Wayne State University Dissertations

January 2018

Hydroxylamines And Hydrazines As Surrogates Of Sp3 Carbons In Medicinal Chemistry

Sandeep Dhanju
Wayne State University, fi1375@wayne.edu

Follow this and additional works at: https://digitalcommons.wayne.edu/oa_dissertations

Part of the Chemistry Commons

HYDROXYLAMINES AND HYDRAZINES AS SURROGATES OF SP^3 CARBONS IN MEDICINAL CHEMISTRY

by

SANDEEP DHANJU

DISSERTATION

Submitted to the Graduate School

of Wayne State University,

Detroit, Michigan

in partial fulfillment of the requirements

for the degree of

DOCTOR OF PHILOSOPHY

2019

MAJOR: CHEMISTRY (Organic)

Approved By:

Advisor	Date

DEDICATION

I dedicate my PhD work to my parents Sanubhai Shrestha and Punamaya Dhanju, for their endless love and support.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF SCHEMES

xiii

LIST OF ABBREVIATIONS

Ac	Acetyl
AIBN	Azobisisobutyronitrile
Ar	Aryl
9-BBN	9-Borabicyclo[3.3.1]nonane
Boc	*tert*-Butyloxycarbonyl
Bn	Benzyl
Bu	Butyl
Bz	Benzoyl
c	Molar concentration
^{o}C	Degree Celsius
CAM	Ceric ammonium molybdate
Cbz	Carboxybenzyl
*m*CPBA	*m*-Chloroperbenzoic acid
DAST	Diethylaminosulfur trifluoride
DCC	*N,N'*-Dicyclohexylcarbodiimide
DEPC	Diethyl phosphorocyanidate
DFT	Density functional theory
DIAD	Diisopropyl azodicarboxylate
DIBAL	Diisobutylaluminium hydride
DIPEA	Diisopropylethylamine
DMAP	4-Dimethylaminopyridine
DMF	Dimethylformamide

DMP	Dess-Martin periodinane
DMS	Dimethyl sulfide
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid
DOS	Diversity-oriented synthesis
EC_{50}	Half maximal effective concentration
EDCI	*N*-Ethyl-*N'*-(3-dimethylaminopropyl)carbodiimide
ESI	Electrospray ionization
Et	Ethyl
ETC	Electron transport chain
FDA	Food and drug administration
Fsp^3	Fraction of sp^3-hydbridized atoms
h	Hour(s)
HATU	Hexafluorophosphate azabenzotriazole tetramethyl uronium
HbO_2	Oxyhemoglobin
HMBC	Heteronuclear multiple bond correlation
HOAt	1-Hydroxy-7-azabenzotriazole
HOBt	1-Hydroxybenzotriazole
HRMS	High resolution mass spectrometry
HRP	Horseradish peroxidase
HTS	High-throughput screening
Hz	Hertz
IC_{50}	Half maximal inhibitory concentration

KHMDS	Potassium bis(trimethylsilyl)amide
LC_{50}	Lethal concentration required to kill 50% of the population
LDA	Lithium diisopropylamide
LiHMDS	Lithium bis(trimethylsilyl)amide
Me	Methyl
mmol	Millimole
MS	Molecular sieves
NaHMDS	Sodium bis(trimethylsilyl)amide
NMDA	*N*-Methyl-D-aspartate
NMO	*N*-Methylmorpholine-*N*-oxide
NMR	Nuclear magnetic resonance
NOE	Nuclear overhauser effect
Ph	Phenyl
PMNs	Polymorphonuclear cells
ppm	Parts per million
py	Pyridine
RNA	Ribonucleic acid
RO3	Rule of three
RO5	Rule of five
SAR	Structure activity relationship
TBAF	Tetrabutylammonium fluoride
TBSCl	*tert*-Butyldimethylsilyl chloride
TCDI	Thiocarbonyldiimidazole

Tf	Trifluoromethanesulfonyl
TFA	Trifluoroacetic acid
TfOH	Trifluoromethanesulfonic acid
THF	Tetrahydrofuran
Tmob	Trimethoxybenzyl
TMSOTf	Trimethylsilyl trifluoromethanesulfonate
US	United States
VEGF	Vascular endothelial growth factor
VT-NMR	Variable temperature nuclear magnetic resonance

CHAPTER 1. INTRODUCTION

1.1. Motives of the research

1.1.1. Medicinal chemistry lacks diversity in compound collections

Drug discovery is a highly complex and multidisciplinary process. The medicinal chemistry community has driven tremendous advances in drug discovery programs in last two decades. The need of screening millions of compounds to develop one lead in the drug development process led to the development of high throughput screening (HTS). HTS can efficiently generate millions of data just by screening large libraries in short periods of time.[1] Both the academic and the industrial communities benefit enormously from HTS in terms of time and cost of identifying preclinical drug candidates (lead compounds). As a result, HTS originated small molecules dominate the collections of the US FDA approved new drug candidates implying that HTS is an essential and necessary frontline tool in drug discovery today.[2]

As HTS and its latest variant, the fragment-based approach, are in full-swing and are key methods in drug discovery programs, the compositions of compound libraries will have a significant impact on the outcomes in pharmaceutical discoveries.[1,3] To assure the quality of the outcomes that can be tractable in clinical phases, medicinal chemists today follow principles of drug-like or lead-like molecules while generating compound libraries. These principles use several molecular descriptors in order to filter the compounds. One of the widely used practices in HTS compound collections is Lipinski's rule of five (RO5).[4] Alternatively, the fragment based approach follows the Astex rule of three (RO3).[5] Several properties like molecular weight, topological polar surface area, rotatable bonds, and numbers of hydrogen bond donors and acceptors are considered

while filtering.[4,5] Another approach in identifying drug-like chemical space is exclusion of undesirable molecular moieties from compound collections.[6] Moieties which are known to generate false positives in screening and are associated with toxicities are undesirable.[6] For example, molecules with heteroatom-heteroatom bonds are excluded from current compound collections based upon their perceived reactivity and toxicity.[6]

Although HTS is a successful approach in addressing multiple biological targets, failures in clinical phases as well as in hitting challenging complex biological targets such as protein-protein interactions are also common.[3] These failures are warnings to medicinal chemists about the lack of the diversity in their current compound libraries and are unavoidable. Also, the descriptors used in the commonly applied principles of drug-like molecules do not address molecular complexity directly.[7] As a result, current HTS libraries are highly populated by low molecular weight and relatively planar compounds with high sp^2 character and little stereochemical complexity.[3,7] Unfortunately, such planar compounds cover only a very small corner of the chemical space. Furthermore, the binding sites in biological targets are rarely planar, which undoubtfully can be viewed as one of the primary reasons of failure in HTS campaigns. In order to overcome the limitations of planarity, the so-called flatland, and to fulfil the need for complexity, the strategy of increasing the fraction of saturated carbons (Fsp^3) within compounds should be considered. In 2009, Lovering et al. conducted a survey[7] on the collection of compounds in different phases of drug discovery and development processes and found that the fractions of sp^3 hybridized atoms and the number of stereogenic centers are higher in those compounds. Therefore, the survey suggested considering the Fsp^3 of the molecules as a descriptor in the compound collections.

Recognizing the need for complexity and diversity, several creative strategies have been designed to date in order to generate more diverse libraries. Diversity oriented synthesis (DOS)[8-11] is one of the approaches that generates architecturally diverse molecules in few steps starting from simple starting materials. These compounds resemble more natural product or drug-like molecules with high Fsp^3 and stereochemical complexity. Other strategies are centered on the use of natural products as the starting point to generate the natural product-like molecules.[12] Even though these creative ideas have introduced natural product-like moieties with complexity in terms of Fsp^3 and the presence of stereogenic centers, several problems are still left unaddressed such as the creation of new complex scaffolds from unexplored parts of chemical space. Furthermore, the synthesis of compounds with the natural product-like scaffolds is still considered as a challenging and time-consuming area of the research.

To overcome these shortcomings, the medicinal chemistry community needs a novel strategy that can open up the new chemical space with new scaffolds. Also, these scaffolds should have the potential of generating large libraries of compounds with reduced synthetic difficulty but high structural complexity.

1.1.2. Hydroxylamines and hydrazines as surrogates of sp^3 carbons

The Crich lab has identified the hydroxylamine and the hydrazine functionalities as new scaffolds that have the potential to enrich compound collections by increasing Fsp^3 in compounds and to address the necessity of diversity from the new chemical space without the synthetic complications arising from the presence of additional stereogenic centers.

With low basicity (pK_a = 5.9 in water)[13,14] and a low inversion barrier (~15 kcal·mol^{-1}),[15-19] hydroxylamines are little protonated under normal physiological conditions and have the potential of changing configuration by rapid inversion at the nitrogen center. Therefore, tri-substituted hydroxylamines, in particular, can serve to mimic both enantiomers of simple stereogenic ethers and alkanes while reducing synthetic and structural complexity (**Figure 1**).
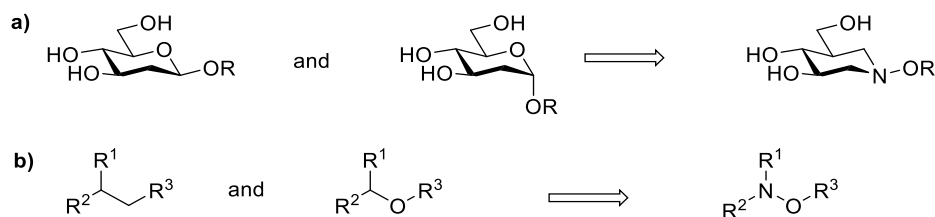
Figure 1. Hydroxylamines as convertible mimics of stereogenic centers. **a)** *N*-Alkoxypiperidines as mimics of axial and equatorial glycosides.[18,19] **b)** Hydroxylamines as mimics of chirally substituted alkanes and ethers.

Hydrazines share similar properties to hydroxylamines including their low inversion barriers (~8 kcal·mol^{-1})[20] and weak basicity (pK_a = 6.4 in water).[21] These properties establish hydrazines as another interesting functional group in which the two nitrogen atoms can substitute for two stereogenic centers and reduce synthetic and structural complexity (**Figure 2**).
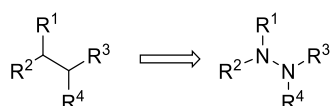
Figure 2. Hydrazines as convertible mimics of two stereogenic centers.

Taking advantage of these unique properties of hydroxylamines and hydrazines, the Crich lab initiated a program to design molecules in which the nitrogen atoms of hydroxylamine and hydrazine functionality substitute important stereogenic centers of

medicinally important molecules, with the goal of reducing synthetic complexity and developing new compositions of matter. As a proof of concept, the Crich lab has recently prepared and evaluated a series of mono-, di-, and tri-meric *N*-alkoxyimino sugars as *β*-(1→3)-glucan mimetics in which the hydroxylamine nitrogen replaces the anomeric carbon and eliminates the need for diastereoselective glycosidic bond formation (**Figure 3**).[18,19]
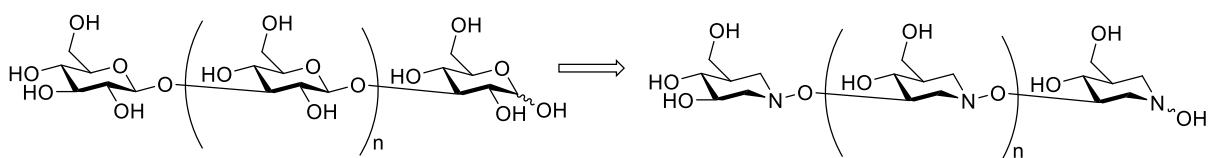
Figure 3. Hydroxylamine based *β*-(1→3)-glucan mimetics as hydroxalogs of *β*-(1→3)-glucans.

1.1.3. Aims of the research

Recognizing the inherent potentials of hydrazines and hydroxylamines in medicinal chemistry, the research presented in this thesis aims to apply the concept of hydrazine and hydroxylamine analogs beyond the carbohydrates studied initially. However, before the broader application of hydroxylamines and hydrazines can be investigated, improved methods for their synthesis are required. Therefore, the work will also focus on developing an effective methodology that can give easy access to diverse sets of tri-substituted hydroxylamines and so potentially be applied in generating libraries of compounds.

Characteristic features of the hydroxylamines and hydrazines including their conformational properties and syntheses, and analog design will be discussed in the following sections.

1.2. Hydroxylamines

1.2.1. Introduction

Hydroxylamines or alkoxyamines represent the class of compounds that are derived from the parent hydroxylamine (H_2N-OH). Depending upon the substitution patterns on the nitrogen and oxygen atoms, they can be classified as mono-substituted, di-substituted, or tri-substituted (**Figure 1**).

Mono-substituted hydroxylamines

Di-substituted hydroxylamines

Tri-substituted hydroxylamines

R = alkyl, aryl

Figure 4. Structures of substituted hydroxylamines.

The N-O bond is also present in oximes and hydroxamic acids (**Figure 5**), which are considered as derivatives of the parent hydroxylamine (NH_2OH). However, oximes have C=N double bonds and hydroxamic acids have *N*-acyl groups giving properties that differentiate them from hydroxylamines particularly in the context of their planar sp^2 nitrogens.[22]

Hydroxylamine

Oxime

Hydroxamic acid

R = alkyl, aryl, H
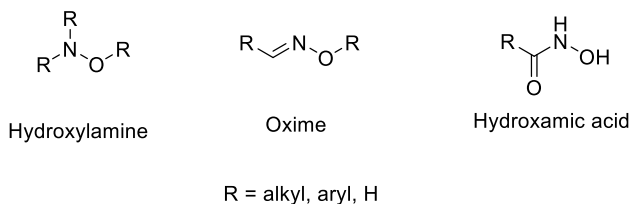
Figure 5. Structures of hydroxylamines, oximes, and hydroxamic acids.

A common confusion in the literature revolves around the use of the word hydroxylamine. Some literature uses it to represent all the derivatives of the parent hydroxylamine including oximes and hydroxamic acids, whereas others have used it to

represent only aryl and alkyl-substituted hydroxylamines. In this thesis, the later representation will be followed.

1.2.2. Conformational properties of hydroxylamines

The presence of an oxygen atom adjacent to nitrogen atom has a substantial effect on the conformation of hydroxylamines. The repulsive interaction between the lone pairs of electrons on these atoms should be minimized in the preferred conformation. In molecular orbital calculations, two energy minima were observed for hydroxylamines representing two stable conformations **1a** and **1b** (**Figure 6**).[22,23] The energy differences between these conformers range from 0.67 kcal·mol^{-1} to 10.79 kcal·mol^{-1} depending upon the substitution patterns.[23] Interestingly, conformer **1a** is more stable with eclipsing of lone pairs with bonds, whereas conformer **1b** is less stable with staggered lone pairs and bonds. This property of hydroxylamines can be viewed as an effect of maximization of separation of the lone pairs on adjacent atoms.[23]

1a   **1b**
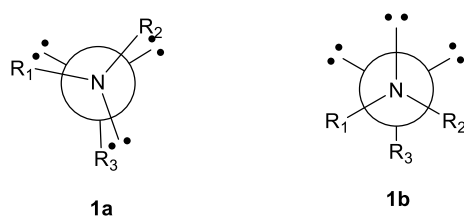
R_1, R_2, R_3 = alkyl, aryl, H

Figure 6. Conformations of hydroxylamine.

Another interesting phenomenon in hydroxylamines is the fast conformational inter-conversion. This stereomutation is attributable to either an individual or a combination of two unique processes; nitrogen inversion (N_{inv}) and N-O bond rotation (N-O$_{rot}$).[17] The energy barriers for nitrogen inversion in hydroxylamines were determined[15-18] to be ~ 15 kcal·mol^{-1}, and are higher than their amine analogs (~ 5 kcal·mol^{-1}),[24] however,

they are low enough to permit rapid inversion at the nitrogen center at ambient temperature. Due to their fast inversion at room temperature, the pyramidal nitrogen atoms in hydroxylamines are not configurationally stable.

The stereomutation of hydroxylamines can be best illustrated as in **Scheme 1**.[17] Owing to the low barrier to inversion, the more stable conformer **1a** converts to the less stable conformer **1b**, which in turn converts to the enantiomer of the more stable conformer **1a*** by rotation of the N-O bond and minimization of the lone pair-lone pair repulsions. The barrier to rotation in the above conversion (**1b → 1a***) is usually small (< 5 kcal.mol^{-1}).[17] Similarly, another enantiomeric less stable conformer **1b*** is the result of N-O bond rotation of **1a** or nitrogen inversion of **1a*** . The barrier to rotation for the conversion of **1a** to **1b*** is unusually high (12 - 15 kcal·mol^{-1}), since the former is more stable than the latter and the high torsional barriers are associated with the process.[17] The more stable conformers **1a** and **1a*** are epimers of each other as are the less stable conformers **1b** and **1b***. As the population of the more stable conformer **1a** and **1a*** are identical and interconvertible at room temperature, a single molecule of a hydroxylamine can represent both epimers of a compound. This is the principle behind the hydroxylamine analog or hydroxalog concept.
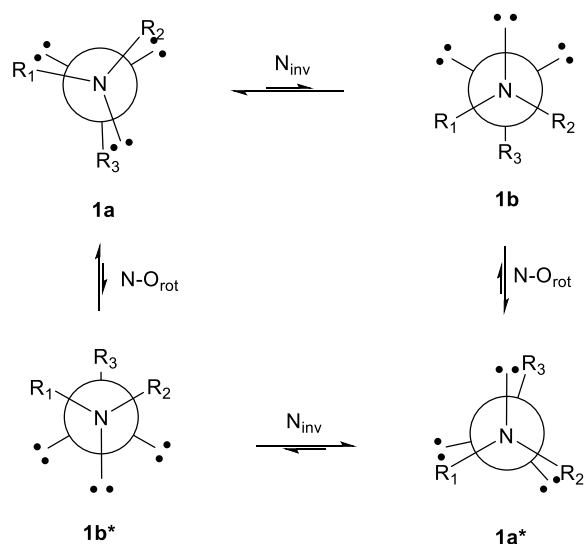
Scheme 1. Stereomutation in hydroxylamines.

1.2.3. Hydroxylamines in medicinal chemistry

The hydroxylamine moiety is an underrepresented functional group in medicinal chemistry, though it has potential to enrich compound collections without synthetic complication from the presence of an additional stereogenic center. Although not common, the hydroxylamine moiety does exist in a small number of bioactive natural products. The antitumor antibiotics esperamicin A1[25,26] and calicheamicins,[27,28] and the selective inhibitor of bacterial tyrosyl tRNA synthase SB-219383[29] are some representative examples containing a hydroxylamine moiety in nature (**Figure 7**).

Anticancer antibiotic esperamicin A1

tRNA synthase inhibitor
SB-219383

Anticancer Antibiotic Calicheamicin γ1
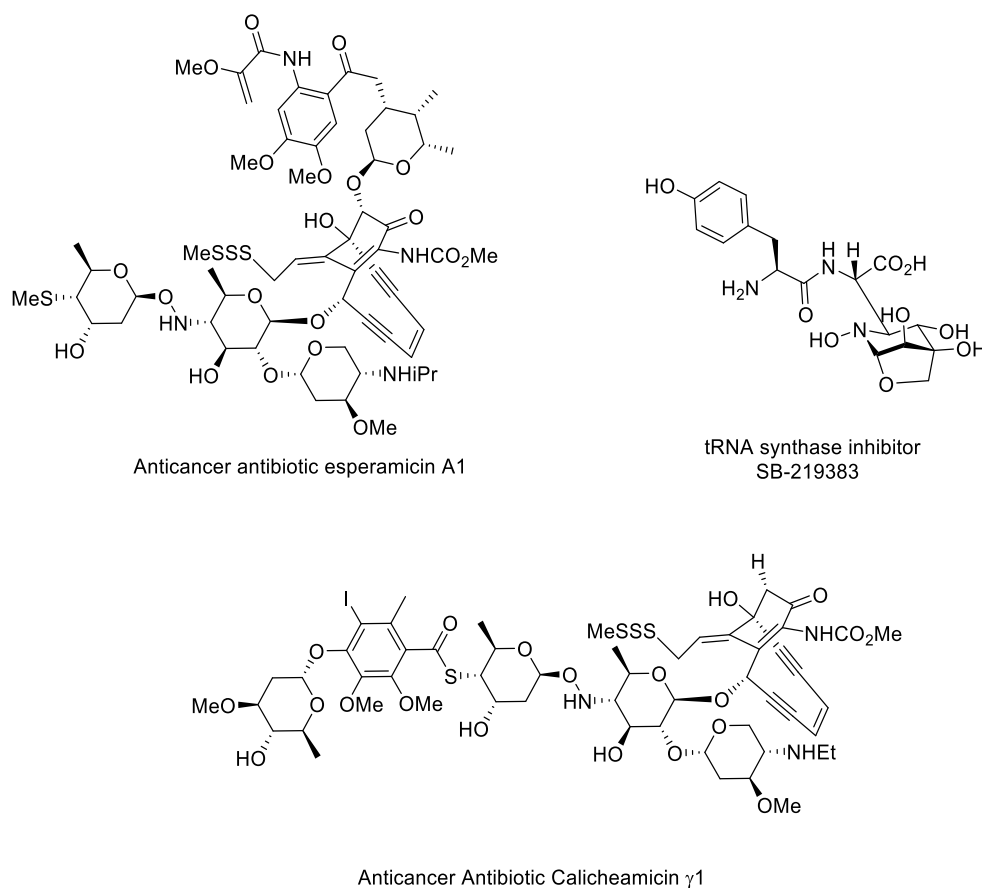
Figure 7. Natural compounds with hydroxylamine moieties.

On the other hand, there are a few examples of the use of hydroxylamines in medicinal chemistry that aim to exploit its rich potential. Two examples are the hydroxylamine-based *β*-(1→3)-glucan mimetics[19] from Crich lab, and a selective inhibitor of human neuraminidase isoenzymes NEU3[30] from the Cairo lab (**Figure 8**).

Hydroxylamine-based *β*-(1→3)-glucan mimetics

Selective inhibitor of human
neuraminidase isoenzymes NEU3
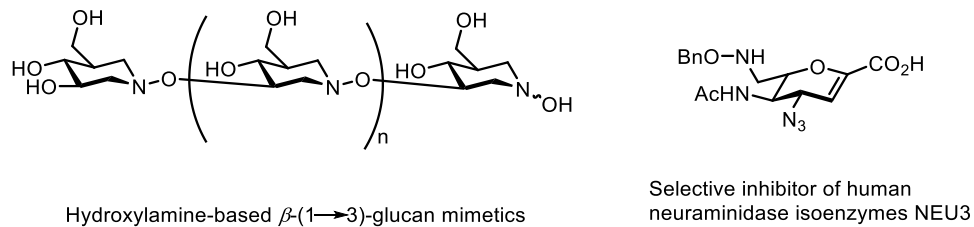
Figure 8. Synthetic compounds with hydroxylamine moieties for use in medicinal chemistry.

Though hydroxylamines have the potential to enrich compound libraries, they have been typically excluded as undesirable moieties[6,31] based upon the perceived problems

with the N-O bond, particularly, its supposed weakness and electrophilicity. However, both DFT[6] and experimentally[32] determined bond dissociation energies for the N-O bond range from 55 to 65 kcal·mol^{-1} depending upon the substitution patterns. Therefore, they are stable entities and are not thermodynamically liable as perceived. On the other hand, hydroxylamines, particularly *N*-arylhydroxylamines, with free -NH or -OH groups, are intermediates on the metabolic pathway of amines, and are converted to electrophilic metabolites by acylation or sulfation as exemplified in **Scheme 2**. [33,34] These metabolites further bind covalently to DNA and induce mutation. In contrast, with no free -NH and -OH groups, tri-substituted hydroxylamines cannot activated to electrophiles in this manner. Therefore, they are not prone to form adducts with DNA to cause genotoxicity and also do not bind covalently with proteins to result in the false positives in screening.[6] Moreover, heteroatom-heteroatom bonds such as N-O and N-N bonds and other electrophilic scaffolds are rather common in marketed drugs.[31] Therefore, discarding hydroxylamines from screening collections based upon the perceived false positives and the weak N-O bond can lead to the loss of quality hits.[6,35]
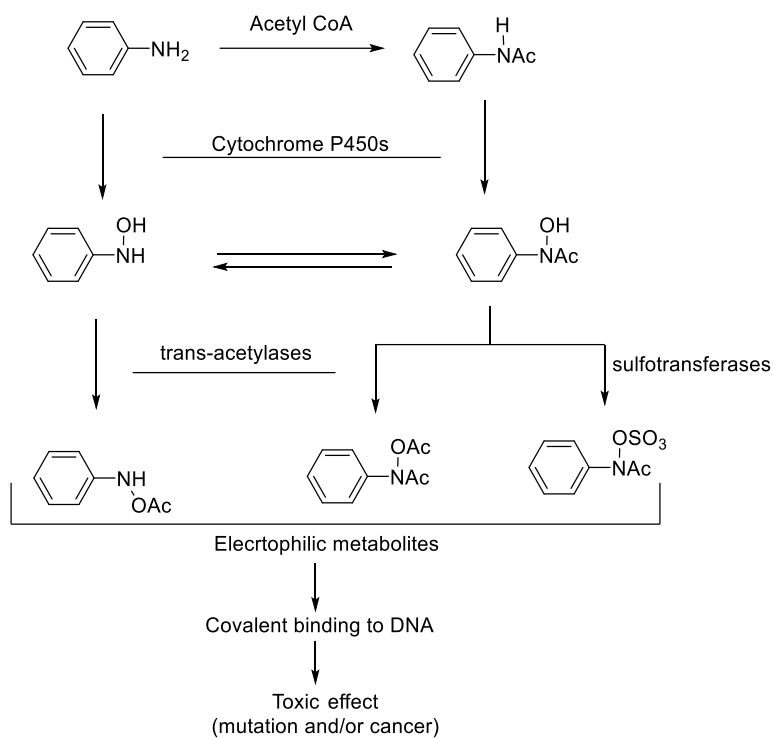
Scheme 2. A representative pathway of metabolic activation of aromatic amines.

1.2.4. Synthesis of tri-substituted hydroxylamines

Depending upon the strategic disconnection approach and the available synthetic methods in the literature, three possible ways for tri-substituted hydroxylamines synthesis can be devised (**Scheme 3**). An ideal route to the substituted hydroxylamines should involve N-O bond formation (**Scheme 3** route a). This ideal route has a huge potential of generating large libraries of compounds starting from commercially available amines and alcohols as starting materials. However, very few examples of tri-substituted hydroxylamine synthesis were reported to date following this route, which involves the displacement of a leaving group from a nitrogen atom by an alcohol or from an oxygen atom by an amine or an amide.[36-39] Progress in this line of work has not been promising due to the combination of low yields and harsh reaction conditions. This approah remains challenging for synthetic chemists.

$$R_1\underset{}{\overset{R_2}{\underset{}{N}}H}$$

a) N-O bond
formation   HO$-$R$_3$

$$R_2\overset{H}{\underset{}{N}}\underset{O}{}\,R_3 \quad \xleftarrow{\substack{b)\ \textit{N-derivatization} \\ R_1-X}} \quad R_1\underset{}{\overset{R_2}{N}}\underset{O}{}\,R_3 \quad \xrightarrow{\substack{c)\ \textit{O-derivatization} \\ X-R_3}} \quad R_1\underset{}{\overset{R_2}{N}}\,OH$$
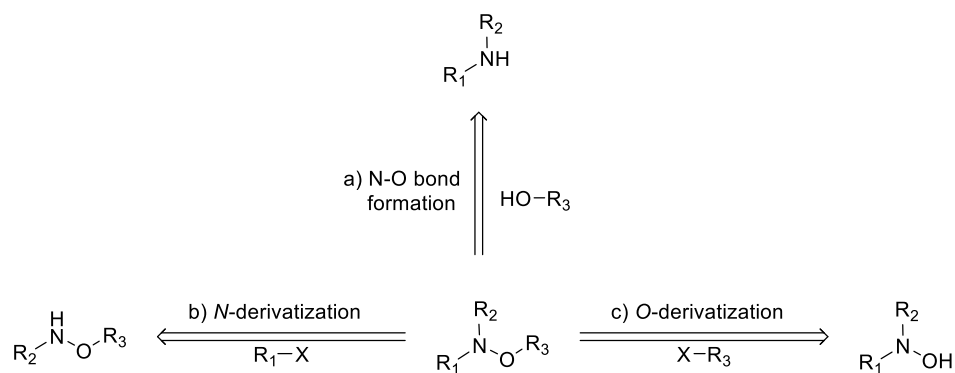
Scheme 3. Possible ways for the synthesis of tri-substituted hydroxylamines.

N-Alkylation (**Scheme 3** route b) of *N,O*-disubstituted hydroxylamines or *O*-alkylation (**Scheme 3** route c) of *N,N*-disubstituted hydroxylamines are viewed as plausible alternative routes. Several synthetic methods for hydroxylamine derivatives have been developed using direct *N*-alkylation and *O*-alkylation routes,[40,41] however methods for the synthesis of tri-substituted hydroxylamines, in particular, are relatively sparse.

One of the traditional and common approaches for the tri-substituted hydroxylamines is *N*-alkylation of *N,O*-disubstituted hydroxylamines via reductive amination (**Scheme 4a**) or S$_N$2 substitution reactions (**Scheme 4b**).[41] Several *N*-alkoxyiminosugar derivatives have been prepared from *O*-substituted hydroxylamines as starting materials by using either double reductive amination (**Scheme 4c**) or combination of reductive amination and S$_N$2 substitution reactions (**Scheme 4d**).[18,19,42,43] Various tri-substituted hydroxylamines have also been synthesized from *N*-methoxyamides in a one pot reaction that involved partial reduction with DIBAL, followed by Lewis acid catalyzed *in situ* generation of *N*-oxy iminium ions and the nucleophilic addition of carbon nucleophiles (**Scheme 4e**).[44] Moreover, bicyclic *N*-oxy amides have also been reduced to tri-substituted hydroxylamines with *in situ* generated AlH$_3$ (**Scheme 4f**).[45] However, all

these methods give access to tri-substituted hydroxylamines with diversity on the nitrogen only.
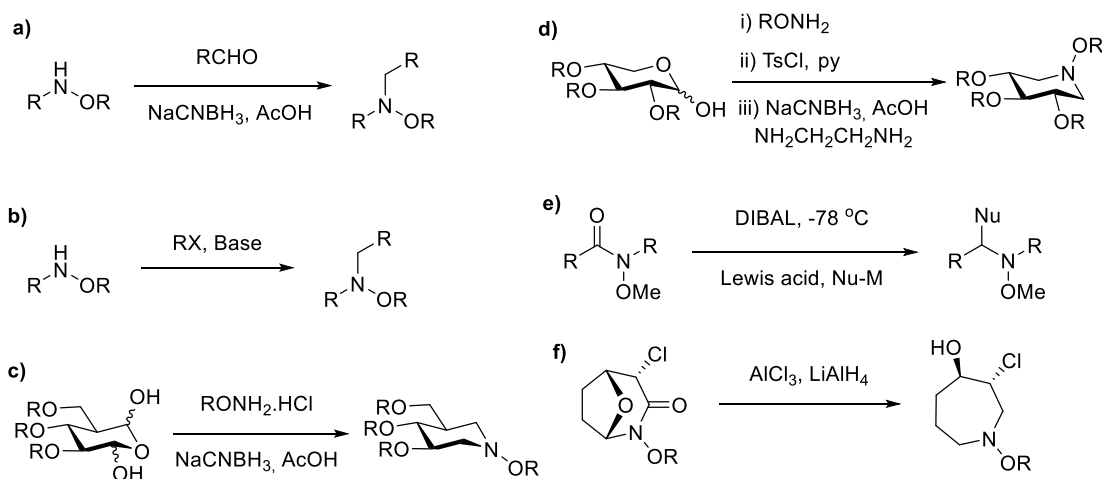
Scheme 4. Synthesis of tri-substituted hydroxylamines.

Alternatively, the approach of *O*-alkylation of *N,N*-disubstituted hydroxylamine has the potential to introduce diversity on the *O*-substituent. However, only alkylation with simple and activated electrophiles such as allyl and aryl halides was reported by this route (**Scheme 5**).[41]

R_1 and R_2 = alkyl
R_3 = allyl, benzyl
X = halogen, OTs, OMs
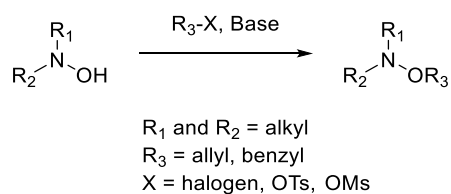
Scheme 5. *O*-Alkylation of *N,N*-disubstituted hydroxylamines.

In conclusion, an efficient method is lacking for the synthesis of tri-substituted hydroxylamines, which particularly, should have the potential of not only the diversification of *O*-substituents but also easy access to these class of compounds, so that the library of compounds with great diversity can be generated for the medicinal chemistry

community. Recognizing this need, an efficient method will be developed to address the existing problem in the synthesis of tri-substituted hydroxylamines.

1.3. Hydrazine

1.3.1. Introduction

Hydrazines represent the class of compounds that are the derivatives of parent hydrazine (H_2N-NH_2). Depending upon the substitution patterns on both nitrogen atoms, they can be classified as mono-substituted, di-substituted, tri-substituted, and tetra-substituted hydrazines (**Figure 9**).
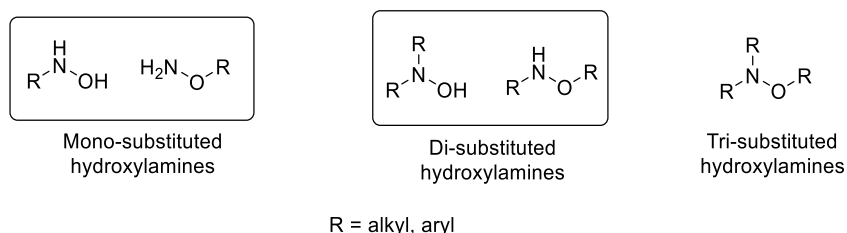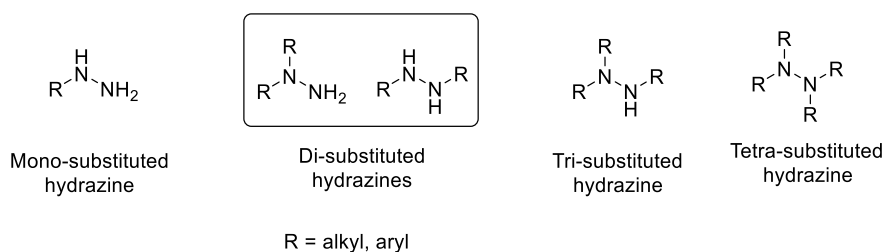

Figure 9. Structures of substituted hydrazines.

If one of the substituents is an acyl group, the compounds are classified as hydrazides, and if one of the nitrogen forms a double bond with sp^2 hybridized carbon atom, the compounds are classified as hydrazones (**Figure 10**). Since N-N bonds are present in both the hydrazones and the hydrazides, they are also considered as hydrazine derivatives, however, they differ significantly from hydrazines in terms of properties. In this thesis, the use of the term hydrazine does not incorporate hydrazones or hydrazides.
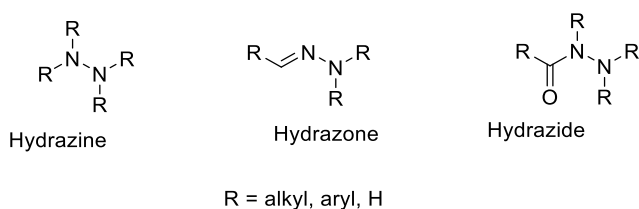
Figure 10. Structures of hydrazines, hydrazones, and hydrazides.

1.3.2. Conformational properties of hydrazines

Alkyl hydrazines are considered bipyramidal, and two diastereomeric staggered conformations, *gauche* (**2**) and *trans* (**3**) (referring to the lone pairs), are possible (**Figure 11**).[46]
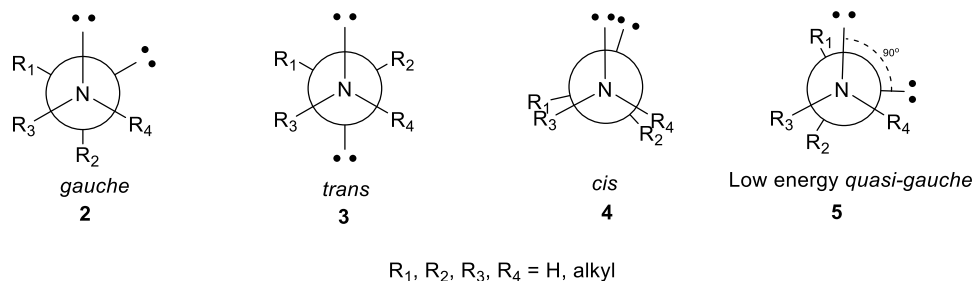
Figure 11. Possible conformations of hydrazine.

The lone pair-lone pair repulsive interaction is a dominant factor in determining the conformation and should be minimized in the most stable one. The low energy lone pair-bond pair and bond pair-bond pair interactions are neglected accordingly. The *trans* conformation (**3**) at first seems to be the more stable one since it offers minimal interaction between the lone pairs. However, the *quasi-gauche* conformation (**5**) (**Figure 11**) with 90° of interorbital angle (Ø) is the most stable conformation according to the spectroscopic studies and molecular orbital calculations.[46] The calculated potential energy function of hydrazine has two energy maxima and one energy minimum (**Figure 12**).[47,48] One maximum corresponds to the *cis* (Ø = 0°) conformation (**4**) and the other to the *trans* (Ø = 180°) conformation (**3**) (**Figure 11**). The energy minimum is the *quasi-gauche* (Ø = 90°) conformation (**5**).
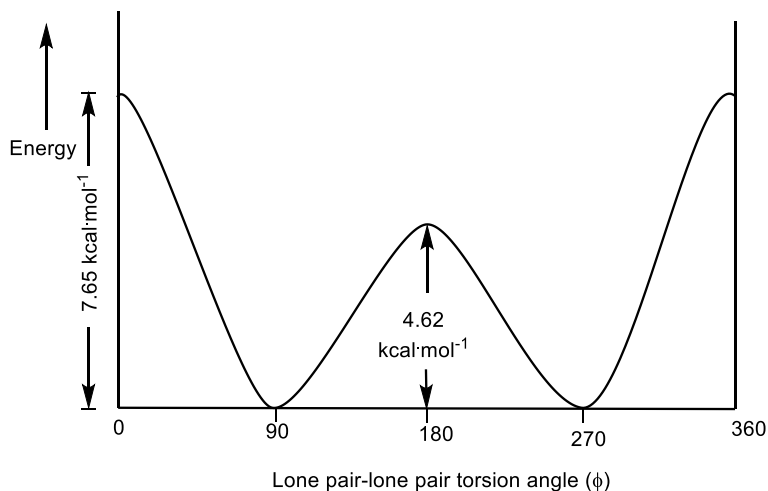
Figure 12. The torsional potential function of $H_2N\text{-}NH_2$.

Several interpretations have been proposed to explain why the *trans* conformation with the least lone pair-lone pair interaction is not adopted by the system.[46-54] The most conceptual and qualitative interpretation[46] is the interaction between the lobes of non-bonding orbitals of two nitrogen atoms is minimal at 90º, while it is maximum at 180º (**Figure 13**). Therefore, two maxima and one minimum are observed in the torsional potential function of hydrazine.

$\phi = 90^\text{o}$ $\phi = 180^\text{o}$

Figure 13. Interactions of non-bonding orbitals of nitrogen atoms at Ø = 90º and 180º.

The interconversion of conformations about the N-N bond is a common phenomenon in hydrazines and is a result of the interplay of internal N-N bond rotations and/or pyramidal nitrogen inversions. The inversion barrier for the parent hydrazine ($H_2N\text{-}NH_2$) is about 7.5 kcal·mol^{-1}.[55] The experimentally calculated barriers for either rotations or inversions in acyclic tetra-alkyl hydrazines range from 5.1 to 11.2 kcal.mol^{-1} depending

upon the nature of alkyl substituents.[20] Though the *quasi-gauche* (Ø = 90°) conformation is the stable conformation, the staggered *gauche* (Ø = 60°) conformation will be used to describe the inversion and rotation processes here after for convenience.

The conformational changes of hydrazine by inversion processes are described in **Scheme 6**.[20] In hydrazines, two distinct types of N-inversion are recognized,[46] which are named as non-eclipsing inversion (non-passing) and eclipsing inversion (passing inversion). The non-eclipsing inversion is a fast and low energy process without eclipsing of substituents. A *gauche* conformation (**6**) leads to another *gauche* conformation (**7**) via a non-eclipsing inversion and the barrier to inversion is around 7.5 kcal·mol^{-1}. The eclipsing inversion, on the other hand, is a high energy process with eclipsing of substituents. A *gauche* conformation (**6**) leads to a *trans* conformation (**8**) via eclipsing inversion process and the barrier to inversion is 10.5 kcal·mol^{-1} (7.5 kcal·mol^{-1} for inversion and 3 kcal·mol^{-1} for eclipsing interactions of alkyl groups). However, the barrier to inversion for the reverse process (*trans* to *gauche*) is 3 kcal·mol^{-1} smaller than the forward process (*gauche* to *trans*).

Non-eclipsing N-inversion
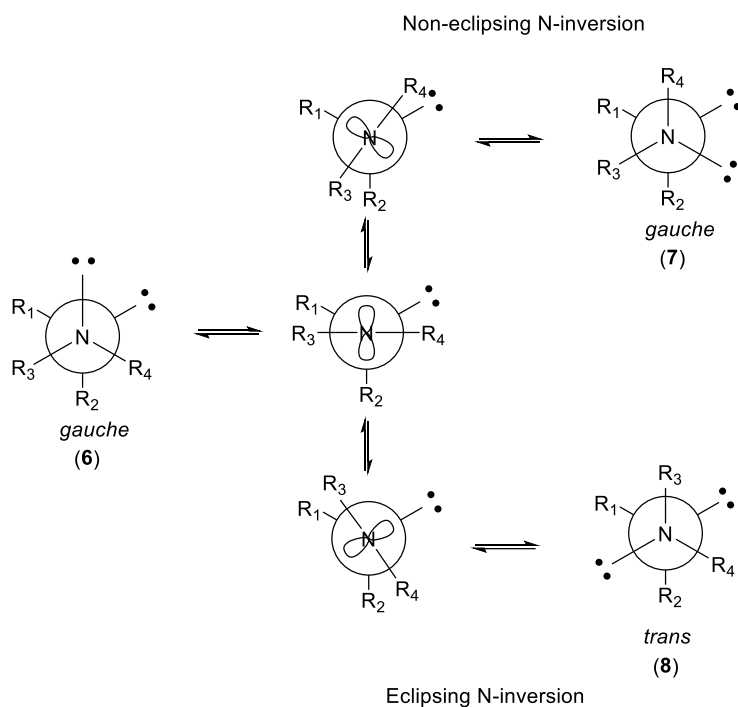
Scheme 6. Diagram showing two different types of N-inversions.

The conformational changes of hydrazine by rotation are described in **Scheme 7**.[20] In an acyclic hydrazine two distinct rotational processes are recognized.[46] The first one is the low energy rotational process that involves eclipsing of one pair of substituents. The conversion of *gauche* (**9**) to *trans* (**10**) conformation is a result of 120° torsional rotation and is a low energy rotational process with an energy barrier of 6-7 kcal·mol⁻¹. However, the barrier to the rotation for reverse transformation (*trans* to *gauche*) is lower by 3 kcal·mol⁻¹ than for the forward transformation (*gauche* to *trans*). The second one is the high energy rotational process that involves eclipsing of two pairs of substituents and two lone pairs. This process occurs in only the *gauche* conformation and leads to another *gauche* conformation. The energy barrier for this type of transformation is around 16 kcal·mol⁻¹ (10 kcal·mol⁻¹ for eclipsing lone pairs, and 6 kcal·mol⁻¹ for two eclipsed interactions of alkyl substituents).
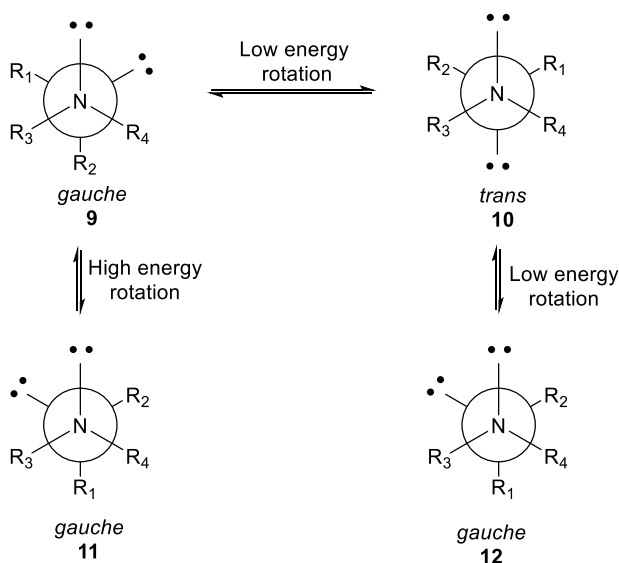
Scheme 7. Diagram showing two types of N-N bond rotations.

1.3.3. **Hydrazines in medicinal chemistry**

N-N bond motifs are relatively scarce in natural products.[56-58] Nevertheless, a survey from Sperry and Blair in 2013[57] showed that over 200 natural products containing the N-N bond motif are known including hydrazines, hydrazones, hydrazides, and many other related functionalities. However, only four of them have the simple hydrazine functionality namely *N*-amino-D-proline, munroniamide, braznitidumine, and ostrerine A (**Figure 14**). Ostrerine A, which was isolated from a mollusk, is the only compound belonging to the tetra-substituted hydrazine class and biological activity of the molecule is still unknown.

N-amino-D-proline

munroniamide

braznitidumine

Ostrerine A
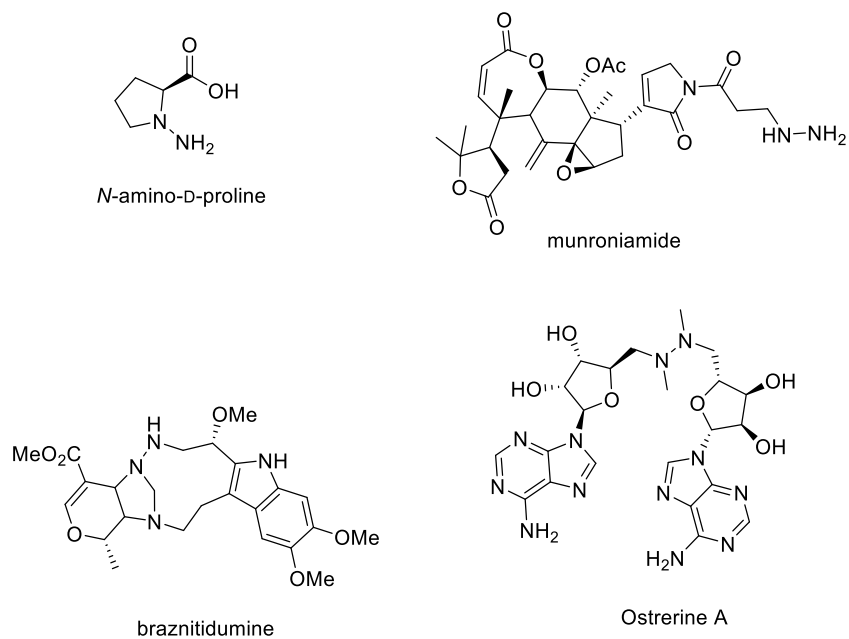
Figure 14. Natural products with the hydrazine moiety.

Recently, Zhang and Hertweck have independently isolated dixiamycins A and B (**Figure 15**), the atropisomeric indoloterpenoid natural products.[59,60] These compounds are the N-N linked dimers of an antibacterial natural product xiamycin A and were found to be more potent than xiamycin A itself (**Figure 15**).[59,60]

Xiamycin A
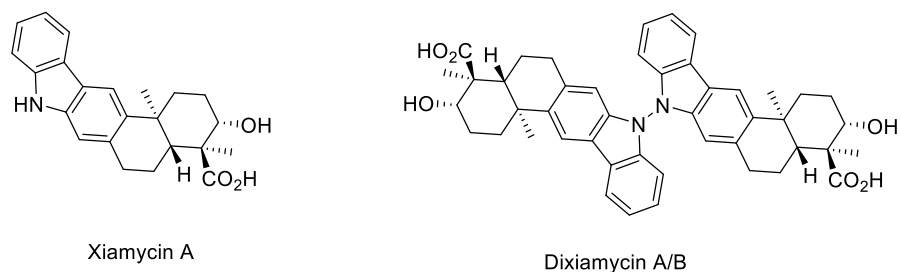
Dixiamycin A/B

Figure 15. Structures of xiamycin A and dixiamycins A and B.

Hydrazine derivatives are an important class of compounds in medicinal chemistry as they are present in several clinical therapeutics. Some of the representative examples are listed in **Figure 16**. Anti-depressant phenelzine[61] and anti-hypertensive agent hydralazine[62] are examples of mono-substituted hydrazines. Di-substituted hydrazine

procarbazine is used to treat Hodgkin's disease, melanoma, and bronchogenic carcinoma as a part of a chemotherapeutic cocktail.[61] Anti-depressant isocarboxazid and anti-tuberculosis antibiotic isoniazid belong to hydrazide class of compounds.[61] Anti-cancer compound PAC-1 is a hydrazone that selectively induces apoptosis in cancerous cells.[63] However, no therapeutic agents with a tetra-substituted hydrazine moiety have been reported to date.
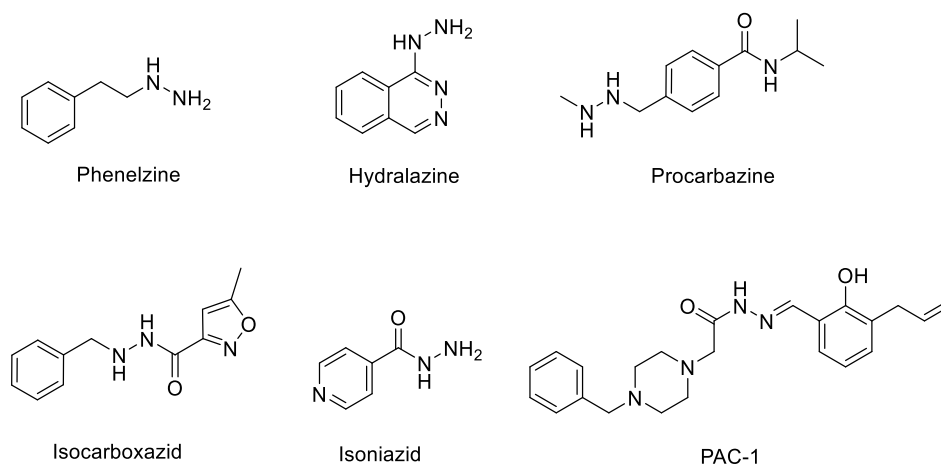
Figure 16. Structures of therapeutic hydrazines and hydrazides.

Although the hydrazine moiety is common in therapeutic agents and has several pharmacological applications in treating pathological diseases, their clinical use is limited due to their toxicity.[34,61] Several hydrazine containing drugs were discontinued in the last decades based on their toxic side effects like hepatotoxicity, induction of systemic lupus erythematosus, and cancinogenesis.[61] Octamoxin, pheniprazine, phenoxypropazine, and safrazine are some of the antidepressants which were withdrawn from the market owing to hepatotoxicity concerns (**Figure 17**).[64] In addition to pharmaceutical agents, toxicity was also observed with several other hydrazine derivatives.[65,66]
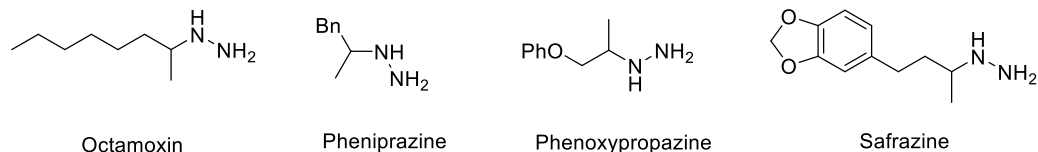
Figure 17. Structures of discontinued therapeutic hydrazines.

The toxicity of the hydrazine derivatives is attributable to the alkylation of DNA by the reactive intermediates like carbocations, carbon-centered radicals, and reactive oxygen species that are generated during the enzyme catalyzed metabolism process.[34,61,66] The metabolisms of mono-substituted and di-substituted derivatives are catalyzed by several enzyme systems like cytochrome P450, horseradish peroxidase (HRP), hepatocytes, microsomes, oxyhemoglobin (HbO_2), myeloperoxidase, and neutrophils (PMNs). **Scheme 8** describes the common mechanism for the formation of the reactive species from *N,N*-disubstituted hydrazines. The carbocations and the alkyl radicals are formed from two distinct pathways (path a and b respectively) after oxidation of hydrazine-derived azo compounds. The so-formed reactive intermediates induce carcinogenesis by alkylating DNA bases.
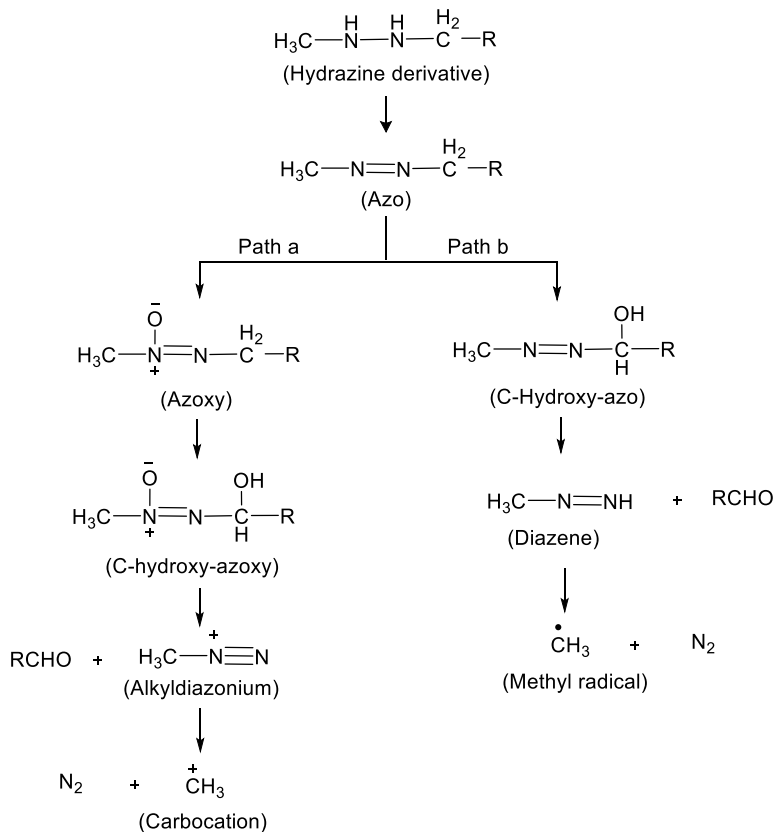
Scheme 8. Metabolic transformation of di-substituted hydrazines.

In case of mono-arylhydrazines, several possible metabolic pathways have been reported.[66] The arenediazonium ion is the reactive intermediate that is supposed to react directly with DNA bases, such as adenine, to generate aryl triazenes (**Scheme 9**).[67] This ion can also react with the DNA backbone to cleave the strand.[66] Alternatively, the diazonium ion can reduce to an alkyl radical which in turn may react with purines (**Scheme 9**) or cleave the DNA strand.[66,68-73]
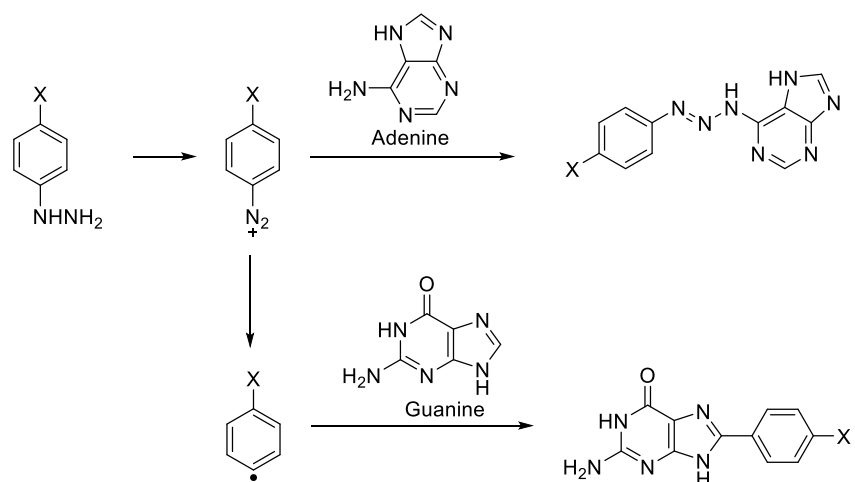
Scheme 9. Metabolic transformation in mono-aryl hydrazines.

Because of these observed toxicities together with the observance of false positives from hydrazine derivatives in the screening process, medicinal chemists have classified hydrazines as undesirable moieties, and they are currently excluded from compound collections.[6,31] However, the exclusion of the whole hydrazine class of compounds based upon the negative results of some compounds is arbitrary. In fact, several therapeutic agents in the market still contain hydrazine moieties and are non-toxic. More importantly, the toxicities are reported only for the mono-substituted, and the di-substituted hydrazines and the key intermediates for the formation of the reactive species are azo-compounds or diazonium ions. However, the formation of these key intermediates is not possible from tetra-substituted hydrazines, which might be one of the reasons why toxicity reports for the tetra-substituted hydrazines are not available. In addition, the false positives in the screening process result from the attack of proteins on electrophilic hydrazine derivatives like hydrazides and *N*-sulfonamides. In contrast, tetra-substituted hydrazines are not electrophilic like their lower congeners. Therefore, exclusion of a whole class of compounds, more importantly, tetra-substituted hydrazines,

from compound collection based on the few toxicity and false positive reports, is reducing the chance of finding quality hits in the drug discovery process. Tetra-substituted hydrazines, in particular, have the potential to enrich compound collections without additional synthetic and stereogenic complexity because of their low basicity and low inversion barriers.

1.3.4. Synthesis of tetra-substituted hydrazines

The development of synthetic routes for substituted hydrazines is an attractive area in synthetic organic chemistry due to their importance in pharmaceutical, agrochemical, polymer, and dye industries and as precursors for organic synthesis.[74] An ideal route for the synthesis of all kinds of substituted hydrazines is the reaction between two amines forming an N-N bond (**Scheme 10**). Since thousands of amines are available commercially, this route would have a huge potential for generating a large library of compounds with diversity. In the flip side, success by this route is considered challenging since forming a heteroatom-heteroatom bond itself remains a challenging goal in the synthetic organic chemistry. Despite the challenge of forming an N-N bond, a few aminations of amines with aminating reagents like chloroamines and sulfonate derivatives of hydroxylamines are documented and are used in laboratory and industrial preparations.[75,76] However, the scope of those methods is narrow due to the limited substrate scope and poor yields. An alternative route is the alkylation or reductive amination of parent hydrazines. However, multiple steps are required to access tri and tetra-substituted hydrazines by this route. Since the synthesis of mono, di, and tri-substituted hydrazines are required as a prelude to the synthesis of tetra-substituted

hydrazines, the available methods for all classes of substituted hydrazines will be discussed briefly including aminations of amines.
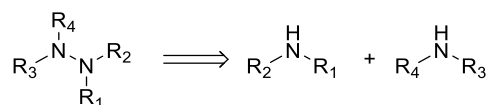
Scheme 10. Ideal route for the synthesis of hydrazines.

1.3.4.1. From hydrazines and their derivaties

Several synthetic methods have been reported aiming at mono-substituted hydrazines.[77] The direct mono-alkylation approach of a free hydrazine, although seems practical, is difficult to control and delivers a mixture of mono-substituted product, over-alkylation, and starting material.[76,77] Therefore, alkylation of *N*-protected hydrazines, followed by deprotection, became the method of choice to access mono-subsituted hydrazines (**Scheme 11a)**.[75] Alternatively, reduction of hydrazones and hydrazides with suitable reducing agents like NaBH$_4$, NaCNBH$_3$, and LiAlH$_4$ was also found to be practical and efficient methods (**Scheme 11b and 11c)**.[75-78]
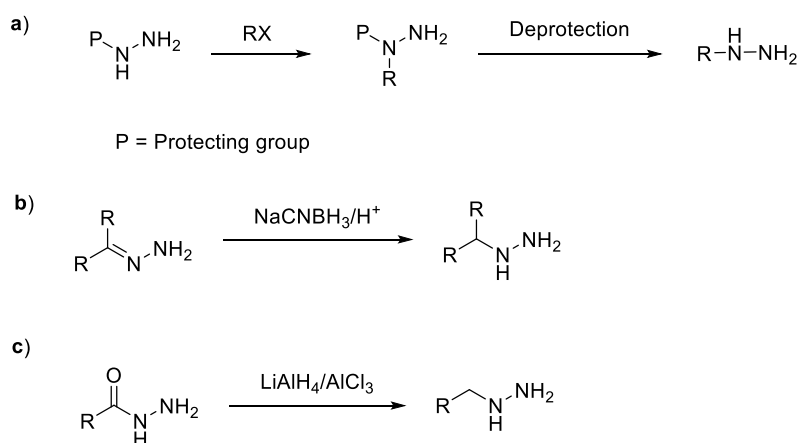
Scheme 11. Synthesis of mono-substituted hydrazines.

The di-substituted hydrazines can be classified into two distinct types namely *N,N-*disubstituted hydrazines (also known as 1,1-disubstituted or unsym-disubstituted

hydrazines) and *N,N*-disubstituted hydrazines (also known as 1,2-disubstituted or sym-disubstituted hydrazines).[76,77] Direct alkylation of mono-substituted hydrazines was generally used to afford *N,N*-disubstituted hydrazines (**Scheme 12a**), whereas strategic protection of the mono-substituted precursor before alkylation and deprotection after alkylation are necessary to generate the *N,N'*-disubstituted hydrazines (**Scheme 12b**).[75,77] Furthermore, the *N,N*-disubstituted hydrazines were also accessed from reduction of hydrazones (**Scheme 12c**) and *N,N'*-diacylhydrazines (hydrazides) (**Scheme 12c**) under similar reduction conditions to those used in reduction approaches for the mono-substituted hydrazines.[75,77,78]
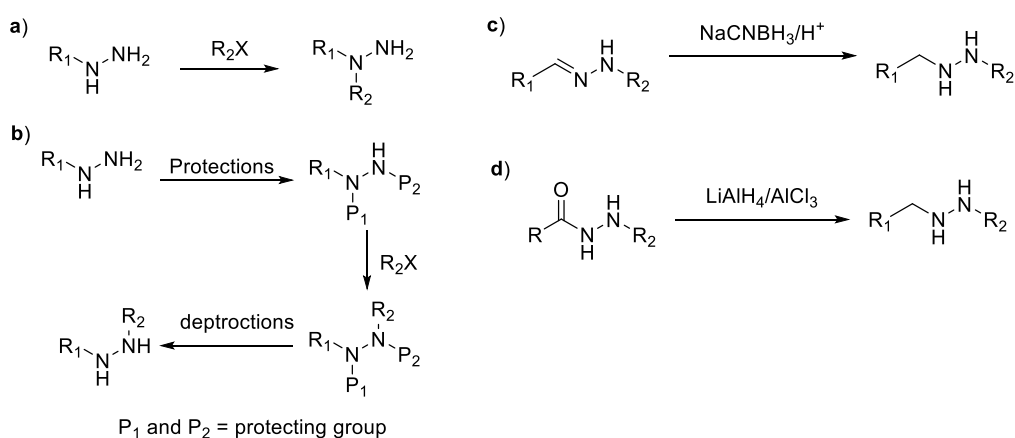
Scheme 12. Synthesis of di-substituted hydrazines.

Similarly, tri and tetra-substituted hydrazines were also prepared from the direct alkylation of di-substituted hydrazines, mostly with activated alkyl halides and alkenes (Michael addition).[77] A recent approach of controlled alkylations of di-anions of *N,N'*-disubstituted hydrazines to generate tri and tetra-substituted hydrazines is a noteworthy example of this kind (**Scheme 13a** and **13b**).[74,79,80] The tri-substituted hydrazines also result from the transformation of *N,N*-disubstituted hydrazines to hydrazone or hydrazide derivatives, followed by reduction (**Scheme 13c** and **13d**).[77] Some of tetra-

alkylhydrazines were also prepared by the sequence of hydrazone formation from *N,N*-disubstituted hydrazines and reduction by NaCNBH$_3$ in the presence of formaldehyde (**Scheme 13c**).[75,77] Reduction of tetra-substituted hydrazides with powerful reducing agents like diborane or LiAlH$_4$ is a common method used in the preparation of simple to complex tetra-substituted hydrazines.[75-77,81]
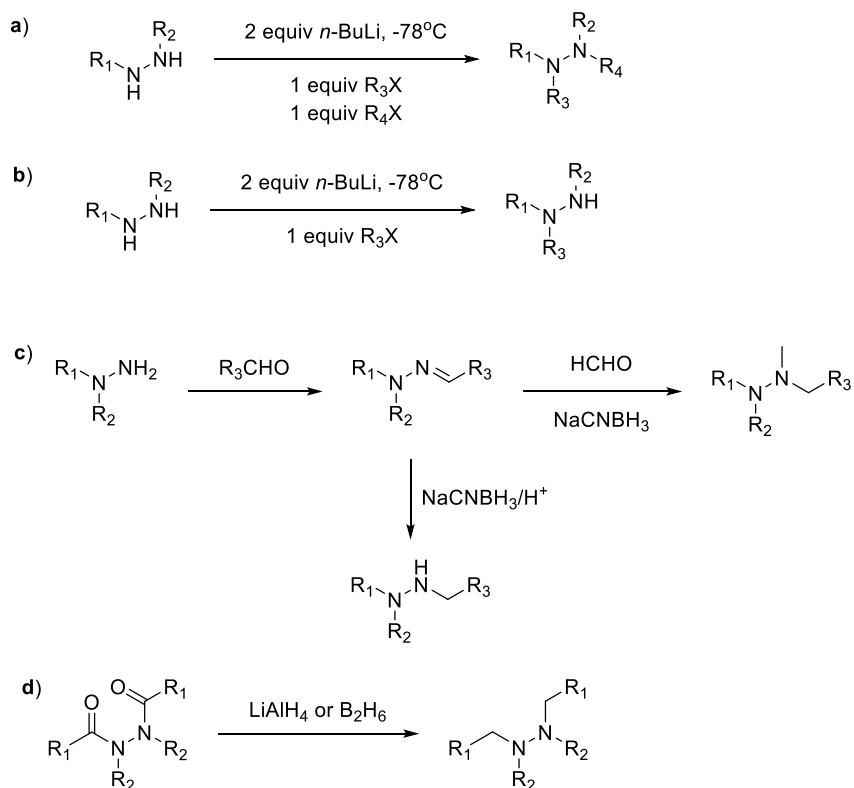
Scheme 13. Synthesis of tri and tetra-substituted hydrazines.

1.3.4.2. From direct amination of amines

Several N-N linked dimers of carbazole derivatives have been generated by electrochemical oxidation via nitrogen centered radical intermediates (**Scheme 14a**).[82] However, this method is limited to symmetric hydrazines only. The reactions of primary and secondary amines with chloroamine[83-85] or hydroxylamine-*O*-sulfonic acid[76,86,87] are useful methods for preparing mono-substituted and *N,N*-disubstituted hydrazines

(**Scheme 14b**). For example, the reaction of dimethylamine with chloroamine is an industrial preparation of *N,N*-dimethylhydrazine (rocket fuel).[75] The *N,N'*-disubstituted hydrazines were prepared from the reaction of *N*-chloro derivatives of primary amines with azomethines followed by the hydrolysis (**Scheme 14c**).[75,77] Moreover, a series of aminating agents based on *N*-protected oxaziridines with various substituents on their carbons has been developed for electrophilic amination of amines (**Scheme 14d**).[75,88] This method was found to be efficient for the synthesis of *N,N*-disubstituted hydrazines only. Several other aminating agents based on hydroxylamine sulfonates can be accessed commercially or synthetically[89] and have the potential to form hydrazines directly from amines.
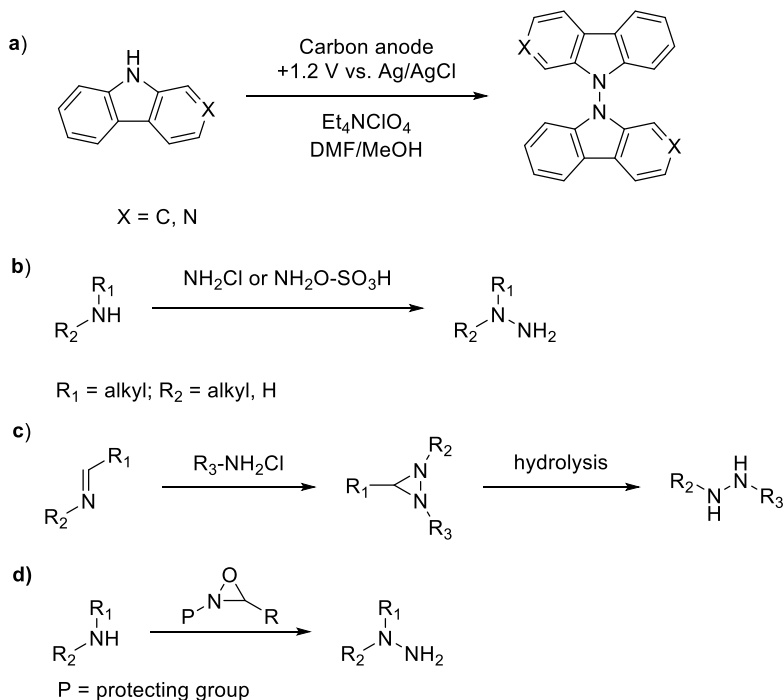
Scheme 14. Synthesis of substituted hydrazines by direct aminations.

In conclusion, *N*-alkylations of protected hydrazines, reductions of hydrazones and hydrazides, and aminations of amines with aminating reagents are some of the important

and widely used methods in the synthesis of substituted hydrazines.[77] Several other methods including transition metal catalyzed reactions are also reported,[90-93] however, short and efficient entries to substituted hydrazines are always in demand.

1.4. Designing hydroxylamine and hydrazine analogs of kalkitoxin

1.4.1. Discovery of kalkitoxin and its biological activitites

Kalkitoxin (**13**) is a natural stretch-peptide that was discovered by Gerwick, Shioiri, and co-workers as a secondary metabolite of *Lyngbya majuscule*, a marine cyanobacterium.[94,95] It was isolated from an extract of *L. majuscule* by the brine shrimp and gold fish toxicity assay guided fractionation process and was disclosed as ichthyotoxic against common gold fish (*Carassius auratus*, LC_{50} 700 nM) and potently toxic against brine shrimp (*Artemia salina*, LC_{50} 170 nm).[95] The potencies of kalkitoxin were also observed in the inhibition of the cell division process in a fertilized sea urchin embryo assay (IC_{50} ~50 nM), in suppression of inflammation as measured in an inflammatory disease model that measures IL-1β-induced $sPLA_2$ secretion from HepG2 cells (IC_{50} 27 nM), and in blockage of voltage sensitive Na^+ channel in mouse neuro-2a cells (EC_{50} 1 nM).[95-98] In addition, an exceptional concentration-dependent neurotoxicity (LC_{50} 3.86 nM) was observed with kalkitoxin when tested in a primary cell culture of rat cerebral granule neurons whose the effects were prevented with non-competitive *N*-methyl-D-aspartate (NMDA) receptor antagonists.[94] Furthermore, The potent anti-cancer property of kalkitoxin was revealed by its excellent cytotoxicity (IC_{50} 1 nM) measured against the human colorectal cancer cell line (HCT-116).[99] In a recent mechanistic study by Nagle, Zhou, and co-workers,[100] kalkitoxin was found to inhibit hypoxia-induced activation of HIF-1 in T47D breast tumor cells (IC_{50} 5.6 nM) selectively by suppressing

mitochondrial oxygen consumption at electron transport chain (ETC) complex I (NADH-ubiquinone oxidoreductase). Further, kalkitoxin was also found to inhibit angiogenesis by blocking the induction of angiogenic factors like vascular endothelial growth factors (VEGF) in tumor cells.
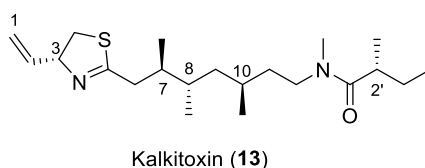
Kalkitoxin (**13**)

Figure 18. Structure of kalkitoxin.

The structure of kalkitoxin is as interesting as its biology. Gerwick, Shioiri and co-workers carried out the total synthesis of natural kalkitoxin and its possible stereoisomers in order to determine the configuration.[95,101] After comparing their NMR spectra and on the basis of the asymmetric synthesis, the absolute configuration of natural (+)-kalkitoxin was assigned as (3*R*,7*R*,8*S*,10*S*,2'*R*). The structure of kalkitoxin includes a 2,4-disubstituted thiazoline with one substituent being an unsaturated side chain and the other a stretch-peptide side chain. The arrangement of the three methyl substituents in the 1,2,4-*anti*,*anti* fashion is the most significant and structurally attractive feature of the stretch-peptide side chain.

1.4.2. Total syntheses of kalkitoxin

With its exceptional biological activities and three methyl substituents in 1,2,4-*anti*,*anti* fashion in its stretch-peptide side chain, kalkitoxin became an attractive target for the synthetic chemists. As a result, five total syntheses have been published to date.

Gerwick, Shioiri, and co-workers reported a first total synthesis of kalkitoxin in 2000 (**Scheme 15**), while determining its absolute configuration.[95] The synthesis commenced with deoxygenation of a known compound **14,**[102] after which the resulting alkene was

subjected to hydroboration, followed by azide **15** formation. The azide **15** was then reduced to an amine, coupled with (*R*)-methylbutyric acid (**16**), and methylated to provide amide **17**. The subsequent *O*-desilylation and oxidation resulted in an aldehyde, which was further subjected to the Horner-Emmons reaction with **18** in order to afford the precursor to asymmetric conjugate addition. Hruby's method[103] of conjugate addition resulted in the adduct **19** with the required 1,2,4-*anti*,*anti* methyl sequence as a single isomer. Conversion of the chiral auxiliary to the acid functionality and coupling with (*R*)-amino alcohol **20** delivered di-amide **21**. Finally, Wipf's oxazoline-thiazoline interconversion reaction sequence[104] was utilized in order to get kalkitoxin (**13**). In this process the amide **21** was first cyclized to a 2-oxazoline in the presence of DAST, then treatment with hydrogen sulfide afforded the thioamide. DAST was again used for the second cyclization to deliver the targeted kalkitoxin (**13**). In conclusion, the synthesis of kalkitoxin was accomplished in 17 linear steps, starting from a known compound **14** in 9% of overall yield. The 1,2,4-*anti*,*anti* sequence was achieved by 1,4-conjugate addition with the aid of a chiral auxiliary and the thiazoline ring was introduced by Wipf's oxazoline-thiazoline interconversion protocol.
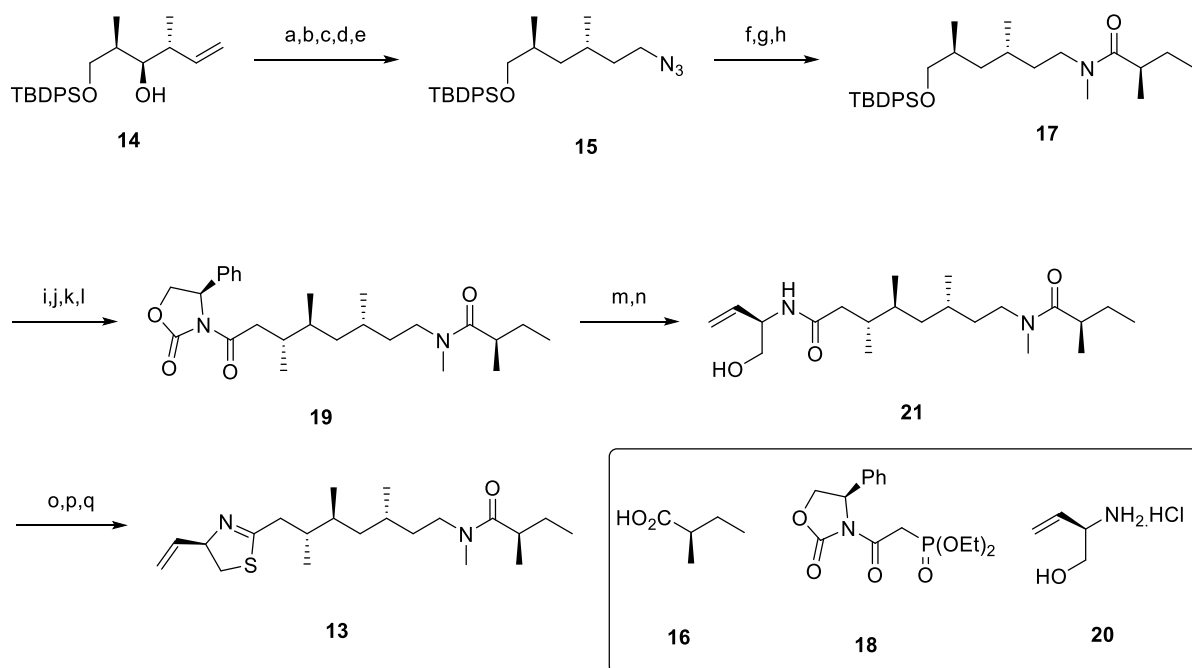
Scheme 15. Total synthesis of kalkitoxin by Gerwick, Shioiri, and co-workers. a) *n*-BuLi, *p*TsCl. b) LiAlH$_4$. c) 9-BBN, ultrasound; NaOH, H$_2$O$_2$. d) MsCl, Et$_3$N, DMAP. e) NaN$_3$, 55 °C. f) H$_2$ (1 atm) 5% Pd-C. g) **16**, DEPC, Et$_3$N. h) *n*-BuLi, MeI. i) TBAF. j) Py.SO$_3$, Et$_3$N, DMSO. k) **18**, NaHMDS. l) MeMgBr, CuBr.DMS, -30 °C. m) LiOH, H$_2$O$_2$. n) **20**, EDCI, DIPEA, DMAP. o) DAST, -20 °C. p) H$_2$S, Et$_3$N. q) DAST, -20 °C.

The second total synthesis was executed by the White group in 2003 (**Scheme 16**).[99,105] A one pot entry to the 1,2,4-*anti,anti* methyl sequence via asymmetric conjugate addition, followed by in situ enolate alkylation, and a TiCl$_4$ induced thiazoline ring formation were the two key steps of the synthesis. The synthesis started with a three-step reaction sequence involving asymmetric alkylation, reduction, and Appel reaction to generate alkyl bromide **23** from literature molecule **22**[106] as a starting material. After transformation of **23** to an organocopper species, it was treated with (*S*)-*N*-(*trans*-crotonyl)-4-phenyloxazolidin-2-one (**24**) and the so-obtained *anti*-1,3-dimethyl enolate was trapped *in situ* with methyl iodide to deliver a mixture of **25** (with the desired 1,2,4-

anti,*anti* configuration) and **26** favoring **25** (3.6:1). After hydrogenation of **25**, the resulting alcohol side chain was extended to amide **27** by Swern oxidation, reductive amination, and coupling with (*R*)-methylbutyric acid (**16**). The chiral auxiliary was removed by reduction and the resulting alcohol was subjected to Swern oxidation, followed by homologation, to give a mixture of alkenes (*E*/*Z*) **28** via the Wittig reaction. The mixture **28** was then hydrolyzed to an aldehyde and oxidized to an acid **29**. Next, amine **32**, the coupling partner of the acid **29**, was synthesized from L-cysteine (**30**) in 5 steps. First, the free thiol and amine groups of **30** were protected as a benzyl thioether and a carbamate, respectively, and the remaining free acid function was transformed to Weinreb amide **31**. Then reduction, Wittig reaction, and selective amine deprotection delivered amine **32**. The resulting amine **32** was coupled with acid **29** and the so-formed thioamide **33** was subjected to Birch reduction conditions to get thioamide **34** with a free thiol group. Finally, the TiCl₄-mediated cyclization of **34** delivered the targeted kalkitoxin. In summary, kalkitoxin was synthesized in 16 linear steps starting from a literature amide **22** in 3% overall yield.
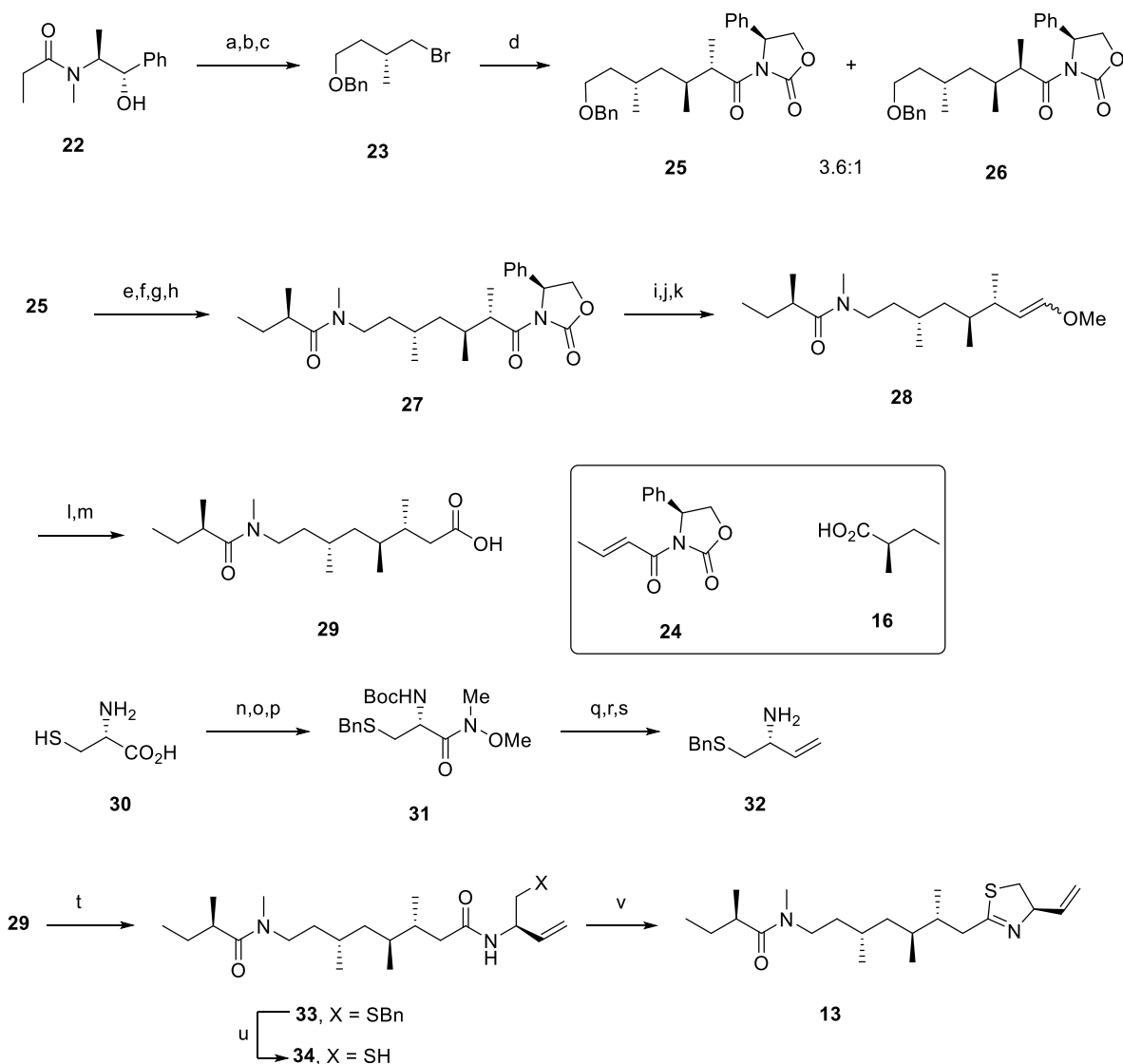
Scheme 16. Total synthesis of kalkitoxin by White *et al.* a) LDA, LiCl, 2-iodoethyl benzyl ether, 0 °C, 98%. b) LDA, BH$_3$.NH$_3$, 0 °C, 90%. c) Ph$_3$P, NBS, 74%. d) Mg; CuBr.SMe$_2$; **24**; MeI, 64%. e) H$_2$ (1 atm), 15% Pd-C, 100%. f) (COCl)$_2$, DMSO, Et$_3$N, 85%. g) MeNH$_2$.HCl, NaCNBH$_3$, 81%. h) **16**, EDCI, HOAt, 84%. i) LiBH$_4$, 80%. j) (COCl)$_2$, DMSO, Et$_3$N. k) Ph$_3$PCH$_2$OMeCl, *n*-BuLi, 52% (2 steps) l) HCl, MeCN. m) NaClO$_2$, NaH$_2$PO$_4$, 2-methyl-2-butene, 96% (2 steps). n) BnBr, NaOH, EtOH, 94%. o) Boc$_2$O, Et$_3$N, 99%. p) MeNHOMe.HCl, EDCI, HOBt, 84%. q) LiAlH$_4$, 91%. r) Ph$_3$PCH$_3$Br, *n*-BuLi, 55%. s) TFA, no purification. t) HATU, DIPEA, **32**, 93% u) Na, NH$_3$, 87%. v) TiCl$_4$, 67%.

The next total synthesis was reported by the Shioiri group in 2004,[101] and was similar to their first total synthesis except for the installation sequence of some of the building blocks in the developing stretch-peptide chain. In 2012, Matsuda and co-workers also reported a total synthesis,[107] in which the stretch-peptide chain was synthesized following the route of the Gerwick and Shioiri synthesis, and the thiazoline ring was installed following White's protocol.

More recently, Aggarwal and co-workers have reported the shortest route to date for kalkitoxin using an iterative synthesis approach (**Scheme 17**).[108] The most attractive feature of this synthesis was the stereoselective achievement of amide **41** from boronic ester **35** in a few steps with a single purification process. First, the boronic ester **35** was converted to a core aliphatic chain **38** by a reaction sequence of two consecutive homologations with (*S*)-**36**, a methylene insertion with **37**, another homologation with (*R*)-**36**, and two methylene insertions. The-so obtained boronic ester **38** was treated with lithiated methoxyamide (**39)** and the resulting amine **40** was coupled with (*R*)-methylbutyric acid (**16**) to furnish amide **41**. After methylation of amide **41**, the resulting amide intermediate was converted to acid **29** in the presence of $RuCl_3$ and $NaIO_4$. Unlike the White synthesis, the coupling partner of acid **29** (i.e., **32**) was prepared from butadiene monoepoxide (**43**) as a starting material. An allylic imide **45** was prepared from the reaction of phthalimide and butadiene monoepoxide under Trost's dynamic kinetic asymmetric transformation conditions. The free hydroxyl group was next converted to a thioether via mesylation and subsequent displacement by benzylmercaptan and the resulting thioether was subjected to aminolysis to produce amine **32**. With acid **29** and

amine **32** in hand, the synthesis of kalkitoxin was completed following White's protocol. The total steps for kalkitoxin synthesis were cut down to thirteen in 12% overall yield.
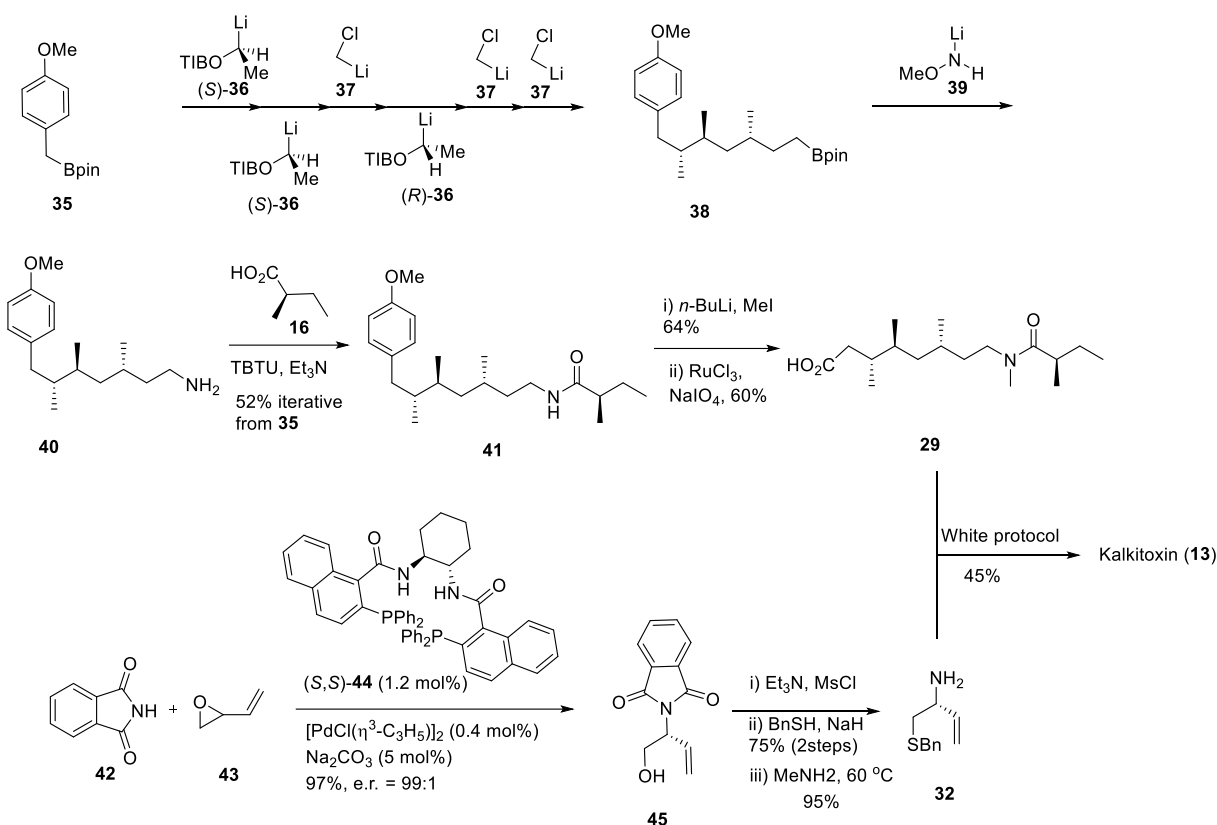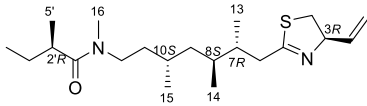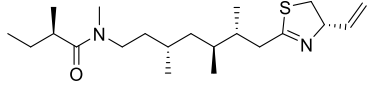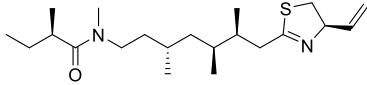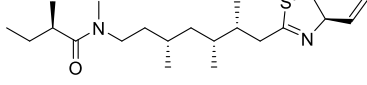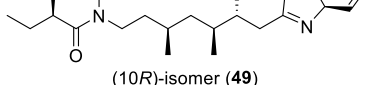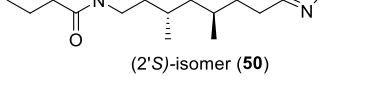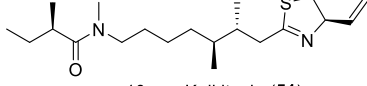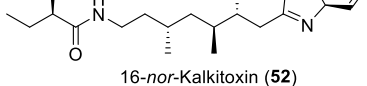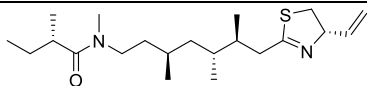
Scheme 17. Total synthesis of kalkitoxin by Aggarwal and co-workers.

1.4.3. SAR studies of kalkitoxin

Several kalkitoxin analogs have been synthesized and screened for their bioactivities in order to understand the role of structural and configurational motifs of kalkitoxin in its observed bioactivity. The structures of these kalkitoxin analogs and their activities against brine shrimp bioassay (LC_{50} data) or against human colon cancer cell line HCT-116 (IC_{50} data) are tabulated in **Table 1**.

Table 1. Biological activities of kalkitoxin and its analogs.

	Kalkitoxin and its isomers	Brine shrimp assay, LC$_{50}$ (nM)	HCT-116 IC$_{50}$ (nM)	Ref.
1	(3R,7R,8S,10S,2'R)-Kalkitoxin (**13**)	170-180	1	95,99,101,107
2	(3S)-isomer (**46**)	15000	-	107
3	(7S)-isomer (**47**)	3600	-	107
4	(8R)-isomer (**48**)	1700	-	107
5	(10R)-isomer (**49**)	620	-	107
6	(2'S)-isomer (**50**)	550	-	101
7	10-nor-Kalkitoxin (**51**)	1800	-	107
8	16-nor-Kalkitoxin (**52**)	Inactive	-	107
9	(3S,7S,8R,10R,2'S)-isomer (**53**)	9300	-	101

10	 (7*S*,8*R*,10*R*,2'*S*)-isomer (**54**)	Inactive	-	101
11	 (7*S*,10*R*,2'*S*)-isomer (**55**)	Inactive	-	101
12	 (3*S*,2'*S*)-isomer (**56**)	1700	-	101
13	 (10*R*,2'*S*)-isomer (**57**)	1100	-	101
14	 **33**	-	400	99
15	 **34**	-	190	99

From these results it is clear that the configuration of the stereogenic center C-3 of kalkitoxin is very important for the activity since the 3*S*-isomer (**46**) of kalkitoxin was found to be ~80 fold less active than kalkitoxin (**Table 1**, entry 2). The activities of 7*S*-isomer (**47**) and 8*R*-isomer (**48**) (**Table 2**, entries 3 and 4) were found to be 20 and 10 fold less than that of kalkitoxin suggesting that the configurations of stereogenic centers C-7 and C-8 in kalkitoxin are also important for the activity. However, the configurations of stereogenic centers C-10 and C-2' in kalkitoxin are comparatively less important for the activity as both the10*R* isomer (**49**) and 2'*S*-isomer (**50**) (**Table 1**, entries 5 and 6) were found to be only about 3 fold less active than kalkitoxin. The isomers **54** and **55** (**Table 1**,

entry 10 and 11) were found to be almost inactive and the enantiomer (**53**) of kalkitoxin (**Table 1**, entry 9) was found to be 50 fold less active than kalkitoxin. Moreover, isomers **56** and **57** (**Table 1**, entry 12 and 13) were also found to be less potent than kalkitoxin. The LC_{50} values for 10-*nor*-kalkitoxin (**51**) and 16-*nor*-kalkitoxin (**52**) suggest that the methyl groups CH_3-15 and CH_3-16 are very important for the activity. White et al. have screened the synthetic precursors of kalkitoxin (**33** and **34**) against human colon cancer cell line (HCT-116) in addition to kalkitoxin (**Table 1,** entries 1, 14, and 15). The precursors **33** and **34** were found to be 400 and 190 fold less potent, respectively than kalkitoxin. Therefore, the thiozoline ring is a very important part of the molecule for the activity. In conclusion, the configurations of all stereogenic centers, methyl groups CH_3-15 and CH_3-16, and thiazoline ring are important for the potency of the kalkitoxin.

1.4.4. Design of hydroxylamine and hydrazine analogs

In order to test the concepts of hydroxylamine and hydrazine analogs, kalkitoxin was chosen as a potential candidate for three reasons. First, kalkitoxin has potent bioactivity as described in **section 1.5.1**. Second, the configurations of stereogenic centers in kalkitoxin are important for the activity as discussed in **section 1.5.3**. And third, kalkitoxin has the potential to serve as a common candidate to test both the hydroxylamine and hydrazine analog concepts since it has two adjacent stereogenic centers.
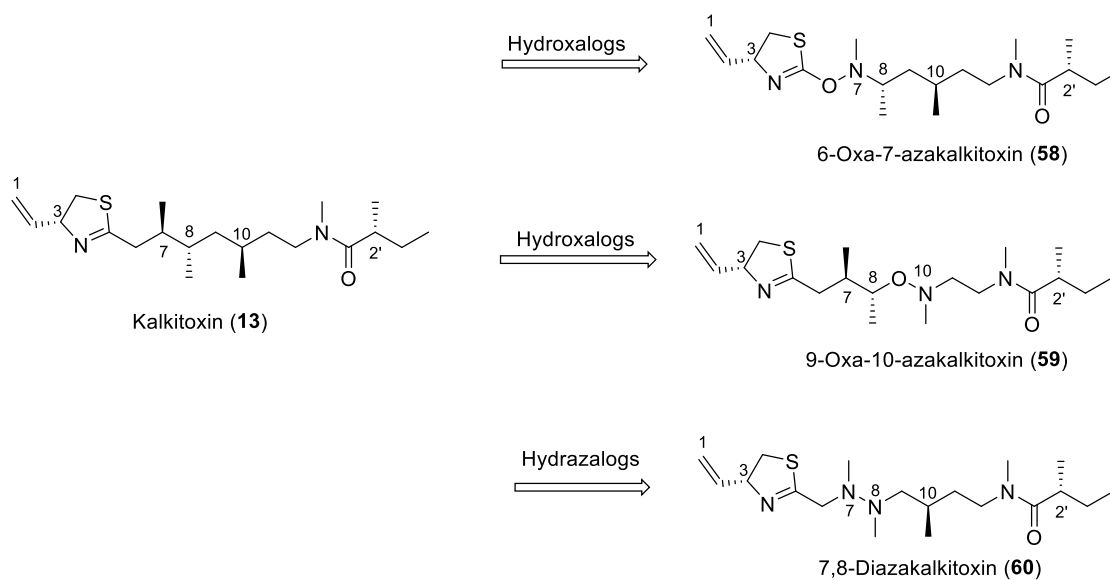
Figure 19. Kalkitoxin and its hydroxalogs and hydrazine analog.

Two hydroxalogs of kalkitoxin were designed, namely, 6-oxa-7-azakalkitoxin (**58**)

and 9-oxa-10-azakalkitoxin (**59**). The former was designed by substituting the C-6 with

an oxygen atom and the stereogenic center C-7 with a nitrogen atom and the latter was

designed by substituting the C-9 with an oxygen atom and the stereogenic center C-10

with a nitrogen atom. The hydrazine analog of kalkitoxin, 7,8-diazakalkitoxin (**60**), was

designed by substituting the stereogenic centers C-7 and C-8 with two nitrogen atoms

thereby introducing the hydrazine moiety. These modifications are intended to reduce the

synthetic as well as the stereogenic complexities due to the rapid inversions at the

nitrogen centers. The synthesis of all three analogs will be executed and the compounds

will be screened against several cancer cell lines in the Valeriote lab at the Henry Ford

Hospital Cancer Center in Detroit.

CHAPTER 2. SYNTHESIS OF *N,N,O*-TRISUBSTITUTED HYDROXYLAMINES

2.1. Design and development of the method

As is described in **section 1.2.4** of **Chapter 1** (**Scheme 3**), an ideal route for the synthesis of tri-substituted hydroxylamies is the N-O bond formation in which commercial amines and alcohols can be utilized to generate a library of compounds. However, owing to the challenges associated with this line of work, chemists have shifted their interest to alternative routes such as *N*-derivatization of *N,O*-disubstituted hydroxylamines and *O*-derivatization of *N,N*-disubstituted hydroxylamines. Several methods for *N*-alkylation have been devised for tri-substituted hydroxylamine synthesis.[18,19,41-43] However, methods for *O*-alkylation of *N,N*-disubstituted hydroxylamine are limited with the exception of *O*-alkylation with simple and activated electrophiles like allyl and aryl halides.[41] In addition to the observed limitation of the substrate scope, the available method for *O*-alkylation also lacks the potential to introduce diversity on the *O*-substituent. To introduce diversity on *O*-substituent, an efficient method for *O*-alkylation of *N,N*-disubstituted hydroxylamines is needed to provide easy access to libraries of tri-substituted hydroxylamines with great diversity.

To solve the existing problem in *O*-alkylation, indirect *O*-alkylation of *N,N*-disubstituted hydroxylamine was designed. Since acylation is much easier than alkylation, *O*-alkylation of *N,N*-disubstituted hydroxylamines was considered analogous to the synthesis of ethers from alcohols by reductive etherification, for which a number of attractive ideas were formulated recently.[109,110] Therefore, by adapting Rychnovsky reductive etherification,[109] acylation of *N,N*-disubstituted hydroxylamines to *N*-acyloxyamines, followed by a two-step method, was considered for the synthesis of tri-

substituted hydroxylamines based upon the stepwise reduction of *N*-acyloxyamines (**Scheme 18**). The *N*-acyloxyamines was first reduced with diisobutylaluminum hydride (DIBAL) at lower temperature and then was treated with acetic anhydride in the presence of pyridine and 4-(dimethylamino)pyridine (DMAP). The resulting *O*-(α-acetoxyalkyl)hydroxylamine was further reduced with triethylsilane in the presence of Lewis acid such as $BF_3.OEt_2$ to provide tri-substituted hydroxylamine.
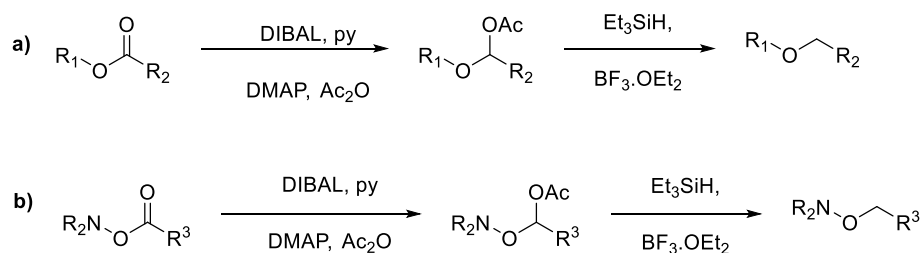
Scheme 18. a) Rychnovsky reductive etherification. b) Synthesis of tri-substituted hydroxylamines from *N*-acyloxyamines.

A possible mechanism for this two-step method is proposed in **Scheme 19**. **Scheme 19a** is the mechanism for the first reduction step, and **Scheme 19b** is the mechanism for the second reduction step. Partial reduction of the *N*-acyloxyamine with DIBAL generates a chelation-stablized tetrahedral intermediate similar to Colby's aluminium complex of hydroxylamine[111] at −78 °C. This tetrahedral intermediate is trapped *in situ* with acetic anhydride in the presence of DMAP generating a more stable *O*-(α-acetoxyalkyl)hydroxylamine. In the second reduction step, an acetoxy anion is released from the *O*-(α-acetoxyalkyl)hydroxylamine with an aid of a Lewis acid and forms an aminoxocarbenium ion intermediate in situ. Donation of hydride from triethylsilane to this intermediate ion generates the tri-substituted hydroxylamine.
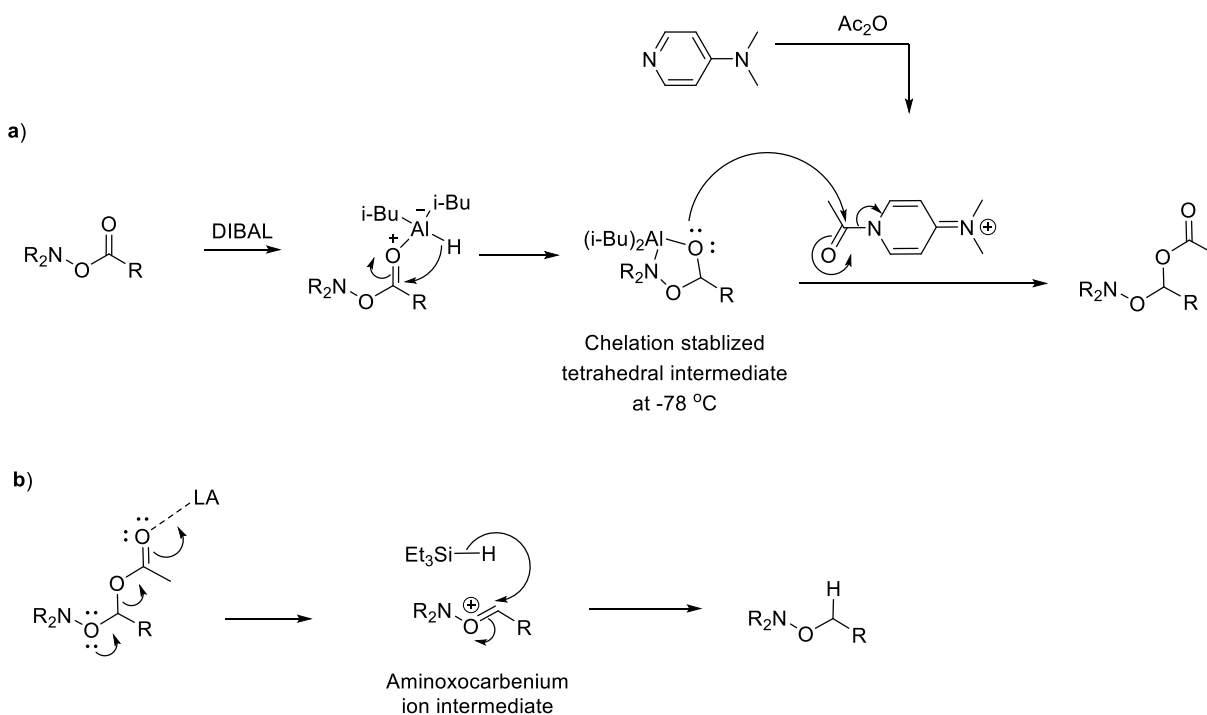
Scheme 19. Proposed mechanism for 2-step reaction in **Scheme 18**.

2.1.1. Synthesis of *N,N*-disubstituted hydroxylamines

A simple protocol to prepare *N,N*-disubstituted hydroxylamines from *N*-(benzoyloxy)amines derived from the reaction of amines with dibenzoyl peroxide was executed (**Table 2**). *N*-(Benzoyloxy)amines (**62a-62e**) were prepared from benzoyl peroxide and commercial secondary amines by adaptation of the Ganem protocol[112] in good yields. On exposure to 2.5 equivalents of DIBAL in ice cold conditions, these *N*-(benzoyloxy)amines were reduced to *N,N*-disubstituted hydroxylamines (**63a-63e**).

Table 2. Synthesis of *N,N*-disubstituted hydroxylamines.

$$R^1R^2NH \xrightarrow[\text{K}_2\text{HPO}_4]{(\text{BzO})_2,} R^1R^2N-OBz \xrightarrow[0\ ^oC]{\text{DIBAL,}} R^1R^2N-OH$$

61a-e   **62a-e**   **63a-e**

Entry	Amine, 61	*N*-Benzoyloxyamine 62, % yield[a]	Hydroxylamine 63, % yield[b]
1	**61a**, $R^1 = R^2$ = Bn	**62a**, 81%	**63a**, 90%
2	**61b**, R^1 =Me, R^2 = Bn	**62b**, 82%	**63b**, 93%
3	**61c**, piperidine	**62c**, 86%	**63c**, 63%
4	**61d**, 4-methylpiperidine	**62d**, 82%	**63d**, 84%
5	**61e**, 4-chloropiperidine	**62e**, 81%	**63e**, 85%

[a] Stirring with (BzO)$_2$ (1.1 equiv) and K$_2$HPO$_4$ (1.5 equiv) in DMF, at rt for 1-22 h. [b] Stirring with DIBAL (2.5 equiv) in CH$_2$Cl$_2$ at 0 ^{o}C for 15 min.
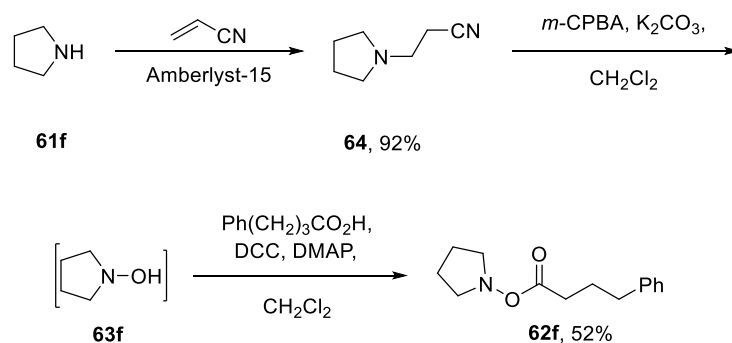
Scheme 20. An alternative synthesis of *N*-acyloxy amines exemplified for *N*-(4-phenylbutyroyl)pyrrolidine.

Alternatively, *N,N*-disubstituted hydroxylamine **63f** was effectively generated from aza-Michael addition of a secondary amine to acrylonitrile, followed by *N*-oxide formation and Cope elimination (**Scheme 20**).[113]   Subsequent treatment of crude **63f** with 4-

phenylbutyric acid in the presence of dicyclohexylcarbodiimide (DCC) and DMAP afforded *N*-acyloxyamine **62f**.

2.1.2. Synthesis of *N*-acyloxyamines

N-acyloxyamines (**62g-62o**) were prepared from *N,N*-disubstituted hydroxylamines (**63a-63f**) and the corresponding carboxylic acids with DCC and DMAP as activating reagents (**Table 3**).[114]

Table 3. Synthesis of *N*-acyloxyamines.

$$R^1R^2N-OH \xrightarrow[\text{DCC, DMAP}^{[a]}]{R^3CO_2H,} R^1R^2N-O-\underset{O}{\overset{O}{C}}-R^3$$

63   **64g-o**

Entry	R^1	R^2	R^3	*N*-Acylhydroxylamine, 62, %yield
1	Bn	Bn	Ph(CH$_2$)$_3$	**62g**, 98%
2	Bn	Bn	C$_{10}$H$_{15}$	**62h**, 72%
3	Bn	Bn	*t*BuCH$_2$	**62i**, 63%
4	Bn	Me	Ph(CH$_2$)$_4$	**62j**, 93%
5	-(CH$_2$)$_5$-		Ph(CH$_2$)$_3$	**62k**, 93%
6	-(CH$_2$)$_2$CHMe(CH$_2$)$_2$-		Ph(CH$_2$)$_3$	**62l**, 91%
7	-(CH$_2$)$_2$CHCl (CH$_2$)$_2$-		Ph(CH$_2$)$_3$	**62m**, 80%
8	Bn	Bn	2-C$_{10}$H$_7$OCH$_2$	**62n**, 89%
9	Bn	Bn	3-C$_5$H$_4$N	**62o**, 97%

[a] R^{3}CO$_2$H (1.25-2.5 equiv), DCC (1.25-2.5 equiv), DMAP (0.2 equiv), CH$_2$Cl$_2$, RT, 0.5-30 h.

2.1.3. Synthesis of *N,N,O*-trisubstituted hydroxylamines

With a series of *N*-acyloxyamines in hand, the efficiency of the two-step by method for the synthesis of tri-substituted hydroxylamines (**Table 4**) was investigated adapting Rychnovsky's protocol[109] for the synthesis of ethers from esters. The *N*-acyloxyamines were reduced with DIBAL at −78 °C, and the tetrahedral intermediates were trapped *in situ* as *O*-(α-acetoxyalkyl)hydroxylamines with acetic anhydride in the presence of pyridine and DMAP, followed by warming to 0 °C. A diverse range of *O*-(α-acetoxyalkyl)hydroxylamines **65** were obtained in good to excellent yields. The reaction is quite general, as exemplified through a diverse set of substrates with alkyl and aryl groups in both nitrogen and oxygen substituents. In addition, substrates with branching at the α- and β-positions of the acyl groups (**62h** and **62i**) were also compatible with the reaction conditions.

Table 4. Synthesis of trisubstituted hydroxylamines.

R^{1}R^{2}N–O–C(=O)–R^3 → (DIBAL, Ac$_2$O, py, DMAP[a]) R^{1}R^{2}N–O–CH(OAc)–R^3 → (Et$_3$SiH, BF$_3$.OEt$_2$[b]) R^{1}R^{2}N–O–CH$_2$–R^3

62 → **65** → **66**

65a, X = OAc, -[c];
66a, X = H, 50%[d]

65b, X = OAc, -[c];
66b, X = H, 53%[d]

65f, X = OAc, 63%;
66f, X = H, 77%

65g, X = OAc, 88%;
66g, X = H, 83%

65h, X = OAc, 65%;
66h, X = H, 89%

65i, X = OAc, 75%;
66i, X = H, 81%

65j, X = OAc, 89%;
66j, X = H, 85%

65k, X = OAc, 55%;
66k, X = H, 75%

65l, X = OAc, 84%;
66l, X = H, 76%

65m, X = OAc, 85%;
66m, X = H, 74%

65n, X = OAc, 90%;
66n, X = H, 38%

65o, X = OAc, -[c];
66o, X = H, 42%[d]

[a] i) DIBAL (1.25-2 equiv), CH$_2$Cl$_2$, -78 °C, time; ii) Ac$_2$O (6 equiv), Py (3 equiv), DMAP (2 equiv), CH$_2$Cl$_2$, -78 ° to 0 °C. [b] Et$_3$SiH (2.5-5 equiv), BF$_3$OEt$_2$ (2.5-6.25 equiv), CH$_2$Cl$_2$, -78 ° to RT. [c] Not isolated. [d] Yield over two steps.

The so-obtained *O*-(α-acetoxyalkyl)hydroxylamines were then treated with triethylsilane in the presence of boron trifluoride etherate at −78 °C, and the reaction was warmed to 0 °C to give the desired *N,N,O*-trisubstituted hydroxylamines in good yields (**Table 4**). As some *O*-(α-acetoxyalkyl)-*N,N*-dialkyl hydroxylamines intermediates (**65a**, **65b**, and **65o**) were found to be quite unstable and decomposed during the purification process, they were transformed to their corresponding tri-substituted hydroxylamines

(**66a**, **66b**, and **66o**) directly without further purification in overall good yield for the two-step process.

On careful monitoring of the reactions, the actual temperature of the Lewis acid-mediated reduction of the *O*-(α-acetoxyalkyl)hydroxylamines by triethylsilane was observed to be substrate dependent, reflecting the stability of the presumed intermediate aminoxocarbenium ions. The β-(2-naphthyloxy)-ethyl system **65n** is an extreme example of the effect of substituent on reduction as it required multiple equivalents of triethylsilane and BF$_3$.OEt$_2$ at room temperature, and took 7 days in order to afford a 38% isolated yield of hydroxylamine **66n**. The relative slowness of this particular reduction reflects the destabilizing influence of alkoxy groups on oxocarbenium ion like intermediates such as is widely appreciated in carbohydrate chemistry.[115-117]

To understand the effect of electron withdrawing groups in the reduction further and to support the existence of assumed aminoxocarbenium ion intermediate, reductions of a series of three *O*-(β,γ, and δ-azido-1-acetoxyalkyl) hydroxylamines were executed (**Scheme 21**).
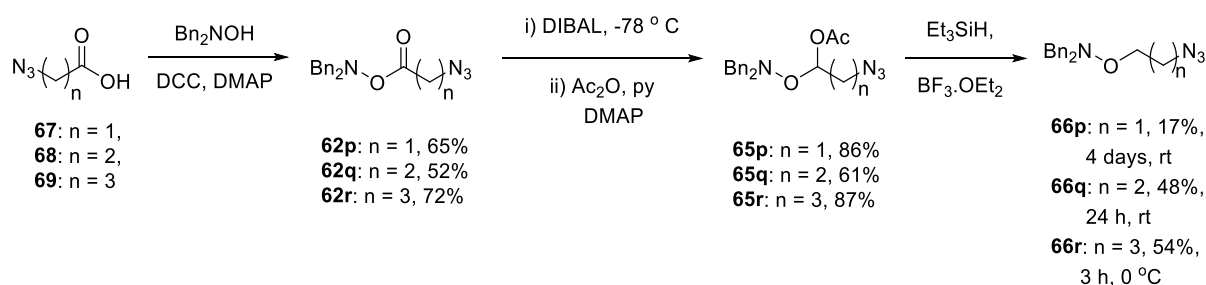
Scheme 21. Synthesis of azidoalkylhydroxylamines.

The synthesis of three *O*-(β,γ, and δ-azido-1-acetoxyalkyl) hydroxylamines were achieved by a similar protocol to the one described previously. As anticipated, the subsequent Lewis acid-mediated reduction by triethylsilane to give the *O*-

(azidoalkyl)hydroxylamines was strongly affected by the proximity of the azido group to the center of reaction. Thus, while the 1-acetoxy-4-azidobutyl system **65r** was reduced to the hydroxylamine **66r** in 54% yield under the standard conditions of treatment with BF$_3$.OEt$_2$ and triethylsilane at −78 °C in dichloromethane followed by warming to 0 °C and workup, a more modest 48% yield of the lower homolog **66q** was obtained from the 1-acetoxy-3-azidopropyl precursor **65q** only when the reaction was warmed to room temperature and stirred for 24 h. Reduction of next lower homolog **65p** was significantly slower, such that **66p** was obtained only in 17% yield after stirring for four days at room temperature. The presence of an electron-withdrawing group vicinal to the reaction center therefore, very strongly retards reduction of the *O*-(1-acetoxyalkyl)hydroxylamines, but the effect falls off rapidly with the insertion of successive methylene groups. The strongly retarding effect of the electron-withdrawing group thus observed in the reduction of both **65n** and **65p** supports the intermediacy of an aminoxocarbenium ion intermediate (**Scheme 22**) in these reactions.
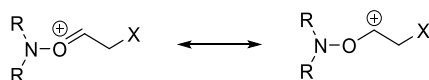
Scheme 22. Intermediate aminoxocarbenium ion destabilized by the electron-withdrawing group X.

2.1.4. Synthesis of tri-substituted hydroxylamines with C-C bond formation

The isolation of the *O*-(α-acetoxyalkyl)hydroxylamines also provided an opportunity to introduce nucleophiles adjacent to the oxygen and increase the molecular complexity of the product hydroxylamines. Therefore, adapting literature methods for the reductive allylation of α-acetoxy ethers,[118,119] various carbon nucleophiles such as allylstannanes, enolethers, and 2-methylfuran were used in this boron-trifluoride etherate-

promoted substitution to afford a number of novel hydroxylamines (**Table 5**) in modest to

excellent yields.

Table 5. Synthesis of trisubstituted hydroxylamines with concomitant C-C bond formation.
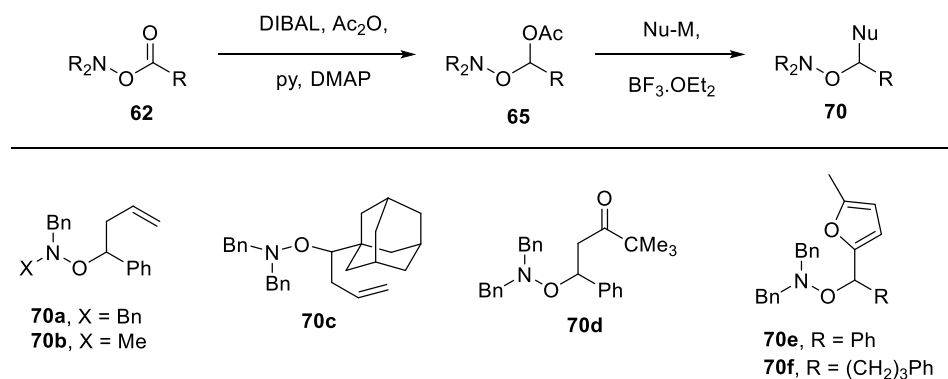
Entry	Ester	*O*-(α-Acyloxy)hydroxyl-amine, 65[a]	Reagent, conditions	Hydroxylamine, 70, %yield
1	**62a**	**65a**, -[b]	Bu₃SnCH₂CH=CH₂ (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 to 0 °C	**70a**, 82%[c]
2	**62b**	**65b**, -[b]	Bu₃SnCH₂CH=CH₂ (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 to 0 °C	**70b**, 54%[c]
3	-	**65h**	Bu₃SnCH₂CH=CH₂ (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 to 0 °C	**70c**, 79%
4	**62a**	**65a**, -[b]	CH₂=C(OTMS)CMe₃, (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 to 0 °C	**70d**, 66%[c]
5	**62a**	**65a**, -[b]	2-methylfuran (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 °C	**70e**, 51%[c]
6	-	**65g**	2-methylfuran (2.5 equiv), BF₃OEt₂ (2.5 equiv), CH₂Cl₂, -78 to 0 °C	**70f**, 20%

[a] i) DIBAL (2 equiv), CH₂Cl₂, -78 °C; ii) Py (3 equiv), DMAP (2 equiv), Ac₂O (6 equiv), CH₂Cl₂, -78 to 0 °C.

[b] Not isolated. [c] Two steps.

2.1.5. Study of stereoselectivity of addition of nucleophiles

As the carbon nucleophile addition on aminoxocarbenium ion intermediate in Lewis acid-mediated reaction creats a new stereogenic center in the molecule, the stereoselectivity of the reaction was investigated by applying the standard conditions in the substrates with pre-existing stereocenters (**Scheme 23**). Intermediate **73** was prepared from Evan's alkylated product **71**.[120] Reaction of hydrogen peroxide and lithium hydroxide with **71** afforded the corresponding acid. The resulting acid was then used for acylation of *N,N*-dibenzylhydroxylamine in the presence of *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide (EDCI) and Hunig's base, and *O*-acyl hydroxylamine **72** was generated. Upon reduction with DIBAL at −78 °C followed by treatment with acetic anhydride and warming to 0 °C, **72** was converted to *O*-(α-acetoxyalkyl)-*N,N*-dialkyl hydroxylamine **73**.
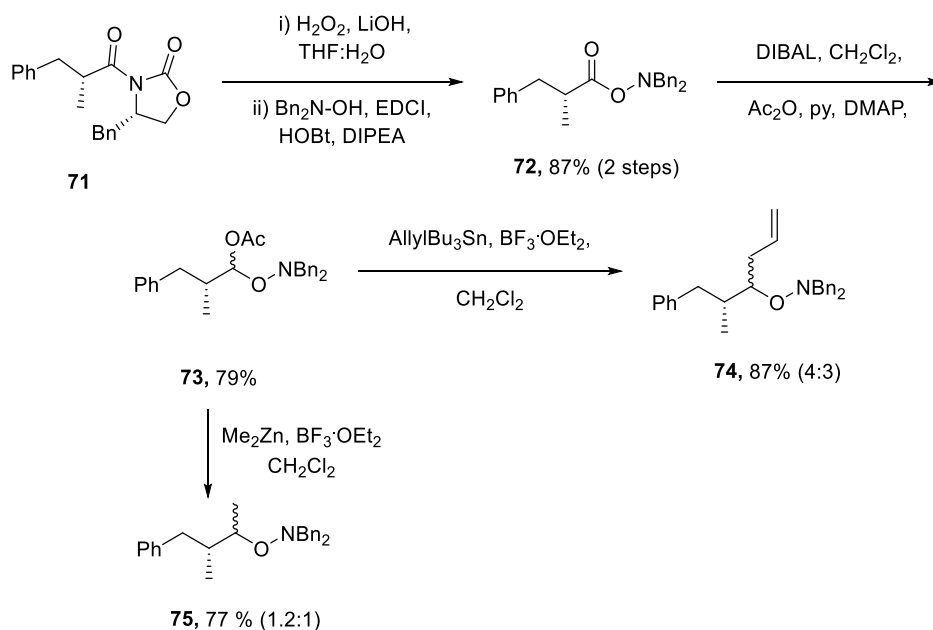
Scheme 23. Study of stereoselectivity of the Lewis acid-mediated reaction.

With the intermediate **73** in hand, the stereoselectivity of the nucleophilic addition was studied. In a reaction of **73** with allyltributylstannane as a nucleophile in the presence of $BF_3.OEt_2$, a mixture of two inseparable diastereomers (**74a** and **74b**) was isolated in 87% yield with poor selectivity (1:1.2). The product **74b** (conformation **D**) and product **74a** (conformation **A**) were identified as the major and minor products, respectively from the NOE experiments and $^3J_{H1,H2}$ values (**Figure 20**). Under similar reaction conditions, an unassigned mixture of inseparable diastereomers (**75a** and **75b**) was obtained in a ratio of 1:1 when using dimethylzinc as a nucleophile. Owing to the observed poor selectivity of these reactions, further investigation towards diastereoselective reaction was aborted.
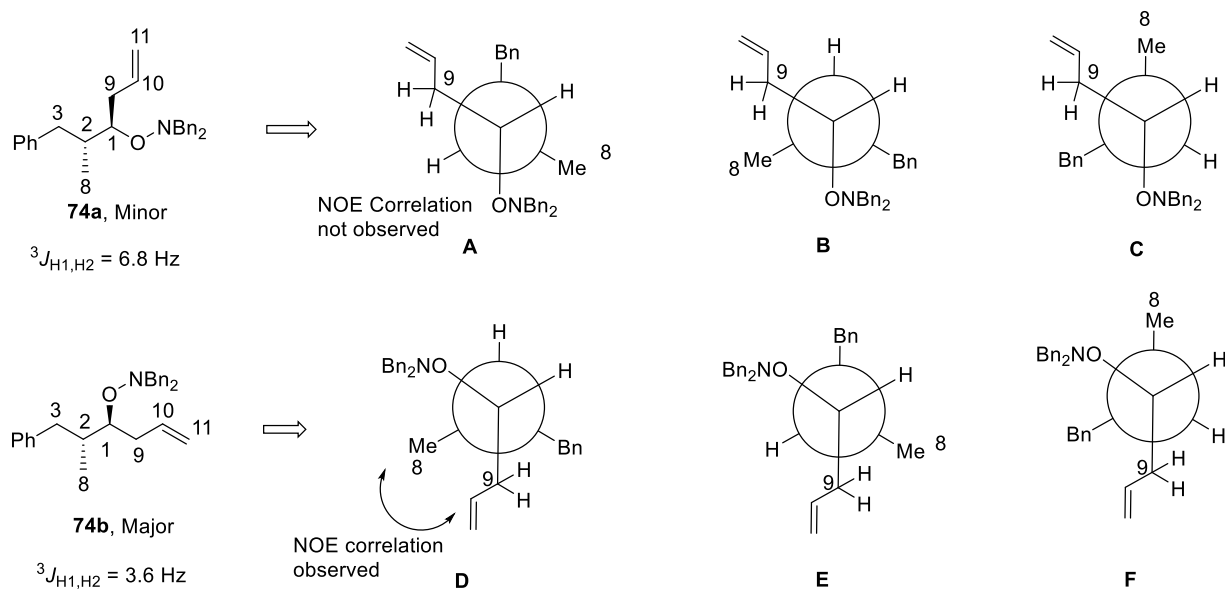
Figure 20. NOE correlation of **74a** and **74b**.

2.1.6. Considering an analogy between tetra-substituted hydrazines and tri-substituted hydroxylamines

The synthesis of tetra-substituted hydrazines was considered analogously to the synthesis of tri-substituted hydroxylamines. Therefore, a two-step reaction method was designed for tetra-substituted hydrazine synthesis based upon stepwise reduction of a

hydrazide, which itself can be accessed from *N*-acylation of a tri-substituted hydrazine (**Scheme 24**.).
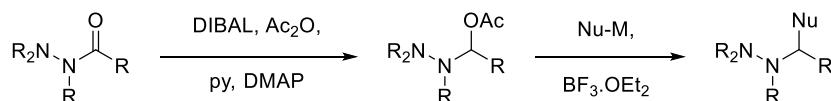
Scheme 24. Synthesis of tetra-substituted hydrazines.

To test the idea, hydrazide **79** was prepared from 1-Boc-1-methylhydrazine **76**[121] (**Scheme 25**). Benzylation of **76** with excess benzyl bromide afforded 1-Boc-1-methyl-2,2-dibenzylhydrazine (**77**), which was then deprotected to 1,1-dibenzyl-2-mehylhydrazine (**78**) by treatment with trifluoroacetic acid. Treatment of **78** with 4-phenylbutyric acid in the presence of DCC and DMAP afforded the hydrazide **79** in excellent yield.
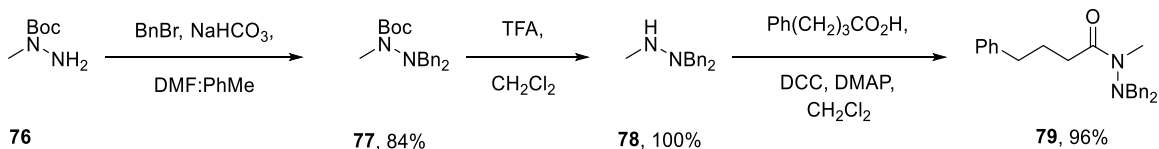
Scheme 25. Synthesis of hydrazide **79**.

Reduction of hydrazide **79** to *N*-(α-acetoxyalkyl)hydrazine **80** was tested by treating with 1.5 equivalents of DIBAL at −78 °C, followed by addition of acetic anhydride, pyridine, and DMAP, and warming to 0 °C. Surprisingly, the anticipated *N*-(α-acetoxyalkyl)hydrazine **80** was not observed, but the tetra-substituted hydrazine **81** and hydrazide **82** were isolated in 33% and 30% yields, respectively. When **79** was treated only with 3 equivalents of DIBAL at lower temperature, the tetra-substituted hydrazine **81** was isolated in 65% yield, and 14% of starting material was recovered. When the reaction was exposed to allyltributylstannane after treatment with 1.25 equivalent of DIBAL at −78 °C followed by warming to room temperature, compound **83** was isolated in 31% yield.
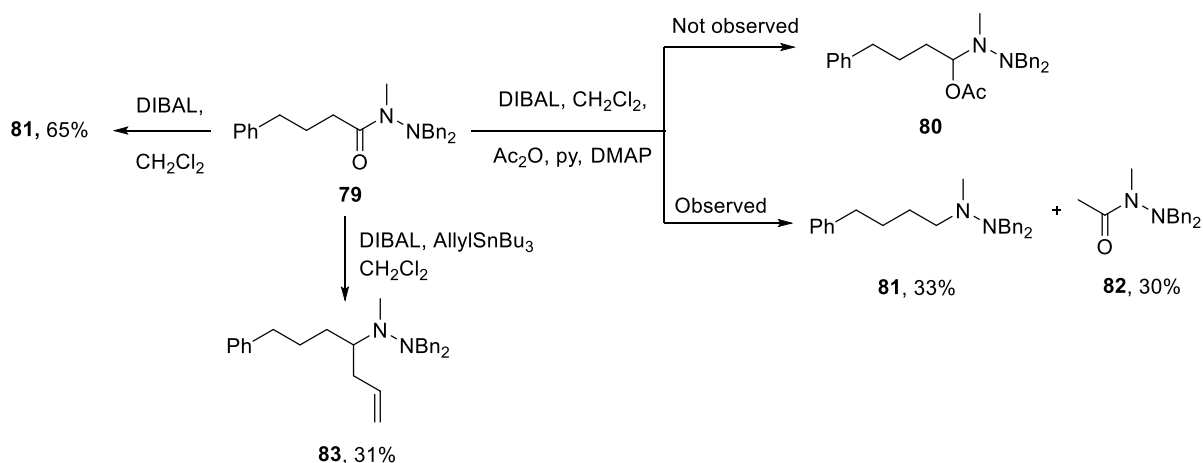
Scheme 26. Results from the reductions of hydrazide **79**.

Consistent with the reduction of **79**, hydrazide **84**, which was prepared from tri-substituted hydrazine **78** and benzoic acid, also resulted in the formation of tetra-substituted hydrazine **85** in 61% yield when reduced with 3 equivalents of DIBAL at −78 °C. Similarly, precursor **86**, which was prepared from *sym*-dimethylhydrazine and an excess of 4-phenylbutyric acid, also afforded tetra-substituted hydrazine **87** in 46% yield with 6 equivalents of DIBAL.
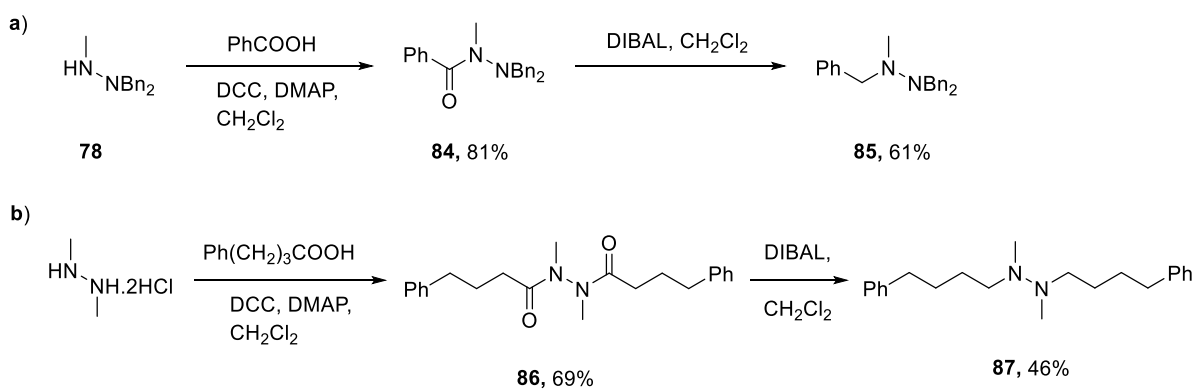
Scheme 27. Synthesis of tetra-substituted hydrazines **85** and **87**.

The failure to observe *N*-(α-acetoxyalkyl)hydrazine **80** from hydrazide **79** in the presence of DIBAL and acetic anhydride, and complete reduction of hydrazides **79**, **84**, and **86** to tetra-substituted hydrazine with DIBAL only, suggests that these reactions

proceeds via azaiminium ion intermediate (**Figure 21**). As azaiminium ion is more stable than aminoxocarbenium ion, at low temperature, the formation of azaiminium ion intermediate was facile and unavoidable in the DIBAL reduction of hydrazide, whereas the formation of aminoxocarbenium ion in the reduction of *O*-acylhydroxylamine was not observed .

azaiminium ion   aminoxocarbenium ion

Figure 21. Structures of azaiminium ion and aminoxocarbenium ion intermediates.

2.2. Conclusion

A straightforward two-step synthesis of *N,N,O*-trisubstituted hydroxylamines has been developed based upon the stepwise reduction of *O*-acylhydroxylamines. The *O*-acylhydroxylamines were easily accessed from acylation of *N,N*-disubstituted hydroxylamines, which themselves were achieved from the benzoylation of commercial amines followed by deacylation. The reductive deacetoxylation of an intermediate *O*-(α-acetoxyalkyl)hydroxylamine is proposed to proceed via intermediacy of an aminoxocarbenium ion. This hypothesis is supported by the diminished reactivity of *O*-(α-acetoxyalkyl)hydroxylamine with electron withdrawing substituents in an alkyl side chain. The replacement of triethylsilane with various carbon nucleophiles in the reductive deacetoxylation step enabled the introduction of substitutions on the α-position of the *O*-substituents. Poor stereoselectivities were observed when the reductive deacetoxylation was conducted on substrates with the pre-existing stereocenters adjacent to the acetoxy group regardless of the nature of carbon nucleophiles used. Overall, the method has

extended the availability of a range of fully substituted hydroxylamines permitting wider application of this under-utilized functionality in medicinal chemistry.

2.3. Future directions

The scope of the hydroxylamine-forming process can be further expanded by manipulating final reduction steps in several ways. Some of the possible manipulations are presented in **Scheme 28**. A well-designed intramolecular cyclization in reductive deacetoxylation of *O*-(*α*-acetoxyalkyl)hydroxylamine has a potential to form a series of compounds with carbocycles on *O*-substituents (**Scheme 28a**). Also, aminoxysugar derivatives are possible if the intramolecular nucleophile is an oxygen atom (**Scheme 28b**). Alternatively, endocyclic hydroxylamines can be achieved if the intramolecular nucleophile is appended to the *N*-substituent (**Scheme 28c**). Overall, the method has the potential to generate diverse sets of tri-substituted hydroxylamines to enrich compound libraries for medicinal chemistry.

$X = CH_2, O$
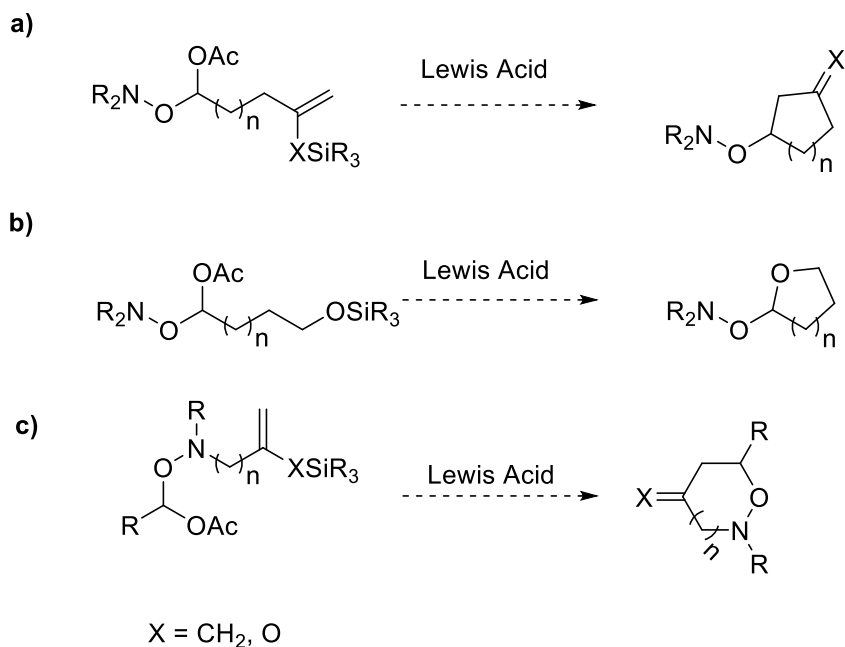
Scheme 28. Some possible future applications for the hydroxylamine-forming method.

CHAPTER 3. CONFORMATIONAL DYNAMICS OF *N*-ALKOXYPIPERIDINES AND PYRROLIDINES

To minimize repulsive interactions between the lone pairs of electrons on nitrogen and oxygen atoms, hydroxylamines adopt two stable conformers (**Figure 22**) with lone pairs of one atom eclipsing bonds of the next atom. The inter-conversion of these conformers at ambient temperature is a rapid process that is attributable to the low inversion barrier at the nitrogen and the low rotation barrier around the N-O bond. Understanding the degree of contributions and the relative energetics of these two processes in the stereomutation of hydroxylamines has been an area of interest for chemists. As a result, extensive studies, both computational and spectroscopic, have been conducted and described in a series of reviews.[17,122-124]
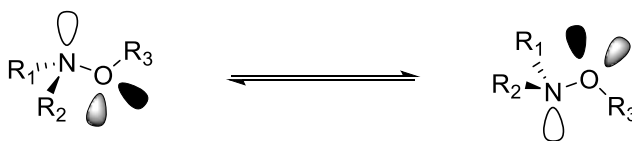
Figure 22. Stereomutation of acyclic hydroxylamine.

The situation is more complex for cyclic hydroxylamines in which the N-O bond is a part of the ring system, and understanding the conformational changes of such systems is rather more complicated due to ring inversion. This complication in stereomutation can be attributable to the individual processes of nitrogen inversion, N-O bond rotation, and ring flip or the interplay of a combination of all three processes. Several studies discussing about this problem have been reported for the endocyclic systems like oxazines in which the N-O bond is a part of the ring system.[17,122-124] However, the studies for the exocyclic hydroxylamines such as *N*-alkoxypiperidines are rare. To shed light on conformational preferences in the *N*-alkoxypiperidine system, variable temperature NMR (VT-NMR) studies of 4-substituted *N*-hydroxypiperieines, *N*-alkoxypiperidines, and *N*-

alkoxypyrrolidines (**Figure 23**) were conducted, and the data were analyzed together with the literature data of analogous comparators.

88, X = H
89, X = Cl
90, X = Me

91, X = H
92, X = Cl
93, X = Me

94
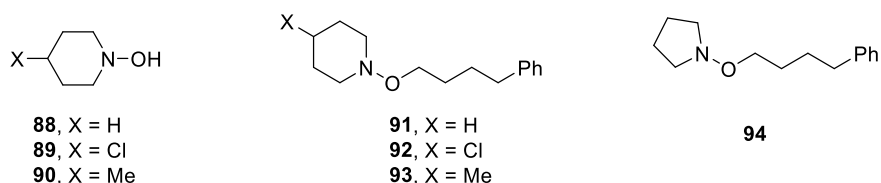
Figure 23. *N*-hydroxypiperidines, *N*-alkoxypiperidines, and *N*-alkoxypyrrolidine studied by variable temperature NMR.
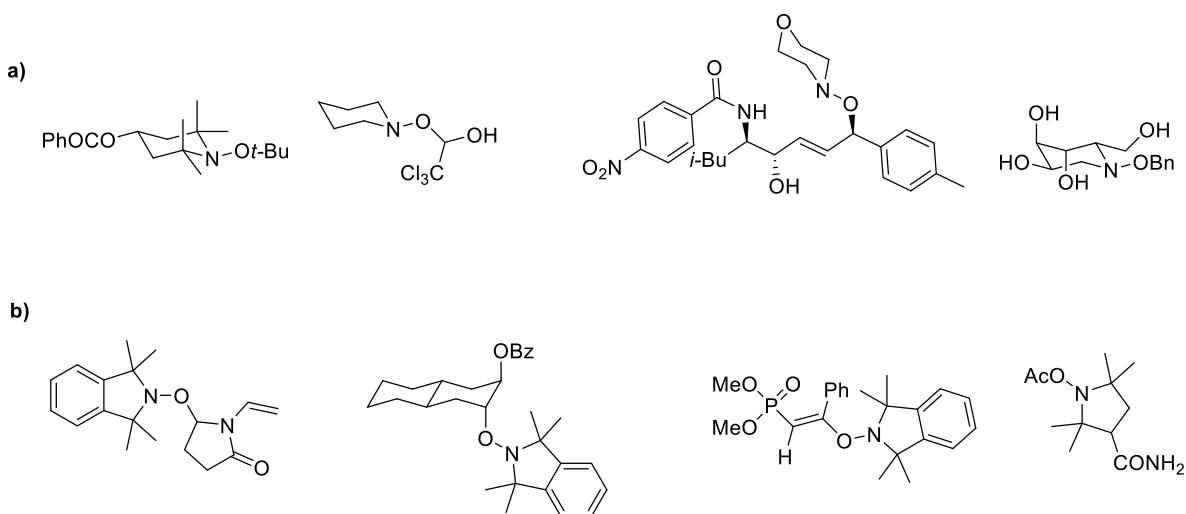
Figure 24. Cyclic hydroxylamines with exocylic N-O bond. a) Six-membered and b) five-membered.

According to the existing literature on saturated six membered hydroxylamines with an exocyclic N-O bond, the nitrogen atoms are predominantly pyramidal as revealed by their X-ray crystal structures,[17,122-124] and the rings adopt chair conformations with the nitrogen substituents in equatorial positions (**Figure 24a**).[45,125-128] The nitrogen atom is also pyramidal in similarly substituted five-membered nitrogen heterocycles as observed in their X-ray crystal structures (**Figure 24b**).[129-133] Consistent with the acyclic and endocyclic hydroxylamines,[17,122-124] these exocyclic hydrxoylamines also prefer

conformations in which the lone pair on nitrogen eclipses with the substituent on oxygen, whether it is a simple alkyl group,[45,125,128,133] a vinyl group,[131] a trifluoromethyl group,[127] an acyl group,[132] or a 1-hydroxyalkyl group.[126] Therefore, *N*-alkoxypiperidines and pyrrolidines are expected to exist in solution as a mixture of two equivalent conformers, and nitrogen inversion, ring inversion, and rotation about the N-O bond is involved in the interconversion of these conformers (**Figure 25**).
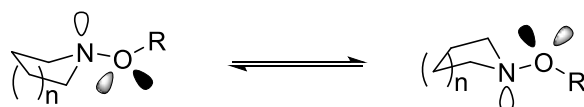
Figure 25. Stereomutation of the *N*-alkoxypiperidines (n = 2) and pyrrolidines (n = 1).

In 1985, Jenkins *et al.* conducted VT-NMR studies in a series of 2-alkoxy-1,1,3,3-tetramethylisoindolines **95** (**Figure 26**) and measured the energy barriers to stereomutation. Based upon the increasing barriers with increasing steric bulk of alkoxy group and decreasing barriers with increasing solvent polarity, the authors inferred that the two processes, the nitrogen inversion and the N-O bond rotation, on these molecules take place at the same time.[134] Later in a VT-NMR study of a series of 1-alkoxy-2,2,6,6-tetramethylpiperidines **96** (**Figure 26**), Anderson *et al.* also observed similar changes in energy barriers with the steric bulk of the alkoxy groups, although not in a linear fashion.[125] Anderson and coworkers further considered that, as each of the three individual processes of N-O bond rotation, ring inversion, and nitrogen inversion have substantial individual barriers, stepwise processes are more likely than composite ones. Finally, they concluded that the main component of the observed barrier in **96** is the barrier to N-O bond rotation.[125] Although VT-NMR studies were performed in a series of *N*-hydroxy and *N*-acyloxy 2,2,6,6-tetramethylpiperidines with the substituents at 4-position in order to

determine the energy barriers,[135] no such studies are reported for 4-substituted *N*-alkoxypiperidines.
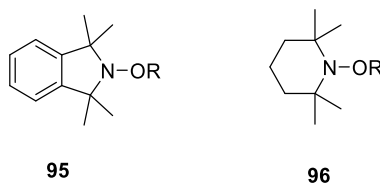
Figure 26. General structures of literature comparators.

To understand the stereomutation process in 4-substituted *N*-alkoxypiperidines and *N*-hydroxylpiperidines in the absence of extreme steric buttressing like in **95** and **96**, VT-NMR studies were performed in *N*-hydroxypiperidines **88**-**90**, *N*-alkoxypiperidines **91**-**93**, and *N*-alkoxypyrrolidine **94**. These exocyclic hydroxylamines were previously synthesized during the methodology development program for trisubstituted hydroxylamine (**Chapter 2**) and are re-numbered in this section for the sake of convenience.

After comparing the ^{1}H NMR spectra of *N*-hydroxypiperidines **88**-**90** at ambient temperature in CDCl$_3$, the effect of substituent at the 4-position on the conformational and configurational equilibria of these systems was clearly noted (**Figure 27**). The unsubstituted *N*-hydroxypiperidine system **88** predominantly adopts a chair-like conformation with a distinct axial and equatorial protons at the 2- and 6-postions, since two sets of two non-equivalent hydrogens germinal to the ring nitrogen are observed in the ^{1}H NMR spectrum. However, the spectrum of 4-chloro-*N*-hydroxypiperidine **89** reveals two isomers, **89a** and **89b**, in a 1:1 ratio. The spectrum of **89a** is similar to that of **88** with the chloride group in an equatorial position of the chair conformation. In contrast, the spectrum of **89b**, has only three signals. One for H4, a second one for all four germinal

protons at the 2- and 6-positions which are merged into a single resonance, and a third one for the combined protons at 3- and 5-postions mixed together.

As in **89**, the spectrum of 4-methyl-*N*-hydroxypiperidine **90** also represents two species **90a** and **90b**, but in a nearly 8.5:1 ratio. The major isomer **90a** adopts a chair-like conformation in which the methyl group is in an equatorial positions. Otherwise, the spectrum is consistent with the unsubstituted system **88** and the isomer **89a** of the 4-chloro system. The spectrum of the minor isomer **90b**, however, is similar to that of the isomer **89b** of the 4-chloro system as all methylene protons at the 2- and 6-positions are aggregated as a single resonance as are other protons at the 3- and 5-positions.
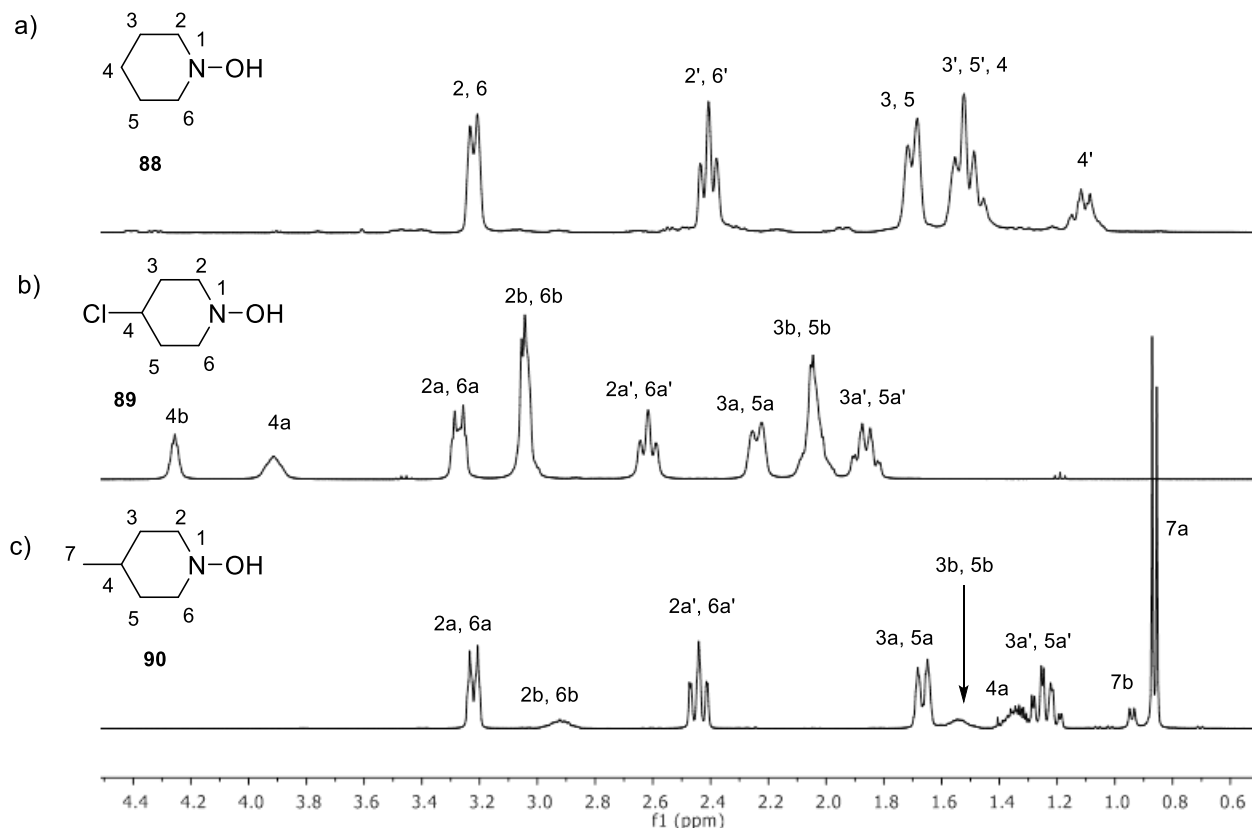
Figure 27. Partial ¹H-NMR spectra of the *N*-hydroxypiperidines **88** (a), **89** (b), and **90** (c).

When the unsubstituted system **88** was heated above 90 °C (coalescence temperature), the signals for the geminal protons at the 2- and 6-positions merged together as they became equivalent. This phenomenon clearly signifies that out of three processes, N-O bond rotation, ring inversion, and nitrogen inversion, at least one of them is slow at room temperature on the NMR time scale. When the system is substituted at the 4-position as in **89** and **90**, the nitrogen inversion process is slow on the NMR timescale and two diastereomers, namely *cis* and *trans*, are observed. However, the ratio of these diastereomers depends upon the size of the substituents present. When the substituent is smaller like a chlorine atom (steric A value = 0.53 – 0.64 kcal.mol^{-1}),[136] the ratio is about 1:1 and when the substituent is larger like a methyl group (steric A value = 1.74 kcal.mol^{-1}),[136] the *trans* isomer is favored. In general, **Figure 28** represents the overall conformational equilibria of a series of *N*-hydroxypiperidines.
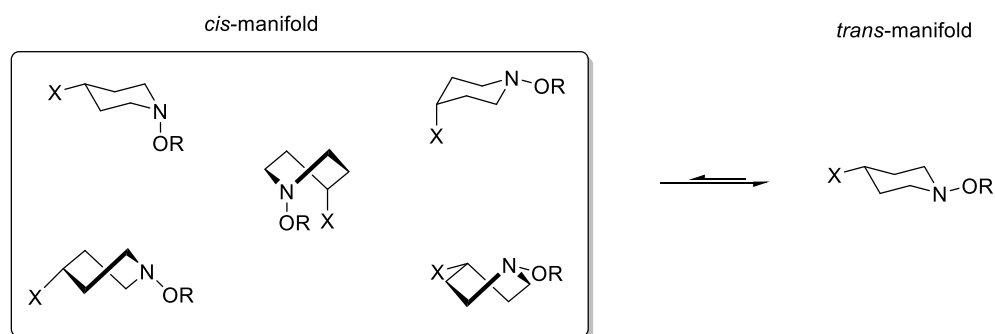
Figure 28. Conformational dynamics of the *N*-hydroxypiperidines (R = H) and *N*-alkoxypiperidines (R = alkyl).

Therefore, unsubstituted *N*-hydroxypiperidine **88**, isomer **89a** of 4-chloro-*N*-hydroxypiperidine, and major isomer **90a** of 4-methyl-*N*-hydroxypiperidine adopt predominantly chair conformations in which both the substituents are in equatorial positions (**Figure 28**, *trans*-manifold). In case of other isomer **89b** of 4-chloro-*N*-

hydroxypiperidine and minor isomer **90b** of 4-methyl-*N*-hydroxypiperidine, a mixture of several rapidly interconverting conformers including twist boats (**Figure 28**, *cis*-manifold) is possible, which results in the aggregation of the four protons geminal to nitrogen atom into a single resonance. Likewise, the four vicinal protons to nitrogen coalesce into a single peak in the minor cis isomers. A similar conclusion can be drawn from the ^{1}H NMR spectra of a series of *N*-alkoxypiperidines **91**-**93** recorded at room temperature in CDCl3 (**Figure 29**).
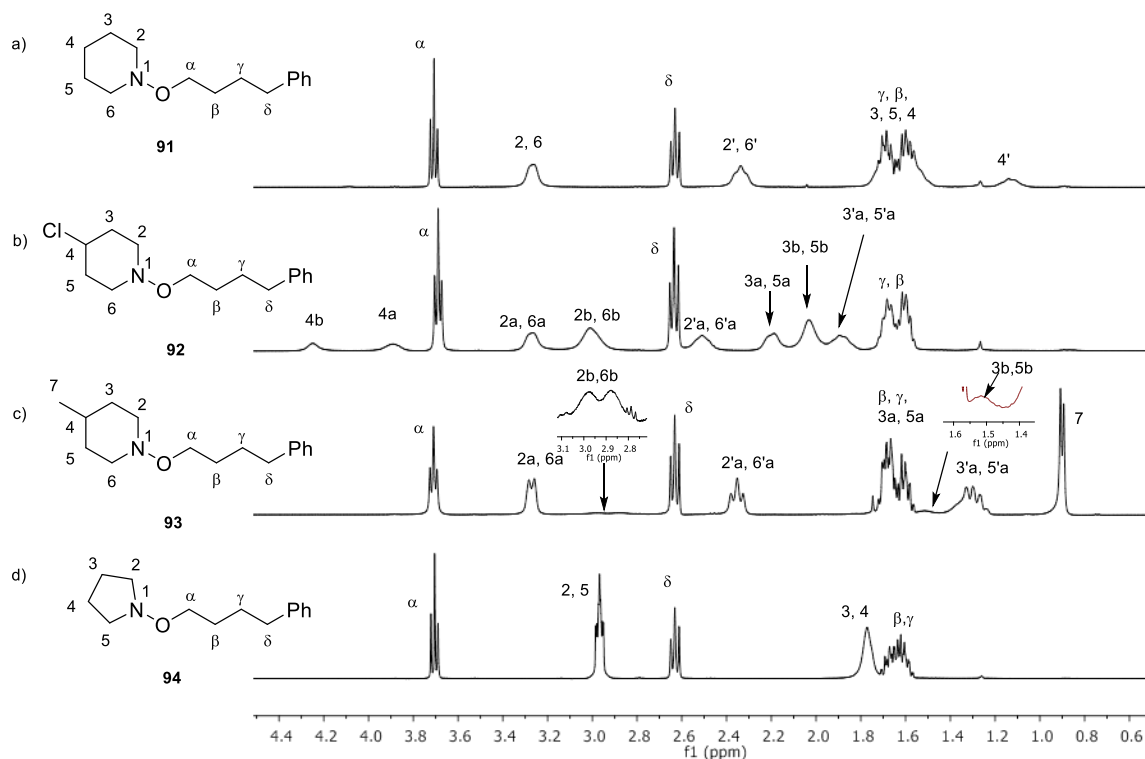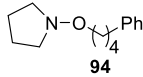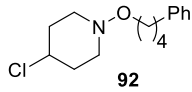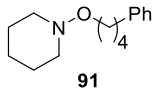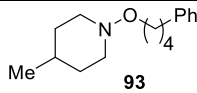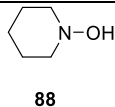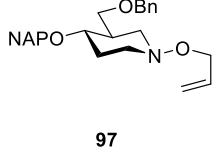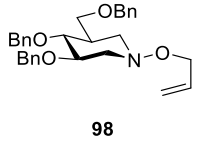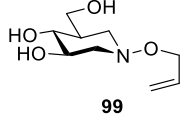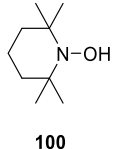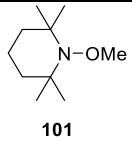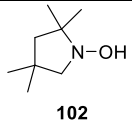
Figure 29. Partial ^{1}H-NMR spectra of the *N*-alkoxypiperidines **91** (a), **92** (b), and **93** (c), and the *N*-alkoxypyrrolidine **94** (d).

After inspection of ^{1}H-NMR spectra of these *N*-alkoxy systems, three crucial pieces of information are instantly discerned. First, all alkoxy groups are fully conformationally equilibrated on the NMR time scale at room temperature, since sharp signals are

observed for all the 4-phenylbutyl protons in each molecule. Second, the *cis* and *trans* isomers are still present in the *N*-alkoxy systems **92** and **93,** with broader line shapes for the piperidine moiety than in the corresponding *N*-hydroxy systems. Similar broadening was also observed for the unsubstituted system **91**. These broader resonances of the piperidine moiety imply that the *N*-alkoxy series is closer to the coalescence temperature for interconversion between the *cis* and *trans* configurations than the *N*-hydroxy series, and the nitrogen inversion barrier is lower for the former than for the latter. This result is in line with the inversion process occurring through a low energy planar transition state due to relief from steric interactions in the case of *N*-alkoxy series. Third, the entire set of NMR signals for the protons in *N*-alkoxypyrrolidine **94** at room temperature were sharp suggesting complete conformational equilibration on the NMR time scale. In order to scrutinize further, VT-NMR studies were conducted in *N*-hydroxypiperidine **88** and *N*-alkoxy systems **91-94** in toluene and DMF as NMR solvents, and the energy barriers to stereomutation were determined. These energy barriers are listed in **Table 6** in increasing order of barrier, together with the barriers for a number of literature comparators.

Table 6. Barriers to stereomutation in **91-94**, **88**, and literature[18,137,138] comparators as determined by VT ^{1}H-NMR.

Entry	Hydroxylamine	Inversion barrier ($\Delta G_c^{\ddagger}$), kcalmol^{-1}	Reference
1	**94**	13.3 (toluene-d$_8$) 12.3 (DMF-d$_7$)	[a]
2	**92**	15.3 (toluene d$_8$) 15.4 (DMF-d$_7$)	[a]
3	**91**	15.9 (toluene d$_8$) 15.9 (DMF-d$_7$)	[a]
4	**93**	[b]	[a]
5	**88**	16.4 (toluene d$_8$) 16.4 (DMF-d$_7$)	[a]
6	**97**	15.3 (DMF-d$_7$)	18
7	**98**	14.6 (DMF-d$_7$)	18
8	**99**	14.5 (DMF-d$_7$)	18
9	**100**	11.5 (THF-d$_8$)	137

10	101	17.0 (CCl$_4$)	137
11	102	12.0 (DMF-d$_7$)	138

[a]: this work. [b]: no coalescence below 120 °C.

The inversion barriers for the series of *N*-alkoxypiperidines **91-93** and **97-99** (**Table 6**, entries 2-4 and 6-8) are significantly higher than for the *N*-alkoxypyrrolidine **94** (**Table 6**, entry 1). This result is consistent with the fact that i) the six-membered ring inversion is a more difficult process than the pseudorotation of the five-membered ring, and ii) the accommodation of an sp^2-hybridized atom is much easier for a five-membered than for six-membered ring. Failure to achieve a coalescence temperature for 4-methyl-*N*-alkoxypiperidine **93** (**Table 6**, entry 4) below 120 °C led to a conclusion that the inversion barrier is significantly higher for the 4-methyl system than for the unsubstituted system **91**, 4-chloro system **92**, and other literature comparators (**Table 6**, entries 6-8). Based on the observations of increasing barrier with increasing steric at 4-postision in the piperidine system and the lower barrier for the pyrrolidine system over the piperidine system, the barrier in the piperidine system can be primarily attributed to the ring flip. The comparatively higher barrier to stereomutation of the literature molecule *N*-methoxy-2,2,6,6-tetramethylpiperidine **101** (**Table 6**, entry 10) than the barriers of the *N*-alkoxypiperidines **91** and **92** (**Table 6**, entries 2 and 3) and the other literature comparators (**Table 6**, entries 6-8) indicates that the main component of the barrier has switched to N-O bond rotation from ring flip due to steric buttressing in **101** as described by Anderson *et al.*[125] A similar conclusion can be drawn for the inversion barriers in the

N-hydroxypiperidine series. The barrier to stereomutation of *N*-alkoxypiperidine **91** (**Table 6**, entry 3) is slightly smaller than that of the corresponding *N*-hydroxy system **88** (**Table 6**, entry 5), implying that the oxygen substituent has minimal effect on stereomutation process. However, the barrier of *N*-methoxy-2,2,6,6-tetramethylpiperidine (**Table 6**, entry 10) is considerably higher than that of its *N*-hydroxy analog (**Table 6**, entry 9), which can be attributed to steric buttressing on N-O bond rotation. The observed lower barrier to stereomutation in the tetra-alkyl system **100** (**Table 6**, entry 9) in comparison to its sterically less hindered *N*-hydroxy and alkoxy analogs **88** and **91** (**Table 6**, entries 3 and 5) is, however, not surprising, since a similar pattern is observed in *N*-alkyl piperidine systems[139-141] and *N*-diazenyl piperidine systems[142,143] (**Figure 30**). The barrier is lower for 2,2,6,6-tetramethyl system **104** than for its unsubstituted congener **103**. A similar trend is also observed for 2,2,6,6-tetramethyl system **106** and its unsubstituted analog **105**. These phenomena reflect the situation in cyclohexane and 1,1,3,3-tetramethyl cyclohexane systems wherein the chair/twist boat energy gap is reduced on tetra-substituted systems in comparison to unsubstituted systems, as in the former case the chair conformation is destabilized by the repulsive *syn* 1,3-diaxial interaction between the alkyl groups.[144]

103   **104**   **105**   **106**
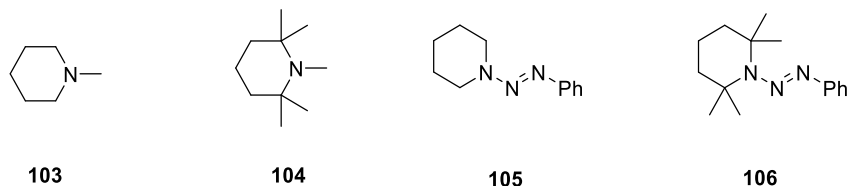
Figure 30. Unsubstituted and 2,2,6,6-tetramethyl substituted systems of *N*-alkyl piperidine and *N*-diazenyl piperidine.

3.1. Conclusion

In order to explore the remained uncertainty in the *N*-alkoxypiperidine system, variable temperature NMR studies of a series of 4-substituted *N*-hydroxypiperidines, *N*-alkoxypiperidines, and *N*-alkoxypyrrolidines were conducted. The observed strong dependence of VT NMR phenomena on the substituent at the 4-position together with other evidence from literature comparators led to the conclusion that the rate-determining step in the stereomutation of such piperidines is the ring flip and not the nitrogen inversion or the N-O bond rotation. When the system is sterically crowded like in *N*-alkoxy-2,2,6,6-tetramethylpiperidines, the barrier to N-O bond rotation becomes the major component of the barrier to stereomutation.

CHAPTER 4. SYNTHESES OF HYDROXYLAMINE AND HYDRAZINE ANALOGS OF KALKITOXIN

As described in **Chapter 1**, hydrazines and hydroxylamines have the potential to enrich compound collections in medicinal chemistry as they can be used to increase the sp^3 fraction of molecule as well as molecular complexity and diversity. As weak bases, hydroxylamines and hydrazines are little protonated under normal physiological conditions. Their low inversion barriers allow them to invert rapidly at nitrogen at ambient temperature, thereby reducing the stereogenic complexity of the molecule. Owing to these unique properties, hydroxylamine and hydrazine moieties can be considered as the surrogates of sp^3 carbons in the carbon frameworks (**Figure 31**), offering new compositions of matter to new libraries representing unexplored areas of chemical space. Taking advantage of these unique properties of the hydroxylamine and hydrazine functionalities, a broad program was initiated where hydroxylamine and hydrazine serve as convertible mimics for stereogenic centers in the carbon framework .
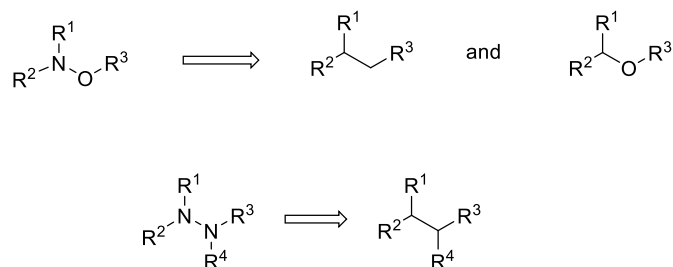
Figure 31. Hydroxylamine and hydrazine as surrogates of sp^3 carbons.

To test the concept of hydrazine and hydroxylamine analogs, kalkitoxin, a natural anticancer compound, was chosen as an initial substrate, because of the combination of its potent bioactivity and the presence of stereogenic centers that are crucial for activity. Two hydroxalogs and a hydrazine analog of kalkitoxin were designed, as shown in **Figure 32**. The hydroxalog 6-oxa-7-azakalkitoxin (**108**) was designed by substituting an oxygen

atom for C-6 and replacing the stereocenter C-7 with a nitrogen atom. The second hydroxalog, 9-oxa-10-azakalkitoxin (**109**), was designed by replacing C-9 with an oxygen atom and the stereogenic center C-10 with a nitrogen atom. The hydrazine analog 7,8-diazakalkitoxin (**110**) was designed by substituting the stereogenic centers C-7 and C-8 with two nitrogen atoms. These hydrazine and hydroxylamine analogs are intended to reduce the synthetic and stereogenic complexity of the target owing to the rapid inversions at nitrogen centers, and to test their influence on biological activity.
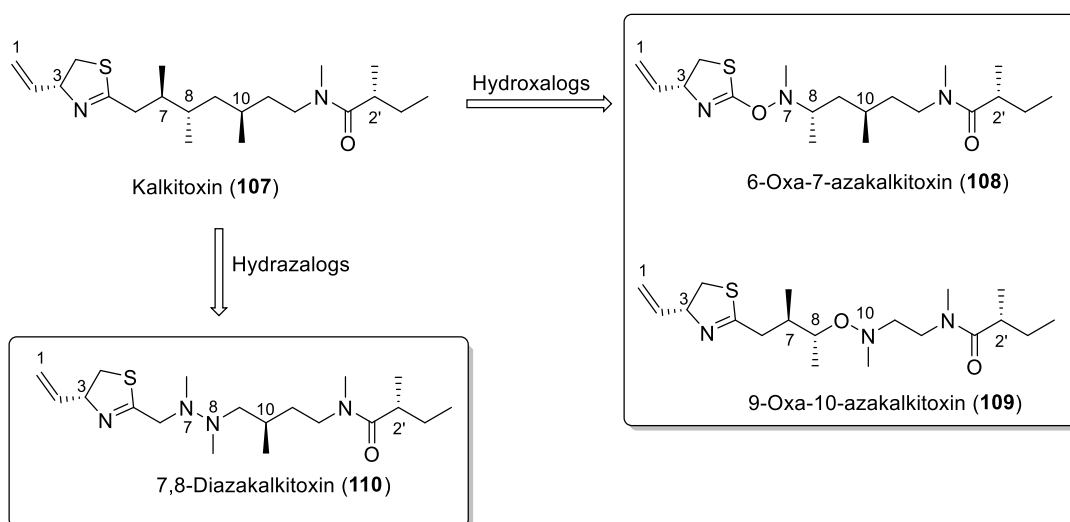
Figure 32. Kalkitoxin and the proposed hydroxylamine and hydrazine analogs.

The program commenced with the synthesis of these analogs. The parent compound, kalkitoxin, was also synthesized for the comparative study. The syntheses of kalkitoxin and its analogs and their biological assessments are described in the following sections.

4.1. Synthesis of kalkitoxin (107)
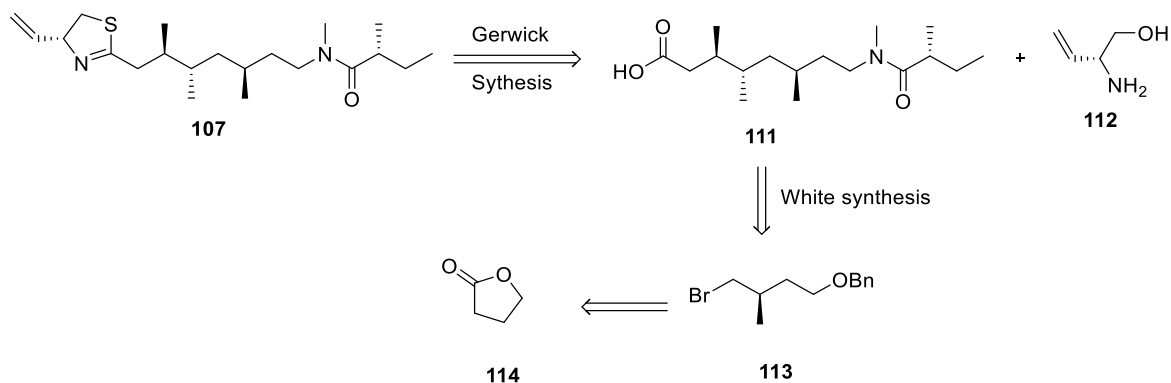
Scheme 29. Retrosynthesis of kalkitoxin.

Kalkitoxin was synthesized by partially adapting previous methods as shown in the retrosynthetic analysis (**Scheme 29**).[95,105] Wipf's oxazoline-thiazoline ring conversion sequence[104] (**Scheme 30**), utilized by Gerwick's synthesis,[95] was selected to construct thiazoline ring. The stretched-peptide chain **111** was, however, was prepared from alkyl bromide **113** by following the reaction conditions from White's kalkitoxin synthesis,[105] relying on their short and efficient pathway to assemble the 1,2,4-*anti,anti*-methyl sequence. γ-Butyrolactone (**114**) was chosen as a starting substrate to synthesize alkyl bromide **113**.
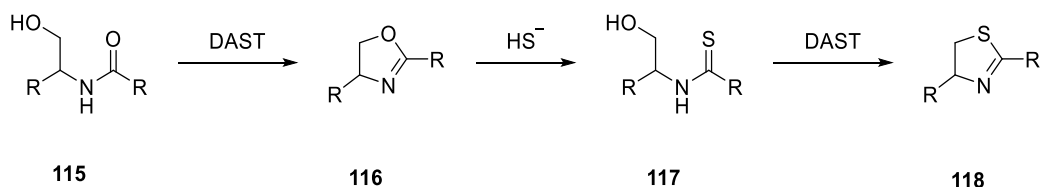
Scheme 30. Wipf's oxazoline-thiazoline conversion strategy.

The synthesis of kalkitoxin (**107**) commenced with hydrolysis of γ-butyrolactone (**114**), followed by benzylation of the resulting alcohol (**Scheme 31**).[145] Acylation of Lithiated (*R*)-4-benzyl-2-oxazolidinone (**120**) with acid **119** delivered *N*-acyloxazolidinone **121**. Enolization of *N*-acyloxazolidinone, followed by alkylation with methyl iodide, gave

122 stereoselectively. Lithium borohydride-mediated reduction afforded next alcohol **123**, which was transformed to bromide **113** upon subjection to Appel's reaction conditions.
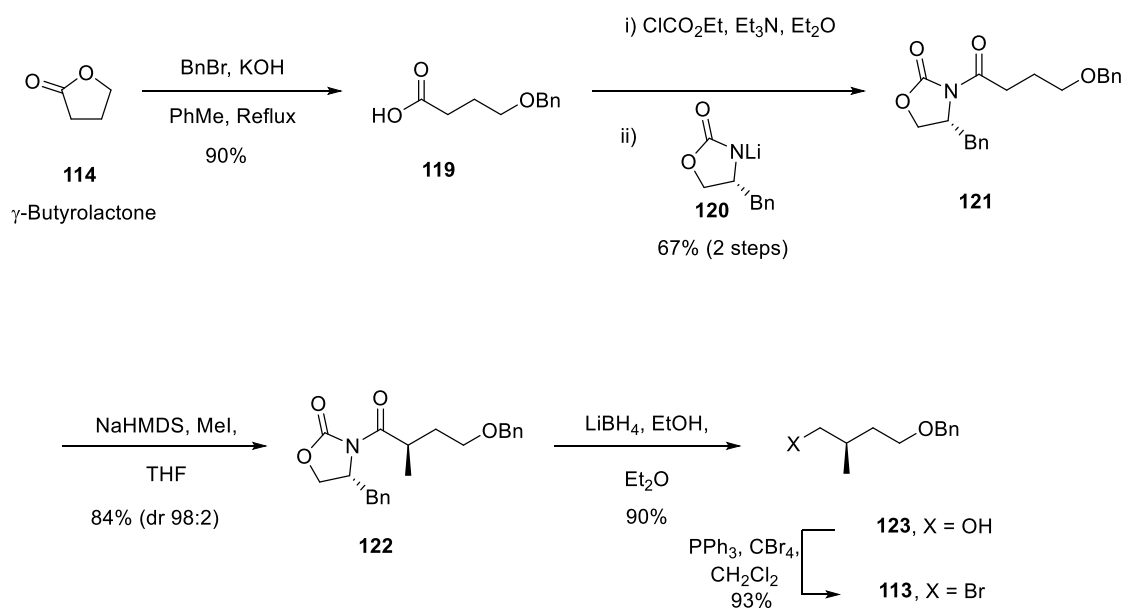
Scheme 31. Synthesis of alkyl bromide **113**.

Alkyl bromide **113** was then used to synthesize the stretch-peptide **111** by White's procedure (**Schemes 32** and **33**). After transformation of alkyl bromide **113** to an organocopper species **124**, it was treated with (*S*)-*N*-(*trans*-crotonyl)-4-phenyloxazolidin-2-one (**125**). The so-obtained product (**126**) with the 1,3-*anti* dimethyl configuration (79% yield) was enolized and further alkylated to deliver **127** (with the desired 1,2,4-*anti*,*anti* configuration) and **128** (with 1,2,4-*syn,anti* configuration) in 62% and 20% yields, respectively. The stereochemistry of compounds **126**, **127**, and **128** were confirmed by comparing their NMR spectra and specific rotation data with the literature data of these compounds synthesized from similar reaction conditions.[99]
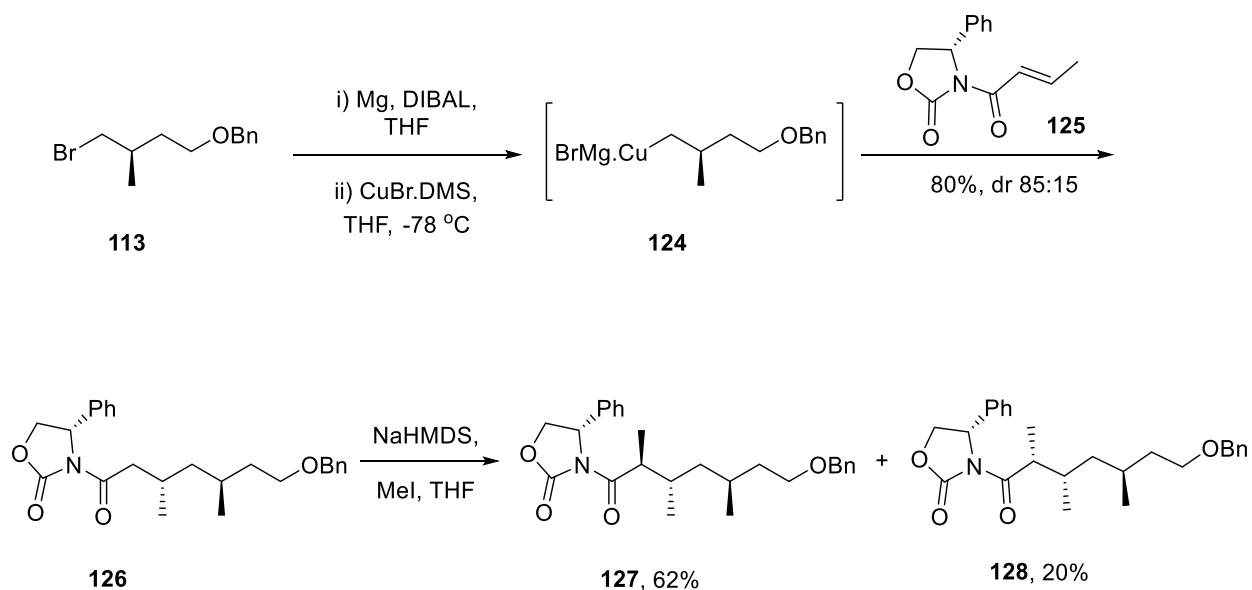
Scheme 32. Construction of the 1,2,4-*anti,anti* methyl sequence.

After hydrogenation of the major diastereomer **127**, Swern oxidation of the resulting alcohol **129** generated corresponding aldehyde **130** (**Scheme 33**). This aldehyde was then extended to amide **133** by reductive amination followed by coupling with (*R*)-methylbutyric acid (**132**). The chiral auxiliary was removed by lithium borohydride-mediated reduction to obtain alcohol (**134**). Subjection of this alcohol to Swern oxidation, followed by homologation by a Wittig reaction, gave a mixture of alkenes (*E/Z*) **135**. The mixture **135** was converted to acid **111** by hydrolyzing to the aldehyde, followed by Pinnick oxidation.
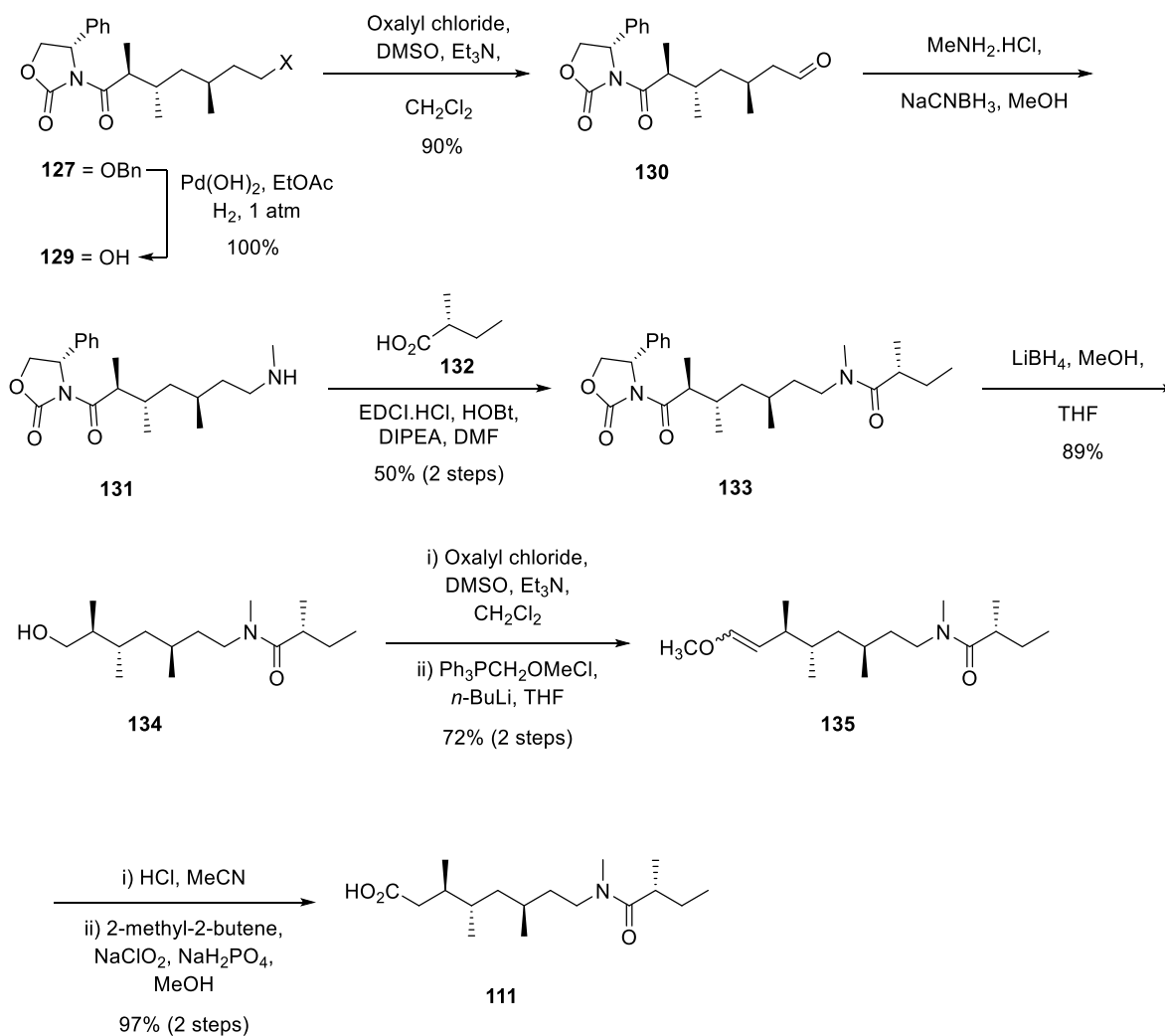
Scheme 33. Synthesis of acid **111**.

Further development of acid **111** to kalkitoxin was achieved by Gerwick's protocol (**Scheme 34**).[95] Acid **111** was coupled with amine **112** to access amido alcohol **136**, the precursor for Wipf's oxazoline-thiazoline reaction sequence. A DAST-mediated cyclization of **136** to 2-oxazoline derivative **137** and subsequent treatment with hydrogen sulfide resulted in thioamide **138**. DAST was again used for the second cyclization to deliver the targeted kalkitoxin (**107**) as a colorless oil. As expected, a mixture of rotamers was observed in its NMR spectra due to amide functionality.
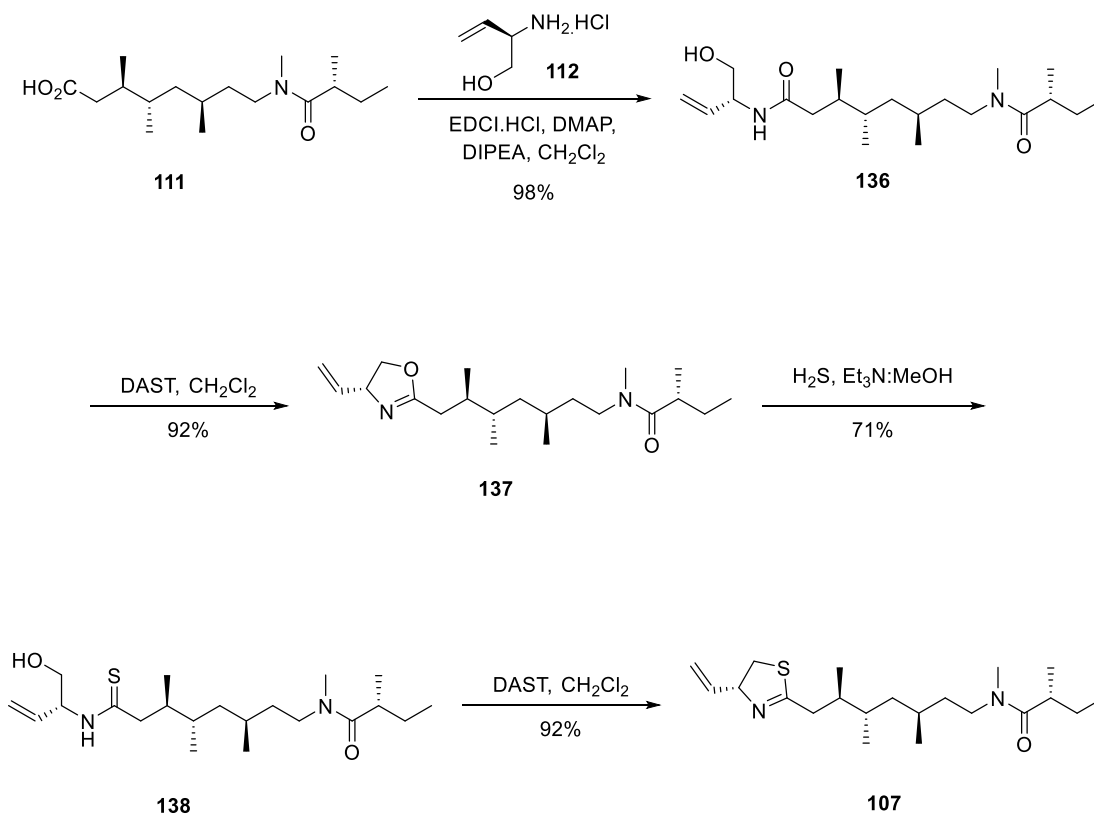
Scheme 34. Synthesis of kalkitoxin from acid **111**.

In summary, kalkitoxin, a stretch-peptide, was synthesized in 21 linear steps from γ-butyrolactone to provide 94 mg of the natural product. The unique 1,2,4-*anti*,*anti* configuration in the alkyl chain was obtained by the protocol used in White's kalkitoxin synthesis. However, the thiazoline ring was constructed efficiently adapting the procedure from the Gerwick's kalkitoxin synthesis. The so-obtained kalkitoxin (**107**) and its oxazoline congener (**137**) were then submitted for biological assessment in the Valeriote lab at the Henry Ford Hospital in Detroit.

4.2. Synthesis of 6-oxa-7-azakalkitoxin (108)
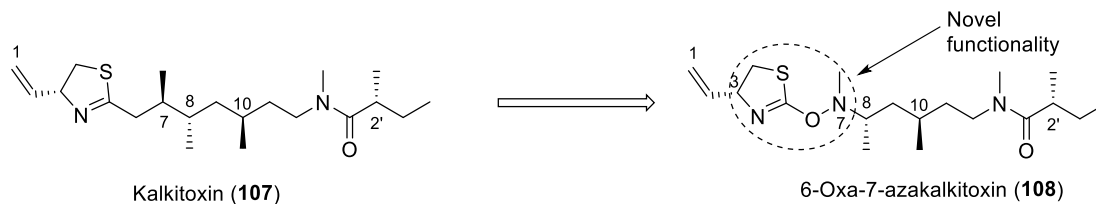
Figure 33. Structure of kalkitoxin and its proposed hydroxalog 6-oxa-7-azakalkitoxin.

The intended modification of kalkitoxin **107** to hydroxalog **108** introduces a novel and unexplored functional moiety, the 2-aminoxy thiazoline, in the molecule. Hence, before undertaking the total synthesis of **108**, it was necessary to find a route to introduce such moiety with the aid of a simple model. Therefore, the model compound **141** was targeted for synthesis (**Scheme 35**). Treatment of ethyl (*R*)-2-oxothiazolidine-4-carboxylate (**139**),[146] an L-cysteine-derived thiazolidinone, with triflic anhydride in the presence of Hunig's base afforded triflate **140**. Owing to its unstable nature, triflate **140** was subsequently treated with the sodium alkoxide of *N,N*-dibenzylhydroxylamine without purification. The targeted compound **141** was observed in the crude reaction mixture by mass spectrometry but decomposed during the workup, leading to the conclusion that the targeted functionality is unstable and difficult to handle. Based on the observed instability of the model compound **141**, the plan to synthesize **108** was halted.
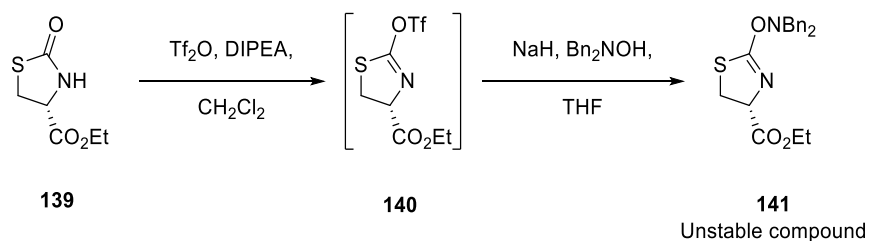
Scheme 35. Synthesis of a model 2-aminoxy thiazoline.

4.3. Synthesis of 9-oxa-10-azakalkitoxin (109)

Kalkitoxin (**107**)

9-Oxa-10-azakalkitoxin (**109**)
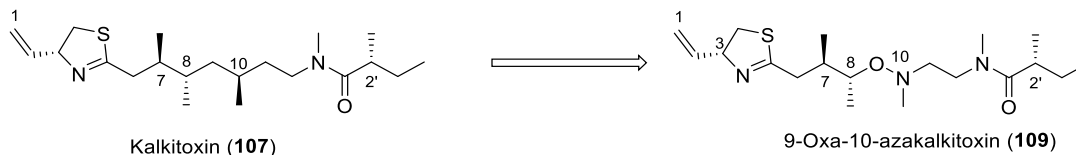
Figure 34. Structure of kalkitoxin and its hydroxalog 9-oxa-10-azakalkitoxin.

The retrosynthetic analysis for hydroxalog **109** was performed relying partially on Gerwick's kalkitoxin synthesis[95] (**Scheme 36**). The synthesis of the target was proposed from three fragments, **112**, **142**, and **132**. As in Gerwick's kalkitoxin synthesis,[95] thiazoline ring was envisaged by adaptation of Wipf's oxazoline-thiazoline conversion strategy[104] (**Scheme 30**). Fragment **112** is a known compound, which was derived from L-serine in literature,[147] whereas fragment **132** is a commercial species. Fragment **142**, with a 1,2-*trans* dimethyl moiety, was proposed to access from acid **143** and hydroxylamine **144** using an indirect alkylation approach of *N,N*-dialkylhydroxylamine (**Chapter 2**), developed previously for tri-substituted hydroxylamines (**Scheme 37**). The pre-existing methyl substituent in the substrate was predicted to direct the reaction to achieve the required diastereoselectivity. Acid **143** is an accessible species from a stereoselective reaction involving a chiral auxiliary system, and *sym*-dimethylethylenediamine **145** was intended to serve as a precursor to *N,N*-disubstituted hydroxylamine **144**.
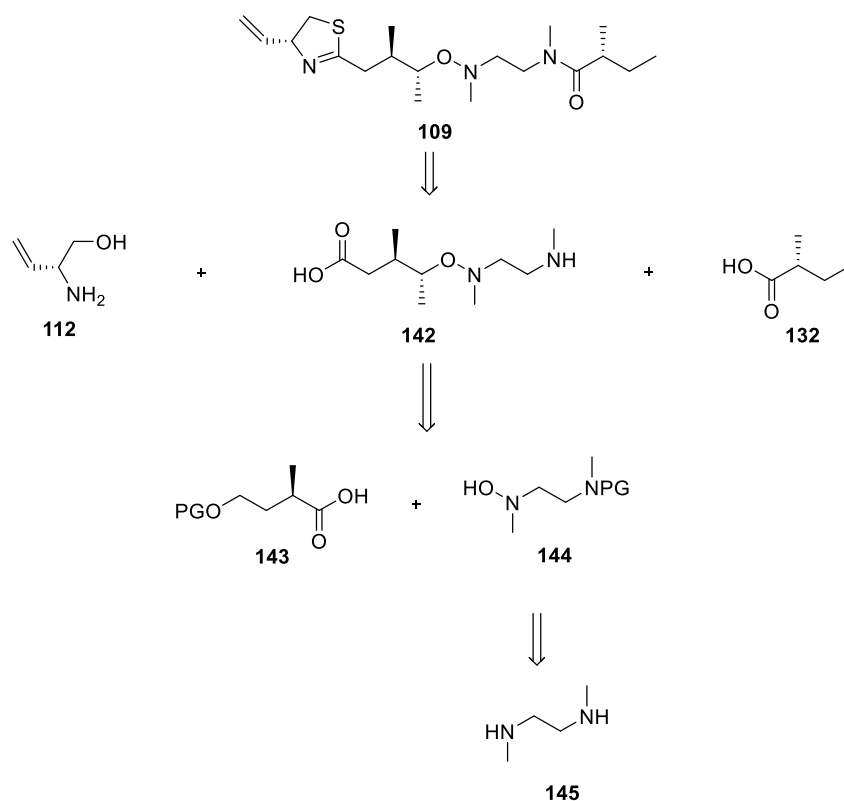
Scheme 36. Retrosynthetic analysis of 9-oxa-10-azakalkitoxin.
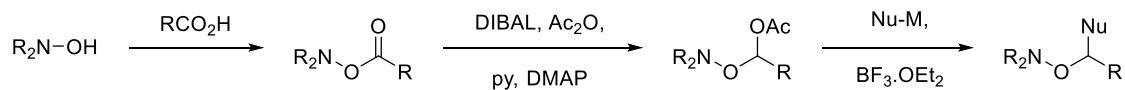
Scheme 37. Indirect alkylation of *N,N*-disubstituted hydroxylamine.

Before approaching the synthesis of hydroxalog **109**, suitable protecting groups for one of the secondary amines in *sym*-dimethylethylenediamine **145** and for the alcohol in fragment **143** were required, and their compatibility under the standard reaction conditions for tri-substituted hydroxylamine synthesis was examined. For the secondary amine, the triazene moiety was considered as a possible protecting group whereas a *tert*-butyldimethylsilyl ether was chosen for the alcohol protection. First, the compatibility was tested for triazene protecting group by targeting tri-substituted hydroxylamine **149** as a

model (**Scheme 38**). To begin the synthesis, one amino group of *sym*-dimethylethylenediamine **145** was protected as a triazene, after which the other was oxidized with benzoyl peroxide to deliver **146**. Upon reduction with DIBAL, compound **146** was converted to a *N,N*-diakylhydroxylamine, which was further acylated *in situ* to give *N*-acyloxyamine **147**. Adapting the protocol for the tri-substituted hydroxylamine synthesis, **147** was then reduced with DIBAL at -78 °C, and the resulting tetrahedral intermediate was trapped with acetic anhydride as *O*-(α-acyloxyalkyl)hydroxylamine **148** in good yield. However, the Lewis acid-mediated conversion of **148** to the tri-substituted hydroxylamine **149** was unsuccessful in the presence of dimethylzinc, as deprotection of the triazene functionality was observed after addition of $BF_3 \cdot OEt_2$, suggesting that the triazene protecting group is incompatible with the reaction conditions for the tri-substituted hydroxylamine synthesis.

Scheme 38. Compatibility test for triazene group.

Next, the carboxybenzyl (Cbz) protecting group was chosen for the secondary amine functionality (**Scheme 39**), considering its tolerance to a wide range of reaction conditions. Intermediate **150** was prepared from *sym*-dimethylethylenediamine, wherein

one amino group was protected as the benzyl carbamate and the other was benzoyloxylated. Debenzoylation of **150** with DIBAL, however, afforded *N,N*-dialkyl hydroxylamine **151** in only 52% yield, as partial deprotection of the carbamate group was observed. Alternatively, debenzoylation was best achieved by a transesterification reaction in an excellent yield.
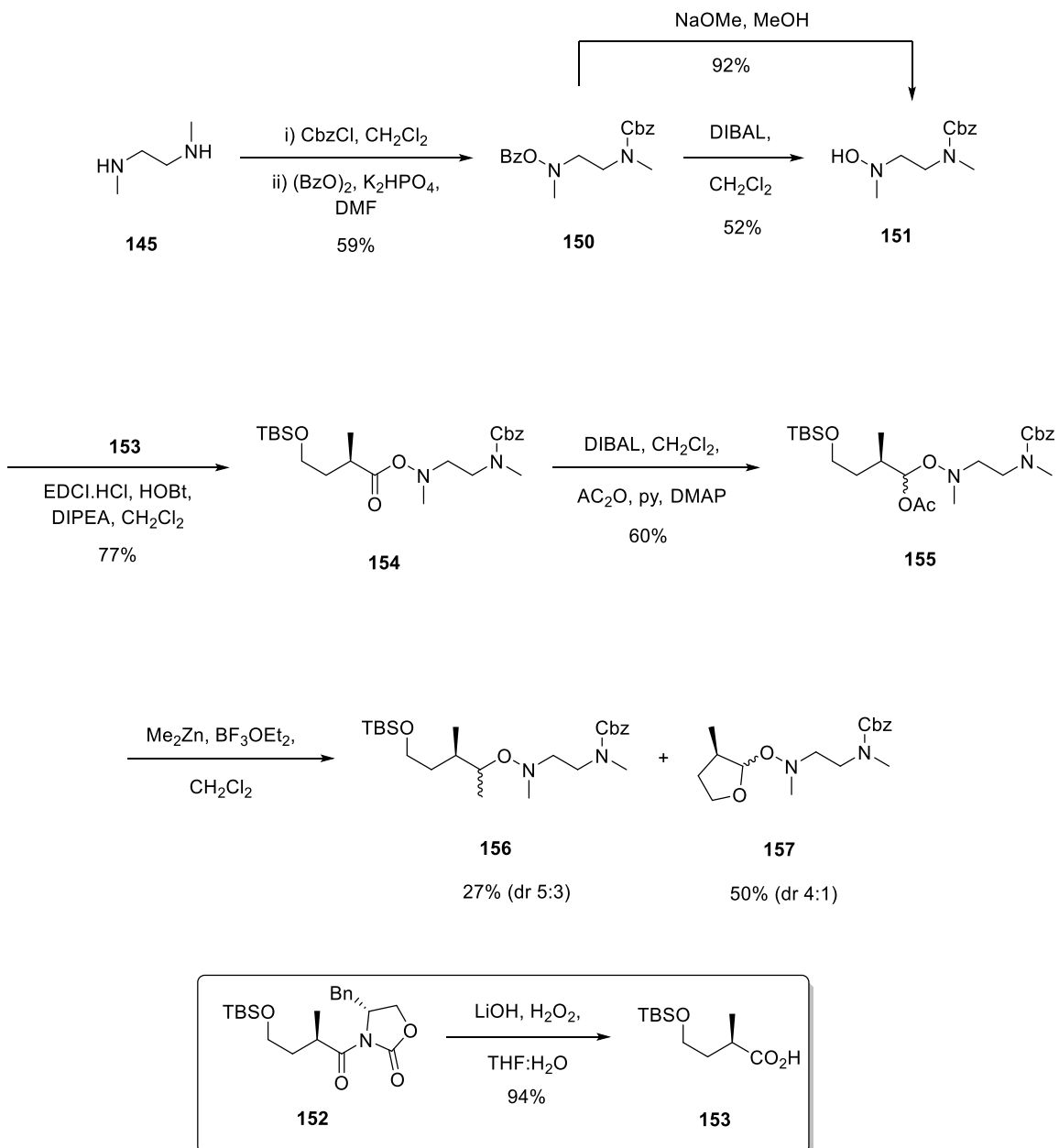
Scheme 39. Compatibility test for carbamate and silyl ether.

Following a known protocol,[148] acid **153** was prepared from the hydrolysis of compound **152**.[148] After purification, the acid **153** was isolated as a colorless oil with $[\alpha]^{20}_D$ −13.21 (*c* 8.25, EtOH). However, the specific rotation of the acid **153** was recorded with opposite sign in literature [lit[148] $[\alpha]^{20}_D$ +13.51 (*c* 1.8, EtOH)]. The so-obtained acid **153** was then used for the acylation of di-substituted hydroxylamine **151** in the presence of EDCI and hydroxylbenzotriazole (HOBt). The resulting *N*-acyloxyamine **154** was reduced with DIBAL at -78 °C followed by treatment with acetic anhydride, DMAP, and pyridine and warmed to 0 °C in order to provide *O*-(α-acyloxyalkyl)hydroxylamine **155**. When **155** was subjected to Lewis acid-mediated deacetoxylation followed by the reaction with dimethylzinc, two products, **156** and **157**, were isolated in 27% and 50% yields, respectively. From the NMR spectra, **156** was analyzed as a mixture of two diastereomers of the desired product in a ratio of 5:3, and **157** was characterized as a mixture of two anomers of a cyclic derivative in a 4:1 ratio. The individual isomers in both of the mixtures were not characterized due to the presence of rotamers in the NMR spectra. The aminoxy sugar derivatives in **157** are the cyclized products resulting from nucleophilic attack of the silyl ether oxygen on the two faces of aminoxocarbenium ion intermediate. After observing the cyclized products as the major outcome, the need for an alternative alcohol protecting group was recognized. However, the carbamate protection for the secondary amine was compatible with the reaction conditions.

To overcome the problem of cyclization, acid **153** was replaced with acid **160**,[149] which carries an olefinic bond, instead of the silyl ether, and which can be converted to the desired alcohol functionality whenever needed (**Scheme 40**). Acid **160** was prepared by stereoselective alkylation of enolate of (*S*)-4-benzyl-3-propionyloxazolidin-2-one

(**158**)[150] with cinnamyl bromide, followed by lithium peroxide-mediated hydrolysis**.** The so-obtained acid **160** was then used for acylation of *N,N*-dialkylhydroxylamine **151** to isolate *N*-acyloxyamine **161**. Upon reduction with DIBAL at a low temperature, followed by the treatment with acetic anhydride, pyridine, and DMAP, **161** was transformed to *O*-(α-acyloxyalkyl) hydroxylamine **162**. The subsequent subjection of **162** to a Lewis acid promoted deacetoxylation, followed by the treatment with dimethylzinc, gave only the desired product **163** in 62% yield as a 4:3 diastereomeric ratio, proving the compatibility of the olefinic double bond to the reaction conditions. However, the poor selectivity, coupled with difficulties in separation and characterization for each diastereomer, necessitated an alternative route for the hydroxalog **109**.
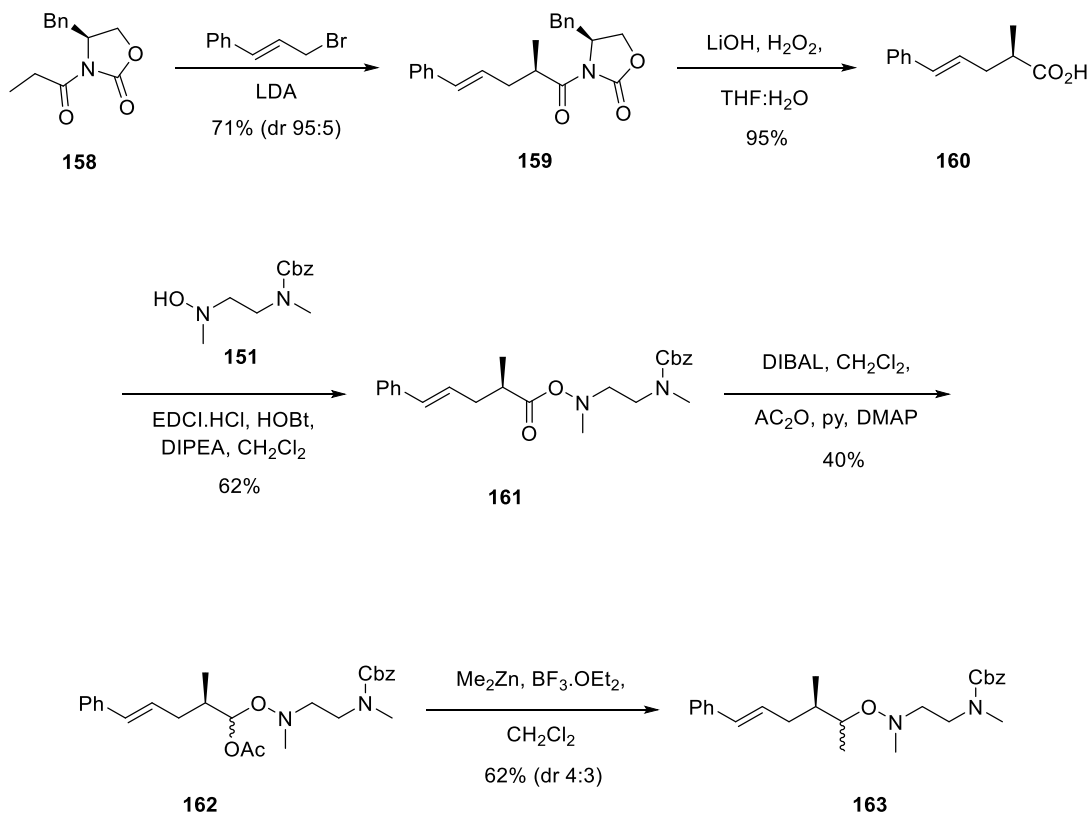
Scheme 40. Synthesis of tri-substituted hydroxylamine **163**.

In a view of the difficulty in installing the hydroxylamine moiety stereoselectively, an alternative retrosynthesis (**Scheme 41**) was proposed for hydroxalog **109**. The strategy for the thiazoline ring construction remained the same as depicted in the previous synthetic plan. Nevertheless, disconnection of acid **164** was proposed such that it could be obtained from *N,O*-disubstituted hydroxylamine **165** and aldehyde **166**, exploiting a classical reductive amination-like reaction. The enantiomerically pure acid **132** was chosen as a precursor to aldehyde **166**. The synthesis of hydroxylamine **165** was planned to begin from a literature compound **167**,[151] wherein the two methyl groups in 1,2-*anti*-fashion will be incorporated directly from **167**, and the hydroxylamine moiety will be introduced by amination of terminal benzylated hydroxyl group after hydrogenolysis.
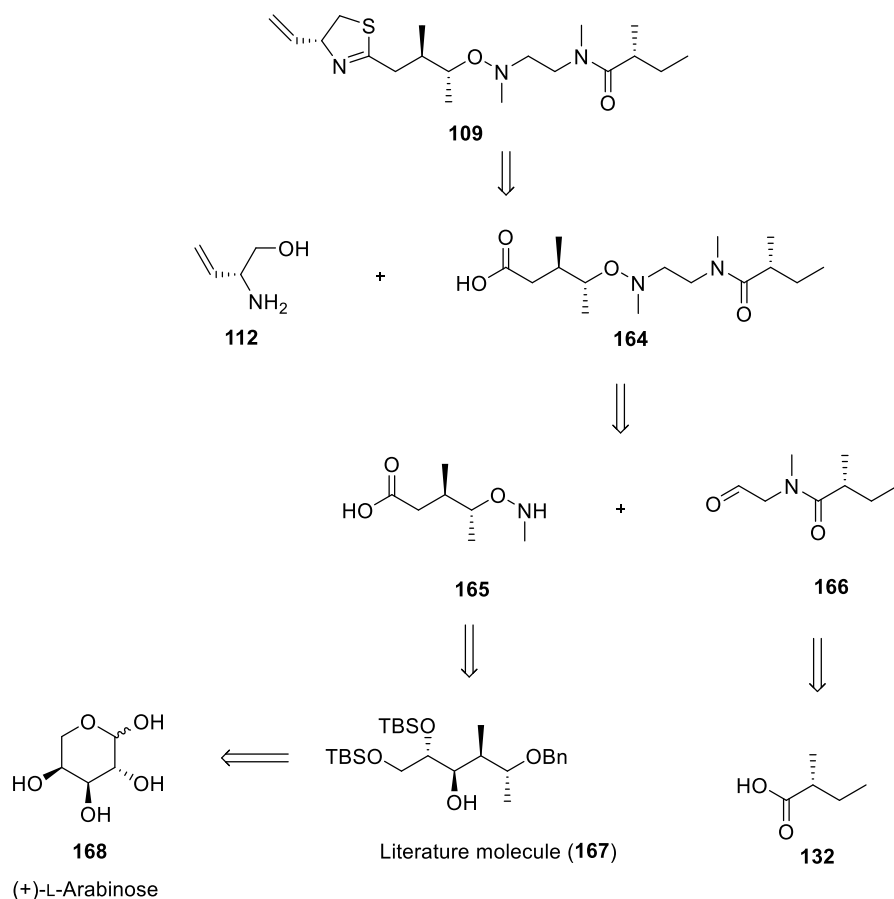
Scheme 41. An alternative retrosynthesis for hydroxalog **109**.

To start the synthesis, literature compound **167** was synthesized on a gram scale in 8 steps following the literature protocol[151] from (+)-L-arabinose in 33% overall yield (**Scheme 42**). Conversion of **167** to imidazolyl thioether **169** was achieved in excellent yield by stirring with thiocarbonyldiimidazole (TCDI) at 50 °C. Upon subjection to Barton's radical deoxygenation, followed by hydrogenolysis, thioether **169** was converted to alcohol **170**.

Scheme 42. Synthesis of alcohol **170**.

In order to introduce the hydroxylamine moiety to alcohol **170**, direct amination of the hydroxyl functionality was conceived. However, methods for hydroxylamine synthesis via the construction of the N-O bond are extremely rare in the current toolbox due to several unsolved challenges as described in **Chapter 1**. Although not common, the Boger lab was successful in forming an N-O bond during the syntheses of the prodrugs of duocarmycin via amination of aryl alcohol with carbamate tosylates and *N*-hydroxyphthalimide as aminating agents (**Scheme 43**).[152]

Scheme 43. Examples of amination of aryl alcohols from the Boger lab.

Before attempting amination on **170**, the feasibility of the reaction was studied on 1-phenyl-2-propanol as a model (**Scheme 44**). A series of aminating reagents, NH_2Cl, H_2N-OTs, CbzHN-OTs, and PhthN-OTs, were synthesized[153-155] and were tested for the substitution reactions with 1-phenyl-2-propanol in basic media. Unfortunately, all attempts at amination of this alcohol were unsuccessful.

Base = NaH, KOtBu, K_2CO_3, NaHMDS

R_2N-X = NH_2Cl, H_2N-OTs, FmocHN-OTs, CbzHN-OTs, PhthN-OTs
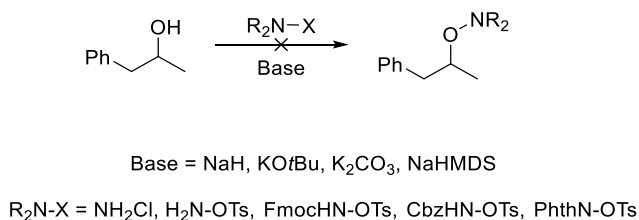
Scheme 44. Test reactions for the amination of alcohol.

As the direct N-O bond formation was unsuccessful in a model substrate, the hydroxylamine functionality was installed by utilizing the Mitsunobu reaction, a classical approach for hydroxylamine synthesis (**Scheme 45**). First, the configuration of the alcohol was inverted by a Mitsunobu reaction with nitrobenzoic acid, followed by transesterification to access alcohol **172**. A second Mitsunobu reaction with *N*-hydroxyphthalimide resulted in hydroxylamine **173** with overall retention of the configuration.
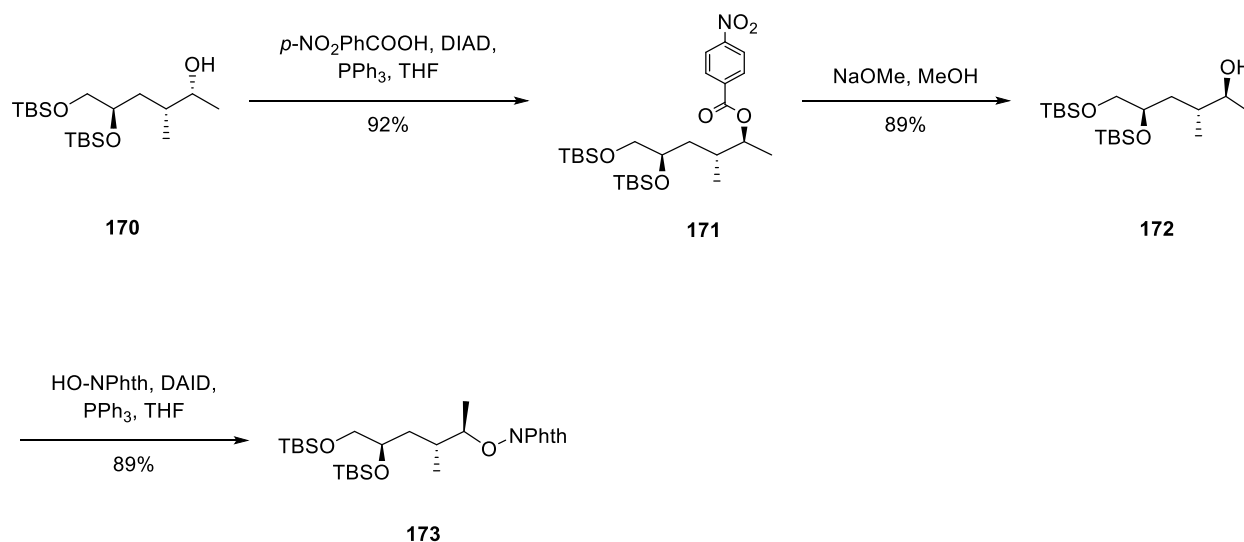
Scheme 45. Installation of the N-O bond by Mitsunobu reaction.

Before moving forward with the hydroxylamine intermediate **173**, aldehyde **166** was synthesized from acid **132** in two steps (**Scheme 46**). Acid **132** was first coupled with *N*-allylmethylamine in the presence of HOBt and EDCI and the resulting amide **174** was then subjected to reductive ozonolysis to access crude aldehyde **166**.
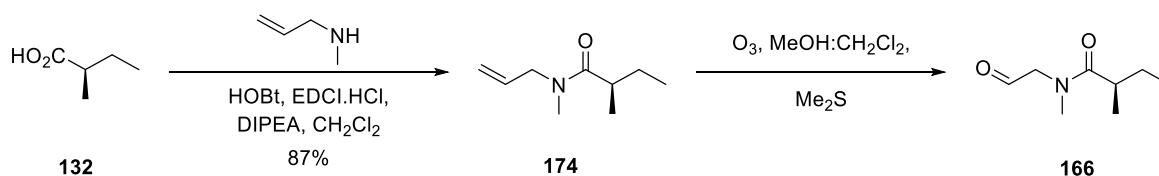
Scheme 46. Synthesis of aldehyde **166**.

After the synthesis of aldehyde **166**, the hydroxylamine intermediate **173** was extended to hydroxalog **109** (**Scheme 47**). Deprotection of the hydrazine functionality in **173** with hydrazine hydrate, followed by the treatment with paraformaldehyde, delivered oxime **175** in an excellent overall yield over 2 steps. Further reduction of this oxime was carried out by using sodium cyanoborohydride as a reductant in an acidic medium, and the resulting *N,O*-disubstituted hydroxylamine intermediate was subjected to reductive amination conditions with crude aldehyde **166** in order to generate tri-substituted

hydroxylamine **176**. Compound **176** was then converted to acid **164** via a three-step reaction sequence of tetrabutylammonium fluoride (TBAF)-mediated deprotection of 1,2-diol, oxidative cleavage to aldehyde, and Pinnick oxidation. Transformation of the so-obtained acid **164** to amide **177** was accomplished by amidation with amino alcohol **112** with the aid of EDCI and DMAP. The conversion of amide **177** to 2-oxazoline **178** was best achieved by diethylaminosulfur trifluoride (DAST)-mediated cyclization at -78 ^{o}C. Finally, Wipf's oxazoline-thiazoline conversion sequence was utilized to convert oxazoline derivate **178** to hydroxalog **109**, wherein the oxazoline ring was first opened to the thioamide by stirring with hydrogen sulfide-saturated methanolic triethylamine and then cyclized to the thiazoline ring in the presence of DAST at -78 ^{o}C. The hydroxalog **109** was isolated as a colorless oil and as a single stereoisomer. Like in kalkitoxin, a mixture of rotamers is observed in the NMR spectra of **109** due to amide functionality.
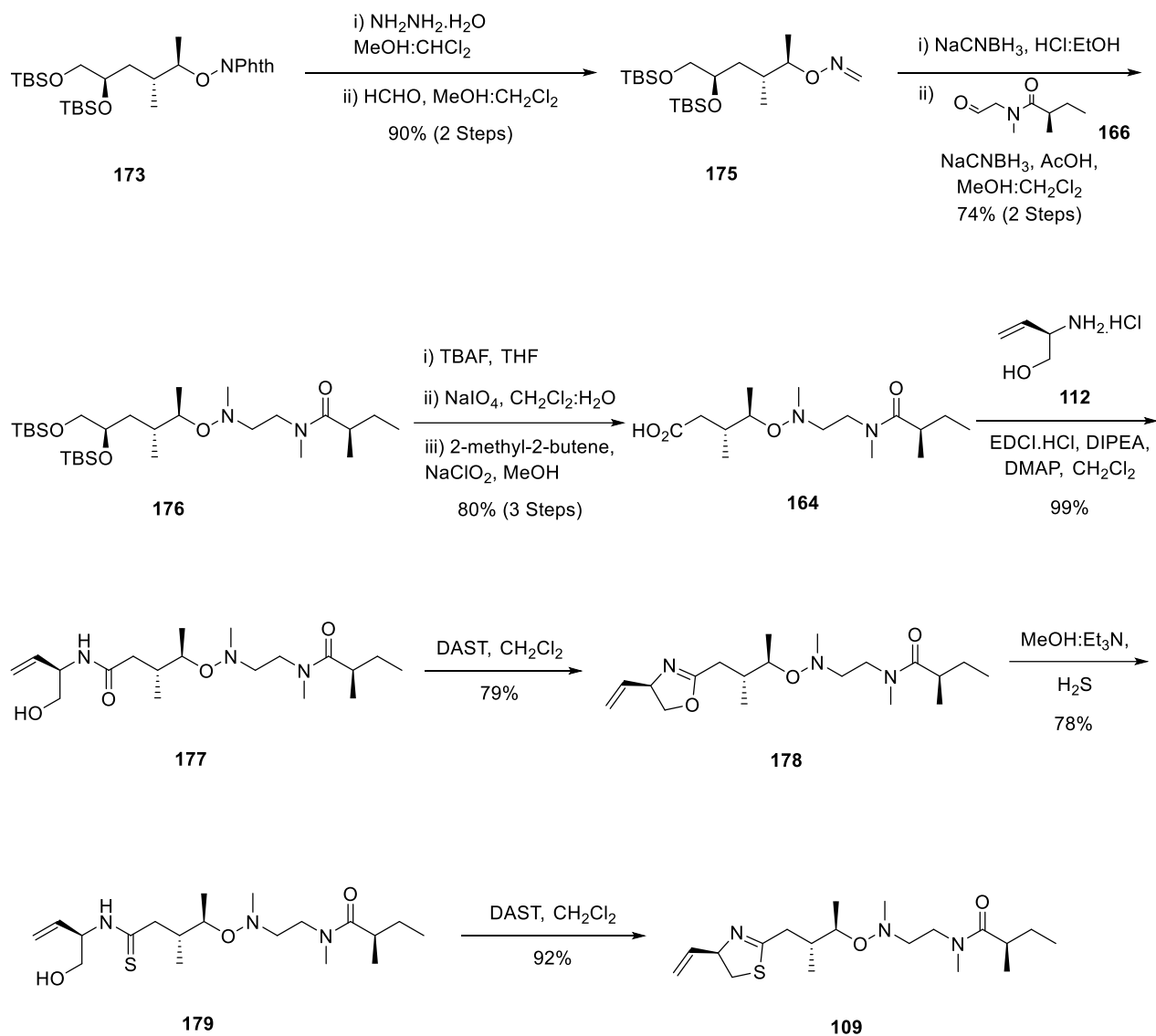
Scheme 47. Synthesis of the hydroxalog **109** from the hydroxylamine intermediate **173**.

In summary, a total synthesis of the novel hydroxalog **109** of natural kalkitoxin was accomplished in 25 linear steps, starting from a commercially available (+)-L-arabinose in 3% overall yield. The hydroxylamine moiety in the molecule was installed by utilizing the Mitsunobu reaction, and reductive amination was applied to generate the tri-substituted hydroxylamine intermediate **176** from its *N,O*-disubstituted precursor. The thiazoline ring in the molecule was constructed by adapting Wipf's oxazoline-thiazoline conversion

strategy from oxazoline derivative **178**, which itself was synthesized via DAST-mediated cyclization of amido alcohol intermediate **177**.

4.4. Synthesis of 7,8-diazakalkitoxin (110)
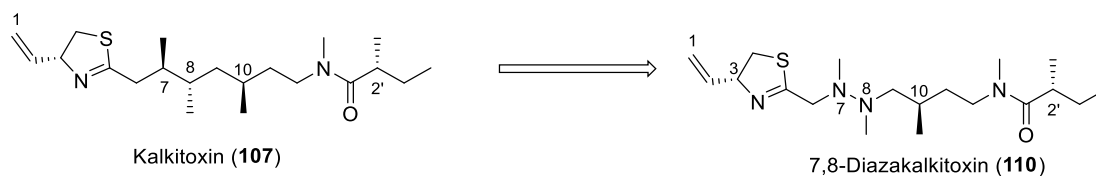
Figure 35. Structure of kalkitoxin and its hydrazine analog.

The retrosynthetic analysis for the hydrazine analog **110** is presented in **Scheme 48**. The synthesis of analog **110** was proposed from fragments **180**, **181**, and **132**. A Lewis acid-mediated cyclization was chosen to construct the thiazoline ring, by adapting protocol from White's kalkitoxin synthesis.[105] Compound **180**, with a suitable protecting group for the thiol functionality, was designed to synthesized from L-cysteine. Reductive amination was planned to deliver tetra-substituted hydrazine core **181** from tri-substituted hydrazine **183** and known aldehyde **184**,[156] whereas **183** itself was proposed to generate from *sym*-dimethylhydrazine **185**, and cinnamyl bromide **186** was chosen as an initial precursor to aldehyde **184**.
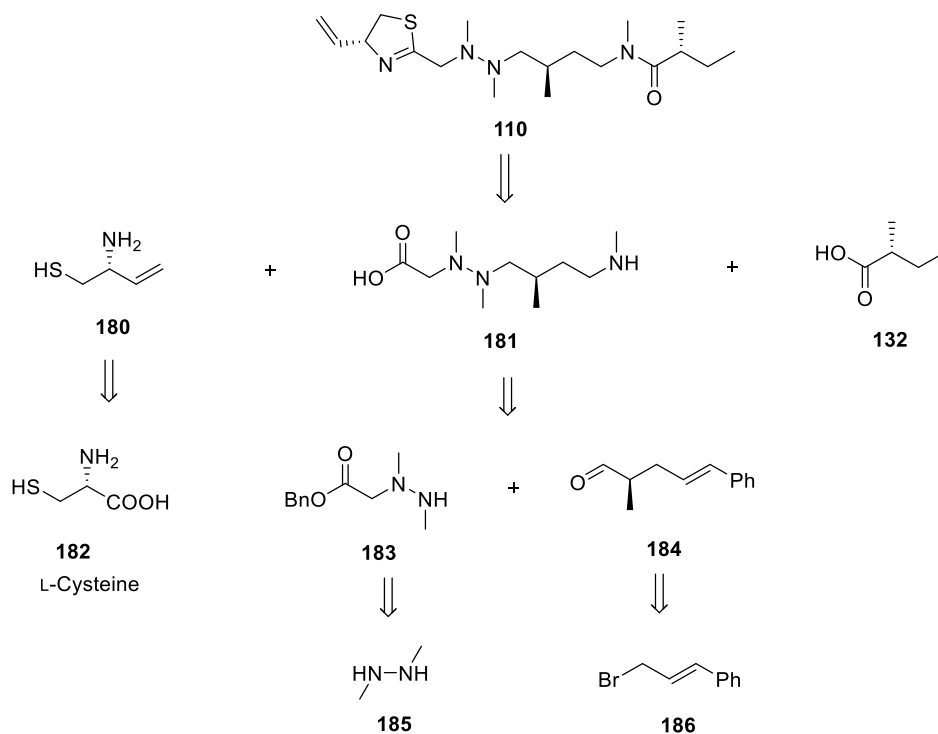
Scheme 48. Retrosynthetic analysis of 7,8-diazakalkitoxin (**110**).

The synthesis of hydrazine analog **110** commenced with the synthesis of enantiomeric aldehyde **184**[156] as described in **Scheme 49**. Alcohol **187**[157] was obtained from reduction of **159**, a cinnamyl bromide-derived oxazoline derivative (**Scheme 40**), with LiBH$_4$ as a reductant. Dess-Martin periodinane (DMP)-mediated oxidation of the resulting alcohol **187** gave the desired aldehyde **184**.
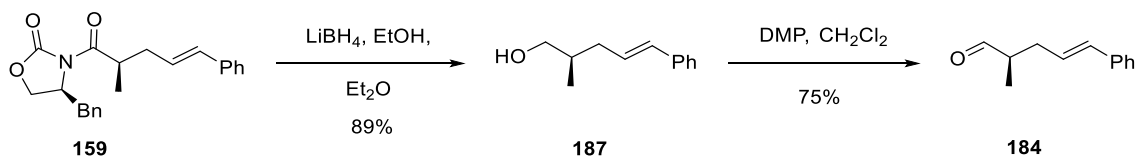
Scheme 49. Synthesis of an aldehyde **184**.

The tri-substituted hydrazine **183**, a product resulting from the controlled mono-alkylation of *sym*-dimethylhydrazine **185** with benzyl bromoacetate,[158] was subjected to NaCNBH$_3$-assisted reductive amination with aldehyde **184** to obtain **188** (**Scheme 50**).

Upjohn dihydroxylation[159] of the resulting tetra-substituted hydrazine backbone **188**, followed by the NaIO$_4$-mediated oxidative cleavage, gave aldehyde **189** in 52% overall yield over two steps. Conversion of this aldehyde **189** to amine **190** was best achieved via a reductive amination reaction. After acylation of the secondary amine **190** with commercial acid **132** in the presence of HOBt and EDCI, amide **191** was furnished in 67% yield.
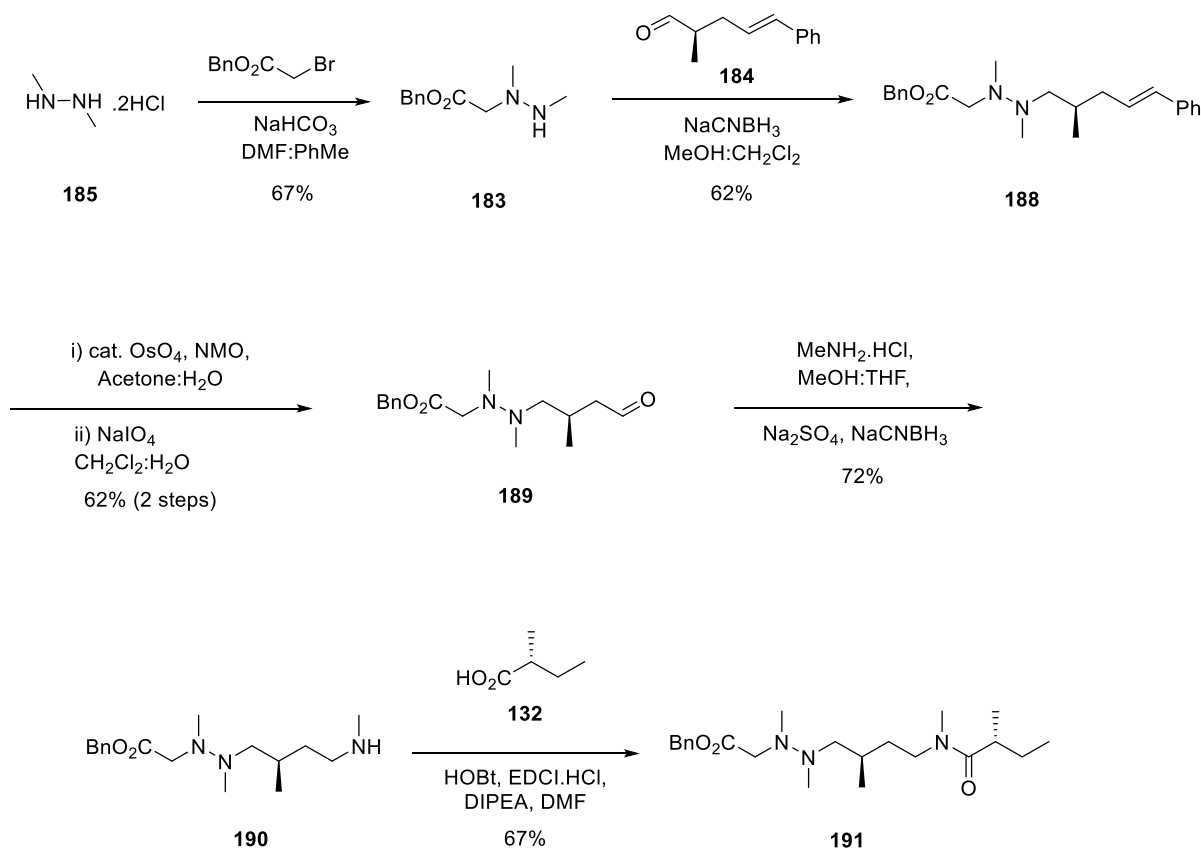
Scheme 50. Synthesis of tetra-substituted hydrazine core **191**.

Before advancing to the next stage, synthesis of vicinal amino thiol **180** (**Scheme 48**) with a suitable protecting group on the thiol functionality was necessary. Earlier, White and co-workers synthesized the benzyl thioether derivative of amino thiol **180** for the synthesis of kalkitoxin.[105] However, application of the harsh reaction conditions for the

benzyl thioether deprotection were predicted to be impractical in the presence of the hydrazine moiety. Therefore, a protecting group with a comparatively milder deprotection conditions was required. The replacement of the benzyl thioether with the trimethoxybenzyl (Tmob) thioether protecting group was considered, as milder reaction conditions (e.g., stirring with a mixture of trifluoroacetic acid and triethylsilane) can be applied for deprotection of such functionality.[160] Therefore, amino trimethoxybenzyl thioether **196** was synthesized (**Scheme 51**) starting from L-cysteine by slight modifications of the protocol of benzyl analog synthesis reported by White and co-workers.[105]
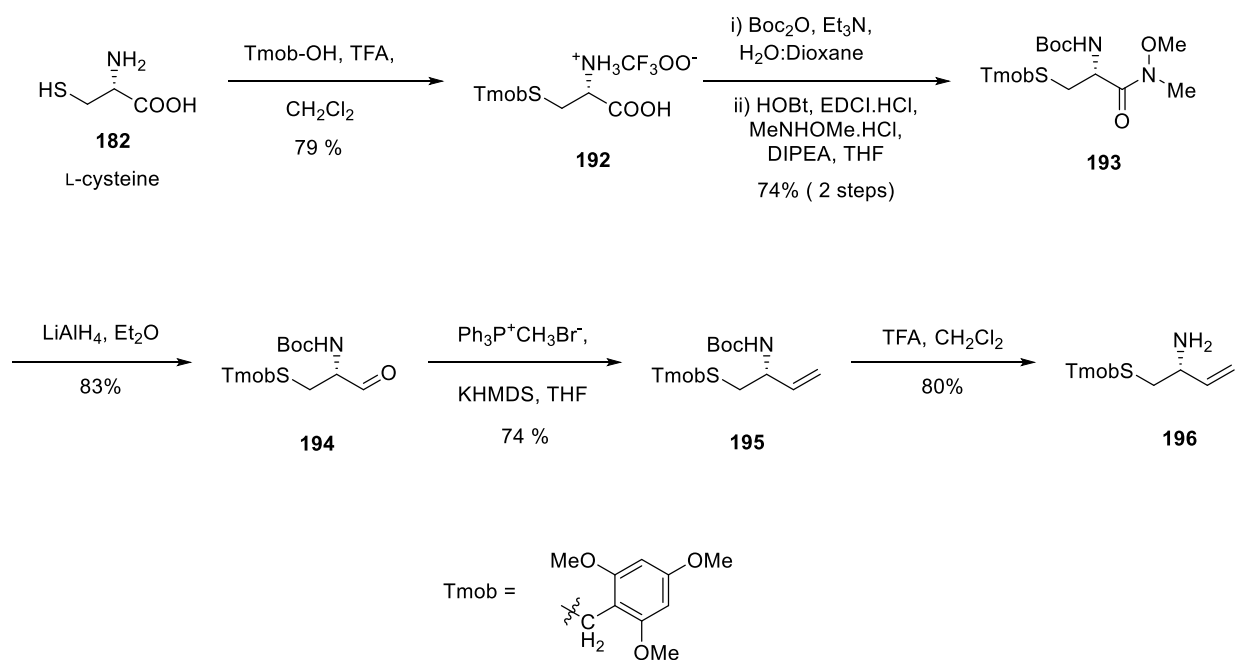
Scheme 51. Synthesis of amino trimethoxybenzyl thioether **196**.

The thiol group of L-cysteine was masked as the trimethoxybenzyl thioether and was isolated as its trifluoroacetate salt **192**.[160] The amino group was protected as the *tert*-butylcarbamate, and the acid functionality was converted to a Weinreb amide. The resulting amide **193** was reduced to aldehyde **194**, which was then homologated to alkene

195 via a Wittig reaction. Subsequent deprotection of the amine functionality with trifluoroacetic acid offered amine **196**.

After unmasking the acid functionality of amide **191** by hydrogenolysis (**Scheme 52**), it was coupled with amine **196** in the presence of hexafluorophosphate azabenzotriazole tetramethyl uranium (HATU) to afford intermediate **197**. Unmasking of the thiol functionality was best achieved by stirring with trifluoroacetic acid in the presence of triethylsilane as a cation scavenger. The resulting amido thiol **198** was exposed to titanium tetrachloride to induce cyclization to obtain targeted analog **110**, by adapting the protocol from White's kalkitoxin synthesis.[105] Unfortunately, no cyclization was detected and only precipitation was observed in the reaction mixture, leading to the conclusion that the hydrazine moiety of the precursor complexed with the titanium salts. Several other reaction conditions (e.g., using TMSOTf as Lewis acid or adding Tf$_2$O in basic condition) were tested, however, no thiazoline ring formation was observed under these conditions.
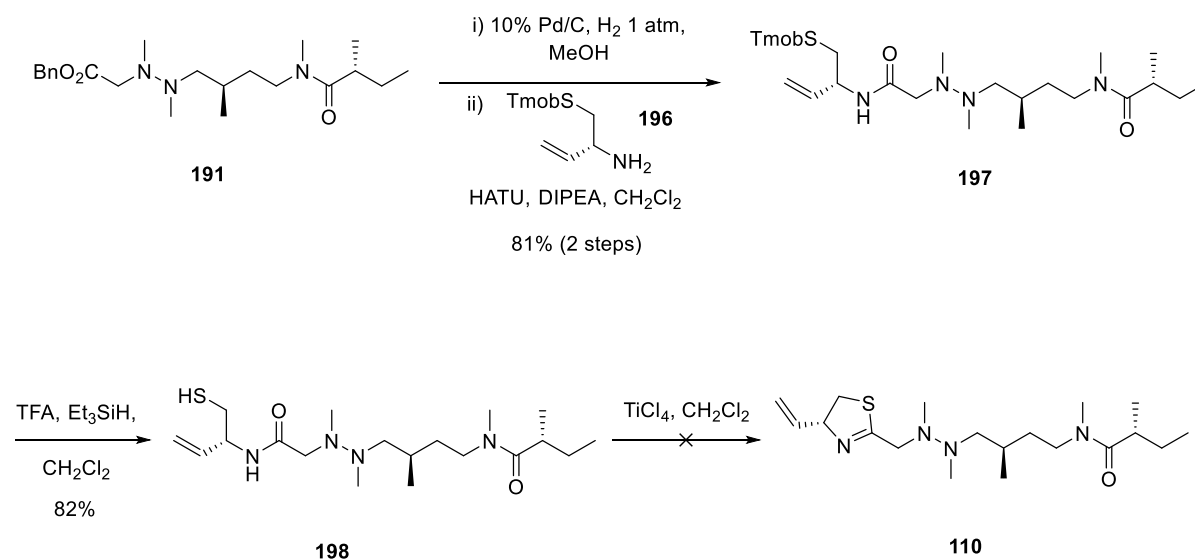
Scheme 52. An unsuccessful attempt at the hydrazine analog synthesis.

The failure of the Lewis acid-mediated cyclization to thiazoline ring necessitated an alternative route. Wipf's oxazoline-thiazoline conversion reaction sequence,[104] which was previously applied in the synthesis of the hydroxalog and a kalkitoxin itself, was chosen as a promising path to deliver the target. However, before using this approach in the target molecule, it was considered prudent to test it in a model substrate with similar functionalities. Therefore, compound **201** (**Scheme 53**) was synthesized as a model from the tri-substituted hydrazine **78** (**Chapter 2**, **Scheme 25**) in a three-step reaction sequence; i) substitution reaction of **78** with benzyl bromoacetate to provide tetra-substituted hydrazine **199**, ii) saponification of ester **199** to afford acid **200**, and iii) amidation of acid **200** to obtain amino alcohol **201**.
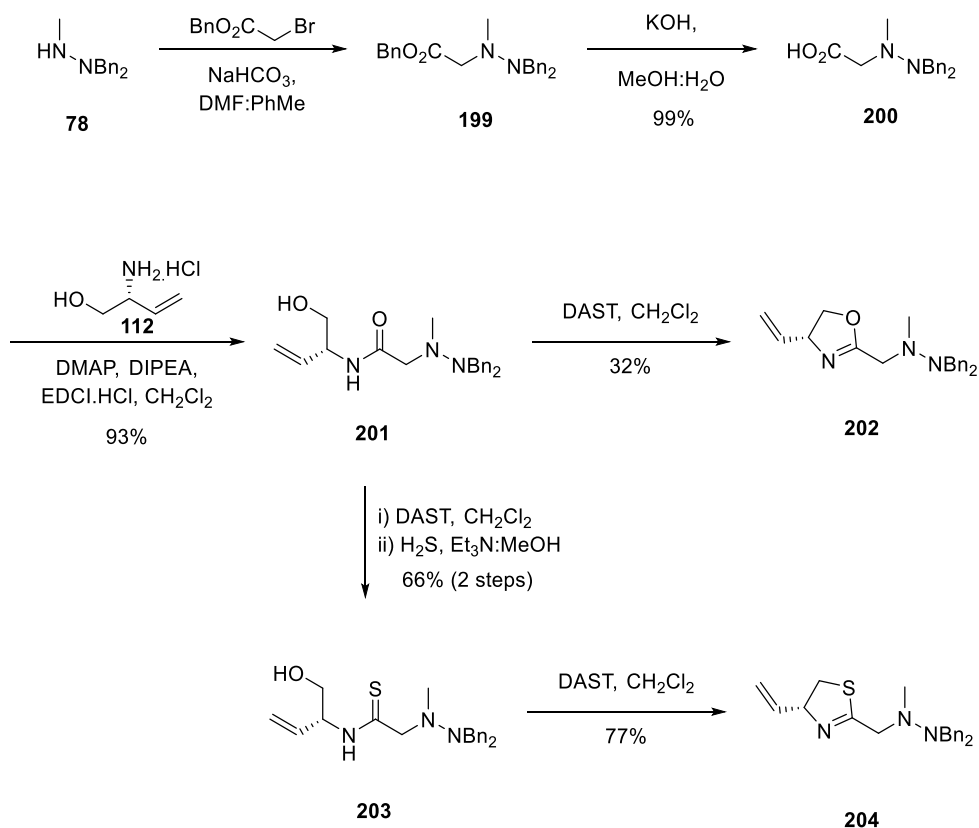
Scheme 53. Synthesis of a model thiazoline derivative.

With substrate **201** in hand, Wipf's oxazoline-thiazoline conversion reaction sequence was tested. On treatment of amide **201** with DAST at -78 °C, cyclization was completed within 5 minutes, but, the cyclized product **202** was isolated in only 32% yield, together with the recovery of 40% of starting material **201** after purification. After careful observation, derivative **202** was found to be unstable due to facile reversion to its precursor alcohol **201**. When product **202** was stirred with hydrogen sulfide saturated methanolic triethylamine without purification, thioamide **203** was isolated in 66% overall yield in two steps, together with traces of alcohol **201**. Upon subsequent DAST-mediated cyclization reaction, thiazoline derivative **204** was isolated in 77% yield. The thiazoline moiety in **204** was found to be more stable than that of the oxazoline moiety in **202**, since ring-opening was not observed during the synthesis. The observed facile ring opening process of the oxazoline derivative can be viewed as the manifestation of the electron withdrawing effect of the hydrazine moiety, which is responsible for high electrophilicity of the sp^2 carbon of oxazoline ring. The stability of the thiazoline derivative compared to the oxazoline derivative is attributable to the difference in electronegativity of sulfur and oxygen atom in the ring.

After a successful cyclization sequence on the model substrate, the reaction conditions were employed for the synthesis of hydrazine analog **110** (**Scheme 54**). Amido alcohol **206** was obtained in 70% yield from the intermediate **191** by hydrogenolysis, followed by coupling the intermediate acid with amine **112**. In DAST-mediated cyclization, oxazoline derivative **207** was isolated in 30% yield, together with the recovery of 35% yield of starting material **206**, since the ring opening phenomenon similar to **202** was observed. Direct conversion to thioamide **208** without purification of **207** was achieved in

52% isolated overall yield over 2 steps. Eventually, the targeted hydrazine analog **110** was isolated in 89% yield from the thioamide **208** upon treatment with DAST. Here the thiazoline derivative **110** was also more stable than of its oxazoline counterpart **207**. The hydrazine analog **110** was isolated as a colorless oil and as a single stereoisomer. Like in kalkitoxin, a mixture of rotamers was observed in the NMR spectra of **110** due to the amide functionality.
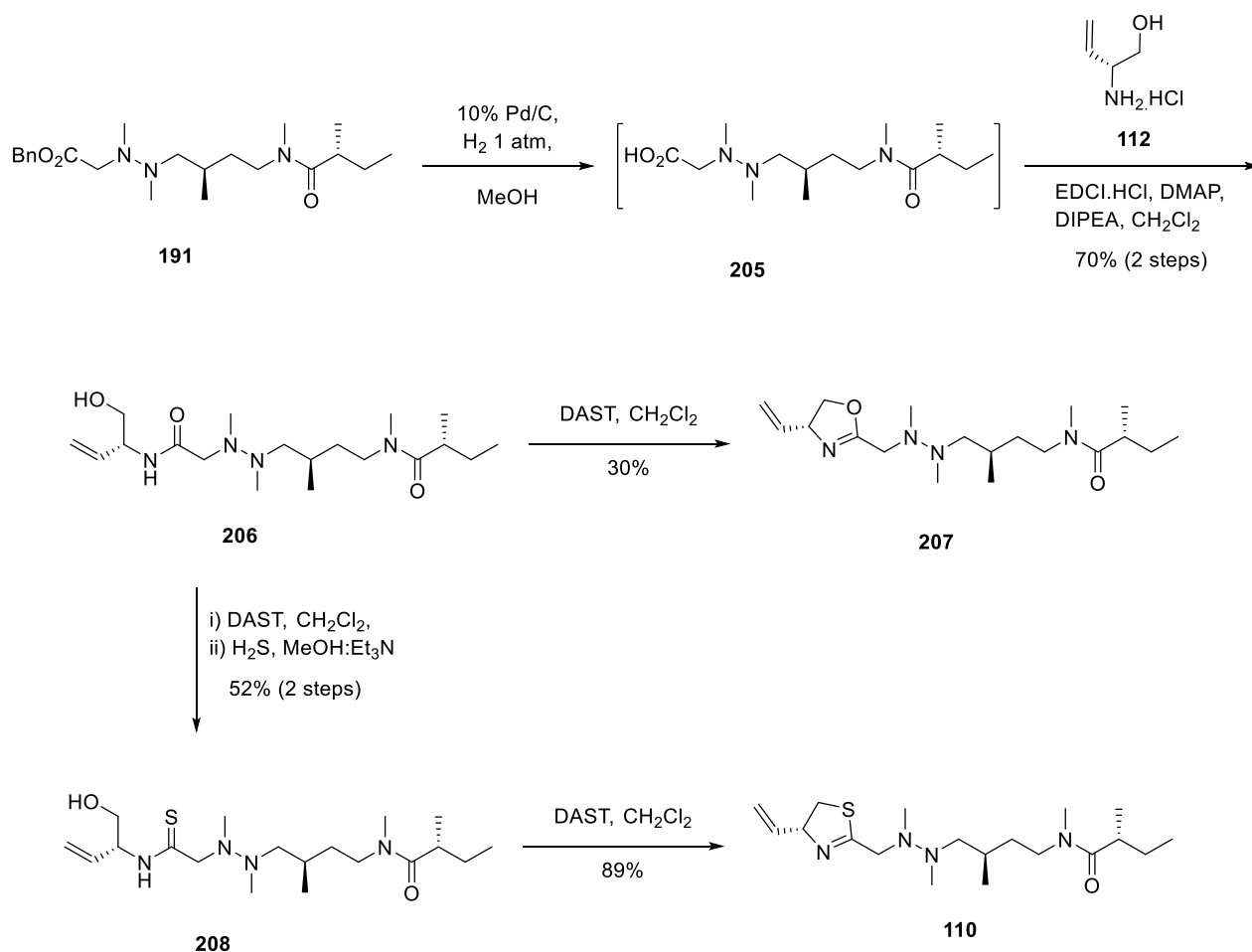
Scheme 54. Synthesis of hydrazine analog **110** of kalkitoxin.

To summarize, the hydrazine analog **110** was synthesized in 11 linear steps starting from commercially available *sym*-dimethylhydrazine in 4% overall yield. As expected, the modification of natural kalkitoxin to the hydrazine analog considerably

simplified the synthesis by reducing the number of steps from 21 to 11. A classical S_N2 substitution reaction was utilized to convert a di-substituted hydrazine to a tri-substituted hydrazine (**185** to **183**), and reductive amination delivered a tetra-substituted hydrazine backbone **188**. Wipf's oxazoline-thiazoline ring conversion strategy was used to construct thiazoline ring. The oxazoline ring was found to be labile to ring opening due to the perceived electron withdrawing effect of the hydrazine moiety, whereas, the thiazoline ring was found to be relatively stable.

4.5. Biological assessments

After completing the syntheses of kalkitoxin (**107**) and its hydroxylamine and hydrazine analogs (**109** and **110**), they were submitted for cytotoxicity assays against various cell lines in the Valeriote laboratory at Henry Ford Hospital in Detroit. The oxazoline analogs (**137**, **178**, **207**) were also submitted to study the importance of the thiazoline ring on the biological activity. All compounds were subjected to *in vitro* disk diffusion assay to quantify differential cell killing against the eleven human cell lines; one leukemia (CCRF-CEM), one normal (CFU-GM), and nine solid tumors. The human solid tumor cell lines were Lung (H125), Ovarian-(OVCAR-5), pancreatic (PANC-1), Liver (HepG2), Brain (U251N), prostate (LNCaP), colon (H-116), and breast (MCF-7 and MDA).

The *in vitro* disk diffusion assay was designed to identify the solid tumor selective compounds by collaborative effort of the Valeriote laboratory at Henry Ford Hospital in Detroit and the Crews laboratory at the University of California in Santa Cruz.[161] Human lymphocytic leukemia cells, CCRF-CEM, were used as a reference tumor for the assay. The second reference cell line chosen was a normal cell (CFU-GM). However, before running assays on expensive human cell lines, the selectivities of the compounds against

murine colon cancer cell (Colon38) in relation to murine leukemia (L1210) and murine normal cell (CFU-GM) were conducted. These murine cell lines were selected as they closely resemble human cell lines. There are four positive results from these assays; i) a compound is selective to the murine solid tumor over leukemia, ii) a compound is selective to the murine solid tumor over normal cells, iii) a compound is selective to the human solid tumor over leukemia, and iv) a compound is selective to the human solid tumor over normal cells. This unique and effective zone assay guides the discovery of novel molecular structures with solid tumor selective anticancer properties.
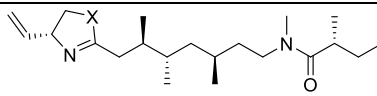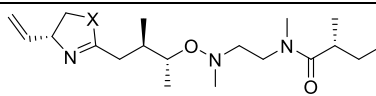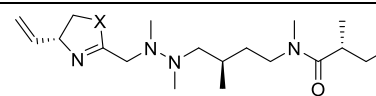
In *in vitro* disk diffusion assay, samples were solubilized in 0.5 mL DMSO; 15 μL of each sample was applied to a 6.5 mm filter disk, which was allowed to dry overnight and then placed close to the edge of the petri dish. Depending upon the cell type, the plates were incubated for 7-10 days and the zones of inhibition were examined by measuring from the edge of the filter disk to the beginning of normal-sized colony formation. The diameter of the filter disk was arbitrarily taken as 200 units. A sample is selective if the zone difference between solid tumor cells and either normal or leukemia cell is greater than 250 units and is expressed as, for example, $_{H-125}\Delta_{CEM}$ = 400, which indicates that there is a 400 unit zone differential between human colon H-125 and leukemia CCRF-CEM. If the compound is excessively toxic at the first dosage, the sample solution was diluted (at 1:4 decrements) and retested against the same tumors. At some dilution, quantifiable cytotoxicity was invariably obtained.

As the assay is under progress in the Valeriote lab, complete data are not available currently. **Table 7** presents the available data from *in vitro* disk diffusion assay. Since the zone inhibition data for kalkitoxin **107** and its analogs **137**, **109**, and **178** against reference

cells (normal cells and leukemia) are not available currently, the quantifications of selectivity of these compounds are not possible. However, kalkitoxin was found to be highly cytotoxic against most of the cancer cell lines as expected. The toxicity of the kalkitoxin decreased slightly when the thiazoline ring was changed to oxazoline. Interestingly, the 9-oxa-10-azakalkitoxin (**109**) was found equally cytotoxic as parent kalkitoxin. The potency of its oxazoline analog (**178**) was reduced against most of the cell line except against hepato carcinoma cell (HepG2).

In contrast, cytotoxicity of hydrazine analogs **110** and **207** were found to be comparatively poor against all cancer cell lines except murine colon tumor cell line (colon 38), against which the potencies were average. The observed weak cytotoxicity of these analogs could be attributable to their less stable thiazoline or oxazoline rings. As the zone inhibition data for hydrazine analogs **110** and **207** against reference cells except human normal cells are available, quantifications of their selectivity are possible. Both the hydrazine analogs **110** and **207** show differential cytotoxicity for murine colon 38 versus normal cell ($_{38}\Delta_{CFU}$ = 550 for **110** and 350 for **207**). In addition to murine colon 38, hydrazine analog **110** also shows differential cytotoxicity for murine colon 38 versus leukemia ($_{38}\Delta_{L1210}$ = 450) and for human brain tumor cell (U251N) versus leukemia ($_{U251N}\Delta_{CEM}$ = 300).

Table 7. *In vitro* disk diffusion assay report for kalkitoxin and its analogs.

107, X = S
137, X = O

109, X = S
178, X = O

110, X = S
207, X = O

Compound	Dilution	Murine			Human										
		L1210	Colon 38	CFU-GM	H-116	H-125	MCF-7	LNCaP	OVC-5	U251N	MDA	PANC-1	HepG2	CEM	CFU-GM
107	10 mg/ml						700	1000	600	950		600	>1000		
	1/4						550	800	500	850		600	>1000		
	1/16						800	700	300	750		650	>1000		
	1/64						550	500		600					
137	6 mg/ml				800	>1000			1000						
	1/4				500	450		600	800	550		600	900		
	1/16				250	300	50		600				500		
109	6 mg/ml				>1000	>1000			>1000						
	1/4				800	900		900	>1000	1000		800	>1000		
	1/16				650	600	50		1000	900		500	950		
	1/64					300			800	700		300	750		
178	6 mg/ml				800	750	750	1000	650	800		800	1000		
	1/4				250	400				500		200	600		
110	10 mg/ml	350	800	250	200	200	100	250	100	500	400	100	300	200	
207	6 mg/ml	400	600	250	100	100	50	200	50	100	250	0	100	200	

IC_{50} values were determined for kalkitoxin **107**, hydroxylamine analog **109,** and oxazoline analogs **137** and **178** against liver cancer cell line (HepG2) and the results are presented in **Table 8**. Comparison of data indicates that 9-oxa-10-azakalkitoxin **109** and kalkitoxin are equally potent, which proves the concept of hydroxylamines as convertible mimics of stereogenic centers. Analog **137** is 50 fold less potent than kalkitoxin and analog **178** is 500 fold less potent than 9-oxa-10-azakalkitoxin **109**. These data together with the zone inhibition data demonstrate that the thiazoline moiety of the kalkitoxin and 9-oxa-10-azakalkitoxin are needed for the potency of the compounds.

Table 8. IC_{50} values for kalkitoxin and its analogs against human liver cancer cell line (HepG2).

Compound	HepG2 (IC_{50}, $ng.mL^{-1}$)
107	3.2
137	1.6×10^2
109	2.4
178	1.5×10^3

4.6. Conclusion

Kalkitoxin was chosen as a candidate to test the idea of hydroxylamine and hydrazine analogs. Analogs **108** and **109** were designed as hydroxylamine analogs and analog **110** was designed as a hydrazine analog. As a model 2-aminoxy thiazoline with similar functionality like in hydroxalog **108** was found unstable, further work on this analog was discontinued. Hydroxalog **109** was synthesized from (+)-L-arabinose, in which the hydroxylamine moiety was introduced by Mitsunobu reaction, and tri-substituted hydroxylamine intermediate was achieved by utilizing a classical reductive amination

condition in di-substituted hydroxylamine. Hydrazine analog **110** was synthesized from *sym*-dimethylhydrazine, and here too reductive amination condition was best applied to construct tetra-substituted backbone. Wipf's oxazoline-thiazoline interconversion strategy was found to be efficient for the thiazoline ring formation for both the analogs, **109** and **110**. For the comparative study of the biological activities, kalkitoxin was synthesized by partially following previously reported syntheses. The stretched-peptide chain of kalkitoxin was best achieved by following White's synthesis whereas, the thiazoline ring was constructed adapting Gerwick's synthesis.

Currently, anti-cancer assays of kalkitoxin and its analogs including oxazoline derivatives are under progress in the Valeriote laboratory at Henry Ford Hospital in Detroit. The hydroxalog 9-oxa-10-azakalkitoxin (IC_{50} = 2.4 nm) was found to be as active as kalktoxin (IC_{50} = 3.2 nm) against the liver cancer cell line, providing the evidence for the hydroxylamine analog concept. However, when the thiazoline ring of both kalkitoxin and hydroxalog were replaced with the oxazoline ring, a remarkable decrease in activity was observed, revealing the importance of the thiazoline ring for the activity.

CHAPTER 5. CONCLUSION

An efficient method for the synthesis of tri-substituted hydroxylamines was developed, adapting Rychnovsky's reductive etherification protocol. In this method *N,N*-dialkylhydroxylamines were first acylated to *O*-acylhydroxylamines, which were then reduced with DIBAL and the intermediate hemiacetals were trapped *in situ* with acetic anhydride as *O*-(α-acyloxyalkyl)hydroxylamines. In a subsequent Lewis acid-mediated deacetoxylation reaction, the *O*-(α-acyloxyalkyl)hydroxylamine was converted to several novel tri-substituted hydroxylamines by conducting the reaction in the presence of triethylsilane as a nucleophile. Various highly substituted tri-substituted hydroxylamine derivatives, with branching to the oxygen substituent, were also isolated when the triethylsilane was replaced with carbon nucleophiles.

In order to understand the relative energetics and contributions of N-inversion, N-O bond rotation, and ring inversion on the stereomutation process, variable temperature (VT) NMR studies were conducted on several 4-substituted *N*-hydroxypiperidines, *N*-alkoxypiperidines, and *N*-alkoxypyrrolidines. As the VT-NMR phenomena were found to be strongly dependent on the substituents at the 4-position, the ring flip process was concluded to be the rate determining step in the stereomutation process of such *N*-alkoxypiperidine systems.

Kalkitoxin, an anti-cancer agent, was chosen to test the concept of hydrazine and hydroxylamine analogs. Hydroxylamine analog 9-oxa-10-azakalkitoxin and hydrazine analog 7,8-diazakalkitoxin were designed by replacing one stereogenic center (C10) and two stereogenic centers (C7 and C8) of kalkitoxin, respectively with nitrogen. Syntheses of kalkitoxin and both analogs were executed successfully on a scale of several

milligrams. Currently, the anti-cancer assays of all these compounds are under progress in the Valeriote laboratory at the Henry Ford Hospital in Detroit. From the available IC_{50} values against hepato-carcinoma cells (HepG2), hydroxalog 9-oxa-10-azakalkitoxin retains bioactivity comparable to kalkitoxin, supporting the concept of hydroxylamines as surrogates of sp^3 carbon centers.

CHAPTER 6. EXPERIMENTAL SECTION

All reactions were conducted in oven dried glasswares under an argon atmosphere. All commercial solvents and reagents were used as purchased without further purification unless otherwise specified. All purifications were performed in flash column chromatography on silica gel (230-400 mesh) or neutral alumina (32-63 μm) as stationary phase unless otherwise noted. Reactions were analyzed by thin layer chromatography technique using pre-coated glass backed plates (w/UV 254), whcih were visualized by UV irradiation (254 nm) or by charring in cerium-ammonium-molybdate (CAM) or ninhydrin solution. 400 MHz, 500 MHz, or 600 MHz instruments were used to record ^{1}H and ^{13}C NMR spectra of all compounds. High-resolution mass spectra were recorded under electrospray conditions with a time of flight (TOF) mass analyzer. Melting points of compounds were determined by using electrothermal melting point apparatus whenever needed.

Experimental section for chapter 2

General procedure (A) for *O*-benzoyl hydroxylamines synthesis.

O-Benzoyl hydroxylamines were synthesized by adapting Ganem protocol.[112] To a stirred suspension of K_2HPO_4 (1.5 mmol), benzoyl peroxide (50% w/w blended with dicyclohexyl phthalate, 1.1 mmol), and dry DMF (2.5 mL) was added secondary amine (1 mmol) at room temperature. The stirring was continued until the completion of the reaction, and water (10 mL) was added followed by stirring vigorously for 1 h. The reaction mixture was extracted with EtOAc, and the resulting EtOAc extract was washed with saturated aq. $NaHCO_3$, water, and brine followed by drying over anhydrous Na_2SO_4 and

concentration. The so-obtained crude was purified by flash column chromatography on silica gel to give the desired *O*- benzoyl hydroxylamine.

O-Benzoyl-*N,N*-dibenzylhydroxylamine (62a).

Following the general procedure **A**, *N,N*-dibenzylamine (0.5 mL, 2.60 mmol) was converted to compound **62a**[162] (0.67 g, 81%) after stirring for 10 h, which was isolated as a white solid (eluent: 0 – 5% of acetone in hexane). M.p. 97 – 98 °C; lit[162] m.p. 96 – 98 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.86 – 7.80 (m, 2H), 7.50 (t, *J* = 7.3 Hz, 1H), 7.46 – 7.44 (m, 4H), 7.39 – 7.22 (m, 8H), 4.21 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.9, 135.9, 132.8, 129.4, 129.3, 128.3, 128.3, 127.6, 62.1.

O-Benzoyl-*N*-benzyl-*N*-methylhydroxylamine (62b).

Following the general procedure **A**, *N*-benzyl-*N*-methylamine (0.5 mL, 3.88 mmol) was converted to compound **62b** (0.77 g, 82%) after stirring for 10 h, which was isolated as a colorless oil (eluent: 0 – 5% of EtOAc in toluene). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.93 – 7.88 (m, 2H), 7.52 (t, *J* = 7.3 Hz, 1H), 7.45 – 7.36 (m, 4H), 7.34 – 7.22 (m, 3H), 4.16 (s, 2H), 2.93 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.9, 135.6, 132.9, 129.4, 129.3, 128.4, 128.3, 127.7, 65.1, 46.1; ESIHRMS calculated for $C_{15}H_{15}NO_2Na$ [M+Na]$^+$, 264.1000; found, 264.1006.

1-Piperidinyloxy benzoate (62c).

Following the general procedure **A**, piperidine (1.0 mL, 10.12 mmol) was converted to compound **62c**[162] (1.80 g, 86%) after stirring for 1.5 h, which was isolated as a white solid (eluent: 0 – 5% of EtOAc in toluene). M.p. 58 – 60 °C; lit[162] m.p. 55 – 59 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.02 – 7.96 (m, 2H), 7.53 (t, *J* = 7.3 Hz, 1H), 7.41 (t, *J* = 7.8 Hz, 2H),

3.48 (br s, 2H), 2.76 (d, J = 5.9 Hz, 2H), 1.86 – 1.76 (m, 4H), 1.64 (br s, 1H), 1.36 – 1.18 (m, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.7, 132.9, 129.6, 129.4, 128.3, 57.5, 25.0, 23.3.

4-Methylpiperidin-1-yl benzoate (62d).

Following the general procedure **A**, 4-methylpiperidine (2.5 mL, 20.57 mmol) was converted to compound **62d**[163] (3.71 g, 82%) after stirring for 1 h. It was isolate as a white solid (eluent: 0 – 10% of EtOAc in toluene). M.p. 91 – 92 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.99 (d, J = 7.3 Hz, 2H), 7.53 (t, J = 7.3 Hz, 1H), 7.41 (t, J = 7.6 Hz, 2H), 3.51 (d, J = 8.8 Hz, 2H), 2.72 (t, J = 10.5 Hz, 2H), 1.75 (d, J = 13.2 Hz, 2H), 1.65 – 1.52 (m, 2H), 1.52 – 1.40 (m, 1H), 0.93 (d, J = 5.9 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.8, 132.9, 129.6, 129.4, 128.3, 57.2, 33.5, 30.1, 21.2.

4-Chloropiperidin-1-oxyl Benzoate (62e).

Following the general procedure **A**, 4-chloropiperidine (1.79 g, 15.0 mmol) was converted to compound **62e** (2.92 g, 81%) after stirring for 1.5 h. It was isolated as a white solid (eluent: 0 – 10% of EtOAc in toluene). M.p. 63 – 65 °C; IR (neat, cm^{-1}) √ 1738; ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.00 (d, J = 6.6 Hz, 2H), 7.55 (t, J = 7.4 Hz, 1H), 7.43 (t, J = 7.7 Hz, 2H), 4.35 (br s, 0.6H), 4.07 (br s, 0.4H), 3.58 (br s, 0.8H), 3.34 (s, 2.4H), 2.97 (br s, 0.8H), 2.37 – 2.24 (m, 2H), 2.20 – 2.02 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.6, 133.1, 129.4, 129.2, 128.4, 55.1, 54.4, 52.8, 51.7, 33.0; ESIHRMS calculated for C$_{12}$H$_{14}$NO$_2$ClNa [M+Na]$^+$, 262.0611; found, 262.0613.

General procedure (B) for the synthesis of *N,N*-disubstituted hydroxylamines

To an ice-cold (<5 °C) solution of *O*-benzoyl hydroxylamine (1 mmol) in dry dichloromethane (6 mL) was added DIBAL (1 M solution in hexane, 2.5 mmol) carefully. The resultant mixture was stirred for 15 min in an ice-bath, and the reaction was quenched

by adding saturated aq. NH$_4$Cl (10 mL) and saturated aq. sodium potassium tartrate (8 mL). The mixture was stirred vigorously at room temperature for 1 h and was extracted with dichloromethane. The resulting organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. Purification was performed by flash column chromatography on silica gel to deliver the desired hydroxylamine.

N,N-Dibenzylhydroxylamine (63a).

The title compound **63a**[164] (60 mg, 90%) was obtained from compound **62a** (100 mg, 0.32 mmol) following the general procedure **B**. It was isolated as a white solid (eluent: 5 – 15% of EtOAc in hexane). M.p. 119 – 121 °C; lit[164] m.p. 118 – 119 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.37 – 7.24 (m, 10H), 6.90 (br s, 1H), 3.69 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.1, 129.8, 128.3, 127.5, 63.7.

N-Benzyl-N-methylhydroxylamine (63b).

The title compound **63b**[16] (265 mg, 93%) was obtained from compound **62b** (500 mg, 2.07 mmol) following the general procedure **B**. It was isolated as a colorless oil (eluent :10 – 30% of EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.36 – 7.24 (m, 5H), 3.70 (s, 2H), 2.51 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 136.9, 129.9, 128.3, 127.5, 66.4, 47.5.

Piperidin-1-ol (63c).

The title compound **63c**[165] (313 mg, 63%) was obtained from compound **62c** (1 g, 4.87 mmol) following the general procedure **B**. It was isolated as a colorless oil (eluent: ether). ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.09 (br s, 1H), 3.22 (d, J = 9.8 Hz, 2H), 2.41 (t, J = 11.0 Hz, 2H), 1.70 (d, J = 13.2 Hz, 2H), 1.60 – 1.42 (m, 3H), 1.18 – 1.02 (m, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 59.0, 25.4, 23.0.

4-Methylpiperidin-1-ol (63d).

The title compound **63d**[166] (443 mg, 84%) was obtained from compound **62d** (1 g, 4.56 mmol) following the general procedure **B**. It was isolated as a colorless oil (eluent: ether). The NMR data suggest a mixture of two stereo-isomers in 8.4:1 ratio. Major isomer; ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.60 (br s, 1H) 3.22 (d, J = 10.8 Hz, 2H), 2.44 (t, J = 11.7 Hz, 2H), 1.71 – 1.62 (m, 2H), 1.42 – 1.30 (m, 1H), 1.30 – 1.17 (m, 2H), 0.86 (d, J = 6.4 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 58.7, 34.0, 29.8, 21.3. The minor isomer was identified by ^{1}H NMR (400 MHz, CDCl$_3$) δ 3.00 – 2.83 (m, 4H), 1.61 – 1.46 (m, 4H), 0.94 (d, J = 6.4 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 54.6, 29.4, 20.0.

4-Chloropiperidin-1-ol (63e).

The title compound **63e** (1.29 g, 85%) was obtained from compound **62e** (2.68 g, 11.2 mmol) following the general procedure **B**. It was isolated as a mixture of two stereoisomers (**63e-1** and **63e-2**) in 1:1 ratio in the form of a white solid (eluent: diethyl ether). M.p. 97 – 99 °C; NMR of isomer **63e-1**: ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.07 (br s, 1H), 3.91 (s, 1H), 3.32 – 3.22 (m, 2H), 2.62 (t, J = 10.7 Hz, 2H), 2.24 (d, J = 12.8 Hz, 2H), 1.93 – 1.80 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 56.5, 55.5, 34.4; NMR of isomer **63e-2**: ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.07 (br s, 1H), 4.26 (s, 1H), 3.09 – 2.98 (m, 4H), 2.11 – 1.97 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 54.9, 53.0, 33.0; ESIHRMS calculated for C$_5$H$_{11}$NOCl [M+H]$^+$, 136.0529; found, 136.0534.

3-(1-Pyrrolidinyl)propanenitrile (64).

To a stirred solution of acrylonitrile (4.0 mL, 60.88 mmol) and Amberlyst-15 (1 g, 30% w/w) was added pyrrolidine (4.0 mL, 48.70 mmol) in an ice-bath. The ice-bath was removed, and the reaction mixture was stirred for 0.5 h. The mixture was then diluted with

dichloromethane and filtered, and the filtrate was concentrated to yield **64**[167] (5.57 g, 92%) as a light yellow oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 2.71 (t, *J* = 7.2 Hz, 2H), 2.53 – 2.44 (m, 6H), 1.79 – 1.68 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 118.9, 53.8, 51.2, 23.5, 17.6.

1-Pyrrolidinyloxy 4-phenylbutanoate (62f).

To a chilled solution of **64** (0.50 g, 4.03 mmol) in dry CH$_2$Cl$_2$ (40 mL) was added *m*-CPBA (0.95 g 77% *m*-CPBA, 6.05 mmol) and K$_2$CO$_3$ (1.11 g, 8.06 mmol) at −78 °C. After stirring for 3 h at −78 °C, the mixture was warmed slowly to ambient temperature, stirred for an additional 12 h, and filtered. 4-phenylbutyric acid (0.99 g, 6.05 mmol), DMAP (0.10 g, 0.81 mmol), and *N,N'*-dicyclohexylcarbodiimide (1.25 g, 6.05 mmol) were added to the resulting filtrate. The resultant mixture was stirred for the next 2 h at room temperature, filtered through a pad of Celite®, and was washed with saturated aq. NaHCO$_3$, brine, dried over anhydrous Na$_2$SO$_4$. The resultant solution was concentrated and purified by flash column chromatography on silica gel (10 – 30% of EtOAc in hexane) to afford **62f** (0.49 g, 52%) as a light red oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.23 (m, 2H), 7.22 – 7.13 (m, 3H), 3.14 (s, 4H), 2.64 (t, *J* = 7.5 Hz, 2H), 2.25 (t, *J* = 7.5 Hz, 2H), 2.00 – 1.90 (m, 2H), 1.85 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 172.0, 141.2, 128.5, 128.4, 128.3, 126.0, 57.4, 35.1, 32.4, 26.6, 22.0; ESIHRMS calculated for C$_{14}$H$_{19}$NO$_2$Na [M+Na]$^+$, 256.1313; found, 256.1319.

General procedure (C) for the synthesis of *O*-acyl hydroxylamines

A solution of *N,N'*-dicyclohexylcarbodiimide (1.25 – 2.5 mmol) in dry dichloromethane (1 mL) was added to a mixture of carboxylic acid (1.25 – 2.5 mmol), *N,N*-disubstituted hydroxylamine (1 mmol), DMAP (0.2 mmol), and dry dichloromethane (10 mL) at ambient temperature. The stirring was continued until completion of the

reaction, and the reaction mixture was filtered through a pad of Celite®. The resultant filtrate was washed with saturated aq. NaHCO$_3$ and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. After purification by flash column chromatography on silica gel, the desired *O*-acyl hydroxylamine was obtained.

N,N-Dibenzyl-O-(4-phenylbutanoyl)hydroxylamine (62g).

Following the general procedure **C**, the title compound **62g** was isolated (1.67 g, 98%) as a colorless oil by stirring **63a** (1 g, 4.69 mmol), *N,N'*-dicyclohexylcarbodiimide (1.22 g, 5.86 mmol), and 4-phenylbutyric acid (0.96 g, 5.86 mmol) for 40 mins. Eluent: 0 – 5% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.42 – 7.41 (m, 4H), 7.34 – 7.23 (m, 8H), 7.19 – 7.16 (m, 1H), 7.02 (d, *J* = 6.9 Hz, 2H), 4.07 (s, 4H), 2.37 (t, *J* = 7.6 Hz, 2H), 2.06 (t, *J* = 7.3 Hz, 2H), 1.75 – 1.62 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.8, 141.3, 136.1, 129.4, 128.4, 128.3, 128.3, 127.7, 125.8, 62.5, 34.7, 31.9, 26.2; ESIHRMS calculated for C$_{24}$H$_{25}$NO$_2$Na [M+Na]$^+$, 382.1783; found, 382.1779.

O-(1-Adamantanecarbonyl)-N,N-dibenzylhydroxylamine (62h).

Following the general procedure **C**, the title compound **62h** was isolated (1.27 g, 72%) as a white solid by stirring **63a** (1 g, 4.69 mmol), *N,N'*-dicyclohexylcarbodiimide (1.94 g, 9.38 mmol), and 1-adamantanecarboxylic acid (1.69 g, 9.38 mmol) for 28 h. Eluent: 0 – 5% of EtOAc in hexane. M.p. 110 – 111 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 (d, *J* = 7.3 Hz, 4H), 7.34 – 7.23 (m, 6H), 4.05 (s, 4H), 1.89 (s, 3H), 1.69 – 1.53 (m, 12H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 175.2, 136.1, 129.4, 128.1, 127.5, 62.1, 40.4, 38.5, 36.4, 27.8; ESIHRMS calculated for C$_{25}$H$_{29}$NO$_2$Na [M+Na]$^+$, 398.2096; found, 398.2103.

N,N-Dibenzyl-O-(3,3-dimethylbutanoyl)hydroxylamine (62i).

Following the general procedure **C**, the title compound **62i** was isolated (0.91 g, 63%) as a white solid by stirring **63a** (1 g, 4.69 mmol), 3,3-dimethylbutanoic acid (0.75 mL, 5.86 mmol), and N,N'-dicyclohexylcarbodiimide (1.22 g, 5.86 mmol) for 1.5 h. Eluent: 0 – 5% of EtOAc in hexane). M.p. 53 – 54 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.42 (d, J = 6.9 Hz, 4H), 7.34 – 7.22 (m, 6H), 4.05 (s, 4H), 1.95 (s, 2H), 0.77 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.2, 136.3, 129.3, 128.3, 127.5, 62.2, 46.2, 30.5, 29.3; ESIHRMS calculated for C$_{20}$H$_{25}$NO$_2$Na [M+Na]$^+$, 334.1783; found, 334.1782.

N-Benzyl-N-methyl-O-(5-phenylpentanoyl)hydroxylamine (62j).

Following the general procedure **C**, the title compound **62j** was isolated (396 mg, 87%) as a colorless oil by stirring **63b** (210 mg, 1.53 mmol), 5-phenylvaleric acid (341 mg, 1.91 mmol) and N,N'-dicyclohexylcarbodiimide (394 mg, 1.91 mmol) for 0.5 h. Eluent: 5 – 20% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.38 – 7.33 (m, 2H), 7.33 – 7.22 (m, 5H), 7.21 – 7.16 (m, 1H), 7.13 (d, J = 7.3 Hz, 2H), 3.99 (s, 2H), 2.81 (s, 3H), 2.54 (t, J = 7.0 Hz, 2H), 2.17 (t, J = 7.0 Hz, 2H), 1.58 – 1.45 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.8, 142.0, 135.9, 129.3, 128.4, 128.3, 128.3, 127.7, 125.8, 65.0, 46.2, 35.4, 32.7, 30.6, 24.5; ESIHRMS calculated for C$_{19}$H$_{23}$NO$_2$Na [M+Na]$^+$, 320.1626; found, 320.1625.

1-Piperidinyloxy 4-phenylbutanoate (62k).

Following the general procedure **C**, the title compound **62k** was isolated (387 mg, 93%) as a colorless oil by stirring **63c** (170 mg, 1.68 mmol), 4-phenylbutyric acid (355 mg, 2.10 mmol,) and N,N'-dicyclohexylcarbodiimide (433 mg, 2.10 mmol) for 0.5 h. Eluent: 5 – 20% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.24 (m, 2H),

7.21 – 7.14 (m, 3H), 3.35 (br s, 2H), 2.70 – 2.51 (m, 4H), 2.28 (t, *J* = 7.3 Hz, 2H), 2.01 – 1.90 (m, 2H), 1.82 – 1.69 (m, 4H), 1.61 (br s, 1H), 1.34 – 1.10 (m, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.5, 141.3, 128.5, 128.4, 126.0, 57.4, 35.1, 32.4, 26.7, 25.0, 23.2; ESIHRMS calculated for C$_{15}$H$_{21}$NO$_2$Na [M+Na]$^+$, 270.1470; found, 270.1465.

4-Methyl-1-piperidinyloxy 4-phenylbutanoate (62l).

Following the general procedure **C**, the title compound **62l** was isolated (740 mg, 91%) as a colorless oil by stirring **63d** (360 mg, 3.13 mmol), 4-phenylbutyric acid (642 mg, 3.91 mmol), and *N,N'*-dicyclohexylcarbodiimide (807 mg, 3.91 mmol) for 0.5 h. Eluent: 10 – 20% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.24 (m, 2H), 7.21 – 7.14 (m, 3H), 3.36 (d, *J* = 8.8 Hz, 2H), 2.65 (t, *J* = 7.6 Hz, 2H), 2.57 (t, *J* = 10.5 Hz, 2H), 2.27 (t, *J* = 7.6 Hz, 2H), 2.02 – 1.90 (m, 2H), 1.70 (d, *J* = 12.7 Hz, 2H), 1.57 – 1.34 (m, 3H), 0.91 (d, *J* = 5.9 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.6, 141.3, 128.5, 128.4, 126.0, 57.1, 35.1, 33.4, 32.4, 30.0, 26.7, 21.2; ESIHRMS calculated for C$_{16}$H$_{23}$NO$_2$Na [M+Na]$^+$, 284.1626; found, 284.1630.

4-Chloro-1-piperidinyloxy 4-Phenylbutanoate (62m).

Following the general procedure **C**, the title compound **62m** was isolated (827 mg, 80%) as a colorless oil by stirring **63e** (500 mg, 3.69 mmol), *N,N'*-dicyclohexylcarbodiimide (951 mg, 4.61 mmol), and 4-phenylbutanoic acid (757 mg, 4.61 mmol) for 1 h. Eluent: 10 – 30% of EtOAc in hexane. IR (neat, cm^{-1}) √ 1757; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.25 (m, 2H), 7.23 – 7.14 (m, 3H), 4.30 (s, 0.6H), 3.97 (br s, 0.4H), 3.42 (br s 0.8H), 3.18 (s, 2.4H), 2.77 (br s, 0.8H), 2.66 (t, *J* = 7.6 Hz, 2H), 2.35 – 2.18 (m, 4H), 2.11 – 1.91 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.4, 141.2, 128.5, 128.4, 126.1,

55.0, 51.5, 35.0, 32.9, 32.3, 26.6; ESIHRMS calculated for $C_{15}H_{20}NO_2ClNa$ [M+Na]$^+$, 304.1080; found, 304.1077.

N,N-Dibenzyl-O-(2-(2-naphthalenyloxy)acetyl)hydroxylamine (62n).

Following the general procedure **C**, the title compound **62n** was isolated (1.67 g, 89%) as a white solid by stirring **63a** (1 g, 4.69 mmol), 2-(naphthalen-2-yloxy)acetic acid (1.19 g, 5.86 mmol), and *N,N'*-dicyclohexylcarbodiimide (1.22 g, 5.86 mmol) for 0.5 h. Eluent: 10 – 45% of EtOAc in hexane. M.p. 87 – 89 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.76 (d, *J* = 7.8 Hz, 1H), 7.69 (d, *J* = 9.3 Hz, 1H), 7.60 (d, *J* = 8.3 Hz, 1H), 7.47 – 7.28 (m, 12H), 6.99 (dd, *J* = 8.8, 2.5 Hz, 1H), 6.66 (d, *J* = 2.5 Hz, 1H), 4.44 (s, 2H), 4.13 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 168.4, 155.6, 135.5, 134.2, 129.6, 129.5, 129.3, 128.6, 128.1, 127.6, 127.0, 126.3, 124.0, 118.2, 107.1, 64.6, 63.1; ESIHRMS calculated for $C_{26}H_{23}NO_3Na$ [M+Na]$^+$, 420.1576; found, 420.1575.

N,N-Dibenzyl-O-nicotinoylhydroxylamine (62o).

Following the general procedure **C**, the title compound **62o** was isolated (1.45 g, 97%) as a white solid by stirring **63a** (1 g, 4.69 mmol), nicotinic acid (0.72 g, 5.86 mmol), and *N,N'*-dicyclohexylcarbodiimide (1.22 g, 5.86 mmol) for 1 h. Eluent: 10 – 45% of EtOAc in hexane. M.p. 117 – 118 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.99 (d, *J* = 1.5 Hz, 1H), 8.70 (dd, *J* = 4.9, 2.0 Hz, 1H), 8.05 (td, *J* = 7.9, 2.0 Hz, 1H), 7.44 (d, *J* = 6.7 Hz, 4H), 7.34 – 7.22 (m, 7H), 4.22 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 163.6, 153.3, 150.3, 136.8, 135.7, 129.4, 128.4, 127.8, 125.3, 123.3, 62.4; ESIHRMS calculated for $C_{20}H_{18}N_2O_2Na$ [M+Na]$^+$, 341.1266; found, 341.1261.

General procedure (D) for the synthesis of *O*-(*α*-acetoxy) hydroxylamines.

A solution of *O*-acyl hydroxylamines (0.5 mmol) in dry dichloromethane (3 mL) was chilled at –78 °C. A 1 M DIBAL solution in hexane (0.63 – 1 mmol) was added dropwise followed by the sequential addition of pyridine (1.5 mmol), a solution of DMAP (1 mmol) in dry dichloromethane (1.5 mL), and Ac$_2$O (3 mmol). After stirring for 12 h (overnight) at –78 °C, the reaction mixture was warmed slowly to 0 °C followed by addition of saturated aq. NH$_4$Cl (5 mL) and saturated aq. sodium potassium tartrate (4 mL). The mixture was stirred vigorously at room temperature for 1 h and was extracted with dichloromethane. The organic extract was washed with brine and dried over anhydrous Na$_2$SO$_4$. The resultant solution was concentrated and purified by column chromatography on silica gel unless otherwise specified.

4-Phenyl-1-(1-pyrrolidinyloxy)butyl acetate (65f).

The title compound **65f** (75 mg, 63%) was isolated as a colorless oil from **62f** (100 mg, 0.43 mmol) and DIBAL (0.64 mL, 1 M in hexane, 0.64 mmol) by following the general procedure **D**. Eluent: 10 – 20% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.24 (m, 2H), 7.21 – 7.14 (m, 3H), 6.13 – 6.08 (m, 1H), 3.00 (s, 4H), 2.67 – 2.59 (m, 2H), 2.09 (s, 3H), 1.76 (s, 4H), 1.71 – 1.64 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.4, 142.0, 128.4, 128.3, 125.8, 100.2, 57.4, 35.5, 32.5, 26.1, 21.9, 21.5; ESIHRMS calculated for C$_{16}$H$_{23}$NO$_3$Na [M+Na]$^+$, 300.1576; found, 300.1582.

O-(1-Acetoxy-4-phenylbutyl-*N,N*-dibenzylhydroxylamine (65g).

The title compound **65g** (102 mg, 88%) was isolated as a colorless oil from **62g** (100 mg, 0.28 mmol) and DIBAL (0.56 mL, 1 M in hexane, 0.56 mmol) by following general procedure **D**. Eluent: 0 – 5% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 –

7.21 (m, 12H), 7.19 – 7.16 (m, 1H), 7.07 (d, J = 7.3 Hz, 2H), 5.88 (t, J = 5.4 Hz, 1H), 4.01 (br s, 2H), 3.78 (d, J = 12.7 Hz, 2H), 2.41 (t, J = 7.3 Hz, 2H), 1.85 (s, 3H), 1.51 – 1.22 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.2, 141.9, 137.1, 129.5, 128.4, 128.3, 128.2, 127.4, 125.7, 100.4, 62.9, 35.3, 32.4, 25.4, 21.2; ESIHRMS calculated for C$_{26}$H$_{29}$NO$_3$Na [M+Na]$^+$, 426.2045; found, 426.2048.

O-(1-Acetoxy-1-adamantanylmethyl)-N,N-dibenzylhydroxylamine (65h).

The title compound **65h** (217 mg, 65%) was isolated as a colorless oil from **62h** (300 mg, 0.80 mmol) and DIBAL (1 mL, 1 M in hexane, 1 mmol) by following the general procedure **D**. Eluent: 0 – 8% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.36 – 7.28 (m, 8H), 7.28 – 7.22 (m, 2H), 5.58 (s, 1H), 3.91 (d, J = 13.2 Hz, 2H), 3.82 (d, J = 13.2 Hz, 2H), 1.89 (s, 3H), 1.82 (s, 3H), 1.65 (d, J = 11.7 Hz, 3H), 1.58 (d, J = 11.7 Hz, 3H), 1.49 (d, J = 12.2 Hz, 3H), 1.33 (d, J = 12.2 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.6, 137.2, 129.6, 128.1, 127.2, 103.1, 62.2, 37.0, 36.7, 36.5, 27.9, 21.1; ESIHRMS calculated for C$_{27}$H$_{33}$NO$_3$Na [M+Na]$^+$, 442.2358; found, 442.2367.

O-(1-Acetoxy-3,3-dimethylbutyl)-N,N-dibenzylhydroxylamine (65i).

The title compound **65i** (85 mg, 75%) was isolated as a colorless oil from **62i** (100 mg, 0.32 mmol) and DIBAL (0.48 mL, 1 M in hexane, 0.48 mmol) by following the general procedure **D**. Eluent: 0 – 5% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.39 – 7.29 (m, 8H), 7.28 – 7.22 (m, 2H), 6.00 (t, J = 5.4 Hz, 1H), 4.00 (br s, 2H), 3.77 (d, J = 13.2 Hz, 2H), 1.88 (s, 3H), 1.40 (dd, J = 14.2, 5.9 Hz, 1H), 1.34 (dd, J = 14.2, 4.9 Hz, 1H), 0.68 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.1, 137.3, 129.4, 128.3, 127.3, 98.5, 62.8, 46.1, 29.6, 29.1, 21.5; ESIHRMS calculated for C$_{22}$H$_{29}$NO$_3$Na [M+Na]$^+$, 378.2045; found, 378.2046.

O-(1-Acetoxy-5-phenylpentyl)-N-benzyl-N-methylhydroxylamine (65j).

The title compound **65j** (51 mg, 89%) was isolated as a colorless oil from **62j** (50 mg, 0.17 mmol) and DIBAL (0.30 mL, 1 M in hexane, 0.30 mmol) by following the general procedure **D**. Eluent: 5 – 10% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.35 – 7.23 (m, 7H), 7.20 – 7.11 (m, 3H), 5.91 (br s, 1H), 3.88 (br s, 1H), 3.76 (d, J = 12.7 Hz, 1H), 2.64 (s, 3H), 2.53 (t, J = 7.6 Hz, 2H), 1.99 (s, 3H), 1.52 (s, 4H), 1.22 (br s, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.3, 142.4, 136.9, 129.6, 128.4, 128.2, 128.2, 127.4, 125.6, 100.3, 65.4, 46.5, 35.7, 32.7, 31.0, 23.6, 21.3; ESIHRMS calculated for C$_{21}$H$_{27}$NO$_3$Na [M+Na]$^+$, 364.1889; found, 364.1885.

4-Phenyl-1-(1-piperidinyloxy)butyl acetate (65k).

The title compound **65k** (64 mg, 55%) was isolated as a colorless oil from **62k** (100 mg, 0.40 mmol) and DIBAL (0.6 mL, 1 M in hexane, 0.60 mmol) by following the general procedure **D**. Flash column chromatography on neutral alumina (0 – 8% of EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.25 (m, 2H), 7.21 – 7.15 (m, 3H), 6.07 (t, J = 4.6 Hz, 1H), 3.24 (br s, 2H), 2.63 (t, J = 6.9 Hz, 2H), 2.54 – 2.36 (m, 2H), 2.06 (s, 3H), 1.79 – 1.62 (m, 6H), 1.55 (br s, 3H), 1.14 (br s, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.3, 142.0, 128.4, 128.3, 125.8, 99.8, 58.2, 57.0, 35.5, 32.6, 26.1, 25.3, 23.4, 21.4; ESIHRMS calculated for C$_{17}$H$_{25}$NO$_3$Na [M+Na]$^+$, 314.1732; found, 314.1739.

1-(4-Methyl-1-piperidinyloxy)-4-phenylbutyl acetate (65l).

The title compound **65l** (98 mg, 84%) was isolated as a colorless oil from **62l** (100 mg, 0.38 mmol) and DIBAL (0.57 mL, 1 M in hexane, 0.57 mmol) by following the general procedure **D**. Eluent: 5 – 10% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.25 (m, 2H), 7.21 – 7.14 (m, 3H), 6.06 (t, J = 4.8 Hz, 1H), 3.32 – 3.17 (m, 2H), 2.63 (t, J

= 6.9 Hz, 2H), 2.53 – 2.36 (m, 2H), 2.06 (s, 3H), 1.77 – 1.58 (m, 6H), 1.40 – 1.19 (m, 3H), 0.88 (d, J = 5.4 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.4, 142.0, 128.4, 128.3, 125.8, 99.9, 57.8, 56.7, 35.5, 34.0, 33.9, 32.6, 30.1, 26.1, 21.4, 21.3; ESIHRMS calculated for C$_{18}$H$_{27}$NO$_3$Na [M+Na]$^+$, 328.1889; found, 328.1888.

1-(4-Chloro-1-piperidinyloxy)-4-phenylbutyl acetate (65m).

The title compound **65m** (98 mg, 85%) was isolated as a colorless oil from **62m** (100 mg, 0.35 mmol) and DIBAL (0.62 mL 1 M in hexane, 0.62 mmol) by following the general procedure **D**. Eluent: 0 – 20% of EtOAc in hexane. IR (neat, cm^{-1}) √ 1738; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.25 (m, 2H), 7.23 – 7.13 (m, 3H), 6.05 (s, 1H), 4.21 (br s, 0.5H), 3.92 (s, 0.5H), 3.28 (br s, 1H), 3.05 (br s, 2H), 2.63 (s, 3H), 2.26 – 1.78 (m, 7H), 1.69 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.3, 141.9, 128.4, 128.4, 125.9, 99.6, 55.9, 55.5, 52.4, 51.3, 35.5, 34.1, 33.0, 32.5, 26.0, 21.4; ESIHRMS calculated for C$_{17}$H$_{24}$NO$_3$ClNa [M+Na]$^+$, 348.1342; found, 348.1346.

***O*-(1-Acetoxy-2-(naphthalen-2-yloxy)ethyl)-*N,N*-dibenzylhydroxylamine (65n).**

The title compound **65n** (100 mg, 90%) was isolated as a colorless oil from **62n** (100 mg, 0.25 mmol) and DIBAL (0.38 mL, 1 M in hexane, 0.38 mmol) by following the general procedure **D**. Eluent: 0 – 10% of EtOAc in hexane. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.76 (d, J = 8.3 Hz, 1H), 7.73 – 7.67 (m, 2H), 7.44 (t, J = 7.1 Hz, 1H), 7.41 – 7.30 (m, 9H), 7.30 – 7.23 (m, 2H), 7.04 (dd, J = 8.8, 2.5 Hz, 1H), 6.95 (d, J = 2.0 Hz, 1H), 6.26 (t, J = 5.4 Hz, 1H), 4.04 (d, J = 13.2 Hz, 2H), 3.97 – 3.87 (m, 3H), 3.84 (dd, J = 10.3, 5.4 Hz, 1H) 1.86 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 169.9, 156.1, 136.9, 134.4, 129.7, 129.3, 129.1, 128.3, 127.6, 127.5, 126.8, 126.3, 123.8, 118.7, 106.9, 96.7, 66.6, 62.8, 21.0; ESIHRMS calculated for C$_{28}$H$_{27}$NO$_4$Na [M+Na]$^+$, 464.1838; found, 464.1826.

General procedure (E) for the direct synthesis of *N,N,O*-trisubstituted hydroxylamines from *O*-acyl hydroxylamines.

A 1 M DIBAL solution in hexane (1 mmol) was added dropwise to a chilled solution of *O*-acyl hydroxylamine (0.5 mmol) in dry dichloromethane (3 mL) at −78 ºC. To this solution were added sequentially, dropwise, pyridine (1.5 mmol), a solution of DMAP (1 mmol) in dry CH_2Cl_2 (1.5 mL), and Ac_2O (3 mmol). The reaction mixture was stirred for 12 h at −78 ºC, warmed slowly to 0 ºC, and added saturated aq. NH_4Cl (5 mL) and saturated aq. sodium potassium tartrate (4 mL). The resultant mixture was vigorously stirred at room temperature for 1 h. Then the mixture was extracted with dichloromethane, and the resultant organic extract was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The so-obtained intermediate residue was dissolved in dry dichloromethane (10 mL) and cooled to −78 ºC. Et_3SiH (1.25 mmol) and $BF_3 \cdot OEt_2$ (1.25 mmol) were added one after the other, and the reaction mixture was warmed slowly to 0 ºC followed by the addition of saturated aq. $NaHCO_3$. The mixture was then extracted with pentane, and the resulting organic extract was dried over anhydrous Na_2SO_4. Subsequent concentration followed by the purification of the resultant residue by flash column chromatography on silica gel (0 – 5% of EtOAc in hexane) gave the desired hydroxylamines.

N,N,O-Tribenzylhydroxylamine (66a).

According to general procedure **E**, compound **66a**[168] (143 mg, 50%) was isolated as a colorless oil from **62a** (300 mg, 0.95 mmol). ^{1}H NMR (400 MHz, CDCl3) δ 7.43 – 7.42 (m, 4H), 7.39 – 7.27 (m, 6H), 7.25 – 7.20 (m, 3H), 6.98 – 6.96 (m, 2H), 4.19 (s, 2H), 3.90

(s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.8, 137.0, 129.8, 129.1, 128.1, 127.7, 127.3, 76.1, 62.9.

N,O-Dibenzyl-N-methylhydroxylamine (66b).

According to general procedure **E**, compound **66b** (34 mg, 53%) was isolated as a colorless oil from **62b** (68 mg, 0.28 mmol). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 – 7.24 (m, 8H), 7.24 – 7.19 (m, 2H), 4.51 (s, 2H), 3.83 (s, 2H), 2.65 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.5, 129.7, 128.8, 128.2, 128.2, 127.7, 127.3, 74.9, 65.2, 45.6; ESIHRMS calculated for C$_{15}$H$_{18}$NO [M+H]$^+$, 228.1388; found, 228.1385.

General procedure (F) for the synthesis of N,N,O-trisubstituted hydroxylamines from O-(α-acetoxy)hydroxylamines.

To a chilled solution of O-(α-acetoxy)hydroxylamine (0.25 mmol) in dry dichloromethane (5 mL) were added Et$_3$SiH (0.63 mmol) and BF$_3$·OEt$_2$ (0.63 mmol) dropwise at –78 °C. The reaction mixture was warmed slowly to 0 °C or room temperature in 2.5 h and stirring was continued until completion. The reaction was quenched by saturated aq. NaHCO$_3$ and was extracted with pentane. The so-obtained organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by flash column chromatography on silica gel to afford the trisubstituted hydroxylamines.

1-(4-Phenylbutoxy)pyrrolidine (66f).

Compound **66f** (30 mg, 77%) was prepared according to general procedure **F** using **65f** (50 mg, 0.18 mmol) with warming to room temperature and stirring for an additional 3 h. It was isolated as a colorless oil (0 – 10% EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.24 (m, 2H), 7.21 – 7.15 (m, 3H), 3.71 (t, J = 6.4 Hz, 2H), 3.01 – 2.92 (m, 4H), 2.63 (t, J = 7.6 Hz, 2H), 1.77 (br s, 4H), 1.71 – 1.55 (m, 4H); ^{13}C NMR (100

MHz, CDCl$_3$) δ 142.5, 128.4, 128.2, 125.7, 72.1, 56.6, 35.8, 28.6, 28.1, 21.9; ESIHRMS calculated for C$_{14}$H$_{22}$NO [M+H]$^+$, 220.1701; found, 220.1702.

N,N-Dibenzyl-*O*-(4-phenylbutyl)hydroxylamine (66g).

Compound **66g** (33 mg, 83%) was prepared according to general procedure **F** using **65g** (47 mg, 0.12 mmol) with warming to 0 °C. It was isolated as a colorless oil (0 – 5% EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 (d, *J* = 6.9 Hz, 4H), 7.34 – 7.24 (m, 8H), 7.22 – 7.14 (m, 1H), 7.09 (d, *J* = 6.9 Hz, 2H), 3.86 (s, 4H), 3.34 (t, *J* = 6.4 Hz, 2H), 2.42 (t, *J* = 7.6 Hz, 2H), 1.44 – 1.37 (m, 2H), 1.34 – 1.27 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 142.5, 137.9, 129.6, 128.4, 128.1, 128.1, 127.2, 125.5, 72.9, 62.6, 35.5, 28.1, 27.8; ESIHRMS calculated for C$_{24}$H$_{28}$NO [M+H]$^+$, 346.2171; found, 346.2177.

O-(1-Adamantanylmethyl)-*N,N*-dibenzylhydroxylamine (66h).

Compound **66h** (39 mg, 89%) was prepared according to general procedure **F** using **65h** (50 mg, 0.12 mmol) with warming 0 °C. It was isolated as a white solid (0 – 5% EtOAc in hexane). M.p. 48 – 49 °C; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.37 (d, *J* = 6.9 Hz, 4H), 7.34 – 7.22 (m, 6H), 3.83 (s, 4H), 2.91 (s, 2H), 1.85 (s, 3H), 1.64 (d, *J* = 12.2 Hz, 3H), 1.56 (d, *J* = 11.3 Hz, 3H), 1.30 (d, *J* = 2.0 Hz, 6H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.9, 129.8, 128.0, 127.0, 83.5, 62.5, 39.6, 37.2, 33.4, 28.2; ESIHRMS calculated for C$_{25}$H$_{32}$NO [M+H]$^+$, 362.2484; found, 362.2498.

N,N-Dibenzyl-*O*-(3,3-dimethylbutyl)hydroxylamine (66i).

Compound **66i** (50 mg, 81%) was prepared according to general procedure **F** using **65i** (75 mg, 0.21 mmol) with warming to 0 °C and stirring for an additional 0.5 h. It was isolated as a colorless oil (0 – 5% EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 (d, *J* = 7.3 Hz, 4H), 7.36 – 7.23 (m, 6H), 3.85 (s, 4H), 3.32 (t, *J* = 7.6 Hz, 2H), 1.17

(t, J = 7.6 Hz, 2H), 0.69 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.9, 129.7, 128.1, 127.1, 70.5, 62.7, 41.5, 29.5, 29.3; ESIHRMS calculated for C$_{20}$H$_{28}$NO [M+H]$^+$, 298.2171; found, 298.2175.

_N_-Benzyl-_N_-methyl-_O_-(5-phenylpentyl)hydroxylamine (66j).

Compound **66j** (35 mg, 85%) was prepared according to general procedure **F** using **65j** (50 mg, 0.15 mmol) with warming to 0 °C and stirring for an additional 45 min at 0 °C. It was isolated as a colorless oil (0 – 5% EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.39 – 7.25 (m, 7H), 7.21 – 7.14 (m, 3H), 3.79 (s, 2H), 3.52 (t, J = 6.4 Hz, 2H), 2.62 (s, 3H), 2.57 (t, J = 7.8 Hz, 2H), 1.60 – 1.51 (m, 2H), 1.51 – 1.42 (m, 2H), 1.32 – 1.21 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 142.7, 137.6, 129.7, 128.4, 128.2, 128.1, 127.2, 125.6, 72.1, 65.1, 45.5, 35.9, 31.3, 28.6, 25.8; ESIHRMS calculated for C$_{19}$H$_{26}$NO [M+H]$^+$, 284.2014; found, 284.2016.

1-(4-Phenylbutoxy)piperidine (66k).

Compound **66k** (37 mg, 75%) was prepared according to general procedure **F** using **65k** (61 mg, 0.21 mmol) with warming to 0 °C and stirring for an additional 2 h. It was isolated as a colorless oil (0 – 10% EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.31 – 7.24 (m, 2H), 7.21 – 7.13 (m, 3H), 3.71 (t, J = 6.4 Hz, 2H), 3.26 (br s, 2H), 2.63 (t, J = 7.6 Hz, 2H), 2.34 (br s, 2H), 1.80 – 1.45 (m, 9H), 1.14 (br s, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 142.5, 128.4, 128.2, 125.6, 71.2, 56.9, 35.8, 28.6, 28.2, 25.5, 23.5; ESIHRMS calculated for C$_{15}$H$_{24}$NO [M+H]$^+$, 234.1858; found, 234.1863.

4-Methyl-1-(4-phenylbutoxy)piperidine (66l).

Compound **66l** (60 mg, 76%) was prepared according to general procedure **F** using **65l** (98 mg, 0.32 mmol) with warming to 0 °C and stirring for an additional 2 h. It

was isolated as a colorless oil (0 – 5% EtOAc in hexane). ^{1}H NMR (500 MHz, toluene-d_8, T = 363 K) δ 7.14 – 6.95 (m, 5H), 3.66 (t, J = 6.2 Hz, 2H), 3.17 (d, J = 10.4 Hz, 2H), 2.50 (t, J = 7.5 Hz, 2H), 2.39 (t, J = 9.6 Hz, 2H), 1.67 – 1.59 (m, 2H), 1.59 – 1.52 (m, 2H), 1.43 (d, J = 13.1 Hz, 2H), 1.30 – 1.13 (m, 3H), 0.76 (d, J = 6.1 Hz, 3H); ^{13}C NMR (125 MHz, toluene-d_8) δ 143.2, 129.1, 128.9, 126.3, 71.7, 57.3, 36.6, 34.9, 31.1, 29.5, 29.1, 22.0; ESIHRMS calculated for $C_{16}H_{26}NO$ [M+H]$^+$, 248.2014; found, 248.2009.

4-Chloro-1-(4-phenylbutoxy)piperidine (66m).

Compound **66m** (52 mg, 74%) was prepared according to general procedure **F** using **65m** (85 mg, 0.26 mmol) with warming to room temperature and stirring for an additional 2 h. It was isolated as a colorless oil (eluent: 0 – 10% of EtOAc in hexane). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.25 (m, 2H), 7.22 – 7.15 (m, 3H), 4.25 (br s, 0.45H), 3.89 (br s, 0.55H), 3.69 (t, J = 6.3 Hz, 2H), 3.26 (br s, 1H), 3.01 (br s, 1.8H), 2.63 (t, J = 7.5 Hz, 2H), 2.51 (br s, 1H), 2.27 – 1.80 (m, 4H), 1.76 – 1.54 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 142.4, 128.4, 128.3, 125.7, 71.5, 56.1, 55.8, 54.4, 51.0, 35.8, 34.7, 33.1, 28.5, 28.1; ESIHRMS calculated for $C_{15}H_{23}NOCl$ [M+H]$^+$, 268.1468; found, 268.1470.

***N,N*-Dibenzyl-*O*-(2-(2-naphthalenyloxy)ethyl)hydroxylamine (66n).**

To an ice-cold solution of **65n** (160 mg, 0.36 mmol) in dry dichloromethane (7.2 mL) were added Et$_3$SiH (290 μL, 1.80 mmol) and BF$_3$·OEt$_2$ (230 μL, 1.80 mmol). The ice-bath was removed and the reaction mixture was stirred for 7 days. After quenching the reaction with saturated aq. NaHCO$_3$, the mixture was extracted with pentane. The resulting organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by flash column chromatography on silica gel (0 – 5% EtOAc in hexane) to give **66n** (52 mg, 38%) as a light yellow oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.75 (d, J = 8.3 Hz, 1H), 7.71

– 7.65 (m, 2H), 7.46 – 7.38 (m, 5H), 7.36 – 7.28 (m, 5H), 7.28 – 7.22 (m, 2H), 7.06 (dd, J = 8.8, 2.5 Hz, 1H), 6.87 (d, J = 2.5 Hz, 1H), 3.93 (s, 4H), 3.78 (t, J = 4.9 Hz, 2H), 3.71 (t, J = 4.9 Hz, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 156.6, 137.4, 134.5, 129.8, 129.1, 128.9, 128.2, 127.6, 127.4, 126.6, 126.2, 123.5, 119.0, 106.5, 71.3, 65.8, 62.7; ESIHRMS calculated for C$_{26}$H$_{25}$NO$_2$Na [M+Na]$^+$, 406.1783; found, 406.1778.

N,N-Dibenzyl-O-(pyridin-3-ylmethyl)hydroxylamine (66o).

A 1M solution of DIBAL in hexane (0.63 mL, 0.63 mmol) was added dropwise to a stirred solution of **62o** (100 mg, 0.31 mmol) in dry dichloromethane (1.9 mL) at –78 °C. Subsequently, pyridine (76 µL, 0.94 mmol), a solution of DMAP (77 mg, 0.63 mmol) in dry dichloromethane (0.9 mL), and Ac$_2$O (178 µL, 1.88 mmol) were added dropwise and sequentially. The reaction mixture was then allowed to stir for 12 h at –78 °C, and it was warmed slowly to 0 °C in 2.5 h followed by the additions of saturated aq. NH$_4$Cl (3.2 mL) and saturated aq. sodium potassium tartrate (2.4 mL). The resultant mixture was vigorously stirred at room temperature for 1 h before the extraction with dichloromethane. The resulting organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The so-obtained crude residue **65o** was dissolved in dry dichloromethane (6.3 mL) and again cooled to –78 °C. Et$_3$SiH (252 µL, 1.58 mmol) and BF$_3$·OEt$_2$ (200 µL, 1.58 mmol) were added dropwise and sequentially to this chilled solution, and the resulting solution was warmed to room temperature and stirring was continued for an additional 36 h. After quenching with saturated aqueous NaHCO$_3$, the reaction mixture was extracted with pentane. The organic extract was then dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by column chromatography on neutral alumina (5 – 20% of EtOAc in hexane) to deliver product **66o** (40 mg, 42%) as a colorless oil. ^{1}H NMR (400

MHz, CDCl$_3$) δ 8.45 (s, 1H), 8.16 (s, 1H), 7.41 – 7.24 (m, 10H), 7.20 – 7.15 (m, 1H), 7.13 – 7.07 (m, 1H), 4.12 (s, 2H), 3.88 (s, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 150.3, 149.0, 137.5, 136.7, 132.4, 129.8, 128.2, 127.4, 123.2, 73.3, 63.1; ESIHRMS calculated for C$_{20}$H$_{21}$N$_2$O [M+H]$^+$, 305.1654; found, 305.1657.

N,N-Dibenzyl-O-(2-azidoacetyl)hydroxylamine (62p).

Following the general procedure **C**, the title compound **62p** was isolated (1.38 g, 65%) as a yellow oil by stirring **63a** (1.53 g, 7.17 mmol), 2-azidoacetic acid **67** (1.45 g, 14.35 mmol), and N,N'-dicyclohexylcarbodiimide (2.97 g, 14.35 mmol) for 2 h. Eluent: 0 – 5% of EtOAc in hexane. IR (neat, cm^{-1}) √ 2108, 1765; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.47 – 7.22 (m, 10H), 4.10 (s, 4H), 3.47 (s, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 167.6, 135.3, 129.5, 128.5, 128.1, 63.0, 49.5; ESIHRMS calculated for C$_{16}$H$_{16}$N$_4$O$_2$Na [M+Na]$^+$, 319.1171; found 319.1169.

N,N-Dibenzyl-O-(3-azidopropionyl)hydroxylamine (62q).

Following the general procedure **C**, the title compound **62q** was isolated (1.30 g, 52%) as a yellow oil by stirring **63a** (1.74 g, 8.15 mmol), 3-azidopropionic acid **68** (1.41 g, 12.23 mmol), and N,N'-dicyclohexylcarbodiimide (2.52 g, 12.23 mmol) for 2 h. Eluent: 0 – 10% EtOAc in hexane. IR (neat, cm^{-1}) √ 2098, 1756; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 (d, J = 6.8 Hz, 4H), 7.37 – 7.25 (m, 6H), 4.08 (s, 4H), 3.26 (t, J = 6.7 Hz, 2H), 2.26 (t, J = 6.7 Hz, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 169.6, 135.7, 129.4, 128.4, 127.8, 62.6, 46.4, 32.4; ESIHRMS calculated for C$_{17}$H$_{19}$N$_4$O$_2$ [M+H]$^+$, 311.1508; found 311.1518.

N,N-Dibenzyl-O-(4-azidobutanoyl)hydroxylamine (62r).

Following the general procedure C, the title compound **62r** was isolated (813 mg, 72%) as a colorless oil by stirring **63a** (687 mg, 3.22 mmol), 4-azidobutyric acid **69** (624

mg, 4.83 mmol), and *N*,*N'*-dicyclohexylcarbodiimide (996 mg, 4.83 mmol) for 2 h. Eluent: 0 – 5% EtOAc in hexane. IR (neat, cm^{-1}) √ 2096, 1756; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.43 – 7.37 (m, 4H), 7.30 (m, 6H), 4.07 (s, 4H), 2.93 (t, *J* = 6.8 Hz, 2H), 2.09 (t, *J* = 7.1 Hz, 2H), 1.57 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 171.2, 135.9, 129.5, 128.3, 127.7, 62.7, 50.0, 29.5, 24.1; ESIHRMS calculated for C$_{18}$H$_{21}$N$_4$O$_2$ [M+H]$^+$, 325.1665; found 325.1673.

N,N-Dibenzyl-O-(1-acetoxy-2-azidoethyl)hydroxylamine (65p).

The title compound **65p** (298 mg, 86%) was isolated as a colorless oil from **62p** (300 mg, 1.01 mmol) and DIBAL (1.77 mL, 1 M in hexane, 1.77 mmol) by following the general procedure **D**. Eluent: 0 – 5% EtOAc in hexane. IR (neat, cm^{-1}) √ 2102, 1751; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.44 – 7.22 (m, 10H), 5.89 (ap t, *J* = 4.9 Hz, 1H), 4.01 (br d, *J* = 13.1 Hz, 2H), 3.87 (d, *J* = 13.1 Hz, 2H), 2.96 (m, 2H), 1.83 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 169.7, 136.8, 129.6, 128.4, 127.6, 97.4, 63.1, 51.2, 21.0; ESIHRMS calculated for C$_{18}$H$_{20}$N$_4$O$_3$Na [M+Na]$^+$, 363.1433; found 363.1434.

N,N-Dibenzyl-O-(1-acetoxy-3-azidopropyl)hydroxylamine (65q).

The title compound **65q** (210 mg, 61%) was isolated as a colorless oil from **62q** (300 mg, 0.97 mmol) and DIBAL (1.70 mL, 1 M in hexane, 1.70 mmol) by following the general procedure **D**. Eluent: 0 – 5% EtOAc in hexane. IR (neat, cm^{-1}) √ 2098, 1743; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.44 – 7.12 (m, 10H), 5.87 (ap t, *J* = 5.4 Hz, 1H), 4.04 (br m, 2H), 3.79 (d, *J* = 13.0 Hz, 2H), 3.00 (dt, *J*= 12.5, 7.2 Hz 1H), 2.89 (dt, *J* = 12.5, 7.2 Hz, 1H), 1.87 (s, 3H), 1.72 – 1.53 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 169.9, 136.9, 129.5, 128.4, 127.5, 98.1, 62.9, 46.1, 32.3, 21.1; ESIHRMS calculated for C$_{19}$H$_{22}$N$_4$O$_3$Na [M+Na]$^+$, 377.1590; found 377.1603.

N,N-Dibenzyl-O-(1-acetoxy-4-azidobutyl)hydroxylamine (65r).

The title compound **65r** (693 mg, 87%) was isolated as a colorless oil from **62r** (700 mg, 2.16 mmol) and DIBAL (3.78 mL, 1 M in hexane, 3.78 mmol) by following the general procedure **D**. Eluent: 0 – 5% EtOAc in hexane. IR (neat, cm^{-1}) √ 2097, 1741; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.44 – 7.16 (m, 10H), 5.80 (dd, J= 6.0, 5.3 Hz, 1H), 4.05 (br m, 2H), 3.77 (d, J = 13.6 Hz, 2H), 3.00 (t, J = 6.6 Hz, 2H), 1.88 (s, 3H), 1.49 – 1.41 (m, 2H), 1.37 – 1.16 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.2, 137.0, 129.4, 128.3, 127.5, 100.0, 63.0, 50.7, 29.9, 23.2, 21.2; ESIHRMS calculated for C$_{20}$H$_{24}$N$_4$O$_3$Na [M+Na]$^+$, 391.1746; found 391.1748.

N,N-Dibenzyl-O-(2-azidoethyl)hydroxylamine (66p).

To an ice-cold solution of **65p** (50 mg, 0.15 mmol) in dry dichloromethane (3 mL) were added Et$_3$SiH (118 μL, 0.74 mmol) and BF$_3$·OEt$_2$ (90 μL, 0.74 mmol). The ice-bath was removed and the reaction mixture was allowed to stir for 4 days. After quenching with saturated aq. NaHCO$_3$, the resultant mixture was extracted with pentane, and the so-obtained organic extract was dried over anhydrous Na$_2$SO$_4$. Concentration followed by the purification of the residue by flash column chromatography on silica gel afforded **66p** (7 mg, 17%) as a yellow oil (eluent: 0 – 5% EtOAc in hexane). IR (neat, cm^{-1}) √ 2102; ^{1}H NMR (600 MHz, CDCl$_3$) δ 7.43 – 7.21 (m, 10H), 3.88 (s, 4H), 3.38 (t, J = 5.2 Hz, 2H), 2.96 (t, J = 5.2 Hz, 2H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 137.5, 129.7, 128.2, 127.4, 71.6, 62.8, 49.7; ESIHRMS calculated for C$_{16}$H$_{19}$N$_4$O [M+H]$^+$, 283.1559; found 283.1567.

N,N-Dibenzyl-O-(3-azidopropyl)hydroxylamine (66q).

Compound **66q** (20 mg, 48%) was prepared according to general procedure **F** using **65q** (50 mg, 0.14 mmol) with warming to room temperature and stirring for an

additional 24 h. It was isolated as a yellow oil (eluent: 0 – 5% EtOAc in hexane). IR (neat, cm^{-1}) √ 2096; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.45 – 7.20 (m, 10H), 3.85 (s, 4H), 3.34 (t, J = 5.9 Hz, 2H), 2.91 (t, J = 7.0 Hz, 2H), 1.51 – 1.41 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.7, 129.6, 128.2, 127.3, 69.7, 62.7, 48.4, 28.0; ESIHRMS calculated for C$_{17}$H$_{21}$N$_4$O [M+H]$^+$, 297.1715; found 297.1716.

N,N-Dibenzyl-O-(4-azidobutyl)hydroxylamine (66r).

Compound **66r** (45 mg, 54%) was prepared according to general procedure **F** using **65r** (100 mg, 0.27 mmol) with warming to 0 °C and stirring for an additional 3 h. It was isolated as a yellow oil (eluent: 0 – 5% EtOAc in hexane). IR (neat,cm^{-1}) √ 2095; ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.42 – 7.36 (m, 4H), 7.36 – 7.24 (m, 6H), 3.85 (s, 4H), 3.28 (t, J = 5.7 Hz, 2H), 2.95 (t, J = 6.5 Hz, 2H), 1.30 – 1.25 (m, 4H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.8, 129.6, 128.1, 127.2, 72.0, 62.7, 50.9, 25.6, 25.5; ESIHRMS calculated for C$_{18}$H$_{23}$N$_4$O [M+H]$^+$, 311.1872; found 311.1885.

N,N-Dibenzyl-O-(1-phenyl-3-buten-1-yl)hydroxylamine (70a).

A solution of **62a** (51 mg, 0.16 mmol) in dry CH$_2$Cl$_2$ (1 mL) was cooled to –78 °C. A 1 M solution of DIBAL in hexane (0.32 mL, 0.32 mmol), pyridine (39 μL, 0.48 mmol), a solution of DMAP (39 mg, 0.32 mmol) in dry CH$_2$Cl$_2$ (0.5 mL), and Ac$_2$O (91 μL, 0.96 mmol) were added sequentially, while maintaining the reaction temperature at –78 °C. The reaction temperature was maintained for the next 12 h and was warmed to 0 °C in 2.5 h. After quenching reaction by the additions of saturated aq. NH$_4$Cl and saturated aq. sodium potassium tartrate at 0 °C, the resultant mixture was vigorously stirred vigorously at room temperature for 1 h. Then the mixture was extracted with dichloromethane, and the obtained organic layer was washed with brine, dried over anhydrous Na$_2$SO$_4$, and

concentrated. The crude residue **65a** was dissolved in dry dichloromethane (3.2 mL) and cooled to –78 °C. Allyltributylstannane (124 µL, 0.40 mmol) and $BF_3 \cdot OEt_2$ (51 µL, 0.40 mmol) were added one after the other, and the reaction mixture was warmed to 0°C in 2.5 h. The reaction was quenched by the addition of saturated aq. $NaHCO_3$ and was extracted with pentane. Then the obtained organic extract was dried over anhydrous Na_2SO_4, concentrated, and purified in flash column chromatography on silica gel (0 – 5% of EtOAc in hexane). The desired product **70a** (45 mg, 82%) was obtained as a colorless oil. ^1H NMR (400 MHz, $CDCl_3$) δ 7.35 – 7.18 (m, 15H), 5.45 – 5.32 (m, 1H), 4.90 – 4.81 (m, 2H), 4.18 (t, J = 7.1 Hz, 1H), 3.83 (d, J = 13.2 Hz, 2H), 3.71 (d, J = 12.7 Hz, 2H), 2.56 – 2.46 (m, 1H), 2.25 – 2.15 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 141.7, 137.8, 134.7, 129.7, 128.1, 128.0, 127.7, 127.5, 127.2, 116.5, 84.7, 62.5, 39.5; ESIHRMS calculated for $C_{24}H_{26}NO$ $[M+H]^+$, 344.2014; found, 344.2012.

N-Benzyl-N-methyl-O-(1-phenylbut-3-en-1-yl)hydroxylamine (70b).

A solution of **62b** (100 mg, 0.41 mmol) in dry CH_2Cl_2 (2.5 mL) was cooled to –78 °C. A 1 M solution of DIBAL in hexane (0.83 mL, 0.83 mmol), pyridine (101 µL, 1.24 mmol), a solution of DMAP (101 mg, 0.83 mmol) in dry CH_2Cl_2 (1.2 mL), and Ac_2O (234 µL, 2.49 mmol) were added sequentially, while maintaining the reaction temperature at –78 °C. The reaction temperature was maintained for the next 12 h and was warmed 0 °C in 2.5 h. After quenching reaction by the additions of saturated aq. NH_4Cl and saturated aq. sodium potassium tartrate at 0 °C, the resultant mixture was vigorously stirred at room temperature for 1 h. Then the mixture was extracted with CH_2Cl_2, and the obtained organic extract was washed with brine, dried over anhydrous Na_2SO_4, and concentrated. The so-obtained crude intermediate **65b** was dissolved in dry CH_2Cl_2 (8.3 mL) and cooled

to –78 °C. Allyltributylstannane (322 µL, 1.04 mmol) and BF$_3$.OEt$_2$ (131 µL, 1.04 mmol) were added one after the other, and the mixture was warmed to 0 °C in 2.5 h. The reaction was quenched by the addition of saturated aq. NaHCO$_3$ and was extracted with pentane. Then the obtained organic layer was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified in flash column chromatography on silica gel (0 – 5% of EtOAc in hexane). The desired product **70b** (59 mg, 54%) as obtained as a colorless oil. ^{1}H NMR (500 MHz, toluene-d$_8$, T = 323 K) δ 7.25 – 6.99 (m, 10H), 5.70 – 5.58 (m, 1H), 4.95 – 4.86 (m, 2H), 4.46 (t, *J* = 6.9 Hz, 1H), 3.66 (s, 2H), 2.62 – 2.53 (m, 1H), 2.40 – 2.25 (m, 4H); ^{13}C NMR (125 MHz, toluene-d$_8$, T = 323 K) δ 143.5, 138.5, 135.7, 130.4, 128.7, 128.6, 127.9, 127.8, 117.0, 84.6, 66.1, 45.9, 41.0; ESIHRMS calculated for C$_{18}$H$_{22}$NO [M+H]$^+$, 268.1701; found, 268.1699.

O-(1-(Adamantan-1-yl)but-3-en-1-yl)-*N,N*-dibenzylhydroxylamine (70c).

A solution of **65h** (100 mg, 0.24 mmol) in dry CH$_2$Cl$_2$ (4.8 mL) was cooled to –78 °C. Allyltributylstannane (186 µL, 0.60 mmol) and BF$_3$.OEt$_2$ (76 µL, 0.60 mmol) were added sequentially maintaining the reaction temperature at –78 °C, and the reaction was warmed to 0 °C in 2.5 h. After quenching reaction by the addition of aq. NaHCO$_3$, the resultant mixture was extracted with pentane. The so-obtained organic layer was dried over anhydrous Na$_2$SO$_4$, concentrated, and passed through a small pad of silica buffered with 2% triethyl amine in 2:98 mixture of EtOAc and hexane (200 mL). Then the filtrate was concentrated and purified by column chromatography on silica gel (eluent: same buffered solution of EtOAc in hexane). The desired product **70c** (76 mg, 79%) was isolated as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.36 – 7.21 (m, 10H), 5.88 – 5.77 (m, 1H), 4.93 – 4.82 (m, 2H), 3.90 (d, *J* = 10.3 Hz, 2H), 3.73 (d, *J* = 13.2 Hz, 2H), 3.04

(dd, *J* = 4.4, 5.3 Hz, 1H), 2.23 – 2.14 (m, 1H), 2.03 – 1.95 (m, 1H), 1.90 (s, 3H), 1.70 – 1.54 (m, 9H), 1.47 – 1.39 (m, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 139.2, 137.9, 129.8, 128.0, 127.1, 113.9, 87.6, 61.4, 38.6, 37.3, 37.0, 33.4, 28.4; ESIHRMS calculated for C$_{28}$H$_{36}$NO [M+H]$^+$, 402.2797; found, 402.2809.

1-(Dibenzylaminoxy)-4,4-dimethyl-1-phenylpentan-3-one (70d).

A solution of **62a** (100 mg, 0.32 mmol) in dry CH$_2$Cl$_2$ (1.9 mL) was cooled to –78 ^{o}C. A 1 M DIBAL solution in hexane DIBAL (0.64 mL, 0.64 mmol), pyridine (78 μL, 0.96 mmol), a solution of DMAP (78 mg, 0.64 mmol) in dry CH$_2$Cl$_2$ (0.9 mL), and Ac$_2$O (182 μL, 1.92 mmol) were added one after the other, while maintaining the reaction temperature at –78 ^{o}C. The reaction temperature was maintained for 12 h at –78 ^{o}C, and then the mixture was warmed to 0 ^{o}C in 2.5 h. After quenching the reaction with saturated aq. NH$_4$Cl (3.2 mL) and saturated aq. sodium potassium tartrate (2.4 mL), the resultant suspension was vigorously stirred at room temperature for 1 h followed by extraction with dichloromethane. Then the obtained organic layer was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The so-obtained crude intermediate **65a** was dissolved in dry CH$_2$Cl$_2$ (6.4 mL) was cooled to –78 ^{o}C, and 3,3-dimethyl-2-(trimethylsilyloxy)butene (173 μL, 0.80 mmol) and BF$_3$·OEt$_2$ (101 μL, 0.80 mmol) were added sequentially, dropwise. The reaction was then warmed to 0 ^{o}C in 2.5 h before quenching with saturated aq. NaHCO$_3$. The reaction mixture extracted with pentane, and the organic layer was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified in flash column chromatography on neutral alumina (10 – 25% of CH$_2$Cl$_2$ in hexane). The desired product **70d** (85 mg, 66%) was isolated as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.19 (m, 13H), 7.17 – 7.13 (m, 2H), 4.82 (dd, *J* = 8.3, 5.4 Hz, 1H), 3.85 (d, *J* = 12.7

Hz, 2H), 3.74 (d, J = 12.7 Hz, 2H), 2.80 (dd, J = 16.6, 5.4 Hz, 1H), 2.38 (dd, J = 16.6, 8.3 Hz, 1H), 0.87 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 213.2, 141.3, 137.5, 129.9, 128.1, 128.0, 127.6, 127.4, 127.2, 79.6, 61.9, 44.1, 42.2, 25.8; ESIHRMS calculated for C$_{27}$H$_{32}$NO$_2$ [M+H]$^+$, 402.2433; found, 402.2431.

N,N-Dibenzyl-O-((5-methylfuran-2-yl)benzyl)hydroxylamine (70e).

A solution of **62a** (100 mg, 0.32 mmol) in dry CH$_2$Cl$_2$ (1.9 mL) was cooled to −78 °C, and DIBAL (0.64 mL, 1 M in hexane, 0.64 mmol), pyridine (78 μL, 0.96 mmol), a solution of DMAP (78 mg, 0.64 mmol) in dry CH$_2$Cl$_2$ (0.9 mL), and Ac$_2$O (182 μL, 1.92 mmol) were added one after the other maintaining the reaction temperature at −78 °C. The reaction was allowed to stir for the next 12 h at −78 °C, and was warmed to 0 °C in 2.5 h. After quenching the reaction by the addition of saturated aq. NH$_4$Cl (3.2 mL) and saturated aq. sodium potassium tartrate (2.4 mL), the resultant suspension was vigorously stirred at room temperature for 1 h. The mixture was then extracted with dichloromethane, and the organic layer was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The so-obtained crude intermediate **65a** was dissolved in dry CH$_2$Cl$_2$ (6.4 mL) and cooled to −78 °C. 2-methylfuran (72 μL, 0.80 mmol) and BF$_3$·OEt$_2$ (101 μL, 0.80 mmol) were added sequentially, and the reaction mixture was quenched by adding saturated aq. NaHCO$_3$ after stirring for 45 min at −78 °C. The resultant suspension was extracted with pentane, and the organic layer was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified in flash column chromatography on neutral alumina (0 – 5% of EtOAc in hexane). Product **70e** (63 mg, 51%) was obtained as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.23 (m, 15H), 5.96 (d, J = 2.9 Hz, 1H), 5.87 (dd, J = 2.9, 1.0 Hz, 1H), 5.16 (s, 1H), 3.86 (d, J = 12.7 Hz, 2H), 3.80 (d, J = 12.7 Hz, 2H), 2.26 (d, J = 0.5

Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 152.2, 151.9, 139.0, 137.8, 129.7, 128.1, 128.0, 127.9, 127.7, 127.1, 110.5, 106.0, 80.3, 62.4, 13.7; ESIHRMS calculated for C$_{26}$H$_{26}$NO$_2$ [M+H]$^+$, 384.1964; found, 384.1965.

N,N-Dibenzyl-O-(1-(5-methylfuran-2-yl)-4-phenylbutyl)hydroxylamine (70f).

A solution of **65g** (50 mg, 0.12 mmol) in dry CH$_2$Cl$_2$ (2.5 mL) was cooled to –78 °C. 2-methylfuran (28 µL, 0.31 mmol) and BF$_3$.OEt$_2$ (40 µL, 0.31 mmol) were added one after the other maintaining the reaction temperature at –78 °C. The reaction was warmed to 0 °C in 2.5 h and was quenched by adding saturated aq. NaHCO$_3$. The resultant suspension was extracted with pentane, and the organic layer was dried over anhydrous Na$_2$SO$_4$, and concentrated. On purification of the residue by flash column chromatography on neutral alumina (0 – 5% of EtOAc in hexane), product **70f** (11 mg, 20%) was obtained as a colorless oil along with the recovered starting material **65g** (35 mg, 70%). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 – 7.19 (m, 12H), 7.16 (t, J = 7.3 Hz, 1H), 7.04 (d, J = 6.9 Hz, 2H), 6.06 (d, J = 2.9 Hz, 1H), 5.87 – 5.85 (m, 1H), 4.12 (t, J = 7.1 Hz, 1H), 3.78 (d, J = 12.7 Hz, 2H), 3.62 (d, J = 13.2 Hz, 2H), 2.45 – 2.30 (m, 2H), 2.24 (s, 3H), 1.76 – 1.64 (m, 1H), 1.61 – 1.49 (m, 1H), 1.44 – 1.31 (m, 1H), 1.30 – 1.13 (m, 1H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 153.1, 151.4, 142.3, 137.9, 129.6, 128.4, 128.1, 128.0, 127.1, 125.5, 109.1, 105.9, 77.7, 62.4, 35.4, 31.6, 27.2, 13.6; ESIHRMS calculated for C$_{29}$H$_{32}$NO$_2$ [M+H]$^+$, 426.2433; found, 426.2426.

(R)-N,N-Dibenzyl-O-(2-methyl-3-phenylpropanoyl)hydroxylamine (72).

To an ice-cold solution (<5 °C) of **71** (1 g, 3.09 mmol) in THF-H$_2$O (3:1, 48 mL) were added LiOH.H$_2$O (0.26 g, 6.18 mmol) and H$_2$O$_2$ (3.50 g, 30 wt% in water, 30.90 mmol). After stirring for 2 h at room temperature, the reaction flask was cooled in ice-bath

(<5 °C), and the reaction was quenched with Na_2SO_3 (30 mL 1.5 M in water, 45.00 mmol). The volatile organic components were evaporated in a rotary evaporator (<25 °C bath temperature), and the aqueous residue was washed out with CH_2Cl_2 to discard non-polar impurities. Then the aqueous layer was cooled in an ice-bath (<5 °C), acidified to pH 1 by addition of 2 M HCl, saturated with NaCl, and extracted with ether. The ether extract was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure. The so-obtained crude intermediate acid was dissolved in dry CH_2Cl_2 (20 mL) together with *N,N*-dibenzylhydroxylamine **63a** (0.99 g, 4.64 mmol) and the resulting solution was cooled in an ice-bath (<5 °C). To this solution were added HOBt (0.78 g wetted with 20 wt% water, 4.64 mmol), EDCI.HCl (0.89 g, 4.64 mmol), and DIPEA (0.81 mL, 4.64 mmol), and the reaction was allowed to stir for 4 h at room temperature. The mixture was washed with water followed by brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (0 − 5% EtOAc in hexane) to give **72** (0.96 g, 87%) as a colorless oil. $[α]^{20}_D$ −8.7 (*c* 0.9, CHCl3); ^1H NMR (400 MHz, CDCl3) δ 7.43 − 7.13 (m, 13H), 7.05 (d, *J* = 7.7 Hz, 2H), 3.99 (ABq, *J* = 13.4 Hz, 4H), 2.73 (dd, *J* = 13.1, 6.7 Hz, 1H), 2.54 − 2.44 (m, 1H), 2.40 (dd, *J* = 13.1, 7.6 Hz, 1H), 0.82 (d, *J* = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CDCl3) δ 174.1, 139.3, 135.9, 129.4, 129.0, 128.3, 128.2, 127.6, 126.3, 62.1, 40.2, 39.3, 16.6; ESIHRMS calculated for $C_{24}H_{25}NO_2Na$ [M+Na]+, 382.1783; found, 382.1798.

(2*R*)-*N,N*-Dibenzyl-*O*-(1-acetoxy-2-methyl-3-phenylpropyl)hydroxylamine (73).

A solution of **72** (200 mg, 0.56 mmol) in dry CH_2Cl_2 (3.5 mL) was chilled at −78 °C. A 1 M DIBAL solution in hexane (0.98 mL, 0.98 mmol), pyridine (135 µL, 1.68 mmol), a solution of DMAP (137 mg, 1.12 mmol) in dry CH_2Cl_2 (1.5 mL), and Ac_2O (317 µL, 3.36

mmol) were added one after the other, while maintaining the reaction temperature at –78 °C . The reaction temperature was maintained for next 12 h at –78 °C, and then it was warmed to 0 °C in 2.5 h followed by the addition of saturated aq. NH$_4$Cl (5 mL) and saturated aq. sodium potassium tartrate (4 mL). The resultant suspension was vigorously stirred for 1 h, and was extracted with CH$_2$Cl$_2$. The organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by flash column chromatography on silica gel (0 – 5% EtOAc in hexane). The desired product **73** (145 mg, 65%, dr 56:44) was obtained as a colorless oil. [α]$^{20}_D$ +7.2 (*c* 2, CH$_2$Cl$_2$); ^{1}H NMR (400 MHz, CDCl$_3$), both diastereomers, δ 7.48 – 6.96 (m, 15H), 5.91 (d, *J* = 4.7 Hz, 0.44H), 5.87 (d, *J* = 4.7 Hz, 0.56H), 4.02 (br s, 1.76H), 3.88 – 3.80 (m, 2.24H), 2.74 (dd, *J* = 13.5, 4.3 Hz, 0.56H), 2.58 (dd, *J* = 13.6, 4.1 Hz, 0.44H), 2.11 (dd, *J* = 13.6, 10.6 Hz, 0.44H), 1.99 (dd, *J* = 13.5, 10.3 Hz, 0.56H), 1.92 – 1.78 (m, 4H), 0.71 – 0.60 (m, 3H); ^{13}C NMR (100 MHz, CDCl$_3$), both diastereomers, δ 170.4, 170.4, 140.4, 140.3, 137.1, 129.5, 129.0, 129.0, 128.3, 128.2, 127.4, 125.8, 125.8, 102.0, 62.6, 38.5, 38.3, 37.3, 37.2, 21.20, 21.17, 13.6, 13.3; ESIHRMS calculated for C$_{26}$H$_{29}$NO$_3$Na [M+Na]$^+$, 426.2045; found, 426.2044.

***N,N*-Dibenzyl-*O*-((2*R*)-2-methyl-1-phenylhex-5-en-3-yl)hydroxylamine (74).**

To a stirred solution of **73** (100 mg, 0.25 mmol) in dry CH$_2$Cl$_2$ (5 mL) were added allyltributylstannane (194 µL, 0.63 mmol) and BF$_3$·OEt$_2$ (77 µL, 0.63 mmol) one after the other at -78 °C. The reaction mixture was warmed slowly to 0 °C in 2.5 h, quenched by saturated aq. NaHCO$_3$, and extracted with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by flash column chromatography on silica gel (0 – 4% EtOAc in hexane). The desired product **74** (83 mg, 87%) was isolated as a colorless oil and was revealed as a mixture of two diastereomers **74a** (minor) and **74b**

(major) in a ratio of 1:1.2 (**74a**:**74b**) from the NMR spectra. $[\alpha]^{20}_D$ –0.6 (c 3.25, CHCl₃);

¹H NMR (600 MHz, CDCl₃), both diastereomers, δ 7.63 – 6.84 (m, 15H), 5.67 – 5.53 (m,

1H), 5.01 – 4.87 (m, 2H), 4.00 – 3.71 (m, 4H), 3.43 (td, J = 6.3, 3.6 Hz, 0.55H), 3.37 (dt,

J = 6.8, 4.9 Hz, 0.45H), 2.65 (dd, J = 13.4, 4.4 Hz, 0.55H), 2.60 (dd, J = 13.6, 6.0 Hz,

0.45H), 2.32 (dt, J = 14.0, 6.3 Hz, 0.55H), 2.26 (dd, J = 13.4, 10.5 Hz, 0.55H), 2.16 (dd,

J = 13.6, 9.0 Hz, 0.45H), 2.13 – 2.08 (m, 0.90H), 2.02 – 1.89 (m, 1.55H), 0.69 (d, J = 6.9

Hz, 1.65H), 0.57 (d, J = 6.8 Hz, 1.35H); ¹³C NMR (150 MHz, CDCl₃) both diastereomers,

δ 141.6, 141.5, 137.8, 136.2, 136.0, 129.7, 129.7, 129.6, 129.04, 129.02, 128.10, 128.09,

128.06, 128.04, 127.2, 127.1, 125.6, 125.5, 116.0, 115.9, 83.71, 83.69, 62.2, 62.1, 39.1,

38.9, 36.5, 36.4, 34.6, 33.3, 14.2, 13.6; ESIHRMS calculated for $C_{27}H_{32}NO$ [M+H]⁺,

386.2484; found, 386.2485.

N,N-Dibenzyl-O-((3R)-3-methyl-4-phenylbutan-2-yl)hydroxylamine (75).

To a stirred solution of **73** (40 mg, 0.10 mmol) in dry CH₂Cl₂ (2 mL) were added

Me₂Zn (250 µL, 1 M in heptane, 0.25 mmol) and BF₃·OEt₂ (31 µL, 0.25 mmol) one after

the other at -78 °C. The reaction mixture was warmed slowly to 0 °C in 2.5 h, quenched

by slow addition of saturated aq. NaHCO₃, and extracted with pentane. The organic

extract was dried over anhydrous Na₂SO₄, concentrated, and purified by flash column

chromatography on silica gel (0 – 5% EtOAc in hexane). Product **75** (28 mg, 77%) was

isolated as a colorless oil and was revealed as an uncharacterized mixture of two

diastereomers in a ratio of 1:1. $[\alpha]^{20}_D$ +2.1 (c 1.25, CHCl₃); ¹H NMR (600 MHz, CDCl₃),

both diastereomers, δ 7.47 – 6.98 (m, 15H), 4.00 – 3.73 (m, 4H), 3.44 (dq, J = 6.3, 3.6

Hz, 0.48H), 3.34 (dq, J = 6.5, 4.1 Hz, 0.52H), 2.73 (dd, J = 13.4, 3.9 Hz, 0.48H), 2.52 (dd,

J = 13.6, 6.0 Hz, 0.52H), 2.16 (dd, J = 13.5, 10.8 Hz, 0.48H), 2.12 (dd, J = 13.7, 9.2 Hz,

0.52H), 1.98 – 1.89 (m, 0.52H), 1.90 – 1.80 (m, 0.48H), 0.89 (d, J = 6.3 Hz, 1.44H), 0.85 (d, J = 6.4 Hz, 1.56H), 0.66 (d, J = 6.9 Hz, 1.44H), 0.60 (d, J = 6.8 Hz, 1.56H); ^{13}C NMR (150 MHz, CDCl$_3$), both diasteromers, δ 141.8, 141.4, 137.9, 129.8, 129.7, 129.1, 128.9, 128.10, 128.06, 128.02, 127.2, 127.1, 125.6, 125.5, 80.6, 80.1, 62.7, 62.5, 39.7, 39.0, 38.1, 37.9, 15.4, 14.6, 14.1, 13.7; ESIHRMS calculated for C$_{25}$H$_{30}$NO [M+H]$^+$, 360.2327; found, 360.2325.

1-Boc-1-methyl-2,2-dibenzylhydrazine (77).

A mixture of *N*-Boc-*N*-methylhydrazine (1 g, 6.84 mmol), NaHCO$_3$ (1.44 g, 17.10 mmol), BnBr (2.03 mL, 17.10 mmol), DMF (2 mL), and toluene (6 mL) was stirred for 21 h at 85 °C. The reaction was cooled to room temperature, diluted with toluene, washed with water followed by brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (0 – 8% EtOAc in hexane) to give **77** (1.88 g, 84%) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.51 – 7.01 (m, 10H), 4.34 (d, J = 11.3 Hz, 1H), 4.01 (d, J = 12.0 Hz, 2H), 3.87 (d, J = 12.5 Hz, 1H), 2.69 (s, 3H, rotamer), 2.45 (s, 3H, rotamer), 1.47 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 156.3, 154.9, 138.5, 137.9, 129.4, 129.3, 128.1, 127.3, 79.8, 79.6, 58.1, 57.6, 39.2, 32.3, 28.6; ESIHRMS calculated for C$_{20}$H$_{26}$N$_2$O$_2$Na [M+Na]$^+$, 349.1892; found, 349.1898.

1,1-Dibenzyl-2-methylhydrazine (78).

Trifluoroacetic acid (15 ml) was added dropwise to a solution of **77** (940 mg, 2.88 mmol) in CH$_2$Cl$_2$ (15 mL) at 0 °C. After stirring for 1 h at 0 °C and 1 h at room temperature, more trifluoroacetic acid (5 mL) was added dropwise. The stirring was continued for the next 2 h at room temperature, and the mixture was concentrated. The residue was

dissolved in CH_2Cl_2 (10 mL), washed with 1 N NaOH followed by brine, dried over anhydrous Na_2SO_4, and concentrated to give desired product **78** (652 mg, 100% yield) as colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.58 – 7.23 (m, 10H), 3.79 (s, 4H), 2.66 (br s, 1H), 2.56 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 138.4, 128.9, 128.3, 127.1, 59.4, 35.1; ESIHRMS calculated for $C_{15}H_{19}N_2$ [M+H]$^+$, 227.1548; found, 227.1545.

N',N'-Dibenzyl-N-methyl-4-phenylbutanehydrazide (79).

To a stirred solution of **78** (347 mg, 1.53 mmol), 4-phenylbutyric acid (251 mg, 1.53 mmol), and DMAP (38 mg, 0.31 mmol) in dry dichloromethane (15 mL) was added DCC (316 mg, 1.53 mmol) at room temperature. The reaction mixture was allowed to stir for 2 h and filtered through a pad of Celite®. The resulting filtrate was concentrated and purified by flash column chromatography on silica gel (0 – 10% EtOAc in hexane) to afford **79** (549 mg, 96% yield) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.37 – 7.06 (m, 15H), 3.81 (ABq, J = 12.8 Hz, 4H), 3.06 (s, 3H), 2.39 (t, J = 7.6 Hz, 2H), 2.16 (t, J = 7.7 Hz, 2H), 1.60 – 1.43 (m, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 176.0, 142.3, 136.4, 129.5, 129.4, 128.6, 128.2, 127.9, 125.7, 57.8, 35.5, 31.3, 26.2, 24.0; ESIHRMS calculated for $C_{25}H_{28}N_2ONa$ [M+Na]$^+$, 395.2099; found, 395.2105.

1,1-Dibenzyl-2-methyl-2-(4-phenylbutyl)hydrazine (81) and N',N'-dibenzyl-N-methylacetohydrazide (82).

To a stirred solution of **79** (93 mg, 0.25 mmol) in dry dichloromethane (1.5 mL) was added DIBAL (0.38 mL 1 M in cyclohexane, 0.38 mmol) dropwise at -78 °C. The reaction mixture was stirred for 30 min, and pyridine (60 µL, 0.75 mmol), a solution of DMAP (61 mg, 0.50 mmol) in dry dichloromethane (0.7 mL), and acetic anhydride (142 µL, 1.5 mmol) were sequentially added. The reaction temperature was maintained at -78

°C for 12 h and allowed to warm to 0 °C in 2.5 h. After addition of saturated aq. NH_4Cl (2.5 mL) and saturated aq. Rochelle's salt (2 mL), the resulting suspension was stirred vigorously at room temperature. The suspension was extracted with dichloromethane and the obtained organic extract was dried over anhydrous Na_2SO_4, concentrated, and purified by flash column chromatography on silica gel (0 – 60% EtOAc in hexane). Two unknown products were isolated, which were characterized as **81** (30 mg colorless oil, 33% yield) and **82** (20 mg colorless oil, 30% yield) from NMR analysis.

Data for **81**; ^1H NMR (400 MHz, $CDCl_3$) δ 7.41 – 7.05 (m, 15H), 3.64 (s, 4H), 2.56 (t, J = 6.4 Hz, 2H), 2.51 – 2.45 (m, 2H), 2.42 (s, 3H), 1.49 – 1.34 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 142.9, 140.2, 129.0, 128.5, 128.1, 128.0, 126.6, 125.5, 54.4, 53.1, 35.8, 35.4, 28.9, 27.4; ESIHRMS calculated for $C_{25}H_{31}N_2O$ [M+H]$^+$, 359.2487; found, 359.2498.

Data for **82**; ^1H NMR (400 MHz, $CDCl_3$) δ 7.44 – 7.18 (m, 10H), 3.82 (ABq, J = 12.7 Hz, 4H), 3.03 (s, 3H), 1.72 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.0, 136.4, 129.4, 128.6, 127.9, 57.9, 23.9, 20.7; ESIHRMS calculated for $C_{17}H_{20}N_2ONa$ [M+Na]$^+$, 291.1473; found, 291.1471.

Alternative synthesis of 1,1-dibenzyl-2-methyl-2-(4-phenylbutyl)hydrazine (81).

To a stirred solution of **79** (50 mg, 0.13 mmol) in dry dichloromethane (1 mL) was added DIBAL (0.39 mL 1 M in cyclohexane, 0.39 mmol) dropwise at -78 °C. The reaction mixture was warmed to 0 °C in 2.5 h and quenched with saturated aq. NH_4Cl (2 mL) and saturated aq. Rochelle's salt (1.5 mL). The resulting suspension was stirred vigorously at room temperature and extracted with dichloromethane. The so-obtained organic extract was dried over anhydrous Na_2SO_4, concentrated, and purified by flash column

chromatography on silica gel (0 – 5% EtOAc in hexane) to afford **81** (31 mg, 65% yield) as a colorless oil. The spectral data were identical with above sample.

1,1-Dibenzyl-2-methyl-2-(7-phenylhept-1-en-4-yl)hydrazine (83).

To an ice-cold (<5 °C) solution of **79** (60 mg, 0.16 mmol) and allyltributylstannane (150 µL, 0.48 mmol) in dry dichloromethane (1 mL) was added DIBAL (0.20 mL 1M DIBAL in hexane, 0.20 mmol) dropwise. After stirring for 5 min, $BF_3.OEt_2$ (59 µL, 0.48 mmol) was added dropwise and the ice-bath was removed. The reaction was allowed to stir for 16 h at room temperature, and then quenched by adding saturated aq. NH_4Cl (2 mL) and saturated Rochelle's salt (1.5 mL). The resulting suspension was stirred vigorously for 1 h and was extracted with dichloromethane. The so-obtained organic extract was concentrated and purified by thin layer chromatography on silica gel (0 – 5% EtOAc in hexane) to give **83** (20 mg, 31% yield) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.37 – 7.02 (m, 15H), 5.49 (ddt, J = 17.1, 10.1, 7.0 Hz, 1H), 4.96 – 4.81 (m, 2H), 3.64 (ABq, J = 13.1 Hz, 4H), 2.65 – 2.56 (m, 1H), 2.52 – 2.33 (m, 6H), 2.08 – 1.96 (m, 1H), 1.58 – 1.42 (m, 1H), 1.38 – 1.19 (m, 3H). ^{13}C NMR (100 MHz, CDCl$_3$) δ 142.8, 139.8, 137.0, 129.4, 128.4, 128.1, 127.9, 126.7, 125.5, 115.4, 62.7, 55.0, 36.1, 34.8, 30.4, 29.9, 26.8; ESIHRMS calculated for $C_{28}H_{35}N_2$ [M+H]$^+$, 399.2800; found, 399.2804.

N',N'-Dibenzyl-N-methylbenzohydrazide (84).

To a stirred solution of **78** (227 mg, 1.23 mmol), benzoic acid (173 mg, 1.41 mmol), and DMAP (31 mg, 0.25 mmol) in dry dichloromethane (12 mL) was added DCC (291 mg, 1.41 mmol) at room temperature. The reaction mixture was allowed to stir for 12 h and filtered through a pad of Celite®. The resulting filtrate was concentrated and purified by flash column chromatography on silica gel (10 – 40% EtOAc in hexane) to afford **84**

(327 mg, 81% yield) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.40 – 7.12 (m, 9H), 6.99 (d, J = 7.0 Hz, 4H), 6.83 (d, J = 7.6 Hz, 2H), 3.99 – 3.72 (ABq, J = 13.0 Hz, 4H), 3.26 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 173.7, 137.0, 136.0, 129.3, 128.7, 128.4, 127.8, 127.2, 127.0, 57.4, 25.0; ESIHRMS calculated for C$_{22}$H$_{22}$N$_2$ONa [M+Na]$^+$, 353.1630; found, 363.1622.

1,1,2-Tribenzyl-2-methylhydrazine (85).

To a stirred solution of **84** (50 mg, 0.15 mmol) in dry dichloromethane (1 mL) was added DIBAL (0.45 mL 1 M in hexane, 0.45 mmol) dropwise at -78 °C. The reaction mixture was warmed to 0 °C in 2.5 h and quenched with saturated aq. NH$_4$Cl (2 mL) and saturated aq. Rochelle's salt (1.5 mL). The resulting suspension was stirred vigorously at room temperature and extracted with dichloromethane. The so-obtained organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated, and purified by flash column chromatography on silica gel (0 – 5% EtOAc in hexane) to afford **85** (29 mg, 61% yield) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.45 – 7.14 (m, 13H), 7.11 – 6.98 (m, 2H), 3.83 (s, 2H), 3.77 (s, 4H), 2.38 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 140.0, 139.4, 128.82, 128.82, 128.02, 127.99, 126.7, 126.6, 58.8, 53.7, 35.4; ESIHRMS calculated for C$_{22}$H$_{25}$N$_2$ [M+H]$^+$, 317.2018; found, 317.2029.

***N,N'*-Dimethyl-4-phenyl-*N'*-(4-phenylbutanoyl)butanehydrazide (86).**

To a stirred solution of *sym*-dimethylhydrazine dihydrochloride (100 mg, 0.75 mmol) and 4-phenylbutyric acid (309 mg, 1.88 mmol) in dry dichloromethane (8 mL) was added triethylamine (230 μL, 1.65 mmol) dropwise at room temperature. After stirring for 5 min, DMAP (37 mg, 0.30 mmol) and DCC (389 mg, 1.88 mmol) were added and the reaction mixture was stirred for next 5 h. The reaction was filtered through a pad of

Celite®, and the resulting filtrate was washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated. After purification via flash column chromatography on silica gel (20 – 60% EtOAc in hexane), the desired hydrazide **86** (181 mg, 69% yield) was isolated as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.42 – 7.03 (m, 10H), 3.04 (s, 6H), 2.62 (t, $J = 7.6$ Hz, 4H), 2.19 (t, $J = 7.4$ Hz, 4H), 2.00 – 1.86 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.6, 141.2, 128.6, 128.4, 126.1, 35.0, 33.5, 30.6, 25.9; ESIHRMS calculated for $C_{22}H_{28}N_2O_2Na$ [M+Na]+, 375.2048; found, 375.2052.

1,2-Dimethyl-1,2-bis(4-phenylbutyl)hydrazine (87).

To a stirred solution of **86** (50 mg, 0.14 mmol) in dry dichloromethane (1.7 mL) was added DIBAL (0.84 mL 1 M in hexane, 0.84 mmol) dropwise at -78 °C. The reaction mixture was warmed to 0 °C in 2.5 h and quenched with saturated aq. NH_4Cl (3 mL) and saturated aq. Rochelle's salt (2 mL). The resulting suspension was stirred vigorously at room temperature and extracted with dichloromethane. The so-obtained organic extract was dried over anhydrous Na_2SO_4, concentrated, and purified by flash column chromatography on silica gel (0 – 20% EtOAc in hexane) to afford **87** (21 mg, 46% yield) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.39 – 7.06 (m, 10H), 2.62 (t, $J = 7.7$ Hz, 4H), 2.46 (t, $J = 7.1$ Hz, 4H), 2.23 (s, 3H), 1.72 – 1.58 (m, 4H), 1.58 – 1.45 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 142.8, 128.4, 128.2, 125.6, 53.8, 35.9, 34.6, 29.3, 27.7; ESIHRMS calculated for $C_{22}H_{33}N_2$ [M+H]+, 325.2644; found, 325.2640.

Experimental section for chapter 3

VT-NMR study of nitrogen inversion in *N*-hydroxy and *N*-alkoxypiperidines:

Variable temperature (VT) NMR spectroscopy technique was used to determine inversion barriers for the compounds **91**, **92**, **94**, and **88**. ^{1}H NMR spectra for each

compound were recorded in 500 MHz instrument over the temperature range of 223 K to 363 K in Toluene-d$_8$ and 233 K to 363 K in DMF-d$_7$ (233 K to 393 K for **93** and **88**). Eyring equation (1) was applied to calculate inversion barriers.

$$\Delta G_C^{\neq} = 4.575 \times 10\text{-}3 \; T_C[9.972 + \log (T_C/\Delta v)] \qquad (1)$$

Table 9. Inversion barriers of *N*-alkoxy and *N*-hydroxypiperidines.
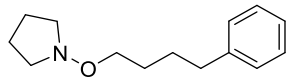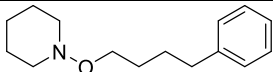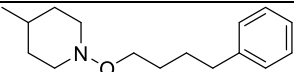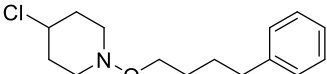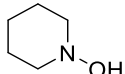
Entry	Compound	Inversion barrier ($\Delta G_c^{\neq}$) in kcal.mol^{-1}	
		Tolune-d$_8$ (NMR solvent)	DMF-d$_8$ (NMR solvent)
1	**94**	13.26	12.91
2	**91**	15.88	15.88
3	**93**	No coalescence below 363 K	No coalescence below 393 K
4	**92**	15.34	15.42
5	**88**	16.42	16.42

Figure 36. ^{1}H NMR spectra (Toluene-d$_8$) of **94** recorded at variable temperatures ranging from 223 K to 303 K (2.50 to 3.30 ppm). Tc is coalescence temperature for H-2.
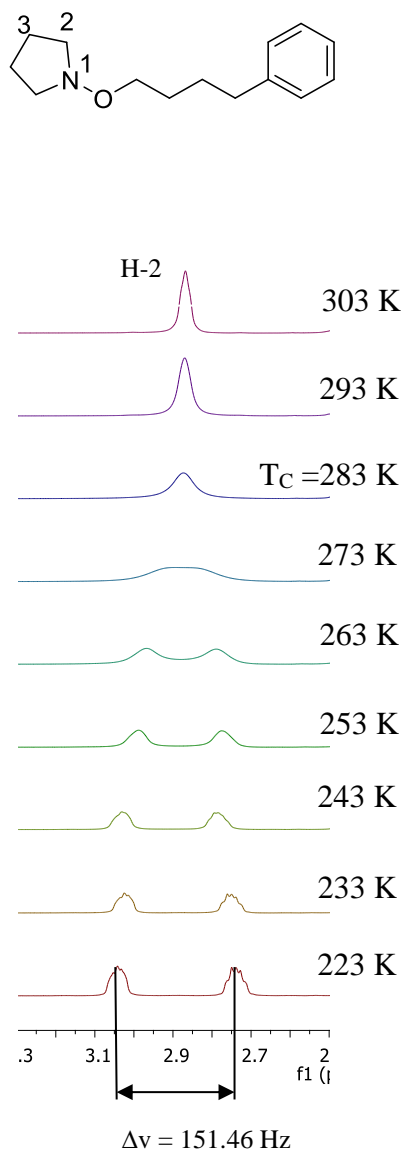
Figure 37. ^{1}H NMR spectra (DMF-d$_7$) of **94** recorded at variable temperatures ranging from 233 K to 283 K (2.40 to 3.20 ppm). Tc is coalescence temperature for H-2.
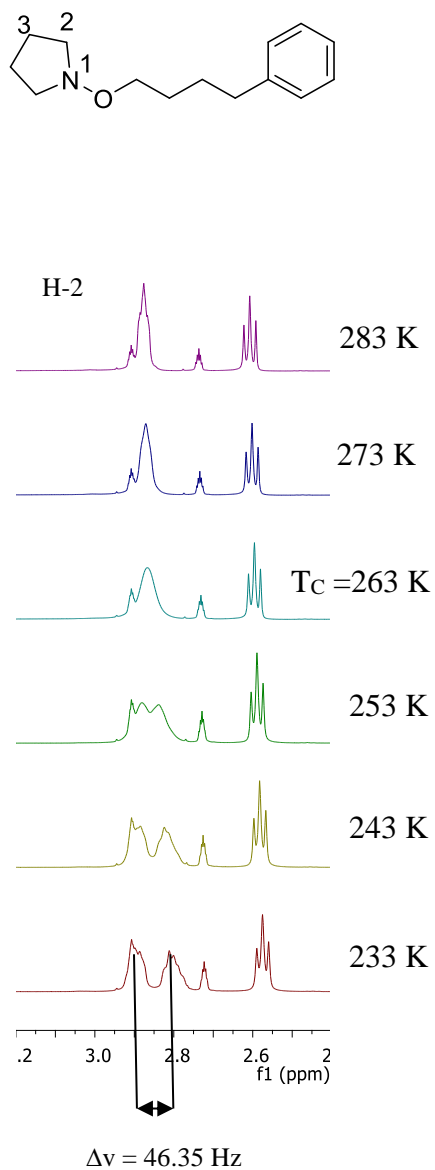
Figure 38. ^{1}H NMR spectra (Toluene-d8) of **91** recorded at variable temperatures ranging from 223 K to 363 K (1.90 to 3.50 ppm). Tc is coalescence temperature for H-2.
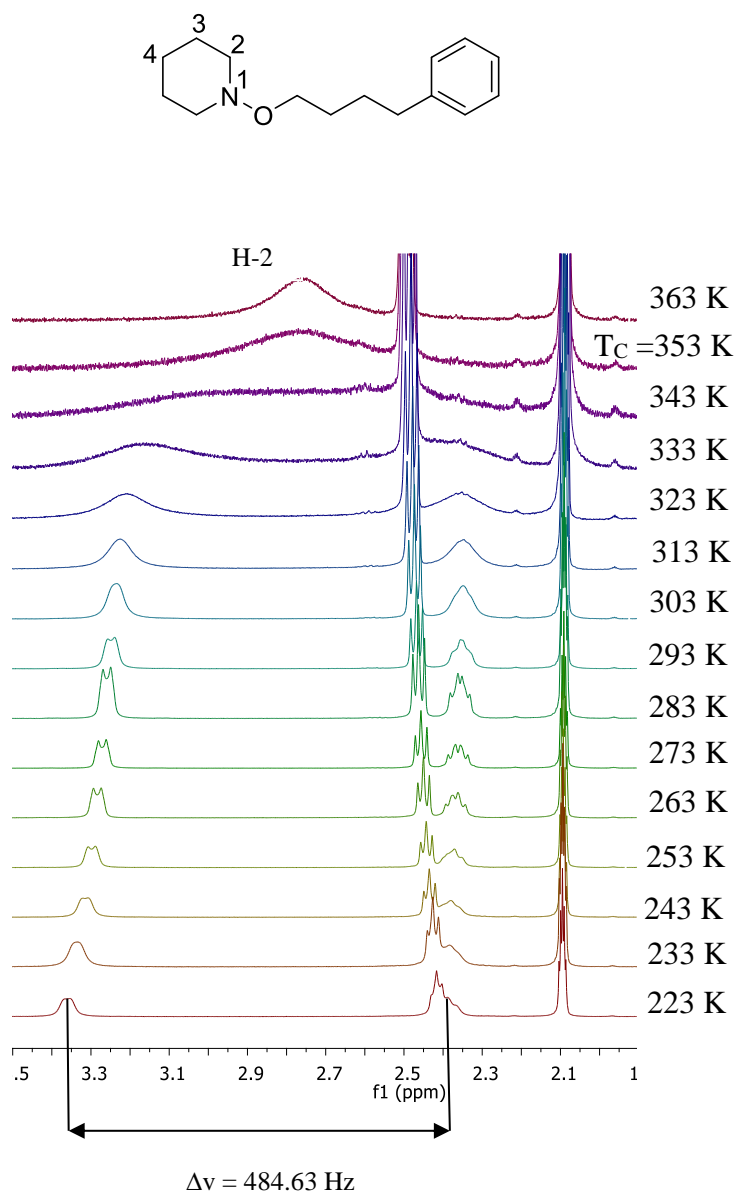
Figure 39. ^{1}H NMR spectra (DMF-d[7]) of **91** recorded at variable temperatures ranging from 263 K to 283 K (1.90 ppm 3.50 ppm). Tc is coalescence temperature for H-2.
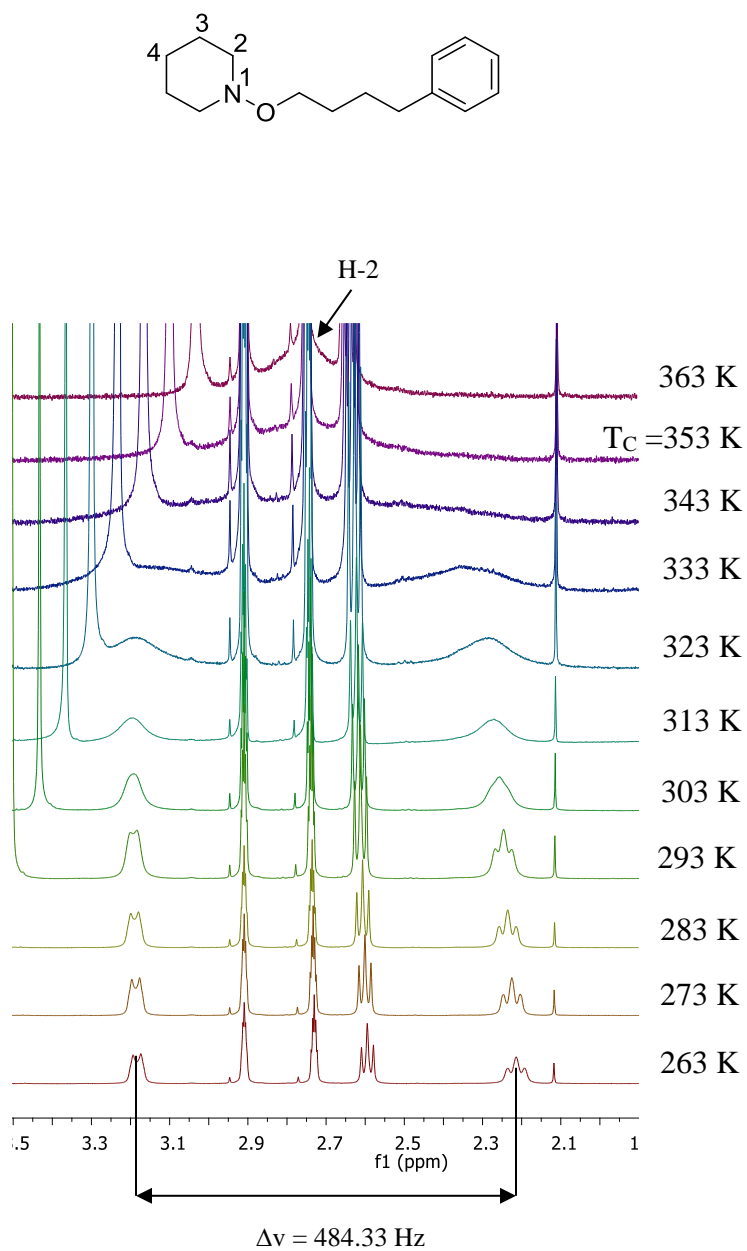
Figure 40. ¹H NMR spectra (Toluene-d₈) of **92** recorded at variable temperatures ranging from 253 K to 343 K (2.80 to 4.00 ppm). Tc is coalescence temperature for H-4.
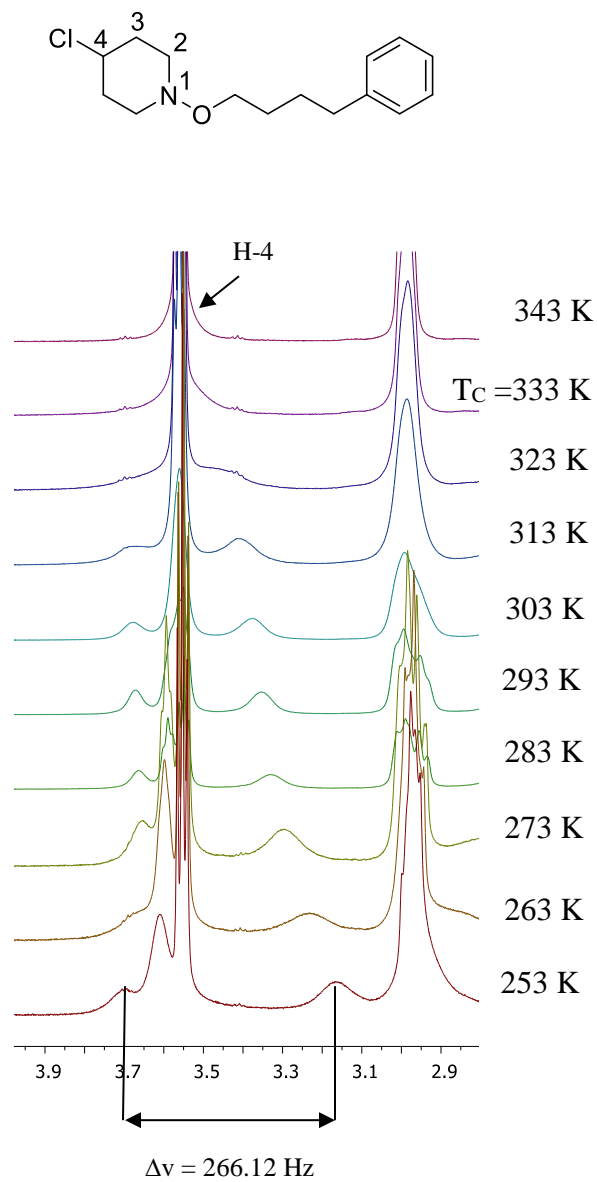
Figure 41. ^{1}H NMR spectra (DMF-d$_7$) of **92** recorded at variable temperatures ranging from 253 K to 343 K (3.80 to 4.90 ppm). Tc is coalescence temperature for H-4.
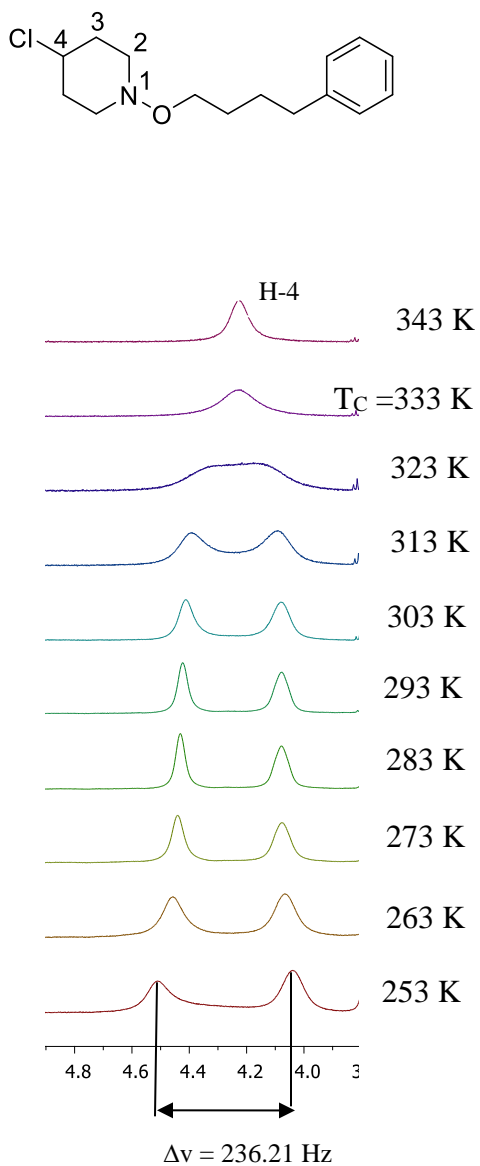
Figure 42. ^{1}H NMR spectra (Toluene-d$_8$) of **88** recorded at variable temperatures ranging from 233 K to 363 K (0.05 to 4.00 ppm). Tc is coalescence temperature for H-2.
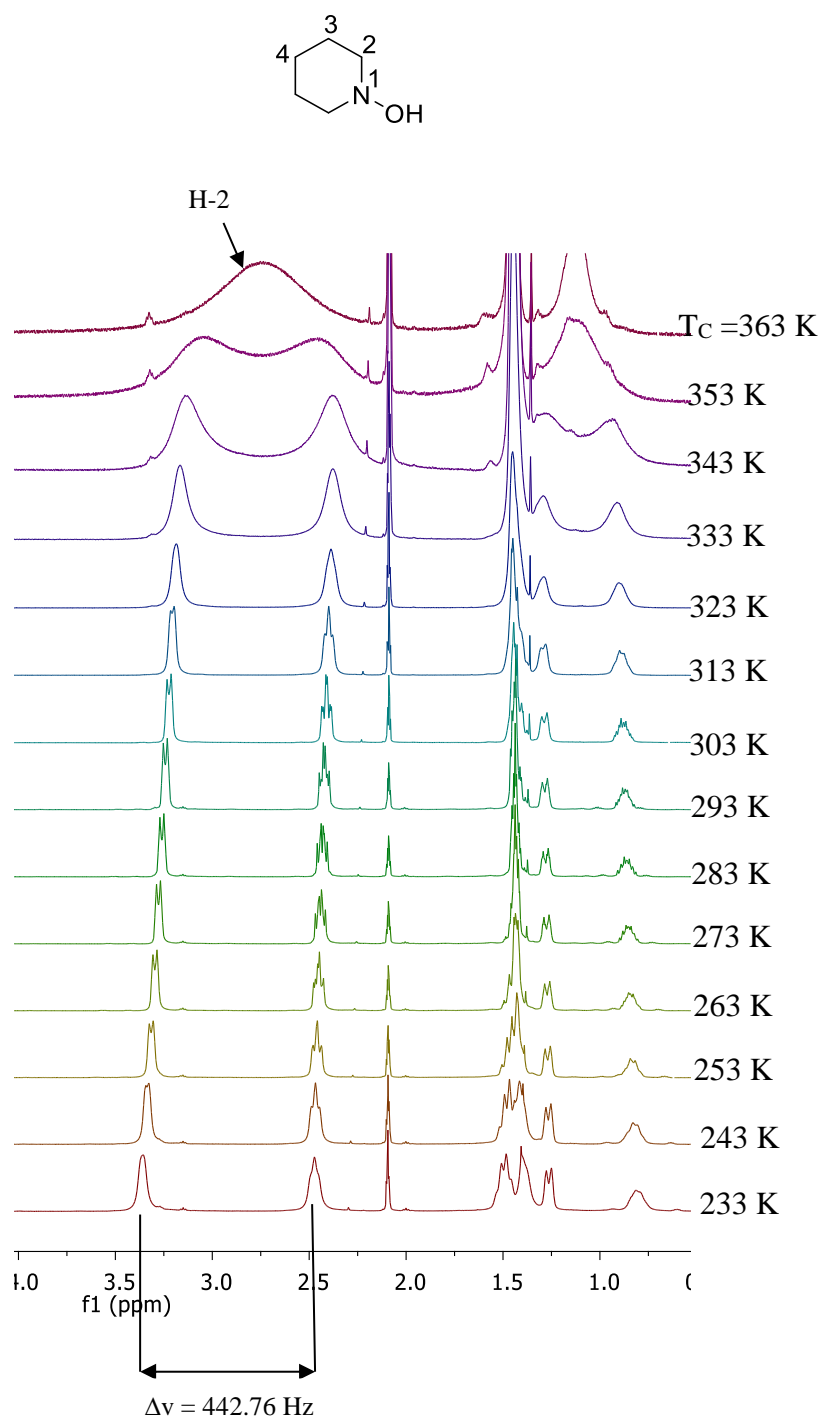
Figure 43. ^{1}H NMR spectra (DMF-d$_7$) of **88** recorded at variable temperatures ranging from 233 K to 363 K (0.05 to 0.40 ppm). Tc is coalescence temperature for H-2.
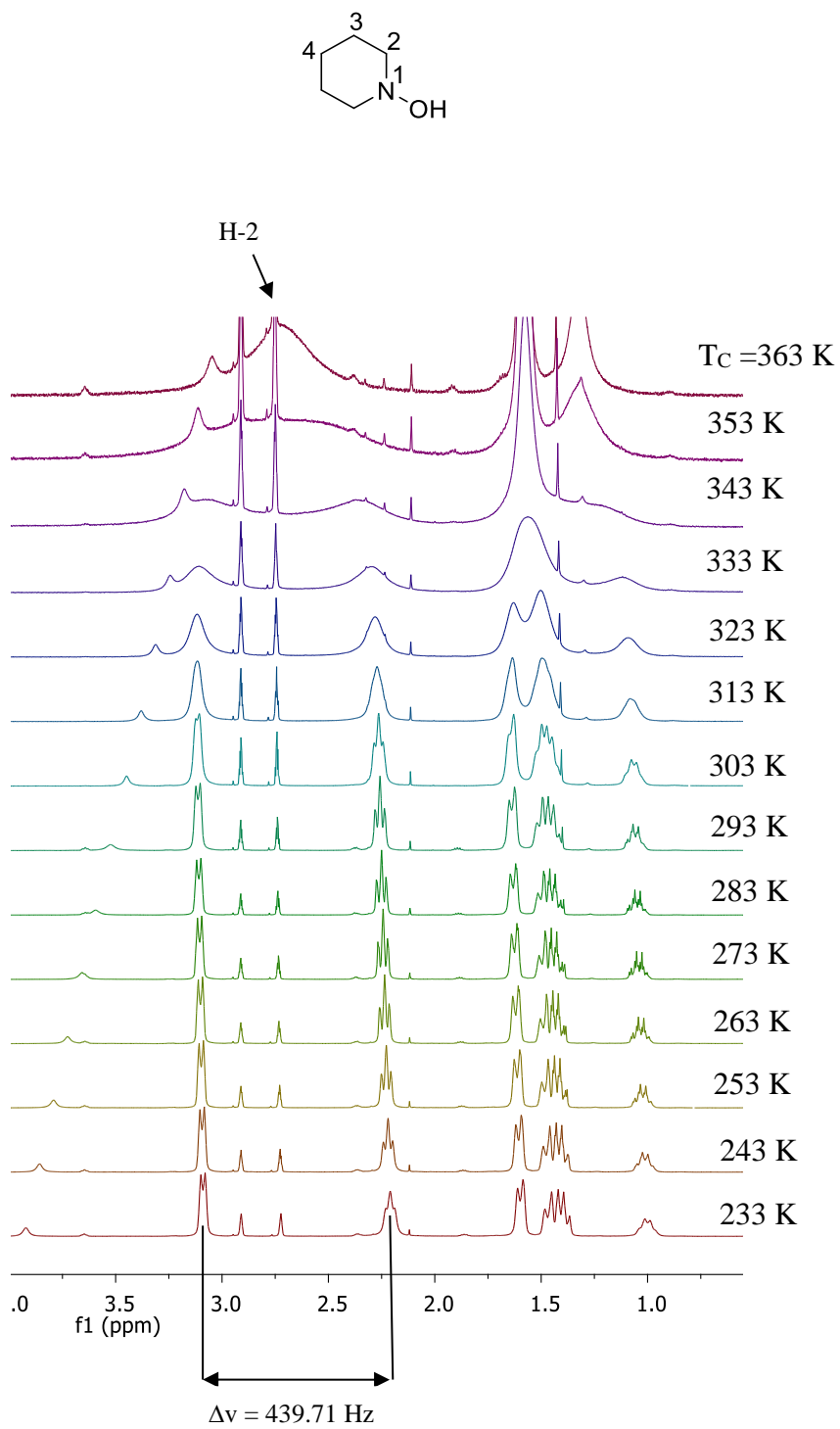
Figure 44. ^{1}H NMR (500 MHz, DMF-d$_7$, T = 393 K) spectrum of (**88**)
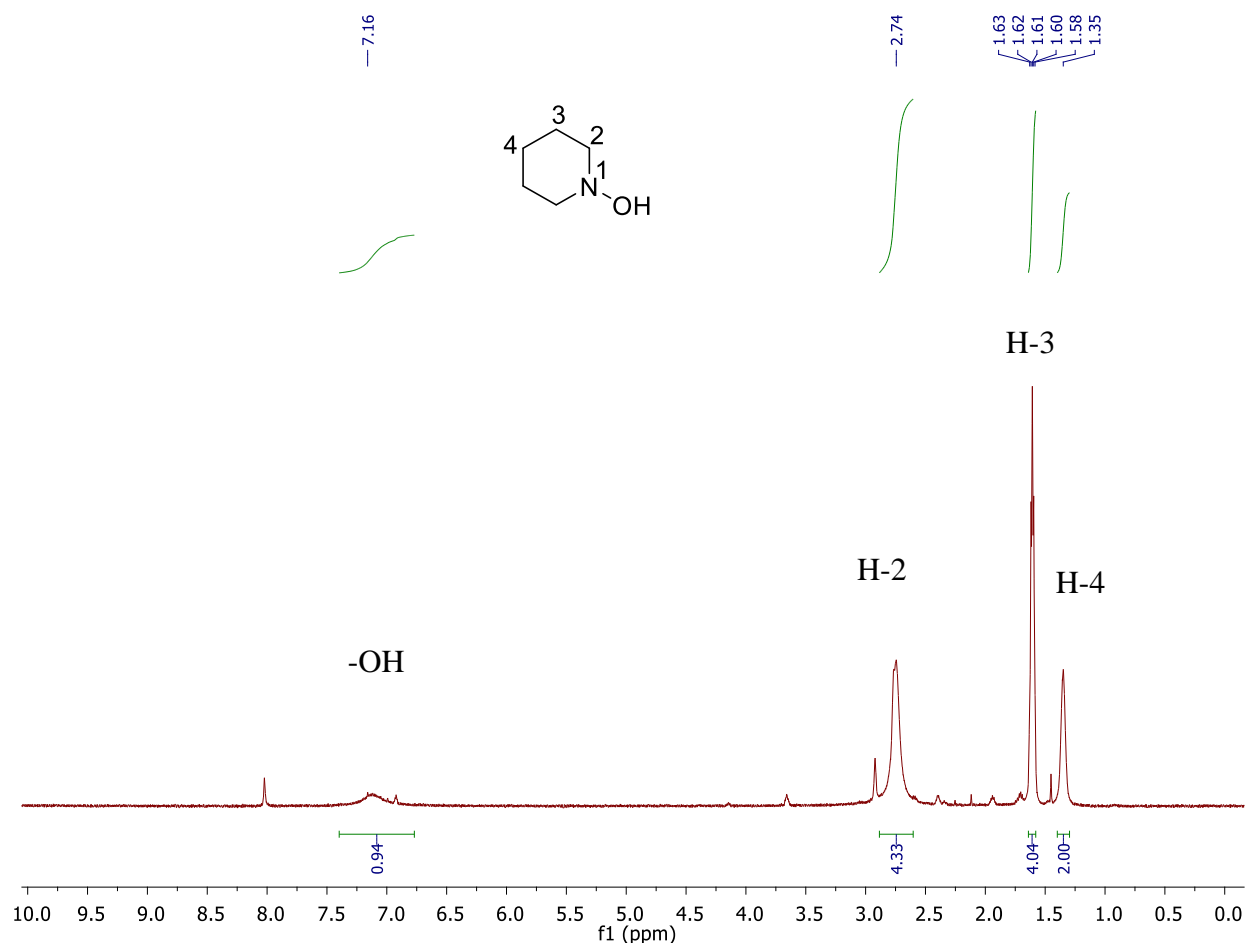
Experimental section for chapter 4

Synthesis of Kalkitoxin (107):

(*R*)-4-Benzyl-3-(4-(benzyloxy)butanoyl)oxazolidin-2-one (121).

Triethylamine (4.17 mL, 29.68 mmol) was added dropwise to a solution of acid **119**[145] in ether (220 mL), and the mixture was stirred for 20 min at room temperature. The reaction flask was cooled in ice-bath and ethyl chloroformate (2.46 mL, 25.74 mmol) was added slowly. After stirring for 1 h at room temperature, the resulting suspension was cooled to −78 °C. In a separate flask *n*-BuLi (12.5 mL, 2.5 M in hexanes, 31.25 mmol)

was added dropwise to a solution of (4*R*)-benzyloxazolidinone (4.56 g, 25.74 mmol) in THF (40 mL) at −78 °C. The resulting mixture was stirred for 30 min and was transferred to a reaction flask containing acid **119** via cannula. After stirring for 1 h at −78 ºC, the reaction was allowed to warm to room temperature in 2.5 h and quenched with half-saturated aq. NH_4Cl (200 mL). The organic phase was separated, and the aqueous phase was extracted with ether. The combined organic phase was washed with brine, dried over anhydrous Na_2SO_4, and concentrated. Purification of the residue by flash column chromatography on silica gel (10 – 20% EtOAc in hexane) afforded **121**[169] (6.07 g, 67% yield) as a colorless oil. $[\alpha]^{20}_D$ −46.8 (*c* 1, $CHCl_3$); lit[169] $[\alpha]^{20}_D$ −42.8 (*c* 1, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.43 – 7.10 (m, 10H), 4.66 – 4.56 (m, 1H), 4.51 (s, 2H), 4.16 – 4.03 (m, 2H), 3.59 (t, *J* = 6.2 Hz, 2H), 3.26 (dd, *J* = 13.4, 3.3 Hz, 1H), 3.07 (t, *J* = 7.2 Hz, 2H), 2.70 (dd, *J* = 13.4, 9.6 Hz, 1H), 2.10 – 1.98 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.1, 153.5, 138.5, 135.4, 129.4, 128.9, 128.4, 127.7, 127.6, 127.3, 72.9, 69.2, 66.1, 55.2, 37.9, 32.5, 24.5.

(*R*)-4-Benzyl-3-((*R*)-4-(benzyloxy)-2-methylbutanoyl)oxazolidin-2-one (122).

To a stirred solution of **121** (12.46 g, 35.26 mmol) in dry THF (50 mL) was added NaHMDS (50 mL, 1 M in THF, 50.00 mmol) via cannula dropwise at −78 ºC. The reaction mixture was stirred for 1 h at −78 ºC and methyl iodide (10.97 mL, 176. 30 mmol) was added dropwise. After stirring for 3 h at −78 ºC, the reaction was quenched with acetic acid (2.5 mL), and the resulting mixture was warmed to room temperature and extracted with EtOAc. The so-obtained organic extract was washed with brine, dried over anhydrous Na_2SO_4, and concentrated. Purification was performed via flash column chromatography on silica gel (10 – 15% EtOAc in hexane) to isolated **122**[169] (10.87 g,

84% yield, dr 95:5) as a colorless oil. $[\alpha]^{20}_D$ –61.6 (*c* 1, CHCl₃); lit[169] $[\alpha]^{20}_D$ –81.3 (*c* 1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 7.42 – 7.09 (m, 10H), 4.53 – 4.31 (m, 3H), 4.01 – 3.86 (m, 2H), 3.73 (t, *J* = 8.4, 1H), 3.61 – 3.46 (m, 2H), 3.19 (dd, *J* = 13.4, 3.4 Hz, 1H), 2.71 (dd, *J* = 13.4, 9.6 Hz, 1H), 2.25 – 2.18 (m, 1H), 1.82 – 1.67 (m, 1H), 1.25 (d, *J* = 6.9 Hz, 3H); ¹³C NMR (100 MHz, CDCl₃) δ 177.1, 153.3, 138.5, 135.4, 129.4, 128.8, 128.3, 127.6, 127.5, 127.2, 72.8, 68.5, 65.8, 55.2, 38.0, 35.1, 33.6, 18.1.

(2*R*)-4-Benzyloxy-2-methylbutanol (123).

To an ice-cold solution of **122** (10.50 g, 28.58 mmol) in anhydrous diethyl ether (275 mL) was added dry ethanol (2.5 mL, 42.86 mmol) and lithium borohydride (0.93 g, 42.86 mmol) portion wise. After stirring for 2 h in ice-bath, the reaction was quenched by slow addition of 1 N NaOH (200 mL). The resulting mixture was stirred for 10 min at room temperature and was extracted with ether. The ether extract was washed with saturated aq. NH₄Cl, water, and brine and dried over Na₂SO₄. After concentration, the resulting residue was purified by flash column chromatography (20 – 30% EtOAc in hexane) to afford the desired alcohol **123**[99] (4.99 g, 90% yield) as a colorless oil. $[\alpha]^{20}_D$ +10.5 (*c* 1, CHCl₃); lit[99] $[\alpha]^{20}_D$ +10.4 (*c* 7.5, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 7.45 – 7.18 (m, 5H), 4.52 (s, 2H), 3.66 – 3.32 (m, 4H), 2.64 (br s, 1H), 1.86 – 1.75 (m, 1H), 1.75 – 1.64 (m, 1H), 1.62 – 1.51 (m, 1H), 0.92 (d, *J* = 6.8 Hz, 3H); ¹³C NMR (101 MHz, CDCL₃) δ 138.0, 128.5, 127.8, 127.7, 73.2, 68.7, 68.1, 34.1, 34.0, 17.2.

(2*R*)-4-Benzyloxy-1-bromo-2-methylbutane (113).

To an ice-cold solution of **123** (5 g, 25.74 mmol) in dry dichloromethane was added CBr₄ (8.95 g, 27.03 mmol) and PPh₃ (7.06 g, 27.03 mmol). After stirring for 30 min in ice-bath and 1 h at room temperature, the reaction mixture was concentrated, and the residue

was dropped in the stirring hexane (50 mL). The resulting white suspension was filtered, and the resulting filtrate was concentrated. After purification of the resulting residue by flash column chromatography on silica gel (0 – 5% EtOAc in hexane), compound **113**[99] (6.16 g, 93% yield) was isolated as a colorless oil. $[\alpha]^{20}_D$ –4.5 (*c* 1, CHCl$_3$); lit[99] $[\alpha]^{20}_D$ –4.7 (*c* 0.32, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.43 – 7.21 (m, 5H), 4.51 (s, 2H), 3.56 – 3.49 (m, 2H), 3.44 (dd, *J* = 9.9, 4.6 Hz, 1H), 3.37 (dd, *J* = 9.9, 5.8 Hz, 1H), 2.12 – 1.95 (m, 1H), 1.87 – 1.72 (m, 1H), 1.64 – 1.47 (m, 1H), 1.04 (d, *J* = 6.7 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 138.4, 128.4, 127.62, 127.60, 73.0, 67.8, 41.6, 34.6, 32.1, 18.7.

(3'*S*,4*S*,5'*S*)-3-(7'-Benzyloxy-3',5'-dimethylheptanoyl)-4-phenyloxazolidin-2-one (126).

To a suspension of magnesium (2.25 g, 61.70 mmol) in dry THF (12 mL) was added a solution of **113** (2.66 g, 10.34 mmol) in dry THF (3 mL) at room temperature. A DIBAL solution (1.56 mL, 1 M in THF, 1.56 mmol) was added dropwise and stirred vigorously for 20 min. This solution was added to a chilled suspension of CuBr.DMS (2.04 g, 10.34 mmol) in dry THF (12 mL) at −78 °C. The resulting suspension was warmed slowly to −20 °C, stirred for 25 min, and cooled back again to −78 °C. A solution of (*S*)-*N*-(*trans*-crotonyl)-4-phenyloxazolidin-2-one **125** in dry THF (9 mL) was added dropwise, and the stirring was continued for 2.5 h at −78 °C. Then the reaction mixture was warmed slowly to −30 °C, quenched with saturated aq. NH$_4$Cl, and extracted with ether. This ether extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentration. Purification of the residual oil by flash column chromatography on silica gel (10 – 20% EtOAc in hexane) gave **126**[99] (1.19 g, 80% yield, dr 85:15) as a colorless oil. $[\alpha]^{20}_D$ +29.5 (*c* 3, CHCl$_3$); lit[99] $[\alpha]^{20}_D$ +20.0 (*c* 2.3, CHCl$_3$) for pure **126**; ^{1}H NMR (400 MHz, CDCl$_3$) δ

7.44 – 7.22 (m, 10H), 5.42 (dd, J = 8.7, 3.6 Hz, 1H), 4.67 (t, J = 8.7 Hz, 1H), 4.48 (s, 2H), 4.26 (dd, J = 9.0, 3.9 Hz, 1H), 3.46 (td, J = 6.7, 1.9 Hz, 2H), 2.92 (dd, J = 15.9, 5.4 Hz, 1H), 2.72 (dd, J = 15.9, 8.1 Hz, 1H), 2.17 – 2.02 (m, 1H), 1.71 – 1.50 (m, 2H), 1.46 – 1.35 (m, 1H), 1.18 (ddd, J = 13.5, 9.6, 4.5 Hz, 1H), 1.05 (ddd, J = 13.8, 9.3, 4.8 Hz, 1H), 0.84 (d, J = 6.6 Hz, 3H), 0.81 (d, J = 6.6 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 172.2, 153.7, 139.2, 138.7, 129.1, 128.7, 128.4, 127.6, 127.5, 126.0, 72.9, 69.8, 68.5, 57.6, 44.3, 43.3, 37.4, 27.11, 27.08, 19.2.

(4S,2'S,3'S,5'S)-3-(7'-Benzyloxy-2',3',5'-trimethylheptanoyl)-4-phenyloxazolidin-2-one (127) and (4S,2'R,3'S,5'S)-3-(7'-benzyloxy-2',3',5'-trimethylheptanoyl)-4-phenyloxazolidin-2-one (128).

To a solution of **126** (826 mg, 2.02 mmol) in dry THF (8 mL) was added NaHMDS (2.32 mL, 1 M in THF, 2.32 mmol) dropwise at −78 °C. The reaction mixture was stirred for 1 h at −78 °C and methyl iodide (0.63 mL, 10.10 mmol) was added dropwise. After stirring for 3 h at −78 °C, the reaction was quenched with saturated aq. NH$_4$Cl, and the resulting mixture was warmed to room temperature and extracted with ether. The so-obtained organic extract was dried over anhydrous Na$_2$SO$_4$, and concentrated. Purification was performed via flash column chromatography on silica gel (10 – 15% EtOAc in hexane) to isolate **127**[99] (529 mg, 62% yield) and **128**[99] (172 mg, 20% yield) as colorless oils.

Data for diastereomer **127**; [$α$]$^{20}_D$ +50.2 (c 1.05, CHCl$_3$); lit[99] [$α$]$^{20}_D$ +49.7 (c 1.65, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.43 – 7.23 (m, 10H), 5.40 (dd, J = 8.4, 3.3 Hz, 1H), 4.63 (t, J = 8.7 Hz, 1H), 4.50 (s, 2H), 4.22 (dd, J = 9.0, 3.3 Hz, 1H), 3.71 – 3.64 (m, 1H), 3.53 – 3.47 (m, 2H), 1.96 – 1.82 (m, 1H), 1.72 – 1.43 (m, 3H), 1.22 – 1.06 (m, 2H), 1.03

(d, J = 6.9 Hz, 3H), 0.92 (d, J = 6.6 Hz, 3H), 0.83 (d, J = 6.6 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 176.3, 153.5, 139.4, 138.6, 129.2, 128.6, 128.4, 127.7, 127.5, 125.7, 73.0, 69.7, 68.5, 57.8, 43.2, 39.7, 38.0, 32.8, 27.1, 19.0, 17.9, 13.7.

Data for diastereomer **128**; $[\alpha]^{20}_D$ -18.5 (*c* 1, CHCl$_3$); lit[99] $[\alpha]^{20}_D$ -20.6 (*c* 1.88, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.45 – 7.23 (m, 10H), 5.45 (dd, J = 9.0, 4.5 Hz, 1H), 4.67 (t, J = 9.0 Hz, 1H), 4.49 (s, 2H), 4.28 (dd, J = 9.0, 4.5 Hz, 1H), 3.70 – 3.62 (m, 1H), 3.45 (t, J = 6.6 Hz, 2H), 1.97 – 1.85 (m, 1H), 1.65 – 1.44 (m, 2H), 1.42 – 1.32 (m, 1H), 1.14 (ddd, J = 13.5, 10.5, 3.6 Hz, 1H), 1.03 (d, J = 6.9 Hz, 3H), 0.89 – 0.79 (m, 1H), 0.71 (d, J = 6.3 Hz, 3H), 0.59 (d, J = 6.6 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 176.6, 153.4, 139.2, 138.7, 129.0, 128.7, 128.3, 127.6, 127.5, 126.4, 72.9, 69.5, 68.5, 57.7, 42.9, 42.5, 37.7, 32.9, 27.2, 18.9, 14.3, 11.9.

(4*S*,2'*S*,3'*S*,5'*S*)-3-(7'-Hydroxy-2',3',5'-trimethylheptanoyl)-4-phenyloxazolidin-2-one (129).

To a solution of **127** (1.85 g, 4.37 mmol) in EtOAc (30 mL) was added Pd(OH)$_2$ on carbon (0.37 g). The resultant heterogeneous mixture was stirred under hydrogen atmosphere at 1 atm pressure for 3 h. Then the reaction mixture was filtered to discard catalyst, and the filtrate was concentrated to give **129**[99] (1.46 g, 100% yield) as a colorless oil. $[\alpha]^{20}_D$ +65.2 (*c* 1, CHCl$_3$); lit[99] $[\alpha]^{20}_D$ +68.6 (*c* 2.5, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.41 – 7.25 (m, 5H), 5.42 (dd, J = 8.4, 3.3 Hz, 1H), 4.66 (t, J = 8.7 Hz, 1H), 4.24 (dd, J = 9.0, 3.3 Hz, 1H), 3.74 – 3.61 (m, 3H), 1.93 – 1.81 (m, 1H), 1.70 – 1.57 (m, 1H), 1.56 – 1.33 (m, 3H), 1.27 – 1.05 (m, 2H), 1.03 (d, J = 6.9 Hz, 3H), 0.92 (d, J = 6.6 Hz, 3H), 0.83 (d, J = 6.6 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 176.4, 153.5, 139.3, 129.2, 128.6, 125.7, 69.7, 60.9, 57.8, 43.1, 41.1, 39.8, 32.7, 26.6, 18.8, 17.8, 13.8.

(4S,2'S,3'S,5'S)-3-(7'-Oxo-2',3',5'-trimethylheptanoyl)-4-phenyloxazolidin-2-one (130).

To a solution of oxalyl chloride (337 µL, 3.93 mmol) in dry dichloromethane (4 mL) was added a solution of dry DMSO (361 µL, 5.08 mmol) in dry dichloromethane (4 mL) dropwise at −78 °C, and the resulting mixture was stirred for 15 min. A solution of **129** (770 mg, 2.31 mmol) in dichloromethane (10 mL) was added dropwise followed by stirring for 15 min while maintaining the reaction temperature at −78 °C. A solution of triethylamine (1.45 mL, 10.40 mmol) in dry dichloromethane (5 mL) was added dropwise, and the reaction was stirred in ice-bath (<5 °C) for 50 min and quenched with saturated aq. NH₄Cl. The resulting mixture was extracted with dichloromethane, and the dichloromethane extract was washed with brine, dried over anhydrous Na₂SO₄, and concentrated. After purification of the residual oil by flash column chromatography on silica gel (20 – 25% EtOAc in hexane), the desired aldehyde **130**[99] (686 mg, 90% yield) was isolated as a colorless oil. $[\alpha]^{20}_D$ +70.0 (c 0.45, CHCl₃); lit[99] $[\alpha]^{20}_D$ +69.3 (c 1, CHCl₃); ¹H NMR (600 MHz, CDCl₃) δ 9.73 (t, J = 2.2 Hz, 1H), 7.40 – 7.22 (m, 5H), 5.41 (dd, J = 8.7, 3.3 Hz, 1H), 4.66 (t, J = 8.8 Hz, 1H), 4.24 (dd, J = 8.8, 3.6, 1.1 Hz, 1H), 3.70 – 3.63 (m, 1H), 2.35 – 2.24 (m, 2H), 2.16 – 2.07 (m, 1H), 1.91 – 1.82 (m, 1H), 1.30 – 1.21 (m, 1H), 1.09 (ddd, J = 13.8, 11.0, 3.6 Hz, 1H), 1.02 (d, J = 7.0 Hz, 3H), 0.94 (d, J = 6.7 Hz, 3H), 0.88 (dd, J = 6.6 Hz, 3H); ¹³C NMR (150 MHz, CDCl₃) δ 202.5, 176.1, 153.4, 139.2, 129.2, 128.6, 125.7, 69.7, 57.8, 52.1, 43.0, 39.6, 32.6, 25.3, 18.9, 17.7, 13.8.

(2*R*,3'*S*,5'*S*,6'*S*,4"*S*)-2,*N*-Dimethyl-*N*-[3',5',6'-trimethyl-7'-oxo-7'-(2"-oxo-4"-phenyloxazolidin-3-yl)heptyl]butyramide (133).

To a stirred solution of **130** (686 mg, 2.07 mmol) in dry MeOH (16 mL) were added methylamine hydrochloride (280 mg, 4.14 mmol), methylamine solution (3.11 mL, 2 M in THF, 6.21 mmol), and anhydrous Na_2SO_4 at room temperature. After stirring for 20 min, $NaCNBH_3$ (195 mg, 3.11 mmol) was added by cooling the reaction mixture in ice-bath. The resulting mixture was stirred for 1 h at room temperature and concentrated. The residual oil was dissolved in DMF (20 mL) and cooled in ice-bath. Then, (*R*)-methylbutyric acid (408 mg, 4.00 mmol), HOBt (688 mg, 80% in water, 4.00 mmol), EDCI.HCl (756 mg, 4.00 mmol), and Hunig's base (0.69 mL, 4.00 mmol) were added to this ice-cold solution one after the other, and the resulting mixture was stirred for 10 h at room temperature. The reaction was diluted with EtOAc, washed with 1 M HCl and brine, dried over anhydrous Na_2SO_4, and concentrated. Purification of the residual oil by flash column chromatography on silica gel (20 – 40% EtOAc in hexane) afforded a mixture of rotamers of compound **133**[99] (50% yield in two steps) as a colorless oil. $[\alpha]^{20}_D$ +43.1 (*c* 0.45, CHCl₃); lit[99] $[\alpha]^{20}_D$ +40.0 (*c* 0.5, CHCl₃); ¹H NMR (600 MHz, CDCl₃), both rotamers, δ 7.39 – 7.25 (m, 5H), 5.42 (dt, *J* = 8.6, 3.2 Hz, 1H), 4.66 (t, *J* = 8.8, 1H), 4.24 (td, *J* = 9.1, 3.5 Hz, 1H), 3.72 – 3.63 (m, 1H), 3.43 – 3.20 (m, 2H), 2.99 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.61 – 2.48 (m, 1H), 1.90 – 1.80 (m, 1H), 1.74 – 1.62 (m, 1H), 1.53 – 1.30 (m, 4H), 1.25 – 1.05 (m, 5H), 1.05 – 0.99 (m, 3H), 0.92 – 0.81 (m, 9H); ¹³C NMR (150 MHz, CDCl₃) , both rotamers, δ 176.5, 176.3, 176.2, 176.1, 153.5, 153.4 139.4, 139.2, 129.20, 129.15, 128.7, 128.6, 125.7, 69.72, 69.68, 57.82, 57.79, 48.0, 46.1, 43.1, 39.81, 39.78, 37.43, 37.35, 37.2, 35.5, 35.2, 33.7, 32.8, 32.6, 28.0, 27.4, 27.0, 18.8, 17.8, 17.7, 17.6, 17.1,

14.0, 13.7, 12.1, 12.0; ESIHRMS calculated for $C_{25}H_{39}N_2O_4$ $[M+H]^+$, 431.2910; found, 431.2914.

(2R,3'S,5'S,6'S)-2,N-Dimethyl-N-[7'-hydroxy-3',5',6'-tri-methylheptyl]butyramide (134).

To an ice-cold solution of **133** (442 mg, 1.03 mmol) in dry THF (11 mL) was added lithium borohydride (67 mg, 3.09 mmol) portion wise. Dry methanol (0.21 mL, 5.15 mmol) was added dropwise in 5 min, and the stirring was continued for next 40 min in ice-bath and 10 min at room temperature. The reaction was quenched with water after cooling the reaction mixture in ice-bath, and the resulting mixture was extracted with EtOAc. The so-obtained organic extract was washed with brine, dried over Na_2SO_4, concentrated, and purified by flash column chromatography (10 – 50% EtOAc in hexane) to afford a mixture of rotamers of alcohol **134**[99] (247 mg, 89% yield) as a colorless oil. $[\alpha]^{20}_D$ –40.9 (c 0.55, CHCl$_3$); lit[99] $[\alpha]^{20}_D$ –36.2 (c 0.5, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) , both rotamers, δ 3.64 – 3.55 (m, 1H), 3.50 – 3.21 (m, 3H), 3.00 (s, 3H, rotamer), 2.92 (s, 3H, rotamer), 2.63 – 2.49 (m, 1H), 1.76 – 1.29 (m, 8H), 1.14 – 1.05 (m, 5H), 0.91 – 0.81 (m, 12H); ^{13}C NMR (100 MHz, CDCl$_3$) , both rotamers, δ 176.5, 176.2, 66.0, 65.9, 48.1, 46.3, 41.1, 41.0, 40.1, 39.8, 37.4, 37.2, 35.32, 35.26, 33.7, 31.4, 31.2, 28.4, 28.2, 27.4, 27.0, 19.2, 19.0, 17.8, 17.11, 17.06, 16.9, 13.3, 13.2, 12.2, 12.0.

(2R,3'S,5'S,6'S)-2,N-Dimethyl-N-[8'-methoxy-3',5',6'-tri-methyloct-7-enyl]-butyramide (135).

To a solution of oxalyl chloride (131 μL, 1.50 mmol) in dry dichloromethane (1.5 mL) was added a solution of dry DMSO (142 μL, 2.00 mmol) in dry dichloromethane (1.5 mL) dropwise at −78 °C, and the resulting mixture was stirred for 15 min. A solution of

134 (272 mg, 1.00 mmol) in dichloromethane (3.5 mL) was added dropwise followed by stirring for 15 min while maintaining the reaction temperature at −78 °C. A solution of triethylamine (566 µL, 4.00 mmol) in dry dichloromethane (1.5 mL) was added dropwise, and the reaction was stirred in ice-bath (<5 °C) for 50 min and quenched with saturated aq. NH_4Cl. The resulting mixture was extracted with dichloromethane, and the dichloromethane extract was washed with brine, dried over anhydrous Na_2SO_4, and concentrated to give a corresponding crude aldehyde intermediate.

(Methoxymethyl)triphenylphosphonium chloride (1.72 g, 5.00 mmol) was dried under high vacuum at 60 °C for 3 h and cooled in argon atmosphere. Then, it was charged with dry THF (8 mL) and the resulting suspension was cooled in ice-bath. After adding *n*-BuLi (1.8 mL, 2.5 M in hexane, 4.50 mmol) dropwise to this suspension, the mixture was stirred for 30 min at room temperature followed by cooling to –2.0 °C. A solution of above prepared crude aldehyde in dry THF (3.5 mL) was added dropwise, and the resulting mixture was stirred for 20 min at –2.0 °C and 1 h at room temperature. The reaction was quenched with MeOH (5 mL), diluted with ether, washed with saturated aq. NH_4Cl and brine, dried over anhydrous Na_2SO_4, and concentrated. After purification of the residue by flash chromatography on silica gel (10 – 20% EtOAc in hexane), the desired alkene **135**[99] (mixture of *E* and *Z* isomers, 215 mg, 72% yield in two steps) was isolated as a yellow oil. $[\alpha]^{20}_D$ –20.88 (*c* 0.9, $CHCl_3$); lit[99] $[\alpha]^{20}_D$ –20.04 (*c* 1.6, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) , both *E/Z* isomers and rotamers, δ 6.17 (dd, *J* = 12.6, 5.4 Hz, 1H), 5.82 (dd, *J* = 9.5, 6.3 Hz, 1H), 4.57 (dd, *J* = 12.6, 8.9 Hz, 1H), 4.15 (dd, *J* = 9.9, 6.3 Hz, 1H), 3.51 (d, *J* = 3.8 Hz, 3H), 3.47 (d, *J* = 4.0 Hz, 3H), 3.39 – 3.18 (m, 4H), 2.98 (s, 6H, rotamer), 2.89 (s, 6H, rotamer), 2.60 – 2.44 (m, 3H), 1.98 – 1.85 (m, 1H), 1.74 – 1.58 (m, 2H), 1.55

– 1.20 (m, 10H), 1.16 – 0.98 (m, 10H), 0.97 – 0.80 (m, 18H), 0.79 – 0.70 (m, 6H); ^{13}C NMR (100 MHz, CDCl$_3$) , both *E/Z* isomers and rotamers, δ 176.5, 176.1, 146.7, 146.5, 145.5, 145.4, 110.8, 110.3, 106.4, 106.1, 59.4, 55.93, 55.86, 48.1, 48.0, 46.2, 42.04, 41.98, 37.8, 37.7, 37.3, 37.2, 37.1, 36.9, 35.51, 35.45, 35.3, 35.2, 33.64, 33.58, 33.4, 28.1, 28.0, 27.4, 27.0, 19.3, 19.23, 19.19 19.1, 18.8, 18.7, 17.7, 17.1, 15.4, 15.3, 15.2, 12.1, 12.0.

(2'*R*,3*R*,4*S*,6*S*)-3,4,6-Trimethyl-8-[methyl-2'-(methyl-butyryl)amino]octanoic acid (111).

To a stirred solution of **135** (180 mg, 0.61 mmol) in acetonitrile (6 mL) was added 1 N HCl (6 mL) at room temperature, and the mixture was allowed to stir for 1 h . The reaction was diluted with ether, washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated to give the crude aldehyde. To a solution of the so-obtained crude aldehyde in MeOH (20 mL) was added 2-methyl-2-butene (3.19 mL, 30.50 mmol) followed by a freshly prepared solution of NaClO$_2$ (1.25 M in 20% NaH$_2$PO$_4$ solution, 2.4 mL, 3.05 mmol) at room temperature. The resultant mixture was stirred vigorously for 1 h at room temperature, diluted with ether, washed with brine, dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (0 – 5 % MeOH in 1:1 mixture of EtOAc and hexane) to afford **111**[99] (175 mg, 97% yield) as a yellow oil. [α]^{20}$_D$ –37.57 (*c* 0.7, CHCl$_3$); lit[99] [α]^{20}$_D$ –35.0 (*c* 0.4, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 3.38 (t, *J* = 7.5 Hz, 1H), 3.34 – 3.22 (m, 1H), 3.00 (s, 3H, rotamer), 2.91 (s, 3H, rotamer), 2.61 – 2.50 (m, 1H), 2.34 (dd, *J* = 15.1, 5.0 Hz, 1H), 2.08 (dt, *J* = 14.9, 9.6 Hz, 1H), 2.01 – 1.88 (m, 1H), 1.73 – 1.62 (m, 1H), 1.58 – 1.28 (m, 5H), 1.15 – 1.01 (m, 5H), 0.94 – 0.89 (m, 3H), 0.89 – 0.83 (m, 6H), 0.83 – 0.77

(m, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 178.73, 178.68, 176.7, 176.4, 48.1, 46.2, 40.4, 40.2, 38.0, 37.9, 37.4, 37.2, 37.1, 35.3, 35.2, 35.0, 34.2, 33.8, 28.2, 27.4, 27.0, 19.09, 19.06, 17.7, 17.0, 16.6, 16.5, 16.02, 16.00, 12.1, 12.0; ESIHRMS calculated for C$_{17}$H$_{34}$NO$_3$ [M+H]$^+$, 300.2539; found, 300.2537.

N-((2R)-3-Butenol-2-yl)-[(3R,4S,6S)-trimethyl-8-((2R)-N-methyl-2-methylbutyramido)]octanamide (136).

To a mixture of **111** (175 mg, 0.58 mmol) and **112** (143 mg, 1.16 mmol) in dry CH$_2$Cl$_2$ (5 mL) were added DIPEA (303 μL, 1.74 mmol) and DMAP (35 mg, 0.29 mmol) at room temperature. After stirring for 10 min, EDCI.HCl (334 mg, 1.74 mmol) was added and stirring was continued for 14 h. The reaction was then diluted with water (5 mL), acidified with 1 M HCl (10 mL), and extracted with EtOAc. The so-obtained organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. Purification of the residue by flash column chromatography on silica gel (0 – 5 % MeOH in CH$_2$Cl$_2$) afforded **136**[101] (210 mg, 98%) as a colorless oil: [α]^{20}$_D$ –10.9 (c 1, CHCl$_3$); lit[101] [α]^{20}$_D$ – 10.8 (c 0.8, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) , both rotamers, δ 6.42 (d, J = 7.9 Hz, 1H, rotamer), 6.27 (d, J = 7.9 Hz, 1H, rotamer), 5.90 – 5.75 (m, 1H), 5.27 – 5.12 (m, 2H), 4.54 (br s, 1H), 3.75 – 3.57 (m, 2H), 3.46 – 3.20 (m, 2H), 2.99 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.63 – 2.47 (m, 1H), 2.30 – 2.18 (m, 1H), 2.05 – 1.85 (m, 2H), 1.74 – 1.29 (m, 6H), 1.19 – 0.99 (m, 5H), 0.91 – 0.74 (m, 12H); ^{13}C NMR (100 MHz, CDCl$_3$) , both rotamers, δ 176.7, 176.6, 173.4, 173.1, 135.5, 135.3, 116.5, 116.4, 65.0, 53.6, 53.5, 48.1, 46.1, 40.6, 40.5, 40.4, 40.0, 37.4, 37.2, 37.1, 35.5, 35.4, 35.3, 35.0, 34.3, 34.1, 33.8, 28.2, 28.1, 27.4, 27.0, 19.3, 19.2, 17.8, 17.1, 16.7, 16.5, 16.2, 15.8, 12.2, 12.0; ESIHRMS calculated for C$_{21}$H$_{41}$N$_2$O$_3$ [M+H]$^+$, 369.3117; found, 369.3118.

(4*R*)-4-Ethenyl-2-[(2*R*,3*S*,5*S*)-2,3,5-trimethyl-7-((2*R*)-*N*-methyl-2-methylbutyramido)-heptyl]oxazoline (137).

To a stirred solution of **136** (210 mg, 0.57 mmol) in dry CH_2Cl_2 (14 mL) was added DAST (150 µL, 1.14 mmol) at −78 °C. The reaction was quenched with saturated aq. $NaHCO_3$ after stirring for 10 min and warmed to room temperature followed by extraction with CH_2Cl_2. The organic extract was dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (50 – 75 % EtOAc in hexane) to give **137**[101] (185 mg, 92%) as a colorless oil. $[\alpha]^{20}_D$ +18.4 (*c* 1, $CHCl_3$); lit[101] $[\alpha]^{20}_D$ +18.6 (*c* 0.76, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) , both rotamers, δ 5.78 (ddd, *J* = 17.2, 10.3, 6.9 Hz, 1H), 5.21 (d, *J* = 17.2 Hz, 1H), 5.11 (dd, *J* = 10.3, 3.5 Hz, 1H), 4.60 – 4.50 (m, 1H), 4.37 – 4.25 (m, 1H), 3.89 (td, *J* = 8.1, 4.4 Hz, 1H), 3.41 – 3.18 (m, 2H), 2.98 (s, 3H, rotamer), 2.89 (s, 3H, rotamer), 2.60 – 2.47 (m, 1H), 2.32 – 2.20 (m, 1H), 2.10 – 1.99 (m, 1H), 1.95 – 1.81 (m, 1H), 1.75 – 1.59 (m, 1H), 1.58 – 1.27 (m, 5H), 1.16 – 1.00 (m, 5H), 0.94 – 0.76 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$) , both rotamers, δ 176.4, 176.1, 168.3, 168.2, 138.2, 138.2, 116.2, 116.1, 71.91, 71.87, 68.2, 48.0, 46.1, 40.1, 37.3, 37.19, 37.16, 36.0, 35.8, 35.4, 35.2, 34.23, 34.15, 33.7, 31.83, 31.77, 28.18, 28.15, 27.4, 27.0, 19.0, 17.8, 17.1, 16.31, 16.25, 16.03, 15.98, 12.2, 12.00; ESIHRMS calculated for $C_{21}H_{39}N_2O_2$ [M+H]$^+$, 351.3012; found, 351.3017.

N-((2*R*)-3-Butenol-2-yl)-[(3*R*,4*S*,6*S*)-trimethyl-8-((2*R*)-*N*-methyl-2-methylbutyramido)]octanethioamide (138).

Compound **137** (170 mg, 0.48 mmol) was dissolved in MeOH–Et$_3$N (1:1, 14 mL, saturated with H_2S), and stirred for 16 h at room temperature. The mixture was concentrated under reduced pressure and purified by flash column chromatography on

neutral alumina (10 – 35 % acetone in hexane) to afford **138**[101] (132 mg, 71%) as a colorless oil. $[\alpha]^{20}_D$ +7.6 (*c* 1, CHCl₃); lit[101] $[\alpha]^{20}_D$ +9.6 (*c* 0.44, CHCl₃); ¹H NMR (400 MHz, CDCl₃) , both rotamers, δ 8.20 (d, *J* = 6.6 Hz, 1H, rotamer), 7.99 (d, *J* = 6.6 Hz, 1H, rotamer), 5.87 (ddd, *J* = 16.4, 10.5, 5.2 Hz, 1H), 5.41 – 5.20 (m, 3H), 3.93 – 3.73 (m, 2H), 3.53 – 3.23 (m, 2H), 3.01 (s, 3H, rotamer), 2.91 (s, 3H, rotamer), 2.77 – 2.50 (m, 2H), 2.44 – 2.30 (m, 1H), 2.30 – 2.19 (m, 1H), 1.75 – 1.31 (m, 6H), 1.19 – 1.00 (m, 5H), 0.93 – 0.75 (m, 12H). ¹³C NMR (100 MHz, CDCl₃) , both rotamers, δ 205.6, 205.4, 176.9, 176.8, 133.7, 133.6, 117.5, 117.3, 63.9, 58.6, 58.5, 50.9, 50.7, 48.2, 46.0, 40.6, 39.6, 38.6, 38.5, 37.4, 37.2, 36.9, 35.3, 34.8, 34.3, 33.9, 33.8, 28.2, 28.1, 27.4, 27.0, 19.4, 19.3, 17.8, 17.0, 16.4, 16.2, 15.7, 12.1, 12.0; ESIHRMS calculated for $C_{21}H_{41}N_2O_2S$ [M+H]⁺, 385.2889; found, 385.2896.

(+)-Kalkitoxin (107).

To a stirred solution of **138** (128 mg, 0.33 mmol) in dry CH_2Cl_2 (8 mL) was added DAST (88 µL, 0.67 mmol) at –78 ℃. The reaction was quenched with saturated aq. NaHCO₃ after stirring for 10 min and warmed to room temperature followed by extraction with CH_2Cl_2. The organic extract was dried over anhydrous Na₂SO₄, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina (10 – 30 % EtOAc in hexane) to give kalkitoxin **107**[95] (94 mg, 77%) as a colorless oil. $[\alpha]^{20}_D$ +9.1 (*c* 1, CHCl₃); lit[95] $[\alpha]^{20}_D$ +16.0 (*c* 0.07, CHCl₃); ¹H NMR (400 MHz, CDCl₃) , both rotamers, δ 5.92 (ddd, *J* = 17.1, 10.3, 6.7 Hz, 1H), 5.27 (dt, *J* = 17.1, 1.4 Hz, 1H), 5.20 – 5.06 (m, 1H), 4.99 – 4.82 (m, 1H), 3.47 – 3.33 (m, 2H), 3.32 – 3.21 (m, 1H), 3.09 – 3.01 (m, 1H), 2.99 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.62 – 2.46 (m, 2H), 2.37 – 2.24 (m, 1H), 1.92 – 1.80 (m, 1H), 1.75 – 1.61 (m, 1H), 1.61 – 1.28 (m, 5H), 1.17 – 1.04 (m, 5H),

0.92 – 0.79 (m, 12H); ^{13}C NMR (100 MHz, CDCl$_3$) , both rotamers, δ 176.5, 176.1, 171.6, 171.4, 137.3, 137.2, 116.1, 116.0, 78.57, 78.55, 48.0, 46.1, 40.0, 38.60, 38.57, 38.3, 38.2, 37.41, 37.36, 37.21, 37.18, 35.4, 35.2, 34.2, 34.1, 33.7, 28.24, 28.22, 27.4, 27.0, 19.09, 19.07, 17.7, 17.1, 16.20, 16.16, 16.00, 15.95, 12.1, 12.0; ESIHRMS calculated for C$_{21}$H$_{39}$N$_2$OS [M+H]$^+$, 367.2783; found, 367.2784.

Synthesis of 9-oxa-10-azakalkitoxin (108):

O-**Benzoyl-*N*-methyl-*N*-(2-(1-methyl-3-phenyltriaz-2-en-1-yl)ethyl)hydroxylamine (146).**

To a stirred solution of *N,N'*-dimethylethylenediamine (1.32 g, 15 mmol) in water (100 mL) were sequentially added a solution of benzenediazonium tetrafluoroborate (0.96 g, 5 mmol) in water (30 mL) dropwise in 20 min and a KOH solution (2 M, 5 mL) dropwise in 10 min at room temperature. The reaction was stirred for 5 min and extracted with CH$_2$Cl$_2$. The organic extract was dried over Na$_2$SO$_4$ and concentrated under reduced pressure to afford crude intermediate. This crude intermediate was then dissolved in dry DMF (2 mL) and was added to a stirred suspension of K$_2$HPO$_4$ (1.31 g, 7.50 mmol) and benzoyl peroxide (2.66 g 50 % w/w blended with dicyclohexyl phthalate, 5.50 mmol) in dry DMF (10 mL) at room temperature. After stirring for 1.5 h, the resultant suspension was diluted with water (50 mL), stirred for 1 h, and extracted with EtOAc. The organic extract was then washed with saturated aq. NaHCO$_3$, water, and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification of the residue by flash column chromatography on silica gel (10 – 40 % EtOAc in hexane) afforded **146** (0.82 g, 53% in 2 steps) as a red oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.02 (d, *J* = 7.3 Hz, 2H), 7.57 (t, *J* = 7.4 Hz, 1H), 7.49 – 7.35 (m, 4H), 7.31 (t, *J* = 7.8 Hz, 2H), 7.14 (t, *J* = 7.2

Hz, 1H), 4.00 (t, J = 6.2 Hz, 2H), 3.38 – 3.21 (m, 5H), 2.94 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 165.0, 150.7, 133.3, 129.5, 128.9, 128.8, 128.5, 125.5, 120.6, 58.9, 53.2, 47.4, 35.8; ESIHRMS calculated for C$_{17}$H$_{20}$N$_4$O$_2$Na [M+Na]$^+$, 335.1484; found, 335.1481.

1-((Methyl(2-(1-methyl-3-phenyltriaz-2-en-1-yl)ethyl)amino)oxy)ethyl acetate (148).

To a stirred solution of **146** (125 mg, 0.40 mmol) in dry CH$_2$Cl$_2$ (2.5 mL) were added sequentially, dropwise DIBAL (0.60 mL 1 M in hexane, 0.60 mmol), pyridine (97 µL, 1.20 mmol), a solution of DMAP (97 mg, 0.80 mmol) in dry CH$_2$Cl$_2$ (1 mL), and Ac$_2$O (227 µL, 2.40 mmol) at −78 °C. The reaction mixture was stirred for 12 h (overnight) at −78 °C, warmed slowly to 0 °C in 2.5 h, and quenched by addition of saturated aq. NH$_4$Cl (4 mL) and saturated aq. sodium potassium tartrate (3 mL) at 0 °C. The resultant mixture was warmed to room temperature, stirred vigorously for 1 h, and extracted with CH$_2$Cl$_2$. The organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. The so-obtained crude intermediate **147** was then forwarded to next reaction without further purification.

To a stirred solution of the crude intermediate **147** in dry CH$_2$Cl$_2$ (2.5 mL) were added sequentially, dropwise DIBAL (0.60 mL 1 M in hexane, 0.60 mmol), pyridine (97 µL, 1.20 mmol), a solution of DMAP (97 mg, 0.80 mmol) in dry CH$_2$Cl$_2$ (1 mL), and Ac$_2$O (227 µL, 2.40 mmol) at −78 °C. The reaction mixture was stirred for 12 h (overnight) at −78 °C, warmed slowly to 0 °C in 2.5 h, and quenched by addition of saturated aq. NH$_4$Cl (4 mL) and saturated aq. sodium potassium tartrate (3 mL) at 0 °C. The resultant mixture was warmed to room temperature, stirred vigorously for 1 h, and extracted with CH$_2$Cl$_2$. The organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (10 –

20 % EtOAc in hexane) to give **148** (91 mg, 78%, uncharacterized single diastereomer) as a yellow oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.42 (d, J = 7.4 Hz, 2H), 7.33 (t, J = 7.7 Hz, 2H), 7.14 (t, J = 7.2 Hz, 1H), 6.13 (q, J = 5.4 Hz, 1H), 3.94 (t, J = 6.3 Hz, 2H), 3.28 (br s, 3H), 2.97 (t, J = 6.3 Hz, 2H), 2.68 (s, 3H), 2.06 (s, 3H), 1.36 (d, J = 5.4 Hz, 3H). ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.1, 150.7, 128.8, 125.5, 120.6, 97.2, 58.9, 52.9, 47.0, 36.1, 21.4, 19.1; ESIHRMS calculated for C$_{14}$H$_{22}$N$_4$O$_3$Na [M+Na]$^+$, 317.1590; found, 317.1589.

Benzyl (2-((benzoyloxy)(methyl)amino)ethyl)(methyl)carbamate (150).

To a solution of *N,N'*-dimethylethylenediamine (4.77 mL, 45 mmol) in dry CH$_2$Cl$_2$ (200 mL) was added a solution of benzyl chloroformate (2.14 mL, 15 mmol) in dry CH$_2$Cl$_2$ (20 mL) in 1 h at 0 °C. The mixture was stirred for 3 h at 0 °C followed by 2 h at room temperature. The reaction was quenched by water (200 mL), and the volatile portion was evaporated under reduced pressure. The aqueous residue was acidified to pH 1 by adding 2 M HCl, and the nonpolar impurities were washed out with CH$_2$Cl$_2$. Then the aqueous phase was basified to pH 9 by adding 10% aq. NaOH and extracted with ether. The ether extract was dried over anhydrous Na$_2$SO$_4$ and concentrated under reduced pressure. The so-obtained intermediate was then dissolved in dry DMF (17 mL). To this solution were added K$_2$HPO$_4$ (1.79 g, 10.32 mmol) and benzoyl peroxide (3.34 g 50 % w/w blended with dicyclohexyl phthalate, 6.88 mmol) at room temperature. After stirring for 24 h, the resultant suspension was diluted with water (50 mL), stirred for 1 h, and extracted with EtOAc. The organic extract was washed with saturated aq. NaHCO$_3$, water, and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification of the residue by flash column chromatography on silica gel (10 – 50 % EtOAc in hexane) afforded **150** (1.40 g, 59% in 2 steps) as a yellow oil ^{1}H NMR (400 MHz,

CDCl$_3$), both rotamers, δ 8.18 – 7.89 (m, 2H), 7.56 (t, J = 7.4 Hz, 1H), 7.49 – 7.16 (m, 7H), 5.09 (s, 2H), 3.52 (t, J = 6.6 Hz, 2H), 3.20 (t, J = 6.1 Hz, 2H, 1 rotamer), 3.12 (t, J = 6.6 Hz, 2H, 1 rotamer), 2.97 (s, 3H), 2.94 – 2.86 (m, 3H); ^{13}C NMR (100 MHz, CDCl$_3$), both rotamers, δ 165.1, 165.0, 156.3, 156.0, 136.7, 133.2, 130.0, 129.4, 128.5, 128.4, 128.3, 127.93, 127.87, 127.79, 67.2, 67.1, 58.8, 58.7, 47.4, 47.2, 46.5, 35.5; ESIHRMS calculated for C$_{19}$H$_{22}$N$_2$O$_4$Na [M+Na]$^+$, 365.1477; found, 365.1478.

Benzyl (2-(hydroxy(methyl)amino)ethyl)(methyl)carbamate (151).

To a stirred solution of **150** (500 mg, 1.46 mmol) in dry CH$_2$Cl$_2$ (9 mL) was added DIBAL (2.92 mL 1 M in hexane, 2.92 mmol) in 20 min at –78 ^{o}C. The reaction was stirred for 20 min at –78 ^{o}C and quenched by addition of saturated aq. NH$_4$Cl (15 mL) and saturated aq. sodium potassium tartrate (10 mL). The resultant mixture was warmed to room temperature, stirred vigorously for 1 h, and extracted with ether. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (50 – 100 % EtOAc in hexane) to give **151** (180 mg, 52%) as a colorless oil: ^{1}H NMR (400 MHz, CDCl$_3$), both rotamers, δ 7.49 – 7.19 (m, 5H), 5.14 (s, 2H), 3.48 (br s, 2H), 2.93 (s, 3H), 2.80 – 2.48 (m, 5H); ^{13}C NMR (100 MHz, CDCl$_3$) both rotamers, δ 157.8, 136.7, 128.5, 128.0, 127.7, 67.4, 67.2, 59.4, 58.8, 48.8, 47.9, 46.8, 46.5, 35.1, 34.2; ESIHRMS calculated for C$_{12}$H$_{19}$N$_2$O$_3$ [M+H]$^+$, 239.1396; found, 239.1398.

Alternative synthesis by transesterification: A solution of **150** (1.30 g, 3.80 mmol) in dry MeOH (15 mL) was treated with NaOMe (47 mg, 0.95 mmol) for 2 h at room temperature. The reaction was quenched by stirring with Amberlyst$^®$ 15 hydrogen form (0.13 g) until the pH 7 was obtained. The mixture was filtered through a pad of Celite$^®$,

concentrated under reduced pressure, and purified by flash column chromatography on silica gel (50 – 100 % EtOAc in hexane) to give **151** (0.83 g, 92%) as a colorless oil. The spectral data were identical with above sample.

(*R*)-4-(*tert*-Butyldimethylsilyloxy)-2-methylbutanoic acid (153).

To an ice-cold solution (<5 °C) of **152**[148] (300 mg, 0.77 mmol) in THF-H_2O (1:1, 4 mL) were added LiOH.H_2O (64 mg, 1.54 mmol) and H_2O_2 (0.34 mL, 30 wt% in water, 3.06 mmol). After stirring for 3 h in ice-bath (<5 °C), the reaction was quenched with Na_2SO_3 (6 mL 1.5 M in water, 8.58 mmol). The resultant mixture was diluted with water, and nonpolar impurities were washed out with CH_2Cl_2. The aqueous layer was acidified to pH 6-7 by addition of 1 M HCl, saturated with NaCl, and extracted with ether. The ether extract was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to give the desired acid **153**[148] (168 mg, 94%) as a colorless oil: $[\alpha]^{20}_D$ –13.21 (*c* 8.25, EtOH); lit[148] $[\alpha]^{20}_D$ +13.51 (*c* 1.8, EtOH) and –14.06 for (*S*)-enantiomer of **153**; ^{1}H NMR (400 MHz, CDCl$_3$) δ 3.72 – 3.61 (m, 2H), 2.63 (td, *J* = 7.5, 6.0 Hz, 1H), 2.03 – 1.85 (m, 1H), 1.68 – 1.53 (m, 1H), 1.20 (d, *J* = 7.1 Hz, 3H), 0.88 (s, 9H), 0.04 (s, 6H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 183.1, 60.7, 36.2, 36.0, 25.7, 18.3, 16.9, -5.5.

Benzyl   (*R*)-(3,6,10,10,11,11-hexamethyl-5-oxo-4,9-dioxa-3-aza-10-siladodecyl)(methyl)carbamate (154).

To an ice-cold solution of hydroxylamine **151** (142 mg, 0.59 mmol) and acid **153** (125 mg, 0.54 mmol) in dry CH_2Cl_2 (3.5 mL) were added HOBt (136 mg wetted with 20 wt% water, 0.81 mmol), EDCI.HCl (155 mg, 0.81 mmol), and DIPEA (141 µL, 0.81 mmol), and the resultant mixture was stirred for 4 h at room temperature. The mixture was washed with water followed by brine, dried over anhydrous Na_2SO_4, concentrated under

reduced pressure, and purified by flash column chromatography on silica gel (20 – 30 %

EtOAc in hexane) to give **154** (187 mg, 77%) as a colorless oil: $[\alpha]^{20}_D$ −12.4 (*c* 6.5, CHCl$_3$);

^{1}H NMR (400 MHz, CDCl$_3$), both rotamers, δ 7.43 – 7.14 (m, 5H), 5.10 (s, 2H), 3.71 –

3.50 (m, 2H), 3.50 – 3.35 (m, 2H), 3.03 (t, *J* = 6.3 Hz, 1H), 2.99 – 2.91 (m, 4H), 2.79 –

2.70 (m, 3H), 2.67 – 2.52 (m, 1H), 1.99 – 1.80 (m, 1H), 1.65 – 1.50 (m, 1H), 1.26 – 1.06

(m, 3H), 0.87 (s, 9H), 0.02 (s, 6H); ^{13}C NMR (101 MHz, CDCl$_3$), both rotamers, δ 174.9,

174.8, 156.2, 156.0, 136.7, 128.4, 127.9, 127.8, 67.2, 67.0, 60.5, 58.6, 58.5, 47.1, 46.8,

46.4, 36.2, 35.5, 35.3, 34.8, 25.9, 18.3, 17.2, 17.1, -5.4; ESIHRMS calculated for

C$_{23}$H$_{41}$N$_2$O$_5$Si [M+H]$^+$, 453.2785; found, 453.2780.

(10*R*)-4,7,10,14,14,15,15-Heptamethyl-3-oxo-1-phenyl-2,8,13-trioxa-4,7-diaza-14-

silahexadecan-9-yl acetate (155).

To a chilled solution of **154** (130 mg, 0.29 mmol) at −78 °C in dry CH$_2$Cl$_2$ (2 mL)

were added sequentially, dropwise DIBAL (0.50 mL1 M in hexane, 0.50 mmol), pyridine

(70 µL, 0.87 mmol), a solution of DMAP (71 mg, 0.58 mmol) in dry CH$_2$Cl$_2$ (0.8 mL), and

Ac$_2$O (164 µL, 1.74 mmol). The reaction mixture was stirred for 12 h (overnight) at −78

°C, warmed slowly to 0 °C in 2.5 h, and quenched by addition of saturated aq. NH$_4$Cl (3

mL) and saturated aq. sodium potassium tartrate (2.5 mL) at 0 °C. The resultant mixture

was warmed to room temperature, stirred vigorously for 1 h, and extracted with CH$_2$Cl$_2$.

The organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, concentrated

under reduced pressure, and purified by flash column chromatography on silica gel (20 –

25 % EtOAc in hexane) to give **155** (86 mg, 60%, uncharacterized single diastereomer)

as a colorless oil. $[\alpha]^{20}_D$ +2.7 (*c* 1.5, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$), rotamers, δ 7.41

– 7.21 (m, 5H), 5.99 – 5.85 (m, 1H), 5.12 (s, 2H), 3.74 – 3.52 (m, 2H), 3.52 – 3.36 (m,

2H), 2.95 (s, 3H), 2.91 – 2.72 (m, 2H), 2.66 – 2.53 (m, 3H), 2.11 – 1.99 (m, 3H), 1.91 (br s, 1H), 1.72 (br s, 1H), 1.37 – 1.21 (m, 1H), 0.97 – 0.85 (m, 12H), 0.04 (s, 6H); ^{13}C NMR (100 MHz, CDCl$_3$), rotamers, δ 170.3, 156.2, 156.1, 136.8, 128.4, 127.9, 127.8, 101.1, 67.04, 67.01, 60.9, 60.82, 60.76, 60.70, 58.6, 58.3, 46.8, 46.4, 35.3, 34.8, 34.3, 33.82, 33.77, 33.2, 33.0, 25.92, 25.91, 21.3, 18.29, 18.26, 14.3, 14.2, 13.8, 13.7, -5.3, -5.4; ESIHRMS calculated for C$_{25}$H$_{44}$N$_2$O$_6$SiNa [M+Na]$^+$, 519.2866; found, 519.2852.

Benzyl ((6*R*)-3,5,6,10,10,11,11-heptamethyl-4,9-dioxa-3-aza-10-siladodecyl)(methyl)carbamate (156) and benzyl methyl(2-(methyl(((3*R*)-3-methyltetrahydrofuran-2-yl)oxy)amino)ethyl)carbamate (157).

To a stirred solution of **155** (50 mg, 0.10 mmol) in dry CH$_2$Cl$_2$ (2 mL) were added Me$_2$Zn (0.25 mL 1 M in heptane, 0.25 mmol) and BF$_3$·OEt$_2$ (31 µL, 0.25 mmol) one after the other at –78 °C. The reaction mixture was warmed slowly to 0 °C in 2.5 h, quenched by slow addition of saturated aq. NaHCO$_3$, and extracted with ether. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (10 – 40 % EtOAc in hexane) to give desired product **156** (12 mg, 27%, dr 63:37) as a colorless oil along with **157** (16 mg, 50%, dr 4:1) as a colorless oil.

Data for **156**: [α]^{20}$_D$ +3.3 (*c* 0.6, CHCl$_3$); ^{1}H NMR (500 MHz, toluene-d$_8$, T = 353 K), both diastereomers, δ 7.25 (d, *J* = 7.4 Hz, 2H), 7.13 (t, *J* = 7.4 Hz, 2H), 7.08 – 7.04 (m, 1H), 5.09 (s, 2H), 3.75 – 3.53 (m, 3H), 3.35 (br s, 2H), 2.82 – 2.69 (m, 5H), 2.41 (s, 3H), 1.87 – 1.79 (m, 1H), 1.40 – 1.26 (m, 2H), 1.09 (d, *J* = 6.2 Hz, 1.9H), 1.06 (d, *J* = 6.2 Hz, 1.1H), 0.96 (s, 9H), 0.92 – 0.88 (m, *J* = 6.8, 3.5 Hz, 3H), 0.08 (s, 6H). ^{13}C NMR (150 MHz, CDCl$_3$), rotamers and both diastereomers, δ 156.2, 136.9, 128.4, 127.9, 127.8, 80.8, 80.5,

67.0, 66.9, 61.6, 61.5, 60.0, 58.7, 58.3, 46.9, 46.5, 46.3, 36.24, 36.17, 35.2, 34.7, 34.6, 33.1, 26.0, 25.9, 18.33, 18.29, 15.5, 15.4, 14.8, 14.3, -5.29, -5.32, -5.34; ESIHRMS calculated for $C_{24}H_{44}N_2O_4SiNa$ [M+Na]⁺, 475.2968; found, 475.2975.

Data for **157**: $[\alpha]^{20}_D$ +60.6 (*c* 0.8, CHCl₃); ^1H NMR (500 MHz, toluene-d₈, T = 263 K) δ 7.21 (d, *J* = 7.0 Hz, 2H), 7.13 – 7.00 (m, 3H), 5.13 (d, *J* = 4.8 Hz, 0.8H), 5.06 (s, 2H), 5.00 (br s, 0.2H), 3.83 (dt, *J* = 8.7, 3.0 Hz, 0.8H), 3.78 – 3.67 (m, 0.4H), 3.64 – 3.56 (m, 0.8H), 3.49 (br s, 1H), 3.37 – 3.26 (m, 1H), 2.80 (s, 3H), 2.75 – 2.65 (m, 2H), 2.53 – 2.45 (m, 3H), 1.88 – 1.76 (m, 1H), 1.69 – 1.56 (m, 1H), 1.50 – 1.38 (m, 1H), 0.95 (d, *J* = 6.7 Hz, 2.4H), 0.85 (d, *J* = 7.0 Hz, 0.6H); ^{13}C NMR (100 MHz, CDCl₃) δ 156.3, 136.9, 128.4, 127.9, 127.8, 106.9, 67.1, 66.9, 66.8, 66.7, 58.8, 58.6, 47.4, 46.7, 46.4, 38.2, 37.9, 37.8, 35.2, 34.7, 32.4, 31.2, 29.7, 12.9, 12.8; ESIHRMS calculated for $C_{17}H_{26}N_2O_4Na$ [M+Na]⁺, 345.1790; found, 345.1792.

(*S*)-4-Benzyl-3-((*R,E*)-2-methyl-5-phenylpent-4-enoyl)oxazolidin-2-one (159).

To a stirred solution of diisopropylamine (5.09 mL, 36.31 mmol) in dry THF (42 mL) was added *n*-BuLi (13.20 mLof 2.5 M solution in hexane, 33.01 mmol) dropwise in 10 min at –78 °C. The resultant mixture was stirred in ice-bath (<5 °C) for 20 min and chilled back to –78 °C. A solution of **158** (7.00 g, 30.01 mmol) in dry THF (42 mL) was added dropwise (40 min) and allowed to stir for additional 30 min at –78 °C. Then a solution of cinnamyl bromide (8.87 g, 45.01 mmol) in dry THF (10 mL) was added dropwise (15 min). Stirring was continued for additional 90 min at –78 °C followed by gradually warming to 0 °C in 2.5 h. The reaction was quenched by saturated aq. NH₄Cl solution (50 mL), and the volatile components were evaporated under reduced pressure. The residue was dissolved in CH₂Cl₂, washed with water followed by brine, dried over anhydrous Na₂SO₄,

and concentrated under reduced pressure. Purification was performed by flash column chromatography on silica gel to afford **159**[170] (7.52 g, 72%, dr 95:5) as a white solid (eluent: 0 – 25% EtOAc in hexane): Mp. 89-91 °C; $[\alpha]^{20}_D$ +10.8 (c 1, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.46 – 7.13 (m, 10H), 6.47 (d, J = 15.8 Hz, 1H), 6.25 (dt, J = 15.8, 7.0 Hz, 1H), 4.78 – 4.60 (m, 1H), 4.18 (t, J = 9.0 Hz, 1H), 4.12 (dd, J = 9.0, 2.9 Hz, 1H), 4.03 – 3.91 (m, 1H), 3.26 (dd, J = 13.4, 3.1 Hz, 1H), 2.73 – 2.64 (m, 1H), 2.62 (dd, J = 13.4, 9.8 Hz, 1H), 2.46 – 2.35 (m, 1H), 1.24 (d, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 176.6, 153.2, 137.3, 135.3, 132.5, 129.3, 128.9, 128.5, 127.3, 127.2, 127.0, 126.1, 66.0, 55.3, 38.0, 37.7, 37.5, 16.6; ESIHRMS calculated for C$_{22}$H$_{24}$NO$_3$ [M+H]$^+$, 350.1756; found, 350.1755.

(*R,E*)-2-Methyl-5-phenylpent-4-enoic acid (160).

To an ice-cold solution (<5 °C) of **159** (1g, 2.86 mmol) in THF-H$_2$O (4:1, 10 mL) were added LiOH.H$_2$O (0.48 g, 11.44 mmol) and H$_2$O$_2$ (0.6 mL, 30 wt% in water, 5.72 mmol). After stirring for 1.5 h in ice-bath (<5 °C), the reaction was quenched with Na$_2$SO$_3$ (17 mL 1M in water, 8.58 mmol). The resultant mixture was diluted with water, and nonpolar impurities were washed out with CH$_2$Cl$_2$. The aqueous layer was acidified to pH 2 by addition of 2 M HCl, saturated with NaCl, and extracted with ether. The ether extract was dried over anhydrous Na$_2$SO$_4$ and concentrated under reduced pressure to give the desired acid **160**[149] (0.37 g, 68%) as a colorless oil: $[\alpha]^{20}_D$ −18.0 (c 1, EtOH); lit[149] $[\alpha]^{20}_D$ −20.3 (c 1.04, EtOH) ^{1}H NMR (400 MHz, CDCl$_3$) δ 11.27 (br s, 1H), 7.53 – 7.11 (m, 5H), 6.47 (d, J = 15.7 Hz, 1H), 6.19 (dt, J = 15.7, 7.0 Hz, 1H), 2.75 – 2.50 (m, 2H), 2.44 – 2.33 (m, 1H), 1.26 (d, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 182.7, 137.3, 132.4, 128.5, 127.2, 126.8, 126.1, 39.5, 36.7, 16.4.

Benzyl (*R,E*)-methyl(2-(methyl((2-methyl-5-phenylpent-4-enoyl)oxy)amino)ethyl)carbamate (161).

To an ice-cold solution of hydroxylamine **151** (695 mg, 2.92 mmol) and acid **160** (370 mg, 1.94 mmol) in dry CH_2Cl_2 (12 mL) were added HOBt (491 mg wetted with 20 wt% water, 2.92 mmol), EDCI.HCl (559 mg, 2.92 mmol), and DIPEA (510 µL, 2.91 mmol), and the resultant mixture was stirred for 5 h at room temperature. The mixture was washed with water followed by brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (20 – 50 % EtOAc in hexane) to give **161** (497 mg, 62%) as a colorless oil. $[\alpha]^{20}_D$ –15.0 (*c* 2.5, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$), both rotamers, δ 7.40 – 7.16 (m, 10H), 6.41 (t, *J* = 14.6 Hz, 1H), 6.22 – 6.03 (m, 1H), 5.10 (s, 2H), 3.43 – 3.31 (m, 2H), 3.01 (t, *J* = 5.4 Hz, 1H), 2.92 (t, *J* = 6.9 Hz, 1H), 2.87 (s, 3H), 2.76 – 2.68 (m, 3H), 2.65 – 2.44 (m, 2H), 2.41 – 2.26 (m, 1H), 1.27 – 1.14 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$), both rotamers, δ 174.34, 174.27, 156.2, 155.9, 137.1, 136.8, 132.5, 132.4, 128.50, 128.45, 127.9, 127.8, 127.3, 127.2, 126.7, 126.6, 126.0, 67.1, 67.0, 58.5, 58.4, 47.2, 46.9, 46.4, 38.7, 37.1, 35.4, 35.2, 16.9; ESIHRMS calculated for $C_{24}H_{31}N_2O_4$ [M+H]$^+$, 411.2284; found, 411.2279.

(10*R,E*)-4,7,10-Trimethyl-3-oxo-1,13-diphenyl-2,8-dioxa-4,7-diazatridec-12-en-9-yl acetate (162).

To a chilled solution of **161** (300 mg, 0.73 mmol) at –78 °C in dry CH_2Cl_2 (4.5 mL) were added sequentially, dropwise DIBAL (1.46 mL1 M in hexane, 0.1.46 mmol), pyridine (176 µL, 2.19 mmol), a solution of DMAP (178 mg, 1.46 mmol) in dry CH_2Cl_2 (2 mL), and Ac_2O (414 µL, 4.38 mmol). The reaction mixture was stirred for 12 h (overnight) at –78 °C, warmed slowly to 0 °C in 2.5 h, and quenched by addition of saturated aq. NH_4Cl (7

mL) and saturated aq. sodium potassium tartrate (5 mL) at 0 °C. The resultant mixture was warmed to room temperature, stirred vigorously for 1 h, and extracted with CH_2Cl_2. The organic extract was washed with brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (20 – 30 % EtOAc in hexane) to give **162** (133 mg, 40%) as a colorless oil. $[\alpha]^{20}_D$ +5.8 (*c* 2.5, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$), both rotamers δ 7.42 – 7.10 (m, 10H), 6.46 – 6.31 (m, 1H), 6.24 – 6.05 (m, 1H), 6.04 – 5.90 (m, 1H), 5.13 (s, 2H), 3.47 (t, *J* = 7.0 Hz, 2H), 2.95 (s, 3H), 2.92 – 2.75 (m, 2H), 2.71 – 2.56 (m, 3H), 2.52 – 2.19 (m, 1H), 2.17 – 1.82 (m, 5H), 1.04 – 0.91 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$), both rotamers, δ 170.3, 156.2, 156.1, 137.5, 136.8, 131.7, 131.6, 128.49, 128.45, 128.2, 128.1, 127.9, 127.8, 127.0, 126.0, 100.7, 67.1, 67.0, 58.6, 58.3, 46.8, 46.4, 36.8, 36.6, 35.3, 34.8, 34.6, 21.3, 14.2, 13.8; ESIHRMS calculated for $C_{26}H_{34}N_2O_5Na$ [M+Na]$^+$, 477.2365; found, 477.2363.

Benzyl methyl(2-(methyl(((3*R*,*E*)-3-methyl-6-phenylhex-5-en-2-yl)oxy)amino)ethyl)carbamate (163).

To a stirred solution of **162** (50 mg, 0.11 mmol) in dry CH_2Cl_2 (2 mL) were added Me_2Zn (0.23 mL 1.2 M in toluene, 0.28 mmol) and $BF_3 \cdot OEt_2$ (34 µL, 0.28 mmol) one after the other at –78 °C. The reaction mixture was warmed slowly to 0 °C in 2.5 h, quenched by slow addition of saturated aq. $NaHCO_3$, and extracted with CH_2Cl_2. The organic extract was dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (10 – 20 % EtOAc in hexane) to give desired product **163** (28 mg, 62%, dr 55:45) as a colorless oil. $[\alpha]^{20}_D$ –6.4 (*c* 1.4, $CHCl_3$); ^{1}H NMR (500 MHz, toluene-d_8, T = 363 K) δ 7.31 – 6.92 (m, 10H), 6.37 (d, *J* = 11.6 Hz, 0.45H), 6.34 (d, *J* = 11.1 Hz 0.55H), 6.20 – 6.08 (m, 1H), 5.08 (s, 2H), 3.70 – 3.56 (m, 1H), 3.40

– 3.29 (m, 2H), 2.83 – 2.68 (m, 5H), 2.47 – 2.36 (m, 3.45H), 2.35 – 2.24 (m, 0.55H), 2.04 – 1.93 (m, 1H), 1.88 – 1.79 (m, 0.45H), 1.79 – 1.67 (m, 0.55H), 1.11 (d, J = 6.3 Hz, 1.65H), 1.09 (d, J = 6.3 Hz, 1.35H), 0.95 – 0.88 (m, J = 7.4 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$), rotamers and diastereomers, δ 156.2, 137.8, 136.9, 131.0, 129.8, 129.6, 129.5, 128.5, 128.4, 127.9, 127.8, 126.9, 125.9, 80.2, 80.0, 67.04, 66.98, 58.7, 58.4, 46.9, 46.5, 46.3, 37.1, 36.9, 36.8, 35.5, 35.2, 34.7, 15.8, 15.3, 15.2, 14.4; ESIHRMS calculated for C$_{25}$H$_{34}$N$_2$O$_3$Na [M+Na]$^+$, 433.2467; found, 433.2460.

_O_-((2_S_,3_R_,4_S_,5_R_)-5-(Benzyloxy)-1,2-bis((_tert_-butyldimethylsilyl)oxy)-4-methylhexan-3-yl) 1H-imidazole-1-carbothioate (169).

A mixture of **167**[151] (1.72 g, 3.56 mmol) and 1,1'-thiocarbonyldiimidazole (3.50 g, 19.66 mmol) was stirred in dry THF (8.5 mL) at 60 °C. After 5 h, additional 1,1'-thiocarbonyldiimidazole (1.50 g, 8.43 mmol) was added, and the resultant mixture was allowed to stir for 15 h. The mixture was then cooled to room temperature, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (5 – 15 % EtOAc in hexane) to give **169** (1.80 g, 85%) as a colorless oil. [α]$^{20}_D$ +2.5 (_c_ 1, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) δ 8.26 (s, 1H), 7.54 (s, 1H), 7.27 – 7.18 (m, 5H), 6.97 (s, 1H), 5.96 (dd, J = 3.9, 3.1 Hz 1H), 4.53 (d, J = 11.6 Hz, 1H), 4.32 (d, J = 11.6 Hz, 1H), 4.14 (td, J = 6.4, 3.9 Hz, 1H), 3.66 (dd, J = 10.5, 6.4 Hz, 1H), 3.60 – 3.52 (m, 2H), 2.35 – 2.27 (m, 1H), 1.19 (d, J = 6.2 Hz, 3H), 1.17 (d, J = 7.0 Hz, 3H), 0.86 (s, 9H), 0.84 (s, 9H), 0.02 (s, 3H), 0.01 (s, 3H), 0.00 (s, 3H), -0.06 (s, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 183.8, 138.6, 136.8, 130.5, 128.1, 127.3, 127.2, 117.8, 85.5, 77.2, 73.4, 70.6, 64.9, 38.3, 25.9, 25.7, 18.3, 17.9, 16.7, 10.6, -4.5, -5.1, -5.5, -5.6; ESIHRMS calculated for C$_{30}$H$_{53}$N$_2$O$_4$SSi$_2$ [M+H]$^+$, 593.3265; found, 593.3260.

(2*R*,3*R*,5*R*)-5,6-Bis((*tert*-butyldimethylsilyl)oxy)-3-methylhexan-2-ol (170).

A degassed solution of **169** (1.78 g, 3.00 mmol), Bu$_3$SnH (2.42 mL, 9.00 mmol), and AIBN (49 mg, 0.3 mmol) in dry toluene (75 mL) was stirred for 2.5 h at 85 °C. The reaction was cooled to room temperature, concentrated under reduced pressure, and eluted through a short column of silica gel (5% EtOAc in hexane). The so-obtained intermediate (impure) was stirred with a mixture of 10% Pd/C and Pd(OH)$_2$ (1:1, 1.6 g) in THF–isopropanol (3:1, 60 mL) under hydrogen gas atmosphere (1 atm) for 18 h. After filtering through a pad of Celite®, the resulting filtrate was concentrated and purified by flash column chromatography on silica gel (5 – 10% EtOAc in hexane) to afford **170** (0.57 g, 50% in 2 steps) as a colorless oil. [α]$^{20}_D$ +30.9 (*c* 1, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) δ 3.76 – 3.65 (m, 2H), 3.55 (dd, *J* = 9.9, 5.2 Hz, 1H), 3.36 (dd, *J* = 9.9, 6.8 Hz, 1H), 1.77 – 1.68 (m, 1H), 1.55 (d, *J* = 4.4 Hz, 1H), 1.47 (ddd, *J* = 13.6, 8.9, 3.8 Hz, 1H), 1.40 – 1.33 (ddd, *J* = 13.6, 9.4, 3.2 Hz, 1H), 1.12 (d, *J* = 6.4 Hz, 3H), 0.89 – 0.87 (m, 12H), 0.86 (s, 9H), 0.06 (s, 3H), 0.05 (s, 3H), 0.04 (s, 3H), 0.03 (s, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 71.8, 71.4, 67.9, 37.4, 35.6, 25.9, 25.9, 19.8, 18.3, 18.1, 14.6, -4.0, -4.8, -5.3, -5.4; ESIHRMS calculated for C$_{19}$H$_{45}$O$_3$Si$_2$ [M+H]$^+$, 377.2907; found, 377.2906.

(2*S*,3*R*,5*R*)-5,6-Bis((*tert*-butyldimethylsilyl)oxy)-3-methylhexan-2-yl   4-nitrobenzoate (171).

To a stirred solution of **170** (350 mg, 0.93 mmol), *p*-nitrobenzoic acid (187 mg, 1.12 mmol), and PPh$_3$ (294 mg, 1.12 mmol) in dry THF (4.5 mL) was added a solution of DIAD (227 mg, 1.12 mmol) in dry THF (0.9 mL) at 0 °C. The resultant mixture was stirred for 3 h at room temperature and concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (0 – 5 % EtOAc in hexane) to afford

171 (450 mg, 92%) as a yellow oil. $[\alpha]^{20}_D$ +37.8 (c 1, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.27 (d, J = 8.8 Hz, 2H), 8.19 (d, J = 8.8 Hz, 2H), 5.18 – 5.00 (m, 1H), 3.80 – 3.68 (m, 1H), 3.57 (dd, J = 9.9, 5.1 Hz, 1H), 3.37 (dd, J = 9.9, 6.9 Hz, 1H), 2.12 – 1.96 (m, 1H), 1.62 – 1.51 (m, 1H), 1.47 – 1.38 (m, 1H), 1.32 (d, J = 6.4 Hz, 3H), 0.99 (d, J = 6.8 Hz, 3H), 0.89 (s, 9H), 0.88 (s, 9H), 0.09 – 0.06 (m, 6H), 0.05 (s, 3H), 0.04 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.2, 150.4, 136.3, 130.6, 123.5, 76.9, 70.7, 67.7, 36.8, 33.6, 25.9, 25.9, 18.3, 18.1, 16.7, 15.4, -3.9, -4.8, -5.31, -5.34; ESIHRMS calculated for C$_{26}$H$_{47}$NO$_6$Si$_2$Na [M+Na]$^+$, 548.2840; found, 548.2833.

(2*S*,3*R*,5*R*)-5,6-Bis((tert-butyldimethylsilyl)oxy)-3-methylhexan-2-ol (172).

To a stirred solution of **171** (440 mg, 0.84 mmol) in dry MeOH (3.5 mL) was added NaOMe (90 mg, 1.67 mmol) in portion wise at room temperature. After stirring for 12 h at room temperature, the reaction mixture was concentrated, and the residue was purified by flash column chromatography on silica gel (5 – 50 % EtOAc in CH$_2$Cl$_2$) to give **172** (280 mg, 89%) as a colorless oil. $[\alpha]^{20}_D$ +33.0 (c 2.7, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 3.77 – 3.66 (m, 1H), 3.65 – 3.58 (m, 1H), 3.56 (dd, J = 9.9, 5.2 Hz, 1H), 3.37 (dd, J = 9.9, 6.8 Hz, 1H), 1.77 (s, 1H), 1.74 – 1.62 (m, 1H), 1.51 (ddd, J = 13.7, 8.9, 3.3 Hz, 1H), 1.35 (ddd, J = 13.7, 9.2, 3.1 Hz, 1H), 1.12 (d, J = 6.3 Hz, 3H), 0.92 – 0.88 (m, 12H), 0.87 (s, 9H), 0.07 (s, 6H), 0.05 – 0.03 (m, 6H). ^{13}C NMR (100 MHz, CDCl$_3$) δ 72.3, 71.4, 67.8, 37.3, 36.1, 25.94, 25.89, 19.9, 18.3, 18.1, 15.7, -4.1, -4.7, -5.32, -5.34; ESIHRMS calculated for C$_{19}$H$_{45}$O$_3$Si$_2$ [M+H]$^+$, 377.2907; found, 377.2908.

2-(((2*R*,3*R*,5*R*)-5,6-Bis((tert-butyldimethylsilyl)oxy)-3-methylhexan-2-yl)oxy)isoindoline-1,3-dione (173).

To a stirred solution of **172** (340 mg, 0.90 mmol), *N*-hydroxyphthalimide (177 mg, 1.08 mmol), and PPh$_3$ (286 mg, 1.08 mmol) in dry THF (5 mL) was added a solution of DIAD (220 mg, 1.08 mmol) in dry THF (1 mL) at 0 °C. After stirring for 30 min at 0 °C and 1.5 h at room temperature, the reaction mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (0 – 5 % EtOAc in hexane) to afford **173** (419 mg, 89%) as a yellow oil. [*α*]$^{20}_D$ +19.6 (*c* 1.3, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.85 – 7.77 (m, 2H), 7.76 – 7.68 (m, 2H), 4.41 – 4.19 (m, 1H), 3.85 – 3.73 (m, 1H), 3.59 (dd, *J* = 10.0, 5.4 Hz, 1H), 3.43 (dd, *J* = 10.0, 6.2 Hz, 1H), 2.16 – 2.00 (m, 1H), 1.72 (ddd, *J* = 13.7, 9.4, 3.1 Hz, 1H), 1.53 (ddd, *J* = 13.7, 10.7, 3.0 Hz, 1H), 1.28 (d, *J* = 6.4 Hz, 3H), 1.06 (d, *J* = 6.8 Hz, 3H), 0.89 (s, 9H), 0.85 (s, 9H), 0.08 (s, 3H), 0.07 (s, 3H), 0.06 – 0.04 (m, 6H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 164.3, 134.3, 129.1, 123.3, 88.1, 71.0, 68.1, 37.0, 33.0, 26.0, 25.9, 18.4, 18.1, 15.8, 14.6, -4.0, -4.8, -5.31, -5.31; ESIHRMS calculated for C$_{27}$H$_{51}$N$_2$O$_5$Si$_2$ [M+NH$_4$]$^+$, 539.3337; found, 539.3335.

(*R*)-*N*-Allyl-*N*,2-dimethylbutanamide (174).

To an ice-cold solution (<5 °C) of an acid (*R*)-2-methylbutyric acid **132** (0.52 g, 5.00 mmol) in dry CH$_2$Cl$_2$ (20 mL) were added *N*-allylmethylamine (530 mg, 7.50 mmol), HOBt (80% in water, 1.35 g, 7.50 mmol), EDCI.HCl (1.44 g, 7.50 mmol), and DIPEA (1.31 mL, 7.50 mmol), and the resulting solution was stirred for 16 h at room temperature. The mixture was diluted with CH$_2$Cl$_2$, washed with 1 M HCl followed by saturated aq. NaHCO$_3$, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification of

the residue by flash column chromatography on silica gel (10 – 30 % EtOAc in hexane) afforded **174** (0.68 g, 87%) as a colorless oil. $[\alpha]^{20}_D$ –38.0 (*c* 1, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$), both rotamers, δ 5.82 – 5.63 (m, 1H), 5.25 – 4.97 (m, 2H), 4.10 – 3.80 (m, 2H), 2.94 (s, 3H, 1 rotamer), 2.88 (s, 3H, 1 rotamer), 2.65 – 2.53 (m, 1H, 1 rotamer), 2.53 – 2.41 (m, 1H, 1 rotamer), 1.74 – 1.57 (m, 1H), 1.44 – 1.27 (m, 1H), 1.09 – 1.02 (m, 3H), 0.88 – 0.78 (m, 3H); ^{13}C NMR (100 MHz, CDCl$_3$), both rotamers, δ 177.1, 176.5, 133.5, 133.2, 117.0, 116.5, 52.1, 50.2, 37.5, 37.3, 34.8, 33.9, 27.5, 27.2, 17.9, 17.3, 12.2, 12.1. ESIHRMS calculated for C$_9$H$_{18}$NO [M+H]$^+$, 156.1388; found, 156.1393.

(*R*)-*N*,2-Dimethyl-*N*-(2-oxoethyl)butanamide (166).

Ozone was bubbled in a solution of **174** (189 mg, 1.20 mmol) in MeOH-CH$_2$Cl$_2$ (1:5, 12 mL) until blue color wasn't observed (20 min) at –78 °C. Argon was bubbled to decolorize the solution, and dimethyl sulfide (0.44 mL, 6.00 mmol) was added. The resultant mixture was stirred in an ice-bath (<5 °C) for 1 h and concentrated under reduced pressure (water bath <25 °C). The so-obtained crude aldehyde **166** was then used immediately in next reaction without purification.

Formaldehyde *O*-((2*R*,3*R*,5*R*)-5,6-bis((*tert*-butyldimethylsilyl)oxy)-3-methylhexan-2-yl) oxime (175).

Hydrazine monohydrate (88 µL, 1.76 mmol) was added to a solution of **173** (230 mg, 0.44 mmol) in MeOH-CH$_2$Cl$_2$ (1:1, 7 mL) at room temperature, and the resultant mixture was stirred for 1 h. The reaction mixture was diluted with CH$_2$Cl$_2$, washed with 5% aq. NaHCO$_3$ solution and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. The so-obtained *O*-alkylhydroxylamine intermediate was then treated with paraformaldehyde (110 µL 37% solution, 1.32 mmol) in MeOH:CH$_2$Cl$_2$ (3:2,

3.3 mL) at room temperature. After 1 h, the resultant mixture was concentrated and purified by flash column chromatography on silica gel (0 – 3 % EtOAc in hexane) to give oxime **175** (161 mg, 90%) as a colorless oil. $[\alpha]^{20}_D$ +25.4 (*c* 1.3, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) δ 6.98 (d, *J* = 8.8 Hz, 1H), 6.35 (d, *J* = 8.8, 1H), 4.17 – 4.05 (m, 1H), 3.74 – 3.66 (m, 1H), 3.53 (dd, *J* = 9.9, 5.4 Hz, 1H), 3.36 (dd, *J* = 9.9, 6.3 Hz, 1H), 2.03 – 1.91 (m, 1H), 1.46 (ddd, *J* = 13.6, 9.1, 3.3 Hz, 1H), 1.33 (ddd, *J* = 13.6, 10.5, 3.2 Hz, 1H), 1.15 (d, *J* = 6.5 Hz, 3H), 0.91 – 0.83 (m, 21H), 0.05 (s, 3H), 0.04 (s, 3H), 0.04 – 0.02 (m, 6H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 136.1, 83.3, 71.1, 68.1, 36.6, 32.9, 26.0, 25.9, 18.3, 18.1, 15.8, 15.1, -4.0, -4.8, -5.30, -5.34; ESIHRMS calculated for C$_{20}$H$_{46}$NO$_3$Si$_2$ [M+H]$^+$, 404.3016; found, 404.3014.

(*R*)-*N*-((5*R*,6*R*,8*R*)-8-((*tert*-Butyldimethylsilyl)oxy)-3,5,6,11,11,12,12-heptamethyl-4,10-dioxa-3-aza-11-silatridecyl)-*N*,2-dimethylbutanamide (176).

NaCNBH$_3$ (75 mg, 1.20 mmol) was added carefully to a stirred solution of **175** (161 mg, 0.40 mmol) in a mixture of 0.1 M HCl and EtOH (1:3, 6 mL, pH = 2-3) at room temperature. After stirring for 1 h, the reaction was quenched with saturated aq. NaHCO$_3$ and extracted with ether. The organic extract was washed with brine, dried over anhydrous NaSO$_4$, and concentrated under reduced pressure. The so-obtained crude *N,O*-dialkylhydroxylamine was then treated with a aldehyde **166** (189 mg, 1.20 mmol) in dry MeOH–CH$_2$Cl$_2$ (5:3, 8 mL) at room temperature. After stirring for 2 h, NaCNBH$_3$ (75 mg, 1.20 mmol) and acetic acid (137 μL, 2.40 mmol) were added sequentially, and the mixture was stirred for next 45 min. The reaction was quenched with saturated aq. NaHCO$_3$ and extracted with EtOAc. The Organic extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification of the

residue by flash column chromatography on silica gel (10 – 30 % EtOAc in hexane) afforded **176** (161 mg, 74%) as a colorless oil. $[\alpha]^{20}_D$ +10.8 (*c* 1, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$), both rotamers, δ 3.73 – 3.31 (m, 6H), 3.05 (s, 3H, 1 rotamer), 2.93 (s, 3H, 1 rotamer), 2.85 – 2.68 (m, 2H), 2.65 – 2.50 (m, 4H), 1.90 (s, 1H), 1.74 – 1.62 (m, 1H), 1.49 – 1.26 (m, 3H), 1.12 – 1.02 (m, 6H), 0.90 – 0.82 (m, 24H), 0.06 – 0.01 (m, 12H); ^{13}C NMR (150 MHz, CDCl$_3$), both rotamers, δ 176.7, 176.4, 81.3, 71.2, 71.1, 68.2, 68.1, 59.5, 58.2, 47.4, 46.5, 46.2, 45.7, 37.3, 37.1, 36.6, 36.4, 36.1, 34.1, 32.3, 32.1, 27.3, 27.0, 26.0, 25.93, 25.91, 25.89, 18.3, 18.1, 17.8, 17.0, 15.7, 15.6, 15.1, 14.9, 12.1, 12.0, -4.0, -4.7, -5.30, -5.34; ESIHRMS calculated for C$_{28}$H$_{63}$N$_2$O$_4$Si$_2$ [M+H]$^+$, 547.4326; found, 547.4328.

(3*R*,4*R*)-4-(((2-((*R*)-*N*,2-Dimethylbutanamido)ethyl)(methyl)amino)oxy)-3-methylpentanoic acid (164).

To an ice-cold solution (< 5 °C) of **176** (161 mg, 0.29 mmol) in dry THF (8 mL) was added TBAF (1 M THF solution, 1.16 mL, 1.16 mmol), and the resultant solution was stirred for 1 h at room temperature. The reaction mixture was diluted with EtOAc, washed with water followed by brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. The so-obtained crude diol intermediate was treated with NaIO$_4$ (94 mg, 0.44 mmol) in CH$_2$Cl$_2$–water (4:1, 5 mL) for 1 h in an ice-bath (<5 °C). The reaction mixture was diluted with CH$_2$Cl$_2$, washed with water followed by brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure to give crude aldehyde intermediate. To a stirred solution of this crude aldehyde in MeOH (11 mL) was added 2-methyl-2-butene (1.54 mL. 14.50 mmol) followed by a freshly prepared solution of NaClO$_2$ (1.25 M in 20% NaH$_2$PO$_4$ solution, 1.16 mL, 1.45 mmol) at room temperature. The resultant mixture was stirred vigorously for 1 h at room temperature, diluted with ether,

washed with brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (0 – 5 % MeOH in CH_2Cl_2) to afford **164** (71 mg, 80%) as a colorless oil. $[\alpha]^{20}_D$ –27.4 (*c* 0.35, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$), both rotamers, δ 3.68 – 3.55 (m, 3H, rotamer), 3.55 – 3.40 (m, 3H, rotamer), 3.04 (s, 3H, rotamer), 2.93 (s, 3H, rotamer), 2.87 – 2.69 (m, 2H), 2.62 – 2.45 (m, 5H), 2.28 – 2.18 (m, 1H), 2.15 – 2.04 (m, 1H), 1.72 – 1.59 (m, 1H), 1.44 – 1.31 (m, 1H), 1.10 – 1.01 (m, 6H), 0.95 – 0.88 (m, 3H), 0.88 – 0.81 (m, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) , both rotamers, δ 178.1, 178.0, 177.0, 176.9, 79.3, 79.2, 59.3, 57.8, 47.4, 46.3, 45.8, 45.5, 37.4, 37.2, 36.8, 36.6, 36.0, 34.2, 33.82, 33.76, 27.2, 27.0, 17.6, 16.9, 15.32, 15.25, 12.0, 11.9; ESIHRMS calculated for $C_{15}H_{31}N_2O_4$ [M+H]$^+$, 303.2284; found, 303.2292.

(3*R*,4*R*)-4-(((2-((*R*)-N,2-Dimethylbutanamido)ethyl)(methyl)amino)oxy)-*N*-((*R*)-1-hydroxybut-3-en-2-yl)-3-methylpentanamide (177).

To a mixture of **164** (71 mg, 0.23 mmol), and **112**[147] (58 mg, 0.47 mmol) in dry CH_2Cl_2 (2 mL) were added DIPEA (123 µL, 0.70 mmol) and DMAP (14 mg, 0.12 mmol) at room temperature. After stirring for 10 min, EDCI.HCl (134 mg, 0.70 mmol) was added. The mixture was stirred for 14 h and concentrated under reduced pressure. Purification of the residue by flash column chromatography on silica gel (25 – 50 % EtOAc in hexane) afforded **177** (86 mg, 99%) as a yellow oil. $[\alpha]^{20}_D$ –5.7 (*c* 0.35, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$), both rotamers, δ 6.82 (s, 1H, rotamer), 6.15 (s, 1H, rotamer), 5.87 – 5.79 (m, 1H), 5.37 – 5.05 (m, 2H), 4.61 – 4.49 (m, 1H), 3.89 – 3.26 (m, 6H), 3.05 (s, 3H, rotamer), 2.94 (s, 3H, rotamer), 2.83 – 2.69 (m, 2H), 2.65 – 2.51 (m, 5H, rotamer), 2.41 (dd, *J* = 14.1, 4.5 Hz, 1H, rotamer), 2.30 – 2.19 (m, 1H), 2.07 (dd, *J* = 14.1, 7.3 Hz, 1H, rotamer), 1.98 (dd, *J* = 14.1, 9.6 Hz, 1H, rotamer), 1.73 – 1.59 (m, 1H), 1.46 – 1.34 (m, 1H), 1.11 – 1.02

(m, 6H), 0.90 (d, J = 7.0 Hz, 3H), 0.88 – 0.84 (m, 3H); ^{13}C NMR (150 MHz, CDCl$_3$), both rotamers, δ 177.1, 177.0, 173.2, 172.6, 135.4, 135.2, 116.6, 116.4, 79.4, 79.0, 65.2, 65.1, 59.5, 57.8, 53.8, 53.5, 47.4, 46.4, 46.0, 45.5, 39.7, 39.6, 37.4, 37.1, 36.1, 34.6, 34.4, 34.3, 27.3, 27.0, 17.6, 17.0, 16.3, 15.9, 14.8, 14.6, 12.1, 11.9; ESIHRMS calculated for C$_{19}$H$_{38}$N$_3$O$_4$ [M+H]$^+$, 372.2862; found, 372.2861.

(R)-N,2-Dimethyl-N-(2-(methyl((($2R,3R$)-3-methyl-4-((R)-4-vinyl-4,5-dihydrooxazol-2-yl)butan-2-yl)oxy)amino)ethyl)butanamide (178).

To a stirred solution of **177** (56 mg, 0.15 mmol) in dry CH$_2$Cl$_2$ (5 mL) was added DAST (40 µL, 0.30 mmol) at –78 °C. The reaction was quenched with saturated aq. NaHCO$_3$ after stirring for 10 min and warmed to room temperature followed by extraction with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina (25 – 50 % EtOAc in hexane) to give **178** (42 mg, 79%) as a colorless oil. [$α$]$^{20}_D$ +26.7 (c 0.27, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 5.79 (ddd, J = 17.2, 10.2, 7.1 Hz, 1H), 5.26 – 5.17 (m, 1H), 5.12 (dd, J = 10.2, 4.6 Hz, 1H), 4.57 (dt, J = 14.8, 7.1 Hz, 1H), 4.38 – 4.27 (m, 1H), 3.94 – 3.87 (m, 1H), 3.68 – 3.34 (m, 3H), 3.05 (s, 3H, rotamer), 2.93 (s, 3H, rotamer), 2.87 – 2.70 (m, 2H), 2.60 – 2.51 (m, 4H), 2.49 – 2.41 (m, 1H), 2.21 – 2.14 (m, 1H), 2.14 – 2.04 (m, 1H), 1.72 – 1.60 (m, 1H), 1.45 – 1.31 (m, 1H), 1.11 – 1.04 (m, 6H), 0.94 – 0.89 (m, 3H), 0.88 – 0.83 (m, 3H); ^{13}C NMR (150 MHz, CDCl$_3$), both rotamers, δ 176.7, 176.4, 168.1, 167.8, 138.2, 138.1, 116.2, 116.1, 79.4, 72.0, 71.9, 68.2, 59.4, 58.1, 47.4, 46.4, 46.0, 45.6, 37.3, 37.1, 36.0, 34.6, 34.5, 34.1, 30.5, 30.4, 27.3, 27.0, 17.7, 17.0, 15.4, 15.3, 15.1, 15.0, 12.1, 12.0; ESIHRMS calculated for C$_{19}$H$_{36}$N$_3$O$_3$ [M+H]$^+$, 354.2757; found, 354.2760.

(*R*)-*N*-(2-(((((2*R*,3*R*)-5-(((*R*)-1-Hydroxybut-3-en-2-yl)amino)-3-methyl-5-thioxopentan-2-yl)oxy)(methyl)amino)ethyl)-*N*,2-dimethylbutanamide (179).

Compound **178** (34 mg, 0.01 mmol) was dissolved in MeOH–Et_3N (1:1, 3 mL, saturated with H_2S), and stirred for 16 h at room temperature. The mixture was concentrated under reduced pressure and purified by flash column chromatography on neutral alumina (25 – 50 % acetone in hexane) to afford **179** (29 mg, 78%) as a colorless oil. $[\alpha]^{20}_D$ +28.8 (*c* 1, $CHCl_3$); ^1H NMR (600 MHz, $CDCl_3$), both rotamers, δ 8.81 (br s, 1H, rotamer), 8.15 (br s, 1H, rotamer), 5.86 (ddd, *J* = 17.3, 10.5, 5.4 Hz, 1H), 5.35 – 5.27 (m, 1H), 5.27 (d, *J* = 17.3 Hz, 1H), 5.24 (d, *J* = 10.5 Hz, 1H), 3.91 – 3.60 (m, 4H), 3.57 – 3.44 (m, 1H, rotamer), 3.40 – 3.30 (m, 1H, rotamer), 3.05 (s, 3H, rotamer), 2.96 (s, 3H, rotamer), 2.92 (dd, *J* = 13.6, 7.4 Hz, 1H), 2.85 – 2.75 (m, 2H), 2.66 – 2.55 (m, 5H), 2.48 – 2.37 (m, 1H), 1.70 – 1.57 (m, 1H), 1.47 – 1.34 (m, 1H), 1.13 – 1.08 (m, 3H), 1.06 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 7.1 Hz, 3H), 0.88 – 0.83 (m, 3H); ^{13}C NMR (150 MHz, $CDCl_3$), major rotamer only, δ 205.1, 177.3, 133.7, 117.2, 78.9, 64.0, 58.9, 57.7, 50.1, 46.6, 45.5, 38.2, 37.4, 36.2, 27.0, 17.1, 17.0, 13.8, 11.9; ESIHRMS calculated for $C_{19}H_{38}N_3O_3S$ [M+H]+, 388.2634; found, 388.2636.

9-Oxa-10-azakalkitoxin (109).

To a stirred solution of **179** (25 mg, 0.07 mmol) in dry CH_2Cl_2 (2.2 mL) was added DAST (17 µL, 0.13 mmol) at −78 °C. The reaction was quenched with saturated aq. $NaHCO_3$ after stirring for 10 min and warmed to room temperature followed by extraction with CH_2Cl_2. The organic extract was dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina (25 % EtOAc in hexane) to give **109** (22 mg, 92%) as a colorless oil. $[\alpha]^{20}_D$ +10.0 (*c* 0.35,

CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 5.92 (ddd, *J* = 17.1, 10.2, 6.8 Hz, 1H), 5.27 (d, *J* = 17.1, 1H), 5.18 – 5.11 (m, 1H), 4.91 (dt, *J* = 14.5, 7.5 Hz, 1H), 3.69 – 3.49 (m, 2H), 3.49 – 3.37 (m, 2H), 3.09 – 3.01 (m, 4H, rotamer), 2.93 (s, 3H, rotamer), 2.87 – 2.71 (m, 2H), 2.70 – 2.62 (m, 1H), 2.62 – 2.51 (m, 4H), 2.40 – 2.31 (m, 1H), 2.20 – 2.12 (m, 1H), 1.74 – 1.62 (m, 1H), 1.44 – 1.33 (m, 1H), 1.12 – 1.02 (m, 6H), 0.91 (d, *J* = 6.9 Hz, 3H, rotamer), 0.89 (d, *J* = 6.9 Hz, 3H, rotamer), 0.88 – 0.84 (m, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 176.7, 176.4, 171.2, 170.8, 137.3, 137.2, 116.1, 116.0, 79.4, 78.6, 78.5, 59.4, 58.1, 47.4, 46.4, 46.1, 45.6, 38.62, 38.61, 37.3, 37.1, 36.8, 36.7, 36.0, 35.9, 35.8, 34.1, 27.3, 27.0, 17.8, 17.0, 15.5, 15.4, 14.9, 14.8, 12.1, 12.0; ESIHRMS calculated for C$_{19}$H$_{36}$N$_3$O$_2$S [M+H]$^+$, 370.2528; found, 370.2541.

Synthesis of 7,8-diazakalkitoxin (110):

(*R,E*)-2-Methyl-5-phenylpent-4-en-1-ol (187).

To a heterogeneous mixture of **159** (7.40 g, 21.18 mmol) and anhydrous diethyl ether (200 mL) were added EtOH (1.85 mL, 31.77 mmol) and LiBH$_4$ (0.69 g, 31.77 mmol) at 0 ºC. After stirring for 2 h at 0 ºC and 2 h at room temperature, the reaction was quenched by careful slow addition of 1M NaOH (180 mL) at 0 ºC followed by stirring for 15 min at room temperature. The resultant mixture was extracted with ether and the ether extract was washed with saturated aq. NH$_4$Cl, water, and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification was performed by flash column chromatography on silica gel to afford **187**[157] (3.33 g, 89%) as a yellow oil (eluent: dichloromethane). [α]^{20}$_D$ +5.4 (*c* 2.5, CH$_2$Cl$_2$); lit[157] [α]$_D$ +6.1 (*c* 1.7, CH$_2$Cl$_2$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.42 – 7.16 (m, 5H), 6.42 (d, *J* = 15.8 Hz, 1H), 6.23 (dt, *J* = 15.8, 7.3 Hz, 1H), 3.57 (dd, *J* = 10.6, 6.2 Hz, 1H), 3.51 (dd, *J* = 10.6, 6.1 Hz, 1H), 2.40 – 2.28 (m, 1H),

2.17 – 2.05 (m, 1H), 1.89 – 1.76 (m, 1H), 1.58 (s, 1H), 0.98 (d, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 137.6, 131.4, 128.7, 128.5, 127.0, 126.0, 68.0, 36.9, 36.1, 16.5.

(*R,E*)-2-Methyl-5-phenylpent-4-en-1-al (184).

To an ice-cold solution of **187** (1g, 5.67 mmol) in dry CH$_2$Cl$_2$ (28 mL) was added Dess-Martin periodinane (4.81 g, 11.35 mmol) portion wise, and the reaction was stirred for 4 h at room temperature. The reaction was diluted with ether (30 mL), quenched with a mixture of water, saturated aq. Na$_2$S$_2$O$_3$, and saturated aq. NaHCO$_3$ (1:1:1, 50 mL), and stirred for 1 h at room temperature. The resultant mixture was extracted with ether, and the ether extract was washed with brine, dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel to afford the desired aldehyde **184**[156] (0.74 g, 75%) as a colorless oil (eluent: 5 – 15% ether in hexane). This material was immediately forwarded to next reaction. [α]^{20}$_D$ –7.12 (*c* 2.5, CH$_2$Cl$_2$); lit[157] [α]$_D$ –6.07 (*c* 2.39, CCl$_4$) ^{1}H NMR (400 MHz, CDCl$_3$) δ 9.72 (d, J = 1.4 Hz, 1H), 7.40 – 7.18 (m, 5H), 6.46 (d, J = 15.8 Hz, 1H), 6.16 (dt, J = 15.8, 7.2 Hz, 1H), 2.71 – 2.59 (m, 1H), 2.59 – 2.48 (m, 1H), 2.38 – 2.25 (m, 1H), 1.17 (d, J = 7.0 Hz, 3H). ^{13}C NMR (100 MHz, CDCl$_3$) δ 204.5, 137.1, 132.5, 128.5, 127.3, 126.6, 126.1, 46.3, 34.0, 13.2.

Benzyl *N*-methyl-*N*-(methylamino)glycinate (183).

To a stirred suspension of *N,N'*-dimethylhydrazine dihydrochloride (1 g, 7.5 mmol) and NaHCO$_3$ (1.89 g, 22.50 mmol) in dry DMF-PhMe (1:3, 10 mL) was added benzyl bromoacetate[158] (1.72 g, 7.5 mmol) dropwise in 1 h at room temperature. The reaction mixture was stirred for 1 h at 85 °C and cooled to room temperature. Then the mixture was diluted with toluene, washed with water followed by brine, dried over anhydrous

Na$_2$SO$_4$, and concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (50 – 100% EtOAc in hexane) to give the desired product **183** (1.04 g, 67%) as a colorless oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.43 – 7.23 (m, 5H), 5.15 (s, 2H), 3.52 (s, 2H), 2.95 (s, 1H), 2.61 (s, 3H), 2.53 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.4, 135.6, 128.5, 128.3, 128.3, 66.2, 59.5, 42.3, 35.5; ESIHRMS calculated for C$_{11}$H$_{17}$N$_2$O$_2$ [M+H]$^+$, 209.1290; found, 209.1289.

Benzyl (*R,E*)-*N*-methyl-*N*-(methyl(2-methyl-5-phenylpent-4-en-1-yl)amino)glycinate (188).

Hydrazine **183** (800 mg, 3.85 mmol) was treated with aldehyde **184** (737 mg, 4.23 mmol) in MeOH-CH$_2$Cl$_2$ (1:1, 21 mL) at room temperature. After stirring for 1 h, NaCNBH$_3$ (724 mg, 11.55 mmol) was added portion wise followed by stirring for 3 h. The reaction was quenched with 1:1 mixture of saturated aq. NaHCO$_3$ and brine (50 mL) and extracted with CH$_2$Cl$_2$. The resultant organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel to afford the desired compound **188** (872 mg, 62%) as a colorless oil (eluent: 0 – 25% EtOAc in hexane). [α]^{20}$_D$ +3.0 (*c* 4.5, CH$_2$Cl$_2$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.43 – 7.25 (m, 9H), 7.23 – 7.15 (m, 1H), 6.37 (d, *J* = 15.8 Hz, 1H), 6.21 (dt, *J* = 15.8, 7.2 Hz, 1H), 5.17 (s, 2H), 3.39 (s, 2H), 2.45 – 2.24 (m, 9H), 2.03 – 1.92 (m, 1H), 1.88 – 1.77 (m, 1H), 0.90 (d, *J* = 6.6 Hz, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.6, 137.9, 135.9, 131.0, 129.5, 128.5, 128.5, 128.3, 128.2, 126.7, 125.9, 66.4, 60.0, 57.3, 38.4, 35.8, 34.3, 31.3, 18.0; ESIHRMS calculated for C$_{23}$H$_{30}$N$_2$O$_2$Na [M+Na]$^+$, 389.2205; found, 389.2204.

Benzyl (*R*)-*N*-methyl-*N*-(methyl(2-methyl-4-oxobutyl)amino)glycinate (189).

To an ice-cold solution of **188** (470 mg, 1.28 mmol) in acetone-water (3:2, 10 mL) was added *N*-methylmorpholine-*N*-oxide (451 mg, 3.84 mmol) and OsO_4 (12.8 mL, 4 µM in PhMe, 0.05 mmol). The reaction flask was rapped with aluminum foil and allowed to stir for 16 h at room temperature. Na_2SO_3 (807 mg, 6.40 mmol) was added and stirred for 5 min followed by concentration under reduced pressure. The resultant residue was dissolved in ethyl acetate, washed with water followed by brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The so-obtained diol residue was treated with $NaIO_4$ (383 mg, 1.80 mmol) in CH_2Cl_2-water (4:1, 10 mL) at 0 °C for 1.5 h. The reaction was diluted with CH_2Cl_2, washed with water followed by brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. Purification was performed by flash column chromatography on silica gel to afford **189** (234 mg, 62%) as a yellow oil (eluent: 10 – 40% EtOAc in hexane). $[\alpha]^{20}_D$ +9.0 (*c* 1.25, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$) δ 9.64 (s, 1H), 7.44 – 7.19 (m, 5H), 5.14 (s, 2H), 3.34 (s, 2H), 2.48 – 2.26 (m, 7H), 2.23 (s, 3H), 2.08 (dd, *J* = 15.7, 4.4 Hz, 1H), 0.90 (d, *J* = 6.0 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 202.9, 170.5, 135.8, 128.5, 128.4, 128.3, 66.4, 60.6, 56.9, 49.4, 35.1, 34.5, 26.9, 18.6; ESIHRMS calculated for $C_{16}H_{25}N_2O_3$ [M+H]$^+$, 293.1865; found, 293.1861.

Benzyl (*R*)-*N*-methyl-*N*-(methyl(2-methyl-4-(methylamino)butyl)amino)glycinate (190).

$MeNH_2$.HCl (11 mg, 0.16 mmol) and $MeNH_2$ (0.13 ml of 2 M in THF, 0.25 mmol) were added sequentially to a stirred mixture of **189** (25 mg, 0.08 mmol), anhydrous Na_2SO_4 (50 mg), and dry MeOH (0.6 mL) at room temperature. After stirring for 30 min at

room temperature, the reaction was cooled in an ice-bath and $NaCNBH_3$ (21 mg, 0.33 mmol) was added. The stirring was continued for 45 min at room temperature and the mixture was concentrated under reduced pressure. Purification was performed by flash column chromatography on silica gel to afford **190** (18 mg, 72%) as a yellow oil (eluent: 0 – 30 % MeOH in CH_2Cl_2). $[\alpha]^{20}_D$ +17.1 (c 1.4, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.43 – 7.27 (m, 5H), 5.16 (s, 2H), 3.43 – 3.30 (m, 2H), 3.11 – 2.99 (m, 1H), 2.90 (dt, J = 12.5, 7.2 Hz, 1H), 2.61 (s, 3H), 2.40 – 2.30 (m, 5H), 2.26 (s, 3H), 1.95 – 1.81 (m, 2H), 1.73 – 1.50 (m, 1H), 0.90 (d, J = 6.5 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.1, 135.5, 128.6, 128.4, 128.3, 66.8, 60.8, 56.2, 47.8, 34.8, 34.6, 32.8, 31.4, 29.0, 19.4. ESIHRMS calculated for $C_{17}H_{30}N_3O_2$ [M+H]$^+$, 308.2338; found, 308.2343.

Benzyl   N-(((R)-4-((R)-N,2-dimethylbutanamido)-2-methylbutyl)(methyl)amino)-N-methylglycinate (191).

To an ice-cold solution (<5 °C) of amine **190** (170 mg, 0.55 mmol) and (R)-2-methylbutanoic acid (113 mg, 1.11 mmol) in DMF (3 mL) were added HOBt (187 mg wetted with 20 wt.% H_2O, 1.11 mmol), EDCI.HCl (213 mg, 1.11 mmol), and DIPEA (193 µL, 1.11 mmol), and the reaction mixture was stirred for 12 h at room temperature. Then the reaction was diluted with EtOAc, washed with water followed by brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on silica gel to give **191** (145 mg, 67%) as a yellow oil (eluent: 20 – 80% ethyl acetate in hexane). $[\alpha]^{20}_D$ –10.4 (c 6.15, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$) , both rotamers, δ 7.44 – 7.27 (m, 5H), 5.13 (s, 2H), 3.47 – 3.08 (m, 4H), 2.95 (s, 3H, rotamer), 2.88 (s, 3H, rotamer), 2.61 – 2.48 (m, 1H), 2.40 – 2.15 (m, 7H), 1.87 – 1.49 (m, 4H), 1.43 – 1.32 (m, 1H), 1.24 – 1.14 (m, 1H), 1.12 – 1.02 (m, 3H), 0.91 – 0.83 (m, 6H); ^{13}C NMR

(150 MHz, CDCl$_3$) , both rotamers, δ 176.5, 176.1, 170.5, 170.5, 135.9, 135.8, 128.5, 128.5, 128.22, 128.15, 66.3, 66.2, 61.1, 60.3, 57.0, 56.9, 48.0, 45.9, 37.3, 37.1, 35.9, 35.3, 35.1, 34.5, 34.4, 34.0, 33.6, 32.2, 32.1, 28.9, 28.8, 27.4, 27.0, 18.2, 18.1, 17.8, 17.10, 17.05, 12.1, 12.0; ESIHRMS calculated for C$_{22}$H$_{38}$N$_3$O$_3$ [M+H]$^+$, 392.2913; found, 392.2917.

***tert*-Butyl** **(*R*)-(1-(methoxy(methyl)amino)-1-oxo-3-((2,4,5-trimethoxybenzyl)thio)propan-2-yl)carbamate (193).**

To a stirred solution of **192**[160] (7.90 g, 19.02 mmol) in dioxane-water (1:1, 38 mL) were added Et$_3$N (6.63 mL, 47.55 mmol) and Boc$_2$O (5.19 g, 23.78 mmol) in 5 min time interval at room temperature. After stirring for 30 min at room temperature, volatile components were evaporated in a rotary evaporator. The residue was diluted with water (50 mL) and the non-polar impurities were washed out with ether and discarded. The resultant aqueous phase was cooled in an ice-bath, acidified with half saturated citric acid to pH 4, and extracted with CH$_2$Cl$_2$. The CH$_2$Cl$_2$ extract was dried over anhydrous Na$_2$SO$_4$ and concentrated under reduced pressure. The so-obtained crude acid intermediate was

Dissolved in dry THF (380 mL) and cooled in an ice-bath. To this ice-cold solution were added HOBt (4.48 g wet in 14 wt% water, 28.53 mmol) and EDCI.HCl (5.47 g, 28.53 mmol). After stirring for 15 min at room temperature, *N,O*-dimethylhydroxylamine (2.78 g, 28.53 mmol) and DIPEA (4.97 mL, 28.53 mmol) were added. The stirring was continued for 20 h and concentrated under reduced pressure. The residue was dissolved in EtOAc, washed with water, saturated aq. NaHCO$_3$, and brine, dried over Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on silica gel (10 – 50 % EtOAc in hexane) to afford desired product **193** (6.23 g, 74%) as a colorless foam:

$[\alpha]^{20}_D$ +63.3 (c 2.3, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 6.11 (s, 2H), 5.46 (d, J = 8.1 Hz, 1H), 4.91 (br s, 1H), 3.84 (d, J = 12.8 Hz, 1H), 3.81 (s, 6H), 3.79 (s, 3H), 3.78 (s, 3H), 3.71 (d, J = 12.8 Hz, 1H), 3.20 (s, 3H), 2.84 (dd, J = 13.9, 5.9 Hz, 1H), 2.73 (dd, J = 13.9, 7.4 Hz, 1H), 1.43 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 172.0, 160.3, 158.7, 155.3, 107.7, 90.6, 79.4, 61.6, 55.7, 55.3, 50.8, 33.7, 32.1, 28.3, 24.6; ESIHRMS calculated for C$_{20}$H$_{32}$N$_2$O$_7$SNa [M+Na]$^+$, 467.1828; found, 467.1825.

tert-Butyl (R)-(1-oxo-3-((2,4,5-trimethoxybenzyl)thio)propan-2-yl)carbamate (194).

To a stirred solution of **193** (5.45 g, 12.26 mmol) in an anhydrous diethyl ether (55 mL) was added LiAlH$_4$ (0.70 g, 18.39 mmol) portion wise at 0 °C, and the mixture was stirred for 40 min. The reaction was diluted with diethyl ether (55 mL) and quenched by addition of ice-cold 1 M HCl (100 mL). The organic layer was separated, and the aqueous layer was extracted with diethyl ether. The combined organic extract was washed with 1 M HCl followed by brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. Purification by flash column chromatography on silica gel (10 – 50% EtOAc in hexane) afforded the desired product **194** (4.37 g, 92%) as a white solid. M.p. 90 – 93 °C $[\alpha]^{20}_D$ −16.97 (c 1.65, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 9.57 (s, 1H), 6.12 (s, 2H), 5.48 (br s, 1H), 4.44 – 4.22 (m, 1H), 3.90 – 3.63 (m, 11H), 3.00 – 2.75 (m, 2H), 1.45 (s, 9H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 199.8, 160.6, 158.8, 155.7, 107.0, 90.6, 80.2, 59.3, 55.7, 55.3, 31.0, 28.3, 24.4; ESIHRMS calculated for C$_{18}$H$_{27}$NO$_6$SNa [M+Na]$^+$, 385.1559; found, 385.1565.

tert-Butyl (R)-(1-((2,4,5-trimethoxybenzyl)thio)but-3-en-2-yl)carbamate (195).

To a stirred suspension of methyltriphenylphosphonium bromide (8.73 g, 24.43 mmol) in dry THF (100 mL) was added KHMDS (44.40 mL 0.5 M in toluene, 22.20 mmol)

slowly, and the mixture was stirred for 1 h at room temperature. The reaction flask was cooled to –78 °C, and a solution of **194** (4.28 g, 11.10 mmol) in dry THF (50 mL) was added drop wise in 30 min. The cooling bath was removed, and the reaction was stirred for 1 h. Then the reaction mixture was diluted with ether (150 mL), washed with half saturated aq. sodium potassium tartrate followed by brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. Purification by flash column chromatography on silica gel (5 – 25% EtOAc in hexane) afforded the desired product **195** (3.15 g, 74%) as a white solid. M.p. 79 – 81 °C; $[\alpha]^{20}_D$ +12.7 (*c* 2, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 6.12 (s, 2H), 5.80 (ddd, *J* = 17.1, 10.4, 5.3 Hz, 1H), 5.18 (dt, *J* = 17.1, 1.4 Hz, 1H), 5.10 (dt, *J* = 10.4, 1.4 Hz, 1H), 5.00 (br s, 1H), 4.38 (br s, 1H), 3.82 (s, 6H), 3.80 – 3.78 (m, 4H), 3.74 (d, *J* = 12.6 Hz, 1H), 2.70 (dd, *J* = 13.8, 5.4 Hz, 1H), 2.64 (dd, *J* = 13.8, 6.4 Hz, 1H), 1.44 (s, 9H); ¹³C NMR (100 MHz, CDCl₃) δ 160.3, 158.8, 155.4, 138.1, 114.9, 107.7, 90.6, 79.3, 55.7, 55.3, 52.3, 36.8, 28.4, 24.6; ESIHRMS calculated for $C_{19}H_{29}NO_5SNa$ [M+Na]⁺, 406.1664; found, 406.1674.

(*R*)-1-((2,4,5-Trimethoxybenzyl)thio)but-3-en-2-amine (196).

To a stirred solution of **195** (1g, 2.61 mmol) in dry CH_2Cl_2 (24 mL) was added trifluoroacetic acid (24 mL) drop wise at 0 °C. After stirring for 30 min, the reaction mixture was concentrated under reduced pressure. The residue was dissolved in CH_2Cl_2 (50 mL) and stirred with 1 N NaOH (50 mL) for 15 min at room temperature. The organic layer was separated, and the aqueous layer was washed extracted with CH_2Cl_2. Combined organic extracts was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (0 – 10% MeOH in CH_2Cl_2) to give the desired product **196** (0.59 g, 80%) as a

colorless oil. $[\alpha]^{20}_D$ –51.5 (c 3.5, CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ 6.11 (s, 2H), 5.83 (ddd, J = 17.1, 10.4, 6.1 Hz, 1H), 5.19 (dt, J = 17.1, 1.3 Hz, 1H), 5.06 (dt, J = 10.4, 1.3 Hz, 1H), 3.84 – 3.74 (m, 10H), 3.71 (d, J = 12.6 Hz, 1H), 3.53 – 3.48 (m, 1H), 2.73 (dd, J = 13.6, 4.1 Hz, 1H), 2.38 (dd, J = 13.6, 9.0 Hz, 1H), 1.80 (s, 2H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 160.3, 158.7, 141.4, 114.3, 107.9, 90.5, 55.7, 55.3, 53.1, 40.1, 23.8; ESIHRMS calculated for C$_{14}$H$_{23}$NO$_3$S [M+H]$^+$, 284.1320; found, 284.1311.

(R)-N-((R)-4-(1,2-Dimethyl-2-(2-oxo-2-(((R)-1-((2,4,5-trimethoxybenzyl)thio)but-3-en-2-yl)amino)ethyl)hydrazineyl)-3-methylbutyl)-N,2-dimethylbutanamide (197).

A heterogeneous mixture of **191** (25 mg, 0.06 mmol), palladium (25 mg 10 wt.% on carbon) and MeOH (1.5 mL) was stirred under hydrogen environment (1 atm) for 15 min and filtered through a pad of Celite®. The resultant filtrate was concentrated under reduced pressure to afford crude acid intermediate. To a solution of this crude acid intermediate and amine **196** (45 mg, 0.16 mmol) in dry CH$_2$Cl$_2$ (0.7 mL) were added HATU (49 mg, 0.13 mmol) and DIPEA (22 µL, 0.13 mmol) at room temperature. The reaction was stirred for 5 h and concentrated under reduced pressure. Purification of the residue by flash column chromatography on silica gel (20 – 70% acetone in CH$_2$Cl$_2$) afforded **197** (29 mg, 81%) as a yellow oil. $[\alpha]^{20}_D$ –22.6 (c 0.5, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 7.40 – 7.26 (m, 1H), 6.09 (s, 2H), 5.90 – 5.78 (m, 1H), 5.16 (dd, J = 17.2, 6.2 Hz, 1H), 5.10 (dd, J = 10.4, 6.2 Hz, 1H), 4.81 – 4.69 (m, 1H), 3.81 – 3.66 (m, 11H), 3.45 – 3.09 (m, 4H), 2.96 (s, 3H, rotamer), 2.89 (s, 3H, rotamer), 2.79 – 2.70 (m, 1H), 2.70 – 2.63 (m, 1H), 2.59 – 2.50 (m, 1H), 2.35 – 2.19 (m, 8H), 1.73 – 1.49 (m, 3H), 1.42 – 1.27 (m, 2H), 1.09 – 1.03 (m, 3H), 0.94 – 0.89 (m, 3H), 0.88 – 0.82(m, 3H). ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 176.4, 176.1, 170.1, 169.8, 160.3, 160.3, 158.73,

158.72, 137.5, 137.4, 115.04, 115.02, 108.0, 107.9, 90.5, 60.3, 59.7, 58.1, 57.6, 55.7, 55.3, 49.98, 49.89, 47.9, 46.0, 37.4, 37.3, 37.2, 36.9, 36.7, 36.2, 35.5, 35.1, 34.1, 33.6, 32.2, 29.33, 29.28, 29.24, 29.19, 27.4, 27.0, 24.18, 24.15, 18.2, 18.1, 17.8, 17.1, 12.1, 12.0; ESIHRMS calculated for $C_{29}H_{51}N_4O_5S$ $[M+H]^+$, 567.3580; found, 567.3582.

(R)-N-((R)-4-(2-(2-(((R)-1-Mercaptobut-3-en-2-yl)amino)-2-oxoethyl)-1,2-dimethylhydrazineyl)-3-methylbutyl)-N,2-dimethylbutanamide (198).

To an ice-cold solution (<5 °C) of **197** (25 mg, 0.04 mmol) in dry CH_2Cl_2 (0.5 mL) were added trifluoroacetic acid (35 μL, 0.44 mmol) and Et_3SiH (35 μL, 0.22 mmol) dropwise one after the other. After stirring for 1 h at room temperature, additional trifluoroacetic acid (35 μL, 0.44 mmol) and Et_3SiH (35 μL, 0.22 mmol) were added dropwise and stirring was continued for additional 1 h. The reaction mixture was concentrated under reduced pressure (<25 °C water-bath) and purified by flash column chromatography on silica gel (0 – 20% MeOH in CH_2Cl_2) to give **198** (14 mg, 82%) as a colorless oil. $[\alpha]^{20}_D$ –15.2 (c 0.25, $CHCl_3$); ^1H NMR (600 MHz, $CDCl_3$) , both rotamers, δ 7.97 (d, J = 8.1 Hz, 1H, rotamer), 7.63 (d, J = 8.4 Hz, 1H, rotamer), 5.99 – 5.59 (m, 1H), 5.33 – 5.08 (m, 2H), 4.76 – 4.54 (m, 1H), 3.63 (d, J = 15.0 Hz, 1H), 3.55 – 3.23 (m, 3H), 3.16 – 3.06 (m, 1H), 3.01 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.80 – 2.71 (m, 3H), 2.69 (s, 3H, rotamer), 2.64 (s, 3H, rotamer), 2.61 – 2.56 (m, 1H), 2.47 (s, 3H, rotamer), 2.45 (s, 3H, rotamer), 1.87 – 1.57 (m, 3H), 1.49 – 1.32 (m, 3H), 1.09 – 0.96 (m, 6H), 0.88 – 0.82 (m, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) major rotamer only; δ 177.1, 167.4, 135.5, 117.2, 59.8, 56.7, 53.1, 45.8, 37.4, 36.2, 35.9, 35.4, 32.0, 28.8, 27.9, 26.9, 18.1, 16.9, 12.0; ESIHRMS calculated for $C_{19}H_{39}N_4O_2S$ $[M+H]^+$, 387.2794; found, 387.2788.

Benzyl *N*-(dibenzylamino)-*N*-methylglycinate (199).

A mixture of 1,1-dibenzyl-2-methylhydrazine **78** (470 mg, 2.08 mmol), NaHCO$_3$ (262 mg, 3.12 mmol), benzyl bromoacetate (714 mg, 3.12 mmol), DMF (0.7 mL), and toluene (2.1 mL) was stirred for 3 h at 85 $^\circ$C. The reaction was cooled to room temperature, diluted with toluene, washed with water followed by brine, dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure. The residue was purified by flash column chromatography on silica gel (0 – 20% EtOAc in hexane) to give **119** (650 mg, 83%) as a colorless oil: ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.51 – 7.09 (m, 15H), 5.04 (s, 2H), 3.71 (s, 4H), 3.45 (s, 2H), 2.57 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.4, 139.2, 135.9, 129.1, 128.5, 128.4, 128.2, 128.0, 126.8, 66.3, 58.2, 54.3, 35.8; ESIHRMS calculated for C$_{24}$H$_{27}$N$_2$O$_2$ [M+H]$^+$, 375.2073; found, 375.2074.

N-(Dibenzylamino)-*N*-methylglycine (200).

To an ice-cold solution of **199** (100 mg, 0.26 mmol) in MeOH (0.4 mL) was added KOH solution (0.4 mL, 7 M in water, 2.80 mmol) dropwise. After stirring for 22 h at room temperature, the volatile components of the reaction were evaporated under reduced pressure. The aqueous residue was diluted with water and non-polar impurities were washed out with CH$_2$Cl$_2$ and discarded. The leftover aqueous residue was acidified to pH 1 by addition of 2 M HCl and then extracted with CH$_2$Cl$_2$. The so-obtained organic extract was dried over anhydrous Na$_2$SO$_4$, and concentrated under reduced pressure to afford the desired acid **200** (75 mg, 99%) as a yellow oil: ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.45 – 7.17 (m, 10H), 3.73 (s, 4H), 3.35 (s, 2H), 2.55 (s, 3H); ^{13}C NMR (100 MHz, CDCl$_3$) δ 170.6, 136.8, 129.2, 128.7, 127.9, 57.5, 54.1, 34.2; ESIHRMS calculated for C$_{17}$H$_{19}$N$_2$O$_2$ [M–H]$^-$, 283.1447; found, 283.1461.

(*R*)-2-(2,2-Dibenzyl-1-methylhydrazineyl)-*N*-(1-hydroxybut-3-en-2-yl)acetamide (201).

To a stirred solution of acid **200** (250 mg, 0.88 mmol) and amino alcohol **112**[147] (280 mg, 2.20 mmol) in dry CH_2Cl_2 (5 mL) were added DIPEA (470 μL, 2.64 mmol), DMAP (55 mg, 0.44 mmol), and EDCI.HCl (520 mg, 2.64 mmol) at room temperature. After stirring for 12 h, the reaction mixture was concentrated under reduced pressure and the residue was purified by flash column chromatography on silica gel to afford **201** (290 mg, 93%) as a colorless oil (eluent: 50 – 100% EtOAc in hexane). $[\alpha]^{20}_D$ +32.8 (*c* 2.4, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.43 – 7.19 (m, 10H), 6.51 (d, *J* = 6.9 Hz, 1H), 5.45 (ddd, *J* = 17.0, 10.5, 5.8 Hz, 1H), 5.05 (d, *J* = 10.5 Hz, 1H), 4.90 (d, *J* = 17.0 Hz, 1H), 4.29 – 4.09 (m, 1H), 3.68 (s, 4H), 3.45 (d, *J* = 16.6 Hz, 1H), 3.41 (dd, *J* = 11.3, 4.2 Hz, 1H), 3.33 (d, *J* = 16.6 Hz, 1H), 3.28 (dd, *J* = 11.3, 6.8 Hz, 1H), 2.64 (br s, 1H), 2.48 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.0, 138.9, 134.2, 128.7, 128.4, 127.2, 116.6, 65.2, 59.2, 54.5, 53.8, 35.7; ESIHRMS calculated for $C_{21}H_{27}N_2O_3Na$ [M+Na]$^+$, 376.2001; found, 376.2007.

(*R*)-2-((2,2-Dibenzyl-1-methylhydrazineyl)methyl)-4-vinyl-4,5-dihydrooxazole (202).

DAST (9 μL, 0.08 mmol) was added to a chilled solution of **201** (20 mg, 0.06 mmol) in dry CH_2Cl_2 (1.2 mL) at –78 °C. The reaction was stirred for 10 min at –78 °C and quenched by saturated aq. $NaHCO_3$. The resultant mixture was warmed to room temperature and extracted with CH_2Cl_2. The organic extract was dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina to afford **202** (6 mg, 32%) as a colorless oil (eluent: 25% EtOAC in hexane): $[\alpha]^{20}_D$ +38.0 (*c* 0.15, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.40 – 7.12 (m, 10H), 5.80 (ddd, *J* = 17.2, 10.2, 7.1 Hz, 1H), 5.25 (d, *J* = 17.2 Hz, 1H), 5.16 (d,

J = 10.2 Hz, 1H), 4.61 – 4.49 (m, 1H), 4.14 (dd, *J* = 9.9, 8.4 Hz, 1H), 3.72 (t, *J* = 8.4 Hz, 1H), 3.66 (ABq, *J* = 13.2 Hz, 4H), 3.47 (ABq, *J* = 13.8 Hz, 2H), 2.55 (s, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 165.8, 139.5, 138.0, 129.2, 127.9, 126.7, 116.3, 72.1, 68.2, 53.6, 52.5, 35.7; ESIHRMS calculated for C$_{21}$H$_{26}$N$_3$O [M+H]$^+$, 336.2076; found, 336.2077.

(*R*)-2-(2,2-Dibenzyl-1-methylhydrazineyl)-*N*-(1-hydroxybut-3-en-2-yl)ethanethioamide (203).

DAST (55 μL, 0.42 mmol) was added to a chilled solution of **201** (100 mg, 0.28 mmol) in dry CH$_2$Cl$_2$ (5.7 mL) at –78 °C. The reaction was stirred for 10 min at –78 °C and quenched with saturated aq. NaHCO$_3$. The resultant mixture was warmed to room temperature and extracted with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$ and concentrated under reduced pressure. The resultant residue was stirred with MeOH-Et$_3$N (1:1, 5.7 mL, saturated with H$_2$S gas) for 13 h at room temperature. Then the reaction was concentrated under reduced pressure and purified by flash column chromatography on silica gel to give **203** (63 mg, 66%, 2 steps) as a yellow oil (eluent: 0 – 50% EtOAc in hexane) along with the starting material **201** (25 mg, 25%) as a colorless oil (eluent: 50 – 100% EtOAc in hexane). Data for **203**: [*α*]^{20}$_D$ +32.1 (*c* 0.95, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) δ 8.54 (d, *J* = 7.5 Hz, 1H), 7.46 – 7.15 (m, 10H), 5.39 (ddd, *J* = 16.9, 10.6, 5.8 Hz, 1H), 5.11 (d, *J* = 10.6 Hz, 1H), 5.07 – 4.99 (m, 1H), 4.95 (d, *J* = 16.9 Hz, 1H), 3.95 (d, *J* = 17.3 Hz, 1H), 3.83 (d, *J* = 17.3 Hz, 1H), 3.70 (s, 4H), 3.45 (dd, *J* = 11.2, 4.7 Hz, 1H), 3.36 (dd, *J* = 11.2, 6.1 Hz, 1H), 2.45 (s, 3H), 1.61 (br s, 1H); ^{13}C NMR (150 MHz, CDCl$_3$) δ 199.1, 138.8, 132.6, 128.6, 128.5, 127.2, 117.8, 67.1, 64.2, 57.5, 54.7, 35.6; ESIHRMS calculated for C$_{21}$H$_{28}$N$_3$OS [M+H]$^+$, 370.1953; found, 370.1950.

(*R*)-2-((2,2-Dibenzyl-1-methylhydrazineyl)methyl)-4-vinyl-4,5-dihydrothiazole (204).

DAST (5 µL, 0.04 mmol) was added to a chilled solution of **203** (10 mg, 0.03 mmol) in dry CH_2Cl_2 (0.5 mL) at −78 °C. The reaction was stirred for 10 min at −78 °C and quenched by addition of saturated aq. $NaHCO_3$. The resultant mixture was warmed to room temperature and extracted with CH_2Cl_2. The organic extract was dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina to afford **204** (7.3 mg, 77%) as a yellow oil (eluent: 0 – 10% EtOAc in hexane): $[\alpha]^{20}_D$ +50.0 (*c* 0.37, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$) δ 7.40 – 7.16 (m, 10H), 5.92 (ddd, *J* = 17.1, 10.3, 6.8 Hz, 1H), 5.27 (dt, *J* = 17.1, 1.2 Hz, 1H), 5.16 (dt, *J* = 10.3, 1.2 Hz, 1H), 4.97 – 4.78 (m, 1H), 3.71 (s, 4H), 3.71 (dd, *J* = 14.0, 1.2 Hz, 1H), 3.64 (dd, *J* = 14.0, 1.2 Hz, 1H), 3.25 (dd, *J* = 11.0, 8.7 Hz, 1H), 2.89 (dd, *J* = 11.0, 8.4 Hz, 1H), 2.45 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.3, 139.3, 137.3, 129.0, 128.0, 126.8, 116.0, 78.3, 57.7, 54.4, 37.2, 36.3; ESIHRMS calculated for $C_{21}H_{26}N_3S$ [M+H]+, 352.1847; found, 352.1844.

(*R*)-*N*-((*R*)-4-(2-(2-(((*R*)-1-Hydroxybut-3-en-2-yl)amino)-2-oxoethyl)-1,2-dimethylhydrazineyl)-3-methylbutyl)-*N*,2-dimethylbutanamide (206).

A heterogeneous mixture of **191** (65 mg, 0.17 mmol), palladium (65 mg, 10 wt.% on carbon), and MeOH (4 mL) was stirred under hydrogen environment (1 atm) for 15 min and filtered through a pad of Celite®. The resultant filtrate was concentrated under reduced pressure to give crude acid intermediate **205**. To a stirred solution of this crude acid **205** and amino alcohol **112**[147] (51 mg, 0.42 mmol) in dry CH_2Cl_2 (0.80 mL) were added DIPEA (89 µL, 0.50 mmol), DMAP (10 mg, 0.08 mmol), and EDCI.HCl (96 mg, 0.50 mmol) at room temperature. After stirring for 12 h, the reaction mixture was

concentrated under reduced pressure and the residue was purified by flash column chromatography on silica gel to afford **206** (43 mg, 70% 2 steps) as a colorless oil (eluent: 25 – 75% acetone in hexane). $[\alpha]^{20}_D$ +12.2 (*c* 0.55, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 7.54 (d, *J* = 6.6 Hz, 1H, rotamer), 7.33 (d, *J* = 8.1 Hz, 1H, rotamer), 5.95 – 5.77 (m, 1H), 5.30 – 5.10 (m, 2H), 4.53 (br s, 1H), 4.33 (br s, 1H), 3.78 – 3.61 (m, 2H), 3.60 – 3.49 (m, 1H), 3.38 – 3.07 (m, 3H), 3.00 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.64 – 2.50 (m, 1H), 2.43 – 2.12 (m, 7H), 1.88 – 1.73 (m, 2H), 1.73 – 1.52 (m, 2H), 1.45 – 1.30 (m, 1H), 1.30 – 1.15 (m, 1H), 1.13 – 0.99 (m, 3H), 0.99 – 0.65 (m, 6H); ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 176.6, 170.7, 135.7, 135.1, 116.6, 115.9, 65.1, 64.7, 60.7, 60.20, 57.9, 57.2, 53.2, 53.0, 47.6, 46.9, 37.4, 37.2, 36.9, 36.6, 36.1, 35.6, 35.4, 34.0, 33.7, 32.2, 29.6, 28.7, 27.3, 26.9, 18.3, 18.0, 17.8, 17.0, 12.1, 11.9; ESIHRMS calculated for C$_{19}$H$_{39}$N$_4$O$_3$ [M+H]$^+$, 371.3022; found, 371.3020.

(*R*)-*N*-((*R*)-4-(1,2-Dimethyl-2-(((*R*)-4-vinyl-4,5-dihydrooxazol-2-yl)methyl)hydrazineyl)-3-methylbutyl)-*N*,2-dimethylbutanamide (207).

DAST (6 μL, 0.04 mmol) was added to a chilled solution of **206** (11 mg, 0.03 mmol) in dry CH$_2$Cl$_2$ (0.9 mL) at –78 ^{o}C. The reaction was stirred for 10 min at –78 ^{o}C and quenched with saturated aq. NaHCO$_3$. The resultant mixture was warmed to room temperature and extracted with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina to afford **207** (3 mg, 30%) as a colorless oil (eluent: 50% EtOAC in hexane). $[\alpha]^{20}_D$ +33.3 (*c* 0.15, CH$_2$Cl$_2$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 5.79 (ddd, *J* = 17.2, 10.2, 7.0 Hz, 1H), 5.22 (dd, *J* = 17.2, 4.5 Hz, 1H), 5.16 – 5.11 (m, 1H), 4.62 – 4.54 (m, 1H), 4.45 – 4.30 (m, 1H), 4.01 – 3.86 (m, 1H), 3.46 – 3.22

(m, 4H), 2.99 (s, 3H, rotamer), 2.90 (s, 3H, rotamer), 2.63 – 2.51 (m, 1H), 2.41 – 2.20 (m, 8H), 1.78 – 1.52 (m, 3H), 1.44 – 1.32 (m, 1H), 1.32 – 1.18 (m, 1H), 1.10 – 1.04 (m, 3H), 0.92 – 0.90 (m, 3H), 0.88 – 0.84 (m, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 176.5, 176.1, 166.0, 165.9, 137.9, 137.8, 116.5, 116.4, 72.28, 72.25, 68.1, 61.2, 61.0, 60.0, 51.5, 51.3, 48.0, 46.0, 37.4, 37.1, 35.2, 35.1, 35.0, 34.9, 34.7, 34.0, 33.6, 32.2, 29.0, 28.9, 27.4, 27.0, 18.18, 18.16, 17.8, 17.1, 12.1, 12.00; ESIHRMS calculated for C$_{19}$H$_{37}$N$_4$O$_2$ [M+H]$^+$, 353.2917; found, 353.2915.

(*R*)-*N*-((*R*)-4-(2-(2-(((*R*)-1-Hydroxybut-3-en-2-yl)amino)-2-thioxoethyl)-1,2-dimethylhydrazineyl)-3-methylbutyl)-*N*,2-dimethylbutanamide (208).

DAST (22 µL, 0.17 mmol) was added to a chilled solution of **206** (42 mg, 0.11 mmol) in dry CH$_2$Cl$_2$ (2.3 mL) at –78 ^{o}C. The reaction was stirred for 10 min at –78 ^{o}C and quenched with saturated aq. NaHCO$_3$. The resultant mixture was warmed to room temperature and extracted with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$ and concentrated under reduced pressure. The resultant residue was stirred with MeOH-Et$_3$N (1:1, 2.3 mL, saturated with H$_2$S gas) for 13 h at room temperature. Then the reaction was concentrated under reduced pressure and purified by flash column chromatography on silica gel to give **208** (23 mg, 52%, 2 steps) as a yellow oil (eluent: 25 – 35% acetone in hexane) along with the starting material **206** (17 mg, 40%) as a yellow oil (eluent: 35 – 75% acetone in hexane). Data for **208**: [α]^{20}$_D$ +9.5 (*c* 0.95, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 9.78 – 9.25 (m, 1H), 5.97 – 5.80 (m, 1H), 5.33 – 5.07 (m, 3H), 4.55 (br s, 1H), 3.91 – 3.75 (m, 2H), 3.76 – 3.51 (m, 3H), 3.40 – 3.25 (m, 1H, rotamer), 3.13 – 3.02 (m, 1H, rotamer), 2.99 (s, 3H, rotamer), 2.89 (s, 3H, rotamer), 2.62 – 2.49 (m, 1H), 2.47 – 2.37 (m, 1H, rotamer), 2.34 – 2.23 (m, 7H), 2.19 – 2.08 (m,

1H), 1.87 – 1.51 (m, 3H), 1.45 – 1.13 (m, 2H), 1.10 – 1.00 (m, 3H), 0.96 – 0.81 (m, 6H); ^{13}C NMR (151 MHz, CDCl$_3$), major rotamer only, δ 199.0, 176.7, 134.0, 116.9, 64.7, 63.4, 61.1, 57.3, 47.0, 37.4, 37.0, 36.4, 35.5, 32.3, 29.8, 26.9, 18.2, 17.0, 11.9; ESIHRMS calculated for C$_{19}$H$_{39}$N$_4$O$_2$S [M+H]$^+$, 387.2794; found, 387.2789.

7,8-Diazakalkitoxin (110).

DAST (5 μL, 0.04 mmol) was added to a chilled solution of **208** (10 mg, 0.03 mmol) in dry CH$_2$Cl$_2$ (0.5 mL) at −78 °C. The reaction was stirred for 10 min at −78 °C and quenched by addition of saturated aq. NaHCO$_3$. The resultant mixture was warmed to room temperature and extracted with CH$_2$Cl$_2$. The organic extract was dried over anhydrous Na$_2$SO$_4$, concentrated under reduced pressure, and purified by flash column chromatography on neutral alumina to afford **110** (8.5 mg, 89%) as a yellow oil (eluent: 25 – 50% EtOAc in hexane): $[\alpha]^{20}_D$ +42.1 (c 0.28, CH$_2$Cl$_2$); ^{1}H NMR (600 MHz, CDCl$_3$) , both rotamers, δ 5.97 – 5.86 (m, 1H), 5.26 (dd, J = 17.1, 4.4 Hz, 1H), 5.16 – 5.11 (m, 1H), 4.96 – 4.82 (m, 1H), 3.59 – 3.46 (m, 2H), 3.45 – 3.26 (m, 3H), 3.08 – 2.81 (m, 4H), 2.65 – 2.50(m, 1H), 2.35 – 2.22 (m, 8H), 1.84 – 1.63 (m, 3H), 1.45 – 1.21 (m, 2H), 1.10 – 1.04 (m, 3H), 0.92 (d, J = 6.5 Hz, 3H), 0.85 (t, J = 7.4 Hz, 3H); ^{13}C NMR (150 MHz, CDCl$_3$) , both rotamers, δ 176.5, 176.1, 173.5, 173.2, 137.3, 137.2, 116.1, 116.0, 78.44, 78.39, 60.9, 60.5, 56.5, 56.4, 48.1, 46.0, 37.4, 37.1, 37.0, 35.9, 35.6, 35.13, 35.09, 34.9, 33.9, 33.6, 32.2, 28.9, 28.8, 27.4, 27.0, 18.18, 18.17, 17.8, 17.1, 12.1, 12.0.; ESIHRMS calculated for C$_{19}$H$_{37}$N$_4$OS [M+H]$^+$, 369.2688; found, 369.2686.

REFERENCES

(1)   Carnero, A. *Clin. Transl. Oncol.* **2006**, *8*, 482-490.

(2)   Swinney, D. C.; Anthony, J. *Nat. Rev. Drug Discov.* **2011**, *10*, 507-519.

(3)   Huigens Iii, R. W.; Morrison, K. C.; Hicklin, R. W.; Flood Jr, T. A.; Richter, M. F.; Hergenrother, P. J. *Nat. Chem.* **2013**, *5*, 195-202.

(4)   Lipinski, C. A.; Lombardo, F.; Dominy, B. W.; Feeney, P. J. *Adv. Drug Deliv. Rev.* **2001**, *46*, 3-26.

(5)   Congreve, M.; Carr, R.; Murray, C.; Jhoti, H. *Drug Discov. Today* **2003**, *8*, 876-877.

(6)   Yu, B.; Reynisson, J. *Eur. J. Med. Chem.* **2011**, *46*, 5833-5837.

(7)   Lovering, F.; Bikker, J.; Humblet, C. *J. Med. Chem.* **2009**, *52*, 6752-6756.

(8)   Burke, M. D.; Schreiber, S. L. *Angew. Chem. Int. Ed.* **2004**, *43*, 46-58.

(9)   Schreiber, S. L. *Science* **2000**, *287*, 1964-1969.

(10)   Galloway, W. R. J. D.; Isidro-Llobet, A.; Spring, D. R. *Nat. Commun.* **2010**, *1*, 80.

(11)   Cui, J.; Hao, J.; Ulanovskaya, O. A.; Dundas, J.; Liang, J.; Kozmin, S. A. *Proc. Natl. Acad. Sci. U.S.A.* **2011**, *108*, 6763-6768.

(12)   Morrison, K. C.; Hergenrother, P. J. *Nat. Prod. Rep.* **2014**, *31*, 6-14.

(13)   Mollin, J.; Kasparek, F.; Lasovsky, J. *Chem. Zvesti* **1975**, *1*, 39-43.

(14)   Bartmess, J. E. In *Chemistry of Hydroxylamines, Oximes, and Hydroxamic Acids*; Rappoport, Z., Liebman, J. F., Eds.; Wiley: Chichester, 2011; Vol. 2, p 115.

(15)   Riddell, F. G.; Turner, E. S.; Boyd, A. *Tetrahedron* **1979**, *35*, 259-261.

(16)   Wazeer, M. M.; Perzanowski, H.; Ali, S. A. *J. Chem. Soc., Perkin Trans. 2* **1997**, 411-418.

(17)   Raban, M.; Kost, D. *Tetrahedron* **1984**, *40*, 3345-3381.

(18)   Malik, G.; Ferry, A.; Guinchard, X.; Cresteil, T.; Crich, D. *Chem. Eur. J.* **2013**, *19*, 2168-2179.

(19)   Ferry, A.; Malik, G.; Guinchard, X.; Vetvicka, V.; Crich, D. *J. Am. Chem. Soc.* **2014**, *136*, 14852-14857.

(20)   Ogawa, K.; Takeuchi, Y.; Suzuki, H.; Yoshida, H. *J. Mol. Struct.* **1985**, *126*, 445-454.

(21)   Nelsen, S. F.; Rumack, D. T.; Sieck, L. W.; Meot-Ner, M. *J. Am. Chem. Soc.* **1988**, *110*, 6303-6308.

(22)   Politzer, P.; Murray, J. S. In *The Chemistry of Hydroxylamines, Oximes, and Hydroxamic Acids, Part 1*; Rappoport, Z., Liebman, J. F., Eds.; Wiley: Chichester, 2009, p 1-28.

(23)   Riddell, F. *Tetrahedron* **1981**, *37*, 849-858.

(24)   Lehn, J. In *Dynamic Stereochemistry*; Springer: Berlin, Heidelberg, 1970; Vol. 15, p 311-377.

(25)   Golik, J.; Dubay, G.; Groenewold, G.; Kawaguchi, H.; Konishi, M.; Krishnan, B.; Ohkuma, H.; Saitoh, K.; Doyle, T. W. *J. Am. Chem. Soc.* **1987**, *109*, 3462-3464.

(26)   Golik, J.; Clardy, J.; Dubay, G.; Groenewold, G.; Kawaguchi, H.; Konishi, M.; Krishnan, B.; Ohkuma, H.; Saitoh, K.; Doyle, T. W. *J. Am. Chem. Soc.* **1987**, *109*, 3461-3462.

(27)   Lee, M. D.; Dunne, T. S.; Siegel, M. M.; Chang, C. C.; Morton, G. O.; Borders, D. B. *J. Am. Chem. Soc.* **1987**, *109*, 3464-3466.

(28)   Lee, M. D.; Dunne, T. S.; Chang, C. C.; Ellestad, G. A.; Siegel, M. M.; Morton, G. O.; McGahren, W. J.; Borders, D. B. *J. Am. Chem. Soc.* **1987**, *109*, 3466-3468.

(29)   Berge, J. M.; Copley, R. C.; Eggleston, D. S.; Hamprecht, D. W.; Jarvest, R. L.; Mensah, L. M.; O'Hanlon, P. J.; Pope, A. J. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 1811-1814.

(30)   Zhang, Y.; Albohy, A.; Zou, Y.; Smutova, V.; Pshezhetsky, A. V.; Cairo, C. W. *J. Med. Chem.* **2013**, *56*, 2948-2958.

(31)   Axerio-Cilies, P.; Castañeda, I. P.; Mirza, A.; Reynisson, J. *Eur. J. Med. Chem.* **2009**, *44*, 1128-1134.

(32)   Luo, Y.-R. *Handbook of Bond Dissociation Energies in Organic Compounds*; CRC press: Boca Raton, 2003.

(33)   Benigni, R.; Giuliani, A.; Franke, R.; Gruska, A. *Chem. Rev.* **2000**, *100*, 3697-3714.

(34)   Benigni, R.; Bossa, C. *Chem. Rev.* **2011**, *111*, 2507-2536.

(35)   Dhanju, S.; Crich, D. *Org. Lett.* **2016**, *18*, 1820-1823.

(36)   Truitt, P.; Long, L. M.; Mattison, M. *J. Am. Chem. Soc.* **1948**, *70*, 2829-2829.

(37)   Sun, R.; Lü, M.; Chen, L.; Li, Q.; Song, H.; Bi, F.; Huang, R.; Wang, Q. *J. Agric. Food. Chem.* **2008**, *56*, 11376-11391.

(38)   Yamawaki, K.; Nomura, T.; Yasukata, T.; Uotani, K.; Miwa, H.; Takeda, K.; Nishitani, Y. *Biorg. Med. Chem.* **2007**, *15*, 6716-6732.

(39)   Foot, O. F.; Knight, D. W. *Chem. Commun.* **2000**, 975-976.

(40)   Chen, N.; Xie, J. *Org. Biomol. Chem.* **2016**, *14*, 11028-11047.

(41)   Melman, A. In *The Chemistry of Hydroxylamines, Oximes, and Hydroxyamic Acids, Part 1*; Rappoport, Z., Liebman, J. F., Eds.; Wiley: Chichester, 2009, p 117-161.

(42)   Malik, G.; Guinchard, X.; Crich, D. *Org. Lett.* **2011**, *14*, 596-599.

(43)   Sun, L.; Li, P.; Amankulor, N.; Tang, W.; Landry, D. W.; Zhao, K. *J. Org. Chem.* **1998**, *63*, 6472-6475.

(44)   Shirokane, K.; Kurosaki, Y.; Sato, T.; Chida, N. *Angew. Chem. Int. Ed.* **2010**, *49*, 6369-6372.

(45)   Barnes, K. L.; Chen, K.; Catalano, V. J.; Jeffrey, C. S. *Org. Chem. Front.* **2015**, *2*, 497-501.

(46)   Shvo, Y. In *The Chemistry of Hydrazo, Azo and Azoxy Groups* Patai, S., Ed.; John Wiley & Sons: New York, 1975, p 1017-1095.

(47)   Penney, W. G.; Sutherland, G. B. B. M. *J. Chem. Phys.* **1934**, *2*, 492-498.

(48)   Veillard, A. *Theor. Chim. Acta* **1966**, *5*, 413-421.

(49)   Gillespie, R.; Nyholm, R. *Q. Rev. Chem. Soc.* **1957**, *11*, 339-380.

(50)   Gillespie, R. *Can. J. Chem.* **1960**, *38*, 818-826.

(51)   Gillespie, R. *J. Chem. Educ.* **1963**, *40*, 295-301.

(52)   Allen, L. *Chem. Phys. Lett.* **1968**, *2*, 597-601.

(53)   Wolfe, S. *Acc. Chem. Res.* **1972**, *5*, 102-111.

(54)   Pople, J. A.; Radom, L.; Hehre, W. J. *J. Am. Chem. Soc.* **1971**, *93*, 289-300.

(55)   Hamada, Y.; Hirakawa, A. Y.; Tamagake, K.; Tsuboi, M. *J. Mol. Spectrosc.* **1970**, *35*, 420-435.

(56)   Waldman, A. J.; Ng, T. L.; Wang, P.; Balskus, E. P. *Chem. Rev.* **2017**, *117*, 5784-5863.

(57)   Blair, L. M.; Sperry, J. *J. Nat. Prod.* **2013**, *76*, 794-812.

(58)   Le Goff, G.; Ouazzani, J. *Biorg. Med. Chem.* **2014**, *22*, 6529-6544.

(59)   Qingbo, Z.; Attila, M.; Sumei, L.; Yuchan, C.; Wenjun, Z.; Xinpeng, T.; Haibo, Z.; Huixian, L.; Weimin, Z.; Si, Z.; Jianhua, J.; Tibor, K.; Changsheng, Z. *Eur. J. Org. Chem.* **2012**, *2012*, 5256-5262.

(60)   Zhongli, X.; Martin, B.; Ling, D.; Christian, H. *Angew. Chem. Int. Ed.* **2012**, *51*, 10293-10297.

(61)   Gamberini, M.; Cidade, M. R.; Valotta, L. A.; Armelin, M. C.; Leite, L. C. *Carcinogenesis* **1998**, *19*, 147-155.

(62)   Klaus, R.; Tawfig, S. *Arch. Pharm.* **1998**, *331*, 308-312.

(63)   Putt, K. S.; Chen, G. W.; Pearson, J. M.; Sandhorst, J. S.; Hoagland, M. S.; Kwon, J.-T.; Hwang, S.-K.; Jin, H.; Churchwell, M. I.; Cho, M.-H.; Doerge, D. R.; Helferich, W. G.; Hergenrother, P. J. *Nat. Chem. Biol.* **2006**, *2*, 543-550.

(64)   Lopez-Munoz, F.; Alamo, C. *Curr. Pharm. Des.* **2009**, *15*, 1563-1586.

(65)   Toth, B. *In Vivo* **2000**, *14*, 299-319.

(66)   Gannett, P. M.; Shi, X.; Lawson, T.; Kolar, C.; Toth, B. *Chem. Res. Toxicol.* **1997**, *10*, 1372-1377.

(67)   Koepke, S. R.; Kroeger-Koepke, M. B.; Michejda, C. J. *Chem. Res. Toxicol.* **1990**, *3*, 17-20.

(68)   Lawson, T.; Gannett, P. M.; Yau, W.-M.; Dalal, N. S.; Toth, B. *J. Agric. Food. Chem.* **1995**, *43*, 2627-2635.

(69)   Gannett, P. M.; Lawson, T.; Miller, M.; Thakkar, D.; Lord, J.; Yau, W.-M.; Toth, B. *Chem. Biol. Interact.* **1996**, *101*, 149-164.

(70)   Hung, M. H.; Stock, L. M. *J. Org. Chem.* **1982**, *47*, 448-453.

(71)   Kikugawa, K.; Kato, T.; Kojima, K. *Mutat. Res.* **1992**, *268*, 65-75.

(72)   Augusto, O. *Free Radical Biol. Med.* **1993**, *15*, 329-336.

(73)   Griffiths, J.; Murphy, J. A. *J. Chem. Soc., Chem. Commun.* **1992**, 24-26.

(74)   Bredihhin, A.; Mäeorg, U. *Tetrahedron* **2008**, *64*, 6788-6793.

(75)   Huddleston, P. R.; Coutts, I. G. In *Comprehensive Organic Functional Group Transformations*; Katritzky, A. R., Meth-Cohn, O., Rees, C. W., Eds.; Pergamon/Elsevier: Oxford, 1995; Vol. 2, p 371-383.

(76)   Stanley, R. S.; Wolf, K.; Academic Press Inc.: San Diego, California, 1992, p 136-146.

(77)   Ragnarsson, U. *Chem. Soc. Rev.* **2001**, *30*, 205-213.

(78)   Hinman, R. L. *J. Am. Chem. Soc.* **1956**, *78*, 1645-1649.

(79)   Bredihhin, A.; Groth, U. M.; Mäeorg, U. *Org. Lett.* **2007**, *9*, 1097-1099.

(80)   Bredihhin, A.; Mäeorg, U. *Org. Lett.* **2007**, *9*, 4975-4977.

(81)   Feuer, H.; Brown Jr, F. *J. Org. Chem.* **1970**, *35*, 1468-1471.

(82)   Rosen, B. R.; Werner, E. W.; O'Brien, A. G.; Baran, P. S. *J. Am. Chem. Soc.* **2014**, *136*, 5571-5574.

(83)   Audrieth, L. F.; Diamond, L. H. *J. Am. Chem. Soc.* **1954**, *76*, 4869-4871.

(84)   Omietanski, G. M.; Melmers, A. D.; Shellman, R. W.; Sisler, H. H. *J. Am. Chem. Soc.* **1956**, *78*, 3874-3877.

(85)   Diamond, L. H.; Audrieth, L. F. *J. Am. Chem. Soc.* **1955**, *77*, 3131-3131.

(86)   Wallace, R. G. *Aldrichimica Acta* **1980**, *13*, 3-11.

(87)   Keller, R.; Smith, P. A. *J. Am. Chem. Soc.* **1944**, *66*, 1122-1124.

(88)   Armstrong, A.; Jones, L. H.; Knight, J. D.; Kelsey, R. D. *Org. Lett.* **2005**, *7*, 713-716.

(89)   Greck, C.; Genêt, J. P. *Synlett* **1997**, 741-748.

(90)   Tsupova, S.; Mäeorg, U. *Org. Lett.* **2013**, *15*, 3381-3383.

(91)   Zhu, Y.; Shi, Y. *Org. Lett.* **2013**, *15*, 1942-1945.

(92)   Vázquez, A. J.; Rodriguez, C.; Nudelman, N. S. *Synth. Commun.* **2009**, *39*, 3958-3972.

(93)   Shestakov, A. N.; Kuznetsov, M. A. *Chem. Commun.* **2016**, *52*, 2398-2400.

(94)   Berman, F. W.; Gerwick, W. H.; Murray, T. F. *Toxicon* **1999**, *37*, 1645-1648.

(95)   Wu, M.; Okino, T.; Nogle, L. M.; Marquez, B. L.; Williamson, R. T.; Sitachitta, N.; Berman, F. W.; Murray, T. F.; McGough, K.; Jacobs, R.; Colsen, K.; Asano, T.; Yokokawa, F.; Shioiri, T.; Gerwick, W. H. *J. Am. Chem. Soc.* **2000**, *122*, 12041-12042.

(96)   Tan, L. T.; Williamson, R. T.; Gerwick, W. H.; Watts, K. S.; McGough, K.; Jacobs, R. *J. Org. Chem.* **2000**, *65*, 419-425.

(97)   LePage, K. T.; Goeger, D.; Yokokawa, F.; Asano, T.; Shioiri, T.; Gerwick, W. H.; Murray, T. F. *Toxicol. Lett.* **2005**, *158*, 133-139.

(98)   Manger, R. L.; Leja, L. S.; Lee, S. Y.; Hungerford, J. M.; Hokama, Y.; Dickey, R. W.; Granade, H. R.; Lewis, R.; Yasumoto, T.; Wekell, M. M. *J. AOAC Int.* **1995**, *78*, 521-527.

(99)   White, J. D.; Xu, Q.; Lee, C.-S.; Valeriote, F. A. *Org. Biomol. Chem.* **2004**, *2*, 2092-2102.

(100)   Morgan, J.; Liu, Y.; Coothankandaswamy, V.; Mahdi, F.; Jekabsons, M.; Gerwick, W.; Valeriote, F.; Zhou, Y.-D.; Nagle, D. *Marine Drugs* **2015**, *13*, 1552-1568.

(101) Yokokawa, F.; Asano, T.; Okino, T.; Gerwick, W. H.; Shioiri, T. *Tetrahedron* **2004**, *60*, 6859-6880.

(102) Roush, W. R.; Palkowitz, A. D.; Ando, K. *J. Am. Chem. Soc.* **1990**, *112*, 6348-6359.

(103) Li, G.; Pattel, D.; Hruby, V. J. *Tetrahedron: Asymmetry* **1993**, *4*, 2315-2318.

(104) Wipf, P.; Miller, C. P.; Venkatraman, S.; Fritch, P. C. *Tetrahedron Lett.* **1995**, *36*, 6395-6398.

(105) White, J. D.; Lee, C.-S.; Xu, Q. *Chem. Commun.* **2003**, 2012-2013.

(106) Myers, A. G.; Yang, B. H.; Chen, H.; McKinstry, L.; Kopecky, D. J.; Gleason, J. L. *J. Am. Chem. Soc.* **1997**, *119*, 6496-6511.

(107) Umezawa, T.; Sueda, M.; Kamura, T.; Kawahara, T.; Han, X.; Okino, T.; Matsuda, F. *J. Org. Chem.* **2012**, *77*, 357-370.

(108) Balieu, S.; Hallett, G. E.; Burns, M.; Bootwicha, T.; Studley, J.; Aggarwal, V. K. *J. Am. Chem. Soc.* **2015**, *137*, 4398-4403.

(109) Kopecky, D. J.; Rychnovsky, S. D. *J. Org. Chem.* **2000**, *65*, 191-198.

(110) Vo, C.-V. T.; Mitchell, T. A.; Bode, J. W. *J. Am. Chem. Soc.* **2011**, *133*, 14082-14089.

(111) Barrios, F. J.; Zhang, X.; Colby, D. A. *Org. Lett.* **2010**, *12*, 5588-5591.

(112) Biloski, A. J.; Ganem, B. *Synthesis* **1983**, 537-538.

(113) O'Neil, I. A.; Cleator, E.; Tapolczay, D. J. *Tetrahedron Lett.* **2001**, *42*, 8247-8249.

(114) Neises, B.; Steglich, W. *Angew. Chem. Int. Ed. Engl.* **1978**, *17*, 522-524.

(115) Overend, W. G.; Rees, C. W.; Sequeira, J. S. *J. Chem. Soc.* **1962**, 3429-3440.

(116)   Zhang, Z.; Ollmann, I. R.; Ye, X.-S.; Wischnat, R.; Baasov, T.; Wong, C.-H. *J. Am. Chem. Soc.* **1999**, *121*, 734-753.

(117)   Martin, A.; Arda, A.; Désiré, J.; Martin-Mingot, A.; Probst, N.; Sinaÿ, P.; Jiménez-Barbero, J.; Thibaudeau, S.; Blériot, Y. *Nat. Chem.* **2015**, *8*, 186.

(118)   Dahanukar, V. H.; Rychnovsky, S. D. *J. Org. Chem.* **1996**, *61*, 8317-8320.

(119)   Kadota, I.; Uyehara, H.; Yamamoto, Y. *Tetrahedron* **2004**, *60*, 7361-7365.

(120)   Tyrrell, E.; Tsang, M. W. H.; Skinner, G. A.; Fawcett, J. *Tetrahedron* **1996**, *52*, 9841-9852.

(121)   Le Grel, P.; Asprogenidi, A.; Huez, P.; Le Grel, B.; Salauen, A.; Roisnel, T.; Potel, M.; Rasti, E.; Hocquet, A. *Chirality* **2013**, *25*, 341-349.

(122)   Riddell, F. G.; Turner, E. S. *J. Chem. Soc., Perkin Trans. 2* **1978**, 707-708.

(123)   Bach, R. D.; Raban, M. In *Acyclic Organonitrogen Sterodynamics*; Lambert, J. B., Takeuchi, Y., Eds.; VCH: New York, 1992, p 63-103.

(124)   Raban, M.; Kost, D. In *Acyclic Organonitrogen Sterodynamics*; Lambert, J. B., Takeuchi, Y., Eds.; VCH: New York, 1992, p 57-88.

(125)   Anderson, J. E.; Tocher, D. A.; Corrie, J. E. T.; Lunazzi, L. *J. Am. Chem. Soc.* **1993**, *115*, 3494-3498.

(126)   Kliegel, W.; Riebe, U.; Patrick, B. O.; Rettig, S. J.; Trotter, J. *Acta Cryst. Sect. E* **2001**, *E57*, o1173-o1174.

(127)   Matousˇek, V.; Pietrasiak, E.; Sigrist, L.; Czarniecki, B.; Togni, A. *Eur. J. Org. Chem.* **2014**, 3087-3092.

(128)   Liew, S. K.; Kaldas, S. J.; Yudin, A. K. *Org. Lett.* **2016**, *18*, 6268-6271.

(129) Busfield, W. K.; Englelhardt, L. M.; Healy, P. C.; Jenkins, I. D.; Thang, S. H.; White, A. H. *Aus. J. Chem.* **1986**, *39*, 357-365.

(130) Busfield, W. K.; Byriel, K. A.; Grice, D.; Jenkins, I. D.; Lynch, D. E. *J. Chem. Soc., Perkin Trans. 2* **2000**, 757-760.

(131) Grice, I. D.; Jenkins, I. D.; Busfield, W. K.; Byrield, K. A.; Kennarde, C. H. L. *Acta Cryst, E.* **2004**, *E60*, o2386-2387.

(132) Liu, Y.-P.; Zhang, X.-F.; Liu, K.-J.; Liu, Y. *Acta Cryst. E.* **2006**, *62*, o3672-o3673.

(133) Morozov, D. A.; Kirilyuk, I. A.; Komarov, D. A.; Goti, A.; Bagryanskaya, I. Y.; Kuratieva, N. V.; Grigor'ev, I. A. *J. Org. Chem.* **2012**, *77*, 10688-10698.

(134) Busfield, W. K.; Jenkins, I. D.; Thang, S. H.; Moad, G.; Rizzardo, E.; Solomon, D. H. *J. Chem. Soc, Chem Commun.* **1985**, 1249-1250.

(135) Anderson, J. E.; Corrie, J. E. T. *J. Chem. Soc., Perkin Trans. 2* **1992**, 1027-1031.

(136) Eliel, E. L.; Wilen, S. H. *Stereochemistry of Organic Compounds*; Wiley: New York, 1994.

(137) Anderson, J. E.; Casarini, D.; Corrie, J. E. T.; Lunazzi, L. *J. Chem. Soc., Perkin Trans. 2* **1993**, 1299-1304.

(138) Perrin, C. L.; Thoburn, J. D.; Elsheimer, S. *J. Org. Chem.* **1991**, *56*, 7034-7038.

(139) Belostotskii, A. M. M. *Chem. Eur. J.* **1999**, *5*, 449-455.

(140) Tafazzoli, M.; Suarez, C.; True, N. S.; LeMaster, C. B.; LeMaster, C. L. *J. Phys. Chem.* **1992**, *96*, 10201-10205.

(141) Anderson, J. E.; John, E. *J. Chem. Soc., Perkin Trans. 2* **1993**, 1299-1304.

(142) Fu, J.; Lau, K.; Barra, M. *J. Org. Chem.* **2009**, *74*, 1770-1773.

(143) Lunazzi, L.; Cerioni, G.; Foresti, E.; Macciantelli, D. *J. Chem. Soc., Perkin Trans. 2* **1978**, 686-691.

(144) Weiser, J.; Golan, O.; Fitjer, L.; Biali, S. E. *J. Org. Chem.* **1996**, *61*, 8277-8284.

(145) Machida, S.; Kato, N.; Harada, K.; Ohkanda, J. *J. Am. Chem. Soc.* **2011**, *133*, 958-963.

(146) Fürstner, A.; De Souza, D.; Turet, L.; Fenster, M. D.; Parra-Rapado, L.; Wirtz, C.; Mynott, R.; Lehmann, C. W. *Chem. Eur. J.* **2007**, *13*, 115-134.

(147) Olivier, K. S.; Van Nieuwenhze, M. S. *Org. Lett.* **2010**, *12*, 1680-1683.

(148) Bajpai, R.; Yang, F.; Curran, D. P. *Tetrahedron Lett.* **2007**, *48*, 7965-7968.

(149) Fuganti, C.; Grasselli, P. *J. Chem. Soc., Chem. Commun.* **1979**, 995-997.

(150) Speltz, T. E.; Fanning, S. W.; Mayne, C. G.; Fowler, C.; Tajkhorshid, E.; Greene, G. L.; Moore, T. W. *Angew. Chem. Int. Ed.* **2016**, *55*, 4252-4255.

(151) Inghardt, T.; Frejd, T.; Svensson, G. *Tetrahedron* **1991**, *47*, 6469-6482.

(152) Jin, W.; Trzupek, J. D.; Rayl, T. J.; Broward, M. A.; Vielhauer, G. A.; Weir, S. J.; Hwang, I.; Boger, D. L. *J. Am. Chem. Soc.* **2007**, *129*, 15391-15397.

(153) Huynh, U.; Uddin, M. N.; Wengryniuk, S. E.; McDonald, S. L.; Coltart, D. M. *Tetrahedron Lett.* **2016**, *57*, 4799-4802.

(154) Qin, L.; Zhou, Z.; Wei, J.; Yan, T.; Wen, H. *Synth. Commun.* **2010**, *40*, 642-646.

(155) Sheikh, M. C.; Takagi, S.; Ogasawara, A.; Ohira, M.; Miyatake, R.; Abe, H.; Yoshimura, T.; Morita, H. *Tetrahedron* **2010**, *66*, 2132-2140.

(156) Evans, D. A.; Ng, H. P.; Clark, J. S.; Rieger, D. L. *Tetrahedron* **1992**, *48*, 2127-2142.

(157) Evans, D. A.; Dow, R. L.; Shih, T. L.; Takacs, J. M.; Zahler, R. *J. Am. Chem. Soc.* **1990**, *112*, 5290-5313.

(158) He, R.; Ching, S. M.; Lam, Y. *J. Comb. Chem.* **2006**, *8*, 923-928.

(159) VanRheenen, V.; Kelly, R. C.; Cha, D. Y. *Tetrahedron Lett.* **1976**, *17*, 1973-1976.

(160) Munson, M. C.; García-Echeverría, C.; Albericio, F.; Barany, G. *J. Org. Chem.* **1992**, *57*, 3013-3018.

(161) Valeriote, F. A.; Tenney, K.; Pietraszkiewicz, H.; Edelstein, M.; Johnson, T. A.; Amagata, T.; Crews, P. *J. Exp. Ther. Oncol.* **2012**, *10*, 119-134.

(162) Berman, A. M.; Johnson, J. S. *J. Org. Chem.* **2006**, *71*, 219-224.

(163) Shang, M.; Zeng, S.-H.; Sun, S.-Z.; Dai, H.-X.; Yu, J.-Q. *Org. Lett.* **2013**, *15*, 5286-5289.

(164) Fernandes, L.; Fischer, F. L.; Ribeiro, C. W.; Silveira, G. P.; Sá, M. M.; Nome, F.; Terenzi, H. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 4499-4502.

(165) Sammelson, R. E.; Kurth, M. J. *Tetrahedron Lett.* **2001**, *42*, 3419-3422.

(166) Windle, J.; Kuhnle, J.; Beck, B. H. *J. Chem. Phys.* **1969**, *50*, 2630-2641.

(167) López-Iglesias, M.; Busto, E.; Gotor, V.; Gotor-Fernández, V. *Adv. Synth. Catal.* **2011**, *353*, 2345-2353.

(168) Ravichandran, R.; Compositions Stablilized with Etheres of Di- and Tri-substituted Hydroxylamines. U.S. Patents 4,696,964: September 29, 1987.

(169) Reddy, B. V. S.; Reddy, B. P.; Swapnil, N.; Yadav, J. S. *Tetrahedron Lett.* **2013**, *54*, 5781-5784.

(170) Evans, D. A.; Ng, H. P. *Tetrahedron Lett.* **1993**, *34*, 2229-2232.

ABSTRACT

HYDROXYLAMINES AND HYDRAZINES AS SURROGATES OF SP^3 CARBONS IN MEDICINAL CHEMISTRY

by

SANDEEP DHANJU

May 2019

Advisor: Dr. David Crich

Major: Chemistry (Organic)

Degree: Doctor of Philosophy

Current compound libraries that are used in high-throughput screening (HTS) are populated by low molecular weight and relatively planar compounds with high sp^2 character and little stereochemical complexity. Unfortunately, such planar compounds cover only a very small corner of chemical space. In order to increase the diversity, structurally complex molecules with high fractions of sp^3-hybridized atoms should be included in compound libraries. However, generating a large number of architecturally complex molecules using the current organic synthesis toolbox is considered tedious due to the requirement of multi-step synthesis. Recognizing this need, several creative strategies, like diversity-oriented synthesis, have been developed with the aim to access structurally complex drug-like scaffolds in a minimum number of steps. However, the efficiency of these strategies is impeded by the time-consuming process of addressing stereogenic complexity. The research presented in this thesis is aimed at providing an alternative strategy that not only enriches the Fsp^3 in compound collections but also reduces the stereogenic complexity.

Chapter one introduces hydroxylamines and hydrazines as interesting functional moieties that are capable of enriching current compound collections by increasing the fraction of sp^3-hybridized atoms without additional stereochemical complexity. As the basis of the research presented in this thesis is the rapid conformational changes of hydroxylamine and hydrazine-related molecules at ambient temperature due to their low barrier to inversion at nitrogen centers, their conformational properties are highlighted in this chapter. Moreover, the present position of hydroxylamines and hydrazines in medicinal chemistry, and the synthetic accessibility of hydroxylamine and hydrazine-related compounds are also discussed. The chapter ends by proposing kalkitoxin, a natural anti-cancer agent, as a substrate to test the concept of hydroxylamines and hydrazines as convertible mimics of the stereogenic centers.

Chapter two presents the discovery of an efficient synthetic method for tri-substituted hydroxylamine synthesis. This method was developed on the basis of acylation of *N,N*-disubstituted hydroxylamines followed by two-step reduction of the resulting *O*-acylhydroxylamines. The first reduction was conducted with DIBAL and was followed by treatment with acetic anhydride to give the *O*-(α-acetoxyalkyl)hydroxylamines; the second reduction was executed with triethylsilane and a Lewis acid. Manipulating the second reduction step, by replacing triethylsilane with a carbon nucleophile afforded a new C-C bond formation, and further expanded the scope of this method. The efficiency of the method is exemplified by synthesis of several novel tri-substituted hydroxylamines.

Chapter three describes the VT-NMR studies that were conducted to understand the contribution of three processes (N-inversion, N-O bond rotation, and ring flip) to the

complex stereomutation of *N*-alkoxypiperidine systems. Since the VT-NMR phenomena were found to be dependent on substituents at the 4-position of the piperidine ring system, the ring inversion process is concluded to be the main component of the barrier to stereomutation of such systems.

Chapter four concerns the design and development of hydrazine and hydroxylamine analogs of kalkitoxin, with the aim of testing the hypothesis of hydrazines and hydroxylamine as convertible mimics of stereogenic centers. The syntheses of kalkitoxin and its convertible hydrazine and hydroxylamine mimetics, 9-oxa-10-azakalkitoxin and 7,8-diazakalkitoxin, are described. Currently, their cytotoxicity studies against several human and murine tumor cell lines are under progress. The hydroxalog 9-oxa-10-azakalkitoxin (IC_{50} = 2.4 nm) was found to be as potent as kalktoxin (IC_{50} = 3.2 nm) against a human liver cell line, so supporting the hydroxylamine analog concept.

The thesis ends with a conclusion and the complete experimental details for the work presented.

AUTOBIOGRAPHICAL STATEMENT

SANDEEP DHANJU

Education Details:

Ph.D.:	Organic Chemistry (2013-present)
Advisor:	Prof. David Crich
Institution:	Wayne State University, Detroit, MI

M.Sc.:	Organic Chemistry (2013)
Advisor:	Prof. Surya Kant Kalauni
Institution:	Tribhuvan University, Kirtipur, Nepal

B.Sc.:	Chemistry (2009)
Institution:	Tribhuvan University, Kirtipur, Nepal

Publications:

- **Dhanju S**.; Blazejewski B; Crich D.* Synthesis of Trialkylhydroxylamines by Stepwise Reduction of *O*-Acyl *N,N*-Disubstituted Hydroxylamines. Substituent Effects on the Reduction of *O*-(1-Acyloxyalkyl)hydroxylamines and on the Conformational Dynamics of *N*-Alkoxypiperidines. *J. Org. Chem.* **2017**, *82*, 5345-5353.
- **Dhanju S**.; Crich D.* Synthesis of *N,N,O*-Trisubstituted Hydroxylamines by Stepwise Reduction and Substitution of *O*-Acyl *N,N*-Disubstituted Hydroxylamines. *Org. Lett.* **2016**, *18*, 1820−1823.

Presentations:

- Presented a poster at **ACS Division of Organic Chemistry 2017 Graduate Research Symposium** at Portland State University, Portland, Oregon, July 20-23, 2017. Title: "Hydroxylamines as Surrogates of Stereogenic Centers in Carbon Frameworks."
- Presented a poster at **18th Annual Chemistry Graduate Symposium**, Wayne State University, 2016. Title: "A Novel Method for the Synthesis of Highly Substituted Hydroxylamines."
- Presented a poster at **251th ACS National Meeting** at San Diego, CA. March 16, 2016. Title: "A Novel Method for the Synthesis of Highly Substituted Hydroxylamines."