TOTAL ASYMMETRIC SYNTHESIS OF RING-A DERIVATIVES OF (+)-*TRANS*-DIHYDRONARCICLASINE

TOTAL ASYMMETRIC SYNTHESIS OF RING-A DERIVATIVES OF (+)-*TRANS*-DIHYDRONARCICLASINE

By JON R. SCATTOLON, H. B. Sc.

A Thesis Submitted to the School of Graduate Studies in Partial Fulfilment of the Requirements for the Master of Degree Master of Science

M.Sc. Thesis (2020); McMaster University, Hamilton Ontario – Chemistry

TITLE: Total asymmetric synthesis of ring-A derivatives of (+)-*trans*-dihydronarciclasine. AUTHOR: Jon R. Scattolon, H. B. Sc. Minor Physics (Brock University). SUPERVISOR: Professor James McNulty. PAGES xvii, 181.

Lay Abstract

This thesis is primarily driven towards the development of four antiviral lycorane structural type alkaloids, and an analogue synthesized via a copper-cocatalyzed Sonogashira reaction, utilizing a labile phenol-derived sulfonated hydroxyl group in its coupling towards an alkyne tagged structure. This method provides easy access for a variety of compounds without a fluorescent tag, taking steps forward in elucidating how the Amaryllidaceae alkaloids are delivering their biological effects.

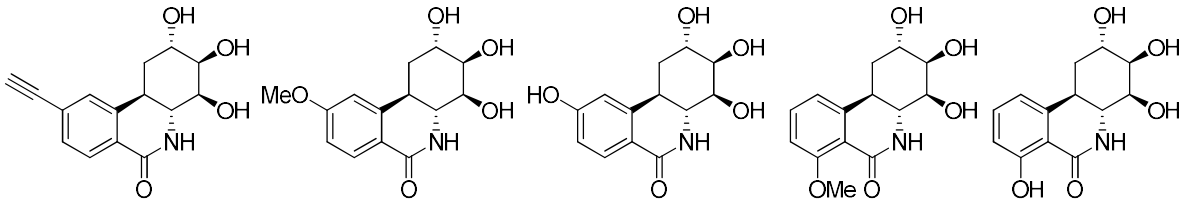
The densely substituted ring-C was obtained via an asymmetric organocatalytic [3+3] sequence for the assembly of the aminocyclitol core and is described. This sequence has provided effective regio, diastereo, and enantioselective access to five unnatural products. Preparation of the precursors were prepared using a Wittig methodology previous reported by the McNulty group that has been used in many syntheses for various Amaryllidaceae alkaloids.

Abstract

Significant attention from the medicinal and pharmaceutical communities has been pushed towards the design and development of natural products for defence against many forms of illnesses. The Amaryllidaceae plant family has shown their prevalence over time aiding towards our needs and becoming viable sources of alkaloids due to their wide variety of bioactivities presented. The low availability towards these often-complex structures with at times comprising up to six contiguous chiral centers have made practical testing scarce. More dominantly the isocarbostyrils are well recognized, being hydroxylated phenanthridones providing increased activities making them model targets to test and develop new synthetic strategies towards. These compounds represent a subset of the Amaryllidaceae alkaloids that lack a basic nitrogen center.

This thesis describes the total synthesis of four derivatives of the antiviral natural product (+)-*trans*-dihydronarciclasine from α-azidoacetone and *m*-anisaldehyde. Herein we demonstrate constructive routes towards ring-A modified, fully functionalized rings-B/C derivatives synthesized via asymmetric chemical syntheses providing further insight into SAR studies. This thesis expands on the organocatalytic [3+3]-cycloaddition sequence to produce aminocyclitol cores providing effective routes towards the development of five stereogenic centers in all targeted ring-C structures. Such studies were attributed to the enal adducts isolated from the Wittig reaction towards four natural product derivatives gaining knowledge related to the targeted molecules mode of action. One additional (+)-*trans*-dihydrolycoricidine analogue will be communicated, that enables the imaging while inside live cells with use of alkyne-tag Raman imaging.

Limitations of the alkaloids include the toxicity that accompanies these agents and the poor aqueous solubilities they provide, eliciting an increased need for new antiviral agents. The syntheses communicated provide effective routes towards unnatural alkaloids and can be pushed towards alternative chiral aminocyclitol targets for future studies. All compounds have been sent away for screening including against coronavirus at Johns Hopkins.



Acknowledgements

I would like to start by thanking Dr. Jim McNulty for his long-standing experience and growth as a chemist I have gained over the years. From volunteering all the way to completing my Master's it has been my great honor and privilege to do so by his side. His encouragement and motivation has helped me push farther along the way. Learning from such a motivated and driven leader has only kept me focused.

I would also like to thank my committee members Dr. Paul Harrison and Dr. Philip Britz-McKibbin for their guidance and feedback related to my work over the years, helping me on improving and giving me valuable words of advice every meeting. Their words of advice were always applied, preparing myself for the next steps I was to face in completing this milestone in my life.

Additionally, I will always appreciate my colleague's assistance throughout my studies, helping me develop a deeper understanding when applying new techniques and helping when asked. A more notable mention to my friend and colleague Chanti for being apart of multiple total syntheses related to natural product derivatives these past few years, it has been a growing experience and I look forward to continuing where I left off.

Grateful towards my family and girlfriend for their much-loved support, and I already can hear my mother saying "you got it from me not your father" so I'll have my smile and nod reaction ready don't you worry.

Table of Contents

List of Figures

List of Schemes

List of Tables

List of Abbreviations and Symbols

[α]	Specific rotation (expressed without units; the implicit units are deg mL g^{-1} dm^{-1})
δ	Chemical shift in ppm IC50 Median inhibitory
λ	Wavelength
ν	Frequency
τ	Retention time
Å	Ångström
Ac_2O	Acetic anhydride
MeCN/ACN	Acetonitrile
Ac	Acetyl
AcCl	Acetyl chloride
$AlCl_3$	Aluminium chloride
Ar	Aryl
ATRI	Alkyne-tag Raman imaging
atm	Atmosphere (unit of pressure)
ca. circa	Approximately
AIBN	Azobisisobutyronitrile
BBN	Banwell modified Bischler-Napieralski
BBr_3	Boron tribromide
BCl_3-DMS	Boron trichloride methyl sulfide
Bn	Benzyl
Bz	Benzoyl
NBS	N-Bromosuccinimide
Boc	*tert*-butoxycarbonyl
t-Bu	*tert*-Butyl
BuLi	Butyllithium
COSY	Correlation spectroscopy
Cy	Cyclohexyl
d	Doublet (NMR) concentration
d.r.	Diastereomeric ratio
DBF	Dibenzylfluorescein
DCM/CH_2Cl_2	Dichloromethane
DIBAL	Diisobutylaluminium hydride
DEPT	Distortionless enhancement of polarization transfer
DIPEA	*N,N*-Diisopropylethylamine (Hünig's base)
DMAP	4-Dimethylaminopyridine
DMDC	Dimethyl dicarbonate
DMF	Dimethylformamide
DMP	2,2-Dimethoxypropane
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid orbital
dppe	1,2-Bis(diphenylphosphino)ethane

dppf	1,1'-Bis(diphenylphosphino)ferrocene
het	Heterocycle
HSV-1	Herpes simplex virus type 1
HCl	Hydrochloric acid
HRMS	High-resolution mass spectrometry
EC50	Effective concentration 50%
ee	Enantiomeric excess
EI	Electron ionization
ent	Enantiomer
Eq.	Equivalent(s)
ESI	Electrospray ionization
Et	Ethyl
EtOAc	Ethyl acetate
EWG	Electron withdrawing group
FTIR	Fourier transform infrared spectroscopy
h	Hour
HPLC	High-performance liquid chromatography
Hz	Hertz
HIV	Human Immunodeficiency
HRMS	High resolution mass spectrometry
i-PrOH	Isopropyl alcohol
IR	Infrared
J	Coupling constants
KOH	Potassium hydroxide
KO*t*Bu	Potassium tert-butoxide
LDA	Lithium diisopropylamide
L	Litre
Ms	Mesylate
m-CPBA	*meta*-Chloroperoxybenzoic acid
mRNA	messenger RNA
Moc	Methoxycarbonyl
MOM	Methoxymethyl
MP	Melting point
MsCl	Methanesulfonyl chloride
MeOH	Methanol
m	Meta
m	Milli
M	Molarity
Ms	Methanesulfonate
µ	Micrometer
MW	Microwave
nM	Nanomolar
NADPH	Nicotinamide adenine dinucleotide phosphate
NOESY	Nuclear Overhauser effect spectroscopy

PEG	Polyethylene glycol
PG	Protecting group
Ph	Phenyl
Pr	Propyl
PMB	*P*-methoxybenzyl
ppm	Parts per million
PPTS	Pyridinium *p*-toluenesulfonate
Py	Pyridine
RBF	Round-bottom flask(s)
Rf	Retention factor
RNA	Ribonucleic acid
rt	Room temperature
NaHMDS	Sodium bis(trimethylsilyl)amide
NaH	Sodium hydride
SF	Sandfly fever-Sicilian
SAR	Structure activity relationship
TBAB	Tetra-*n*-butylammonium bromide
TBAF	Tetra-*n*-butylammonium fluoride
TBAI	Tetra-*n*-butylammonium iodide
TBSCl	tert-Butyldimethylsilyl chloride
TBSOTf	*tert*-Butyldimethylsilyl trifluoromethanesulfonate
TI	Therapeutic index
BuSnH	Tributyltin hydride
Tf	Triflic
Tf_2O	Trifluoromethanesulfonic anhydride
THF	Tetrahydro furan
TLC	Thin layer chromatography
TsOH	*p*-Toluenesulfonic acid
PhI=NTs	*p*-(Tosylimino)phenyliodinane
UV	Ultraviolet light
VZV	Varicella zoster virus
w/w	Weight by weight
Zn	Zinc

Declaration of Academic Achievement

The total syntheses of the four *trans*-dihydronarciclasine derivatives and one *trans*-dihydrolycoricidine analogue were completed collaboratively by myself and Chanti Dokuburra.

1.0 Introduction

Natural products are defined as chemicals produced by living organisms, many of which are used in the treatment of common and life-threatening illnesses and ailments today. Before the advancement of synthetic chemistry and antimicrobial agents' plants provided our major source of defence. Developing countries are often subject to shortages of funds, medical facilities, and newly developed medicine, so much that they become more reliant on their natural sources. This is seen among various African, Asian, and Polynesian communities that still use traditional remedies for their primary health care (Louw, Regnier, & Korsten, 2002). For hundreds of years natural products have served their purpose in the treatment towards human illnesses, playing a vital role in medicine today (Jin, 2013; Bastida, et al., 2011; Evidente, et al., 2009). Human disease over time has been commonly treated by natural products (Cragg & Snader, 1997) which was particularly evident in a study held between 1981-2002 showing that 60% of drugs related to the cancer field were of natural origin (Newman, Cragg, & Snader, 2003). It has been estimated that more than half of all cancer drugs and antibiotics originated from a natural product, including Paclitaxel (sold under the brand name Taxol, a diterpene alkaloid) that was derived from the bark of the Pacific yew tree; penicillin, one of the first antibiotics, from mold; and drugs which lower cholesterol from compounds in fungi (Demain & Vaishnav, 2011). Natural products have and still do act as an immense source of environmentally friendly alternatives in comparison to commercial products. Within this, some worthy to mention for their antineoplastic properties include vinca alkaloids (vinblastine, vincristine, vindesine, vinorelbine), taxanes (paclitaxel, docetaxel), podophyllotoxin and its derivatives

(topotecan, irinotecan), and anthracyclines (doxorubicin, daunorubicin, epirubicin, idarubicin) (Mukherjee, Basu, Sarkar, & Ghosh, 2001; Wang, Wang, Chen, & Shen, 2012; Mridul, et al., 2018). An estimated 1, 000, 000 natural products have been found, with 500, 000 - 600,000 being of plant origin (Berdy, 1995; Mendelson & Balick, 1995). With only 15% of the world's known plant resources screened thus far for their therapeutic value including analgesic, anticancer, antimutagenic, immunostimulatory, anti-infective, antimalarial, antivascular, and respiratory system effects (Fennell & van Staden, 2001), there still remains room to grow for future studies. Between 1984 – 1995 antiviral agents reported mainly in the annual reports of medicinal chemistry that 70% of synthetic agents approved by the FDA were modeled by a natural product parent, affirming that natural products remain our best resource leads for future potent yet safe antiviral agents (Cragg, Newman, & Sander, 1997).

1.0.1 References

J. Berdy. *Biotechnologia*. **1995**, 13.

J. Bastida, S. Berkov, N. B. Pigni, J. P. Andrade, V. Martínez, C. Codina, F. Viladomat, D. Muñoz-Torrero. *Recent Advances in Pharmaceutical Sciences*. **2011**, 65.

G. M. Cragg, D. J. Newman, K. M. Snader. *Journal of Natural Products*. **1997**, 52.

A. Demain, P. Vaishnav. *Microbial Biotechnology*. **2011**, 687.

A. Evidente, A. Kireev, A. Jenkins, A. Romero, W. Steelant, S. Slambrouck, A. Kornienko. *Planta Medica*. **2009**, 501.

C. W. Fennell, J. van Staden. *Journal of Ethnopharmacology*. **2001**, 15.

Z. Jin. *Natural Product Reports*. **2013**, 849.

C. A. M. Louw, T. J. C. Regnier, L. Korsten. *Journal of Ethnopharmacology.* **2002**, *82*, 147.

R. Mendelson, M. J. Balick. *Economic Botany*. **1995**, 223.

R. Mridul, L. Liang, X. Xiao, P. Feng, M. Ye, J. Liu. *Biomedicine & Pharmacotherapy*. **2018**, 682.

M. K. Mukherjee, S. Basu, N. Sarkar, A. C. Ghosh. *Current Medicinal Chemistry*. **2001**, 1467.

D. J. Newman, G. M. Cragg, K. M. Snader. *Journal of Natural Products*. **2003**, 1022.

Z. Wang, N. Wang, J. Chen, J. Shen. *Evidence-Based Complementary and Alternative Medicine*. **2012**, 650.

1.1 Amaryllidaceae Plant Family History

The monocotyledonous Amaryllidaceae family was originally found in 1805 and consists of ca. 85 genera, and 1100 species. The plant family has three subfamilies, the Agapanthoideae (agapanthus), Alliodeae (onions and chives), Amaryllidoideae (amaryllis, daffodils, and snowdrops representing the largest of the group), and is one of the 20 most important alkaloid-containing plant families (Cahlíková, et al., 2019). Active research over the last 200 years has been pushed towards the Amaryllidaceae plant family and their isoquinoline type alkaloids they produce (Acosta, Pigni, Oleas, & Bastida, 2014). Plants in

this family are seen as bulbous perennial or biennial, more commonly found in the tropics, and South Africa (Naira, Bastidab, Codinab, Viladomatb, & Stadena, 2013), while to a lesser degree in the Andean South America and the Mediterranean. Increased attention was directed towards the plant family due to their ornamental appeal of their beautiful flowers, production of a volatile oil, horticultural value, use in traditional medicinal practices, and economic exploitation. The bulbous plant family is eminent for its ornamental appeal which underpins a vibrant floriculture sector showcasing popular varieties including "daffodils" (*Narcissus pseudonarcissus*), "snowdrops" (*Galanthus nivalis*), and "snowflakes" (*Leucojum aestivum*).

Figure 1. Amaryllidaceae family plant.

Trade in the Amaryllid flower varieties is a multi-million dollar revenue generator in the floriculture industry, highlighted more by the daffodils (Nair, Bastida, Codina, Viladomat, & Staden, 2013). Many plants within the diverse family have been cultivated for their ornamental appeal, including *Narissus L.* in Europe (Torras-Claveria, Berkov, Codina, Viladomat, & Bastida, 2013), *Hippeastrum Herb.* in South America and the Indian

subcontinent (Ponnamma, 1978), *A. Cristophii* in Turkey, Iran, Turkmenistan, *A. giganteum* in central and southwestern Asia, *A. hollandium* in Iran and Kyrgyzstan, *Amaryllis belladonna L.* in Egypt alongside *N. tazetta L.* (Pettit G. , Gaddamidi, Goswami, & Cragg, 1984), while others are wild sourced ornamentals from the genus *Galanthus L.* (Ronsted, Zubov, Bruun-Lund, & A.P., 2013). Indigenous people have as well utilized different species in the plant family in traditional medicine for the cure of many ailments and diseases (Fennell & Staden, 2001; Nair & Staden, 2014). *Narcissus* oil was used in the middle ages in Chinese, North African, Central American, and Arabian medicine by individuals with cancer (Hartwell, 1967), and is seen in the bible with multiple references to the Mediterranean *Narcissus tazetta L.* (Duke & Duke, 1983; Hartwell J. , 1967). Extracts from the Amaryllidaceae family for medicinal purposes can be traced back to the times of Hippocrates and Pliny (Pettit G. , Pettit, Backhaus, Boyd, & Meerow, 1993), and near the fourth century B.C. oil of the daffodil *Narcissus poeticus L.* (Amaryllidaceae) was already known by the Greek physician Hippocrates of Cos ("Father of Medicine") for cancer treatment. It was Hippocrates of Cos (ca. B.C. 460-370) who recommended a pessary prepared from *Narcissus* oil to treat individuals suffering from uterine tumors (Kornienko & Evidente, 2008). Following his time, two physicians from ancient Greek Pedanius Dioscorides (ca. A.D. 40-90) and Soranus of Ephesus (A.D. 98-138) continued his practice during the first and second centuries A.D. towards the treatment of cancer (Pettit G. , et al., 1986). The Roman natural philosopher Gaius Plinius Secundus (A.D. 23-79), better known as Pliny the Elder applied the topical anticancer uses from extracts of the plant (Kornienko & Evidente), as well as from *Narcissus pseudonarcissus* (Tojo, 1991; Noumbissie,

Kapnang, Fomum, Martin, & Bodo, 1992; Bastida, et al., 1992) during the first century A.D.. Pliny the Elder recorded that by the first century A.D. *N. poeticus* was used in the Middle East and Roman Empire for the same reason (Pettit G. , et al., 1986). However, continuing towards the development of potent, yet safe antiviral agents, natural products still hold as our best resource for chemically diverse new leads (Sayed, 2000). With the aforementioned uses around the world, in addition to different ethnic groups in South Africa including Khoi-San, Sotho, Tswana, Xhosa, and Zulu (Fennell & Staden, Journal of Ethnopharmacology , 2001), widespread use of the Amaryllidaceae plant family is well established throughout history (Martin S. , 1987; Fox & Tram, 1991; Furusawa, Irie, Combs, & Wildman, 1980; Baez & Vasquez, 1978; Jimenez, Santos, Alonso, & Vasquez, 1976) and by its nature serving as a prevailing element towards the fight against a multitude number of illnesses. The medicinal value of the Amaryllidaceae plant extracts are well recognized for their high therapeutic value. The fact that we share no efficient means of supplying these compounds keeps researchers pursuing new methods to solve the problems we face each day. The use of such remedies by the ethnic groups described falls on the fact of traditional medicines forming an integral part of southern African culture, and how such cures can be of greater significance in developing countries (Elgorashi & Staden, 2009). Amongst the illnesses and various ailments are skin disease, headaches, wounds, renal and liver complaints, aching joints, stomach ailments, rheumatism, dizziness, chest and bladder pain, snake bites, facilitation of child birth during labor, hysteria, and as narcotics (Hutchings, 1989). The rational supporting the plant family's use is reasoned through the

unique type of metabolites that they produce which has led to their medicinal efficacy (figure 2).

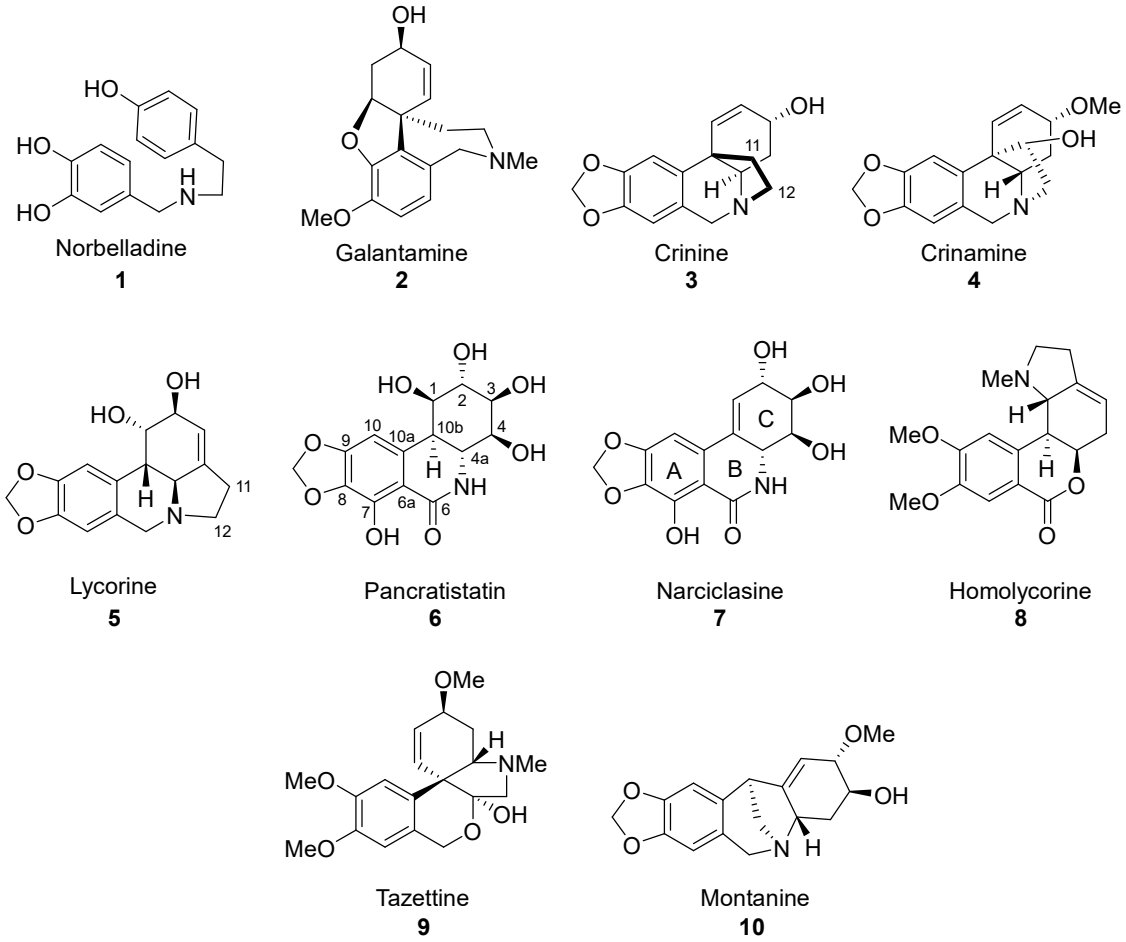


Figure 2. Structurally diverse alkaloids of the Amaryllidaceae plant family.

The alkaloids produced are exclusively synthesized by the Amaryllidaceae family, with notable exceptions by the genus *Hosta*, in the order Asparagales which has been shown to produce a collection of similar alkaloids (Chase, Reveal, & Fay, 2009). The alkaloids produced by the Amaryllidaceae family (Bastida J. , et al., 2011; Evidente A. , et al., 2009; Martin S. , 1987; Jin, 2013; Wildman, 1968; Cook & Loudon, 1952) are constantly under

study for their antiviral (He, et al., 2012), antibacterial (Nair, Staden, Bonnet, & Wilhelm, 2017; Nair, Wilhelm, Bonnet, & Staden, 2017), bacteriostatic (Ceriotti, 1967), antitumor (Ding, et al., 2016), antifungal (Nair & Staden, 2017), antimalarial (Uzor, 2020), antimitotic (Ceriotti, Nature, 1967), antiplasmodial (Cedron, Gutierrez, Flores, Ravelo, & Braun, 2010), antitrypanosomal (Toriizuka, et al., 2008), anti-inflammatory (Fürst R. , 2016), antiparasitic (Ouarzane-Amara, et al., 2001), antidiabetic (Mukhopadhyay, Pai, Babu, & Lobo, 2019), analgesic, emetric (Kretzing, et al., 2011), cytotoxic, cholinesterase inhibition (Dalecká, Havelek, Královec, Brucková, & Cahlíková, 2013; He, Qu, Gao, Hu, & Hong, 2015; Jin, 2016), insecticidal properties (Desgagné-Penix, 2020), as well as activity on the central nervous system (CNS) related to hallucinogenic effects, mental disorders, and age-related dementia (Lopez, Bastida, Viladomat, & Codina, 2002; Ding, et al., 2016; Tram, Titorenkova, Bankova, Handjieva, & Popov, 2000). The most well recognized and human used of the Amaryllidaceae plant family for the treatment of human disease is galantamine (**2**) a tertiary alkaloid from the bulbs of the Caucasian snowdrops, *Galanthus woronowii* (F. & Yakovleva, 1952) and several other Amaryllidaceae plants (Sramek, Frackiewicz, & Cutler, 2000). Alkaloids can be distinguished as a class of nitrogenous organic compounds, but frequently contain a nitrogen-containing base, and the Amaryllidaceae plant family is very prominent in this respect. Galantamine hydrobromide is a drug which has been developed and approved in many countries for the treatment of mild-to-moderate Alzeimer's disease (AD). High value is attributed to such a treatment because AD represents the most common form of dementia, and in 1998 an estimated 15 million people suffered with the condition (Bosanquet, 1998). Its impressive tolerability profile, while

showing low potential for clinically significant drug-drug interactions makes it a significant addition to the armamentarium of drugs available for human treatments. It behaves as an acetylcholinesterase inhibitor (AChEI) with the ability to amplify the nerve-muscle transfer (Zakirov & Umarova, 1971), and is produced commercially as a treatment from cultivated plants (Marco-Contelles, Carreiras, Rodriguez, Villarroya, & Garcia, 2006). Its bioavailability has been exploited therapeutically to raise the depressed levels of acetylcholine located in the brain, giving patients with AD relief while remaining innocuous through the inhibition of acetylcholinesterase (Houghton, Agbedahunsi, & Adegbulugbe, 2004). Its first asymmetric synthesis towards the enantiomerically pure (+)- and (-)-galantamine (**2**) had been reported from Koga et. al. (Tomioka, et al., 1878; Shimizu, Tomioka, Yamada, & Koga, 1977; Tomioka, Shimizu, Yamada, & Koga, 1977).

To this day new alkaloids are being extracted out of the Amaryllidaceae family of plants, including jonquailine (Masi, et al., 2015), and crinsarnine (Masi, et al., 2016). The Amaryllidaceae family of plants have long ago established themselves as an extensive source of pharmacologically active alkaloids (Karakoyun, et al., 2019) that are structurally related due to their biogenesis from a common amino acid derived precursor norbelladine **1** (see Chapter 1.2). It was not until 1971 when the biosynthesis of the Amaryllidaceae alkaloids were first presented (Fuganti, Staunton, & Battersby, 1971). Lycorine (**5**) (pyrrolo[de]phenanthridine subgroup) in 1877 was the first alkaloid isolated from the plant *Narcissus pseudonarcissus* and has since been shown to be the most abundant alkaloid in the family possessing antitumor activity (Hulcova, et al., 2018; Cedron, Gutierrez, Flores, Ravelo, & Estevez-Braun, 2010). The Amaryllidaceae plant family has had more than 600

structurally diverse alkaloids extracted from its members, pulling attention from scientists over the years due to their challenging isolation in aim of completing a synthesis feasible to scale for clinical trials. Increased attention from researchers in this field towards the lycorane isocarbostyrils including (+)-*trans*-dihydrolycoricidine (**13**), narciclasine (**7**), and pancratistatin (**6**) has been drawn because of their biological activities observed (Banwell, Gao, Schwartz, & White, 2012; Rinner & Hudlicky, 2005; Chapleur, Chretien, Ibn-Ahmed, & Khaldi, 2006; Manpadi & Kornienko, 2008; Danishefsky & Lee, 1989; Tian, Hudlicky, & Königsberger, 1995; Trost & Pulley, 1995; Keck, McHardy, & Murry, 1995; Hudlicky T. , et al., 1996; Doyle, Hendrix, Van-Derveer, Javanmard, & Haseltine, 1997) (Magnus & Sebhat, 1998; Magnus & Sebhat, Tetrahedron, 1998; Magnus & Sebhat, 1998; Rigby, Maharoof, & Mateo, 2000; Ko, Kim, Park, Kim, & Kim, 2004). These selected alkaloids have been reviewed and studied to show potent cytotoxicity in the NCI 60 human tumor cell line panel having GI50 values in the nanomolar range (Pettit & Melody, 2005; Pettit G. R., Pettit III, Backhaus, & Meerow, 1993; Paull, et al., 1989). A standout feature of natural products lies in their ability to effect other organisms, take self-defence for example. Not all organisms have the ability of locomotion or movement through their life spam, while at times are also the prey in the environment they live. Through the production of specific natural products, they have developed mechanisms to defend themselves. Such mechanisms led to the production of information rich molecules for researchers to learn and further study in aim of increasing their efficacy. The Amaryllidaceae family has long been notorious for their medicinal value, and admitted to other toxic properties as well (Louw, Regnier, & Korsten, 2002; Fennell & van Staden, 2001; Foukaridis, Osuch,

Mathibe, & Tsipa, 1995; Gaillard & Pepin, 1999). Toxicological profiles have communicated adverse effects from a number of Amaryllidaceae alkaloids causing a transient fall in blood pressure from animal studies when administered at elevated levels including galantamine, *epi*-galantamine, and narwedine all of which have been shown to cause significant hypotensive effects in mice (Bezhenova, Aliev, & Zairov, 1972). While in ancient Greece and Rome the oil from the *Narcissus* species were being used for the treatment of cancer, ongoing research today is uncovering new leads to help save lies every year (Hartwell J. , 1967). The most recognized natural products from the Amaryllidaceae family are narciclasine (**7**)**,** lycoricidine (**11**), and pancratistatin (**6**), more commonly known as the isocarbostyrils (Figure 3) (Pettit G. R., Pettit, Backhaus, Boyd, & Meerow, 1993).

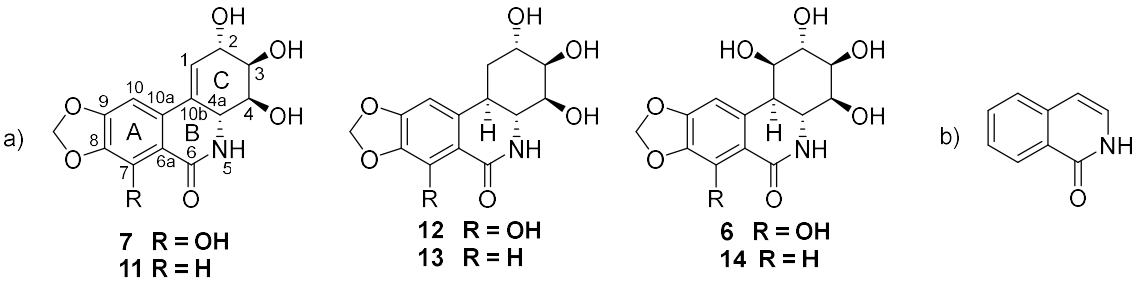


Figure 3. a) Three major isocarbostyrils congeners of the Amaryllidaceae family, and their respective 7-deoxy analogues; b) Isocarbostyril moiety.

These important metabolites and their congeners contribute most towards the plant family's therapeutic benefits related to folk medicine treatment. Among the 860 - 1100 species of plants belonging to the Amaryllidaceae family, 30 or more species have shown use in ancient folk medicine for cancer treatment (Martin & Brossi, 1987). Notably, *N. poeticus L.* used by the ancient Greek physicians mentioned earlier, is now known to contain 0.12g of narciclasine per kilogram of fresh bulbs (Piozzi, Marino, Fuganti, & Di Martino, 1969;

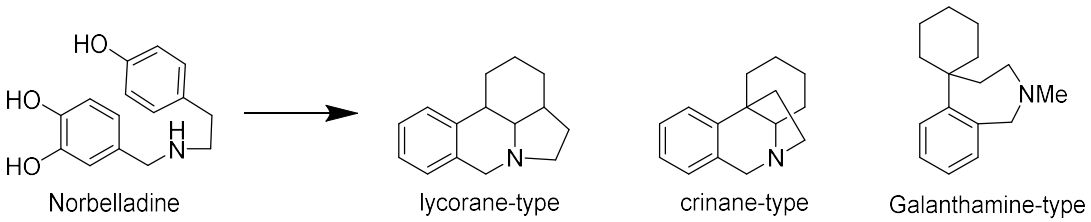
Kornienko & Evidente, 2008). Plants within the Amaryllidaceae genus are responsible for the production of alkaloids, and other non-basic molecules. With the isolation process not providing sufficient enough material for clinical studies, coupled with the remarkable biological activities presented, the chemistry community remains focused on coming forward with new syntheses of the natural products to one day be granted the medicinal benefits from the Amaryllidaceae family. The total synthesis of challenging molecules such as narciclasine (**7**), 7-deoxynarciclasine (also known as lycoricidine **11**), 7-deoxy-*trans*-dihydronarciclasine (also known as *trans*-dihydrolycoricidine **13**), and pancratistatin (**6**), have been performed (table 1) and continue to stimulate interest. Providing sufficient quantities of the natural products themselves, or other derivatives/analogues is a goal researchers strive for.

Table 1. Selected total syntheses of Amaryllidaceae isocarbostyrils (Kornienko & Evident, 2008).

Entry	Isocarbostyril	Research group	Publication year	# of steps	Total yield
1	(+)-narciclasine	Rigby	1997	23	0.2%
2	(+)-narciclasine	Hudlicky	1999	9	0.6%
3	(+)-narciclasine	Keck	1999	14	16%
4	(+)-narciclasine	Yan	2002	13	17%
5	(+)-pancratistatin	Danishefsky	1989	27	0.16%
6	(+)-pancratistatin	Trost	1995	19	8%
7	(+)-7-deoxynarciclasine	Ogawa	1991	23	0.04%
8	(+)-7-deoxynarciclasine	Keck	1999	11	27%
9	(+)-7-deoxynarciclasine	Yan	2002	15	11%
10	(+)-7-deoxypancratistatin	Paulsen	1982	9	6.5%
11	(+)-7-deoxypancratistatin	Keck	1998	15	12%
12	(+)-7-deoxypancratistatin	Madsen	2006	15	4.3%
13	(+)-*trans*-dihydronarciclasine	Cho	2007	15	11%
14	(+)-*trans*-dihydronarciclasine	Studer	2008	17	5.6%
15	(+)-*trans*-dihydronarciclasine	McNulty	2016	9	4.7%
16	(+)-7-deoxy-*trans*-dihydronarciclasine	Iwabuchi	2005	24	1.4%

***One-pot procedures counted as one step. If the precursors were prepared and not commercially available, the number of steps and yields were taken from the cited references.

With respect to the alkaloids' structures, they are related as a consequence of their biogenesis from norbelladine but are later divided into subgroups based on their ring structures. Three classic structural types that are more recognized include the lycorane-type (isocarbostyril *trans*-dihydrolycoricidine **13**), galantamine-type (galantamine **2**), and the crinane-type (crinamine **4**) (McKillop, 1969) but many other complex modifications are present (Jin, 2005).



Scheme 1. Biosynthesis origin of the Amaryllidaceae alkaloids.

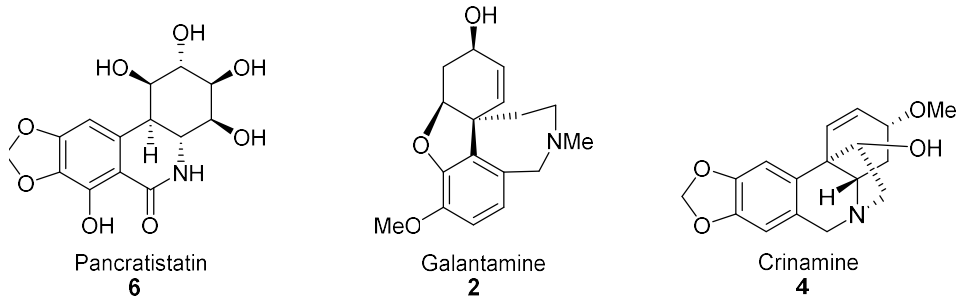


Figure 4. Selected biologically active Amaryllidaceae constituents.

The reasons behind their synthetic pursuit falls on the importance they each share towards medicine, agriculture, along with other commercial and humanitarian ventures. Their limited isolation quantities, along with the inherent expense in gathering the natural products from their source has prevented their further development. If these molecules wish to continue to go through more screenings, or to be used clinically, synthesis may be our best option in obtaining enough material. Viral disease is a rising concern, especially when herpes simplex virus (HSV) is the topic of discussion which has been shown to be a problem for society and is heightened in terms of sexually transmitted diseases. Solutions to this problem can be addressed through the study of terrestrial and marine organisms that have previously functioned as great sources for many chemotherapeutic agent sources (Pettit, Herald, & Smith, 1989; Martin S. F., 1987; Fox & R., 1991). DNA and RNA viral

pathogens (McNulty, et al., 2016) have previously been combatted against using lycorine type alkaloids with their effective antiviral activities. Activity of this branch of structurally related alkaloids against flaviviruses including Japanese encephalitis (JE), yellow fever and Dengue is of great importance after the recent outbreaks of the genetically related Zika flavivirus (ZIKV) (Gabrielsen B. , et al., 1992; Revu, et al., 2016). Other examples include 7-deoxy-*trans*-dihydronarciclasine (also known as *trans*-dihydrolycoricidine **13**) found in bulbs of *Hymenocallis littoralis*, *Hymenocallis caribaea*, and *Hymenocallis latifolia* which inhibited the cytopathicity and/or replication of various viruses, in addition to lycorine (**5**) being responsible for exerting antiviral activity extracted from the Amaryllidaceae plant leaves and roots of Clivia miniata Regel against Herpes simplex, Semliki forest, polio, Coxsackie, and measles viruses in Vero cells (leven, Vlietinck, Vanden, & Berghe, 1982). These biosynthetic products assert such effects mainly through the inhibition of protein synthesis when the peptide bond is forming. This can be seen with lycorine's effects on viral protein formation in poliovirus infected HeLa cells (Vrijsen, Vanden Berghe, Vlietinck, & Boeye, 1986). Other alkaloids following this type of behavior include pseudolycorine (**16**) and pretazettine (**17**) that show activity against murine Rauscher leukemia virus and neurotropic RNA viruses (Robert, Zee-Cheng, & Cheng, 1978).

1.1.1 References

K. Acosta, N. Pigni, N. Oleas, J. Bastida. *Pharmacologyonline*. **2014**, 178.

M. G. Banwell, N. Y. Gao, B. D. Schwartz, L. V. White. *Topics in Current Chemistry*.

2012, 163.

J. Bastida, S. Berkov, L. Torras, N. B. Pigni, J. P. Andrade, V. Martinez, C. Codina, F. Viladomat. *Transworld Research Network*. **2011**, 65.

J. Bastida, C. Codina, F. Viladomat, M. Rubiralta, J. C. Quirion. B. Weniger. *Journal of Natural Products*. **1992**, 122.

E. D. Bezhenova, K. V. Aliev, V. B. Zairov. *Farmakol*. **1972**, 100.

N. Bosanquet. *Health Policy Unit, Imperial School of Medicine*. **1998**, 98.

L. Cahlíková, N. Vaněčková, M. Šafratová, K. Breiterová, G. Blunden, D. Hulcová, L. Opletal. *Molecules*. **2019**, 4238.

J. C. Cedron, D. Gutierrez, N. Flores, G. Ravelo, A. Braun. *Bioorganic & Medicinal Chemistry*. **2010**, 4694.

G, Ceriotti. *Nature*. **1967**, 595.

Y. Chapleur, F. Chretien, S. Ibn-Ahmed, M. Khaldi. *Current Organic Synthesis*. **2006**, 169.

M. W. Chase, J. L. Reveal, M. F. Fay. *Botanical Journal of the Linnean Society*. **2009**, 132.

J. W. Cook, J. D. Loudon. *Academic Press, New York*. **1952**, 331.

S. Danishefsky, J. Y. Lee. *Journal of the American Chemical Society*. **1989**, 111, 4829.

I. Desgagné-Penix. *Phytochemistry Review*. **2020**, 9678.

Y. Ding, D. Qu, K. M. Zhang, X. X. Cang, Z. N. Kou, W. Xiao, J. B. Zhu. *Journal of Asian Natural Products Research*. **2016**, 53.

T. J. Doyle, M. Hendrix, D. Van-Derveer, S. Javanmard, J. Haseltine. *Tetrahedron*. **1997**, 11153.

J. A. Duke, P. K. Duke. *Medicinal Plants of the Bible*. **1983**, 98.

E. E. Elgorashi, J. Staden. *Journal of the American Chemical Society*. **2009**, 151.

A. Evidente, A. S. Kireev, A. R. Jenkins, A. E. Romero, W. F. A. Steelant, A. Slambrouck. *Planta Medica*. **2009**, 501.

C. W. Fennell, J. J. van Staden. *Journal of Ethnopharmacology*. **2001**, 15.

G. N. Foukaridis, G. E. Osuch, L. Mathibe, P. Tsipa. *Journal of Ethnopharmacology*. **1995**, 77.

B. W. Fox, R. Tram. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. **1991**, 22.

R. Fürst. *Planta Medica*. **2016**, 1389.

E. Furusawa, H. Irie, H. Combs. W. Wildman. *Chemotherapy*. **1980**, 36.

B. Gabrielsen, T. P. Monath, J. W. Huggins, D. F. Kefauver, G. R. Pettit, G. Groszek, M. Hollingshead, J. J. Kirsi, M. W. Shannon, E. M. Schubert, J. DaRe, B. Ugarkar, M. Ussery, M. J. Phelan, J. Dare. *Natural Products*. **1992**, 1369.

Y. Gaillard, G. Pepin. *Journal of Chromatography*. **1999**, 181.

J. L. Hartwell. *Lloydia*. **1967**, 379.

J. He, W. Qi, L. Wang, J. Tian, P. Jiao, G. Liu, W. Ye, M. Liao. *Influenza and Other Respiratory Viruses*. **2012**, 922.

M. He, C. Qu, O. Gao, X. Hu, X. Hong. *Royal Society of Chemistry Advances*. **2015**, 16562.

P. J. Houghton, J. M. Agbedahunsi, A. Adegbulugbe. *Phytochemistry*. **2004**, *65*, 2893.

T. Hudlicky, X. R. Tian, K. Königsberger, R. Maurya, J. Rouden, B. Fan. *Journal of the American Chemical Society*. **1996**, 10754.

D. Hulcova, K. Breiterova, T. Siatka; K. Klimova; L. Davani, M. Safratova, A.

Hostalkova, A. De Simone, V. Andrisano, L. Cahlikova. *Molecules*. **2018**, 719.

A. Hutchings. *Bothalia, African Biodiversity & Conservation*. **1989**, 112.

Z. Jin. *Natural Product Reports*. **2005**, 111.

Z. Jin. *Natural Products*. **2013**, 839.

Z. Jin. *Natural Product Reports*. **2016**, 1318.

C. Karakoyun, M. Masi, A. Cimmino, M. Önür, N. Somer, A. Kornienko, A. Evidente. *Natural Product Communitions*. **2019**, 1.

A. Kornienko. *Planta Medica*. **2009**, 501.

S. Kretzing, G. Abrahama, B. Seiwert, F. R. Ungemach, U. Krügel, R. Regenthal. *Toxicon*. **2001**, 117.

J. L. Hartwell. *Lloydia*. **1967**, 379.

A. Jimenez, A. Santos, D. Alonso, D. Vasquez. *Biochimica et Biophysica Acta*. **1976**, 342.

A. Kornienko, A. Evidente. *Chemical Reviews*. **2008**, 1982.

G. E. Keck, S. F. McHardy, J. A. Murry. *Journal of the American Chemical Society*. **1995**, 7289.

H. Ko, E. Kim, J. E. Park, D. Kim, S. Kim. *Journal of the American Chemical Society*. **2004**, 112.

A. Kornienko, A. Evidente. *Chemical Reviews*. **2008**, 1982.

I. Leven, A. J. Vlietinck, D. A. Vanden, J. Berghe, R. Totte, E. Dommisse, F. Alderweireldt. *Natural Products*. **1982**, 564.

S. Lopez, J. Bastida, F. Viladomat, C. Codina. *Life Sciences*. **2002**, 2521.

C. A. M. Louw, T. J. C. Regnier, L. Korsten. *Journal of Ethnopharmacology*. **2002**, 147.

P. Magnus, I. K. Sebhat. *Journal of the American Chemical Society*. **1998**, 5341.

P. Magnus, I. K. Sebhat. *Tetrahedron*. **1998**, 15509.

M. Manpadi, A. Kornienko. *Organic Preparations and Procedures International*. **2008**, 107.

S. F. Martin. *Academic Press, New York*. **1987**, 251.

S. F. Martin, A. R. Brossi. *The Alkaloids*. **1987**, 352.

S. F. Martin. *Academic Press, New York*. **2002**, 287.

M. Masi, A. Cala, N. Tabanca, A. Cimmino, I. R. Green, J. R. Bloomquist, W. Otterlo, F. F. A. Macias, A. Evidente. *Molecules*. **2016**, 1432.

M. Masi, L. V. Frolova, X. Yu, V. Mathieu, A. Cimmino, A. D. Carvalho, R. Kiss, S. Rogelj, A. Pertsemlidis, A. Kornienko, A. Evidente. *Fitoterapia*. **2015**, 41.

A. McKillop. *Butterworths, London*. **1969**, 149.

J. McNulty, L. D'Aiuto, Y. Zhi, L. McClain, C. Zepeda-Velezquez, S. Ler, H. A. Jenkins, M. B. Yee, P. Piazza, R. H. Yolken, P. R. Kinchington, V. L. Nimgaonkar. *ACS Medicinal Chemistry Letters*. **2016**, 46.

N. Mukhopadhyay, S. Pai , U. Babu, R. Lobo. *International Research Journal of Pharmacy*. **2019**, 31.

J. Nair, J. Staden. *Natural Product Communications*. **2017**, 1953.

J. Nair, J. Staden, S. Bonnet, A. Wilhelm. *Natural Product Communications*. **2016**, 635.

J. Nair, A. Wilhelm, S. L. Bonnet, J. Staden. *Bioorganic & Medicinal Chemistry Letters*. **2017**, 4943.

J. Naira, J. Bastidab, C. Codinab, F. Viladomatb, J. Stadena. *Natural Product*

Communications. **2013**, 1335.

B. E. Noumbissie, H. Kapnang, Z. T. Fomum, M. T. Martin, B. Bodo. *Journal of Natural Products*. **1992**, 137.

M. Ouarzane-Amara, J. F. Franetich, D. Mazier, G. R. Pettit, L. Meijer, C. Doerig, I. Desportes-Livage. *Antimicrobial Agents and Chemotherapy*. **2001**, 3409.

K. D. Paull, R. H. Shoemaker, L. Hodes, A. Monks, D. A. Scudiero, L. Rubinstein, J. Plowman, M. R. Boyd. *Journal of the National Cancer Institute*. **1989**, *81*, 1088.

G. R. Pettit, V. Gaddamidi, A. Goswami, G. M. Cragg. *Journal of Natural Products*. **1984**, 796.

G. R. Pettit, V. Gaddamidi, D. L. Herald, S. B. Singh, G. M. Cragg, J. Schmidt, F. E. Boettner, M. Williams, Y. Sagawa. *Natural products*. **1986**, 995.

G. R. Pettit, C. L. Herald, C. R. Smith. *Elsevier*. **1989**.

G. R. Pettit, N. Melody. *Journal of Natural Products*. **2005**, *68*, 207.

G. R. Pettit, G. R. Pettit, III, R. A. Backhaus, M. R. Boyd, A. W. Meerow. *Journal of Natural products*. **1993**, 1682.

F. Piozzi, M. L. Marino, C. Fuganti, A. Di Martino. *Phytochemistry*. **1969**, 1749.

N. F. Proskurnina, A. P. Yakovleva. *Russian Journal of General Chemistry*. **1952**, *22*, 1899.

O. Revu, C. Zepeda-Velázquez, A. J. Nielsen, J. McNulty, R. H. Yolken, L. Jones-Brando. *ChemistrySelect*. **2016**, 5895.

J. H. Rigby, U. S. M. Maharoof, M. E. Mateo. *Journal of the American Chemical Society*. **2000**, 6624.

U. Rinner, T. Hudlicky. *Synlett*. **2005**, 365.

K. Y. Robert, S. J. Zee-Cheng, C. C. Cheng. *Journal of Medicinal Chemistry*. **1978**, 199.

N. Ronsted, D. Zubov, S. Bruun-Lund, A.P. Davis. *Molecular Phylogenetics and Evolution*. **2013**, 205.

K. Shimizu, K. Tomioka, S. I. Yamada, K. Koga. *Heterocycles*. **1977**, *8*, 277.

K. Shimizu, K. Tomioka, S. I. Yamada, K. Koga. *Chem. Pharm. Bull.* **1978**, *26*, 3765.

J. J. Sramek E. J. Frackiewicz, N. R. Cutler. *Expert Opinion on Investigational Drugs*. **2000**, *9*, 2393.

X. R. Tian, T. Hudlicky, K. Königsberger. *Journal of the American Chemical Society*. **1995**, 3643.

E. J. Tojo. *Journal of Natural Products*. **1991**, 1387.

K. Tomioka, K. Shimizu, S. I. Yamada, K. Koga, K. *Heterocycles*. **1977**, *6*, 1752.

Y. Toriizuka, E. Kinoshita, N. Kogure, M. Kitajima, A. Ishiyama, K. Otoguro, H. Yamada, S. Omura, H. Takayama. *Bioorganic & Medicinal Chemistry*. **2008**, 10182.

L. Torras-Claveria, S. Berkov, C. Codina, F. Viladomat, J. Bastida. *Industrial Crops and Products*. **2013**, 237.

N. T. N. Tram, T. V. Titorenkova, V. S. Bankova, N. V. Handjieva, S. S. Popov. *Fitoterapia*. **2000**, 183.

B. M. Trost, S. R. Pulley. *Journal of the American Chemical Society*. **1995**, 10143.

P. Uzor. *Evidence-Based Complementary and Alternative Medicine*. **2020**, 1.

R. Vrijsen, D. Vanden Berghe, A. Vlietinck, A. Boeye. *Biological Chemistry*. **1986**, 505.

U. B. Zakirov, S. S. Umarova. *Farmakol. Alkaloid Ser. Glik.* **1971**, 96.

1.2 Discovery and Biosynthesis

Exploration in 1877 marked the introduction of the first alkaloid from the Amaryllidaceae family, isolated from *N. pseudonarcicuss* and named lycorine (**5**). Since then over 600 structurally diverse alkaloids have been extracted from its members. From these findings, investigations into the biosynthesis of the natural products belonging to variable ring types have been performed (Bernfeld, 1963). But it was not until the 20th century when natural products exhibiting the highest level of anticancer activity were isolated and characterized. The congeners were found to be all highly oxygenated compounds sharing the isocarbostyril moiety (see figure 3). Narciclasine (**7**) being the first of this class isolated in 1968 (Piozzi, Fuganti, Mondelli, & Ceriotti, 1968), and soon after lycoricidine (**11**) (Okamoto, Torii, & Isogai, 1968). Almost two decades later pancratistatin (**6**) was isolated in 1968 by Pettit et. al. (Pettit G. , Gaddamidi, Cragg, Herald, & Sagawa, 1984), 7-deoxypancratistatin (**14**) in 1989 by Ghosal (Ghosal, Singh, Kumar, & Srivastava, 1989), and the following year *trans*-dihydronarciclasine (**12**) was isolated by Pettit et. al. (Pettit, Cragg, Singh, Duke, & Doubek, 1990). The more recent isolation of 7-deoxy-*trans*-dihydronarciclasine (**13**) was accomplished in 1993 by the same group (Pettit, Backhaus, Boyd, & Meerow, 1993).

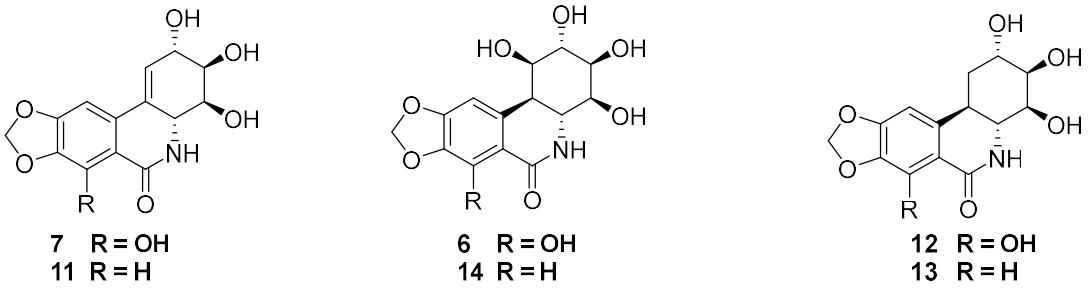
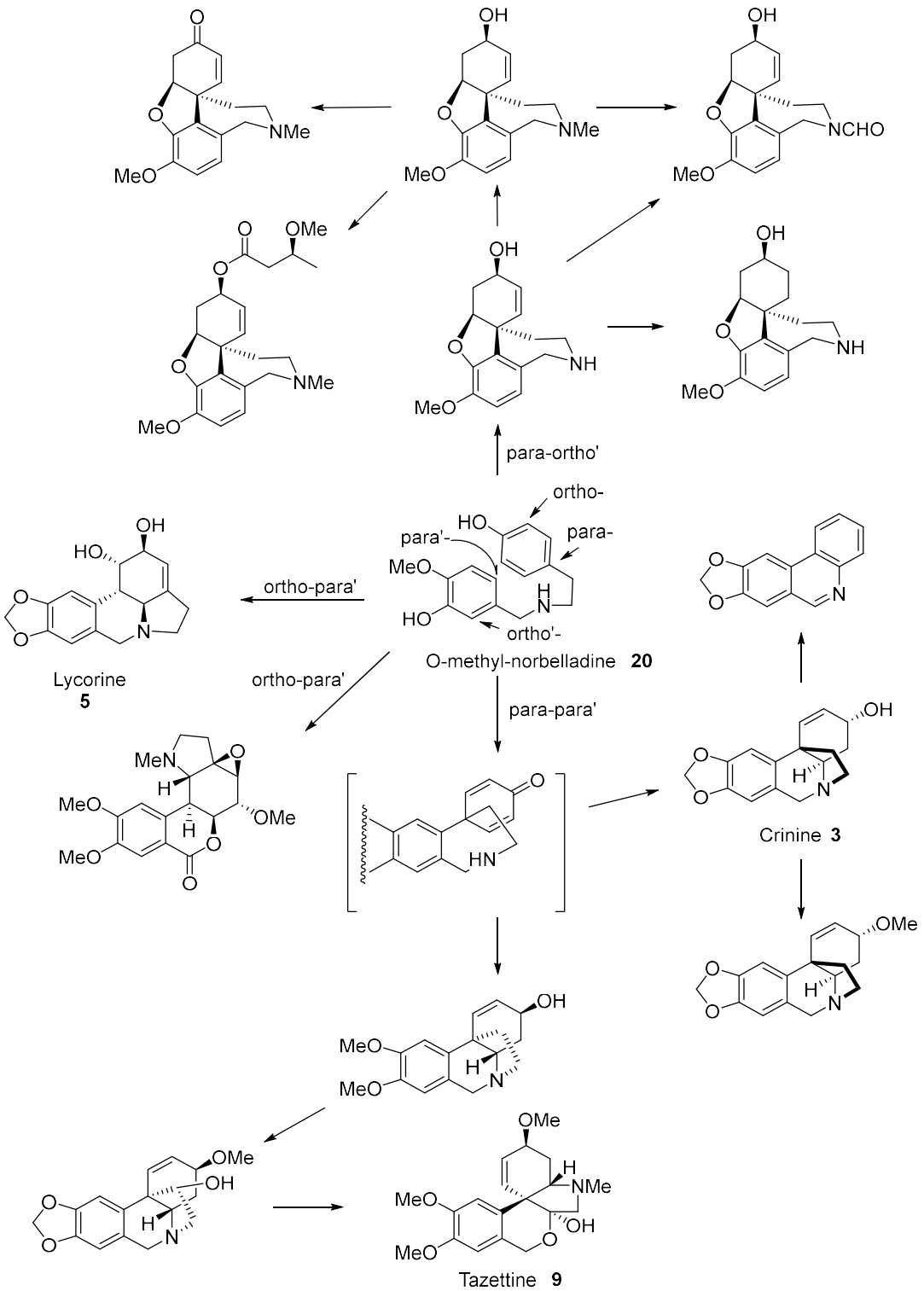


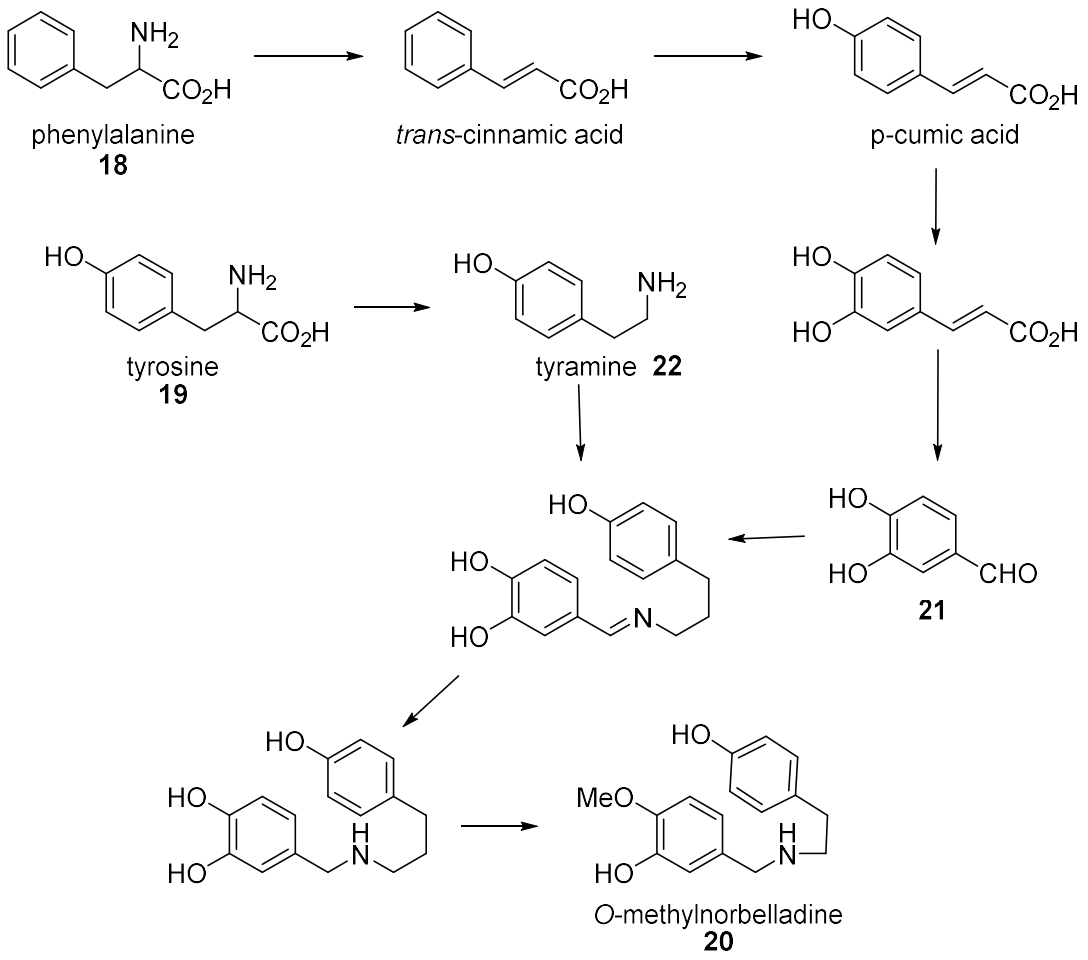
Figure 5. Three major isocarbostyril congeners of the Amaryllidaceae family, as well as their 7-deoxy analogues.

The Amaryllidaceae alkaloids are related because of their biogenesis from a common amino acid derived precursor norbelladine **1**, through which diverse skeletal rearrangements each alkaloid and its ring type are derived. Three classical structural types are recognized (McKillop, 1969) through which they contain or are derived from lycorine, crinane, or galantamine cores, although many other complex modifications are still prevalent (see figure 4 and scheme 1).



Scheme 2. Biogenesis of various alkaloids within the Amaryllidaceae family.

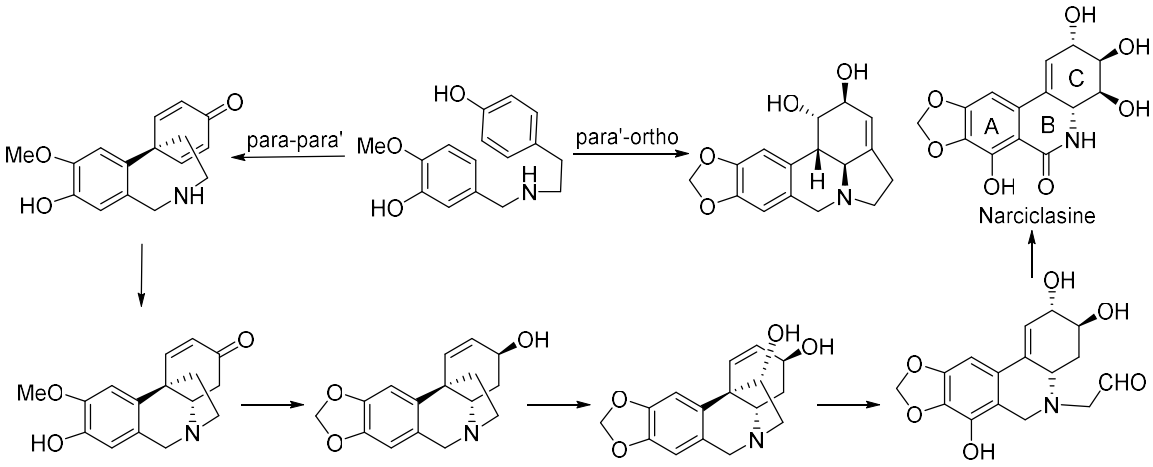
Although the explicit biosynthetic pathway for these alkaloids has yet to be discovered, it is rational to presume that they each share a common biosynthetic origin. What has been identified are the two major starting materials for the biogenesis of the alkaloids being phenylalanine (**18**) and tyrosine (**19**) (scheme 3).



Scheme 3. Biosynthesis of *O*-methylnorbelladine.

The two starting amino acids phenylalanine (**18**) and tyrosine (**19**) undergo transformations into protocatechuic aldehyde (**21**) and tyramine (**22**) respectfully. Subsequent coupling, reduction, and methylation leading to the central intermediate in the biosynthesis for the

Amaryllidaceae alkaloids *O*-methylnorbelladine (**20**). Through previous analyses there are at least three different types of intramolecular phenol oxidative couplings with *O*-methylnorbelladine **20** that take place (Jin, 2005). Phenanthridine type alkaloids are formed by the pathway following an oxidative cyclization of **20** but divergent processes involving regioselective oxidation cyclizations have led to a wide variety of structural types of alkaloids within the Amaryllidaceae family. Studies related to narciclasine (**7**) with labeled markers (Fuganti, Staunton, & Battersby, 1971; Fuganti & Mazza, 1971; Battersby, Hebert, McDonald, Ramage, & Clements, 1966; Fuganti, 1973) including tritium incorporation (Fuganti, 1975) and other tracer experiments have suggested that it is cogently produced by a para-para′ phenol oxidation process (Kornienko & Evidente, 2008)(scheme 4).



Scheme 4. Biosynthesis of narciclasine via para-para′ coupling.

Following all the studies performed and observing the similar structures among the congeners of narciclasine (**7**) it is believed they follow the same biosynthetic pathway.

1.2.1 References

A. R. Battersby, R. B. Hebert, E. McDonald, R. Ramage, J. H. Clements. *Chemical Communications*. **1966**, 603.

P. Bernfeld. *Pergamon Press: Oxford*. **1963**, 985.

C. Fuganti. *Academic Press: New York*. **1975**, 83.

C. Fuganti. *Gazzetta Chimica Italiana*. **1973**, 1255.

C. Fuganti, Mazza, M. *Chemical Communications*. **1971**, 1388.

C. Fuganti, Staunton, J, Battersby, A.R. *Chemical Communications*. **1971**, 1154.

Z. Jin. *Natural Products*. **2005**, 111.

A. Kornienko, A. Evidente. *Chemical Reviews*. **2000**, 1982.

A. McKillop. *Butterworths, London*. **1969**, 149.

G. R. Pettit, R. A. Backhaus, M. R. Boyd, A. W. J. Meerow. *Natural Products*. **1993**, 1682.

G. R. Pettit, G. M. Cragg, S. B. Singh, J. A. Duke, D. L. Doubek. *Natural Products*. **1990**, 176.

G. R. Pettit, V. Gaddamidi, G. M. Cragg, D. L. Herald, Y. Sagawa. *Chemical Communications*. **1984**, 1693.

F. Piozzi, C. Fuganti, r. Mondelli, G. Ceriotti. *Tetrahedron*. **1968**, 1119.

T. Okamoto, Y. Torii, Y. O. Isogai. *Chemical and Pharmaceutical Bulletin*. **1968**, 1860.

1.3 Antiviral Activities from Amaryllidaceae Alkaloids and Their Analogues

In the chemotherapy field, antiviral activity of 5-iodo-2'-deoxyuridine was first reported by Herrman and Gabliks in vitro (Herrman & Gabliks, 1961) and by Kaufman et al. (Kaufman, Martola, & Dohlman, 1962) for the treatment of herpes keratitis. Following the original success of an antiviral agent against HSV, several nucleoside analogues have been synthesized treating this viral infection. The smallest molecule-based antivirals are represented by nucleoside analogues, which have formed the backbone of chemotherapy of chronic infections due to HIV, hepatitis B or C viruses, and HSV. With favorable pharmacokinetic parameters and high antiviral potencies, a selection of nucleoside analogues have been used against acute infections caused by other medically important RNA and DNA viruses (Eyer, Nencka, Clercq, Seley-Radtke, & Růže, 2018). Additionally, the anti-HSV activity of two antibiotics cytochalasine B (Dix & Courtney, 1976) and tunicamycin (Pizer, Cohen, & Eisenberg, 1980) have been released, although the administration of such agents over time has led to resistance. Other limitations include the toxicity that comes with these agents, forming more of a need for new antiviral agents. Conventional antiviral drugs which interfere with viral proteins and/or functions have often led to the selection of drug resistance in virus populations evolving under selective pressures (Cuypers, et al., 2016; Maldonado & Mansky, 2018). Knowing these limitations, plants possessing physiologically active components directs research into gaining information from these rich sources of natural products. Previous studies relating to antiviral screening analyses of crude extracts from the Amaryllidaceae plant family were shown to deliver pronounced effects against several viruses under study (leven, et al., 1979;

Vanden Berghe, leven, Mertens, Vlietinck, & Lammens, 1978). The aforementioned Clivia miniate was shown to be one of the more active antiviral Amaryllidaceae plants, providing activities in tissue cultures against poliomyelitis, Coxsackie, Semliki forest, measles and HSV (Ieven, Vlietinck, Vanden Berghe, & Totte, 1982) caused by the alkaloids the plant was producing (Ieven & Vlietinck, 1981). This was later vindicated by further studies which showed that the only fractions containing the alkaloids provided any antiviral effects against Semliki forest, HSV and poliomyelitis viruses (Gorter, 1920; Boit, 1954; Briggs, Highet, Highet, & Wildman, 1956; Boit & Mehlis, Naturwissenschaften, 1961). Only six alkaloids were extracted out of Clivia miniate, but of them only **5** provided effective antiviral activity while the remainder were devoid of any notable potencies (Gorter, Bulletin du Jardin Botanique de Buitenzorg, 1920; Boit, 1954; Briggs, Highet, Highet, & Wildman, 1956). After performing a large scale extraction of the alkaloids from the leaves and roots of Clivia miniate, **5** and clivimine were seen as the two major constituents in the plant isolating in a 0.043% and 0.0085% yield respectfully (Ieven, Vlietinck, Vanden Berghe, & Totte, 1982). Although **5** shows antiviral activities, it comes with an effective range (maximum non-toxic dose to minimum effective dose) of 10 to 1 ug/mL on the viruses studied. Experiments also revealed that **5** was not inactivating the viruses directly, even when administered in higher concentrations that would exert cytotoxic activities. Together with **7** and **11**, **6** as well displayed strong in vitro RNA antiviral activity (Gabrielsen B. , et al., 1992). Other notable mentions of other plants in the Amaryllidaceae plant family showing antiviral activities include perennial plants of the *Haemanthus* genus (Ghosal, Saini, & Razdan, 1985; Louw, Regnier, & Korsten, 2002), *Narcissus tazetta L.*

(Martin S. F., 1987; Fuganti, 1975; Wildman, 1968), and the bulbs of *Hymenocallis littoralis*, *Hymenocallis caribaea*, and *Hymenocallis latifolia* (Pettit G. , et al., 1986; Pettit G. , Pettit, Backhaus, Boyd, & Meerow, 1993).

1.3.1 References

H. G. Boit. *Chemische Berichte*. **1954**, 1704.

C. K. Briggs, P. F. Highet, R. J. Highet, W. C. *Wildman. Journal of the American Chemical Society*. **1956**, 2899.

L. Cuypers, J. Snoeck, L. Kerremans, P. Libin, R. Crabbé, S. Van Dooren, G. Vuagniaux, A. M. Vandamme. *Infection, Genetics and Evolution*. **2016**, 169.

R. D. Dix, R. J. Courtney. *Virology*. **1976**, 127.

L. Eyer, R. Nencka, E. Clercq, K. Seley-Radtke, D. Růže. *Antiviral Chemistry and Chemotherapy*. **2018**, 1.

C. Fuganti. *Academic Press: New York*. **1975**, 83.

B. Gabrielsen, T. P. Monath, J. W. Huggins, D. F. Kefauver, G. R. Pettit, G. Groszek, M. J. J. Hollingshead, W.M. Kirsi, E. M. Shannon, E. M.Schubert. *Natural products*. **1992**, 1569.

S. Ghosal, K. S. Saini, S. Razdan. *Phytochemistry*. **1985**, 2141.

K. Gorter. *Bulletin du Jardin botanique de Buitenzorg*. **1920**, 331.

E. C. Herrman, Jr. J. Gabliks. *Cancer Chemotherapy Reports*. **1961**, 85.

M. Ieven, D. Vanden, A. Berghe, F. Mertens, A. Vlietinck, E. Lammens. *Planta Medica*. **1979**, 311.

M. Ieven, A. J. Vlietinck. *Pharmaceutisch Weekblad*. **1981**, 169.

M. Ieven, A. J. Vlietinck, D. A. Vanden Berghe, J. Totte. *Natural products*. **1982**, 564.

H. E. Kaufman, E. L. Martola, C. Dohlman. *Archives of Ophthalmology*. **1962**, 236.

C. A. Louw, T. J. Regnier, L. Korsten. *Journal of Ethnopharmacology*. **2002**, 47.

S. F. Martin. *Academic Press, New York*. **1987**, 251.

J. O. Maldonado, L. M. Mansky. *Viruses*. **2018**, 376.

G. R. Pettit, V. Gaddamidi, D. L. Herald, S. B. Singh, G. M. Cragg, J. M. Schmidt, F. E. M. Boettner, Y. Williams. Sagawa. *Natural Products*. **1986**, 995.

G. R. Pettit, Pettit, III, R. A. Backhaus, M. R. Boyd, A. W. Meerow. *Journal of Natural Products*. **1993**, 1682.

L. I. Pizer, G. H. Cohen, R. J. Eisenberg. *Virology*. **1980**, 142.

D. A. Vanden Berghe, M. Leven, F. Mertens, A. J. Vlietinck, E. Lammens. *Natural Products*. **1978**, 463.

W. C. Wildman. *Academic Press, New York*. **1968**, 307.

1.3A Antiviral Studies by Gabrielsen et. al. in 1992

Antiviral structure-activity relationships (SAR) have not been studied frequently in relation to lycorine derivatives, but in 1992 Gabrielsen et. al. conducted a large screen study of 23 Amaryllidaceae isoquinoline alkaloids (Figure 6) and related synthetic analogues by evaluating in cell cultures against RNA-containing flaviviruses (Japanese encephalitis, yellow fever, and dengue viruses), bunyaviruses (Punta Toro, sandfly fever, and Rift Valley fever viruses), alphavirus (Venezuelan equine encephalomyelitis virus), lentivirus (human immunodeficiency virus-type 1) and the DNA-containing vaccinia virus. The study established a multitude number of findings including (+)-***12*** being the most effective anti-flaviviral analogue exhibiting IC50 values of <0.003 - 0.015 ug/mL. Findings related to the antiviral pharmacophore found that by exchanging the trans-fused B/C-ring junction with a cis-fused junction, and loss or epimeric alcohols in the cyclohexane ring would result in a significant drop, or loss all together in antiviral activity (Gabrielsen B. , et al., 1992; Gabrielsen B. , et al., 1992).

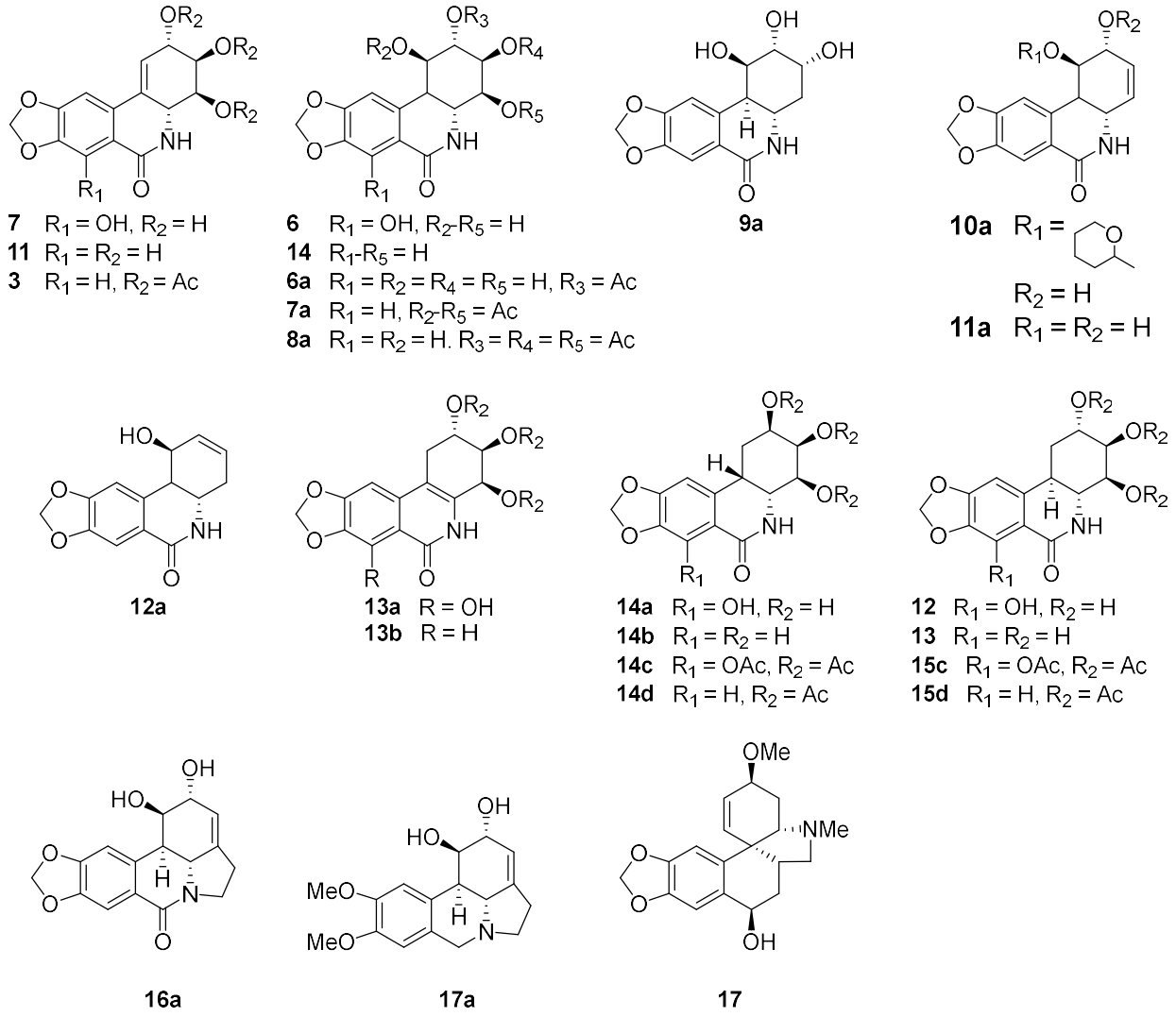


Figure 6. Amaryllidaceae isoquinoline alkaloids studied by Gabrielsen.

Narciclasine **7**, lycoricidine **11**, pancratistatin **6**, 7-deoxypancratistatin **14**, and acetates **6a-8a**, isonarciclasine **13a**, cis-dihydronarciclasine **14a**, trans-dihydronarciclasine **12**, their 7-deoxy analogues **13b**/**13**, lycorines **16a** and **17a**, and pretazettine **17** exhibited consistent in vitro activities against all three flaviviruses and against the bunyaviruses, Punta Toro and Rift Valley fever virus. Important to note was the activity seen against the sandfly fever-Sicilian (SF) virus only being observed with the 7-deoxy analogues. Negative

findings included the poor selectivity and the toxicity in uninfected cells (TC_{50}) at concentrations 10-fold that of the viral inhibitory concentration (IC_{50}). The study did confirm in the antiviral activities of **4** and **5** culminating their application towards mice infected with Japanese encephalitis, but only after approaching near toxic concentrations. Antiviral activity was consistently shown with the tested flaviviruses (JE, YF, dengue-4), and to a lesser degree the bunyaviruses (PT, SF, RVF) (Tables 2 and 3).

Table 2. Antiviral activity in vitro against flaviviruses: Japanese Encephalitis, yellow fever, and Dengue-Type 4.

Compound	Virus								
	Japanese Encephalatis (JE)[a]			Yellow Fever[a]			Dengue-4[b]		
	TC_{50}	IC_{50}	TI_{50}	TC_{50}	IC_{50}	TI_{50}	TC_{50}	IC_{50}	TI_{50}
7	0.031	0.008	4.08	0.037	0.006	6.1	0.06	0.015	4.0
11	0.27	0.056	4.9	0.29	0.053	5.6	0.25	0.059	4.2
3a	>1.0	inactive		>1.0	inactive		>25	inactive	
6	0.092	0.022	4.2	0.079	0.016	4.9	0.5	0.063	8.0
14	2.8	0.48	5.9	2.6	0.4	6.6	2.5	0.67	3.7
6a	10.8	3.3	3.3	21.0	4.8	4.3	50.0	1.5	33.3
7a	2020	724	2.8	>1000	262	3.8	not tested		
8a	16.4	4.5	3.6	8.3	2.2	3.8	100	<5.0	>20.0
9a	2000	inactive		1800	inactive		not tested		
10a	>100	inactive		>100	inactive		not tested		
11a	630	inactive		500	inactive		not tested		
12a	>100	inactive		>100	inactive		not tested		
13a	1.5	0.72	2.1	0.90	0.22	4.1	5.0	0.27	18.5
13b	>100	inactive		23.8	5.7	4.2	50.0	8.5	5.9
14a	4.9	0.96	5.1	5.2	1.3	3.9	>5.0	2.5	>2.0
14b	62.5	12.7	4.9	64.0	9.6	6.6	25.0	4.4	5.7
14c	29.0	8.1	3.5	97.0	[d]	[d]	not tested		
12	0.025	0.004	5.6	0.027	<0.003	>8.5	0.063	0.015	4.2
13	0.22	0.039	5.6	0.28	0.037	7.5	2.5	0.5	5.0
16a	2.7	0.33	8.2	2.04	0.28	7.3	2.5	0.24	10.4
17a	1.4	0.28	5.0	1.3	0.35	3.7	1.0	0.39	2.6
17	2.3	0.60	3.8	2.8	0.5	5.6	not tested		

[a]TC50 and IC50 obtained by MTT assay
[b]IC50 measured by plaque reduction; TC50 measured by cytopathic effect
[c]In ug/mL
[d]Viral cytopathic effect reduced 25-49% only

Table 3. Antiviral activity in vitro against Bunyaviruses: Punta Toro, Rift Fever, Sandfly Fever, and Dengue-Type 4.

Compound	Virus								
	Punta Toro[a]			Rift Valley Fever[b]			Sandfly Fever-Sicilian[a]		
	TC_{50}	IC_{50}	TI_{50}	TC_{50}	IC_{50}	TI_{50}	TC_{50}	IC_{50}	TI_{50}
7	0.029	0.0074	3.9	0.022	inactive		0.028	inactive	
11	0.27	0.042	6.3	0.83	0.15	5.5	0.26	0.058	4.5
3a	>1.0	inactive		<2.5	inactive		>1.0	inactive	
6	0.10	d	d	0.5	0.16	3.1	0.13	inactive	
14	2.9	0.66	4.3	21.5	5.1	4.3	4.5	1.7	2.7
6a	9.6	4.7	2.1	24.0	5.5	4.4	5.2	inactive	
7a	>320	inactive		>250	inactive		>320	inactive	
8a	10.6	2.5	4.2	<250	inactive		21.5	inactive	
9a	1200	inactive		>250	inactive		>320	inactive	
10a	>100	inactive		<250	inactive		>100	inactive	
11a	500	inactive		<250	inactive		>320	inactive	
12a	>320	inactive		250	inactive		>320	inactive	
13a	1.4	0.28	5.1	25.0	3.3	7.6	0.72	inactive	
13b	26.2	7.2	3.7	50.0	10.0	5.0	17.6	inactive	
14a	8.0	2.2	3.6	5.0	1.4	3.6	8.0	[d]	[d]
14b	68.0	14.0	4.8	Not tested			73.0	25	3.0
14c	77.0	12.0	6.4	not tested			21	[d]	[d]
12	0.026	0.008	3.3	Not tested			0.027	inactive	
13	0.34	0.057	5.9	0.5	0.25	2.0	0.25	inactive	
16a	2.3	0.50	4.6	5.0	0.93	5.4	1.4	inactive	
17a	2.3	0.60	3.9	3.8	0.63	6.0	2.5	inactive	
17	2.3	0.61	3.7	10.0	2.9	3.5	2.5	0.82	3.0

[a]TC50 and IC50 obtained by MTT assay
[b]IC50 measured by plaque reduction; TC50 measured by cytopathic effect
[c]In ug/mL
[d]Viral cytopathic effect reduced 25 - 49% only

Compounds **1 - 18** generally inhibited PT and RVF viruses but suffered from their poor selectivity, while viral cytopathic effects were unable to be reduced 50% by Pancratistatin against the PT virus. An important finding was found with relation to the SF virus that showed resistance against many of the alkaloids in the present study. The four alkaloids showing activities all shared a common structural feature with the loss of the C-7 hydroxy substituent (TC_{50} values). Other disadvantages were seen in **7** and its 7-deoxy analogue (lycoricidine) **11** being found to show the highest toxicity to host cells of any compounds in this series, and that by shifting the double bond from the C-1-C-10b position of the C-ring to the C-lOb-C-4a position (isonarciclasines **13a** and **13b**) that increased the TC50 values by 24 - 200-fold. By comparing pancratistatin **6** with its 7-deoxy analogue **14** (and similarly narciclasine **7** with lycoricidine **11**), host cell toxicity (TC50) was reduced by 8-32-fold after replacement of the C-7 hydroxy substituent with a hydrogen. The aforementioned value in ring-C hydroxylation towards antiviral efficacy was confirmed by comparing triol **9a**, diol **11a**, and mono-hydroxy **12a** with 7-deoxypancratistatin **14**. Only the latter compound **14** showed any antiviral activity. The study then conducted in-vivo experiments with Pancratistatin **6** and its 7-deoxy analogue **14** by evaluating in one or both variants of the murine JE virus models. Prophylaxis with **14** partially protected against JE viral infections in mice, but following their previous findings with many other alkaloids, the doses required approached unsafe levels in accomplishing its efficacy. Pancratistatin shared similar results, showing that at 6 mg/kg/day, it was toxic to 60% of the uninfected mice while a dose of 2 mg/kg/day was not sufficient in reducing JE virus induced mortality. The study communicated that pancratistatin's insolubility in aqueous media and limited

bioavailability may have been the cause of their poor reproducibility of pharmacokinetics from the in vivo test results. Overall, the chemotherapeutic doses bordered with the toxic doses so new analogues can be aimed in removing this liability. Sidwell et al. would later confirm **7** activity against Punta Toro virus in both in vitro and in vivo models for this infection (Sidwell, et al., 1994). Narciclasine's narrow therapeutic range although has been found against Rauscher leukemia virus in an in vitro model in NIH/3T3 cells due to its host cell toxicity (Furusawa, Irie, Combs, & Wildman, 1980).

1.3A.1 References

E. Furusawa, H. Irie, D. Combs, W. C. Wildman. *Chemotherapy*. **1980**, 36.

B. Gabrielsen, T. P. Monath, J. W. Huggins, J. J. Kirsi, M. Hollingshead, W. M. Shannon, G. R. Pettit. *New York: Plenum Press*. **1992**, 121.

B. Gabrielsen, T. P. Monath, J. W. Huggins, D. F. Kefauver, G. R. Pettit, G. Groszek, M. J. J. Hollingshead, W. M. Kirsi, E. M. Shannon, J. Schubert, B. DaRe. Ugarkar, M. Ussery, M. J. Phelan, J. Dare. *Natural Products*. **1992**, 1569.

S. C. Sidwell, R. W. Huffman, J. H. Barnard, D. L. Smee, D. F. Warren, R. P. Chirigos, M. M. Kende, J. W. Huggins. *Antiviral Research*. **1994**, 105.

1.3B Antiviral Studies by Kobayashi et. al. in 1989

The effects of alkaloids isolated from the Amaryllidaceae plant family on herpes simplex virus type 1 (HSV-1) were studied by Kobayashi et. al. in 1989 towards the relationship between the alkaloid structure and the mechanism of action for each

biosynthesized product (Renard-Nozaki, Kim, Imakura, Kihara, & Kobayashi, 1989). HSV-1 is a ubiquitous DNA virus from the Herpesviridae family that has been responsible for the substantial human morbidity and mortality. Its high prevalence in seropositivity rates surmounting 70% in elders, leading to recurrent cold sores, corneal infection, blindness, and seldomly encephalitis (Kinchington, Leger, Guedon, & Hendricks, 2012). The study was conducted using 36 alkaloids including five of the lycorine, seven of the tazettine, three of the lycorenine, seven of the galantamine, five of the cherylline, and nine compounds from the apogalanthamine group were synthesized. Of these tested alkaloids, only four were determined to have antiviral action based on having a favorable antiviral index of >5 (Table 4). These were lycorine-HCl, **16**, **17**, and hippeastrine.

Table 4. Cytotoxicity and antiviral activity of select Amaryllidaceae alkaloids.

	Toxicity (A) µg	Effective dose (B) µg	AI (A/B)
Lycorine group (2/5)			
- Lycorine	25	2	12.5
- Pseudolycorine	12.5	2	6.25
Tazettine group (2/7)			
- Tazettine	> 200	100	
- Pretazettine	20	4	5
Lycorenine group (1/3)			
- Hippeastrine	200	25	8
Galantamine group	Antiviral activity > 200		
Apogalanthamine group	Strong toxicity		
Cherylline group (3/5)			
- *O*,*O*-dimethyl-cherylline	>200	200	
- Lafinine	>200	200	
- *O*,*O*-dimethyl-lafinine	200	100	2

***MIC (toxicity) = maximum concentration at which 100% of the cells were still alive post treatment for a duration of 48 h, with no morphological difference with untreated cells. Effective dose = minimal concentration without traces of CPE 48 h post-HIV-infection. AI = antiviral index.

The study was successful in determining key features that were apparent for the more active alkaloids, showing a trend with the antiviral agents including a hexahydroindole ring with two functional hydroxy groups being established. In contrast to these findings, galantamine-type compounds containing benzazepine rings were neither cytotoxic nor antiviral, while many structures having dibenzazocine consequently showed toxicity profiles at low concentrations. The report showed that the antiviral activities from the alkaloids were caused via the inhibition of multiplication and not related to the direct inactivation of the extracellular viruses. A full explanation of its biological activity was not communicated, rather noting the mechanism of the antiviral effect could be partially reasoned as the blocking of viral DNA polymerase activity resulting in the reduction of viral DNA.

1.3B.1 References

P. R. Kinchington, A. J. Leger, J. M. Guedon, R. L. Hendricks. *Herpesviridae*. **2012**, *3*, 5.

J. Renard-Nozaki, T. Kim, Y. Imakura, M. Kihara, S. Kobayashi. *Virology Research Journal*. **1989**, 115.

1.3C Antiviral Studies by McNulty et. al. in 2016

Continuing forward with studies related to HSV-1, in addition to new research related to varicella zoster virus (VZV) McNulty et. al. studied **13** and three analogues **24**-**26** (figure 7) that were produced via asymmetric chemical syntheses (McNulty, et al., 2016).

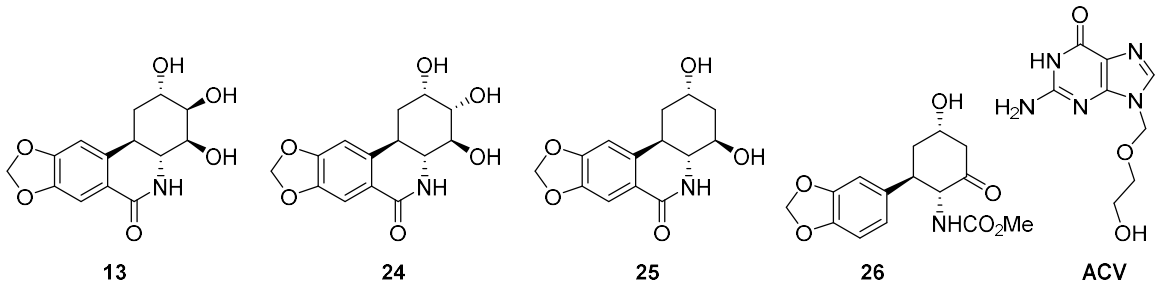


Figure 7. **13** and three analogues screened against HSV-1.

HSV-1 is a growing concern, leading to latent infections in sensory neurons that are refractory to clearance, forming an intractable lifelong reservoir that cycles with lytic infections. HSV-1 has yet to be treated to the extent which successfully eliminates the latent infection, although acute HSV-1 is readily treatable with nontoxic nucleoside analogues including acyclovir (**ACV**). This prevailing imbalance, on top of the growing ACV drug resistance has led to new antiviral solutions being proposed to remove the rising concern (Burrel, et al., 2013). Alkaloid **13** proved superior over the current standard for HSV-1 infection ACV, potentially inhibiting lytic HSV-1 infection, significantly reducing HSV-1 reactivation, and more potently inhibiting VZV lytic infection. The study involving the natural and non-natural lycorine-type alkaloids furthered the identification of an increased SAR, communicating the importance of the configurationally defined (R)-secondary alcohol at C3 for the efficacious inhibition of the lytic HSV-1 infection, as well as other antiviral activities. This was confirmed by comparing the activities of three analogues

synthesized with **6**, **7**, and **13** all of which possessing the (3R)-stereochemistry provided potent antiviral activities. This acknowledged the identification of **13** as the functionally minimum antiviral pharmacophore.

1.3C.1 References

S. Burrel, C. Aime, L. Hermet, Z. Ait-Arkoub, H. Agut, D. Boutolleau. *Antiviral Research*. **2013**, 365.

J. McNulty, L. D'Aiuto, Y. Zhi McClain, C. Zepeda-Velezquez, S. Ler, H. A. Jenkins, M. B. Yee, P. Piazza, R. H. Yolken, P. R. Kinchington, V. L. Nimgaonkar. *ACS Medicinal Chemistry Letters*. **2016**, 46.

1.3D Antiviral Studies by McNulty et. al. in 2018

Following the potent antiviral activities discovered with **12** and **13** (high therapeutic index) against Zika virus (Revu, et al., 2016; D'Aiuto, et al., 2018), HSV-1, and varicella zoster virus (McNulty, et al., 2016), more findings surfaced related to a series of quinazoline derivatives against HSV-1 (Brown C. E., et al., 2017). The investigation was initiated by a rapid six-step (four processes) convergent route to quinazolinone-Amaryllidaceae alkaloid hybrid molecules from L-arabinose (figure 8) (Brown C. , et al., 2018).

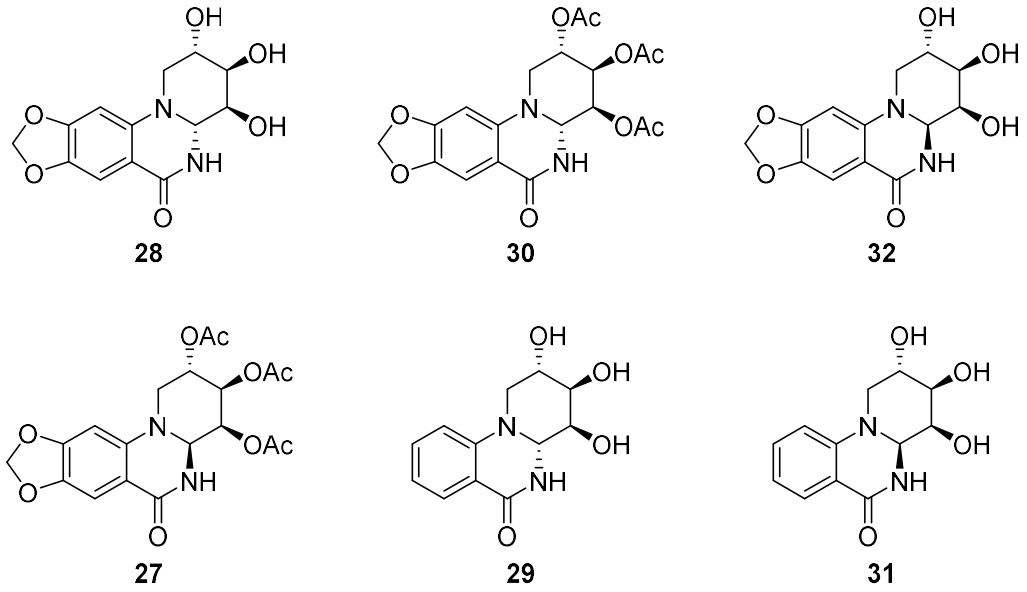


Figure 8. 10b-aza-analogues synthesized all devoid of antiviral activity.

Unexpectedly with the complete conformational and functional group overlap the synthesized molecules share, 10b-aza-analogues all showed no antiviral activity. Chair conformational analysis on ring-C revealed compound **32** adopting a different conformation than naturally observed in the cabocyclic natural series, identical to that observed in the X-ray structure of compound **31** with the hydroxy groups acylated. Such findings revealed the critical importance in the phenanthridone type chair conformation for antiviral activity, and the importance of *trans*-dihydrolycoricidine's electronic properties in ring-C for delivering potent activities. Investigation into the molecule's electrostatic potential heat maps discovered that although *trans*-dihydrolycoricidine displays an electropositive hole over ring-C, aza-analogue **28** is less electropositive, shedding light into the possible role played by ring-C with π-type secondary orbital interactions with the antiviral target. The study demonstrated the impotence use with the exchange of the 10b-C

with a nitrogen atom, presenting findings heavily set associated with ring-C, while minute changes were reflected in rings-A and B.

1.3D.1 References

L. D'Aiuto, J. McNulty, C. Hartline, M. Demers, R. Kalkeri, J. Wood, L. McClain, A. Chattopadhyay, Y. Zhi, J. Naciri, A. Smith, R. Yolken, K. Chowdari, C. Zepeda-Velazquez, C. Babu Dokuburra, E. Marques, R. Ptak, P. Kinchington, S. Watkins, M. Prichard, D. Bloom, V. Nimgaonkar. *Scientific Reports*. **2018**, *8*, 16662.

C. Brown, T. Kong, J. Britten, N. Werstiuk, J. McNulty. L. D'Aiuto. Demers, M. Nimgaonkar, V. *ACS Omega*. **2018**, 11467.

C. E. Brown, T. Kong, J. McNulty, L. D'Aiuto, K. Williamson, L. McClain, P. Piazza. *Bioorganic & Medicinal Chemistry Letters*. **2017**, 4601.

O. Revu, C. Zepeda-Velázquez, A. J. Nielsen, J. McNulty, R. H. Yolken, L. Jones-Brando. *ChemistrySelect*. **2016**, 5895.

J. McNulty, L. D'Aiuto, Y. Zhi, L. McClain, C. Zepeda-Velázquez, S. Ler, H. A. Jenkins, M. B. Yee, P. Piazza, R. H. Yolken, P. R. Kinchington, V. L. Nimgaonkar. *ACS Medicinal Chemistry Letters*. **2016**, 46.

1.4 First Isolation, Biological Activities, and Syntheses

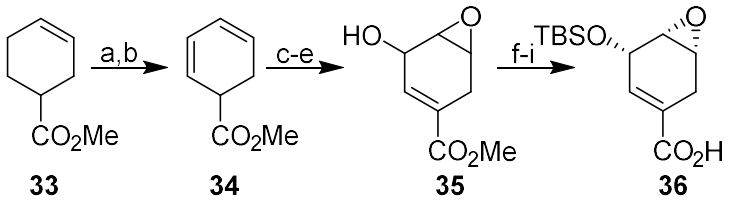
Active research over the last 200 years has advanced in regard to the Amaryllidaceae plant family due to their unique type of metabolites they produce, attracting

more attention to their syntheses. Their medicinal efficacy has made them valuable hosts for pharmacologically active compounds. To this day new alkaloids are still being extracted, but already more than 600 structurally diverse alkaloids have been found, covering 11 major classes (see figure 2 and scheme 2). Discoveries from 1877 marked the introduction of these metabolites from *N. pseudonarcicuss* named lycorine, but more have been extracted to see their promising biological activities. This section will go over select examples of the more well recognized isocarbostyrils. Their first enantioselective asymmetric syntheses will be presented, including one addition total synthesis of pancratistatin that features a sequence generating five new stereocenters in one step.

1.4A (+)-Narciclasine

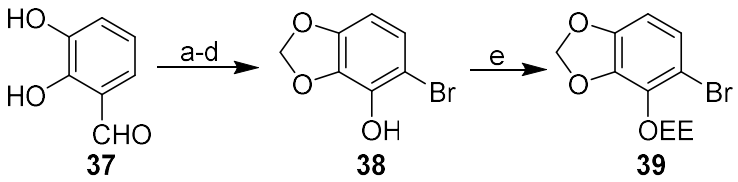
Narciclasine (also known as lycoricidinol **7**) is a non-basic constituent of the Amaryllidaceae plant family and was the first isocarbostyril isolated in 1967 from *Narcissus* bulbs (Fuganti & Mondelli, 1968). It has been isolated at concentrations between 100 to 120 mg/kg in *Narcissus* bulbs, but during peak times in the year it has been isolated in the bulbs of *Narcissus incomparabilis Mill. Var. Helios* harvested in the month of March at 200 mg/kg (Ingrassia, Lefranc, Mathieu, Darro, & Kiss, 2008). Despite narciclasine being the earliest isocarbostyril congener discovered it has seen little attention from a synthetic standpoint. Narciclasine targets the 60S subunit of ribosomes affecting protein biosynthesis, in addition to GTPase elongation factors Eef1A and RhoA that damages cytoskeleton organization (Fürst, 2016; Lefranc, et al., 2009; Van Goietsenoven, et al., 2013). Modern studies related to the Zika virus (ZIKV) recognized narciclasine as a potent

anti-Zika compound inhibiting the cytopathic effect of the virus by 90% at sub-micromolar concentrations. The compound does present negative effects, including the inhibition of human cytochrome P450 3A4, and not providing effective treatments towards mouse tumor models. When removing the double bond in ring-C, inhibition of CYP450 3A4 now becomes diminished, opening up the possibility for its potential as a human chemotherapeutic (Mcnulty, et al., 2011; Mcnulty, et al., 2009; Fürst, 2016). It was not until 1997 when the first total synthesis of (+)-narciclasine in enantiomerically pure form was reported (Rigby & Mateo, 1997). Their synthetic strategy involved the transformation of commercially available 3-cyclohexene-1-carboxylic acid employing a modification of the Berchtold sequence performed earlier for the synthesis of chorismite derivatives (scheme 5) (Pawlak & Berchtold, 1987).



Scheme 5. Preliminary preparation of the precursors for the first total synthesis of (+)-narciclasine: a) NBS, AIBN, PhI, reflux; b) AIBN, Bu_3SnH, PhH, 75% over two steps; c) O_2, *hv*, rose bengal,; d) $(PhP)_2RuCl_2$, CH_2Cl_2; e) NaOMe, MeOH, 30% over three steps; f) butyryl chloride, TEA; g) cholesterol esterase; h) TBSCl, imidazole; i) LiOH, MeOH, H_2O, 42% over four steps.

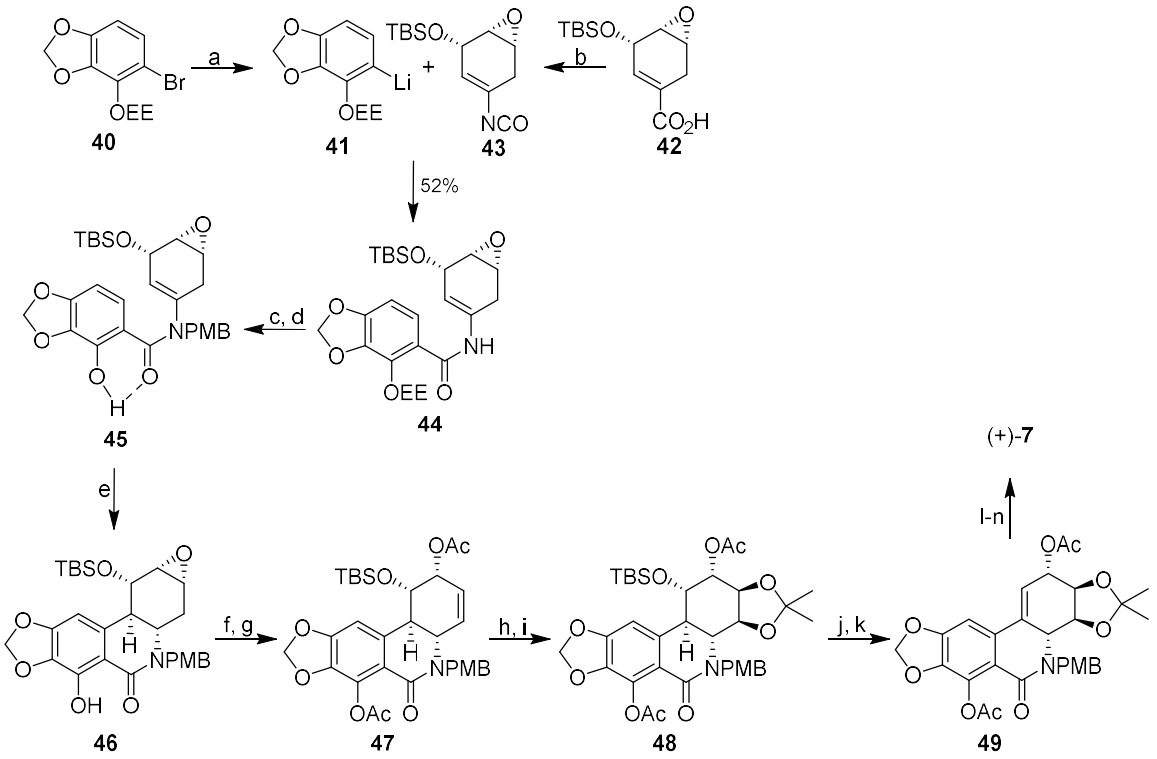
The corresponding ring-A fragment was prepared in four steps from commercially available 2,3-dihydroxybenzaldehyde **37** employing previously reported chemistry (Brown, Loriot, & Robin, 1982).



37 **38** **39**

Scheme 6. Initial stages in the synthesis towards the formation of ring-A for (+)-narciclasine: a) CH_2Br_2, K_2CO_3; b) *m*-CPBA; c) KOH/EtOH; d) CF_3CO_2Ag, Br_2, 45% over four steps; e) ethyl vinyl ether, PPTS, 70%.

The synthesis of **36** and **39** in doing so arranged the reagents required for the coupling between rings-A and C fragments prior to cyclization. Metalation of **40** followed by the addition of isocyanate **43** derived from **36** via lipase resolution afforded the enamide **44** then subsequently protected with PMB and selective deprotection forming the free phenol **45** which served as a vital component for the cyclization in the next step forming the *trans*-fused phenanthridone skeleton. Irradiation using 254nm in benzene (Rayonet photochemical reactor) generated the desired *trans*-fused phenanthridone **46** as a single diastereomer in 46% yield based on the recovery of starting material. This key step incorporated a hydrogen-bond-directed aryl enamide photocyclization tethering each ring for the final product. Exploiting the intramolecular hydrogen bonding between the C7 hydroxyl substituent, with the proximate enamide carbonyl oxygen to establish conformational control throughout the course of the cyclization. In their studies methanol was tested and found to work as well, reinforcing the notion that intramolecular hydrogen bonding was serving its purpose in controlling the course of the reaction forming the specific regioisomer.



Scheme 7. Total synthesis of (+)-narciclasine by Rigby and Matteo: a) *n*-BuLi, THF, -78 °C, DPPA, TEA, PhH, c) PMBBr, NaH; d) PPTS, MeOH, 76% over two steps; e) *hv*, PhH, 46%; f) $(PhSe)_2$, $NaBH_4$, H_2O_2; g) NaH, AcCl, 48% over two steps; h) OsO_4, TMNO, *t*-BuOH; i) TsOH, $(CH_3)_2C(OMe)_2$, 76% over two steps; j) TBAF, THF; k) Burgess Rgnt, 64% over two steps; l) K_2CO_3, MeOH; m) *n*-BuLi, THF, O_2; n) TsOH, 37% over three steps.

Following the isolation of the desired *trans*-fused B/C-ring junction the synthesis was carried through until reaching the final steps involving the deprotection of the C1 TBS group, and immediate dehydration with Burgess reagent in refluxing benzene to provide (+)-**7**.

1.4A.1 References

E. Brown, M. Loriot, J. P. Robin. *Tetrahedron Letters*. **1982**, 949.

C. Fuganti, R. Mondelli. *Tetrahedron*. **1968**, 1119.

R. Fürst. *Planta Medica*. ***2016***, 1389.

L. Ingrassia, F. Lefranc, V. Mathieu, F. Darro, R. Kiss. *Translational Oncology*. **2008**, *1*, 1.

F. Lefranc, S. Sauvage, G. Van Goietsenoven, V. Megalizzi, D. Lamoral-Theys, O. Debeir, S. Spiegl-Kreinecker, W. Berger, V. Mathieu, C. Decaestecker, R. Kiss. *Molecular Cancer Therapeutics*. **2009**, 1739.

J. Mcnulty, J. J. Nair, M. Singh, D. J. Crankshaw, A. C. Holloway, J. Bastida. *Bioorganic & Medicinal Chemistry Letters*. **2009**, 3233.

J. Mcnulty, A. Thorat, N. Vurgun, J. J. Nair, E. Makaji, D. J. Crankshaw, A. C. Holloway, S. J. Pandey. *Natural Products*. **2011**, 106.

J.C. Pawlak, G. A. Berchtold. *Journal of Organic Chemistry*. **1987**, 1765.

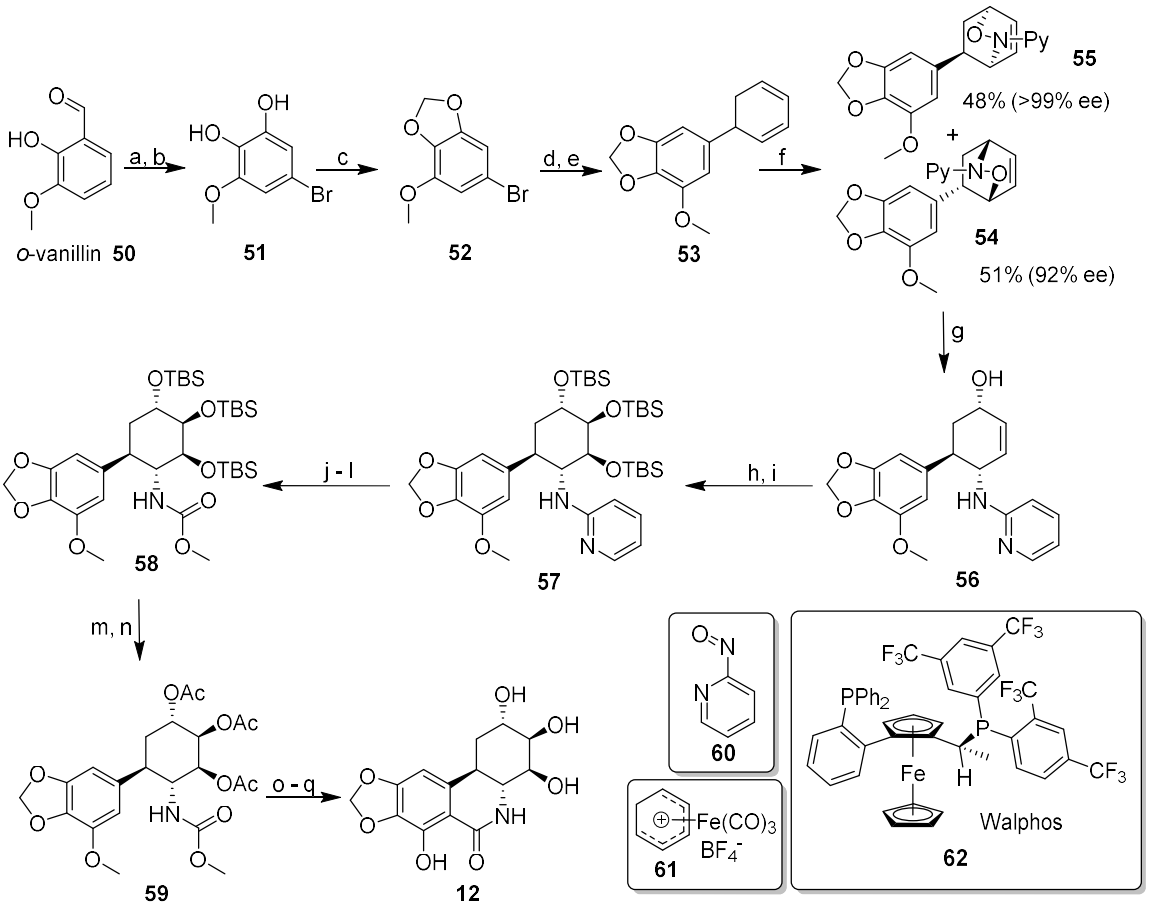
J. H. Rigby, M. E. Mateo. *Journal of the American Chemical Society*. **1997**, 12655.

G. Van Goietsenoven, V. Mathieu, F. Lefranc, A. Kornienko, A. Evidente, A. Kiss. *Medicinal Research Reviews*. **2013**, 439.

1.4B (+)-*trans*-Dihydronarciclasine

With cancer being the second leading cause of disease worldwide, significant attention has been directed to the phenanthridone alkaloids. Although **12** has been reviewed showing its anticancer activity to be higher when compared to other more well investigated congeners, not as much attention has been pushed in its direction for biological and synthetic studies. Increased attention although has picked up, and more studies are being directed to learning about its biological activities (Zhao, et al., 2019), in addition to the total synthesis approaches being pursued (Shin, Choi, & Cho, 2007; Shin, Choi, & Cho, 2007; Jana & Studer, 2008; Cho & Cho, 2008). The alkaloid has seen recent studies related to the Zika virus (ZIKV) and was recognized as a potent anti-Zika compound inhibiting the cytopathic effect of the virus by 90% at sub-micromolar concentrations evaluated. Research from the National Cancer Institute reports **12** showing the highest anticancer activity of the phenanthridone alkaloids. A large screen comparison of **12** and **13** derivatives each showing a modified ring-A towards the compound's antiproliferative activity against 60 human tumour cell lines (NCI60), representing leukemia, melanoma, and cancers of the lung, colon, brain, ovary, breast, prostate, as well as kidney in vitro found that (±)-**12** has the highest potency as a cytotoxic molecule among the 13 alkaloids screened (Pettit, Pettit III, Backhaus, Boyd, & Meerow, 1993; Varró, et al., 2019). It was also reported that the SAR results revealed that the presence of the hydroxy substituent at C7 and the rigid 1,3-benzodioxiole scaffold were essential for the corresponding antiproliferative activities. It was not until 1990 when the new biosynthetic product (+)-**12** was first discovered in a Chinese medicinal plant *Zephyranthes candida*. In 2007 the first total synthesis of the

racemate was reported by Cho et. al. (Shin, Choi, & Cho, 2007; Shin, Choi, & Cho, 2007), and in 2008 the first total synthesis of enantiomerically pure (+)-**12** was reported by Jana and Studer (scheme 8).



Scheme 8. First total synthesis of enantiomerically pure (+)-**12** reported by Jana and Studer: a) Br_2, AcOH; b) NaOH, H_2O_2, H_2O; c) DBM, K_2CO_3, DMF, 80 °C, 73% over three steps; d) *n*-BuLi, CuCN, THF, -78 °C; e) $Ce(NH_4)_2(NO_3)_6$, acetone, 0 °C, 85% over two steps; f) $CuPF_6(MeCN)_4$, Walphos (10%), CH_2Cl_2, -78 °C, **54** 51%, **55** 48%; g) $[Mo(CO)_6]$, $NaBH_4$, $MeOH/H_2O$, 90%; h) $K_2OsO_2(OH)_4$, NMO, acetone/H_2O; i) TBSCl, imidazole, DMF, 75 °C, 77% over two steps; j) MeMgCl, MeOCOCl, THF; k) MeOTf, CH_2Cl_2, 0 °C; l) NaOH, $MeOH/H_2O$, 50 °C, 88% over three steps; m) TBAF, THF; n) Ac_2O, Py, 95% over two steps; o) Tf_2O, DMAP, CH_2Cl_2; p) BBr_3, CH_2Cl_2; q) NaOMe, MeOH, 33% over three steps.

Both syntheses utilized an important step for the formation of the phenanthridone skeleton involving the Banwell modified Bischler-Napieralski reaction during the later stages of their syntheses (see chapter 1.5). The key step in the synthesis involves the enantioselective nitroso Diels-Alder reaction of racemic diene **53** to separable adducts **54** and **55** that had been previously developed by the group (Jana & Studer, 2007). The synthesis begins with a bromoanisole derivative **52** previously optimized from *o*-vanillin in three steps (Magnus & Sebhat, 1998). Subsequent transmetalation via a bromine-lithium exchange using copper to enable the cross-coupling with an iron-complexed cyclohexadienyl cation **61**, with subsequent oxidative decomplexation providing adduct diene **53** in an 85% overall yield. Succeeding the production of both rings-A and C, a copper catalyzed regiodivergent Diels-Alder reaction between diene **53** and 2-nitrosopyridine **60** using a chiral Walphos ligand **62** was executed converting each enantiomer of the racemic diene into separable regioisomeric cycloadducts (Jana & Studer, 2007; Sturm & Weissensteiner, 2003). The procedure provided a moderate 48% yield towards the desired regioisomer **55** but was successful in providing high enantioselectivity of >99% *ee*. Reductive nitrogen-oxygen bond cleavage employed by molybdenum hexacarbonyl and sodium borohydride established the correct stereochemistry for the C2 and C4a substituents in a 90% yield. Diastereoselective dihydroxylation with potassium osmate and global TBS protection of ring-C hydroxyl substituents provided **57** in a 77% yield. Carbamoylation of the amino group set up the sequential methylation and basic hydrolysis of the pyridine ring. Subsequent desilylation and O-acetylation afforded the triacetate derivative **59** in a 95% yield, setting the stage for Banwell's modified Bischler-Napieralski reaction to produce

ring-B which underwent good regioselectivity. Following subsequent deprotection steps, the synthesis was completed in 17 linear steps with a 5.6% overall yield.

1.4B.1 References

Y.S. Cho, C.G. Cho. *Tetrahedron*. **2008**, 64, 2172.

C. K. Jana, A. Studer. *Chemistry - A European Journal*. **2008**, *14*, 6326.

C. K. Jana, A. Studer. *Angewandte Chemie International Edition*. **2007**, 6542.

C. K. Jana, A. Studer. *Angewandte Chemie International Edition*. **2007**, 6662.

P. Magnus, I. K. Sebhat. *Tetrahedron*. **1998**, 15509.

G. R. Pettit, G. R. Pettit III, R. A. Backhaus, M. R. Boyd, A. W. Meerow. *Natural Products*. **1993**, 1682.

I. J. Shin, E.S. Choi, C.G. Cho. *Angewandte Chemie International Edition*. **2007**, *46*, 2303.

I. J. Shin, E.S. Choi, C.G. Cho. *Angewandte Chemie.* **2007**, *119*, 2353.

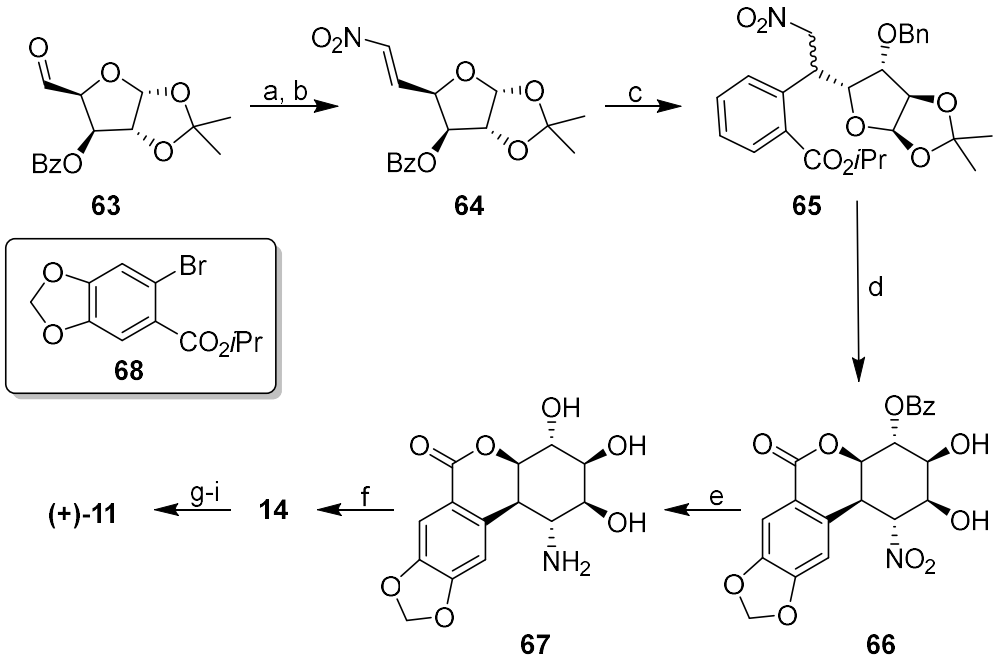
T. Sturm, W. Weissensteiner. *Advanced Synthesis & Catalysis*. **2003**, 160.

G. Varró, P. Pálchuber, B. Pogrányi, A. Simon, L. Hegedûs, L. Kádas. *European Journal of Medicinal Chemistry*. **2019**, 76.

D. Zhao, M. Y. Gu, L. J. Zhang, H. J. Jeon, Y. B. Cho, H. O. Yang**.** *Journal of Agricultural and Food Chemistry*. **2019**, *67*, 9796.

1.4C (+)-Lycoricidine

Lycoricidine **11** (also known as 7-deoxynarciclasine) was first discovered in *L. radiata* in 1968 (Okamoto, Torii, & Isogai, 1968). The compound was isolated in 3.2 mg/kg but was found in higher concentrations in the bulbs of *H. littoralis* collected in Hawaii at 118 mg/kg (Pettit G. R., Pettit, Backhaus, & Boettner, 1995). Ensuing studies revealed the alkaloid to show antitumor activities, as well as being a plant growth inhibitor. In contrast to narciclasine's limited synthetic pursuit, it would seem lycoricidine has attracted some of the voided attention. In 1975 Ohta and Kimoto reported its first racemic synthesis making it the first polyhydroxylated phenanthridone to be synthesized (Ohta & Kimoto, 1975). Apart of the lycorane isocarbostyrils, interest continues in its direction because of the nanomolar anticancer activity it provides. Soon following Paulsen and Stubbe reported the first enantioselective synthesis of (+)-lycoricidine in 1982 (Paulsen & Stubbe, 1982). The synthesis made use of a chiral pool starting material being a glucose derivative **63** because it already had installed four stereocenters matching (+)-lycoricidine (scheme 9).



Scheme 9. The first enantioselective synthesis of (+)-**11** by Paulsen and Stubbe: a) (i) CH_3NO_2, NaOH; (ii) Ac_2O, TsOH; b) K_2CO_3, benzene, 71% over two steps; c) **68**, *n*-BuLi, THF; d) (i) AcOH, H_2O; (ii) $NaHCO_3$, MeOH, 34%; e) H_2, Pd/C, MeOH, 77%; f) K_2CO_3, MeOH, 72%; g) BzCl, Py, DMAP; h) $SOCl_2$, Py; 70% over two steps; i) NH_3, MeOH.

This derivative was subsequently transformed into nitroolefin **64**, and afterwards reacted with the lithium derivative of Piperonylic acid, isopropyl ester **68** to give **65** through conjugate addition. Following the acidic deprotection the compound was cyclized to cyclohexane **66**, at which point reduction and recyclization synthesized **14** (7-deoxypancratistatin). The isocarbostyril congener was subsequently set forth to a selective reprotection followed by an elimination to isolate **(+)-11**.

1.4C.1 References

S. Ohta, S. Kimoto. *Tetrahedron*. **1975**, 2279.

T. Okamoto, Y. Torii, Y. Isogai. *Chemical and Pharmaceutical Bulletin*. **1968**, 1850.

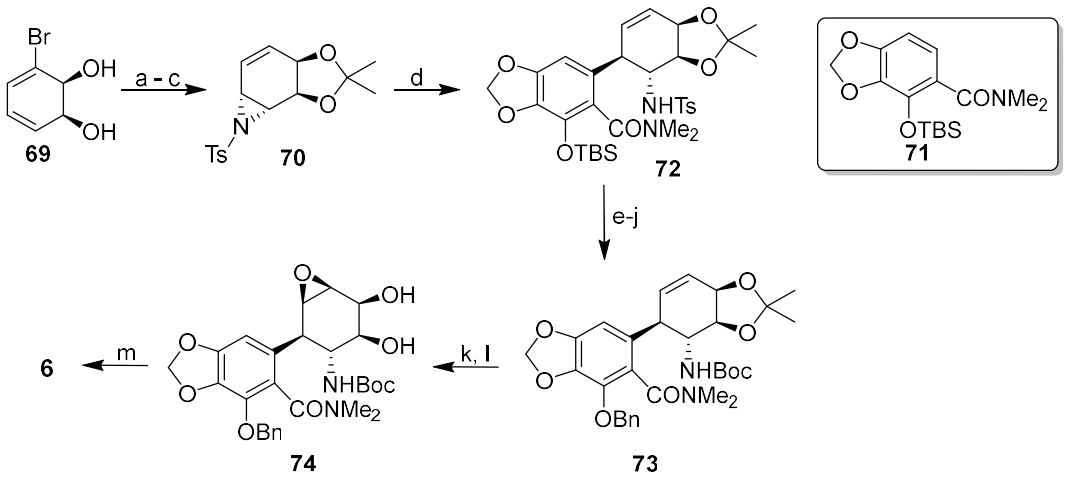
H. Paulsen, M. Stubbe. *Tetrahedron Letters*. **1982**, 3171.

G. R. Pettit, G. R. Pettit III, R. A. Backhaus, F. E. Boettner. *Journal of Natural Products*. **1995**, *58*, 37.

1.4D (+)-Pancratistatin

Pancratistatin was first discovered by Pettit, et. al. in a Hawaiian species reclassified as *H. littoralis* in a low yield of 0.028% in 1984 (Pettit G. , Gaddamidi, Cragg, Herald, & Sagawa, 1984). The structure was elucidated by NMR spectral analysis, and later confirmed by the X-ray crystal structure of pancratistatin monomethyl ether. Pancratistatin has since received a significant amount of attention from the synthetic community (Manpadi & Kornienko, 2008) for its biological activities including its strong *in vitro* cancer cell growth inhibitory activities against the US National Cancer Institute (NCI) panel of cancer cell lines (Pettit, Backhaus, Boyd, & Meerow, 1993; Pettit G. , et al., 1986). Pancratistatin was shown to target complex I and III of the mitochondria revealing its selectivity and mode of action affecting cancerous cells inducing apoptosis (programmed cell death) (McLachlan, Kekre, McNulty, & Pandey, 2005). Studies have shown its efficacy in mouse models using human xenografts for different cancers (Pandey & McNulty, 2011; Griffin, Karnik, McNulty, & Pandey, 2011; Ingrassia, Lefranc, Mathieu, Darro, & Kiss, 2008; D. Ma, et al.,

2017). A vast amount of studies were performed in determining its minimum pharmacophore, communicating that the C2, C3, and C4 hydroxyls with a trans-fused ring-B/C junction play an important role in its anticancer activities (Manpadi & Kornienko, 2008). From its structural similarity to narciclasine, pancratistatin has been surmised to exert its antitumor potency by disrupting protein synthesis and has been shown in recurring studies for its selective toxicity to cancer cells over normal cells (Kekre, Griffin, McNulty, & Pandey, 2005; McLachlan, Kekre, McNulty, & Pandey, 2005; Pandey S. , Kekre, Naden, & McNulty, 2005; Pettit G. R., et al., 1986). Pancratistatin and its 7-deoxy analogue are currently the only known agents (not including interferon inducers) with the ability to show significant chemotherapeutic efficacy in a Japanese encephalitis virus-infected mouse model. In 1989 Danishefsky and Lee reported the first racemic synthesis for pancratistatin (Danishefsky & Lee, 1989), while the first enantioselective semi-synthesis would be reported by Hudlicky et. al. that utilized a microbial metabolite to produce their starting material **69** in 1995 (Tian, Hudlicky, & Konigsberger, 1995; Hudlicky, et al., 1996). Despite diol **69** being commercially available, the group prepared its starting material by whole-cell oxidation of bromobenzene with *Pseudomonas putida* 39/D (Hudlicky, Boros, & Boros, 1992; Gibson, Hensley, Yosioka, & Mabry, 1970). Tosylaziridine **70** was generated following an Evans procedure (Yamada, Yamamoto, & Okawara, 1975; Evans, Paul, & Bilodeau, 1991), and was subsequently subjected to nucleophilic opening with a cuprate derived from *ortho*-lithiation of amide **71** affording exclusively the desired S_N2 opening.



Scheme 10. The first semi-synthesis of (+)-**6** by Hudlicky: a) 2,2-DMP, TsOH, acetone; b) PhI=NTs, $Cu(acac)_2$, MeCN, 27% over two steps; c) *n*-Bu_3SnH, AIBN, THF, reflux, 78%; d) (i) **71**, *s*-BuLi, TMEDA, THF; (ii) CuCN, (iii) **70**, BF_3, Et_2O, 49%; e) (i) *s*-BuLi, THF; (ii) $(Boc)_2O$, 68%; f) Na/anthracene, DME, 62%; g) SMEAH, morpholine, THF, 72%; h) BnBr, K_2CO_3, DMF, 83%; j) (i) $NaClO_2$, KH_2PO_4, 2-methyl-2-butene, *t*-BuOH, H_2O; (ii) CH_2N_2, 98%; k) HOAc, THF, H_2O, 73%; l) *t*-BuOOH, $VO(acac)_2$, benzene, 53%; m) $PhCO_2Na$, H_2O, reflux, 51%.

They reported finding that a higher-order phenyl cyanocuprate accomplished this from previous reports related to epoxides between the selectivity between S_N2 and S_N2' openings (Lipshutz, Kozlowski, & Wilhelm, 1982; Hudlicky T. , Tian, Kónigsberger, & Rouden, 1994; Marshall, 1989). This involved amide **71** being subjected to ortho-metalation below -90 °C following Snieckus's protocol being converted in situ into the lithium cyanocuprate $Ar_2Cu(CN)Li_2$ (Lipshutz, Kozlowski, & Wilhelm, 1982) whose addition provided cleanly tosylamide **72** in 75% yield almost exclusively as one atropisomer (equilibrated slowly at room temperature to its more stable form, presumably being α-form). This is where the synthesis took a turn in the other direction because detosylation followed by cyclization to form ring-B should have permitted access to the phenanthridone skeleton for the final product, but due to the unforeseen difficulties associated by the atropoisomerism this was

unsuccessful. Four alternative routes were pursued but all came back unsuccessful. This egregious issue was circumvented at the expense of a reduced efficacy. Additional steps towards the phenanthridone skeleton included many functionalizations lengthening the total synthesis, showing that the benzamide and the methods of its subsequent transformations were incompatible with the functional group sensitivities present. Upon the generation of epoxide **74** being available to diaxial opening, the almost neutral conditions applied at 100 °C over the course of 6 days (an altered procedure towards D-*chiro*-inositol preparation) (Mandel & Hudlicky, 1993) accomplished not only the stereospecific epoxide opening, but a remarkable series of events including thermal cleavage of the Boc group and cyclization to the lactam, as well as debenzylation to **6** in 13 steps with a final step isolation of 51% yield. Allowing the reaction to continue for 6 days was also shown to play an important role, because from reactions of allowing only 2 days to react this produced the benzyl-protected pancratistatin over the formation of the free hydroxy substituent on C7 which required an additional step of hydrogenation to the final product.

1.4D.1 References

S. Danishefsky, J. Y. Lee. *Journal of the American Chemical Society*. **1989**, 4829.

D. A. Evans, M. Paul, T. J. Bilodeau. *The Journal of Organic Chemistry*. **1991**, 6744.

D. T. Gibson, M. Hensley, H. Yosioka, T. J. Mabry. *Biochemistry*. **1970**, 1626.

C. Griffin, A. Karnik, J. McNulty, S. Pandey. *Molecular Cancer Therapeutics*. **2011**, 57.

T. Hudlicky, E. E. Boros, C. H. Boros. *Synthesis*. **1992**, 174.

T. Hudlicky, X. R. Tian, K. Konigsberger, R. Maurya, J. Rouden, B. Fan. *Journal of the American Chemical Society*. **1996**, 10752.

T. Hudlicky, X. Tian, K. Kónigsberger, J. Rouden. *The Journal of Organic Chemistry*. **1994**, 4037.

L. Ingrassia, F. Lefranc, V. Mathieu, F. Darro, R. Kiss. *Translational Oncology*. **2008**, 1.

N. Kekre, C. Griffin, J. McNulty, S. Pandey. *Cancer Chemother Pharmacology*. **2005**, 29.

B. H. Lipshutz, J. Kozlowski, R. S. Wilhelm. *Journal of the American Chemical Society*. **1982**, 2305.

D. Ma, C. Pignanelli, D. Tarade, T. Gilbert, M. Noel, F. Mansour, D. Adams, K. Dowhayko, S. Stokes, S. Vshyvenko, T. Hudlicky, J. McNulty, S. Pandey, S. *Pandey. Scientific Reports*. **2017**, 42957.

M. Mandel, T. Hudlicky. *The Journal of Organic Chemistry*. **1993**, 2331.

M. Manpadi, A. Kornienko. *Organic Preparations and Procedures International*. **2008**, 107.

J. A. Marshall. *Chemical Reviews*. **1989**, 1503.

A. McLachlan, N. Kekre, J. McNulty, S. Pandey. *Apoptosis*. **2005**, 619.

S. Pandey, N. Kekre, J. Naden, J. McNulty. *BioTechnologia*. **2005**, 279.

S. Pandey, J. McNulty. *International Journal of Oncology*. **2011**, 1549.

G. R. Pettit, V. Gaddamidi, G. M. Cragg, D. L. Herald, Y. Sagawa. *Journal of the Chemical Society, Chemical Communications*. **1984,** 1693.

G. R. Pettit, V. Gaddamidi, D. L. Herald, S. B. Singh, G. M. Cragg, J. M. Schmidt, F. E. Boettner, M. Williams, Y. Sagawa. *Journal of Natural Products*. **1986**, 995.

G. R. Pettit, R. A. Backhaus, M. R. Boyd, A. J. W. Meerow. *Natural Products*. **1993**, 1682.

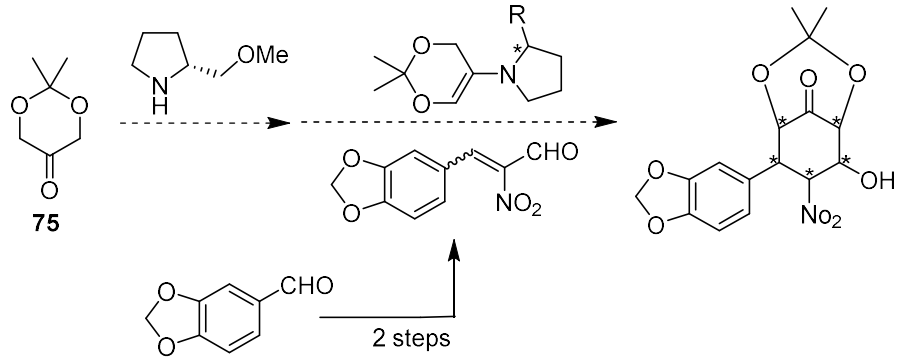
X. R. Tian, T. Hudlicky, K. Konigsberger. *Journal of the American Chemical Society*. **1995**, 3644.

Y. Yamada, T. Yamamoto, M. Okawara. *Chemistry Letters*. **1975**, 361.

1.4DD Selected Total Synthesis of (+)-Pancratistatin

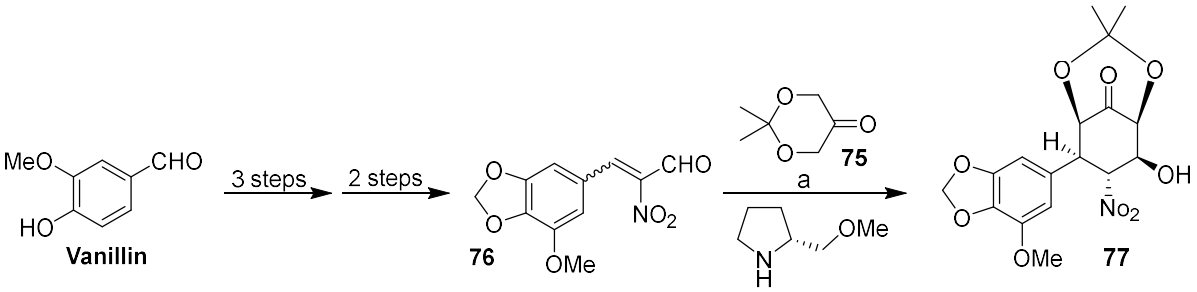
Synthesis strategies towards complex natural products commonly face many obstacles along the way. This was explicit in the previous semi-synthesis which required many changes lowering the overall efficacy after failing to garner efficient alternative routes to rectify their issues. New reactions that have the potential to build complex chiral structures from simple achiral starting materials in an enantioselective fashion would help alleviate this distress from a synthetic standpoint. After isolating hundreds of alkaloids from the Amaryllidaceae family, such routes would enable future syntheses towards enantioselective pathways to their corresponding aminocyclitol cores. Taking into consideration the minute quantities each alkaloid has from their plant sources (as previously explained in the earlier sections in this chapter), success in this regard would lead to increased overall yields and help fill libraries of unnatural alkaloids trying to increase a molecule's bioavailability. Alonso et. al. in 2012 set this plan into action by reporting an enantioselective annulation of β-(hetero)aryl-α-nitro-α,β-enals with enamines generated using pyrrolidine catalysts to form protected nitrocyclohexitols. The process can be used towards a wide-variety of natural products generating five new stereocenters from two

asymmetric non-chiral precursors (scheme 11 and 12) (Cagide-Fagín, Nieto-García, Lago-Santome, & Alonso, 2012).



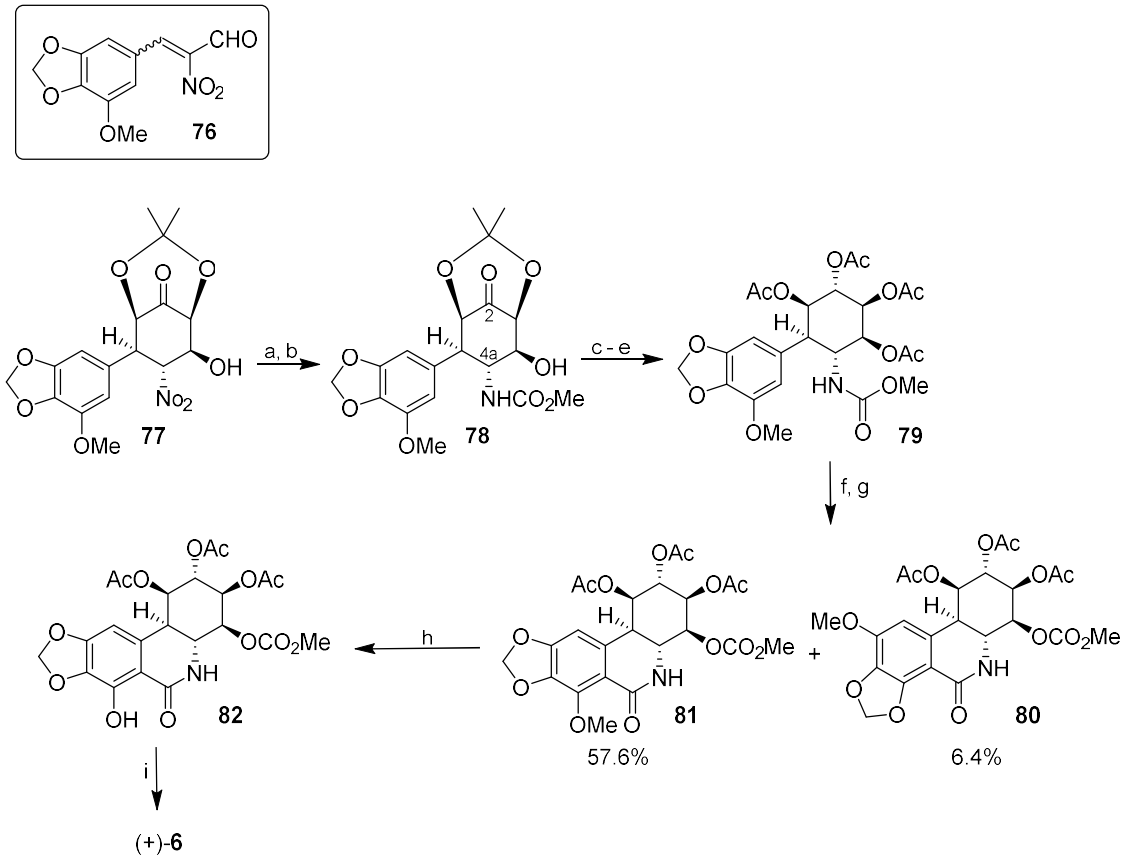
Scheme 11. Enantioselective annulations of nitroenals.

The steps shown in scheme 11 explain how compound **75** and catalyst (R)-2-(methoxymethyl)pyrrolidine being commercially available, and a β-(hetero)aryl-α-nitro-α,β-enal requiring only two steps to be synthesized from commercial aldehydes and 2-nitroethanol, how aminocyclitol cores can be isolated in a robust fashion with five new stereocenters. Such routes serve natural product syntheses with there being more than 20, 000 anti-infective, antitumor, immune, anti-inflammatory, peptidomimetric, anti-ulcer, dermatological, and nervous system agents having a cyclohexane ring with (at least) one nitrogen and three oxygens attached being reported in more than 82, 000 references (Cagide-Fagín, Nieto-García, Lago-Santome, & Alonso, 2012).



Scheme 12. Transformation of vanillin to **76** had been optimized from previous studies with 57% over three steps towards 5-methoxypiperonal, and an ensuing 51% over two steps. Reaction conditions: a) DMF, 75%, >99% *ee*.

The subsequent cyclization moved very effectively over three steps proceeding by an enantioselective annulation of nitroenal **76** with 2,2-dimethyl-1,3-dioxan-5-one. The sequence proceeds via an enamine intermediate generated by (R)-2-(methoxymethyl)pyrrolidine as the chiral catalyst with 2,2-dimethyl-1,3-dioxan-5-one towards the convergent synthesis of the aminocyclitol **77** without incident. This strategy went into effect for the total synthesis of (+)-**6**, demonstrating the versatility for such a development.



Scheme 13. Alonso's total synthesis of (+)-**6**: a) H_4NCOOH, 10% Pd/C, MeOH; b) $ClCO_2Me$, DMAP, CH_2Cl_2, 90% over two steps; c) Dowex 50WX, MeOH; d) $Na(AcO)_3BH$, $C_2H_4Cl_2$/THF; e) Ac_2O, DMAP, Et_3N, CH_2Cl_2, 87% over three steps; f) Tf_2O, DMAP, CH_2Cl_2, O °C; g) HCl, 1,4-dioxane, rt, 64% over two steps; h) BBr_3, CH_2Cl_2, -78 °C, 50%; i) NaOMe, MeOH/THF, 86%.

The synthesis begins with the preparation of **76** that had been previously optimized from vanillin (three steps, 57% yield) (Boisnard & Carbonnelle, 2001; Ellis & Lenger, 1998; McKittrick & Stevenson, 1984), and subsequently enal (two steps, 51%) (Martínez-Bescos, et al., 2008). Annulation followed between dioxanone **75** with nitroenal **76** using (R)-2-(methoxymethyl)-pyrrolidine assembling ring-C in an enantioselective manner with all carbons having the correct stereochemistry of the target molecule with a 38% yield and 75% *ee* (>99% *ee* after crystallization in diethyl ether). Excluding DMSO (49% *ee*),

halogenated, protic, and other aprotic solvents performed similarly with respect to the enantioselectivity (70-75% *ee*) when compared to DMF and acetonitrile. Acetonitrile and DMF provided the highest yielding annulations, so DMF was chosen to be used. For the conversion of nitrocyclitol **77** into **79**, the **C4a** nitro group was reduced to the amine, and protected by acylation to **78**. The following three steps were done in sequence involving the stereoselective reduction of the C2 keto group and changing the protecting group pattern to make it more compatible for the following step towards the electrophilic aromatic substitution of **79**. Treatment with triflic anhydride and 4-dimethylaminopyridine (DMAP) at 0 °C, followed by acidic hydrolysis of the iminoether intermediate provided lactam **81**, with its regioisomer **80** in a 9:1 ratio (**81**/**80**) and a 64% yield for the formation of ring-B. The synthesis was completed following two deprotection steps affording (+)-**6**.

1.4DD.1 References

S. Boisnard, A. C. Carbonnelle. *Organic Letters*. **2001**, 2061.

F. Cagide-Fagín, O. Nieto-García, H. Lago-Santome, R. Alonso. *The Journal of Organic Chemistry*. **2012**, 11377.

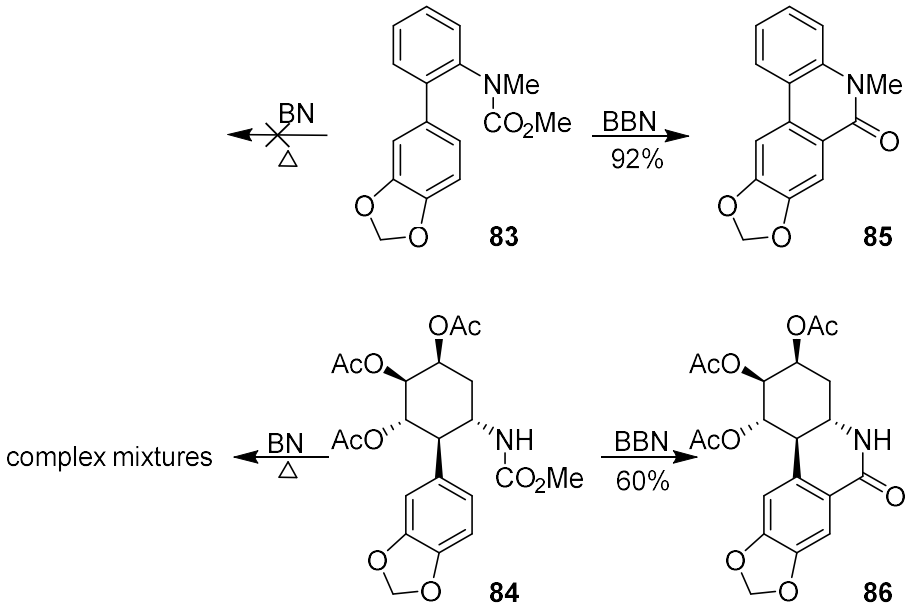
J. E. Ellis, S.R.P.D. Lenger. *Synthetic Communications*. **1998**, 1517.

P. Martínez-Bescos, F. Cagide-Fagín, L. F. Roa, J. C. Ortiz-Lara, K. Kierus, L. Ozores-M. M. Viturro, R. Fernández-González, R. Alonso. *The Journal of Organic Chemistry*. **2008**, 3745.

1.5 Banwell's modification of the Bischler-Napieralski reaction

In 1893 two scientists by the names of Bischler and Napieralski first reported a new method for the cyclization of β-arylethylamides and β-arylethylcarbamates by intramolecular electrophilic aromatic substitution, using phosphorus oxychloride ($POCl_3$) as a condensing agent now known as the Bischler-Napieralski (BN) reaction (Bischler & Napieralki, 1893). Since their discovery, the BN cyclization of β-phenethylamides have delivered an effective method for the construction of 3,4-dihydroisoquinolines and related heterocyclic molecules (Fodor & Nagubandi, 1980). Like many chemical processes, such conversions required both an increase in temperatures and use of aggressive reagents. Consequently, substrates with sensitive functional groups were often unable to be used towards such harsh reaction conditions. Although this was the case, the reaction was used for the synthesis of naturally occurring alkaloids and thiophene derivatives. From increased attention by the chemistry community, Banwell undertook research specifically directed towards the synthesis of alkaloids and reported a modified approach over the acidic conditions in combination with high temperatures and a dehydrating agent (Banwell & Wu, 1994; Banwell & Cowden, 1994; Banwell, Cowden, & Ho, 1994; Banwell, Cowden, & Gable, 1994; Lewis, 1995). Following the additional studies related to the reaction conditions, Banwell et. al. changed the reaction medium to a basic medium and saw great success. The reaction is now known as the Banwell modified Bischler-Napieralski (BBN) reaction and uses a combination of trifluoromethanesulfonic (triflic) anhydride (Tf_2O) and DMAP for the cyclization of both β-phenethylcarbamates and β-phenethylamides at or below room temperature. Triflic anhydride has been used in many syntheses as an

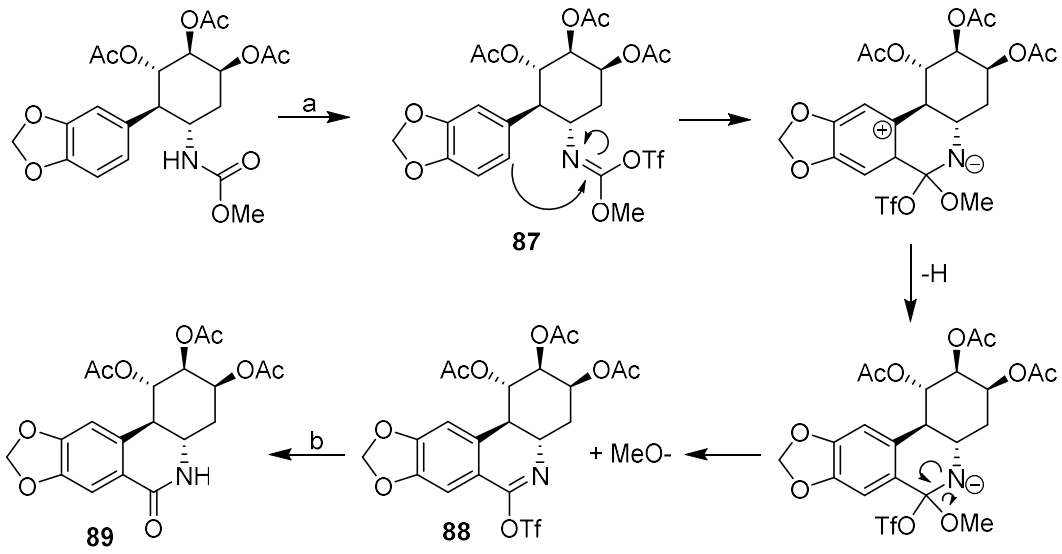
electrophilic reagent for the activation of a wide range of functional groups and is utilized in this reaction because of the high efficacy it brings with activating carbamates and amides (Stang, Hanack, & Subramanian, 1872; Baraznenok & Nenajdenko, 2000). The reaction continues via the cyclodehydration of the carbonyl group. The refined BBN was performed in the synthesis of two Amaryllidaceae alkaloids lycoricidine and pancratistatin in 1994 (Banwell, Cowden, & Gable, 1994). During related studies, it was shown that while using the BBN reaction conditions, successful cyclizations were achieved while $POCl_3$ (BN conditions) failed to produce any cyclized products for some of the investigated compounds (scheme 14) (Banwell, et al., 1995).



Scheme 14. Comparison between the intramolecular cyclization reaction of BN and BBN pathways showing how Banwell's modification has increased the reaction's efficacy.

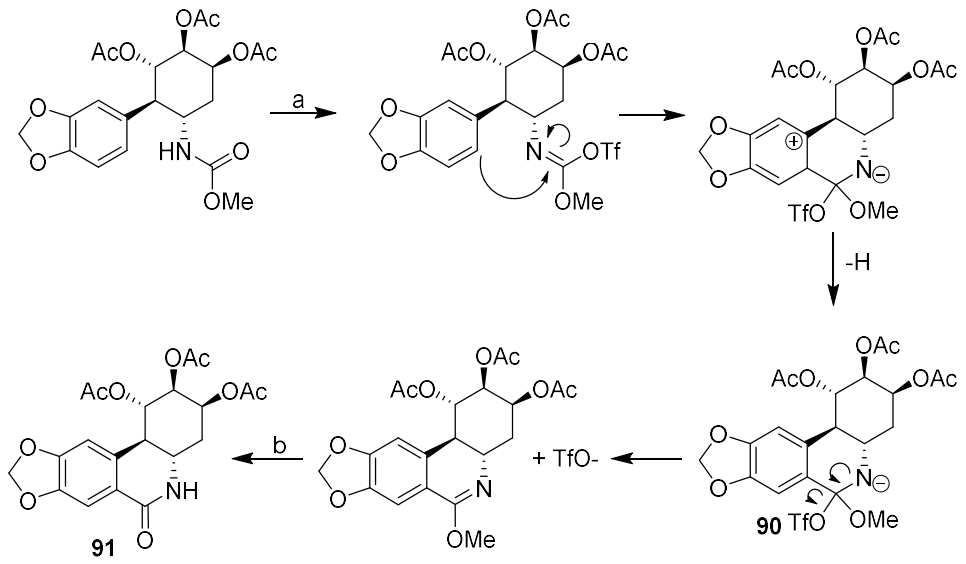
The modifications improved the reaction scope and is highlighted in scheme 14 where no reaction took place in the first reaction after allowing 13 hours to react, in addition to

increasing the temperature to 200 °C. This same reaction when performed following BBN reaction conditions provided a 92% yield, demonstrating increased utility for such a procedure. Treatment of **83** with Tf_2O-DMAP (5:3 molar ratio with respect to the substrate) at 0 - 15 °C for 10 hours isolated, after aqueous work-up, *N*-methylcrinasiadine **85** in a 92% yield (Banwell & Cowden, 1994; Mondon & Krohn, 1972; Zee-Cheng, Yan, & Cheng, 1978). The modifications leading to the successful cyclizations were found to be very reliant on the molar ratios of Tf_2O and DMAP being ca. 5 molar equivalents of Tf_2O and ca. 3 molar equivalents of DMAP. When attempting to see similar results with $POCl_3$ only extensive decomposition was observed. After identifying the optimal reaction conditions, mechanistic insight was pursued to determine the reactive intermediates throughout the cyclization. A reactive carbonimidic intermediate **87** is generated, which then cyclizes via electrophilic aromatic substitution producing ring-B. Elimination, and regeneration of aromaticity of the resulting species would lead to the formation of **89**.



Scheme 15. Proposed sequence for the BBN reaction: a) Tf_2O/DMAP (5:3), CH_2Cl_2, 0 °C – rt; b) 1M HCl, 60% over two steps.

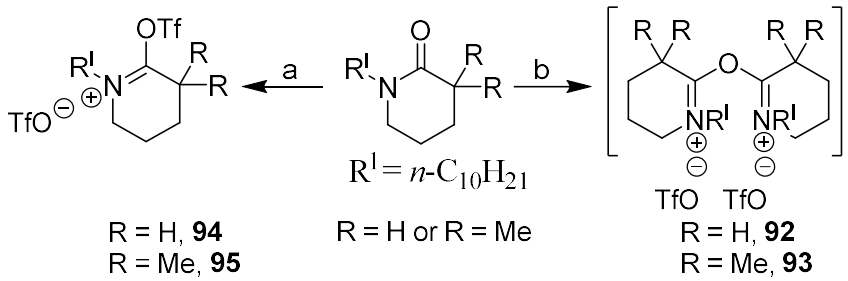
After continued studies by Kim (Hwang, Kim, & Kim, 2012), an alternate elimination pathway (scheme 16) was proposed by compound **90** to be the final step prior to aqueous work-up with the addition of 1M HCl isolating **91**.



Scheme 16. Alternative elimination mechanism for the BBN proposed by Kim: a) Tf_2O/DMAP (5:3), CH_2Cl_2, 0 °C – rt; b) 1M HCl, 60% over two steps.

The combination of Tf_2O and DMAP provided effective intramolecular cyclizations under very mild conditions.

More recently, Kolby et. al. reported a detailed analysis monitoring the reaction between the electrophilic amide activation of tertiary lactams (White, Mewald, & Movassaghi, 2014). Throughout their studies they identified another more stable intermediate for tertiary lactams as being a diiminium ether **92**/**93** (scheme 17).



Scheme 17. Lactam activation via electrophilic triflic anhydride, forming two reactive intermediates of sulfonyl iminium ions **94**/**95** and diiminium ethers **92**/**93**: a) Tf_2O (1 eq.), CH_2Cl_2, −78 °C; b) Tf_2O (0.5 eq.), CH_2Cl_2, −78 °C.

The previously established lactams react with the electrophilic triflic anhydride to form compounds **94**/**95**, but after further analysis through in situ IR monitoring, they identified the formation of a diiminium ether **92**/**93**. Their studies communicated that depending on the tertiary lactam, increased heating was required to see the formation of the corresponding diiminium ether formation, but its specific IR intensities were observed, nonetheless.

1.5.1 References

M. G. Banwell, C. J. Cowden, I. C. S. Ho. *Journal of Natural Products*. **1994,** 1746.

M. G. Banwell, A. Cowden. *Australian Journal of Chemistry*. **1994**, 2235.

M. G. Banwell, B. D. Bissett, S. Busato, C. J. Cowden, D. C. R. Hockless, J. W. Holman, M. G. Banwell, C. J. Cowden, R. W. Gable. *Journal of the Chemical Society, Perkin Transactions*. **1994**, 3515.

I. L. Baraznenok, V. G. Nenajdenko, E. S. Balenkova. *Tetrahedron*. **2000**, *56*, 3077.

A. Bischler, B. Napieralki. *Ber*. **1983**, 1903.

M. G. Banwell, A. Wu. *Journal of the Chemical Society, Perkin Transactions*. **1994**, 2671.

G. Fodor, S. Nagubandi. *Tetrahedron*. **1980**, 1279.

S. Hwang, D. Kim, S. Kim. *Chemistry - A European Journal - Chemistry Europe*. **2012**, 9977.

J. R. Lewis. *Natural Product Reports*. **1995**, 339.

A. Mondon, K. Krohn. *Chemische Berichte*. **1972**, 3726.

R. Read, A. W. Wub. *Journal of the Chemical Society, Chemical Communications*. **1995**, 2551.

P. J. Stang, L. R. Hanack, L. R. Subramanian. *Synthesis*. **1982**, 85.

K. White, M. Mewald, M. Movassaghi. *The Journal of Organic Chemistry*. **2014**, *16*, 358.

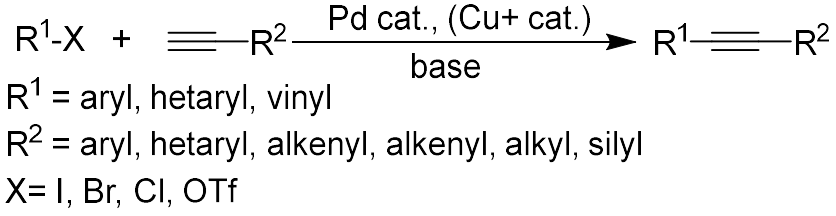
R. K. Y. Zee-Cheng, S. J. Yan, C. C. Cheng. *Journal of Medicinal Chemistry*. **1979**, 199.

1.6 Copper-Cocatalyzed Sonogashira Reaction

Formation of new carbon-carbon (C-C) bonds play a pivotal role throughout the chemistry community and are industrially important. Transition metal catalyzed C-C coupling is frequently seen in pharmaceuticals, agrochemicals, and industrially important compounds as they commonly encompass aromatic and heteroaromatic units in their structures (Busch, Wodrich, & Corminboeuf, 2017; Trost & Fleming, 1991; Doucet & Hierso, 2007). When comparing to conventional non-catalytic processes, these coupling reactions permit the use of a much broader substrate scope towards the isolation of more diverse molecules. Palladium incorporated catalyst systems are customarily useful for such approaches with their tunability, selectivity, and reactivities they are associated with. Such

use often delivers a high turnover number and turnover frequency allowing for minimal catalyst loadings (Veerakumar, Thanasekaran, Lu, Lin, & Rajagopal, 2017). From the array of transition metal catalyzed cross-coupling reactions reported today, they can easily be considered a cornerstone in organic synthesis (Cornils & Herrmann, 1996; Negishi & de Meijere, 2002; Miyaura, 2000; Diederich & de Meijere, 2004; Beller & Bolm, 2004; Tsuji, 1995; Zapf. & Beller, 2002; Tucker & de Vries, 2002; van de Weghe, 2005). The Sonogashira cross-coupling reaction has been employed in many areas, exhibiting its effectiveness in the formation of new C-C bonds. Such value can be rationalized through the mild reaction conditions used during these types of procedures including room temperature, aqueous media, and use of mild bases, which has led to the synthesis of a large assortment of complex target molecules. The Sonogashira cross-coupling reaction is one of the most valued and widely utilized routes towards sp^2-sp C-C bond formations in organic synthesis over the last 50 years (Sonogashira, Tohda, & Hagihara, 1975; Sonogashira K. , 2002; Chinchilla & Nájera, 2007; Chinchilla & Najera, 2011). Its application covers many fields including pharmaceuticals, natural products, organic materials, and nanomaterials (Shang, Wang, Sun, Dai, & Yu, 2014; Mahapatra & Carter, 2013; Doucet & Hierso, 2007; Nájera, Gil-Moltó, Karlström, & Falvello, 2003; Chinchilla & Najera, 2011). Palladium catalyzed cross-coupling reactions can be seen across many fields in synthetic chemistry, but the palladium catalyzed Sonogashira reaction has pushed itself to the top of the list for preparing arylacetylenes and conjugated enynes (Chinchilla & Nájera, Chemical Reviews, 2007; Chinchilla & Najera, 2011). Its efficacy is carried by the generation of high yields, while tolerant towards a variety of functional groups. In 1975, three independent

contributors reported the alkynylation reaction of aryl halides using aromatic acetylenes from Heck (Dieck & Heck, 1975), Cassar (Cassar, 1975), as well as Sonogashira, Tohda and Hagihara (Sonogashira, Tohda, & Hagihara, 1975). The first two reports required temperatures around 100 °C, with the addition of organic or inorganic bases for the successful cross-coupling reactions with palladium. Heck's procedure was based on the known Mizoroki-Heck reaction (Knowlesa & Whiting, 2007; Nakashima, Hirata, Sheppard, & Nishikata, 2020; Mizoroki, Mori, & Ozaki, 1971) which consisted of performing the coupling while making use of a phosphane-palladium complex as the catalyst and triethylamine or piperidine as the corresponding base and solvent. While for Cassar's procedure, he utilized a phosphane-palladium catalyst with sodium methoxide as the base and DMF as the solvent. Sonogashira and Hagihara showed how with the addition of a catalytic amount of copper(I) iodide the reaction would dramatically improve the rate of alkynylation while at room temperature. The procedure has since become the generally accepted method involving aryl or alkenyl halides or triflates (scheme 18) and was related to the previously reported coupling among copper acetylides and phenyl or vinyl halides. The reaction was called the Stephens-Castro reaction (Stephens & Castro, 1963).



Scheme 18. Generalized conditions for the Sonogashira reaction.

The general reactivity order of sp^2 species is vinyl iodide ≥ vinyl triflate > vinyl bromide > vinyl chloride > aryl iodide > aryl triflate ≥ aryl bromide ≫ aryl chloride, making processes run smoothly when the more expensive but unstable starting materials are used. Procedures to navigate around this, can be performed by functionalizing with activating substituents on the aryl ring, reducing the electron density between the C-X bond (fig. 9), and in doing so allowing an easier oxidative addition to occur (Littke & Fu, 2002).

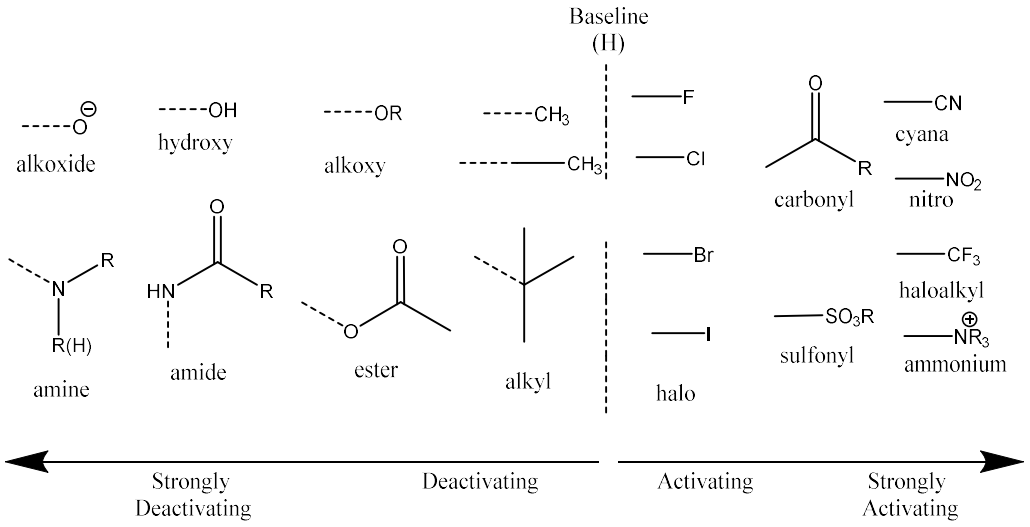
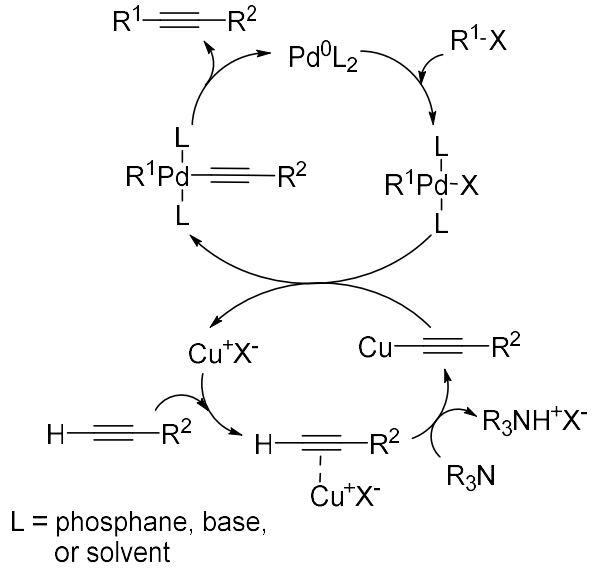


Figure 9. Reactivity order of sp^2 species.

Additional studies related to the first step of the palladium cycle have also shown that by exchanging ligands such as triphenylphosphine with more electron rich phosphane ligands leads to an easier oxidative addition as well. These studies support the successful Sonogashira cross-coupling reactions when using low reacting-bromo or chloro arenes. It has been demonstrated that by using ligands possessing high steric constraints promotes an easier dissociation from the Pd^0L_2 resting state, necessary before oxidative insertion (Barrios-Landeros & Hartwig, 2005). Reports with the combination of $Pd(PhCN)_2Cl_2$, CuI,

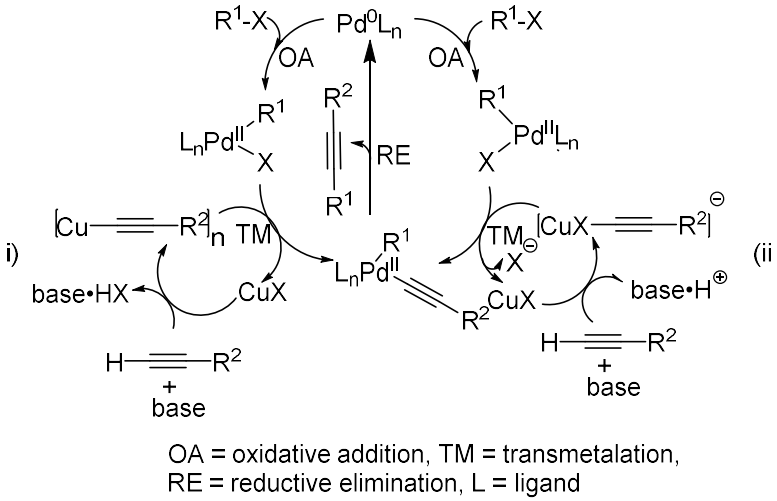
and P(*t*-Bu)$_3$ for the Sonogashira reaction of aryl bromides at room temperature at a 3% palladium loading have illustrated these benefits (Hundertmark, Littke, Buchwald, & Fu, 2000). The copper-cocatalyzed Sonogashira reaction is believed to take place through two independent catalytic cycles (scheme 19). The tertiary amine represents the base, but this can be performed with alternative amines or inorganic bases which perform similarly.



Scheme 19. Two independent Copper-cocatalyzed Sonogashira cycles with palladium proceeding along synergistically operating catalytic cycles.

Palladium catalysis starts off with the oxidative addition between the R^1-X bond where R^1 can be aryl, hetaryl, or vinyl while 'X' is often I, Br, Cl, or OTf but other examples such as OTs are used. The copper-cycle then generates the reactive species to perform the trans-metalation but is not fully understood how mechanistically it takes place. A copper salt is generally used in the reaction, and the base has been thought to abstract the acetylic proton from the alkyne, thus forming the copper acetylide species, although there is no evidence for this taking place (Bertus, Fe´court, Bauder, & Pale, 2004). Debate has risen with the

knowledge of the bases used not being strong enough to deprotonate the alkyne. But this has been retorted by claims involving a π-alkyne-copper complex being responsible for increasing the acidity of the alkyne proton, and enabling the formation of the copper acetylide species (Bertus, Fe´court, Bauder, & Pale, 2004). With many still trying to uncover the exact mechanism, recent NMR studies with silver have shown that the π-alkyne-silver complexes are formed after the generation of the silver acetylides when performing a silver-cocatalyzed Sonogashira reaction (Le´tinois-Halbes, Pale, & Berger, 2005). These findings could be extrapolated to the reactions with copper, since the assumed in situ formation of a copper acetylide intermediate has never been proven, leaving questions still about the nature of the real catalyst involved (Stambuli, Buhl, & Hartwig, 2002). Other reports (Amatore & Jutand, 2000; Grosshenny, Romero, & Ziessel, 1997) have illustrated that in the presence of anions and halides, anionic palladium species have been observed insinuating the formation of anionic palladium species as the real reason for the catalytic processes taking place. For example, it is known that $Pd^0(PPh_3)_2$ does not exist in solution when in the presence of halide anions because they coordinate to the palladium(0) center and form the anionic species (Amatore & Jutand, 2000; Grosshenny, Romero, & Ziessel, 1997). During the past decade many computational studies have been undertaken trying to unravel the precise mechanism (Xue & Lin, 2010; Goossen, Koley, Hermann, & Thiel, 2005), and a recent one by Wang et. al. proposed an alternative anionic mechanism as previously mentioned (scheme 20). Alkynes bearing electron withdrawing groups were more in favor of going through the anionic mechanism than their electron donating counterparts (Ljungdahl, Bennur, Dallas, Emtenas, & Martensson, 2008).

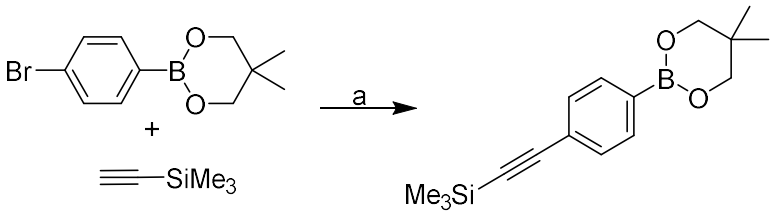


Scheme 20. Copper-cocatalyzed Sonogashira reaction pathways: *i*) neutral mechanism; *ii*) anionic mechanism.

The study reports an iodide ion (I^-) coordinated copper acetylide being formed in situ from acetylene in the presence of base and copper iodide. This reactive species would then be held accountable for the transmetalation with the Pd(II) species (Wang, Song, Qu, & Luo, 2017). Commonly seen catalysts for such procedures are triphenylphospane related complexes such as $Pd(PPh_3)_4$, and the more stable and soluble $Pd(PPh_3)_2Cl_2$, while other bidentate ligands include $Pd(dppe)Cl_2$, $Pd(dppp)Cl_2$, or $Pd(dppf)Cl_2$.

With many Sonogashira reactions being set-up using polar solvents (microwave active), microwave heating has been used in the homogenous-phase reactions following typical reaction conditions for the successful coupling of different iodides, triflates, bromides, and 2-chloropyridine with trimethylsilylacetylene (TMSA) in only 5 - 25 minutes (Erdélyi & Gogoll, 2001; Erdélyi, Langer, Karlén, & Gogoll, 2002; Miljanić, Vollhardt, & Withener, 2003; Han, Castro, & Burgess, 2003; Gong & He, 2004; Kuang, Yang, Senboku, & Tokuda, 2005). An example of this application was reported for the

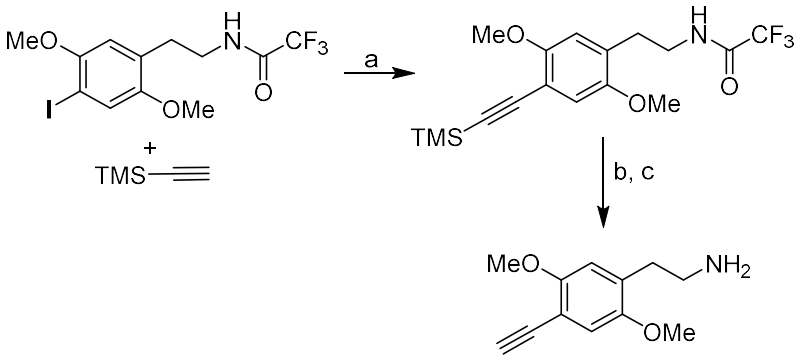
bromoaryl boronate being coupled to TMSA in almost a quantitative yield in 25 minutes (scheme 21) (Zheng, Reid, Lin, & Wang, 2006). When comparing this result with two more traditional set-ups in refluxing DMF for 6 hours and another heating at 130 °C for 30 minutes, they afforded no reaction and only a 60% yield respectfully.



Scheme 21. Sonogashira reaction conditions inside a microwave: a) $PdCl_2(PPh_3)_2$ (5 mol%), CuI (5 mol%), PPh_3, Et_2NH, DMF, MW, 120 °C, 98%.

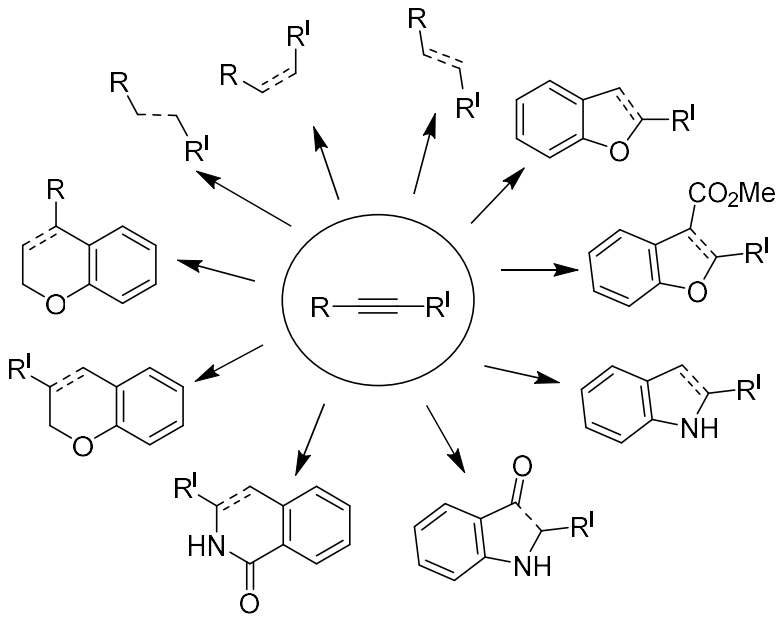
Recent examples showing reactions between aryl iodides or bromides and arylated (Collins, et al., 2004; Jessen & Pedersen, 2004; Chen, Rao, & Knaus, 2005; Kauch, Snieckus, & Hoppe, 2005; Anana, Rao, Chen, & Knaus, 2006), alkylated (Xu, Li, & Ma, 2003; Lane & Halcomb, 2005; Lo, Neumann, Nagayama, Perlstein, & Schreiber, 2004), or conjugated alkenylated acetylenes (Hoshi, Nakayabu, & Shirakawa, 1991; Fouad, et al., 2005), even when the aryl halide is supported on a solid (Kashiwagi, Chiba, Ikezoe, & Anzai, 2004; Knepper, Vanderheiden, & Brase, 2006), or PEG-soluble phase (Xia & Wang, 2003), can be found. The synthesis towards terminal alkynes using palladium is a very efficient route to isolate final products of interest by themselves, or for subsequent couplings to diarylalkynes with recurrent applications for such things as building blocks in electrooptical devices and material sciences (Nagy, Novak, & Kotschy, 2005) towards the synthesis of aromatics, heterocycles, alkenes, 1,3 diynes, ynones and allyl ethers (Chinchilla & Najera,

2013). An example of this can be seen in the synthesis of phenethylamine (PEA) (scheme 22), which had been undertaken for SAR investigations.



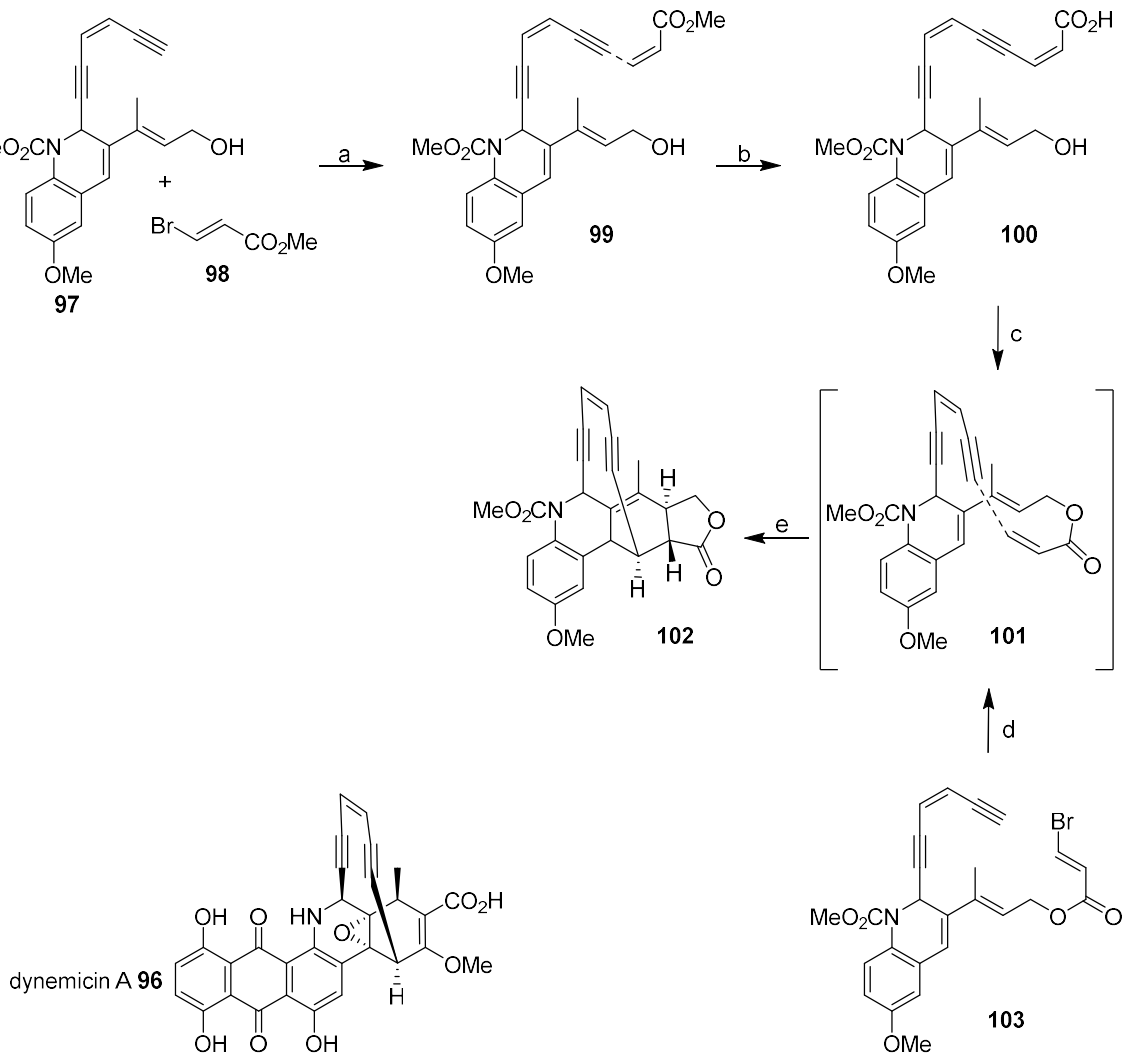
Scheme 22. General Sonogashira reaction conditions, with a subsequent deprotection step for the synthesis of a monoamine alkaloid PEA: a) $Pd(PPh_3)_2Cl_2$ (2 mol%), 97%; b) TBAB, THF, rt; c) aq. NaOH, MeOH, 88% over two steps.

A typical example of the Sonogashira reaction is seen with the synthesis of PEA (Traschsel, 2003). Production of arylalkynes and conjugated enynes have further application towards the formation of cyclic or polycyclic molecules, as well as being subsequently reduced providing extra incentives for its applications (scheme 23).



Scheme 23. Transformations of arylalkynes and conjugated enynes generated by Sonogashira coupling (Wang & Gao, 2014).

A synthesis featuring these applications include the likes of Schreiber et. al. towards the total synthesis of dynemicin A (**96**) (Taunton, Wood, & Schreiber, 1993; Wood, et al., 1992; Chikashita, Porco Jr., Stout, Clardy, & Schreiber, 1991; Porco Jr., Schoenen, Stout, & Schreiber, 1990), a potent enediyne containing antibiotic. The compound being a hybrid antitumor antibiotic has enediyne and anthracycline cores, which mediates strand breakage of DNA, that is enhanced in the presence of NADPH or dithiothreitol.



Scheme 24. Sonogashira coupling followed by the transannular Diels–Alder reaction to construct the core skeleton of dynemicin A (**96**). Reaction conditions: a) $Pd(PPh_3)_4$, CuI, toluene, 25 °C; b) LiOH, THF/H_2O, 65% over two steps; c) 2,4,6-$Cl_3C_6H_2COCl$, DMAP, toluene, 50%; d) $Pd(PPh_3)_4$, CuI, toluene, 25 °C, 25%; e) transannular Diels-Alder reaction.

Their work presented in scheme 24 included two separate experiments involving Sonogashira cross-coupling conditions. Coupling between enediyne **97** and vinyl bromide **98** in the presence of CuI and $Pd(PPH_3)_4$ produced polyunsaturated ester **99**, which was then hydrolyzed under alkaline conditions giving the corresponding acid **100**. Subsequent Yamaguchi macrocyclization led to the isolation of pentacyclic **102** in a 50% yield. The

proposed mechanism involves an intermediate macrocyclic ring **99** that is transformed via the spontaneous Diels-Alder reaction. Their studies also demonstrated how an intramolecular Sonogashira coupling of **103** in the presence of $Pd(PPh_3)_4$ and CuI had as well produced **102** in a 25% yield. The impressive one-pot set-up was well recognized because of the efficient production of three rings with a highly strained enediyne motif with four contiguous stereocenters.

When mapping out a synthesis for a target molecule, including steps with feasible aryl halides at times can involve harsh reaction conditions, as well as increased waste production (Stang, Hanack, & Subramanian, 1982). From the extensive studies involving compounds containing hydroxyl groups, phenol-derived sulfonated hydroxyl groups have been investigated showing how well versatile they are as alternatives to aryl halides. Aryl and alkenyl sulfonates use have shown an increase in the past few decades, exhibiting them as valuable coupling partners for successful cross-coupling reactions. Their syntheses are performed with ease from phenols and carbonyl enolates providing reliant routes towards aryl and 1-alkenyl electrophiles (Stang, Hanack, & Subramanian, 1982). Such reactive species provide significant utility for the synthetic chemist regarding the wide-ranging phenolic (Huang & T., 2010; Tungmunnithum, Thongboonyou, Pholboon, & Yangsabai, 2018; Kumar & Goel, 2019) and carbonyl groups existing ubiquitously in nature allowing for the preparation of a broad range of electrophilic sites towards pharmaceutically attractive building blocks (figure 10).

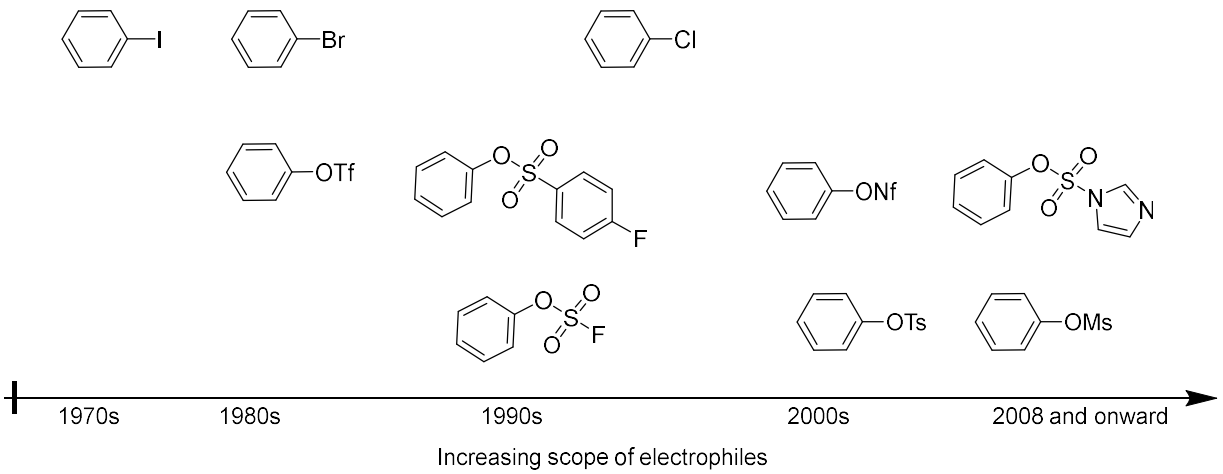


Figure 10. The development of aryl halides and sulfonate electrophiles involved in cross-coupling reactions (So & Kwong, 2011).

One of the earliest studies involving an aryl sulfonate involved a cross-coupling reaction with an aryl triflate in 1987 (Echavarren & Stille, 1987). Alternatives to triflates include aryl fluorosulfonates (Roth & Fuller, 1991), aryl *p*-fluorobenzenesulfonates (Badone, Cecchi, & Guzzi, 1992) and aryl nonaflates (Anderson, Mendez-Perez, Priego, & Buchwald, 2003). Aryl triflates are not easy to handle being very labile compounds easily hydrolyzed and degrading gradually at room temperature. In contrast to these characteristics, aryl tosylates and enol tosylates are less prone to degradation being notably more stable, while easily being prepared by inexpensive tosyl chloride in many syntheses (Gelman G. B., 2003; R'Kyek, et al., 2012; Gelman, L., & Buchwald, 2003; Fu, Zhang, Yin, & Schumacher, 2002; Gebauer & Bruckner, 2000; Hayford, et al., 2005; Steinhuebel, Baxter, Palucki, & Davies, 2005). Other notable syntheses that incorporated the Sonogashira reaction are shown in figure 11 (Kaur, et al., 2009; Yepremyan & Minehan, 2012; Enders, Fronert, Bisschops, & Boeck, 2012; Yuen & Brimble, 2012; Tietze, et al.,

2013; Duffey, LeTiran, & Morken, 2003; Maezaki, et al., 2004; Cramer, Laschat, Baro, Schwalbe, & Richter, 2005; Bock, Dehn, & Kirschning, 2008).

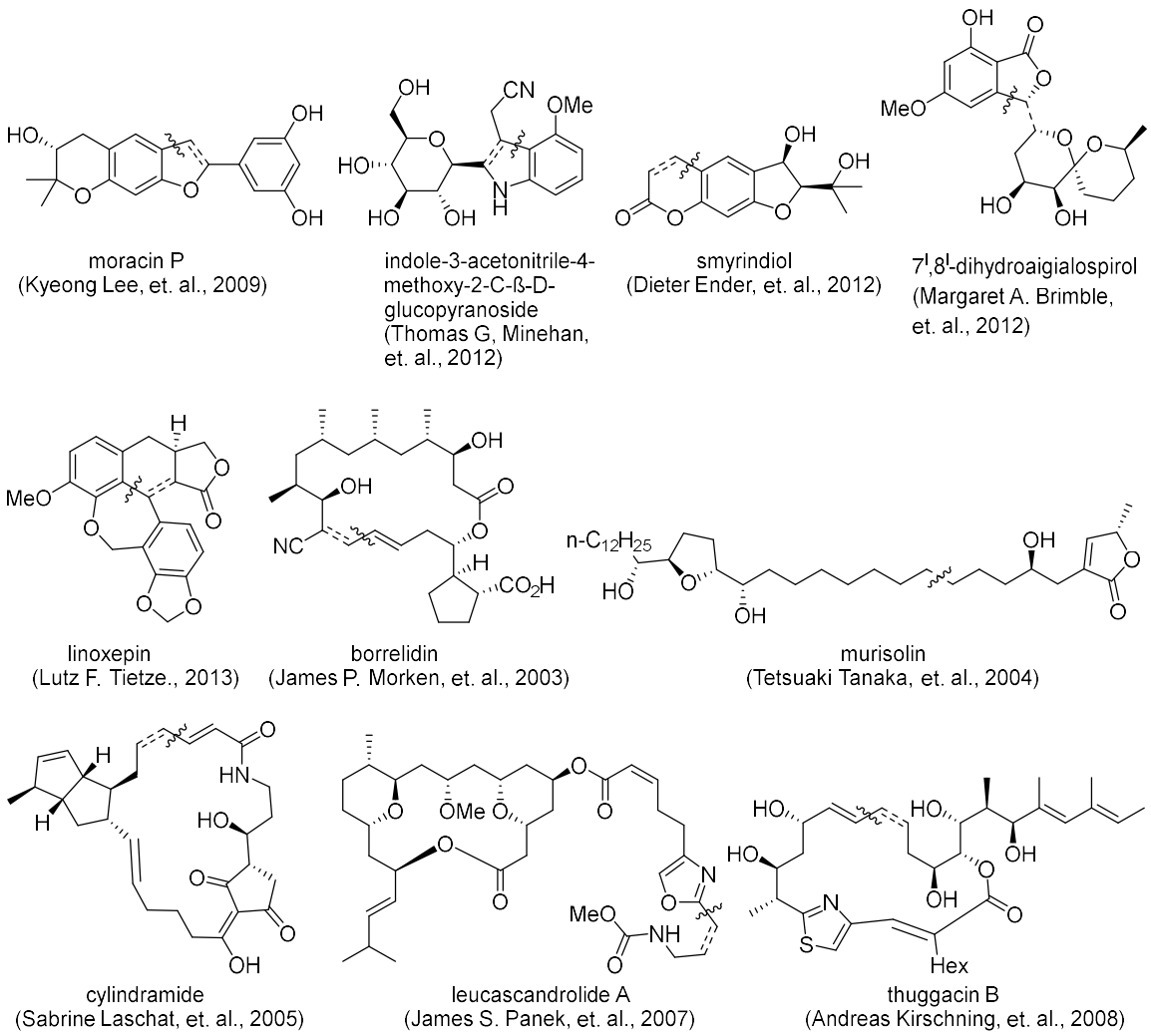


Figure 11. Total syntheses that utilized the copper-cocatalyzed Sonogashira reaction towards their final target molecules.

1.6.1 References

C. Amatore, A. Jutand. *Accounts of Chemical Research*. **2000**, 314.

R. Anana, P. N. P. Rao, Q. H. Chen, E. Knaus. *Bioorganic & Medicinal Chemistry*. **2006**, 5259.

K. W. Anderson, M. Mendez-Perez, J. Priego, S. L.Buchwald. *The Journal of Organic Chemistry*. **2003**, 9563.

D. Badone, R. Cecchi, U. Guzzi. *The Journal of Organic Chemistry*. **1992**, 6321.

P. Bertus, F. Fe´court, C. Bauder, P. Pale. *New Journal of Chemistry*. **2004**, 12.

M. Bock, R. Dehn, A. Kirschning. *Angewandte Chemie International Edition*. **2008**, 9134.

M. Busch, M. D. Wodrich, C. Corminboeuf. *ACS Catalysis*. **2017**, 5643.

L. Cassar. *Journal of Organometallic Chemistry*. **1975**, 253.

Q. H. Chen, P. N. P. Rao, E. Knaus. *Bioorganic & Medicinal Chemistry Letters*. **2005**, 6425.

H. Chikashita, J. A. Porco Jr., T. J. Stout, J. Clardy, J. L. Schreiber. *The Journal of Organic Chemistry*. **1991**, 1692.

R. Chinchilla, C. Nájera. *Chemical Reviews*. **2007**, 874.

R. Chinchilla, C. Najera. *Chemical Society Reviews*. **2011**, 5084.

R. Chinchilla, C. Najera. *Chemical Society Reviews*. **2013**, 1783.

J. C. Collins, J. M. Burke, P. S. Amith, A. S. Batsanov, J. A. K. Howard, T. B. Marder. *Organic & Biomolecular Chemistry*. **2004**, 3172.

N. Cramer, S. Laschat, A. Baro, H. Schwalbe, C. Richter. *Angewandte Chemie International Edition*. **2005**, 820.

H. Dieck, F. Heck. *Journal of Organometallic Chemistry*. **1975**, 259.

M. O. Duffey, A. LeTiran, J. P. Morken. *Journal of the American Chemical Society*. **2003**, 1458.

A. M. Echavarren, J. K. Stille. *Journal of the American Chemical Society*. **1987**, 5478.

D. Enders, J. Fronert, T. Bisschops, F. Boeck, F. Beilstein. *Journal of Organic Chemistry*. **2012**, 1112.

M. Erdélyi, A. Gogoll. *The Journal of Organic Chemistry*. **2001**, 4165.

M. Erdélyi, V. Langer, A. Karlén, A. Gogoll. *New Journal of Chemistry*. **2002**, 834.

H. Doucet, J. C. Hierso. *Angewandte Chemie International Edition*. **2007**, 834.

F. S. Fouad, J. M. Wright, G. Plourde II, A. D. Purohit, J. K. Wyatt, A. El-Shafey, G. Hynd, C. F. Crasto, Y. Lin, G. B. Jones. *Journal of Organic Chemsitry*. **2005**, 9789.

X. Fu, S. Zhang, J. Yin, D. P. Schumacher. *Tetrahedron Letters*. **2002**, 6673.

O. Gebauer, R. Bruckner. *Synthesis*. **2000**, 588.

G. Gelman, S. Buchwald. *Angewandte Chemie International Edition*. **2003**, 5993.

Y. Gong, W. He. *Heterocycles*. **2004**, 851.

J. L. Goossen, D. Koley, H. L. Hermann, W. Thiel. *Organometallics*. **2005**, 2398.

V. Grosshenny, F. M. Romero, R. Ziessel. *Organic Chemistry Frontiers*. **1997**, 1491.

J. W. Han, J. C. Castro, K. Burgess. *Tetrahedron Letters*. **2003**, 9359.

A. Hayford, J. Kaloko, S. El-Kazaz, G. Bass, C. Harrison, T. Corpew. *Organic Letters*. **2005**, 2671.

M. Hoshi, H. Nakayabu, K. Shirakawa. *Synthesis*. **1991**, 199.

W. Huang, T. Cai. *Nutrition and Cancer*. **2010**, *62*, 1

T. Hundertmark, A. F. Littke, S. L. Buchwald, G. C. Fu. *Organic Letters*. **2000**, 1729.

C. H. Jessen, E. B. Pedersen. *Helvetica Chimica Acta*. **2004**, 2465.

M. Kaur, Y. Xia, Y. L. Jin, N. T. Dat, K. Gajulapati, Y. Choi, Y. S. Hong, J. J. Leea, K. Lee. *Chemical Communications*. **2009**, 1879.

Y. Kashiwagi, S. Chiba, H. Ikezoe, J. Anzai. *Synlett*. **2005**, 2513.

M. Kauch, V. Snieckus, D. Hoppe. *The Journal of Organic Chemistry*. **2005**, 7149.

K. Knepper, S. Vanderheiden, S. Brase. *European Journal of Organic Chemistry*. **2006**, 1886.

J. P. Knowlesa, A. Whiting. *Organic & Biomolecular Chemistry*. **2007**, 31.

C. Kuang, Q. Yang, H. Senboku, M. Tokuda. *Tetrahedron*. **2005**, 4043.

N. Kumar, N. Goel. *Biotechnology Reports*. **2019**, *24*, 1.

O. R'Kyek, N. Halland, A. Lindenschmidt, J. Alonso, P. Lindemann, M. Urmann, M. Nazare. *Chemistry - A European Journal*. **2012**, 9986.

J. W. Lane, R. L. Halcomb. *Organic Letter*. **2005**, 4017.

U. Le´tinois-Halbes, P. Pale, S. Berger. *The Journal of Organic Chemistry*. **2005**, 9185.

A. F. Littke, G. C. Fu. *Angewandte Chemie International Edition*. **2002,** 4176.

T. Ljungdahl, T. Bennur, A. Dallas, H. Emtenas, J. Martensson. *Organometallics*. **2008**, 2490.

M. M. C. Lo, C. S. Neumann, S. Nagayama, E. O. Perlstein, S. L. Schreiber. *Journal of the American Chemical Society*. **2004**, 16077.

N. Maezaki, H. Tominaga, N. Kojima, M. Yanai, D. Urabe, T. Tanaka. *Chemical Communications*. **2004**, 406.

S. Mahapatra, R. G. J. Carter. *Journal of the American Chemical Society*. **2013**, 10792.

O. Sÿ. Miljanić, K. P. C. Vollhardt, G. C. Withener. *Synlett*. **2003**, 29.

T. Mizoroki, K. Mori, A. Ozaki. *Short Communications*. **1971**, 581.

A. Nagy, Z. Novak,A. Kotschy. *Journal of Organometallic Chemistry*. **2005**, 4453.

C. Nájera, J. Gil-Moltó, S. Karlström, L. R. Falvello. *Organic Letters*. **2003**, 1451.

Y. Nakashima, G. Hirata, T. D. Sheppard, T. Nishikata. *Asian Journal of Organic Chemistry*. **2020**, 480.

J. A. Porco Jr., F. J. Schoenen, J. Stout, S. L. Schreiber. *Journal of the American Chemical Society*. **1990**, 7410.

G. P. Roth, C. E. Fuller. *The Journal of Organic Chemistry*. **1991**, 3493.

M. Shang, H. L. Wang, S. Z. Sun, H. X. Dai, J.Q. Yu. *Journal of the American Chemical Society*. **2014**, 11590.

K. Sonogashira. *Journal of Organometallic Chemistry*. **2002**, 46.

K. Sonogashira, Y. Tohda, N. Hagihara. *Tetrahedron Letters*. **1975**, 4467.

J. P. Stambuli, M. Buhl, J. F. Hartwig. *Journal of the American Chemical Society*. **2002**, 9346.

P. J. Stang, M. Hanack, L. R. Subramanian. *Synthesis*. **1982**, 85.

D. Steinhuebel, J. M. Baxter, M. Palucki, I. W. Davies. *The Journal of Organic Chemistry*. **2005**, 10124.

R. D. Stephens, C. E. J. Castro. *The Journal of Organic Chemistry*. **1963**, 2163.

J. Taunton, J. L. Wood, S. L. Schreiber. *Journal of the American Chemical Society*. **1993**, 10378.

L. F. Tietze, S. C. Duefert, J. Clerc, M. Bischoff, C. Maass, D. Stalke. *Angewandte Chemie International Edition*. **2013**, 3191.

D. Traschsel. *Helvetica Chimica Acta*. **2003**, 2754.

B. M. Trost, I. Fleming. *Pergamon - Oxford*. **1991**, 521.

C. E. Tucker, J. G. de Vries. *Topics in Catalysis*. **2002**, 111.

D. Tungmunnithum, A. Thongboonyou, A. Pholboon, A. Yangsabai. *Medicines*. **2018**, *5*, 93.

P. Veerakumar, P. Thanasekaran, K. L. Lu, K. C. Lin, S. Rajagopal. *ACS Sustainable Chemistry & Engineering*. **2017**, 8475.

D. Wang, S. Gao. *Organic Chemistry Frontiers*. **2014**, 556.

X. Wang, Y. Song, J. Qu, Y. Luo. *Organometallics*. **2017**, 1042.

P. van de Weghe. *Letters in Organic Chemistry*. **2005**, 113.

J. L. Wood, J. A. Porco Jr., J. Taunton,A. Y. Lee, J. Clardy, S. L. Schreiber. *Journal of the American Chemical Society*. **1992**, 5898.

D. Xu, Z. Li, S. Ma. *Tetrahedron: Asymmetry*. **2003**, 3657.

L. Xue, Z. Lin. *Chemical Society Reviews*. **2010**, 1692.

A. Yepremyan, T. G. Minehan. *Organic & Biomolecular Chemistry Impact factor*. **2012**, 5194.

T. Y. Yuen, M. A. Brimble. *Organic Letters*. **2012**, 5154.

A. Zapf, M. Beller. *Topics in Catalysis*. **2002**, 101.

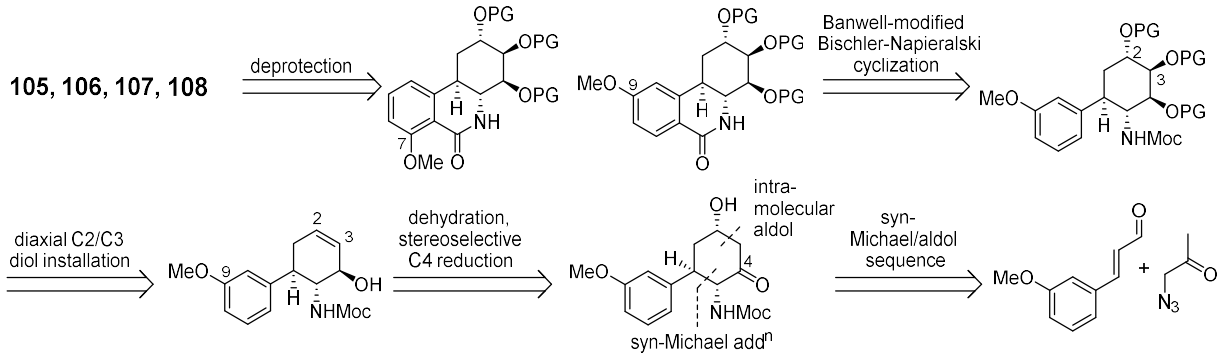
S. L. Zheng,S. Reid, N. Lin, B. Wang. *Tetrahedron Letters*. **2006**, 2331.

2.0 Thesis Introduction

From the aforementioned studies seen throughout history, including references dating back to the bible, Amaryllidaceae alkaloids have raised the expectations of what we see in a natural product's potential (Wallin, Wattmo, & Minthon, 2011). Rising from the increase in screenings performed on new derivatives, libraries of alkaloids have been created and at times showing an increase in activity (Vshyvenko, Scattolon, Hudlicky, Romero, & Kornienko, 2011; Vshyvenko, et al., 2012). In addition, technology has advanced to help further understand a target molecule, aiding in the discovery towards a compound's mode of action. Of the three major structurally distinct type of alkaloids, our study focuses on lycorane type alkaloids. Although the phenanthridone size is rather small when compared to other prescribed treatments (Manpadi, et al., 2009), important and/or interesting molecules are unfortunately isolated from their natural source in very minute quantities, *trans*-dihydronarciclasine is a great illustrative natural product reflecting this point, making its isolation from plant material impractical (Pettit, Cragg, Singh, Duke, & Doubek, 1990). Over the last 3 decades, our lab has focused on the development and discovery of natural and unnatural cytotoxic alkaloids and their derivatives in the Amaryllidaceae family (McNulty & Mo, 1998; Pandey S. , Kekre, McNulty, & Naderi, 2005; Revu, et al., 2016). In 2012 Yamakoshi et. al. reported a promising approach for visualizing non-immobilized small molecules in live cells (Yamakoshi, 2012). This new technique operates using Raman imaging, and is now known as alkyne-tag Raman imaging (ATRI). Imaging of molecules in live cells is an important tool for biology, chemical biology, biochemistry, structural biology studies, cell biology, neurobiology,

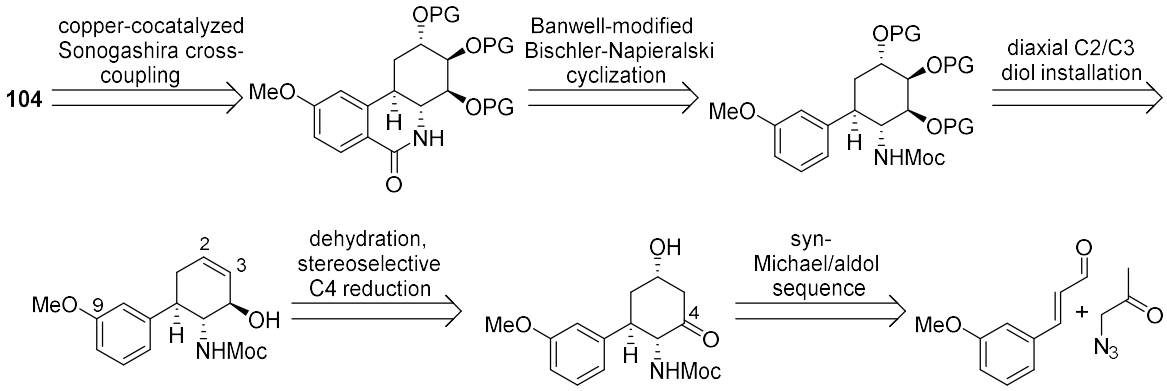
pharmaceutical, and medical sciences. Specifically, the use of fluorescent labels has allowed for the sensitive detection of small molecules in living cells (Jensen, 2013). Although this has been previously explored, a flawless technique with no short comings has yet to prevail. Problems arise when the tag/marker has a molecular weight significantly larger than the parent small molecule. Additional concerns include interfering with its biological activity, cellular localization, and dynamics of the parent molecule. ATRI permits the visualization of small molecules in living cells by using very small Raman-active tags that lack the disadvantages of target fluorescent tags, that can markedly influence the chemical and biological properties of the parent molecule. Raman microscopy can visualize the location of molecules without a fluorescent tag because of its ability to detect molecular vibrations. The vibrational spectroscopic technique is an effective tool for non-invasive optical tissue diagnosis. The technique is simple, reproducible, non-destructive to the tissue, and only requires small quantities of material with little sample preparation. Spectral bands are molecule specific and supply direct information about the biochemical composition. Modern Raman spectroscopy commonly applies a laser as its excitation light source, but this had not always been the case before lasers were available. Theoretically, if the energy from the incident photon is unchanged after collision with the molecule, the scattered photon will share the same frequency, and is known by Rayleigh or elastic scattering. When this is not the case, and the energy of the released photon has increased (anti-Stokes-Raman) or decreased (Stokes-Raman), this is called Raman or inelastic scattering. The change in photon energy corresponds to the difference in the final and initial vibrational energy levels for the molecule involved. This was first described in

1982 by Raman, who received the Nobel Prize two years after (Singh & Riess, 1998). Raman peaks often are narrow, and in many cases are associated with the vibration of a specific bond, or functional group in the molecule (Choo-Smith, et al., 2002; Huang, et al., 2003; Shafer-Peltier, et al., 2002; Gazi, et al., 2003). This method of detection was more facilitated towards molecular species existing only in large quantities in the cell ie. proteins, and lipids. Raman signals from smaller molecules tend to be weak and can easily be masked by intense overlapping Raman signals from dominant intracellular species. Alkyne functional groups although show a distinct and strong Raman scattering peak in the cellular silent region (1800-2800cm^{-1}), where most endogenous molecules show no Raman scattering. This technique's advantages include the application of a small tag(s), alkyne signals not degrading over repeated scans versus photobleaching and other forms of degradation, and ATRI having the ability to delineate many other cellular structures in addition to the alkyne-tagged structure in only one scan versus fluorescent only showing stained structures. Considering these elements, and as part of an ongoing total synthesis program, we became interested in one *trans*-dihydrolycoricidine C9 analogue **104**. Such an analogue would allow for the further understanding of ring-A activities having all the requisite functionalities present on rings-B and C. Herein, we report the first concise, chemo- and regioselective synthesis of one *trans*-dihydrolycoricidine analogue **104** and four *trans*-dihydronarciclasine derivatives **105**, **106**, **107**, **108** (scheme 25 and scheme 26).



Scheme 25. Retrosynthetic analysis of four (+)-*trans*-dihydronarciclasine derivatives. The natural-product numbering system is employed. PG = protecting group, Moc = methoxycarbonyl.

We anticipated that compounds **105**-**108** would be derived from the methoxycarbonyl-substituted aminocyclitols, and advanced using BBN reaction conditions generating a mixture of regioisomers from the fully functionalized ring-C compound. This precursor would be most convenient for the ring closing step following the generation of all correctly functionalized carbon atoms in ring-C. Fully functionalized ring-C production from routes including the stereoselective reduction of the C4 enone, and 2,3-diaxial diol formation from the ensuing epoxidation with *m*-CPBA would align the correct stereocenters to match our final target molecules. A key step involved in synthesizing ring-C would hinge upon the regiocontrolled syn-stereoselective Michael addition of α-azidoacetone and subsequent intramolecular aldol cyclization with *trans*-*m*-methoxycinnamaldehyde using Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether as the catalyst.



Scheme 26. Retrosynthetic analysis of the (+)-*trans*-dihydrolycoricidine C9-analogue. The natural-product numbering system is employed. PG = protecting group, Moc = methoxycarbonyl.

With this strategy in mind, we traced back our steps to the C9-methoxy BBN product and envisioned pushing the synthesis towards the production of a labile phenol-derived sulfonated hydroxyl group setting up a reactive cross-coupling partner for the ensuing copper-cocatalyzed Sonogashira reaction towards **104**.

With the focal point of the synthesis falling on the substitution of an alkyne-tag on C9 for visual imaging purposes, in addition to the antiviral activities of four *trans*-dihydronarciclasine ring-A modified, fully functionalized rings-B and C derivatives synthesized via asymmetric chemical syntheses will be presented. These derivatives will add to the growing library of studies related to *trans*-dihydronarciclasine. The synthesis of unnatural derivatives/analogues is often required to improve bioavailability and lower any side effects associated if applicable. Encouraged by the recent enantioselective organocatalytic approaches toward six-membered carbocycles (see chapter 1.4DD), our group developed a [3+3]-type Michael-aldol sequence towards (+)-*trans*-dihydrolycoricidine (McNulty & Zepeda-Velazquez, 2014; McNulty, et al., 2016). Studies involved an α-nitrogen substituted acetone reagent reacting with an enal to provide the

rapid entry to the Amaryllidaceae core. Our work was used to continue such procedures and expand further the asymmetric entry to amino inositols. Natural product syntheses in recent years have taken steps forward in the direction towards their practical production of their target molecules, which was demonstrated by the (+)-*trans*-dihydrolycoricidine synthesis by McNulty et. al. being performed in only nine steps with an overall yield of 12% and greater than 98% *ee* (McNulty & Zepeda-Velazquez, 2014). Considering these elements, ring-A was modified following the previous SAR studies performed. Gabrielsen found that (+)-*trans*-dihydronarciclasine was the most effective anti-flaviviral analogue, exchanging the trans-fused B/C-ring junction with a cis-fused junction and loss or epimeric alcohols in ring-C would result in a significant drop, or loss all together in antiviral activity. It was also discovered that only the 7-deoxy analogues provided activity against the SF virus, leaving our C9 alkyne-tag analogue, C7, C9 methoxy and C9 hydroxy substituted derivatives with promising expectations for select viruses. With the limited amount of antiviral studies performed, McNulty collaborated with Stanley Medical Research Institute researching anti-HSV-1 activities with various Amaryllidaceae alkaloids and their analogues. McNulty's discoveries included *trans*-dihydrolycoricidine to be superior over the current standard for HSV-1 infection (ACV), configurationally defined (R) secondary alcohol at C3 to be vital for antiviral activities, the critical importance in the phenanthridone type chair conformation for antiviral activity, and established *trans*-dihydrolycoricidine as the functionally minimum antiviral pharmacophore. The studies report of the essential moieties were conserved, while modifying ring-A to advance SAR studies by exchanging the methylenedioxy bridge. Rapid entry to structures similar to this coming from

inexpensive starting materials is in high demand after seeing the significant medical potential attributed to these alkaloids. Since the cellular macromolecular target(s) is still yet to be identified, advancing our analysis towards structurally simplified potent derivatives with the potential to maintain or increase activity will be communicated. Key elements within the syntheses will be discussed more in depth related to the [3+3] method towards an asymmetric entry to the aminocyclitol core, reduction of the azide to the amine and how this influenced our starting material selection, α- and β-epoxide transformations when producing triol **123**, BBN cyclization for ring-B, and the copper-cocatalyzed Sonogashira reaction towards the C9 alkyne-tagged *trans*-dihydrolycoricidine analogue.

2.01 References

L. P. Choo-Smith, H. G. M. Edwards, H. P. Endtz, J. M. Kros, F. Heule, H. Barr, H. Robinson, H. A. Bruining, G. J. Pupells. *Biopolymers (Biospectroscopy)*. **2002**, 1.

L. Díaz, A, Delgado. *Current Medicinal Chemistry*. **2010**, 2393.

A. Fürst, P. A. Plattner. *Helvetica Chimica Acta*. **1949**, 275.

E. Gazi, J. Dwyer, P. Gardner, A. Ghanbari-Siakhani, A. P. Wde, N. P. Lockyer, J. C. Vickerman, N. W. Clarke, J. H. Shanks, L. J. Scott, C. A. Hart, M. Brown. *Journal of Pathology*. **2003**, 99.

Z. Huang, A. McWilliams, M. Lui, D. I. McLean, S. Lam, H. Zeng. *National Journal of Cancer*. **2003**, 1047.

H. Hussain, A. Al-Harrasi, I. Green, I. Ahmed, G. Abbas, U. Rehman. *RSC Advances*. **2014**, *25*, 12882.

E. Jensen. *The Anatomical Record*. **2013**, 1.

S. R. S. S Kotti, C. Timmons, G. Li. *Chemical Biology & Drug Design*. **2006**, 101.

T. Kouko, K. Matsumura, T. Kawasaki. *Tetrahedron*. **2005**, 2309.

J. Kroutil, T. Trnka, M. Budesinsky, M. Černý. *European Journal of Organic Chemistry*. **2002**, 2449.

C. Kumar, Studer, J. A. *Chemistry - A European Journal*. **2008**, 6326.

M. Manpadi, A. S. Kireev, I. V. Magedov, J. Altig, P. Tongwa, M. Y. Antipin, A. Evidente, W. van Otterlo, A. Kornienko. *The Journal of Organic Chemistry*. **2009**, *74*, 7122.

J. McNulty, L. D'Aiuto, Y. Zhi, L. McClain, C. Zepeda-Velázquez, S. Ler, H. Jenkins, M. B. Yee, P. Piazza, P. R. Yolken, V. Kinchington, L. Nimgaonkar. *ACS Medicinal Chemistry Letters*. **2016**, 46.

J. McNulty, J. Mao, R. Gibe, R. Mo, S. Wolf, G. R. Pettit, D. L. Herald, M. R. Boyd. *Bioorganic & Medicinal Chemistry Letters*. **2001**, 169.

J. McNulty, R. Mo. *Journal of the Chemical Society, Chemical Communications*. **1998**, 933.

J. McNulty, C. Zepeda-Velazquez. *Angewandte Chemie International Edition*. **2014**, 8450.

S. Pandey, N. Kekre, J. McNulty, J. Naderi. *Cancer Chemotherapy and Pharmacology*. **2005**, 29.

D. J. Pasto, J. A. Gontarz. *Journal of the American Chemical Society*. **1970**, 7480.

G. R. Pettit, G.M. Cragg, S. B. Singh, J. A. Duke, D. L. Doubek. *Natural Products*. **1990**, 176.

O. Revu, C. Zepeda-Velázquez, A. J. Nielsen, J. McNulty, R. H. Yolken, L. Jones-Brando. *ChemistrySelect*. **2016**, 5895.

K. E. Shafer-Peltier, A. S. Haka, M. Fitzmaurice, J. Crowe, R. R. Dasar, M. S. Feld. *Journal of Raman Spectroscopy*. **2002**, 552.

R. Singh, F. Riess. *Current Science*. **1998**, 965.

M. Squillacote, R. S. Sheridan, O. L. Chapman, F. A. L. Anet. *Journal of the American Chemical Society*. **1975**, 3244.

H. Tanimoto, K. Kakiuchi. *Natural Product Communications*. **2013**, 1021.

B. M. Trost, D. L. A. Van Vranken. *Journal of the American Chemical Society*. **1993**, 444.

S. Vshyvenko, J. Scattolon, T. Hudlicky, A. E. Romero, A. Kornienko. *Bioorganic & Medicinal Chemistry Letters*. **2011**, 4750.

S. Vshyvenko, J. Scattolon, T. Hudlicky, A. E. Romero, A. Kornienko, D. Ma, I. Tuffley, S. Pandey. *Canadian Journal of Chemistry*. **2012**, 932.

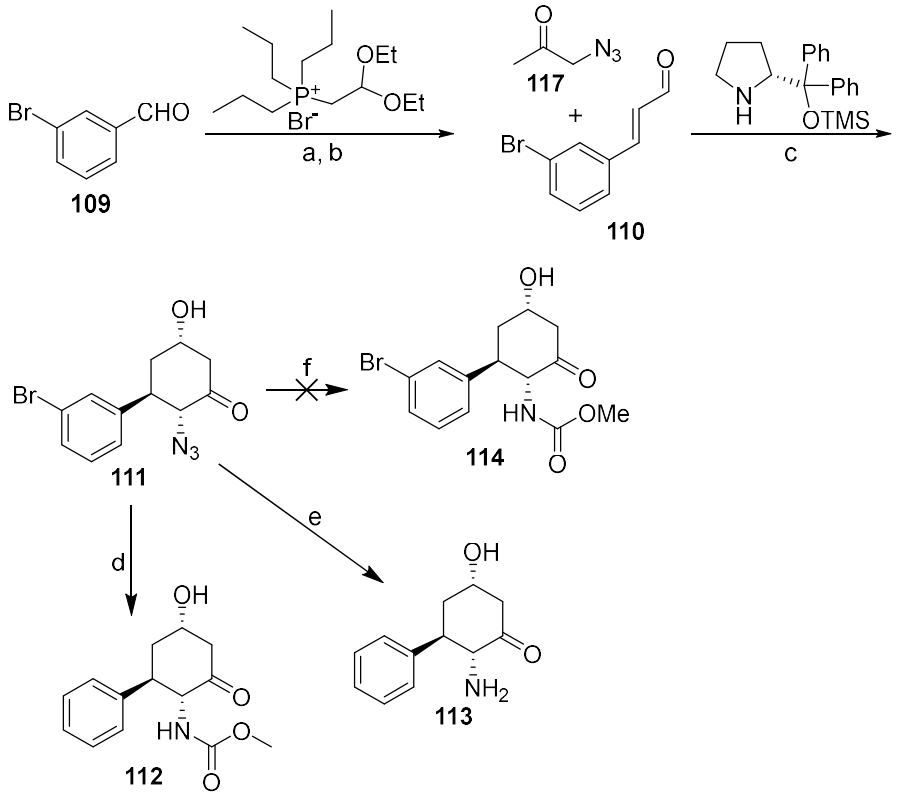
A. K. Wallin, C. Wattmo, L. Minthon. *Neuropsychiatric Disease and Treatment*. **2011**, *7*, 565.

H. Wang, K. N. Houk, *Chemical Science*. **2014**, 462.

H. Yamakoshi. *Journal of the American Chemical Society*. **2012**, 20681.

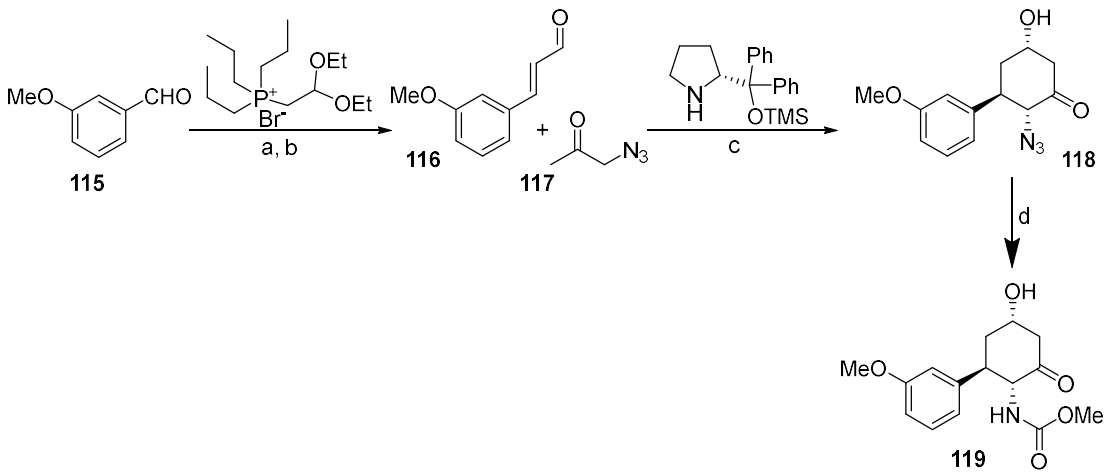
2.1 Synthesis and Functionalization of Ring-C

We started our synthesis from commercially available *m*-bromobenzaldehyde **109** (scheme 27), in sight of an easy handle for cross-coupling reactions towards the isolation of the C9 alkyne-tagged analogue. This was then subjected to a two-carbon aldehyde to enal homologation using a diethyl acetal-functionalized Wittig reagent yielding *trans-m*-bromocinnamaldehyde **110** in a 90% yield. After production of Michael acceptor **110**, subsequent syn-stereoselective addition with a succeeding intramolecular aldol was achieved using Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether (14 mol%) a secondary amine catalyst, in combination with the bulky chiral tertiary amine base quinidine (14 mol%), producing cycloadduct **111** in a 57% yield (McNulty & Zepeda-Velazquez, 2014). Reduction and subsequent protection of azide **111** while not effecting the aryl bromide moiety was more challenging than expected due to unforeseen complications with the starting material **109**. While starting off with a mild procedure using Staudinger reaction conditions (Tanimoto & Kakiuchi, 2013), and following-up with two alternative routes using 10% Pd/C, H_2, DMDC and Zn/aq. NH_3, all came back unsuccessful (scheme 27). The Staudinger reaction conditions have been applied towards the total syntheses of many other marine bisindole alkaloids that shared structural similarities including an aryl bromide moiety during the reduction steps (Kouko, Matsumura, & Kawasaki, 2005), communicating that our substrate was the issue at hand. Although reduction of the azide was successful with 10% Pd/C, H_2, DMDC and Zn/aq. NH_3, unwanted debromination took place eliminating the aryl halide moiety that is required for the later steps in the synthesis producing carbamate **112** and primary amine **113** respectfully.



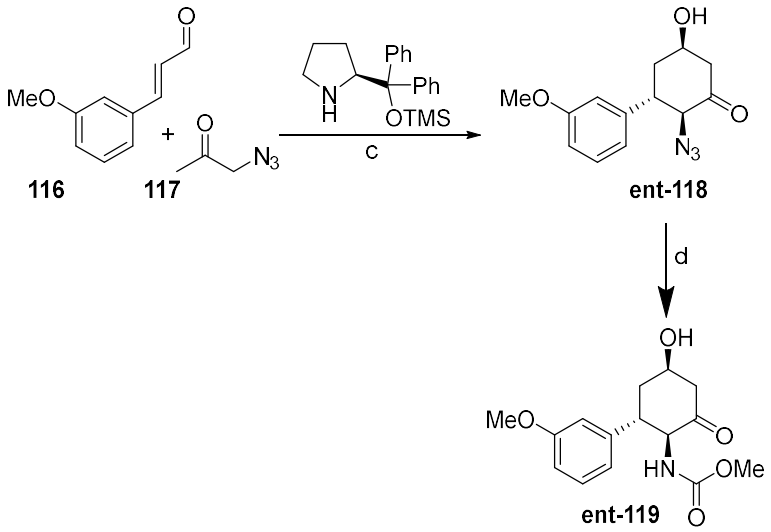
Scheme 27. Initial findings when using *m*-bromobenzaldehyde as our precursor: a) NaH, THF, **109**; b) 1M HCl, rt, 90%; c) quinidine, CH_2Cl_2, -10 °C - rt, 57%; d) 10% Pd/C, DMDC, EtOAc, 95%; e) (i) Zn dust, EtOH/H_2O (4:1), 0.5 h; (ii) aq. NH_3, EtOAc, 30%; f) PEt_3, THF, H_2O, 0%.

Unsuccessful attempts towards **114** led us to change our starting material to commercially available *m*-anisaldehyde **115**, setting up the total synthesis of four additional targets for antiviral studies. Other benefits for using this precursor will be mentioned in chapter 2.3. *m*-Anisaldehyde **115** was subjected to a two-carbon aldehyde to enal homologation using a diethyl acetal-functionalized Wittig reagent yielding *trans-m*-methoxycinnamaldehyde **116** in a 95% yield (see scheme 28).



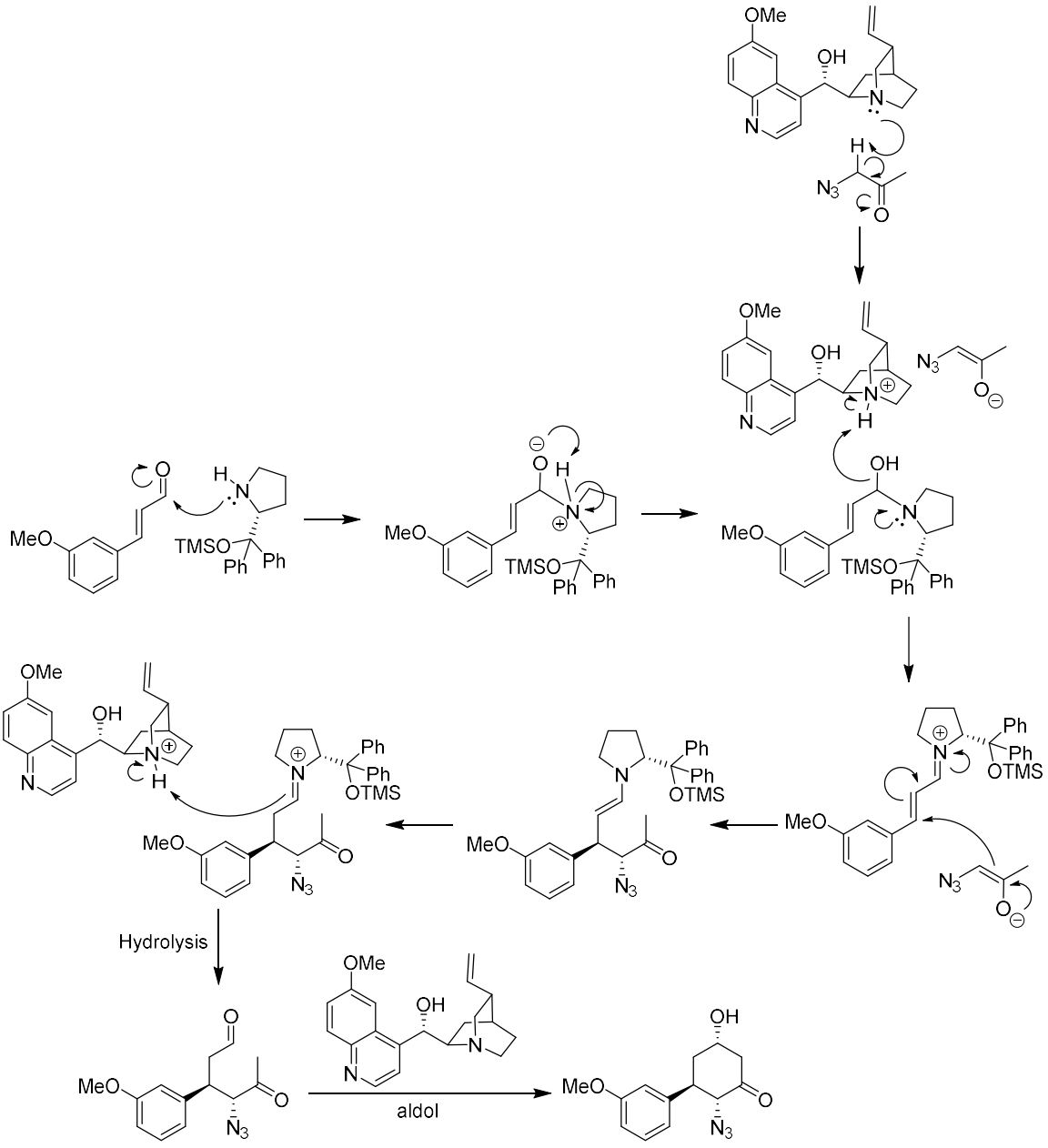
Scheme 28. Synthesis towards the aminocyclitol core using *m*-anisaldehyde as an alternative starting material: a) NaH, THF, **115**; b) 1M HCl, rt, 95%; c) quinidine, CH_2Cl_2, -10 - 0 °C, 24 h, 55%; d) DMDC, H_2, 10% Pd/C, MeOH, rt, 32 h, 88%.

The vital iminium ion-mediated [3+3]-Michael-aldol sequence (McNulty & Zepeda-Velazquez, 2014; Revu, et al., 2016) of **116** with α-azidoacetone **117** proved highly effective once again using Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether (10 mol%) in combination with quinidine (10 mol%), producing cycloadduct **118** in a 55% yield. A duplicate reaction was set-up, but this time while using Jørgensen (*S*)-diphenylprolinol trimethylsilyl ether instead of Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether (scheme 29).



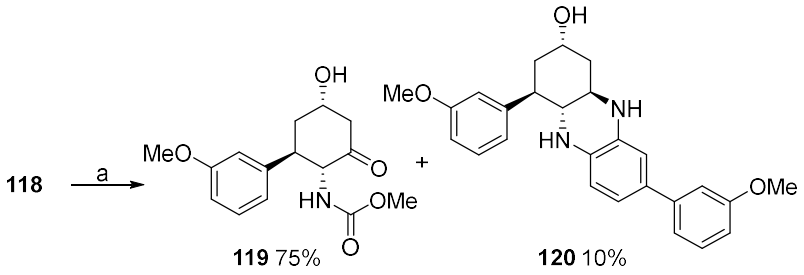
Scheme 29. Duplicate reaction with Jørgensen (*S*)-diphenylprolinol trimethylsilyl ether instead of using Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether: c) quinidine, CH_2Cl_2, -10 - 0 °C, 24 h, 55%; d) DMDC, H_2, 10% Pd/C, MeOH, rt, 32 h, 88%.

The results followed as when performed with Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether, but this time isolating the enantiomer of **118** (***ent*-118**). Mechanistic insight for the production of the aminocyclitol (Díaz & Delgado, 2010) ring-C is shown in scheme 30.



Scheme 30. A proposed mechanism using Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether.

Following the production of the six-membered carbocycle **118**, reduction and subsequent protection of azide **118** to the corresponding carbamate **119** was performed, but only after being optimized from our initial discovery of only isolating a 75% yield.



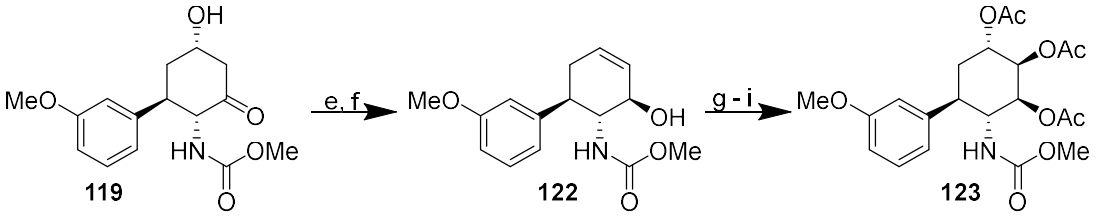
Scheme 31. Reduction and carbamoylation leading to by-product formation from our initial trials **120**: a) H_2, 10% Pd/C, DMDC, MeOH, 16 h, rt, 75% **119**, 10% **120**.

After using only 1 eq. of 10% Pd/C in the presence of DMDC over 16 h an unforeseen vicinal diamine (Kotti, Timmons, & Li, 2006) by-product **120** was isolated in a 10% yield (Scheme 31). Although the anticipated reduction of the azide **118** to moc-protected amine **119** was successful, this free amine could then participate in a condensation reaction via homocoupling with the ketone functional group and followed hydroxy elimination producing **120**. Smooth transformation of azide **118** to carbamate **119** was performed by advancing the catalyst loading and reaction time. When using 1.5 eq. of 10% Pd/C and allowing 36 h to react, carbamate **119** was isolated in an 88% yield. Carbamate **118** and carbamate ***ent*-118** each provided >99% *ee*. The enantiomers were cleanly baseline resolved using chiral HPLC (AD-H) column (see appendix D).

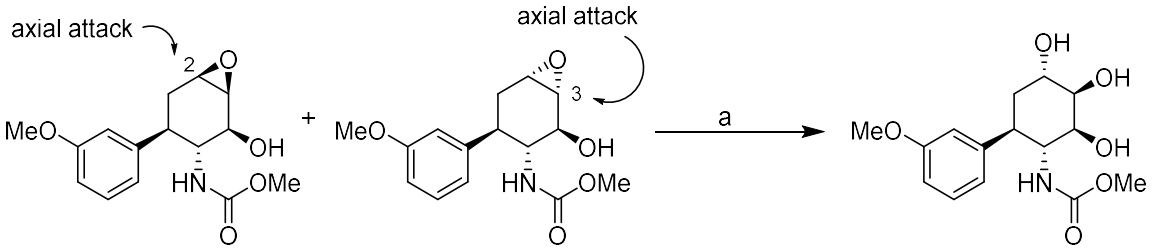
Ensuing the formation of each of the targeted molecules ring-C structures, the following steps were constructed towards the formulation of ring-C's five stereogenic carbon centers matching the configurations required by the final targets. Dehydration of **119** with Methanesulfonyl chloride and Hünig's base (DIPEA) produced **121** a translucent oil providing a crude yield of 95% after partitioning. This product was used without further

purification and reduced to the equatorial alcohol **122** in a 95% yield with lithium tri-*tert*-butoxyaluminium hydride (Li(*t*-BuO)$_3$AlH) (scheme 32).

Scheme 32. Synthesis towards a protected but fully functionalized ring-C: e) DIPEA, MsCl, CH_2Cl_2, rt, 10 h; f) Li(*t*-BuO)$_3$AlH, THF, O °C – rt, 20 h, 95% over two steps; g) *m*-CPBA, $NaHCO_3$, CH_2Cl_2, rt, 24 h; h) NaOBz/H_2O, 90 - 95 °C, 16 h; i) Ac_2O, Py, rt, 16 h, 77% over three steps.

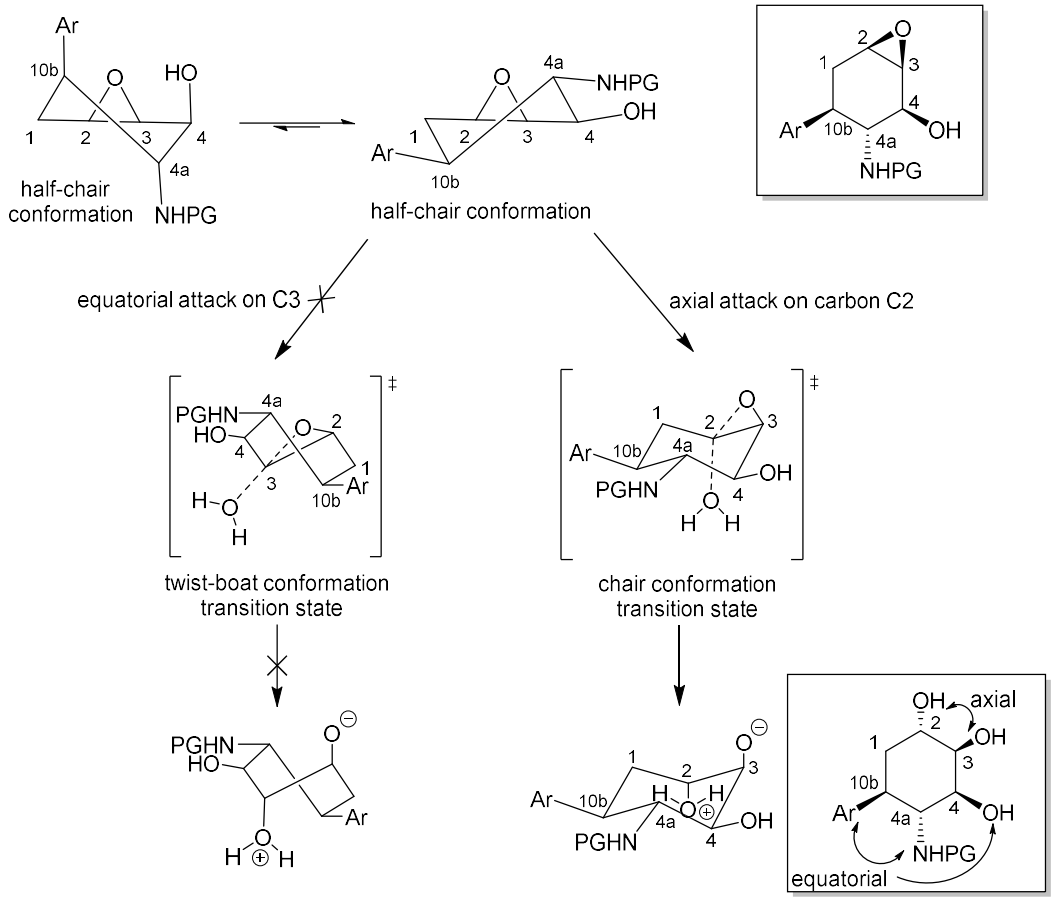


Epoxidation of the allylic alcohol **122** using *m*-CPBA (Hussain, et al., 2014; Henbest & Wilson, 1957) produced two diastereomers, a mixture of α- and β-epoxides that were directly transformed in a stereospecific manner to the single diastereomer 2,3-diaxial diol **123** (Revu, et al., 2016). Although epoxidation of the electron rich allylic alcohol produced two diastereomers, no separation was required following the ensuing base mediated *trans*-diaxial epoxide opening with NaOBz/H_2O leading to a single *trans*-diaxial product **123** without incident (Fürst & Plattner, 1949). Similar procedures from the McNulty group have been performed related to lycorane derivatives in the formation of the 2,3-diol subunit (McNulty, et al., 2001; McNulty & Zepeda-Velázquez, 2014; Revu, et al., 2016).



Scheme 33. Nucleophilic attack by H_2O on each of the corresponding diastereomers proceeding via a *trans*-diaxial attack forming the same product: a) NaOBz, H_2O, 90 - 95 °C, 16 h.

From these cases, the nucleophile (H_2O) and the epoxide oxygen will always be *trans* to one another in the final product assuring the correct stereogenic centers for every carbon in ring-C (Niederer, Fodor, & Catino, 2018; Doitomi, Xua, & Hirao, 2017). In both cases, the ring opens out to give a stable diaxial chair conformation product (scheme 34) (Murphy, 1969; Wang H. H., 2011; Clayden, 2001; Fürst A. P., 1949; Deora & Carlier, 2019). Nucleophilic attack takes place through the chair transition state over the higher in energy twist-boat transition state due to the unfavorable torsional interactions in the ring. These interactions influence the stereoselectivity for many synthetically important transformations (Wang & Houk, 2014; Martínez, Vega, Aguirre, Lantaño, & Moltrasio, 2012). In terms of relative energy, the difference between the twist-boat and chair conformations of cyclohexane is 5.5 kcal/mol (Squillacote, Sheridan, Chapman, & Anet, 1975), inevitably leaving more complex molecules to favor the chair transition state even more. Other examples including halonium ions (Pasto & Gontarz, 1970), aziridines (Kroutil, Trnka, Budesinsky, & Černý, 2002), and five-membered ring systems (Trost & Van Vranken, 1993) have followed the Fürst−Plattner rule (Fürst & Plattner, 1949) in their stereoselective additions.



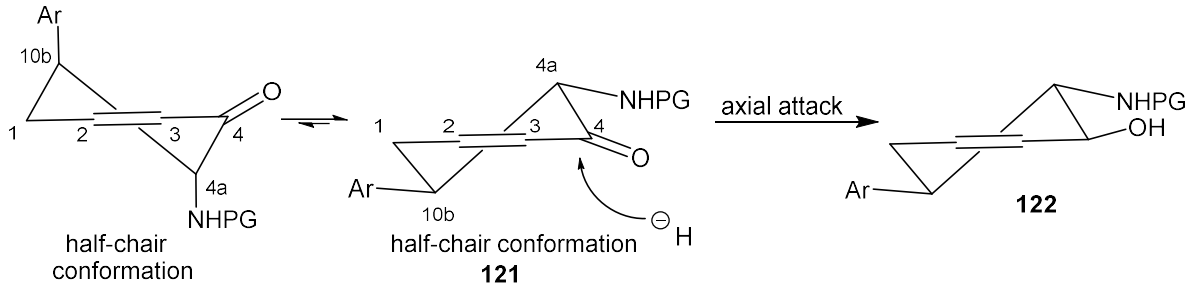
Scheme 34. Stereoelectronic requirements for backside displacement leads to axial attack over an equatorial attack (one of the epoxides diastereomer's illustrated), favoring the *trans* diaxial product towards **123** following the Fürst−Plattner rule or the *trans* diaxial effect. Both diastereomers follow in this rule/effect. PG = protecting group.

The ensuing monomeric triol was acetylized in situ with acetic anhydride in the presence of pyridine to give triacetate **123** in a 77% yield over three sequential steps without incident.

2.11 Conclusion

Succeeding unbeknownst complications with *m*-bromobenzaldehyde **109**, early stage synthesis of the fully functionalized ring-C was developed from readily available starting material **115** without incident. Key components involved the organocatalytic regiocontrolled syn-stereoselective Michael-aldol addition of α-azidoacetone **117** with *trans*-*m*-methoxycinnamaldehyde **116** acting as the Michael acceptor, followed by an intramolecular aldol reaction. The annulation materialized with high enantioselectivity in the presence of Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether and quinidine (bulky chiral cinchona alkaloid base) reducing the number of steps required towards the final products. The sequence proceeds by iminium ion or enamine activation, but the precise role played by the base is not fully understood. The asymmetric Michael/aldol sequence provided swift formation of ring-C, with high efficacy. Using this well optimized route, permitted access to our most densely functionalized subunit of the molecule with great efficacy when only requiring two synthons bearing no chiral centers. Such access from readily available material highlights the advantages seen in our [3+3] Michael-aldol sequence towards the production of aminocyclitol cores. Manipulation via Jørgensen (*R*)-diphenylprolinol trimethylsilyl ether and our base additive has led to an increased selectivity which we are able to apply and study towards the formation of other naturally occurring products bearing cyclic or polycyclic cores. Subsequent dehydration proceeded smoothly, setting up the equatorial alcohol **122** formation without further purification. The reaction was slow, but due to steric hindrance, and reactivity in the more stable half-chair conformer selective reduction was observed over the higher in energy half-chair conformer,

leaving the axial attack dominant when all substituents are equatorial (scheme 35) (Cherest M. F., 1968; Cherest M. F., 1968; Wu, 1991).

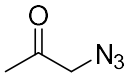


Scheme 35. Reduction to the equatorial alcohol **122** via axial attack with a bulky reducing agent Li(*t*-BuO)$_3$AlH occurring when the molecule is in its most stable half-chair conformation.

While the aryl substituent is axial oriented, this leads to a steric interaction with the C4 ketone substituent, leaving the C4 ketone substituent favoring pointing away from the ring and equatorial. Epoxidation of the allylic alcohol **122** led to the formation of a mixture of α- and β-epoxides that were directly transformed in a stereospecific manner assuring the correct stereogenic centers for every carbon in ring-C with the natural products. Diaxial opening with NaOBz/H_2O producing the *trans*-diaxial product was an effective one pot reaction towards the isolation of the fully functionalized ring-C molecule.

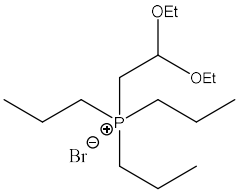
2.12 Experimental

1-Azidopropan-2-one [115]



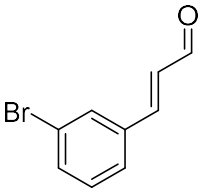
Sodium azide (2.485 g, 38.23 mmol, 1 eq.) was added to a solution of 1-bromopropan-2-one (2.54 mL, 31.63 mmol, 1.2 eq.) in acetone (10 mL) and water (20 mL). The reaction mixture was stirred at rt for 24 h and acetone was removed using a nitrogen stream. The remaining aqueous solution was extracted with CH_2Cl_2 (2 x 25 mL). The combined organic extracts were washed with brine, dried over anhydrous Na_2SO_4, and filtered. The filtrate had a nitrogen stream blown over it until all solvent was removed, yielding (2.505 g, 80% **yield**) of the title compound as a colorless liquid. This compound is known and matches the reported spectroscopic data (McNulty & Zepeda-Velázquez, 2014).

Tripropyl-(2,2-diethoxyethyl)-phosphonium bromide



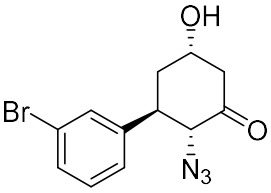
A solution of bromoacetaldehyde diethyl acetal (9.60 g, 48.7 mmol, 1 eq.) and tri-*n*-propylphosphine (8.58 g, 53.6 mmol, 1.10 eq.) in THF (27 mL) was heated for 24 h at 60 °C. After cooling, the solvent and the residual tri-*n*-propylphosphine were removed on the rotary evaporator (75 °C at 5 mbar). The obtained yellow oil was dried *in vacuo* (0.1 mbar) isolating 16.38 g of phosphonium bromide as a white solid. No further purification was necessary. Storage temperature: 0 °C. This compound is known and matches the reported spectroscopic data (McNulty & Zepeda-Velázquez, 2014).

(*E*)-3-(3-bromophenyl)acrylaldehyde [110]



m-Bromobenzaldehyde **109** (2.47 g, 13.4 mmol, 1 eq.) and tri-*n*-propyl-(2,2-diethoxyethyl)-phosphonium bromide (5.99 g, 16.7 mmol, 1.25 eq.) were dissolved in anhydrous THF (20 mL). Sodium hydride (0.97 g, 40.3 mmol, 3 eq., 60% dispersed in mineral oil) was added slowly to the reaction mixture over a period of 10 min, maintaining the temperature below 30 °C, and the suspension was stirred for 24 h at rt. Water (25 mL) was added to the mixture, and extracted with CH_2Cl_2 (3 × 25 mL). The extracts were combined and washed again with water (2 x 25 mL). The organic layers were dried using Na_2SO_4, filtered and concentrated *in vacuo* to afford diethyl acetal 3.19 g as red oil. Subsequently 20 mL of 2 M HCl was added and stirred for 1 h at rt. When the reaction was completed, the reaction mixture was extracted with CH_2Cl_2 (3 × 25 mL) and was separated by column chromatography to afford *trans-m*-bromocinnamaldehyde **110** (2.54 g, 90% **yield**). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 9.63 (d, *J* = 7.6 Hz, 1H), 7.63 (s, 1H), 7.48 (d, *J* = 7.9 Hz, 1H), 7.42 (d, *J* = 7.2 Hz, 1H), 7.33 (d, *J* = 15.8 Hz, 1H), 7.24 (t, *J* = 8.21 Hz, 1H), 6.63 (dd, *J* = 15.8, 7.6 Hz, 1H). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 193.29, 150.69, 136.06, 133.98, 131.23, 130.61, 129.66, 126.93, 123.24. **HRMS** (EI): exact mass calculated for C_9H_7BrONa [$(M + Na)^+$], 232.9578; found 232.9562.

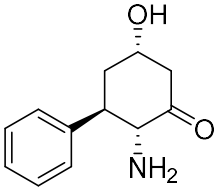
(2*R*,3*R*,5*S*)-2-azido-3-(3-bromophenyl)-5-hydroxycyclohexanone [111]



A solution of *trans-m*-bromocinnamaldehyde **110** (0.978 g, 4.68 mmol, 1 eq.) and (*R*)-(+)-α,α-diphenyl-2-pyrrolidinemethanol trimethylsilyl ether (0.145 g, 0.45 mmol, 0.14 eq.) in CH_2Cl_2 (6.2 mL) was stirred for 10 min, after which it was cooled to -10 °C. 1-Azidopropan-2-one **117**

(0.442 g, 4.46 mmol, 1.05 eq.) was added dropwise over 5 min. The brown solution was stirred for 20 min at rt and quinidine (0.145 g, 0.45 mmol, 0.14 eq.) was added in one portion. The reaction was stirred at rt for 24 h, after which TLC (CH_2Cl_2/MeOH 98:2) showed full conversion. The CH_2Cl_2 was carefully evaporated (30 °C, 32 mbar) yielding a brown oil that was purified by flash chromatography (eluent CH_2Cl_2/MeOH 100:0 to 95:5), to afford **111** (0.825 g, 57% **yield**). **$[\alpha]^{23}_D$** = +68 (c = 1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.49-7.43 (m, 2H, Ar-*H*), 7.27-7.25 (m, 2H, Ar-*H*), 4.60-4.57 (m, 1H, C*H*-OH), 4.13 (d, *J* = 12.3 Hz, 1H, C*H*-N_3), 3.53-3.38 (td, *J* = 12.3 Hz, 1H, Ph-C*H*), 2.82 - 2.75 (m, 2H, CO-CH_2), 2.23-2.16 (m, 2H, Ph-CH-CH_2). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 203.37, 142.84, 130.61, 130.43, 130.32, 126.25, 122.85, 77.21, 67.53, 47.96, 44.27, 39.03. **HRMS** (ESI): exact mass calculated for $C_{12}H_{12}BrN_3O_2Na$ [$(M + Na)^+$], 332.0011; found 332.0019.

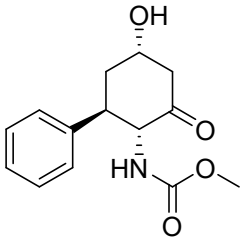
(2*R*,3*R*,5*S*)-2-amino-5-hydroxy-3-phenylcyclohexanone [113]



Zinc powder (0.013g, 0.20 mmol, 0.7 eq.) was added to a solution of azide **111** (0.092g, 0.30 mmol, 1 eq.) and ammonium chloride (37.8g, 0.7 mmol, 3.5 eq.) in ethyl alcohol (10 mL) and water (2.7 mL). The solution was stirred vigorously at rt for 30 min. Ethyl acetate (EtOAc) (200 mL) and aqueous ammonia (10 mL) were then added. The mixture was filtered, and the filtrate was washed with brine, and dried over anhydrous Na_2SO_4. After removal of the solvent under reduced pressure, the residue was purified by flash chromatography to afford amine **113** (0.018 g, 30% **yield**). $[\alpha]^{23}_D$ = -10 (c = 0.36, MeOH, l =1 dm).**^{1}H NMR** (600 MHz, DMSO) δ 7.98 (s, 2H, NH_2), 7.46-7.37 (m, 4H Ar-*H*), 7.32 (t, *J* = 7.1 Hz, 1H, Ar-*H*), 5.26 (bs, 1H, O*H*|), 4.59 (d, *J* = 9.8 Hz, 1H, C*H*-NH_2), 4.36-4.35 (m, 1H, C*H*-OH), 3.43 (td, *J* = 12.4,

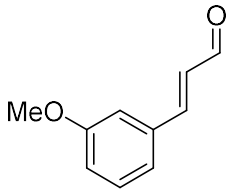
2.6 Hz, 1H, COCH_2), 3.06 (dd, J = 2.5, 12.4Hz, 1H, COCH_2), 2.39-2.33 (m, 1H, Ph-CH_2), 1.90 (dt, J = 29.1, 14.5 Hz, 1H, Ph-CH_2). **^{13}C NMR** (151 MHz, DMSO) δ 204.27, 139.62, 128.90 (2C), 127.96 (2C), 127.53, 66.63, 60.69, 47.13, 43.19, 39.25. **HRMS** (ESI): exact mass calculated for $C_{12}H_{15}NO_2Na$ [$(M + Na)^+$], 228.1000; found 228.1009.

methyl ((1*R*,4*S*,6*R*)-4-hydroxy-2-oxo-6-phenylcyclohexyl)carbamate [112]



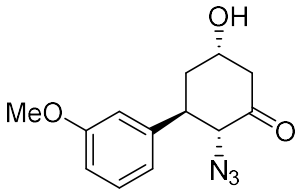
In a 50 ml RBF, azide **111** (0.046 g, 0.15 mmol, 1 eq.), dimethyl dicarbonate (0.060 g, 0.45 mmol, 3.0 eq.), and 10% Pd/C (0.012 g, 0.113 mmol, 0.75 eq.) were suspended in EtOAc (15 mL). The vessel was sealed and subjected to hydrogen balloon (1 atm) with vigorous stirring for 12 h, after which TLC (CH_2Cl_2/MeOH 95:5) showed full conversion. The suspension was filtered through a celite® pad and carefully evaporated (20 °C, 0.1 mbar). The translucent grey oil was purified by flash chromatography (eluent CH_2Cl_2/MeOH 100:0 to 97:3) giving the product as a translucent oil **112** (0.037g, 95% **yield**). **^{1}H NMR** (600 MHz, DMSO) δ 7.30 (dt, J = 15.0, 7.4 Hz, 4H, Ar-*H*), 7.19 (t, J = 7.0 Hz, 1H, Ar-*H*), 7.09 (d, J = 9.2 Hz, 1H, CON*H*), 5.02 (s, 1H, CH-O*H*), 4.47 (dd, J = 12.2, 9.4 Hz, 1H, C*H*-NH), 4.29 (m, 1H,C*H*-OH), 3.38 (s, 3H, NHCOOCH_3), 2.92 (dd, J = 13.5, 2.7 Hz, 1H, COCH_2), 2.39 (dd, J = 13.6, 2.8 Hz, 1H, COCH_2), 2.27 (dt, J = 13.2 2.5 Hz, 1H, PhCHCH_2), 1.89 (dd, J = 13.5, 2.8 Hz, 1H, PhCHCH_2). **^{13}C NMR** (151 MHz, DMSO) δ 206.29, 156.62, 142.79, 128.12 (2C), 127.63 (2C), 126.28, 66.85, 62.94, 51.05, 47.56, 43.10 39.61. **HRMS** (ESI): exact mass calculated for $C_{14}H_{17}NO_4Na$ [(M + Na)+], 185.0578; found 185.0566. **$[\alpha]^{23}_D$** = -12 (c = 1, MeOH, l = 1 dm).

(*E*)-3-(3-methoxyphenyl) acrylaldehyde [116]



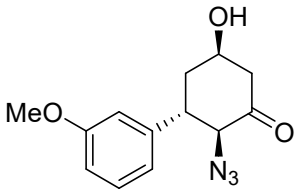
m-Anisaldehyde **115** (1.822 g, 13.4 mmol, 1 eq.) and tri-*n*-butyl-(2,2-diethoxyethyl)-phosphonium bromide (5.99 g, 16.7 mmol, 1.3 eq.) were dissolved in anhydrous THF (20 mL). Sodium hydride (0.97 g, 40.3 mmol, 3 eq.) was added to the reaction mixture over a period of 10 min, maintaining the temperature below 30 °C, and the suspension was stirred for 24 h at rt. Water (25 mL) was added to the mixture, and the mixture was extracted with CH_2Cl_2 (3 × 25 mL). The extracts were combined and washed again with water (2 x 25 mL). The organic layers were dried using Na_2SO_4, filtered and concentrated *in vacuo* to afford diethyl acetal (3.19 g) as a red oil. After 20 mL of 2 M HCl was added and stirred for 1 h at rt. When the reaction was completed, the reaction mixture was extracted with CH_2Cl_2 (3 × 25 mL), and the compound was separated by column chromatography to afford *trans*-*m*-methoxycinnamaldehyde **116** (2.063 g, 95% **yield**). This compound is known and matches the reported spectroscopic data. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 9.67 (d, *J* = 7.7 Hz, 1H, *H*C=O), 7.41 (d, *J* = 15.9 Hz, 1H, C*H*=CH-CHO), 7.31 (t, *J* = 7.9 Hz, 1H, Ar-H), 7.13 (d, *J* = 7.6 Hz, 1H, Ar-H), 7.05 (s, 1H, Ar-*H*), 6.96 (dd, *J* = 8.2, 1.8 Hz, 1H, Ar-H), 6.67 (dd, *J* = 15.9, 7.7 Hz, 1H, CH=*CH*-CHO), 3.81 (s, 3H, -OC*H*$_3$). **HRMS** (ESI): exact mass calculated for $C_{10}H_{10}O_2Na$ [(M + Na)+], 185.0578; found 185.0566.

(2*R*,3*R*,5*S*)-2-azido-5-hydroxy-3-(3-methoxyphenyl)cyclohexanone [118]



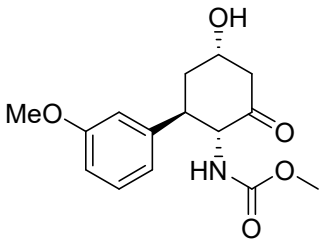
A mixture of *trans-m*-methoxycinnamaldehyde **116** (0.758 g, 4.68 mmol, 1 eq.) and (R)-(+)-α,α-diphenyl-2-pyrrolidinemethanol trimethylsilyl ether (0.145 g, 0.45 mmol, 0.1 eq.) in CH_2Cl_2 (6.2 mL) was stirred for 10 min, after which it was cooled to -10 °C. 1-Azidopropan-2-one **117** (0.442 g, 4.46 mmol, 0.95 eq.) was added dropwise over 5 min. The brown solution was stirred for 20 min at rt and quinidine (0.145 g, 0.45 mmol, 0.1 eq.) was added in one portion. The reaction was stirred at rt for 24 h, after which TLC (CH_2Cl_2/MeOH 98:2) showed full conversion. The CH_2Cl_2 was carefully evaporated (30 °C, 32 mbar) yielding a brown oil that was purified by flash chromatography (eluent CH_2Cl_2/MeOH 100:0 to 95:5) to afford product **118** (0.671g, 55% **yield**). $[\alpha]^{23}_{D}$ = +87 (c = 0.82, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.30 (t, *J* = 7.9 Hz, 1H, Ar-*H*,), 6.89 (d, *J* = 7.6 Hz, 1H, Ar-*H*), 6.86-6.84 (m, 1H, Ar-*H*), 6.83 (s, 1H, Ar-*H*), 4.58 (m, 1H, C*H*-OH), 4.10 (d, *J* = 12.0 Hz, 1H, C*H*-N_3), 3.82 (s, 3H, -OC*H*$_3$), 3.46 (td, *J* = 12.1, 4.3 Hz, 1H, Ph-C*H*), 2.83-2.69 (m, 2H, C*H*$_2$CO), 2.27-2.13 (m, 2H, Ph-CH-C*H*$_2$), 1.72 (s, 1H,-O*H*). **^{13}C NMR** (151 MHz, $CDCl_3$) *δ* 202.92, 159.99, 142.09, 130.02, 119.52, 113.59, 112.61, 71.21, 67.88, 55.27, 48.14, 44.70, 39.33. **HRMS** (ESI): exact mass calculated for $C_{13}H_{15}N_3O_3Na$ [$(M + Na)^+$], 284.1011; found 284.1006.

(2*S*,3*S*,5*R*)-2-azido-5-hydroxy-3-(3-methoxyphenyl)cyclohexanone [*ent*-118]



The title compound was synthesized following the above procedure, but (S)-(+)-α,α-diphenyl-2-pyrrolidinemethanol trimethylsilyl ether was used as the catalyst. **[α]$^{23}_{D}$** = -82 (c = 1, MeOH, l = 1 dm).

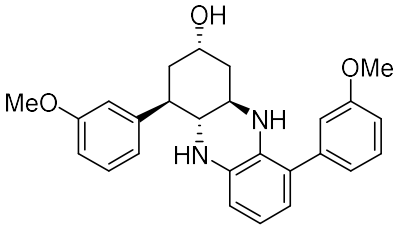
Methyl((1*R*,2*R*,4*S*)-4-hydroxy-2-(3-methoxyphenyl)-6-oxocyclohexyl)carbamate [119]



In a 50 mL RBF, **118** (0.391 g, 1.5 mmol, 1 eq.), dimethyl dicarbonate (0.600 g, 4.5 mmol, 3 eq.) and 10% Pd/C (0.240 g, 2.25 mmol, 1.5 eq.) were suspended in methanol (15 mL). The vessel was sealed and subjected to hydrogen balloon (1 atm) with vigorous stirring for 12 h, after which TLC (CH_2Cl_2/MeOH 95:5) showed full conversion. The suspension was filtered through a celite® pad and evaporated *in vacuo* (20 °C, 0.1 mbar). The translucent grey oil was purified by flash chromatography (eluent CH_2Cl_2/MeOH 100:0 to 97:3) giving the product as a translucent oil carbamate **119** (0.325 g, 88% **yield**) **[α]$^{23}_{D}$** = -23 (c = 2, MeOH, l = 1 dm). *ee*: τmajor = 7.49 min, τminor = 12.3 min (>99.5% ***ee***), AD-H column (150 x 4.6 mm, 5μ), *n*-hexane/*i*-PrOH (80:20) as a mobile phase; flow rate 0.75 mL/min, column temperature 25°C, λ236 nm, sample 1 mg/mL dissolved in the mobile phase. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.24 (t, *J* = 7.9 Hz, 1H, Ar-*H*), 6.86 (d, *J* = 7.5 Hz, 1H, Ar-*H*), 6.80 (d, *J* = 12.7 Hz, 1H, Ar-*H*), 6.78 (dd, *J* = 8.2, 2.3 Hz, 1H, Ar-*H*), 5.25 (d, *J* = 8.7 Hz, 1H), 4.61 (t, *J* = 9.2 Hz, 1H), 4.53 (m, 1H), 3.78 (s, 3H, Ar-OCH_3), 3.48 (s, 3H, NHCOOCH_3), 3.28 (dd, *J* = 19.7, 8.5 Hz, 1H, Ph-C*H*), 3.04-2.95

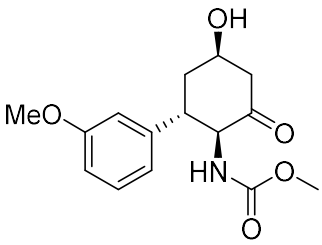
(bs, 1H, O*H*), 2.83 (d, *J* = 13.1 Hz, 1H, COCH_2), 2.70 (d, *J* = 13.9 Hz, 1H, COCH_2), 2.19 (t, *J* = 11.2 Hz, 2H, Ph-CH-CH_2). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 206.01, 159.64, 157.02, 142.30, 129.62, 119.76, 113.56, 112.33, 68.17, 63.17, 55.16, 52.16, 48.19, 45.44, 40.29. **HRMS** (ESI): exact mass calculated for $C_{15}H_{19}NO_5Na$ [(M + Na)$^+$], 316.1161; found 316.1158.

(2*S*,4*R*,4*aR*,10*aR*)-4,9-bis(3-methoxyphenyl)-1,2,3,4,4*a*,5,10,10*a*-octahydrophenazin-2-ol [120]



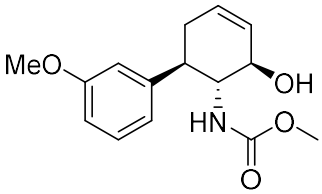
Biproduct **120** (0.062g, 10% **yield**) isolated when using 1 eq. of 10% Pd/C catalyst loading on **118**. **[α]$^{23}_D$** = +97 (c = 1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.40 (t, *J* = 7.9 Hz, 1H, Ar-*H*), 7.31 (t, *J* = 7.8 Hz, 1H, Ar-*H*), 7.03 (d, *J* = 7.5 Hz, 1H, Ar-*H*), 7.00 (s, 1H, Ar-*H*), 6.95 (d, *J* = 8.3 Hz, 1H, Ar-*H*), 6.90 (d, *J* = 7.5 Hz, 1H, Ar-*H*), 6.86 (d, *J* = 8.9 Hz, 1H, Ar-*H*), 6.84 (s, 1H, Ar-*H,*), 6.75 (t, *J* = 7.7 Hz, 1H, Ar-*H*), 6.61 (d, *J* = 7.5 Hz, 1H, Ar-*H*), 6.40 (d, *J* = 7.9 Hz, 1H, Ar-*H*), 4.49-4.29 (bs, 1H, N*H*), 4.15-4.11 (m, 1H, Ph-C*H*), 3.41 (td, *J* = 3.2, 12.0 Hz, 1H, Ph-CH-CH(NH)-C*H*-NH), 2.21 (d, *J* = 14.0 Hz, 1H, Ph-CH-CH_2), 2.13 (d, *J* = 15.0 Hz, 1H, CH(NH)-CH_2), 1.96-1.87 (m, 2H, CH(NH)-CH_2, Ph-CH-CH_2), 1.70 (s, 1H, CH-O*H*). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 159.96 (2C), 144.20, 140.16, 132.64, 130.04, 129.80, 129.20, 127.83, 121.33, 120.60, 120.37, 119.57, 114.03, 113.93, 113.91 (2C), 113.65, 112.34, 67.03, 56.14, 55.28 (2C), 49.52, 39.43, 39.26, 36.68. **HRMS** (ESI): exact mass calculated for $C_{26}H_{28}NO_3Na$ [(M + Na)$^+$], 439.1988; found 439.1978.

methyl ((1*S*,2*S*,4*R*)-4-hydroxy-2-(3-methoxyphenyl)-6-oxocyclohexyl)carbamate (*ent*-119)



The title compound was made from *ent*-azide ***ent*-118**. **$[\alpha]^{23}_D$** = +21 (c = 2, MeOH, l = 1 dm**).** ***ee***: τminor = 7.59 min, τmajor = 12.3 min (>99.5% ***ee***), AD-H column (150 x 4.6 mm, 5μ), *n*-hexane/*i*-PrOH (80:20) as a mobile phase; flow rate 0.75 mL/min, column temperature 25°C, λ236 nm, sample 1 mg/mL dissolved in the mobile phase.

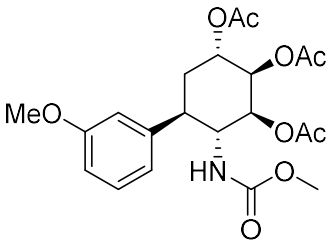
methyl ((1*R*,2*R*,3*R*)-3-hydroxy-3'-methoxy-1,2,3,6-tetrahydro-[1,1'-biphenyl]-2-yl)carbamate [122]



In a 50 mL RBF, carbamate **119** (0.360 g, 1.23 mmol, 1 eq.) was dissolved in CH_2Cl_2 (18 mL) under nitrogen and cooled in an ice bath. Methanesulfonyl chloride (0.123 mL, 1.59 mmol, 1.3 eq.) was added in one portion, and then Hünig's base (0.642 mL, 3.69 mmol, 3 eq.) was added dropwise. The resulting mixture was stirred for 10 h at rt, then poured into water (10 mL) and the layers separated. The organic phase was washed with 1 M HCl (1 mL) then brine (10 mL) and dried over Na_2SO_4. Concentration under reduced pressure gave the crude product as a translucent oil (0.305g, 95% **yield**). This product was used with out further purification. In a 50 mL RBF, enone (0.305 g, 1.10 mmol, 1 eq.) was dissolved in THF (15 mL), cooled to 0 °C, and lithium tri-*tert*-butoxyaluminum hydride (0.846 g, 3.30 mmol, 3 eq.) was added. After 12 h of stirring, saturated aqueous NH_4Cl was added to quench the reaction. The product was extracted with EtOAc (3 x 15 mL) and the combined organic layers were washed with brine (10 mL) and dried over Na_2SO_4. The solvent was removed

in vacuo and the residue was purified by column chromatography (CH_2Cl_2/MeOH; 100:0 to 97:3) to afford the product **122** (0.291 g, 95% **yield**) as a translucent oil. **$[\alpha]^{23}_D$** = -104.3 (c = 1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.24 (t, *J* = 6.9 Hz, 1H, *Ar-H*), 6.79 (m, 2H, Ar-*H*), 6.76 (dt, *J* = 3.9, 1.8 Hz, 1H, Ar-*H*), 5.82-5.75 (m, 1H, CH2-C*H*=), 5.72 (dt, *J* = 3.7, 2.1 Hz, 1H, =C*H*-CH-OH), 4.59 (brs, 1H, NH), 4.33 (brs, 1H, C*H*-OH), 4.25 (brs, -O*H*), 3.85 (ddd, 1H, J = 13.4, 7.8, 6.5 Hz, 1H, C*H*-NH), 3.80 (s, 3H, Ar-OCH_3), 3.55 (s, 3H, HNCOO-CH_3), 2.86 (td, J = 10.3, 6.5 Hz, 1H, Ar-C*H*), 2.42-2.29 (m, 2H, Ar-CH-CH_2). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 160.03, 158.54, 142.65, 130.05, 129.40, 126.92, 120.00, 113.51, 112.57, 74.09, 58.60, 55.22, 52.44, 44.82, 34.96. **HRMS** (ESI): exact mass calculated for $C_{15}H_{19}NO_4Na$ [$(M + Na)^+$], 300.1212; found 300.1208.

(1*S*,2*R*,3*S*,4*R*,5*R*)-4-((methoxycarbonyl)amino)-5-(3-methoxyphenyl)cyclohexane-1,2,3-triyl triacetate [123]



In a 50 mL RBF, allyl alcohol **122** (0.260 g, 0.94 mmol, 1 eq.) was dissolved in CH_2Cl_2 (20 mL) along with $NaHCO_3$ (0.158 g, 1.88 mmol, 2 eq.). To the stirred suspension was added *m*-CPBA (0.422g, 1.88 mmol, 2 eq.) at rt. The resulting suspension was stirred vigorously for 24 h. A 20% (w/v) aqueous solution of sodium sulfite (10 mL) was added, and the resulting two-phase mixture was stirred vigorously for 15 min. The two layers were separated, and the aqueous layer was extracted with CH_2Cl_2 (2 x 10 mL). The combined organic layers were washed with a 20% (w/v) aqueous solution of sodium sulfite (10 mL) and a 5% (w/v) aqueous solution of $NaHCO_3$ (2 x 10 mL), dried with anhydrous Na_2SO_4 and evaporated under reduced pressure (20 °C, 32 mbar) to give

a 4:1 diastereomeric mixture (0.234 g) that was used towards the next step without further purification.

To a solution of epoxides (0.234 g, 0.80 mmol, 1 eq.) in water (5.1 mL) was added sodium benzoate (0.009 g, 0.06 mmol, 0.08 eq.) and was heated at 90 - 95 °C for 16 h. After TLC showed full conversion, the solution was cooled to rt, the water was removed *in vacuo*, and the light pink residue single trihydroxy compound (0. 249 g) was used for next step, without further purification.

To a solution of the trihydroxy compound (0.140 g, 0.80 mmol, 1 eq.) in pyridine (0. 386 mL, 4.80 mmol, 6 eq.) was added acetic anhydride (0.243 mL, 4.80 mmol, 6 eq.). The reaction mixture was stirred at rt for 16 h. After the pyridine was removed *in vacuo* (0.1 mbar). The residue was dissolved in EtOAc (20 mL) and washed with saturated $NaHCO_3$ (2 x 10 mL) and water (10 mL). The solvent was removed *in vacuo* and the product purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford triacetate **123** (0.316 g, over 3 steps **77% yield**). **$[\alpha]^{23}_D$** = -12 (c = 1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.22 (t, *J* = 8.0 Hz, 1H, Ar-*H*), 6.84 (t, *J* = 8.0 Hz, 1H, Ar-*H*), 6.80-6.75 (m, 2H, Ar-*H*), 5.37 (t, *J* = 2.9 Hz, 1H, C*H*-OAc), 5.23 (d, *J* = 9.2 Hz, 1H, C*H*-OAc), 5.15 -4.96 (m, 1H, C*H*-OAc), 4.44 (d, *J* = 33.5 Hz, 1H, N*H*), 4.25 (m, 1H, C*H*NH), 3.80 (s, 3H Ar-OCH_3), 3. 46 (s, 3H, O=COCH_3), 2.95 (m, 1H, Ar-C*H*), 2.38 (s, 3H, O=C-CH_3), 2.17 (s, 3H, O=C-CH_3), 2.17-2.12 (m, 1H, Ar-CH-CH_2), 2.03-2.02 (m, 1H, Ar-CH-CH_2), 2.01 (s, 3H, O=C-CH_3). **HRMS** (ESI): exact mass calculated for $C_{21}H_{27}NO_9Na$ [$(M + Na)^+$], 460.1584; found 460.1578.

2.12.1 References

M. Cherest, H. Felkin, Prudent, N. *Tetrahedron Letters*. **1968**, 2204.

J. Clayden, N. Greeves, S. Warren. *Organic Chemistry*. **2001**, 859.

N. Deora, P. Carlier. *Organic & Biomolecular Chemistry*. **2019**, *17*, 8628.

K. Doitomi, K. Xua, H. Hirao. *Royal Society of Chemistry*. **2017**, 3470.

A. Fürst, A. Plattner. *Helvetica Chimica Acta*. **1949**, 275.

S. R. S. S. Kotti, C. Timmons, G. Li. *Chemical Biology & Drug Design*. **2006**, 101.

T. Kouko, K. Matsumura, T. Kawasaki. *Tetrahedron*. **2005**, 2309.

J. Kroutil, T. Trnka, M. Budesinsky, M. Černý. *European Journal of Organic Chemistry*. **2002**, 2449.

A. Martínez, D. Vega, J. Aguirre, B. Lantaño, G. Moltrasio. *Synthesis*. **2012**, *44*, 125.

J. McNulty, J. Mao, R. Gibe, R. Mo, S. Wolf, G. R. Pettit, D. L. Herald, M. R. Boyd. *Bioorganic & Medicinal Chemistry Letters*. **2001**, 169.

J. McNulty, C. Zepeda-Velazquez. *Angewandte Chemie International Edition*. **2014**, 8450.

D. K. Murphy, R. L. Alumbaugh, B. Rickborn. *The Journal of The American Chemical Society*. **1969**, 2649.

D. J. Pasto, J. A. Gontarz. *Journal of the American Chemical Society*. **1970**, 7480.

O. Revu, C. Zepeda-Velázquez, A. J. Nielsen, J. McNulty, R. H. Yolken, L. Jones-Brando. *ChemistrySelect*. **2016**, 5895.

M. Squillacote, R. S. Sheridan, O. L. Chapman, F. A. L. Anet. *Journal of the American Chemical Society*. **1975**, 3244.

H. Tanimoto, K. Kakiuchi. *Natural Product Communications*. **2013**, 1021.

B. M. Trost, D. L. A. Van Vranken. *Journal of the American Chemical Society*. **1993**, 444.

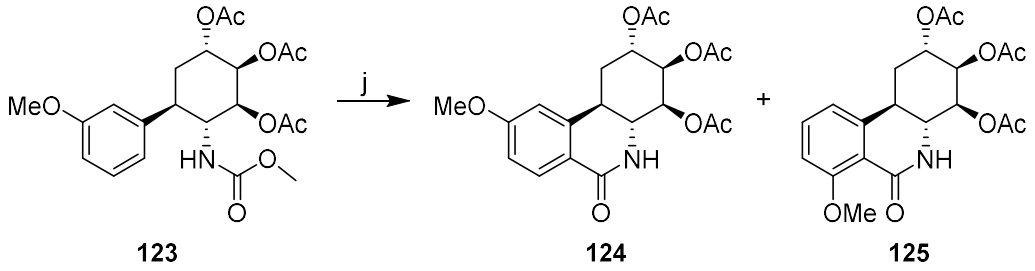
H. Wang, K. N. Houk, D. A. Allen, M. E. Jung. *Organic Letters*. **2011**, 3238.

H. Wang, K. N. Houk. *Chemical Science*. **2014**, 462.

Y. D. Wu, J. A. Tucker, K. N. Houk. *The Journal of the American Chemical Society*. **1991**, 5018.

2.2 Total Synthesis of Four (+)-*trans*-dihydronarciclasine Derivative targets

Succeeding the functionalization of all key requisites pertaining to ring-C, our attention turned to the formation of the phenanthridone ring following BBN reaction conditions (scheme 36). The BBN reaction was used during the later stages of the syntheses on triacetate **123** producing two regioisomeric tricycles, **124** as the major product in a 51% yield, and **125** as the minor product in a 15% yield.

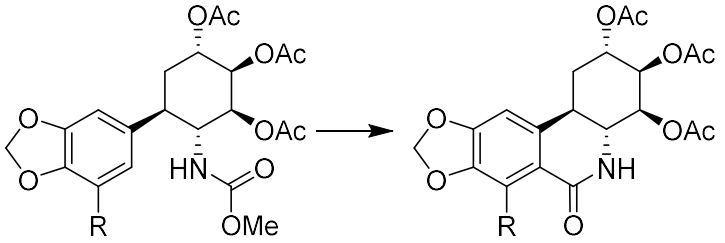


123 **124** **125**

Scheme 36. Synthesis of the phenanthridone skeleton after closure of ring-B via BBN reaction conditions: j) 1M Tf_2O in CH_2Cl_2, DMAP, CH_2Cl_2, O °C - rt, 16 h, 51% **124**, 15% **125**.

The intramolecular annulation proceeded smoothly isolating a 66% overall yield. These results showed similar findings to other substrates under similar reaction conditions (table 5) (Kumar & Studer, 2008; McNulty & Zepeda-Velazquez, 2014).

Table 5. Comparisons to other similar published reports of Banwell's modified Bischler-Napieralski reaction, including yield and conditions.



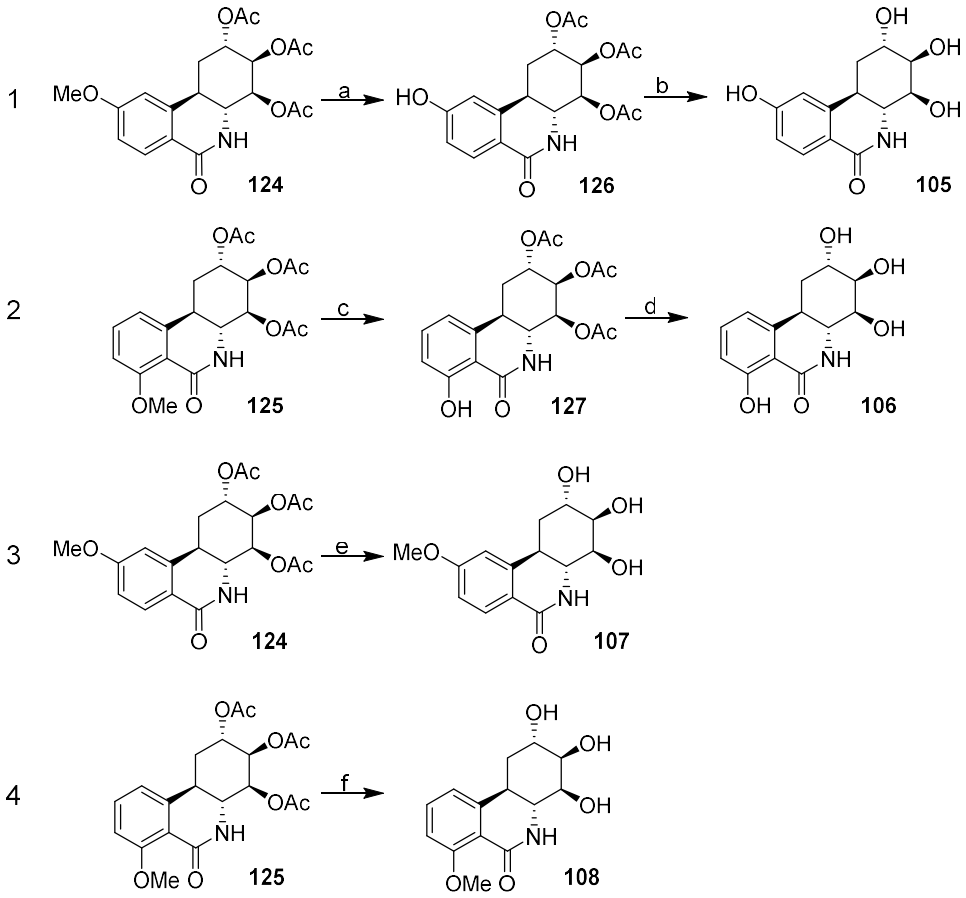
X1: R = OMe
Y1: R = H

X2: R = OMe
Y2: R = H

Entry	Substrate	Product	Tf_2O equiv.	DMAP equiv.	Solvent Conc.	Temp.	Time	Yield	d.r.
1	**X1**	**X2**	8	4.5	25mL, 0.0124 M	0-5°C	24 h	82%	64 : 18
2	**Y1**	**Y2**	5	3	8mL, 0.04M	0°C-rt	16 h	52%	
3	**123**	**124**, **125**	5	3	8mL, 0.04M	0°C-rt	16 h	66%	51 : 15

Our next steps focused on the deprotection of all protecting groups towards the isolation of the four (+)-*trans*-dihydronarciclasine derivatives. Starting from the C9-methyl ether **124**, we were faced with a small number of problems right to begin with. Initial attempts using trimethylsilyl iodide (TMSI) and boron tribromide (BBr_3) did not proceed smoothly, while boron trichloride methyl sulfide complex (BCl_3-DMS) was only successful towards an ester cleavage on ring-C, not producing the phenol moiety for ring-A. Further studies would reveal that when using aluminum chloride with tetra-*n*-butylammonium iodide ($AlCl_3$/TBAI), the phenolic compound **126** would be isolated in an 88% yield. The same reaction conditions were repeated for the cleavage of the C7-methyl ether 1**25,** isolating **127** in an 89% yield. Acetate deprotection using K_2CO_3/MeOH on compounds

124, **125**, **126**, and **127** proceeded smoothly isolating the four antiviral target molecules **5** (96% yield), **7** (96% yield), **6** (94% yield), and **8** (96% yield) respectively.



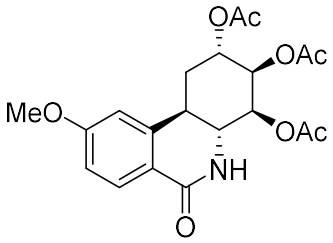
Scheme 37. Completed synthesis of four antiviral *trans*-dihydronarciclasine derivatives*:* 1 - a) $AlCl_3$/TBAI (1:2), CH_2Cl_2/benzene (1:1), 88%; b) K_2CO_3, MeOH, 94%; 2 - c) $AlCl_3$/TBAI (1:2), CH_2Cl_2/benzene (1:1), 89%; d) K_2CO_3, MeOH, 96%; 3 – e) K_2CO_3, MeOH, 96%; 4 – f) K_2CO_3, MeOH, 96%.

2.21 Conclusion

Isolation of our four antiviral target molecules was achieved in high yields following the final deprotection steps, communicating great utility in the synthesis's starting substrate. The syntheses towards two methyl ether antiviral target molecules **107** and **108** were completed in 10 steps, with an overall yield of 13.9% and 4.1% respectfully both with greater than 98% *ee*. While syntheses towards two phenol containing antiviral targets **105** and **106** were completed in 11 steps, with an overall total yield of 12.0% and 3.65% respectfully both with greater than 98% *ee*. The value observed in the asymmetric organocatalytical [3+3] sequence delivering the densely substituted ring-C provided great utility towards the production of all four antiviral target molecules. This minimized the number of steps required to furnish such a complex moiety in the targets, which upon functionalizing in stereospecific manners enabled the full construction of the aminocyclitol core. This sequence has provided effective regio, diastereo, and enantioselective access to ring-A modified, fully functionalized rings-B/C *trans*-dihydronarciclasine derivatives. Success in this regard, has now expanded the growing library of derivatives related to (+)-*trans*-dihydronarciclasine from precursors α-azidoacetone and *m*-anisaldehyde. The compounds have been sent away for antiviral analyses at John Hopkins, and we await their communication before further disclosure of the effects seen with the four antiviral targets of (+)-*trans*-dihydronarciclasine.

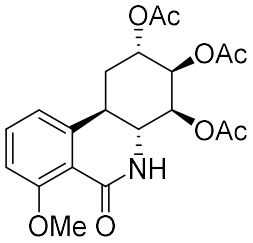
2.22 Experimental

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-9-methoxy-6-oxo-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [124]



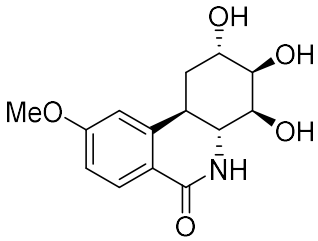
Into a 50 mL RBF, **123** (0.140 g, 0.32 mmol, 1 eq.) and DMAP (0.117 g, 0.96 mmol, 3 eq.) were dissolved in CH_2Cl_2 (8 mL) at 0 °C. A 1.0 M solution of Tf_2O in CH_2Cl_2 (1.597 mL, 1.60 mmol, 5 eq.) was added dropwise to the reaction mixture over a period of 10 min. The reaction was stirred for 16 h at rt. The solvent was evaporated, and the residue was treated with a mixture of THF (5 mL) and 1 M HCl (1 mL). After stirring for 1 h at rt, the mixture was partitioned between a saturated aqueous solution of $NaHCO_3$ (1 mL) and CH_2Cl_2 (20 mL). The organic phases were combined, dried with anhydrous Na_2SO_4 and concentrated. The Regio isomers **124** and **125** were purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2). The major regio isomer isolated was the 9-methoxy Banwell product **124 (**0.066 g, 51% **yield**), $\mathbf{R_f}$ = 0.35 (HEX/EA; 1:1); $\mathbf{[\alpha]^{23}_D}$ = +34 (c = 0.1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) **δ** 8.096 (d, J = 8.65 Hz, 1H), 6.939 (dd, J = 8.54, 2.09 HZ, 1H), 6.778 (m, 1H), 6.582 (s, 1H), 5.513 (t, J = 2.94 HZ, 1H), 5.254 (m, 2H), 3.884 (s, 3H), 3.855 (dd, J = 11.1, 12.4 HZ, 1H), 3.287 (d, J = 3.74, 12.23 HZ, 1H), 2.561 (dt, J = 3.3, 14.6 HZ, 1 H), 2.70 (s, 3H), 2.126 (s, 3H), 2.088 (s, 3H), 2.008 (m, 1H); **^{13}C NMR** (151 MHz, CDCl3) δ 170.27, 169.46, 169.20, 166.13, 163.35, 141.90, 130.75, 121.32, 112.07, 109.67, 77.23, 77.02, 76.81, 71.76, 68.67, 67.51, 55.53, 52.69, 35.02, 26.45, 21.08, 20.85, 20.72. **HRMS** (ESI): exact mass calculated for $C_{20}H_{23}NO_8Na$ [(M + Na)+], 428.1321; found 428.1316.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-7-methoxy-6-oxo-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [125]



Minor product (0.019 g, 15% **yield**), **Rf:** 0.30 (HEX/EA 1:1), **[α]^{23}D** = +41 (c = 1, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.50 (m, 1H), 7.00(d, J = 8.57 HZ, 1H), 6.869 (d, J = 7.76 HZ, 1H), 6.386 (s, 1H), 5.441 (t, 3H), 5.219 (m, 2H), 3.957 (s, 3H), 3.746 (t, J = 11.35 HZ, 1H), 3.223 (td, J = 3.98, 12.6 HZ, 1H), 2.545 (dt, J = 3.4, 14.3 HZ, 1H), 2.163 (s, 3H), 2.08 (s, 3H), 2.06 (s, 3H), 1.962 (m, 1H). **^{13}C NMR** (151 MHz, $CDCl_3$) δ 170.39, 169.46, 169.19, 164.37, 160.35, 142.56, 133.39, 117.27, 115.42, 111.50, 71.66, 68.66, 67.46, 56.28, 52.01, 35.87, 26.79, 21.08, 20.78, 20.72. **HRMS** (ESI): exact mass calculated for $C_{20}H_{23}NO_8Na$ [(M + Na)+], 428.1321; found 428.1314.

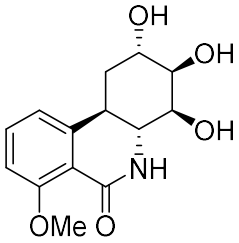
(2*S*,3*R*,4*S*,4*aR*,10*bR*)-2,3,4-trihydroxy-9-methoxy-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one [107]



9-Methoxy Banwell synthesized compound **124 (**0.005 g, 0.012 mmol, 1 eq.) and potassium carbonate (0.002 g, 0.01 mmol, 0.8 eq.) were dissolved in MeOH (1 mL) and stirred at rt. TLC analysis showed full conversion after 6 h. The mixture was concentrated under a flow of N_2. The white product was dissolved in 9:1 CH_2Cl_2/MeOH and filtered through a pad of silica to afford **107** (0.0032 g, 96% **yield**). **[α]^{23}D** = +52 (c = 0.25, MeOH, l = 1 dm). **^{1}H NMR** (700 MHz, DMSO) δ 7.81 (d, *J* =8.5 Hz, 1H), 6.90 (d, *J* = 8.5, Hz, 1H), 6.87 (s, 1H), 6.82 (s, 1H), 5.14 (bm, 3H), 3.91 (d, *J* = 2.4 Hz, 1H), 3.82 (s, 3H), 3.76-3.71 (m, 2H), 2.95 (dd, *J* = 11.9, 9.3 Hz, 1H), 2.18 (dt, *J* = 13.0, 3.2 Hz, 1H), 1.70 (td, *J* = 13.0, 2.2 Hz, 1H). **^{13}C NMR** (176 MHz, DMSO) δ 164.60, 162.35, 144.41,

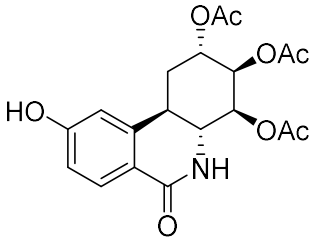
129.45, 122.07, 111.69, 109.23, 71.72, 69.89, 68.66, 55.37, 55.07, 40.06, 34.53, 28.03. **HRMS** (ESI): exact mass calculated for $C_{14}H_{17}NO_5Na$ [(M + Na)+], 302.1004; found 302.0986.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-2,3,4-trihydroxy-7-methoxy-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one [108]



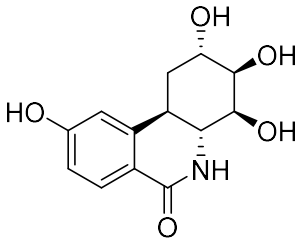
7-Methxoy Banwell synthesized compound **125** (0.005 g, 0.012 mmol, 1 eq.) and potassium carbonate (0.002 g, 0.01 mmol, 0.8 eq.) were dissolved in MeOH (1 mL) and stirred at rt. TLC analysis showed full conversion after 6 h. The mixture was concentrated under a flow of N_2. The white product was dissolved in 9:1 CH_2Cl_2/MeOH and filtered through a pad of silica isolating trihydroxy **108** (0.0032 g, 96% yield). **$[\alpha]^{23}_D$** = +56 (c = 0.25, DMSO, l = 1 dm). **^{1}H NMR** (600 MHz, DMSO) δ 7.47 (s, 1H, CON*H*), 7.45 (t, *J* = 8.0 Hz, 2H, Ar-*H*), 7.02 (d, *J* = 8.5 Hz, 1H, Ar-*H*), 6.91 (d, *J* = 7.8 Hz, 1H, Ar-*H*), 3.91 (dd, *J* = 3.2, 6,25 Hz, 1H, CH_2CH-OH), 3.79 (s, 3H, Ar-OMe), 3.77-3.75 (m, 1H, $CH_2CHOHCHOH$), 3.25-3.19 (dd, *J* = 10.2, 3.0 Hz, 1H, CHNHC*H*OH), 2.85 (td, *J* = 12.3, 3.8 Hz, 1H,), 2.11 (dd, *J* = 10.0, 3.2 Hz, 1H), 1.67 (td, J= 3.2 13.7 Hz, 1H). **^{13}C NMR** (151 MHz, DMSO) δ 163.04, 159.12, 145.01, 132.35, 118.24, 115.49, 111.17, 71.70, 69.40, 68.63, 55.70, 54.73, 35.55, 28.55. **HRMS** (ESI): exact mass calculated for $C_{14}H_{17}NO_5Na$ [(M + Na)$^+$], 302.1004; found 302.0988.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-9-hydroxy-6-oxo-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [126]



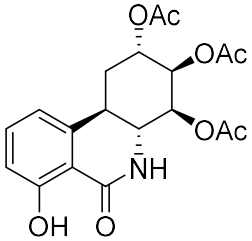
124 (0.050 g, 0.123 mmol, 1 eq.) was dissolved in 1:1 ratio of dry CH_2Cl_2 and dry Benzene. After, the addition of Aluminium chloride (0.049 g, 0.369 mmol, 3 eq.) under argon at 0 °C, *n*-$Bu_4N^+I^-$ (0.27g, 0.738 mmol, 6 eq.) was added under argon to the reaction mixture over a period of 10 min at 0 °C. Reaction mixture turned to a red colour and stirred for 3 h at rt. After confirming with TLC, reaction was quenched with water, and stirred with 2 M HCl for 0.5 h, and then extracted with EtOAc (3 x 10 mL). The organic layers were washed with brine, dried over Na_2SO_4, and filtered through Dowex® 50WX8 hydrogen form. The mixture was concentrated *in vacuo* and purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford the **126** (0.042g, 88% **Yield**). **$[\alpha]^{23}_D$** = +45 (c = 0.9, MeOH, l = 1 dm). **^{1}H NMR** (700 MHz, $CDCl_3$) δ 7.99 (t, *J* = 7.8 Hz, 1H, ArH), 6.86 (dd, *J* = 8.4, 1.9 Hz, 1H, ArH), 6.85-6.77 (bs, 1H, CON*H*), 6.75 (s, 1H, Ar*H*), 5.46 (t, *J* = 3.1 Hz, 1H, C*H*-OAc), 5.23 (dd, *J* = 10.8, 2.9 Hz, 1H, C*H*-OAc), 5.21 (dd, *J* = 5.9, 2.8 Hz, 1H, C*H*-OAc), 3.85 (dd, *J* = 12.5, 11.2 Hz, 1H C*H*-N), 3.67 (dd, *J* = 25.2, 9.1 Hz, 1H, Ph-O*H*), 3.24 (td, *J* = 12.7, 3.5 Hz, 1H, Ar-C*H*), 2.50 (dt, *J* = 14.4, 3.0 Hz, 1H, CH_2), 2.15 (s, 3H, COCH_3), 2.11 (s, 3H, COCH_3), 2.08 (m, 3H, COCH_3) 1.98-1.93 (m, 1H, CH_2). **^{13}C NMR** (176 MHz, $CDCl_3$) δ 170.27, 169.46, 169.17, 166.72, 161.01, 142.60, 131.19, 114.66, 114.66, 110.94, 71.48, 68.53, 67.46, 52.78, 34.69, 26.40, 21.07, 20.84, 20.71. **HRMS** (ESI): exact mass calculated for $C_{19}H_{21}NO_8Na$ [$(M + Na)^+$], 414.1165; found 414.1156.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-2,3,4,9-tetrahydroxy-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one [105]



127 (0.0047 g, 0.012 mmol, 1 eq.) and K_2CO_3 (0.002 g, 0.010 mmol, 0.8 eq.) in MeOH (1 mL) were stirred at rt until a white solid precipitated. TLC analysis showed full conversion after 6 h. The mixture was concentrated under a flow of N_2, leaving a white product that was dissolved in 9:1 CH_2Cl_2/MeOH and filtered through a pad of silica. The white solid was recrystallized from MeOH to afford trihydroxy **105** (0.003 g, 94% yield). **$[\alpha]^{23}_D$** = + 31 (c = 0.25, MeOH, l = 1 dm). **^{1}H NMR** (700 MHz, DMSO) δ 7.75 (d, *J* = 8.54 Hz, 1H), 6.75(bs, 1H), 6.74 (dd, *J* = 8.54, 2.3 Hz, 1H), 6.71 (s, 1H), 3.94 (dd, *J* = 3.1, 5.6 Hz, 1H), 3.77-3.74 (m, 2H), 3.36 (dd, *J* = 12.4, 9.4 Hz, 1H), 2.94 (td, *J* = 12.4, 3.5 Hz, 1H), 2.13-2.08 (dt, *J* = 3.5, 13.2 Hz, 1H), 1.70 (td, *J* = 13.2, 2.5 Hz, 1H). **^{13}C NMR** (176 MHz, DMSO) δ 165.28, 161.45, 144.84, 130.03, 120.93, 113.74, 110.61, 72.14, 70.37, 69.10, 55.42, 34.78, 28.55. **HRMS** (ESI): exact mass calculated for $C_{13}H_{15}NO_5Na$ [$(M + Na)^+$], 288.0848; found 288.0842.

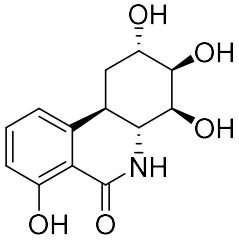
(2*S*,3*R*,4*S*,4*aR*,10*bR*)-7-hydroxy-6-oxo-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [127]



125 (0.005 g, 0.012 mmol, 1 eq.) was dissolved in 1:1 ratio of dry CH_2Cl_2 (0.5 mL) and dry Benzene (0. 5 mL). After, the addition of Aluminium chloride (0.005 g, 0.037 mmol, 3 eq.) under argon at 0 °C, and *n*-$Bu_4N^+I^-$ (0. 0.027 g, 0.074 mmol, 6 eq.) was added also under argon slowly to the reaction mixture over a period of 10 min. Reaction mixture turned to a red colour and was allowed to stir for an additional 1 h at rt. After confirming with TLC,

reaction mixture was quenched with water, stirred with 2 M HCl for 0.5 h, and was extracted with EtOAc (3 x 10 mL). The organic layers were washed with brine, dried over Na_2SO_4, filtered through Dowex® 50WX8 hydrogen form and concentrated *in vacuo*. It was purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford 7-phenol triacetate **127** (0.0043 g, 89% yield). **$[\alpha]^{23}_D$** = +45 (c = 0.5, MeOH, l = 1 dm). (2S,3R,4S,4aR,10bR)-7-hydroxy-6-oxo-1,2,3,4,4a,5,6,10b-octahydrophenanthridine-2,3,4-triyl triacetate 9-phenol (7-phenol) **^{1}H NMR** (700 MHz, $CDCl_3$) δ 12.21 (s, 1H Ph-O*H*), 7.41 (t, *J* = 7.75 Hz, 1H, Ar-*H*), 6.91 (d, *J* = 8.4 Hz, 1H), 6.70 (d, *J* = 7.6 Hz, 1H, Ar-*H*), 5.98 (s, 1H, CON-*H*), 5.47-5.44 (m, 1H, C*H*-OAc), 5.23-5.19 (m, 2H, C*H*-OAc, C*H*-OAc), 3.84 (dd, *J* = 12.7, 10.9 Hz, 1H, C*H*-N), 3.21 (td, *J* = 12.7, 3.7 Hz, 1H, Ph-C*H*), 2.53 (d, *J* = 14.5 Hz, 1H, CH_2), 2.15-2.14 (m, 3H, COCH_3), 2.10 (d, *J* = 5.3 Hz, 3H, COCH_3), 2.08 (m, 3H, COCH_3), 1.97 (ddd, *J* = 15.0, 9.5, 3.1 Hz, 1H, CH_2). **^{13}C NMR** (176 MHz, $CDCl_3$) δ 170.18, 170.15, 169.37, 169.18, 162.14, 140.17, 135.01, 116.91, 113.87, 110.69, 71.84, 68.55, 67.38, 52.72, 34.62, 26.53, 21.05, 20.80, 20.71. **HRMS** (ESI): exact mass calculated for $C_{19}H_{21}NO_8Na$ [$(M + Na)^+$], 414.1165; found 414.11564.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-2,3,4,7-tetrahydroxy-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one [106]



105 (0.0043 g, 0.011 mmol, 1 eq.) and K_2CO_3 (0.002 g, 0.010 mmol, 0.8 eq.) were dissolved in MeOH (1 mL) and stirred at rt until a white solid precipitated. TLC analysis showed full conversion after 6 h. The mixture was concentrated under a flow of N_2. The white product was dissolved in 9:1 CH_2Cl_2/MeOH and filtered through a pad of silica. The white solid was recrystallized from methanol to afford trihydroxy **106** (0.0026 g, 96% **yield**). **$[\alpha]^{23}_D$** = + 28

(c = 0.25, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, CD_3OD) δ 8.45 (s, 1H, Ph-O*H*, 80% was deuterated), 7.29 (t, *J* = 7.81 Hz, 1H, Ar*H*), 6.70 (d, *J* = 7.7 Hz, 1H, ArH), 6.68 (d, *J* = 8.3 Hz, 1H, ArH), 4.00 (dd, *J* = 2.9, 6.3 Hz, 1H, CH_2CHOH), 3.84-3.82 (m, 1H, $CH_2CHCHOH$), 3.79 (dd, *J* = 10.3, 2.8 Hz, 1H, C*H*OH), 3.44-3.38 (dd, *J* = 10.3, 12.7 Hz, 1H, C*H*NH), 2.98 (td, *J* = 11.0, 3.69 Hz, 1H, PhC*H*), 2.20 (dt, *J* = 13.4, 3.19 Hz, 1H, CH_2), 1.77 (m, 1H, CH_2). **^{13}C NMR** (151 MHz, DMSO) δ 169.54, 160.95, 143.10, 134.27, 115.03, 113.83, 111.02, 71.69, 69.34, 68.45, 55.32, 33.75, 28.05. **HRMS** (ESI): exact mass calculated for $C_{13}H_{15}NO_5Na$ [$(M + Na)^+$], 288.0848; found 288.0840.

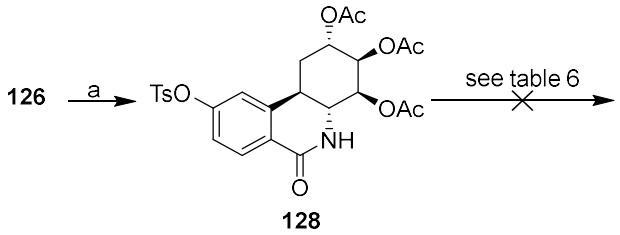
2.22.1 References

C. Kumar, J. A. Studer. *Chemistry – A European Journal*. **2008**, 6326.

J. McNulty, C. Zepeda-Velazquez. *Angewandte Chemie International Edition*. **2014**, 8450.

2.3 Total Synthesis of (+)-*trans*-Dihydrolycoricidine Analogue Target

With access to our four antiviral targets secured, compound **126** was used towards the synthesis of our final target molecule **104**. As previously mentioned in chapter 2.1, the change in starting material was also based around the cross-coupling partners for the Sonogashira reaction towards the C-9 alkyne tagged *trans*-dihydrolycoricidine analogue **104**. Our initial attempts were directed around the cross-coupling between stable **128** (see chapter 1.6) and TMSA (scheme 38, table 6).



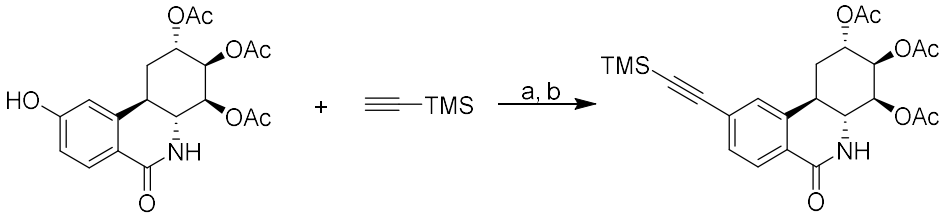
Scheme 38. Initial attempts using stable **128** towards synthesizing analogue **104**: a) TsCl, Py, CH_2Cl_2, rt, 16 h, 95%.

Phenolic compound **126** was activated to the corresponding tosylate **128** smoothly via tosyl chloride and pyridine in a 95% yield. In contrast, the succeeding Sonogashira cross-coupling was very problematic, **128** was inert to all reaction conditions examined, leaving the envisioned leaving group nonlabile and palladium unable to perform oxidative addition between the aryl-OTs bond with only the recovery of starting material observed (table 6).

Table 6. Reaction conditions for the copper-cocatalyzed Sonogashira reaction involving tosylate **128** and triflate.

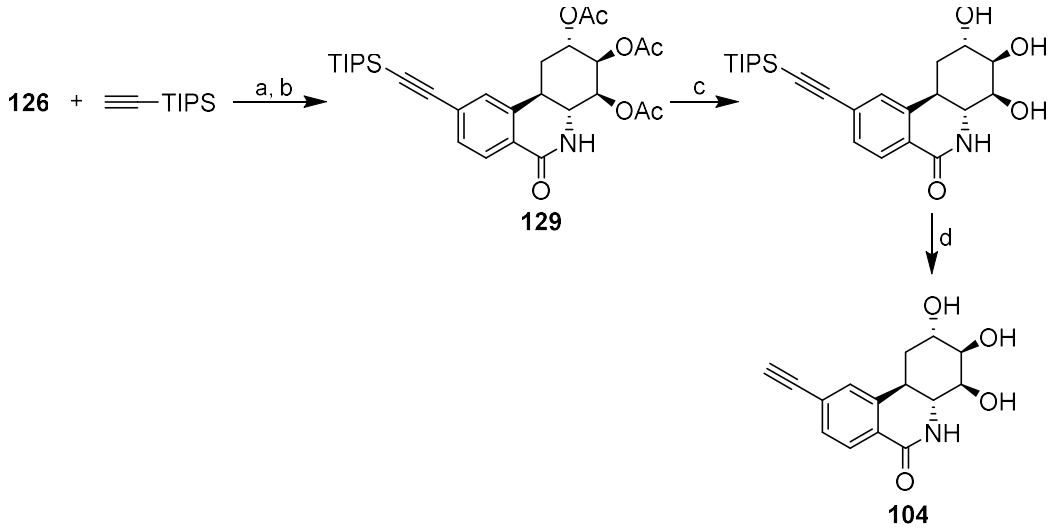
Reagent	Alkyne Reagent	Solvent	Temp.	Catalysts	Base	Result
R-OTs	TMS-acetylene	THF	rt – 70 °C	CuI, $PdCl_2(PPh_3)_2$	DIPA	N.R.
R-OTs	TMS-acetylene	Toluene	rt – 90 °C	CuI, $PdCl_2(PPh_3)_2$	DIPA	N.R.
R-OTs	TMS-acetylene	THF	rt – 70 °C	CuI, $PdCl_2(PPh_3)_2$	TEA	N.R.
R-OTs	TMS-acetylene	THF	rt – 70 °C	CuI, $PdCl_2(PPh_3)_2$	Pyrrolidine	N.R.
R-OTs	TMS-acetylene	THF	rt – 70 °C	CuI, $PdCl_2(PPh_3)_2$	DIPEA	N.R.
R-OTs	TMS-acetylene	THF	rt – 70 °C	CuI, $Pd(PPh_3)_4$	DIPA	N.R.
R-OTf	TMS-acetylene	THF	rt – 40 °C	CuI, $PdCl_2(PPh_3)_2$	DIPA	10% unstable TMS alkyne
R-OTf	TIPS-acetylene	THF	rt – 40 °C	CuI, Cy_3P, $PdCl_2(PPh_3)_2$	DIPA	91% stable TIPS alkyne

An alternative cross-coupling reagent was prepared (scheme 39), designed to increase the reactivity and show greater opportunity for successful oxidative insertion by palladium(0) (see chapter 1.6). Efficient cross-coupling reactivity is observed with aryl triflates (Kanwal, Mujahid, Rasool, Rizwan, & Malik, 2020) setting up more reliable reaction conditions, while also having an activating substituent on the aryl ring aiding towards palladium's first step in the catalytic cycle. We first reacted phenol **126** with triflic anhydride and pyridine producing the corresponding triflate (scheme 39). Without characterizing the C9-triflate due to instability concerns, subsequent coupling with TMSA led to the production of an unstable product losing all material due to degradation.



Scheme 39. Synthesis of unstable Sonogashira product: a) Tf_2O, Py, CH_2Cl_2, 12 h; b) CuI, Cy_3P, $PdCl_2(PPh_3)_2$, THF:DIPA (7:3).

We then prepared a more stable coupling partner (Triisopropylsilyl)acetylene (TIPSA) and reacted phenol **126** with triflic anhydride and pyridine synthesizing the corresponding triflate as previously performed (scheme 40). Subsequent removal of pyridine *in vacuo* and partitioning with EtOAc the unstable triflate was used immediately in the next step without further purification. Finally following optimized Sonogashira conditions, **126** was transformed into **129** in a 91% yield over two steps with TIPSA.



Scheme 40. Synthesis of *trans*-dihydrolycoricidine analogue **104**: a) Tf_2O, Py, CH_2Cl_2, 16 h; b) CuI, Cy_3P, $PdCl_2(PPh_3)_2$, THF:DIPA (7:3), 3 h, 91% over two steps **129**; c) K_2CO_3, MeOH, rt, 95%; d) TBAF, THF, 0.5 h, 95% **104**.

2.31 Conclusion

The concise total synthesis exemplifies the use of *m*-anisaldehyde derivatives as precursors towards the isolation of multiple target molecules, expanding the scope of studies to a growing library of unnatural products. In this last case only one analogue was prepared but following additional investigations control over a variety of additional target molecules can be accomplished. The syntheses presented effective routes towards five new unnatural alkaloids of the isocarbostyril group, while incorporating key steps with the [3+3] Michael aldol sequence, a BBN intramolecular cyclization towards the synthesis of two additional antiviral target molecules, and featured a unique late stage copper-cocatalyzed Sonogashira reaction towards the final target molecule for ATRI purposes. The alkynylation reactions involved following Sonogashira reaction conditions were successful towards the production of arylalkynes showing us the greater use of precursor *m*-anisaldehyde **115**. Although our initial attempts were futile, the generation of a stable tosylate **128** was our intended route because this compound could be characterized before continuing further and was less prone to degradation. Regardless of other procedures using this reactive species in their cross-coupling procedures, the isolation at the least communicated the production of a more active partner for the ensuing transmetalation. Such access allowed us to control our transformation via the production of a more labile cross-coupling triflate partner. Overall, we were successful in the isolation of (+)-*trans*-dihydrolycoricidine analogue **104** in 14 steps, with an overall yield of 10.5% with a greater than 98% *ee*.

2.32 Future Work

2.32A Analysis of the BBN Studies (1)

With the syntheses proceeding smoothly after working around two unforeseen complications, my most strategic steps moving forward would involve optimization of the lowest yielding sequence in generating the phenanthridone skeleton. Although a moderate yielding process, the BBN reaction did present the lowest yielding step of the synthesis. When looking at the results from previous studies by McNulty, as well as from other groups, ring-C and the level at which it is functionalized effects the production of the ensuing lactam (compounds reflect N-moc protected precursors) (Hudlický, et al., 2002; McNulty & Zepeda-Velazquez, 2014).

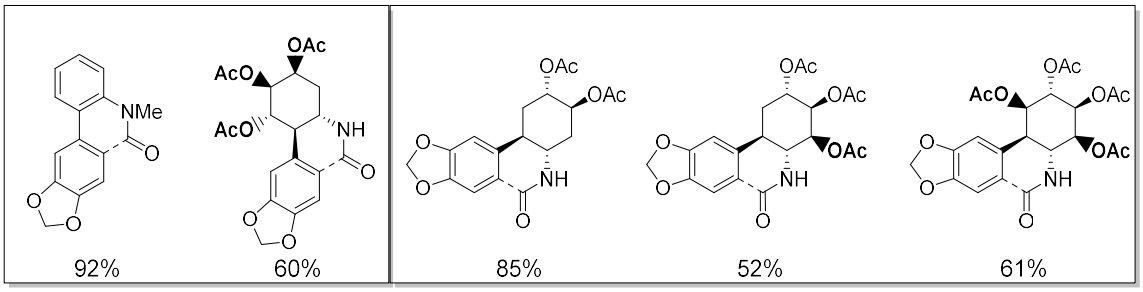
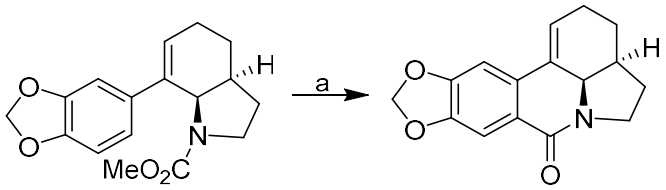


Figure 12. Comparison of results from the BBN reaction. Bonds/atoms shown in bold identify the additional functionalizations present.

Comparing these results with the total synthesis of (±)-γ-lycorane from Banwell et. al. (Banwell, Harvey, & Hockless, 2000), less functionalization has provided higher yields.



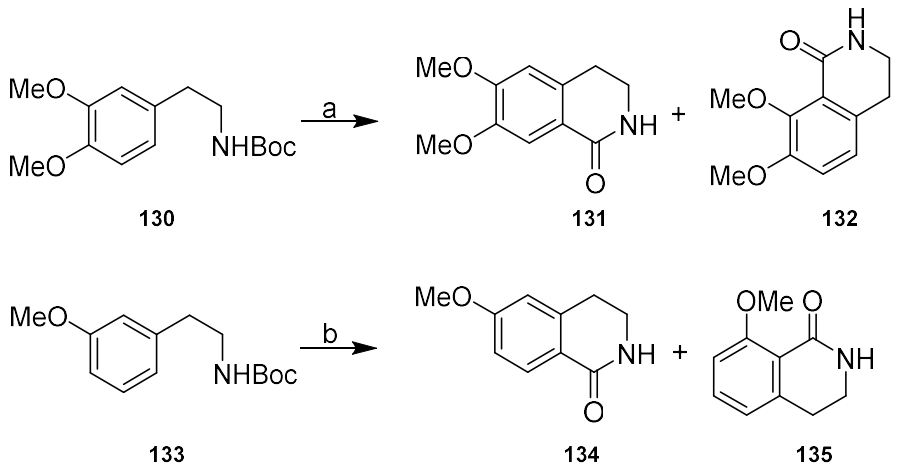
Scheme 41. BBN reaction performed in the total synthesis of (±)-γ-lycorane being the third last step by Banwell: a) Tf_2O, DMAP, CH_2Cl_2, 0 °C, 2 h, 85%.

Frequently this has been encountered with similar substrates (scheme 41, 42), and it is to show that even when comparing to other studies with similar structures this trend can be seen. But it is important to notice the pattern in the reactions performed to mark their weaknesses/limitations, to then analyze alternative routes which may provide increased efficacy. Their studies first attempted the BN reaction but observed double-bond migration leading to a mixture of products. This issue was mitigated following the improved BBN conditions using 5 eq. of triflic anhydride and 3.1 eq. DMAP isolating their cyclized product in an 85% yield, again showing the increased efficacy this modification has developed. An alternative route to pursue would be to close ring-B prior to the reduction step of the enone (Martínez, Vega, Aguirre, Lantaño, & Moltrasio, 2012; Ohta & Kimoto, 1975), and not having to use the lowest yielding step during the final few steps in the synthesis. Although this step is commonly featured during the final steps in many syntheses of Amaryllidaceae alkaloids, interest in developing a more refined route would be of my interest. Studies may elicit C4a epimerization or aromatization of ring-C so optimizations would have to be carefully performed and monitored to prevent this from occurring if possible. If such unwanted by-products are spontaneously formed with no control over, other reaction variables (equivalents, concentration, reaction time) may be best to adjust rather than a change in the order of steps altogether. Additionally, alternative protecting

groups, or lower temperatures can as well be evaluated, which may deliver increased efficacy.

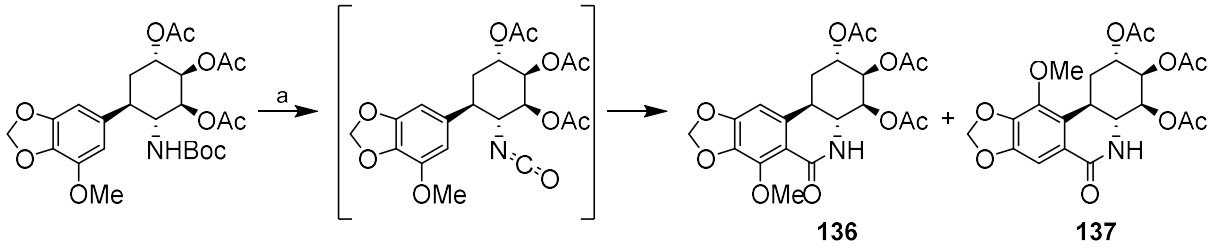
2.32B Analysis of the BBN Studies (2)

An alternative route via the formation of isocyanate intermediates towards in situ Friedel-Crafts-type cyclizations has been studied towards the synthesis of ring-B (but without a fully functionalized ring-C) (In, Hwang, Kim, Seo, & Kim, 2013). The reaction conditions required by a study performed by Kim et. al. were milder than the ones applied towards the BBN reaction and included increased regioselectivity when using unsymmetrical aryl substrates. Enhanced efficacy was also observed with studies by the addition of a Lewis acid enabling a more unperturbed Friedel-Craft cyclization of the isocyanate intermediate to take place. Such routes could be explored to investigate access to higher ratios of regioisomeric products than the ones obtained in these syntheses (3.4:1). Increased yields/regioselectivity were rationalized through the comparison of the intermediates involved in the ring closing step, and how with use of isocyanate intermediates provide a less sterically inhibited species than the imino triflate intermediate seen in traditional BBN reactions. Optimized conditions utilized an N-Boc carbamate with triflic anhydride (1.1 eq.) and 2-chloropyridine (1.5 eq.) as their mild base between temperatures of –78 °C to room temperature.



Scheme 42. Substrate scope using the refined procedures include the reaction with **130**: a) Tf_2O (1.1 eq.), 2-ClPy (1.5 eq.), CH_2Cl_2, -78 °C - rt, **131** 82.9%, **132** 4.1%; and **133**: b) Tf_2O (1.1 eq.), 2-ClPy (1.5 eq.), $BF_3{\cdot}Et_2O$ (5.0 eq.), CH_2Cl_2, -78 °C - rt, **134** 78.6%, **135** 4.4%.

Results from the stereocontrolled total synthesis of (+)-*trans*-dihydronarciclasine (scheme 43) communicated good regioselectivities when following these adjusted conditions, providing high yields after optimization (Hwang, Kim, & Kim, 2012).

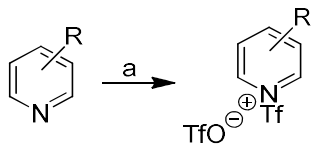


Scheme 43. Increased regioselectivity by Friedel–Crafts-type cyclization towards the production of ring-B via an isocyanate intermediate derived from an N-Boc group: a) Tf_2O, 2-ClPy, CH_2Cl_2, –78 °C to 35 °C, 20 h, 76% **136**, 6% **137**.

The reaction was set-up by adding triflic anhydride (1.1 eq.) and 2-chloropyridine (1.5 eq.) at -78 °C and letting stir for 30 min, then allowing the remaining time to proceed at 35 °C. Such procedures could be applied to our systems, to investigate the change in reactivities observed.

2.32C Analysis of the BBN Studies (3)

As introduced in chapter 5.1, White and colleagues (White, Mewald, & Movassaghi, 2014) investigated the BBN reaction performing in-depth studies related to the pyridines used, and the effects electron rich versus electron poor bases provided. The previous report utilized 2-chloropyridine, that was labelled "unique" from White's study due to its resistance to undergo N-sulfonylation under reaction conditions (scheme 44) (Movassaghi & Hill, 2006). This characteristic was important for the tertiary lactam substrates his group studied, because of the competing unwanted deactivations seen with electron rich pyridines.



Scheme 44. N-sulfonylation of pyridine derivatives. When R = H, N-sulfonylation as shown occurs, but when R = 2-Cl no N-sulfonylation is present: a) Tf_2O, CH_2Cl_2, -78 - 23 °C.

Minimization of this unwanted set-back may be necessary to investigate upon reaching further results from said reactions, because in some instances the electron-rich base completely shut down the lactam activation pathway. Electron-rich pyridines can react with triflic anhydride even at temperatures as low as -78 °C, while the combination of steric hindrance and an electron-withdrawing group substituent can eradicate this from taking place ie. 2-chloropyridine, and why it was labelled "unique".

2.32D Starting Material Selection & New Target Molecules

Although the syntheses presented were carried out in good yields, an alternative precursor for the total synthesis of the C9 *trans*-dihydrolycoricidine analogue would provide additional targets for additional studies. If I were to design another synthesis, I would be interested in starting from benzene-1,3-dicarbaldehyde. My reasoning behind this would be to start the synthesis with a protection step of one of the aldehyde groups. A step including the formation of a cyclic thiolacetal which can be manipulated in the later steps of the synthesis is an example. Deprotection of a 1,3-dithiane protecting group regenerates the aldehyde and can then be transformed to the alkyne via the Bestmann–Ohira reagent producing our terminal alkyne (Dhameja & Pandey., 2018). This route would eliminate the need for cross-coupling reactions and open new routes towards new target molecules. Pursuing this approach would provide an additional electrophilic center towards the production of more C9 analogues (aldehyde). Staying on topic with the C9 alkyne-tagged target, an example towards this use would be the azide-alkyne Huisgen cycloaddition (Amblard, Cho, & Schinazi, 2009; Huisgen, 1963; Huisgen, 1963; Huisgen, 1963; Huisgen, 1963; Gothelf & Jorgenson, 1998; Rostovtsev, Green, Fokin, & Sharpless, 2002) towards the development of a 1,2,3-triazole moiety, or the application of a homogeneous silver(I) catalyst for the cycloaddition of azides onto terminal alkynes delivering the corresponding substituted 1,4-triazole unit (McNulty & Keskar, 2012). These last two examples are additional analogues that may be pursued after pushing more material forward. These targets possessing triazole moieties have been shown to have active pharmacological properties which is the driving force for synthesizing these new target molecules. Another

attractive feature is the use of click chemistry, where reaction conditions have been conducted under ambient conditions. An example of an active compound with a triazole moiety is shown in figure 13 (Kharb, Sharma, & Yar, 2011; Bohacek & Guida, 1996).

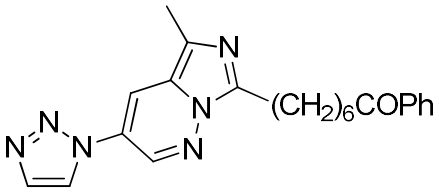


Figure 13. Anti-HIV active compound featuring an 1,2,3-triazole moiety.

The 1,2,3-triazole moieties show a lot of promise because they are stable from metabolic degradation and capable of hydrogen bonding which is a favorable feature for the binding of biomolecular targets, while also showings good solubility measures (Dalvie, Kalgutkar, Khojasteh-Bakht, Obach, & O'Donnell, 2002; Horne, Yadav, Stout, & Ghadiri, 2004). Other potential pharmaceuticals based around 1,2,3-triazoles include the anticancer compound carboxyamidotriazole (CAI) (Soltis, et al., 1996), the nucleoside derivative non-nucleoside reverse transcriptase inhibitor *t*-butyldimethylsilylspiroamino-oxathioledioxide (TSAO) (Sheng & Zhang, 2011), and the cephalosporin antibiotic Cefatrizine (RBusto, et al., 1976).

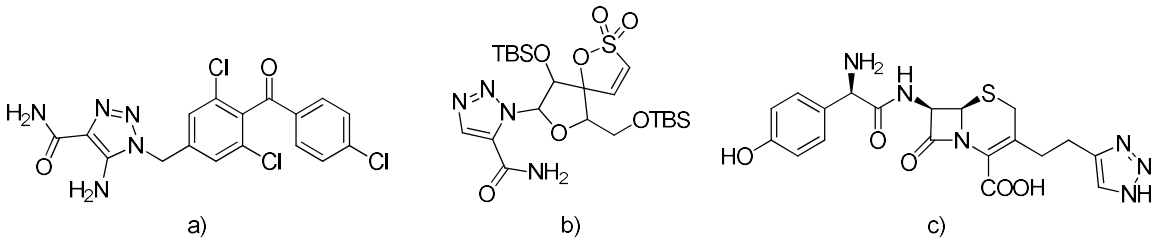


Figure 14. Biologically active compounds all containing the 1,2,3-triazole moiety: a) CAI; b) TSAO; c) Cefatrizine.

Another underlining attraction for incorporating these moieties are their use as linkers, and showing bioisosteric effects on peptide linkages, aromatic rings, alkenes, and imidazole rings. Chemical characteristics including dipole-dipole, π-stacking interactions, hydrogen bond formation, low toxicity, and improved solubility of triazole compounds have increased their awareness in the medicinal field because of how they are binding with their biological targets with such high affinity (Dheer, Singh, & Shankar, 2017). An array of alternate applications are illustrated in scheme 23 with future target molecules. Alternative other targets to be investigated would include the reactions of the C7 and C9 hydroxy-substituted *trans*-dihydroxynarciclasine derivatives with propagyl bromide and other reagents as such containing alkyne functional groups. Upon gathering our results from the *trans*-dihydrolycoricidine analogue studies via ATRI, further studies can be addressed including structures which resemble more like their parent compound to observe the differences between the compound's mode of action.

2.32D.1 References

F. Amblard, J. H. Cho, R. F. Schinazi. *Chemical Reviews*. **2009**. *109*, 4207.

R. D. Busto, E. Haas, T. Madhavan, K. Burch, F. Cox, E. Fisher, E. Quinn, D. Pohlod. *Antimicrobial Agents and Chemotherapy*. **1976**, 376.

D. K. Dalvie, A. S. Kalgutkar, S. C. Khojasteh-Bakht, R. S. Obach, J. P. O'Donnell. *Chemical Research in Toxicology*. **2002**, *15*, 269.

M. Dhameja, J. Pandey. *Asian Journal of Organic Chemistry*. **2018**, *7*, 1502.

D. Dheer, V. Singh, R. Shankar. *Bioorganic Chemistry*. **2017**, *71*, 30.

K. V. Gothelf, K. A. Jorgenson, Chemical Reviews. **1998**, *98*, 863.

W. S. Horne, M. K. Yadav, C. D. Stout, M. R. Ghadiri. *Journal of the American Chemical Society*. **2004**, *126*, 15366.

R. Huisgen. *Angewandte Chemie*. **1963**, *75*, 604.

R. Huisgen. *Angewandte Chemie International Edition*. **1963**, *2*, 565.

R. Huisgen. *Angewandte Chemie*. **1963**, *75*, 742.

R. Huisgen. *Angewandte Chemie International Edition*. **1963**, *2*, 633.

S. Hwang, D. Kim, S. Kim. *Chemistry – A European Journal*. **2012**, *18*, 9977.

J. In, S. Hwang, C. Kim, J. H. Seo, S. Kim. *European Journal of Organic Chemistry*. **2013**, 965.

R. Kharb, P. C. Sharma, M. S. Yar. *Journal of Enzyme Inhibition and Medicinal Chemistry*. **2011**, *26*, 1.

H. C. Kolb, K. B. Sharpless. *Drug Discovery Today*. **2003**, *8*, 1128.

A. Martínez, D. Vega, J. Aguirre, B. Lantaño, G. Moltrasio. *Synthesis*. **2012**, 44, 125.

J. McNulty, K. Keskar. *European Journal of Organic Chemistry*. **2012**, 5462.

J. McNulty, C. Zepeda-Velazquez. *Angewandte Chemie International Edition*. **2014**, *53*, 8450.

M. Movassaghi, M. D. Hill. *Journal of the American Chemical Society*. **2006**, *128*, 14254.

T. Hudlický, U. Rinner, D. Gonzalez, H. Akgun, S. Schilling, P. Siengalewicz, T. A.

G. R. Martinot, G. R. Pettit. *The Journal of Organic Chemistry*. **2002**, *67*, 8726.

S. Ohta, S. Kimoto. *Tetrahedron Letters*. **1975**, 2279.

V. V. Rostovtsev, L. G. Green, V. V. Fokin, K. B. Sharpless. *Angewandte Chemie*. **2002**, *114*, 2708.

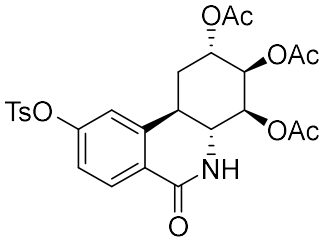
C. Sheng, W. Zhang, *Current Medicinal Chemistry*. **2011**, *18*, 733.

M. J. Soltis, H. J. Yeh, K. A. Cole, N. Whittaker, R. P. Wersto, E. C. Kohn. *Drug Metabolism & Disposition*. **1996**, *24*, 799.

K. White, M. Mewald, M. Movassaghi. *The Journal of Organic Chemistry*. **2014**, *16*, 358.

2.33 Experimental

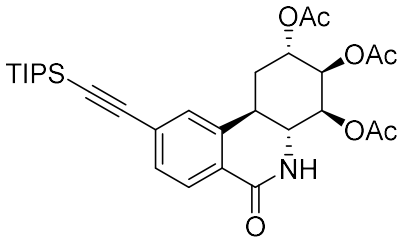
(2*S*,3*R*,4*S*,4*aR*,10*bR*)-6-oxo-9-(tosyloxy)-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [128]



Pyridine (0.024 mL, 0.306 mmol, 6 eq.) was added to a mixture of 4-toluenesulfonyl chloride (0.011 g, 0.056 mmol, 1.1 eq.) in CH_2Cl_2 (0.5 mL). Phenolic compound **126** (0.02 g, 0.051 mmol, 1 eq.) was dissolved in CH_2Cl_2, and added to reaction mixture and stirred at rt for 16 h. After that the pyridine was removed *in vacuo* (0.1 mbar). The residue was dissolved in EtOAc (2 mL) and washed with saturated $NaHCO_3$ (2 x 10 mL) and water (10 mL). The solvent was removed *in vacuo* and the product purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford tosylate compound **128** (0.048 g, 95% **yield**) of clear yellow oil. $\mathbf{[\alpha]^{23}_D}$ = +76 (c = 0.58 $CHCl_3$, l = 1 dm). **^{1}H NMR:** (600 MHz, $CDCl_3$) **δ** 8.0 (d, J = 8.68 Hz, 1H), 7.75 (d, J = 7.98 Hz, 2H), 7.36 (d, J = 7.98 Hz, 2H), 7.06 (s, 1H), 6.86 (d, J = 8.4 Hz, 1H), 8.21 (s, 1H), 5.45 (t, J = 2.99 Hz, 1H), 5.23-5.19 (m, 2H), 3.83 (t, J = 11.75 Hz, 1H), 3.25 (td, J = 3.39, 12.9 Hz, 1H), 2.48 (s, 3H), 2.42 (d, J = 14.3 Hz, 1H), 2.15 (s, 3H), 2.10 (s, 3H), 2.09 (s, 3H), 1.92 (m, 1H). **^{13}C**

NMR (151 MHz, $CDCl_3$) δ 170.23, 169.27, 169.15, 164.82, 152.75, 145.93, 141.97, 132.25, 130.33, 129.98, 128.51, 127.51, 121.14, 118.31, 71.60, 68.36, 67.32, 52.58, 35.05, 26.34, 21.76, 21.05, 20.82, 20.73. **HRMS** (ESI): exact mass calculated for $C_{26}H_{27}NO_{10}SNa$ [$(M + Na)^+$], 568.1253; found 568.1246.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-6-oxo-9-((triisopropylsilyl)ethynyl)-1,2,3,4,4*a*,5,6,10*b*-octahydrophenanthridine-2,3,4-triyl triacetate [129]

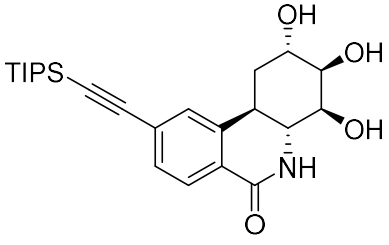


Phenolic compound **126** (0.010 g, 0.025 mmol, 1 eq.) was dissolved in CH_2Cl_2. Pyridine (0.012 mL, 0.153 mmol, 6 eq.) and a 1M solution of triflic anhydride (0.025 mL, 0.025 mmol, 1 eq.) in CH_2Cl_2 was added dropwise to the reaction mixture over 10 min at 0 °C and stirred at rt for 16 h. After that the pyridine was removed *in vacuo* (0.1 mbar). The residue was dissolved in EtOAc (2 mL) and water (3 mL) was added to extract with EtOAc (3 x 5 mL). Organic phases were concentrated *in vacuo* to afford triflate compound (0.013 g, 0.025 mmol) which was used for next step without further purification.

Copper iodide (0.001 g, 0.005 mmol, 0.2 eq.) was charged in a 5 mL two neck RBF, heated with a heat gun under vacuum, and flushed with argon. $PdCl_2(PPh_3)_2$ (0.0018 g, 0.0025 mmol, 0.1 eq.), tricyclohexylphosphine (0.0003 g, 0.0013 mmol, 0.05 eq.), and a stir bar were added to the RBF that was then sealed with a septum in the glove box. A mixture of triflate (0.013 g, 0.025 mmol, 1 eq.) in THF:DIPA (0.7 mL/0.3 mL) was stirred for 10 min. TIPS acetylene (0.017 mL, 0.076 mmol, 3 eq.) was added dropwise to the reaction mixture over a period of 10 min, and let stir for 15 min and the reaction mixture turned to a red

color. Reaction mixture was stirred for 3 h at 35 ºC. After confirming by TLC, reaction mixture was quenched with water, and extracted with EtOAc (3 x 10 mL), organic phases were concentrated *in vacuo*, and the product purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford Sonogashiara product **129** (0.013 g, 91% **yield**) as a clear yellow oil. **$[\alpha]^{23}_D$** = +34 (c = 1, THF, l = 1 dm). **^{1}H NMR** (700 MHz, $CDCl_3$) δ 8.04 (d, *J* = 7.9 Hz, 1H, Ar-*H*), 7.51 (d, *J* = 7.9 Hz, 1H, Ar-*H*), 7.34 (s, 1H, Ar-*H*), 6.26 (s, 1H, CON*H*), 5.46-5.45 (m, 1H, C*H*-OAc), 5.26-5.21 (m, 2H, C*H*-OAc, C*H*-OAc), 3.85-3.80 (dd, *J* =12.3, 10.9 Hz, 1H, C*H*-NH), 3.25 (td, *J* = 12.6, 3.6 Hz, 1H), 2.59 (dd, *J* = 14.4, 3.6 Hz, 1H, Ph-CH-C*H*$_2$), 2.15 (s, 3H, $COCH_3$), 2.10 (d, *J* = 7.6 Hz, 3H, $COCH_3$), 2.07 (s, 3H, $COCH_3$), 2.04-1.99 (m, 1H, Ph-CH-C*H*$_2$), 1.15-1.11 (m, 21H). **^{13}C NMR** (176 MHz, $CDCl_3$) δ 170.21, 169.43, 169.18, 165.26, 139.62, 131.15, 128.42, 128.13, 126.92, 126.92, 105.93, 94.62, 71.76, 68.56, 67.45, 52.58, 34.83, 26.37, 21.07, 20.81, 20.71, 18.67, 11.26. **HRMS** (ESI): exact mass calculated for $C_{30}H_{41}NO_7SiNa$ [$(M + Na)^+$], 578.2550; found 578.2538.

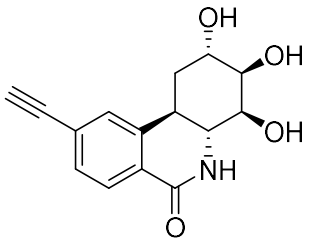
(2*S*,3*R*,4*S*,4*aR*,10*bR*)-2,3,4-trihydroxy-9-((triisopropylsilyl)ethynyl)-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one



129 (0.0066 g, 0.012 mmol, 1 eq.) and K_2CO_3 (0.002 g, 0.010 mmol, 0.8 eq.) in MeOH (1 mL) were stirred with the TLC analysis showing full conversion after 6 h. Extracted with EtOAc (3 x 10 mL) after adding water (2 mL), and the organic phases were concentrated *in vacuo*, and the product was purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 98:2) to afford TIPS triol (0.0048 g, 95% **yield**). **$[\alpha]^{23}_D$** = +48 (c = 0.33, MeOH, l = 1 dm). **^{1}H NMR** (600 MHz, CD_3OD) δ

7.99 (d, *J* = 8.0 Hz, 1H), 7.49 (d, *J* = 8.0 Hz, 1H), 7.44 (s, 1H), 4.16 (dd, *J* = 5.9, 2.9 Hz, 1H), 3.98-3.97 (m, 1H), 3.94 (dd, *J* = 10.1, 3.0 Hz, 1H), 3.57 (dd, *J* = 12.5, 10.1 Hz, 1H), 3.18 (td, *J* = 12.5, 3.6 Hz, 1H), 2.36 (dt, *J* = 13.6, 3.6 Hz, 1H), 1.93 (td, *J* = 7.6, 3.8 Hz, 1H), 1.20 (s, 21H). **^{13}C NMR** (151 MHz, CD_3OD) δ 167.36, 143.55, 131.23, 129.82, 128.89, 128.79, 127.84, 107.78, 94.19, 73.27, 71.49, 70.44, 55.99, 35.31, 29.13, 18.97, 12.40. **HRMS** (ESI): exact mass calculated for $C_{24}H_{35}NO_4SiNa$ [(M + Na)$^+$], 452.2233; found 452.2226.

(2*S*,3*R*,4*S*,4*aR*,10*bR*)-9-ethynyl-2,3,4-trihydroxy-1,3,4,4*a*,5,10*b*-hexahydrophenanthridin-6(2H)-one [104]



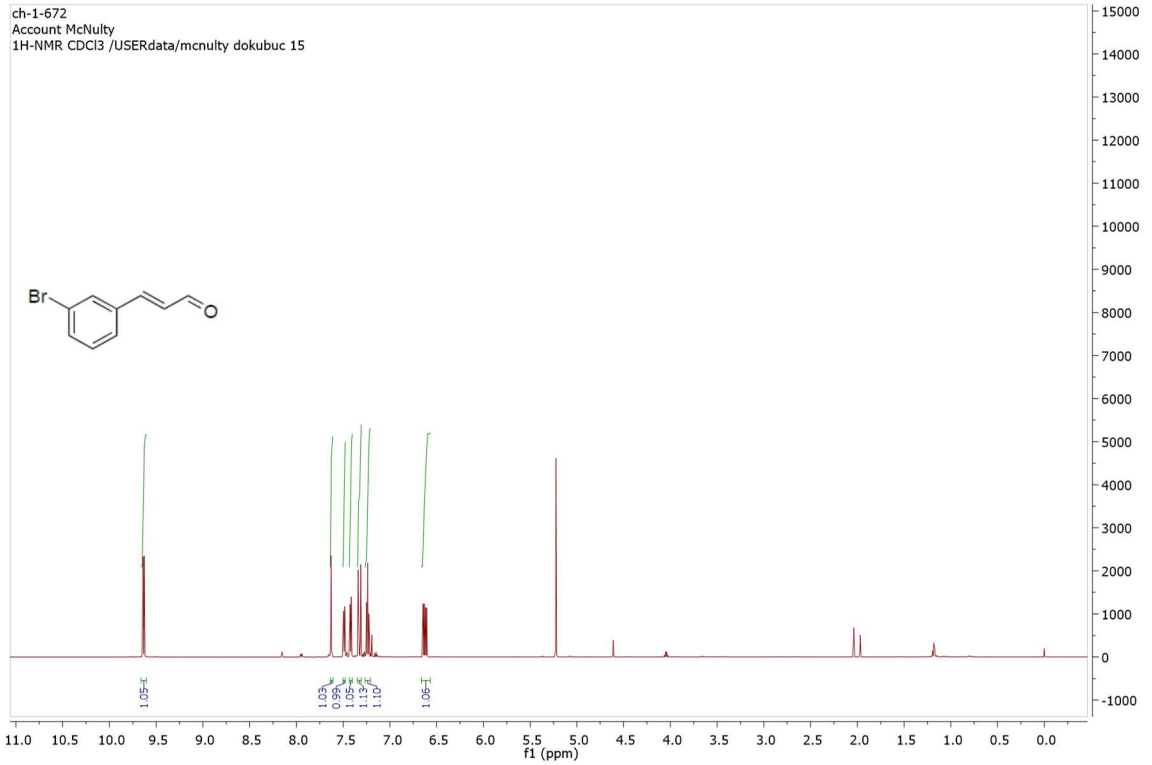
TIPS triol (0.002 g, 0.0046 mmol, 1 eq.) was dissolved in THF (0.5 mL), and a 1 M solution of TBAF (0.01mL, 2.2 eq.) in THF was added at 0 ºC, and stirred for 0.5 h. Reaction was quenched with 1 M HCl (1mL) and allowed to stir 0.5 h and extracted with EtOAc (3 x 10 mL), organic phases were concentrated *in vacuo*. The product was purified by flash column chromatography (CH_2Cl_2/MeOH; 100:0 to 90:10) to afford alkyne triol **104** (0.0012 g, 95% **yield**). **$[\alpha]^{23}_D$** = +25 (c = 0.04, MeOH, l = 1 dm). **^{1}H NMR** (700 MHz, CD_3OD) δ 7.97 (t, *J* = 10.0 Hz, 1H), 7.51 (t, *J* = 10.0 Hz, 1H), 7.50 (s, 1H), 4.14 (dd, *J* = 5.8, 2.8 Hz, 1H), 3.96 (t, *J* = 3.2 Hz, 1H), 3.93 (dd, *J* = 10.1, 3.0 Hz, 1H), 3.73 (s, 1H), 3.55 (dd, *J* = 12.5, 10.1 Hz, 1H), 3.17 (td, *J* = 12.5, 3.5 Hz, 1H), 2.35 (dt, *J* = 13.5, 3.2 Hz, 1H), 1.95-1.89 (m, 1H). **^{13}C NMR** (176 MHz, MeOD) δ 167.45, 143.65, 131.41, 130.09, 128.90, 128.36, 128.21, 79.47, 79.28, 73.38, 71.58, 70.54, 56.12, 35.43, 29.21. **HRMS** (ESI): exact mass calculated for $C_{15}H_{15}NO_4Na$ [(M + Na)+], 296.0899; found 296.0886.

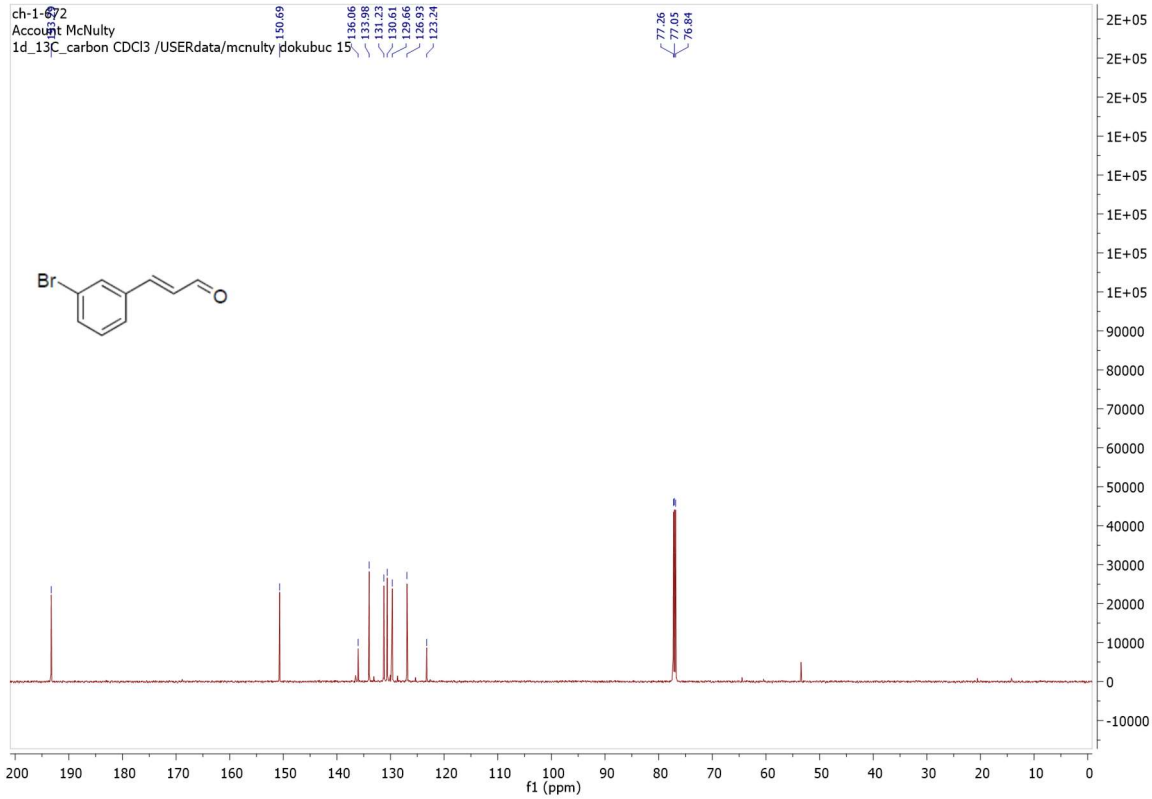
2.33.1 References

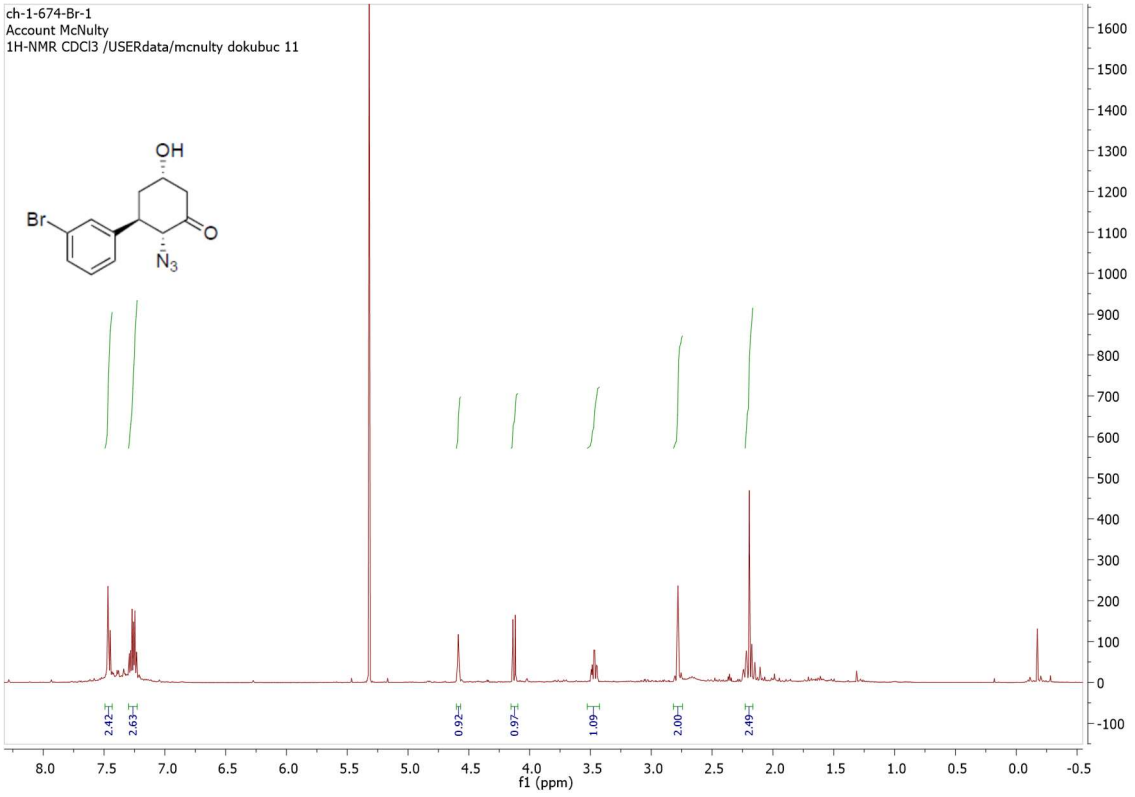
I. Kanwal, A. Mujahid, N. Rasool, K. Rizwan, A. Malik. *Catalysts*. **2020**, 443.

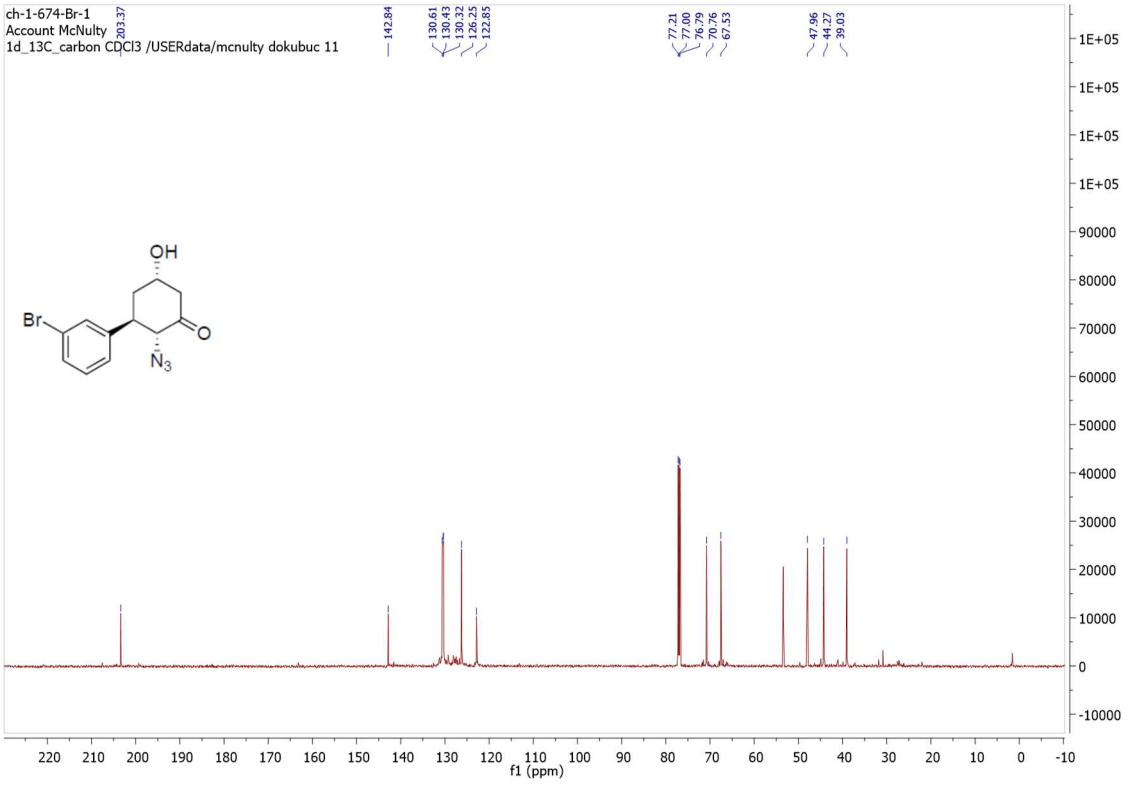
3.0 Appendices

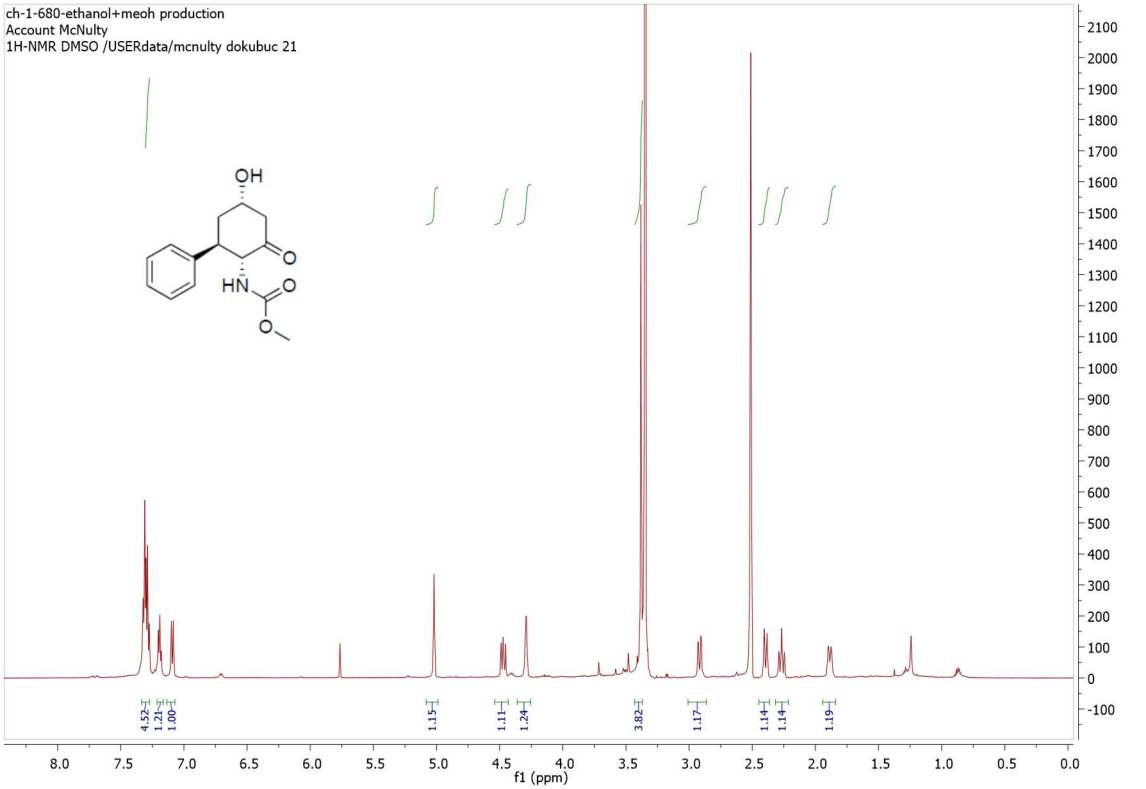
3.1 Appendix A: NMR Spectra Pertaining to Chapter 2.1

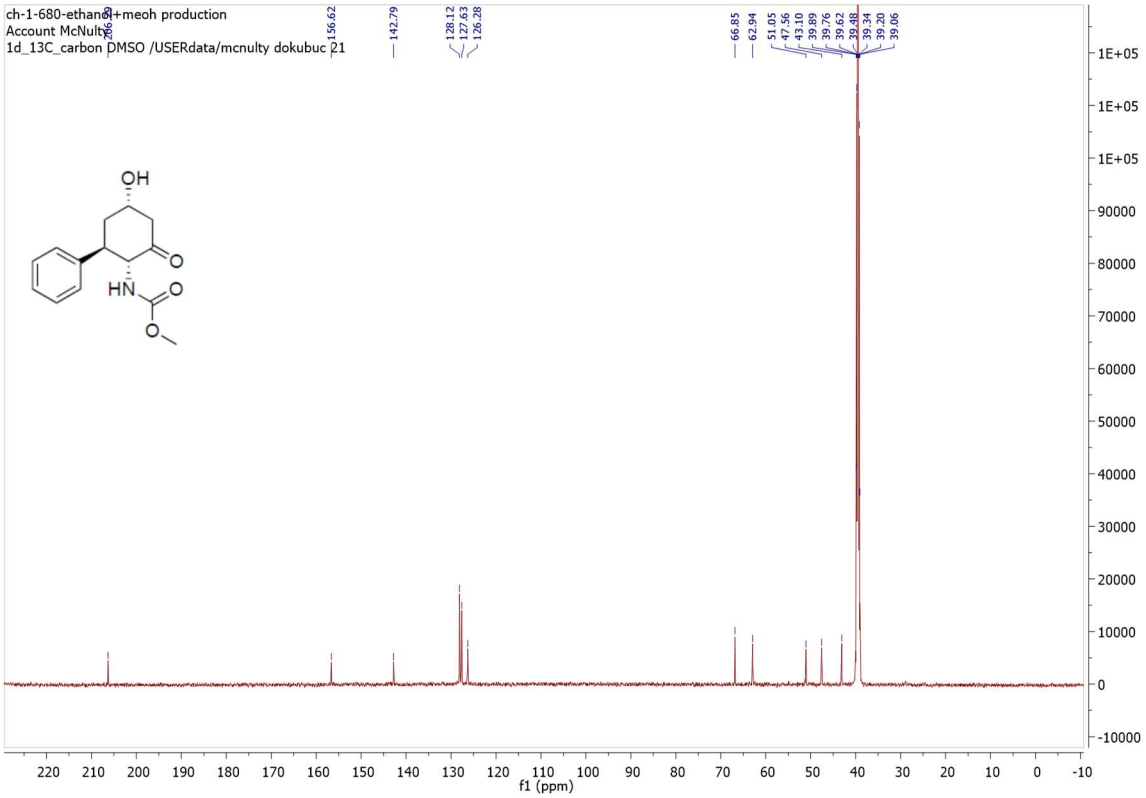


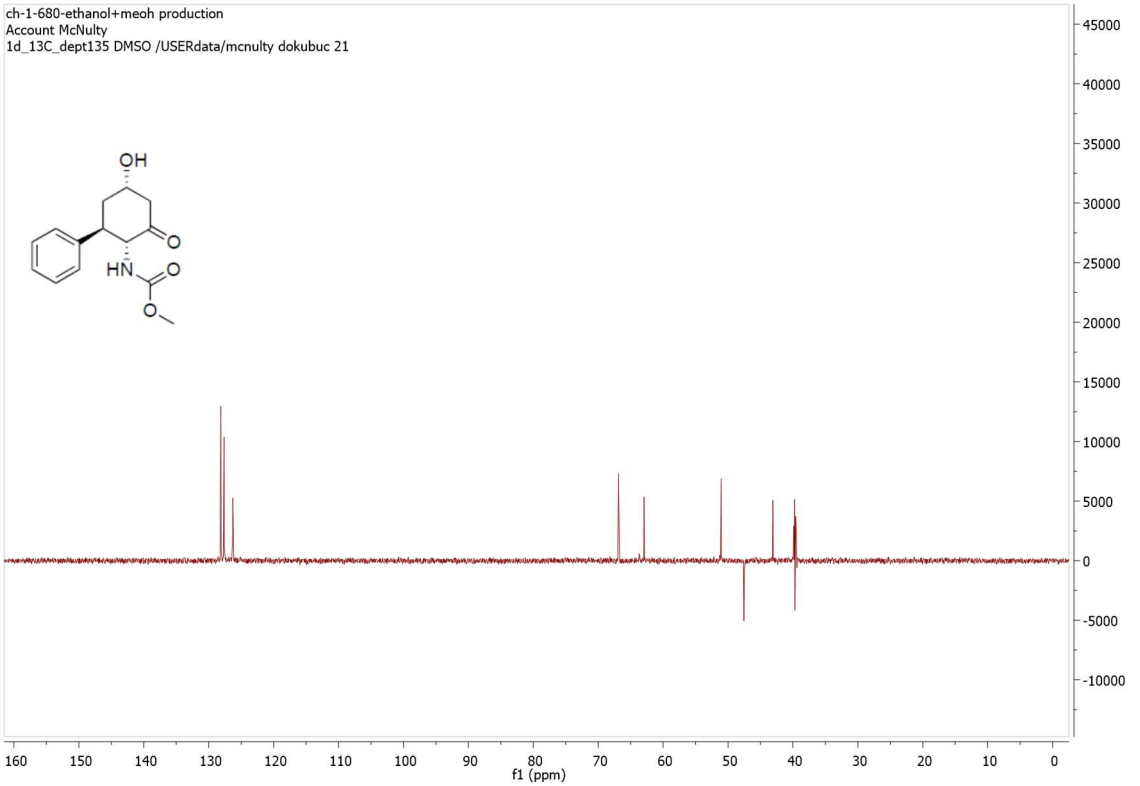


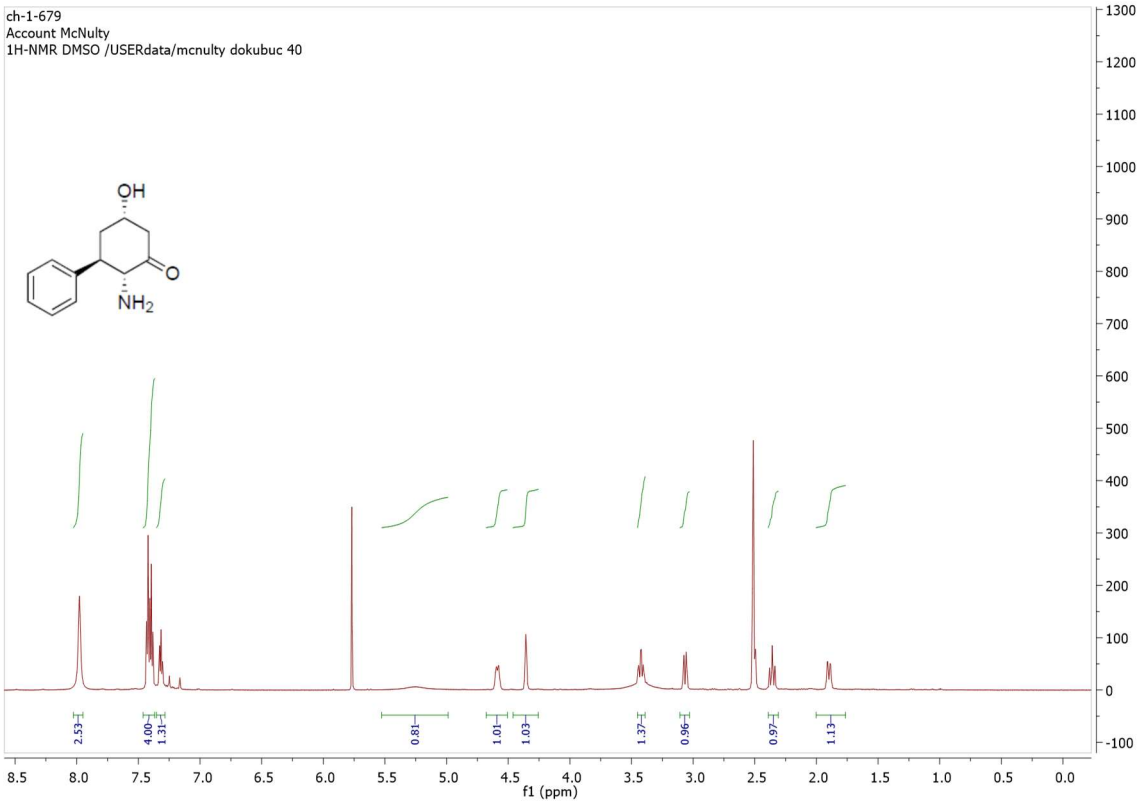

ch-1-674-Br-1
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 11
OH
Br
O
N3
2.42
2.63
0.92
0.97
1.09
2.00
2.49
f1 (ppm)

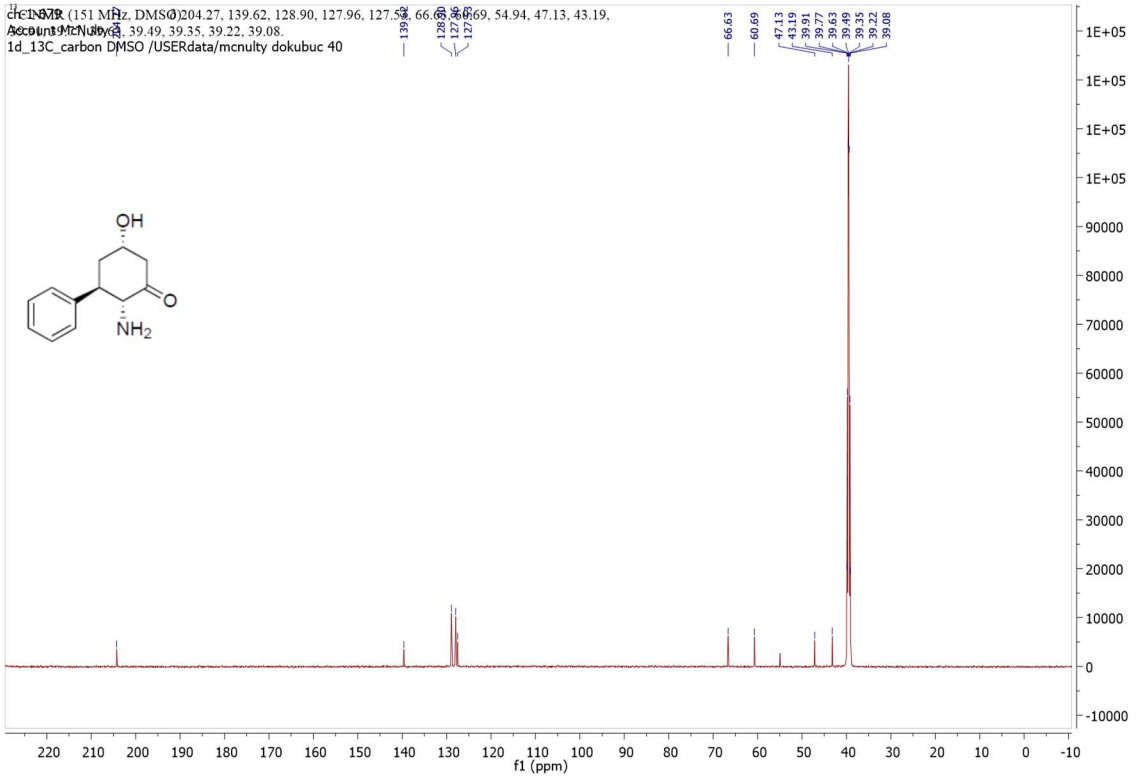

ch-1-674-Br-1
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty dokubuc 11
203.37
142.84
130.61
130.43
130.32
126.25
122.85
77.21
77.00
76.79
70.76
67.53
47.96
44.27
39.03
OH
Br
O
N3
f1 (ppm)

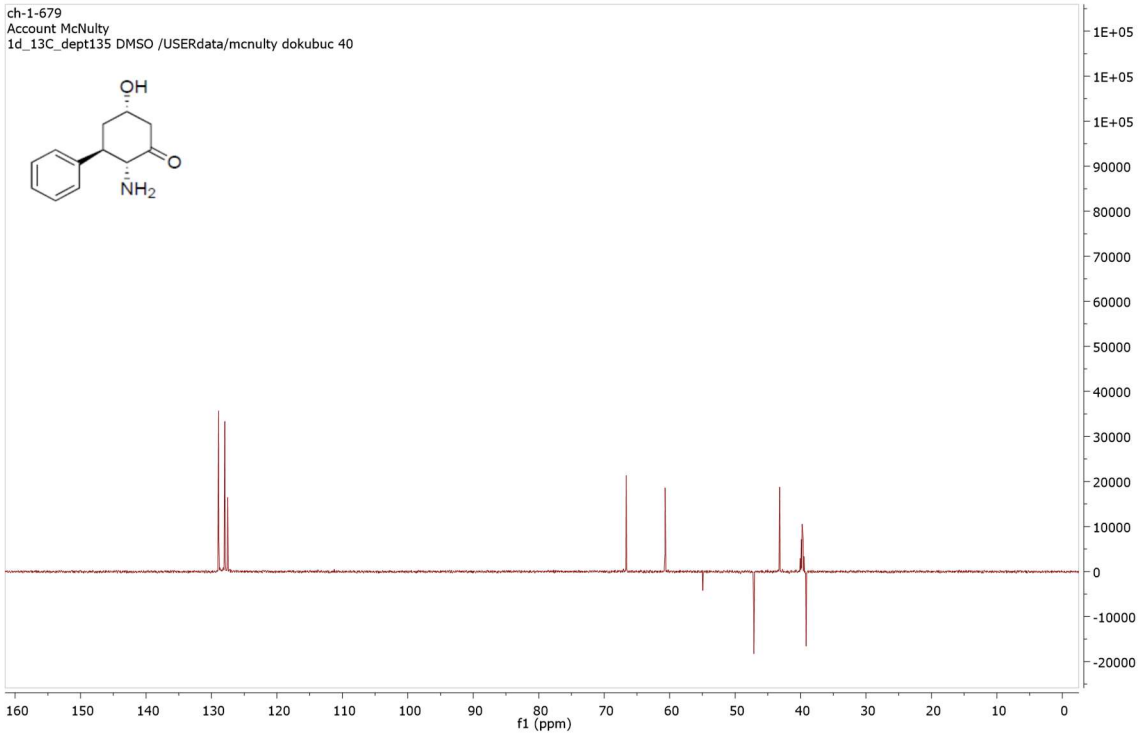

ch-1-680-ethanol+meoh production
Account McNulty
1H-NMR DMSO /USERdata/mcnulty dokubuc 21
OH
O
HN
O
O
4.52
1.21
1.00
1.15
1.11
1.24
3.82
1.17
1.14
1.14
1.19
f1 (ppm)

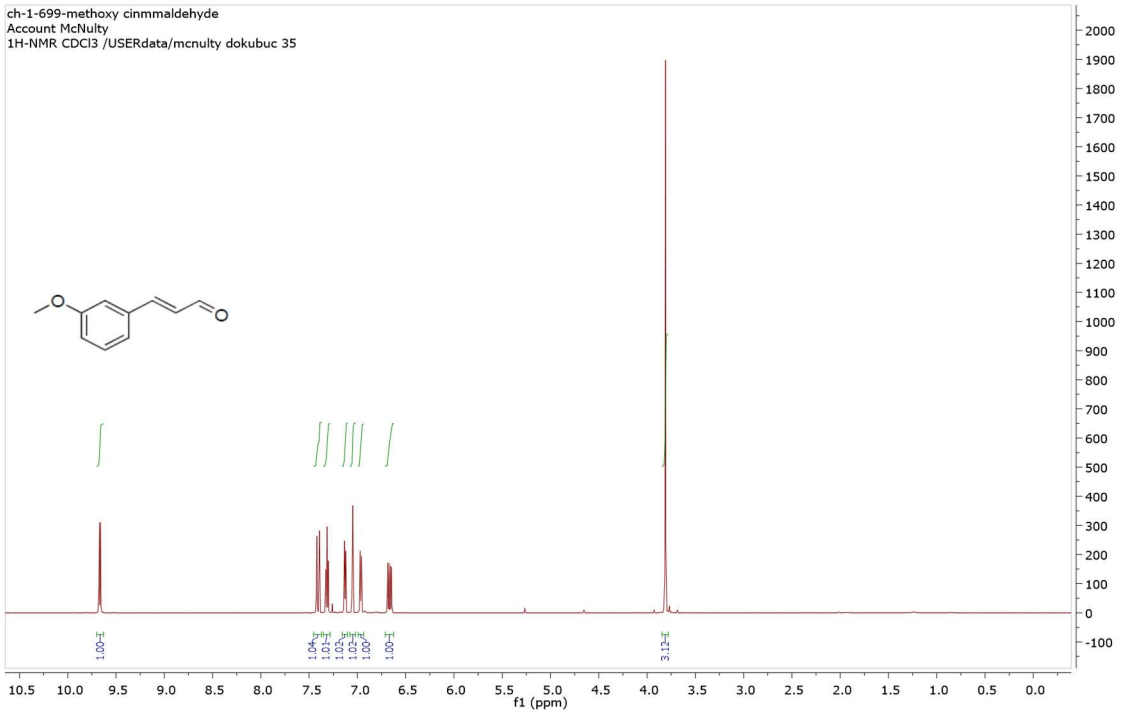

ch-1-680-ethanol+meoh production
Account McNulty
1d_13C_carbon DMSO /USERdata/mcnulty dokubuc 21
156.62
142.79
128.12
127.63
126.28
66.85
62.94
51.05
47.56
43.10
39.89
39.76
39.62
39.48
39.34
39.20
39.06
OH
O
HN
O
O
f1 (ppm)

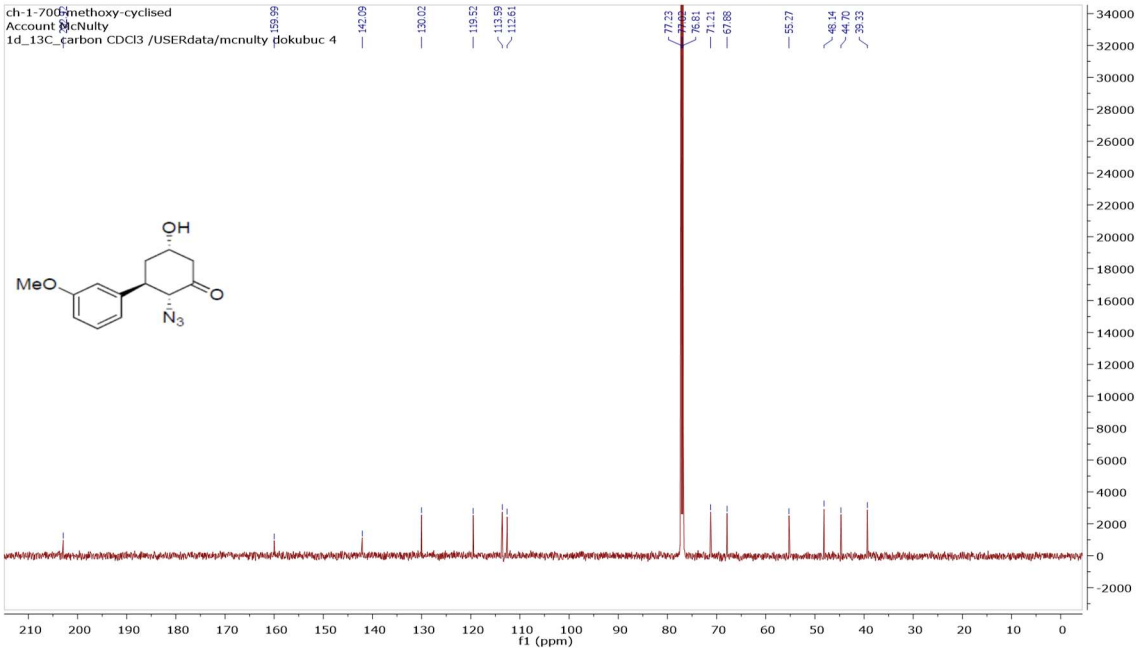

ch-1-680-ethanol+meoh production
Account McNulty
1d_13C_dept135 DMSO /USERdata/mcnulty dokubuc 21
OH
O
HN
O
O
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
-5000
-10000
160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

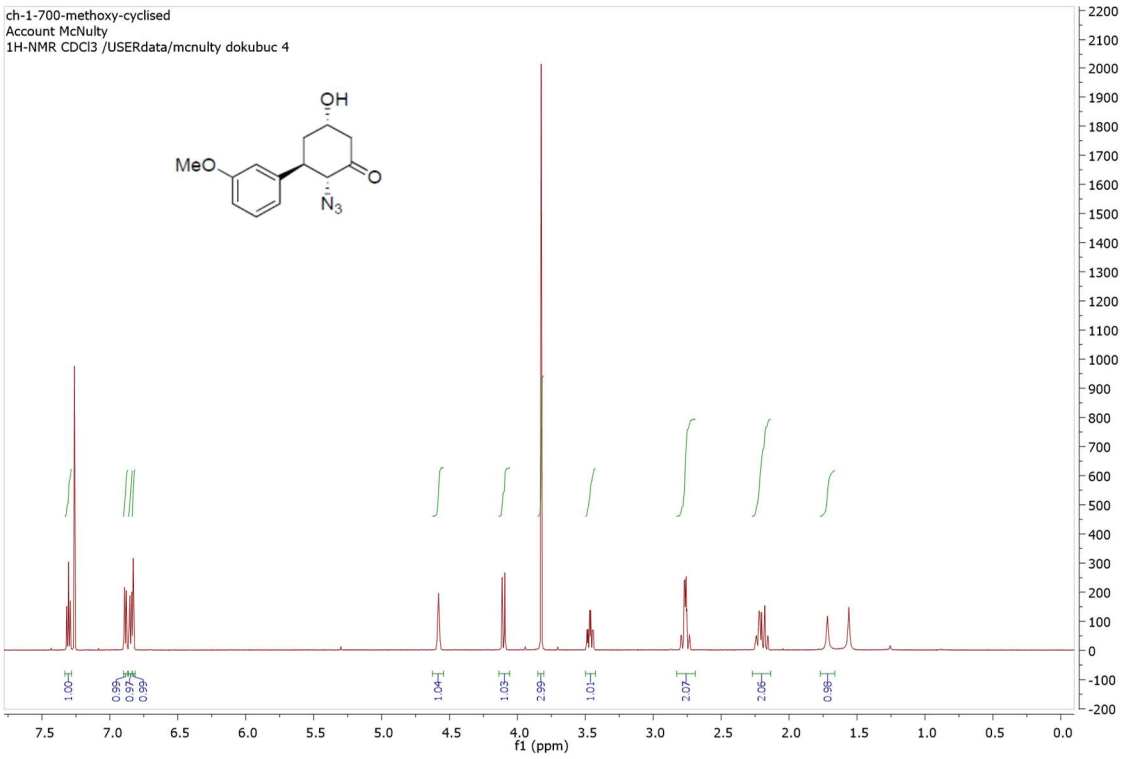

ch-1-679
Account McNulty
1H-NMR DMSO /USERdata/mcnulty dokubuc 40
OH
O
NH2
1300
1200
1100
1000
900
800
700
600
500
400
300
200
100
0
-100
2.53
4.00
1.31
0.81
1.01
1.03
1.37
0.96
0.97
1.13
8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)

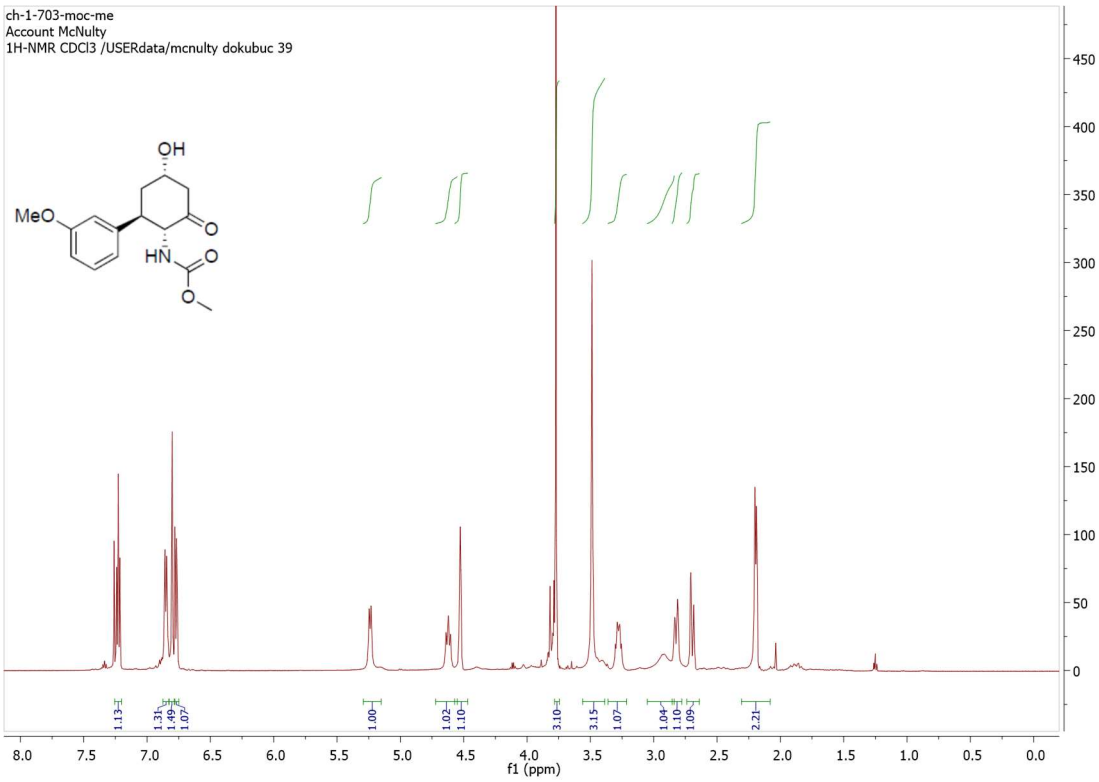

ch-1-679
Account McNulty
1d_13C_carbon DMSO /USERdata/mcnulty dokubuc 40
OH
O
NH2
f1 (ppm)

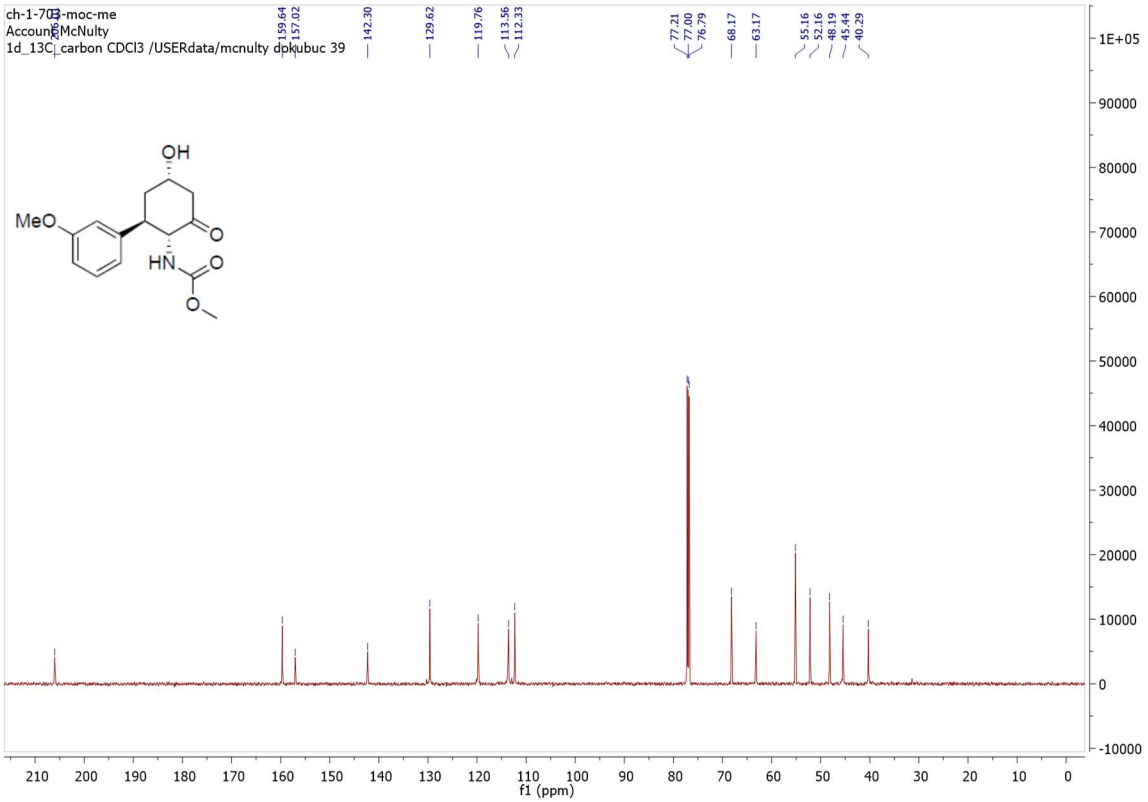

ch-1-679
Account McNulty
1d_13C_dept135 DMSO /USERdata/mcnulty dokubuc 40
OH
O
NH2
f1 (ppm)

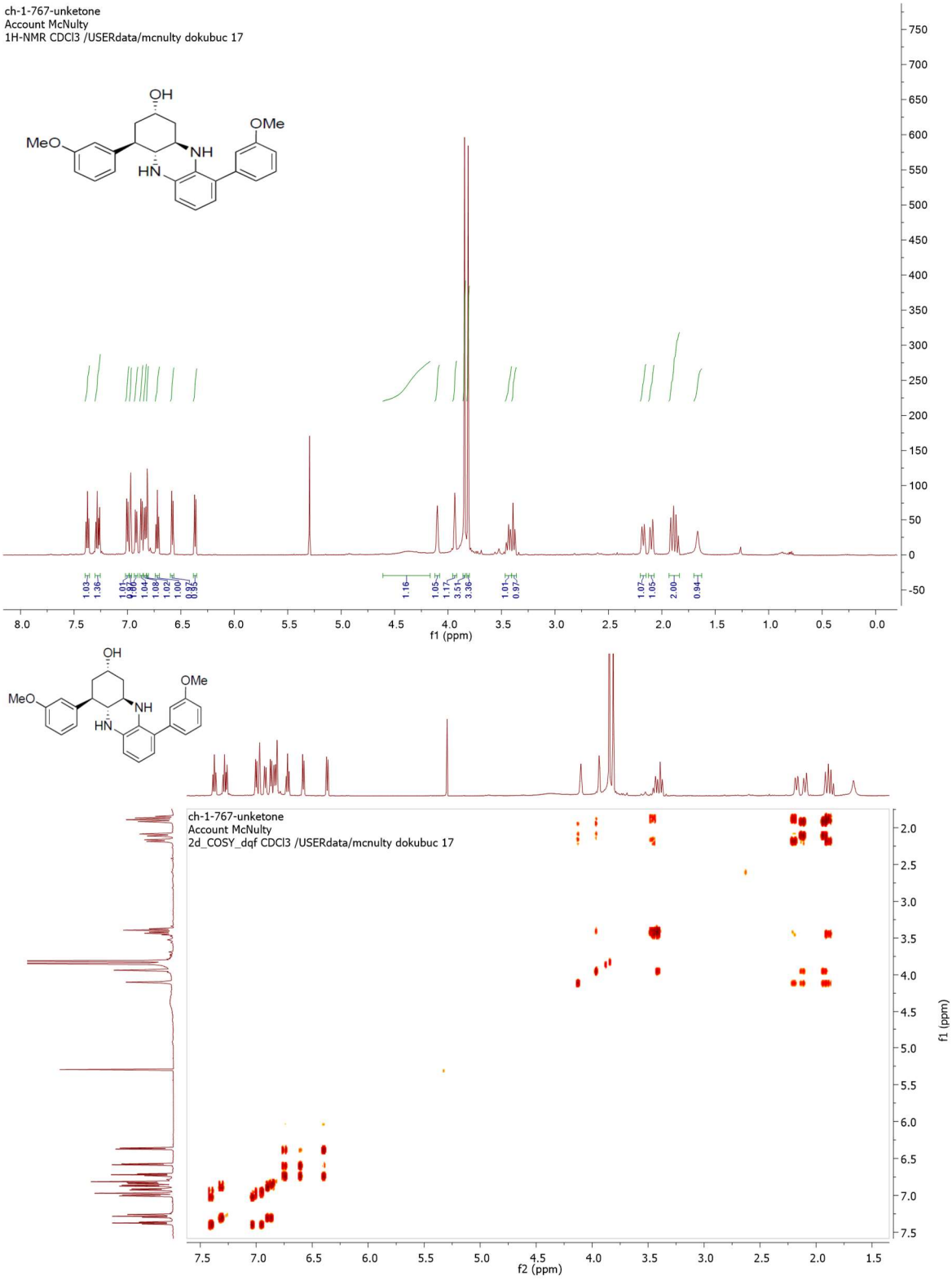

ch-1-699-methoxy cinmmaldehyde
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 35
1.00
1.04
1.01
1.05
1.02
1.00
1.00
3.12
f1 (ppm)

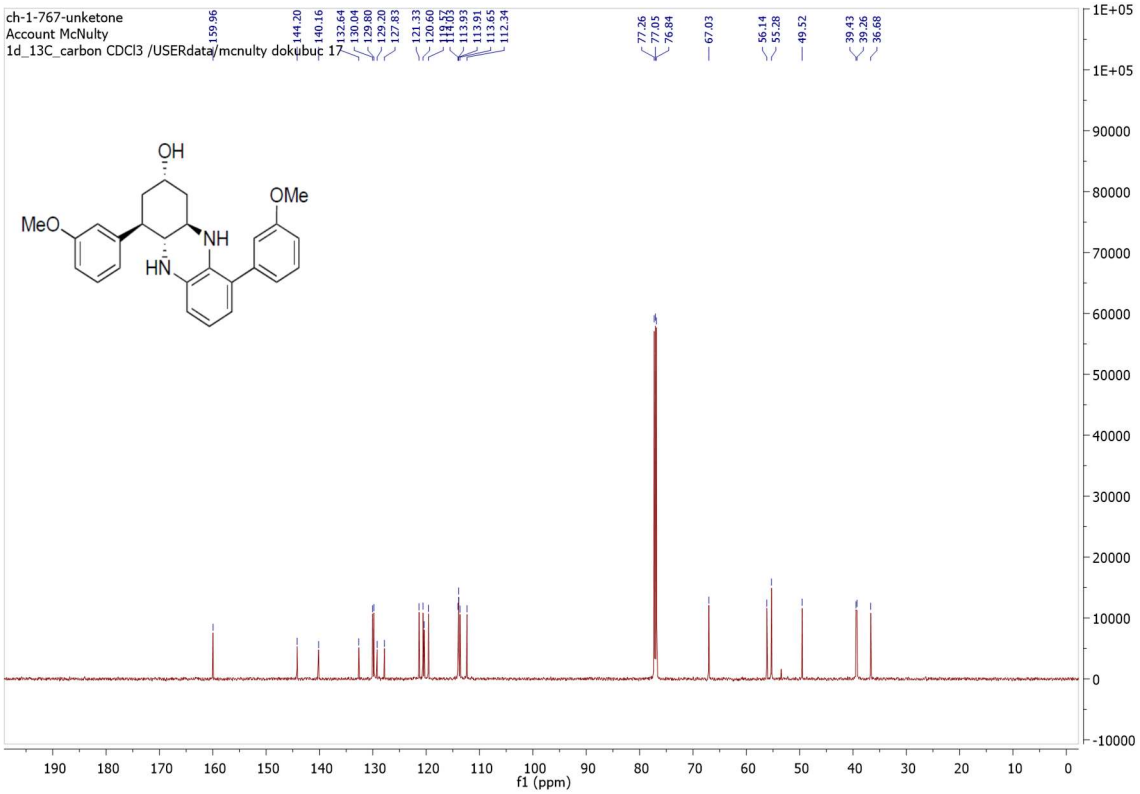

ch-1-700-methoxy-cyclised
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty dokubuc 4
159.99
142.09
130.02
119.52
113.59
112.61
77.23
76.81
71.21
67.88
55.27
48.14
44.70
39.33
OH
MeO
O
N3
f1 (ppm)

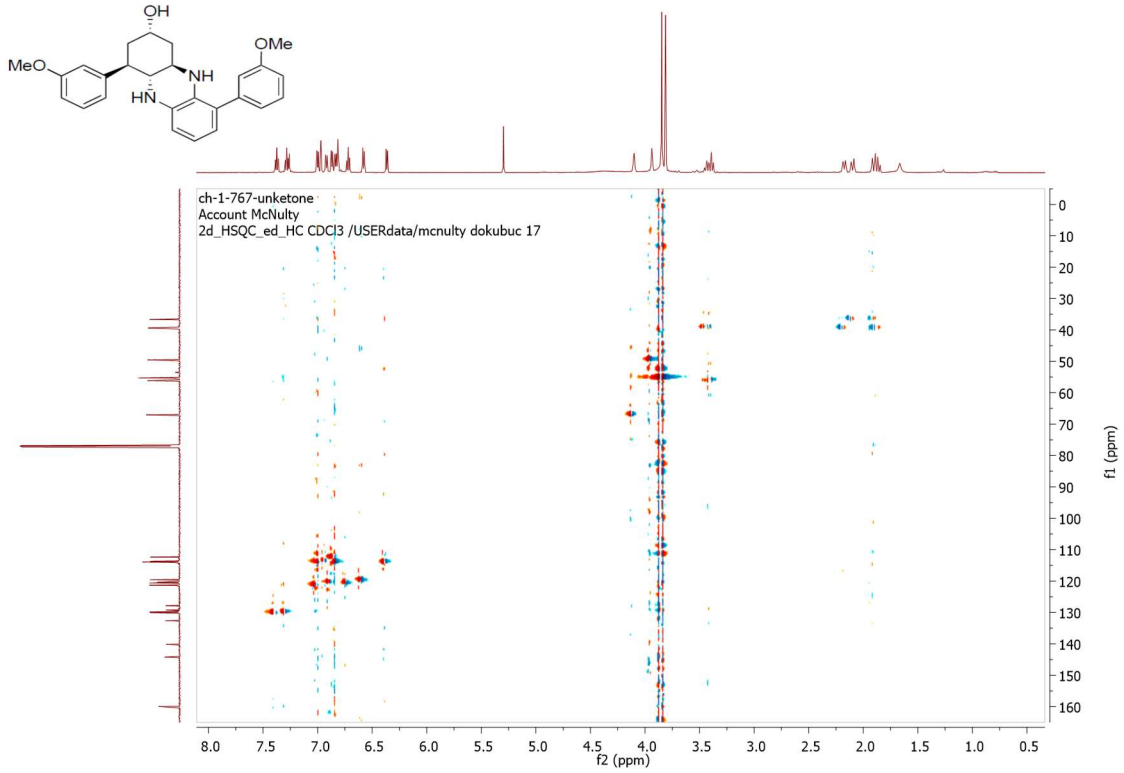

ch-1-700-methoxy-cyclised
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 4
OH
MeO
O
N3
1.00
0.99
0.97
0.99
1.04
1.03
2.99
1.01
2.07
2.06
0.98
f1 (ppm)

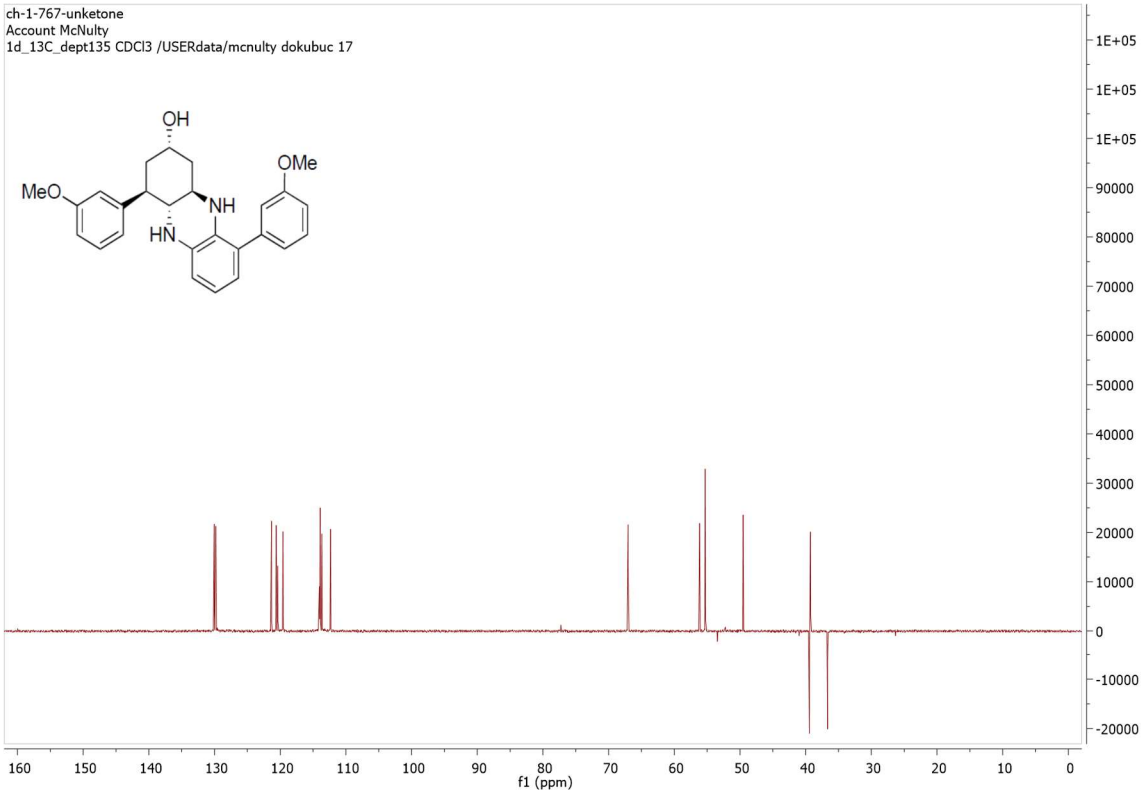

ch-1-703-moc-me
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 39
OH
MeO
O
HN
O
O
1.13
1.31
1.49
1.07
1.00
1.02
1.10
3.10
3.15
1.07
1.04
1.10
1.09
2.21
f1 (ppm)

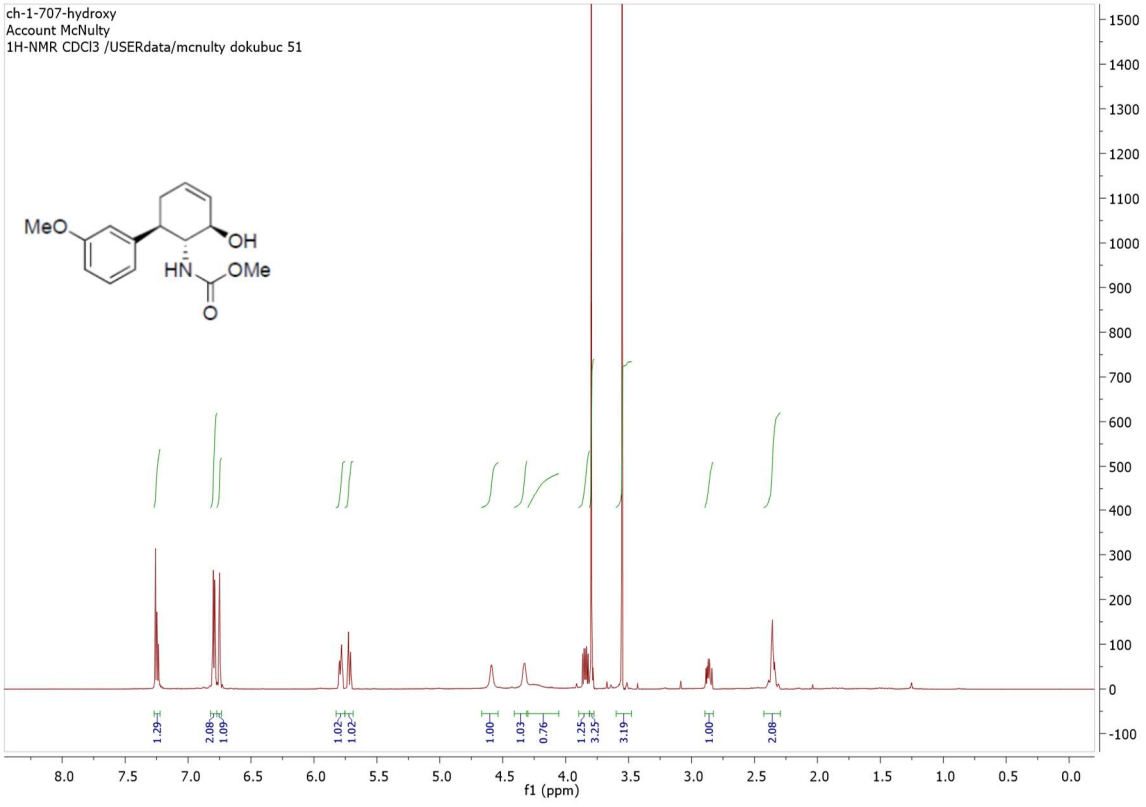

ch-1-703-moc-me
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty dokubuc 39
159.64
157.02
142.30
129.62
119.76
113.56
112.33
77.21
77.00
76.79
68.17
63.17
55.16
52.16
48.19
45.44
40.29
OH
MeO
O
HN
O
O
f1 (ppm)

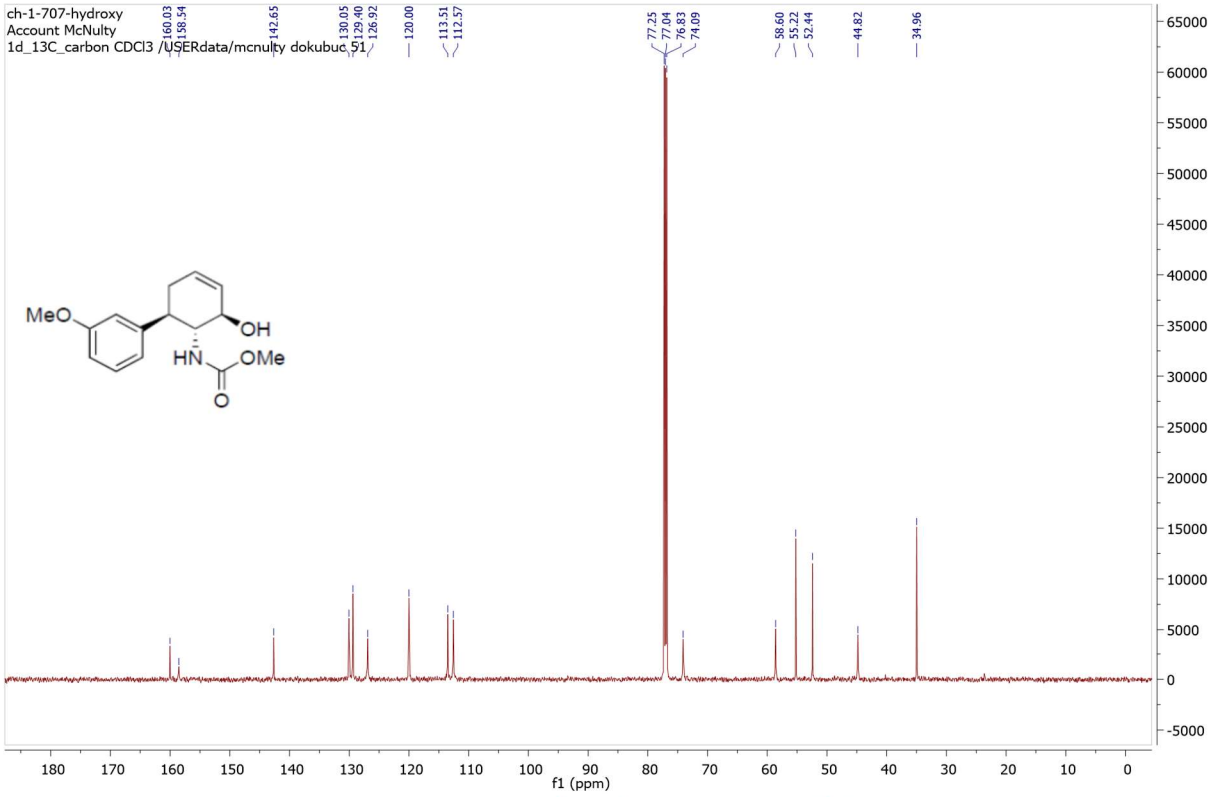

ch-1-767-unketone
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 17
OH
OMe
MeO
NH
HN
f1 (ppm)
ch-1-767-unketone
Account McNulty
2d_COSY_dqf CDCl3 /USERdata/mcnulty dokubuc 17
f2 (ppm)
f1 (ppm)

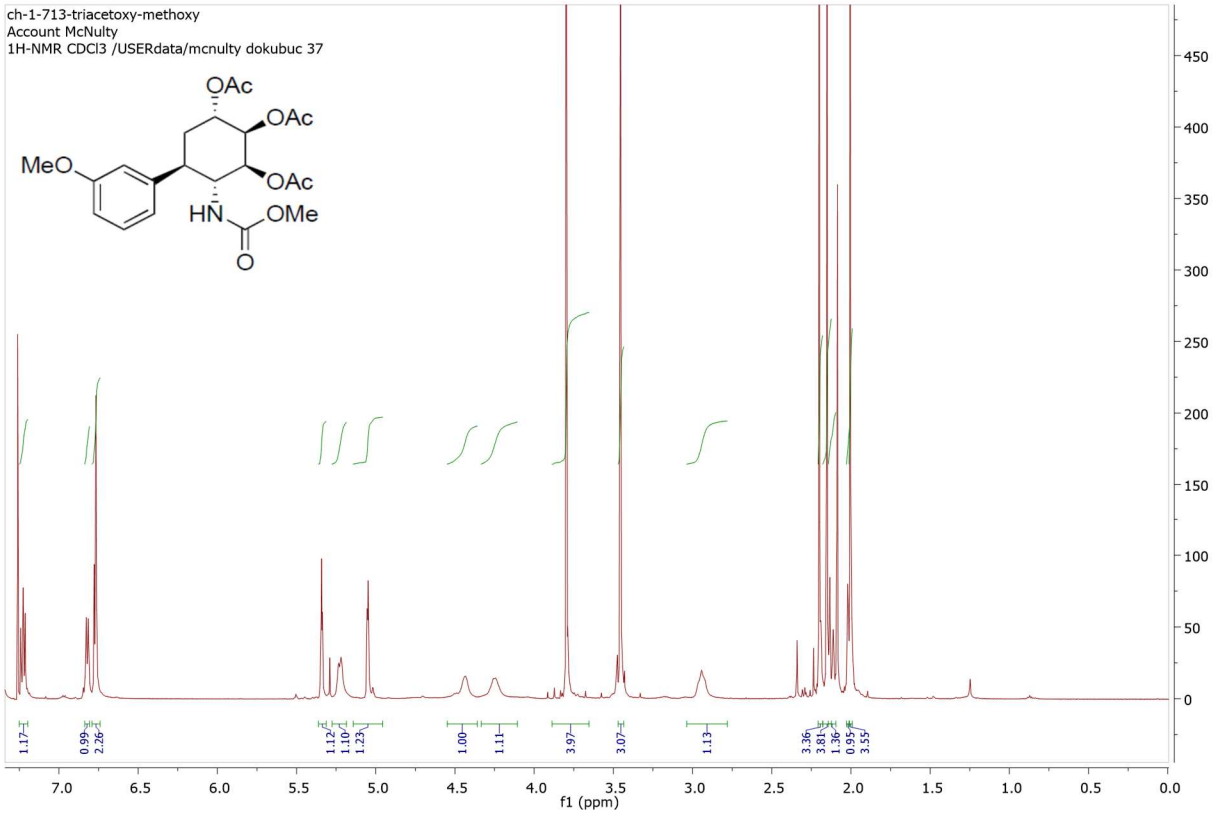

ch-1-767-unketone
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty dokubuc 17
159.96
144.20
140.16
132.64
130.04
129.80
129.20
127.83
121.33
120.60
118.57
118.03
113.93
113.91
113.55
112.34
77.26
77.05
76.84
67.03
56.14
55.28
49.52
39.43
39.26
36.68
OH
MeO
OMe
NH
HN
f1 (ppm)

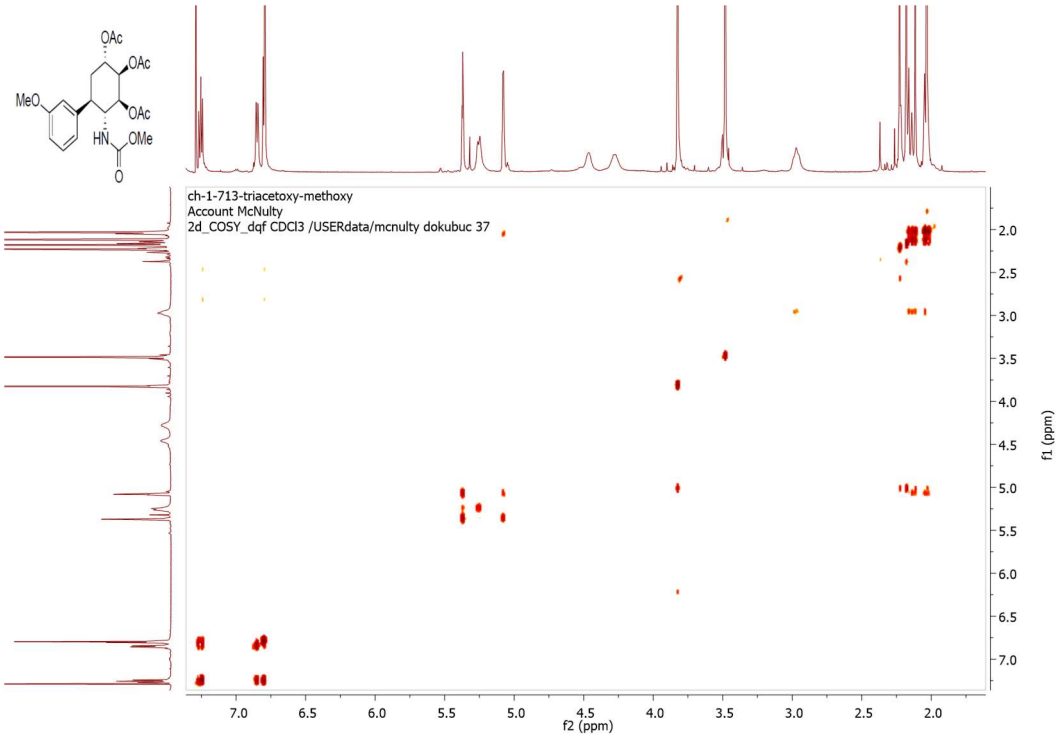

OH
MeO
OMe
NH
HN
ch-1-767-unketone
Account McNulty
2d_HSQC_ed_HC CDCl3 /USERdata/mcnulty dokubuc 17
f2 (ppm)
f1 (ppm)


ch-1-767-unketone
Account McNulty
1d_13C_dept135 CDCl3 /USERdata/mcnulty dokubuc 17
OH
MeO
OMe
NH
HN
f1 (ppm)

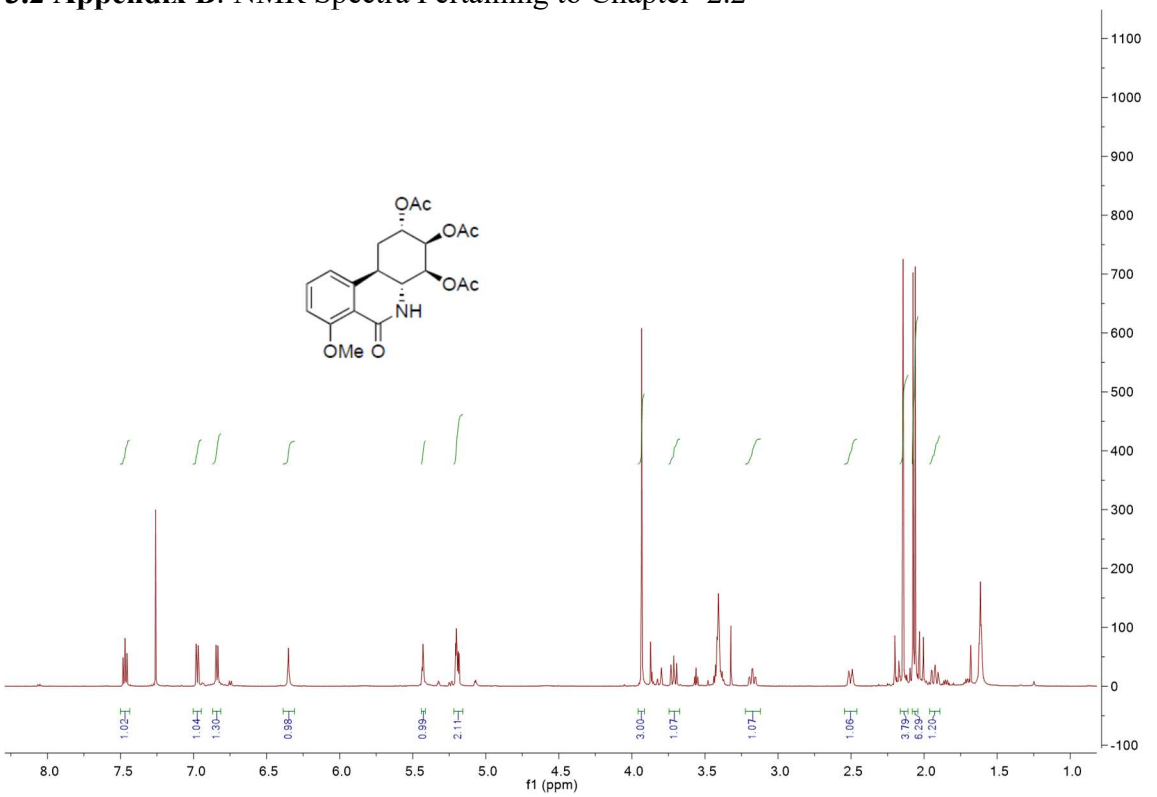

ch-1-707-hydroxy
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 51
MeO
OH
HN
OMe
O
1.29
2.08
1.09
1.02
1.02
1.00
1.03
0.76
1.25
3.25
3.19
1.00
2.08
f1 (ppm)

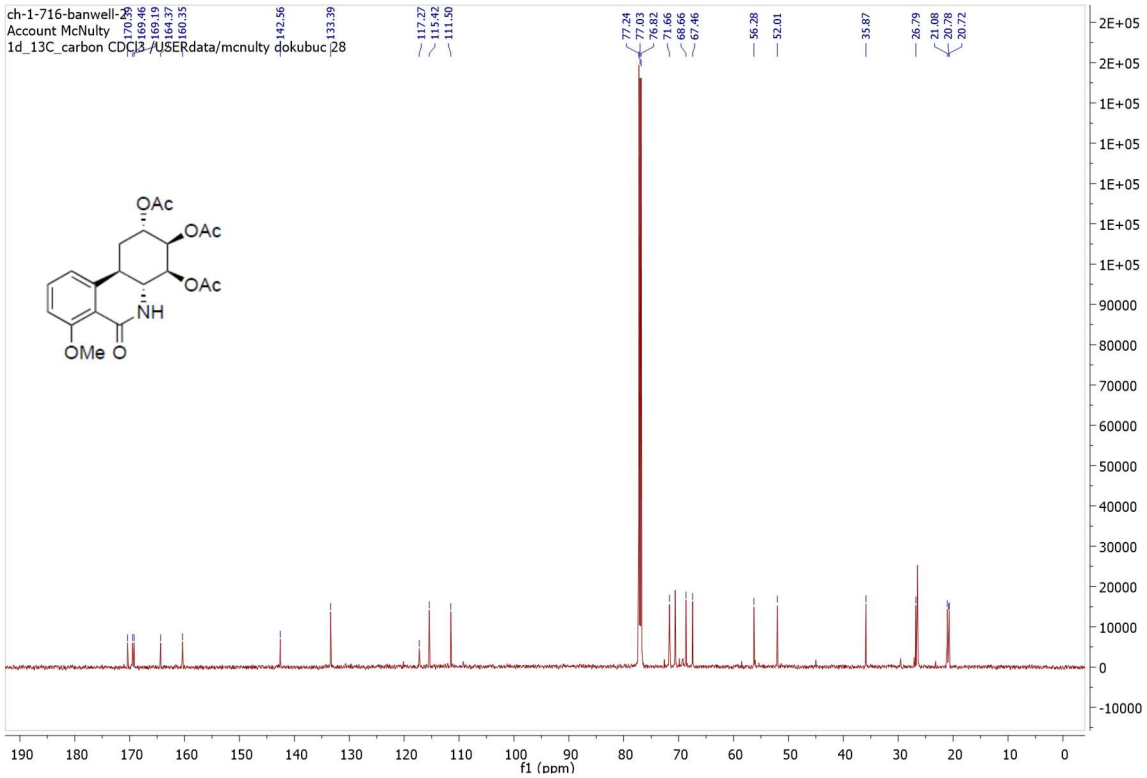

ch-1-707-hydroxy
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty dokubuc 51
160.03
158.54
142.65
130.05
129.40
126.92
120.00
113.51
112.57
77.25
77.04
76.83
74.09
58.60
55.22
52.44
44.82
34.96
MeO
OH
HN
OMe
O
f1 (ppm)

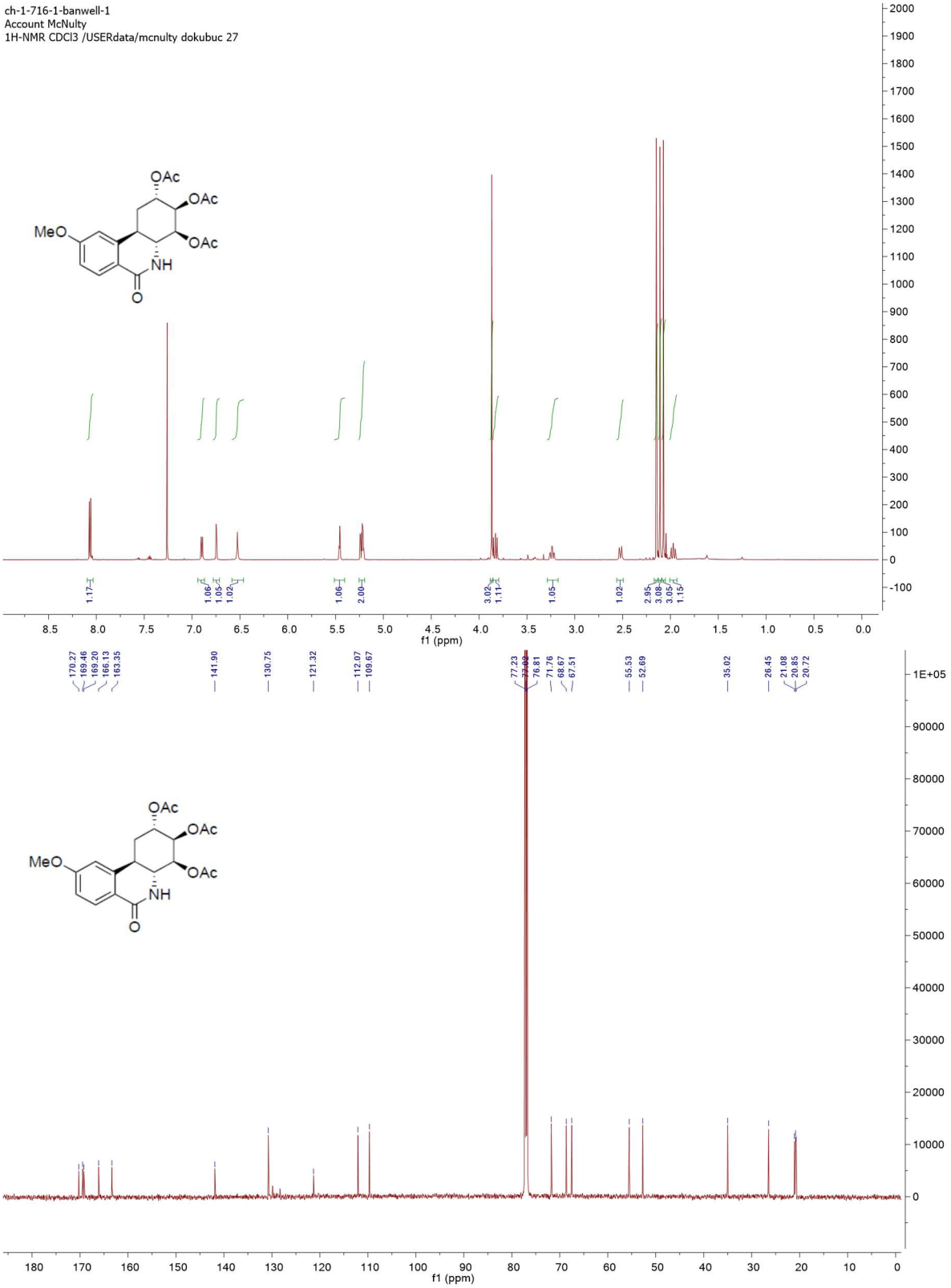

ch-1-713-triacetoxy-methoxy
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 37
OAc
OAc
OAc
MeO
HN
OMe
O
1.17
0.99
2.26
1.12
1.10
1.23
1.00
1.11
3.97
3.07
1.13
3.36
3.81
1.36
0.95
3.55
f1 (ppm)

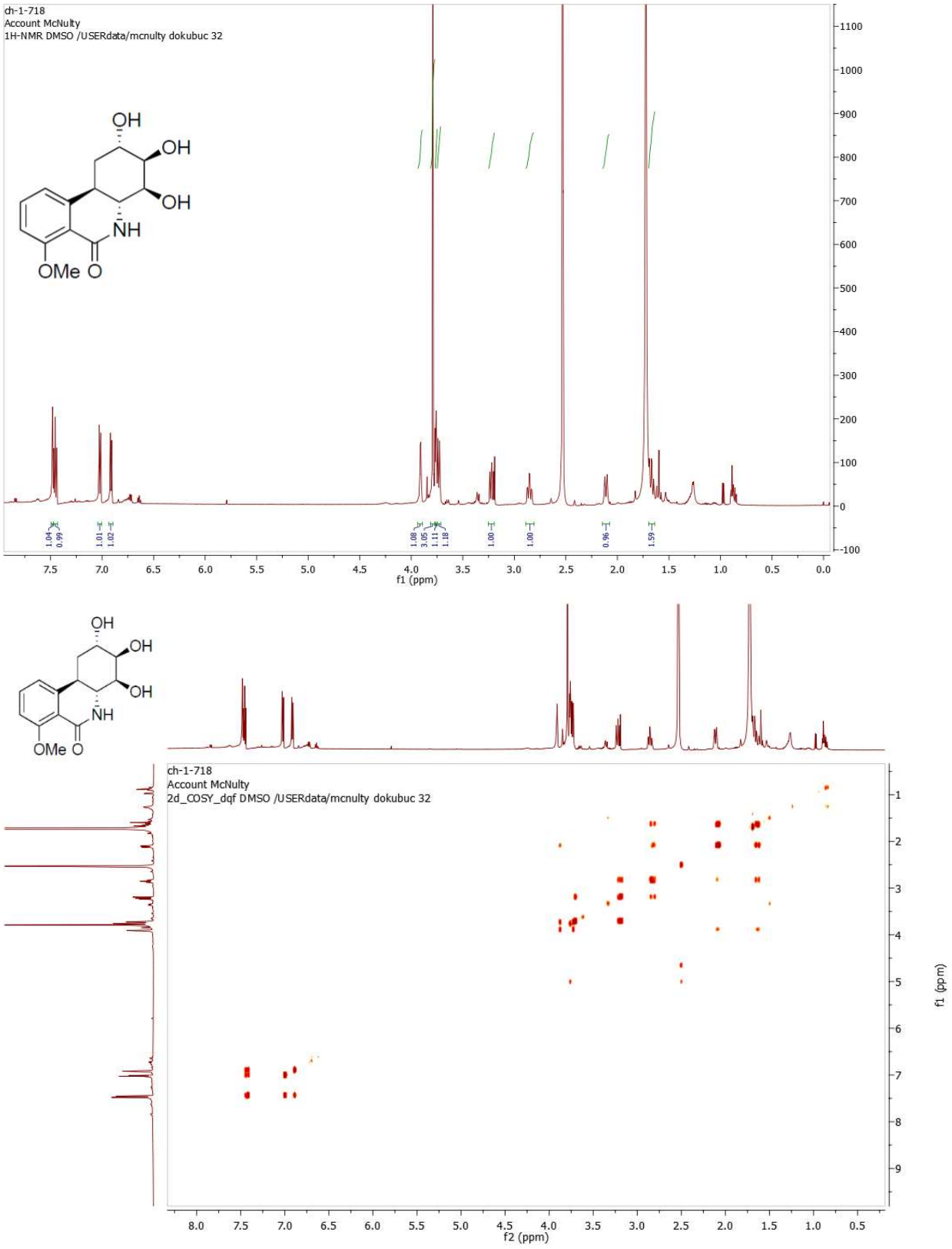

OAc
OAc
MeO
OAc
HN
OMe
O
ch-1-713-triacetoxy-methoxy
Account McNulty
2d_COSY_dqf CDCl3 /USERdata/mcnulty dokubuc 37
f2 (ppm)
f1 (ppm)

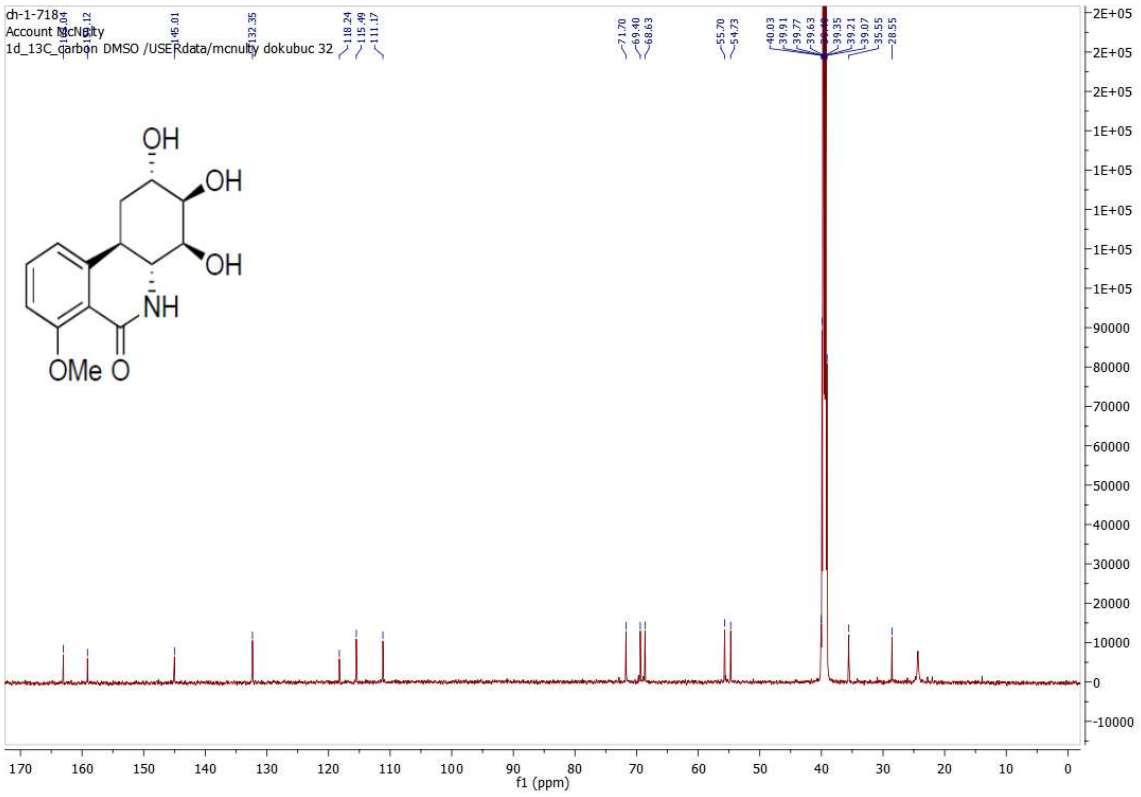
3.2 Appendix B: NMR Spectra Pertaining to Chapter 2.2

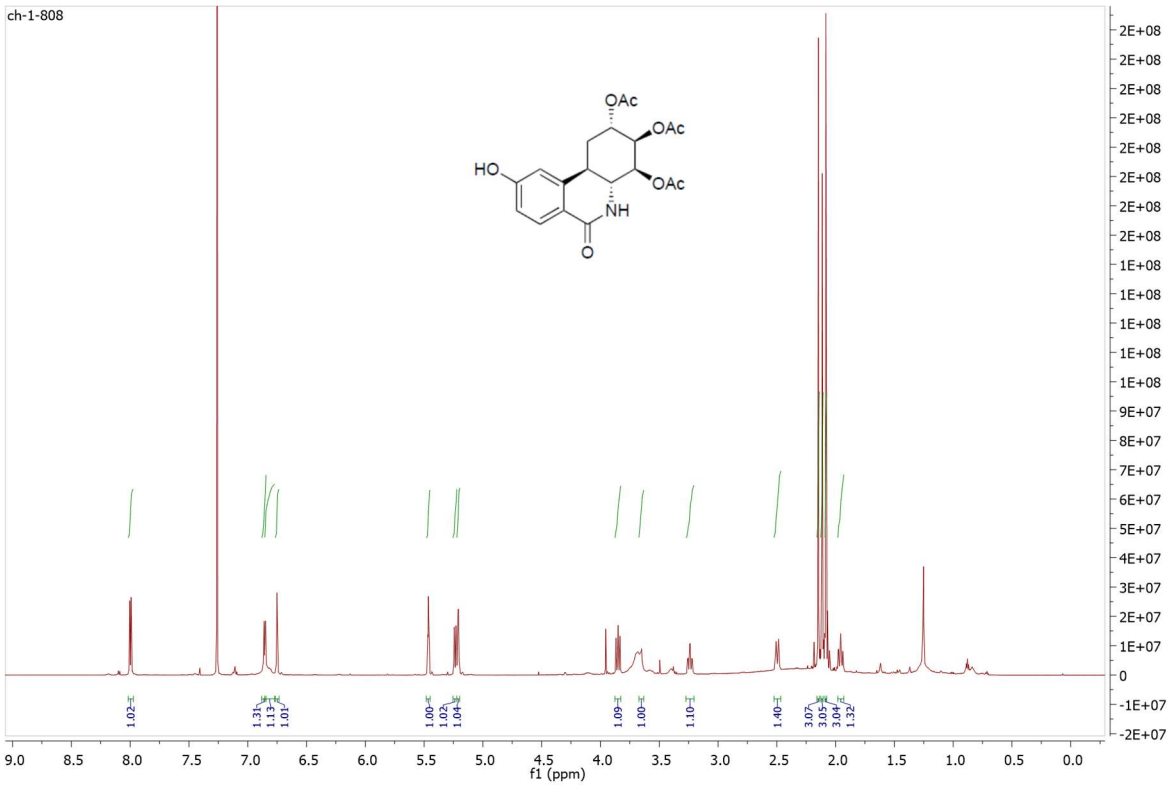


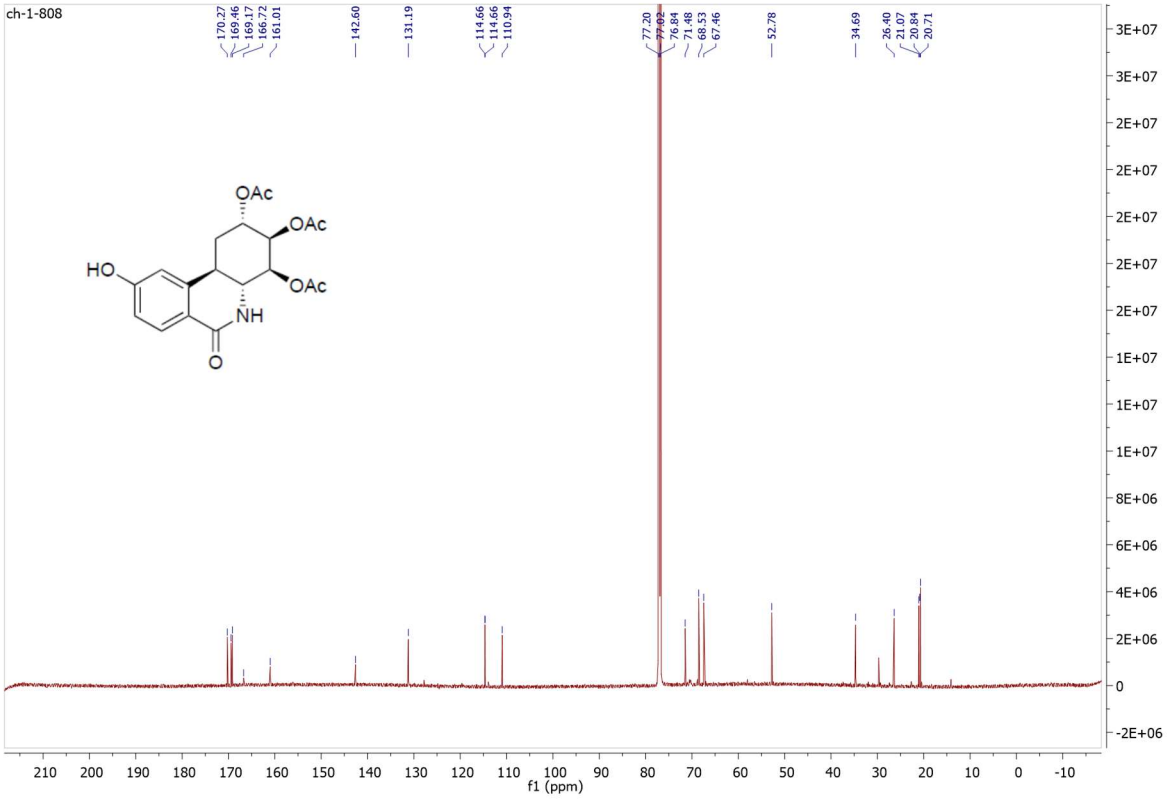


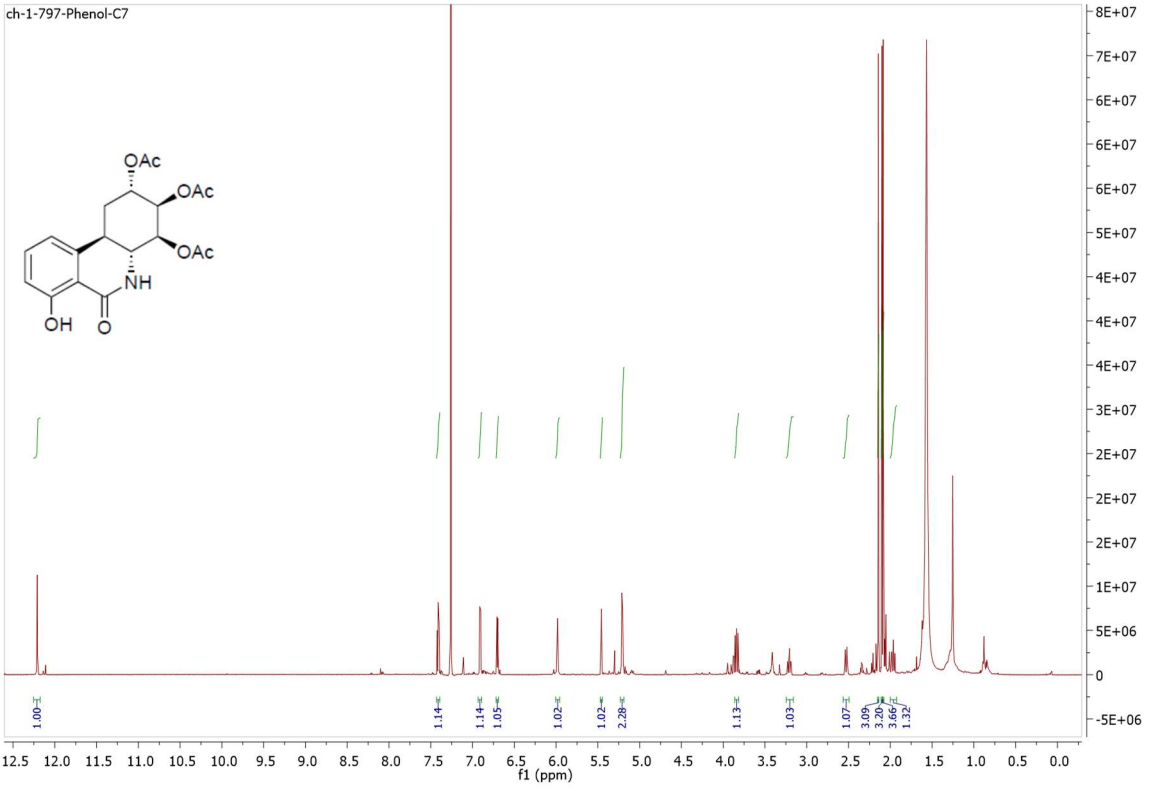

ch-1-716-1-banwell-1
Account McNulty
1H-NMR CDCl3 /USERdata/mcnulty dokubuc 27
OAc
OAc
OAc
MeO
NH
O
1.17
1.06
1.05
1.02
1.06
2.00
3.02
1.11
1.05
1.02
2.95
3.08
3.05
1.15
f1 (ppm)
170.27
169.46
169.20
166.13
163.35
141.90
130.75
121.32
112.07
109.67
77.23
76.81
71.76
68.67
67.51
55.53
52.69
35.02
26.45
21.08
20.85
20.72
f1 (ppm)

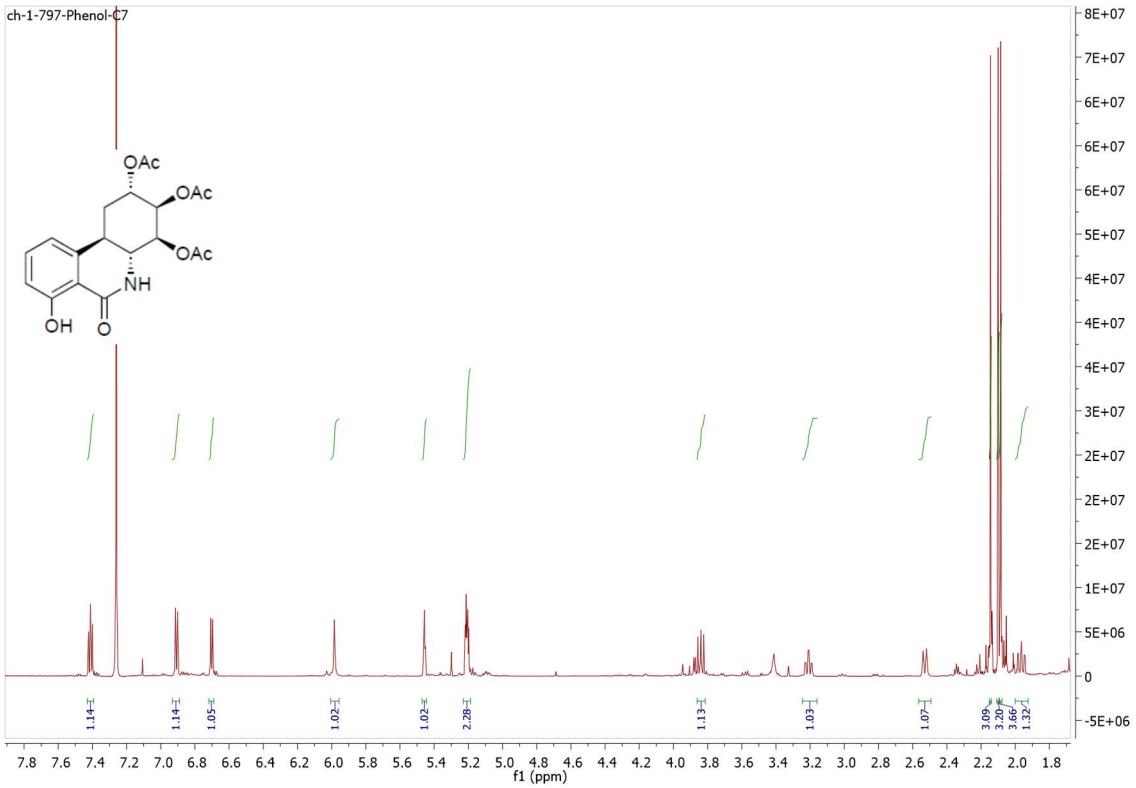

ch-1-718
Account McNulty
1H-NMR DMSO /USERdata/mcnulty dokubuc 32
OH
OH
OH
NH
OMe O
f1 (ppm)
ch-1-718
Account McNulty
2d_COSY_dqf DMSO /USERdata/mcnulty dokubuc 32
f2 (ppm)
f1 (ppm)

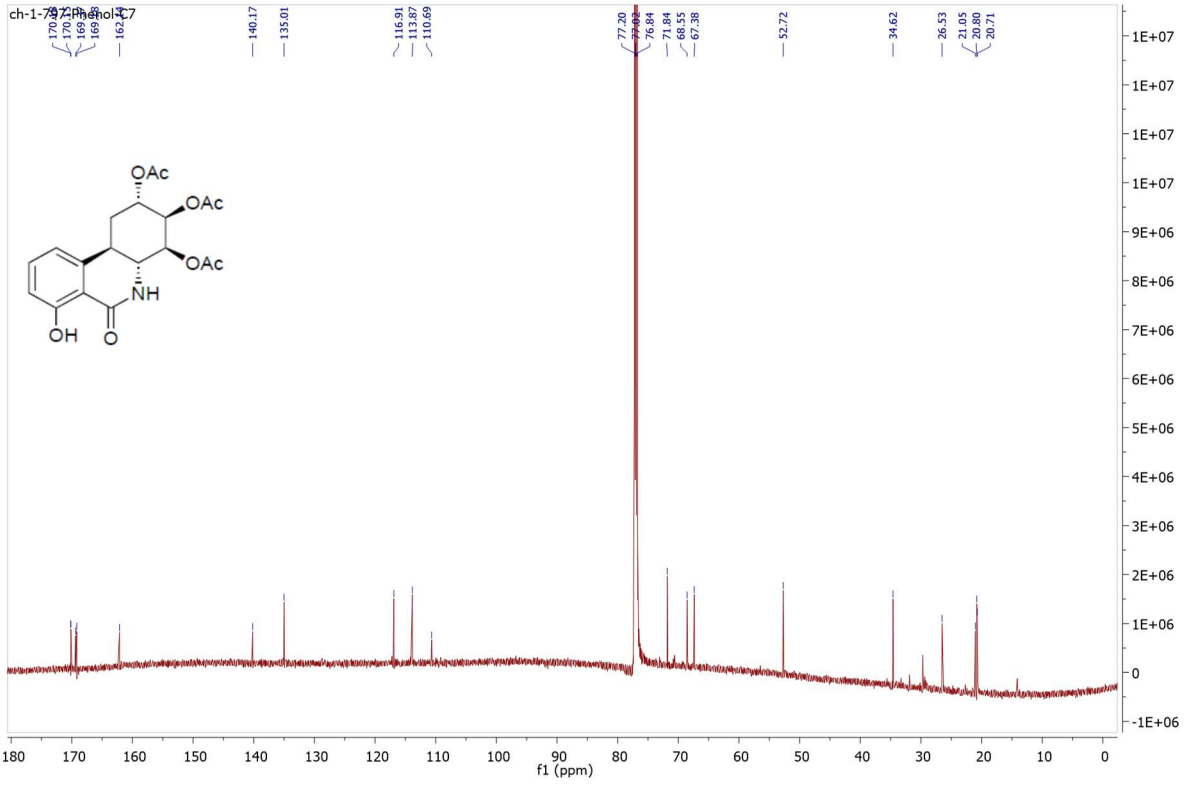

ch-1-718
Account McNulty
1d_13C_carbon DMSO /USERdata/mcnulty dokubuc 32
OH
OH
OH
NH
OMe O
f1 (ppm)

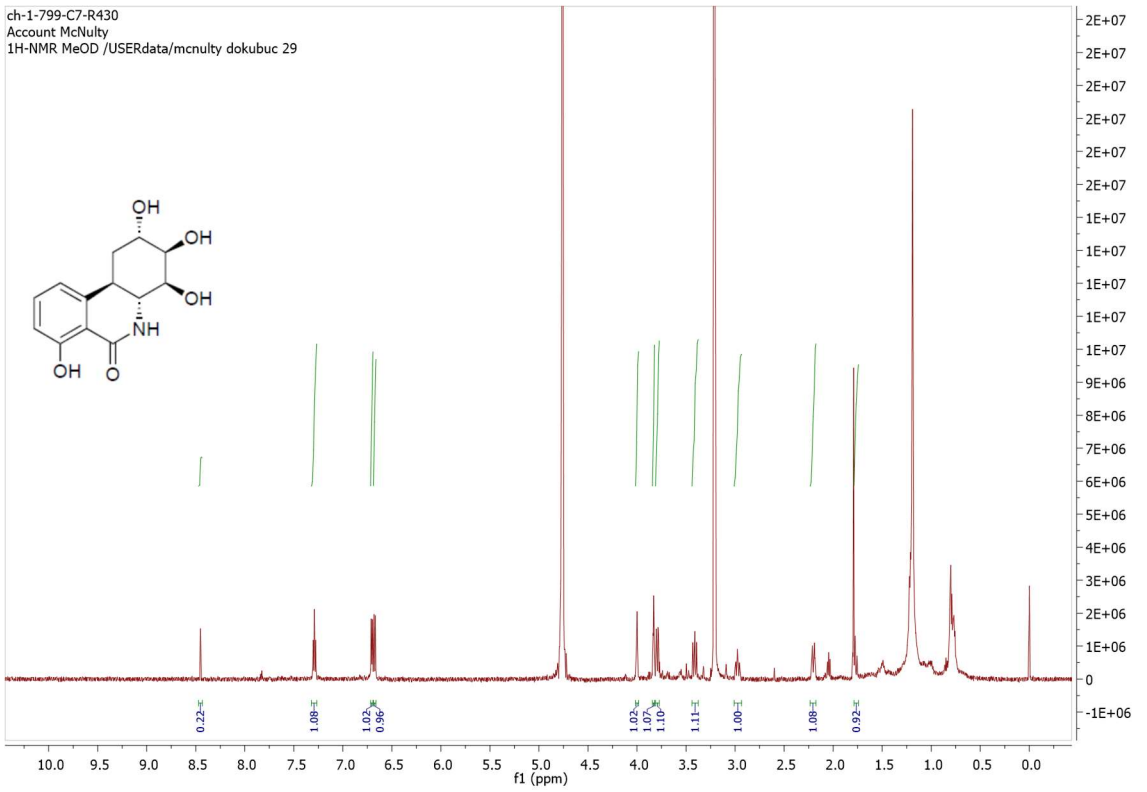

ch-1-808
OAc
OAc
OAc
HO
NH
O
f1 (ppm)

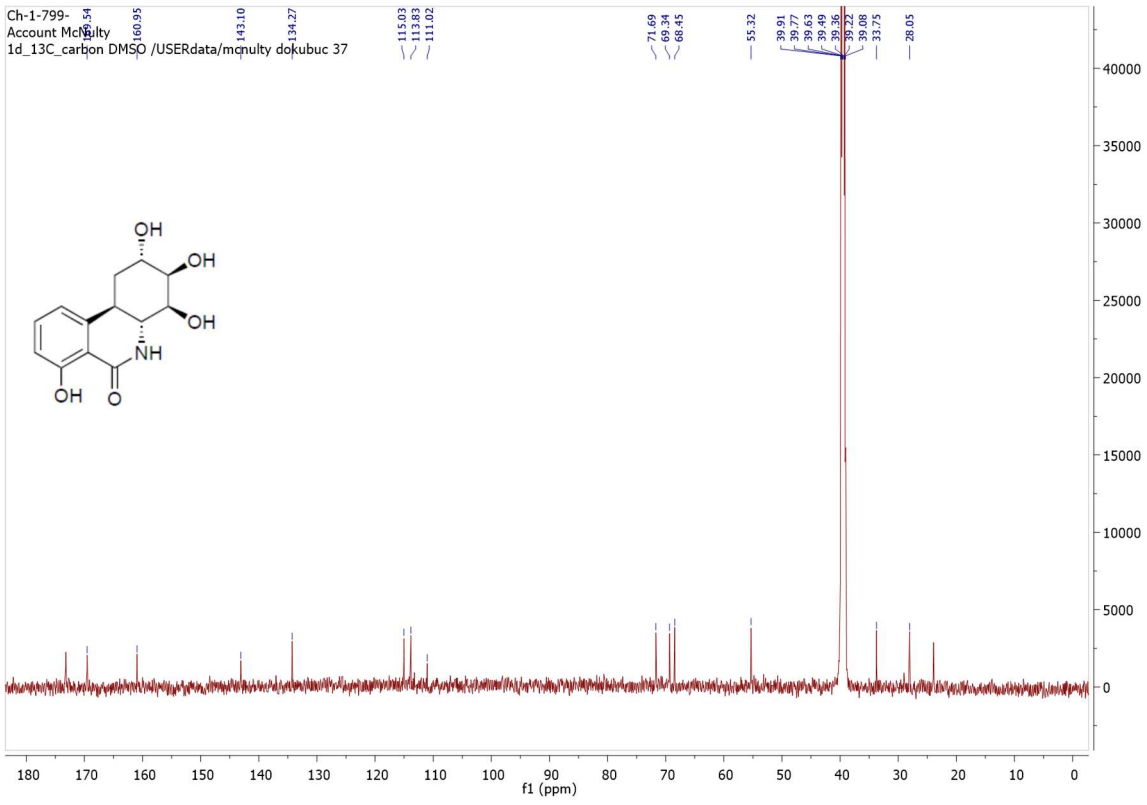

ch-1-808
170.27
169.46
169.17
166.72
161.01
142.60
131.19
114.66
114.66
110.94
77.20
77.02
76.84
71.48
68.53
67.46
52.78
34.69
26.40
21.07
20.84
20.71
OAc
OAc
OAc
HO
NH
O
f1 (ppm)


ch-1-797-Phenol-C7
OAc
OAc
OAc
NH
OH
O
1.00
1.14
1.14
1.05
1.02
1.02
2.28
1.13
1.03
1.07
3.09
3.20
3.66
1.32
f1 (ppm)

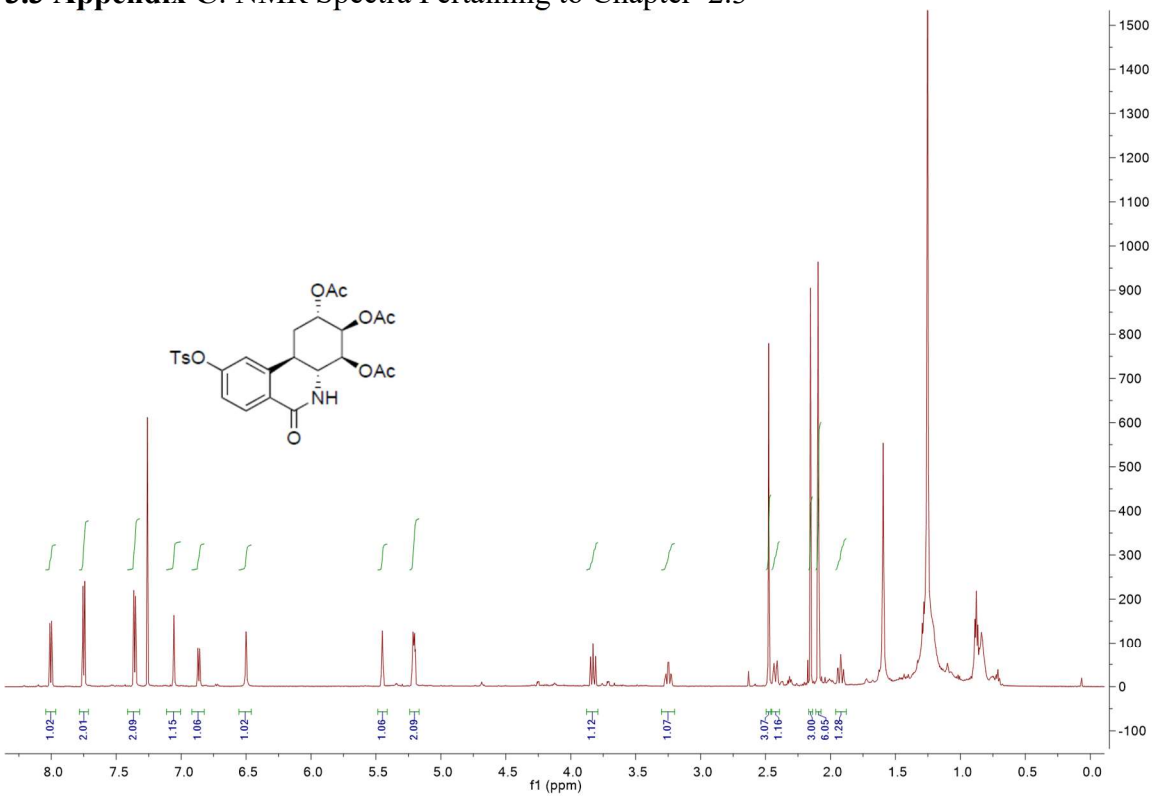

ch-1-797-Phenol-C7
OAc
OAc
OAc
NH
OH
O
f1 (ppm)

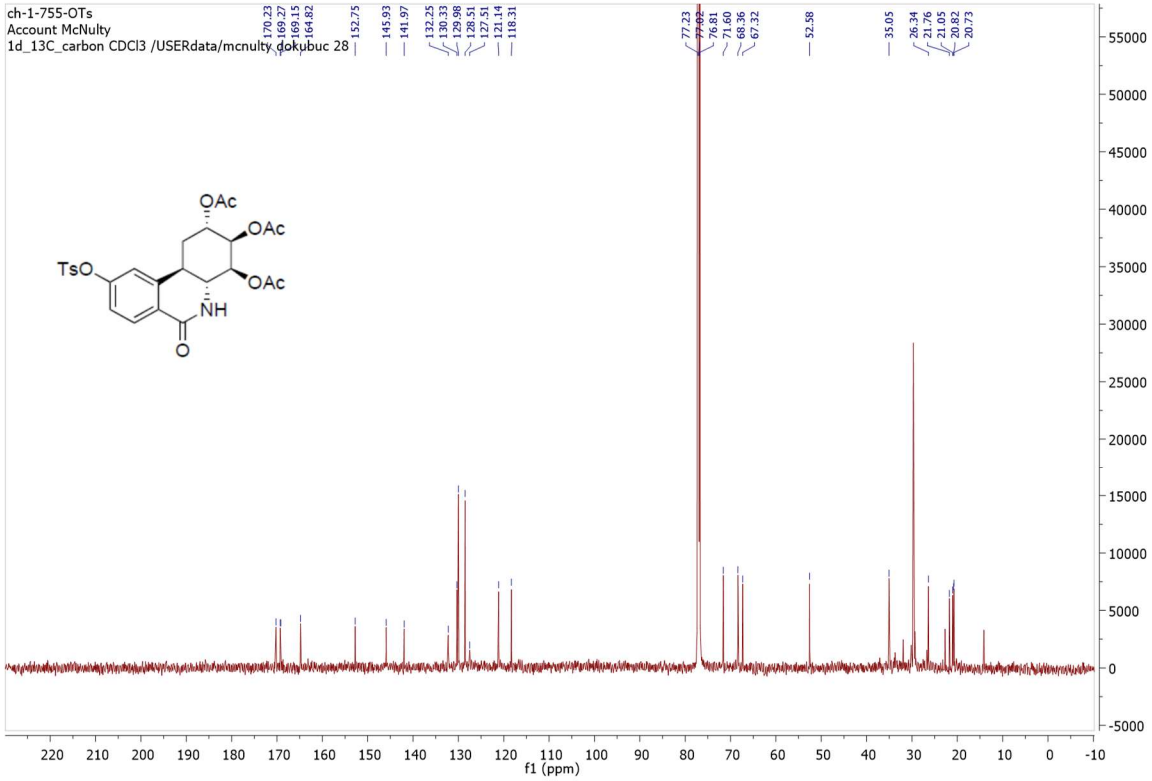

ch-1-797-Phenol-C7
OAc
OAc
OAc
NH
OH
O
f1 (ppm)

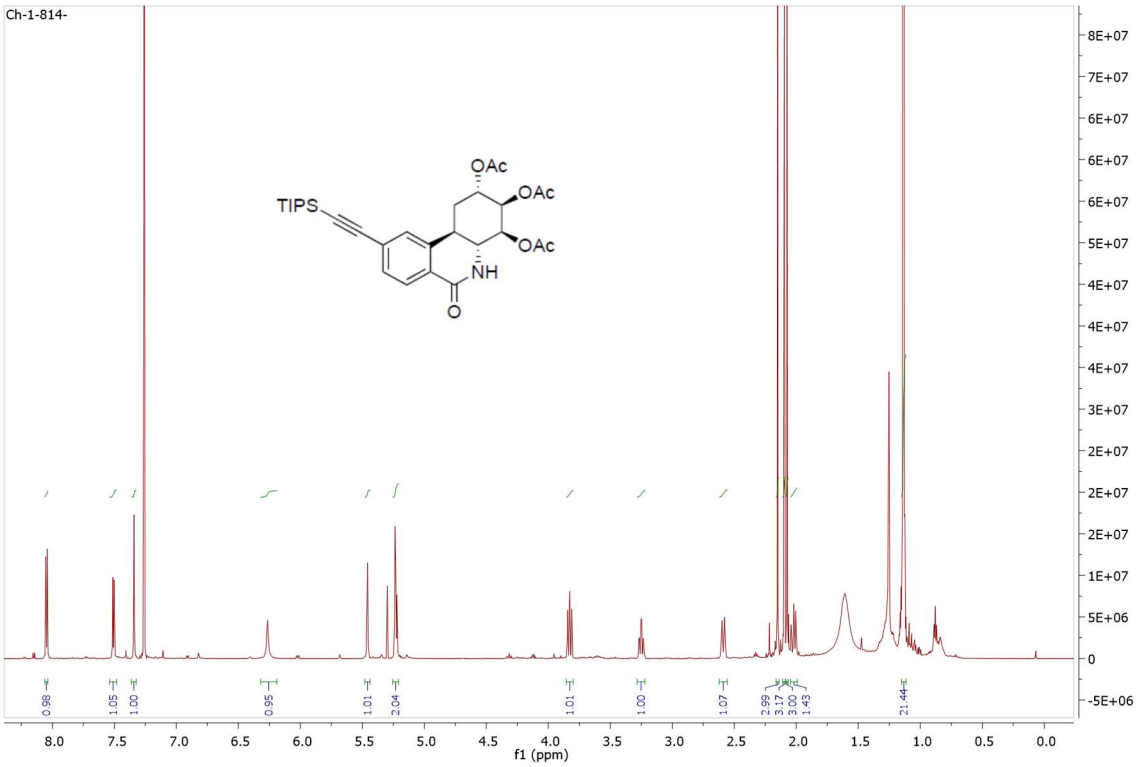

ch-1-799-C7-R430
Account McNulty
1H-NMR MeOD /USERdata/mcnulty dokubuc 29
f1 (ppm)

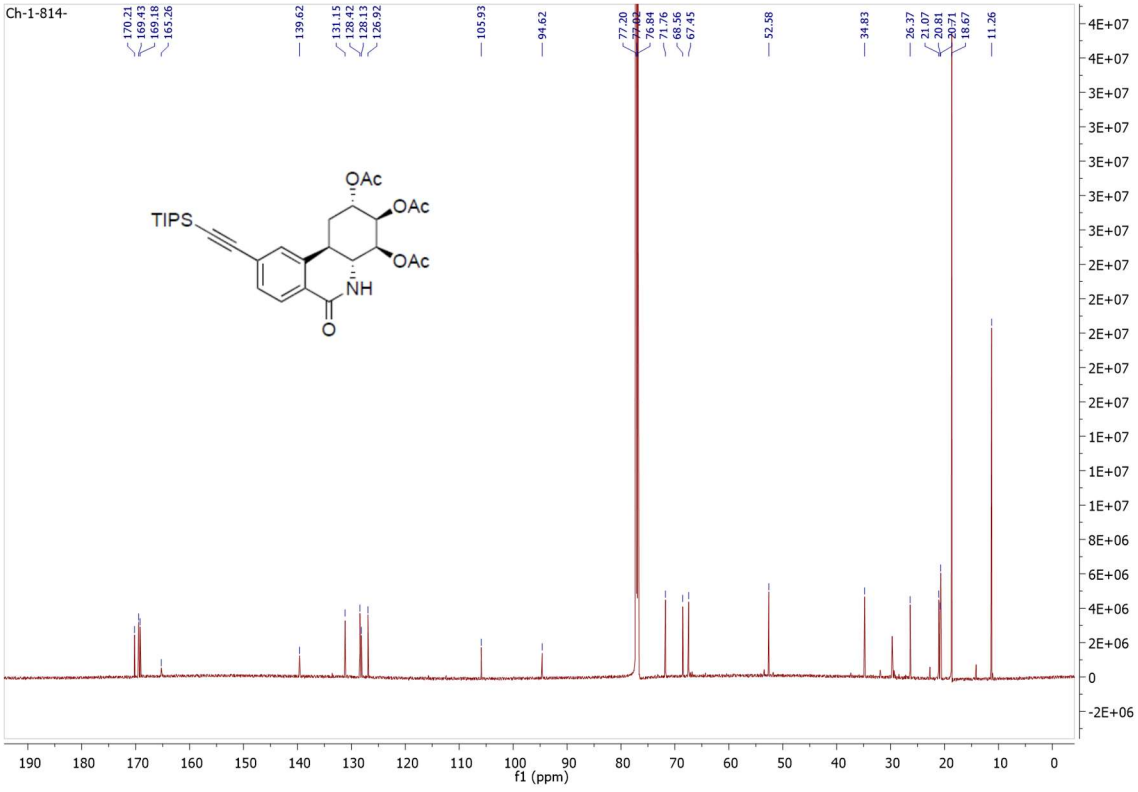

Ch-1-799-
Account McNulty
1d_13C_carbon DMSO /USERdata/mcnulty dokubuc 37
f1 (ppm)

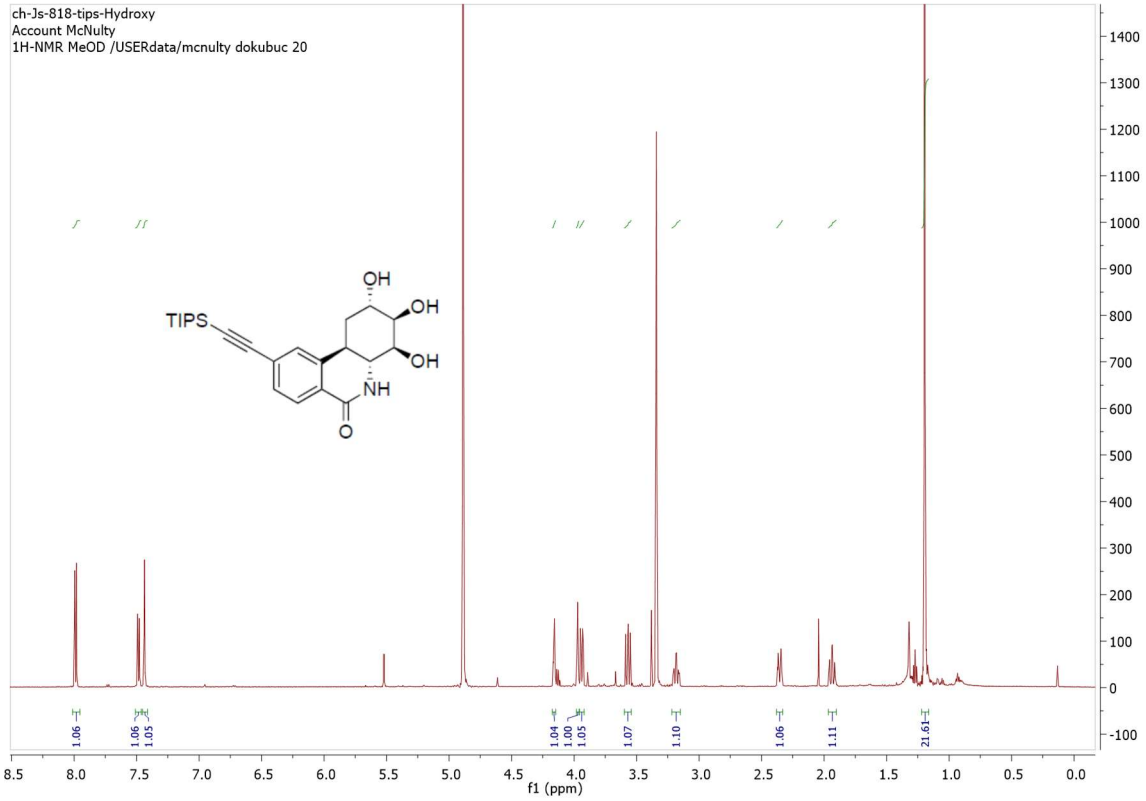
3.3 Appendix C: NMR Spectra Pertaining to Chapter 2.3

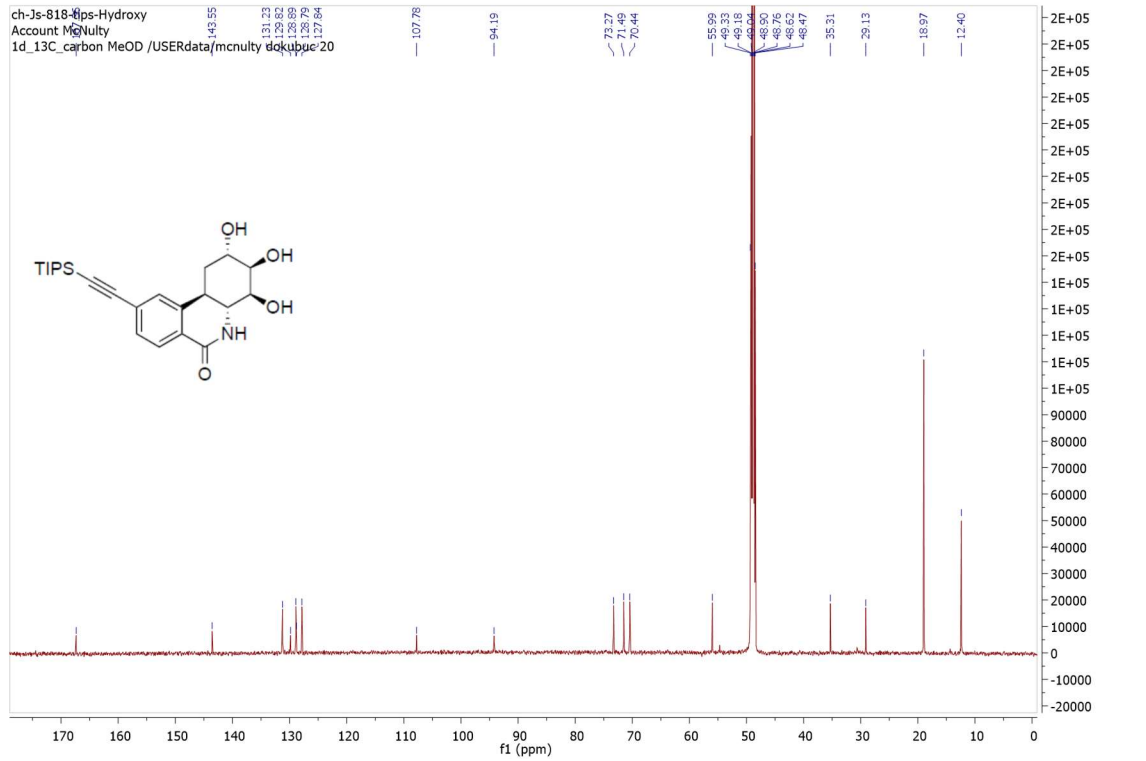


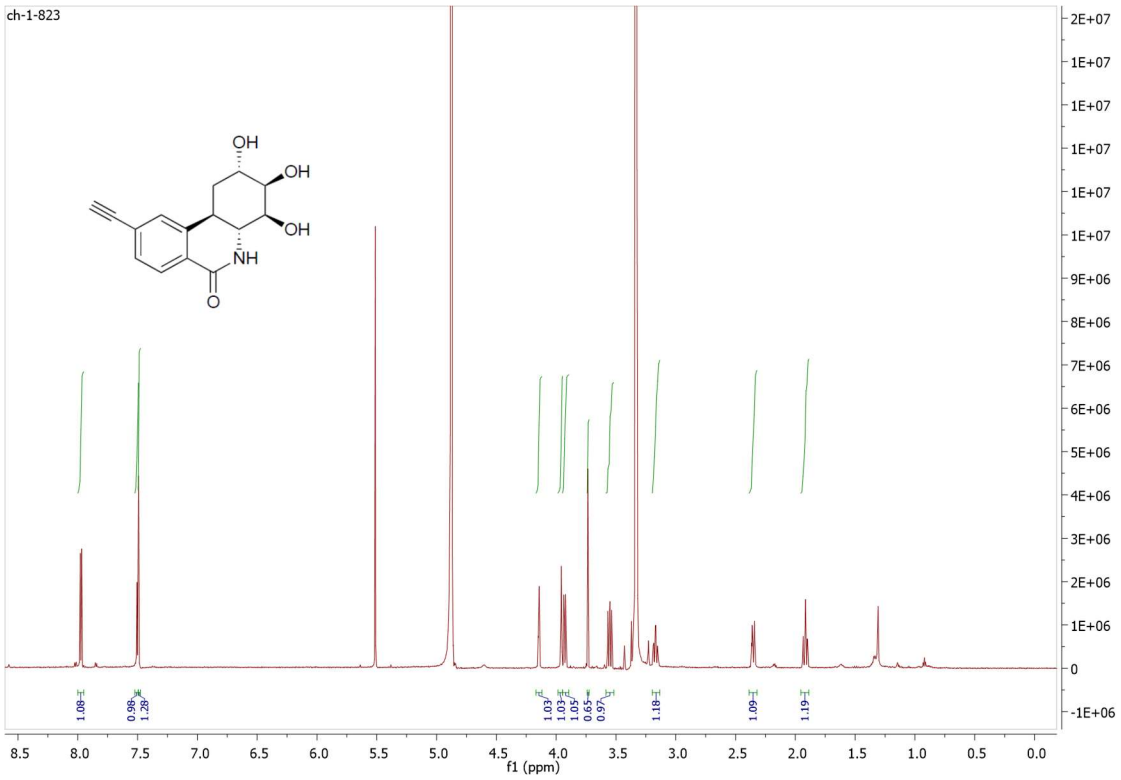



Ch-1-814-
TIPS
OAc
OAc
OAc
NH
O
f1 (ppm)

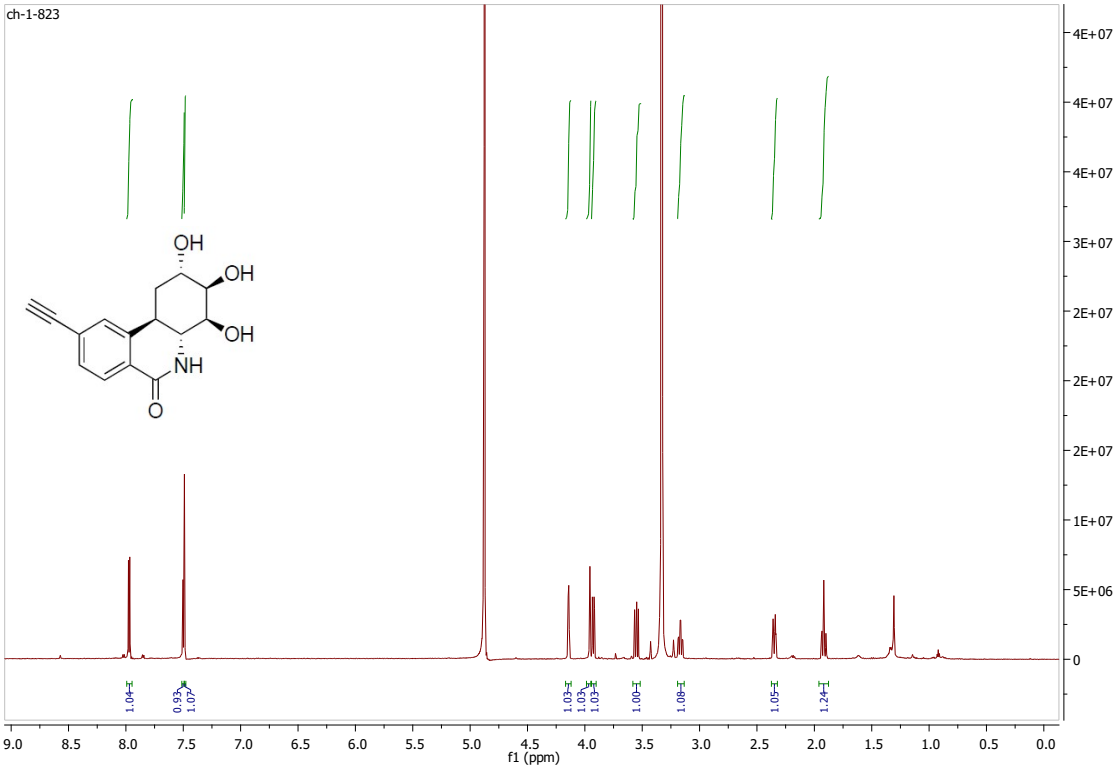

Ch-1-814-
170.21
169.43
169.18
165.26
139.62
131.15
128.42
128.13
126.92
105.93
94.62
77.20
77.03
76.84
71.76
68.56
67.45
52.58
34.83
26.37
21.07
20.91
20.71
18.67
11.26
TIPS
OAc
OAc
OAc
NH
O
f1 (ppm)


ch-Js-818-tips-Hydroxy
Account McNulty
1H-NMR MeOD /USERdata/mcnulty dokubuc 20
TIPS
OH
OH
OH
NH
O
f1 (ppm)

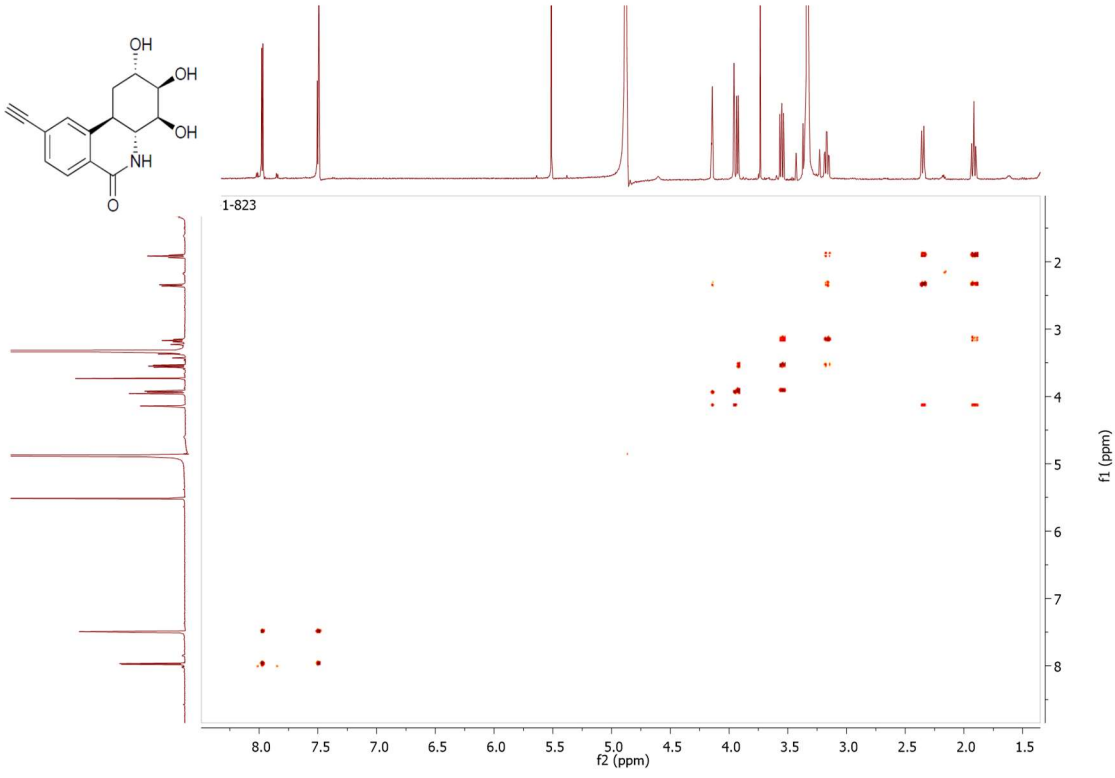

ch-Js-818-tips-Hydroxy
Account McNulty
1d_13C_carbon MeOD /USERdata/mcnulty dokubuc 20
TIPS
OH
OH
OH
NH
O
f1 (ppm)

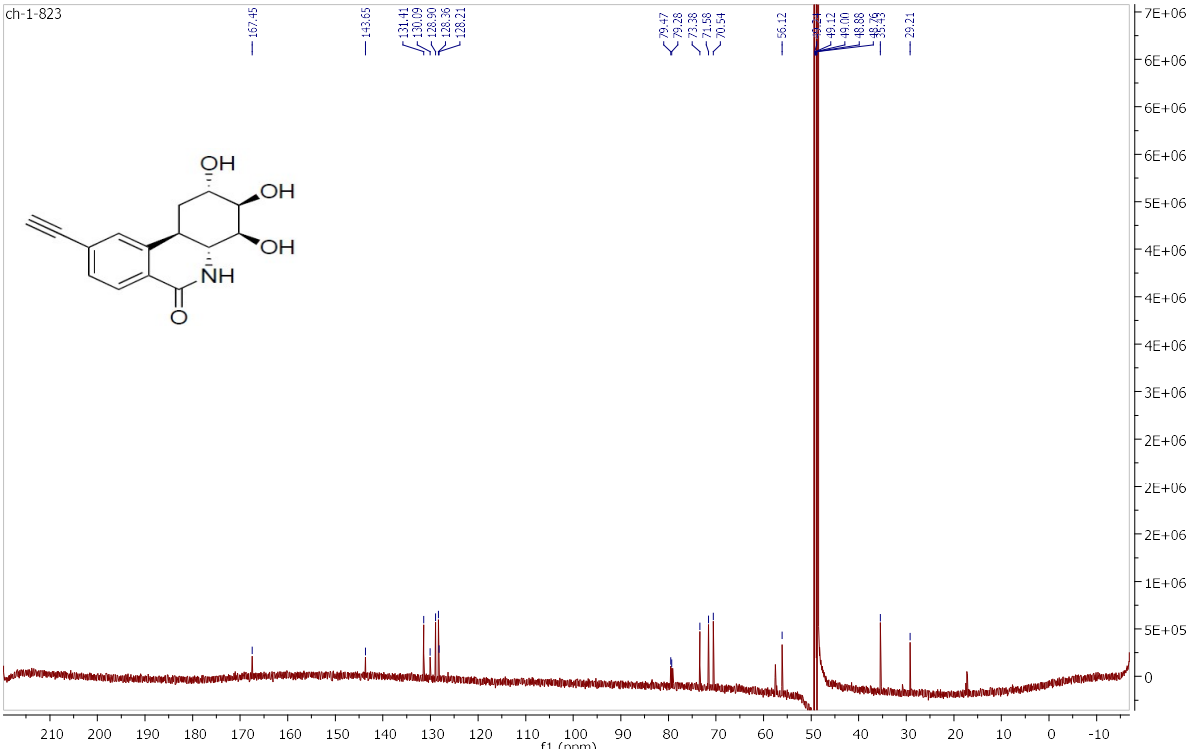


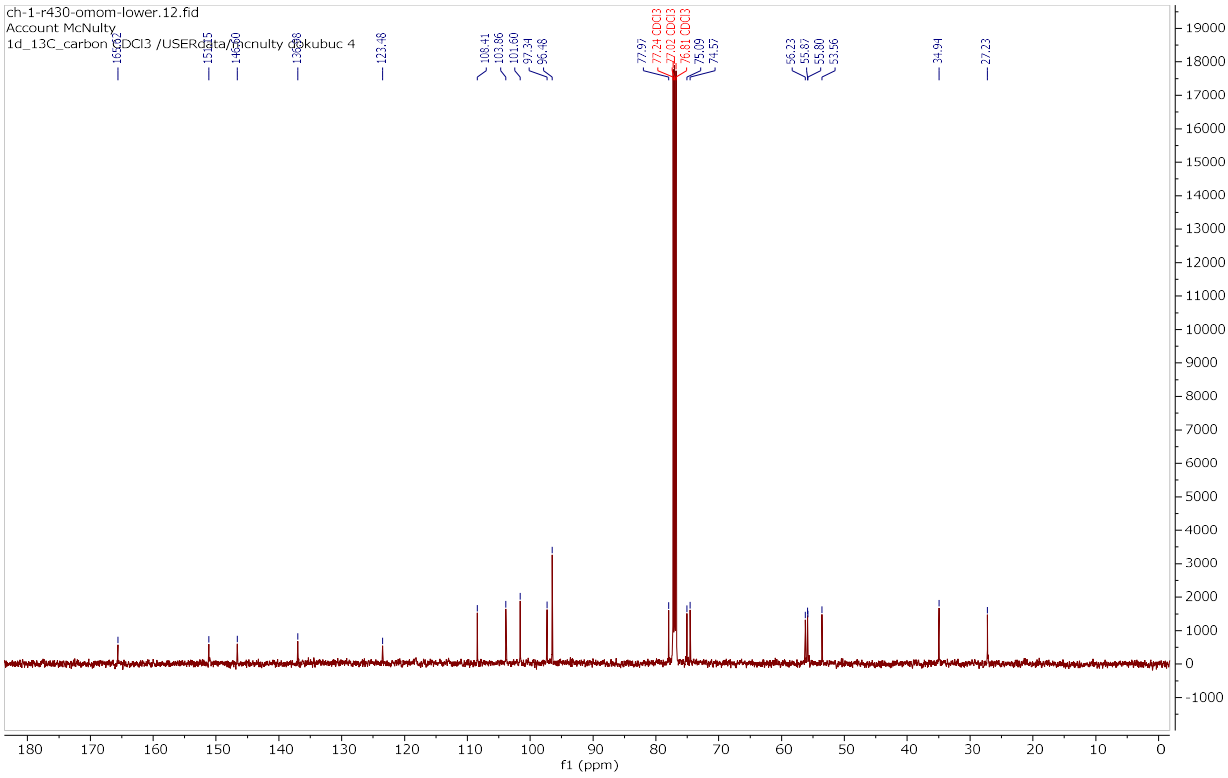
Alkyne -H was 33% was deuterated



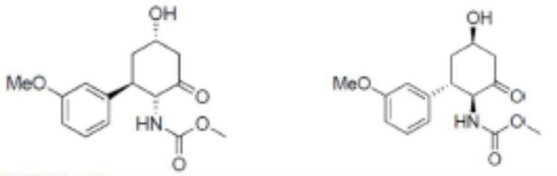
After 24 hours the alkyne H was completely deuterated

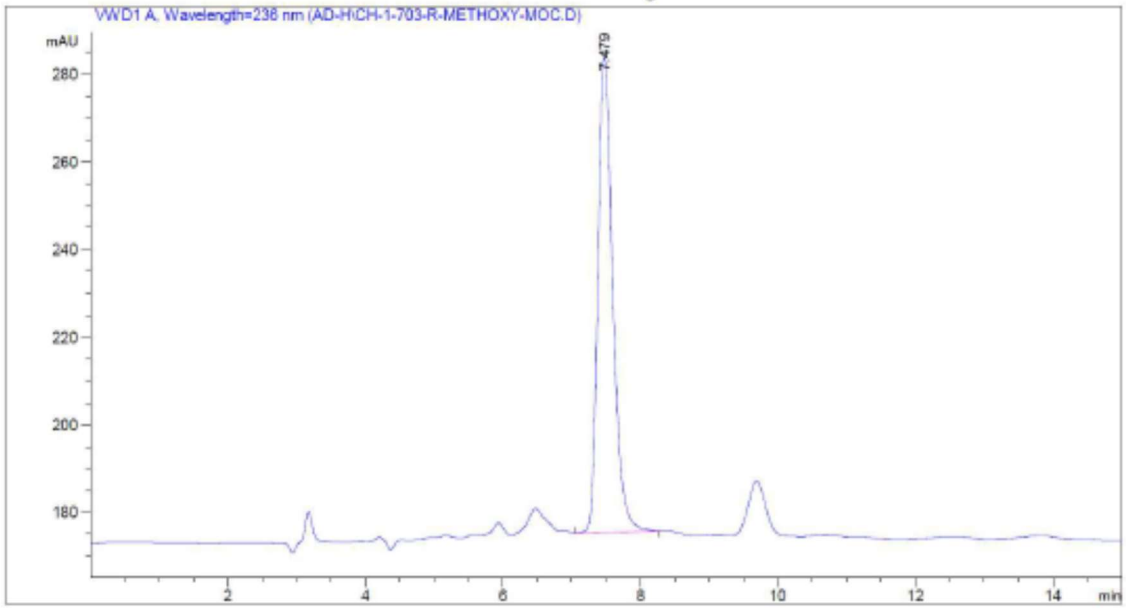



ch-1-823
167.45
143.65
131.41
130.09
128.90
128.36
128.21
79.47
79.28
73.38
71.58
70.54
56.12
49.12
49.00
48.88
35.45
29.21
OH
OH
OH
NH
O
f1 (ppm)


ch-1-r430-omom-lower.12.fid
Account McNulty
1d_13C_carbon CDCl3 /USERdata/mcnulty cookubuc 4
123.48
108.41
103.86
101.60
97.34
96.48
77.99
77.24 CDCl3
76.81 CDCl3
75.09
74.57
56.23
55.87
55.80
53.56
34.94
27.23
f1 (ppm)

3.4 Appendix D: HPLC Chromatograms Pertaining to Chapter 2.1





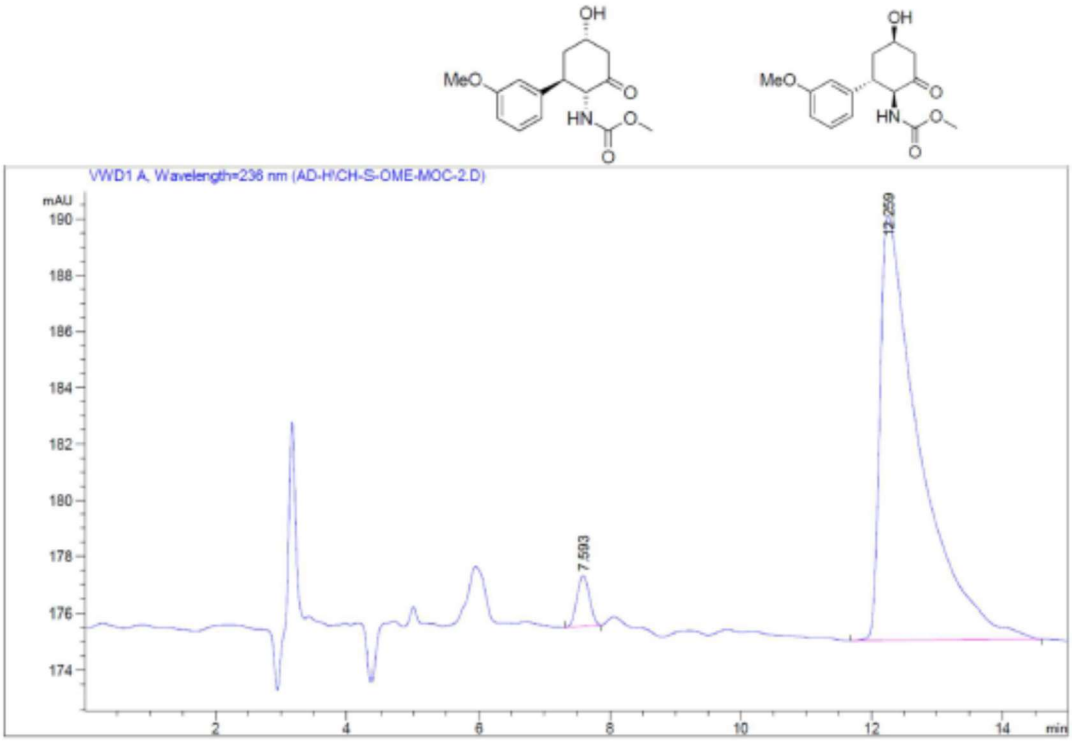

```
=====================================================================
                        Area Percent Report
=====================================================================

Sorted By             :      Signal
Multiplier            :      1.0000
Dilution              :      0.2000
Sample Amount:                :     20.00000  [ng/ul]   (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs


Signal 1: VWD1 A, Wavelength=236 nm

Peak RetTime Type  Width     Area      Height     Area
  #   [min]        [min]   [mAU*s]     [mAU]       %
----|-------|----|-------|----------|----------|--------|
   1   7.479 VB    0.2315 1641.82373  108.50411 100.0000

Totals :                  1641.82373  108.50411
```

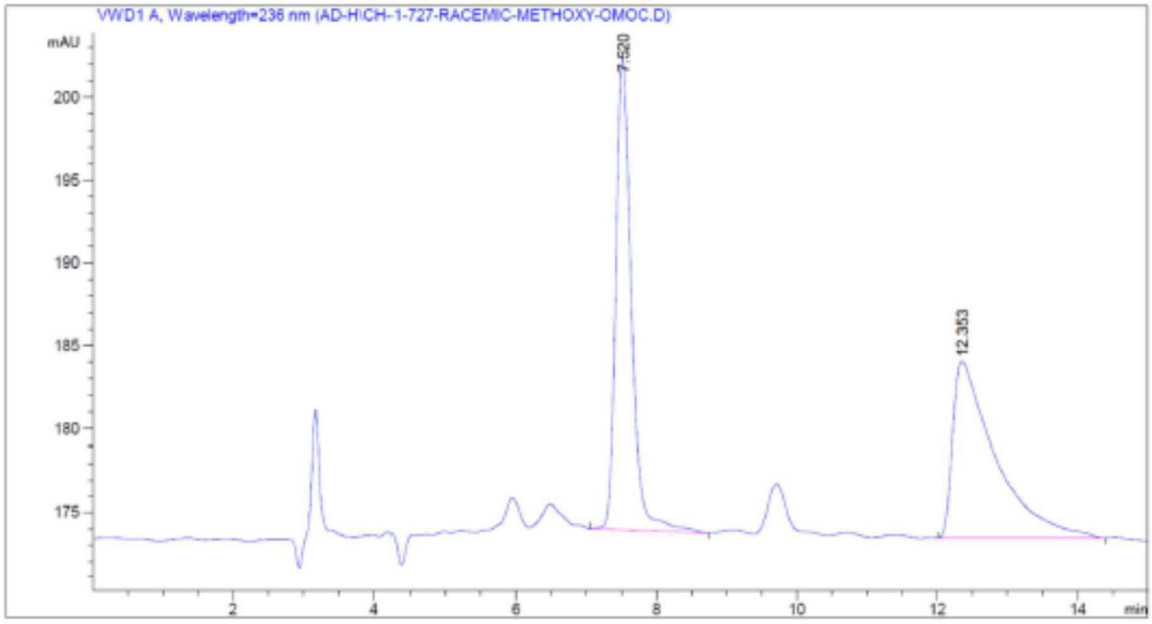



```
=====================================================================
                        Area Percent Report
=====================================================================

Sorted By             :      Signal
Multiplier            :      1.0000
Dilution              :      1.0000
Sample Amount:                :   5.00000e-1  [ng/ul]   (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs


Signal 1: VWD1 A, Wavelength=236 nm

Peak RetTime Type  Width     Area      Height     Area
  #   [min]       [min]   [mAU*s]     [mAU]       %
----|-------|----|-------|----------|----------|--------|
   1   7.593 BB    0.2115   23.55928    1.76734   3.5941
   2  12.259 BB    0.5754  631.93164   15.06400  96.4059
```




```
=====================================================================
                    Area Percent Report
=====================================================================

Sorted By             :      Signal
Multiplier            :      1.0000
Dilution              :      0.2000
Sample Amount:        :     20.00000  [ng/ul]   (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs


Signal 1: VWD1 A, Wavelength=236 nm

Peak RetTime Type  Width     Area      Height     Area
  #   [min]       [min]   [mAU*s]     [mAU]       %
----|-------|----|-------|----------|----------|--------|
   1   7.520 BB    0.2361  440.86270   28.39894  49.5915
   2  12.353 BB    0.5746  448.12558   10.61332  50.4085
```