

8-6-2009

# Synthesis of Amphibian Alkaloids and Development of Acetaminophen Analogues

Lei Miao  
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Synthesis of Amphibian Alkaloids and Development of Acetaminophen Analogues

A Dissertation

Submitted to the Graduate Faculty of the  
University of New Orleans  
in partial fulfillment of the  
requirements for the degree of

Doctor of Philosophy  
in  
The Department of Chemistry

by

Lei Miao

B.S., Chemistry, University of Science & Technology of China, China, 2003

August 2009

*To my family*

## **ACKNOWLEDGMENTS**

I would like to express my gratitude to my advisor, Professor Mark L. Trudell, for his support, guidance and encouragement during my doctoral study. His confidence and knowledge helped me to stay on track and achieve my goals.

I am also grateful to Professor Bruce C. Gibb , Professor Branco S. Jursic, Professor Guijun Wang and Professor John B. Wiley for being part of my committee.

I wish to thank Corinne Gibb for her kind help on the NMR all the time.

I wish to thank Professor Edwin D. Stevens for the X-Ray crystallographic data.

I wish to thank Professor Edwin Vedejs at the University of Michigan in Ann Arbor for the assistance during the aftermath of Hurricane Katrina.

I wish to thank Johanna and Kenneth Ludema for their kindness and help when I was in Ann Arbor, Michigan.

I wish to thank some of the past and present members of the Trudell group for their help and support: Dr. Liang Xu, Dr. Suhong Zhang, Dr. Shaine Cararas, Dr. Harneet Kaur, Hong Shu, April Noble, Xiaobo Gu, Andrea Forsyth, Abha Verma, Kim Slaughter and Dr. Murali Reddy.

Finally, I wish to thank my lovely wife Ying Long, my family and all my friends. They have been my real inspiration and my strength to succeed in my doctoral studies.

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## ABSTRACT

The focus of these studies has been toward the development of new synthetic methods and procedures for the synthesis of novel compounds with unique biological properties. This research has led to the development of two new synthetic strategies for the construction of two novel amphibian alkaloids. In addition, the efforts have led to the large-scale process for the preparation of a novel analgesic compound.

The regioselective ring opening of lactones ( $\delta$ -valerolactone and  $\gamma$ -butyrolactone) with aryllithium reagents is reported for the construction of a series of  $\delta$ -hydroxyarylketones and  $\gamma$ -hydroxyarylketones.

Both the *R* and *S* enantiomers of the amphibian alkaloid noranabasamine were prepared in >30% overall yield with 80% ee and 86% ee, respectively. An enantioselective iridium-catalyzed *N*-heterocyclization reaction with either (*R*)- or (*S*)-1-phenylethylamine and 1-(5-methoxypyridin-3-yl)-1,5-pentanediol was employed to generate the 2-(pyridin-3-yl)-piperidine ring system in 69-72% yield.

A cis-2,5-disubstituted pyrrolidine building block derived from (-)-Cocaine•HCl was prepared. We utilized this compound as a chiral building block for the formal synthesis of (+)-gephyrotoxin. Using this pyrrolidine building block, Kishi's intermediate was obtained

enantiospecifically in 15 steps and 9.4% overall yield.

A large-scale process for the preparation of the analgesic compounds SCP-123 and its sodium salt, SCP-123ss•monohydrate has been developed. The process for the preparation of SCP-123 required three synthetic steps with no chromatography, while the process for the preparation of SCP-123ss required four synthetic steps and no chromatography. The overall yields for both SCP-123 and SCP-123ss were 47% and 46%, respectively, and both compounds were obtained in exceptionally high purity (>99%).

Keywords: ketones, lactones, nucleophilic addition, ring-opening, enantioselective, noranabasamine, *N*-heterocyclization, gephyrotoxin, pyrrolidine, Kishi's intermediate, analgesic, acetaminophen, propacetamol, saccharin, hydrolysis, parenteral administration.

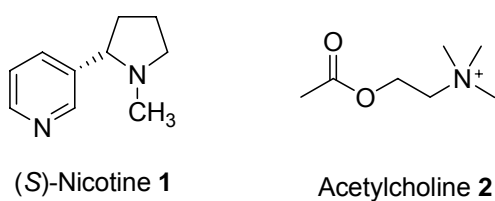
# CHAPTER 1

## INTRODUCTION

### 1.1. Nicotinic acetylcholine receptors

Nicotinic acetylcholine receptors (nAChRs) are cholinergic receptors. Cholinergic receptors form ionotropic channels (i.e. ligand-gated) in the cell membranes of certain neurons. As ligand-gated ion channel receptors, nAChRs are connected to an ion channel directly; but they don't employ a second messenger like metabotropic receptors.<sup>1</sup>

**Figure 1.1.** Structure of (*S*)-nicotine and acetylcholine



Muscarinic acetylcholine receptors (mAChRs) are acetylcholine receptors as well. Both nAChR and mAChR are triggered by the binding of the neurotransmitter acetylcholine (ACh). However, nicotinic receptors are also activated by nicotine. Likewise, muscarinic receptors are

also opened by muscarine.<sup>1-3</sup> Nicotinic acetylcholine receptors can be found in many tissues in the body, and are present in the peripheral nervous system (PNS) and the central nervous system (CNS).

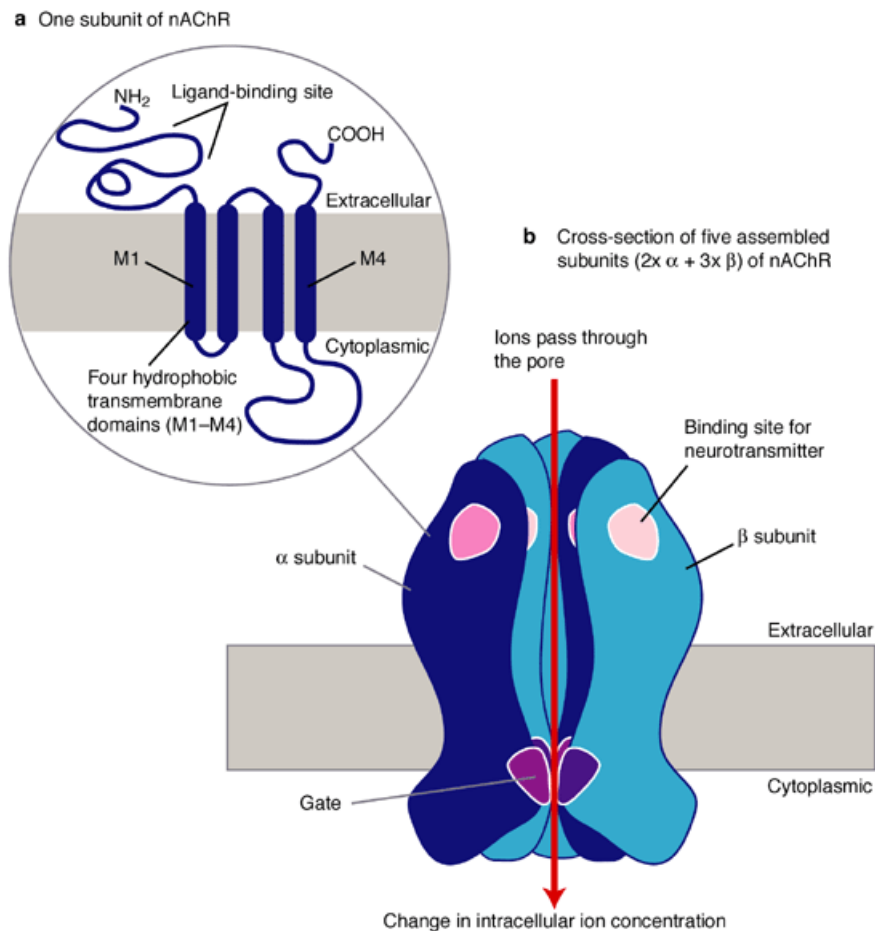
Although the nAChR has been cloned, the full mechanism by which these receptors function is still unclear. However, despite the lack of a definitive pharmacophore, nAChRs are attractive pharmacological targets for medication development for the treatment of a variety of disorders and disease states.

## **1.2. Nicotinic acetylcholine receptor structure and receptor subtypes**

Nicotinic receptors have a molecular mass of 290 kDa<sup>4</sup>. A central pore is surrounded by five subunits symmetrically (Figure 1.2).<sup>5,6</sup> The nAChRs are ligand-gated ion channels. The structures are composed with five ( $\alpha$  or  $\beta$  or  $\gamma$ ) subunits. The subunit can be homomeric (i.e. all  $\alpha$ ) or heteromeric (i.e. a mixture of  $\alpha$  and  $\beta$ , Figure 1.2).

One subunit of the nAChR comprises an N-terminal extracellular domain, four hydrophobic transmembrane domains (in dark blue), a long cytoplasmic loop and other shorter loops. The N-terminal extracellular domain is involved in the binding of (neurotransmitter) ligands. Four hydrophobic transmembrane domains (in dark blue) are named M1, M2, M3 and M4. The long cytoplasmic loop is connected to the M3 and M4 domains. The other shorter loops are between the other domains.

**Figure 1.2.** Structure of a neuronal nicotinic acetylcholine receptor (nAChR)<sup>7</sup>



As seen in a cross-section (Figure 1.2), each subunit has a gate region and a neurotransmitter binding site. As a ligand-gated ion channel, it binds a specific neurotransmitter [in this case, nicotine (**1**)]. The binding process changes the conformation of the receptor ending with opening of the channel. The channel pore is lined with charged amino acids. These amino acids selectively transfer the ions through cell membrane and into the cell. Compared to all the other ionotropic receptors (GABA<sub>A</sub> receptors, glycine receptors and the 5HT 3 serotonin



receptors), or the signature Cys-loop proteins, they all have similar properties.<sup>8</sup>

Based on the nicotinic receptor primary sites of expression, they are generally divided to two subtypes. The two subtypes are muscle type and neuronal type. The muscle type receptors are found at the neuromuscular junction. These receptors have two different forms. One is the embryonic form which is made up of  $\alpha 1$ ,  $\beta 1$ ,  $\delta$ , and  $\gamma$  subunits with a 2:1:1:1 ratio; The other is the adult form which is made up of  $\alpha 1$ ,  $\beta 1$ ,  $\delta$ , and  $\epsilon$  subunits in a 2:1:1:1 ratio.<sup>1-3,9</sup> The neuronal subtypes are combinations of twelve different nicotinic receptor subunits:  $\alpha 2$  through  $\alpha 10$  and  $\beta 2$  through  $\beta 4$ . The combinations are either homomeric or heteromeric. Several examples of the neuronal subtypes include  $(\alpha 4)_3(\beta 2)_2$ ,  $(\alpha 4)_2(\beta 2)_3$ , and  $(\alpha 7)_5$ . In the hydrophobic regions, all the subunits are the same, no matter whether it is a neuronal type receptor or a muscle type receptor.<sup>1</sup>

### **1.3. Nicotinic acetylcholine receptor subunits**

There have been 17 nAChR subunits identified to date. All the subunits belong to either muscle type or neuronal type subunits. Among those 17 subunits,  $\alpha 2$ - $\alpha 7$  and  $\beta 2$ - $\beta 4$  can be cloned in human cells. All the other genes are present in chick and rat genomes.<sup>10</sup> On the basis of similar protein sequence, the 17 nAChR subunits have been divided into 4 subfamilies (I-IV) (Table 1.1).<sup>11</sup> The subfamily III has been further divided into 3 types.

**Table 1.1.** Nicotinic acetylcholine receptor subunits

Neuronal type					Muscle type
I	II	III			IV
$\alpha 9, \alpha 10$	$\alpha 7, \alpha 8$	1	2	3	$\alpha 1, \beta 1, \delta, \gamma, \epsilon$
		$\alpha 2, \alpha 3, \alpha 4, \alpha 6$	$\beta 2, \beta 4$	$\beta 3, \alpha 5$	

#### 1.4. Nicotinic acetylcholine receptor notable variations

Each nicotinic receptor ion channel complex has five subunits to form the pentameric structure. So there are variety of combinations of these subunits. Among all the pentamers, several combinations are more prominent than others. For example,  $(\alpha 1)_2\beta 1\delta\epsilon$  (muscle type),  $(\alpha 3)_2(\beta 4)_3$  (ganglion type),  $(\alpha 4)_2(\beta 2)_3$  (CNS type) and  $(\alpha 7)_5$  (another CNS type) are shown in Table 1.2.<sup>12</sup>

**Table 1.2.** Comparison of notable nicotinic acetylcholine receptor

Receptor type	Location	Effect	Nicotinic agonists	Antagonists
Muscle type: ( $\alpha 1$ ) <sub>2</sub> $\beta 1\delta\epsilon$ <sup>12</sup> or ( $\alpha 1$ ) <sub>2</sub> $\beta 1\delta\gamma$	Neuromuscular junction	EPSP, mainly by increased Na <sup>+</sup> and K <sup>+</sup> permeability	acetylcholine <sup>1</sup> carbachol suxamethonium	$\alpha$ -bungarotoxin $\alpha$ -conotoxin tubocurarine <sup>1</sup> pancuronium
Ganglion type: ( $\alpha 3$ ) <sub>2</sub> ( $\beta 4$ ) <sub>3</sub>	autonomic ganglia	EPSP, mainly by increased Na <sup>+</sup> and K <sup>+</sup> permeability	acetylcholine <sup>1</sup> carbachol nicotine <sup>1</sup> epibatidine dimethylphenyl-piperazinium varenicline	$\alpha$ -bungarotoxin <sup>1</sup> mecamylamine trimetaphan hexamethonium bupropion dextromethorphan ibogaine 18-methoxycoronaridine
CNS type: ( $\alpha 4$ ) <sub>2</sub> ( $\beta 2$ ) <sub>3</sub>	Brain	Post- and presynaptic excitation, <sup>12</sup> mainly by increased Na <sup>+</sup> and K <sup>+</sup> permeability	nicotine epibatidine acetylcholine cytisine	mecamylamine methylcaconitine $\alpha$ -conotoxin
(another) CNS type: ( $\alpha 7$ ) <sub>5</sub>	Brain	Post- and presynaptic excitation, <sup>12</sup> mainly by increased Ca <sup>2+</sup> permeability	epibatidine dimethylphenyl-piperazinium	mecamylamine $\alpha$ -bungarotoxin <sup>1</sup>

### 1.5. Nicotinic acetylcholine receptor binding the channel

All ligand-gated ion channels are opened by an agonist chemical messenger binding to the nAChR. Acetylcholine is one of the endogenous agonists. There are other examples of

agonists of the nAChR including nicotine, epibatidine and choline. The binding site of acetylcholine is generated from the accumulation of amino acid residues. These residues are from both the  $\alpha$  and  $\beta$  subunits. In the case of homomeric receptors, those residues will be between two  $\alpha$  subunits near the N terminus in the extracellular domain.<sup>2</sup> All present subunits will exhibit a conformational change when an agonist binds to the site. The channel then opens to generate a pore with a diameter of about 0.65 nm.<sup>2,13</sup>

### **1.6. Nicotinic acetylcholine receptor opening the channel**

Nicotinic AChRs may have different conformational states that are interconvertible. With an agonist bound, the open process is stabilized and those states are desensitized. When the channel is open, positively charged ions can cross through the channel. Normally,  $\text{Na}^+$  enters the cell and  $\text{K}^+$  exits the cell. The net flow of ions with positive charge is inward. The nAChR is a cation channel with no selectivity. It means several ions with positive charge can move across the cell membrane.<sup>1</sup> The ion channels are permeable to  $\text{Na}^+$  and  $\text{K}^+$  and the level of permeability depends upon combinations of the subunits. These subunits are also permeable to  $\text{Ca}^{2+}$ .<sup>2</sup> The amount of  $\text{Na}^+$  and  $\text{K}^+$  the channels can accommodate in their pores (their conductance) varies from 50-110 pS (pico Siemens,  $10^{-12}$  S). The conductance is also dependent upon the individual subunit composition as well as the permeant ion.<sup>14</sup>

Interestingly, because  $\text{Ca}^{2+}$  can pass through some neuronal nAChRs, the release of other neurotransmitters can be affected at the same time.<sup>3</sup> The channel normally opens rapidly and

closes when the agonist diffuses away. The whole process generally takes about 1 millisecond.<sup>2</sup>

### **1.7. Nicotinic acetylcholine receptor effects**

The state of neurons is modified through the activation of receptors by nicotine. There are two main mechanisms during the process. First, the plasma membrane is depolarized by the movements of cations (an excitatory postsynaptic potential in neurons is generated in this case), and other voltage-gated ion channels are been activated as well. Second, when  $\text{Ca}^{+}$  moves along (directly or indirectly) the entrance, the action on different intracellular cascades will lead to the release of neurotransmitters or the regulation of some other gene activity.

### **1.8. Nicotinic acetylcholine receptor regulation of desensitization**

Katz and Thesleff first characterized ligand-bound desensitization of receptors in the nicotinic acetylcholine receptor.<sup>15</sup> The responsiveness of a receptor toward a stimulus generally decreased after prolonged or repeated exposure to a stimulus. It is called desensitization. The activation of second messenger-dependent protein kinases can modulate nAChR function through phosphorylation.<sup>16</sup> The desensitization of nAChR has been observed by protein kinase A and protein kinase C through phosphorylation.<sup>15,17</sup> Receptor desensitization has also been observed that the agonist itself results in an agonist-induced conformational change in the receptor after prolonged receptor exposure to the agonist.<sup>18</sup> When an agonist is bound with a

positive allosteric modulator, desensitized receptors can be restored back to a prolonged open state.<sup>19</sup>

### **1.9. Nicotinic acetylcholine receptor roles**

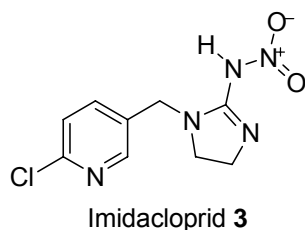
The subunits of the nicotinic receptors are the members of a multigene family (17 members in human). The combinations of those subunits create a huge number of different receptor subtypes. These receptor subtypes have highly variable kinetic, electrophysiological and pharmacological properties and exhibit different responses to nicotine dependent on very different effective concentrations. This functional diversity divides them in two major types of neurotransmission. One is classical synaptic transmission (wiring transmission) that involves acting on immediately neighbouring receptors and the release of a neurotransmitter at high concentrations. Second is paracrine transmission (volume transmission). This involves the release of neurotransmitters by synaptic buttons. These synaptic buttons try to reach their receptors by diffusion through the extra-cellular medium and may be distant. The muscular nicotinic receptor generally functions post-synaptically. This is one of the examples in which the nicotinic receptors exist in different synaptic locations.<sup>20</sup>

### **1.10. Nicotine**

Nicotine is one of the alkaloids that exist in the nightshade family of plants (*Solanaceae*).

Nicotine concentration is approximately 0.6-3.0% of the dry weight of tobacco.<sup>21,22</sup> It is biosynthesized in the roots, but accumulated in the leaves. One of its functions is to serve as an antiherbivore chemical defense with particular specificity to insects. In the past nicotine has been widely used as an insecticide. Today certain nicotine analogs, such as imidacloprid (**3**) (Figure 1.3) are still in use for such purpose.

**Figure 1.3.** Structure of imidacloprid



The name of nicotine came from the tobacco plant *Nicotiana tabacum*. In 1560, a French ambassador in Portugal named Jean Nicot de Villemain sent tobacco and seeds from Brazil to Paris and started their medicinal use. Then the plant was called *Nicotiana tabacum* after the name of Jean Nicot de Villemain. German chemists Posselt & Reimann first isolated nicotine from the tobacco plant in 1828, but it was not until 1843, that its chemical empirical formula was described by Melsens.<sup>23</sup> The structure of nicotine was confirmed by Garry Pinner in 1893. The first synthesis of nicotine was done by A. Pictet and Crepieux in 1904.

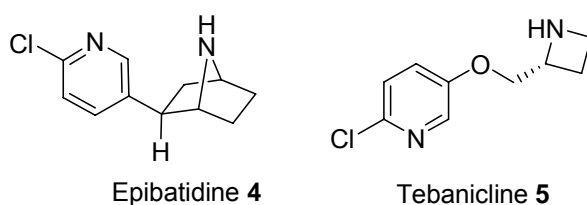
At low concentration (an average cigarette gives 1 mg of nicotine absorbed during smoking), nicotine plays a role as a stimulant in mammals. It is one of the main reasons that

tobacco smoking has the dependence-forming properties. American Heart Association says that: "Nicotine addiction has historically been one of the hardest addictions to break." The pharmacological and behavioral characteristics indicate that tobacco addiction is similar to addictions caused by other drugs such as heroin and cocaine.<sup>24</sup>

### 1.11. Epibatidine

Epibatidine (**4**) is an amphibian alkaloid that was first isolated from the skin of *Epipedobates tricolor*, a neotropical poisonous frog, found in the rainforest of Ecuador. The first isolation was done by John Daly at the National Institutes of Health. Later it was discovered that epibatidine was about 200 times more potent than morphine as an analgesic.<sup>25</sup> Because of the scarcity of epibatidine from natural sources, a number of research groups have reported the syntheses of this unique alkaloid.<sup>26</sup>

**Figure 1.4.** Structure of epibatidine and tebanicline



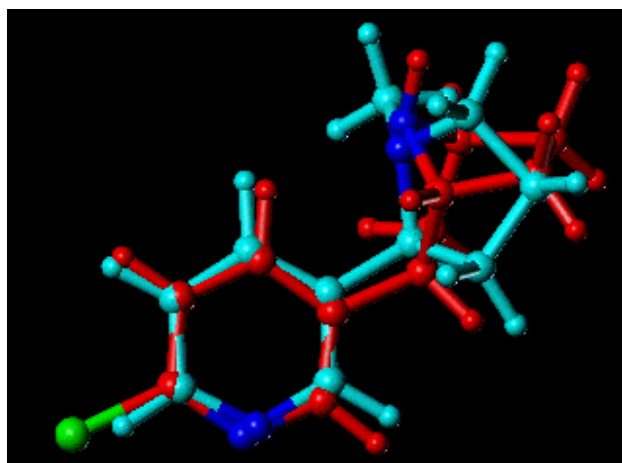
Studies with epibatidine have shown that the compound acts more like nicotine and



shows binding and activation of nicotinic acetylcholine receptors and has very little affinity at opioid receptors. Because of the toxicity of epibatidine, it is not an ideal candidate for clinical use. However the compound has provided new leads for the design of new analgesics.<sup>27</sup>

Among all the epibatidine derivatives that have been tested, the most promising one reported up to date is ABT-594 [Tebanicline (**5**), Abbott Laboratories] (Figure 1.4). ABT-594 was found to be 50 times more potent than morphine and there was no paralysis or depression of muscle action indicated in animal tests. Phase II clinical trials have been completed in Europe. When humans with neuropathic pain were treated by ABT-594, clinical efficacy did show up as well as some unacceptable incidence of gastrointestinal side effects.<sup>28</sup> As a result further development was halted. More research in this area is ongoing.<sup>29</sup>

**Figure 1.5.** Superposition of nicotine and epibatidine<sup>25</sup> [Nicotine (cyan) and epibatidine (red). Nitrogens are blue; chlorine is green.]



Nicotine and epibatidine have very similar structures. They both have a pyridine ring. They both have a basic nitrogen that contains ring system linked to the pyridine ring. In nicotine, there is one carbon between the nitrogen and pyridine ring, while the linkage in epibatidine is two carbons. The basic nitrogen is a part of a five-membered ring in both molecules. The five membered ring is a part of the azabicycloheptane structure in epibatidine. Dukat *et al.* have explored energy-minimized molecular models of these two compounds.<sup>30</sup> The optimized structures of nicotine and epibatidine could be overlayed with similar position of the various structural features in space (Figure 1.5). This modeling experiment was the first to show in a three dimensional fashion that epibatidine and nicotine may interact with similar receptor features.

### **1.12. Half maximal inhibitory concentration (IC<sub>50</sub>)**

The half maximal inhibitory concentration (IC<sub>50</sub>) is a measure to determine the effectiveness of a compound in performing biological or biochemical inhibitor function. This quantitative measure shows the concentration of a special drug or other compounds (inhibitors) that is required to inhibit an existing biological process (or parts of a process, for instance: an enzyme, cell, cell receptor microorganism) by half. This is called the half maximal (50%) inhibitory concentration (IC) of a substance (50% IC, or IC<sub>50</sub>). It is widely used to measure the potency of an agonist or antagonist drug in pharmacological research. The pIC<sub>50</sub> is converted from IC<sub>50</sub> by a log function of IC<sub>50</sub>. In the scale of pIC<sub>50</sub>, higher value shows exponentially

greater potency of a drug candidate. Referring to the FDA,  $IC_{50}$  demonstrates the concentration of a drug that is needed for 50% inhibition *in vitro*. While  $EC_{50}$  demonstrates the plasma concentration needed for gaining 50% of a maximum effect *in vivo*.  $IC_{50}$  and  $EC_{50}$  are comparable for an agonist drug.

Functional antagonist assays are used to determine the  $IC_{50}$  of a drug. A dose-response curve needs to be constructed. The effect of different concentrations of antagonist on reversing agonist activity is required for examination. These procedures determine the  $IC_{50}$  of a drug. The values of  $IC_{50}$  can be figured out for an existing antagonist by determining the concentration required to inhibit half of the maximum biological response of the agonist.

The  $IC_{50}$  values are often dependent on the conditions when these measurements are taken. Normally, the higher  $IC_{50}$  value of inhibitor, the lower activity of an agonist will be. The increasing of enzyme concentration results the increasing of  $IC_{50}$  value.  $IC_{50}$  value may also be affected by other factors on the type of inhibition. For example,  $IC_{50}$  value for dependent enzymes doesn't depend on the concentration of ATP. The potency of two antagonists can be compared by  $IC_{50}$  values.

In the type of competition binding assay, a single concentration of radiolabelled ligand (normally an agonist) is employed in every assay tube. A low concentration below its  $K_d$  (dissociation constant) is used for the ligand. A range of concentrations of other competing non-radioactive compounds (normally antagonists) is presented. The scale of specific binding of the radiolabelled ligand is determined. The potency of these compounds that compete for the binding of the radiolabelled ligand can be measured. In order to compare *in vitro* potencies

among different ligands from different laboratories, the  $K_i$  value is often used. Derived by Cheng-Prusoff, the  $K_i$  value is not as dependent upon experimental conditions and allows direct comparison of data obtained from different laboratories.

$$K_i = IC_{50} / \{ [L] / K_d + 1 \} \quad \text{Equation 1}$$

As shown in Equation 1, the  $K_i$  value is determined from the  $IC_{50}$  value.  $[L]$  = the concentration of free radioligand used in the assay, and  $K_d$  = the dissociation constant of the radioligand for the receptor.

$IC_{50}$  value means the concentration of competing ligand which displaces 50% of the specific binding of the radiolabelled ligand in this case.  $IC_{50}$  value can not directly indicate the affinity, but the Cheng-Prusoff equation can relate  $IC_{50}$  and affinity at least for competitive agonists and antagonists.<sup>31</sup>

### **1.13. Half maximal effective concentration ( $EC_{50}$ )**

The half maximal effective concentration ( $EC_{50}$ ) is the concentration of a drug or antibody which generates a response halfway between the baseline and maximum value. It is widely used as a measure of drug potency as well as  $IC_{50}$ . In a graded dose response curve,  $EC_{50}$  means the concentration of a compound or a drug where 50% of its maximal effect is occurred.

While in a quantitative dose response curve,  $EC_{50}$  indicates the concentration of a compound or a drug where 50% of the population give a response.

$IC_{50}$  and  $EC_{50}$  are related but different.  $IC_{50}$  is routinely used for the summary measure of the dose-response curve for competition binding assays and functional antagonist assays. While  $EC_{50}$  is the widely used one for the summary measure of agonist/stimulator assays. The concentration normally follows a sigmoidal curve and a small change in concentration results in a rapid increase. Alternately, the  $IC_{50}$  value is the point at which the effectiveness slows with increasing concentration.

#### **1.14. Amphibian alkaloids**

Plants normally produce and store nitrogenous secondary metabolites that are called alkaloids. Beyond the plant kingdom, there are a diverse array of alkaloids that come from amphibian skins. Amphibian alkaloids are the lipophilic alkaloids that have been detected in amphibian skin. Normally these skin alkaloids are toxic and are obviously used in chemical defense against predators.<sup>32</sup> The most recent review summarized about 24 classes of over 800 amphibian alkaloids in 2005.<sup>33</sup> As time goes on, more and more amphibian alkaloids will be detected and characterized. A lot of structures have been confirmed and established and there have been revisions of some previously proposed structures as spectroscopic techniques improve.

In 1978 less than 100 amphibian alkaloids had been classified.<sup>34</sup> More than 200 alkaloids were classified by 1987,<sup>35</sup> and the number went up to 300 in 1993.<sup>36</sup> A review in 1999 included

about 500 alkaloids.<sup>37</sup> The classification was named after the nominal molecular weight and an identifying letter all in bold style. Many alkaloids were extracted from amphibian skin and characterized using gas chromatographic (GC) mass spectrometry and GC-Fourier-transform infrared (FTIR) spectral analyses.

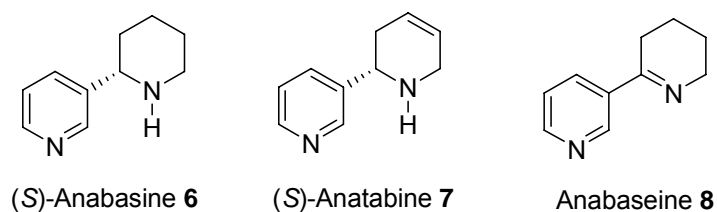
In general, evidence indicates amphibian alkaloids are sequestered from dietary sources. Except for the European fire salamander and the pseudophrynamines, amphibian alkaloids are not synthesized by the skin of amphibians themselves. Diet is the main source for amphibian to obtain and store these alkaloids. For example, ants, beetles, millipedes, and other small arthropods, even some unknown creatures are food chain for amphibians. Among the 800 amphibian skin alkaloids, only a few have been discovered in arthropods.<sup>33</sup> It is believed that beetles possess the batrachotoxins and coccinelline-like tricyclics; ants and mites possess the pumiliotoxins; ants also possess the decahydroquinolines, izidines, pyrrolidines, and piperidines; millipedes possess the spiropyrrolizidines. Histrionicotoxins, lehmizidines, and tricyclic gephyrotoxins are also very likely from ants. The source of epibatidine is unknown but generally believed not to be from a dietary source.

All the skin alkaloids compose an extraordinary chemical ecology in amphibian skin. The alkaloids derived from dietary arthropods are the secretions for defensive purpose. Dendrobatid alkaloids were specially named after the family frogs of Dendrobatidae in which those alkaloids were found. The batrachotoxins, the histrionicotoxins, the decahydroquinolines, the gephyrotoxins, the cyclopentaquinolizidines, epibatidine, the pumiliotoxins and related congeners are all belong to the class dendrobatid alkaloids.<sup>32</sup>

These alkaloids have aroused tremendous academic and pharmaceutical interest due to their structural diversity and biological activity. However, the paucity of these alkaloids from natural resources have made total synthesis the only practical method to provide sufficient material for intensive structural and biological activity studies. An ongoing project in our laboratory has developed synthetic strategies for the construction of amphibian alkaloids that exhibit pharmacological activity mediated by nicotinic receptor ion channels.<sup>38,39</sup>

### 1.15 Anabasine

(*S*)-Anabasine (**6**), (*S*)-anatabine (**7**) and anabaseine (**8**) are all pyridine alkaloids (Figure 1.6). These alkaloids can be found in cigarette tobacco and have a high potency at the nAChR.<sup>39</sup> (*S*)-Anabasine (**6**) was found in the plant of Tree Tobacco (*Nicotiana glauca*). *Nicotiana glauca* and *Nicotiana tabacum* are close relatives of the tobacco plant. Its structure is similar to nicotine (**1**), and at one time it was widely used as an insecticide. There is a trace presence of anabasine in



**Figure 1.6.** Structure of anabasine, anatabine and anabaseine

smoking tobacco, and a person's exposure to tobacco smoke can be indicated by anabasine.<sup>41</sup>

The affinity of anabasine (**6**) for mouse brain is 30-fold lower than nicotine. However the efficacy of anabasine reaches 40% that of nicotine. Anabasine is a nAChR agonist. The  $K_i$  value at the subunit of  $\alpha 4\beta 2$  is 210 nM. A depolarizing block of nerve transmission will be generated in high doses, and nicotine poisoning like symptoms will be exhibited. Accumulating doses could cause death because of asystole.<sup>42</sup> It is believed that teratogenesis in swine comes from large amounts of ingested anabasine.<sup>43</sup> Both enantiomers exhibit an intravenous  $LD_{50}$  value from 11 mg/kg to 16 mg/kg in mice.<sup>44</sup>

(*S*)-Anatabine (**7**) was found in the plant of *Nicotiana tabacum* and exhibits approximately half the potency of anabasine. Anabaseine (**8**) is a paralytic toxin discovered in the marine worm *Paranemertes peregina*.<sup>40</sup> It is a partial agonist at the  $\alpha 4\beta 2$  subtype nAChRs. The affinity of anabaseine is 20-fold weaker for the  $\alpha 4\beta 2$  subunit nAChRs and 10% efficacy of nicotine was observed. At the ganglionic  $\alpha 7$  subtype receptor, high efficacy was observed as well as high selectivity for this particular receptor subtype.<sup>40</sup>

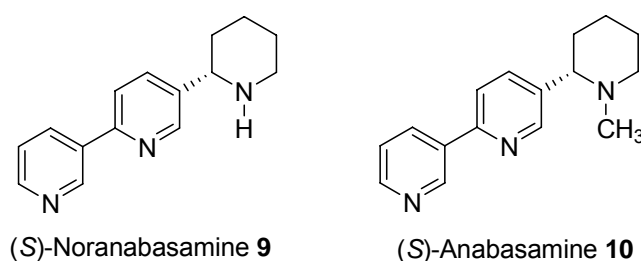
### 1.16 Noranabasamine

(*S*)-Noranabasamine (**9**) (Figure 1.7) was isolated from a Colombian poison-dart frog.<sup>45</sup> Because only trace amount of this alkaloid can be obtained from the nature, the biological activity of noranabasamine has not been investigated. The dietary sources for frogs to accumulate noranabasamine in their skin remains a mystery. Based on the structure shown in



Figure 1.7, noranabasamine is a demethylated form of another alkaloid anabasamine (**10**).

**Figure 1.7.** Structure of noranabasamine and anabasamine



(S)-Anabasamine (**10**) was found in the poisonous semi-shrub *Anabasis aphylla* of Central Asia.<sup>46</sup> Because of the scarcity of anabasamine from the nature, only limited biological studies have been conducted in the Soviet Union in 1980s.<sup>47</sup> Recent studies showed that the catalytic activity of the enzyme acetylcholinesterase can be inhibited by anabasamine.<sup>48</sup> Anti-inflammatory activity similar to indomethain was observed when anabasamine was orally applied to rats.<sup>49</sup> Similar inhibitory effects has been observed for the steroids such as hydrocortisone.<sup>50</sup> This observed effect should be the outcome from the activation of the adrenal cortex-hypothalamus-pituitary system. The piperidine alkaloid maybe play a role in strengthening the adrenergic system when it decreases the ptosis induced by reserpine. Reserpine is a compound that can blocks the dopamine-norepinephrine transformation in mice.<sup>50</sup>

Another interesting aspect for anabasamine biological activity is that when anabasamine was administered to rats, the activity of hepatic alcohol dehydrogenase was increased and

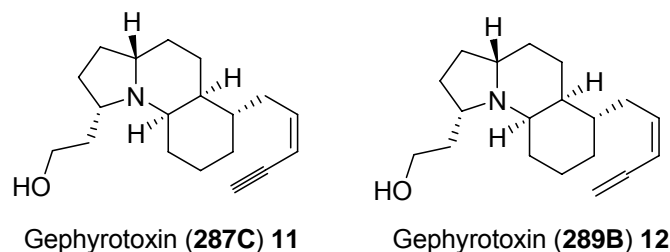
ethanol levels were decreased in the blood stream.<sup>51</sup> In addition, the adrenal regulated production of tryptophan pyrrolase was stimulated in the liver of those rats that were administered anabasamine.<sup>52</sup>

All the previous studies with (*S*)-noranabasamine (**9**) and (*S*)-anabasamine (**10**) mostly focused on the isolation and purification of this alkaloid from other related alkaloids found in amphibian skin and plants specimen. The low concentrations in plants and amphibians, the difficulty in isolation and the limited source in nature make these compounds attractive targets for synthesis. Future studies on these alkaloids and their analogues would be easier if there were more practical ways to make them available to scientists.

### 1.17. Gephyrotoxin

Gephyrotoxin was first isolated and characterized in 1977 from the skin of tropical frogs *Dendrobates histrionicus*.<sup>53</sup> The absolute configuration was based on X-ray analysis of the hydrobromide salt of gephyrotoxin (**11**). However, questions remain about the absolute configuration of gephyrotoxin isolated from frog skin.<sup>36,37,54</sup> Only two gephyrotoxins **287C** and **289B** (Figure 1.8) were discovered in nature. Gephyrotoxins are only found in very rare dendrobatid frog species of the genus *Dendrobates*. They were shown as minor alkaloids along with 19-carbon histrionicotoxins as major alkaloids in extracts. Histrionicotoxins and gephyrotoxins are always isolated from the same source. It is believed that ants and other small arthropods are most likely the origin.<sup>32</sup> Gephyrotoxin **287C** (**11**) showed relatively low toxicity

**Figure 1.8.** Structure of gephyrotoxin **287C** and **289B**



when mice were treated with a minimal toxic dose much higher than 500  $\mu$ g. Initial studies revealed this compound as muscarinic antagonist with low activity.<sup>55</sup> Recent studies have indicated it as a nontoxic noncompetitive blocker of nicotinic receptors.<sup>37</sup> Gephyrotoxin was also revealed to have an association with a more complex and interesting array of neurological activities.<sup>56</sup> The low natural abundance and unusual chemical and biological activities make this compound a attractive target for synthesis.

### 1.18. References

1. Dale, P.; Augustine, G. J.; Fitzpatrick, D.; Hall, W. C.; LaMantia, A.-S.; McNamara, J. O.; White, L. E. *Neuroscience 4th ed.* **2008**, Sinauer Associates, 122-126.
2. Siegel, G. J.; Agranoff, B. W.; Fisher, S. K.; Albers, R. W.; and Uhler, M. D. *GABA Receptor Physiology and Pharmacology* **1999**, American Society for Neurochemistry, Retrieved on 2008-10-01.
3. Itier V.; Bertrand D. *FEBS Letters* **2001**, *504*, 118-125.
4. Unwin, N. *J. Mol. Biol.* **2005**, *346*, 967-989.

5. Bate, L.; Williamson, M.; Gardiner, M. The major susceptibility locus for myoclonic epilepsy on chromosome 15q. *In Juvenile Myoclonic Epilepsy (Schmitz, B. and Sander, S., eds), Wrightson Biomedical Publications, Petersfield, UK* (in press)
6. Bate, L.; Gardiner, M. Genetics of idiopathic epilepsy. *In Techniques in the Behavioural and Neural Sciences*. (Crusio, W. E. and Gerlai, R. T., eds), **1999**, 13, 820-840, Elsevier Science, UK.
7. [http://journals.cambridge.org/fulltext\\_content/ERM/ERM1\\_19/S1462399499001349sup015.pdf](http://journals.cambridge.org/fulltext_content/ERM/ERM1_19/S1462399499001349sup015.pdf)
8. Cascio, M. *J. Biol. Chem.* **2004**, 279, 19383-19386.
9. Giniatullin, R.; Nistri, A.; Yakel, J. L. *Trends Neurosci.* **2005**, 28, 371-378.
10. Graham, A.; Court, J.A.; Martin-Ruiz, C. M.; Jaros, E.; Perry, R.; Volsen, S. G.; Bose, S.; Evans, N.; Ince, P.; Kuryatov, A.; Lindstrom, J.; Gotti, C.; Perry, E. K. *Neuroscience* **2002**, 113, 493-507.
11. Le Novère, N.; Changeux, J.-P. *J. Mol. Evol.* **1995**, 40, 155-172.
12. Rang, H. P.; Dale, M. M.; Ritter, J. M.; Moore, P. K. *Pharmacology 5th ed.* **2003**, Edinburgh: Churchill Livingstone.
13. Colquhoun, D.; Sivilotti, L. G. *Trends Neurosci.* **2004**, 27, 337-44.
14. Mishina, M.; Takai, T.; Imoto, K.; Noda, M.; Takahashi, T.; Numa, S.; Methfessel, C.; Sakmann, B. *Nature* **1986**, 321, 406-411.
15. Pitchford, S.; Day, J. W.; Gordon, A.; Mochly-Rosen, D. *J. Neurosci.* **1992**, 12, 4540-4544.
16. Huganir, R. L.; Greengard, P. *Proc. Natl. Acad. Sci. USA* **1983**, 80, 1130-1134.
17. Safran, A.; Sagi-Eisenberg, R.; Neumann, D.; Fuchs, S. *J. Biol. Chem.* **1987**, 262, 10506-10510.
18. Barrantes, F. J. *J. Mol. Biol.* **1978**, 124, 1-26.
19. Hurst, R. S.; Hajós, M.; Raggenbass, M.; Wall, T. M.; Higdon, N. R.; Lawson, J. A.; Rutherford-Root, K. L.; Berkenpas, M. B.; Hoffmann, W. E.; Piotrowski, D. W.; Groppi, V.

- E.; Allaman, G.; Ogier, R.; Bertrand, S.; Bertrand, D.; Arneric, S. P. *J. Neurosci.* **2005**, *25*, 4396-4405.
20. Wonnacott, S. *Trends Neurosci.* **1997**, *20*, 92-98.
  21. Siegmund, B.; Leitner, E.; Pfannhauser, W. *J. Agric. Food Chem.* **1999**, *47*, 3113-3120.
  22. Hoffmann, D.; Hoffmann, I. *Smoking and Tobacco Control Monograph No. 9* **2009**, 1-50.  
[http://dcccps.nci.nih.gov/tcrb/monographs/9/m9\\_3.PDF](http://dcccps.nci.nih.gov/tcrb/monographs/9/m9_3.PDF)
  23. Melsens J. *Prakt. Chem.* **1844**, *32*, 372-377.
  24. *American Heart Association and Nicotine addiction* **2009**  
<http://www.americanheart.org/presenter.jhtml?identifier=4753>
  25. Dowd, M. J. *Epibatidine - A review* <http://www.phc.vcu.edu/Feature/oldfeature/epi/>
  26. Olivo, H. F.; Hemenway, M. S. *Org. Prep. Proc. Int.* **2002**, *34*, 1-26.
  27. Carroll, F. I. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 1889-1896.
  28. Meyer, M. D. *Drug. Dev. Res.* **2006**, *67*, 355-359.
  29. Bunnelle, W. H.; Daanen, J. F.; Ryther, K. B.; Schrimpf, M. R.; Dart, M. J.; Gelain, A.; Meyer, M. D.; Frost, J. M.; Anderson, D. J.; Buckley, M.; Curzon, P.; Cao, Y. J.; Puttfarcken, P.; Searle, X.; Ji, J.; Putman, C. B.; Surowy, C.; Toma, L.; Barlocco, D. *J. Med. Chem.* **2007**, *50*, 3627-3644.
  30. Dukat, M.; Damaj, M. I.; Glassco, W.; Dumas, D.; May, E. L.; Martin, B. R.; Glennon, R. A. *Med. Chem. Res.*, **1994**, *4*, 131-139.
  31. Cheng, Y.; Prusoff, W. H. *Biochem. Pharmacol.* **1973**, *22*, 3099-3108.
  32. Daly, J. W. In *The Alkaloids*; Cordell, G. A.; Ed.; Academic Press: San Diego, 1998; Vol. 50, pp 141-169.
  33. Daly, J. W.; Spande, T. F.; Garraffo, H. M. *J. Nat. Prod.* **2005**, *68*, 1556-1575.
  34. Daly, J. W.; Brown, G. B.; Mensah-Dwumah, M.; Myers, C. W. *Toxicon* **1978**, *16*, 163-188.
  35. Daly, J. W.; Myers, C. W.; Whittaker, N. *Toxicon* **1987**, *25*, 1023-1095.

36. Daly, J. W.; Garraffo, H. M.; Spande, T. F. In *The Alkaloids*; Cordell, G. A., Ed.; Academic Press: New York, 1993; Vol. 43, pp 185-288.
37. Daly, J. W.; Garraffo, H. M.; Spande, T. F. In *Alkaloids: Chemical and Biological Perspectives*; Pelletier, S. W., Ed.; Pergamon: New York, 1999; Vol. 13, pp 1-161.
38. Banner, E. J.; Stevens, E. D.; Trudell, M. L. *Tetrahedron Lett.* **2004**, 45, 4411-4414.
39. Zhang, C.; Trudell, M. L. *J. Org. Chem.* **1996**, 61, 7189-7191.
40. Schmitt, J. D.; *Curr. Med. Chem.* **2000**, 7, 749-800.
41. Jacob, P. 3rd.; Yu, L.; Shulgin, A. T.; Benowitz, N. L. *Am. J. Public Health* **1999**, 89, 731-736.
42. Mizrachi, N.; Levy, S.; Goren, Z. *J. Forensic Sci.* **2000**, 45, 736-741.
43. *Canadian Biodiversity Information Facility*. Government of Canada. 2008-03-18. Retrieved on 2008-05-01.  
[http://www.cbif.gc.ca/pls/pp/ppack.info?p\\_psn=186&p\\_type=all&p\\_sci=sci&p\\_x=px](http://www.cbif.gc.ca/pls/pp/ppack.info?p_psn=186&p_type=all&p_sci=sci&p_x=px)
44. Stephen, L.; Wildeboer, K.; Panter, K.; Kem, W.; Gardner, D.; Molyneux, R.; Chang, C.-W.; Soti, F.; Pfister, J. USDA. 2006-02-20. Retrieved on 2008-05-01.  
[http://www.ars.usda.gov/research/publications/Publications.htm?seq\\_no\\_115=182063&pf=1](http://www.ars.usda.gov/research/publications/Publications.htm?seq_no_115=182063&pf=1)
45. Tokuyama, T.; Daly, J. W. *Tetrahedron* **1983**, 39, 41-47.
46. Sadykov, A. S. *Dokl Akad. Nauk. Uzb. SSR* **1967**, 24, 34-35.
47. Mukhamedzhanova, K. S. *et al. Dokl Akad. Nauk. Uzb. SSR* **1984**, 8, 45-47.
48. Tilyabaez, Z.; Abduvakhabov, A. A. *Chem. Nat. Comp.* **1998**, 34, 295-297.
49. Mukhamedzhanova, K. S. *Dokl Akad. Nauk. Uzb. SSR* **1983**, 20, 47-49.
50. Khnychenko, L. K. *Dokl Akad. Nauk. Uzb. SSR* **1978**, 4, 72-73.
51. Muzaev, S. *Dokl Akad. Nauk. Uzb. SSR* **1982**, 9, 47-48.
52. Muzaev, S. *Dokl Akad. Nauk. Uzb. SSR* **1977**, 7, 60-61.

53. Daly, J. W.; Witkop, B.; Tokuyama, T.; Nishikawa, T.; Karle, I. L. *Helv. Chim. Acta* **1977**, *60*, 1128-1140.
54. Daly, J. W.; Spande, T. F. In *Alkaloids: Chemical and Biological Perspectives*; Pelletier, S. W., Ed.; Wiley: New York, 1986; Vol. 4, pp 1-274.
55. Mensah-Dwumah, M.; Daly, J. W. *Toxicon* **1978**, *16*, 189-194.
56. Souccar, C.; Varanda, W. A.; Aronstam, R. S.; Daly, J. W.; Albuquerque, E. X. *Mol. Pharmacol.* **1984**, *25*, 384-400.

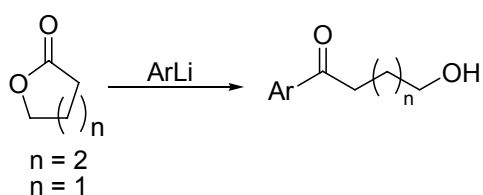
## CHAPTER 2

### Hydroxyarylketones via Ring-Opening Reactions of Lactones with Aryllithium Reagents

#### 2.1. Abstract

The regioselective ring opening of lactones ( $\delta$ -valerolactone and  $\gamma$ -butyrolactone) with aryllithium reagents is reported for the construction of a series of  $\delta$ -hydroxyarylketones and  $\gamma$ -hydroxyarylketones.

#### Scheme 2.1. Abstract scheme



#### 2.2. Introduction

$\delta$ -Hydroxyketones have been reported to be useful building blocks for the construction of



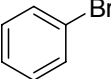
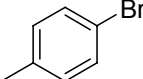
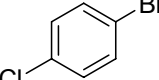
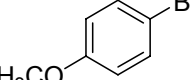
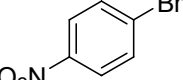
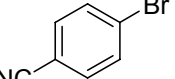
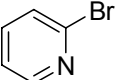
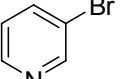
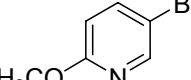
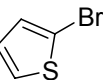
both natural and non-natural compounds.<sup>1,2</sup> The synthesis of these versatile intermediates has been achieved via a variety of methods that include the photooxidation of aryldihydropyrans,<sup>3</sup> oxidation of  $\beta$ -hydroxy-sulfones,<sup>4</sup> and nucleophilic ring opening of  $\delta$ -valerolactone (**1**).<sup>1,2,5-7</sup> Of these methods, the latter has been the most widely used, however, little attention has been given to organolithium nucleophiles.

Previous work in our laboratories led to the development of a synthetic process for the construction  $\delta$ -hydroxy-pyridinylketone derivatives via the addition of pyridinyl-lithium reagents with  $\delta$ -valerolactone.<sup>5</sup> The success of this reaction has prompted a broader study of the scope and limitations. Herein we report the reactivity of a series of aryllithium and heteroaryllithium reagents with  $\delta$ -valerolactone and  $\gamma$ -butyrolactone for the preparation of  $\delta$ -hydroxyarylketo and  $\gamma$ -hydroxyarylketo derivatives.

### 2.3. Results and discussion

As summarized Table 2.1, a series of  $\delta$ -hydroxyketones **3** were readily prepared from the reaction of **1** with a variety of aryllithium and heteroaryllithium reagents (General Method A). The organolithium reagents were initially generated in situ by treatment of the corresponding bromide with either *n*-butyllithium or *tert*-butyllithium in Et<sub>2</sub>O at -78 °C. The lactone **1** was then added to the organolithium solution. The reaction was then quenched with brine to furnish the  $\delta$ -hydroxyketones **3** in high yields (Table 2.1).

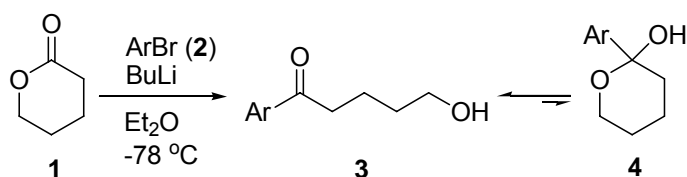
**Table 2.1.** Ring-opening of  $\delta$ -valerolactone (**1**)

entry	compound	ArBr ( <b>2</b> )	BuLi (1.2 equiv)	<b>3</b> (%yield) <sup>a</sup>
1	<b>a</b>		<i>t</i> -BuLi	79
2	<b>b</b>		<i>t</i> -BuLi	87
3	<b>c</b>		<i>t</i> -BuLi	72
4	<b>d</b>		<i>t</i> -BuLi	90
5	<b>e</b>		<i>t</i> -BuLi	-- <sup>b</sup>
6	<b>f</b>		<i>t</i> -BuLi	-- <sup>b</sup>
7	<b>g</b>		<i>n</i> -BuLi	81
8	<b>h</b>		<i>n</i> -BuLi	63
9	<b>i</b>		<i>n</i> -BuLi	98 <sup>c,d</sup>
10	<b>j</b>		<i>n</i> -BuLi	79

<sup>a</sup>General Method A, isolated yields. <sup>b</sup>Intractable mixture. <sup>c</sup>1.33 equiv of **1**. <sup>d</sup>See reference 5.

The  $\delta$ -hydroxyketones generally existed as a mixture of chain-ring tautomers (**3**:**4**, > 90:10), with the equilibrium favoring the open-chain form **3** (Scheme 2.2).<sup>8</sup>

**Scheme 2.2.** Ring-opening of  $\delta$ -valerolactone (**1**)

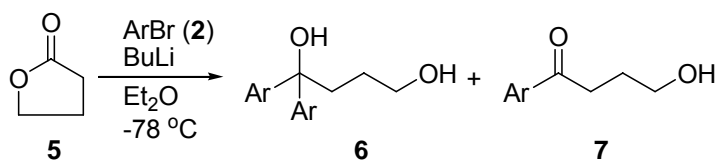


It is noteworthy that the reaction conditions did not tolerate strong electron-withdrawing groups on the aryl bromide (Table 2.1, entry 5,  $\text{NO}_2$ ; entry 6,  $\text{CN}$ ). Presumably the sensitivity of these groups to the strongly basic and nucleophilic organolithium reagents led to the formation of intractable mixtures. In addition, no diol products formed from the subsequent addition of the organolithium to the hydroxyketones were observed. These results suggest that under the low temperature conditions the lactone **1** was more reactive toward nucleophilic attack than the newly formed ketones **3**. As such it is possible to obtain the  $\delta$ -hydroxyarylketones in high yield by simple control of the reaction stoichiometry.

The ring-opening reaction was also explored with  $\gamma$ -butyrolactone (**5**) (Table 2.2). Using the reaction conditions established for **1** (General Method A), the reactivity of **5** differed significantly from that of the homologous lactone **1**. In general, the major products from the reaction of **5** with the various aryllithium reagents were the 1,1-diaryl-1,4-butanediols **6**,<sup>9</sup> while

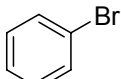
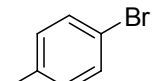
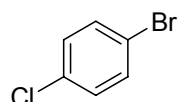
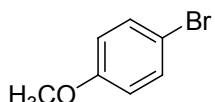
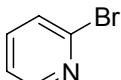
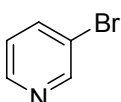
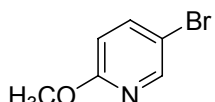
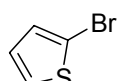
the corresponding  $\gamma$ -hydroxyketones **7** were the minor products (Scheme 2.3).

**Scheme 2.3.** Ring-opening of  $\gamma$ -butyrolactone (**5**)



The formation of diols **6a-d,j** resulted from a second addition of the organolithium reagent to the corresponding ketone that was formed initially. These results suggest that the  $\gamma$ -hydroxyketones **7a-d,j** are more reactive toward nucleophilic addition than the lactone **5**. It was interesting to discover that the pyridinyl lithium reagents **2g**, **2h** and **2i** did not readily undergo the secondary addition reaction. Only the corresponding  $\gamma$ -hydroxyketones **7** were obtained in good yield with only trace amounts ( $< 10\%$ ) of the diols **6** present. Presumably the pyridinyl moiety of the ketones **7g**, **7h** and **7i** sufficiently deactivated the carbonyl toward the addition of a second equivalent of the organolithium reagent. As a result the more reactive lactone **5** is consumed prior to the competing second addition. Even employing an excess of the pyridyllithium reagents (2.0 equiv) did not lead to increased production of the corresponding diols **6** but did lead to slightly diminished yields.

**Table 2.2.** Ring-opening of  $\gamma$ -butyrolactone (**5**)

entry	compound	ArBr ( <b>2</b> )	BuLi (1.2 equiv)	<b>6/7</b> <sup>a</sup>	<b>6/7</b> <sup>b</sup>
1	<b>a</b>		<i>t</i> -BuLi	65/16	9/71
2	<b>b</b>		<i>t</i> -BuLi	72/18	10/67
3	<b>c</b>		<i>t</i> -BuLi	68/13	11/63
4	<b>d</b>		<i>t</i> -BuLi	77/15	14/68
5	<b>g</b>		<i>n</i> -BuLi	<sup>c</sup> --/70	
6	<b>h</b>		<i>n</i> -BuLi	<sup>c</sup> --/66	
7	<b>i</b>		<i>n</i> -BuLi	<sup>c</sup> --/89	
8	<b>j</b>		<i>n</i> -BuLi	70/20	23/75

<sup>a</sup>General Method A, isolated yields. <sup>b</sup>General Method B, isolated yields. <sup>c</sup>Trace amounts of **6** (< 10%) observed by NMR.

With these results in hand, it was envisaged that the order of addition of the lactone **5** may effect the product **6/7** distribution. To this end, the order of addition was reversed by

addition of a pre-cooled (-78 °C) solution of the aryllithium reagent in Et<sub>2</sub>O to a solution of **5** in Et<sub>2</sub>O at -78 °C (General Procedure B). For the aryl bromides **2a-d,j** that were tested, all gave improved yields of the  $\gamma$ -hydroxyarylketone **7** over the corresponding diols **6** (Table 2.2). The  $\gamma$ -hydroxyarylketones **7** existed nearly exclusively in the open-chain tautomer form. Only trace amounts of the ring tautomers could be observed by nmr for **7g** and **7h**.

## 2.4. Conclusion

In summary, a series of  $\delta$ -hydroxyarylketones and  $\gamma$ -hydroxyarylketones were synthesized through our general method A /B using  $\delta$ -valerolactone and  $\gamma$ -butyrolactone. These hydroxyarylketones can be used for the construction of both natural and non-natural compounds.

## 2.5. Acknowledgment

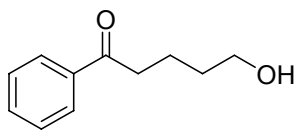
This research was funded by the National Institute on Drug Abuse (DA11528), the Louisiana Board of Regents and the University of New Orleans.

## 2.6. Experimental section

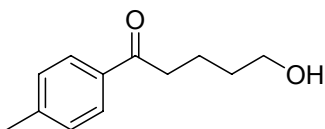
**General Method A.** Under an atmosphere of nitrogen, to a stirred solution of aryl bromide **2** (5 mmol, 1.0 equiv) in dry Et<sub>2</sub>O (80 mL) was added a solution of BuLi in hexanes (6 mmol, 1.2

equiv) drop-wise over 15 minutes at -78 °C. The solution was stirred for an additional 15 minutes at -78 °C and then a solution of lactone (**1** or **5**) (5 mmol, 1.0 equiv) in Et<sub>2</sub>O (20 mL) was added drop-wise. The reaction was allowed to warm to room temperature and stirred for 2 hours. Brine (75 mL) was added to quench the reaction and the organic layer was removed. The aqueous layer was extracted with EtOAc (2 × 50 mL) and CHCl<sub>3</sub> (2 × 50 mL). All the combined organic portions were dried over MgSO<sub>4</sub>, filtered and concentrated under reduced pressure. The residue was purified by flash column chromatography (SiO<sub>2</sub>, hexanes/EtOAc) to afford hydroxyarylketone **3** or **7**.

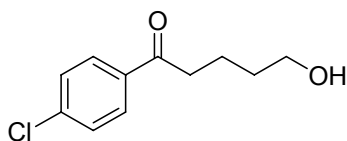
**General Method B.** Under an atmosphere of nitrogen, to a stirred solution of aryl bromide **2** (5 mmol, 1.0 equiv) in dry Et<sub>2</sub>O (40 mL) was added a solution of BuLi in hexanes (6 mmol, 1.2 equiv) drop-wise over 15 minutes at -78 °C. The solution was stirred for an additional 15 minutes at -78 °C and then added vial cannula to a solution of lactone (**5**) (5 mmol, 1.0 equiv) in Et<sub>2</sub>O (40 mL) at -78 °C. The reaction was allowed to warm to room temperature and stirred for 2 hours. Brine (75 mL) was added to quench the reaction and the organic layer was removed. The aqueous layer was extracted with EtOAc (2 × 50 mL) and CHCl<sub>3</sub> (2 × 50 mL). All the combined organic portions were dried over MgSO<sub>4</sub>, filtered and concentrated under reduced pressure. The residue was purified by flash column chromatography (SiO<sub>2</sub>, hexanes/EtOAc) to afford the  $\gamma$ -hydroxyarylketone **7**.



**5-Hydroxy-1-phenylpentan-1-one (3a).**<sup>2</sup> Compound **3a** was prepared by General Method A and obtained as a slight yellow oil (565 mg, 79% yield). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.61-1.68 (m, 2H), 1.79-1.87 (m, 2H), 1.99 (s, 1H), 3.02 (t, *J* = 7.1, 2H), 3.66 (t, *J* = 6.3, 2H), 7.45 (t, *J* = 7.9, 2H), 7.55 (t, *J* = 7.4, 1H), 7.95 (m, 2H). <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 20.4, 32.4, 38.3, 62.6, 128.3, 128.8, 133.3, 137.1, 200.7. Anal. Calcd. for C<sub>11</sub>H<sub>14</sub>O<sub>2</sub>: C, 74.13; H, 7.92. Found: C, 73.63; H, 7.98.



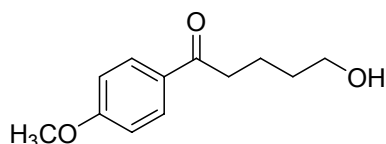
**5-Hydroxy-1-*p*-tolylpentan-1-one (3b).**<sup>1c</sup> Compound **3b** was prepared by General Method A and obtained as a white solid (837 mg, 87% yield), mp 36-38 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.56-1.75 (m, 3H), 1.84 (m, 2H), 2.38 (s, 3H), 3.00 (t, *J* = 7.1, 2H), 3.67 (dd, *J* = 11.6, 6, 2H), 7.26 (d, *J* = 7.3, 2H), 7.87 (d, *J* = 8.2, 2H). <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 20.5, 21.9, 32.5, 38.2, 62.5, 128.4, 129.5, 134.6, 144.1, 200.5. Anal. Calcd. for C<sub>12</sub>H<sub>16</sub>O<sub>2</sub>: C, 74.97; H, 8.39. Found: C, 74.75; H, 8.49.



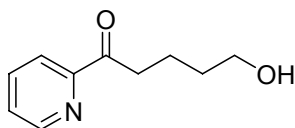
**1-(4-Chlorophenyl)-5-hydroxypentan-1-one (3c).** Compound **3c** was prepared by General



Method A and obtained as a white solid (0.77 g, 72% yield), mp 59-61 °C.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.61-1.69 (m, 3H), 1.80-1.88 (m, 2H), 3.00 (t,  $J = 7.1$ , 2H), 3.68 (dd,  $J = 11.6$ , 5.8, 2H), 7.43 (d,  $J = 8.4$ , 1H), 7.90 (d,  $J = 8.5$ , 1H).  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  20.4, 32.4, 38.3, 62.6, 129.1, 129.7, 135.4, 139.7, 199.3. Anal. Calcd. for  $\text{C}_{11}\text{H}_{13}\text{ClO}_2$ : C, 62.12; H, 6.16. Found: C, 62.17; H, 6.10.

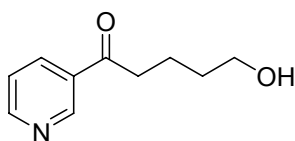


**5-Hydroxy-1-(4-methoxyphenyl)pentan-1-one (3d).**<sup>3</sup> Compound **3d** was prepared by General Method A and obtained as a colorless oil (933 mg, 90% yield).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.60-1.68 (m, 2H), 1.77-1.85 (m, 2H), 2.01 (s, 1H), 2.96 (t,  $J = 7.1$ , 2H), 3.65 (dd,  $J = 10.7$ , 5.9, 2H), 3.85 (s, 3H), 6.89-6.93 (m, 2H), 7.91-7.95 (m, 2H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  20.6, 32.5, 38.0, 55.7, 62.5, 113.9, 127.5, 130.5, 163.7, 199.3. Anal. Calcd. for  $\text{C}_{12}\text{H}_{16}\text{O}_3$ : C, 69.21; H, 7.74. Found: C, 69.63; H, 7.74.

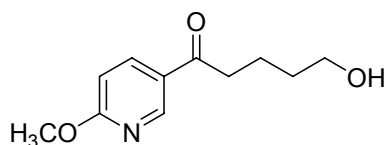


**5-Hydroxy-1-(pyridin-2-yl)pentan-1-one (3g).**<sup>10</sup> Compound **3g** was prepared by General Method A and obtained as a yellow oil (730 mg, 81% yield).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.63-1.71 (m, 2H), 1.77-1.87 (m, 2H), 1.92 (s, 1H), 3.25 (t,  $J = 7.3$ , 2H), 3.69 (t,  $J = 6.1$ , 2H), 7.46 (ddd,  $J = 7.5$ , 4.8, 1.0, 1H), 7.83 (td,  $J = 7.7$ , 1.7, 1H), 8.03 (d,  $J = 7.9$ , 1H), 8.66 (d,  $J = 4.7$ ,

1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 19.5, 20.3, 25.5, 32.4, 35.8, 37.4, 62.5, 62.6, 95.1, 120.2, 122.1, 123.6, 127.3, 137.2, 137.6, 147.7, 149.1, 153.5, 161.1, 202.1. Anal. Calcd. for C<sub>10</sub>H<sub>13</sub>NO<sub>2</sub>: C, 67.02; H, 7.31; N, 7.82. Found: C, 66.96; H, 7.46; N, 7.73.

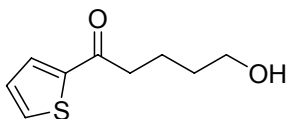


**5-Hydroxy-1-(pyridin-3-yl)pentan-1-one (3h).**<sup>1d</sup> Compound **3h** was prepared by General Method A and obtained as a yellow oil (350 mg, 63% yield). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.64-1.72 (m, 2H), 1.83-1.99 (m, 3H), 3.06 (t, *J* = 7.1, 2H), 3.70 (t, *J* = 6.1, 2H), 7.43 (dd, *J* = 8.0, 4.8, 1H), 8.25 (m, 1H), 8.77 (dd, *J* = 4.8, 0.8, 1H), 9.17 (d, *J* = 2.1, 1H). <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 20.3, 32.2, 38.7, 62.3, 124.0, 132.4, 135.8, 149.6, 153.4, 199.4. Anal. Calcd. for C<sub>10</sub>H<sub>13</sub>NO<sub>2</sub>: C, 67.02; H, 7.31; N, 7.82. Found: C, 65.74; H, 7.39; N, 7.67.

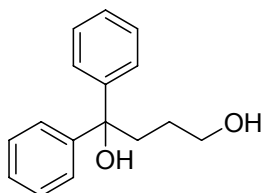


**5-Hydroxy-1-(6-methoxypyridin-3-yl)-pentan-1-one (3i).**<sup>5</sup> Compound **3i** was prepared by General Method A and obtained as a pale yellow solid (1.0 g, 98% yield), mp 42-44 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.80 (d, *J* = 2.4 Hz, 1H), 8.14 (dd, *J* = 8.7, 2.4 Hz, 1H), 6.79 (d, *J* = 8.7 Hz, 1H), 4.01 (s, 3H), 3.68 (t, *J* = 6.3 Hz, 2H), 2.97 (t, *J* = 7.1 Hz, 2H), 1.92 (brs, 1H), 1.81-1.87 (m, 2H), 1.64-1.70 (m, 2H). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ, 198.3, 166.9, 149.2, 138.4, 126.8, 111.4, 62.5, 54.3, 38.2, 32.4, 20.4. Anal. Calcd. for C<sub>11</sub>H<sub>15</sub>NO<sub>3</sub>: C, 63.14; H, 7.23; N, 6.69. Found: C,

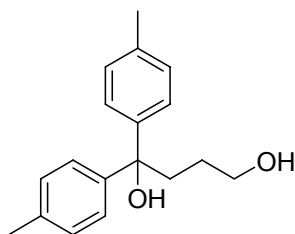
62.97; H, 7.19; N, 6.64.



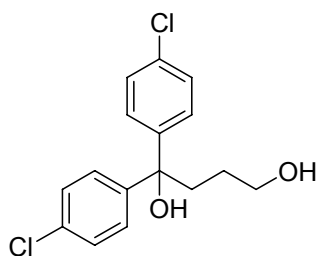
**5-Hydroxy-1-(thiophen-2-yl)pentan-1-one (3j).** Compound **3j** was prepared by General Method A and obtained as a yellow oil (726 mg, 79% yield).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.62-1.71 (m, 3H), 1.82-1.90 (m, 2H), 2.97 (t,  $J = 7.2$ , 2H), 3.67 (dd,  $J = 10.4$ , 6, 2H), 7.13 (dd,  $J = 4.9$ , 3.8, 1H), 7.63 (dd,  $J = 4.9$ , 1.1, 1H), 7.73 (dd,  $J = 3.8$ , 1.1, 1H).  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  20.9, 32.3, 39.1, 62.4, 128.4, 132.2, 133.9, 144.4, 193.8. Anal. Calcd. for  $\text{C}_9\text{H}_{12}\text{O}_2\text{S}$ : C, 58.67; H, 6.56. Found: C, 58.50; H, 6.70.



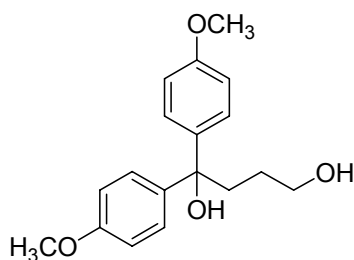
**1,1-Diphenylbutane-1,4-diol (6a).**<sup>1a</sup> Compound **6a** was prepared by General Method A and obtained as a white solid (393 mg, 65% yield), mp 104-106 °C.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.54-1.61 (m, 2H), 1.94 (s, 1H), 2.41 (t,  $J = 7.2$ , 2H), 3.24 (s, 1H), 3.63 (t,  $J = 5.6$ , 2H), 7.18-7.23 (m, 2H), 7.27-7.32 (m, 4H), 7.39-7.43 (m, 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.4, 39.2, 63.3, 78.1, 126.3, 127.0, 128.4, 147.3. Anal. Calcd. for  $\text{C}_{16}\text{H}_{18}\text{O}_2$ : C, 79.31; H, 7.49. Found: C, 79.06; H, 7.55.



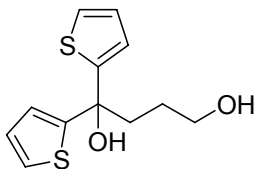
**1,1-Ditolylbutane-1,4-diol (6b).** Compound **6b** was prepared by General Method A and obtained as a white solid (486 mg, 72% yield), mp 106-108 °C.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.53-1.62 (m, 2H), 1.83 (s, 1H), 2.31 (s, 6H), 2.38 (t,  $J = 7.2$ , 2H), 2.94 (s, 1H), 3.64 (s, 2H), 7.10 (d,  $J = 8.0$ , 4H), 7.29 (d,  $J = 8.2$ , 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  21.2, 27.5, 39.2, 63.4, 78.0, 126.2, 129.1, 136.5, 144.5. Anal. Calcd. for  $\text{C}_{18}\text{H}_{22}\text{O}_2$ : C, 79.96; H, 8.20. Found: C, 79.66; H, 8.19.



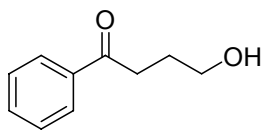
**1,1-Di(4-chlorophenyl)butane-1,4-diol (6c).**<sup>9b</sup> Compound **6c** was prepared by General Method A and obtained as a white solid (529 mg, 68% yield), mp 125-127 °C.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.50-1.60 (m, 2H), 2.11 (s, 1H), 2.37 (t,  $J = 7.1$ , 2H), 3.64 (t,  $J = 5.6$ , 2H), 3.83 (s, 1H), 7.24-7.28 (m, 4H), 7.30-7.34 (m, 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.0, 39.3, 63.2, 76.5, 127.7, 128.6, 133.0, 145.6. Anal. Calcd. for  $\text{C}_{16}\text{H}_{16}\text{Cl}_2\text{O}_2$ : C, 61.75; H, 5.18. Found: C, 61.76; H, 5.36.



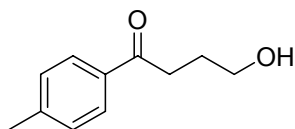
**1,1-Di(4-methoxyphenyl)butane-1,4-diol (6d).** Compound **6d** was prepared by General Method A and obtained as a colorless oil (584 mg, 77% yield).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.54-1.61 (m, 2H), 1.73 (s, 1H), 2.36 (t,  $J = 7.6$ , 2H), 2.83 (s, 1H), 3.66 (m, 2H), 3.78 (d,  $J = 6.0$ , 6H), 6.80-6.85 (m, 4H), 7.29-7.33 (m, 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.5, 39.4, 55.5, 63.4, 100.2, 113.6, 127.5, 139.8, 158.5. Anal. Calcd. for  $\text{C}_{18}\text{H}_{22}\text{O}_4$ : C, 71.50; H, 7.33. Found: C, 70.69; H, 7.26.



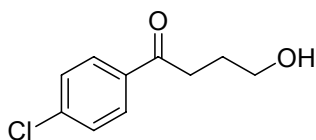
**1,1-Di(thiophen-2-yl)butane-1,4-diol (6j).** Compound **6j** was prepared by General Method A and obtained as a white solid (445 mg, 70% yield, mp 94-96 °C).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.67-1.74 (m, 2H), 1.92 (t,  $J = 4.9$ , 1H), 2.48 (t,  $J = 7.0$ , 2H), 3.70 (dd,  $J = 10.8$ , 5.7, 2H), 4.33 (s, 1H), 6.93-6.97 (m, 4H), 7.22 (dd,  $J = 4.9$ , 1.4, 2H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.5, 42.9, 63.2, 76.1, 123.9, 124.8, 126.9, 152.4. Anal. Calcd. for  $\text{C}_{12}\text{H}_{14}\text{O}_2\text{S}_2$ : C, 56.66; H, 5.55. Found: C, 56.91; H, 5.83.



**4-Hydroxy-1-phenylbutan-1-one (7a).**<sup>6</sup> Compound **7a** was prepared by General Method B and obtained as a colorless oil (582 mg, 71% yield). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.84 (t, *J* = 5.3, 1H), 1.99-2.06 (m, 2H), 3.14 (t, *J* = 6.9, 2H), 3.75 (dd, *J* = 5.9, 11.3, 2H), 7.43-7.48 (m, 2H), 7.54-7.59 (m, 1H), 7.96-7.99 (m, 2H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 27.1, 35.5, 62.6, 128.3, 128.8, 133.4, 137.1, 200.8. Anal. Calcd. for C<sub>10</sub>H<sub>12</sub>O<sub>2</sub>: C, 73.15; H, 7.37. Found: C, 72.88; H, 7.42.

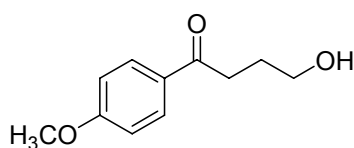


**4-Hydroxy-1-p-tolylbutan-1-one (7b).**<sup>4b</sup> Compound **7b** was prepared by General Method B and obtained as a white solid (593 mg, 67% yield), mp 42-44 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.86 (s, 1H), 1.98-2.05 (m, 2H), 2.41 (s, 3H), 3.11 (t, *J* = 6.9, 2H), 3.75 (s, 2H), 7.27 (d, *J* = 8.0, 2H), 7.88 (d, *J* = 8.2, 2H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 21.9, 27.2, 35.5, 62.6, 128.4, 129.5, 134.6, 144.2, 200.4. Anal. Calcd. for C<sub>11</sub>H<sub>14</sub>O<sub>2</sub>: C, 74.13; H, 7.92. Found: C, 73.98; H, 7.98.

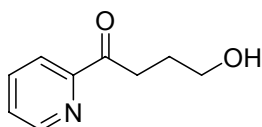


**1-(4-Chlorophenyl)-4-hydroxybutan-1-one (7c).**<sup>11</sup> Compound **7c** was prepared by General Method B and obtained as a colorless oil (628 mg, 63% yield). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ

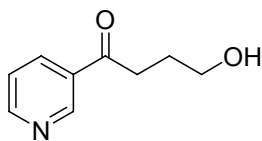
1.77 (s, 1H), 1.96-2.04 (m, 2H), 3.10 (t,  $J = 6.9$ , 2H), 3.74 (t,  $J = 6.0$ , 2H), 7.44 (d,  $J = 8.5$ , 2H), 7.92 (d,  $J = 8.5$ , 2H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.0, 35.4, 62.4, 129.1, 129.7, 135.4, 139.8, 199.5. Anal. Calcd. for  $\text{C}_{10}\text{H}_{11}\text{ClO}_2$ : C, 60.46; H, 5.58. Found: C, 60.66; H, 5.81.



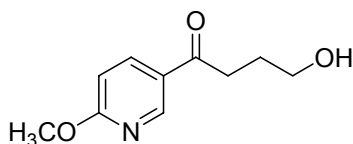
**4-Hydroxy-1-(4-methoxyphenyl)butan-1-one (7d).**<sup>12</sup> Compound **7d** was prepared by General Method B and obtained as a white solid (657 mg, 68% yield), mp 46-48 °C.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.93 (t,  $J = 5.2$ , 1H), 1.97-2.04 (m, 2H), 3.08 (t,  $J = 6.9$ , 2H), 3.74 (q,  $J = 5.7$ , 2H), 3.87 (s, 3H), 6.93 (d,  $J = 9.0$ , 2H), 7.96 (d,  $J = 9.0$ , 2H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.3, 35.2, 55.7, 62.6, 114.0, 127.5, 130.6, 163.7, 199.4. Anal. Calcd. for  $\text{C}_{11}\text{H}_{14}\text{O}_3$ : C, 68.02; H, 7.27. Found: C, 67.97; H, 7.37.



**4-Hydroxy-1-(pyridin-2-yl)butan-1-one (7g).**<sup>7</sup> Compound **7g** was prepared by General Method A and obtained as a yellow oil (577 mg, 70% yield).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.98-2.05 (m, 2H), 2.57 (t,  $J = 5.7$ , 1H), 3.31 (t,  $J = 7.0$ , 2H), 3.70 (q,  $J = 6.0$ , 2H), 7.48 (ddd,  $J = 7.6$ , 4.8, 1.2, 1H), 7.84 (td,  $J = 7.7$ , 1.7, 1H), 8.03 (d,  $J = 7.9$ , 1H), 8.66 (d,  $J = 4.7$ , 1H).  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  27.8, 34.5, 62.2, 122.1, 127.5, 137.3, 149.1, 153.6, 202.7. Anal. Calcd. for  $\text{C}_9\text{H}_{11}\text{NO}_2$ : C, 65.44; H, 6.71; N, 8.48. Found: C, 64.76; H, 7.04; N, 8.29.

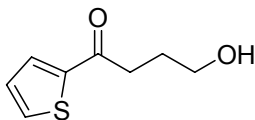


**4-Hydroxy-1-(pyridin-3-yl)butan-1-one (7h).**<sup>1d</sup> Compound **7h** was prepared by General Method A and obtained as a light yellow solid (545 mg, 66% yield), mp 36-38 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.98-2.05 (m, 2H), 2.40 (t, *J* = 5.1, 1H), 3.13 (t, *J* = 7.0, 2H), 3.74 (dd, *J* = 11.3, 5.8, 2H), 7.40 (dd, *J* = 8.0, 4.8, 1H), 8.23 (dt, *J* = 8.0, 2.0, 1H), 8.74 (dd, *J* = 4.8, 1.7, 1H), 9.16 (d, *J* = 2.2, 1H). <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 26.8, 35.6, 61.7, 124.0, 132.4, 135.8, 149.6, 153.4, 199.4. Anal. Calcd. for C<sub>9</sub>H<sub>11</sub>NO<sub>2</sub>: C, 65.44; H, 6.71; N, 8.48. Found: C, 65.85; H, 6.82; N, 8.45.



**4-Hydroxy-1-(6-methoxypyridin-3-yl)butan-1-one (7i).** Compound **7i** was prepared by General Method A and obtained as a pale yellow solid (0.87 g, 89% yield), mp 35-37 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.82 (d, *J* = 2.4 Hz, 1H), 8.15 (dd, *J* = 8.7, 2.5 Hz, 1H), 6.79 (d, *J* = 8.7 Hz, 1H), 4.01 (s, 3H), 3.75 (d, *J* = 4.4 Hz, 2H), 3.08 (t, *J* = 6.9 Hz, 2H), 2.14 (s, 1H), 1.98-2.05 (m, 2H). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ 198.5, 167.0, 149.3, 138.4, 126.8, 111.4, 62.3, 54.3, 35.3, 27.0. Anal. Calcd. for C<sub>10</sub>H<sub>13</sub>NO<sub>3</sub>: C, 61.53; H, 6.71; N, 7.18. Found: C, 61.70; H, 6.82; N, 7.03.





**4-Hydroxy-1-(thiophen-2-yl)butan-1-one (7j).**<sup>13</sup> Compound **7a** was prepared by General Method B and obtained as a colorless oil (641 mg, 75% yield). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.87 (t, *J* = 5.3, 1H), 1.99-2.06 (m, 2H), 3.08 (t, *J* = 7.0, 2H), 3.75 (q, *J* = 5.8, 2H), 7.12-7.15 (m, 1H), 7.64 (dd, *J* = 4.9, 1.0, 1H), 7.75 (dd, *J* = 3.8, 1.0, 1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 27.4, 36.2, 62.4, 128.4, 132.3, 133.9, 144.4, 193.7. Anal. Calcd. for C<sub>8</sub>H<sub>10</sub>O<sub>2</sub>S: C, 56.44; H, 5.92. Found: C, 56.05; H, 6.10.

## 2.7. References

1. (a) Descotes, G.; Soula, J.-C. *Bull. Soc. Chim. Fr.* **1964**, *5*, 2636-2639. (b) Crich, D.; Huang, X.; Newcomb, M. *Org. Lett.* **1999**, *1*, 225-227. (c) Bailey, W. F.; Khanolkar, A. D. *Tetrahedron* **1991**, *47*, 7727-7738. (d) Ohkawa, S.; Terao, S.; Terashita, Z.; Shibouta, Y.; Nishikawa, K. *J. Med. Chem.* **1991**, *34*, 267-276.
2. Rosenblum, S. B.; Bihovsky, R. *J. Am. Chem. Soc.* **1990**, *112*, 2746-2748.
3. (a) Atkinson, R. S. *Chem. Commun.* **1970**, 177. (b) Atkinson, R. S. *J. Chem. Soc.* **1971**, 784-788.
4. (a) Fuji, K.; Node, M.; Usami, Y.; Kiryu, Y. *J. Chem. Soc. Chem Commun.* **1987**, 449-450. (b) Fuji, K.; Usami, Y.; Kiryu, Y.; Node, M. *Synthesis* **1992**, 852-858.
5. Miao, L.; DiMaggio, S. C.; Shu, H.; Trudell, M. L. *Org. Lett.* **2009**, *11*, 1579-1582.
6. Yang, S.-B.; Gan, F.-F.; Chen, G.-J.; Xu, P.-F. *Synlett* **2008**, *16*, 2532-2534.
7. Gómez, I.; Alonso, E.; Ramón, D. J.; Yus, M. *Tetrahedron* **2000**, *56*, 4043-4052.

8. Whiting, J. E.; Edward, J. T. *Can. J. Chem.* **1971**, *49*, 3799-3806.
9. (a) Vozza, J. F. *J. Org. Chem.* **1959**, *24*, 720-722. (b) Umio, S.; Ueda, I.; Nojima, H. *J. Med. Chem.* **1972**, *15*, 855-856.
10. Shimizu, J.; Tsurki, T.; Yamagishi, Y.; Inchino, T. Japanese Patent JP06157459; *Chem. Abstr.* **1994**, *121*:179498.
11. Huang, N.; Xu, L. *Youji Huaxue* **1989**, *9*, 436-437. *Chem. Abstr.* **1989**, *113*:5301.
12. Ramadas, S. R.; Sukumaran, K. B. *Ind. J. Chem.* **1970**, *8*, 470-471.
13. Acheson, R. M.; Cooper, M. W. *J. Chem. Soc. Perkin I*, **1980**, 1185-1193.

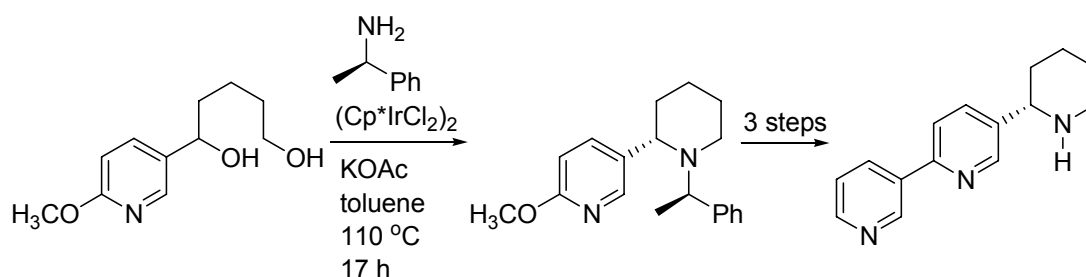
## CHAPTER 3

### Enantioselective Syntheses of Both Enantiomers of Noranabasamine

#### 3.1. Abstract

Both the *R* and *S* enantiomers of the amphibian alkaloid noranabasamine were prepared in > 30% overall yield with 80 %ee and 86 %ee, respectively. An enantioselective iridium-catalyzed *N*-heterocyclization reaction with either (*R*)- or (*S*)-1-phenylethylamine and 1-(5-methoxypyridin-3-yl)-1,5-pentanediol was employed to generate the 2-(pyridin-3-yl)-piperidine ring system in 69-72 % yield.

**Scheme 3.1.** Noranabasamine synthesis



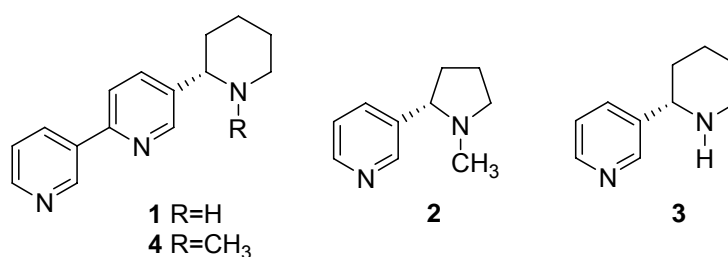
### 3.2. Introduction

The pharmacology of amphibian alkaloids has generated significant interest in these molecules over the past decade. Many of these compounds have aided in the elucidation of biological mechanisms and the development of lead compounds for the treatment of a wide variety of pathologies mediated by nicotinic acetylcholine receptors (nAChRs) and corresponding ion channels.<sup>1</sup> However, the paucity of useful quantities of isolated amphibian alkaloids has led to a flurry of synthetic activity to make these compounds available for biological study. While many of the amphibian alkaloids possess unique chemical structures,<sup>1</sup> the similarity between noranabasamine (**1**) isolated from the columbian poison dart frog *Phyllobates terribilis*,<sup>2</sup> and plant alkaloids isolated from the tobacco species *Nicotian tabacum* [e.g., nicotine (**2**), anabasine (**3**)],<sup>3</sup> as well as the central asian shrub *Anabasis aphylla* [anabasamine (**4**)],<sup>4</sup> is noteworthy. The plant-derived piperidine alkaloids **2** and **3** are widely known to elicit their pharmacological effects via nAChRs.<sup>5</sup> Anabasamine (**4**) has been much less studied but has been reported to inhibit acetylcholine esterase and exhibit anti-inflammatory activity.<sup>6</sup>

Our interests in the development of new pharmacotherapies for nAChR mediated disorders and disease states<sup>7</sup> prompted an investigation into the synthesis of the enantiomers of noranabasamine (**1**). It was our aim to develop an efficient synthesis of **1**, that would provide sufficient quantities for biological evaluation. In addition, it was envisaged that the preparation of both antipodes of **1** would aid in the confirmation of the absolute configuration of the natural product which has yet to be unequivocally established.<sup>4</sup> Herein we describe the first

enantioselective syntheses of both enantiomeric forms of noranabasamine.

**Figure 3.1.** Noranabasamine (**1**) and related plant alkaloids

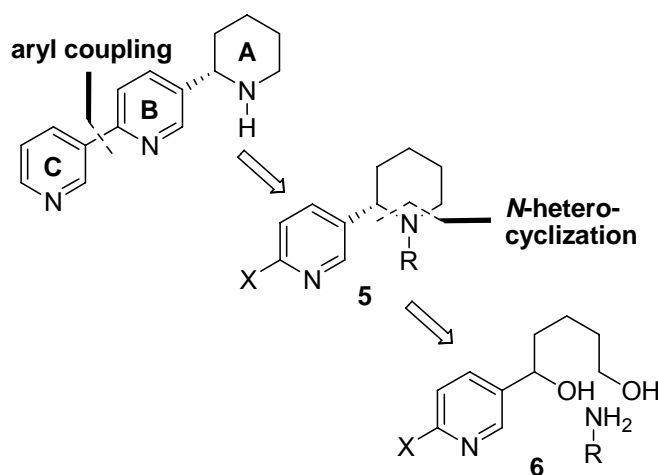


### 3.3. Results and discussion

Our retrosynthetic analysis illustrated in Scheme 3.2 focused on the disconnection of the terminal pyridyl group (ring C) to afford a 2-substituted piperidine fragment **5** as our initial target. There are a variety of methods for the enantioselective construction of 2-substituted piperidines,<sup>8</sup> but the iridium-complex-catalyzed *N*-heterocyclization of primary amines with diols recently reported by Yamaguchi and co-workers seemed to be exceptionally well suited for the construction of the AB-ring system of **5** and has not been explored for the preparation of natural products.<sup>9</sup> A diastereoselective *N*-heterocyclization with the appropriate chiral primary amine was envisaged for introduction of the single stereogenic carbon atom of the noranabasamine skeleton. The approach not only was deemed straightforward but also offered the flexibility for the preparation of various derivatives and analogues if structure-activity studies were warranted.

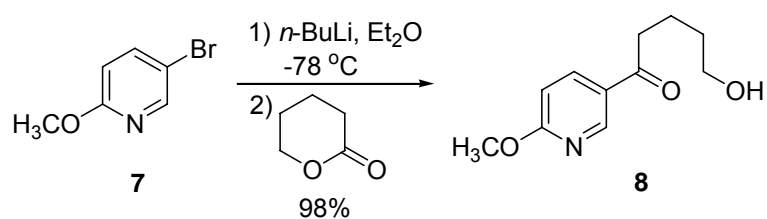
in the future.

**Scheme 3.2.** Retrosynthetic analysis of noranabasamine (**1**)



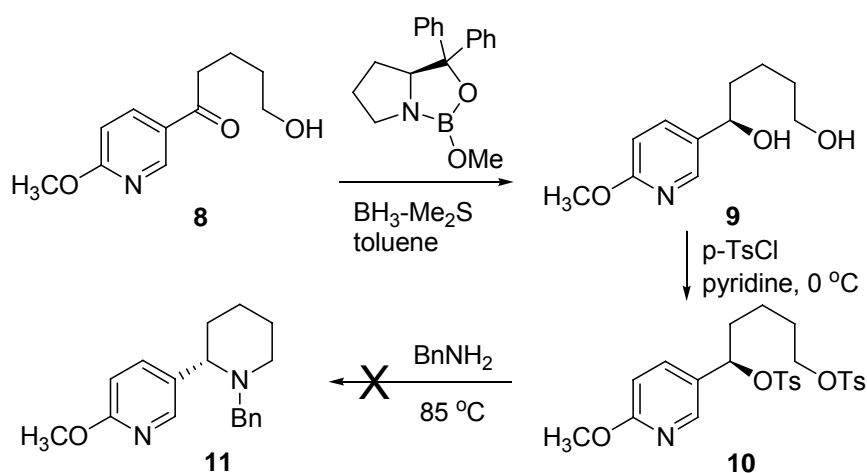
As illustrated in Scheme 3.3, the ketone **8** was prepared from 5-bromo-2-methoxypyridine (**7**). Treatment of **7** with *n*-butyllithium followed by addition of  $\delta$ -valerolactone to the lithiated pyridine solution afforded the ketone **8** in 98% yield. The

**Scheme 3.3.** Preparation of ketone **8**



ring-opening reaction proceeded regioselectively to give **8** without further nucleophilic addition to the carbonyl.

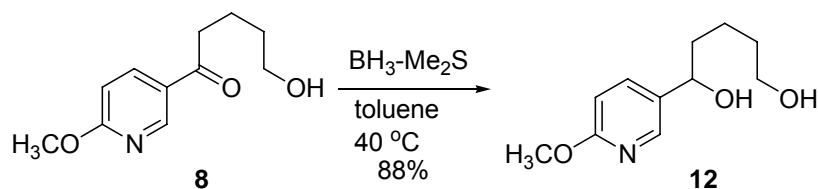
**Scheme 3.4.** Initial proposed *N*-heterocyclization



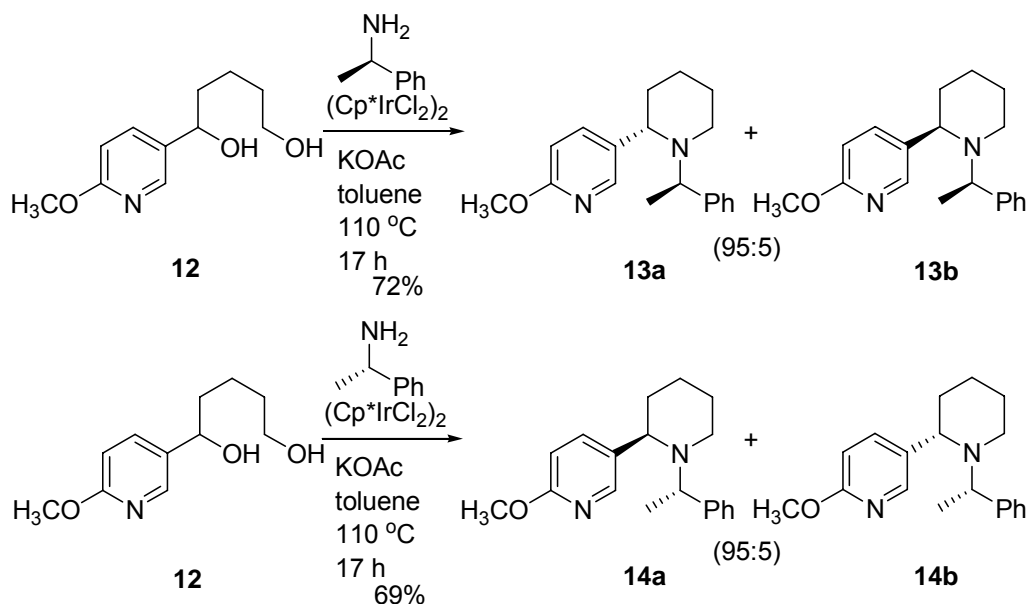
Our initial approach was to generate a secondary chiral alcohol at the ketone position of **8**.<sup>10</sup> As shown in Scheme 3.4, ketone **8** was reduced to the corresponding diol **9** using (*S*)-(-)- $\alpha,\alpha$ -diphenyl-2-pyrrolidinemethanol as a chiral catalyst. The diol **9** was converted to ditosylate **10**.<sup>11,12</sup> We envisaged that the ditosylate **10** would undergo the *N*-heterocyclization with the treatment benzylamine.<sup>12</sup> However after several attempts, none of the desired product **11** was obtained through this route.

The carbonyl group of **8** was then reduced to the hydroxyl moiety with  $\text{BH}_3\cdot\text{SMe}_2$  to furnish the racemic diol **12** in 88% yield (Scheme 3.5). With the diol **12** in hands, our attention focused on the enantioselective construction of the piperidine ring using *N*-heterocyclization

**Scheme 3.5.** Preparation of racemic diol **12**



**Scheme 3.6.** Diastereoselective *N*-Heterocyclization

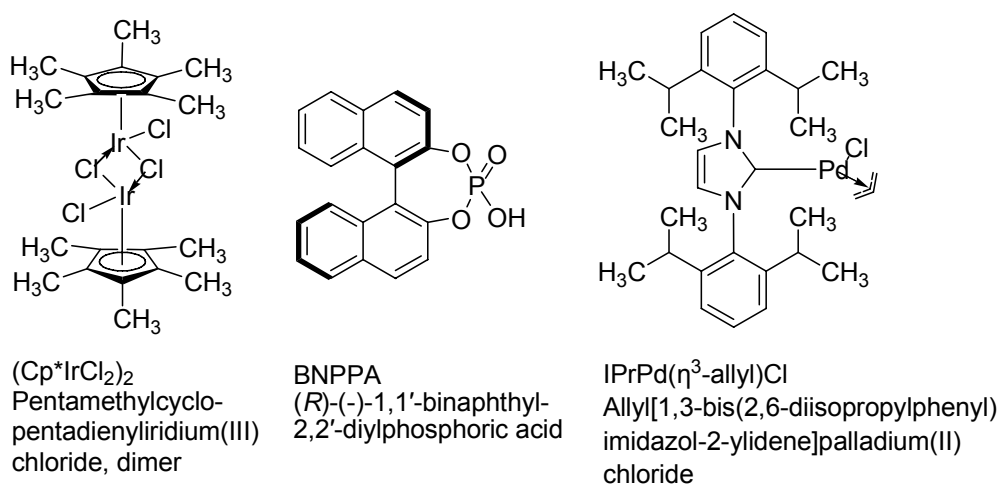


chemistry (Scheme 3.6). The diol **12** was heated at  $110\text{ }^\circ\text{C}$  in toluene with *(R)*-1-phenylethylamine<sup>13</sup> in the presence of a catalytic amount (1.5 mol%) of  $(\text{Cp}^*\text{IrCl}_2)_2$  (Figure 3.2) in a sealed reaction tube. The *N*-heterocyclization then proceeded diastereoselectively to provide the 2-substituted piperidine **13ab** in 72% yield (**13a**:**13b**, dr, 95:5). The diastereoisomers were easily separated by column chromatography. The 2-substituted



piperidine **14ab** was prepared in similar fashion from diol **12** using (*S*)-1-phenylethylamine (Scheme 3.6). The piperidine **14ab** was obtained in 69% yield with a diastereomeric ratio of **14a:14b** equal to 95:5. On the basis of the work of Yamaguchi and co-workers, we initially assigned the stereochemistry at C2 of **13a** as possessing an *S*-configuration and **14a** as having an *R*-configuration.

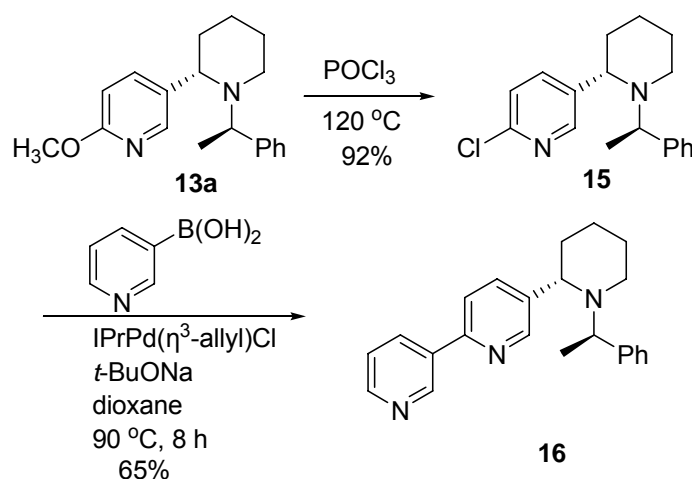
**Figure 3.2.** Structure of  $(\text{Cp}^*\text{IrCl}_2)_2$ , BNPPA and  $\text{IPrPd}(\eta^3\text{-allyl})\text{Cl}$



Presumably *N*-heterocyclization proceeds through the formation of various imine and enamine intermediates.<sup>9</sup> To ensure ourselves that epimerization/racemization of the two stereogenic centers had not occurred during the ringenerating process, it would be necessary to establish the enantiomeric integrity of the piperidine ring. Crooks and co-workers recently reported a procedure for the determination of the enantiopurity of anabasine and related alkaloids using NMR spectroscopy and the chiral shift reagent 1,1'-binaphthyl-2,2'-diylphosphoric acid

(BNPPA) (Figure 3.2).<sup>14</sup> With this technique in mind, it was envisaged that the issue of the enantiopurity of the piperidine ring system would be more easily resolved with **1**, due to the similar secondary amine structure of noranabasamine to the anabasine.

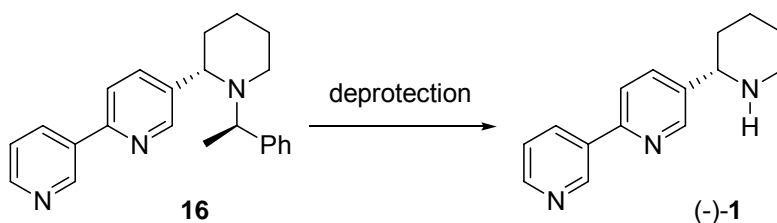
**Scheme 3.7.** Synthesis of tricyclic compound **16**



With the piperidine ring system **13a** in hand, our attention was directed toward completing the synthesis of noranabasamine (**1**). On the basis of the initial stereochemical assignment of **1**,<sup>2</sup> it was assumed that the *N*-heterocyclization product **13a** possessed the correct stereochemistry at C2. However, the conversion of **13a** into **1** would require the manipulation of the methoxy group into a more suitable moiety to facilitate an aryl cross-coupling reaction. To this end, treatment of **13a** with  $\text{POCl}_3$  furnished the 5-chloropyridin-3-yl derivative **15** in 92% yield (Scheme 3.7). The Suzuki-Miyaura coupling of **15** with 3-pyridineboronic acid could be achieved using several different types of palladium/ligand systems.<sup>15</sup> Initially we employed the

catalytic system reported by Nolan and co-workers for the coupling sequence using allyl[1,3-bis(2,6-diisopropylphenyl)imidazol-2-ylidene]palladium(II) chloride [IPrPd( $\eta^3$ -allyl)Cl] (Figure 3.2) as a catalyst.<sup>16</sup> This furnished the tricyclic compound **16** in 65% isolated yield.

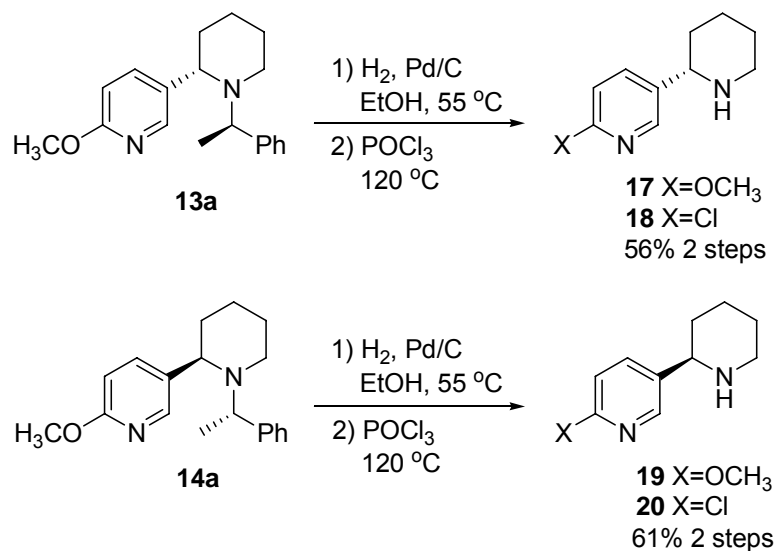
**Scheme 3.8.** Attempt to deprotect the tricyclic compound **16**



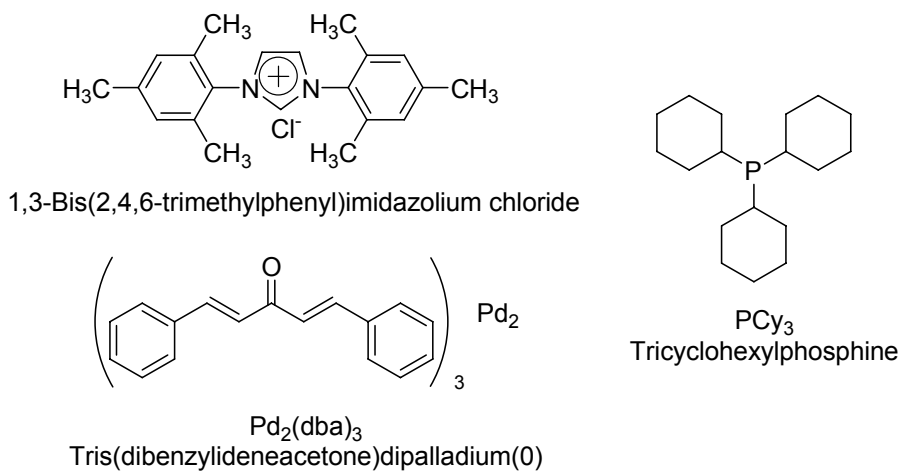
As shown in Scheme 3.8, various conditions were attempted to deprotect the the tricyclic compound **16**. They were: I: 10% (or 20%) Pd/C [or Pd(OH)<sub>2</sub>/C], H<sub>2</sub> (1 atm or 50 PSI), 6N HCl (0 equiv or 2.0 equiv), EtOH, RT or 65 °C, 24 hr or 48 hr.<sup>9,17</sup> II: TMSCH<sub>2</sub>CH<sub>2</sub>OCOCl, THF; Bu<sub>4</sub>N<sup>+</sup>F<sup>-</sup>.<sup>18,19</sup> III: 10% Pd/C, NH<sub>4</sub><sup>+</sup>HCOO<sup>-</sup>, Et<sub>2</sub>O, RT.<sup>20</sup> IV: Na/NH<sub>3</sub>.<sup>21</sup> Unfortunately, despite numerous attempts to remove the *N*-phenylethyl auxiliary group, none were successful. The increased basicity of the molecule and the additional steric hindrance around the nitrogen atom completely shut down the hydrogenolysis of the *N*-1-phenylethyl group. High pressure, high temperature, and extended reaction times either resulted in recovery of unreacted starting material or decomposition and formation of intractable mixtures.

To avoid the problematic hydrogenolysis of **16**, an alternative sequence of reactions was devised to prepare **1** (Scheme 3.9). The methoxy derivative **13a** was subjected to hydrogenolysis

**Scheme 3.9.** Revised procedure of hydrogenation



**Figure 3.3.** Structure of Pd catalysts and ligands

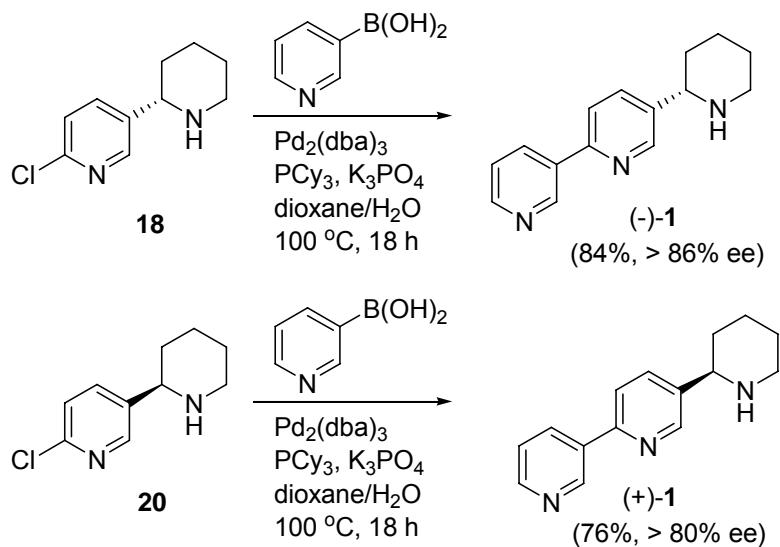


conditions to furnish **17** and concomitant treatment with POCl<sub>3</sub> provided the chloro analogue **18** in 56% yield over the two-step procedure. This sequence was also applied to **14a** and furnished

the corresponding **20** in 61% yield.

For the final step, several conditions were attempted to optimize the Suzuki-Miyaura coupling reactions. They were: I: IPrPd( $\eta^3$ -allyl)Cl (Figure 3.2), t-BuONa, dioxane (Used for the synthesis of **16**).<sup>16</sup> II: Pd(OAc)<sub>2</sub>, K<sub>2</sub>CO<sub>3</sub>, DMF/H<sub>2</sub>O.<sup>22</sup> III: Pd<sub>2</sub>(dba)<sub>3</sub> (Figure 3.3), 1,3-bis(2,4,6-trimethylphenyl)imidazolium chloride (Figure 3.3), Cs<sub>2</sub>CO<sub>3</sub>, dioxane.<sup>23</sup> All the above conditions gave low yields and difficulties of chromatography separations.

**Scheme 3.10.** Synthesis of noranabasamine

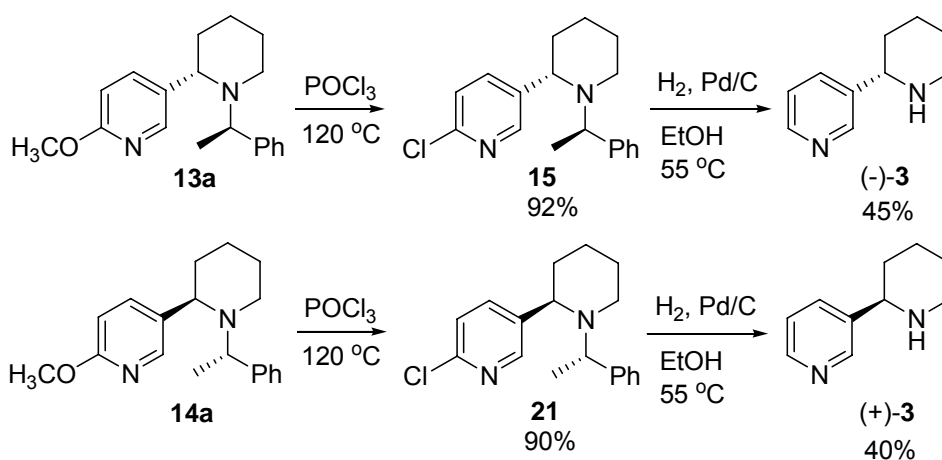


We utilized Fu and co-workers' report using Pd<sub>2</sub>(dba)<sub>3</sub> (Figure 3.3) as coupling catalyst and PCy<sub>3</sub> (Figure 3.3) as ligand (Scheme 3.10).<sup>24</sup> These conditions for the coupling of 3-pyridineboronic acid with **18** were found to be superior to other methods because of the ease of the workup and purification steps. This afforded the (*S*)-noranabasamine **(-)-1** in 84% yield,

$[\alpha]_D^{25} = -32.9$  ( $c$  0.33,  $\text{CH}_3\text{OH}$ ). The NMR data of (-)-**1** was identical to the reported data of the isolated material, and the optical rotation was also levorotatory.<sup>25</sup> The synthesis of the (*R*)-noranabasamine (+)-**1**  $\{[\alpha]_D^{25} +34.6$  ( $c$  0.5,  $\text{CH}_3\text{OH}$ ) from **20** provided additional support of a 2*S*-configuration of the natural antipode of noranabasamine.

At this point we sought to establish the enantiopurity of the piperidine ring systems. As expected use of the chiral shift reagent BNPPA (Figure 3.2) afforded baseline resolution of the proton signals for H2, H6, and H2'' of both enantiomers of **1**.<sup>26</sup> From the NMR study it was quite clear that the enantiopurity of (-)-**1** was greater than 86% ee, and that of (+)-**1** was greater than 80% ee. From these results it can be inferred that the diastereoselective *N*-heterocyclization reactions that furnished **13a** and **14a** (Scheme 3.6) are highly enantioselective (>80% ee) and consistent with previous studies.<sup>9</sup>

### Scheme 3.11. Synthesis of anabasine



Altering the sequence of chlorination and hydrogenation on **13a** afforded another natural product, (*S*)-anabasine (-)-**3** (Scheme 3.11). This afforded the (*S*)-anabasine (-)-**3** in 41% yield over two steps,  $[\alpha]^{25}_{\text{D}} = -36.5$  (*c* 0.5, CH<sub>3</sub>OH). The synthesis of the (*R*)-anabasine (+)-**3**  $\{[\alpha]^{25}_{\text{D}} +36.7$  (*c* 0.6, CH<sub>3</sub>OH) from **21** was performed as the manner.

### 3.4. Conclusion

In summary, we have shown that the iridium-catalyzed *N*-heterocyclization reaction is a facile method for the efficient and enantioselective construction of 2-(pyridin-3-yl)-piperidine alkaloids. This reaction was a key step in the first total synthesis of both enantiomers of the amphibian alkaloid noranabasamine (**1**) in greater than 30% overall yield and has allowed us to establish the absolute configuration of the natural product as levorotatory. Additional studies with regard to the scope and limitations of this reaction system are ongoing and will be reported in due course. The biological evaluation of both enantiomeric forms of noranabasamine is currently under investigation and will be reported elsewhere.

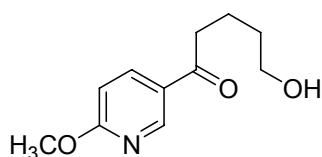
### 3.5. Acknowledgment

This research was funded by the National Institute on Drug Abuse (DA11528) and the University of New Orleans.

### 3.6. Experimental section

#### General Experimental Methods

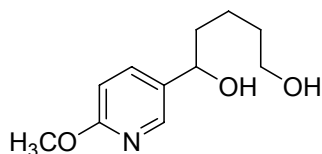
All chemicals were purchased from Aldrich Chemical Company and used as received unless otherwise noted. Anhydrous toluene was purchased from Mallinckrodt Baker, Inc. Proton and carbon NMR were recorded on a Varian-400 MHz nuclear magnetic resonance spectrometer at ambient temperature in deuterated chloroform ( $\text{CDCl}_3$ ), or methanol ( $\text{CD}_3\text{OD}$ ) from Cambridge Isotope Laboratories, Inc.  $^1\text{H}$  NMR chemical shifts are reported as  $\delta$  values (ppm) relative to tetramethylsilane.  $^{13}\text{C}$  NMR chemical shifts are reported as  $\delta$  values (ppm) relative to chloroform-*d* (77.0 ppm). For chiral shift NMR, more accurate integration was obtained using MestReNova® software. Optical rotations were measured on Autopol III autopolarimeter at the sodium D line (2 mL sample cell). Melting points (mp) were measured with an Electrothermal R Mel-Temp apparatus and are uncorrected.



**4-Hydroxybutyl-6-methoxypyridin-3-yl ketone (8).** Under an atmosphere of nitrogen, to a stirred solution of 5-bromo-2-methoxypyridine (7, 2.50 g, 1.72 mL, 13.3 mmol, 1.0 equiv) in dry  $\text{Et}_2\text{O}$  (80 mL) was added a solution of *n*-BuLi in hexanes (1.6M, 9.14 mL, 14.6 mmol, 1.1 equiv) dropwise over 15 minutes at  $-78^\circ\text{C}$ . The solution was stirred for an additional 15 minutes at  $-78^\circ\text{C}$  and then a solution of  $\delta$ -valerolactone (1.77 g, 17.7 mmol, 1.3 equiv) in  $\text{Et}_2\text{O}$  (20 mL) was

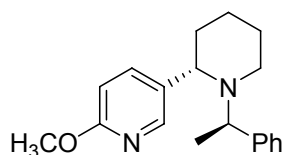


added dropwise. The reaction was stirred at room temperature for 2 hours. Brine (75 mL) was added to quench the reaction and the organic layer was removed. The aqueous layer was extracted with EtOAc (2 × 50 mL) and CHCl<sub>3</sub> (2 × 50 mL). All the combined organic portions were dried over MgSO<sub>4</sub>, filtered and concentrated using a rotary evaporator under reduced pressure. The residue was purified by flash column chromatography (SiO<sub>2</sub>, 20:80 hexanes/EtOAc) to afford 2.75 g (98% yield) of **8** as a pale yellow solid, mp 42-44 °C. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.64-1.70 (m, 2H), 1.81-1.87 (m, 2H), 1.92 (b, 1H), 2.97 (t, *J* = 7.1, 2H), 3.68 (t, *J* = 6.3, 2H), 4.01 (s, 3H), 6.79 (d, *J* = 8.7, 1H), 8.14 (dd, *J* = 8.7, 2.4, 1H), 8.80 (d, *J* = 2.4, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 20.4, 32.4, 38.2, 54.3, 62.5, 111.4, 126.8, 138.4, 149.2, 166.9, 198.3. Anal. Calcd. for C<sub>11</sub>H<sub>15</sub>NO<sub>3</sub>: C, 63.14; H, 7.23; N, 6.69. Found: C, 62.97; H, 7.19; N, 6.64.



**1-(6-Methoxypyridin-3-yl)pentane-1,5-diol (12).** Under an atmosphere of nitrogen, to a stirred solution of ketone **8** (1.0 g, 4.78 mmol, 1.0 equiv) in dry toluene (30 mL) was added borane dimethyl sulfide complex (10M, 0.956 mL, 9.56 mmol, 2.0 equiv) dropwise at 40 °C. The mixture was stirred at 40 °C for 2 hours, quenched with methanol (20 mL) in an ice bath and concentrated under reduced pressure to remove the solvent. The residue was dissolved in brine (25 mL) and extracted with EtOAc (2 × 25 mL) and CHCl<sub>3</sub> (2 × 25 mL). All the organic portions were combined, dried over MgSO<sub>4</sub>, filtered and concentrated using a rotary evaporator under

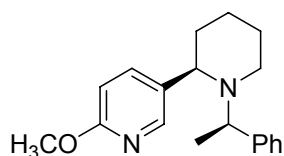
reduced pressure. The residue was purified by flash column chromatography (SiO<sub>2</sub>, EtOAc) to afford 0.88 g (88% yield) of **12** as a colorless oil. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.21-1.87 (m, 6H), 2.33 (b, 2H), 3.62 (t, *J* = 6.3, 2H), 3.92 (s, 3H), 4.65 (t, *J* = 6.6, 1H), 6.74 (d, *J* = 8.6, 1H), 7.61 (dd, *J* = 8.6, 2.3, 1H), 8.07 (d, *J* = 2.1, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 22.1, 32.3, 38.5, 53.8, 62.4, 71.6, 111.0, 133.4, 137.2, 144.5, 163.8. Anal. Calcd. for C<sub>11</sub>H<sub>17</sub>NO<sub>3</sub>: C, 62.54; H, 8.11; N, 6.63. Found: C, 61.56; H, 8.19; N, 6.52.



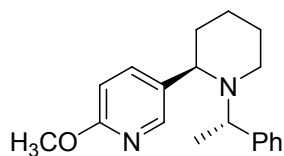
#### General Procedure A: *N*-Heterocyclization.

**(2*S*)-*N*-[(*R*)-1-Phenylethyl]-2-(5-methoxypyridin-3-yl)-piperidine (**13a**).** The diol **12** (211 mg, 1.0 mmol, 1.0 equiv.), [Cp\*IrCl<sub>2</sub>]<sub>2</sub> (3.0% Ir, 12 mg, 0.015 mmol, 0.015 equiv), KOAc (6.0%, 5.88 mg, 0.06 mmol, 0.06 equiv), (*R*)-1-phenylethylamine (99% ee) (121 mg, 0.127 mL, 1.0 mmol, 1.0 equiv) and toluene (1.0 mL) were placed under an atmosphere of argon in a high-pressure tube. The tube was sealed, and the mixture was stirred at the temperature of 110 °C for 17 hours. The resulting mixture was purified by preparative TLC (SiO<sub>2</sub>, 12:88 hexanes/EtOAc) to afford 214 mg (72% yield) of **13a** as slight yellow oil. [α]<sub>D</sub><sup>25</sup> +69.6 (*c* 1.15, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.19 (d, *J* = 6.8, 3H), 1.26-1.80 (m, 6H), 2.22 (td, *J* = 11.5, 2.4, 1H), 2.56 (d, *J* = 11.4, 1H), 3.49 (dd, *J* = 10.7, 2.8, 1H), 3.78 (q, *J* = 6.8, 1H), 3.92 (s, 3H), 6.75 (d, *J* = 8.5, 1H), 7.17-7.42 (m, 5H), 7.77 (dd, *J* = 2.3, 8.5, 1H), 8.16 (d, *J* = 2.0, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 8.4, 25.7, 26.4, 37.2, 45.3, 53.6, 55.2, 62.1, 111.5, 126.4, 127.7, 128.1,

133.4, 138.1, 144.6, 145.7, 163.8. Anal. Calcd. for C<sub>19</sub>H<sub>24</sub>N<sub>2</sub>O: C, 76.99; H, 8.16; N, 9.45. Found: C, 76.89; H, 8.09; N, 9.38.

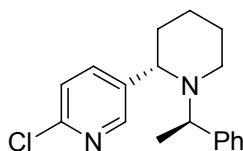


**(2R)-N-[(R)-1-Phenylethyl]-2-(5-methoxypyridin-3-yl)-piperidine (13b).** <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.06-1.19 (m, 1H), 1.35 (d, *J* = 7.2, 3H), 1.39-1.72 (m, 5H), 1.78 (td, *J* = 11.7, 2, 1H), 3.11 (m, 2H), 3.87 (q, *J* = 7.1, 1H), 3.97 (s, 3H), 6.79 (d, *J* = 8.5, 1H), 7.02 (d, *J* = 6.9, 2H), 7.21-7.31 (m, 3H), 7.70 (dd, *J* = 8.5, 2.3, 1H), 8.12 (d, *J* = 2.2, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 18.9, 25.2, 26.5, 38.3, 46.1, 53.6, 56.9, 62.5, 111.4, 127.0, 127.8, 129.0, 134.2, 138.2, 138.7, 146.1, 163.6.



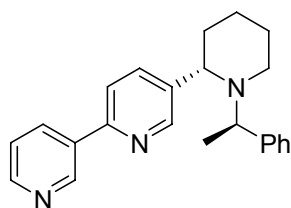
**(2R)-N-[(S)-1-Phenylethyl]-2-(5-methoxypyridin-3-yl)-piperidine (14a).** General procedure A was employed with **12** and (*S*)-1-phenylethylamine (99.5% ee) to afford 204 mg (69% yield) of **14a** as slight yellow oil. [ $\alpha$ ]<sub>D</sub><sup>25</sup> -62.8 (*c* 1.0, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.19 (d, *J* = 6.8, 3H), 1.30-1.80 (m, 6H), 2.22 (td, *J* = 11.6, 2.4, 1H), 2.56 (d, *J* = 11.4, 1H), 3.50 (dd, *J* = 10.8, 2.4, 1H), 3.78 (q, *J* = 6.8, 1H), 3.92 (s, 3H), 6.74 (d, *J* = 8.5, 1H), 7.17-7.41 (m, 5H), 7.76 (dd, *J* = 8.5, 1.6, 1H), 8.16 (d, *J* = 2.2, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 8.4, 25.7, 26.4, 37.2, 45.3, 53.6, 55.2, 62.2, 111.4, 126.4, 127.7, 128.1, 133.3, 138.1, 144.6, 145.7, 163.8. Anal. Calcd. for C<sub>19</sub>H<sub>24</sub>N<sub>2</sub>O:

C, 76.99; H, 8.16; N, 9.45. Found: C, 76.75; H, 8.04; N, 9.30.

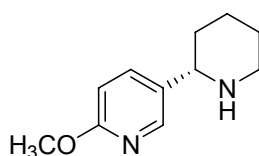


#### General Procedure B: Chlorination.

**(2S)-N-[(R)-1-Phenylethyl]-2-(5-chloropyridin-3-yl)piperidine (15).** The piperidine **13a** (0.40 g, 1.4 mmol) was dissolved in POCl<sub>3</sub> (3 mL) and sealed in a high-pressure reaction tube. The reaction was stirred at 120 °C for 17 hours. The cooled reaction mixture was slowly dripped into an ice-cold NaOH solution (2N, 75 mL) with continuous shaking to ensure each drop was well dissolved. The resulting mixture was extracted with EtOAc (2 × 50 mL) and CHCl<sub>3</sub> (2 × 50 mL). All the organic portions were combined, dried over MgSO<sub>4</sub>, filtered and concentrated using a rotary evaporator under reduced pressure. The residue was purified by preparative TLC (SiO<sub>2</sub>, 12:88 hexanes/EtOAc) to afford 373 mg (92% yield) of **15** as yellow solid, mp 66-69 °C.  $[\alpha]_D^{25} +48.4$  (c 0.5, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.20 (d, *J* = 6.8, 3H), 1.28-1.81 (m, 6H), 2.23 (td, *J* = 10.3, 2.4, 1H), 2.58 (d, *J* = 11.5, 1H), 3.56 (dd, *J* = 10.8, 2.4, 1H), 3.71 (q, *J* = 6.8, 1H), 7.17-7.39 (m, 6H), 7.82 (dd, *J* = 8.2, 2.2, 1H), 8.43 (d, *J* = 1.6, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 8.6, 25.4, 26.1, 37.3, 45.1, 55.6, 62.1, 124.7, 126.6, 127.6, 128.2, 138.1, 139.8, 144.0, 149.2, 150.2. Anal. Calcd. for C<sub>18</sub>H<sub>21</sub>ClN<sub>2</sub>: C, 71.87; H, 7.04; N, 9.31. Found: C, 71.71; H, 6.97; N, 9.16.

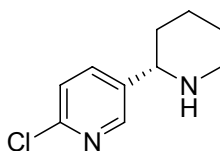


**(2S)-N-(R)-1-Phenylethyl-noranabasamine (16).** Under an atmosphere of argon, the piperidine **15** (184 mg, 0.61 mmol, 1.0 equiv), pyridine-3-boronic acid (113 mg, 0.92 mmol, 1.5 equiv), allyl[1,3-bis(2,6-diisopropylphenyl)imidazol-2-ylidene]palladium(II) chloride (35 mg, 0.0612 mmol, 0.1 equiv), sodium *tert*-butoxide (118 mg, 1.2 mmol, 2.0 equiv ) were added to dry dioxane (10 mL). The reaction mixture was stirred vigorously at 90 °C for 8 hours. The cooled reaction mixture was filtered through celite 545 (2 g) and rinsed with EtOAc (5 mL) and CHCl<sub>3</sub> (5 mL). The filtrate was concentrated using a rotary evaporator under reduced pressure. The residue was purified by preparative TLC (SiO<sub>2</sub>, 30:70 hexanes /EtOAc) to afford 137 mg (65% yield) of **16** as light yellow oil. <sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 1.24 (d, *J* = 6.8, 3H), 1.92-1.33 (m, 6H), 2.27 (td, 11.6, 2.8, 1H), 2.61 (d, *J* = 11.5, 1H), 3.64 (dd, *J* = 10.8, 2.4, 1H), 3.82 (q, *J* = 6.8, 1H), 7.20 (t, *J* = 7.3, 1H), 7.30 (t, *J* = 7.6, 2H), 7.39 (dd, *J* = 4.8, 7.9, 1H), 7.44 (d, *J* = 7.5, 2H), 7.74 (d, *J* = 8.1, 1H), 7.95 (dd, *J* = 8.0, 2.0, 1H), 8.31 (dt, *J* = 8.0, 1.6, 1H), 8.64 (d, *J* = 3.2, 1H), 8.78 (d, *J* = 1.6, 1H), 9.18 (s, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 8.6, 25.6, 26.3, 37.4, 45.2, 55.6, 62.7, 120.9, 123.8, 126.6, 127.6, 128.2, 134.4, 135.0, 136.3, 140.1, 144.2, 148.4, 149.8, 150.0, 153.9.



#### General Procedure C. Hydrogenolysis.

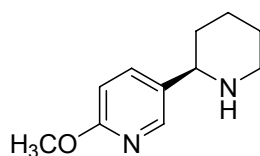
**(S)-2-(5-Methoxypyridin-3-yl)-piperidine (17).** The piperidine **13a** (166 mg, 0.56 mmol), Pd on carbon (10%) (65 mg) and ethanol (20 mL) were stirred in a 100 mL round bottom flask under an atmosphere of hydrogen (1 atm) for 2 hours at 55 °C. The cooled reaction mixture was filtered through celite 545 (2 g) and rinsed with EtOAc (2 × 10 mL). The filtrate was concentrated using a rotary evaporator under reduced pressure to afford **17** as a light yellow oil that was carried on to next step without further purification.  $[\alpha]_D^{25}$  -22.9 (*c* 1.0, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.40-1.89 (m, 7H), 2.78 (td, *J* = 11.6, 2.4, 1H), 3.16 (d, *J* = 11.3, 1H), 3.55 (dd, *J* = 10.4, 2.8, 1H), 3.91 (s, 3H), 6.70 (d, *J* = 8.5, 1H), 7.63 (dd, *J* = 8.5, 2.4, 1H), 8.09 (d, *J* = 2.4, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 25.4, 25.9, 34.8, 47.9, 53.6, 59.4, 110.9, 133.7, 137.6, 145.3, 163.8.



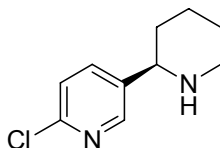
#### General Procedure D: Chlorination.

**(S)-2-(5-Chloropyridin-3-yl)piperidine (18).** The unpurified piperidine **17** was dissolved in POCl<sub>3</sub> (3 mL) and sealed in a high-pressure reaction tube. The reaction was stirred at 120 °C for 1.5 hours. The cooled reaction mixture was slowly dropped into an ice-cold NaOH solution (2N, 75 mL) with continuous shaking to ensure each drop was well dissolved. The resulting mixture was extracted with EtOAc (2 × 50 mL) and CHCl<sub>3</sub> (2 × 50 mL). All the organic portions were combined, dried over MgSO<sub>4</sub>, filtered and concentrated using a rotary evaporator under reduced pressure. The residue was purified by preparative TLC (SiO<sub>2</sub>, 1:99 Et<sub>3</sub>N/EtOAc) to afford 62

mg (56% yield, 2 steps) of **18** as a yellow solid, mp 53-57 °C.  $[\alpha]_D^{25}$  -27.8 (*c* 1.0, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.39-2.14 (m, 7H), 2.77 (td, *J* = 11.6, 2.8, 1H), 3.17 (d, *J* = 11.6, 1H), 3.62 (dd, *J* = 10.5, 2.8, 1H), 7.24-7.27 (m, 1H), 7.69 (dd, *J* = 2.5, 8.2, 1H), 8.34 (d, *J* = 2.1, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 25.3, 25.7, 35.1, 47.7, 59.2, 124.3, 137.5, 139.8, 148.6, 150.3. Anal. Calcd. For C<sub>10</sub>H<sub>13</sub>ClN<sub>2</sub>: C, 61.07; H, 6.66; N, 14.24. Found: C, 60.53; H, 6.62; N, 13.66.

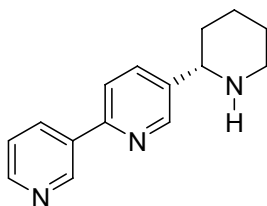


**(R)-2-(5-Methoxypyridin-3-yl)-piperidine (19).** General Procedure C using the piperidine **14a** (158 mg, 0.53 mmol) afforded **19** as a light yellow oil that was carried on to next step without further purification.  $[\alpha]_D^{25}$  +24.2 (*c* 1.0, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.41-1.97 (m, 7H), 2.79 (td, *J* = 11.6, 2.8, 1H), 3.17 (d, *J* = 11.3, 1H), 3.56 (dd, *J* = 10.0, 2.4, 1H), 3.92 (s, 3H), 6.70 (d, *J* = 8.5, 1H), 7.63 (dd, *J* = 2.4, 8.5, 1H), 8.09 (d, *J* = 2.4, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 25.5, 25.9, 34.9, 48.0, 53.6, 59.5, 110.9, 133.9, 137.6, 145.2, 163.8.



**(R)-2-(5-Chloropyridin-3-yl)-piperidine (20).** General Procedure D using the unpurified piperidine **19** afforded 64 mg (61% yield, 2 steps) of **20** as yellow solid, mp 55-57 °C.  $[\alpha]_D^{25}$  +28.4 (*c* 0.5, CH<sub>3</sub>OH). <sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 1.40-1.92 (m, 7H), 2.79 (td, *J* = 11.6, 2.8, 1H), 3.19 (d, *J* = 11.6, 1H), 3.64 (dd, *J* = 10.8, 2.8, 1H), 7.28 (d, *J* = 8.1, 1H), 7.70 (dd, *J* = 8.2, 2.5, 1H),

8.36 (d,  $J = 2.4$ , 1H).  $^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  25.3, 25.8, 35.2, 47.8, 59.3, 124.3, 137.5, 140.0, 148.6, 150.2. Anal. Calcd. For  $\text{C}_{10}\text{H}_{13}\text{ClN}_2$ : C, 61.07; H, 6.66; N, 14.24. Found: C, 61.27; H, 6.86; N, 13.78.

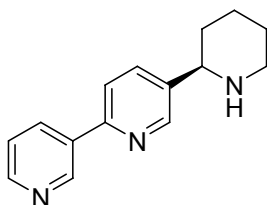


#### General Procedure E: Suzuki-Miyaura Coupling Reaction.

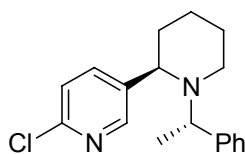
**(S)-Noranabasamine [(-)-1].** The piperidine **18** (110 mg, 0.56 mmol, 1.0 equiv), 3-pyridine boronic acid (83 mg, 0.68 mmol, 1.2 equiv),  $[\text{Pd}_2(\text{dba})_3]$  (5.2 mg, 0.0056 mmol, 0.01 equiv),  $\text{PCy}_3$  (3.8 mg, 0.014 mmol, 0.024 equiv), dioxane (1.5 mL) and aqueous  $\text{K}_3\text{PO}_4$  (1.27M, 0.75 mL, 0.95 mmol, 1.7 equiv) were placed under an atmosphere of argon in a high-pressure tube. The pressure tube was sealed and heated in an oil bath at 100 °C for 18 hours with vigorous stirring. The cooled mixture was diluted with EtOAc (10 mL) and  $\text{CHCl}_3$  (10 mL). The resulting mixture was dried over  $\text{MgSO}_4$ , filtered through celite 545 (2 g) and rinsed with EtOAc (5 mL) and  $\text{CHCl}_3$  (5 mL). The filtrate was concentrated using a rotary evaporator under reduced pressure. The residue was purified by preparative TLC ( $\text{SiO}_2$ , 2:98  $\text{Et}_3\text{N}/\text{EtOAc}$ ) to afford 113 mg (84% yield) of (-)-**1** as a yellow solid, mp 81-83 °C.  $[\alpha]_{\text{D}}^{25}$  -32.9 ( $c$  0.33,  $\text{CH}_3\text{OH}$ ).  $^1\text{H}$  NMR ( $\text{CDCl}_3$ )  $\delta$  1.46-1.98 (m, 6H), 2.25 (bs, 1H), 2.83 (td,  $J = 11.6, 2.8$ , 1H), 3.23 (d,  $J = 11.4$ , 1H), 3.71 (dd,  $J = 10.4, 2.4$ , 1H), 7.39 (dd,  $J = 8.0, 4.8$ , 1H), 7.72 (d,  $J = 8.1$ , 1H), 7.85 (dd,  $J = 8.2, 2.2$ , 1H), 8.30 (dt,  $J = 8.1, 2.4$ , 1H), 8.64 (dd,  $J = 4.8, 1.6$ , 1H), 8.68 (d,  $J = 2.1$ , 1H), 9.18 (d,  $J =$



2.4, 1H).  $^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  25.4, 25.8, 35.0, 47.8, 59.7, 120.6, 123.8, 134.5, 135.0, 135.5, 140.0, 148.4, 149.2, 150.0, 154.0.

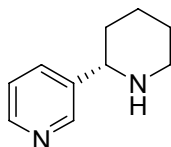


**(R)-Noranabasamine [(+)-1].** General Procedure E using **20** (94 mg, 0.48 mmol) afforded 69 mg (76% yield, based upon recovered starting material) of (+)-**1** as yellow solid, mp 78-80 °C.  $[\alpha]_{\text{D}}^{25} +34.6$  ( $c$  0.5,  $\text{CH}_3\text{OH}$ ).  $^1\text{H}$  NMR ( $\text{CDCl}_3$ )  $\delta$  1.46-1.98 (m, 6H), 2.49 (bs, 1H), 2.83 (td,  $J$  = 11.6, 2.4, 1H), 3.23 (d,  $J$  = 11.7, 1H), 3.72 (dd,  $J$  = 10.8, 2.4, 1H), 7.39 (dd,  $J$  = 8.0, 4.8, 1H), 7.72 (d,  $J$  = 8.2, 1H), 7.87 (dd,  $J$  = 8.2, 2.2, 1H), 8.30 (dt,  $J$  = 8.0, 2.0, 1H), 8.64 (dd,  $J$  = 4.8, 1.6, 1H), 8.69 (d,  $J$  = 2.0, 1H), 9.17 (d,  $J$  = 2.2, 1H).  $^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  25.3, 25.7, 34.8, 47.8, 59.7, 120.6, 123.8, 134.4, 135.0, 135.5, 139.8, 148.4, 149.2, 150.0, 154.0.



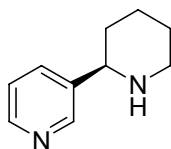
**(2R)-N-[(S)-1-Phenylethyl]-2-(5-chloropyridin-3-yl)piperidine (21).** General Procedure B using the piperidine **14a** (90 mg, 0.304 mmol) afforded 90 mg (90% yield) of **21** as yellow solid, mp 66-69 °C.  $[\alpha]_{\text{D}}^{25} -50.8$  ( $c$  1.0,  $\text{CH}_3\text{OH}$ ).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.20 (d,  $J$  = 6.8, 3H), 1.24-1.82 (m, 6H), 2.23 (td,  $J$  = 11.6, 2.6, 1H), 2.59 (d,  $J$  = 11.5, 1H), 3.57 (dd,  $J$  = 10.8, 2.7, 1H), 3.71 (q,  $J$  = 6.8, 1H), 7.18-7.40 (m, 6H), 7.82 (dd,  $J$  = 8.2, 2.4, 1H), 8.43 (d,  $J$  = 2.3, 1H).  $^{13}\text{C}$

NMR (CDCl<sub>3</sub>)  $\delta$  8.6, 25.4, 26.2, 37.4, 45.1, 55.6, 62.1, 124.7, 126.6, 127.5, 128.2, 138.1, 139.8, 144.0, 149.1, 150.2. Anal. Calcd. for C<sub>18</sub>H<sub>21</sub>ClN<sub>2</sub>: C, 71.87; H, 7.04; N, 9.31. Found: C, 71.66; H, 7.03; N, 9.19.



#### General Procedure F. Hydrogenolysis.

**(S)-Anabesine [(-)-3].** The piperidine **15** (131 mg, 0.44 mmol), Pd on carbon (10%) (65 mg) and ethanol (20 mL) were stirred in a 100 mL round bottom flask under an atmosphere of hydrogen (1 atm) for 2 hours at 55 °C. The cooled reaction mixture was filtered through celite 545 (2 g) and rinsed with EtOAc (2 × 10 mL). The filtrate was concentrated using a rotary evaporator under reduced pressure. The residue was purified by preparative TLC plates (2:98 Et<sub>3</sub>N/EtOAc) to afford 32 mg (45% yield) of (-)-**3** as sight yellow oil.  $[\alpha]_D^{25}$  -35.6 (*c* 0.5, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  1.44-1.99 (m, 7H), 2.81 (td, *J* = 11.6, 2.4, 1H), 3.21 (d, *J* = 11.4, 1H), 3.64 (dd, *J* = 10.0, 2.4, 1H), 7.25 (dd, *J* = 8.2, 5.3, 1H), 7.73 (dt, *J* = 7.9, 1.6, 1H), 8.49 (dd, *J* = 4.8, 1.2, 1H), 8.58 (d, *J* = 1.6, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  25.4, 25.8, 34.9, 47.8, 60.0, 123.7, 134.5, 140.8, 148.8, 149.9.



**(R)-Anabesine [(+)-3].** General Procedure F using the piperidine **21** (141 mg, 0.48 mmol)

afforded 31 mg (40% yield) of (+)-**3** as sight yellow oil.  $[\alpha]_D^{25} +36.7$  ( $c$  0.6, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  1.44-2.00 (m, 7H), 2.81 (td,  $J$  = 11.6, 2.8, 1H), 3.21 (d,  $J$  = 11.4, 1H), 3.64 (dd,  $J$  = 10.2, 2.6, 1H), 7.25 (dd,  $J$  = 8.0, 4.8, 1H), 7.72 (dt,  $J$  = 8.0, 1.8, 1H), 8.49 (dd,  $J$  = 4.8, 1.6, 1H), 8.59 (d,  $J$  = 4.8, 1.6, 1H). <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  25.4, 25.9, 35.0, 47.9, 60.0, 123.7, 134.4, 148.8, 148.9.

### 3.7. References and notes

1. For a review see: (a) Daly, J. W.; Spande, T. F.; Garraffo, H. M. *J. Nat. Prod.* **2005**, *68*, 1556-1575. (b) Daly, J. W. *J. Med. Chem.* **2003**, *46*, 445-452. (c) Gomes, A.; Giri, B.; Saha, A.; Mirsha, R.; Dasgupta, S. C.; Debnath, A.; Gomes, A. *Indian J. Exp. Biol.* **2007**, *45*, 579-593. (d) *Neuronal Nicotinic Receptors: Pharmacology and Therapeutic Opportunities*; Arneric, S. P., Brioni, J. D., Eds.; Wiley-Liss Inc.: New York, 1999.
2. Tokuyama, T.; Daly, J. W. *Tetrahedron* **1983**, *39*, 41-47.
3. Leete, E.; Mueller, M. E. *J. Am. Chem. Soc.* **1982**, *104*, 6440-6444.
4. Lovkova, M. Y.; Nurimov, E. *Isv. Akad. Nauk. SSSR Ser. Biol.* **1978**, 545-557.
5. Crooks, P. A.; Dwoskin, L. P. *Biochem. Pharmacol.* **1997**, *54*, 743-753.
6. (a) Tilyabaev, Z.; Abduvakhobov, A. A. *Chem. Nat. Compd.* **1998**, *34*, 295-297. (b) Tilyabaev, Z.; Aabd Mukhamedzhanova, Kh. S. *Dokl. Akad. Nauk UzSSR* **1984**, *8*, 45-47. (c) Mukhamedzhanova, Kh. S. *Dokl. Akad. Nauk UzSSR* **1983**, *7*, 47-49.
7. (a) Cheng, J.; Izenwasser, S.; Zhang, C.; Zhang, S.; Wade, D.; Trudell, M. L. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 1775-1778. (b) Nishiyama, T.; Gyermek, L.; Trudell, M. L.; Hanaoka, K. *Eur. J. Pharmacol.* **2003**, *470*, 27-31. (c) Cheng, J.; Zhang, C.; Stevens, E. D.; Izenwasser, S.; Wade, D.; Chen, S.; Paul, D.; Trudell, M. L. *J. Med. Chem.* **2002**, *45*, 3041-3047. (d) Cheng, J.; Izenwasser, S.; Wade, D.; Trudell, M. L. *Med. Chem. Res.* **2001**, *10*, 356-365.
8. (a) Hande, S. M.; Kawai, N.; Uenishi, J. *J. Org. Chem.* **2009**, *74*, 244-253. (b) Spangenberg,

- T.; Breit, B.; Mann, A. *Org. Lett.* **2009**, *11*, 261-264. (c) Castro, A.; Ramírez, J.; Juárez, J.; Terán, J. L.; Orea, L.; Galindo, A.; Gnecco, D. *Heterocycles* **2007**, *71*, 2699-2708. (d) Amat, M.; Bassas, O.; Llor, N.; Cantó, M.; Pérez, M.; Molins, E.; Bosch, J. *Chem. Eur. J.* **2006**, *12*, 7872-7881. (e) Ayers, J. T.; Xu, R.; Dwoskin, L. P.; Crooks, P. A. *AAPS J.* **2005**, *7*, E752-E758. (f) Amat, M.; Cantó, M.; Llor, N.; Bosch, J. *Chem. Commun.* **2002**, *5*, 526-527. (g) Felpin, F.-X.; Girard, S.; Vo-Thanh, G.; Robins, R. J.; Villiéras, J.; Lebreton, J. *J. Org. Chem.* **2001**, *66*, 6305-6312. (h) Felpin, F.-X.; Vo-Thanh, G.; Robins, R. J.; Villiéras, J.; Lebreton, J. *Synlett* **2000**, *11*, 1646-1648. (i) Hattori, K.; Yamamoto, H. *Tetrahedron* **1993**, *49*, 1749-1760. (j) Kunz, H.; Pfrengle, W. *Angew. Chem., Int. Ed. Engl.* **1989**, *101*, 1041-1042. (k) Pfrengle, W.; Kunz, H. *J. Org. Chem.* **1989**, *54*, 4261-4263. (l) Giovannini, A.; Savoia, D.; Umani-Ronchi, A. *J. Org. Chem.* **1989**, *54*, 228-234.
9. Fujita, K.-I.; Fujii, T.; Yamaguchi, R. *Org. Lett.* **2004**, *6*, 3525-3528.
  10. Xu, J.; Wei, T.; Zhang, Q. *J. Org. Chem.* **2003**, *68*, 10146-10151.
  11. Yoshida, Y.; Sakakura, Y.; Aso, N.; Okada, S.; Tanabe, Y. *Tetrahedron* **1999**, *55*, 2183-2192.
  12. Najdi, S.; Kurth, M. J. *Tetrahedron Lett.* **1990**, *31*, 3279-3282.
  13. Commercially available from Alfa Aesar Chemical Co. with enantiopurity of 99% ee. The (*S*)-enantiomer was available in 99.5% ee.
  14. Ravard, A.; Crooks, P. A. *Chirality* **1996**, *8*, 295-299.
  15. For a review, see: Li, J. J.; Gribble, G. W. *Palladium in Heterocyclic Chemistry*; Pergamon: Amsterdam, 2000; pp 191-197.
  16. Viciu, M. S.; Germaneau, R. F.; Navarro-Fernandez, O.; Stevens, E. D.; Nolan, S. P. *Organometallics* **2002**, *21*, 5470-5472.
  17. Tsai, M. R.; Chen, B. F.; Cheng, C. C.; Chang, N. C. *J. Org. Chem.* **2005**, *70*, 1780-1785.
  18. Sekine, M.; Tobe, M.; Nagayama, T.; Wada, T. *Lett. Org. Chem.* **2004**, *1*, 179-182.
  19. Campbell, A. L.; Pilipauskas, D. R.; Khanna, I. K.; Rhodes, R. A. *Tetrahedron Lett.* **1987**, *28*, 2331-2334.
  20. Laschat, S.; Fox, T. *Synthesis* **1997**, 475-479.
  21. Amat, M.; Escolano, C.; Gómez-Esqué, A.; Lozano, O.; Llor, N.; Grier, R.; Molins, E.; Bosch, J. *Tetrahedron: Asymmetry* **2006**, *17*, 1581-1588.

22. Miao, G.; Ye, P.; Yu, L.; Baldino, C. M. *J. Org. Chem.* **2005**, *70*, 2332-2334.
23. Zhang, C.; Huang, J.; Trudell, M. L.; Nolan, S. P. *J. Org. Chem.* **1999**, *64*, 3804-3805.
24. Kudo, N.; Perseghini, M.; Fu, G. C. *Angew. Chem., Int. Ed.* **2006**, *45*, 1282-1284.
25. The specific rotation for the natural material was reported as  $[\alpha]_{\text{D}}$  -14.4 (CH<sub>3</sub>OH). See reference 2.
26. See experimental section and appendix for experimental details and spectra.

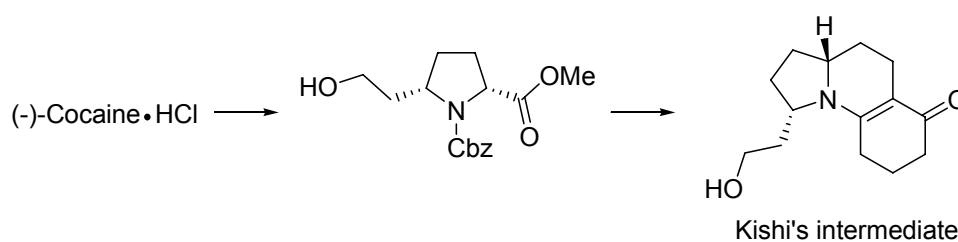
## CHAPTER 4

### A Formal Synthesis of (+)-Gephyrotoxin-Kishi's Intermediate

#### 4.1. Abstract

A cis-2,5-disubstituted pyrrolidine building block derived from (-)-cocaine•HCl was prepared. We utilized this compound as a chiral building block for the formal synthesis of (+)-gephyrotoxin. Using this pyrrolidine building block, Kishi's intermediate was obtained enantiospecifically in 15 steps and 9.4% overall yield.

**Scheme 4.1.** General approach for the formal synthesis of Kishi's intermediate

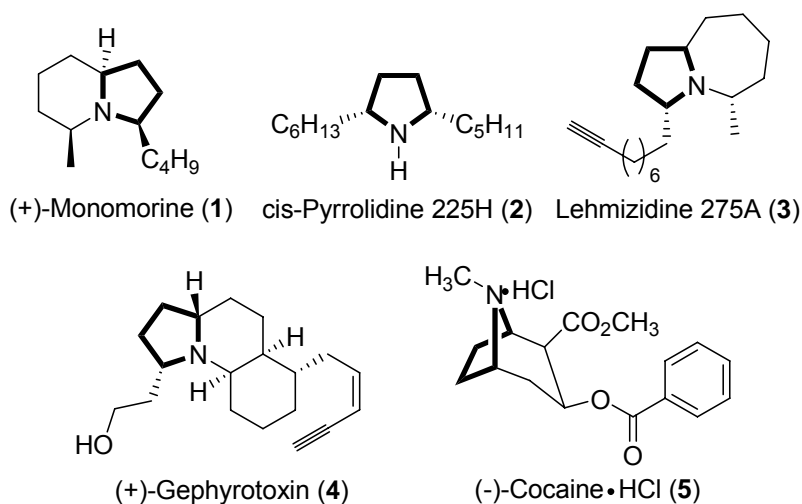


#### 4.2. Introduction

Lipophilic alkaloids detected in amphibian skin have aroused tremendous academic and

pharmaceutical interest due to their structural diversity and biological activity. Over 800 amphibian alkaloids comprising over 20 structural classes of alkaloids have been reviewed through 2005.<sup>1</sup> However, the paucity of these alkaloids from natural resources have made total synthesis the only practical method to provide sufficient material for intensive structural and biological activity studies. An ongoing project in our laboratory has developed synthetic strategies for the construction of amphibian alkaloids that exhibit pharmacological activity mediated by nicotinic receptor ion channels.<sup>2,3</sup>

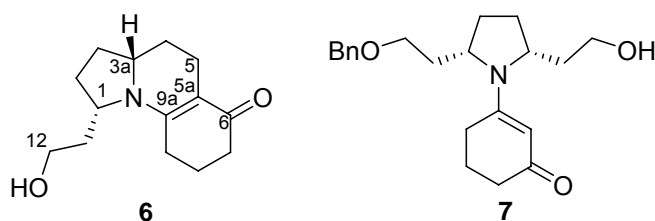
**Figure 4.1.** Structure of amphibian alkaloids



At least four classes of these alkaloids are found to share the common structural feature of a cis-2,5-disubstituted pyrrolidine ring system. As shown in Figure 4.1, they are represented by the natural products: (+)-monomorine (1), cis-pyrrolidine 225H (2), lehmizidine 275A (3) and

(+)-gephyrotoxin (**4**). The structural similarity encouraged us to design a general and effective synthetic method that would allow enantioselective access to these compounds as well as their analogues. Our approach utilized the abundant natural product cocaine (**5**) as the starting material. Cocaine has four chiral centers, two of which can be directly introduced into the target molecules. We have previously completed and reported a synthesis of (-)-monomorphine, the enantiomer of the natural product (+)-monomorphine.<sup>4</sup>

**Figure 4.2.** Structure of Kishi's intermediate



Gephyrotoxin was first isolated and characterized in 1977 from the skin of tropical frogs *Dendrobates histrionicus*.<sup>5</sup> Initial studies revealed this compound as muscarinic antagonist with low activity.<sup>6</sup> Recent studies have indicated it as a nontoxic noncompetitive blocker of nicotinic receptors.<sup>7</sup> Due to its interesting array of neurological activities and scarcity of this product in nature, several groups have conducted and reported the synthesis of gephyrotoxin in racemic or enantiopure forms.<sup>8-11</sup> A few reported syntheses involved the common enantiopure tricyclic intermediate known as Kishi's intermediate (**6**) (Figure 4.2). Compound **7** was synthesized both in Kishi and Lhomment's work to get intermediate **6**.<sup>10,12</sup> We report herein an efficient formal

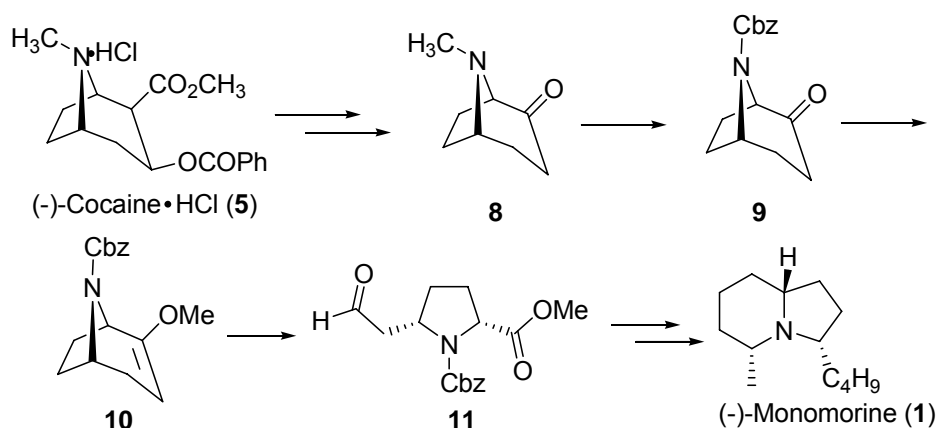


synthesis of (+)-gephyrotoxin (**1**) with a different approach to Kishi's intermediate (**6**) other than the structure of **7**.

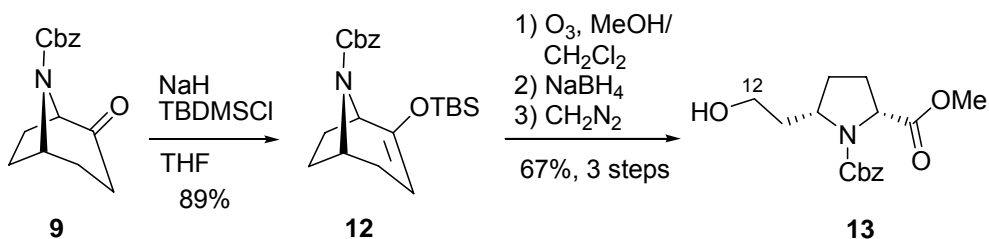
### 4.3. Results and discussion

We have reported the synthesis of (-)-monomorphine using Cbz-carbamate **9** derived for (-)-cocaine•HCl (**5**).<sup>4,13</sup> The pyrrolidine building block **11** has been developed from Cbz-carbamate **8** through the intermediate methyl enol ether **10** (Scheme 4.2). While the instability of compound **9** encouraged us to revise the procedure of generating cis-2,5-disubstituted pyrrolidine building block.

**Scheme 4.2.** Pyrrolidine building block in the synthesis of (-)-monomorphine



**Scheme 4.3.** Revised procedure of pyrrolidine building block

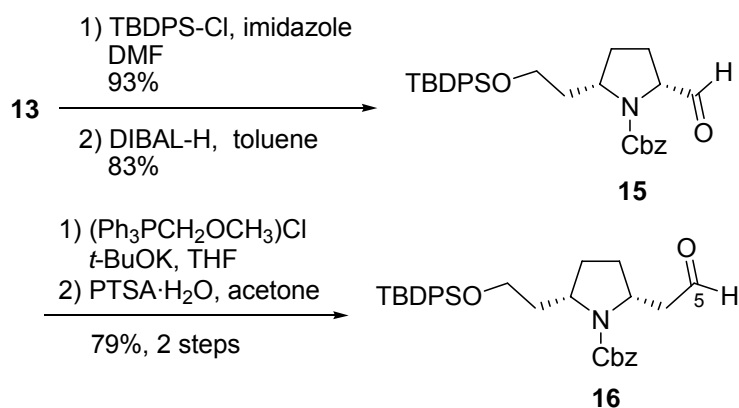


As shown in Scheme 4.3, Cbz-carbamate **9** was readily available in our lab and was treated with NaH and TBDMSCl to furnish silyl enol ether **12** according to the procedure reported by Rassat and coworkers.<sup>14</sup> Ether **12** was stable to chromatography for an excellent yield of 89%. The enol ether **12** was subjected to ozonolysis conditions the double bond was cleaved by ozone at  $-78^\circ\text{C}$ . The ozonide was reduced with  $\text{NaBH}_4$  in the subsequent step, then the reaction mixture was treated with  $\text{CH}_2\text{N}_2$  to furnish our new pyrrolidine building block **13** with 67% yield over three steps.<sup>4,14</sup> It is noteworthy that the functional moiety at the C12 position was introduced by the reduction with  $\text{NaBH}_4$  in the step after the ozonolysis. We still could use triphenylphosphine as we used before to keep the aldehyde moiety group ending with the same pyrrolidine building block **11**.<sup>4</sup> While in this case,  $\text{NaBH}_4$  was chosen as reduction reagent to generate alcohol moiety at C12 position.

In order to install the C5 to our pyrrolidine building block **13**, the alcohol function was converted to silyl ether **14** using TBDPS-Cl (Scheme 4.4).<sup>15</sup> Then the protected building block **14** was subjected to reduction using DIBAL-H to generate the corresponding aldehyde **15**.<sup>4</sup> These two reactions went smoothly with a yield of 93% and 83% respectively. A Wittig

olefination reaction was employed to install the C5 unit using  $(\text{Ph}_3\text{PCH}_2\text{OCH}_3)\text{Cl}$  and  $t\text{-BuOK}$ . The subsequent step was treated with  $\text{PTSA}\cdot\text{H}_2\text{O}$  and acetone ending with the desired aldehyde **16** with 79% yield over two steps.<sup>4,14</sup> The above reactions have been optimized and proved to be useful substrates for our pyrrolidine synthesis.

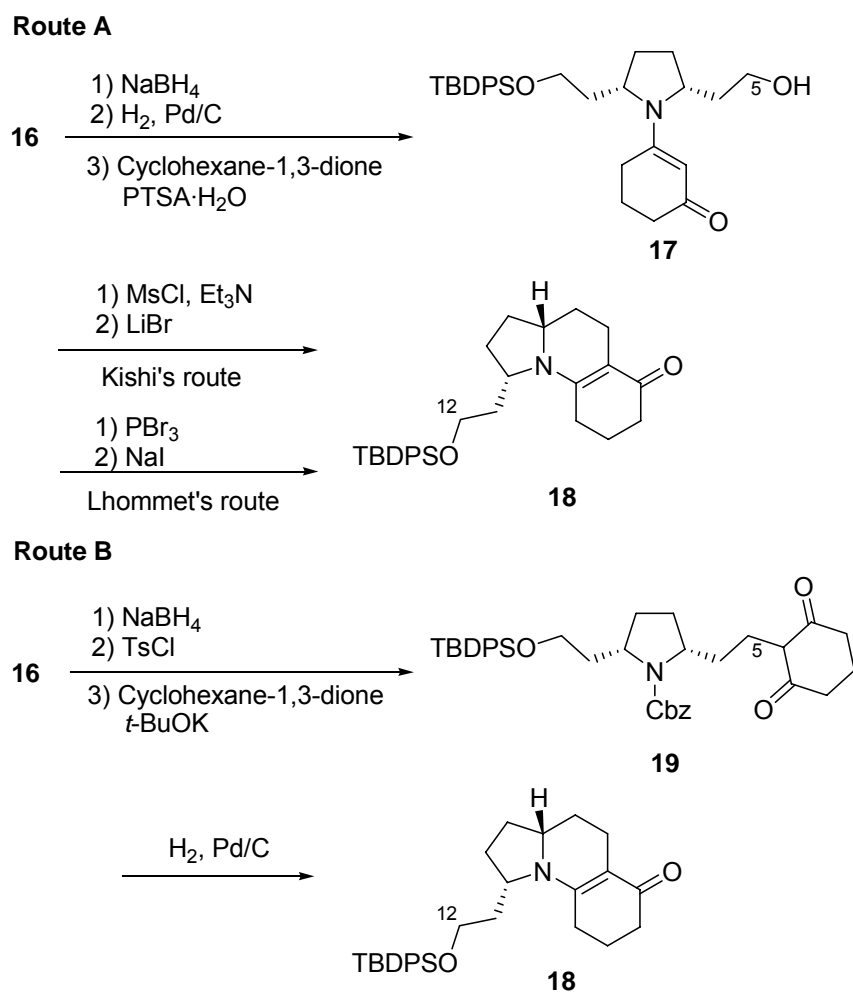
**Scheme 4.4.** Installation C5 into pyrrolidine building block



With aldehyde **16** in hand, we could follow Kishi and Lhommet's procedure for the construction of the tricyclic ring system (Scheme 4.5, Route A).<sup>8a,10,12</sup> First, the aldehyde at the C5 position could be converted into the alcohol using  $\text{NaBH}_4$ . Second, the Cbz protecting group would be removed by  $\text{Pd/C}$  under hydrogen atmosphere. Third, cyclohexane-1,3-dione could be coupled in the presence of  $\text{PTSA}\cdot\text{H}_2\text{O}$  to form compound **17**. Enolamine **17** is a similar structure compared to **7** which was generated through both Kishi and Lhommet's routes, respectively. Using either Kishi or Lhommet's route, there will be a two step reaction sequence

for converting compound **17** into the tricyclic compound **18** (the C12 silyl ether protected Kishi's intermediate). The total synthesis requires five steps from **16** to **18** in Route A.

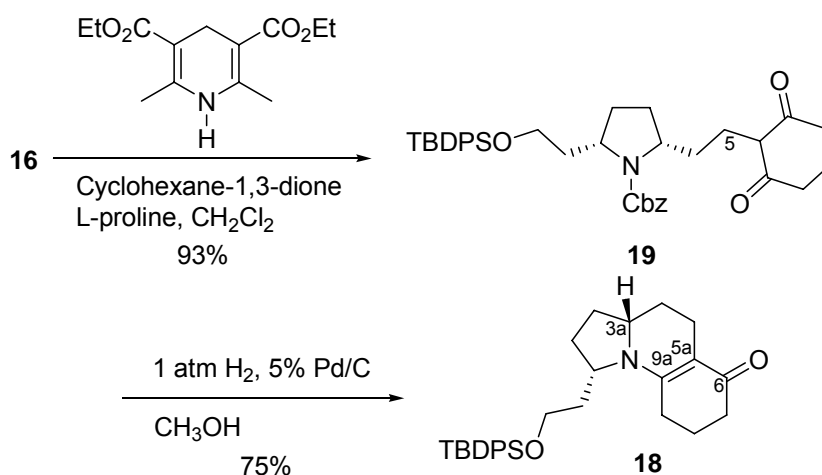
**Scheme 4.5.** Proposed routes for cyclization



As shown in Scheme 4.5, Route B was our original proposed route for synthesizing the silyl ether protected Kishi's intermediate (**18**). First, the aldehyde was treated by  $\text{NaBH}_4$  to

generate the corresponding alcohol at the C5 position. Second, the alcohol at C5 was converted to tosylate group by using TsCl. Third, cyclohexane-1,3-dione was attached using *t*-BuOK to get compound **19** which is our new approach to the Kishi's intermediate (**6**). Structure **19** was different from compound **7** which was included in Kishi's and Lhomment's reports. Therefore, 3 steps were employed already here. Compound **19** was subjected to cyclization under 1 atmosphere of hydrogen to furnish the tricyclic compound **18** as the silyl ether protected Kishi's intermediate. Route B was a 4 step procedure from **16** to **18**.

**Scheme 4.6.** Revised route for cyclization



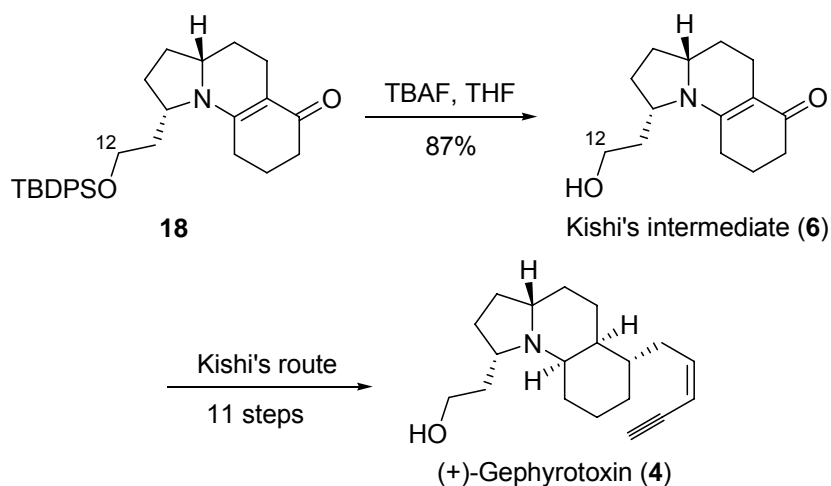
In 2007, Kishor and Ramachary developed a methodology of coupling aldehydes with cyclohexane-1,3-dione with good yields based on different aldehyde substrates.<sup>16</sup> Kishor and Ramachary's methodology was employed in our pyrrolidine building block aldehyde **16** (Scheme 4.6). Upon dissolving aldehyde **16** dissolved in dichloromethane, diethyl

2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxylate and cyclohexane-1,3-dione were added to the mixture in one portion. Subsequently, L-proline was added as a catalyst in this reaction. The reaction was monitored by TLC and proceeded to completion within one hour. The reaction mixture was directly purified by chromatography to furnish compound **19** with an excellent yield of 93% without an aqueous workup. Then compound **19** was subjected to hydrogenation yielding the tricyclic compound **18** in 75%. The Cbz deprotection, cyclization and enamine elimination occurred as one step. Our new synthetic route converted **16** to **18** in two steps with excellent yields compared to Routes A and B (Scheme 4.5). Structure **18** was confirmed by  $^{13}\text{C}$  NMR at positions C5a, C6 and C9a. Proton and carbon NMR indicated that the other diastereomer at the C3a position was present in 10%. The isomerization may have occurred during ozonolysis, DIBAL-H reduction or hydrogen cyclization. Our new route has less steps, is more efficient, and produces higher yields. In addition, our pyrrolidine building block is amenable to all of the above mentioned reactions with good yields.

According to Gerasyuto and coworkers, the two diastereomers can be separated by chromatography after the deprotection of TBDPS group.<sup>11</sup> Compound **18** was subjected to cleavage of TBDPS protection group at C12 position (Scheme 4.7).<sup>17</sup>

Typical treatment with TBAF in THF resulted in Kishi's intermediate (**6**) in 87% yield. NMR spectra and the melting point of **6** matched the literature data,<sup>10</sup> and the absolute configuration of Kishi's intermediate (**6**) has been confirmed by X-Ray crystallography (See appendix for details).

**Scheme 4.7.** Synthesis of (+)-gephyrotoxin (**4**)-Kishi's intermediate (**6**)



**4.4. Conclusion**

In summary, our pyrrolidine building block derived from (-)-cocaine•HCl (**5**) can be used for the synthesis of complex amphibian alkaloids. With this strategy, we have prepared Kishi's intermediate (**6**) of (+)-gephyrotoxin (**4**) from (-)-cocaine•HCl (**5**) in 15 steps and 9.4% overall yield. Compared to previous syntheses of Kishi's intermediate, our approach had less steps than Kishi's route (18 steps) and a higher diastereoselective ratio than the route of Gerasyuto and coworkers. In addition, condensation reaction with cyclohexane-1,3-dione and subsequent intramolecular cyclization employed in Kishi's route and Lhomme's route were significantly improved by our new synthetic approach.

#### **4.5. Acknowledgment**

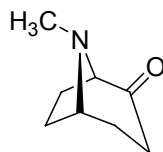
This research was funded by the National Institute on Drug Abuse (DA11528) and the University of New Orleans.

#### **4.6. Experimental section**

##### **General Experimental Methods**

All chemicals were purchased from Aldrich Chemical Company and used as received unless otherwise noted. Anhydrous dichloromethane was purchased from Mallinckrodt Baker, Inc. Confiscated grade (-)-cocaine hydrochloride was provided by NIDA Drug Supply System, Research Technology Branch, National Institute on Drug Abuse. Proton and carbon NMR were recorded on a Varian-400 MHz nuclear magnetic resonance spectrometer at ambient temperature in deuterated chloroform ( $\text{CDCl}_3$ ) from Cambridge Isotope Laboratories, Inc.  $^1\text{H}$  NMR chemical shifts are reported as  $\delta$  values (ppm) relative to tetramethylsilane.  $^{13}\text{C}$  NMR chemical shifts are reported as  $\delta$  values (ppm) relative to chloroform-*d* (77.0 ppm). Optical rotations were measured on Autopol III autopolarimeter at the sodium D line (2 mL sample cell). Melting points (mp) were measured with an Electrothermal R Mel-Temp apparatus and are uncorrected.

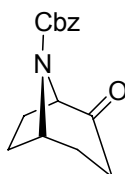




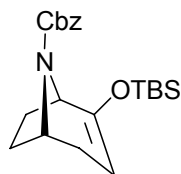
**(1*R*,5*S*)-8-Methyl-8-azabicyclo[3.2.1]octan-2-one (8).** A solution of (-)-cocaine hydrochloride (**5**) (34.0 g, 100 mmol) in concentrated hydrochloric acid (276 mL) was refluxed for 24 hours. After the mixture was cooled to room temperature, it was diluted with H<sub>2</sub>O (255 mL) and extracted with Et<sub>2</sub>O (2 × 255 mL) to remove benzoic acid. The aqueous phase was then evaporated under vacuum to dryness. The white solid was further dried under vacuum at 100 °C for 24 hours. This afforded crude white solid which without further purification was used in the next step.

To finely powdered the previous step crude white solid (20.0 g, 98.2 mmol) in a 2 L round bottom flask were added Na<sub>2</sub>CO<sub>3</sub> (25.4 g, 240 mmol) and 4-dimethylaminopyridine (DMAP) (305 mg, 2.50 mmol), and the vessel was sealed under an atmosphere of nitrogen. Dried CH<sub>2</sub>Cl<sub>2</sub> (366 mL) was added to the flask followed by addition of diphenylphosphorylazide (DPPA) (25.9 mL, 0.12 mol). The reaction mixture was stirred vigorously for 48 hours. The solvent was removed under vacuum, and the resulting residue was then dissolved in H<sub>2</sub>O (106 mL) followed by the careful addition of 1N HCl (604 mL). The solution was then heated in a preheated oil bath (120 °C) for 35 minutes (until the carbon dioxide and nitrogen evolution ceased). The aqueous HCl was removed under vacuum, and the residue was made basic (pH 9.5-10.0) with a saturated solution of Na<sub>2</sub>CO<sub>3</sub>. The aqueous solution was extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 × 500 mL). The combined organic fractions were dried (Na<sub>2</sub>SO<sub>4</sub>) and the solvent was removed under vacuum. The resulting liquid was purified by vacuum bulb-to-bulb distillation (Kugelrohr). This afforded

**8** (10.6 g, 76% yield, 3steps) as a colorless liquid.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.67-1.78 (m, 3H), 2.13-2.25 (m, 4H), 2.28-2.37 (m, 1H), 2.39 (s, 3H), 3.25-3.30 (m, 2H).

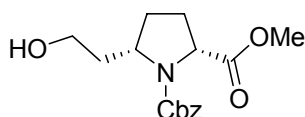


**(1R,5S)-Benzyl 2-oxo-8-azabicyclo[3.2.1]octane-8-carboxylate (9).** Benzyl chloroformate (18 mL, 128 mmol) was added to a solution of **8** (3.6 g, 26 mmol) and potassium carbonate (180 mg, 1.3 mmol) in toluene (80 mL). The solution was heated to reflux for 48 hours. The solvent was removed under reduced pressure, and the residue was dissolved in water (50 mL). The aqueous mixture was extracted with  $\text{CH}_2\text{Cl}_2$  ( $3 \times 50$  mL), and the combined organic layers were dried over  $\text{Na}_2\text{SO}_4$ . The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography ( $\text{SiO}_2$ , 50:50 hexanes/EtOAc) to afford **9** (3.42 g, 56% yield) as a colorless oil.  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.77-1.86 (m, 2H), 2.18-2.25 (m, 2H), 2.32-2.38 (m, 2H), 2.42-2.48 (m, 2H), 4.45-4.51 (m, 2H), 5.11-5.17 (m, 2H), 7.27-7.36 (m, 5H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  27.8, 30.4, 32.4, 52.8, 64.1, 67.0, 127.8, 128.0, 128.2, 128.4, 136.2, 153.8, 205.3. Anal. Calcd for  $\text{C}_{15}\text{H}_{17}\text{NO}_3$ : C, 69.48; H, 6.61; N, 5.40. Found: C, 69.65; H, 6.74; N, 5.62.



**(1R,5S)-Benzyl 2-(tert-butyldimethylsilyloxy)-8-azabicyclo[3.2.1]oct-2-ene-8-**

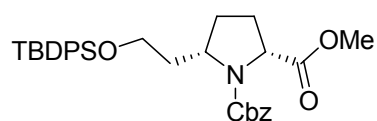
**carboxylate (12).** NaH (60 mg, 2.5 mmol, 5.0 equiv) was suspended in dry THF (4 mL) under nitrogen at 0 °C with ice bath. A solution of compound **9** (130 mg, 5 mmol, 1.0 equiv) in dry THF (2 mL) was added dropwise. The stirring was continued for 2 hours. Then TBDMSCl (1.0M in THF, 1 mL, 1 mmol, 2.0 equiv) was added dropwise at 0 °C. The stirring was continued overnight. At 0 °C, water (5 mL) was added slowly. The solution was extracted with Et<sub>2</sub>O (3 × 10 mL). All combined organic portions were dried over MgSO<sub>4</sub> and evaporated. The crude product was purified by flash column chromatography (SiO<sub>2</sub>, 95:5 hexanes/EtOAc) to afford **12** (166 mg, 89% yield) as a colorless oil.  $[\alpha]_D^{25}$  -43.5 (*c* 1.2, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 0.14 (m, 6H), 0.90 (s, 9H), 1.58-1.67 (m, 2H), 1.74 (dd, *J* = 16.4, 4.6, 1H), 1.94-2.18 (m, 3H), 2.61-2.79 (m, 0.5H), 4.11-4.49 (m, 2.5H), 5.09-5.19 (m, 2H), 7.28-7.35 (m, 5H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ -4.5, -4.1, 18.2, 25.8, 29.4, 30.2, 31.4, 32.4, 33.7, 34.4, 52.5, 57.7, 66.8, 97.3, 128.0, 128.1, 128.6, 137.1, 154.4, 154.8. Anal. Calcd for C<sub>21</sub>H<sub>31</sub>NO<sub>3</sub>Si: C, 67.52; H, 8.36; N, 3.79. Found: C, 67.73; H, 8.58; N, 3.79.



**(2*R*,5*S*)-1-Benzyl 2-methyl 5-(2-hydroxyethyl)pyrrolidine-1,2-dicarboxylate (13).** Compound **12** (781 mg, 2.1 mmol, 1.0 equiv) was dissolved in CH<sub>2</sub>Cl<sub>2</sub> (50 mL) and CH<sub>3</sub>OH (5 mL). At -78 °C, O<sub>3</sub> was bubbled into the solution. Soon the solution showed a slight blue color. O<sub>3</sub> was continued to bubble through the solution for 15 more minutes and then removed. N<sub>2</sub> was bubbled through for 10 minutes. The solution was still kept at -78 °C. NaBH<sub>4</sub> (250 mg) was added by one portion. After 30 minutes, another portion NaBH<sub>4</sub> (300 mg) was added. The mixture was warmed

to room temperature. The solvent was removed under reduced pressure. The residue was triturated with 2N HCl (25 mL). The aqueous layer was extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 × 20 mL), and the combined organic layers were dried over MgSO<sub>4</sub> and evaporated to afford an oil, which was used directly in the next step.

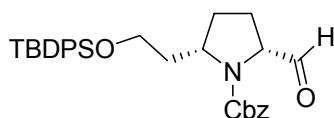
To a stirred solution of the previous oil in Et<sub>2</sub>O (20 mL) at 0 °C, CH<sub>2</sub>N<sub>2</sub> was bubbled through the solution until the yellow color shown up. Then the CH<sub>2</sub>N<sub>2</sub> was stopped and N<sub>2</sub> was bubbled through the solution for 30 minutes. The cold bath was removed and the resulting mixture was purified by flash column chromatography (SiO<sub>2</sub>, 40:60 hexanes/EtOAc) to afford 430 mg (67% yield, 3 steps) of **13** as a colorless oil.  $[\alpha]_D^{25} +52$  (*c* 0.6, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.61-1.82 (m, 3H), 1.95-2.11 (m, 2H), 2.30-2.37 (m, 1H), 3.60 (s, 3H), 3.64-3.82 (m, 3H), 3.93 (dd, *J* = 9.8, 4.6, 1H), 4.37 (t, *J* = 8.3, 1H), 5.03-5.22 (m, 2H), 7.28-7.37 (m, 5H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 29.2, 30.9, 37.8, 52.4, 55.8, 59.1, 59.9, 67.8, 127.9, 128.3, 128.7, 136.4, 156.1, 173.6. Anal. Calcd for C<sub>16</sub>H<sub>21</sub>NO<sub>5</sub>: C, 62.53; H, 6.89; N, 4.56. Found: C, 62.28; H, 7.00; N, 4.49.



**(2*R*,5*S*)-1-Benzyl 2-methyl 5-(2-(*tert*-butyldiphenylsilyloxy)ethyl)pyrrolidine-1,2-**

**dicarboxylate (14).** Compound **13** (374 mg, 1.22 mmol, 1.0 equiv) and imidazole (166 mg, 2.44 mmol, 2.0 equiv) was dissolved in dry DMF (15 mL) under nitrogen at 0 °C. TBDPS-Cl (402 mg, 0.374 mL, 1.46 mmol, 1.2 equiv) was added drop wise to the mixture. The mixture was allowed to warm to room temperature and stirred overnight. At 0 °C, H<sub>2</sub>O (15 mL) was added to the

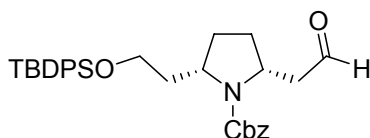
mixture to quench the reaction. The mixture was extracted with Et<sub>2</sub>O (2 × 30 mL). The combined organic layers were dried over MgSO<sub>4</sub> and evaporated under reduced pressure. The resulting mixture was purified by flash column chromatography (SiO<sub>2</sub>, 85:15 hexanes/EtOAc) to afford 617 mg (93% yield) of **14** as a colorless oil.  $[\alpha]_D^{25} +22.1$  (*c* 0.73, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.03 (d, *J* = 11.0, 9H), 1.61-2.02 (m, 4H), 2.15-2.40 (m, 2H), 3.58 (s, 1H), 3.65-3.79 (m, 4H), 4.09-4.16 (m, 1H), 4.31-4.41 (m, 1H), 5.01-5.20 (m, 2H), 7.26-7.44 (m, 11H), 7.63 (t, *J* = 6.4, 4H). <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 19.4, 27.0, 28.4, 29.4, 29.9, 30.2, 36.5, 37.2, 52.2, 52.4, 56.5, 57.7, 59.9, 60.2, 61.7, 62.1, 67.0, 67.3, 127.9, 128.1, 128.6, 128.7, 129.8, 134.0, 135.8, 136.9, 155.1, 173.6. Anal. Calcd for C<sub>32</sub>H<sub>39</sub>NO<sub>5</sub>Si: C, 70.43; H, 7.20; N, 2.57. Found: C, 70.64; H, 7.23; N, 2.56.



**(2*S*,5*R*)-Benzyl 2-(2-(*tert*-butyldiphenylsilyloxy)ethyl)-5-formylpyrrolidine-1-**

**carboxylate (15).** Compound **14** (547 mg, 1 mmol, 1.0 equiv) was dissolved in toluene (6 mL) under nitrogen. At -78 °C, DIBAL-H (1.0M in toluene, 1.5 mL, 1.5 equiv) was added dropwise over a period of 45 minutes. The stirring at -78 °C was continued for additional 15 minutes and then the cold bath was removed. Et<sub>2</sub>O (10 mL), H<sub>2</sub>O (4 mL) and 15% NaOH (6 mL) were added one by one. After warmed to room temperature, the mixture was extracted with Et<sub>2</sub>O (2 × 20 mL). The combined organic layers were dried over MgSO<sub>4</sub> and concentrated under vacuum. The resulting mixture was purified by flash column chromatography (SiO<sub>2</sub>, 80:20 hexanes/EtOAc) to

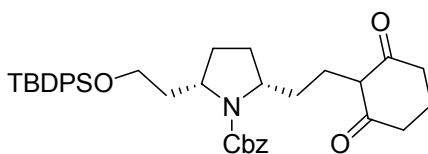
afford 431 mg (83% yield) of **15** as a colorless oil.  $[\alpha]_{\text{D}}^{25} +17.3$  ( $c$  0.95, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  1.06 (d,  $J$  = 6.7, 9H), 1.43-2.45 (m, 6H), 3.59-3.80 (m, 2H), 4.13-4.27 (m, 2H), 5.08-5.22 (m, 2H), 7.26-7.46 (m, 11H), 7.67 (s, 4H), 9.35 (s, 0.5H), 9.48 (s, 0.5H) <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  19.4, 24.9, 26.0, 27.1, 29.8, 30.1, 37.3, 37.8, 56.5, 57.7, 61.6, 61.8, 65.9, 66.3, 67.4, 67.6, 127.9, 128.1, 128.3, 128.8, 129.9, 133.9, 135.8, 136.5, 154.7, 155.9, 200.5. Anal. Calcd for C<sub>31</sub>H<sub>37</sub>NO<sub>4</sub>Si: C, 72.20; H, 7.23; N, 2.72. Found: C, 71.88; H, 7.37; N, 2.61.



**(2S,5R)-Benzyl 2-(2-(*tert*-butyldiphenylsilyloxy)ethyl)-5-(2-oxoethyl)pyrrolidine-1-**

**carboxylate (16).** (Ph<sub>3</sub>PCH<sub>2</sub>OCH<sub>3</sub>)Cl (1.25 g, 3.64 mmol, 1.33 equiv) was suspended in dry THF (50 mL) under nitrogen protection. *t*-BuOK (1.0M in THF, 3.42 mL, 3.42 mmol, 1.25 equiv) was added dropwise the solution. After 10 minutes, a solution of aldehyde **15** (1.412 g, 2.74 mmol, 1.0 equiv) in dry THF (10 mL) was added dropwise. After the addition, the mixture was continued to stir for 2 hours. Then water (50 mL) was added to the mixture. The resulting solution was extracted with Et<sub>2</sub>O (2 × 50 mL). The combined organic layers was dried over MgSO<sub>4</sub> and concentrated under vacuum to afford an oil, which was used directly in the next step. To a stirred solution of the previous oil in acetone (50 mL) at 0 °C, PTSA·H<sub>2</sub>O (260 mg, 1.37 mmol, 0.5 equiv) was added as one portion. The cold bath was removed after the addition. The stirring was continued for additional 30 minutes. Most of the solvent was evaporated and water (50 mL) was added to the resulting mixture. The resulting solution was extracted with CH<sub>2</sub>Cl<sub>2</sub> (2

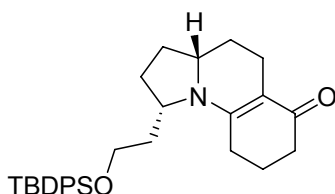
× 50 mL). The combined organic layers was dried over MgSO<sub>4</sub> and concentrated under vacuum. The crude product was purified by flash column chromatography (SiO<sub>2</sub>, 80:20 hexanes/EtOAc) to afford 1.145 g (79% yield, 2 steps) of **16** as a colorless oil.  $[\alpha]_{\text{D}}^{25} +2.44$  (*c* 1.31, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 1.03 (s, 9H), 1.39-2.27 (m, 6H), 2.36-2.45 (m, 1H), 2.78-3.11 (m, 1H), 3.69 (s, 2H), 4.04 (s, 1H), 4.22-4.36 (m, 1H), 5.10 (d, *J* = 6.2, 2H), 7.25-7.44 (m, 11H), 7.63 (s, 4H), 9.71 (d, *J* = 52.8, 1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 19.4, 27.1, 27.7, 28.5, 29.7, 30.4, 36.5, 38.4, 48.1, 50.3, 52.9, 54.2, 55.8, 56.4, 61.8, 67.0, 67.1, 127.2, 127.9, 128.1, 128.7, 129.9, 133.9, 135.8, 136.8, 200.9 Anal. Calcd for C<sub>32</sub>H<sub>39</sub>NO<sub>4</sub>Si: C, 72.55; H, 7.42; N, 2.64. Found: C, 72.55; H, 7.39; N, 2.57.



**(2*S*,5*S*)-Benzyl 2-(2-(*tert*-butyldiphenylsilyloxy)ethyl)-5-(2-(2,6-dioxocyclohexyl)**

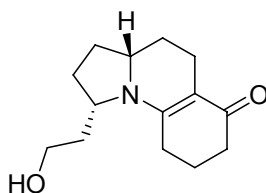
**ethyl)pyrrolidine-1-carboxylate (19).** Aldehyde **16** (462 mg, 0.872 mmol, 1.0 equiv) was dissolved in CH<sub>2</sub>Cl<sub>2</sub> (1.75 mL). Diethyl 2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxylate (221 mg, 0.872 mmol, 1.0 equiv) and cyclohexane-1,3-dione (98 mg, 0.872 mmol, 1.0 equiv) were added subsequently. Then L-proline (20 mg, 0.175 mmol, 0.2 equiv) was added to the mixture. The stirring was continued for 1 hour. The resulting mixture was directly subjected to purify by flash column chromatography (SiO<sub>2</sub>, 50:50 hexanes/EtOAc) to afford 509 mg (93% yield) of **19** as a colorless oil.  $[\alpha]_{\text{D}}^{25} +42.1$  (*c* 0.4, CH<sub>3</sub>OH). <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 0.98 (s, 9H), 1.35-1.97 (m, 11H), 2.15-2.54 (m, 6H), 3.60-3.81 (m, 3H), 4.00-4.09 (m,

1H), 5.15 (d,  $J = 2.0$ , 2H), 7.26-7.45 (m, 11H), 7.60-7.64 (m, 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  14.4, 19.2, 19.4, 21.1, 27.0, 29.7, 30.0, 31.7, 37.0, 37.2, 39.2, 56.9, 58.7, 61.7, 67.8, 114.7, 127.9, 128.2, 128.3, 128.7, 129.9, 133.8, 135.7, 199.4. Anal. Calcd for  $\text{C}_{38}\text{H}_{47}\text{NO}_5\text{Si}\cdot\text{H}_2\text{O}$ : C, 70.88; H, 7.67; N, 2.18. Found: C, 70.98; H, 7.46; N, 2.18.



**(1S,3aS)-1-(2-(*tert*-Butyldiphenylsilyloxy)ethyl)-1,2,3,3a,4,5,8,9-octahydropyrrolo[1,2-a]quinolin-6(7H)-one (18).** Compound **19** (473 mg, 0.756 mmol, 1.0 equiv) was dissolved in methanol (100 mL). 10% Pd/C (245 mg) was added to the solution. The mixture was subjected to hydrogenation with hydrogen balloon at room temperature for 24 hours. The resulting mixture was filtered through celite 545 (5 g) and rinsed with methanol ( $2 \times 50$  mL). All the combined filtrates were concentrated under vacuum. The residue was purified by preparative TLC ( $\text{SiO}_2$ , 95:5 EtOAc/ $\text{CH}_3\text{OH}$ ) to afford 270 mg (75% yield) of **18** as a yellow oil.  $[\alpha]_{\text{D}}^{25} +317$  ( $c$  0.31,  $\text{CH}_3\text{OH}$ ).  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  1.06 (d,  $J = 7.7$ , 9H), 1.13-1.28 (m, 1H), 1.38-2.20 (m, 10H), 2.26-2.43 (m, 3H), 2.60-2.75 (m, 2H), 3.20-3.28 (m, 1H), 3.58-3.78 (m, 2H), 4.03 (t,  $J = 8.6$ , 1H), 7.37-7.47 (m, 6H), 7.65 (dt,  $J = 8.0, 1.6$ , 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  19.4, 20.3, 21.6, 22.1, 27.1, 27.4, 27.6, 27.8, 28.6, 29.1, 29.8, 30.7, 36.3, 36.6, 38.5, 38.9, 55.7, 56.4, 57.3, 59.4, 61.0, 61.4, 107.3, 128.0, 130.1, 133.6, 135.7, 135.8, 158.9, 193.9. Anal. Calcd for  $\text{C}_{30}\text{H}_{39}\text{NO}_2\text{Si}\cdot 0.5\text{H}_2\text{O}$ : C, 74.64; H, 8.35; N, 2.90. Found: C, 74.51; H, 8.28; N, 2.95.





**(1*S*,3*aS*)-1-(2-Hydroxyethyl)-1,2,3,3*a*,4,5,8,9-octahydropyrrolo[1,2-*a*]quinolin-6(7*H*)-one (6).**

Compound **18** (174 mg, 0.367 mmol, 1.0 equiv) was dissolved in THF (5 mL). Tetrabutylammoniumfluoride (1.0M in THF, 0.55 mL, 0.55 mmol, 1.5 equiv) was added dropwise to the solution. The stirring was continued for 3 hours at room temperature. Then saturated Na<sub>2</sub>CO<sub>3</sub> solution (0.5 mL) was added to the reaction mixture. After 10 minutes, Et<sub>2</sub>O (10 mL) was added to the mixture. The resulting mixture was dried over MgSO<sub>4</sub> and filtered. The filtrate was concentrated under vacuum. The residue was purified by preparative TLC (SiO<sub>2</sub>, 92:8 CH<sub>2</sub>Cl<sub>2</sub>/CH<sub>3</sub>OH) to afford 75 mg (87% yield) of **6** as a white solid, mp 178-180 °C (recrystallization from EtOAc/Cyclohexanes, 1:1) {lit.<sup>10</sup>: mp 176-179 °C}. [ $\alpha$ ]<sub>D</sub><sup>25</sup> +798 (*c* 0.29, EtOH) {lit.<sup>10</sup>: [ $\alpha$ ]<sub>D</sub><sup>25</sup> +538 (*c* 1.40, EtOH)}. <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  1.18-1.29 (m, 1H), 1.49-2.20 (m, 11H), 2.32 (t, *J* = 6.5, 2H), 2.40-2.48 (m, 1H), 2.61-2.68 (m, 2H), 3.23-3.31 (m, 1H), 3.61-3.78 (m, 2H), 4.03 (t, *J* = 8.0, 1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  21.4, 21.9, 27.3, 28.4, 29.0, 29.6, 36.3, 38.5, 55.6, 59.3, 60.0, 107.1, 158.9, 193.7.

#### 4.7. References

1. Daly, J. W.; Spande, T. F.; Garraffo, H. M. *J. Nat. Prod.* **2005**, *68*, 1556-1575.

2. Banner, E. J.; Stevens, E. D.; Trudell, M. L. *Tetrahedron Lett.* **2004**, *45*, 4411-4414.
3. Zhang, C.; Trudell, M. L. *J. Org. Chem.* **1996**, *61*, 7189-7191.
4. Zhang, S.; Xu, L.; Miao, L.; Shu, H.; Trudell, M. L. *J. Org. Chem.* **2007**, *72*, 3133-3136.
5. Daly, J. W.; Witkop, B.; Tokuyama, T.; Nishikawa, T.; Karle, I. L. *Helv. Chim. Acta* **1977**, *60*, 1128-1140.
6. Mensah-Dwumah, M.; Daly, J. W. *Toxicon* **1978**, *16*, 189-194.
7. Daly, J. W.; Garraffo, H. M.; Spande, T. F. In *Alkaloids: Chemical and Biological Perspectives*; Pelletier, S. W.; Ed.; Pergamon: New York, 1999; Vol. 13, pp 1-161.
8. For previous total syntheses of racemic gephyrotoxin, see: (a) Fujimoto, R.; Kishi, Y.; Blount, J. F. *J. Am. Chem. Soc.* **1980**, *102*, 7154-7156. (b) Hart, D. J.; Kanai, K. *J. Am. Chem. Soc.* **1983**, *105*, 1255-1263. (c) Overman, L. E.; Lesuisse, D.; Hashimoto, M. *J. Am. Chem. Soc.* **1983**, *105*, 5373-5379.
9. For previous formal syntheses of racemic gephyrotoxin, see: (a) Ito, Y.; Nakajo, E.; Nakatsuka, M.; Saegusa, T. *Tetrahedron Lett.* **1983**, *24*, 2881-2884. (b) Pearson, W. H.; Fang, W.-K. *J. Org. Chem.* **2000**, *65*, 7158-7174.
10. Fujimoto, R.; Kishi, Y. *Tetrahedron Lett.* **1981**, *42*, 4197-4198.
11. Wei, L.-L.; Hsung, R. P.; Sklenicka, H. M.; Gerasyuto, A. I. *Angew. Chem., Int. Ed.* **2001**, *40*, 1516-1518.
12. Santarem, M.; Vanucci-Bacqué, C. Lhomme, G. *J. Org. Chem.* **2008**, *73*, 6466-6469.
13. Zhang, C.; Lomenzo, S. A.; Ballay, C.; Trudell, M. L. *J. Org. Chem.* **1997**, *62*, 7888-7889.
14. Michel, P. Rassat, A.; Daly, J. W.; Spande, T. F. *J. Org. Chem.* **2000**, *65*, 8908-8918.
15. Jamison, T. F.; Moslin, R. M. *J. Org. Chem.* **2007**, *72*, 9736-9745.
16. Ramachary, D. B.; Kishor, M. *J. Org. Chem.* **2007**, *72*, 5056-5068.
17. Hanessian, S.; Lavalley, P. *Can. J. Chem.* **1975**, *53*, 2975-2977.

## **CHAPTER 5**

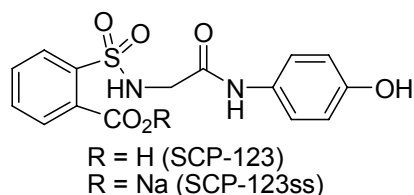
### **First Multi-gram Preparation of SCP-123, A Novel Water Soluble Analgesic**

#### **5.1. Abstract**

A large-scale process for the preparation of the analgesic compounds SCP-123 and its sodium salt, SCP-123ss•monohydrate has been developed. The process for the preparation of SCP-123 required three synthetic steps with no chromatography, while the process for the preparation of SCP-123ss required four synthetic steps and no chromatography. The overall yields for both SCP-123 and SCP-123ss were 47% and 46%, respectively, and both compounds were obtained in exceptionally high purity (>99%).

Keywords: analgesic, acetaminophen, propacetamol, saccharin, hydrolysis, parenteral administration.

**Figure 5.1.** Structure of SCP-123



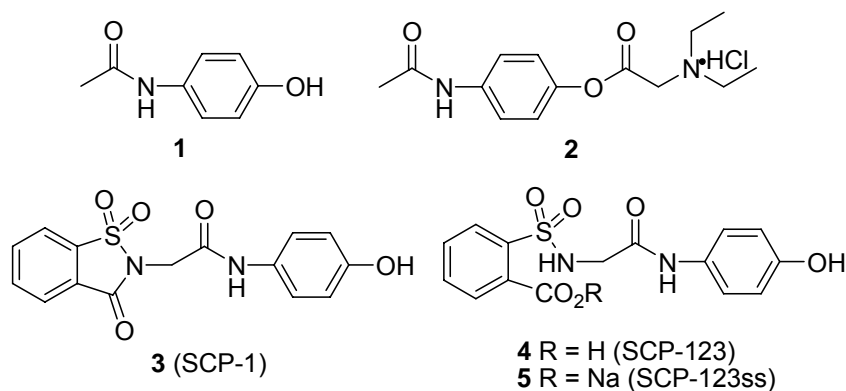
## 5.2. Introduction

The analgesic acetaminophen (**1**) (Figure 5.2) is widely used for the acute and chronic control of pain.<sup>1</sup> While the therapeutic window of **1** is quite broad, it does have some significant limitations. The low water solubility is problematic for some delivery applications, chronic use has significant hepatotoxicity, and acute use with alcohol can be lethal.<sup>2-4</sup> A water-soluble analogue of **1** is the prodrug propacetamol hydrochloride (**2**).<sup>5,6</sup> This form of acetaminophen is rapidly and completely hydrolyzed by plasma esterases to release **1**.<sup>7</sup> The pharmacological effects in clinical trials have shown that **2** possesses similar efficacy to **1**, but due to its greater water-solubility can be parenterally administered and thus can be employed when oral administration is not possible.<sup>5,6</sup> However, since **2** is a prodrug of **1**, it still exhibits a similar pharmacological profile of side effects and toxicity.

The recent discovery that the saccharin derivative of acetaminophen, SCP-1 (**3**) possesses analgesic properties of equal potency to acetaminophen with significantly diminished hepatotoxicity has prompted an extensive investigation into this class of compounds as a new

generation of analgesic drugs.<sup>8-12</sup> The lead compound **3**, was found to possess an analgesic and antipyretic profile similar to **1**.<sup>10-12</sup> However, recent studies with **3** have shown that it is extensively and rapidly hydrolyzed in vivo.<sup>10</sup> The metabolite SCP-123 (**4**) and corresponding sodium salt SCP-123ss (**5**) are equipotent on a molar basis with **3** in analgesic models.<sup>13</sup> Presumably, the efficacy of **3** is derived from the hydrolysis products **4** and **5**. Therefore, it was of interest to develop large-scale syntheses of **4** and **5** for further drug development studies.

**Figure 5.2.** Structure of acetaminophen analogue



## 5.3. Results and discussion

### 5.3.1. SCP-1 and analogues

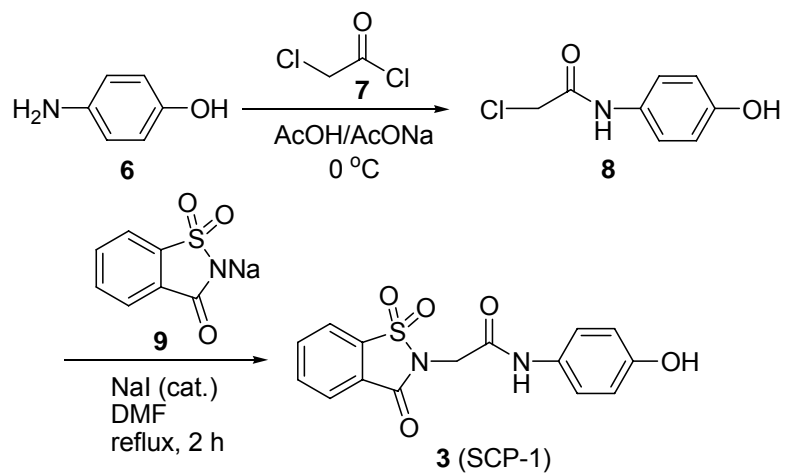
For the proposed initial studies, multigram quantities of **4** and **5** were required. Previous work with these compounds in our laboratories had revealed that the most efficient way to

prepare the gram quantities of these metabolites was via the hydrolysis of the saccharin ring of **3**.<sup>10</sup> Therefore the design of a large-scale synthesis focused on the initial preparation of **3**, followed by the subsequent hydrolysis to afford either **4** or **5**. Two synthetic routes have been established for the preparation of gram quantities of **3**.<sup>8-10</sup> As illustrated in Scheme 5.1, the two routes primarily differ in the sequence in which the saccharin moiety is added to the acetyl unit. In Route A,<sup>9,10</sup> the saccharin moiety is added in the last step to the 2-chloroacetamide intermediate **8** that has been previously generated from 4-aminophenol (**6**) and 2-chloroacetyl chloride (**7**). This yields **3** via a two-step process. Alternatively in Route B,<sup>8</sup> the intermediate acetic acid intermediate **11** is formed initially from sodium saccharin (**9**) and bromoacetic acid (**10**). The intermediate **11** is then coupled to 4-aminophenol to furnish **3** also via a two-step process.

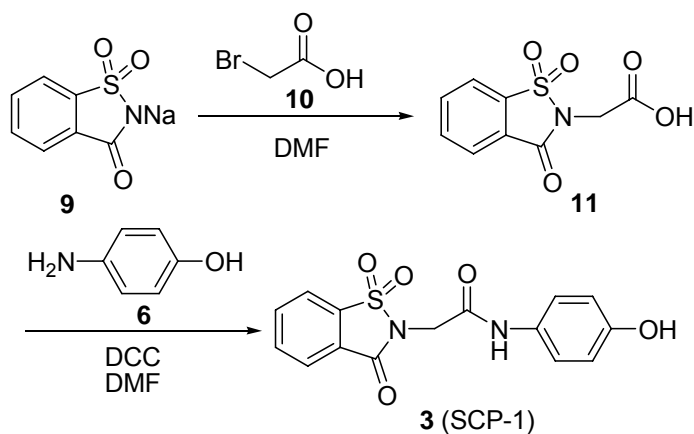
Upon evaluation of both routes on a gram scale synthesis, Route A was deemed to be of greater merit due the low cost of the commercially available starting materials and the ease of purification of the intermediate **8** and SCP-1 (**3**). In Route A, the intermediate **8** precipitated cleanly from the reaction medium and could be obtained in a state of high purity (>95 %). Likewise in Route A, **3** could be obtained in greater than 95% purity by precipitation from the reaction media with ice water. Subsequently, a single recrystallization from ethanol/water routinely afforded **3** in pure form (>99%). Alternatively, Route B seemed to be limited by the hygroscopic intermediate acid **11**, which at times has been difficult to handle. In addition, the purification of **3** derived from Route B was found to require multiple recrystallizations to remove the impurity dicyclohexylurea, that is a by-product of the coupling reaction.

**Scheme 5.1.** Synthetic routes to SCP-1 (**3**)

**Route A**



**Route B**

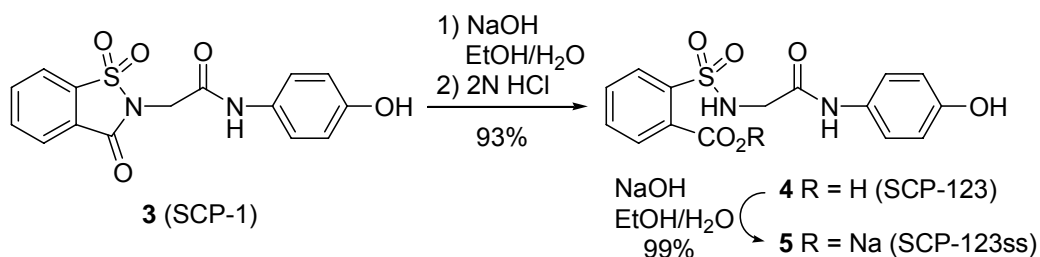


Based upon our evaluation, Route A was scaled 15-fold and run on a mole scale based upon 4-aminophenol (**6**). The 2-chloroacetyl chloride (**7**) was added at a controlled rate to a suspension of **6** in a buffered solution of acetic acid [HOAc: NaOAc (sat.) (1:1, v:v)] such that the reaction temperature did not exceed 5 °C. Mechanical stirring was required to maintain adequate mixing throughout the reaction process. As the addition of the acid chloride progressed,

the suspension dissipated and the reaction mixture became clear. However, prior to the end of the addition of **7**, precipitation of the intermediate 2-chloroacetamide **8** was observed. From this point on, mechanical stirring was essential to obtain consistent and high yields. The 2-chloroacetamide intermediate **8** was obtained in 70% yield and no further purification was required for advancement to the next step. Preparation of **3** was routinely performed on a mole scale. The 2-chloroacetamide **8** and saccharin sodium salt (**9**) were heated to reflux in DMF with a catalytic amount of NaI (0.40 mol %). The saccharin derivative was then easily obtained by precipitation in ice water. A single recrystallization from ethanol/water furnished **3** in 72% yield.

As illustrated in Scheme 5.2, the hydrolysis of **3** was readily achieved with NaOH solution followed by concomitant treatment with 2N hydrochloric acid.<sup>10,14</sup> This afforded the corresponding acid **4** (SCP-123) in 93% yield.

**Scheme 5.2.** Synthesis of SCP-123 (**4**) and SCP-123ss (**5**)

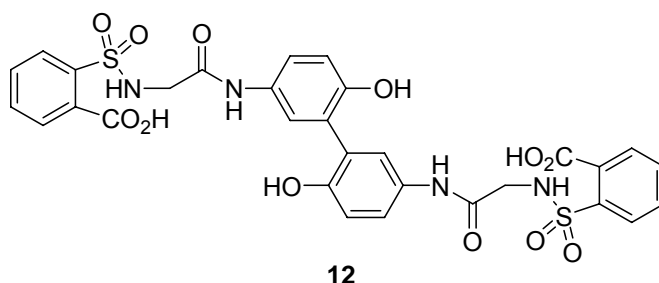


The hydrolysis reaction was not as easily scaled-up as the previous steps. This step in the sequence was routinely performed on a 50-gram scale. This limitation was due primarily to the



sensitivity of the hydrolysis step to the concentration of the saccharin derivative **3** in the basic solution. If the reaction mixture was not sufficiently dilute the formation of a side-product **12**, that resulted from oxidative phenolic coupling, was obtained.<sup>15,16</sup>

**Figure 5.3.** Structure of side-product **12**



This impurity **12** was present in varying amounts, ranging from 10-25%, depending upon the concentration of the reaction mixture relative to **3**. The coupling product **12** was difficult to detect by NMR and could only be identified and separated from the product **4** by HPLC. An optimized concentration of **3** in 0.5N NaOH was determined to be 0.25 M. At this concentration, the hydrolysis of **3** proceeded cleanly and the oxidative-coupling product **12** was not observed. These conditions were preferred to using tedious degassing procedures and performing the reaction under anaerobic conditions. Due to the large reaction volumes at this concentration we were limited by our equipment and thus typically performed the hydrolysis on a 50-gram scale. The resultant hydrolysis product could be manipulated easily by precipitation with acid to give **4** in > 99% purity. Despite the smaller scale of the hydrolysis reaction, this step was typically

executed in multiple simultaneous batches that could be combined to rapidly generate sub-kilogram quantities of **4**. However, we have no evidence to suggest that this reaction is limited to this scale and could not be performed on a larger scale if needed.

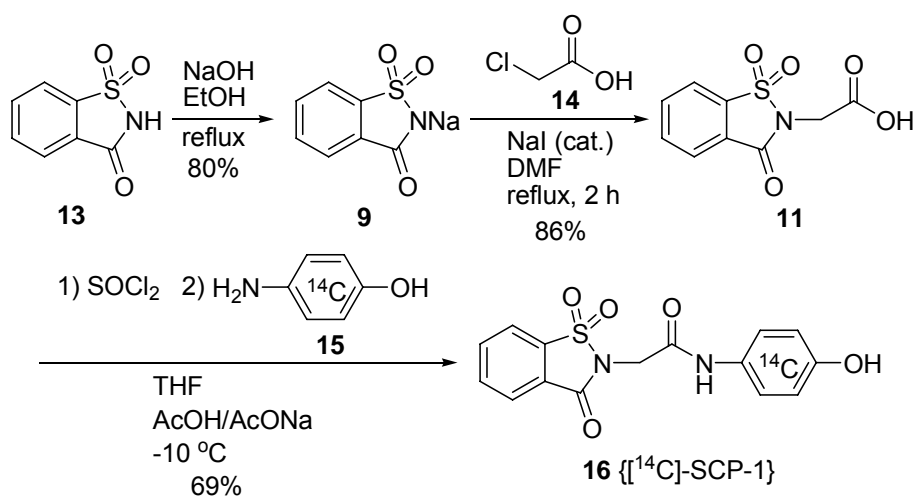
The preparation of the sodium salt **5** was achieved by titration of the acid **4** with one equivalent of sodium hydroxide (Scheme 5.2). The advantage of this procedure over the direct conversion of **3** into **5** was that the direct method gave an unquantifiable mixture of mono- and di-sodium salts due to the acidic phenol moiety. Alternatively, the titration of **4** with one equivalent of NaOH afforded the sodium carboxylate **5**, which could be precipitated cleanly out solution as the mono-sodium salt. Filtration and vacuum drying gave **5** as the monohydrate (**5•H<sub>2</sub>O**) in quantitative yield and exceptionally high purity (>99%) as determined by HPLC and combustion analysis. The monohydrate **5•H<sub>2</sub>O**, albeit somewhat hygroscopic, was stable to extensive drying and gave consistent combustion analysis when stored in dry environment.

### 5.3.2. Synthesis of radiolabelled SCP-123 [<sup>14</sup>C]-SCP-123}

Because the price for radioactive 4-aminophenol (**15**) is so expensive, Route A (Scheme 5.1) is not suitable for the synthesis of [<sup>14</sup>C]-SCP-1. On the other hand, Route B (Scheme 5.1) employed the *N,N'*-dicyclohexylcarbodiimide (DCC) as a coupling reagent which generated a complicated work-up procedure and ended with a lower yield.<sup>8,17</sup> Our new synthesis route started with commercial available saccharin (**13**). As shown in Scheme 5.3, we need to generate the intermediate 2-saccharin acetyl chloride using thionyl chloride. More dry the materials, the better

the yield of the reactions. In order to obtain anhydrous saccharin sodium salt (**9**) without any hydrate, saccharin (**13**) was treated with NaOH in reflux EtOH to afford saccharin sodium salt (**9**) precipitating out the solution at room temperature.<sup>1</sup> Saccharin sodium salt (**9**) was coupled with 2-chloroacetic acid (**14**) with catalytic amount of NaI in reflux DMF to form 2-saccharin acetic acid (**11**) in 86% yield.

**Scheme 5.3.** Synthesis of [<sup>14</sup>C]-SCP-1

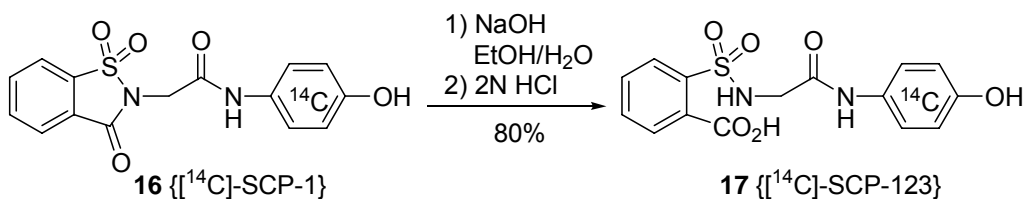


Compound **11** was treated with thionyl chloride under reflux for 2 hours to afford the intermediate 2-saccharin acetyl chloride which was used in the next step without further purification. The intermediate was dissolved in dry THF and dropwised to a precooled buffer solution of saturated NaOAc, HOAc and 4-amino-[<sup>14</sup>C(U)]phenol (1 mCi) at -10 °C over a period of 15 minutes. The amidation reaction went to complete within 30 minutes to furnish

[ $^{14}\text{C}$ ]-SCP-1 (**16**) in 69% overall yield. Higher yield come from the slow addition of the acid chloride solution and 1.35 equivalents of the acid **11** relative to the 4-aminophenol. The reaction mixture was directly poured into ice water to form white precipitate which was filtered and dried under vacuum to afford NMR pure (> 95%) product. The whole procedure required no chromatography or recrystallizations.

The synthesis of the metabolite [ $^{14}\text{C}$ ]-SCP-123 (**17**) was achieved by treatment of [ $^{14}\text{C}$ ]-SCP-1 (**16**) with 0.5N NaOH aqueous solution (Scheme 5.4). The hydrolysis reaction went to complete within 1 hour ending the ring open product [ $^{14}\text{C}$ ]-SCP-123 (**17**) in 80% yield. NMR pure (> 95%) product was obtained by adding 2N HCl to the reaction mixture followed by general work-up and recrystallization.

**Scheme 5.4.** Synthesis of [ $^{14}\text{C}$ ]-SCP-123

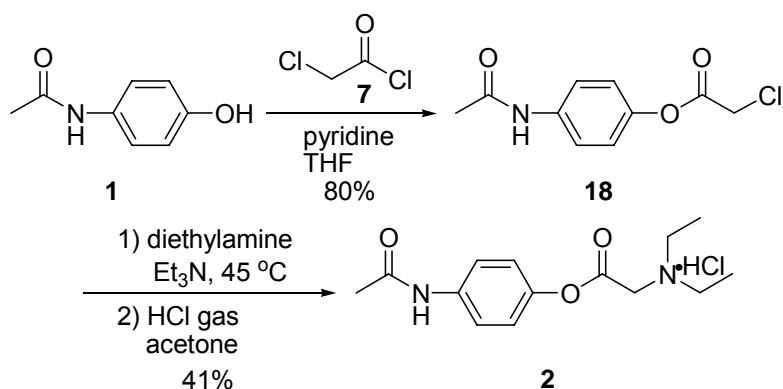


### 5.3.3. Propacetamol hydrochloride

In addition to the SCP compounds, propacetamol hydrochloride was synthesized to be used as standard in the biological evaluations (Scheme 5.5).<sup>18,19</sup> Acetaminophen (**1**) was treated

with chloroacetyl chloride (**7**) in THF in the presence of pyridine to furnish *p*-acetamidophenyl chloroacetate (**18**) with 80% yield.

**Scheme 5.5.** Synthesis of propacetamol hydrochloride



Compound **18** was coupled to diethylamine in the solvent of  $\text{Et}_3\text{N}$  to form an intermediate *p*-acetamidophenyl diethylamino acetate. When HCl gas was bubbled through the solution of the intermediate in dry acetone, white crystals precipitated out the solution to afford propacetamol hydrochloride (**2**) with 41% yield.

#### 5.4. Conclusion

In conclusion, we have developed a large-scale process for the preparation of **4** (SCP-123) and its sodium salt, **5**• $\text{H}_2\text{O}$  (SCP-123ss• $\text{H}_2\text{O}$ ). The overall yields for both **4** and **5**• $\text{H}_2\text{O}$  were 47% and 46% respectively. The process for the preparation of **4** required three synthetic steps

with no chromatography, while the process for the preparation of **5•H<sub>2</sub>O** required four synthetic steps and no chromatography. In both processes, the desired compounds **4** and **5•H<sub>2</sub>O** were isolated in high purity (> 99%) as determined by HPLC and combustion analysis.

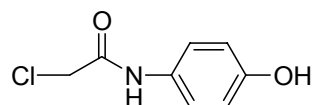
### 5.5. Acknowledgment

We are grateful to the National Institute on Neurological Disorders and Stroke for the support of this research. The project described was supported by Grant Number U44NS046891 from the National Institute of Neurological Disorders And Stroke. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institute of Neurological Disorders And Stroke or the National Institutes of Health.

### 5.6. Experimental section

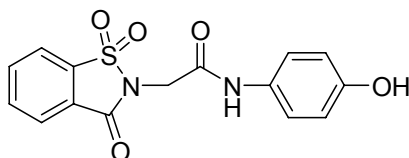
General Methods. All chemicals were purchased from Aldrich Chemical Company and used as received unless otherwise noted. All reactions were performed in glass reactors. Proton and carbon NMR were recorded on a Varian-400 MHz nuclear magnetic resonance spectrometer at ambient temperature in deuterated dimethylsulfoxide (DMSO-*d*<sub>6</sub>) from Cambridge Isotope Laboratories, Inc. <sup>1</sup>H NMR and <sup>13</sup>C NMR chemical shifts are reported as δ values (ppm) relative to tetramethylsilane. Melting points (mp) were measured with an Electrothermal ® Mel-Temp apparatus and are uncorrected. HPLC was used to monitor the purity of all

intermediates using standard HPLC equipment with PDA detection and data system. Separations were performed with a Waters Nova-Pak C18 (3.9 × 150 mm) steel analytical column. The mobile phases for isocratic and gradient separations were prepared using 0.01% TFA in water and 0.01% TFA in CH<sub>3</sub>CN. All the compounds were monitored against the reference materials including the starting materials. Combustion analysis (C, H, N) was performed by Atlantic Microlabs Inc., Norcross, GA.

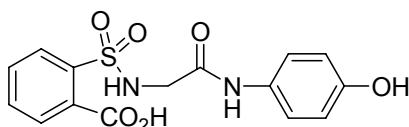


**N-(4-Hydroxyphenyl)-2-chloroacetamide (8).** The 4-aminophenol (**6**, 150 g, 1.37 mol,) was added to a saturated solution of sodium acetate (500 mL) in a three-neck 2 L-round-bottom flask fitted with a thermometer, addition funnel and mechanical stirrer. Acetic acid (500 mL) was added to the mixture and the suspension was cooled in an ice bath. The 2-chloroacetyl chloride (**7**, 155 g, 109 mL, 1.37 mol) was added portion wise to the suspension such that the reaction temperature did not exceed 5 °C. As the addition of **7** progressed the suspension dissipated and the mixture clarified. Prior to completion of the addition of **7**, a white precipitate began to form. Upon completion of the addition, the heterogenous the mixture was stirred at room temperature for 2 hours. The white precipitate was filtered, washed with distilled water solution (2 × 100 mL) and dried under vacuum to afford 177 g of **8** as a white solid (70% yield), mp 142-144 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 4.17 (s, 2H), 6.70 (d, *J* = 8.8, 2H), 7.35 (d, *J* = 8.8, 2H), 9.26 (s, 1H), 10.02 (s, 1H). <sup>13</sup>C NMR (DMSO) δ 44.2, 115.9, 121.9, 130.7, 154.5, 164.6. Anal. Calcd. for

C<sub>8</sub>H<sub>8</sub>ClNO<sub>2</sub>: C, 51.77; H, 4.34; N, 7.55. Found: C, 51.87; H, 4.31; N, 7.49.



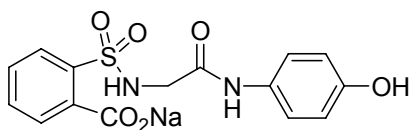
**SCP-1 (3).** The 2-chloroacetamide (**8**, 326 g, 1.75 mol) and saccharin sodium salt hydrate **9** (433 g, 2.10 mol, purchased from Acros Organics) were mixed together in the presence of NaI (1.0 g, 0.0067 mol, 0.4 mol %) in DMF (1 L). The mixture was heated to reflux for 2 hours, cooled and poured into ice water (500 mL). A white precipitate formed and more ice was added until no additional precipitate formed. The sticky white precipitate was collected by vacuum filtration and allowed to dry in air for 30 minutes. The filter cake was dissolved in 50% ethanol-water (2000 mL) and recrystallized to furnish 419 g of **3** as white crystals (72% yield), mp 204-207 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 4.54 (s, 2H), 6.73 (d, *J* = 8.8, 2H), 7.36 (d, *J* = 8.8, 2H), 8.00 (dt, *J* = 6.8, 14.3, 2H), 8.11 (d, *J* = 7.5, 1H), 8.30 (d, *J* = 7.6, 1H), 9.26 (s, 1H), 10.07 (s, 1H). <sup>13</sup>C NMR (DMSO) δ 41.2, 115.9, 121.8, 122.3, 125.8, 127.2, 130.8, 135.9, 136.5, 137.6, 154.4, 159.4, 163.3. Anal. Calcd. for C<sub>15</sub>H<sub>12</sub>N<sub>2</sub>O<sub>5</sub>S: C, 54.21; H, 3.64; N, 8.43. Found: C, 54.15; H, 3.58; N, 8.41.



**SCP-123 (4).** A suspension of SCP-1 (**3**, 50 g, 0.15 mol) and aqueous 0.5N NaOH (600 mL, 0.30

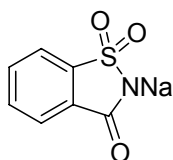


mol) was stirred at room temperature for 1 hour. Ethanol (400 mL) was added to the mixture until the solution become clear. Stirring was continued for an additional 1 hour. The solution was acidified with 2N HCl (500 mL) solution to a pH of 1 (pH meter). The white precipitate that formed was filtered and washed with distilled water (100 mL). The filter cake was dried under vacuum to afford 49 g of **4** as a white solid (93%, yield), mp 184-186 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 3.72 (d, *J* = 5.1, 2H), 6.65 (d, *J* = 8.8, 2H), 7.20 (d, *J* = 8.8, 2H), 7.39 (s, 1H), 7.65-7.77 (m, 3H), 7.93 (m, 1H), 9.21 (s, 1H), 9.71 (s, 1H), 13.81 (b, 1H). <sup>13</sup>C NMR (DMSO) δ 46.5, 115.8, 121.7, 129.3, 130.4, 130.7, 131.7, 133.2, 133.4, 138.2, 154.2, 166.0, 169.5. Anal. Calcd. for C<sub>15</sub>H<sub>14</sub>N<sub>2</sub>O<sub>6</sub>S: C, 51.42; H, 4.03; N, 8.00. Found: C, 51.42; H, 4.15; N, 7.86.

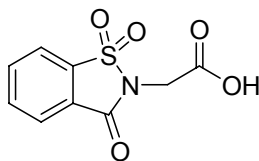


**SCP-123ss·H<sub>2</sub>O (5·H<sub>2</sub>O).** The acid SCP-123 (**4**, 71.5 g, 0.24 mol) was suspended in ethanol (400 mL) and cooled in an ice bath. A pre-cooled (0 °C) solution of NaOH (8.2 g, 0.24 mol) in distilled water (40 mL) was added drop wise to the ethanolic suspension. After the addition of the basic solution was complete more ethanol was added to the mixture, if needed, to dissolve all the solids. The clear reaction mixture was stirred for an additional 2 hours. The reaction mixture was then evaporated by 10% (~50 mL) on a rotoevaporator without a water bath. Once a precipitate started to form, the mixture was removed from the rotoevaporator and cooled in an ice bath for 1 hour. The white precipitate was filtered and washed with distilled water (100 mL). The filter cake was dried under vacuum at 60 °C to afford 93 g of **5·H<sub>2</sub>O** as white solid (99%

yield). mp 188-190 °C.  $^1\text{H}$  NMR (400 MHz, DMSO)  $\delta$  3.57 (s, 2H), 6.65 (d,  $J$  = 8.8, 2H), 7.24 (d,  $J$  = 8.8, 2H), 7.39 (t,  $J$  = 7.0, 1H), 7.51 (t,  $J$  = 7.5, 1H), 7.63 (d,  $J$  = 6.6, 1H), 7.74 (d,  $J$  = 7.7, 1H), 9.00 (s, 1H), 9.58 (s, 1H), 10.03 (s, 1H).  $^{13}\text{C}$  NMR (DMSO)  $\delta$  47.1, 115.7, 121.7, 128.0, 128.1, 130.7, 130.8, 132.9, 136.3, 142.5, 154.3, 166.4, 171.3. Anal. Calcd. for  $\text{C}_{15}\text{H}_{13}\text{N}_2\text{NaO}_6\text{S}\cdot\text{H}_2\text{O}$ : C, 46.15; H, 3.87; N, 7.18. Found: C, 46.01; H, 3.89; N, 7.14.

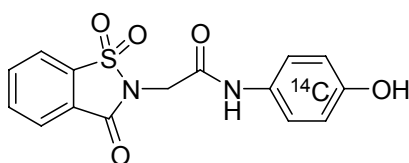


**Saccharin sodium salt (9).** The saccharin **13** (20.0 g, 109.2 mmol, 1.0 equiv) was dissolved in refluxing ethanol (120 mL). Another solution of sodium hydroxide (4.367 g, 109.2 mmol, 1.0 equiv) in hot ethanol (50 mL) was added dropwise. The mixture was refluxed for 10 minutes, cooled to room temperature. And the precipitate was filtered and dried under vacuum to afford 17.86 g (80% yield) of saccharin sodium salt (**9**) as a white solid, mp 355-357 °C.  $^1\text{H}$  NMR (400 MHz, DMSO)  $\delta$  7.54-7.67 (m, 4H).  $^{13}\text{C}$  NMR (DMSO)  $\delta$  119.8, 123.2, 131.7, 132.3, 135.4, 146.0, 168.7. Anal. Calcd. for  $\text{C}_7\text{H}_4\text{NNaO}_3\text{S}$ : C, 40.98; H, 1.97; N, 6.83. Found: C, 41.05; H, 1.90; N, 6.76.



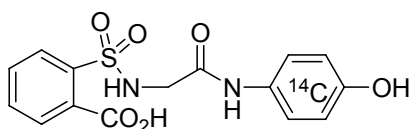
**2-[2,3-Dihydro-3-oxo-1,2-benzisothiazol-2-yl-1,1-dioxide]-acetic acid (11).** 2-chloroacetic

acid (**14**) (4.725 g, 50 mmol, 1.0 equiv) and saccharin sodium salt (**9**) (16.46 g, 80 mmol, 1.6 equiv) were mixed together in the presence of NaI (20 mg) in DMF (64 mL). The mixture was heated to reflux for 2 hours. The reaction was allowed to cool to room temperature, the precipitate was filtered and the filtrate was evaporated under reduced pressure. The residue was dissolved in EtOAc (200 mL), washed with brine (2 × 150 mL) and dried with Na<sub>2</sub>SO<sub>4</sub>. The solvent was removed under reduced pressure and the residue was recrystallized from EtOAc to afford 10.31 g (86% yield) of **11** as a white solid, mp 208-210 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 4.47 (s, 2H), 7.98-8.14 (m, 3H), 8.33 (d, *J* = 7.6, 1H), 13.35 (s, 1H). <sup>13</sup>C NMR (DMSO) δ 39.7, 122.5, 125.9, 126.7, 136.1, 136.8, 137.7, 159.2, 168.2. Anal. Calcd. for C<sub>9</sub>H<sub>7</sub>NO<sub>5</sub>S: C, 44.81; H, 2.92; N, 5.81. Found: C, 45.05; H, 2.88; N, 5.82.



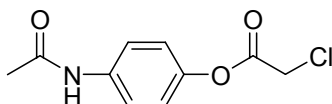
[<sup>14</sup>C]-SCP-1 (**16**). A mixture of acetic acid (**11**) (358 mg, 1.48 mmol) and thionyl chloride (6 mL) was heated at reflux (external oil bath temperature of 85 °C) for 1.5 hours. The excess SOCl<sub>2</sub> was removed under reduced pressure and the resulting acid chloride (1.48 mmol) was used without further purification in the subsequent reaction. The acid chloride (*ca* 1.48 mmol) was dissolved in dry THF (3 mL) and added dropwise over a period of 15 minutes to a mixture of 4-aminophenol (119 mg, 1.09 mmol), 4-amino-[<sup>14</sup>C(U)]-phenol (2 mL, 1 mCi/mL, 77 mCi/mmol, in 0.01N HCl), NaOAc•3H<sub>2</sub>O (644 mg), HOAc (2.25 mL) and saturated NaOAc solution (0.9

mL) at -10 °C. After the addition, the reaction mixture was stirred at -10 °C for 1 hour and poured into icewater (65 g). The precipitate was collected by vacuum filtration and dried under vacuum overnight to furnish 249 mg (69% yield, specific activity: 1.09 mCi/mol) of [ $^{14}\text{C}$ ]-SCP-1 (**16**) as a white solid.  $^1\text{H}$  NMR (400 MHz, DMSO)  $\delta$  4.50 (s, 2H), 6.69 (d,  $J$  = 8.9, 2H), 7.32 (d,  $J$  = 8.9, 2H), 8.00-8.15 (m, 3H), 8.33 (d,  $J$  = 7.4, 1H), 9.33 (s, 1H), 10.07 (s, 1H).  $^{13}\text{C}$  NMR (DMSO)  $\delta$  41.2, 115.8, 121.7, 122.4, 125.8, 127.1, 130.7, 136.0, 136.6, 137.6, 154.4, 159.4, 163.2.

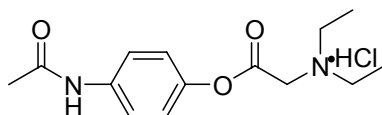


[ $^{14}\text{C}$ ]-SCP-123 (**17**). A mixture of [ $^{14}\text{C}$ ]-SCP-1 (**16**) (241 mg, 0.725 mmol) and aqueous 0.5N NaOH (2.9 mL) was stirred at room temperature for 1 hour. More ethanol was added to the mixture until the solution become clear. Keep stirring for an additional 1 hour. The solution was acidified with 2N HCl solution to PH = 1. Ethanol was removed as less as possible under reduced pressure. The resulting aqueous solution was extracted with EtOAc (2  $\times$  15 mL). The combined extracts were washed with brine and dried over  $\text{MgSO}_4$ . The solvent was removed under reduced pressure and the residue was triturated with hexanes. The resulting solid was recrystallized from ethyl acetate to afford 204 mg (80% yield, specific activity: 0.99 mCi/mol) of [ $^{14}\text{C}$ ]-SCP-123 (**17**) as a white solid.  $^1\text{H}$  NMR (400 MHz, DMSO)  $\delta$  3.70 (d,  $J$  = 5.0, 2H), 6.63 (d,  $J$  = 8.8, 2H), 7.19 (d,  $J$  = 8.9, 2H), 7.38 (s, 1H), 7.64-7.75 (m, 3H), 7.91 (m, 1H), 9.18 (s, 1H), 9.69 (s, 1H), 13.74 (b, 1H).  $^{13}\text{C}$  NMR (DMSO)  $\delta$  46.5, 115.7, 121.6, 129.3, 130.4, 130.6, 131.6,

133.4, 138.2, 154.2, 165.9, 169.4.



***p*-Acetamidophenyl chloroacetate (18).** Chloroacetyl chloride (0.2 mol, 22.6 g, 16 mL, 1.0 equiv) was added to the solution of acetaminophen (**1**) (0.2 mol, 30.2 g, 1.0 equiv) in THF (120 mL) in the presence of pyridine (0.2 mol, 15.82 g, 26.2 mL, 1.0 equiv). The mixture was kept stirring at room temperature for 2 hours, filtered, washed with water (50 mL). Recrystallization in ethanol afforded 36.2 g (80% yield) of *p*-acetamidophenyl chloroacetate (**18**) as white crystals, mp 187-189 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 2.03 (s, 3H), 4.65 (s, 2H), 7.08 (d, *J* = 8.8, 2H), 7.60 (d, *J* = 8.8, 2H), 10.02 (s, 1H). Anal. Calcd. for C<sub>10</sub>H<sub>10</sub>ClNO<sub>3</sub>: C, 52.76; H, 4.43; N, 6.15. Found: C, 52.93; H, 4.45; N, 6.17.



**Propacetamol hydrochloride (2).** *p*-Acetamidophenyl chloroacetate (**38**) (0.03 mol, 7.6 g, 1.0 equiv) was added in small portions to a solution of diethylamine (0.034 mol, 2.5 g, 3.54 mL, 1.13 equiv) in triethylamine (10 mL) with stirring, while the temperature was maintained at 45 °C. The mixture was stirred for an additional 2 hours at 45 °C, cooled, poured into iced water (20 mL), then extracted with ether (3 × 25 mL), dried over MgSO<sub>4</sub> overnight and the ether was evaporated under vacuum to yield *p*-acetamidophenyl diethylamino acetate as an intermediate in the form of a thick oil. Hydrogenchloride gas was bubbled into the solution of the thick oil in dry

acetone (50 mL) at PH =1 to give white precipitate, washed with acetone and dried under vacuum to afford 4.1 g (41% yield) of propacetamol hydrochloride (**2**) as white solid mp 210-212 °C. <sup>1</sup>H NMR (400 MHz, DMSO) δ 1.25 (t, *J* = 7.2, 6H), 2.04 (s, 3H), 3.26 (d, *J* = 6.8, 4H), 4.44 (s, 2H), 7.15 (d, *J* = 8.9, 2H), 7.66 (d, *J* = 8.9, 2H), 10.25 (s, 1H), 10.51 (b, 1H). Anal. Calcd. for C<sub>14</sub>H<sub>21</sub>ClN<sub>2</sub>O<sub>3</sub>: C, 55.90; H, 7.04; N, 9.31. Found: C, 55.63; H, 7.16; N, 9.06.

## 5.7. References

1. Prescott, L. F. *Am. J. Ther.* **2000**, 7, 143-147.
2. Watkins, P. B.; Kaplowitz, N.; Slattery, J. T.; Colonese, C. R.; Colucci, S. V.; Stewart, P. W.; Harris, S. C. *J. Amer. Med. Assoc.* **2006**, 296, 87-93.
3. Slattery, J. T.; Nelson, S. D.; Thummel, K. E. *Clin. Pharmacol. Ther.* **1996**, 60, 241-246.
4. McGoldrick, M. D.; Bailie, G. R. *Ann. Pharmacother.* **1997**, 31, 221-227.
5. Depré, M.; Van Hecken, A.; Verbesselt, R.; Tjandra-Maga, T. B.; Gerin, M.; Schepper P. J.; *Fund. Clin. Pharmacol.* **1992**, 6, 259-262.
6. Flouvat, B.; Levene, A.; Fitoussi, S.; Delhotel-Landes, B.; Gendron, A.; *Int. J. Clin. Pharmacol. Ther.* **2004**, 42, 505-507.
7. Barcia, E.; Martin, A.; Azuara, M. L.; Negro, S. *Chem. Pharm Bull.* **2005**, 53, 277-280.
8. Bazan, N. G.; Alvarez-Builla, J., U.S. Patent 5,554,636, 1996. *Chem. Abstr.* **1996**, 125, 266037.
9. Bazan, N. G.; Alvarez-Builla, J., U.S. Patent 5,621,110, 1997. *Chem. Abstr.* **1996**, 125, 266037.
10. Vaccarino, A. L.; Paul, D.; Mukherjee, P. K.; Rodríguez de Turco, E. B.; Marcheselli, V. L.; Xu, L.; Trudell, M. L.; Minguez, J. M.; Matía, M. P.; Sunkel, C.; Alvarez-Builla, J.; Bazan,

- N. G. *Bioorg. Med. Chem.* **2007**, *15*, 2206-2215.
11. Cui, J.-G.; Zhang, X.; Zhao, Y. H.; Chen, C.; Bazan, N. *Biochem. Biophys. Res. Commun.* **2006**, *350*, 358-363. Erratum in: *Biochem. Biophys. Res. Commun.* **2007**, *359*, 187-188.
  12. Bazan, N. G.; Cui, J.-G., WIPO patent application WO 2008/021896 A2. *Chem. Abstr.* **2008**, *148*, 276766.
  13. Narducy, K. W. Unpublished results. St. Charles Pharmaceuticals.
  14. Xu, L.; Trudell, M. L. *J. Label. Compd. Radiopharm.* **2005**, *48*, 219-222.
  15. Weiss, A. C.; Kuhnle, J. A.; Windle, J. J.; Wierseman, A. K. *Tetrahedron Lett.* **1966**, *50*, 6251-6255.
  16. Whiting, D. A. in *Comprehensive Organic Synthesis*, Trost, B. M.; Fleming, I.; Pattenden, G. (Eds.) Pergamon Press: Oxford, 1991, Vol. 3, pp 659-700 and references cited therein.
  17. González-Martin, G.; Lyndon, C.; Sunkel, C. *Eur. J. Pharm. Biopharm.* **1998**, *46*, 293-297.
  18. Li, T.; Chen, Y.; Li, Y. *Lanzhou Daxue Xuebao* **1990**, *26*, 165-166.
  19. Cognacq; Jean-Claude, U.S. Patent 4,127,671, 1978.

## APPENDIX

### CHAPTER 3

HNMR of (-)-**1**

HNMR of (+)-**1**

Expanded HNMR of (-)-**1** + (+)-**1** + BNPPA (1 equiv)

HNMR of (-)-**1** + BNPPA (1 equiv)

HNMR of (+)-**1** + BNPPA (1 equiv)

### CHAPTER 4

X-ray Crystallographic Data, Positional Parameters, General Displacement, Parameter Expressions, Bond Distances, and Bond Angles for (1*S*,3*aS*)-1-(2-hydroxyethyl)-1,2,3,3*a*,4,5,8,9-octahydropyrrolo[1,2-*a*]quinolin-6(7*H*)-one (**6**)-Kishi's intermediate

### CHAPTER 5

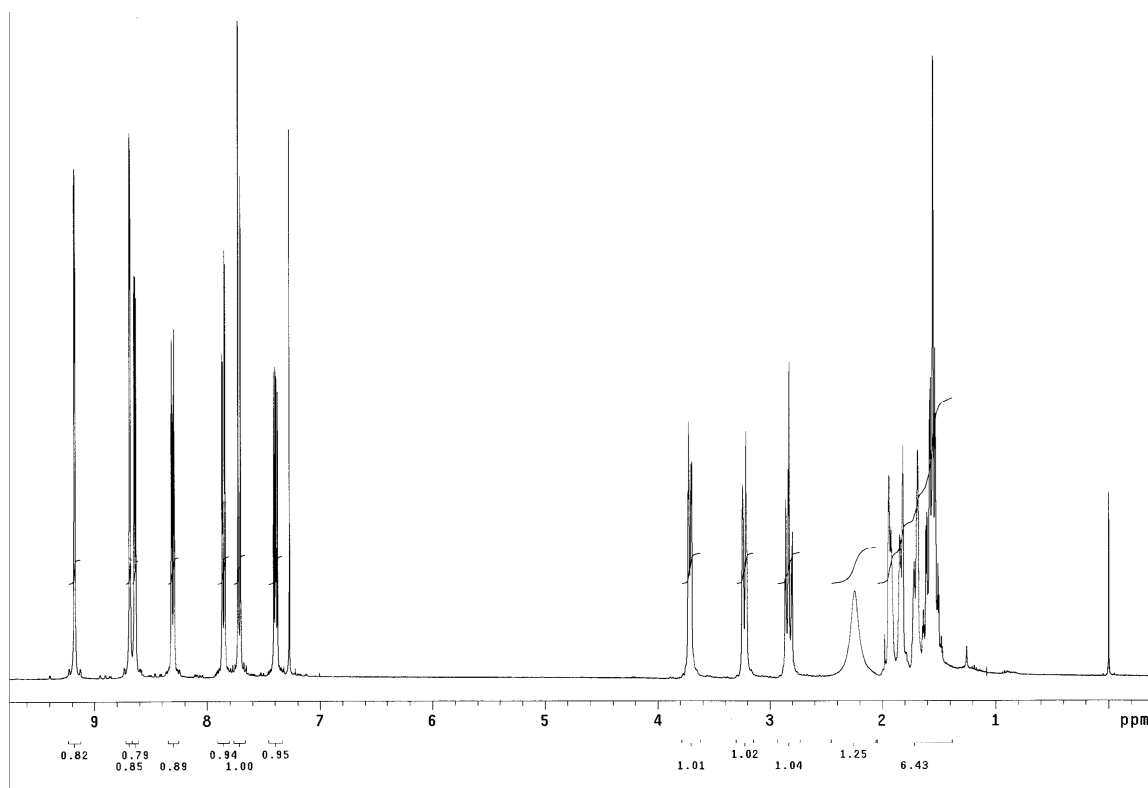
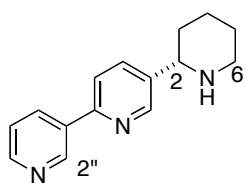
<sup>1</sup>H NMR **4**, **5** and **12**

HPLC Conditions

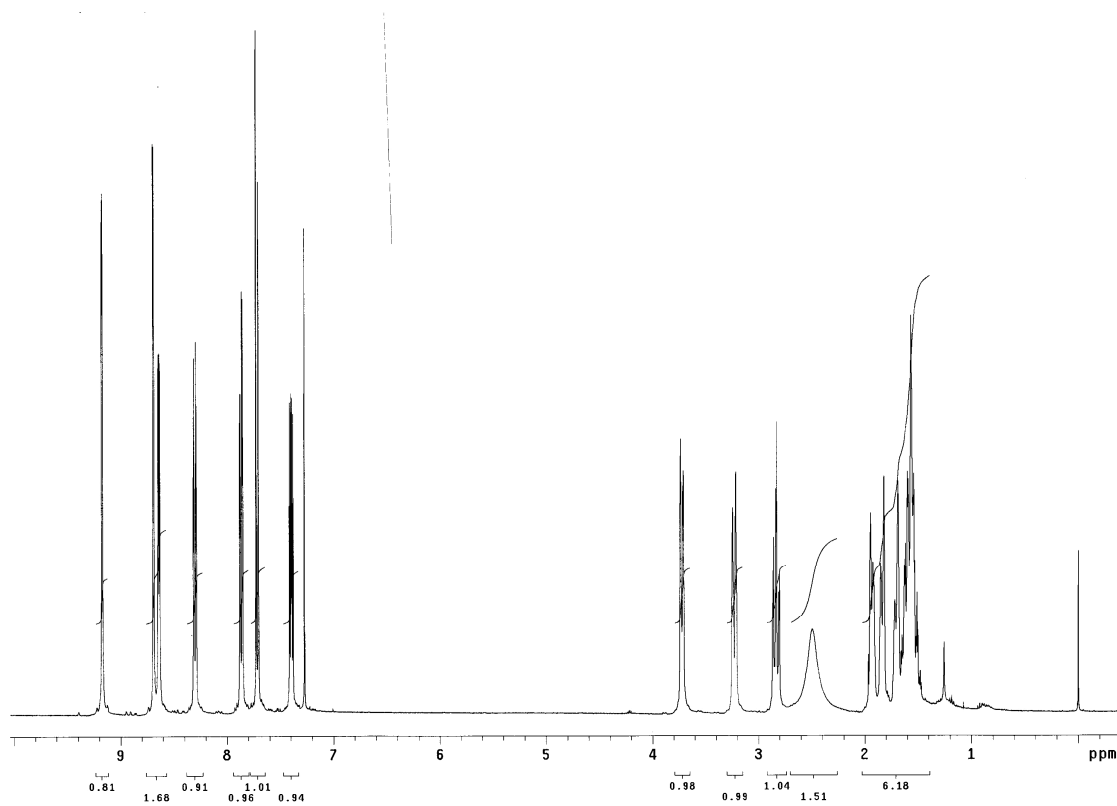
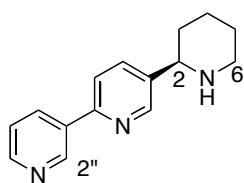
LC-ESI-MS spectrum of **12**



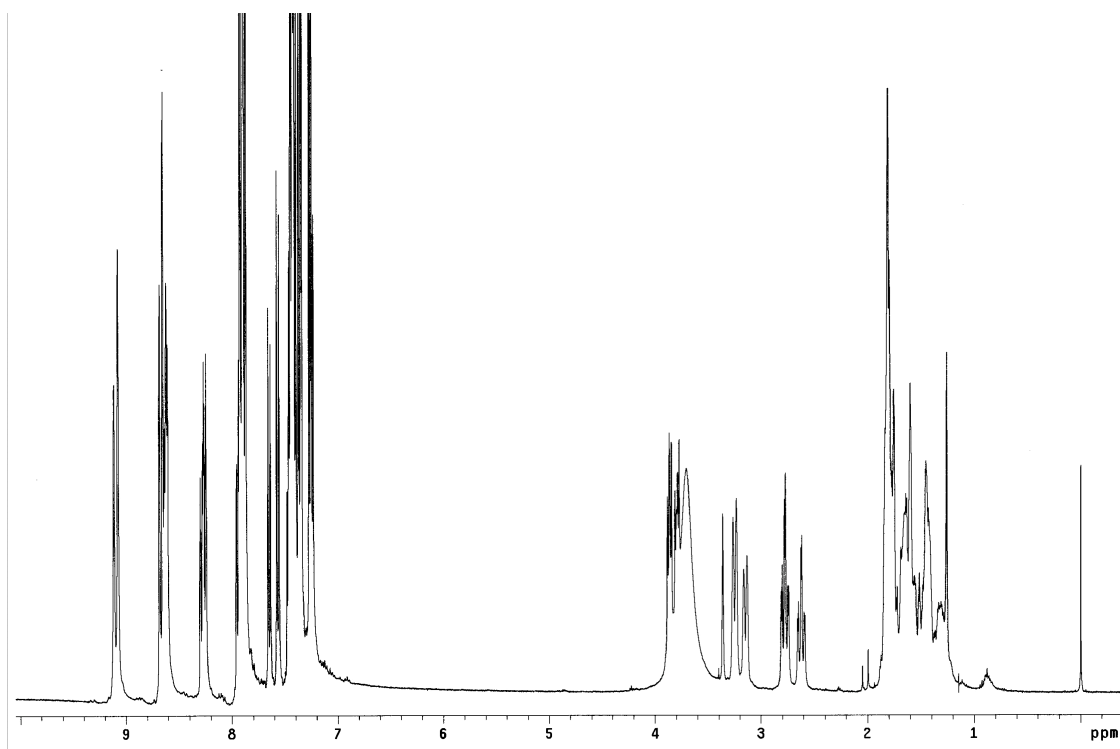
# **HNMR of (-)-1**



**HNMR of (+)-1**



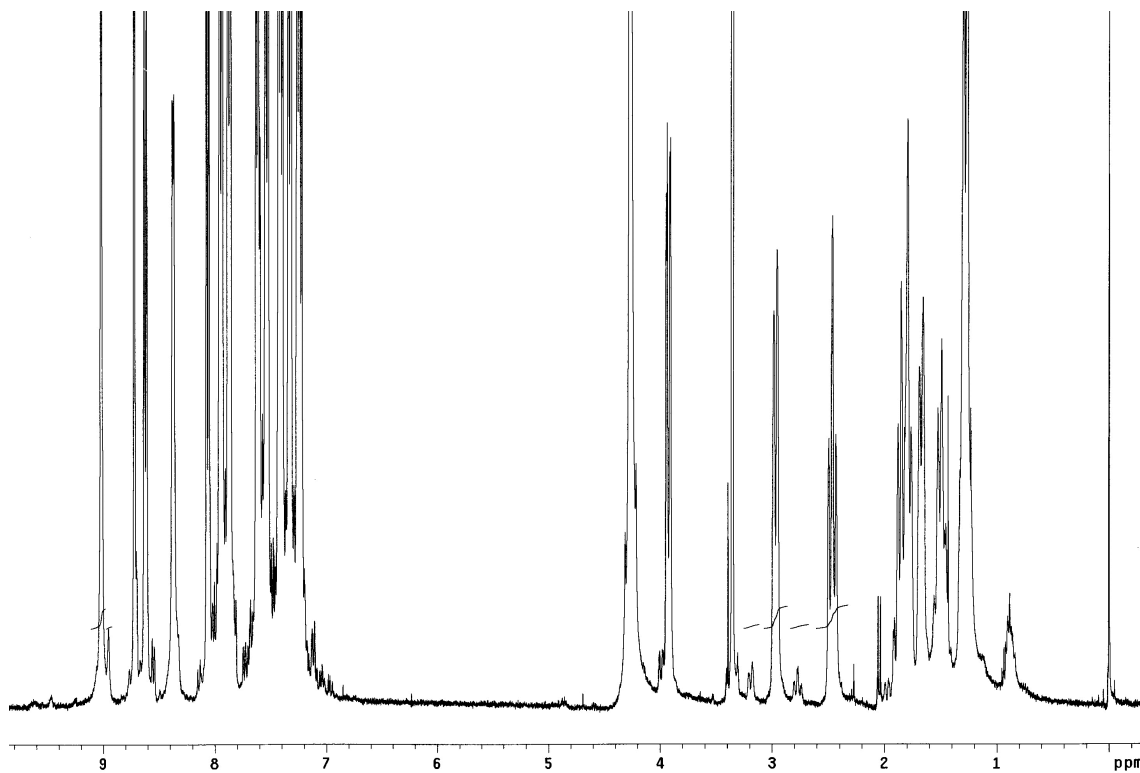
**Expanded HNMR of (-)-1 + (+)-1 + BNPPA (1 equiv)**



**Resolved HNMR signals**

Assigned Proton	Chemical Shift	Enantiomer
H2''	9.11 (s)	<b>(-)-1</b>
H2''	9.08 (s)	<b>(+)-1</b>
H6eq	3.25 (d, J = 12 Hz)	<b>(+)-1</b>
H6eq	3.15(d, J = 12 Hz	<b>(-)-1</b>
H6ax	2.77 (dt, J = 12, 4 Hz)	<b>(+)-1</b>
H6ax	2.62 (dt, J = 12, 4 Hz)	<b>(-)-1</b>

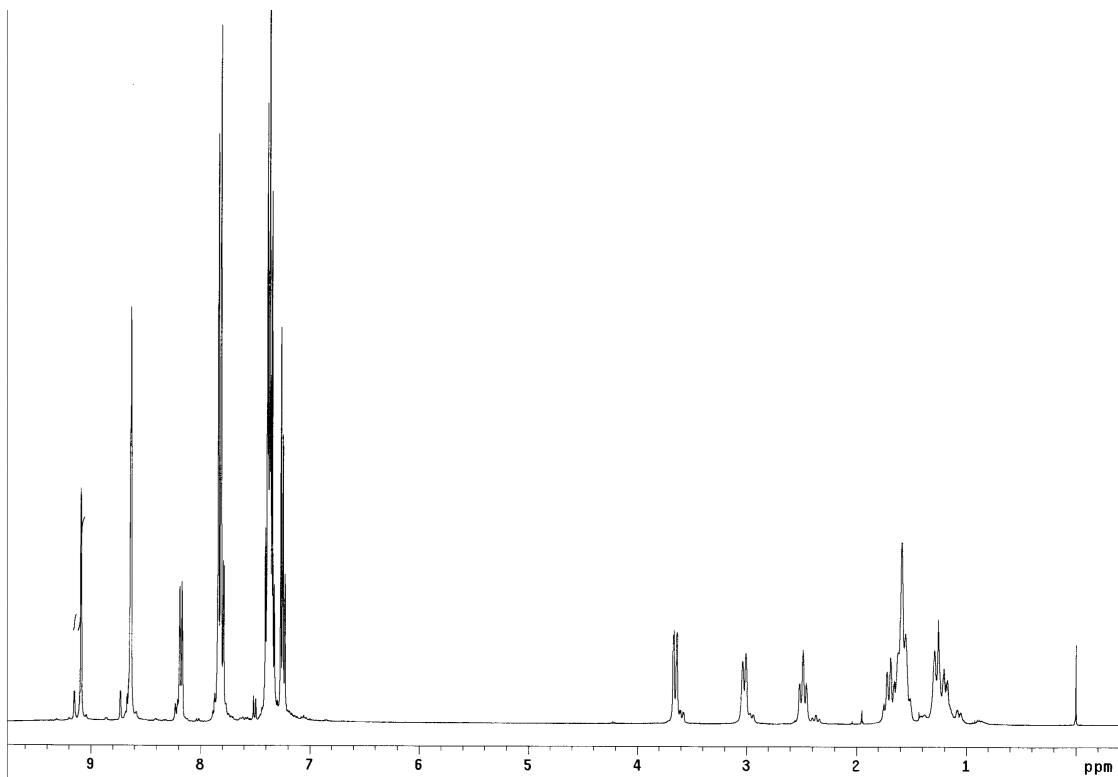
**HNMR of (-)-1 + BNPPA (1 equiv)**



Enantiomer ratio (-)-1/(+)-1 was determined using the signal for H6ax = 13:1\* (86% ee).

\*Integration was obtained using MestReNova® software.

**HNMR of (+)-1 + BNPPA (1 equiv)**



Enantiomer ratio (-)-**1**/(+)-**1** was determined using the signal for H2'' = 9:1\* (80 %ee).

\*Integration was obtained using MestReNova® software.

Crystal Structure of  
 (1*S*,3*aS*)-1-(2-hydroxyethyl)-1,2,3,3*a*,4,5,8,9-octahydropyrrolo[1,2-*a*]quinolin-6(7*H*)-one (6)-  
 Kishi's intermediate

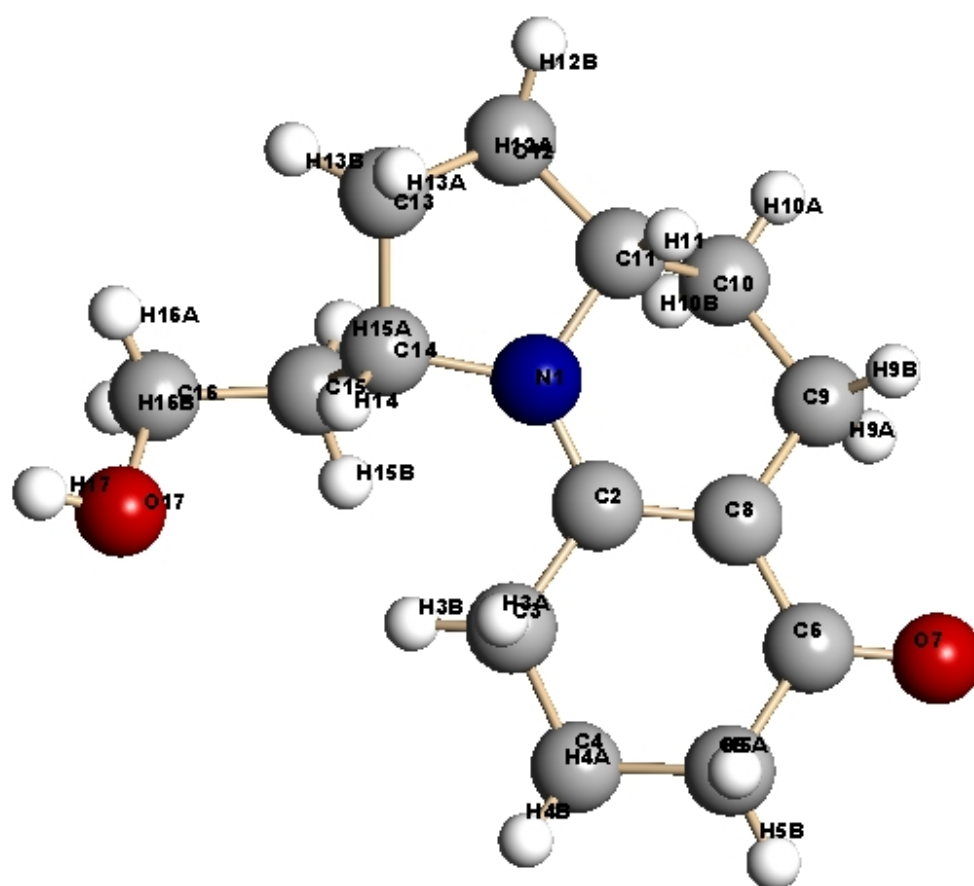


Table 1. Crystal data and structure refinement for Kishi's intermediate (6)

Empirical formula	C <sub>14</sub> H <sub>21</sub> NO <sub>2</sub>	
Formula weight	235.32	
Temperature	120(2) K	
Wavelength	0.71073 Å	
Crystal system	Orthorhombic	
Space group	P2(1)2(1)2(1)	
Unit cell dimensions	a = 8.83600(10) Å	α = 90°.
	b = 10.49690(10) Å	β = 90°.
	c = 13.3180(2) Å	γ = 90°.
Volume	1235.25(3) Å <sup>3</sup>	
Z	4	
Density (calculated)	1.265 Mg/m <sup>3</sup>	
Absorption coefficient	0.084 mm <sup>-1</sup>	
F(000)	512	
Crystal size	0.80 x 0.40 x 0.20 mm <sup>3</sup>	
Theta range for data collection	2.47 to 32.49°.	
Index ranges	-13 ≤ h ≤ 13, -15 ≤ k ≤ 15, -20 ≤ l ≤ 20	
Reflections collected	52997	
Independent reflections	4454 [R(int) = 0.0208]	
Completeness to theta = 32.49°	100.0 %	
Absorption correction	None	
Max. and min. transmission	0.9834 and 0.9360	
Refinement method	Full-matrix least-squares on F <sup>2</sup>	
Data / restraints / parameters	4454 / 0 / 238	
Goodness-of-fit on F <sup>2</sup>	1.080	
Final R indices [I > 2σ(I)]	R1 = 0.0285, wR2 = 0.0805	
R indices (all data)	R1 = 0.0291, wR2 = 0.0814	
Absolute structure parameter	-0.4(6)	
Largest diff. peak and hole	0.371 and -0.182 e. Å <sup>-3</sup>	

Table 2. Atomic coordinates ( $\times 10^4$ ) and equivalent isotropic displacement parameters ( $\text{\AA}^2 \times 10^3$ ) for Kishi's intermediate (**6**). U(eq) is defined as one third of the trace of the orthogonalized  $U^{ij}$  tensor.

	x	y	z	U(eq)
N(1)	4153(1)	1342(1)	10115(1)	17(1)
C(2)	3935(1)	2114(1)	9329(1)	15(1)
C(3)	4005(1)	1532(1)	8296(1)	18(1)
C(4)	3347(1)	2399(1)	7496(1)	22(1)
C(5)	3991(1)	3732(1)	7589(1)	23(1)
C(6)	3814(1)	4264(1)	8634(1)	19(1)
O(7)	3770(1)	5442(1)	8756(1)	30(1)
C(8)	3741(1)	3415(1)	9468(1)	17(1)
C(9)	3499(1)	3977(1)	10499(1)	21(1)
C(10)	3235(1)	2945(1)	11285(1)	23(1)
C(11)	4368(1)	1876(1)	11127(1)	20(1)
C(12)	4237(1)	717(1)	11810(1)	28(1)
C(13)	4756(1)	-383(1)	11141(1)	25(1)
C(14)	4075(1)	-59(1)	10114(1)	18(1)
C(15)	2438(1)	-520(1)	10005(1)	20(1)
C(16)	2333(1)	-1923(1)	9743(1)	25(1)
O(17)	2895(1)	-2090(1)	8753(1)	30(1)



Table 3. Bond lengths [ $\text{\AA}$ ] and angles [ $^\circ$ ] for Kishi's intermediate (**6**).

N(1)-C(2)	1.3386(8)
N(1)-C(11)	1.4716(8)
N(1)-C(14)	1.4719(8)
C(2)-C(8)	1.3881(8)
C(2)-C(3)	1.5069(8)
C(3)-C(4)	1.5174(10)
C(4)-C(5)	1.5161(11)
C(5)-C(6)	1.5079(10)
C(6)-O(7)	1.2480(8)
C(6)-C(8)	1.4257(9)
C(8)-C(9)	1.5093(9)
C(9)-C(10)	1.5238(11)
C(10)-C(11)	1.5185(10)
C(11)-C(12)	1.5236(11)
C(12)-C(13)	1.5287(12)
C(13)-C(14)	1.5328(10)
C(14)-C(15)	1.5323(9)
C(15)-C(16)	1.5154(10)
C(16)-O(17)	1.4202(10)
C(2)-N(1)-C(11)	120.27(5)
C(2)-N(1)-C(14)	126.66(6)
C(11)-N(1)-C(14)	112.84(5)
N(1)-C(2)-C(8)	120.59(6)
N(1)-C(2)-C(3)	117.54(5)
C(8)-C(2)-C(3)	121.75(5)
C(2)-C(3)-C(4)	112.46(5)
C(5)-C(4)-C(3)	110.61(6)
C(6)-C(5)-C(4)	112.26(6)
O(7)-C(6)-C(8)	121.18(7)
O(7)-C(6)-C(5)	119.32(7)
C(8)-C(6)-C(5)	119.48(6)
C(2)-C(8)-C(6)	120.35(6)
C(2)-C(8)-C(9)	121.56(6)

C(6)-C(8)-C(9)	118.06(6)
C(8)-C(9)-C(10)	111.63(6)
C(11)-C(10)-C(9)	109.21(6)
N(1)-C(11)-C(10)	108.87(6)
N(1)-C(11)-C(12)	103.45(6)
C(10)-C(11)-C(12)	117.22(6)
C(11)-C(12)-C(13)	103.40(6)
C(12)-C(13)-C(14)	103.61(6)
N(1)-C(14)-C(15)	111.12(5)
N(1)-C(14)-C(13)	101.62(6)
C(15)-C(14)-C(13)	112.63(6)
C(16)-C(15)-C(14)	112.74(6)
O(17)-C(16)-C(15)	108.17(6)

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Symmetry transformations used to generate equivalent atoms:

Table 4. Anisotropic displacement parameters ( $\text{\AA}^2 \times 10^3$ ) for Kishi's intermediate (**6**). The anisotropic displacement factor exponent takes the form:  $-2\pi^2 [h^2 a^{*2} U^{11} + \dots + 2 h k a^* b^* U^{12}]$

	$U^{11}$	$U^{22}$	$U^{33}$	$U^{23}$	$U^{13}$	$U^{12}$
N(1)	20(1)	16(1)	16(1)	0(1)	1(1)	-1(1)
C(2)	14(1)	15(1)	16(1)	-1(1)	1(1)	-1(1)
C(3)	22(1)	17(1)	16(1)	-3(1)	2(1)	-1(1)
C(4)	25(1)	25(1)	16(1)	-1(1)	0(1)	-1(1)
C(5)	28(1)	22(1)	21(1)	4(1)	4(1)	0(1)
C(6)	17(1)	16(1)	25(1)	1(1)	3(1)	0(1)
O(7)	33(1)	15(1)	42(1)	2(1)	5(1)	2(1)
C(8)	16(1)	14(1)	19(1)	-2(1)	2(1)	-1(1)
C(9)	23(1)	19(1)	22(1)	-7(1)	4(1)	0(1)
C(10)	24(1)	27(1)	18(1)	-4(1)	4(1)	1(1)
C(11)	19(1)	26(1)	16(1)	-1(1)	0(1)	-1(1)
C(12)	30(1)	34(1)	19(1)	6(1)	0(1)	2(1)
C(13)	22(1)	27(1)	27(1)	9(1)	0(1)	4(1)
C(14)	17(1)	16(1)	23(1)	2(1)	4(1)	2(1)
C(15)	17(1)	16(1)	28(1)	0(1)	4(1)	0(1)
C(16)	31(1)	17(1)	28(1)	0(1)	8(1)	-3(1)
O(17)	48(1)	18(1)	25(1)	0(1)	8(1)	2(1)

Table 5. Hydrogen coordinates ( $\times 10^4$ ) and isotropic displacement parameters ( $\text{\AA}^2 \times 10^3$ ) for Kishi's intermediate (**6**).

	x	y	z	U(eq)
H(3A)	5047(13)	1343(11)	8157(9)	21(3)
H(3B)	3453(15)	673(14)	8274(10)	36(3)
H(4A)	2260(16)	2424(13)	7558(9)	29(3)
H(4B)	3589(15)	2044(12)	6844(9)	28(3)
H(5A)	5042(16)	3695(13)	7403(10)	31(3)
H(5B)	3483(18)	4322(16)	7074(12)	47(4)
H(9A)	2623(15)	4558(13)	10474(9)	27(3)
H(9B)	4380(16)	4526(14)	10678(10)	34(3)
H(10A)	3306(17)	3293(14)	11959(11)	37(3)
H(10B)	2229(15)	2564(13)	11225(9)	27(3)
H(11)	5376(15)	2220(13)	11158(10)	31(3)
H(12A)	3163(16)	623(13)	12017(10)	33(3)
H(12B)	4827(18)	767(15)	12450(11)	43(4)
H(13A)	5821(16)	-366(13)	11117(10)	32(3)
H(13B)	4379(16)	-1255(13)	11387(10)	33(3)
H(14)	4667(13)	-394(12)	9554(9)	22(3)
H(15A)	1885(14)	-316(12)	10605(9)	25(3)
H(15B)	1951(14)	-30(12)	9475(9)	24(3)
H(16A)	2888(15)	-2455(14)	10195(9)	28(3)
H(16B)	1311(16)	-2197(13)	9780(10)	30(3)
H(17)	3181(17)	-2868(16)	8715(11)	41(4)

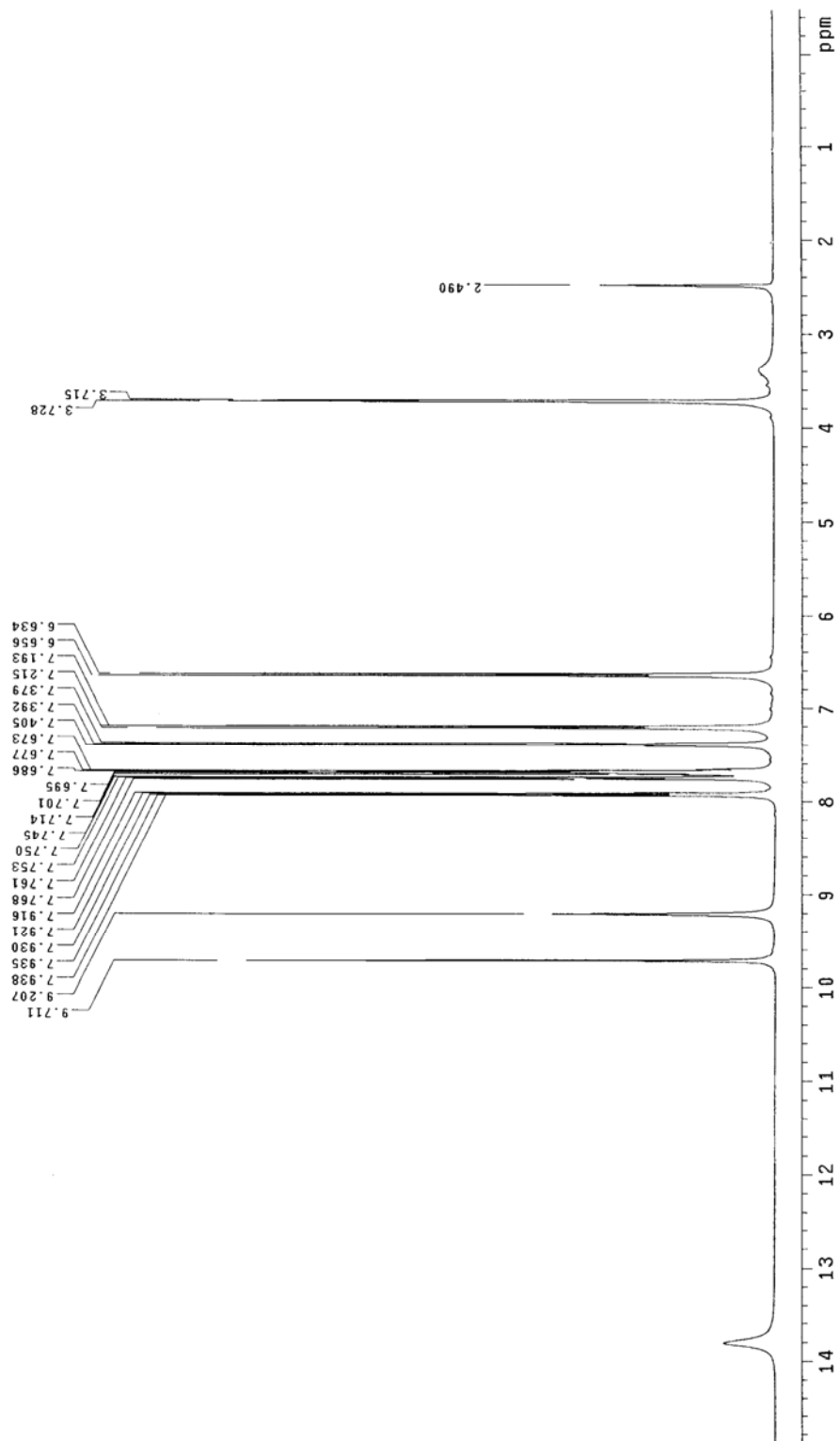
Table 6. Torsion angles [ $^{\circ}$ ] for Kishi's intermediate (**6**).

C(11)-N(1)-C(2)-C(8)	-7.10(9)
C(14)-N(1)-C(2)-C(8)	166.99(6)
C(11)-N(1)-C(2)-C(3)	169.16(6)
C(14)-N(1)-C(2)-C(3)	-16.76(9)
N(1)-C(2)-C(3)-C(4)	165.03(6)
C(8)-C(2)-C(3)-C(4)	-18.76(9)
C(2)-C(3)-C(4)-C(5)	48.90(8)
C(3)-C(4)-C(5)-C(6)	-53.12(8)
C(4)-C(5)-C(6)-O(7)	-154.50(7)
C(4)-C(5)-C(6)-C(8)	27.16(9)
N(1)-C(2)-C(8)-C(6)	167.29(6)
C(3)-C(2)-C(8)-C(6)	-8.80(9)
N(1)-C(2)-C(8)-C(9)	-10.72(9)
C(3)-C(2)-C(8)-C(9)	173.19(6)
O(7)-C(6)-C(8)-C(2)	-173.93(6)
C(5)-C(6)-C(8)-C(2)	4.38(9)
O(7)-C(6)-C(8)-C(9)	4.15(10)
C(5)-C(6)-C(8)-C(9)	-177.54(6)
C(2)-C(8)-C(9)-C(10)	-9.66(9)
C(6)-C(8)-C(9)-C(10)	172.29(6)
C(8)-C(9)-C(10)-C(11)	44.21(8)
C(2)-N(1)-C(11)-C(10)	42.99(8)
C(14)-N(1)-C(11)-C(10)	-131.86(6)
C(2)-N(1)-C(11)-C(12)	168.36(6)
C(14)-N(1)-C(11)-C(12)	-6.50(7)
C(9)-C(10)-C(11)-N(1)	-60.16(7)
C(9)-C(10)-C(11)-C(12)	-177.05(6)
N(1)-C(11)-C(12)-C(13)	27.82(8)
C(10)-C(11)-C(12)-C(13)	147.62(6)
C(11)-C(12)-C(13)-C(14)	-39.02(8)
C(2)-N(1)-C(14)-C(15)	-71.79(8)
C(11)-N(1)-C(14)-C(15)	102.66(6)
C(2)-N(1)-C(14)-C(13)	168.17(6)
C(11)-N(1)-C(14)-C(13)	-17.37(7)

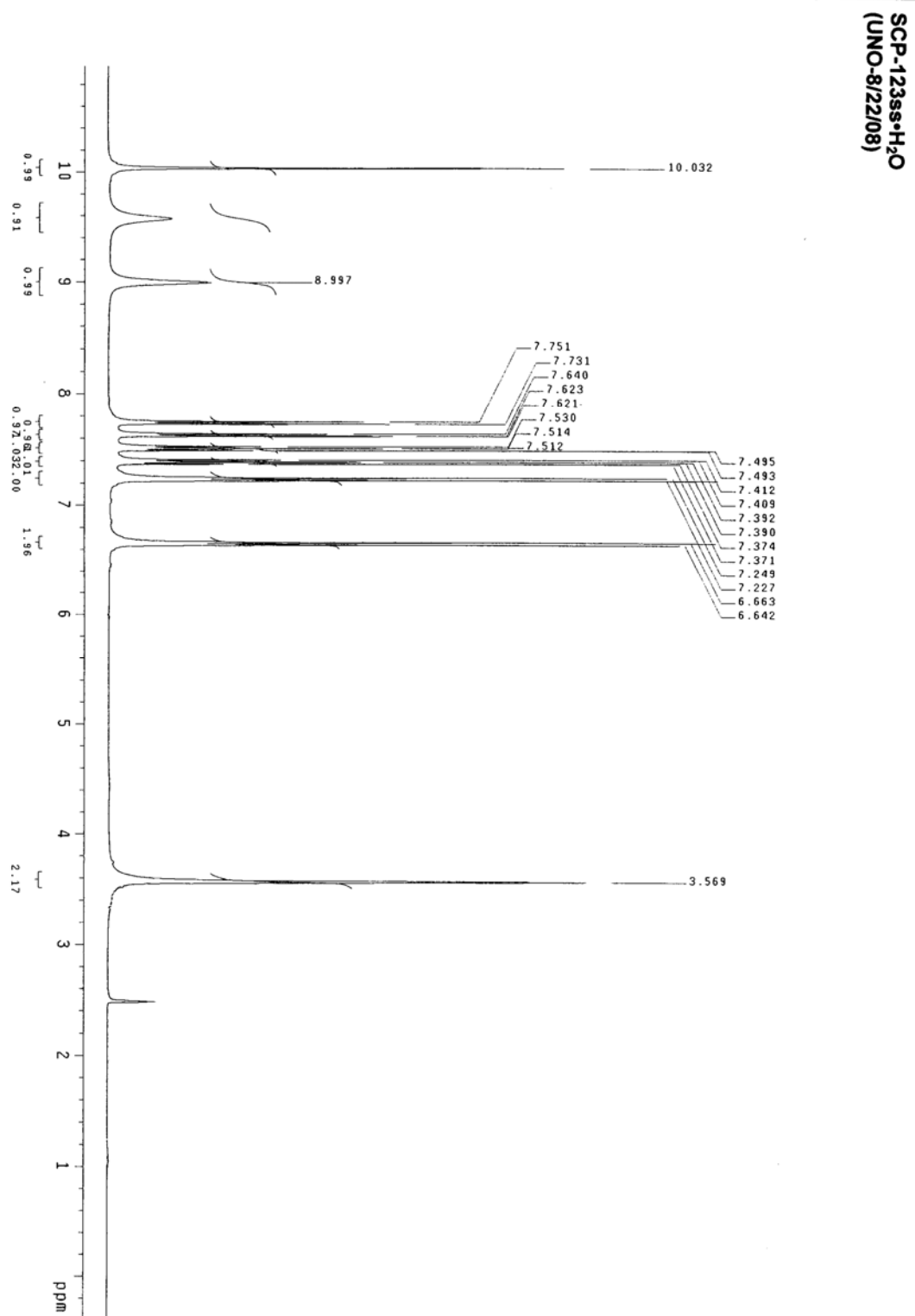
C(12)-C(13)-C(14)-N(1)	34.11(7)
C(12)-C(13)-C(14)-C(15)	-84.86(7)
N(1)-C(14)-C(15)-C(16)	165.84(6)
C(13)-C(14)-C(15)-C(16)	-80.89(7)
C(14)-C(15)-C(16)-O(17)	-68.29(8)

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Symmetry transformations used to generate equivalent atoms:

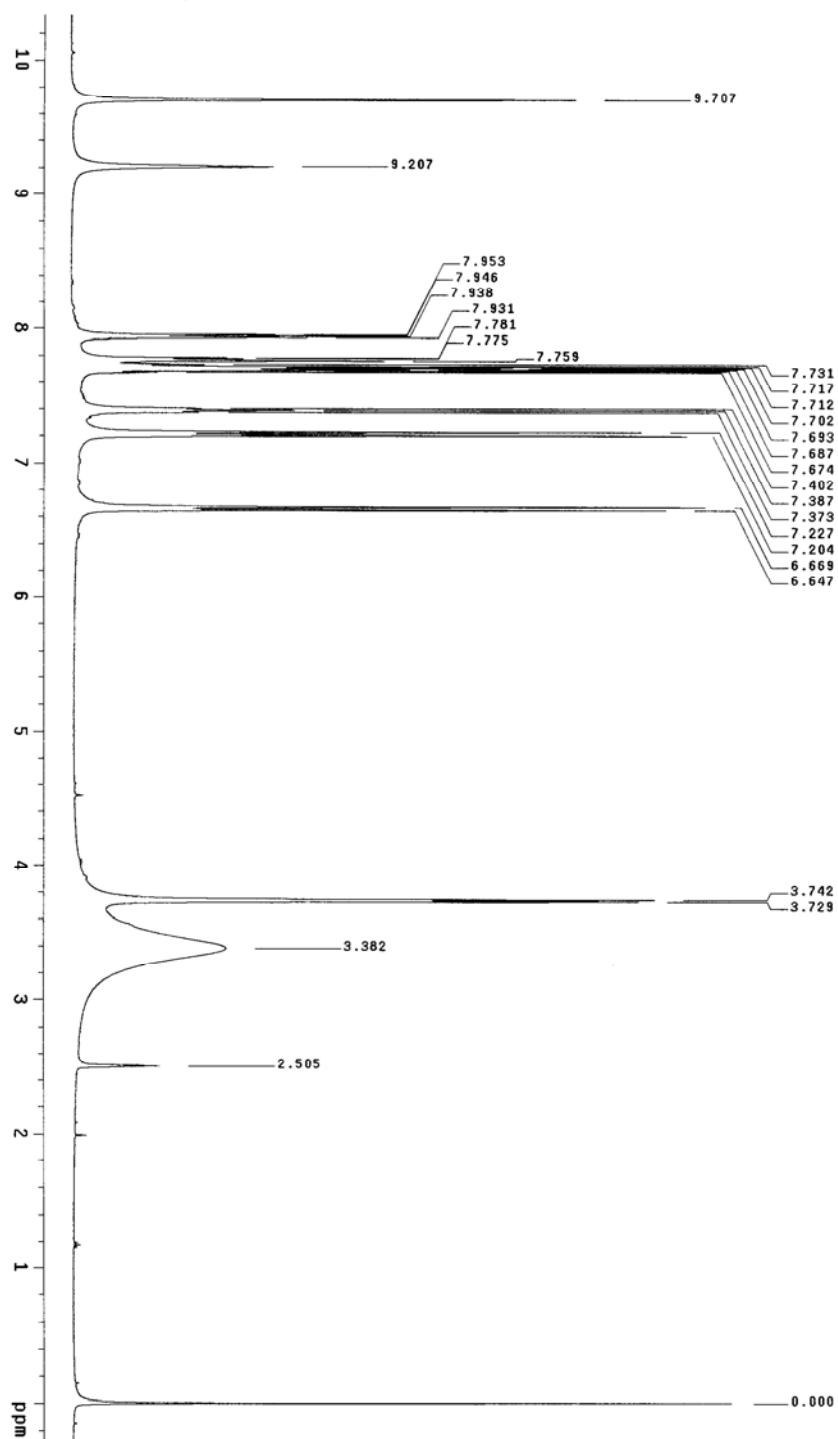
<sup>1</sup>H NMR Spectrum of SCP-123 (**4**) in DMSO-d<sub>6</sub>.

$^1\text{H}$  NMR Spectrum of SCP-123ss (**5**) in DMSO- $\text{d}_6$ .





$^1\text{H}$  NMR Spectrum of **12** in DMSO- $d_6$ .



## **HPLC ANALYSIS**

### **1. REAGENTS AND INSTRUMENTATION**

- Trifluoroacetic acid, HPLC Grade Aldrich.
- Acetonitrile HPLC grade EM Science.
- HPLC Waters 501/486 Tunable Detector
- Column: Waters Nova-Pak C18 (3.9 x 150 mm) Steel Analytical.

### **2. CROMATOGRAPHIC CONDITIONS**

- Flow: 1.0 ml/min
- Column Temperature 25<sup>0</sup>C
- Volume injection: 5 µl
- Detector: Ultraviolet absorption- wavelength 254 nm.

Mobile phase: Gradient, **Table 1.**

A. TFA 0.1 % in acetonitrile

B. TFA 0.1% in water.

**Table 1: Gradient.**

Time (min)	%A	%B
0	0	100
5	0	100
30	100	0
32	100	0
33	0	100
45	0	100

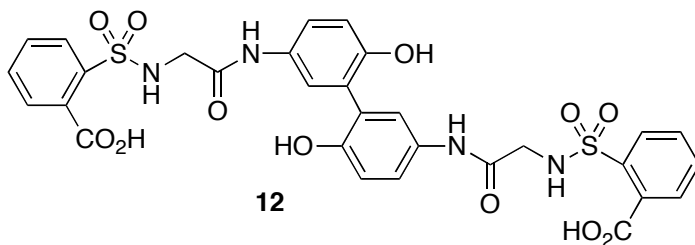
### **3. RETENTION TIME**

Compound	t <sub>R</sub> (min)*
<b>3</b>	3.1
<b>4</b>	15.8
<b>12</b>	17.0

\*Average of three runs.

## LC-ESI-MS IDENTIFICATION OF DIMER 12

LC-TSQ Quantum Instrument (Thermo-Finnigan) coupled to Surveyor, equipped with electro-spray ionization (ESI)

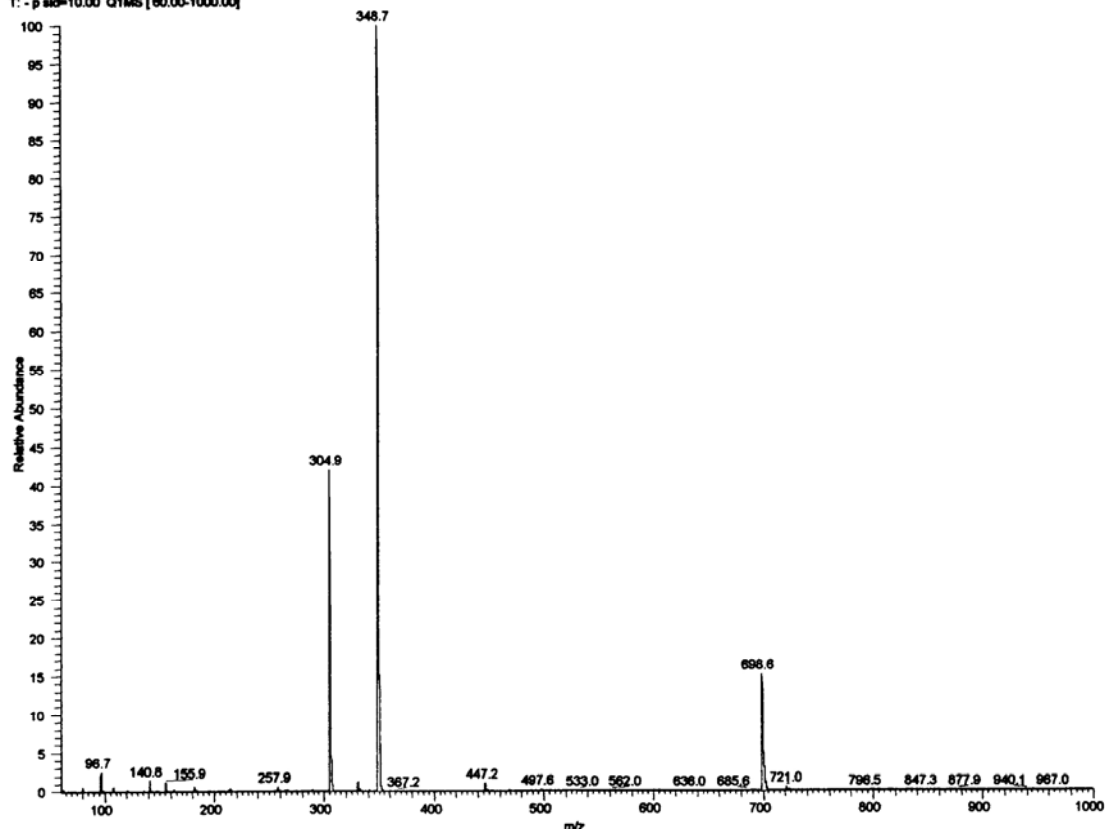


12

Chemical Formula: C<sub>30</sub>H<sub>26</sub>N<sub>4</sub>O<sub>12</sub>S<sub>2</sub>  
MW: 698.68

### Q1 Full Scan

JMS-1-28 Mark-2 13Feb04 #43 RT: 0.64 AV: 1 NL: 1.24E7  
T: - p acd=10.00 Q1MS [60.00-1000.00]



## **VITA**

Lei Miao was born in Jinzhai county, Anhui province, China on September 27, 1982. He graduated from Lu'an No.1 high school, Lu'an, Anhui, China in July 1998. He received his bachelor in Science degree in Chemistry at the University of Science & Technology of China, Hefei, China in 2003. He continued his education at the University of New Orleans to pursue a PhD degree in organic synthesis under the supervision of Professor Mark L. Trudell in 2004. He went on to complete the requirements for this degree in August 2009.