SYNTHESIS OF THE ANTIMALARIAL FLINDERSIAL ALKALOIDS

A Dissertation Presented to

the Faculty of the Department of Chemistry

University of Houston

In Partial Fulfillment

of the Requirements for the Degree

Doctor of Philosophy

By

Ravi Krishna Vallakati

May 2015

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Dedicated to my family

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ABSTRACT

The concept of mimicking nature to synthesize natural compounds has been successfully demonstrated in the synthesis of all the anti-malarial *Flindersia* alkaloids. Previous work on the biosynthesis of the related natural products borreverine and isoborreverine inspired us to synthesize racemic flinderoles A, B, and C in as few as three synthetic steps from simple and readily available materials. The development of reaction conditions for the dimerization of borrerine in the presence of acid is discussed in detail.

The progress toward the enantioselective total synthesis of flinderoles A, B, and C is also presented. The retrosynthetic plan was inspired by the biomimetic synthesis of the flinderoles, in which an acid mediated amino cyclization would form the flinderole framework. Tryptamine and 3-butyn-2-one undergo a vinylogous Pictet-Spengler reaction to give a β-carboline, which opens to a 2-enone tryptamine in the presence of base. A BINOL catalyzed enantioselective conjugate addition of a vinyl boronic acid to the 2-enone tryptamine has been achieved. A pyrrolo[a]indole unit could be connected to another indole unit by a Sonagashira coupling reaction in a convergent manner.

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ABBREVIATIONS AND ACRONYMS

app.	apparent
aq.	aqueous
$BF_3{\cdot}OEt_2$	boron trifluoride diethyl etherate
Bn	benzyl
Bu	butyl
Cbz	carboxybenzyl
˚C	degree Celcius
DCE	1,2-dichloroethane
DCM	dichloromethane
DCM	dichloromethane
DIBAL-H	diisobutylaluminumhydride
DMAP	dimethylaminopyridine
DME	1,2-dimethoxyethane
DMF	N,N-dimethylformamide
DMSO	dimethylsulfoxide
dr	diastereomeric ratio
dt	doublet of triplets
ee	enantiomeric excess
eq	equivalent
er	enantiomeric ratio
Et	ethyl
Et_2O	ether
Et_3N	triethyl amine

EtOAc	ethylacetate
EtOH	ethanol
EWG	electron withdrawing group
h	hour
H_2O	water
HPLC	high performance liquid chromatography
Hz	hertz
I_2	iodine
IBX	2-iodoxybenozoicacid
IC_{50}	50% inhibitory concentration
IR	infrared
i-Pr	isopropyl
J	coupling constant
L	liter
LAH	lithium aluminum hydride
LDA	lithium di-isopropyl amide
m	multiplet or milli
Me	methyl
MeCN	acetonitrile
MeI	methyl iodide
MeOH	methanol
MeOH	methanol
MeOTf	methyltriflate
Mg	magnesium

min	minute(s)
mol	mole(s)
µM	micro molar
mol	mole
MOM	methoxymethyl
MS	molecular sieves
MsCl	methane sulfonyl chloride or mesyl chloride
NMR	nuclear magnetic resonance
OTf	triflate
Ph	phenyl
PhH	benzene
PhMe	toluene
ppm	part per million
q	quartet
RT	room temperature
s	singlet
t	triplet
td	triplet of dublets
TBAF	tetrabutylammonium fluoride
TBDPSCl	tert-butylchlorodiphenylsilane
TBSCl	tert-butyldimethylsilyl chloride
t-Bu	tert-butyl
TFA	trifluoroacetic acid
THF	tetrahydrofuran

CHAPTER 1

ANTIMALARIAL FLINDERSIAL ALKALOIDS

1.1 Background

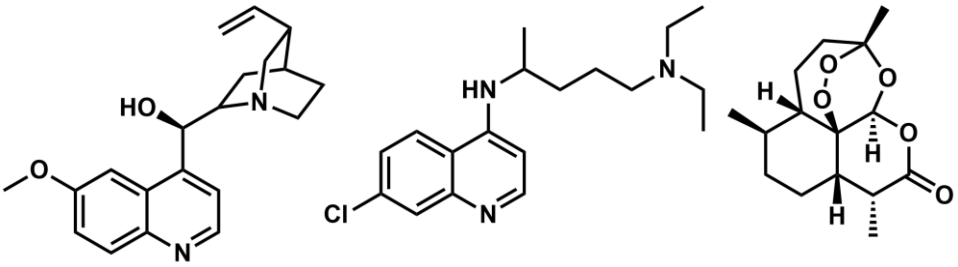
1.1.1 Malaria

Diseases caused by pathogenic microorganisms have been a persistent threat to human life since ancient days. Malaria is an infectious disease affecting humankind worldwide. Malaria is caused by five *Plasmodium* protozoal parasites: *P. falciparum, P. vivax, P. ovale, P. malariae and P. knowelski*. More than 95% of malarial infections are due to *P. falciparum* and *P. vivax*. Of these two parasites, *Plasmodium falciparum* has been responsible for the majority of malaria-related deaths. About half of the world's population lives in malaria-threatened areas.[1] Based on a statistical analysis, in 2010 malaria caused about 216 million clinical cases and approximately 665,000 deaths.[2] Presently, a number of effective antimalarial drugs and treatments are available, out of which artemisinin combination therapy is most effective. However, the malarial parasites are developing drug resistance. Moreover, current antimalarial drugs are still unaffordable for underdeveloped Asian and African countries where people are most susceptible to malaria. Hence, there is a tremendous demand for the discovery of novel and cost-economical antimalarial agents. Though the development of an antimalarial vaccine has been a goal for decades, the prevention and treatment of malaria has been immensely reliant on small-molecule-based treatments.[3] Historically, natural products and their derivatives have proven to be resource of potential candidates in developing successful malaria treatments.[4]

1.1.2 Natural products and potent small molecules against malaria

Quinine (**1,** Figure 1.1), an alkaloid natural product isolated from the bark of *Cinchona* trees in South America, has been an important antimalarial drug since its serendipitous discovery in the 1600s. In fact, quinine is considered to be the first successful chemical compound to treat an infectious disease. Parasite resistance to quinine was first observed in 1910, however. As the development of resistance to quinine was slow, it remained the main drug to treat malaria until the 1920's. Thereafter more effective synthetic antimalarials were available. Of these synthetic compounds, chloroquine (**2**) was the most important, as it had fewer side effects compared to quinine, and it has been widely used since the 1940s. Extensive use of chloroquine resulted in the slow development of parasite resistance, which appeared in parts of Southeast Asia and South America by the late 1950s. The resistance was more prominent by the 1980s.[5] Due to the development of parasite resistance to chloroquine, malaria was then treated using another natural product, artemisinin (**3**), isolated in 1972 from the Chinese herb *Artemisia annua*. Artemisinin and its derivatives have been proven to be the most effective drugs against malaria in modern days. However, development of multi-drug resistance by *Plasmodium falciparum* has been a constant impediment to eradicate malaria. Recently, researchers found that the malarial parasite is developing resistance to the monotherapy of artemisinin in South-East Asia, hence presently it is administered in combination with other antimalarial compounds to slow down the development of resistance to artemisinin-derived drugs.[6]

Figure 1.1 Antimalarials



1 Quinine ***2*** (±)Chloroquine ***3*** Artemisinine

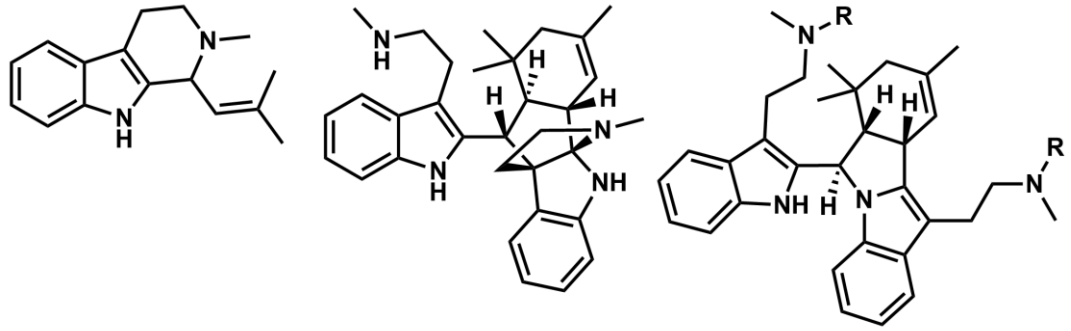
As the malarial parasite is constantly developing resistance to existing drugs, the search for new anti-malarial agents with novel modes of action has become a significant goal. In this scenario, the recently isolated anti-malarial flindersial alkaloids that may possess orthogonal biological action compared to chloroquine encourages the scientific community to foresee the development of new drugs to eradicate malaria in the near future.[7]

1.2 Introduction

1.2.1 Borreverines

Flindersia alkaloids have been known since the 1970's. Borreverine (**5**, Figure 1.2), a bis-indole alkaloid was the first flindersial alkaloid isolated from the aerial parts of *Borreria verticillata*[8] along with borrerine (**4**), a mono-indole alkaloid. Later, isoborreverine (**6**) and various analogues of borreverine were isolated from *Flindersia fournieri* along with borrerine and borreverine.[9] Of these compounds, isoborreverine exhibits prominent and diverse antimicrobial activity, whereas borreverine possesses moderate and narrow antimicrobial effect.[10]

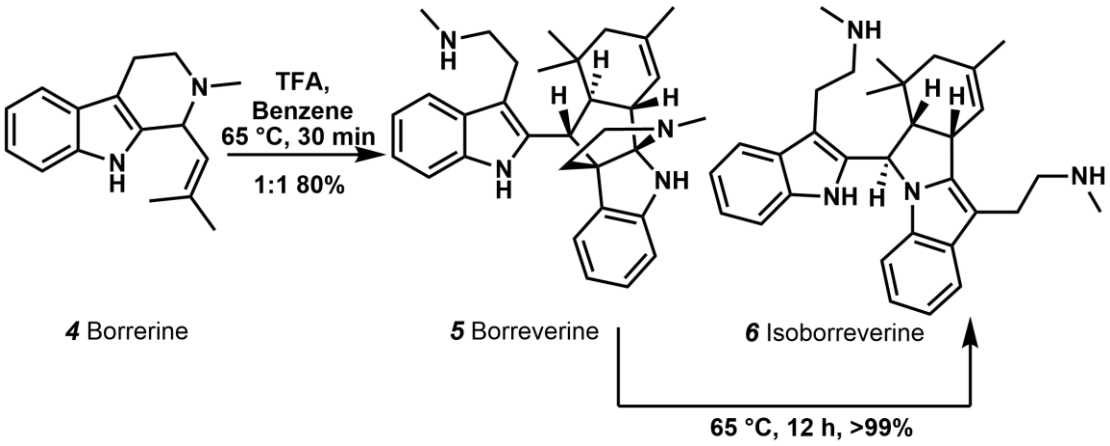
Figure 1.2: Borrerine and Borreverines



4 Borrerine ***5*** Borreverine ***6*** R=H Isoborreverine
7 R=CH_3 Dimethylisoborreverine

The co-occurrence of the indole alkaloid borrerine and its dimer borreverine in *Borreria verticillata* suggests borreverine might be produced by borrerine dimerization in the presence of acid. To confirm this hypothesis in 1979, Koch and Cavé reported a biomimetic synthesis of borreverine and isoborreverine in equal amounts with overall 80% yield when borrerine was exposed to trifluoroacetic acid (TFA) (Scheme 1.1).[11]

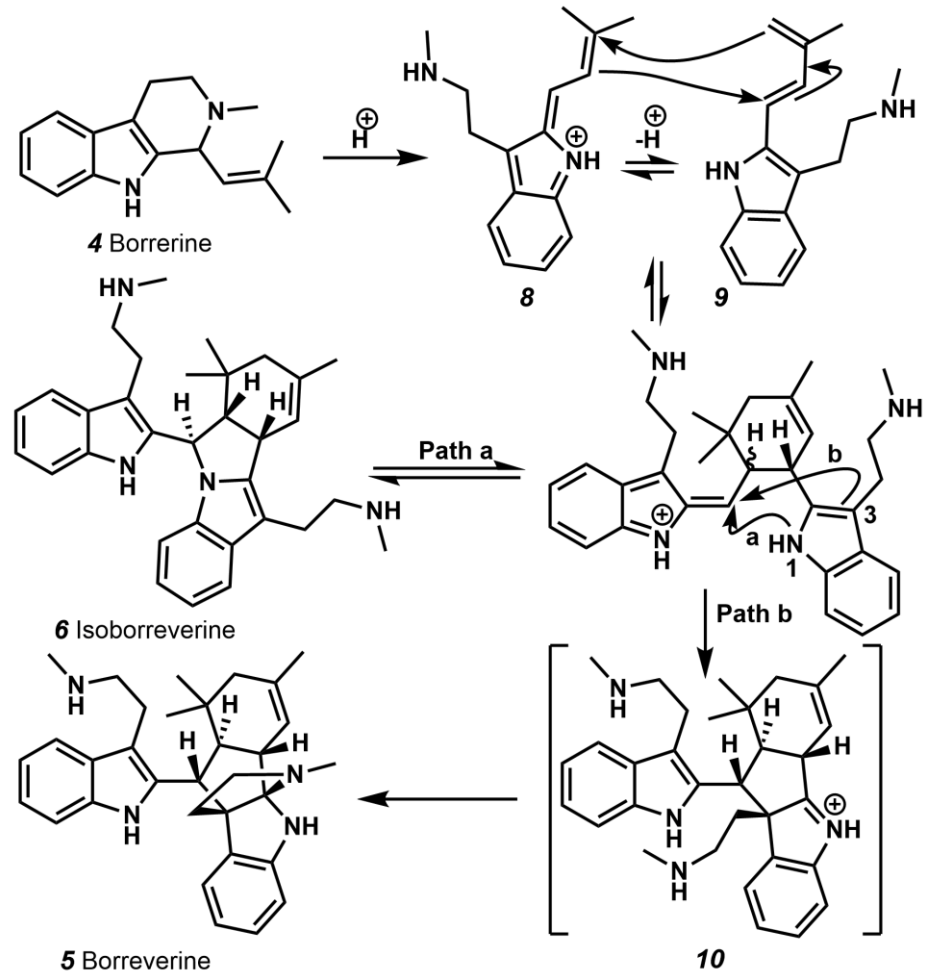
Scheme 1.1: Koch and Cave's biomimetic synthesis of borreverines



This process might proceed through initial protonation of the tertiary amine of borrerine. The lone pair of the indole nitrogen facilitates breaking the aromaticity to open

up the pyrrolidine ring to give the extended iminium **8** (Scheme 1.2). In the presence of acid, iminium **8** can be in equilibrium with diene **9**. These two intermediates can undergo a Diels-Alder cycloaddition followed by the nucleophilic attack of either N1 (path a) or C3 (path b) at the electrophilic olefin conjugated with the iminium of the pendant indole to produce isoborreverine or borreverine's precursor, respectively. Finally, the pendant secondary amine would attack the iminium to make the propellane core of borreverine. It is interesting to note that, if the reaction was run for 12 hours, only isoborreverine was formed. This result indicates that equilibrium exists between borreverine and isoborreverine in an acidic environment, favoring the latter product. The lack of aromaticity in one of the indole rings in the borreverine **5** might be a possible reason for this conversion.

Scheme 1.2 Koch and Cave's proposed mechanism for borreverines synthesis

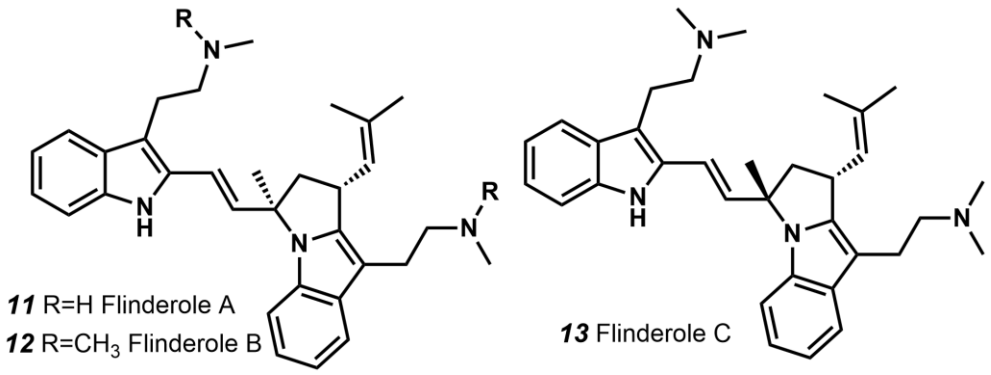


1.2.2 Flinderoles

In 2008 and 2009, Carroll, Quinn, Avery and coworkers isolated and elucidated the structures of novel bis-indole alkaloids with the aim of finding new antimalarial agents. Flinderole A (**11**, Figure 3), was extracted from the bark of *Flindersia acuminata* that was collected in Lake Bernie National Park, Queensland, Australia along with the previously known flindersial alkaloid, isoborreverine (**6**, Figure 1.2). Flinderoles B (**12**) and C (**13**), as well as dimethyl isoborreverine (**7**), were isolated from the bark of

Flindersia amboinensis that was collected in Papua New Guinea.[12] The structural elucidation revealed that the flinderoles are structural isomers to the borreverines. Two major components, tryptamine and pyrroloindole, were attached through a trans olefin. Two stereocenters are present on the pyrrolidine ring, one of which is a quaternary stereocenter. A pendant isobutenyl on the pyrrolidine ring is a characteristic feature in the flinderole structure.

Figure 1.3 Flinderoles



1.2.3 Antimalarial activity of flindersial alkaloids

Avery and coworkers also conducted a preliminary antimalarial assay using Dd2, a chloroquine-resistant *P. falciparum* strain, and the selectivity was assessed with the HEK-293 mammalian cell line (Table 1.1). The IC_{50} values of 0.15-1.42μM for the flindersia alkaloids are comparable with the most popular antimalarial small molecule chloroquine. In general, the flindersial alkaloids with dimethyl substituents on the amine groups are more potent than monomethyl amine containing alkaloids. On the other hand, the borreverines selectively inhibited the parasite cell lines when compared to mammalian cell lines. Based on the data, dimethylisoborreverine was the most active

alkaloid against the malarial cell line, whereas flinderole B is the most potent among flinderoles.

Table 1.1 Antimalarial Activity and Cytotoxicity of Flindersial Alkaloids[12b]

compound	IC_{50} (μM)		selectivity index $\left(\frac{IC_{50}\ HEK\text{-}293}{IC_{50}\ Dd2}\right)$
	Dd2	HEK-293	
Flinderole A	1.42 ± 0.07	19.97 ± 1.26	14
Flinderole B	0.15 ± 0.02	2.13 ± 0.08	14
Flinderole C	0.34 ± 0.03	9.75 ± 0.46	29
Isoborreverine	0.32 ± 0.02	8.99 ± 0.73	28
Dimethyliso borreverine	0.08 ± 0.01	4.09 ± 0.69	51
Chloroquine	0.22 ± 0.04	23.91 ± 2.21	108
Artemisinine	0.02 ± 0.01	>100	>6250

Dd2: Chloroquine resistant *Plasmodium falciparum* strain

HEK-293: mammalian cell line

Subsequently, more details of the biological activities of the flindersia alkaloids were described by Avery and coworkers.[7] Several *Plasmodium falciparum* strains were tested (Table 1.2). In addition, further biological experiments performed on the most active alkaloid, dimethylisoborreverine, gave some insight on the cellular pathway of the anti-malarial activity. Based on food vacuole morphology studies, the authors suggested that the reduction of hemozoin formation observed in the dimethylisoborreverine-treated parasites might be due to the hemoglobin degradation in the earlier stages of parasite growth rather than directly binding to the heme as in the case of chloroquine.

Table 1.2 Antimalarial Activity and Cytotoxicity of Flindersial Alkaloids II[7]

compound	*P. falsiparum* IC_{50} (μM)				Mammalian cells		SI
	3D7	FCR3[a]	HB3[b]	K1[a,b]	HeLa IC_{50} (μM)	HEK293 IC_{50} (μM)	
Flinderole A	0.75	0.92	1.16	1.61	19.93	19.97	12–26
Flinderole B	0.21	0.11	0.64	0.08	2.79	2.13	3–27
Flinderole C	1.10	0.36	1.17	0.33	19.21	9.75	8–29
Isoborreverine	0.24	0.16	0.47	0.33	10.68	8.99	19–56
Dimethyliso borreverine	0.22	0.02	0.81	0.06	5.37	4.09	5–205
Chloroquine	0.02	0.09	0.02	0.30	103.35	26.96	300–1348
Artemisinine	0.02	0.02	0.02	0.01	>100	>100	>5000

IC_{50} 50% Inhibitory concentration; [a] chloroquine-resistant strain;
[b] pyrimethamine-resistant strain; SI selective index (HEK 293/Pf)

1.3 Total syntheses of the flinderoles

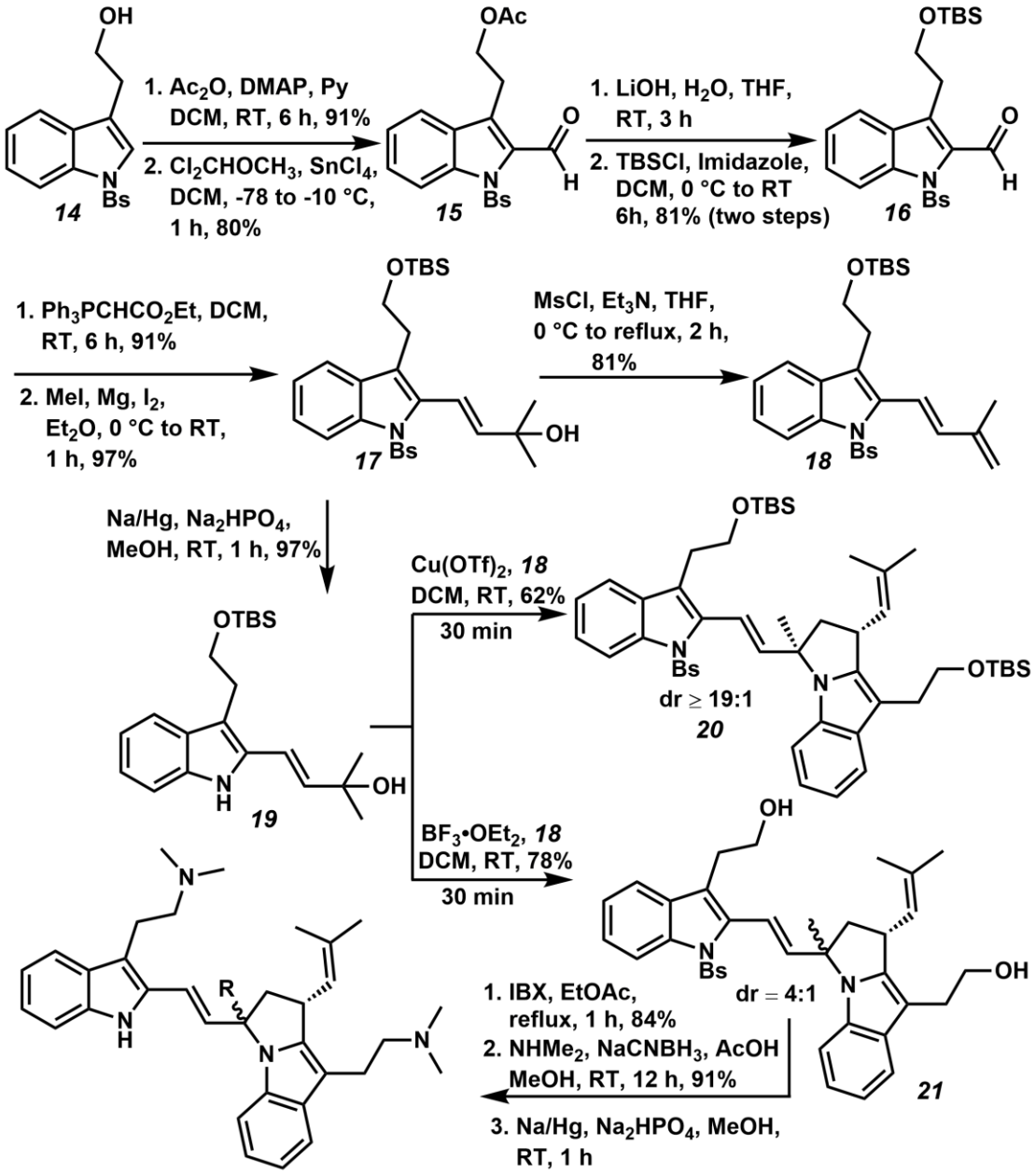
Soon after their isolation, two total syntheses of flinderoles B and C were reported. In 2011, Dethe reported a biosynthesis-inspired total synthesis of the flinderoles in 14 synthetic steps starting from tryptophol.[13] In the same year, Toste reported a convergent synthesis of the flinderoles using a versatile gold-catalyzed hydroarylation reaction to make the pyrroloindole framework.[14]

1.3.1 Dethe's total synthesis of flinderoles B and C

The first total synthesis of flinderoles B and C was achieved by the Dethe group. A regioselective formal [3+2] cycloaddition between strategically protected indole diene **18** and tertiary alcohol **19** (Scheme 1.3) constructed the flinderole framework. Sulfonated diene **18** was prepared from the tryptophol **17** by activating and eliminating the tertiary hydroxyl group. Alcohol **19** was prepared from the same tryptophol **17** by single electron

initiated desulfonylation. The substitution at the C-2 position on indole was successfully installed by formylation of **14** using dichloromethyl methyl ether and stannic chloride followed by Wittig olefination and Grignard addition. The formal [3+2] cycloaddition was highly diastereoselective (dr >19:1) in the presence of copper(II)triflate, whereas moderate (dr=4:1) selectivity was achieved with $BF_3{\cdot}OEt_2$. However the TBS group was removed in these acidic conditions. The resulting diastereomers were oxidized to their respective aldehydes, and the total synthesis of flinderoles B and C was completed by reductive amination in a total of 14 steps.

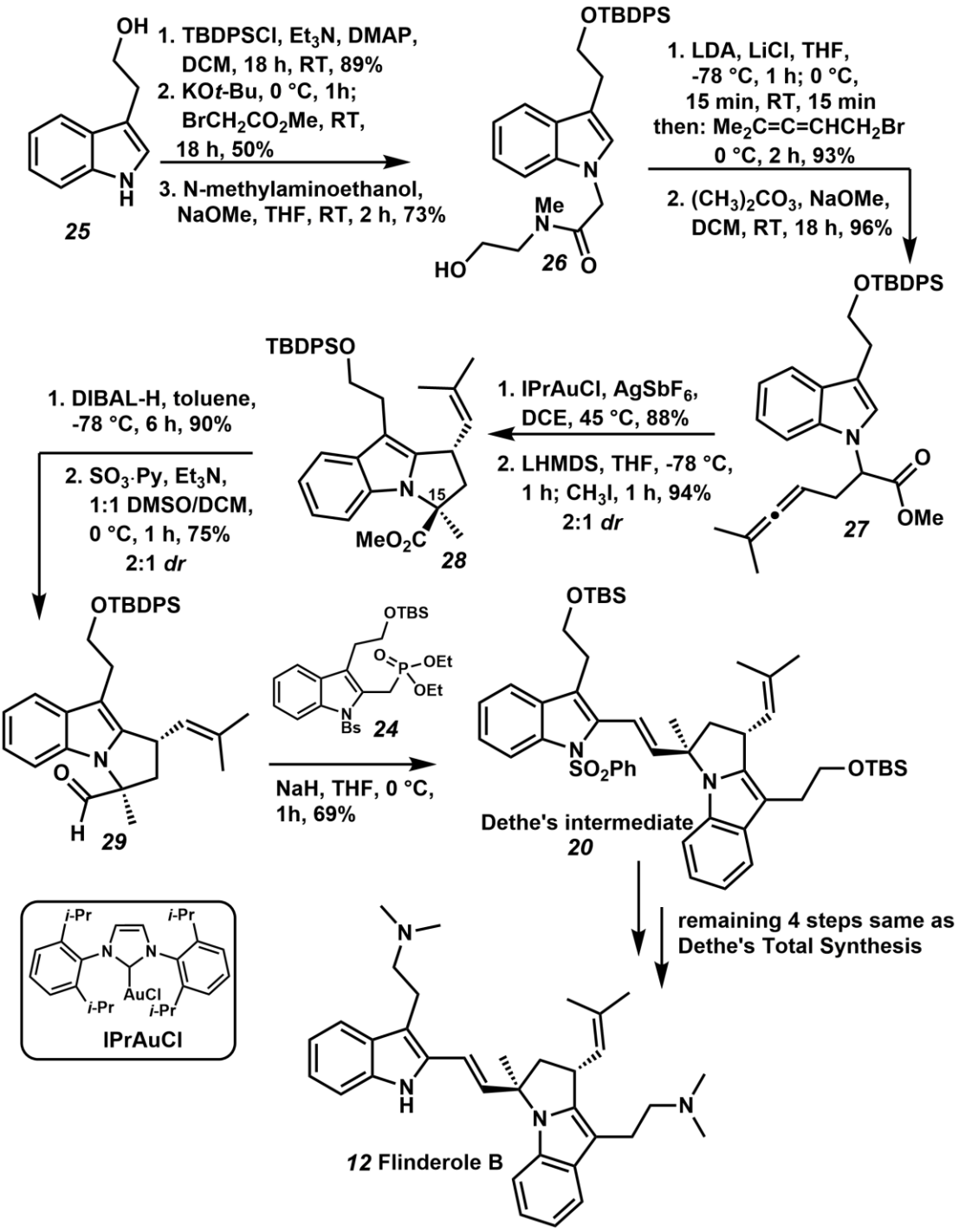
Scheme 1.3 Bioinspired synthesis of flinderoles by Dethe group[13]



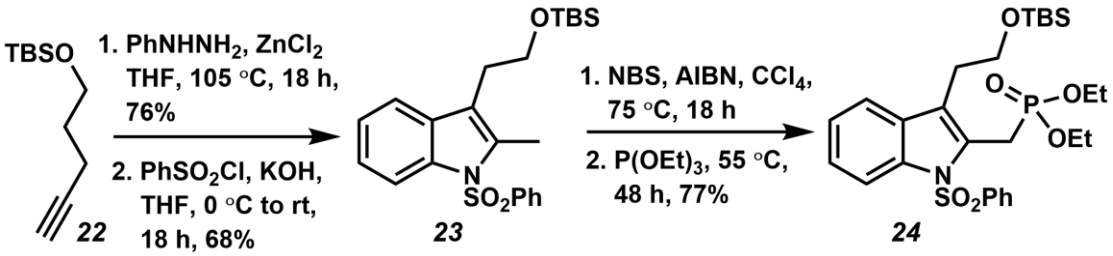
1.3.2 Toste's total synthesis of flinderoles B and C

Toste's group used a novel synthetic methodology to synthesize the flinderoles. They have shown that the versatile gold catalyzed intramolecular hydroarylation of a pendant allene can install the pyrrolidine ring and isobutenyl side chain—the unique structural features of the flinderole molecular framework–in one reaction. This convergent synthesis involves the Horner-Emmons-Wadsworth olefination reaction to combine phosphonate **24** and pyrroloindole **29** (Scheme 1.4). The pyrroloindole **29** was prepared from indole-allene **27** through a gold(I)-catalyzed cyclization. The quaternary center at C-15 was installed by enolate alkylation. Indole-allene **27** was prepared by enolate alkylation of the amide **26** and the amide in turn prepared from commercially available typtophol **25**. Synthesis of phosphonate **24** began with zinc promoted Fisher indole synthesis using TBS protected pentyn-1-ol **22** and phenyl hydrazine followed by indole sulfonylation to yield the tryptophol **23** (Scheme 1.5). Finally, radical bromination of methyl group and an Arbuzov reaction sequence afforded phosphonate **24**. Pyrroloindole **29** and phosphonate **24** were combined through an olefination reaction to yield Dethe's intermediate **20,** and the total synthesis flinderoles B and C was completed thereafter using Dethe's approach.

Scheme 1.4 Total synthesis of flinderoles B and C by Toste's group[14]



Scheme 1.5 Synthesis of phosphonate 24[14]



1.3 Conclusions

In conclusion, the emergence of multi-drug resistance in parasites is posing a tremendous challenge to find new anti-malarial agents. In the search of new anti-malarials, the flindersial alkaloids seem to have a novel mechanism of action compared to chloroquine. Structural novelty also makes *Flindersia* alkaloids as interesting targets to synthesize. Dethe[14] and Toste[15] synthesized flinderoles B and C in 14 and 19 total steps respectively.

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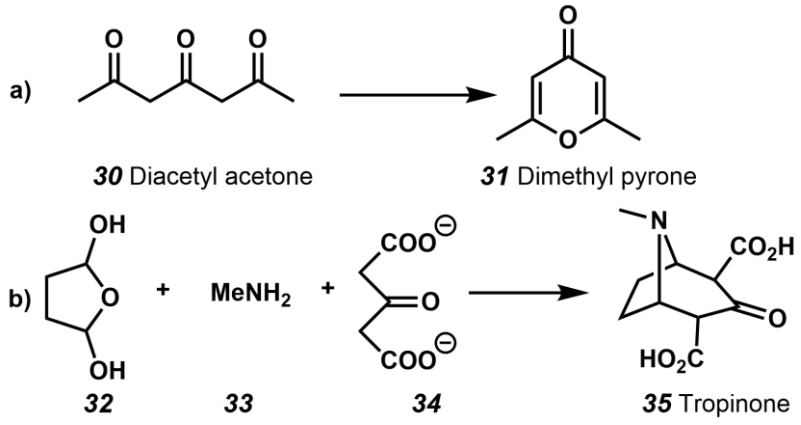
CHAPTER 2

BIOMIMETIC SYNTHESIS OF THE FLINDEROLES[14,16,17]

2.1 Introduction

"For all natural products, there exists a synthesis from ubiquitous biomolecules. The inherent interconnectivity of natural products implies that a truly biomimetic total synthesis represents a general solution not to the preparation of a compound, but to the preparation of all similarly derived natural products (discovered and undiscovered)".[1] These words by Skyler and Heathcock enunciate the remarkable impact of biomimetic synthesis of natural products. Development of one pot procedures mimicking nature to prepare complex molecules from small building blocks has been well recognized. In the 1890s Collie and coworkers synthesized dimethyl pyrone from diacetyl acetone, which later led them to develop a biosynthetic hypothesis for polyketides. In the same manner, alkaloids were also prepared from simple starting materials in one pot, such as the preparation of tropinone (**35**, Figure 2.1).[2]

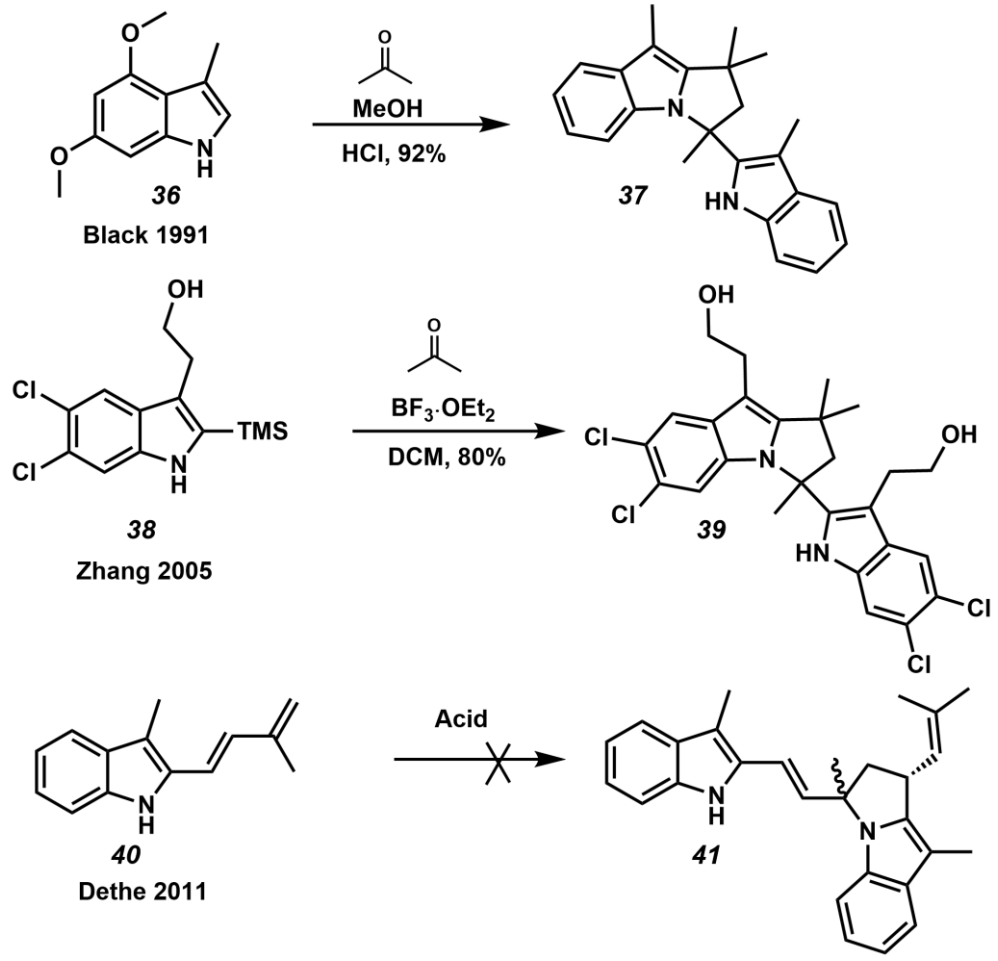
Figure 2.1 Seminal biomimetic syntheses[2]



2.2 Synthetic methods for pyrroloindole framework

The synthesis of pyrroloindole derivatives using unprotected indoles and methyl ketones in the presence of Lewis acid or Brønsted acid conditions has been known for decades.[3] More relevant work to synthesize the pyrroloindole framework present in the flinderoles was done by Black in the early 1990's using 3-methyl indole **37** and acetone (Scheme 2.1).[4] Acetone is the electrophile and serves as intermolecular partner for the whole cyclization. Recently, Xuqing Zhang reported an unusual dimerization of unprotected 2-silyl tryptophols **38** and acetone in the presence of a Lewis acid to give pyrroloindole **39**.[5] However, all these examples are intermolecular using an external dimerization partner like acetone. To our knowledge, no examples of intramolecular dimerizations have been reported. Interestingly, Dethe reported that intramolecular dimerization on unprotected indoles had not been successful. In his attempt to synthesize the flinderole framework, it was found that in the presence of acid in various conditions 2-diene-appended 3-methyl indole **40** gave only an intractable polymeric mixture instead of intramolecular dimerization products.[6]

Scheme 2.1 Reports to prepare pyrroloindole derivatives using unprotected indoles

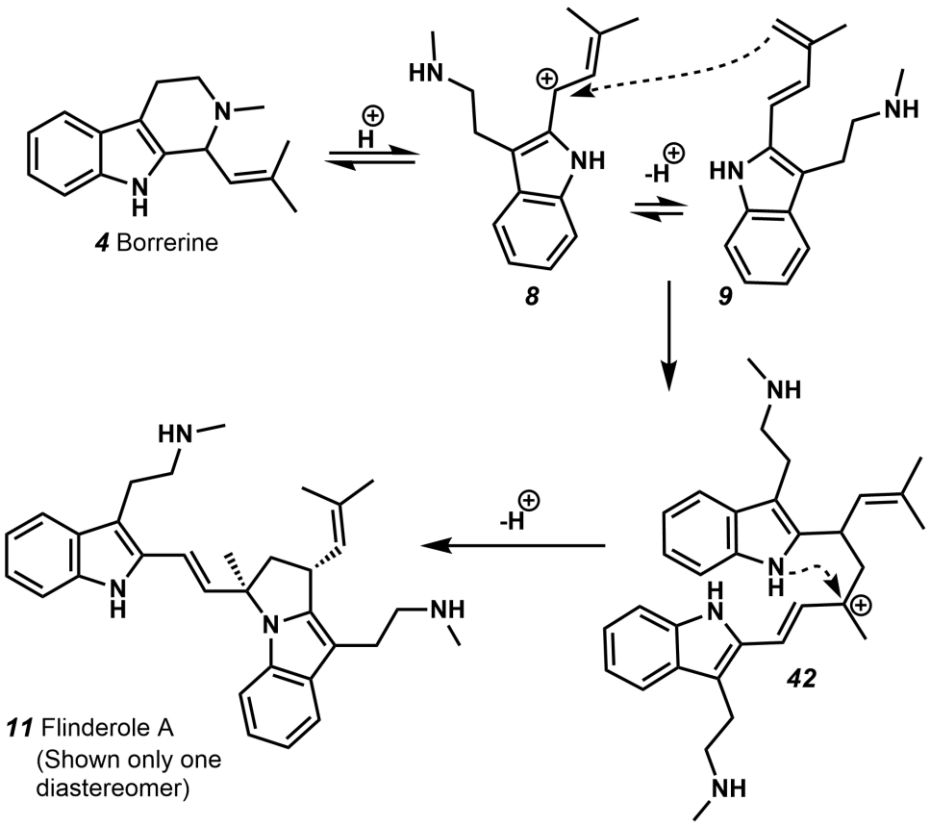


2.3 Biomimetic synthetic plan for the flinderoles

Intrigued by the biomimetic synthesis of the borreverines from borrerine (**4**) by Koch and Cavé in the 1970's, we hypothesized that the flinderoles could also be prepared similarly from borrerine through an acid dimerization.[7] It is noteworthy to mention that borrerine, a *borreria* alkaloid, was also isolated along with a few other flindersial alkaloids from plants like *Flindersia fourinieri*,[8] suggesting a possible biosynthetic relation between borrerine and the flindersial alaklods. In the presence of acid, the

tertiary amine of borrerine gets protonated to open the carboline ring to a stable benzylic cation **8**. Deprotonation of the isobutenyl methyl can then lead to indole diene **9**. Borreverine's formation was postulated to occur through [4+2] cycloaddition of the intermediates **8** and **9** (Scheme 2.2). However, if the dimerization can occur though a formal [3+2] cycloaddition, then flinderoles could be prepared in one pot (Scheme 2.2). Further support for this possibility was shown by Dethe and coworkers in their successful total synthesis of flinderoles B **12** and C **13**.[6] They developed and utilized a formal [3+2] cycloaddition between two strategically protected alkene-appended indoles in the presence of a Lewis acid (Scheme 1.3).

Scheme 2.2 Hypothesis for flinderole synthesis



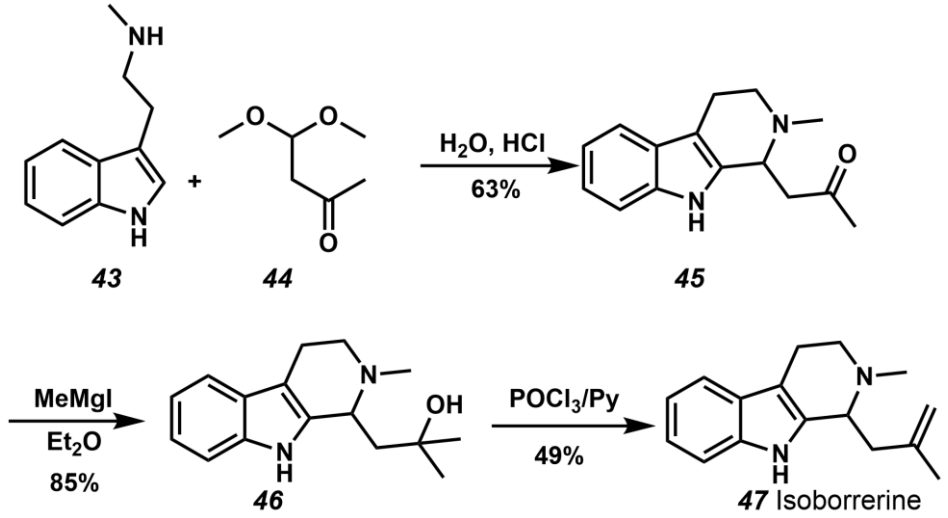
2.4 Synthesis of borrerine (4)

As malaria is more wide spread in underdeveloped countries, present medicines for treating malaria are still unaffordable for most of the malarial patients. Concise synthetic routes using readily available chemicals and few or no column chromatographic purifications in the synthesis of antimalarial intermediates may reduce the price of antimalarial medicine in the future. To meet this goal, we wanted to synthesize the potent antimalarial flindersial alkaloids in as few synthetic steps as possible. As borrerine is not commercially available and isolation is not practical, we decided to synthesize borrerine from readily available tryptamine.

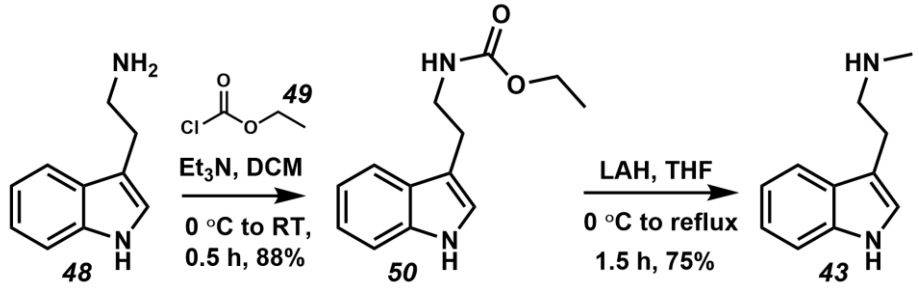
2.4.1 Initial attempt to synthesize borrerine (4)

Soon after borrerine (**4**) was isolated from *Borreria verticellata* by Cavé and coworkers in 1973,[9] several syntheses were reported.[10] But most of them suffered from a long synthetic sequence. Initially, we were interested in the Koch and Tillequin synthesis of isoborrerine (**47,** Figure 2.2),[10a] another mono indole natural product and structural isomer of borrerine which was isolated from *Flindersia fourinieri*. With a few changes in reagent selection and using fewer steps, borrerine could be synthesized in a similar way as isoborrerine **47**. We started our synthesis of borrerine from *N*-methyl tryptamine (**43)**, which is commericially available or readily synthesized from tryptamine **48** in two steps (Scheme 2.3).[11]

Figure 2.2 Synthesis of isoborrerine (**47**) by Koch[10a]



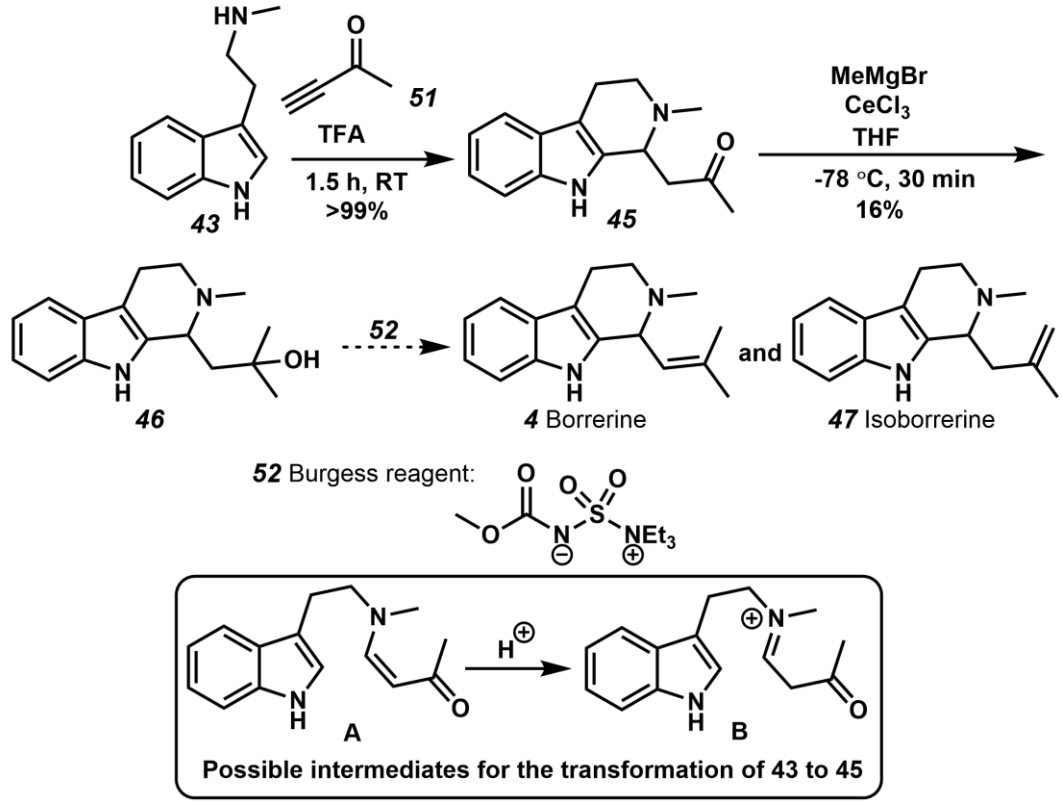
Scheme 2.3 Synthesis of *N*-methyl tryptamine (**43**)



N-methyl tryptamine **43** was then subjected to a modified Pictet-Spengler reaction using 3-butyn-2-one (**51**) instead of acetylacetaldehyde dimethylacetal (**44**) to yield methyl carbolineindole ketone **45** in quantitative yield with a pendant acetylmethylene group (Scheme 2.4).[10a] Presumably, the reaction could proceed through the enamine intermediate **A** and iminium intermediate **B**. In our hands, the nucleophilic 1,2-addition of methyl magnesium bromide to ketone **45** was an incomplete reaction and the chromatography purification of the tertiary alcohol **46** from the remaining starting material was difficult. Methyl cerate addition consumed all the starting material;

however, the isolated yield of the product **46** was only 16%. We envisioned that Burgess reagent via dehydration would produce selectively the internal olefin as in borrerine rather than the terminal olefin as in isoborrerine (**47**).

Scheme 2.4 Initial attempt to synthesize borrerine (**4**)

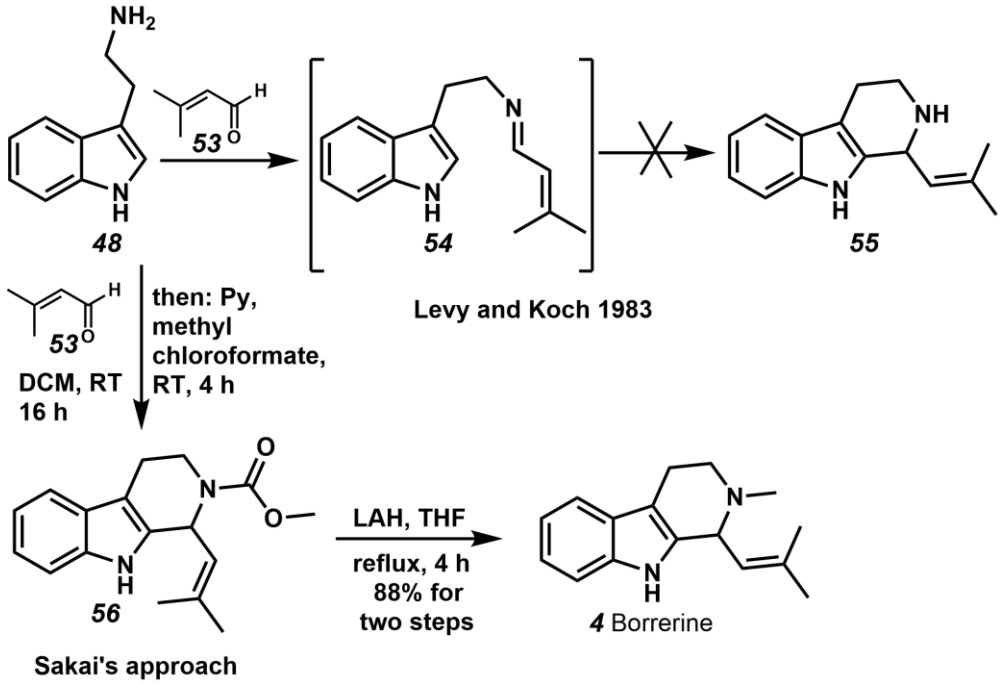


2.4.2 Sakai's approach to synthesize borrerine (4)

Levy and Koch reported that the reaction between tryptamine (**48**) with butenal **53** gave no desired carboline **55**.[10b] Presumably the reaction might have stopped at the imine intermediate **54**. Sakai elegantly solved this problem by trapping the imine with chloroformate in the presence of pyridine, which can act as the acid scavenger.[12] We followed the exact procedure developed by Sakai. However, we improved the yield of

carbamate **56** by avoiding column chromatography. Instead, trituration of the crude material using 10% ether in hexane gave the pure carbamate **56** in excellent yield. Reduction of the carbamate **56** with LAH proceeded smoothly to yield borrerine **4** (Scheme 2.5).

Scheme 2.5 Synthesis of borrerine (**4**)



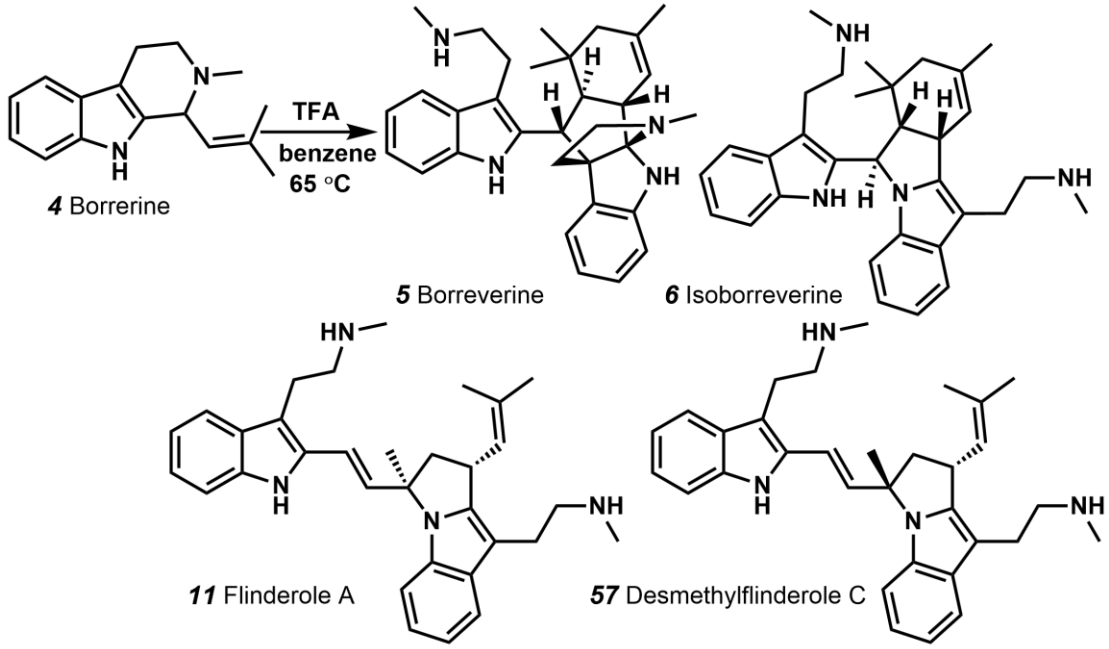
2.5 Dimerization of borrerine in the presence of acid

With borrerine (**4**) in hand, our next goal was to reproduce the biomimetic synthesis of borreverine (**5**) and isoborreverine (**6**) from borrerine in 80% yield as reported by Koch and Cavé.[13] We were interested to see whether the remaining 20% of the mass balance might be flinderoles. As the flinderole structures were not known in the 1970's, the authors might not have noticed the 20% yield of flinderole formation. To our

disappointment, the reaction conditions reported were not adequate, as the number of equivalents of TFA used and concentration were not mentioned.

To find the exact number of equivalents of TFA necessary to reproduce the 80% yield of the borreverines, we started testing the dimerization reaction from one equivalent to a maximum of six equivalents in different reaction attempts. There was no reaction with 1 equiv of TFA at 65 °C in benzene for 2 h. The starting material was totally consumed with 2 equiv of TFA at 65 °C for 40 min as observed by TLC. ^{1}H NMR showed the presence of isoborreverine as the major product. However, we were surprised to see only trace peaks of borreverine. With careful analysis of the ^{1}H NMR we discovered that there were two more compounds present in the crude material. We were ecstatic to realize that we had synthesized flinderole A (**11**) and its unprecedented diastereomer desmethylflinderole C (**57**), presumably through [3+2] cycloaddition.[14] As we envisioned, the possibility of a [3+2] along with the [4+2] cycloaddition was confirmed by the formation of the flinderoles and borreverines in the same reaction. In fact, the NMR integration shows the formation of isoborreverine (**6**) and flinderole diastereomers in almost 30% and 70% respectively, favoring [3+2] cycloaddition over [4+2]. This result is contrary to the findings of Koch and Cavé under TFA conditions.

Scheme 2.6 Dimerization of borrerine (**4**) with TFA



entry	TFA (equiv)	time	Ratio of Products[a] *5*: *6*: *11*:*57*
1[b]	unknown	30 min	50: 50: 0: 0
2[b]	unknown	12 h	0: 100: 0: 0
3	1.0	2 h	0: 0: 0: 0
4	2.0	40 min	1: 28: 41: 30

[a] Determined by the integration of ^{1}H NMR peaks.
[b] reported result[13]

2.5.1 Extraction and purification challenges

Initially, extraction as well as separation of all these structural isomers was a conundrum as all the alkaloids were produced presumably in salt form, each with 4 basic nitrogens. Quenching the TFA salts and excess TFA in the solution to free the alkaloids using traditional methods like aq $NaHCO_3$ or aq NaOH resulted in inseparable emulsions. Only a small portion of the products were recovered in polar organic solvents like ethyl

acetate. In search of efficient non-aqueous quenching methods for TFA, we found a protocol developed by Ganesan and coworkers to quench the TFA in Boc protecting group removal using basic amberlyst 21 A.[15] Applying this method let us recover greater than 95% of the crude material. With the crude material in hand, our next challenge was to separate the alkaloids from each other. Conventional methods of purification like column chromatography using silica gel or alumina were not successful. However, the TFA salts of each alkaloid were separable by semi-preparative HPLC to yield enough compound for spectroscopic characterization. Loading small portions of crude material on neutral alumina TLC plates eluting with 2-3% of methanol in DCM also gave reasonably good separation. Repeated alumina TLC purification successfully separated all the alkaloids produced in the reaction.

Motivated by the formation of the flinderole diastereomers from borrerine, we decided to screen a range of TFA equivalents, different acids, solvents, and temperature to find the reaction conditions that favored flinderole formation over the borreverines.

2.5.2 TFA

As the number of equivalents of TFA was increased, the formation of isoborreverine was seen more than the flinderoles. At lower temperatures, even with a higher concentration of TFA, the reaction was slow and incomplete. Interestingly, neat TFA gave only isoborreverine. This indicates that at a higher concentration of acid, the kinetically favored flinderoles presumably, transformed to the more thermodynamically stable isoborreverine (**6**). In fact, a control experiment of treating purified flinderoles with

TFA gave exclusively isoborreverine, confirming that isoborreverine can be formed from the flinderoles and that it is the most thermodynamically stable of the *flindersia* alkaloids. This is also evident from the findings of Koch and Cavé, wherein borreverine (**5**) and isoborreverine (**6**) were observed in equal ratio after 30 minutes of reaction time, but only isoborreverine was obtained after 12 hours. This suggests that the kinetically favored borreverine (**5**) is completely transformed to the thermodynamically stable isoborreverine (**6**) after extended reaction time.[13]

Table 2.1 TFA screening

4 Borrerine

entry	acid (equiv)	temp	time	product ratio[a] *5* : *6* : *11* : *57*
1	TFA (2.0)	55 °C	80 min	1 : 29 : 38 : 32
2	TFA (2.0)	40 °C	2.5 h	1 : 30 : 30 : 23[b]
3	TFA (3.0)	40 °C	2 h	0: 34 : 34 :32
4	TFA (3.0)	65 °C	0.5 h	1 : 43 : 28 : 28
5	TFA (6.0)	0–22 °C	24 h	0 : 34 : 16: 13[b]
6	TFA (6.0)	40 °C	0.5 h	0 : 21 : 17 : 17[b]

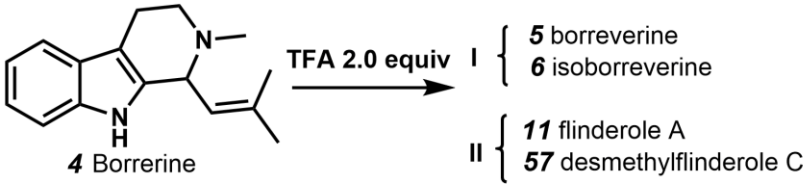
[a] Determined by integration of ^{1}H NMR peaks.
[b] The balance of the material was 4.

2.5.3 Solvents

Other solvents like tetrahydrofuran (THF), dioxane, or *N*, *N*-dimethylformamide (DMF) resulted in no reaction (Table 2.2). Protic polar solvent like methanol gave a significant ratio of flinderoles; however, the reaction was incomplete. Changing the

solvent from benzene to chloroform gave flinderoles as the majority of the crude mixture; however, borrerine was less reactive in these conditions as evident by the isolation of unreacted starting material. Presumably, the halogenated solvent may dilute the TFA to a certain extent to suppress any further interconversion of flinderoles to isoborreverine. However, reaction with TFA in dichloromethane (DCM) was very slow. Based on these results, we can predict that acid strength is crucial for the dimerization reaction to get selectivity either for the [3+2] or for the [4+2] cycloadditions.

Table 2.2 Sovents



entry	solvent	temp	time	product ratio[a] *I* : *II*
1	benzene	65°C	40 min	40: 60
2	chloroform	45 °C	1.2 h	10: 40[b]
3	DCM	22 °C	12 h	5: 5[b]
4	MeOH	60 °C	2 h	10: 40[b]
5	DMF, THF, Dioxane	60 °C	1 h	0: 0[b]

[a]Determined by integration of ^{1}H NMR peaks. The combined yields were all above 95%. [b]The balance of the material was 4.

2.6 Acids

Several Brønsted and Lewis acids were examined in the dimerization of borrerine (**4**). Many of these acids either failed to promote the reaction or resulted in decomposition to unidentified intractable mixtures.

2.6.1 Lewis acids

When dimerization occurred with Lewis acids, isoborreverine was almost exclusively produced. Reaction with aluminum chloride at room temperature failed to give any product. There was no reaction even at 60℃ with TMSOTf. Reaction was incomplete even at elevated temperatures with scandium triflate or aluminum chloride, however isoborreverine **6** was the only dimerization product observed in those reactions. When $BF_3{\cdot}OEt_2$ in DCM was used at room temperature, complete starting material consumption was observed only after 3 days. This condition led to the formation of exclusively isoborreverine (**6**) with full conversion (Table 2.3).

Table 2.3 Lewis acids

4 Borrerine

entry	acid[a]	temp	time	product ratio[b] *5* : *6* : *11* : *57*
1	TMSOTf	60 °C	40 min	0 : 0 : 0 : 0
2	$AlCl_3$	0–22 °C	120 min	0 : 0 : 0 : 0
3	$AlCl_3$	60 °C	40 min	0 : 40 : 0: 0[c]
4	$Sc(OTf)_3$	60 °C	40 min	0 : 38 : 0 : 0[c]
5	Tf_2O	0 °C	60 min	0 : 50 : 0 : 0[c]
6	$BF_3{\bullet}OEt_2$	22 °C	3 days	0 : 100 : 0 : 0

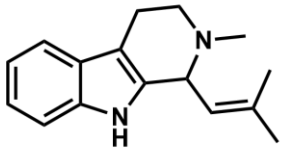
[a]Unless otherwise noted, 2 equiv of acid was used.
[b]Determined by integration of ^1H NMR peaks.
The combined yields were all above 95%.
[c]The balance of the material was 4.

2.6.2 Brønsted acids

Borrerine (**4**) was unreactive at 0 °C or at room temperature with Brønsted acids. Triflic acid turned out to be too strong at 60 °C for borrerine dimerization. Interestingly, 1N HCl in methanol produced a majority of the flinderoles compared to isoborreverine (**6**). Borreverine (**5**) was observed in significant amounts only with acetic acid, which is a very weak acid compared to trifluoroacetic acid (Table 2.4).

The formation of different flindersial alkaloids in the dimerization of borrerine (**4**) was determined by the crude ^{1}H NMR integration of significant peaks. Initially, in the crude ^{1}H NMR of the acetic acid reaction, the very close significant peaks at 5.40–5.45 in DMSO-d^6 of both isoborreverine (**6**) and borreverine (**5**) mislead us to overlook the formation of borreverine in greater amount than isoborreverine. However, after purifying the crude material, with the full spectrum in hand, we found that for the first time we produced borreverine in a significant amount. In the literature, the ^{1}H NMR of borreverine was reported only in $CDCl_3$. We confirmed the identity of borreverine by comparing the synthetic borreverine ^{1}H NMR in $CDCl_3$ with the reported borreverine ^{1}H NMR. Once we had the ^{1}H NMR of pure borreverine in DMSO-d^6, we again revisited all the crude ^{1}H NMRs and find out that no other acid produced borreverine more than a trace amount.

Table 2.4 Brønsted acids



4 Borrerine

conditions

5 borreverine
6 isoborreverine
11 flinderole A
57 desmethylflinderole C

entry	acid[a]	solvent	temp	time	product ratio[b] ***5*** : ***6*** : ***11*** : ***57***
1	TfOH	benzene	0–22 °C	40 min	0 : 0 : 0 : 0[c]
2	TfOH	benzene	60 °C	40 min	decomp.
3	*p*TsOH	benzene	0–22 °C	60 min	0 : 0 : 0 : 0[c]
4	1N HCl[d]	methanol	55 °C	2 hr	1 : 30 : 43 : 26
5	CH_3COOH[e]	None	55 °C	18 hr	26 :12 : 30 : 32
6	CH_3COOH[f]	None	55 °C	18 hr	31 :21 : 24 : 24
7	TFA	None	65 °C	48 h	0 : 0 : 100 : 0

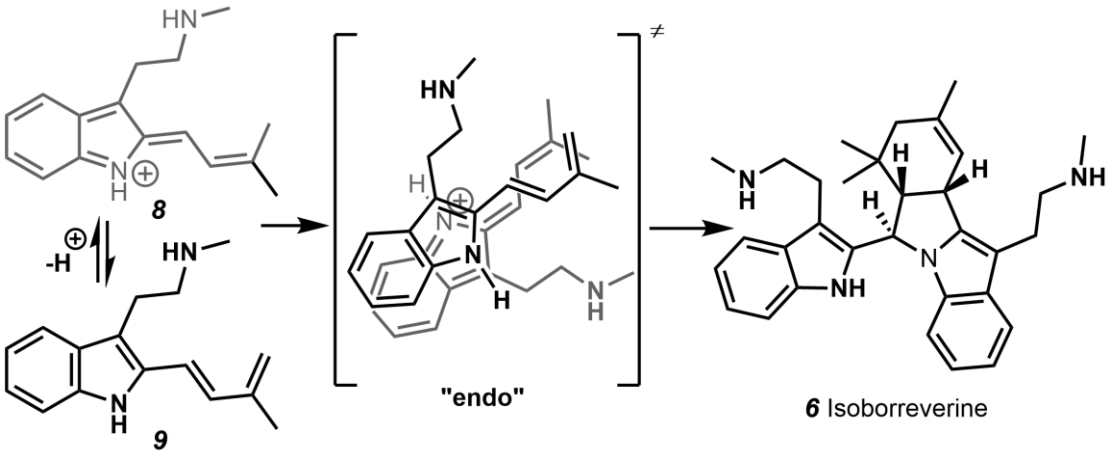
[a]Unless otherwise noted, 2 equiv of acid was used. [b]Determined by integration of ^{1}H NMR peaks. The combined yields were all above 95%. [c]The balance of the material was 4. [d]6 equiv of acid. [e]147 equiv of acid. [f]210 equiv of acid.

2.7 Borreverine formation

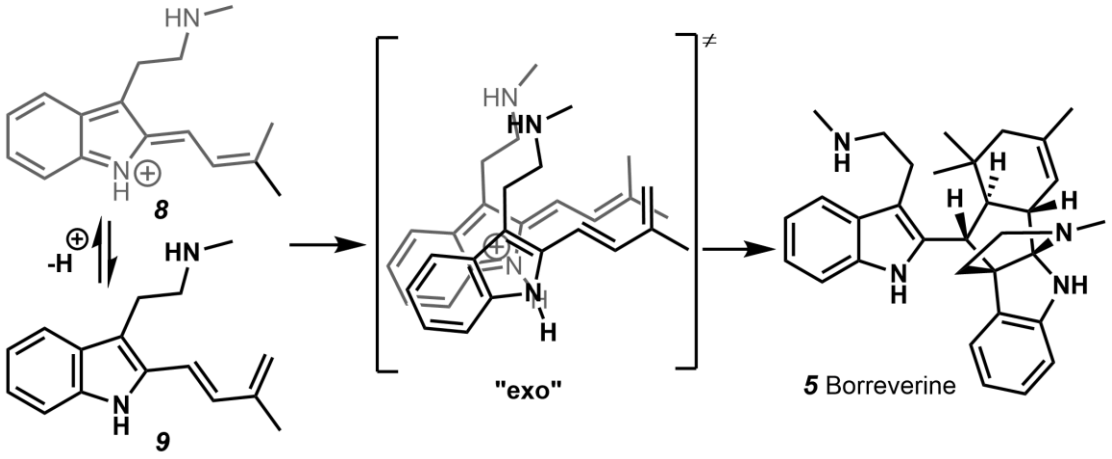
In our attempt to reproduce Koch and Cavé's result of a 1:1 ratio of borreverine and isoborreverine from borrerine with TFA, we always saw only a trace amount of borreverine formation.[14,16] This can be explained in terms of kinetic and thermodynamic arguments. For kinetic reasons, if we look at the transition state arrangement, it is very clear that borreverine formation would have to arise from a less favorable exo-Diels–Alder reaction in the initial step (Scheme 2.7). However, isoborreverine has to arise from a more favorable endo-Diels–Alder transition state (Scheme 2.8).[17] For thermodynamic reasons, the lack of aromaticity in one of the indoles makes it less stable than isoborreverine. However, in the case of a weak acid like acetic acid, borreverine is

produced in a significant ratio (Table 2.4). Further study is underway to demonstrate the effects of pH on product formation in the borrerine dimerization reaction.

Scheme 2.8 Formation of isoborreverine ***6***



Scheme 2.7 Formation of borreverine ***5***



2.8 Synthesis of flinderole B and C and dimethylisoborreverine

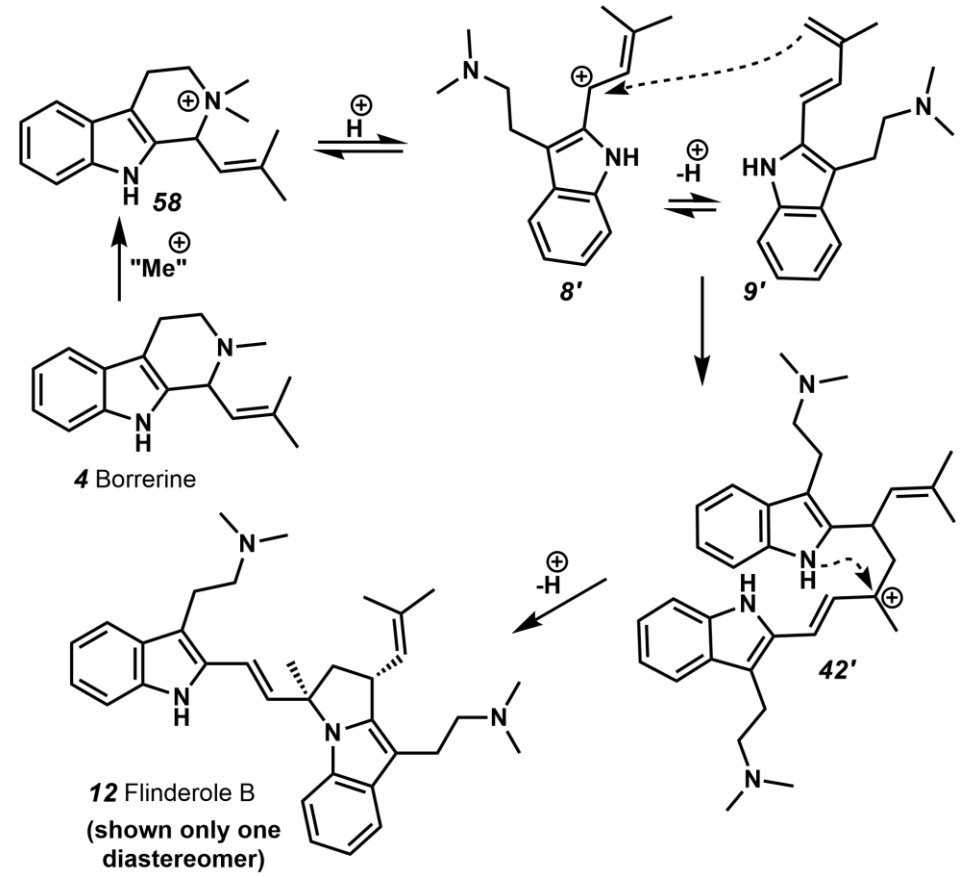
After the successful execution of a biomimetic synthesis of the secondary amine flindersial alkaloids through borrerine dimerization, our focus then shifted to synthesize the tertiary amine congeners flinderole B (**12**), C (**13**) and dimethylisoborreverine (**7**).

These tertiary amine alkaloids were more desired because of greater antimalarial activity. Two different synthetic routes were considered. Initially, we thought about a straightforward base-promoted methylation of synthetically prepared flinderole A (**11**), its diastereomer desmethylflinderole C (**57**), and isoborreverine (**6**). However, we were intrigued by the presence of tertiary amine alkaloids only in *F. amboinensis*, whereas the secondary amine alkaloids occurred exclusively in *F. acuminata*. This observation prompted us to propose a possible biosynthesis of the tertiary amine flindersial alkaloids. We envisioned that in *F. amboinensis* a direct methylation of borrerine could make a quaternary ammonium **58** (Scheme 2.9), which initiates the dimerization reaction in the presence of acid. However, borrerine was not isolated from *F. amboinensis* to support this hypothesis. Synthetically, this hypothesis would be very efficient, as all the tertiary amine congeners could be prepared in one-pot from borrerine. With readily available strong electrophilic methylating reagents like Meerwein's salt or methyl triflate, we decided to test the borrerine methylation and acid dimerization cascade.

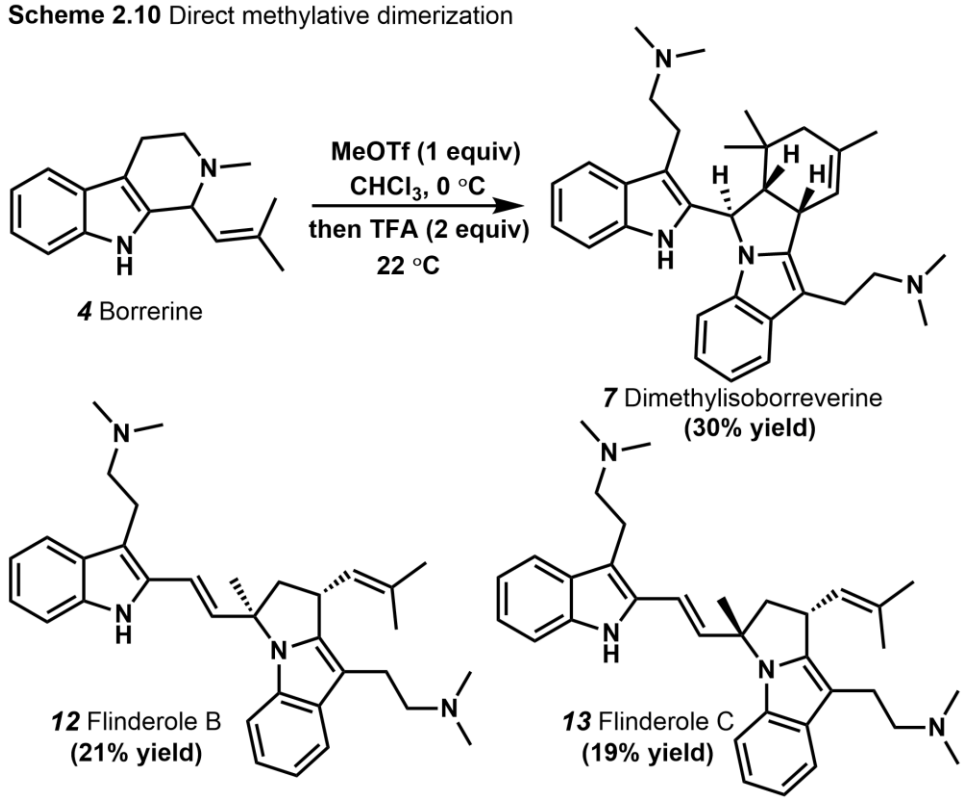
Treatment of borrerine with 1 equivalent of methyl triflate at 0 °C produced the ammonium **58**, which could be opened into **8'** and **9'** in the presence of 2 equivalents of TFA at room temperature. The ammonium **58** was not reactive enough to initiate the dimerization cascade at 0 °C with TFA. There was no reaction with 1 equivalent of TFA. At ambient temperature, the formation of the tertiary amine flindersial alkaloids **12** and **13** was very fast (Scheme 2.10). An extended reaction time produced dimethylisoborreverine exclusively, suggesting its structural stability. Work-up and

purification of these less polar alkaloids was less cumbersome than the more polar secondary amine congeners.

Scheme 2.9 Hypothesis for flinderole B and C synthesis



Scheme 2.10 Direct methylative dimerization



2.9 Conclusion

Our hypothesis of flinderole biosynthesis, which mirrored the borreverines' biosynthesis, proved to be successful in vitro as we were able to synthesize all the antimalarial flindersial alkaloids from borrerine (**4**). The formation of borreverine (**5**) was observed only with weak acetic acid. This 3-step synthesis of racemic flinderoles gives an excellent opportunity to synthesize their analogues. Their biological screening would give more insight into their antimalarial potency and allow the development of a potential new antimalarial in the future.

2.10 Experimental section

2.10.1 General considerations

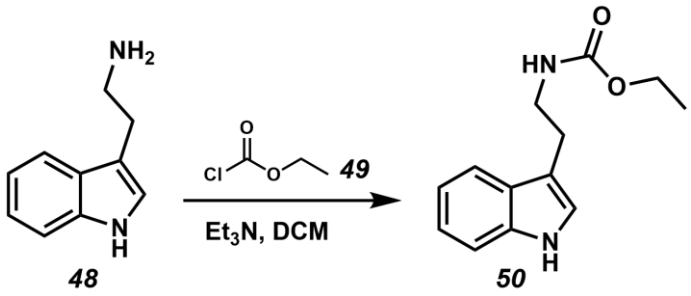
All the reactions were performed in flame- or oven-dried glassware. Benzene, THF, and DCM were purged with argon and dried over activated alumina columns. Column chromatography was performed on 60 Å silica gel (Sorbent Technologies) and commercially available neutral alumina. The ^{1}H and ^{13}C NMR spectra were recorded on a JEOL ECA-500 or ECX-400 spectrometer. Analytical thin layer chromatography was performed on EMD silica gel/TLC plates with fluorescent indicator 254 nm. HPLC was performed using a Gilson 321 pump with UV/VIS-155 detector and FC204 fraction collector. Semi-preparative HPLC was performed using an XBridge^{TM} C_{18} 5 μM, 19 x 150 mm column.

2.10.2 Materials

Commercially available compounds were purchased from Aldrich and were used without further purification.

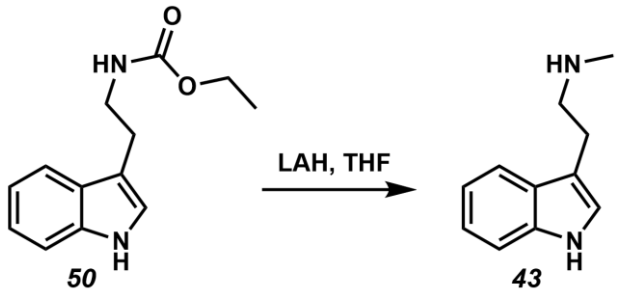
2.10.3 Experimental procedures

2.10.3.1 Synthesis of ethyl 2-(1H-indol-3-yl)ethylcarbamate (50)



In a flame-dried (under vacuum) three-necked flask, tryptamine (3.00 g, 18.7 mmol) was dissolved in 47 mL of freshly distilled DCM. Triethylamine (1.89 g, 18.7 mmol) was added to the reaction mixture and upon cooling to 0 ºC, ethyl chloroformate (2.03 g, 18.7 mmol) was syringed in dropwise maintaining the same temperature. When the addition was complete, the mixture was warmed to ambient temperature, at which time the organic layer was washed sequentially with water, 1M HCl, 5% $NaHCO_3$ solution, followed by washes with water and brine. The organic layer was dried over Na_2SO_4, solvent evaporated under vacuum, and purified by flash column chromatography 7:3 hexanes/ethylacetate give the product as an orange oil (3.8 g, 88%); ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.26 (br. s, 1H), 7.61 (d, *J* = 7.7 Hz, 1H), 7.37 (dt, *J* = 8.7 Hz, 1H), 7.22 (td, *J* = 7.7, 0.9 Hz, 1H), 7.14 (td, *J* = 7.7, 0.9 Hz, 1H), 7.00 (s, 1H), 4.78 (br. s, 1H), 4.13 (q, *J* = 7.3 Hz, 2H), 3.55 (m, 2H), 2.98 (t, *J* = 6.8 Hz, 2H), 1.23 (t, *J* = 7.3 Hz, 3H) ppm.

2.10.3.2 Synthesis of 2-(1H-indol-3-yl)-N-methylethanamine (43)

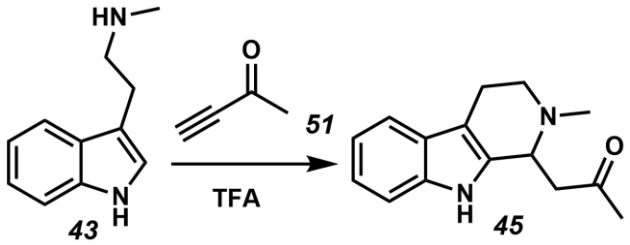


50 *43*

Lithium aluminum hydride (1.71 g, 45.0 mmol) was added to a THF (30mL) solution of carbamate (3.50 g, 15.0 mmol) at 0 ºC under nitrogen atmosphere. The reaction mixture was refluxed for 1.5 h. When the reaction is over, the reaction mixture was cooled to -78 ºC and diluted with THF (about 10 mL) and the excess of lithium

aluminum hydride was hydrolyzed by subsequent addition of water (6 mL), 15 % NaOH (6 mL) and water (3x6 mL). The suspension was warmed to room temperature and filtered. The white solid was washed with THF (30 mL). The organic layer was evaporated under vacuum, residual water was removed by the addition of brine and repeated extraction with DCM. Organic layers were combined and dried over Na_2SO_4, and the solvent removed under vacuum to give a off-white amorphous solid (1.94 g, 74.3 %). The crude product was deemed sufficiently pure and subjected to the next step without further purification. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.2 (br. s, 1H), 7.64 (d, *J* = 7.7 Hz, 1H), 7.37 (d, *J* = 8.2 Hz, 1H), 7.21 (td, *J* = 8.2, 0.9 Hz, 1H), 7.14 (td, *J* = 8.2, 0.9 Hz, 1H), 7.04 (bs, 1H), 3.00-2.97 (m, 2H), 2.93-2.90 (m, 2H), 2.44 (s, 3H) ppm.

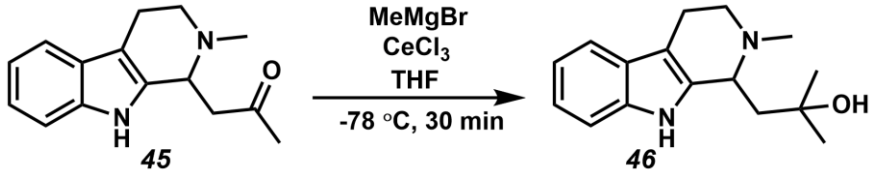
2.10.3.3 Synthesis of 1-(2-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indol-1-yl)propan-2-one (45)



To the solution of tryptamine **43** (1.1 g, 6.3 mmol) in 50 mL of DCM was added propargyl ketone **51** (1.17 mL, 6.6 mmol, 1.05 eq) and stirred for 1 h at room temperature. The reaction mixture was then treated with excess TFA (7.2 g, 63 mmol, 10.0 eq) for 15 minutes after which it was quenched by pouring into 50 mL of cold water. To this mixture, was slowly added a concentrated solution of sodium hydroxide (10 N)

until the pH is basic followed by extraction with DCM. The organic layer was washed with water, dried over anhydrous sodium sulfate (Na_2SO_4), filtered and concentrated under vacuum. The crude product was purified by silicagel chromatography using 9.6:0.4 DCM/methanol as eluent to give the product (1.52 g, 99%); ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.40 (bs, 1H), 7.49 (d, *J* = 7.7 Hz, 1 H), 7.31 (d, *J* =7.7 Hz, 1H), 7.16 (td, *J* = 7.7, 1.3 Hz, 1H), 7.09 (td, *J* = 8.2, 1.3 Hz, 1H), 4.09 (dd, *J* = 10.0, 2.7 Hz, 1H), 3.20 (dd, *J* = 18.3, 3.2 Hz, 1H), 3.11-3.00 (m, 1H), 2.99-2.85 (m, 3H), 2.50 (s, 3H), 2.21 (s, 3H) ppm; ^{13}CNMR (100.52 MHz, $CDCl_3$) δ 210.2, 135.65, 134.5, 126.8, 121.6, 119.2, 118.1, 111.0, 107.0, 54.4, 49.6, 47.9, 42.0, 30.7, 18.1 ppm; IR (methanol) 3396, 3272, 2939, 2844, 1705, 1466, 1447, 1417, 1362, 1303, 1156, 1106, 1059, 1026, 741, 691 cm^{-1}.

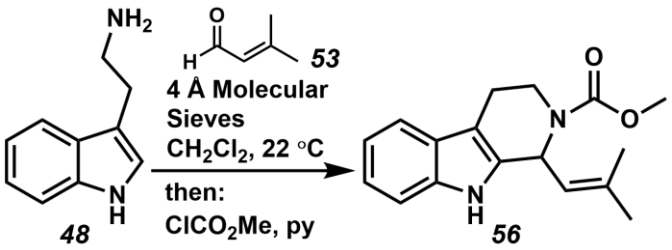
2.10.3.4 Synthesis of 2-methyl-1-(2-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indol-1-yl)propan-2-ol (46)



The solution of anhydrous cerium chloride (1.53 g, 4.12 mmol, 2.0 eq) in 20 mL of dry THF was stirred for 21 hours at room temperature. At -78 °C, was added t-BuLi (1.7 M in pentane, 1.2 mL, 2.06 mmol, 1.0 eq). Solution turns to pale pink color. Methyl magnesium bromide (3 M in hexane, 1.37 mL, 4.12 mmol, 2.0 eq)) was then added at the same temperature and let it stir at -78 °C for 15 minutes and then at 0 °C for 20-30 minutes. The reaction was brought back to -78 °C and the starting material (0.50 mg, 2.06

mmol, 1.0 eq) in 10 mL of THF was added. After 15 minutes the reaction was quenched with saturated ammonium chloride, extracted with ethyl acetate (2 x 40 mL). Organic layer was then washed with brine, dried with sodium sulfate, filtered and evaporated under vacuum. The crude material was purified by silicagel chromatography using 9.6:0.4 DCM/methanol as eluent to give the product (85 mg, 16%); ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.86 (bs, 1H), 7.51 (d, *J* = 7.3 Hz, 1H), 7.31 (d, *J* = 7.7 Hz, 1H), 7.18 (td, *J* = 7.3, 1.3 Hz, 1H), 7.12 (td, *J* = 7.3, 1.3 Hz, 1H), 4.01 (dd, *J* = 11.4, 3.2 Hz, 1H), 3.34-3.26 (m, 1H), 2.96-2.87 (m, 2 H), 2.64-2.56 (m, 1H), 2.53 (s, 3H), 2.06 (dd, *J* = 11.4, 3.2 Hz, 1H), 1.74 (dd, *J* = 14.6, 3.6 Hz, 1H), 1.39 (s, 3H), 1.22 (s, 3H) ppm; ^{13}CNMR (100.52 MHz, $CDCl_3$) δ 135.8, 133.3, 127.1, 121.8, 119.5, 118.2, 110.8, 107.4, 70.4, 56.8, 44.6, 41.0, 31.3, 28.9, 16.0 ppm; IR (DCM) 3200, 2966, 2937, 1447, 1468, 1363, 1264, 1298, 1237, 1190, 1162, 1097, 1060, 1029, 1009, 981, 956, 908, 789, 732 cm^{-1}.

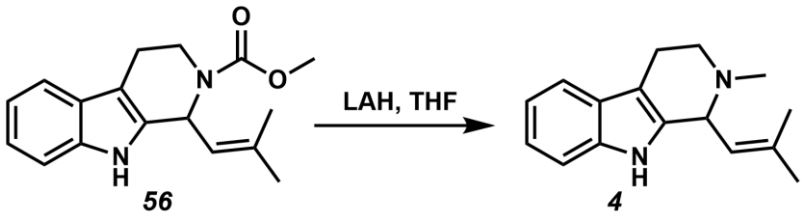
2.10.3.5 Synthesis of methyl 1-(2-methylprop-1-enyl)-3,4-dihydro-1H-pyrido[3,4-*b*]indole-2(9*H*)-carboxylate (56)



To a solution of tryptamine (1.28 g, 7.98 mmol) and 3-methyl 2-butenal (0.84 mL, 8.78 mmol) in DCM (40 mL) was added 15 g of powdered 4 Å molecular sieves. The suspension was stirred for 16 hr at room temperature. Pyridine (2.48 mL, 31.9 mmol) and

methyl chloroformate (1.35 mL, 17.5 mmol) were added at 0 °C, and stirring continued for 5 hr at room temperature. The solution was filtered through celite, and the celite bed was washed with an additional DCM (40 mL). The organic layer was extracted with 1N HCl (2x30 mL) and then washed with brine solution (20 mL). The organic layer was dried over Na_2SO_4 and then was evaporated under vacuum. The solid was triturated with 10% ether in hexane (20 mL) to afford a brown amorphous solid (1.97 g, 87% yield). The triturated material was sufficiently pure to carry on to the next step. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.64 (bs, 1H), 7.47 (d, *J* = 7.45 Hz, 1H), 7.29 (d, *J* = 8.0 Hz, 1H), 7.14 (dt, *J* = 6.8 Hz, 1.15 Hz, 1H), 7.09 (dt, *J* = 6.8 Hz, 1.15 Hz, 1H), 5.93 (bs, 1H), 5.35 (dt, *J* = 9.1 Hz, 1.7 Hz), 4.45 (brs, 1H), 3.74 (s, 3H), 2.79-2.85 (dt, *J* = 15.4, 5.1, 1H), 2.72-2.75 (dd, *J* = 14.8 Hz, 2.8 Hz, 1H), 1.76 (s, 3H), 1.98 (bs, 3H); ^{13}CNMR (100.52 MHz, $CDCl_3$) δ 156.1, 136.1, 127.0, 122.2, 121.9, 119.6, 118.2, 110.9, 52.8, 49.7, 39.3, 26.0, 21.5, 18.4 ppm; IR (neat) 3398, 2906, 2849, 11682, 1460, 1442, 1406, 1374, 1361, 1295, 1264, 1189, 1165, 1143, 1111, 1095, 1029, 998, 984, 909, 879, 838, 804, 760, 750, 736, 690, 666 cm^{-1}.

2.10.3.6 Synthesis of borrerine: (2-methyl-1-(2-methylprop-1-enyl)-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole (4)



56 *4*

To a solution of methyl carbamate **56** (0.50 g, 1.67 mmol) in 25 mL of THF was added lithium aluminum hydride (0.38 g, 10.06 mmol) portion-wise at 0 °C. The reaction mixture was stirred while refluxing for 3 h, and then quenched with 2 mL of water, 4 mL 15% NaOH, and 2 mL of water in sequence at -78 °C. The resulting suspension was brought to room temperature and filtered using a buchner funnel. The solid was washed with 10 mL of THF. The combined organic solvents were evaporated under vacuum. The water layer was extracted with DCM (3x20 mL). The organic layer was washed with brine (10 mL) and then dried over Na_2SO_4. The solvents were evaporated under vacuum. The crude material was purified via flash column chromatography using 96:4 DCM/ methanol as an eluent on silica gel. Borrerine (352 mg, 88% yield) was obtained as off-white amorphous solid. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.55 (bs, 1H), 7.46 (d, *J* = 7.45 Hz, 1H), 7.27 (d, *J* = 8.0 Hz, 1H), 7.0-7.13 (m, 2H), 5.20 (dt, *J* = 9.1 Hz, 1.1 Hz, 1H), 4.05(d, *J* = 9.1 Hz, 1H), 3.16 (dddd, *J* = 5.1, 1.7 Hz, 1 H), 2.91-2.98 (m, 1H), 2.72-2.77 (dq, *J* =15.4, 2.2 Hz, 1H), 2.59-2.65 (dt, *J* = 10.8, 4.0 Hz, 1H), 2.43 (s, 3H), 1.85 (s, 3H), 1.87 (s 3H) ppm; ^{13}CNMR (125.77 MHz, $CDCl_3$) δ 137.3, 136.0, 134.5, 127.6, 124.6, 121.3, 119.3, 118.2, 110.8, 108.0, 60.0, 53.3, 43.4, 26.2, 21.6, 18.6 ppm; IR (DCM) 2909, 2843, 2788, 1465, 1445, 1365, 1306, 1279, 1263, 1222, 1184, 1163, 1113, 1056, 1037, 1009, 966, 920, 801, 736, 701 cm^{-1}.

2.10.3.7 General procedure for the optimization of the acid dimerization of borrerine to give isoborreverine, flinderole A, or desmethylflinderole C (Table 2.1). To a solution of borrerine (50 mg, 0.208 mmol) dissolved in 0.5 mL benzene or chloroform was added trifluoroacetic acid (31.8 μL, 0.416 mmol) at room temperature.

The reaction was brought to 65 °C, and stirring was continued for 30 min. The solvents were then evaporated under vacuum. The crude material was passed through a silica gel column using methanol as the eluent. This material was then analyzed via ^{1}H NMR. Relative product percentages were determined by comparing the δ 5.40 peaks of isoborreverine, the δ 5.26 peaks of desmethylflinderole C and the δ 5.21 peaks of flinderole A. Individual products could be isolated using semi-preparative HPLC equipped with a reverse phase column.

The spectral characteristics for isoborreverine[8,18] and flinderole A[19] were identical to those previously reported.

Desmethylflinderole C: ^{1}H NMR (500 MHz, $CDCl_3$) δ 11.16 (s, 1H), 8.54 (m, 1H), 8.47 (m, 1H), 7.48-7.54 (m, 2H), 7.22-7.26 (m, 2H), 7.07 (t, *J* = 8.02 Hz, 1H), 6.94-6.99 (m, 3H), 6.81 (d, *J* = 16.0 Hz, 1H), 6.59 (d, *J* = 16.0 Hz, 1H), 5.21 (d, *J* = 9.7 Hz, 1H), 4.34 (q, *J* = 8.8 Hz, 1H), 2.85-3.0 (m, 8H), 2.68 (m, 1H), 2.57 (t, *J* = 5.7 Hz, 6H), 2.26 (m, 1H), 1.81 (s, 3H), 1.76 (s, 3H), 1.69 (s, 3H)

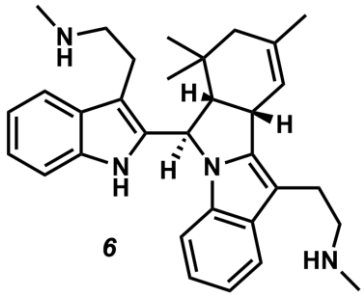
2.10.3.8 General procedure for the optimization of the acid dimerization of borrerine to give isoborreverine, flinderole A, or desmethylflinderole C (Table 2.3 and 2.4). To a solution of borrerine (10 mg, 0.0416 mmol) in 0.2 mL benzene was added Lewis acid or Brønsted acid and stirred for reported time and temperatures. The solvent was then evaporated under vacuum. The crude material was passed through a silica gel

column using methanol as the eluent. This material was then analyzed via ^{1}H NMR. Relative product percentages were determined by comparing the δ 5.40 peaks of isoborreverine, the δ 5.26 peaks of desmethylflinderole C and the δ 5.21 peaks of flinderole A.

2.10.3.9 General procedure for the preparation of flinderole B, C, and dimethylisoborreverine (Scheme 2.10). To a solution of borrerine (10 mg, 0.0416 mmol) dissolved in 0.2 mL chloroform was added MeOTf (4.56 μL, 0.0416 mmol) at 0 °C. TFA (6.37 μL, 0.0832) was added at 0 °C, and then the reaction mixture was brought to room temperature. Stirring was continued for 20 min. The reaction was then quenched with amberlyst 21-A resin (100 mg) and stirred for 20 min at room temperature. After filtration of the solution through cotton, the solvents were evaporated under vacuum. This material was then analyzed via ^{1}H NMR. Relative product percentages were determined by comparing the δ5.48 peaks of dimethylisoborreverine, the δ 5.28 peaks of flinderole B and the δ 5.23 peaks of flinderole C. Individual products could be isolated using semi-preparative HPLC equipped with a reverse phase column.

The spectral characteristics for dimethylisoborreverine[8,18], flinderole B and flinderole C[19] were identical to those previously reported.

2.10.3.10 Synthesis of isoborreverine (6)

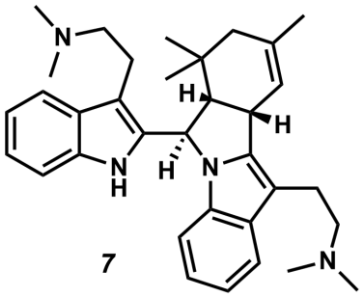


6

The solution of borrerine (10 mg, 0.0416 mmol) and trifluoroacetic acid (6.37 μL, 0.0832 mmol) was stirred at 65 °C for 48 hr. The reaction mixture was diluted with DCM (2 mL). Amberlyst 21 Å resin (100 mg) was added and stirring continued for 20 minutes to quench excess trifluoroacetic acid. The solution was filtered through a cotton plug and then evaporated under vacuum to afford isoborreverine as an off-white amorphous solid (8 mg, 80% yield).

Alternatively: To a solution of borrerine (50 mg, 0.208 mmol) in DCM (6 mL) at 0 °C was added $BF_3{\cdot}OEt_2$ (204.1 μL, 1.66 mmol). Stirring was continued at 22 °C for 3 days. The reaction mixture was diluted with DCM (6 mL) and quenched with saturated $NaHCO_3$ solution until a pH 7 was achieved. The crude product was extracted with 2% methanol in DCM (2x10 mL). The organic layer was dried over sodium sulfate and evaporated under vacuum to yield isoborreverine as a yellow amorphous solid (20 mg, 40% yield).

2.10.3.11 Synthesis of dimethylisoborreverine (7)



7

To a solution of borrerine (100 mg, 0.416 mmol) in chloroform (0.5 mL) at 0 °C was added methyl triflouromethane sulfonate (45.6 μL, 0.416 mmol). After 2 minutes, trifluoroacetic acid (63.7 μL, 0.832 mmol) was added at the same temperature. The reaction was brought to 22 °C and held at that temperature for 35 min. The reaction mixture was diluted with DCM (5 mL) and amberlyst 21 Å resin (1.0 g) was added and stirring continued for 20 minutes to quench the excess trifluoroacetic acid. The organic layer was filtered through a cotton plug, and the solvents were evaporated under vacuum. The crude material was purified using silicagel chromatography with 8:2 DCM/methanol as eluent to give a brown amorphous solid (60 mg, 56% yield).

2.11 References

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APPENDIX ONE

Spectra Relevant to Chapter 2:

Biomimetic Synthesis of Flinderoles

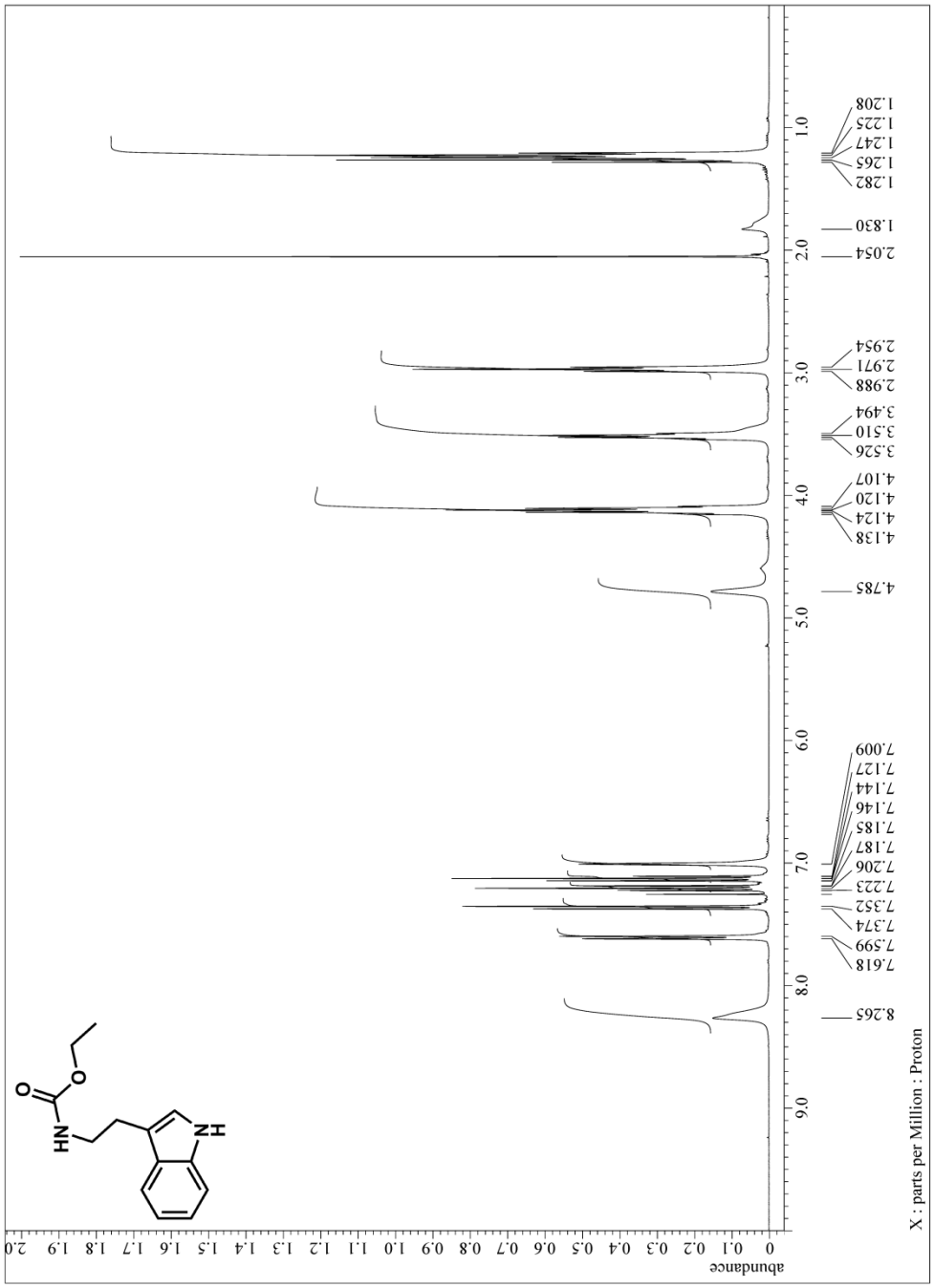


Figure A.1.1 ^{1}H NMR ($CDCl_3$) of compound **50**

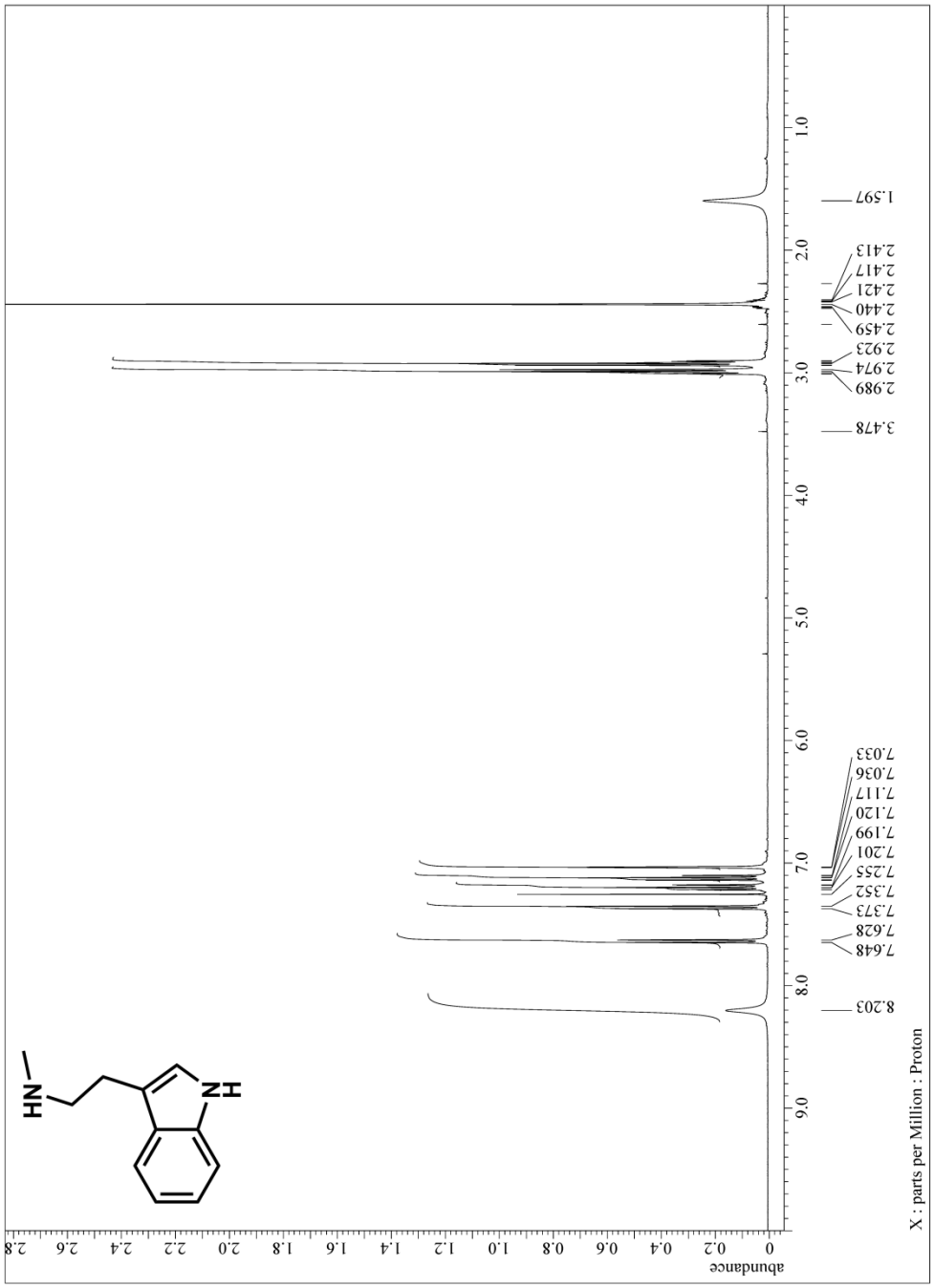


Figure A.1.2 ^{1}H NMR ($CDCl_3$) of compound **43**

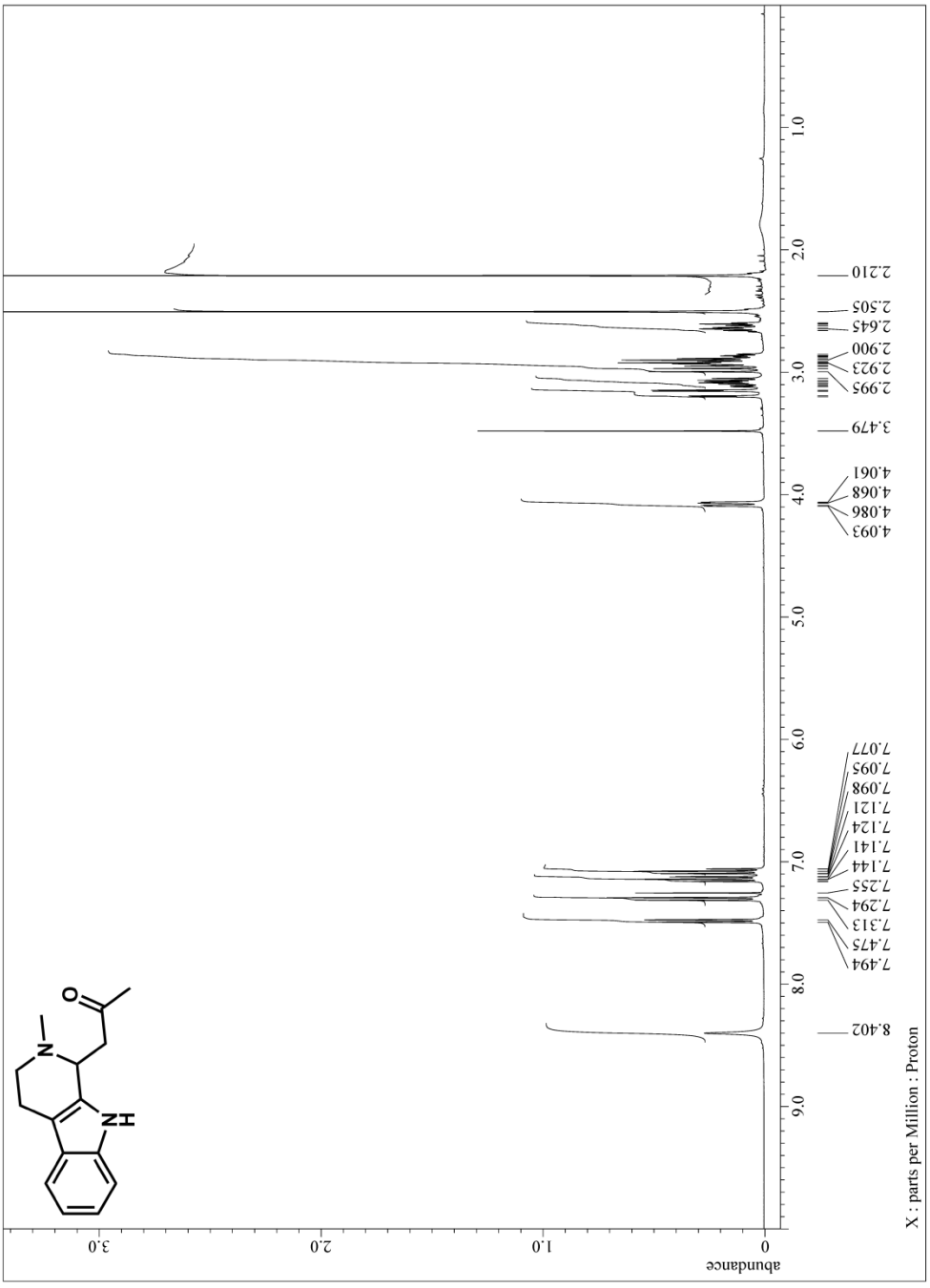


Figure A.1.3 ^{1}H NMR ($CDCl_3$) of compound **45**

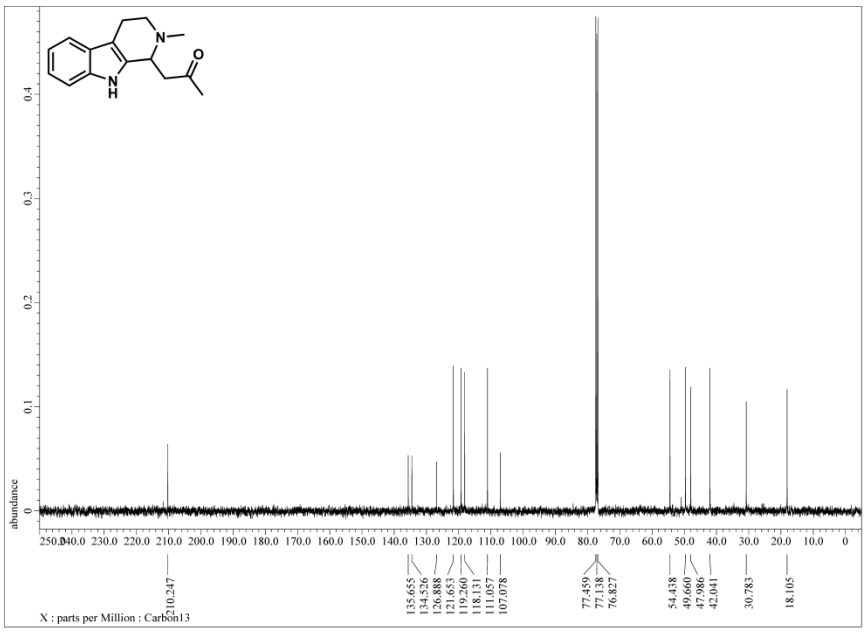


Figure A.1.4 ^{13}C NMR ($CDCl_3$) of compound **45**

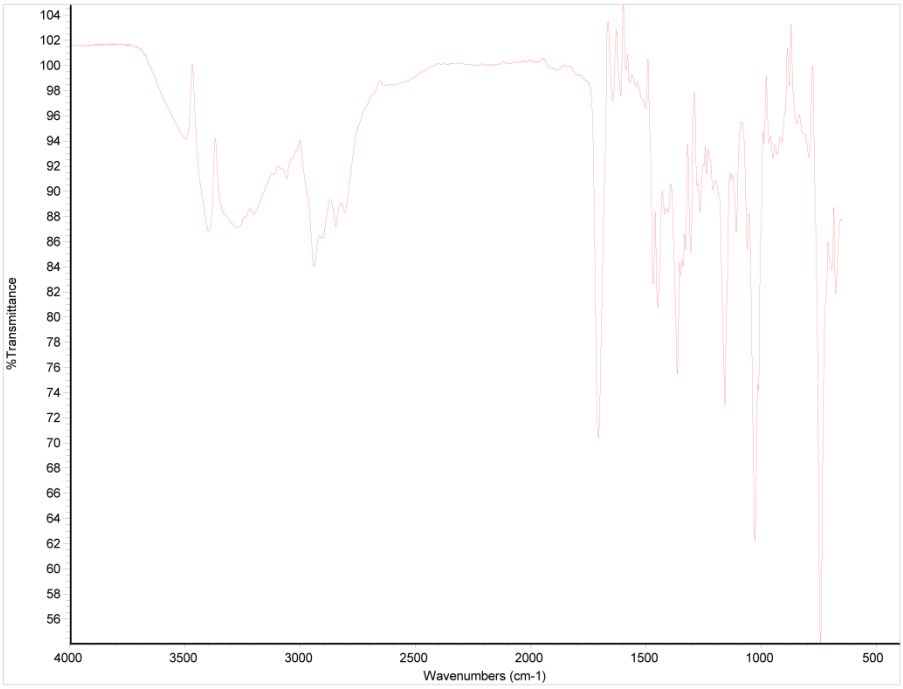


Figure A.1.5 IR spectrum of compound **45**

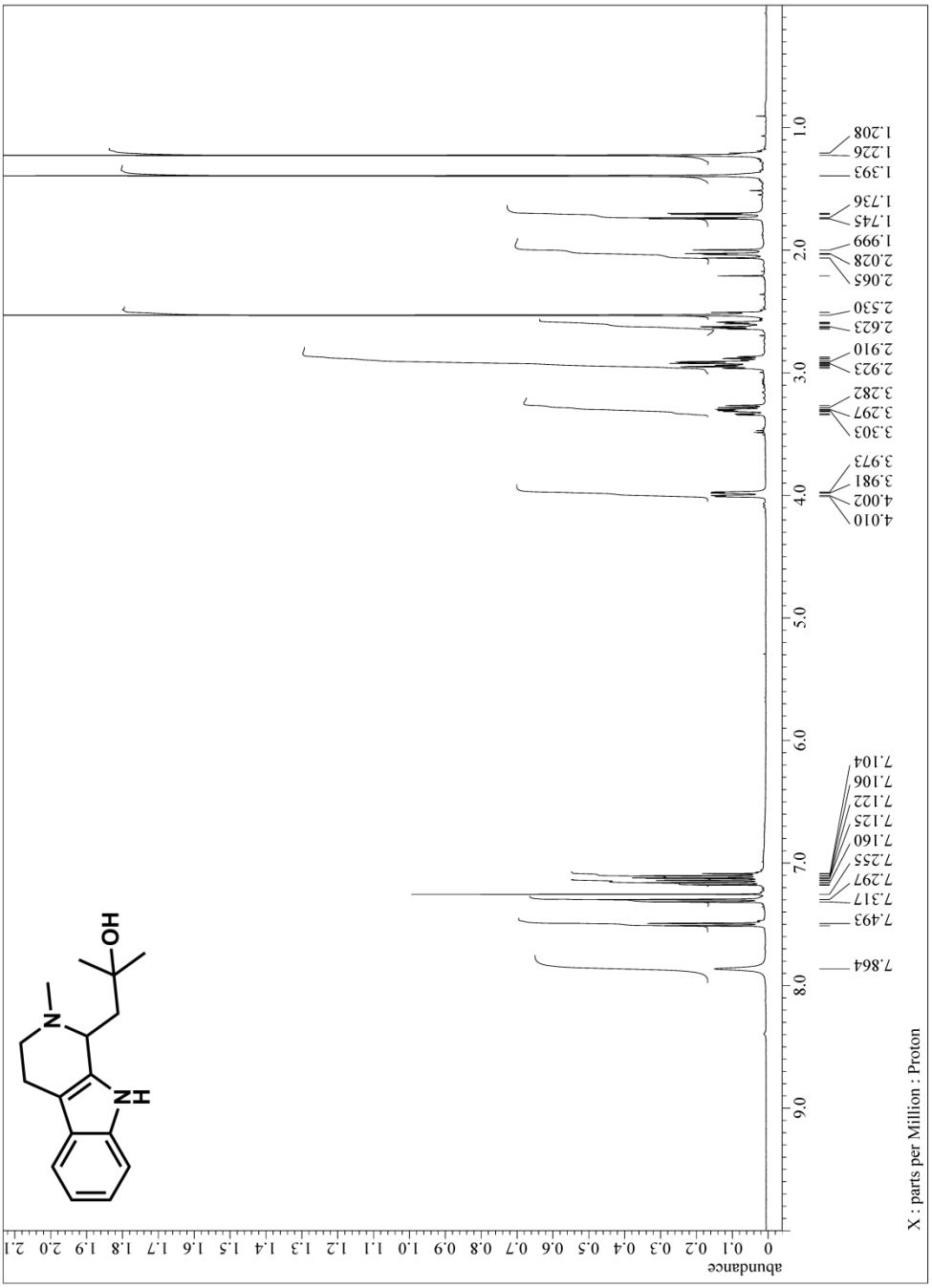


Figure A.1.6 ^{1}H NMR ($CDCl_3$) of compound **46**

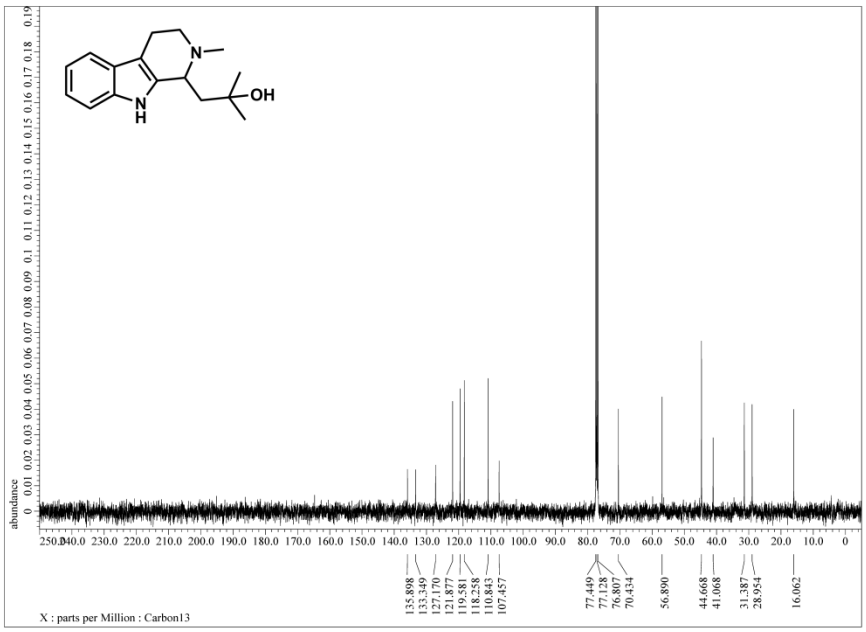


Figure A.1.7 ^{13}C NMR ($CDCl_3$) of compound **46**

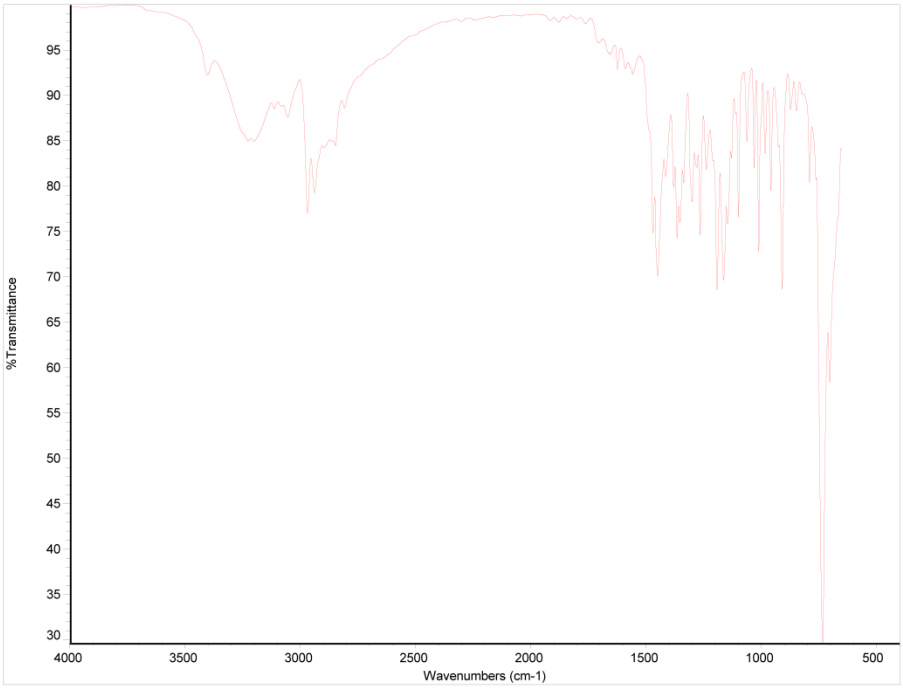


Figure A.1.8 IR spectrum of compound **45**

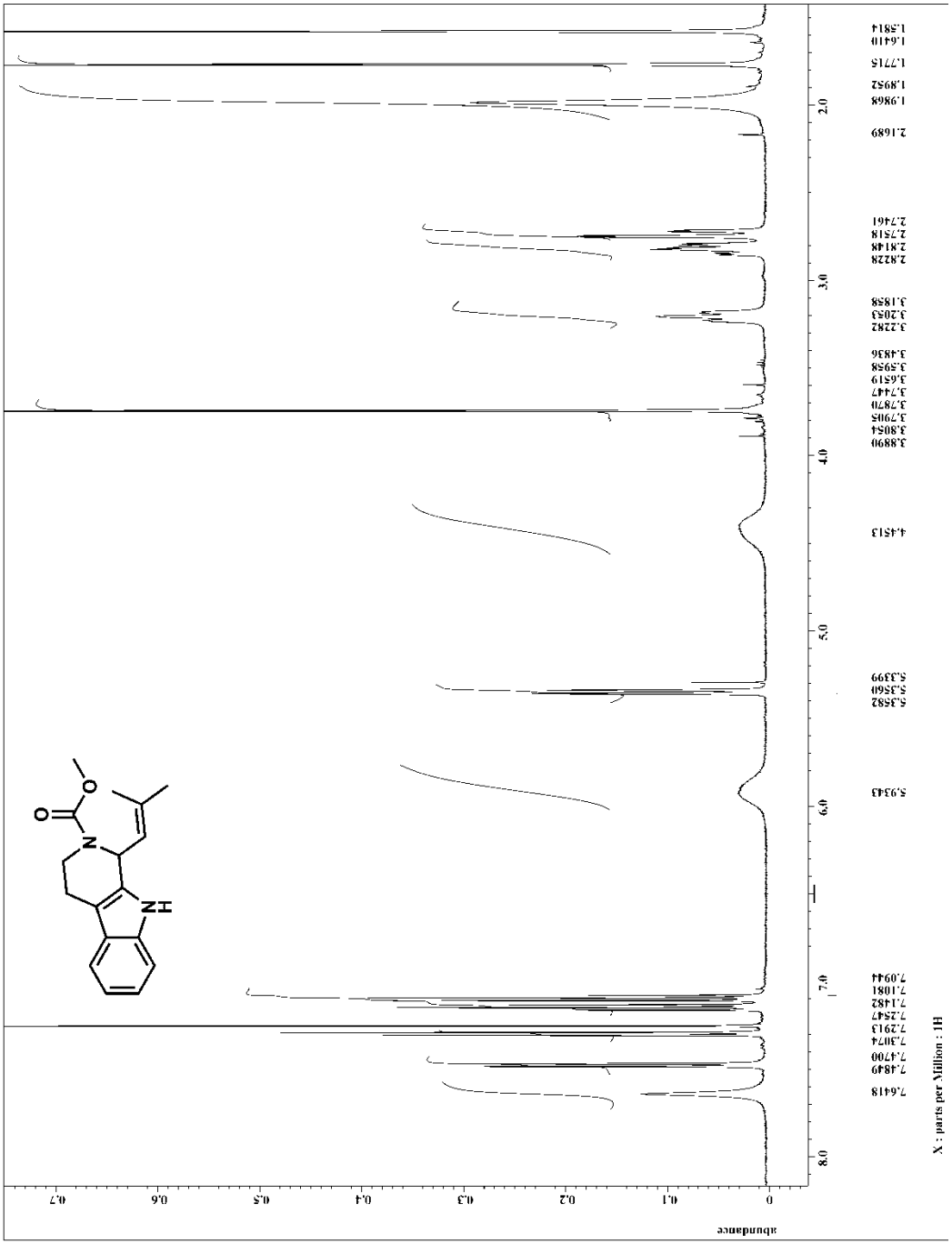


Figure A.1.9 ^{1}H NMR ($CDCl_3$) of compound **56**

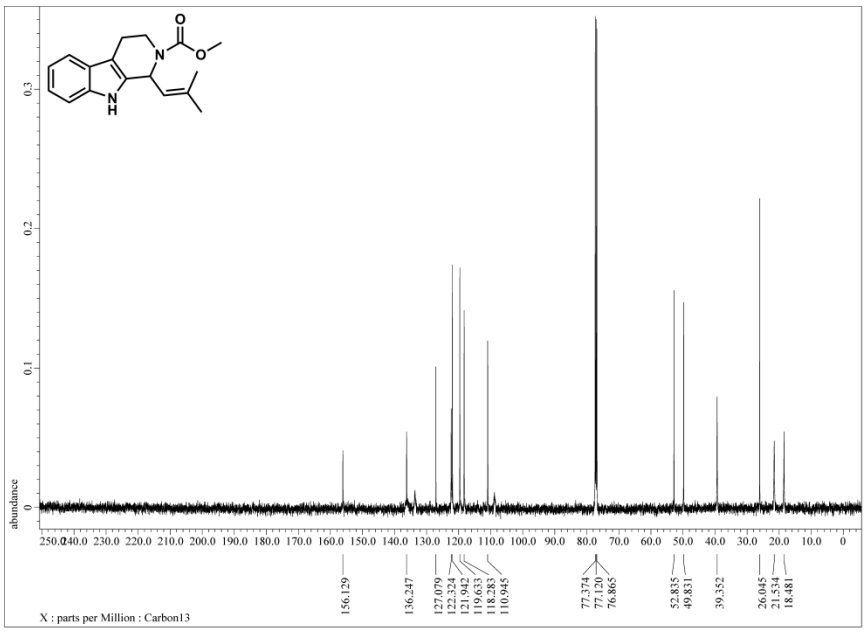


Figure A.1.10 ^{13}C NMR ($CDCl_3$) of compound **56**

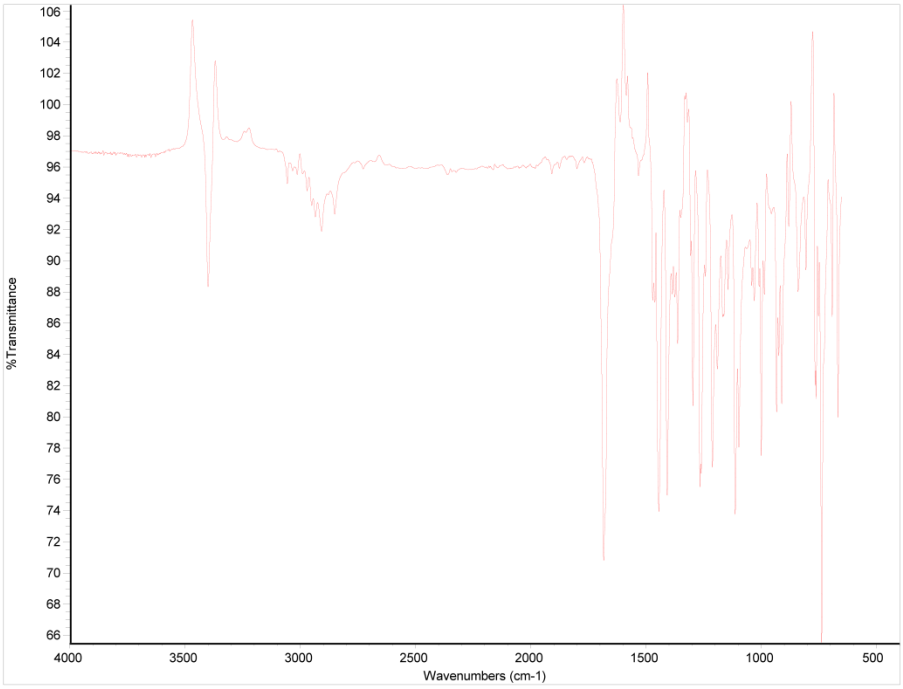


Figure A.1.11 IR spectrum of compound **56**

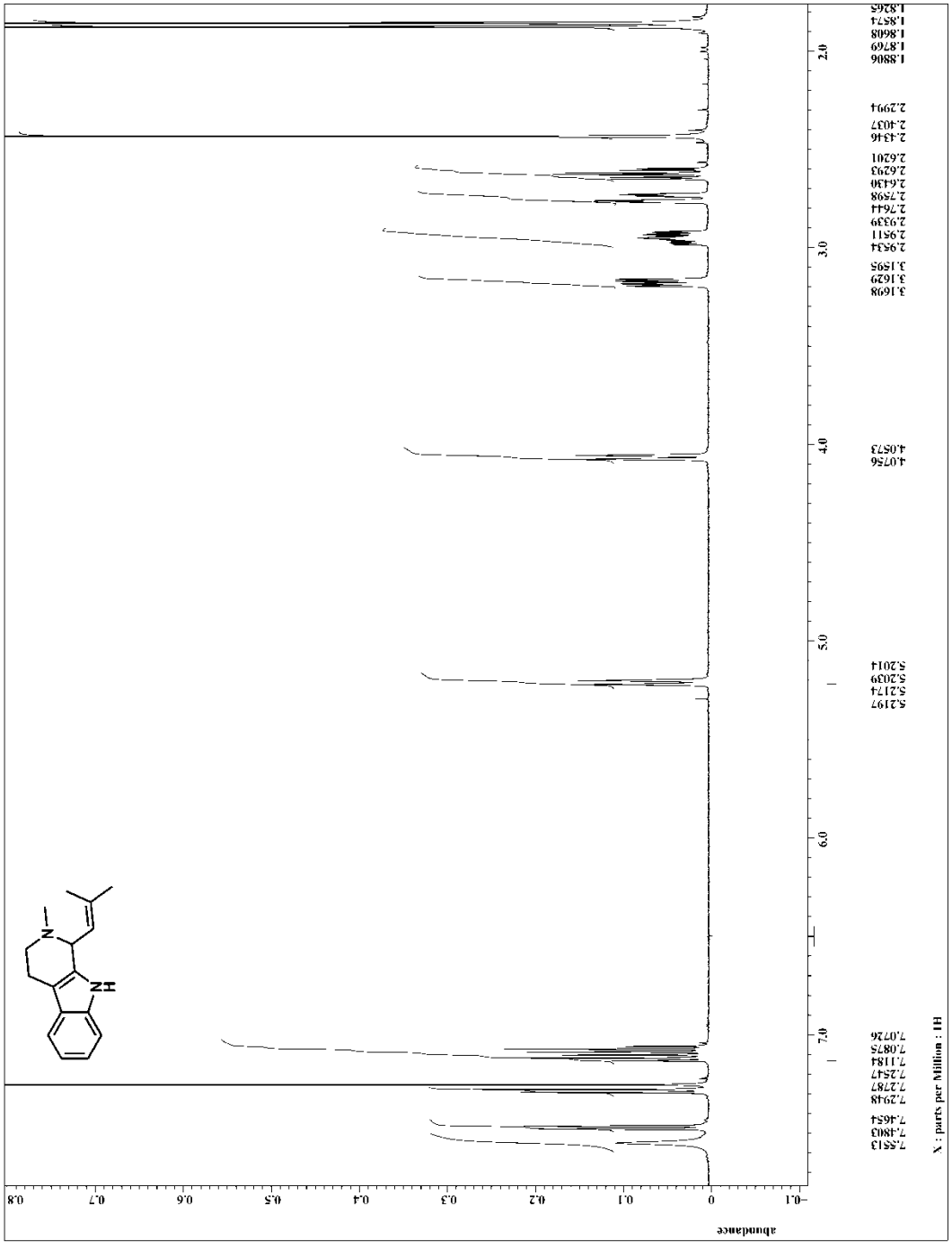


Figure A.1.12 ^{1}H NMR ($CDCl_3$) of compound **4**

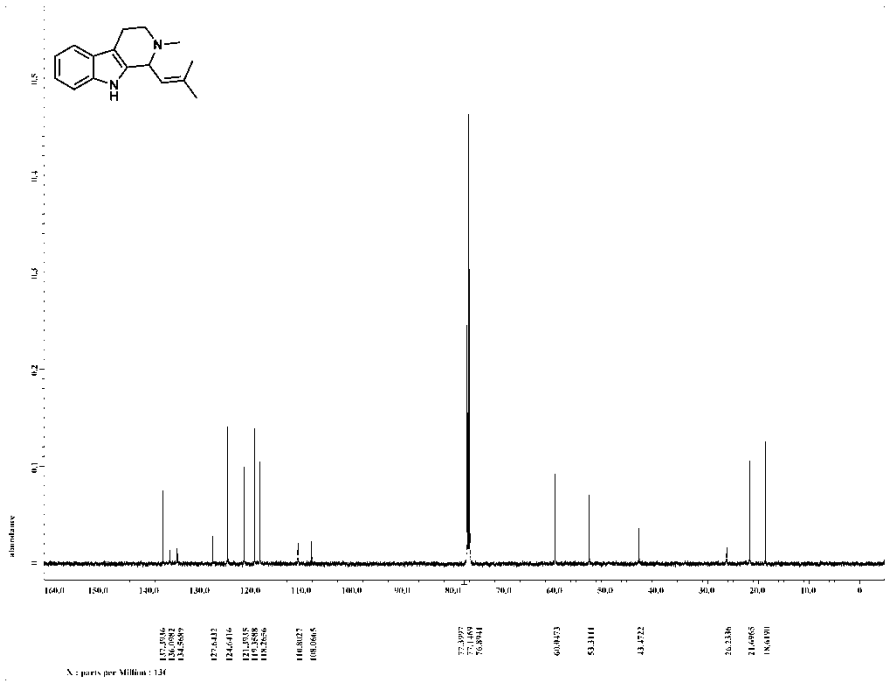


Figure A.1.13 ^{13}C NMR ($CDCl_3$) of compound **4**

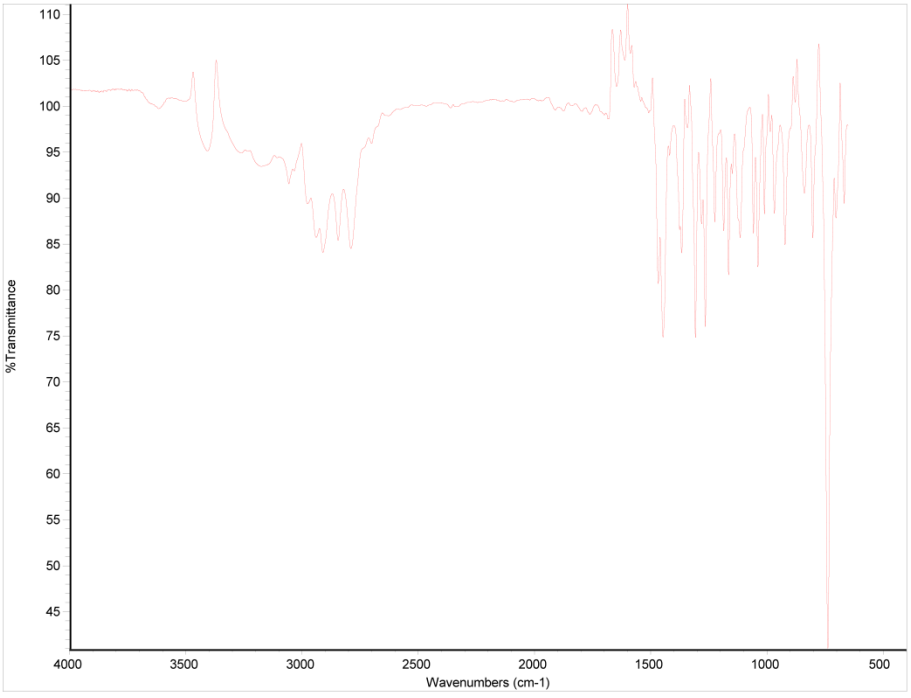


Figure A.1.14 IR spectrum of compound **4**

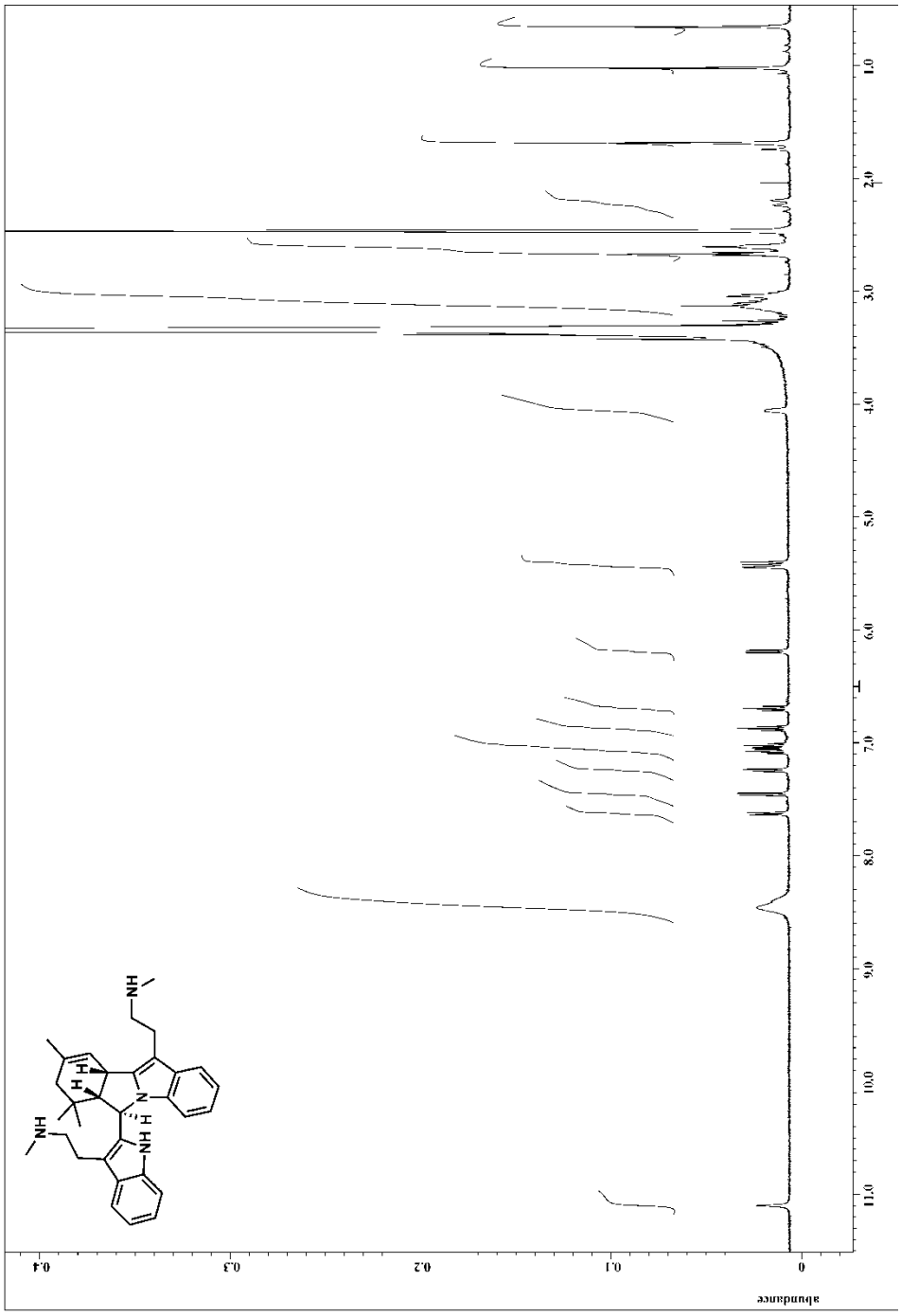


Figure A.1.15 ^{1}H NMR (DMSO-d6) of compound **6**

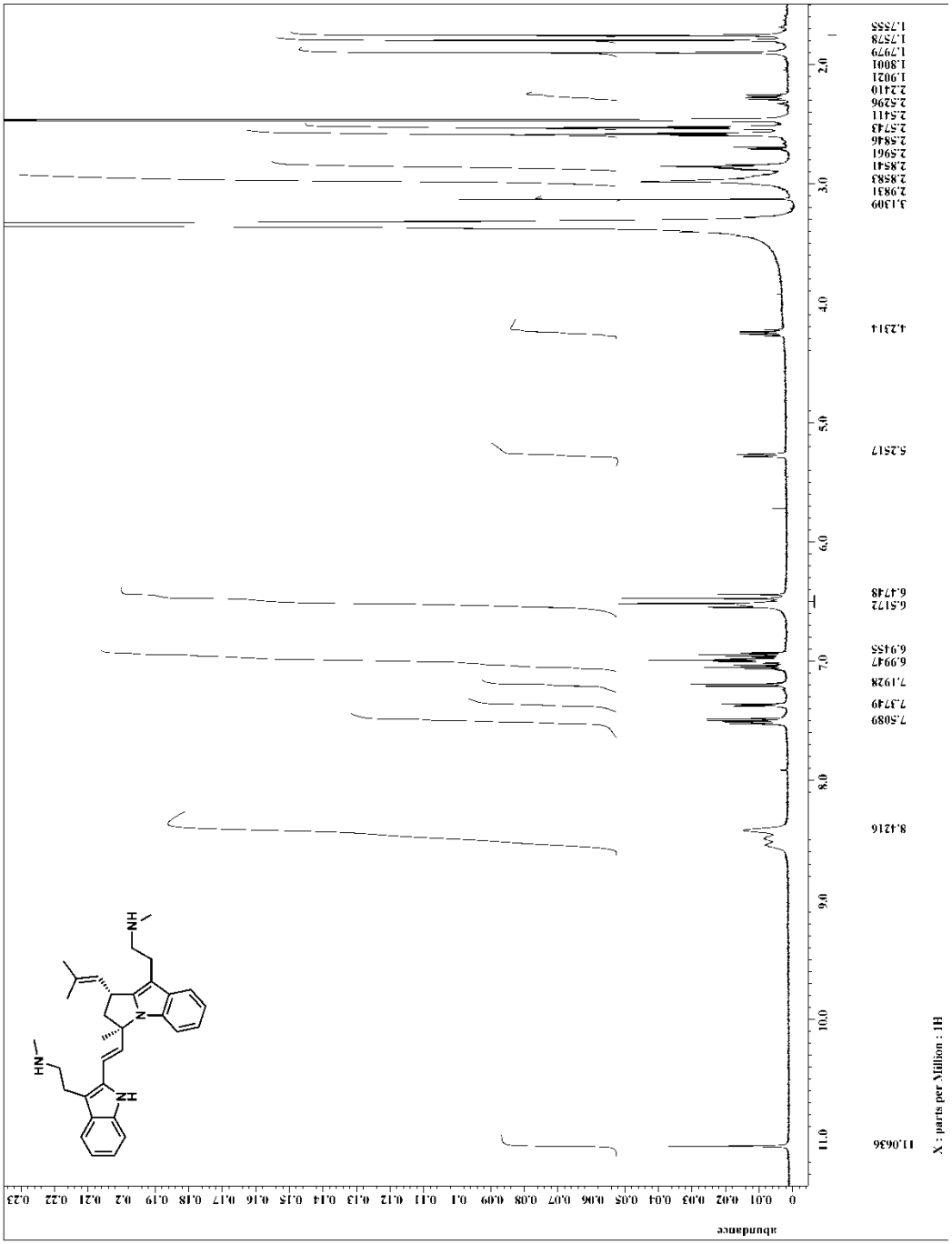


Figure A.1.16 ^{1}H NMR (DMSO-d6) of compound **11**

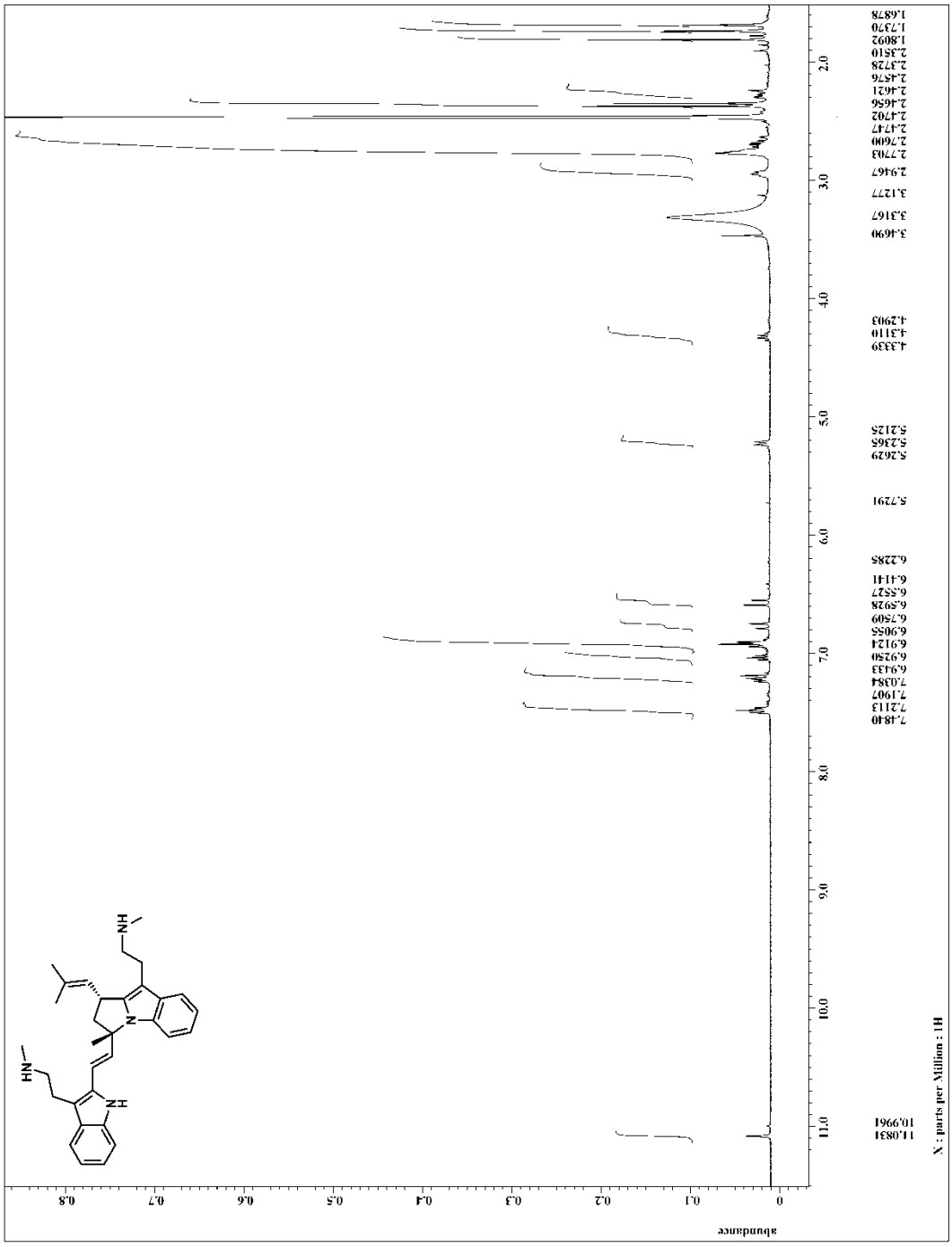


Figure A.1.17 ^{1}H NMR (DMSO-d6) of compound **57**

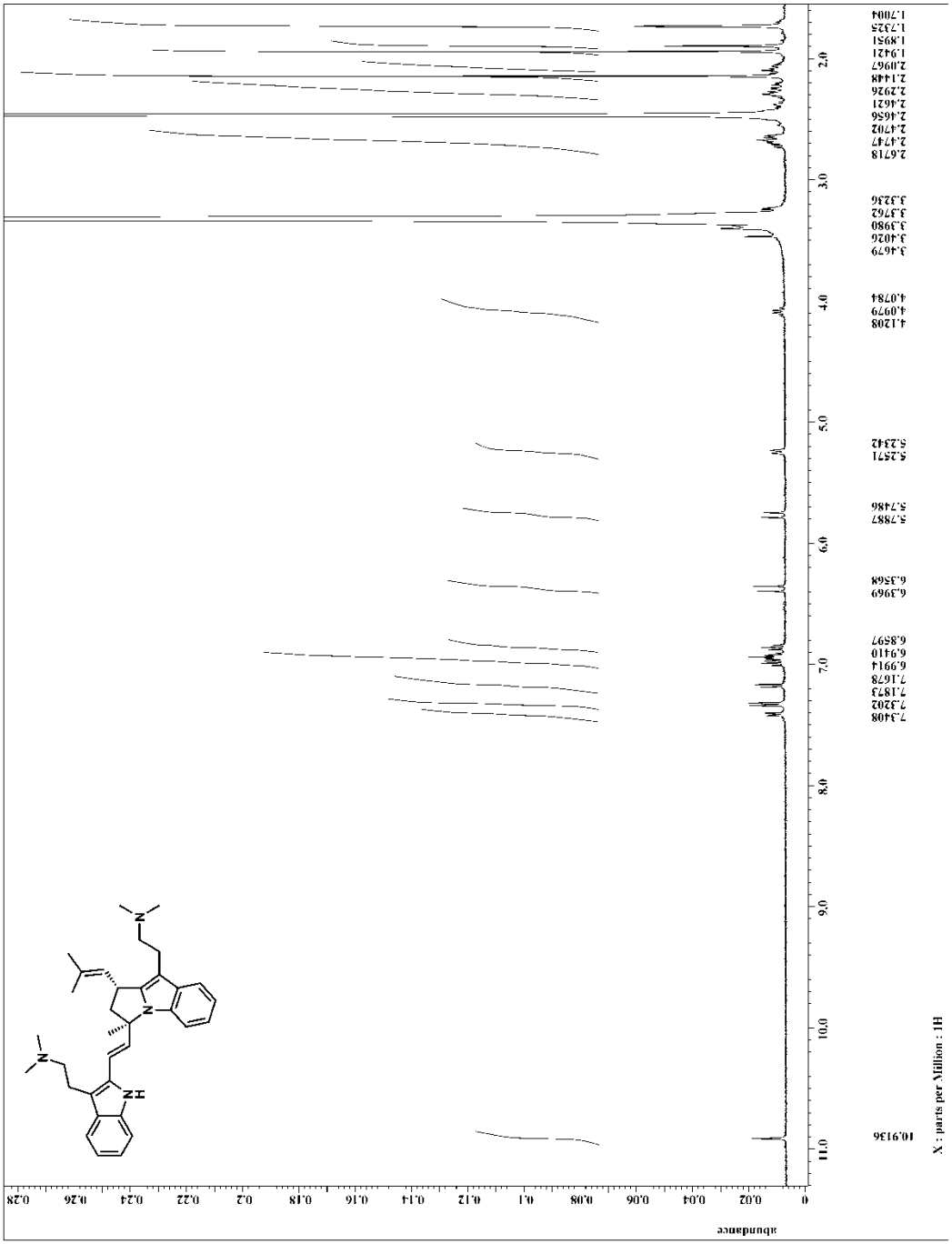


Figure A.1.18 ^{1}H NMR (DMSO-d6) of compound **12**

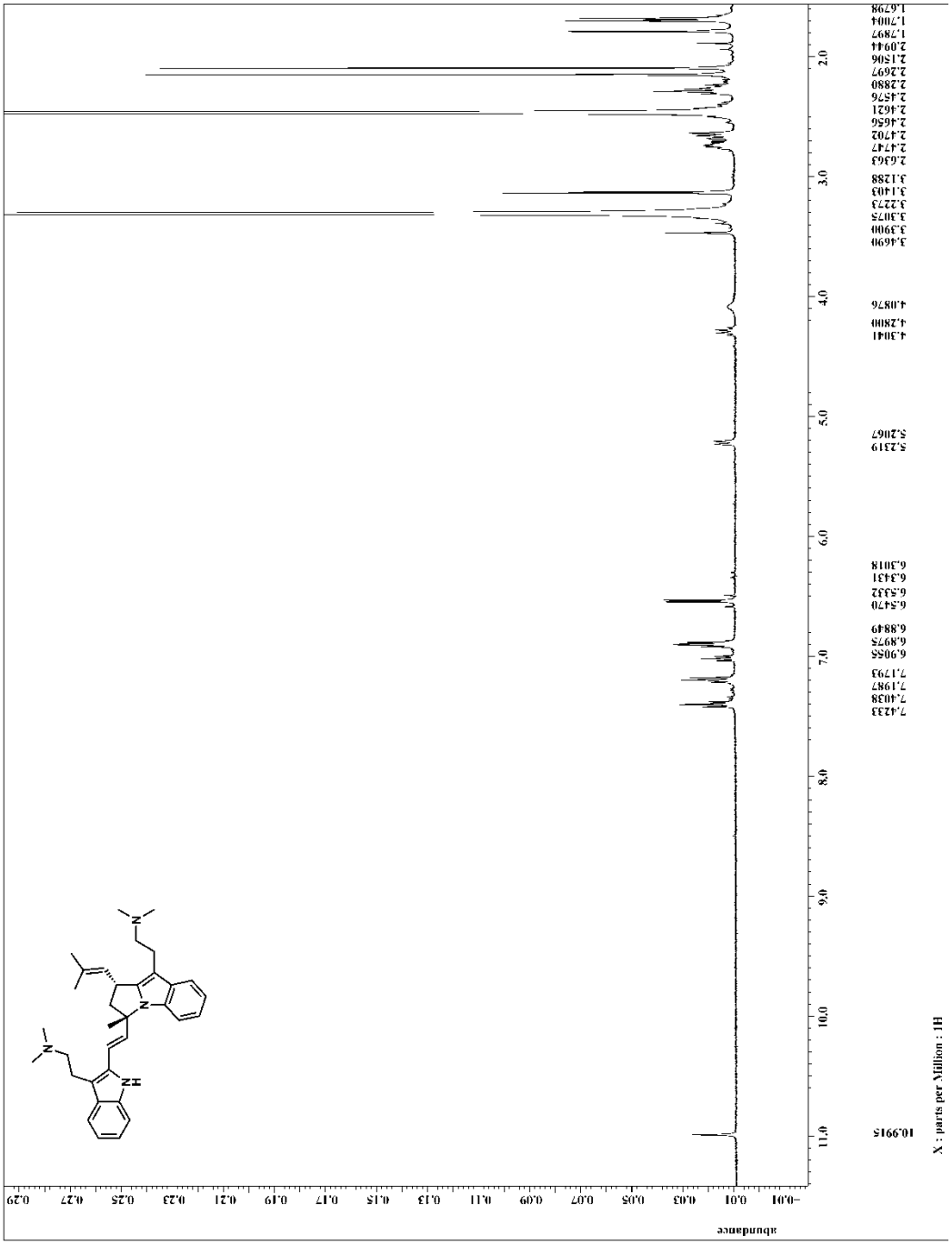


Figure A.1.19 ^{1}H NMR ($CDCl_3$) of compound **13**

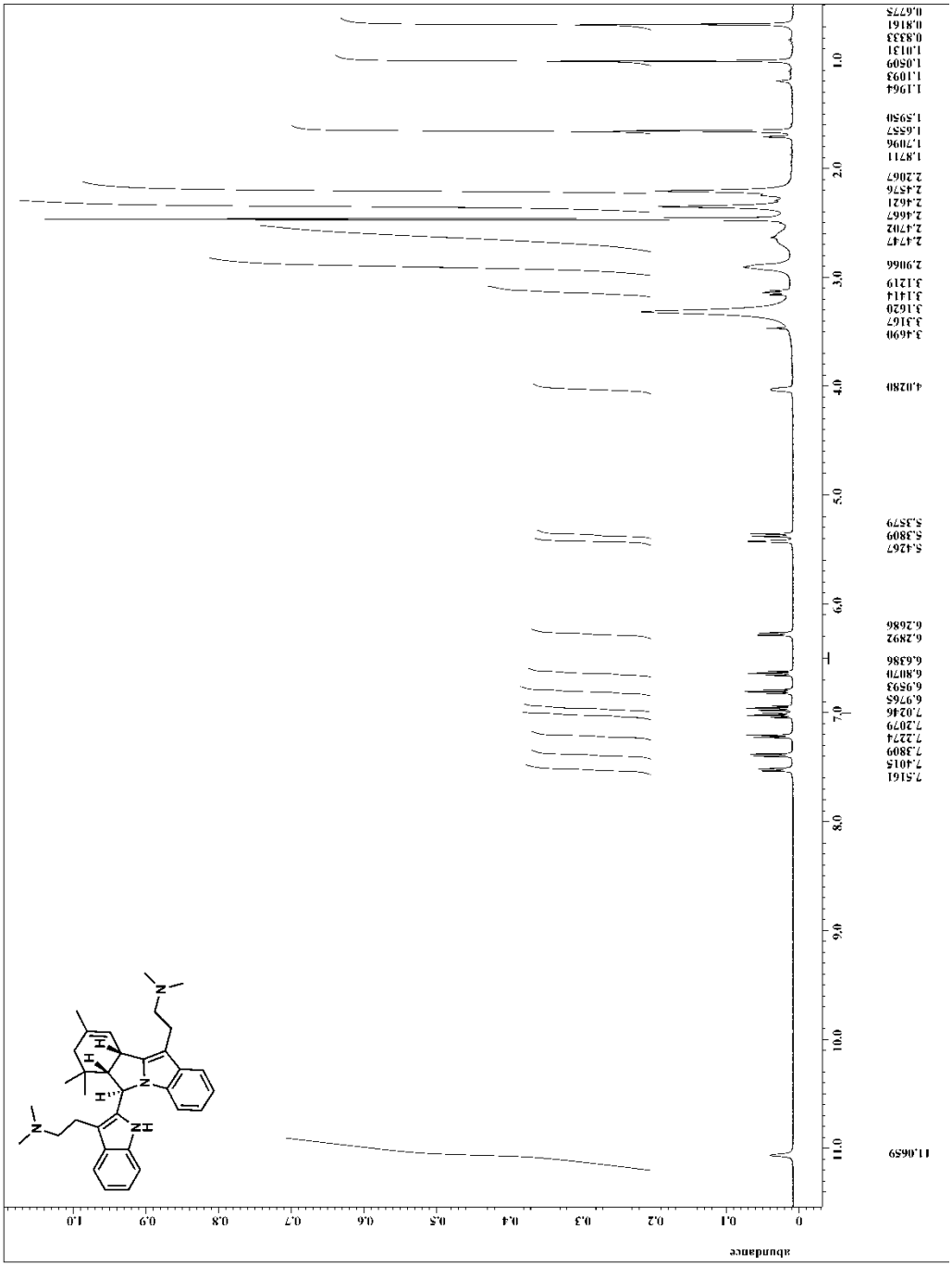


Figure A.1.20 ^{1}H NMR (DMSO-d6) of compound **7**

CHAPTER 3

BIOSYNTHESIS AND OPTICAL ACTIVITY OF THE FLINDEROLES[10,11]

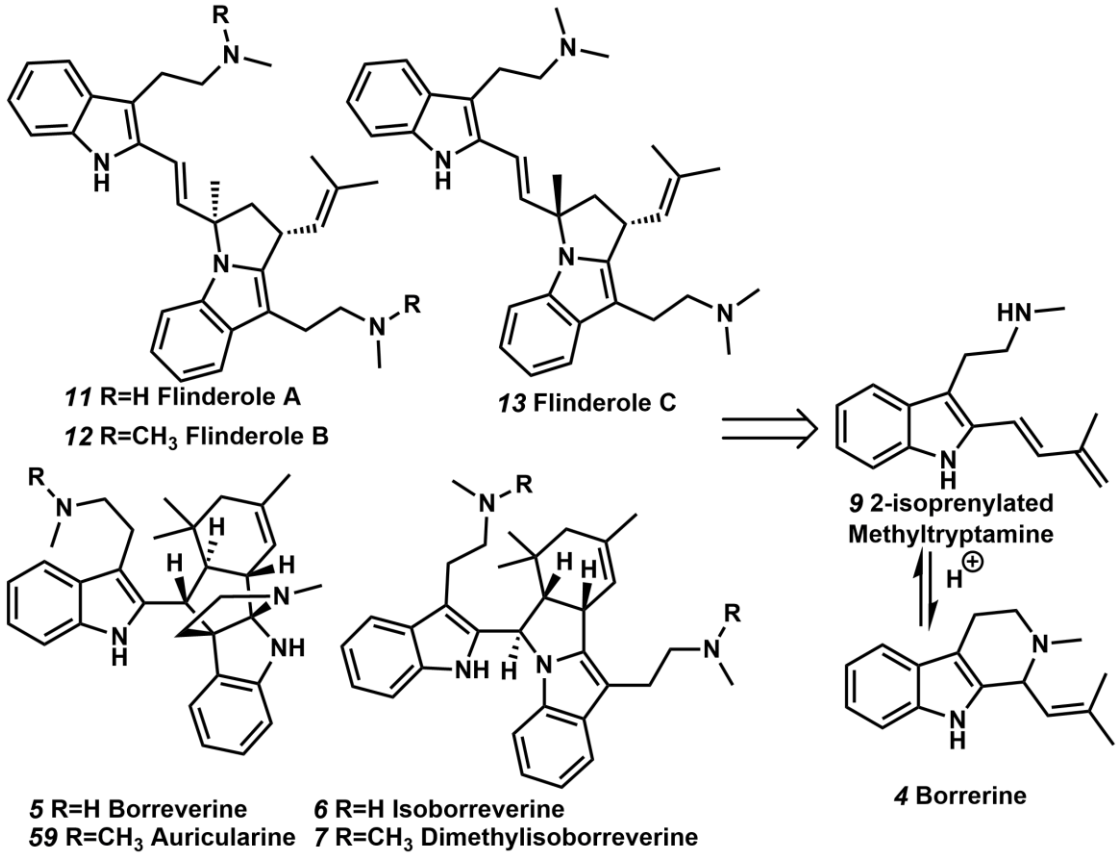
3.1 Introduction

Generally, the synthesis of complex natural products in a laboratory is achieved in multiple steps comprised of stepwise chemical transformations, isolations, and purifications.[1] However, the extraction of complex but structurally similar natural products from a specific genus of fauna or flora suggests that nature can generate complex structural scaffolds using simple precursors. For example, a 2-isoprenylated methyltryptamine building block may be hypothesized for the biosynthesis of flinderoles A–C (**11**–**13**),[2] borreverine (**5**)[3], isoborreverine (**6**),[4] dimethylisoborreverine (**7**),[5] auricularine (**59**),[6] and their derivatives (Scheme 3.1).[5,7]

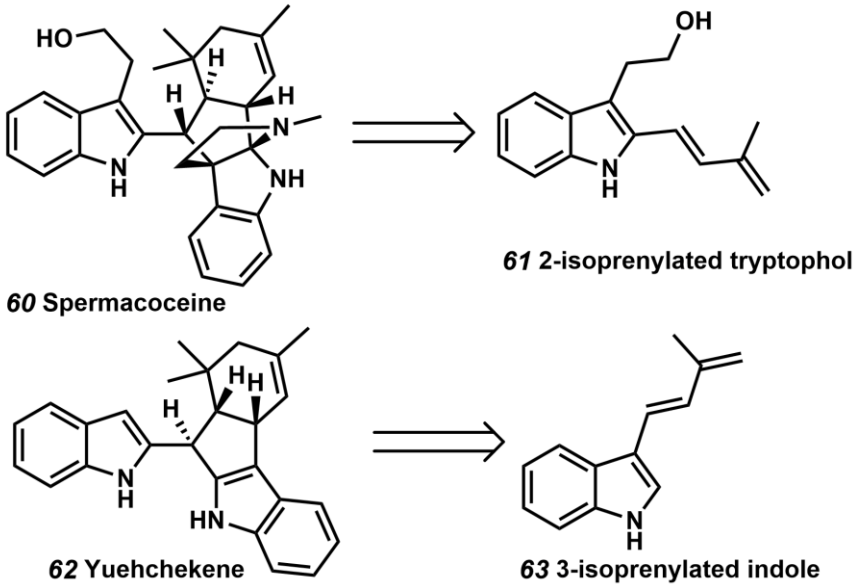
Work in the 1970's on the *Flindersia* alkaloids provides valuable information about the biosynthesis of the borreverines. The lack of optical activity of natural borreverines and the co-occurrence of borrerine (**4**) may suggest that acid was the only reagent required to dimerize borrerine to generate borreverines.[4] This hypothesis does not require the participation of any enzyme in the dimerization, and that hypothesis was validated by a synthetic attempt wherein borrerine (**4**), a cyclized congener of 2-isoprenyl methyltryptamine **9**, was treated with TFA to yield borreverine (**5**) and isoborreverine (**6**) in equal amounts. Similarly, the 3-isoprenylated indole building block **63** could generate yuehchukene (**62**) in the presence of acid.[8] However, indole **63** has not been isolated from natural sources. It is noteworthy to mention that yuehchukene was also isolated as a racemic mixture. Consistent with these previous reports, we successfully synthesized the

flinderoles A–C from borrerine, which generates 2-isoprenyl methyltryptamine **9** in the presence of acid.[9]

Scheme 3.1 2-isoprenylated methyltryptamine as the precursor for *Flindersia* alkaloids



Scheme 3.2 Spermacoceine and yuehchukene biosynthesis



3.2 Discrepancy in the specific rotation data

Based on our biomimetic synthesis of the flinderoles, we hypothesized that the formation of flinderoles in plants may take place without any enzymatic action. Hence, we postulated that a racemic mixture of the flinderoles occurs in plants. However, in contrast to our proposal, the isolated flinderoles A-C were reported as optically active. Interestingly, the borreverines, the structural isomers of the flinderoles, were isolated as racemates from both *Borreria* and *Flindersia* plants. If the flinderoles are produced in nature through an enzyme-free acid dimerization as we demonstrated in vitro, then no optical activity would be expected. Hence, a contradiction arose from specific rotation data that does not correlate with our hypothesis and would indicate that the flinderoles arise from different mechanisms than borreverines and yuehchukene. Avery and coworkers reported the specific rotation values for flinderoles A–C of $[\alpha]_D^{18}$= −6.5 (c

0.03, MeOH), $[\alpha]_D^{22}$ −7.4 (c 0.03, MeOH) and $[\alpha]_D^{22}$ −7.3 (c 0.03, MeOH), respectively. The concentrations they used, which were a consequence of the small quantities of samples isolated from the tree bark, were very low for optical rotation determination. Considering the sensitivity and inaccuracy of polarimeters at such low concentrations, these low absolute rotation values needed to be either corrected or confirmed. To resolve this discrepancy, the best method would be to access natural flinderoles and to repeat these rotation experiments. Unfortunately, we could not get access to the natural material. Alternatively, we could separate the enantiomers of the synthetic flinderoles and repeat the rotation experiments. We envisioned that these experiments may give insight into the biosynthesis of flinderoles. To achieve this goal, we collaborated with the group of Prof. Daniel Armstrong, a pioneer in chiral separations at the University of Texas, Arlington. After extensive experimentation, Mr. Jonathan Smuts from Prof. Armstrong's group found that a macrocyclic glycopeptide (vancomycin)-based stationary phase (Chirobiotic V) was suitable to separate the flinderole B enantiomers. Using preparative-scale vancomycin columns, sufficient amounts of the pure enantiomers of flinderole B were obtained. ^{1}HNMR and mass spectrometry confirmed the identity and purity of the separated enantiomers.[10]

Initially, our goal was to repeat the optical rotation experiment at the same concentration reported by Avery and coworkers (c = 0.028 g/100 mL). However, the optical rotation could not be determined effectively as it was close to the error of the instrument. As we suspected, the concentration was too dilute to influence the polarized light. The use of higher concentrations (0.325 g/100 mL and 1.400 g/100 mL) of pure

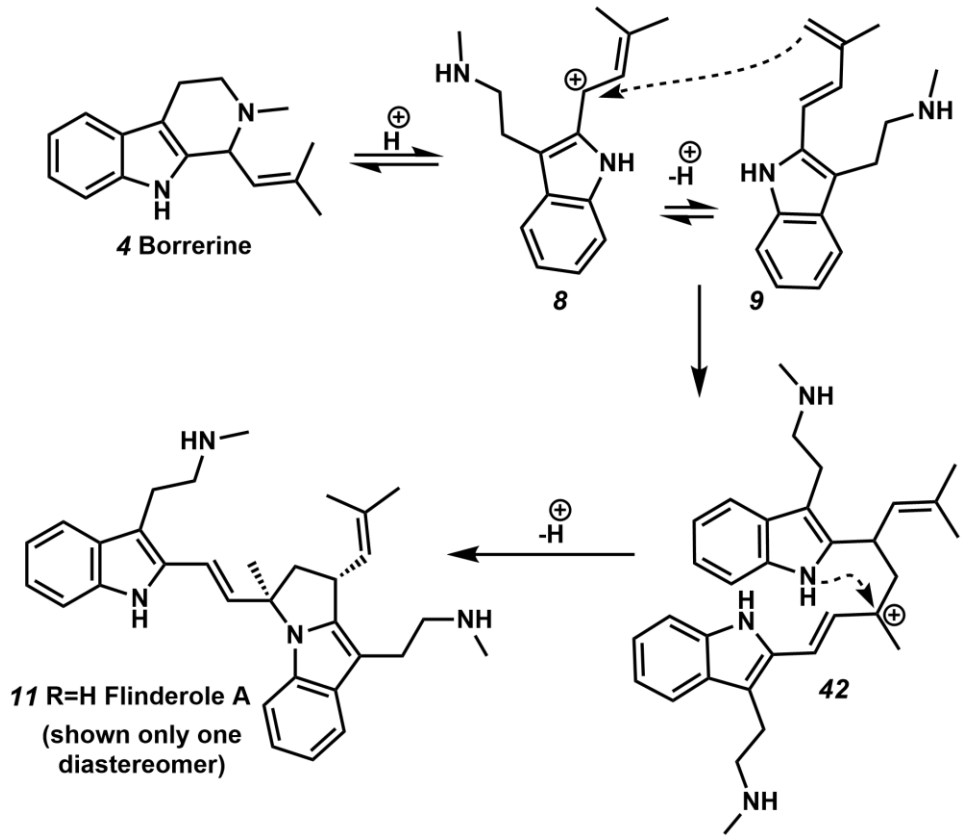
enantiomers gave consistent optical rotation values for a series of 10 measurements. The faster eluting enantiomer had a specific rotation of −48.8, and the slower eluting enantiomer had a specific rotation of +48.6 (Table 3.1).

Table 3.1 Specific rotation data for (–)-and (+)-flinderole B

entry	enantiomer	concentration	α	error	$[\alpha]^{D}_{24}$
1	Natural Flinderole B	0.03 g/100 mL	Not Reported	N/A	–7.4°
2	First Eluted	0.028 g/100 mL	–0.006°	0.008	ND
3	First Eluted	1.400 g/100 mL	–0.342°	0.005	–48.9
4	Second Eluted	0.325 g/100 mL	0.079°	0.002	48.6

The specific rotation experiments shown here suggest that the flinderoles in plants may exist as a racemic mixture rather than enantiopure. Based on our results, we decided to propose a biosynthesis of flinderoles. As demonstrated by the biomimetic synthesis, the dimerization of borrerine might occur with the help of only acid in the plants, excluding any enzyme role to induce chirality. Hence, we clarified data that potentially conflicted with our hypothesis of flinderole biosynthesis.[11] As hypothesized in Chapter 2, and as shown here in Scheme 3.1, in the presence of acid the tertiary amine of borrerine is protonated to open the carboline ring to a stable benzylic carbocation **8**. Deprotonation of the isobutenyl methyl can then lead to indole diene **9**. The terminal double bond of the diene **9** could attack the benzylic carbocation to make a carbon-carbon single bond as shown in structure **42**, followed by the attack of indole nitrogen to give flinderole A (**11**) or desmethyl flinderole C (**57**).

Scheme 3.1 Proposed biosynthesis of flinderoles



3.3 Conclusions

With the current specific rotation data for the synthetic flinderole B, it appears that the original literature data for the natural flinderole B is not sufficiently reliable to disqualify enzyme-free biosynthesis of the flinderoles. This study does not prove that the natural flinderoles are produced as racemates. The ambiguity will remain until we can access the natural flinderoles and analyze them with the HPLC assay. We predict that the naturally occurring samples are likely to be racemic. This prediction is consistent with the lack of optical activity for the borreverines and yuehchukene and with our proposed biosynthesis.

3.4 Supporting information

3.4.1 Materials:

Methanol was HPLC grade (VWR, Sugarland, TX). acetic acid (>99.7%), and triethylamine (>99%) were purchased from Sigma-Aldrich (Bellefonte, PN, USA). Flinderole B samples were synthesized as shown in chapter II.

3.4.2 Instrumentation:

Optical rotations were measured on a Digipol 781 Automatic Polarimeter (Rudolph Instruments, Fairfield, NJ) at 589nm and calibrated with a double quartz control plate rotation standard (Rudolph Instruments). The sample cell was 50mm long and had a volume of 800µL.

Preparative HPLC was performed on a Jasco 2000 series HPLC using a Chirobiotic V (Astec; 250mm x 21mm x 5µm) column. The pump (PU-2086) was set at 20mL/min with the mobile phase consisting of methanol with 0.5% acetic acid and 0.5% triethylamine. Detection was monitored at 280 nm with a high pressure UV/vis VWD (UV-2075). The enantiomeric fractions were collected using the Jasco SCF-Vch-Bp 6-valve change unit in the manual mode. Sample injection was performed using an autosampler (AS-2059-SFC) with a 1mL injection loop in the partial fill loop mode (injection volume=500µL).

Analytical HPLC separations were performed on an Agilent 1200 series HPLC (Agilent Technologies, Palo Alto, CA, USA) equipped with a diode array detector and a temperature controlled column chamber, auto sampler, and quaternary pump. All separations were carried out on a Chirobiotic V (Astec; 250mm x 4.6mm x 5µm) at room temperature (~20^{o} C) unless stated otherwise. For all HPLC experiments, the injection

volume was 5 µL and flow rate was 1.0 ml/min in isocratic mode. The following UV wavelengths were monitored for detection: 230, 254 and 280nm.

Mass spectra were acquired on a Thermo Finnigan LXQ (Thermo Electron Corp., San Jose, CA) operating in the positive mode by direct infusion.

3.4.3 Results:

	α (optical rotation)				
	E1 set 1	**E1 set 2**	**E2 set 1**	**E2 set 2**	**Quartz**
Value 1	-0.339	-0.344	0.078	0.077	1.805
Value 2	-0.339	-0.343	0.08	0.079	1.807
Value 3	-0.352	-0.345	0.081	0.075	1.806
Value 4	-0.333	-0.342	0.079	0.08	1.806
Value 5	-0.339	-0.342	0.081	0.079	1.807
Average, α	-0.340	-0.343	0.080	0.078	1.806
Std dev,α	0.007	0.001	0.001	0.002	0.001
Average $[\alpha]_D^{24}$	-48.6	-49.0	49.1	48.0	-
d (mm)	50				
c (E1) in g/100 mL	1.4				
c (E2) in g/100 mL	0.325				
Quartz lit. α	1.807				

Table 1: The optical rotation of each enantiomer was acquired as two sets of five replicate measurements. E1 refers to the first eluting enantiomer from the preparative HPLC and E2 the second eluting enantiomer.

The ESI-MS data for each enantiomer (E1 and E2): $[M+H]^+$ = 509.4, $[M+Na]^+$ = 531.4, $[M+2H]^{2+}$ = 255.2.

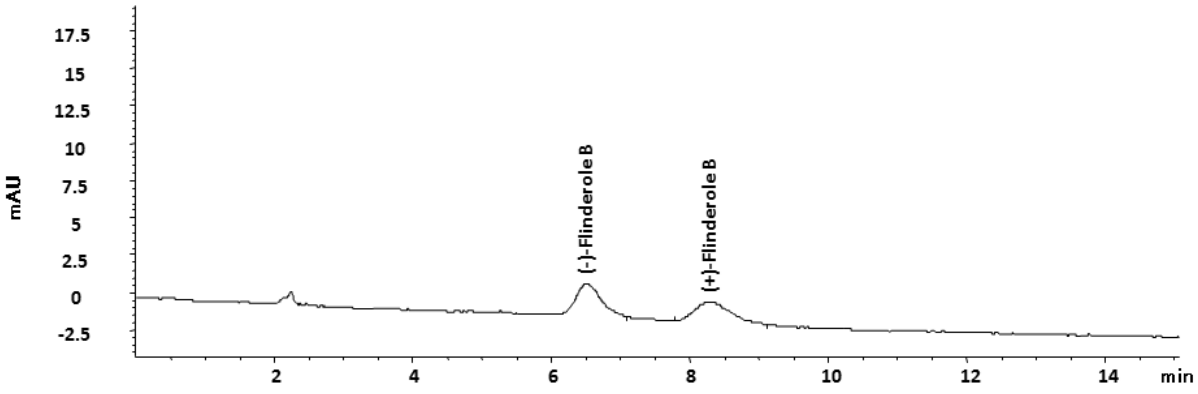


Figure 1: An analytical HPLC separation of flinderole B on a Chirobiotic V column. Mobile phase=methanol (0.5% acetic acid/0.5% triethylamine). Detection at 280nm.

3.5 References

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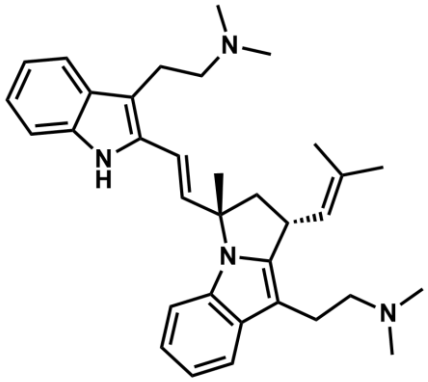
CHAPTER 4

PROGRESS TOWARD THE ENANTIOSELECTIVE TOTAL SYTNTHESIS OF FLINDEROLES A–C

4.1 Introduction

Soon after their isolation from *Flindersia* plants, the flinderoles A–C became an attractive target for total synthesis.[1,2] We were not only intrigued by the flinderoles' excellent antimalarial activity but also by their structural novelty. These bisindole alkaloids have a highly functionalized pyrrolidine ring which is fused to one of the indole rings to make a pyrrolo[a]indole framework as shown in Figure 4.1. In addition, two stereocenters on the pyrrolidine ring offer a great challenge to synthetic chemists. We saw this as an opportunity to develop synthetic methodologies to install these two stereocenters enantioselectively. The first stereocenter is an α-chiral center at the indole 2-position, created by the isobutylene substitution. The second stereocenter is tetrasubstituted and is located adjacent to the indole nitrogen. In this chapter the following will be discussed: first and foremost is the retrosynthesis of flinderoles A, B and C. Second, the forward synthesis, which mainly includes the detailed description for the installation of the first stereocenter through an enantioselective BINOL-catalyzed conjugate addition of boronic acids, a synthetic methodology previously developed in our lab.[3] Third, the progress towards the diastereoselective installation of the second stereocenter.

Figure 4.1 Flinderole C structure

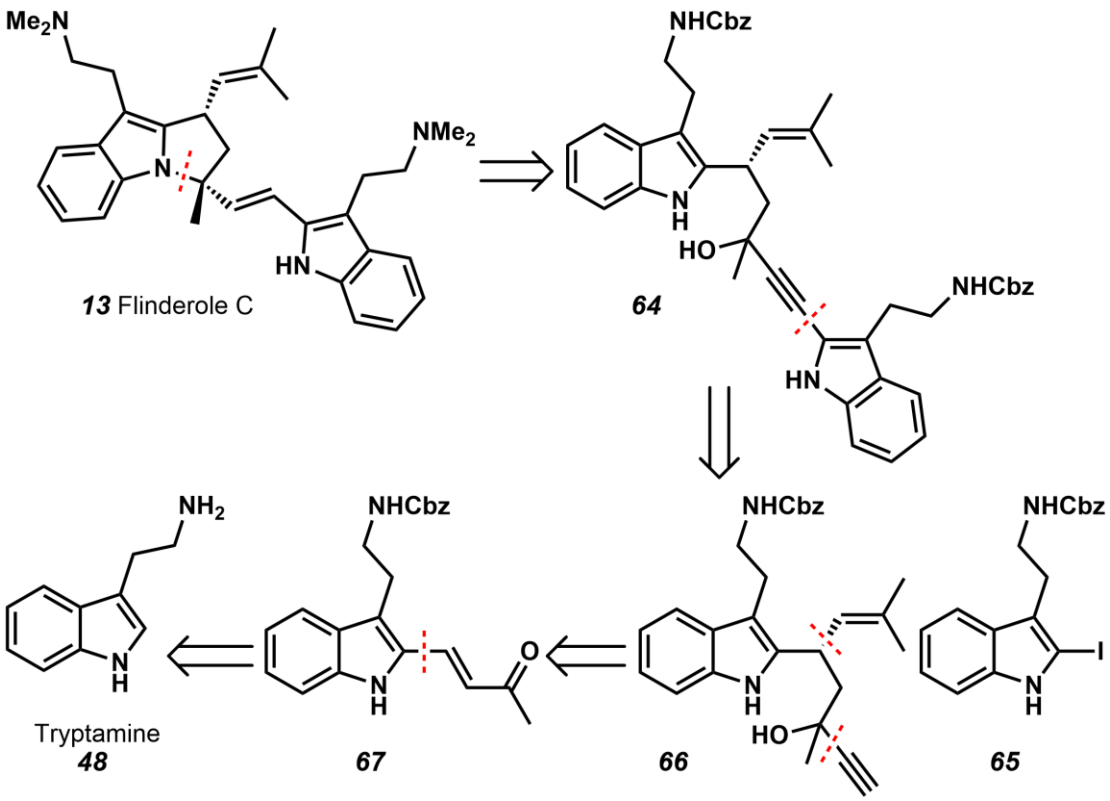


13 Flinderole C

4.2 Retrosynthesis of flinderoles B and C

The first disconnection between the indole nitrogen and tetrasubstituted carbon on the pyrrolidine ring was inspired by our biosynthetic proposal of flinderoles.[4] We envisioned that the advanced intermediate **64** would generate a stable cation in the presence of acid (**42**, Scheme 3.1, Chapter 3) which could be attacked by the indole nitrogen in S_N^1 fashion. The next disconnection is between the indole 2-position and the triple bond of propargylic alcohol **64**. This can be achieved in a convergent manner through a Sonagashira coupling between 2-iodo tryptamine **65** and propargylic alcohol **66**. The third disconnection is the installation of the isobutylene side chain in **66** that could be achieved through an enantioselective boronic acid conjugate addition on enone **67**. The enone **67** could be generated from tryptamine (**48**) by 2-position functionalization of the indole ring.

Scheme 4.1 Retrosynthetic approach for the total synthesis of flinderole C

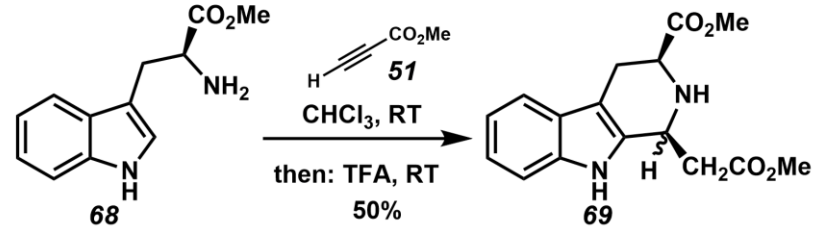


4.3 Forward synthesis

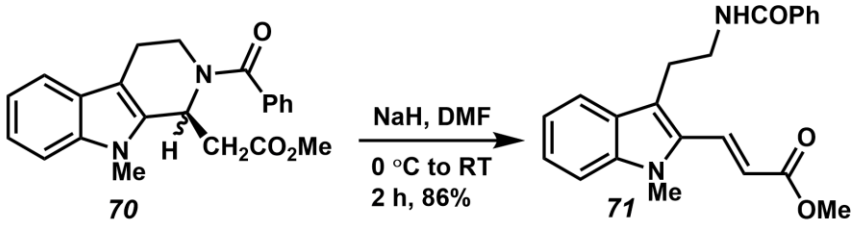
4.3.1 Pictet-Spengler reaction

The Bailey group developed a modified Pictet-Spengler reaction using (*S*)-tryptophan (**68**) and 3-butyn-2-one (**51**) as the Michael acceptor to produce 1,3-disubstituted tetrahydro-β-carboline **69** (Scheme 4.2).[5] They also demonstrated that, in the presence of a base, these carbolines with electron withdrawing groups on the carboline nitrogen and methyl group on indole nitrogen can open into 2-enone tryptophan derivatives (Scheme 4.3).[6] We wanted to test this sequence of reactions without methylating the indole nitrogen.

Scheme 4.2 Modified Pictet-Spengler reaction to prepare β-carboline **69** by Bailey[5]

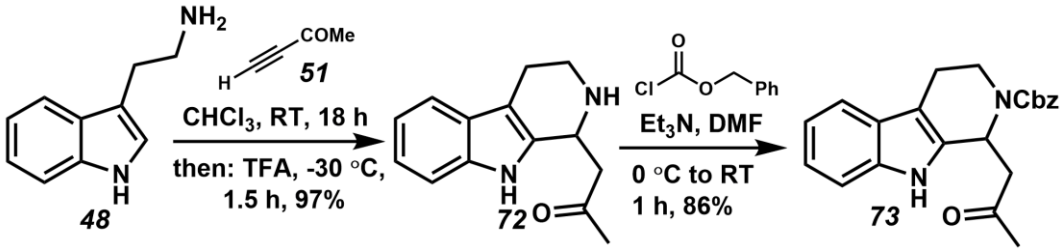


Scheme 4.3 Base induced ring opening of β-carboline **71** Bailey[6]

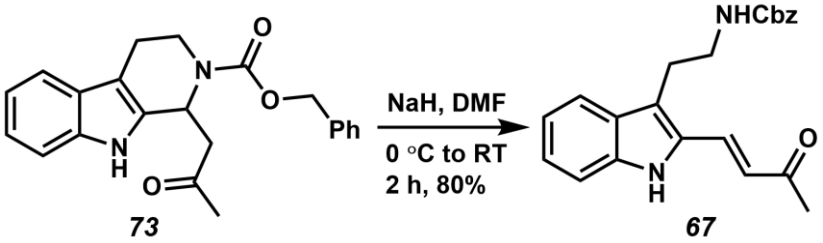


In a similar manner, we wanted to synthesize enone **67** starting from readily available tryptamine (**48**) (Scheme 4.4). The modified Pictet-Spengler reaction of tryptamine (**48**) and 3-butyn-2-one (**51**) gave the β-carboline **72** (similar reaction was used in the scheme 2.4, chapter 2). Immediately after work up and without purification it was treated with benzyl chloroformate in the presence of triethyl amine. There are two reasons to use the Cbz group. First, it acts as an electron withdrawing group, which would facilitate the base induced ring opening of β-carboline to the desired enone **67** (Scheme 4.5). The second reason is that the Cbz group is a masked methyl group. Upon reduction, the carbamate can be transformed to the secondary amine of the flinderoles at the end of the total synthesis.

Scheme 4.4 Synthesis of β-carboline **73**



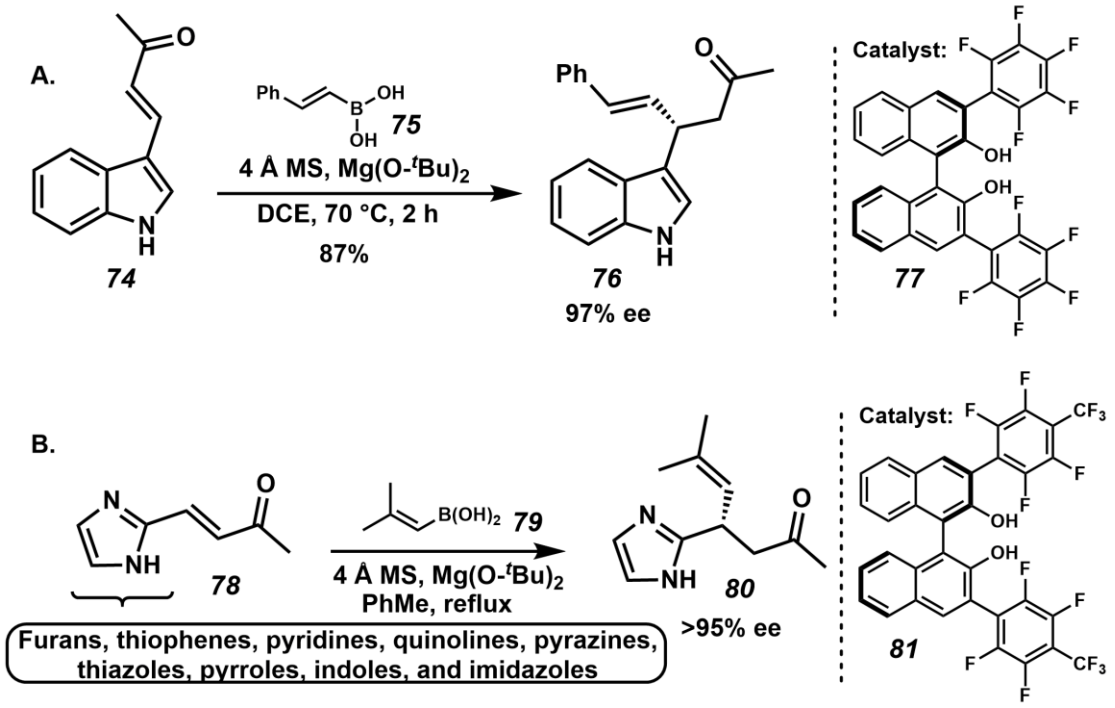
Scheme 4.5 Synthesis of enone 67



4.3.2 May lab enantioselective conjugate addition methodologies on heterocycles

Previously in our lab, Dr. Brian Lundy and Dr. Santa Jansone-Popova developed a BINOL-catalyzed enantioselective boronic acid conjugate addition on 2-enone appended unprotected indoles to produce α-chiral indoles with high yield and excellent enantioselectivity.[3a] For this transformation, BINOL catalyst **77** with pentafluoro phenyl groups at the 3 and 3' positions was used (Scheme 4.6). The chiral center produced is very stable and there was no epimerization observed. Later on, Phong Le and Thien Nguyen expanded the scope of this novel methodology to various heterocyclic enones.[3b] However, reaction was sluggish with the BINOL catalyst **77**. To increase the reactivity of the substrates, a modified BINOL catalyst **81** was developed to achieve excellent enatioselectivities and high yields.

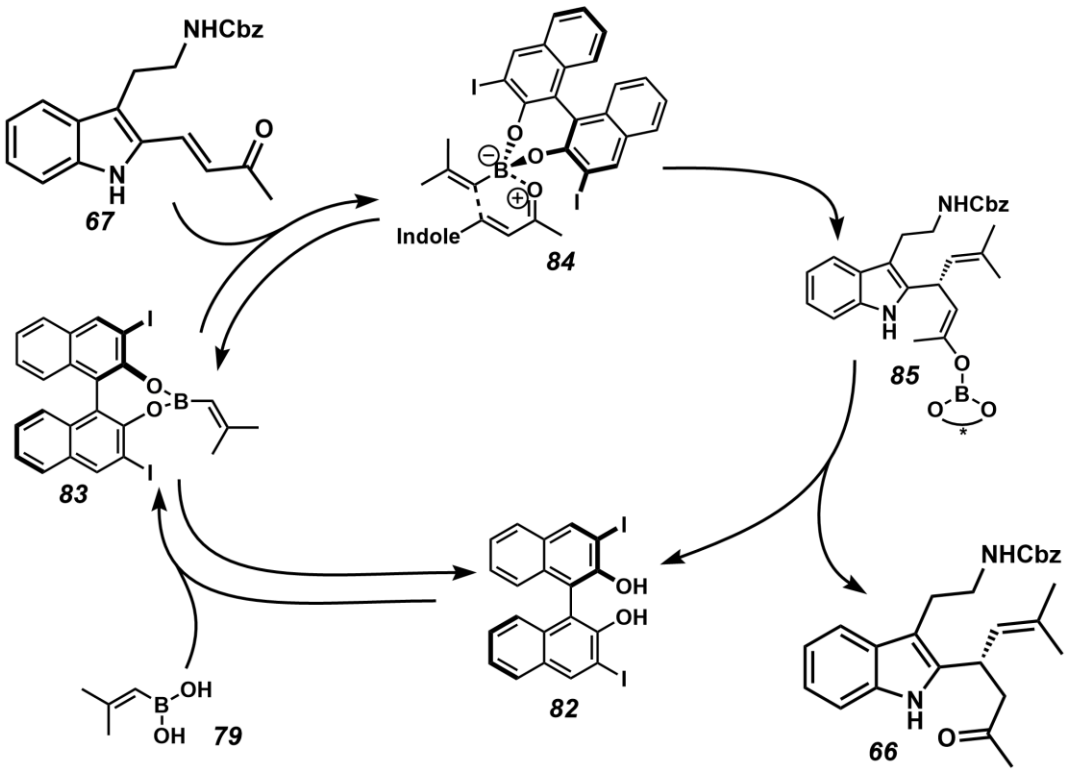
Scheme 4.6 May lab enantioselective conjugate addition methodologies[3a,3b]



4.3.3 Proposed mechanism for the BINOL-catalyzed conjugate additions

Based on a proposal by Chong and coworkers, the mechanism of the BINOL-catalyzed conjugate addition reaction begins with the formation of boronate ester **83** from BINOL **82** and boronic acid **79** (Scheme 4.7). The boron of the ester has to be Lewis acidic to coordinate with the indole enone to make the boron "ate" complex **84**. Presumably, the electron withdrawing substituents at the 3 and 3' positions of BINOL serve this purpose through an inductive effect. In addition, these electron withdrawing substituents also stabilize the negative charge on the boron in the boron "ate" complex **84**. Conjugate addition can then occur to make the boron-enolate **85**. Hydrolysis of the boron enolate would then regenerate the catalyst, and give the product **66**.[7]

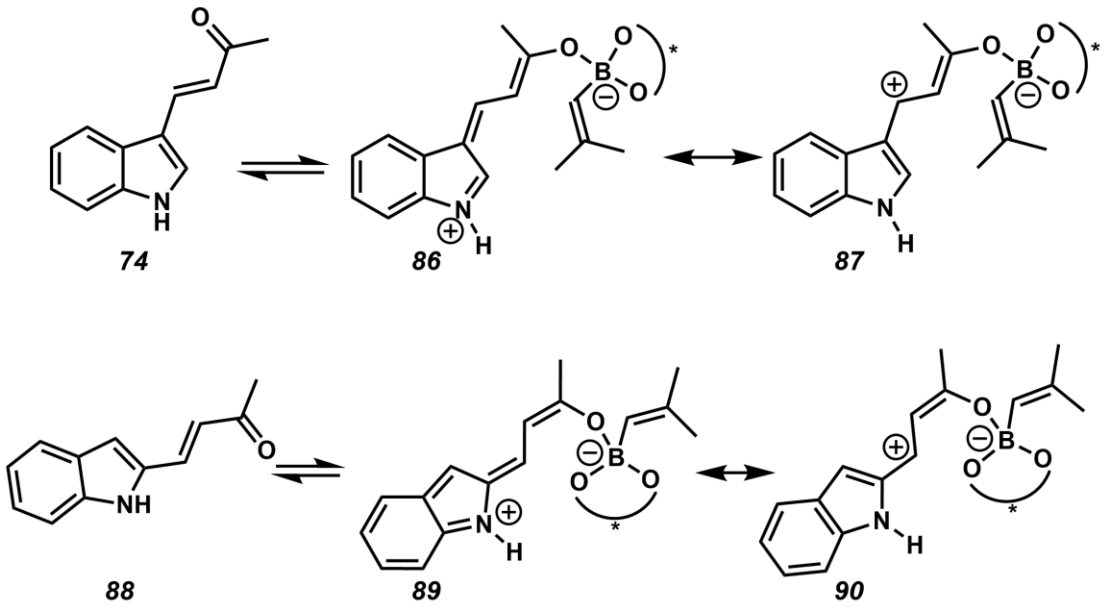
Scheme 4.7 Proposed organocatalysis mechanism



4.3.4 Reactivity difference between 3-enone appended indoles and 2-enone appended indoles

There is a difference in reactivity between a 3-enone indole and a 2-enone indole that results from electronic effects in the formation of the boron "ate" complex. If we analyze the boron "ate" complexes **86** and **89** (Scheme 4.8), we can see the formation of **89** has to disrupt the aromaticity in the benzene ring of the indole, which makes it a highly energetic species, whereas in the formation of **86** that aromaticity is not disturbed. Hence, there is a challenge to overcome the indole electronics to achieve boronic acid conjugate addition on 2-enone indole substrates.

Scheme 4.8 Reactivity difference between 3-enone indole and 2-enone indole

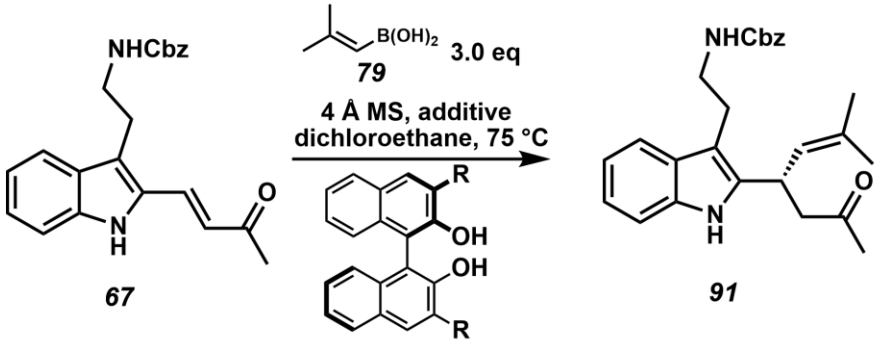


4.3.5 Optimization of the conjugate addition reaction

Based on the aforementioned diminished reactivity, we anticipated that stoichiometric BINOL may be necessary to promote the conjugate addition on 2-enone appended tryptamine **67**. We started testing the reaction using 1 equivalent of BINOL, which gave only trace amounts of the conjugate addition product **91** (entry 1, Table 4.1). Upon using various BINOL catalysts with electron withdrawing groups at the 3 and 3' positions, the enantioselectivity was excellent, but the yields were not satisfactory (entries 2-4). Dr. Brian Lundy reported a 60% yield of the product **91** based on the recovered starting material with 20 mol% di-I_2-BINOL as the catalyst and 3 equivalents of boronic acid (entry 5). Modifying those conditions by using 1 equivalent of di-I_2-binol and 1 equivalent of $Mg(O\text{-}^tBu)_2$ gave 60% yield with high enantioselectivity (entry 6). However, a catalytic amount of di-I_2-BINOL with $Mg(O\text{-}^tBu)_2$ gave only 35% yield

(entry 7). Consumption of starting material was very fast with $Mg(O\text{-}^tBu)_2$, and before all the starting material was consumed rapid decomposition of the product was observed. This prompted us to use a less basic additive like Cs_2CO_3 instead of the $Mg(O\text{-}^tBu)_2$, which improved the yield of **91** to our satisfaction and the enantioselectivity was also excellent (entry 8).

Table 4.1 Developing an enantioselective conjugate addition of boronic acids



entry	R		additive		time	isolated yield (SM)	er
1	H	(1.0 eq)	$Mg(O^tBu)_2$	(1.0 eq)	26 hours	<2% (85%)	--
2	CF_3	(1.0 eq)	$Mg(O^tBu)_2$	(1.0 eq)	3 hours	35% (55%)	--
3	$C_6(p\text{-}CF_3)F_4$	(1.0 eq)	$Mg(O^tBu)_2$	(1.0 eq)	3 hours	35% (50%)	99:1
4	C_6F_5	(1.0 eq)	$Mg(O^tBu)_2$	(1.0 eq)	3 hours	45% (40%)	99:1
5[a]	I	(0.2 eq)	$Mg(O^tBu)_2$	(0.2 eq)	48 hours	60%[b] (30%)	97:3
6	I	(1.0 eq)	$Mg(O^tBu)_2$	(1.0 eq)	2 hours	60% (20%)	98:2
7	I	(0.2 eq)	$Mg(O^tBu)_2$	(0.2 eq)	2hours	35% (30%)	--
8	I	(0.2 eq)	Cs_2CO_3	(1.0) eq	24 hours	67%(15%)	98:1

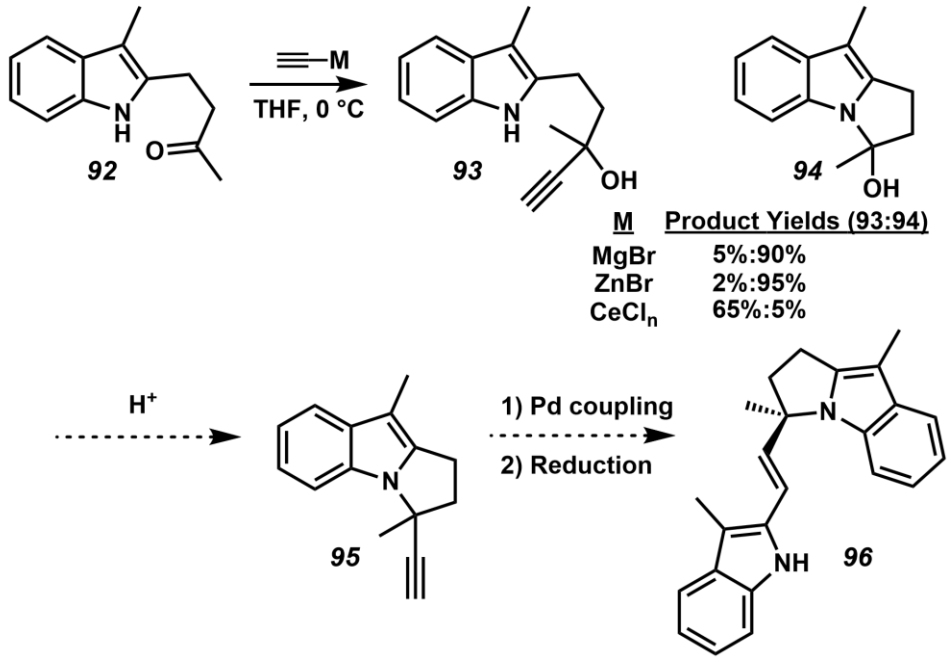
[a]Dr. Brian Lundy result[15] [b]yield based on recovered starting material

4.3.6 Propargyl alcohol formation

For the forward synthesis, we chose model substrate **92** to test the 1,2-addition to the ketone with an alkyne nucleophile to form **93** (Scheme 4.9). The unprotected indole

could be deprotonated in basic conditions and form thermodynamically stable hemiaminal **94**. With a Grignard reagent and organo zincate only hemiaminal **94** was formed. However, the organo cerate gave the desired propargylic alcohol **93** as a majority of the crude product along with a trace amount of the undesired hemiaminal **94**. Acid mediated cyclization of **93** did not give any product. If it had been successful, we would have continued with a Sonagashira coupling and reduction of the triple bond to yield the flinderole scaffold **96**.

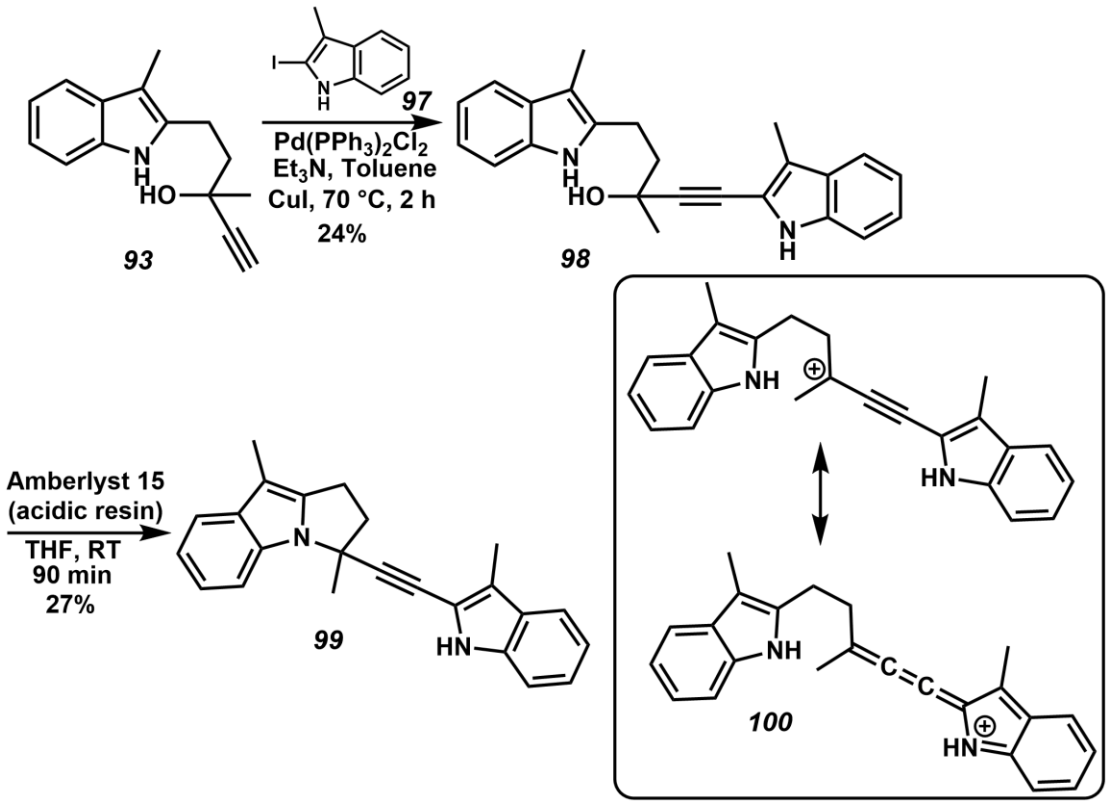
Scheme 4.9 Synthetic attempts on the model substrate



However, this problem was solved by changing the reaction sequence. Sonagashira coupling of the propargylic alcohol **93** and 2-iodoindole **97** gave the advanced intermediate **98**. In the presence of a mild acid like amberlyst 15A resin, **98** could produce stabilized cation shown as resonance forms of **100**, so that the indole lone

of indole nitrogen can attack it to make pyrroloindole **99**. Indeed, a preliminary test of the reaction under acidic conditions gave the flinderole framework in low yield.

Scheme 4.10 Formation of flinderole skeleton

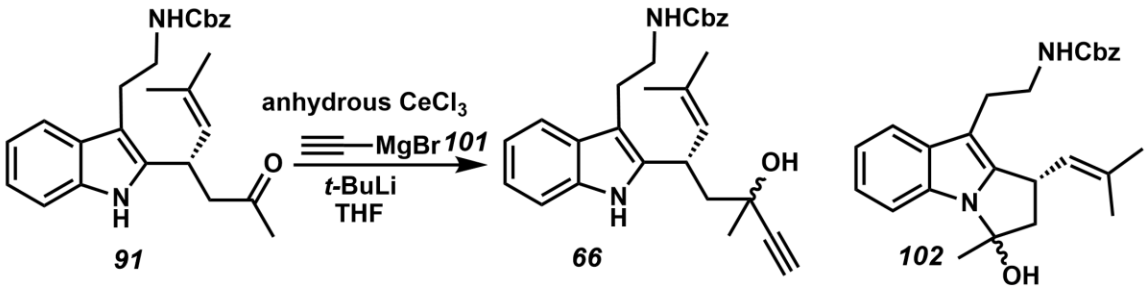


4.3.7 Synthesis of propargylic alcohol 66

Implementation of the exact reaction conditions developed on the model substrate to synthesize propargylic alcohol **93** did not work on the real substrate **91**. Various reaction conditions were tested by changing the equivalents of anhydrous cerium chloride. In most cases only the undesired hemiaminal **102** was formed as the major product. The carbamate on the sidechain might be affecting the cerate formed in the reaction mixture. However, after making the cerate at -78 °C, then stirring at 0 °C for 20-

30 minutes followed by the addition of starting material at −78 °C gave the product **66** as two diastereomers in almost a 1:1 ratio in 10-15% isolated yield. However, these highly moisture sensitive cerate reaction conditions suffered with low isolated yield of the product and poor reproducibility of the reaction.

Table 4.2 Propargylic Alcohol **66**



entry	$CeCl_3$	Ethynyl MgBr	Procedure[a]	66: 102: 91[b,c]
1	0	2.5	-78 °C	0: 90: 10
2	2.0	2.0	-78 °C	0: 80: 10
3	3.0	2.5	-78 °C	0: 70: 20
4	3.0	2.5	-78 °C 15 min then 0 °C 20-30 min	50: 20: 20[d]
5	3.0	2.5	Premixing SM with $CeCl_3$ -78 °C	0: 80: 10
6	3.0	2.5	No *t*-BuLi -78 °C	0: 0: 90

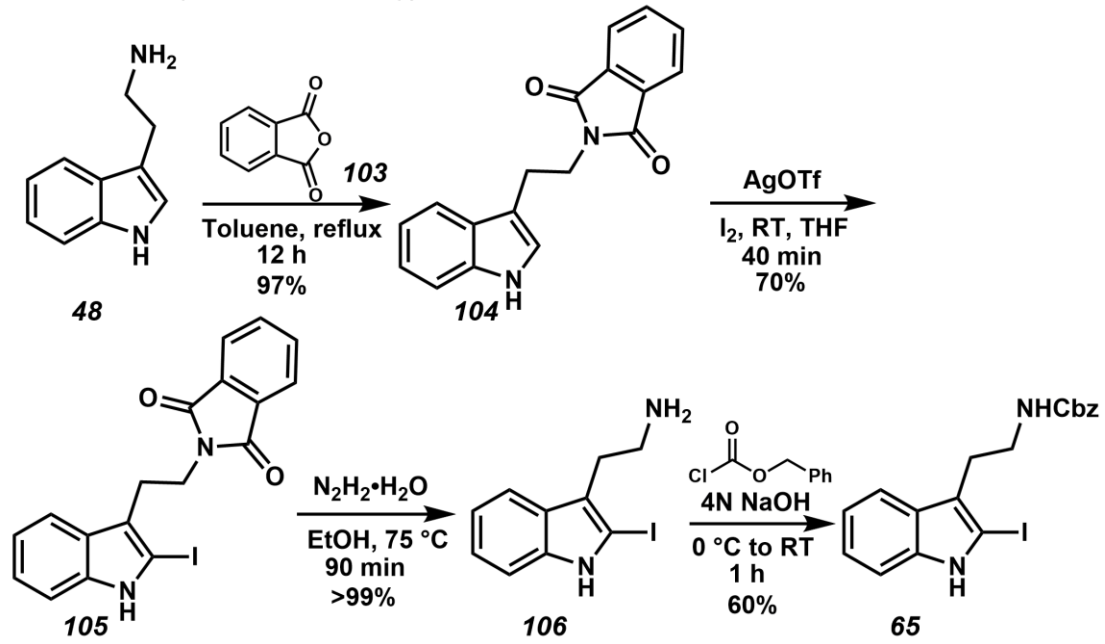
[a]Reaction was quenched with aq. sat. NH_4Cl
[b]90% of the crude material was collected
[c] Determined by ^1H NMR integration
[d]10-15% of product was isolated

4.3.8 Synthesis of 2-iodo tryptamine 65

Tryptamine (**48**) was protected first with pthalimide and then iodinated by a silver triflate mediated iodination to produce 2-iodo-N-pthaloyl-tryptamine (**105**).[8] Cleavage of

pthalimide gave 2-iodo tryptamine (**106**), which was immediately reacted with benzylchloroformate to yield Cbz protected 2-iodo tryptamine **65**.

Scheme 4.11 Synthesis of 2-Iodotryptamine **65**

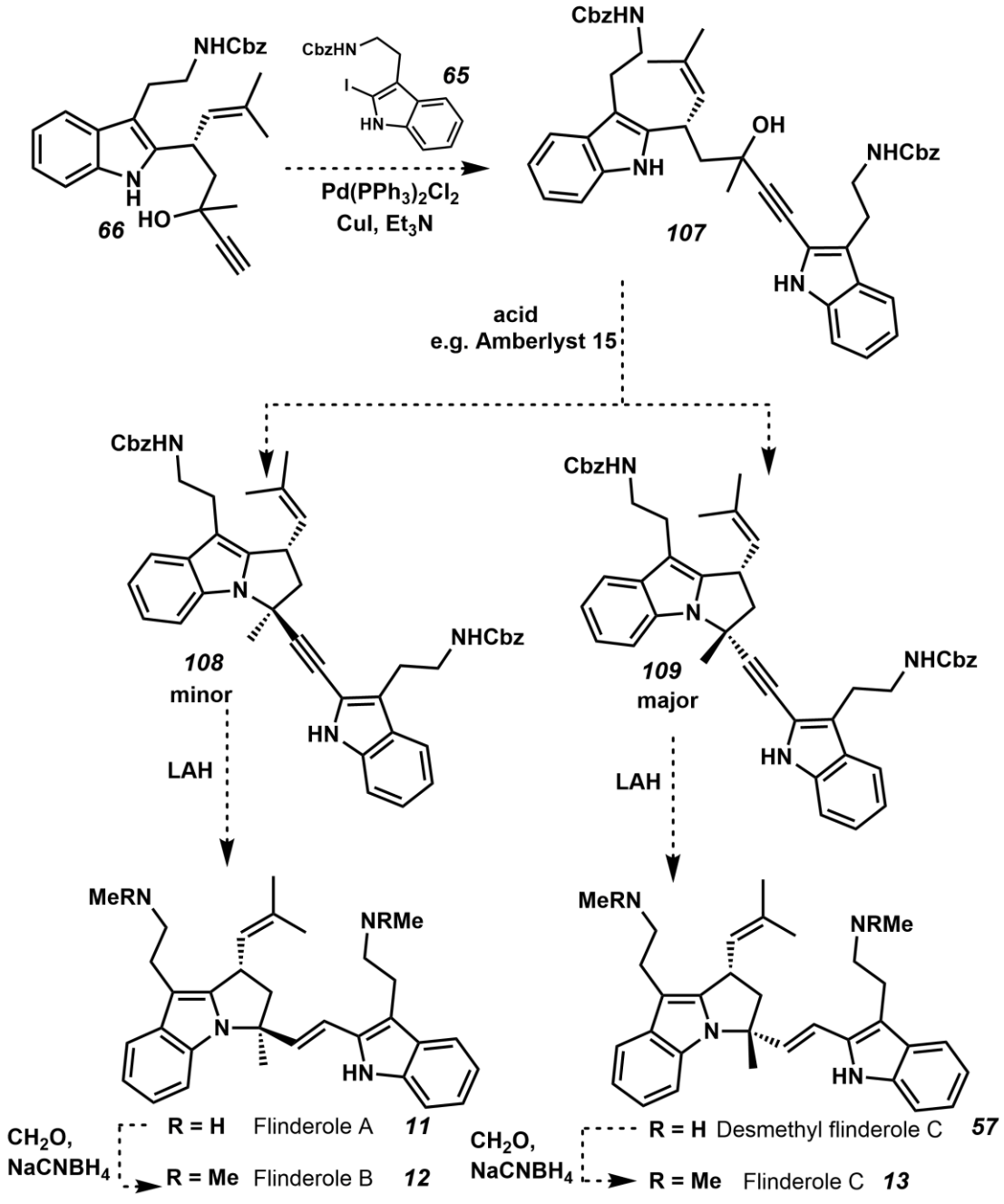


4.3.9 Proposed completion of the enantioselective synthesis of flinderoles A–C

The Sonagashira coupling of propargylic alcohol **66** and 2-iodo tryptamine **65** would give **107** (Scheme 4.12).[9] From this advanced intermediate **107** we envisioned that the diastereoselectivity of the synthesis of flinderole A (**11**) and desmethyl flinderole C (**57**) could be controlled by the acid mediated amino cyclization. We assume that the amino cyclization in the presence of acid is a reversible process, and so a more stable product would be preferred. The steric sizes of methyl, akenyl (isobutylene group), and alkynyl (ethynyltryptamine) substituents could be estimated based on their A-values (conformational free energies reported in Kcal/mol).[10] Methyl and alkenyl substituents

have an equal A value of 1.7 whereas an alkynyl substituent has an A value of 0.5. Therefore, we predict that the product **109** with the alkenyl and methyl groups in a trans configuration would be preferred over the product **108** which has those groups in cis configuration. Reduction of the carbamate and triple bond could be achieved using strong reducing reagents like LAH. Aluminum could be coordinated to indole N-H and the alkyne to direct the hydride transfer to produce the trans olefin.[11]

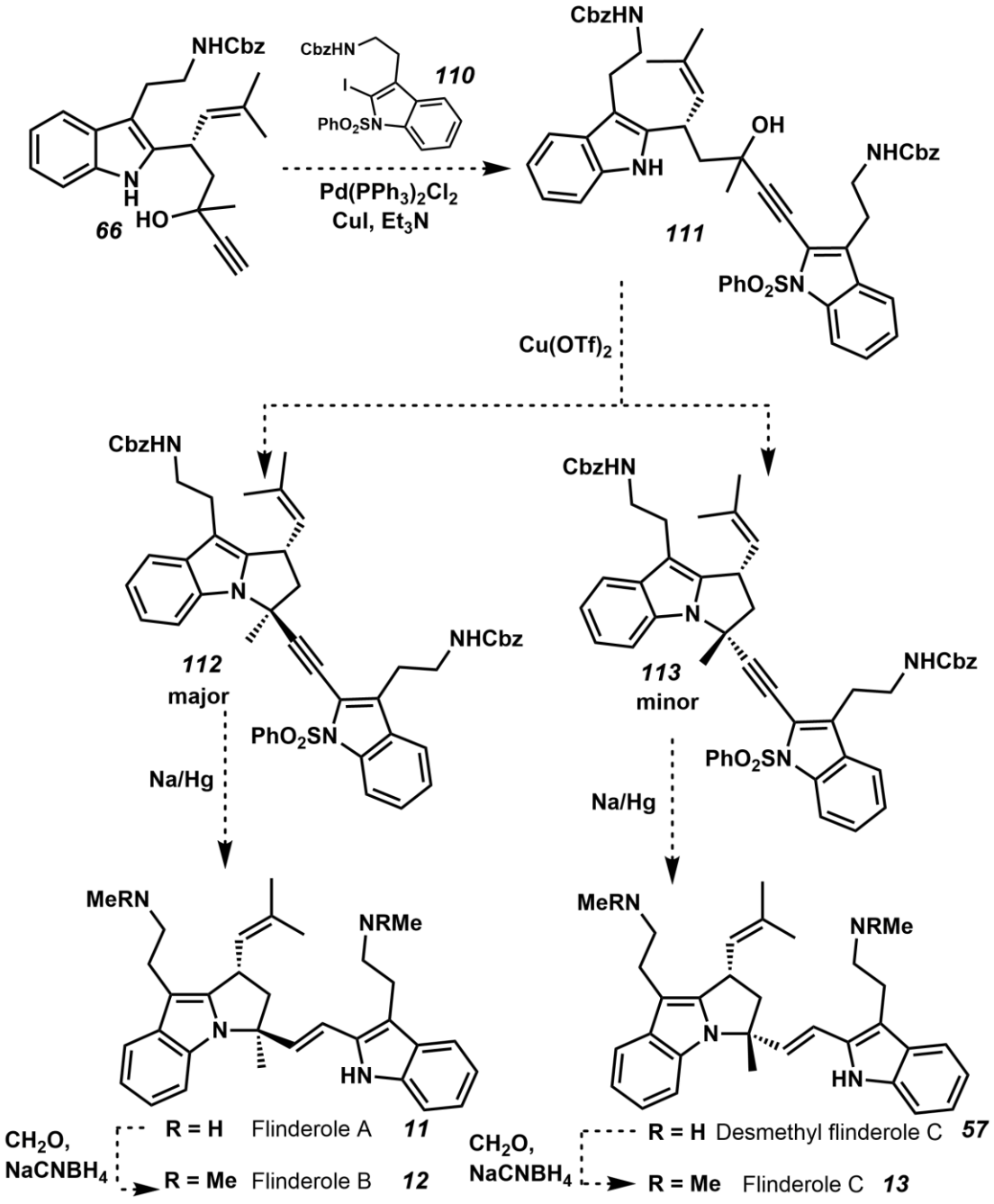
Scheme 4.12 Completing the enantioselective synthesis of the flinderoles



However, $Cu(OTf)_2$ might give an opposite selectivity (Scheme 4.13).[2a] According to Dethe, copper could coordinate to the benzene sulphonyl group of one indole ring and nitrogen of the other indole ring. The approach of the alkyne could be

endo to prefer the formation of **112** over **113**. Finally, reduction of carbamate, alkyne and benzene sulphonyl groups would give flinderole A (**11**) and desmethyl flinderole C (**57**), which upon reductive amination would complete the flinderole B (**12**) and flinderole C (**13**) synthesis.

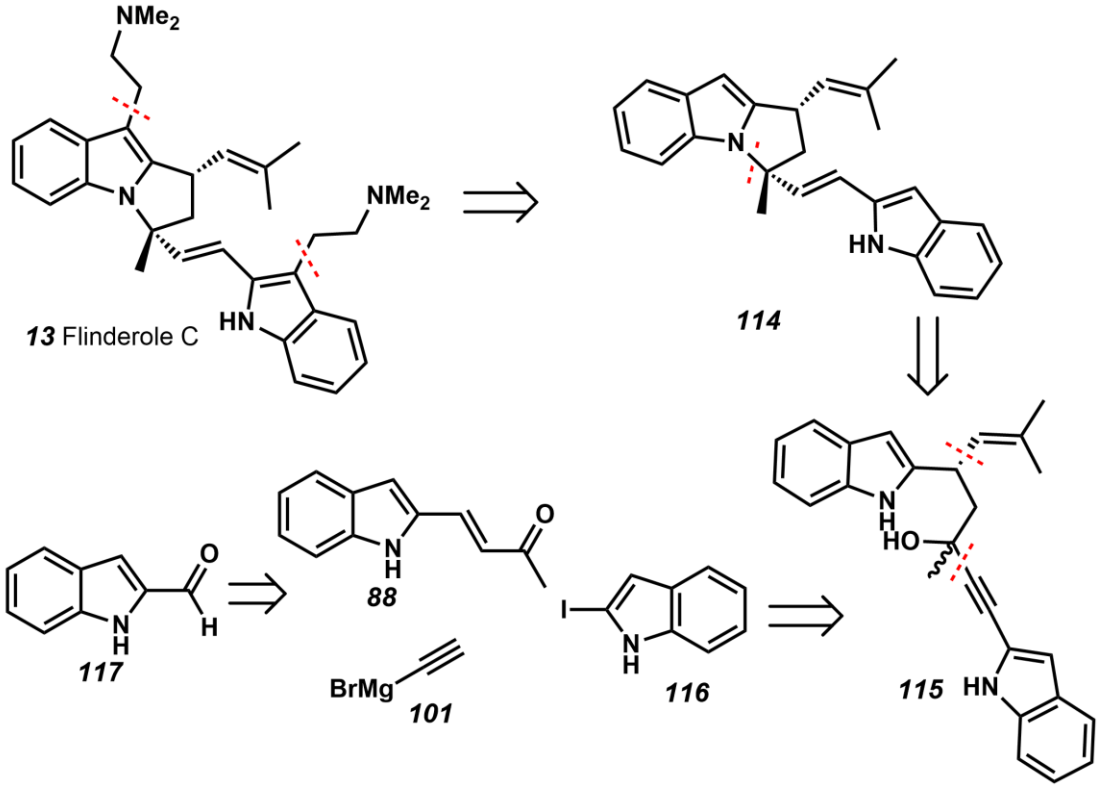
Scheme 4.13 Completing the enantioselective synthesis of the flinderoles II



4.4 Alternative retrosynthesis for enantioselective synthesis of flinderoles

We proposed an alternate retrosynthesis by performing a side chain disconnection first. The rest of the retrosynthesis remains the same as the earlier retrosynthesis. The enone **88** can be prepared from indole 2-carboxaldehyde (**117**).

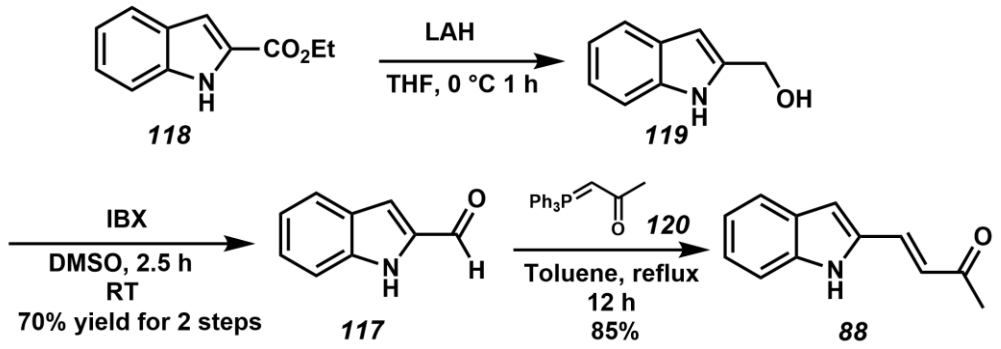
Scheme 4.14 Alternative retrosynthesis of flinderoles for the enantioselective synthesis



4.4.1 Synthesis of 2-enone indole 88

Reduction of the indole 2-ethylcarboxylate (**118**) with LAH gave the indole alcohol **119**, and then oxidation using IBX resulted in indole 2-carboxaldehyde (**117**) (Scheme 4.15). Wittig olefination using ylide **120** and the carboxaldehyde **117** gave the 2-enone indole **88** in excellent yield.

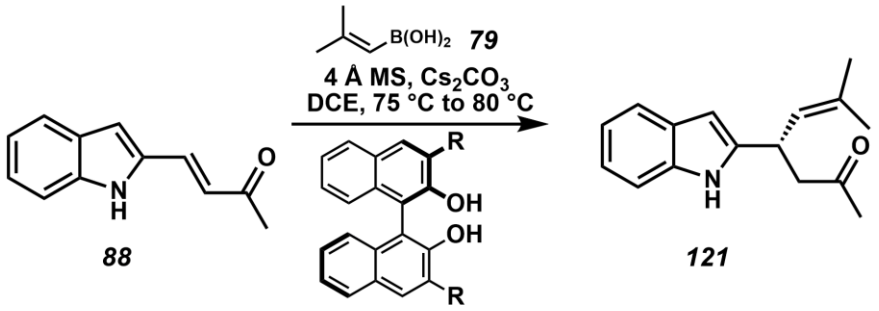
Scheme 4.15 Synthesis of 2-enone indole **88**



4.4.2 Enantioselective conjugate addition on 2-enone indole 88

Initially, BINOL with different halides substituted at the 3 and 3'-positions were tested for the conjugate addition of boronic acid **82** on the 2-enone indole. As the size of the halide increased, the yield and the enantioselectivity increased accordingly (entries 1-3, Table 4.3). A trifluoromethyl-substituted BINOL gave similar result as the di-I_2-BINOL (entry 5). However, the pentafluoro-substituted BINOL proved to be effective for this substrate, which gave excellent enantioselectivity and yield (entry 6).

Table 4.3 Conjugate addition on 2-enone indole

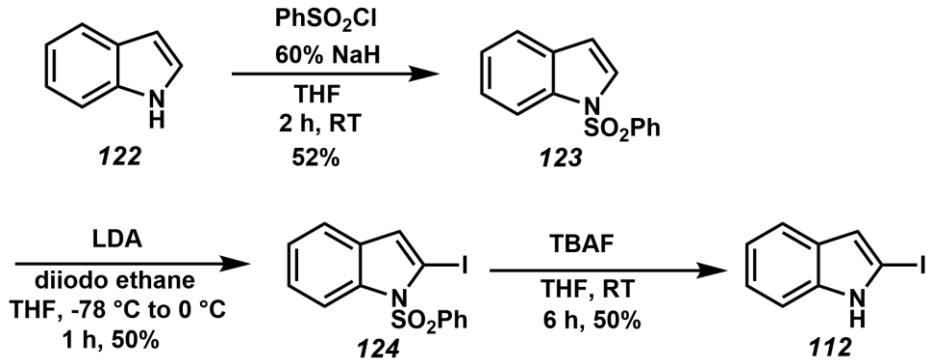


entry	R	time	yield (SM)	er
1	F	24hours	40% (50%)	53 : 47
2	Cl	24 hours	60% (35%)	69 : 31
3	I	20 hours	60% (10%)	82 : 18
4	I	17 hours	75% (10%)	ND
5	CF_3	24 hours	55% (10%)	81 : 19
6	C_6F_5	20 hours	75% (10%)	96 : 4
7	C_6F_5racemic	43 hours	48% (0%)	50 : 50

4.4.3 Synthesis of 2-iodoindole (112)

The synthesis of 2-iodo indole (**112**) was achieved following a literature procedure.[12] Indole (**122**) was first protected with a phenylsulfonyl group, followed by iodination using LDA and diiodo ethane. Finally, the phenyl sulphonyl group was removed using fluoride to yield 2-iodo indole (**112**).

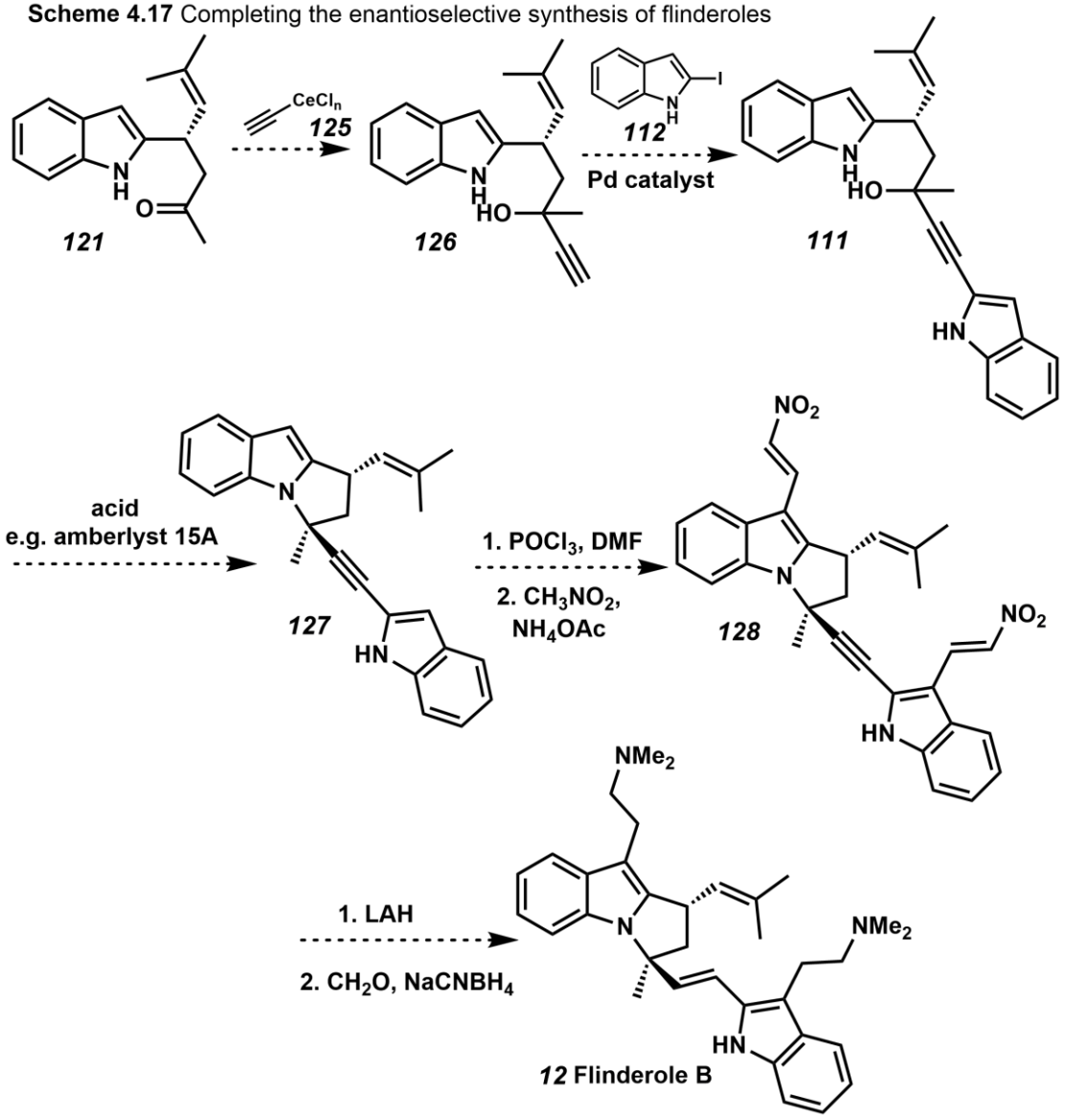
Scheme 4.16 Synthesis of 2-iodo indole (**112**)



4.4.4 Proposed completion of the enantioselective synthesis of flinderoles A–C

As the cerate addition to ketone **91** had been giving inconsistent results, we thought that excluding the tryptamine side chain would improve formation of propargylic alcohol **126**. Advanced intermediate **117** then could be synthesized in a convergent manner using a Sonagashira coupling reaction of 2-iodoindole (**112**) and the propargylic alcohol **126**. An acid promoted S_N1 type amino cyclization would make the pyrrolo[a] indole framework with diastereoselectivity. At this point, the side chain could be installed using Vilsmeir-Haack conditions, followed by nitroolefination. Reduction of the nitro olefin and triple bond could be achieved using strong reducing reagents like LAH.[12] Aluminum could be coordinated to indole N-H and the alkyne to direct the hydride transfer to produce the trans olefin. Finally mono-methylation would give the secondary amine congeners of flinderoles and di-methylation would give the tertiary amine congeners of flinderoles.

Scheme 4.17 Completing the enantioselective synthesis of flinderoles



4.5 Conclusions

The progress towards a concise, protecting group-free enantioselective synthesis of flinderoles A, B, and C has been shown in this chapter. The acid-promoted amino cyclization strategy would make our total synthesis novel compared to previously known total syntheses of flinderoles. Enantioselectivity is introduced by a BINOL-catalyzed

boronic acid conjugate addition. The rest of the synthesis would be diastereoselective. This route would help us to prepare the flinderoles and flinderole analogue enantiopure, which can be used for biological studies and antimalarial drug development. Finally, functional group interconversion would complete the enantioselective synthesis of flinderoles A–C.

4.6 Experimental section

4.6.1 General considerations

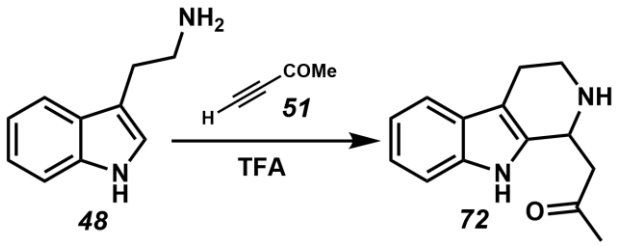
All the reactions were performed in flame- or oven-dried glassware. Benzene, THF, and DCM were purged with argon and dried over activated alumina columns. Column chromatography was performed on 60 Å silica gel (Sorbent Technologies) and commercially available neutral alumina. The ^{1}H and ^{13}C NMR spectra were recorded on a JEOL ECA-500 or ECX-400 spectrometer. Analytical thin layer chromatography was performed on EMD silica gel/TLC plates with fluorescent indicator 254 nm. HPLC was performed using a Gilson 321 pump with UV/VIS-155 detector and FC204 fraction collector.

4.6.2 Materials

Commercially available compounds were purchased from Aldrich and were used without further purification.

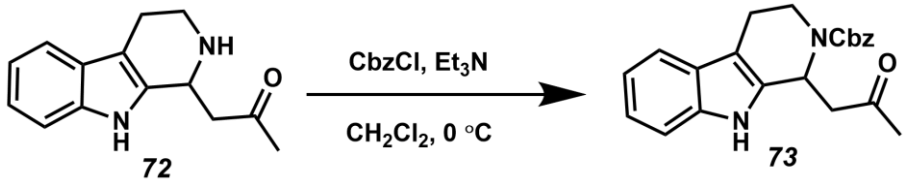
4.6.3 Experimental procedures

4.6.3.1 Synthesis of 1-(2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indol-yl)propan-2-one (72)



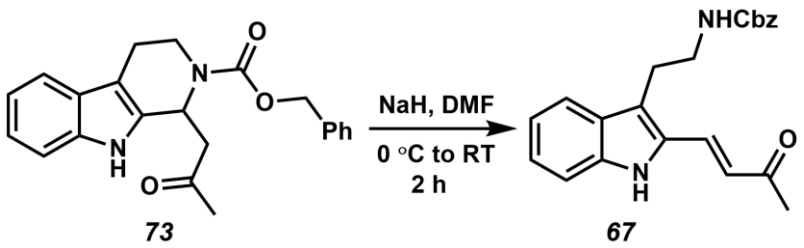
A stirred solution of tryptamine (2.00 g, 124 mmol) in 50 mL of DCM was treated with propargyl ketone **51** (1.17 mL, 149 mmol, 1.2 eq) and stirred for 18 h at room temperature. To the reaction mixture was added TFA (1.91 mL, 249 mmol, 2.0 eq) at -30 °C for 30 min and then it stirred at the same temperature for 1 more hour. It was then quenched by pouring into 100 mL of cold water. To this mixture was slowly added a concentrated solution of sodium hydroxide (10 N) until the pH was 10, followed by extraction with DCM. The organic layer was washed with water, dried over anhydrous sodium sulfate (Na_2SO_4), filtered, and concentrated under reduced pressure. The crude product was obtained (2.82 g, 99%) and used directly for the next step.

4.6.3.2 Synthesis of benzyl 1-(2-oxopropyl)-3,4-dihydro-1H-pyrido[3,4-b]indole-2(9H)-carboxylate (73)



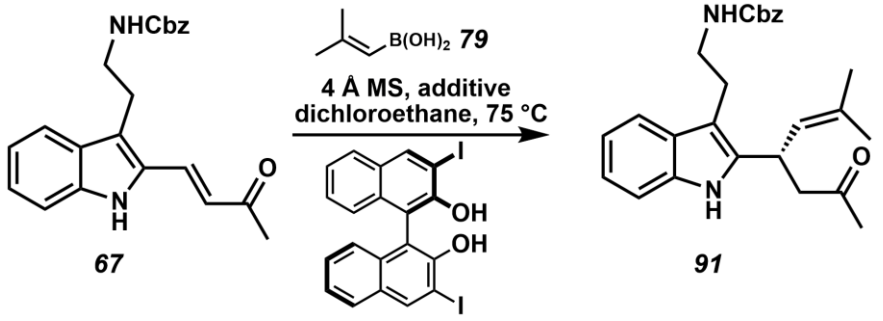
A well stirred solution of **72** (2.62 g, 11.5 mmol) and triethylamine (4.8 mL, 34.4 mmol, 3.0 eq) in DCM (50 mL) was cooled to 0 ^{o}C and slowly treated with benzyl chloroformate (2.46 mL, 17.2 mmol, 1.5 eq). The reaction mixture was stirred at the same temperature for about 30 min and then for 1 h at room temperature. The reaction mixture was quenched with water, extracted with DCM, and washed with saturated $NaHCO_3$ and then brine. The extract was dried with Na_2SO_4 and concentrated under reduced pressure. The residual crude product was purified by basic alumina chromatography using 7:3 hexanes/ethyl acetate (3.57 g, 78%): ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.78 (s, 1H), 8.61 (s, 1H), 7.46 (t, *J* = 9.1 Hz, 1H), 7.38-7.32 (m, 12 H), 7.25 (t, *J* = 7.4 Hz, 2H), 7.09 (m, 2H), 5.69 (m, 1H), 5.64 (d, *J* = 10.3 Hz, 1H), 5.29 (d, *J* = 12.6 Hz, 1H), 5.18 (s, 2H), 5.14 (d, *J* = 12.6 Hz, 1H), 4.57 (dd, *J* = 8.0, 5.1 Hz, 1H), 4.46 (dd, *J* = 8.0, 5.1 Hz, 1H), 3.49 (s, 2H), 3.15-2.7 (m, 12H), 2.2 (s, 3H), 2.1 (s, 3H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): δ 209.6, 209.5, 155.5, 154.9, 136.6, 135.7, 135.6, 133.4, 132.9, 128.6, 128.2, 128.0, 122.1, 122.0, 119.5, 119.4, 118.2, 118.1, 111.2, 108.4, 107.9, 67.5, 49.5, 48.9, 47.0, 46.9, 39.6, 39.4, 30.5, 21.6, 21.1 ppm; IR ($CHCl_3$): 3425, 3032, 2917, 1693, 1467, 1422, 1360, 1324, 1305, 1262, 1230, 1211, 1158, 1099, 1056, 994, 907, 729, 698 cm^{-1}.

4.6.3.3 Synthesis of (E)-benzyl 2-(2-(3-oxobut-1-enyl)-1H-indol-3-yl)ethylcarbamate (67)



To the solution of compound **73** (3.39 g, 10.8 mmol) in DMF (80 mL) at 0 ^{o}C was added NaH (60% dispersion, 0.65 g, 16.3 mmol, 1.5 eq). The mixture was stirred at 0 ^{o}C for 30 min and then at room temperature for 2 h. After completion, the reaction was quenched with water, diluted with ether (100 mL), and the organic phase was extracted. The organic layer was washed with water (5 x 50 mL) and then brine, dried with sodium sulfate, filtered and concentrated under reduced pressure. Residual product was purified by silica gel chromatography using 7.5:2.5 hexanes/ethyl acetate to give 3.14 g (80%): ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.78 (br s, H), 7.64-7.60 (m, 2H), 7.35-7.25 (m, 7H), 7.12 (t, *J* = 7.3 Hz, 1H), 6.54 (d, *J* = 16.4 Hz, 1H), 5.09 (s, 2H), 4.83 (m, 1H), 3.48 (dd, *J* = 13.2, 6.4 Hz, 2H), 3.15 (t, *J* = 6.8 Hz, 2H), 2.37 (s, 3 H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): δ 198.0, 156.1, 137.6, 136.5, 130.6, 128.6, 128.2, 128.1, 125.6, 123.8, 120.5, 119.8, 111.4, 66.7, 42.0, 27.4, 24.8 ppm; IR ($CHCl_3$): 3317, 3062, 2942, 1695, 1605, 1638, 1585, 1520, 1454, 1325, 1259, 1130, 1072, 1004, 967, 908, 733, 697 cm^{-1}.

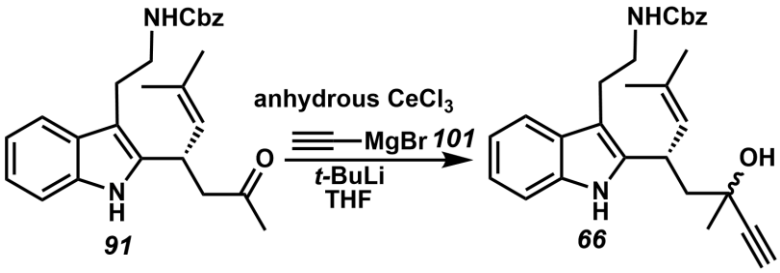
4.6.3.4 General procedure for conjugate addition to synthesize (S)-benzyl 2-(2-(2-methyl-6-oxohept-2-en-4-yl)-1H-indol-3-yl)ethylcarbamate (91)



To a flask equipped with a stir bar was added powdered molecular sieves (3.28 g), and the flask was flame-dried under high vacuum. The flask was then back filled with Argon. Tryptamine enone **67** (0.250 g, 0.68 mmol), Cs_2CO_3 (0.224 g, 0.68 mmol, 1.0 eq), BINOL catalyst (0.074 g, 0.13 mmol, 0.2 eq), and boronic acid (0.206 g, 2.06 mmol, 3.0 eq) were then added. The flask was sealed with a rubber septum, flushed with Argon and filled with freshly distilled dichloroethane (25 mL) and the reaction was heated to 75 ^{o}C. After 23 hours, the reaction was monitored closely by TLC and stopped when decomposition of the product was observed. Reaction was then cooled to room temperature, filtered using Buchner funnel, washed with ethyl acetate, concentrated and purified by silica gel chromatography using 8:2 hexane/ethyl acetate to give 0.19 g (67%) product and 0.037 g (15%) of starting material. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.40 (bs, 1H), 7.43 (d, *J* = 7.7 Hz, 1H), 7.26-7.19 (m, 5H), 7.05 (td, *J* = 7.7, 0.9 Hz, 1H), 6.98 (td, *J* = 7.7, 0.9 Hz, 1H), 5.35 (d, *J* = 9.1 Hz, 1H), 5.05-4.98 (m, 2H), 4.81 (bt, *J* = 5.5 Hz, 1H), 4.20-4.15 (m, 1H), 3.44-3.34 (m, 2H), 2.91-2.81 (m, 3H), 2.71 (dd, *J* = 16.9, 5.5 Hz, 1H), 2.00 (s, 3H), 1.59 (s, 3H), 1.57 (s, 3H) ppm; ^{13}C NMR (125.77 MHz, $CDCl_3$): δ 208.4, 156.7, 138.2, 135.7, 134.7, 128.8, 128.5, 128.4, 124.1, 121.8, 119.7, 118.6, 111.0,

107.6, 66.9, 49.7, 41.8, 31.9, 31.1, 26.0, 25.1, 18.3 ppm; IR ($CHCl_3$): 3341, 2931, 1698, 1640, 1585, 1517, 1448, 1378, 1356, 1246, 1136, 1070, 1003, 908, 839, 731, 697 cm^{-1}.

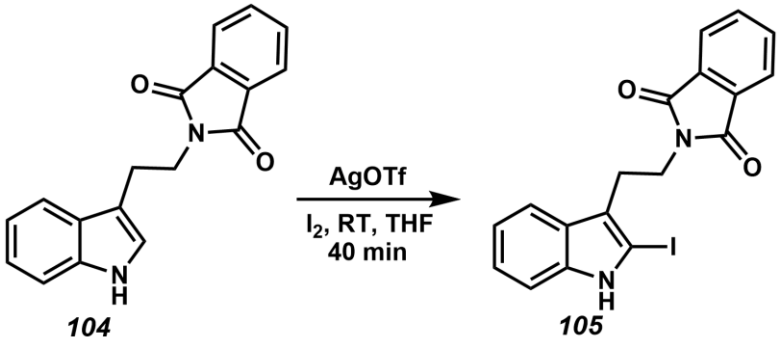
4.6.3.5 Synthesis of (S)-benzyl2-(2-(6-hydroxy-2,6-dimethyloct-2-en-7-yn-4-yl)-1H-indol-3-yl)ethylcarbamate (66)



A solution of anhydrous cerium chloride (159 mg, 0.645 mmol) in 2 mL of dry THF was stirred for 21 hours at room temperature. *t*-BuLi (1.7 M in pentane, 0.12 mL, 0.215 mmol, 1.0 eq) was added at -78 °C . The solution turned to a pale pink color. Ethynyl magnesium bromide (0.5 M in hexane, 1.07 mL, 0.537 mmol) was then added at the same temperature, and the reaction was stirred at -78 °C for 15 minutes and then at 0 °C for 20-30 minutes. The reaction mixture was brought to -78 °C, and the starting material (90 mg, 0.215 mmol) in 2 mL of THF was added. After 15 minutes, the reaction was quenched with saturated aqueous ammonium chloride, and extracted with ethyl acetate (2 x 4 mL). The organic layer was then washed with brine, dried with sodium sulfate, filtered, and evaporated under vacuum. The crude material was purified on silica gel using 7.5:2.5 hexane/ethylacetate as eluent to give 10.5 mg (11%) of the product, which contain two diastereomers, and 10 mg (11%) of the starting material. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.08 (bs, 1H), 8.03 (bs, 1H), 7.53 (d, *J* = 7.3 Hz, 2H), 7.34-7.30 (m,

10H), 7.27 (t, *J* = 7.7 Hz, 2H), 7.20 (t, *J* = 7.7 Hz, 2H), 7.09 (m, 2H), 5.48 (d, *J* = 9.6 Hz, 1H), 5.36 (d, *J* = 8.7 Hz, 1H), 5.09 (bs, 1H), 5.05 (bs, 2H), 5.01 (app.t, *J* = 5.04 Hz, 1H), 4.91 (app.t, *J* = 5.04 Hz, 1H), 4.48-4.27 (m, 2H), 3.54 (d, 5.94 *J* = Hz, 4H), 3.07-2.97 (tt, *J* = 12.3 Hz, 6.8 Hz, 4H), 2.55 (s, 1H), 2.49 (s, 1H), 2.37 (s, 6H), 1.9-2.2 (m, 4H), 1.77 (s, 3H), 1.71 (s, 3H), 1.69 (s, 3H), 1.66 (s, 3H) ppm; IR ($CHCl_3$): 3302, 2975, 2931, 1701, 1613, 1585, 1517, 1461, 1375, 1245, 1132, 1072, 1008, 909, 734, 698 cm^{-1}.

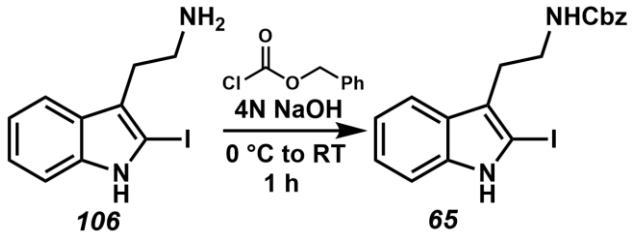
4.6.3.6 Synthesis of 2-(2-(2-iodo-1H-indol-3-yl)ethyl)isoindoline-1,3-dione (105)



To the solution of starting material (500 mg, 1.72 mmol) and iodine (482 mg, 1.90 mmol, 1.1 eq) in 16 mL of THF was added AgOTf dissolved in 5 mL of THF drop wise for 10 minutes. After 30 minutes of stirring, the reaction was quenched with saturated aqueous $Na_2S_2O_3$ and extracted with ethyl acetate (2 x 15mL). The organic layer was then washed with brine, dried with sodium sulfate, filtered, and concentrated under reduced pressure. Residual product was purified by trituration with hexane to give a yellow amorphous solid (500 mg, 70%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.11 (bs, 1H), 7.81-7.79 (m, 2H), 7.69-7.67(m, 2H), 7.62 (d, *J* = 7.7 Hz, 1H), 7.25 (d, *J* = 9.6 Hz, 1H), 7.14-7.05(m, 2 H), 3.96 (t, *J* = 7.3 Hz, 2H), 3.11 (t, *J* = 7.7 Hz, 2H) ppm; ^{13}C NMR

(125.77 MHz, CDCl3): δ 168.3, 136.09, 133.9, 132.2, 127.7, 123.2, 122.5, 122.4, 120.3, 118.1, 112.0, 110.5, 108.7, 37.5, 24.1 ppm; IR ($CHCl_3$): 3385, 3333, 3059, 2944, 1769, 1698, 1615, 1552, 1448, 1467, 1434, 1395, 1362, 1336, 1302, 1230, 1187, 1171, 1126, 1104, 1076, 1014, 1003, 907, 870, 715 cm^{-1}.

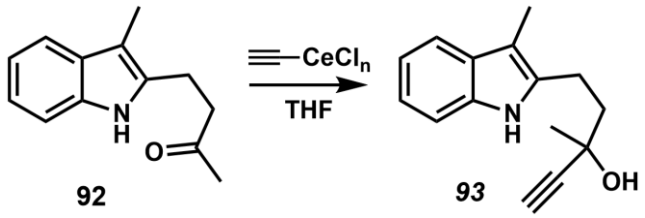
4.6.3.7 Synthesis of benzyl 2-(2-iodo-1H-indol-3-yl)ethylcarbamate (65)



To the solution of starting material (150 mg, 0.524 mmol) in DCM:MeOH (3mL:3mL) was added benzylchloroformate and 4 N NaOH at 0℃. After stirring it for 1 hr at room temperature, the reaction mixture was quenched with water and extracted with DCM (2 x 10 mL). The organic layer was then washed with brine, dried it over sodium sulfate, filtered and the solvent was evaporated under vacuum. The residual product was purified by silicagel chromatography using 7:3 Hexane/Ethyl acetate to give 130 mg (60%) of product. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.46 (bs, 1H), 7.51 (d, *J* = 7.7 Hz, 1H), 7.40-7.36 (m, 5H), 7.29 (t, *J* = 7.3 Hz, 1H), 7.13 (t, *J* =7.3 Hz, 1H), 7.10 (t, *J* = 7.7 Hz, 1 H), 5.18 (s, 2H), 4.8 (bs, 1H), 3.81 9s, 3H), 3.49 (m, 2H), 2.95 (t, *J* = 6.8 Hz, 2H) ppm; ^{13}C NMR (125.77 MHz, CDCl3): δ 156.6, 155.9, 139.0, 137.0, 136.0, 128.7, 128.69, 128.64, 128.4, 128.3, 128.2, 122.4, 120.0, 118.7, 117.9, 110.6, 78.9, 69.8, 66.7, 55.0,

41.2, 27.3 ppm; IR (DCM): 3401, 3327, 2945, 1693, 1607, 1515, 1466, 1454, 1416, 1348, 1309, 1247, 1199, 1106, 1043, 1002, 912, 803, 733, 696 cm^{-1}.

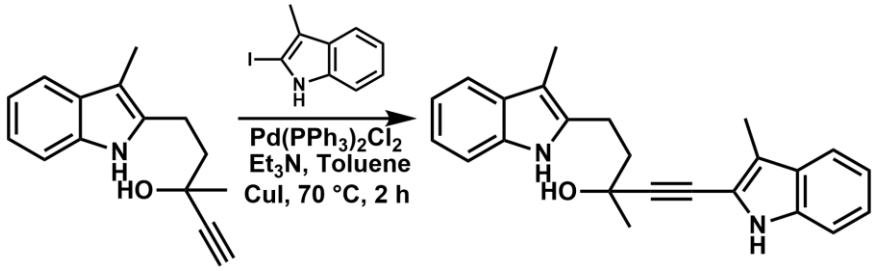
4.6.3.8 Synthesis of 3-methyl-5-(3-methyl-1H-indol-2-yl)pent-1-yn-3-ol (93)



A solution of anhydrous cerium chloride (182 mg, 0.496 mmol) in 1.5 mL of dry THF was stirred for 21 hours at room temperature. *t*-BuLi (1.7 M in pentane, 0.3 mL, 0.215 mmol, 1.0 eq) was added at -78 °C. The solution turned to a pale pink color. Ethynyl magnesium bromide (0.5 M in hexane, 1.24 mL, 0.620 mmol) was then added at the same temperature and allowed to stir at -78 °C for 15 minutes and then at 0 °C for 20-30 minutes. Then the starting material (50 mg, 0.248 mmol) in 2 mL of THF was added at -78 °C. The reaction was quenched with a saturated aqueous ammonium chloride after 15 minutes and then extracted with ethyl acetate (2x4 mL). Organic layer was then washed with brine, dried with sodium sulfate, filtered, and evaporated under vacuum. The crude material was purified on silica gel using 7.5:2.5 hexane/ethylacetate as eluent to give 36.6 mg (65%) of product, and 6.5 mg (13%) of starting material. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.94 (bs, 1H), 7.49 (d, *J* = 7.4 Hz, 1H), 7.27 (d, *J* = 8.0 Hz, 1H), 7.07-7.12 (m, 2H), 3.05-2.96 (m, 2H), 2.55 (s, 1H), 2.26 (s, 3H), 2.0 (t, *J* = 8.0 Hz, 2H), 1.56 (s, 3H) ppm; ^{13}C NMR (125.77 MHz, CDCl3): δ 135.3, 134.3, 129.4, 121.2, 119.1, 118.2,

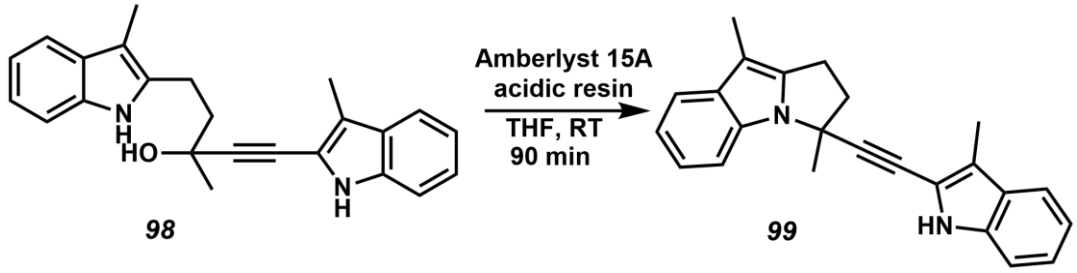
110.3, 107.2, 87.1, 72.3, 68.1, 42.7, 30.4, 21.6, 8.6 ppm; IR (benzene): 3288, 3090, 3035, 2979, 1615, 1464, 1478, 1373, 1334, 1240, 1161, 1087, 1035, 1005, 858, 747, 676 cm^{-1}.

4.6.3.9 Synthesis of 3-methyl-1,5-bis(3-methyl-1H-indol-2-yl)pent-1-yn-3-ol (98)



To a degassed mixture of the palladium catalyst (4.3 mg, 0.006 mmol) and copper iodide (2.3 mg, 0.012 mmol) was added 3 mL of freshly distilled triethylamine followed by a solution of propargylic alcohol (70 mg, 0.30 mmol) and 2-iodo-3-methylindole (79 mg, 0.30 mmol) in toluene (0.35 mL). The reaction was heated to 70 °C and stirred it for 2 h. After all the starting material was consumed, the reaction mixture was quickly filtered through a small silica gel pad to remove the catalyst. The crude material was then chromatographycally purified on silicagel using 80:20 Hexane/ethylacetate as eluent to give 27.0 mg (24%) of product. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.15 (bs, 1H), 7.54-7.52 (m, 1H), 7.47 (d, *J* = 8.0 Hz, 1H), 7.29-7.26 (m, 3H), 7.20-7.15 (m, 1H), 7.13 (d, *J* = 8.0 Hz, 1H), 7.09-7.05 (m, 1H), 3.5 (s, 3 H), 3.14-3.19 (m, 2H), 2.31 (s, 3H), 2.28 (s, 3H), 2.16 (t, *J* = 7.4 Hz, 2H) ppm; ^{13}C NMR (125.77 MHz, CDCl3): δ 135.7, 135.2, 134.5, 129.2, 127.6, 123.5, 121.2, 119.6, 119.2, 119.1, 118.3, 116.0, 110.90, 111.71, 110.48, 107.5, 97.61, 76.0, 68.2, 50.9, 42.4, 31.4, 21.6, 9.6, 8.6 ppm.

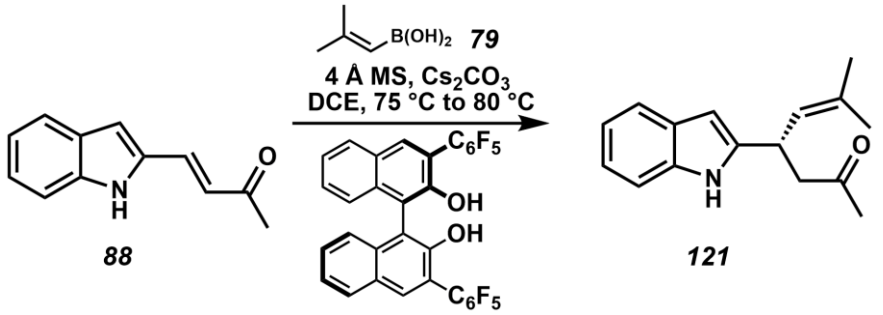
4.6.3.10 Synthesis of 3,9-dimethyl-3-((3-methyl-1H-indol-2-yl)ethynyl)-2,3-dihydro-1H-pyrrolo[1,2-a]indole (99)



98 *99*

To a solution of starting material (8.7 mg, 0.024 mmol) in 0.3 mL of THF was added excess acidic amberlyst 15A (40 mg) and the reaction was stirred for 1.5 h at room temperature. The Amberlyst resin was removed by filtration through cotton and the solution was quenched with basic Amberlyst 21A basic resin. The solvent was evaporated under vacuum. The crude material was purified using preparative thin layer chromatography using 85:15 Hexane/Ethylacetate as eluent to give 2.2 mg (27%) of product. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.87 (bs, 1H), 7.57 (d. *J* = 7.4 Hz, 1H), 7.51-7.49 (dd, *J* = 8.5, 3.4 Hz, 2H), 7.25-7.12 (m, 3H), 7.15-7.08 (m, 3H), 3.06-3.00 (m, 2H), 2.72-2.70 (m, 2H), 2.34 (s, 3H), 2.24 (s, 3H), 1.96 (s, 3H) ppm; ^{13}C NMR (125.77 MHz, CDCl3): δ 139.6, 135.7, 133.7, 131.3, 127.8, 123.6, 120.3, 119.8, 119.2, 118.8, 18.6, 118.3, 115.9, 110.7, 109.8, 101.5, 96.3, 56.6, 44.8, 27.5, 21.8, 9.6, 8.9 ppm.

4.6.3.11 Synthesis of (S)-4-(1H-indol-2-yl)-6-methylhept-5-en-2-one (121)



To a flask equipped with a stir bar was added powdered molecular sieves (1.35 g), and the flask was flamed-dried under high vacuum. The flask was then back-filled with Argon. Indole enone **88** (0.100 g, 0.540 mmol), Cs_2CO_3 (0.176 g, 0.540 mmol, 1.0 eq), BINOL catalyst (0.077 g, 0.108 mmol, 0.2 eq), and boronic acid (0.162 g, 1.62 mmol, 3.0 eq) were then added. The flask was sealed with a septum, flushed with Argon and filled with freshly distilled dichloroethane (10 mL), and the reaction was heated to 75 ^{o}C. After 20 hours, the reaction was monitored closely by TLC and stopped when decomposition of the product was observed. The reaction was then cooled to room temperature, filtered using Buchner funnel, washed with ethyl acetate, concentrated under reduced pressure and purified by silica gel chromatography using 8:2 hexane/ethyl acetate to give 0.093 g (72%) of product and 10 mg (10%) of starting material. ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.35 (bs, 1H), 7.54 (d, *J* = 7.7 Hz, 1H), 7.30 (d, *J* = 7.7 Hz, 1H), 7.12 (td, *J* = 6.8, 1.3 Hz, 1H), 7.06 (td, *J* = 6.8, 1.3 Hz, 1H), 6.19 (s, , 1H), 5.34 (dt, *J* = 9.1, 1.3 Hz, 1H), 4.28 (app.q, *J* = 7.6 Hz, 1H), 2.99 (dd, *J* = 17.8, 7.7 Hz, 1H), 2.84 (dd, *J* = 17.4, 5.5 Hz, 1H), 2.16 (s, 3H), 1.76 (d, *J* = 1.3 Hz, 3H), 1.71 (d, *J* = 1.3 Hz, 3H) ppm; ^{13}C NMR (100 MHz, CDCl3): δ 208.6, 142.0, 135.9, 134.3, 128.2, 124.4, 121.3, 120.0, 119.6, 110.6, 98.3,

49.9, 32.8, 30.7, 25.9, 18.2 ppm; IR (DCM); 3392, 2969, 2927, 1705, 1617, 1487, 1456, 1416, 1375, 1295, 1232, 1155, 1112, 1048, 1013, 782, 749 cm^{-1}.

4.7 References

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APPENDIX TWO

Spectra Relevant to Chapter 4:

Progress Toward The Enantioselective Total Sytnthesis of Flinderoles A–C

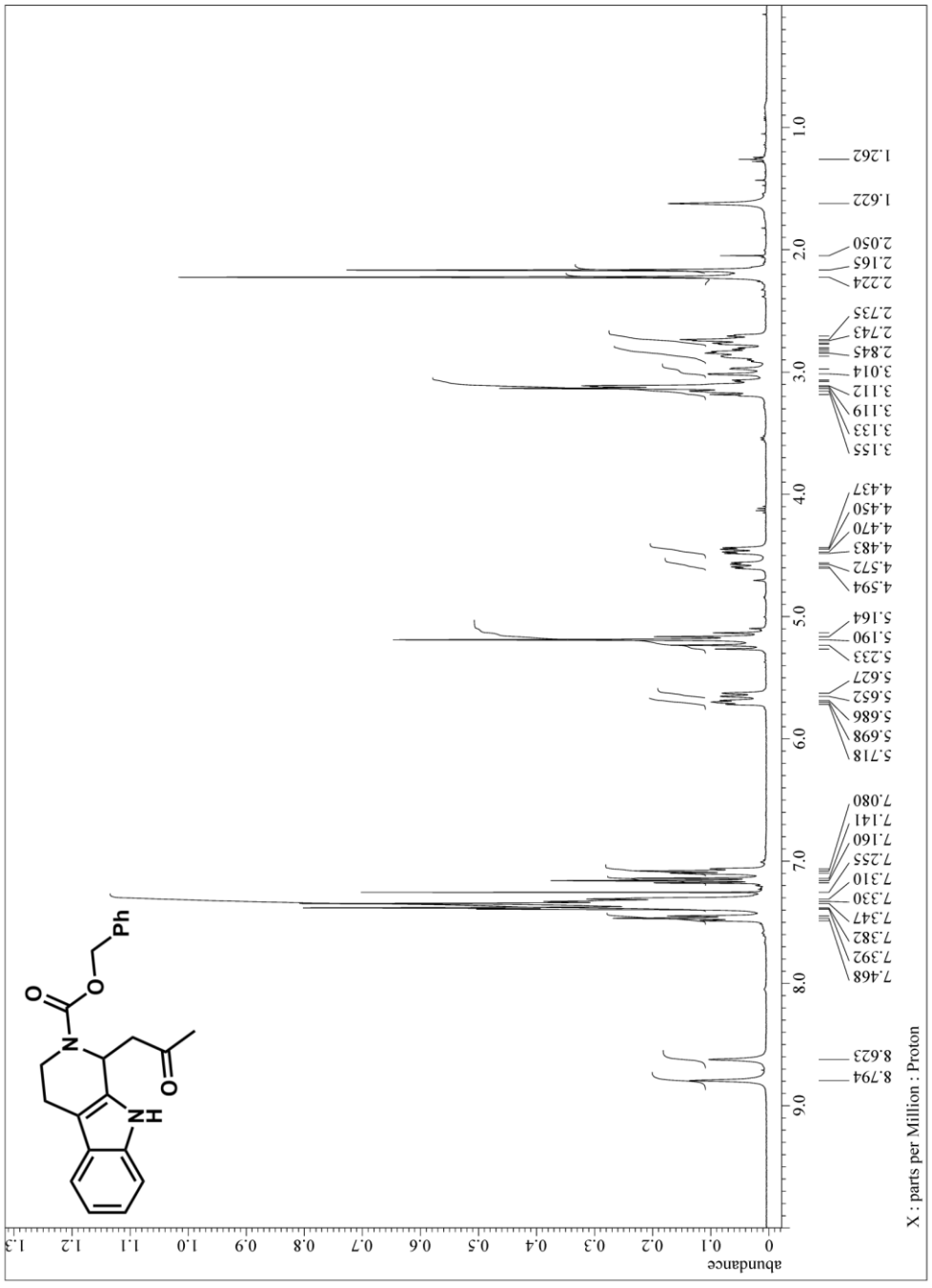


Figure A.2.1 ^{1}H NMR ($CDCl_3$) of compound **73**

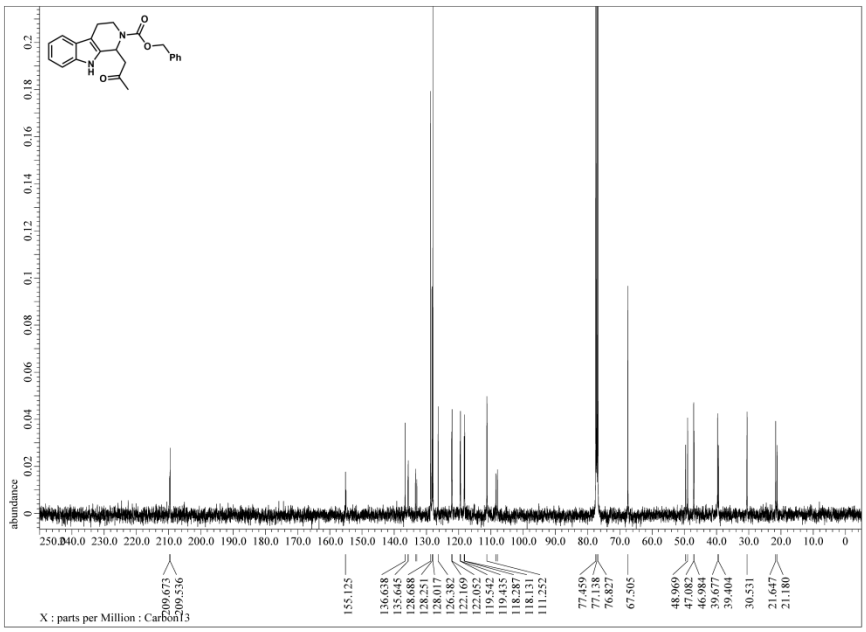


Figure A.2.2 ^{13}C NMR ($CDCl_3$) of compound **73**

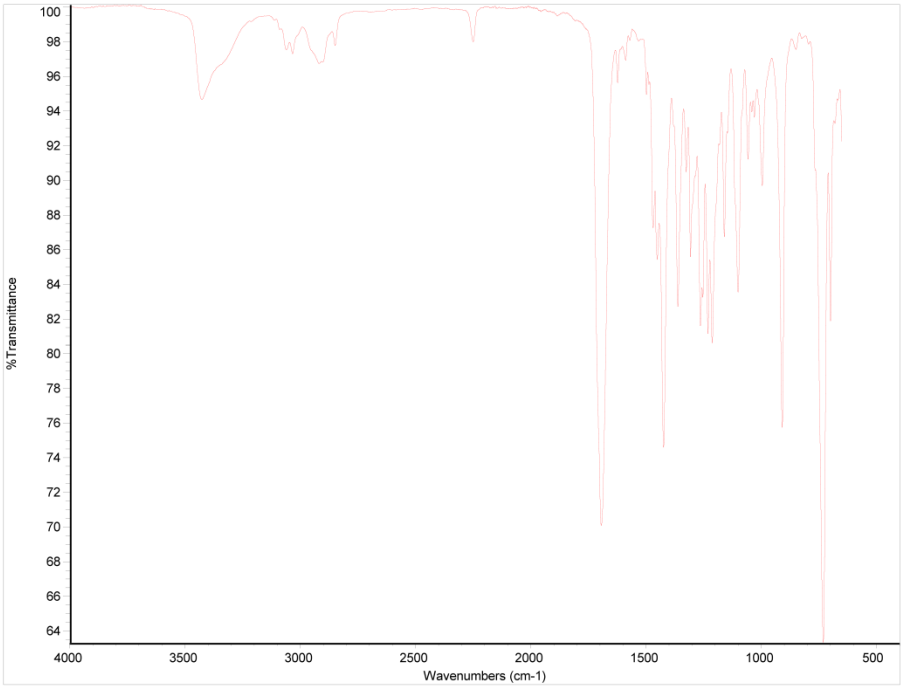


Figure A.2.3 IR spectrum of compound **73**

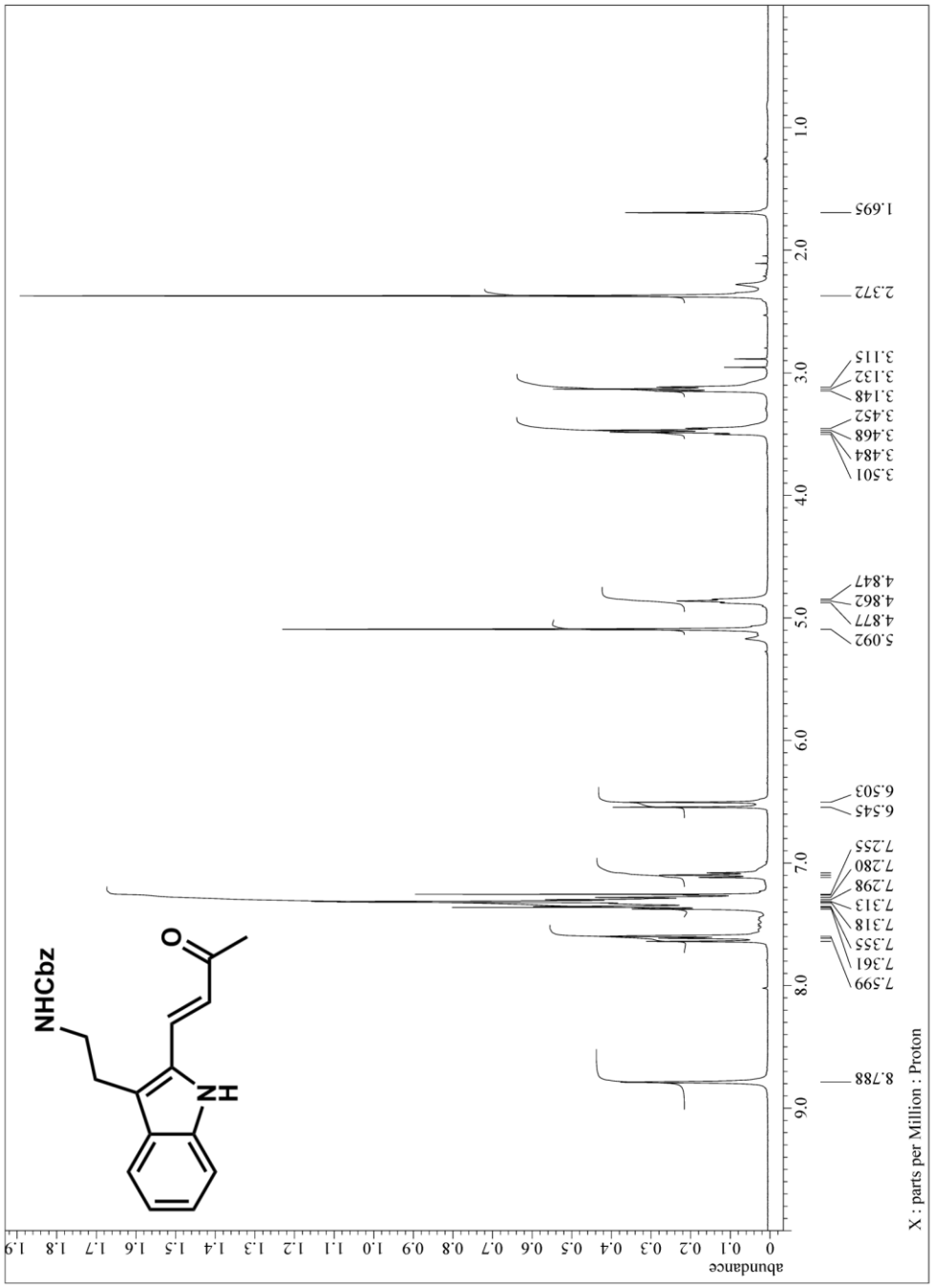


Figure A.2.4 ^{1}H NMR ($CDCl_3$) of compound **67**

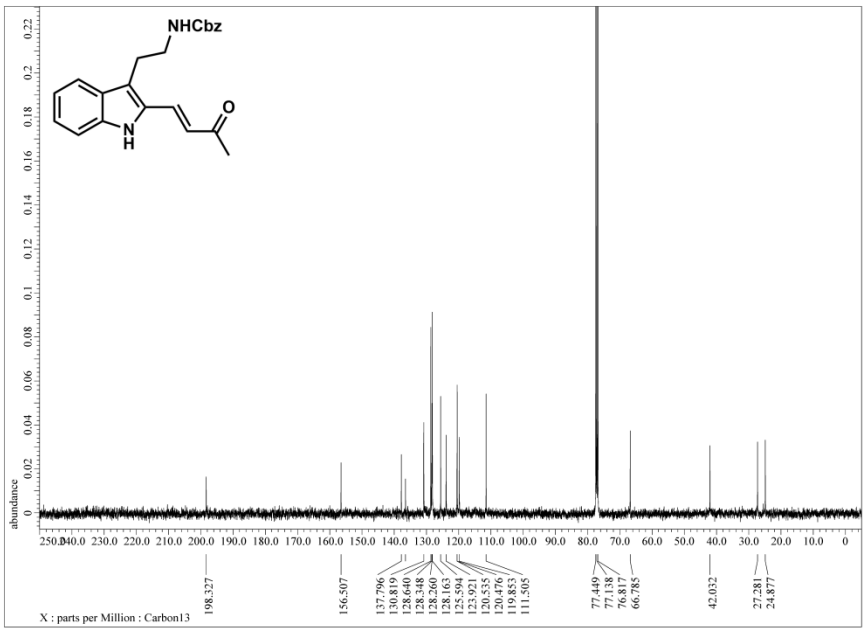


Figure A.2.5 ^{13}C NMR ($CDCl_3$) of compound **67**

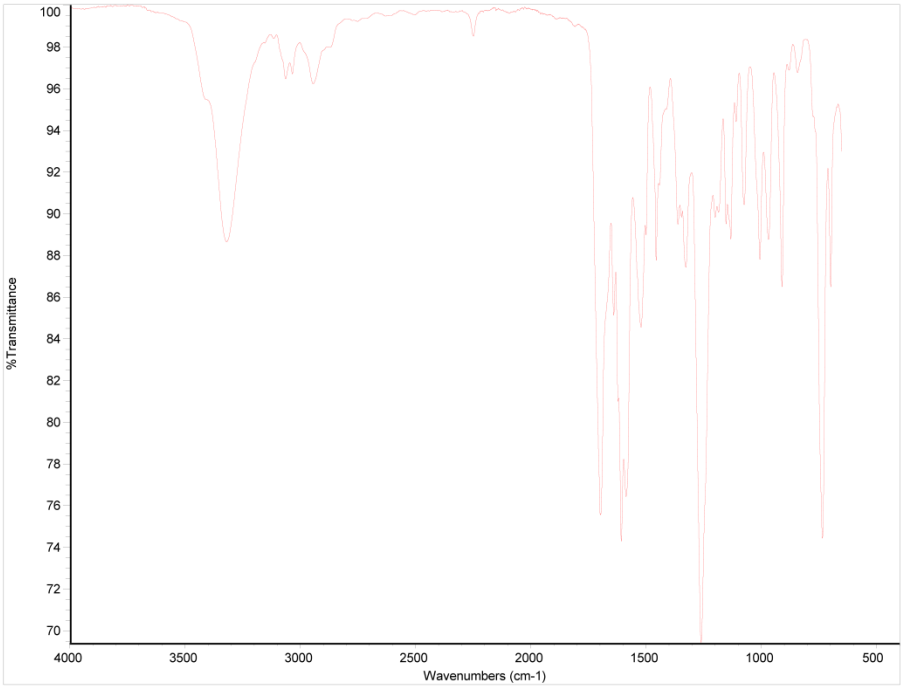


Figure A.2.6 IR spectrum of compound **67**

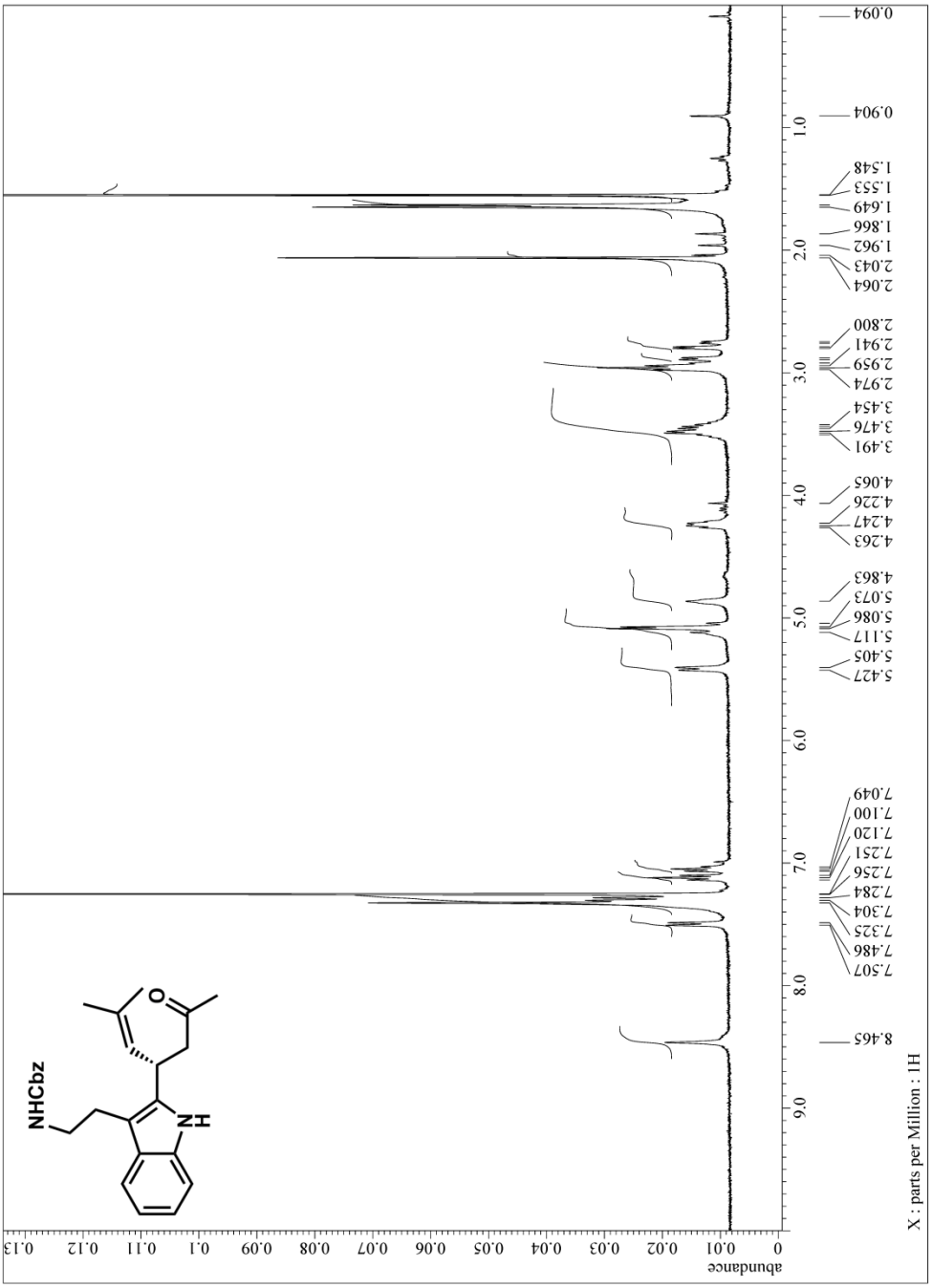


Figure A.2.7 ^{1}H NMR ($CDCl_3$) of compound **91**

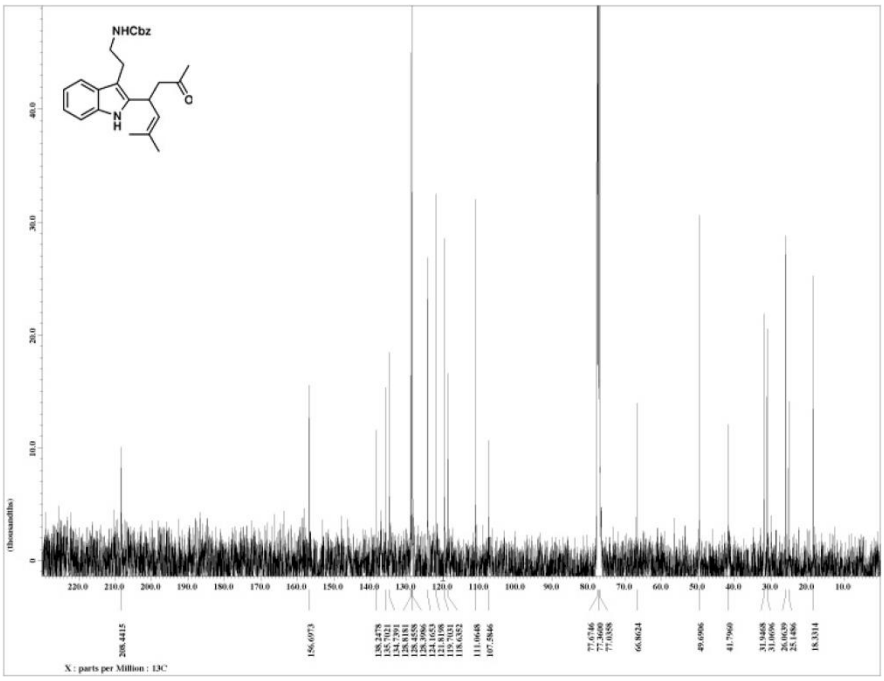


Figure A.2.8 ^{13}C NMR ($CDCl_3$) of compound **91**

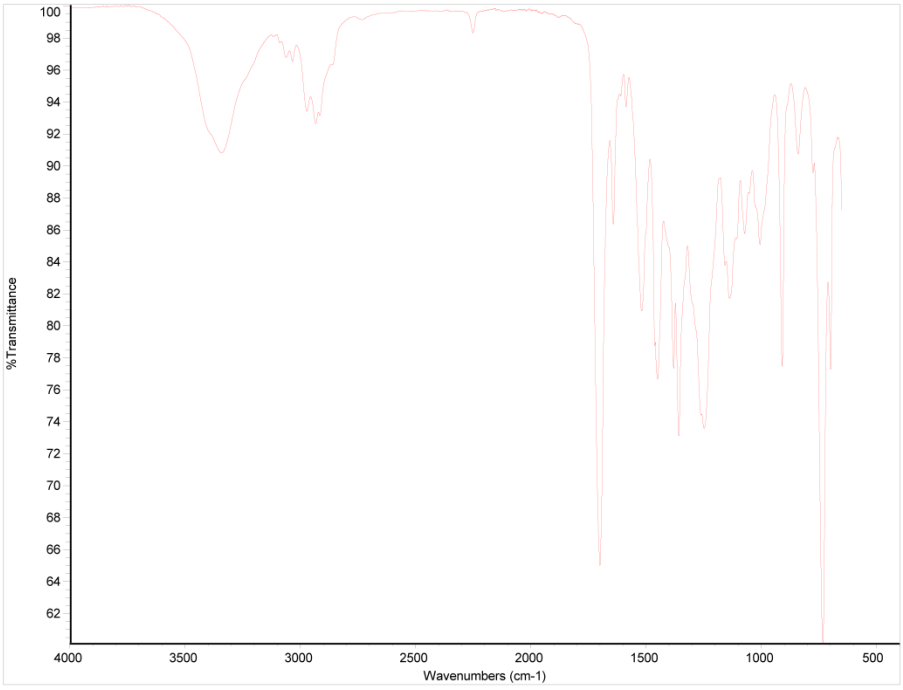
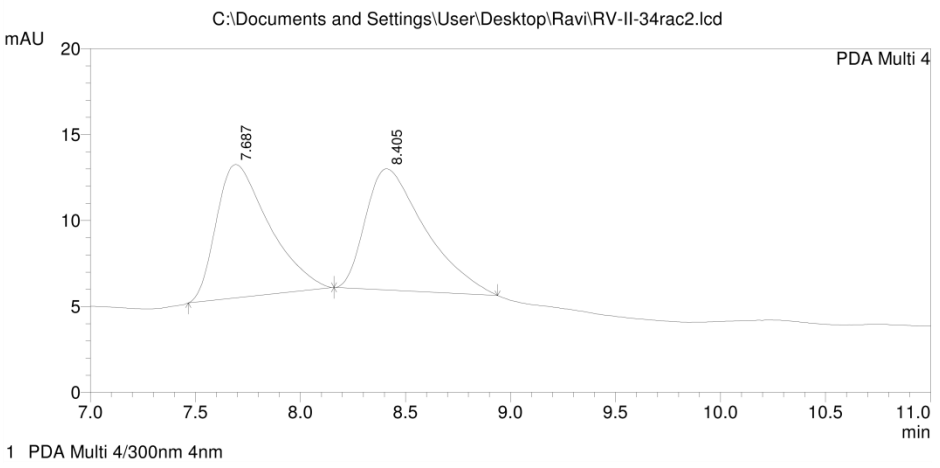


Figure A.2.9 IR spectrum of compound **91**

<Chromatogram>

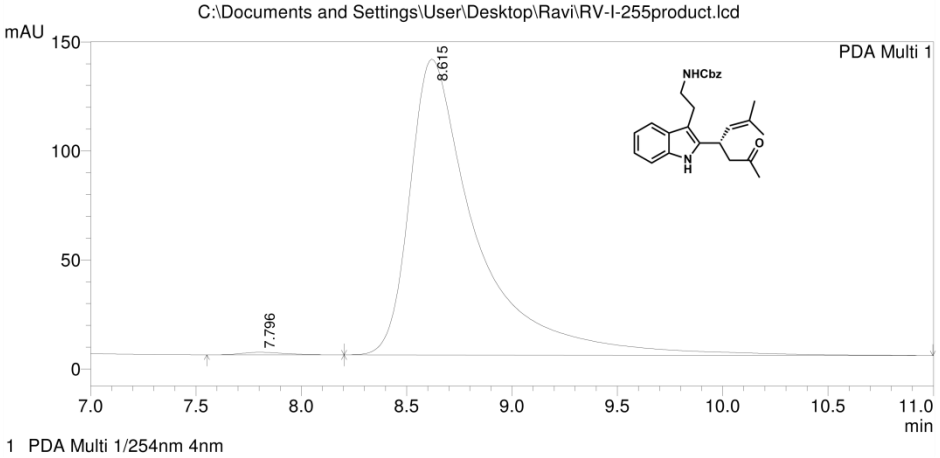


PeakTable

PDA Ch4 300nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	7.687	135959	7779	49.336	52.462
2	8.405	139621	7049	50.664	47.538
Total		275580	14828	100.000	100.000

Fig A.2.10 HPLC trace for racemic compound **91**

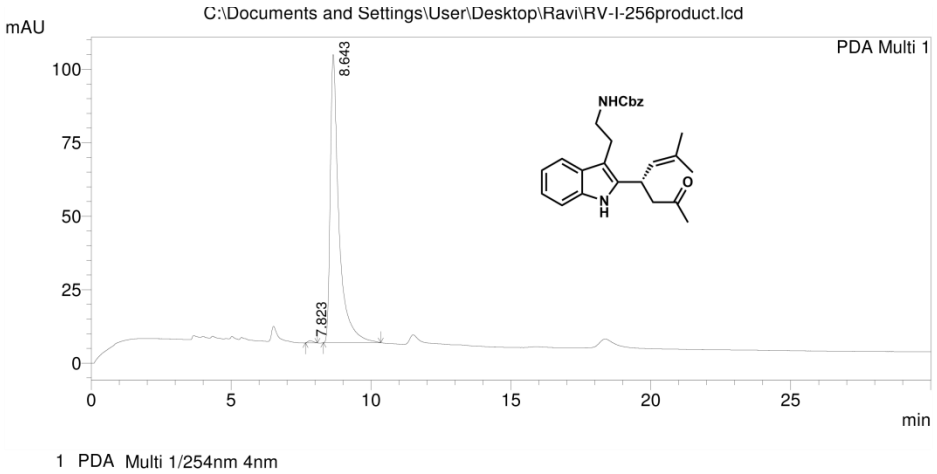


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	7.796	16955	1179	0.561	0.861
2	8.615	3005324	135715	99.439	99.139
Total		3022279	136895	100.000	100.000

Fig A.2.11 HPLC trace for compound **91** (entry 3 Table 4.1)

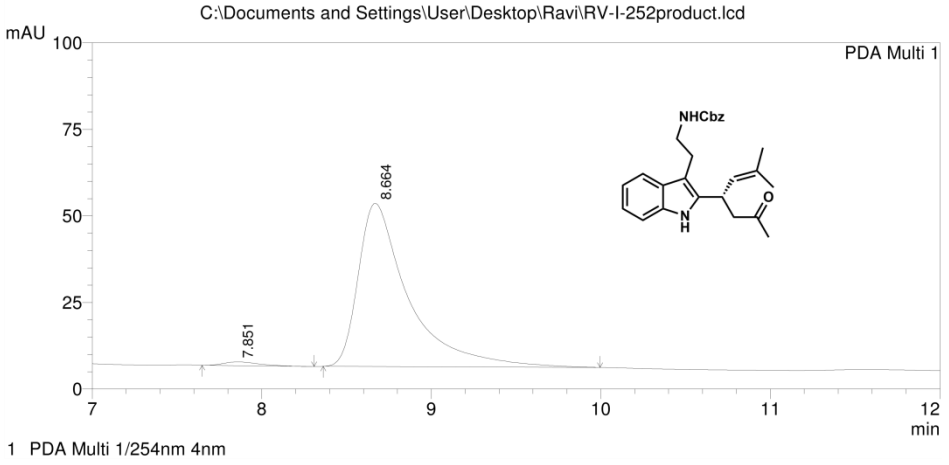


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	7.823	10065	779	0.473	0.788
2	8.643	2116767	98077	99.527	99.212
Total		2126832	98855	100.000	100.000

Fig A.2.12 HPLC trace for compound **91** (entry 4 Table 4.1)

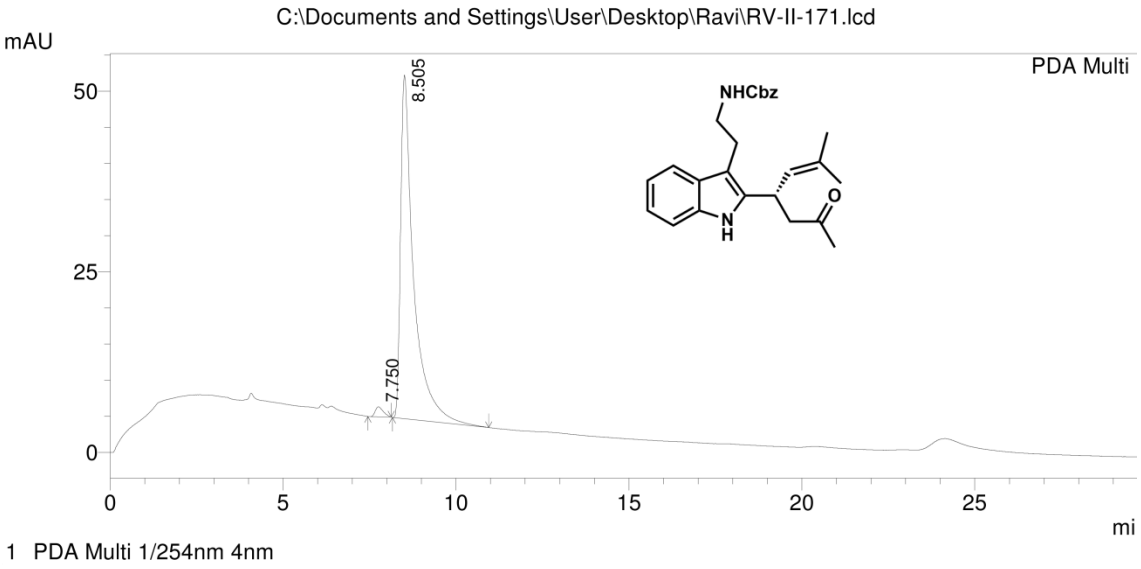


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	7.851	15592	1109	1.580	2.297
2	8.664	971389	47183	98.420	97.703
Total		986981	48292	100.000	100.000

Fig A.2.13 HPLC trace for compound **91** (entry 6 Table 4.1)



PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	7.750	21613	1373	1.718	2.807
2	8.505	1236329	47558	98.282	97.193
Total		1257942	48932	100.000	100.000

Fig A.2.14 HPLC trace for compound **91** (entry 8 Table 4.1)

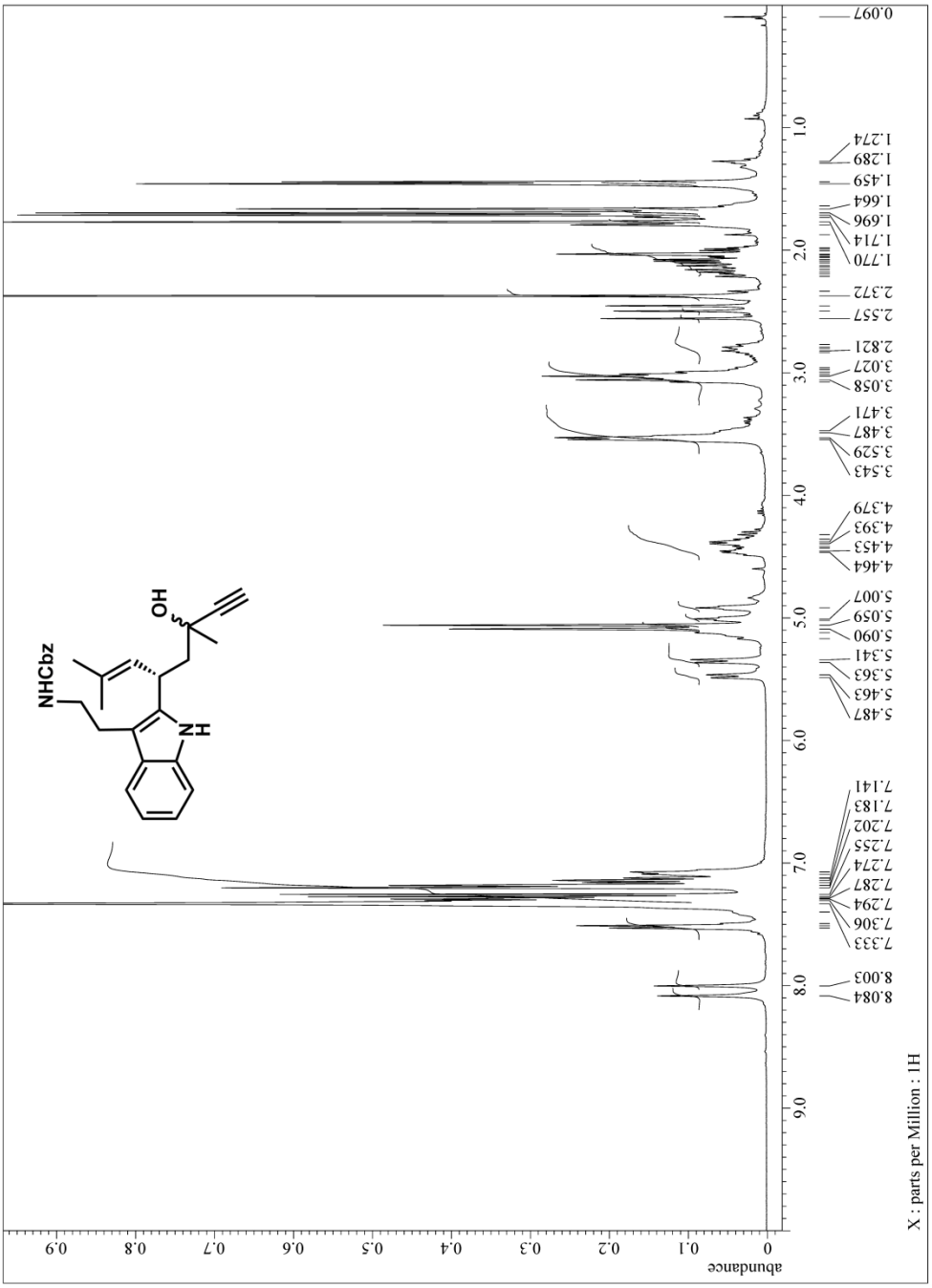


Figure A.2.15 ^{1}H NMR ($CDCl_3$) of compound **66**

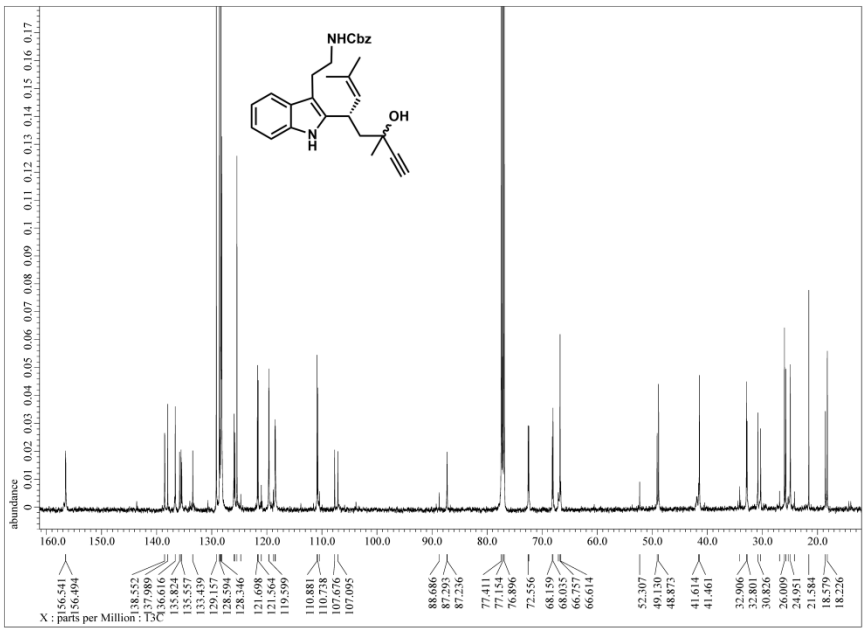


Figure A.2.16 ^{13}C NMR ($CDCl_3$) of compound **66**

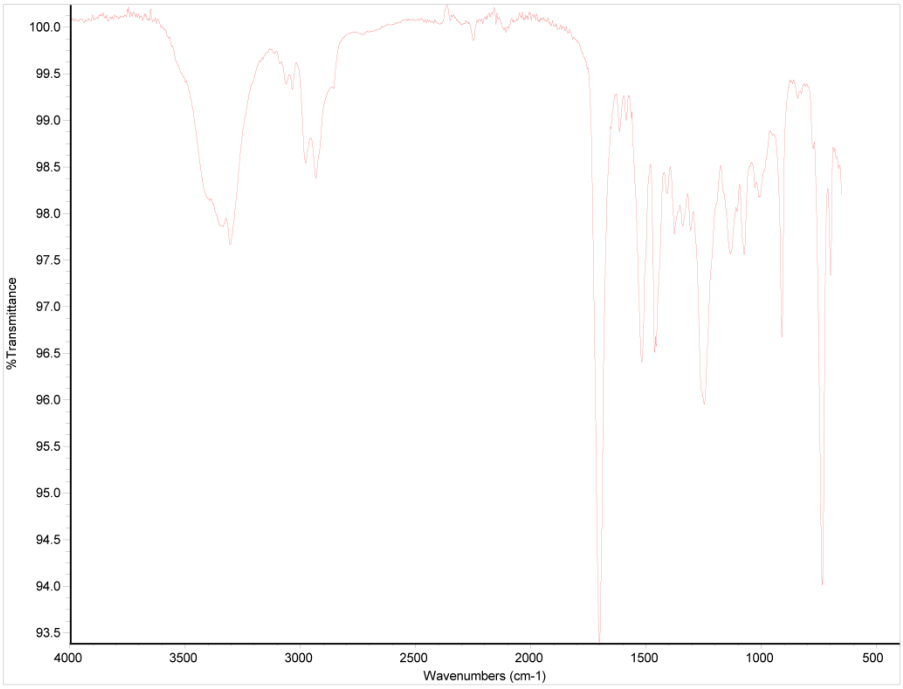


Figure A.2.17 IR spectrum of compound **66**

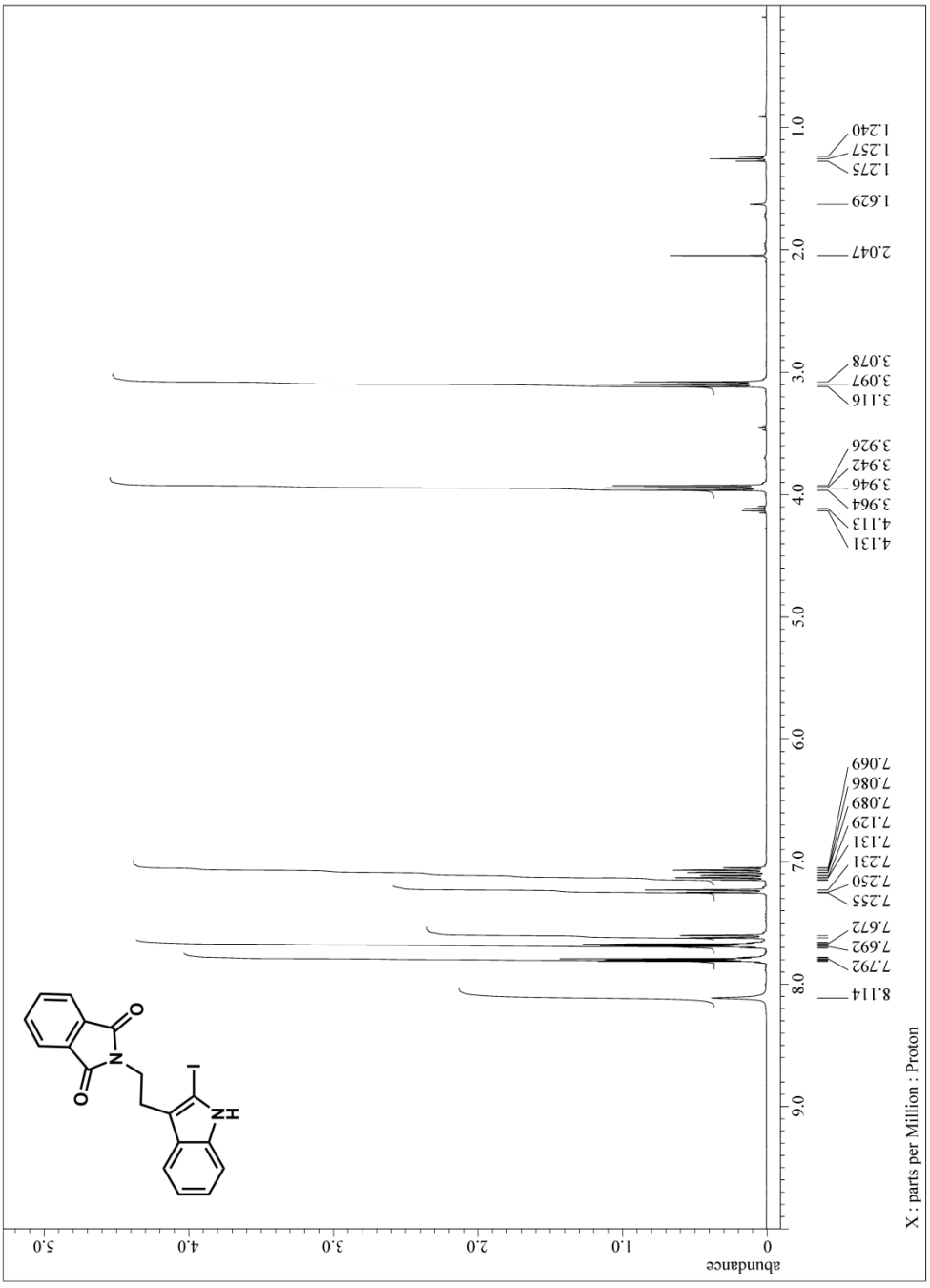


Figure A.2.18 ^{1}H NMR ($CDCl_3$) of compound **105**

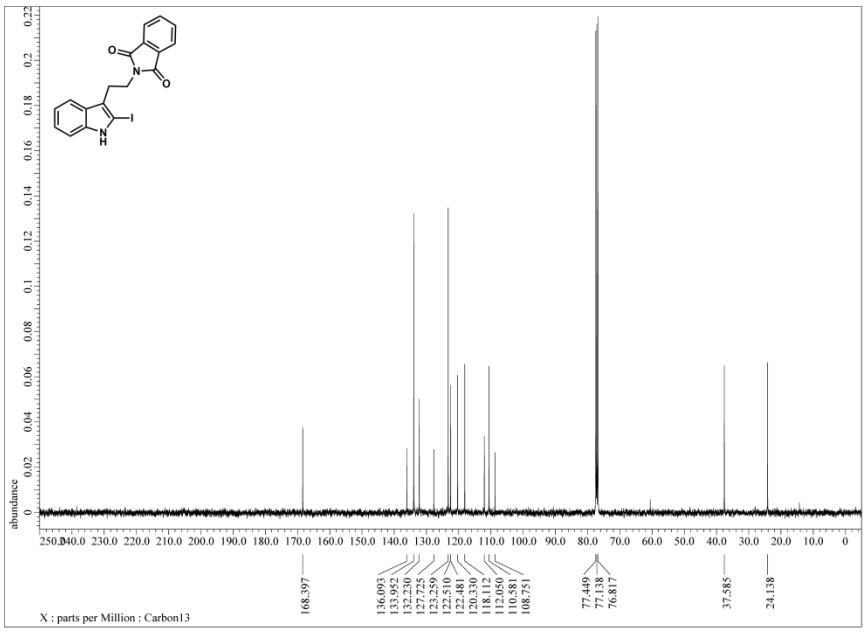


Figure A.2.19 ^{13}C NMR ($CDCl_3$) of compound **105**

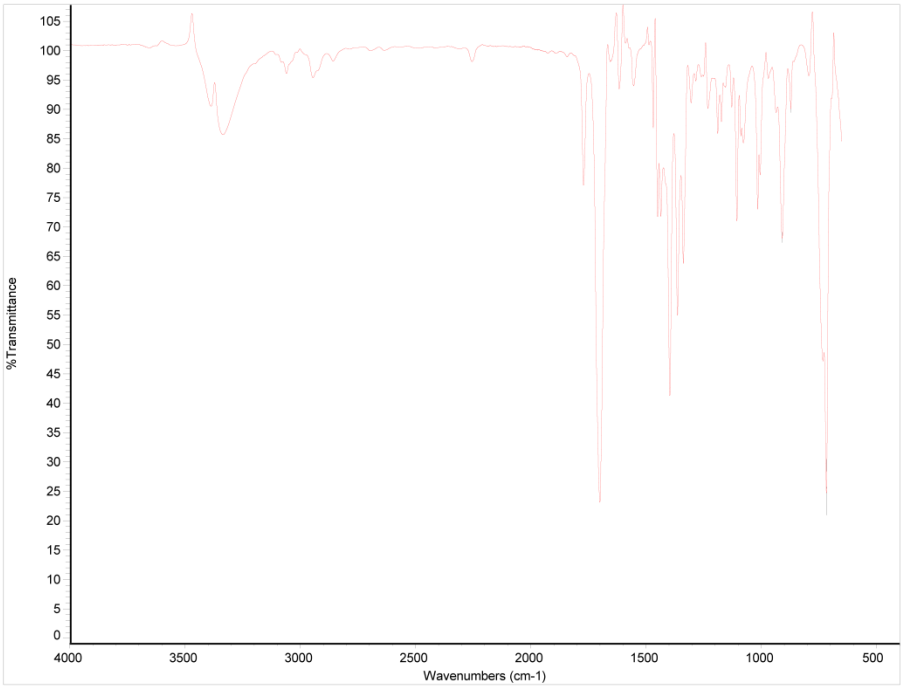


Figure A.2.20 IR spectrum of compound **105**

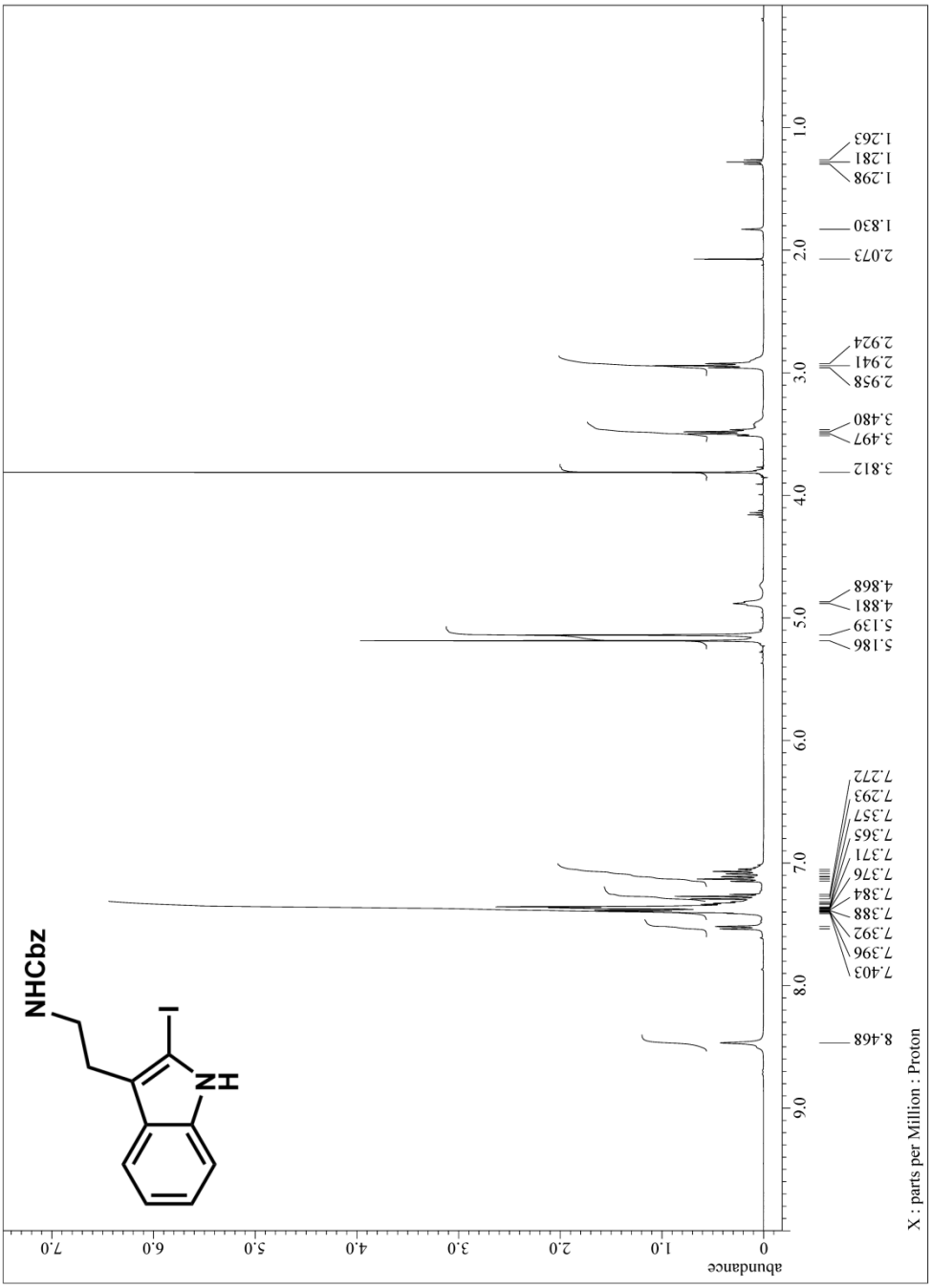


Figure A.2.21 ^{1}H NMR ($CDCl_3$) of compound **65**

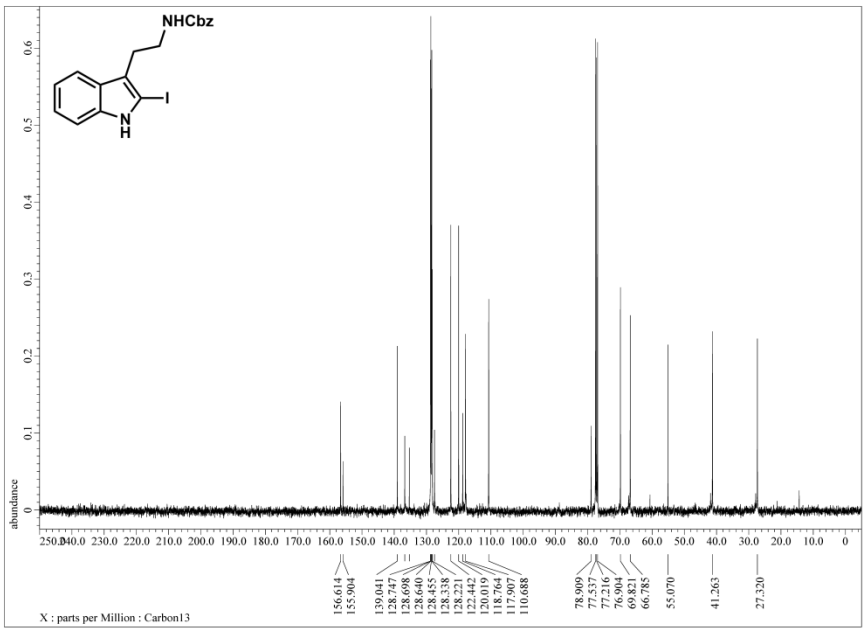


Figure A.2.22 ^{13}C NMR ($CDCl_3$) of compound **65**

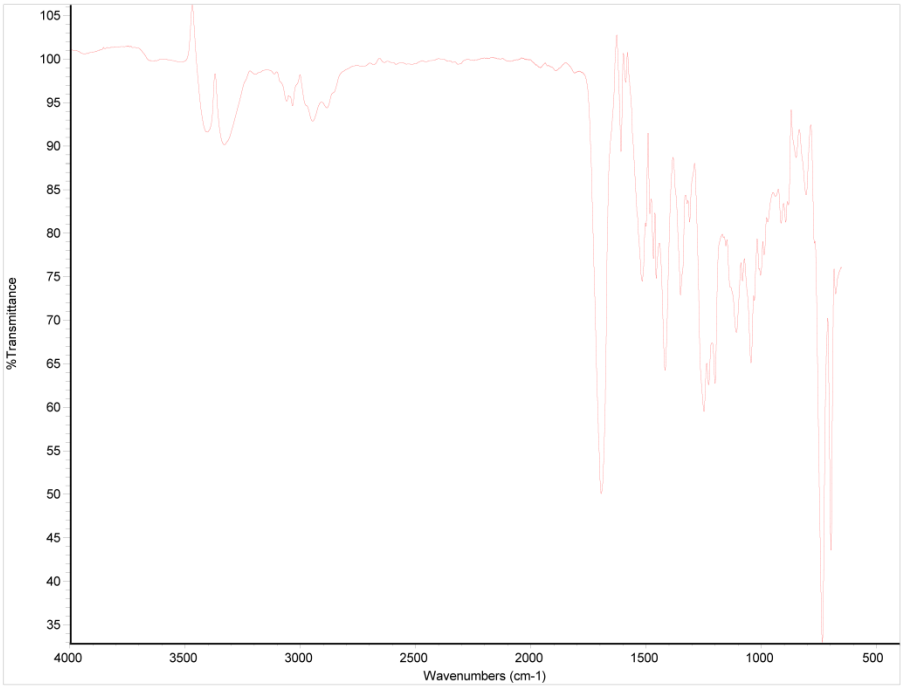


Figure A.2.23 IR spectrum of compound **65**

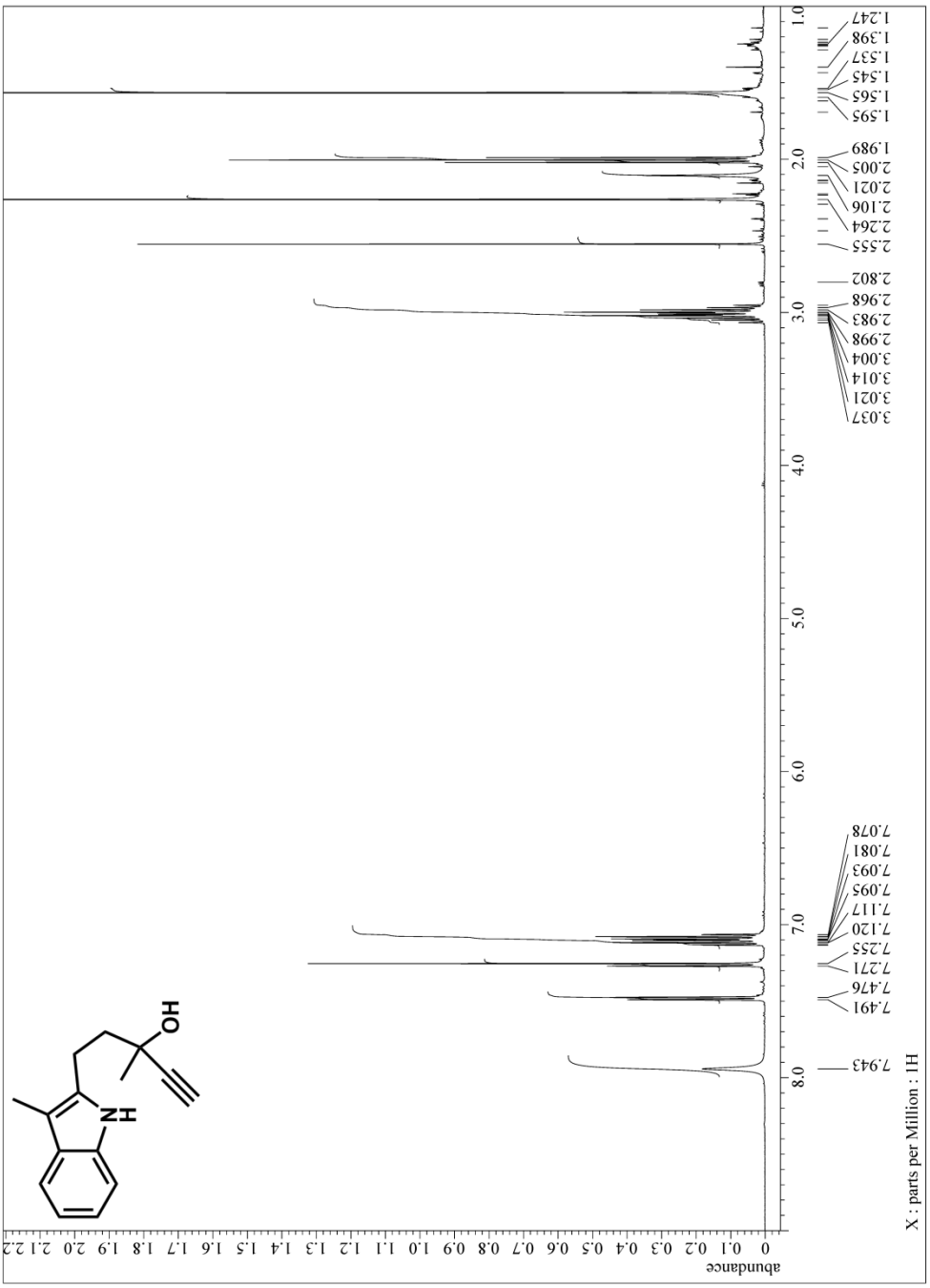


Figure A.2.24 ^{1}H NMR ($CDCl_3$) of compound **93**

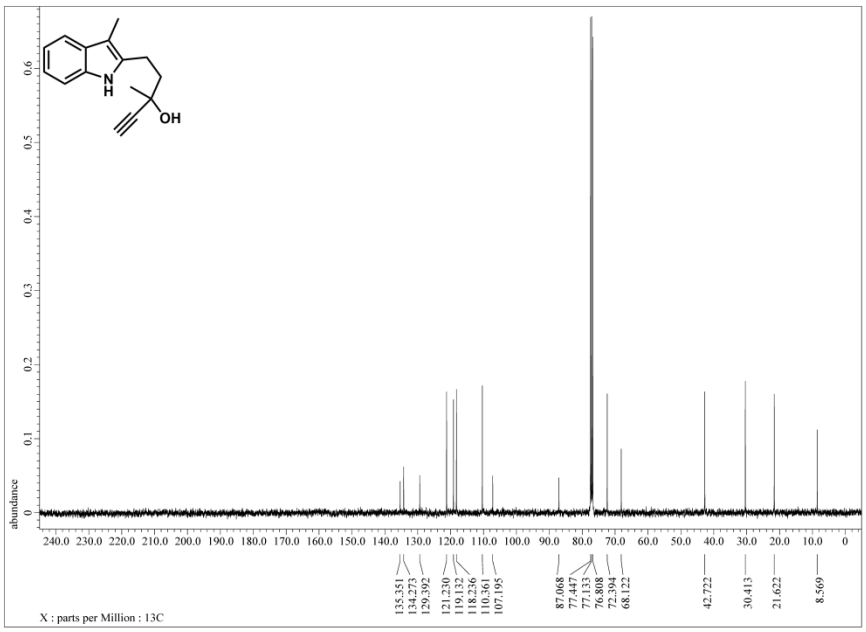


Figure A.2.25 ^{13}C NMR ($CDCl_3$) of compound **93**

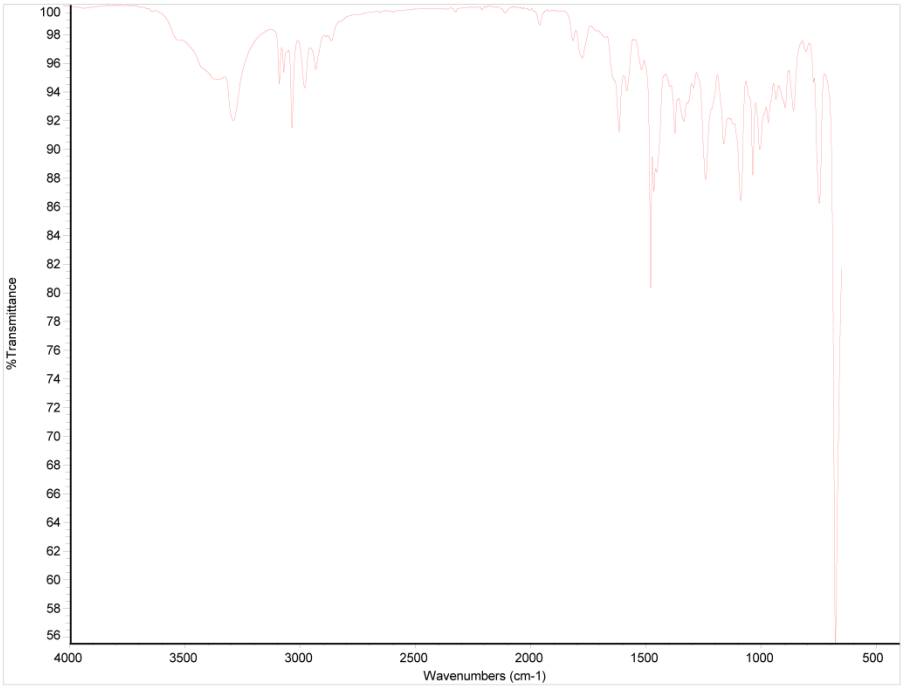


Figure A.2.26 IR spectrum of compound **93**

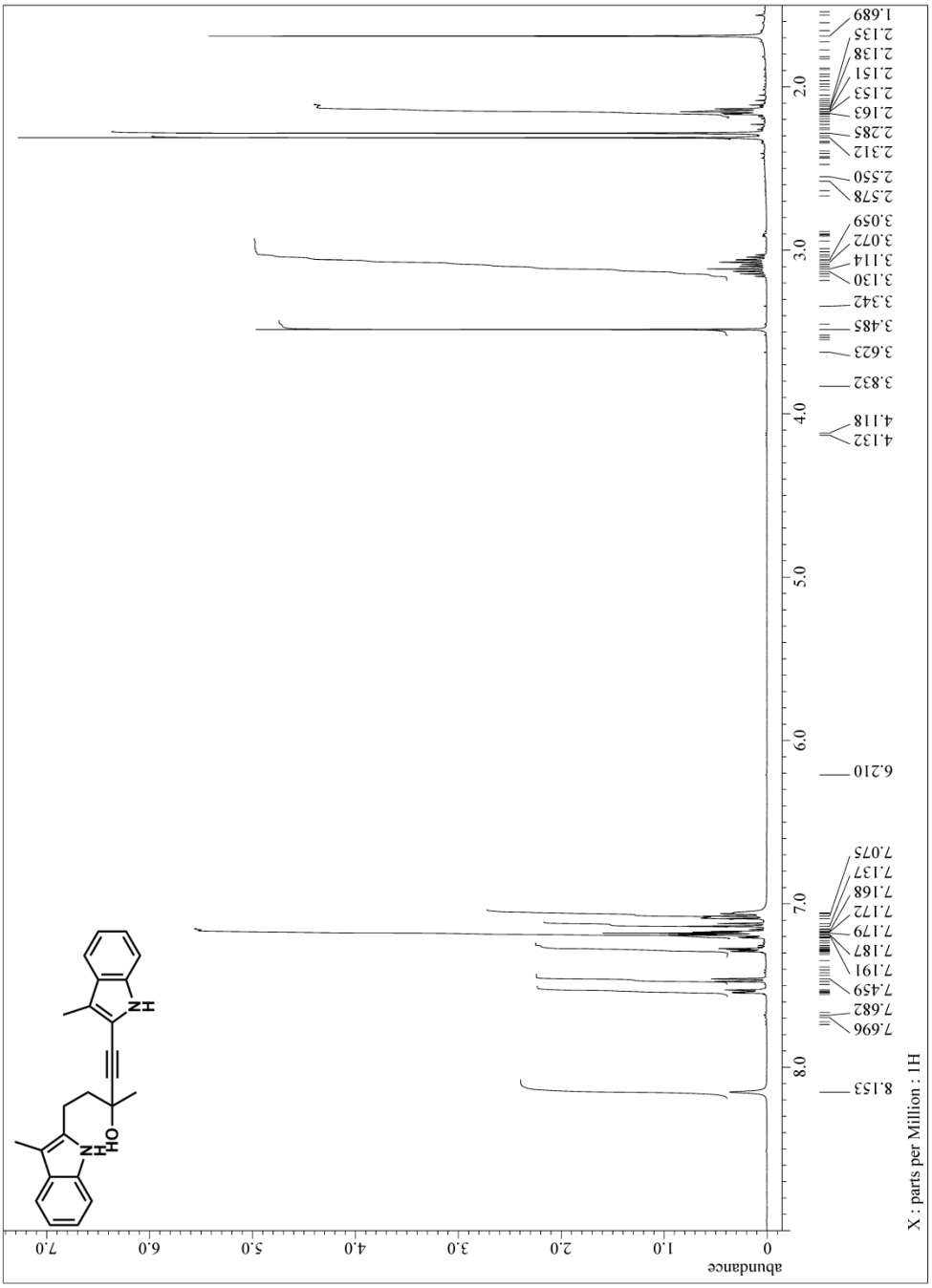


Figure A.2.27 ^{1}H NMR ($CDCl_3$) of compound **98**

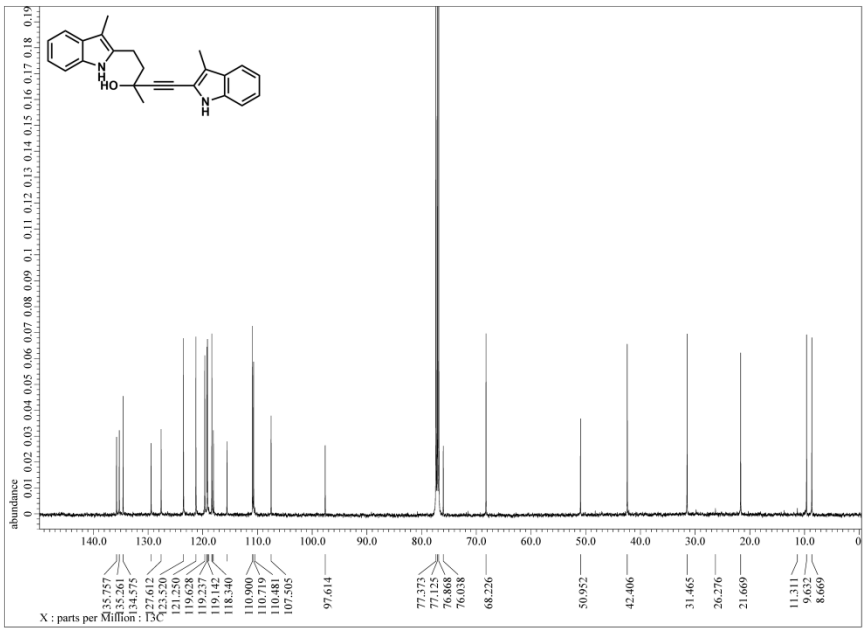


Figure A.2.28 ^{13}C NMR ($CDCl_3$) of compound **98**

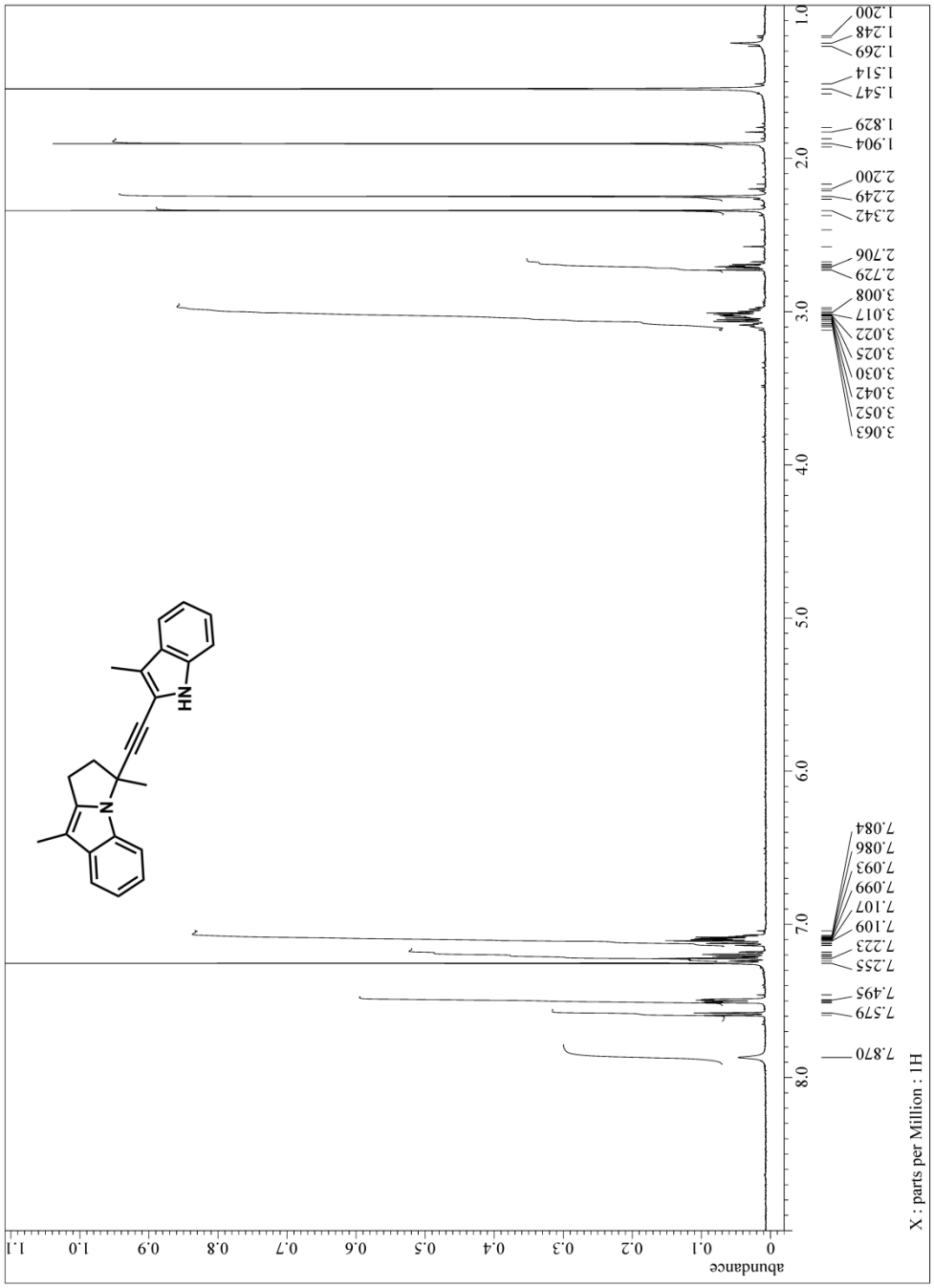


Figure A.2.29 ^{1}H NMR ($CDCl_3$) of compound **99**

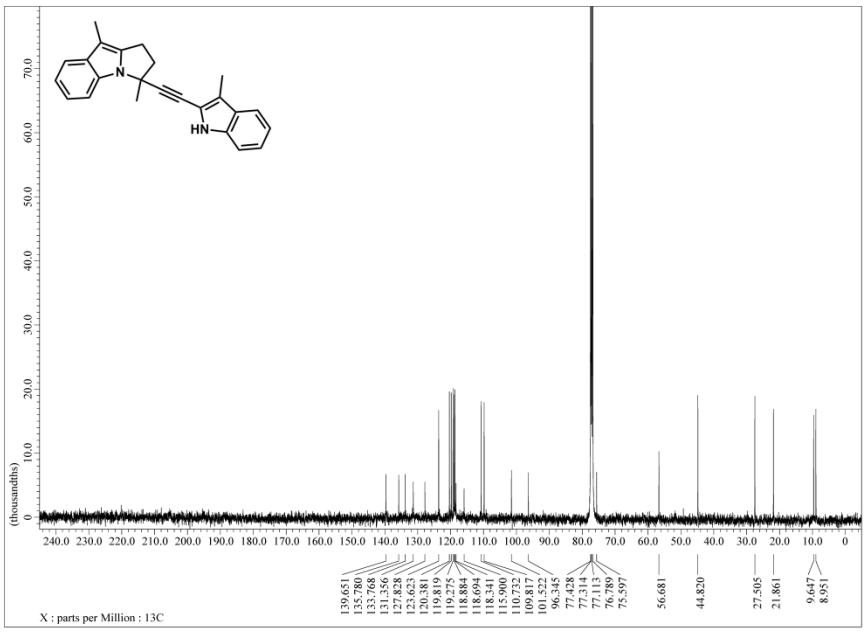


Figure A.2.30 ^{13}C NMR ($CDCl_3$) of compound **99**

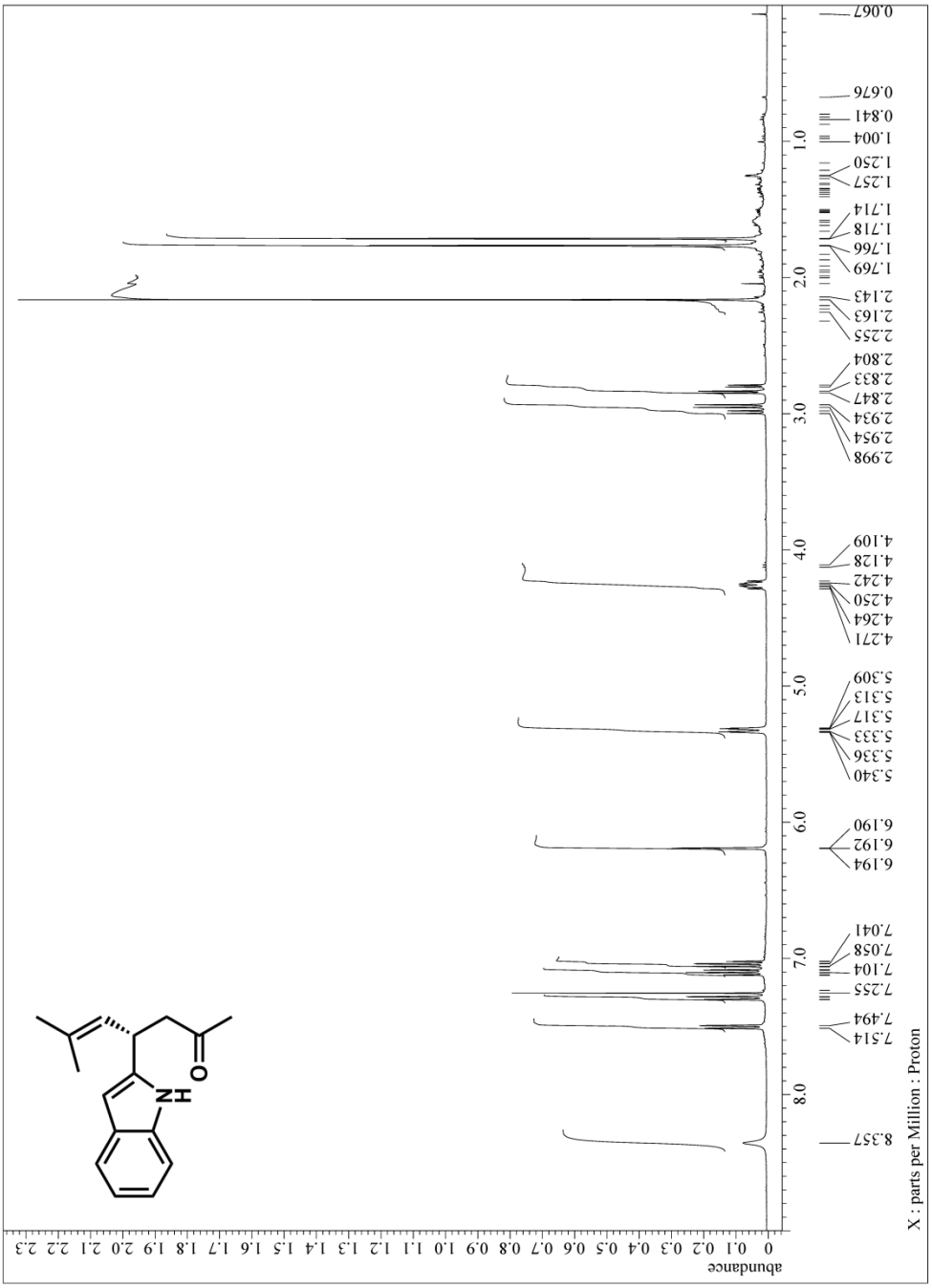


Figure A.2.31 ^{1}H NMR ($CDCl_3$) of compound **121**

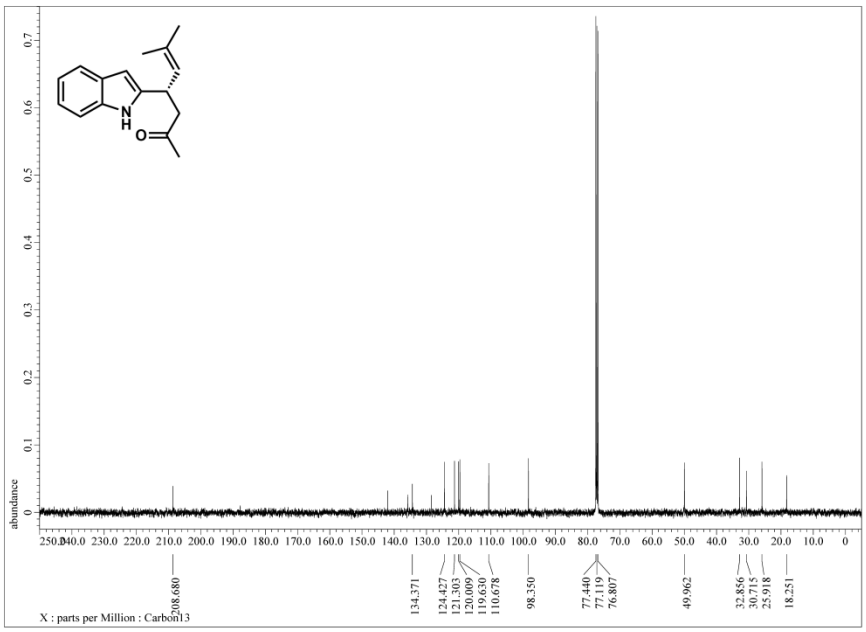


Figure A.2.32 ^{13}C NMR ($CDCl_3$) of compound **121**

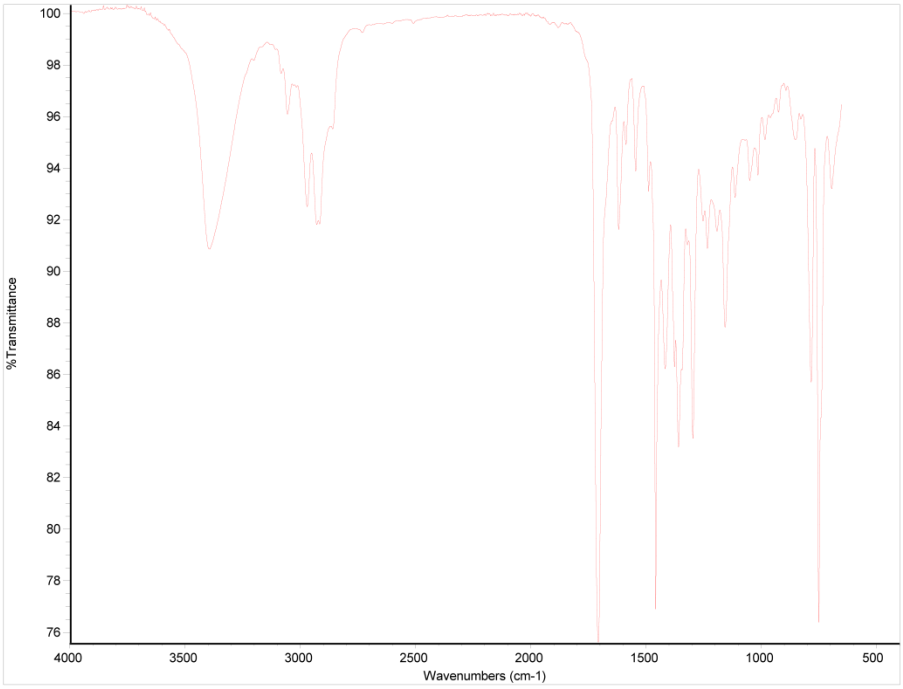
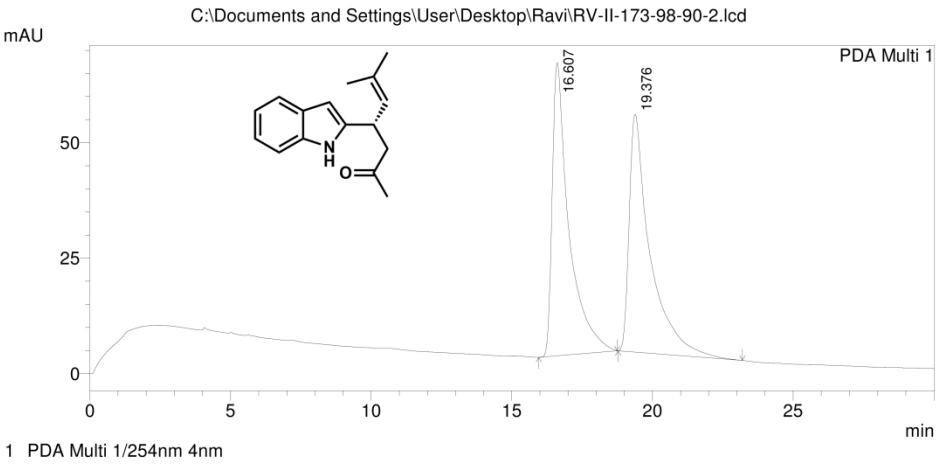


Figure A.2.33 IR spectrum of compound **121**

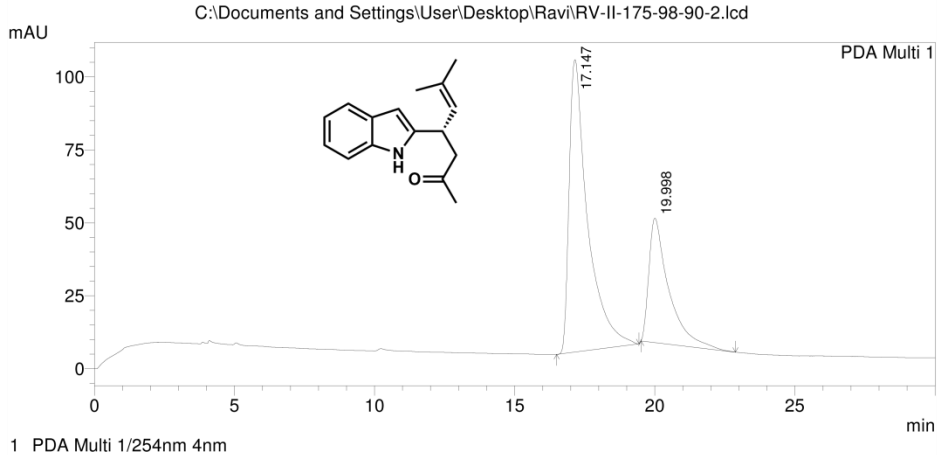


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	16.607	2624433	63442	49.766	55.183
2	19.376	2649110	51524	50.234	44.817
Total		5273543	114967	100.000	100.000

Fig A.2.34 HPLC trace for compound **121** (entry 7 Table 4.3)

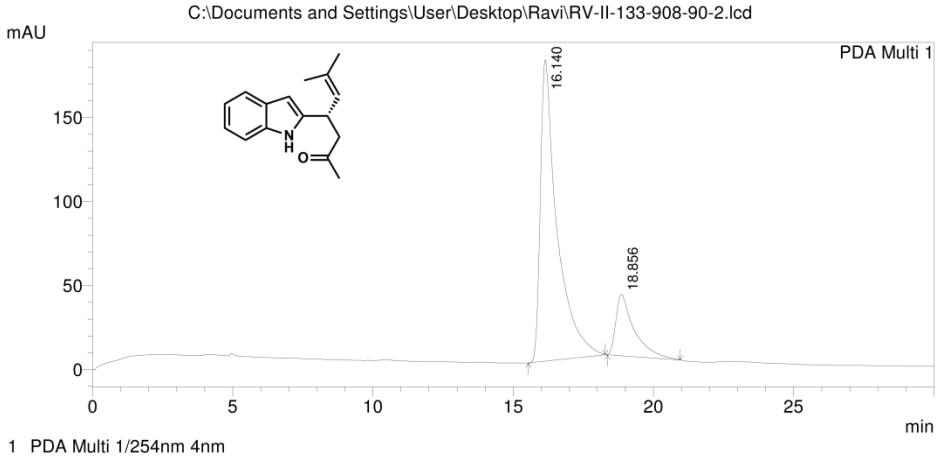


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	17.147	4590191	100216	68.590	70.140
2	19.998	2101987	42663	31.410	29.860
Total		6692178	142879	100.000	100.000

Fig A.2.35 HPLC trace for compound **121** (entry 2 Table 4.3)

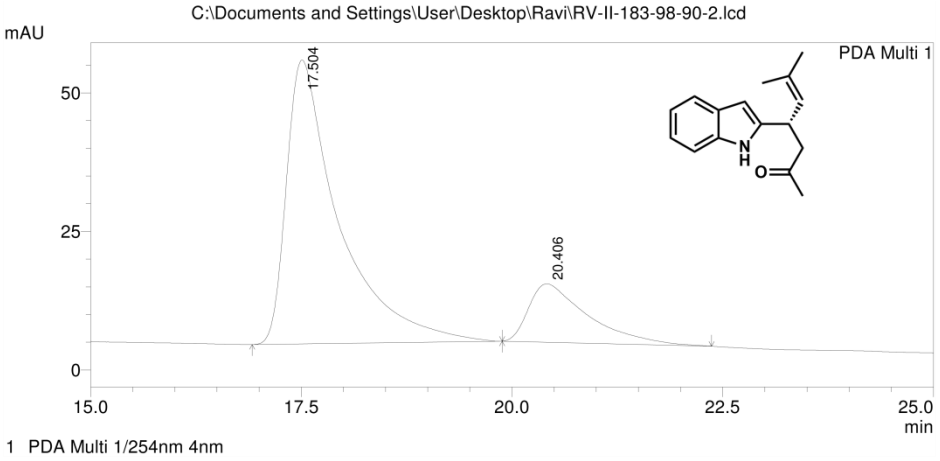


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	16.140	7442270	179461	81.923	83.101
2	18.856	1642236	36495	18.077	16.899
Total		9084506	215956	100.000	100.000

Fig A.2.36 HPLC trace for compound **121** (entry 3 Table 4.3)

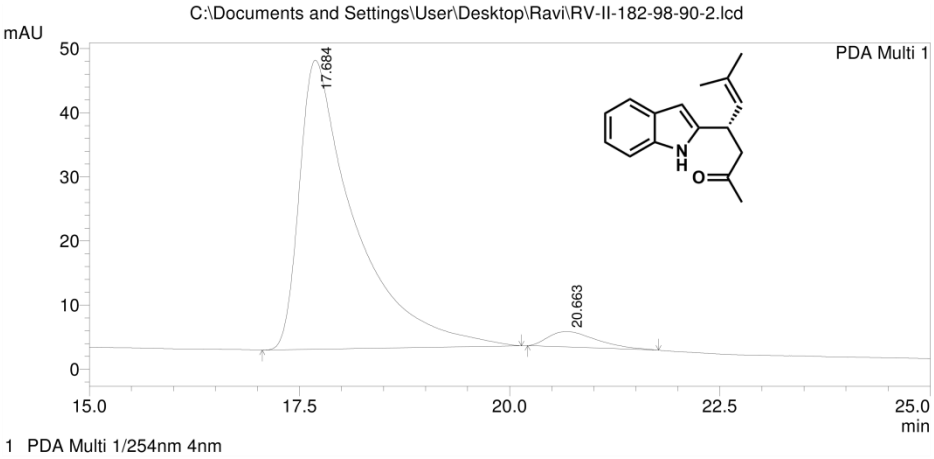


PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	17.504	2212672	51198	81.213	82.929
2	20.406	511851	10539	18.787	17.071
Total		2724522	61737	100.000	100.000

Fig A.2.37 HPLC trace for compound **121** (entry 5 Table 4.3)



PeakTable

PDA Ch1 254nm 4nm

Peak#	Ret. Time	Area	Height	Area %	Height %
1	17.684	2071616	45078	95.542	94.979
2	20.663	96670	2383	4.458	5.021
Total		2168286	47461	100.000	100.000

Fig A.2.38 HPLC trace for compound **121** (entry 6 Table 4.3)