UNIVERSITY OF CALIFORNIA,
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Assignment of Configuration Using Kinetic Resolution Reagents

and

Total Synthesis of (+)-Fastigiatine

DISSERTATION

submitted in partial satisfaction of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

in Chemistry

by

Renzo Alexander Samamé

Dissertation Committee:
Professor Scott D. Rychnovsky, Chair
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2015

DEDICATION

To my mother and sister

&

*my wife Katrina and the kid*s

for their support

"A bit of Science distances one from God, but much science nears one to Him...
The more I study nature, the more I stand amazed at the work of the Creator."

– Louis Pasteur

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LIST OF ABBREVIATIONS

Å	Angstroms
Ac	Acetyl
Atm	Atmosphere
ax	Axial
Bn	Benzyl
Boc	*tert*-butoxycarbonyl
Bp	Boiling point
Bu	Butyl
°C	Degree Celsius
cat.	Catalytic
CSA	Camphorsulfonic acid
cis	L., on the same side
d	day(s)
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
δ	Chemical shift
DIBAL-H	Diisobutylaluminum hydride
DMAP	4-Dimethylaminopyridine
DMF	*N,N*-Dimethylformamide
DMPU	1,3-dimethyl-3,4,5,6-tetrahydro-2(1H)-pyrimidinone
DMSO	dimethyl sulfoxide
dppf	1,1'-bis(diphenylphosphino)ferrocene
dr	Diastereomeric ratio

ee	Enantiomeric excess
er	Enantiomeric ratio
eq	Equatorial
Eq.	Equation
equiv	Equivalents
ESI	Electrospray ionization
Et	Ethyl
GC	Gas chromatography
h	hour(s)
HMPA	*N, N, N', N', N'', N''*-hexamethylphosphoramide
HRMS	High resolution mass spectrometry
Hz	Hertz
IR	Infrared spectrometry
J	Coupling constant
LAH	Lithium aluminium hydride
LiDBB	Lithium di-*tert*-butylbiphenylide
LiN	Lithium naphthalenide
LDA	Lithium diisopropylamide
µ	micro
m	milli
M	Molar
m-CPBA	3-Chloroperoxybenzoic acid
min	minute(s)

Me	Methyl
MPLC	Medium pressure liquid chromatography
MHz	Megahertz
Ms	methanesulfonyl
NMR	Nuclear magnetic resonance
Ph	Phenyl
ppm	parts per million
rt	Room temperature
sec	secondary
t	*tert*
TBAF	Tetra-n-butylammonium fluoride
TBS	*tert*-butyldimethylsilyl
TES	triethylsilyl
Tf	trifluoromethanesulfonyl
TFA	trifluoroacetic acid
THF	Tetrahydrofuran
THP	Tetrahydropyran
TIPS	Triisopropylsilyl
TLC	Thin layer chromatography
TMS	Trimethylsilyl
trans	L., across
Ts	4-Toluenesulfonyl
TsOH	4-Toluenesulfonic acid

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Many positive events have happened to me since my arrival to Irvine and this document is one of them. Of course, none of these lines would have been written without the support of family and friends. I would like to start by thanking my mother and sister who were always there for me whenever I needed them.

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Publications

"The Lycopodium Alkaloids: Total Synthesis of (+)–Fastigiatine." (Manuscrip in Preparation)

Samame, R. A.; Owens, C. M.; Rychnovsky, D. R. "Concise Synthesis of (+)–Fastigiatine." *Chem. Sci*, **2015**, DOI: 10.1039/c5sc03262h.

Miller, S. M; Samame, R. A.; Rychnovsky, D. R. "Nanomole-scale Assignment of Configuration for Primary Amines Using a Kinetic Resolution Strategy." *J. Am. Chem. Soc.* **2012**, *134*, 20318–20321.

ABSTRACT OF THE DISSERTATION

Assignment of Configuration Using Kinetic Resolution Reagents

and

Total Synthesis of (+)-Fastigiatine

By

Renzo Alexander Samamé

Doctor of Philosophy in Chemistry

University of California, Irvine, 2015

Professor Scott Douglas Rychnovsky, Chair

The first part of this thesis illustrates the application of kinetic resolution reagents for the determination of absolute configuration. A dual-catalytic approach based on ion pair recognition was explored using the combined action of 4-(*N*,*N*-dimethylamino)pyridine (DMAP) and a chiral thiourea receptor co-catalyst. After difficulties were encountered with a dual-catalytic mode, an alternative approach using enantioselective acyl transfer reagents was investigated. The new strategy led to a successful development of a new and efficient method to rapidly establish the absolute configuration of primary amines using mass spectrometry.

Part two of this thesis describes the development of a modular approach toward the synthesis of the *Lycopodium* alkaloids. A highly concise six-step total synthesis of the complex alkaloid (+)-fastigiatine was accomplished using a transannular Mannich reaction that generated two quaternary carbons at a late stage. The modular approach to fastigiatine will be expanded to other members of the family including himeradine A and lyconadin A.

Chapter 1

Investigation of a Catalytic Approach to Determine Absolute Configuration

I. Introduction: Determining the relative and absolute configuration of organic molecules is a critical aspect in the synthesis and isolation of organic compounds.[1] Several methods have been developed to determine the absolute configuration of molecules including the circular dichroism exciton method,[2] optical rotary dispersion,[3] Kishi's NMR spectroscopy method,[4] Horeau's method,[5] the Mosher analysis,[6] and X-ray crystallographic analysis.[7]

The Mosher analysis is a common technique used by chemists to determine the configuration of secondary alcohols and primary amines.[8] With the Mosher analysis, the optically pure amine in question is derivatized to its 2-methoxy-2-phenyl-2-trifluoromethylacetyl (MTPA) amide with the treatment of (*R*) and (*S*) MTPA acid and the absolute configuration is established by measuring the chemical shift differences (Δδ's) of the resultant diastereomers via ^{1}H NMR spectroscopy.

With X-ray crystallography, beams of X-ray are shot to a crystalline sample that generates a diffraction pattern. By measuring the intensities and angles of these diffraction patterns one can establish an electron density map. The substance in question is then correlated to fit the map in order to identify the absolute configuration. While these analyses are reliable, each has their limitation. The Mosher method requires significant material and time to conduct the derivatization, purification and spectroscopic measurements. X-ray crystallography requires the analyte to exist in its crystalline form with appropriate particle size and quality, which could take hours to months to grow. Because these analyses can be difficult and time-consuming processes, the development of a new method that could overcome such issues would be ideal.

II. Introduction to the Competing Enantioselective Conversion (CEC) Method:

A new strategy that uses kinetic resolution catalysts to determine the absolute configuration of secondary alcohols was developed in our laboratory and was named the Competing Enantioselective Conversion (CEC) Method.[9] Like kinetic resolution, this method relies on the difference in reaction rates between an enantioenriched alcohol **1.1** and enantiomers of a chiral catalyst **1.2** and **1.3** (Figure 1.1). The relative rate of the fast-reacting catalyst as observed in ^{1}H NMR spectroscopy determines the absolute configuration of the enantioenriched alcohol. This new method facilitated routine assignment of chiral secondary alcohols using only a few milligrams (1-3 mg) of the alcohol without any purification or isolation of its derivative.

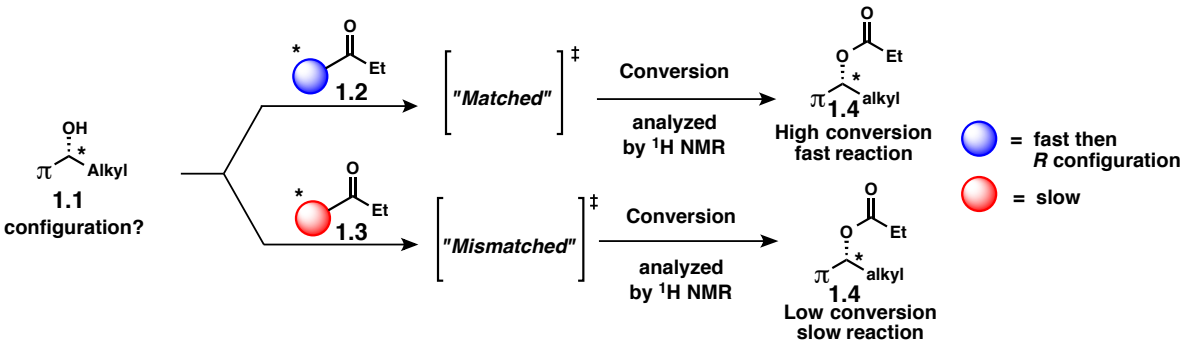


Figure 1.1 Representative scheme of the CEC method.

In theory, this technology can be extended to any functional group for which a kinetic resolution catalyst has been developed. Another class of functional groups of interest to the synthetic community are amines. We sought to extend this method for determining absolute configuration to primary amines, which seemed like a logical extension and since amines are one the most common functional groups in natural products and pharmaceuticals.[10] One concern associated with the amines substrates is their high reactivity, which often results in high background reactions rates. In recognition of this, we opted to investigate a dual catalytic approach for our method development. Seidel developed a dual catalytic system with DMAP and

chiral thiourea for the kinetic resolution of benzylic amines[11], propargylic amines[12] and allylic amines.[13] We envisioned using Seidel's catalyst to develop a mnemonic for assigning the absolute configuration to primary amines as shown in Figure 1.2.

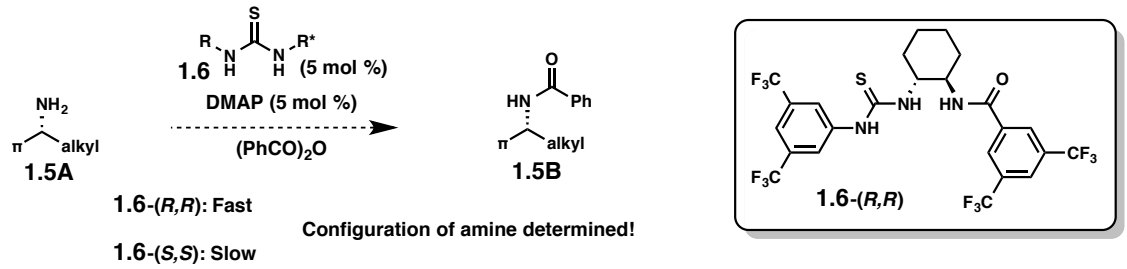
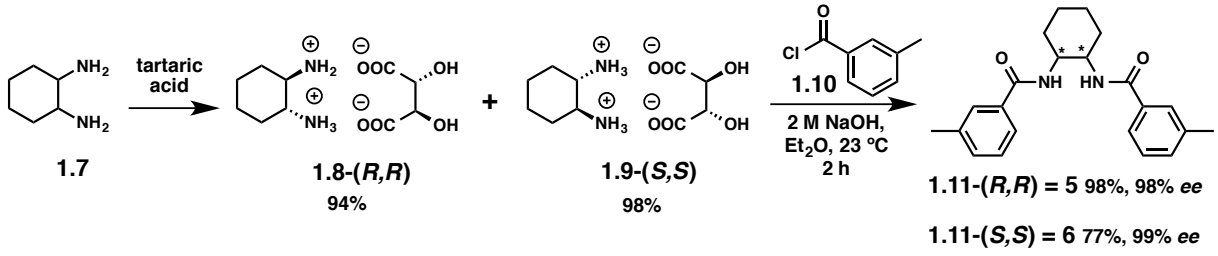


Figure 1.2. Proposed determination of the absolute configuration of primary amines.[14]

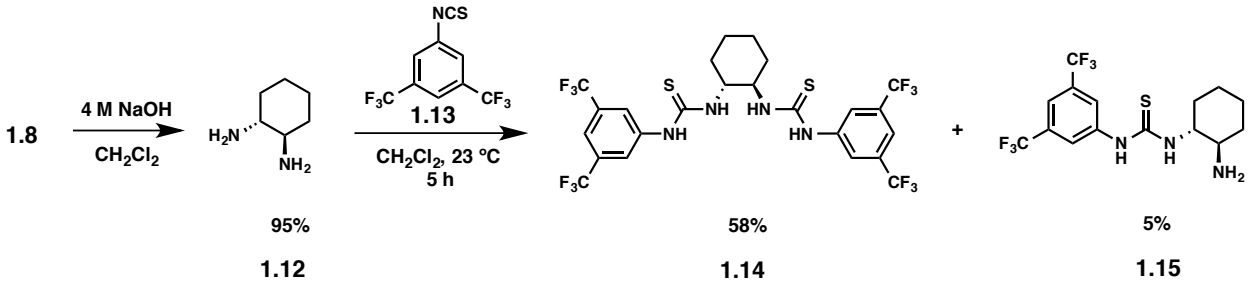
III. Results and Discussion: The first part of the project was to synthesize Seidel's catalyst (**1.6**). Some of the steps were modified to use the least expensive starting materials (**1.7** and **1.17**). The synthesis began with commercially available (±)-1,2-cyclohexanediamine (**1.7**), which was resolved using (*L*) and (*D*) tartaric acid to afford enantioenriched cyclohexanediamine tartrate salts **1.8** and **1.9** (Scheme 1).[15] In order to determine the enantiomeric excess of the resolved amines, tartrate salts **1.8** and **1.9** were derivatized using *m*-toluoyl chloride in the presence of sodium hydroxide to provide bisamides **1.11**-(*R,R*) and **1.11**-(*S,S*). HPLC analysis of **1.11**-(*R,R*) and **1.11**-(*S,S*) indicated 98% ee and >99% ee respectively.[16]

Scheme 1.1. Kinetic resolution of *trans*-(±)-1,2-cyclohexanediamine.



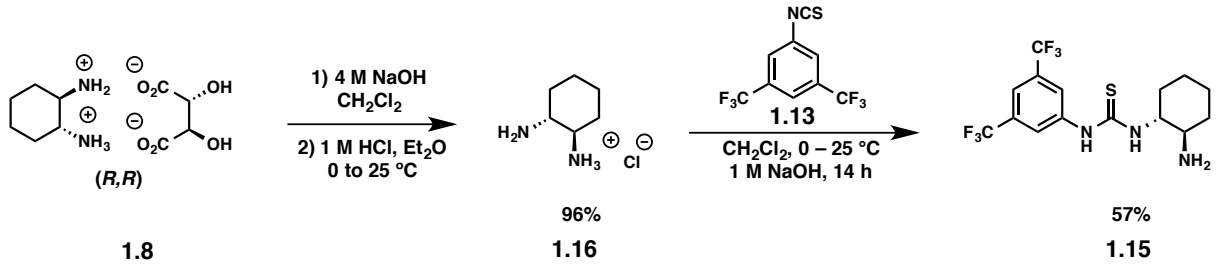
With each enantioenriched diamine salt in hand, the salts were converted to the free amines under basic conditions (Scheme 1.2). However, due to sublimation and solubility issues, substantial loss of the diamine occurred during early attempts of isolation. Dissolution in a strong solution of sodium hydroxide, followed by rapid extraction using methylene chloride and evaporation at 0 °C *in-vacuo* afforded over 95% yield of the diamine **1.12**.

Scheme 1.2. Synthesis of disubstituted thiourea **1.14** and monothiourea **1.15**.



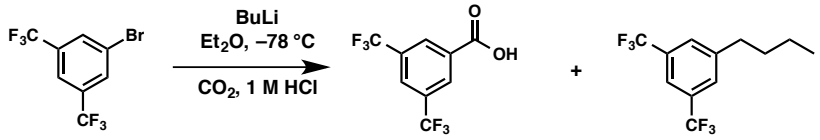
Monofunctionalization of the enantioenriched cyclohexanediamine was attempted to access cost-efficient and multigram scale production of the catalyst, but undesired disubstituted aminothiourea **1.14** was formed as the major product despite the slow addition of the isothiocyanate (Scheme 1.2). The free diamine **1.12** turned into an oil within hours of being generated, making its stoichiometric handling difficult. For easier handling, the mono-hydrochloride diamine salt **1.16** was made using 1 equivalent of HCl in diethyl ether at 0 °C (Scheme 1.3).[17] Slow addition of 3,5-bis(trifluoromethyl)isothiocyanate at low concentration to the mono-hydrochloride diamine salt **1.16** gave the desired mono substituted aminothiourea **1.15** in a 57% yield (Scheme 1.3).

Scheme 1.3. Synthesis of monosubstituted aminothiourea **1.15**.



To install the amide moiety of Seidel's catalyst, NHS-coupling reactant **1.21** was prepared by first carboxylating 1-bromo-3,5-bis(trifluoromethyl)benzene **1.17** with butyllithium to provide 3,5-bis (trifluoromethyl)benzoic acid **1.18**. The use of *n*-butyllithium produced **1.18** in 23% yield, along with with 1-butyl-3,5- bis(trifluoromethyl)benzene **1.19** as a side product in a 51% yield. Replacement of *n*-butyllithium with *sec*-butyllithium provided 70–79% yield of the acid **1.18** on up to a 10-gram scale (Table 1.1).

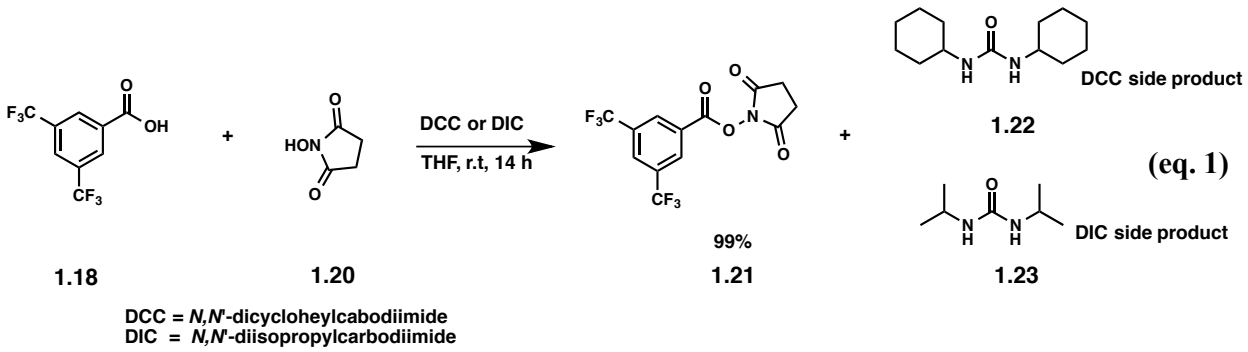
Table 1.1. Carboxylation of 1-bromo-3,5-bis(trifluoromethyl)benzene **1.17**.



1.17 **1.18** **1.19**

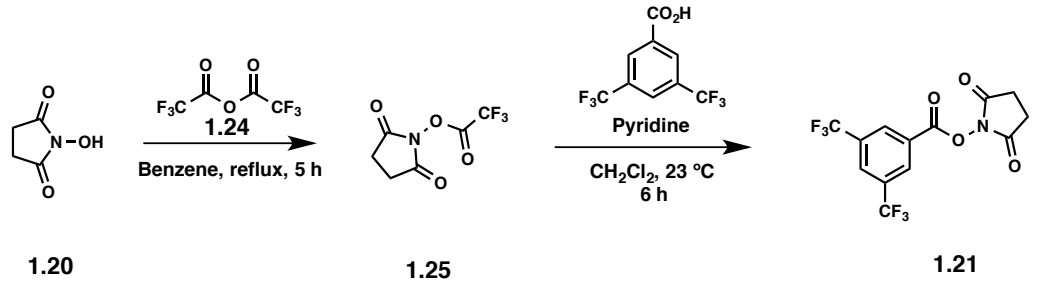
entry	base	**1.18** % yield	**1.19** % yield
1	n-BuLi	23	51
2	sec-BuLi	79	0

Coupling of acid **1.18** to *N*-hydroxylsuccinimide **1.20** using either *N,N'*-dicycloheylcabodiimide (DCC) or *N,N'*-diisopropylcarbodiimide (DIC) provided the desired NHS-ester **1.21**.[18] However, purification proved to be problematic due to the formation of urea by-products **1.22** and **1.23** respectively (eq. 1).

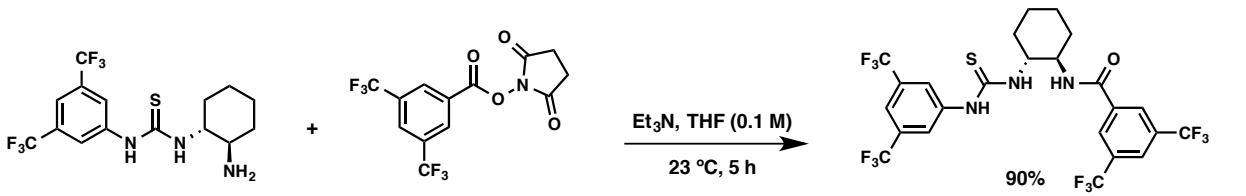


To circumvent this issue, an alternative method to couple the benzoic acid that employed an NHS-TFA adduct **1.25** was accessed by reacting *N*-hydroxylsuccinimide **1.20** with trifluoroacetic anhydride.[19] NHS-ester **1.21** was observed by TLC chromatography, however extensive hydrolysis was observed during flash chromatography purification. To minimize decomposition, when the reaction mixture was completed as observed by TLC, quick soft acid-base extraction followed by evaporation afforded NHS-ester product **1.21** in quantitative yield (Scheme 1.4).

Scheme 1.4. NHS-ester bond formation **1.21**.

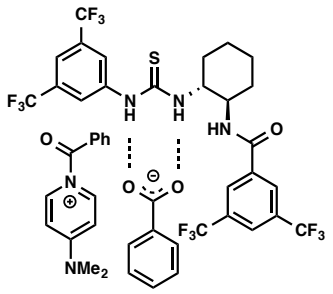


Addition of NHS-ester **1.21** to aminothiourea **1.15** under basic conditions afforded the desired catalyst **1.6-**(*R,R*) in a 90-95% yield (eq. 2). Its enantiomer **1.6**-(*S,S*) was obtained in a similar 90-94% yield. The catalysts were synthesized in 4 linear steps using a convergent synthesis from inexpensive starting materials in a 37-40% overall yield.



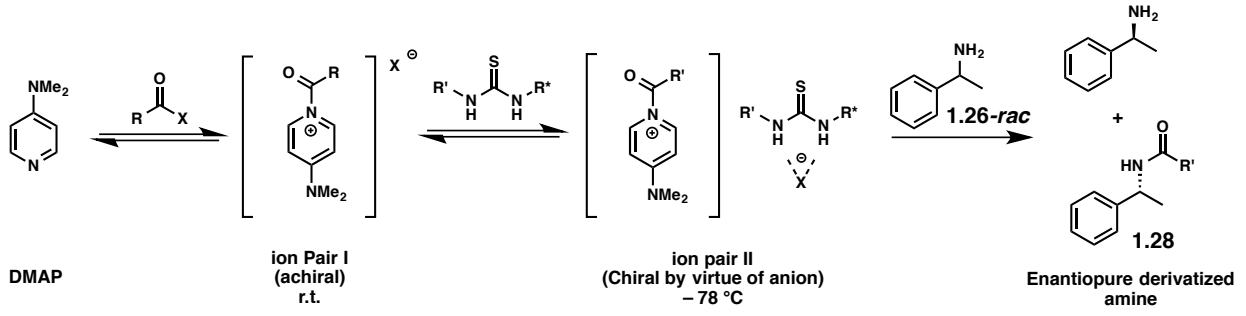
1.15 **1.21** **1.6-(*R, R*)** **(eq. 2)**

Kinetic Resolution: Before testing the new method for determining configuration, catalyst **1.6** was used to attempt to reproduce Seidel's reported kinetic resolution conditions. In this catalytic system, a simple achiral acylpyridinium salt (ion pair **I**) formed in situ from a benzoic anhydride and DMAP is rendered chiral upon binding of the associated anion to thiourea **1** to form ion pair **II** (Scheme 1.5). Ion pair **II** directs acylation to one enantiomer of the racemic amine, providing an efficient kinetic resolution. It is worth noting that although Seidel reported the chiral ion pair intermediate **II**, it was not reported how ion pair **II** induced chiral information on the acylation reaction when our investigations were conducted. Seidel reported selectivity factors in the range of 12-56 for benzylic amines, propargylic amines and allylic amines.[11–13] Initial validation studies using (±)-phenylethylamine **1.26** and (±)-1-(1-naphthyl)ethylamine **1.27** failed to give the reported selectivities, presumably due to insufficient formation of ion pair **II** prior to the addition of the amine or under the reaction conditions.



ion pair II

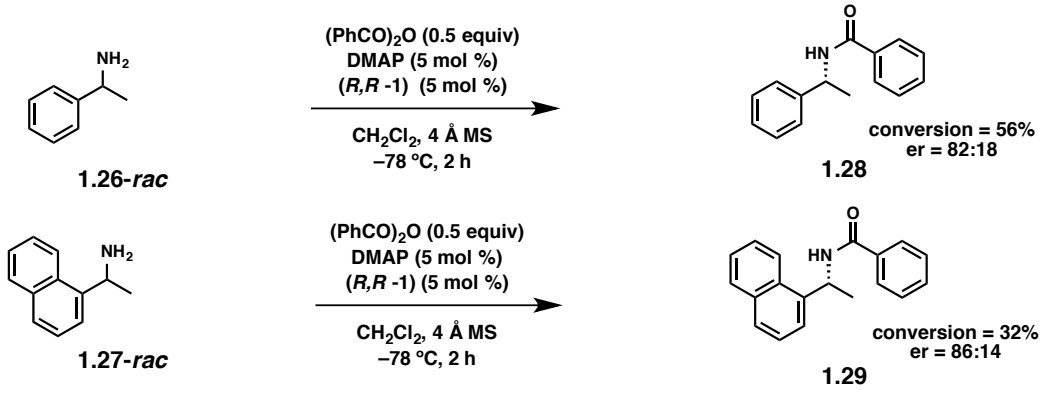
Scheme 1.5. Seidel's dual catalytic approach to the kinetic resolution of amines.



DMAP | ion Pair I (achiral) r.t. | ion pair II (Chiral by virtue of anion) – 78 °C | Enantiopure derivatized amine

To ensure ion pair **II** formation, chiral thiourea **1.6** catalyst was added to the acylpyridinium salt (Ion pair **I**). The temperature was lowered to –78 ºC and stirred for a longer period of 25 minutes, followed by addition of the amine. Seidel's procedure employed a methylmagnesium chloride (MeMgCl) addition to quench the remaining anhydride, however the side products generated from this method could not be fully separated by column chromatography. A methanolic ammonia quench reduced the formation of side products and allowed for the purification of the enantioenriched amide products. Control experiments using either MeMgCl or NH_3/MeOH at fixed periods of time showed that both are effective quenching reagents.

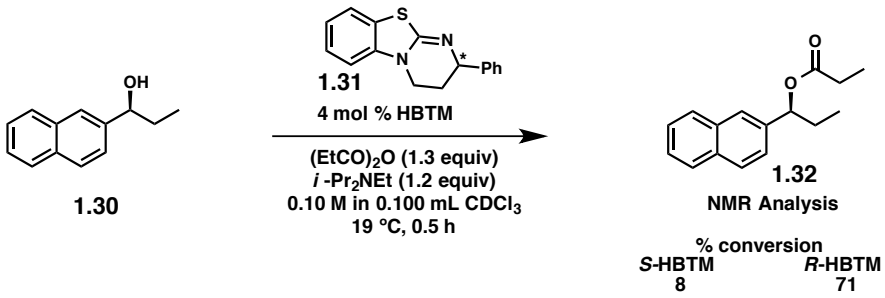
Scheme 1.6. Kinetic resolution of **1.26-*rac*** and **1.27-*rac***.



Kinetic resolution of substrates **1.26** and **1.27** afforded **1.28-**(*R*) and **1.29**-(*R*) in 56% and 32% yield, respectively. Chiral HPLC analysis of the resolved products **1.28** and **1.29** were in agreement with the literature values.[11]

Method development: In our previous studies determining the absolute configuration of secondary alcohols, the absolute configuration of 1-(naphthalen-2-yl)propan-1-ol **1.30** was determined using Birman's kinetic resolution catalysts **1.31**.[20] Birman's (*R)*-homobenzotetramisole (*R*-HBTM) was the fast-reacting catalyst confirming the absolute configuration of the alcohol in question (Table 1.2).[9]

Table 1.2. Configuration assignment of (*S*)-1-(naphthalen-2-yl)propan-1-ol **1.30**.



In order to demonstrate proof of concept with primary amines, three parallel reactions using optically pure (*R*)-phenylethylamine **1.26**-(*R*) with different catalysts were conducted. In principle, the fast reacting catalysts **1.6**-(*R,R*) would provide a matched scenario that would lead to a fast reaction (Figure 1.3). Alternatively, the slow-reacting catalyst **1.6**-(*S,S*) would be a mismatched case and lead to slow reactivity. Lastly, the reaction of **1.26**-(*R*) with DMAP in the absence of catalyst would result in even slower conversion to amide **1.28**.

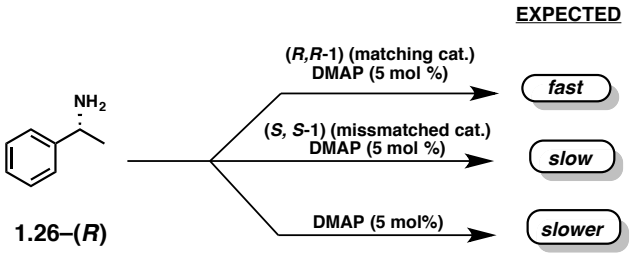


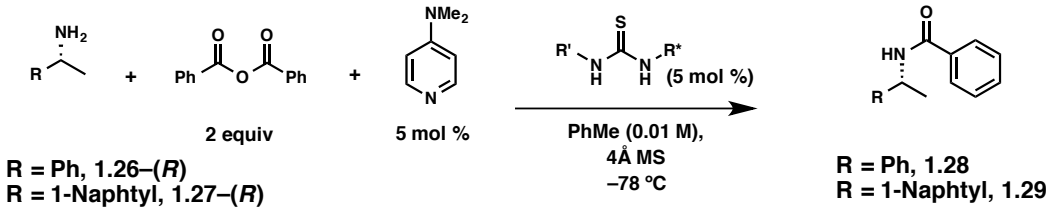
Figure 1.3. Expected rates of acylation of amine **1.26**.

After 1 hour, the following yields were observed: **1.6**-(*R,R*), **1.6**-(*S,S*), and no chiral catalyst provided amide **1.28** in 83%, 66% and 89% yields, respectively (Table 1.3, entry 1). While a noticeable difference in conversion with the **1.6**-(*R,R*) and **1.6**-(*S,S*) catalysts occurred, the achiral reaction proceeded the fastest of the three reactions. Reaction times were reduced in search of the expected rate conversions.

However, only minimal difference, in yields between **1.6**-(*R,R*), **1.6**-(*S,S*) and DMAP alone were observed (Table 1.3, entries 2 and 3). Since DMAP alone catalyzed the reaction faster

than with catalyst **1.6**, the DMAP equivalent was reduced from 5 mol% to 1 mol%. Surprisingly, similar conversions were observed for these reactions (Table 1.3, entry 4). In order to confirm these results, optically pure (*R*)-1-(1-naphthyl)ethylamine **1.27**, which showed higher selectivity in kinetic resolution, was examined.[11–12]

Table 1.3. Studies on the acylation of **1.26** and **1.27**.



Entry	Amine	Time (h)	Conversion (%)		
			(*R,R*-**1**)	(*S,S*-**1**)	DMAP alone
1	1.26	1.0	83	66	89
2	1.26	0.5	62	57	46
3	1.26	0.25	60	57	38
4	1.26	1.0	61	67	52
5	1.27	1.0	37	32	13
6	1.27	1.0	36	34	25

[a] Percent conversions were determined by ^{1}H NMR analysis of the crude mixture using 4,4'-di-*tert*-butylbiphenyl (DBB) as an internal standard. [b] DMAP equivalent was reduced to 1 mol %. [c] Benzoic anhydride was reduced to 1 equiv.

Using naphthyl amine **1.27**-(*R*), three parallel reactions were investigated to understand the role of benzoic anhydride. One equivalent of benzoic anhydride using **1.6**-(*R,R*) afforded 37% conversion (Table 1.3, entry 5). The **1.6**-(*S,S*) catalyzed reaction led to 32% conversion while DMAP alone provided 13% conversion. Two equivalents of benzoic anhydride using the **1.6**-(*R,R*) catalyst gave 36% conversion, the **1.6**-(*S,S*) catalyzed reaction yielded 34% conversion, and DMAP alone gave 25% conversion (Table 1.3, entry 6). The absence of a distinct rate difference using each catalyst was again observed using naphthyl amine **1.27**.

IV. Conclusions: Although we were able to reproduce Seidel's kinetic resolution of primary amines, an adaptation of the system to assign absolute configuration was not successful. When this project was conducted, the mechanism for Seidel's dual-catalytic system had not been reported, which made elucidating the difference in reactivity of racemic amines versus enantiopure amines challenging.[28] A rationale for the lack of selectivity can be explained by the stoichiometric use of anhydride. During our previous studies in the determination of absolute configuration of secondary alcohols, the use of two equivalents of anhydride proved to be ideal for the method development. However, in the case of amine moieties, two equivalents may have led to significant background reactions.

Amines are notorious for their high levels of reactivity with simple acylating reagents (e.g., anhydrides); this reactivity obviates the need of a catalyst additive (e.g., DMAP) for the installation of an acyl group. In Seidel's catalytic system, primary amines have the opportunity to react with three acylating sources: 1) benzoic anhydride, 2) acylpiridinium salt **I** and 3) ion pair **II**. It is possible that reducing the amounts of anhydride may have resulted in more pronounced rate differences for the reaction when using opposite enantiomers of the catalyst. However, the conditions of the system were not ideal (e.g., low temperatures, low concentrations and molecular sieves addition) for the development of a simple and straightforward method. Due to complications with the dual-catalytic system described in this chapter, we decided to investigate an alternative kinetic resolution system. The details on the development and successful application of kinetic resolution reagents to assign absolute configuration are described in the upcoming chapter of this thesis.

Supporting Information

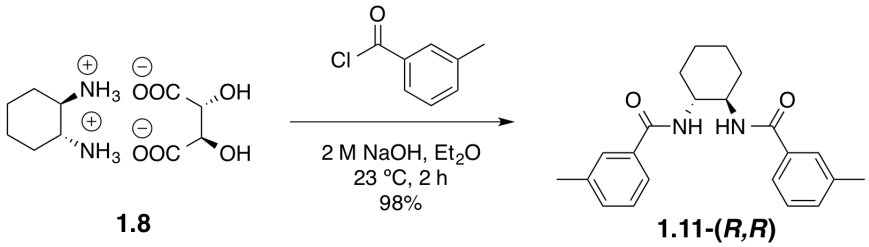
I. General Experimental Details

All reactions were performed under an atmosphere of argon unless stated otherwise. All glassware was oven- or flame-dried and cooled under an inert atmosphere of argon unless stated otherwise. All commercially available reagents were used as received except the following: tetrahydrofuran, dichloromethane, toluene, and diethyl ether were degassed with argon and dried by vacuum filtration through activated alumina according to the procedure by Grubbs.[21] Triethylamine was distilled from CaH_2 and 4-dimethylaminopyridine was recrystallized from distilled toluene prior to use. Benzoic anhydride was washed with Na_2CO_3 and extracted in dichloromethane prior to use. Molarities of organolithium reagents were determined by titration.[22] Methylmagnesium chloride was titrated with I_2 according to the procedure described by Krasovskiy and Knochel.[23] Thin-layer chromatography (TLC) was performed on Whatman 250 μm layer 6 Å glass-baked silica gel plates or Merck 250 μm layer 6 Å glass-backed neutral aluminum oxide plates. Eluted plates were visualized using UV light, iodine, vanillin, *p*-anisaldehyde, Dragendorff's reagent or potassium permanganate stains. Silica gel chromatography was performed according to the method by Still, Khan and Mitra.[24]

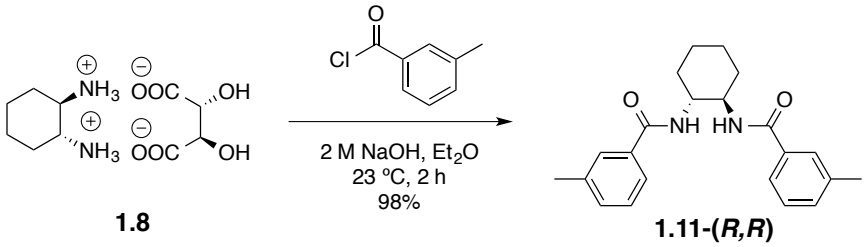
II. Instrumentation

Infrared spectra were recorded on a MIDAC Prospect FT-IR spectrometer. Optical rotations were measured on a JASCO DIP-370 digital polarimeter. ^1H and ^{13}C NMR spectra were recorded at 500 and 125 MHz, respectively. ^1H NMR spectra were reported in ppm on the δ scale and referenced to tetramethylsilane (0.00 ppm) or residual solvent signal ($CDCl_3$ at 7.26 ppm, CD_2Cl_2 at 5.32 ppm). The data are presented as follows: chemical shift, multiplicity (s =

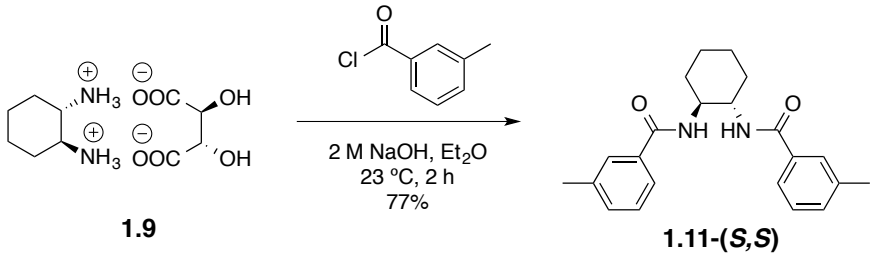
singlet, d = doublet, t = triplet, q = quartet, quint = quintet, m = multiplet, br = broad, app = apparent), coupling constant(s) in Hertz (Hz), and integration. ^{13}C NMR spectra were reported in ppm relative to $CDCl_3$ (77.07 ppm) or CD_2Cl_2 (53.80). Unless otherwise stated, NMR spectra were collected at 25 ºC. Melting points were obtained using an electrothermal melting point appartatus and are uncorrected. Enantioselectivities were determined using an analytical HPLC instrument with Daicel™ chiralpack® column. High resolution mass spectrometry was peformed by the University of California, Irvine Mass Spectrometry Center.



***N*,*N*'-((1*R*,2*R*)-Cyclohexane-1,2-diyl)bis(3-methylbenzamide) (1.11).** To a solution of tartrate diamine salt **1.8**-(*R*,*R*) (0.200 g, 0.757 mmol) in Et_2O (10 mL) was added a NaOH (2 M) solution (6 mL) at room temperature and stirred until the resultant biphasic mixture was clear. *m*–Toluoyl chloride (1.29 mL, 1.51 mmol) was added dropwise via syringe. Upon addition of the acid chloride, a precipitate formed. The resulting mixture was allowed to stir for 2 hours. The reaction was extracted with CH_2Cl_2 (3 x 10 mL). The combined organic extracts were dried over Na_2SO_4 and concentrated *in vacuo* to provide a white solid. The solid was purified via column chromatography (30:70 EtOAc/Hex), and the pure product was obtained in 98% yield (260 mg). **mp** = 197–198 ºC; R_f = 0.25 (30:70 EtOAc/Hex); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 7.53 (s, 2H), 7.50 (app d, *J* = 7.0 Hz, 2H), 7.23–7.19 (m, 4H), 6.86 (app d, *J* = 5.0 Hz, 2H), 4.02 (s, 2H), 2.31 (s, 6H), 2.21 (app d, *J* = 8.5 Hz, 2H), 1.84 (app s, 2H), 1.42 (app d, *J* = 6.5 Hz, 4H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 168.6, 138.5, 134.4, 132.4, 128.6, 127.9, 124.1, 54.7, 32.6, 25.0, 21.5; **IR** (thin film) 3309, 3082, 2927, 2858, 1631, 1604, 1585 cm^{-1}; **HRMS** (ESI/methanol) *m / z* calcd for $C_{22}H_{26}N_2O_2Na$ $(M + Na)^+$ 373.1892, found 373.1892; **HPLC**: Daicel chiralpak AD-H, *i*-PrOH/*n*-hexane=10/90, flowrate = 1mL/min, UV = 254 nm, t_R= 7.0 min (minor) and t_R= 17.2 min (major), 98 % *ee*. Spectral data were consistent with those previously reported.[25]

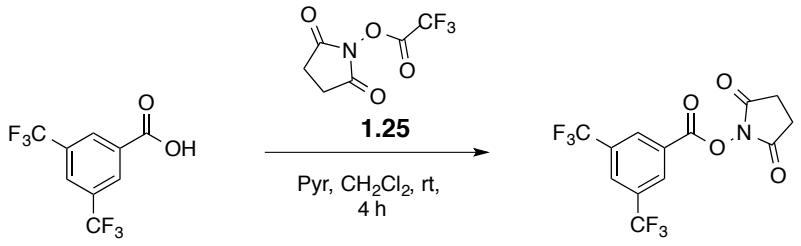


***N*,*N*'-((1*S*,2*S*)-Cyclohexane-1,2-diyl)bis(3-methylbenzamide) (6).** Following the procedure for compound **1.8**-(*R*,*R*) the product was obtained in 77% yield (189 mg). **mp** = 200–202 ºC; $\mathbf{R}_f$ = 0.25 (30:70 EtOAc/Hex); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 7.54 (s, 2H), 7.49 (app d, *J* = 7.0 Hz, 2H), 7.26–7.19 (m, 4H), 6.79 (app s, 2H), 4.01 (s, 2H), 2.32 (s, 6H), 2.22 (app d, *J* = 6.0 Hz, 2H), 1.84 (app s, 2H), 1.43 (app s, 4H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 168.6, 138.5, 134.4, 132.4, 128.6, 127.9, 124.1, 54.7, 32.7, 25.0, 21.5; **HPLC**: Daicel chiralpak AD-H, *i*PrOH/*n*-hexane=10/90, flowrate = 1mL/min, UV = 254 nm, t_R= 7.0 min (major) and 17.3 (minor), >99% *ee*. Spectral data were consistent with those previously reported of compound **6** enantiomer.[5]



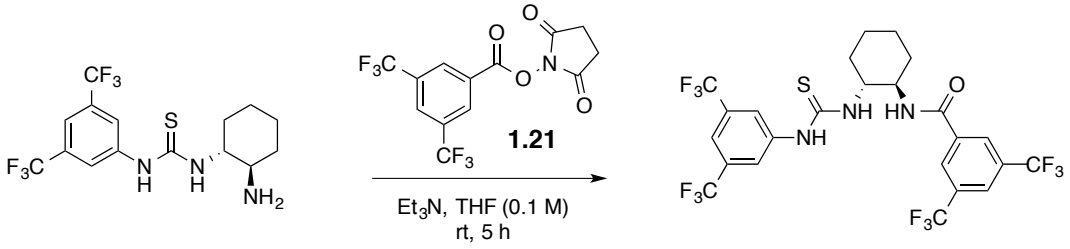
3,5-Bis(trifluoromethyl)benzoic acid (1.18). Anhydrous diethyl ether (31 mL) was added to a solution of *sec*–BuLi (0.98 M) so that the final molarity was 0.5 M. The resultant solution was cooled to –78 ºC and 1-bromo-3,5-bis(trifluoromethyl)benzene **1.17** (5.0 g, 15.8 mmol) was added dropwise as a solution in diethyl ether (4.5 mL, 3.5 M). After 12 minutes, CO_2 gas was bubbled through the solution for 25 minutes. The gas line was then removed and the mixture was allowed to warm to room temperature. Hydrochloric acid (25 mL, 1 M) was directly poured into

the solution and stirred for 15 minutes. The mixture was extracted with CH_2Cl_2 (3 x 25 mL), dried over $NaSO_4$ filtered and concentrated in vacuo to afford **1.18** in 77% yield (3.38 g). $\mathbf{R_f}$ = 0.35 (45:55 EtOAc/Hex); **mp** = 142–144 ºC; **^{1}H NMR** (500 MHz, $CDCl_3$) δ 12.30–11.70 (bs, 1H), 8.58 (s, 2H), 8.15 (s, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 169.8, 132.8 (q, $J_{C\text{-}F}$ = 34.0 Hz), 131.5, 131.5, 130.8–130.2 (m), 127.7 (quint, $J_{C\text{-}F}$ = 3.7 Hz), 123.0 (q, $J_{C\text{-}F}$ = 271.2 Hz); **IR** (thin film) 2890, 2360, 1709, 1620 cm^{-1}; **HRMS** (ESI/methanol) *m / z* calcd for $C_9H_3F_6O_2$ $(M - H)^-$ 257.0037, found 257.0040.[26]



2,5-Dioxopyrrolidin-1-yl 3,5-bis(trifluoromethyl)benzoate (1.21). Acid **1.18** (2.0 g, 7.72 mmol) was suspended in a solution of pyridine and anhydrous CH_2Cl_2 (1.65 mL : 6.6 mL) and stirred to dissolution for 5 minutes. TFA-NHS[27] ester **1.25** was added to the solution and stirred for 4 hours. Upon completion as observed by TLC, the mixture was quickly washed with 5% $NaHCO_3$ (50 mL), washed with NH_4Cl (50 mL), and extracted with CH_2Cl_2 (3 x 25 mL). The combined organic extracts were dried over Na_2SO_4, filtered, and concentrated in vacuo to afford NHS-ester **1.21** in 99 % yield (0.678 g) for immediate use. $\mathbf{R_f}$ = 0.30 (45:55 EtOAc/Hex); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 8.58 (s, 2H), 8.19 (s, 1H), 2.88 (s, 4H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 168.8, 159.9, 133.1 (q, $J_{C\text{-}F}$ = 34.3 Hz), 130.8, 130.8, 128.5 (q, $J_{C\text{-}F}$ = 3.6 Hz), 127.7,

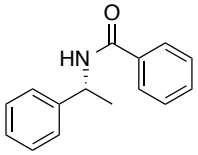
122.7 (q, J_{CF} = 271.5 Hz), 25.9. Further characterization was not possible due to facile NHS–ester bond hydrolysis.



***N*-((1*R*,2*R*)-2-(3-(3,5-Bis(trifluoromethyl)phenyl)thioureido)cyclohexyl)-3,5 bis(trifluoromethyl)-benzamide (1).** Aminothiourea **9** (1.0g, 2.59 mmol, 1.0 equiv) was dissolved in anhydrous THF (2.6 mL, 0.1 M) and stirred for 5 minutes. NHS-ester **15** (1.38g, 3.94 mmol, 1.5 equiv) was then added. Upon completion after 5 hours as observed by TLC (30:70 EtOAc/Hex), the mixture was concentrated in vacuo and purified via column chromatography (30:70 EtOAc/Hex) to afford thiourea **1** as a white solid in 90 % yield (1.48 g). **mp** = 145–147 ºC; $\mathbf{R}_f$ = 0.32 (30:70 EtOAc/Hex); $[\alpha]_D^{25}$ +20.5 (c 0.1, $CHCl_3$); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 8.58 (s, 1H), 8.24 (s, 1H), 7.95 (s, 1H), 7.66 (s, 2H), 7.63 (s, 2H), 7.21–7.06 (m, 1H), 4.80–4.55 (m, 1H), 4.11–3.81 (m, 1H), 2.35–2.15 (m, 2H), 1.98–1.81 (m, 2H), 1.58–1.34 (m, 4H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 181.8, 165.9, 139.2, 135.9, 132.7 (q, J_{C-F} = 33.9 Hz), 132.6 (q, J_{C-F} = 33.8 Hz), 127.6, 125.7, 124.1, 122.9 (q, J_{C-F} = 272.4 Hz), 122.9 (q, J_{C-F} = 272.40 Hz), 119.6, 57.5, 56.9, 32.3, 24.8; **IR** (thin film) 3278, 3074, 2943, 2866, 1651, 1547, 1385, 1281, 1180, 1134, 702 cm^{-1}. **HRMS** (ESI/methanol) *m / z* calcd for $C_{24}H_{19}F_{12}N_3OSNa$ $(M + Na)^+$ 648.0955, found 648.0952. Spectral data were consistent with those previously reported.[11-13]

General Procedure for the Kinetic Resolution of Primary Amines:

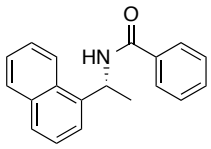
A 250 mL round-bottomed flask was charged with benzoic anhydride (378 mg, 1.65 mmol) and 4 Å MS (320 mg) which was followed by the addition of DMAP (5 mg, 0.04 mmol) as a solution in toluene (3.2 mL). Freshly distilled toluene was added to the mixture (68 mL) and the reaction mixture was placed in a dry ice/acetone bath at –78 ºC. After 15 minutes, thiourea catalyst **1** was added (25 mg, 0.04 mmol) as a solution in toluene (6.4 mL), and the solution allowed to stir for an additional 20 minutes. The primary amine (100 mg, 0.8 mmol) was added as a solution in toluene (3.2 mL) and the reaction mixture was allowed to stir for 2 hours before the addition of a 7.0 M methanolic ammonia solution (2.4 mL, 16.0 mmol). The cooling bath was removed, and the reaction mixture was allowed to warm to room temperature over 30 minutes. The crude mixture was concentrated in vacuo, and purified via column chromatography (15:85 EtOAc/Hex) to afford desired enantioenriched amide product.



1.28

(*R*)-*N*-(1-Phenylethyl)benzamide (21). Following the general procedure, compound **21** was obtained as white crystals in 56 % yield (104 mg). **mp** = 115–118 ºC; R_f = 0.33 (30:70 EtOAc/Hex); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 7.77 (d, *J* = 8.0 Hz, 2H), 7.49 (t, *J* = 7.25 Hz, 1H), 7.43–7.35 (m, 6H), 7.28 (app t, *J* = 7.0 Hz, 1H), 5.35 (quint, *J* = 7.0 Hz, 1H), 1.61 (d, *J* = 6.5 Hz, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 166.8, 143.3, 134.8, 131.7, 129.0, 128.8, 127.7, 127.1, 126.5, 49.4, 21.9; **IR** (thin film) 3298, 3062, 2931, 2958, 2360, 2341, 1635, 1535, 1489 cm^{-1};

HRMS (ESI/methanol) *m / z* calcd for $C_{15}H_{15}NONa$ $(M + Na)^+$ 248.1051, found 248.1060; **HPLC**: Daicel chiralpak OD-H, *i*PrOH/*n*-hexane=10/90, Flowrate = 1 mL/min, UV = 254 nm, t_R= 14.4 min (major) and t_R= 20.2 min (minor), e.r = 82:18. Spectral data were consistent with those previously reported in the literature.[11]



1.29

(*R*)-*N*-(1-(Naphthalen-1-yl)ethyl)benzamide (22). Following the general procedure, compound **22** was obtained as white crystals in 32 % yield (62 mg). **mp** = 192–194 ºC; $\mathbf{R_f}$ = 0.25 (30:70 EtOAc/Hex); **^{1}H NMR** (500 MHz, $CDCl_3$) δ 8.18 (d, *J* = 8.5 Hz, 1H), 7.88 (d, *J* = 8.0 Hz, 1H), 7.83 (d, *J* = 8.0 Hz, 1H), 7.73 (d, *J* = 7.5 Hz, 2H), 7.60 (d, *J* = 7.0 Hz, 1H), 7.56–7.45 (m, 4H), 7.40 (t, *J* = 7.8 Hz, 2H), 6.33 (d, *J* = 7.5 Hz, 1H), 6.14 (quint, *J* = 7.3 Hz, 1H), 1.79 (d, *J* = 6.5 Hz, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 166.6, 138.3, 134.7, 134.2, 131.7, 131.4, 129.0, 128.8, 127.1, 126.9, 126.2, 125.4, 123.7, 122.9, 45.4, 20.8; **IR** (thin film) 3298, 3059, 2978, 2927, 1631, 1535 cm^{-1}; **HRMS** (ESI/methanol) *m / z* calcd for $C_{19}H_{17}NONa$ $(M + Na)^+$ 298.1208; found 298.1202. **HPLC**: Daicel chiralpak OD-H, *i*PrOH/*n*-hexane=10/90, Flowrate = 1mL/min, UV = 254nm, t_R= 12.8 min (major) and t_R= 30.5 min (minor), e.r = 87:13. Spectral data were consistent with those previously reported.[11]

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[14] Figure adapted from our previous studies on the determination of absolute configuration using kinetic resolution catalysts.

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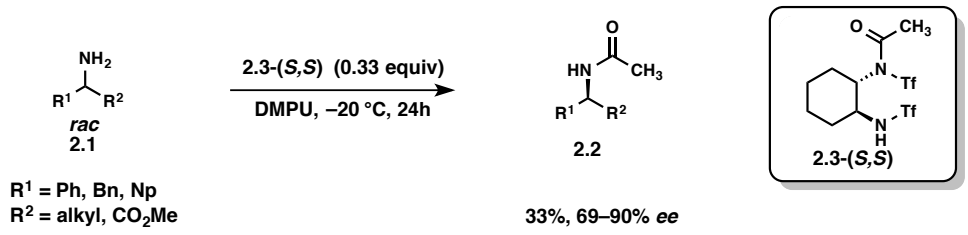
Chapter 2

Utilizing Kinetic Resolution Reagents to Assign Absolute Configuration

I. Abstract: Herein is described a new method to determine the absolute configuration of primary amines. Our strategy combines Mioskowski's enantioselective acylation reagents with strategic deuterium incorporation and Electrospray Ionization-Mass Spectrometry (ESI-MS), which has produced a rapid and accurate approach to determine the absolute configuration of amines based on a mass difference.[1]

II. Introduction: In 2004, Mioskowski and co-workers introduced the use of chiral bis(sulfonamide) **2.3-(*S,S*)** for the kinetic resolution of primary amines (Scheme 2.1).[2] These kinetic resolution reagents take advantage of the trans-1,2-diaminocyclohexane **2.4-(*S,S*)** as a chiral scaffold and are effective on a broad scope of primary amines. Furthermore, the reagent's stability to hydrolysis as well as long bench stability and ease of preparation attracted us to investigate its applications for assigning the absolute configuration of primary amines.

Scheme 2.1. Mioskowski's kinetic resolution method.

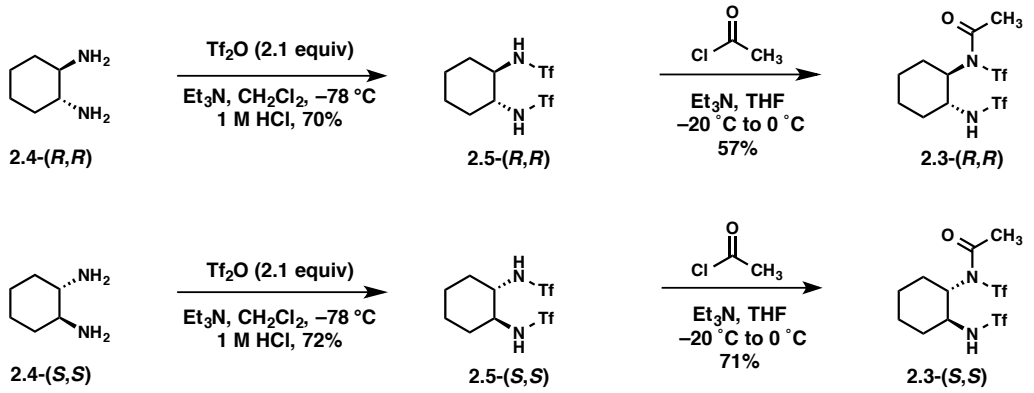


III. Absolute Configuration Assignment of Primary Amines Using ^{1}H NMR Spectroscopy Performed by Dr. Shawn M. Miler.

The Rychnovsky group's early attempts to assign the absolute configuration of primary amines via the Competing Enantioselective Conversion (CEC) method were performed by Dr.

Shawn Miller.[4] Initial studies began with the preparation of Mioskowski's kinetic resolution reagents **2.3-(*R*,*R*)** and **2.3-(*S*,*S*)** according to a literature procedure.[2a] Bistrifluoromethanesulfonylation of *trans*-cyclohexyldiamines **2.4-(*R*,*R*)** and **2.4-(*S*,*S*)** provided both **2.5-(*R*,*R*)** and **2.5-(*S*,*S*)**-bis(sulfonamides) in 70 and 72% yield, respectively (Scheme 2.2). Acylation of the bis(sulfonamides) with acetyl chloride afforded reagents **2.3-(*R*,*R*)** and **2.3-(*S*,*S*)** in 57% and 71% yields, respectively. This reagent combination was used to develop effective reaction conditions for the selective acylation of amines.

Scheme 2.2. Synthesis of enantiopure Miowskowski's reagents.



The approach to determining the absolute configuration of primary amines using ^{1}H NMR was conducted in a similar fashion to our reported method for secondary alcohols.[5] Initial studies to determine the feasibility of the method involved two separate reactions of amine **2.6-(*R*)** with reagents **2.3-(*R*,*R*)** and **2.3-(*S*,*S*)** conducted in triplicates. Conversion to amide product in the reactions was measured by integration of the H atom adjacent to the amine or amide (Figure 2.1).

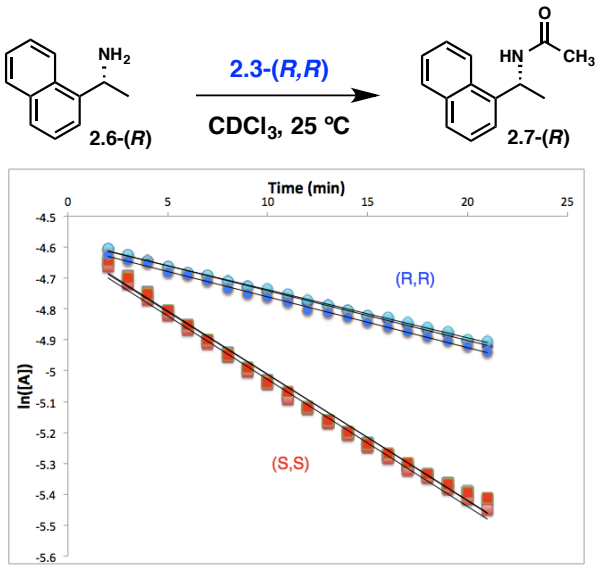


Figure 2.1. Kinetic studies using Mioskowski's reagents.

Dr. Miller's initial experiments utilized 500.0 µL of chloroform solvent to ensure sufficient volume for the spectrometers to lock the sample. Earlier studies on secondary alcohols used a total concentration of 0.1 M, which proved sufficient for ^{1}H NMR resolution and reaction rates. Because amines are inherently more nucleophilic, a concentration of 0.01 M of amine substrate **2.6-(*R*)** was instead used. Lastly, three equivalent of reagents **2.3-(*R*,*R*)** and **2.3-(*S*,*S*)** were used to maintain pseudo-first-order kinetics. Analysis of the data determined that the faster reacting reagent was **2.3-(*S*,*S*)** by a factor of 2.6 (Figure 2.1). Control studies using the opposite enatiomer, amine **2.6-(*S*)**, provided similar results with an expected switch in selectivity. With a selectivity demonstrated for both enantiomers of **2.6** as well as other primary amine substrates, a mnemonic was established to predict the absolute configuration of primary amines (Figure 2.2). The predictive mnemonic places the larger substituent to the left of the amine and the smaller substituent to the right. If the reagent **2.3-(*R*,*R*)** reacts faster, the amine is facing forward. Alternatively, when the **2.3-(*S*,*S*)** reagent reacts faster, the amine is back.

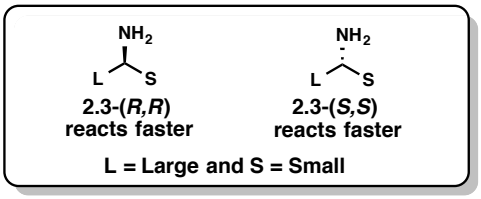


Figure 2.2. Predictive mnemonic for primary amines.

Conclusions: Dr. Miller demonstrated that ^{1}H NMR could be used to assign the absolute configuration of primary amines after the side-by-side reactions with each enantiomer Mioskowski's reagents. A variety of amine substrates were investigated for the method and a predictive mnemonic was established. While the approach of using NMR to determine the faster reaction was straightforward, a few limitations further prevented its use. The protocol required significant instrument time and sufficient amounts of material for the analysis. Furthermore, well-resolved ^{1}H NMR signals were not always obtained, complicating the data analysis. A new approach that could circumvent these limitations was envisioned and its development is presented in the following section of this chapter.

IV. Nanomole-Scale Assignment of Absolute Configuration of Primary Amines Using Electrospray Ionization Mass-Spectrometry

A new approach using mass spectrometry (MS) was envisioned to simplify the analysis. MS would allow for the rapid detection of species while only requiring small quantities of amine for the analysis. The new strategy featured the use of isotopically labeled pseudoenantiomers of Mioskowski's enantioselective reagents. With the new approach, the amine **2.4** to be evaluated of "unknown" absolute configuration is treated with an excess of equimolar mixture of **2.3-(*R,R*)** and **2.3-(*S,S*)-*d*$_3$** to afford a mixture of unlabeled acetamide **2.5**-(M) and deuterium labeled acetamide **2.5**-(M+3) (Figure 2.3A). By maintaining pseudo-first-order kinetics, the

relative ratios of the **2.5**-$(M)^+$ and **2.5**-$(M+3)^+$ peaks in the MS-ESI spectrum would then be used to determine which reagent reacted faster. If the relative abundance of $(M)^+$ is higher than $(M+3)^+$, then **2.3-(*R,R*)** reacted faster (Figure 2.3B). Alternatively, if the $(M+3)^+$ is more pronounced than $(M)^+$ then **2.3-(*S,S*)-*d*$_3$** reacted faster (Figure 2.3C). It is important to acknowledge that the work of my colleague, Dr. Shawn Miller, provided groundwork for the ESI-MS approach, which was presented in section II of this chapter. I joined the mass spectrometry project at an early stage of its development and our combined efforts are described here in.

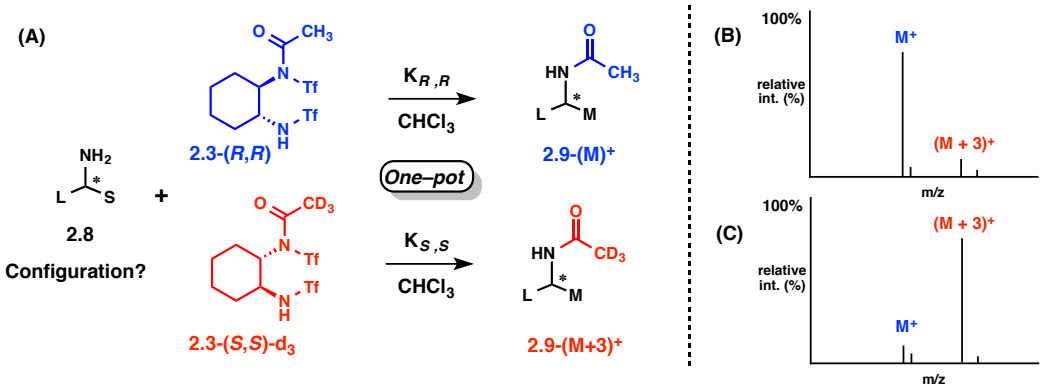
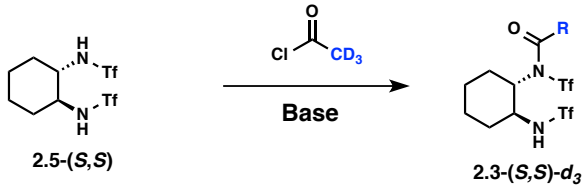


Figure 2.3. Proposed absolute configuration method of amines via mass-spectrometry.

Results and Discussion: The project began with the preparation of isotopically labeled reagent **2.3-(*S,S*)-*d*$_3$** using a modified protocol.[6] Initial experiments showed significant deuterium loss during the installation of the d_3-acetyl group (Table 2.1, entries 1 and 2). The deuterium loss presumably occurs from the generation of a ketene intermediate formed *in situ*. As an alternative to avoid ketene formation, catalytic amounts of DMAP as well as excess was employed (entries 3 and 4), though no reaction was observed. After careful screening, it was found that pyridine and acetyl chloride-d_3 yielded the desired reagent albeit with 5–22% deuterium loss (entry 5).

Table 2.1. Optimization of deuterated reagent **2.3-(*S,S*)-d_3**.



2.5-(*S,S*) 2.3-(*S,S*)-d_3

entry	Base	Conditions	Product (%)
1	Et_3N (1.5 equiv)	THF, 25 °C	R = CHD_2 (75 %)
2	Et_3N (0.9 equiv)	THF, –20 to 0 °C	R = CHD_2 (57%)
3	DMAP (10 mol%)	THF, –20 to 25 °C	No Reaction
4	DMAP (7 equiv)	THF, –20 to 25 °C	No Reaction
5	Pyridine (10 equiv)	Et_2O, –20 to 25 °C	R = CD_3 (71%)

With the desired reagents **2.3-(*R,R*)** and **2.3-(*S,S*)-d_3** in hand, the method development using ESI-MS began. Initially, the reactions were run with 5.0 μmol of amine and 1.5 μmol of both **2.3-(*R,R*)** and **2.3-(*S,S*)-d_3** in a total volume of 500.0 μL. Due to the sensitivity of ESI-MS, reactions could be reduced in scale to 0.5 μmol with no observed change in rates.

Reactions were carried out in MS vials for 60 minutes, at which point the reaction was quenched with methanol and directly subjected to ESI-MS. As predicted, the selectivity of the reaction could be determined from the MS readout (Figure 2.4). The reaction of **2.10-(*R*)** with **2.3-(*R,R*)** and **2.3-(*S,S*)-d_3** showed a more intense $(M+3)^+$ signal indicating a faster reaction with **2.3-(*S,S*)-d_3** (Figure 2.4A)

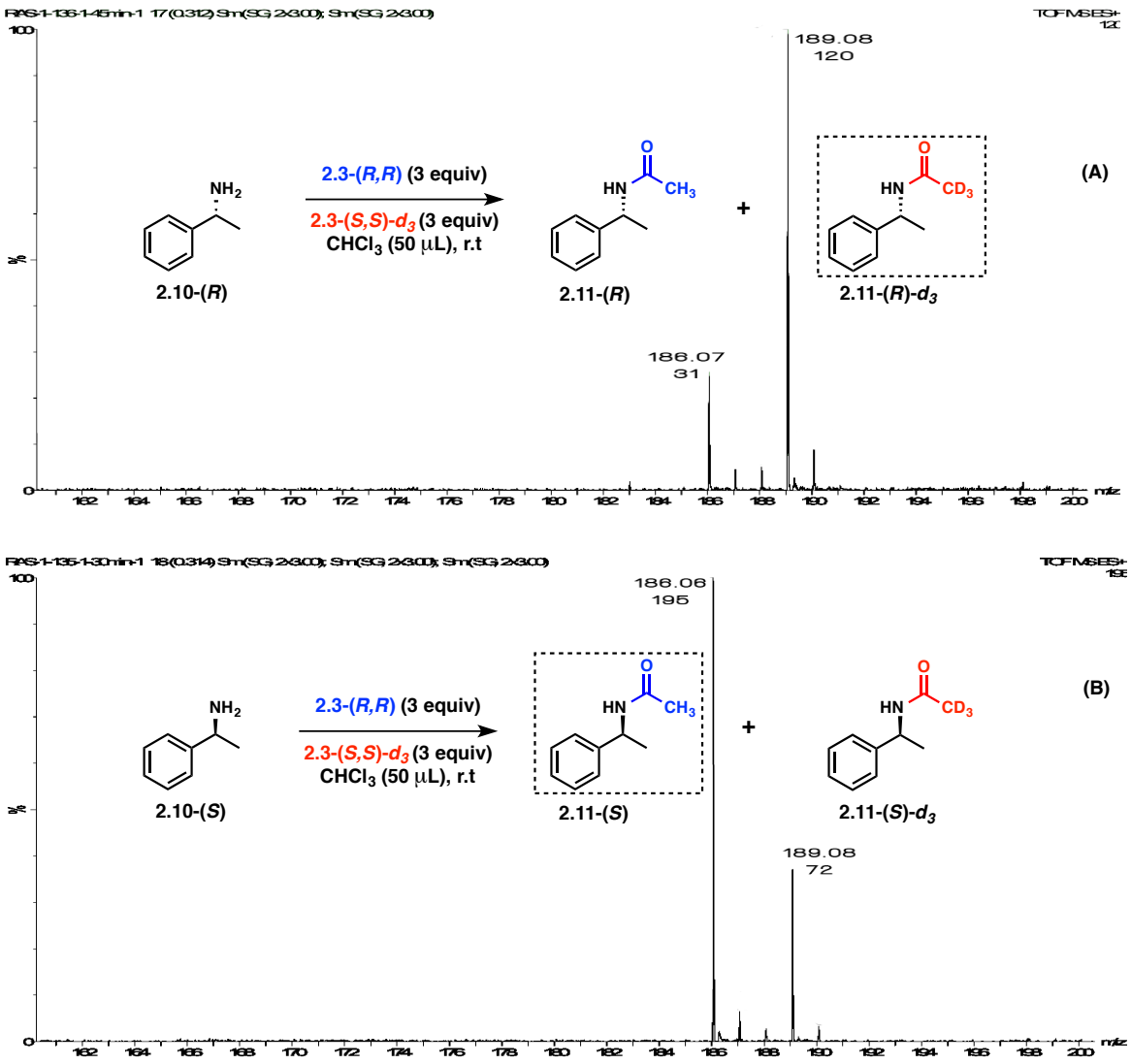


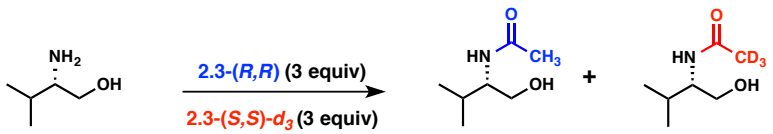
Figure 2.4. (A) MS spectra of the reaction with **2.10-(*R*)**. (B) MS spectra of the reaction with **2.10-(*S*)**.

As a proof concept, the opposite enantiomer **2.10-(*S*)** was subjected to the same conditions described previously (Figure 2.4B). Gratifyingly, the opposite selectivity was observed, therefore confirming that absolute configuration could indeed be assigned using mass spectrometry.

Because the acylated products of the reaction existed as both the protonated and sodiated ion species in MS-ESI, it was rationalized that generation of single ion-bound peaks (either

protonated or sodiated) would improve the analysis. During preliminary studies, quenching of the acylation reaction with methanol showed both of the protonated and sodiated species (Table 2.2, entry 1). Initial attempts focused on suppressing the sodiated ions to generate the protonated species only. When the reaction was quenched with a solution of 10% formic acid in methanol (entry 2), both species continued to be observed in the readout. Increasing the acidity using 20% acetic acid in methanol resulted in minimal decrease of the sodiated ions (entry 3). Further increase in acidity may have eventually led to formation of protonated ions only, but concerns of decomposition or fragmentation during the analysis prevented further exploration. Instead, the use of 50 mM of NaOAc in methanol ensured full conversion to the sodiated ions.

Table 2.2. Generation of sodium-bound peaks.[a]



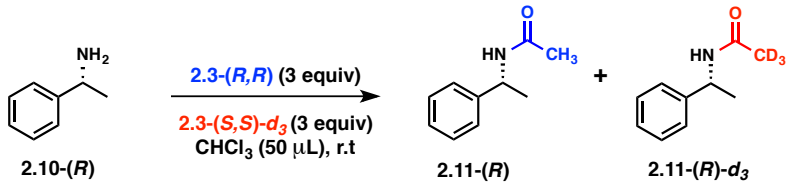
entry	Quenching Source		M+ Intensity	:	(M+3)+ Intensity	# trials / δ
1	MeOH	H^+	1	:	4.55	3 / 0.18
		Na^+	1	:	2.87	3 / 0.02
2	10% HCO_2H / MeOH	H^+	1	:	4.32	3 / 0.01
		Na^+	1	:	2.85	3 / 0.12
3	20 % AcOH / MeOH	H^+	1	:	3.02	3 / 0.26
		Na^+	1	:	2.80	3 / 0.03
4	50 mM NaOAc / MeOH	H^+	–	:	–	–
		Na^+	1	:	2.94	3 / 0.05

a) The alcohol group behaves as small groups therefore the 2.3-(*S*,*S*)-d_3 reacts faster. See Table 2.5

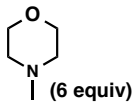
In the report by Mioskowski and co-workers, the amine to be resolved is present in two-fold excess relative to the kinetic resolution reagent.[2] Such amine excess creates a basic environment under the reaction conditions. With this in mind, a base additive was added to our reaction conditions in an attempt to optimize the methodology (Table 2.3). Three non-nucleophilic bases were initially selected for the studies. Under standard conditions, the reaction of amine **2.10-(*R*)** with **2.3-(*R,R*)** and **2.3-(*S,S*)-*d*$_3$** produced a more intense $(M+3)^+$ signal with a selectivity factor of ~ 1 : 2.68 (Table 2.3, entry 1). When the reaction was conducted in the presence of Et_3N, the selectivity factor decreased significantly to a 1:1.13 ratio that favored **2.11-(*R*)-*d*$_3$** (entry 2). A slight enhance in selectivity, however, was observed when 4-methylmorpholine was employed as an additive (entry 3). Surprisingly, switching to Hünig's base additive showed a reversal in selectivity that gave a relative ratio of ~ 1.51:1 that favored **2.11-(*R*)** (entry 4). Finally, analogous kinetic resolution conditions using sub-stoichiometric amounts of **2.3-(*R,R*)** and **2.3-(*S,S*)-*d*$_3$** were carried out without improvement of selectivity (entries 5 and 6).

It appears that increasing the basicity around the nitrogen atom of **2.10-(*R*)** reduces the selectivity of the acylation process (entries 2, 4, 5 and 6). These observations maybe associated with an increase in nucleophilicity that affects the enantioselectivity of the reaction. While the use of 4-methylmorpholine showed a modest increase in selectivity, it was not significant enough to warrant adding an extra component to the reaction.

Table 2.3. Attempts at improving selectivity using base additives.



2.10-(*R*) 2.11-(*R*) 2.11-(*R*)-d_3

entry	Conditions		M^+ Intensity	:	$(M+3)^+$ Intensity	# trials / δ
1	No Base	Na^+	1	:	2.68	5 / 0.08
2	Et_3N (6 equiv)	Na^+	1	:	1.13	3 / 0.03
3	 (6 equiv)	Na^+	1	:	2.84	3 / 0.03
4	Hünig's Base (6 equiv)	Na^+	1.51	:	1	3 / 0.12
5 [a]	(*R*, *R*) : (*S*, *S*)-d_3 (0.5 equiv)	Na^+	1	:	1.99	2 / 0.01
6 [b]	(*R*, *R*) : (*S*, *S*)-d_3 (0.25 equiv)	Na^+	1	:	1.90	2 / 0.13

a) 0.5 equiv. of each reagent 2.3-(*R*,*R*) and 2.3-(*S*,*S*)-d_3 were employed. (b) 0.25 equiv. of each reagent 2.3-(*R*,*R*) and 2.3-(*S*,*S*)-d_3 were employed.

As discussed earlier, depending on the batch of synthesized of **2.3-(*S*,*S*)-d_3**, deuterium incorporation often results in less than 100%. As such, a correction factor to account for deuterium loss needed to be established in order to obtain accurate ratios of the $(M)^+$ and $(M+3)^+$ relative abundances. A correction factor was determined by reacting amine **2.10-(*R*)** with reagent **2.3-(*S*,*S*)-d_3** under the established optimized conditions (Figure 2.5). Analysis of the MS readout revealed an $(M+2)^+$ signal with an ion count of 2687, corresponding to **2.11-(*R*)-d_2** and the $(M+3)^+$ that belonged to **2.11-(*R*)-d_3** signal with a count of 12444. The ratio of $(M+2)^+$ to the $(M+3)^+$ signal is the correction factor by which the peak $(M+3)^+$ is increased. In this particular case, the true value of $(M+3)^+$ is 15131 which results in a 22% increase to the raw data. This

correction factor was applied to the analysis of compounds shown on Table 2.4 that employed this particular **2.3-(*S,S*)-d_3** batch.

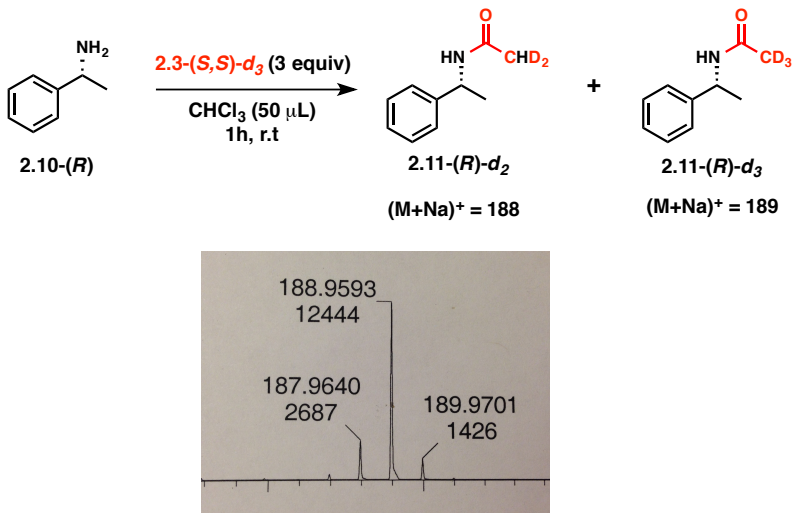
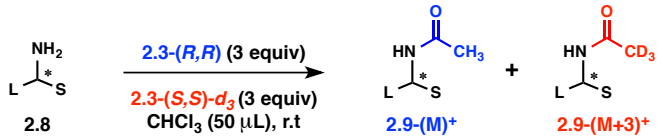


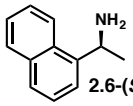
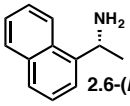
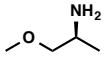
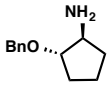
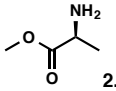
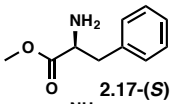
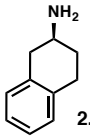
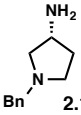
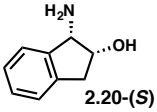
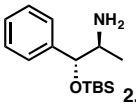
Figure 2.5. Determining a correction factor for loss of deuterium.

With optimized conditions at hand and a procedure to compensate for deuterium loss, the assignment of the absolute configuration of a variety of primary amines was undertaken. Multiple trials were conducted and the averaged ratios, as well as standard deviations, are listed on the Table 2.4.

As expected, enantiomeric pair **2.6-(*S*)** and **2.6-(*R*)** showed complimentary selectivities, although a small difference in ratios was observed (Table 2.4, entries 1 and 2). This small difference may result from mechanical errors such as the weighing of starting materials. The method was effective for amino ester derivatives in which the ester behaves as a larger group compared to the adjacent sp^3 carbon (entries 5 and 6). Amines with remote substituents can also be analyzed using the CEC method (entries 7 and 8).

Table 2.4. Determination of the absolute configuration of primary amines.[a]



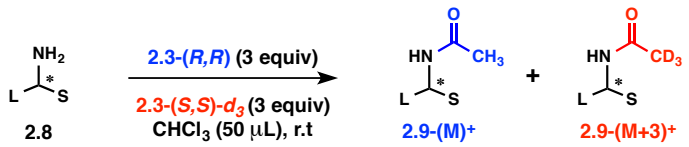
entry	amine	M^+ Intensity[b]	:	$(M+3)^+$ Intensity[b]	# trials / δ [c]
1	 2.6-(*S*)	**3.13**	:	1	5 / 0.03
2	 2.6-(*R*)	1	:	**3.38**	5 / 0.08
3	 2.14-(*S*)	**1.40**	:	1	3 / 0.02
4	 2.15-(*S*)	**1.22**	:	1	5 / 0.006
5	 2.16-(*S*)	**3.67**	:	1	3 / 0.04
6	 2.17-(*S*)	**4.16**	:	1	3 / 0.12
7	 2.18-(*S*)	**1.44**	:	1	3 / 0.02
8	 2.19-(*R*)	1	:	**1.10**	3 / 0.02
9	 2.20-(*S*)	1	:	**1.03**	3 / 0.003
10	 2.21-(*S*)	**1.12**	:	1	3 / 0.01

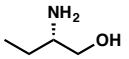
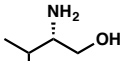
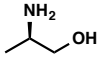
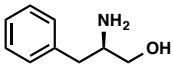
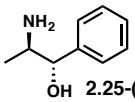
(a) All reactions were run for 1h and analyzed by ESI-MS. (b) The sodium peaks were analyzed in all cases. (c) The standard deviation (δ) for multiple runs is included.

It worth noting that caution should be taken when encountering smaller selectivities during the analysis (entries 8 and 9). For example, ratio values of less than 1.2:1 are considered to be inconclusive and alternative methods for assigning the absolute configuration should be

considered. The method was also applied to substrates that contained additional protic moieties, such as the amino alcohols shown in Table 2.5. In the case of alcohols, they behave as small groups even when they carry substituents (entry 5).

Table 2.5. Absolute configurations of α-amino alcohols.[a]



entry	amine	M^+ Intensity[b]	:	$(M+3)^+$ Intensity[b]	# trials / δ [c]
1	 2.22-(*S*)	1	:	**1.93**	5 / 0.04
2	 2.12-(*S*)	1	:	**2.87**	3 / 0.02
3	 2.23-(*R*)	**1.66**	:	1	5 / 0.03
4	 2.24-(*R*)	**2.30**	:	1	3 / 0.02
5	 2.25-(*R*)	**1.22**	:	1	3 / 0.02

[a–c] See the corresponding footnotes in Table 2.4

Based on these studies, a predictive mnemonic to assign absolute configuration using ESI-MS is presented in Figure 2.6. The mnemonic places the "large" group to the left and the "small" group to the right of the amine. If the reagent **2.3-(*R,R*)** reacts faster, the amine is facing forward, and when reagent **2.2-(*S,S*)-*d*$_3$** reacts faster, the amine is back. Carbonyl and aromatic groups, both which contain sp^2 carbons, behave as large groups. Alcohols behave as small groups (Table 2.5), but when protected behave as bulky substituents (Table 2.4, entries 2.9 and 2.10).

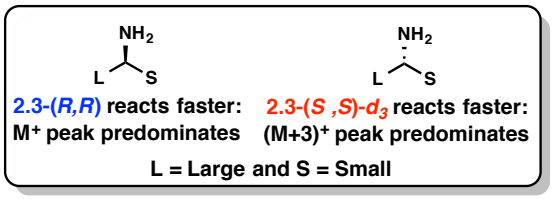
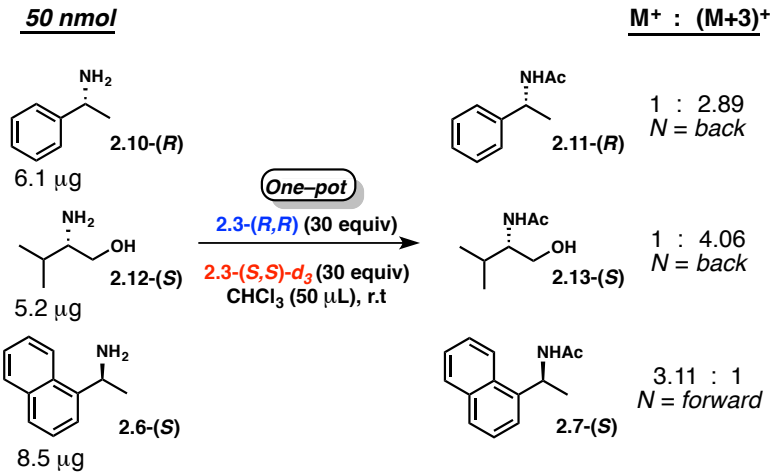


Figure 2.6. Mnemonic for assigning absolute configuration of primary amines.

The standard analysis uses 0.5 μmol of amine, which translates to ~75 μg of material, consumed. In order to further exploit the potential of ESI-MS for determining configuration, one additional experiment was performed (Scheme 2.3). Because amines that contained different functional groups afforded acylated products of different masses, it was rationalized that the analysis could be carried out in a single experiment as opposed to several experiments. With this in mind, a mixture containing 50 nmol (<10 μg) of each of the three amines was subjected to the optimized CEC conditions. After one hour, ESI-MS analysis of the crude mixture provided the absolute configuration of all three amines in a single experiment.

Scheme 2.3. Configuration assignment of various amines in a single experiment.



V. Conclusions: A new method for assigning absolute configuration was presented in this chapter. The new strategy uses a pseudoenantiomeric pair of kinetic resolution reagents and mass spectrometry to rapidly establish the absolute configuration of a variety of primary amines based

on mass differences of the acylated products. Based on the experimental data, a predictive mnemonic was established. Furthermore, the analytical tool developed in this chapter was effective on micromole down to nanomole-scale and was applied to mixture of amines. Finally, other members of the Rychnovsky group are expanding the core concepts of this methodology to secondary amines.

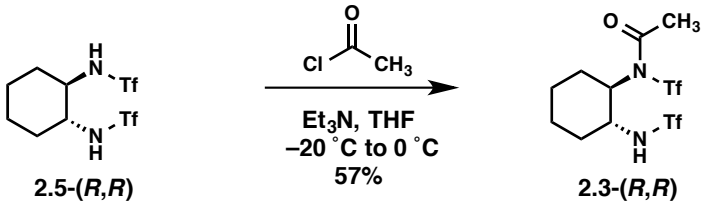
Supporting Information

VI. General experimental and laboratory conditions:

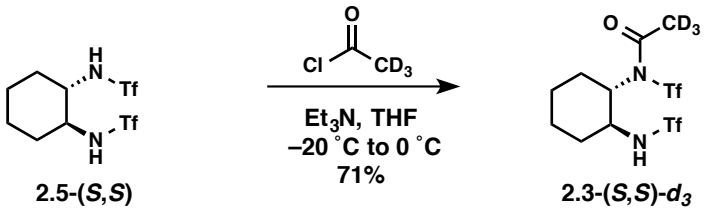
All glassware was flame- or oven-dried and cooled under argon unless otherwise stated. All reactions and solutions were conducted under argon unless otherwise stated. All commercially available reagents were used as received, unless otherwise stated. Toluene (PhMe), tetrahydrofuran (THF), dimethylformamide (DMF), diethyl ether (Et_2O) and dichloromethane (CH_2Cl_2) were degassed and dried by filtration through activated alumina under vacuum according to the procedure by Grubbs.[7] Diisopropylamine (DIPA), acetonitrile (MeCN), 1,3-Dimethyl-3,4,5,6-tetrahydro-2-pyrimidinone (DMPU) were distilled from CaH_2 prior to use. All reactions involving LiDBB were conducted with glass stirbars. Thin layer chromatography (TLC) was performed with Millipore 60 F_{254} glass-backed silica gel plates and visualized using potassium permanganate, Dragendorff-Munier, ceric ammonium molybdate (CAM) or vanillin stains. Flash column chromatography was performed according to the method by Still, Kahn, and Mitra[8] using Millipore Geduran Silica 60 (40-63 μm).

Instrumentation

All data collected at ambient temperature unless noted. ^{1}H NMR spectra were taken at 500 or 600 MHz, calibrated using residual NMR solvent or TMS and interpreted on the δ scale. Peak abbreviations are listed: s = singlet, d = doublet, t = triplet, q = quartet, pent = pentet, dd = doublet of doublets, ddd = doublet of doublet of doublets dt = doublet of triplets, ddt = doublet of doublet of triplets, dq = doublet of quartets, m = multiplet, app = apparent, br = broad. ^{13}C NMR spectra were taken at 125 MHz, calibrated using the NMR solvent, and interpreted on the δ scale.



***N*-((1*R*,2*R*)-2-(trifluoromethylsulfonamido)cyclohexyl)-*N*-(trifluoromethylsulfonyl) ethanamide (2.3-(*R*,*R*))** Freshly distilled acetyl chloride (94.4 μL, 1.32 mmol) was added dropwise to a stirred solution of 1,2-bis(trifluoromethanesulfonamide) cyclohexane (0.50 g, 1.32 mmol) and triethylamine (276 μL, 1.98 mmol) in tetrahydrofuran (10 mL) at –20 ^{o}C. The resulting mixture was allowed to stirred for 5 hours at 0 ^{o}C. Upon completion after 5 hours as observed by TLC (15:85 EtOAc/Hex), the mixture was concentrated under vacuo and purified via column chromatography (8:92 EtOAc/Hex) to afford the desired compound as a white solid in 57 % yield (0.32 g). **^{1}H NMR** (500 MHz, $CDCl_3$) δ 5.01 (d, J = 10.0 Hz, 1H), 4.27 (br s, 1H), 3.76 (br s, 1H), 2.51 (s, 3H), 2.49–2.42 (m, 1H), 2.27–2.23 (m, 1H), 1.86–1.83 (m, 2H), 1.81–1.75 (m,1H), 1.45–1.18 (m, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 170.2, 119.7 (q, J_{C-F} = 318.5), 119.6 (q, J_{C-F} = 318.4), 67.2, 54.7, 35.5, 29.3, 27.1, 25.7, 24.6; IR (thin film) 3301, 2949, 2868, 1716, 1457, 1388, 1195, 1148, 1131, 1071, 1015, 971, 941, 919, 896, 862, 729; **HRMS** (ESI/methanol) m / z calcd for $C_{10}H_{14}F_6N_2O_5S_2Na$ $[M+Na]^+$ 443.0146, found 443.0141; **mp** = 107–109 ^{o}C; $\mathbf{R}_f$ = (15:85 EtOAc/Hex).



***N*-((1*S*,2*S*)-2-(trifluoromethylsulfonamido)cyclohexyl)-*N*-(trifluoromethylsulfonyl) ethanamide-d_3 (2.3-(*S*,*S*)-d_3)** Acetyl chloride-d_3 (121 μL, 1.65 mmol) was added dropwise to a stirred solution of 1,2-bis(trifluoromethanesulfonamide)cyclohexane (0.50g , 1.32 mmol) and freshly distilled pyridine (1.06 mL, 13.2 mmol) in diethylether (20 mL) at –20 °C. The mixture was allowed to stirred overnight at room temperature. Upon completion as observed by TLC (15:85 EtOAc/Hex), the mixture was concentrated under vacuo and purified via column chromatography (8:92 EtOAc/Hex) to afford the desired compound as a white solid in 71 % yield (0.40 g). **^{1}H NMR** (500 MHz, $CDCl_3$) δ 5.11 (s, 1H), 4.27 (br s, 1H), 3.76 (br s, 1H), 2.55–2.38 (m, 1H), 2.28–2.21 (m, 1H), 1.90–1.83 (m, 2H), 1.81–1.74 (m,1H), 1.44–1.18 (m, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 170.2, 119.6 (q, J_{C-F} = 318.5), 119.5 (q, J_{C-F} = 318.4), 67.2, 54.7, 35.6, 29.3, 27.1, 25.7, 24.6; **IR** (thin film) 3299, 2948, 2867, 1712, 1458, 1415, 1383, 1194, 1146, 1131, 1074, 1000, 955, 919, 818, 792, 740; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{10}H_{14}F_6N_2O_5S_2Na$ $[M+Na]^+$ 446.0337, found 446.0347; **mp** = 110–112 °C; $\mathbf{R}_f$ = (15:85 EtOAc/Hex).

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Chapter 3

Lycopodium Alkaloids: Background and Synthesis Strategies

I. Introduction: The *Lycopodium* alkaloids are a diverse group of structurally complex natural products isolated from the *Lycopodium* club mosses.[1] Over 250 *Lycopodium* alkaloids had been reported and many were shown to possess interesting biological activities ranging from neurotropic to anticancer properties.[2,3] Initial studies by Bödeker in 1881 led to the isolation of lycopodine (**3.1**) from the *Lycopodium complanatum*.[4] Since then, the *Lycopodium* alkaloid family has attracted extensive attention from the scientific community.

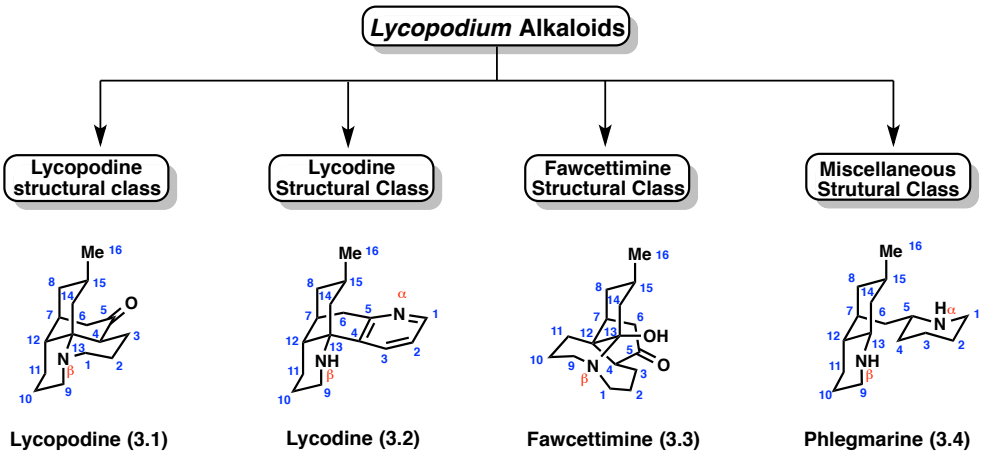


Figure 3.1. Structural classes of the *Lycopodium* alkaloids.

The *Lycopodium* alkaloids are classified into four structural classes: the lycopodine, the lycodine, the fawcettimine, and the miscellaneous class (Figure 3.1).[1e] The lycopodine class is the largest class of the family with over 79 isolated compounds. In terms of structural features, the lycopodines contain a tetracyclic core composed of four connected 6-membered rings (e.g., lycopodine (**3.1**)). The lycodine structural class contains a modified tetracyclic core where the αN and C1 are connected forming an annulated pyridine or pyridinone ring (e.g., lycodine (**3.2**)).

The fawcettimine class also contains a tetracyclic ring system with a migrated C4-C13 bond that forms a C4-C12 linkage (e.g., fawcettimine (**3.4**)). Any remaining alkaloids that do not belong to the first three groups are classified under the miscellaneous class, with phlegmarine being a representative case (**3.4**). Structural numbering of the *Lycopodium* alkaloids is presented in accordance to Conroy's biogenetic proposal.[5]

II. Proposed Biosynthesis: The biogenetic origin of these intriguing molecules has attracted the attention of several groups in the past. However, due to unsuccessful cultivation and limited access to *in*-vitro propagation, the biosynthesis of these molecules has not been fully established.[1f] Through ^{14}C and ^{13}C feeding studies a proposed biosynthesis was reported (Figure 3.2).[6] Initially, the amino acid lysine (**3.5**) is decarboxylated to form cadaverine (**3.6**), which then undergoes an oxidative cyclization to form piperideine (**3.7**). Coupling of piperideine (**3.7**) with dicarboxylic acid (**3.8**), or its bisCoA ester (**3.9**), followed by decarboxylation forms pelleterine (**3.11**), a key intermediate in the biosynthesis of the *Lycopodium* alkaloids.[6b] Pelleterine (**3.11**), or some derivative thereof, then dimerizes via an intermolecular aldol reaction to form dimer **3.12**. Oxidation of **3.12** provides intermediate **3.13**, which upon cyclization produces the phlegmarine skeleton **3.14**. Studies suggest that the phlegmarine skeleton is a found in all *Lycopodium* alkaloids and serves as common intermediate in the biosynthesis of the family.[6c] Tricycle **3.14** undergoes an intramolecular Mannich reaction to form the C13–C4 bond of compound **3.15**, which reacts further to produce lycodine (**3.2**). Cleavage of αN–C1 and rearrangement of **3.15** forms the lycopodine (**3.1**). Skeletal rearrangement of C13–C4 bond to form a new C4–C12 bond allows access to Fawcettimine (**3.3**).[7]

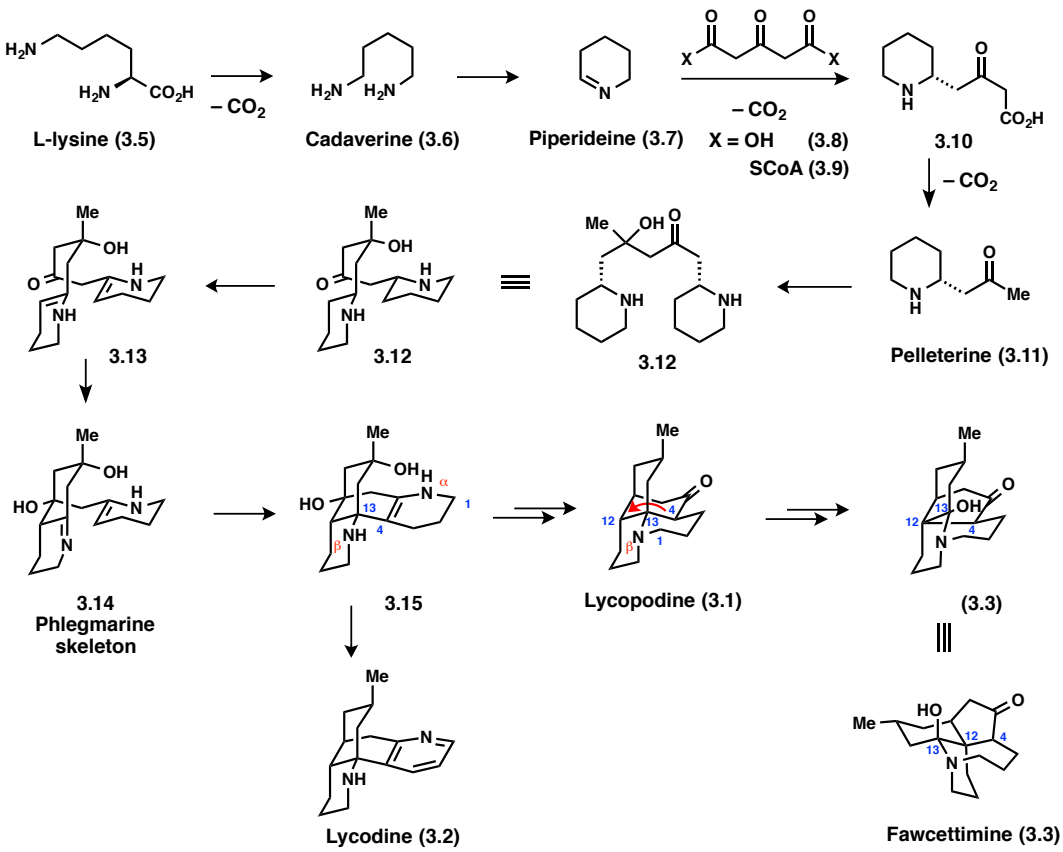


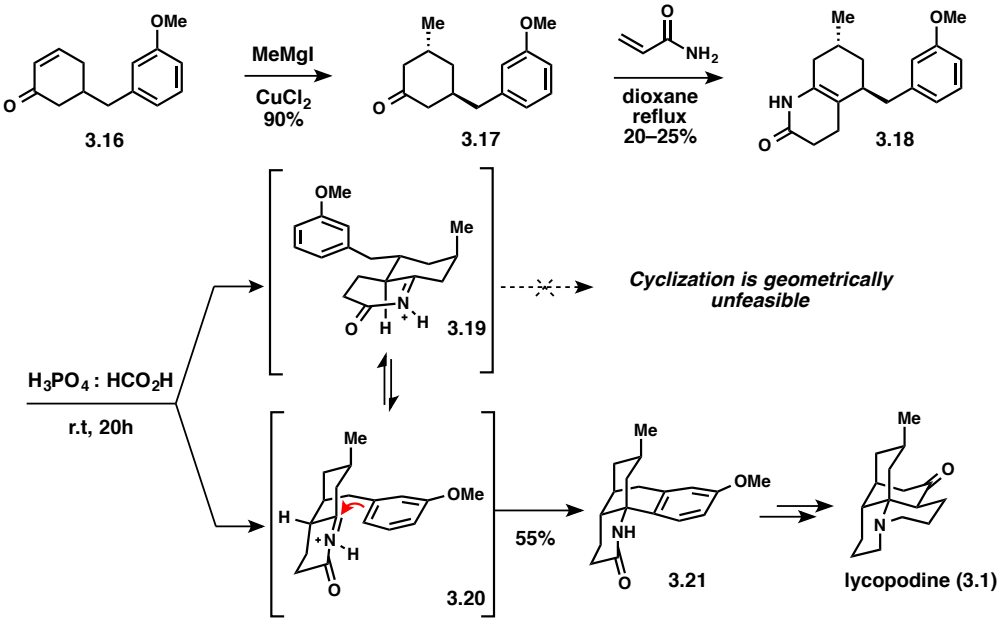
Figure 3.2. Proposed biosynthesis of the *Lycopodium* alkaloids.

Strategies for synthesizing the *Lycopodium* alkaloids: Numerous approaches towards making these molecules led to the discovery of new and efficient synthetic methods. In the following sections of this chapter four selected total syntheses will be discussed to provide background of the strategies that motivated us to pursue our studies described in chapter 4.

III. Highlights of Stork's Synthesis of (±)-Lycopodine: In 1968, Stork and coworkers reported the first total synthesis of lycopodine (**3.1**), which was developed on the basis of a Mannich reaction variant.[8] The synthesis began with a 1,4-addition reaction of an organocuprate reagent derived from methyl magnesium bromide into **3.16**. The conjugate addition reaction proceeded

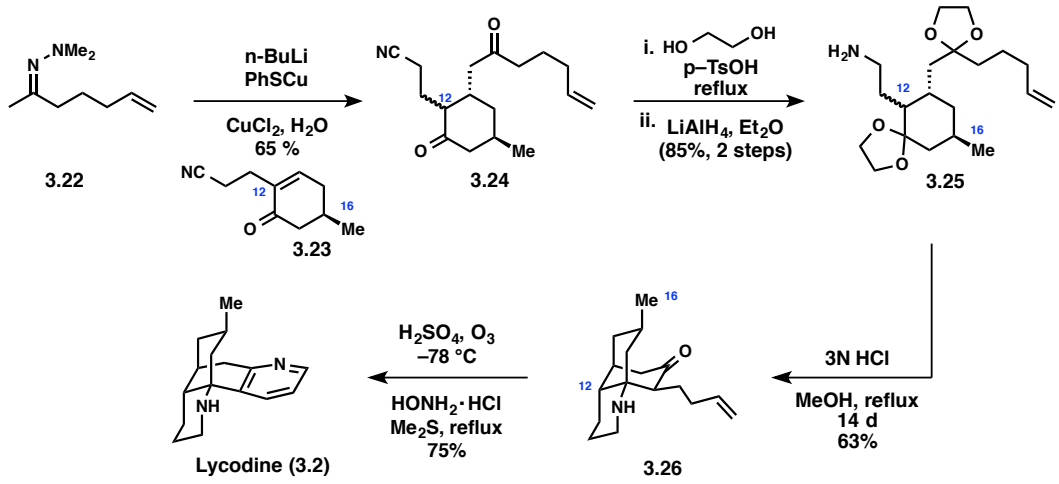
with the correct stereochemistry to afford *trans* **3.17** in 90% yield. Condensation of acrylamide with pyrrolidinenamine of **3.17** produced quinolone **3.18** in 20% yield, along with its undesired regioisomer, which was purified via recrystallization. Stork postulated that upon reaction with acid, **3.18** could produce either of the protonated species **3.19** or **3.20**; however, only one of the two reversibly protonated species would cyclize with the appended aromatic ether. This idea proved to be correct, and treatment of **3.18** with acid at room temperature led to amidoalkylation product **3.21** in 55% yield. The authors argued that the appended aromatic nucleophile exists in the equatorial conformation in isomer **3.19** and axial conformation in isomer **3.20**. The acyliminium ion **3.20** can undergo Mannich cyclization due to proper orbital overlap whereas cyclization of **3.19** is geometrically unfeasible. Further elaboration of the aromatic unit in **3.21** afforded lycopodine (**3.1**) in 17 steps with a 1.1% overall yield resulting in the first total chemical synthesis of a member of the *Lycopodium* alkaloids.

Scheme 3.1. Stork Synthesis of (±)-lycopodine (**3.1**).



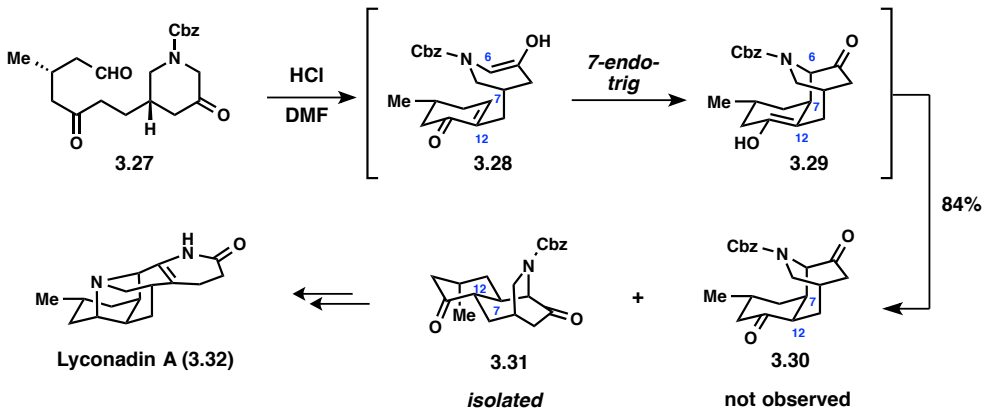
IV. Highlights of Heathcock's Synthesis of (±)-Lycodine: In 1978, Heathcock and coworkers reported a direct approach for the construction of lycodine (**3.2**) which also utilized a Mannich reaction as a key step.[9] Heathcock's efforts began with a conjugate addition of the cuprate derived from the anion of hydrazone **3.22** to afford diketone **3.24** as an equimolar mixture of C12 epimers. It was later shown that the stereochemistry at C12 would be inconsequential during the intramolecular Mannich reaction that formed the core of (**3.2**). Formation of the diketal, followed by reduction of the nitrile provided amine **3.25** in 85% yield. The core of (**3.2**) was then assembled in a single chemical operation. Exposure of **3.25** to aqueous HCl triggered a cascade reaction that involved: ketal deprotection, iminium condensation, and Mannich cyclization to deliver tricycle **3.26**. An additional one-pot procedure yielded Lycodine (**3.2**) in 11 total steps with an overall 3.2% yield. Heathcock's synthesis reiterated that the 1,4-addition reaction proceeds anti to the C16 methyl group and that epimers at C12 are inconsequential. Heathcock's endeavors led to a highly concise synthesis that gave access to the lycodine class of *Lycopodium* alkaloids using conceptually similar bond disconnections to Stork's lycopodine synthesis.

Scheme 3.2. Heathcock's synthesis of (±)-lycodine (**3.2**).



V. Highlights of Smith Synthesis of (+)-Lyconadine A: Since Stork's inaugural synthesis of lycopodine (**3.1**), many other syntheses have been reported and new members of the family have been isolated. Lyconadin A (**3.32**), which belongs to the miscellaneous class, contains an unprecendeted pentacyclic framework with an embedded 7-membered ring.[10] In 2007, Smith and coworkers reported the first total synthesis of lyconadin A (**3.32**).[11] The Smith synthesis featured an impressive cascade reaction that involved: (1) an intramolecular aldol condensation that formed eneone **3.28** and (2) a *7-endo-trig* conjugate addition that forged the embedded 7-membered ring system in (**3.32**) (Scheme 3.3). Unfortunately, while the 7-membered formation proceeded with the desired C7 stereochemistry, protonation of enol **3.29** resulted in the exclusive formation of the undesired *trans*-fused ring **3.31** and not the expected product **3.30**. Stereochemical correction at C12 and further elaboration led to the first total synthesis of lyconadin A.

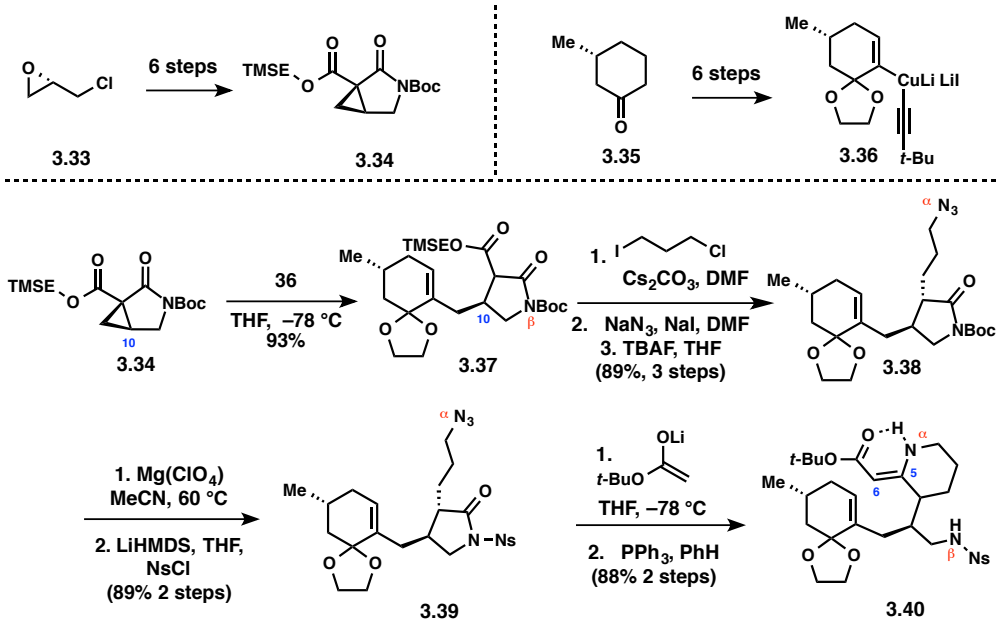
Scheme 3.3. Smith's synthesis of (+)-lyconadine A (**3.27**).



VI. Shair's Synthesis of (+)-Fastigiatine: In 2010, Shair and co-workers reported the first total synthesis of (+)-fastigiatine (**3.47**).[12] The authors devised an elegant approach in 19 steps from commercial epichlorohydrin (**3.33**) or cuprate (**3.36**). Coupling of advanced intermediates **3.34**

and **3.36** introduced the cyclohexane moiety found in the natural product with the correct stereochemistry at C10. Next, the authors installed the appended αN in three steps: (1) alkylation with 1-chloro-3-idopropane, (2) S_N2 displacement of chloride with azide, and (3) decarboxylative cleavage with TBAF to furnish **3.38** in good yields. In four additional steps, the cascade precursor **3.40** that contained the C5–C6 linkage was formed.

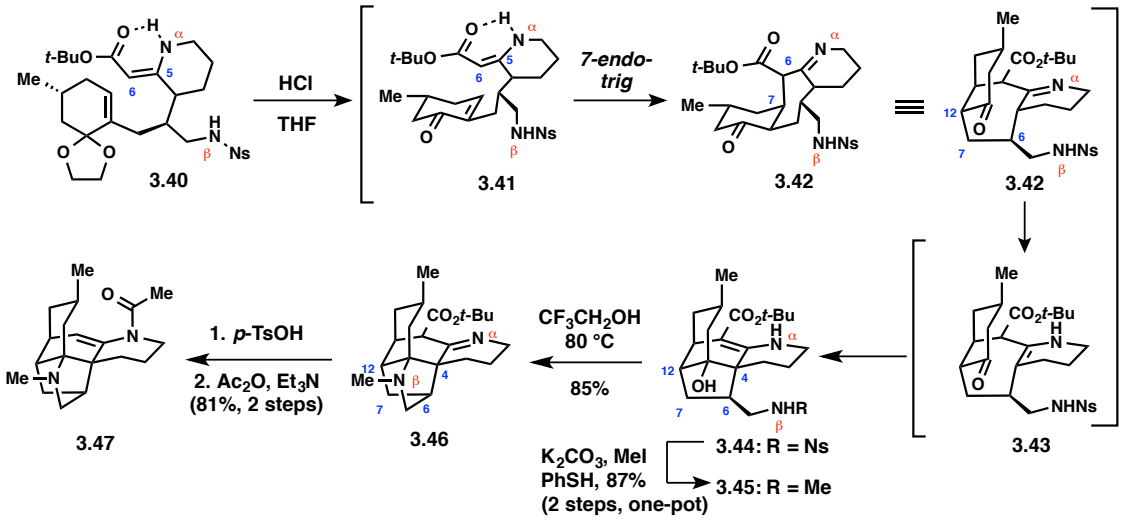
Scheme 3.4. Shair's synthesis of enamine **3.40**.



Utilizing a related strategy to the synthesis of Lyconadin A (**3.32**),[11] Shair and co-workers constructed the 7-membered ring of (**3.47**) (Scheme 3.4). Exposure of **3.40** to aqueous HCl triggered the following tandem reaction sequence: (1) ketal deprotection **3.41**, (2) *7-endo-trig* conjugate addition **3.42** and (3) transannular aldol to produce **3.44**. While the aldol product **3.44** was unexpected, the authors were delighted to find out **3.44** contained the correct connectivity found in fastigiatine. Functionalization of βN produced **3.45**, which underwent a

Mannich reaction upon exposure to elevated temperatures forming **3.46**. Further functional group manipulation afforded fastigiatine (**3.47**) in 15 steps from intermediates **3.34** and **3.36**.

Scheme 3.5. Completion of (+)-fastigiatine (**3.47**).



VII. Conclusion: The aforementioned syntheses embody only a small portion of information pertaining to the *Lycopodium* alkaloids. The seminal work of Stork and Heathcock illustrated the feasibility of intramolecular Mannich reactions in alkaloid synthesis. The contemporary work of Smith and Shair showed effective strategies for the construction of 7-membered ring *Lycopodium* alkaloids. These selected syntheses have inspired the work of others, including our group.

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Chapter 4

Total Synthesis of (+)-Fastigiatine

I. Abstract: A modular approach towards the fastigiatine-type alkaloids is described herein. A concise synthesis of fastigiatine was accomplished using a biomimetic transannular Mannich reaction that generated two quaternary carbons at a late stage. The strategic approach described in this chapter should be applicable to other members of the *Lycopodium* family.

II. Introduction: Fastigiatine (**4.1**) was first isolated from the *Lycopodium Fastigatum* in 1986 by MacLean and coworkers.[1] Its structure was solved by X-ray analysis of its free base, which allowed for the determination of a pentacyclic ring system that contained two fully substituted carbons and six stereogenic centers. Although the biological relevance of (**4.1**) has not been described, the newly isolated congeners (**4.2**)[2] and (**4.4**)[3,4] were described to possess interesting pharmacological properties (Figure 4.1). Himeradine A (**4.4**) contains a scaffold similar to fastigiatine, but features an appended quinolizidine fragment connected via a methylene linker. Biological studies of himeradine A by Kobayashi and coworkers showed modest cytotoxicity against murine lymphoma L1210 cells (IC_{50} = 10 μg/mL). The structural complexity and biological activity of these molecules inspired us to develop a modular approach toward this series of alkaloids.

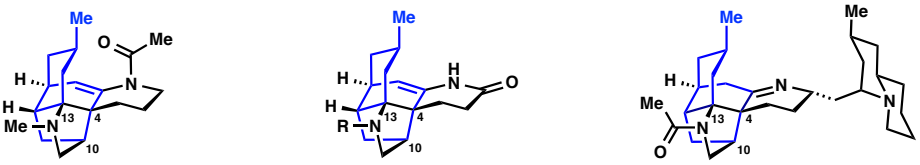


Figure 4.1. Fastigiatine and related alkaloids.

III. Proposed Biosynthesis: The biogenetic origin of (**4.1**) was first proposed by MacLean, and is illustrated in figure 4.2.[1] Tricycle **4.5**, derived from phlegmarine, undergoes an intramolecular Mannich reaction that forges the C4–C13 linkage. An enzyme-mediated oxidation of tetracycle **4.6** at the C10 position installs the necessary leaving group on **4.7** for the subsequent ring formation. MacLean argues that an intramolecular S_N2 reaction stitches together the C4–C10 linkage, thereby forming the core structure of fastigiatine (**4.1**). Inspired by the original work of Heathcock[5] and Shair,[4a] a modular approach towards **4.1** was initiated, with the goal of eventually applying the developed route towards himeradine A and the lyconadin alkaloids. During the course of these studies, Shair and co-workers disclosed the first total synthesis of himeradine A.[4b]

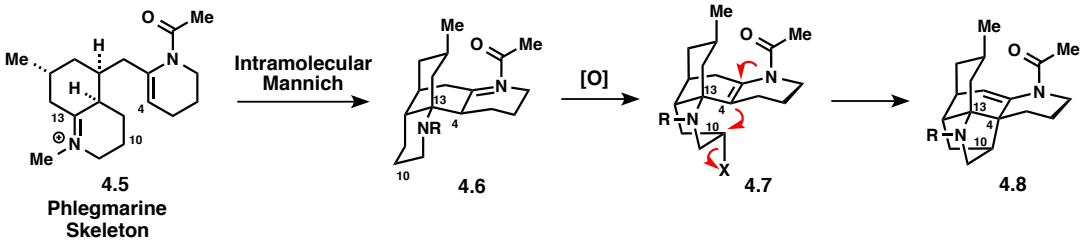


Figure 4.2. MacLean's proposed biosynthesis of fastigiatine.

The overall strategy was designed around a transannular Mannich ring closure proceeding through intermediate **4.9**, which leads to a dramatic simplification of the pentacyclic scaffold as shown in the retrosynthesis plan (Figure 4.3).[6] Diamine **4.10** was envisioned to be accessed from *cis* benzo[7]annulene **4.13** via cross coupling with **4.11,** although more complex fragments could be introduced as part of the himeradine synthesis. Conjugate addition with organometallic **4.12** would then provide precursor **4.10**. Different protecting groups and reaction sequence was considered at this stage, and if necessary, protection of C13 and C15 carbonyls was contemplated. Enone **4.13** contains twelve of the carbon atoms and three of the stereogenic centers in the correct absolute configuration as found in fastigiatine. It also maps onto the core of

the lyconadin and himeradine A targets. The hypothetical compound **4.13** could be prepared by a Diels-Alder reaction and ring expansion from known cyclohexenone **4.14**.

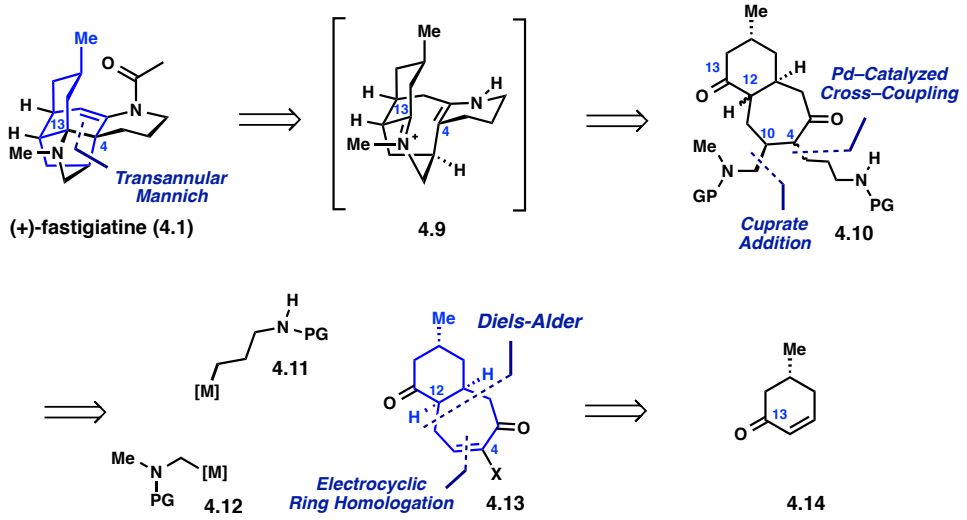
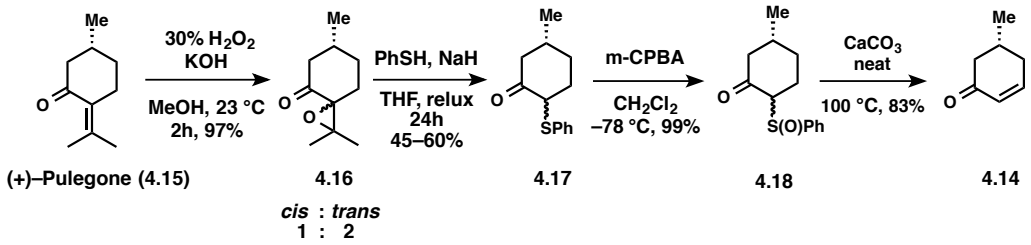


Figure 4.3. Retrosynthesis plan for fastigiatine.

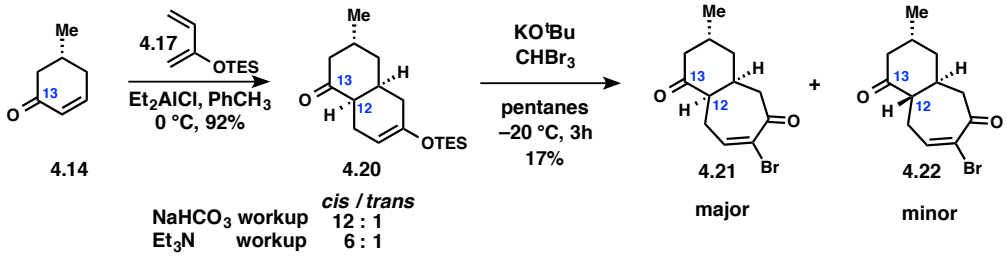
IV. Results and Discussion: Initial work consisted on the preparation of known enone **4.14** using a slightly modified protocol as outlined in Scheme 4.1.[7] The synthesis begins with a large-scale oxidation of commercially available (*R*)-(+)-Pulegone (**4.15**) using hydrogen peroxide to produce epoxide **4.16** in excellent yields.[8] Reaction of pulegone oxide **4.16** and thiophenol in the presence of sodium hydride afforded sulfide **4.17** in 45-60% yield.[7b] The sulfenylation reaction of **4.16** was eventually re-examined by undergraduate student Christina Owens, and while the yields were not significantly improved, the reaction allowed for gram-scale preparation of **4.17**. Oxidation of sulfide **4.17** at low temperature using *m*-CPBA afforded sulfoxide **4.18** in good yields.[7a] Thermal elimination of neat **4.16** in the presence of $CaCO_3$ led to the desired enone **4.14** on gram scale.[9]

Scheme 4.1. Synthesis of cyclohexenone **4.12**.



With the enone building block in hand, a diastereoselective Diels-Alder reaction between **4.14** and TES enol ether **4.19** was conducted.[10] Quenching of the cycloaddition reaction with aqueous $NaHCO_3$ prevented loss of silyl group, to afford decalin **4.20** as a 12:1 ratio of diastereomers favoring the C12 *cis* isomer. It is worth mentioning that a detrimental ratio (~6:1) was observed when using a stronger base such as EtN_3. A ring expansion was next attempted on TES enol ether **4.20** using dibromocarbene, which afforded the unoptimized ring expansion products **4.21** and **4.22** in 17% combined yield.[11] As anticipated due to the basic reaction conditions, tandem equilibration and epimerization at C12 occurred during the ring expansion. These preliminary results suggested the configurational lability at C12 that needed to be addressed before moving forward.

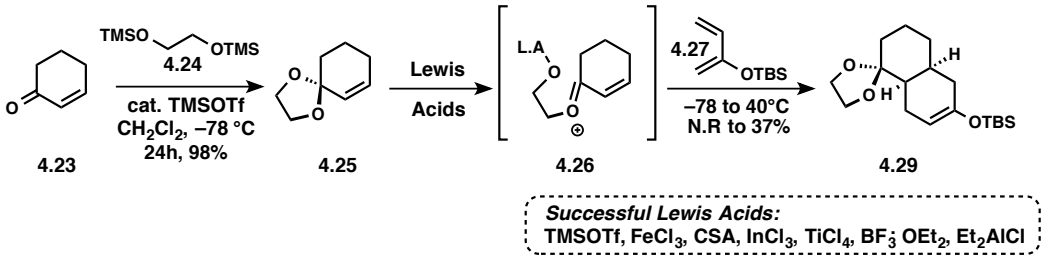
Scheme 4.2. Preparation of bromo enone **4.21**.



It became clear that C13 carbonyl protection was necessary to circumvent undesired C12 epimerization downstream. One approach towards this goal involved an ionic Diels-Alder strategy, rather than the Lewis acid catalyzed Diels-Alder reaction previously described.[12] If

successful, making the C13 carbonyl as a ketal would prevent any undesired epimerization during the subsequent reaction sequence. To test this idea, a model system was developed, using achiral enone **4.23** as an inexpensive building block. Ketalization of **4.23** using Noyori's conditions led to Diels-Alder precursor **4.25** in excellent yield.[13] Cycloaddition of coupling partners **4.25**, and more robust TBS-modified enol ether **4.27**, in the presence of a Lewis acid led to produce **4.29**, proceeding via intermediacy of oxonium ion **4.26**. A variety of conditions were investigated to increase the formation of **4.29**, with unsuccessful results. While this approach was attractive because addition and removal of ketal could proceed in two steps, the low yields obtained for the cycloaddition reaction discouraged us to further pursue this strategy.

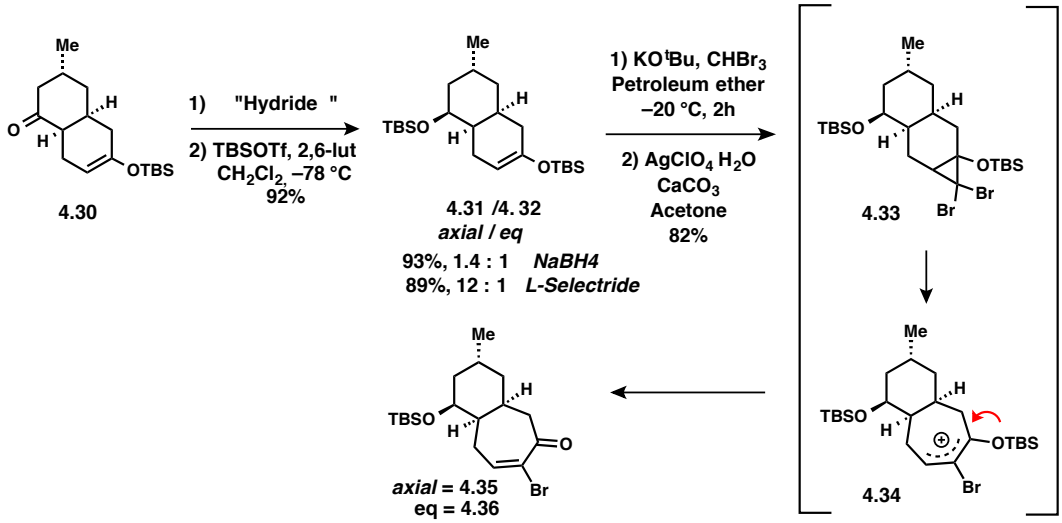
Scheme 4.3. Synthesis of protected decalin **4.29**.



Attention was next turned to a reduction–protection sequence using a metal hydride and a silyl-protecting group (Scheme 4.4). Reduction of TBS modified decalin **4.30** with sodium borohydride produced the corresponding alcohol in a ~1.4:1 mixture favoring the axial product, which was protected as the TBS ether **4.31** using TBSOTf.[14] A diastereoselective reduction that favored the axial product ~12:1 was accomplished using the bulkier reducing agent *L*-selectride. Although the C13 stereochemistry is inconsequential for the downstream cascade, it was decided to prepare both C13 diatereomers to understand their influence in the upcoming key conjugate addition reaction. For brevity, only the axial C13 diastereomer is presented in Scheme 4.4. Ring

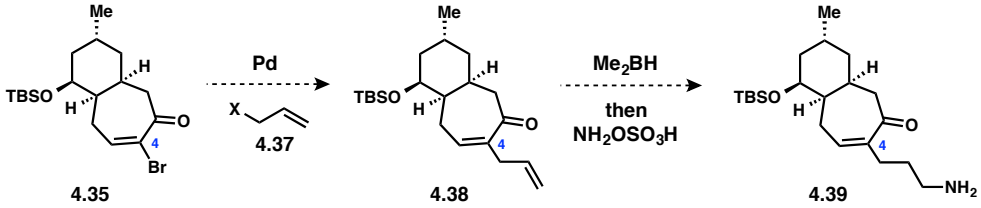
expansion of TBS-ether **4.31** with dibromocarbene followed by exposure to silver (I) salt allowed for the mild formation of bromo enone **4.35** on gram scale.[15]

Scheme 4.4. Preparation of bromoenones **4.35** and **4.36**.



Fastigiatine required the installation of a three-carbon chain onto C4, with an appended terminal nitrogen atom. Towards that goal, an allylation reaction was envisioned to introduce the necessary carbon chain on **4.38**, which would then allow for the hydroamination of the resultant terminal alkene to afford amine **4.39**. This would install the first nitrogen atom present in fastigiatine, although more complex fragments could be introduced as part of the himeradine synthesis (Scheme 4.5).

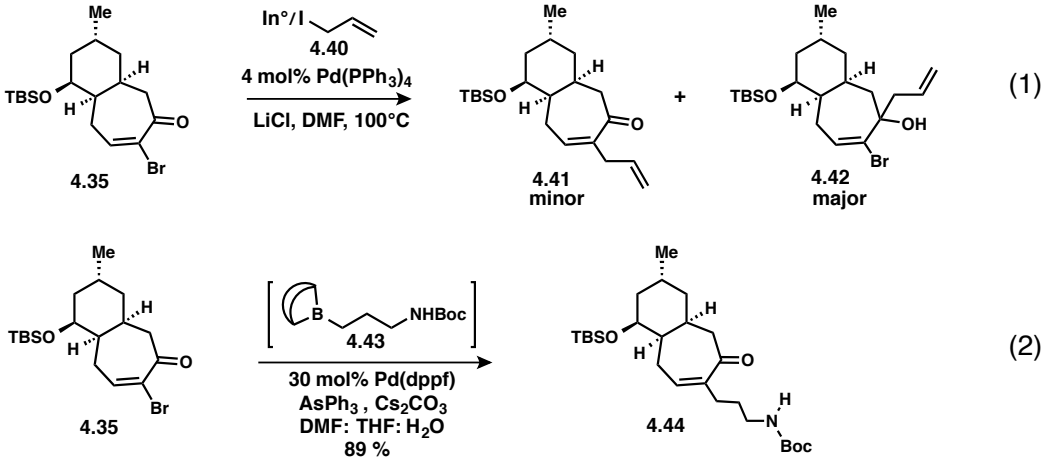
Scheme 4.5. Proposed allylation-hydroamination sequence.



While an allyl cross-coupling onto bromo enone seemed a reasonable approach, no examples were found in the literature involving such groups. I decided to utilize the mild reagent allylindium **4.40** generated *in situ* from allyl iodide and indium metal to effect the desired

transformation.[16] Unfortunately, a competing 1,2-addition pathway led to alcohol **4.42** as the major product on several occasions (Scheme 4.6, Eq. 1). In order to suppress the ketone addition, various conditions were investigated including portion-wise addition of palladium, allylindium generation protocols, and different temperatures, but the reaction remained problematic and unreliable. As an alternative, sp^2-sp^3 Suzuki coupling with a protected allylamine to introduce the three-carbon chain was simultaneously investigated (Scheme 4.6, Eq. 2).[17] Fortunately, coupling of bromo enone **4.35** with the borane **4.43** derived from *N*-Boc allylamine allowed for the direct installation of the three-carbon chain, to afford amide **4.44** in high yields and gram-quantities.

Scheme 4. 6. Cross-coupling reaction of bromo enone **4.35**.

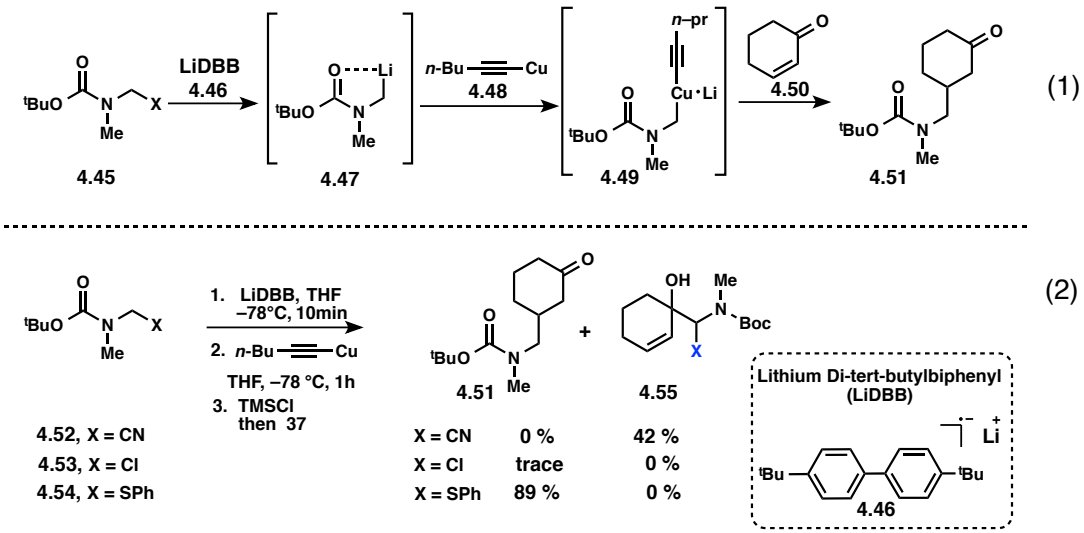


A key step in the synthesis of fastigiatine involved a conjugate addition of a methylene amine building block at the beta position of enone **4.44**. Several alternatives were considered, including reagents as simple as diethylaluminum cyanide, but most of them would require extensive manipulation after the addition.[18] The development of a new, more elaborated reagent would permit the installation of a functionalized synthon **4.49**, obviating circuitous chemical operations downstream (Scheme 4.7, Eq. 1). With this idea in mind, pronucleophile **4.45** would undergo reductive lithiation to produce alkyl lithium **4.47**;[19] subsequent treatment with cuprous

acetylide **4.48** would generate mixed cuprate **4.49**,[20] which could then engage in a conjugate addition.

Investigations began with nitrile **4.52** and model substrate **4.50** (Scheme 4.7, Eq. 2).[21] Nitrile **4.52** did not afford the desired product, but instead underwent a 1,2-addition reaction to produce alcohol **4.55**. This suggested that reductive decyanation is not facile with primary nitriles, as alpha deprotonation occurs first, precluding nitrile cleavage and therefore undergoes a 1,2-addition reaction. The 1,4-addition reaction, however, proceeded when alkyl chloride **4.53** was used, albeit in low yields. This suggested its potential towards the formation of **4.51**.[22] Unfortunately, carbamate **4.53** proved to be quite unstable to air and handling, therefore preventing us from its further use. In the search of a bench-stable reagent that could still participate in the required chemistry, I elected to investigate sulfide **4.54**.[23] Gratifyingly, the thioether **4.54** performed remarkably, producing compound **4.51** in great yields.

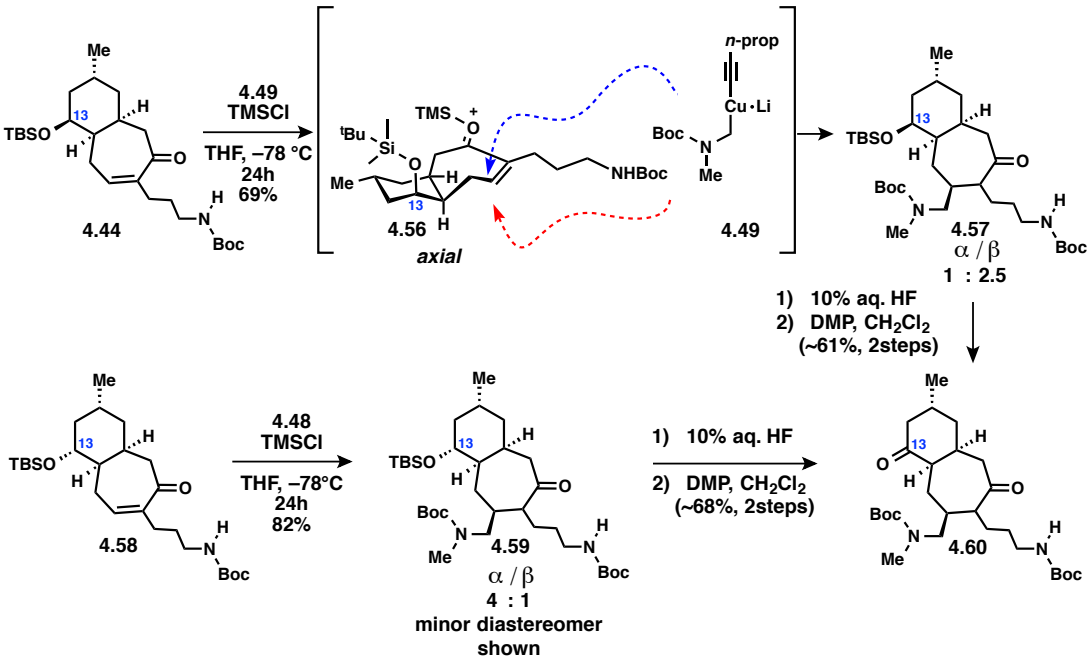
Scheme 4.7. Development of cuprate reagent **4.49**.



With methylene synthon **4.54** in hand, the conjugate addition reaction was tested on C13 stereoisomers **4.44** and **4.58** (Scheme 4.8). Two plausible scenarios were anticipated for the addition: 1) attack could proceed from either top or bottom face of the acceptor, *e.g.*, structure

4.56; and 2) the C13 substituent might influence the stereochemical outcome of the reaction. It was soon discovered that axial TBS ether **4.44** led to dicarbamate **4.57**, favoring the desired beta adduct (~1:2.5), whereas equatorial **4.58** gave product **4.59** with reversed selectivity (~4:1). While the reactions proceeded in high yields, their diastereoselectivities remained moderate. Equally important was the identification of **4.44** as a better substrate for the desired beta attack transformation. At this junction, the factors controlling the selectivity remain unclear, and further experimentation should address these questions. A deprotection and oxidation of compounds **4.57** and **4.59** led to cascade precursor **4.60** in 61% and 68% yield, respectively.

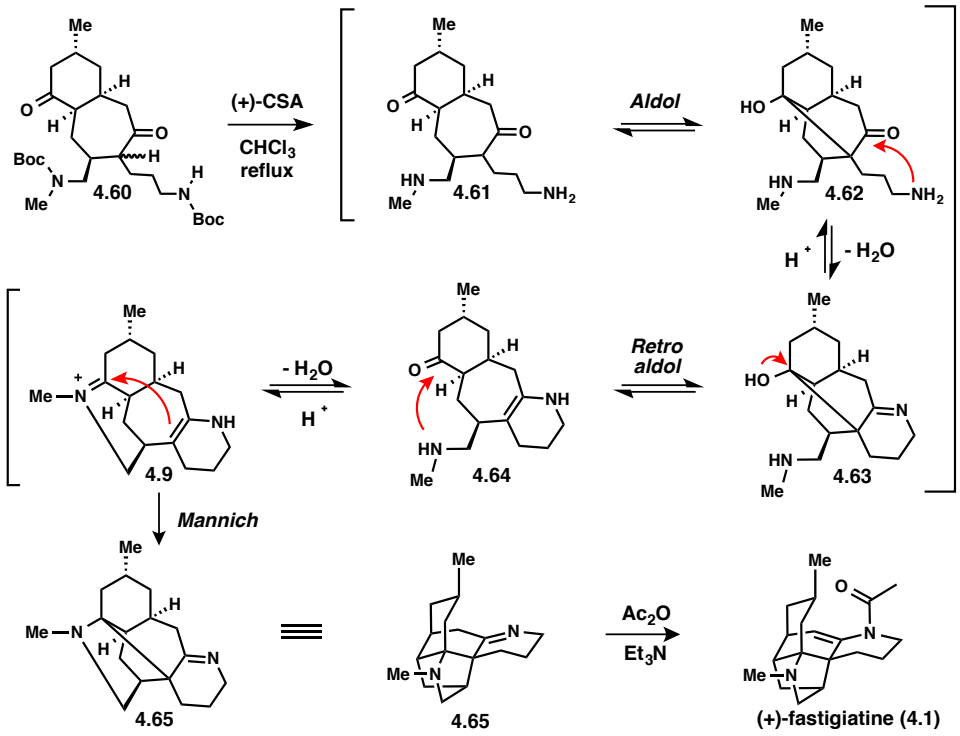
Scheme 4.8. Conjugate addition of mixed cuprate **4.49**.



The overall strategy for synthesizing fastigiatine was designed around a transannular Mannich ring closure to assemble the core of the natural product **4.65** as illustrated in Scheme 4.9. This one-step transformation would involve deprotection of both Boc groups in **4.60** with CSA to induce a series of intramolecular cascade reactions. Upon deprotection, the free amines in **4.61** were thought to occur in equilibrium between the diketone **4.61** and aldol product **4.62**.

Imine formation **4.63** followed by a retro-aldol reaction would allow for iminium condensation **4.9** setting the stage for the transannular Mannich reaction to form the core **4.65**. The sequence of events may occur in a different fashion, but Scheme 4.9 provides an overview of the assembly and follows precedent of Shair's work.[4]

Scheme 4.9. Cascade cyclization of fastigiatine (**4.1**).

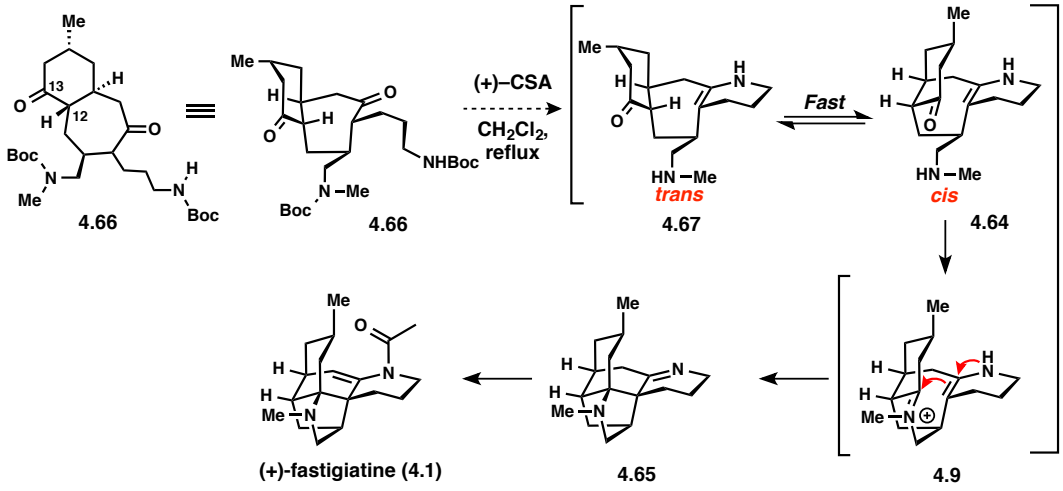


Initial studies on the deprotection of **4.60** utilized TFA, which was then treated with CF_3CH_2OH to afford trace formation of **4.65**. Switching to (+)-CSA at elevated temperatures however, permitted the formation of **4.65**.[24] Acylation of the crude reaction mixture produced fastigiatine in excellent yields.

With a completed synthesis of (**4.1**), an improved second-generation route was realized which would obviate extensive C13 manipulation sequences. Stereochemical erosion at C12 was observed in the early stages of synthesis. This may have led to undesired *trans*-6,7–fused isomer **4.66** which possesses the wrong geometry for the cyclization. However, **4.66** could undergo a

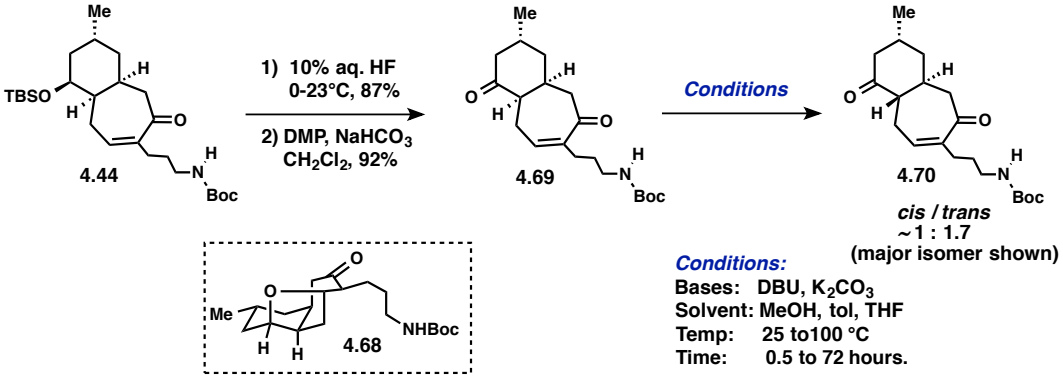
series of rearrangements to afford the natural product (**4.1**) as shown in Scheme 4.10. Upon deprotection, diketone **4.66** would cyclize to enamine **4.67**, which could equilibrate *in-situ* to *cis*-isomer **4.64** and participate in the cascade previously described.[25]

Scheme 4.10. Alternative cascade toward fastigatine.



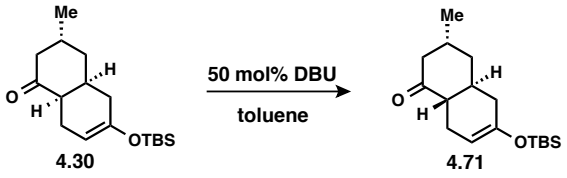
V. C12 epimerization Studies: To verify whether or not **4.66** could participate in the formation of (**4.1**), preparation of *trans* **4.70** was next pursued. Carbamate **4.70** would then be converted to **4.66** via conjugate addition. Towards this end, desilylation of **4.44** with aqueous HF followed by a mild oxidation gave diketone **4.69**. It is worth noting that attempts at deprotection of **4.44** under basic TBAF conditions led to the exclusive formation of oxa-michael product **4.68**, while buffering the basicity of TBAF using AcOH or H_2O additives afforded no deprotection. Next, C12 epimerization of **4.69** was conducted utilizing a variety of conditions. In all cases, product **4.70** was isolated as an inseparable thermodynamic mixture with its *cis* isomer (~1:1.7). Resubjecting the mixture to the same conditions led to unchanged ratios suggesting plausible thermodynamic equilibria.

Scheme 4.11. Epimerization studies of benzo[7]annulene.



In contrast, reaction of **4.30** with DBU at high temperatures yielded **4.71** in a 3:1 ratio favoring the *trans* isomer, which is readily isolable. Initially, the epimerization proceeded slowly at ambient temperatures (Table 4.1, entry 1), however heating with a microwave reactor afforded **4.71** in 1 hour (Table 4.1, entry 3). Control experiments with longer reaction times or other bases did not increase formation of **4.71**, nor was decomposition observed under the described conditions.

Table 4.1. Epimerization of decalin **4.30**.

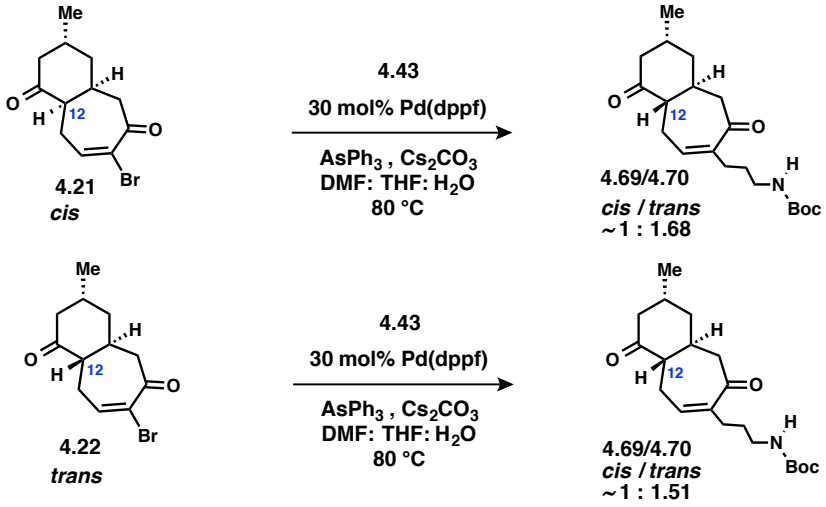


entry	temperature	time	*cis* : *trans*
1	25 °C	7 d	1 : 1.3
2	100 °C	35 h	1 : 2.75
3	190 °C, μwaves	1 h	1 : 3.0

Concurrent to the isomerization studies, a Suzuki coupling of diastereomeric *cis* **4.21** and *trans* **4.22** was conducted (Scheme 4.12). As reminiscent of before, the ratio of the resultant Suzuki products were similar to those of the epimerization studies observed in Scheme

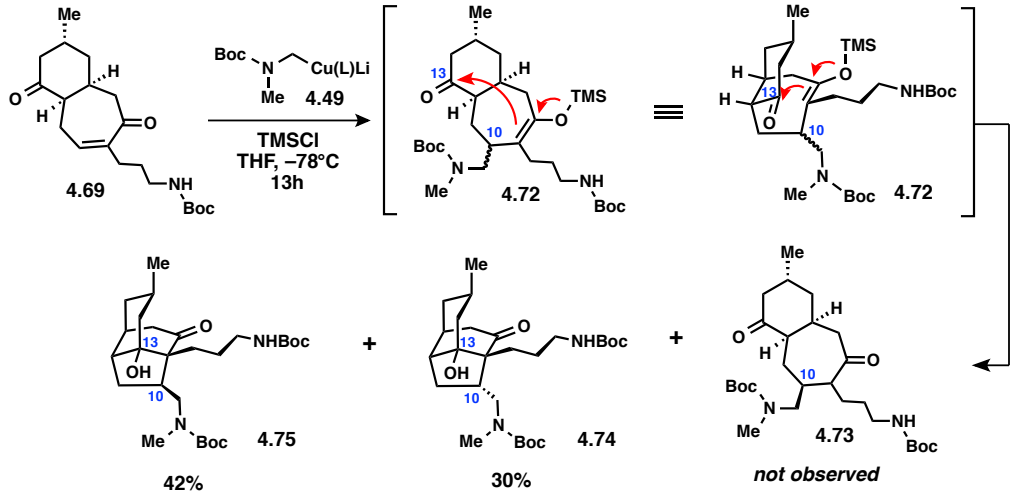
4.11. While it is clear that decalin **4.30** could be driven towards *trans* **4.71** upon epimerization, this situation became more complex for benzo[7]annulene **4.69**, as diastereomeric **4.70** (1:1.5) always resulted from a thermodynamically controlled process. Furthermore, the C13 unprotected synthetic sequence poses a serious concern, since the desired *cis* fusion continuously erodes, and generation of epimeric **4.69** and **4.70** cannot be separated nor equilibrated to a single C12 diastereomer.

Scheme 4.12. Suzuki coupling on diketones.



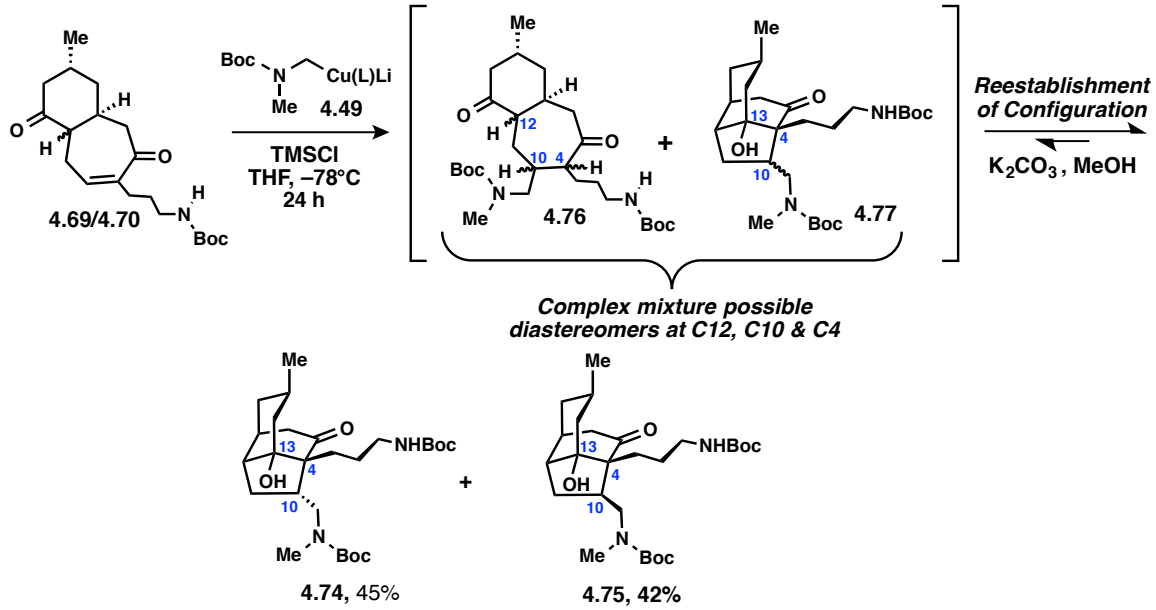
The next step involved addition into diastereomeric **4.69**/**4.70**, however I opted to perform the addition using single diastereomer *cis* **4.69** and attempt epimerization in the subsequent step (Scheme 4.13). At first glance, the addition proceeded well, but further NMR inspection of the isolated products **4.74** and **4.75** revealed the disappearance of the C13 carbonyl group, and generation of two quaternary carbons. This transformation presumably occurs via conjugate addition and concomitant transannular aldol of **4.72**, producing tricycle **4.75** along with its C10 epimer in 42% and 30% yield, respectively. While **4.74** could not be recycled, the major tricycle **4.75** contained the correct absolute stereochemistry and could be carried on to synthesize fastigiatine.

Scheme 4.13. Generation of tricycle products **4.74** and **4.75**.



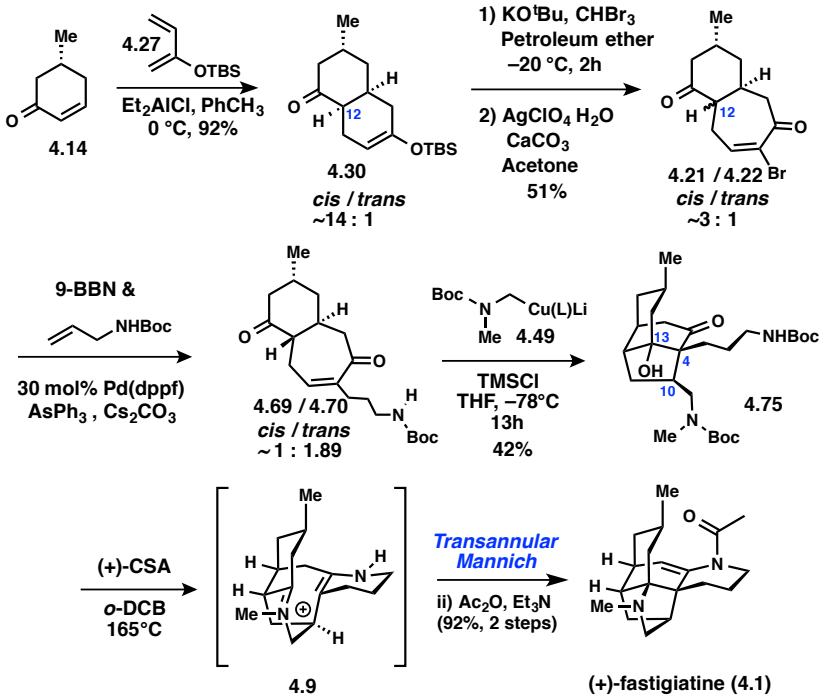
The conjugate addition was next performed on an inseparable mixture of **4.69** and **4.70**, which gave a complex mixture with possible epimers at C4, C10 and C12 (Scheme 4.14). Prof Rychnovsky suggested a base-mediated equilibration upon completion of the reaction to produce tricycles **4.74** and **4.75** via epimerization and transannular aldol reaction. With this idea in mind, after painstaking isolation of various TLC spots, treatment of the crude conjugate addition reaction mixture with K_2CO_3 in methanol allowed for convergence of the isolated products to tricycles **4.74** and **4.75**. Exploiting a transannular aldol reaction between C4 and C13 at a late-stage reestablished the absolute configuration, simplifying the otherwise unwieldy epimeric mixture.

Scheme 4.14. Conjugate addition-transannular aldol reaction.



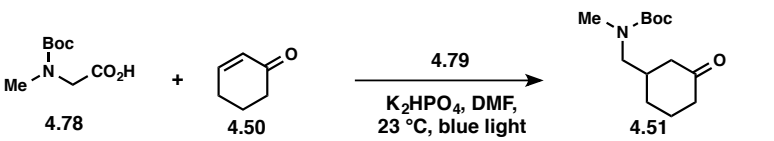
VI. A Six-Step sequence to fastigiatine: With a good understanding of the C13 protecting group free route, the total synthesis of fastigiatine was accomplished in six steps (Scheme 4.15). Diels-Alder reaction of **4.14** and **4.27** gave decalin **4.30** as a 14:1 ratio favoring the *cis* isomer. Dibromocarbene ring expansion produced bromo enones **4.21** and **4.22** in a 3:1 ratio favoring the *cis* isomer. Suzuki coupling led to a thermodynamic mixture of **4.69** and **4.70** in excellent yield. Conjugate addition led to a complex mixture that was simplified by working up the reaction with K_2CO_3 and methanol to afford tricycle **4.75**, isolated as ca. 1:1 mixture with its C10 epimer in high yield. Compound **4.75** contains the correct C10 configuration needed for the Mannich cyclization and can be taken forward directly to the cascade cyclization. Treatment of **4.75** with CSA in *o*-DCB removed the two Boc protecting groups, and set up a retro-aldol equilibrium that permitted the formation of intermediate **4.9** en route to a transannular Mannich reaction. Acylation of the crude reaction mixture produced fastigiatine (**4.1**) in excellent yield.

Scheme 4.15. A six-step sequence to fastigiatine (**4.1**)

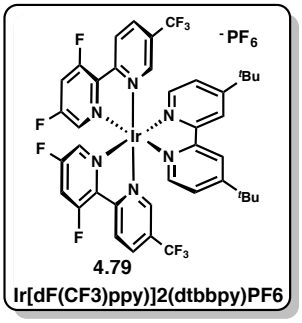


VII. Attempts at Improving the Conjugate Addition: During the course of our studies, MacMillan and coworkers reported the photon-induced decarboxylation of α-amino acids to produce Michael donors that participated in conjugate addition reactions.[26,27] Inspired by these new protocols, we set out to explore the photoredox coupling of N-Boc amino acid **4.78** into model acceptor **4.50** using photocatalyst **4.79** with visible light (Scheme 4.17). After reaction optimization, the addition underwent with operational ease and in excellent yield.

Table 4.2. Decarboxylative conjugate addition.

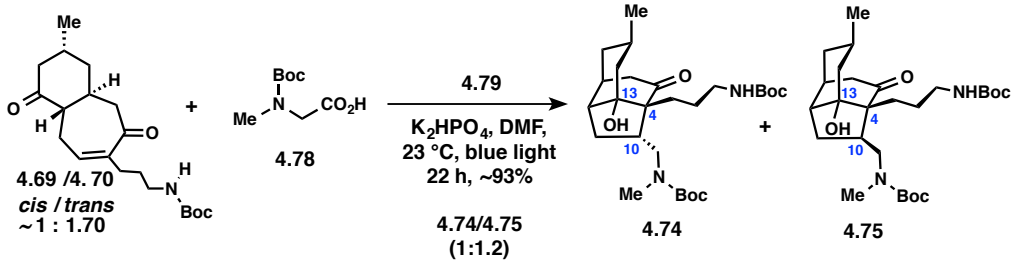


entry	78 (equiv)	50 (equiv)	79 (cat.)	K_2HPO_4 (equiv)	time	Yield % (isolated)
1	1.0	1.0 (25 mg)	1 mol% (~ 2.8 mg)	1.2	48 h	36 %
2	1.5	1.0	4 mol% (~11 mg)	1.7	24 h	96 %



With an alternative protocol to install the requisite methylene amine fragment of fastigiatine, the addition was next performed onto a mixture of **4.69** and **4.70**. Preliminary results demonstrated high yields for the reaction, however the selectivity remained moderate when applied to a diastereomeric mixture of **4.69**/**4.70**. Further applications of the photoredox coupling to the *Lycopodium* alkaloids are underway and will be reported in due time.

Scheme 4.17. Decarboxylative conjugate addition into diastereomeric **4.69** /**4.70**.



VIII. Conclusions on the Total Synthesis of fastigiatine

In conclusion, a new synthesis of fastigiatine was accomplished by two different methods. The first approach employed protection of C13 carbonyl group to avoid isomerization. A second-generation route proved challenging, as C12 stereochemistry underwent configurationally changes, but a transannular aldol reaction corrected the configuration at a late-stage, obviating the need for protecting groups. A novel phenylthio carbamate reagent for

conjugate additions was developed and applied to the synthesis of (**4.1**). Further investigations utilized photoredox chemistry to incorporate a glycine derivative as an alternative approach of the "methylene amine" synthon. The strategic approach developed for fastigiatine is currently being expanded to himeradine A and the lyconadin alkaloids. The landmark syntheses by Heathcock and Shair served as groundwork for our investigations.

IX. General experimental and laboratory conditions

All glassware was flame- or oven-dried and cooled under argon unless otherwise stated. All reactions and solutions were conducted under argon unless otherwise stated. All commercially available reagents were used as received, unless otherwise stated. Toluene (PhMe), tetrahydrofuran (THF), dimethylformamide (DMF), diethyl ether (Et_2O) and dichloromethane (CH_2Cl_2) were degassed and dried by filtration through activated alumina under vacuum according to the procedure by Grubbs.[28] Diisopropylamine (DIPA), acetonitrile (MeCN), 1,3-Dimethyl-3,4,5,6-tetrahydro-2-pyrimidinone (DMPU) were distilled from CaH_2 prior to use. All reactions involving LiDBB were conducted with glass stirbars. Thin layer chromatography (TLC) was performed with Millipore 60 F_{254} glass-backed silica gel plates and visualized using potassium permanganate, Dragendorff-Munier, ceric ammonium molybdate (CAM) or vanillin stains. Flash column chromatography was performed according to the method by Still, Kahn, and Mitra[29] using Millipore Geduran Silica 60 (40-63 μm).

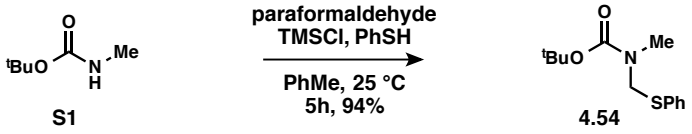
Instrumentation

All data collected at ambient temperature unless noted. ^{1}H NMR spectra were taken at 500 or 600 MHz, calibrated using residual NMR solvent or TMS and interpreted on the δ scale. Peak abbreviations are listed: s = singlet, d = doublet, t = triplet, q = quartet, pent = pentet, dd = doublet of doublets, ddd = doublet of doublet of doublets dt = doublet of triplets, ddt = doublet of doublet of triplets, dq = doublet of quartets, m = multiplet, app = apparent, br = broad. ^{13}C NMR spectra were taken at 125 MHz, calibrated using the NMR solvent, and interpreted on the δ scale.

Some samples were analyzed above room temperature to minimize line broadening due to rotamers.

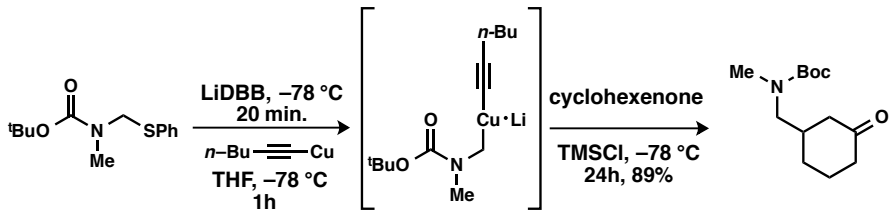
General procedure for preparation of LiDBB stock solution.

A round-bottom flask equipped with a glass stir bar was charged with 4,4'-di-*tert*-butylbiphenyl (1 equiv) and the flask was flame-dried under vacuum until 4,4'-di-*tert*-butylbiphenyl melted, at which point it was cooled to room temperature under argon. Lithium wire (10 equiv) was clipped in a stream of argon. Dry THF (0.5 M) was added and the solution stirred to give a dark green solution within 2-3 min. The mixture was cooled to 0 °C and stirred for 5 h to produce lithium di-*tert*-butylbiphenyl (LiDBB) at full molarity.



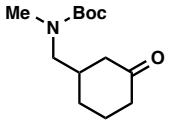
***tert*-butyl methyl((phenylthio)methyl)carbamate (4.54):** Dry toluene (149 mL) was added to a 500 mL round-bottom flask containing *tert*–butyl methyl carbamate **S1** (5.86 g, 0.044 mol, 1 equiv), paraformaldehyde (1.55 g, 0.051 mol, 1.15 equiv) and magnesium sulfate (15 g) at room temperature. After 5 min, TMSCl (16.9 mL, 0.134 mol, 3 equiv) was added dropwise via syringe. The solution was allowed to stir for 15 min and then thiophenol (5.07 mL, 0.025 mol, 1.1 equiv) was added, and the resulting mixture was allowed to stir until starting material was consumed as observed by TLC. After 5 h, the crude reaction mixture was filtered, concentrated and purified via chromatography (15% EtOAc in hexanes) to afford product **4.54** (10.62 g, 94%) as a crystalline white solid. **^{1}H NMR** (500 MHz, $CDCl_3$, 65 °C) δ 7.49 (d, J = 7.0 Hz, 1H), 7.31–7.20 (m, 3H), 4.75 (s, 2H), 2.90 (s, 3H), 1.33 (s, 9H); **^{13}C NMR** (125 MHz, $CDCl_3$, 65 °C)

δ 155.0, 134.5, 133.4, 129.0, 127.6, 80.2, 55.5, 33.3, 28.3; **IR** (thin film) 2972, 2929, 1699, 1478, 1443, 1389, 1265, 1230, 1172, 1133, 1052, 869, 745 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{13}H_{19}NO_2SNa$ $(M + Na)^+$: 276.1034, found: 276.1029. **mp** = 60–63 °C; TLC (20% EtOAc in hexanes) $\mathbf{R}_f$ = 0.42 ($KMnO_4$ stain).



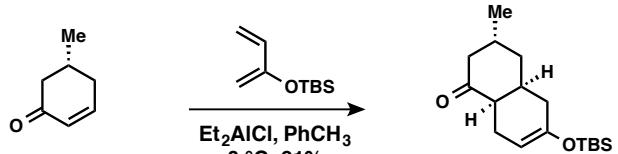
***tert*-butyl methyl((3-oxocyclohexyl)methyl)carbamate (4.51):** A round bottom flask containing **4.54** (211 mg, 0.83 mmol) and 1,10-phenanthroline (2-3 crystals) was dried by azeotroping three times with freshly distilled benzene. The flask was then equipped with a glass stir bar and THF (15 mL) was introduced under Ar. The mixture was cooled to –78 °C and *n*–BuLi/hexanes (2-3 M) was added until a brown dark color persisted (~0.3–0.4 mL). This procedure was performed to quench adventitious proton sources. LiDBB (4.7 mL, 1.86 mmol, 2.2 equiv) was then added dropwise over 10 min at –78 °C until a dark-green color persisted, and the mixture was allowed to stir for 20 min. A separate flask containing 1-hexynyl copper (240 mg, 1.67 mmol) and tetrahydrofuran (3 mL) was cooled to –78 °C and trimethyl phosphite (0.44 mL, 3.75 mmol) was introduced; the mixture was stirred until a clear solution developed. The resulting homogeneous solution was added via syringe to the organolithium reagent down the flask wall over 3 min and stirring was continued for 1 h to produce 15 as deep red solution. The cyclohexenone (40 mg, 0.42 mmol) was added as a solution in THF (0.3 mL) with freshly distilled TMSCl (263 μL, 2.08 mmol). The resulting mixture was stirred at –78 °C for 24 h and quenched with 10% concentrated ammonium hydroxide/saturated ammonium chloride (20 mL),

followed by warming to room temperature. After 1 h, the organic layers were separated and the aqueous layers were extracted with ethyl acetate (20 mL) three times. The organic layers were combined, dried and concentrated under vacuum. The resulting mixture was filtered through a plug silica with 20% CH_2Cl_2 in hexanes to remove excess of 4,4'-di-*tert*-butylbiphenyl, at which point ethyl acetate was used to flushed the plug. The material was concentrated under vacuum. The mixture was concentrated, loaded onto silica gel with DCM and purified by column chromatography, eluting with 25% EtOAc/hexanes gradient, to afford **4.51** (89.2 mg, 89%).



4.51

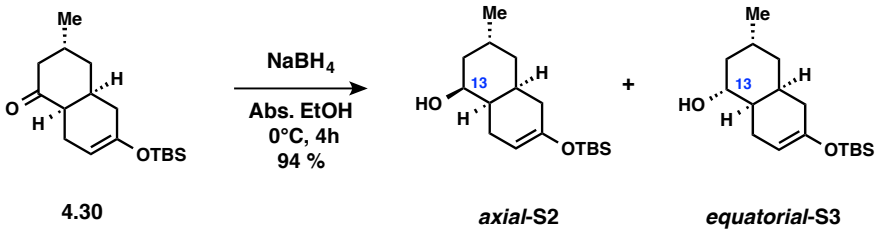
^{1}H NMR (500 MHz, $CDCl_3$) δ 3.24–3.03 (m, 2H), 2.84–2.74 (m, 3H), 2.39–2.28 (m, 2H), 2.27–2.18 (m, 1H), 2.11–1.90 (m, 3H), 2.67 (s, 3H), 1.88–1.75 (m, 1H), 1.67–1.53 (m, 1H), 1.40 (s, 9H), 1.38–1.27 (m, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 211.3, 210.9, 156.2, 155.8, 79.8, 79.6, 54.5, 53.9, 45.8, 45.7, 41.5, 38.5, 38.1, 35.2, 29.1, 28.5, 25.3, 25.2; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{13}H_{23}NO_3Na$ $(M + Na)^+$: 264.1576, found: 264.1572; **TLC** (25 % EtOAc in Hexanes) $\mathbf{R_f}$ = 0.33 (CAM stain). Spectral data matched those reported in the literature.[30]



4.14 **4.30**

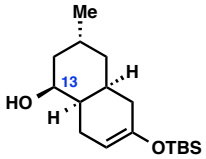
(3*R*,4a*S*,8a*R*)-6-((*tert*-butyldimethylsilyl)oxy)-3-methyl-3,4,4a,5,8,8a-hexahydronaphthalen-1(2*H*)-one (4.30): A round-bottom flask was charged with (+)-5-methylcyclohex-2-en-1-one (2.01 g, 18.26 mmol) and 2-*tert*-butyldimethylsiloxy-1,3-butadiene (4.68 g, 25.44 mmol) and

purged 4 times via vacuum/argon cycles. Dry toluene (75 mL) was added and the solution was cooled to 0 °C. Diethyl aluminum chloride (19.1 mL, 1.0 M in toluene 19.1 mmol) was then added dropwise over a 10 min period. The resulting mixture was allowed to reach room temperature with stirring. After 1.5 h, the mixture was cooled to 0 °C and the reaction was quenched by addition of saturated $NaHCO_3$ (250 mL) and 10% potassium sodium tartrate (20 mL). The aqueous layer was separated and extracted with Et_2O (3 x 200 mL). The combined organic layers were washed with saturated $NaHCO_3$ (3 x 200 mL), brine (3 x 200 mL), dried over Na_2SO_4 and concentrated under reduced pressure. Volatile materials were removed under high vacuum (ca. 1 Torr) overnight to afford the desired product **4.30** (4.91 g, 91%) as light yellow oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 4.81–4.78 (app. m, 1H), 2.61 (t, J = 5.8 Hz, 1H), 2.57–2.48 (m, 2H), 2.39 (ddd, J = 13.5, 4.8, 2.0 Hz, 1H), 2.25–2.17 (m, 1H), 2.20–1.95 (m, 2H), 1.86 (app. dd, J = 8.0, 1.5 Hz, 2H), 1.80 (d, J = 14.5 Hz, 1H), 1.64 (ddd, J = 13.5, 11.5, 4.0 Hz, 1H), 1.03 (d, J = 6 Hz, 3H), 0.89 (s, 9H), 0.11 (s, 3H), 0.08 (s, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 211.1, 148.2, 102.0, 49.7, 47.0, 38.2, 36.1, 31.9, 30.8, 25.9, 22.6, 22.2, 18.1, -4.1, -4.4; **HRMS** (ESI/methanol) m / z calcd for $C_{17}H_{31}O_2Si$ $(M + H)^+$: 295.2093, found: 295.2095. TLC (10% EtOAc in hexanes) $\mathbf{R}_f$ = 0.60 (CAM Stain). Spectral data were consisted with those reported in the literature.[10]



4.30 *axial*-S2 *equatorial*-S3

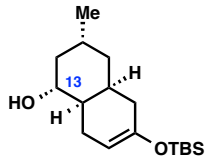
(3*R*,4a*S*,8a*R*)-6-((*tert*-butyldimethylsilyl)oxy)-3-methyl-1,2,3,4,4a,5,8,8a-octahydronaphthalen-1-ol (S2) and (S3): To a solution of decalin **4.30** (2.95 g, 10.03 mmol) in absolute ethanol (33 mL, 0.3 M) at 0 °C was added sodium borohydride (1.89 g, 50.13 mmol) in three portions over 30 minutes. Upon completion as observed by TLC, the reaction mixture was partitioned between EtOAc (50 mL) and H_2O (50 mL) and allowed to reach room temperature. The organic layer was separated and the aqueous layer was extracted with EtOAc (3 x 50 mL). The combined organic layers were washed with brine, dried over Na_2SO_4 and concentrated *in vacuo* to give a colorless oil. Purification by column chromatography (10% EtOAc in hexanes) afforded a mixture of separable diastereomers **S2** and **S3** (2.79 g, 94%) as yellow oil (~1.4:1 *ax*/*eq* mixture of C13 epimers).



axial-S2

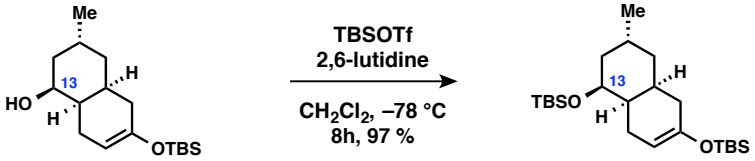
^{1}H NMR (500 MHz, $CDCl_3$) δ 4.86 (app. s, 1H), 3.99 (app. s, 1H), 2.41–2.27 (m, 1H), 2.22–2.13 (m, 1H), 2.12–2.01 (m, 4H), 1.98–1.87 (m, 1H), 1.85–1.73 (m, 1H), 1.65–1.67 (m, 1H), 1.27–1.12 (m, 1H), 0.94 (d, J = 6.5 Hz, 3H), 0.91 (s, 9H), 0.12 (d, J = 5.1 Hz, 6H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 151.8, 102.1, 72.8, 39.7, 37.2, 34.9, 31.0, 26.0, 21.6, 18.2, -4.1, -4.2;

IR (thin film) 3393, 2926, 2856, 1674, 1462, 1378, 1250, 1192, 1176, 1084, 1013, 881, 834, 777, 679 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{17}H_{32}O_2SiNa$ $(M + Na)^+$: 319.2069, found: 319.2061; **TLC** (10% EtOAc in hexanes) $\mathbf{R}_f = 0.41$ (CAM Stain); $[\alpha]^{24}_D = +29$ (*c* 2.94, $CHCl_3$).



***equatorial*-S3**

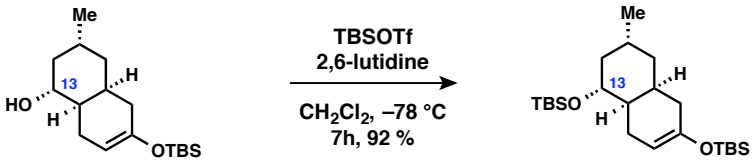
^{1}H NMR (500 MHz, $CDCl_3$) δ 4.77 (app. d, *J* = 5.7 Hz, 1H), 3.57 (ddd, *J* = 15.2, 10.6, 4.5 Hz, 1H), 2.42 (dd, *J* = 17.6, 5.8 Hz, 1H), 2.23–2.16 (m, 1H), 2.15–2.08 (m, 1H), 1.98 (app. d, *J* = 11.6 Hz, 2 H), 1.86–1.76 (m, 2H), 1.52 (d, *J* = 14.0, 2H), 1.45 (ddd, *J* = 10.9, 5.4 Hz, 1H), 1.31–1.24 (br. s, 1H), 1.18 (td, *J* = 13.3, 4.8 Hz, 1H), 0.93 (d, *J* = 6.6 Hz, 3H), 0.91 (s, 9H), -0.12 (d, *J* = 5.8 Hz, 6H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 141.1, 101.7, 67.7, 54.1, 45.1, 41.8, 39.1, 33.7, 31.7, 26.6, 25.9, 24.2, 22.5, 18.2, -4.1, -4.3; **IR** (thin film) 3373, 2926, 1701, 1666, 1513, 1463, 1365, 1250, 1171, 1103, 1058, 835, 774 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{17}H_{32}O_2SiNa$ $(M + Na)^+$: 319.2069, found: 319.2076; **TLC** (10% EtOAc in hexanes) $\mathbf{R}_f = 0.31$ (CAM Stain); $[\alpha]^{25}_D = -4.0$ (*c* 1.57, $CHCl_3$).



***axial*-S2** **4.31**

(((1*S*,3*R*,4a*S*,8a*R*)-3-methyl-1,2,3,4,4a,5,8,8a-octahydronaphthalene-1,6-diyl)bis(oxy))bis(*tert*-butyldimethylsilane) (4.31): To a solution of **S2** (1.48 g, 5.0 mmol) in CH_2Cl_2 (7.1 mL) at –78 °C was added 2,6-lutidine (1.16 mL, 9.98 mmol, 2 equiv) and TBSOTf

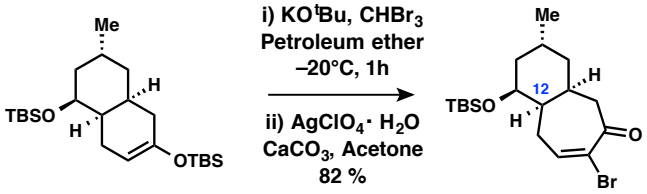
(1.38 mL, 5.99 mmol, 1.2 equiv). The mixture was stirred for 8 h, and the reaction was quenched with Et_3N (0.84 mL, 5.99 mmol, 1.2 equiv) and $NaHCO_3$ (15 mL) at –78 °C. The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 x 15 mL). The combined organic layers were washed with brine, dried over Na_2SO_4 and concentrated *in vacuo* to give a yellow oil. Purification by column chromatography (5% CH_2Cl_2 in hexanes) gave product **4.31** (1.98 g, 97%) as a clear oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 4.77 (s, 1H), 3.94 (dt, *J* = 8.9, 4.2, 1H), 2.23–2.12 (m, 1H), 2.09–1.98 (m, 4H), 1.88–1.78 (br. s, 2H), 1.69 (ddd, *J* = 13.9, 9.5, 5.1 Hz, 1H), 1.61 (ddd, *J* = 14.1, 10.5, 4.7 Hz, 1H), 1.28 (dt, *J* = 13.6, 3.8 Hz, 1H), 1.06 (dt, *J* = 13.3, 4.1 Hz, 1H), 0.97 (d, *J* = 7.3 Hz, 3H), 0.90 (s, 9H), 0.86 (s, 9H), 0.11 (s, 6H), 0.01 (s, 6H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 151.2, 149.2, 109.7, 102.3, 68.5, 41.3, 38.7, 36.0, 35.5, 34.8, 30.6, 30.4, 28.3, 26.13, 26.06, 25.9, 20.5, 18.9, 18.4, 18.32, 18.26, 18.2, 15.9, 7.03, 6.98, 5.3, -4.0, -4.3, -4.4, -4.5, -4.6; **IR** (thin film) 2955, 2911, 1673, 1461, 1360, 1255, 1180, 1152, 1097, 1068, 1005, 963, 892, 859, 835, 773, 745 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{23}H_{46}O_2Si_2H$ $(M + H)^+$: 411.3115, found: 411.3128; **TLC** (5% CH_2Cl_2 in hexanes) $\mathbf{R}_f$ = 0.36 (CAM Stain).



equatorial-S3 4.32

(((1*R*,3*R*,4a*S*,8a*R*)-3-methyl-1,2,3,4,4a,5,8,8a-octahydronaphthalene-1,6-diyl)bis(oxy))bis(*tert*-butyldimethylsilane) (4.32): To a solution of **S3** (286 mg, 0.97 mmol) in CH_2Cl_2 (1.4 mL) at –78 °C was added 2,6-lutidine (0.22 mL, 1.93 mmol, 2 equiv) and TBSOTf (0.29 mL, 1.26 mmol, 1.3 equiv). The mixture was stirred for 7 h, and the reaction was quenched

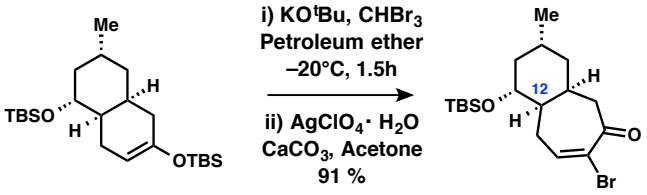
with Et_3N (0.17 mL, 1.26 mmol, 1.3 equiv) and $NaHCO_3$ (5 mL) at –78 °C. The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 x 5 mL). The combined organic layers were washed with brine, dried over Na_2SO_4 and concentrated *in vacuo* to give a yellow oil. Purification by column chromatography (5% CH_2Cl_2 in hexanes) gave the product (364 mg, 92%) as a clear oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 4.76–4.72 (m, 1H), 3.58–3.50 (m, 1H), 2.37 (dd, *J* = 16.2, 5.6 Hz, 1H), 2.21–2.13 (m, 2H), 2.06–1.93 (m, 2H), 1.88–1.72 (m, 3H), 1.52–1.45 (m, 2H), 1.15 (td, *J* = 12.8, 4.5 Hz, 1H), 1.00–0.95 (m, 6H), 0.94–0.85 (m, 16H), 0.70–0.62 (m, 4H), 0.12 (d, *J* = 9.3 Hz, 2H), 0.03 (s, 6H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 149.0,102.2, 101.6, 68.4, 45.4, 41.8, 39.2, 33.7, 31.7, 26.5, 26.1, 25.9, 24.4, 22.6, 18.3, 18.2, 7.0, 5.2, -3.9, -4.0, -4.3, -4.5, -4.6; **IR** (thin film) 2954, 2910, 1670, 1461, 1362, 1250, 1170, 1100, 1090, 1065, 1001, 961, 895, 831, 771, 744 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{23}H_{46}O_2Si_2H$ $(M + H)^+$: 411.3115, found: 411.3132; **TLC** (5% CH_2Cl_2 in hexanes) $\mathbf{R}_f$ = 0.24 (CAM Stain); $[\alpha]^{23}_D$ = -8.0 (*c* 1.80, $CHCl_3$).



4.31 **4.35**

(1*S*,3*R*,4a*S*,9a*R*)-7-bromo-1-((*tert*-butyldimethylsilyl)oxy)-3-methyl-1,2,3,4,4a,5,9,9a-octahydro-6*H*-benzo[7]annulen-6-one (4.35): To a solution of compound **4.31** (1.24 g, 3.03 mmol) in petroleum ether (16 mL) at –20 °C was added KO*t*-Bu (1.02 g, 9.08 mmol) and freshly distilled bromoform (0.79 mL). The reaction mixture was allowed to stir at –20 °C until starting material was consumed as observed by TLC. After 1h, the mixture was poured into 12 mL of water. The organic layer was separated and the aqueous layer was extrated with EtOAc (3 x 20

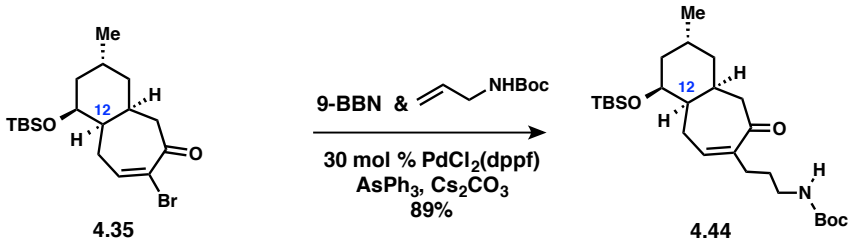
mL). The combined organic layers were dried over Na_2SO_4 and concentrated under vacuo. The residue was dissolved in acetone (33 mL), to this solution was added calcium carbonate (1.51 g, 15.14 mmol) and silver perchlorate monohydrate (3.14 g, 15.14 mmol), and the mixture was stirred at 25 °C overnight, during which time a dark precipitate developed. The mixture was quenched by addition of Et_3N (2.11 mL, 15.14 mmol) and silica gel (~1.0 g), and the mixture concentrated under vacuo. The resulting crude mixture was flushed through a plug of silica using Et_2O. Purification by column chromatography gave the product (0.96, 82%) as a yellow oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 7.23 (dd, *J* = 9.8, 4.8 Hz, 1H), 3.89–3.84 (m, 1H) 3.05 (dd, *J* = 15.5, 11.0 Hz, 1H), 2.58 (ddd, *J* = 15.7, 11.1, 4.7 Hz, 1H), 2.51 (dd, *J* = 16.0, 3.0 Hz, 1H), 2.37 (ddd, *J* = 15.8, 9.7, 4.3 Hz, 1H), 2.24–2.1(app. m, 1H), 1.95–1.85 (app. m, 1H), 1.80 (dq, *J* = 10.0, 4.0 Hz, 1H), 1.70–1.62 (app. m, 1H), 1.56 (dt, *J* = 13.7, 3.9 Hz, 1H), 1.31 (ddd, *J* = 13.8, 9.0, 5.3 Hz, 1H), 1.21–1.14 (app. m, 1H), 0.91 (d, *J* = 7 Hz, 3H), 0.85 (s, 9H), 0.02 (s, 3H), -0.02 (s, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 197.8, 145.9, 125.6, 69.9, 47.0, 40.7, 40.4, 38.6, 31.0, 28.1, 26.0, 23.4, 21.1, 18.1, -4.4, -4.8; **IR** (thin film) 2952, 2926, 2852, 1681, 1462, 1253, 1059, 834, 735 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{18}H_{31}BrO_2Si$ $(M + NH_4)^+$: 404.1620, found: 411.1613; **TLC** (40% CH_2Cl_2 in hexanes) $\mathbf{R_f}$ = 0.34 (CAM Stain); $[\alpha]^{23}_D$ = +45 (*c* 0.9, $CHCl_3$).



4.32 **4.36**

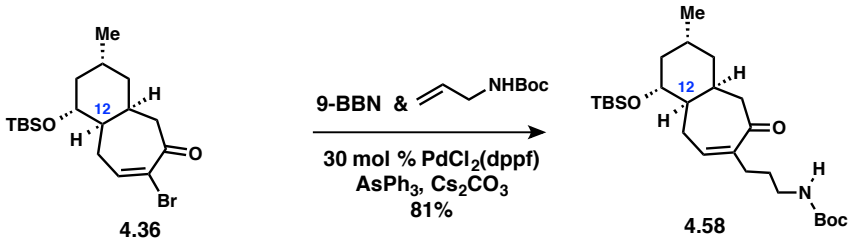
(1*R*,3*R*,4a*S*,9a*R*)-7-bromo-1-((*tert*-butyldimethylsilyl)oxy)-3-methyl-1,2,3,4,4a,5,9,9a-octahydro-6*H*-benzo[7]annulen-6-one (4.36): To a solution of compound **4.32** (364 mg, 0.89 mmol) in petroleum ether (4.7 mL) at –20 °C was added KO*t*-Bu (297 mg, 2.66 mmol) and freshly distilled bromoform (0.23 mL). The reaction mixture was allowed to stir at –20 °C until starting material was consumed as observed by TLC. After 1.5h, the mixture was poured into 5 mL of water. The organic layer was separated and the aqueous layer was extrated with EtOAc (3 x 5 mL). The combined organic layers were dried over Na_2SO_4 and concentrated under vacuo. The residue was dissolved in acetone (9.9 mL), to this solution was added calcium carbonate (444 mg, 4.44 mmol) and silver perchlorate monohydrate (993 mg, 4.44 mmol), and the mixture was stirred at 25 °C overnight, during which time a dark precipitate developed. The mixture was quenched by addition of Et_3N (0.62 mL, 4.44 mmol) and silica gel (~0.5 g), and the mixture concentrated under vacuo. The resulting crude mixture was flushed through a plug of silica using Et_2O. Purification by column chromatography gave the product (311 mg, 91%) as a yellow oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 7.16 (dd, J = 9.3, 6.2 Hz, 1H), 3.47 (td, J = 10.6, 4.4 Hz, 1H), 2.78 (ddd, J = 15.3, 9.0, 5.8 Hz, 1H), 2.63 (dd, J = 17.0, 11.0 Hz, 1H), 2.46 (app. d, 15.5 Hz, 1H), 2.33–2.26 (m, 1H), 2.09 (ddd, J = 14.9, 10.3, 6.3 Hz, 1H), 1.85–1.79 (m, 1H), 1.73 (tt, J = 9.5, 6.3 Hz, 1H), 1.57 (dd, J = 13.5, 2.0 Hz, 1H), 1.51–1.39 (m, 1H), 1.30–1.19 (m, 2H), 1.06 (q, J = 11.7 Hz, 1H), 0.91 (d, J = 6.5 Hz, 1H), 0.88 (s, 9H), 0.06 (s, 3H), 0.04 (s, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 197.7, 143.9, 126.0, 73.6, 45.4, 44.2, 41.6, 40.6, 32.8, 32.3, 26.9, 26.0, 22.3, 18.2, -3.6, -4.5; **IR** (thin film) 2957,

2925, 2852, 1681, 1456, 1253, 1101, 1064, 834, 772 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{18}H_{31}BrO_2Si$ $(M + NH_4)^+$: 404.1620, found: 411.1623; **TLC** (40% CH_2Cl_2 in hexanes) $\mathbf{R_f}$ = 0.28 (CAM Stain).



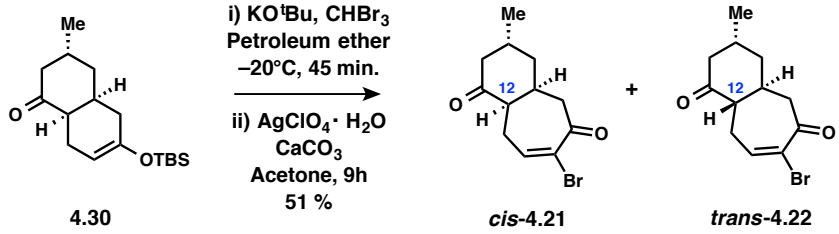
***tert*-butyl(3-((1*S*,3*R*,4a*S*,9a*R*)-1-((*tert*-butyldimethylsilyl)oxy)-3-methyl-6-oxo 2,3,4,4a,5,6,9,9a-octahydro-1*H*-benzo[7]annulen-7-yl)propyl)carbamate (4.44):** To a solution of *tert*–butyl allylcarbamate (239 mg, 1.51 mmol) in degassed THF (2.5 mL) was added a solution of 9-BBN (0.5 M in THF, 4.3 mL, 2.13 mmol) at room temperature. After stirring for 4 h, the solution was treated with degassed water (365 μL, 20.25 mmol) for 20 min. In a separate Schlenk flask, bromo enone **4.35** (391 mg, 1.01 mmol), $CsCO_3$ (725 mg, 2.23 mmol), $AsPh_3$ (124 mg, 0.41 mmol), and $Pd(dppf)Cl_2$ (296 mg, 0.41 mmol) were degassed via high-vacuum/argon cycles (4x) and diluted in degassed DMF (6.5 mL). The resulting mixture was then stirred for 15 min before the borane solution was added. The reaction was stirred for 4 h at 80 °C, at which point the mixture turned black. The mixture was cooled to room temperature, diluted with Et_2O (15 mL) and filtered through a plug of alumina. Concentration in *vacuo* followed by purification via flash column chromatography (eluent, gradient 15% EtOAc in hexanes) the desired product (419 mg, 89%) as a colorless oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 6.39–6.21 (m, 1H), 4.80–4.55 (br. s, 1H), 3.88–3.83 (m, 1H), 3.09–3.02 (br. s, 2H), 2.91–2.84 (m, 1H), 2.52–2.42 (m, 1H), 2.33–2.27 (m, 1H), 2.26–2.21 (m, 1H), 2.20–2.10 (m, 1H), 1.94–1.86 (m, 1H), 1.73–1.67 (m, 1H), 1.65–1.60 (m, 1H), 1.56–1.49 (m, 3H), 1.41 (s, 9H),

1.31–1.23 (m, 1H), 1.21–1.14 (m, 1H), 0.91 (d, 7.0 Hz, 3H), 0.84 (s, 9H), 0.00 (s, 3H), -0.02 (s, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 206.7, 156.2, 142.5, 140.6, 79.1, 70.0, 48.6, 41.7, 40.2, 40.1, 38.3, 31.1, 29.9, 29.7, 28.6, 26.0, 25.9, 23.9, 21.0, 18.2, -4.4, -4.7; IR (thin film) 3362, 2925, 2857, 1694, 1515, 1451, 1410, 1388, 1364, 1299, 1251, 1166, 1105, 1050, 636, 775, 676 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{26}H_{47}NO_4SiNa$ $(M + Na)^+$: 488.3172, found: 488.3174; TLC (15% EtOAc in hexanes) $\mathbf{R}_f$ = 0.31 (CAM Stain); $[\alpha]^{23}_D$ = +34 (*c* 0.83, $CHCl_3$).



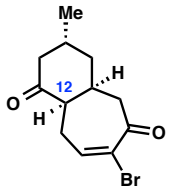
***tert*-butyl(3-((1*R*,3*R*,4a*S*,9a*R*)-1-((*tert*-butyldimethylsilyl)oxy)-3-methyl-6-oxo-2,3,4,4a,5,6,9,9a-octahydro-1*H*-benzo[7]annulen-7-yl)propyl)carbamate (4.58):** To a solution of *tert*–butyl allylcarbamate (131 mg, 0.83 mmol) in degassed THF (1.4 mL) was added a solution of 9-BBN (0.5 M in THF, 2.3 mL, 1.16 mmol) at room temperature. After stirring for 4 h, the solution was treated with degassed water (199 μL, 11.08 mmol) for 20 min. In a separate Schlenk flask, bromo enone **4.36** (214 mg, 0.55 mmol), $CsCO_3$ (397 mg, 1.22 mmol), $AsPh_3$ (50 mg, 0.17 mmol), and $Pd(dppf)Cl_2$ (122 mg, 0.17 mmol) were degassed via high-vacuum/argon cycles (4x) and diluted in degassed DMF (3.6 mL). The resulting mixture was then stirred for 15 min before the borane solution was added. The reaction was stirred for 4 h at 80 °C, at which point the mixture turned black. The mixture was cooled to room temperature, diluted with Et_2O (15 mL) and filtered through a plug of alumina. Concentration in *vacuo* followed by purification via flash column chromatography (eluent, gradient 15% EtOAc in hexanes) the desired product (209 mg, 81%) as a colorless oil. **^{1}H NMR** (500 MHz, $CDCl_3$) δ 6.38–6.33 (m, 1H), 4.70-4.62

(br. s, 1H), 3.46 (td, J = 10.6, 4.4 Hz, 1H), 3.11–3.04 (m, 2H), 2.70 (ddd, J = 14.7, 9.8, 6.0 Hz, 1H), 2.52 (dd, J = 17.3, 11.6 Hz, 1H), 2.35–2.27 (m 1H), 2.26–2.19 (m, 2H), 2.17–2.09 (m, 1H), 2.01–1.93 (m, 1H), 1.82–1.76 (m, 1H), 1.67–1.59 (m, 1H), 1.58–1.50 (m, 4H), 1.43 (s, 9H), 1.22 (dt, J = 13.0, 5.2 Hz, 1H), 0.99 (q, J = 11.8 Hz, 1H), 0.90 (d, J = 6.6 Hz, 3H), 0.88 (s, 9H), 0.05 (s, 3H), 0.03 (s, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 207.0, 156.2, 142.6, 137.8, 79.2, 74.0, 46.7, 44.4, 41.7, 40.8, 40.2, 32.9, 30.6, 30.0, 29.7, 28.6, 27.0 26.0, 22.4, 18.2, -3.6, -4.5; **IR** (thin film) 3373, 2926, 1701, 1665, 1512, 1463, 1364, 1249, 1171, 1103, 1058, 835, 774 cm^{-1}; **HRMS** (ESI/methanol) m / z calcd for $C_{26}H_{47}NO_4SiNa$ $(M + Na)^+$: 488.3172, found: 488.3181; **TLC** (15% EtOAc in hexanes) $\mathbf{R}_f$ = 0.30 (CAM Stain); $[\alpha]^{23}_D$ = +22 (c 1.14, $CHCl_3$).



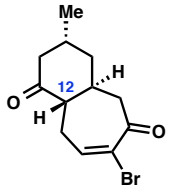
(3*R*,4a*S*)-7-bromo-3-methyl-3,4,4a,5,9,9a-hexahydro-1*H*-benzo[7]annulene-1,6(2*H*)-dione (4.21) and (4.22): To a solution of decalin **4.30** (1.21 g, 4.12 mmol) in petroleum ether (110 mL) at –20 °C was added potassium *tert*-butoxide (1.39 g, 12.37 mmol) in 3 portions. The heterogeneous mixture turned yellow within 2 min. After 2 min, freshly distilled bromoform (1.08 mL, 12.37 mmol) was added dropwise in petroleum ether (20 mL) over 4 min. The reaction mixture was allowed to stir at –20 °C until starting material was consumed as observed by TLC. After 45 min, the mixture was removed from the cooling bath and filtered through a silica plug with 25% EtOAc in petroleum ether. The filtrate was concentrated under vacuum and the resulting yellow oil was dissolved in acetone (45 mL). Calcium carbonate (2.06 g, 20.63 mmol) and silver perchlorate monohydrate (1.85 g, 8.25 mmol) were added. The reaction was allowed

to stir at 25 °C for 9 h, during which time a dark precipitate developed. The reaction was quenched by addition of Et_3N (1.15 mL, 8.25 mmol) and silica gel (1.5 g), and the mixture concentrated under vacuo. The resulting crude mixture was flushed through a plug of silica using Et_2O. The material was concentrated under vacuo and purified via chromatography (eluent, gradient 15% → 25% EtOAc in hexanes) to afford a mixture of diastereomers **4.21** and **4.22** (0.57 g, 51%) as yellow oil (~3:1 *cis*/*trans* mixture of C-12 epimers). A small sample of the mixture was purified by MPLC to separate the cis and trans isomers for characterization.



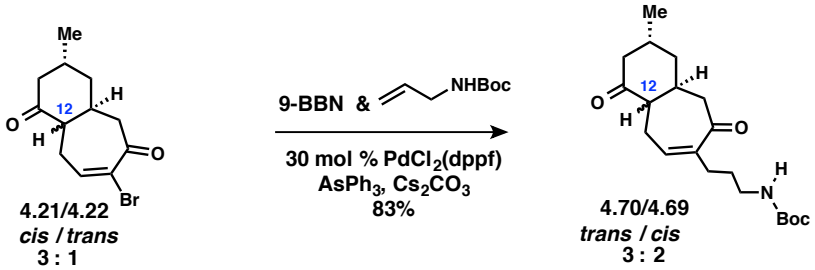
***cis*-4.21**

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.24 (dd, *J* = 10.5, 4.8 Hz, 1H), 2.75–2.65 (m, 3H), 2.57–2.46 (m, 3H), 2.4 (dd, *J* = 16.3, 10.3 Hz, 1H), 2.07 (t, *J* = 12.8 Hz, 1H), 1.97–1.87 (m, 1H), 1.84 (dt, *J* = 13.5, 3.3 Hz, 1H), 1.75 (ddd, *J* = 14.8, 11.5, 4.3 Hz, 1H), 1.05 (d, *J* = 6.5 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 209.8, 195.7, 144.1, 125.6, 49.7, 47.4, 45.3, 39.4, 34.7, 29.9, 27.0, 22.1; **IR** (thin film) 3444, 2955, 2924, 1705, 1685, 1600, 1452, 1379, 1231, 1111, 1041, 916 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{12}H_{15}BrO_2Na$ $(M + Na)^+$: 293.0153, found: 293.0161; **TLC** (20% EtOAc in hexanes) R_f = 0.33 (CAM Stain).



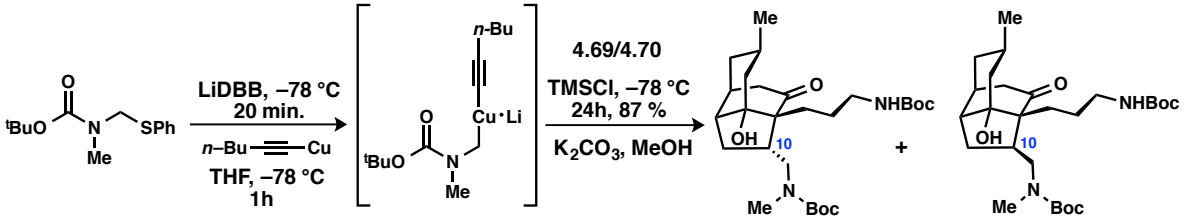
trans-4.22

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.22 (dd, *J* = 8.3, 4.3 Hz, 1H), 2.92 (dd, *J* = 14.0, 6.0 Hz, 1H), 2.87–2.78 (m, 1 Hz), 2.60 (dd, *J* = 14.0, 5.5 Hz, 1H), 2.53–2.36 (m, 3H), 2.27–2.21 (m, 1H), 2.18 (d, *J* = 13 Hz, 1H), 1.94 (td, *J* = 13.0, 4.8 Hz, 1H), 1.68 (d, *J* = 14.5 Hz, 1H), 0.98 (d, *J* = 7Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 209.4, 195.3, 145.3, 125.9, 54.9, 48.0, 46.9, 38.3, 35.6, 29. 8, 28.4, 20.0; **IR** (thin film) 3437, 2957, 2826, 1602, 1711, 1687, 1459, 1385, 1238, 1090, 912 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{12}H_{15}BrO_2Na$ $(M + Na)^+$: 293.0153, found: 293.0161; **TLC** (20% EtOAc in hexanes) $\mathbf{R}_f$ = 0.32 (CAM Stain).



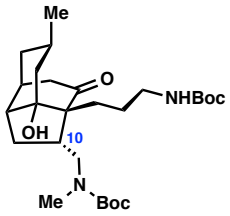
***tert*-butyl (3-((3*R*,4a*S*)-3-methyl-1,6-dioxo-2,3,4,4a,5,6,9,9a-octahydro-1*H*-benzo[7]annulen-7-yl)propyl)carbamate (4.70/4.69):** To a solution of *tert*–butyl allylcarbamate (402 mg, 2.56 mmol) in degassed THF (4.3 mL) was added a solution of 9-BBN (0.5 M in THF, 7.2 mL, 3.58 mmol) at room temperature. After stirring for 4 h, the solution was treated with degassed water (615 µL, 34.14 mmol) for 20 min. In a separate Schlenk flask, bromo enone **4.21**/**4.22** (461 mg, 1.71 mmol), $CsCO_3$ (1.22 g, 3.76 mmol), $AsPh_3$ (157 mg, 0.51 mmol), and $Pd(dppf)Cl_2$ (375 mg, 0.51 mmol) were degassed via high-vacuum/argon cycles (4x) and diluted in degassed

DMF (11 mL). The resulting mixture was then stirred for 15 min before the borane solution was added. The reaction was stirred for 4 h at 80 °C, at which point the mixture turned black. The mixture was cooled to room temperature, diluted with Et_2O (15 mL) and filtered through a plug of alumina. Concentration in *vacuo* followed by purification via flash column chromatography (eluent, gradient 30% → 40% EtOAc in hexanes) afforded inseparable diastereomers (494 mg, 83%) of **4.70** and **4.69** as a colorless oil (~ 3:2 *trans*/*cis* epimers at C-12). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 6.50–6.40 (m, 1H), 4.67–4.55 (br. s, 1H), 3.13–3.01 (app. m, 2H), 2.83–2.73 (app. m, 1H), 2.66–2.60 (m, 0.5H), 2.59–2.54 (m, 1H), 2.49–2.40 (m, 2.5H), 2.34–2.25 (m, 2.5H), 2.20–2.11 (m, 2H), 2.03 (t, J = 12.5 Hz, 0.5H), 1.97–1.87 (m, 1H), 1.82–1.75 (m, 0.5H), 1.73–1.69 (m, 0.5H), 1.68–1.64 (m, 1H), 1.54 (p, J = 5.9 Hz, 2H), 1.42 (s, 9H), 1.02 (d, J = 6.5 Hz, 1.5H), 0.97 (d, J = 7.0, Hz, 1.5H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 211.1, 210.5, 204.6, 204.1, 156.2, 142.6, 139.5, 138.4, 79.3, 55.6, 53.3, 49.8, 49.1, 48.0, 47.04, 46.99, 40.5, 40.1, 39.5, 39.0, 38.3, 36.0, 34.8, 30.5, 29.99, 29.97, 29.92, 29.6, 28.6, 26.7, 25.6, 22.1, 20.1; **IR** (thin film) 3373, 2953, 2921, 2881, 1708, 1664, 1517, 1454, 1391, 1363, 1252, 1173, 875 cm^{-1}; **HRMS** (ESI/methanol) m / z calcd for $C_{20}H_{31}NO_4Na$ $(M + Na)^+$: 372.2151, found: 372.2157; **TLC** (40% EtOAc in hexanes) $\mathbf{R}_f$ = 0.32 (CAM Stain).



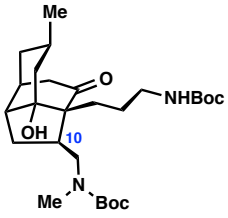
Tricycle 4.75 and its C10 epimer: A round bottom flask containing **4.54** (0.62 g, 2.44 mmol) and 1,10-phenanthroline (2-3 crystals) was dried by azeotroping three times with freshly distilled benzene. The flask was then equipped with a glass stir bar and THF (27 mL) was introduced

under Ar. The mixture was cooled to –78 °C and *n*–BuLi/hexanes (2-3 M) was added until a brown dark color persisted (~0.3–0.4mL). This procedure was performed to quench adventitious proton sources. LiDBB (12.8 mL, 5.12 mmol, 2.1 equiv) was then added dropwise over 10 min at –78 °C until a dark-green color persisted, and the mixture was allowed to stir for 20 min. A separate flask containing 1-hexynyl copper (0.71 g, 4.94 mmol) and tetrahydrofuran (6.2 mL) was cooled to –78 °C and trimethyl phosphite (1.8 mL, 14.6 mmol) was introduced; the mixture was stirred until a clear solution developed. The resulting homogeneous solution was added via syringe to the organolithium reagent down the flask wall over 3 min and stirring was continued for 1 h to produce 15 as deep red solution. The carbamate 10 (213 mg, 0.61 mmol) was added as a solution in THF (0.5 mL) with freshly distilled TMSCl (0.39 mL, 3.05 mmol). The resulting mixture was stirred at –78 °C for 24 h and quenched with 10% concentrated ammonium hydroxide/saturated ammonium chloride (120 mL), followed by warming to room temperature. After 1 h, the organic layers were separated and the aqueous layers were extracted with ethyl acetate (40 mL) three times. The organic layers were combined, dried and concentrated under vacuum. The resulting mixture was filtered through a plug silica with 20% CH_2Cl_2 in hexanes to remove excess of 4,4'-di-*tert*-butylbiphenyl, at which point ethyl acetate was used to flushed the plug. The material was concentrated under vacuum. The crude product was dissolved in methanol (15 mL) with potassium carbonate (627 mg, 4.54 mmol) and stirred for 4 h. The mixture was concentrated, loaded onto silica gel with DCM and purified by MPLC, eluting with 20% to 40% EtOAc/hexanes gradient, to deliver tricycle **4.75** (126.1 mg, 42%) and its C10 epimer (134.5 mg, 45%).



4.74

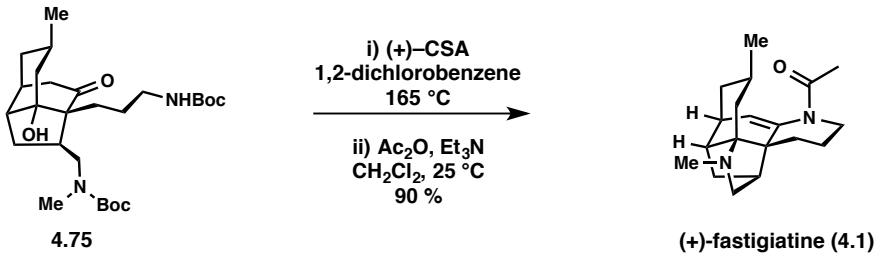
^{1}H NMR (500 MHz, tol-d_8, 85 °C) δ 4.46–4.37 (br s, 1H), 3.42–3.27 (br s, 1H), 3.10 (m, 2H), 2.77–2.72 (m, 1H), 2.71–2.68 (m, 1H), 2.67 (s, 3H), 2.63–2.52 (m, 1H), 2.25 (ddd, *J* = 13.5, 11.2, 7.6 Hz, 1H), 2.01 (d, *J* = 17.5 Hz, 1H), 1.87 (app d, *J* = 13.6 Hz, 1H), 1.85–1.80 (m, 1H), 1.76–1.67 (m, 1H), 1.66–1.56 (m, 5H), 1.45 (s, 18H), 1.37–1.29 (m, 2H), 1.12–1.00 (br s, 1H), 0.86 (q, *J* = 13.7 Hz, 2H), 0.70 (d, *J* = 6.5 Hz, 3 H); **^{13}C NMR** (125 MHz, $CDCl_3$, 25 °C) δ 213.5, 213.3, 156.5, 156.3, 155.8, 80.1, 79.6, 79.3, 65.4, 48.4, 48.0, 47.1, 43.7, 43.2, 43.0, 42.7, 41.8, 41.1, 35.5, 35.3, 34.5, 34.2, 32.1, 29.9, 29.8, 29.6, 28.7, 28.6, 26.0, 25.5, 24.5, 22.9, 22.5, 14.3 ; **IR** (thin film) 3364, 2962, 2925, 1686, 1519, 1482, 1451, 1393, 1367, 1247, 1163, 1043, 870, 771 cm^{-1}; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{27}H_{46}N_2O_6Na$ $(M + Na)^+$: 517.3254, found: 517.3261; **TLC** (44 % EtOAc in Hexanes) R_f = 0.34 (CAM stain); $[\alpha]^{24}_D$ = –74 (*c* 1.23, $CHCl_3$).



4.75

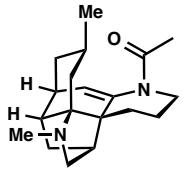
^{1}H NMR (500 MHz, tol-d_8, 85 °C) δ 4.51–4.43 (br s, 1H), 3.75 (dd, *J* = 13.3, 11.3 Hz, 1H), 3.17 (dd, *J* = 13.8, 4.3 Hz, 1H), 3.12–3.05 (m, 2H), 2.71 (s, 3H), 2.29 (dd, *J* = 16.5, 8.0 Hz, 1H), 2.17 (ddd, *J* = 12.8, 7.0, 5.3 Hz, 1H), 2.03–1.96 (m, 1H), 1.91–1.85 (m, 1H), 1.79 (d, *J* = 16.5 Hz, 1H) 1.74 (dd, *J* = 14.0, 4.5 Hz, 1H), 1.69–1.64 (m, 1H), 1.61–1.51 (m,

4H), 1.46 (s, 9H), 1.43 (s, 9H), 1.34–1.26 (m, 3H), 1.06–1.00 (br s, 1H), 0.83 (td, J = 12.8, 3.0 Hz, 1H), 0.73 (t, J = 13.0 Hz, 1H), 0.65 (d, J = 6.0 Hz, 3H), 0.51–0.43 (br s, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$, 25 °C) δ 214.4, 214.2, 156.8, 156.4, 156.22, 83.9, 83.8, 79.75, 79.66, 79.12, 79.06, 65.2, 65.1, 52.6, 51.7, 48.0, 43.0, 42.1, 41.9, 41.3, 40.7, 35.4, 35.3, 34.9, 32.1, 31.9, 31.6, 29.9, 29.8, 29.5, 28.6, 25.6, 22.9, 22.7, 21.8, 14.3; **IR** (thin film) 3380, 2957, 2920, 1961, 1514, 1456, 1393, 1362, 1252, 1168, 1033, 876, 771 cm^{-1}; **HRMS** (ESI/methanol) m / z calcd for $C_{27}H_{46}N_2O_6Na$ $(M + Na)^+$: 517.3254, found: 517.3234; **TLC** (44 % EtOAc in Hexanes) $\mathbf{R_f}$ = 0.38 (CAM stain); $[\alpha]^{24}_D$ = –41 (c 1.82, $CHCl_3$).



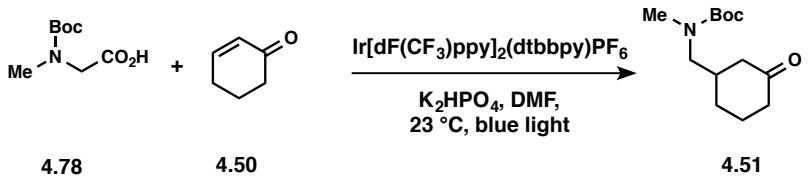
(+)-Fastigiatine (4.1): A 10 mL Schlenk flask was charged with tricycle **4.75** (57.1 mg, 0.12 mmol) and purged three times with argon/vacuum. Freshly distilled and degassed 1,2-dichlorobenzene (5.9 mL) was introduced and the solution cooled to 0 °C, at which point (+)-10-camphorsulfonic acid (402.5 mg, 1.73 mmol) was added. The reaction was removed from the ice bath and warmed to 165 °C in a sealed atmosphere for 1 h. The mixture was cooled to 0 °C, quenched with saturated $NaHCO_3$ (5 mL) and extracted with $CHCl_3$ (5 mL) two times. The combined organic layers were dried over Na_2SO_4 and concentrated to remove $CHCl_3$. To the resulting solution were added Et_3N (0.16 mL, 1.16 mmol) and Ac_2O (0.11 mL, 1.16 mmol), and the mixture was stirred for 5 h. The reaction was quenched by addition of methanol (2 mL). Concentration under vacuum and purification by silica gel chromatography (gradient 1% → 10%

MeOH in $CHCl_3$ with 0.5% Ammonium hydroxide) afforded (+)-fastigiatine (34.6 mg, 90% yield) as a white crystalline solid. The data for the synthetic natural product matched that reported by Shair.[4]



(+)-fastigiatine (4.1)

^{1}H NMR (500 MHz, $CDCl_3$, 25 °C) δ 5.19 (d, *J* = 5.5 Hz, 1H), 3.82 (dt, *J* = 11.5, 6.0 Hz, 1H), 3.30–3.21 (m, 2H), 2.42–2.37 (m, 1H), 2.32 (s, 3H), 2.19 (d, 9.0 Hz, 1H), 2.18–2.16 (m, 1H), 2.15 (s, 3H), 2.07 (br. app. d, *J* = 14.5 Hz, 1H), 2.06–1.96 (m, 1H), 1.93–1.89 (m, 1H), 1.81–1.72 (m, 1H), 1.68 (dd, *J* = 14.0, 4.5 Hz, 1H), 1.63–1.53 (m, 3H), 1.43–1.32 (m, 2H), 1.20 (app. t, *J* = 12.0 Hz, 1H), 1.02 (app. dt, *J* = 12.8, 3.3 Hz, 1H), 0.91 (d, 6.5 Hz, 3H); **^{13}C NMR** (125 MHz, $CDCl_3$, 25 °C) δ 170.5, 139.6, 123.6, 65.7, 60.0, 55.4, 45.9, 45.8, 40.6, 38.7, 37.8, 35.4, 35.0, 34.3, 25.9, 23.4, 22.7, 22.0, 21.6; **HRMS** (ESI/methanol) *m* / *z* calcd for $C_{19}H_{28}N_2ONa$ $(M + Na)^+$: 323.2099, found: 323.2106; **TLC** (10 % MeOH in $CHCl_3$) $\mathbf{R_f}$ = 0.33 (UV or $KMnO_4$);); $[\alpha]^{24}_D$= +310 (*c* 1.32, $CHCl_3$).



4.78 **4.50** **4.51**

A 2 mL oven-dried dram vial equipped with a magnetic stirrer was charged with **4.78** (75 mg, 0.40 mmol),[32] cyclohexenone (25 mg, 0.26 mmol), K_2HPO_4 (78 mg, 0.45 mmol), $Ir[dF(CF_3)ppy]_2(dtbbpy)PF_6$ (11.8 mg, 0.01 mmol) and distilled DMF (0.66 mL, 0.4 M). The reaction mixture was degassed by bubbling argon for 15 min and the vial was sealed and irradiated with (2 x 34 W blue LED lamps) for 24 hours. The crude mixture was purified via column chromatography to afford the desired product **4.51** (61.2 mg, 96%) as a yellow oil. Spectral data matched those reported in the literature.[30]

X. References:

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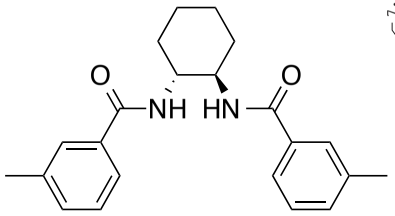
APPENDIX

NMR Spectra of Compounds

1H spectrum



1.11-(*R,R*)

7.533
7.502
7.488
7.260
7.233
7.219
7.205
7.190
6.867
6.857

4.015

2.361
2.306
2.223
2.206

1.835

1.427
1.414

0.069
-0.003

```
Current Data Parameters
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NAME       RAS_1_82_PURE_2
EXPNO                     1
PROCNO                    1

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Time                   9.54
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PROBHD   5 mm CPTCI 1H-
PULPROG                zg30
TD                    81728
SOLVENT               CDCl3
NS                       16
DS                        2
SWH                8012.820 Hz
FIDRES             0.098043 Hz
AQ                 5.0998774 sec
RG                       5.7
DW                    62.400 usec
DE                     6.00 usec
TE                    298.0 K
D1                0.10000000 sec
MCREST            0.00000000 sec
MCWRK             0.01500000 sec

======== CHANNEL f1 ========
NUC1                      1H
P1                     7.50 usec
PL1                    1.60 dB
SFO1           500.2235015 MHz

F2 - Processing parameters
SI                     65536
SF             500.2200321 MHz
WDW                       no
SSB                        0
LB                      0.00 Hz
GB                         0
PC                      4.00
```
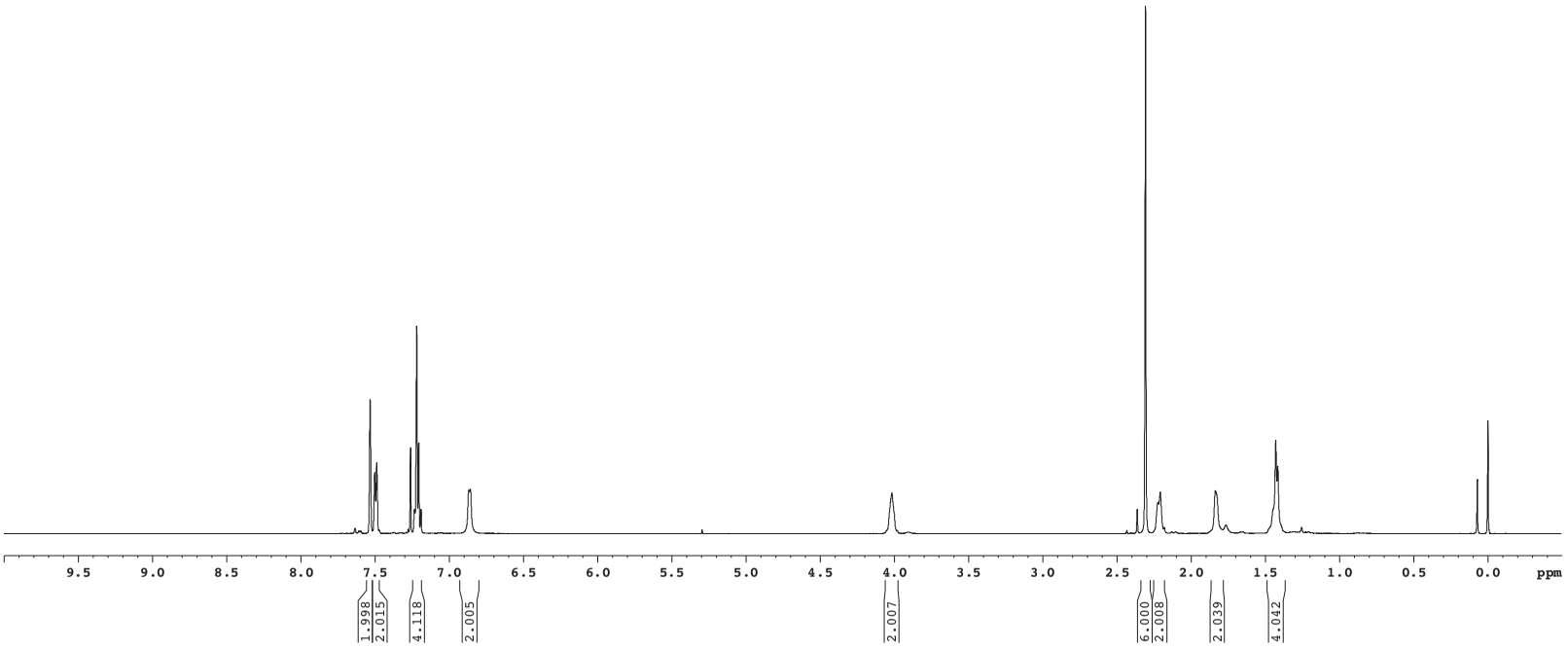
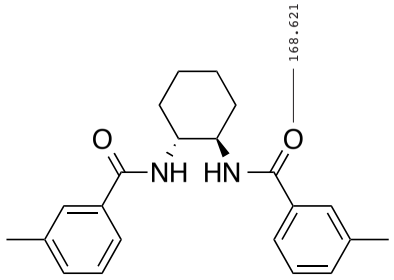



Z–restored spin–echo 13C spectrum with 1H decoupling



1.11-(*R,R*)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PROCNO                1

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PROBHD   5 mm CPTCI 1H-
PULPROG  SpinEchopg30gp.prd
TD                65536
SOLVENT             DMSO
NS                  210
DS                   16
SWH           30303.031 Hz
FIDRES         0.462388 Hz
AQ            1.0813940 sec
RG               3649.1
DW               16.500 usec
DE                6.00 usec
TE                298.0 K
D1           0.25000000 sec
d11          0.03000000 sec
D16          0.00020000 sec
d17          0.00019600 sec
MCREST       0.00000000 sec
MCWRK        0.01500000 sec
P2                31.00 usec

======== CHANNEL f1 ========
NUC1                13C
P1                15.50 usec
P11              500.00 usec
P12             2000.00 usec
PL0              120.00 dB
PL1               -1.00 dB
SFO1        125.7942548 MHz
SP1               3.20 dB
SP2               3.20 dB
SPNAM1    Crp60,0.5,20.1
SPNAM2      Crp60comp.4
SPOFF1               0.00 Hz
SPOFF2               0.00 Hz

======== CHANNEL f2 ========
CPDPRG2          waltz16
NUC2                1H
PCPD2            100.00 usec
PL2               1.60 dB
PL12             24.60 dB
SFO2        500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1         SINE.100
GPNAM2         SINE.100
GPX1              0.00 %
GPX2              0.00 %
GPY1              0.00 %
GPY2              0.00 %
GPZ1             30.00 %
GPZ2             50.00 %
p15             500.00 usec
p16            1000.00 usec

F2 - Processing parameters
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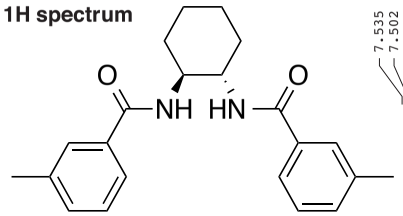
SF          125.7803997 MHz
WDW                  no
SSB                   0
LB                0.00 Hz
GB                    0
PC                2.00
```

1H spectrum



1.11-(*S,S*)


```
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EXPNO                     1
PROCNO                    1

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Time                   9.59
INSTRUM            cryo500
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PULPROG                zg30
TD                    81728
SOLVENT               CDCl3
NS                       16
DS                        2
SWH                8012.820 Hz
FIDRES             0.098043 Hz
AQ                 5.0998774 sec
RG                       6.3
DW                    62.400 usec
DE                     6.00 usec
TE                    298.0 K
D1                 0.10000000 sec
MCREST             0.00000000 sec
MCWRK              0.01500000 sec

======== CHANNEL f1 ========
NUC1                      1H
P1                     7.50 usec
PL1                    1.60 dB
SFO1           500.2235015 MHz

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SSB                           0
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GB                            0
PC                     4.00
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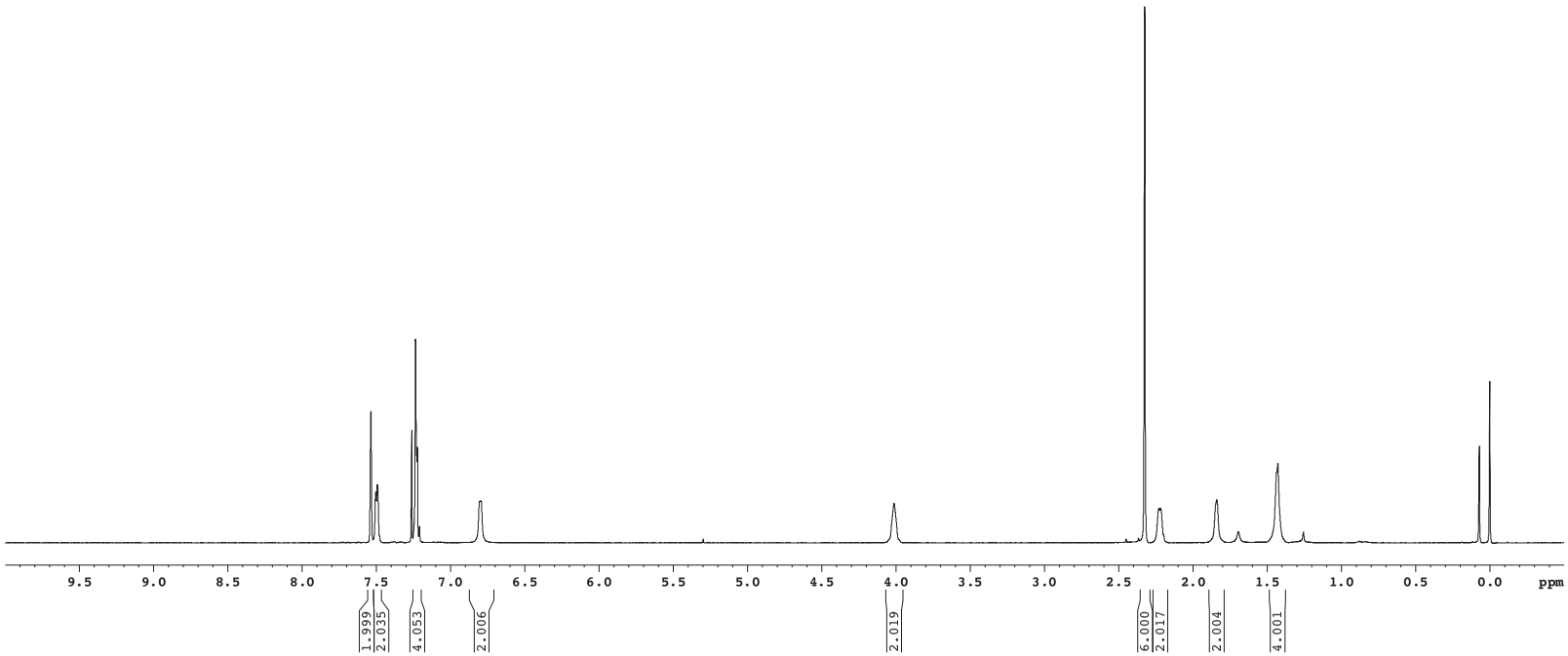
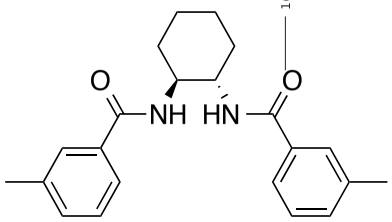


Z–restored spin–echo 13C spectrum with 1H decoupling



1.11-(*S,S*)


```
Current Data Parameters
USER                samame
NAME          RAS_1_83_pure
EXPNO                     2
PROCNO                    1

F2 - Acquisition Parameters
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Time                17.12
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PULPROG  SpinEchopg30gp.prd
TD                  65536
SOLVENT             CDCl3
NS                    444
DS                     16
SWH             30303.031 Hz
FIDRES          0.462388 Hz
AQ              1.0813940 sec
RG                 7298.2
DW                16.500 usec
DE                 6.00 usec
TE                298.0 K
D1             0.25000000 sec
d11            0.03000000 sec
D16            0.00020000 sec
d17            0.00019600 sec
MCREST         0.00000000 sec
MCWRK          0.01500000 sec
P2                31.00 usec

======== CHANNEL f1 ========
NUC1                 13C
P1                15.50 usec
P11              500.00 usec
P12             2000.00 usec
PL0              120.00 dB
PL1               -1.00 dB
SFO1         125.7942548 MHz
SP1                3.20 dB
SP2                3.20 dB
SPNAM1    Crp60,0.5,20.1
SPNAM2      Crp60comp.4
SPOFF1               0.00 Hz
SPOFF2               0.00 Hz

======== CHANNEL f2 ========
CPDPRG2           waltz16
NUC2                 1H
PCPD2             100.00 usec
PL2                1.60 dB
PL12              24.60 dB
SFO2         500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1          SINE.100
GPNAM2          SINE.100
GPX1                0.00 %
GPX2                0.00 %
GPY1                0.00 %
GPY2                0.00 %
GPZ1               30.00 %
GPZ2               50.00 %
p15               500.00 usec
p16              1000.00 usec

F2 - Processing parameters
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SF           125.7803992 MHz
WDW                   no
SSB                    0
LB                  0.00 Hz
GB                     0
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```
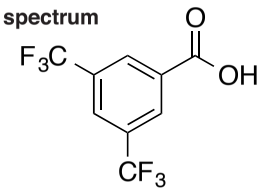

1H spectrum



1.18

8.580
8.146
7.260
7.258

```
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EXPNO                    1
PROCNO                   1

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Time                 12.11
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TD                  81728
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NS                      8
DS                      2
SWH              8012.820 Hz
FIDRES           0.098043 Hz
AQ               5.0998774 sec
RG                    6.3
DW                 62.400 usec
DE                  6.00 usec
TE                298.0 K
D1             0.10000000 sec
MCREST         0.00000000 sec
MCWRK          0.01500000 sec

======== CHANNEL f1 ========
NUC1                   1H
P1                   7.50 usec
PL1                  1.60 dB
SFO1          500.2235015 MHz

F2 - Processing parameters
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WDW                     no
SSB                      0
LB                    0.00 Hz
GB                       0
PC                    4.00
```
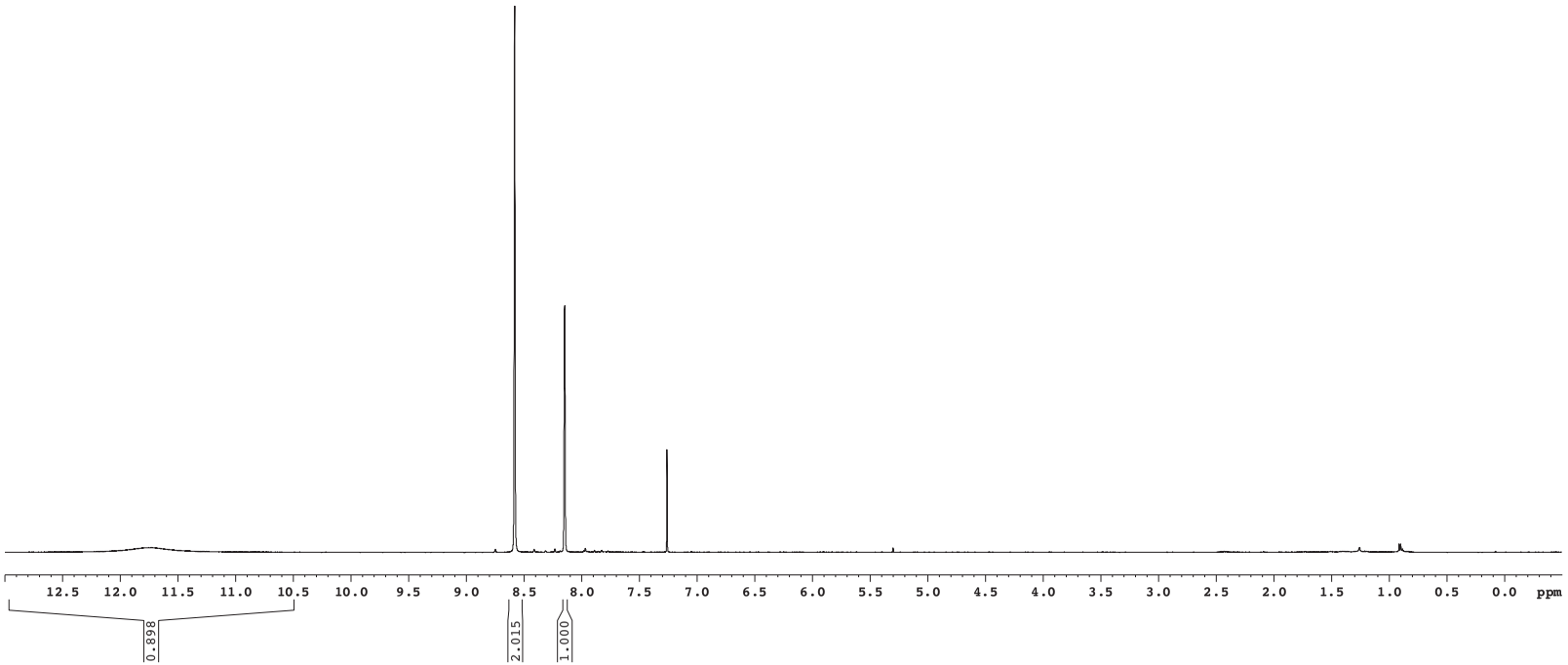
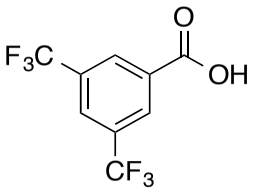



Z–restored spin–echo 13C spectrum with 1H decoupling



1.18

169.811

133.164
132.902
132.891
132.630
132.619
132.347
131.482
131.465
130.601
130.578
130.572
127.705
127.675
127.646
127.617
127.588
126.213
124.043
121.873
119.704

77.483
77.229
76.975

```
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PROCNO                   1

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TD                   65536
SOLVENT              CDCl3
NS                     295
DS                      16
SWH              30303.031 Hz
FIDRES           0.462388 Hz
AQ               1.0813940 sec
RG                  7298.2
DW                  16.500 usec
DE                   6.00 usec
TE                  298.0 K
D1               0.25000000 sec
d11              0.03000000 sec
D16              0.00020000 sec
d17              0.00019600 sec
MCREST           0.00000000 sec
MCWRK            0.01500000 sec
P2                  31.00 usec

======== CHANNEL f1 ========
NUC1                   13C
P1                  15.50 usec
P11                500.00 usec
P12               2000.00 usec
PL0                120.00 dB
PL1                 -1.00 dB
SFO1          125.7942548 MHz
SP1                  3.20 dB
SP2                  3.20 dB
SPNAM1     Crp60,0.5,20.1
SPNAM2       Crp60comp.4
SPOFF1               0.00 Hz
SPOFF2               0.00 Hz

======== CHANNEL f2 ========
CPDPRG2            waltz16
NUC2                   1H
PCPD2               100.00 usec
PL2                  1.60 dB
PL12                24.60 dB
SFO2          500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1           SINE.100
GPNAM2           SINE.100
GPX1                 0.00 %
GPX2                 0.00 %
GPY1                 0.00 %
GPY2                 0.00 %
GPZ1                30.00 %
GPZ2                50.00 %
p15                500.00 usec
p16               1000.00 usec

F2 - Processing parameters
SI                   65536
SF            125.7803955 MHz
WDW                    no
SSB                     0
LB                   0.00 Hz
GB                      0
PC                   2.00
```
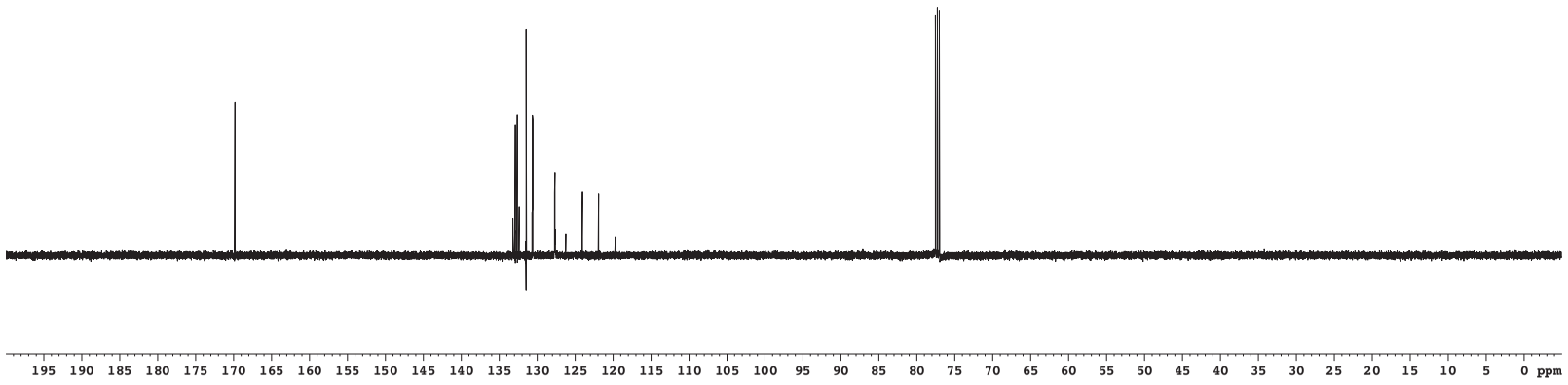
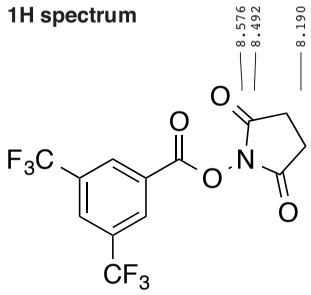
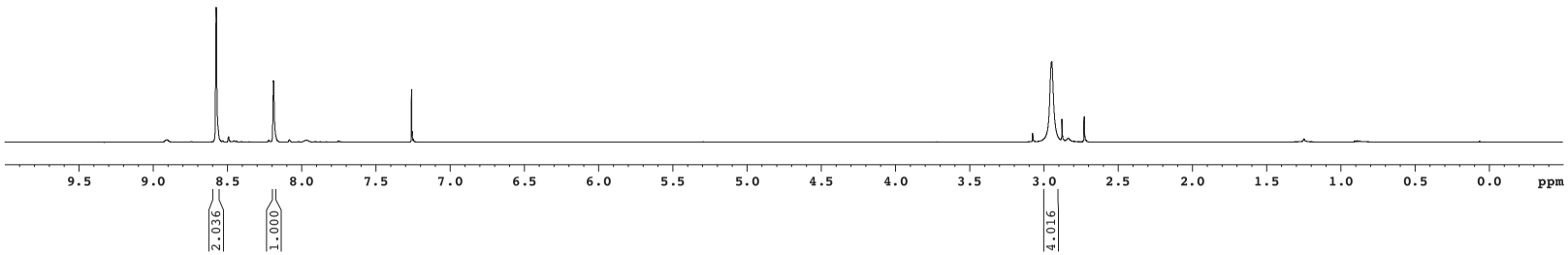



1H spectrum

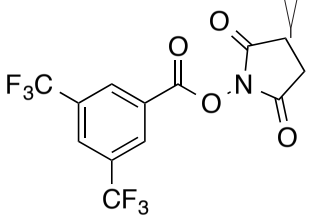


1.21

```
Current Data Parameters
USER                  samame
NAME               RAS_1_52
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
Date_              20110725
Time                 18.55
INSTRUM            cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG               zg30
TD                   81728
SOLVENT              CDCl3
NS                      10
DS                       2
SWH                8012.820 Hz
FIDRES             0.098043 Hz
AQ                 5.0998774 sec
RG                       7.1
DW                    62.400 usec
DE                     6.00 usec
TE                    298.0 K
D1                 0.10000000 sec
MCREST             0.00000000 sec
MCWRK              0.01500000 sec

======== CHANNEL f1 ========
NUC1                     1H
P1                    7.50 usec
PL1                   1.60 dB
SFO1           500.2235015 MHz

F2 - Processing parameters
SI                       65536
SF             500.2200305 MHz
WDW                      no
SSB                       0
LB                     0.00 Hz
GB                        0
PC                     4.00
```



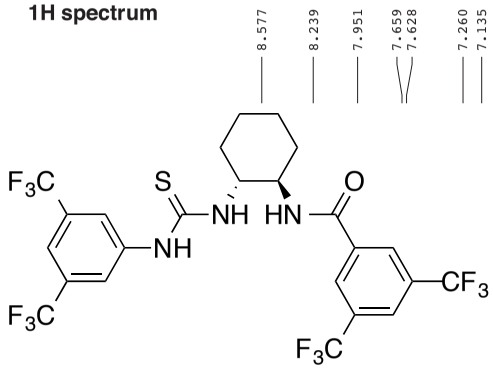
Z–restored spin–echo 13C spectrum with 1H decoupling



1.21


```
Current Data Parameters
USER              samame
NAME            RAS_1_52
EXPNO                  2
PROCNO                 1

F2 - Acquisition Parameters
Date_           20110725
Time              19.01
INSTRUM          cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG  SpinEchopg30gp.prd
TD                65536
SOLVENT            CDCl3
NS                   224
DS                    16
SWH           30303.031 Hz
FIDRES        0.462388 Hz
AQ            1.0813940 sec
RG               2580.3
DW               16.500 usec
DE                6.00 usec
TE                298.0 K
D1           0.25000000 sec
d11          0.03000000 sec
D16          0.00020000 sec
d17          0.00019600 sec
MCREST       0.00000000 sec
MCWRK        0.01500000 sec
P2                31.00 usec

======== CHANNEL f1 ========
NUC1                13C
P1                15.50 usec
P11              500.00 usec
P12             2000.00 usec
PL0              120.00 dB
PL1               -1.00 dB
SFO1        125.7942548 MHz
SP1               3.20 dB
SP2               3.20 dB
SPNAM1    Crp60,0.5,20.1
SPNAM2      Crp60comp.4
SPOFF1               0.00 Hz
SPOFF2               0.00 Hz

======== CHANNEL f2 ========
CPDPRG2          waltz16
NUC2                1H
PCPD2             100.00 usec
PL2                1.60 dB
PL12              24.60 dB
SFO2        500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1          SINE.100
GPNAM2          SINE.100
GPX1               0.00 %
GPX2               0.00 %
GPY1               0.00 %
GPY2               0.00 %
GPZ1              30.00 %
GPZ2              50.00 %
p15              500.00 usec
p16             1000.00 usec

F2 - Processing parameters
SI                 65536
SF          125.7803988 MHz
WDW                   no
SSB                    0
LB                0.00 Hz
GB                     0
PC                2.00
```

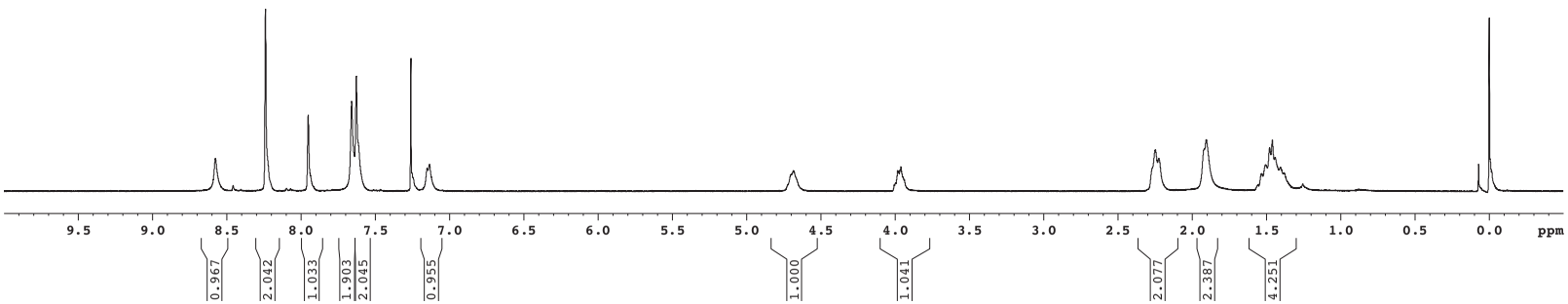
1H spectrum



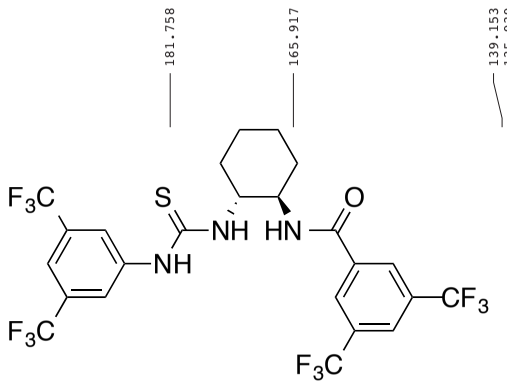
1.6-(*R,R*)

8.577 8.239 7.951 7.659 7.628 7.260 7.135 4.685 3.982 3.960 2.248 2.225 1.903 1.531 1.506 1.477 1.458 1.403 1.253 0.069 -0.002

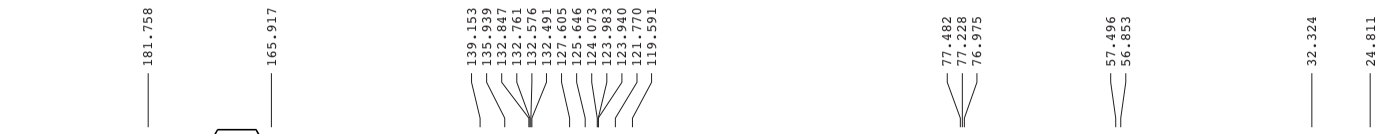
```
Current Data Parameters
USER                samame
NAME         RAS_1_77pure
EXPNO                  1
PROCNO                 1

F2 - Acquisition Parameters
Date_           20111029
Time               11.35
INSTRUM          cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG             zg30
TD                 81728
SOLVENT            CDCl3
NS                     8
DS                     2
SWH             8012.820 Hz
FIDRES             0.098043 Hz
AQ              5.0998774 sec
RG                   7.1
DW                  62.400 usec
DE                   6.00 usec
TE                  298.0 K
D1              0.10000000 sec
MCREST          0.00000000 sec
MCWRK           0.01500000 sec

======== CHANNEL f1 ========
NUC1                   1H
P1                   7.50 usec
PL1                  1.60 dB
SFO1          500.2235015 MHz

F2 - Processing parameters
SI                    65536
SP             500.2200324 MHz
WDW                   no
SSB                    0
LB                  0.00 Hz
GB                     0
PC                  4.00
```



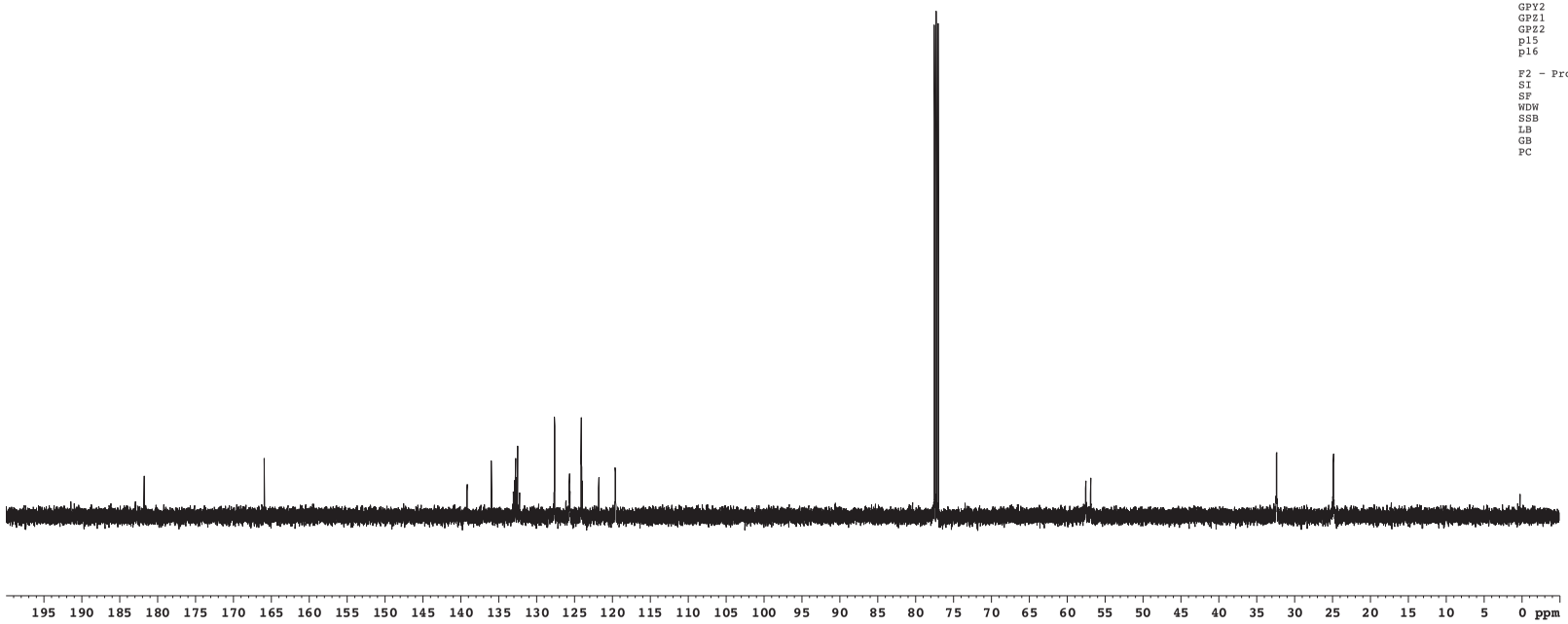
Z–restored spin–echo 13C spectrum with 1H decoupling



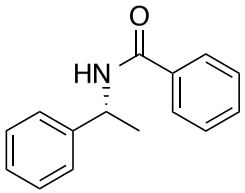
1.6-(*R,R*)




```
Current Data Parameters
USER              samame
NAME        RAS_1_77pure
EXPNO                2
PROCNO               1

F2 - Acquisition Parameters
Date_          20111029
Time              11.41
INSTRUM         cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG  SpinEchopg30gp.prd
TD               65536
SOLVENT           CDCl3
NS                 784
DS                  16
SWH          30303.031 Hz
FIDRES        0.462388 Hz
AQ            1.0813940 sec
RG             4597.6
DW              16.500 usec
DE               6.00 usec
TE              298.0 K
D1           0.25000000 sec
d11          0.03000000 sec
D16          0.00020000 sec
d17          0.00019600 sec
MCREST       0.00000000 sec
MCWRK        0.01500000 sec
P2              31.00 usec

======== CHANNEL f1 ========
NUC1               13C
P1              15.50 usec
P11            500.00 usec
P12           2000.00 usec
PL0            120.00 dB
PL1             -1.00 dB
SFO1       125.7942548 MHz
SP1              3.20 dB
SP2              3.20 dB
SPNAM1   Crp60,0.5,20.1
SPNAM2     Crp60comp.4
SPOFF1           0.00 Hz
SPOFF2           0.00 Hz

======== CHANNEL f2 ========
CPDPRG2         waltz16
NUC2               1H
PCPD2          100.00 usec
PL2              1.60 dB
PL12            24.60 dB
SFO2       500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1         SINE.100
GPNAM2         SINE.100
GPX1             0.00 %
GPX2             0.00 %
GPY1             0.00 %
GPY2             0.00 %
GPZ1            30.00 %
GPZ2            50.00 %
p15            500.00 usec
p16           1000.00 usec

F2 - Processing parameters
SI               65536
SF         125.7803997 MHz
WDW                no
SSB                 0
LB               0.00 Hz
GB                  0
PC               2.00
```



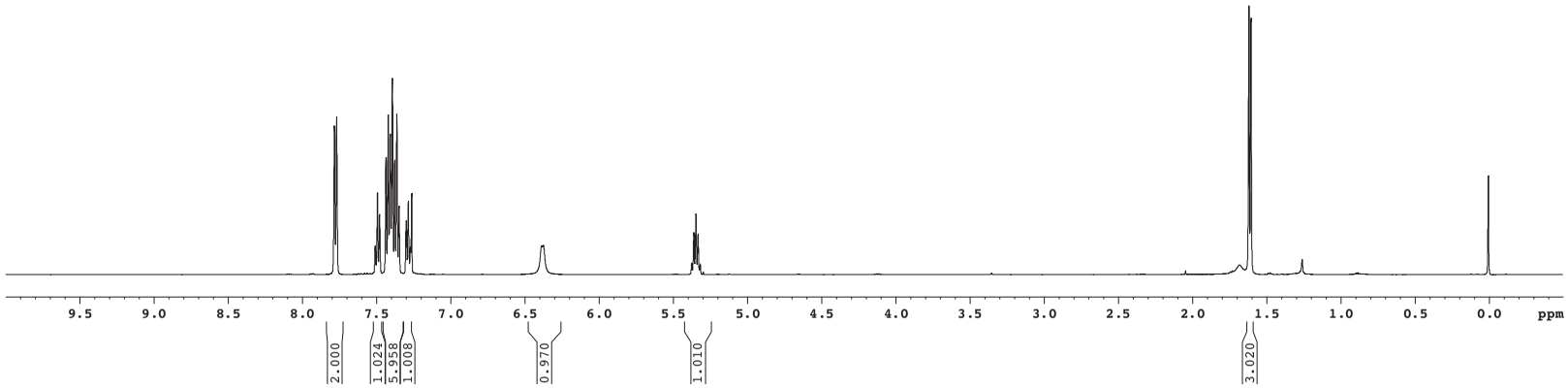
1H spectrum



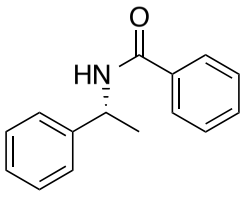
1.28

7.782
7.766
7.505
7.491
7.476
7.434
7.419
7.405
7.391
7.375
7.361
7.345
7.297
7.283
7.269
7.260
6.385
6.373
5.373
5.359
5.345
5.330
5.316
1.681
1.617
1.604
1.259
0.004

```
Current Data Parameters
USER                samame
NAME        RAS_1_90_PURE
EXPNO                   1
PROCNO                  1

F2 - Acquisition Parameters
Date_            20111025
Time                10.00
INSTRUM           cryo500
PROBHD    5 mm CPTCI 1H-
PULPROG              zg30
TD                  81728
SOLVENT             CDCl3
NS                      8
DS                      2
SWH              8012.820 Hz
FIDRES           0.098043 Hz
AQ               5.0998774 sec
RG                      8
DW                 62.400 usec
DE                  6.00 usec
TE                 298.0 K
D1              0.10000000 sec
MCREST          0.00000000 sec
MCWRK           0.01500000 sec

======== CHANNEL f1 ========
NUC1                   1H
P1                  7.50 usec
PL1                 1.60 dB
SFO1          500.2235015 MHz

F2 - Processing parameters
SI                  65536
SF            500.2200319 MHz
WDW                    no
SSB                     0
LB                   0.00 Hz
GB                      0
PC                   4.00
```



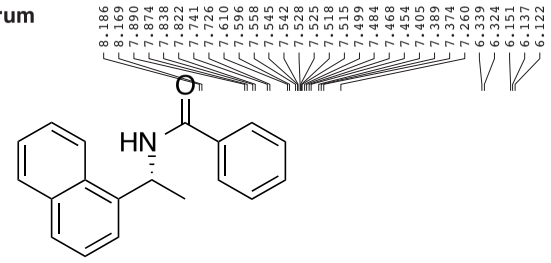
Z–restored spin–echo 13C spectrum with 1H decoupling



1.28

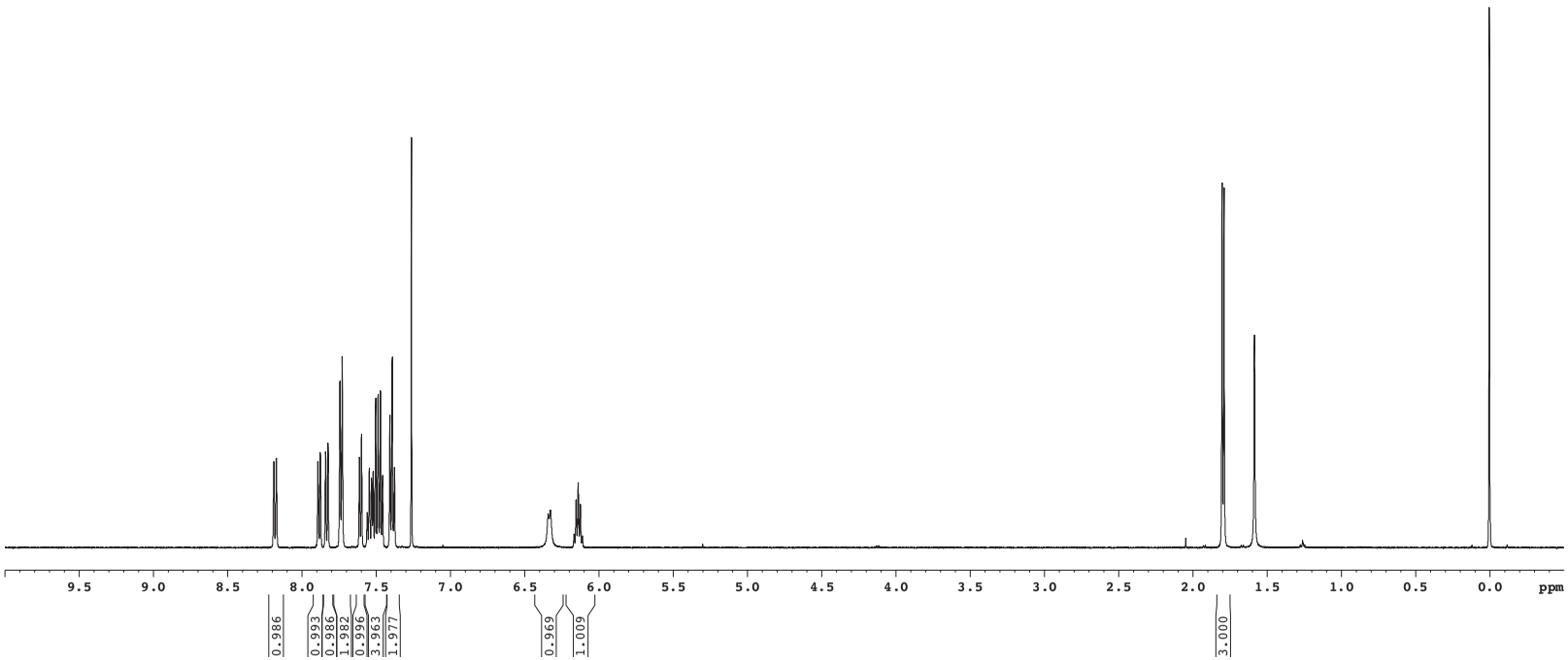

```
Current Data Parameters
USER              samame
NAME        RAS_1_90_PURE
EXPNO                    2
PROCNO                   1

F2 - Acquisition Parameters
Date_              20111025
Time                 10.01
INSTRUM            cryo500
PROBHD    5 mm CPTCI 1H-
PULPROG   SpinEchopg30gp.prd
TD                   65536
SOLVENT              CDCl3
NS                     350
DS                      16
SWH             30303.031 Hz
FIDRES          0.462388 Hz
AQ              1.0813940 sec
RG                 11585.2
DW                 16.500 usec
DE                  6.00 usec
TE                 298.0 K
D1              0.25000000 sec
d11             0.03000000 sec
D16             0.00020000 sec
d17             0.00019600 sec
MCREST          0.00000000 sec
MCWRK           0.01500000 sec
P2                 31.00 usec

======== CHANNEL f1 ========
NUC1                   13C
P1                  15.50 usec
P11                500.00 usec
P12               2000.00 usec
PL0                120.00 dB
PL1                 -1.00 dB
SFO1         125.7942548 MHz
SP1                  3.20 dB
SP2                  3.20 dB
SPNAM1    Crp60,0.5,20.1
SPNAM2       Crp60comp.4
SPOFF1               0.00 Hz
SPOFF2               0.00 Hz

======== CHANNEL f2 ========
CPDPRG2            waltz16
NUC2                   1H
PCPD2              100.00 usec
PL2                  1.60 dB
PL12                24.60 dB
SFO2         500.2225011 MHz

===== GRADIENT CHANNEL =====
GPNAM1           SINE.100
GPNAM2           SINE.100
GPX1                 0.00 %
GPX2                 0.00 %
GPY1                 0.00 %
GPY2                 0.00 %
GPZ1                30.00 %
GPZ2                50.00 %
p15                500.00 usec
p16               1000.00 usec

F2 - Processing parameters
SI                   65536
SF           125.7804025 MHz
WDW                     no
SSB                      0
LB                  0.00 Hz
GB                       0
PC                    2.00
```

1H spectrum

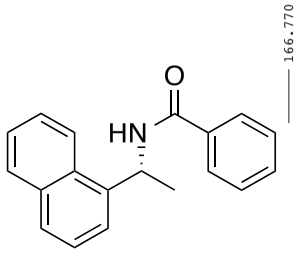


1.29


```
Current Data Parameters
USER                samame
NAME          RAS_1_89_PURE
EXPNO                   1
PROCNO                  1

F2 - Acquisition Parameters
Date_            20111025
Time                9.44
INSTRUM          cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG             zg30
TD                 81728
SOLVENT            CDCl3
NS                     8
DS                     2
SWH             8012.820 Hz
FIDRES          0.098043 Hz
AQ              5.0998774 sec
RG                   6.3
DW                62.400 usec
DE                 6.00 usec
TE                298.0 K
D1           0.10000000 sec
MCREST       0.00000000 sec
MCWRK        0.01500000 sec

======== CHANNEL f1 ========
NUC1                  1H
P1                 7.50 usec
PL1                1.60 dB
SFO1         500.2235015 MHz

F2 - Processing parameters
SI                 65536
SF           500.2200323 MHz
WDW                   no
SSB                    0
LB                  0.00 Hz
GB                     0
PC                  4.00
```



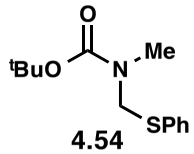
Z–restored spin–echo 13C spectrum with 1H decoupling



1.29


```
Current Data Parameters
USER                samame
NAME          RAS_1_90_PURE
EXPNO                     2
PROCNO                    1

F2 - Acquisition Parameters
Date_              20111025
Time                 10.01
INSTRUM            cryo500
PROBHD   5 mm CPTCI 1H-
PULPROG  SpinEchopg30gp.prd
TD                    65536
SOLVENT               CDCl3
NS                      350
DS                       16
SWH               30303.031 Hz
FIDRES            0.462388 Hz
AQ                1.0813940 sec
RG                  11585.2
DW                   16.500 usec
DE                    6.00 usec
TE                  298.0 K
D1                0.25000000 sec
d11               0.03000000 sec
D16               0.00020000 sec
d17               0.00019600 sec
MCREST            0.00000000 sec
MCWRK             0.01500000 sec
P2                   31.00 usec

======== CHANNEL f1 ========
NUC1                    13C
P1                   15.50 usec
P11                 500.00 usec
P12                2000.00 usec
PL0                 120.00 dB
PL1                  -1.00 dB
SFO1           125.7942548 MHz
SP1                   3.20 dB
SP2                   3.20 dB
SPNAM1    Crp60,0.5,20.1
SPNAM2       Crp60comp.4
SPOFF1                0.00 Hz
SPOFF2                0.00 Hz

======== CHANNEL f2 ========
CPDPRG2             waltz16
NUC2                    1H
PCPD2                100.00 usec
PL2                   1.60 dB
PL12                 24.60 dB
SFO2           500.2225011 MHz

====== GRADIENT CHANNEL =====
GPNAM1            SINE.100
GPNAM2            SINE.100
GPX1                  0.00 %
GPX2                  0.00 %
GPY1                  0.00 %
GPY2                  0.00 %
GPZ1                 30.00 %
GPZ2                 50.00 %
p15                 500.00 usec
p16                1000.00 usec

F2 - Processing parameters
SI                    65536
SF             125.7804025 MHz
WDW                     no
SSB                      0
LB                    0.00 Hz
GB                       0
PC                    2.00
```

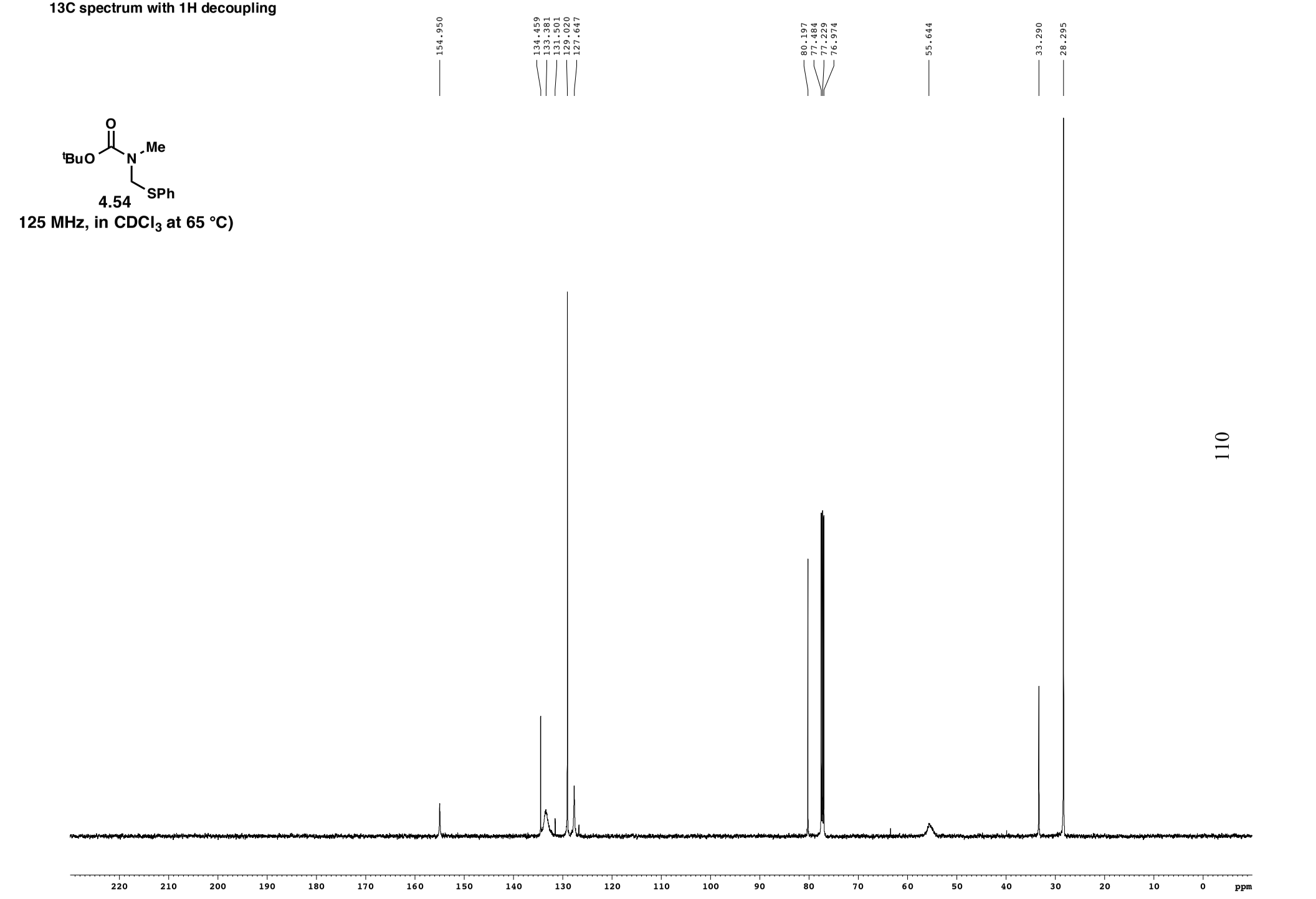
1H spectrum



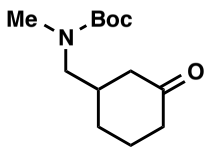
4.54

(500 MHz, in $CDCl_3$ at 65 °C)

13C spectrum with 1H decoupling



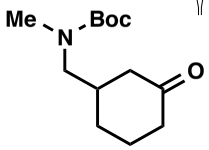
1H spectrum



4.51

(500 MHz, in $CDCl_3$ at 25 °C)

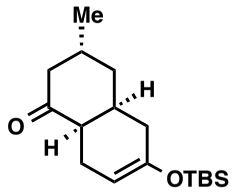
Z-restored spin-echo 13C spectrum with 1H decoupling



4.51

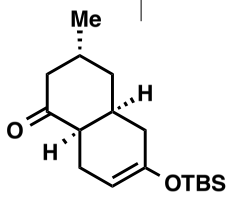
125 MHz, in $CDCl_3$ at 25 °C)

1H spectrum



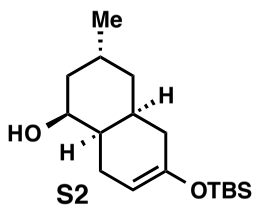
4.30
(500 MHz, in $CDCl_3$ at 25 °C)

Z–restored spin–echo 13C spectrum with 1H decoupling



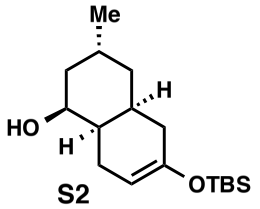
4.30
(125 MHz, in $CDCl_3$ at 25 °C)

1H spectrum



(500 MHz, in $CDCl_3$ at 25 °C)

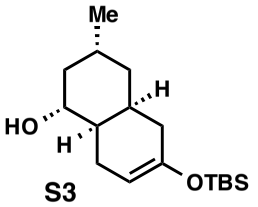
Z-restored spin-echo 13C spectrum with 1H decoupling



S2

125 MHz, in $CDCl_3$ at 25 °C)

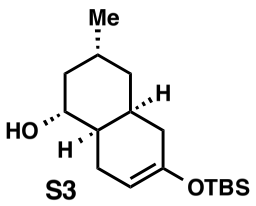
1H spectrum



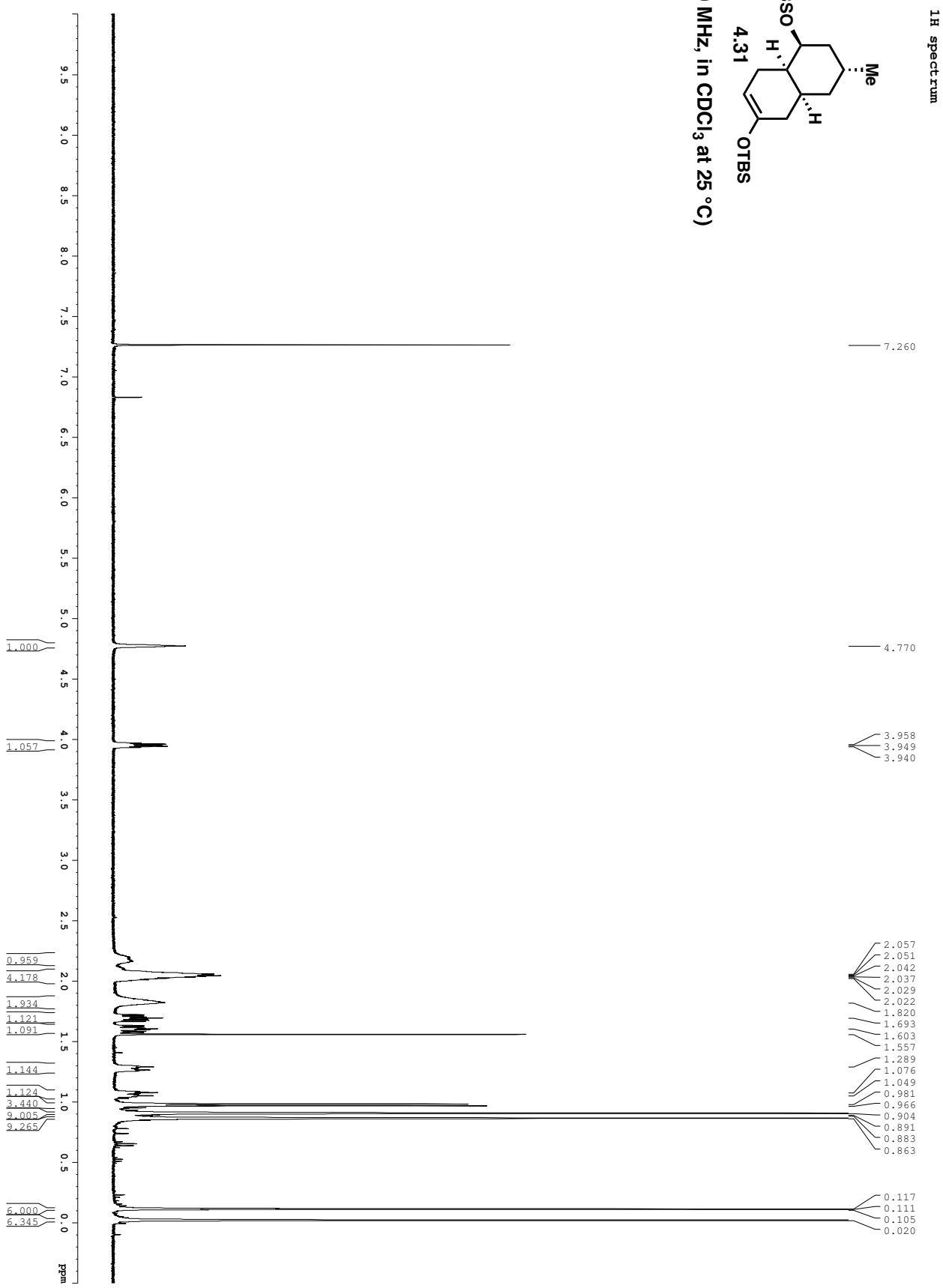
S3

(500 MHz, in $CDCl_3$ at 25 °C)

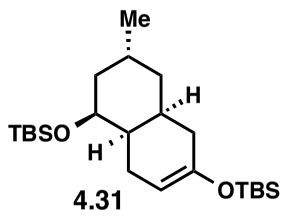
Z-restored spin-echo 13C spectrum with 1H decoupling



(125 MHz, in $CDCl_3$ at 25 °C)

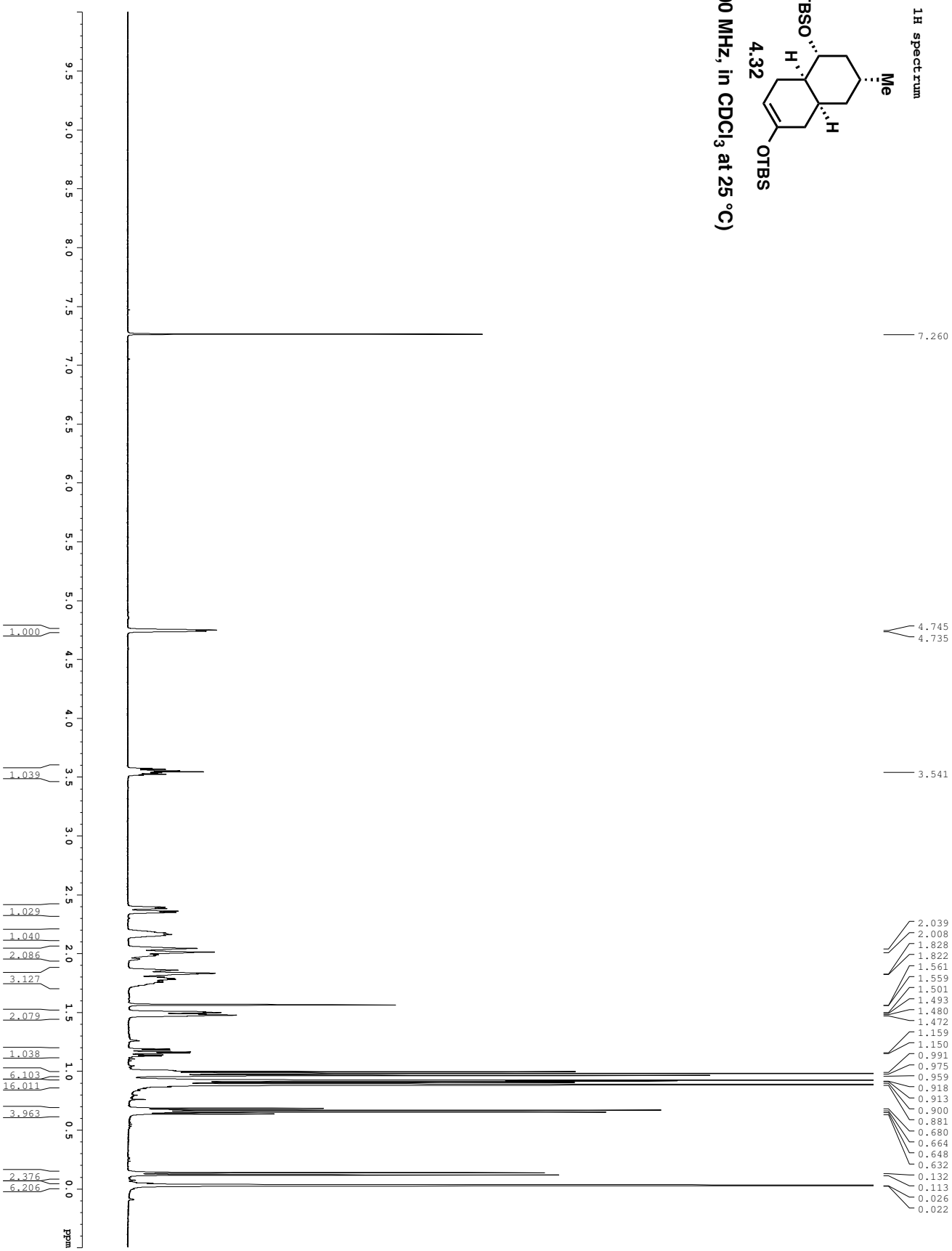

1H spectrum
TBSO
H
Me
H
OTBS
4.31
(500 MHz, in CDCl3 at 25 °C)
7.260
4.770
3.958
3.949
3.940
2.057
2.051
2.042
2.037
2.029
2.022
1.820
1.693
1.603
1.557
1.289
1.076
1.049
0.981
0.966
0.904
0.891
0.883
0.863
0.117
0.111
0.105
0.020
1.000
1.057
0.959
4.178
1.934
1.121
1.091
1.144
1.124
3.440
9.005
9.265
6.000
6.345
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling

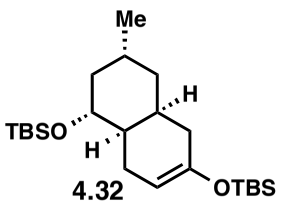


4.31

(125 MHz, in $CDCl_3$ at 25 °C)

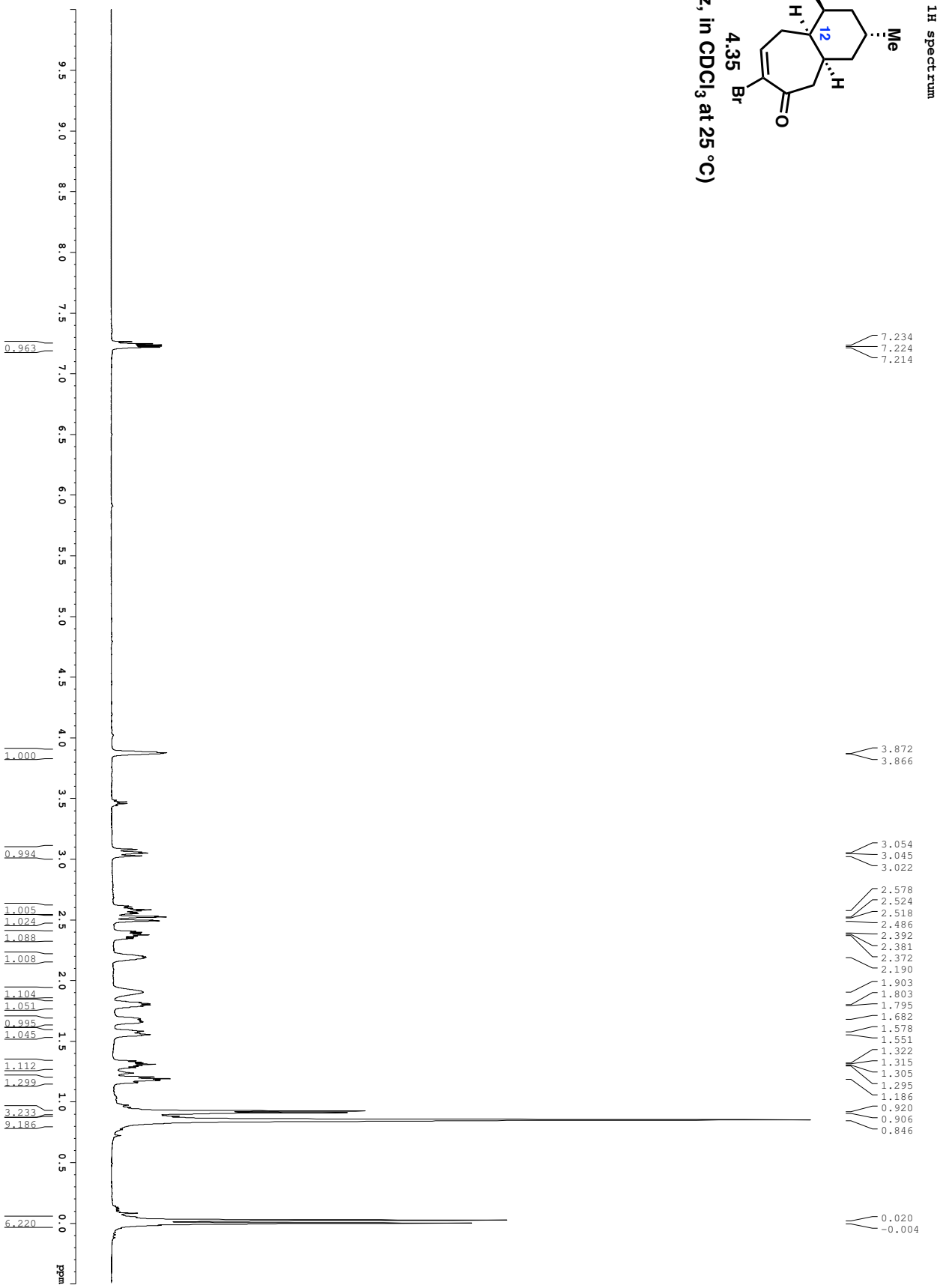

1H spectrum
Me
TBSO
H
H
OTBS
4.32
(500 MHz, in CDCl3 at 25 °C)
7.260
4.745
4.735
3.541
2.039
2.008
1.828
1.822
1.561
1.559
1.501
1.493
1.480
1.472
1.159
1.150
0.991
0.975
0.959
0.918
0.913
0.900
0.881
0.680
0.664
0.648
0.632
0.132
0.113
0.026
0.022
1.000
1.039
1.029
1.040
2.086
3.127
2.079
1.038
6.103
16.011
3.963
2.376
6.206
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling

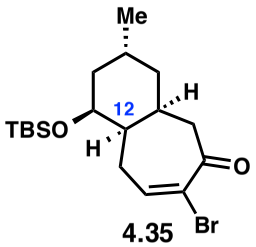


4.32

125 MHz, in $CDCl_3$ at 25 °C)


1H spectrum
TBSO
H
12
Me
H
4.35
Br
O
(500 MHz, in CDCl3 at 25 °C)
0.963
1.000
0.994
1.005
1.024
1.088
1.008
1.104
1.051
0.995
1.045
1.112
1.299
3.233
9.186
6.220
7.234
7.224
7.214
3.872
3.866
3.054
3.045
3.022
2.578
2.524
2.518
2.486
2.392
2.381
2.372
2.190
1.903
1.803
1.795
1.682
1.578
1.551
1.322
1.315
1.305
1.295
1.186
0.920
0.906
0.846
0.020
-0.004
ppm

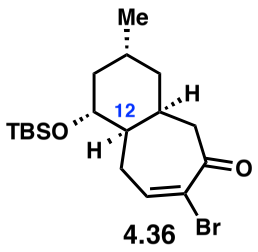
Z-restored spin-echo 13C spectrum with 1H decoupling



4.35

(125 MHz, in $CDCl_3$ at 25 °C)

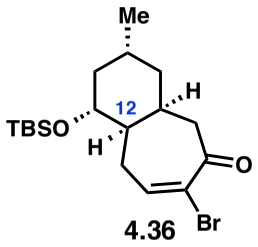
1H spectrum



4.36

00 MHz, in $CDCl_3$ at 25 °C)

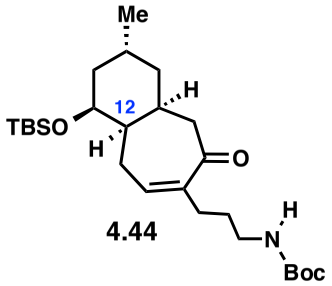
Z-restored spin-echo 13C spectrum with 1H decoupling



4.36

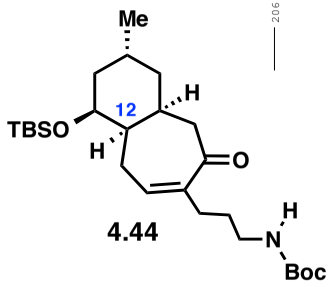
25 MHz, in $CDCl_3$ at 25 °C)

1H spectrum



500 MHz, in $CDCl_3$ at 25 °C)

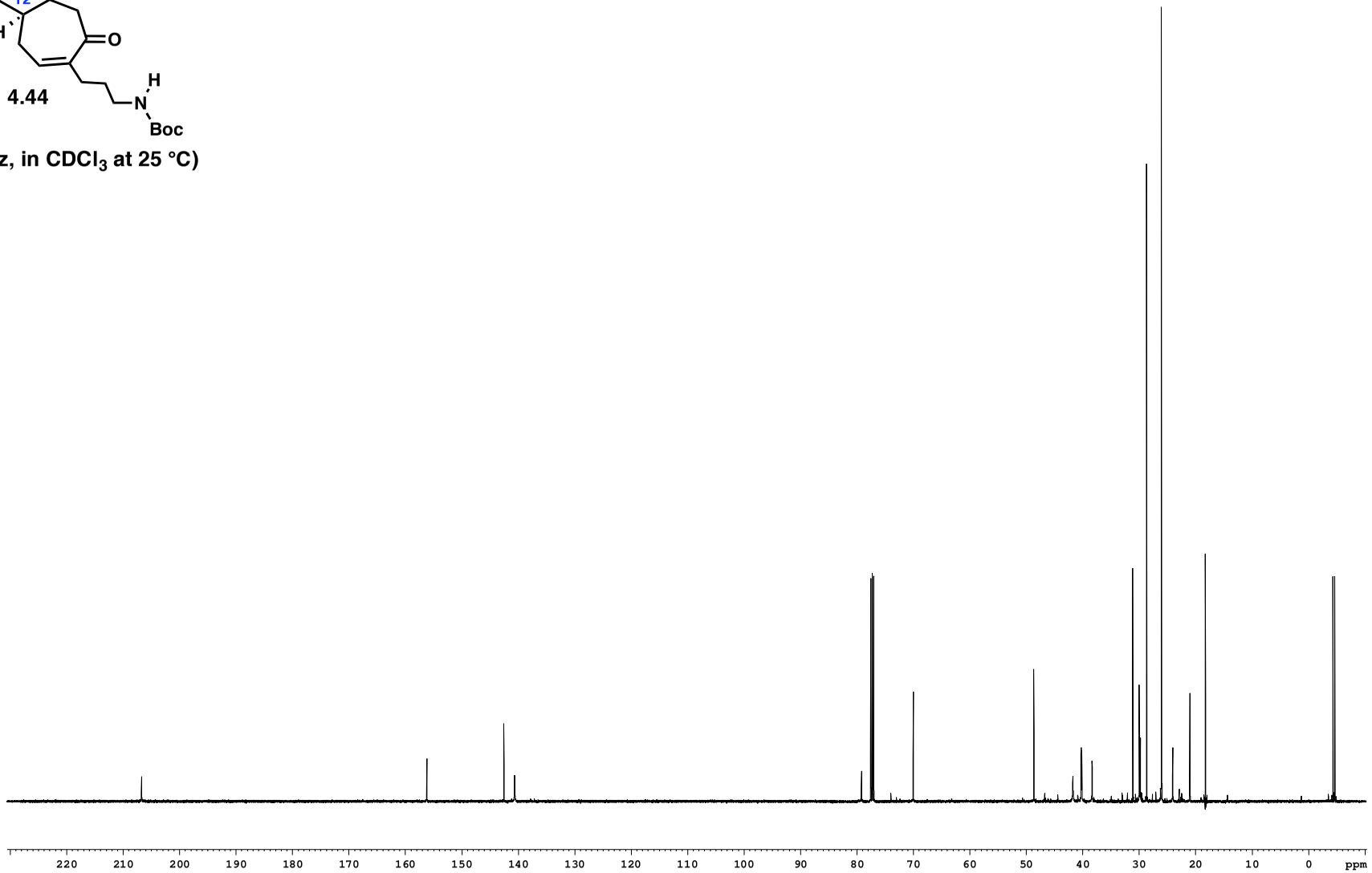
Z-restored spin-echo 13C spectrum with 1H decoupling

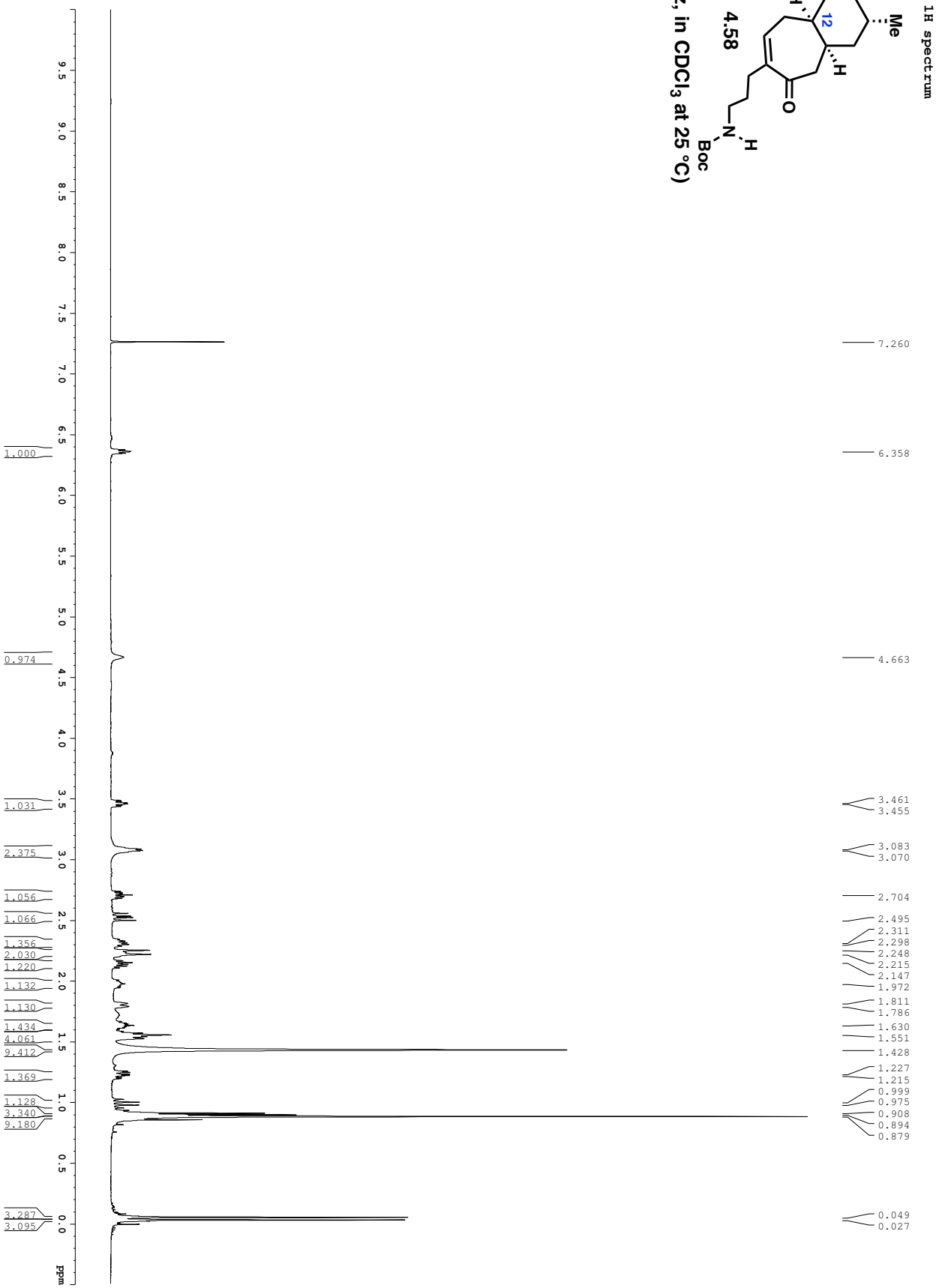


4.44

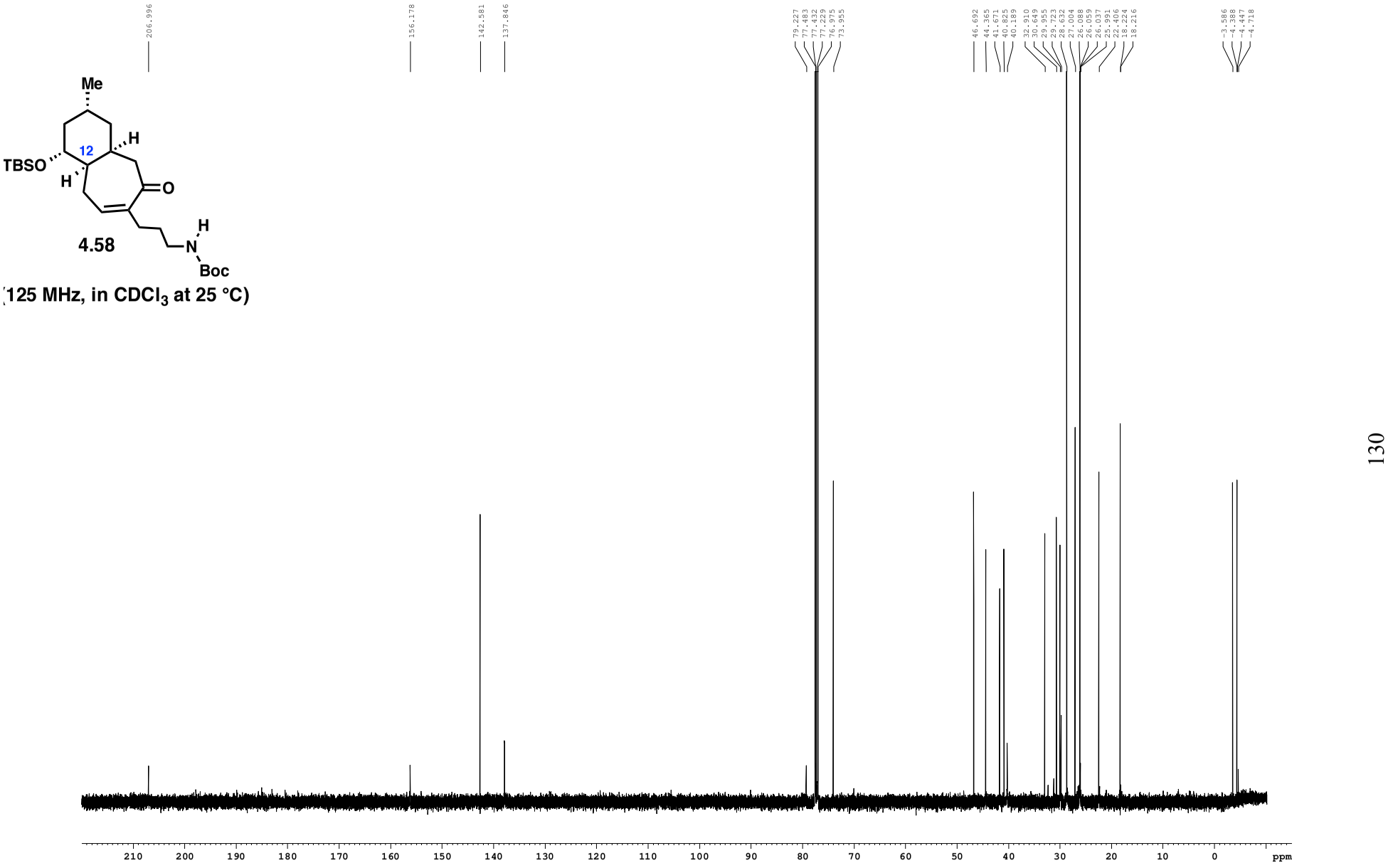
(125 MHz, in $CDCl_3$ at 25 °C)

206.713
156.158
142.502
140.609
79.126
77.485
77.231
76.977
69.959
48.591
41.657
40.222
40.098
38.271
31.111
29.930
29.701
28.609
28.034
26.011
25.964
25.925
23.982
20.930
18.184
18.172
-4.413
-4.745



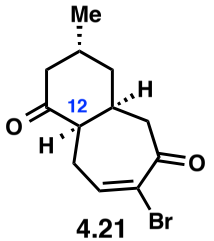

1H spectrum
Me
TBSO
H
12
H
4.58
O
N
H
Boc
500 MHz, in CDCl3 at 25 °C)
7.260
6.358
4.663
3.461
3.455
3.083
3.070
2.704
2.495
2.311
2.298
2.248
2.215
2.147
1.972
1.811
1.786
1.630
1.551
1.428
1.227
1.215
0.999
0.975
0.908
0.894
0.879
0.049
0.027
1.000
0.974
1.031
2.375
1.056
1.066
1.356
2.030
1.220
1.132
1.130
1.434
4.061
9.412
1.369
1.128
3.340
9.180
3.287
3.095
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling



(125 MHz, in $CDCl_3$ at 25 °C)

1H spectrum

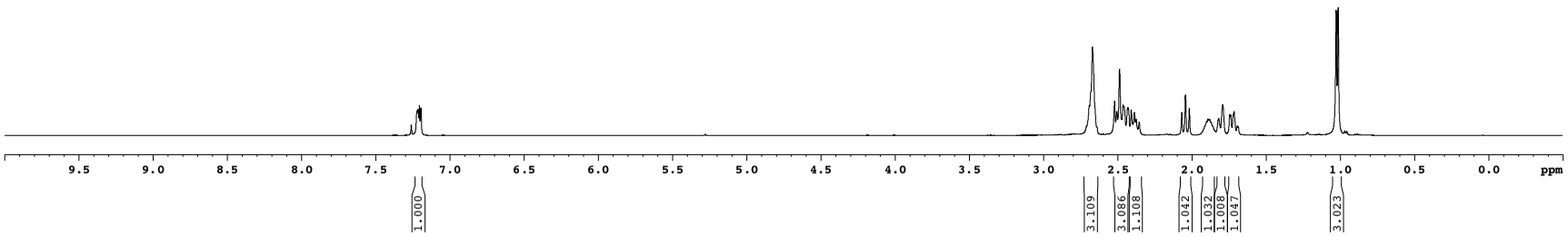


4.21

00 MHz, in $CDCl_3$ at 25 °C)

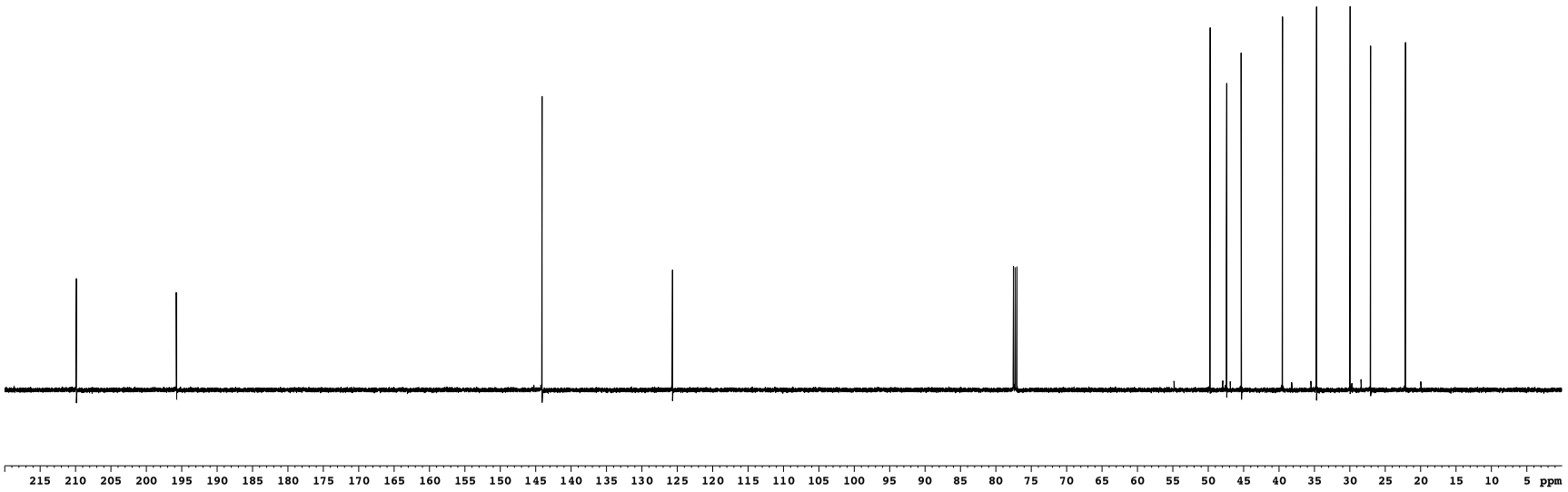
7.260
7.223
7.217
7.205
7.195

2.691
2.669
2.520
2.504
2.486
2.462
2.431
2.407
2.387
2.374
2.354
2.067
2.042
2.016
1.891
1.879
1.870
1.820
1.792
1.744
1.737
1.715
1.694
1.687
1.027
1.014
0.969

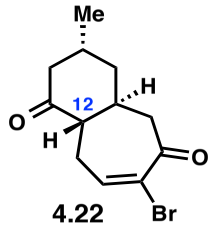


Z–restored spin–echo 13C spectrum with 1H decoupling

25 MHz, in $CDCl_3$ at 25 °C)

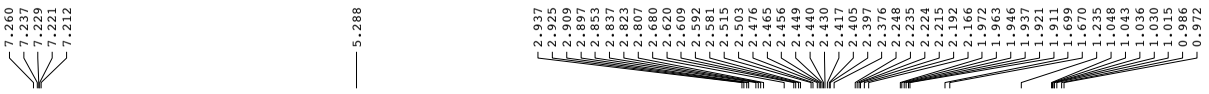


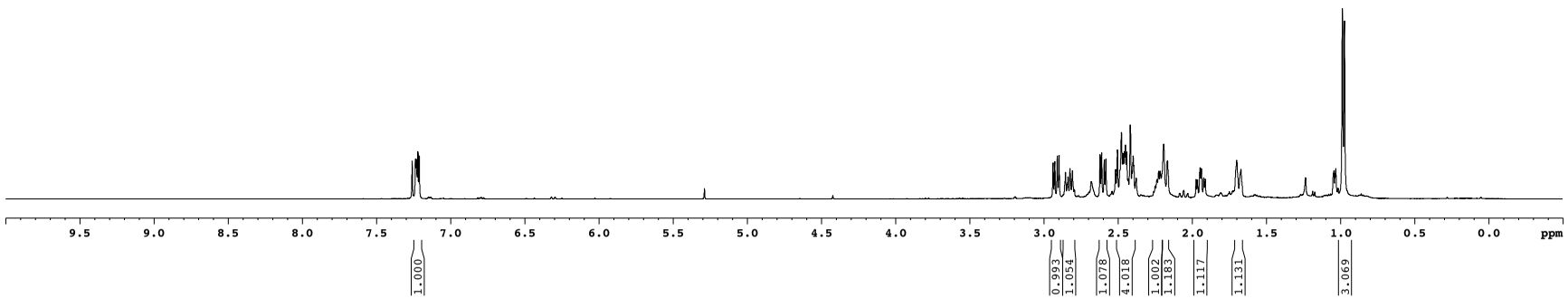
1H spectrum



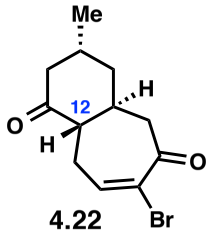
4.22

500 MHz, in $CDCl_3$ at 25 °C)





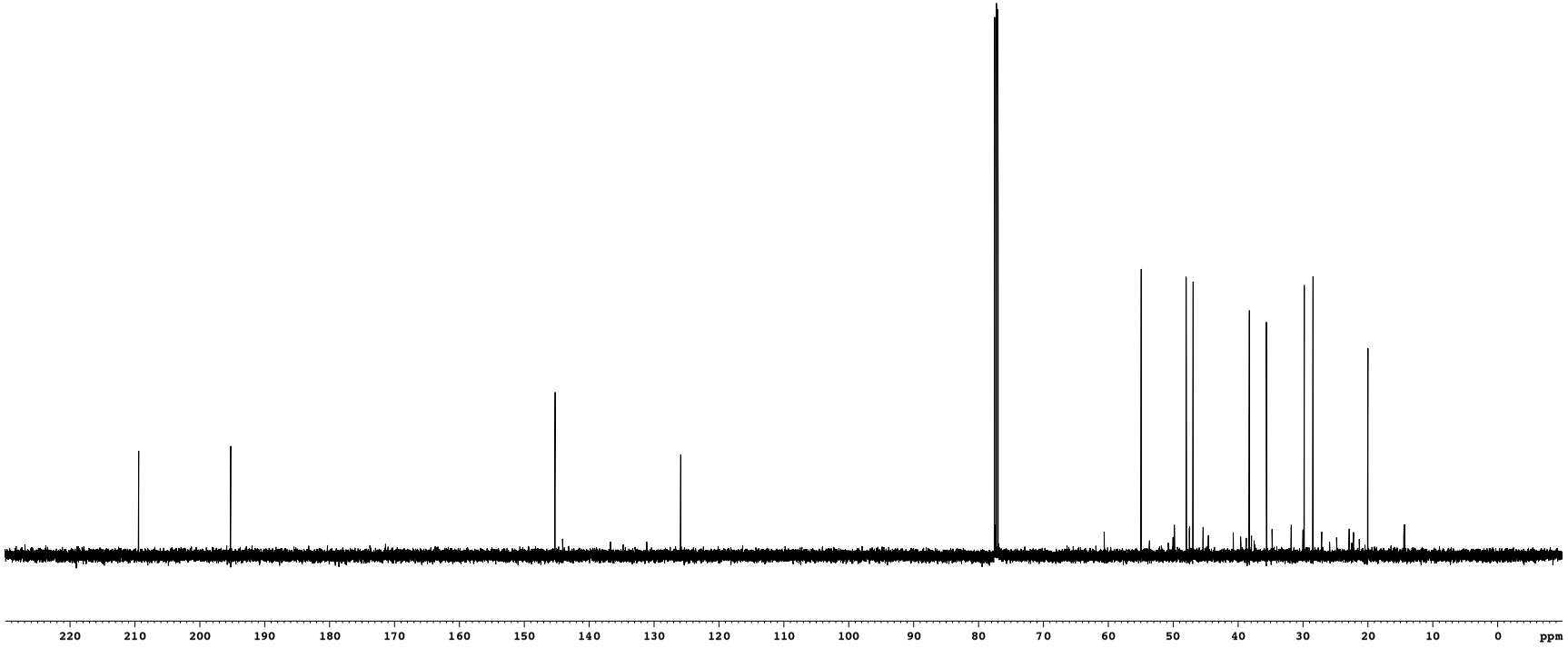
Z–restored spin–echo 13C spectrum with 1H decoupling

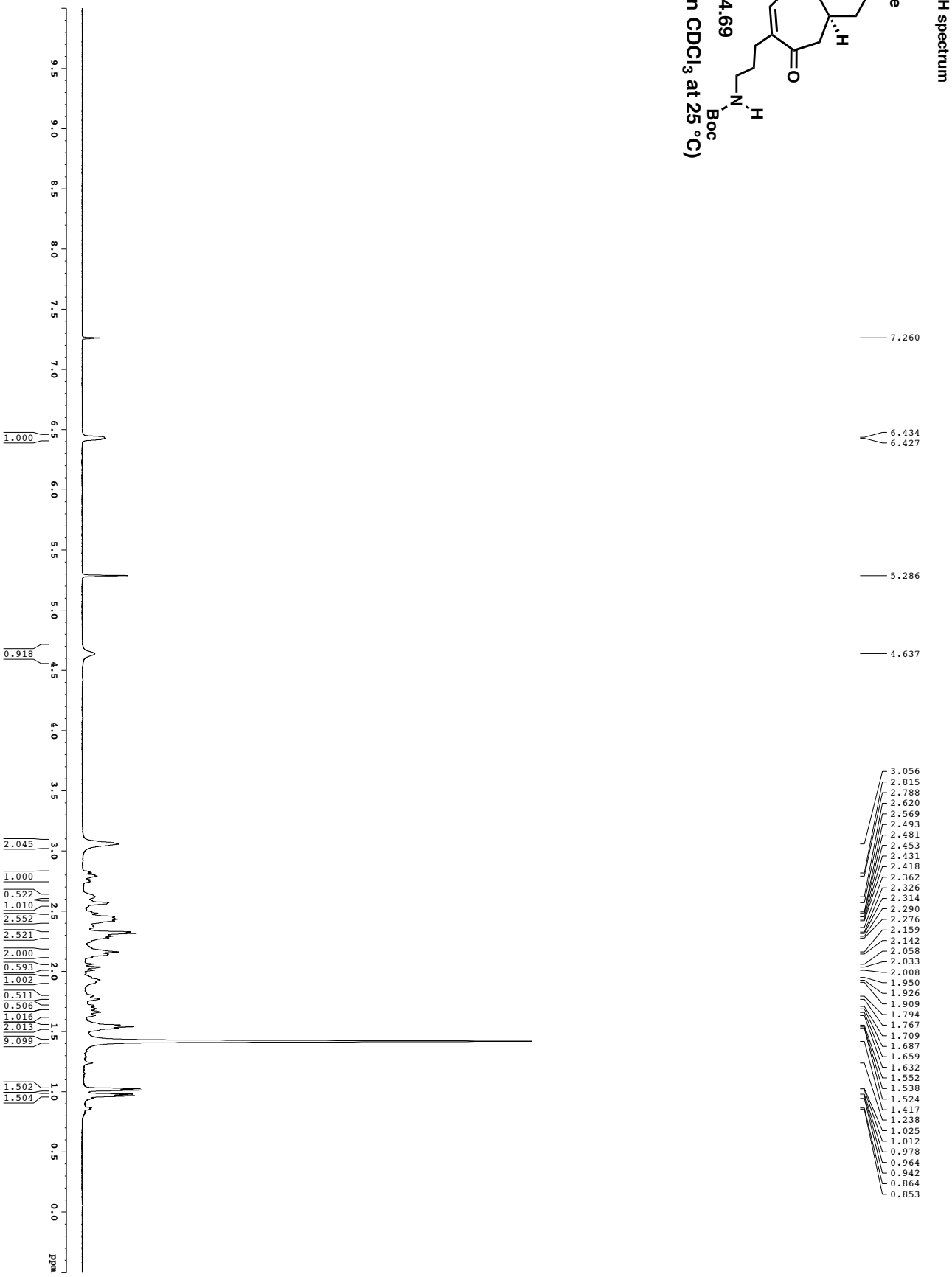


4.22

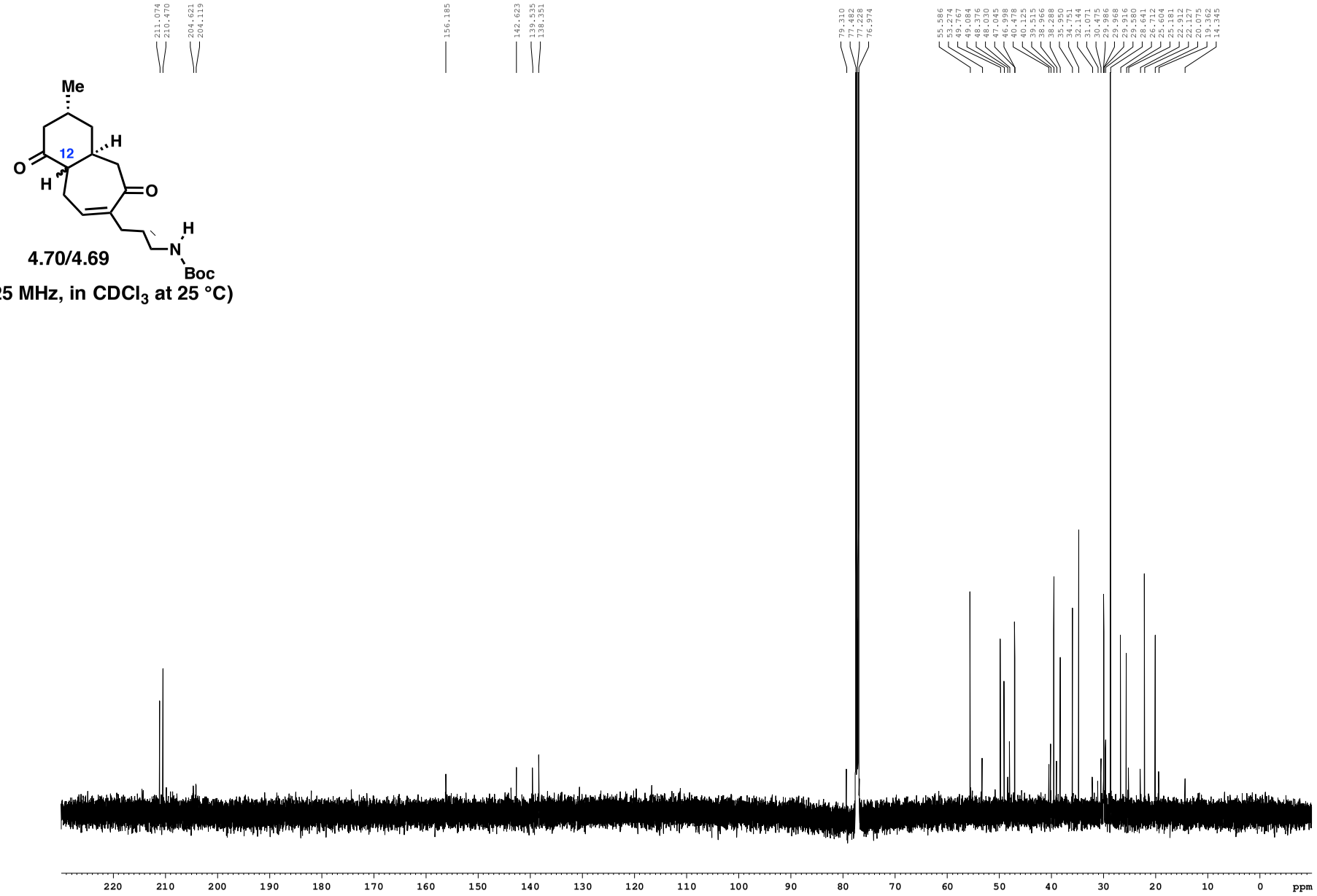
25 MHz, in $CDCl_3$ at 25 °C)

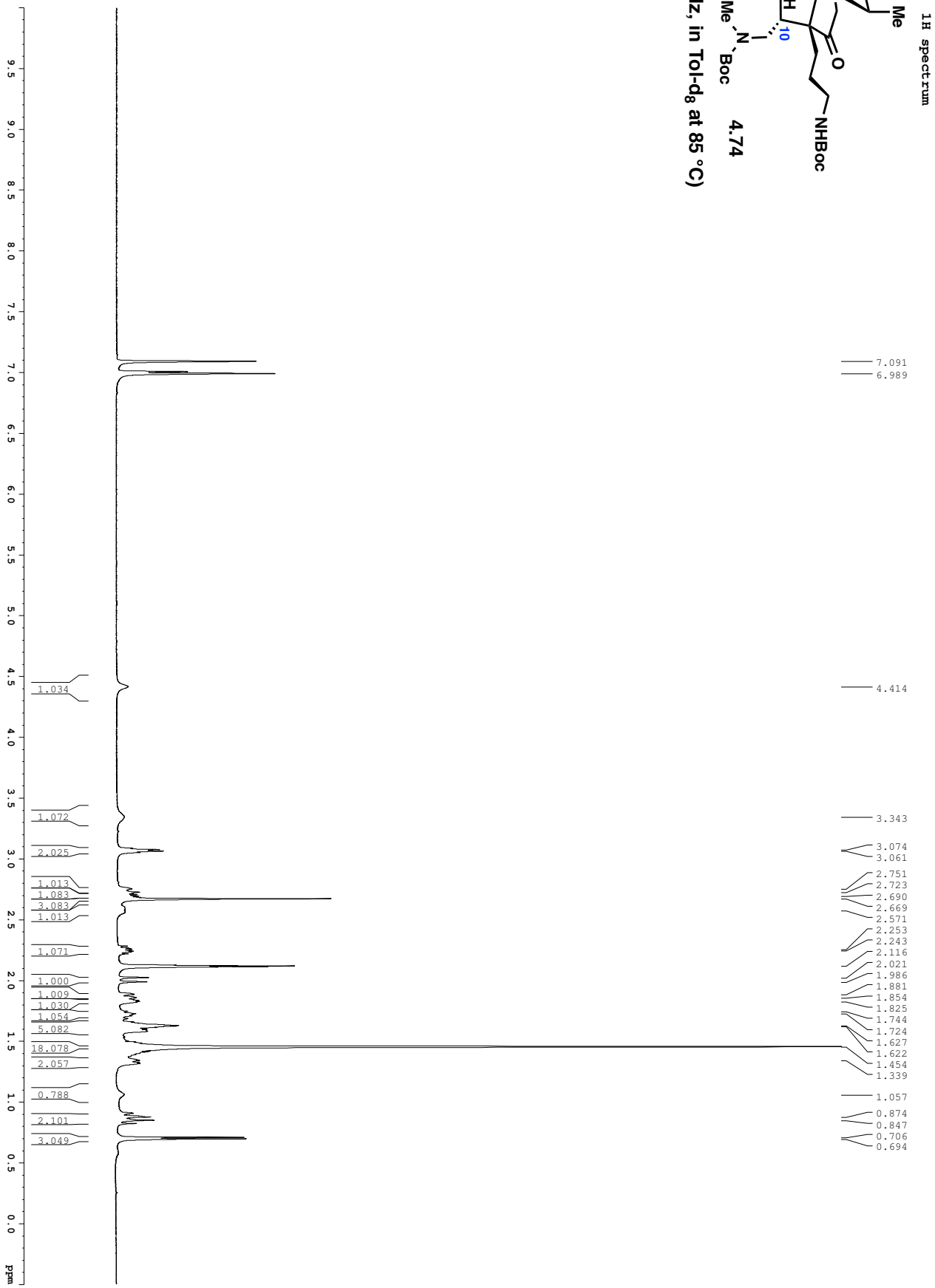
209.424
195.252
145.252
125.854
77.484
77.230
76.976
54.890
47.964
46.913
38.251
35.573
29.767
28.436
19.973



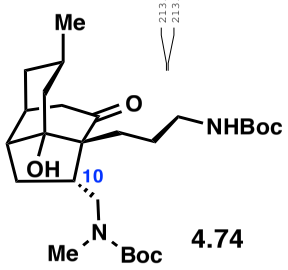

1H spectrum
Me
H
12
H
O
O
N
H
Boc
4.70/4.69
00 MHz, in CDCl3 at 25 °C)
7.260
6.434
6.427
5.286
4.637
3.056
2.815
2.788
2.620
2.569
2.493
2.481
2.453
2.431
2.418
2.362
2.326
2.314
2.290
2.276
2.159
2.142
2.058
2.033
2.008
1.950
1.926
1.909
1.794
1.767
1.709
1.687
1.659
1.632
1.552
1.538
1.524
1.417
1.238
1.025
1.012
0.978
0.964
0.942
0.864
0.853
1.000
0.918
2.045
1.000
0.522
1.010
2.552
2.521
2.000
0.593
1.002
0.511
0.506
1.016
2.013
9.099
1.502
1.504
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling



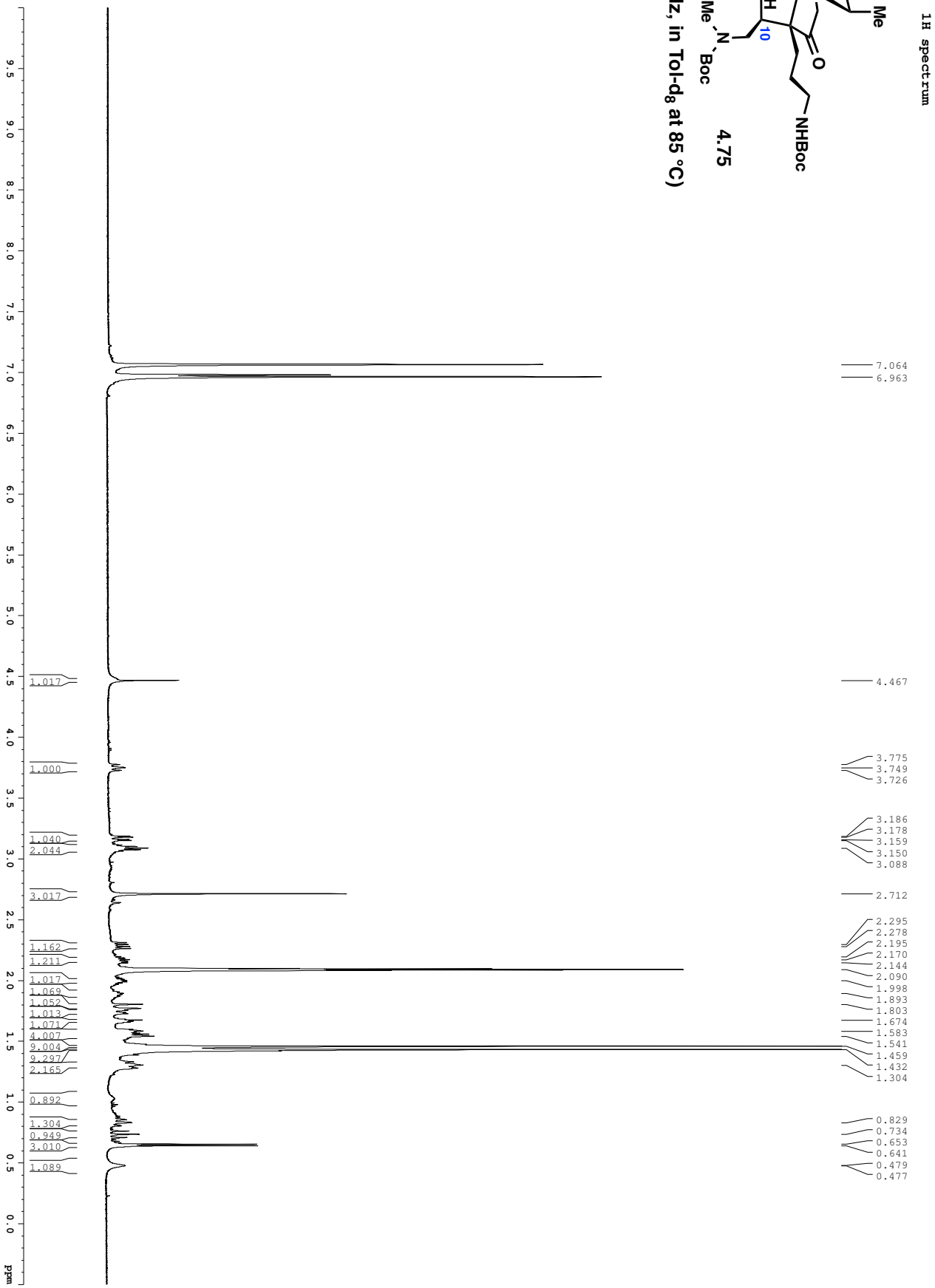

1H spectrum
Me
OH
10
O
NHBoc
Me
N
Boc
4.74
(500 MHz, in Tol-d8 at 85 °C)
7.091
6.989
4.414
3.343
3.074
3.061
2.751
2.723
2.690
2.669
2.571
2.253
2.243
2.116
2.021
1.986
1.881
1.854
1.825
1.744
1.724
1.627
1.622
1.454
1.339
1.057
0.874
0.847
0.706
0.694
1.034
1.072
2.025
1.013
1.083
3.083
1.013
1.071
1.000
1.009
1.030
1.054
5.082
18.078
2.057
0.788
2.101
3.049
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling

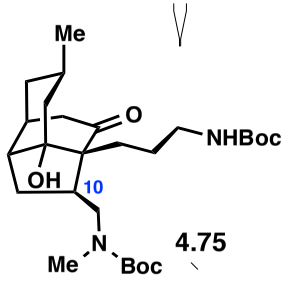


4.74

125 MHz, in $CDCl_3$ at 25 °C)


1H spectrum
Me
OH
10
O
NHBoc
Me
N
Boc
4.75
(500 MHz, in Tol-d8 at 85 °C)
7.064
6.963
4.467
3.775
3.749
3.726
3.186
3.178
3.159
3.150
3.088
2.712
2.295
2.278
2.195
2.170
2.144
2.090
1.998
1.893
1.803
1.674
1.583
1.541
1.459
1.432
1.304
0.829
0.734
0.653
0.641
0.479
0.477
1.017
1.000
1.040
2.044
3.017
1.162
1.211
1.017
1.069
1.052
1.013
1.071
4.007
9.004
9.297
2.165
0.892
1.304
0.949
3.010
1.089
ppm

Z-restored spin-echo 13C spectrum with 1H decoupling

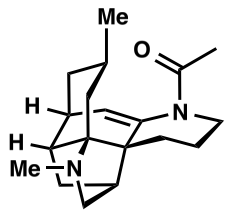


125 MHz, in $CDCl_3$ at 25 °C)

214.363
214.225
156.753
156.426
156.225
83.852
83.767
79.747
79.603
79.315
79.058
77.486
77.435
77.232
76.978
65.245
65.127
52.576
51.717
48.012
43.046
42.089
41.916
41.305
40.714
35.438
35.318
34.850
32.104
31.908
31.617
29.835
29.736
29.541
28.634
25.628
22.874
22.706
21.810
14.315

220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm

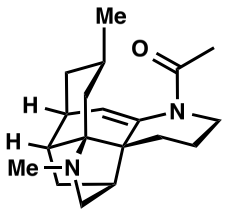
1H spectrum



(+)-fastigiatine (4.1)

500 MHz, in $CDCl_3$ at 25 °C)

Z-restored spin-echo 13C spectrum with 1H decoupling



(+)-fastigiatine (4.1)

125 MHz, in $CDCl_3$ at 25 °C)