A New Total Synthetic Pathway to Cryptophycin-3 and an Analogue as well as an Efficient Approach to the Total Synthesis of the Proteasome Inhibitor Epoxomicin

Ein neuer kompletter Syntheseweg für Cryptophycin-3 und ein Analogon sowie ein effizienter Zugang zur Totalsynthese des Proteasom Inhibitors Epoxomicin

DISSERTATION

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ABBREVIATIONS

abs.	absolute
Ac	Acetyl
arom.	aromatic
BBN (9-)	9-Borabicyclo[3.3.1]nonane
Bn	Benzyl
b.p.	boiling poinz
br	broad (NMR)
Bu	Butyl
*t*Bu	*tertiary* Butyl
Boc	*tertiary* Butoxy carbonyl
BuLi	Butyllithium
c	Concentration
COSY	Correlation Spectroscopy
CSA	Camphor sulfonic acid
δ	Chemical shift in ppm (NMR)
d	Doublet (NMR)
DCC	Dicyclohexylcarbodiimide
DCM	Dichloromethane
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
de	Diastereomeric excess
DEPT	Distortionless Enhancement
DIBAL	Diisobutylaluminium hydride
DIEA	Diisopropylethylamine
DMAP	4-Dimethylaminopyridin
DMF	N,N-Dimethylformamid
DMSO	Dimethylsulfoxide
dr	Diastereomeric ratio
E	trans
EDC	1-Ethyl-3-(3'-dimethylaminopropyl)carbodiimide
ee	Enantiomeric excess
EI	Electron impact

eq	equation
equiv	equivalents
ESI	Electrospray ionization
Et	Ethyl
Et_2O	Diethyl ether
EtOAc	Ethyl acetate
Fig.	Figure
Fmoc-Cl	Fluorenylmethoxy-carbonyl chloride
FDPP	Pentafluorophenyl-diphenylphosphinate
g	gram(s)
GC	Gas chromatography
h	hour(s)
HBTU	2-(1H-Benzotriazole-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate
HOBT	Hydroxybenzotriazole
HPLC	high performance liquid chromatography
HRMS	high resolution mass spectrometry
Hz	hertz
imid.	Imidazole
IR	Infrared
i-Pr	isopropyl
J	coupling constant
L	liter(s)
LA	Lewis acid
LAH	Lithium aluminium hydride
LDA	Lithium diisopropylamide
m	Multiplet (NMR)
Me	Methyl
MeOH	Methanol
mg	milligram(s)
m/z	Mass to charge ratio (MS)
NMO	N-Methylmorpholin-N-Oxide
NMR	Nuclear magnetic resonance
PE	Petroleum ether
Ph	Phenyl

PMB	*p*-Methoxybenzyl
PPTS	Pyridinium *para*-toluene sulfonate
Py	Pyridine
PyBoP	Benzotriazole-1-yl-oxy-tris-pyrrolidino-phosphonium hexafluorophosphare
PyBoP	Bromo-tris-pyrrolidino-phosphonium hexafluorophosphate
q	Quartet (NMR)
RCM	Ring closing Metathesis
R_f	Retention factor (TLC)
rt	Room temperature (ca. 23 °C)
s	Singlet (NMR)
s	second
t	Triplet (NMR)
TBAF	Tetrabutylammonium fluoride
TBDMS	*tertiary* Butyldimethylsilyl
TBDPS	*tertiary* Butyldiphenylsilyl
TFA	Trifluoroacetic acid
THF	Tetrahydrofuran
TfO	Trifluoromethanesulfonate
Triflate	Trifluoromethanesulfonate
UV	Ultraviolet
Z	cis

I. INTRODUCTION

The importance of natural products for medicine is undisputed. Since the earliest time mankind has been using extracts from natural sources like plants, animals and fungi for treatment of different diseases without knowing their relation between chemical structures and biological activity. During the development of modern natural sciences it was discovered that many of the earlier used natural extracts contain in fact biological active compounds which can be used as drugs or therapeutics and applied for medical treatment. Quite often natural products serve as biochemical tools or they led to the discovery of a hitherto unknown biochemical pathway or molecular target. Nevertheless, in the context of the human genome project speculations arose that there might be not enough natural products to probe the functions of all genes via selective interaction of natural products with the corresponding proteins.[1] On the other hand natural products offer structurally interesting platforms that might be used to produce natural product-like molecules. This way the chances of discovering novel modes of biological activity might be higher.[2,3]

The search for bioactive natural compounds from marine organisms is still a relatively new field because of the difficulties involved in collecting samples and the fact that there is virtually no ethnobotanical data to draw on. However the biodiversity of the marine environment exceeds by far that of its terrestrial counterpart, thus the oceans represent a huge resource for new biologically active compounds. Many of these molecules possess unique structural features which are enormous and specific enough to protect the organisms in a very hostile environment. New drug discoveries indicate that marine organisms have a vast potential for new pharmaceuticals and will become a much more profound source than any other group of terrestrial organisms.

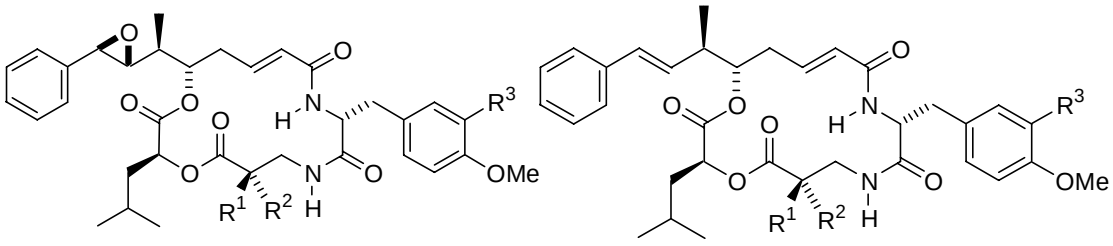
In the present work two topics concerning natural product synthesis are being discussed. The first topic represents a novel convergent synthetic way to a biologically active cyclodepsipeptide, Cryptophycin-3, whereas the second represents the synthesis of an epoxide containing protein-reactive natural compound, Epoxomicin.

Depsipeptides or depsides, *i.e.* heterodic peptides, which contain not only an amide bond but also other types of bonds (usually an ester, *i.e.* a depside bond or a disulphide bridge) as part of their backbone, are another group of peptide derivatives with interesting biologi-

cal properties. Although their occurrence and pharmacological values are incomparable with those of peptides, they still represent an interesting target for drug research and development.

Cyclodepsipeptides are cyclic depsipeptides containing at least one hydroxy acid. In addition cyclodepsipeptides often also contain unusual amino acids, such as extended, *N*-methylated, hydroxylated, or halogenated ones. The hydroxy acids are frequently derived from the polyketide pathway, so many cyclodepsipeptides are macrocyclic hybrid peptide/polyketide-like molecules. Compounds of this type have quite recently been accessed by a chemoenzymatic route.[4] Thanks to their modular natures and the presence of peptide subunits, cyclodepsipeptides are ideal lead compounds for the generation of libraries of natural product-like compounds or compound collections of interest in the context of chemical genetics and target discovery.[5,6,7] One can also argue that an organism, by producing cyclodepsipeptides, has a flexible tool available for responding to a possible hostile environment by simple combination of post-translationally modified amino acids and one larger building block from another biosynthetic pathway. A quite prominent family of cyclodepsipeptides are the Cryptophycins.

The first representative of these cyclic depsipeptides, Cryptophycin-1 (**1**) (Figure 1), was isolated in 1990 from a marine blue-green algae.[8] Some later, the isolation and structure of Cryptophycin-24 (**3**), also known as arenastatin A, was described.[9] Initially Cryptophycin-1 was classified as an antifungal agent, but it was later discovered to have powerful antitumor activity caused by disruption of microtubule assembly.[10] It blocks the cell cycle at the G2/M phase apparently through inhibition of tubulin polymerization into microtubules.[11,12] Its extreme potency has led to additional studies investigating other possible modes of action.[12,13,14] Of particular interest was the fact that Cryptophycin showed activity against some multiple drug resistance (MDR) cell lines. Because of their interesting biological activities and challenging molecular structures, the total synthesis of Cryptophycin-3 (**5**) and its an analogue has been reported.



	R^1	R^2	R^3
Cryptophycin 1 (**1**)	Me	H	Cl
Cryptophycin 2 (**2**)	Me	H	H
Cryptophycin 24 (**3**)	H	H	H
Cryptophycin 52 (**4**)	Me	Me	Cl

	R^1	R^2	R^3
Cryptophycin 3 (**5**)	Me	H	Cl
Cryptophycin 4 (**6**)	Me	H	H

Figure 1: Important structures of Cryptophycins

The sequencing of the human genome has transformed the way in which scientists think about biology. However there is a vast gulf between the wealth of genome sequence information and our knowledge of genome function. Researchers are now confronting the task of understanding the cellular and molecular functions of thousands of predicted gene products. The genome gives rise to the proteome, and the combinatorial interactions among proteins make living organisms so complex at the molecular level. To understand these interactions the development of new technologies and concepts to expedite global analysis of protein function is required.[15] A growing number of research laboratories are exploring biology with chemistry-based strategies that are capable of yielding insight into the role of individual proteins in complex biological systems. The field of chemical synthesis often plays a major role in this process, perhaps most visible through simplification in identifying protein targets of bioactive natural products.[16]

Nature engages protein targets with reactive small molecules in many ways, and the number of natural products that covalently modifies proteins is likely to be very large. A provocative subset of biologically active natural products is endowed with electrophilic functional groups that covalently modify nucleophilic residues in specific protein targets. Lipstatin[17] and fumagillin[18] embody the chemistry of the carbonyl group and the epoxide respectively and they are prominent examples of protein-reactive natural products (Figure 2). These protein reactive natural products are highly attractive as molecular probes for protein activity profiling experiments because they provide information about enzyme active sites in complex proteomes.[19]

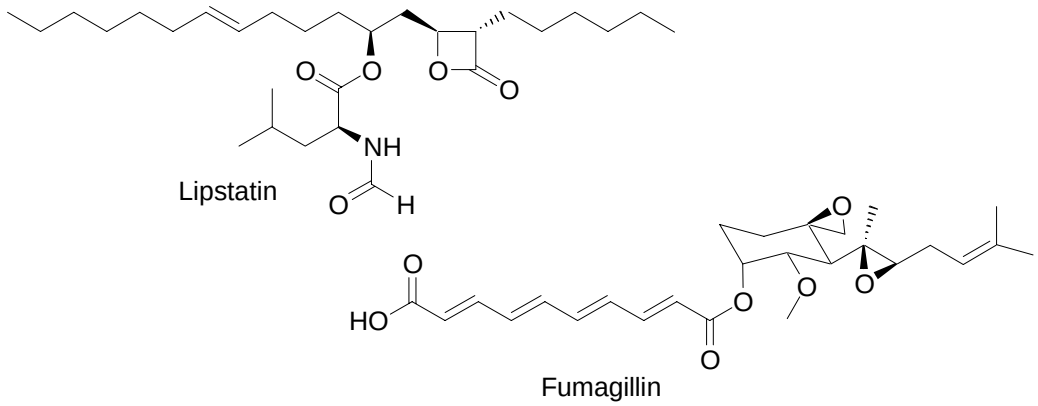


Figure 2: The structures of Lipstatin and Fumagillin

Numerous biologically active natural products that mediate their activities via inhibition of specific intracellular protein targets have been isolated. The second part of this work represents the synthesis of Epoxomicin (**7**) (Figure 3), a peptidyl epoxide natural product that acts as a covalent inhibitor of the proteasome.[20]

Epoxomicin was isolated based on its potent in vivo antitumor activity against solid tumors derived from B16 melanoma.[21] Epoxomicin belongs to a small family of linear peptides, of which all share two structural features: a threonine or serine residue and an α,β-epoxy-ketone derived from leucine or γ,δ-dehydroleucine.

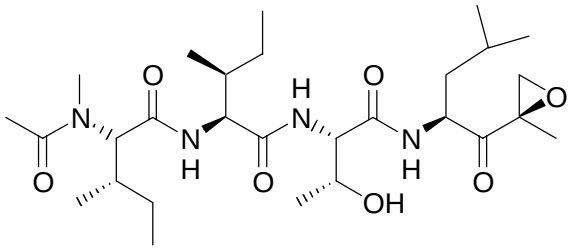


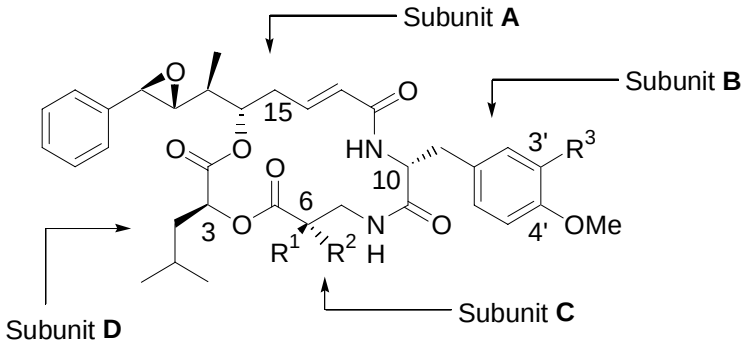
Figure 3: The structure of Epoxomicin (**7**)

These α,β-epoxyketone containing peptides exert their biological activities via adduct formation with intracellular target proteins since the reduction of this reactive epoxide results in loss of biological activity.[22]

II. THE FAMILY OF CRYPTOPHYCINS

1. Biological activity of Cryptophycins

The Cryptophycins are potent tumor-selective cytotoxins, isolated from blue-green algae *Nostoc sp.* ATCC 53789 and *Nostoc sp*.SV 224.[8] When Cryptophycin-1 was first isolated by Schwartz and co-workers, they chose the name Cryptophycin because of the compound's highly potent fungicidal activity against filamentous fungi and yeast of the genus *Cryptococcus*. Because of the narrow range of anti-fungal activity of Cryptophycin-1 (**1**) and its low therapeutic index,[23] the development of Cryptophycin-1 as an anti-fungal compound was not pursued. Cryptophycin-1 was later isolated by Moore and co-workers, who screened over 1000 extracts of blue-green algae in search of compounds that exhibited antitumor activity.[8b] Cryptophycin-1, the most abundant cytotoxin of the Cryptophycins,[24] has potent activity in the pM range in the multi-drug-resistant (MDR) cell lines SKVLB1 and MCF-7/ADR, [25] thereby surpassing vinblastine, paclitaxel, and colchicine in the ability to retain cytotoxic activity in MDR cell lines.



Cryptophycin-1 (**1**) R^1=Me, R^2=H, R^3=Cl

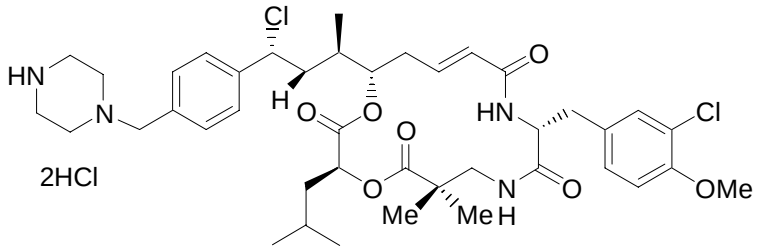
Cryptophycin-24 (**3**) R^1=H, R^2=H, R^3=H

Cryptophycin-52 (**4**) R1=Me, R2=Me, R3=Cl

Figure 4: Important biologically active Cryptophycins

A structurally simpler Cryptophycin, Cryptophycin-24 (arenastatin A **3**), was isolated from an Okinawan marine sponge, *Dysidea arenaria*.[26] Although Cryptophycin-24 lacks the chlorine substituent on the aryl ring of the C10 side chain and the methyl group at C6, it is extremely potent against KB cells (Figure 4).[27] Unfortunately, Cryptophycin-24 has a

half-life of approximately 10 min in mouse serum due to ester hydrolysis.[28] Therefore Cryptophycin-52 (**4**), the C6 *gem*dimethyl analogue of Cryptophycin-1, was prepared to decrease the rate of ester hydrolysis in vivo.[29,30] This synthetic analogue is the first Cryptophycin to undergo clinical trials for the treatment of cancer.[31] Out of around 450 analogues, Cryptophycin-52 has emerged as a promising clinical candidate, advancing even to phase 2 clinical studies. Table 1 shows IC_{50} values for some representative Cryptophycines. [32]



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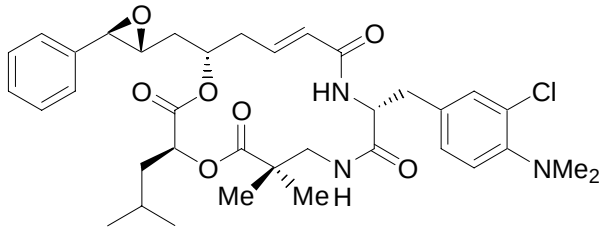

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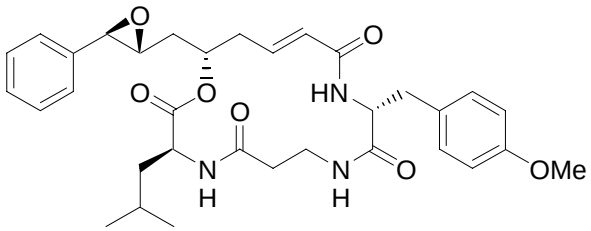

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Figure 5: Structure of some other important Cryptophycin analogues

The Cryptophycins are microtubule-destabilizing agents that elicit cytotoxic effects by binding noncovalently to a site at, or overlapping, the *Vinca* site on tubulin at the rhizoxin/ maytansine site.[25,27,33-35] Cryptophycin-1 also induces the phosphorylation of Bcl-2[36] and activates the apoptosis process.[12,37] Similar to other known antimitotic agents,[38] Cryptophycin-1 slows microtubule dynamics at low concentrations[10,13] while at subnanomolar concentrations it causes a complete loss of microtubule within cells. Through its action on

microtubules and possibly other yet to be discovered targets, Cryptophycin induces potent cytotoxicity by activating the apoptotic pathway.[12,37]

Table 1: [a] Data from references: (compounds 1,2,3,5,10)[8h], (compounds 4,8)[30], (compound 6)[8b], (compound 9)[29]
KB = human nasopharyngeal carcinoma cell line
CCRF-CEM = human leukemia cell line

Compound	IC_{50} [nM][a]	Cell type
1	0.0092	KB
2	0.057	KB
3	0.198	KB
4	0.022	CCRF-CEM
5	3.23	KB
6	2.15	KB
8	0.021	CCRF-CEM
9	0.054	CCRF-CEM
10	6.0	KB

One of the strong features of Cryptophycin is that it lacks cross resistance with paclitaxel, doxorubicin,[39] and other families of drugs such as DNA alkylators, topoisomerase II inhibitors, and antimetabolites[40] which are generally used in combination protocols for the treatment of cancer. Cryptophycin-1 has also been found to exhibit excellent antiproliferative activity in vivo against five solid tumors of murine origin (i.e. colon adenocarcinomas 38 and 51, taxol-sensitive and taxol-resistant mammary adenocarcinoma M16, and pancreatic ductal adenocarcinoma) that were subcutaneously transplanted into mice.[8b,41] In vitro, Cryptophycin-1 was shown to be highly toxic to the human nasopharyngeal carcinoma and the human colorectal adenocarcinoma cell lines.[8b]

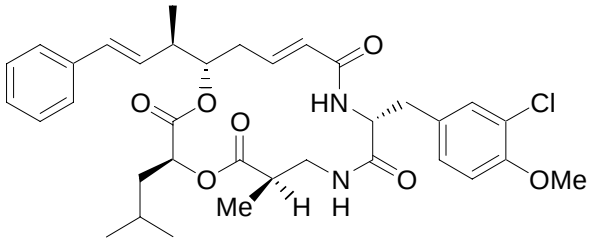
2. Structure-activity relationships of Cryptophycins

Structure-activity relationship (SAR) studies of the C10 side chain of Cryptophycin-1 demonstrated that introduction of a second chloro substituent at the C5′ position of the C10 side chain decreased activity by a factor of 120, while absence of the C3′-Cl decreased activity by a factor of 10.[25] A factor of 30 decrease inactivity was observed when the C4′-*O*-methyl group was absent.[41] These data suggested that the 3′-C1 and 4′-MeO are required for optimal activity. More recently, it was revealed that the cytotoxicity of Cryptophycin can be more dramatically affected by altering the C10 side chain.

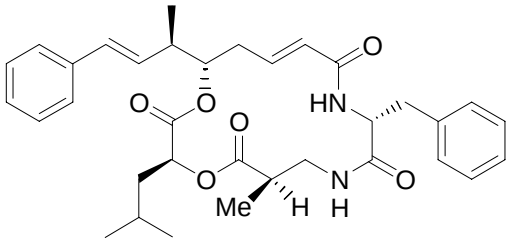
Patel and co-workers prepared analogues of Cryptophycin-52 (**4**) that were tested in the human leukemia CCRF-CEM tumor cell line.[31] Substitutions for the benzyl moiety and alternative substitution patterns on the aromatic ring were also made. Substitution of the C10 aryl ring with a naphthyl ring, altering the tether length between the macrocycle and the aryl ring, removing the aromatic ring, or replacing it with a cyclohexyl group, led to analogues with varying degrees of reduced activity. Variations in the substitution pattern of the benzyl ring of the Cryptophycin-52 analogues revealed that the 3′-Cl substituent provides an increased in vitro activity, whereas compounds lacking the 3′-Cl substituent were active only in the 4 to 120 nM range with the exception of the analogue containing an unsubstituted benzyl ring at C10 which was active and had an ED_{50} value of 183 pM. The compound with the most activity was the 3′-C1, 4′-NMe_2 analogue **9**, which had an IC_{50} value 2.5 times that of Cryptophycin-52 (Figure 5).[31]

The structure activity studies also showed that variations in the fragment A epoxide are well tolerated, revealing even compounds that are active against murine Panc-03 tumors at much lower doses.[31] The chlorohydrin **8** is one example. Compounds, such as **5** or **6**, that lack the epoxide ring still are quite active which shows that there is no covalent bond upon binding to the tubulin. The triamide **10** is characterized by poor solubility and low bio-availability but still shows reasonable activity.

The SAR determined from isolated analogues of Cryptophycin-1 and synthesized Cryptophycin analogues of Cryptophycin-52 suggested that any analogue synthesized should retain the following on the C10 side chain for optimal activity: (1) the *R* stereochemistry at C10, (2) a benzyl moiety, and (3) a chloro substitution at the 3′ (meta) position. Therefore we have chosen to synthesize Cryptophycin-3 (**5**) and its an analouge(**11**)(Figure 6), they both contain the side at C10 with benzyl moiety and *R* stereochemistry.



Cryptophycin-3 (**3**)



analogue **11**

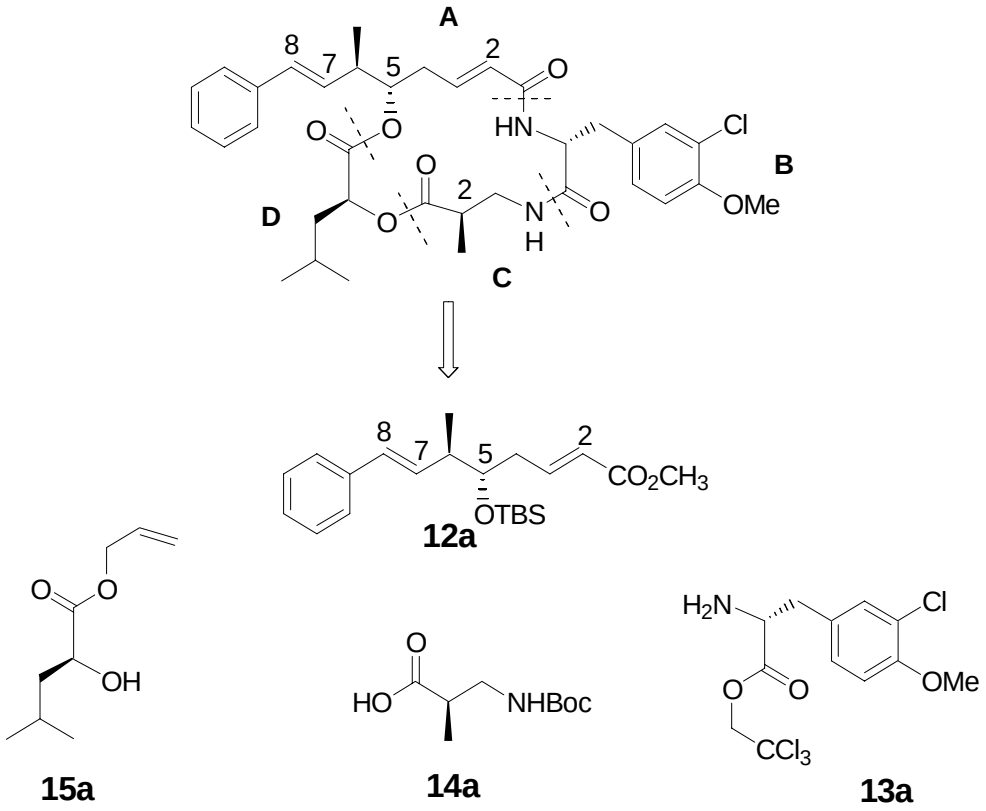
Figure 6: Structure of Cryptophycin-3 (**5**) and analouge **11**

While the in-vitro and in-vivo studies with Cryptophycin-52 were very promising, the clinical studies revealed significant neurological toxicity and only weak or no therapeutic response. Nevertheless, from a chemical point of view the ω-hydroxy acid of Cryptophycin should be an interesting building block allowing to restrict the conformation of tri- and tetrapeptides that are inserted in between the hydroxyl and carboxyl function.

3. Previous total and formal syntheses of Cryptophycins

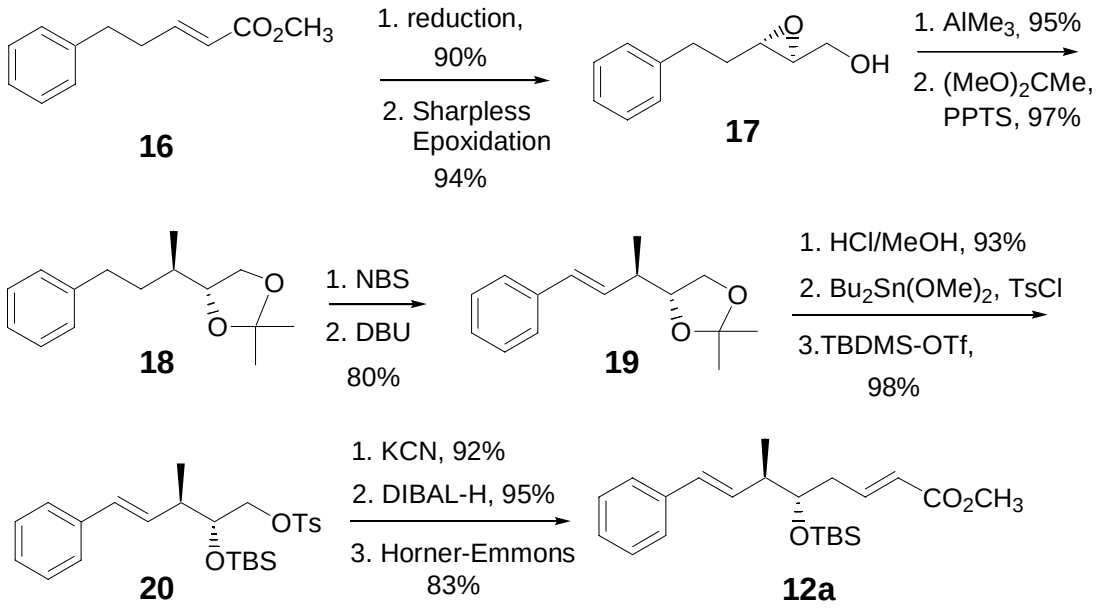
3.1 The first Tius synthesis

The Tius group was first to publish the relative and absolute stereochemistry for the crytophycins in 1994.[24] In 1995 they synthesized and corrected the structures of Cryptophycin-1 and -3. For the purposes of retrosynthetic analysis, Cryptophycin-1 was disconnected into four units (Scheme 1). Unit A **12a** is a polyketide-derived molecular fragment, unit B **13a** is an O-methyl-D-chlorotyrosine derivative, whereas units C **14a** and D **15a** correspond to (*R*)-3-amino-2-methyl-propanoic acid and (*S*)-2-hydroxy-4-methylvaleric (L-leucic) acid derivative. The macrocyclisation occurs by amide bond formation between unit B and unit C.



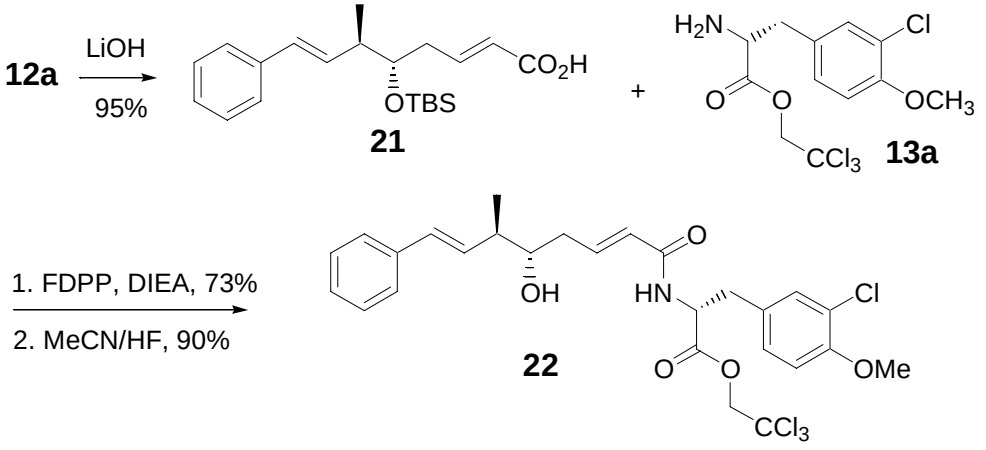
Scheme 1: Retrosynthetic analysis by Tius group

Scheme 2 summarizes the first successful approch to unit A, **12a**. In this synthesis compound **16** was prepared from commercially available dihydrocinnamaldehyde and trimethyl-phosphonoacetate in the presence of tetramethylguanidine. Reduction with DIBAL-H and Sharpless asymmetric epoxydation using L-(+)-diethyl tartrate of **16** led to epoxy alcohol **17**. Regio- and stereo-specific epoxide ring opening with trimethyl aluminium resulted in the desired diol which was protected as acetonide **18**. Benzylic bromination with NBS led to an unstable bromide which was immediately dehydrobrominated with DBU and became styrene **19**. But in a large scale reaction **19** was eroded by the formation of diastereomeric tetrahydrofuryl alcohol. Hydrolysis of the acetonide group in **19** with aqueous methanolic HCl led to diol on which the primary OH was selectively protected as tosylate by Ley's procedure[42] and secondary OH was protected as TBS ether and led to **20**. Displacement of tosylate by cyanide was followed by the reduction with DIBAL-H generating thecorresponding aldehyde, which on Horner-Emmons homologation with trimethylphosphonoacetate led to **12a**.



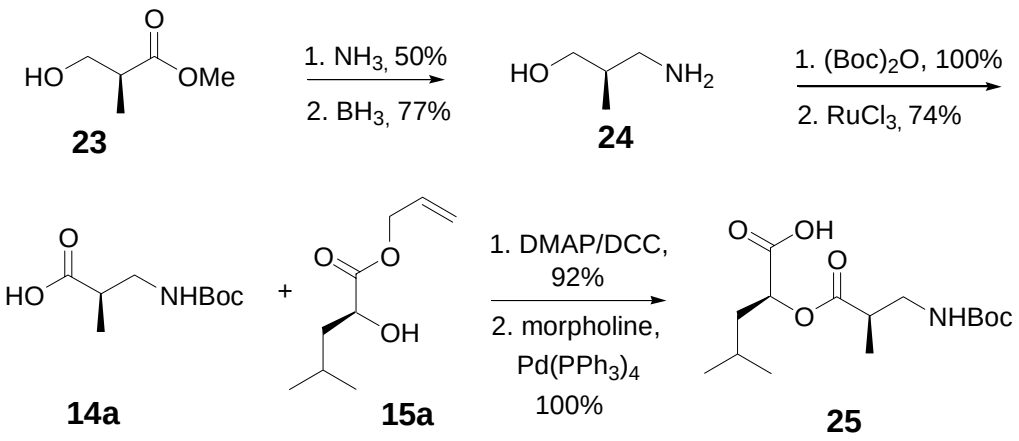
Scheme 2: The synthetic pathway to unit A **12a**

Scheme 3 shows the assembly of unit A and unit B. Before coupling, the methyl ester of **12a** was hydrolysed to acid **21** with lithium hydroxide and the protected unit B **13a** was prepared from commercially available D-tyrosine in five steps. Chlorination[43] with sulfuryl chloride was followed by protection of amine with BOC. Then methylation of the phenol took place with dimethyl sulfate in presence of potassium carbonate. Hydrolysis of the methyl ester with aqueous NaOH gave back to acid which on reesterrification with 2,2,2-trichloroethanol in presence of DCC and pyridine gave the Troc ester. Cleavage of BOC with trifluoroacetic acid was leading to the trifluoroacetate salt of **13a**. Mixing a solution of **21** in anhydrous DMF with pentafluorophenyl diphenylphosphonate (FDPP), the trifluoroacetate salt of **13a** and DIEA at room temperature led to the amide which after cleavage of the silyl ether protecting group with HF produced **22** in good yield.



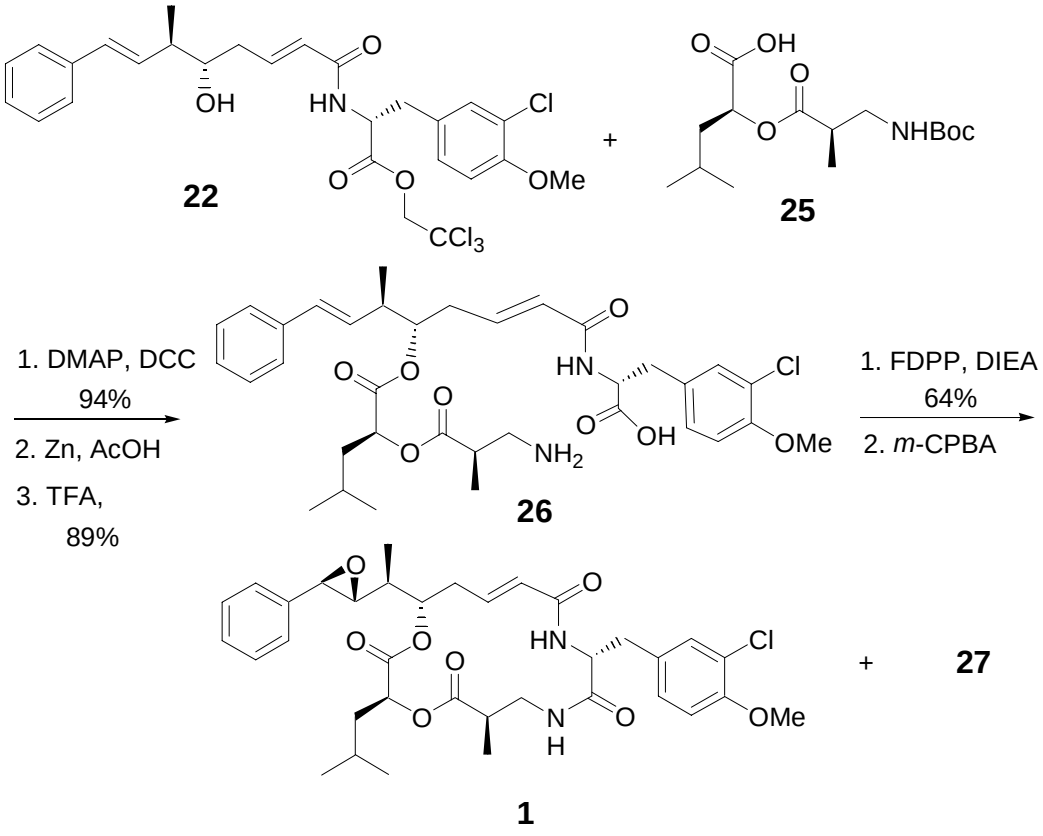
Scheme 3: Coupling of unit A and unit B

The Tius synthesis follows a highly convergent strategy to join unit C **14a** and unit D **15a**, (Scheme 4). The β-amino acid **14a** was prepared from commercially available (*S*)-(+)-3-hydroxy-2-methylpropanoate **23**, which was converted to the amide with ammonia and on reduction with borane led to amine **24**. Protection of the amino group as Boc derivative, followed by oxidation of the primary alcohol with ruthenium tetroxide, led to carboxylic acid **14a**. Commercially available L-leucic acid was converted to allyl ester **15** under phase-transfer conditions by stirring with allyl bromide and tetra-*n*-butylammonium chloride. The coupling of **14a** with **15a** was accomplished with DCC and DMAP to produce a dipeptide analogue which on deprotection with morpholine and catalytic $Pd(PPh_3)_4$[44] led to **25**.



Scheme 4: Coupling of unit C and unit D

The ester linkage between **22** and **25** was installed through the use of DCC/DMAP to produce a seco compound (Scheme 5), which the sequential removal of Troc and Boc protecting groups occured on by treatment with zinc and acetic acid, followed by neat trifluoro-acetic acid and providing **26**. Macrolactamization of **26** with FDPP in presence of DIEA in DMF gave Cryptophycin-3 (**5**) in 64% yield. Epoxidation with *m*-CPBA led to a ca. 2/1 mixture of Cryptophycin-1 (**1**) and the corresponding (7*S*,8*S*)-*trans*-epoxide **27**.

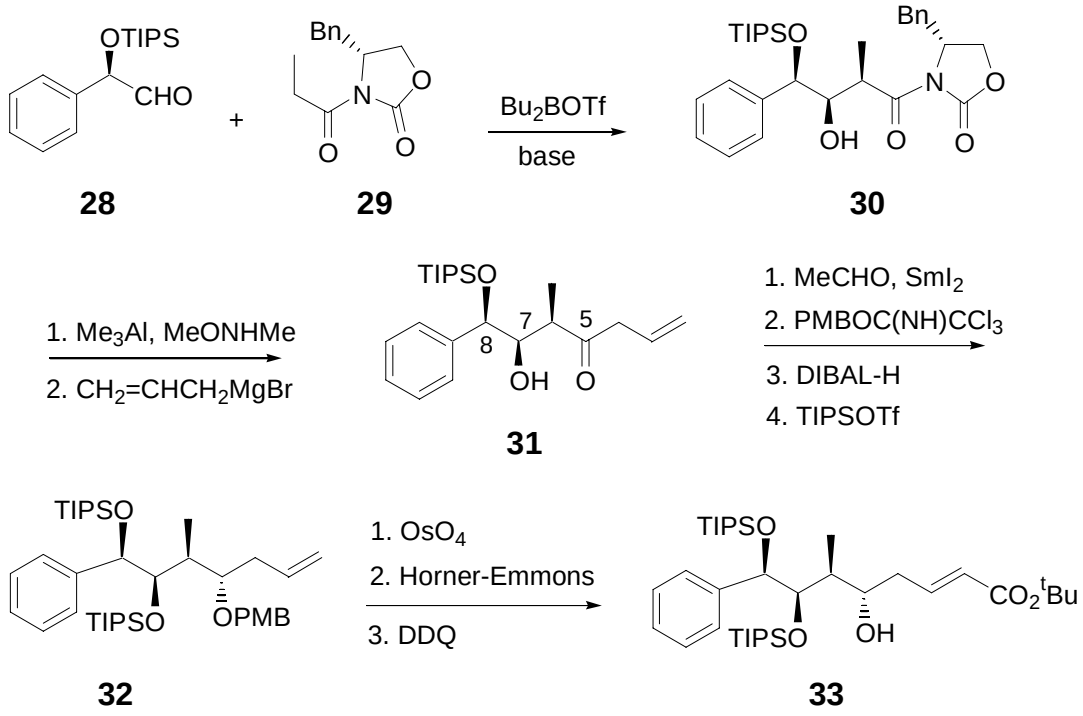


Scheme 5: Coupling of unit C + D and unit A + B

3.2 The Leahy-Gardinier synthesis

The first synthesis to address the stereochemical problem posed by the epoxide was conducted by Leahy and Gardinier.[45] A diol was used as a convenient synthetic equivalent of the epoxide. For the synthesis of unit A (Scheme 6) two asymmetric fragments, (*R*)-mandelaldehyde derivative **28** and the boron enolate derived from Evans chiral imide **29**[46] were coupled and led to a single product **30**. Trimethylaluminium-mediated transamidation produced the corresponding Weinreb amide, which on treatment with allylmagnesium bromide provided allyl ketone **31**. Exposure of **31** to acetaldehyde and samarium iodide reduced the C5 carbonyl group to alcohol and at the same time converted C7 hydroxyl to corresponding acetate.[47] Protection of the C5 OH as p-methoxybenzyl (PMB) ether was followed by reductive cleavage of the acetete with DIBAL-H and protection as a TIPS derivative of C7 to give **32**. The conversion of **32** to **33** was accomplished by oxidative cleavage of the terminal alkene to the aldehyde, followed by Horner–Emmons homologation using Masamune-Roush conditions and oxidative cleavage of the PMB protecting group with DDQ.

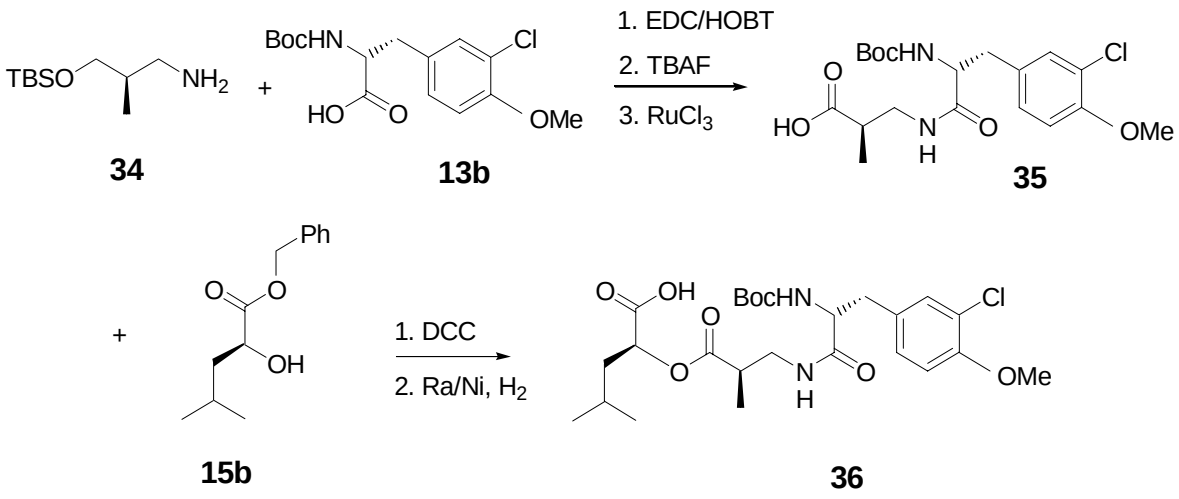
Leahy and Gardinier followed a rather different strategy for the assembly of units B, C, and D (Scheme 7). Protected aminoalcohol **34** was prepared from the reduction of known amide.[48]



Scheme 6: Leahy-Gardiner's synthesis of unit A

The amine **34** was then coupled with acid unit B **13b** in presence of HOBT and EDC. Deprotection of TBS with TBAF and oxidation of primary the alcohol function with ruthenium tetroxide resulted in unit B + C compound **35**. After deprotection coupling of **35** with the benzyl ester of L-leucic acid, unit D **15b** took place with DCC. A final cleavage of benzyl ester with Raney nickel led to unit B + C + D compound **36** in good yield.

The coupling of unit A **33** with unit B+C+D fragment, **36** under Yamaguchi conditions produced **37** (Scheme 8).[49] Cleavage of Boc and *tert*-butyl ester protecting groups took place in a single operation, by exposure of **37** to HCl without disturbing the silyl ether protecting groups. Macrolactamization was carried out with *O*-benzotriazole-1-yl-N,N,N`,N`-bis-(pentamethylene)uronium hexafluorophosphate[50] in good yield and finally after deprotection of the silyl protecting groups the cyclic compound **38** was obtained.



Scheme 7: Leahy- Gardiner's assembly of tripeptide

For the formation of epoxide the Leahy and Gardinier developed a very ingenious modification of the Sharpless method. 4-Azido-1,1,1-trimethoxybutane, prepared from commercially available 4-chlorobutyronitrile in two steps, was allowed to react with diol **38** in presence of trimethylchlorosilane according to Sharpless' conditions and led to **39**. Exposure of **39** to triphenylphosphine and water generated to the C7-C8 chlorohydrin which on reaction with potassium carbonate led to Cryptophycin-1 (**1**) in overall good yield.

3.3 The Tius-Li synthesis

In this synthesis (*R*) Mandelic acid is an attractive starting material as a *sole* source of asymmetry for unit A (Scheme 9). [51] (*R*)-Methyl mandelate (**40**) was converted to the *O*-ethoxyethyl derivative and then reduction of the ester group with DIBAL gave protected mandelaldehyde **41**, which was combined with diene **42** by a hetero Diels-Alder reaction.[52] Exposure of the crude Diels-Alder **43** product to trifluoroacetic acid, followed by workup, gave a 10/1 mixture of **44** and **45**. Stirring a solution of this product mixture over KF/alumina[53] epimerized **44** to **45** as a 1/4 mixture.

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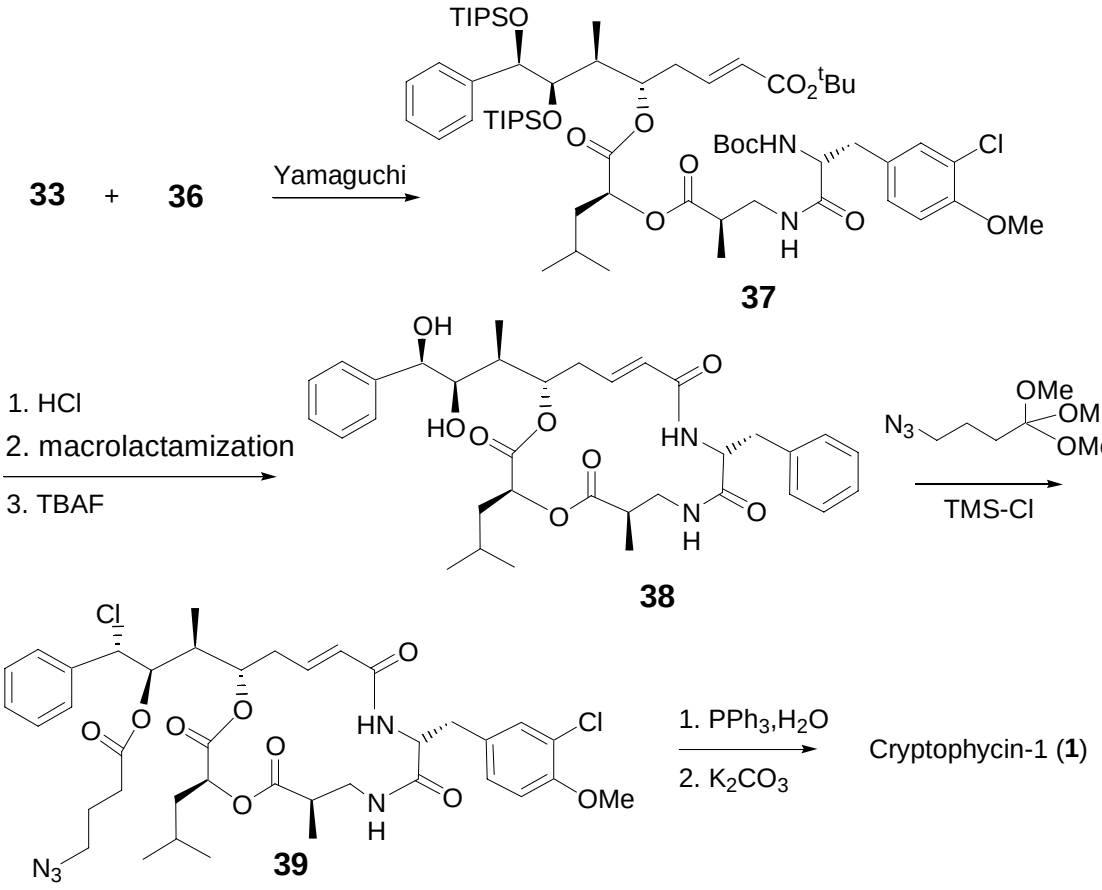
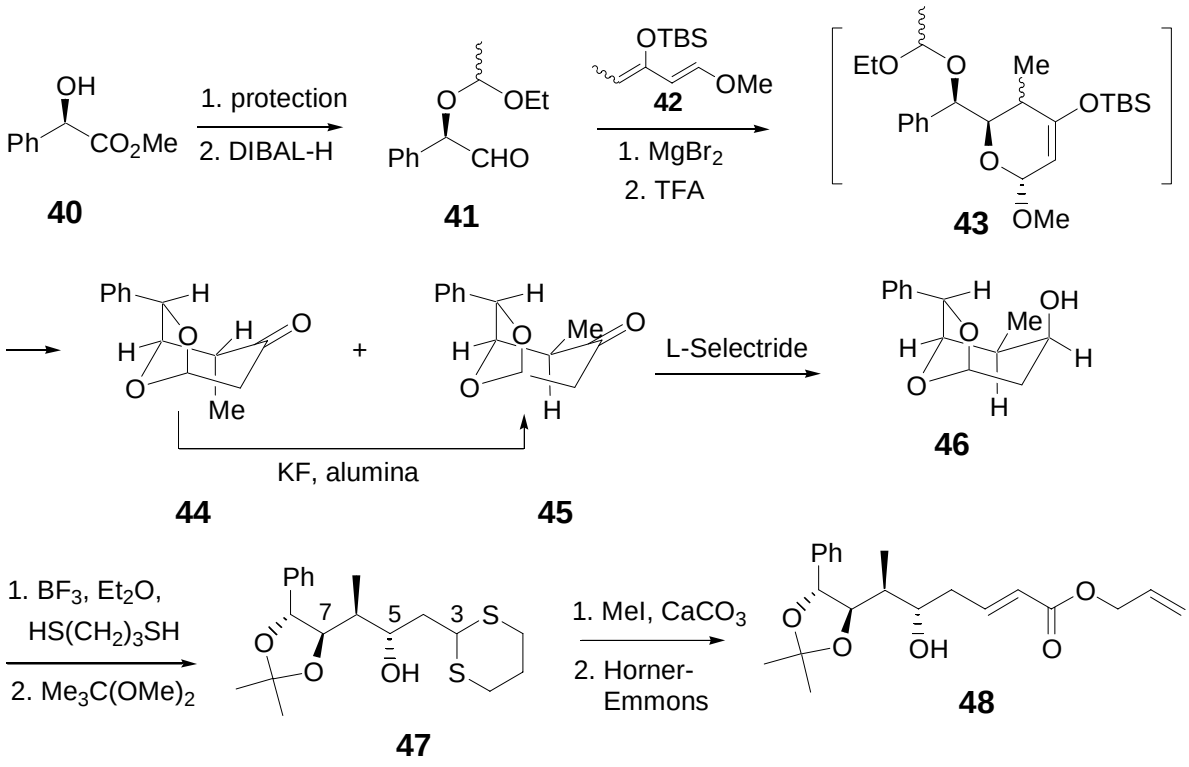


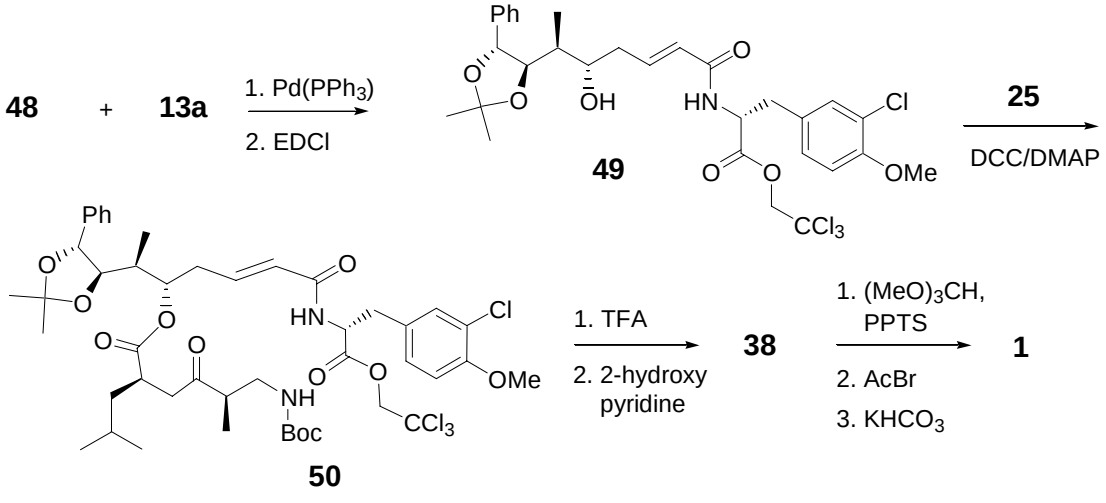
Scheme 8: Leahy-Gardiner's final steps of synthesis of Cyrptophycin-1

Reduction of the 1/4 mixture with L-selectride gave alcohol **46** as a single isomer. Reaction of **46** with boron trifluoride etherate and 1,3-propanedithiol converted the mixed acetal todithioketal. ubsequent proctection of the syn C7-C8 diol as acetonide gave **47**. Treatment of this dithioketal with iodomethane and calcium carbonate led to the β-hydroxyaldehyde, which on Horner-Emmons reaction with allyldiethylphosphonoacetate led to unit A **48**. The allyl ester in **48** was cleaved in presence of catalytic palladium(0) and morpholine (Scheme 10)[44] and coupling with unit B compound **13** took place with EDCl and triethylamine to produce **49**. Esterification of the C5 hydroxyl group with unit C + D compound **25** occurred in the presence of DCC and DMAP and led to seco compound **50**.



Scheme 9: The Tius-Li synthesis of **48**

Simultaneous removal of the Boc and acetonide protecting groups in **50** took place with trifluoroacetic acid. The macrolactamization,[50] developed by Eli Lilly Co. was a simple treatment of the seco compound in toluene with 2-hydroxypyridine producing **38**, the same intermediate from Leahy's synthesis. Treatment of **38** with trimethyl orthoformate and PPTS, followed by addition of acetyl bromide led to a sensitive bromohydrin formate, which on exposure to potassium bicarbonate gave Cryptophycin-1 (**1**) in overall good yield.



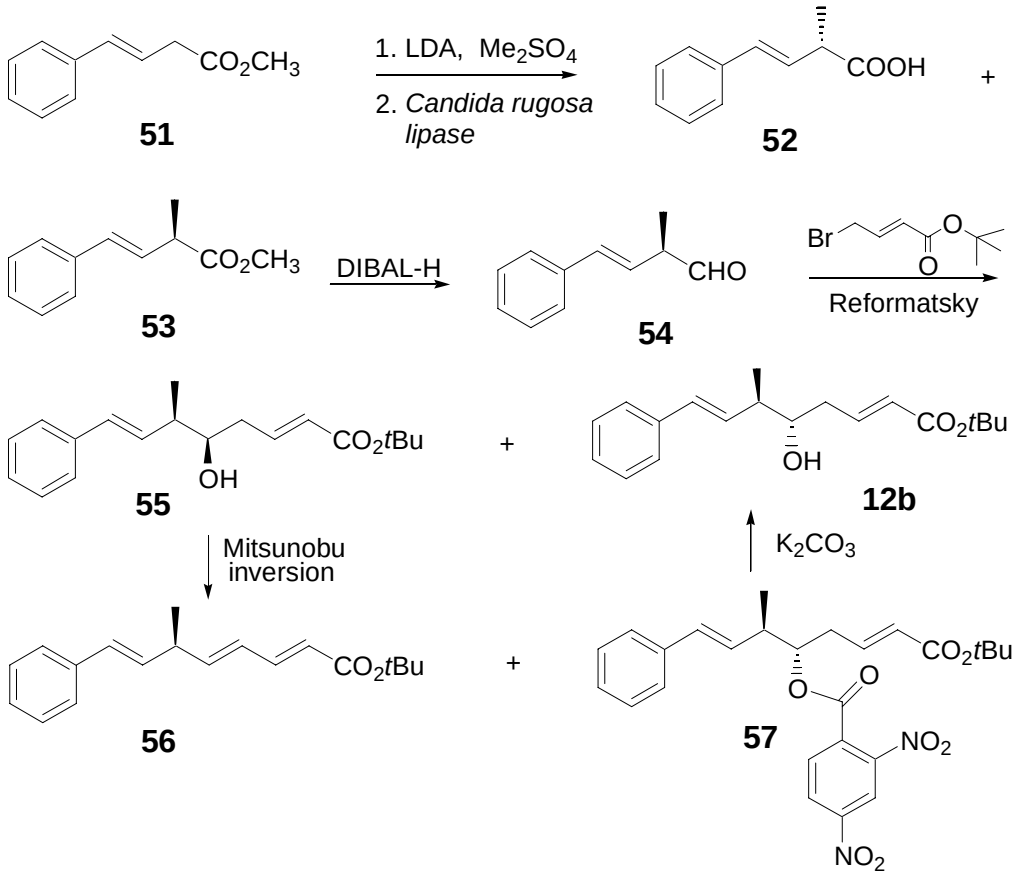
Scheme 10: Tius-Li's final steps of synthesis of Cryptophycin-1

3.4. The synthesis of unit A

Since the subunit A is the most challening from a synthetic point of view, the following section summarizes various synthesis of this hydroxy acid. These synthesis illustrate various ways to establish the *anti*-OH / Me vicinal stereochemical pattern.

3.4.1 The Sih chemoenzymatic synthesis

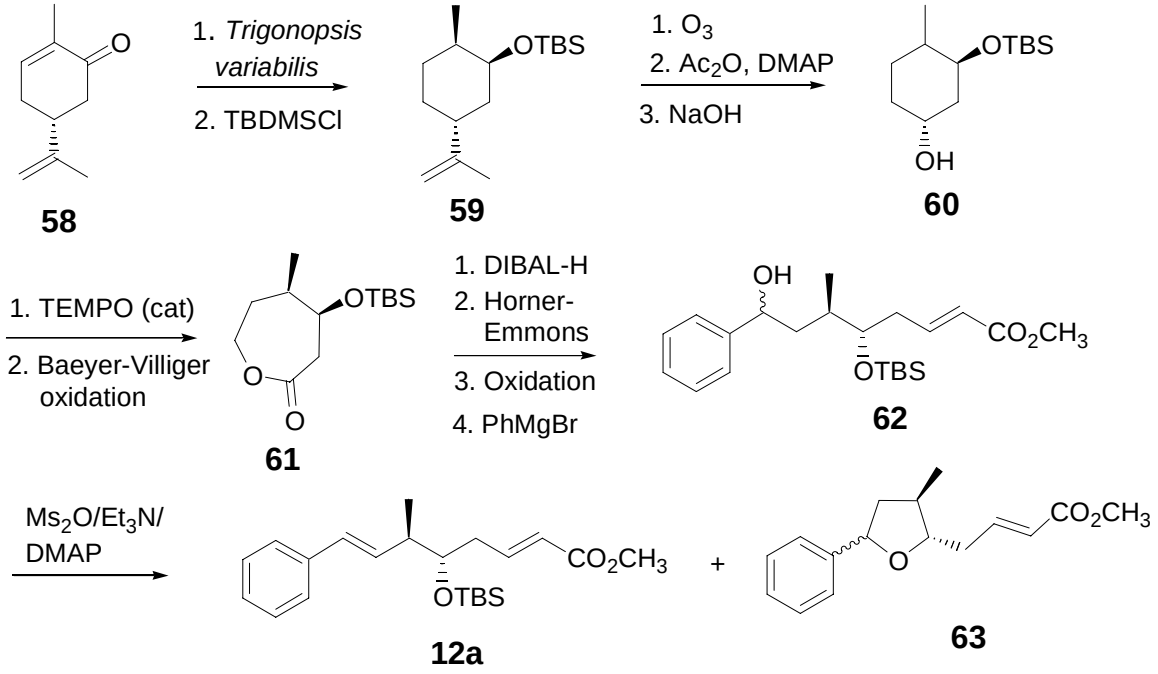
The Sih synthesis of unit A relies upon an enzymatic hydrolysis of a racemic ester (Scheme 11).[54] (*E*)-Methyl styryl acetate **51** was first treated with LDA, followed by dimethyl sulfate to produce a racemic ester which on exposure to *Candida rugosa lipase* in phosphate buffer led to carboxylic acid (*S*)-**52** in 45% and methyl ester (*R*)-**53** in 48% yield. After separation of the methyl ester (*R*)-**53** from the carboxylic acid the ester was reduced with DIBAL-H to aldehyde **54**. The introduction of the remaining carbons of unit A was accomplished by means of a Reformatsky reaction with *tert*-butyl 4-bromocrotonate. In this reaction the undesired *syn*-γ-adduct **55** was isolated in 22%, along with the desired *anti*-γ-adduct **12b** in 18% yield. The undesired major product **55** was converted to desired *anti*-product through a Mitsunobu inversion. Exposure of **55** to 2,4-dinitrobenzoic acid, diethylazodicarboxylate (DEAD) and triphenylphosphine led to nitrobenzoate **57** with correct C5 stereochemistry along with triene **56**. The nitrobenzoate **57** was hydrolysed to **12b** with methanolic potassium carbonate. The overall yield of **55** from **12c** was 13% yield. Although Sih's synthesis of unit A provides an illustration of the synthetic utility of an enzyme-mediated kinetic resolution, the yield is inferior to that of the first Tius synthesis.



Scheme 11: The Sih chemoenzymatic synthesis of unit A

3.4.2 The Lilly chemoenzymatic synthesis

Commercially available (*R*)-carvone **58** was bioreduced with *Trigonopsis variables* (ATCC 10679) using glucose as a carbon source to produce saturated alcohol (de>98%, as judged by GC)[55], which on protection of the secondary hydroxyl group as the TBS derivative led to **59** (Scheme 12). After ozonolysis, Criegee rearrangement and hydrolytic cleavage of intermediate acetate **59** alcohol **60** was produced.[56] Oxidation of **60** took place in a two phase system in presence of catalytic TEMPO, to cyclohexanone which on Baeyer-Villiger oxidation with 30% aqueous hydrogen peroxide and sufficient trifluoroacetic acid led to **61**. The electron-with-drawing effect of the β-silyloxy substituent was expected to favor the rearrangement to lactone **61**.[57] The conversion of **61** to unit A **12a** followed a conventional strategy. Reduction of lactone **61** to lactol with DIBAL-H was succeeded by a Horner-Emmons reaction and then oxidation of the primary hydroxyl group gave the aldehyde. After that addition of phenylmagnesium bromide produced diastereomeric benzylic alcohol **62**.



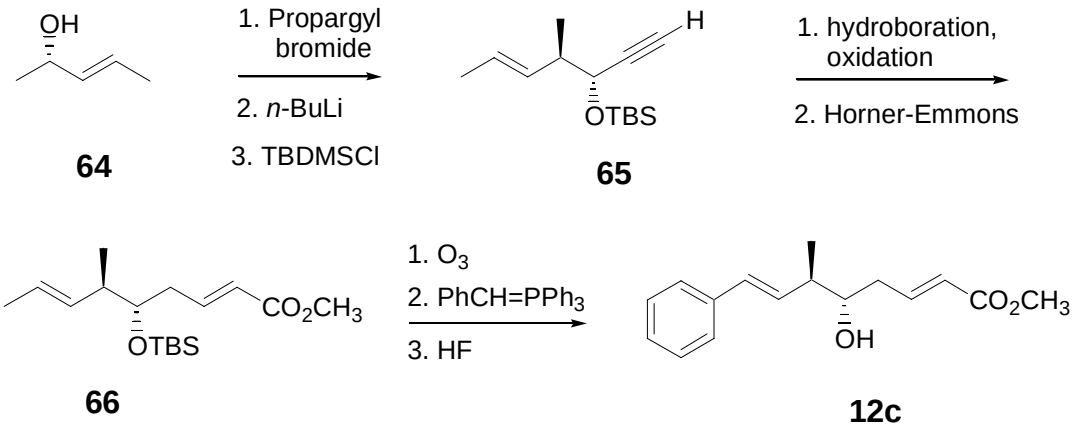
Scheme 12: The Lilly's chemoenzymatic synthesis

The elimination of water from **62** was done with 3 equivalents of methanesulfonic anhydride and 9 equivalents of triethylamine in presence of catalytic DMAP leding to styrene **12a** and tetrahydrofuran **63** in a 4/1 ratio. This chemoenzymatic synthesis of unit A is interesting because it relies upon a non-obvious strategy.

3.4.3 Synthesis via [2,3]-Wittig rearrangement

Nakai and Mikami[58] synthesized unit A via an anionic [2,3]-Wittig rearrangement as a key step. As shown in Scheme 13 they started commercially available (*E*)-3-penten-2-ol (**64**).[59] On reaction with propargyl bromide and strong base under phase transfer condition compound **64** was converted to the corresponding allyl propargyl ether. The subsequent anionic [2,3]-Wittig rearrangement with *n*-butyllithium gave a 9/1 mixture of di-astereomers and the protection of the C5 alcohol of the major product led to **65** as TBS ether. Selective hydroboration of the terminal alkyne in the presence of 1,2-disubstituted alkene with disamylborane, followed by oxidation of the intermediate vinylborane led to aldehyde which on Horner-Emmons homologation gave the α,β-unsaturated ester **66**. On ozonolysis[60] of **66** followed by reductive workup, Wittig reaction with benzylidene

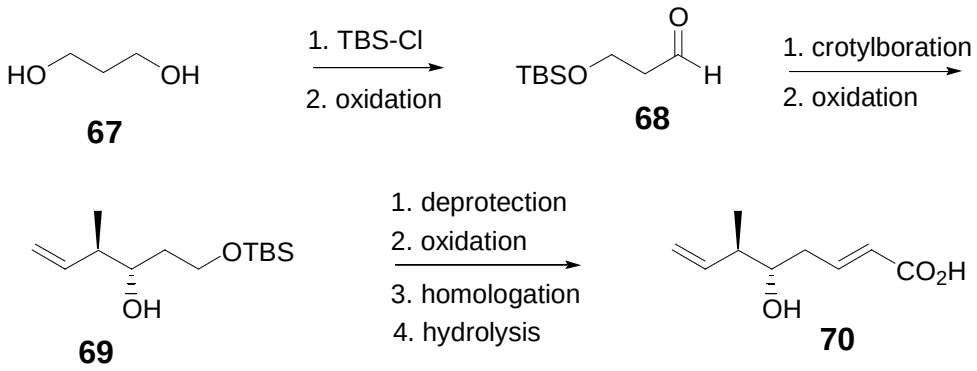
triphenylphosphorane and finally deprotection of the TBS protectinting group with HF gave unit A compound **12c** in an good overall yield.



Scheme 13: Synthesis via a Wittig rearrangement

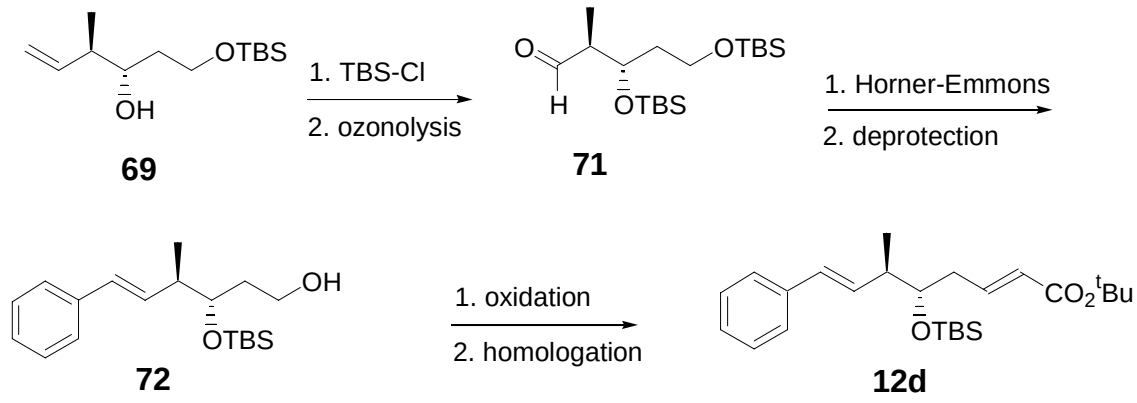
3.4.4 Asymmetric crotylboration

Michael Martinelli and co-workers in Eli Lilly Company have described a very brief synthesis of unit A[61-63] based on an asymmetric crotylboration reaction.[64] After *mono* protection of 1,3-diol **67** (Scheme 14) the free hydroxyl group was oxidized with sodium hypochlorite in presence of TEMPO and led to aldehyde **68**.[65] The enantiometrically pure Brown asymmetric crotylboron reagent[61-63] was added to **68**, followed by oxidation with hydrogen peroxide providing homoallylic alcohol **69** in ≥ 99% ee and de. After quantitative removal of the silyl protecting group with fluoride the compound **69** was again oxidized to an aldehyde. A methyl ester homologue was made by phosphonate, then the ester group hydrolysed with base produced carboxylic acid **70**. The Eli Lilly group introduced the phenyl ring of Cryptophycin by means of a Heck reaction with iodobenzene at the end of the synthetic sequence that led to Cryptophycin-52 (**4**).



Scheme 14: Synthesis by asymmetric crotylboration reaction

White [66,67] and co-workers applied a similar strategy for the synthesis of unit A (Scheme15). After preparing compound **69** they protected the free hydroxyl group with TBS and cleaved the terminal alkene group by ozonolysis, which led to aldehyde **71**. The styryl group was introduced through a Horner-Emmons reaction[68] and showed E isomer as the exclusive reaction product on which the selective removal of the primary siliyl group took place with HF-pyridine complex leading to alcohol **72**. The Dess-Martin periodinane oxidized the alcohol to aldehyde, which was converted to **12d** and protected unit A **12b** by homologation with (*tert*-butoxycarbonylmethylene)triphenylphosphorane[69] in an good overall yield.

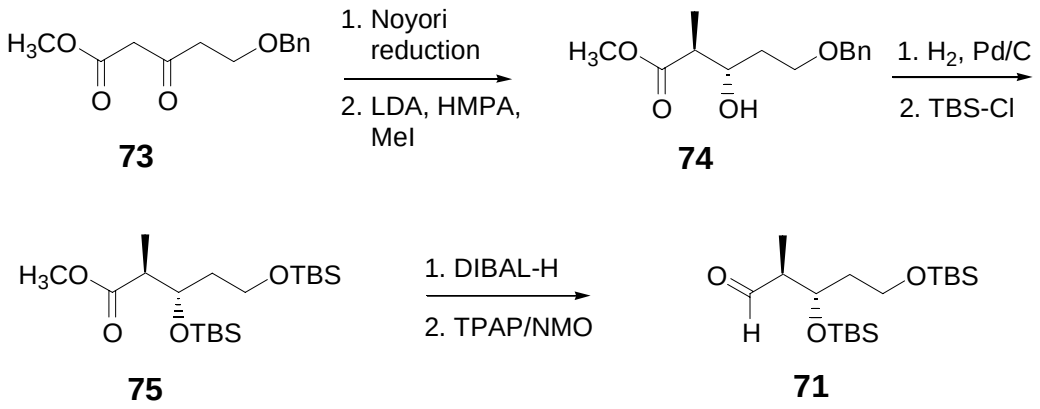


Scheme 15: Synthesis of **12d** from intermediate **69**

On addition to the enantioselective crotylboration approach, White has also disclosed a related strategy based on a diastereoselective allylstannation of a chiral, non-racemic aldehyde.[66] In this synthesis they introduced the styryl part by Takai reaction [70] followed by Stile coupling with phenyltrimethylstannane to produce protected unit A **12d**.[71]

3.4.5 Noyori hydrogenation – Frater alkylation

Georg and co-workers[72] have later made clever use of the Noyori asymmetric hydrogenation to control the absolute stereochemistry at C5 of unit A (Scheme16).[73] Methyl-5-benzyloxy-3-oxopentanone **73** was hydrogenated in presence of (*S*)-BINAP/$RuBr_2$ to the (*R*) alcohol with 97% ee, which on exposure to LDA in THF, followed by a solution of iodomethane in HMPA led to *anti*-product **74**.[74] Hydrogenolytic cleavage of the benzyl ether protecting group, followed by protection of the primary alcohol group as TBS ether derivative gave ester **75**. The reduction of the ester group with DIBAL-H led to the primary alcohol which on oxidation with TPAP/NMO[75] produced aldehyde **71**, the same intermediate that White used, and from this point on, Georg's and White's synthesis were parallel.

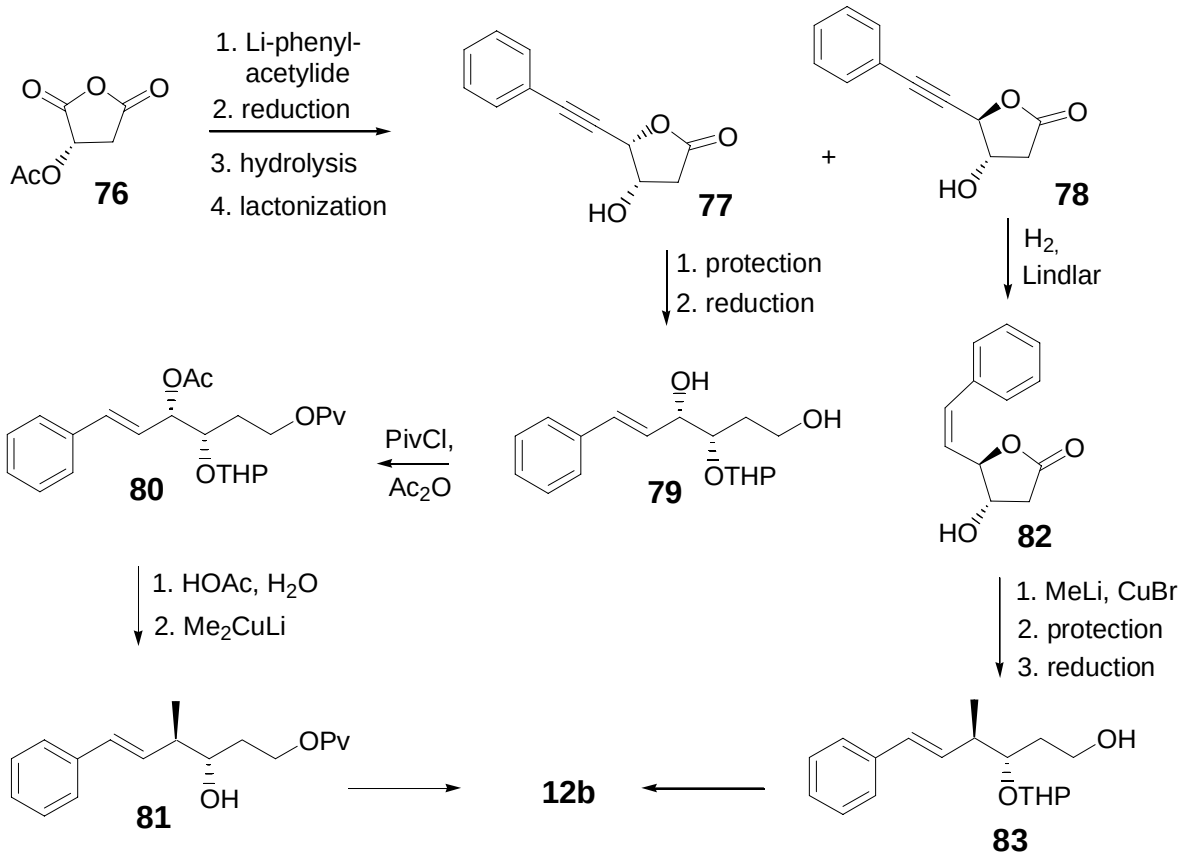


Scheme 16: Synthetic pathway by Noyori hydrogenation **–** Frater alkylation

3.4.6 (*S*)-(−)-2-Acetoxysuccinic anhydride as starting material

This is a very unconventional synthetic pathway for unit A, described by Lavalee and co-workers (Scheme 17).[76] Regioselective ring opening at C1 of (*S*)-2-acetoxysuccinic anhydride **76** with lithium phenylacetylide was followed by non-stereoselective reduction by sodium borohydride which on base-mediated hydrolysis led to 1/1 mixture diastereomeric diols. Exposure of this mixture to tosic acid led to diastereomeric lactones **77** and **78**. In the compound **77** the hydroxyl group was protected as ether was followed by quantative reduction with $LiAlH_4$ to produce diol **79**. Sequencial pivaloylation of the primary hydroxyl group, followed by acetylation of secondary hydroxyl group in situ, led

to fully protected triol **80**. After selective hydrolysis of the THP protecting group with aqueous acetic acid, this allylic acetate was exposed to lithium dimethyl cuprate and gave **81**. The conversion of **81** to unit A compound **12c** took place in five steps. Protection of C5 alcohol by THP, the cleavage of pivaloate by $LiAlH_4$, Swern oxidation of the primary alcohol, exposure to (*tert*-butoxycarbonylmethylene)triphenyl phosphorane and finally hydrolytic cleavage of THP group led to **12b** in a good yield.

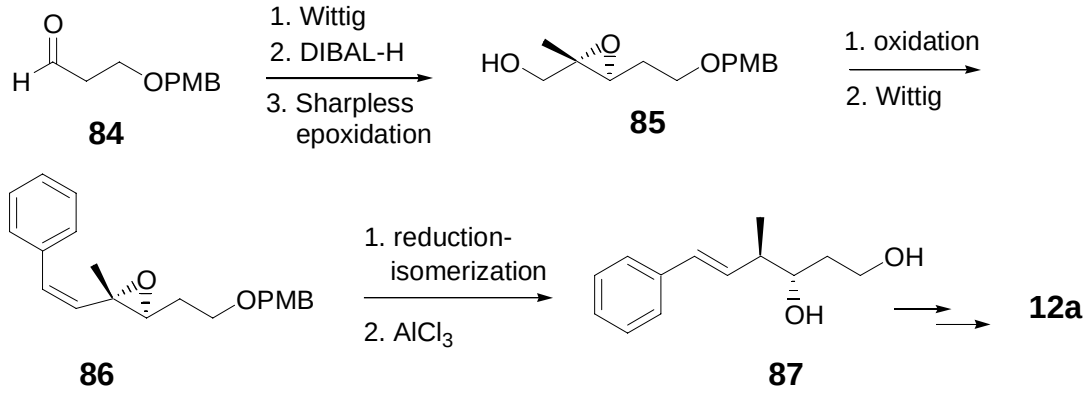


Scheme17: Synthetic pathway of unit A from acetoxysuccinic anhydride

The compound **78** was semihydrogenated over Lindar's catalyst and led to alkene **82**. On methyl cuprate addition to **82**, protection of the C5 hydroxyl group as THP ether and reduction of the carboxylic acid to primary alcohol with $LiAlH_4$ led to **83**. Compound **83** was converted to **12b** through the same last three steps (Swern oxidation; Wittig homologation; hydrolysis) as has been described for the conversion of **12b** from **81**.

3.4.7 Vinyl epoxide reduction

Like the original Tius publication, Furuyama and Shimizu made use of the Sharpless asymmetric epoxidation for control of the absolute stereochemistry of unit A.[77] The *para*-methoxybenzyl derivative **84** of 3-hydroxypropanal (Scheme 18) was treated with [1-(ethoxycarbonyl)ethyl]triphenylphosphorans, followed by reduction of the ester function with DIBAL-H to the primary alcohol which was converted to the non-racemic epoxide **85** under Sharpless condition, using (+)-diethyl tartrate the chiral inducer. After Swern oxidation, the epoxyaldehyde was exposed to benzylidenetriphenylphosphorane and warming to room temperature led to *Z*-styryl compound **86**. After that the key step in the sequence was the palladium mediated reduction-isomerisation which set the relative and absolute stereochemistry at C5 and C6 (unit A). The isomerization process was initiated by the epoxide ring opening with formation of a π-allyl palladium species (Figure 7). The conversion took place by reaction with catalytic Pd(0), tri-*n*-butyl-phosphine and triethyl ammonium formate.



Scheme 18: Synthetic pathway by vinyl epoxide reduction

Interconversion between this π-allyl intermediate and a σ-boned palladium species was reversible. Relief of non-bonding interactions between phenyl and methyl groups favored one of the two rotamers of the σ- bound intermediate. Conversion to another π-allyl species, followed by reductive elimination of palladium, completed the catalytic cycle which after deprotection of PMB group led to **87**. Protection of both hydroxyl groups as TBS ether and selective hydrolysis of primary TBS with acetic acid gave **72** (Scheme 15) which on Swern oxidation and Horner-Emmons homologation leading to **12a**, the unit A compound of the first Tius synthesis.

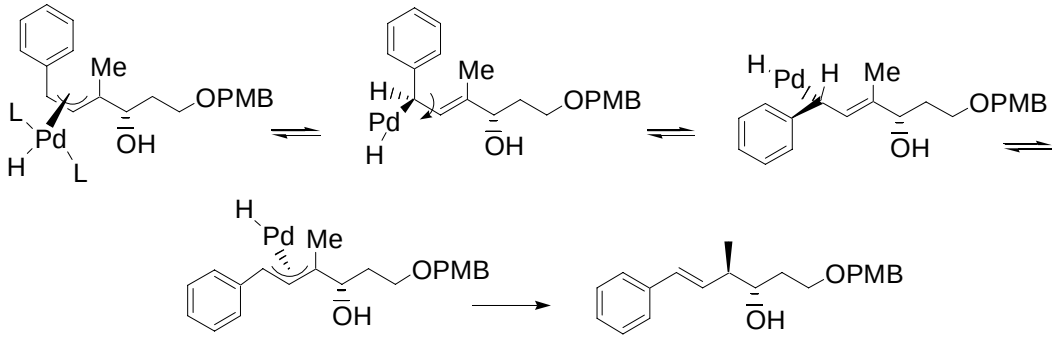
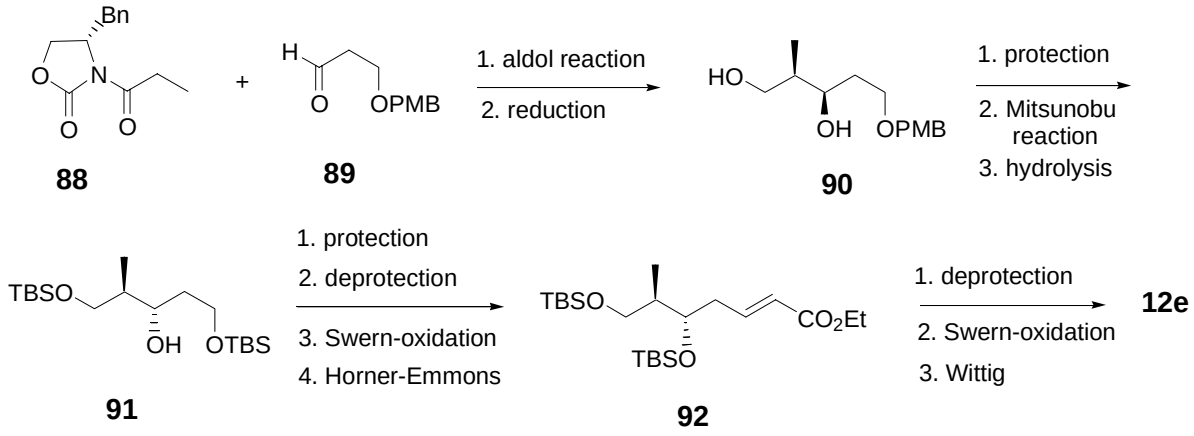


Figure 7: The proposed mechanism for the reduction-isomerisation step

3.4.8 Synthesis via Mitsunobu reaction.

In this synthesis, the unit A was prepared by *syn*-aldol methodology followed by Mitsunobu reaction (Scheme 19).[78] The Evans aldol reaction was done between the auxiliary **88** and the protected aldehyde **89**. After reductive removal of the chiral auxiliary the primary hydroxyl group of **90** was protected by silylation to allow the inversion of the secondary hydroxyl group by Mitsunobu reaction.[79] Basic ester hydrolysis delivered an alcohol, which was protected giving **91**. Extension of the carbon chain on the other terminus was done by oxidative removal of the *p*-methoxy benzyl group, followed by Swern oxidation and Horner-Wadsworth-Emmons reaction to give compound **92**. Construction of the styrene part was done by selective removal of the primary silyl protecting group, Swern oxidation and Wittig reaction between the obtained aldehyde and benzyl phosphonate providing the target compound **12d.**

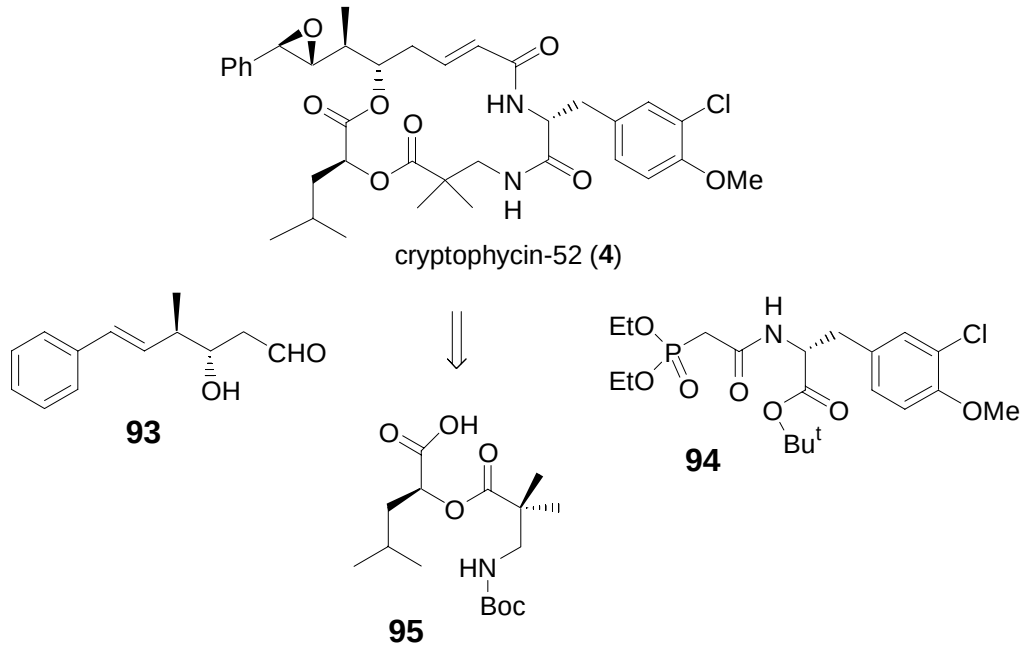


Scheme 19: Synthesis by Mitsunobu reaction

3.5. Syntheses of some important Cryptophycin analogues

3.5.1. Synthesis of stable Cryptophycin-52 (**4**)

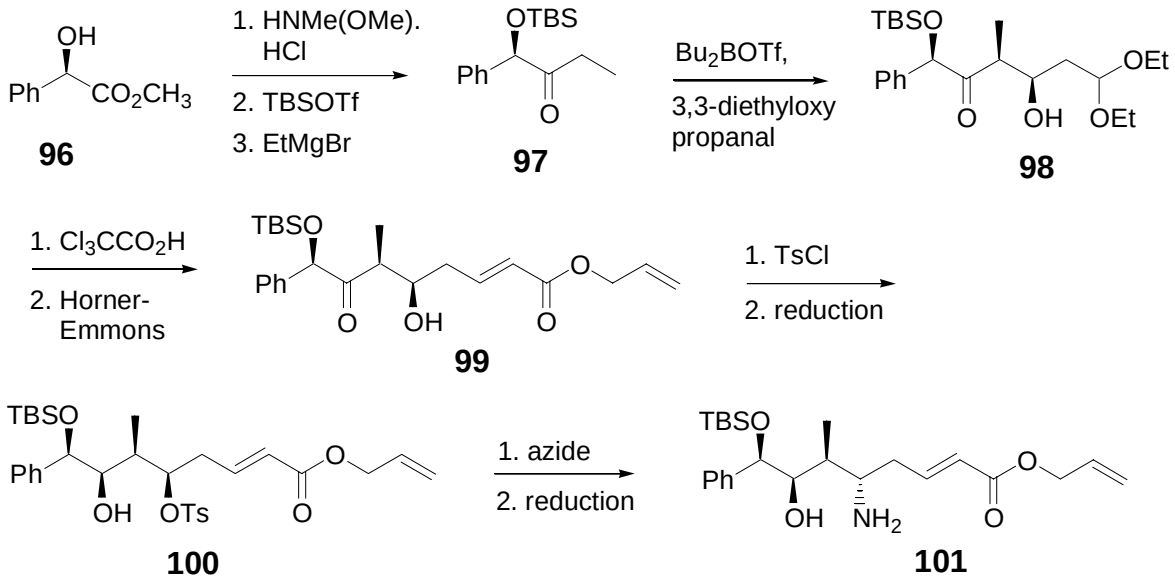
In comparison with Cryptophycin-1 (**1**), Cryptophycin-52 (**4**) contains an additional methyl group in unit C, which increases the steric hindrance and diminishes the rate of hydrolytic cleavage of the unit C-unit D ester linkage. Synthesis of Cryptophycin-52 was done by several groups and one of them was the group of A. K. Ghosh.[80] They synthesised by assembly of three fragments (Scheme 20), phenyl hexenal **93**, D-tyrosine phosphonate **94**, and protected β-amino acid derivative of **95**. In the fragment **93** both of the stereogenic centres at C3 and C4 were derived from optically active 4-phenylbutyrolactone, synthesised enantioselectively by Corey-Bakshi-Shibata reduction.[81] A Horner-Emmons olefination between aldehyde **93**, and phosphonate **94** generated the key tyrosine octadienamide subunit, which on esterification with acid of **95** offered the corresponding protected acyclic precursor for Cryptophycin-52. Removal of the appropriate protecting groups followed by cycloamidation of the resulting amino acid constructed the 16-membered macrocyclic ring. They generated the sensitive epoxide at the final step of the synthesis with *m*-CPBA as a 2/1 mixture of two diastereomers.



Scheme 20: Retrosynthetic analysis of Cryptophycin-52

3.5.2 1-Aza-Cryptophycin-1, an unstable Cryptophycin

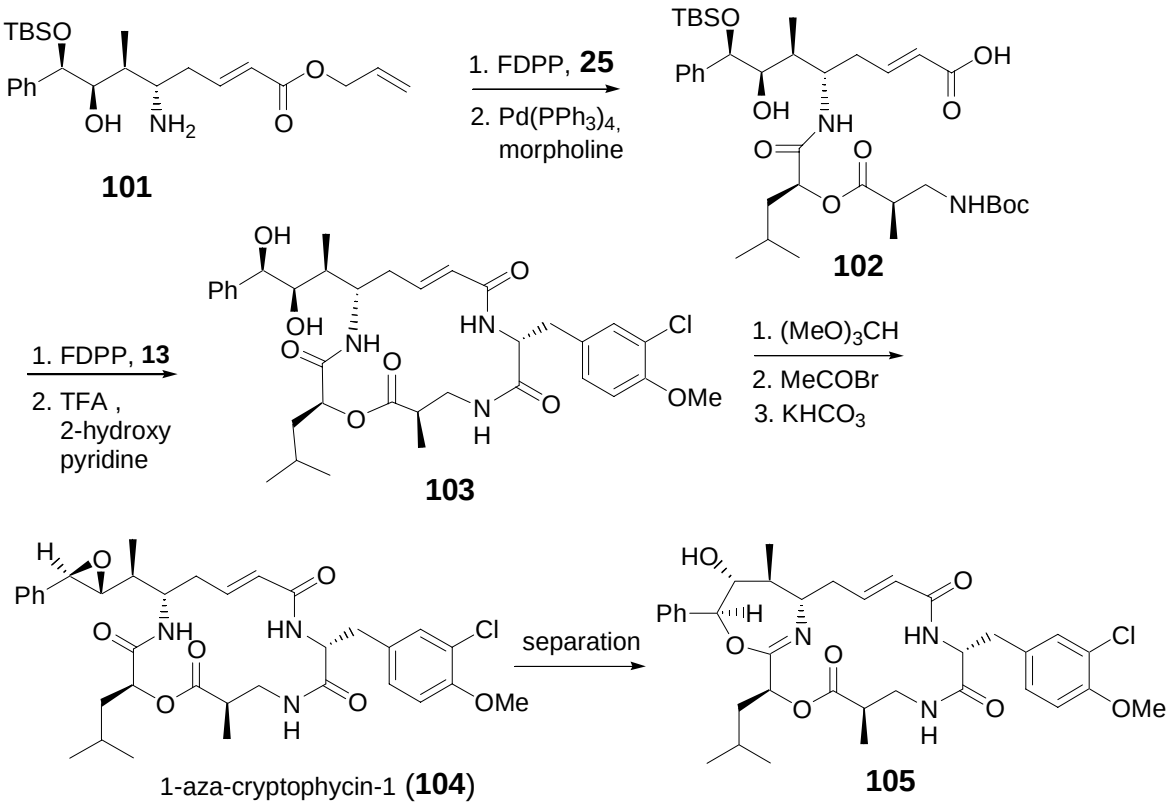
An alternative strategy for stabilizing the Cryptophycins against hydrolytic cleavage is by replacement of labile ester links for amide links between unit A and unit D. In this context the Tius group discussed the synthesis and aspects of the reactivity of 1-*aza*-Cryptophycin-1.[82] The synthesis of *aza* analogue of unit A (Scheme 21) controls the epoxide stereochemistry by using (*R*)-methyl mandelate **96** as a source of asymmetry. Conversion of methyl ester **96** to Weinreb amide was done in presence of trimethylaluminium, and the hydroxyl group was protected as TBS ether, followed by the reaction with ethylmagnesium bromide which led to ethyl ketone **97**. Condensation of di-*n*-butylboryl enolate[83,84,] derived from **97** with 3,3-dihydroxypropanol, [85,86] followed by oxidative work up, led exclusively to *syn* diol **98**. Acid hydrolysis of the acetal function to the aldehyde and Horner-Emmons reaction under Masamune-Roush conditions gave allyl ester **99**. Conversion of alcohol to tosylate was followed by Luche reduction of the keto group and led to alcohol **100** with 20/1 stereoselectivity.[87] Exposure of **100** to tetramethylguanidinium azide and reduction of azide with triphenylphosphine led to primary amine **101**, an analogue of unit A.



Scheme 21: Synthesis of analogue of unit A of 1-Aza Cryptophycin-1

The condensation of **101** with fragment **25** (unit C + D) was mediated by FDPP, which on palladium catalyst cleavage of the allyl ester produced carboxylic acid **102** (Scheme 22). The carboxylate group of **102** was condensed by the ame conditions with protected unit B

13a to give the seco compound. Exposure of this compound to TFA, followed by evaporation to dryness and reaction with 2-hydroxypyridine led to cyclic syn-diol compound **103**. The conversion of *syn*-diol in **103** to epoxide was accomplished through orthoformate, which was treated with acetyl bromide and then with potassium bicarbonate. leading to an approximately equimolar mixture of desired epoxide **104** and **105**.



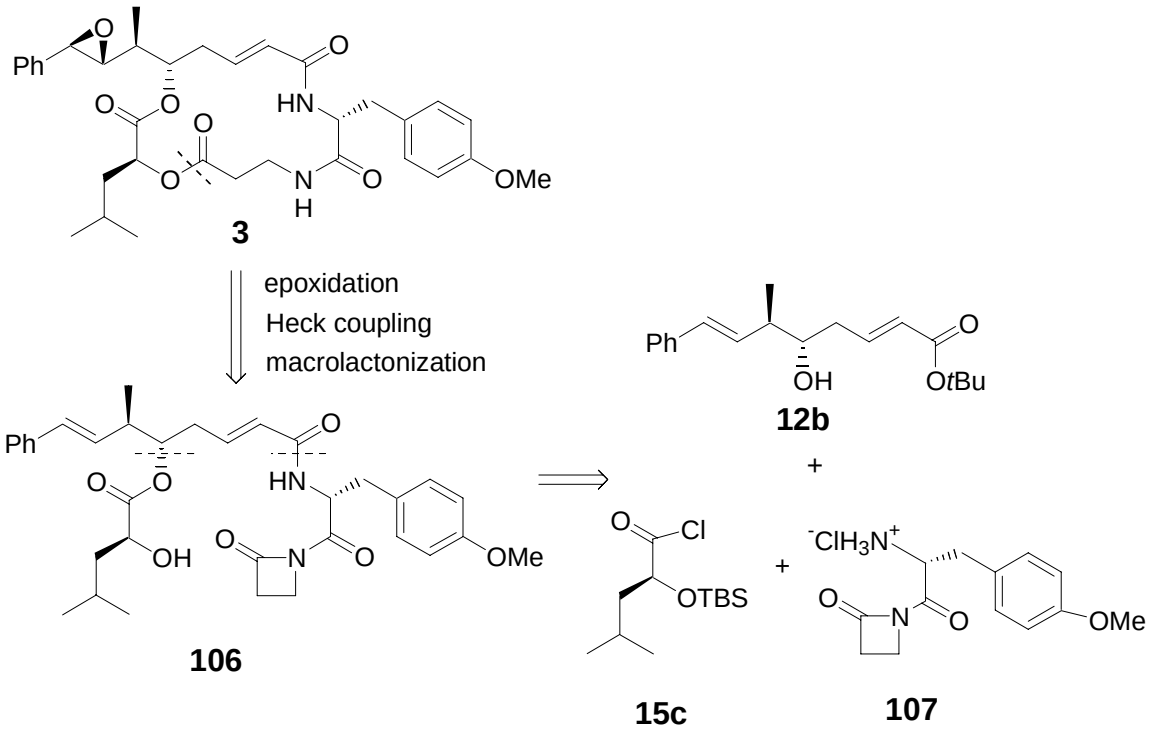
Scheme 22: Synthesis of 1-aza-Cryptophycin-1

3.5.3. Synthesis of Cryptophycin-24 (**3**)

3.5.3.1 Synthesis via N-Acyl-β-lactam macrolactonization

The simplest of the macrolides, Cryptophycin-24 (**3**) which lacks a chiral centre at C6 and a chlorine substituent in the tyrosine moiety, was synthesized by George and co-workers.[88,89] In this synthesis a efficient and concise approach, involving a cyanide-initiated acyl-β-lactam ring opening followed by cyclization was applied successfully. The retrosynthetic analysis for Cryptophycin-24 reveals that the molecule **106** be assembled from three basic building blocks, octadienoate esters **12b**, L-leucic derivative **15c** and N-acylazetidinones **107** (Scheme 23). The introduction of the 3′-phenyl group of Cryptophy-

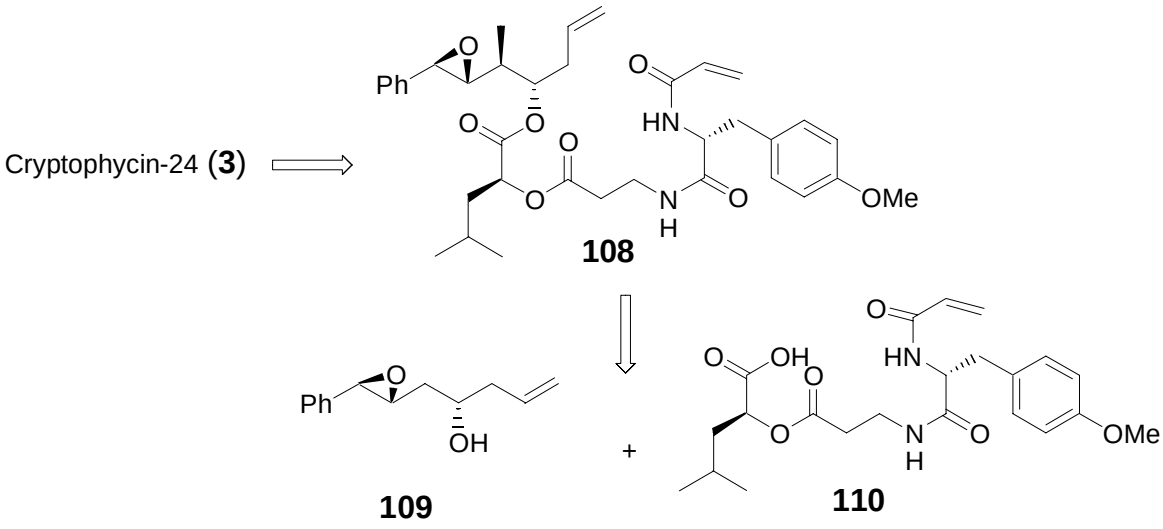
cin-24 could be accomplished by a Heck reaction, at the early stage in the synthesis, or later in the synthetic scheme. This approach made the synthesis flexible for incorporation of various aryl substituents at the C3′ position.



Scheme 23: Retrosynthetic pathway to Cryptophycin-24 (**3**)

3.5.3.2 Synthesis via RCM approach

Georg and co-workers later also synthesized Cryptophycin-24 (**3**) from diene **108** using RCM conditions.[90] The intermediate diene **108** was prepared by assembly of epoxy alcohol **109**[15,16] and acid **110** (Scheme 24).



Scheme 24: RCM approach for synthesis of Cryptophycin-24 (**3**)

In the final step the macrocyclization was done by metathesis reaction of diene **108** with the Grubbs 1 catalyst[91] with exclusive formation of the *E*-isomer to provide Cryptophycin-24 in good yield and the synthesis has been achieved in the presence of the chemically reactive styrene epoxide moiety.

There are some other reports of total and formal synthesis[92-94] but they are not discussed here because of their low selectivity and moreover they are more or less similar to other pathways of synthesis.

4. Key reactions and mechanisms

4.1. Aldol reaction

The aldol reaction is one of the most important methods of forming carbon-carbon bonds. The addition of an enolate to an aldehyde leads to the formation of at least one chiral center. In the case of α-substituted enolates it has been possible to control the product stereochemistry to a large extent, while the aldol reaction of α-unsubstituted chiral enolates was for many years a "problem child" for synthetic chemists because of its insufficient stereoselectivity. Progress in this area has only been made in the last few years using either new chiral auxiliaries or alternatives to the aldol reaction.[95]

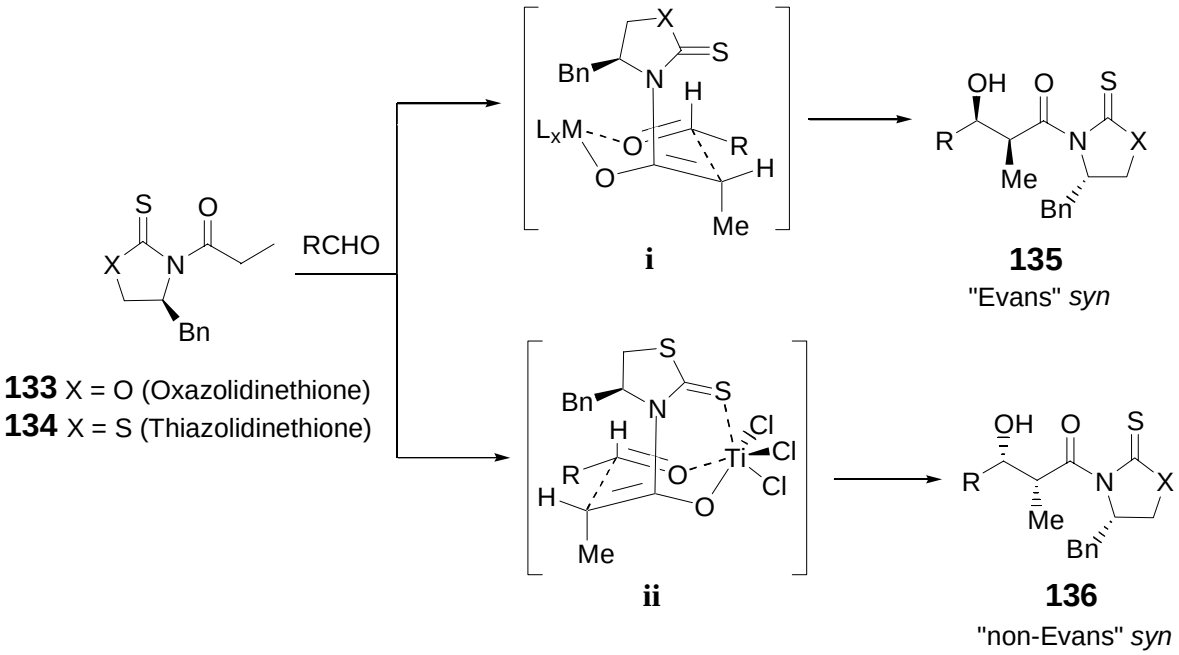
Aldol reaction, a classical method for the construction of carbon chains with oxygen functionality in 1,3-positions,[96] has undergone remarkable changes in the last twenty years. The impulse for this development was given by the increasingly ambitious synthetic goals, which were provided in particular by the macrolide and polyether antibiotics with their many functional groups. New and particularly stereoselective variants of the aldol reaction have proved to provide the key to success.

4.1.1 *Syn* selective Crimmins aldol

An important application of chiral oxazolidinethiones and thiazolidinethiones is the asymmetric aldol condensation of their corresponding *N*-acyl derivatives with suitable aldehyde partners. The use of *N*-acyloxazolidinethiones and thiazolidinethiones in asymmetric aldol additions complements very well other existing methodologies such as the dialkylboron enolates of *N*-acyloxazolidinones pioneered by Evans,[97] and the tin(II) enolates of *N*-acyl sultams developed by Oppolzer.[98]

Since the first report of the use of chiral *N*-acyloxazolidinethiones in asymmetric aldol-type reactions by Nagao in 1985,[99] the number of examples of their application in the total synthesis of natural products has grown exponentially. More recently, Crimmins and coworkers published a detailed account of their work in asymmetric aldol additions employing titanium(IV) enolates of *N*-acyloxazolidinethiones and *N*-acylthiazolidinethiones.[100] Crimmins addressed the importance of the dialkylboron enolates of *N*-acyloxazolidinones as the most commonly used enolates for the preparation of the Evans *syn* products.

The reaction with boron enolates proceeds via the non-chelated transition state **i** to deliver the well-known Evans *syn* products **113** (Scheme 25). However, the use of titanium(IV) enolates of *N*-acyloxazolidinethiones and *N*-acylthiazolidinethiones allows the reaction to proceed via the chelated transition state **ii** to deliver the non-Evans *syn* products **114**. Furthermore, Crimmins reported the potential of titanium(IV) enolates of both *N*-acyloxazoli-dinethiones **111** and *N*-acylthiazolidinethiones **112** for the preparation of both Evans and non-Evans *syn* aldol products by variation of the reaction conditions.



Scheme 25: Non-chelated (**i)** and chelated **(ii)** transition states in the asymmetric aldol addition of titanium(IV) enolates of *N*-acyloxazolidinethiones and thiazolidinethiones.

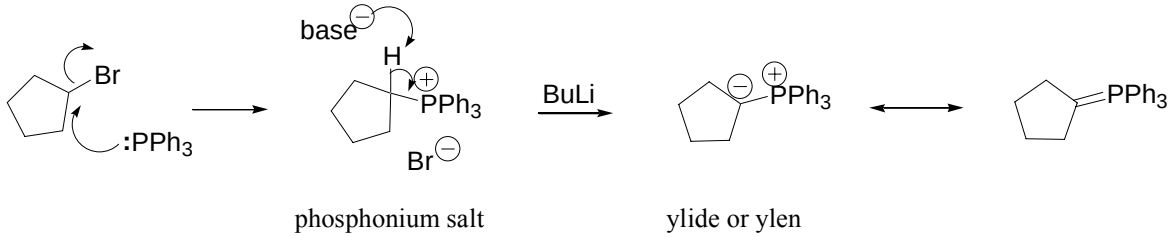
Crimmins found that the diastereoselectivity of the titanium(IV) enolates of *N*-acyloxazolidinethiones and *N*-acylthiazolidinethiones to deliver the Evans **113** or non-Evans *syn* products **114** is dependent on the nature and amount of the base used to generate the enolates.[100] The Evans *syn* products were obtained, via the non-chelated transition state **i**, when the titanium enolates were formed in the presence of two equivalents of a base such as (–)-sparteine. It was suggested that the second equivalent of amine coordinates to the metal center preventing further coordination of the imide or thioimide carbonyl to the metal center. Non-Evans *syn* products were obtained when only one equivalent of amine was used to generate the enolates. In this case the imide carbonyl or the thiocarbonyl coordinated to the metal center to produce the highly ordered chelated transition state **ii**. Coordination of the imide carbonyl or thiocarbonyl to the metal center led to reversal of the π-facial orientation of the enolate in the transition state.[100]

4.2 Wittig reaction

It is a reaction between a carbonyl compound (aldehyde or ketone only) and a species known as phosphonium ylide to form an alkene. Unlike with other elimination reactions, the Wittig elimination step occurs only from an intermediate not from isolated starting ma-

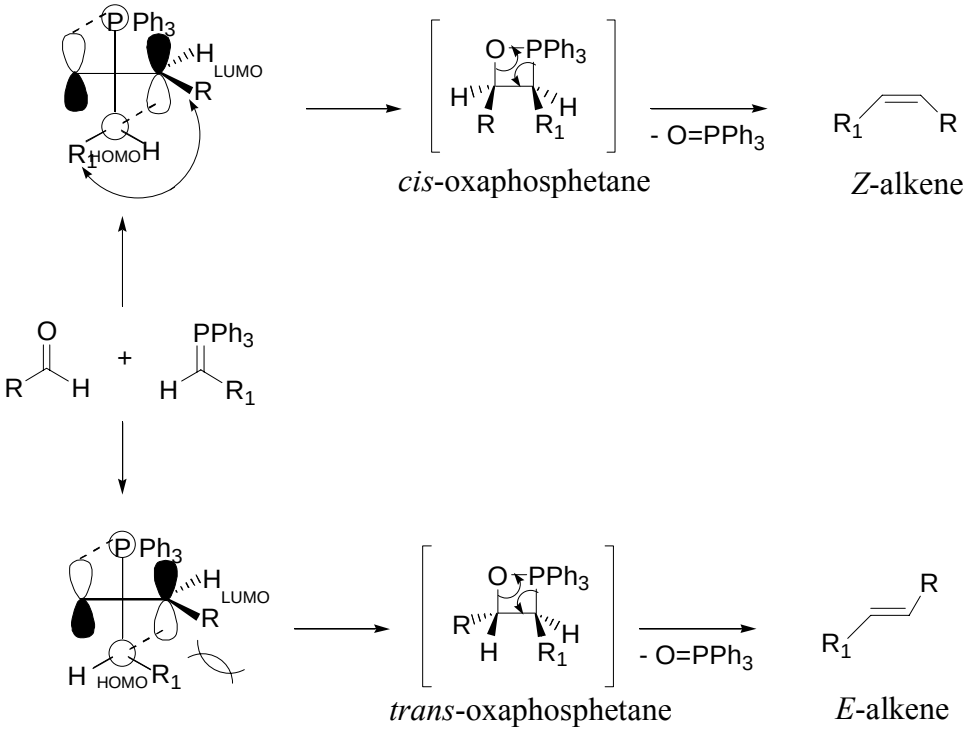
terials. This intermediate is made in situ in the reaction and decomposes spontaneously. The Wittig reaction is therefore a connective alkene-forming reaction. It is much more widely used than the Julia or Peterson reaction, because the Wittig reaction only needs one step.

An ylide is a species with positive and negative charges on adjacent atoms, and a phosphonium ylide carries its positive charge on phosphorous. They are made from phosphonium salts by deprotonation them with a strong base (Scheme 26).



Scheme 26: Synthesis of ylide

The stereochemistry of the Wittig reaction can be affected by solvent, cation, temperature, and type of aldehyde. *Z*-stereoselectivity is maximized by polar aprotic solvent, exclusion of lithium salts, and low reaction temperature.[101] It is assumed that, the reaction occurs through a [2+2] cycloaddition of the ylide with the aldehyde, which can result in a *cis*- or *trans*-configuration of the oxaphosphetane (Scheme 27). Because of the four participating electrons, the thermal cycloaddition has to occur via a MÖBIUS transition state or a [π^2 s + π^2a] approach, respectively.[102] The two double bonds approach each other in a perpendicular fashion with the R groups as far as possible apart from each other.



Scheme 27: Mechanism of Wittig reaction

Therefore, the transition state leading to the *cis*-oxaphosphetane is favoured. The collapse of the four membered oxaphosphetane is stereoselective and the *cis*-oxaphosphetane exclusively forms a *cis*-olefin and *trans*-oxaphosphetane only *trans*-olefin. The formation of the 1,2-substituted oxaphosphetane is kinetically controlled, but when stabilized ylide are used the thermodynamic stability of the oxaphosphetane is increased and therefore, the selectivity of the *cis*-oxaphosphetane is decreased. Consequently in this case the *E*-alkene can be favored.

4.3 Coupling reactions

A key step in the peptide production process is the fomation of the peptide bond. This requires the activation of a carboxylic acid which is generally done by a so-called coupling reagent (Figure 8). The peptide coupling reagent field has clearly evolved in the last decades from carbodiimides to onium (phosphonium and uronium) salts.

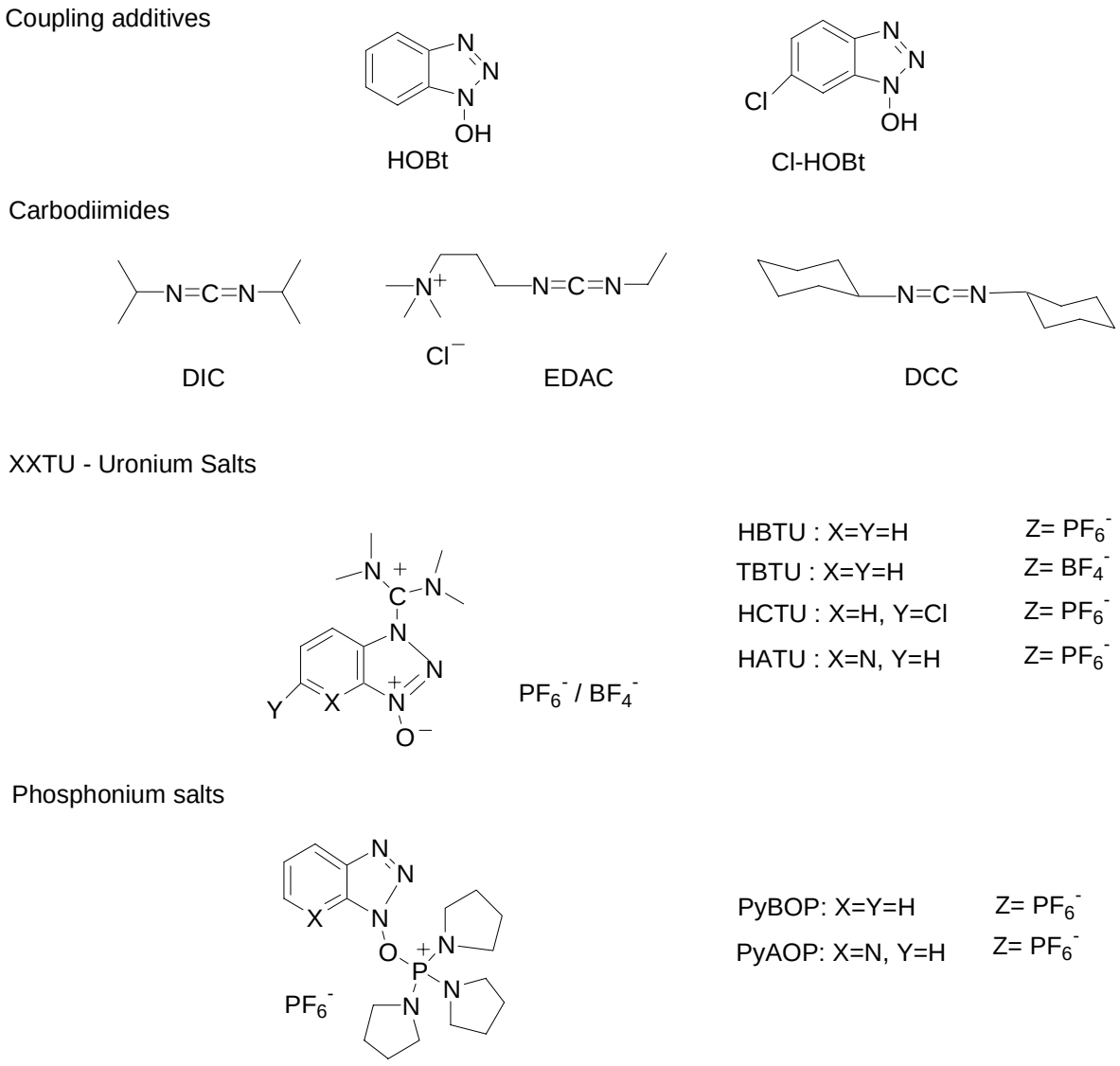
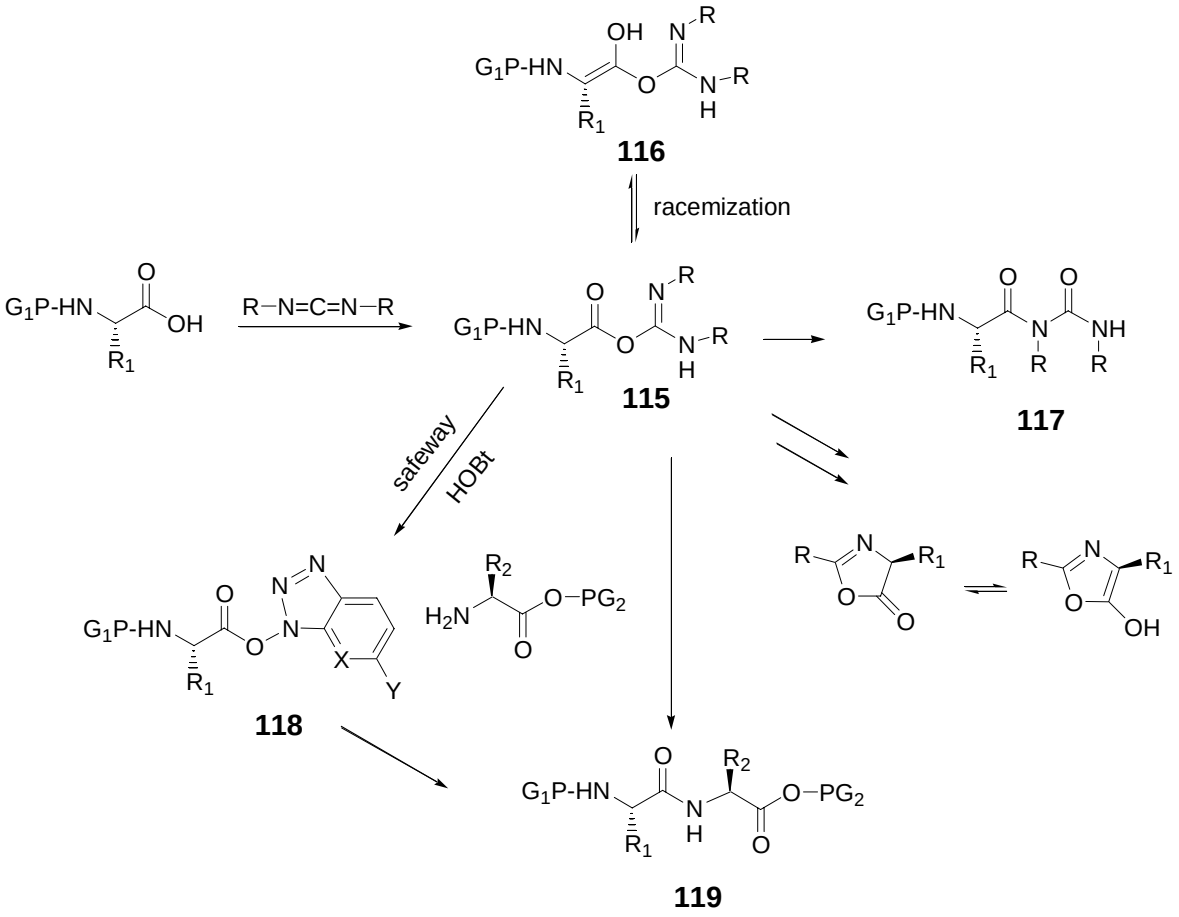


Figure 8: Coupling additives and coupling reagents

4.3.1. Coupling by carbodiimides

The area of industrial coupling reagents began in 1955 with the introduction of dicyclohexylcarbodiimide (DCC), which at that time was already known and well studied as a reagent for formation of amide bonds. The mechanism of the carbodiimide activation is complex and depending on the solvent, starts by a proton transfer, followed by addition of carboxylate to form the *O*-acylisourea **115** (Scheme 28). This is the most reactive species which can attack the amino component to give the corresponding amide. However, the *O*-acylisourea **115** can undergo a rearrangement to give the *N*-acyl urea **117**, which is not reactive or sustains an intramolecular cyclization to give a 5(4*H*)-oxazolone. This is less

reactive than the*O*-acylisourea and can tautomerize with the corresponding loss of chirality.[103]

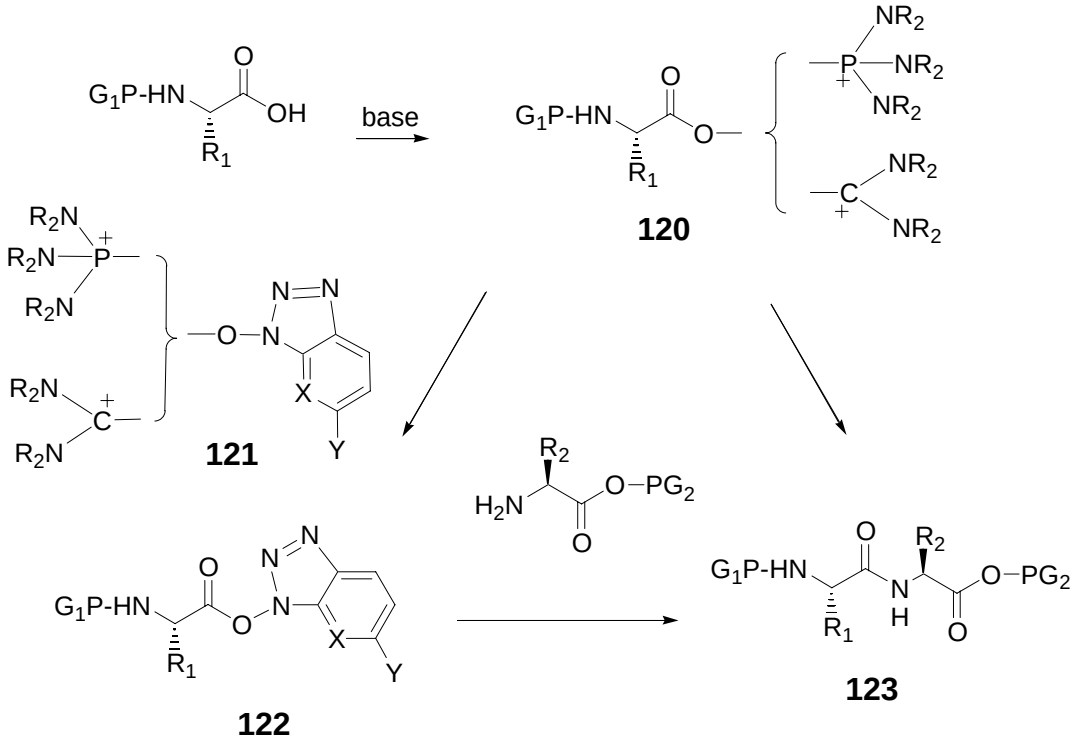


Scheme 28: Mechanism of peptide bond formation by carbodiimide activation

Another problem is racemization of the activated ester **115** to **116** before attacking **115** directly with the amino group of the second amino acid. Therefore it is better to have plenty of 1-Hydroxybenzotriazole (HOBt) which intercepts the activated ester **115** first so the new intermediate **118** does not racemize, mostly because the reaction is highly accelerated by the addition of HOBt. The second amino acid, protected on the carboxyl group, then attacks the HOBt **118** ester and gives the dipeptide **119** in a very fast reaction without racemization. All these factors make the addition of a benzotriazole derivative almost mandatory to maintain high yields and chiral configuration during the peptide bond formation by carbodiimide activation.

4.3.2 Coupling by uronium and phosphonium salts of benzotriazoles

Other widely used coupling reagents include uronium and phosphonium salts such as HATU, HBTU,TBTU and PyBOP, PyAOP respectively. Amino acids react with onium salt in the presence of bases (Scheme 29) and form the hypothetic intermediate species of acyloxy-onium salts **120.**



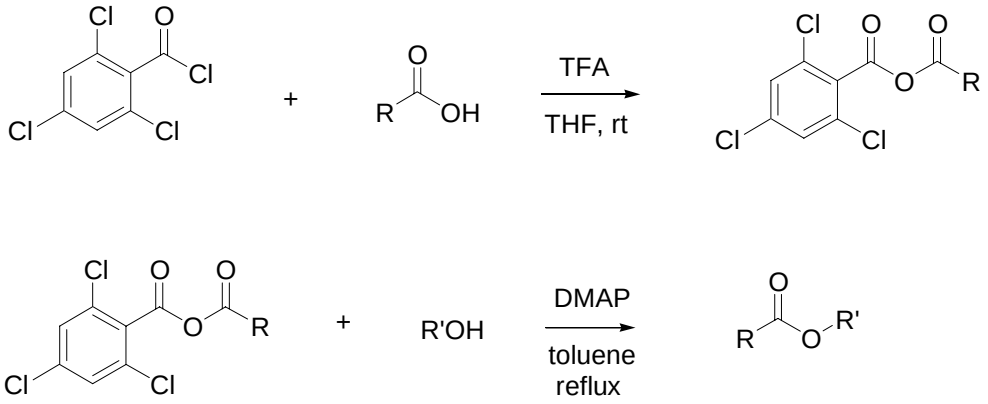
Scheme 29: Mechanism of peptide bond formation through onium salts activation

Until now these intermediate species have not been detected or isolated, as they react immediately with the benzotriazole derivative **121** to give the hydroxybenzotriazole ester **122**. To accelerate this an extra equivalent **121** is added in some synthetic protocols. Ester **122** reacts with the the amino component to lead to the corresponding amide **123**.[103]
Uronium salts having the counterion hexafluorophosphate are more soluble than tetrafluoroborates salts, which allow the preparation of more concentrated solutions.

4.4 Yamaguchi esterification

The coupling of carboxylic acids with alcohols to produce esters is a fundamental synthetic process. A variety of conditions have been developed to accommodate different synthetic scenarios.[104-106] In these procedures equimolar amounts of carboxylic acid and alcohol,

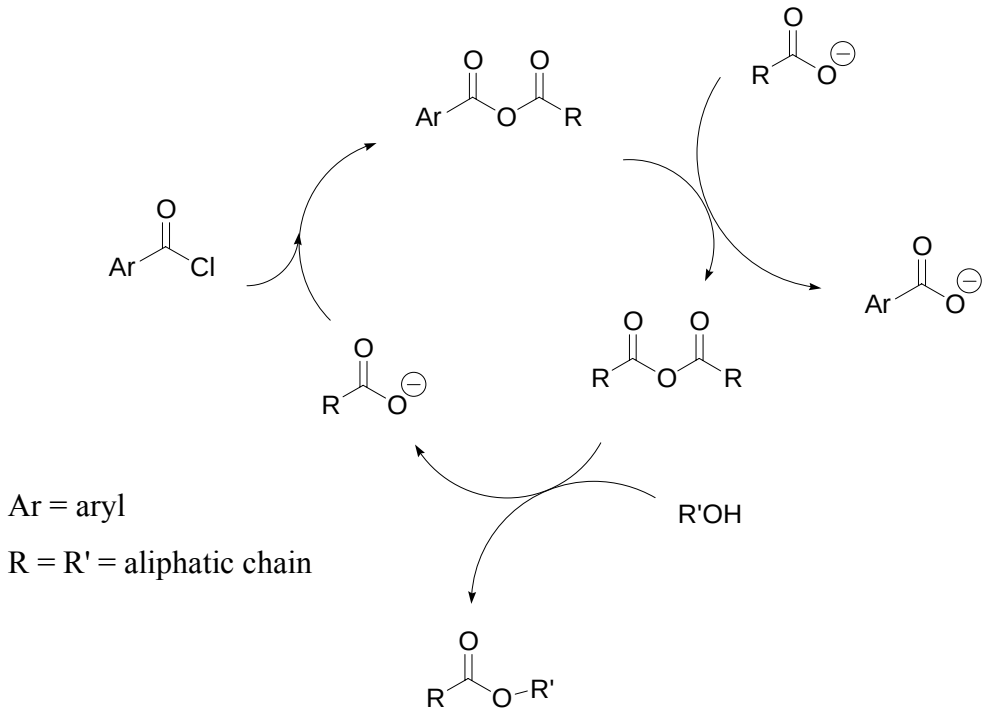
mild conditions, and the elimination of toxic byproducts were established. Of these, the Yamaguchi esterification is very effective and commonly used in the synthesis of macrolactones. It has found great use in regioselective synthesis of highly functionalized esters.[107] The Yamaguchi esterification involves the reaction of an acid with 2,4,6-trichlorobenzoyl chloride to form the mixed 2,4,6-trichlorobenzoyl anhydride.[49] This isolated mixed anhydride, upon reaction with an alcohol, in the presence of DMAP, produces the ester regioselectively (Scheme 30).



Scheme 30: Yamaguchi esterification

In this mechanism all the reactions involved are reversible, except for the last ester formation step. The byproduct of this step is the aliphatic carboxylate, which reenters the cycle. Thus, until the regioselective completion of the reaction, there is always aliphatic carboxylate remaining, competing with the aromatic carboxylate and the alcohol. The mechanism is based on the assumption that the aliphatic carboxylates are better nucleophiles than the aromatic carboxylates and the alcohols.

This proposed mechanism suggests that any aromatic acid chloride capable of producing preferentially and in situ the symmetric aliphatic anhydrides could be used in the regioselective synthesis of aliphatic esters. It is more important to consider the relationship between steric effects, electronic effects, and reactivity. The aliphatic anhydride produced in situ must be more electrophilic towards the alcohol than the aromatic carbonyl of the mixed aliphatic-aromatic anhydride for this procedure to succeed.



Scheme 31: Postulated mechanism of the Yamaguchi esterification

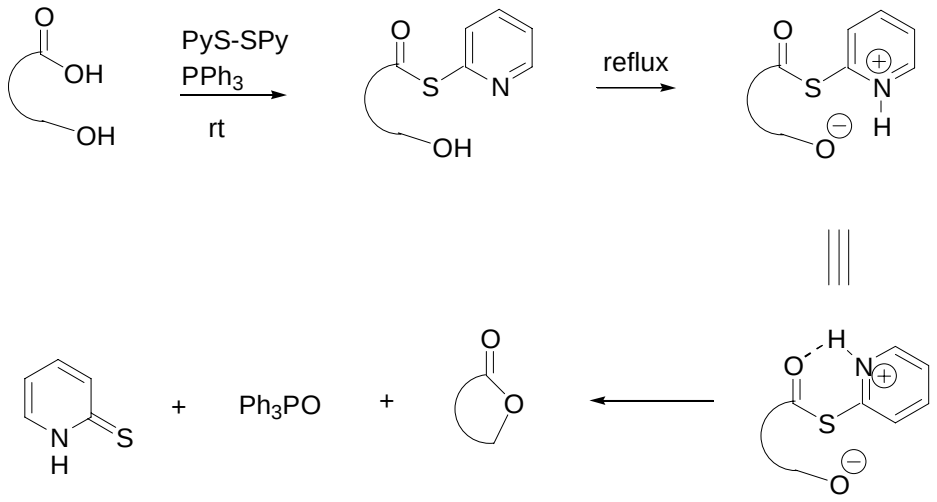
4.5 Macrolactonization

Macrolactones are quite common in nature. The lactone ring imposes conformational constraint and its general improves binding to a receptor. Due to the ring size, the synthesis of macrolactones requires special activation reagents combined with high dilution conditions.

4.5.1 Corey and Nicolaou reactions

The macrolactonization of thioesters corresponds to the biosynthetic pathway for the formation of macrolides.[108-112] It is therefore not surprising that this strategy has also been one of the most popular chemical ways to obtain such macrolactones. The most famous reaction involving a thioester is the "double activation" method described in 1974 by Corey and Nicolaou.[113] This mechanism involves the initial formation of a 2-pyridine thioester of the ω-hydroxy acid via a Mukaiyama oxidation-reduction condensation with PySSPy and triphenylphosphine.[114] Internal proton transfer then affords an intermediate in which both the carbonyl and the hydroxyl group have been activated, leading to the electrostatically driven macrolactonization (Scheme 32). This "double activation" has been

confirmed and a mechanism involving ketene formation was ruled out by deuterium labeling and kinetic studies.[115,116]

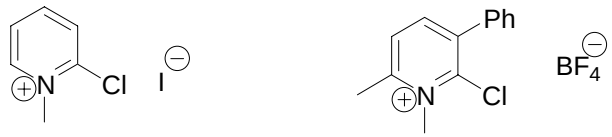


Scheme 32: Proposed mechanism of Corey-Nicolaou synthesis

The "classical" Corey-Nicolaou method has been used in large a number of total syntheses and synthetic applications: *e.g.* zearalenone,[113] brefeldin,[117-119] tylonolide.[120,121]

4.5.2 The Mukaiyama methods

The use of 1-methyl-2-chloropyridinium iodide **124** (Figure 9) as an efficient reagent for the macrolactonization of ω-hydroxy acids was introduced by Mukaiyama in 1976.[122] The mechanism involves (Scheme 33) chloride substitution by the carboxylate ion to give a highly activated acyloxypyridinium species **125** which then undergoes macrolactonization.

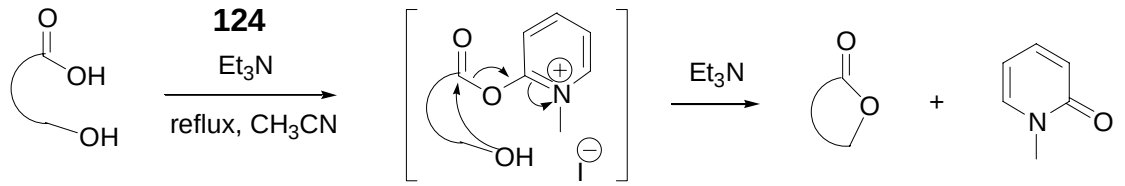


124 **124a**

Figure 9: Structure of Mukaiyama's reagents

Mukaiyama has described further developments of this reagent to suppress under the cyclization conditions the decomposition of the pyridinium salt by attack of triethylamine either

on the 1-methyl group to form 2-chloropyridine or on the pyridinium ring to form pyridinium salts. They synthesized a new pyridinium salt, 2-chloro-6-methyl-1,3-diphenylpyridinium tetrafluoroborate **124a** to avoid these side reaction.[114,123] Indeed, in the presence of benzyltriethylammonium chloride and a hindered non-nucleophilic base such as a 2,6-disubstituted pyridine, this reagent gave better yields in the macrolactonization.



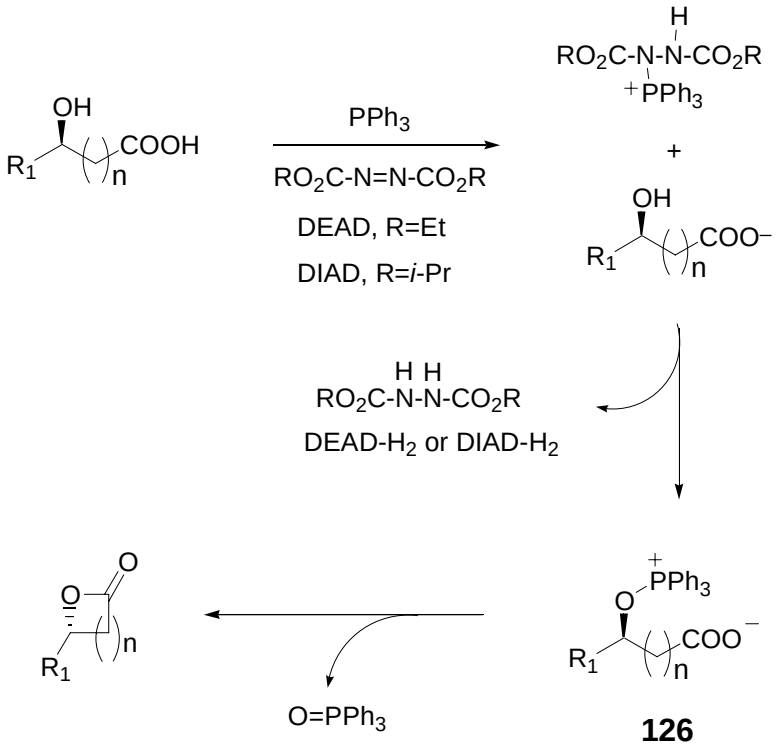
125

Scheme 33. Mechamism of Mukaiyama macrolactonization

4.5.3 Mitsunobu reaction

In 1976 Mitsunobu described a macrolactonization protocol to obtain medium and large macrolactones. This methodology is based on the activation of the seco-acid alcohol using diethyl azodicarboxylate (DEAD) and triphenylphosphine.[123-127] Initially diolides were usually obtained as the major products for the medium ring lactones,[128,129] and the Mitsunobu reaction has long been considered as a selective method to obtain diolides. A modification was introduced by Steglich in 1991 during the synthesis of combrestatin analogues.[130] Using the classical Mitsunobu protocol, the diolide was obtained as the major product (diolide 40%, macrolactone 25%), but with slow addition of the seco-acid to DEAD-triphenylphosphine, the macrolactone was the major product (macrolactone 59%, diolide trace yield) (Scheme 34).

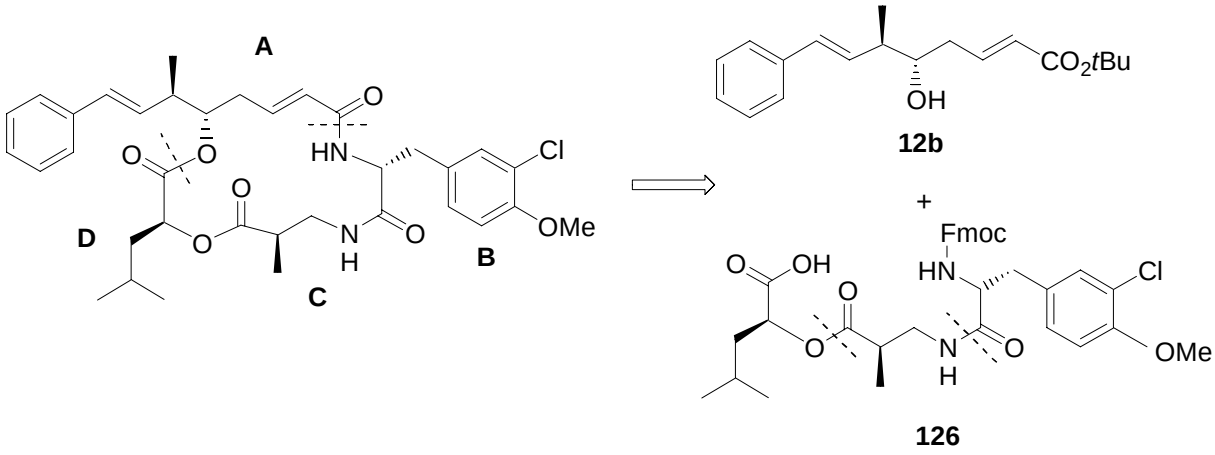
In the reaction mechanism, the key intermediate is an alkoxyphosphonium salt **126** produced in situ, and the macrolactonization proceeds via an intramolecular S_N^2 reaction with inversion of the alcohol configuration.



Scheme 34: Reaction steps and mechanism of the Mitsunobu lactonization

5. Goal of research

As depsipeptides, the Cryptophycins are of particular interest in the context of the synthesis of natural product-like libraries. In principle, synthesis of this type of cyclic ring systems can be approached using various strategies,[131] such as the classical solution head-to-tail cyclization, the solid phase cyclorelease strategy[132] or on-support cyclization using backbone or side chain attachment.[133] The molecule of Cryptophycin-3 offers vast scope to arrange different amino or hydroxy acids to test its biological activity. According to previous total syntheses, it can be acquired by many different retrosynthetic pathways. The most convenient one is the division of fragment A **12b** and the tripeptide fragment **161**, which can be the assembly of different amino acids or hydroxy acids (Scheme 35). In this point we need a strategy where fragments A and various types of tripeptide fragment can be produced efficiently and in large scale.



Scheme 35. Retrosynthetic anylysis of Cryptophycin-3 (**5**)

Several strategies for the total synthesis of Cryptophycins and its analouges were reported by different working groups, which all use the classical solution phase head-to-tail cyclisation.

In the first synthesis by Tius et al.[24] there is a drawback for the synthesis of fragment A. According to them, for the large scale synthesis of compound **19** from **18** by bromination with NBS, a diastereomeric tetrahydrofuryl alcohol was formed as byproduct (Scheme 2). In the assembly of fragments they coupled first the expensive fragment A with fragment B, but this is not so clever for the synthesis of such a macrocycle.

In the first steps of the synthesis of fragment A Leahy-Gardiner established the stereochemistry of C7 and C8 by coupling two asymmetric compounds: (*R*)-mandeldehyde derivative **29** and boron enolate **30** (Scheme 6).[45] But the starting materials are expensive and the Evans aldol reaction is limited to a 2 g scale, in a large scale it gives a diastereomeric mixture of **31**.

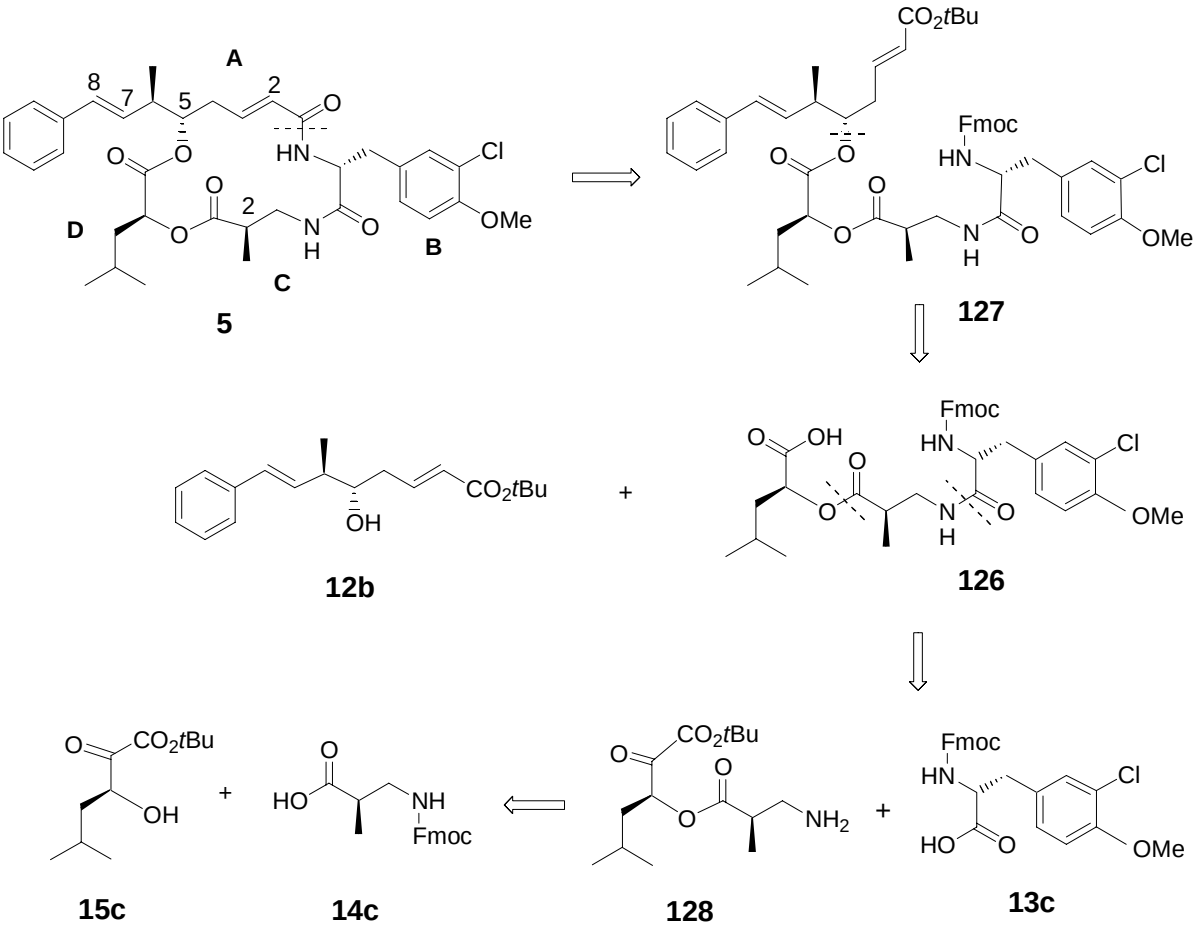
Tius and Li used a hetero Diels-Alder reaction for the synthesis of A, where diene **43** was used as 9/1 mixture of geometrical isomer which is reflected in the ratio of Diels-Alder reaction products.[51] The exposure of crude Diels-Alder product **44** to trifluoroacetic acid gave 4/1 mixture of bicyclic products **45** and **46**. Furthermore the protection reaction of syn C7-C8 diol in compound **47** is very sensitive, which easily gives a tetrrahydrofuran derivative with final compound up to 30% yield (Scheme 9).

Therefore, the objective of our study is the development of simple and reliable synthetic strategy, where the fragment A can be made in an efficient way as well as the synthesis of tripeptide fragment allows the solid phase assembly to make a large number of such macrocycles.

6. Results and discussion

6.1 Retrosynthetic analysis of Cryptophycin-3 (5)

In our retrosynthetic analysis, Cryptophycin-3 (**5**), was obtained from the protected *seco*-compound **127** which was partitioned into two halves: 5-hydroxy ester **12b** (unit A) and tripeptide derivative, acid **126** (Scheme 36). The latter is composed of the depsipeptide fragment **128**, and phenylalanine derivative **13c** (unit B). Ester **128** could be synthesized by the coupling between β-amino acid **14c** (unit C) and α-hydroxy ester **15c** (unit D).



Scheme 36: Retrosynthetic disconnection of Cryptophycin-3 (**5**)

6.2 Synthesis of 5-hydroxy ester 12b (unit A)

The synthesis of fragment A has to address the attachment of the aryl ring and the establishment of the *anti*-stereorelationship at C5 and C6. Therefore we sought to develop a

route where the stereochemistry is based on aldol methodology, followed by *anti* selective hydroboration and flexible introduction of the aryl group by Wittig olefination. The remainder of the carbon atoms of fragment A were added by Wittig-Horner homologation (Figure 10).

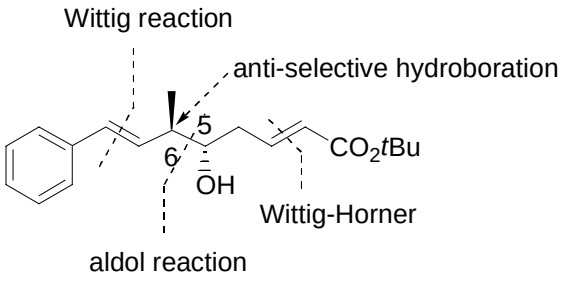
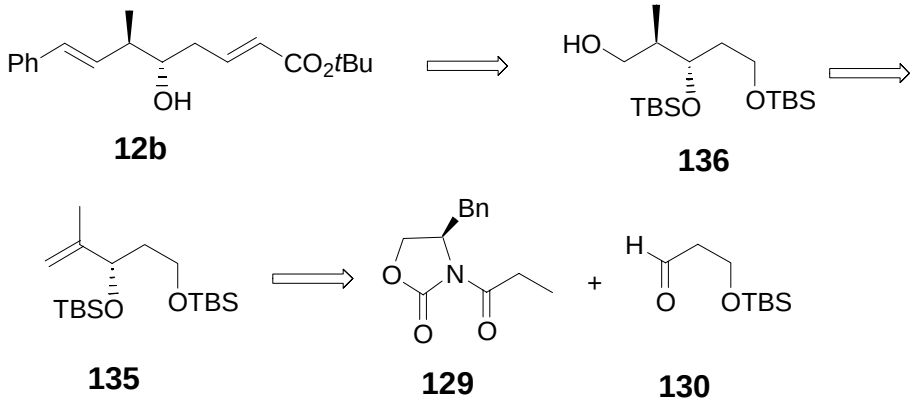


Figure 10: Overview of the synthesis of unit A

6.2.1 Retrosynthetic analysis of 5-hydroxy ester 12b (unit A)

The retrosynthetic analysis of 5-hydroxy ester **12b** is shown in the Scheme 37. The α,β-unsaturated erter **12b** was acquired from the *anti*-alcohol **136** which was prepared from the disubstituted allyl ether **135** by hydroboration reaction. The terminal alkene **170** was produced from the aldol product of aldehyde **130** and chiral auxiliary **129**.

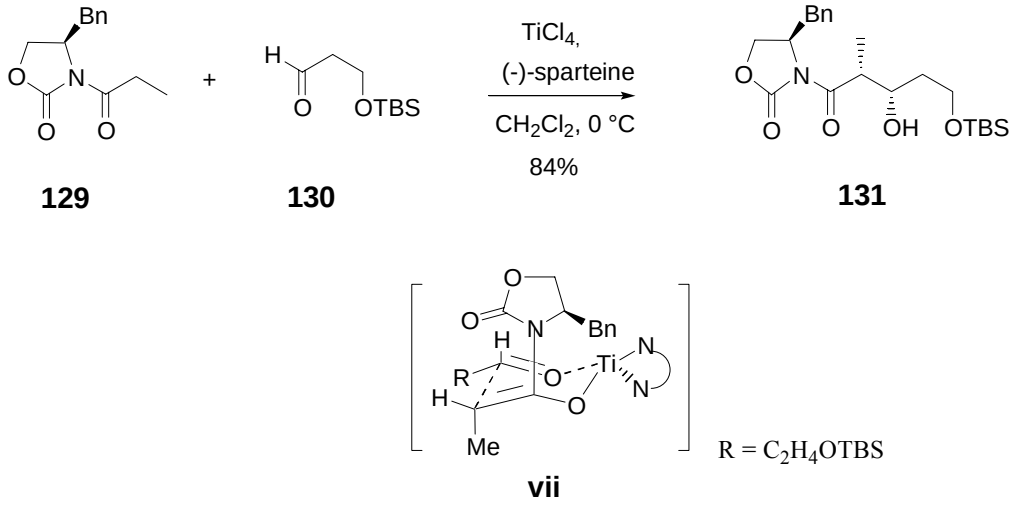


Scheme 37: Retrosynthetic pathway to 5-hydroxy ester

6.2.2 Aldol reaction with Evans methodology

The key reaction in the synthesis of 5-hydroxy ester **12b** was an asymmetric Evans aldol

reaction,[134-136] which provided the two stereocenters at C5 and C6. In our synthesis we exploited a modified Evans procedure which according to the literature also results in "Evans" *syn*-type products.[100,78] The N-propionyloxazolidinone **129** was treated with $TiCl_4$ (1 eq.) and (−)-spartein (2.5 eq.) at −78°C and led to *Z*-enolate, which on further reaction with the protected aldehyde **130** produced the *syn*-aldol product **131**. This reaction was carried out through the non-chelated transition state **vii** with an excellent diastereoselectivity (>99% *de*) (Scheme 38).



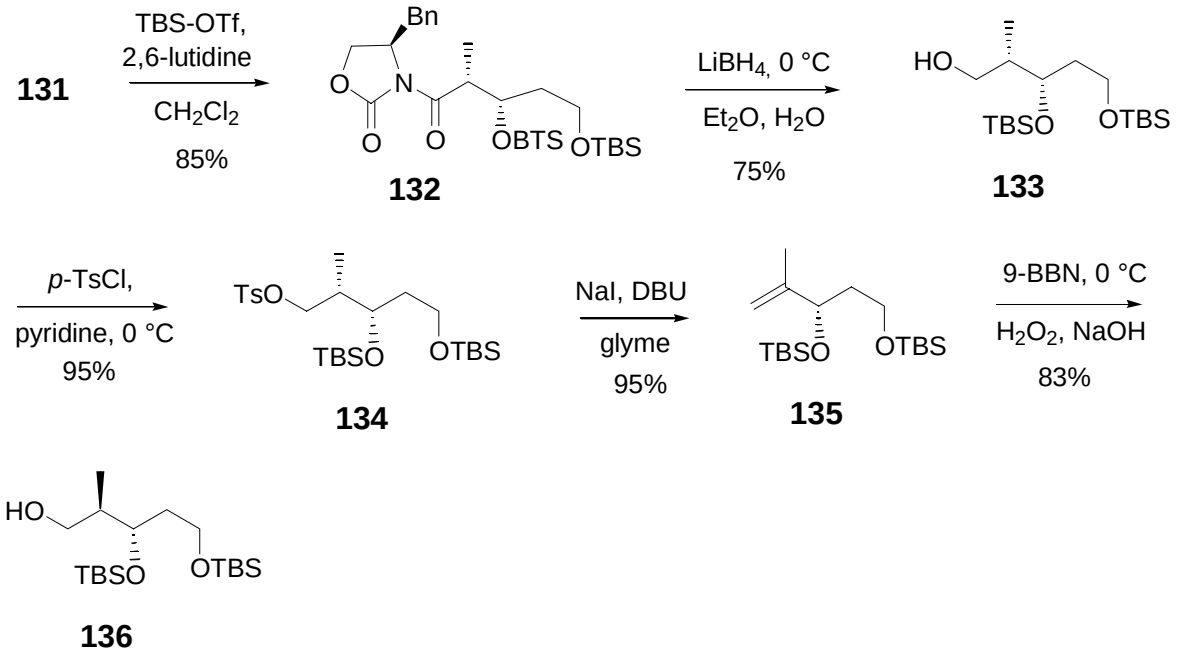
Scheme 38: Evans aldol reaction

In this reaction we used 2.5 eq. (−)-sparteine instead of diisopropylethylamine or tetramethylethylenediamine (TMEDA) to generate the titanium enolate of N-propionyl-oxazolidi-none **129**, because it changed the rate of the reaction and diastereoselectivities.[100] The isolated yield with (−)-sparteine was improved substantially compared to TMEDA. Another important point is that $TiCl_4$ and (−)-sparteine were used directly as received without further purification in contrast with *n*-Bu_2BOTf (now it can also possible to buy), which was needed to be prepared in advance.[100]

6.2.3 Further synthetic pathway to 5-hydroxy ester **12b**

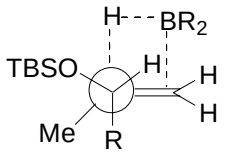
After Evan's aldol reaction, the *syn*-aldol product **131** was purified by flash chromatography and the hydroxyl group was protected with *tert*-butyldimethylsilyl triflate (TBSOTf) in presence of lutidine leading to **132** (Scheme 39). This protection reaction was followed by reductive removal of the auxiliary with lithium borohydride producing *syn*-alcohol **133**.

Thereafter a terminal double bond had to be introduced, which was accomplished in high yield method by forming tosylate and base induced elimination.[137,78] The tosylation of alcohol **133** was done by *para*-toluenesulfonyl chloride (*p*-TsCl) in pyridine and Subsequent heating of the tosylate **134** at reflux in glyme (1,2-dimethoxyethane) in presence of sodium iodide and DBU effected clean elimination to provide the disubstituted allyl ether **135**. The stereocenter destroyed in this proces was reintroduced through diastereoselective hydroboration,[138,139] by using 9-BBN (bicyclic 9-borabicyclo[3.3.1]nonane), which gave the *anti* product **136** with high diastereoselectivity (9:1) and high yield.



Scheme 39: Synthetic pathway to compound **136**

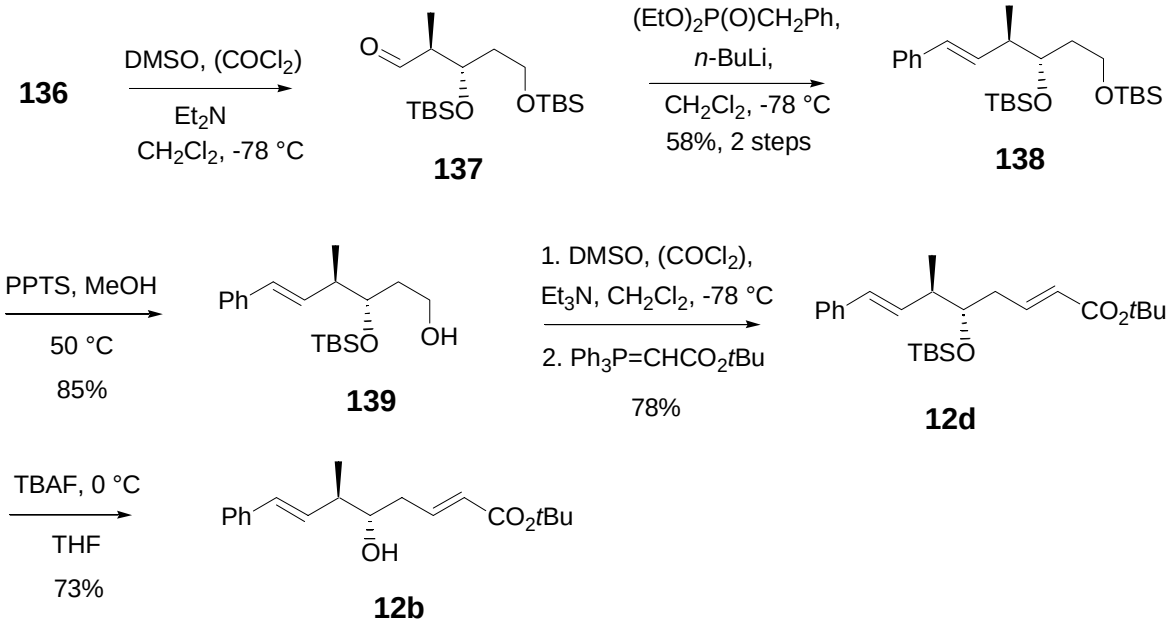
The stereochemistry of the hydroboration can be explained by the Houk model (Figure 11).[139] According to this model, the *anti* diol is produced via a staggered transition state **xi** where the substituents R and OTBS occupy the *anti* and outside positions, respectively. Attack of the borane takes place *anti* to the R group which is the best electron doner among the three substituents.



xi

Figure 11: Probable T S of hydroboration reaction

Then the terminal hydroxyl group was oxidized to aldehyde **137** by Swern oxidation, which was then extended by reaction with diethylbenzyl phosphonate, $(EtO)_2P(O)CH_2Ph$ under Wittig-Horner conditions to produce the styrene derivative **138** (Scheme 40). Selective cleavage of the primary silyl protecting group from **138** was done by pyridiniumtoluene-4-sulfonate (PPTS) leading to alcohol **139**. After that, one-pot Swern oxidation of **139** and Wittig treatment[140] with $Ph_3P{=}CHCO_2tBu$[141-143] provided the unsaturated ester **12d** in good yield. Finally, the removal of the secondary silicon protecting group by using tetra-*n*-butylammonium fluoride (TBAF) furnished the key building block **12b**.



Scheme 40: Synthetic pathway to the 5-hydroxy ester **12b**

According to the ^{1}H-NMR spectrum (Figure 12), the hydroxy acid **12b** was diastereomerically pure. The expanded region clearly showed the chemical shift value of methyl group (1.07 ppm) and the proton at C-5 (3.58 ppm).

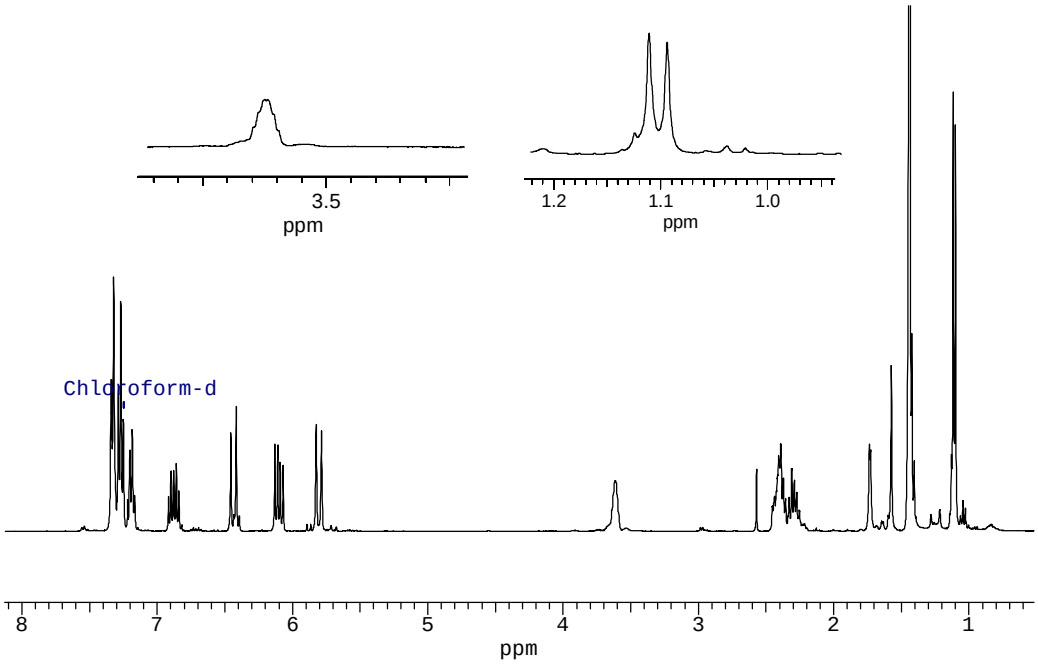
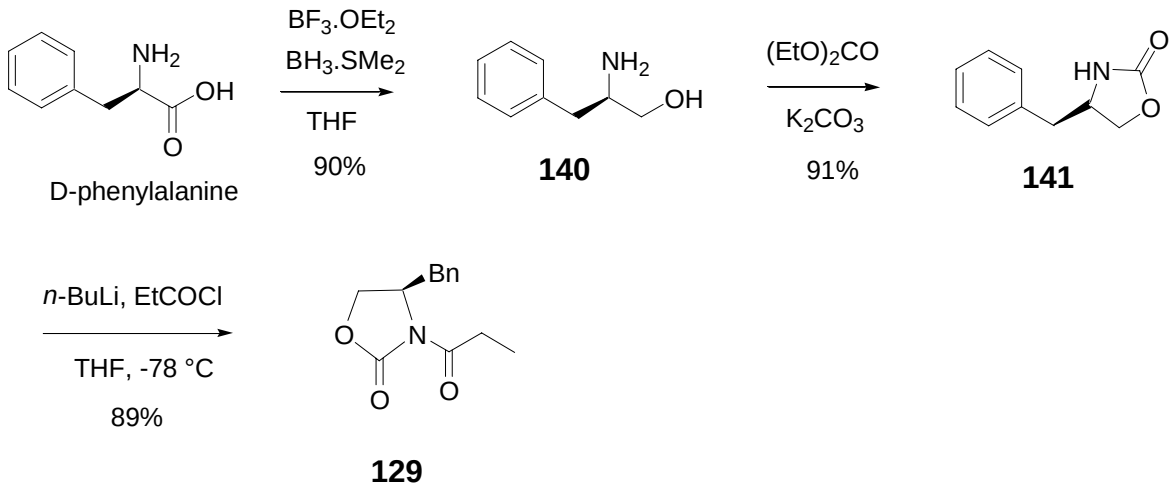


Figure 12: ^{1}H-NMR spectrum of hydroxy acid **12b**

The preparation of oxazolidinone **141** was done by reduction of D-phenylalnine with borane dimethylsulfide to amino alcohol **140**, followed by condensation with diethylcarbonate in presence of catalytic amount of potassium. Further deprotonation with *n*-butyllithium and subsequent acylation with propionyl chloride led to the N-propionyloxazolidinone **129** in good overall yield for the three steps (Scheme 41).[134]



Scheme 41: Synthesis of acylated Evans auxiliary **129**

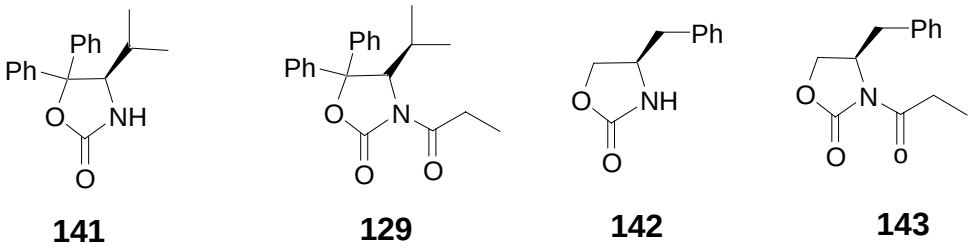
6.2.4 Aldol reaction using the Seebach auxiliary

For the synthesis of 5-hydroxy ester **12d** we also attempted the aldol reaction using the Seebach auxiliary, 5,5-diphenyl-oxazolidinone **142** (Figure 13). This chiral auxiliary is easily accessible from valine and a useful alternative to the widely employed Evans auxiliary **141**. We chose this auxiliary because it has a number of advantages:[134]

i) The N-acyl derivative **143** of this auxiliary is more prone to crystallize than the corresponding derivative of other auxiliaries.

ii) Recycling of this auxiliary after cleavage can be achieved by simple filtration, because of the high insolubility of auxiliary **142** in most organic solvents.

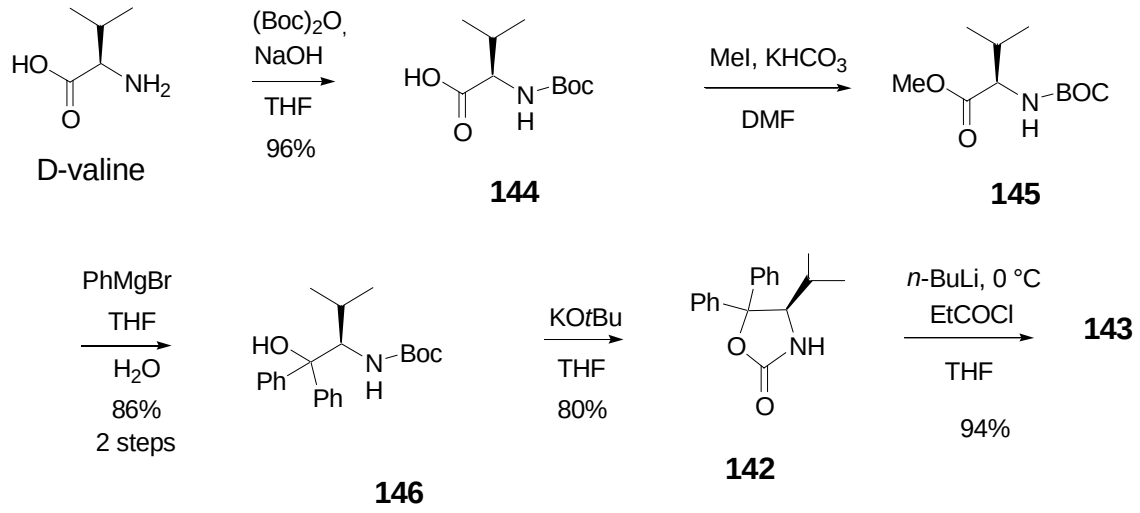
iii) Acylation of the auxiliary **142** with *n*-BuLi can be performed at elevated temperature (0° as compared to –78 °C for Evans auxiliary **141**), which is an energetical and apparative advantage, especially in large scale applications.



141 **129** **142** **143**

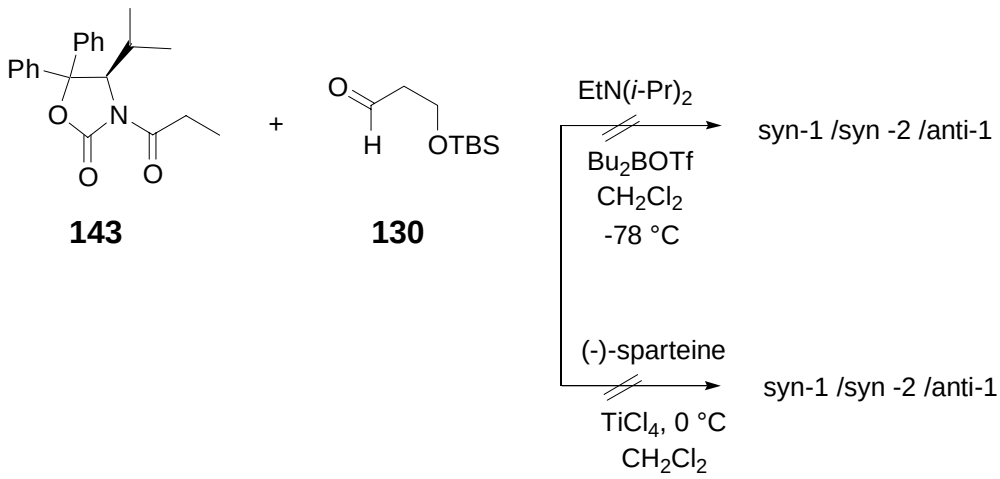
Figure 13: Structure of Seebach and Evans auxiliary

For the synthesis of N-propionyl-oxazolidin-2-one **143** (Scheme 42) we first converted D–valine to (*R*)-N-(t-Butyloxycarbonyl) valine (**144**) with Boc-anhydride which was then methylated by methyl iodide and potassium hydrogen carbonate to get the fully protected valine **145**. This was added to freshly prepared phenyl magnesium bromide (prepared from phenyl bromide and magnesium) and led to tertiary alcohol **146**. This addition reaction was followed by the cyclization with potassium *tert*-butoxide and created oxazolidinone **142** in good yield. Acylation of oxazolidinone **142** was done with propanoyl chloride and *n*-BuLi to provide the required N-propanoyl-oxazolidin-2-one **143**.[144,134]



Scheme 42: Synthesis of N-acyloxazolidin-2-one **143**

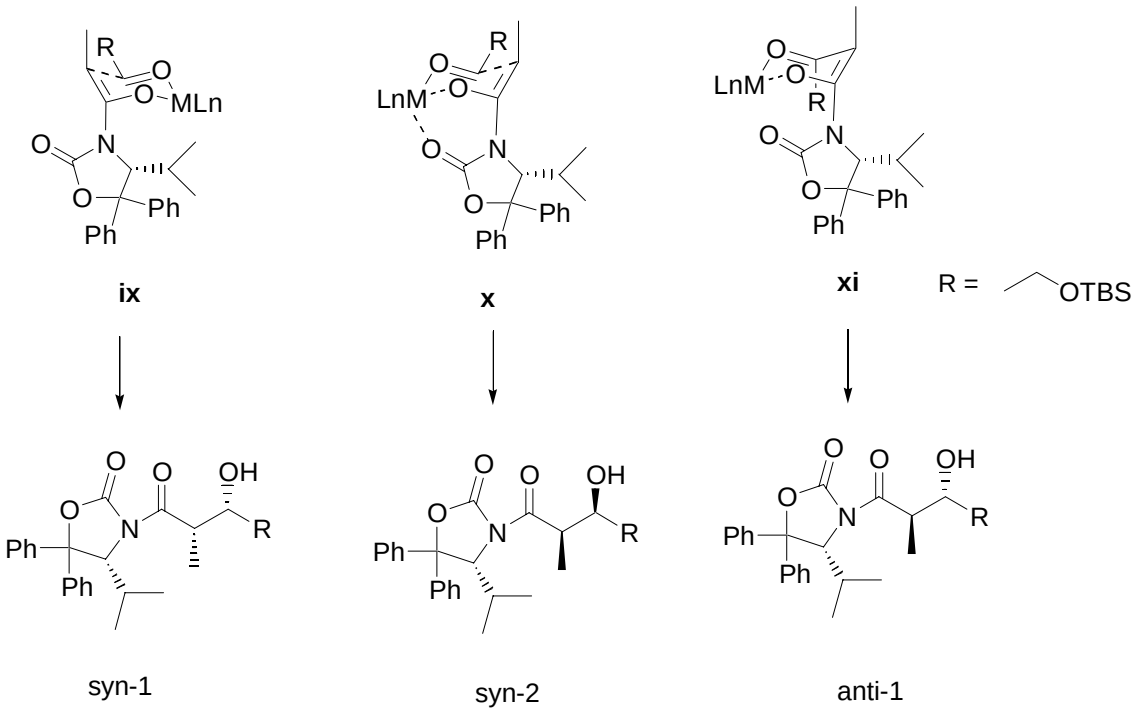
In the aldol reaction N-acyloxazolidin **143** reacted with protected aldehyde **130** (Scheme 43) at –78 °C using *n*-BuB_2OTf and *i*-Pr_2NEt. This resulted in a mixture of aldol products with low diastereose-lectivity and low yield. Then we tried different reagents, $TiCl_4$ and (–)-sparteine, but in this case we also got a mixture of diastereomers with low *de*.



Scheme 43: Aldol reactions using Seebach auxiliary

These stereochemical results of the aldol reactions could be explained by a chair-like transition-state model.[144] It was assumed that the Bu_2B- and $TiCl_4$-enolate derivatives gave syn-1 and syn-2 aldol products via intermediate **ix** and **x** (Scheme 44). The anti-1 aldol product moght have formed via intermediate **xi**, which can be explained by an attractive

interaction between the silicon protecting hydroxyl group in **130** and the two Ph groups in the oxazolidinone **143**.

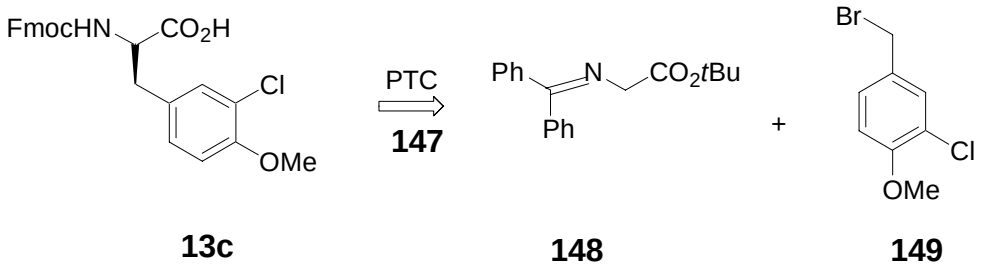


Scheme 44: T. S. of Seebach aldol reaction

Therefore we decided to proceed our synthesis of unit A using Evans aldol methodology, inspite of many advantages of oxazolidinone **142**.

6.3. Synthesis of amino acid 13c (unit B) by enantioselective alkylation

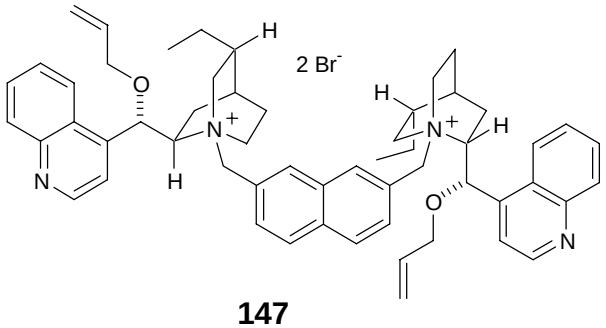
The phenylalanine derivative **13c** is usually prepared from D-tyrosine through chlorination of the aromatic core. Since these conditions would probably not be compatible with the Fmoc protecting group, a new synthesis based on the highly enantioselective alkylation of the glycine derivative **148** was developed.[145-149] The alkylation reaction was done with glycine imine **148** with compound **149** in presence of the chiral cinchona derived catalyst **147** (Figure 14) under phase-transfer conditions (Scheme 45).



13c **148** **149**

Scheme 45: Retrosynthetic pathway to amino acid **13c**

Phase-transfer catalysis (PTC) is a useful method for practical synthesis, because of its operational simplicity and mild reaction conditions, which enables this method to be applied to industrial processes.[150-154] This asymmetric alkylation is based on the fact that the introduction of a bulky subunit at the nitrogen atom of cinchona alkaloids leads to an enhancement of the stereoselectivity. This enhancement of the stereoselectivity is a result of the scereening effect between each cinchona unit, which can make the substrate approach from preferentially one direction. Among the dimeric catalysts, cinchona-alkaloid-derived catalysts, which have a naphthalene moiety as a bridging ligand turned out to provide good *ee*-values for the alkylation of glycine imine derivatives. High selectivities are also obtained if the tertiary nitrogen atom is alkylated with fluorinated benzyl[155] or anthracenylmethyl halides.[156] This electronic effect is attributed to the formation of tighter ion pairs.



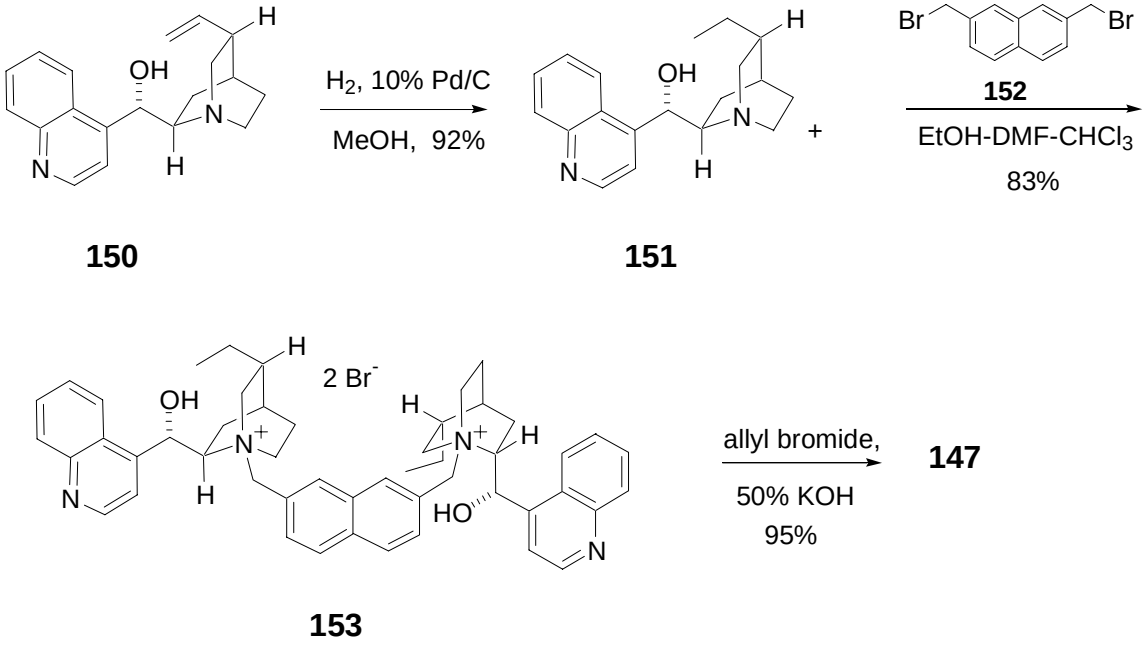
147

Figure 14. Structure of chiral catalyst **147**

6.3.1 Synthesis of chiral catalyst **147**

For the synthesis of chiral catalyst **147**[145] we used commercially available (+)-cinchonidine **150** which was reduced by Pd/C under hydrogen atmosphere to hydrocin-

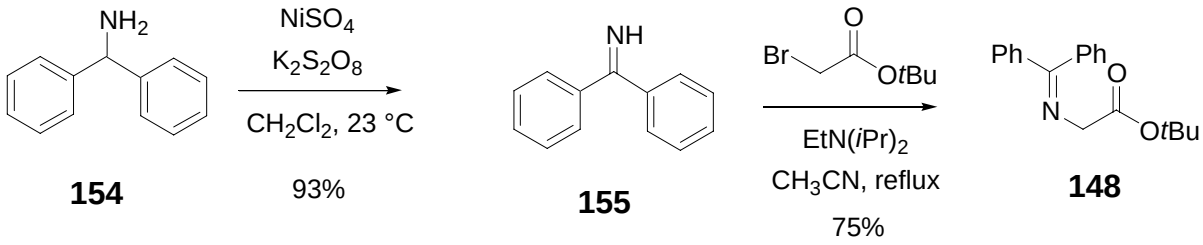
chonidine **151** (Scheme 46). Amide **151** then reacted with dibromonaphthalene **152** at 100 °C to generate dimeric compound **153**. In this reaction two molecules of **151** reacted with one molecule of dibromonaphthalene **152**, which was prepared from dimethylnaphthalene by bromination with *n*-bromosuccinimide (NBS). After that, the hydroxyl group of **153** was reacted with allyl bromide in presence of 50% potassium hydroxide to get the desired chiral catalyst **147.**



Scheme 46: Synthesis of chiral catalyst **147**

6.3.2 Preparation of N-(Diphenylmethylene)glycine *tert*-butyl ester **148**

The synthesis of N-(Diphenylmethylene)glycine *tert*-butyl ester **148**[157] was started by nickel catalyzed oxidation of amine **154**. First commertially available **154** was dehydrogenated under catalytic conditions with nickel sulfate $NiSO_4$ and aqueous basic solution of potassium thiosulfate ($K_2S_2O_8$) to diphenylmethanimine **155** quantitatively. Then **155** reacted with *tert*-butyl-2-bromo-acetate in presence of diisopropylethylamine to get the desired glycine imine **148** (Scheme 47).



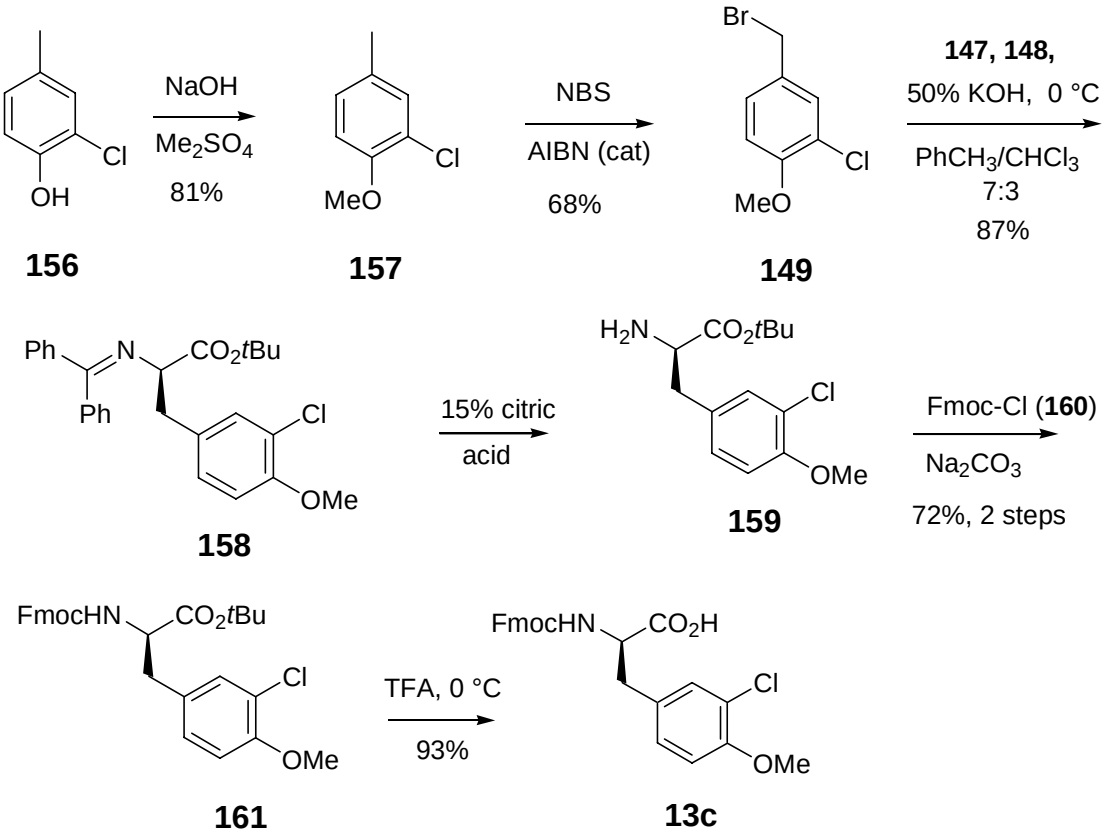
154 **155** **148**

Scheme 47: Synthesis of glycine derivative **148**

6.3.3. Synthetic pathway to amino acid **13c** (unit B)

In the synthetic pathway to phenylalanine derivative **13c** (Scheme 48), the hydroxyl group of phenol **156** was converted to methyl ether **157** with dimethyl sulfate for the making of the tyrosine aromatic core. The methyl group of **157** was then brominated with N-bromosuccinimide (NBS) to form benzylic bromide **149**. The enantioselective alkylation[145] of the glycine imine **148** with benzylic bromide **149** was done with the use of the chiral cinchona-derived phase-transfer catalyst **147** to produce benzylated imine **158** in good yield. The reaction was carried out with 50% aqueous potassium hydroxide in toluene and chloroform (7:3). The *ee* was determined to be 96 % by HPLC using a chiral column (DAICEL Chiral OB-H, 250 × 2.6 mm; 98:02 heptane and isopropanol, flow 0.5 mL min^{-1}, t_{minor} = 8.82 min, t_{major} = 9.79 min).

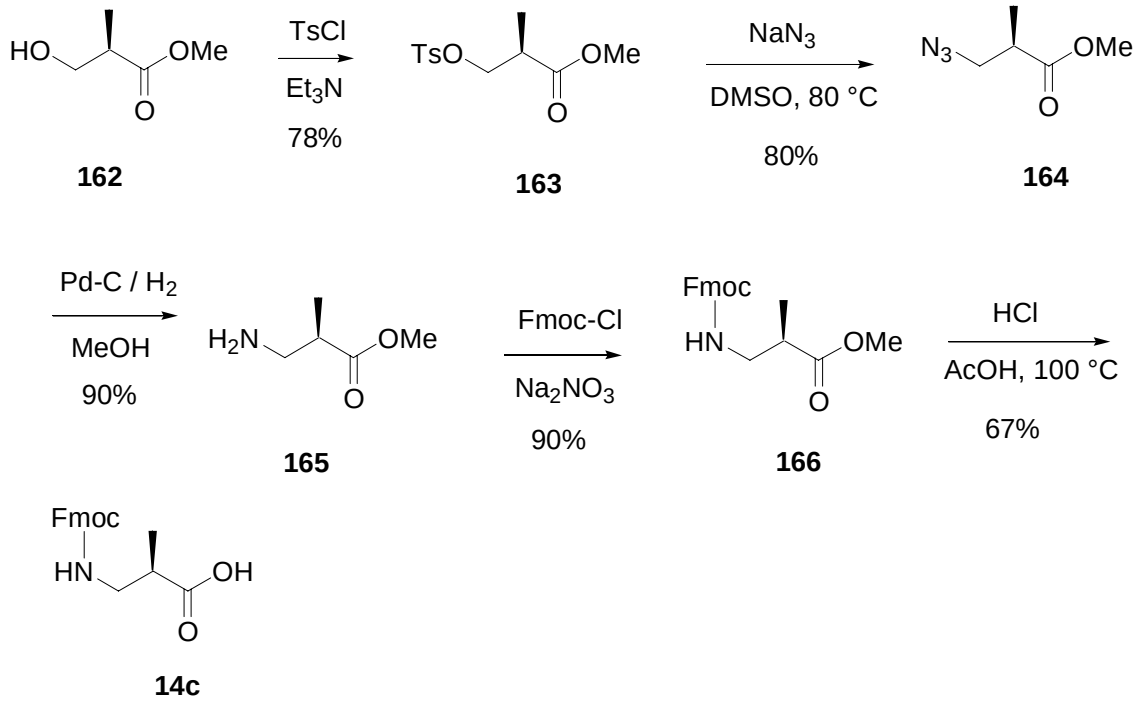
Hydrolysis of imine **158** was done by 15% citric acid to give free amine **159**. The free amino group of **159** was then immediately protected with (fluorenylmethoxy)carbonyl chloride (Fmoc-Cl) (**160**) and provided fully protected amino acid **161**. Treatment of **161** with trifluoroacetic acid (TFA) furnished the desired amino acid **13c**.



Scheme 48: Synthetic pathway to phenyl alanine derivative **13c**

6.4 Synthesis of β-amino acid 14c (unit C)

The synthesis of β-amino acid **14c** started from commercially available (–)-methyl D-hydroxy isobutyrate **162** (Roche ester) as shown in Scheme 49. The hydroxyl group of **162** was first tosylated with tosyl chloride and led to tosylate **163**.[158-161] Substitution of the tosyl group with azide was done by using sodium azide in DMSO forming azide **164**. Then, catalytic hydrogenation[162] of the azide function with Pd/C under hydrogen atmosphere provided the free amine **165**.[163,164] The amino group was then immediately protected under Schotten-Baumann conditions using (fluorenylmethoxy)carbonyl chloride (Fmoc-Cl) in methanol and sodium carbonate and resulted in Fmoc protected ester **166**. Hydrolysis of the ester function of **166** to derive the amino acid **14c** was then achieved under acidic conditions.[165]

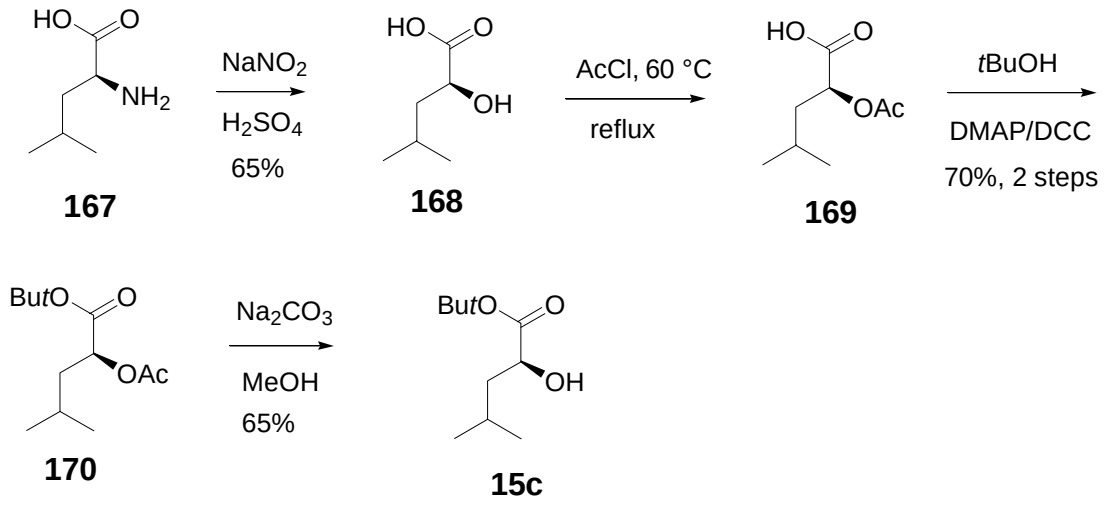


162 **163** **164** **165** **166** **14c**

Scheme 49: Synthetic pathway to β-amino acid **14c**

6.5 Synthesis of α-hydroxy ester 15c (unit D)

The α-hydroxy ester **15c** was prepared from commercially available L-isoleucine **167** by diazotazation[166,167] as shown in Scheme 50. L-isoleucine was diazotized with sodium nitrite and sulfuric acid giving α-hydroxy carboxylic acid **168** (isoleucic acid) with retention of configuration at the α-carbon due to the neighboring-group participation. The hydroxyl group of **168** was protected with acetyl chloride to give hydroxyl protected acid **169**. After that the protection of the carboxylic group as *t*-butyl ester with *t*-butyl alcohol using dicyclohexylcarbodiimide (DCC) and 4-(N,N-dimethylamino)pyridine (DMAP) produced fully protected **170**. Finally the acyl group was deprotected with sodium carbonate in methanol to give the required α-hydroxy ester **15c**.

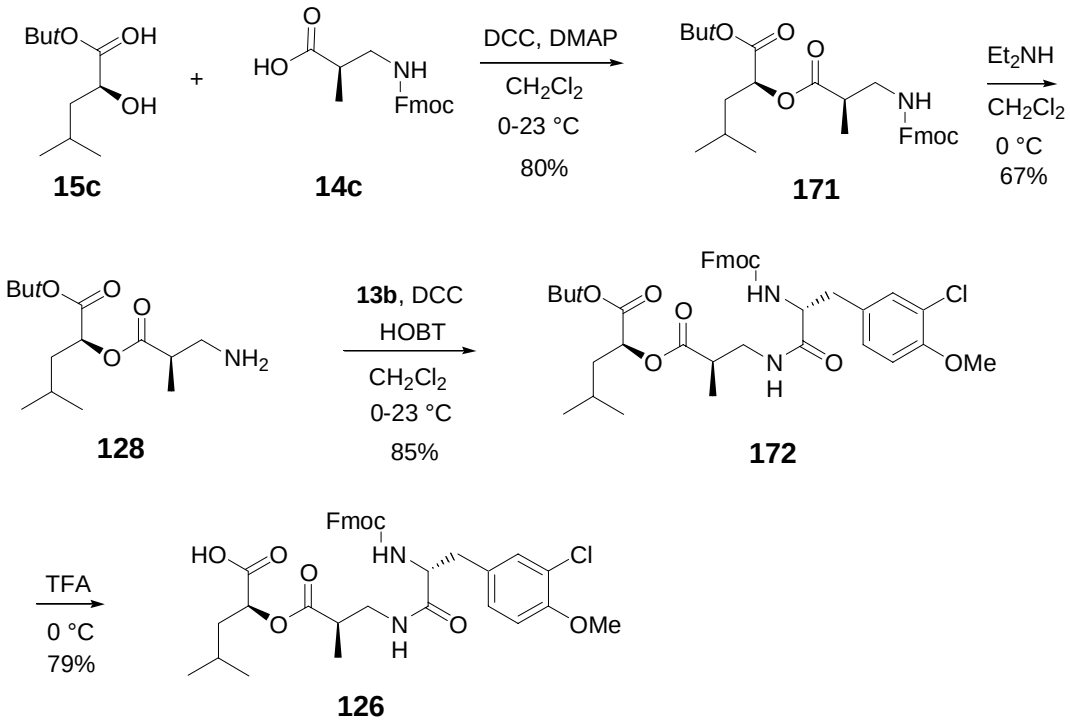


Scheme 50: Synthetic pathway to α-hydroxy ester **15c**

6.6 Assembly of tripeptide derivative acid 126 (D + C + B section)

The assembly of tripeptide derivative **126** began with ester formation between the secondary hydroxyl group of **15c** (unit D) and the carboxylic acid function of protected β-amino acid **14c** (unit C) (Scheme 51). Use of dicyclohexylcarbodiimide (DCC) and (N,N-dimethylamino)pyridine (DMAP) in the esterification provided an excellent yield of the dipeptide analogue **171** (D + C section). The treatment of **171** with diethylamine in THF caused cleavage of the Fmoc protecting group, resulting in the primary amine **128**. This compound turned out to be stable towards intramolecular amide formation. After that amide formation in presence of dicyclohexylcarbodiimide (DCC) and 1-hydroxybenzotriazole (HOBT) coupled **128** with the Fmoc protected amino acid **13c** (unit B) and provided the D + C + B section **172**, in good yield.

The next stage in the plan was forming an ester bond between the alcohol of unit A **12b** and the carboxylic group of tripeptide derivative D + C + B section **172**. Accordingly, treatment of **172** with trifluoroacetic acid liberated the C-terminal carboxyl group, giving the tripeptide acid **126**.



Scheme 51: Assembly of tripeptide unit **126**

6.7 Synthesis of protected seco-compound 127 and macrocyclization

For the synthesis of seco compound **127**, coupling through ester bond formation between the secondary hydroxyl group of **12b** and the carboxylic acid function of **126** was tried with different condensing reagents as shown in Table 2.

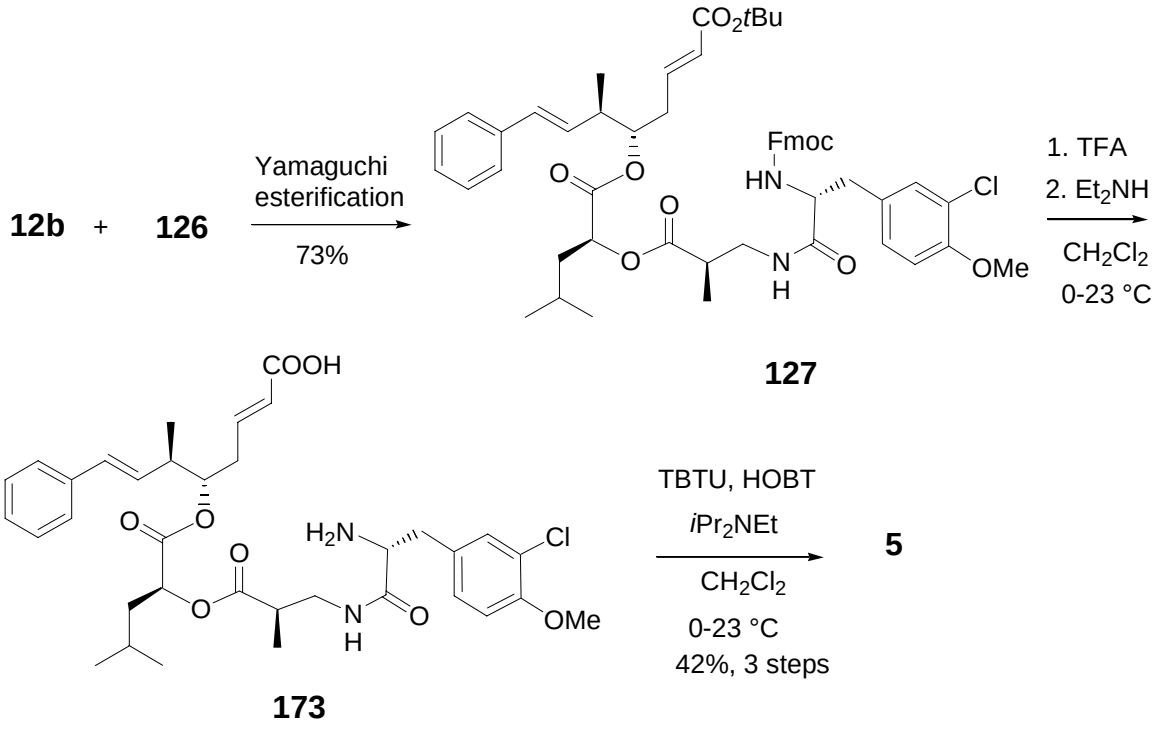
Table 2: 1) by using DCC / DMAP, yield was very poor due to racemization; the excess DCC and the byproduct was also dificult to separate by column chromatography
2) by using EDCl/ DMAP, yield was also poor
3) by Yamaguchi esterification, the reaction was completely finished with very good yield

Entry	Coupling reagent	Reaction conditions	Yield
1	DCC / DMAP	CH_2Cl_2, 0 °C	25%
2	EDCl / DMAP	CH_2Cl_2, rt	31%
3	2,4,6-trichlorobenzoylchloride/DMAP	DIEA, THF, rt	73%

In the Yamaguchi esterification, the carboxyl group of the tripeptide derivative **126** was activated using theYamaguchi reagent, 2,4,6-trichlorobenzoyl chloride,[167] in presence of

DMAP and diisopropylethylamine (DIEA), and coupled with the hydroxyl group of **12b**. This resulted in fully protected seco compound **127** (Scheme 52).

The *tert*-butyl ester group was then removed with trifluoroacetic acid (TFA) and after evaporating the solvent and excess TFA, the Fmoc group was cleaved with diethylamine (DEA) in the same step to get the intemediate *seco*-compound **173**. The macrocyclization of **173** was achieved using the coupling reagent 2-(1*H*-benzotriazol-1-yl)-1,1,3,3-tetramethyluronium tetrafluoroborate (TBTU) in presence of 1-hydroxybenzo-triazole (HOBT) and diisopropylethylamine (DIEA) leding to the macrocycle **5** in good overall yield.



Scheme 52: Final steps of the synthesis of Cryptophycin-3 (**5**)

The ^{13}C-NMR spectrum shows the pure compound of **5** in Figure 15.

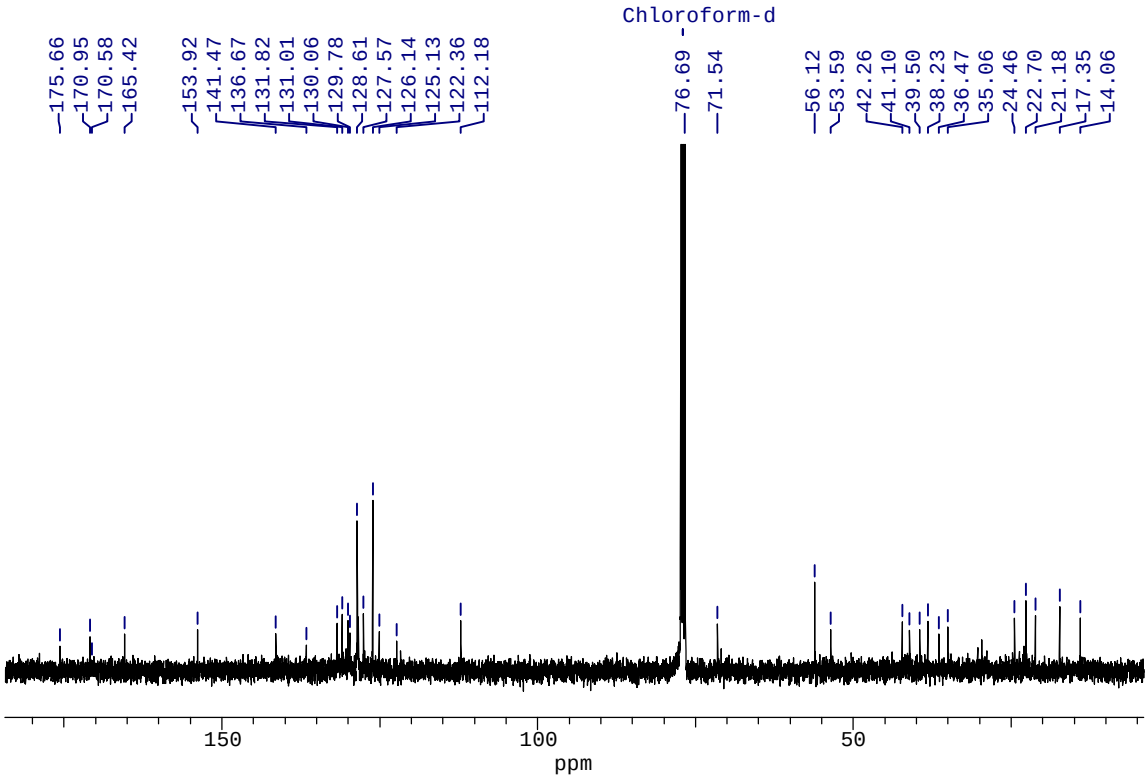
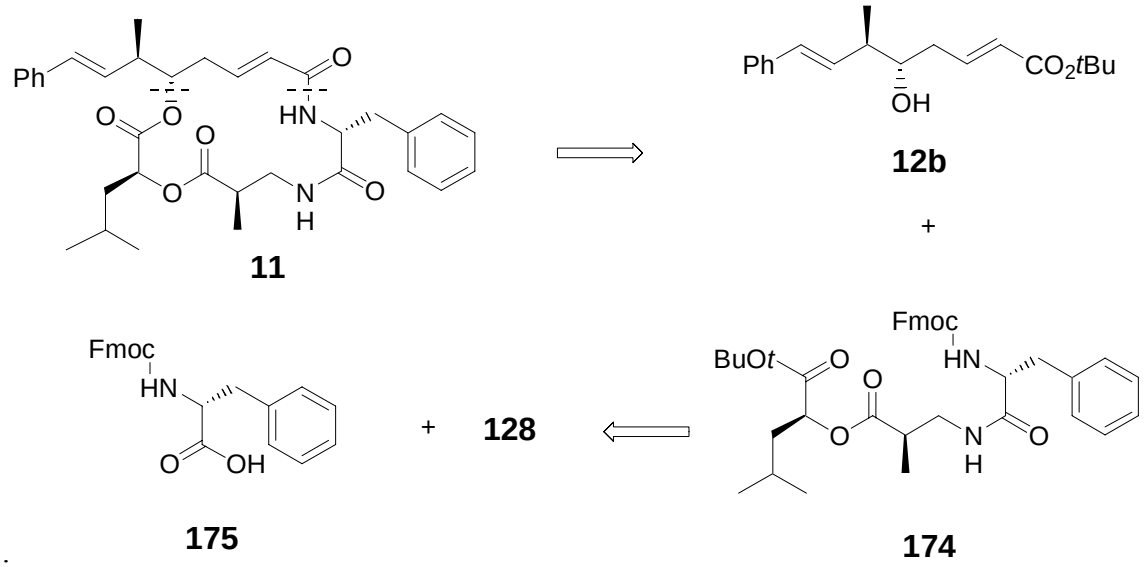


Figure 15: ^{13}C-NMR spectrum of compound **5**

6.8 Synthesis of Cryptophycin analogue 11

6.8.1 Retrosynthetic analysis

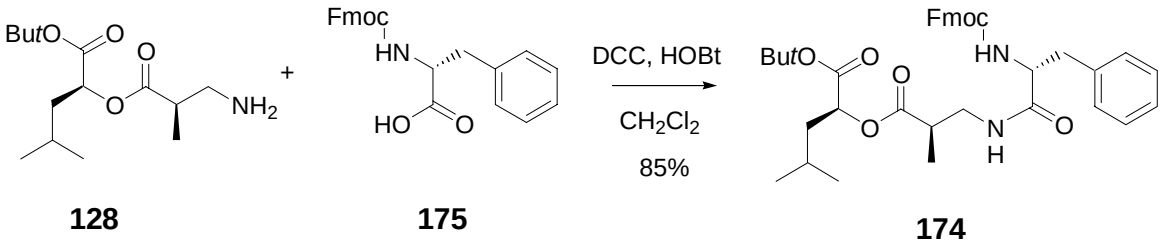
The Cryptophycin analogue **11** was prepared in similar fashion, by the combination of unit A (**12b**) and the tripeptide derivative **174**. This tripeptide derivative **174** is the combination of the dipeptide **128** and Fmoc protected phenylalanine **175** as shown in Scheme 53.



53: Retrosynthetic pathway to compound **11**

6.8.2 Synthesis of the tripeptide derivative **174**

For the synthesis of the tripeptide derivative **174**, we replaced the Fmoc-protected amino acid **13b** with commercially available Fmoc protected (*R*)-phenylalanine **175** (Scheme 54). Acid **175** was coupled with dipeptide **128** in a similar fashion using DCC and HOBt to gave **174**.

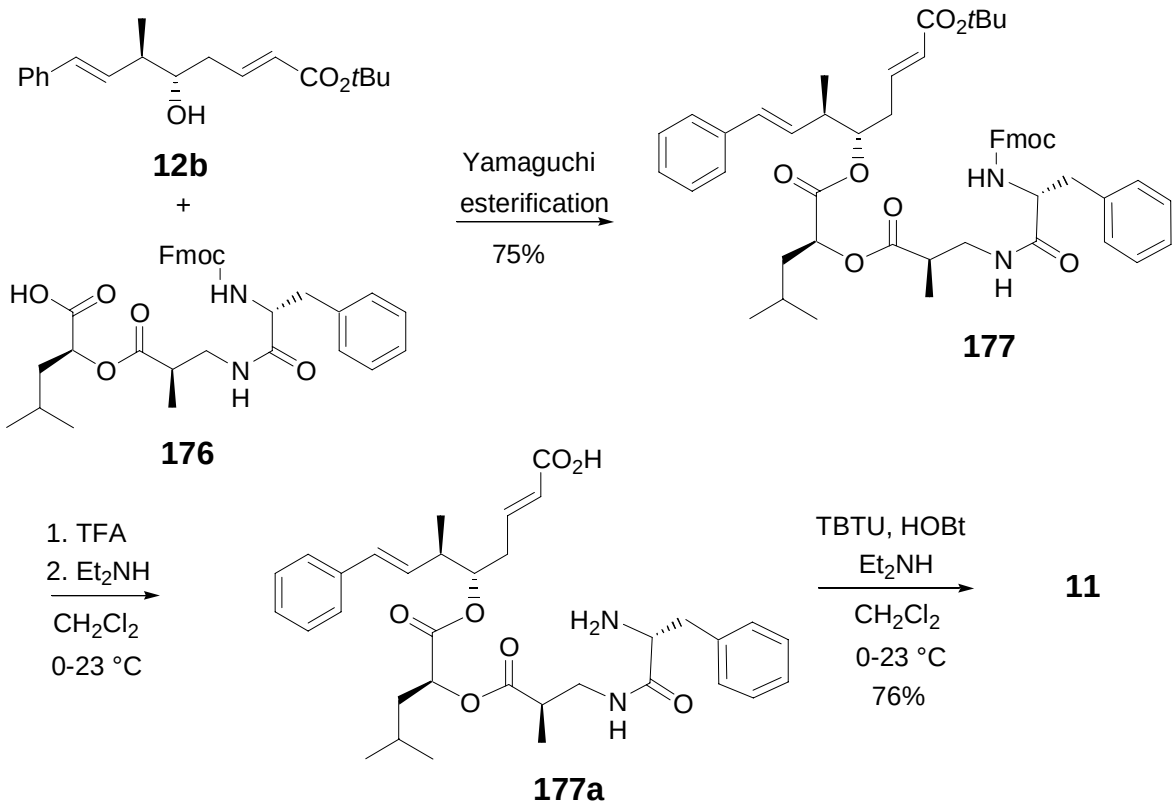


128 175 174

Scheme 54: Synthetic pathway to the tripeptide derivative **174**

6.8.3 Synthesis of *seco*-compound **177** and final cyclization

The ester group of the tripeptide derivative **174** was cleaved with TFA providing acid **176** which was immediately coupled without further purification. Acid **176** was coupled with the hydroxyl group of unit A **12b** using the Yamaguchi reagent and produced ester **177** (Scheme 55). For cyclization first the *tert*-butyl group was cleaved with TFA, followed by removal of Fmoc protecting group with diethylamine resulting in the *seco*-compound **177a**. By using the same coupling reagent TBTU and HOBt in presence of diethylamine the final cyclization led to depsipeptide **11** in good yield.



Scheme 55: Final steps of synthesis of compound **11**

Figure 16 shows the ^{13}C-NMR spectrum of the compound of **11**.

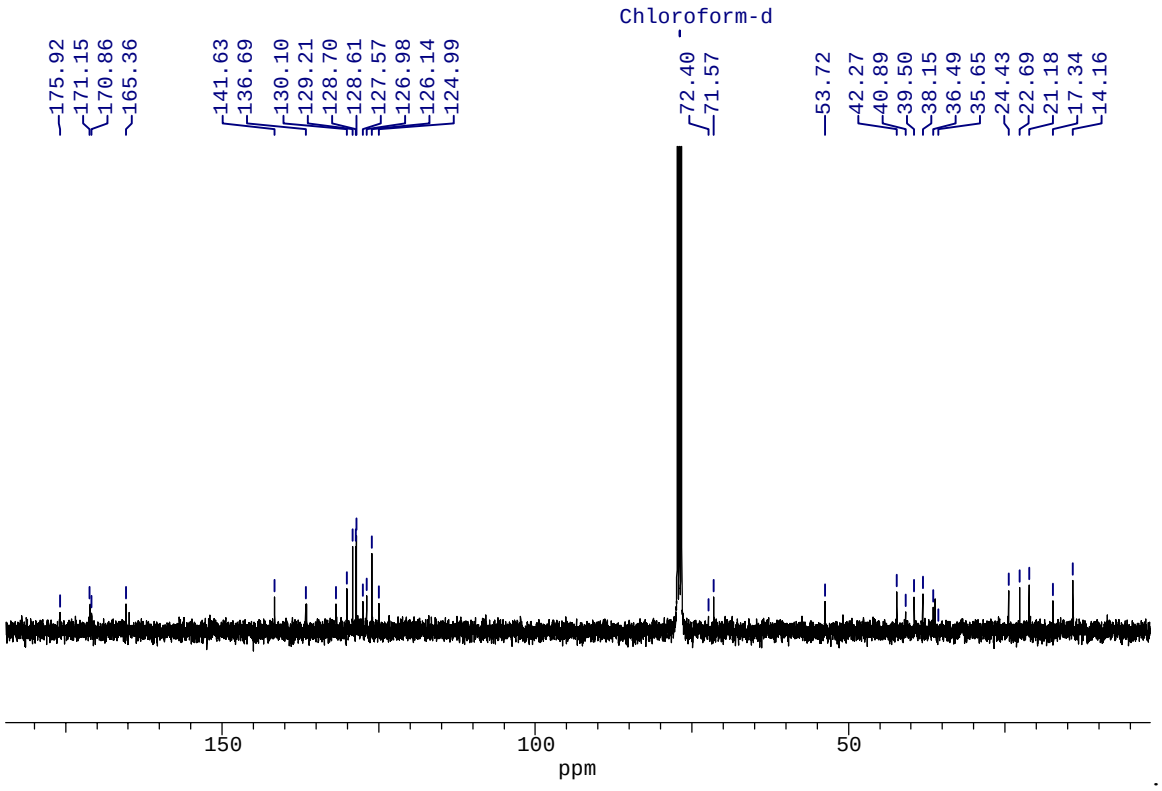


Figure 16: ^{13}C-NMR-spectrum of compound **11**

III. THE PROTEASOME INHIBITOR EPOXOMICIN

1. Biological activity

Proper function of cells is governed by well-coordinated regulatory mechanisms involving numerous interactions among millions of cellular proteins with high degree of specificity. Many important eukaryotic regulatory proteins that are involved in the regulation of various cellular processes are synthesized when required and degraded after performing the desired function in a signal dependent manner. So precise timing of synthesis and degradation is the key to the regulation of many important cellular processes such as cell cycle progression, antigen presentation and cytokine-stimulated signal transduction. Therefore, deregulation of some of these mechanisms may lead to improper functioning of the cell that might lead to adverse consequences in these cellular processes.

Intracellular protein degradation in eukaryotic cells occurs predominantly through a highly regulated proteasomal pathway in which proteins targeted for destruction are ligated to the 76 amino acid polypeptide ubiquitin. Poly-ubiquitinated proteins are recognized by the 19S regulatory domain of the 26S proteasome and through a series of ATP hydrolysis-dependent processes, are deubiquitinated and threaded into the core proteolytic complex, the 20S proteasome, where they are finally proteolyzed into short peptides.[20] Eukaryotic proteasome, an approximately 700 kDa cylindrical-shaped multicatalytic protease complex composed of 28 subunits, is a dimeric assembly of two symmetrical discs, each consisting of 7 alpha-type subunits and 7 beta-type subunits. Alpha subunits form the outer rings and the beta subunits form the inner rings. Besides expressing β-subunits ubiquitously, higher vertebrates also possess three interferon inducible β-subunits (LMP7, LMP2, MECL1), which replace normal counterparts X, Y and Z respectively and alter the catalytic activities of the proteasome.[20]

In order to understand the proteasome function and its role in cell biology researchers have employed ‘chemical genetics’ approaches using various proteasome inhibitors. These inhibitors include the natural products and small cell membrane-permeable peptides with aldehyde, vinyl sulfone, boronic acid and glyoxal functional groups. Specific peptide aldehydes of substrate analogues have been found to form reversible covalent adducts with the proteasome and inhibit certain proteolytic activities. However, these inhibitors also hinder

lysosomal and Ca^{2+} activated proteases, thus limiting their use in understanding their precise effects on cells. In spite of great interest in identifying specific novel targets as therapeutics for various diseases, many natural products, with unknown mode of actions, have been identified that have therapeutic potentials such as anti-tumor activity. Epoxomicin, a natural product isolated from an *Actinomycetes* species, is an α′,β′-epoxyketone-tetrapeptide and has potent *in vivo* anti-tumor activity against solid tumors. Crews et al. have found that Epoxomicin targets proteasomal pathways to exert its anti-tumor activity. Epoxomicin primarily inhibits chymotrypsin-like activity 80-fold faster than lactacystin, another proteasome inhibitor. It is a cell membrane permeable, selective and irreversible proteasome inhibitor. Crews et al. demonstrated that Epoxomicin covalently binds to the LMP7, X, Z and MECL1 catalytic β-subunits of the proteasome and selectively inhibits the proteasome. In contrast to peptide aldehyde inhibitors, Epoxomicin does not significantly inhibit nonproteasomal proteases such as trypsin, chymotrypsin, papain, calpain and cathepsin B. It has also been shown to inhibit NF-κB activation and can be used as a potential anti-inflammatory agent.[20, 168].

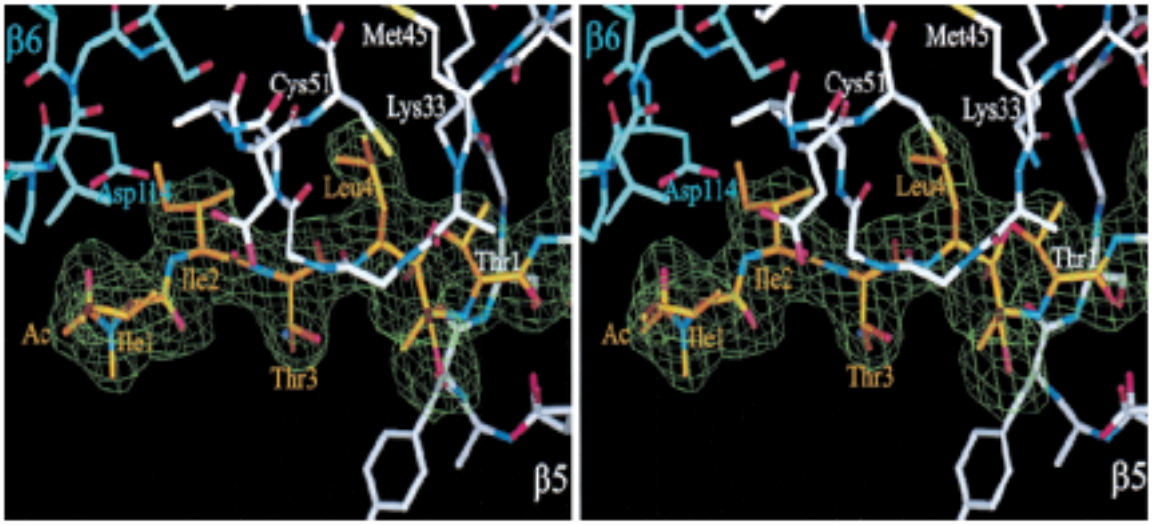
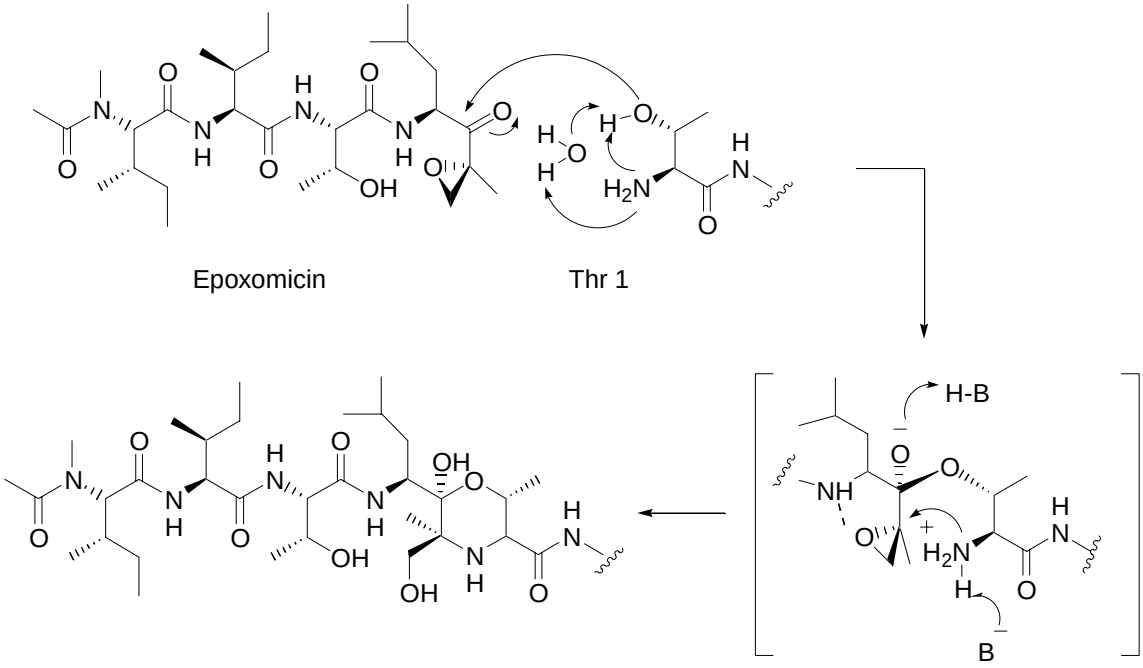


Figure 17. Stereoview of the electron density map of the Epoxomicin adduct at β5

To address the unique specificity of Epoxomicin for the proteasome, a co-crystallized α′,β′-epoxyketone-containing natural product with the yeast S cerevisrae 20S proteasome (Figure 17) has been reported.[169] A comparison between that structure and a crystal stucture of 20S proteasome complexed with acetyl-Leu-norleucinal[170] showed that both the Epoxomicin and the peptide aldehyde bind similarly to the catalytic subunits, completing

an antiparallel β-sheet.[170] However, a striking difference is the covalent adduct formed by each inhibitor with the amino terminal threonine (Thr 1). Whereas the peptide aldehyde is attacked by the threonyl Oγ to form a hemiacetal, a well-defined electron density map of the β5/Pre2 subunit complexed with Epoxomicin reveals the presence of a unique 6-atom morpholino ring.

This morpholino derivative results from adduct formation between the α′,β′-epoxyketone of Epoxomicin and the amino terminal threonyl Oγ and N of the β5 subunit (Scheme 56). Formation of an irriversible morpholino adduct upon Epoxomicin addition is consistent with the observed kinetic profile of the Epoxomicin and 20S proteasome interaction.[20, 168]



Scheme 56: The proposed mechanism of morpholino derivative adduct formation

The mechanism of the morpholino derivative formation is most likely a two-step process. First, activation of the threonyl Oγ by its N-terminal amino group directly[171] or via a neighboring water molecule, which is positioned between the nucleophilic oxygen of the side chain and the α-amino nitrogen, acting as a base (B). The second step is the formation of a hemiacetal by intramolecular cyclization, where Thr1 N opens the epoxide ring with consequent inversion of the C2 carbon. Activation of the epoxide may be facilitated by the hydrogen bond formation between the N5 hydrogen and the oxygen of the epoxide.

Since other proteases, which are common targets for many proteasome inhibitors, do not have an amino terminal nucleophilic residue as part of their active sites, Epoxomicin cannot form the same morpholino adduct with those proteases as it does with 20S proteasome, thus explaining why Epoxomicin does not inhibit those proteases.[169] The unique specificity of Epoxomicin towards proteasome inhibition allowed the Crews group to evaluate its therapeutic potential against various diseases involving proteasome function.

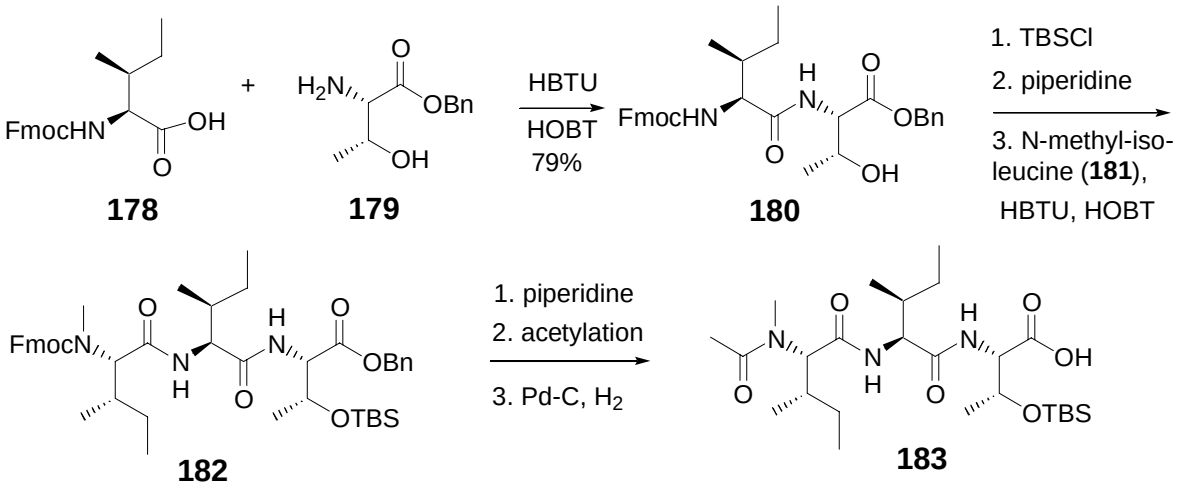
2. Previous total syntheses of Epoxomicin

The total synthesis of Epoxomicin (**7**) has been reported by Crews et al. in 1999[20] and Williams et al. in 2004.[172] Here in this thesis we describe those two syntheses briefly.

2.1. Total synthesis of Epoxomicin by Crews *et al.*

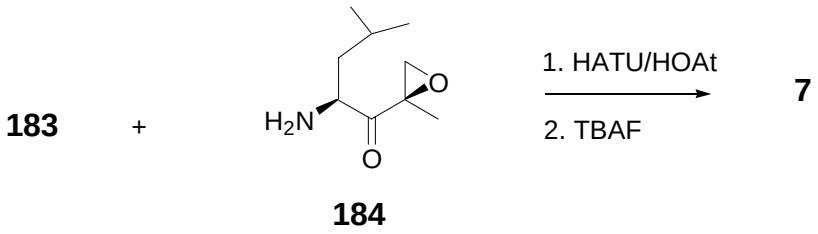
In the total synthesis by Crews and his group α,β-epoxyketone Epoxomicin (**7**) and biotinylated Epoxomicin[20] were synthesized, but here we describe only the total synthesis of Epoxomicin (**7**). They made first the tripeptide molecule which was coupled with the epoxide moiety, prepared from an unsaturated ketone.

For the synthesis ot the tripeptide fragment (Scheme 57), Fmoc-N-isoleucine **178** was coupled with protected hydroxy ester **179** in presence of HBTU and HOBT to give dipeptide **180.** The hydroxyl group of the dipeptide was protected with TBSCl and removal of the Fmoc group was done with piperidine. This was followed by coupling of the amine with Fmoc-N-methyl-isoleucine (**181**) with HBTU/HOBT and led to the fully protected fragment **182**. Removal of the Fmoc group and subsequent acetylation followed by catalytic hydrogenolysis gave the target tripeptide fragment **183**.



Scheme 57: Crews' synthetic pathway to tripeptide fragment **183**

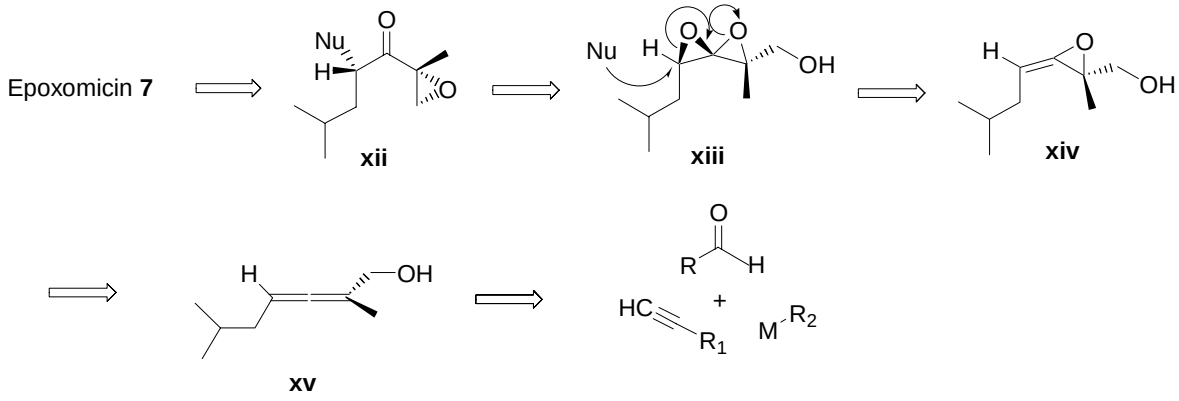
After synthesis of tripeptide fragment **183**, it was coupled with α,β-epoxy leucine **184**, in presence of HATU/HOAt (Scheme 58).[173] Finally, the TBS protecting group was removed with TBAF to provide the desired Epoxomicin (**7**).



Scheme 58: Crews' final steps of the synthesis of Epoxomicin

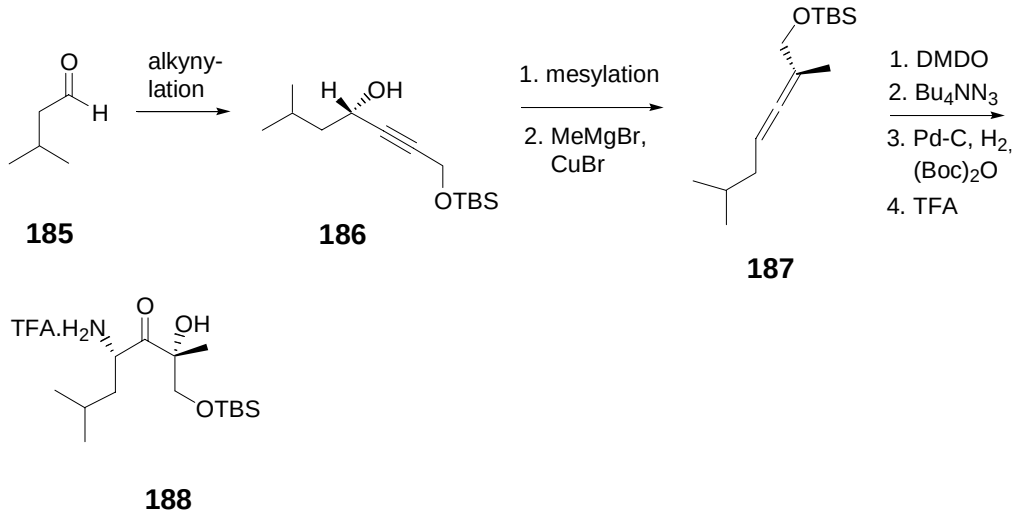
2.2 Total synthesis of Epoxomicin by Williams *et al.*

Williams and his group developed an efficient route to Epoxomicin (**7**) and established the first use of the spirodiepoxide functional group in total synthesis.[172] In this synthesis they described, that the spirodiepoxide of type **xiii** could serve as a precursor of simplified structure **xii**, present in **7** (Scheme 59). In the presence of a suitable nucleophile, species **xiii** should undergo a regioselective S_N2 reaction. Enantiomerically pure **xiv** would be derived from oxidation of optically pure, appropriately protected allene **xv**, which could arise from aldehyde, alkyne and organometallic precursors.



Scheme 59: Williams' retrosynthetic pathway to Epoxomicin (**7**)

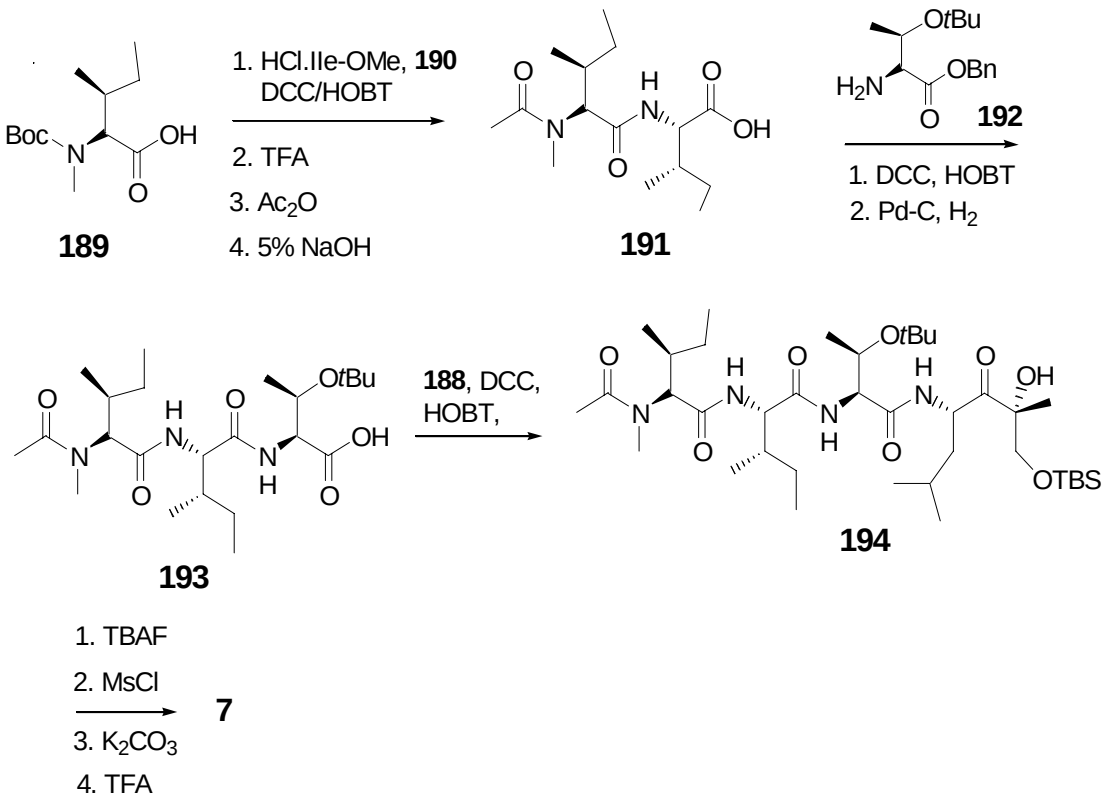
The optimized synthesis of Epoxomicin is represented in Scheme 60. Isovaleraldehyde **185** was subjected to asymmetric alkynylation[174] to form **186** (>95% ee). The alcohol was converted to mesylate and subsequently transformed into allene **187**. Treatment of **187** with DMDO[175] and exposure to sodium azide produced an azide, which in situ after reduction, protection and treatment with TFA led to stable amine salt **188**.



Scheme 60: Synthesis of amino salt **188**

Now DCC promoted coupling between acid **189** and HCl.II-OMe **190** led to a dipeptide which on Boc removal, acetylation and saponification gave acid **191** (Scheme 61). Then this compound was coupled with threonine **192** and on hydrogenolysis resulted in **193**, which was again coupled with amine salt **188** and furnished hydroxy ketone **194**. Exposure of **194** to fluoride cleaved the TBS group and the resulting primary alcohol was converted

to mesylate and then finally after cyclization and deprotection of the *tert*-butyl group produced Epoxomicin (**7**).



Scheme 61: Williams' final steps of Epoxomicin synthesis

3. Goal of research

Structural studies with the co-crystallization of the 20S proteasome-Epoxomicin complex revealed that the unique specifity of the natural product toward proteasome is mainly due to the α,β-epoxyketone pharmacophore, which forms an unusual six-membered morpholino ring with the amino terminal catalytic threonine (Thr-1) of the 20S proteasome. [169] The observed selectivity of Epoxomicin for the proteasome is rationalized by the requirements for both N-terminal amino group and side chain nucleophile for adduct formation.

In most eukaryotic organisms Ntn-Hydrolases, which maintain the control over protein quality are represented by the three active sub-units of the proteasome (β1, β2, and β5). In addition to the proteasome, plasmodium falciparum, which causes malaria tropica, contains

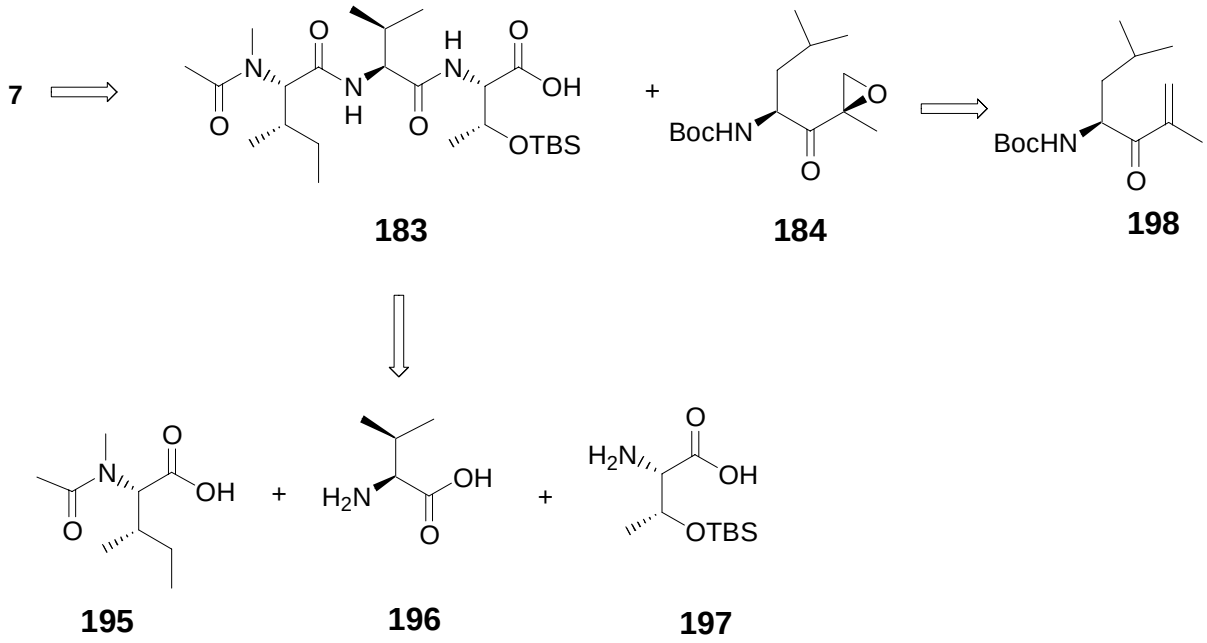
another Ntn-Hydrolase (PFL1465c) similar to prokaryotic proteasome progenitors. Therefore four different activities can be targeted by selective inhibitors. A naturally occurring epoxide containing peptide (Epoxomicin) and synthetic derivatives block P. falciparum growth in the low nanomolar range. Changes of this peptide moiety of the drug can be used to target the inhibitor to individual proteasome subunits. B. Mordmüller et al. found that this property of epoxide-containing peptides can be used to target selectively plasmodial Ntn-Hydrolases[176], facilitating a new approach in the treatment of malaria tropica.
In addition proteasome inhibition is currently being evaluated for a variety of therapeutic purposes enabled by e.g. antitumor activity.[177]

However, apart from the aforementioned studies, relatively little has been reported regarding the synthesis of this α,β-epoxyketone inhibitor. Current efforts are focused on the synthesis of additional peptide epoxyketone proteasome inhibitors that display specifity for each of the three proteolytic activities of the proteasome.[20,178] For this reason the synthesis method will enable future in vivo studies using this reagent.

4. Results and discussion

4.1 Retrosynthetic analysis of Epoxomicin (7)

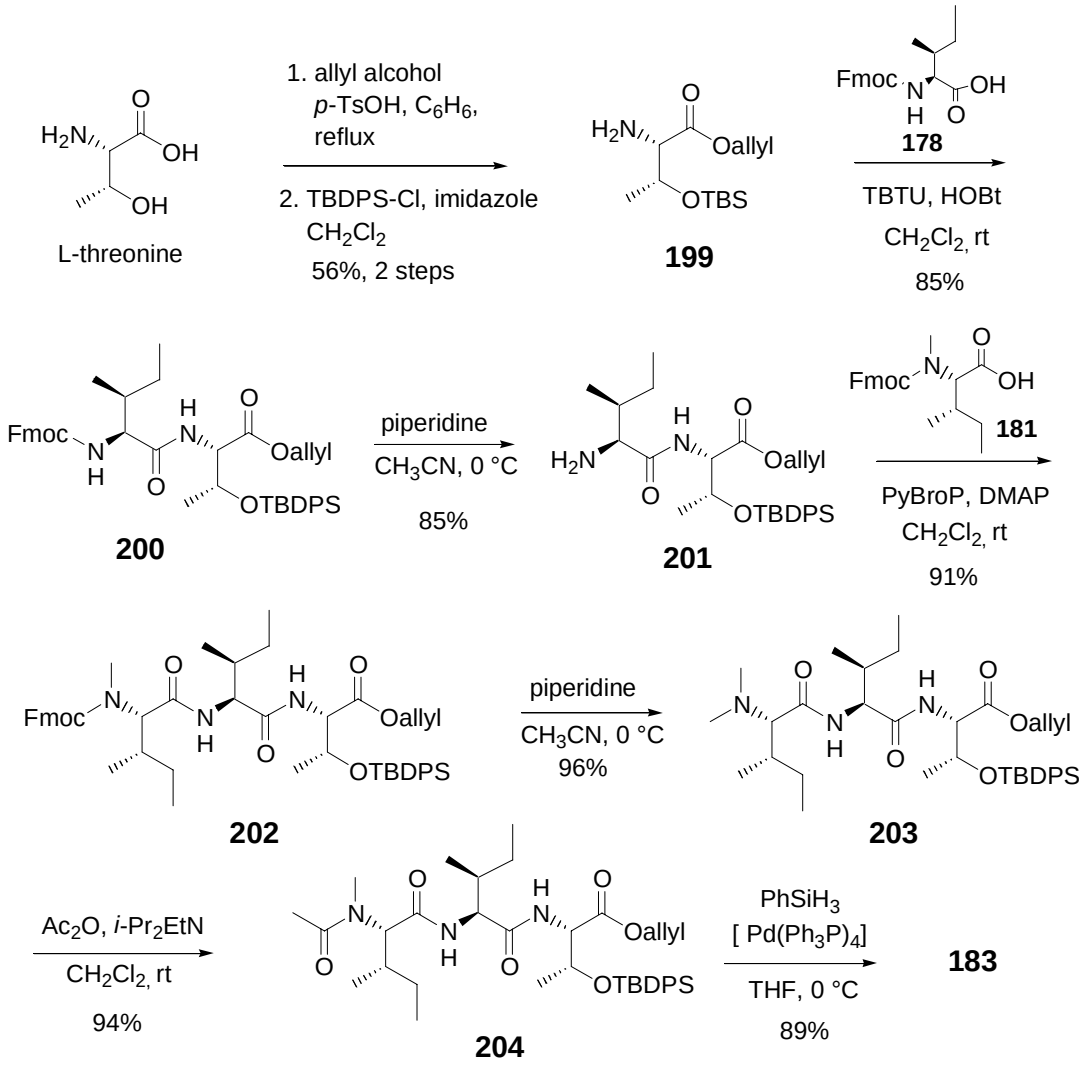
Epoxomicin is a tetrapeptide molecule with the first amino acid ending with an acetyl protecting group and the last peptide containing an epoxide. In the retrosynthetic analysis the Epoxomicin molecule would be obtained from the amino group protected tripeptide, left hand fragment **183**, which would be coupled with the α,β-epoxy leucine, right hand fragment **184** to yield the complete backbone of the target compound **7** (Scheme 62). The left hand fragment **183** is the combination of protected threonine **195**, isoleucine **196** and hydroxy group protected N-methyl isoleucine **197**. The epoxide containing right-hand fragment **184** could be prepared from the precursor α,β-unsaturated ketone **198**.



Scheme 62: Retrosynthetic pathway to Epoxomicin (**7**).

4.2 Synthesis of the left hand tripeptide fragment 183

The synthesis of left hand tripeptide fragment **183** started from commercially available L-threonine (Scheme 63). The carboxyl group was first protected by formation of ester with allyl alcohol in presence of *p*-toluenesulfonic acid (*p*-TsOH) followed by protection of the hydroxyl group with TBDPS-Cl giving ester **199**. This amine was coupled with Fmoc protected isoleucine **178** and produced dipeptide **200**. Here we used the coupling reagent *O*-benzotriazo-1-yl-*N,N,N,N*-tetramethyl-uronium hexafluoroborate (TBTU) and 1-hydroxybenzotroazole (HOBt). After removal of the Fmoc protecting group with piperidine, the free amine **221** was again coupled with the commercially available Fmoc-*N*-methyl-isoleucine **181** using the coupling reagent Bromo-tris-pyrrolidino-phosphonium hexafluorophosphate (PyBroP) with dimethylaminopyridine (DMAP) to get tripeptide **202**. The Fmoc protecting group of **202** was then removed again with piperidine and led to **203**. Subsequent acetylation with acetyl chloride and diisopropylethyl amine resulted in tripeptide **204.** The allyl ester of compound **204** was then cleaved using phenyl silane ($PhSiH_3$) and tetrakistriphenylphosphine palladium [$Pd(PPh_3)_4$] to get the desired compound, acid **183**.

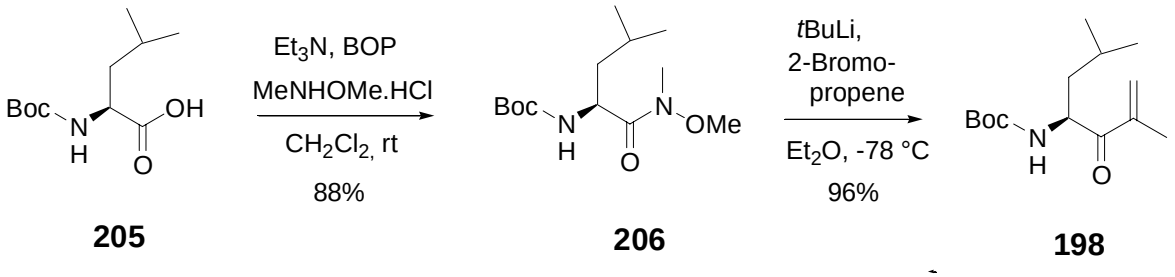


Scheme 63: Synthesis of the tripeptide fragment **183**

4.3 Synthesis of the right hand fragment 184

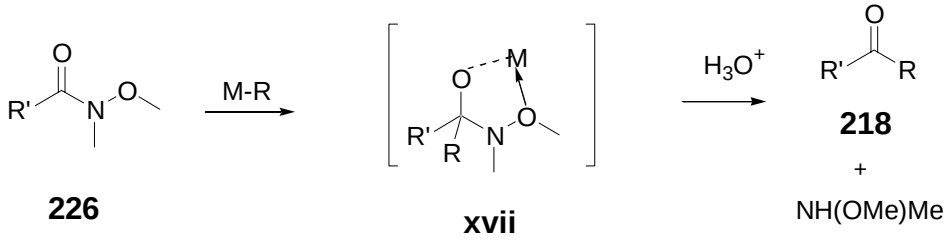
4.3.1 Synthesis of α,β-unsaturated ketone **198**

For the synthesis of the right-hand fragment **184**, first we synthesized α,β-unsaturated ketone **198** (Scheme 64). This synthesis was initiated from Boc-N-leucine **205**, which was converted to the Boc-leucine Weinereb amide **206** by reaction with benzotriazol-1-yloxy-tris[dimethylamino]phosphonium hexafluorophosphate (BOP) and *O,N*-dimethylhydroxyl-amine hydrochloride (OMe-N-MeH_2Cl). The addition of propen-2-yl lithium to **206** was done by using 2-bromopropene and *t*BuLi resulting in the formation of α,β-unsaturated ketone **198** in almost quantitative yield.



Scheme 64: Synthesis of enone **198**

In this transformation the Weinereb amide **206** was attacked by an organolithium reagent (propen-2-yl lithum) and led to the formation of a tetrahedral intermediate (**xvii**) which is stabilized by *chelation* of the lithium atom by two oxygen atoms (Scheme 65). This intermediate is stable under the reaction conditions thus preventing double addition. Upon aqueous work up, the desired ketone **198** is obtained.



Scheme 65: Mechanism of the Weinreb amide reduction

The α,β-unsaturated ketone **198** was confirmed from the ^{13}C-spectrum data in Figure 18. It clearly showed the chemical shift value of α,β-unsaturated carbonyl carbon (201.5 ppm), as well as the olefinic carbon atoms.

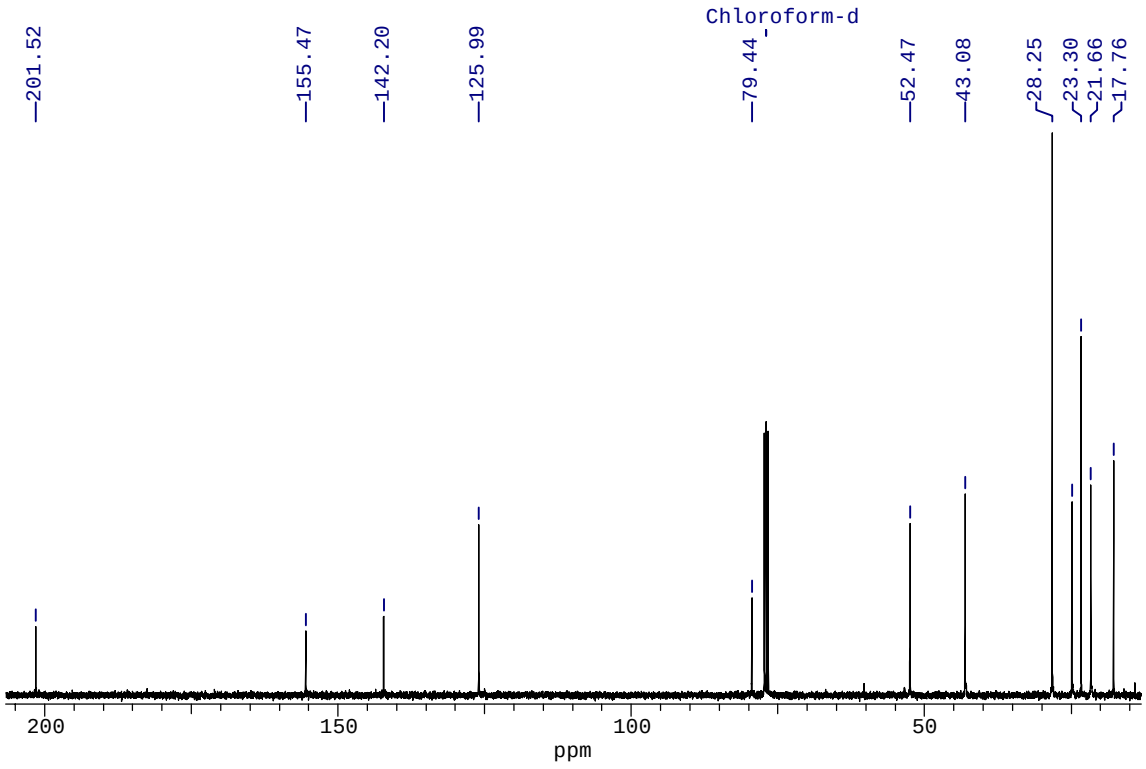
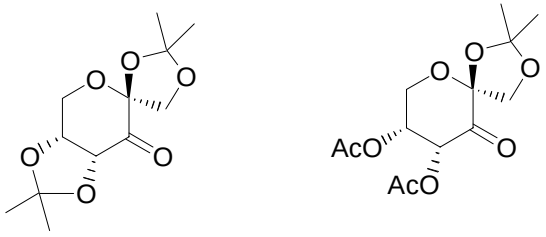


Figure 18: ^{13}C-NMR spectrum of α,β-unsaturated ketone **198**

4.3.2 Synthesis of the epoxide using chiral dioxiranes

Dioxiranes generated in situ from chiral ketones have been shown to be highly enantioselective for the asymmetric epoxidation of a variety of olefins.[179] It was reported that fructose-derived ketone **207** and its acetate analogue **208** (Figure 19) are effective epoxidation catalysts and give high ee's for a variety of trans- and trisubstituted olefins.[180] Therefore, in our synthetic route we first synthesized the acetate **208** from **207** and used both of them for epoxidation of α,β-unsaturated ketone **198**.



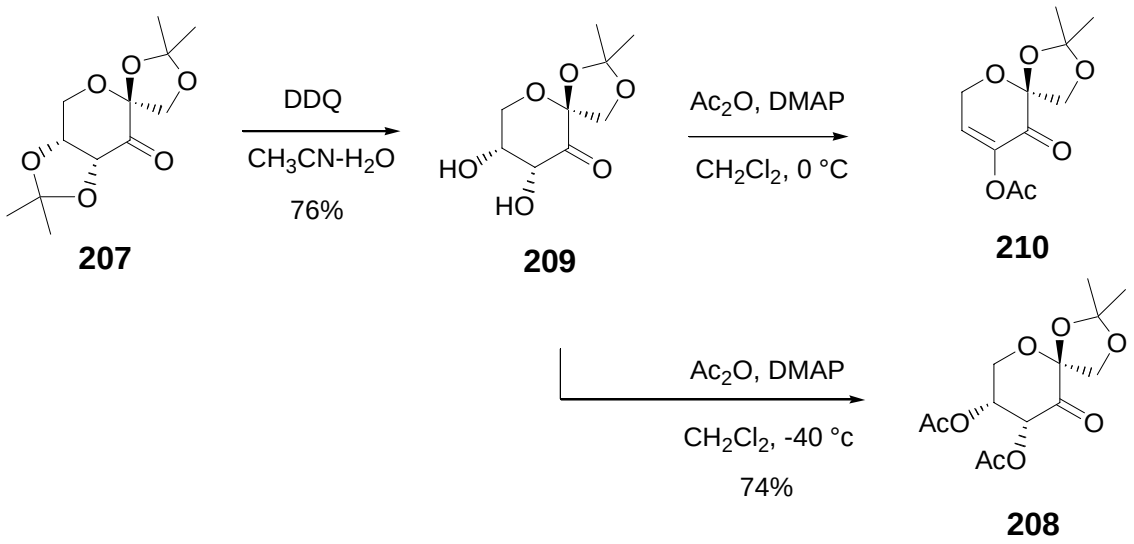
207 **208**

Figure 19: Structures of chiral ketone

4.3.2.1 Preparation of ketone **208**

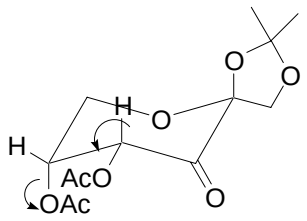
The fructose derivative ketone **207** was purchased from Aldrich and reacted with di-chlorodicyano benzoquinone (DDQ), which deprotected selectively only adjacent hydroxyl

groups, to produce diol **209** (Scheme 66). The acetylation of both hydroxyl groups was done by using acetic anhydride (Ac_2O) and DMAP. In this step, it was interesting to note that we got predominantly eliminated product **210**, when the reaction was carried out at 0 °C but the desired ketone **208**, when the reaction was done at –40 °C. We assumed that in situ the product **208** was converted to eliminated product **210** through its chair like conformation **xviii**, because of the higher temperature (Figure 20).



207 **209** **210** **208**

Scheme 66: Preparation of chiral ketone **208**



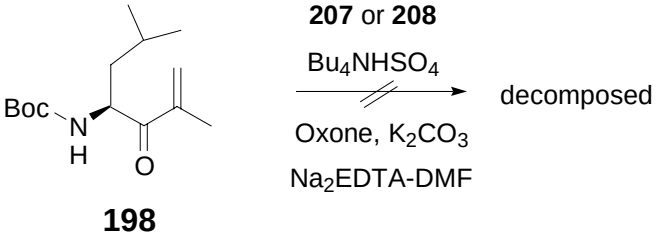
xviii

Figure 20: Probable structure of **xviii**

4.3.2.2 Asymmetric epoxidation through ketone **207** and **208**

For the epoxidation reaction unsaturated ketone **198** was treated separately with chiral ketone **207** and **208** in presence of tetrabutylammonium hydrogen sulfate (Bu_4NHSO_4) and

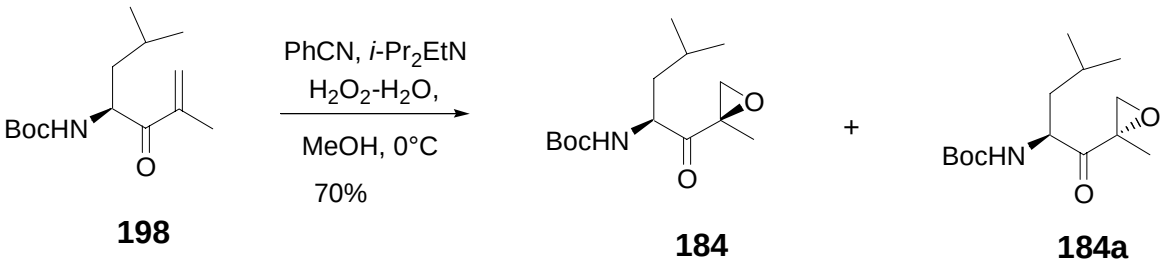
oxone in buffer solution (Scheme 67).[180] But unfortunately in both cases the reaction products were decomposed.



Scheme 67: Asymmetric epoxidation of **198**

4.3.3 Synthesis of the epoxide through alkaline peroxide

After unsatisfactory results of selective epoxide formation by chiral ketones, we proceeded our synthesis by epoxide formation with alkaline hydrogen peroxide (H_2O_2) (Scheme 68). The α,β-unsaturated ketone **198** was treated with hydrogen peroxide in presence of diisopropylethylamine (*i*-Pr_2EtN) and benzonitrile (PhCN) to furnish a mixture of the epoxides **184** and **184a** in a ratio of 3:1. The major epoxide **184**, which produced the final product identical to Epoxomicin (**7**) was separated by column chromatography very easily and used in the next steps.

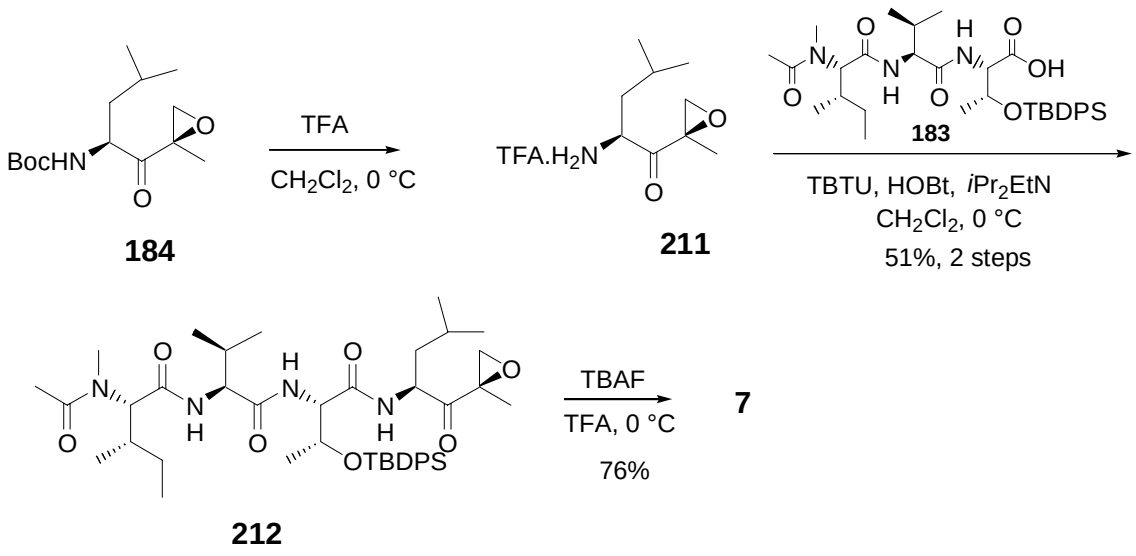


Scheme 68: Synthesis of epoxide **184** with hydrogen peroxide

4.4. Coupling of fragments and final steps of synthesis of Epoxomicin (7)

Before coupling of two fragments, the Boc protecting group of the right-hand fragment **184** was removed with trifluoroacetic acid (TFA) to get the salt of amine **211** (Scheme 69).

After removing the solvent and excess TFA, the salt **211** was directly coupled with left-hand fragment **183** using *O*-benzotriazo-1-yl-*N,N,N,N*-tetramethyluronium hexafluoroborate (TBTU) and 1-hydroxybenzotroazole (HOBt) in presence of diisopropylethylamine (*i*-Pr_2EtN). In the resulting compound **212,** the TBDPS protecting group was lastly removed with tetrabutylammonium fluoride (TBAF) to get our target molecule Epoxomicin (**7)** in very good overall yield.



Scheme 69: Final steps of synthesis of Epoxomicin (**7**)

The optical rotation value of compound **7** $[\alpha]_D^{24} = -64.4$ (c = 0.36, MeOH), lit[172] −66.1(c = 0.50, MeOH), confirmed that the stereochemistry of epoxide is identical with the natural Epoxomicin.

IV. CONCLUSION

This dissertation includes two parts, the first one is a convergent total synthesis of the natural product Cryptophycin-3 (**5**) and its analogue and the second part contains the synthesis of a proteasome inhibitor, α,β-epoxy ketone Epoxomicin (**7**).

In the first part, the present synthetic route consists of the synthesis of the four building blocks (fragment A, B, C and D), the assembly of these building blocks and the final cyclization. The most interesting building block A **12b** contains two stereocenters at C5 and C6 with *anti*-stereorelationship, which were created by an Evans *syn* aldol reaction. The necessary inversion of the methyl–bearing stereocenter was done by hydroboration of a terminal 2,2-disubstituted double bond (Scheme 52). The aryl part and the remaining carbons of the molecule were introduced by Wittig-Horner olefination and a Wittig reaction, respectively. The building block B was prepared by a very efficient enantioselective alkylation between the glycine derivative **148** and benzyl bromide **149** in presence of chiral catalyst **147** with high stereoselectivity. The other two building blocks C and D were prepared by very short and efficient synthetic pathways, which make the whole synthesis short. The assembly of three fragments (D, C, B) was very advantageous because of the Fmoc amino protecting group, which was deprotected very easily by diethylamine. The final coupling between tripeptide **126** and building block A was done by a high yield Yamaguchi esterification. The final cyclization was performed by using coupling reagent TBTU and HOBT which completed the total synthesis nicely and drives efficient enough to produce the whole molecule in large scale. The total synthesis of an analogue **11** was made in a similar fashion but with different tripeptide **174** instead of **127**.

Therefore, from the new synthetic point of view and according to our goal of total synthesis of Cryptophycins, the whole synthesis can be described as follows:

1) The synthesis of fragment A is very efficient, also in large scale where the *anti*-stereo-relationship at C5 and C6 is based on aldol methodology, followed by *anti*-selective hydroboration and flexible introduction of aryl group was done by Wittig olefination.

2) The synthesis of α-amino acid B by an enantioselective alkylation is a totally new approach, especially for the use of the Fmoc protecting group.

3) The use of the Fmoc protecting group should in principle allow a solid-phase assembly of a suitable seco compound which also means a new approach for the synthesis of Cryptophycins and natural product like libraries.

The second goal of the present thesis was the synthesis of the very promising epoxide reactive molecule Epoximicin (**7**). We made the whole molecule in such a way that the tripeptide left-hand fragment **183** was coupled with the epoxide containing right-hand fragment **211**. By using a suitable coupling reagent (TBTU/HOBT) destruction of the epoxide could be prevented (Scheme 69). All the protecting groups were removed very easily in good yield. Efforts were made to form the epoxide stereoselectively using chiral dioxiranes. However, under these conditions the epoxide did not form. Finally, we synthesized the epoxide in a 3:1 ratio by using alkaline peroxide, which are separated easily by flash chromatography. The whole synthesis was done in only 13 steps in good overall yield. Our synthesis of Epoxomicin is improved concerning efficiency and described in more detail than the previous reports for Epoxomicin or related compounds. This way we established a facile method for the synthesis of multi-gram quantities. Key features of the synthesis of the tripeptide part include the use of an allyl protecting group for the right-handed carboxyl function, combined with Fmoc and *tert*-butylsilyl protectin groups for the amino and alcohol functions, respectively.

V. EXPERIMENTAL PART

1. General Remarks

1.1 Chemicals and Working Techniques

The chemicals were purchased from the firms Acros, Aldrich, Fluka, Lancaster, Avocado and Merck. All reagents were obtained from commercial suppliers and used without further purification unless otherwise stated. All solvents were distilled and/or dried prior to use by standard methodology except for those, which were reagent grades. The applied petroleum ether fraction had a b.p. of 40–60 ^{o}C. Anhydrous solvents were obtained as follows: THF, diethyl ether and toluene by distillation from sodium and benzophenone, dichloromethane and chloroform by distillation from calcium hydride, acetone by distillation from phosphorous pentoxide. Absolute triethylamine and pyridine and diisopropylethylamine were distilled over calcium hydride prior to use. Unless otherwise mentioned, all reactions were carried out under a nitrogen atmosphere and the reaction flasks were pre-dried by heat gun under high vacuum. All air or water sensitive chemicals were stored under inert atmosphere. Compounds that are not described in the experimental part were synthesized according to the literature.

1.2 NMR-spectroscopy

All the spectra were measured on a Bruker Avance 400 spectrometer, which operated at 400 MHz for ^{1}H and 100 MHz for ^{13}C nuclei, respectively. ^{1}H- and ^{13}C-NMR spectra were performed in deuterated solvent and chemical shifts were assigned by comparison with the residual proton and carbon resonance of the solvent and tetramethylsilane (TMS) as an internal reference ($\delta = 0$). Data are reported as follows: chemical shift (multiplicity: s = singlet, d = doublet, t = triplet, ddd = doublet of doublet of doublet, dt = doublet of triplet, td = triplet of doublet, m = multiplet, br = broadened, *J* = coupling constant (Hz), integration, peak assignment in italic form).

1.3 Mass Spectrometry

Mass spectra were recorded on Finnigan Triple-Stage-Quadrupol Spectrometer (TSQ-70)

from Finnigan-Mat. High-resolution mass spectra were measured on a modified AMD Intectra MAT 711 A from the same company. The used mass spectrometric ionization methods were electron-impact (EI), fast-atom bombardment (FAB) or field desorption (FD). FT-ICR-mass spectrometry and HR-FT-ICR mass spectra were measured on an APEX 2 spectrometer from Bruker Daltonic with electrospray ionization method (ESI). Some of the mass spectra were also measured on an Agilent 1100 series LC-MSD. Analytical HPLC-MS: HP 1100 Series connected with an ESI MS detector Agilent G1946C, positive mode with fragmentor voltage of 40 eV, column: Nucleosil 100–5, C-18 HD, 5 µm, 70 × 3 mm Machery Nagel, eluent: NaCl solution (5 mM)/acetonitrile, gradient: 0/10/15/17/20 min with 20/80/80/99/99% acetonitrile, flow: 0.6 mL min^{-1}. High resolution mass (HRMS) are reported as follows: (ESI): calculated mass for the related compound followed by found mass.

1.4 Infrared Spectroscopy

The FT-IR spectra were recorded on a Fourier Transformed Infrared Spectrometer model FT/IR-430 from the company JASCO. Solid samples were pulverized with potassium bromide and percent reflection (R%) was measured. The percent transmittance (T%) of liquid substances was measured in film between potassium bromide plates. Absorption band frequencies are reported in cm^{-1}.

1.5 Polarimetry

Optical rotations were measured on a JASCO Polarimeter P-1020. They are reported as follows: $[\alpha]^{temperature}D$ (concentration, solvent). The unit of *c* is g/100 mL. Anhydrous CH_2Cl_2 or $CHCl_3$ was used as a solvent. For the measurement the sodium D line = 589 nm was used.

1.6 Melting Points

Melting points were determined with a Büchi Melting point B-540 apparatus and were not corrected.

1.7 Chromatographic Methods

Flash column chromatography was performed using flash silica gel (40–63 µm, 230–400 mesh ASTM) from Macherey-Nagel.

Gas chromatography was performed on a CHROMPACK CP 9000 using a flame ionization detector, and carrier gas H_2. For GC-MS coupled chromatography, a GC-system series 6890 with an injector series 7683 and MS-detector series 5973 from Hewlett Packard was used, with EI method and carrier gas He. Analytical HPLC was performed on a Hewlett Packard HP 1100 system.

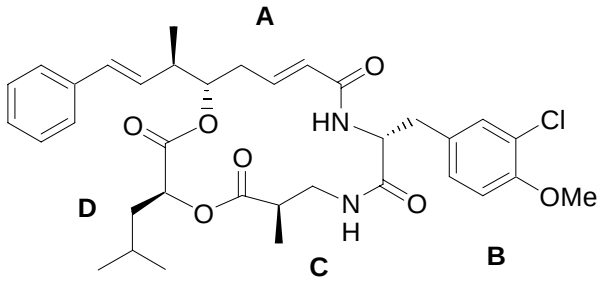
Analytical thin layer chromatography (TLC) was performed on silica gel 60 precoated F_{254} plates (Merck) or Polygram Sil G/UV_{254} (Macherey Nagel). The compounds were visualized by UV_{254} light and the chromatography plates were developed with an aqueous solution of molybdophosphorous acid or an aqueous solution of potassium permanganate by heating with the hot gun. For preparation of the molybdate solution 20 g ammonium molybdate [$(NH_4)_6Mo_7O_{24}.4H_2O$] and 0.4 g $Ce(SO_4)_2.4H_2O$ were dissolved in 400 mL of 10% H_2SO_4. The potassium permanganate solution was prepared from 2.5 g $KMnO_4$ and 12.5 g Na_2CO_3 in 250 mL H_2O.

2. Experimental procedures

The experimental procedures are arranged in ascending order of the number of the compounds.

2.1 Synthesis of Cryptophycin-3

Cryptophycin-3 (5)

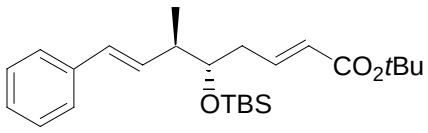


Trifluoroacetic acid (5 mL) was added slowly at 0 °C to a solution of the protected *seco* compound **127** (80 mg, 0.085 mmol) in CH_2Cl_2 (2 mL) and the mixture was stirred for 2 h at room temperature. The solvent was removed in vacuo and toluene (5 mL) was added.

After removal of the solvent, the residue was redissolved in THF (3 mL) and diethylamine (3 mL) was added dropwise at 0 °C. The reaction mixture was stirred again 2 h at room temperature followed by the removal of the solvents in vacuo. The crude *seco* acid **173** was dissolved in dry DMF (15 mL), then TBTU (30 mg, 0.081 mmol), HOBT (2 mg), DIEA (33 µl, 0.19 mmol) were successively added at room temperature. The reaction mixture was stirred for 2 h at room temperature, saturated $NaHCO_3$ solution (5 mL) was added and stirring continued for 1 hour. The mixture was extracted with CH_2Cl_2 (15 mL) and the organic layer dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (50% EtOAc in petroleum ether) to give compound **5** (22 mg, 42% from **127**).

$\boldsymbol{R_f}$ = 0.31 (50% EtOAc in petroleum ether);
$\boldsymbol{[\alpha]_D^{27}}$ = + 24.68 (*c* = 0.40, $CHCl_3$), {ref.[66]; $[\alpha]_D^{22}$ = + 29.5 (*c* = 0.20, $CHCl_3$)};
IR (neat): ν_{max} = 3430, 3310, 2959, 1727, 1667, 1504, 1250, 1173 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): unit A δ = 7.34–7.17 (m, 5H, Ph), 6.65 (ddd, *J* = 15.2, 9.8, 5.5 Hz, 1H, CH_2*CH*CH), 6.37 (d, *J* = 15.9 Hz, 1H, Ph*CH*), 5.97 (dd, *J* = 15.8, 8.7 Hz, 1H, PhCH*CH*), 5.74 (d, *J* = 15.4 Hz, 1H, *CH*CO), 5.08–4.93 (m, 1H, *CH*OCH_2), 2.60–2.45 (m, 2H, *CH*Me/CHCH_2), 2.41–2.28 (m, 1H, CHCH_2), 1.10 (d, *J* = 6.8 Hz, 3H, Me); unit B δ= 7.04 (dd, *J* = 8.3, 1.8 Hz, 1H, C_{ortho}), 6.96–6.91 (m, 1H, C'$_{ortho}$), 6.80 (d, *J* = 8.3 Hz, 1H, C'$_{meta}$), 5.66 (d, *J* = 8.6 Hz, N–H), 4.87–4.72 (m, 1H, NH*CH*), 3.83 (s, 3H, OMe), 3.10 (dd, *J* = 14.4, 5.3 Hz, 1H, CH_2Ph), 3.00 (dd, *J* = 14.4, 7.0 Hz, 1H, CH_2Ph); unit C δ = 7.34–7.17 (m, N–H), 3.51–3.41 (m, 1H, CH_2NH), 3.32–3.22 (m, 1H, CH_2NH), 2.71–2.62 (m, 1H, CO*CH*), 1.19 (d, *J* = 7.0 Hz, 3H, Me); unit D δ = 4.87–4.72 (m, 1H, CO*CH*), 1.75–1.49 (m, 2H, CHCH_2/*CH*Me), 1.38–1.26 (m, 1H CHCH_2), 0.74 (d, *J* = 6.3 Hz, 3H, Me), 0.69 (d, *J* = 6.3 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): unit A δ = 165.4 (CO), 141.4 (CH_2*CH*CH), 136.6 (C_{quart}), 131.8 (Ph*CH*), 129.7 (PhCH*CH*), 128.6 (C_{ortho}), 127.5 (C_{meta}), 126.1 (C_{para}), 125.1 (*CH*CO), 76.9 (*CH*OCH_2), 42.2 (*CH*Me) 36.4 (CHCH_2), 17.3 (Me); unit B δ = 170.2 (CO), 153.9 (C_{para}), 131.0 (C_{ortho}), 130.0 (C'$_{ortho}$), 128.6 (C_{quart}), 122.3 (C'$_{meta}$), 112.1 (C_{meta}), 56.1 (NH*CH*), 53.5 (OMe), 35.0 (CH_2Ph); unit C δ = 175.6 (CO), 41.1 (CH_2NH), 38.2 (CO*CH*), 14.0 (Me); unit D δ = 170.9 (CO), 71.5 (CO*CH*), 39.5 (CHCH_2), 24.4 (*CH*Me), 22.7 (Me), 21.1 (Me) ppm;
HRMS (FT-ICR): calcd. for $C_{35}H_{43}N_2O_7Cl$ $[M + Na]^+$ 661.2651, found 661.2654.

tert-Butyl(2*E*,5*S*,6*R*,7*E*)-5-{[_tert_-Butyl(dimethyl)silyl]oxy}-6-methyl-8-phenylocta-2,7-dienoate (12d)



A solution of dimethyl sulfoxide (0.086 mL, 1.2 mmol) in CH_2Cl_2 (5 mL) was added to a stirred solution of oxalyl chloride (0.050 mL, 0.6 mmol) in CH_2Cl_2 (8 mL) dropwise at –78 °C. A solution of alcohol **139** (0.160 g, 0.50 mmol) in CH_2Cl_2 (5 ml) was added 10 min later. After being stirred for 30 min at –78 °C, the reaction mixture was treated with Et_3N (0.346 mL, 2.50 mmol) and allowed to warm to 0 °C. At this point *tert*-butyl(triphenyl-phosphoranylidene)acetate (0.546 g, 0.150 mmol) in CH_2Cl_2 (6 mL) was added and the mixture stirred at room temperature overnight. The reaction mixture was poured into half-saturated NH_4Cl solution (50 mL) and the layers were separated. The organic layer was dried with $MgSO_4$, filtered and concentrated in vacuo. Purification of the residue by flash chromatography (5% EtOAc in petroleum ether) gave the enoate **12d** (180 mg, 78%) as a colorless oil.

$\boldsymbol{R}_f$ = 0.25 (5% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{24}}$ = +50.1 (*c* = 0.95, CH_2Cl_2);

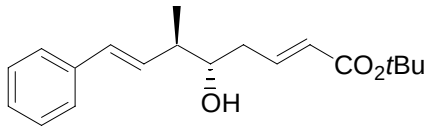
IR (neat): ν_{max} = 3350, 2950, 2856, 1715, 1471, 1366, 1256 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.48–7.11 (m, 5H, Ph), 6.78 (dt, *J* = 15.0, 7.0 Hz, 1H, CH_2CHCH), 6.31 (d, *J* = 15.9 Hz, 1H, Ph*CH*), 6.11 (dd, *J* = 15.9, 8.0 Hz, 1H, PhCH*CH*), 5.69 (d, *J* = 15.6 Hz, 1H, CH*CH*CO), 3.72–3.64 (m, 1H, CH*CH*OCH_2), 2.46–2.34 (m, 1H, *CH*CH_3), 2.31–2.19 (m, 2H, CHCH_2), 1.41 (s, 9H, CMe_3), 1.04 (d, *J* = 6.8 Hz, 3H, Me), 0.85 (s, 9H, $SiCMe_3$), 0.0 (s, 6H, $SiCMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 165.7 (CO), 144.8 (CH_2CHCH), 137.6 (C_{quart}), 132.0 (Ph*CH*), 130.3 (PhCH*CH*), 128.4 (C_{meta}), 127.0 (C_{para}), 126.0 (C_{ortho}), 125.0 (CH*CH*CO), 80.0 (*C*Me_3), 75.1 (CH*CH*OCH_2), 42.8 (*CH*CH_3), 37.2 (CHCH_2), 28.1 (OCMe_3), 25.8 (SiCMe_3), 18.0 (Si*C*Me_3), 16.0 (Me), –4.3 (SiMe), –4.4 (SiMe) ppm;

HRMS (FT-ICR): calcd. for $C_{25}H_{40}O_3Si$ $[M + Na]^+$ 439.2639, found 439.2638.

tert-Butyl(2*E*,5*S*,6*R*,7*E*)-5-Hydroxy-6-methyl-8-phenylocta-2,7-dienoate (12b)



Tetra-*n*-butylammonium fluoride (1 M in THF, 0.87 mL, 0.87 mmol) was added at 0 °C to a solution of enoate **12d** (110 mg, 0.26 mmol) in dry THF (4 mL) and the reaction mixture was stirred at room temperature for 3 h. The mixture was washed with brine, dried with $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromato-graphy (first 30% and then 50% EtOAc in petroleum ether) to give the hydroxy ester **12b** (55 mg, 73%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.29 (30% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{25}}$ = +40.0 (*c* = 1.42, CH_2Cl_2);

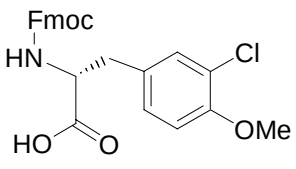
IR (neat): ν_{max} = 3444, 3025, 2976, 2931, 1712, 1652, 1494, 1361 cm^{-1};

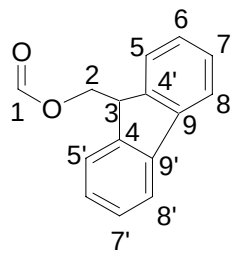
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.70–7.22 (m, 5H, Ph), 6.89–6.81 (m, 1H, CH_2*CH*CH), 6.41 (d, *J* = 15.9 Hz, 1H, Ph*CH*), 6.06 (dd, *J* = 15.9, 8.5 Hz, 1H, PhCH*CH*), 5.77 (d, *J* = 15.6 Hz, 1H, *CH*CO), 3.61–3.56 (m, 1H, *CH*OCH_2), 2.42–2.22 (m, 3H, *CH*Me/CHCH_2), 1.41 (s, 9H, $C(Me)_3$), 1.07 (d, *J* = 6.8 Hz, 6H, $(Me)_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 165.7 (CO), 143.9 (CH_2*CH*CH), 137.0 (C_{quart}), 131.9 (Ph*CH*CH), 130.9 (PhCH*CH*), 128.5 (C_{ortho}), 127.4 (C_{meta}), 126.1 (C_{para}), 125.9 (*CH*CO), 80.2 (*C*Me_3), 73.8 (*CH*OCH_2), 43.2 (*CH*Me), 37.1 (CHCH_2), 28.1 (CMe_3), 16.7 (Me) ppm;

RMS (FT-ICR): calcd. for $C_{19}H_{26}O_3$ $[M + Na]^+$ 325.1774, found 325.1775.

3-Chloro-*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-*O*-methyl-D-tyrosine (13c)





Fmoc

The *tert*-butyl ester **161** (300 mg, 0.59 mmol) was dissolved in CH_2Cl_2 (5 mL) and trifluoroacetic acid (5 mL) was added dropwise at 0 °C. After stirring at room temperature for 5 h, the reaction mixture was concentrated in vacuo. To the residue was added toluene (5 mL) and this mixture was concentrated again. The residue was purified by flash chromatography (5% MeOH in CH_2Cl_2) to yield the Fmoc-protected acid **13c** (245 mg, 93%) as a white solid product.

$\boldsymbol{R_f}$ = 0.25 (5% MeOH in CH_2Cl_2);

$\boldsymbol{[\alpha]_D^{25}}$ = –20.71 (*c* = 0.38, CH_2Cl_2);

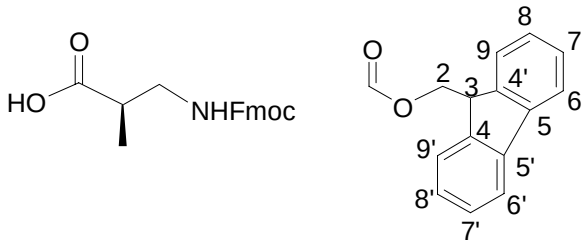
IR (neat): ν_{max} = 3410, 3316, 3065, 2953, 2930, 1722, 1716, 1606, 1504, 1450, 1280, 1259, 1065 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76–7.18 (m, 9H, Fmoc, C_{ortho}), 6.98 (d, *J* = 8.0 Hz, 1H, C'_{ortho}), 6.80 (d, *J* = 8.0 Hz, 1H, C'_{meta}), 5.27 (d, *J* = 7.0 Hz, NH), 4.67–4.56 (m, 1H, NH*CH*), 4.47–4.29 (m, 2H, Fmoc), 4.24–4.11 (m, 1H, Fmoc), 3.84 (s, 3H, OMe), 3.16–2.96 (m, 2H, CH_2Ph) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 175.0 (CO), 155.7 (CO, Fmoc), 154.1 (C_{para}), 143.5 (4/4', Fmoc), 141.2 (5/5', Fmoc), 131.0 (C_{ortho}), 128.6 (C'_{ortho}), 127.7 (C_{quart}), 127.0 (7/7', Fmoc), 125.3 (8/8', Fmoc), 125.0 (9/9', Fmoc), 122.3 (C_{meta}), 119.8 (6/6',Fmoc), 112.1 (C'_{meta}), 67.0 (2, Fmoc), 56.0 (NH*CH*), 54.5 (OMe), 47.0 (3, Fmoc), 36.6 (CH_2Ph) ppm;

HRMS (FT-ICR): calcd. for $C_{25}H_{22}ClNO_5$ $[M + Na]^+$ 474.1079, found 474.1077.

(2*R*)-3-{[(9*H*-Fluoren-9-ylmethoxy)carbonyl]amino}-2-methylpropanoic acid (14c)



A solution of the ester **166** (500 mg, 1.4 mmol) in acetic acid (50 mL) was treated with concentrated HCl (5 mL) followed by heating of the mixture at 100 °C for 15 h. After cooling it was poured into water (500 mL) and the colorless precipitate collected by filtration. The product was purified by flash chromatography (5% MeOH in CH_2Cl_2) to provide 320 mg (67%) of the acid **14c** as a colorless solid substance.

$\boldsymbol{R_f}$ = 0.38 (5% MeOH in CH_2Cl_2 + 2 drops of AcOH);

$\boldsymbol{[\alpha]_D^{25}}$ = –10.85 (*c* = 0.27, CH_2Cl_2);

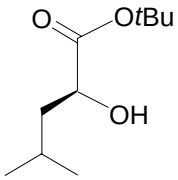
IR (neat): ν_{max} = 3344, 2360, 1715, 1520, 1450, 1247 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.71–7.19 (m, 8H, Fmoc), 5.16 (s, br, OH), 4.46–4.14 (m, 3H, Fmoc), 3.39–3.23 (m, 2H, *CH*$_2$NH), 2.72–2.69 (m, 1H, CO*CH*), 1.16 (d, *J* = 7.1 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 177.4 (CO), 156.7 (CO, Fmoc), 146.2 (4/4', Fmoc), 143.8 (5/5', Fmoc), 127.6 (7/7', Fmoc), 127.0 (8/8', Fmoc), 125.0 (9/9', Fmoc), 119.9 (6/6', Fmoc), 66.7 (2, Fmoc), 47.9 (3, Fmoc), 43.5 (CH*CH*$_2$NH), 39.7 (CO*CH*CH_2), 15.0 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{19}H_{19}NO_4$ $[M + Na]^+$ 348.1206, found 348.1204.

tert-Butyl(2*S*)-2-Hydroxy-4-methylpentanoate (15c)



To the solution of Na_2CO_3 (2.76 g, 26.1 mmol) in methanol (12 mL) and water (20 mL) acetate **170** (2 g, 8.7 mmol) was added. The resulting solution was stirred vigorously at room temperature for 12 h. Methanol was removed under vacuum, and the resulting aqueous solution was extracted with CH_2Cl_2 twice. The organic layer was then dried over anhydrous Na_2SO_4, filtered and concentrated under vacuum. The residue was purified by flash chromatography (15% EtOAc in petroleum ether) to produce **15c** (1.30 g, 79%) as a pale yellow oil.

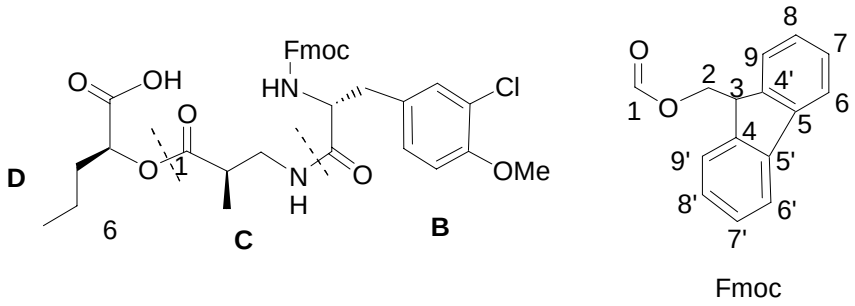
$\boldsymbol{R_f}$ = 0.27 (15% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{25}}$ = –8.16 (*c* = 1.47, CH_2Cl_2);

^{1}H NMR (400 MHz, $CDCl_3$): δ = 3.96 (q, *J* = 6.4 Hz, 1H, *CH*OH), 2.93 (s, 1H, OH), 1.87–1.75 (m, 1H, *CH*Me), 1.45–1.35 (m, 11H, CH*CH*$_2$/C(*Me*)$_3$), 0.85 (d, *J* = 6.8 Hz, 6H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 175.0 (CO), 81.8 (*C*Me$_3$), 69.1 (CHOH), 43.4 (CH*CH*$_2$), 27.8 (C*Me*$_3$), 24.3 (*CH*Me), 23.1 (Me), 21.4 (Me) ppm;

(2*S*)-2-{[(2*R*)-3-({3-Chloro-*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-*O*-methyl-D-tyrosyl}amino)-2-methylpropanoyl]oxy}-4-methylpentanoic acid (126)



The *tert*-butylester **172** (100 mg, 0.14 mmol) was dissolved in CH_2Cl_2 (5 mL) followed by the addition of trifluoroacetic acid (5 mL) at 0 °C. The reaction mixture was stirred for 3 h at room temperature. Then toluene (5 mL) was added and the mixture concentrated in vacuo. This procedure was repeated twice to give the crude acid which was purified by flash chromatography (5% MeOH in CH_2Cl_2 + 2 drops of AcOH) resulting in acid **126** (82 mg, 79%) as a white solid product.

$\boldsymbol{R_f}$ = 0.21 (5% MeOH in CH_2Cl_2 + 2 drops of AcOH);

$\boldsymbol{[\alpha]_D^{24}}$ = –34.87 (*c* = 0.44, CH_2Cl_2);

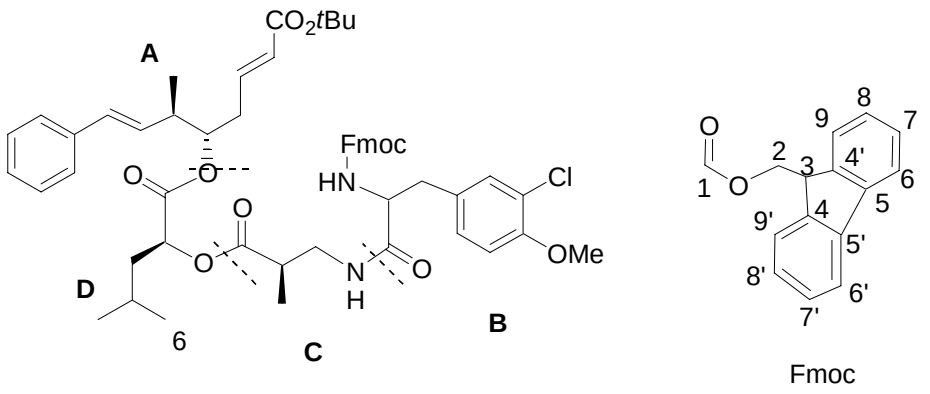
IR (neat): ν_{max} = 3416, 3310, 2850, 2051, 1700, 1630, 1502, 1180 cm^{-1};

^{1}H NMR (400 MHz, CD_3OD): unit B δ = 7.67–6.58 (m, 11H, Ph), 4.24–4.00 (m, 4H, NH*CH*/Fmoc), 3.58 (s, 3H, OMe), 2.72–2.43 (m, 2H, *CH*$_2$Ph), unit C δ = 3.45–3.30 (m, 2H, *CH*$_2$NH), 3.08–2.89 (m, H, CO*CH*), unit D δ = 4.86 (dd, *J* = 9.8, 4.5, Hz, 1H, CO*CH*), 1.73–1.52 (m, 2H, CH*CH*$_2$) 1.25–1.18 (m, 1H, *CH*Me), 1.04 (d, *J* = 6.8 Hz, 3H, Me), 0.82 (d, *J* = 6.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, CD_3OD): unit B δ = 169.6 (CO), 156.4 (CO, Fmoc), 153.7 (C_{para}), 142.5 (4, Fmoc), 140.7 (5, Fmoc), 131.3 (C_{ortho}), 128.7 (C'_{ortho}), 128.1 (C_{quart}), 127.9 (7/7, Fmoc), 127.5 (8/8', Fmoc), 126.8 (9/9', Fmoc), 126.2 (C'_{meta}), 120.0 (6/6', Fmoc), 114.8 (C_{meta}), 63.1 (2, Fmoc), 57.4 (NH*CH*), 55.5 (OMe), 44.7 (3, Fmoc), 36.1 (*CH*$_2$Ph), unit C δ = 172.2 (CO), 43.8 (*CH*$_2$NH), 41.1 (CO*CH*), 21.9 (Me), unit D δ =174.0 (CO), 74.5 (CO*CH*), 38.2 (CH*CH*$_2$), 23.7 (*CH*Me), 16.8 (Me), 15.0 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{35}H_{39}ClN_2O_8$ $[M + Na]^+$ 674.1350, found 674.1343.

tert-Butyl(2_E_,5_S_,6_R_,7_E_)-5-[((2_S_)-2-{[(2_R_)-3-({3-chloro-_N_-[(9_H_-fluoren-9-ylmethoxy)carbonyl]-_O_-methyl-D-tyrosyl}amino)-2-methylpropanoyl]oxy}-4-methylpentanoyl)oxy]-6-methyl-8-phenylocta-2,7-dienoate (127)



To a solution of the tripeptide derivative amino acid **126** (90 mg, 0.153 mmol) in THF (3 mL) DIEA (35 µL, 0.201 mmol), 2,4,6-trichlorobenzoyl chloride (27 µL, 0.173 mmol) and DMAP (2 mg) were added at room temperature. After 30 min the alcohol **12b** (25 mg, 0.082 mmol), dissolved in THF (1 mL) was added slowly in a dropwise fashion. After being stirred for an additional 2 h, saturated aqueous $NaHCO_3$ solution (5 mL) was added. The layers were separated and the aqueous layer was extracted with CH_2Cl_2 (3 × 20 mL). The combined organic layers were dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (30% EtOAc in petroleum ether) to give the seco compound **127** (94 mg, 73%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.23 (30% EtOAc in petroleum ether);

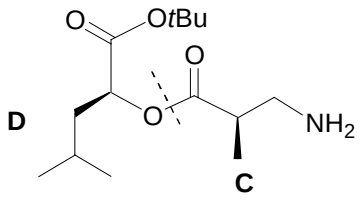
$\boldsymbol{[\alpha]_D^{24}}$ = 1.19 (*c* = 0.28, CH_2Cl_2);

IR (neat): ν_{max} = 3566, 2925, 2900, 1750, 1730, 1653, 1506, 1456, 1258 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): unit A δ = 7.57–7.16 (m, 5H, Ph), 6.82–6.73 (m, 1H, CH_2*CH*CH), 6.14 (d, *J* = 8.6 Hz, 1H, Ph*CH*), 6.00 (dd, *J* = 15.9, 8.5 Hz, 1H, PhCH*CH*), 5.81 (d, *J* = 15.9 Hz, 1H, *CH*CO), 5.12–4.88 (m, 1H *CH*OCH_2), 2.82–2.30 (m, 3H, *CH*Me /CH*CH_2*), 1.44 (s, 9H, CMe_3), 1.10 (d, *J* = 7.1 Hz, Me); unit B δ = 7.74 (d, *J* = 7.3 Hz, 2H, Ph-Fmoc), 7.57–7.16 (m, 6H, Ph-Fmoc), 7.11 (dd, *J* = 8.5, 2.0 Hz, 1H, C_{ortho}), 7.08–7.02 (m, 1H C'_{ortho}), 6.82–6.73 (m, 1H C'_{meta}), 5.12–4.88 (m, N–H), 4.57–4.29 (m, 1H, NH*CH*), 4.21–3.97 (m, 2H, Fmoc), 3.78 (s, 3H, OMe), 3.74–3.61 (m, 1H, Fmoc), 3.01–2.85 (m, 2H, *CH_2*Ph); unit C δ = 7.57–7.16 (m, N–H), 3.29–3.06 (m, 2H, *CH_2*NH), 2.82–2.30 (m, 1H,

CO*CH*), 1.15 (d, *J* = 7.1 Hz, 3H, Me); unit D δ = 4.57–4.29 (m, 1H, CO*CH*), 1.80–1.52 (m, 3H, CH_2CH/*CH*Me), 0.80 (d, *J* = 6.3 Hz, 3H, Me), 0.74 (d, *J* = 6.3 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): unit A δ = 164.5 (CO), 143.7 (CH_2*CH*CH), 137.5 (C_{quart}), 137.2 (Ph*CH*), 135.6 (PhCH*CH*), 133.0 (C_{ortho}), 130.7 (C_{meta}), 127.6 (C_{para}) 126.1 (*CH*CO), 80.1 (*C*(Me)$_3$), 73.7 (*CH*OCH_2), 40.4 (*CH*Me), 33.6 (CH*CH_2*), 28.0 (C*Me_3*), 16.7 (Me); unit B δ = 171.0 (CO), 158.4 (CO, Fmoc), 157.5 (C_{para}), 144.0 (4, Fmoc), 141.1 (5, Fmoc), 131.7 (C_{ortho}), 130.2 (C'$_{ortho}$), 129.7 (C_{quart}), 128.3 (7/7', Fmoc), 127.9 (8/8', Fmoc), 126.1 (9/9', Fmoc), 125.0 (C'$_{meta}$), 119.8 (6/6', Fmoc), 113.8 (C_{meta}), 60.3 (2, Fmoc), 55.0 (NH*CH*), 53.5 (OMe), 52.5 (3, Fmoc), 37.1 (*CH_2*Ph); unit C δ = 173.8 (CO), 47.0 (*CH_2*NH), 43.1 (2, CO*CH*), 14.1 (Me); unit D δ = 171.8 (CO), 70.6 (CO*CH*), 39.3 (*CH_2*CH), 24.6 (*CH*Me), 22.9 (Me), 21.4 (Me) ppm;
HRMS (FT-ICR): calcd. for $C_{54}H_{63}ClN_2O_{10}$ $[M + Na]^+$ 957.4063, found 957.4061.

tert-Butyl(2*S*)-2-{[(2*R*)-3-Amino-2-methylpropanoyl]oxy}-4-methylpentanoate (128)



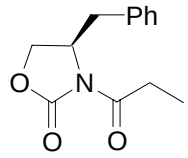
A solution of ester **171** (300 mg, 0.60 mmol) in THF (85 mL) was treated with diethylamine (5 mL) at 0 °C. The reaction mixture was stirred for 15 min at 0 °C and 12 h at room temperature. After evaporation of the solvent in vacuo, the residue was purified by flash chromatography (2% MeOH in CH_2Cl_2) to give the amine **128** (110 mg, 67%) as a colorless liquid. The amine **128** was immediately used for the next step.

***R*$_f$** = 0.19 (2% MeOH in CH_2Cl_2);
IR (neat): ν_{max} = 3286, 2959, 2872, 2285, 1738, 1651, 1556, 1369, 1141 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): unit C δ = 2.98–2.90 (m, 1H, *CH_2*NH_2) 2.82–2.75 (m, 1H, *CH_2*NH_2), 2.62–2.53 (m, 1H, CO*CH*), 1.18 (d, *J* = 7.0 Hz, Me); unit D δ = 4.89 (dd, *J* = 9.3, 4.5 Hz, 1H, CO*CH*), 1.80–1.69 (m, 2H, CH*CH_2*), 1.66–1.54 (m, 1H, *CH*Me), 1.43 (s, 9H, (Me)$_3$), 0.94 (d, *J* = 6.3 Hz, 3H, Me), 0.90 (d, *J* = 6.3 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): unit C δ =175.0 (CO), 45.5 (*CH_2*NH_2), 39.6 (CO*CH*), 14.6 (Me); unit D δ = 169.8 (CO), 81.9 (*C*(Me)$_3$), 71.2 (CO*CH*), 43.4 (CH*CH_2*), 27.9 (C(Me)$_3$),

24.7 (*CH*Me), 23.0 (Me), 21.5 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{14}H_{27}NO_4$ $[M + H]^+$ 274.2013, found 274. 2014.

(4*R*)-4-Benzyl-3-propionyl-1,3-oxazolidin-2-one (129)



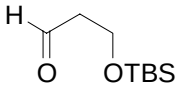
To a cooled solution of Evans chiral auxiliary **141** (10 g, 56 mmol) in THF (150 mL) at –78 °C was added dropwise *n*-BuLi (22.6 mL, 56.5 mmol, 2.5 M in hexane). After 15 min propionyl chloride (5.4 mL, 58.9 mmol) was added dropwise at –78 °C. The reaction mixture was stirred at the same temperature for 1 h, warmed to room temperature within 5 h and stirred again at room temperature for 90 min. The reaction mixture was quenched with saturated aqueous NH_4Cl (50 mL) and diluted with water (50 mL). The layers were separated and the organic layer was concentrated at vacuo. The crude product was dissolved in 5:1 hexane and EtOAc containing 1% MeOH. This solution was washed with saturated aqueous NH_4Cl solution (50 mL) and water (50 mL). The organic layer was dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (30% EtOAc/petroleum ether) yielding N-propionyl-oxazolidinone **129** (11.7 g, 89%) as a colorless oil.

R_f = 0.42 (50% EtOAc/petroleum ether);

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.37–7.24 (m, 5H, Ph), 4.66 (ddd, *J* = 3.3, 7.1, 13.1 Hz, 1H, CH_2*CH*NCH_2), 4.26–4.13 (m, 2H, CH*CH_2*O), 3.29 (dd, *J* =13.4, 3.3 Hz, 1H, Ph*CH_2*CH), 2.03–2.85 (m, 2H, CO*CH_2* Me), 2.79 (dd, *J* = 13.4, 9.6 Hz, 1H, Ph*CH_2*CH), 1.19 (t, *J* =7.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 174.0 (CO), 153 (CO, oxazolidinone), 135.3 (C_{quart}), 129.3 (C_{meta}), 128.9 (C_{ortho}), 127.9 (C_{para}), 66.1 (CH*CH_2*O), 55.1 (CH_2*CH*CH_2), 37.9 (Ph*CH_2*CH), 29.1 (CO*CH_2* Me), 8.2 (CH_2 *Me*) ppm.

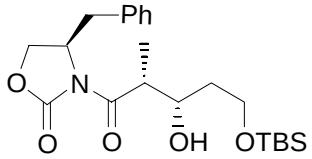
3-{[*tert*-Butyl(dimethyl)silyl]oxy}propanal (130)



Sodium hydride (1.92 g, 80 mmol) was added to THF (200 mL) after being washed with hexane. The propanediol (6 g, 78 mmol) was added to this mixture at room temperature followed by stirring for 45 min, resulting in a large amount of an opaque white precipitate. Then, *tert*-butyldimethylsilyl chloride (11.8 g, 78 mmol) was added and vigorous stirring was continued for 45 min. The mixture was poured into ether (1 L), washed with 10% aqueous K_2CO_3 solution (2 x 200 mL) and brine (2 x 200 mL), dried with $MgSO_4$, filtered and concentrated in vacuo. The crude residue was purified by flash chromatography to get the monoprotected alcohol (13.9 g, 92% yield), which was used for the next step.
To this solution of monoprotected alcohol (2 g, 10.5 mmol) and dried trimethylamine (13.8 g, 10.1 mmol) in dried DMSO (20 mL) a solution of pyridine sulfur trioxide complex (5 g, 31 mmol) in DMSO (15 mL) was added. After 10 min of stirring the mixture was dropped in ice and diluted HCl was added to adjust the pH to about 7. The solution was extracted with ether (2 x 100 mL) and the organic layer was dried with $MgSO_4$, filtered and concentrated in vacuo. The crude residue was purified by flash chromatography (10% EtOAc in petroleum ether) and produced the protected aldehyde **130** (1.64 g, 82%) as a colorless oil.

IR (neat): ν_{max} = 2954, 1397, 1241 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ =9.73–9.66 (s, 1H, CHO), 3.93–3.90 (m, 2H, CH_2), 2.55–2.51 (m, 2H, CH_2), 0.82 (s, 9H, $SiCMe_3$), 0.01 (s, 6H, $SiMe_2$)ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 202.7 (CO), 48.0 (CH_2), 47.2 (CH_2), 26.5 (SiC*Me*$_3$), 18.6 (Si*C*Me_3), –4.7 ($SiMe_2$);

(4*R*)-4-Benzyl-3-[(2*R*,3*S*)-5-{[(tert-butyl)dimethylsilyl]oxy}-3-hydroxy-2-methylpentanoyl]-1,3-oxazolidin-2-one (131)



Titanium(IV) chloride (0.49 mL, 4.50 mmol) was added dropwise to a cooled (0 °C) solution of the oxazolidinone **129** (1.0 g, 4.29 mmol) in CH_2Cl_2 (50 mL). The mixture was stirred for 5 min. Subsequently (–)-sparteine (2.51 g, 10.72 mmol) was added to the yellow slurry. The dark red enolate solution was stirred at 0 °C for 20 min, after which aldehyde **130** (0.89 gm, 4.72 mmol) dissolved in CH_2Cl_2 (10 mL) was added dropwise. The reaction mixture was stirred at 0 °C for 1 h and then quenched with half-saturated NH_4Cl (10 mL). After separation of the layers the organic layer was dried (Na_2SO_4), filtered and concentrated in vacuo. Purification of the residue by flash chomatography (20% EtOAc in petroleum ether) afforded **131** (1.58 g, 87%) as a colorless oil.

R_f = 0.26 (15% EtOAc in petroleum ether);

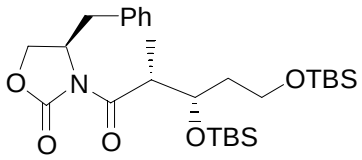
$[\alpha]_D^{24}$ = –42.0 (*c* = 0.98, $CHCl_3$);

IR (neat): ν_{max} = 3504, 2954, 2857, 1782, 1737, 1697, 1241 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.30–7.05 (m, 5H, Ph), 4.65–4.55 (m, 1H, oxazolidinoneCH), 4.17–4.01 (m, 3H, oxazolidinone*CH_2*/CH*CH*OCH_2), 3.82–3.66 (m, 3H, CO*CH*Me /CH_2*CH_2*O), 3.51 (d, *J* = 1.8 Hz, OH), 3.18 (dd, *J* = 8.3, 13.1 Hz, 1H, Ph*CH_2*), 2.70 (dd, *J* = 9.4, 13.4 Hz, 1H, Ph*CH_2*), 1.75–1.51 (m, 2H, CH*CH_2*), 1.21 (d, *J* =7.1 Hz, Me), 0.82 (s, 9H, $SiCMe_3$), 0.17 (s, 6H, $SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 176.3 (CO), 153.1 (CO, oxazolidinone), 135.1 (C_{quart}), 129.4 (C_{meta}), 128.9 (C_{ortho}), 127.3 (C_{para}), 71.2 (CH*CH*OCH_2), 66.0 (oxazolidinoneCH), 61.9 (CH_2*CH_2*O), 55.3 (oxazolidinone*CH_2*), 42.8 (CO*CH*Me), 37.7 (Ph*CH_2*CH), 35.8 (2H, CH*CH_2*), 25.8 (Si*C*Me_3), 18.1 (SiC*Me_3*), 11.2 (Me), –5.6 ($Si(Me)_2$) ppm.

(4*R*)-4-Benzyl-3-[(2*R*,3*S*)-3,5-bis{[(*tert*-butyl)dimethylsilyl]oxy}-2-methylpentanoyl]-1,3-oxazolidin-2-one (132)



tert-Butyldimethylsilyltriflate (0.47 g, 2.0 mmol) was added to a solution of compound **131** (0.60 g, 1.4 mmol) and lutidine (0.46 g, 3.9 mmol) in CH_2Cl_2 (10 mL) and the solution was stirred overnight. Water (5 mL) was added, the mixture stirred for 20 min and the organic layer separated. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried (Na_2SO_4), filtered and concentrated in vacuo. Purification of the residue by flash chromatography (10% EtOAc in petroleum ether) afforded 0.70g (92%) of **132** as a colourless gummy product.

$\boldsymbol{R}_f$ = 0.21 (10% EtOAc in petroleum ether);

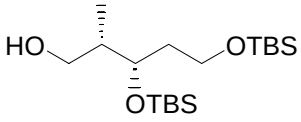
$\boldsymbol{[\alpha]_D^{24}}$ = –56.5 (*c* = 0.98, $CHCl_3$);

IR (neat): ν_{max} = 2954, 2857, 1784, 1703, 1383, 1252, 1209 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.36–7.15 (m, 5H, Ph), 4.63–4.55 (m, 1H, oxazolidinoneCH), 4.19–4.06 (m, 3H, oxazolidinone*CH*$_2$/*CH*OH), 3.93–3.84 (m, 1H, *CH*Me), 3.75–3.61 (m, 2H, CH_2*CH*$_2$O), 3.28 (dd, *J* = 3.0, 13.4 Hz, 1H, PH*CH*$_2$), 2.72 (dd, *J* = 9.6, 13.4 Hz, 1H, PH*CH*$_2$), 1.85–1.74 (m, 1H, CH*CH*$_2$$CH_2$), 1.21 (d, *J* = 6.8 Hz, 3H, Me), 0.87 (s, 18H, 2SiCMe_3), 0.17 (s, 12H, $SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 175.1 (CO), 152.9 (CO, oxazolidinone), 135.3 (C_{quart}), 129.4 (C_{meta}), 128.9 (C_{ortho}), 127.3 (C_{para}), 70.4 (*CH*OH), 65.9 (oxazolidinone*CH*$_2$), 59.4 (CH_2*CH*$_2$O), 55.7 (oxazolidinoneCH), 43.1 (*CH*Me), 38.2 (PH*CH*$_2$), 37.6 (CH*CH*$_2$$CH_2$), 25.9 (Si*C*$Me_3$), 18.1 (SiC*Me*$_3$), 12.3 (Me), –4.8 ($SiMe_2$) ppm.

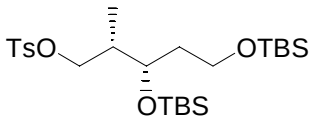
(2*S*,3S)-3,5-Bis{[*tert*-Butyl(dimethyl)silyl]oxy}-2-methylpentan-1-ol (133)



A solution of 2M $LiBH_4$ in THF (0.63 mL, 1.26 mmol) was added dropwise to a solution of aldol product **132** (450 mg, 0.8 mmol) in Et_2O (8 mL) and H_2O (25 μL) at 0 °C. The reaction mixture was stirred for 1 h at 0 °C and then 2 h at room temperature. Then the reaction was quenched by addition of saturated NH_4Cl (5 mL) and the mixture was stirred for 1 hr. After that the organic layer was separated and the aqueous layer extracted with CH_2Cl_2 (2 x 20 mL). The combined organic extracts were dried over MgSO4, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (with 10% EtOAc in petroleum ether) to get pure alcohol **133** (230 mg, 75%) as a colorless oil.

$\boldsymbol{R_f}$ = 18.0 (10% EtOAc in petroleum ether);
$\boldsymbol{[\alpha]_D^{25}}$ = –19.0 (*c* = 0.72, $CHCl_3$);
IR (neat): ν_{max} = 3394, 2955, 2930, 2858, 1470, 1254, 1065 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 3.95–3.88 (m, 1H, *CH*$_2$OH), 3.72–3.56 (m, 3H, *CH*$_2$OH/ *CH*$_2$OTBS), 3.52 (dd, *J* = 10.9, 5.1 Hz, 1H, *CH*OTBS), 2.87 (s, br, 1H, OH), 2.02–1.91 (m, 1H, *CH*Me), 1.68 (q, *J* = 6.3 Hz, 2H, *CH*$_2$CH$_2$), 0.86 (s, 18H, $SiCMe_3$), 0.78 (d, *J* = 7.1 Hz, 3H, Me), 0.08 (s, 3H, SiMe), 0.06 (s, 3H, SiMe), 0.02 (s, 6H, $SiMe_3$) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 72.6 (*CH*OTBS), 65.8 (*CH*$_2$OH) 59.7 (*CH*$_2$OTBS) 39.7 (*CH*CH_3), 35.0 (*CH*$_2$$CH_2$), 25.8 ($SiCMe_3$), 18.1 ($SiCMe_3$), 17.9 ($SiCMe_3$), 12.4 ($CH_3$), –4.4 (SiMe), –4.7 (SiMe), –5.7 ($SiMe_2$) ppm;
HRMS (FT-ICR): calcd. for $C_{18}H_{42}O_3Si_2Na$ $[M + Na]^+$ 385.2564, found 385.2558.

(2*S*,3*S*)-3,5-Bis{[*tert*-butyl(dimethyl)silyl]oxy}-2-methylpentyl-4-methylbenzenesulfonate (134)



p-Toluenesulfonyl chloride (420 mg, 2.20 mmol) was added at 0 °C to a stirred solution of alcohol **133** (400 mg, 1.10 mmol) in pyridine (3 mL). After being stirred for 2 hr, the reaction mixture was quenched by addition of water (10 mL), diluted with Et_2O (50 mL) and washed with 1 N HCl, saturated $NaHCO_3$ solution and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (with 5% EtOAc in petroleum ether) to give pure tosylate **134** (543 mg, 95%) as a slightly yellow oil.

$\boldsymbol{R_f}$ = 0.33 (10% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{25}}$ = –15.7 (*c* = 1.28, $CHCl_3$);

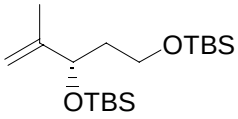
IR (neat): ν_{max} = 3394, 2955, 2930, 2858, 1470, 1254, 1065 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76 (d, *J* = 8.3 Hz, 2H, Ph), 7.31 (d, *J* = 7.8 Hz, 2H, Ph), 4.02 (dd, *J* = 9.1, 5.8 Hz, 1H, *CH*OTBS), 3.86–3.75 (m, 2H, CH_2OTBS), 3.60–3.45 (m, 2H, CH_2OTs), 2.42 (s, 3H, Me), 1.97–1.83 (m, 1H, CH_2CH_2), 1.65–1.53 (m, 1H, CH_2CH_2), 1.50–1.41 (m, H, *CH*Me), 0.85 (s, 9H, $SiCMe_3$), 0.77 (s, 9H, $SiCMe_3$), 0.00 (s, 6H, $SiMe_2$), 0.01 (s, 3H, Me), –0.07 (s, 6H, $SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 144.5 (Ph), 133.0 (Ph), 129.0 (Ph), 127.9 (Ph), 72.6 (*CH*OTBS), 69.1 (CH_2OTs), 59.5 (CH_2OTBS) 37.9 ($CHCH_3$), 36.3 (CH_2CH_2), 25.8 (SiC*Me*$_3$), 25.7 (SiC*Me*$_3$), 21.5 (Me), 18.1 (Si*C*Me_3), 17.9 (Si*C*Me_3), 11.1 (CH_3), –4.5 (SiMe), –4.8 (SiMe), –5.7 ($SiMe_2$) ppm;

HRMS (FT-ICR): calcd. for $C_{25}H_{48}O_5SSi_2Na$ $[M + Na]^+$ 539.2653, found 539.2662.

(3*S*)-3,5-Bis{[*tert*-butyl(dimethyl)silyl]oxy}-2-methylpent-1-ene (135)



A mixture of tosylate **134** (400 mg, 0.81 mmol), NaI (365 mg, 2.44 mmol), DBU (371 mg, 2.44 mmol) in glyme (10 mL) was heated at reflux with stirring for 3 h. After the reaction mixture had cooled to room temperature, Et_2O (50 mL) and H_2O (50 mL) were added and the layers were separated. The combined organic layers were washed with saturated $NaHCO_3$ solution (30 mL), 1 N HCl (30 mL) and brine (50 mL). Then it was dried over MgSO4, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (with 5% EtOAc in petroleum ether) to give pure alkene **135** (266 mg, 95% yield) as a slightly yellow oil.

$\boldsymbol{R_f}$ = 0.31 (5% EtOAc in petroleum ether);

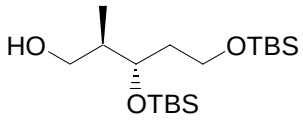
$\boldsymbol{[\alpha]_D^{25}}$ = –14.0 (*c* = 0.88, $CHCl_3$);

IR (neat): ν_{max} = 2954, 2930, 2857, 1471, 1254, 1092 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 4.85 (s, 1H, CH_2=CMe), 4.75 (s, 1H, CH_2=CMe), 4.20 (dd, *J* = 7.3, 5.3 Hz, 1H, *CH*OTBS), 3.68–3.55 (m, 2H, CH_2OTBS), 1.57–1.17 (m, 5H, CH_2CH_2/C*Me*), 0.88 (s, 18H, $2SiCMe_3$), 0.03 (s, 12H, $2SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 147.7 (CH_2=*C*Me), 110.4 (*C*H_2=CMe), 73.3 (*CH*OTBS), 59.7 (*C*H_2OTBS), 39.5 (*C*H_2CH_2), 25.8 (SiC*Me*$_3$), 18.2 (Si*C*Me$_3$), 17.0 (Si*C*Me$_3$), –4.8 (Me), –5.2 ($SiMe_2$), –5.3 ($SiMe_2$) ppm.

(2*R*,3*S*)-3,5-Bis-{[*tert*-butyl(dimethyl)silyl]oxy}-2-methylpentan-1-ol (136)



A solution of 9-BBN in THF (0.5 M, 4.3 mL, 2.17 mmol) was added dropwise at 0 °C to a solution of alkene **135** (250 mg, 0.72 mmol) in THF (2 mL). After that the reaction mixture was stirred at ambient temperature for 3 h and was treated with EtOH (1.35 mL), aqueous NaOH (3 N, 0.9 mL) and 30% aqueous H_2O_2 (0.9 mL) and stirred at rt for additional 2 h. The mixture was saturated with solid K_2CO_3 and extracted with Et_2O (20 mL). The combined organic layers were washed with brine (20 mL), dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (with 10% EtOAc in petroleum ether) and resulted in pure alcohol **136** (218 mg, 83% yield) as a colorless oil.

***R*$_f$** = 0.24 (5% EtOAc in petroleum ether);

[α]$_D^{25}$ = –4.0 (*c* = 0.89, $CHCl_3$);

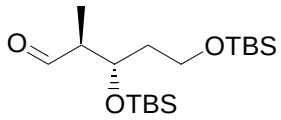
IR (neat): ν_{max} = 3394, 2955, 2930, 2858, 1470, 1254, 1065 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 3.86 (dd, *J* = 10.9, 6.1 Hz, 1H, *C*H_2OH), 3.73 (dd, *J* = 10.9, 4.3 Hz, 1H, *C*H_2OH), 3.62 (t, *J* = 6.6 Hz, 2H, *C*H_2OTBS), 3.49 (dd, *J* = 10.9, 5.3 Hz, 1H, *CH*OTBS), 2.68 (s, br, 1H, OH), 1.90–1.79 (m, 0.5H, *CH*Me), 1.78–1.69 (m, 2H, *C*H_2CH_2), 1.56–1.45 (m, 0.5H, *CH*Me), 0.98 (d, *J* = 7.1 Hz, 3H, CH_3), 0.86 (s, 9H, SiCMe_3), 0.85 (s, 9H, SiCMe_3), 0.05 (s, 6H, $SiMe_2$), 0.00 (s, 6H, $SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 74.1 (*CH*OTBS), 65.1 (*C*H_2OH), 59.6 (*C*H_2OTBS) 38.5 (*CH*CH_3), 37.5 (*C*H_2CH_2), 25.8 (SiC*Me*$_3$), 18.2 (Si*C*Me$_3$), 17.9 (Si*C*Me$_3$), 14.3 (CH_3), –4.4 ($SiMe_2$), –5.3 ($SiMe_2$) ppm;

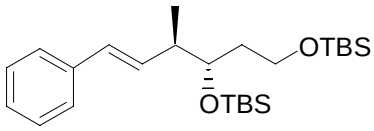
HRMS (FT-ICR): calcd. for $C_{18}H_{42}O_3Si_2Na$ $[M + Na]^+$ 385.25647, found 385.25613.

(2*R*,3*S*)-3,5-Bis-{[*tert*-butyl(dimethyl)silyl]oxy}-2-methylpentananal(137)



Dimethyl sulfoxide (0.22 mL, 3.12 mmol) dissolved in CH_2Cl_2 (5 mL) was added dropwise at –78 °C to a solution of oxalyl chloride (0.139 mL, 1.6 mmol) in CH_2Cl_2 (10 mL). After 5 min, alcohol **136** (0.5 g, 1.3 mmol) in CH_2Cl_2 (10 mL) was added dropwise to the reaction mixture, and stirring was continued at –78° C for 1 h. Triethylamine (0.91 mL, 6.5 mmol) was then added dropwise, and the reaction mixture was warmed to room temperature over 3 h. For the workup the mixture was treated with water (15 mL) and the layers were separated. The aquous layer was extracted with CH_2Cl_2 (2 x 15 mL). The combined organic layers were washed with brine (30 mL), dried with $MgSO_4$, filtered and concentrated in vacuo to give crude aldehyde **137**, which was used for the next step without further purification.

[(1*E*,3*R*,4*S*)-4,6-Di-{[*tert*-butyl(dimethyl)silyl]oxy}-3-methylhex-1-enyl]benzene (138)



A solution of diethyl benzylphosphonate (0.77 mL, 3.71 mmol) in THF (15 mL) was treated at –78 °C with *n*-BuLi (2.5 M in hexane, 0.83 mL, 2.07 mmol). Stirring was continued at –78 °C for 1 h before a solution of aldehyde **137** (from the previous step), dissolved in THF (7 mL) was added dropwise. After being stirred at –78 °C for 1 h, the reaction mixture was allowed to warm gradually to room temperature within 6 h. Aqueous NH_4Cl solution (25 mL) was added and the mixture extracted with Et_2O (3 × 40 mL). The combined organic layers were washed with water (30 mL) and brine (30 mL), dried over $MgSO_4$, filtered and concentrated. The residue was purified by flash chromato-graphy (5% EtOAc in petroleum ether) to give the styrene **138** (345 mg, 58%, 2 steps) as a colorless oil.

$\boldsymbol{R_f}$ = 0.55 (5% EtOAc in petroleum ether);

$[\alpha]_D^{25}$ = +17.8 (*c* = 1.00, CH_2Cl_2);

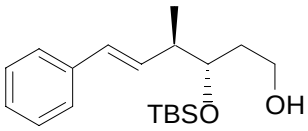
IR (neat): ν_{max} = 2955, 2928, 1471, 1463, 1256, 1067 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.39–7.10 (m, 5H, Ph), 6.37 (d, *J* = 15.9 Hz, 1H, PH*CH*), 6.18 (dd, *J* = 15.6, 7.8 Hz, 1H, PhCH*CH*), 3.88–3.79 (m, 1H, CH*CH*OCH_2), 3.74–3.59 (m, 2H, CH_2OH), 2.54–2.43 (m, 1H, *CH*CH_3), 1.65 (q, *J* = 6.5 Hz, 2H, CH_2CH_2OH), 1.11 (d, *J* = 7.0 Hz, 3H, CH_3), 0.95 (s, 9H, $SiCMe_3$), 0.85 (s, 9H, $SiCMe_3$), 0.11 (s,6H, $SiMe_2$), 0.00 (s, 6H, $SiMe_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 137.8 (C_{quart}), 132.8 (PhCH*CH*), 129.7 (PH*CH*), 128.4 (C_{meta}), 126.8 (C_{para}), 126.0 (C_{ortho}), 72.6 (CH*CH*OCH_2), 60.1 (CH_2OH), 42.7 (*CH*CH_3), 36.7 (CH_2CH_2OH), 25.9 (Si*C*Me_3), 18.1 (SiC*Me*$_3$), 15.5 (CH_3), –4.5 ($SiMe_2$), –5.3 ($SiMe_2$) ppm;

HRMS (FT-ICR): calcd. for $C_{25}H_{46}O_2Si_2$ $[M + Na]^+$ 457.7920, found 457.7925.

(3*S*,4*R*,5*E*)-3-{[*tert*-Butyl(dimethyl)silyl]oxy}-4-methyl-6-phenylhex-5-en-1-ol (139)



A solution of disilyl ether **138** (0.36 g, 0.85 mmol) and pyridinium *para*-toluenesulfonate (70 mg, 0.27 mmol) in MeOH (20 mL) was stirred for 4 h at 50 °C. Thereafter most of the MeOH was removed under reduced pressure and the mixture partitioned between water and Et_2O. The aqueous layer was extracted with Et_2O and the combined organic layers were washed with saturated $NaHCO_3$ solution and brine, dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (15% EtOAc in petroleum ether) to give the primary alcohol **139** (225 mg, 85%) as a colorless oil.

R_f = 0.25 (15% EtOAc in petroleum ether);

$[\alpha]_D^{25}$ = +44.4 (*c* = 0.75, CH_2Cl_2);

IR (neat): ν_{max} = 3352, 2955, 2930, 1465, 1378, 1255, 1093 cm^{-1};

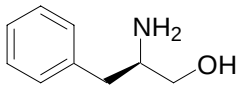
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.26–7.06 (m, 5H, Ph), 6.27 (d, *J* = 16.1 Hz, 1H, PH*CH*), 6.02 (dd, *J* = 15.9, 7.5 Hz, 1H, PhCH*CH*), 3.81–3.73 (m, 1H, *CH*OCH_2), 3.68–3.57 (m, 2H, CH_2OH), 2.50–2.37 (m, 1H, *CH*Me), 1.84 (s, br, OH), 1.61 (q, *J* = 5.8 Hz,

2H, *CH*$_2$CH$_2$OH), 0.99 (d, *J* = 6.8 Hz, 3H, Me), 0.80 (s, 9H, Si–CMe$_3$), 0.00 (2 s, 6H, SiMe$_2$) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 137.5 (C_{quart}), 132.5 (PhCH*CH*), 130.0 (PH*CH*). 128.4 (C_{meta}), 127.0 (C_{para}), 125.9 (C_{ortho}), 74.5 (CH*CH*OCH$_2$), 60.4 (*CH*$_2$OH), 42.6 (*CH*CH$_3$), 34.9 (*CH*$_2$CH$_2$OH), 25.8 (SiC*Me*$_3$), 18.0 (Si*C*Me$_3$), 14.7 (Me), –4.3 (SiMe), –4.5 (SiMe) ppm;

HRMS (FT-ICR): calcd. for $C_{19}H_{32}O_2Si$ $[M + Na]^+$ 343.5310, found 343.5314.

(*R*)-Phenylalanol (140)

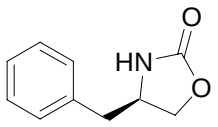


D-Phenyl alanine (20 g, 121.0 mmol) was dissolved in absolute THF (60 mL) followed by the dropwise dropwise addition of boron trifluoride etherate (15.7 mL). After complete addition, the reaction mixture was heated at reflux for 2 h and the resulting solution became colorless and homogeneous. Thereafter to the vigorously refluxed reaction mixture borone-dimethyl sulfide complex (12.4 mL, 131.6 mmol) was added carefully and dropwise over a 30 min period. During the course of addition there was continuous evolution of dimethyl sulfide and hydrogen, and the solution turned from orange to light brown. The reaction mixture was stirred for additional 6 h under reflux condition and then allowed to cool to ambient temperature. The excess borane was quenched by the slow addition of a 1:1 mixture of THF and water (20 mL), followed by the addition of 5M aqueous solution of sodium hydroxide (90 ml). The resulting two-phase mixture was heated to reflux for additional 12 h, cooled to room temperature and filtered through a glass funnel. The residual solid product was washed with small portions of THF and the filtrate was concentrated under reduced pressure to remove the bulk of the THF. The resulting slurry was then extracted with CH_2Cl_2 and the combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated in vacuo. The solid residue was crystallized from boiling EtOAc to give aminoalcohol **140** (16.5 g, 90%) as white needles (m.p.: 91–92 °C, in literature: 88–91 °C[134]).

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.33–7.16 (m, 5H, Ph), 3.63 (dd, *J* = 10.8, 7.2 Hz, 1H, CH*CH*$_2$OH), 3.37 (dd, *J* = 10.8, 7.2 Hz, 1H, CH*CH*$_2$OH), 3.05–3.15 (m, 1H,

CH_2CHNCH_2), 2.78 (dd, *J* = 13.4, 5.2 Hz, 1H, PhCH_2CH), 2.46 (dd, *J* = 13.3, 5.2 Hz, 1H, PhCH_2CH), 2.47-2.56 (m, 3H, NH_2, OH,) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 138.6 (C_{quart}), 129.1 (Ph), 128.5 (Ph), 126.4 (Ph), 66.2 ($CHCH_2OH$), 54.1 (CH_2CHCH_2), 40.8 (PhCH_2CH) ppm.

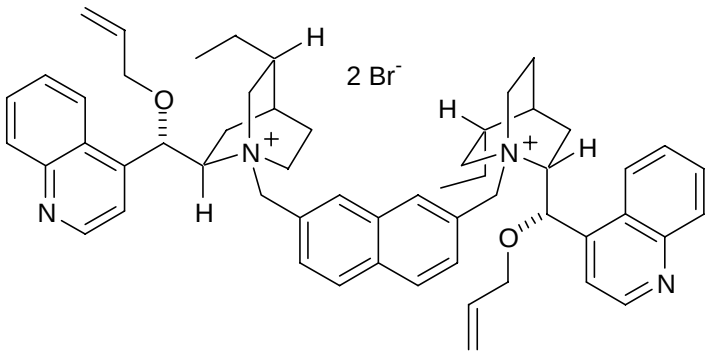
(4R)-4-Benzyl-1,3-oxazolidin-2-one (141)



A mixture of (*R*)-phenylalanol **140** (15.0 g, 92 mmol), anhydrous K_2CO_3 (1.37 g, 99.2 mmol) and diethyl carbonate (25 mL, 203.6 mmol) was placed into a round bottom flask and lowered into a preheated (150 °C) oil bath with distillation flask. The distillation receiver was cooled in an ice bath and the ethanol (ca. 2 mL) was collected. After the distillation, the reaction mixture was allowed to cool to room temperature, diluted with CH_2Cl_2 (400 mL) and washed with water (100 mL). The resulting organic phase was dried over anhydrous $MgSO_4$, filtered and concentrated. The white solid product was crystallized with EtOAc and hexane to produce oxazolidinone **129** (16 g, 91%) as white crystals (m.p.: 87–88.3 °C, in literature: 84.5–86.5 °C[134]).

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.37–7.09 (m, 5H, Ph), 5.51 (br, s, NH), 4.35–4.45 (m, 1H, CH_2CHNCH_2), 4.02–4.16 (m, 2H, $CHCH_2O$), 2.85 (d, *J* =6.8 Hz, 2H, PhCH_2CH) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 159.2 (CO), 135.9 (C_{quart}), 128.9 (Ph), 128.8 (Ph), 127.2 (Ph), 69.6 ($CHCH_2O$), 53.7 (CH_2CHCH_2), 41.4 (PhCH_2CH) ppm.

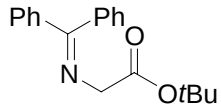
2,7-Bis-[*O*(9)-allylhydrocinchonidinium-*N*-methyl]naphthalene-dibromide (147)



To a suspension of dimer **153** (1.8 g, 1.99 mmol) in CH_2Cl_2 (10 mL) allyl bromide (1.03 mL, 11.9 mmol) and 50% aqueous KOH (2.23 mL, 19.9 mmol) were added. The resulting mixture was stirred vigorously at room temperature for 4 h, in which all solid products were dissolved. The mixture was diluted with water (20 mL) and extracted with dichloromethane (2 x 50 mL). The combined organic extracts were dried over $MgSO_4$, filtered and concentrated in vacuo. The crude solid product was recrystallized from dichloromethane-ether to afford 2.95 g (95% yield) of desired product **147** as a light yellow solid product.

^{1}H NMR (400 MHz, CD_3OD): δ = 8.92 (d, *J* = 4.5 Hz, 2H), 8.52 (s, 2H), 8.32 (d, *J* = 8.3 Hz, 2H), 8.12 (d, *J* = 8.3 Hz, 2H), 8.05 (d, *J* = 8.3 Hz, 2H), 7.96 (d, *J* = 4.5 Hz, 2H), 7.91 (d, *J* = 8.3 Hz, 2H), 7.81–7.69 (m, 4H), 6.64 (s, 2H), 5.33 (d, *J* = 12.3 Hz, 2H), 5.18 (d, *J* = 12.1 Hz, 2H), 4.83–4.51 (m, 2H), 4.34–4.21 (m, 2H), 4.06 (d, *J* = 9.6 Hz, 2H), 3.68 (s, 2H), 3.28 (d, *J* = 15.4 Hz, 2H), 3.07 (d, *J* = 9.8 Hz, 2H), 2.94 (m, 2H), 2.80 (m, 2H), 2.40 (m, 2H), 1.90 (m, 2H), 1.80–1.72 (m, 4H), 1.62–1.57 (m, 2H), 1.33–1.0 (m, 6H), 0.88 (t, *J* = 7.2Hz, 6H) ppm;
^{13}C NMR (100 MHz, CD_3OD): δ = 150.1, 148.6, 147.4, 136.1, 135.6, 134.2, 132.7, 131.1, 130.2, 129.9, 129.2, 127.5, 126.1, 124.5, 121.3, 69.2, 66.9, 64.3, 58.1, 49.6, 49.4, 49.2, 49.0, 48.7, 48.5, 48.6, 36.9, 25.9, 25.3, 22.1, 11.6 ppm;

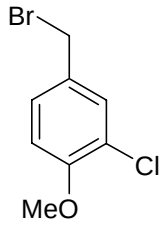
tert-Butyl-N-(diphenylmethylene)glycinate (148)



A solution of *tert*-butyl-2-bromo acetate (7.0 g, 35.9 mmol) in acetonitrile (40 mL) was treated with benzophenonimine **155** (6.5 g, 35.8 mmol) and diisopropylethylamine (6.2 mL, 4.6 g, 35.6 mmol) followed by refluxing of the mixture for 12 h. After cooling to room temperature most of the acetonitrile was removed in vacuo. The remainder was partitioned between water (40 mL) and diethyl ether (60 mL) and the phases were separated. The organic layer was dried with $MgSO_4$, filtered and concentrated in vacuo until the mixture became turbid. Cooling in an icebath provided a first fraction of 4.1 g. Concentration of all the filtrates resulted in another crop; total yield 10.2 g of **148** (96% yield), as a slightly yellow solid product.

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.71–7.20 (m, 10H, Ph), 4.17 (s, 2H, N*CH*$_2$), 1.44 (s, 9H, CMe_3) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.3 (CO), 169.6 (C=N), 139.2 (Ph), 136.0 (Ph), 132.2 (Ph), 130.2 (Ph), 129.9 (Ph), 128.6 (Ph), 128.4 (Ph), 128.3 (Ph), 128.1 (Ph), 127.9 (Ph), 127.5 (Ph), 80.8 (*C*Me$_3$), 56.2 (N*CH*$_2$), 27.9 (C*Me*$_3$) ppm;

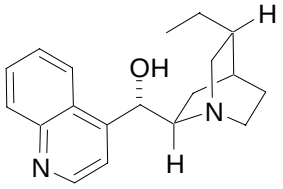
4-(Bromomethyl)-2-chloro-1-methoxybenzene (149)



A mixture of 2-chloro-1-methoxy-4-methylbenzene **157** (4.0 g, 25.5 mmol), NBS (5.0 g, 28.1 mmol) and AIBN (190 mg, 1.15 mmol) in dry CCl_4 (160 mL) was refluxed overnight. After being cooled to room temperature the mixture was washed with NaOH solution (1.5 N, 75 mL) and water (75 mL). The organic layer was dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (3:1 petroleum ether and diethyl ether) to provide the benzyl bromide **149** (4.1 g, 68% yield) as a colorless oil.

***R*$_f$** = 0.52;
IR (neat): ν_{max} = 2946, 1698, 1603, 1503, 1261, 1065 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.40 (d, *J* = 2.0 Hz, 1H, Ph), 7.22 (dd, *J* = 8.4, 2.1 Hz, 1H, Ph), 6.85 (d, *J* = 8.5 Hz, 1H, Ph), 4.43 (s, 2H, *CH*$_2$Br), 3.86 (s, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 154.7(C_{para}), 130.6 (C_{ortho}), 128.4 (C'_{ortho}), 125.6 (C_{quart}), 122.2 (C'_{meta}), 112.5 (C_{meta}), 56.6 (OMe), 32.5 (Me) ppm;
HRMS (FT-ICR): calcd. for C_8H_8BrClO $[M + Na]^+$ 258.5050, found 258.5034.

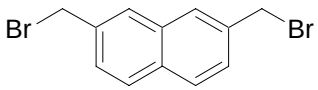
(+)-Hydrocinchonidine (151)



A mixture of (+)-cinchonidine **150** (5.0 g, 16.98 mmol) and 10% Pd/C (1 g) in methanol (130 mL) was stirred under hydrogen atmosphere at room temperature for 10 h. The reaction mixture was filtered through a pad of Celite and the filtrate was concentrated in vacuo. The residue was suspended in hexane (200 mL) and stirred at room temperature for 1 h and then filtered. The solid products were collected to afford 4.6 g **151** (92% yield) of the desired product as a white solid product.

^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.73 (d, *J* = 4.5 Hz, 1H), 7.94 (d, *J* = 8.0 Hz, 1H), 7.81 (d, *J* = 8.0 Hz, 1H), 7.50 (m, 2H), 7.2 (t, *J* =7.2 Hz, 1H), 5.56 (d, *J* = 3.9, 1H), 3.53–3.30 (m, 1H), 2.94 (m, 2H), 2.76–2.69 (m, 2H), 1.84–1.79 (m, 3H), 1.37–1.28 (m, 3H), 1.13–0.99 (m, 2H), 0.72 **(**t, *J* = 6.9Hz, 6H) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 150.1, 148.1, 130.3, 128.9, 126.6, 125.5, 122.9, 118.3, 60.1, 51.0, 50.8, 50.07, 37.9, 26.0. 24.9, 11.9 ppm.

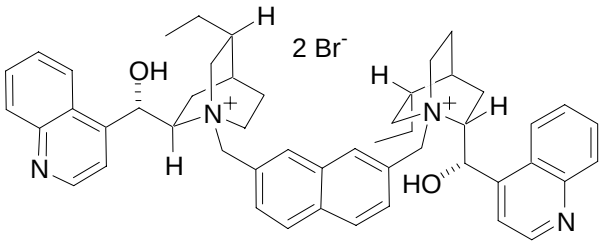
2,7-Bis(bromomethyl)naphthalene (152)



A mixture of 2,7-dimethylnaphthalene (2.0 g, 12.80 mmol), N-bromosuccinimide (5.0 g, 28.16 mmol) and 2,2'-azobisisobutyronitrile (190 mg, 1.15 mmol) in CCl_4 (160 mL) was stirred at reflux for 10 min after which the mixture was cooled to 0 °C. The precipitated succinimide was filtered off and the filtrate evaporated under reduced pressure. The residue was recrystallized from chloroform to give 3.5 g (88% yield) of the desired product **152** as a white solid product.

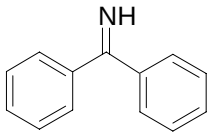
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.82–7.75 (m, 4H, Ph), 7.50 (dd, *J* = 8.3, 1.5 Hz, 2H, Ph), 4.64 (s, 4H, CH_2Br) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 135.8, 132.9, 132.6, 129.5, 127.8, 127.4, 33.6 ppm.

2,7-Bis(hydrocinchonidinium-*N*-methyl)naphthalene-dibromide (153)



A mixture of (+)-hydrocinconidine **151** (2.0 g, 6.75 mmol) with 2,7-bis(bromomethyl)-naphthalene (**152**) (1.04 g, 3.31 mmol) in a mixture of ethanol (5mL), DMF (6 mL) and chloroform (2 mL) was stirred at 100 °C for 6 h. After cooling the reaction mixture to room temperature, the resulting suspension was diluted with methanol (20 mL) and ether (60 mL) and stirred for 1 h. The solid product was filtered and washed with ether. The crude product **153** (2.5 g, 83%) was used for the next step without further purification.

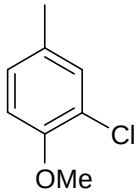
1,1-Diphenylmethanimine (155)



An aqueous solution of 1 mol% of $NiSO_4$ (13.6 mL, 0.54 mmol) was added to a stirring mixture of benzhydrylamine **154** (20 g, 109 mmol) in 250 mL CH_2Cl_2 and an aqueous solution of (1.5 L) of $K_2S_2O_8$ / NaOH (74.2 g, 0.28 mol / 22 g, 0.80 mol). The resulted mixture was stirred vigorously at room temperature for 6 h. Then the mixture was passed through a short Celite column to remove the black precipitates of nickel peroxide. The filtrate was extracted with CH_2Cl_2 (2 x 200 mL). The combined organic extracts were dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography to give pure imine **155** (18.5 g, 93%).

^{1}H NMR (400 MHz, $CDCl_3$): δ = 9.54 (s, NH), 7.79 (d, *J* = 7.0 Hz, 2H, C_{ortho}), 7.56 (d, *J* = 7.0 Hz, 2H, C'_{ortho}), 7.56–7.37 (m, 6H, Ph) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 178.0 (CN), 139.1 (C_{quart}), 132.1 (C_{ortho}), 130.0 (C'_{ortho}), 129.8 (C_{ortho}), 128.2 (C_{meta}), 128.1 (C'_{meta}) ppm.

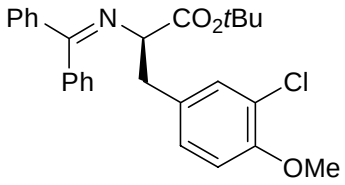
2-Chloro-1-methoxy-4-methylbenzene (157)



A suspension of 2-chloro-4-methylphenol **156** (5.0 g, 35 mmol) in 2.5 N NaOH (15 mL) was cooled to 10 °C and dimethylsulfate (3.2 mL, 38 mmol) was added slowly. Then the reaction mixture was heated overnight under reflux condition, cooled and extracted with ether (3 x 25 mL). The combined organic extracts were dried over $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography to give the amisol derivative **157** (4.4 g, 80%) as a dark yellow liquid.

^{1}H NMR (400 MHz, CD_3OD): δ = 7.17 (s, 1H, C_{ortho}), 7.00 (d, *J* = 8.3 Hz, 1H, C_{meta}), 6.80 (d, *J* = 8.3 Hz, 1H, C'_{ortho}), 3.85 (s, 3H, OMe), 2.26 (s, 3H, Me) ppm;
^{13}C NMR (100 MHz, CD_3OD): δ = 154.5 (C_{para}), 137.8 (C_{quart}), 129.6 (C_{ortho}), 121.7 (C'_{ortho}), 119 (C'_{meta}), 112.8 (C_{meta}), 55.8 (OMe), 21.2 (Me) ppm.

***tert*-Butyl-3-chloro-*N*-(diphenylmethylene)-*O*-methyl-D-tyrosinate (158)**

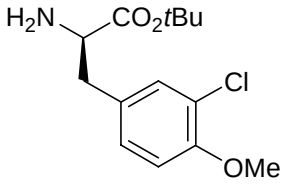


To a stirred mixture of *N*-(diphenylmethylene)glycine *tert*-butyl ester **148** (500 mg, 1.7 mmol) and chiral catalyst **147** (17.0 mg, 0.017 mmol) in toluene/chloroform (7:3, 10 mL) was added the benzyl bromide **149** (500 mg, 2.1 mmol). The reaction mixture was cooled to 0 °C and treated with 50% aqueous KOH (2.5 mL). The mixture stirred at room temperature for approximately 20 h (TLC control). The suspension was diluted with ether (100 mL), washed with water (2 × 50 ml) and the phases were separated. The organic layer was dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (2:1 petroleum ether / diethyl ether) to give the alkylation product **158**

(0.65 g, 87%) as a colorless oil. The *ee* was determined to be 96% by HPLC using a chiral column (DAICEL Chiral OB-H, 250 × 2.6 mm; 98:02 heptane and isopropanol, flow 0.5 mL min^{-1}, t_{minor} = 8.82 min, t_{major} = 9.79 min).

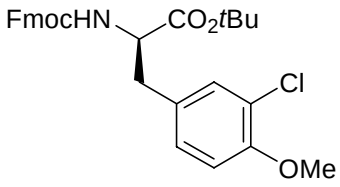
$\boldsymbol{R_f}$ = 0.29 (2:1 petroleum ether / diethyl ether);
$\boldsymbol{[\alpha]_D^{23}}$ = 155.41 (*c* = 0.62, CH_2Cl_2);
IR (neat): ν_{max} = 2975, 2928, 1731, 1622, 1502, 1445, 1256, 1150 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.48 (d, *J* = 7.0 Hz, 2H, $CH_{ortho,}$ CH'_{ortho}), 7.30–7.16 (m, 6 H, Ph,), 6.94–6.83 (m, 2H, Ph, CH_{meta}), 6.70–6.58 (m, 3H, Ph), 4.00 (dd, *J* = 8.9, 4.4 Hz, 1H, *CH*NH), 3.72 (s, 3H, OMe), 3.08–2.93 (m, 2H, CH*CH_2*), 1.35 (s, 9H, CMe_3) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.0 (CO), 153.8 (C_{para}), 139.8 (C_{ortho}), 136.7 (C'_{ortho}), 131.7 (C_{quart}), 130.6 (Ph), 129.5 (Ph), 129.1 (Ph), 128.8 (Ph), 128.6 (Ph), 128.3 (Ph), 128.0 (Ph), 122.2 (C_{meta}), 112.0 (C'meta), 81.6 (*C*Me_3), 68.0 (*CH*NH), 56.5 (OMe), 38.8 (CH*CH_2*), 28.4 (C*Me_3*) ppm;
HRMS (FT-ICR): calcd. for $C_{27}H_{28}ClNO_3$ $[M + Na]^+$ 472.5035, found 472.5030.

tert-Butyl-3-chloro-*O*-methyl-D-tyrosinate (159)



A solution of the alkylated imine **158** (500 mg, 1.1 mmol) in THF (10 mL) and 15% aqueous citric acid (5 mL) was stirred at room temperature for 14 h. Then the mixture was diluted with Et_2O (10 mL) and extracted with 1 M HCl (3 × 10 mL). The combined aqueous layers were washed with Et_2O (10 mL), basified with solid K_2CO_3 and then extracted with EtOAc (3 × 15 mL). The organic extracts were dried over $MgSO_4$, filtered and concentrated in vacuo resulting in the crude amino acid **159** which was used in the next step without further purification.

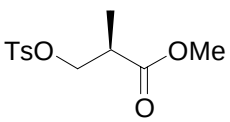
tert-Butyl-3-chloro-_N_-[(9_H_-fluoren-9-ylmethoxy)carbonyl]-_O_-methyl-D-tyrosinate (161)



The crude amino ester **159** (400 mg, 1.4 mmol) was dissolved in THF (10 mL) and 10% aqueous Na_2CO_3 (10 mL) was added, followed by FmocCl (**160**) (0.4 g, 1.5 mmol). The reaction mixture was stirred 14 h at room temperature. The aqueous layer was extracted with EtOAc (3 × 10 mL), dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (10% EtOAc in petroleum ether) to give **161** (510 mg, 72%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.30 (10% EtOAc in petroleum ether);
$\boldsymbol{[\alpha]_D^{25}}$ = –25.07 (*c* = 1.22, CH_2Cl_2);
IR (neat): ν_{max} = 3421, 3335, 3064, 2978, 2933, 1732, 1606, 1503, 1450, 1368, 1280 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76 (d, *J* = 7.3 Hz, 2H, Fmoc), 7.61–7.17 (m, 7H, Ph, Fmoc), 6.98 (d, *J* = 8.0 Hz, 1H, Ph), 6.81 (d, *J* = 8.3 Hz, 1H, Ph), 5.30 (d, *J* = 7.8 Hz, 1H, NH), 4.53–4.41 (m, 2H, Fmoc), 4.36–4.28 (m, 1H, *CH*NH), 4.21 (t, *J* = 6.9 Hz, 1H, Fmoc), 3.86 (s, 3H, OCH_3), 3.05–2.96 (m, 2H, CH*CH_2*), 1.43 (s, 9H, CMe_3) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 170.3 (CO), 155.4 (CO, Fmoc), 154.0 (C_{para}), 143.7 (Fmoc), 141.2 (Fmoc), 131.2 (C_{ortho}), 128.7 ($C_{'quart}$), 127.7 ($C_{'ortho}$), 127.0 (Fmoc), 125.0 (Fmoc), 122.1 (C_{meta}), 119.9 (Fmoc), 111.9 (C'_{meta}), 82.7 (*C*Me_3), 66.9 (Fmoc), 56.1 (OMe), 55.0 (*CH*NH), 47.1 (Fmoc), 37.1 (CH*CH_2*), 27.9 (C*Me_3*) ppm;
HRMS (FT-ICR): calcd. for $C_{25}H_{22}ClNO_5$ $[M + Na]^+$ 474.1078, found 474.1077.

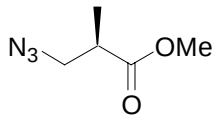
Methyl-(2_R_)-2-methyl-3-{[(4-methylphenyl)sulfonyl]oxy}propanoate (163)



D-(–)-Methyl-β-hydroxy-iso-butyrate **162** (2.0 g, 16.9 mmol), TsCl (5.0 g, 26.2 mmol) and Et_3N (4.48 mL, 44.2 mmol) were stirred in CH_2CL_2 (40 mL) for 3h. Water (100 mL) and diethyl ether (100 mL) were added and the mixture stirred for further 5 min. The aqueous layer was separated and washed with ether. The organic layers were washed with 1 N HCl (100 mL), $NaCO_3$ solution (100 mL) and brine (100 mL) respectively and dried over $MgSO_4$. The solvent was removed and the crude residue was purified (10% then 20% EtOAc in petroleum ether) to produce **163** (3.6 g, 78%) as a colorless oil.

R_f = 0.20 (20% EtOAc in petroleum ether);
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.72 (d, *J* = 8.3 Hz, 2H, Ph), 7.31 (d, *J* = 8.3 Hz, 2H, Ph), 4.18–4.12 (m, 1H, *CH*$_2$OTs), 4.03–3.99 (m, 1H, *CH*$_2$OTs), 3.58 (s, 3H, OMe), 2.80–2.75 (m, 1H, *CH*Me), 2.40 (s, 3H, MePh), 1.12 (d, *J* =7.3 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 170.9 (CO), 144.8 (C_{quart}), 132.5 (C_{ortho}), 129.7 (C_{meta}), 127.8 (C_{para}), 70.6 (*CH*$_2$OTs), 51.9 (OMe), 39.0 (*CH*CH_3), 21.4 (MePh), 13.4 (Me) ppm.

Methyl (2*R*)-3-azido-2-methylpropanoate (164)



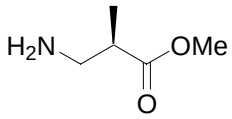
A mixture of tosylate **163** (2.0 g, 7.3 mmol) and sodium azide (1.0 g, 15.3 mmol) in DMSO (30 mL) was heated at 80 °C for 2–3 h.After being cooled to room temperature, 30 mL of water were added and the mixture was extracted with CH_2Cl_2 (3 × 30 mL). The combined extracts were dried with $MgSO_4$, filtered and concentrated in vacuo. The crude product was purified by flash chromatography (20% EtOAc in petroleum ether) resulting in azide **164** (0.9 g, 85%) as a colorless oil.

***R*$_f$** = 0.47 (20% EtOAc in petroleum ether);
$[\alpha]_D^{22}$ = –14.32 (*c* = 0.99, CH_2Cl_2);
IR (neat): ν_{max} = 2982, 2103, 1732, 1463, 1381, 1199 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 3.68 (s, 3H, OMe), 3.56–3.46 (m, 1H, *CH*$_2$N$_3$), 3.35 (dd, *J* = 12.1, 5.8 Hz, 1H, *CH*$_2$N$_3$), 2.73–2.60 (m, 1H, *CH*Me), 1.28 (d, *J* = 7.0 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 174.7 (CO), 54.1 (OMe), 52.9 (CH_2N_3), 40.0 (*CH*Me), 15.1 (Me) ppm;

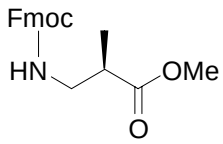
HRMS (FT-ICR): calcd. for $C_5H_9N_3O_2$ $[M + Na]^+$ 166.0587, found 166.0585.

Methyl-(2*R*)-3-amino-2-methylpropanoate (165)



A mixture of azide **164** (0.28 g, 2.0 mmol) and 10% Pd–C (140 mg) in MeOH (2 mL) was stirred under hydrogen atmosphere at room temperature for 15 h. The reaction mixture was filtered through a pad of celite. Concentration of the filtrate gave the crude amine 1**65** which was immediately protected without further purification.

Methyl-(2*R*)-3-{[(9H-Fluoren-9-ylmethoxy)carbonyl]amino}-2-methylpropanoate (166)



The crude amino ester **165** (300 mg, 2.5 mmol) was dissolved in THF (10 mL), 10% aqueous Na_2CO_3 (10 mL) was added, followed by FmocCl (1.0 g, 3.8 mmol). The reaction mixture was stirred at room temperature for 14 h. The aqueous layer was extracted with EtOAc (2 × 30 mL) and the combined organic extracts were dried with $MgSO_4$, filtered and concentrated. The residue was purified by flash chromatography (20% EtOAc in petroleum ether) providing the protected amine **166** (0.61 g, 90%) as a colorless solid product.

$\boldsymbol{R_f}$ = 0.25 (20% EtOAc in petroleum ether);

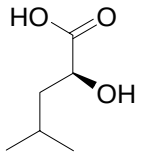
$\boldsymbol{[\alpha]_D^{22}}$ = –13.2 (*c* = 0.962, CH_2Cl_2);

IR (neat): ν_{max} = 3310, 2250, 1760, 1520, 1350, 1250 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.77–7.28 (m, 8H, Fmoc), 5.43–5.33 (m, 1H, NH), 4.40

(d, *J* = 6.3 Hz, 2H, Fmoc), 4.22 (t, *J* = 6.8 Hz, 1H, Fmoc), 3.71 (s, 3H, OMe), 3.46–3.29 (m, 2H, *CH*$_2$NH), 2.79–2.68 (m, 1H, *CH*Me), 1.20 (d, *J* = 7.3 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 176.1 (CO), 156.8 (Fmoc), 144.3 (Fmoc), 141.7 (Fmoc), 128.0 (Fmoc), 127.4 (Fmoc), 125.4 (Fmoc), 120.3 (Fmoc), 67.0 (Fmoc), 52.2 (OMe), 47.6 (Fmoc), 43.7 (*CH*$_2$NH), 40.2 (*CH*Me), 15.1 (Me) ppm;
HRMS (FT-ICR): calcd. for $C_{20}H_{21}NO_4$ $[M + Na]^+$ 362.3850, found 362.3855.

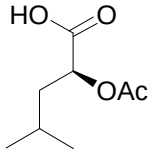
(2*S*)-2-Hydroxy-4-methylpentanoic acid (168)



L-Leucine **167** (5.0 g, 38 mmol) was dissolved in diluted 0.5 M H_2SO_4 (150 mL) at 0 °C. Sodium nitrite (15.7 g, 228 mmol) was added. The mixture was allowed to stir at 0 °C for 6 h. The reaction mixture was extracted with diethyl ether. The combined organic layers were dried over $MgSO_4$ and concentrated in vacuo. The residue was azeotroped with toluene twice to yield a yellow syrup. After removal of residual toluene under high vacuum, the residue was washed again with hexane and filtered to give **168** (3.27 g, 65%) as a colorless solid product.

^{1}H NMR (400 MHz, $CDCl_3$): δ = 4.28 (dd, *J* = 8.4, 4.9 Hz, 1H, *CH*OH), 1.94–1.87 (m, 1H, *CH*$(Me)_2$), 1.45–1.35 (m, 2H, CH*CH*$_2$,) 0.96 (d, *J* = 6.8 Hz, 6H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 175.0 (CO), 68.8 (CHOH), 43.2 (CH*CH*$_2$), 24.4 (*CH*Me), 23.1 (Me), 21.4 (Me) ppm.

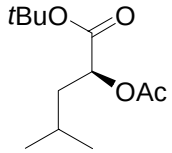
(2*R*)-2-(Acetyloxy)-4-methylpentanoic acid (169)



To compound **168** (1.2 g, 9 mmol) acetyl chloride (10 mL) was added at 0 °C. The reaction mixture was then refluxed at 60 °C for 4 hr. Excess acetyl chloride was removed under

vacuum. Diethyl ether was added and the solution was washed with water. The organic layer was dried and concentrated under vacuum to give acid **169** as a crude oil, which was used in the next step without further purification.

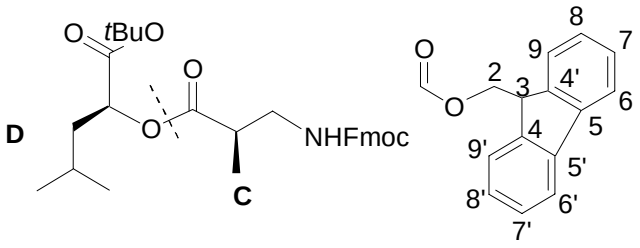
tert-butyl-(2*S*)-2-(acetyloxy)-4-methylpentanoate (170)



Acid **169** and *tert*-butyl alcohol (1.31 g, 18.9 mmol) were dissolved in CH_2Cl_2 (30 mL) and DMAP (361mg, 2.97 mmol) was added. Then DCC (2.45 g, 11.9 mmol) in CH_2Cl_2 (10 mL) was added at 0 °C. The reaction mixture was stirred at room temperature for 12 h. After the urea was filtered the organic layer was washed with water, dried over $MgSO_4$ and evaporated in vacuo. The crude residue was purified by flash column chromatography (15% EtOAc in petroleum ether) to give ester **170** as a light yellow oil (1.45 g, 70%, 2 steps).

$\boldsymbol{R_f}$ = 0.35 (15% EtOAc in petroleum ether);
^{1}H NMR (400 MHz, $CDCl_3$): δ = 4.75 (d, *J* = 9.3 Hz, 1H, *CH*OAc), 1.98 (s, 3H, CO*Me*), 1.65–1.60 (m, 2H, CH*CH*$_2$), 1.49–1.45 (m, 1H, *CH*Me), 1.33 (s, 9H, CMe_3), 0.82 (d, *J* = 6.8 Hz, 6H, CH_3) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 170.2 (CO), 169.6 (CO, Ac), 81.5 (*C*Me_3), 71.2 (*CH*OAc), 39.5 (CH*CH*$_2$), 27.6 (C*Me*$_3$), 24.4 (CHMe), 22.7 (Me), 21.3 (Me), 20.3 (CO*Me*) ppm.

tert-Butyl(2*S*)-2-[((2*R*)-3-{[(9*H*-Fluoren-9-ylmethoxy)carbonyl]amino}-2-methylpropanoyl)oxy]-4-methylpentanoate (171)



To a solution of hydroxy ester **15c** (180 mg, 0.95 mmol), the amino acid **14c** (373 mg, 1.14 mmol) and DMAP (50 mg, 0.40 mmol) in CH_2Cl_2 (3 mL) at 0 °C was added dropwise a solution of DCC (293 mg, 1.42 mmol) in CH_2Cl_2 (3 mL). The clear solution was stirred 30 min at 0 °C and then 5 h at room temperature. The white precipitate was filtered off and the filtrate concentrated. The residue was dissolved in Et_2O and washed with 0.5 N HCl (10 mL), saturated $NaHCO_3$ solution (10 mL) and brine (10 mL). The ether layer was dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash chromatography (10% of EtOAc in petroleum ether) to give the ester **171** (378 mg, 80%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.25 (10% of EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{24}}$ = –32.41 (*c* = 0.44, CH_2Cl_2);

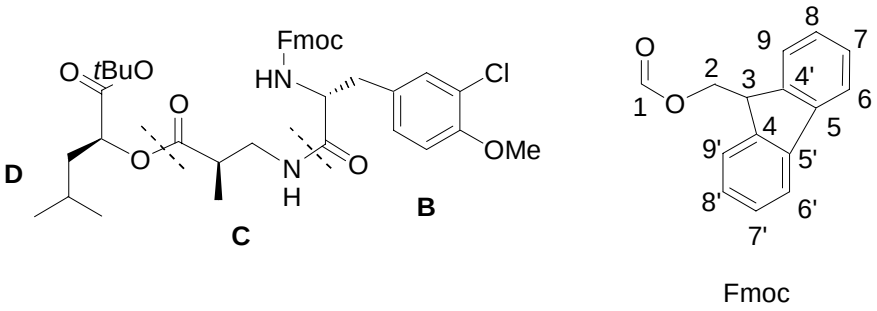
IR (neat): ν_{max} = 3366, 2958, 1732, 1522, 1450, 1369, 1249, 1159 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): unit C δ = 7.77–7.22 (m, 8H, Fmoc), 6.03–5.92 (m, N–H), 4.38–4.26 (m, 2H, Fmoc), 4.18 (t, *J* = 7.3 Hz, 1H, Fmoc), 3.59–3.50 (m, 1H, *CH*$_2$NH), 3.35–3.25 (m, 1H, *CH*$_2$NH), 2.83–2.73 (m, 1H, CO*CH*), 1.21 (d, *J* = 7.0 Hz, 3H, Me); unit D δ = 4.92 (dd, *J* = 9.3, 4.5 Hz, 1H, CO*CH*), 1.83–1.57 (m, 3H, *CH*$_2$Me/*CH*Me), 1.47 (s, 9H, CMe_3), 0.94 (d, *J* = 6.3 Hz, 3H, Me), 0.91 (d, *J* = 6.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): unit C δ = 174.3 (CO), 156.5 (CO, Fmoc), 144.0 (4,4', Fmoc), 141.2 (5,5', Fmoc), 127.5 (7,7', Fmoc), 126.9 (8,8', Fmoc), 125.2 (9,9', Fmoc), 119.8 (6,6', Fmoc), 66.7 (2, Fmoc), 47.1 (3, Fmoc), 43.6 (*CH*$_2$NH), 40.8 (CO*CH*), 14.5 (Me), unit D δ = 170.3 (CO), 82.5 (*C*Me_3), 71.3 (CO*CH*), 39.5 (CH*CH*$_2$), 27.9 (C*Me*$_3$), 24.7 (CH_2*CH* Me), 23.0 (Me), 21.5 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{29}H_{37}NO_6$ $[M + Na]^+$ 518.2513, found 518.2511.

tert-Butyl(2*S*)-2-{[(2*R*)-3-({3-chloro-*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-*O*-methyl-D-tyrosyl}amino)-2-methylpropanoyl]oxy}-4-methylpentanoate (172)



The amine **128** (150 mg, 0.54 mmol), the amino acid **13c** (243 mg, 0.54 mmol) and HOBT (73 mg, 0.54 mmol) were dissolved in dry THF (3 mL), followed by the addition of DCC (166 mg, 0.81 mmol), dissolved in THF (2 mL) at 0 °C. The mixture was stirred for 7 h at room temperature before it was filtered and concentrated. The residue was diluted with Et_2O and washed with water. The organic layer was dried over $MgSO_4$, filtered and concentrated. The residue was purified by flash chromatography (30% EtOAc in petroleum ether) to give the tripeptide analogue **172** (325 mg, 84%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.32 (30% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{24}}$ = –21.93 (*c* = 1.25, CH_2Cl_2);

IR (neat): ν_{max} = 3317, 2950, 2152, 1738, 1670, 1504, 1257 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): unit B δ = 7.74 (d, *J* = 7.5 Hz, 2H, Fmoc), 7.56–7.15 (m, 7H, Fmoc, C_{ortho}), 7.03 (d, *J* = 8.0 Hz, 1H, C'$_{ortho}$), 6.79 (d, *J* = 8.3 Hz, 1H, C'$_{meta}$), 5.61 (d, *J* = 8.3 Hz, NH), 4.51–4.38 (m, 1H, NH*CH*), 4.22–4.07 (m, 2H, Fmoc), 3.81 (s, 3H, OMe), 3.78–3.70 (m, 1H, Fmoc), 3.20–3.08 (m, 2H, CH_2Ph), unit C δ = 5.00–4.95 (m, NH), 3.04 (dd, *J* = 13.9, 6.1, 1H, CH_2NH), 2.95 (dd, *J*= 13.9, 6.8 Hz, 1H, CH_2NH), 2.82–2.70 (m, 1H, COCH), 1.17 (d, *J* = 6.8 Hz, Me), unit D δ = 4.51–4.38 (m, 1H, CO*CH*), 1.78–1.53 (m, 3H, CH_2CH/*CH*Me), 1.43 (s, 9H, CMe_3), 0.93 (d, *J* = 6.3 Hz, 3H, Me), 0.90 (d, *J* = 6.3 Hz, 3H, Me) ppm;

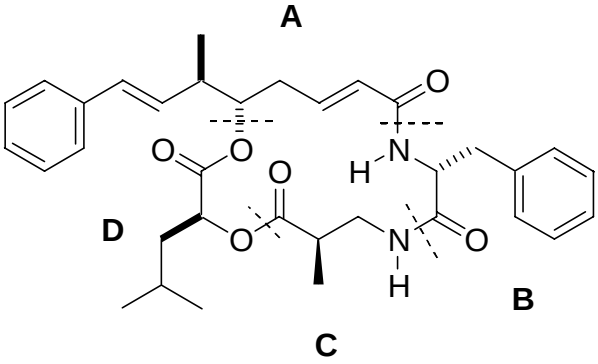
^{13}C NMR (100 MHz, $CDCl_3$): unit B δ =170.0 (CO), 155.6 (CO, Fmoc), 153.8 (C_{para}), 143.7 (4/4', Fmoc), 141.2 (5/5', Fmoc), 131.0 (C'$_{ortho}$), 129.5 (C_{ortho}), 127.6 (C_{quart}), 127.2 (7/7', Fmoc), 125.6 (8/8', Fmoc), 125.0 (9/9', Fmoc), 122.1 (C'$_{meta}$), 119.9 (6/6', Fmoc), 112.1 (C_{meta}), 67.0 (2, Fmoc), 60.3 (NH*CH*), 56.0 (OMe), 41.8 (3, Fmoc), 38.0 (CH_2Ph), unit C δ = 173.6 (CO), 47.0 (CH_2NH), 40.6 (CO*CH*), 14.7 (Me), unit D δ =171.0 (CO),

83.1 (*C*Me$_3$), 71.0 (CO*CH*), 39.4 (CH*CH$_2$*), 27.9 (CMe$_3$), 124.8 (*CH*Me), 22.9 (Me), 21.1 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{39}H_{47}ClN_2O_8$ $[M + Na]^+$ 729.2913, found 729.2912.

2.2 Synthesis of a Cryptophycin analogue 11

Cryptophycin (11)



To a solution of seco compound **177** (100 mg, 0.11 mmol) in CH_2Cl_2 (3 mL) trifluoroacetic acid (5 mL) was added slowly at 0 °C and the mixture stirred for 2 h at room temperature. The solvent was removed in vacuo and toluene (5 mL) was added. The residue was dissolved in THF (3 mL) and diethylamine (3 mL) was added dropwise at 0 °C. The reaction mixture was stirred for 2 h at room temperature, followed by removal of the solvents in vacuo. This crude product **177a** was dissolved in dry DMF (15 mL) and the mixture was treated successively with TBTU (60 mg, 0.16 mmol), HOBT (2 mg), and DIEA (66 µl, 0.38 mmol) at room temperature. The reaction mixture was stirred for 2 h before saturated $NaHCO_3$ solution (10 mL) was added and stirring continued for 1 h. After extraction with CH_2Cl_2 (3 × 15 mL) the combined organic layers were dried over $MgSO_4$, filtered and concentrated. The residue was purified by flash chromatography (50% EtOAc in petroleum ether) to provide macrocycle **11** (47 mg, 74%) as a slightly yellow oil.

$\boldsymbol{R_f}$ = 0.29 (50% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{24}}$ = 7.04 (*c* = 0.32, CH_2Cl_2);

IR (neat): ν_{max} = 3271, 2960, 1744, 1714, 1675, 1540, 1457, 1341, 1175 cm^{-1};

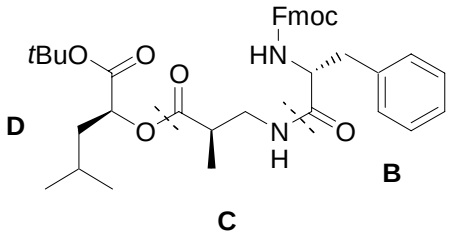
^{1}H NMR (400 MHz, $CDCl_3$): unit A δ = 7.34–7.13 (m, 5H, Ph), 6.66 (ddd, *J* = 15.1, 10.0, 4.9 Hz, 1H, CH_2*CH*CH), 6.36 (d, *J* = 15.9 Hz, 1H, Ph*CH*), 5.97 (dd, *J* = 15.9, 8.8 Hz, 1H,

PhCH*CH*), 5.70 (d, *J* = 15.6 Hz, 1H, *CH*CO), 5.05–4.94 (m, 1H, *CH*OCH$_2$), 2.57–2.44 (m, 2H, *CH*Me/CH*CH$_2$*), 2.39–2.26 (m, 1H, CH*CH$_2$*), 1.09 (d, *J* = 6.8 Hz, 3H, Me); unit B δ = 7.34–7.13 (m, 5H, Ph), 5.62 (d, *J* = 8.3 Hz, 1H, N–H), 4.86–4.74 (m, 1H, NH*CH*), 3.18 (dd, *J* = 14.1, 5.3 Hz, 1H, *CH$_2$*Ph), 3.08 (dd, *J* = 14.4, 7.3 Hz, 1H, *CH$_2$*Ph); unit C δ = 7.06–6.99 (m, 1H, N–H), 3.26–3.22 (m, 2H, *CH$_2$*NH), 2.70–2.60 (m, 1H, CO*CH*), 1.20 (d, *J* = 8.0 Hz, 3H, Me); unit D δ= 4.86–4.74 (m, 1H, CO*CH*), 1.67–1.54 (m, 2H, CH*CH$_2$*), 1.43–1.36 (m, 1H, *CH*Me), 0.71 (d, *J* = 6.3 Hz, 3H, Me), 0.68 (d, *J* = 6.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): unit A δ =165.3 (CO), 141.6 (CH$_2$*CH*CH), 136.7 (C$_{quart}$), 131.8 (Ph*CH*), 129.2 (PhCH*CH*), 128.6 (C$_{ortho}$), 127.5 (C$_{meta}$), 126.1 (C$_{para}$), 125.0 (*CH*CO), 72.4 (*CH*OCH$_2$), 42.3 (*CH*Me), 38.1 (CH*CH$_2$*), 17.3 (Me); unit B δ = 170.8 (CO), 130.1 (C$_{quart}$), 128.6 (C$_{ortho}$), 126.9 (C$_{meta}$), 126.1 (C$_{para}$), 53.7 (NH*CH*), 35.6 (*CH$_2$*Ph); unit C δ = 175.9 (CO), 40.8 (*CH$_2$*NH), 39.5 (CO*CH*), 14.2 (Me); unit D δ = 171.1 (CO), 71.1 (CO*CH*), 36.4 (CH*CH$_2$*), 24.4 (*CH*Me), 22.7 (Me), 21.2 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{34}H_{42}N_2O_6$ $[M + Na]^+$ 597.2935, found 597.2934.

tert-Butyl(2*S*)-2-{[(2*R*)-3-({*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-D-phenylalanyl}amino)-2-methylpropanoyl]oxy}-4-methylpentanoate (174)



A mixture of the amine **128** (150 mg, 0.54 mmol), the Fmoc-protected D-phenylalanine **175** (212 mg, 0.54 mmol) and HOBT (50 mg, 0.37 mmol) in dry THF (3 mL) was treated with DCC (166 mg, 0.81 mmol) dissolved in THF (2 mL) at 0 °C. The reaction mixture was stirred for 7 h at room temperature, filtered and concentrated. The residue was diluted with Et_2O and the resulting mixture washed with water. The organic layer was dried over $MgSO_4$, filtered and concentrated. The residue was purified by flash chromatography (30% EtOAc in petroleum ether) to give the tripeptide analogue **174** (280 mg, 81%) as a colorless oil.

***R*$_f$** = 0.33 (30% EtOAc in petroleum ether);

$[\alpha]_D^{24}$ = –23.63 (*c* = 1.25, CH_2Cl_2);

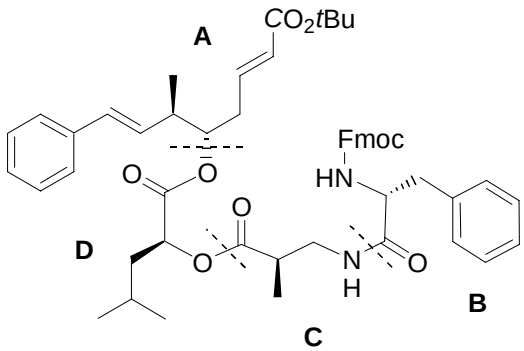
IR (neat): ν_{max} = 3064, 2957, 2341, 1732, 1665, 1539, 1450, 1246, 1106 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): unit B δ = 7.65 (d, *J* = 7.3 Hz, 2H, Fmoc), 7.51–6.95 (m, 11H, Ph/Fmoc), 5.49 (d, *J* = 8.0 Hz, 1H, NH), 4.45–4.27 (m, 1H, NH*CH*), 4.16–3.98 (m, 2H, Fmoc), 3.69–3.55 (m, 1H, Fmoc), 3.09–3.00 (m, 2H, CH_2Ph); unit C δ = 4.91–4.84 (m, 1H, NH), 2.95 (dd, *J* = 13.7, 7.2 Hz, 1H, CH_2NH), 2.85–2.75 (m, 1H, CH_2NH), 2.70–2.61 (m, 1H, COCH), 1.06 (d, *J* = 7.0 Hz, 3H, Me); unit D δ = 4.45–4.27 (m, 1H), 1.69–1.40 (m, 3H, CH_2CH/*CH*Me), 1.34 (s, 9H, CMe_3), 0.84 (d, *J* = 6.3 Hz, 3H, Me), 0.81 (d, *J* = 6.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): unit B δ = 170.7 (CO), 155.6 (CO, Fmoc), 143.7 (4/4', Fmoc), 141.2 (5/5', Fmoc), 136.4 (C_{quart}), 129.5 (C_{ortho}), 129.2 (C_{para}), 128.5 (C_{meta}), 127.6 (7/7', Fmoc), 127.0 (8/8', Fmoc), 125.0 (9/9', Fmoc), 119.9 (6/6', Fmoc), 66.9 (2, Fmoc), 56.1 (NH*CH*), 41.8 (3, Fmoc), 39.1 (CH_2Ph); unit C δ = 173.7 (CO), 47.0 (CH_2NH), 40.5 (CO*CH*), 14.7 (Me); unit D δ = 171.0 (CO), 82.9 (*C*Me_3), 71.0 (CO*CH*), 39.5 (CHCH_2), 27.9 (CMe_3), 24.7 (*CH*Me), 23.0 (Me), 21.5 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{38}H_{46}N_2O_7$ $[M + Na]^+$ 665.3197, found 665.3202.

tert-Butyl(2*E*,5*S*,6*R*,7*E*)-5-[((2*S*)-2-{[(2*R*)-3-({*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-D-phenylalanyl}amino)-2-methylpropanoyl]oxy}-4-methylpentanoyl)oxy]-6-methyl-8-phenylocta-2,7-dienoate (177)



To a solution of the fully protected amino acid **174** (230 mg, 0.35 mmol) in CH_2Cl_2 (7 mL) trifluoroacetic acid (7 mL) was added at 0 °C followed by stirring of the reaction mixture for 3 h at room temperature. Then toluene (5 mL) was added and the mixture concentrated. This was repeated twice and the crude acid **176** was subjected to the next reaction without further purification.

To a solution of the *N*-protected crude acid **176** (200 mg, 0.34 mmol) in THF (3 mL) DIEA (45 µL, 0.40 mmol), 2,4,6-trichlorobenzoyl chloride (91 µL, 0.374 mmol) and DMAP (2 mg) were added. After stirring for 30 min the alcohol **12b** (51 mg, 0.17 mmol), dissolved in THF (1 mL) was added slowly in a dropwise fashion. After 2 h saturated aqueous $NaHCO_3$ solution (5 ml) was added and the layers were separated. The aqueous layer was extracted with CH_2Cl_2 (10 mL). The combined organic layer was dried over $MgSO_4$, filtered and concentrated. The crude product was purified by flash chromatography (30% EtOAc in petroleum ether) to give the protected seco compound **177** (231 mg, 75%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.21 (30% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{25}}$ = 1.88 (*c* = 0.84, CH_2Cl_2);

IR (neat): ν_{max} = 3318, 3063, 2959, 1735, 1600, 1545, 1450, 1254, 1152, 1081 cm^{-1};

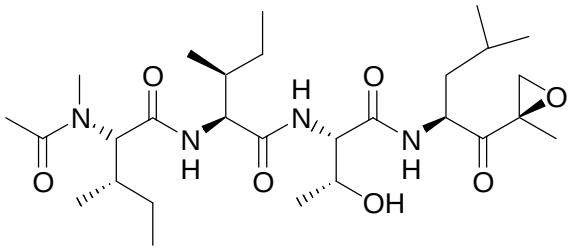
^{1}H NMR (400 MHz, $CDCl_3$): unit A δ = 7.47–7.02 (m, 5H, Ph), 6.97–6.90 (m, 1H, CH_2*CH*CH), 6.23 (d, *J* = 15.6 Hz, 1H, Ph*CH*), 6.03 (dd, *J* = 15.9, 8.4 Hz, 1H, PhCH*CH*), 5.81 (d, *J* = 8.4 Hz, 1H, *CH*CO), 5.76–5.69 (m, 1H, *CH*OCH_2), 3.13–2.93 (m, 2H, CHCH_2), 2.70–2.23 (m, 1H, *CH*Me), 1.41 (s, 9H, CMe_3), 1.06 (d, *J* = 7.0 Hz, 3H, Me); unit B δ = 7.65 (d, *J* = 7.6 Hz, 2H, Fmoc), 7.47–7.02 (m, 11H, Ph/Fmoc), 5.04–4.79 (m, 1H, NH), 4.52–4.26 (m, 2H, Fmoc), 4.13–3.85 (m, 2H, NH*CH*/Fmoc), 2.70–2.23 (m, 2H, CH_2Ph); unit C δ = 6.83–6.74 (m, 1H, NH), 3.69–3.57 (m, 1H, CH_2NH), 3.30–3.20 (m, 1H, CH_2NH), 2.70–2.23 (m, 1H, CO*CH*), 1.15 (d, *J* = 8.0 Hz, 3H, Me); unit D δ = 5.04–4.79 (m, 1H, NH), 1.76–1.43 (m, 3H, CH_2CH/*CH*Me), 0.72 (d, *J* = 6.3 Hz, 3H, Me), 0.67 (d, *J* = 6.3 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): unit A δ = 165.9 (CO), 142.0 (CH_2*CH*CH), 137.2 (C_{quart}), 136.6 (Ph*CH*), 133.7 (PhCH*CH*), 131.7 (C_{ortho}), 129.3 (C_{meta}), 127.0 (C_{para}), 126.0 (*CH*CO), 80.5 (*C*Me_3), 70.6 (*CH*OCH_2), 42.0 (*CH*Me), 34.8 (CHCH_2), 28.1 (CMe_3), 16.8 (Me); unit B δ = 171.0 (CO), 156.0 (CO, Fmoc), 144.0 (4, Fmoc), 141.1 (5, Fmoc), 131.7 (C_{ortho}), 129.8 (C_{quart}), 128.5 (7/7', Fmoc), 127.5 (8/8', Fmoc), 126.6 (9/9', Fmoc), 125.0 (C_{meta}) 119.8 (6/6', Fmoc), 67.0 (2, Fmoc), 56.5 (NH*CH*), 52.5 (3, Fmoc), 39.4 (CH_2Ph); unit C δ = 173.5 (CO), 47.1 (CH_2NH), 40.6 (CO*CH*), 14.5 (Me); unit D δ = 171.2 (CO), 40.4 (CO*CH*), 38.5 (CH_2CH), 24.6 (*CH*Me), 22.8 (Me), 21.2 (Me) ppm;

HRMS (FT-ICR): calcd. for $C_{53}H_{62}N_2O_9$ $[M + Na]^+$ 893.4347, found 893.4344.

2.3 Synthesis of Epoxomicin

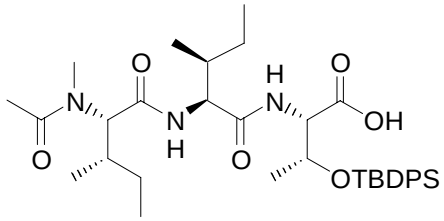
Epoxomicin (7)



To the solution of TBDPS-ether **212** (70 mg, 0.088 mmol) in dry THF (2 mL) TBAF (100 µl, 1 M solution in THF) was added at 0 °C and the mixture stirred for 1 h (0 °C to room temperature). The solvent was evaporated at room temperature and the residue was purified by flash chromatography (70% EtOAc in hexene) to generate **7** (34 mg, 75%) as a colorless solid product.

$\boldsymbol{R_f}$ = 0.34 (70% EtOAc in hexane);
$\boldsymbol{[\alpha]_D^{24}}$ = –62.4 (*c* = 0.36, MeOH), {ref[170] $[\alpha]_D^{24}$ = –66.1 (*c* = 0.50, MeOH)},
IR (neat): ν_{max} = 3270, 3079, 1675, 1540, cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.46 (d, *J* = 7.5 Hz, 1H, NH), 7.30 (d, *J* = 8.0 Hz, 1H, NH), 6.96 (d, *J* = 7.5 Hz, 1H, NH), 4.69 (d, *J* = 11.3 Hz, 1H, *CH*OH), 4.54–4.50 (m, 1H, N*CH*), 4.48 (dd, *J* = 7.7, 2.9 Hz, 1H, NH*CH*CH_2), 4.29–4.17 (m, 2H, NH*CH*), 3.29 (d, *J* = 5.0 Hz, 1H, *CH_2*epoxide), 2.97 (s, 3H, NMe) 2.86 (d, *J* = 5.0, 1H, *CH_2*epoxide), 2.10 (s, 3H, CO*Me*), 2.07–2.00 (m, 2H, 2NCH*CH*), 1.98–1.88 (m, 1H, *CH*Me_2), 1.75–1.57 (m, 2H, CH*CH_2*), 1.51 (s, 3H, *Me*epoxide), 1.37–1.18 (m, 4H, 2CH*CH_2*), 1.08 (s, 3H, *Me*CHOH), 0.91–0,70 (m, 18H, 6Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 207.0 (CO), 170.8 (CO), 170.4 (CO), 169.9 (*CO*Me), 169.4 (CO), 65.2 (CHOH), 61.3 (Cepoxide), 59.1 (NHCH), 57.9 (NCH), 56.5 (NHCH), 51.1 (CH_2epoxide), 49.2 (NHCH), 38.1 (*CH_2*$CHMe_2$), 35.0 (NHCH*CH*), 30.8 (NHCH*CH*), 30.7 (N*Me*), 23.8 (C*Me_2*), 23.4 (*CH_2*Me), 23.3 (*CH_2*Me), 22.0 (*CH_2*Me), 20.8 (CO*Me*), 19.8 (*Me*CHOH), 16.4 (C*Me_2*), 15.5 (*Me*), 14.2 (*Me*), 14.2 (Meepoxide), 9.7 ($MeCH_2$), 9.2 (*Me*CH_2) ppm;
HRMS (FT-ICR): calcd. for $C_{28}H_{50}N_4O_7$ $[M + Na]^+$ 577.7090, found 577.7088.

_N_-Acetyl-_N_-methyl-L-isoleucyl-L-isoleucyl-_O_-[_tert_-butyl(diphenyl)silyl]-L-threonine (183)



$PhSiH_3$ (0.14 mL, 1.15 mmol) was added dropwise at 0 °C to a of solution of allyl ester **204** (390 mg, 0.57 mmol) and $[Pd(Ph_3P)_4]$ (20 mg) in THF (3 mL). The resulting solution was stirred and maintained at 0 °C for 45 min. After that the reaction was quenched with H_2O (0.1 mL) and concentrated. The crude residue was purified by flash chromatography (hexane and Et_2O first 8:1, then 3:1, 1:1, 1:3 followed by pure Et_2O and pure EtOAc) to give the tripeptide derivative **183** (325 mg, 88% yield) as a colorless oil.

$\boldsymbol{R_f}$ = 0.17 (50% EtOAc in petroleum ether);

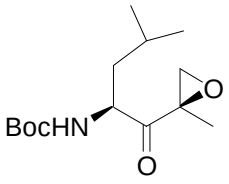
$\boldsymbol{[\alpha]_D^{24}}$ = –70.4 (*c* = 1.3, CH_2Cl_2);

IR (neat): ν_{max} = 3300, 3054, 2962, 1743, 1639, 1519 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.64–7.60 (m, 4H, Ph), 7.45–7.34 (m, 6H, Ph), 6.69–6.64 (m, 2H, 2NH), 4.67–4.38 (m, 4H, 2NH*CH*/*CH*OTBDPS/*CH*NMe), 2.94 (s, 3H, NMe), 2.11 (s, 3H, CO*Me*), 1.56–1.20 (m, 6H, 2*CH*Me/2*CH*$_2$Me), 1.00 (s, 9H, CMe_3), 0.98–0.82 (m, 15H, 5Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 172.3 (CO), 172.0 (CO), 171.2 (CO), 166.7 (CO), 135.8 (C_{quart}Ph), 132.0 (Ph) 129.8 (Ph), 128.5 (Ph), 127.7 (Ph), 119.1 (Ph), 69.6 (*CH*OTBDPS), 62.2 (NMe*CH*), 61.2 (NHCH), 60.3 (NHCH), 32.6 (*CH*Me), 31.4 (*CH*Me), 28.9 (CO*Me*), 27.8 (N*Me*), 26.9 (SiC*Me*$_3$), 24.6 (*CH*$_2$Me), 24.5 (*CH*$_2$Me), 21.9 (*Me*), 19.3 (Si*C*Me$_3$), 15.5 (*Me*), 14.1 (*Me*), 11.2 (Me), 10.3 (Me) ppm.

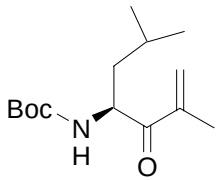
***tert*-Butyl (1*S*)-3-methyl-1-{[(2*R*)-2-methyloxiran-2-yl]carbonyl}butylcarbamate (184)**



To a solution of unsaturated ketone **198** (500 mg, 1.9 mmol) in MeOH (5 mL) benzonitril (235 mg, 2.28 mmol) and *i*Pr_2NEt (328 mg, 2.28 mmol) were added at 0 °C. Then 30% H_2O_2 (1.21 g, 19 mmol) was added and the reaction mixture was stirred for 40 h at 0 to 4 °C. After that the reaction mixture was diluted with ether (100 mL) and H_2O (25 mL). The aqueous layer was separated and extracted with CH_2Cl_2 (100 mL). The combined organic layers were washed with brine (100 mL), dried with $MgSO_4$, filtered and concentrated. The crude residue was purified and separated from **184a** by flash chromatography (10% then 5% EtOAc in petroleum ether) to provide pure epoxide **184** (6.5 g, 56%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.21 (10% EtOAc in petroleum ether);
$\boldsymbol{[\alpha]_D^{24}}$ = 134.0 (*c* = 0.23, CH_2Cl_2);
IR (neat): ν_{max} = 3374, 2965, 1708, 1508, 1369 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 4.85 (d, *J* = 8.3 Hz, 1H, NH), 4.30 (t, *J* = 9.6 Hz, NH*CH*), 3.25 (d, *J* = 5.0 Hz, 1H, *CH_2*–epoxide), 2.85 (d, *J* = 5.0 Hz, 1H, *CH_2*–epoxide), 1.74–1.64 (m, 1H, *CH*Me_2), 1.48 (s, 3H, Me–epoxide), 1.37 (m, 9H, C*Me_3*), 1.18–1.11 (m, 2H, CH*CH_2*), 0.93 (d, *J* = 6.5 Hz, 3H, Me), 0.89 (d, *J* = 6.5 Hz, 3H, Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 209.5 (*CO*CH=CH_2), 155.5 (CO), 79.6 (*C*Me_3), 58.9 (C_{quart}, epoxide), 52,2 (CH_2, epoxide), 51.3 (NH*CH*), 40.3 (CH_2), 28.2 (C*Me_3*), 25.0 (*CH*Me_2), 23.3 (Me), 21.2 (Me), 16.7 (*Me*CH=CH_2) ppm.

tert-Butyl(1*S*)-1-isobutyl-2,3-dioxobutylcarbamate (198)



2-Bromopropene (0.87 g, 7.3 mmol) was taken in THF (5 mL) and *t*-BuLi (1.5 M, 9.73 mL, 14.6 mmol) was added at –78 °C. The reaction mixture was stirred for 1 h and Weinreb amide **206** (500 mg, 1.82 mmol) in THF (1 mL) was added. After stirring 3 h at –78 °C, the reaction was quenched with NH_4Cl solution (10 mL) and extracted with ether (2 x 50 mL). The combined organic layers were dried with $MgSO_4$, filtered and concentrated in vacuo. The crude residue was purified by flash chromatography (20% EtOAc in petroleum ether) to give pure enone **198** (435 mg, 93%) as a dark red colored oil.

$\boldsymbol{R_f}$ = 0.43 (20% EtOAc in petroleum ether);

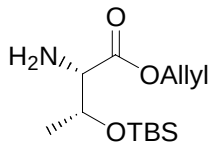
$\boldsymbol{[\alpha]_D^{24}}$ = 52.3 (*c* = 0.30, CH_2Cl_2);

IR (neat): ν_{max} = 3351, 2969, 2094, 1708, 1660, 1508 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 6.03 (s, 1H, CH=*CH*$_2$), 5.83 (s, 1H, CH=*CH*$_2$), 5.59 (d, *J* = 8.5 Hz, 1H, NH), 5.02 (t, *J* = 9.6 Hz, NH*CH*), 1.85 (s, 3H, CH_2=CH*Me*), 1.73–1.64 (m, 1H, *CH*Me_2), 1.38 (m, 9H, C*Me*$_3$), 1.35–1.21 (m, 2H, CH*CH*$_2$), 0.95 (d, *J* = 6.5 Hz, 3H, Me), 0.85 (d, *J* = 6.5 Hz, 3H, Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 201.5 (*CO*CH=CH_2), 155.4 (CO), 142.2 (*CH*=CH_2), 125.9 (CH=*CH*$_2$), 79.4 (*C*Me_3), 52.4 (NH*CH*), 43.0 (CH_2), 28.2 (C*Me*$_3$), 24.8 (*CH*Me_2), 23.3 (Me), 21.6 (Me), 17.7 (*Me*CH=CH_2) ppm.

Allyl-*O*-[*tert*-butyl(diphenyl)silyl]-L-threoninate (199)



A mixture of allyl alcohol (29 mL, 420 mmol), L-threonine (5.0 g, 42 mmol), *p*-TsOH (9.6 g, 50 mmol) and C_6H_6 (150 mL) was refluxed for 45 h using a Dean-Stark trap and then concentrated. TBDPS-Cl (17.31 g, 63 mmol) was added to a 0 °C mixture of the crude residue, imidazole (15 g, 210 mmol) and CH_2Cl_2 (100 mL). The mixture was allowed to come to room temperature overnight and concentrated. The residue was dissolved in EtOAc (300 mL), washed with 1 M NaOH (3 x 50 mL) and brine (2 x 50 mL), dried with $NaSO_4$, filtered and concentrated. The residue was purified by flash chromatography (30% then 50% EtOAc in petroleum ether) to give pure amine **199** (6.5 g, 56%) as a light yellow oil.

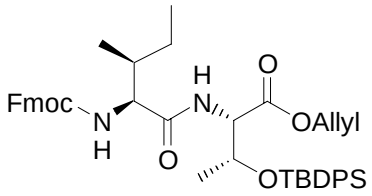
$\boldsymbol{R_f}$ = 0.20 (50% EtOAc in petroleum ether);

IR (neat): ν_{max} = 3300, 2935, 1430, 1369, 1160 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.70–7.60 (m, 4H, Ph), 7.44–7.34 (m, 6H, Ph), 5.87–5.76 (m, 1H, *CH*=CH_2), 5.27–5.20 (m, 2H, CH=*CH*$_2$), 4.59 (dd, *J* = 13.0, 5.9 Hz, 1H, CH_2, allyl), 4.39 (dd, *J* = 13.0, 5.9 Hz, 1H, CH_2, allyl), 4.37–4.33 (m, 1H, *CH*OTBDPS), 3.39 (d, *J* = 2.5 Hz, 1H, NH_2*CH*), 1.73 (m, 2H, NH_2), 1.12 (d, *J* = 6.3 Hz, 3H, Me), 1.00 (s, 9H, CMe_3) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 174.2 (CO), 135.8 (C_{quart}–Ph), 134.0 (*CH*=CH_2), 133.2 (Ph), 131.8 (Ph), 129.6 (Ph), 127.5 (Ph), 118.7 (Ph), 70.8 (NHCH), 65.7 (*CH*OTBDPS), 60.5 (CH_2, allyl), 26.7 (SiC*Me*$_3$), 20.5 (Me), 19.2 (Si*C*Me$_3$) ppm.

Allyl (2*S*,3*R*)-3-{[*tert*-butyl(diphenyl)silyl]oxy}-2-[((2*S*,3*S*)-2-{[(9*H*-fluoren-9-ylmethoxy)carbonyl]amino}-3-methylpentanoyl)amino]butanoate (200)



Compound **199** (400 mg, 1 mmol), Fmoc protected amino acid **178** (356 mg, 1 mmol), TBTU (375 mg, 1 mmol) and HOBT (136 mg, 1 mmol) were taken in CH_2Cl_2 (5 mL). The reaction mixture was allowed to warm to room temperature overnight and concentrated. The crude residue was then taken up in EtOAc (20 mL), dried with $MgSO_4$ and concentrated. The crude product was purified by flash chromatography (30% EtOAc in petroleum ether) to furnish dipeptide **200** (315 mg, 85%) as a colorless thick oil.

***R*$_f$** = 0.37 (30% EtOAc in petroleum ether);

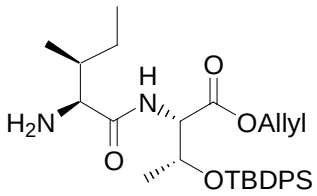
[α]$_D^{24}$ = –2.9 (*c* = 1.0, CH_2Cl_2);

IR (neat): ν_{max} = 3430, 3313, 2958, 1731, 1673, 1519, 1376, 1241 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.71 (d, *J* = 7.5 Hz, 2H, Fmoc), 7.60–7.52 (m, 6H, Fmoc), 7.38–7.20 (m, 10H, Ph), 6.48 (d, *J* = 8.8 Hz, NH), 5.80–5.70 (m, 1H, *CH*=CH_2), 5.50 (d, *J* = 8.3 Hz, 1H, NH), 5.25–5.16 (m, 2H, CH=*CH*$_2$), 4.56–4.46 (m, 3H, CH_2, allyl/*CH*OTBDPS), 4.41–4.35 (m, 4H, 2NH*CH*/Fmoc), 4.18 (t, *J* = 6.9 Hz, 1H, Fmoc), 1.92–1.87 (m, 1H, *CH*Me), 1.66–1.55 (m, 1H, *CH*$_2$Me), 1.33–1.21 (m, 1H, *CH*$_2$Me), 1.00–0.90 (m, 18H, CMe_3/ 3Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.2 (CO), 169.0 (CO), 156.0 (CO, Fmoc), 143.9 (Fmoc), 141.2 (Fmoc), 135.8 (C_{quart}–Ph), 133.4 (*CH*=CH_2), 132.5 (Ph), 131.3 (Ph), 129.9 (Ph), 129.8 (Fmoc), 127.5 (Ph), 127.0 (Fmoc), 125.0 (Fmoc), 119.9 (Fmoc), 118.9 (Ph), 69.9 (*CH*OTBDPS), 67.0 (NHCH), 66.2 (NHCH), 59.4. (Fmoc), 57,8 (*CH*$_2$allyl), 47.1 (Fmoc), 37.9 (*CH*Me), 26.7 (SiC*Me*$_3$), 25.1 (*CH*$_2$Me), 20.9 (*Me*), 19.1 (Si*C*Me$_3$), 15.0 (*Me*), 11.6 (*Me*) ppm.

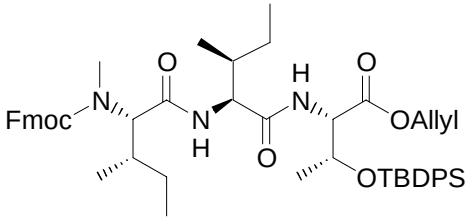
Allyl-L-isoleucyl-*O*-[*tert*-butyl(diphenyl)silyl]-L-threoninate (201)



Piperidine (0.078 mL, 0.80 mmol) was added to a 0 °C solution of the Fmoc-tripeptide **200** (300 mg, 0.40 mmol) and CH_3CN (5 mL). The resulting mixture was allowed to warm to room temperature over 3 h and then concentrated. The crude was purified by chromatography (hexane / Et_2O first 8:1, then 3:1, 1:1 and 1:3 followed by pure Et_2O and pure EtOAc) to give dipeptide amine **201** (178 mg, 85%) as a colorless oil.

***R*$_f$** = 0.19 (50% EtOAc in petroleum ether);
IR (neat): ν_{max} = 3374, 3066, 2958, 1743, 1673, 1504, 1376, 1311, 1103 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.18 (d, *J* = 9.6 Hz, 1H, NH), 7.62–7.52 (m, 5H, NH/Ph), 7.44–7.34 (m, 6H, Ph), 5.85–5.75 (m, 1H, *CH*=CH_2), 5.25 (dd, *J* = 17.1, 1.2 Hz, 1H, CH=*CH*$_2$), 5.20 (dd, *J* = 10.4, 1.0 Hz, 1H, CH=*CH*$_2$), 4.60–4.51 (m, 3H, *CH*$_2$allyl/ *CH*OTBDPS), 4.41 (dd, *J* = 13.1, 5.8 Hz, 1H, NH*CH*), 3.44 (d, *J* = 3.9 Hz, 1H, *CH*NH_2), 2.06–1.98 (m, 1H, *CH*Me), 1.54–1.44 (m, 1H, *CH*$_2$Me), 1.27–1.16 (m, 1H, *CH*$_2$Me), 1.04–0.91 (m, 18H, CMe_3/3Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 174.7 (CO), 170.4 (CO), 143.9 (Fmoc), 135.7 (C_{quart}Ph), 133.8 (*CH*=CH_2), 132.8 (Ph), 131.5 (Ph), 129.8 (Ph), 127.6 (Ph),118.7 (Ph), 70.2 (*CH*OTBDPS), 65.9 (NHCH), 60.0 (NH_2CH), 57,5 (CH_2allyl), 37.9 (*CH*Me), 26.7 (SiC*Me*$_3$), 23.9 (*CH*$_2$Me), 21.0 (Me), 19.1 (Si*C*Me_3), 10.0 (Me), 11.9 (Me) ppm.

Allyl-*N*-[(9*H*-fluoren-9-ylmethoxy)carbonyl]-*N*-methyl-L-isoleucyl-L-isoleucyl-*O*-[*tert*-butyl(diphenyl)silyl]-L-threoninate (202)



Amide **201** (400 mg, 0.78 mmol), Fmoc protected acid **181** (288 mg, 0.78 mmol) and DMAP (100 mg, 0.78 mmol) were taken in CH_2Cl_2 (5 mL). PyBrop (363 mg, 0.78 mmol) was added and the reaction mixture was stirred for 20 h at room temperature. The resulting mixture was concentrated and the residue was purified by flash chromatography (10% then 30% EtOAc in petroleum ether) to give the fully protected tripeptide **202** (612 mg, 91%) as a light yellow oil.

$\boldsymbol{R_f}$ = 0.33 (30% EtOAc in petroleum ether);

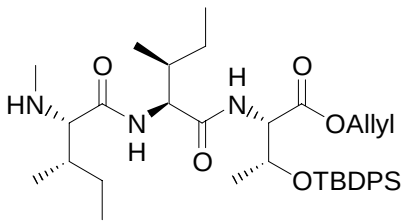
$\boldsymbol{[\alpha]_D^{24}}$ = –55 (*c* = 0.41, CH_2Cl_2);

IR (neat): ν_{max} = 3324, 2961, 1673, 1515, 1307, 1241, 1160 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76 (d, *J* = 7.3 Hz, 2H, Fmoc), 7.64 (d, *J* = 7.3 Hz, 2H, Fmoc), 7.61–7.56 (m, 4H, Fmoc), 7.45–7.29 (m, 10H, Ph), 6.61 (d, *J* = 9.1 Hz, 2H, 2NH), 5.85–5.74 (m, 1H, *CH*=CH_2), 5.21 (d, *J* = 9.6 Hz, 1H, CH=*CH_2*), 5.20 (d, *J* = 9.6 Hz, 1H, CH=*CH_2*), 4.59–4.37 (m, 7H, *CH_2*allyl/*CH*OTBDPS/*CH*NMe/Fmoc), 4.26 (d, *J* = 8.0 Hz, 2H, 2NH*CH*), 2.91 (s, 3H, NMe), 2.20–2.00 (m, 1H, *CH*Me), 1.98–1.94 (m, 1H, *CH*Me), 1.54–1.39 (m, 2H, *CH_2*Me), 1.27–1.18 (m, 2H, *CH_2*Me), 1.01 (s, 9H, CMe_3), 0.98–0.80 (m, 15H, 5Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.0 (CO), 169.9 (CO), 169.8 (CO), 157.3 (CO, Fmoc), 143.8 (Fmoc), 141.3 (Fmoc), 135.8 (C_{quart}Ph), 133.5 (*CH*=CH_2), 132.5 (Ph), 131.3 (Ph), 129.9 (Ph), 129.9 (Fmoc), 127.6 (Ph), 127.5 (Fmoc), 124.8 (Fmoc), 120.0 (Fmoc), 119.1 (Ph), 69.9 (*CH*OTBDPS), 67.9 (NHCH), 66.2 (NHCH), 63.4 (NMe*CH*), 57.7 (Fmoc), 57.4 (*CH_2*allyl), 47.1 (Fmoc), 36.9 (*CH*Me), 31.5 (*CH*Me), 29.8 (N*Me*), 26.8 (SiC*Me_3*), 24.7 (*CH_2*Me), 24.4 (*CH_2*Me), 20.9 (*Me*), 19.1 (Si*C*Me_3), 15.6 (*Me*), 15.0 (*Me*), 11.6 (Me), 10.4 (Me) ppm.

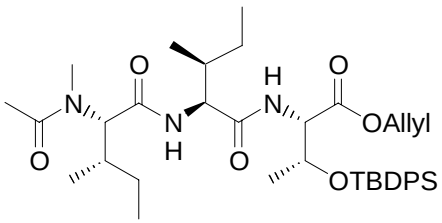
Allyl-*N*-methyl-L-isoleucyl-L-isoleucyl-*O*-[*tert*-butyl(diphenyl)silyl]-L-threoninate (203)



Piperidine (0.09 mL, 0.92 mmol) was added to a 0 °C solution of the Fmoc-tripeptide **202** (400 mg, 0.46 mmol) and CH_3CN (5 mL). The resulting mixture was allowed to warm to room temperature over 3 h and then concentrated. The crude was purified by chromatography (hexane / Et_2O first 8:1, then 3:1, 1:1, and 1:3 followed by pure Et_2O and pure EtOAc) to produce the tripeptide derivative amine **203** (285 mg, 96%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.18 (50% EtOAc in petroleum ether);
IR (neat): ν_{max} = 3320, 3058, 2962, 2873, 2680, 1747, 1658, 1515, 1307, 1160 cm^{-1};
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76 (d, *J* = 8.5 Hz, 1H, NH), 7.64–7.59 (m, 4H, Ph), 7.44–7.34 (m, 6H, Ph), 6.61 (d, *J* = 8.8 Hz, 1H, NH), 5.83–5.73 (m, 1H, *CH*=CH_2), 5.25 (d, *J* = 9.0 Hz, 1H, CH=*CH_2*), 5.20 (d, *J* = 9.0 Hz, 1H, CH=*CH_2*), 4.58–4.37 (m, 6H, *CH_2*allyl/*CH*OTBDPS/*CH*NMe/2*CH*NH), 3.11 (s, *NH*Me), 2.47 (s, 3H, NMe), 1.98–1.92 (m, 2H, 2*CH*Me), 1.68–1.53 (m, 2H, *CH_2*Me), 1.38–1.21 (m, 2H, *CH_2*Me), 1.00 (s, 9H, CMe_3), 0.98–0.80 (m, 15H, 5Me) ppm;
^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.0 (CO), 169.9 (CO), 169.8 (CO), 135.8 (C_{quart}Ph), 133.5 (*CH*=CH_2), 132.5 (Ph), 131.4 (Ph), 129.8 (Ph), 127.5 (Ph), 119.1 (Ph), 69.9 (*CH*OTBDPS), 69.4 (NHCH), 66.2 (NHCH), 63.4 (NMe*CH*), 57.8 (*CH_2*allyl), 37.2 (*CH*Me), 35.1 (*CH*Me), 29.8 (N*Me*), 26.8 (SiC*Me_3*), 25.4 (*CH_2*Me), 24.2 (*CH_2*Me), 20.7 (*Me*), 19.1 (Si*C*Me_3), 15.5 (*Me*), 15.3 (*Me*), 11.6 (Me), 11.4 (Me), ppm;

Allyl-*N*-acetyl-*N*-methyl-L-isoleucyl-L-isoleucyl-*O*-[*tert*-butyl(diphenyl)silyl]-L-threoninate (204)



Compound **203** (200 mg, 0.31 mmol) was taken in CH_2Cl_2 (2 mL) and diisopropylethyl amine (0.072 mL, 0.63 mmol) was added. Then Ac_2O (0.075 mg, 0.63 mmol) was added dropwise at 0 °C and the reaction mixture was stirred at room temperature for 4 h. After evaporation of the solvent in vacuo the residue was purified by flash chromatography (30% EtOAc in petroleum ether) to provide acetamide **204** (201 mg, 94%) as a slightly yellow oil.

$\boldsymbol{R_f}$ = 0.24 (30% EtOAc in petroleum ether);

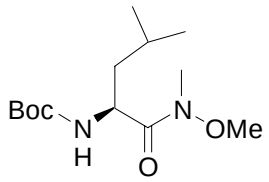
$\boldsymbol{[\alpha]_D^{24}}$ = –73.4 (*c* = 1.4, CH_2Cl_2);

IR (neat): ν_{max} = 3436, 3301, 3058, 2962, 2545, 1743, 1639, 1515, 1307, 1160 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.60 (dd, *J* = 19.3, 7.2 Hz, 4H, Ph), 7.42–7.31 (m, 6H, Ph), 6.74 (d, *J* = 8.3 Hz, 1H, NH), 6.65 (d, *J* = 9.1 Hz, 1H, NH), 5.82–5.71 (m, 1H, *CH*=CH_2), 5.20 (d, *J* = 9.3 Hz, 1H, CH=*CH_2*), 5.18 (d, *J* = 9.6 Hz, 1H, CH=*CH_2*), 4.67–4.45 (m, 4H, *CH_2*allyl/*CH*OTBDPS/*CH*NMe), 4.36 (d, *J* = 8.0 Hz, 2H, 2NH*CH*), 2.94 (s, 3H, NMe), 2.09 (s, 3H, CO*Me*), 2.00–1.97 (m, 2H, 2*CH*Me), 1.52–1.43 (m, 1H, *CH_2*Me), 1.35–1.28 (m, 1H, *CH_2*Me), 1.25–1.12 (m, 2H, *CH_2*Me), 0.99 (s, 9H, CMe_3), 0.91–0.82 (m, 15H, 5Me) ppm;

^{13}C NMR (100 MHz, $CDCl_3$): δ = 171.0 (CO), 171.0 (CO), 170.0 (CO), 169.8 (CO), 135.8 (C_{quart}–Ph), 133.5 (*CH*=CH_2), 132.5 (Ph), 131.3 (Ph), 129.6 (Ph), 127.5 (Ph), 118.9 (Ph), 69.9 (*CH*OTBDPS), 66.0 (NMe*CH*), 60.7 (NHCH), 60.2 (NHCH), 57.6 (*CH_2*allyl), 36.4 (*CH*Me), 31.4 (*CH*Me), 31.3 (CO*Me*), 29.8 (N*Me*), 26.7 (SiC*Me_3*), 24.5 (*CH_2*Me), 24.4 (*CH_2*Me), 20.6 (*Me*), 19.1 (Si*C*Me_3), 15.5 (*Me*), 15.2 (*Me*), 11.2 (Me), 10.3 (Me) ppm;

N^2-(*tert*-Butoxycarbonyl)-*N^1*-methoxy-*N^1*-methyl-L-leucinamide (206)



Triethylamine (0.43 g, 4.3 mmol) was added to a stirred solution of Boc-amino acid **205** (1.0 g, 4.3 mmol) in CH_2Cl_2 (10mL). Then BOP (1.9 g, 4.3 mmol) was added, followed after a few minutes by O,N-dimethylhydroxylamine hydrochloride (0.45 g, 4.4 mmol) and Et_3N (1 mL). The reaction was monitored by TLC and pH paper (due to the pH smaller than 7 neutralization by adding a few drops of Et_3N enabled a complete reaction). The mixture was then diluted with CH_2Cl_2 (100 mL) and washed successively with 3 N HCl (3 x 20 mL) and saturated NaCl solution (3 x 20 mL). The resulting organic layer was dried with $MgSO_4$, filtered and concentrated in vacuo. The crude residue was purified by flash chromatography (30% EtOAc in petroleum ether) to give pure amide **206** (105 mg, 88%) as a colorless oil.[181]

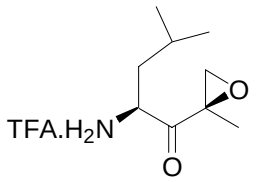
$\boldsymbol{R_f}$ = 0.28 (30% EtOAc in petroleum ether);

IR (neat): ν_{max} = 3325, 2935, 1710, 1660, 1500 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 5.03 (d, *J* = 9.1 Hz, 1H, NH), 4.68 (d, *J* = 3.2 Hz, 1H, NH*CH*), 3.75 (s, 3H, OMe), 3.16 (s, 3H, NMe), 1.73–1.63 (m, 1H, *CH*Me_2), 1.38–1.35 (m, 11H, *CH_2*/C*Me_3*), 0.93 (d, *J* = 6.5 Hz, 3H, Me), 0.89 (d, *J* = 6.5 Hz, 3H, Me) ppm;

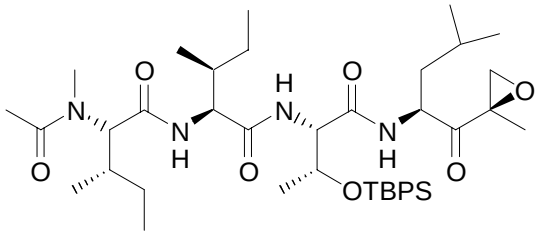
^{13}C NMR (100 MHz, $CDCl_3$): δ = 173.8 (*CO*NMe), 155.5 (CO), 79.1 (*C*Me_3), 61.5 (OMe), 48.9 (NH*CH*), 42.0 (CH_2), 32.0 (NMe), 28.2 (C*Me_3*), 24.6 (*CH*Me_2), 23.2 (Me), 21.5 (Me) ppm.

(2*S*)-2-amino-4-methyl-1-(2-methyloxiran-2-yl)pentan-1-one trifluoroacetate (211)



Compound **184** (100 mg, 0.36 mmol) was taken in CH_2Cl_2 (3 mL) and TFA was added dropwise at 0 °C. The reaction mixture was stirred 1.5 h (TLC controlled) and concentrated. The crude residue was azeotroped twice with toluene and the solvent evaporated. This residue of **211** was used in the next step without further purification.

***N*-acetyl-*N*-methyl-L-isoleucyl-L-isoleucyl-*O*-[tert butyl(diphenyl)silyl]-*N*^{1}-((1*S*)-3-methyl-1-{[(2*R*)-2-methyloxiran-2-yl]carbonyl}butyl)-L-threoninamide (212)**



Compound **183** (233 mg, 0.36 mmol), salt of amine **211** (100 mg, 0.36 mmol), TBTU (135 mg, 0.36 mmol) and HOBT (50 mg, 0.36 mmol) were taken in CH_2Cl_2 and DMF (3:2, 5mL). Diisopropylethylamine was added dropwise at 0 °C and the reaction mixture was stirred at room temperature for 20 h. After dilution with CH_2Cl_2 (15 mL) it was washed successively with H_2O (10 mL), $NaHCO_3$ solution (10mL) and brine (10mL). Then the

organic layer was dried with $MgSO_4$, filtered and concentrated in vacuo. The crude residue was purified (30% EtOAc in petroleum ether) and produced compound **212** (153 mg, 51%) as a colorless oil.

$\boldsymbol{R_f}$ = 0.34 (30% EtOAc in petroleum ether);

$\boldsymbol{[\alpha]_D^{24}}$ = –39.0 (*c* = 0.41, CH_2Cl_2);

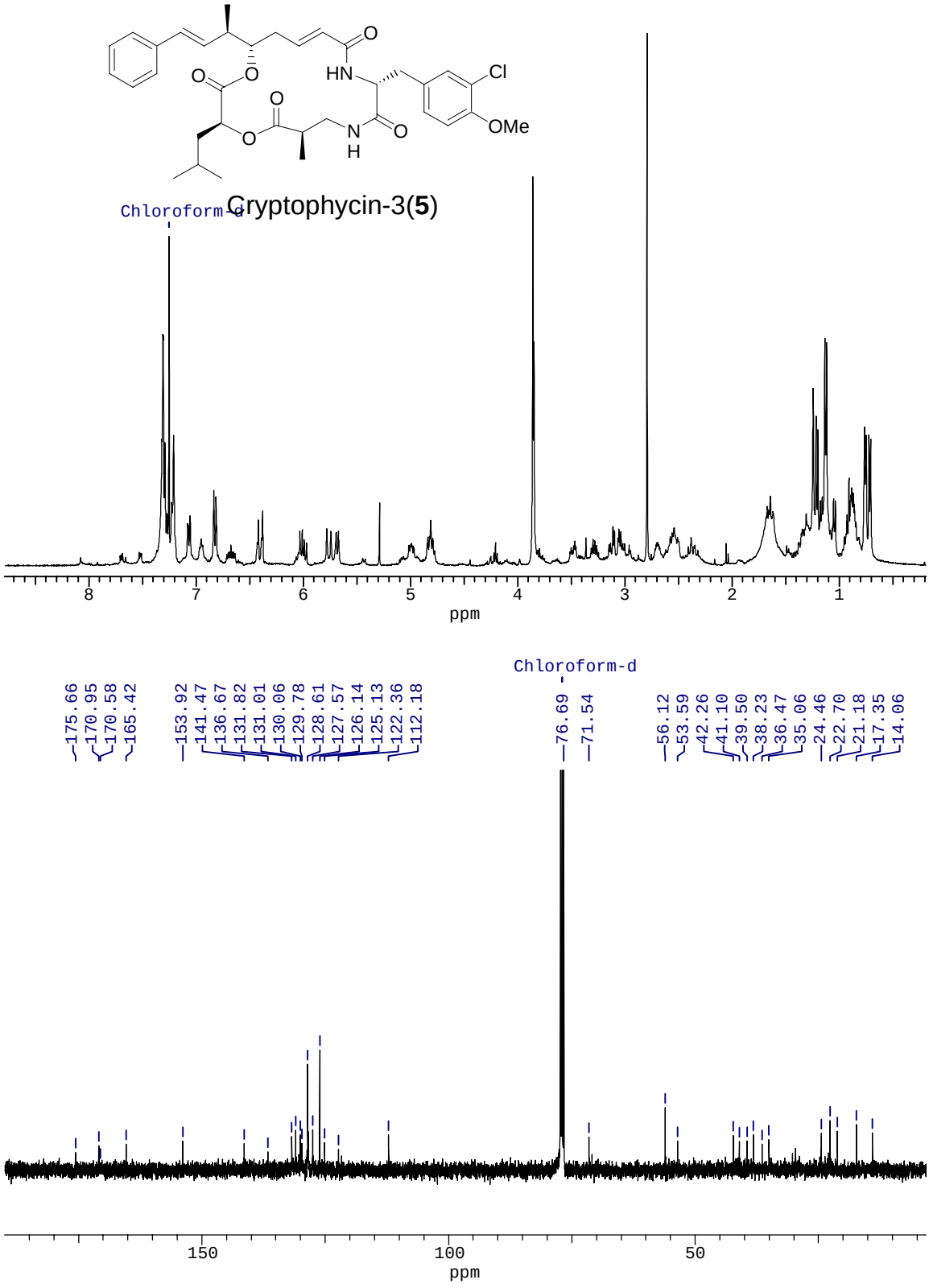
IR (neat): ν_{max} = 3294, 3080, 1636, 1540 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.76–7.71 (m, 4H, Ph), 7.43–7.35 (m, 6H, Ph), 7.25 (d, *J* = 6.3 Hz, 1H, NH), 6.80 (d, *J* = 6.3 Hz, 1H, NH), 6.53 (d, *J* = 6.8 Hz, 1H, NH), 4.58–4.50 (m, 3H, 3NH*CH*), 4.34–4.28 (m, 1H, NMe*CH*), 4.19–4.15 (m, 1H, *CH*OTBDPS), 3.39 (d, *J* = 5.0 Hz, 1H, CH_2epoxide), 2.95–2.90 (m, 1H, CH_2epoxide), 2.89 (s, 3H, NMe), 2.07 (s, 3H, CO*Me*), 1.97–1.87 (m, 1H, *CH*Me_2), 1.71–1.65 (m, 2H, 2*CH*Me), 1.53 (s, 3H, *Me*epoxide), 1.36–1.18 (m, 6H, CHCH_2), 1.10 (s, 9H, CMe_3), 0.91–0,70 (m, 21 H, 7Me) ppm;

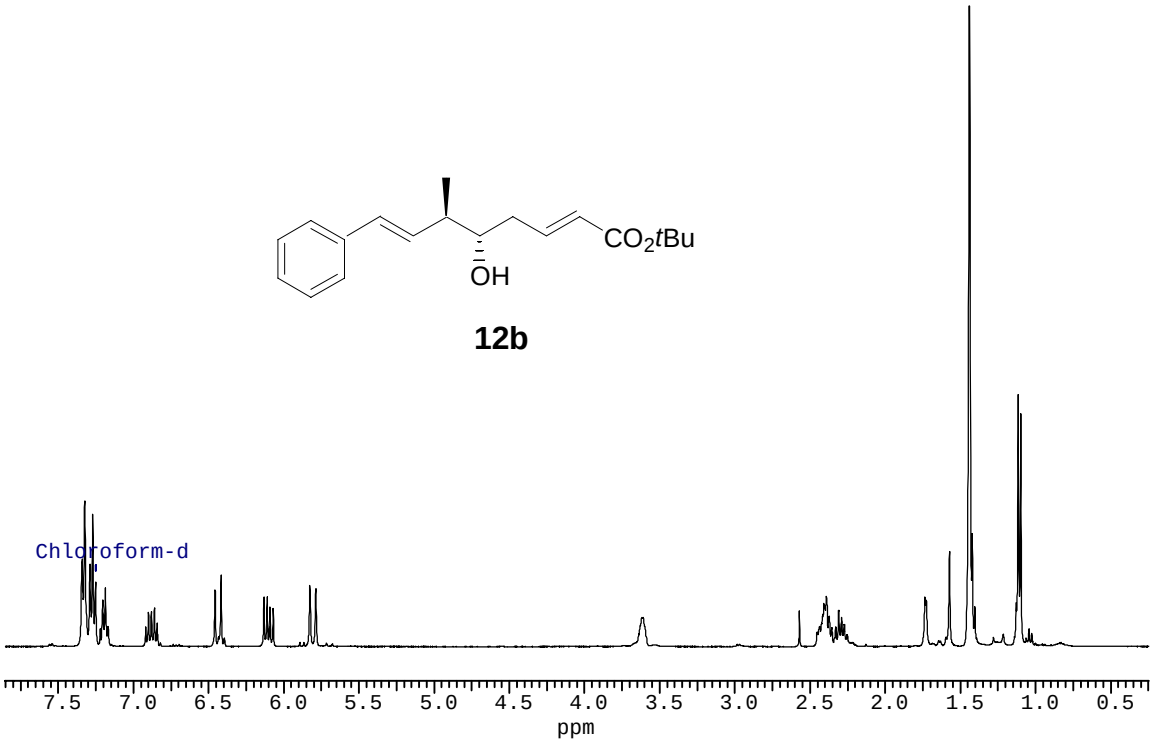
^{13}C NMR (100 MHz, $CDCl_3$): δ = 208.3 (CO), 172.1 (CO), 170.4 (CO), 169.6 (*CO*Me), 135.8 (C_{quart}Ph), 133.3 (Ph), 132.1 (Ph), 130.1 (Ph), 127.7 (Ph), 119 (Ph), 69.1 (*C*HOTBDPS), 59.3 (C_{quart}epoxide), 57.9 (NMeCH), 56.9 (NHCH), 52.4 (NHCH), 50.6 (NHCH), 44.4 (CH_2epoxide), 39.5 (CH_2CHMe_2), 36.2 (*CH*Me), 33.8 (*CH*Me), 31.3 (*CH*Me_2), 27.0 (CMe_3), 25.1 (*CH*Me_2), 24.3 (CH_2Me), 23.2 (CH_2Me), 22.0 (CO*Me*), 21.1 (*Me*CHOTBDPS), 19.2 (CMe_3), 16.7 (Meepoxide), 15.7 (*Me*), 15.4 (*Me*), 11.2 (*Me*), 10.3 (*Me*) ppm.

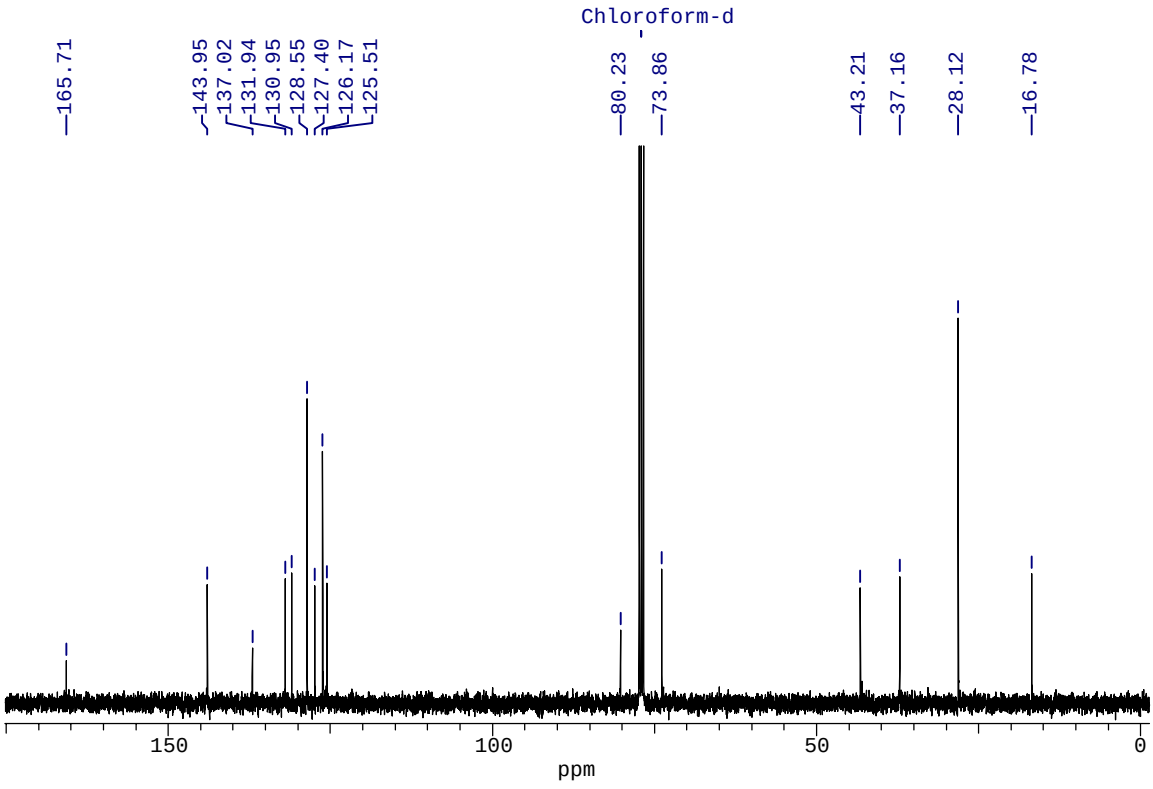
VI. APPENDIX

1. NMR-Spectra for important compounds

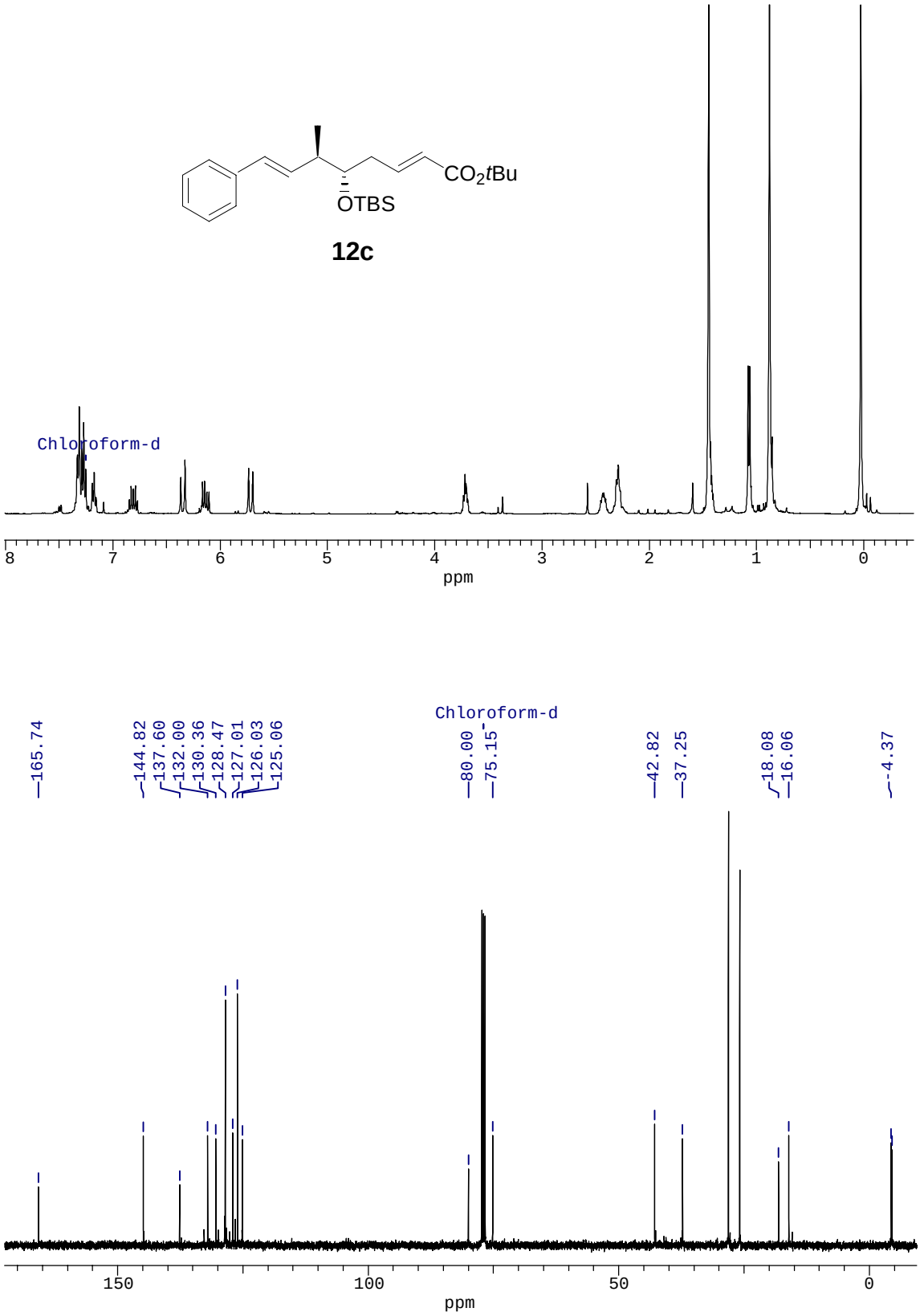


^{1}H- and ^{13}C-NMR spectra of compound **5**

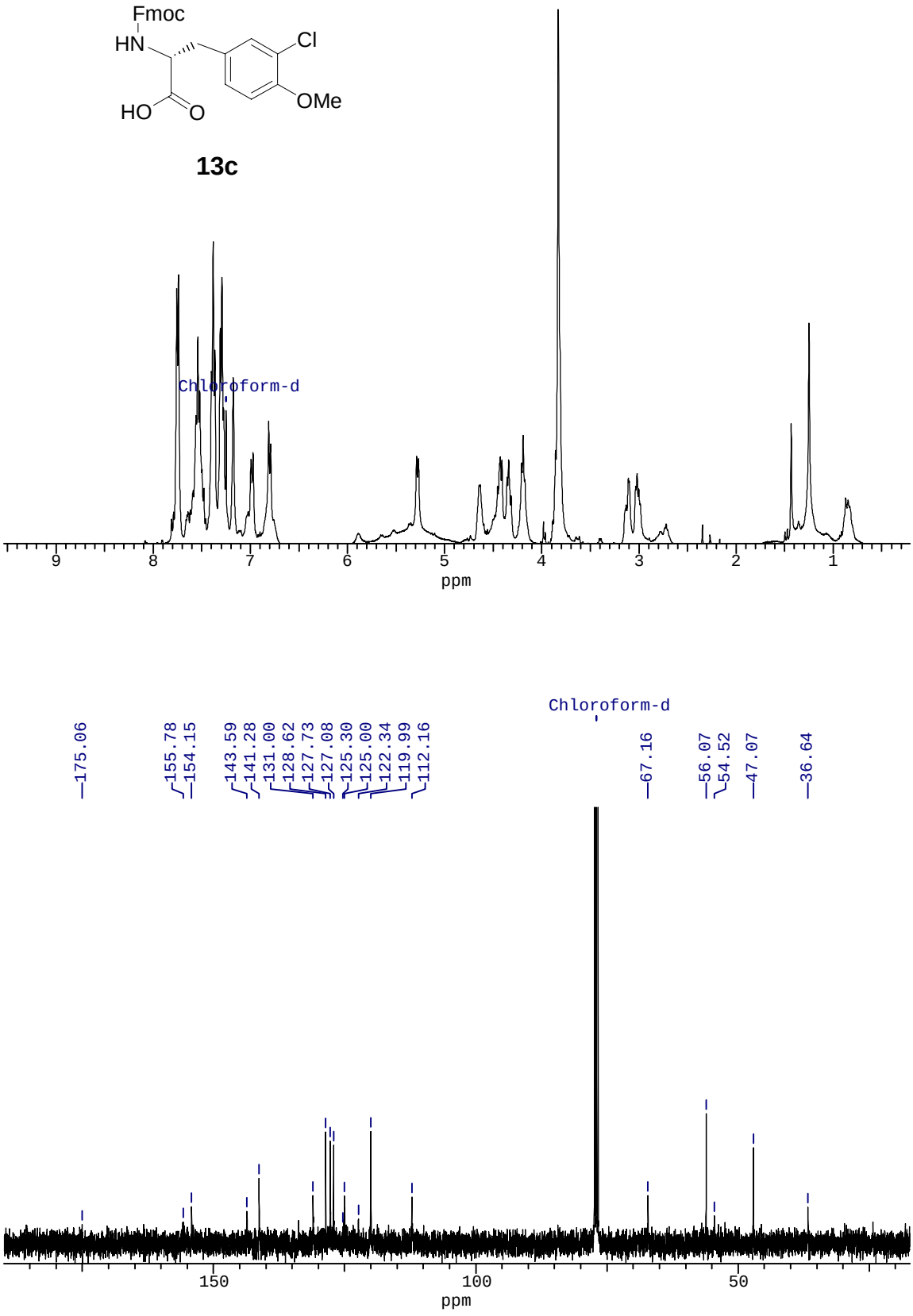




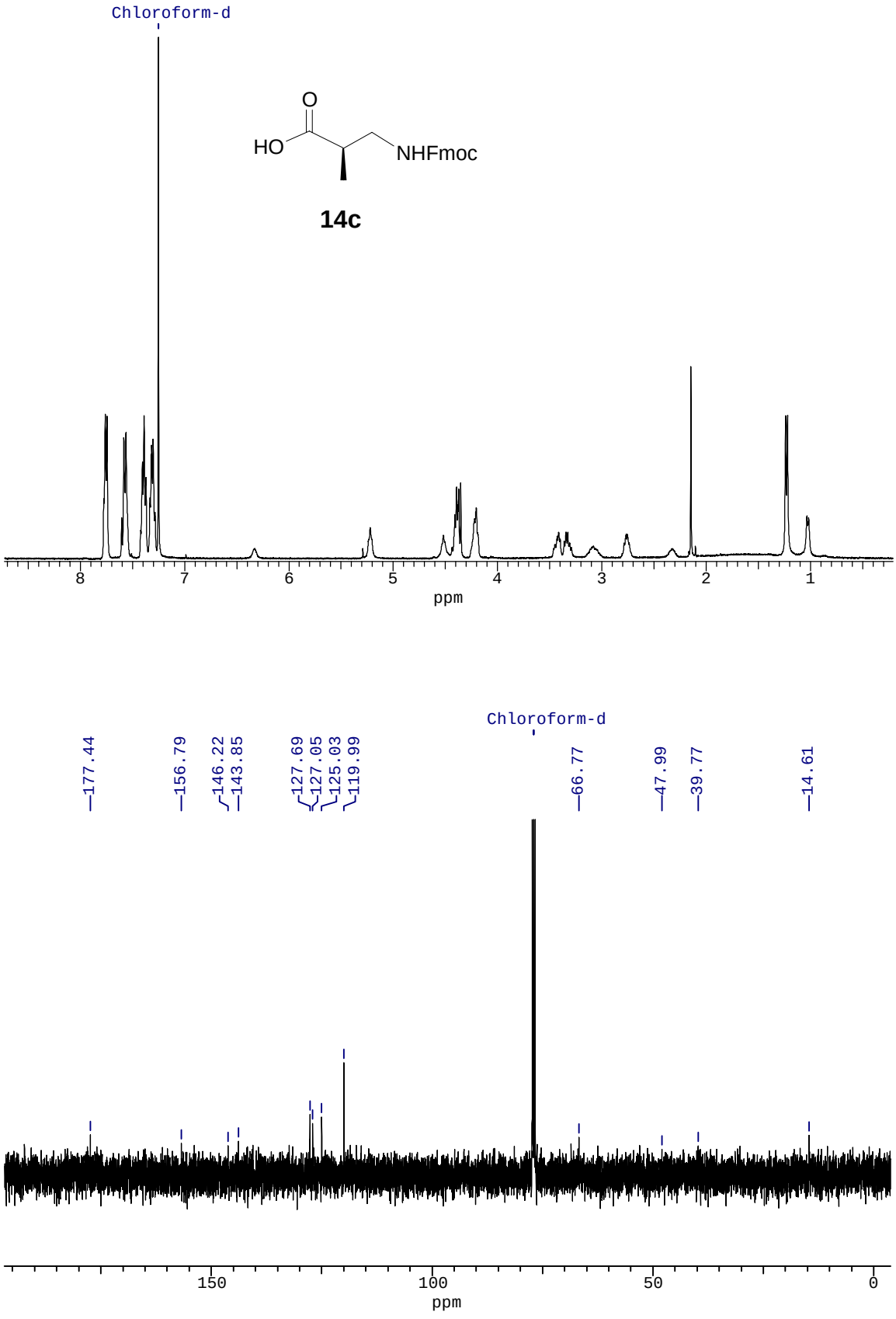
^{1}H- and ^{13}C-NMR spectra of compound **12b**



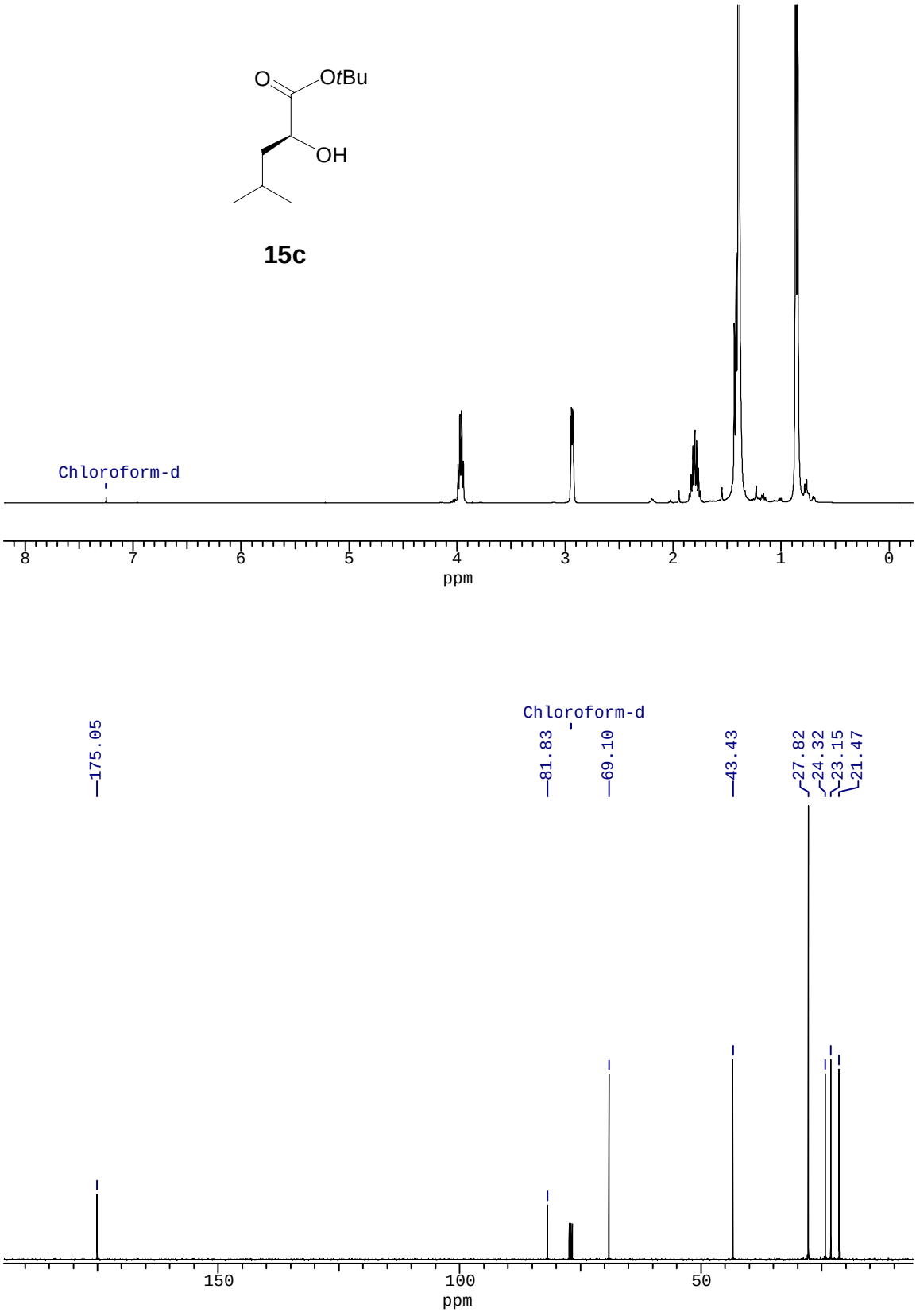
^{1}H- and ^{13}C-NMR spectra of compound **12c**



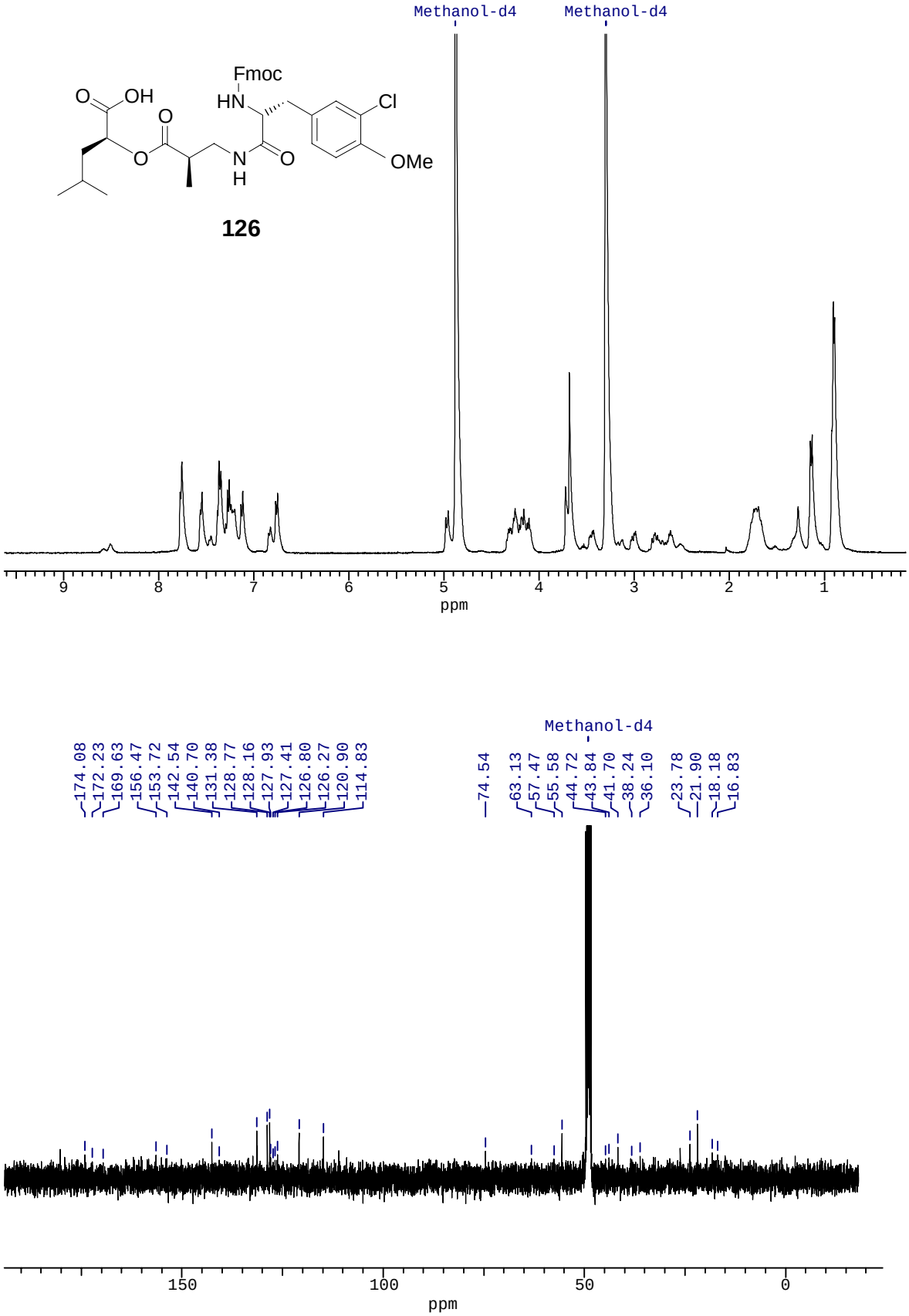
^{1}H- and ^{13}C-NMR spectra of compound **13c**



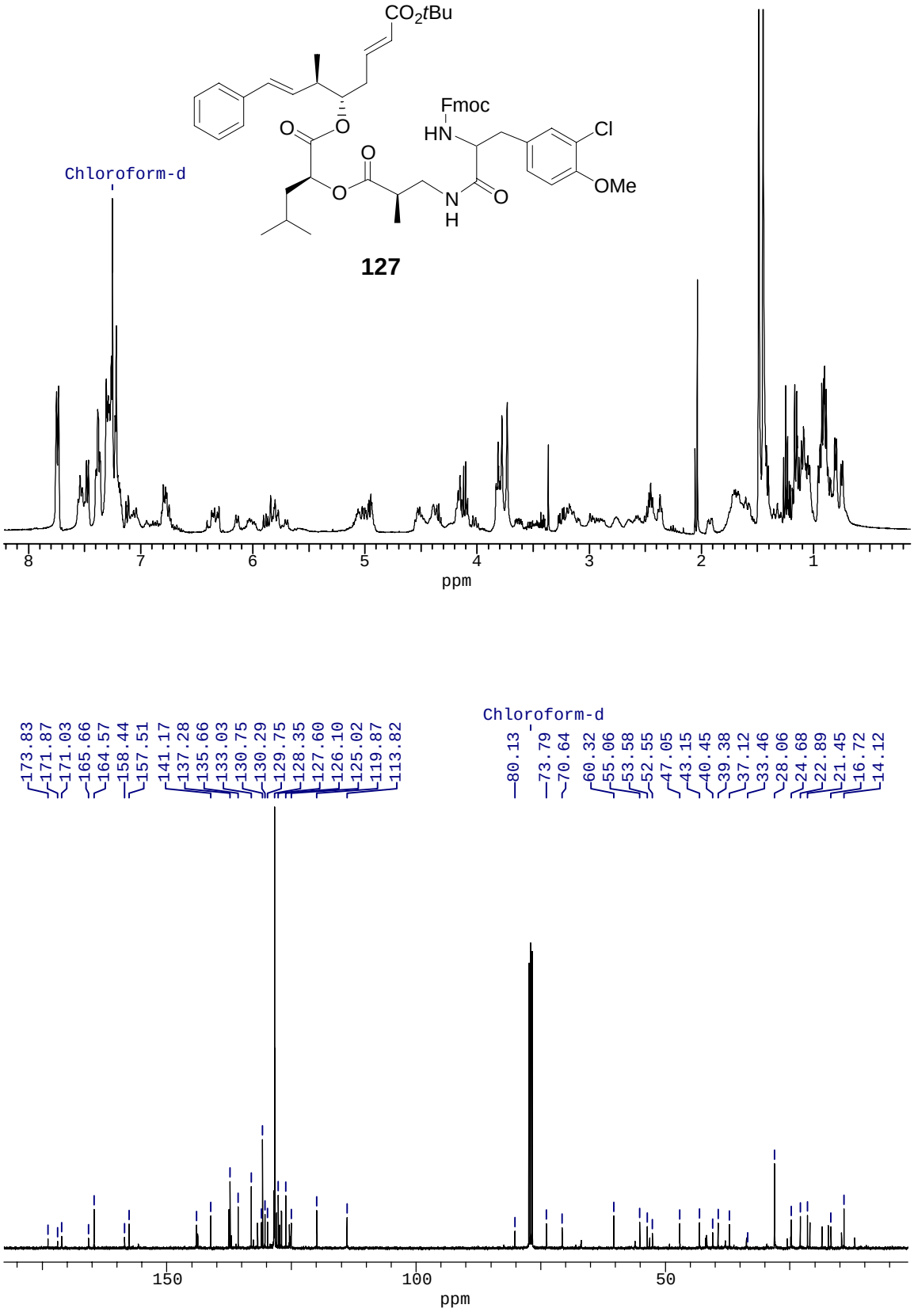
^{1}H- and ^{13}C-NMR spectra of compound **14c**



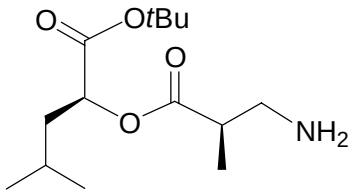
^{1}H- and ^{13}C-NMR spectra of compound **15c**



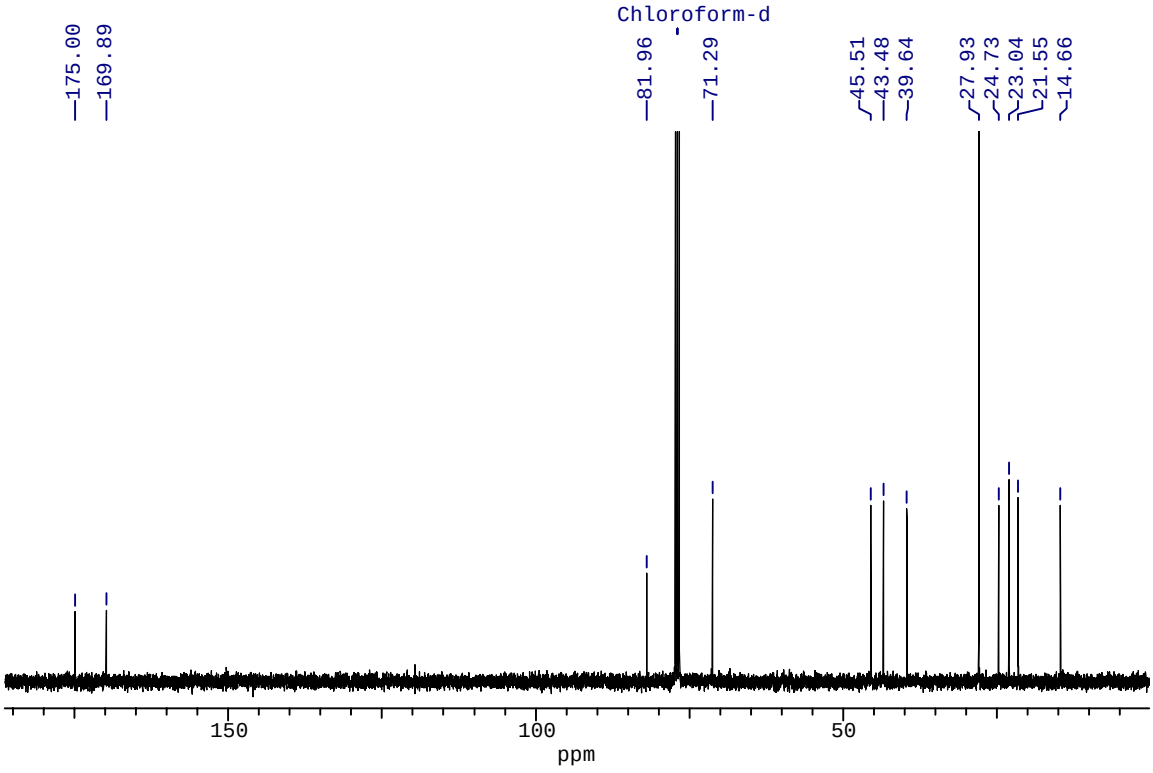
^{1}H- and ^{13}C-NMR spectra of compound **126**



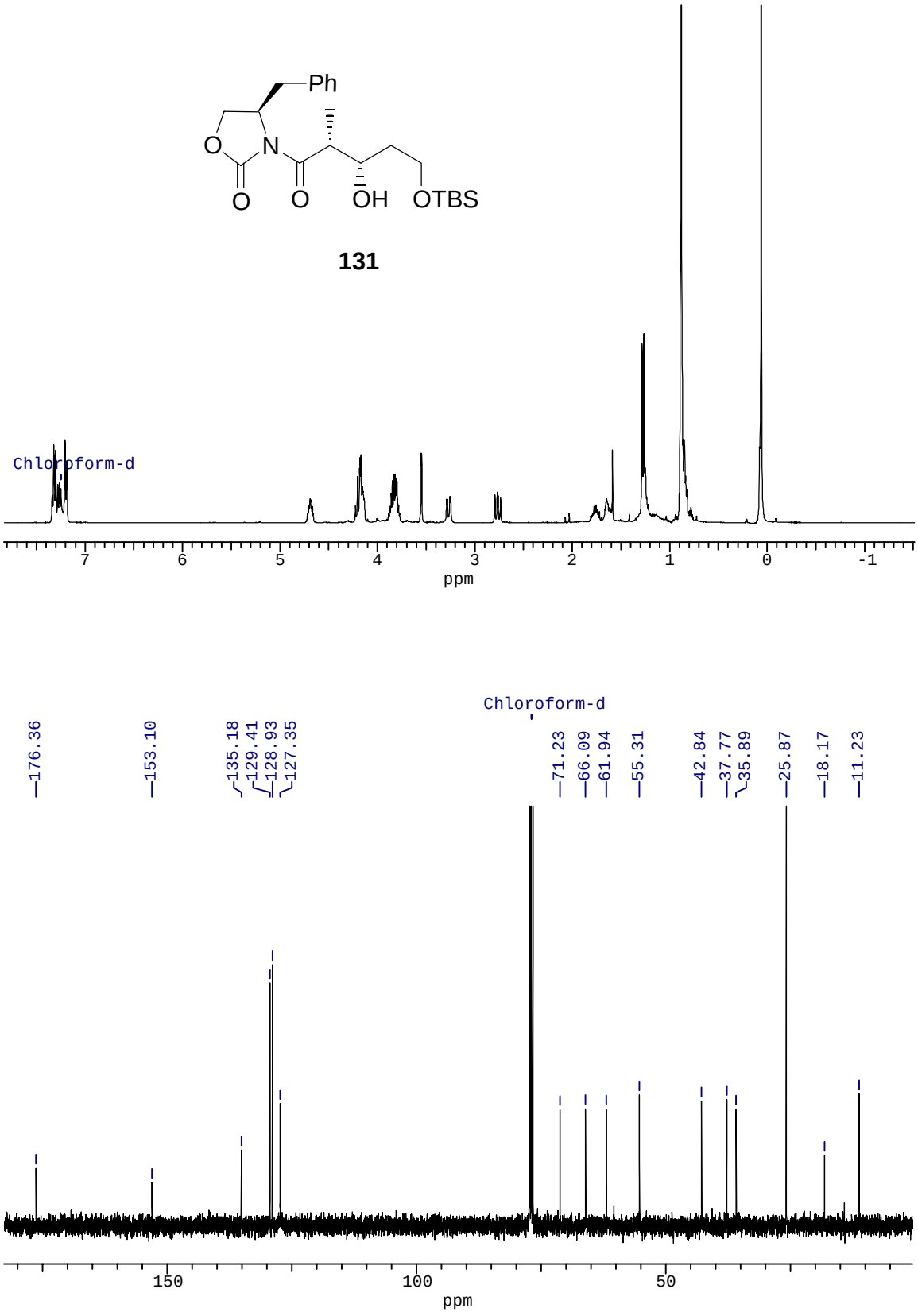
^{1}H- and ^{13}C-NMR spectra of compound **127**



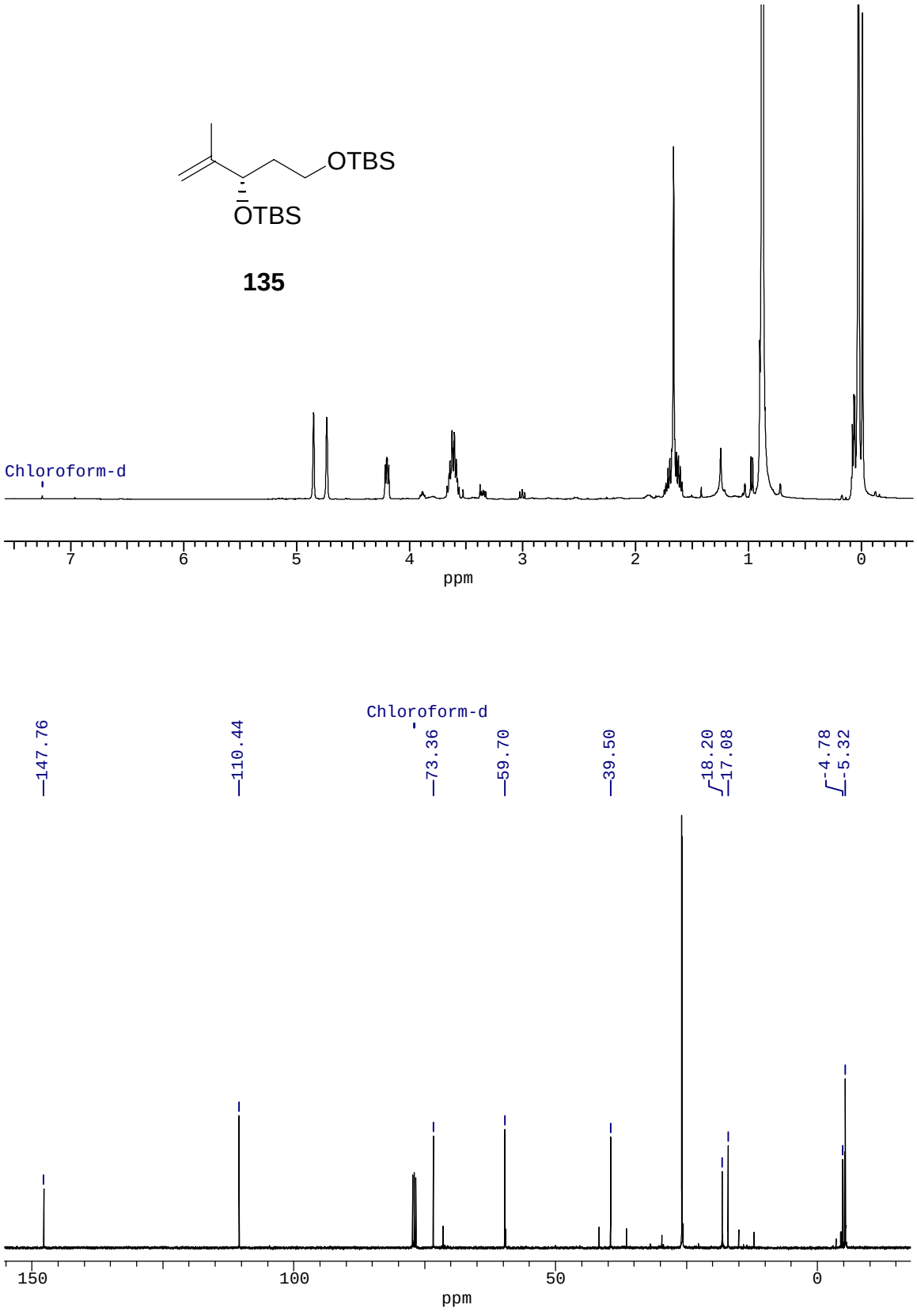
128



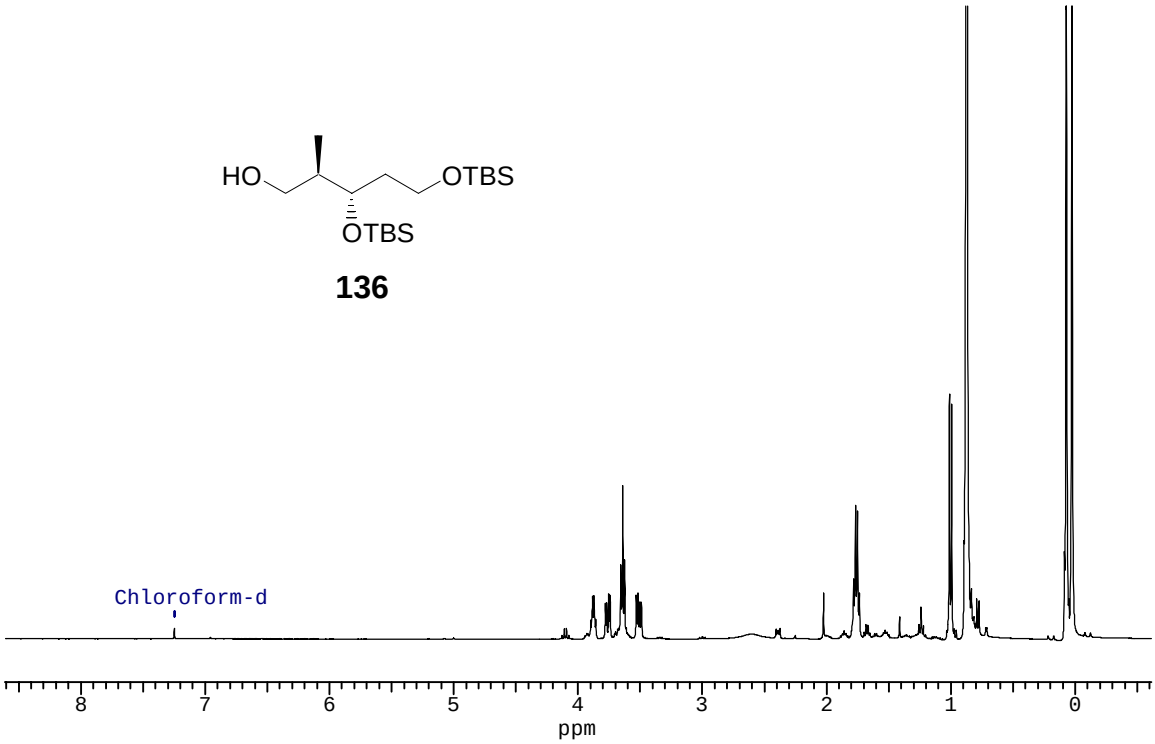
^{1}H- and ^{13}C-NMR spectra of compound **128**



^{1}H- and ^{13}C-NMR spectra of compound **131**



^{1}H- and ^{13}C-NMR spectra of compound **135**



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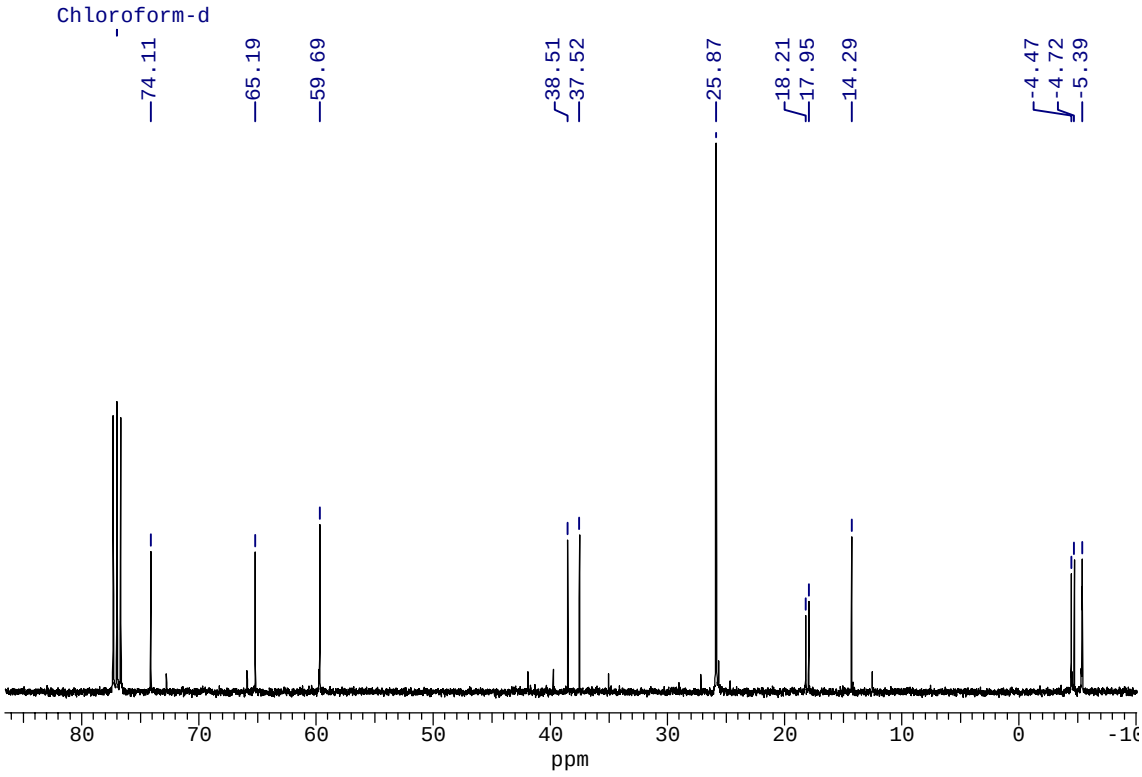
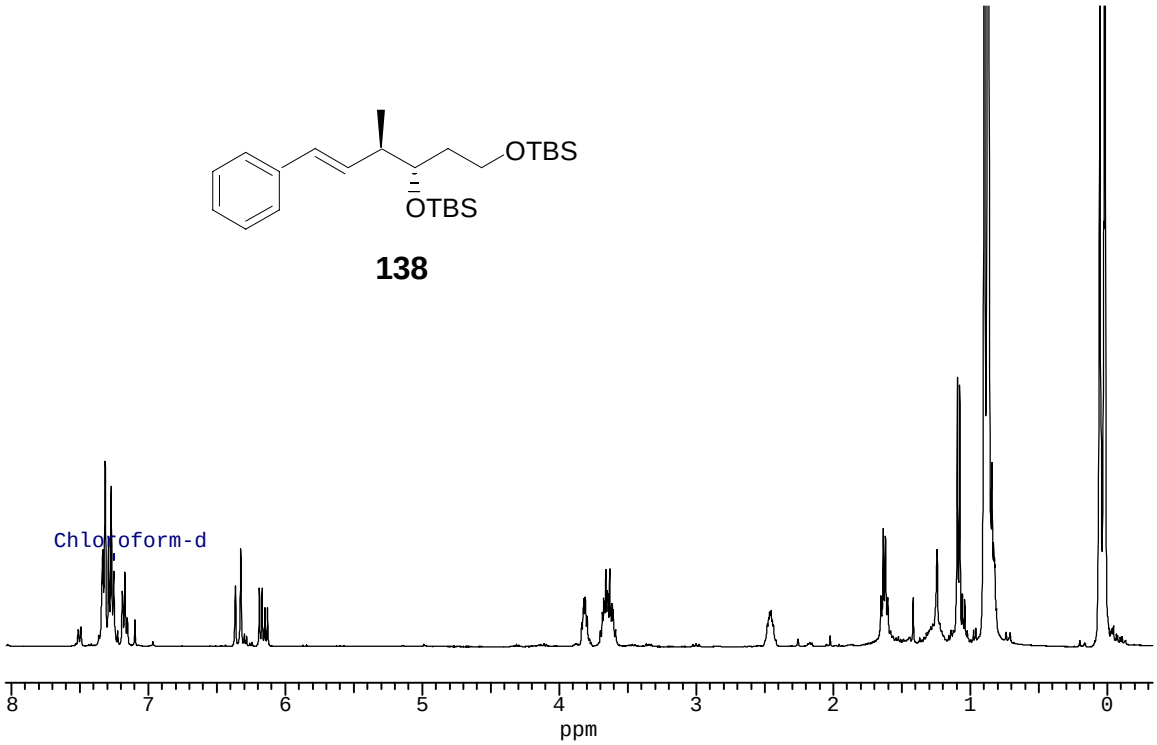


^{1}H- and ^{13}C-NMR spectra of compound **136**



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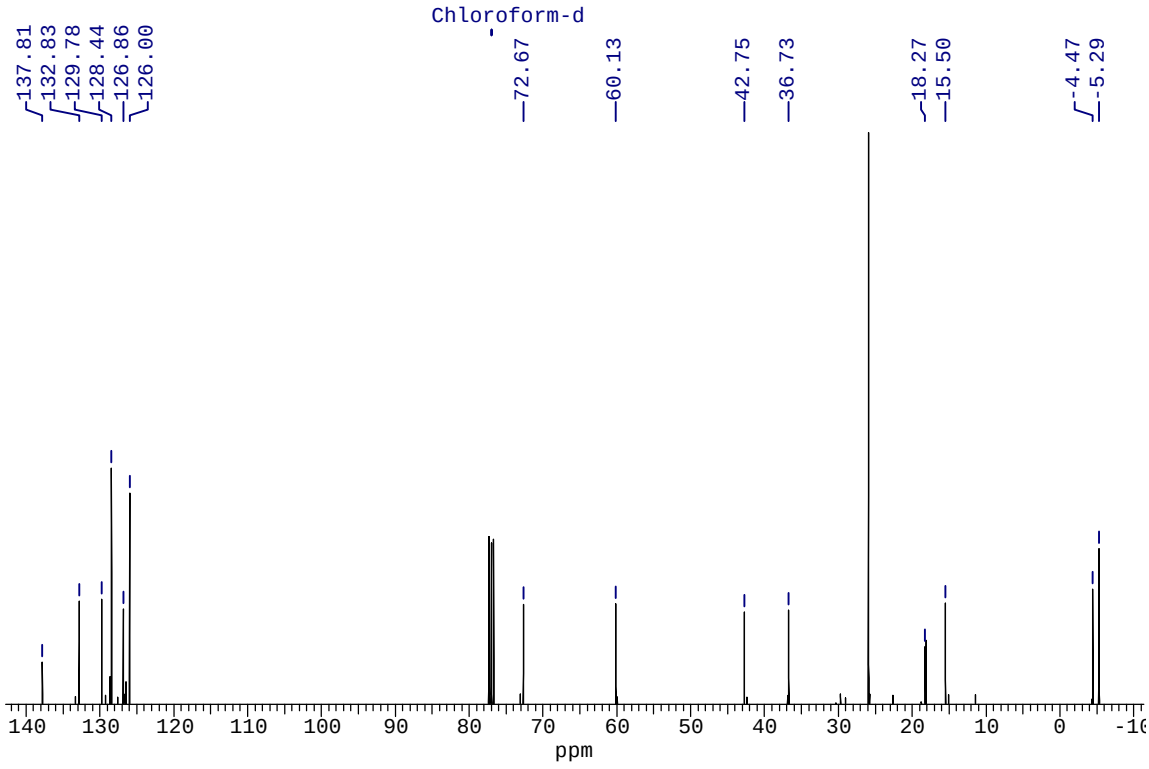
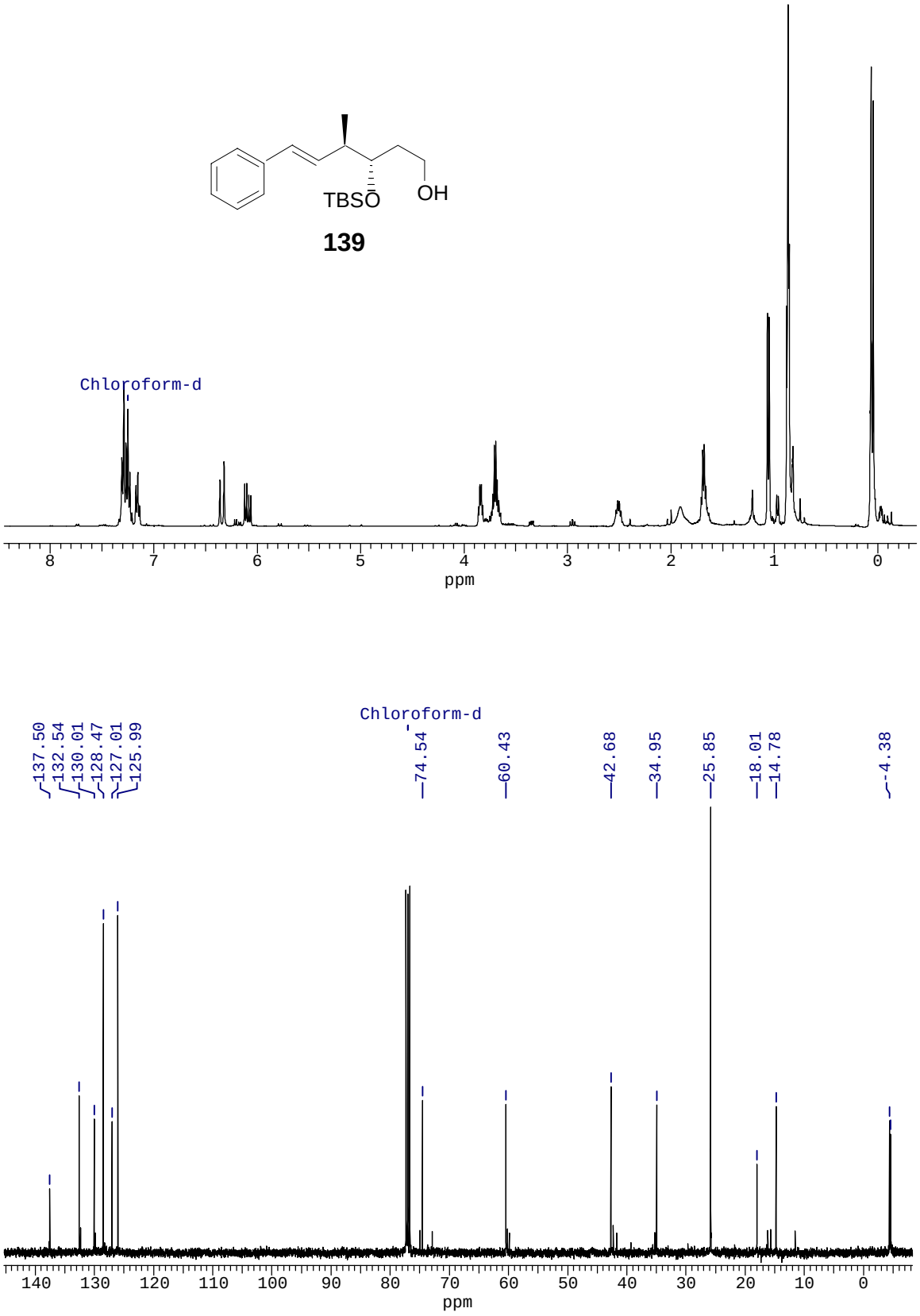
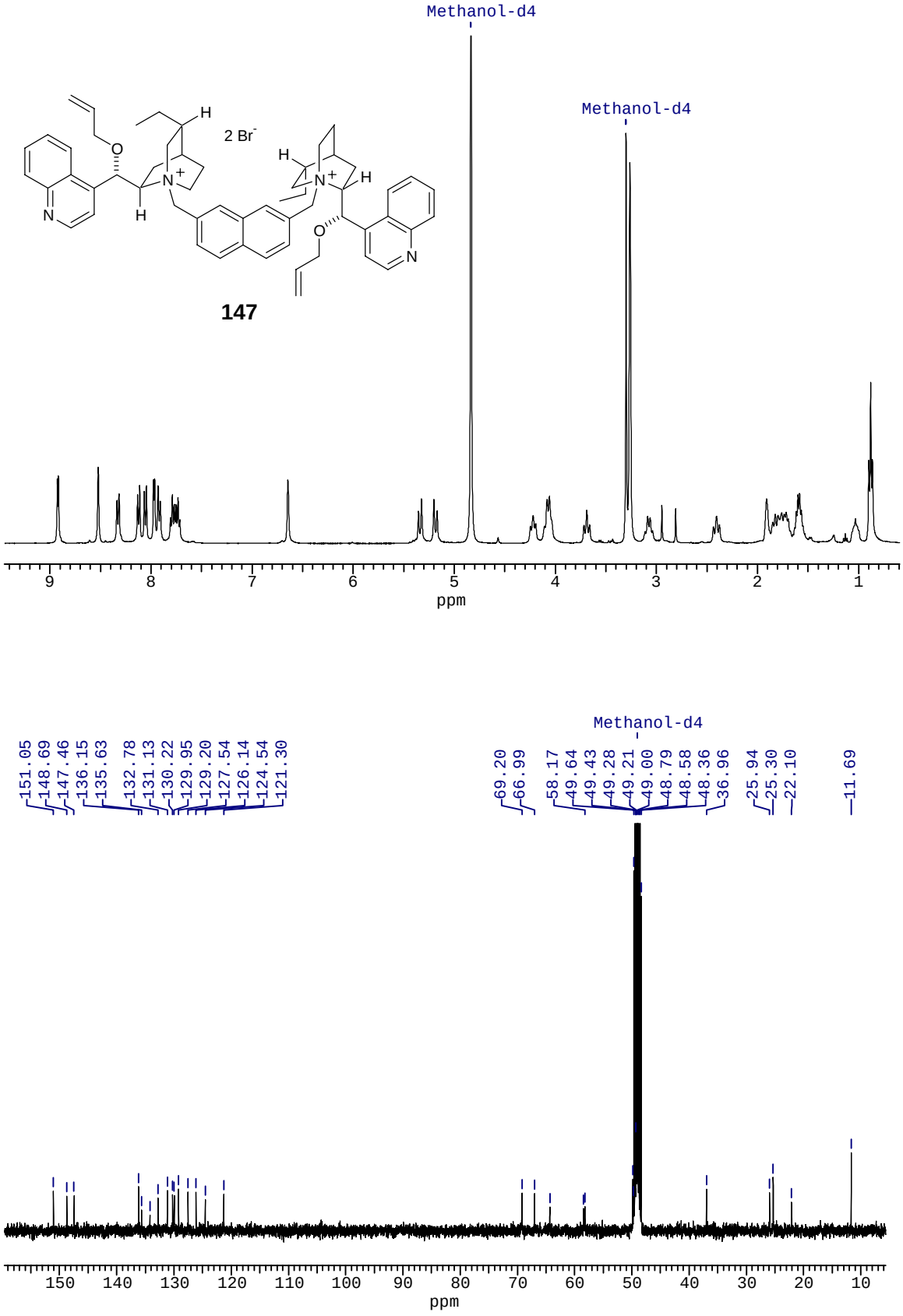


^{1}H- and ^{13}C-NMR spectra of compound **138**



^{1}H- and ^{13}C-NMR spectra of compound **139**



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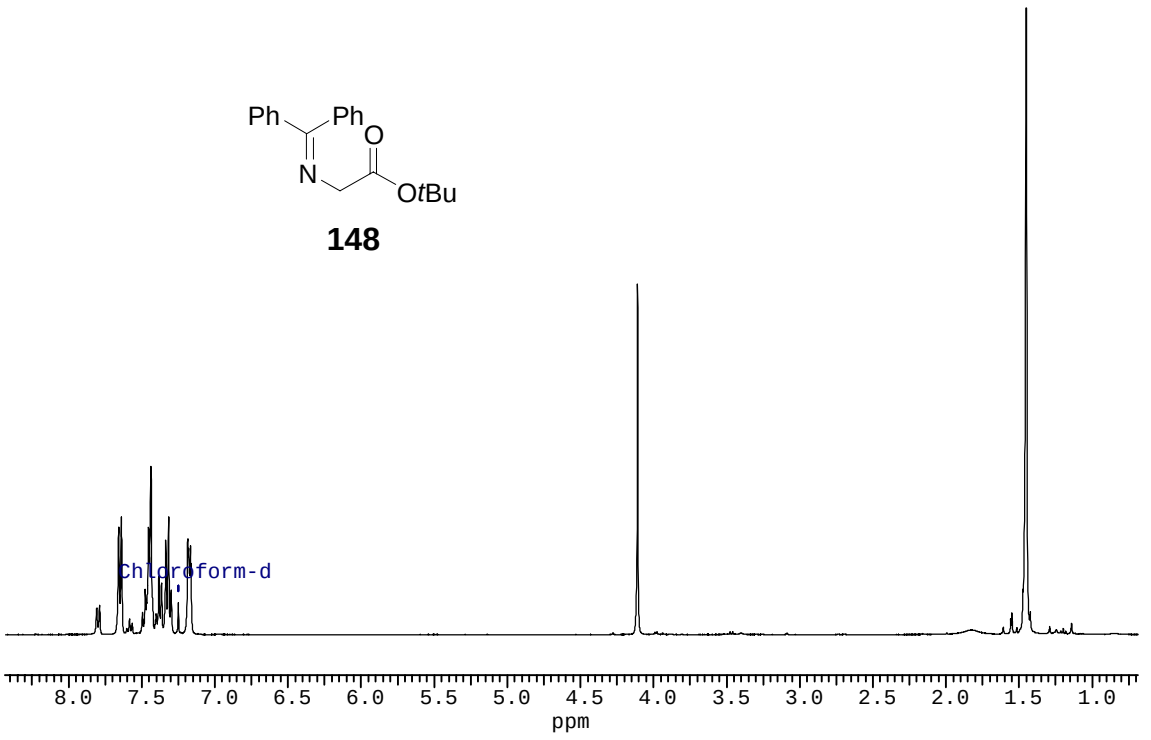
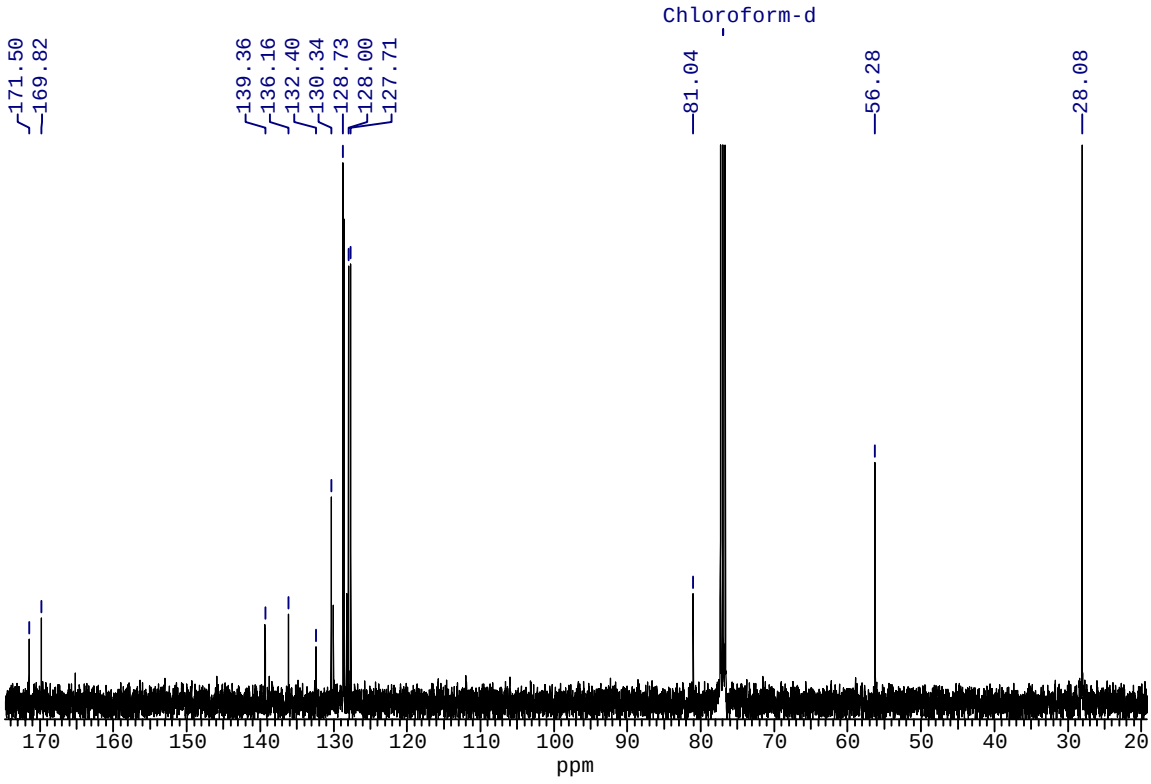
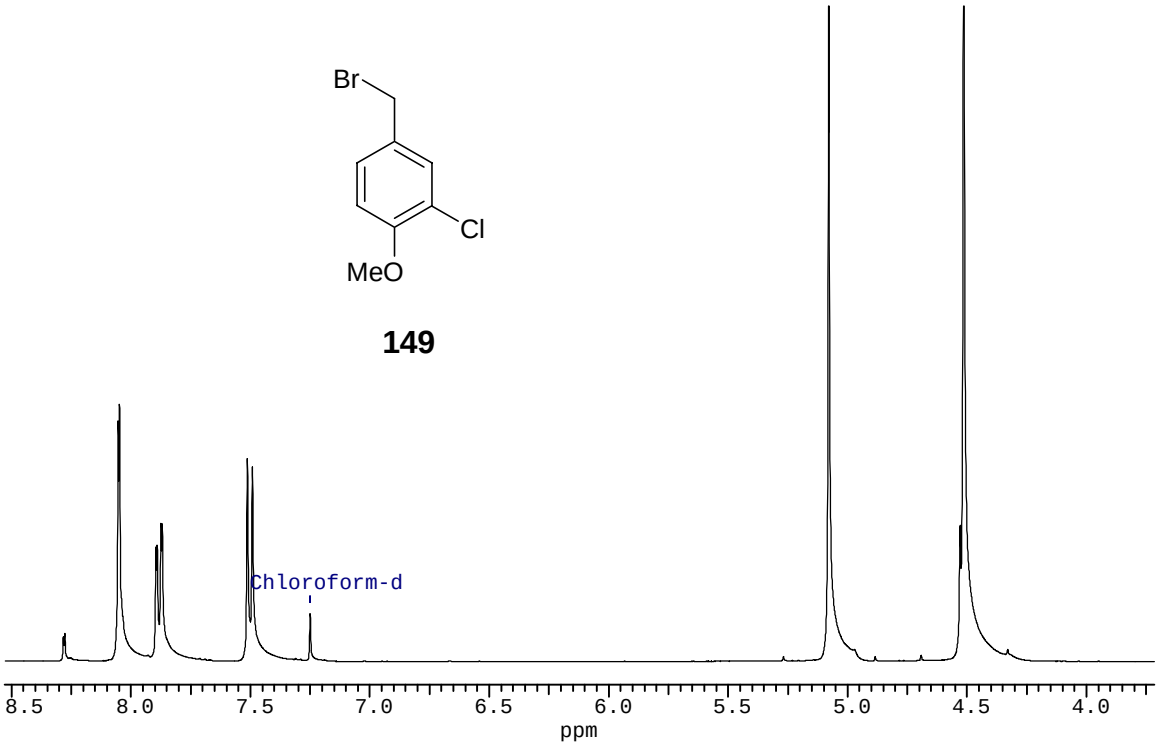
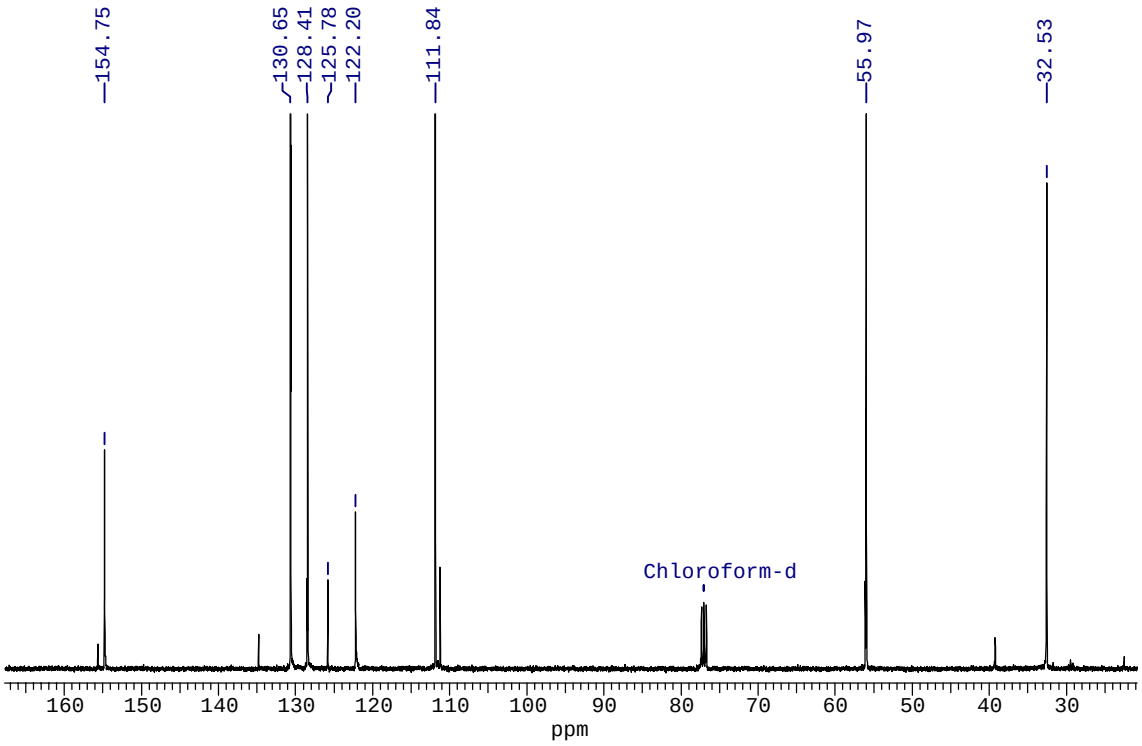


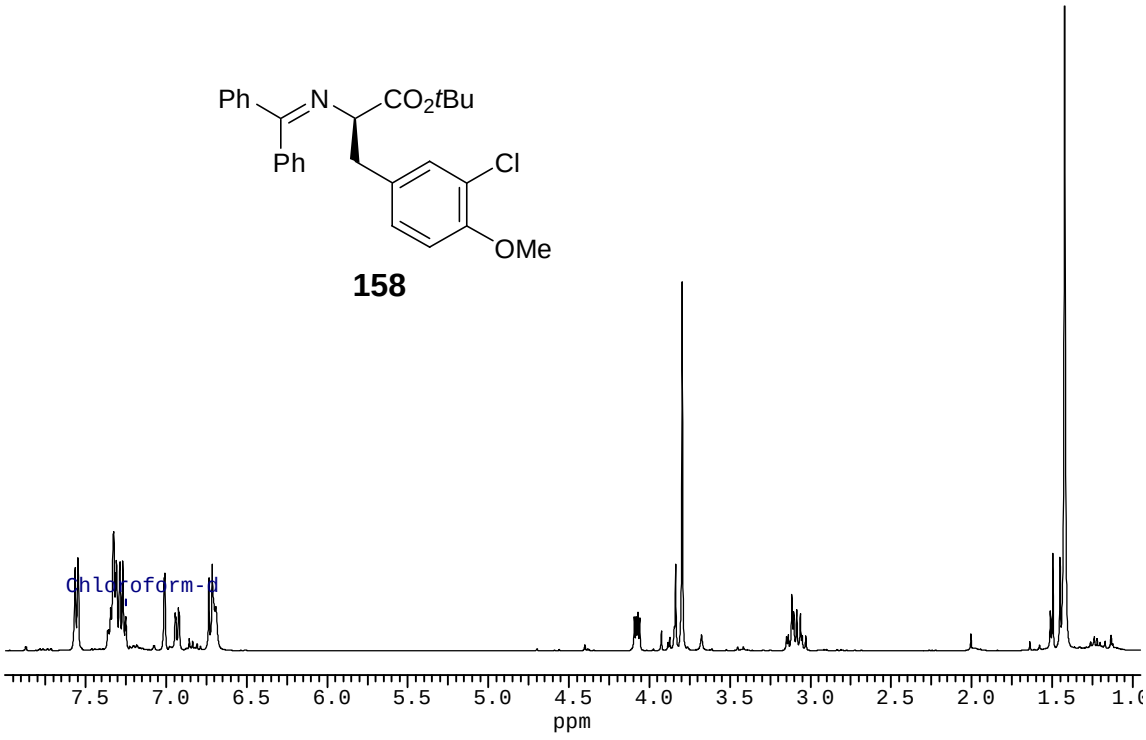


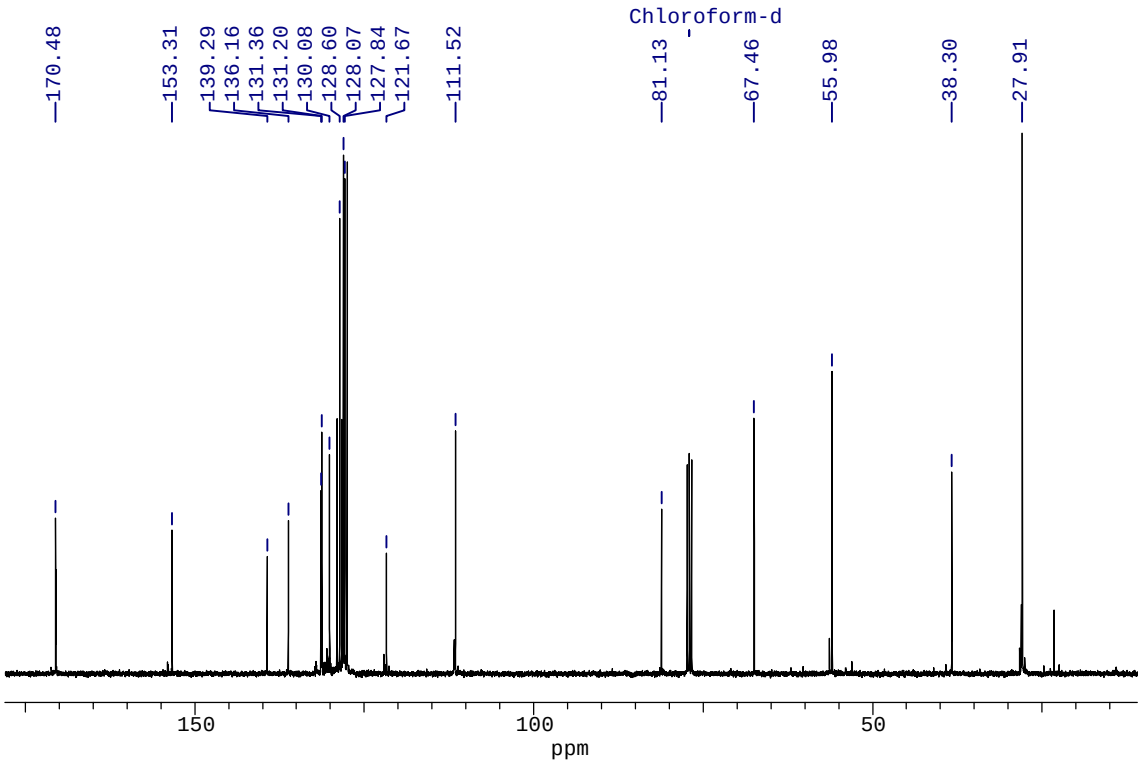
^{1}H- and ^{13}C-NMR spectra of compound **148**



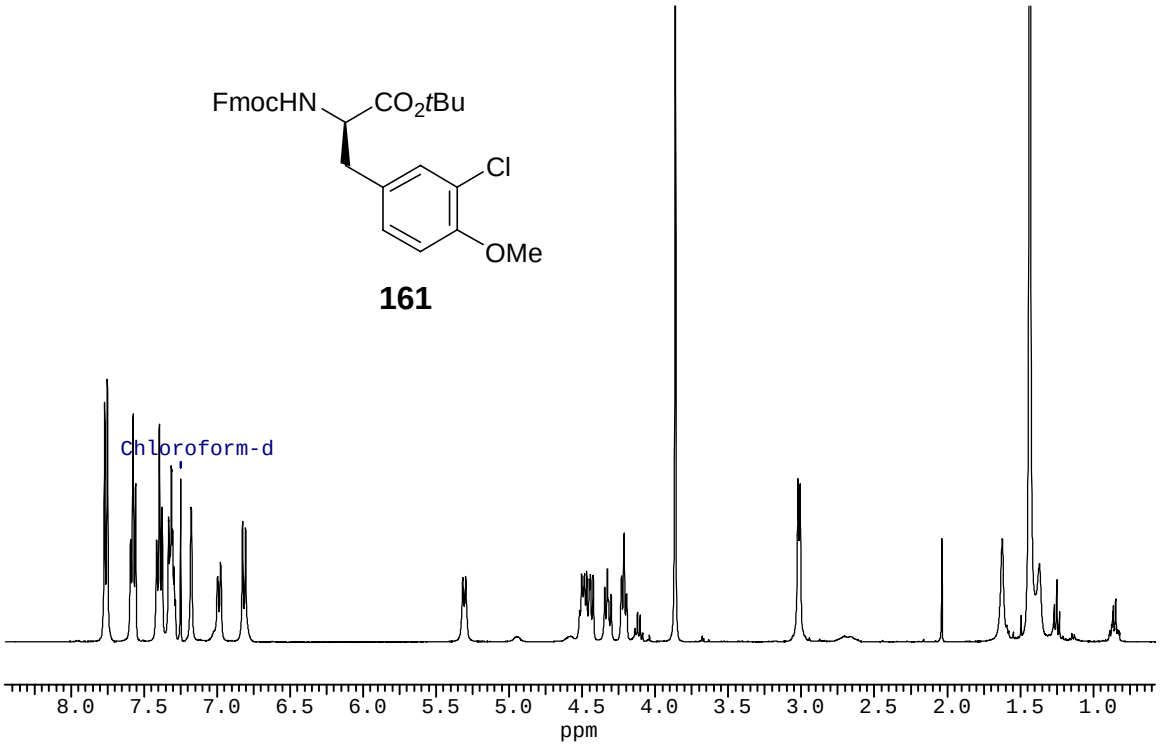


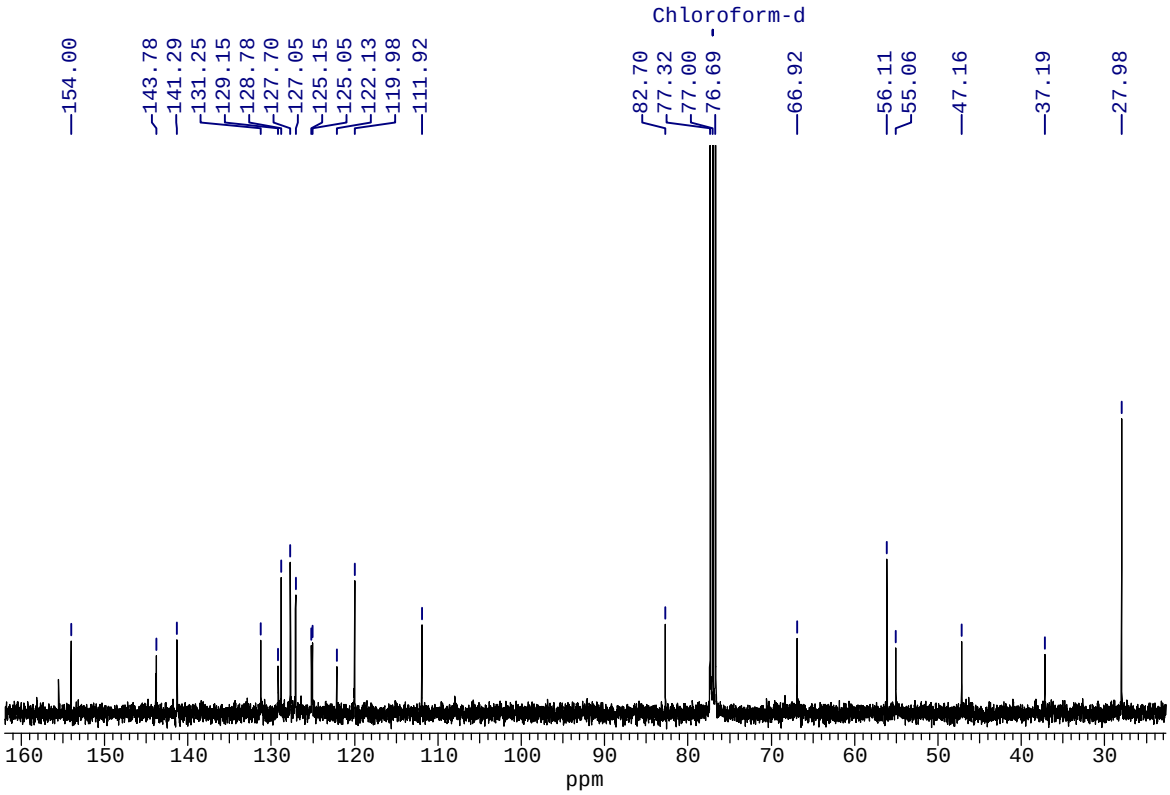
^{1}H- and ^{13}C-NMR spectra of compound **149**



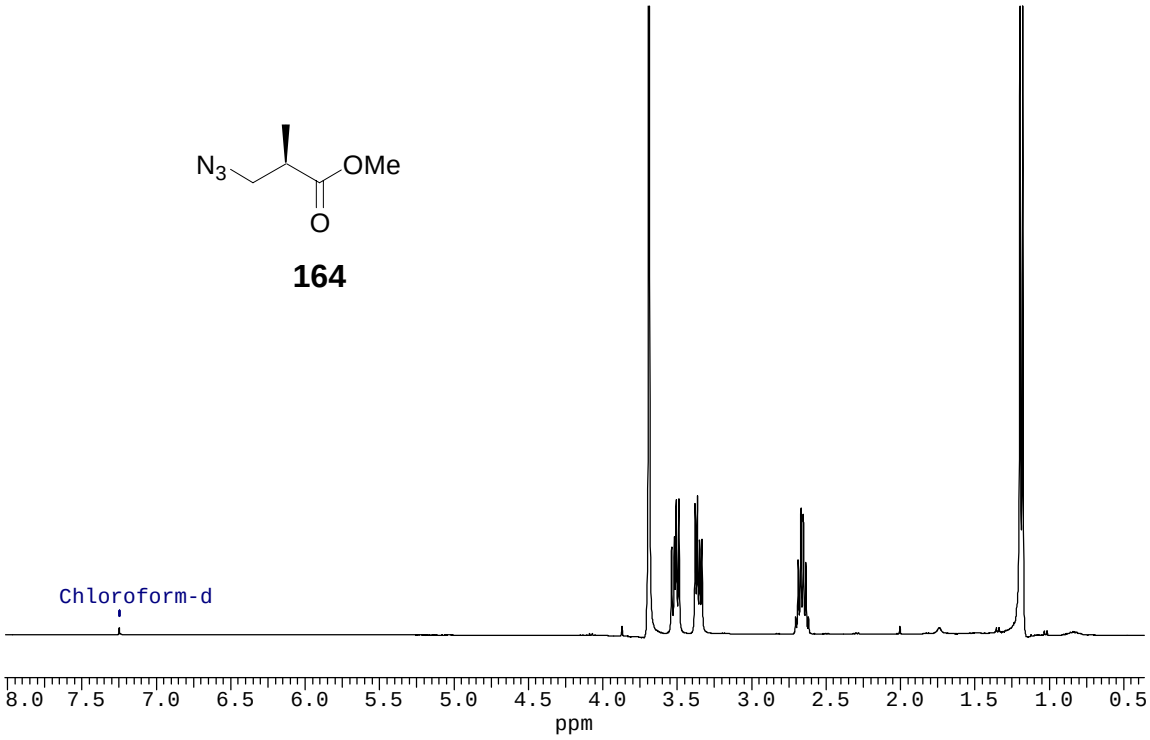


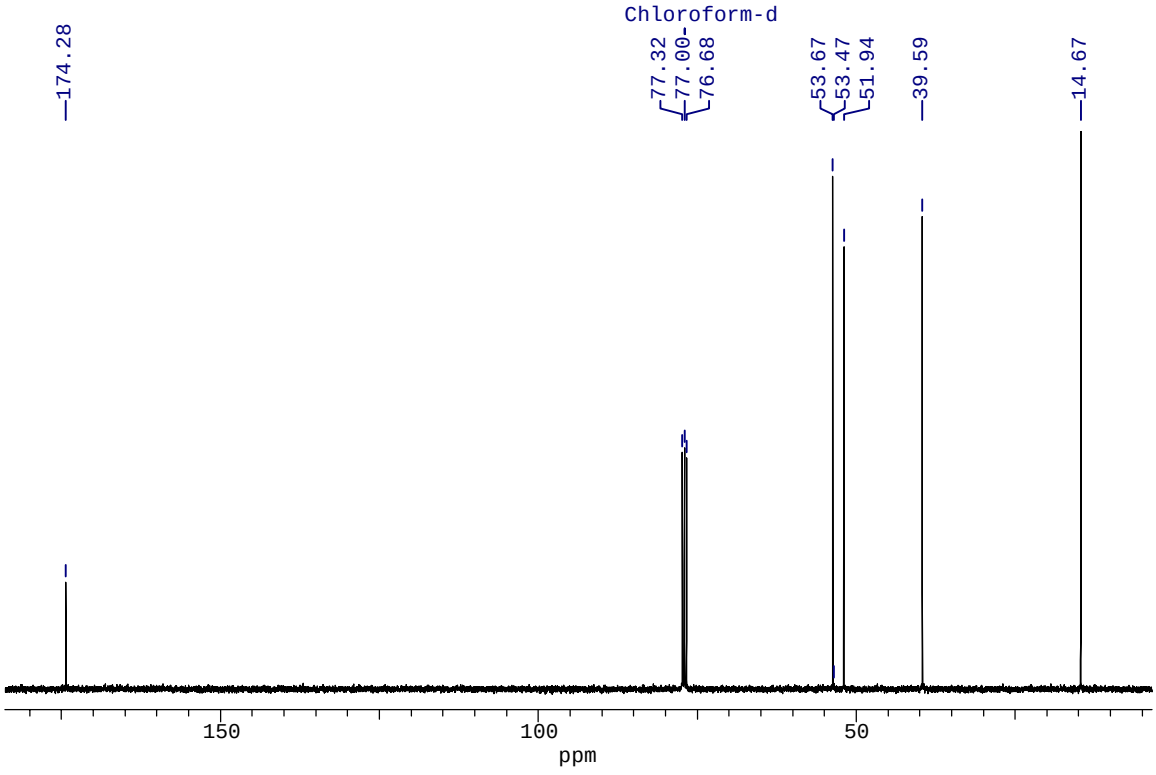
^{1}H- and ^{13}C-NMR spectra of compound **158**



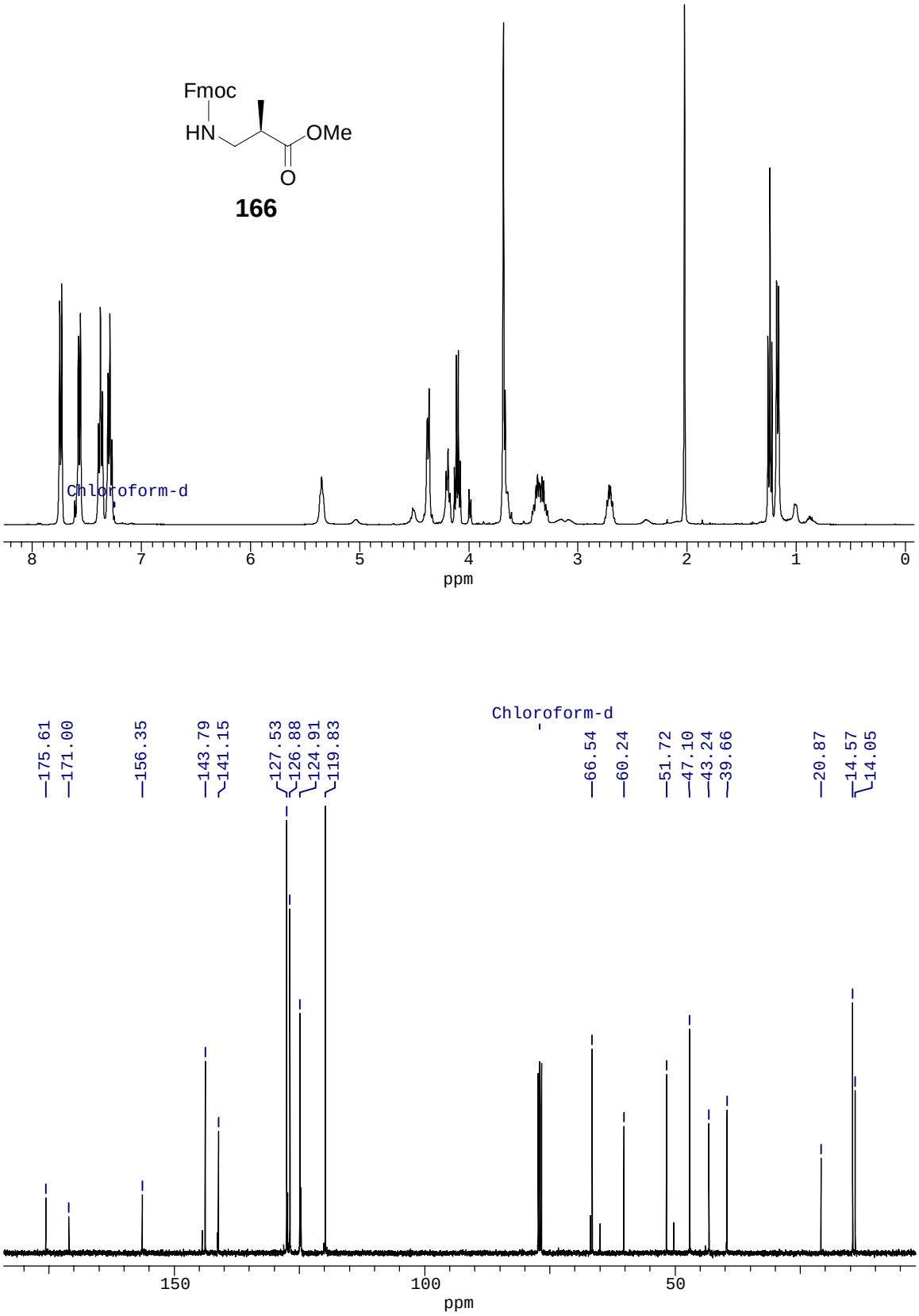


^{1}H- and ^{13}C-NMR spectra of compound **161**

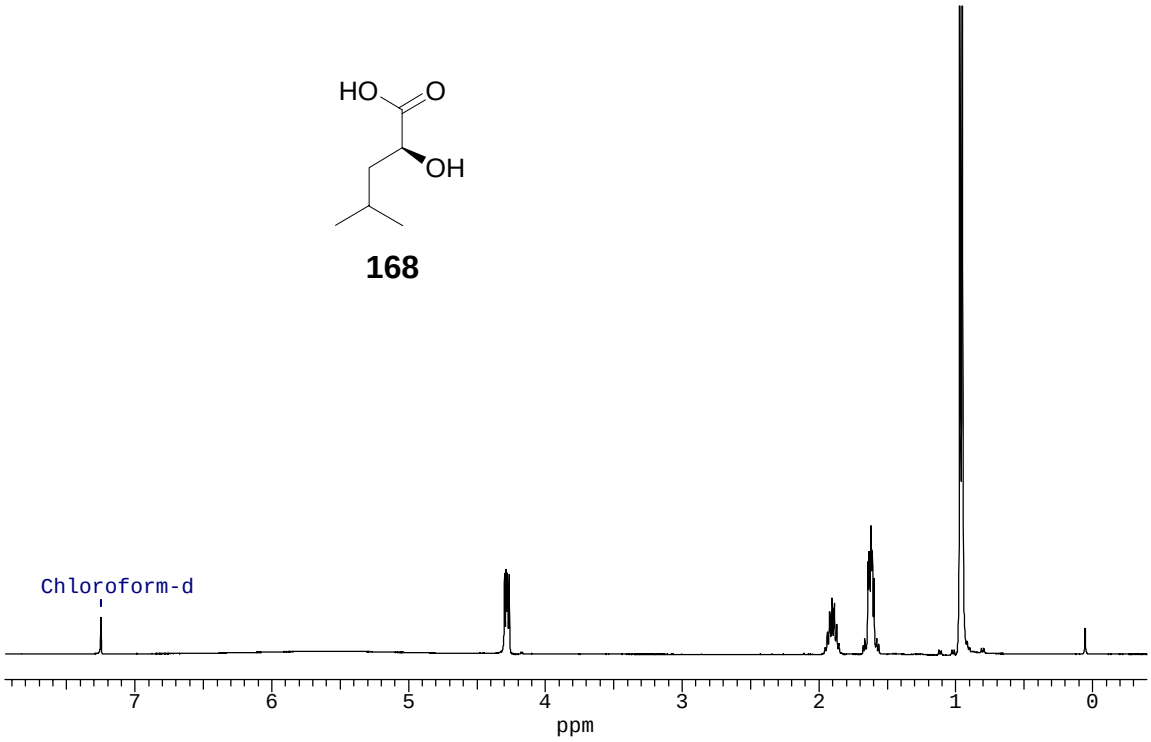




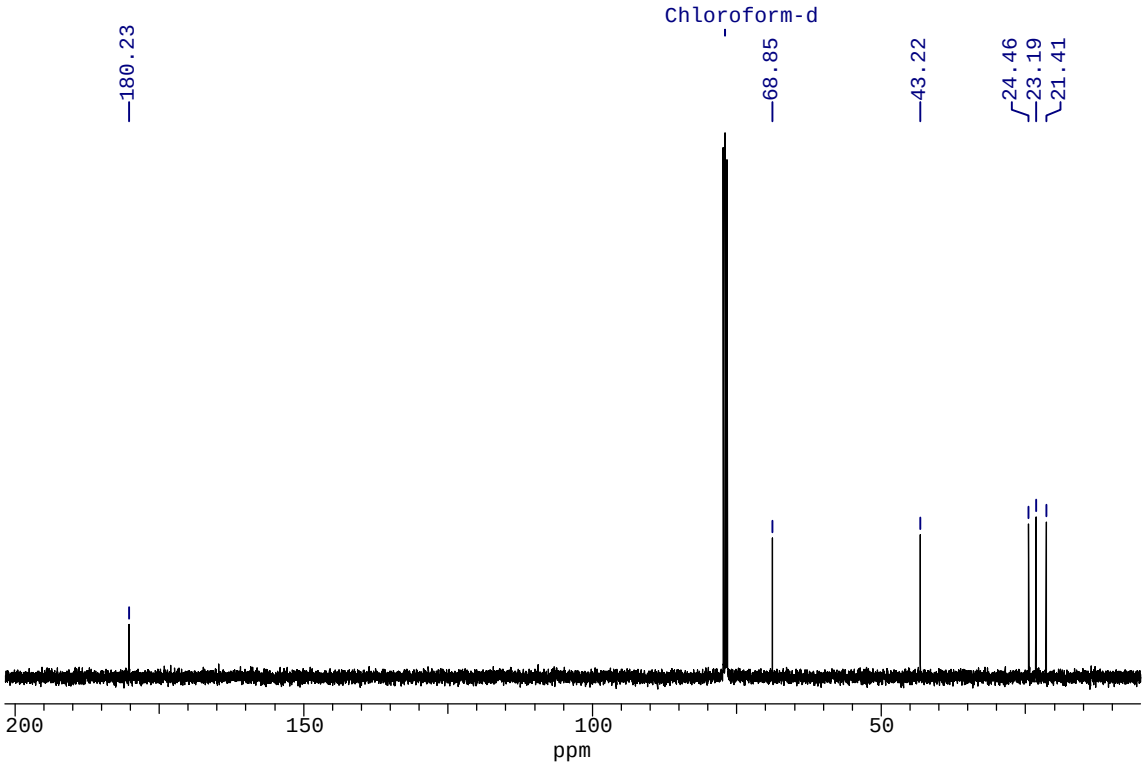
^{1}H- and ^{13}C-NMR spectra of compound **164**



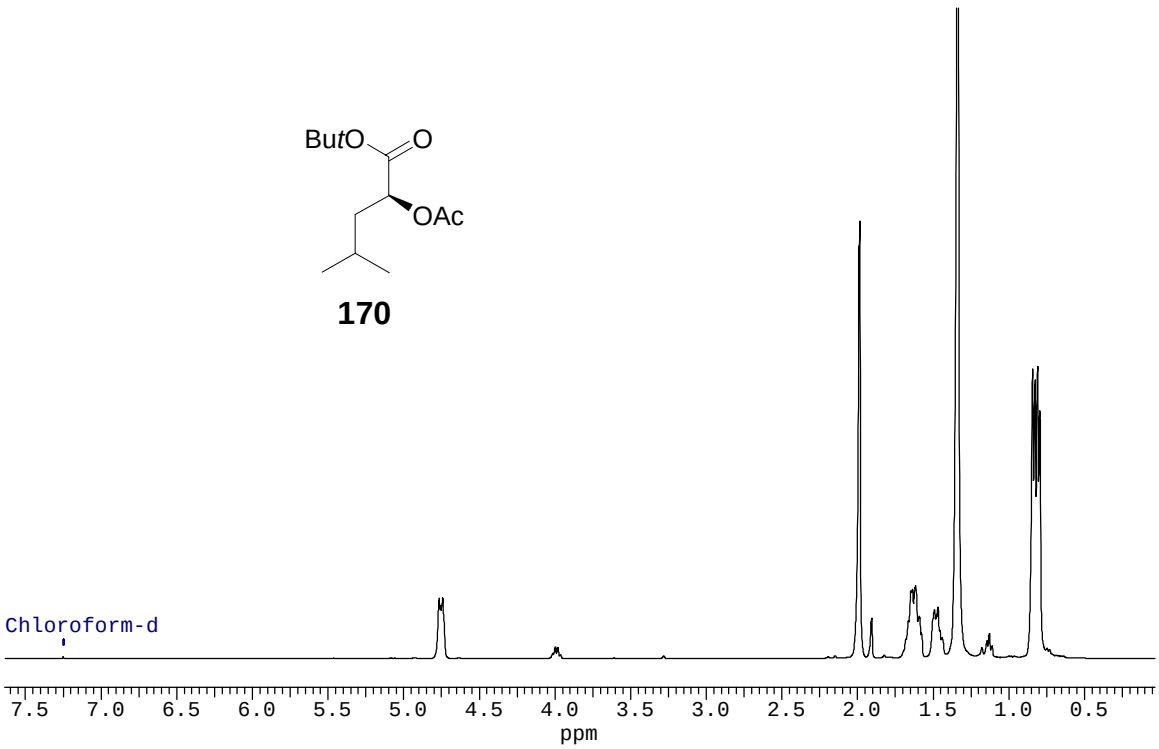
^{1}H- and ^{13}C-NMR spectra of compound **166**

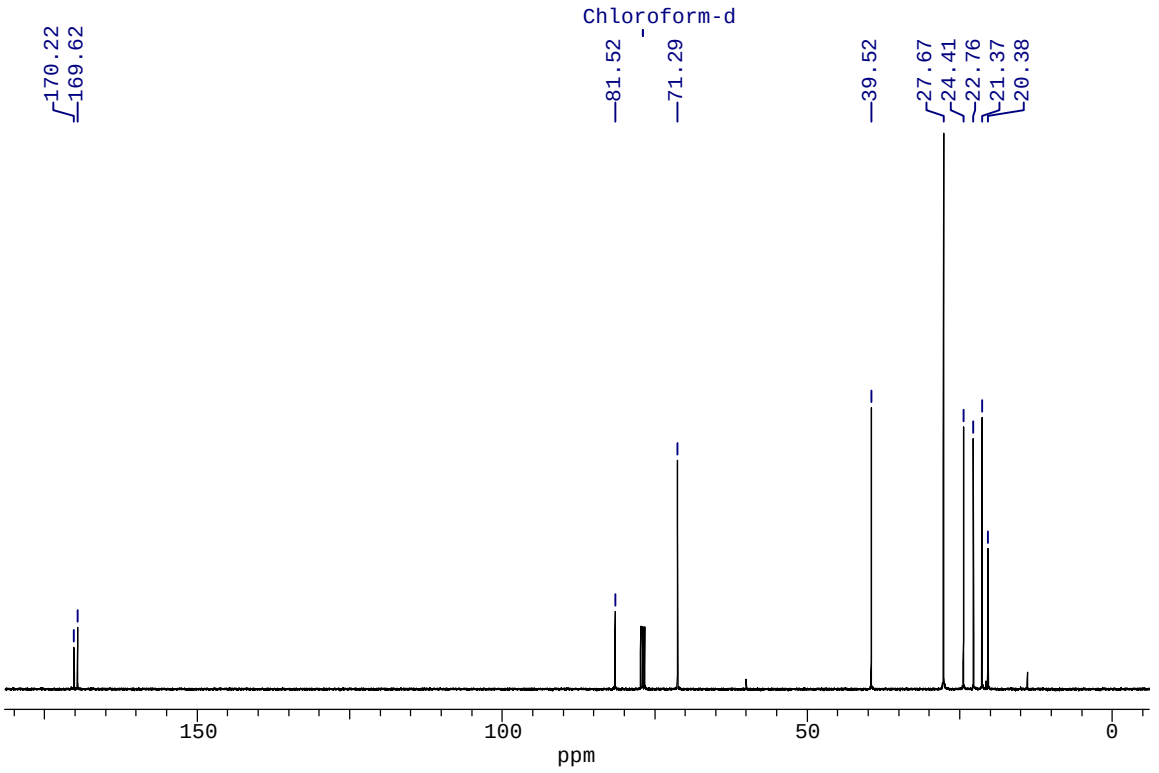


168

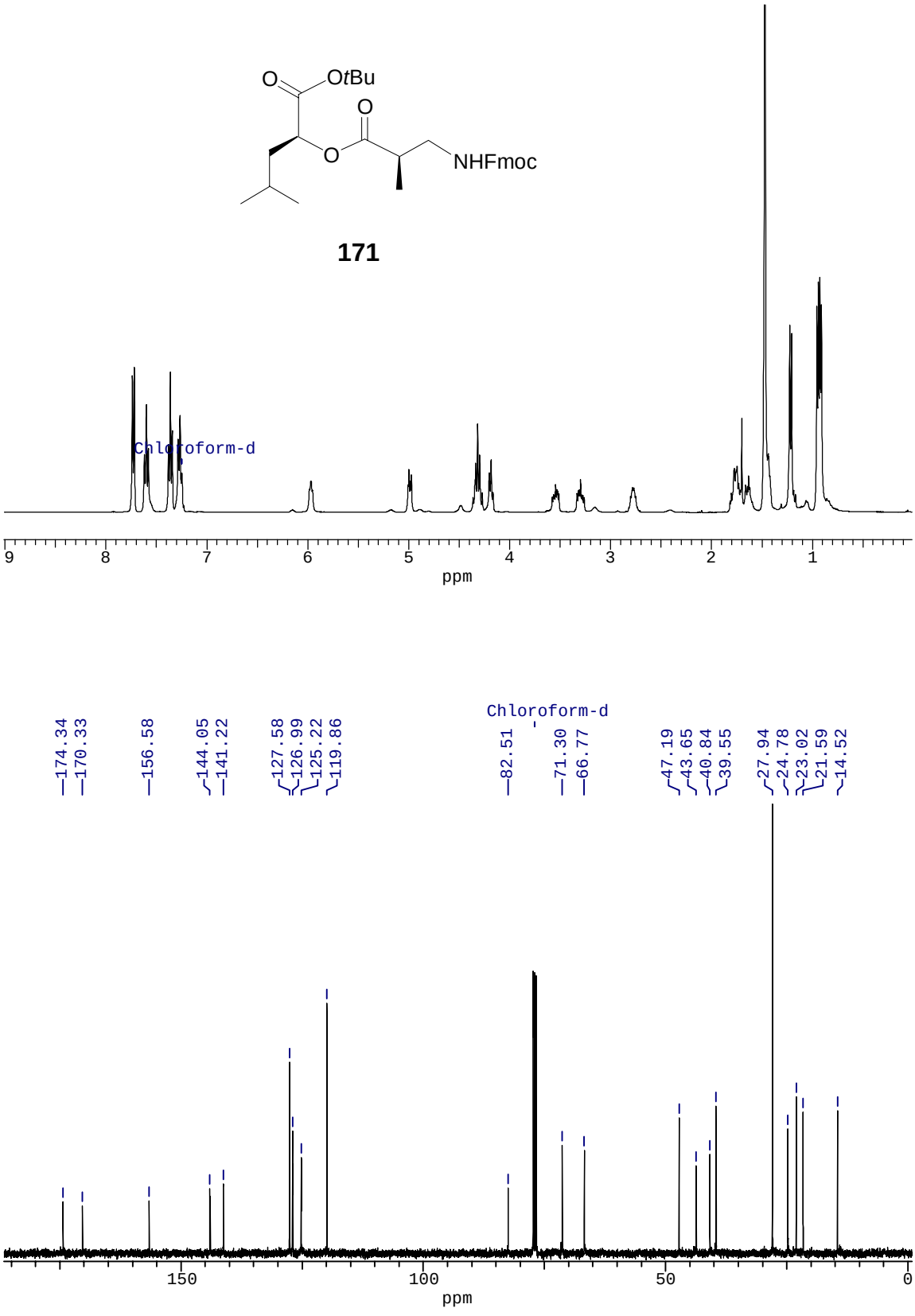


^{1}H- and ^{13}C-NMR spectra of compound **168**

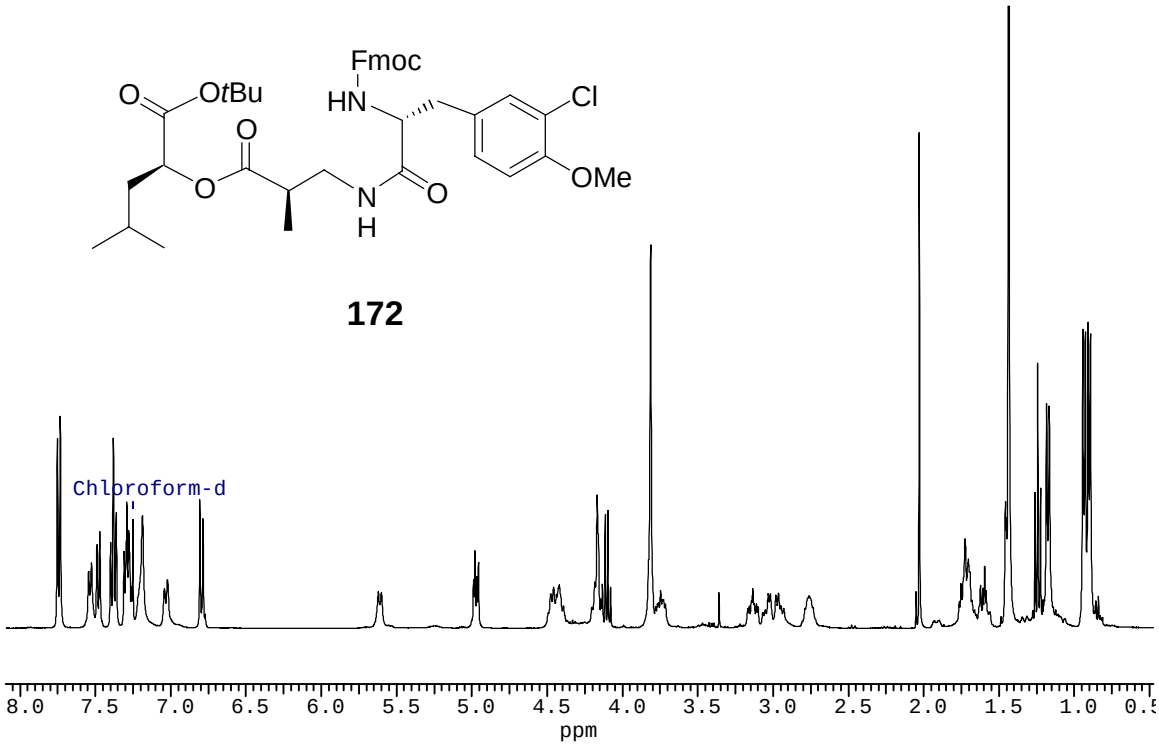




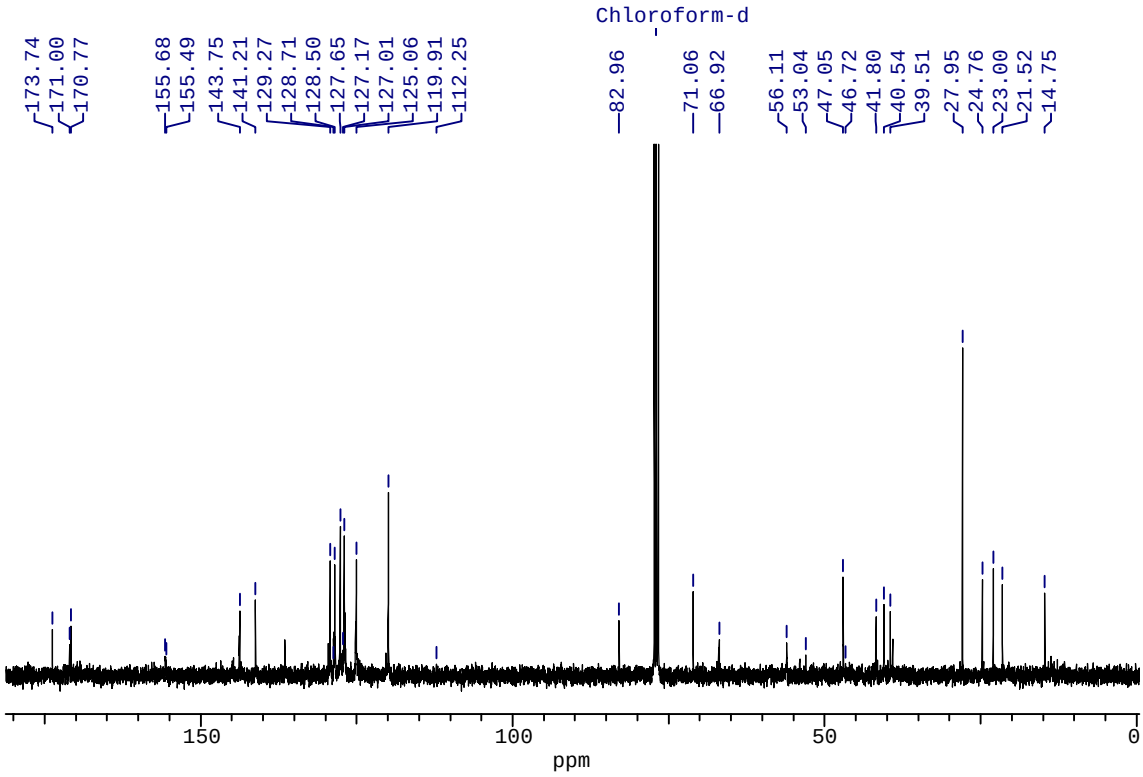
^{1}H- and ^{13}C-NMR spectra of compound **170**



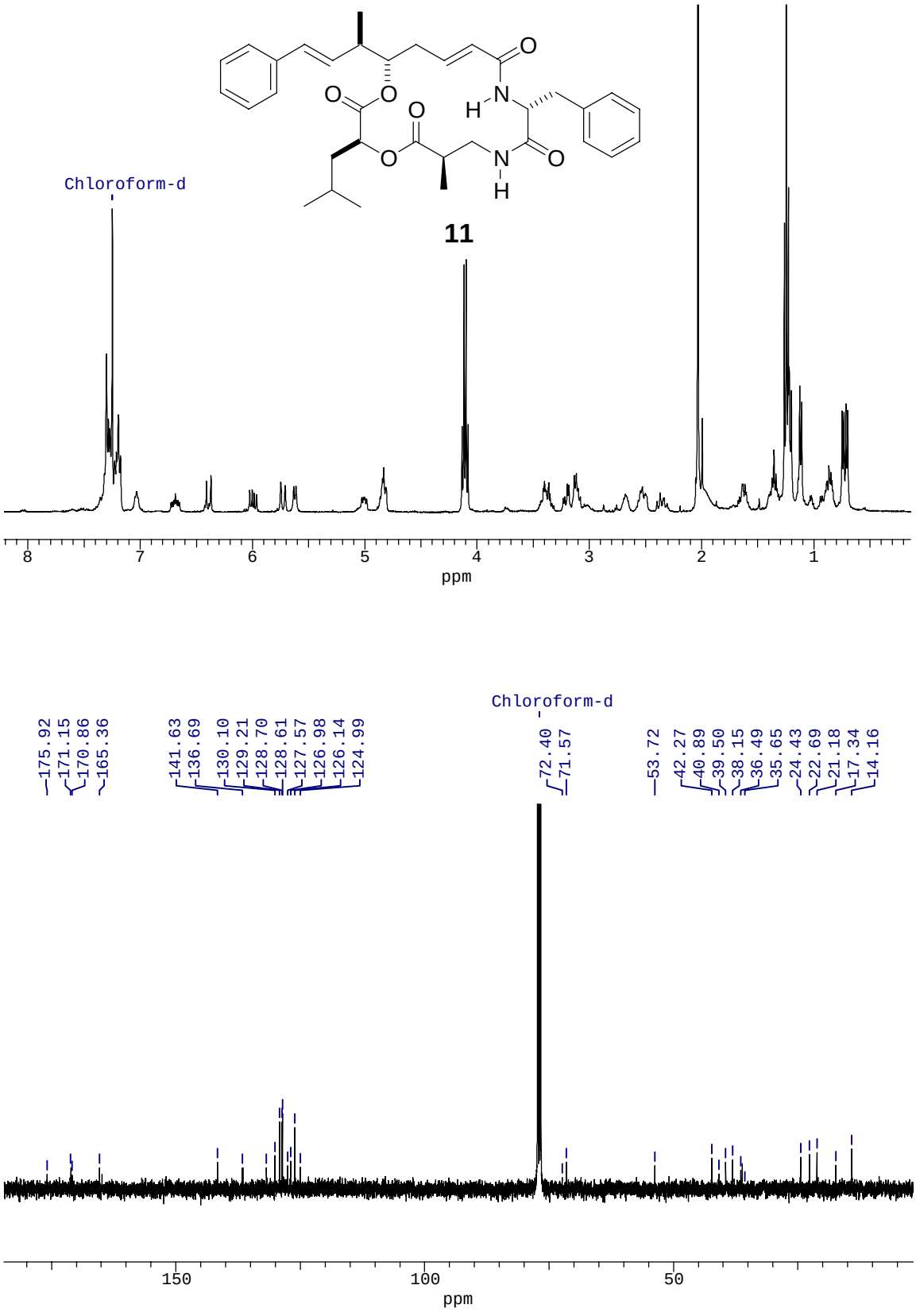
^{1}H- and ^{13}C-NMR spectra of compound **171**



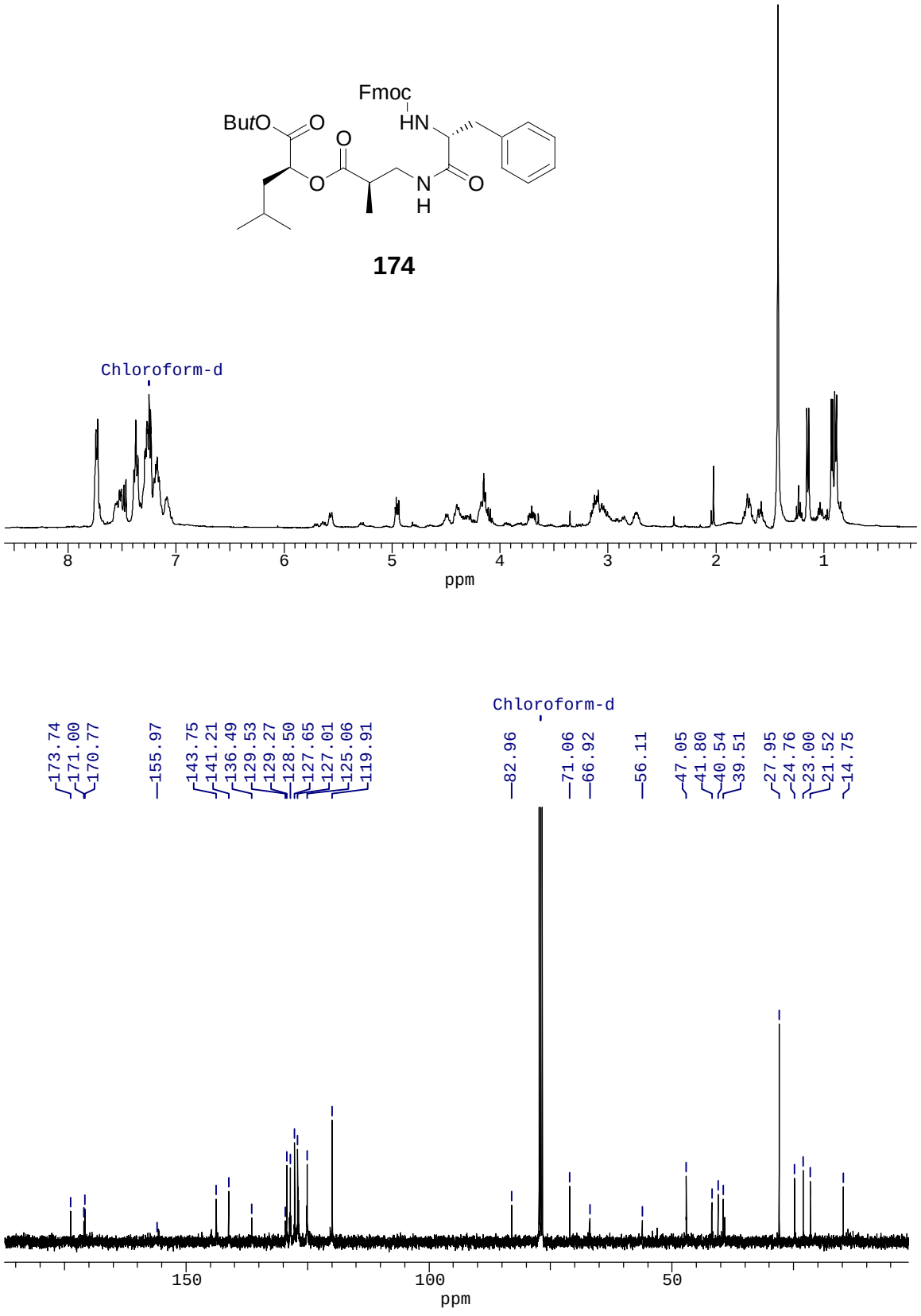
172



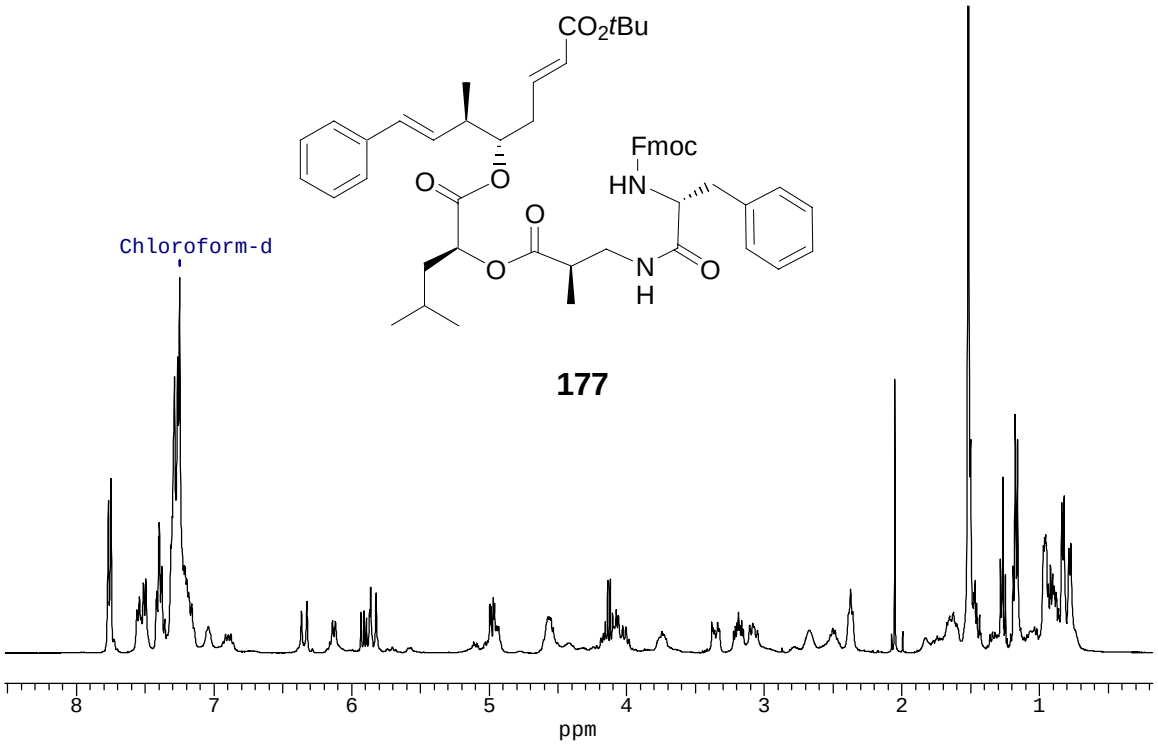
^{1}H- and ^{13}C-NMR spectra of compound **172**



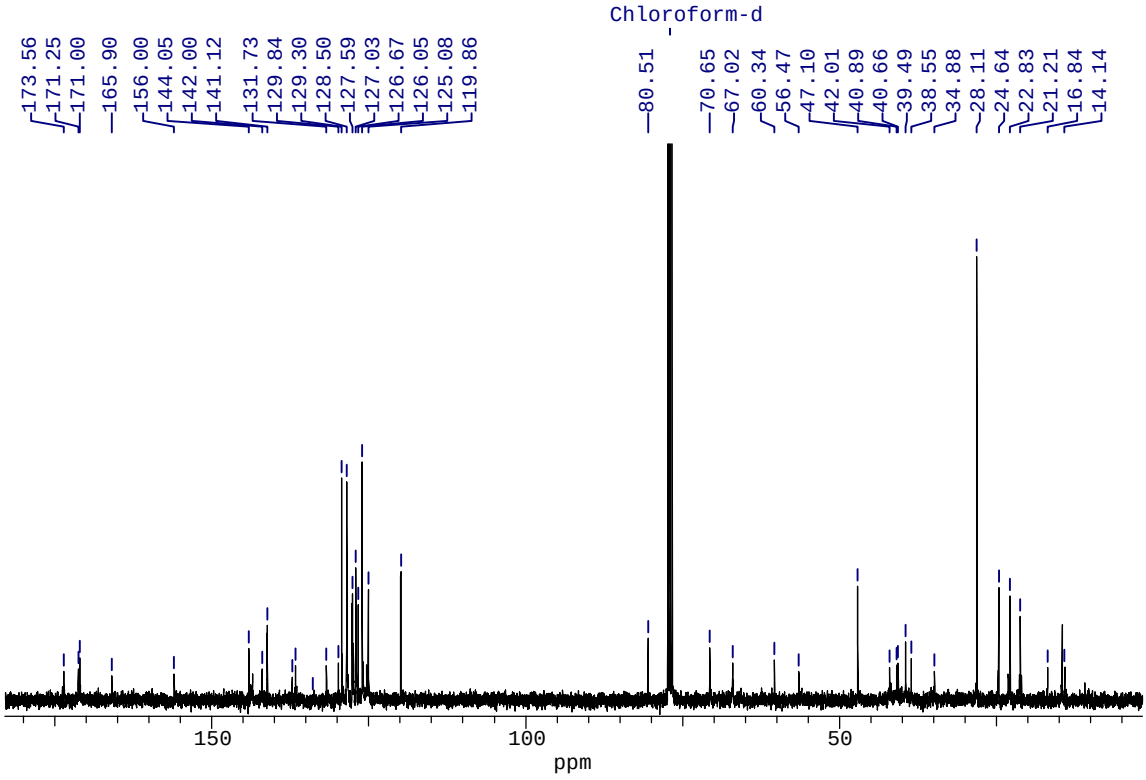
^{1}H- and ^{13}C-NMR spectra of compound **11**



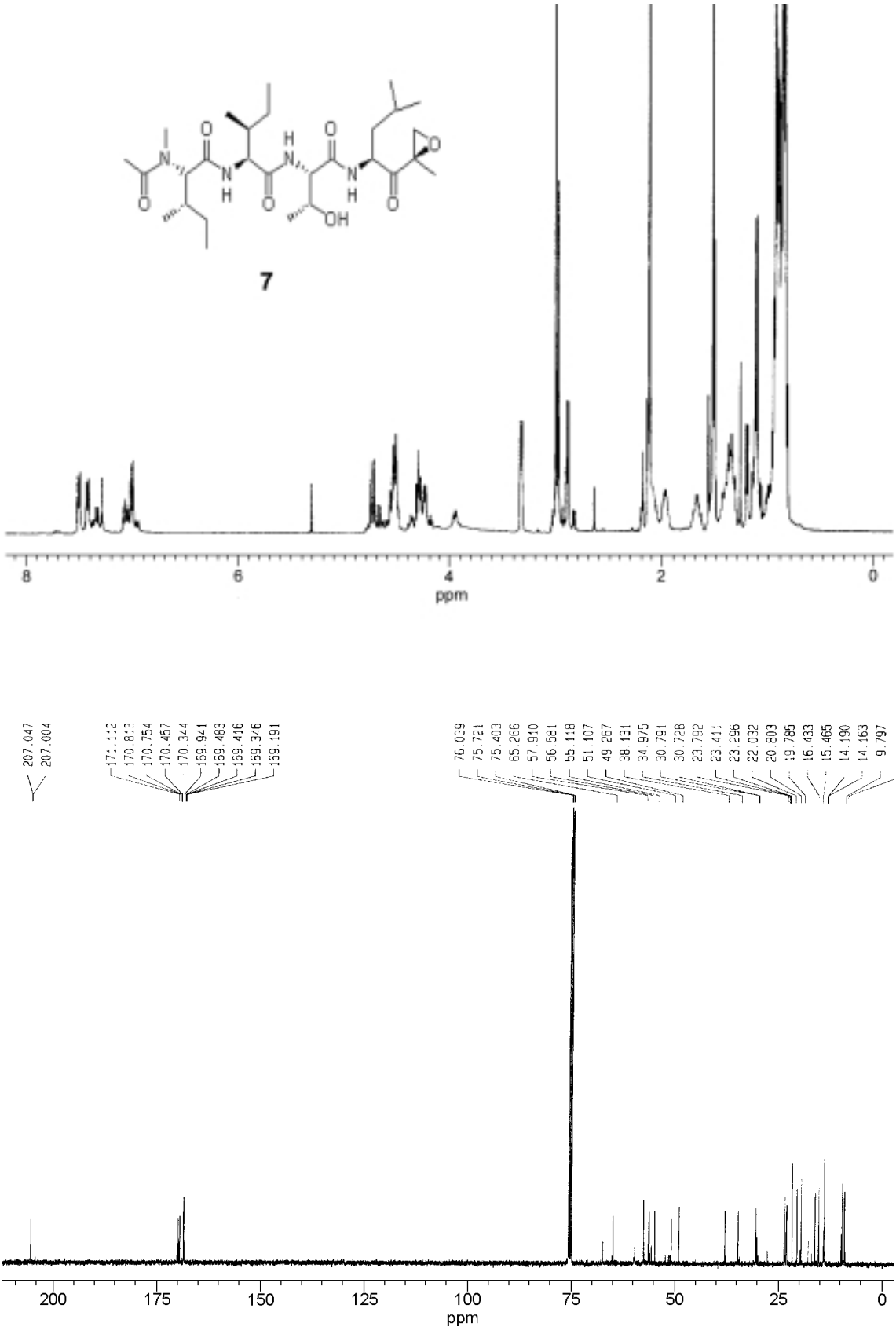
^{1}H- and ^{13}C-NMR spectra of compound **174**



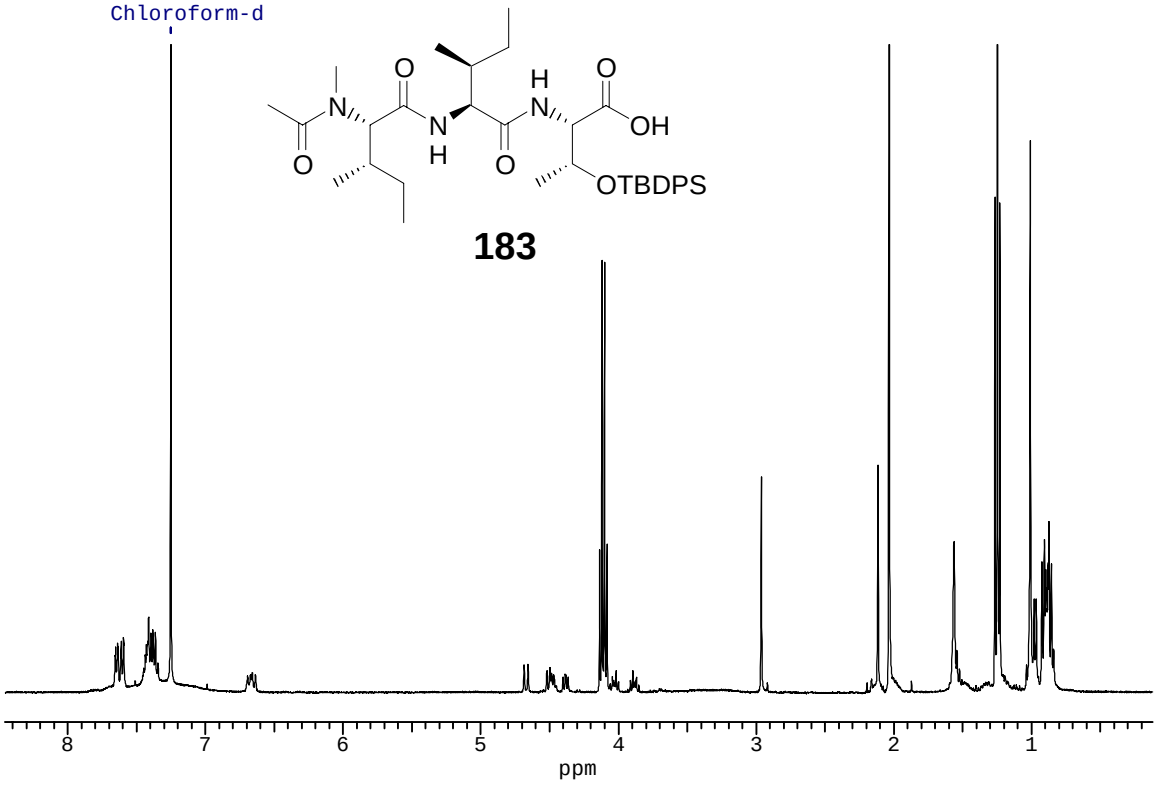
177

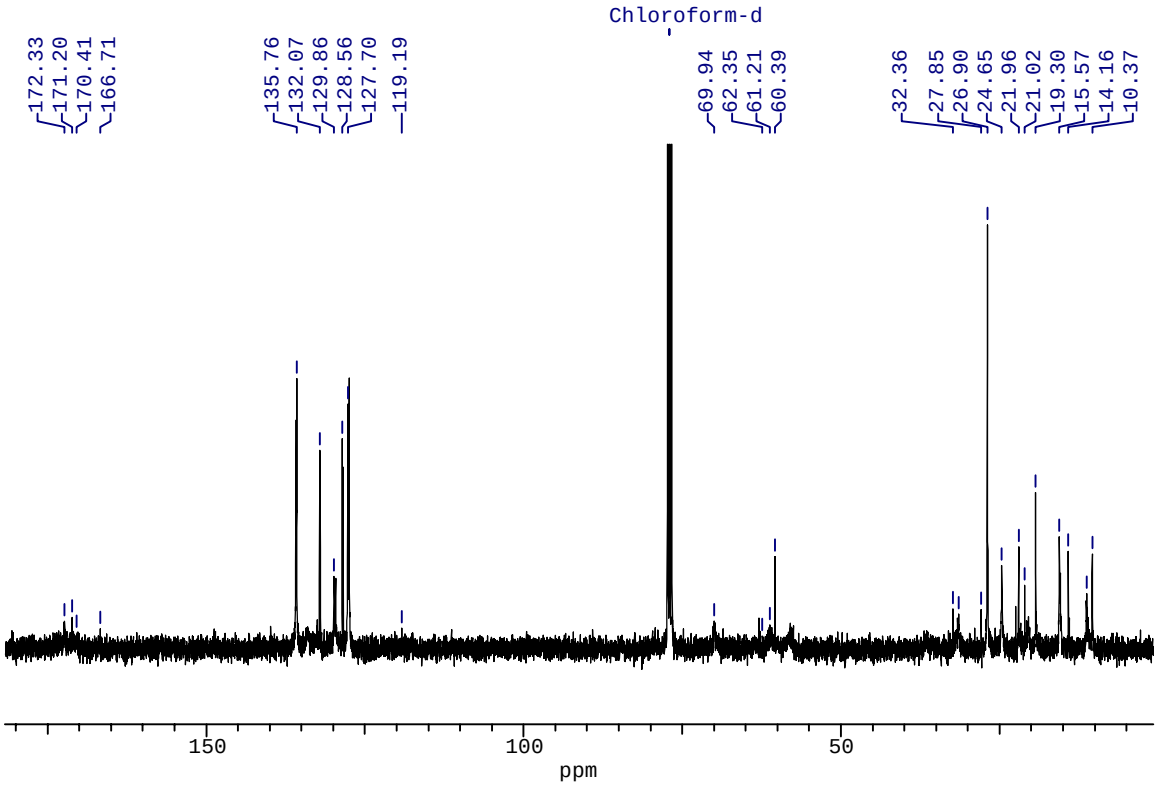


^{1}H- and ^{13}C-NMR spectra of compound **177**

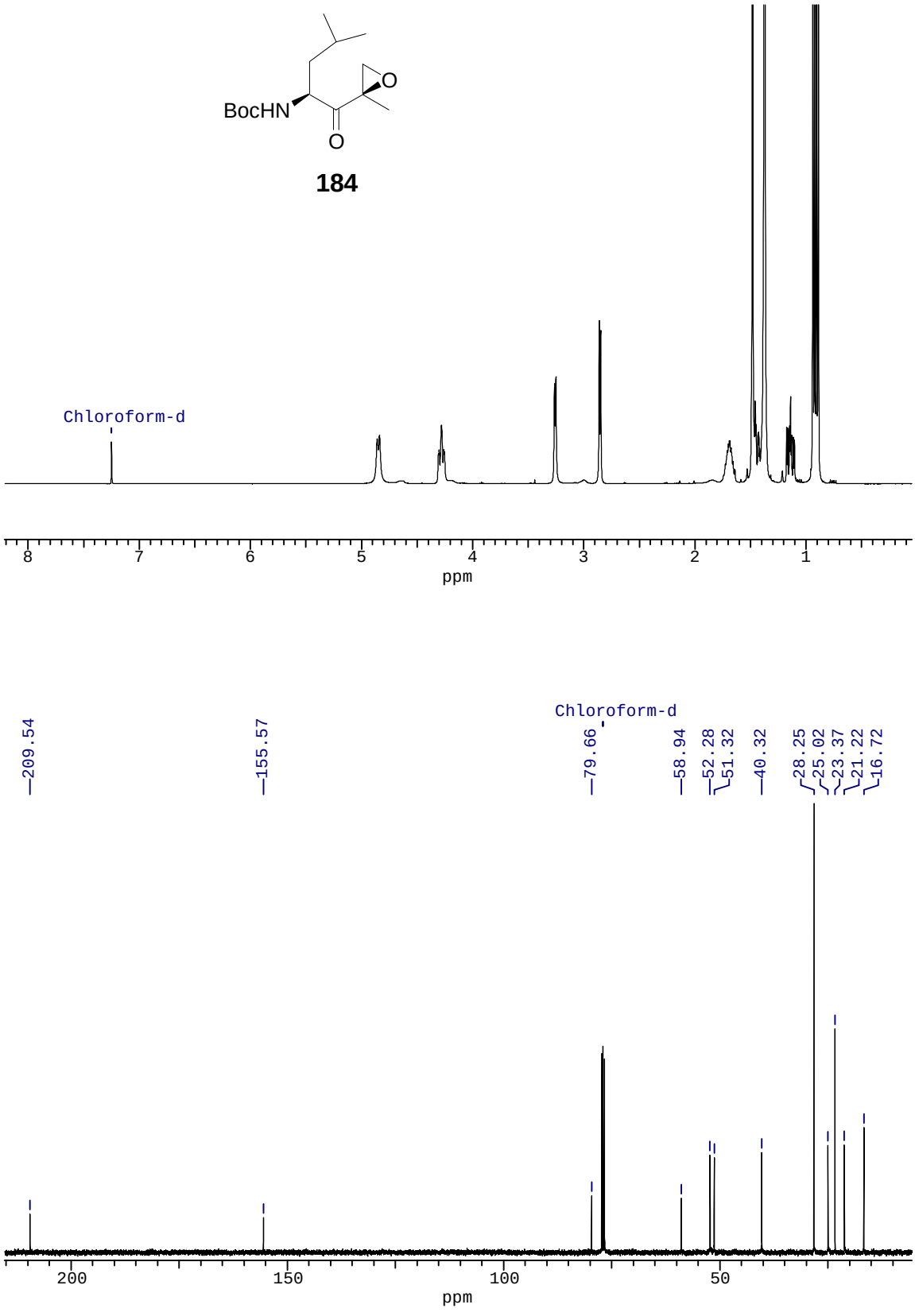


^{1}H- and ^{13}C-NMR spectra of compound **7**

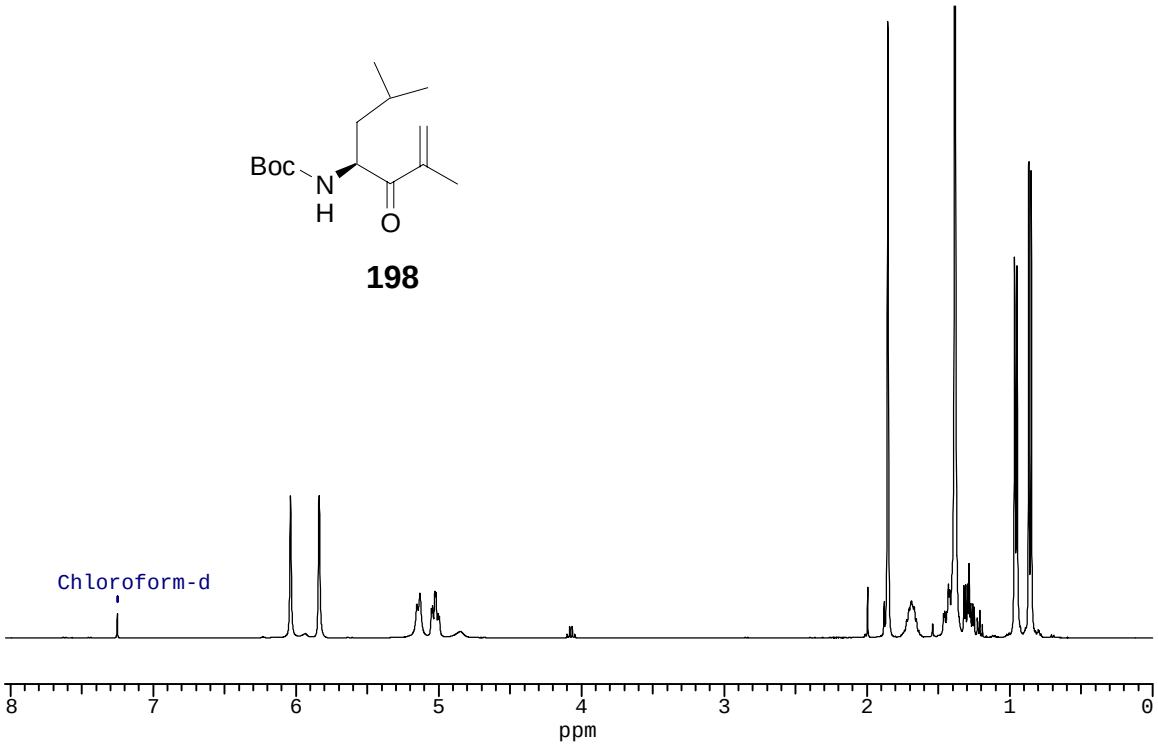


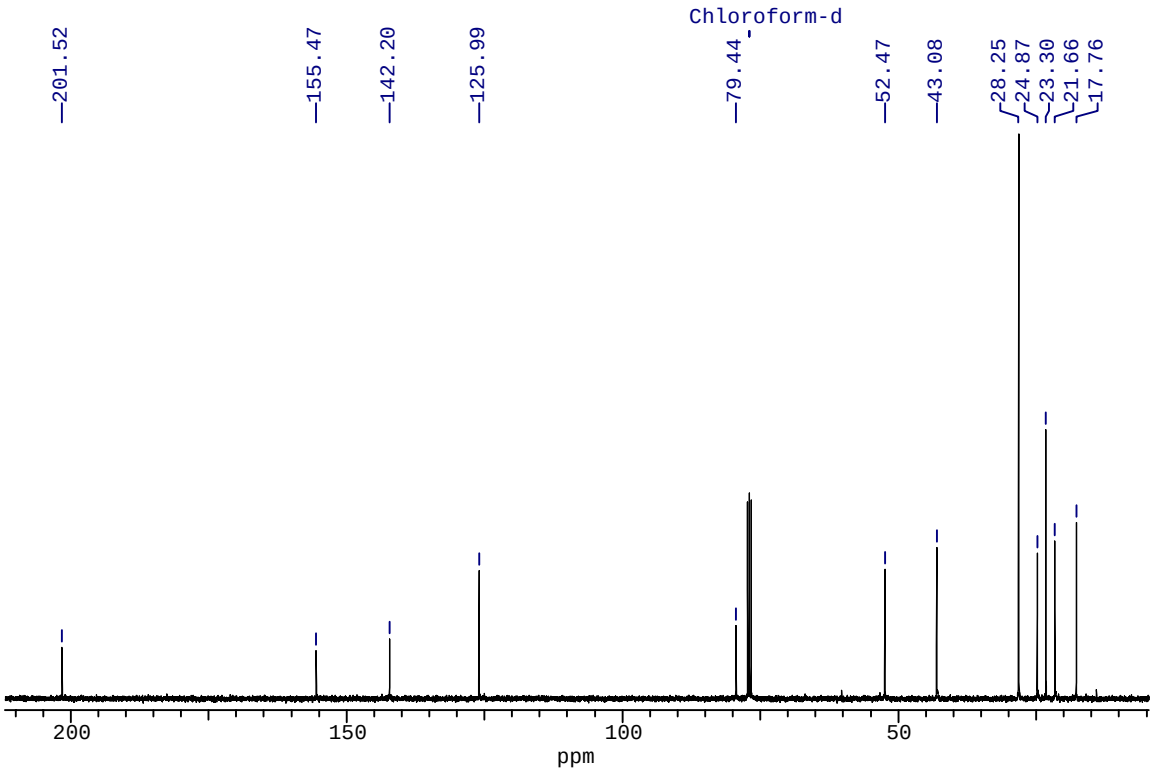


^{1}H- and ^{13}C-NMR spectra of compound **183**

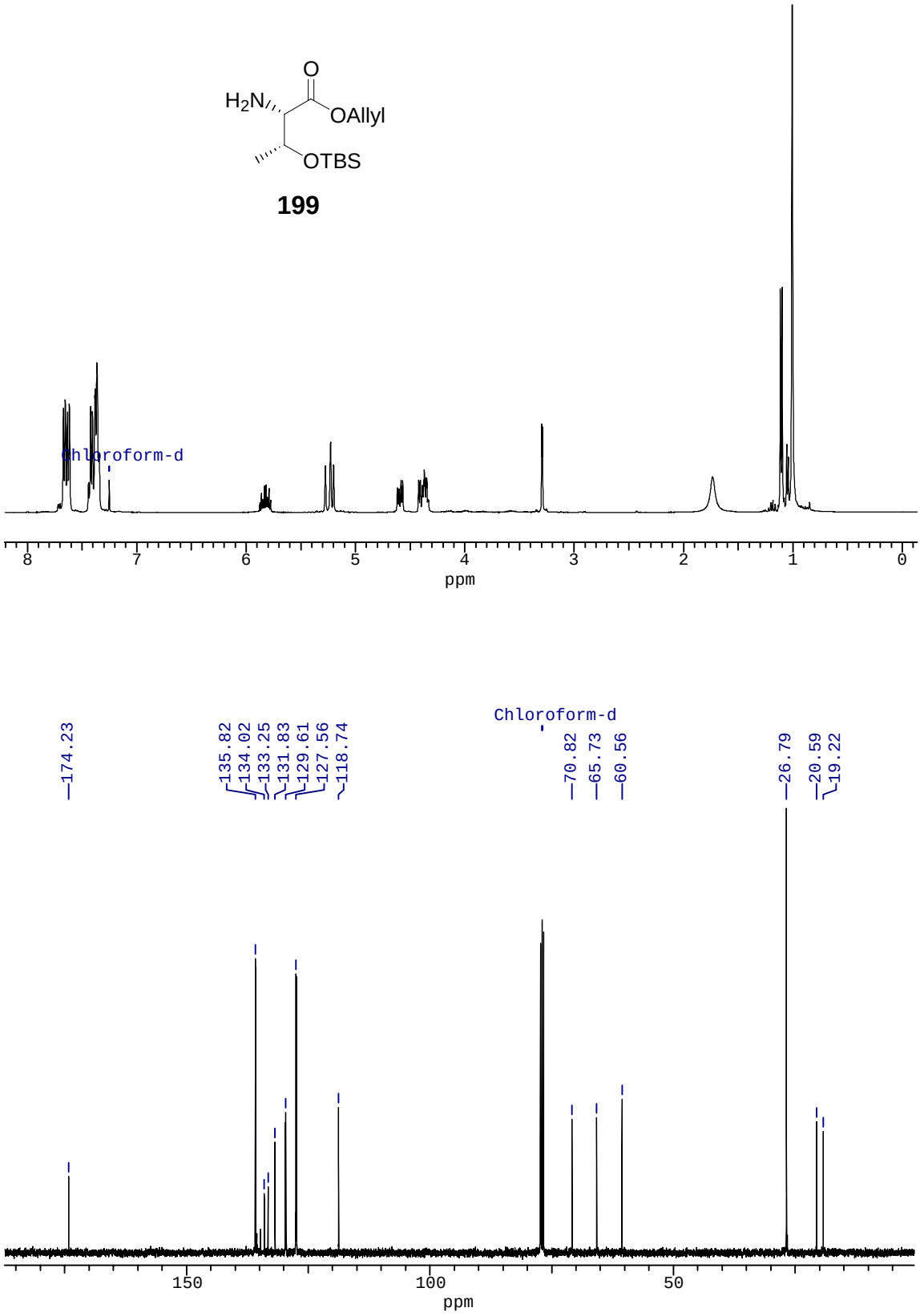


^{1}H- and ^{13}C-NMR spectra of compound **184**

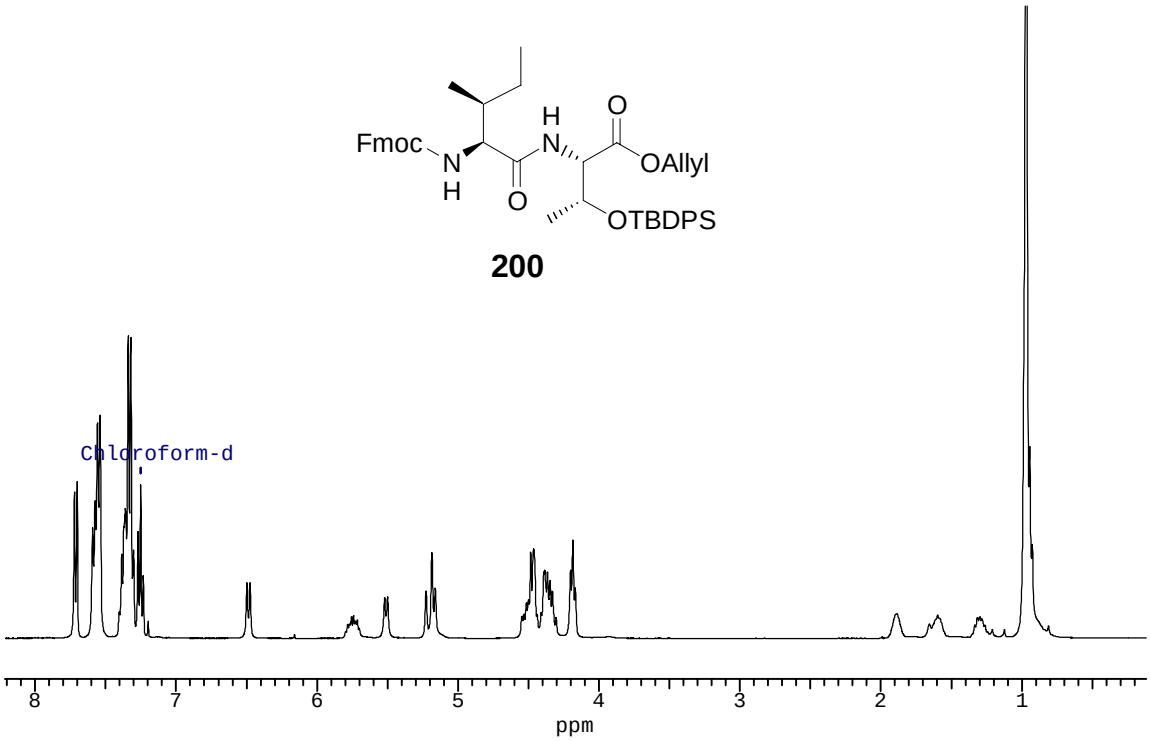


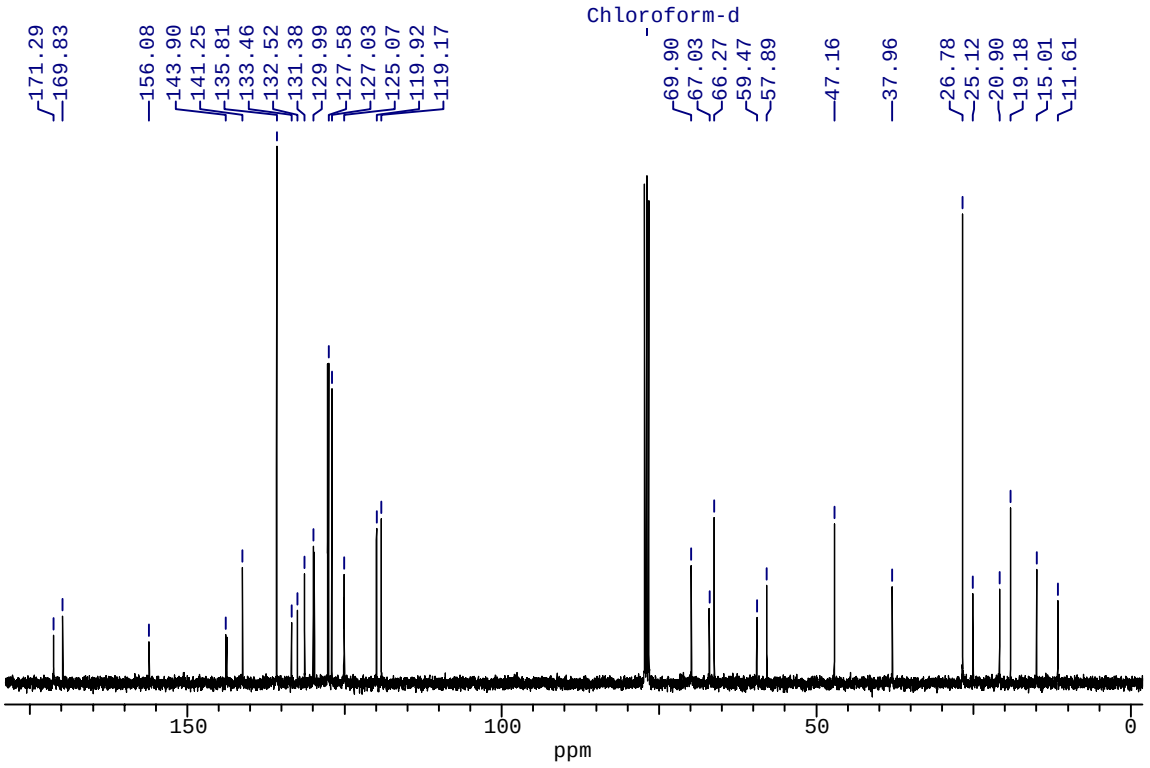


^{1}H- and ^{13}C-NMR spectra of compound **198**

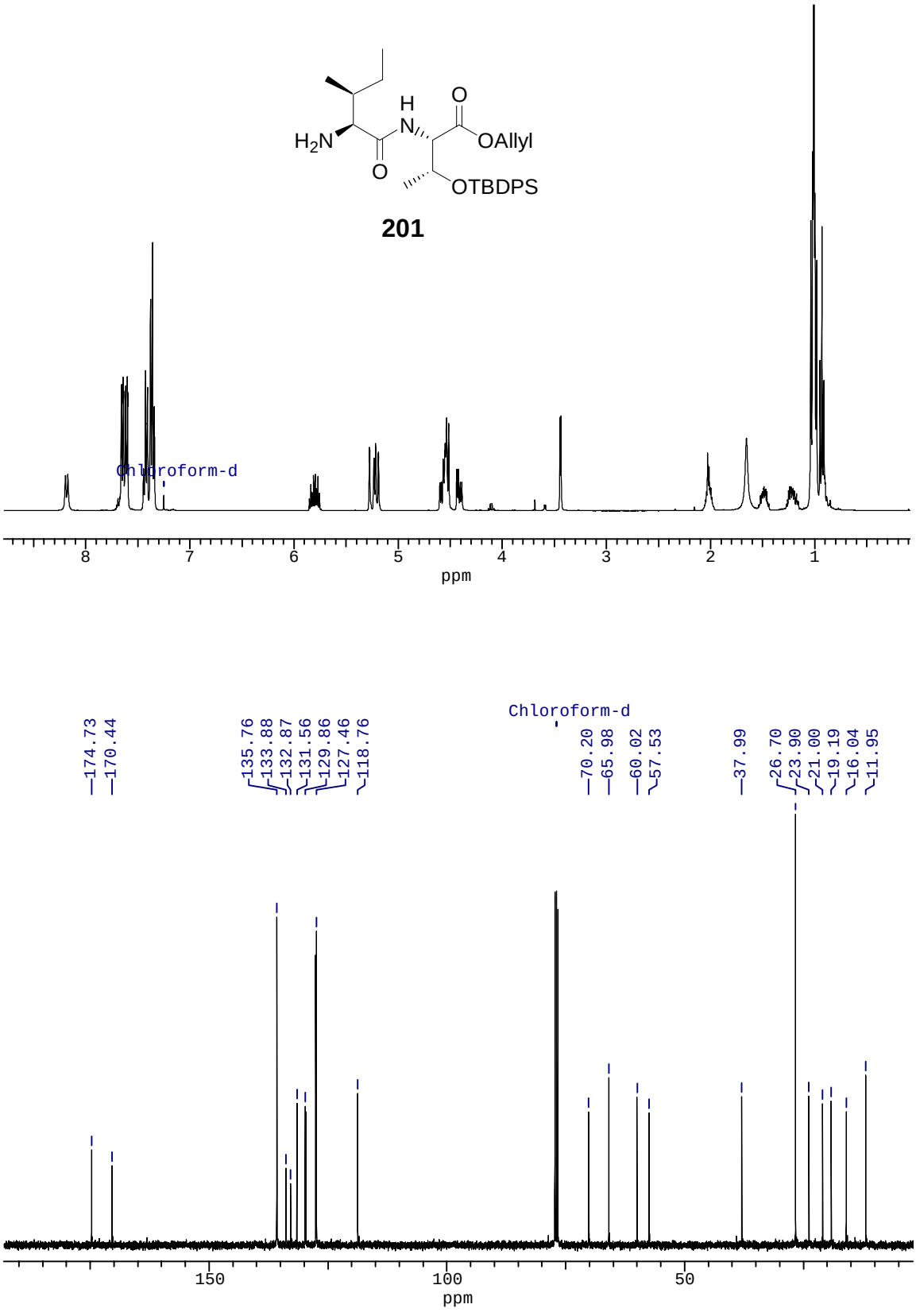


^{1}H- and ^{13}C-NMR spectra of compound **199**

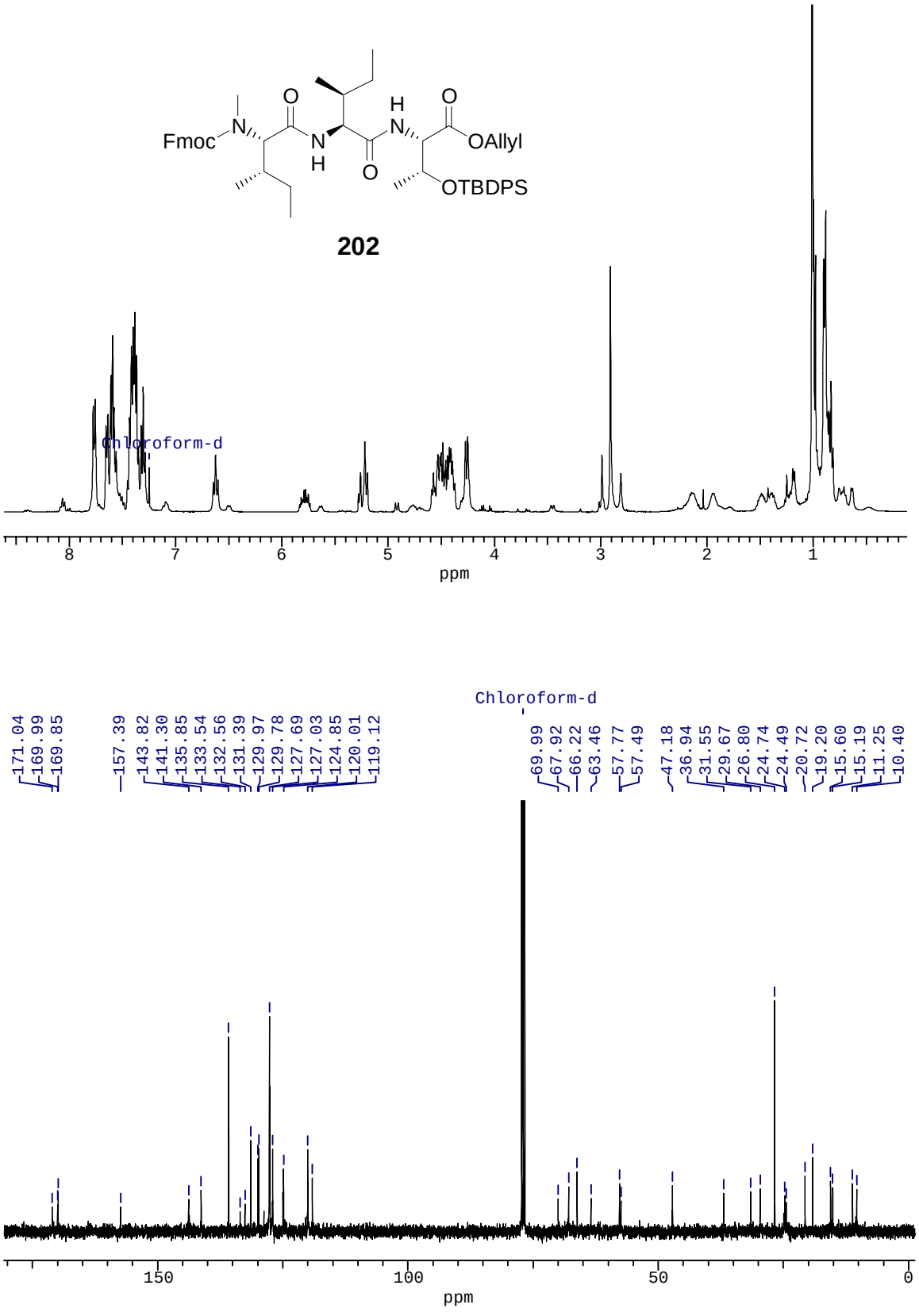




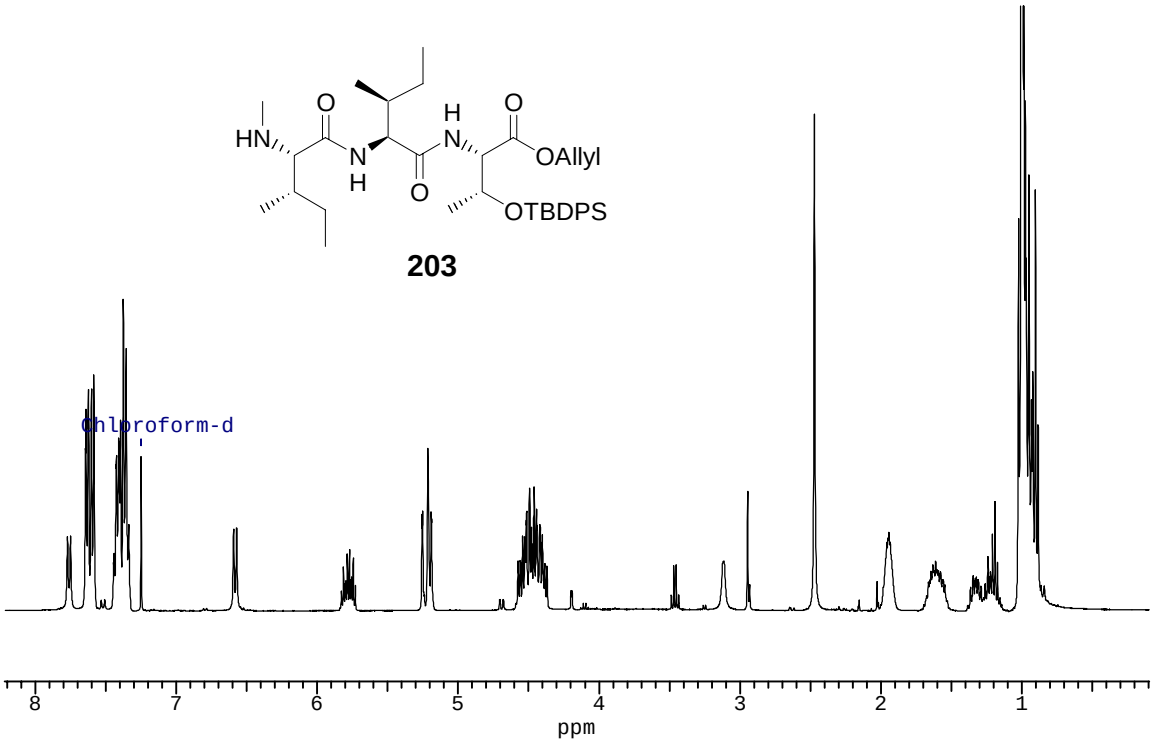
^{1}H- and ^{13}C-NMR spectra of compound **200**

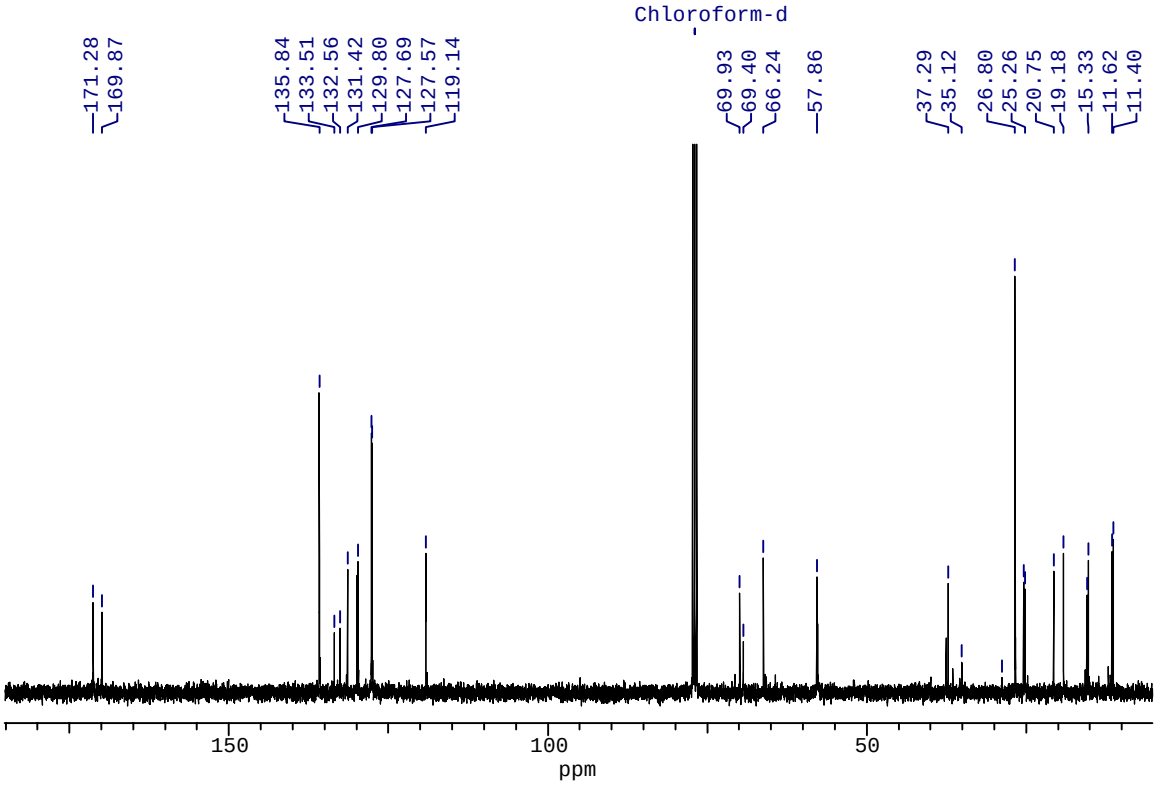


^{1}H- and ^{13}C-NMR spectra of compound **201**

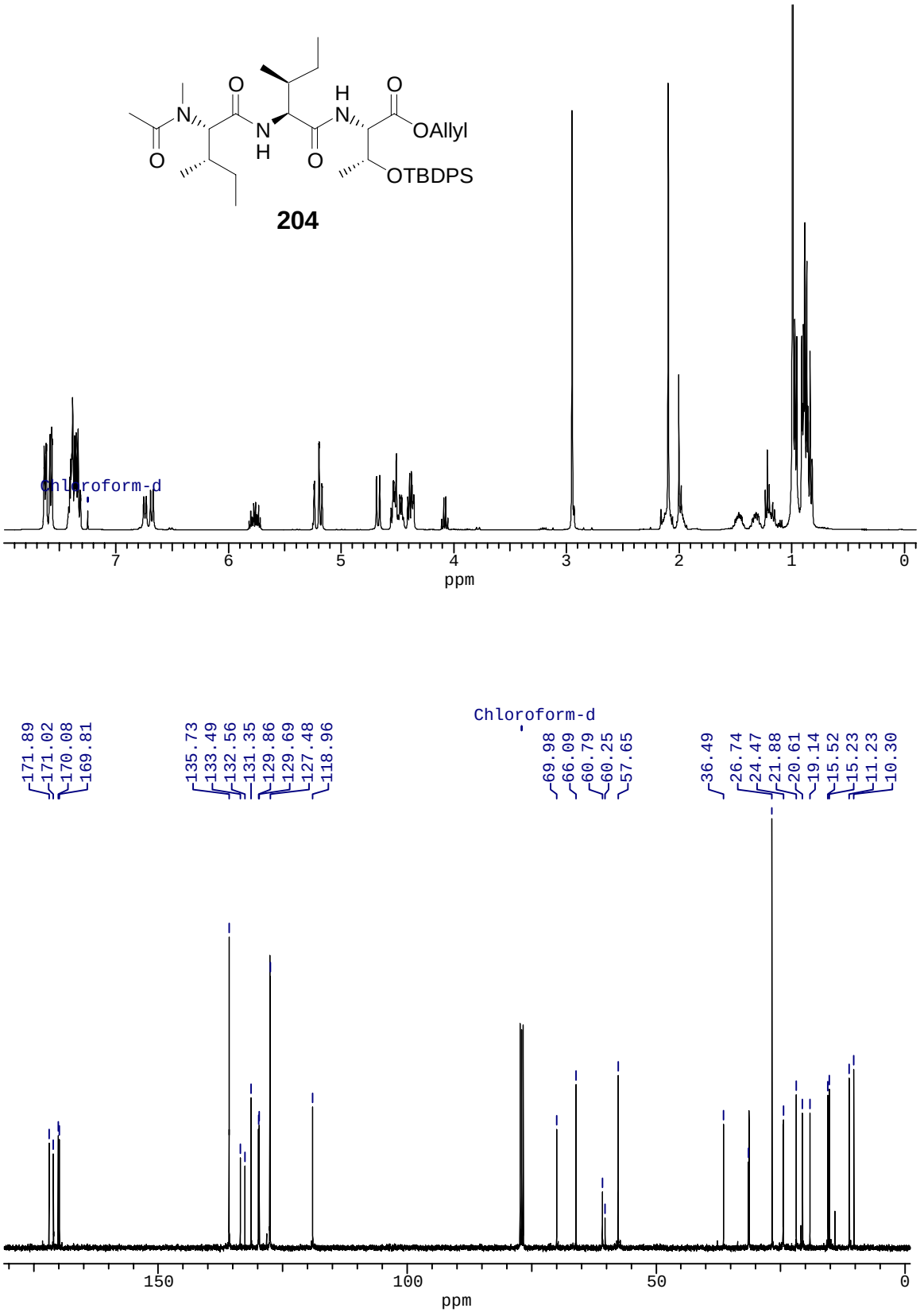


^{1}H- and ^{13}C-NMR spectra of compound **202**

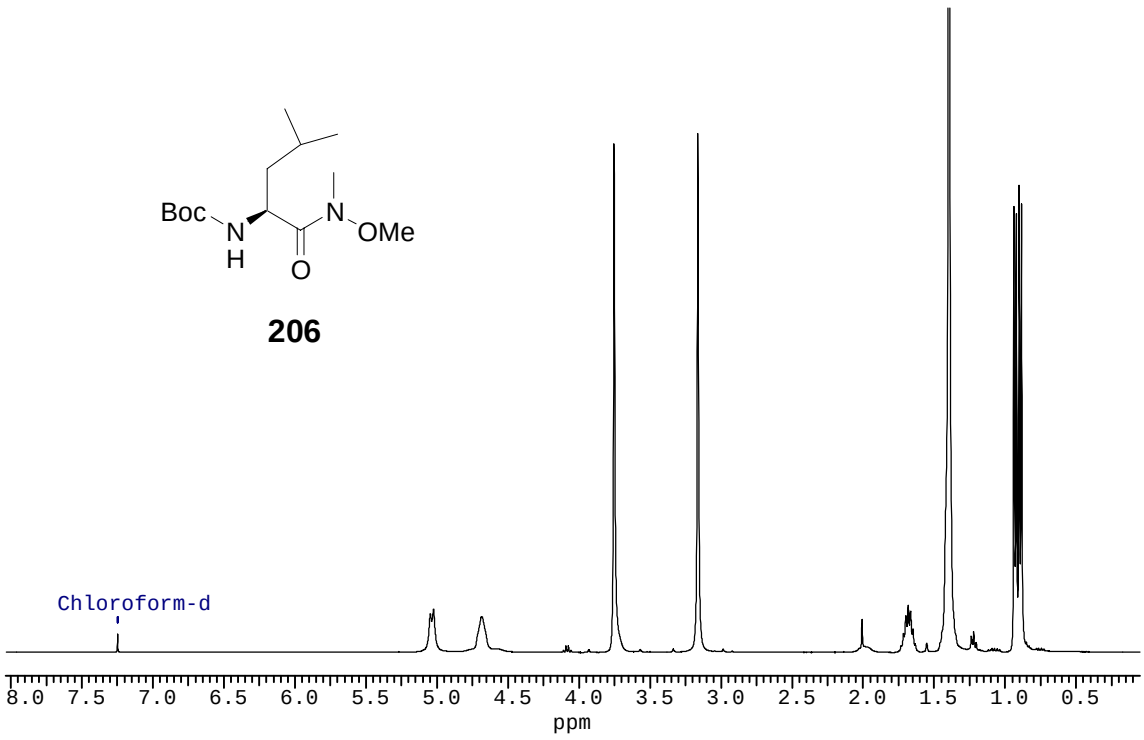


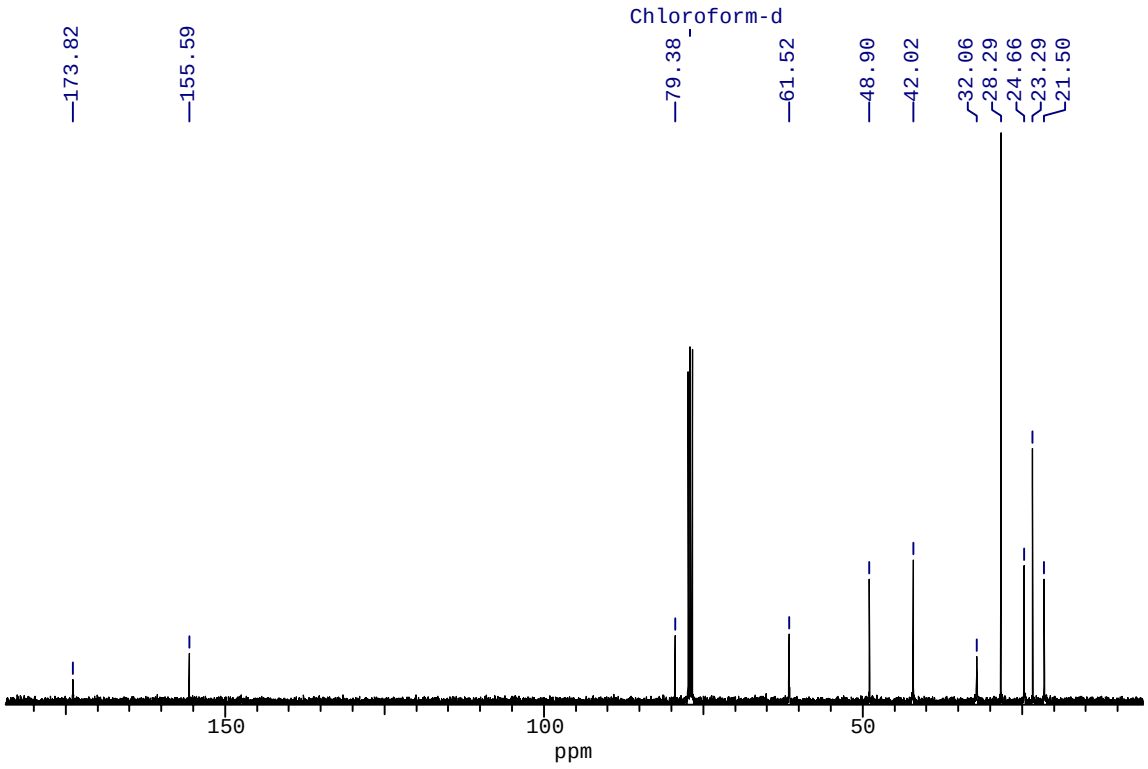


^{1}H- and ^{13}C-NMR spectra of compound **203**

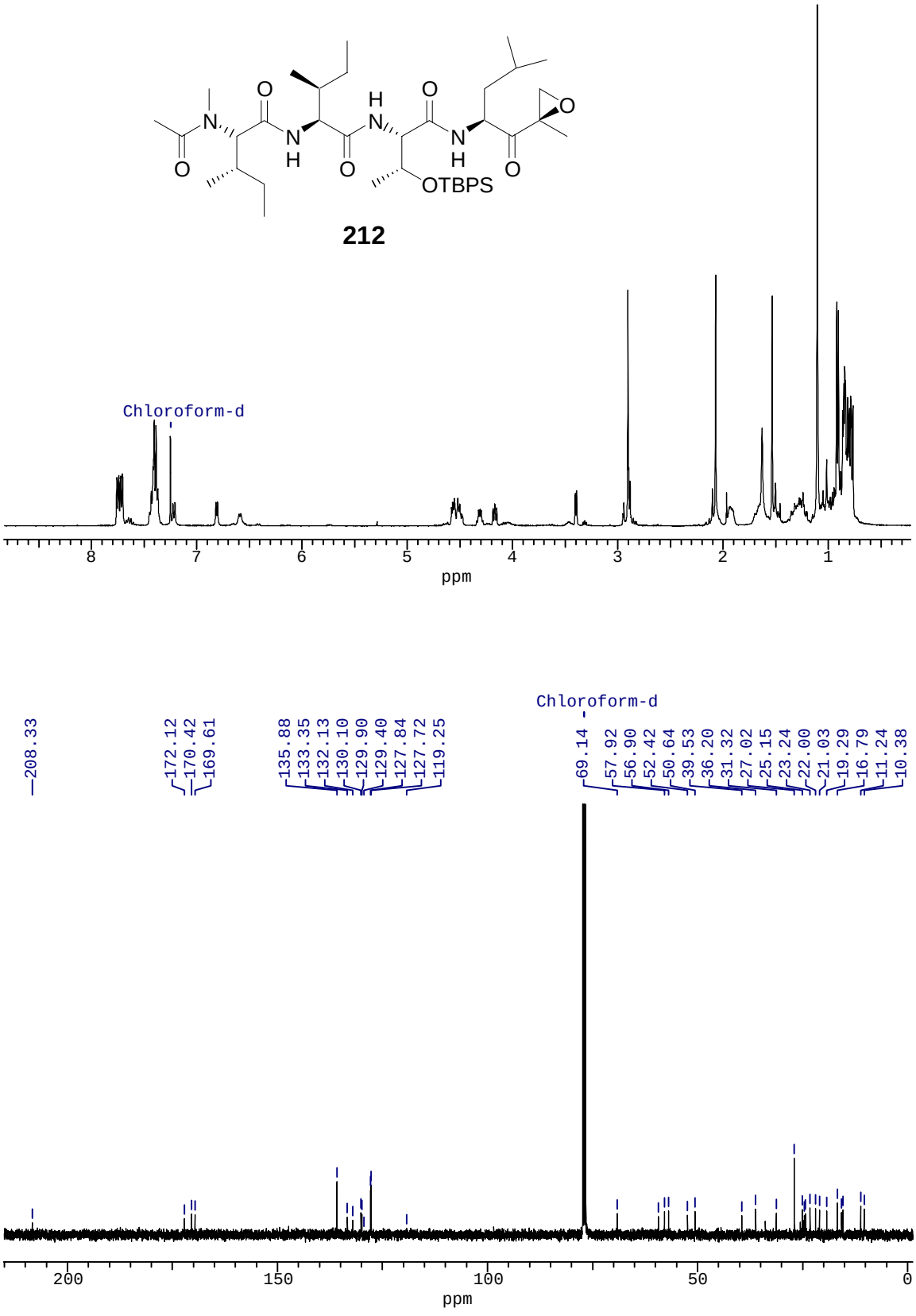


^{1}H- and ^{13}C-NMR spectra of compound **204**





^{1}H- and ^{13}C-NMR spectra of compound **206**



^{1}H- and ^{13}C-NMR spectra of compound **212**

2. References

1. D. Tan, M. A. Foley, B. R. Stockwell, M. D. Shair, S. L. Schreiber, *J. Am. Chem. Soc*. **1999**, *121*, 9073-9087
2. C. Hermann, C. Giammasi, A. Geyer, M. E. Maier, *Tetrahedron* **2001**, *57*, 8999-9010
3. C. Watson, *Angew. Chem.* **1999**, *111*, 2025-2031, *Angew. Chem. Int. Ed.* **1999**, *38*, 1903-1908
4. R. M. Khole, M. D. Bruke, J. Tao, C. T. Walsh, *J. Am. Chem. Soc*. **2003,** *125*, 7160-7161
5. S. L. Schreiber, *Chem. Eng. News* **2003**, 51-61
6. R. W. King, *Chem. Biol*. **1999**, *6*, R327-R333
7. S. Chen, Q. Zhang, X. Wu, P. G. Schultz, S. Ding, *J. Am. Chem. Soc*. **2004**, *126*, 410-411

8.a) R. E. Schwartz, C. F. Hirsch, D. F. Sesin, J. E. Flor, M. Chartrain, R. E. Fromtling, G. H. Harris, M. J. Salvatore, J. M. Liesch, K. Yudin, *J. Ind. Microbiol*. **1990**, *5*, 113-123

b) G. Trimurtulu, I. Ohtani, G. M. L. Patterson, R.E. Moore, T. H. Corbett, F. A. Valeriote, L. Demchik, *J. Am. Chem. Soc*. **1994**, *116*, 4729-4737

c) R. E. Moore, *J. Indust. Microbiol.* **1996**, *16*, 134-143

d) A. Jordan, J. A. Hadfield, N. J. Lawrence, A. T. McGown, *Med. Res. Rev.* **1998**, *18*, 259-296

e) C. Shih, B. A. Teicher, *Curr. Pharmacet. Des.* **2001**, *7*, 1259-1276

f) M. A. Tius, *Tetrahedron* **2002**, *58*, 4343-4367

g) T. Li, C. Shih, *Front. Biotechnol. Pharmaceut.* **2002**, *3*, 172-192

h) M. Eggen, G. I. Georg, *Med. Res. Rev.* **2002**, *22*, 85-101

i) S. Eißler, A. Stoncius, M. Nahrwold, N. Sewald, *Synthesis* **2006**, 3747-3789

9. M. Kobayashi, W. Wang, N. Ohyabu, M . Kurosu, I. Kitagawa, *Chem. Pharm. Bull*. **1995,** *43*, 1598-2198
10. D. Panda, K. DeLuca, D. Williams, M. A. Jordan, L. Wilson, *Proc. Natl. Acad. Sci.* U. S. A. **1998**, *95*, 9313-9318
11. K. Kerksiek, M. Mejillano, R. E. Schwartz, G. I. Georgy, R. Himes*, FEBS Lett*. **1995**, *377*, 59-61

12. S. L. Mooberry, L. Busquets, G. Tien. *Intl. J. Cancer* **1997**, *73*, 440-448
13. D. Panda, R. H. Himes, R. E. Moore, L. Wilson, M. A. Jordan, *Biochemistry* **1997**, *36*, 12048-12953
14. D. Kessel, *J. Chromatogr B.* **1999**, *735*, 121
15. a) A. Pandey, M. Mann, *Nature* **2000**, *405*, 837-846
    b) N. L. Anderson, N. G. Anderson, *Electrophoresis* **1998,** *19*, 1853-1861
16. a) D. T. Hung, T. F. Jamison, S. L. Schreiber, *Chem. Biol*. **1996**, *3*, 623-639
    b) R. F. Standaert, A. Galat, G. L. Verdine, S. L. Schreiber, *Nature* **1990**, *346*, 671-674
17. P. Hadvary, W. Sidler, W. Meister, W. Vetter, H. Wolfer*, J. Biol. Chem*. **1991**, *266*, 2021-2027
18. W. T. Lowther, D. A. McMillen, A. M. Orville, B. W. Matthews, *Proc. Natl. Acad. Sci*. USA. **1997**, *94*, 6099-6103
19. C. Drahl, F. C. Benjamin, E. J. Sorensen, *Angew Chem. Int. Ed.* **2005**, *44*, 5788-5809
20. N. Sin, K. B. Kim, M. Elofsson, L. Meng, H. Auth, H. B. Benjamin, Kwok, M. C. Crews, *Bioorg. Med. Chem. Lett*. **1999**, *9*, 2283-2288
21. M. Hanada, K. Sugawara, K. Kaneta, S. Toda, Y. Nishiyama, K. Tomita, H. Yamamoto, M. Konishi, T. Oki, *J. Antibiot*. **1992**, *45*, 1747-1752
22. K. Sugawara, M. Hatori, Y. Nishiyama, K. Tomita, H. Kamei, M. Konishki, T. Oki, *J. Antibiot.* **1990**, *43*, 8-11
23. T. H. Corbett, F. A. Valeriote, L. Demchik, L. Polin, C. Panchapor, S. Pugh, K. White, J. Knight, J. Jones, L. Jones, P. LoRusso, B. Foster, R. A. Wiegand, L. Lisow, T. Golakoti, C. E. Helzel, J. Ogino, G. M. Patterson, R. E. Moore, *J. Exp. Ther. Oncol*. **1996**, *1*, 95-108
24. R. A. Barrow, T. Hemscheidt, J. Liang, S. Paik, R. E. Moore, M. A. Tius, *J. Am. Chem. Soc.* **1995**, *117*, 2479-2490
25. C. D. Smith, X. Zhang, S. L. Mooberry, G. M. L. Patterson, R. E. Moore, *Cancer Res.* **1994**, *54*, 3779-3784
26. M. Kabayashi, S. Aoki, N. Ohyabu, M. Kuroso, W. Wang, I. Kitigawa, *Tetrahydron Lett*. **1994**, *35*, 7969-7972
27. S. B. Buck, J. K. Huff, R. H. Himes, G. I. Georg, *J. Med. Chem*. **2004**, *47*, 696-702
28. K. Morita, Y. Koiso, Y. Hashimoto, M. Kabayashi, W. Wang, N. Ohyabu, S. Iwasaki, *Biol. Pharm. Bull*. **1997**, 20, 171-174

29. V. F. Patel, S. L. Andis, J. H. Kennedy, J. E. Roy, R. M. Schultz, *J. Med. Chem.* **1999**, *42*, 2588-2603
30. R. S. Al-Awar, J. E. Ray, R. M. Schultz, S. L. Andis, J. H. Kennedy, R. E. Moore, T. Golakoti, G. V. Subbaraju, T. H. Corbett, *J. Med. Chem*. **2003**, *46*, 2985-3007
31. M. M. Wagner, D. C. Paul, C. Shih, M. A. Jordan, L. Wilson, D. C. Willums, *Cancer Chemother. Pharmacol*. **1999**, *43*, 115-125
32. J. Edelman Martin, R. Gandara David, P. Husner, V. Israel, D. Thornton, J. DeSanto, L. A. Doyle, *Lung Cancer* **2003**, *39*, 197-199
33. C. D. Smith, X. Zhang, *J. Biol. Chem*. **1996,** *271*, 6192-6198
34. S. L. Moobery, C. R. Taoka, L. Busquets, *Cancer Lett*. **1996**, *107*, 53-57
35. R. Bai, R.E. Schartz, J. A. Kepler, G. R. Pettit, E. Hamel, *Cancer Res*. **1996**, *56*, 4398-4406
36. K. Lu, J. Dempsey, R. M. Schultz, C. Shih, B. A. Teicher, *Cancer Chemother. Pharmacol*. **2001**, *47*, 170-178
37. D. Kessel, Y. Luo, *Cancer Lett*. **2000**, *151*, 25-29
38. M. A. Jordan, *Curr. Med. Chem*. **2002**, *2*, 1-17
39. B. A. Teicher, P. Forler, K. Menon, V. Phares, T. Amsrud, C. Shih, *In Vivo* **2000**, *14*, 471-480
40. K. Menon, E. Alvarez, P. Forler, V. Phares, T. Amsrud, C. Shih, R. Al-Awar, B. A. Teicher, *Cancer Chemother. Pharmacol*. **2000**, *46*, 142-149
41. T. Golakoti, J. Ogino, C. E. Heltzel, T. L. Hosebo, C. M. Jensen, L.K. Larsen, G. L. M. Patterson. R. E. Moore, S. L. Mooberry, T. H. Corbett, F. A. Valeriote, *J. Am. Chem. Soc*. **1995,** *117*, 12030-12049
42. G. J. Boons, G. H. Castle, J. A. Clase, P. Grice, S. V. Ley, C. Pinel, *Synlett* **1993**, 913-914
43. R. Zeynek, Z. Hoppe-Seyler, *Physiol. Chem*. **1926**, *144*, 1-247
44. P. D. Jefferey, S. W. McCombie, *J. Org. Chem*. **1982**, *47*, 587-590
45. K. M. Gardinier, J. W. Leahy, *J. Org. Chem*. **1997**, *62*, 7098-7099
46. D. A. Evans, S. W. Koldor, T. K. Jones, J. Clardy, T. J. Stout, *J. Am. Chem. Soc*. **1990**, *112*, 7001-7031
47. D. A. Evans, A. H. Hoveyda, *J. Am. Chem. Soc.* **1990**, *112*, 6447-6449
48. H. Hoiki, M. Okuda, W. Miagi, S. Oto, *Tetrahedron Lett.* **1993**, *34*, 6131-6134
49. J. Inanaga, K. Hirata, H. Saeki, T. Katsuki, M. Yamaguchi, *Bull. Chem. Soc. Jpn.* **1979,** *52,* 1989-1993

50. A. H. Fray, *Tetrahedron: Asymmetry* **1998**, *8*, 2777-1779
51. L. H. Li, M. A. Tius, *Org*. *Lett*. **2002**, *4*, 1637-1740
52. S. J. Danishefsky, *Chemtracts* **1989**, 1-273
53. M. A. Tius, J. Petersen, *Synlett* **1997**, 531-533
54. G. M. Salamonczyk, K. Han, Z-W. Guo, C. Sih, *J. Org. Chem*. **1996**, *61*, 6893-6900
55. D. L. Varie, J. Benjamin, B. Briggs, J. S. Cronin, D. A. Hay, J. A. Rieck, M. J. Zmijewski, *Tetrahedron Lett*. **1998**, *39*, 8405-8408
56. a) S. L. Schreiber, W.-F. Liew, *Tetrahedron Lett*. **1983**, *24*, 2363-2366
    b) H. J. Swarts, A. A. Verstegen-Hacksma, B. J. M. Jansen, A. deGroot, *Tetrahedron* **1994**, *43*, 251-256
57. G. R. Krow, *Org. React*. **1993**, *43*, 251-775
58. T. Nakai, K. Mikami, *Org. React*. **1994**, *46*, 105-207
59. B. Morgan, A. C. Oelschlager, T. M. Stokes, *J. Org. Chem*. **1992**, *57*, 3231-3236
60. Y. Veysoglu, L. A. Mitscher, J. K. Swayze, *Synthesis* **1980**, 807-810
61. H. C. Brown, K. S. Bhat, R. S. Randad, *J. Org. Chem*. **1989**, *54*, 1570-1576
62. H. C. Brown, K. S. Bhat, *J. Am. Chem. Soc*. **1986**, *108*, 5919-5923
63. H. C. Brown, N. N. Joshi, *J. Org. Chem*. **1988**, *53*, 4059-4062
64. U. P. Dhokte, V. V. Khau, D. R. Hutchison, M. J. Martinelli, *Tetrahedron Lett*. **1998**, *39*, 8771-8774
65. P. Anelli, F. Montanari, S. Quici, *Org. Synth*. **1990**, *69*, 212-215
66. J. D. White, J. Hong, L. A. Robarge, *J. Org. Chem*. **1999**, *64*, 6206-6216
67. J. D. White, J. Hong, L. A. Robarge, *Tetrahedron Lett*. **1998**, *39*, 8779-8782
68. A. Johns, J. A. Murphy, M. S. Sherburn, *Tetrahedron* **1989**, *45*, 7835-7857
69. C. P. Chuang, D. J. Hart, *J. Org. Chem*. **1983**, *48*, 1782-1784
70. K. Takai, K. Nitta, K. Utimoto, *J. Am. Chem. Soc*. **1986**, *108*, 7408-7410
71. J. K. Stille, B. Groh, *J. Am. Chem. Soc*. **1987**, *109*, 813-817
72. M. Eggen, C. Mossman, S. B. Buck, S. K. Nair, L. Bhat, S. M. Ali, E. A. Reiff, T. C. Boge, I. G. George, *J. Org. Chem.* **2000**, *65*, 7792-7799
73. R. Noyori, *Asymmetric Catalysis in Organic Synthesis*, Wiley, New York, **1994**
74. G. Fra'ter*, Helv. Chim. Acta* **1979**, *62*, 2825-2828
75. S. V. Ley, J. Norman, W. P. Griffith, *Synthesis* **1994**, 639-666
76. R. Rej, D. Nguyen, B. Go, S. Fortin, J. F. Lavalée, *J. Org. Chem.* **1996**, *61*, 6289-6295
77. M. Furuyama, I. Shimizu, *Tetrahedron: Asymmetry* **1998**, *9*, 1351-1357

78. P. Pukan, S. Sasmal, M. E. Maier, *Eur. J. Org. Chem.* **2003**, 1733-1740
79. F. Martin, J. A. Dodge, *Tetrahedron Lett.* **1991**, *32*, 3017-3032
80. A. K. Ghosh, L. Swanson, *J. Org. Chem.* **2003**, *68*, 9823-9826
81. J. E. Corey, R. K. Bakshi, S. Sibata, C. Chen V. K: Singh, *J. Am. Chem. Soc*. **1987**, *109*, 7925-7926
82. R. Barrow, R.E. Moore, L. H. Li, M. A. Tius, *Tetrahedron* **2000**, *56*, 3339-3351
83. S. Masamune, W, Choy, F. A. J. Kerdesky, B. Imperiali, *J. Am. Chem. Soc*. **1981**, *103*, 1566-1568
84. D. A. Evans, J. V. Nelson, E. Vogel, T. R. Taber, *J. Am. Chem. Soc*. **1981**, *103*, 3099-3111
85. S. M. Makin, Y. E. Raifel's, O. V. Limanova, O. A. Shavrygina, L. M. Kosheleva, *J. Org. Chem*. *USSR* **1979**, 1665
86. B. M. Trost, J. Dumas, M. Villa, *J. Am. Chem. Soc*. **1992**, *114*, 9836-9845
87. A. L. Gemal, J. L. Luche, *J. Am. Chem. Soc*. **1981**, *103*, 5454-5459
88. R. Vidya, M. J. Eiggen, S. K. Nair, G. I. Georg, R. H. Rimes, *J. Org. Chem.* **2003**, *68*, 9687-9693
89. M. J. Eiggen, S. K. Nair, G. I. Georg, *Org. Lett,* **2001**, *3*, 1813-1815
90. N. K. Tripathy, G. I. Georg, *Tetahedron Lett*. **2004**, *45*, 5309-5311
91. P. Schwab, R. H. Grubbs, J. W. Ziller, *J. Am. Chem. Soc*. **1996**, *118*, 100-110
92. B. H. Norman, T. Schultz, M. Richard, S. L. Andis, *J. Org. Chem*. **1998**, *63*, 5288-5294
93. J. A. Christopher, P. J. Kocienski, A. Kuhl, R. Bell, *Synlett* **2000**, 463-466
94. C. Pousset, M. Haddad, M. Larchevéque, *Tetrahedron* **2001**, *57*, 7163-7167
95. F. Velazquez, H. F. Olivo, *Curr. Org. Chem.* **2002**, *6*, 303-340
96. A. T. Nielsen, W. J. Houlihan, *Org. React.* **1968**, *16*, 1-438
97. D. A. Evans, J. Bartroli, T. L. Shih, *J. Am. Chem. Soc.* **1981**, *103*, 2127-2109
98. W. Oppolzer, J. Blagg, I. Rodriguez, E. Walther, *J. Am. Chem. Soc.* **1990**, *112*, 2767-2772
99. Y. Nagao, S. Yamada, T. Kumagai, M. Ochiai, E. Fujita, *J. Chem. Soc. Chem.* **1981**, *103*, 2127-2109
100. M. T. Crimmins, B. W. King, E. A. Tabet, K. Chaudhary, *J. Org. Chem.* **2001**, *66*, 894-902
101. B. E. Maryanoff, A. B. Reitz, B. A. Duhl-Emswiler, *J. Am. Chem. Soc.* **1985**, *107*, 217-226

102. D. L. Boger, The Scripps Research Institute, in *Modern Organic Synthesis*, 1st. ed (Eds. R. M. Garbaccio), TSRI Press, La Jolla, CA, **1999**, 359
103. T. Bruckdorfer, O. Marder, F. Albericio, *Curr. Pharmaceut. Biotech.*, **2004**, *5*, 29-43
104. R. C. Larock, *Comprehensive Organic Transformation*; Wiley-VCH: NewYork, London, **1999**
105. *Comprehensive Organic Synthesis*: B. M. Trost, I. Fleming, Eds: Pergamon Press: Oxford, U. K. **1991**, *6*, 1-309
106. *Comprehensive Organic Group Transformation*: A. R. Katritzky, O. Meth-Kohn, C. W. Rees, Eds; Pergamon Press: Oxford, U. K. **1995**, *5*, 1-182
107. K. Ishihara, S. Ohara, H. Yamamoto, *Science* **2000**, *290*, 1140-1144
108. L. Katz, *Chem. Rev*. **1997**, *97*, 2557-2575
109. C. Khosla, *Chem. Rev*. **1997**, *97*, 2577-2590
110. J. Staunton, K. J. Weissman, *Nat. Prod. Rep*. **2001**, *18*, 1-380
111. J. F. Aporicio, M. V. Mendes, N. Anton, R. E. Rico, J. F. Martin, *Curr. Med. Chem*. **2004**, *11*, 1645-1659
112. A. S. Willams, *Synthesis* **1999**, 1707-1709
113. E. J. Corey, K. C. Nicolaou, *J. Am. Chem. Soc*. **1974**, *96*, 5614-5616
114. T. Mukaiyama, *Angew. Chem.* **1979**, 91, 798-812, *Angew. Chem. Int. Ed.* **1979**, *18*, 707-710
115. R. H. Wollenberg, J. S. Nimitz, D. Y. Gokcek, *Tetrahedron Lett.* **1980**, *21*, 2791-2794
116. E. J. Corey, D. J. Brunelle, P. J. Stork, *Tetrahedron Lett*. **1976**, *17*, 3405-3408
117. T. Kitahara, K. Mori, *Tetrahedron* **1984**, *40*, 2935-2944
118. D. F. Taber, L. J. Silberberg, E. D. Robunson, *J. Am. Chem. Soc*. **1991**, *113*, 6639-6645
119. E. J. Corey, P. Carpino, *Tetrahedron Lett*. **1990**, *31*, 7555-7558
120. K. Tatsuta, Y. Amemiya, Y. Kanemura, M. Kinoshita, *Tetrahedron Lett*. **1981**, *22*, 3997-4000
121. P.A. Grieco, J. Inanga, N. H. Lin, T. Yanami, *J. Am. Chem. Soc*. **1982**, *104*, 5781-5781
122. T. Mukaiyama, M. Usui, K. Saigo, *Chem. Lett.* **1979**, 491
123. K. Narasaka, K. Maruyama, T. Mukaiyama, *Chem. Lett*. **1978**, 885-888
124. T. Kurihara, Y. Nakajima, O. Mitsunobu, *Tetrahedron Lett.* **1976**, *17*, 2455-2458
125. O. Mitsunobu, *Synthesis* **1981**, 1-28

126. D. L. Hughes, *Org. React*. **1992**, *42*, 335-669
127. D. L. Hughes, *Org. Prep. Proced. Int*. **1996**, *28*, 1-127
128. K. Ohta, O. Miyagawa, H. Tsutsui, O. Mitsunobu, *Bull. Chem. Soc. Jpn.* **1993**, *66*, 523-535
129. K. Ohta, O. Mitsunobu, *Tetrahedron Lett.* **1991**, *32*, 517-520
130. K. Justus, W. Steglich, *Tetrahedron Lett.* **1991**, *32*, 5781-5784
131. J. N. Lambert, J. P. Mitchell, K. D. Roberts, *J. Chem. Soc. Perkin Trans.1* **2001**, 471-484
132. K. C. Nicolaou, N, Winssiger, J. Pastor, F. Morphy, *Angew. Chem*. **1998**, *110*, 2677-2680; *Angew. Chem. Int. Ed*. **1998**, 37, 2534-2537
133. J. Alsina, K. C. Jensen, F. Alberico, G. Barany, *Chem. Eur. J.* **1999**, *5*, 2787-2795
134. J. R. Gage, D.A. Evans, *Org. Synth*. **1989**, *68*, 83-91
135. C. Palomo, M. Oiarbide, J. M. Garcia, *Chem. Eur. J*. **2002**, *8*, 36-44
136. P. Arya, H. Qin, *Tetrahedron*, **2000**, *56*, 917-947
137. M. Bauer, M. E. Maier, *Org. Lett.* **2002**, *4*, 2205-2208
138. W. C. Still, J. C. Barrish, *J. Am. Chem. Soc*. **1983**, *105*, 2487-2489
139. K. N. Houk, N. G. Rondan, Y.-D. Wu, J. T. Metz, M. N. Padon-Row, *Tetrahedron*, **1984**, *40*, 2257-2274
140. R. E. Ireland, D. W. Norbeck, *J. Org. Chem*. **1985**, *50*, 2198-2200
141. G. F. Griffiths, G. W. Kenner, S. W. McCombie, K. M. Smith, *Tetrahedron* **1976**, *32*, 275
142. M. P. Cooke, Jr., D. L. Burman, *J. Org. Chem.* **1982**, *47*, 4955-4963
143. D. Guiney, C. L. Gibson, C. J. Suckling, *Org. Biomol. Chem.* **2003**, *1*, 664-675
144. T. Hintermann, D. Seebach, *Helv. Chim. Acta* **1998**, *81*, 2093-2126
145. H.-G. Park, B.-S. Jeong, M.-S. Yoo, J.-H. Lee, M.-K. Park, Y.-J. Lee, M.-J. Kim, S-S. Jew, *Angew. Chem,***2002**, *114*, 3162-3164; *Angew. Chem. Int. Ed*. **2002**, *41*, 3036-3038
146. E. J. Corey, M. C. Noe, *Org. Synth*. **2002,** *41*, 3036-3038
147. T. Ooi, M. Kameda, K. Maruoka, *J. Am. Chem. Soc*. **2003,** *125*, 5139-5151
148. C. Lemaire, S, Gillet, S. Guillouet, A. Plenevaux, J, Aerts, A. Luxen, *Eur. J. Org. Chem.* **2004**, 2899-2904
149. A. H. G. Siebum, W. S. Woo, J. Raap, J. Lugtenburg, *Eur. J. Org. Chem*. **2004**, 2905-2913
150. A. Nelson, *Angew. Chem*. **1999**, *111*, 1685-1687; *Angew. Chem*. **1999**, *38*, 1538-1541

151. *Phase-Transfer Catalysis: Mechanism and Synthesis* (Ed: M. E. Halpern) American Chemical Society, Washington, DC, **1997**

152. *Handbook of Phase Transfer Catalysis* (Eds: Y. Sasson, R. Neuman), Blackie A&M. London, **1997**

153. T. Shiori, S. Arai, *in Stimulating Concepts in Chemistry* (Eds: F. Vögtle, J. F. Stoddart, M. Shibasaki), Wiley-VCH; Weinhein, **2000**, 123-143

154. M. J. O'Donnel *in Catalytic Asymmetric Synthesis* (Ed: I. Ojima), Wiley-VCH, New York, **2000**

155. S. Jew, M. Yoo, B.-S. Jeong, I. Park, H.-G. Park, *Org. Lett*. **2002,** *24*, 4245-4248

156. H. Buller-Bunz, P. J. Cruiry, *Tetrahedron Lett*. **2005**, *46*, 3839-3842

157. S. Yamazaki, Y. Yamazaki, *Bull. Chem. Soc. Jpn.* **1990**, *63*, 301-303] and *tert*-butyl-2-bromo acetate according to EP 1 207 151 A1.

158. T. Sato, K. Hanayama, T. Fujisawa, *Tetrahedron Lett*. **1988**, *29*, 2197-2200

159. A. Fürstner, C. Aissa, R. Riveiros, J. Ragot, *Angew. Chem*. **2002**, *114*, 4958-4960; *Angew. Chem. Int. Ed*. **2002**, *41*, 4763-4766

160. A. K. Ghosh, A. Bischoff, *Org. Lett*. **2000**, *2*, 1573-1576

161. J. H. V. Maarseveen, P. H. H. Hermkens, E. D. Clercq, J. Balzarini, H. W. Schreen, C. G. Kruse, *J. Med. Chem.* **1992**, *35*, 3223-3230

162. B. T. Cho, S. K. Kang, S. H. Shin, *Tetrahedron: Asymmetry* **2002**, *13*, 1209-1217

163. a) O. Munoz-Muniz, E. Juaristi, *Tetrahedron* **2003**, *59*, 4223-4230

b) G. V. Shustov, A. Rauk, *Tetrahedron: Asymmetr,* **1996**, 699-708

c) G. M. Salamonczyk, K. Han, Z.-W. Guo, C. J. Sih, *J. Org. Chem*. **1996**, *61*, 6893-6900

d) L. E. Janes, R. J. Romas, *Tetrahedron: Asymmetry* **1997**, *8*, 3719-3734

164. D. Seebach, Alois. Boog, W. B. Schweizer, *Eur. J. Org. Chem.* **1999**, *1*, 335-360

165. a) D. Saylic, E. M. Campi, A. C. Donohue, W. R. Jackson, A. J. Robinson, *Tetrahedron: Asymmetry* **2001**, *12*, 657-668

166. F. Degerbeck, B. Fransson, F. Grehn, U. Ragnarsson, *J. Chem. Soc*. *Perkin Trans*. *1*, **1993**, 11-14

167. D. Yang, B. Li, F.-F. Ng, Y.-L. Yan, J. Qu, Y.-D. Wu, *J. Org. Chem*. **2001**, *66*, 7303-7312

168. L. Meng, R. Mohan, B. H. Kwok, M. Elofsson, N, Sin, C. M. Crews, *Proc. Natl. Acad. Sci. USA,* **1999**, *96*, 10403-10408

169. M. Groll, K. B. Kim, N. Kairies, R. Huber, C. M. Crews, *J. Am. Chem. Soc*. **2000**, *122*, 1237-1238.

170. M. Groll, L. Ditzel, J. Löwe, D. Stock, M. Bochtler, H. D. Bartunik, R. Huber, *Nature* **1997**, *386*, 463-471

171. M. Groll, W, Heinemyer, S. Jager, T. Ullrich, M. Bochtler, D. Wolf, R. Huber, *Proc Natl Acad Sci. U S A,* **1999**, *96*, 10976-10983

172. S. Katukojvala, K. N. Berlett, S. D. Lotesta, L. J. Williams, *J. Am. Chem. Soc*. **2004**, *126*, 15348-15349

173. L. A. Carpino, *J. Am. Chem. Soc*. **1993**, *115*, 4397-4398

174. D. Frantz, R. Fassler, E. M. Carreira, *J. Am. Chem. Soc.* **2000**, *122*, 1806-1807

175. R. W. Murray, R. Jeyaraman, *J. Org. Chem*. **1985**, *50*, 2847-2853

176. B. Mordmüller, *Development of selective Nth-hydrolase inhibitors,* University of Tübingen, Germany

177. V. Spataro, C. Norbury, A. Harris, *Br. J. Cancer* **1998**, *77*, 448-455

178. M. Elofssen, U. Splittgerber, J. Myung, R. Mohan, C. M. Crews, *Chem. Biol*. **1999,** *6*, 811-822

179. a) Y. Tu, Z.-X. Wang, Y. Shi, *J. Am. Chem. Soc*. **1996**, *118*, 9806-9807

b) Z.-X. Wang, Y. Tu, M. Frohn, J.-R. Zhang, Y. Shi, *J. Am. Chem. Soc*. **1997**, *119*, 11224-11235

c) H. Tian, X. She, Y. Shi, *Org. Lett*. **2001**, *3*, 715-717

d) Z.-X. Xian, Y. Shi, *J. Org. Chem*. **1997,** *62*, 8622-8623

180. X.-Y. Wu, X. She, Y. Shi, *J. Am. Chem. Soc*. **2002**, *124*, 8792-8793

181. T. Raghuram, S. Vijaysaradhi, I. Singh, J. Singh, *Synth. Commun.* **1999**, *29*, 3215-3219

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