3.08 mmol) and triethyl phosphate (1.02 g, 6.17 mmol) was heated up to 160 °C for 6h. The mixture was then cooled and the excess of triethyl phosphate was removed under reduced pressure. Purification of the residue by flash column chromatography (EtOAc) gave 0.98 g (87%) of phsphonate **175** as a colorless oil.

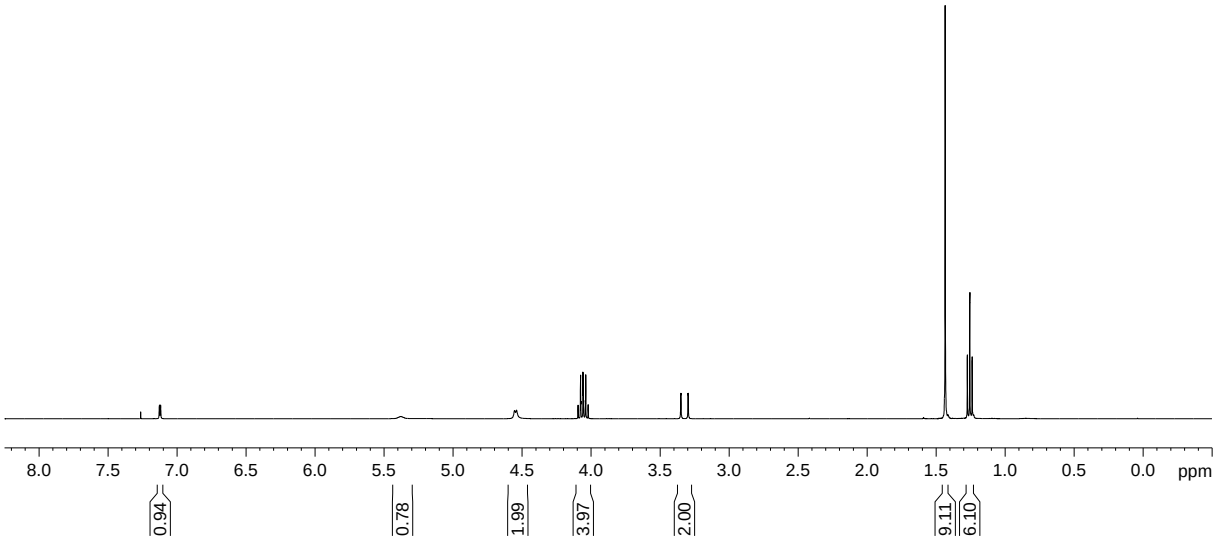
TLC: R_f 0.11 (EtOAc, UV, CPS).

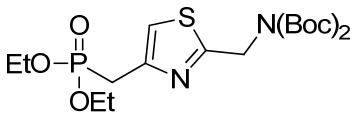
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.12 (d, *J* = 3.6 Hz, 1H), 5.38 (br s, 1H), 4.55 (d, *J* = 5.7 Hz, 2H), 4.06 (dq, *J* = 8.0. 7.1 Hz, 4H), 3.32 (d, *J* = 21.0 Hz, 2H), 1.43 (s, 9H), 1.26 (t, *J* = 7.1 Hz, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 169.9, 156.0, 151.7, 102.8, 83.6, 62.6 (d, *J* = 6.5 Hz), 41.9, 28.1, 23.8 (d, *J* = 142.3 Hz), 16.5 (d, *J* = 6.0 Hz).

IR (film): ν 3285, 2979, 2931, 2910, 1715, 1521, 1392, 1366, 1250, 1166, 1052, 1026, 966, 933, 791.

HRMS (ESI): calculated for $C_{19}H_{34}N_2O_7PS$ $[M+H]^+$ 365.1295, found 365.1294.





145f

Di-*tert*-butyl ((4-((diethoxyphosphoryl)methyl)thiazol-2-yl)methyl)dicarbamate (145f). To a solution of phosphonate **175** (150 mg, 0.41 mmol) in MeCN (4.0 mL) was added Boc_2O (180 mg, 0.82 mmol) and DMAP (25 mg, 0.21 mmol) at room temperature and the reaction mixture was stirred for 13 h. The reaction mixture was then quenched by addition of saturated aqueous NH_4Cl (5 mL) and the aqueous phase was extracted with EtOAc (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc) to yield 186 mg (97%) of double-Boc protected thiazole **145f** as a slightly yellow oil.

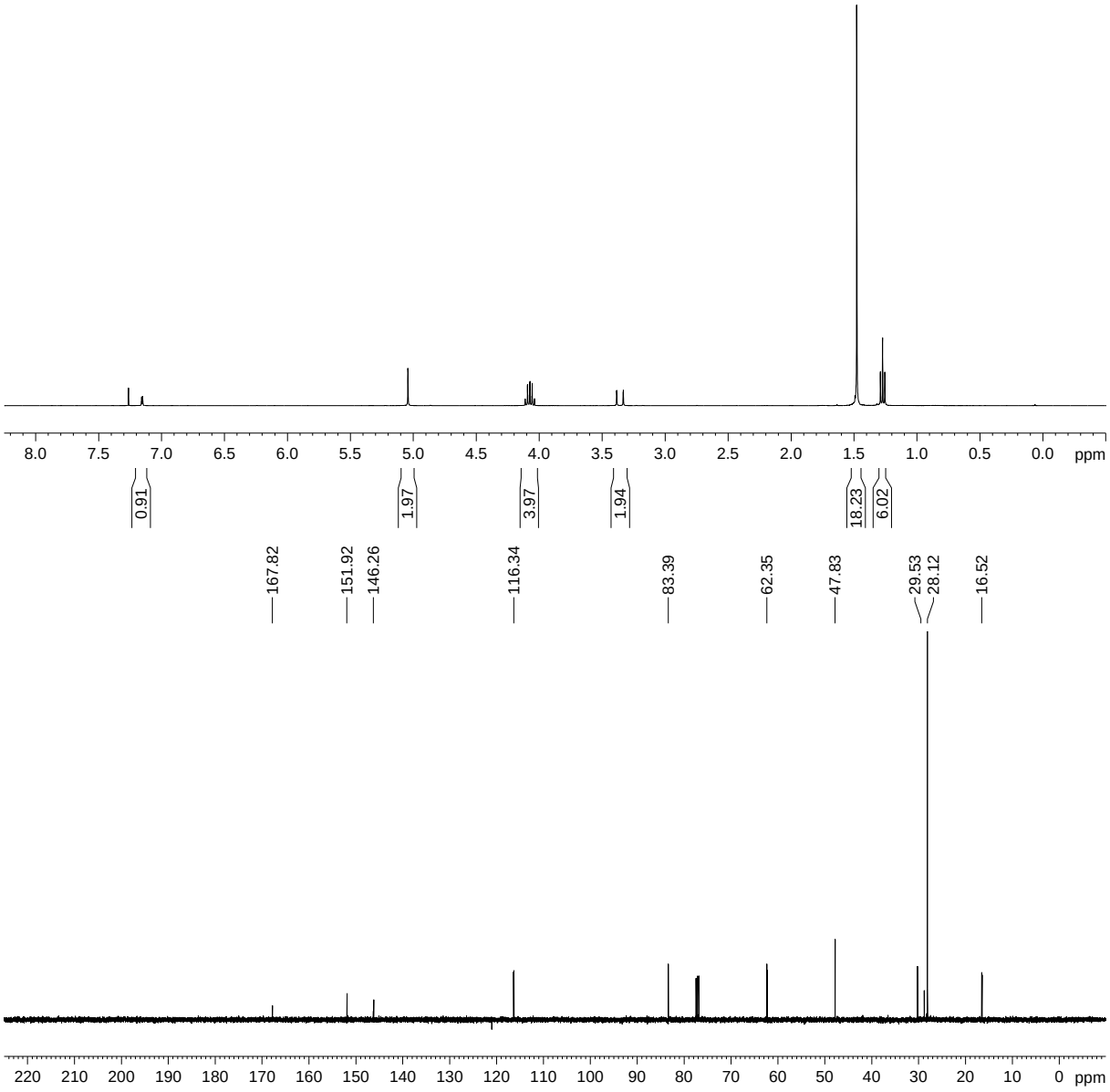
TLC: R_f 0.24 (EtOAc, UV, CPS).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.15 (d, *J* = 3.5 Hz, 1H), 5.05 (s, 2H), 4.08 (dq, *J* = 8.0. 7.1 Hz, 4H), 3.36 (d, *J* = 21.0 Hz, 2H), 1.48 (s, 18H), 1.27 (t, *J* = 7.1 Hz, 6H).

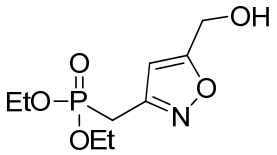
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 167.8, 151.9, 146.3 (d, *J* = 7.7 Hz), 116.3 (d, *J* = 7.2 Hz), 83.4, 62.3 (d, *J* = 6.4 Hz), 47.8, 29.5 (d, *J* = 141.1 Hz), 28.1, 16.5 (d, *J* = 6.0 Hz).

IR (film): ν 2980, 2933, 1794, 1753, 1700, 1520, 1479, 1458, 1422, 1392, 1367, 1341, 1254, 1228, 1165, 1129, 1129, 1054, 964, 854, 782.

HRMS (ESI): calculated for $C_{19}H_{34}N_2O_7PS$ $[M+H]^+$ 465.1819, found 465.1818.



4.2.7 Functionalized Isoxazole Heterocycles



184

Diethyl ((5-(hydroymethyl))isoxazol-3-yl)methyl)phosphonate (184). To a solution of oxime **183** (3.80 g, 19.47 mmol) and pyridine (15 drops) in $CHCl_3$ (60 mL) is added NCS (3.38 g, 25.31 mmol) in portions at room temperature. The reaction mixture is then heated up to 45 °C and stirred for 24 h at this temperature. Propargyl alcohol (1.31 g, 23.37 mmol) in

$CHCl_3$ (20 mL) is added dropwise and after 10 min NEt_3 (3.25 mL, 23.37 mmol) is added over a period of 40 min at 45 °C. Stirring is continued for 4 h at the same temperature. The reaction mixture is then allowed to reach room temperature washed with water (2 x 40 mL). The organic phase is dried over $MgSO_4$, removed under reduced pressure and the residue was purified by flash column chromatography (EtOAc) to give 1.91 g (40%) of isoxazole **184** as a colorless oil.

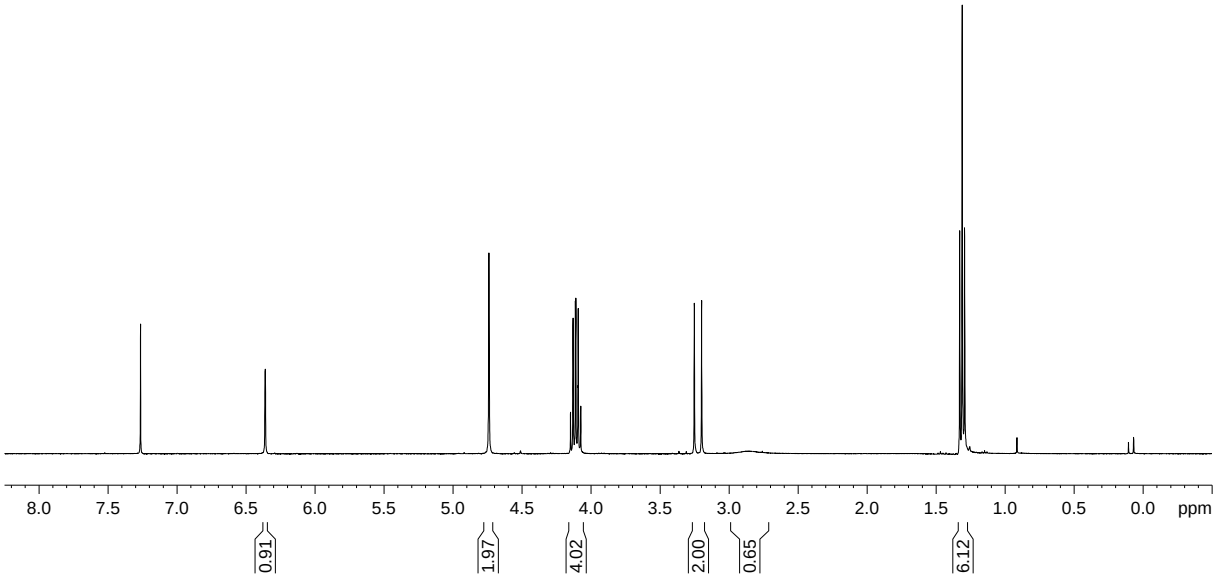
TLC: R_f 0.17 (EtOAc, CPS).

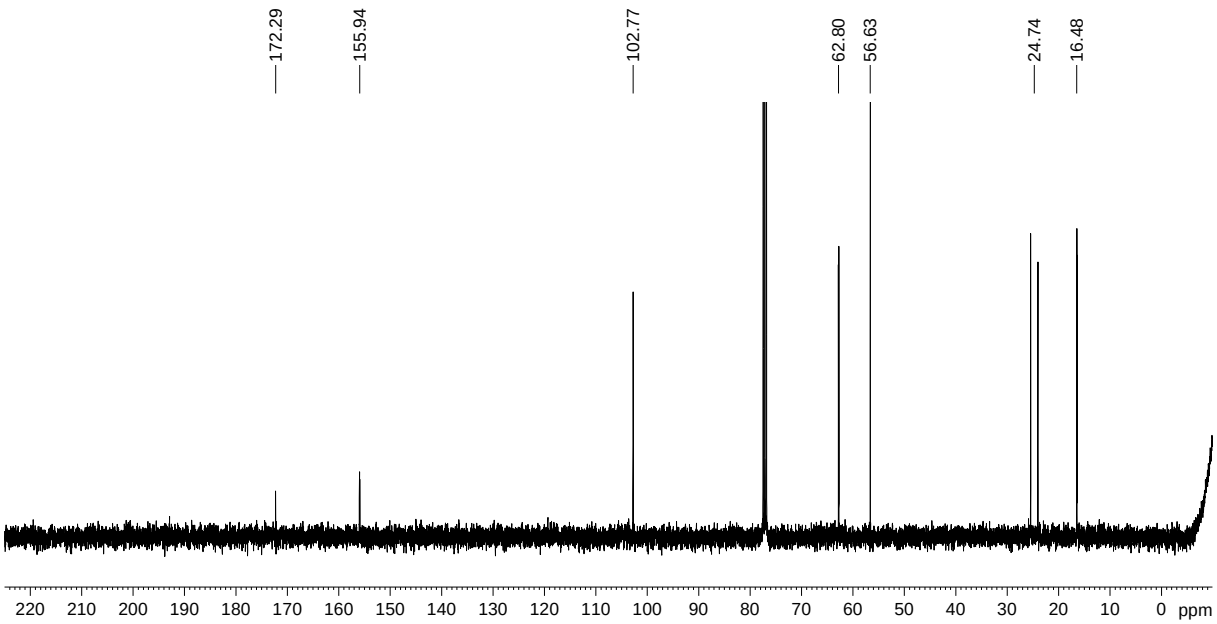
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.36 (s, 1H), 4.74 (s, 2H), 4.11 (dq, *J* = 8.1, 7.1 Hz, 4H), 3.22 (d, *J* = 21.0 Hz, 2H), 2.86 (br s, 1H), 1.31 (t, *J* = 7.1 Hz, 6H).

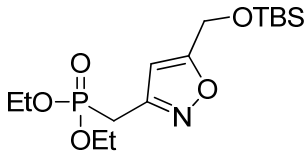
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 172.3 (d, *J* = 2.0 Hz), 155.9 (d, *J* = 6.9 Hz), 102.8 (d, *J* = 2.4 Hz), 62.8 (d, *J* = 6.7 Hz), 56.6, 25.9, 24.7 (d, *J* = 142.6 Hz), 18.4, 16.5 (d, *J* = 5.9 Hz).

IR (film): ν 3361, 2986, 2912, 1606, 1428, 1395, 1371, 1237, 1163, 1050, 1024, 905, 847, 816, 756, 529.

HRMS (ESI): calculated for $C_9H_{17}NO_5P$ $[M+H]^+$ 250.0839, found 250.0836.







140b

Diethyl ((5-((*tert*-butyldimethylsilyloxy)methyl)isoxazol-3-yl)methyl)phosphonate (140b). To a solution of alcohol **184** (1.90 g, 7.62 mmol) and imidazole (1.30 g, 19.06 mmol) in DMF (10 mL) was added TBSCl (1.38 g, 9.15 mmol) in one portion at room temperature and the mixture was stirred for 24 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (10 mL). The aqueous phase was extracted with EtOAc (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (EtOAc) afforded 2.60 g (94%) of silyl ether **140b** as a colorless oil.

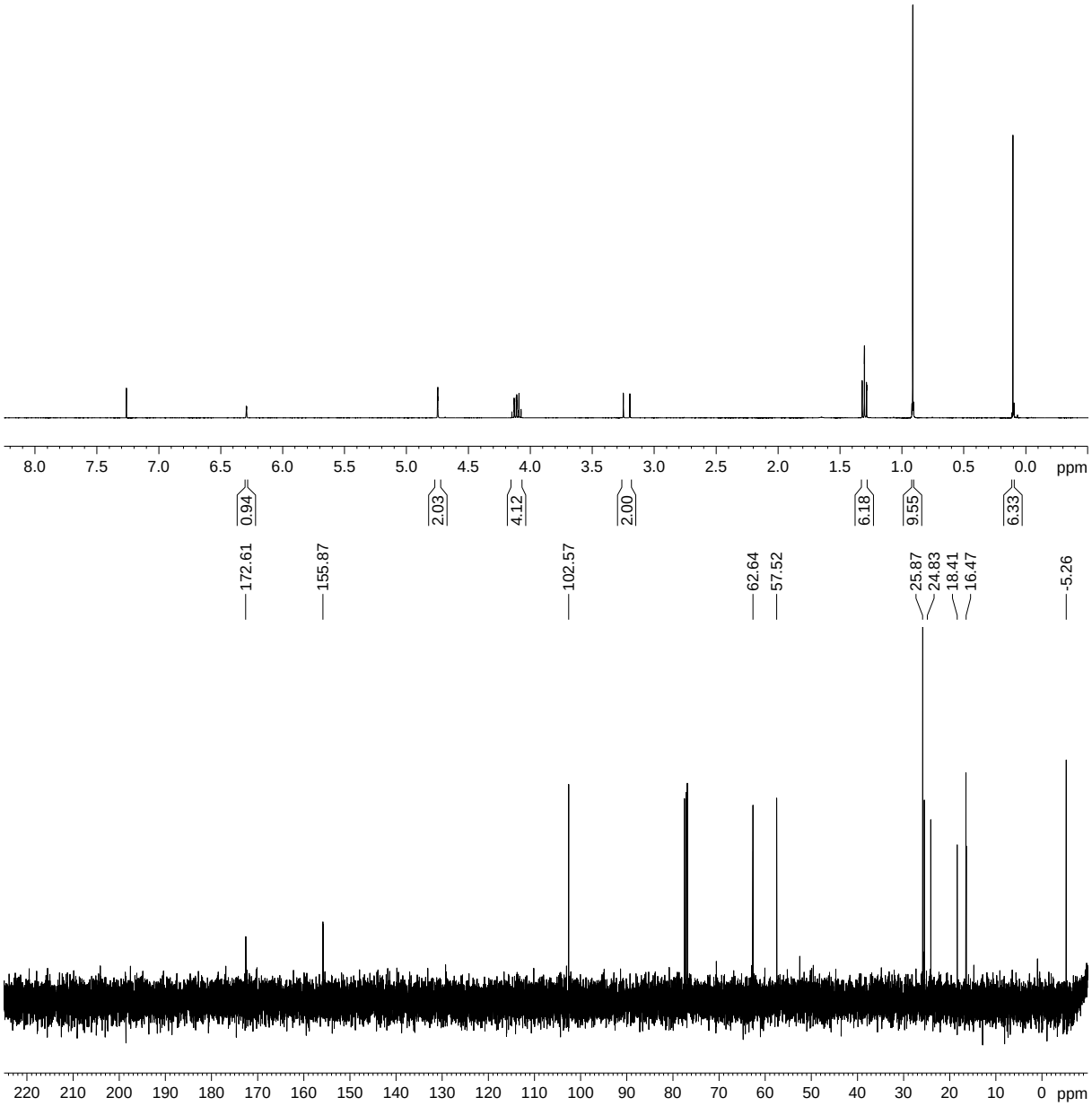
TLC: R_f 0.46 (EtOAc, $KMnO_4$, CPS).

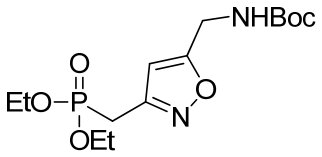
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.29 (q, *J* = 0.9 Hz, 1H), 4.75 (t, *J* = 0.6 Hz, 2H), 4.11 (dq, *J* = 8.1, 7.1 Hz, 4H), 3.22 (d, *J* = 21.0 Hz, 2H), 1.30 (t, *J* = 7.1 Hz, 6H), 0.91 (s, 9H), 0.10 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 172.6 (d, *J* = 2.4 Hz), 155.9 (d, J = 7.2 Hz), 102.6 (d, *J* = 2.4 Hz), 62.6 (d, *J* = 6.6 Hz), 57.5, 25.9, 24.8 (d, *J* = 142.4 Hz), 18.4, 16.5 (d, *J* = 6.2 Hz), -5.3.

IR (film): ν 2956, 2930, 2858, 1608, 1473, 1463, 1428, 1391, 1256, 1144, 1128, 1097, 1053, 1025, 966, 907, 837, 780, 539, 530.

HRMS (ESI): calculated for $C_{15}H_{31}NO_5PSi$ $[M+H]^+$ 364.1704, found 364.1709.





185

***tert*-Butyl ((3-((diethoxyphosporyl)methyl)ixoxazol-5-yl)methyl)carbamate (185).** To a solution of oxime **183** (1.85 g, 9.47 mmol) and pyridine (10 drops) in $CHCl_3$ (30 mL) is added NCS (1.52 g, 11.37 mmol) in portions at room temperature. The reaction mixture is then heated up to 45 °C and stirred for 20 h at this temperature. *Tert*-butyl prop-2-yn-1-ylcarbamate (1.76 g, 11.37 mmol) in $CHCl_3$ (10 mL) is added dropwise and after 10 min NEt_3 (1.58 mL, 11.37 mmol) is added over a period of 40 min at 45 °C. Stirring is continued for 4 h

at the same temperature. The reaction mixture is then allowed to reach room temperature washed with water (2 x 40 mL). The organic phase is dried over $MgSO_4$, removed under reduced pressure and the residue was purified by flash column chromatography (EtOAc) to give 1.73 g (55%) of isoxazole **185** as a colorless oil.

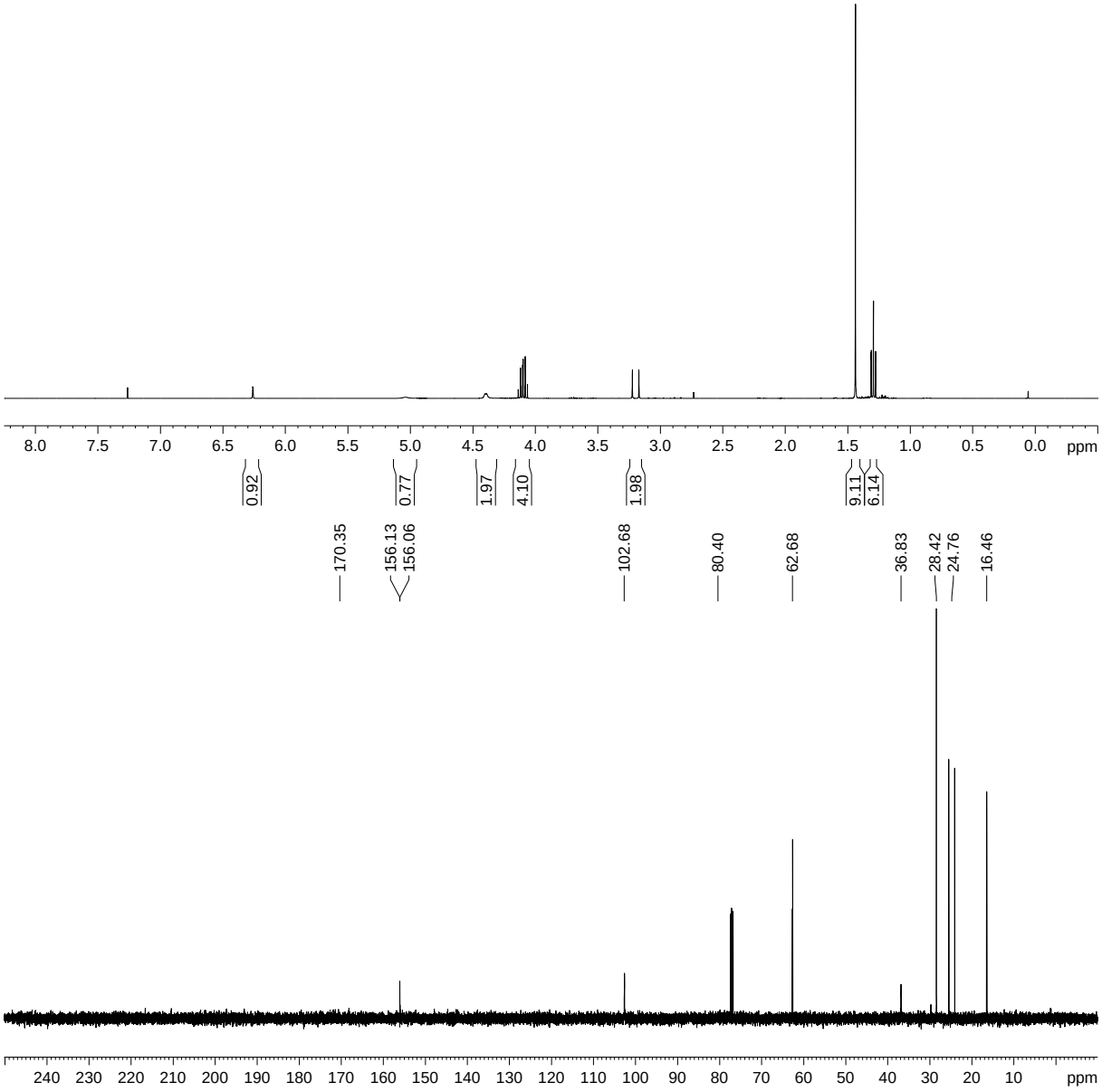
TLC: R_f 0.17 (EtOAc, UV, CPS).

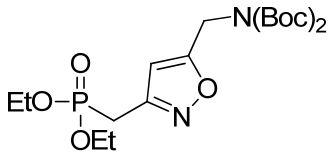
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.26 (s, 1H), 5.04 (br s, 1H), 4.40 (d, *J* = 4.8 Hz, 2H), 4.10 (dq, *J* = 8.2, 7.1 Hz, 4H), 3.20 (d, *J* = 21.0 Hz, 2H), 1.44 (s, 9H), 1.30 (t, *J* = 7.1 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.3, 156.2 (d, *J* = 2.3 Hz), 156.1 (d, *J* = 1.6 Hz), 102.7, 80.4, 62.7 (d, *J* = 6.6 Hz), 36.8, 28.4, 24.8 (*J* = 142.2), 16.5 (d, *J* = 5.9 Hz).

IR (film): ν 3287, 2979, 2932, 1714, 1605, 1521, 1509, 1432, 1366, 1251, 1164, 1051, 1023, 967, 902, 850, 791, 758, 404.

HRMS (ESI): calculated for $C_{14}H_{25}N_2O_6P$ $[M+H]^+$ 349.1523, found 349.1529.





145g

Di-*tert*-butyl ((3-((diethoxyphosporyl)methyl)ixoxazol-5-yl)methyl)dicarbamate (145g). To a solution of mono-Boc protected isoxazole 1**85** (175 mg, 0.50 mmol) in MeCN (4.5 mL) was added Boc_2O (219 mg, 1.00 mmol) and DMAP (31 mg, 0.25 mmol) at room temperature and the reaction mixture was stirred for 10 h. The reaction mixture was then quenched by addition of saturated aqueous NH_4Cl (5 mL) and the aqueous phase was extracted with EtOAc (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc) to yield 192 mg (85%) of double-Boc protected isoxazole **145g** as a slightly yellow oil.

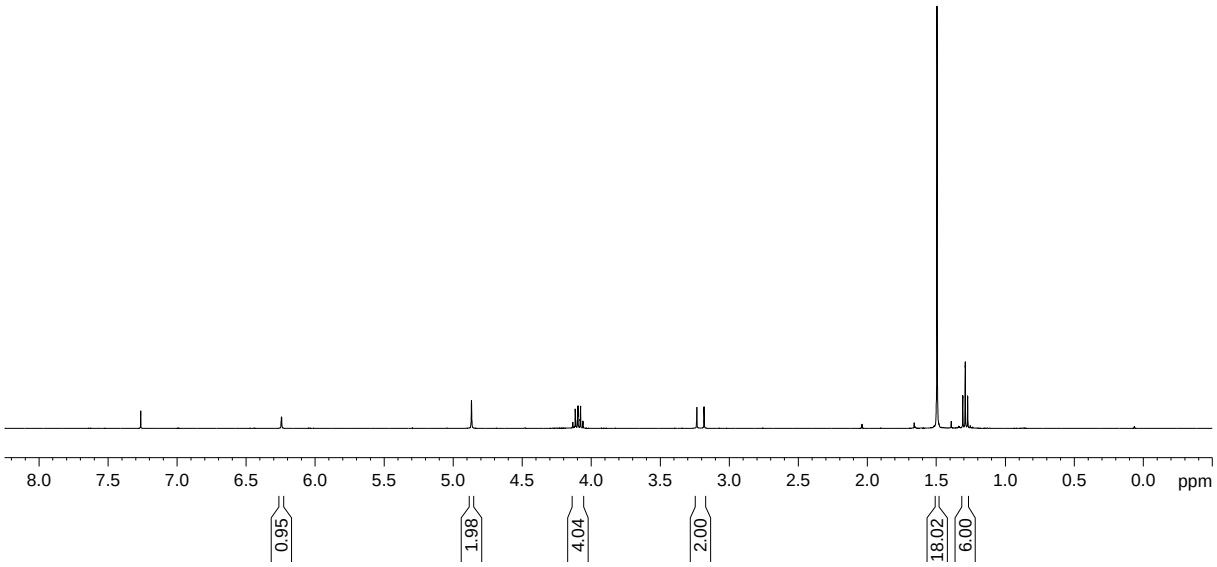
TLC: R_f 0.30 (EtOAc, CPS).

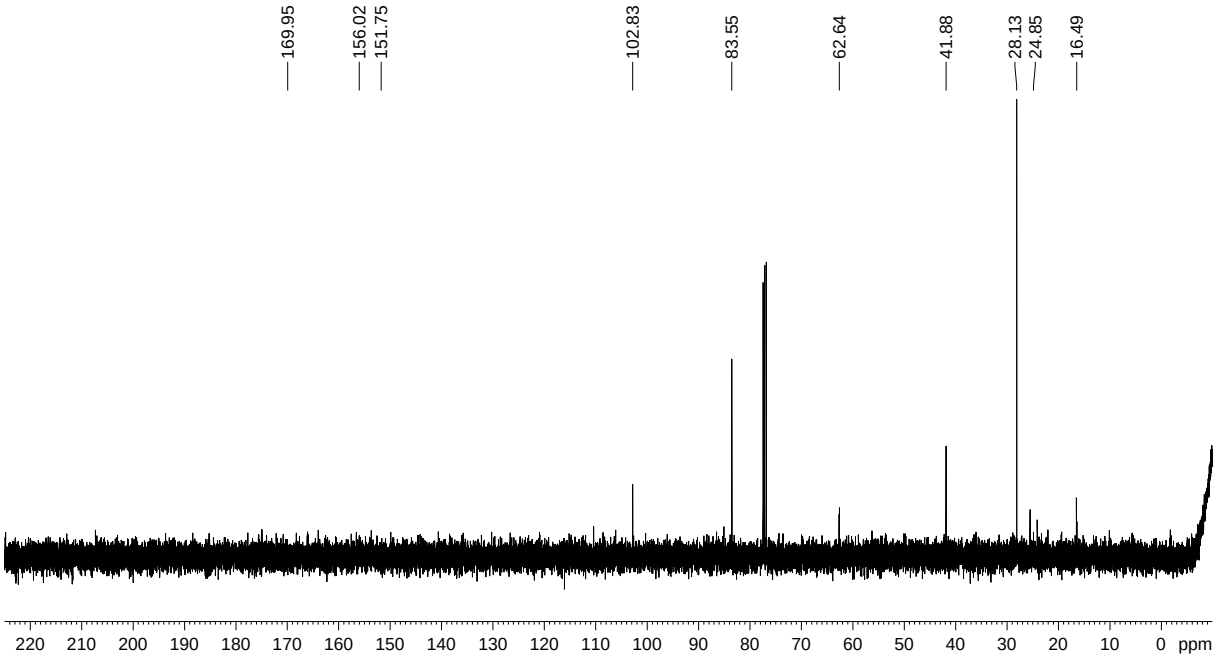
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.24 (d, *J* = 0.7 Hz, 1H), 4.87 (d, *J* = 0.7 Hz, 2H), 4.10 (dq, *J* = 8.1, 7.1 Hz, 4H), 3.21 (d, *J* = 21.0 Hz, 2H), 1.49 (s, 18H), 1.29 (td, *J* = 7.1, 0.5 Hz, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 169.9, 156.0 (d, *J* = 7.2 Hz), 151.7, 102.8, 83.6, 62.6 (d, *J* = 6.5 Hz), 41.9, 28.1, 24.8 (d, *J* = 142.2 Hz), 16.5 (d, *J* = 5.5 Hz).

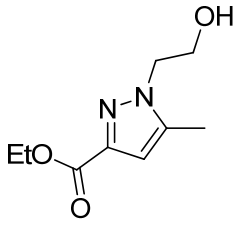
IR (film): ν 2981, 2935, 1793, 1754, 1701, 1606, 1480, 1428, 1392, 1369, 1349, 1257, 1223, 1141, 1054, 1026, 966, 852, 793, 763.

HRMS (ESI): calculated for $C_{19}H_{37}N_3O_8P$ $[M+NH_4]^+$ 466.2313, found 466.2313.





4.2.8 Functionalized Pyrazole Heterocycles



198

Ethyl 1-(2-hydroxyethyl)-5-methyl-1*H*-pyrazole-3-carboxylate (198). To a solution of ethyl acetopyruvate (**157**) (1.78 mL, 12.65 mmol) in EtOH (50 mL) was added 2-hydroxyethylhydrazine (0.86 mL, 12.65 mmol) at room temperature and the reaction mixture was heated to 60 °C for 1.5 h. The reaction mixture was allowed to reach room temperature and the reaction was then quenched by addition of saturated aqueous NH_4Cl (50 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 60 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 1.41 g (56%) of pyrazole **198** as a slightly yellow oil.

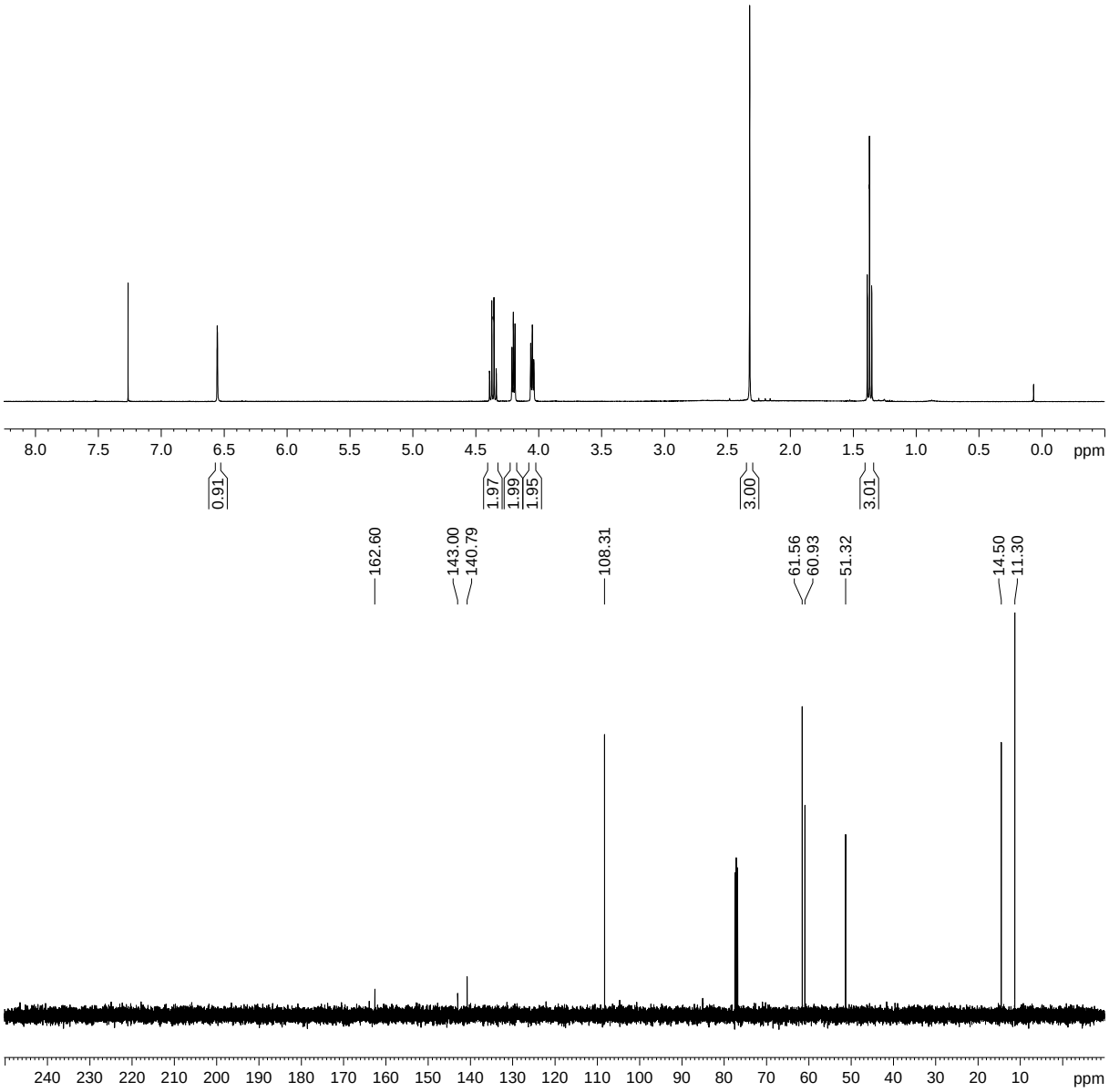
TLC: R_f 0.09 (hexane/EtOAc 1:1, UV, $KMnO_4$, CPS).

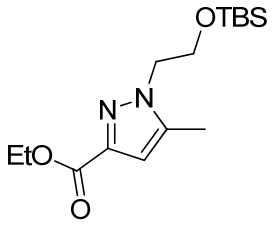
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.55 (s, 1H), 4.36 (q, *J* = 7.1, 2H), 4.20 (t, *J* = 5.0 Hz, 2H), 4.05 (t, *J* = 5.0 Hz, 2H), 2.32 (s, 3H), 1.37 (t, *J* = 7.1 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.6, 143.0, 140.8, 108.3, 61.6, 60.9, 51.3, 14.5, 11.3.

IR (film): ν 3398, 2980, 2957, 2937, 2878, 1714, 1550, 1443, 1429, 1389, 1215, 1137, 1106, 1056, 1027, 866, 840, 777.

HRMS (ESI): calculated for $C_9H_{15}N_2O_3$ $[M+H]^+$ 1991077, found 199.1076.





201

Ethyl 1-(2-(*tert*-butyldimethylsilyloxy)ethyl)-5-methyl-1*H*-pyrazole-3-carboxylate (201). To a solution of alcohol **198** (1.41 g, 7.10 mmol) and imidazole (1.06 g, 15.63 mmol) in DMF (20 mL) was added TBSCl (1.18 g, 7.81 mmol) in one portion at room temperature and the mixture was stirred for 12 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (20 mL). The aqueous phase was extracted with EtOAc (3 x 25 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 4:1) afforded 2.08 g (94%) of silyl ether **201** as a colorless oil.

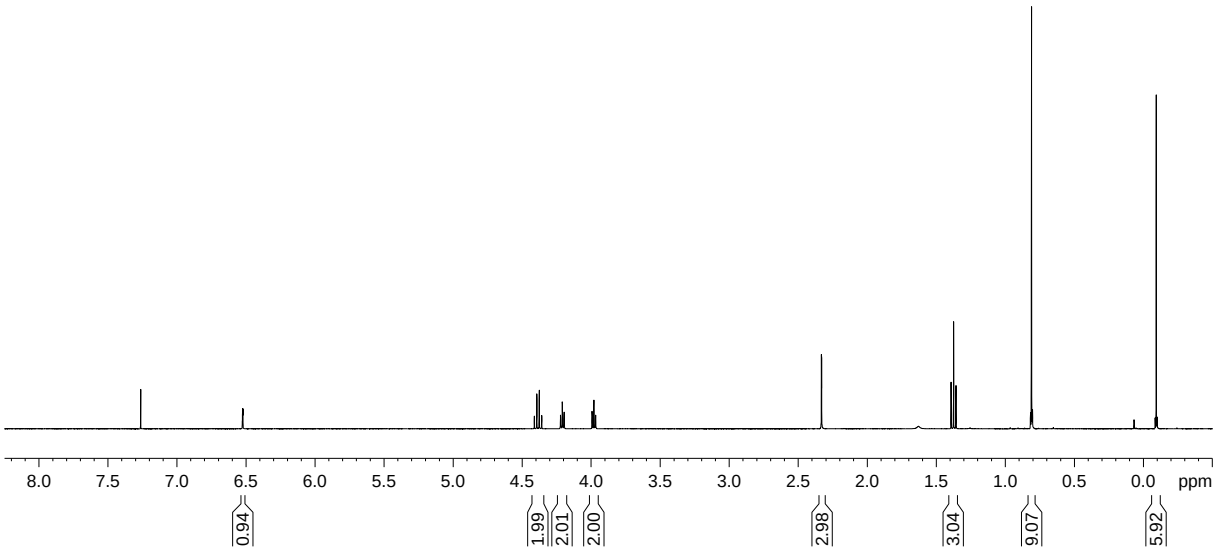
TLC: R_f 0.25 (hexane/EtOAc 4:1, UV, $KMnO_4$).

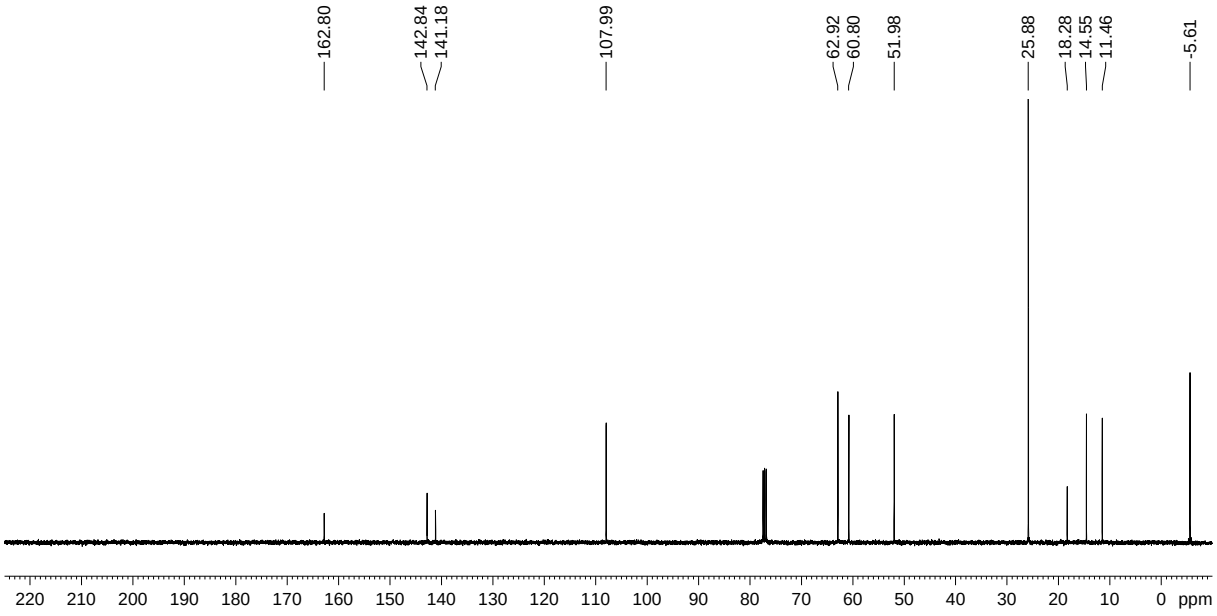
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.52 (q, *J* = 0.7 Hz, 1H), 4.39 (q, *J* = 7.1 Hz, 2H), 4.21 (t, *J* = 5.2 Hz, 2H), 3.98 (t, *J* = 5.2 Hz, 3H), 2.33 (d, *J* = 0.7 Hz, 3H), 1.38 (t, *J* = 7.1 Hz, 3H), 0.82 (s, 9H), -0.09 (s, 6H).

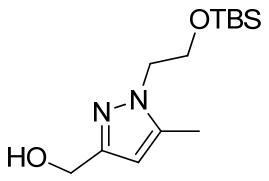
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.8, 142.8, 141.2, 108.0, 62.9, 60.8, 52.0, 25.9, 18.3, 14.6, 11.5, -5.6.

IR (film): ν 2954, 2929, 2857, 1732, 1716, 1472, 1443, 1389, 1382, 1362, 1253, 1218, 1209, 1146, 1105, 1062, 1029, 927, 834, 828, 811, 776.

HRMS (ESI): calculated for $C_{15}H_{29}N_2O_3Si$ $[M+H]^+$ 313.1942, found 313.1945.







202

(1-(2-((*tert*-butyldimethylsilyloxy)ethyl)-5-methyl-1*H*-pyrazol-3-yl)methanol (202). To a solution of ester **201** (2.05 g, 6.56 mmol) in CH_2Cl_2 (100 mL) was added DIBAL-H (1 M in CH_2Cl_2, 16.04 mL, 14.43 mmol) dropwise at -78 °C. The reaction mixture was stirred for 0.5 h at this temperature, was then allowed to warm to -20 °C slowly and stirred for 0.5 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (150 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases got transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to give 1.73 g (98%) of alcohol **202** as a white solid.

TLC: R_f 0.26 (hexane/EtOAc 1:1, $KMnO_4$, CPS).

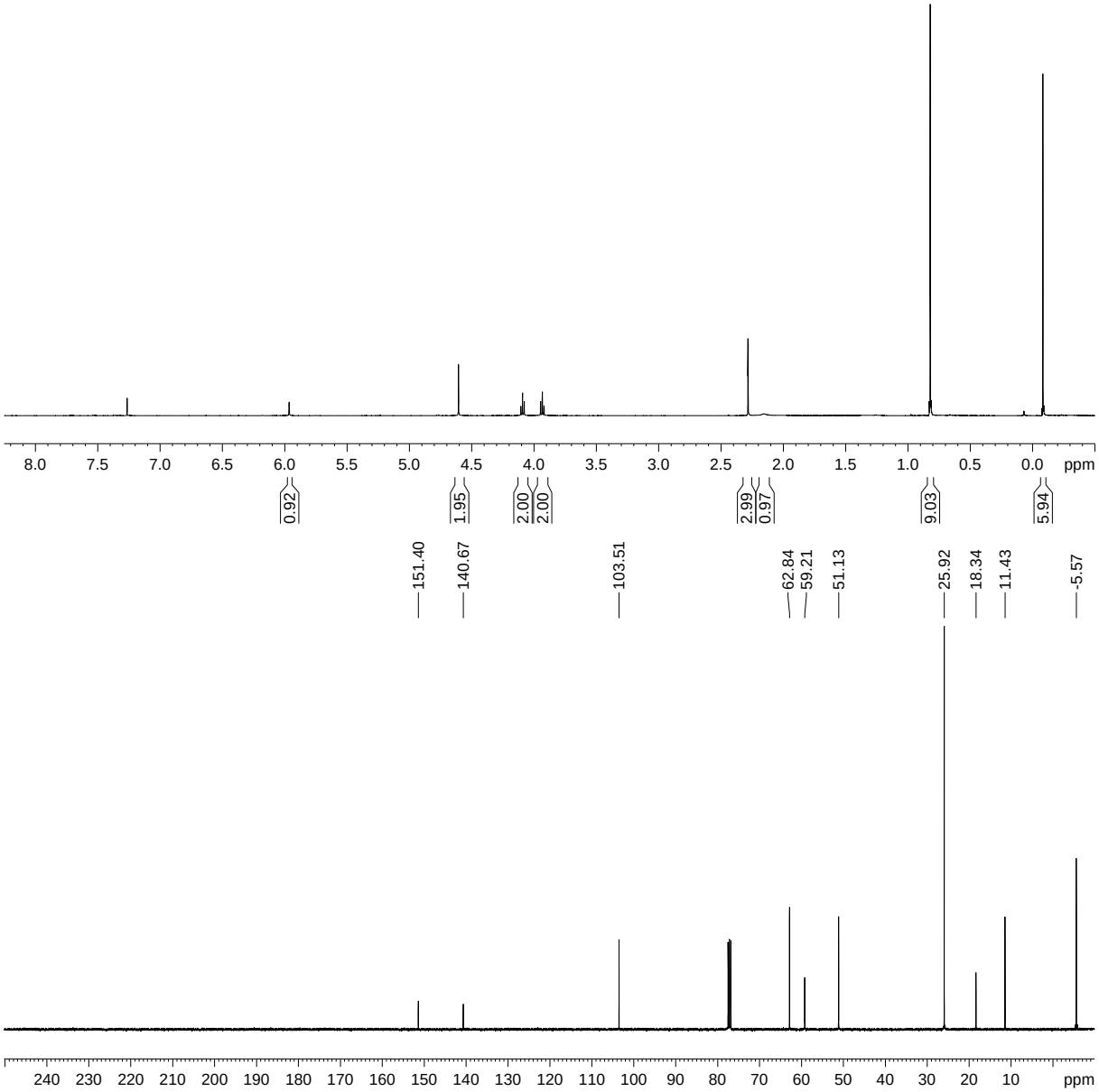
mp: 45-47 °C.

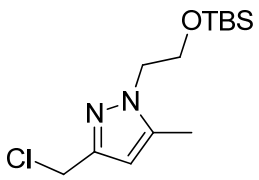
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.96 (s, 1H), 4.60 (s, 2H), 4.09 (t, *J* = 5.4 Hz, 2H), 3.93 (t, *J* = 5.4 Hz, 2H), 2.28 (d, *J* = 0.7 Hz, 3H), 2.15 (br s, 1H), 0.82 (s, 9H), -0.08 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 151.4, 140.7, 103.5, 62.8, 59.2, 51.1, 25.9, 18.3, 11.4, -5.6.

IR (film): ν 3316, 2953, 2929, 2857, 1151, 1471, 1463, 1440, 1451, 1361, 1256, 1114, 1068, 1039, 1007, 924, 834, 780.

HRMS (EI): calculated for $C_{13}H_{27}N_2O_2Si$ $[M+H]^+$ 271.1836, found 271.1836.





203

1-(2-(*tert*-butyldimethylsilyloxy)ethyl)-3-(chloromethyl)-5-methyl-1*H*-pyrazole (203). To a solution of alcohol **202** (20 mg, 0.07 mmol), 2,6-lutidine (30 uL, 0.30 mmol) and LiCl (13 mg, 0.30 mmol) in DMF (0.5 mL) was added MsCl (17.2 uL, 0.22 mmol) dropwise at 0 °C.

The reaction mixture was stirred for 2 h at this temperature, was then allowed to reach room temperature and stirred for 1 h. The reaction mixture was then diluted with water (1 mL) and the aqueous phase was extracted with Et_2O (3 x 2 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to give 15.5 mg (73%) of chloride **203** as a colorless oil which turned into a white solid upon storage at -20 °C.

TLC: R_f 0.30 (hexane/EtOAc 9:1, $KMnO_4$).

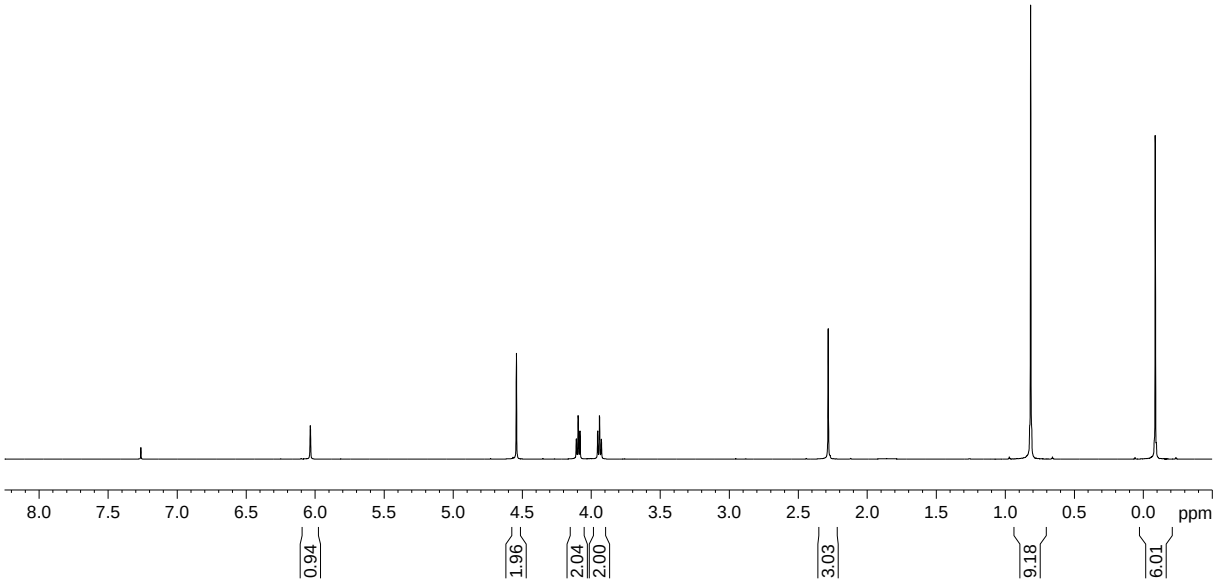
mp:

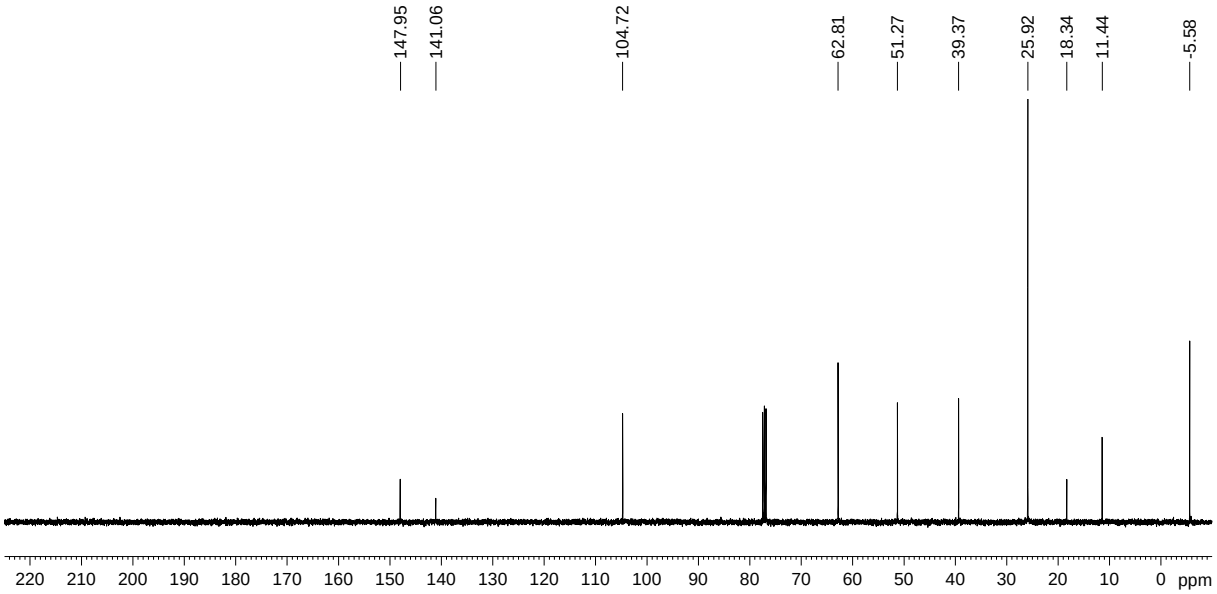
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.04 (s, 1H), 4.54 (s, 2H), 4.09 (t, *J* = 5.3 Hz, 2H), 3.94 (t, *J* = 5.3 Hz, 2H), 2.28 (d, *J* = 0.7 Hz, 3H), 0.82 (s, 9H), -0.09 (s, 6H).

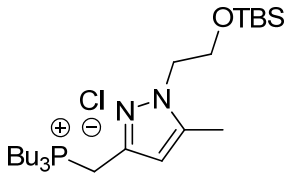
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 148.0, 141.6, 104.7, 62.8, 51.3, 39.4, 25.9, 18.3, 11.4, -5.6.

IR (film): ν 2953, 2929, 2857, 1552, 1471, 1463, 1438, 1400, 1389, 1362, 1258, 1166, 1112, 1063, 1019, 1006, 926, 830, 812, 778, 734, 663.

HRMS (EI): calculated for $C_{13}H_{26}ClN_2OSi$ $[M+H]^+$ 2891497, found 289.1495.







140c

Tributyl((1-(2-(*tert*-butyldimethylsilyloxy)ethyl)-5-methyl-1*H*-pyrazol-3-yl)methyl)phophonium chloride (140c). To a solution of chloride **203** (0.50 g, 1.73 mmol) in DMF (30 mL) was added Bu_3P (0.74 mL, 2.94 mmol) at room temperature and the reaction mixture was left stirring for 24 h at this temperature. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1 → 10:1) to give 0.78 g (98%) of Wittig salt **140c** as a colorless oil which turns into a white solid upon storage at -20 °C.

TLC: R_f 0.30 (CH_2Cl_2 /MeOH 10:1, CPS).

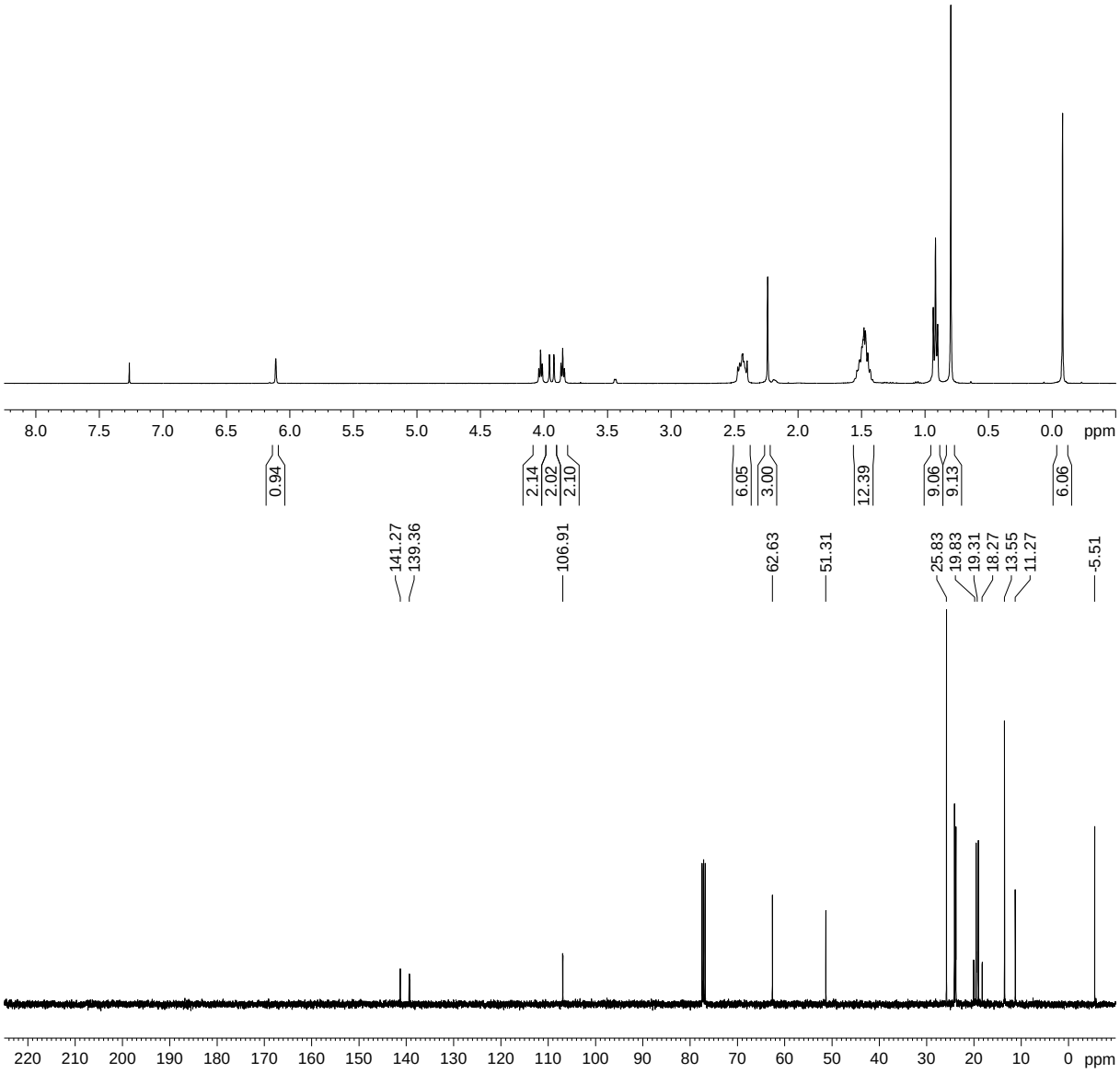
mp: 91-95 °C

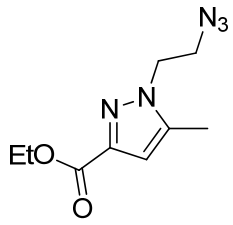
^{1}H-NMR (400 MHz, $CDCl_3$): δ 6.11 (s, 1H), 4.03 (t, *J* = 5.3 Hz, 2H), 3.94 (d, *J* = 14.2 Hz, 2H), 3.85 (t, *J* = 5.3 Hz, 2H), 2.49-2.38 (m, 6H), 2.24 (s, 3H), 1.56-1.40 (m, 12H), 0.92 (t, *J* = 7.0 Hz, 9H), 0.80 (s, 9H), -0.08 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ =

IR (film): ν 2956, 2929, 2872, 2858, 1548, 1463, 1453, 1398,1389, 1252, 1101, 930, 834, 812, 777, 724.

HRMS (ESI): calculated for $C_{25}H_{52}N_2OPSi$ $[M]^+$ 455.3581, found 455.3589.





204

Ethyl 1-(2-azidoethyl)-5-methyl-1*H*-pyrazole-3-carboxylate (204). To a solution of alcohol **198** (0.60 g, 3.03 mmol) and NEt_3 (1.26 mL, 9.09 mmol) in CH_2Cl_2 (30 mL) was added MsCl (0.35 mL, 4.55 mmol) dropwise at 0 °C. The reaction mixture was stirred for 1 h at this temperature and was then cautiously quenched by addition of saturated aqueous NH_4Cl (30 mL) The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were washed with brine (100 mL), dried over $MgSO_4$ and concentrated under reduced pressure. The residue was dissolved in DMF (30 mL)

and sodium azide (187 mg, 2.87 mmol) was added in one portion at room temperature. The reaction mixture was heated to 80 °C, stirred for 14 h at this temperature and was then cooled to room temperature. Water (30 mL) was added and the aqueous phase was extracted with EtOAc (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to give 0.64 g (96%) of chloride **204** as a yellow oil

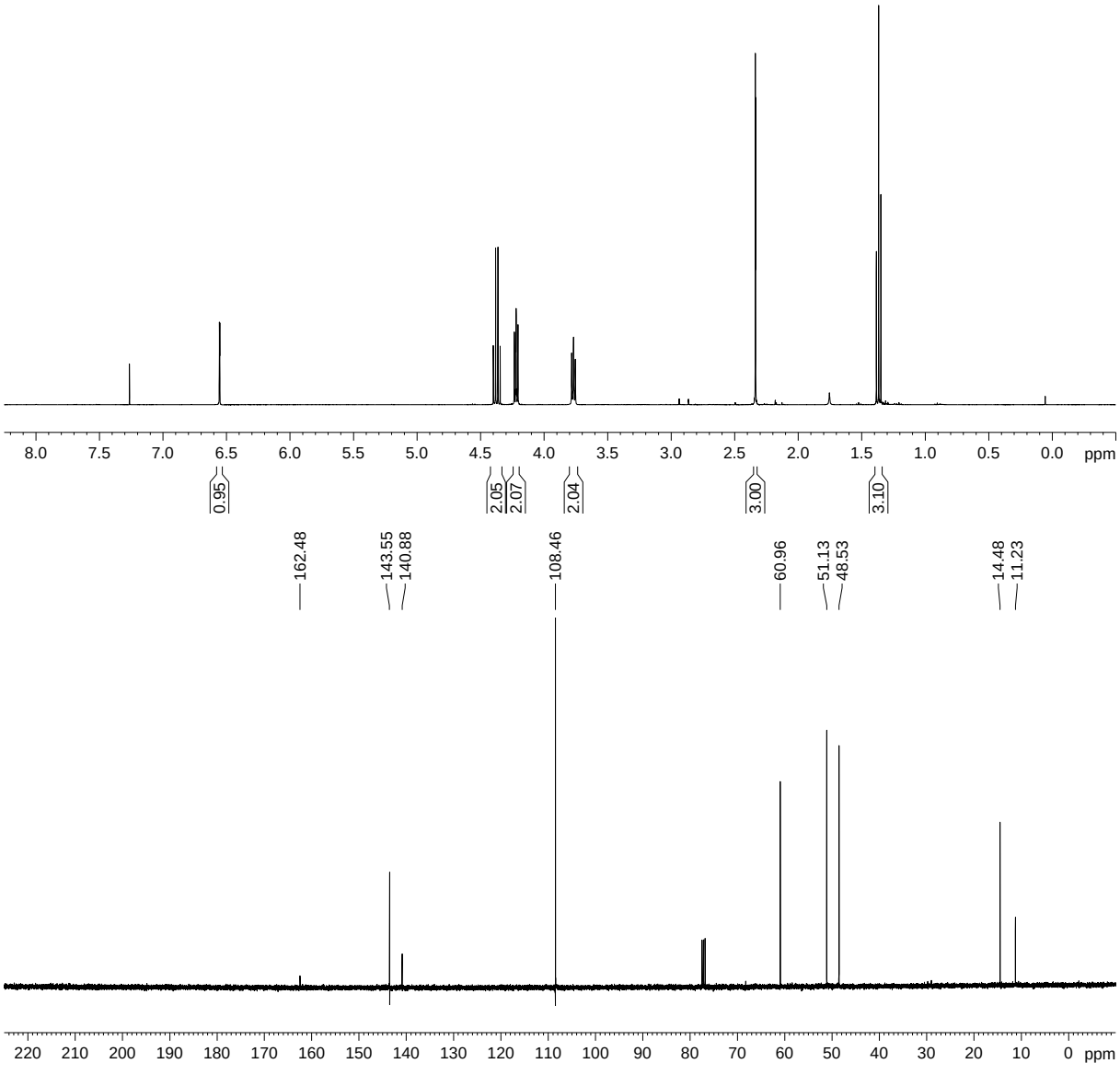
TLC: R_f 0.45 (hexane/EtOAc 1:1, UV).

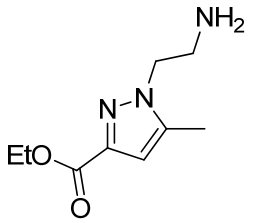
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.56 (d, *J* = 0.7 Hz, 1H), 4.37 (q, *J* = 7.1 Hz, 2H), 4.22 (t, *J* = 5.8 Hz, 2H), 3.77 (t, *J* = 5.8 Hz, 2H), 2.34 (d, *J* = 0.7 Hz, 3H), 1.37 (t, *J* = 7.1 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.5, 142.7, 140.1, 108.3, 60.7, 52.3, 41.9, 14.4, 11.2.

IR (film): ν 3347, 2980, 2955, 2939, 1716, 1657, 1536, 1443, 1427, 1386, 1300, 1217, 1105, 1028, 980, 915, 840, 816, 777, 729.

HRMS (EI): calculated for $C_9H_{14}N_5O_2$ $[M+H]^+$ 224.1142, found 224.1139.





205

Ethyl 1-(2-aminoethyl)-5-methyl-1*H*-pyrazole-3-carboxylate (205). To a solution of azide **204** (31 mg, 0.14 mmol) in EtOH (1 mL) was added Palladium on activated carbon (15 mg, 0.014 mmol, 10%) in one portion at room temperature and the reaction mixture was stirred under atmosphere of H_2 for 22h. The solution was then filtered through a small plug of celite and the precipitate was rinsed with EtOH (5 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc/MeOH/NEt_3 17:2:1) to afford 24 mg (88%) of amine **205** as a yellow oil.

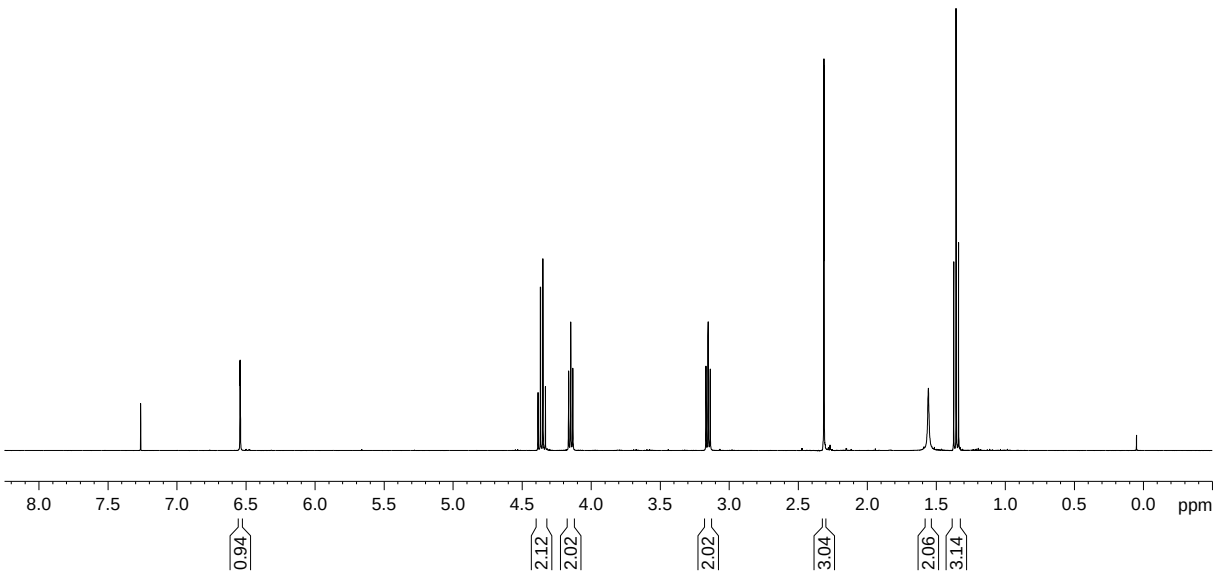
TLC: R_f 0.23 (EtOAc/MeOH/NEt_3 17:2:1, UV, Ninhydrin).

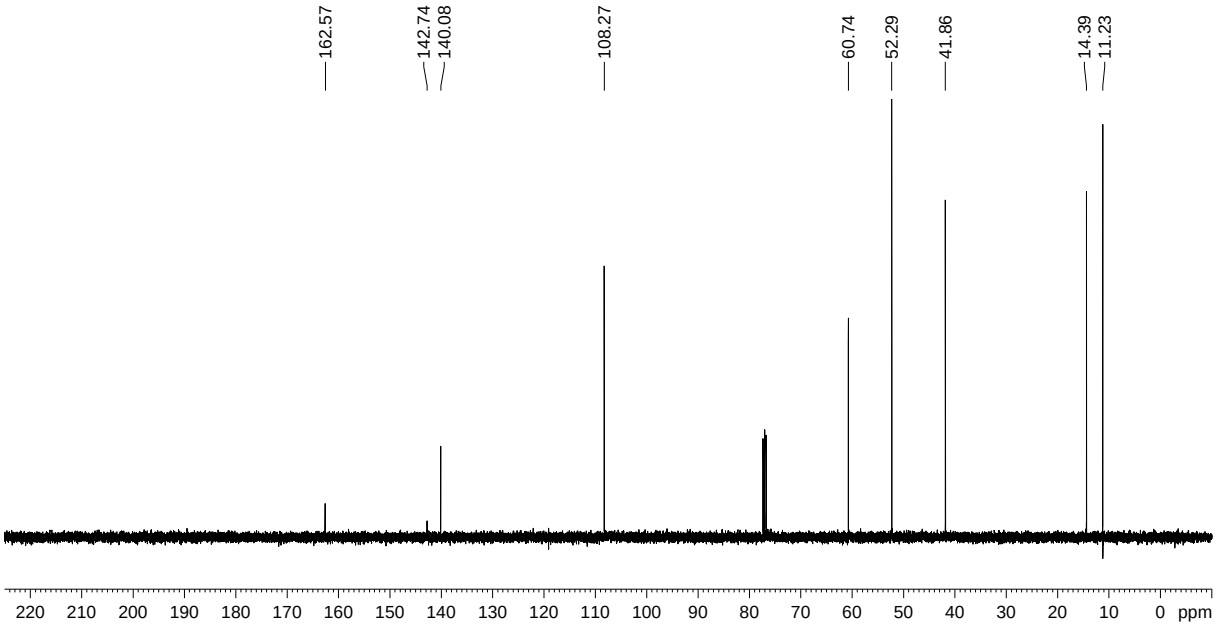
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.54 (q, *J* = 0.7 Hz, 1H), 4.36 (q, *J* = 7.1 Hz, 2H), 4.15 (t, *J* = 6.1 Hz, 2H), 3.15 (t, *J* = 6.1 Hz, 2H), 2.31 (d, *J* = 0.7 Hz, 3H), 1.56 (br s, 2H), 1.36 (t, *J* = 7.1 Hz, 3H).

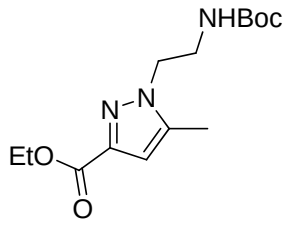
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.5, 143.5, 140.9, 108.5, 61.0, 51.1, 48.5, 14.5, 11.2.

IR (film): ν 2982, 2960, 2934, 2100, 1714, 1550, 1442, 1428, 1390, 1348, 1296, 1213, 1138, 1105, 1056, 1026, 981, 844, 819, 778.

HRMS (EI): calculated for $C_9H_{16}N_3O_2$ $[M+H]^+$ 198.1237, found 198.1234.







206

Ethyl 1-(2-((*tert*-butoxycarbonyl)amino)ethyl)-5-methyl-1*H*-pyrazole-3-carboxylate (206). To a solution of amine **205** (290 mg, 1.47 mmol) and NEt_3 (1.02 mL, 7.35 mmol) in CH_2Cl_2 (12 mL) was added Boc_2O (353 mg, 1.62 mmol) at room temperature and the reaction mixture was stirred for 5 h. The reaction mixture was then quenched by addition of saturated aqueous NH_4Cl (10 mL) and the aqueous phase was extracted with EtOAc (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc1:1) to yield 380 mg (87%) of Boc protected amine **206** as a white solid.

TLC: R_f 0.25 (hexane/EtOAc 1:1, UV, Ninhydrin, CPS).

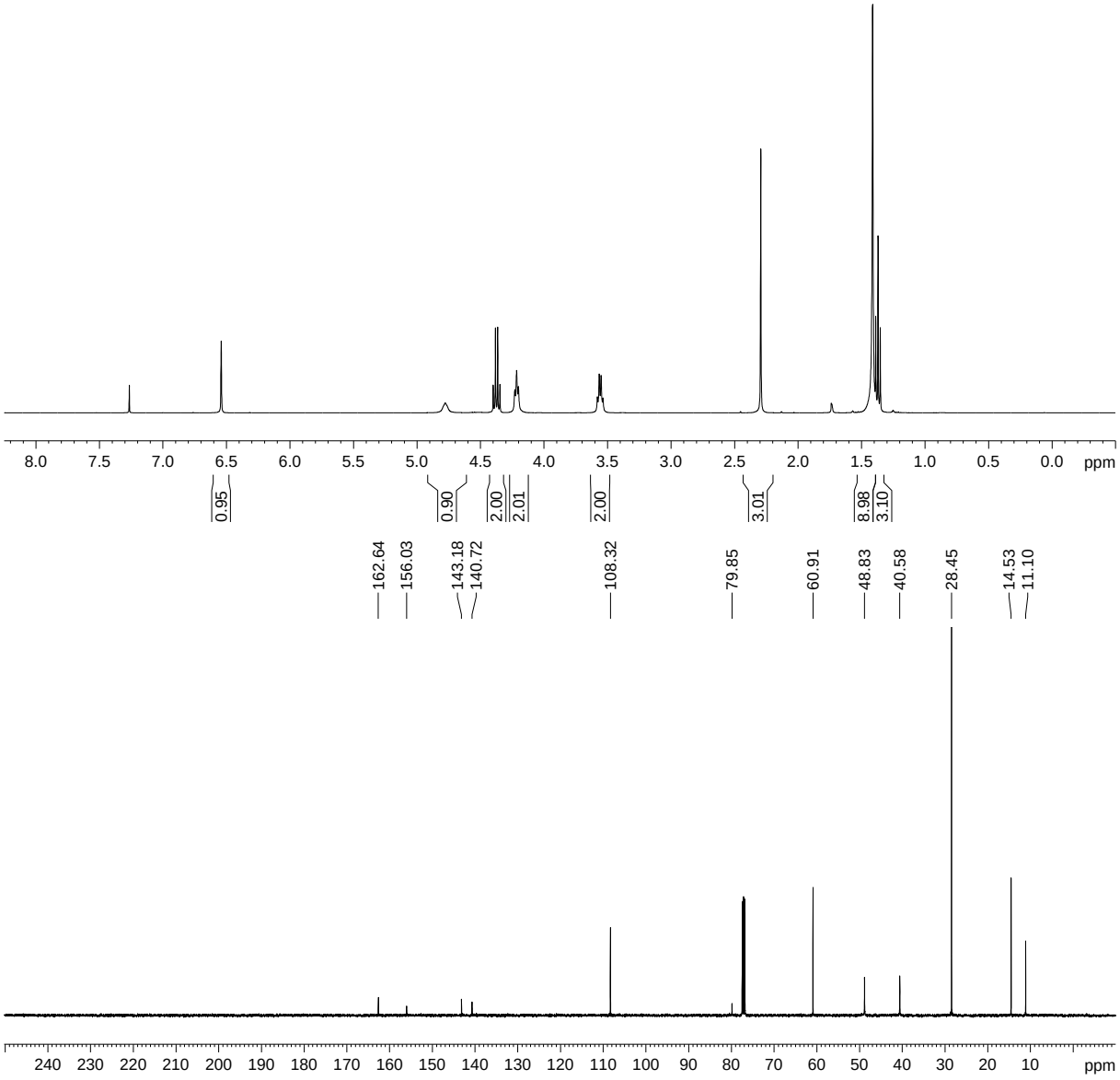
mp: 105-106 °C

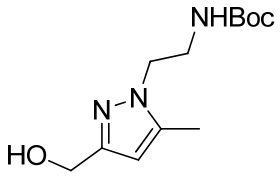
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.54 (s, 1H), 4.78 (t, *J* = 5.3 Hz, 1H), 4.37 (q, *J* = 7.1 Hz, 2H), 4.22 (t, *J* = 5.9 Hz, 2H), 3.56 (q, *J* = 5.9 Hz, 2H), 2.29 (s, 3H), 1.41 (s, 9H), 1.37 (t, *J* = 7.1 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 162.6, 156.0, 143.2, 140.7, 108.3, 79.9, 60.9, 48.8, 40.6, 28.5, 14.5, 11.1.

IR (film): ν 3356, 2978, 2935, 1706, 1515, 1447, 1390, 1365, 1250, 1213, 1165, 1106, 1065, 1028, 982, 859, 777.

HRMS (EI): calculated for $C_{14}H_{23}N_3NaO_4$ $[M+Na]^+$ 320.1581, found 320.1579.





207

***tert*-Butyl (2-(3-(hydroxymethyl)-5-methyl-1*H*-pyrazol-1-yl)ethyl)carbamate (207).** To a solution of ester **206** (295 mg, 0.99 mmol) in CH_2Cl_2 (17 mL) was added DIBAL-H (1 M in CH_2Cl_2, 2.98 mL, 2.98 mmol) dropwise at -78 °C. The reaction mixture was stirred for 0.5 h at this temperature, was then allowed reach room tempeature slowly and stirred for 2 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous

Rochelle salt (30 mL). The solution was left stirring rigorously until the two phases got transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 20 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (EtOAc → EtOAc/MeOH 20:1) to give 221 g (87%) of alcohol **207** as a colorless oil.

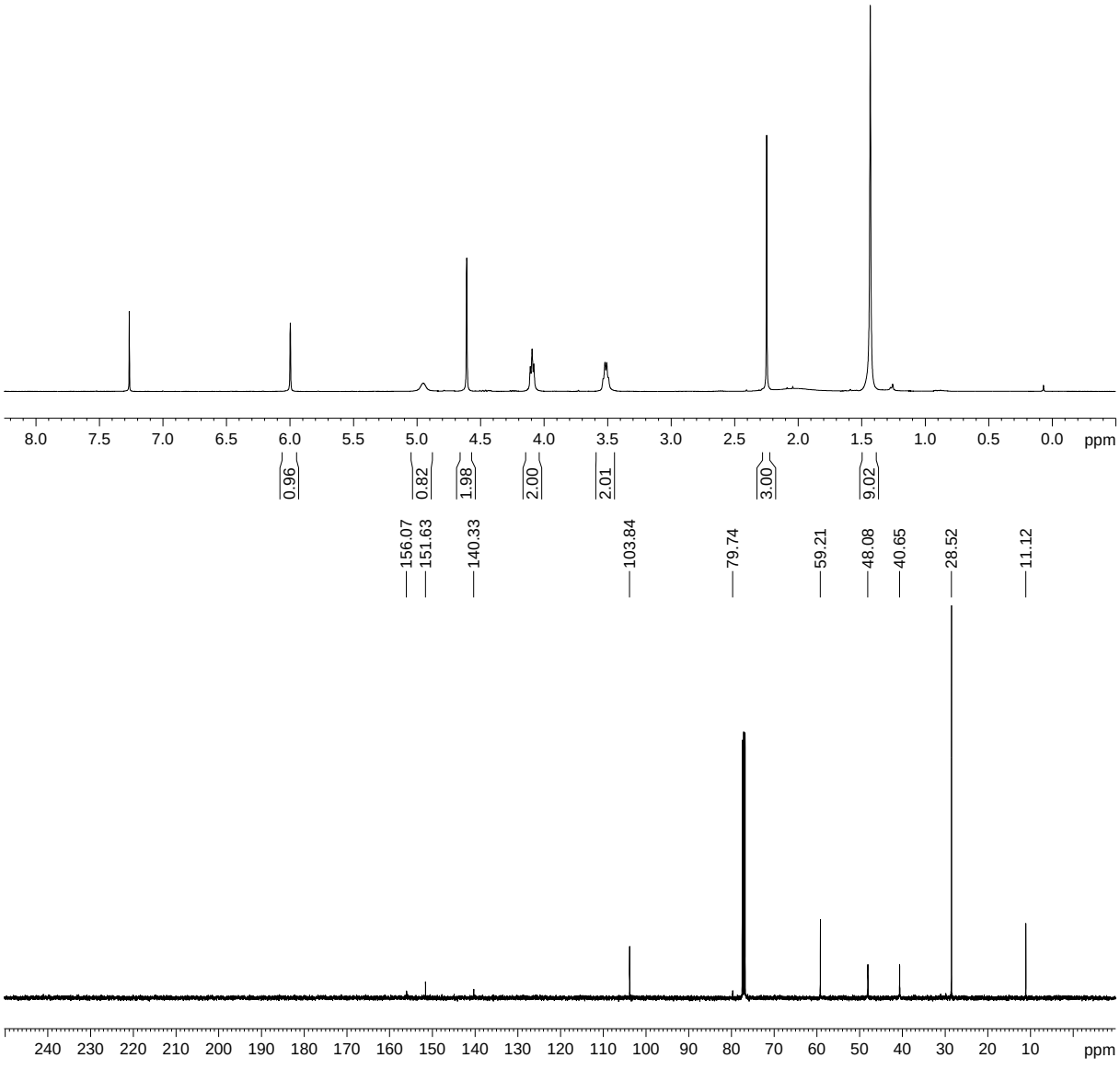
TLC: R_f 0.12 (EtOAc, $KMnO_4$).

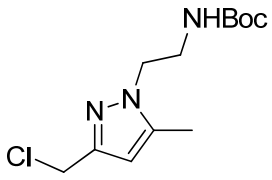
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.00 (s, 1H), 4.95 (br s, 1H), 4.61 (s, 2H), 4.09 (t, *J* = 5.6 Hz, 1H), 3.52 (q, *J* = 5.6 Hz, 2H), 2.25 (s, 3H), 1.43 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 156.1, 151.6, 140.3, 103.8, 79.7, 59.2, 48.1, 40.6, 28.5, 11.1.

IR (film): ν 3327, 2976, 2929, 2871,1697, 1520, 1510, 1455, 1394, 1252, 1170, 1075, 1040, 1007, 985, 859, 785.

HRMS (EI): calculated for $C_{12}H_{21}N_3NaO_3$ $[M+Na]^+$ 278.1475, found 278.1474.





208

***tert*-Butyl (2-(3-(chloromethyl)-5-methyl-1*H*-pyrazol-1-yl)ethyl)carbamate (208).** To a solution of alcohol **207** (60 mg, 0.48 mmol) in CH_2Cl_2 (4 mL) was added thionyl chloride (36 µL, 0.50 mmol) dropwise at 0 °C and the reaction mixture was stirred for 45 min at this temperature. The reaction mixture was then quenched by addition of saturated aqueous Na_2CO_3 (5 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 2:1) to give 48 mg (75%) of chloride **208** as a white solid.

TLC: R_f 0.23 (hexane/EtOAc 2:1, UV, $KMnO_4$).

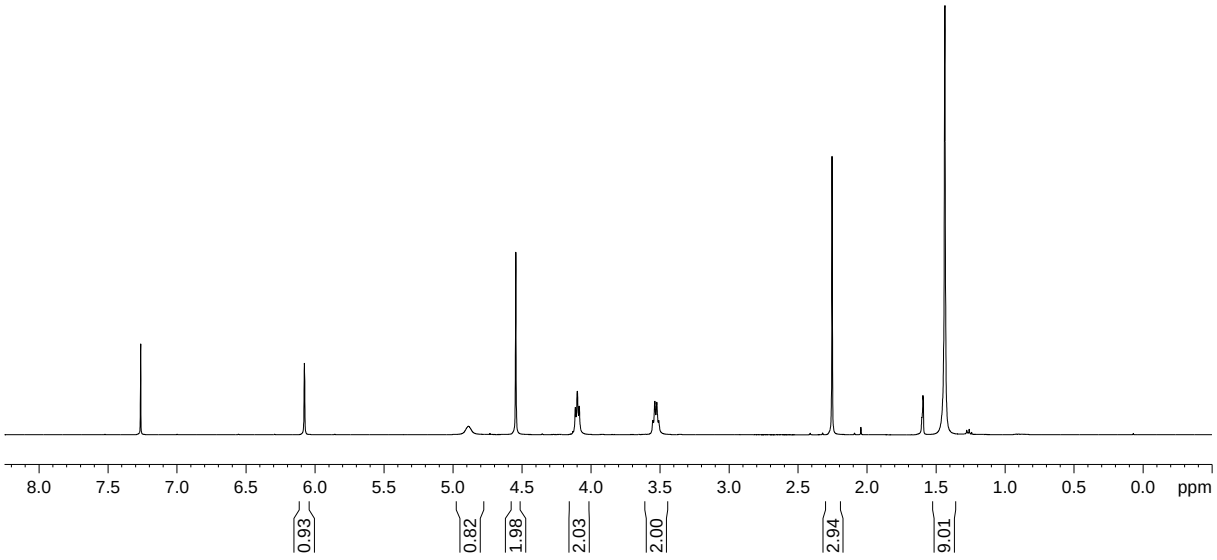
mp: 107-108 °C

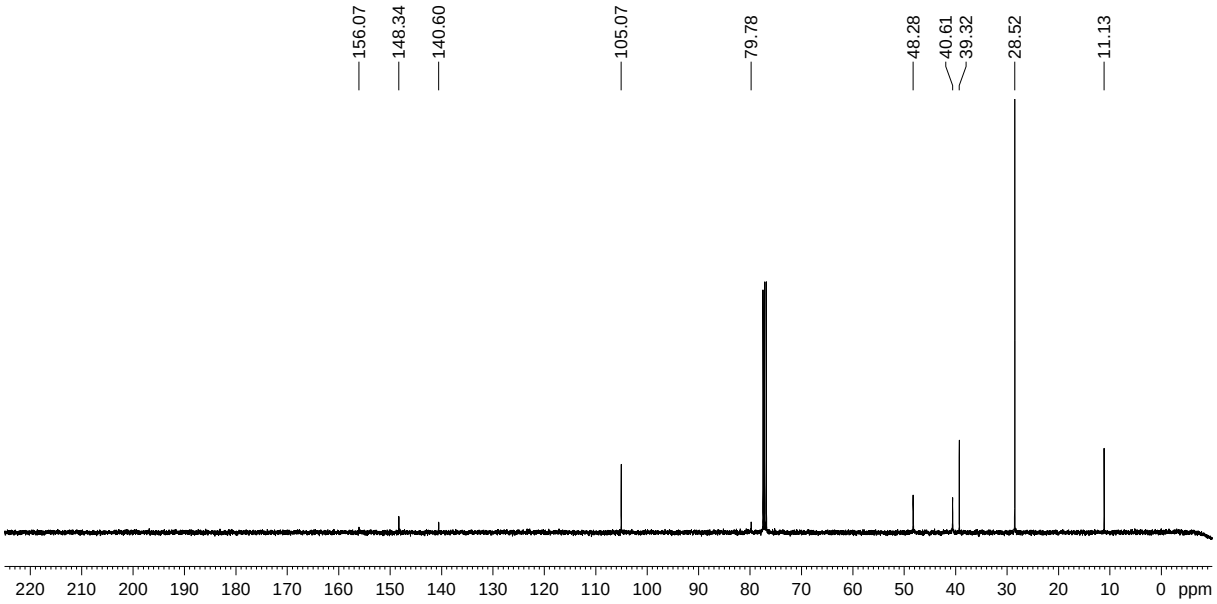
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.08 (s, 1H), 4.89 (br s, 1H), 4.55 (s, 2H), 4.10 (t, *J* = 5.6 Hz, 1H), 3.53 (q, *J* = 5.6 Hz, 2H), 2.25 (s, 3H), 1.44 (s, 9H).

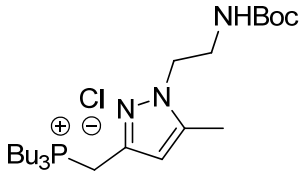
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 156.1, 148.3, 140.6, 105.1, 79.8, 48.3, 40.6, 39.3, 28.5, 11.1.

IR (film): ν 3301, 2980, 2969, 2939, 1701, 1553, 1532, 1456, 1393, 1365, 1317, 1280, 1266, 1255, 1172, 1146, 1069,1023, 959, 872, 803, 736, 713, 658.

HRMS (EI): calculated for $C_{12}H_{21}ClN_3O_2$ $[M+H]^+$ 274.1317, found 274.1316.







209

((1-(2-((*tert*-butoxycarbonyl)amino)ethyl)-5-methyl-1*H*-pyrazol-3-yl)methyl)tributylphosphonium chloride (209). To a solution of chloride **208** (13 mg, 0.05 mmol) in DMF (0.3 mL) was added Bu_3P (59 µL, 0.24 mmol) at room temperature. The reaction mixture was heated to 40 °C and stirred for 72 h at this temperature. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1 → 10:1) to give 21 mg (93%) of Wittig salt **209** as a colorless oil.

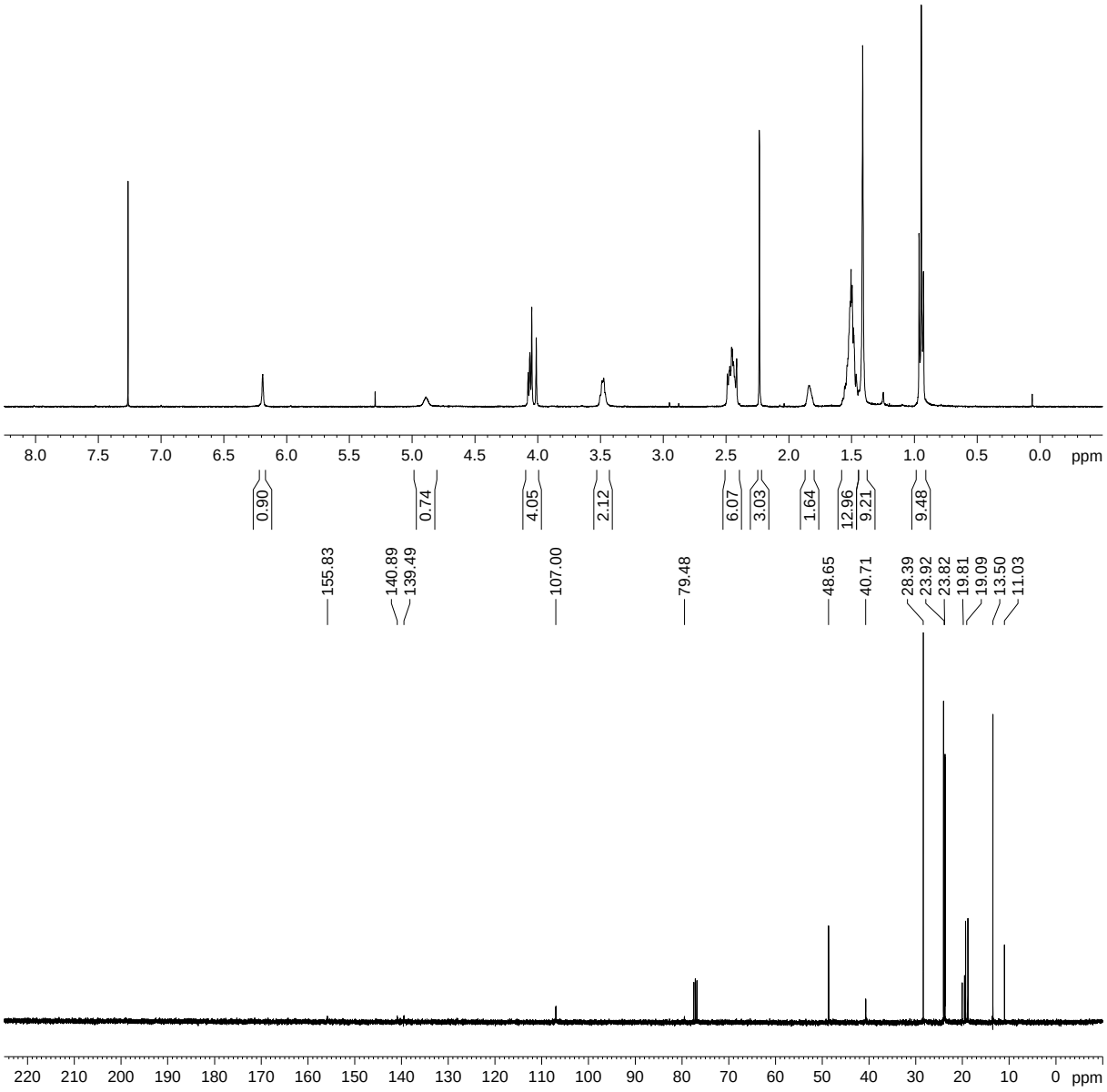
TLC: R_f 0.39 (CH_2Cl_2 /MeOH 9:1, CPS).

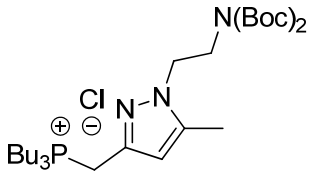
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.09 (s, 1H), 5.12 (d, *J* = 5.4 Hz, 1H), 4.03 (t, *J* = 5.6 Hz, 2H), 3.93 (d, *J* = 14.1 Hz, 2H), 3.43 (q, *J* = 5.6 Hz, 2H), 2.44-2.33 (m, 12H), 2.20 (s, 6H), 1.53-1.40 (m, 12H), 1.35 (s, 9H), 0.90 (t, *J* = 7.1 Hz, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 155.8, 140.9, 139.5 (d, *J* = 8.3 Hz), 107.0 (d, *J* = 4.0 Hz), 79.5, 48.7, 40.7, 28.4, 23.9 (d, *J* = 27.1 Hz), 23.8 (d, *J* = 16.8 Hz), 19.8 (d, *J* = 48.4 Hz), 19.1 (d, *J* = 47.1 Hz), 13.5, 11.0.

IR (film): ν 3337, 3236, 2960, 2932, 2873,1705, 1546, 1514, 1454, 1390, 1364, 1274, 1250, 1173, 1126, 991, 969, 917, 780, 724.

HRMS (EI): calculated for $C_{24}H_{47}N_3O_2P$ $[M]^+$ 440.3400, found 440.3401.





145g

((1-(2-((di-*tert*-butoxycarbonyl)amino)ethyl)-5-methyl-1*H*-pyrazol-3-yl)methyl)tributylphosphonium chloride (145g). To a solution of phosphonate **209** (53 mg, 0.11 mmol) in MeCN (1.5 mL) was added Boc_2O (49 mg, 0.22 mmol) and DMAP (7 mg, 0.06 mmol) at room temperature and the reaction mixture was stirred for 12 h. The reaction mixture was then quenched by addition of saturated aqueous NH_4Cl (2 mL) and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over

$MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 20:1) to yield 186 mg (97%) of double-Boc protected pyrazole **145g** as a slightly yellow oil.

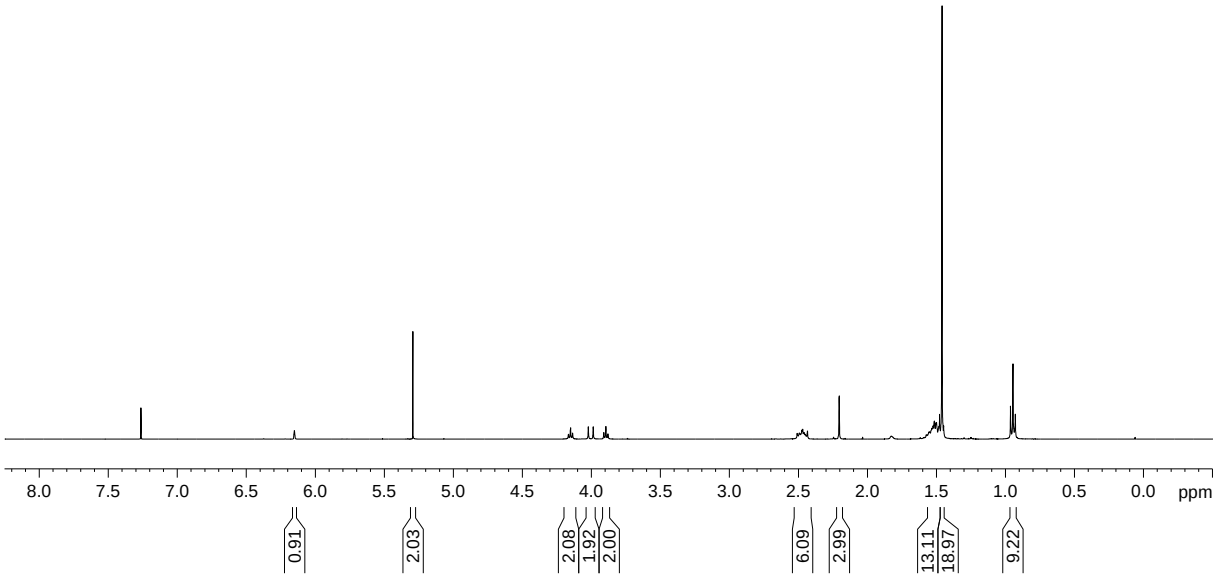
TLC: R_f 0.11 (CH_2Cl_2 /MeOH 20:1, CPS).

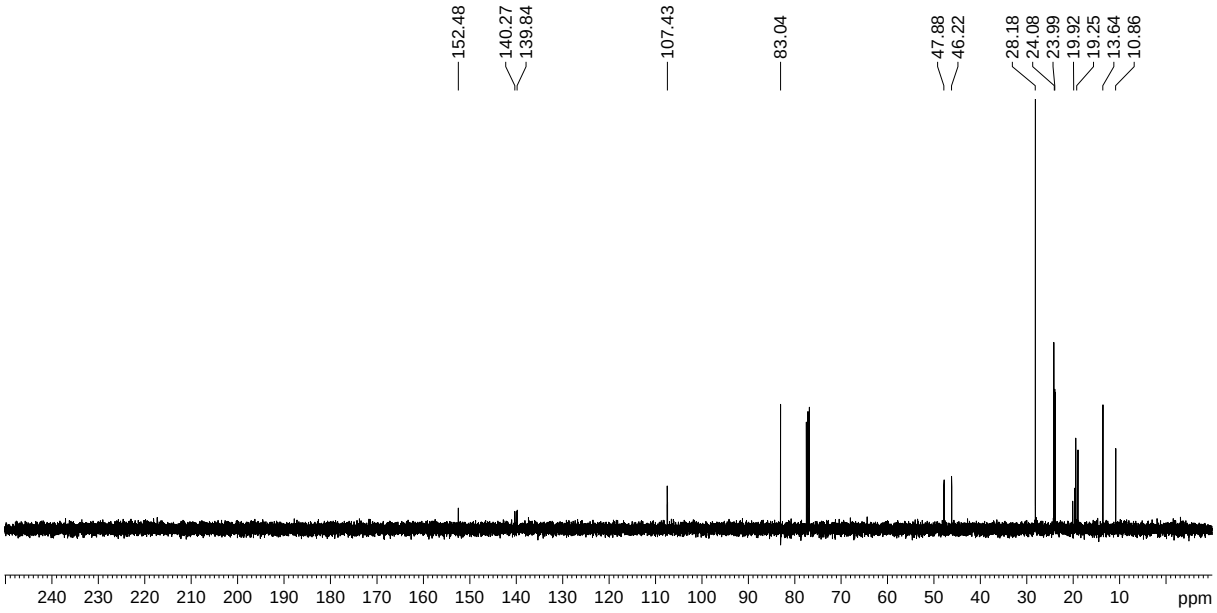
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.15 (s, 1H), 5.29 (s, 2H), 4.15 (t, *J* = 6.0 Hz, 2H), 4.00 (d, *J* = 14.3 Hz, 2H), 3.89 (t, *J* = 6.0 Hz, 2H), 2.51-2.43 (m, 6H), 2.20 (s, 3H), 1.57-1.47 (m, 12H), 1.46 (s, 18H), 0.94 (t, *J* = 7.0, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 152.5, 140.3, 139.8 (d, *J* = 9.0 Hz), 107.4 (d, *J* = 5.3 Hz), 83.0, 47.9, 46.2, 28.2, 24.1 (d, *J* = 20.9 Hz), 24.0 (d, *J* = 10.4 Hz), 19.9 (d, *J* = 48.7 Hz), 19.3 (d, *J* = 47.0 Hz), 13.6, 10.9.

IR (film): ν 2960, 2933, 2873, 1787, 1736, 1696, 1549, 1456, 1394, 1367, 1345, 1248, 1218, 1170, 11.35, 849.

HRMS (ESI): calculated for $C_{29}H_{55}N_3O_4P$ $[M]^+$ 540.3925, found 540.3919.

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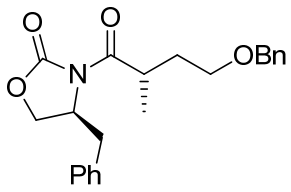
140.27
139.84
107.43
83.04
47.88
46.22
28.18
24.08
23.99
19.92
19.25
13.64
10.86
240 230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 ppm

4.3 Synthesis of Hypermodified Epothilone A Analogs

4.3.1 First Synthesis



241

(*S*)-4-benzyl-3-((*S*)-4-(benzyloxy)-2-methylbutanoyl)oxazolidin-2-one (241). To a solution of acyloxazolidinone (**240**) (8.81 g, 24.93 mmol) in THF (60 mL) was added NaHMDS (1 M in THF, 33 mL, 32.41 mmol) dropwise at -78 °C and the reaction mixture was stirred for 1 h at this temperature. Iodomethane (9.3 mL, 149.6 mmol) in THF (40 mL) was then added over a period of 1 h at -78 °C and the reaction mixture was stirred for 1 h at this temperature. The reaction mixture was then allowed to reach room temperature over a period of 1 h and stirred for additional 0.5 h at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (40 mL). The phases were separated and the aqueous phase was extracted with Et_2O (2 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 6:1) to yield 8.07 g (88%) of methylated acyloxazolidinone **241** as a colorless oil.

TLC: R_f 0.45 (hexane/EtOAc 3:1, UV, CPS).

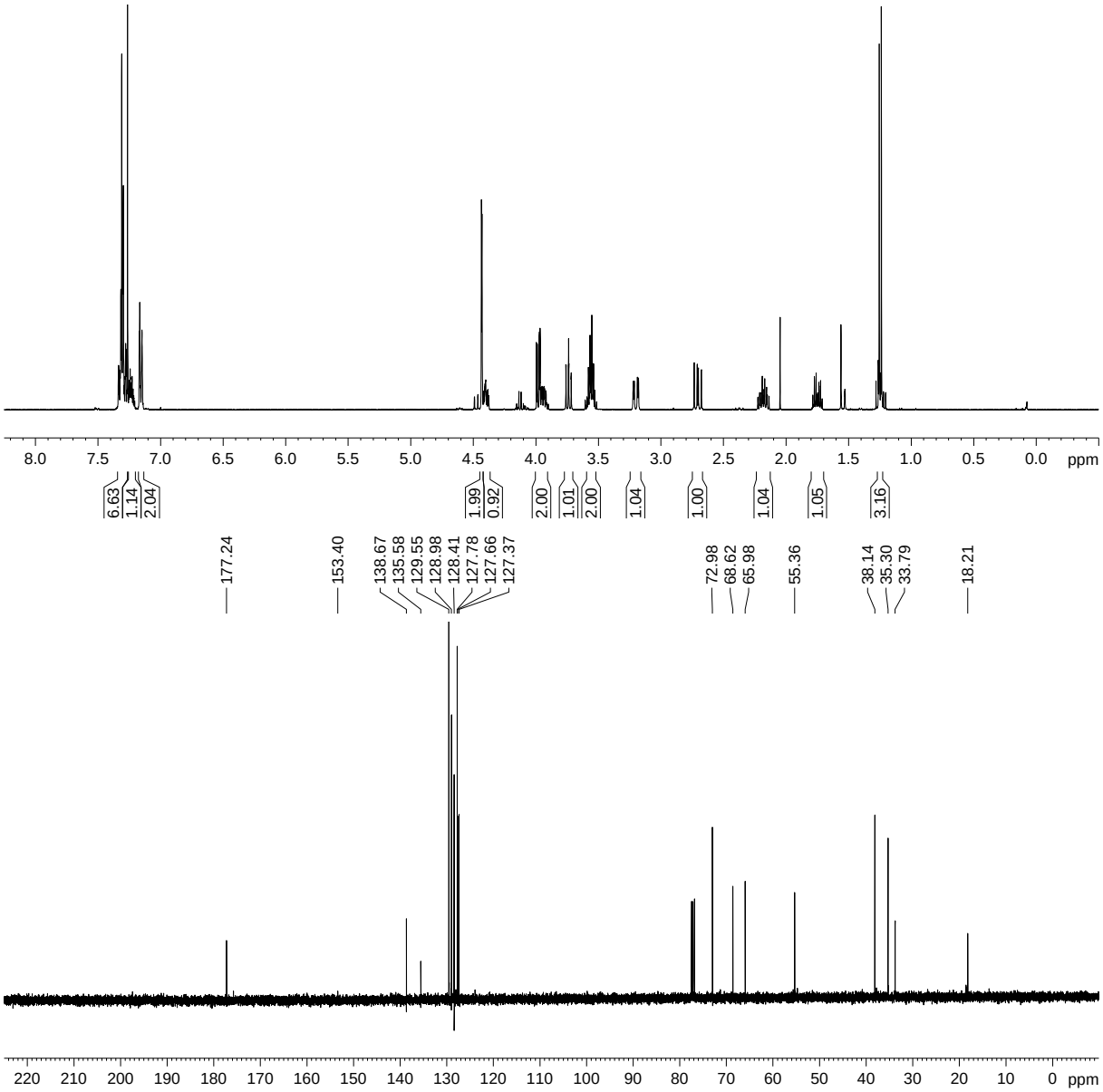
$[\alpha]^{20}_D$: = +61.1 (*c* = 0.82, $CHCl_3$).

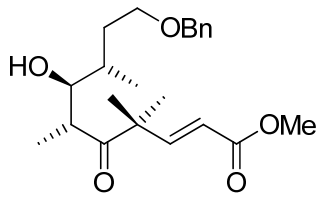
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.35-7.19 (m, 8H), 7.18-7.13 (m, 2H), 4.43 (d, *J* = 1.9 Hz, 2H) 4.42-4.36 (m, 1H), 3.98 (dd, *J* = 9.0, 2.7 Hz, 1H), 3.94 (dddd, *J* = 13.3, 8.3, 7.0, 5.0 Hz, 1H), 3.73 (ddd, *J* = 8.8, 8.0, 0.7 Hz, 1H), 3.61-3.51 (m, 2H), 3.20 (dd, *J* = 13.4, 3.3 Hz, 1H), 2.70 (dd, *J* = 13.4, 9.6 Hz, 1H), 2.24-2.13 (m, 1H), 1.75 (dq, *J* = 14.1, 5.3 Hz, 1H), 1.25 (d, *J* = 7.0 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 177.2, 153.4, 138.7, 135.6, 129.6, 129.0, 128.4, 127.8, 127.7, 127.4, 73.0, 68.6, 66.0, 55.4, 38.1, 35.3, 33.8, 18.2.

IR (film): ν □3030, 2967, 2926, 2862, 1776, 1695, 1496, 1455, 1385, 1351, 1289, 1241, 1206, 1094, 1015, 973, 741, 700, 595.

HRMS (ESI): calculated for $C_{22}H_{25}NNaO_4$ $[M+Na]^+$ 390.1676, found 390.1672.





248

(6*R*,7*S*,8*S*,*E*)-methyl 10-(benzyloxy)-7-hydroxy-4,4,6,8-tetramethyl-5-oxodec-2-enoate (248). To a solution of ethyl ketone **83** (1.82 g, 9.91 mmol) in CH_2Cl_2 (30 mL) was added $TiCl_4$ (1 M in CH_2Cl_2, 9.91 mL, 9.91 mmol) and DIPEA (1.94 mL, 11.23 mmol) dropwise at -78 °C. The deep red reaction mixture was stirred for 1 h at this temperature. Aldehyde **232** (1.27 g, 6.61 mmol) in CH_2Cl_2 (10 mL) was then added over a period of 0.5 h and the reaction mixture was stirred for 0.5 h at -78 °C. The reaction was then cautiously quenched by

addition pH 7 phosphate buffer (60 mL), the mixture was filtered through a filter paper and the precipitate was rinsed extensively with CH_2Cl_2 (150 mL). The phases of the filtrate were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 6:1 → 5:1) to yield 1.40 g (56%) of aldol product **248** as a colorless oil.

TLC: R_f 0.26 (hexane/EtOAc 3:1, CPS).

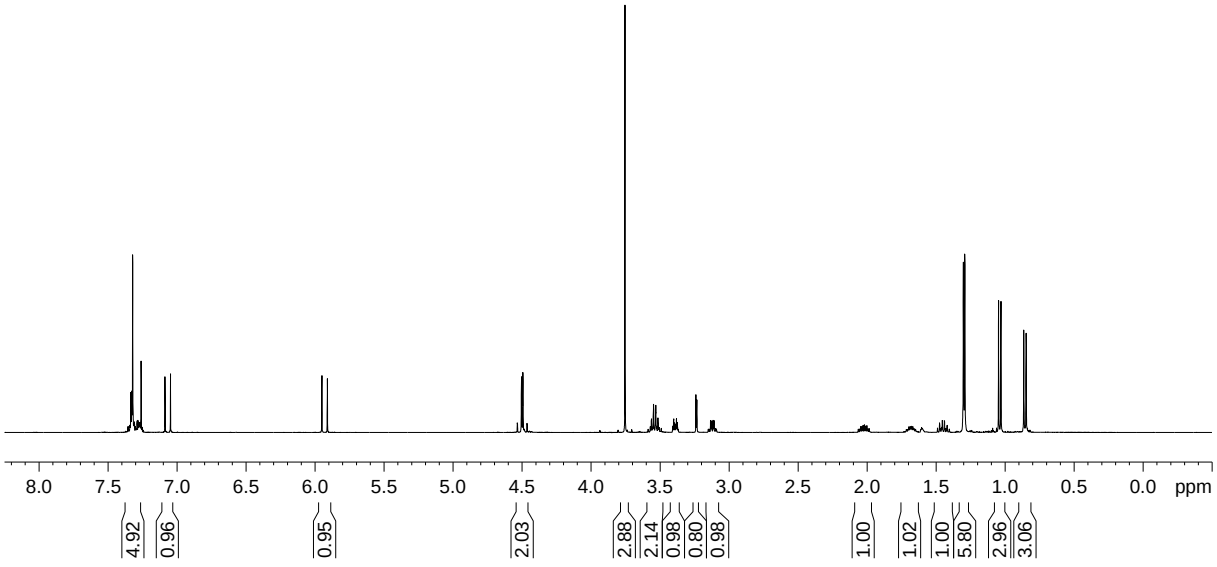
$[\alpha]^{20}_D$: = -13.1 (*c* = 0.82, $CHCl_3$).

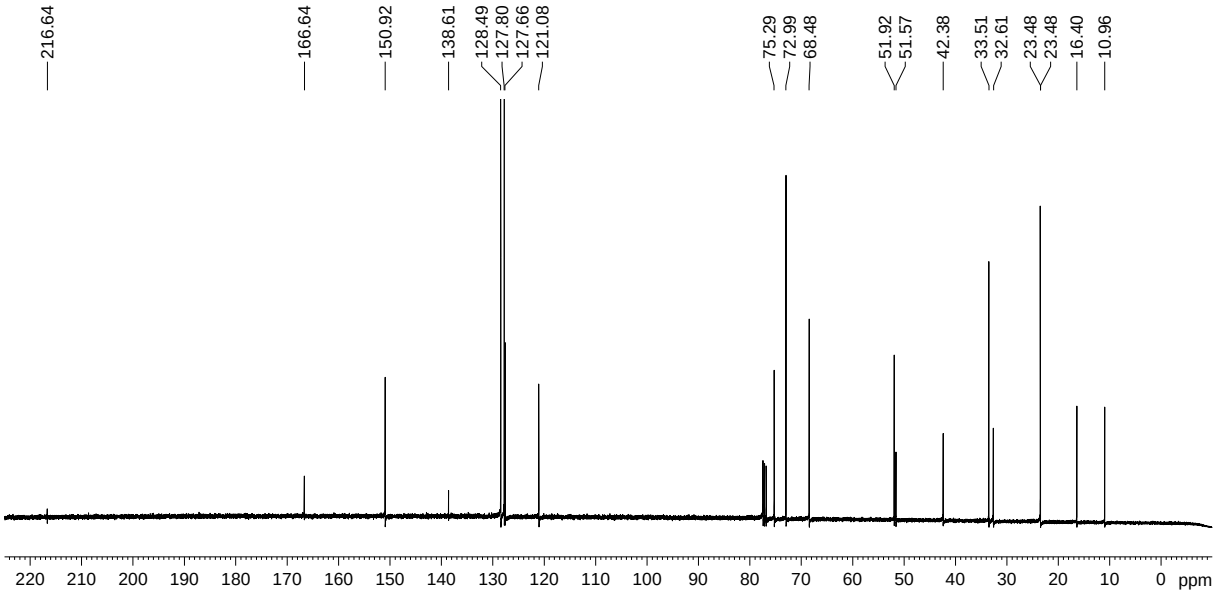
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.35-7.24 (m, 5H), 7.07 (d, *J* = 15.9 Hz, 1H), 5.93 (d, *J* = 15.9 Hz, 1H), 4.52 (d, *J* = 12.0 Hz, 1H), 4.48 (d, *J* = 12.0 Hz, 1H), 3.75 (s, 3H), 3.56 (ddd, *J* = 9.4, 6.5, 6.1 Hz, 1H), 3.52 (ddd, *J* = 9.4, 7.1, 6.2 Hz, 1H), 3.39 (dt, *J* = 8.3, 2.6 Hz, 1H), 3.24 (d, *J* = 2.6 Hz, 1H), 3.12 (qd, *J* = 6.9, 2.6 Hz, 1H), 2.02 (dddd, *J* = 14.0, 7.1, 6.5, 4.0 Hz, 1H), 1.68 (dqd, *J* = 8.3, 6.8, 4.0 Hz, 1H), 1.45 (dddd, *J* = 14.0, 8.3, 6.2, 6.1 Hz, 1H), 1.30 (s, 3H), 1.29 (s, 3H), 1.04 (d, *J* = 6.9 Hz, 3H), 0.86 (d, *J* = 6.8 Hz, 3H).

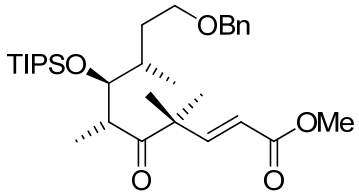
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 216.6, 166.6, 150.9, 138.6, 128.5, 127.8, 127.7, 121.1, 75.3, 73.0, 68.5, 51.9, 51.6, 42.4, 33.5, 32.6, 23.5, 23.5, 16.4, 11.0.

IR (film): ν □3521, 2969, 2937, 2874, 1724, 1702, 1647, 1456, 1437, 1366, 1315, 1298, 1281, 1200, 1177, 1096, 986, 739, 700.

HRMS (ESI): calculated for $C_{22}H_{32}NaO_5$ $[M+Na]^+$, 399.2142, found 399.2128.







249

(6*R*,7*S*,8*S*,*E*)-methyl 10-(benzyloxy)-4,4,6,8-tetramethyl-5-oxo-7 ((triisopropylsilyl)oxy)dec-2-enoate (249). To a solution of alcohol **248** (0.35 g, 0.92 mmol) in CH_2Cl_2 (2 mL) was added 2,6-lutidine (0.74 mL, 6.41 mmol) and TIPSOTf (0.62 mL, 2.29 mmol) dropwise at -78 °C. The reaction mixture was allowed to reach room temperature and stirred overnight. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (3 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 9:1) to yield 0.46 g (94%) of silyl ether **249** as a colorless oil.

TLC: R_f 0.37 (hexane/EtOAc 9:1, UV, CPS).

$[\alpha]^{20}_D$: = -2.0 (*c* = 0.69, $CHCl_3$).

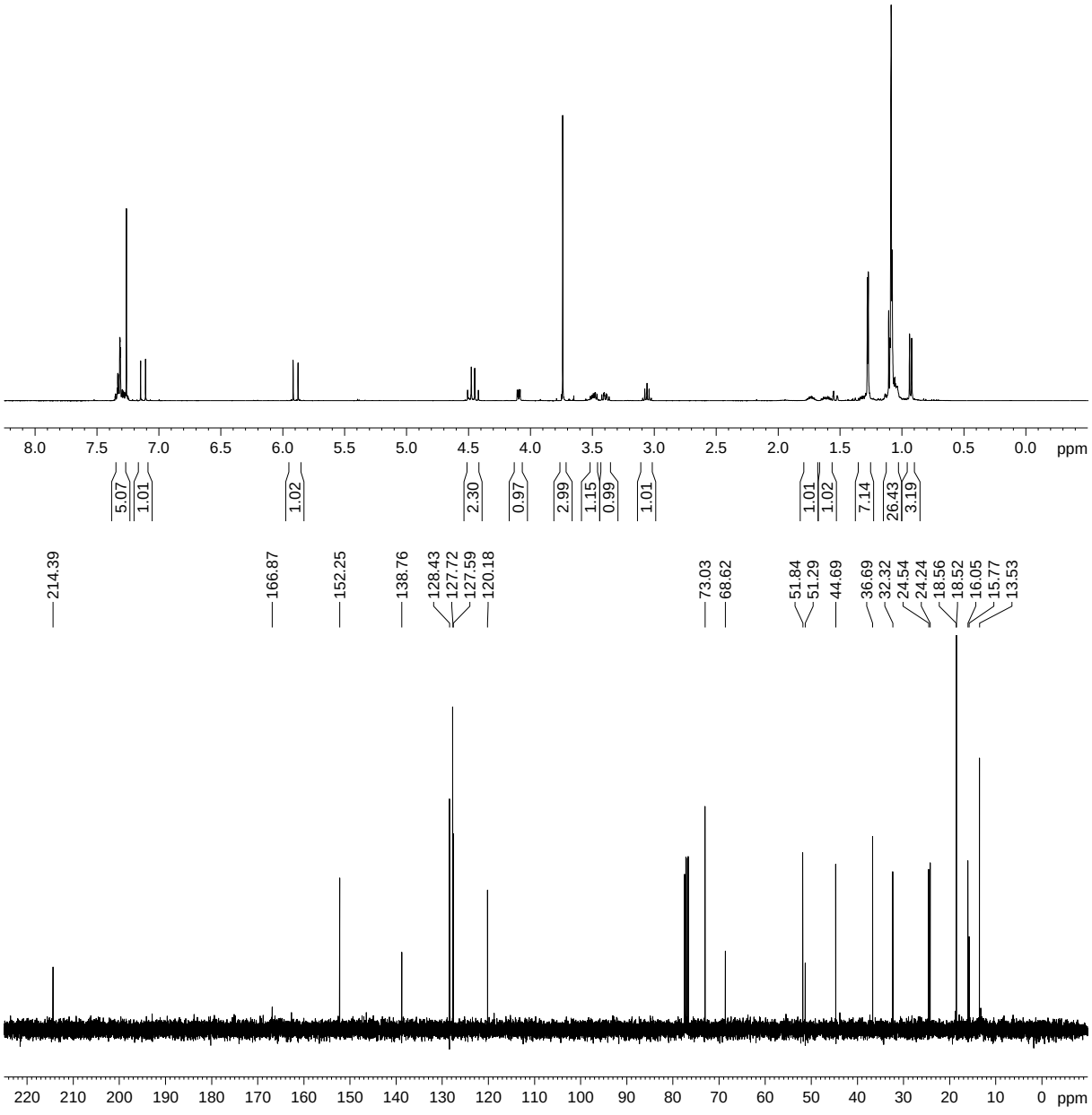
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.35-7.24 (m, 5H), 7.07 (d, *J* = 15.9 Hz, 1H), 5.93 (d, *J* = 15.9 Hz, 1H), 4.49 (d, *J* = 12.0 Hz, 1H), 4.43 (d, *J* = 12.0 Hz, 1H), 4.09 (dd, *J* = 6.8, 2.6 Hz, 1H), 3.74 (s, 3H), 3.49 (ddd, *J* = 9.3, 6.9, 5.0 Hz, 1H), 3.39 (ddd, *J* = 9.3, 7.3, 6.3 Hz, 1H), 3.06 (dq, *J* = 7.1, 6.8 Hz, 1H), 1.74 (dqd, *J* = 13.6, 6.8, 2.6 Hz, 1H), 1.61 (dddd, *J* = 13.6,

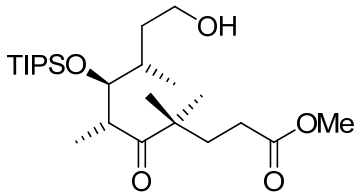
7.3, 6.9, 3.9 Hz, 1H), 1.35-1.28 (m, 1H), 1.28 (s, 3H), 1.27 (s, 3H), 1.10 (d, J = 6.9 Hz, 3H), 1.09 (s, 18H), 1.08 (s, 3H), 1.04 (d, J = 6.9 Hz, 3H), 0.93 (d, J = 6.9 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 214.4, 166.9, 152.2, 138.8, 128.4, 127.7, 127.6, 120.2, 73.0, 68.6, 51.8, 51.3, 44.7, 36.7, 32.3, 24.5, 24.2, 18.6, 18.5, 16.0, 15.8, 13.5.

IR (film): ν 2943, 2866, 1727, 1708, 1647, 1459, 1384, 1365, 1313, 1298, 1276, 1198, 1175, 1098, 1016, 988, 965, 883, 818, 737, 676

HRMS (ESI): calculated for $C_{31}H_{53}O_5Si$ $[M+H]^+$ 533.3657, found 533.3641





250

(6*R*,7*S*,8*S*)-methyl 10-hydroxy-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)decanoate (250). To a solution of benzyl protected alcohol **249** (0.36 g, 0.57 mmol) in MeOH (5 mL) was added Pd/C (10% palladium on activated charcoal, 0.12 g, 0.11 mmol) in one portion at room temperature and the reaction mixture was stirred under an atmosphere of H_2 for 0.5 h. The suspension was then filtered through a small plug of celite and the precipitate was rinsed with EtOAc (20 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to afford 0.24 g (93%) of alcohol **250** as a colorless oil.

TLC: R_f 0.21 (hexane/EtOAc 3:1, CPS).

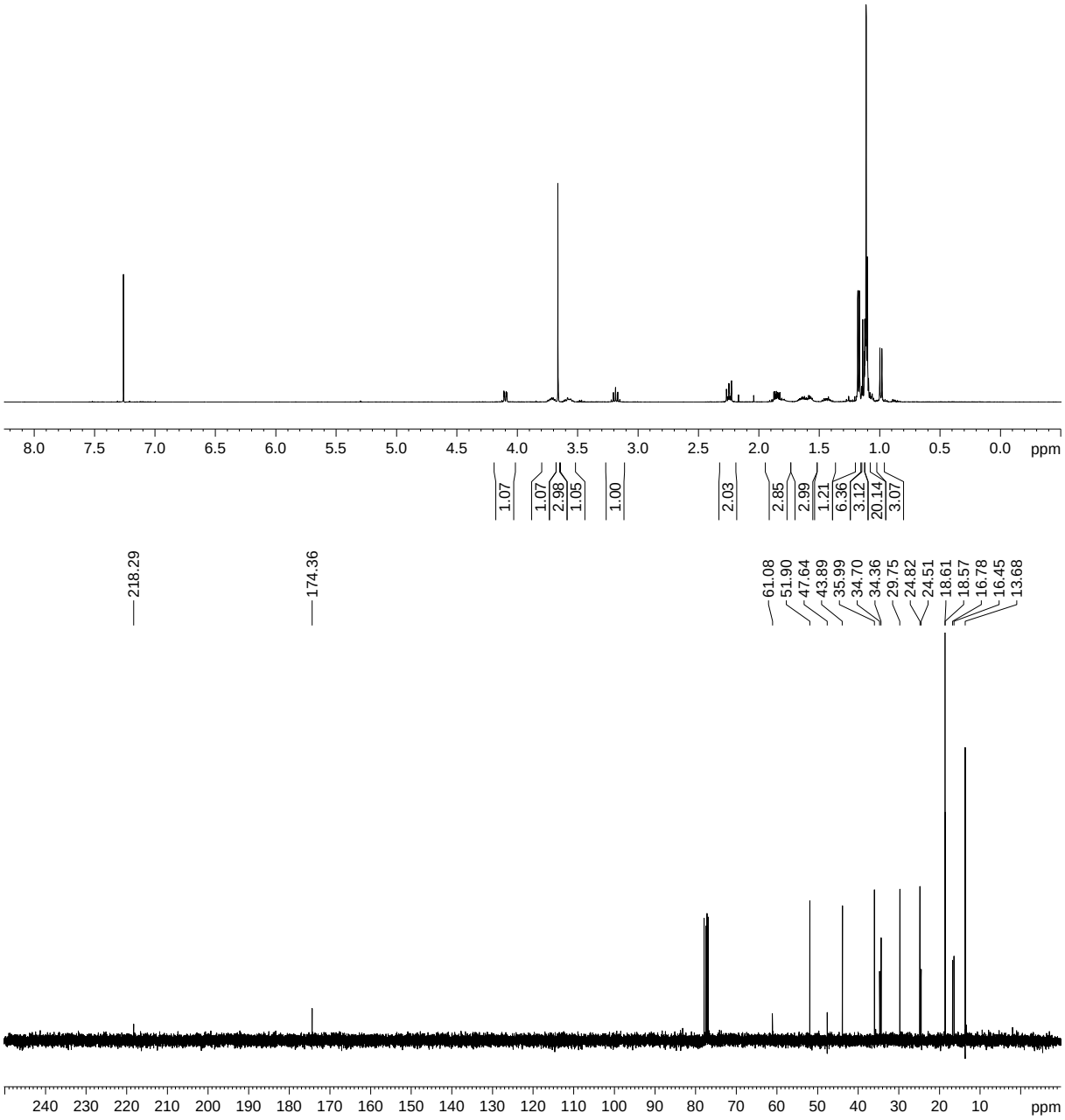
$[\alpha]^{20}_D$: = -2.4 (*c* = 0.71, $CHCl_3$).

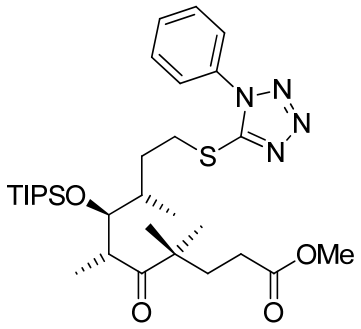
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.10 (dd, *J* = 7.3, 2.0 Hz, 1H), 3.72 (ddd, *J* = 10.8, 7.1, 5.0 Hz, 1H), 3.66 (s, 3H), 3.57 (ddd, *J* = 10.8, 7.1, 7.0 Hz, 1H), 3.19 (dq, *J* = 7.3, 6.9 Hz, 1H), 2.26 (d, *J* = 7.4 Hz, 1H), 2.24 (dd, *J* = 7.4, 4.6 Hz, 1H), 1.89-1.82 (m, 2H), 1.80 (br s, 1H), 1.69-1.59 (m, 2H), 1.57 (dd, *J* = 7.4, 3.8 Hz, 1H), 1.18 (s, 3H), 1.17 (s, 3H), 1.13 (d, *J* = 7.0 Hz, 3H), 1.18 (s, 18H), 1.10 (s, 3H), 0.99 (d, *J* = 6.9 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.3, 174.4, 61.1, 51.9, 47.6, 43.9, 36.0, 34.7, 34.4, 29.7, 24.8, 24.5, 18.6, 18.6, 16.8, 16.4, 13.7.

IR (film): ν □3466, 2944, 2867, 1740, 1696, 1463, 1371, 1296, 1257, 1202, 1159, 1111, 1054, 990, 883, 820, 676.

HRMS (ESI): calculated for $C_{24}H_{48}NaO_5Si$ $[M+Na]^+$ 467.3163, found 467.3161.


8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 ppm
1.07 1.07 2.98 1.05 1.00 2.03 2.85 2.99 1.21 6.36 3.12 20.14 3.07
218.29 174.36
61.08 51.90 47.64 43.89 35.99 34.70 34.36 29.75 24.82 24.51 18.61 18.57 16.78 16.45 13.68
240 230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 ppm



251

(6*R*,7*S*,8*S*)-methyl 4,4,6,8-tetramethyl-5-oxo-10-((1-phenyl-1*H*-tetrazol-5-yl)thio)-7-((triisopropylsilyl)oxy)decanoate (251). To a solution of alcohol **250** (1.51 g, 3.40 mmol) in THF (30 mL) was added 1-phenyl-1*H*-tetrazole-5-thiol (1.21 g, 6.80 mmol) and triphenyl phosphine (1.34 g, 5.10 mmol) in one portion at room temperature. The reaction mixture was then cooled to 0 °C and DEAD (0.94 mL, 5.95 mmol) was added dropwise. The reaction mixture was allowed to reach room temperature and stirred for 1 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (30 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to yield 1.97 g (96%) of sulfide **251** as a colorless oil.

TLC: R_f 0.39 (hexane/EtOAc 4:1, UV, CPS).

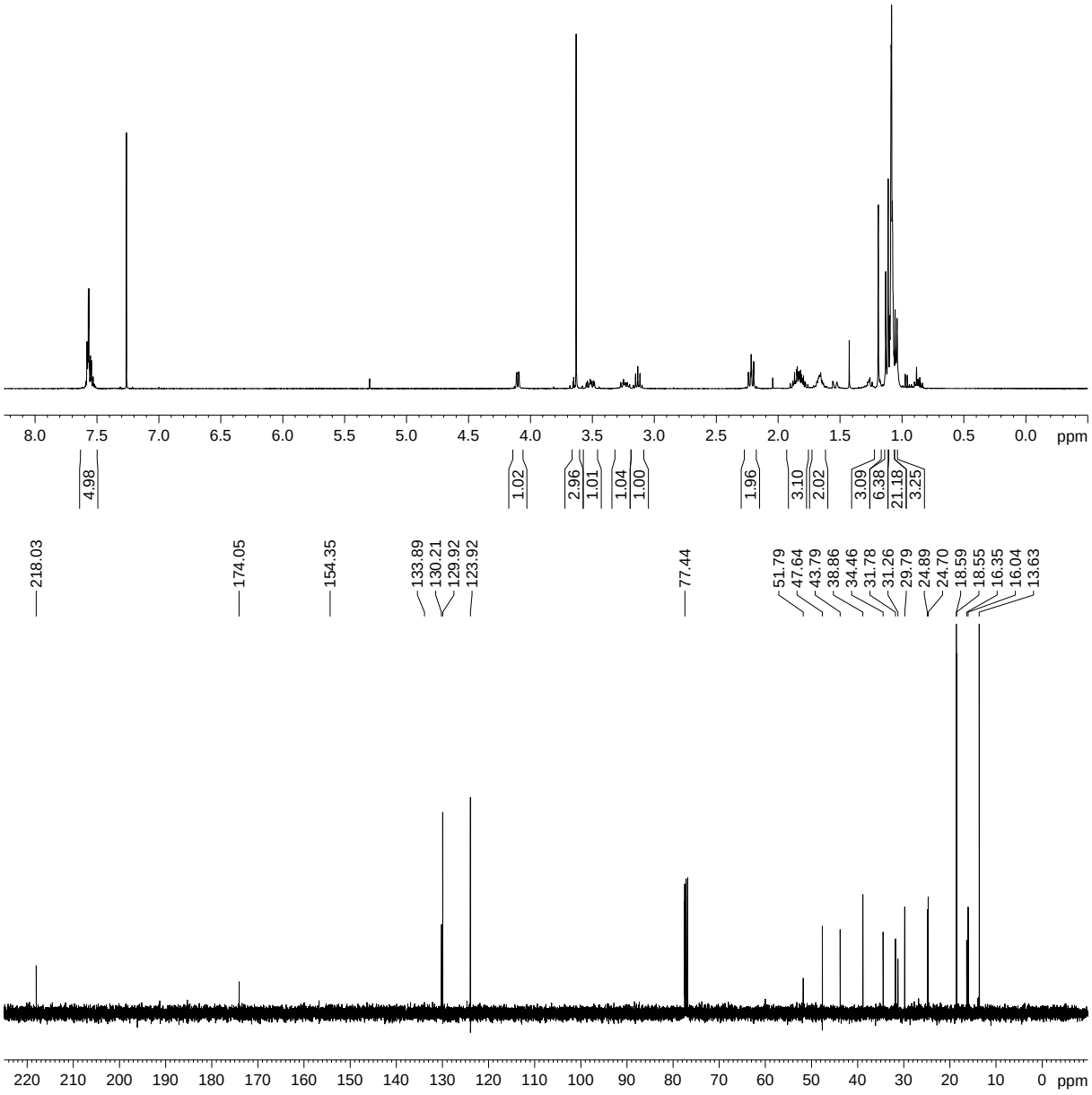
$[\alpha]^{20}_D$: = -14.6 (*c* = 0.44, $CHCl_3$).

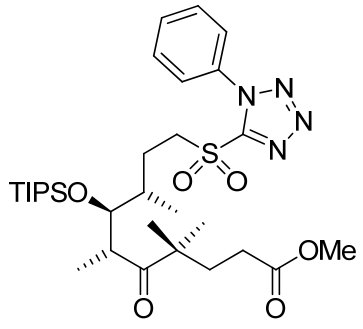
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.63-7.49 (m, 5H), 4.10 (dd, *J* = 7.1, 1.2 Hz, 1H), 3.63 (s, 3H), 3.52 (ddd, *J* = 13.1, 9.8, 4.4 Hz, 1H), 3.24 (ddd, *J* = 13.1, 8.7, 6.8 Hz, 1H), 3.13 (dq, *J* = 7.1, 6.9 Hz, 1H), 2.23 (dd, *J* = 6.9, 1.3 Hz, 1H), 2.21 (d, *J* = 6.9 Hz, 1H), 1.90-1.75 (m, 3H), 1.72-1.61 (m, 2H), 1.19 (s, 3H), 1.12 (d, *J* = 6.9 Hz, 3H), 1.11 (s, 3H), 1.10-1.06 (m, 21H), 1.05 (d, *J* = 6.9 Hz, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 154.3, 133.9, 130.2, 129.9, 123.9, 77.4, 51.8, 47.6, 43.8, 38.9, 34.5, 31.8, 31.3, 29.8, 24.9, 24.7, 18.6, 18.6, 16.3, 16.0, 13.6.

IR (film): ν □2945, 2867, 1738, 1695, 1597, 1501, 1463, 1386, 1295, 1244, 1200, 1174, 1091, 1011, 984, 884, 847, 820, 762, 678.

HRMS (ESI): calculated for $C_{31}H_{53}N_4O_4SSi$ $[M+H]^+$ 605.3551, found 605.3555.





227

(6*R*,7*S*,8*S*)-methyl 4,4,6,8-tetramethyl-5-oxo-10-((1-phenyl-1*H*-tetrazol-5-yl)sulfonyl)-7-((triisopropylsilyl)oxy)decanoate (251). To a solution of sulfide **227** (1.97 g, 3.26 mmol) in CH_2Cl_2 (30 mL) was added *m*-CPBA (2.81 g, 11.40 mmol) in one portion at room

temperature and the reaction mixture was stirred for 6 h. The reaction mixture was diluted with EtOAc (15 mL), washed successively 2M aqueous Na_2SO_3 solution (30 mL) and saturated aqueous $NaHCO_3$ (30 mL). The organic phase was dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to yield 1.96 g (94%) of sufon **251** as a viscous colorless oil.

TLC: R_f 0.39 (hexane/EtOAc 4:1, UV, CPS).

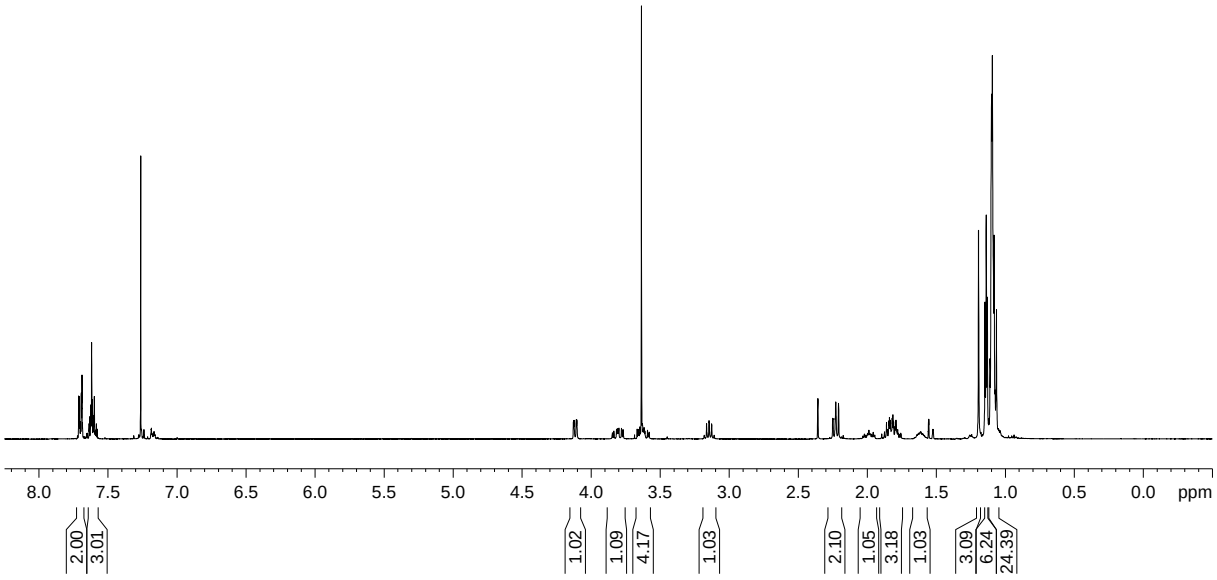
$[\alpha]^{20}_D$: = -10.9 (*c* = 0.61, $CHCl_3$).

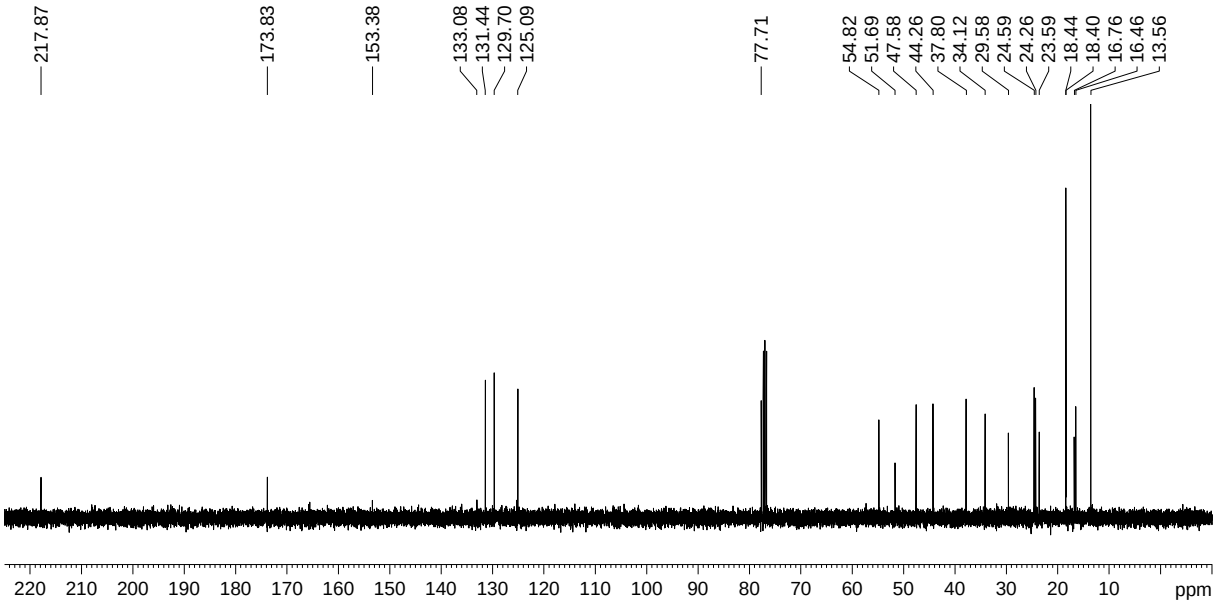
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.73-7.67 (m, 2H), 7.64-7.57 (m, 3H), 4.12 (dd, *J* = 8.0, 1.8 Hz, 1H), 3.81 (ddd, *J* = 14.3, 12.1, 4.5 Hz, 1H), 3.67-3.57 (m, 1H), 3.64 (s, 3H), 3.15 (dq, *J* = 8.1, 6.9 Hz, 1H), 2.24 (dd, *J* = 7.2, 0.9 Hz, 1H), 2.21 (d, *J* = 7.2 Hz, 1H), 1.99 (dddd, *J* = 13.3, 12.5, 4.2, 4.2 Hz, 1H), 1.86 (dd, *J* = 14.0, 8.1 Hz, 1H), 1.85 (dd, *J* = 14.0, 8.0 Hz, 1H), 1.84-1.78 (m, 1H), 1.61 (dddd, *J* = 10.4, 7.1, 3.9, 2.1 Hz, 1H), 1.19 (s, 3H), 1.14 (d, *J* = 6.9 Hz, 3H), 1.14 (s, 3H), 1.12-1.08 (m, 21H), 1.07 (d, *J* = 6.9 Hz, 3H).

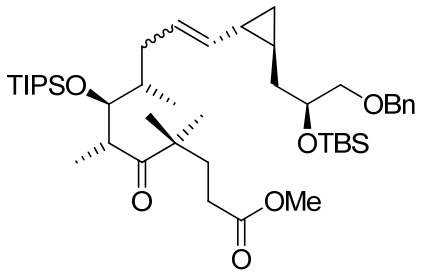
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 217.9, 173.8, 153.4, 133.1, 131.4, 129.7, 125.1, 77.7, 54.8, 51.7, 47.6, 44.3, 37.8, 34.1, 29.6, 24.6, 24.3, 23.6, 18.4, 18.4, 16.8, 16.5, 13.6.

IR (film): ν □2946, 2867, 1737, 1694, 1498, 1463, 1342, 1297, 1198, 1152, 1093, 1030, 1011, 990, 884, 849, 821, 763, 682, 627.

HRMS (ESI): calculated for $C_{31}H_{53}N_4O_6SSi$ $[M+H]^+$ 637.3450, found 637.3461.







253

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-3-(benzyloxy)-2-((*tert*-butyldimethylsilyl)oxy) propyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undec-10-enoate (253). To a solution of sulfon **227** (1.94 g, 3.04 mmol) and aldehyde **228** (1.28 g, 3.68 mmol) in THF (30 mL) was added LiHMDS (3.65 mL, 3.65 mmol) dropwise at -78 °C and the reaction mixture was stirred for 40 min at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (40 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 40 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 12:1) afforded 2.08 g (90%, *E*/*Z* 3:1) of olefine **253** as a colorless oil.

Note: The NMR data are given for the major (*E*) isomer only.

TLC: R_f 0.34 (hexane/EtOAc 9:1, UV (weak), CPS).

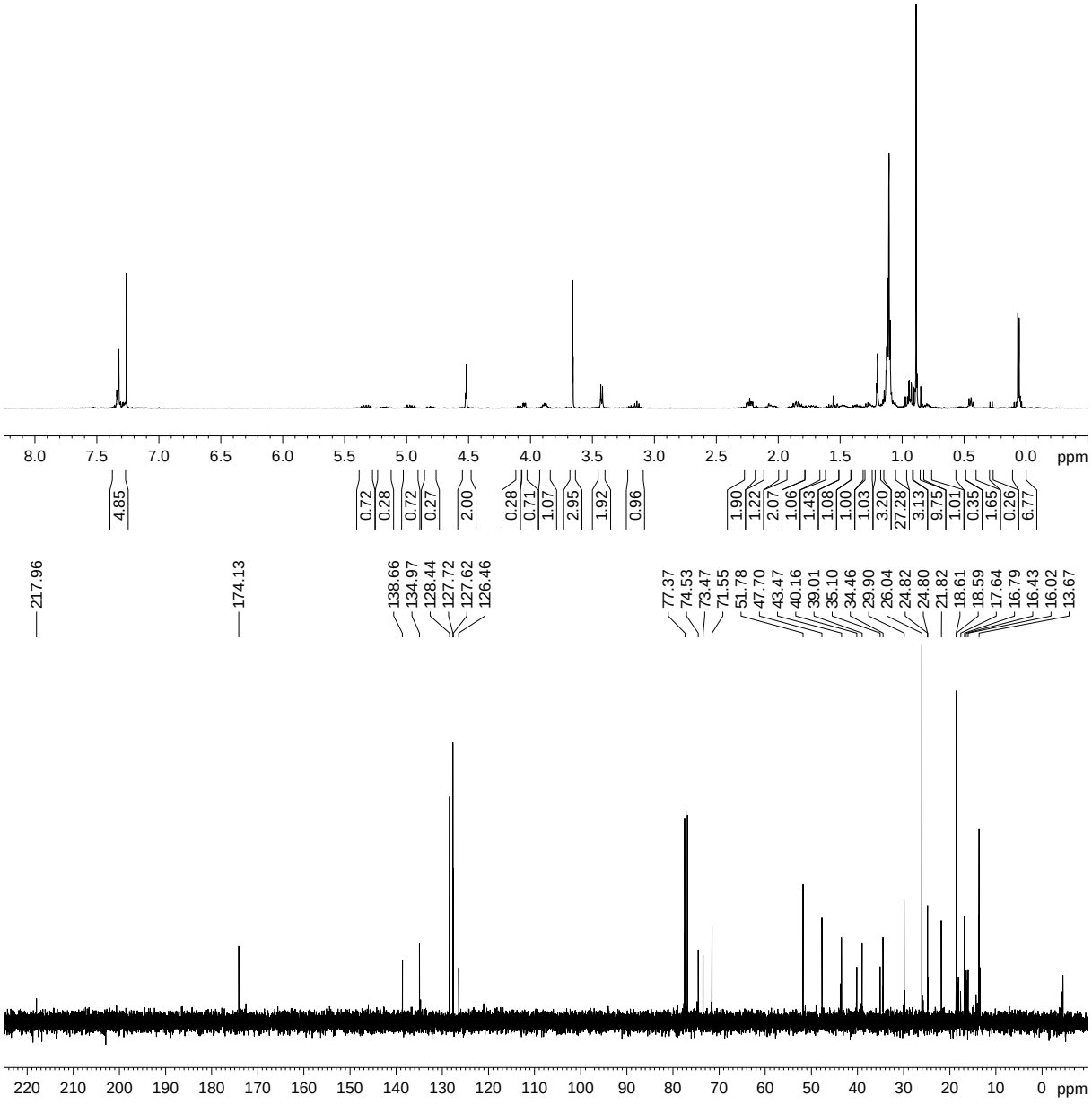
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.27 (m, 5H), 5.32 (ddd, *J* = 15.1, 7.8, 6.7 Hz, 1H), 4.96 (dd, *J* = 15.1, 8.5 Hz, 1H), 4.52 (s, 2H), 4.05 (dd, *J* = 6.7, 2.2 Hz, 1H), 3.92-3.86 (m,

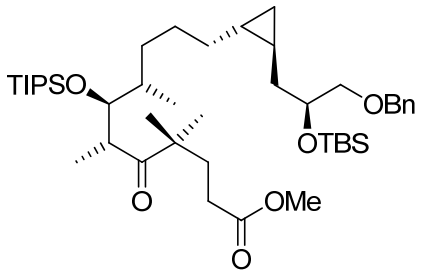
1H), 3.67 (s, 3H), 3.42 (d, *J* = 5.5 Hz, 2H), 3.14 (qd, *J* = 6.9, 6.7 Hz, 1H), 2.24 (dd, *J* = 6.4, 4.8 Hz, 1H), 2.22 (dd, *J* = 6.4, 3.8 Hz, 1H), 2.10-2.02 (m, 1H), 1.92-1.79 (m, 2H), 1.78-1.67 (m, 1H), 1.57 (dt, *J* = 13.7, 6.4 Hz, 1H), 1.51-1.41 (m, 1H), 1.36 (ddd, *J* = 13.7, 7.5, 5.0 Hz, 1H), 1.30-1.24 (m, 1H), 1.20 (s, 3H), 1.13-1.08 (m, 27H), 0.93 (d, *J* = 6.9 Hz, 3H), 0.89 (s, 9H), 0.83-0.76 (m, 1H), 0.45 (d, *J* = 7.9 Hz, 1H), 0.44 (d, *J* = 6.9 Hz, 1H), 0.06 (s, 3H), 0.05 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 138.7, 135.0, 128.4, 127.7, 127.6, 126.5, 77.4, 74.5, 73.5, 71.6, 51.8, 47.7, 43.5, 40.2, 39.0, 35.1, 34.5, 29.9, 26.0, 24.8, 24.8, 21.8, 18.6, 18.6, 17.6, 16.8, 16.4, 16.0, 13.7, 13.5, -4.3, -4.6.

IR (film): ν □2946, 2932, 2864, 1741, 1698, 1467, 1366, 1296, 1254, 1201, 1122, 1098, 1056, 1027, 991, 884, 835, 776, 738, 675.

HRMS (ESI): calculated for $C_{44}H_{82}NO_6Si_2$ $[M+NH_4]^+$ 776.5675, found 776.5674.





254

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-3-(benzyloxy)-2-((*tert*-butyldimethylsilyl)oxy)propyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoate (254). To a solution of olefine **253** (1.97 g, 2.59 mmol) and TPSH (11.62 g, 38.92 mmol) in 1,2-Dichloroethane (150 mL) was added Et_3N (5.41 mL, 38.92 mmol) over a period of 4 h at 50 °C and the reaction mixture was stirred overnight. The reaction mixture was diluted with EtOAc (50 mL) and filtered through a small plug of celite. The precipitate was rinsed extensively with EtOAc (200 mL) and the filtrate was concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 12:1) afforded 1.88 g (95%) of saturated **254** as a colorless oil.

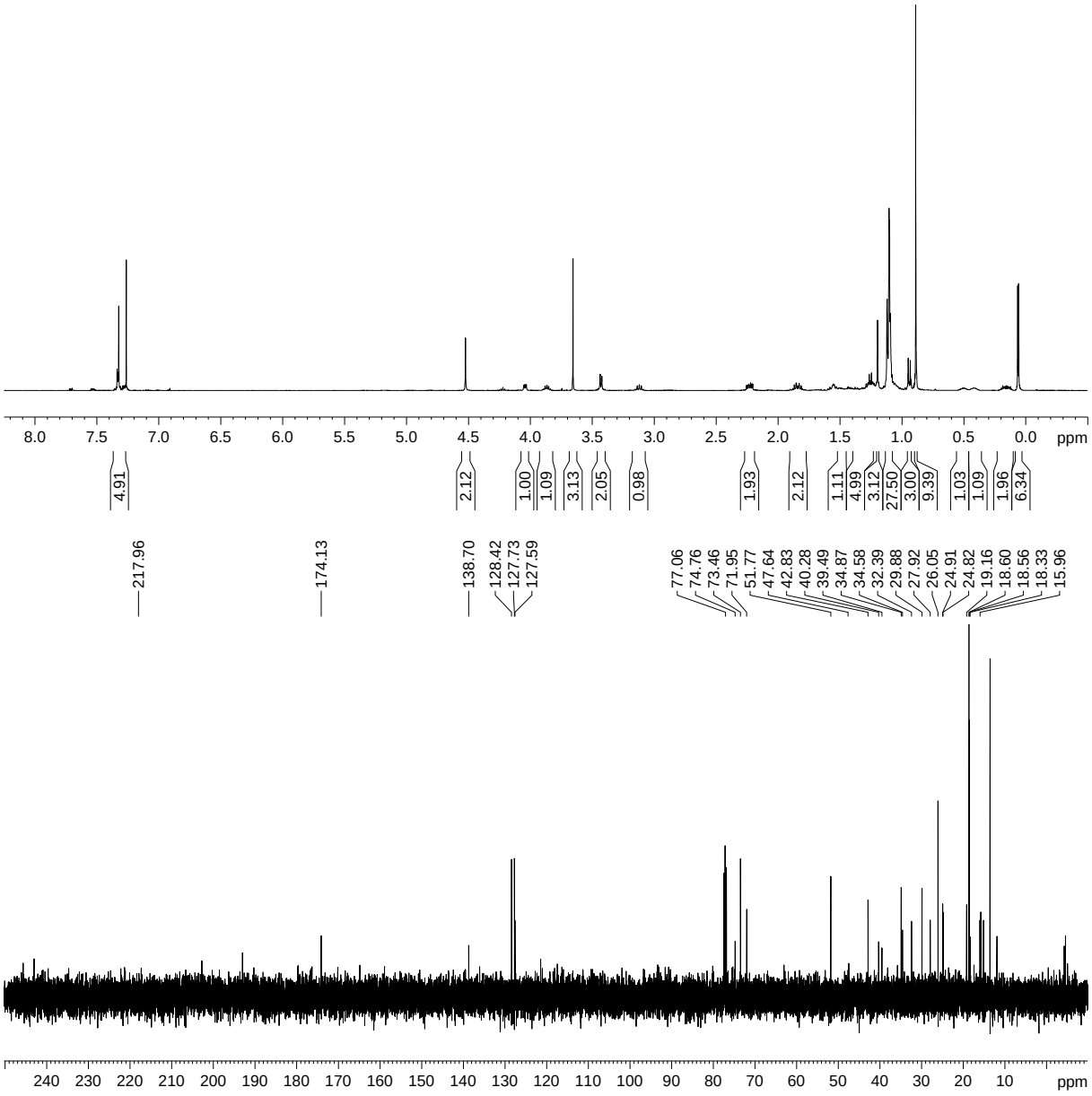
TLC: R_f 0.48 (hexane/EtOAc 9:1, UV (weak), CPS).

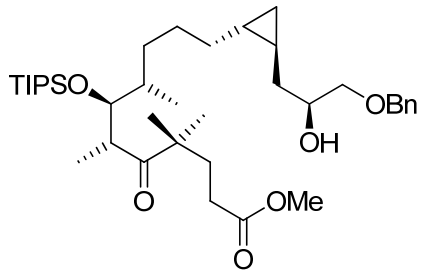
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.34-7.27 (m, 5H), 4.52 (s, 2H), 4.04 (dd, *J* = 6.2, 2.6 Hz, 1H), 3.87 (dq, *J* = 7.0, 5.6 Hz, 1H), 3.66 (s, 3H), 3.43 (d, *J* = 6.4 Hz, 1H), 3.43 (d, *J* = 6.2, 4.4 Hz, 1H), 3.12 (qd, *J* = 7.0, 6.0 Hz, 1H), 2.24 (dd, *J* = 9.0, 3.7 Hz, 1H), 2.22 (dd, *J* = 9.0, 3.1 Hz, 1H), 1.86 (dd, *J* = 10.5, 4.8 Hz, 1H), 1.82 (dd, *J* = 10.5, 4.5 Hz, 1H), 1.54-1.45 (m, 1H), 1.44-1.22 (m, 5H), 1.20 (s, 3H), 1.14-1.07 (m, 27H), 0.94 (d, *J* = 6.9 Hz, 3H), 0.89 (s, 9H), 0.50 (dddd, *J* = 10.7, 8.1, 7.3, 4.9 Hz, 1H), 0.42 (dddd, *J* = 11.0, 8.1, 5.9, 4.9 Hz, 1H), 0.17 (ddd, *J* = 12.2, 4.6, 4.6 Hz, 1H), 0.13 (ddd, *J* = 12.2, 4.5, 4.5 Hz, 1H), 0.07 (s, 3H), 0.06 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 138.7, 128.4, 127.7, 127.6, 77.1, 74.8, 73.5, 72.0, 51.8, 47.6, 42.8, 40.3, 39.5, 34.9, 34.6, 32.4, 29.9, 27.9, 26.1, 24.9, 24.8, 19.2, 18.6, 18.6, 18.3, 16.0, 15.8, 15.1, 13.6, 11.9, -4.3, -4.6.

IR (film): ν □2947, 2928, 2863, 1742, 1697, 1463, 1365, 1254, 1120, 1093, 1063, 990, 978, 883, 835, 809, 776, 734, 698, 677.

HRMS (ESI): calculated for $C_{44}H_{84}NO_6Si_2$ $[M+NH_4]^+$ 778.5832, found 778.5821.





255

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-3-(benzyloxy)-2-hydroxypropyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoate (255). To a solution of silyl ether **254** (2.00 g, 2.62 mmol) in CH_2Cl_2 (80 mL) and MeOH (40 mL) was added CSA

(0.92 g, 3.94 mmol) in one portion at room temperature. The reaction mixture was stirred at for 3 h and the reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (50 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 4:1) afforded 1.62 g (95%) of alcohol **255** as a colorless oil.

TLC: R_f 0.24 (hexane/EtOAc 4:1, UV (weak), CPS).

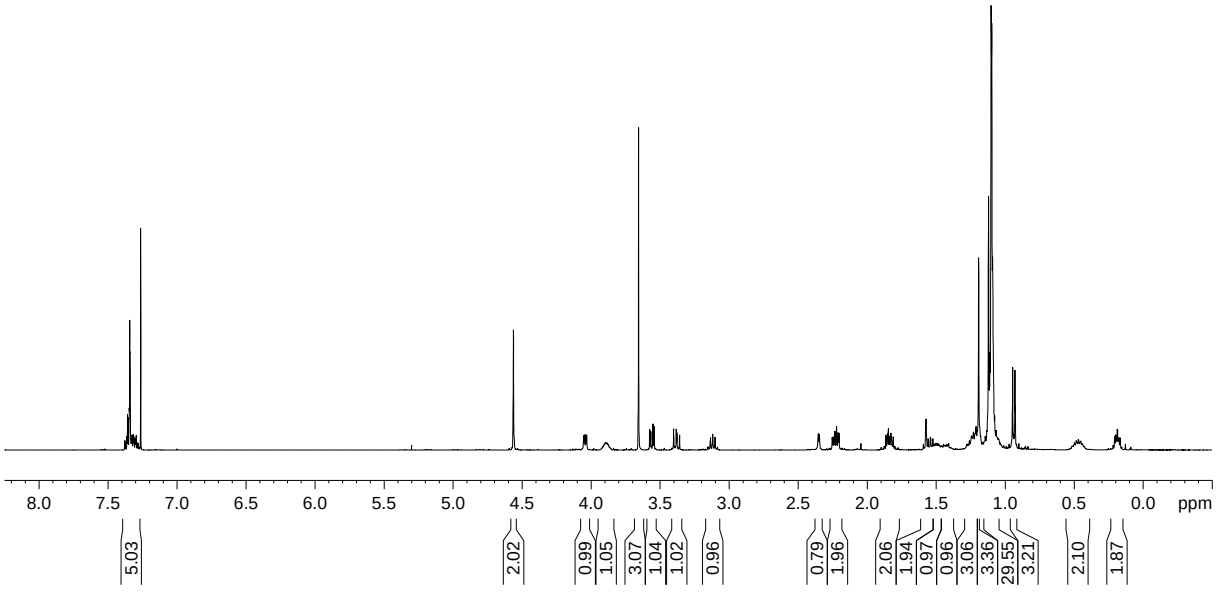
$[\alpha]^{20}_D$: = (*c* = 0.60, $CHCl_3$).

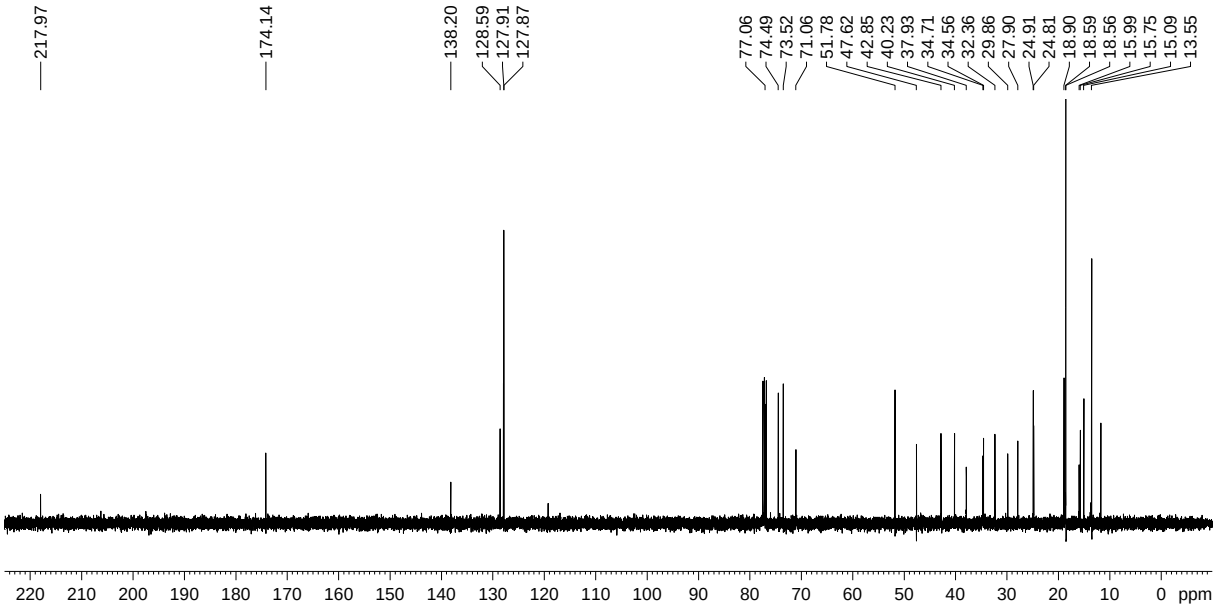
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.38-7.27 (m, 5H), 4.56 (s, 2H), 4.04 (dd, *J* = 6.2, 2.6 Hz, 1H), 3.89 (qdd, *J* = 7.0, 3.2, 3.2 Hz, 1H), 3.65 (s, 3H), 3.56 (d, *J* = 9.4, 3.2 Hz, 1H), 3.43 (d, *J* = 9.4, 7.7 Hz, 1H), 3.12 (qd, *J* = 7.0, 6.1 Hz, 1H), 2.35 (d, *J* = 3.4 Hz, 1H), 2.24 (dd, *J* = 8.4, 3.5 Hz, 1H), 2.22 (dd, *J* = 8.4, 2.5 Hz, 1H), 1.85 (dd, *J* = 6.9, 4.6 Hz, 1H), 1.83 (dd, *J* = 6.9, 5.0 Hz, 1H), 1.59-1.52 (m, 1H), 1.52-1.47 (m, 1H), 1.46-1.38 (m, 1H), 1.28-1.20 (m, 3H), 1.19 (s, 3H), 1.13-1.07 (m, 30H), 0.94 (d, *J* = 6.9 Hz, 3H), 0.55-0.40 (m, 2H), 0.20 (dd, *J* = 7.3, 4.5 Hz, 1H), 0.17 (ddd, *J* = 7.9, 4.5 Hz, 1H).

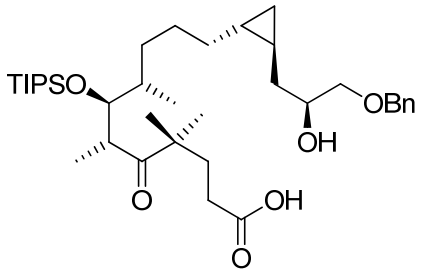
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 138.2, 128.6, 127.9, 127.9, 77.1, 74.5, 73.5, 71.1, 51.8, 47.6, 42.8, 40.2, 37.9, 34.7, 34.6, 32.4, 29.9, 27.9, 24.9, 24.8, 18.9, 18.6, 18.6, 16.0, 15.8, 15.1, 13.5, 11.8.

IR (film): ν □3511, 2963, 2943, 2865, 1740, 1696, 1456, 1438, 1368, 1298, 1258, 1201, 1118, 1092, 1065, 990, 883, 847, 814, 735, 698, 678.

HRMS (ESI): calculated for $C_{38}H_{70}NO_6Si$ $[M+NH_4]^+$ 664.4967, found 664.4981.







256

(6*R*,7*S*,8*S*)-11-((1*R*,2*S*)-2-((*S*)-3-(benzyloxy)-2-hydroxypropyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoic acid (256). To a solution of ester **255** (1.55 g, 2.40 mmol) in *t*-BuOH (48 mL) and H_2O (12 mL) was added LiOH monohydrate (0.60 g, 14.40 mmol) in one portion at room temperature. The reaction mixture was stirred for 2 h at this temperature. The reaction mixture diluted with water (50 mL) and CH_2Cl_2 (50 mL) whereupon the solution turned milky. The solution was then acidified by addition of 1 M aqueous HCl solution until pH 5 was reached. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced to yield 1.55 g (100%) of acid **256** as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -11.2 (*c* = 0.60, $CHCl_3$).

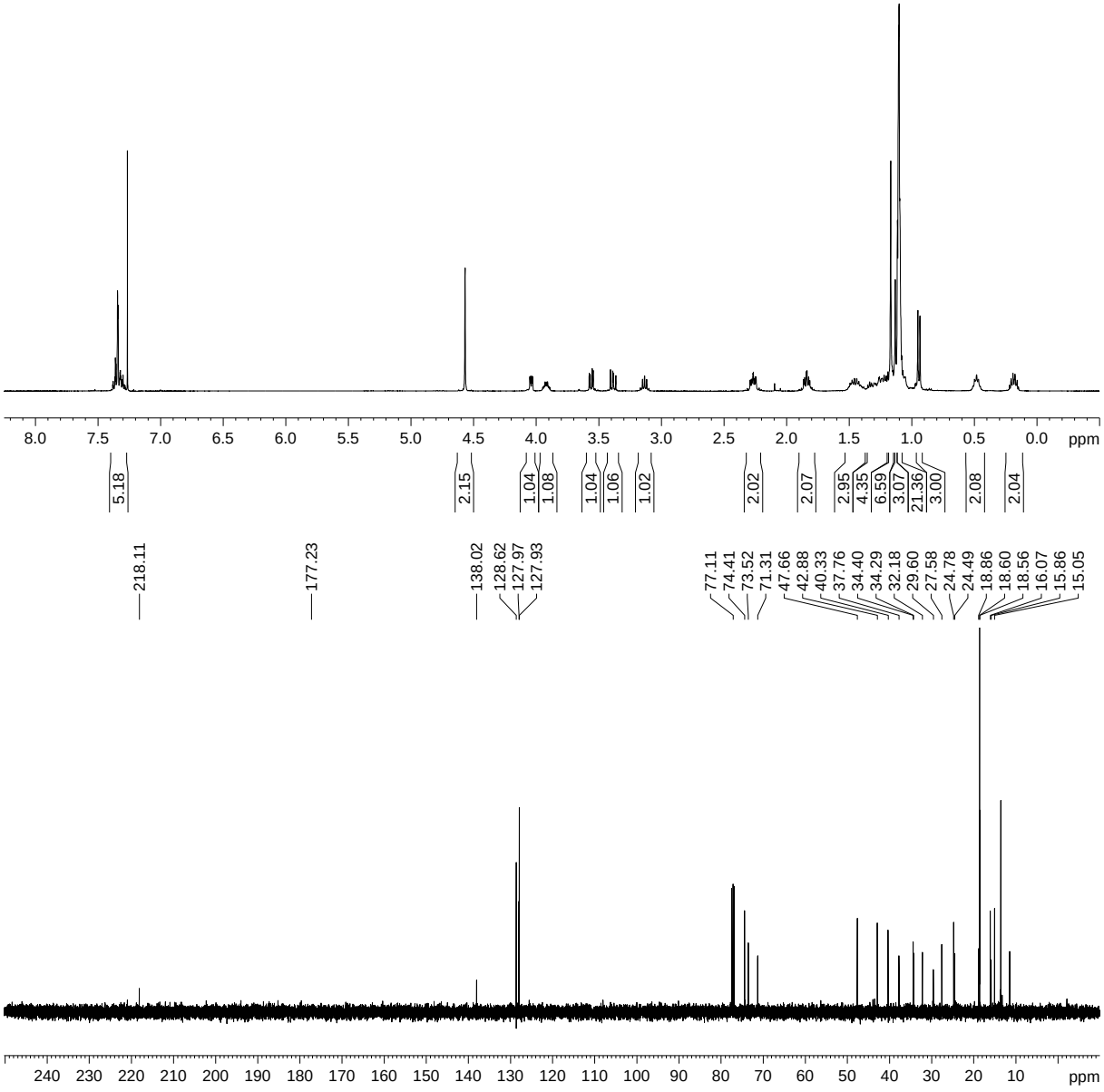
[^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.40-7.27 (m, 5H), 4.52 (s, 2H), 4.04 (dd, *J* = 6.1, 2.5 Hz, 1H), 3.92 (dddd, *J* = 7.8, 7.2, 5.3, 3.1 Hz, 1H), 3.56 (dd, *J* = 9.5, 3.1 Hz, 1H), 3.38 (dd, *J* = 9.5, 7.6 Hz, 1H), 3.13 (qd, *J* = 6.9, 6.3 Hz, 1H), 2.28 (dd, *J* = 5.9, 3.1 Hz, 1H), 2.25 (dd,

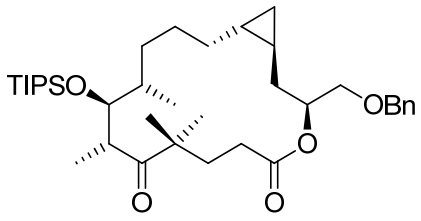
J = 5.9, 2.4 Hz, 1H), 1.85 (dd, *J* = 8.7, 3.1 Hz, 1H), 1.83 (dd, *J* = 8.7, 3.8 Hz, 1H), 1.51-1.37 (m, 3H), 1.35-1.19 (m, 5H), 1.17 (s, 6H), 1.12 (d, *J* = 6.9 Hz, 3H), 1.11-1.08 (m, 21H), 0.94 (d, *J* = 6.9 Hz, 3H), 0.94-0.89 (m, 1H), 0.88-0.86 (m, 1H), 0.48 (dddd, *J* = 12.7, 9.7, 5.1, 4.8 Hz, 2H), 0.18 (ddd, *J* = 12.4, 10.7, 4.4 Hz, 2H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 177.2, 138.0, 128.6, 128.0, 127.9, 77.1, 74.4, 73.5, 71.3, 47.7, 42.9, 40.3, 37.8, 34.4, 34.3, 32.2, 29.6, 27.6, 24.8, 24.5, 18.9, 18.6, 18.6, 16.1, 15.9, 15.1, 13.6, 11.5.

IR (film): ν □3470, 2929, 2865, 1704, 1459, 1386, 1367, 1292, 1240, 1207, 1117, 1093, 998, 982, 883, 816, 738, 699, 676.

HRMS (ESI): calculated for $C_{37}H_{64}NaO_6Si$ $[M+Na]^+$ 655.4364, found 655.4359.





218

(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-3-((benzyloxy)methyl)-8,8,10,12-tetramethyl-11-((triisopropylsilyl)oxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (218). To a solution of NEt_3 (0.68 mL, 4.90 mmol), 2,4,6-trichlorobenzoyl chloride (0.42 mL, 2.69 mmol) and DMAP (0.39 g, 3.18 mmol) in THF (900 mL) was added acid **256** (1.55 g, 2.45 mmol) in THF (50 mL) over a period of 45 min at room temperature and the reaction mixture was stirred for additional 45 min. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (150 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 12:1) to yield 1.13 g (75%) of macrolactone **218** as a colorless oil.

TLC: R_f 0.21 (hexane/EtOAc 12:1, CPS).

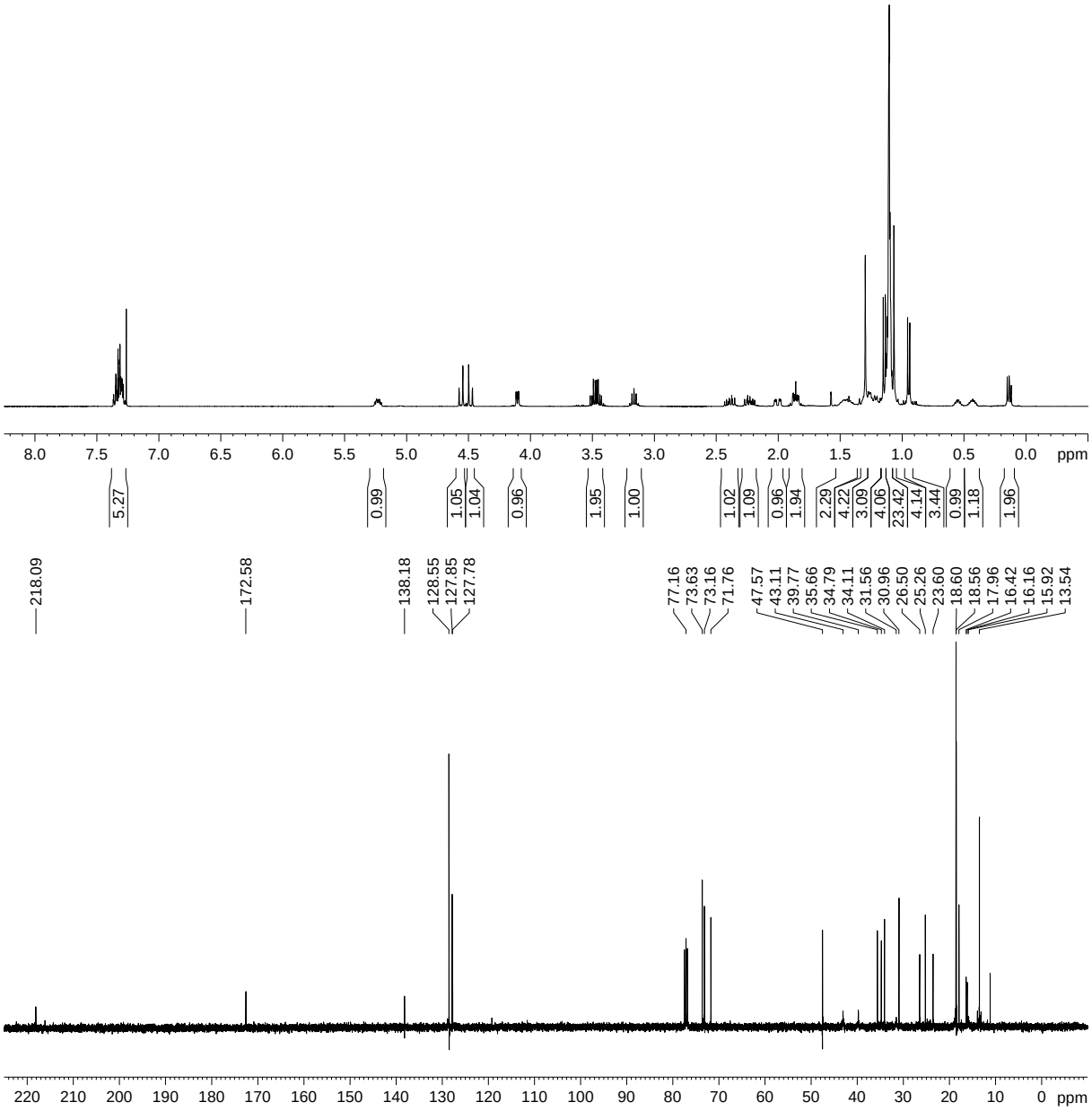
$[\alpha]^{20}_D$: = +11.8 (*c* = 1.64, $CHCl_3$).

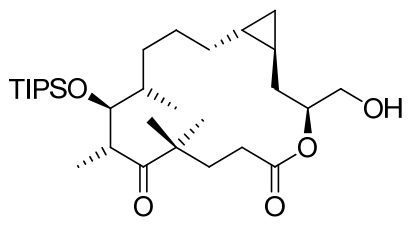
[^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.27 (m, 5H), 5.23 (dddd, *J* = 10.0, 5.2, 5.2, 1.8 Hz, 1H), 4.56 (d, *J* = 12.1 Hz, 1H), 4.48 (d, *J* = 12.1 Hz, 1H), 4.11 (dd, *J* = 6.4, 2.4 Hz, 1H), 3.50 (dd, *J* = 10.3, 5.7 Hz, 1H), 3.44 (dd, *J* = 10.7, 4.8 Hz, 1H), 3.17 (qd, *J* = 6.9, 6.8 Hz, 1H), 2.39 (ddd, *J* = 15.2, 11.1, 6.2 Hz, 1H), 2.23 (ddd, *J* = 15.2, 11.4, 6.0 Hz, 1H), 2.01 (dd, *J* = 14.9, 4.1, 1.8 Hz, 1H), 1.87 (dd, *J* = 6.0, 1.9 Hz, 1H), 1.85 (dd, *J* = 6.2, 2.4 Hz, 1H), 1.53-1.36 (m, 2H), 0.55 (dddd, *J* = 12.4, 7.0, 6.3, 4.3 Hz, 1H), 0.43 (dddd, *J* = 10.5, 9.5, 6.3, 4.1 Hz, 1H), 0.15 (d, *J* = 6.3 Hz, 1H), 0.13 (d, *J* = 6.3 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 172.6, 138.2, 128.5, 127.9, 127.8, 77.2, 73.6, 73.2, 71.8, 47.6, 43.1, 39.8, 35.7, 34.8, 34.1, 31.6, 31.0, 26.5, 25.3, 23.6, 18.6,18.6, 18.0, 16.4, 16.2, 15.9, 13.5, 11.2.

IR (film): ν □2939, 2865, 1734, 1697, 1459, 1367, 1286, 1254, 1204, 1121, 998, 980, 883, 819, 737, 698, 676.

HRMS (ESI): calculated for $C_{37}H_{63}O_5Si$ $[M+H]^+$ 615.4439, found 615.4456.





257

(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-3-(hydroxymethyl)-8,8,10,12-tetramethyl-11-((triisopropylsilyl)oxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (257). To a solution of protected alcohol **218** (45 mg, 0.073 mmol) in CH_2Cl_2 (1.3 mL) was added $BCl_3{\cdot}SMe_2$ (55 µL, 0.336 mmol) dropwise at room temperature and the reaction mixture was stirred for 5.5

h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 21.3 mg (48%) of alcohol **257** as a colorless oil.

TLC: R_f 0.21 (hexane/EtOAc 3:1, CPS).

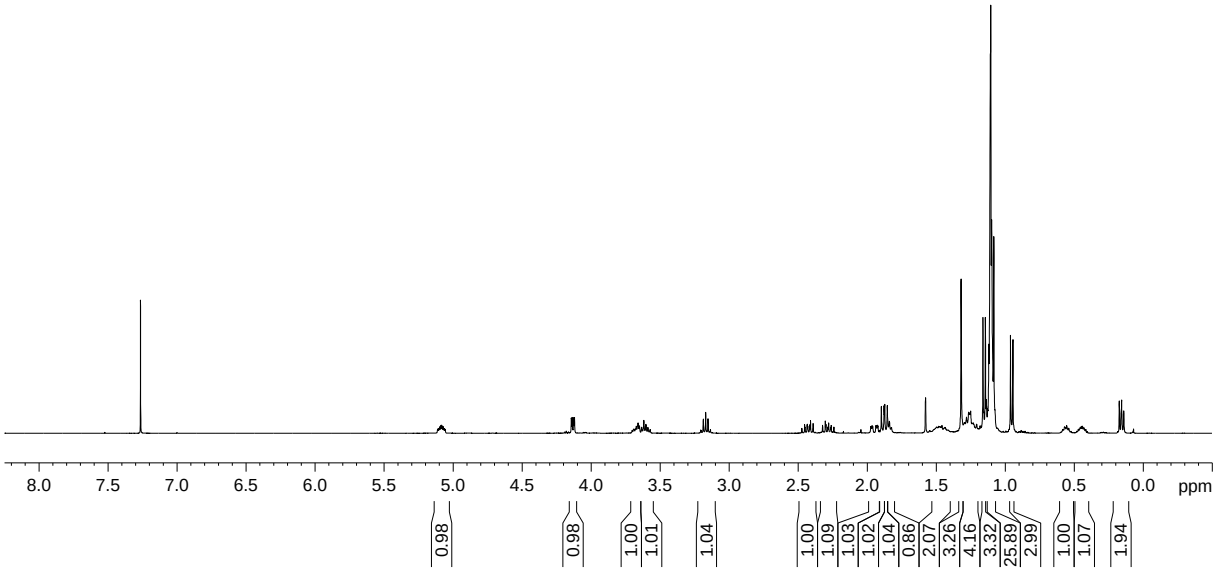
$[\alpha]^{20}_D$: = +15.2 (*c* = 0.69, $CHCl_3$).

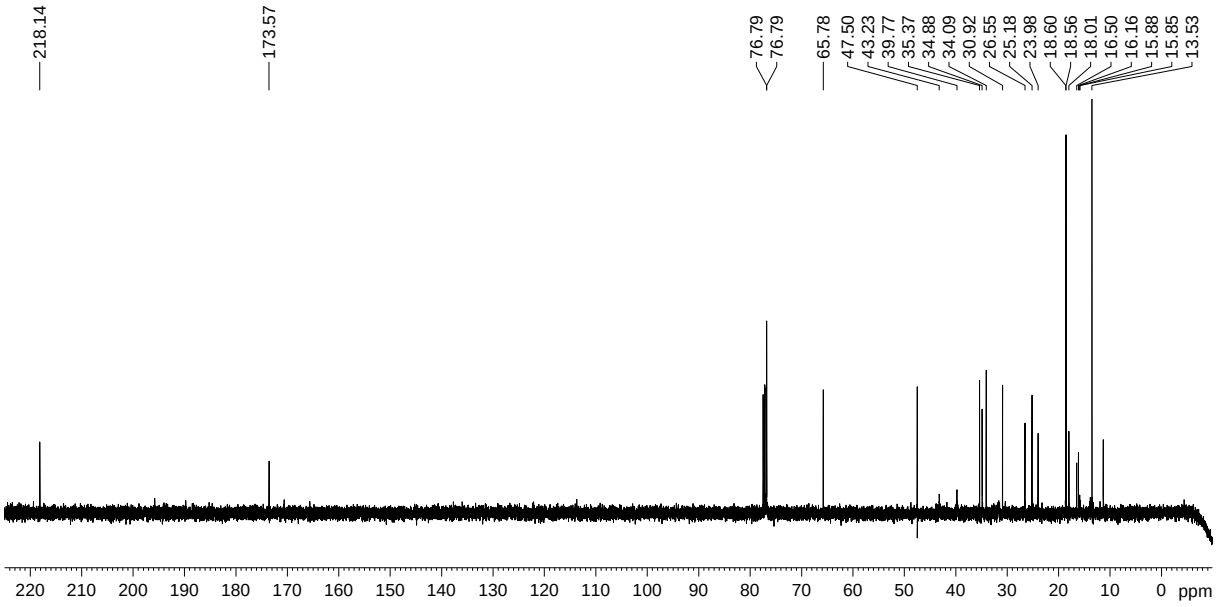
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.08 (dddd, *J* = 10.0, 5.9, 3.8, 1.7 Hz, 1H), 4.13 (dd, *J* = 6.6, 2.3 Hz, 1H), 3.67 (ddd, *J* = 11.8, 5.2, 3.9 Hz, 1H), 3.60 (ddd, *J* = 11.8, 6.4, 6.3 Hz, 1H), 3.17 (dq, *J* = 7.0, 6.9 Hz, 1H), 2.43 (dddd, *J* = 9.3, 7.4, 7.3, 7.3 Hz, 1H), 2.28 (dddd, *J* = 9.7, 7.8, 7.5, 7.4 Hz, 1H), 1.95 (ddd, *J* = 14.9, 4.1, 1.7 Hz, 1H), 1.89 (d, *J* = 7.4 Hz, 1H), 1.86 (d, *J* = 7.8 Hz, 1H), 1.53-1.39 (m, 2H), 1.32 (s, 3H), 1.30-1.20 (m, 4H), 1.15 (d, *J* = 6.9 Hz, 3H), 1.13-1.07 (m, 26H), 0.95 (d, *J* = 6.8 Hz, 3H), 0.56 (ddd, *J* = 13.6, 11.0, 6.1 Hz, 1H), 0.44 (dddd, *J* = 10.6, 9.6, 6.5, 4.3 Hz, 1H), 0.16 (d, *J* = 7.5 Hz, 1H), 0.15 (d, *J* = 7.4 Hz, 1H).

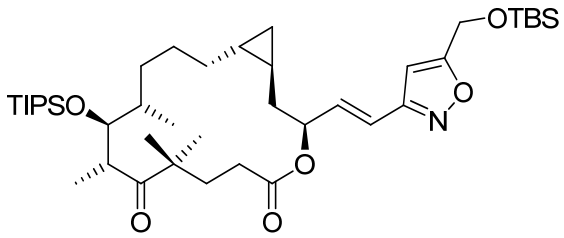
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 173.6, 76.8, 76.8, 65.8, 47.5, 43.2, 39.8, 35.4, 34.9, 34.1, 30.9, 26.6, 25.2, 24.0, 18.6, 18.6, 18.0, 16.5, 16.2, 15.9, 15.9, 13.5, 11.3.

IR (film): ν □3464, 2940, 2866, 1732, 1699, 1462, 1384, 1368, 1285, 1258, 1123, 1087, 1043, 999, 982, 883, 820, 674.

HRMS (ESI): calculated for $C_{30}H_{57}O_5Si$ $[M+H]^+$ 525.3970, found 525.3952.







258

(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-3-((*E*)-2-(5-(((*tert*-butyldimethylsilyl)oxy)methyl)isoxazol-3-yl)vinyl)-8,8,10,12-tetramethyl-11-((triisopropylsilyl)oxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (258). To a solution of alcohol **257** (21.3 mg, 0.041 mmol) in CH_2Cl_2 (2 mL) was added DMP (0.12 mL, 0.060 mmol) dropwise at 0 °C whereupon the reaction mixture turned milky. The reaction mixture was allowed to reach room temperature and stirred for 0.5 h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (2 mL) and saturated aqueous $Na_2S_2O_3$ (2 mL). Stirring was continued for 0.5 h, when two almost clear phases had formed. The phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the obtained crude aldehyde was directly used in the subsequent Wittig reaction.

To a solution of phosphonate **140b** (62 mg, 0.170 mmol) in THF (2 mL) was added *t*-BuOK (18.2 mg mL, 0.162 mmol) in one portion at 0 °C and the reaction mixture was stirred for 0.5 h. The reaction mixture was cooled to -78 °C and crude aldehyde (ca. 21 mg, 0.041 mmol) in THF (0.5 mL) was added dropwise. The reaction mixture was allowed to

warm to -20 °C over a period of 2 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (2 mL), the phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 12:1) to afford 12.0 mg (40%) of an inseparable 1:1 mixture of **258** and its C16-C17 *Z* isomer as a colorless oil.

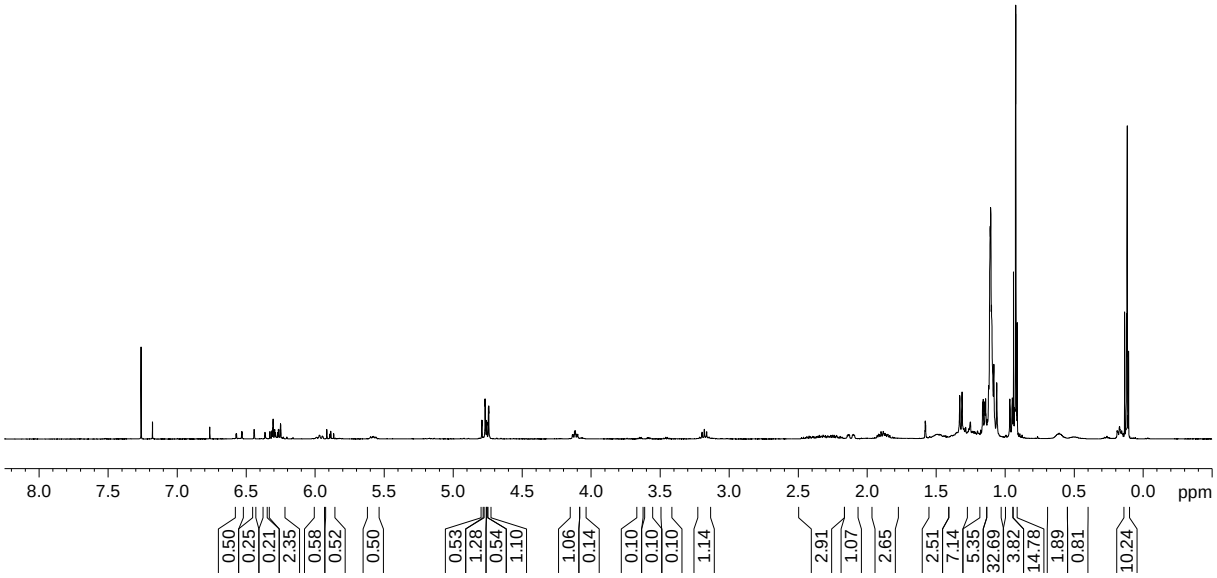
TLC: R_f 0.32 (hexane/EtOAc 9:1, UV, CPS).

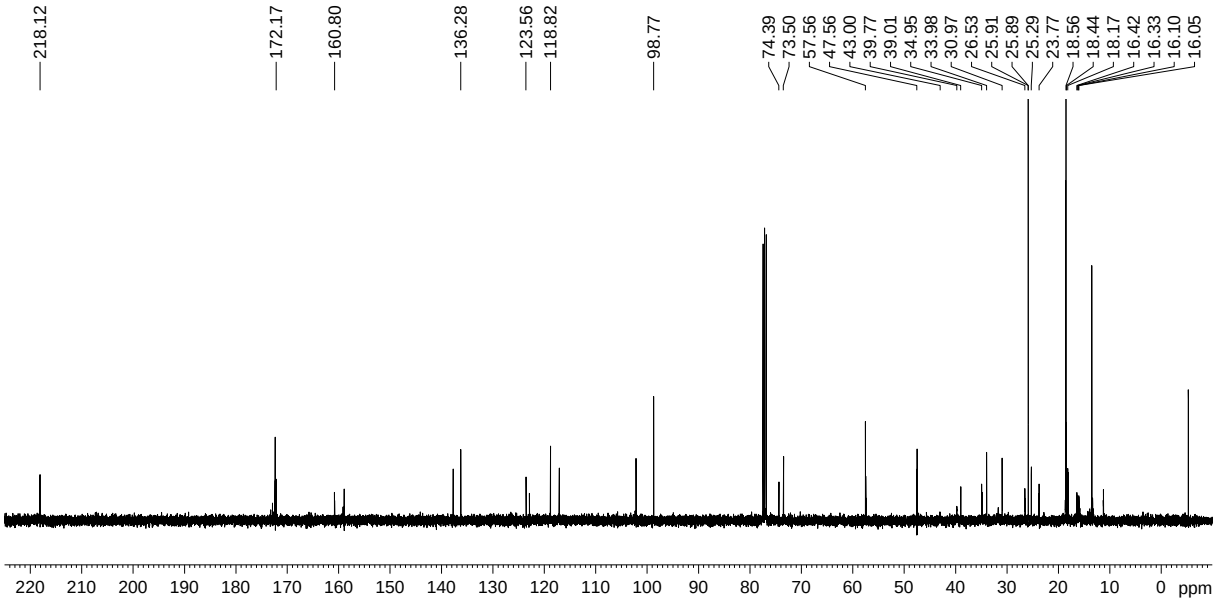
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.30 (dd, *J* = 16.2, 5.8 Hz, 1H), 6.28 (d, *J* = 16.2 Hz, 1H), 6.25 (s, 1H), 5.58 (dddd, *J* = 7.7, 7.6, 1.7, 1.7 Hz, 1H), 4.77 (d, *J* = 0.7 Hz, 2H), 4.12 (dd, *J* = 6.6, 2.3 Hz, 1H), 3.18 (qd, *J* = 6.9, 6.6 Hz, 1H), 2.48-2.17 (m, 3H), 2.12 (ddd, *J* = 14.8, 4.0, 2.1 Hz, 1H), 1.95-1.85 (m, 2H), 1.54-1.40 (m, 2H), 1.31 (s, 3H), 1.30-1.20 (m, 2H), 1.15 (d, *J* = 6.9 Hz, 3H), 1.13-1.04 (m, 27H), 0.96 (d, *J* = 6.9 Hz, 3H), 0.92 (s, 9H), 0.65-0.55 (m, 2H), 0.19-0.13 (m, 2H), 0.13 (s, 3H), 0.11 (s, 3H).

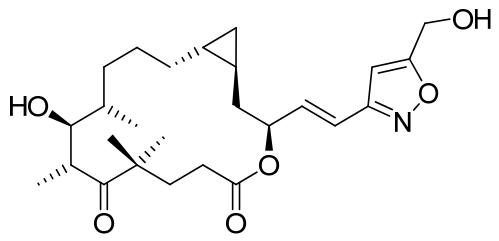
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 172.2, 160.8, 136.3, 123.6, 118.8, 98.8, 74.4, 73.5, 57.6, 47.6, 43.0, 39.8, 39.0, 34.9, 34.0, 31.0, 26.5, 25.9, 25.9, 25.3, 23.8, 18.6, 18.4, 18.2, 16.4, 16.3, 16.1, 16.0, 13.5, 11.2, -5.2, -5.3.

IR (film): ν □2929, 2862, 1736, 1696, 1604, 1463, 1386, 1364, 1287, 1255, 1136, 1092, 1002, 980, 917, 883, 837, 815, 779, 677.

HRMS (ESI): calculated for $C41H_{74}NO_6Si2$ $[M+H]^+$ 732.5049, found 732.5036.







3b

(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-11-hydroxy-3-((*E*)-2-(5-(hydroxymethyl)isoxazol-3-yl)vinyl)-8,8,10,12-tetramethyl-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (3b). To a 1:1 mixture of protected cyclopropyl-Epo B **258** and its C16-C17 *Z* isomer (38 mg, 0.052 mmol) in THF (2 mL) was added HF/pyridine (2 mL, ca. 70% HF) dropwise at room temperature and the reaction mixture was stirred for 2 h. The reaction mixture was then carefully poured into a cold saturated aqueous solution of $NaHCO_3$ (150 mL). The aqueous phase was extracted with EtOAc (3 x 40 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:1) to afford 18.4 mg (77%) of hypermodified Epo A analog **3b** as a colorless oil.

TLC: R_f 0.41 (hexane/EtOAc 1:2, UV, CPS).

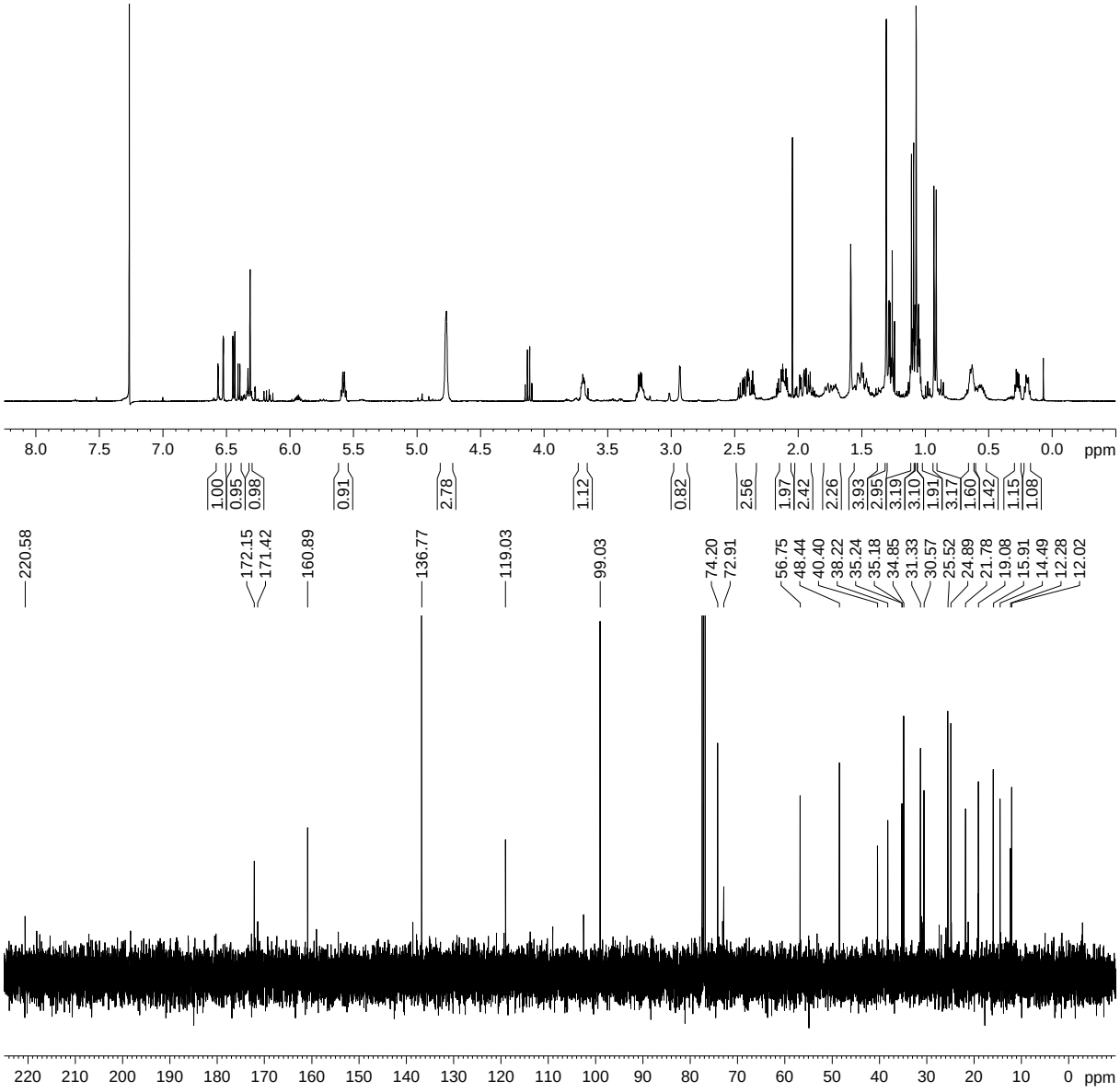
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.54 (dd, *J* = 16.3, 1.2 Hz, 1H), 6.42 (dd, *J* = 16.3, 5.9 Hz, 1H), 6.31 (s, 1H), 5.58 (ddd, *J* = 10.2, 4.9, 1.1 Hz, 1H), 4.77 (s, 1H), 4.77 (s, 2H), 3.71-3.66 (m, 1H), 3.24 (dd, *J* = 6.8. 3.5 Hz, 1H), 2.93 (d, *J* = 1.5 Hz, 1H), 2.43 (ddd, *J* = 14.3, 12.7, 5.5 Hz, 1H), 2.38 (ddd, *J* = 14.3, 4.2, 4.2 Hz, 1H), 2.14-2.07 (m, 2H), 1.98 (ddd, *J* = 14.0, 13.7, 4.1 Hz, 1H), 1.93 (dd, *J* = 12.5, 5.3 Hz, 1H), 1.80-1.67 (m, 2H), 1.55-

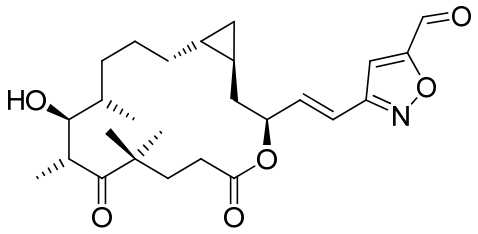
1.37 (m, 4H), 1.31 (s, 3H), 1.10 (d, *J* = 6.8 Hz, 3H), 1.07 (s, 3H), 0.92 (d, *J* = 7.0 Hz, 3H), 0.65-0.61 (m, 1H), 0.60-0.52 (m, 1H), 0.28 (dd, *J* = 8.5, 4.7 Hz, 1H), 0.22-0.17 (m, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 220.6, 172.1, 171.4, 160.9, 136.8, 119.0, 99.0, 74.2, 72.9, 56.7, 48.4, 40.4, 38.2, 35.2, 35.2, 34.8, 31.3, 30.6, 25.5, 24.9, 21.8, 19.1, 15.9, 14.5, 12.3, 12.0.

IR (film): ν □3432, 2969, 2932, 2876, 1731, 1691, 1603, 1458, 1438, 1371, 1288, 1259, 1200, 1132, 1075, 1040, 974, 913, 803, 733.

HRMS (ESI): calculated for $C_{26}H_{39}NNaO_6$ $[M+Na]^+$ 484.2670, found 484.2665.

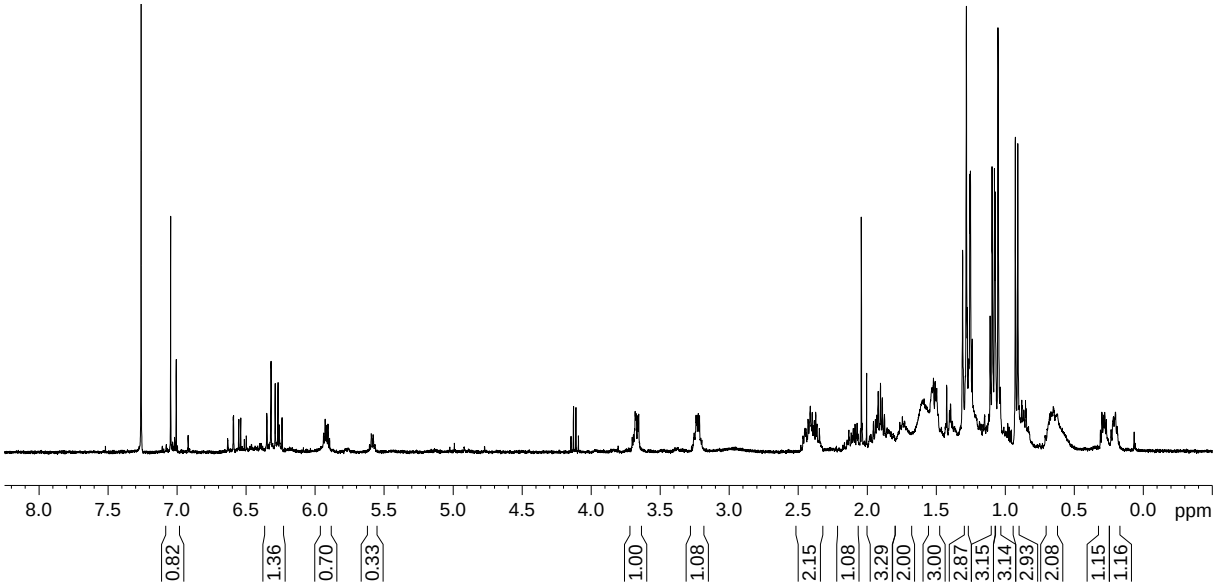


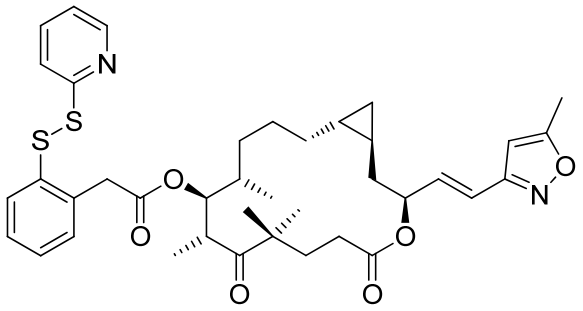


277

3-((*E*)-2-((1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-11-hydroxy-8,8,10,12-tetramethyl-5,9-dioxo-4-oxabicyclo[14.1.0]heptadecan-3-yl)vinyl)isoxazole-5-carbaldehyde (277). To a solution of alcohol **3b** (1.9 mg, 0.004 mmol) in CH_2Cl_2 (0.5 ml) was added BAIB (1.5 mg, 0.005 mmol) and TEMPO (0.14 mg, 0.001 mmol) at 0 °C. The reaction mixture was allowed to reach room temperature and was stirred for 2 h. The reaction mixture was then concentrated under reduced pressure the residue was purified by silica gel column chromatography using hexane/EtOAc (1: 1) as eluent to give 1.2 mg (63%) of aldehyde **277** as a colorless oil.

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.98 (d, *J* = 1.4 Hz, 1H), 7.04 (s, 1H), 6.33 (d, 11.8 Hz, 1H), 6.26 (dd, *J* = 11.8, 8.2 Hz, 1H), 5.92 (ddd, *J* = 8.2, 4.8, 4.2 Hz, 1H), 3.67 (dd, *J* = 6.8, 3.2 Hz, 1H), 3.23 (dd, *J* = 6.9, 3.2 Hz, 1H), 2.48-2.33 (m, 2H), 2.16-2.07 (m, 1H), 1.99-1.80 (m, 3H), 1.79-1.67 (m, 2H), 1.55-1.47 (m, 2H), 1.44-1.33 (m, 2H), 1.28 (s, 3H), 1.09 (d, *J* = 6.9 Hz, 3H), 1.05 (s, 3H), 0.91 (d, *J* = 7.0, 3H), 0.70-0.63 (m, 2H), 0.29 (ddd, *J* = 8.0, 4.8, 4.6 Hz, 1H), 0.23-0.18 (m, 1H).





276

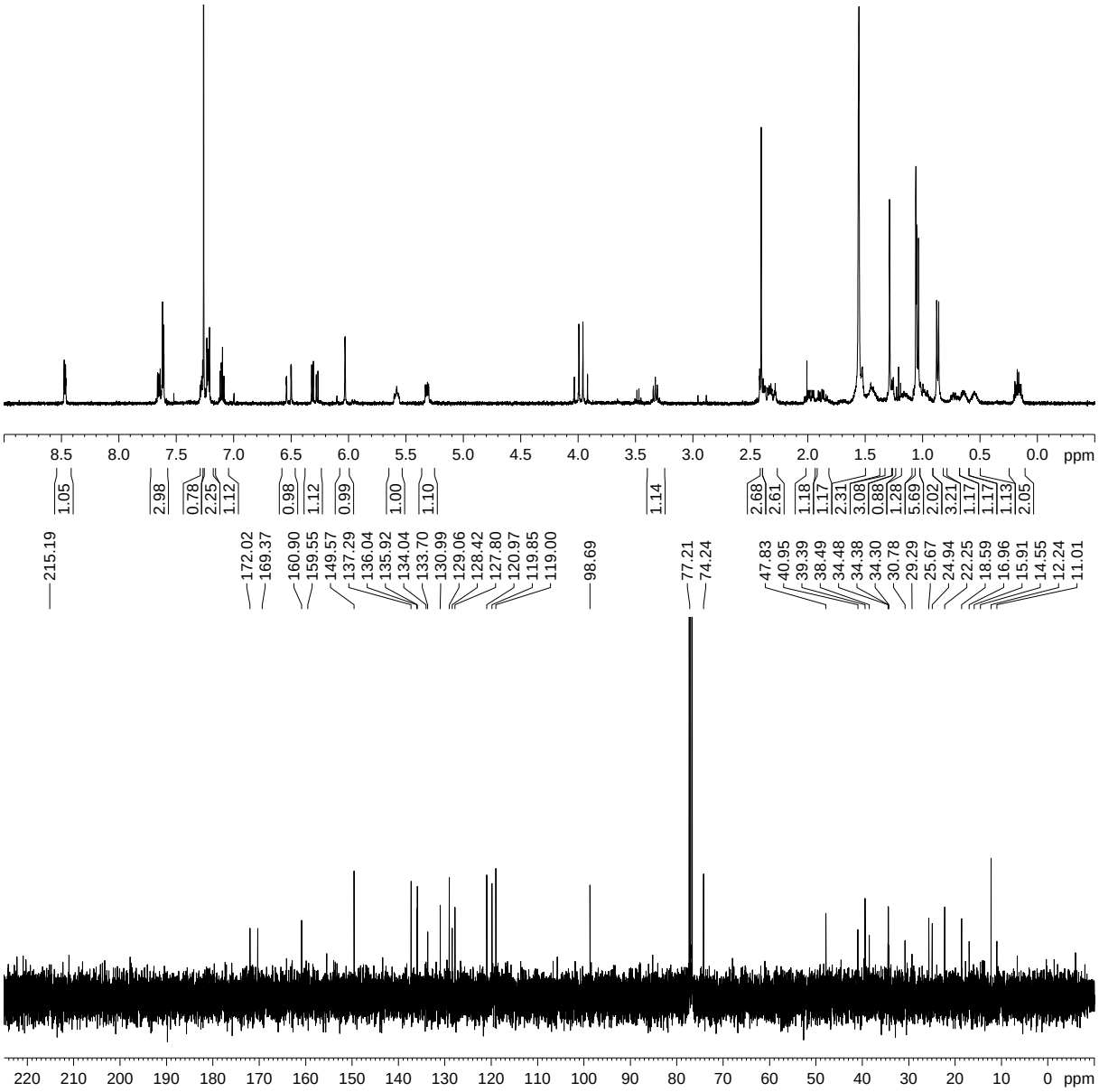
(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-8,8,10,12-tetramethyl-3-((*E*)-2-(5-methylisoxazol-3-yl)vinyl)-5,9-dioxo-4-oxabicyclo[14.1.0]heptadecan-11-yl 2-(2-(pyridin-2-yldisulfanyl)phenyl)acetate (276). To a solution of acid (10.1 mg, 0.036 mmol) and alcohol **3a** (8.1 mg, 0.018 mmol) in CH_2Cl_2 (1 mL) was added DCC (11.3 mg, 0.055 mmol) and DMAP (2.2 mg, 0.018 mmol) at room temperature and the mixture was stirred for 2 h. The reaction mixture was then filtered through a small plug of silica. The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 2:1) to afford 2.0 mg (16%) of ester **276** as a colorless oil.

TLC: R_f 0.32 (hexane/EtOAc 3:2, UV, CPS).

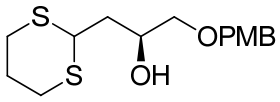
$[\alpha]^{20}_D$: = +3.9 (*c* = 0.31, $CHCl_3$).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.45 (dt, *J* = 4.6, 1.3 Hz, 1H), 7.66-7.64 (m, 1H), 7.61 (dd, *J* = 4.6, 1.3 Hz, 2H), 7.29-7.20 (m, 3H), 7.10 (dd, *J* = 8.6, 4.6 Hz, 1H), 6.52 (dd, *J* = 16.0, 1.2 Hz, 1H), 6.29 (dd, *J* = 16.0, 6.0 Hz, 1H), 6.03 (s, 3H), 5.60-5.56 (m, 1H), 5.32 (dd, *J* = 7.4, 3.8 Hz, 1H), 3.33 (qd, *J* = 6.9, 6.8 Hz, 1H), 2.40 (s, 3H), 2.39-2.27 (m, 3H), 1.99 (ddd, *J* = 13.4, 13.1, 4.9, 1H), 1.87 (ddd, 13.6, 13.2, 5.6 Hz, 1H), 1.54-1.49 (m, 2H), 1.50-1.37 (m, 2H), 1.29 (s, 3H), 1.27-1.23 (m, 1H), 1.18-1.10 (m, 1H), 1.06 (s, 3H), 1.04 (d, J = 6.9 Hz, 3H), 1.02-0.91 (m, 2H), 0.87 (d, *J* = 6.9 Hz, 3H), 0.76-0.67 (m, 1H), 0.66-0.60 (m, 1H), 0.58-0.50 (m, 1H), 0.19 (dd, *J* = 8.7, 4.3 Hz, 1H), 0.15 (dd, *J* = 8.2, 4.7 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 215.2, 172.0, 169.4, 160.9, 159.5, 149.6, 137.3, 136.0, 135.9, 134.0, 133.7, 131.0, 129.1, 128.4, 127.8, 121.0, 119.9, 119.0, 98.7, 77.2, 74.2, 47.8, 41.0, 39.4, 38.5, 34.5, 34.4, 34.3, 30.8, 29.3, 25.7, 24.9, 22.3, 18.6, 17.0, 15.9, 14.5, 12.2, 11.0.


8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 ppm
1.05 2.98 0.78 2.25 1.12 0.98 1.12 0.99 1.00 1.10 1.14 2.68 2.61 1.18 1.17 2.31 3.08 0.88 1.28 5.69 2.02 3.21 1.17 1.17 1.13 2.05
215.19 172.02 169.37 160.90 159.55 149.57 137.29 136.04 135.92 134.04 133.70 130.99 129.06 128.42 127.80 120.97 119.85 119.00 98.69 77.21 74.24 47.83 40.95 39.39 38.49 34.48 34.38 34.30 30.78 29.29 25.67 24.94 22.25 18.59 16.96 15.91 14.55 12.24 11.01
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm

4.3.2 Optimized Synthesis



262

(*S*)-1-(1,3-dithian-2-yl)-3-((4-methoxybenzyl)oxy)propan-2-ol (262). To a solution of 1,3-dithiane (11.6 g, 70.83 mmol) in THF (250 mL) was added *n*-BuLi (1.6 M in hexanes, 55.3 mL, 88.54 mmol) dropwise at -30 °C. The reaction mixture was stirred for 1 h at this temperature and was then allowed to reach 0 °C. Epoxide **261** (11.63 g, 70.83 mmol) in THF (20 mL) was added and stirred for 1 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (150 mL). It was extracted with Et_2O (2 × 75 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 3:1 to 2:1) afforded 17.52 g (79%) of a secondary alcohol **262** as a white solid.

TLC: R_f 0.20 (hexane/EtOAc 3:1, UV (weak), CPS).

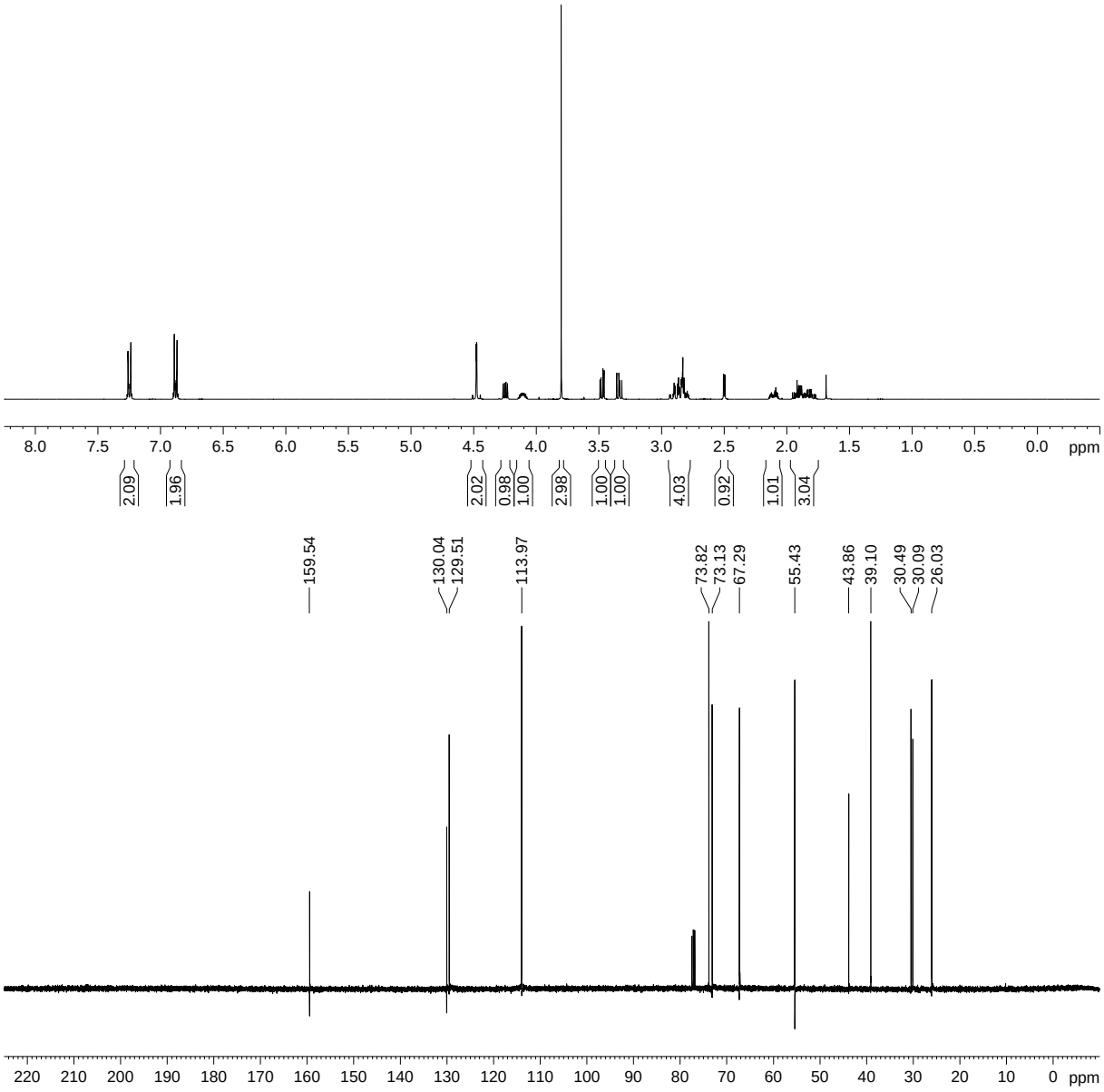
$[\alpha]^{20}_D$: = -4.3 (*c* = 1.68, $CHCl_3$).

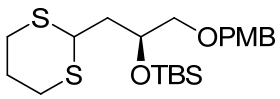
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.25 (dt, *J* = 8.6, 2.4 Hz, 2H), 6.88 (dt, *J* = 8.6, 2.4 Hz, 2H), 4.49 (d, *J* = 11.6 Hz, 1H), 4.46 (d, *J* = 11.6 Hz, 1H), 4.25 (dd, *J* = 9.6, 5.0 Hz, 1H), 4.11 (ddddd, J = 10.1, 7.2, 4.4, 3.7, 3.5 Hz, 1H), 3.80 (s, 3H), 3.47 (dd, *J* = 9.5, 3.5 Hz, 1H), 3.34 (dd, *J* = 9.5, 6.9 Hz, 1H), 2.90 (td, *J* = 14.6, 2.9 Hz, 1H), 2.88-2.77 (m, 3H), 2.50 (d, *J* = 4.4 Hz, 1H), 2.10 (dtt, *J* = 14.1, 5.0, 2.7 Hz 1H), 1.96-1.76 (m, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 159.5, 130.0, 129.5, 114.0, 73.8, 73.1, 67.3, 55.4, 43.9, 39.1, 30.5, 30.1, 26.0.

IR (film): ν □□3448, 2932, 2899, 2858, 2834, 1612, 1585, 1511, 1463, 1422, 1363, 1302, 1276, 1244, 1173, 1105, 1081, 1030, 907, 872, 818, 771, 759, 664.

HRMS (ESI): calculated for $C_{15}H_{22}NaO_3S_2$ $[M+Na]^+$ 337.0903, found 337.0904.





263

(*S*)-((1-(1,3-dithian-2-yl)-3-((4-methoxybenzyl)oxy)propan-2-yl)oxy)(*tert*-butyl)dimethylsilane (263). To a solution of alcohol **262** (17.5 g, 55.65 mmol) in DMF (50 mL) was added imidazole (8.33 g, 122.4 mmol) and TBSCl (9.23 g, 61.22 mmol) and the reaction mixture was stirred for 14 h. The reaction was then quenched by addition of saturated aqueous NH_4Cl (100 mL), H_2O (30 mL) and Et_2O (100 mL). The phases were separated and the aqueous phase was extracted with Et (3 × 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by

flash column chromatography (hexane/EtOAc 9:1) afforded 23.8 g (quantitative) of silyl ether **263** as a colorless oil.

TLC: R_f 0.30 (hexane/EtOAc 9:1, UV, CPS).

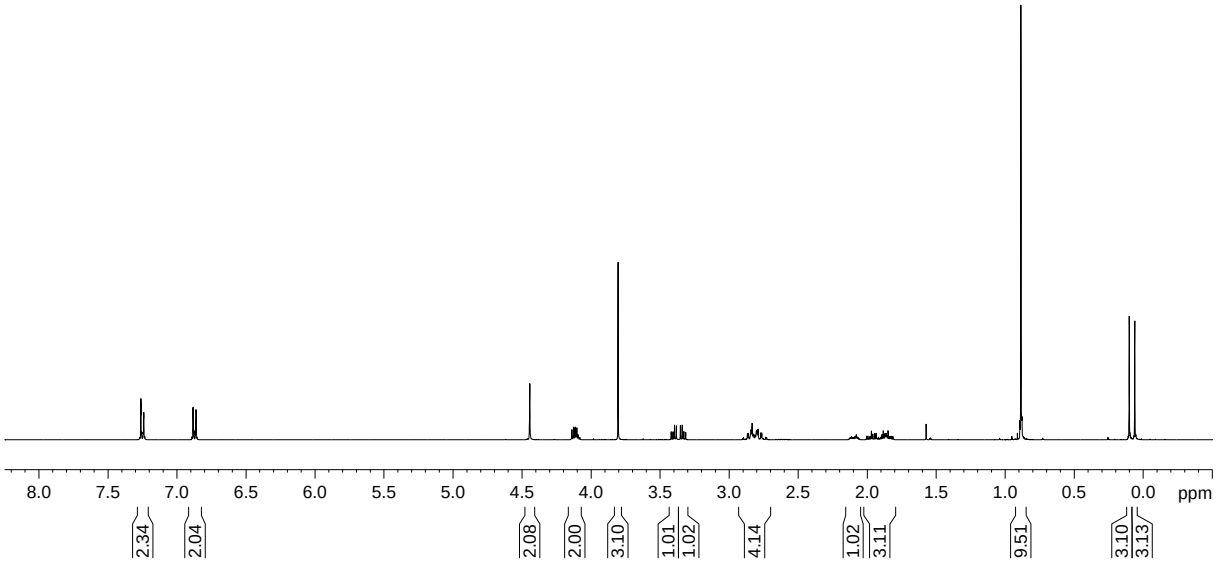
$[\alpha]^{20}_D$: = -19.4 (*c* = 1.21, $CHCl_3$).

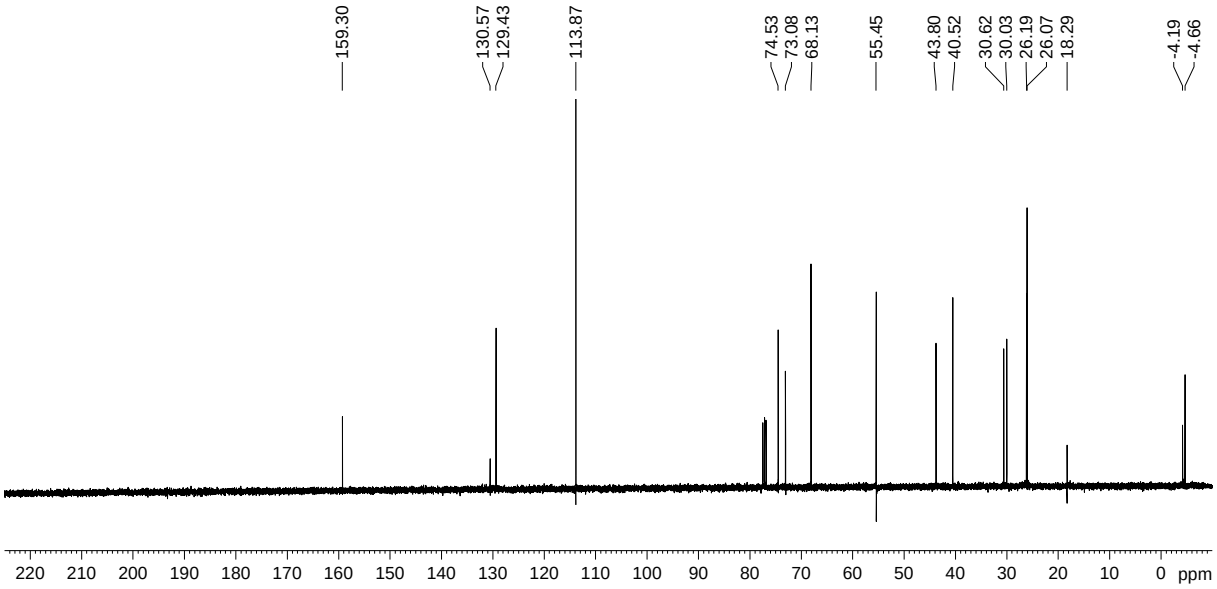
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.25 (dt, *J* = 8.6, 2.4 Hz, 2H), 6.87 (dt, *J* = 8.6, 2.4 Hz, 2H), 4.44 (s, 2H), 4.12 (dd, *J* = 9.5, 5.0 Hz, 2H), 3.80 (s, 3H), 3.40 (dd, *J* = 9.7, 5.3 Hz, 1H), 3.33 (dd, *J* = 9.7, 5.3 Hz, 1H), 2.85 (td, *J* = 14.1, 2.8 Hz, 1H), 2.85-2.79 (m, 2H), 2.77 (td, *J* = 14.1, 2.8 Hz, 1H), 2.10 (dtt, *J* = 14.1, 5.0, 2.8 Hz, 1H), 1.97 (ddd, *J* = 14.0, 9.8, 3.9 Hz, 1H), 1.91-1.80 (m, 2H), 0.89 (s, 9H), 0.10 (s, 3H), 0.06 (s, 3H).

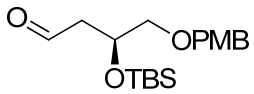
^{13}C-NMR (100 MHz, $CDCl_3$): δ =159.3, 130.6, 129.4, 113.9, 74.5, 73.1, 68.1, 55.5, 43.8, 40.5, 30.6, 30.0, 26.2, 26.1, 18.3, -4.2, -4.7.

IR (film): ν □□2951, 2929, 2897, 2855, 1613, 1512, 1470, 1462, 1422, 1362, 1302, 1246, 1173, 1126, 1108, 1087, 1036, 967, 834, 826, 809, 775, 666.

HRMS (ESI): calculated for $C_{21}H_{36}NaO_3S_2Si$ $[M+Na]^+$ 451.1767, found 451.1765.







264

***(S)*-3-((*tert*-butyldimethylsilyl)oxy)-4-((4-methoxybenzyl)oxy)butanal (264).** To a solution of thioacetal **263** (23.91 g, 55.77 mmol) in THF/H_2O (10:1, 550 mL) was added $CaCO_3$ (14 g, 139.4 mmol) and $Hg(ClO_4)_2 \cdot 3H_2O$ (50 g, 111.3 mmol) in portions over 5 h. The reaction mixture was then diluted with Et_2O (250 mL) and filtrated over a pad of celite. The filtrate filtrated a second time over celite. The phases were separated and the aqueous phase was extracted with Et_2O (2 x 100 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 9:1) afforded 17.01 g (90%) of aldehyde **264** as a colorless oil.

TLC: R_f 0.29 (hexane/EtOAc 9:1, UV, CPS).

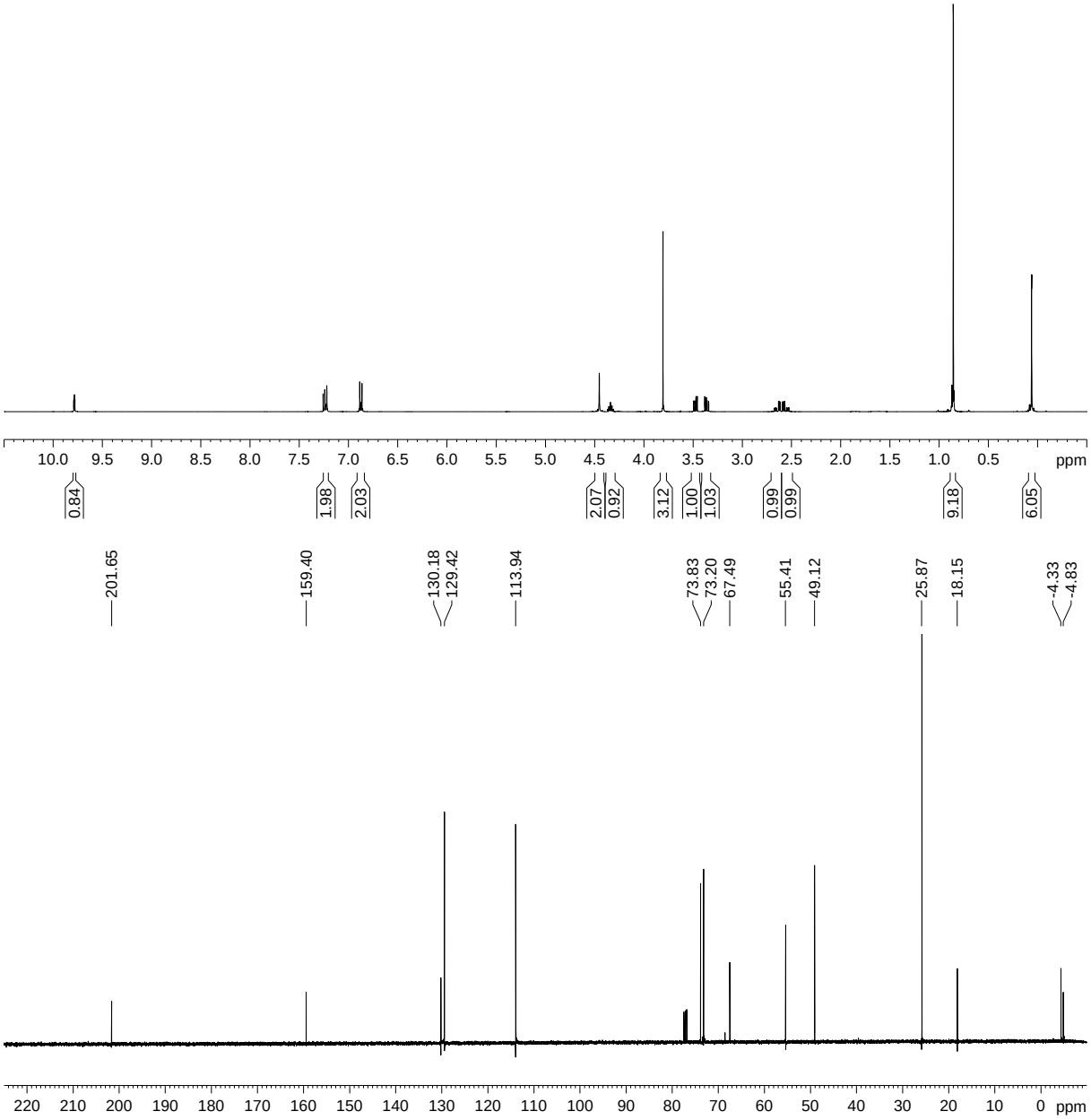
$[\alpha]^{20}_D$: = -12.9 (*c* = 1.50, $CHCl_3$).

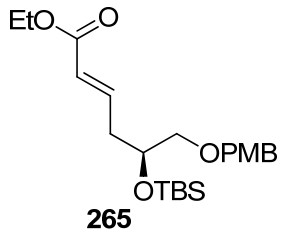
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.79 (dd, *J* = 2.7, 2.2 Hz, 1H), 7.23 (dt, *J* = 8.5, 2.5 Hz, 2H), 6.87 (dt, *J* = 8.5, 2.5 Hz, 2H), 4.45 (s, 2H), 4.34 (ddd, *J* = 11.5, 6.5, 5.1 Hz, 1H), 3.81 (s, 3H), 3.47 (dd, *J* = 9.6, 5.1 Hz, 1H). 3.36 (dd, *J* = 9.6, 6.4 Hz, 1H), 2.64 (ddd, *J* = 15.9, 5.1, 2.1 Hz, 1H), 2.56 (ddd, *J* = 15.9, 6.6, 2.7 Hz, 1H), 0.86 (s, 9H), 0.06 (s, 3H), 0.06 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 201.7, 159.4, 130.2, 129.4, 113.9, 73.8, 73.2, 67.5, 55.4, 49.1, 25.9, 18.1, -4.3, -4.8.

IR (film): ν □□2953, 2929, 2897, 2856, 1726, 1613, 1513, 1471, 1463, 1362, 1302, 1247, 1173, 1098, 1034, 1006, 833, 777.

HRMS (ESI): calculated for $C_{18}H_{30}NaO_4Si$ $[M+Na]^+$ 361.1806, found 361.1809.





265

(*S*,*E*)-ethyl 5-((*tert*-butyldimethylsilyl)oxy)-6-((4-methoxybenzyl)oxy)hex-2-enoate (265). To a solution of aldehyde **264** (17.0 g, 50.22 mmol) in CH_2Cl_2 (250 mL) was added (Triphenylphosphoranylidene)acetic acid ethyl ester (17.5 g, 50.22 mmol) in one portion at

0 °C. The reaction mixture was stirred for 3 h at 0 °C. A second portion of (Triphenylphosphoranylidene)acetic acid ethyl ester (2.6 g, 7.53 mmol) was added and the reaction mixture allowed to reach rt and stirred for additional 16 h at this temperature. The solution was concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 9:1) afforded 17.68 g (86%) of α,β-unsaturated ester **265** as a colorless oil.

TLC: R_f 0.37 (hexane/EtOAc 9:1, UV, CPS).

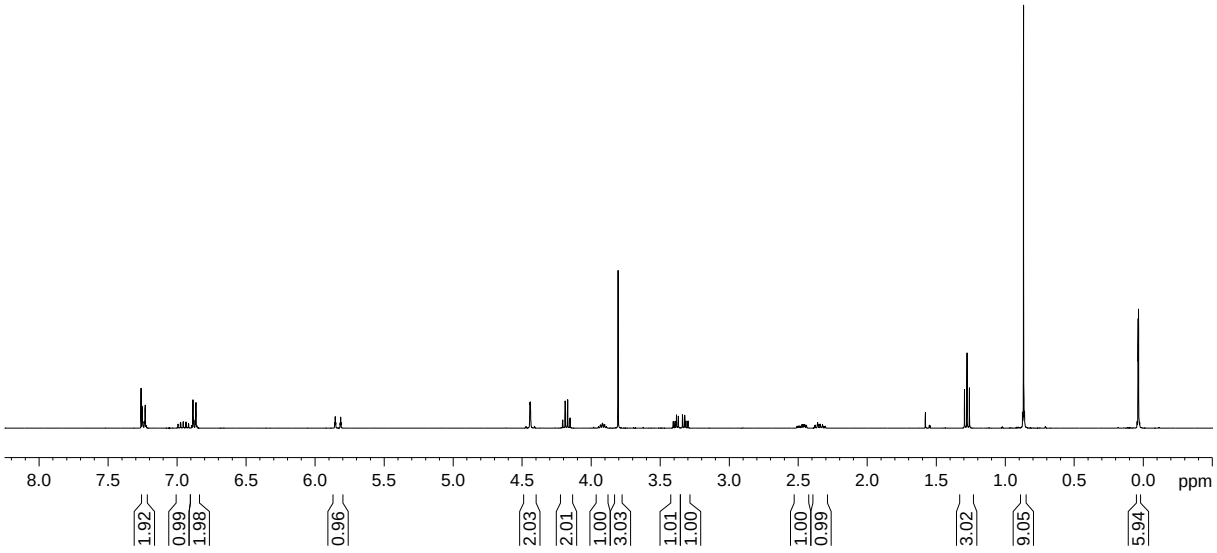
[α]$^{20}_{D}$: = -12.1 (*c* = 0.80, $CHCl_3$).

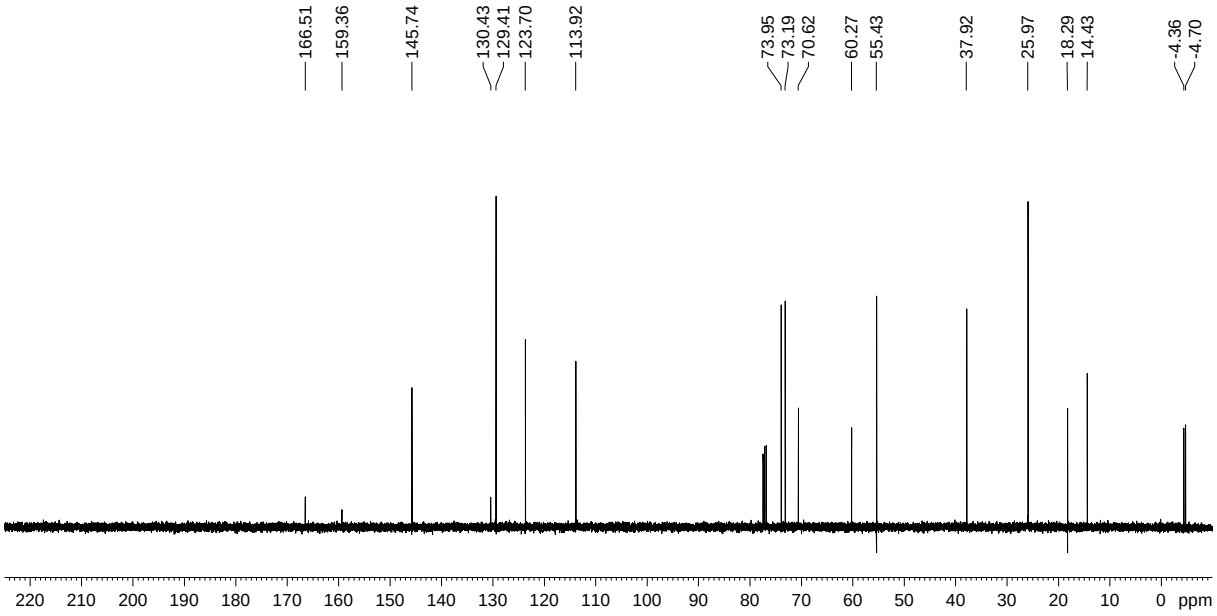
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.23 (dt, *J* = 8.6, 2.5 Hz, 2H), 6.96 (ddd, *J* = 15.4, 7.9, 7.4 Hz, 1H), 6.87 (dt, *J* = 8.6, 2.5 Hz, 2H), 5.83 (dt, *J* = 15.7, 1.4 Hz, 1H), 4.46 (d, *J* = 11.6 Hz, 1H), 4.43 (d, *J* = 11.6 Hz, 1H), 4.18 (q, *J* = 7.1 Hz, 2H), 3.92 (ddd, *J* = 11.5, 6.2, 5.3 Hz, 1H), 3.81 (s, 3H), 3.39 (dd, *J* = 9.5, 5.3 Hz, 1H), 3.32 (dd, *J* = 9.5, 6.2 Hz, 1H), 2.47 (dddd, *J* = 14.2, 7.1, 4.7, 1.5 Hz, 1H), 2.34 (dddd, *J* = 14.2, 7.9, 6.9, 1.3 Hz, 1H), 1.28 (t, *J* = 7.2 Hz, 3H), 0.87 (s, 9H), 0.04 (s, 3H), 0.04 (s, 3H).

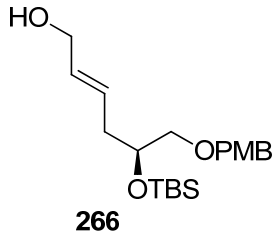
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 166.5, 159.4, 145.7, 130.4, 129.4, 123.7, 113.9, 74.0, 73.2, 70.6, 60.3, 55.4, 37.9, 26.0, 18.3, 14.4, -4.4, -4.7.

IR (film): ν □□2954, 2930, 2899, 2857, 1719, 1655, 1613, 1513, 1472, 1464, 1366, 1317, 1302, 1248, 1210, 1172, 1097, 1037, 1006, 985, 836, 810.

HRMS (ESI): calculated for $C_{22}H_{36}NaO_5Si$ $[M+Na]^+$ 431.2224, found 431.2226.







266

(*S*,*E*)-5-((*tert*-butyldimethylsilyl)oxy)-6-((4-methoxybenzyl)oxy)hex-2-en-1-ol (266). To a solution of α,β-unsaturated ester **265** (17.68 g, 43.27 mmol) in CH_2Cl_2 (250 mL) was added DIBAL (1 M in CH_2Cl_2, 100 mL, 99.52 mmol) dropwise over 30 min at -78 °C and the reaction mixture was stirred for 30 min at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle solution (300 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). Phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 3:1) afforded 14.88 g (94%) of allylic alcohol **266** as a colorless liquid.

TLC: R_f 0.33 (hexane/EtOAc 3:1, UV, CPS).

$[\alpha]^{20}_D$: = -6.00 (*c* = 0.92, $CHCl_3$).

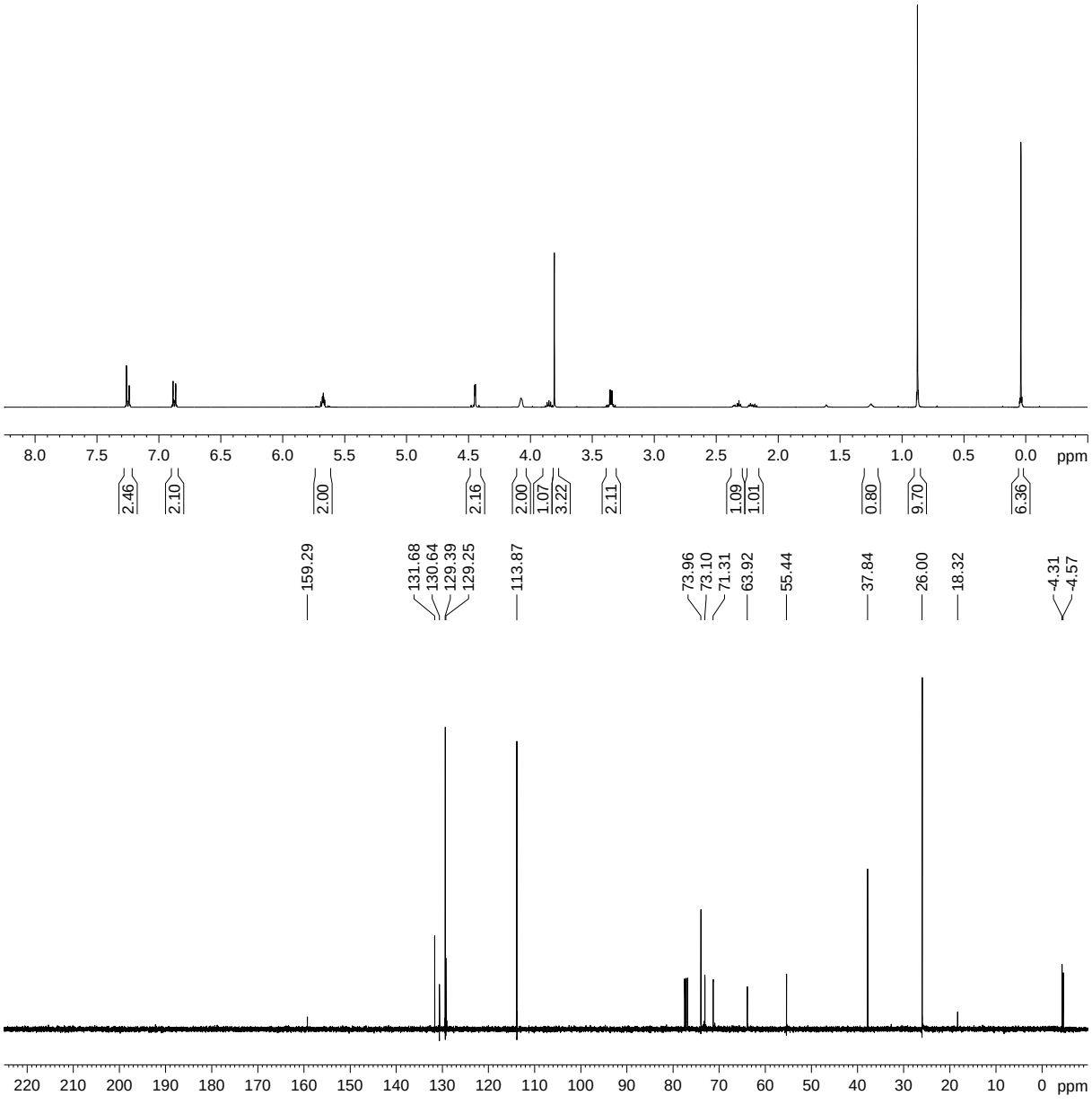
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.25 (dt, *J* = 8.6, 2.6 Hz, 2H), 6.87 (dt, *J* = 8.6, 2.6 Hz, 2H), 5.70 (dd, *J* = 15.5, 5.8 Hz, 1H), 5.65 (dd, *J* = 15.5, 4.8 Hz, 1H), 4.46 (d, *J* = 11.6 Hz, 1H), 4.46 (d, *J* = 11.6 Hz, 1H), 4.07 (br s, 2H), 3.85 (dt, *J* = 11.6, 5.6 Hz, 1H), 3.80 (s, 3H), 3.35

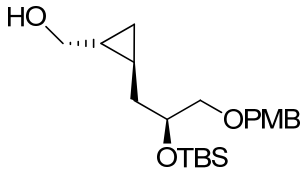
(dd, *J* = 5.5, 2.7 Hz, 2H), 2.39-2.29 (m, 1H), 2.25-2.16 (m, 1H) 1.25 (br s, 1H), 0.87 (s, 9H), 0.04 (s, 3H), 0.04 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 159.3, 131.7, 130.6, 129.4, 129.2, 113.9, 74.0, 73.1, 71.3, 63.9, 55.4, 37.8, 26.0, 18.3, -4.3, -4.6.

IR (film): ν □□3374, 2953, 2928, 2897, 2856, 1613, 1513, 1471, 1462, 1361, 1302, 1247, 1173, 1101, 1036, 1005, 972, 831, 811, 776, 668.

HRMS (ESI): calculated for $C_{20}H_{34}NaO_4Si$ $[M+Na]^+$ 389.2119, found 389.2121.





267

((1*R*,2*S*)-2-((*S*)-2-((*tert*-butyldimethylsilyl)oxy)-3-((4-methoxybenzyl)oxy)propyl) cyclopropyl)methanol (267). To a solution of $ZnEt_2$ (1 M in hexane, 10.0 mL, 10.0 mmol) in CH_2Cl_2 (32 mL) was added CH_2I_2 (1.6 mL, 20.0 mmol) carefully dropwise at 0 °C. The mixture was stirred at 0 °C for 20 min and a preformed mixture of (+)-(*S*,*S*)-2-butyl-*N*,*N*,*N'*,*N'*-tetramethyl-1,3,2-dioxaborolane-4,5-dicarboxamide (Charette ligand, 0.83 mg, 3.06 mmol) and allylic alcohol **266** (0.92 g, 2.51 mmol) in CH_2Cl_2 (8 mL) was added *via* syringe. The reaction mixture was stirred for 4 h at 0 °C and then quenched by addition of saturated aqueous NH_4Cl (75 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (2 x 50 mL). The combined organic extracts were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 5:1) provided 0.98 g of cyclopropyl alcohol **267** (quantitative yield) in a diastereomeric ratio of 15:1 as a colorless oil.

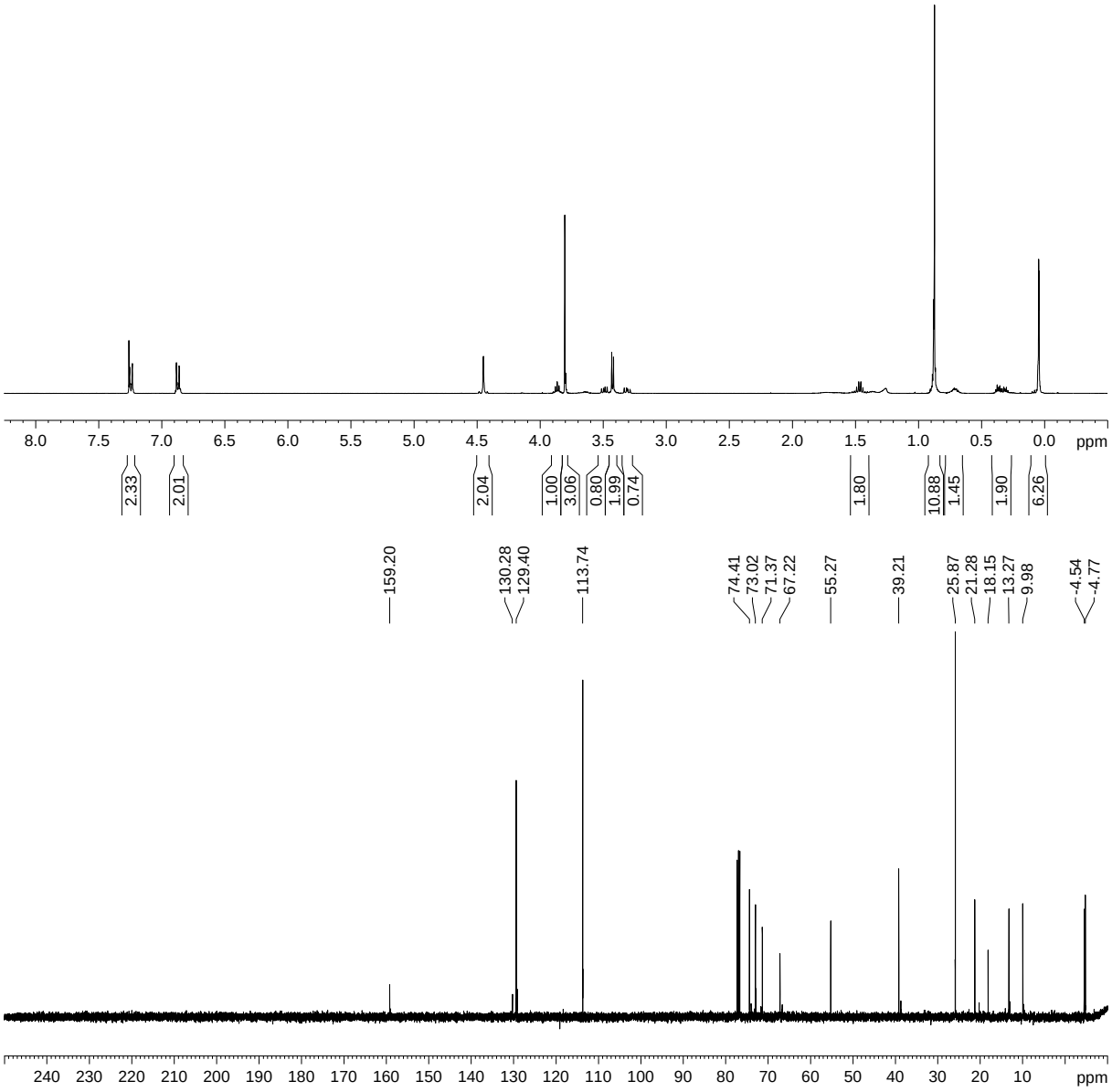
TLC: R_f 0.33 (hexane/EtOAc 3:1, UV, CPS).

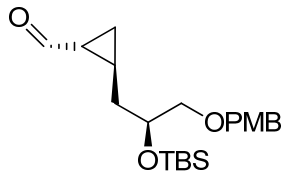
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.25 (dt, *J* = 8.6, 2.5 Hz, 1H), 6.87 (dd, *J* = 8.6, 2.5 Hz, 1H), 4.46 (d, *J* = 11.6 Hz, 1H), 4.43 (dd, *J* = 11.6 Hz, 1H), 3.86 (dd, *J* = 11.6, 5.8 Hz, 1H), 3.81 (s, 3H), 3.49 (dd, *J* = 11.2, 6.5 Hz, 1H), 3.42 (d, *J* = 5.6 Hz, 2H), 3.31 (dd, J = 11.1, 7.5 Hz, 1H), 1.49 (dd, *J* = 13.9, 6.7 Hz, 1H), 1.43 (dd, *J* = 13.9, 6.7 Hz, 1H), 0.87 (s, 9H), 0.77-0.66 (m, 2H), 0.36 (dt, *J* = 8.4, 4.7 Hz, 1H), 0.31 (dt, *J* =, 8.2, 5.1 Hz, 1H), 0.05 (s, 3H), 0.04 (s, 3H)

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 159.2, 130.3, 129.4, 113.7, 74.4, 73.0, 71.4, 67.2, 55.3, 39.2, 25.9, 21.3, 18.2, 13.3, 10.0, -4.5, -4.8.

IR (film): ν □□3419, 2953, 2927, 2856, 1613, 1587, 1513, 1470, 1463, 1361, 1302, 1247, 1173, 1085, 1055, 1036, 1006, 834, 809, 775, 666.

HRMS (ESI): calculated for $C_{21}H_{36}NaO_4Si$ $[M+Na]^+$ 403.2275, found 403.2272.





268

(1*R*,2*S*)-2-((*S*)-2-((*tert*-butyldimethylsilyl)oxy)-3-((4-methoxybenzyl)oxy)propyl) cyclopropanecarbaldehyde (268). To a solution of alcohol **267** (4.18 g, 10.98 mmol) in CH_2Cl_2 (75 ml) was added DMP (15%, 22.83 mL, 10.98 mmol) at 0 °C, the reaction mixture was allowed to reach room temparature and stirred for 1h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (50 mL) and saturated aqueous $Na_2S_2O_3$ (50 mL). Stirring was continued for 30 min, when two almost clear phases had formed. The

phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 60 mL). The combined organic phases were washed with brine (100 mL), dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (hexane/EtOAc (9:1) afforded 3.91 g (94%) of aldehyde **268** as a colorless oil.

TLC: R_f 0.33 (hexane/EtOAc 12:1, UV, CPS).

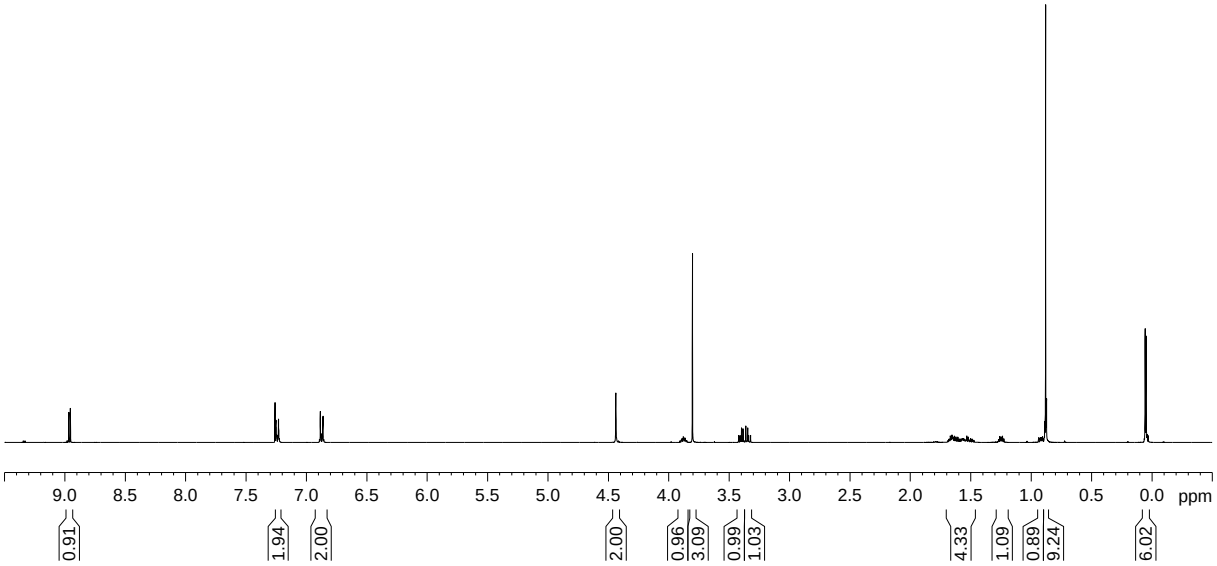
$[\alpha]^{20}_D$: = -32.3 (*c* = 0.93, $CHCl_3$).

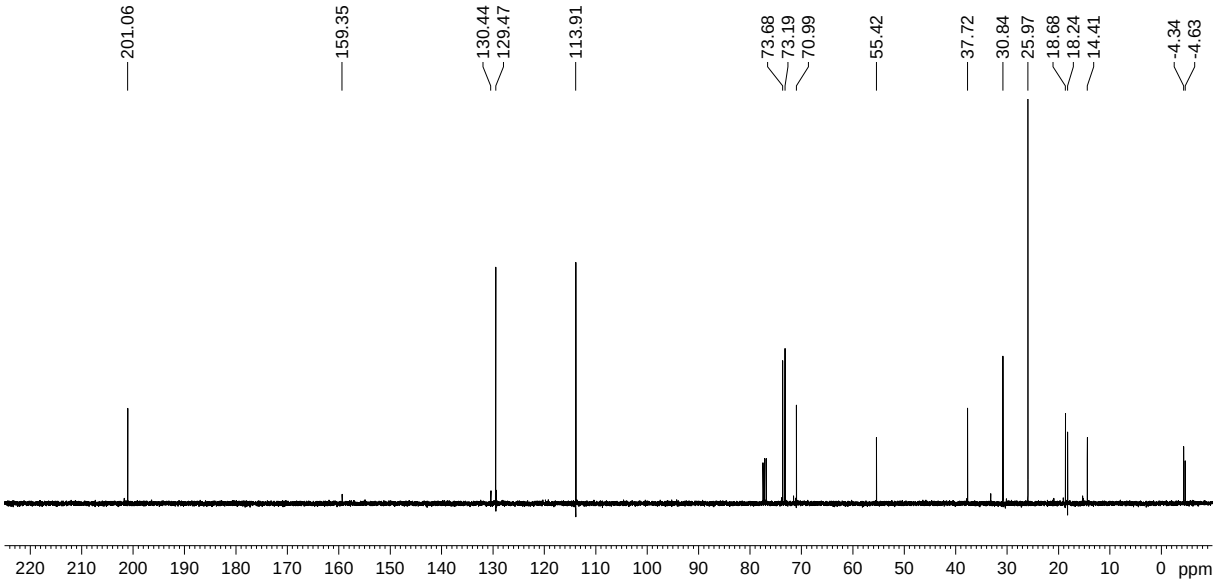
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.96 (d, *J* = 5.6 Hz, 1H), 7.25 (dt, *J* = 8.6, 2.5 Hz, 1H), 6.87 (dd, *J* = 8.6, 2.5 Hz, 1H), 4.44 (s, 2H), 3.88 (ddd, *J* = 11.1, 6.3, 5.1 Hz, 1H), 3.81 (s, 3H), 3.41 (dd, *J* = 9.5, 5.1 Hz, 1H), 3.34 (dd, *J* = 9.5, 6.3 Hz, 1H), 1.69-1.53 (m, 3H), 1.50 (ddd, 12.4, 6.7, 4.4 Hz, 1H), 1.25 (ddd, *J* = 8.3, 5.1, 4.6 Hz, 1H), 0.91 (ddd, *J* = 8.3, 6.1, 4.6 Hz, 1H), 0.88 (s, 9H), 0.06 (s, 3H), 0.05 (s, 3H).

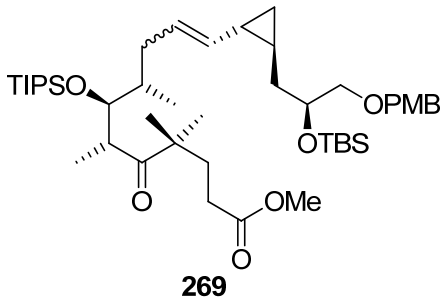
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 201.1, 159.4, 130.4, 129.5, 113.9, 73.7, 73.2, 71.0, 55.4, 37.7, 30.8, 26.0, 18.7, 18.2, 14.4, -4.3, -4.6.

IR (film): ν □□□2953, 2929, 2897, 2897, 2856, 1706, 1613, 1513, 1470, 1463, 1362, 1302, 1247, 1173, 1122, 1086, 1036, 1006, 834, 809, 776, 666.

HRMS (ESI): calculated for $C_{21}H_{34}NaO_4Si$ $[M+Na]^+$ 401.2119, found 401.2115.







269

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-2-((*tert*-butyldimethylsilyl)oxy)-3-((4-methoxybenzyl)oxy)propyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undec-10-enoate (269). To a solution of sulfone **227** (3.07 g, 4.82 mmol) and aldehyde **268** (2.19 g, 5.78 mmol) in THF (48 mL) was added LiHMDS (1M in THF, 5.8 mL, 5.78 mmol) dropwise at -78 °C and the reaction mixture was stirred for 40 min at this temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (40 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 30 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 9:1) afforded 3.50 g (92%, *E*/*Z* 4.5:1) of olefine **269** as a colorless oil.

Note: The NMR data are given for the major (*E*) isomer only.

TLC: R_f 0.30 (hexane/EtOAc 9:1, CPS).

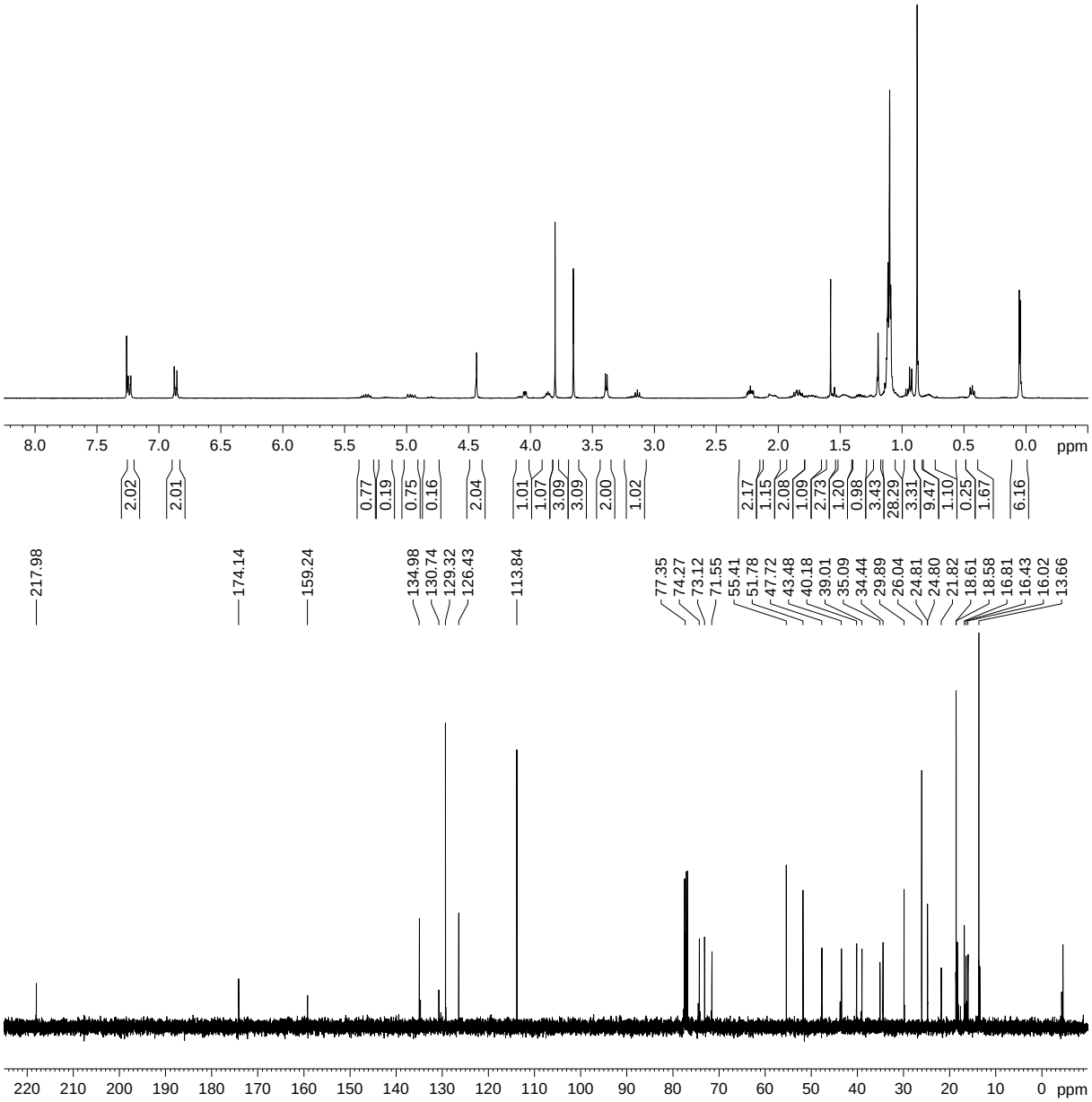
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.26-7.21 (m, 2H), 6.87 (dt, *J* = 8.6, 2.5 Hz, 2H), 5.33 (ddd, *J* = 15.1, 7.9, 6.6 Hz, 1H), 4.96 (dd, *J* = 15.1, 8.5 Hz, 1H), 4.44 (s, 2H), 4.05 (dd, *J* = 6.7, 2.2 Hz, 1H), 3.86 (ddd, *J* = 10.8, 6.1 5.6 Hz, 1H), 3.80 (s, 3H), 3.65 (s, 3H), 3.39 (d,

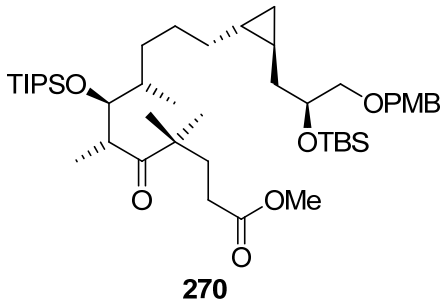
J = 5.6 Hz, 2H), 3.15 (qd, *J* = 6.9, 6.8 Hz, 1H), 2.24 (dd, *J* = 6.4, 4.6 Hz, 1H), 2.21 (dd, *J* = 6.1, 3.8 Hz, 1H), 2.10-2.00 (m, 1H), 1.93-1.76 (m, 2H), 1.79-1.65 (m, 1H), 1.61-1.52 (m, 1H), 1.52-1.40 (m, 1H), 1.34 (ddd, *J* = 14.1, 7.5, 5.0 Hz, 1H), 1.19 (s, 3H), 1.14-1.07 (m, 28H), 0.93 (d, *J* = 6.9 Hz, 3H), 0.88 (s, 9H), 0.84-0.73 (m, 1H), 0.44 (m, 2H), 0.05 (s, 3H), 0.05 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 159.2, 135.0, 130.7, 129.3, 126.4, 113.8, 77.4, 74.3, 73.1, 71.5, 55.4, 51.8, 47.7, 43.5, 40.2, 39.0, 35.1, 34.4, 29.9, 26.0, 24.8, 24.8, 21.8, 18.6, 18.6, 16.8, 16.4, 16.0, 13.7, 13.4, -4.3, -4.6.

IR (film): ν 2947, 2865, 1741, 1696, 1623, 1513, 1463, 1438, 1388, 1364, 1301, 1248, 1173, 1122, 1097, 1056, 1036, 1005, 990, 978, 883, 834, 776, 678.

HRMS (ESI): calculated for $C_{45}H_{80}NO_7Si_2$ $[M+NH_4]^+$ 806.5781, found 806.5776.





270

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-2-((*tert*-butyldimethylsilyl)oxy)-3-((4-methoxybenzyl)oxy)propyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoate (270). To a solution of olefine **269** (2.2 g, 2.59 mmol) and *o*-nitrobenzenesulfonylhydrazide (12.11 g, 55.75 mmol) in CH_2Cl_2 (150 mL) was added NEt_3 (7.75 mL, 55.75 mmol) over a period of 4 h at room temperature. The reaction mixture was left stirring for 12 h, was then diluted with hexane/EtOAc (3:1, 50 mL) and filtered through a plug of silica. The precipitate was washed extensively with hexane/EtOAc (3:1, 200 mL) and the filtrate was concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 9:1) afforded 2.10 g (95%) of silyl ether **270** as a colorless oil.

TLC: R_f 0.30 (hexane/EtOAc 9:1, CPS).

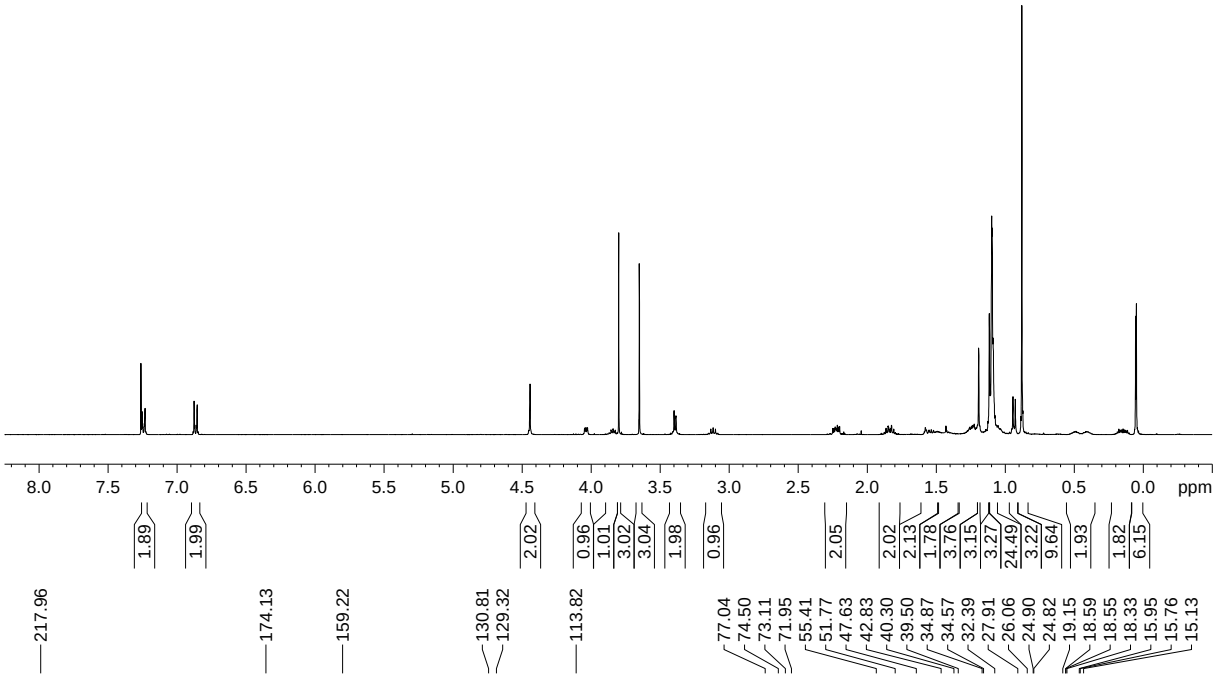
$[\alpha]^{20}_D$: = -20.2 (*c* = 1.10, $CHCl_3$).

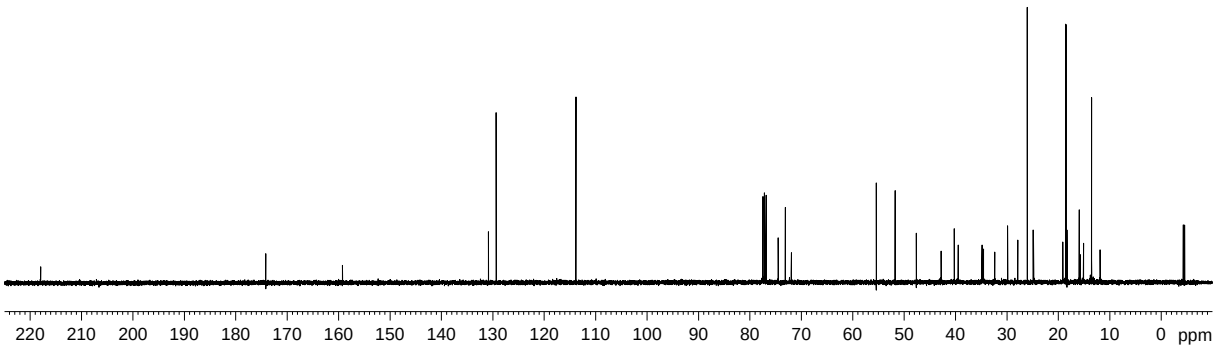
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.24 (dt, *J* = 8.6, 2.4 Hz, 2H), 6.86 (dd, *J* = 8.6, 2.4 Hz, 2H), 4.44 (s, 2H), 4.04 (dd, *J* = 6.1, 2.6 Hz, 1H), 3.84 (dq, *J* = 6.4, 5.4 Hz, 1H), 3.80 (s, 3H), 3.65 (s, 3H), 3.39 (d, *J* = 5.5 Hz, 2H), 3.12 (qd, *J* = 7.0, 6.0 Hz, 1H), 2.24 (dd, *J* = 8.8, 3.8 Hz, 1H), 2.21 (dd, *J* = 8.8, 3.1 Hz, 1H), 1.86 (dd, *J* = 10.8, 4.8 Hz, 1H), 1.82 (dd, *J* = 10.8, 4.5 Hz, 1H), 1.58-1.46 (m, 2H), 1.48-1.35 (m, 2H), 1.29-1.20 (m, 4H), 1.19 (s, 3H), 1.12 (s, 3H), 1.12-1.07 (m, 24H), 1.05-1.02 (m, 1H), 0.94 (d, *J* = 6.9 Hz, 3H), 0.88 (s, 9H), 0.50 (dddd, *J* = 12.7, 8.0, 5.0, 4.5 Hz, 1H), 0.45-0.36 (m, 1H), 0.17 (dd, *J* = 8.0, 4.7 Hz, 1H), 0.12 (dd, *J* = 8.89, 4.4 Hz, 1H), 0.05 (s, 3H), 0.05 (s, 3H).

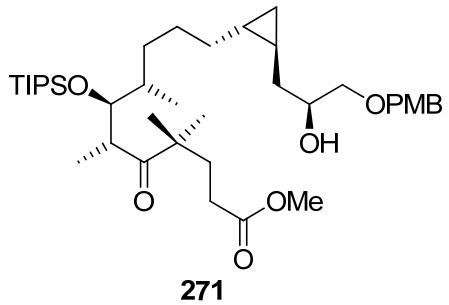
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 159.2, 130.8, 129.3, 113.8, 77.0, 74.5, 73.1, 71.9, 55.4, 51.8, 47.6, 42.8, 40.3, 39.5, 34.9, 34.6, 32.4, 27.9, 26.1, 24.9, 24.8, 19.2, 18.6, 18.6, 18.3, 15.9, 15.8, 15.1, 13.5, 11.9, -4.3, -4.6.

IR (film): ν □□2928, 2864, 1742, 1697, 1613, 1513, 1463, 1438, 1388, 1365, 1301, 1248, 1173, 1120, 1092, 1038, 1005, 992, 978, 883, 834, 810, 776, 677.

HRMS (ESI): calculated for $C_{45}H_{86}NO_7Si_2$ $[M+NH_4]^+$ 808.5937, found 808.5929.







271

(6*R*,7*S*,8*S*)-methyl 11-((1*R*,2*S*)-2-((*S*)-2-hydroxy-3-((4-methoxybenzyl)oxy)propyl) cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoate (271). To a solution of silyl ether **270** (3.99 g, 5.04 mmol) in CH_2Cl_2 (166 mL) and MeOH (84 mL) was added CSA (2.34 g, 10.1 mmol) in one portion at room temperature. The reaction mixture was

stirred for 2 h and was then quenched by addition of saturated aqueous $NaHCO_3$ (150 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 5:1) afforded 3.03 g (89%) of secondary alcohol **270** as a colorless oil.

TLC: R_f 0.25 (hexane/EtOAc 3:1, CPS).

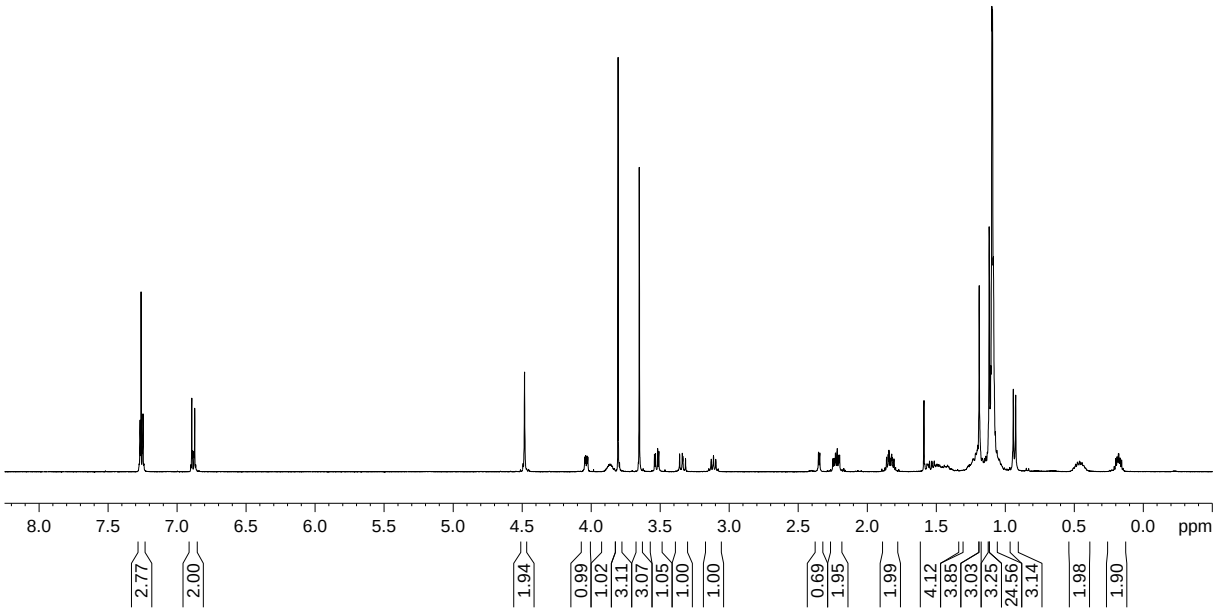
$[\alpha]^{20}_D$: = -17.9 (*c* = 0.81, $CHCl_3$).

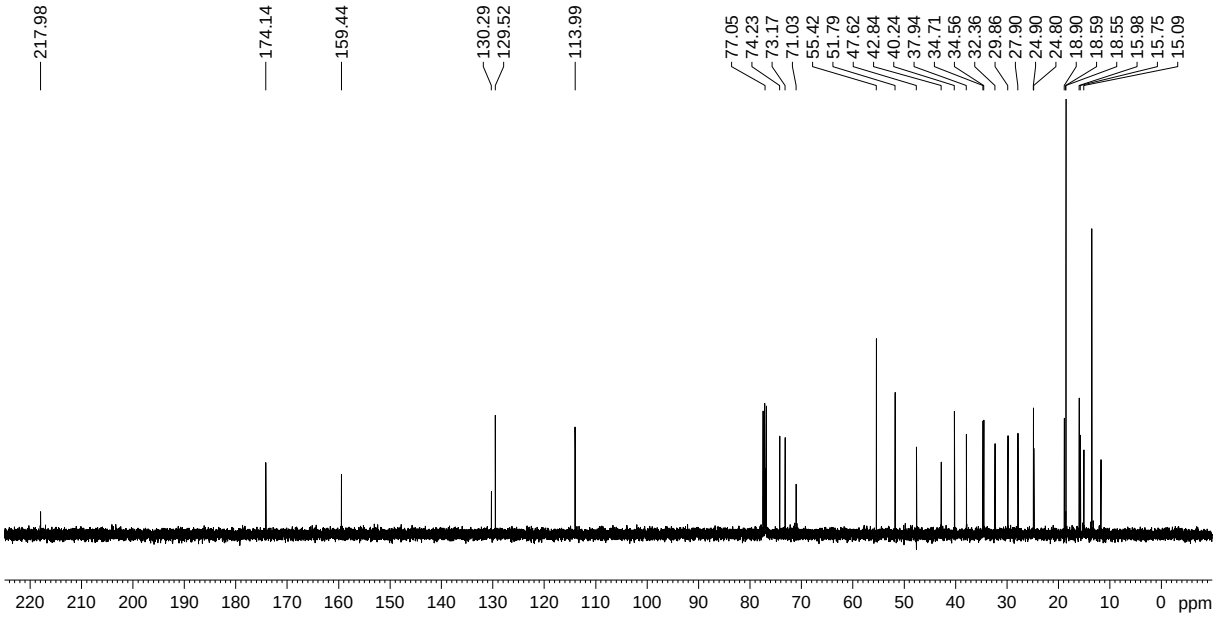
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.26 (dt, J = 8.6, 2.4 Hz, 2H), 6.88 (dd, J = 8.6, 2.4 Hz, 2H), 4.50 (d, *J* = 11.6 Hz, 1H), 4.47 (d, *J* = 11.6 Hz, 1H), 4.04 (dd, J = 6.2, 2.6 Hz, 1H), 3.91-3.82 (m, 1H), 3.81 (s, 3H), 3.65 (s, 3H), 3.53 (dd, *J* = 9.5, 3.1 Hz, 1H), 3.34 (dd, *J* = 9.5, 7.7 Hz, 1H), 3.11 (qd, *J* = 6.9, 6.1 Hz, 1H), 2.35 (d, *J* = 3.5 Hz, 1H), 2.24 (dd, *J* = 8.4, 3.5 Hz, 1H), 2.21 (dd, *J* = 8.4, 2.6 Hz, 1H), 1.85 (dd, *J* = 6.9, 4.7 Hz, 1H), 1.82 (dd, *J* = 6.9, 5.1, 1H), 1.55-1.36 (m, 4H), 1.28-1.19 (m, 4H), 1.19 (s, 3H), 1.17-1.12 (m, 2H), 1.13-1.07 (m, 24H), 1.07-1.01 (m, 2H), 0.93 (d, *J* = 6.9 Hz, 3H), 0.53-0.39 (m, 2H), 0.22-0.14 (m, 2H).

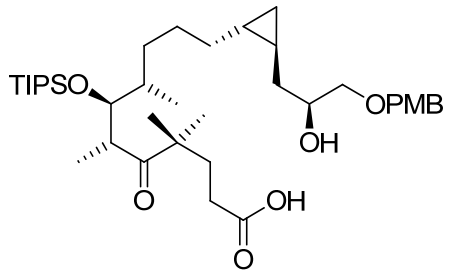
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.0, 174.1, 159.4, 130.3, 129.5, 114.0, 77.1, 74.2, 73.2, 71.0, 55.4, 51.8, 47.6, 42.8, 40.2, 37.9, 34.7, 34.6, 32.4, 29.9, 27.9, 24.9, 24.8, 18.9, 18.6, 18.6, 16.0, 15.7, 15.1, 13.5, 11.8.

IR (film): ν □□3479, 2941, 2865, 1739, 1696, 1613, 1513, 1463, 1438, 1388, 1367, 1301, 1247, 1205, 1173, 1119, 1091, 1065, 1035, 1012, 990, 883, 846, 821, 677.

HRMS (ESI): calculated for $C_{39}H_{68}NO_7Si$ $[M+NH_4]^+$ 694.5073, found 694.5062.







272

(6*R*,7*S*,8*S*)-11-((1*R*,2*S*)-2-((*S*)-2-hydroxy-3-((4-methoxybenzyl)oxy)propyl)cyclopropyl)-4,4,6,8-tetramethyl-5-oxo-7-((triisopropylsilyl)oxy)undecanoic acid (272). To a solution of methyl ester **271** (3.03 g, 4.48 mmol) in *t*-BuOH (90 mL) and H_2O (22 mL) was added LiOH monohydrate (1.13 g, 26.85 mmol) in one portion at room temperature. The reaction mixture was stirred for 2 h and then diluted with water (50 mL) and CH_2Cl_2 (50 mL) whereupon the solution turned milky. The solution was then acidified by addition of 1 M aqueous HCl solution until pH 5 was reached. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced to yield 2.96 g (100%) of acid **272** as a colorless oil.

TLC: R_f : streak (hexane/EtOAc 2:1, UV, CPS).

$[\alpha]^{20}_D$: = -10.9 (*c* = 0.80, $CHCl_3$).

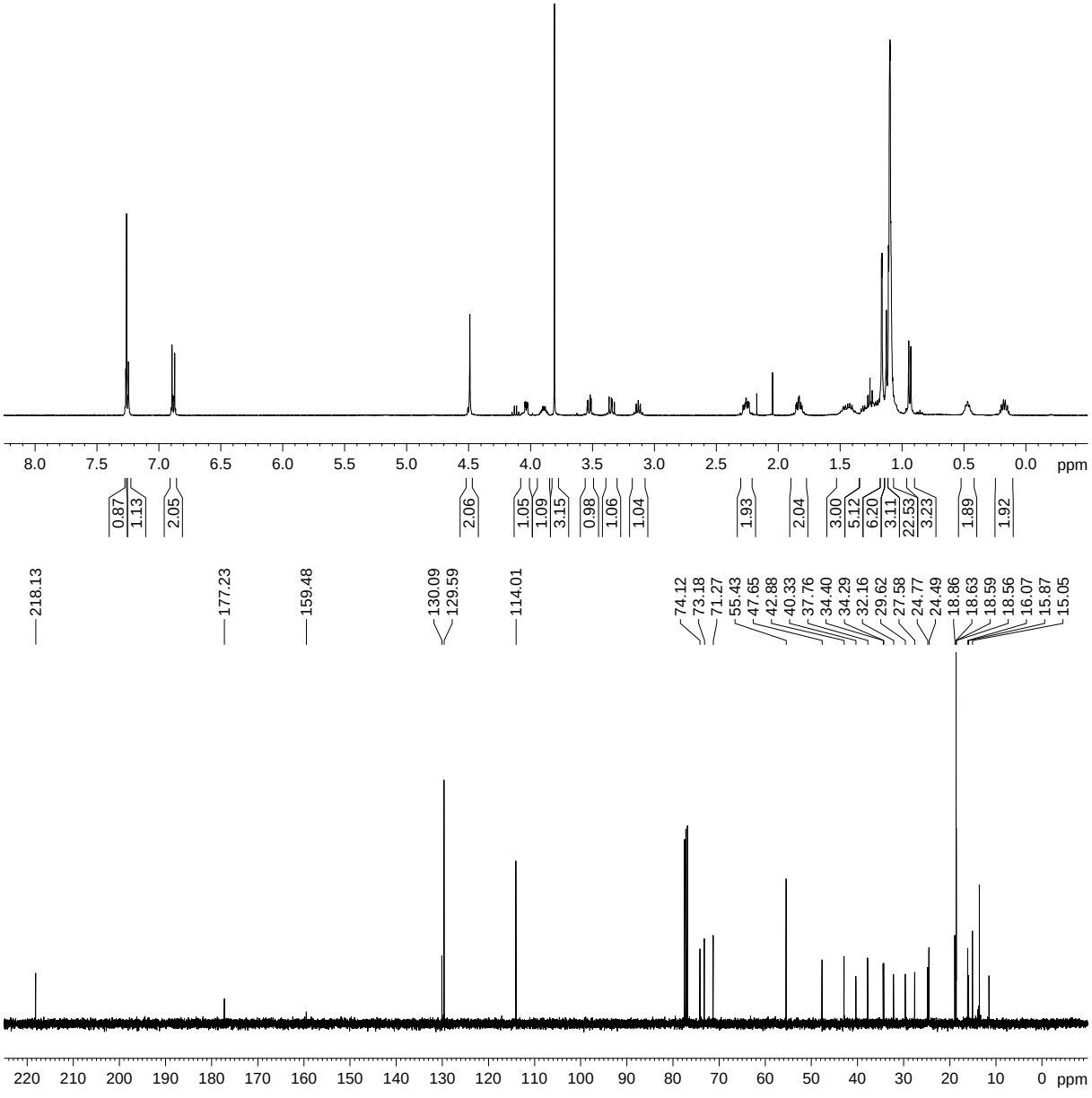
[^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.26 (dt, *J* = 8.6, 2.5 Hz, 2H), 6.88 (dt, *J* = 8.6, 2.5 Hz, 2H), 4.49 (s, 2H), 4.03 (dd, *J* = 6.2, 2.5 Hz, 1H), 3.89 (dddd, *J* = 10.5, 5.5, 5.4, 3.2 Hz, 1H), 3.80 (s, 3H), 3.53 (dd, *J* = 9.5, 3.1 Hz, 1H), 3.34 (dd, *J* = 9.5, 7.8 Hz, 1H), 3.13 (qd, *J* = 7.0,

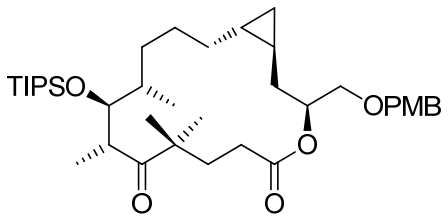
6.0 Hz, 1H), 2.27 (dd, *J* = 9.1, 3.5 Hz, 1H), 2.25 (dd, *J* = 5.5, 2.5 Hz, 1H), 1.84 (dd, *J* = 7.5, 3.1 Hz, 1H), 1.82 (dd, *J* = 9.1, 3.6 Hz, 1H), 1.50-1.36 (m, 3H), 1.33-1.17 (m, 5H), 1.16 (s, 3H), 1.16 (s, 3H), 1.13 (s, 3H), 1.11-1.07 (m, 22H), 0.94 (d, *J* = 7.0 Hz, 3H), 0.51-0.42 (m, 2H), 0.19 (dd, *J* = 10.4, 4.3 Hz, 1H), 0.15 (dd, *J* = 11.0, 4.3 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 177.2, 159.5, 130.1, 129.6, 114.0, 74.1, 73.2, 71.3, 55.4, 47.7, 42.9, 40.3, 37.8, 34.4, 34.3, 32.2, 29.6, 27.6, 24.8, 24.5, 18.9, 18.6, 18.6, 18.6, 16.1, 15.9, 15.1, 13.6, 11.5.

IR (film): ν □2962, 2939, 2865, 1697, 1613, 1513, 1464, 1387, 1366, 1301, 1248, 1173, 1114, 1090, 1065, 1036, 1014, 998, 980, 883, 847, 820, 735, 677.

HRMS (ESI): calculated for $C_{38}H_{66}NaO_7Si$ $[M+Na]^+$ 685.4470, found 685.4471.





273

(1*S*,3*S*,10*R*,11*S*,12*S*,16*R*)-3-(((4-methoxybenzyl)oxy)methyl)-8,8,10,12-tetramethyl-11-((triisopropylsilyl)oxy)-4-oxabicyclo[14.1.0]heptadecane-5,9-dione (273). To a solution of 2-methyl-6-nitrobenzoic anhydride (0.80 g, 2.32 mmol) and DMAP (0.57 g, 4.64 mmol) in CH_2Cl_2 (480 mL) was added seco acid **272** (1.02 g, 1.55 mmol) in CH_2Cl_2 (290 mL) over a period of 15 h using a dropping funnel and the reaction mixture was stirred for another 30 min. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (500 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 250 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 9:1) to yield 0.93 g (94%) of macrolactone **273** as a colorless oil.

TLC: R_f 0.20 (hexane/EtOAc 9:1, CPS).

$[\alpha]^{20}_D$: = +15.3 (*c* = 0.87, $CHCl_3$).

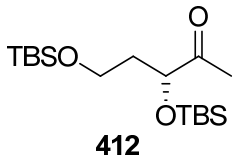
[^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.23 (dt, *J* = 8.6, 2.5 Hz, 2H), 6.87 (dt, *J* = 8.6, 2.5 Hz, 2H), 5.21 (dddd, *J* = 10.3, 5.0, 4.7, 1.8 Hz, 1H), 4.48 (d, *J* = 11.6 Hz, 1H), 4.40 (d, *J* = 11.6 Hz, 1H), 4.10 (dd, *J* = 6.5, 2.4 Hz, 1H), 3.80 (s, 3H), 3.46 (dd, 10.4, 5.9 Hz, 1H), 3.40 (dd, *J* = 10.4, 4.9 Hz, 1H), 3.16 (qd, *J* = 6.9, 6.7 Hz, 1H), 2.43-2.33 (m, 1H), 2.27-2.17 (m, 1H), 1.99 (ddd, *J* = 15.0, 4.0, 1.6 Hz, 1H), 1.86 (dd, 5.9, 2.3 Hz, 1H), 1.83 (dd, *J* = 6.0, 2.9 Hz, 1H), 1.67-1.45 (m, 2H), 1.29 (s, 3H), 1.28-1.18 (m, 4H), 1.14 (d, *J* = 6.9 Hz, 3H), 1.12-1.07 (m, 22H), 1.06 (s, 3H), 1.05-1.01 (m, 1H), 0.94 (d, *J* = 6.9 Hz, 3H), 0.89-0.83 (m, 2H), 0.54 (ddd, *J* = 12.3, 10.7, 6.5 Hz, 1H), 0.42 (dddd, *J* = 9.8, 9.5, 6.6, 4.2 Hz, 1H),

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 218.1, 172.6, 159.4, 130.2, 129.4, 113.9, 73.6, 72.8, 71.4, 55.4, 47.6, 43.1, 39.8, 35.7, 34.8, 34.1, 31.7, 31.0, 27.1, 26.5, 25.2, 23.6, 18.6, 18.6, 18.6, 18.0, 16.4, 16.2, 13.5, 11.2.

IR (film): ν □2925, 2865, 1735, 1697, 1613, 1513, 1464, 1366, 1248, 1121, 1093, 1037, 999, 979, 883, 820, 678.

HRMS (ESI): calculated for $C_{38}H_{64}O_6Si$ $[M+H]^+$ 645.4545, found 645.4534.

4.4 Towards the Total Synthesis of Michaolide E



412

(*R*)-3,5-bis((*tert*-butyldimethylsilyl)oxy)pentan-2-one (412). To a solution of alcohols **419** and **420** (2.51 g, 10.8 mmol) and imidazole (1.62 g, 23.78 mmol) in DMF (18 mL) was added TBSCl (1.79 g, 11.9 mmol) at room temperature and the mixture was stirred for 20 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (15 mL). The aqueous phase was extracted with Et_2O (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 30:1) afforded 3.68 g (98%) of silyl ether **412** as a colorless oil.

TLC: R_f 0.33 (hexane/EtOAc 30:1, CPS).

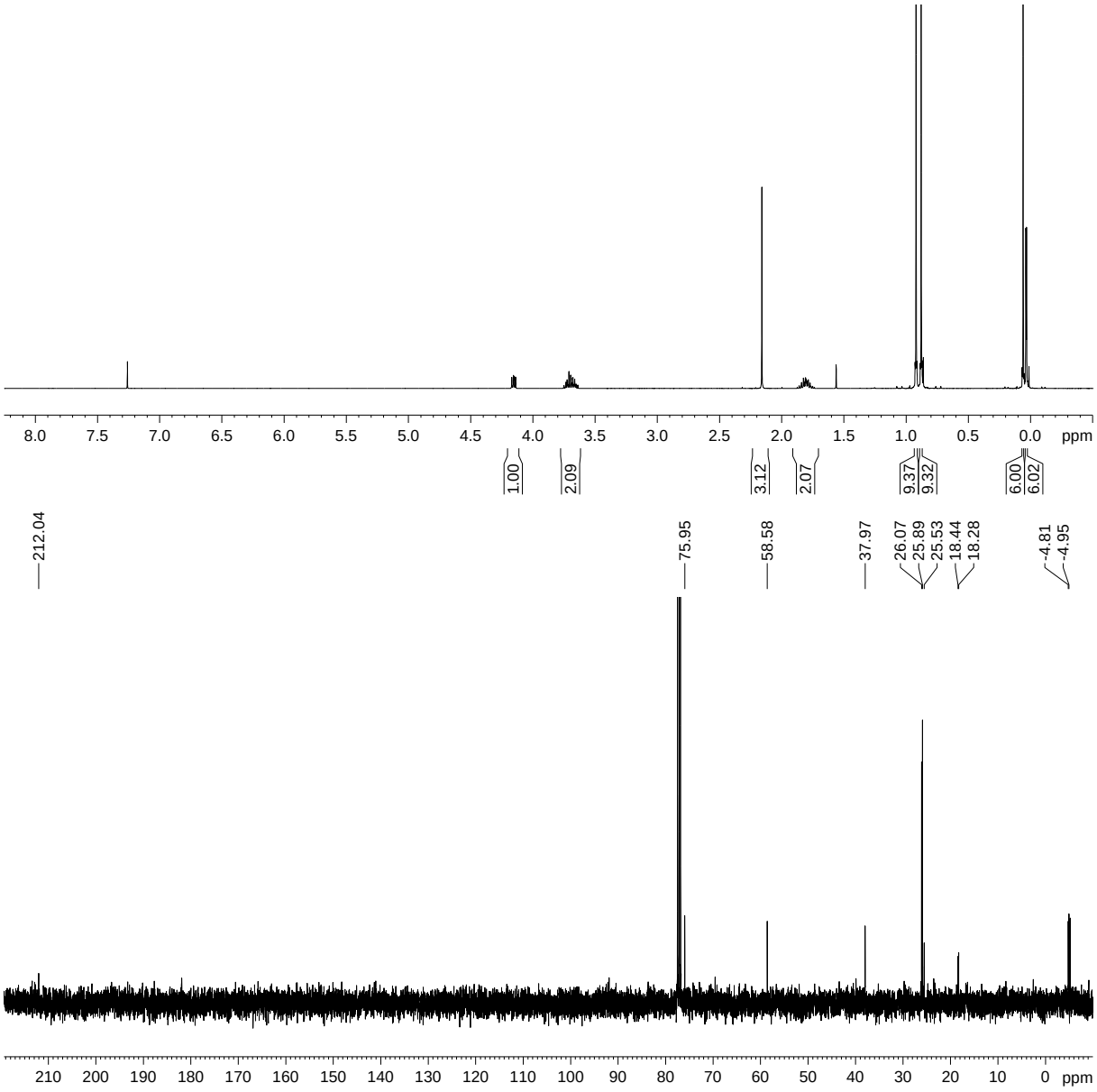
$[\alpha]^{20}_D$: = -12.6° (*c* = 0.64, $CHCl_3$).

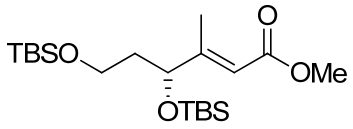
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.15 (dd, *J* = 6.7, 5.3 Hz, 1H), 3.71 (ddd, *J* = 10.2, 6.0, 6.0 Hz), 3.67 (ddd, *J* = 10.2, 6.9, 5.7 Hz, 1H), 2.16 (s, 3H), 1.84 (dddd, *J* = 13.7, 6.9, 6.7, 6.0 Hz, 1H), 1.77 (dddd, *J* = 13.7, 6.0, 5.7, 5.3, 1H), 0.92 (s, 9H), 0.88 (s, 9H), 0.06 (s, 6H), 0.04 (s, 3H), 0.03 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 212.0, 75.9, 58.6, 38.0, 26.1, 25.9, 25.5, 18.4, 18.3, -4.8, --5.0, -5.3, -5.3.

IR (film): ν 2954, 2929, 2886, 1719, 1471, 1463, 1418, 1389, 1361, 1254, 1103, 1044, 1005, 938, 917, 835, 811, 776, 711, 669.

HRMS (ESI): calculated for $C_{17}H_{38}O_3Si_2$ $[M+Na]^+$ 369.2252, found 369.2262.





422

(*R*,*E*)-methyl 4,6-bis((*tert*-butyldimethylsilyl)oxy)-3-methylhex-2-enoate (422). To a solution of NaH (0.33 mg, 8.22 mmol, 60% in mineral oil) in THF (40 mL) was added trimethyl phosphonacetate (1.19 mL, 8.22 mmol) at 0 °C and the reaction mixture was stirred for 1 h at this temperature (the solution did not clear off). The reaction mixture was allowed to reach room temperature and ketone **412** (0.95 g, 2.74 mmol) in THF (5 mL) was added. The reaction mixture heated to 45 °C and stirred for 16.5 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (50 mL). The aqueous

phase was extracted with Et_2O (3 x 50 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 50:1) afforded 0.95 g (91%, *E*/*Z* 12:1) of unsaturated ester **422** as a single isomer as a colorless oil.

TLC: R_f 0.27 (hexane/EtOAc 50:1, UV, CPS).

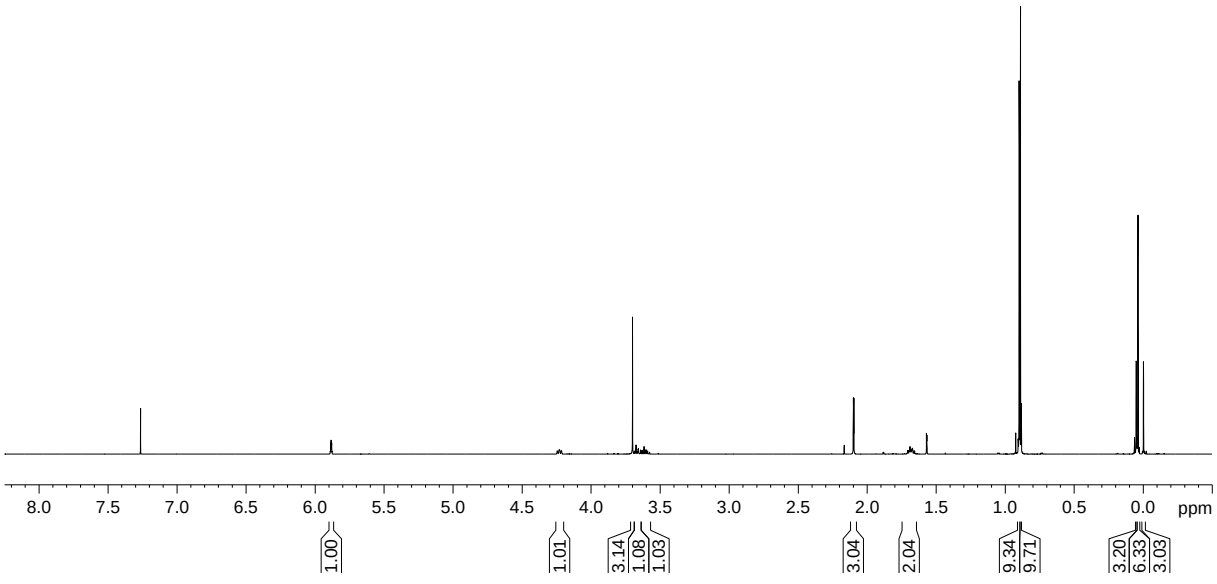
$[\alpha]^{20}_D$: -13.4° (*c* = 1.00, $CHCl_3$).

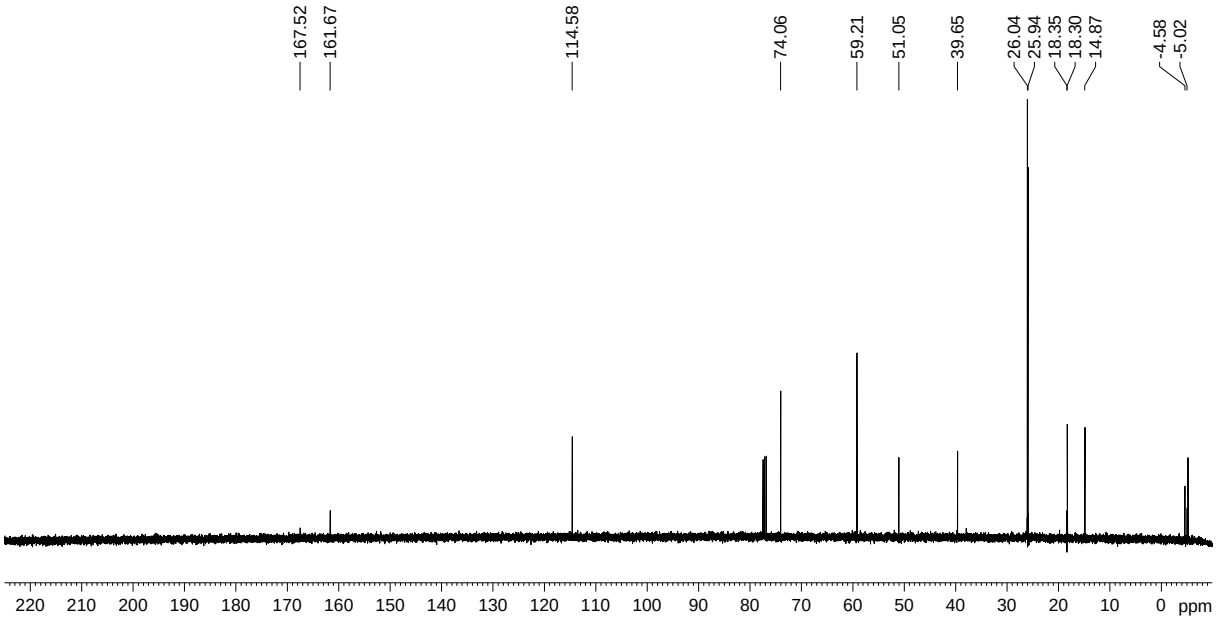
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.88 (p, *J* = 1.3 Hz, 1H), 4.23 (dd, *J* = 6.9, 5.1 Hz, 1H), 3.70 (s, 3H), 3.67 (ddd, *J* = 10.2, 7.2, 6.2 Hz, 1H), 3.60 (ddd, *J* = 10.2, 5.8, 5.6 Hz, 1H), 2.10 (d, *J* = 1.3 Hz, 1H), 2.26 (s, 3H), 1.72-1.64 (m, 2H), 0.90 (s, 9H), 0.89 (s, 9H), 0.05 (s, 3H), 0.04 (s, 6H), 0.00 (s, 3H).

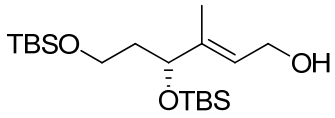
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 167.5, 161.7, 114.6, 74.1, 59.2, 51.1, 39.7, 26.0, 25.9, 18.3, 18.3, 14.9, -4.6, -5.0, -5.2, -5.2.

IR (film): ν 2953, 2929, 2886, 2828, 1723, 1657, 1472, 1463, 1435, 1406, 1389, 1361, 1332, 1255, 1225, 1154, 1092, 1036, 1006, 940, 892, 835, 776.

HRMS (ESI): calculated for $C_{20}H_{42}O_4Si_2$ $[M+Na]^+$ 425.2514, found 425.2505.







423

(*R*,*E*)-4,6-bis((*tert*-butyldimethylsilyl)oxy)-3-methylhex-2-en-1-ol (423). To a solution of ester **422** (1.07 g, 2.67 mmol) in CH_2Cl_2 (25 mL) was added DIBAL-H (1 M in CH_2Cl_2, 5.87 mL, 5.87 mmol) dropwise at -78 °C and the reaction mixture was stirred for 30 min at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (50 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 30:1 → 10:1) to yield 0.79 g (89%) of allylic alcohol **423** as a colorless oil.

TLC: R_f 0.34 (hexane/EtOAc 9:1, CPS).

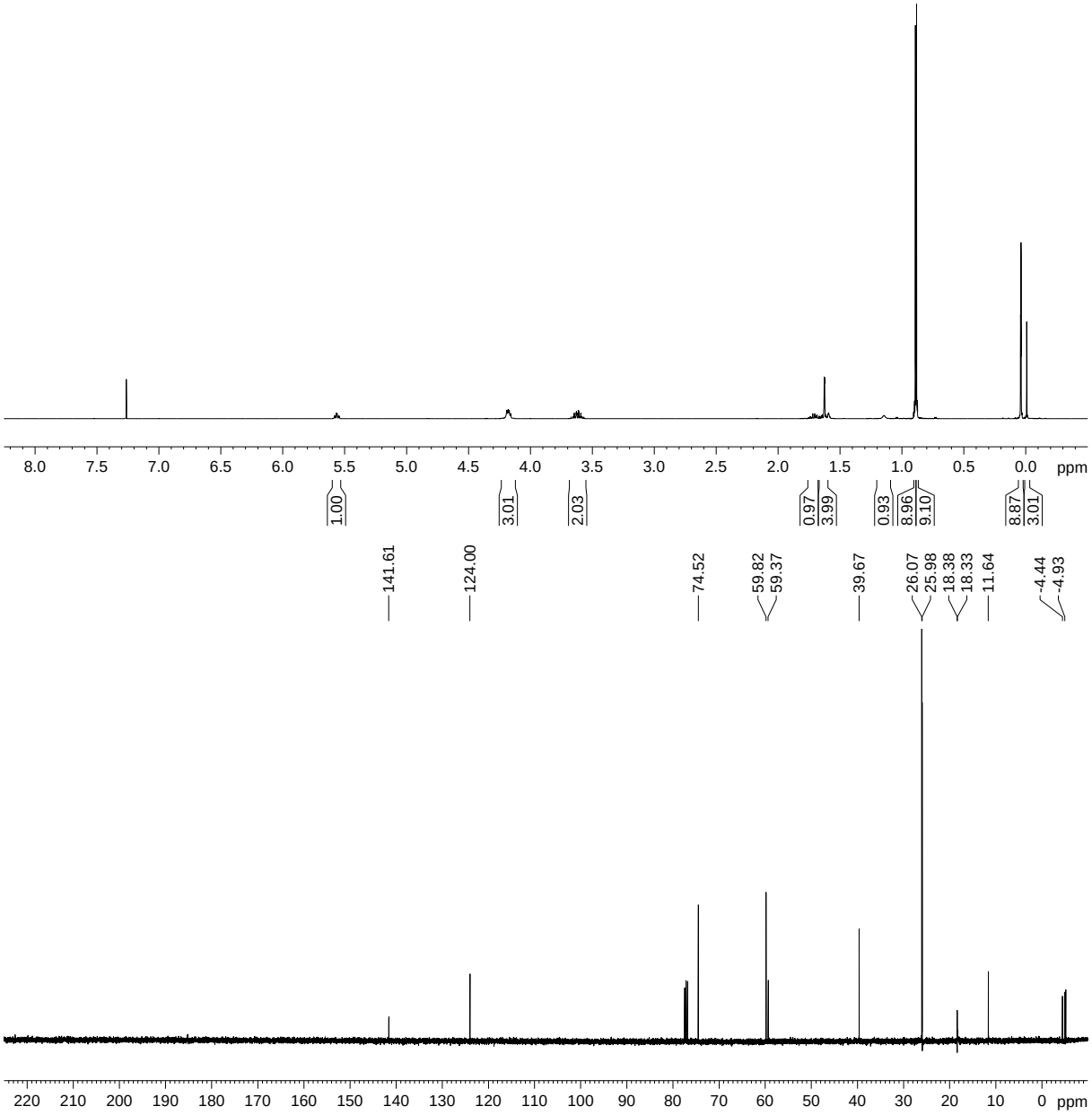
$[\alpha]^{20}_D$: -5.2° (*c* = 1.33, $CHCl_3$).

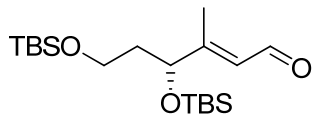
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.56 (tt, *J* = 6.8, 1.2 Hz, 1H), 4.22-4.12 (m, 3H), 3.64 (ddd, *J* = 10.2, 7.4, 6.1 Hz, 1H), 3.60 (ddd, *J* = 10.2, 6.8, 5.6 Hz, 1H), 1.71 (dddd, *J* = 11.7, 8.0, 5.8, 5.6 Hz, 1H), 1.65-1.58 (m, 1H), 1.62 (d, *J* = 1.2 Hz, 3H), 1.15 (br s, 1H), 0.89 (s, 9H), 0.88 (s, 9H), 0.04 (s, 3H), 0.03 (s, 6H), -0.01 (s, 3H)..

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 141.6, 124.0, 74.5, 59.8, 59.4, 39.7, 26.1, 26.0, 18.4, 18.3, 11.6, -4.4, -4.9, -5.1, -5.2.

IR (film): ν 3349, 2954, 2929, 2886, 2857, 1472, 1463, 1388, 1361, 1254, 1085, 1004, 939, 834, 774, 665.

HRMS (ESI): calculated for $C_{19}H_{42}O_3Si_2$ $[M+Na]^+$ 397.2565, found 397.2567.





411

(*R*,*E*)-4,6-bis((*tert*-butyldimethylsilyl)oxy)-3-methylhex-2-enal (411). To a solution of allylic alcohol **423** (0.79 g, 2.10 mmol) in Et_2O (25 mL) was added MnO_2 (2.92 g, 33.60 mmol) in one portion at room temperature and the solution was stirred for 2 h. The reaction

mixture was then filtered through a small plug of celite and the precipitate was rinsed with Et_2O (50 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 25:1) to yield 0.71 g of unsaturated aldehyde **411** (91%) as a colorless oil.

TLC: R_f 0.22 (hexane/EtOAc 30:1, UV, CPS).

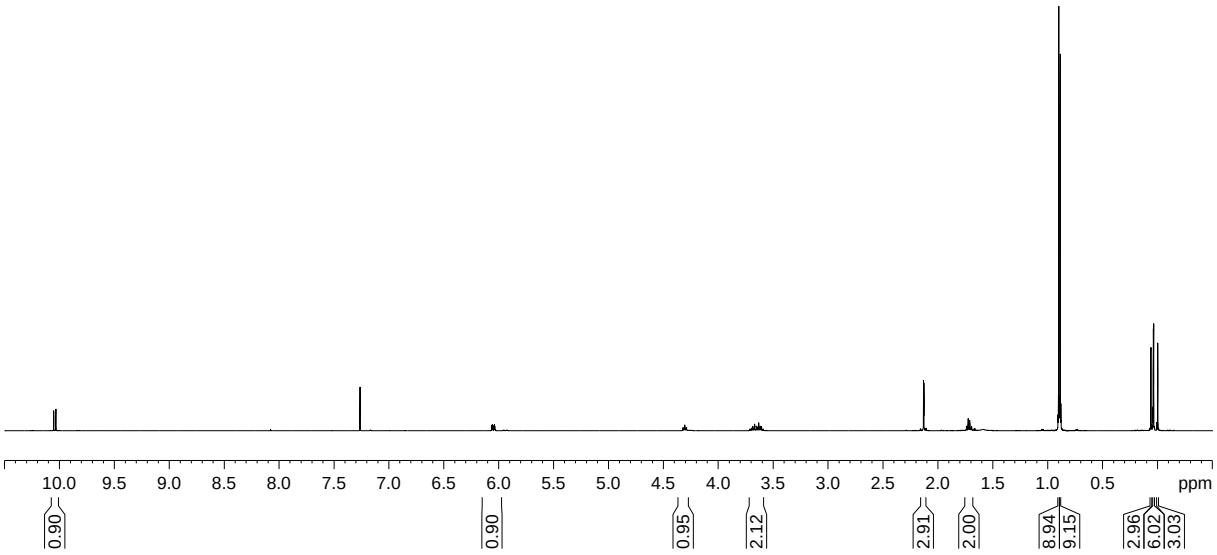
$[\alpha]^{20}_D$: = -8.1° (*c* = 0.91, $CHCl_3$).

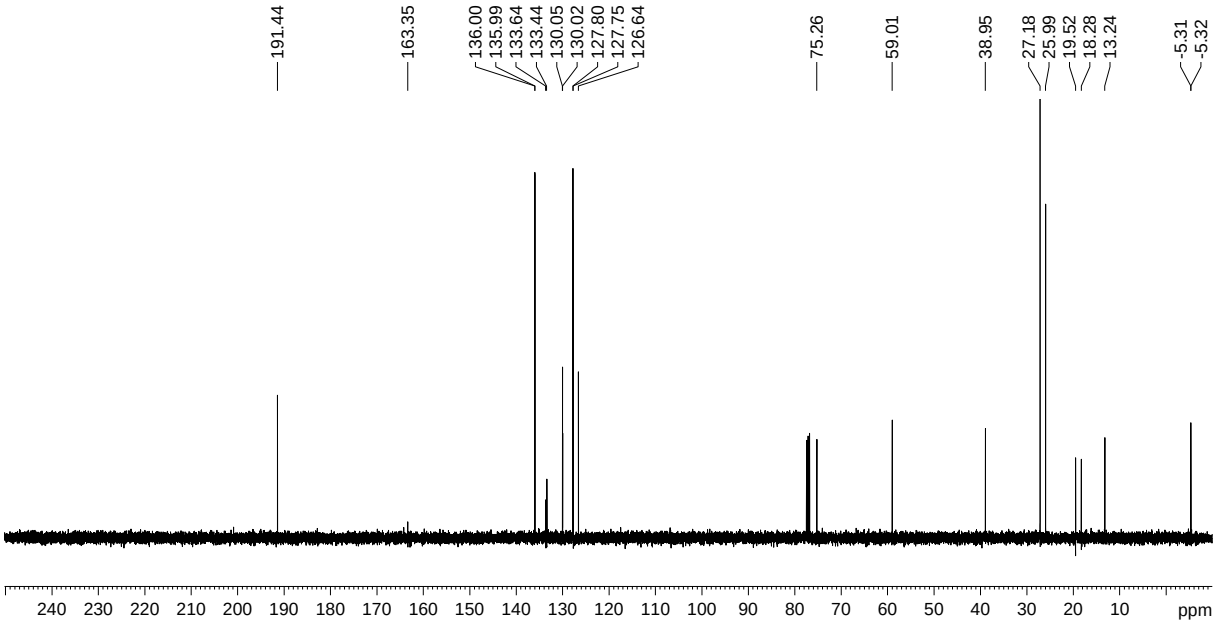
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 10.04 (d, *J* = 8.0 Hz, 1H), 6.05 (dp, *J* = 8.0, 1.2 Hz, 1H), 4.31 (dd, *J* = 6.9, 5.1 Hz, 1H), 3.68 (ddd, *J* = 10.2, 7.2, 6.2, 1H), 3.62 (ddd, *J* = 10.2, 5.8, 5.4 Hz, 1H), 2.13 (d, *J* = 1.2 Hz, 3H), 1.75-1.68 (m, 2H), 0.90 (s, 9H), 0.89 (s, 9H), 0.06 (s, 3H), 0.04 (s, 3H), 0.03 (s, 3H), 0.00 (s, 3H).

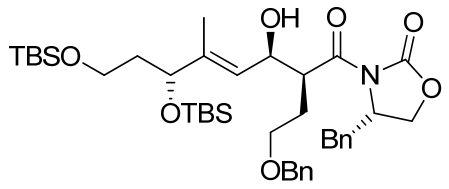
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 191.7, 165.2, 125.8, 73.7, 59.0, 39.6, 26.0, 25.9, 18.3, 18.3, 13.4, -4.6, -5.0, -5.2, -5.2.

IR (film): ν 2954, 2929, 2886, 2858, 1693, 1649, 1472, 1463, 1419, 1389, 1362, 1254, 1186, 1163, 1099, 1006, 939, 893, 835, 776, 711, 669.

HRMS (ESI): calculated for $C_{19}H_{40}O_3Si_2$ $[M+Na]^+$ 397.2565, not found.







426

(*S*)-4-benzyl-3-((2*S*,3*R*,6*R*,*E*)-2-(2-(benzyloxy)ethyl)-6,8-bis((*tert*-butyldimethylsilyl)oxy)-3-hydroxy-5-methyloct-4-enoyl)oxazolidin-2-one (426). To a solution of Evans-auxiliary derivate **425** (0.53 g, 1.50 mmol) in CH_2Cl_2 (10 mL) was added *n*-Bu_2OTf (1.65 mL, 1M in CH_2Cl_2, 1.65 mmol) and DIPEA (0.31 mL, 1.80 mmol) dropwise at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 0.5 h. The reaction mixture was then cooled to -78 °C. Aldehyde **411** (0.73 g, 1.95 mmol) in CH_2Cl_2 (3 mL) was added over a period of 15 min and the reaction mixture was stirred for 1.5 h at -78 °C. The reaction was then cautiously quenched by addition of pH 7 buffer (4 mL), MeOH (20 mL) and 30% aqueous H_2O_2 (4 mL). The resulting mixture was stirred at 0 °C for 1 h, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 8:1) to yield 0.78 g (73%) of aldol product **426** as a single isomer and colourless oil.

Note: The excess of the aldehyde can be reisolated.

TLC: R_f 0.45 (hexane/EtOAc 4:1, UV, CPS).

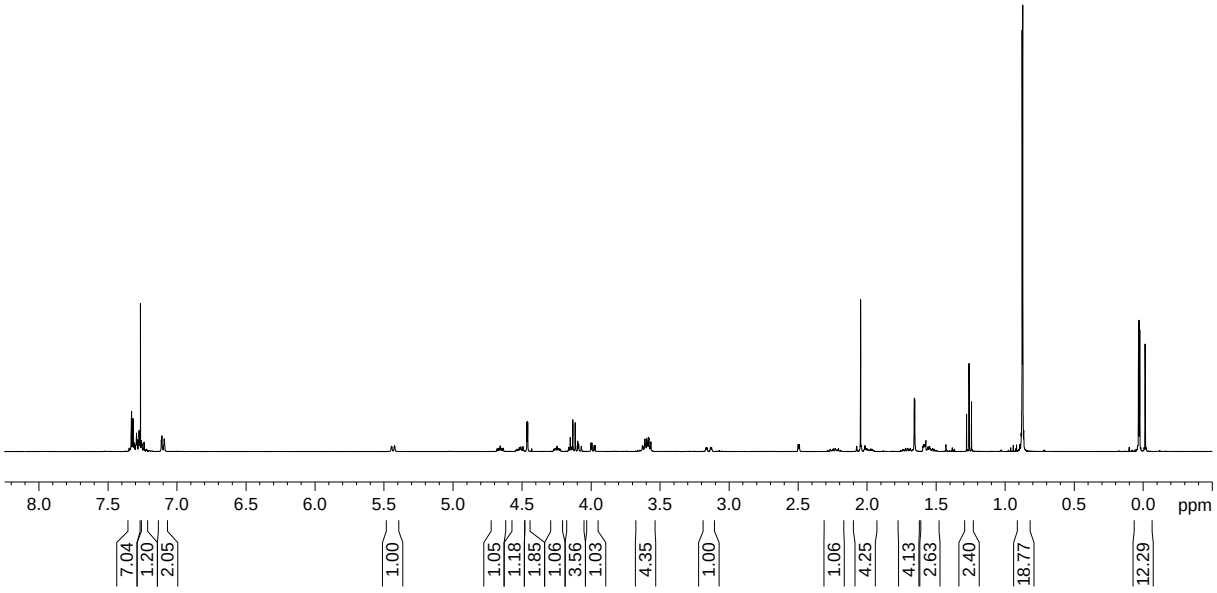
$[\alpha]^{20}_D$: = 9.9 (*c* = 0.73, $CHCl_3$).

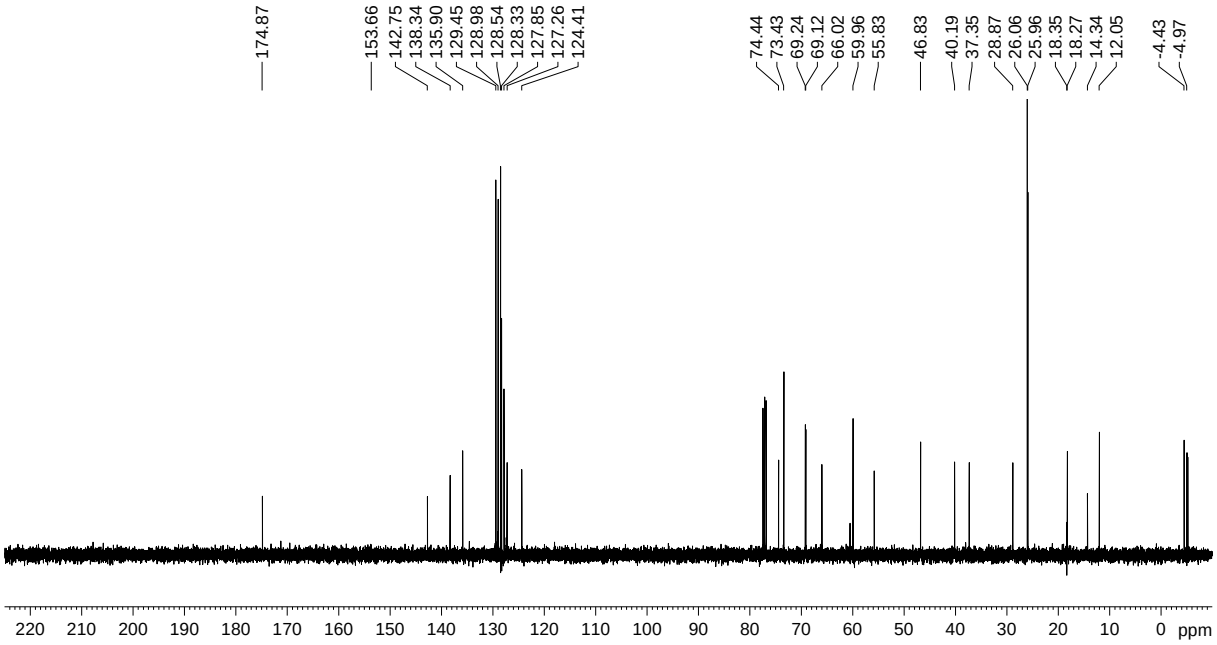
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.34-7.22 (m, 8H), 7.12-7.08 (m, 2H), 5.44 (dp, *J* = 8.7, 1.2 Hz, 1H), 4.66 (ddd, *J* = 8.9, 5.9, 3.3 Hz, 1H), 4.52 (dddd, *J* = 10.6, 7.8, 2.9, 2.9 Hz, 1H), 4.46 (d, *J* = 2.0 Hz, 2H), 4.25 (ddd, *J* = 9.4, 5.9, 3.3 Hz, 1H), 4.15 (dd, *J* = 8.3, 4.1 Hz, 1H), 4.09 (dd, *J* = 8.7, 8.6 Hz, 1H), 3.99 (dd, *J* = 9.1, 2.8 Hz, 1H), 3.63-3.56 (m, 4H), 3.15 (dd, *J* = 13.3, 3.0 Hz, 1H), 2.48 (d, *J* = 3.2 Hz, 1H), 2.24 (dddd, *J* = 14.6, 9.1, 6.5, 6.5 Hz, 1H), 2.05 (dd, *J* = 13.6, 10.6 Hz, 1H), 1.98 (dddd, *J* = 14.6, 5.2, 5.1, 2.8 Hz, 1H), 1.72 (dddd, *J* = 13.4, 8.3, 6.3, 5.7 Hz, 1H), 1.66 (d, *J* = 1.2 Hz, 3H), 1.54 (ddd, *J* = 13.4, 7.3, 4.1 Hz, 1H), 0.88 (s, 9H), 0.87 (s, 9H), 0.04 (s, 3H), 0.03 (s, 3H), 0.03 (s, 3H), -0.01 (s, 3H).

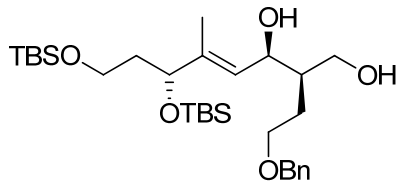
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 174.9, 153.7, 142.8, 138.3, 135.9, 129.5, 129.0, 128.5, 128.3, 127.9, 127.3, 124.4, 124.4, 74.4, 73.4, 69.2, 69.1, 66.0, 60.0, 55.8, 46.8, 40.2, 37.3, 28.9, 26.1, 26.0, 18.4, 18.3, 12.1, -4.4, -5.0, -5.1, -5.1.

IR (film): ν 3509, 3030, 2954, 2929, 2857, 1782, 1696, 1472, 1360, 1252, 1206, 1096, 1006, 939, 836, 776, 741, 700, 667.

HRMS (ESI): calculated for $C_{40}H_{63}NO_7Si_2$ $[M+Na]^+$ 748.4035, found 748.4040.







438

(2*R*,3*R*,6*R*,*E*)-2-(2-(benzyloxy)ethyl)-6,8-bis((*tert*-butyldimethylsilyl)oxy)-5-methyloct-4-ene-1,3-diol (438). To a solution of imide **426** (2.45 g, 3.38 mmol) in THF (50 mL) was added MeOH (0.19 mL, 4.73 mmol) and $LiBH_4$ (103 mg, 4.73 mmol) in one portion at 0 °C. The solution was stirred for 3 h at 0 °C, a second portion of MeOH (0.12, 2.68 mmol) and $LiBH_4$ (62 mg, 2.83 mmol) was then added and stirring was continued for further 3 h at 0 °C. The reaction was then cautiously quenched by addition of 15% NaOH aqueous solution (20 mL) and the aqueous phase was extracted with EtOAc (3 x 20 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to afford 1.37 g (75%) of diol **438** as a colorless oil.

TLC: R_f 0.31 (hexane/EtOAc 3:1, UV, CPS).

$[\alpha]^{20}_D$: = 1.2 (*c* = 0.77, $CHCl_3$).

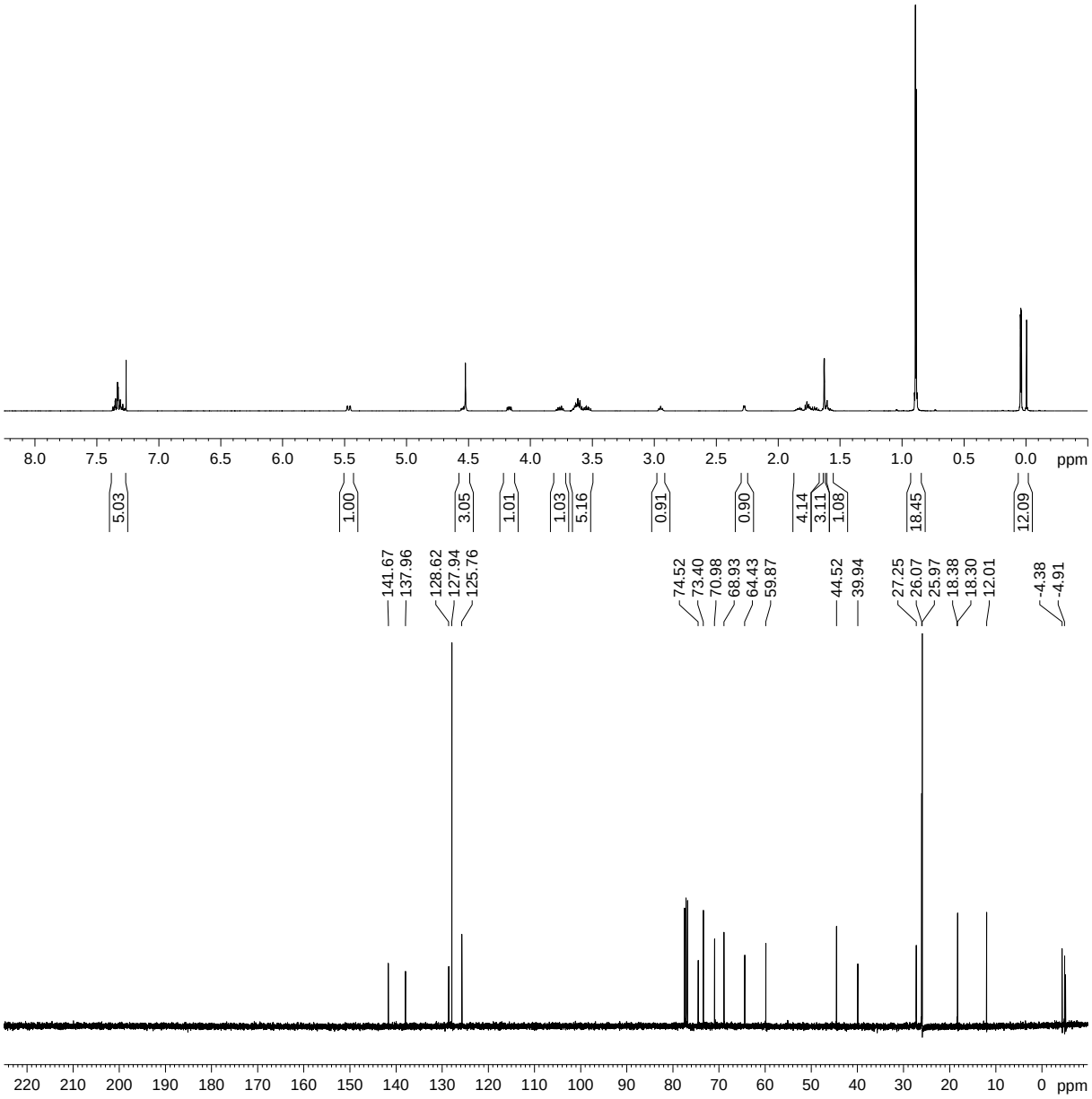
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.26 (m, 5H), 5.47 (dp, *J* = 8.9, 1.2 Hz, 1H), 4.55 (dd, *J* = 4.7, 3.6 Hz, 1H), 4.52 (s, 2H), 4.17 (dd, *J* = 7.8, 4.8 Hz, 1H), 3.76 (ddd, *J* = 11.2, 5.4, 5.3 Hz, 1H), 3.66-3.57 (m, 4H), 3.55 (dddd, *J* = 9.7, 9.3, 6.5, 5.0 Hz, 1H), 2.95 (t, *J* = 5.8 Hz, 1H), 2.27 (d, *J* = 3.6 Hz, 1H), 1.87-1.79 (m, 1H), 1.79-1.73 (m, 2H), 1.70 (ddd, *J* = 7.7, 6.0,

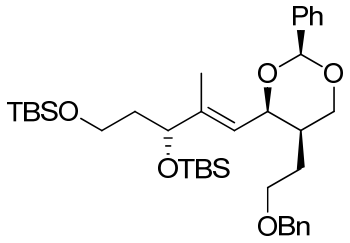
5.9 Hz, 1H), 1.63 (d, *J* = 1.2 Hz, 3H), 1.59 (ddd, *J* = 13.7, 7.0, 4.9 Hz, 1H), 0.89 (s, 9H), 0.88 (s, 9H), 0.04 (s, 3H), 0.04 (s, 3H), 0.03 (s, 3H), -0.01 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 141.7, 138.0, 128.6, 128.6, 127.9, 125.8, 74.5, 73.4, 71.0, 68.9, 64.4, 59.9, 44.5, 39.9, 27.3, 26.1, 26.0, 18.4, 18.3, 12.0, -4.4, -4.9, 5.1, -5.1.

IR (film): ν 3389, 2953, 2928, 2884, 2857, 1472, 1388, 1361, 1254, 1087, 1005, 940, 896, 835, 775, 745, 732, 697.

HRMS (ESI): calculated for $C_{30}H_{56}O_5Si_2$ $[M+Na]^+$ 575.3558, found 575.3548.





458

(*R*)-5-((*E*)-1-((2*S*,4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-phenyl-1,3-dioxan-4-yl)prop-1-en-2-yl)-2,2,3,3,9,9,10,10-octamethyl-4,8-dioxa-3,9-disilaundecane (458). To a solution of diol **457** (1.08 g, 1.95 mmol) in CH_2Cl_2 (20 mL) was added benzaldehyde dimethyl acetale (0.44 mL, 2.93 mmol) and CSA (36 mg, 0.16 mmol) at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 1.5 h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (20 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 20 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 20:1) gave 0.88 g (70%) of acetal **458** as a colorless oil.

TLC: R_f 0.31 (hexane/EtOAc 20:1, UV, CPS).

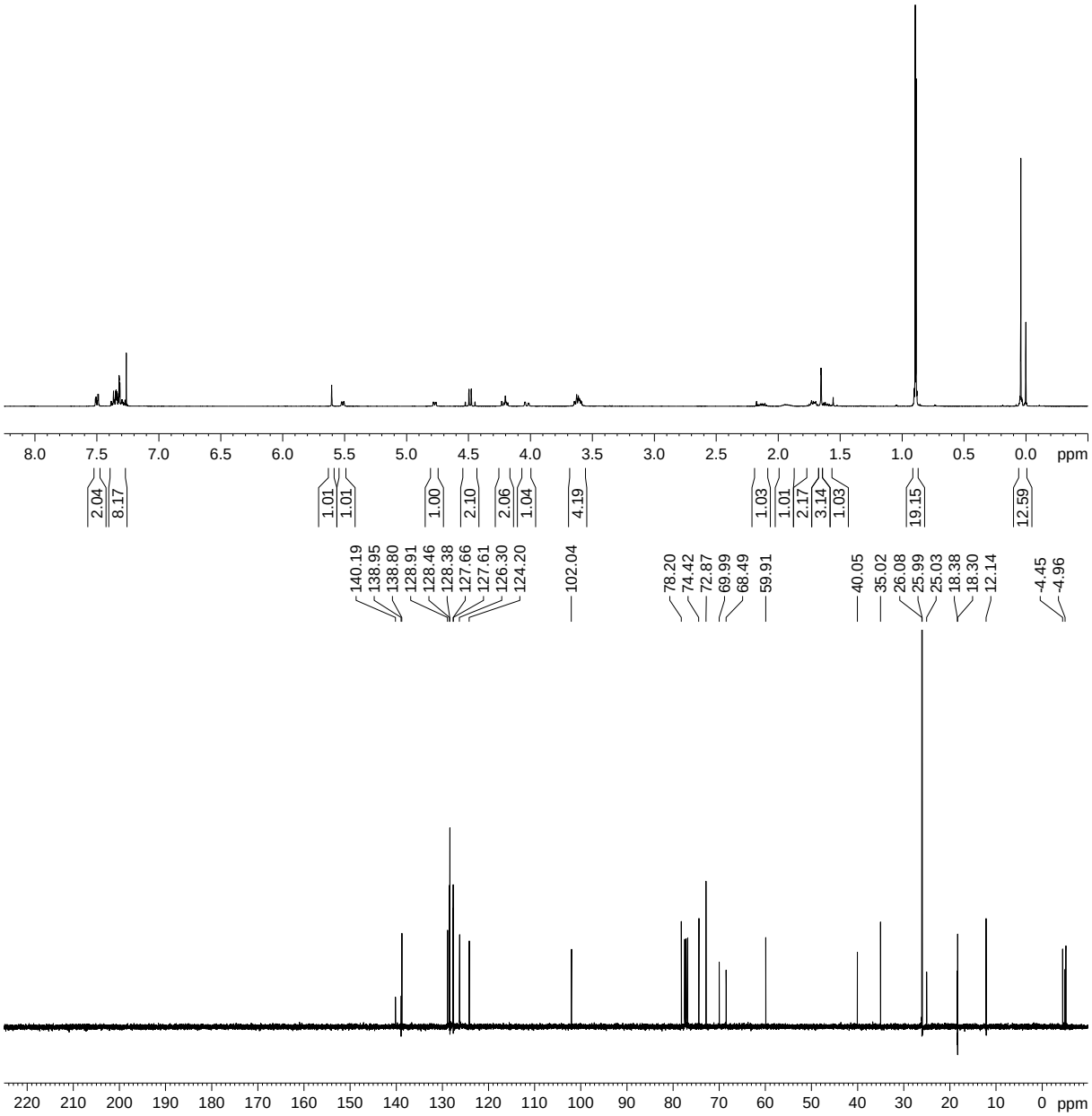
$[\alpha]^{20}_D$: = 4.0 (*c* = 0.88, $CHCl_3$).

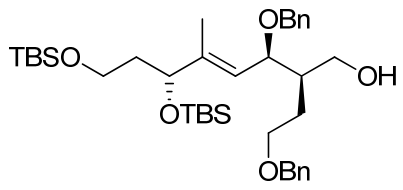
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.52-7.48 (m, 2H), 7.39-7.27 (m, 8H), 5.60 (s, 1H), 5.51 (dp, *J* = 6.9, 1.2 Hz, 1H), 4.77 (dd, *J* = 6.9, 2.5 Hz, 1H), 4.51 (d, *J* = 12.0 Hz, 1H), 4.46 (d, *J* = 12.0 Hz, 1H), 4.22 (dd, *J* = 11.6, 1.2 Hz, 1H), 4.20 (dd, *J* = 8.0, 4.7 Hz, 1H), 4.03 (ddd, *J* = 11.6, 2.3, 1.0 Hz, 1H), 3.68-3.56 (m, 4H), 2.14 (dddd, *J* = 15.3, 10.2, 5.1, 5.0 Hz, 1H), 1.93 (dddd, *J* = 14.6, 9.3, 7.0, 3.5 Hz, 1H), 1.77-1.67 (m, 2H), 1.65 (d, *J* = 1.2 Hz, 3H), 1.62 (dddd, *J* = 13.6, 12.0, 7.1, 4.8 Hz, 1H), 0.90 (s, 9H), 0.89 (s, 9H), 0.04 (s, 9H), 0.00 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 140.2, 138.9, 138.8, 128.9, 128.5, 128.4, 127.7, 127.6, 126.3, 124.2, 102.0, 78.2, 74.4, 72.9, 70.0, 68.5, 59.9, 40.0, 35.0, 26.1, 26.0, 25.0, 18.4, 18.3, 12.1, -4.5, -5.0, -5.1, -5.2.

IR (film): ν 2953, 2928, 2856, 1471, 1462, 1362, 1306, 1253, 1213, 1146, 1087, 1051, 1028, 1006, 982, 939, 899, 835, 776, 748, 734, 697, 662.

HRMS (ESI): calculated for $C_{37}H_{60}O_5Si_2$ $[M+Na]^+$ 663.3871, found 663.3876.





453

(2*R*,3*R*,6*R*,*E*)-3-(benzyloxy)-2-(2-(benzyloxy)ethyl)-6,8-bis((*tert*-butyldimethylsilyl)oxy)-5-methyloct-4-en-1-ol (453). To a solution of acetale **458** (50 mg, 0.08 mmol) in CH_2Cl_2 (1.5 mL) was added DIBAL-H (1.2 M in toluene, 0.13 mL, 0.16 mmol) dropwise at -78 °C and the reaction mixture was allowed to reach -25 °C. Me_2AlCl (1 M in hexane, 0.10 mL, 0.10 mmol) was added dropwise and the reaction mixture was allowed to reach 0 °C over a period

of 1 h. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (5 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to yield 38 mg (76%) of alcohol **453** as a colorless oil.

TLC: R_f 0.26 (hexane/EtOAc 20:1, UV, CPS).

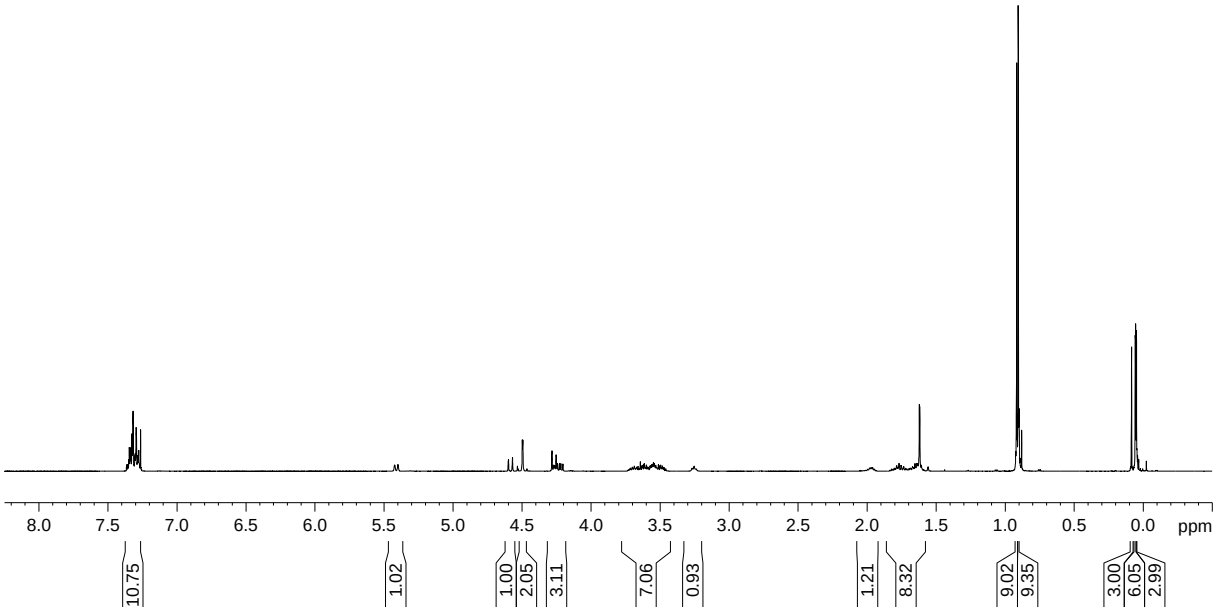
$[\alpha]^{20}_D$: = -18.1 (*c* = 2.46, $CHCl_3$).

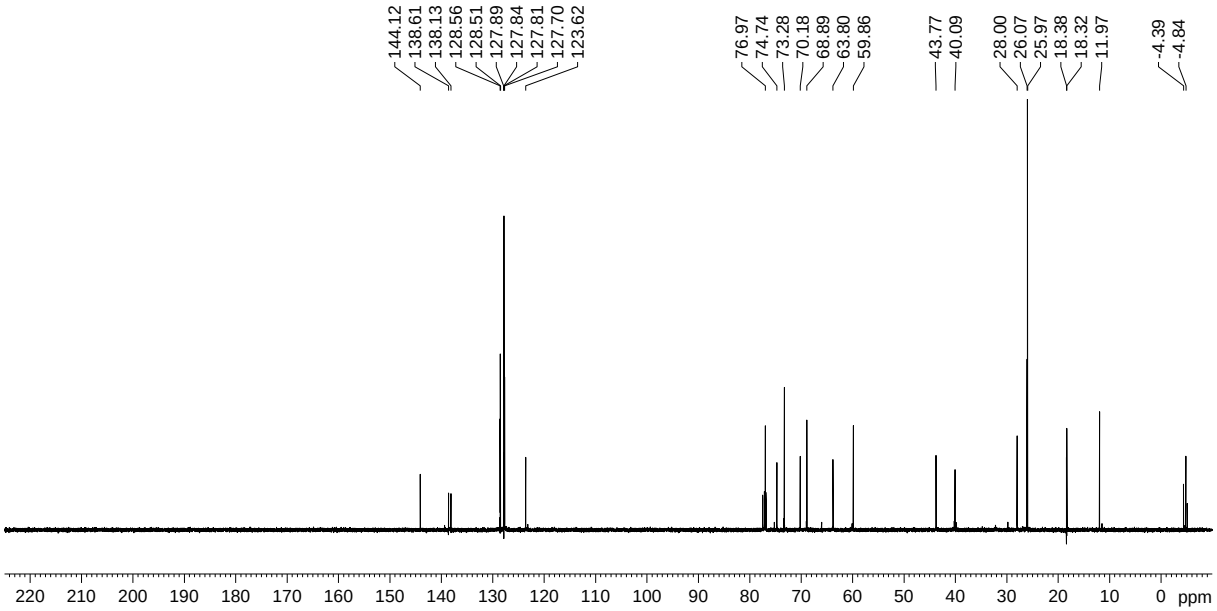
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.36-7.25 (m, 10H), 7.39-7.27 (m, 8H), 5.41 (dp, *J* = 9.5, 1.2 Hz, 1H), 4.58 (d, *J* = 11.8 Hz, 1H), 4.49 (d, *J* = 1.5 Hz, 1H), 4.26 (d, *J* = 11.8 Hz, 1H), 4.25 (dd, *J* = 8.0, 2.8 Hz, 1H), 4.22 (dd, *J* = 9.5, 6.0 Hz, 1H), 4.70 (dd, *J* = 11.2, 5.6 Hz, 1H), 3.68-3.44 (m, 6H), 3.25 (dd, *J* = 6.2, 5.8 Hz, 1H), 2.03-1.92 (m, 1H), 1.81 (ddd, *J* = 7.2, 5.0, 4.6 Hz, 1H), 1.78-1.62 (m, 3H), 1.62 (d, *J* = 1.2 Hz, 3H), 1.61-1.57 (m, 1H), 0.91 (s, 9H), 0.90 (s, 9H), 0.08 (s, 3H), 0.05 (s, 3H), 0.05 (s, 3H), 0.04 (s, 3H).

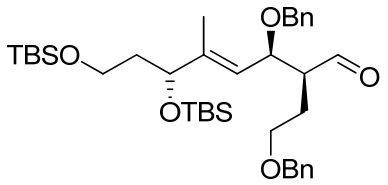
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 144.1, 138.6, 138.1, 128.6, 128.5, 127.9, 127.8, 127.8, 127.7, 123.6, 77.0, 74.7, 73.3, 70.2, 68.9, 63.8, 59.9, 43.8, 40.1, 28.0, 26.1, 26.0, 18.4, 18.3, 12.0, -4.4, -4.8, -5.1, -5.1.

IR (film): ν 3436, 2953, 2928, 2884, 2856, 1496, 1471, 1388, 1361, 1252, 1206, 1088, 1005, 940, 897, 834, 812, 775, 733, 697, 665.

HRMS (ESI): calculated for $C_{37}H_{62}O_5Si_2$ $[M+Na]^+$ 665.4028, found 665.40280.







448

(2*S*,3*R*,6*R*,*E*)-3-(benzyloxy)-2-(2-(benzyloxy)ethyl)-6,8-bis((*tert*-butyldimethylsilyl)oxy)-5-methyloct-4-enal (448). To a solution of alcohol **453** (16 mg, 0.02 mmol) in CH_2Cl_2 (0.3 mL) was added BAIB (11 mg, 0.03 mmol) and TEMPO (1.0 mg, 0.01 mmol) at 0 °C and the reaction mixture was stirred for 4 h at this temperature. A second portion of BAIB (11 mg, 0.03 mmol) and TEMPO (1.0 mg, 0.01 mmol) was then added and stirring was continued for further 5 h at 0 °C. The reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 25:1) to yield 13.5 mg (85%) of aldehyde **448** as a colorless oil.

TLC: R_f 0.15 (hexane/EtOAc 25:1, UV, CPS).

$[\alpha]^{20}_D$: = -18.1 (*c* = 2.46, $CHCl_3$).

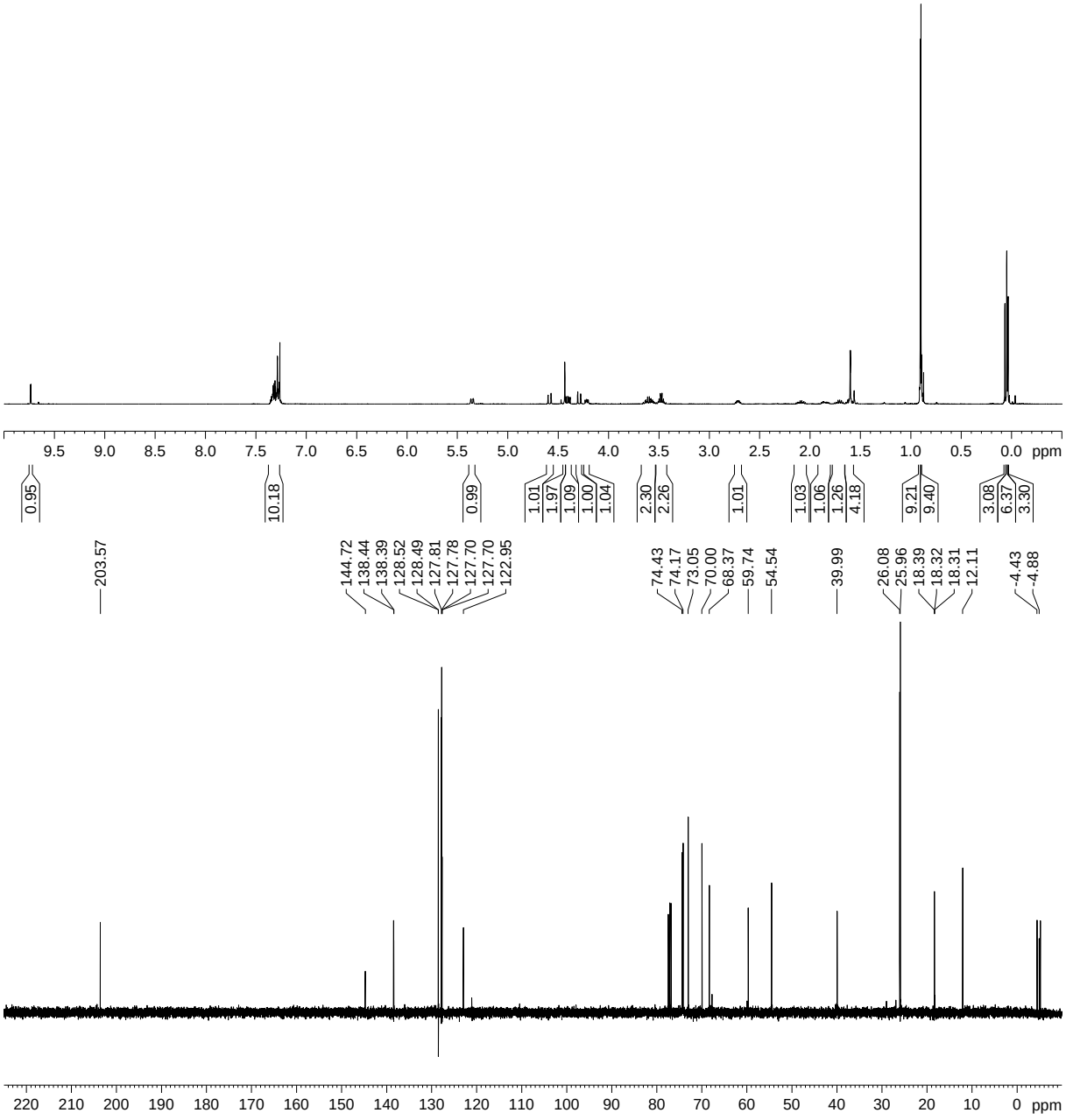
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.73 (d, *J* = 2.3 Hz, 1H), 7.37-7.25 (m, 10H), 5.36 (dp, *J* = 9.7, 1.2 Hz, 1H), 4.59 (d, *J* = 11.8 Hz, 1H), 4.49 (d, *J* = 1.5 Hz, 1H), 4.44 (s, 2H), 4.40 (dd, *J* = 9.6, 6.4 Hz, 1H), 4.29 (d, *J* = 11.8 Hz, 1H), 4.22 (dd, *J* = 7.7, 4.9 Hz, 1H), 3.63 (ddd, *J* = 10.2, 7.0, 6.8 Hz, 1H), 3.58 (ddd, *J* = 10.2, 7.0, 5.5 Hz, 1H), 3.49 (ddd, *J* = 9.2, 6.8, 5.7 Hz, 1H), 3.46 (ddd, *J* = 9.2, 6.4, 6.2 Hz, 1H), 2.72 (dddd, *J* = 8.9, 6.3, 4.0, 2.3 Hz, 1H), 2.09 (dddd, *J* = 14.5, 8.9, 6.7, 5.7 Hz, 1H), 1.85 (dddd, *J* = 14.6, 6.0, 6.0, 4.0 Hz, 1H), 1.71

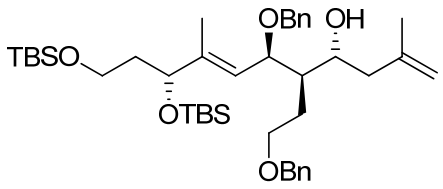
(dddd, J = 13.5, 7.8, 6.0, 6.0 Hz, 1H), 1.61 (ddd, J = 13.5, 7.0, 4.9 Hz, 1H), 1.60 (d, J = 1.2 Hz, 3H), 0.91 (s, 9H), 0.90 (s, 9H), 0.07 (s, 3H), 0.05 (s, 6H), 0.04 (s, 3H)

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 203.6, 144.7, 138.4, 138.4, 128.5, 128.5, 127.8, 127.8, 127.7, 127.7, 123.0, 74.4, 74.2, 73.1, 70.0, 68.4, 59.7, 54.5, 40.0, 26.1, 26.0, 18.4, 18.3, 18.3, 12.1, -4.4, -4.9, -5.1, -5.2.

IR (film): ν 2953, 2928, 2857, 1724, 1471, 1388, 1361, 1254, 1090, 1028, 1005, 939, 896, 836, 776, 736, 698.

HRMS (ESI): calculated for $C_{37}H_{60}O_5Si_2$ $[M+Na]^+$ 663.3871, found 663.3880.





463

(4*R*,5*R*,6*R*,9*R*,*E*)-6-(benzyloxy)-5-(2-(benzyloxy)ethyl)-9,11-bis((*tert*-butyldimethylsilyl)oxy)-2,8-dimethylundeca-1,7-dien-4-ol (463). To a solution of aldehyde **448** (91 mg, 0.14 mmol) in CH_2Cl_2 (3 mL) was added $SnCl_4$ (16.6 µg, 0.14 mmol) and after 2 min methallyl silyl (24.3 µg, 0.14 mmol) at -90 °C and the reaction mixture was stirred for 15 min at this temperature. The reaction was then cautiously quenched by addition of water (3 mL). The solution was allowed to warm to room temperature and, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 5 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 12:1) to yield 78 mg (76%) of alcohol **463** as a single isomer as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 10:1, UV, CPS).

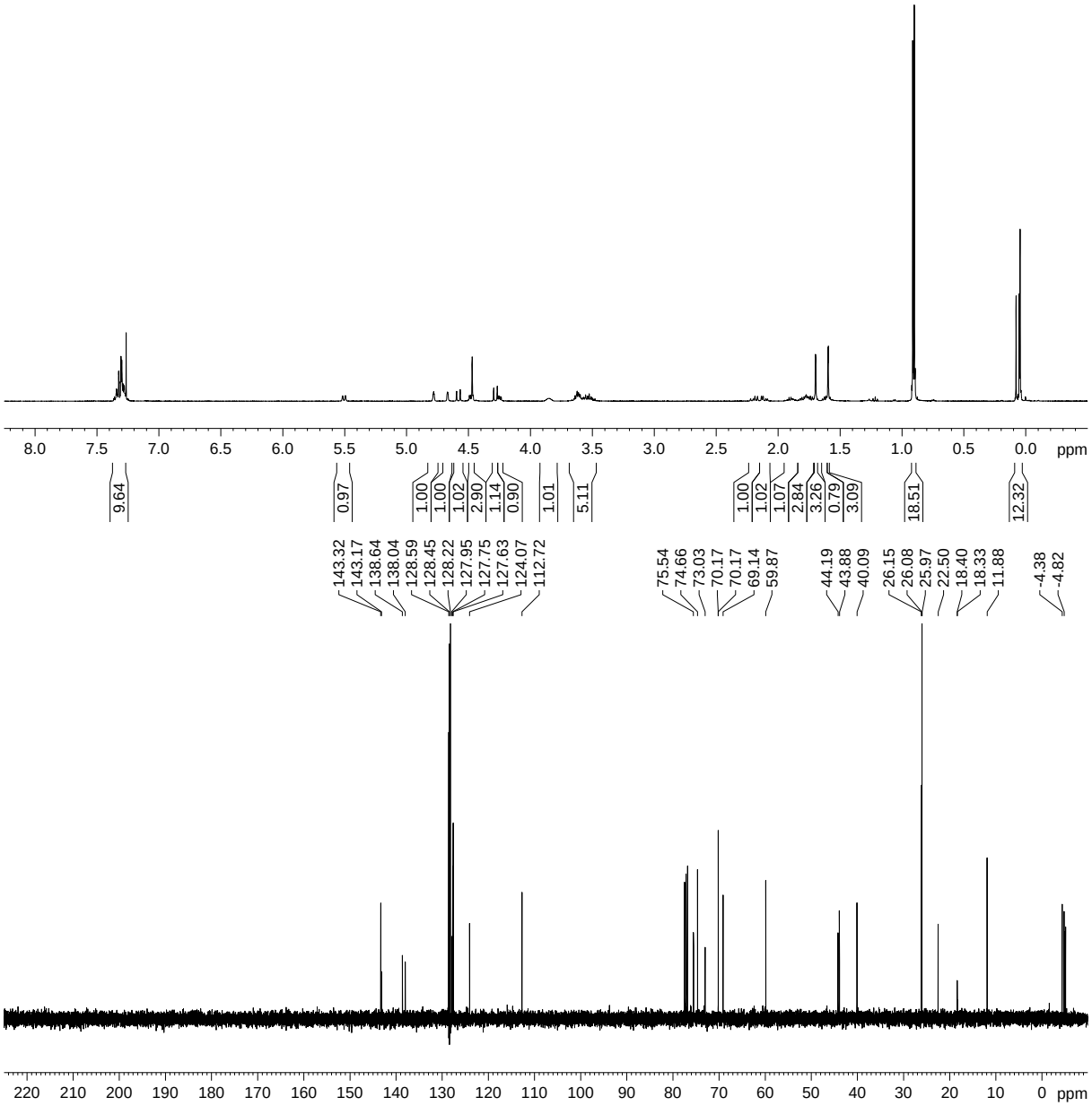
$[\alpha]^{20}_D$: = (*c* = 2.46, $CHCl_3$).

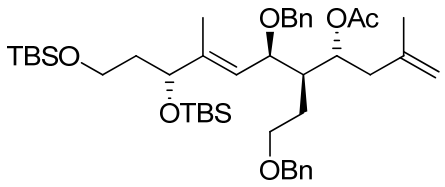
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.25 (m, 10H), 5.50 (dp, *J* = 9.1, 1.2 Hz, 1H), 4.78 (m, 1H), 4.67 (m, 1H), 4.58 (d, *J* = 11.6 Hz, 1H), 4.47 (dd, *J* = 8.8, 3.4 Hz, 1H), 4.47 (s, 2H), 4.28 (d, *J* = 11.6 Hz, 1H), 4.25 (dd, *J* = 7.8, 4.8 Hz, 1H), 3.85 (m, 1H), 3.67-3.47 (m, 5H), 2.19 (dd, *J* = 13.8, 8.4 Hz, 1H), 2.11 (dd, *J* = 13.8, 5.2 Hz, 1H), 1.95-1.85 (m, 1H), 1.83-1.72 (m, 3H), 1.70 (s, 3H), 1.63 (dd, *J* = 7.1, 4.9 Hz, 1H), 1.59 (d, *J* = 1.2 Hz, 3H), 0.91 (s, 9H), 0.90 (s, 9H), 0.08 (s, 3H), 0.05 (s, 3H), 0.04 (s, 6H) .

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 143.3

143.2, 138.6, 138.0, 128.6, 128.5, 128.2, 127.9, 127.7, 127.6, 124.1, 112.7, 75.5, 74.7, 73.0, 70.2, 70.2, 69.1, 59.9, 44.2, 43.9, 40.1, 26.1, 26.1, 26.0, 22.5, 18.4, 18.3, 11.9, -4.4, -4.8, -5.1, -5.1.

HRMS (ESI): calculated for $C_{37}H_{60}O_5Si_2$ $[M+Na]^+$ 663.3871, found 663.3880.





466

(4*R*,5*R*,6*R*,9*R*,*E*)-6-(benzyloxy)-5-(2-(benzyloxy)ethyl)-9,11-bis((*tert*-butyldimethylsilyl)oxy)-2,8-dimethylundeca-1,7-dien-4-yl acetate (466). To a solution of alcohol **463** (310 mg, 0.45 mmol), NEt_3 (0.19 mL, 1.34 mmol) and DMAP (5.4 mg, 0.04 mmol) in MeCN (10 mL) was added Ac_2O (0.13 mL, 1.34 mmol) dropwise at 0 °C and the reaction mixture was allowed to reach room temperature and stirred for 7 h at this

temperature. The reaction was then quenched by addition of saturated aqueous NH_4Cl (10 mL), the phases were separated and the aqueous phase was extracted with ether (3 x 10 mL). The combined organic phases were washed with brine (30 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 25:1) afforded 300 mg (91%) of protected alcohol **466** as a colorless oil.

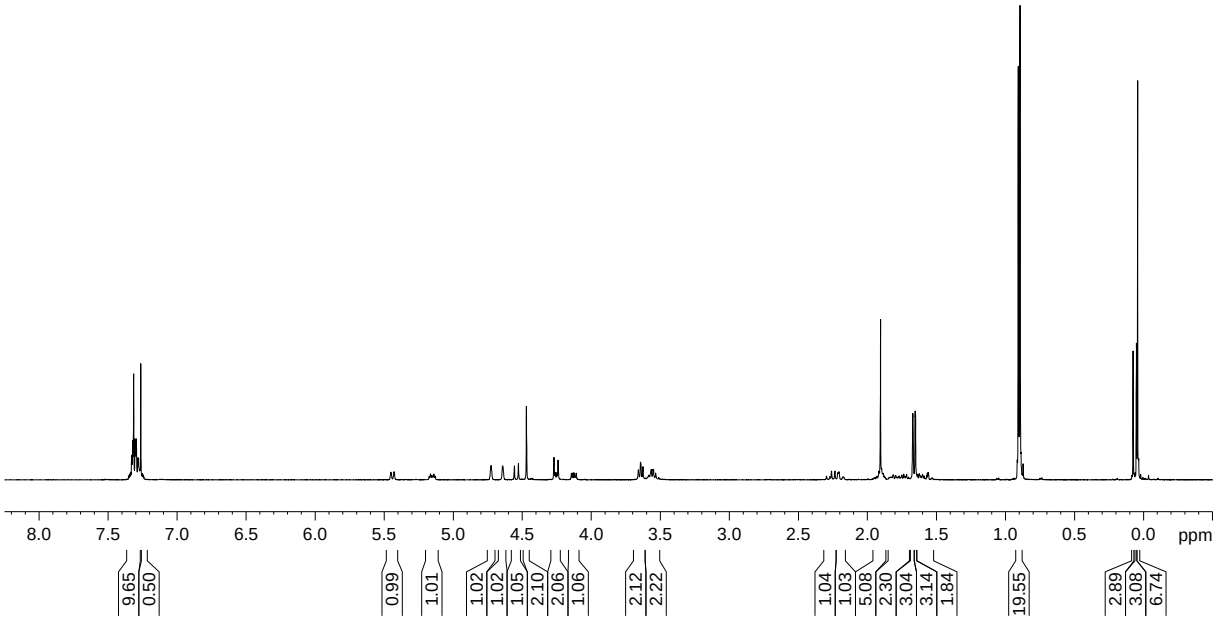
TLC: R_f 0.18 (hexane/EtOAc 12:1, UV, CPS).

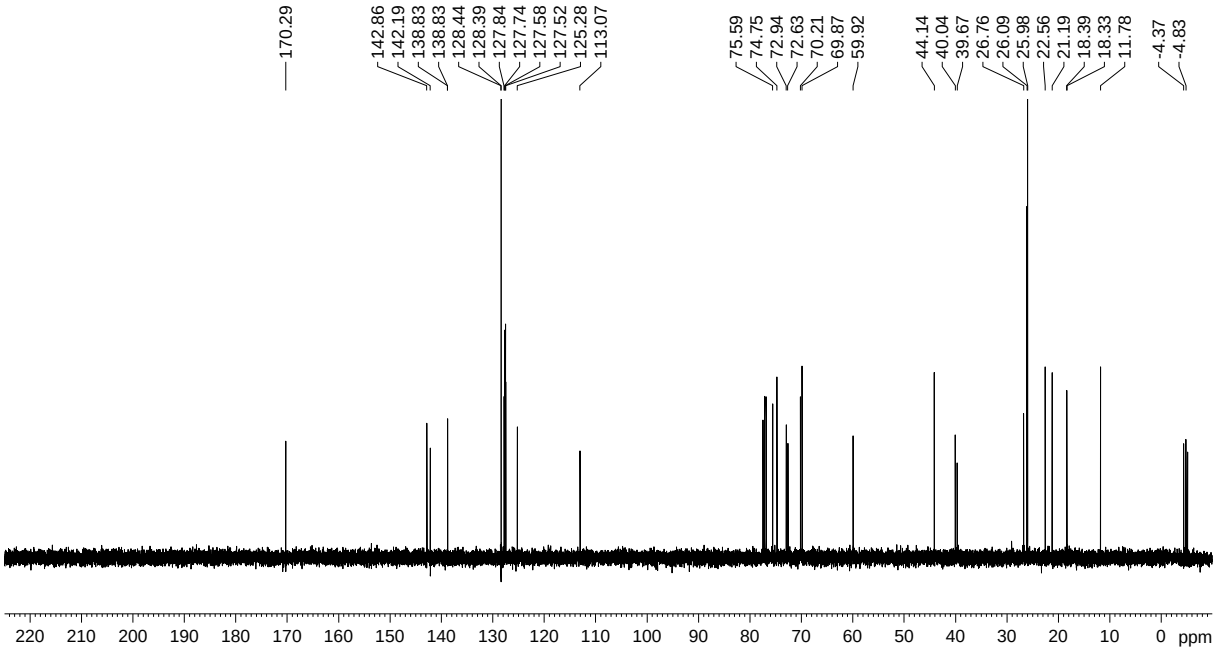
$[\alpha]^{20}_D$: = (*c* = 2.46, $CHCl_3$).

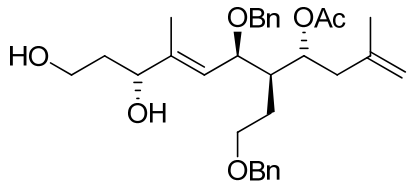
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.21 (m, 10H), 5.44 (d, *J* = 9.0 Hz, 1H), 5.15 (ddd, *J* = 10.0, 3.7, 3.6 Hz, 1H), 4.73 (m, 1H), 4.64 (m, 1H), 4.54 (d, *J* = 11.6 Hz, 1H), 4.47 (s, 2H), 4.26 (d, *J* = 11.6 Hz, 1H), 4.26 (m, 1H), 4.13 (dd, *J* = 9.1, 5.1 Hz, 1H), 3.65 (d, *J* = 6.3 Hz, 1H), 3.63 (d, *J* = 6.3 Hz, 1H), 3.60-3.52 (m, 2H), 2.27 (dd, *J* = 14.3, 10.0 Hz, 1H), 2.19 (dd, *J* = 14.3, 3.0 Hz, 1H), 1.96-1.87 (m, 2H), 1.91 (s, 3H), 1.85-1.77 (m, 1H), 1.75 (dddd, *J* = 13.8, 8.3, 5.9, 5.9 Hz, 1H), 1.67 (s, 3H), 1.65 (d, *J* = 1.2 Hz, 3H), 1.63-1.57 (m, 1H), 0.91 (s, 9H), 0.89 (s, 9H), 0.08 (s, 3H), 0.05 (s, 3H), 0.04 (s, 6H) .

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.3, 142.9, 142.2, 138.8, 138.8, 128.4, 128.4, 127.8, 127.7, 127.6, 127.5, 125.3, 113.1, 75.6, 74.8, 72.9, 72.6, 70.2, 69.9, 59.9, 44.1, 40.0, 39.7, 26.8, 26.1, 26.0, 22.6, 21.2, 18.4, 18.3, 11.8, -4.4, -4.8, -5.1, -5.1.

HRMS (ESI): calculated for $C_{37}H_{60}O_5Si_2$ $[M+Na]^+$ 663.3871, found 663.3880.







469

(4*R*,5*R*,6*R*,9*R*,*E*)-6-(benzyloxy)-5-(2-(benzyloxy)ethyl)-9,11-dihydroxy-2,8-dimethylundeca-1,7-dien-4-yl acetate (469). To a solution of silyl ether **466** (300 mg, 0.41 mmol) in THF (8 mL) was added TBAF•3H$_2$O (256 Mg, 0.81 mmol) in one portion at 0 °C and the reaction mixture was allowed to reach room temperature. After 12 h at this temperature the reaction was then cautiously quenched by addition of saturated aqueous NH$_4$Cl (10 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 10 mL). The combined organic phases were dried over MgSO$_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:3) to yield 176 mg (86%) of diol **469** as a colorless oil.

TLC: R$_f$ 0.10 (hexane/EtOAc 10:3, CPS).

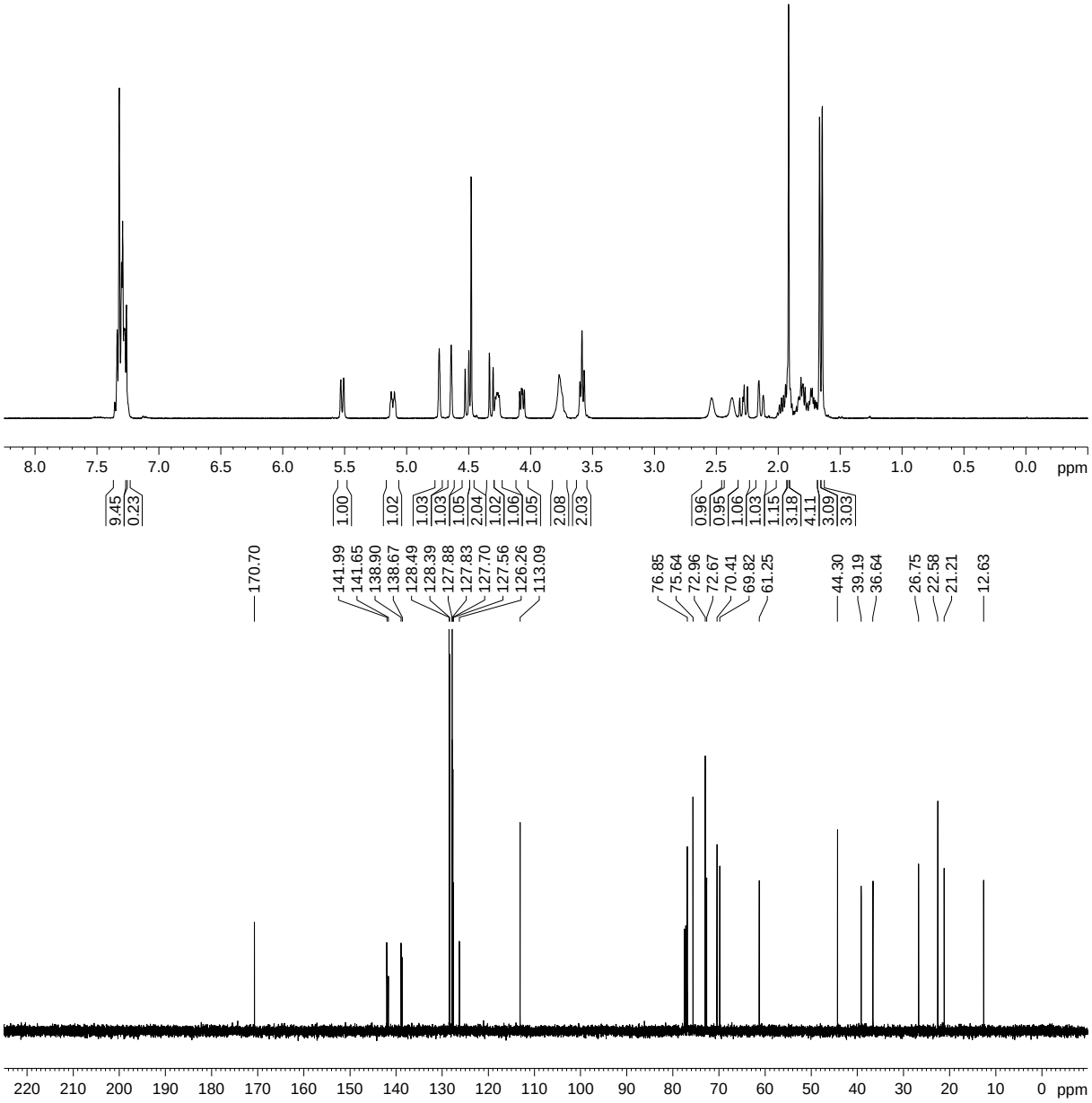
[α]$^{20}_D$: = ° (*c* = 0.45, CHCl$_3$).

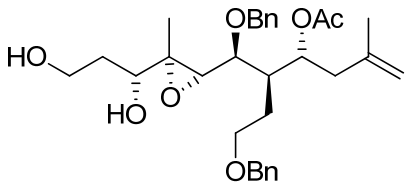
^{1}H-NMR (400 MHz, CDCl$_3$): δ = 7.37-7.22 (m, 10H), 5.52 (d, *J* = 9.1 Hz, 1H), 5.11 (ddd, *J* = 10.6, 3.0, 2.9 Hz, 1H), 4.74 (s, 1H), 4.64 (m, 1H), 4.51 (d, *J* = 12.0 Hz, 1H), 4.48 (s, 2H), 4.32 (d, *J* = 12.0 Hz, 1H), 4.27 (dd, *J* = 7.8, 4.4 Hz, 1H), 4.07 (dd, *J* = 9.1, 5.9 Hz, 1H), 3.82-3.70 (m, 2H), 3.59 (d, *J* = 6.8 Hz, 1H), 3.57 (d, *J* = 6.8 Hz, 1H), 2.54 (br s, 1H), 2.37 (br s,

1H), 2.28 (dd, *J* = 14.3, 10.6 Hz, 1H), 2.14 (d, *J* = 14.3 Hz, 1H), 2.01-1.93 (m, 1H), 1.92 (s, 3H), 1.89-1.76 (m, 3H), 1.76-1.68 (m, 1H), 1.67 (s, 3H), 1.65 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.7, 142.0, 141.7, 138.9, 138.7, 128.5, 128.4, 127.9, 127.8, 127.7, 127.6, 126.3, 113.1, 76.8, 75.6, 73.0, 72.7, 70.4, 69.8, 61.3, 44.3, 39.2, 36.6, 26.7, 22.6, 21.2, 12.6.

HRMS (ESI): calculated for $C_{13}H_{22}O_3Si$ $[M+Na]^+$ 249.1461, found 249.1457.





471

(4*R*,5*R*)-7-(benzyloxy)-5-((*S*)-(benzyloxy)((2*S*,3*S*)-3-((*R*)-1,3-dihydroxypropyl)-3-methyloxiran-2-yl)methyl)-2-methylhept-1-en-4-yl acetate (471).

To a stirred solution of Ti(O*i*Pr)$_4$ (36 µL, 0.12 mmol) and molecular sieves-3Å (160 mg) in CH_2Cl_2 (0.5 mL) were added D-(+)-diethyl tartrate (20 µL, 0.12 mmol) and alcohol **470** (30 mg, 0.06 mmol) in CH_2Cl_2 (0.3 mL) at -35 °C. The reaction mixture was stirred for 15 min and *t*BuOOH (5.33 M in decane, 43 µL, 0.23 mmol) was added dropwise at same temperature. The reaction mixture was then stirred for 3 h at -35 °C. The reaction quenched by addition of water (0.87 mL), the reaction mixture was allowed to reach 0 °C and stirred for 30 min. The reaction mixture was then filtered through a small plug of celite and the precipitate was rinsed with CH_2Cl_2 (3 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (5 x 3 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:2) to yield 20 mg (65%) of epoxide **471** as a colorless oil.

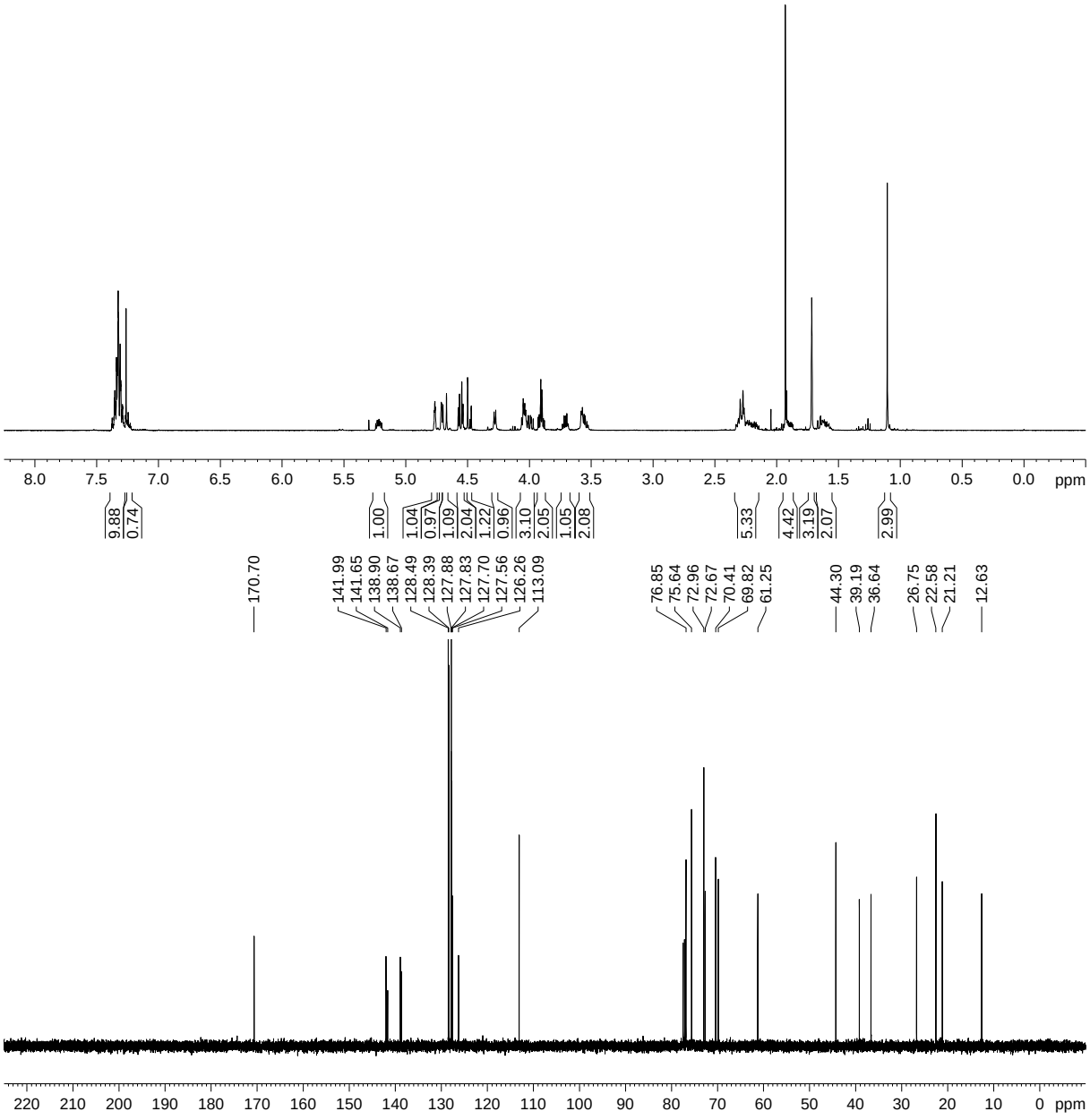
TLC: R_f 0.33 (hexane/EtOAc 3:5, UV (weak), CPS).

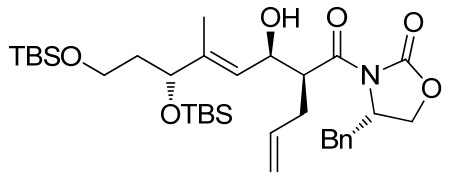
$[\alpha]^{20}_D$: = ° (*c* = 0.45, $CHCl_3$).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.22 (m, 10H), 5.52 (d, *J* = 9.1 Hz, 1H), 5.11 (ddd, *J* = 10.6, 3.0, 2.9 Hz, 1H), 4.74 (s, 1H), 4.64 (m, 1H), 4.51 (d, *J* = 12.0 Hz, 1H), 4.48 (s, 2H), 4.32 (d, *J* = 12.0 Hz, 1H), 4.27 (dd, *J* = 7.8, 4.4 Hz, 1H), 4.07 (dd, *J* = 9.1, 5.9 Hz, 1H), 3.82-3.70 (m, 2H), 3.59 (d, *J* = 6.8 Hz, 1H), 3.57 (d, *J* = 6.8 Hz, 1H), 2.54 (br s, 1H), 2.37 (br s, 1H), 2.28 (dd, *J* = 14.3, 10.6 Hz, 1H), 2.14 (d, *J* = 14.3 Hz, 1H), 2.01-1.93 (m, 1H), 1.92 (s, 3H), 1.89-1.76 (m, 3H), 1.76-1.68 (m, 1H), 1.67 (s, 3H), 1.65 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.7, 142.0, 141.7, 138.9, 138.7, 128.5, 128.4, 127.9, 127.8, 127.7, 127.6, 126.3, 113.1, 76.8, 75.6, 73.0, 72.7, 70.4, 69.8, 61.3, 44.3, 39.2, 36.6, 26.7, 22.6, 21.2, 12.6.

HRMS (ESI): calculated for $C_{13}H_{22}O_3Si$ $[M+Na]^+$ 249.1461, found 249.1457.





410

(*S*)-3-((2*S*,3*R*,6*R*,*E*)-2-allyl-6,8-bis((*tert*-butyldimethylsilyl)oxy)-3-hydroxy-5-methyloct-4-enoyl)-4-benzyloxazolidin-2-one (410). To a solution of Evans-auxiliary derivate **425** (0.51 g, 1.96 mmol) in CH_2Cl_2 (20 mL) was added n-Bu_2OTf (2.14 mL, 1M in CH_2Cl_2, 2.14 mmol) and DIPEA (0.40 mL, 2.32 mmol) dropwise at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 0.5 h. The reaction mixture was then cooled to -78

°C. Aldehyde **411** (0.67 g, 1.78 mmol) in CH_2Cl_2 (5 mL) was added over a period of 15 min and the reaction mixture was stirred for 1 h at -78 °C. The reaction was then cautiously quenched by addition of pH 7 buffer (5 mL), MeOH (25 mL) and 30% aqueous H_2O_2 (5 mL). The resulting mixture was stirred at 0 °C for 1 h, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 8:1) to yield 0.90 g (80%) of aldol product **410** as a single isomer.

Note: The excess of the aldehyde can be reisolated.

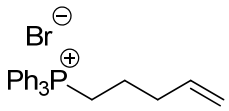
TLC: R_f 0.23 (hexane/EtOAc 9:1, UV, CPS).

$[\alpha]^{20}_D$: = 24.9 (*c* = 0.24, $CHCl_3$).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37-7.27 (m, 3H), 7.24-7.19 (m, 2H), 5.85 (dddd, *J* = 17.0, 10.2. 7.9, 6.2 Hz, 1H), 5.47 (dp, *J* = 8.8, 1.2 Hz, 1H), 5.11 (dm, *J* = 17.0 Hz, 1H), 5.04 (dm, *J* = 10.2 Hz, 1H), 4.71 (ddd, *J* = 11.8, 5.8, 3.3 Hz, 1H), 4.65 (ddd, *J* = 13.1, 6.8, 3.3 Hz, 1H), 4.25 (ddd, *J* = 9.5, 5.6, 4.8 Hz, 1H), 4.21-4.11 (m, 3H), 3.63 (dd, *J* = 7.1, 3.6 Hz, 1H), 3.61 (dd, *J* = 7.1, 2.4 Hz, 1H), 3.30 (ddd, *J* = 13.4, 3.5, 3.4 Hz, 1H), 2.62 (dd, *J* = 13.3, 10.2 Hz, 1H), 2.54-2.44 (m, 2H), 2.16 (d, *J* = 3.4 Hz, 1H), 1.76-1.68 (m, 1H), 1.67 (d, *J* = 1.2 Hz, 3H), 1.59 (ddd, *J* = 7.5, 7.4, 4.6 Hz, 1H), 0.88 (s, 18H), 0.04 (s, 6H), 0.04 (s, 3H), -0.01 (s, 3H).

IR (film): ν 2954, 2928, 2857, 1783, 1699, 1472, 1387, 1252, 1209, 1099, 1006, 915, 836, 777, 744, 701.

HRMS (ESI): calculated for $C_{40}H_{63}NO_7Si_2$ $[M+Na]^+$ 654.3617, found 654.3631.



484

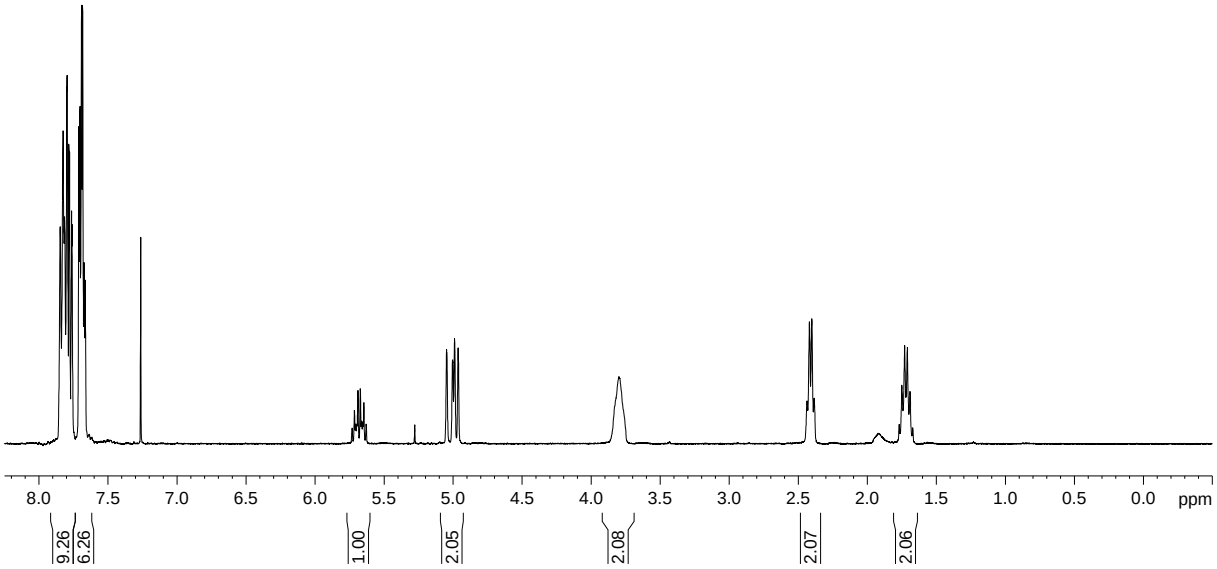
Pent-4-en-1-yltriphenylphosphonium bromide (484). To a solution of bromide **483** (0.1 mL, 0.85 mmol) in MeCN (6 mL) was added triphenylphosphine (0.33 g, 1.27 mmol) at room temperature and the reaction was heated up to reflux for 31 h. The solvent was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 50:1 → 10:1) to give 0.28 g (81%) of Wittig salt **484** as a white solid.

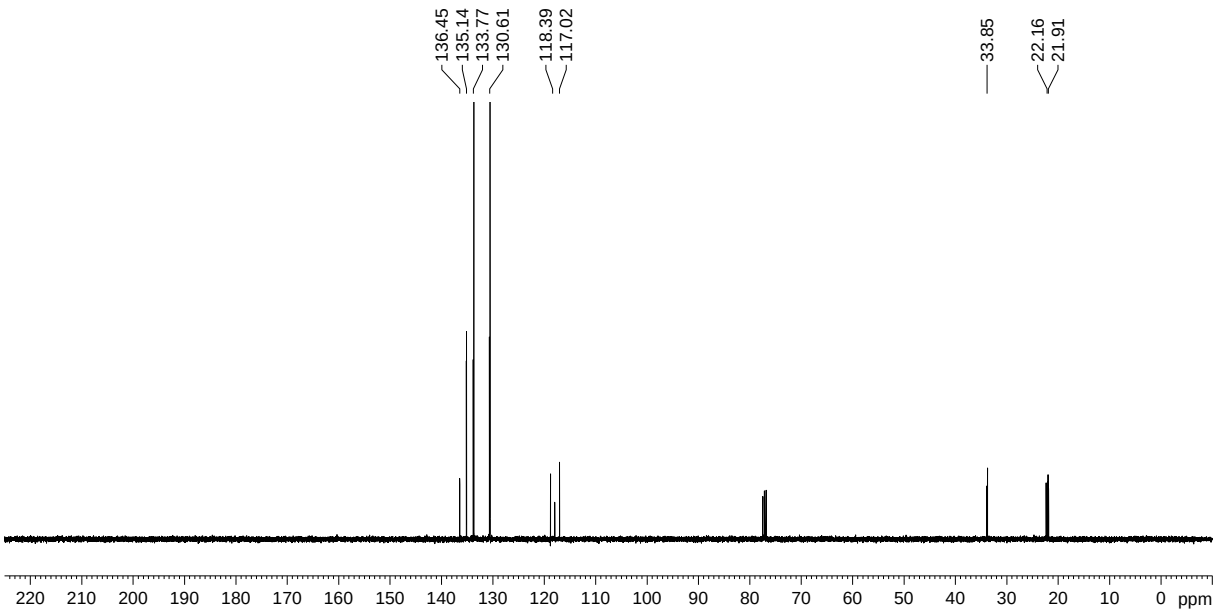
TLC: R_f 0.50 (CH_2Cl_2 /MeOH 10:1, UV, CPS).

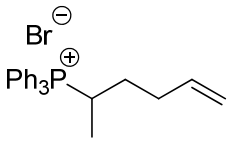
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.86-7.75 (m, 9H), 7.69 (t, *J* = 7.8, 3.4 Hz, 6H), 5.68 (ddd, *J* = 17.1, 10.2, 6.7, 6.7 Hz, 1H), 5.02 (d, *J* = 17.1 Hz, 1H), 4.97 (d, *J* = 10.2 Hz, 1H), 3.86-3.75 (m, 2H), 2.41 (td, *J* = 7.1, 6.7 Hz, 2H), 1.72 (tdt, *J* = 7.8, 7.8, 7.1 Hz, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 136.5, 135.1 (d, *J* = 3.2 Hz), 133.8 (d, *J* = 10.0 Hz), 130.6 (d, *J* = 12.0 Hz,), 118.4 (d, *J* = 86.0 Hz), 117.0, 33.9 (d, *J* = 16.2 Hz,), 22.2 (d, *J* = 35.1 Hz), 21.9 (d, *J* = 11.6 Hz).

HRMS (ESI): calculated for $C_{13}H_{22}O_3Si$ $[M+Na]^+$ 249.1461, found 249.1457.







485

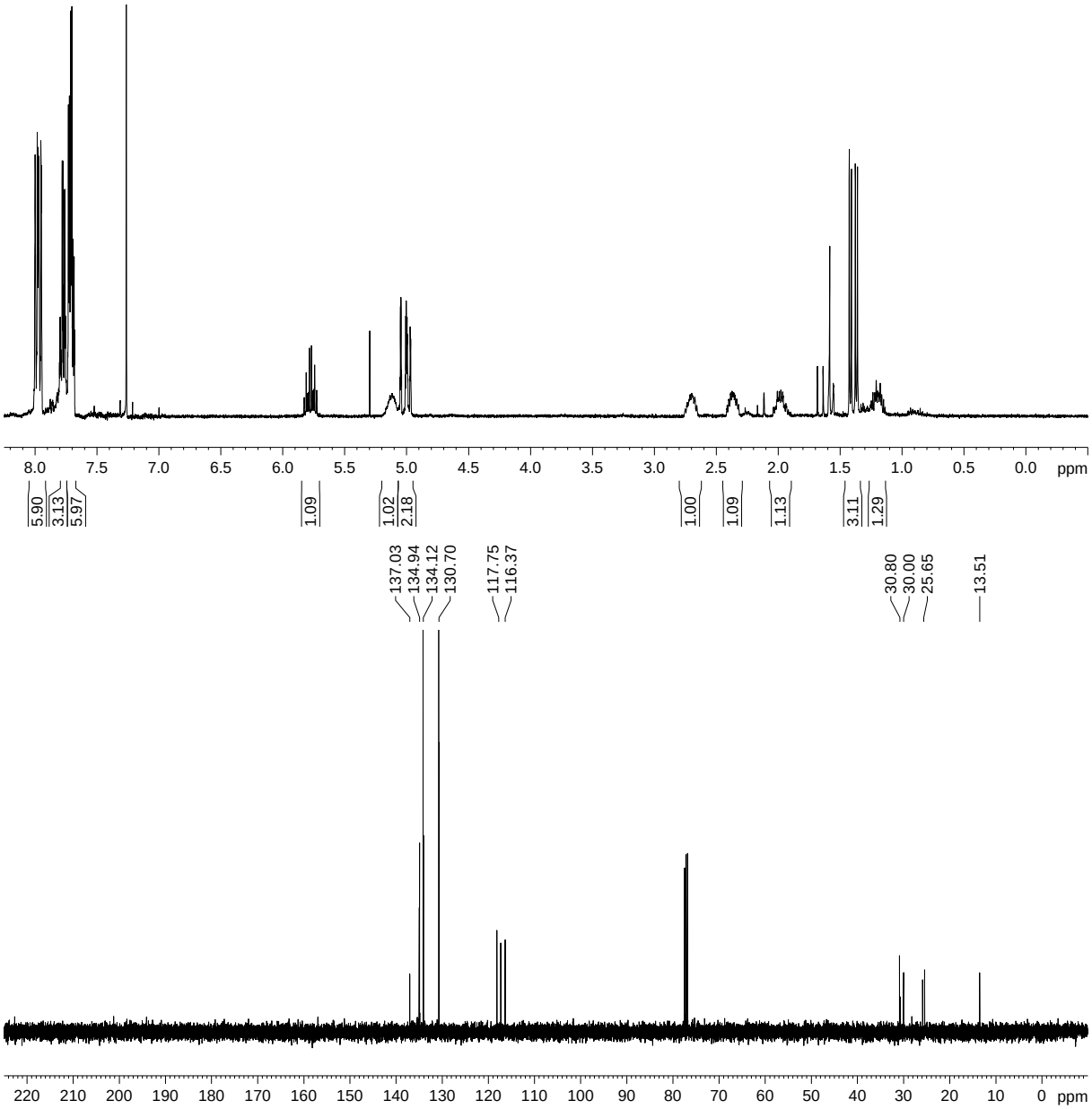
Hex-5-en-2-yltriphenylphosphonium bromide (485). To a solution of wittig salt **484** (0.28 g, 0.68 mmol) in THF (7 mL) was added *n*-BuLi (1.6 M in hexane, 0.47 mL, 0.75 mmol) at 0 °C and the orange reaction mixture was stirred for 1 h at this temperature. MeI (0.17 mL, 2.72 mmol) was then added and the reaction mixture was allowed to reach room temperature and stirred for 1.5 h. The white suspension was then concentrated under reduced pressure and the residue was purified by flash column chromatography (CH_2Cl_2 /MeOH 50:1 → 20:1) to give 0.28 g (97%) of Wittig salt **485** as a white solid.

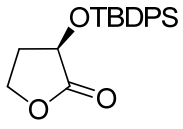
TLC: R_f 0.34 (CH_2Cl_2 /MeOH 20:1, UV, CPS).

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.98 (d, *J* = 11.8 Hz, 3H), 7.96 (dd, *J* = 11.8, 1.3 Hz, 3H), 7.79-7.75 (m, 3H), 7.73-7.68 (m, 6H), 5.78 (ddd, *J* = 17.1, 10.2, 6.7, 6.7 Hz, 1H), 5.19-5.07 (m, 1H), 5.03 (dq *J* = 17.1, 1.6 Hz, 1H), 4.98 (dq, *J* = 10.2, 1.2 Hz, 1H), 2.76-2.65 (m, 1H), 2.42-2.31 (m, 1H), 2.04-1.92 (m, 1H), 1.39 (dd, *J* = 19.6, 7.1 Hz, 3H), 1.26-1.13 (m, 1H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 137.0, 134.9 (d, *J* = 3.1 Hz), 134.1 (d, *J* = 9.3 Hz), 130.7 (d, *J* = 12.2 Hz), 117.8 (d, *J* = 83.1 Hz), 116.4, 30.8 (d, *J* = 14.8 Hz), 30.0, 25.6 (d, *J* = 46.0 Hz), 13.5 (d, *J* = 2.2 Hz).

HRMS (ESI): calculated for $C_{13}H_{22}O_3Si$ $[M+Na]^+$ 249.1461, found 249.1457.


8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 ppm
5.90
3.13
5.97
1.09
1.02
2.18
1.00
1.09
1.13
3.11
1.29
137.03
134.94
134.12
130.70
117.75
116.37
30.80
30.00
25.65
13.51
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm



500

(*R*)-3-((*tert*-butyldiphenylsilyl)oxy)dihydrofuran-2(3*H*)-one (500). To a solution of (*R*)-α-hydroxybutyrolactone **417** (1.18 g, 11.56 mmol) and imidazole (1.97 g, 28.90 mmol) in DMF (10 mL) was added TBDPSCl (3.61 mL, 13.87 mmol) at room temperature and the mixture was stirred for 19 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (20 mL). The aqueous phase was extracted with Et_2O (3 x 30 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 10:1) afforded 3.75 g (95%) of silyl ether **500** as a white solid.

TLC: R_f 0.18 (hexane/EtOAc 15:1, UV, CPS).

$[\alpha]^{20}_D$: = 38.1 (*c* = 1.60, $CHCl_3$).

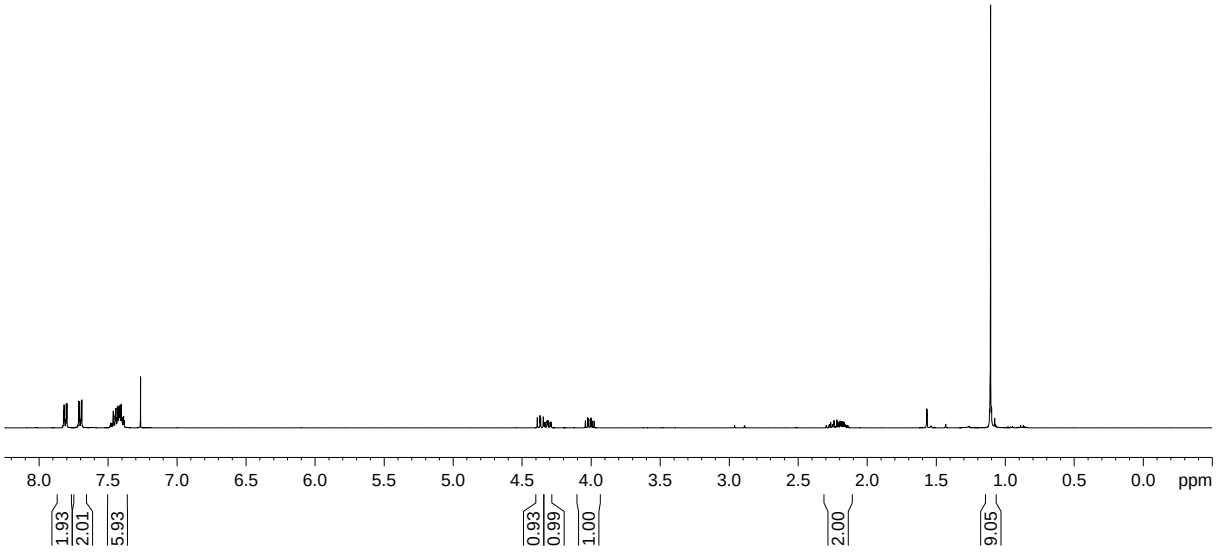
mp: 85-87 °C.

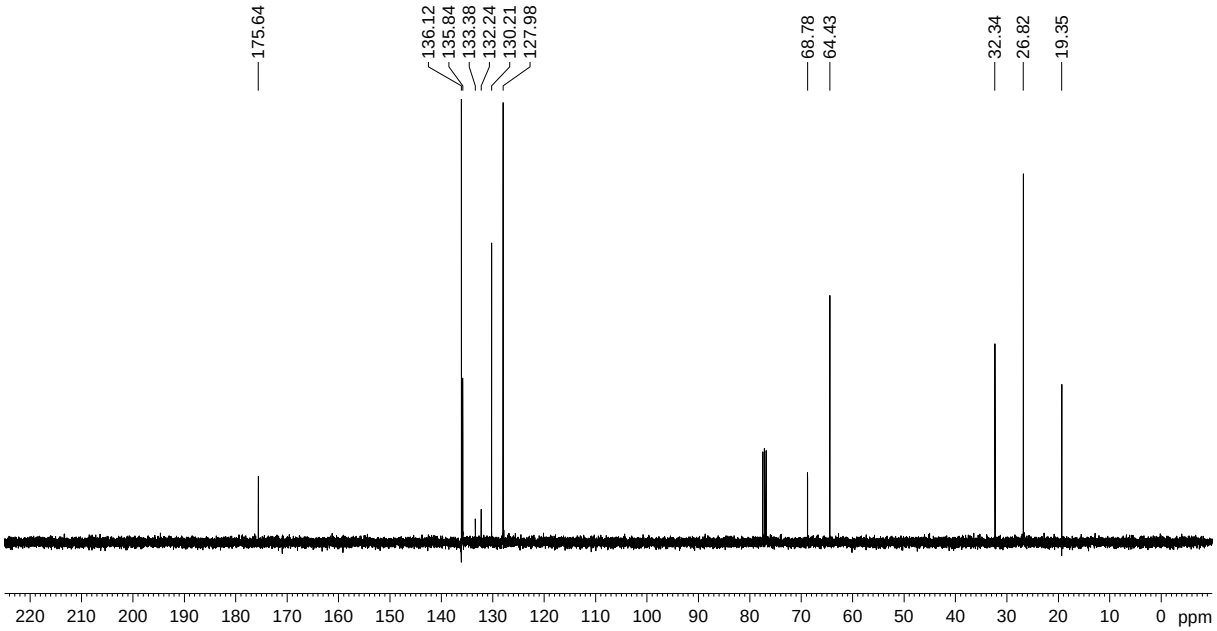
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.87 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.70 (ddd, *J* = 6.5, 1.4, 1.4 Hz, 2H), 7.49-7.38 (m, 6H), 4.37 (dd, *J* = 9.5, 8.0 Hz, 1H), 4.32 (ddd, *J* = 9.2, 8.7, 2.7 Hz), 4.00 (ddd, *J* = 9.8, 9.5, 6.4 Hz, 1H), 2.23 (dddd, *J* = 12.5, 9.7, 9.7, 8.3 Hz, 1H), 2.17 (dddd, *J* = 9.2, 8.0, 6.4, 2.8 Hz, 1H), 1.11 (s, 9H).

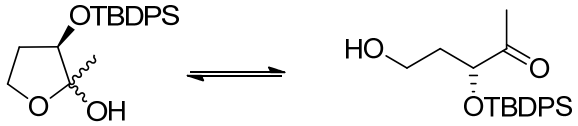
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 175.6, 136.1, 135.8, 133.4, 132.2, 130.2, 128.0, 68.8, 64.4, 32.3, 26.8, 19.4.

IR (film): ν 3072, 3049, 2932, 2858, 1787, 1487, 1472, 1428, 1391, 1361, 1284, 1218, 1148, 1106, 1021, 998, 948, 885, 840, 822, 742, 702, 656, 613, 518, 501

HRMS (ESI): calculated for $C_{20}H_{24}O_3Si$ $[M+Na]^+$ 363.1387, found 363.1395.







501 **501**

(2*R*, 3*R*)-3-((*tert*-butyldiphenylsilyl)oxy)-2-methyltetrahydrofuran-2-ol (501).

(2*S*, 3*R*)-3-((*tert*-butyldiphenylsilyl)oxy)-2-methyltetrahydrofuran-2-ol (501).

(*R*)-3-((*tert*-butyldiphenylsilyl)oxy)-5-hydroxypentan-2-one (501. To a solution of silyl ether **500** (3.72 g, 10.93 mmol) in THF (50 mL) was added MeLi (3.0 M in Et_2O, 4.01 mL, 12.02 mmol) dropwise at -78 °C and the reaction mixture was stirred for 2 h at this temperature. The cooling bath was removed and the reaction was quenched by addition of saturated aqueous NH_4Cl (30 mL). The mixture was diluted with saturated aqueous Rochelle salt (20 mL) and Et_2O (50 mL). The phases were separated and the aqueous phase was extracted with Et_2O (2 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 3.39 g (87%) of a mixture of cyclic acetal **501** and linear alcohol **501** as white crystals.

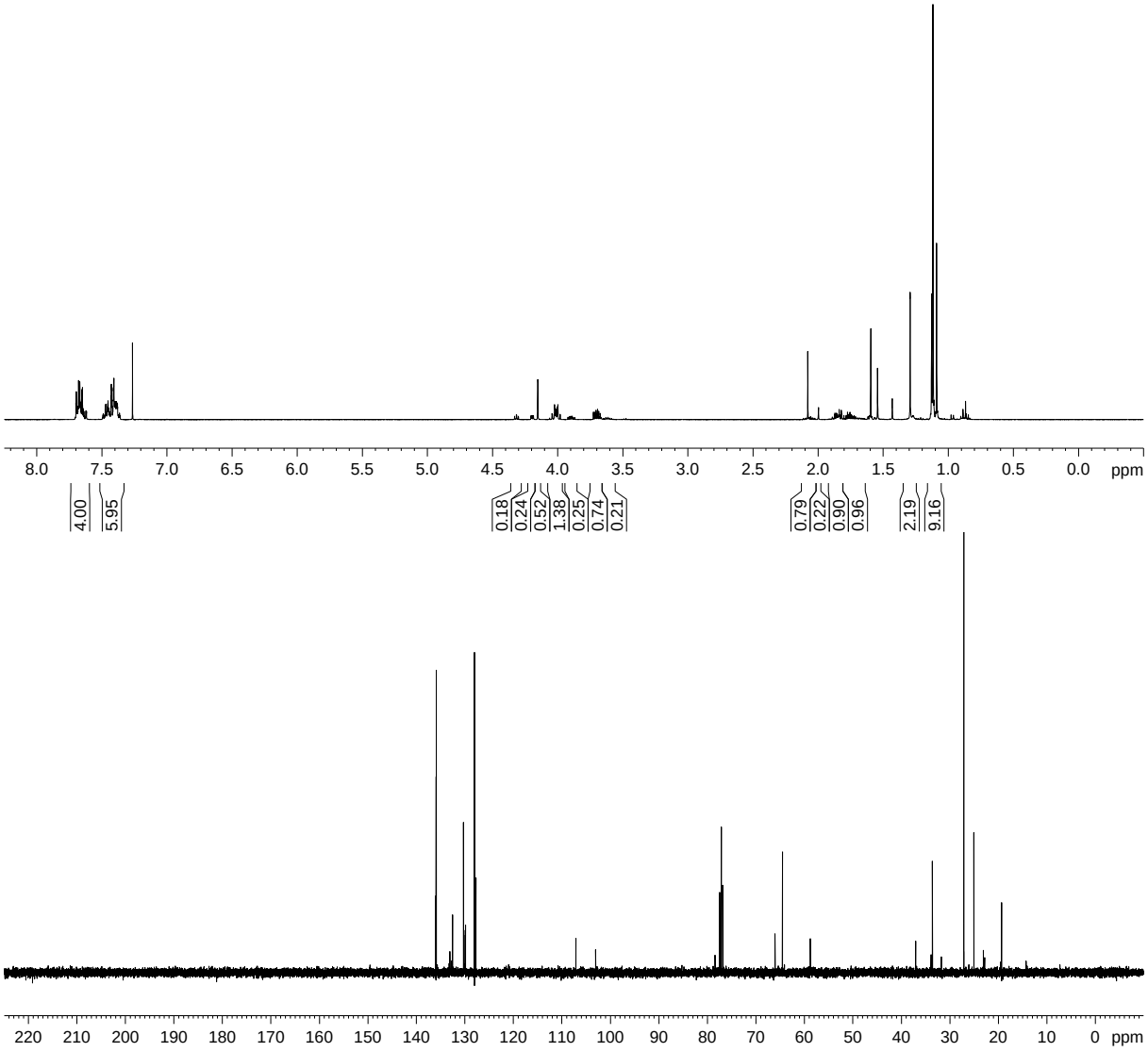
Note: No investigations have been carried out to assign the ^1H and ^{13}C-NMR signals to the single alcohols.

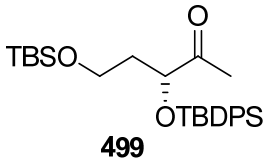
TLC: R_f 0.20 (hexane/EtOAc 7:1, UV, CPS).

$[\alpha]^{20}_D$: -25.3° (*c* = 3.33, $CHCl_3$).

IR (film): ν 3421, 3071, 3048, 2932, 2891, 1717, 1487, 1472, 1426, 1391, 1376, 1362, 1188, 1107, 1054, 1022, 998, 923, 893, 822, 740, 701, 612, 508.

HRMS (ESI): calculated for $C_{21}H_{28}O_3Si$ $[M+Na]^+$ 379.1700, found 379.1704.





499

(*R*)-5-((*tert*-butyldimethylsilyl)oxy)-3-((*tert*-butyldiphenylsilyl)oxy)pentan-2-one (499). To a solution of alcohol **501** (3.29 g, 9.23 mmol) and imidazole (1.45 g, 21.22 mmol) in DMF (8 mL) was added TBSCl (1.60 g, 10.61 mmol) at room temperature and the mixture was stirred for 13 h. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (15 mL). The aqueous phase was extracted with Et_2O (3 x 20 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification

of the residue by flash column chromatography (hexane/EtOAc 30:1) afforded 3.68 g (98%) of silyl ether **499** as a colorless oil.

TLC: R_f 0.27 (hexane/EtOAc 50:1, UV, CPS).

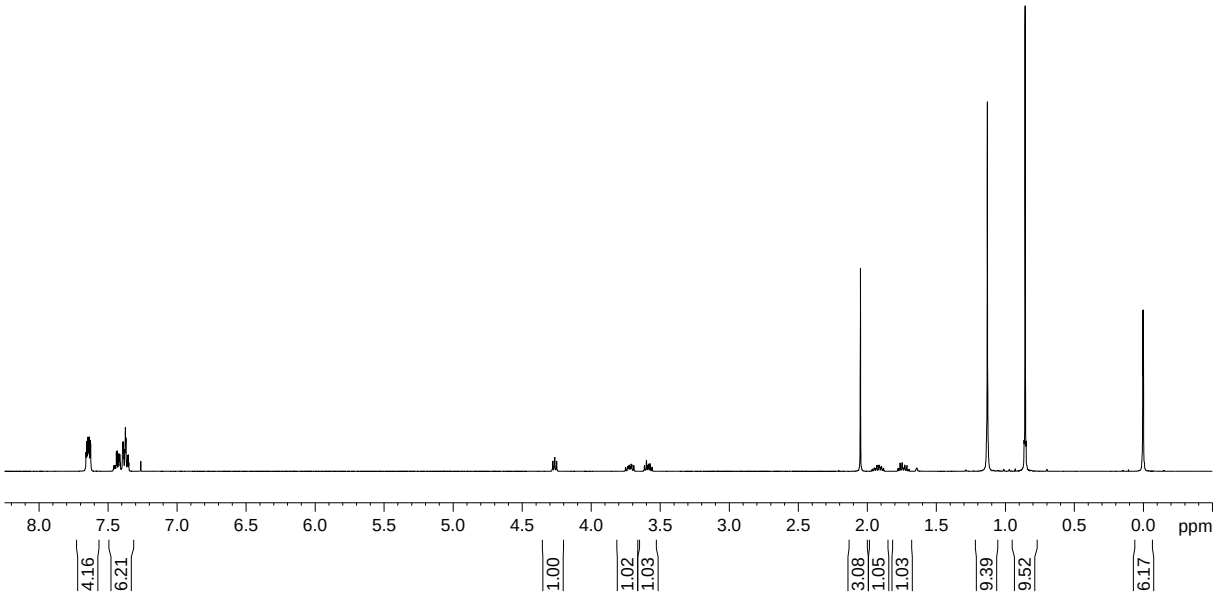
$[\alpha]^{20}_D$: = -15.5° (*c* = 3.40, $CHCl_3$).

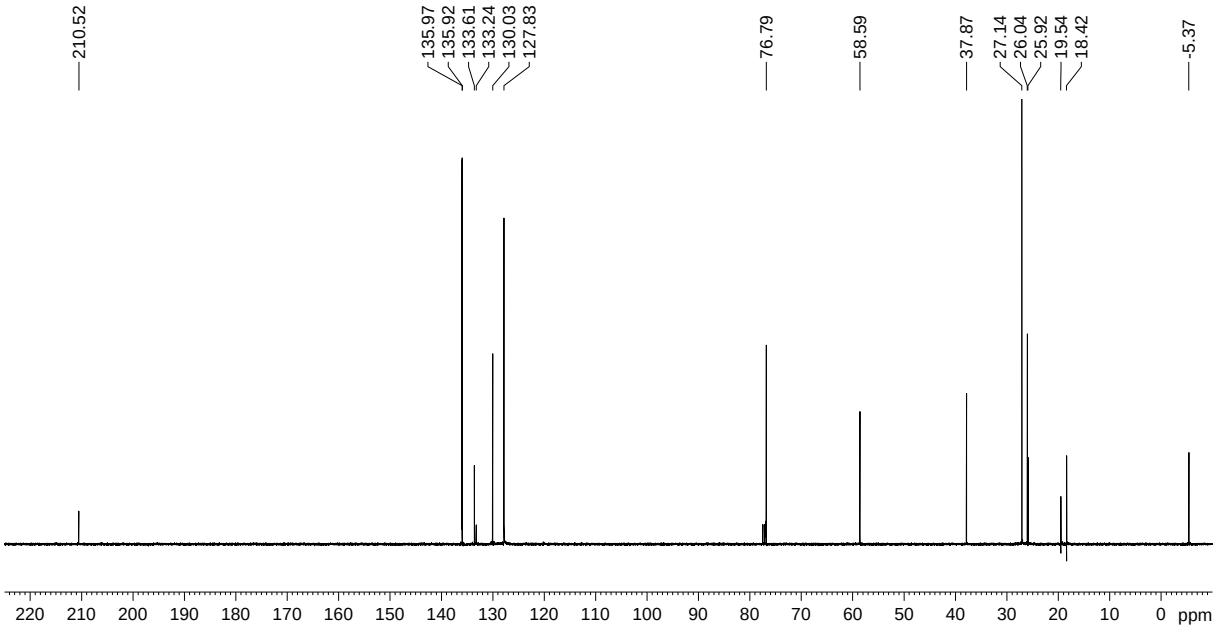
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.67-7.62 (m, 4H), 7.46-7.35 (m, 6H), 4.26 (dd, *J* = 6.1, 5.3 Hz, 1H), 3.72 (ddd, *J* = 10.2, 7.5, 5.8 Hz), 3.59 (ddd, *J* = 10.2, 5.8, 5.8 Hz, 1H), 2.05 (s, 3H), 1.92 (dddd, *J* = 13.8, 7.5, 6.1, 5.4 Hz, 1H), 1.74 (dddd, *J* = 13.8, 6.1, 5.8, 5.8, 1H), 1.13 (s, 9H), 0.86 (s, 9H), 0.88 (s, 9H), 0.01 (s, 3H), 0.00 (s, 3H).

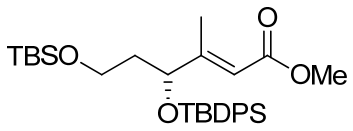
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 210.5, 136.0, 135.9, 133.6, 133.2, 130.0, 127.8, 76.8, 58.6, 37.9, 27.1, 26.0, 25.9, 19.5, 18.4, -5.4, -5.4.

IR (film): ν 3072, 2955, 2930, 2887, 2857, 1732, 1718, 1472, 1428, 1391, 1361, 1350, 1254, 1107, 1006, 916, 834, 776, 740, 701, 663, 610, 503

HRMS (ESI): calculated for $C_{27}H_{42}O_3Si_2$ $[M+Na]^+$, found.







502

(*R*,*E*)-methyl 6-((*tert*-butyldimethylsilyl)oxy)-4-((*tert*-butyldiphenylsilyl)oxy)-3-methylhex-2-enoate (502). To a solution of NaH (1.14 mg, 28.55 mmol, 60% in mineral oil) in THF (130 mL) was added trimethyl phosphonacetate **421** (4.26 mL, 29.44 mmol) at 0 °C and the reaction mixture was stirred for 1 h at this temperature (the solution did not clear off). The reaction mixture was allowed to reach room temperature and ketone **499** (4.20 g, 8.92 mmol) in THF (20 mL) was added. The reaction mixture heated to 45 °C and stirred for 21 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (100 mL). The aqueous phase was extracted with Et_2O (3 x 100 mL), the combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 50:1) afforded 4.70 g (99%, *E*/*Z* 20:1) of unsaturated ester **502** as a single isomer as a colorless oil.

TLC: R_f 0.19 (hexane/EtOAc 30:1, UV, CPS).

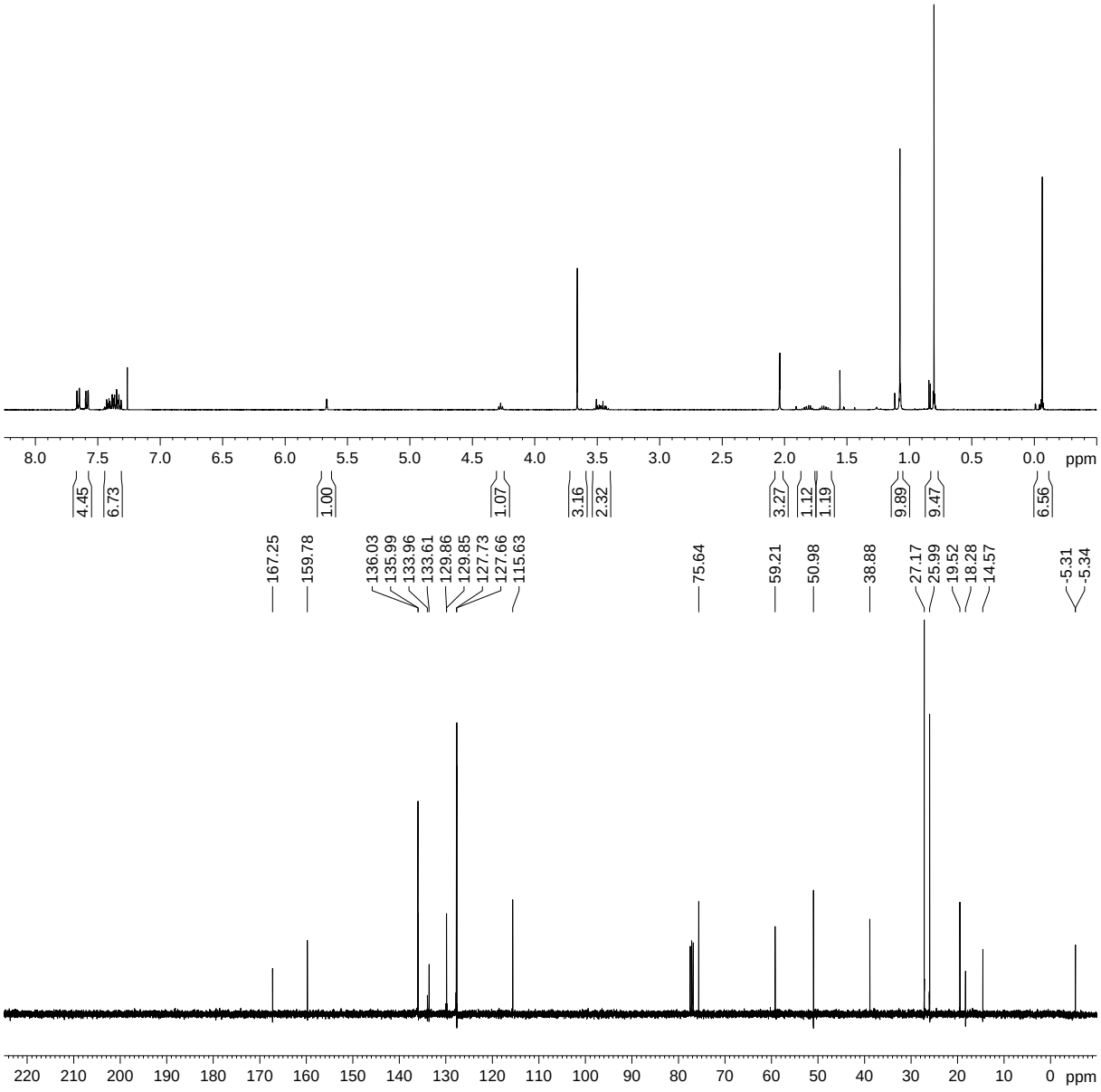
$[\alpha]^{20}_D$: -13.1° (*c* = 2.00, $CHCl_3$).

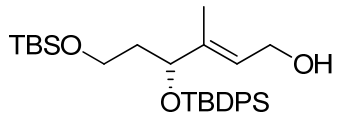
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.66 (dd, *J* = 6.5, 1.5 Hz, 2H), 7.59 (dd, *J* = 6.5, 1.5 Hz, 2H), 7.45-7.30 (m, 6H), 5.67 (p, *J* = 1.2 Hz, 1H), 4.27 (dd, *J* = 6.9, 5.1 Hz, 1H), 3.66 (s, 3H), 3.49 (ddd, *J* = 10.2, 7.0, 5.8 Hz, 1H), 3.60 (ddd, *J* = 10.2, 7.0, 6.9 Hz, 1H), 2.04 (d, *J* = 1.2 Hz, 1H), 1.82 (dqd, *J* = 13.7, 7.0, 5.8 Hz, 1H), 1.68 (dqd, *J* = 13.7, 6.6, 5.9Hz, 1H), 1.08 (s, 9H), 0.80 (s, 9H), -0.06 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 167.2, 159.8, 136.0, 136.0, 134.0, 133.6, 129.9, 129.8, 127.7, 127.7, 115.6, 75.6, 59.2, 51.0, 38.9, 27.2, 26.0, 19.5, 18.3, 14.6, -5.3, -5.3.

IR (film): ν 2953, 2929, 2893, 2858, 1722, 1655, 1472, 1429, 1390, 1362, 1255, 1223, 1156, 1105, 1036, 1007, 940, 890, 834, 776, 739, 701, 613, 509.

HRMS (ESI): calculated for $C_{30}H_{46}O_4Si_2$ $[M+Na]^+$ 527.3007, found 527.3003.





503

(*R*,*E*)-6-((*tert*-butyldimethylsilyl)oxy)-4-((*tert*-butyldiphenylsilyl)oxy)-3-methylhex-2-en-1-ol (503). To a solution of ester **502** (4.70 g, 8.92 mmol) in CH_2Cl_2 (100 mL) was added

DIBAL-H (1 M in CH_2Cl_2, 19.63 mL, 19.63 mmol) dropwise at -78 °C and the reaction mixture was stirred for 30 min at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (150 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to yield 4.20 g (94%) of allylic alcohol **503** as a colorless oil.

TLC: R_f 0.24 (hexane/EtOAc 9:1, UV, CPS).

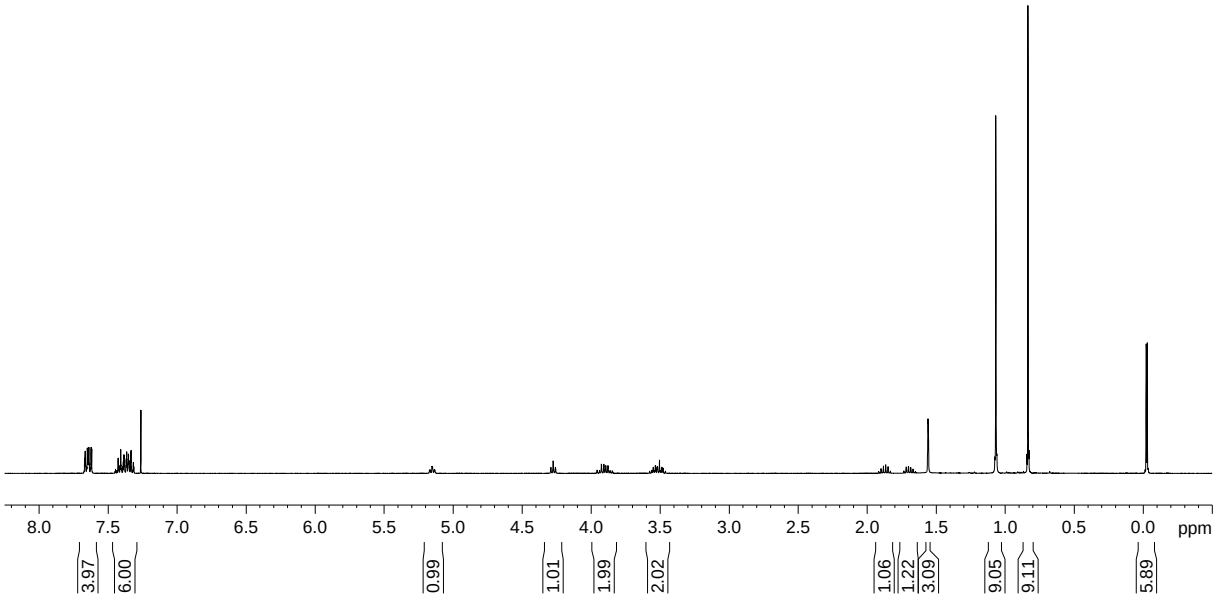
$[\alpha]^{20}_D$: -3.1° (*c* = 1.33, $CHCl_3$).

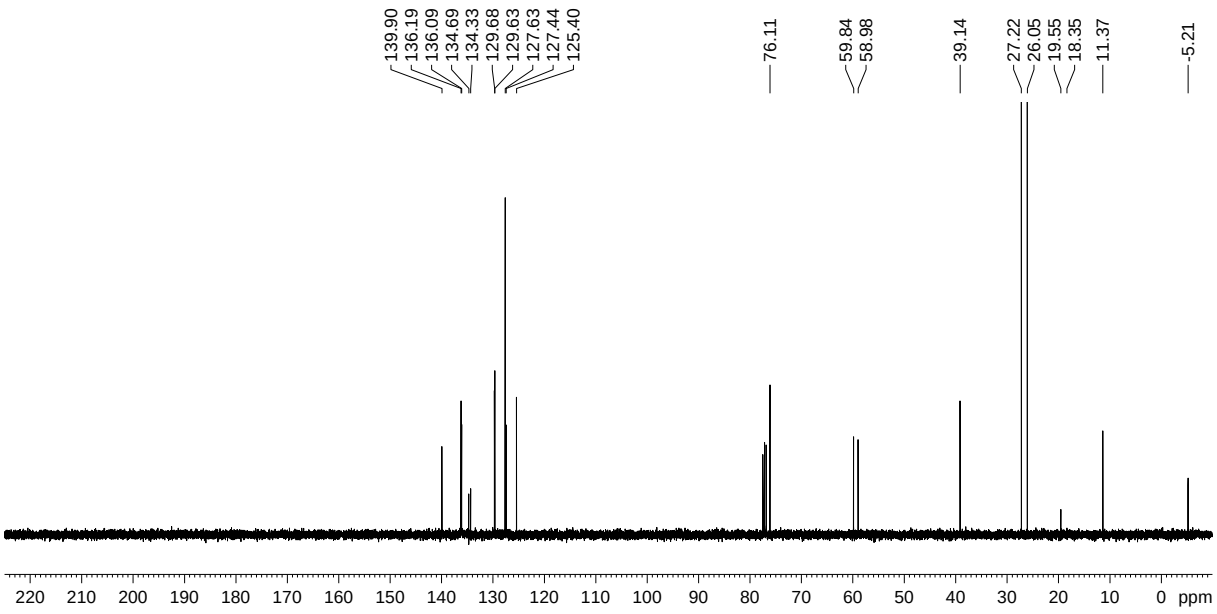
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.66 (ddd, *J* = 6.5, 1.5, 1.5 Hz, 2H), 7.63 (ddd, *J* = 6.5, 1.5, 1.5 Hz, 2H), 7.45-7.31 (m, 6H), 5.15 (tt, *J* = 6.7, 1.2 Hz, 1H), 427 (t, *J* = 6.5 Hz, 1H), 3H), 3.90 (dd, *J* = 12.6, 7.1 Hz, 1H), 3.87 (dd, *J* = 12.6, 6.5 Hz, 1H), 3.54 (ddd, *J* = 10.2, 7.1, 6.1 Hz, 1H), 3.48 (dd, *J* = 10.2, 6.8 Hz, 1H), 1.87 (ddd, *J* = 13.4, 6.7, 6.7 Hz, 1H), 1.69 (ddd, *J* = 13.4, 6.9, 6.3 Hz, 1H), 1.56 (d, *J* = 1.2 Hz, 3H), 1.07 (s, 9H), 0.80 (s, 9H), 0.04 (s, 3H), -0.02 (s, 6H), -0.03 (s, 3H).

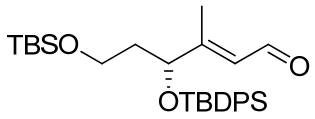
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 139.9, 136.2, 136.1, 134.7, 134.3, 129.7, 129.6, 127.6, 127.4, 125.4, 76.1, 59.8, 59.0, 39.1, 27.2, 26.1, 19.6, 18.4, 11.4, -5.2.

IR (film): ν 3358, 3072, 2954, 2929, 2887, 2857, 1472, 1428, 1389, 1361, 1255, 1109, 1079, 1006, 940, 835, 776, 754, 702, 665, 613, 508..

HRMS (ESI): calculated for $C_{29}H_{46}O_3Si_2$ $[M+Na]^+$ 521.2878, found 521.2878.







498

(*R*,*E*)-6-((*tert*-butyldimethylsilyl)oxy)-4-((*tert*-butyldiphenylsilyl)oxy)-3-methylhex-2-enal (498). To a solution of allylic alcohol **503** (1.35 g, 2.70 mmol) in Et_2O (25 mL) was added MnO_2 (3.75 g, 43.15 mmol) in one portion at room temperature and the solution was stirred for 3 h. The reaction mixture was then filtered through a small plug of celite and the precipitate was rinsed with Et_2O (50 mL). The filtrate was concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 25:1) to yield 1.28 g of unsaturated aldehyde **498** (96%) as a colorless oil.

TLC: R_f 0.22 (hexane/EtOAc 25:1, UV, CPS).

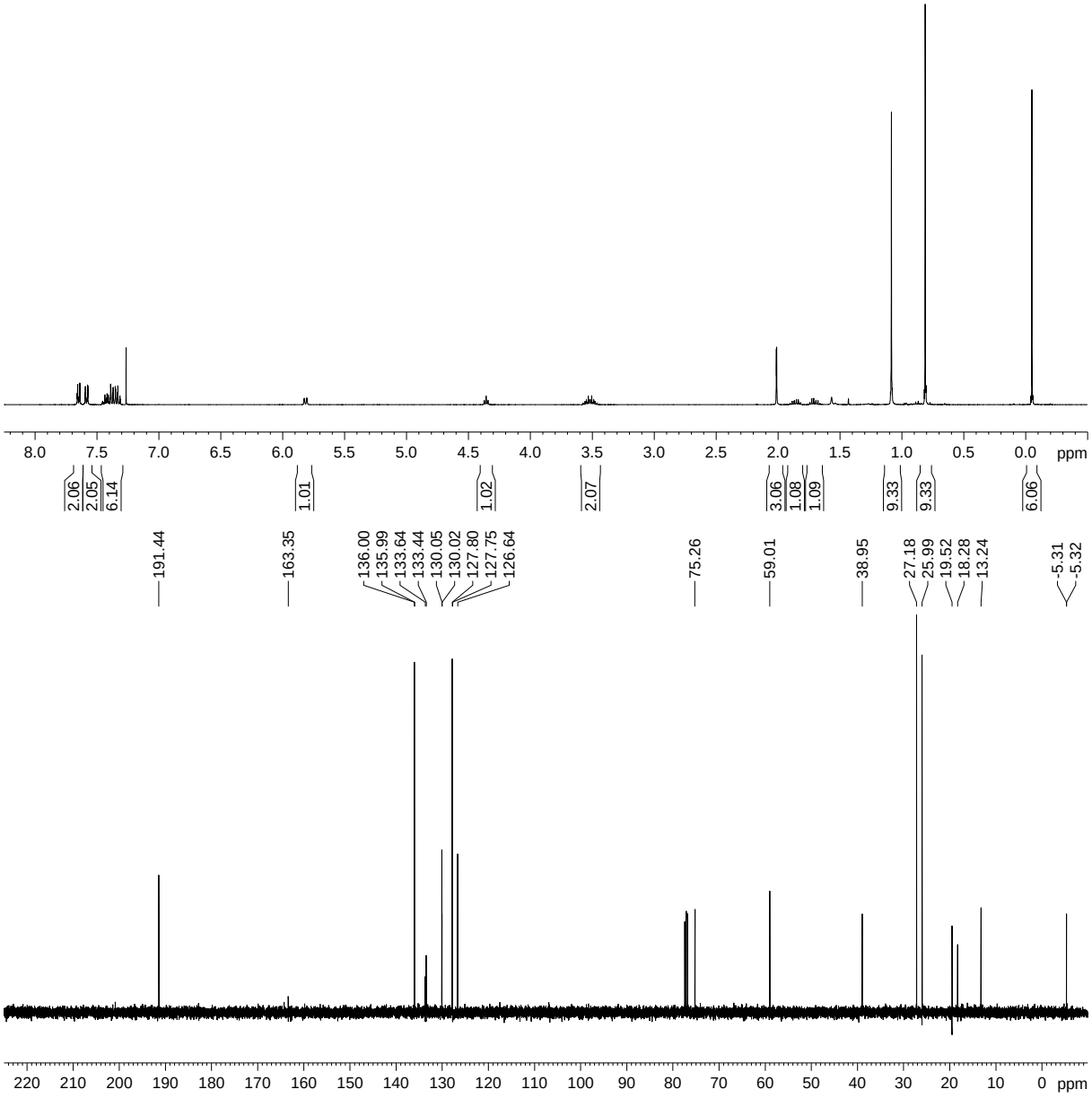
$[\alpha]^{20}_D$: = 4.3° (*c* = 1.00, $CHCl_3$).

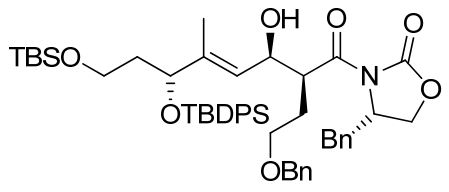
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.87 (d, *J* = 8.0 Hz, 1H), 7.65 (ddd, *J* = 6.5, 1.3, 1.3 Hz, 2H), 7.58 (ddd, *J* = 6.5, 1.3, 1.3 Hz, 2H), 7.46-7.30 (m, 6H), 5.81 (dp, *J* = 8.0, 1.0 Hz, 1H), 4.35 (dd, *J* = 6.5, 6.5 Hz, 1H), 3.54 (ddd, *J* = 10.3, 6.3, 6.2, 1H), 3.49 (ddd, *J* = 10.3, 6.6, 6.5 Hz, 1H), 2.01 (d, *J* = 1.2 Hz, 3H), 1.86 (dddd, *J* = 13.6, 6.8, 6.8, 6.1 Hz, 1H), 1.70 (dddd, *J* = 13.6, 6.3, 6.3, 6.3 Hz, 1H), 1.08 (s, 9H), 0.81 (s, 9H), -0.05 (s, 6H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 191.4, 163.4, 136.0, 136.0, 133.6, 133.4, 130.1, 130.0, 127.8, 127.8, 126.6, 75.3, 59.0, 39.0, 27.2, 26.0, 19.5, 18.3, 13.2, -5.3, -5.3.

IR (film): ν 2937, 2889, 2860, 1676, 1467, 1433, 1386, 1254, 1103, 942, 831, 76, 738, 101, 612, 505.

HRMS (ESI): calculated for $C_{29}H_{44}O_3Si_2$ $[M+Na]^+$ 519.2721, found 519.2728.





504

(*S*)-4-benzyl-3-((2*S*,3*R*,6*R*,*E*)-2-(2-(benzyloxy)ethyl)-8-((*tert*-butyldimethylsilyl)oxy)-6-((*tert*-butyldiphenylsilyl)oxy)-3-hydroxy-5-methyloct-4-enoyl)oxazolidin-2-one (504). To a solution of Evans-auxiliary derivate **240** (0.50 g, 1.40 mmol) in CH_2Cl_2 (20 mL) was added *n*-Bu2BOTf (1.54 mL, 1M in CH_2Cl_2, 1.54 mmol) and DIPEA (0.29 mL, 1.68 mmol)

dropwise at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 0.5 h. The reaction mixture was then cooled to -78 °C. Aldehyde **498** (0.84 g, 1.68 mmol) in CH_2Cl_2 (5 mL) was added dropwise the reaction mixture was stirred for 1 h at -78 °C. The reaction was then cautiously quenched by addition of pH 7 buffer (5 mL), MeOH (25 mL) and 30% aqueous H_2O_2 (5 mL). The resulting mixture was stirred at 0 °C for 1 h, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 8:1 to 5:1) to yield 1.07 g (90%) of aldol product **504** as a single isomer and colourless oil.

TLC: R_f 0.32 (hexane/EtOAc 5:1, UV, CPS).

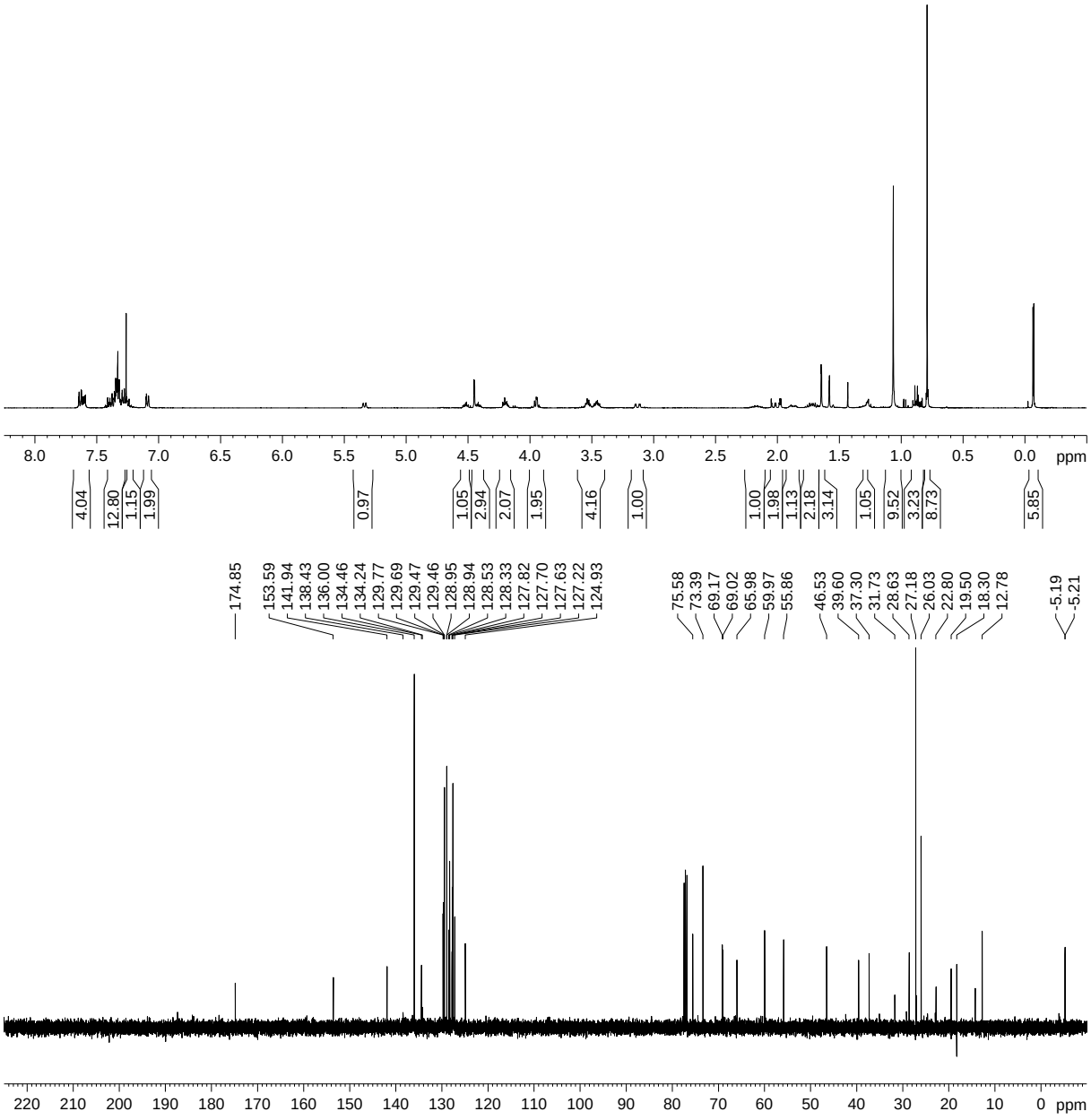
$[\alpha]^{20}_D$: = 29.6° (*c* = 0.68, $CHCl_3$).

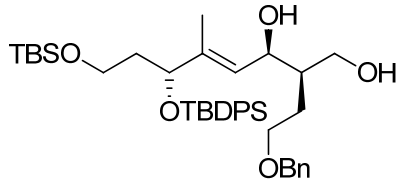
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.66-7.57 (m, 4H), 7.44-7.18 (m, 14H), 7.09 (ddd, *J* = 6.6, 1.8, 1.7 Hz, 2H), 5.34 (dp, *J* = 8.7, 1.0 Hz, 1H), 4.51 (dd, *J* = 5.7, 5.7, 2.7 Hz, 1H), 4.45 (d, *J*°= 1.8 Hz, 2H), 4.42 (ddd, *J* = 7.1, 3.6, 3.2 Hz, 1H), 4.21 (dd, *J* = 6.0, 3.3 Hz, 1H), 4.18 (dd, *J* = 6.2, 3.2 Hz, 1H), 3.95 (dd, *J* = 14.4, 9.1 Hz, 1H), 3.93 (dd, *J* = 9.1, 3.4 Hz, 1H), 3.56-3.50 (m, 2H), 3.49-3.42 (m, 2H), 3.13 (dd, *J* = 13.4, 3.1 Hz, 1H), 2.17 (dddd, *J* = 14.8, 9.8, 8.1, 5.7° Hz, 1H), 2.02 (dd, *J* = 13.4, 10.5 Hz, 1H), 1.96 (d, *J* = 3.3 Hz, 1H), 1.87 (dddd, *J* = 14.8, 4.4, 4.3, 3.3 Hz, 1H), 1.77-1.65 (m, 2H), 1.65 (d, *J* = 1.0 Hz, 3H), 1.34-1.25 (m, 2H), 1.06 (s, 9H), 0.91-0.82 (m, 3H), 0.79 (s, 9H), -0.06 (s, 3H), -0.07 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 174.8, 153.6, 141.9, 138.4, 136.0, 134.5, 134.2, 129.8, 129.7, 129.5, 129.5, 128.9, 128.9, 128.5, 128.3, 127.8, 127.7, 127.6, 127.2, 124.9, 75.6, 73.4, 69.2, 69.0, 66.0, 60.0, 55.9, 46.5, 39.6, 37.3, 31.7, 28.6, 27.2, 26.0, 22.8, 19.5, 18.3, 12.8, -5.2, -5.2.

IR (film): ν 3496, 2936, 2860, 1781, 1694, 1464, 1426, 1386, 1249, 1203, 1102, 1017, 834, 773, 741, 702, 613, 507.

HRMS (ESI): calculated for $C_{50}H_{67}O_7Si_2$ $[M+Na]^+$ 872.4348, found 872.4337.





505

(2*R*,3*R*,6*R*,*E*)-2-(2-(benzyloxy)ethyl)-8-((*tert*-butyldimethylsilyl)oxy)-6-((*tert*-butyldiphenylsilyl)oxy)-5-methyloct-4-ene-1,3-diol (505). To a solution of amide **504** (1.04 g, 1.22 mmol) in THF (12 mL) was added MeOH (0.15 mL, 3.67 mmol) and $LiBH_4$ (80 mg, 3.67 mmol) in one portion at 0 °C. The solution was stirred for 2 h at this temperature and the reaction was then cautiously quenched by addition of 15% NaOH aqueous solution (10 mL)

and the aqueous phase was extracted with EtOAc (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to afford 0.62 g (76%) of diol **505** as a colorless oil.

TLC: R_f 0.34 (hexane/EtOAc 3:1, UV, CPS).

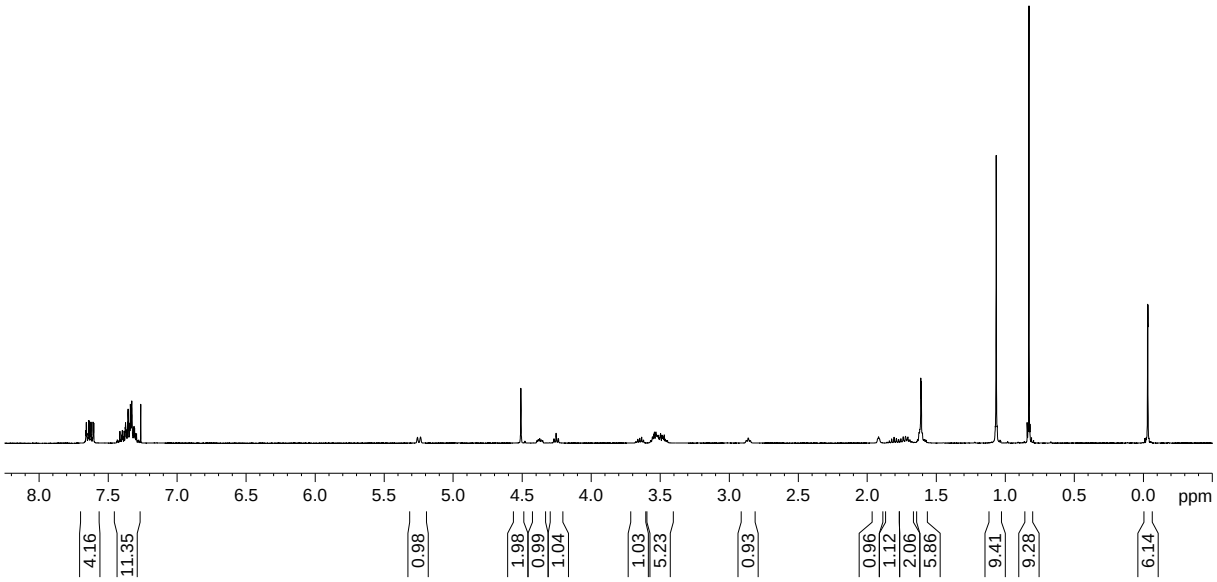
$[\alpha]^{20}_D$: = 17.5 (*c* = 1.80, $CHCl_3$).

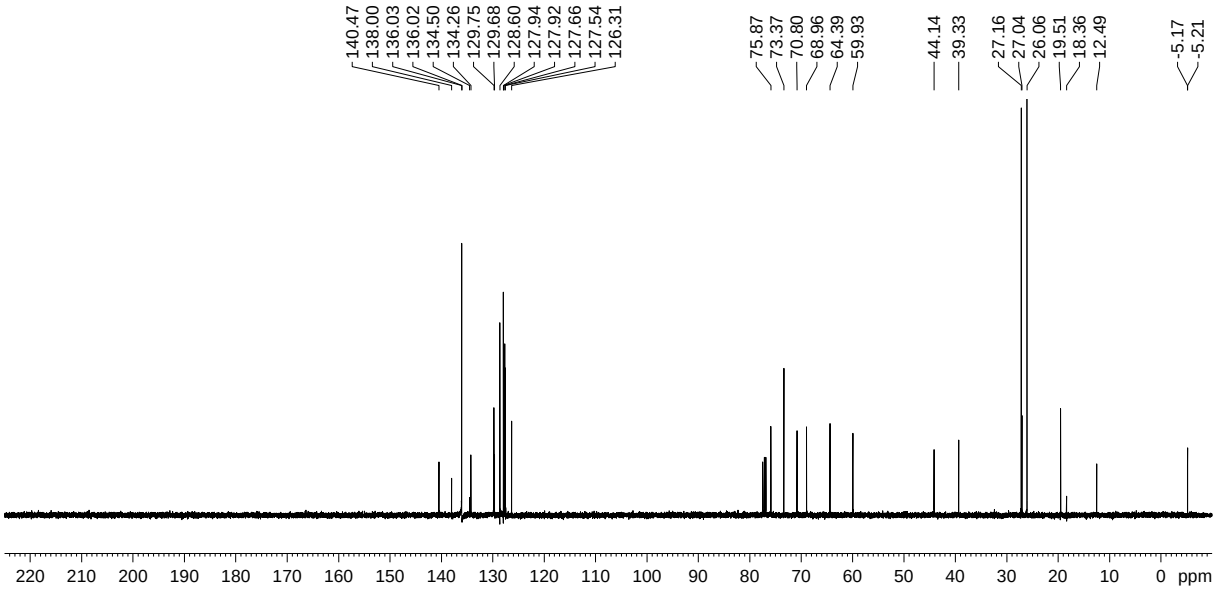
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.65 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.62 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.44-7.26 (m, 11H), 5.25 (dp, *J* = 8.9, 1.2 Hz, 1H), 4.51 (s, 2H), 4.37 (ddd, *J* = 8.4, 4.7, 3.5 Hz, 1H), 4.25 (t, *J* = 6.4 Hz, 1H), 3.65 (ddd, *J* = 11.1, 5.4, 5.3 Hz, 1H), 3.58-3.45 (m, 5H), 2.86 (t, *J* = 5.7 Hz, 1H), 2.27 (d, *J* = 3.6 Hz, 1H), 1.92 (t, *J* = 2.6 Hz, 1H), 1.82 (ddt, *J* = 13.4, 6.7, 6.5 Hz, 1H), 1.76-1.67 (m, 2H), 1.61 (d, *J* = 1.2 Hz, 3H), 1.59 (ddd, *J* = 13.7, 7.0, 4.9 Hz, 1H), 1.07 (s, 9H), 0.83 (s, 9H), - 0.03 (s, 3H), -0.03 (s, 3H).

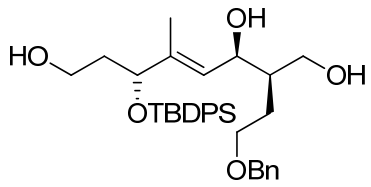
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 140.5, 138.0, 136.0, 136.0, 134.5, 134.3, 129.8, 129.7, 128.6, 127.9, 127.9, 127.7, 127.5, 126.3, 75.9, 73.4, 70.8, 69.0, 64.4, 59.9, 44.1, 39.3, 27.2, 27.0, 26.1, 19.5, 18.4, 12.5, -5.2, -5.2.

IR (film): ν 3384, 2952, 2929, 2886, 2857, 1468, 1428, 1389, 1362, 1254, 1105, 1076, 1003, 942, 833, 776, 739, 701, 610, 552, 506.

HRMS (ESI): calculated for $C_{40}H_{60}O_5Si_2$ $[M+Na]^+$ 699.3871, found 699.3868.







541

(2*R*,3*R*,6*R*,*E*)-2-(2-(benzyloxy)ethyl)-6-((*tert*-butyldiphenylsilyl)oxy)-5-methyloct-4-ene-1,3,8-triol (541). To a solution of silyl ether **505** (21 mg, 0.03 mmol) in THF (0.80 mL) and water (0.24 mL) was added AcOH (0.80 mL) at room temperature and the reaction mixture was stirred for 24 h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (8 mL), the phases were separated and the aqueous phase was extracted with EtOAc (3 x 10 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:4) to yield 14 mg (81%) of triol **541** as a colourless oil.

TLC: R_f 0.10 (hexane/EtOAc 1:4, UV, CPS).

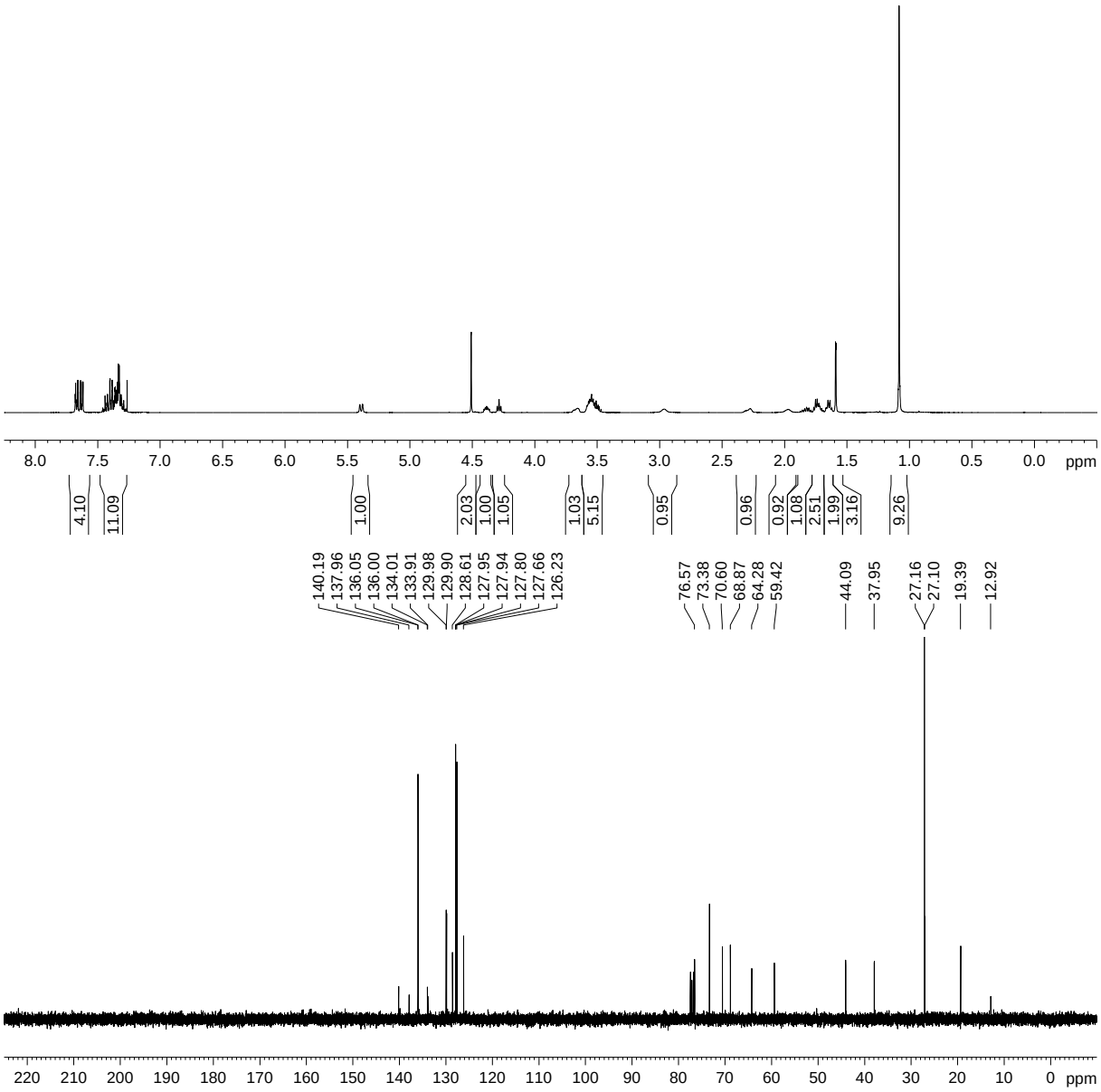
$[\alpha]^{20}_D$: = 46.8 (*c* = 1.67, $CHCl_3$).

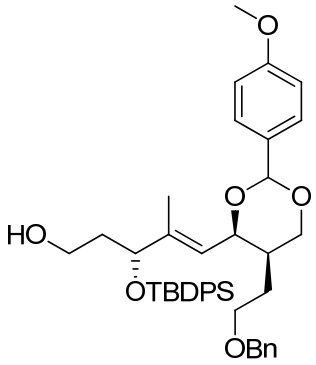
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.67 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.63 (ddd, *J* = 6.4, 1.5, 1.4 Hz, 2H), 7.47-7.27 (m, 11H), 5.39 (dp, *J* = 8.8, 1.0 Hz, 1H), 4.51 (s, 2H), 4.39 (ddd, *J* = 8.9, 4.5, 3.6 Hz, 1H), 4.29 (t, *J* = 5.7 Hz, 1H), 3.71-3.64 (m, 1H), 3.60-3.46 (m, 5H), 2.97 (br s, 1H), 2.34-2.24 (m, 1H), 1.97 (br s, 1H), 1.83 (ddt, *J* = 14.3, 7.1, 5.8 Hz, 1H), 1.78-1.68 (m, 2H), 1.68-1.62 (m, 2H), 1.59 (d, *J* = 1.0 Hz, 3H), 1.08 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 140.2, 138.0, 136.0, 136.0, 134.0, 133.9, 130.0, 129.9, 128.6, 128.0, 127.9, 127.8, 127.7, 126.2, 76.6, 73.4, 70.6, 68.9, 64.3, 59.4, 44.1, 37.9, 27.2, 27.1, 19.4, 12.9.

IR (film): ν 3369, 2931, 2885, 2858, 1469, 1455, 1427, 1389, 1362, 1107, 1068, 1004, 822, 741, 700, 669, 610, 552, 505.

HRMS (ESI): calculated for $C_{34}H_{46}O_5Si$ $[M+Na]^+$ 585.3007, found 585.3002.





542

(3*R*,*E*)-5-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-3-((*tert*-butyldiphenylsilyl)oxy)-4-methylpent-4-en-1-ol (542). To a solution of triol **541** (3.55 g, 6.31 mmol) in CH_2Cl_2 (65 mL) was added *para*-methoxybenzaldehyde dimethyl acetale (1.31 mL, 7.58 mmol) and CSA (7 mg, 0.03 mmol) at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 6 h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (30 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 30 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 4:1) gave 3.64 g (85%) of an inseparable 8:1 mixture of acetale isomers **542**.

Note: The spectroscopic data are given for the major acetale isomer.

TLC: R_f 0.14 (hexane/EtOAc 4:1, UV, CPS).

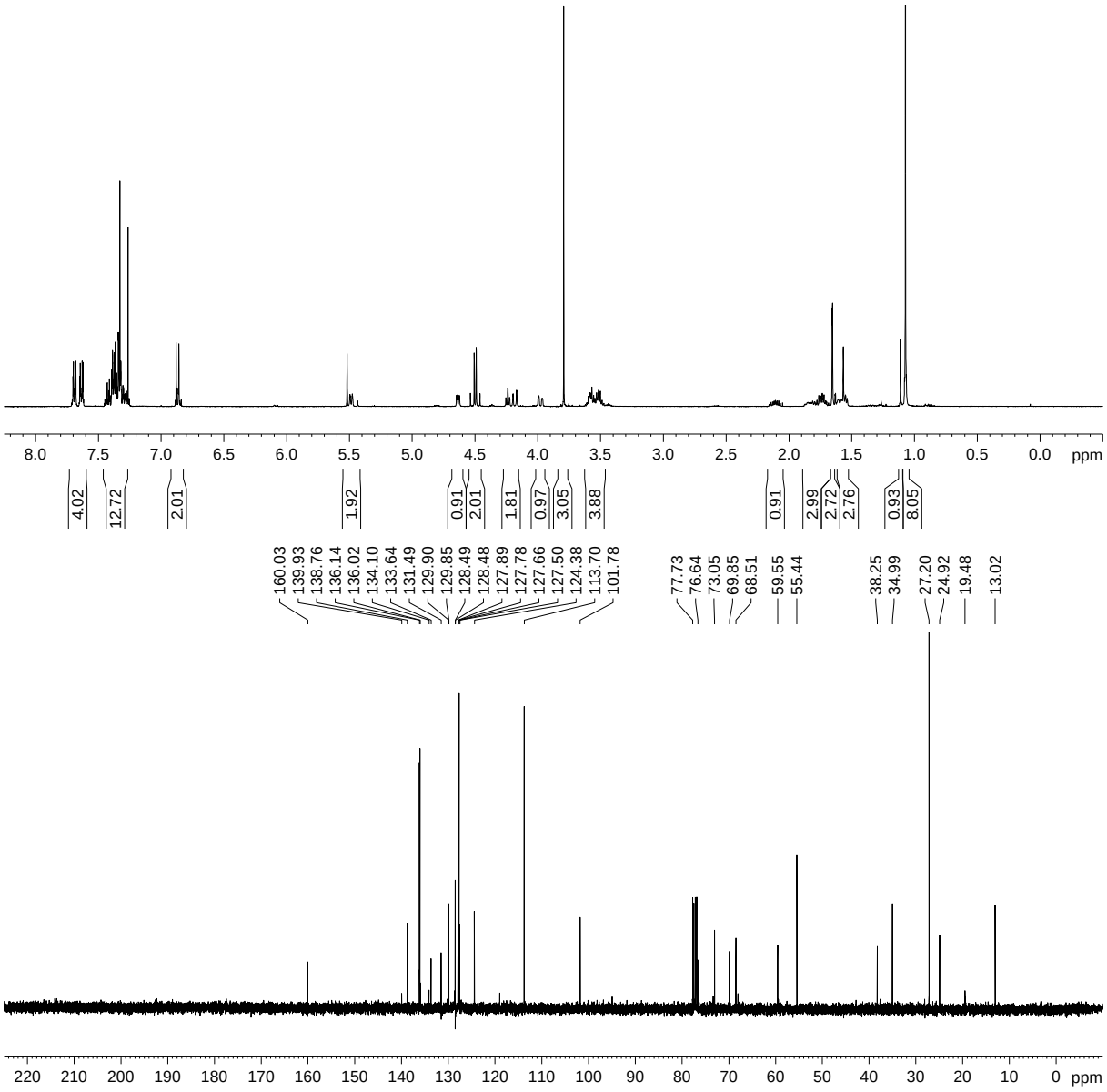
$[\alpha]^{20}_D$: = 55.1 (*c* = 0.60, $CHCl_3$).

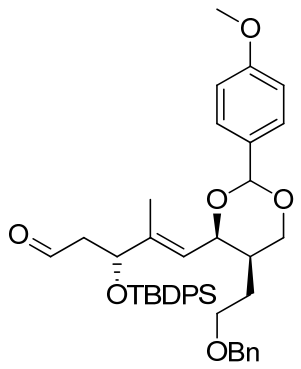
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.69 (ddd, *J* = 6.4, 1.4, 1.3 Hz, 2H), 7.63 (ddd, *J* = 6.4, 1.4, 1.3 Hz, 2H), 7.45-7.27 (m, 13H), 6.87 (ddd, *J* = 8.9, 2.4, 2.2 Hz, 2H), 5.52 (s, 1H), 5.49 (dp, *J* = 7.3, 1.2 Hz, 1H), 4.64 (dd, *J* = 7.3, 2.4 Hz, 1H), 4.52 (d, *J* = 12.0 Hz, 1H), 4.48 (d, *J* = 12.0 Hz, 1H), 4.24 (t, *J* = 5.8 Hz, 1H), 4.18 (dd, *J* = 11.5, 1.2 Hz, 1H), 3.98 (ddd, *J* = 11.5, 2.4, 1.0 Hz, 1H), 3.79 (s, 3H), 3.63-3.46 (m, 4H), 2.11 (dddd, *J* = 14.5, 10.4, 5.3, 5.2 Hz, 1H), 1.84 (dddd, *J* = 14.5, 9.1, 7.5, 3.4 Hz, 1H), 1.78-1.67 (m, 2H), 1.65 (d, *J* = 1.2 Hz, 3H), 1.62-1.56 (m, 1H), 1.56-1.52 (m, 1H), 1.07 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 160.0, 139.9, 138.8, 136.1, 136.0, 134.1, 133.6, 131.5, 129.9, 129.8, 128.5, 128.5, 127.9, 127.8, 127.7, 127.5, 124.4, 113.7, 101.8, 77.7, 76.6, 73.0, 69.8, 68.5, 59.5, 55.4, 38.3, 35.0, 27.2, 24.9, 19.5, 13.0.

IR (film): ν 2956, 2931, 2890, 2857, 1724, 1615, 1589, 1517, 1470, 1463, 1456, 1428, 1390, 1364, 1303, 12448, 1213, 1172, 1145, 1105, 1054, 1032, 998, 936, 825, 740, 701, 612, 552, 504, 487.

HRMS (ESI): calculated for $C_{42}H_{52}O_6Si$ $[M+K]^+$ 719.3165, found 719.3165.





543

(3*R*,*E*)-5-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-3-((*tert*-butyldiphenylsilyl)oxy)-4-methylpent-4-enal (543). To a solution of alcohol **542** (3.57 g, 5.24 mmol) in CH_2Cl_2 (65 mL) was added BAIB (3.37 g, 10.49 mmol) and TEMPO (164 mg,

1.05 mmol) at 0 °C. The reaction mixture was slowly allowed to reach room temperature and stirred for 8 h at this temperature. The reaction was then cautiously quenched by addition of 10% aqueous $Na_2S_2O_3$ (40 mL). Stirring was continued for 10 min, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure.The residue was purified by flash column chromatography (hexane/EtOAc 5:1) to yield 3.25 g (91%) of inseparable 4.5:1 mixture of acetale isomers of aldehyde **543** as a colorless oil.

Note: The spectroscopic data are given for the major acetale isomer.

TLC: R_f 0.22 (hexane/EtOAc 5:1, UV, CPS).

$[\alpha]^{20}_D$: = 45.6 (*c* = 3.93, $CHCl_3$).

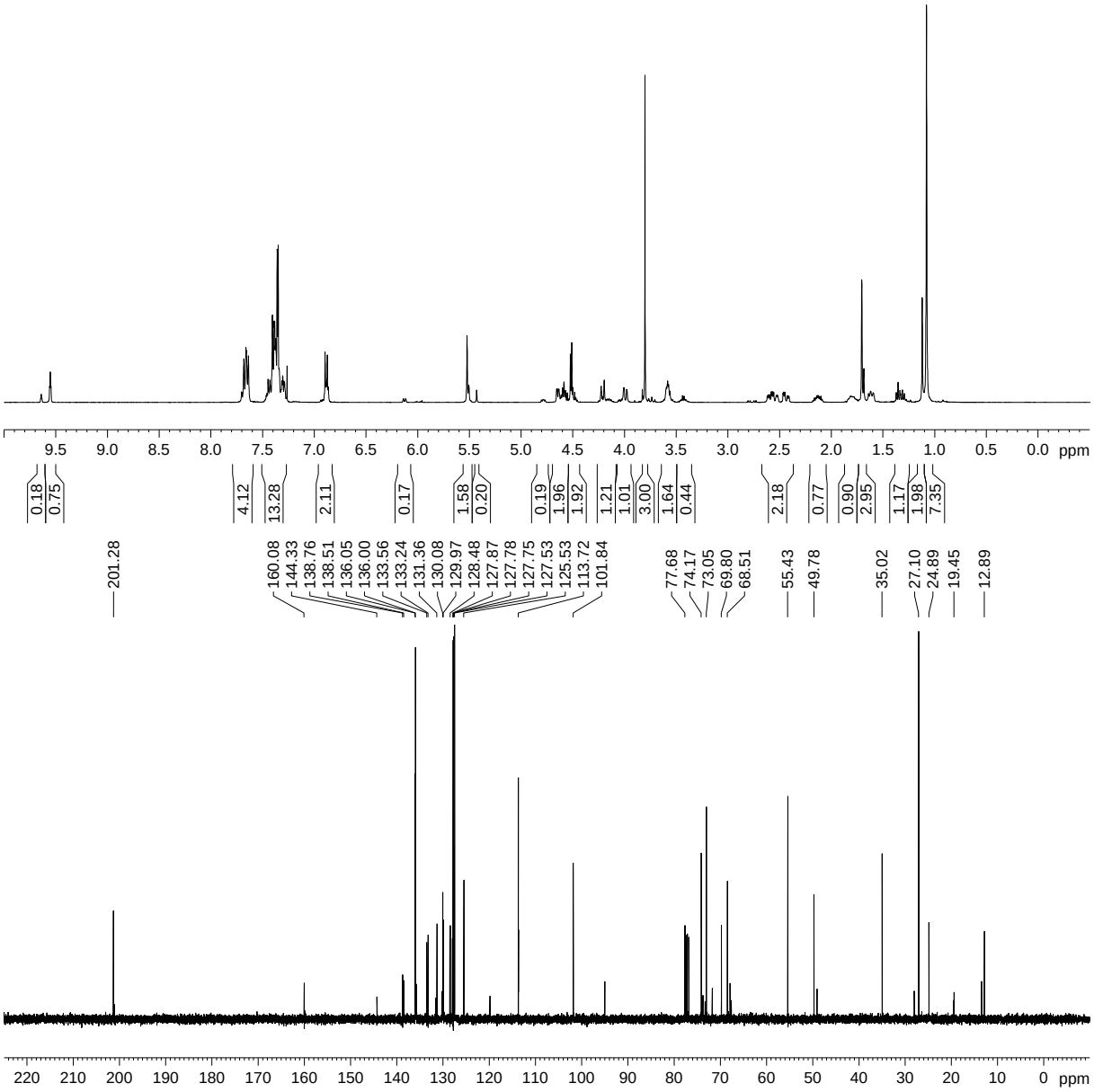
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.54 (t, *J* = 2.7 Hz, 1H), 7.65 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.63 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.46-7.27 (m, 13H), 6.87 (ddd, *J* = 8.8, 2.4, 2.2 Hz, 2H), 5.51 (s, 1H), 5.49 (dp, *J* = 7.3, 1.2 Hz, 1H), 4.63 (dd, *J* = 7.3, 2.4 Hz, 1H), 4.57 (t, *J* = 5.8 Hz, 1H), 4.53 (d, *J* = 12.0 Hz, 1H), 4.49 (t, *J* = 12.0 Hz, 1H), 4.20 (dd, *J* = 11.6, 1.2 Hz, 1H), 3.98 (ddd, *J* = 11.6, 2.1, 1.0 Hz, 1H), 3.80 (s, 3H), 3.57 (dd, *J* = 5.1, 2.3 Hz, 1H), 3.55 (dd, *J* = 5.8, 4.3 Hz, 1H), 2.57 (ddd, *J* = 15.7, 6.6, 3.1 Hz, 1H), 2.42 (ddd, *J* = 15.7, 5.4, 2.4 Hz, 1H), 2.12 (dddd, *J* = 14.5, 10.5 5.3, 5.2 Hz, 1H), 1.78 (dddd, *J* = 14.5, 7.4, 7.3, 3.3 Hz, 1H), 1.69 (d, *J* = 1.2 Hz, 3H), 1.63-1.58 (m, 1H), 1.06 (s, 9H).

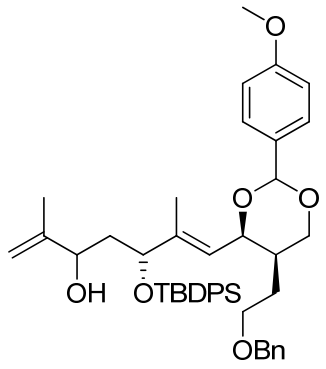
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 201.3, 160.1, 144.3, 138.8, 138.5, 136.1, 136.0, 133.6, 133.2, 131.4, 130.1, 130.0, 128.5, 127.9, 127.8, 127.7, 127.5, 125.5, 113.7, 101.8, 77.7, 74.2, 73.1, 69.8, 68.5, 55.4, 49.8, 35.0, 27.1, 24.9, 19.4, 12.9.

IR (film): ν 3444, 2953, 2931, 2886, 2857, 1615, 1589, 1517, 1470, 1463, 1428, 1389, 1364, 1303, 1249, 1172, 1144, 1106, 1075, 1052, 1034, 1009, 935, 825, 738, 102, 612.

HRMS (ESI): calculated for $C_{42}H_{52}O_6Si$ $[M+Na]^+$ 701.3274, not found.

LRMS (ESI): calculated for $C_{42}H_{52}O_6Si$ $[M+Na]^+$ 701.3274, found 701.3.





544

(5*R*,*E*)-7-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-5-((*tert*-butyldiphenylsilyl)oxy)-2,6-dimethylhepta-1,6-dien-3-ol (544).

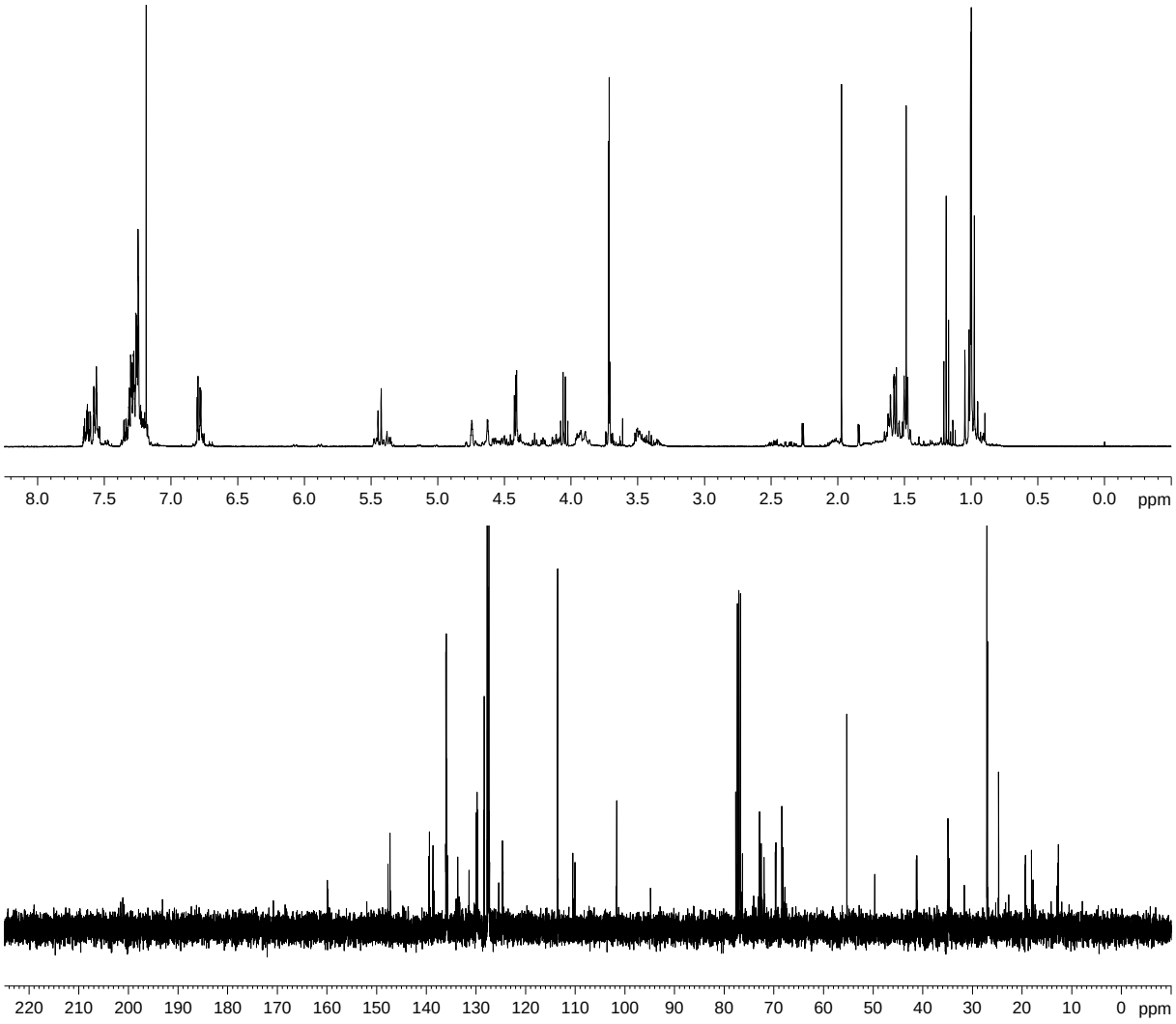
To a solution of aldehyde **543** (3.50 g, 5.16 mmol) in THF (50 mL) was added freshly prepared isopropenylmagnesium bromide (1 M in THF, 6.96 mL, 6.96 mmol) dropwise at −78 °C and the reaction was stirred for 0.5 h at this temperature. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (50 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 4:1) gave 3.24 g (94%) of a mixture of four diastereomers **544**.

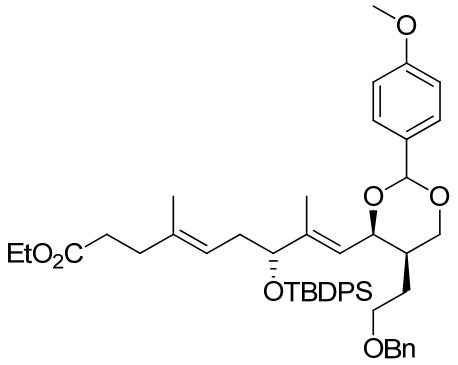
Note: The spectroscopic data are not given because of the complex sets of signals.

TLC: R_f 0.26-0.16 (hexane/EtOAc 5:1, UV, CPS).

IR (film): ν 3497, 2931, 2856, 1615, 1589, 1517, 1455, 1428, 1389, 1365, 1303, 1248, 1172, 1144, 1105, 1052, 1033, 1008, 932, 901, 824, 740, 701, 612, 542, 507.

HRMS (ESI): calculated for $C_{45}H_{56}O_6Si$ $[M+K]^+$ 759.3478, found 759.3467.





545

(4*E*,7*R*,8*E*)-ethyl 9-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-7-((*tert*-butyldiphenylsilyl)oxy)-4,8-dimethylnona-4,8-dienoate (545). To a solution of allylic alcohol **544** (2.80 g, 3.88 mmol) in toluene (40 mL) was added triethyl orthoacetate (3.37 g, 10.49 mmol) and propionic acid (5.8 uL, 0.08 mmol) at room temperature. The reaction mixture was then heated to reflux for 24 h. The reaction was then allowed to reach 40 °C and the reaction mixture was concentrated under reduced pressure. The residue was dissolved in ether (50 mL) and the organic phase was washed with saturated aqueous $NaHCO_3$ (30 mL). The organic phases was dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (hexane/EtOAc 7:1) to yield 2.61 g (85%) of a separable 4.5:1 mixture of acetale isomers of ester **545** as a colorless oil.

Note: The spectroscopic data are given for the major acetale isomer.

TLC: R_f 0.29, 0.24 (hexane/EtOAc 5:1, UV, CPS).

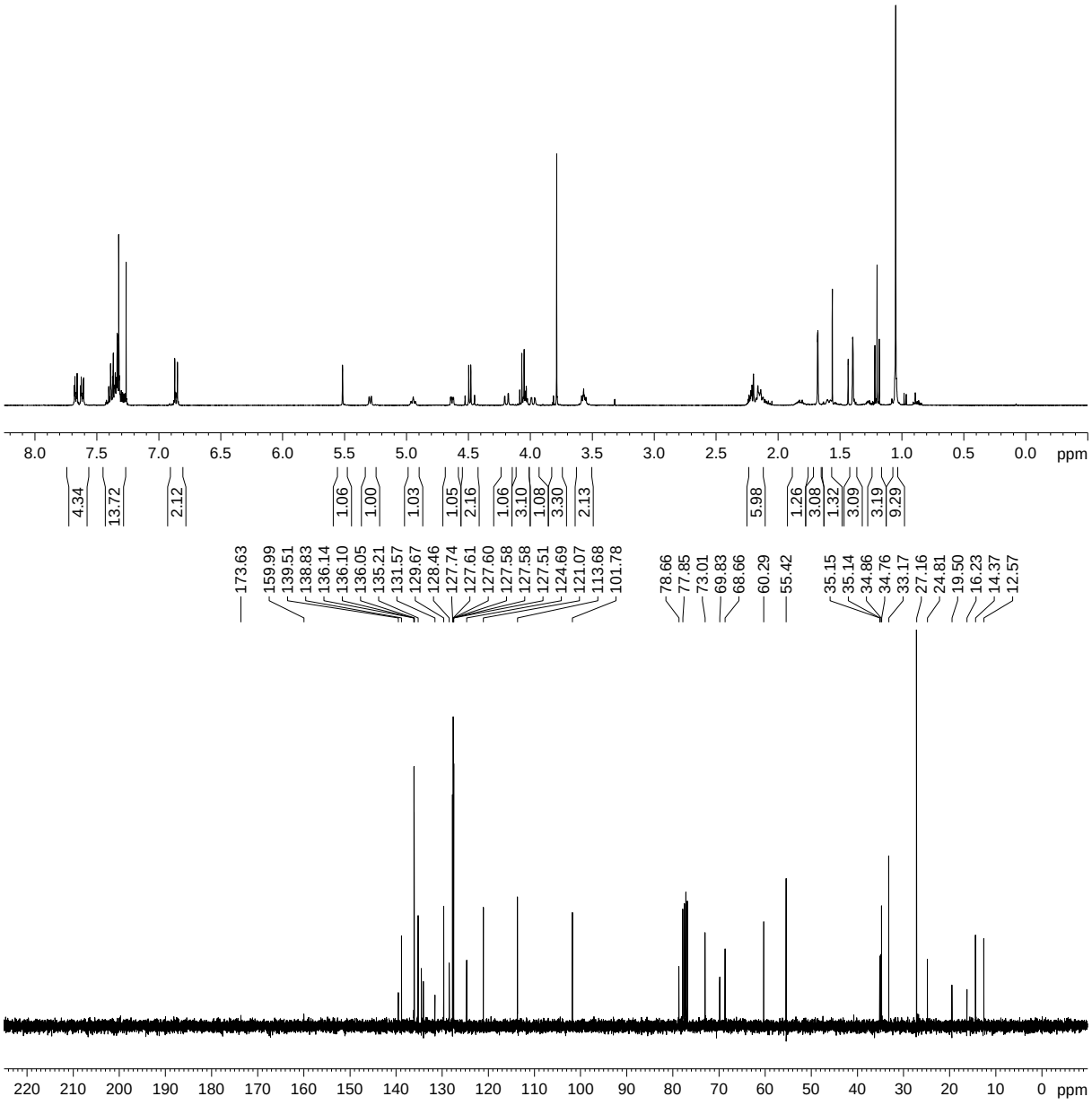
$[\alpha]^{20}_D$: = 22.1 (*c* = 0.67, $CHCl_3$).

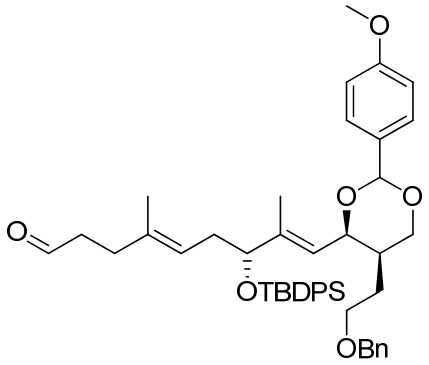
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.67 (ddd, *J* = 6.5, 1.4, 1.4 Hz, 2H), 7.62 (ddd, *J* = 6.5, 1.3, 1.2 Hz, 2H), 7.43-7.27 (m, 13H), 6.86 (ddd, *J* = 8.8, 2.4, 2.0 Hz, 2H), 5.52 (s, 1H), 5.29 (dp, *J* = 7.0, 1.2 Hz, 1H), 4.95 (tq, *J* = 7.3, 1.2 Hz, 1H), 4.63 (dd, *J* = 7.1, 2.4 Hz, 1H), 4.51 (d, *J* = 11.9 Hz, 1H), 4.47 (t, *J* =11.9 Hz, 1H), 4.19 (dd, *J* = 11.6, 1.2 Hz, 1H), 4.06 (q, *J* = 7.1 Hz, 2H), 4.05-4.02 (m, 1H), 3.98 (dd, *J* = 11.6, 1.6 Hz, 1H), 3.79 (s, 3H), 3.58 (dd, *J* = 4.7, 0.9 Hz, 1H), 3.56 (dd, *J* = 5.8, 2.7 Hz, 1H), 2.57 (ddd, *J* = 15.7, 6.6, 3.1 Hz, 1H), 2.25-2.07 (m, 6H), 1.87-1.78 (m, 1H), 1.68 (d, *J* = 1.2 Hz, 3H), 1.63-1.57 (m, 1H), 1.40 (d, *J* = 1.2 Hz, 3H), 1.20 (t, *J* = 7.1 Hz, 3H), 1.05 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 173.6, 160.0, 139.5, 138.8, 136.1, 136.1, 136.0, 135.2, 131.6, 129.7, 128.5, 127.7, 127.6, 127.6, 127.6, 127.6, 127.5, 124.7, 121.1, 113.7, 101.8, 78.7, 77.9, 73.0, 69.8, 68.7, 60.3, 55.4, 35.1, 35.1, 34.9, 34.8, 33.2, 27.2, 24.8, 19.5, 16.2, 14.4, 12.6.

IR (film): ν 3070, 3031, 2955, 2931, 2889, 2856, 1733, 1616, 1589, 1517, 1455, 1444, 1428, 1389, 1367, 1302, 1248, 1213, 1171, 1147, 1107, 1074, 1052, 1036, 1009, 983, 939, 824 ,741, 702, 612, 547, 530, 503.

HRMS (ESI): calculated for $C_{49}H_{62}O_7Si$ $[M+K]^+$ 829.3896, found 829.3893.





546

(4*E*,7*R*,8*E*)-9-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-7-((*tert*-butyldiphenylsilyl)oxy)-4,8-dimethylnona-4,8-dienal (546). To a solution of ester **545** (2.45 g, 1.58 mmol) in CH_2Cl_2 (50 mL) was added DIBAL-H (1M in hexane, 1.58 mL, 1.58 mmol) dropwise at -78 °C. The reaction mixture was stirred for 30 min at this temperature and the reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (80 mL). The solution was allowed to warm to room temperature and was left stirring vigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 8:1) to give 2.12 g (90%) of a separable 4.5:1 mixture of acetale isomers of aldehyde **546** as a colorless oil.

Note: The spectroscopic data are given for the major acetale isomer.

TLC: R_f 0.39, 0.29 (hexane/EtOAc 5:1, UV, CPS).

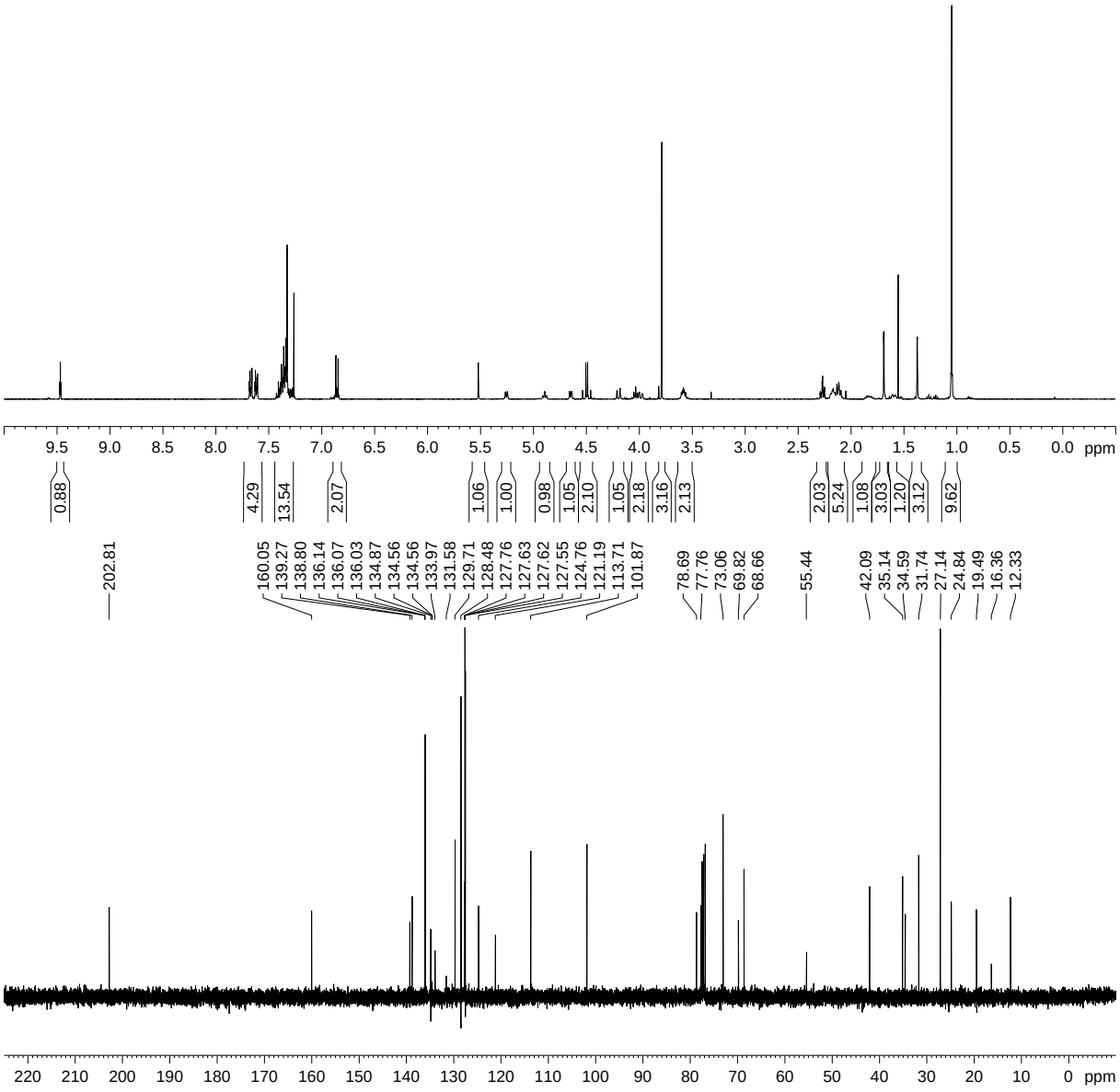
$[\alpha]^{20}_D$: = 25.6 (*c* = 0.57, $CHCl_3$).

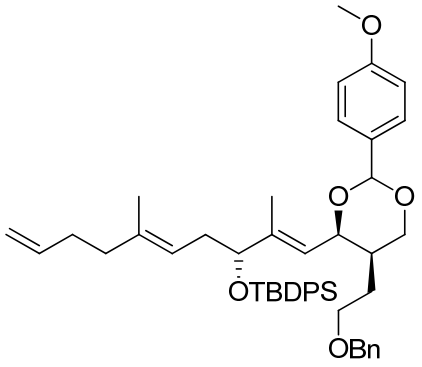
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.47 (t, *J* = 1.7 Hz, 1H), 7.67 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.62 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.44-7.27 (m, 13H), 6.86 (ddd, *J* = 8.8, 2.8, 2.0 Hz, 2H), 5.52 (s, 1H), 5.25 (dp, *J* = 7.0, 1.2 Hz, 1H), 4.89 (tq, *J* = 7.3, 1.2 Hz, 1H), 4.65 (dd, *J* = 7.4, 2.5 Hz, 1H), 4.52 (d, *J* = 11.9 Hz, 1H), 4.47 (t, *J* =11.9 Hz, 1H), 4.20 (dd, *J* = 11.7, 1.2 Hz, 1H), 4.03 (t, *J* = 6.8 Hz, 1H), 3.98 (dd, *J* = 11.9, 1.6 Hz, 1H), 3.79 (s, 3H), 3.59 (dd, *J* = 5.1, 2.3 Hz, 1H), 3.57 (dd, *J* = 5.9, 3.9 Hz, 1H), 2.27 (ddd, *J* = 15.7, 1.7, 1.1 Hz, 1H), 2.27 (t, *J* = 1.7 Hz, 1H), 2.20-2.07 (m, 5H), 1.83 (dddd, *J* = 14.6, 7.7, 6.8, 3.8 Hz, 1H), 1.69 (d, *J* = 1.2 Hz, 3H), 1.63-1.57 (m, 1H), 1.38 (d, *J* = 1.2 Hz, 3H), 1.05 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 202.8, 160.1, 139.3, 138.8, 136.1, 136.1, 136.0, 134.9, 134.6, 134.6, 134.0, 131.6, 129.7, 128.5, 127.8, 127.6, 127.6, 127.5, 124.8, 121.2, 113.7, 101.9, 78.7, 77.8, 73.1, 69.8, 68.7, 55.4, 42.1, 35.1, 34.6, 31.7, 27.1, 24.8, 19.5, 16.4.

IR (film): ν 3070, 3030, 2998, 2955, 2931, 2890, 2856, 1723, 1616, 1289, 1518, 1488, 1471, 1456, 1426, 1389, 1365, 1340, 1303, 1249, 1214, 1172, 1145, 1107, 1074, 1052, 1035, 1009, 982, 939, 824, 740, 702, 622, 612, 548, 507.

HRMS (ESI): calculated for $C_{47}H_{58}O_6Si$ $[M+K]^+$ 785.3634, found 785.3653.





547

(((1*E*,3*R*,5*E*)-1-((4*R*,5*R*)-5-(2-(benzyloxy)ethyl)-2-(4-methoxyphenyl)-1,3-dioxan-4-yl)-2,6-dimethyldeca-1,5,9-trien-3-yl)oxy)(*tert*-butyl)diphenylsilane (547). To a supension of methyltrimethylphosphonium bromide (1.01 g, 2.85 mmol) in THF (35 mL) was added *n*-BuLi (1M in hexane, 2.58 mL, 2.58 mmol) dropwise at 0 °C and the reaction mixture was stirred for 30 min at this temperature. Aldehyde **546** (1.77 g, 2.37 mmol) in THF (5 mL) was then added dropwise and the reaction mixture was stirred for 3 h at 0 °C. The reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (50 mL). The phases were separated and the aqueous phase was extracted with ether (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to give 1.59 g (90%) of a separable 4.5:1 mixture of acetale isomers of olefine **547** as a colorless oil.

Note: The spectroscopic data are given for the major acetale isomer.

TLC: R_f 0.14 (hexane/EtOAc 10:1, UV, CPS).

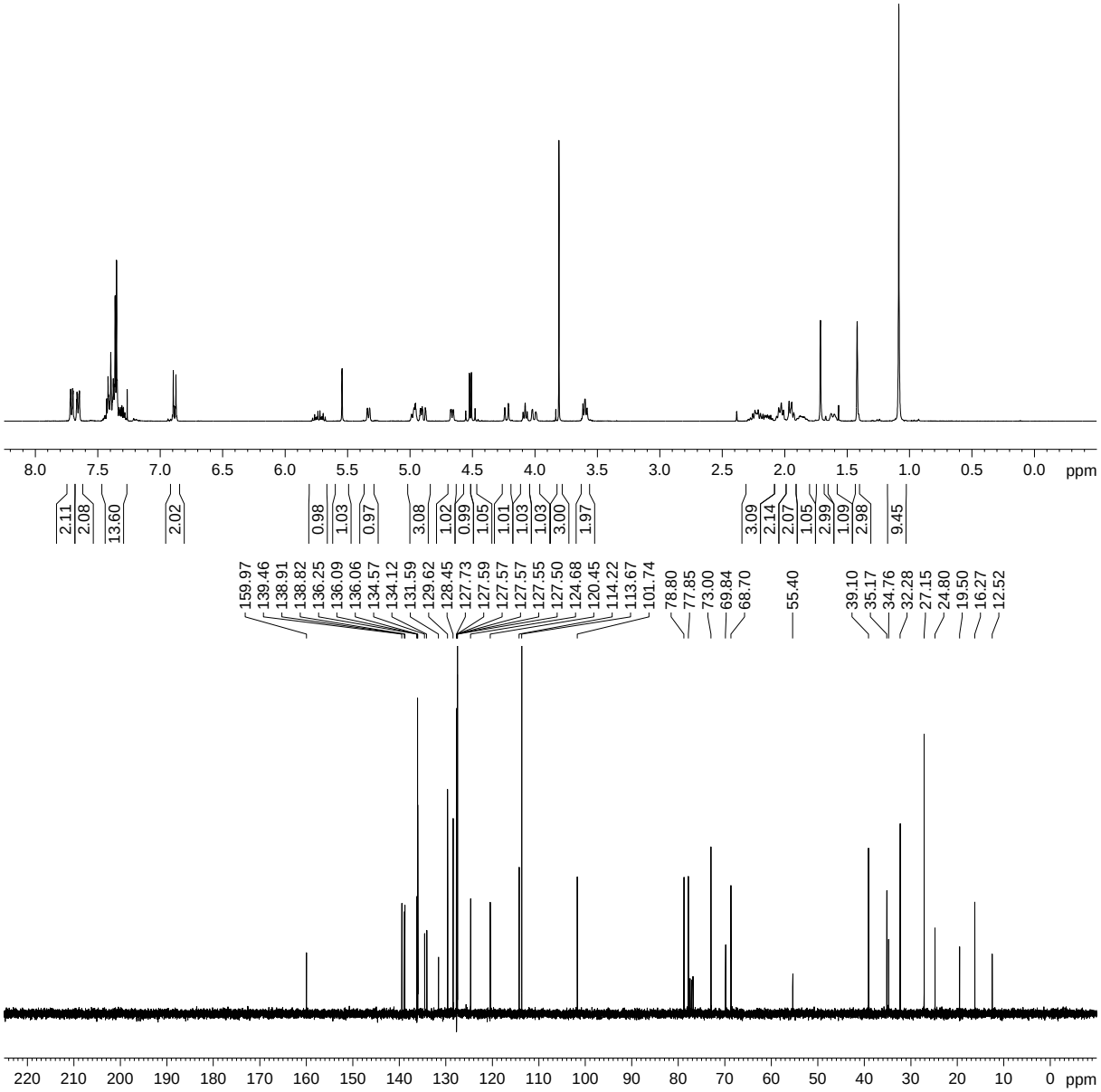
$[\alpha]^{20}_D$: = (*c* = 0.57, $CHCl_3$).

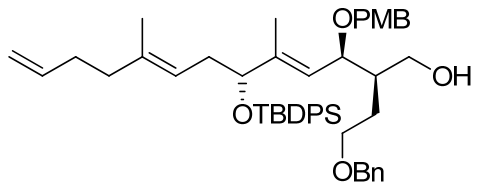
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.71 (ddd, *J* = 6.5, 1.3, 1.2 Hz, 2H), 7.66 (ddd, *J* = 6.5, 1.3, 1.2 Hz, 2H), 7.46-7.27 (m, 13H), 6.86 (ddd, *J* = 8.8, 2.8, 2.0 Hz, 2H), 5.57 (dt, *J* = 16.7, 10.3, 6.4 Hz, 2H), 5.54 (s, 1H), 5.33 (d, *J* = 7.1 Hz, 1H), 4.98 (d, *J* = 8.2 Hz, 1H), 4.94 (dq, *J* = 17.1, 1.7 Hz, 1H), 4.89 (dd, *J* = 9.9, 1.2 Hz, 1H), 4.66 (dd, *J* = 7.1, 2.1 Hz, 1H), 4.54 (d, *J* = 11.9 Hz, 1H), 4.49 (t, *J* =11.9 Hz, 1H), 4.22 (d, *J* = 11.4 Hz, 1H), 4.08 (t, *J* = 6.5 Hz, 1H), 4.00 (dd, *J* = 11.4, 1.6 Hz, 1H), 3.81 (s, 3H), 3.60 (dd, *J* = 7.4, 5.9 Hz, 1H), 2.26 (dd, *J* = 14.7, 6.8 Hz, 1H), 2.20 (dd, *J* = 14.7, 8.1 Hz, 1H), 2.16-2.09 (m, 1H), 2.07-1.99 (m, 2H), 1.98-1.91 (m, 2H), 1.86 (dddd, *J* = 14.1, 7.7, 7.5, 3.3 Hz, 1H), 1.72 (s, 3H), 1.64-1.58 (m, 1H), 1.41 (s, 3H), 1.08 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 160.0, 139.5,138.9, 138.8, 136.3, 136.1, 136.1, 134.6, 134.1, 131.6, 129.6, 128.4, 127.7, 127.6, 127.6, 127.6, 127.5, 127.5, 124.7, 120.5, 114.2, 113.7, 101.7, 78.8, 77.9, 73.0, 69.8, 68.7, 55.4, 39.1, 35.2, 34.8, 32.3, 27.2, 24.8, 19.5, 16.3, 12.5.

IR (film): ν 3070, 2955, 2930, 2856, 1616, 1589, 1518, 1471, 1455, 1428, 1389, 1364, 1303, 1249, 1172, 1145, 1107, 1074, 1054, 1036, 1009, 939, 908, 824, 740, 702, 613, 506.

HRMS (ESI): calculated for $C_{48}H_{60}O_5Si$ $[M+Na]^+$ 767.4102, found 767.4096.





548

(2*R*,3*R*,4*E*,6*R*,8*E*)-2-(2-(benzyloxy)ethyl)-6-((*tert*-butyldiphenylsilyl)oxy)-3-((4-methoxybenzyl)oxy)-5,9-dimethyltrideca-4,8,12-trien-1-ol (548). To a solution of acetale **547** (1.71 g, 2.30 mmol) in CH_2Cl_2 (35 mL) was added DIBAL-H (1.0 M in hexane, 5.98 mL,

5.98 mmol) dropwise at -78 °C and the reaction mixture was allowed to reach -70 °C. Me_2AlCl (1 M in hexane, 3.45 mL, 3.45 mmol) was added dropwise and the reaction mixture was allowed to reach -30 °C over a period of 1 h. The reaction was then cautiously quenched by addition of saturated aqueous Rochelle salt (100 mL). The solution was allowed to warm to room temperature and was left stirring rigorously until the two phases became transparent. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 7:1) to yield 1.69 g (99%) of alcohol **548** as a colorless oil.

TLC: R_f 0.38 (hexane/EtOAc 5:1, UV, CPS).

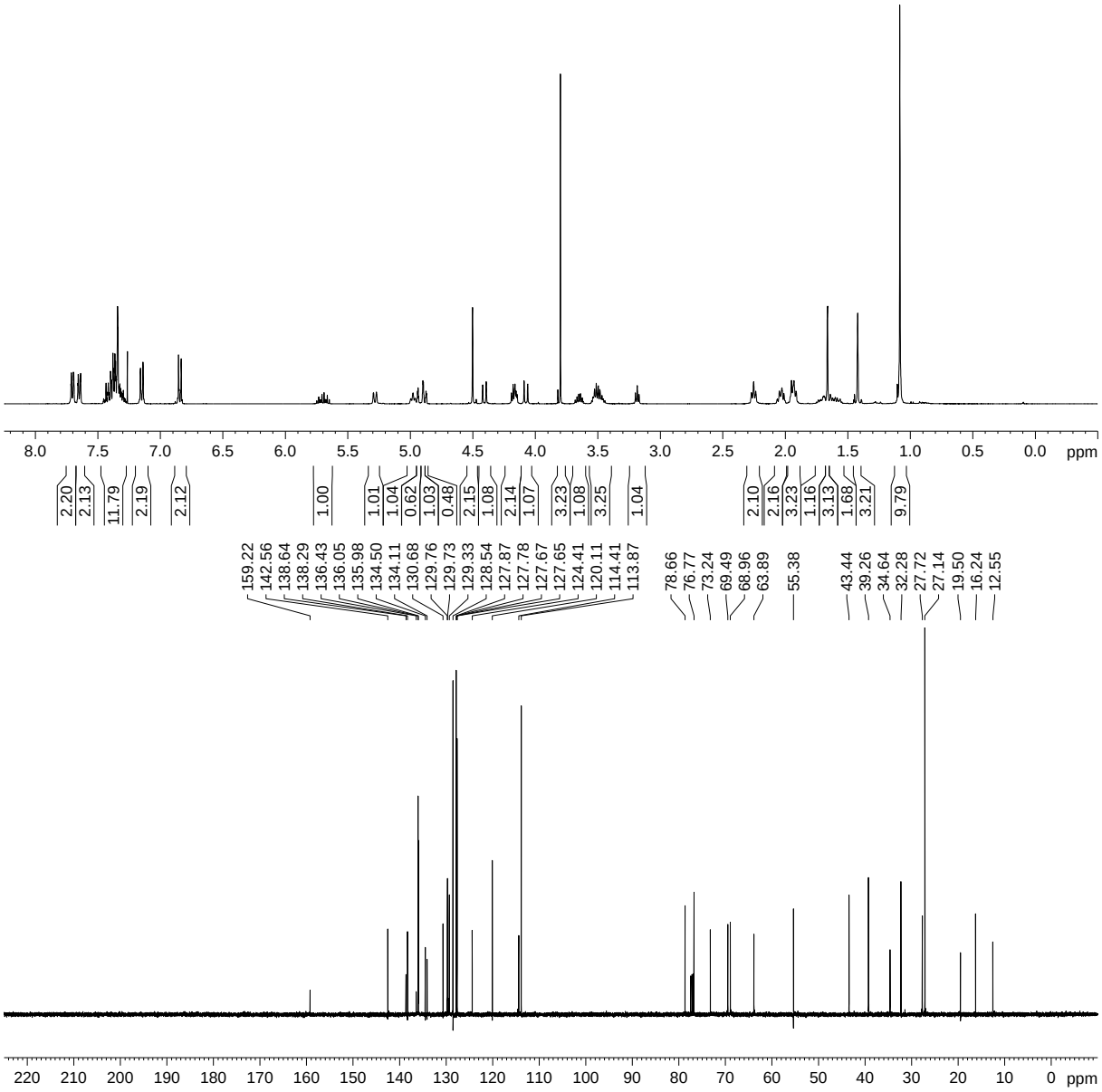
$[\alpha]^{20}_D$: = 19.7 (*c* = 0.43, $CHCl_3$).

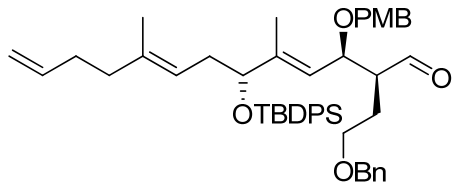
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.70 (dd, *J* = 7.2, 1.4 Hz, 2H), 7.65 (dd, *J* = 7.2, 1.4 Hz, 2H), 7.46-7.27 (m, 11H), 7.14 (d, *J* = 8.5 Hz, 2H), 6.84 (d, *J* = 8.5 Hz, 2H), 5.70 (ddt, *J* = 17.0, 10.3, 6.4 Hz, 1H), 5.28 (d, *J* = 9.7 Hz, 1H), 4.98 (dd, *J* = 7.2, 6.8 Hz, 1H), 4.91 (dq, *J* = 17.0, 1.7 Hz, 1H), 4.88 (dt, *J* = 9.6, 1.2 Hz, 1H), 4.50 (s, 2H), 4.41 (d, *J* = 11.3 Hz, 1H), 4.18 (t, *J* = 5.3 Hz, 1H), 4.16 (dd, *J* = 5.6, 3.1 Hz, 1H), 4.08 (d, *J* = 11.3 Hz, 1H), 4.00 (dd, *J* = 11.4, 1.6 Hz, 1H), 3.80 (s, 3H), 3.65 (tt, *J* = 11.2, 5.7 Hz, 1H), 3.56-3.43 (m, 3H), 3.18 (t, *J* = 6.0 Hz, 1H), 2.25 (t, *J* = 6.8 Hz, 2H), 2.07-2.00 (m, 2H), 1.97-1.90 (m, 3H), 1.75-1.55 (m, 3H), 1.42 (s, 3H), 1.08 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 159.2, 142.6, 138.6, 138.3, 136.4, 136.0, 136.0, 134.5, 134.1, 130.7, 129.8, 129.7, 129.3, 128.5, 127.9, 127.8, 127.7, 127.7, 124.4, 120.1, 114.4, 113.9, 78.7, 76.8, 73.2, 69.5, 69.0, 63.9, 55.4, 43.4, 39.3, 34.6, 32.3, 27.7, 27.1, 19.5, 16.2. 12.6

IR (film): ν 3450, 3070, 3032, 2930, 2892, 2857, 1613, 1513, 1471, 1455, 1428, 1389, 1362, 1302, 1248, 1173, 1111, 1061, 1037, 1008, 999, 940, 912, 822, 740, 702, 611, 583, 570, 558, 542, 506.

HRMS (ESI): calculated for $C_{48}H_{60}O_5Si$ $[M+Na]^+$ 767.4102, found 767.4113.





549

(2*S*,3*R*,4*E*,6*R*,8*E*)-2-(2-(benzyloxy)ethyl)-6-((*tert*-butyldiphenylsilyl)oxy)-3-((4-methoxybenzyl)oxy)-5,9-dimethyltrideca-4,8,12-trienal (549). To a solution of alcohol **548** (1.58 g, 2.11 mmol) in CH_2Cl_2 (40 mL) was added DMP (15% in CH_2Cl_2, 6.59 mL, 3.17 mmol) dropwise at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 2.5 h. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (25 mL) and saturated aqueous $Na_2S_2O_3$ (25 mL). Stirring was continued

for 30 min, when two almost clear phases had formed. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were washed with brine (100 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the residue by flash column chromatography (hexane/EtOAc 10:1) yielded 1.28 g (82%) of aldehyde **549** as a colorless oil.

TLC: R_f 0.18 (hexane/EtOAc 10:1, UV, CPS).

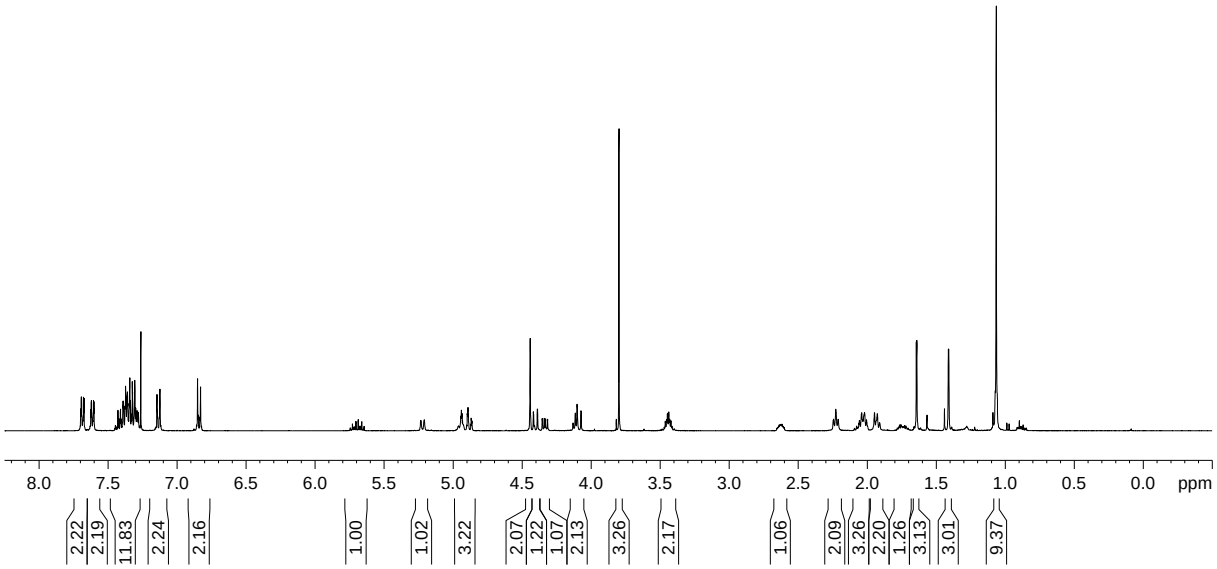
$[\alpha]^{20}_D$: = -24.5 (*c* = 0.46, $CHCl_3$).

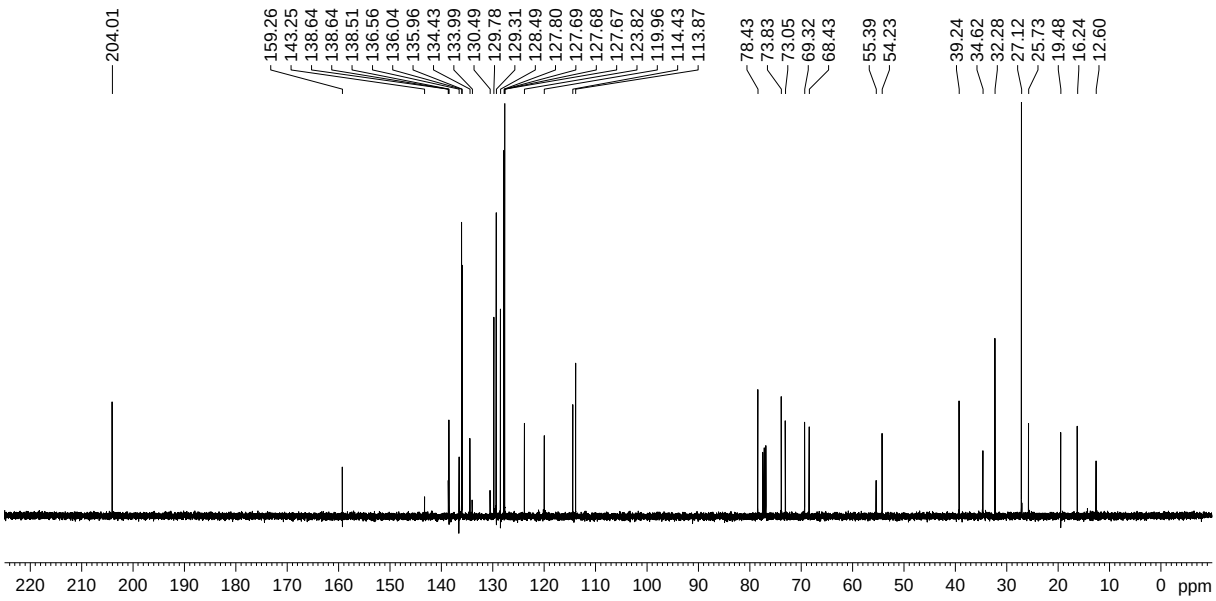
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 9.69 (d, *J* = 2.1 Hz, 1H), 7.69 (dd, *J* = 6.5, 1.4 Hz, 2H), 7.61 (dd, *J* = 6.5, 1.4 Hz, 2H), 7.45-7.27 (m, 11H), 7.13 (d, *J* = 8.6 Hz, 2H), 6.84 (d, *J* = 8.6 Hz, 2H), 5.69 (ddt, *J* = 17.0, 10.3, 6.4 Hz, 1H), 5.22 (d, *J* = 9.7 Hz, 1H), 4.98-4.92 (m, 1H), 4.92 (dq, *J* = 17.0, 1.7 Hz, 1H), 4.88 (dm, *J* = 9.6 Hz, 1H), 4.44 (s, 2H), 4.40 (d, *J* = 11.4 Hz, 1H), 4.34 (dd, *J* = 9.5, 5.7 Hz, 1H), 4.12 (t, *J* = 6.4 Hz, 1H), 4.09 (d, *J* = 11.4 Hz, 1H), 3.80 (s, 3H), 3.44 (ddd, *J* = 13.0, 6.7, 3.0 Hz, 2H), 2.63 (dddd, *J* = 9.5, 4.1, 4.1, 2.3 Hz, 1H), 2.23 (t, *J* = 6.7 Hz, 2H), 2.10-2.00 (m, 3H), 1.96-1.90 (m, 2H), 1.74 (dddd, *J* = 12.7, 6.4, 6.2, 4.4 Hz, 1H), 1.64 (d, *J* = 1.1 Hz, 3H), 1.41 (d, *J* = 1.2 Hz, 3H), 1.06 (s, 9H).

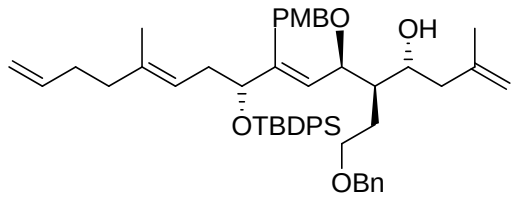
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 204.0, 159.3, 143.3, 138.6, 138.6, 138.5, 136.6, 136.0, 136.0, 134.4, 134.0, 130.5, 129.8, 129.3, 128.5, 127.8, 127.7, 127.7, 127.7, 123.8, 120.0, 114.4, 113.9, 78.4, 73.8, 73.1, 69.3, 68.4, 55.4, 54.2, 39.2, 34.6, 32.3, 27.1, 25.7, 19.5, 16.2, 12.6.

IR (film): ν 3450, 3071, 2998, 2956, 2931,2895, 2857, 1721, 1612, 1587, 15113, 1471, 1455, 1428, 1389, 1362, 1302, 1248, 1173, 1110, 1064, 1037, 1008, 999, 939,911, 845, 821, 740, 701, 612, 566, 506.

HRMS (ESI): calculated for $C_{48}H_{60}O_5Si$ $[M+Na]^+$ 767.4102, found 767.4113.







550

(4*R*,5*R*,6*R*,7*E*,9*R*,11*E*)-5-(2-(benzyloxy)ethyl)-9-((*tert*-butyldiphenylsilyl)oxy)-6-((4-methoxybenzyl)oxy)-2,8,12-trimethylhexadeca-1,7,11,15-tetraen-4-ol (550). To a solution of aldehyde **549** (0.37 g, 0.50 mmol) in CH_2Cl_2 (35 mL) was added $SnCl_4$ (123 µg, 0.47 mmol) and after 2 min methallyl silyl (70 µg, 0.55 mmol) at -90 °C and the reaction mixture was stirred for 15 min at this temperature. The reaction was then cautiously quenched by addition of water (10 mL) and saturated aqueous $NaHCO_3$ (30 mL). The solution was allowed to warm to room temperature, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 10:1) to yield 0.29 g (73%) of alcohol **550** as a single isomer as a colorless oil.

TLC: R_f 0.17 (hexane/EtOAc 8:1, UV, CPS).

$[\alpha]^{20}_D$: = -22.7 (*c* = 0.48, $CHCl_3$).

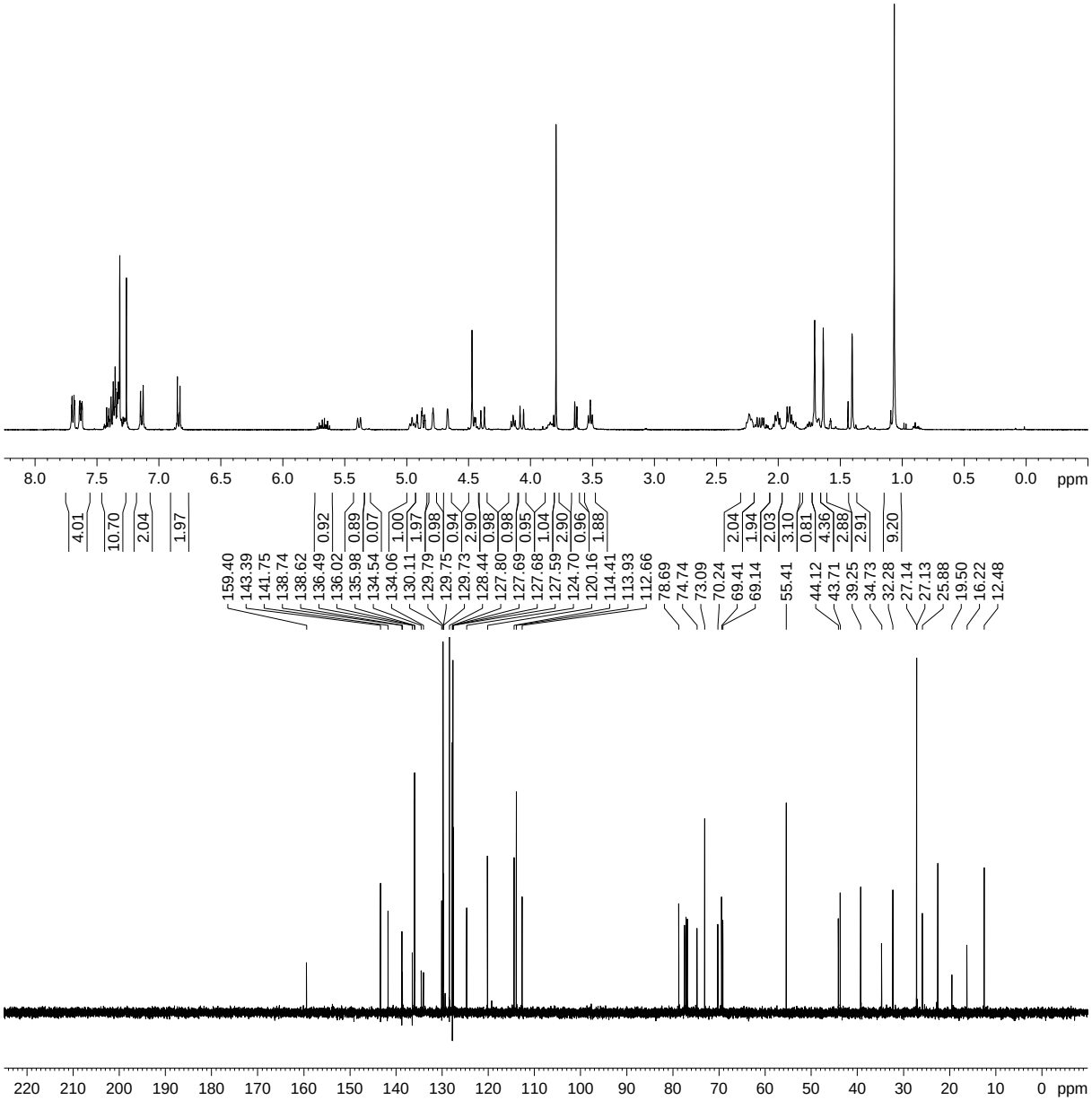
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.70 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.63 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.45-7.27 (m, 11H), 7.14 (d, *J* = 8.6 Hz, 2H), 6.84 (d, *J* = 8.6 Hz, 2H), 5.67 (dddd, *J* = 17.0, 10.1, 6.7, 6.4 Hz, 1H), 5.39 (d, *J* = 8.9 Hz, 1H), 4.96 (dd, *J* = 7.2, 6.8 Hz, 1H), 4.90 (dq, *J* = 17.0, 1.3 Hz, 1H), 4.86 (dq, *J* = 9.3 Hz, 1H), 4.79 (t, *J* = 1.5 Hz, 1H), 4.67

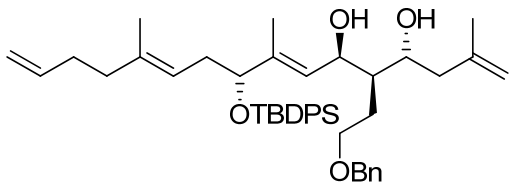
(s, 1H), 4.47 (s, 2H), 4.47-4.44 (m, 1H), 4.39 (d, J = 11.3 Hz, 1H), 4.14 (t, J = 6.5 Hz, 1H), 4.07 (d, J = 11.3 Hz, 1H), 3.84 (ddd, J = 12.6, 7.8, 5.7 Hz, 1H), 3.79 (s, 3H), 3.63 (d, J = 7.0 Hz, 1H), 3.52 (t, J = 6.7 Hz, 2H), 2.29-2.20 (m, 2H), 2.18 (dd, J = 14.0, 8.0 Hz, 1H), 2.11 (dd, J = 14.0, 5.7 Hz, 1H), 2.05-1.97 (m, 2H), 1.94-1.86 (m, 3H), 1.75 (ddd, J = 14.1, 7.1, 5.2 Hz, 1H), 1.71 (s, 3H), 1.70-1.66 (m, 1H), 1.64 (s, 3H), 1.40 (s, 3H), 1.06 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 159.4, 143.4, 141.7, 138.7, 138.6, 136.5, 136.0, 136.0, 134.5, 134.1, 130.1, 129.8, 129.7, 129.7, 128.4, 127.8, 127.7, 127.7, 127.6, 124.7, 120.2, 114.4, 113.9, 112.7, 78.7, 74.7, 73.1, 70.2, 69.4, 69.1, 55.4, 44.1, 43.7, 39.3, 34.7, 32.3, 27.1, 27.1, 25.9, 19.5, 16.2, 12.5

IR (film): ν 3480, 2997, 2931, 2857, 1612, 1513, 1471, 1454, 1428, 1390, 1377, 1362, 1302, 1249, 1174, 1110, 1059, 1038, 1009, 999, 940, 910, 888, 847, 822, 740, 702, 612, 505.

HRMS (ESI): calculated for $C_{52}H_{68}O_5Si$ $[M+Na]^+$ 823.4728, found 823.4734.





557

(4*R*,5*R*,6*R*,7*E*,9*R*,11*E*)-5-(2-(benzyloxy)ethyl)-9-((*tert*-butyldiphenylsilyl)oxy)-2,8,12-trimethylhexadeca-1,7,11,15-tetraene-4,6-diol (557). To a solution of PMB-ether **550** (0.51 g, 0.64 mmol) in MeCN (22.5 mL) and water (2.5 mL) was added CAN (0.91 g, 1.67 mmol) in one portion at room temperature and the reaction mixture was stirred for 2 hours. The reaction was then cautiously quenched by addition of saturated aqueous $NaHCO_3$ (30 mL and the aqueous phase was extracted with EtOAc (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 5:1) to yield 0.24 g (55%, 74% brsm) of diol **557** as a colorless oil.

Note: The reaction was quenched prior to full conversion because the yield would not be any better. If the starting material was resubjected to the above described conditions, 74% yield can be achieved.

TLC: R_f 0.42 (hexane/EtOAc 4:1, UV, CPS).

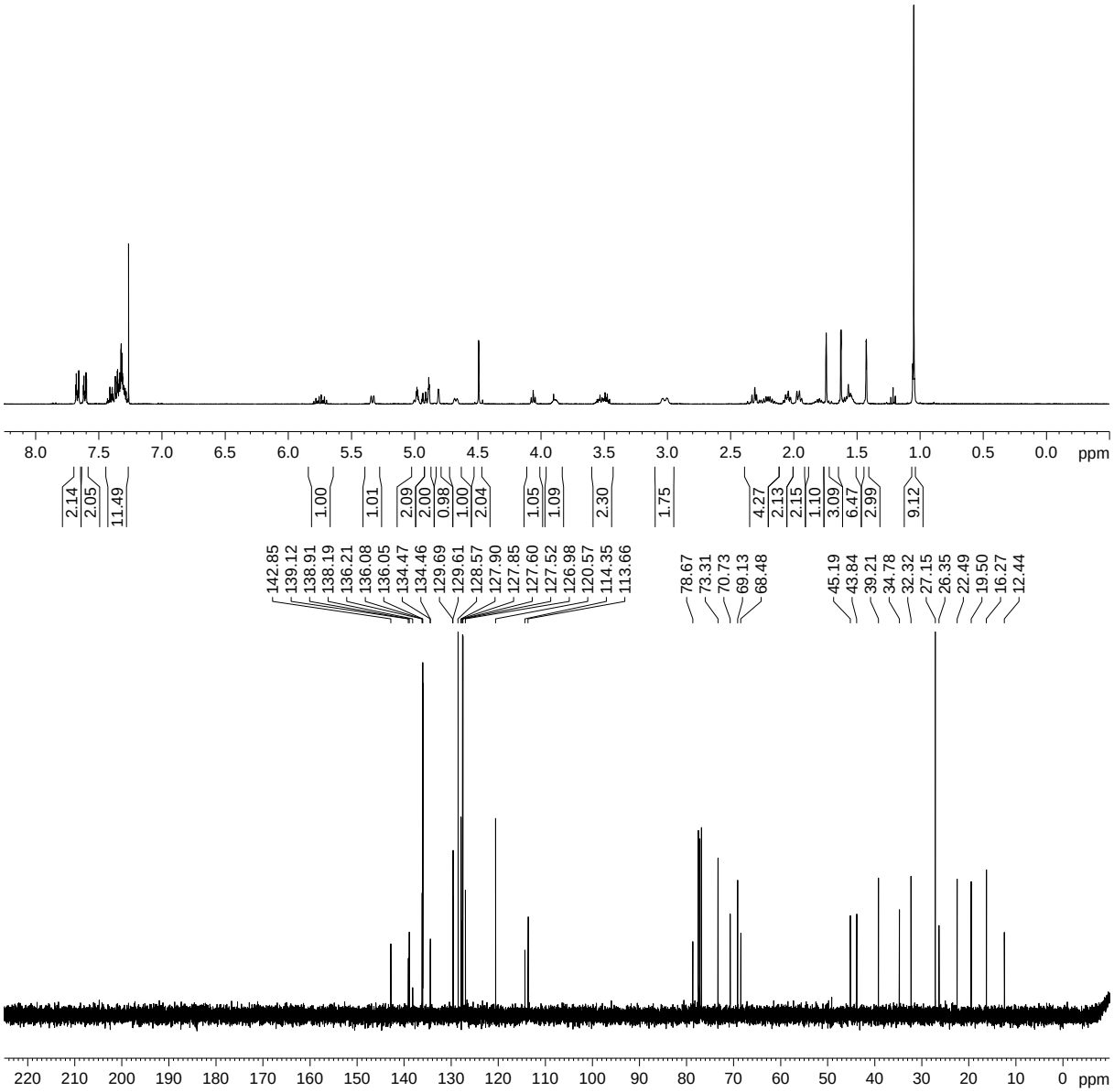
$[\alpha]^{20}_D$: = 17.8 (*c* = 0.60, $CHCl_3$).

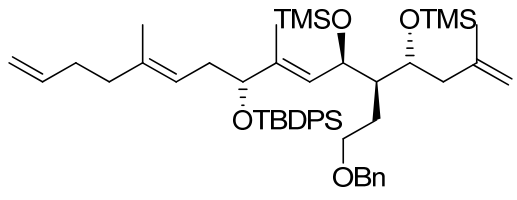
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.67 (ddd, *J* = 6.6, 1.4, 1.3 Hz, 2H), 7.61 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.44-7.27 (m, 11H), 5.67 (dddd, *J* = 16.8, 10.2, 6.4, 6.4 Hz, 1H), 5.33 (d, *J* = 8.5 Hz, 1H), 4.98 (dm, *J* = 10.2 Hz, 1H), 4.96 (dm, *J* = 16.8 Hz, 1H), 4.90 (dq, *J* = 17.0, 1.3 Hz, 1H), 4.90 (dm, *J* = 9.3 Hz, 1H), 4.89 (m, 1H), 4.81 (q, *J* = 1.0 Hz, 1H), 4.67 (d, *J* = 8.2 Hz, 1H), 4.49 (s, 2H), 4.06 (t, *J* = 6.2 Hz, 1H), 3.93-3.85 (m, 1H), 3.58-3.52 (m, 1H), 3.49 (ddd, *J* = 9.4, 7.0, 5.4 Hz, 1H), 3.02 (dd, *J* = 14.1, 3.0 Hz, 2H), 2.34 (dd, *J* = 14.1, 8.9 Hz, 1H), 2.28 (dd, *J* = 14.1, 5.3 Hz, 1H), 2.21 (dd, *J* = 13.1, 6.8 Hz, 1H), 2.16 (dd, *J* = 14.3, 6.7 Hz, 1H), 2.05 (dq, *J* = 7.1, 1.3 Hz, 2H), 1.98-1.93 (m, 2H), 1.81 (dq, *J* = 12.4, 7.0 Hz, 1H), 1.74 (s, 3H), 1.60-1.50 (m, 1H), 1.63 (d, *J* = 1.3 Hz, 3H), 1.43 (s, 3H), 1.05 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 142.8, 139.1, 138.9, 138.2, 136.2, 136.1, 136.1, 134.5, 134.5, 129.7, 129.6, 128.6, 127.9, 127.8, 127.6, 127.5, 127.0, 120.6, 114.3, 113.7, 78.7, 73.3, 70.7, 69.1, 68.5, 45.2, 43.8, 39.2, 34.8, 32.3, 27.1, 26.3, 22.5, 19.5, 16.3, 12.4.

IR (film): ν 3363, 3071, 2931, 2894, 2857, 1642, 1471, 1453, 1389, 1362, 1109, 1065, 1029, 1003, 941, 907, 40, 702, 611, 505.

HRMS (ESI): calculated for $C_{44}H_{60}O_4Si$ $[M+Na]^+$ 701.4153, found 701.4146.





561

(4*R*,5*R*,6*R*,9*R*,*E*)-5-(2-(benzyloxy)ethyl)-2,2,8,12,12-pentamethyl-4-(2-methylallyl)-9-((*E*)-3-methylhepta-2,6-dien-1-yl)-11,11-diphenyl-6-((trimethylsilyl)oxy)-3,10-dioxa-2,11-disilatridec-7-ene (561). To a solution of diol **557** (0.31 g, 0.46 mmol) in CH_2Cl_2 (35 mL) and was added 2,6-lutidine (0.27 mL, 2.31 mmol) at 0 °C and the reaction mixture was stirred for 10 min. The reaction mixture was then cooled to -78 °C and TMSOTf (0.21 mL, 1.16 mmol) was added dropwise. The reaction mixture was stirred at this temperature for 20 min.

and the reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (30 mL). The phases were separated and the aqueous phase was extracted with ether (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 50:1) to give 0.37 g (97%) of silyl ether **561** as a colorless oil.

TLC: R_f 0.38 (hexane/EtOAc 30:1, UV, CPS).

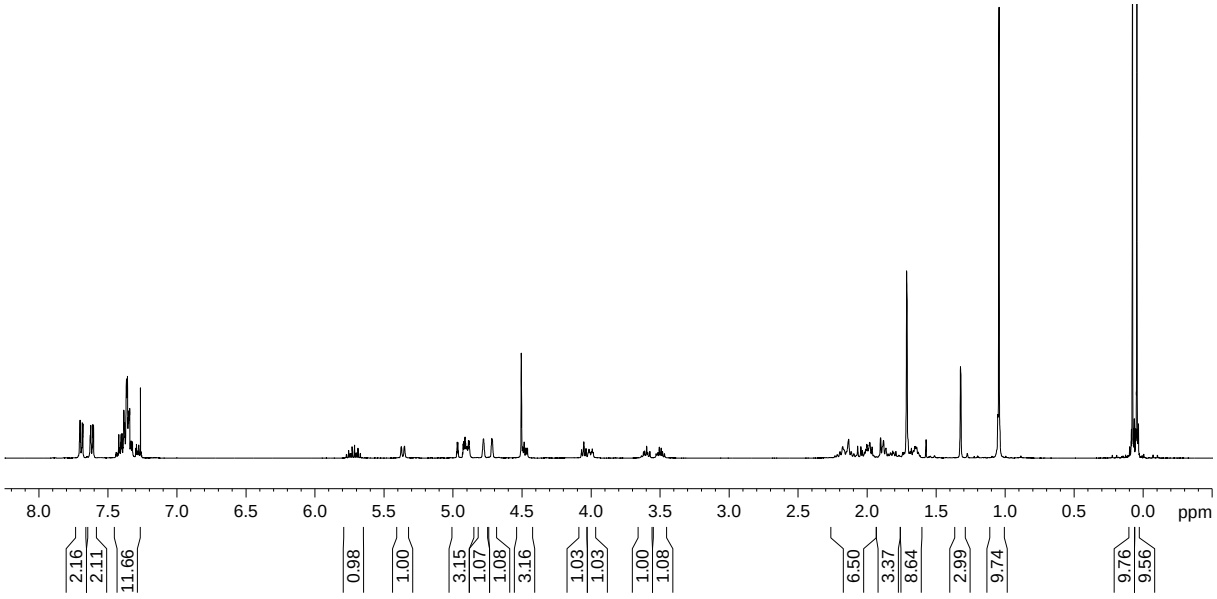
$[\alpha]^{20}_D$: = 2.6 (*c* = 2.21, $CHCl_3$).

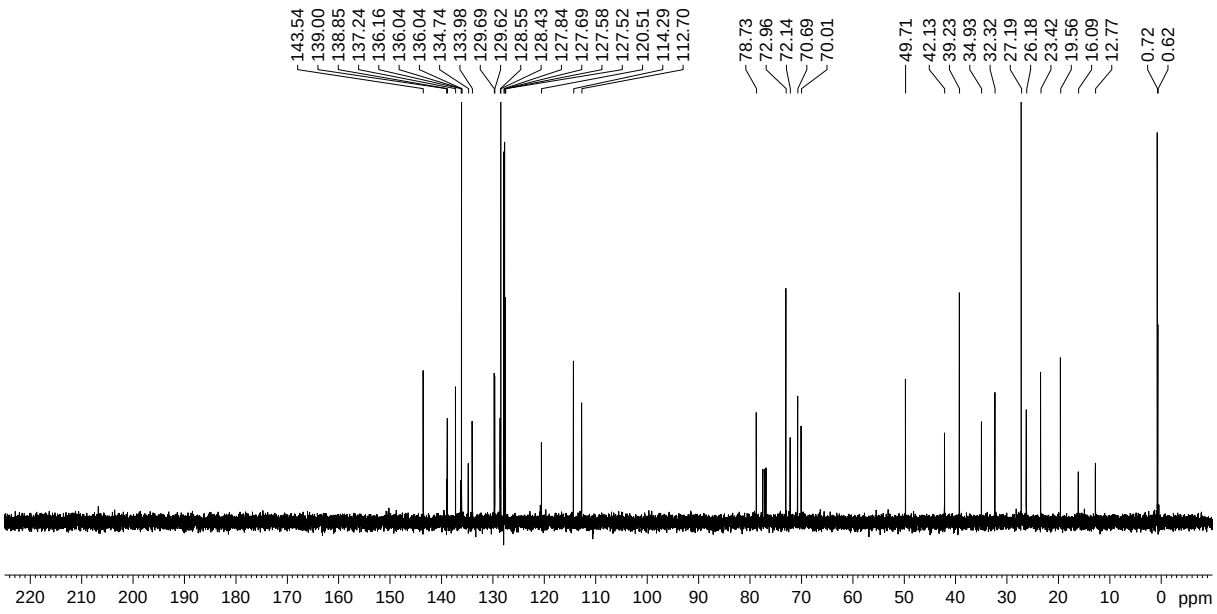
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.69 (ddd, *J* = 6.6, 1.4, 1.3 Hz, 2H), 7.62 (ddd, *J* = 6.5, 1.4, 1.3 Hz, 2H), 7.45-7.25 (m, 11H), 5.72 (dddd, *J* = 17.0, 10.3, 6.3, 6.3 Hz, 1H), 5.36 (d, *J* = 9.1 Hz, 1H), 4.94 (dq, *J* = 17.0, 1.6 Hz, 1H), 4.92-4.88 (m, 1H), 4.90 (dm, *J* = 10.3 Hz, 1H), 4.78 (t, *J* = 1.6 Hz, 1H), 4.72 (s, 1H), 4.50 (s, 2H), 4.48 (dd, *J* = 9.3, 4.2 Hz, 1H). 4.05 (t, *J* = 6.2 Hz, 1H), 4.00 (ddd, *J* = 9.5, 2.9, 2.6 Hz, 1H), 3.60 (td, *J* = 9.3, 5.7 Hz, 1H), 3.50 (td, *J* = 9.5, 5.7 Hz, 1H), 2.24-1.94 (m, 6H), 1.90-1.76 (m, 3H), 1.74-1.61 (m, 2H), 1.71 (s, 3H), 1.71 (s, 3H), 1.32 (s, 3H), 1.05 (s, 9H), 0.08 (s, 9H), 0.05 (s, 9H).

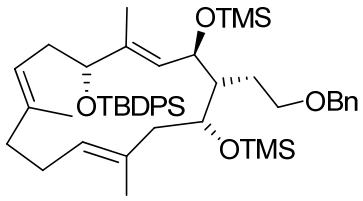
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 143.5, 139.0, 138.8, 137.2, 136.2, 136.0, 136.0, 134.7, 134.0, 129.7, 129.6, 128.5, 128.4, 127.8, 127.7, 127.6, 127.5, 120.5, 114.3, 112.7, 78.7, 73.0, 72.1, 70.7, 70.0, 49.7, 42.1, 39.2, 34.9, 32.3, 27.2, 26.2, 23.4, 19.6, 16.1, 12.8, 0.7, 0.6.

IR (film): ν 3071, 2956, 2932, 2897, 2857, 1452, 1428, 1387, 1363, 1250, 1108, 1065, 938, 879, 840, 741, 701, 612, 507.

HRMS (ESI): calculated for $C_{50}H_{76}O_4Si_3$ $[M+Na]^+$ 847.4944, found 847.4949.







562

(((1*R*,2*R*,3*R*,4*E*,6*R*,8*E*,12*E*)-2-(2-(benzyloxy)ethyl)-6-((*tert*-butyldiphenylsilyl)oxy)-5,9,13-trimethylcyclotetradeca-4,8,12-triene-1,3-diyl)bis(oxy))bis(trimethylsilane) (562).

To a solution of diene **561** (0.34 g, 0.41 mmol) in benzene (310 mL) was added *Hoveyda-Grubbs* 2nd generation catalyst (31 mg, 0.05 mmol) at room temperature and the reaction mixture was heated to 65 °C and stirred for 15 hours at this temperature. The reaction mixture was then filtered through a small plug of silica and the precipitate was rinsed with Et_2O (100 mL). The filtrate was then concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 50:1) to yield 0.31 g (94%) of RCM-product **562** as a single isomer as a colorless oil.

TLC: R_f 0.15 (hexane/EtOAc 50:1, UV, CPS).

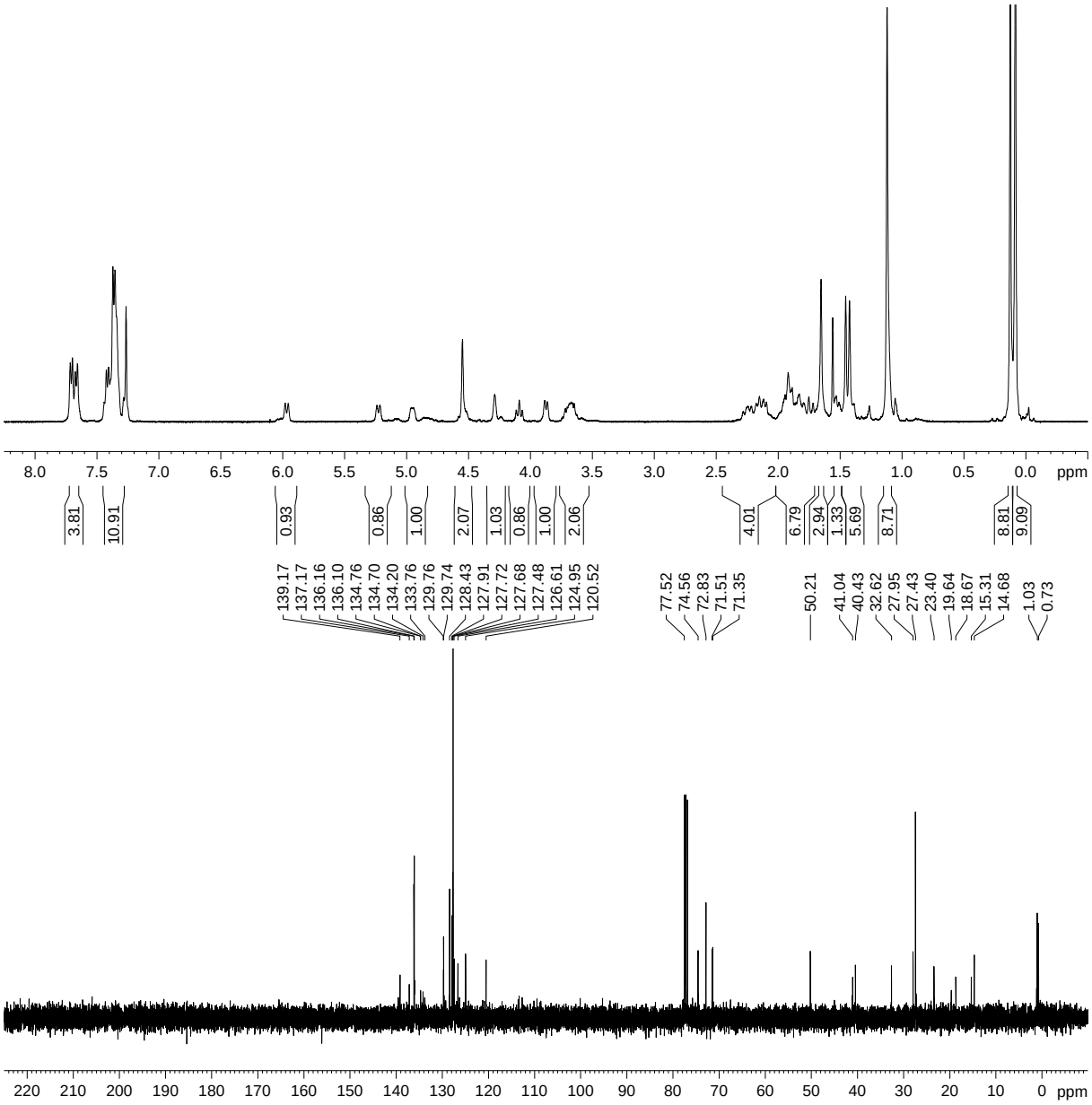
$[\alpha]^{20}_D$: = -25.7 (*c* = 0.50, $CHCl_3$).

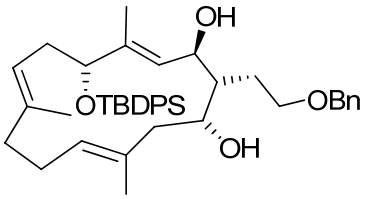
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.71 (d, *J* = 7.1 Hz, 2H), 7.66 (d, *J* = 7.1 Hz, 2H), 7.45-7.23 (m, 11H), 5.97 (d, *J* = 9.8 Hz, 1H), 5.23 (d, *J* = 9.8 Hz, 1H), 4.95 (d, *J* = 6.9 Hz, 1H), 4.55 (s, 2H), 4.29 (s, 1H), 4.09 (t, *J* = 9.7 Hz, 1H), 3.87 (d, *J* = 8.3 Hz, 1H), 3.76-3.61 (m, 2H), 2.30-2.08 (m, 4H), 2.00-1.70 (m, 7H), 1.65 (s, 3H), 1.55-1.48 (m, 1H), 1.45 (s, 3H), 1.42 (s, 3H), 1.12 (s, 9H), 0.13 (s, 9H), 0.08 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 139.2, 137.2, 136.2, 136.1, 134.8, 134.7, 134.2, 133.8, 129.8, 129.7, 128.4, 127.9, 127.7, 127.7, 127.5, 126.6, 125.0, 120.5, 77.5, 74.6, 72.8, 71.5, 71.4, 50.2, 41.0, 40.4, 32.6, 27.9, 27.4, 23.4, 19.6, 18.7, 15.3, 14.7, 1.0, 0.7.

IR (film): ν 3071, 3050, 3031, 2955, 2932, 2857, 1472, 1454, 1428, 1362, 1250, 1109, 1081, 1042, 869, 839, 741, 701, 611, 507.

HRMS (ESI): calculated for $C_{48}H_{72}O_4Si_3$ $[M+Na]^+$ 819.4631, found 819.4642.





563

(1*R*,2*R*,3*R*,4*E*,6*R*,8*E*,12*E*)-2-(2-(benzyloxy)ethyl)-6-((*tert*-butyldiphenylsilyl)oxy)-5,9,13-trimethylcyclotetradeca-4,8,12-triene-1,3-diol (563). To a solution of diol **562** (0.34 g, 0.43 mmol) in MeOH (21 mL) was added citric acid (0.27 g, 1.28 mmol) in one portion at room temperature and the reaction mixture was stirred for 1.5 hours. The reaction mixture was then diluted with CH_2Cl_2 (20 mL) and the reaction was cautiously quenched by addition of saturated aqueous $NaHCO_3$ (30 mL). The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 30 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 4:1) to yield 0.25 g (89%) of diol **563** as a white foam.

TLC: R_f 0.25 (hexane/EtOAc 3:1, UV, CPS).

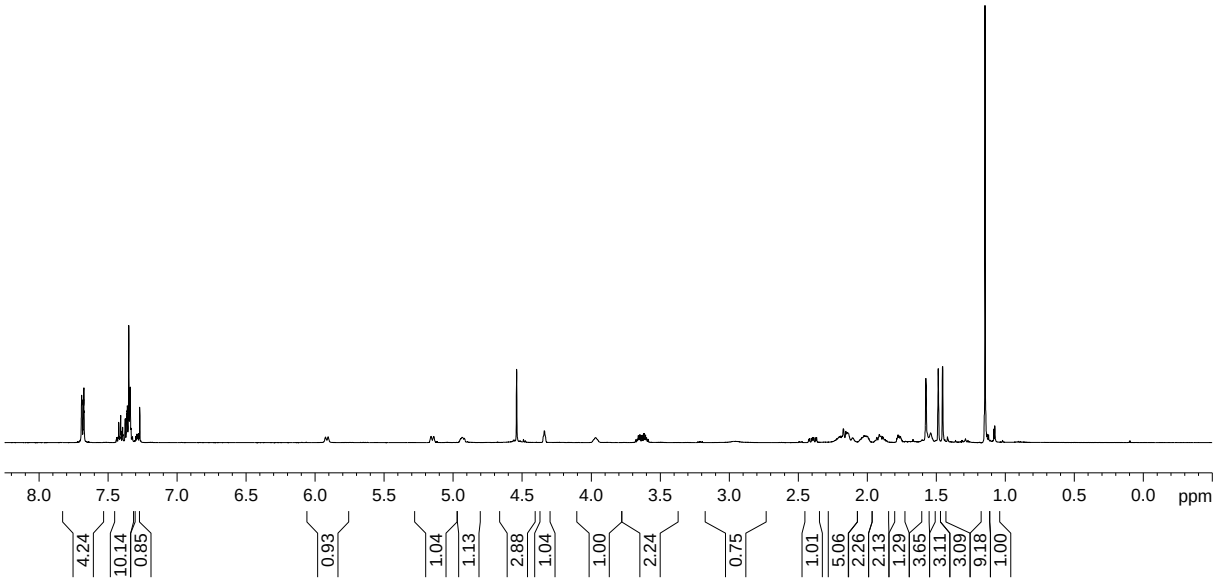
$[\alpha]^{20}_D$: = -21.6 (*c* = 0.31, $CHCl_3$).

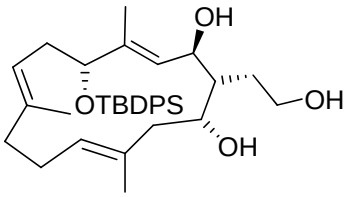
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.69-7.65 (m, 4H), 7.43-7.26 (m, 11H), 5.91 (d, *J* = 9.8 Hz, 1H), 5.14 (d, *J* = 9.8 Hz, 1H), 4.95-4.90 (m, 1H), 4.55-4.48 (m, 1H), 4.53 (s, 2H), 4.33 (t, *J* = 3.0 Hz, 1H), 3.99-3.94 (m, 1H), 3.65 (ddd, *J* = 9.4, 7.3, 4.9 Hz, 1H), 3.60 (ddd, *J* = 9.4, 6.5, 5.3 Hz, 1H), 2.93 (br s, 1H), 2.39 (ddd, *J* = 15.2, 9.8, 3.3 Hz, 1H), 2.24-2.11 (m, 4H), 2.05-1.97 (m, 2H), 1.94-1.85 (m, 2H), 1.76 (dddd, *J* = 8.7, 3.6, 3.5, 1.8 Hz, 1H), 1.57 (s, 3H), 1.55-1.49 (m, 2H), 1.48 (s, 3H), 1.45 (s, 3H), 1.14 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 139.5, 138.6, 136.1, 136.1, 134.6, 134.5, 134.0, 134.0, 129.8, 129.8, 128.5, 127.8, 127.7, 127.7, 127.7, 126.3, 125.1, 120.6, 74.7, 74.7, 73.2, 72.5, 70.0, 40.1, 33.3, 32.4, 27.4, 27.4, 26.4, 24.2, 19.6, 16.6, 14.8, 14.6.

IR (film): ν 3398, 2930, 2893,2856, 1472, 1454, 1428, 1386, 1363, 1206, 1107, 1077, 1028, 1007, 941, 823, 796, 741, 702, 612, 506.

HRMS (ESI): calculated for $C_{42}H_{56}O_4Si$ $[M+Na]^+$ 675.3840, found 675.3840.





494

(1*R*,2*R*,3*R*,4*E*,6*R*,8*E*,12*E*)-6-((*tert*-butyldiphenylsilyl)oxy)-2-(2-hydroxyethyl)-5,9,13-trimethylcyclotetradeca-4,8,12-triene-1,3-diol (494). To a solution of liquid ammonia (34 mL) was added benyl ether **563** (0.51 g, 0.78 mmol) in THF (34 mL) at -78 °C and the reaction mixture was stirred for 10 min. Sodium (38 mg, 1.56 mmol) was then added in one portion and the reaction was stirred for 20 min. The addition of sodium was repeated in the way described above until the reaction mixture turned blue and the reaction was immediately quenched by addition of saturated aqueous NH_4Cl (30 mL) after this observation. The reaction mixture was allowed to reach room temperature so that the ammonia would evaporate. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 1:2) to yield 0.43 mg (98%) of triol **563** as a colourless oil.

TLC: R_f 0.22 (hexane/EtOAc 1:1, UV, CPS).

$[\alpha]^{20}_D$: = -19.3 (*c* = 0.07, $CHCl_3$).

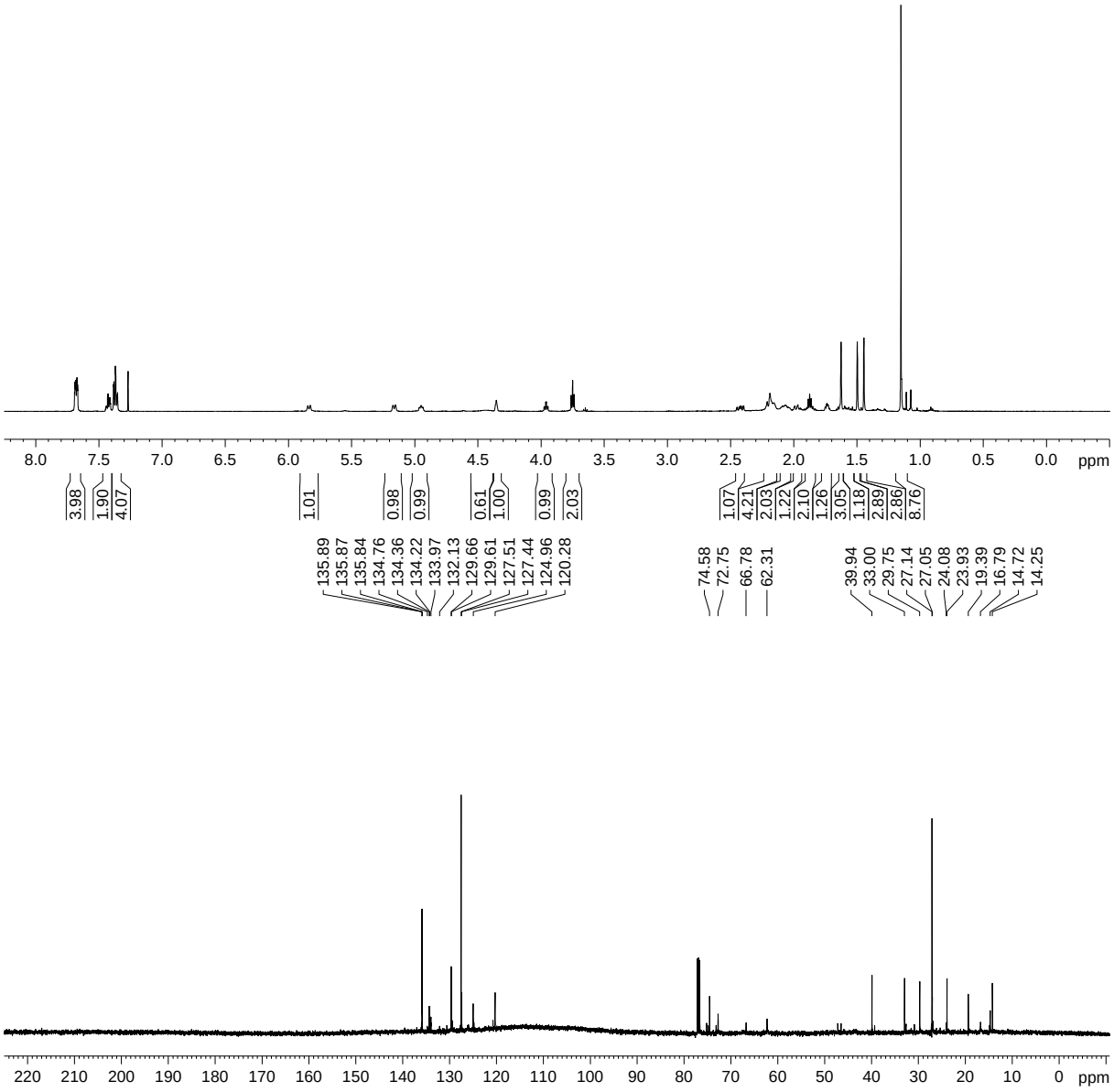
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.68 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.67 (ddd, *J* = 6.5, 1.5, 1.4 Hz, 2H), 7.45-7.41 (m, 2H), 7.37 (tdd, *J* = 7.1, 1.5, 1.4 Hz, 4H), 5.84 (d, *J* = 9.8 Hz, 1H),

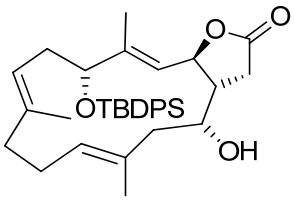
5.16 (d, *J* = 9.5 Hz, 1H), 4.95 (t, *J* = 7.0 Hz, 1H), 4.44 (br s, 1H), 4.36 (t, *J* = 3.2 Hz, 1H), 3.96 (td, *J* = 6.7, 2.1 Hz, 1H), 3.75 (t, *J* = 6.0 Hz, 2H), 2.42 (ddd, *J* = 15.3, 9.9, 3.1 Hz, 1H), 2.23-2.13 (m, 4H), 2.11-2.02 (m, 2H), 1.98 (dd, *J* = 11.9, 2.4 Hz, 1H), 1.91-1.83 (m, 2H), 1.73 (ddd, *J* = 11.9, 5.1, 2.0 Hz, 1H), 1.63 (s, 3H), 1.60-1.53 (m, 1H), 1.50 (s, 3H), 1.45 (s, 3H), 1.15 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 135.9, 135.9, 135.8, 134.8, 134.4, 134.2, 134.0, 132.1, 129.7, 129.6, 127.5, 127.4, 125.0, 120.3, 74.6, 72.8, 66.8, 62.3, 39.9, 33.0, 29.8, 27.1, 27.1, 24.1, 23.9, 19.4, 16.8, 14.7, 14.3.

IR (film): ν 3367, 2928, 2856, 1682, 1670, 1651, 1471, 1457, 1445, 1428, 1388, 1363, 1261, 1105, 1078, 1048, 1025, 942, 820, 798, 753, 741, 702, 665, 611, 506, 488.

HRMS (ESI): calculated for $C_{35}H_{50}O_4Si$ $[M+Na]^+$ 585.3371, found 585.3362





564

(3a*R*,4*R*,6*E*,10*E*,13*R*,14*E*,15a*R*)-13-((*tert*-butyldiphenylsilyl)oxy)-4-hydroxy-6,10,14-trimethyl-3,3a,4,5,8,9,12,13-octahydrocyclotetradeca[b]furan-2(15a*H*)-one (564). To a solution of triol **494** (95 mg, 0.17 mmol) in CH_2Cl_2 (7.5 mL) was added added Yttberbium triflate (4.2 mg, 0.007 mmol), BAIB (136 mg, 0.42 mmol) and TEMPO (4 mg, 0.025 mmol) at 0 °C. The reaction mixture was slowly allowed to reach room temperature and stirred for 4 h at this temperature. The reaction was then cautiously quenched by addition of 10% aqueous $Na_2S_2O_3$ (7 mL). Stirring was continued for 10 min, the phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 10 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (hexane/EtOAc 5:2) to yield 71 mg (75%) of lactone **564** as a white foam.

TLC: R_f 0.21 (hexane/EtOAc 5:2, UV, CPS).

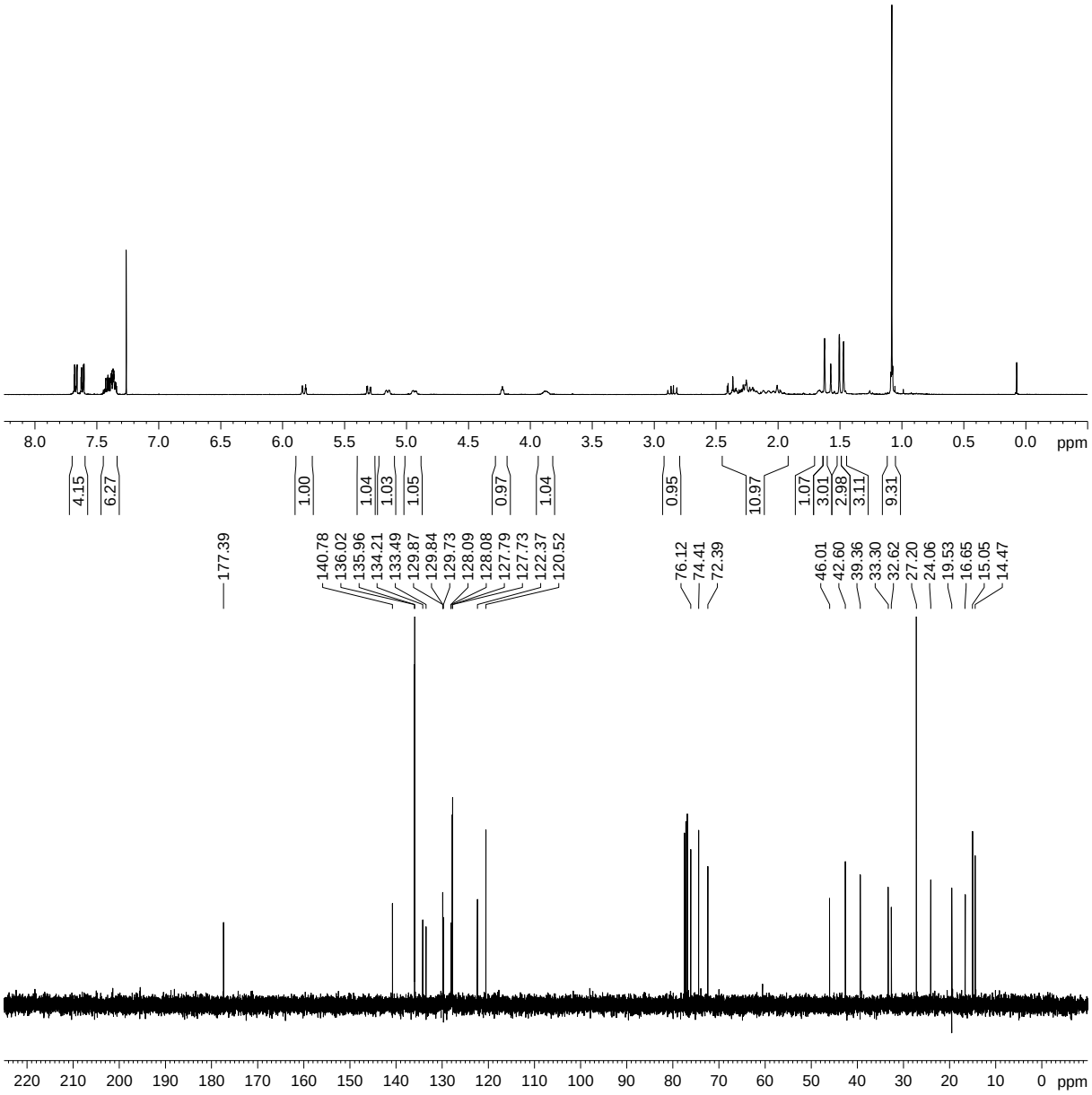
$[\alpha]^{20}_D$: = -3.2 (*c* = 0.37, $CHCl_3$).

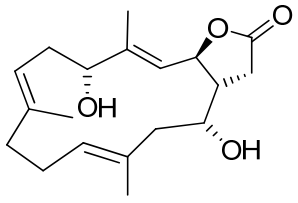
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.67 (ddd, *J* = 6.4, 1.5, 1.4 Hz, 2H), 7.62 (d, *J* = 6.4, 1.5, 1.4 Hz, 2H), 7.45-7.34 (m, 6H), 5.87 (dp, *J* = 10.4, 1.3 Hz, 1H), 5.30 (d, *J* = 10.4, 2.3 Hz, 1H), 5.15 (dd, *J* = 10.2, 2.2 Hz, 1H), 4.94 (dd, *J* = 9.3, 2.9 Hz, 1H), 4.23 (t, *J* = 3.3 Hz, 1H), 3.88 (dddd, *J* = 9.3, 5.5, 5.4, 2.3 Hz, 1H), 2.85 (dd, *J* = 18.0, 10.5 Hz, 1H), 2.40-2.13 (m, 7H), 2.10-1.93 (m, 3H), 1.70-1.64 (m, 1H), 1.63 (s, 3H), 1.51 (d, *J* = 1.3 Hz, 3H), 1.47 (s, 3H), 1.08 (s, 9H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 177.4, 140.8, 136.0, 136.0, 134.2, 133.5, 129.9, 129.8, 129.7, 128.1, 128.1, 127.8, 127.7, 122.4, 120.5, 76.1, 74.4, 72.4, 46.0, 42.6, 39.4, 33.3, 32.6, 27.2, 24.1, 19.5, 16.6, 15.0, 14.5.

IR (film): ν 3440, 2930, 2896, 2857, 1771, 1472, 1428, 1388, 1362, 1340, 1186, 1108, 1082, 1047, 994, 955, 822, 703, 611, 504, 487.

HRMS (ESI): calculated for $C_{35}H_{47}O_4Si$ $[M+H]^+$ 559.3238, found 559.3243.





565

(3a*R*,4*R*,6*E*,10*E*,13*R*,14*E*,15a*R*)-4,13-dihydroxy-6,10,14-trimethyl-3,3a,4,5,8,9,12,13-octahydrocyclotetradeca[b]furan-2(15a*H*)-one (565). To a solution of silyl ether **564** (5.2 mg, 0.009 mmol) in MeCN (0.5 mL) was added added TASF (15.4 mg, 0.06 mmol) in one portion at room temperature. The reaction mixture was heated up to 80 °C and was stirred for 24 hours at this temperature. The reaction mixture was allowed to reach room temperature and the reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl

(1 mL). The phases were separated and the aqueous phase was extracted with EtOAc (3 x 2 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure.The residue was purified by flash column chromatography (hexane/EtOAc 1:1) to yield 2.3 mg (77%) of diol **565** as a colourless oil.

TLC: R_f 0.30 (hexane/EtOAc 1:1, CPS).

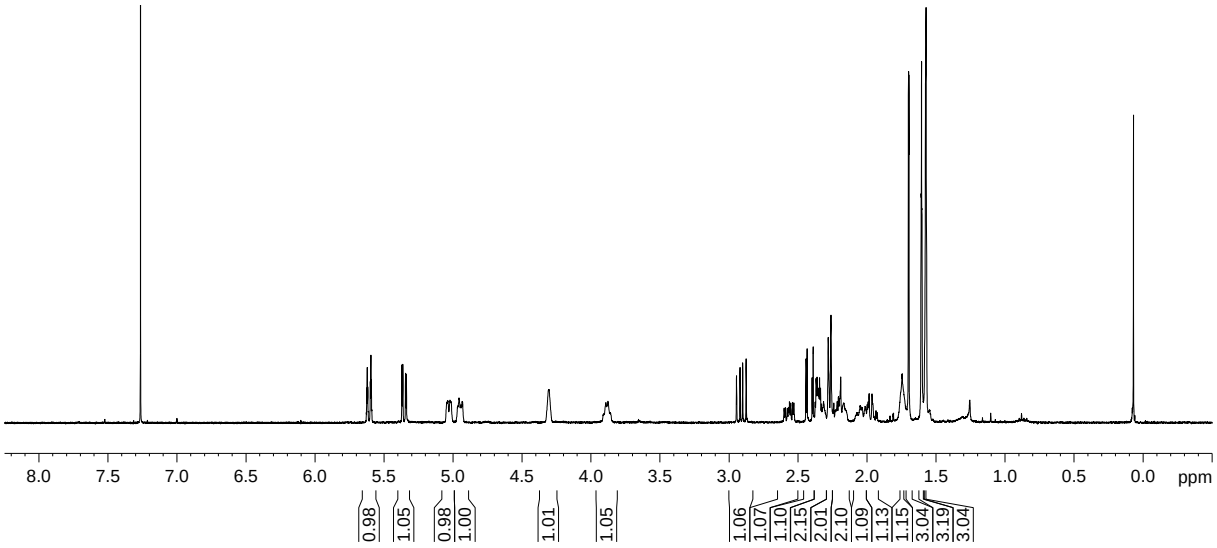
$[\alpha]^{20}_D$: = -119.9 (*c* = 0.15, $CHCl_3$).

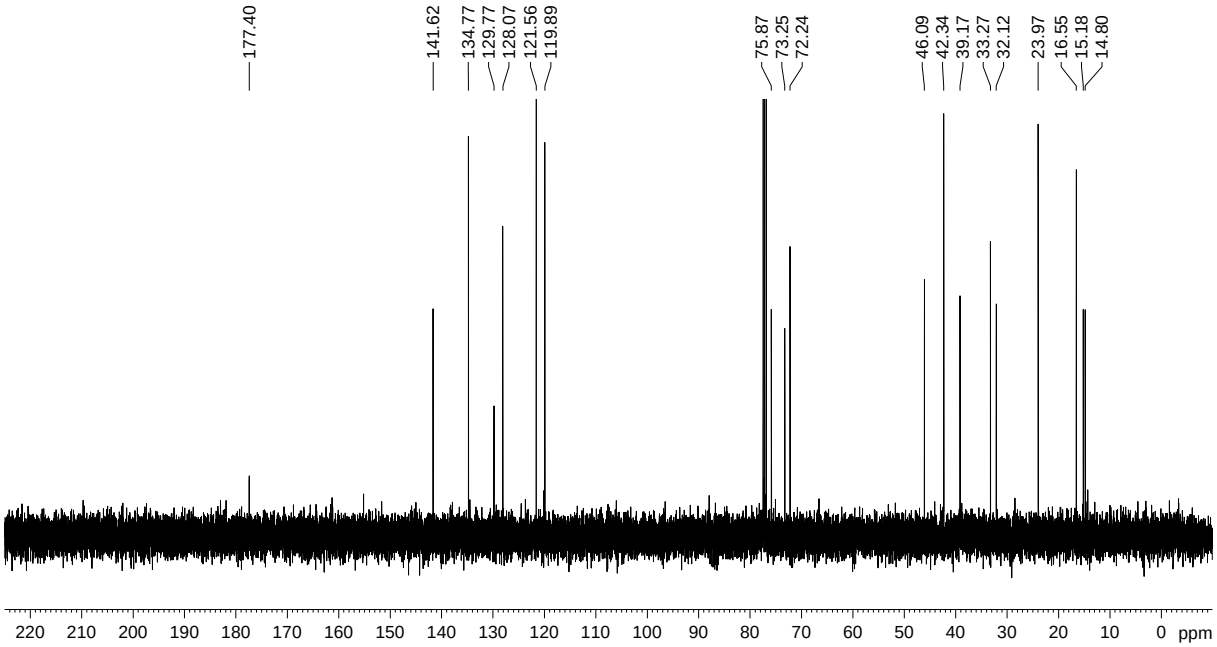
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.61 (dp, *J* = 10.3, 1.3 Hz, 1H), 5.30 (d, *J* = 10.3, 2.5 Hz, 1H), 5.03 (ddd, *J* = 9.4, 2.6, 1.2 Hz, 1H), 4.94 (ddd, *J* = 9.4, 4.6, 1.2 Hz, 1H), 4.31 (s, 1H), 3.88 (ddd, *J* = 14.2, 5.7, 2.5 Hz, 1H), 2.91 (dd, *J* = 17.7, 10.1 Hz, 1H), 2.57 (ddd, *J* = 15.7, 9.3, 3.5 Hz, 1H), 2.41 (dd, *J* = 17.7, 3.0 Hz, 1H), 2.35 (ddd, *J* = 10.1, 5.4, 2.6 Hz, 1H), 2.34 (dd, *J* = 5.4, 2.6 Hz, 1H), 2.34-2.29 (m, 1H), 2.27 (d, *J* = 8.0 Hz, 2H), 2.25-2.13 (m, 2H), 2.04 (dd, *J* = 13.9, 9.3 Hz, 1H), 2.00-1.92 (m, 1H), 1.76-1.70 (br s, 1H), 1.70 (d, *J* = 1.7 Hz, 3H), 1.61 (t, *J* = 1.2 Hz, 3H), 1.57 (s, 3H).

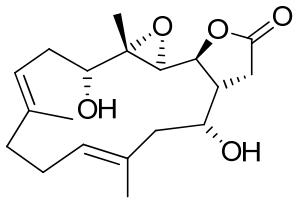
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 177.4, 141.6, 134.8, 129.8, 128.1, 121.6, 119.9, 75.9, 73.2, 72.2, 46.1, 42.3, 39.2, 33.3, 32.1, 24.0, 16.6, 15.2, 14.8.

IR (film): ν 3420, 2926, 2853, 1759, 1439, 1417, 1385, 1340, 1259, 1197, 1047, 991, 956, 925, 912, 733.

HRMS (ESI): calculated for $C_{19}H_{28}O_4$ $[M+Na]^+$ 343.1880, found 343.1884.







566

(1a*S*,2*R*,4*E*,8*E*,11*R*,11a*R*,14a*S*,14b*S*)-2,11-dihydroxy-1a,5,9-trimethyl-2,3,6,7,10,11,11a,12,14a,14b-decahydrooxireno[2',3':13,14]cyclotetradeca[1,2-b]furan-13(1a*H*)-one (566). To a solution of allylic alcohol **565** (22 mg, 0.069 mmol) in benzene (2.5 mL) was added added *t*-BuOOH (11.3 uL, 5.5 in decane, 0.062 mmol) in benzene (0.1 mL) and $VO(acac)_2$ (1.1 mg, 4.1 umol) in benzene (0.1 mL) at 0 °C whereupon the colour of the reaction mixture turned red. The reaction mixture was stirred for 10 min and was then allowed to reach room temperature and stirred for 0.5 hours at this temperature. The colour of the reaction mixture went from red to pale yellow during this time. The reaction was then quenched with saturated aqueous $Na_2S_2O_3$ (3 mL) and Et_2O (3 mL) was added. The phases were separated and the aqueous phase was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (hexane/EtOAc 3:2) to yield 12.0 mg (52%) of epoxide **566** as a colourless oil.

TLC: R_f 0.18 (hexane/EtOAc 3:2, CPS).

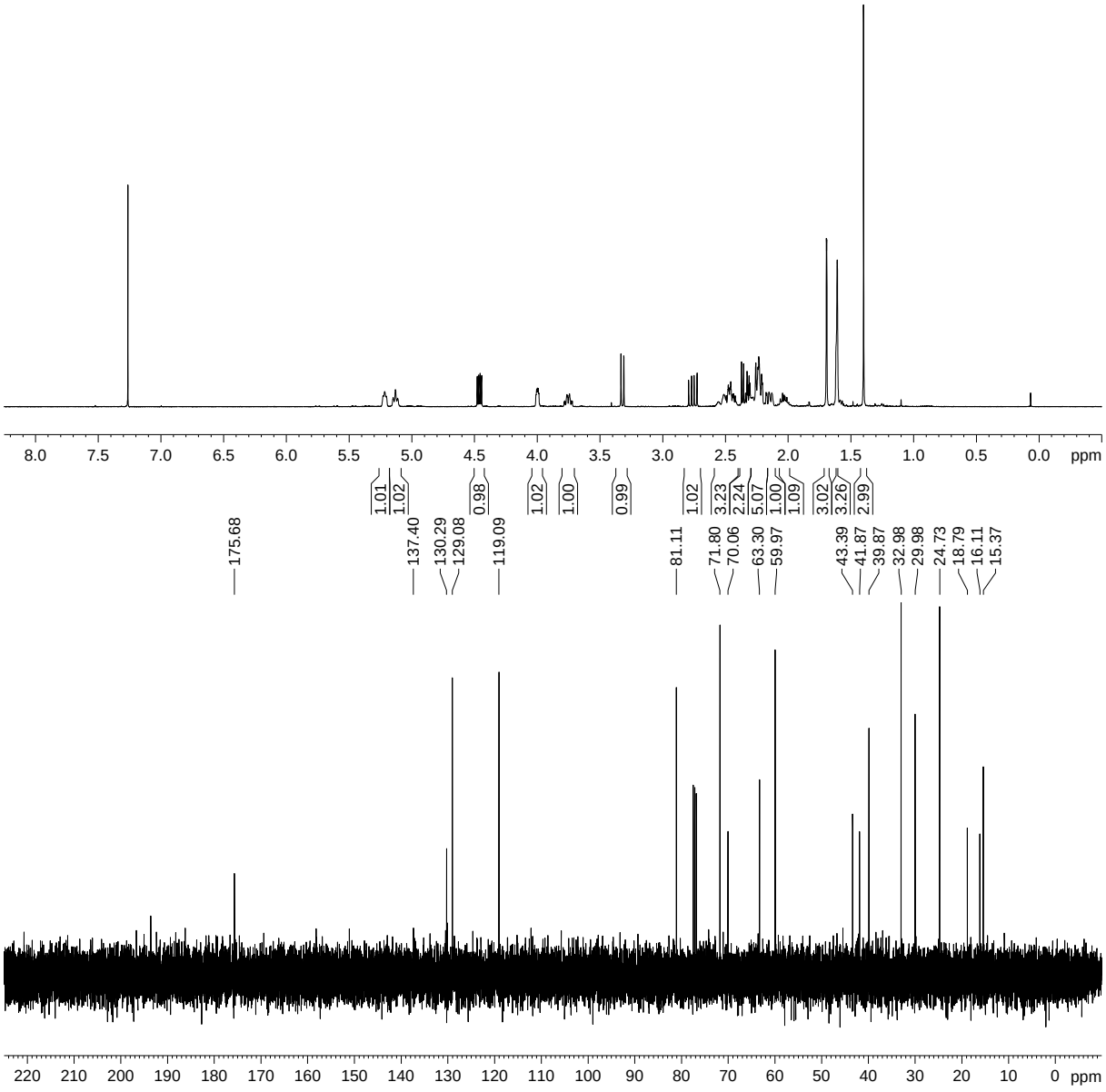
$[\alpha]^{20}_D$: = -38.8 (*c* = 0.77, $CHCl_3$).

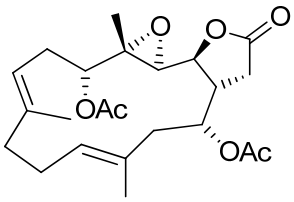
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.22 (t, *J* = 5.2 Hz, 1H), 5.13 (t, *J* = 7.3 Hz, 1H), 4.46 (dd, *J* = 9.1, 4.9 Hz, 1H), 4.00 (dt, *J* = 4.8, 2.0 Hz, 1H), 3.75 (dddd, *J* = 8.6, 8.3, 6.6, 2.6 Hz, 1H), 3.32 (d, *J* = 9.1 Hz, 1H), 2.76 (dd, *J* = 17.5, 9.1 Hz, 1H), 2.57-2.40 (m, 3H), 2.34 (dd, *J* = 17.5, 6.3 Hz, 1H), 2.34-2.30 (m, 1H), 2.29-2.20 (m, 5H), 2.14 (d, *J* = 8.6 Hz, 1H), 2.07-1.99 (m, 1H), 1.69 (d, *J* = 1.1 Hz, 3H), 1.61 (s, 3H), 1.40 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 175.7, 137.4, 130.3, 129.1, 119.1, 81.1, 71.8, 70.1, 63.3, 60.0, 43.4, 41.9, 39.9, 33.0, 30.0, 24.7, 18.8, 16.1, 15.4.

IR (film): ν 3449, 2923, 2856, 1770, 1422, 1384, 1345, 1231, 1182, 1050, 998, 965, 861, 819, 755, 701, 639, 545.

HRMS (ESI): calculated for $C_{19}H_{28}O_5$ $[M+Na]^+$ 359.1829, found 359.1824.





567

(1a*S*,2*R*,4*E*,8*E*,11*R*,11a*R*,14a*S*,14b*S*)-1a,5,9-trimethyl-13-oxo-1a,2,3,6,7,10,11,11a,12,13,14a,14b-dodecahydrooxireno[2',3':13,14]cyclotetradeca[1,2-b]furan-2,11-diyl diacetate (567). To a solution of diol **566** (11.5 mg, 0.03 mmol) in MeCN (3.0 mL) was added added NEt_3 (14.3 uL, 0.10 mmol), DMAP (0.42 mg, 0.003 mmol) and Ac_2O (9.7 uL, 0.10 mmol) at 0 °C. The reaction mixture was allowed to reach room temperature and stirred for 4 hours. The reaction mixture was allowed to reach room temperature and the reaction was then cautiously quenched by addition of saturated aqueous NH_4Cl (3 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (hexane/EtOAc 5:2) to yield 11.5 mg (80%) of diacetate **567** as a colourless oil.

TLC: R_f 0.29 (hexane/EtOAc 5:2, CPS).

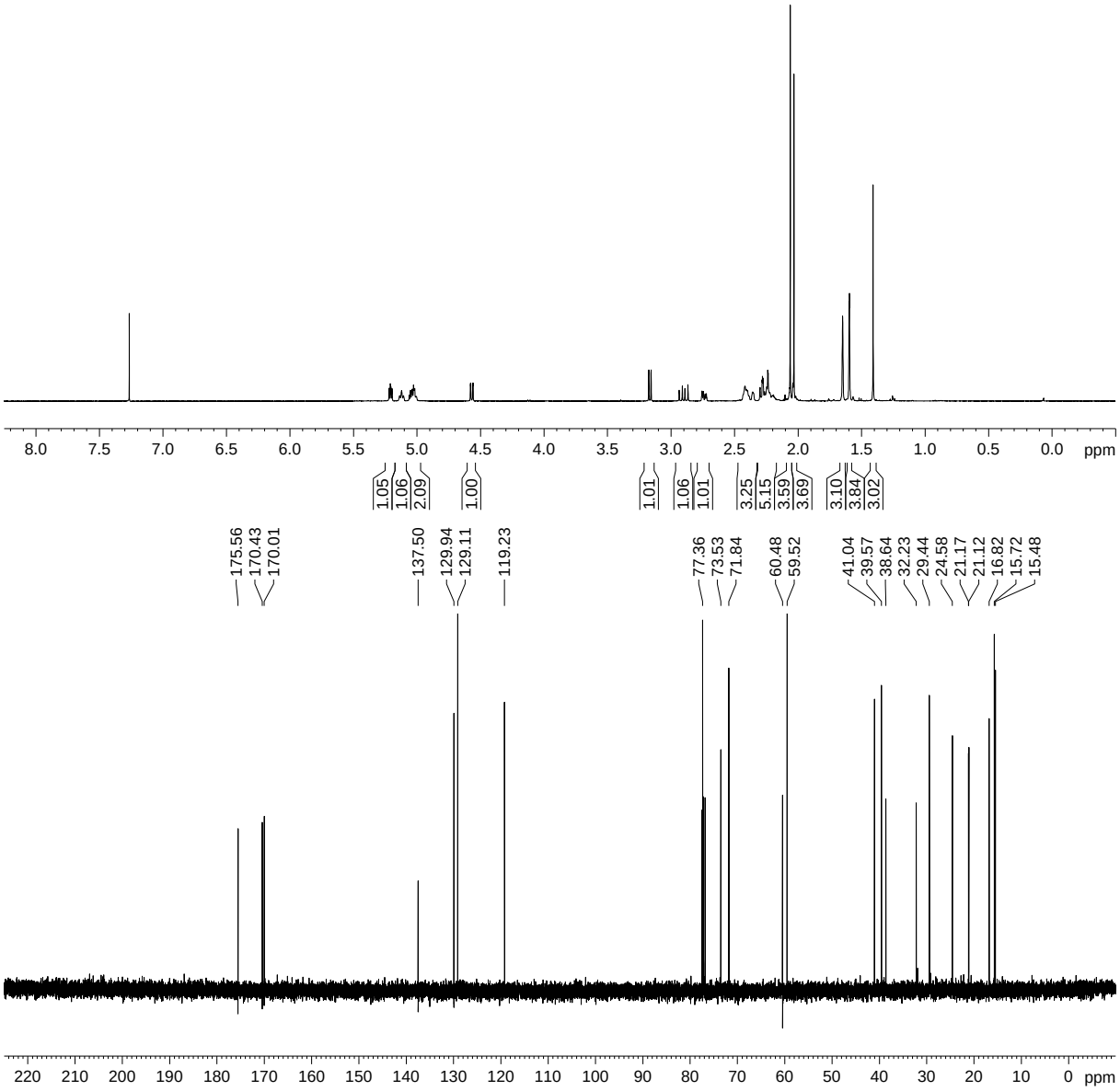
$[\alpha]^{20}_D$: = -61.4 (*c* = 0.67, $CHCl_3$).

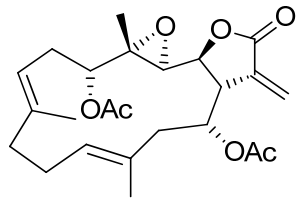
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.21 (t, *J* = 4.0 Hz, 1H), 5.12 (t, *J* = 5.8 Hz, 1H), 5.06-4.99 (m, 2H), 4.57 (d, *J* = 7.3 Hz, 1H), 3.17 (d, *J* = 7.3 Hz, 1H), 2.90 (dd, *J* = 17.8, 9.7 Hz, 1H), 2.74 (dd, *J* = 9.6, 4.1 Hz, 1H), 2.46-2.34 (m, 3H), 2.30-2.15 (m, 5H), 2.06 (s, 3H), 2.06-2.02 (m, 1H), 2.03 (s, 3H), 1.65 (s, 3H), 1.59 (s, 3H), 1.41 (s, 3H).

^{13}C-NMR (100 MHz, $CDCl_3$): δ = 175.6, 170.4, 170.0, 137.5, 129.9, 129.1, 119.2, 77.4, 73.5, 71.8, 60.5, 59.5, 41.0, 39.6, 38.6, 32.2, 29.4, 24.6, 21.2, 21.1, 16.8, 15.7, 15.5.

IR (film): ν 2933, 2856, 1780, 1739, 1429, 1376, 1231, 1174, 1080, 1042, 981, 934, 934, 857, 829, 755.

HRMS (ESI): calculated for $C_{23}H_{32}O_7$ $[M+Na]^+$ 421.2221, found 421.2225.





5

(1a*S*,2*R*,4*E*,8*E*,11*R*,11a*R*,14a*S*,14b*S*)-1a,5,9-trimethyl-12-methylene-13-oxo-1a,2,3,6,7,10,11,11a,12,13,14a,14b-dodecahydrooxireno[2',3':13,14]cyclotetradeca[1,2-b]furan-2,11-diyl diacetate (5). To a solution of diacetate **567** (7.4 mg, 0.018 mmol) in THF (0.5 mL) was added added LiHMDS (21.1 uL, 0.021 mmolat -78 °C, the reaction mixture was allowed to reach 0 °C and was stirred for 15 min at this temperature. The reaction mixture was then cooled to -78 °C and Eschenmoser salt (9.8 mg, 0.053 mmol) was added in one

portion. The reaction mixture was allowed to reach 0 °C over a period of 1.5 hours. The reaction was then cautiously quenched by of saturated aqueous NH_4Cl (3 mL). The phases were separated and the aqueous phase was extracted with Et_2O (3 x 3 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. The residue was dissolved in MeOH (0.5 mL) and MeI (3.3 uL, 0.053 mmol) was added at 0 °C. The reaction mixture was then allowed to reach room temperature and 0.5 hours. The reaction was cautiously quenched by addition of saturated aqueous $NaHCO_3$ (1 mL) and CH_2Cl_2 (2 mL) was added. The phases were separated and the aqueous phase was extracted with CH_2Cl_2 (3 x 2 mL). The combined organic phases were dried over $MgSO_4$, concentrated under reduced pressure and the residue was purified by flash column chromatography (hexane/EtOAc 3:1) to yield 3.0 mg (40%, 49% brsm) of enone **5** as a colourless oil.

TLC: R_f 0.35 (hexane/EtOAc 5:2, CPS).

$[\alpha]^{20}_D$: = -8.8 (*c* = 0.067, $CHCl_3$).

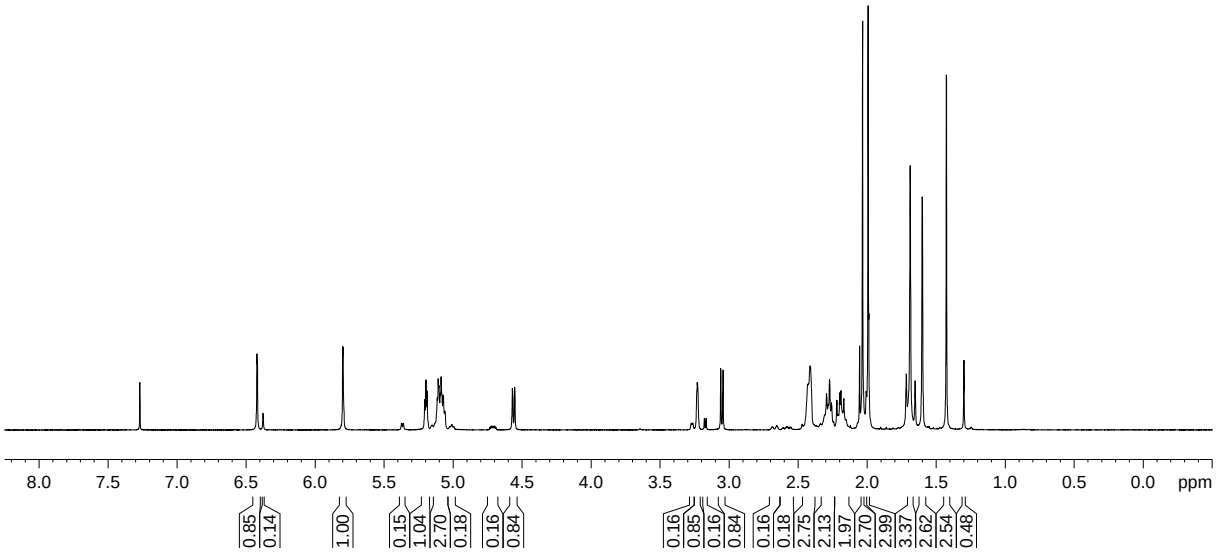
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.42 (d, *J* = 1.5 Hz, 1H), 5.80 (d, *J* = 1.5 Hz, 1H), 5.20 (t, *J* = 4.6 Hz, 1H), 5.13-5.05 (m, 3H), 4.56 (d, *J* = 8.3 Hz, 1H), 3.23 (q, *J* = 1.5 Hz, 1H), 3.05 (d, *J* = 8.3 Hz, 1H), 2.45-2.40 (m, 3H), 2.32-2.25 (m, 2H), 2.22-2.15 (m, 1H), 2.19 (dd, *J* = 14.8, 10.8 Hz, 1H), 2.03 (s, 3H), 2.01-1.97 (m, 1H), 1.99 (s, 3H), 1.69 (s, 3H), 1.60 (s, 3H), 1.43 (s, 3H).

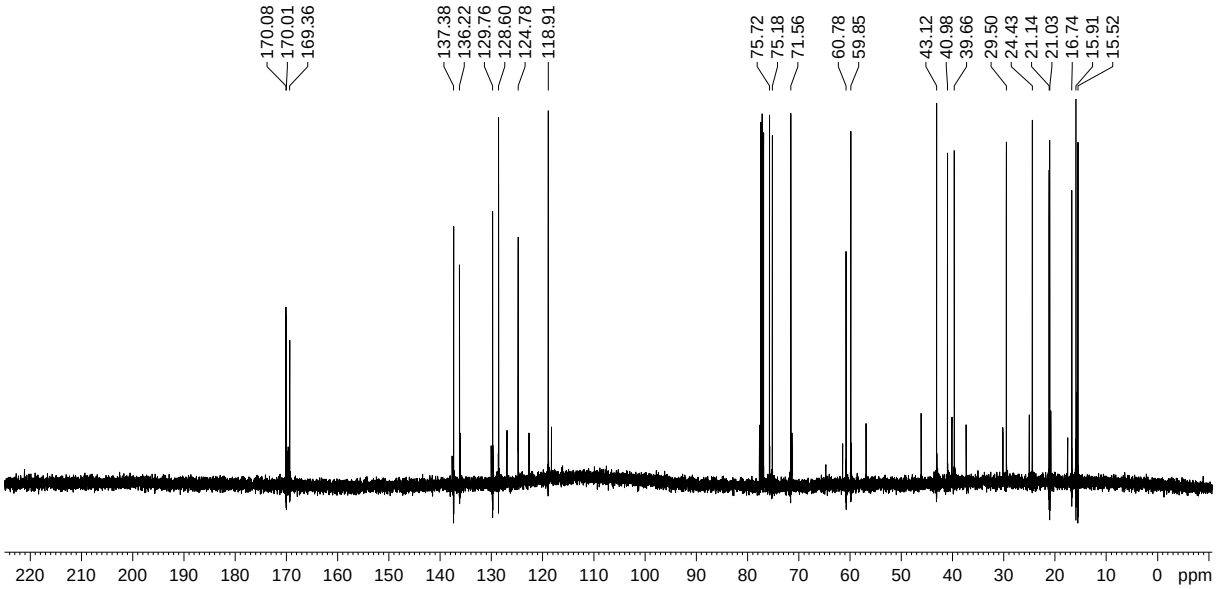
^{13}C-NMR (100 MHz, $CDCl_3$): δ = 170.1, 170.0, 169.4, 137.4, 136.2, 129.8, 128.6, 124.8, 118.9, 75.7, 75.2, 71.6, 60.8, 59.8, 43.1, 41.0, 39.7, 29.5, 24.4, 21.1, 21.0, 16.7, 15.9, 15.5.

IR (film): ν 2927, 1771, 1744, 1651, 1555, 1539, 1511, 1454, 1374, 1229, 1038, 772.

HRMS (ESI): calculated for $C_{24}H_{32}O_7$ $[M+H]^+$ 433.2221, found 433.2224.

HPLC: 0-2: 40%, 2-10: 40-70%, 10-12: 70-90%, Ref: 10.17 Min.

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