ABSTRACT

SHYMANSKA, NATALIIA VASYLIVNA. Total Synthesis of the Synoxazolidinone Family of Natural Products and Novel Approaches for the Construction of Nitrogen and Oxygen Containing Heterocycles. (Under the direction of Dr. Joshua G. Pierce).

A plethora of natural products and pharmaceutical agents incorporate nitrogen and oxygen-containing heterocycles as their core structural moieties. The successful synthesis of biologically active heterocyclic compounds not only leads to desired scaffolds but also enables their functionalization, providing applications for medicinal chemistry and chemical biology studies. Development of new synthetic methods for the construction of novel and existing heterocycles is very important, especially in the context of natural product synthesis. This dissertation describes the development of several new approaches for the synthesis of novel nitrogen and oxygen-containing heterocycles, 4-oxazolidinones. Our synthetic efforts aimed at the construction of 4-oxazolidinone core scaffolds produced two alternative methods and enabled the first total synthesis of the synoxazolidinone family of antimicrobial natural products. A key transformation we employed in our synthesis was an intramolecular addition of an enol to \(N\)-acyliminium ion. Through modification of the \(N\)-acyliminium ion precursors and cyclization conditions we were able to prepare a variety of novel 4-oxazolidinones which we used to establish an initial antimicrobial activity profile of these heterocycles against clinically relevant strains of bacteria. In addition to natural products, we identified promising inhibitors of growth of methicillin-resistant \textit{Staphylococcus aureus} (MRSA). Our attempts to determine the mechanism of biological action of the synoxazolidinones led to the discovery of biofilm inhibition properties of 4-oxazolidinones.

During the optimization of the synthetic procedure for previously inaccessible functionalized 4-oxazolidinones we also discovered another class of heterocyclic compounds,
dihydropyrrolones. These medicinally relevant scaffolds originated from an alternative cyclization pathway from our cascade transformation towards 4-oxazolidinones. We subsequently developed a high yielding synthetic protocol for alkyl-containing dihydropyrrolones. During further explorations of the reactivity of dihydropyrrolones we utilized a sigmatropic Claisen rearrangement of their allyl ethers to access novel pyrrolidine-2,3-dione heterocycles containing a quaternary stereocenter. Finally, pyrrolidine-2,3-diones were efficiently converted to unprecedented, sterically crowded, β-amino acids which are described in the final section of this dissertation.
Total Synthesis of the Synoxazolidinone Family of Natural Products and Novel Approaches for the Construction of Nitrogen and Oxygen Containing Heterocycles.

by

Nataliia Vasylivna Shymanska

A dissertation submitted to the Graduate Faculty of North Carolina State University in partial fulfillment of the requirements for the Degree of Doctor of Philosophy

Chemistry

Raleigh, North Carolina
2016

APPROVED BY:

_______________________________
Dr. Joshua G. Pierce
Committee Chair

_______________________________
Dr. David A. Shultz

_______________________________
Dr. Christian Melander

_______________________________
Dr. Jonathan S. Lindsey
Ця робота присвячена моїй родині: батькам- Ользі та Василю - за підтримку та заохочення до академічних злетів; бабусі та дідуся - Стефанії та Володимиру - за те, що прививали любов до віршів Тараса Шевченка; моєму брату - Ігореві - за його взірцевий сильний характер. Я також присвячую цю роботу своїй вчителці хімії - Наталії Леонтівні Масник- за її невтомну працю з учнями і всі ті хімічні олімпіади, до яких вона мене готувала. Ці дорогі мені люди відіграли неабияку роль на шляху до знань і без них я ніколи б не змогла стати доктором Шиманською.

This work is dedicated to the members of my family: my Parents, Olga and Vasyl, for providing me with support and encouragements for my academic achievements; my Grandparents, Stefania and Volodymyr, for teaching me many Taras Shevchenko’s poems; and my Brother, Igor, for his inspiring strong character. I would also like to dedicate this work to my school chemistry teacher - Natalia Leontivna Masnyk, for her tireless work with students and all the chemistry Olympiads she helped me to prepare for. They make me who I am today and without them I could never have become Dr. Shymanska.
BIOGRAPHY

Nataliia Vasylivna Shymanska was born on April 16, 1989 in the small town of Zolochiv, on the west of Ukraine (Lviv Region) to the family of a mechanic and seamstress. Nataliia grew up in Zolochiv where she attended high school №1 from which she graduated with multiple honors. Throughout her school years she enjoyed studying science and competed in numerous Olympiads in Physics, Mathematics and Chemistry. In August of 2006, Nataliia moved to Kyiv to pursue her science education at the Department of Chemistry of Kyiv National Taras Shevchenko University. There, she explored chemical reactivity of isocoumarines and related flavonoid systems in the natural product research group led by Professors Volodymyr P. Khilya and Valentyna V. Ishchenko. After obtaining a Bachelor’s degree in Chemistry in June of 2010, she transitioned to the Institute of High Technologies at the Kyiv National University to pursue an interdisciplinary Master’s degree with a focus on bioorganic chemistry under the supervision of Prof. Igor V. Komarov. During that time, Nataliia was also receiving additional training in organic synthesis and worked on medicinal chemistry projects at the Institute of Bioorganic and Petroleum Chemistry (National Academy of Sciences of Ukraine, Kyiv). In June of 2011, Nataliia was awarded a Fulbright Scholarship to pursue a Ph.D. degree at the department of Chemistry at North Carolina State University, USA. There, she joined the research group of Prof. Joshua Pierce and worked on the total synthesis of antimicrobial natural products and the development of novel methods for the synthesis of functionalized heterocycles and amino acids. In December of 2015 she received Marie Curie Fellowship to continue her academic journey as postdoctoral fellow (EIPOD) at the European Molecular Biology Laboratory in Heidelberg, Germany.
ACKNOWLEDGMENTS

I would like to acknowledge my Ph.D. advisor Dr. Joshua Pierce for his time and valuable support, immediate help and his commitment to promote scientific curiosity. I am very thankful to Dr. Pierce for enabling my sometimes ambitious research ideas and inspiring scientific discussions as well as providing resources and funding. Next, I would like to acknowledge the members of my advisory committee, Drs. David Shultz, Christian Melander, and Jonathan Lindsey, for contributing to my education in chemistry through classes and discussions. I am thankful to Dr. Shultz for having confidence in my growth as a scientist and his inspirational teaching charisma. Also, thanks to Dr. Lindsey and graduate school representative Dr. Matthews for being scrupulous in reviewing my preliminary defense document and their comprehensive feedback on corrections. I would also like to acknowledge Dr. Christian Melander for discussion of antimicrobial testing results and his research group members, especially Dr. Roberta Melander and Dr. Robert Furlani, for sharing their experience in working with bacteria and teaching me how to perform various antimicrobial assays. I would also like to acknowledge Dr. Comins for providing invaluable practical synthetic advice and allowing me to borrow reagents on Saturday afternoons. Thank you Dr. Sommer, Dr. Sun, and Danielle Lehman for help with X-Ray analysis, managing important NMR experiments and obtaining high resolution mass-spectra as well as discussions valuable for the characterization of my compounds.

I am very thankful to Pierce group members, especially Yasamin Moazami, Berenice Lewandowski-Lemercier and Yunlong Shi for being kind and hard working colleagues with whom I went through all challenges of graduate school. I am very thankful to my friends
Xingci Situ, Nikolette Lucas McCombs (Ghiladi group) and Monica Enamorado (Comins group) for coffee break discussions and sharing their encouragement and support. I am also thankful to my friends from Raleigh cycling and running community for sharing over six thousand stress-relieving miles together on the bike and on foot exploring North Carolina.

Lastly, I am much obliged to my boyfriend Harris Bassett and his family: Denise and Bill Doucettes, Carlton Bassett and Natalie Harris for sharing their love and family warmth that helped me stay balanced through life’s ups and downs during Ph.D.. They have made my life in the USA more enjoyable and meaningful.
TABLE OF CONTENTS

LIST OF TABLES .............................................................................................................. xi

LIST OF FIGURES ............................................................................................................ xiii

LIST OF SCHEMES .......................................................................................................... xiv

LIST OF ABBREVIATIONS ............................................................................................... xix

CHAPTER 1: General Introduction ...................................................................................... 1

1.1 Purpose and Scope ....................................................................................................... 2

1.2 References .................................................................................................................... 5

CHAPTER 2: Natural Products as a Driving Force for Chemical and Biological Discovery .......................................................................................................................... 6

2.1 Abstract ....................................................................................................................... 7

2.2 Introduction and Background ...................................................................................... 7

  2.2.1 Importance of Natural Products ........................................................................... 7

  2.2.2 Antimicrobial Natural Products and the Threat of Antibiotic Resistnace .......... 10

  2.2.3 Marine Natural Products ..................................................................................... 16

      2.2.3.1 Isolation and Biological Activity of Synoxazolidinones A-C ............... 19

      2.2.3.2 Isolation and Biological Activity of Lipoxazolidinones A-C .............. 23

2.3 Conclusions and Outlook ........................................................................................... 24

2.4 References ................................................................................................................... 26

CHAPTER 3: Previous Syntheses and Applications of Oxazolidine-4-ones .................. 29

3.1 Abstract ....................................................................................................................... 30
3.2 Introduction and Background ........................................................................................................... 30

3.2.1 Classification and Nomenclature of Oxazolidinones ................................................................. 30

3.2.2 Previous Syntheses of 2,5-dialkyloxazolidin-4-ones of Type I ........................................... 33

3.2.3 Previous Syntheses of 5-alkylidene-2-alkyloxazolidin-4-ones of Type II ... 45

3.2.4 Previous Syntheses of 2-alkylidene-5-alkyloxazolidin-4-ones of Type III. 48

3.3 Conclusions and Outlook ...................................................................................................................... 50

3.4 References ..................................................................................................................................................... 51

CHAPTER 4: Our Approaches to 5-alkylidene-2-alkyl-4-oxazolidinones ...... 53

4.1 Abstract ........................................................................................................................................................... 54

4.2 Introduction and Background ............................................................................................................. 54

4.2.1 Oxazolidin-4-one Containing Natural Products ........................................................................ 54

4.2.2 Synthesis and Properties of N-acyliminium Ions ................................................................... 56

4.2.3 Our Approaches to 4-oxazolidinones .................................................................................... 59

4.3 Results and Discussion .............................................................................................................................. 60

4.3.1 Synthesis of Arylpyruvic Acids and their Amides ................................................................. 60

4.3.2 Acid Promoted Cyclization/Dehydration Cascade of Primary Keto Amides and Aldehydes ......................................................................................................................................................... 64

4.3.2.1 Cyclization/Dehydration of Unsubstituted Aliphatic Aldehydes ........................................ 64

4.3.2.2 Cyclization/Dehydration of Substituted Aliphatic Aldehydes ... 67

4.4 Conclusions and Outlook ....................................................................................................................... 74

4.5 Experimental ....................................................................................................................................................... 75
CHAPTER 5: First Total Synthesis of Synoxazolidinones A and B .......... 96

5.1 Abstract ......................................................................................... 97
5.2 Introduction and Background ......................................................... 97
5.3 Results and Discussions .................................................................. 99
   5.3.1 Acylation of N-substituted Aldimines with Activated α-keto-acid
       Derivatives ...................................................................................... 99
       5.3.1.1 Acylation of Diisobutylaluminum- and Trimethylsilyl-imines .. 100
       5.3.1.2 Acylation of Chiral N-tert-butanesulfinimines ...................... 102
       5.3.1.3 Acylation of N-alkylimines ............................................... 110
   5.3.2 The First Total Synthesis of Synoxazolidinones A and B .............. 115
   5.3.3 Studies Towards Total Synthesis of Synoxazolidinone C ............... 120

5.4 Conclusions and Outlook ................................................................. 125
5.5 Experimental .................................................................................... 126
5.6 References ....................................................................................... 149

CHAPTER 6: Antimicrobial Activity of 4-oxazolidinones .................... 152

6.1 Abstract .......................................................................................... 153
6.2 Introduction and Background .......................................................... 153
6.3 Results and Discussions .................................................................. 155
   6.3.1 MIC Values of 4-oxazolidinones Against Pathogenic Bacteria ....... 155
   6.3.2 Biofilm Inhibition Properties of 4-oxazolidinones ....................... 162
CHAPTER 7: Synthesis and Application of 1,5-dihydro-2H-pyrrol-2-ones as Precursors for Novel Highly Functionalized N-containing heterocycles and β-amino acids

7.1 Abstract

7.2 Introduction and Background

7.3 Results and Discussions

7.3.1 Optimization of the Synthesis of Alkyl Substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones

7.3.2 An Entry to Pretazettine via Functionalized 1,5-dihydro-2H-pyrrol-2-ones

7.3.3 Attempts at Asymmetric Synthesis of Alkyl Substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones

7.3.4 Synthesis of Novel 4-allyl-pyrroliidine-2,3-diones

7.3.4.1 Claisen Rearrangement of 3-(allyloxy)-1,5-dihydro-2H-pyrrol-2-ones

7.3.4.2 Tsuji-Trost Allylation of 3-(hydroxy)-1,5-dihydro-2H-pyrrol-2-ones

7.3.5 Properties of Novel 4-allyl-pyrroliidine-2,3-diones

7.4 Conclusions and Outlook

7.5 Experimental

7.6 References
7.3.5.1 Synthesis of Novel $\beta^{2,2,3}$-Amino Acids Containing Quaternary Stereocenter ................................................................. 213

7.3.5.2 Synthesis of Highly Substituted Polycyclic $N$-heterocycles Containing Quaternary Stereocenter ................................................................. 216

7.4 Conclusions and Outlook .................................................................................................................................................................................. 218

7.5 Experimental ................................................................................................................................................................................................ 219

7.6 References ................................................................................................................................................................................................ 274
LIST OF TABLES

Table 2.1: Introduction of classes of antibacterial agents with new mechanisms of action. 11

Table 4.1: Attempts at synthesis of 2-oxo-3-phenylpropanamide. 61

Table 4.2: Acid promoted debenzylation of 4-12 obtained via coupling of phenylpyruvic acid with 2,4-dimethoxybenzylamine. 63

Table 4.3: Substrate scope for amide coupling/DMB cleavage protocol. 64

Table 4.4: Acid concentration screen for 4-oxazolidinone formation via dehydration/cyclization approach. 65

Table 4.5: Substrate scope for dehydration/cyclization reaction. 66

Table 4.6: Dehydration of 2-oxo-3-phenylpropanamide 4-11 with enolizable aldehydes 4-28, 4-16. 67

Table 4.7: Acid mediated dehydration/cyclization of 2-oxo-3-phenylpropanamide 4-11 with guanidine containing aldehydes 4-30 and 4-31. 70

Table 4.8: Dehydration of 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanamide (4-15g) with aldehydes 4-30 and 4-31. 73

Table 5.1: Acylation of N-tert-butanesulfinimines 5-23 and 5-24 with phenylpyruvic acid chloride 5-17. 104

Table 5.2: Reaction of N-tert-butanesulfinimine 5-23 with various phenylpyruvic acid activated species. 106

Table 5.3: Diastereoslectivity of the N-alkyl imine acylation/enol addition. 113

Table 5.4: Attempts at isomerization of cyclic amides 5-81 and 5-82 to the corresponding enamides 5-83 and 5-84. 123

Table 6.1: Antimicrobial activity of guandine-containing 4-oxazolidinones. 156

Table 6.2: Antimicrobial activity of commercially available antibiotics. 157

Table 6.3: Antimicrobial activity of aliphatic 4-oxazolidinones containing varied aroamatic substituent. 158

Table 6.4: Antimicrobial activity of miscellaneous 4-oxazolidinones. 160
Table 6.5: Effects of synoxazolidinone A (2-26) on adhesion and growth of fouling marine microorganisms
........................................................................................................................................... 164

Table 6.6: MIC and IC_{50} values of 4-oxazolidinone against MRSA. ........................................... 169

Table 6.7: Effect of media on MIC of methicillin in presence of varied amounts of 5-60. 173

Table 7.1: Formal [3+2] cycloaddition between pyruvates and imines. .................... 187

Table 7.2: Reductive Heck reaction of O-beznyl-1,5-dihydro-2H-pyrrol-2-one 7-69 for the synthesis of the model [6.5] ring system of pretazettine. ................................................................. 195

Table 7.3: C-alkylation of 1,5-dihydro-2H-pyrrol-2-ones via Tsuji-Trost reaction. ......... 211
LIST OF FIGURES

Figure 2.1: Examples of biologically active natural products isolated from various plants... 8

Figure 2.2: Representative examples of antibiotics derived from natural products. .......... 13

Figure 2.3: Timeline of antibiotic introduction and resistance........................................ 14

Figure 2.4: Antibiotic resistance mechanisms. ................................................................. 15

Figure 2.5: Bioactive natural products isolated from various marine species ................ 18

Figure 2.6: Synoxazolidinone family of natural products and their biological activity. ..... 19

Figure 2.7: Family of lipoxazolidinone natural products and their biological activity .......... 23

Figure 4.1: Natural products containing a 4-oxazolidinone heterocycle with unsaturation at C2 or C5.......................................................... 55

Figure 6.1: Inhibition of MRSA biofilm formation by 2-26.............................................. 166

Figure 6.2: Inhibition of MRSA biofilm formation by 5-59.............................................. 166

Figure 6.3: Inhibition of MRSA biofilm formation by 4-22.............................................. 167

Figure 6.4: Inhibition of MRSA biofilm formation by 5-64.............................................. 167

Figure 6.5: Inhibition of MRSA biofilm formation by 5-61.............................................. 168

Figure 6.6: Inhibition of MRSA biofilm formation by 5-63.............................................. 168

Figure 6.7: Inhibition of MRSA biofilm formation by 5-60.............................................. 169

Figure 6.8: Time-kill curves for MRSA (BAA 44) with 5-60............................................. 171

Figure 6.9: Resensitization of MRSA to methicillin by 5-60........................................... 172

Figure 6.10: Effects 4-oxazolidinones on protein synthesis.......................................... 174

Figure 7.1: Examples of naturally occurring biologically active dihydropyrrolones...... 184

Figure 7.2: Representative members of the crinine class of Amaryllidaceae alkaloids .... 189
LIST OF SCHEMES

Scheme 2.1: Key observations in the synthesis of vitamin B\textsubscript{12} which drove Woodward’s motivation to investigate the mechanism of pericyclic reactions. ................................................. 9

Scheme 2.2: Proposed biosynthetic route to synoxazolidinones A, B and C......................... 22

Scheme 2.3: Degradation pathways for lipoxazolidinone A................................................ 24

Scheme 3.1: Structures and nomenclature of oxazolidine-4-ones. ................................... 31

Scheme 3.2: Synthesis of optically active $\alpha$-hydroxy acids and diols from oxazolin-4-one derivatives. ............................................................................................................. 31

Scheme 3.3: Synthesis of 4-oxazolidinones from $O$-acyl-mandelamides......................... 32

Scheme 3.4: Synthetic application of the diastereoselective 4-oxazolidinone ring-opening towards synthesis of (+)-tashiromine................................................................. 32

Scheme 3.5: Retrosynthetic disconnections 2,5-dialkyl-4-oxazolidinones and common starting materials for their synthesis. ................................................................. 33

Scheme 3.6: Fischer’s oxazole synthesis and its extension. ................................................ 34

Scheme 3.7: Synthesis and relative configuration of oxazolidine-4-ones obtained via condensation of lactamide 3-15 with various aldehydes. ....................................................... 34

Scheme 3.8: Cyclocondensation of cyanohydrins with aldehydes and ketones. ............... 35

Scheme 3.9: Example of $p$TsOH mediated dehydration of $\alpha$-hydroxy amides with aldehydes for preparation of 4-oxazolidinones. ................................................................................. 36

Scheme 3.10: Example of Lewis acid mediated dehydration of $\alpha$-hydroxy amides with aldehydes for preparation of 4-oxazolidinones...................................................... 36

Scheme 3.11: Example of acid mediated dehydration of $\alpha$-hydroxy amides with $\alpha$-chloroethers for preparation of 4-oxazolidinones.................................................. 37

Scheme 3.12: One pot preparation of 4-oxazolidinones via addition to an $N$-acyliminium ion. ......................................................................................................................... 37

Scheme 3.13: Synthesis of 4-oxazolidinones from nitro- substituted cyclohexane.......... 38

Scheme 3.14: Substrate scope and proposed mechanism of enantioselective synthesis of 4-oxazolidinones via [3+2] cycloaddition................................................................. 39
Scheme 3.15: The enantioselective synthesis of α-disubstituted 4-oxazolidinones via palladium-catalyzed allylic alkylation. .......................................................... 40

Scheme 3.16: Diastereoselective synthesis of 4-oxazolidinones from O-acyl-mandelamides. .................................................................................................................................................................................. 41

Scheme 3.17: Challenge in ring closing metathesis of acyclic amides ......................... 41

Scheme 3.18: Oxidative cleavage of 2-azetidinones .......................................................... 42

Scheme 3.19: Light promoted transformations of secondary α-keto-amides leading to 4-oxazolidinones .................................................................................................................................................. 42

Scheme 3.20: Access to 4-oxazolidinones via ring modification of oxazoles .................. 43

Scheme 3.21: Intramolecular cyclizations leading to 4-oxazolidinones ......................... 44

Scheme 3.22: Unsaturated 5-alkylidene-2-alkyl-4-oxazolidinones and common precursors for their synthesis ........................................................................................................................................ 45

Scheme 3.23: Synthesis of 4-oxazolidinone 3-89 via reduction of oxazolone 3-88 .......... 45

Scheme 3.24: Synthesis of (Z)-5-benzyldiene-2-phenyloxazol-4-(5H)-one (3-88) .............. 46

Scheme 3.25: Synthesis of 5-alkylidene-2-alkyl-4-oxazolidineone 3-96 and Diels-Alder reaction leading to formation of 2,5-dialkyloxazolidin-4-ones 3-98 and 3-99 ....................... 47

Scheme 3.26: Photooxygenation of the 4-pyrrolin-3-one resulting in 4-oxazolidinone formation.................................................................................................................................................................................. 48

Scheme 3.27: Photolysis of secondary α-ketoamides leading to 4 oxazolidinones ............. 48

Scheme 3.28: Synthesis of fused oxazolidin-4-ones via dipolar cycloaddition reactions. ... 49

Scheme 4.1: Proposed transformation to prepare 4-oxazolidinones ............................... 56

Scheme 4.2: Common precursors to preparation of N-acyliminium ions in situ ............ 57

Scheme 4.3: Relative electrophilicity of various iminium ions ........................................... 58

Scheme 4.4: Example of intramolecular addition to an iminium ion ............................... 58

Scheme 4.5: Approaches to 4-oxazolidinones developed in our laboratory .................. 59

Scheme 4.6: Preparation of N², N³-Bis(tert-butoxycarbonyl)-N¹-(3-chloro-4-oxybutyl)guanidine (4-31). .............................................................................................................................................................................. 69
Scheme 4.7: Synthesis of 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanoic acid (4-13g) and its primary amide 4-15g. .............................................................. 72

Scheme 5.1: Potential addition pathways for acyliminium intermediate 4-1. ......................... 97

Scheme 5.2: Synthesis of 3-hydroxy-1,5-dihydro-2H-pyrrol-2-one 5-8.................................. 98

Scheme 5.3: Possible origin of lack of the diastereoselectivity and low reaction yields for α-substituted aldehydes in our previous approach described in Chapter 4. ......................... 99

Scheme 5.4: Synthesis of N-silylimines. .................................................................................. 100

Scheme 5.5: Acylation of N-silylimines with phenylpyruvic acid chloride 5-17. ................. 101

Scheme 5.6: Acylation of N-diisobutylaluminium imine with phenylpyruvic acid chloride. ........................................................................................................ 102

Scheme 5.7: Preparation of N-tert-butanesulfinimine 5-23 and 5-24. ................................. 103

Scheme 5.8: Proposed mechanism of loss of the auxiliary during the acylation of chiral tert-butanesulfinyl aldimines. ............................................................... 107

Scheme 5.9: Reaction of phenylpyruvic acid with Ghosez’s reagent (5-27) ......................... 107

Scheme 5.10: Synthesis of synoxazolidinone B via acylation of guanidine containing N-tert-butanesulfinimine 5-34. ................................................................. 108

Scheme 5.11: Attempts at the synthesis of synoxazolidinone A via acylation of guanidine containing N-tert-butanesulfinimine 5-36................................................. 109

Scheme 5.12: Synthesis of β-lactams via [2+2] cycloaddition of benzylaldimines and ketenes derived from acid chlorides. ......................................................... 110

Scheme 5.13: Chemoselectivity of N-alkyl imine acylation/enol addition. ......................... 111

Scheme 5.14: Proposed transition state of N-acyliminium ion intermediate leading to the formation of the major diastereomer of 4-oxazolidinone 5-54c. ......................... 114

Scheme 5.15: Deprotection of 2,4-DMB oxazolidin-4-one 5-45c........................................ 115

Scheme 5.16: Total synthesis of synoxazolidinone B........................................................... 116

Scheme 5.17: The first total synthesis of synoxazolidinone A. ............................................. 117

Scheme 5.18: Small library of 4-oxazolidinones containing α-Cl and terminal guanidine substitution............................................................... 119
Scheme 5.19: Retrosynthetic analysis of synoxazolidinone C .......................................................... 121

Scheme 5.20: Synthesis of cyclic amides 5-81 and 5-82 as isomerization precursors. ...... 122

Scheme 5.21: Model halolactonization studies................................................................. 125

Scheme 6.1: Sites of modification in our 4-oxazolidinone library. ................................. 155

Scheme 6.2: Antimicrobial activity of guanidine-containing 4-oxazolidinones (structures of compounds from Table 6.1). ................................................................. 157

Scheme 6.3: Antimicrobial activity of aliphatic 4-oxazolidinones containing varied aromatic substituent (structures of compounds from Table 6.3). .................................................. 159

Scheme 6.4: Antimicrobial activity of miscellaneous 4-oxazolidinones (structures of compounds from Table 6.4). ................................................................. 161

Scheme 6.5: Natural (6-12) and synthetic (6-13) biofilm inhibitors and methicillin........ 163

Scheme 7.1: Potential equilibrium in addition pathways of N-acyliminium intermediates 7-1 and 7-2 leading to various ratios of regioisomeric cyclization (C- vs O-) products 7-4 and 7-3 ................................................................................................................. 183

Scheme 7.2: O-protection and N-deprotection of dihydropyrrolones............................... 185

Scheme 7.3: O-protection and N-deprotection of dihydropyrrolones ............................... 189

Scheme 7.4: Substrate scope of O-allyl-1,5-dihydro-2H-pyrrol-2-ones. ......................... 189

Scheme 7.5: Limitations of our methodology: 1,5-dihydro-2H-pyrrol-2-ones inaccessible via standard conditions (Table 7.1) .................................................................................................................. 190

Scheme 7.6: Interconversion between 7-54, 7-51 and 7-52. ............................................ 192

Scheme 7.7: Retrosynthetic analysis of our approach to pretazettine (7-51) ................. 192

Scheme 7.8: Synthesis of model substrates bearing key functional moieties required for synthesis of pretazettine. ...................................................................................... 193

Scheme 7.9: Overman’s synthesis of 6a-epipretazettine via an intramolecular Heck reaction. .................................................................................................................. 194

Scheme 7.10: Possible products in our reductive Heck approach (7-79 desired, 7-80, 7-81 undesired) .............................................................................................. 196
Scheme 7.11: Observed reactivity during condensation of 2,4-DMBA, enolizable aldehydes and phenyl pyruvic ester. ................................................................. 198

Scheme 7.12: Proposed mechanisms for the formation 1,5-dihydro-2H-pyrrol-2-one....... 200

Scheme 7.13: Asymmetric entry to N-tosyl aryl-containing pyrrolones 7-26. ............... 201

Scheme 7.14: Conditions and chiral reagents explored when attempting asymmetric synthesis of 5-43. ............................................................................. 203

Scheme 7.15: Chiral phenylpyruvic derivatives explored in attempts towards the asymmetric synthesis of 5-43. ............................................................................. 205

Scheme 7.16: Chiral amine derivatives explored in attempts at diastereoselective preparation of 7-48. ................................................................. 206

Scheme 7.17: Claisen rearrangement of 3-(allyloxy)-1,5-dihydro-2H-pyrrol-2-ones .... 208

Scheme 7.18: Novel 4-allyl-pyrrolidine-2,3-diones via Claisen rearrangement of O-allyl-1,5-dihydro-2H-pyrrol-2-ones................................................................. 209

Scheme 7.19: Removal of DMB-protecting group. .......................................................... 212

Scheme 7.20: Designation of amino acids. .................................................................... 214

Scheme 7.21: Our approach to novel β²,³-amino acids. ................................................ 214

Scheme 7.22: Synthesis of novel β²,³-amino acids via ring opening of 4-allyl-pyrrolidine-2,3-diones ................................................................................. 215

Scheme 7.23: Gram-scale synthesis of β²,³-amino acid 7-105a ...................................... 216

Scheme 7.24: Ring-closing metathesis reaction of 4-allyl-pyrrolidin-2,3-diones for the synthesis of N-containing polycyclic compounds................................. 217
LIST OF ABBREVIATIONS

A2058: melanoma cell line
Ac: Acetyl
Ac-D-Ala-D-Ala: Acetyl-D-alanyl-D-alanine
Ac₂O: Acetic anhydride
AcOH: Acetic acid
AllylOAc: Allyl acetate
4 Å MS: Molecular sieves, 4 Å
3 Å MS: Molecular sieves, 3 Å
aq.: Aqueous
BINAP: 2,2'-bis(Diphenylphosphino)-1,1'-binaphthyl
BINOL: 1,1'-Bi-2-naphthol
BNPPA: 1,1'-Binaphthalene-2,2'-diyl hydrogen phosphate
Boc: tert-Butyloxycarbonyl
BOX: Bisoxazoline
BRSM: Based on the recovery of the starting material
Bz: Benzoyl
CAN: Ceric ammonium nitrate
cat.: Catalytic
CDI: Carboxyldiimidazole
CFU: Colony-forming unit
CLSI: Clinical and Laboratory Standards Institute
COD: 1,5-Cyclooctadiene
CyHex: Cyclohexyl
DABCO: 1,4-Diazabicyclo[2.2.2]octane
DBU: 1,8-Diazabicycloundec-7-ene
DCC: N,N'-dicyclohexylcarbodiimide
DCM: Dichloromethane
DDQ: 2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
DIBAL-H: Diisobutylaluminum hydride
DIEPA: N,N'-Diisopropylethylamine
DMAD: Dimethyl acetylenedicarboxylate
DMAP: N,N'-Dimethyl-4-aminopyridine
DMB: Dimethoxybenzyl
DMBA: Dimethoxybenzyl amine
2,4-DMBA: 2,4-Dimethoxybenzyl amine
DME: 1,2-Dimethoxyethane
DMF: Dimethylformamide
DMSO: Dimethylsulfoxide
DMXBA: 3-(2,4-dimethoxybenzyldiene)-anabaseine
DNA: Deoxyribonucleic acid
DPPE: 1,2-Bis(diphenylphosphino)ethane
dr: Diastereomeric ratio
DTBMP: 2,6-Di-tert-butyl-4-methylpyridine
Δ: Heat

ee: Enantiomeric excess

EDCI, EDC: 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide

EPS: extracellular polymeric substance

EWG: Electron withdrawing group

equiv.: Equivalent

FDA: Food and Drug Administration

h: Hour

hv: Light

HCT-116: Human colon carcinoma cell line

HESI: heated electrospray ionization

Hex: Hexane

HMDS: Hexamethyldisilazane

HOBt: Hydroxybenzotriazole

HOMO: Highest occupied molecular orbital

HRMS: High-resolution mass spectrometry

HT-29: Colon carcinoma cell line

HTS: High-throughput screening

HPLC: High pressure liquid chromatography

LAH: Lithium aluminium hydride

LB: Lewis base

LC: Liquid chromatography
LG: Leaving group
LLC: Lewis lung carcinoma
LC-MS: Mass spectrometer coupled liquid chromatography
LiHMDS: Lithium hexamethyldisilazide
L-Pro: L-Proline
MCF-7: Breast adenocarcinoma cell line
mCPBA: meta-Chloroperoxybenzoic acid
MCR: Multicomponent reaction
MDR: Multi-drug resistance
MHB: Mueller Hinton broth
MIC: Minimum Inhibitory Concentration
MRC-5: Lung fibroblast cell line
MRSA: Methicillin-resistant Staphylococcus aureus
MW: Microwave
N-Ac-Gly: N-Acetylglycine
NBS: N-Bromosuccinimide
NCS: N-Chlorosuccinimide
ND: Not determined
NMM: N-methylmorpholine
NMR: Nuclear magnetic resonance
NR: No reaction
Nu: Nucleophile
MS: Mass spectrometry

[O]: Oxidation

OAc: Acetate

OD: Optical density

P388: Murine leukemia cell line

PBB: para-Bromobenzyl

PCC: Pyridinium chlorochromate

Piv: Pivaloyl

pmdba: Bis(4-methoxybenzylidene)acetone

PTLC: Preparative thin layer chromatography

$p$TsOH: para-Toluene sulfonic acid

Py: Pyridine

pyBOP: Benzo(1-yl-oxytrypyrrolidinophosphonium hexafluorophosphate

quant.: Quantitative

RCM: Ring-Closing Metathesis

RNA: Ribonucleic acid

rt: Room temperature

SAR: Structure-activity relationship

SM: Starting material

TADDOL: (4R,5R)-4,5-Bis(diphenylhydroxymethyl)-2,2-dimethyldioxolane

TBACl: Tetrabutylammonium chloride

TBS, TBDMS: tert-Butyldimethylsilyl
TBSOTf: tert-Butyldimethylsilyl trifluoromethanesulfonate
TEA: Triethylamine
Tf: Triflyl
TFA: Trifluoroacetic Acid
TFE: Trifluoroethanol
THF: Tetrahydrofuran
TLC: Thin layer chromatography
TMS: Trimethylsilyl
TMSCl: Trimethylsilyl chloride
T3P: Propylphosphonic anhydride
Ts: Tosyl
TSBG: Tryptic soy broth supplemented with glucose
UV: Ultraviolet
vs.: Versus
v/v: Volume ratio
CHAPTER 1

General Introduction

In a time when one can generally state that given enough resources and the best of human beings in terms of creativity and fortitude any molecule can be prepared, why does the field of complex molecule synthesis require further development? Why is there a need for small tweaks on reactions that have been developed years earlier or additional solutions to functional group manipulations? The answer is that in fact we do not have the methods to deliver practical approaches for the synthesis of the remarkable structures which Nature continuously provides us with.¹ These structures bear characteristics of a high chemical diversity and biochemical specificity, making them a favorable source of chemical probes and lead compounds for drug discovery.²
1.1 Purpose and Scope

Heterocycles are often the core structural fragments of biologically active compounds and natural products. The overwhelming prevalence of nitrogen and oxygen containing heterocycles amongst pharmaceutical agents is associated with their capabilities to mimic interactions of important biological targets such as proteins, nucleic acids and carbohydrates. Structural diversity of heterocyclic compounds is also implicated in the large arsenal of synthetic procedures for their preparation. New ways to assemble biologically relevant nitrogen containing heterocycles are always needed in order to update and expand the natural product chemists’ toolkit.

Natural products have inspired chemists through their design and synthesis to open doors to new opportunities in chemical biology, medicine, and many other scientific disciplines that rely on new molecules and molecular insight for future innovation. This symbiotic relationship between natural product synthesis and new methods development is demonstrated in this dissertation. Our aim is to develop new methods for the synthesis of rare nitrogen and oxygen containing heterocycles present in novel biologically active natural products. Driven by the goal to pursue the first total synthesis of synoxazolidinones we focused our efforts on new disconnections around 4-oxazolidinone ring.

Chapter 2 will describe the importance of natural products in antimicrobial drug discovery. The paradigm of resistance of pathogenic bacteria to antibiotics will be discussed. The need for the discovery of new sources of chemical diversity and small molecule scaffolds in order to overcome a threatening burden of resistance of bacteria to antibiotics
will be highlighted. Novel marine antimicrobial natural products, namely synoxazolidinones and lipoxazolidinones, will be introduced in the light of new core 4-oxazolidinone heterocycle for antimicrobial drug discovery. Chapter 3 will reveal significance and current knowledge about 4-oxazolidinone containing compounds, methods for their synthesis, applications and scope. In Chapter 4, we will discuss our ideas and efforts directed towards the rapid construction of 4-oxazolidinone ring via N-acyliminium intermediates employing dehydration/cyclization cascade. Optimization of cyclization conditions for two alternative strategic disconnections to obtain 4-oxazolidinones pushed us to explore a variety of the aryl pyruvic acid derivatives and imines and their reactivity will be also discussed in Chapter 4. Our exploration of the tandem N-acyliminium ion formation/enol addition culminated in the first total synthesis of synoxazolidinones A and B via imine acylation/cyclization cascade will be described in Chapter 5.

By utilizing our newly developed methods, we were able to generate a wide variety of 4-oxazolidinone containing analogues of synoxazolidinones. In Chapter 6 we describe biological studies focused on antimicrobial structure activity relationships of the synthesized library of 4-oxazolidinones against clinically relevant bacteria (primarily against Methicillin-resistant *Staphylococcus aureus*). We demonstrated that promising levels of antimicrobial activity against MRSA can be achieved by greatly simplified 4-oxazolidinone containing analogues. In Chapter 7 we discuss the discovery of an alternative cyclization pathway during 4-oxazolidinone ring formation. We observed that certain conditions also yielded regioisomeric 3-hydroxy-1,5-dihydro-2H-pyrrolones, a class known medicinally relevant heterocycles. Chapter 7 will describe our efforts to switch the reaction selectivity for the
exclusive formation of these regioisomeric heterocycles. Therein we will discuss reactivity of stable 3-allyloxy-1,5-dihydro-2H-pyrrolones and their unexplored sigmatropic rearrangement chemistry. An optimized methodology for the synthesis novel pyrrolidine-2,3-diones containing quaternary stereocenters will be also described in Chapter 7 as well as their use in the synthesis of unprecedented $\beta^{2,2,3}$ amino acids and sterically crowded $N$-containing fused heterocycles.
1.2 References


CHAPTER 2

Natural Products as a Driving Force for Chemical and Biological Discovery
2.1 Abstract

In this chapter the importance of natural products as a guide-post for chemical and biological discovery is highlighted. The role of natural products in antimicrobial drug development is discussed in the light of antibiotic resistance. Examples of novel marine natural products as a promising sources of structural and functional diversity are provided. The biological activity of rare oxazolidinone containing marine natural products is described followed by the introduction of our synthesis targets - synoxazolidinone A and B.

2.2 Introduction and Background

2.2.1 Importance of Natural Products

Historically, natural products derived from plants, animals and bacteria were the source of all medicinal compounds used by humans to treat illnesses and the revisiting of natural products for drug discovery continues today. Natural products are increasingly employed in clinical trials or provide advanced leads for compounds which entered clinical trials, especially as anticancer and antimicrobial agents. Since the 1940’s to date, 131 out of 175 small molecule scaffolds of anticancer drugs are natural products or molecules that have been inspired by natural products. The success of these molecules arises from the fact that natural products possess structural and chemical diversity which is unsurpassed by synthetic libraries and have evolved in the presence of complex architectures of biological molecules. Altogether, more than 200,000 compounds have already been isolated from terrestrial sources such as plants, animals and microorganisms.¹ The remarkable diversity of structures
and activities of plant-derived drugs (Figure 2.1) are represented by examples of vinblastine (2-1), camptothecin (2-2), morphine (2-3) and paclitaxel (2-4).²

**Figure 2.1:** Examples of biologically active natural products isolated from various plants.

Total synthesis of structurally complex and biologically active natural products has been a major stimulus for the progress in organic chemistry. Synthetic plans even towards simple natural products often lead to serendipitous discoveries of novel reactivity and structures. In addition, many innovations and tweaks of existing synthetic methodologies have been made in the pursuits of natural products’ core structures. As a result, the capacity of organic synthesis to deal with molecules of considerable complexity has increased significantly in recent years.
Not only have natural products contributed to drug discovery, they have also served useful as substrates for the investigation of reaction mechanisms. The fundamental concept of the conservation of molecular orbital symmetry in pericyclic reactions was first explained with evidence that originated from Woodward’s synthetic work towards vitamin B$_{12}$ (2-10) (Scheme 2.1). To approach intermediate 2-6 Woodward had planned to utilize an intramolecular Michael addition which was envisioned to proceed via 2-7 upon treatment of 2-5 with base; however, this transformation did not take place under basic conditions.

Scheme 2.1: Key observations in the synthesis of vitamin B$_{12}$ which drove Woodward’s motivation to investigate the mechanism of pericyclic reactions.

When 2-5 was heated at its melting point temperature the opposite diastereomer 2-8 of the product formed via disrotatory ring closure. When treated with light, 2-8 underwent ring
opening to form alkene 2-9 which appeared to have the opposite double bond geometry relative to 2-5.

When subjected to thermal cyclization conditions intermediate 2-9 proceeded to give desired diastereomer 2-6. After consideration of similar experiments performed by other scientists in the field⁸,⁹ Woodward noticed that symmetry elements of the starting material HOMO were conserved in the product molecular orbitals, and therefore he proposed that symmetry had to be conserved in the transition state. In 1964, together with computational chemist Roald Hoffmann, Woodward developed and justified by additional experiments a very important set of rules, now used to predict the outcome of pericyclic reactions and for which the Nobel Prize was awarded in 1981.¹⁰

2.2.2 Antimicrobial Natural Products and the Threat of Antibiotic Resistance

Chemical synthesis, along with biological evaluation of natural products and their analogues, followed by the elucidation of the mechanism by which these molecules function in biological systems, represents an exciting area of chemical biology. The high selectivity of natural products is a result of specific non-covalent or covalent interactions with their biological targets. For example, very often natural products incorporate reactive structural moieties, such as α,β-unsaturated carbonyl groups or lactams, and therefore can irreversibly modify proteins. A famous example is the antibiotic penicillin which covalently inhibits formation of peptidoglycan crosslinks in the bacterial cell wall.

Antimicrobial natural products are weapons utilized by various species in the fight for life, space and resources. Microorganisms have evolved these secondary metabolites in the
presence of their targets (provided by coexistence with other competitor or aggressor microorganisms) as a means of protection. In fact, an enormous success of natural products as leads for new drugs can be observed in the antimicrobial drug development field. It has been roughly estimated that in the areas of cancer and infectious disease, 75% of FDA approved pharmaceutical agents are natural products and/or their derivatives.11 Interestingly, many of these molecules have reached clinical trials without substantial structural modifications (Table 2.1). Representative structures of antibiotics derived from natural products are shown on Figure 2.2.

**Table 2.1:** Introduction of classes of antibacterial agents with new mechanisms of action.

<table>
<thead>
<tr>
<th>Year</th>
<th>Class</th>
<th>Target</th>
<th>Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>1935</td>
<td>Sulfonamides (synthetic)</td>
<td>Folate pathway - pterate synthetase</td>
<td>Prontosil (2-11)</td>
</tr>
<tr>
<td>1940</td>
<td>β-lactams</td>
<td>Cell wall - transpeptidase</td>
<td>Penicillin G (2-12)</td>
</tr>
<tr>
<td>1949</td>
<td>Polyketides</td>
<td>Protein biosynthesis - ribosomal A site</td>
<td>Tetracycline (2-13)</td>
</tr>
<tr>
<td>1949</td>
<td>Phenylpropanoids</td>
<td>Protein biosynthesis - peptidyl transferase</td>
<td>Chloramphenicol (2-14)</td>
</tr>
<tr>
<td>1950</td>
<td>Aminoglycosides</td>
<td>Protein biosynthesis - initiation complex</td>
<td>Tobramycin (2-15)</td>
</tr>
<tr>
<td>1952</td>
<td>Macrolides</td>
<td>Protein biosynthesis - translocation</td>
<td>Erythromycin A (2-16)</td>
</tr>
<tr>
<td>1958</td>
<td>Glycopeptides</td>
<td>Cell wall - Ac-D-Ala-D-Ala</td>
<td>Vancomycin (2-17)</td>
</tr>
<tr>
<td>1962</td>
<td>Quinolones (synthetic)</td>
<td>DNA replication - DNA gyrase</td>
<td>Ciprofloxacin (2-18)</td>
</tr>
<tr>
<td>1962</td>
<td>Streptogramins</td>
<td>Protein biosynthesis - 50S ribosomal subunit</td>
<td>Pristinamycins (2-19, 2-20)</td>
</tr>
<tr>
<td>2000</td>
<td>Oxazolidinones (synthetic)</td>
<td>Protein biosynthesis - 70S initiation complex</td>
<td>Linezolid (2-21)</td>
</tr>
<tr>
<td>2003</td>
<td>Lipopeptides</td>
<td>Bacterial membrane - phospholipids</td>
<td>Daptomycin (2-22)</td>
</tr>
</tbody>
</table>

Ref. (11)
Antibiotic chemotherapy is based on Paul’s Ehrlich concept of selective toxicity.\textsuperscript{12,13} Many antimicrobial drugs can successfully access fundamental differences between bacterial and host cells. In principle, there are many ways to achieve selective killing of bacteria due to their significantly different metabolism and increased rates of multiplication. Antibiotics can either kill bacteria (bacteriocidal effect) or inhibit their growth (bacteriostatic effect). Inhibition of bacterial growth can take place via several mechanisms, such as interfering with cell wall synthesis, perturbing DNA replication or RNA transcription, as well as stalling protein synthesis (Table 2.1). Antibiotics can also exert bacteriostatic action by affecting essential metabolic pathways in bacterial metabolism (for example the folic acid pathway). In practice, the bacterial cell wall is distinct in structure, whereas intracellular processes and vital structures such as ribosomes, DNA and cytoplasmic components are similar to mammalian systems. For this reason, some of the most successful antibiotics (\(\beta\)-lactams, vancomycin) to date are those that interfere with cell wall biosynthesis.
Figure 2.2: Representative examples of antibiotics derived from natural products.
Infections are overwhelmingly the leading cause of death among humans. According to the World Health Organization, about 26% of the 57 million deaths worldwide in 2002 were caused solely by infectious disease.\textsuperscript{14} Our large arsenal of antibiotics have posed a strong selective pressure on bacteria causing them to develop advanced defense mechanisms to resist toxic antibiotic action. In today’s growing movement of people and globalization of the economy the rate of spread of resistant pathogens is significantly higher than the ability of traditional pharmaceutical approaches to develop effective antimicrobial treatments (Figure 2.3).

\textbf{Figure 2.3:} Timeline of antibiotic introduction and resistance.

Resistance has been an issue ever since the introduction into clinical use of the first antimicrobial agents in the 1940s (Figure 2.3).\textsuperscript{15} During the 20 years following the introduction of the sulfonamides and β-lactams, scientists developed many new classes of antimicrobial compounds including the aminoglycosides, tetracyclines, nitroimidazoles, and
quinolones to treat resistant bacteria (Table 2.1). Since this time, the pharmaceutical industry has largely been exploring incremental structural changes within known antibiotic classes. The introduction of new classes of antibacterial drugs that possess a completely novel mechanism of action has slowed since the late 1960s. The last new class of antibiotics prior to 2000 (Linezolid) was approved in 1968 (Trimethoprim). The majority of antimicrobials introduced since that time have been modifications of previously discovered classes of drugs. Over time as the use of antimicrobial drugs has increased so has the level of bacterial resistance. To date, bacteria exist that are resistant to all known antibiotics and employ various, and sometimes multiple mechanisms of resistance (Figure 2.4).

**Figure 2.4: Antibiotic resistance mechanisms.**

To achieve resistance bacteria can lower the intracellular concentration of antibiotic by lowering membrane permeability, by reducing influx of the antibiotic or by increasing efflux of the antibiotic via specific protein pumps. Bacteria can also mutate or modify the
antibiotic target (or antibiotic itself) to eliminate selective interactions. Some bacteria have evolved to overproduce the target mimic which will scavenge active antimicrobial drug. It also uses factor associated protection of the binding site to avoid antibiotic. Drug degradation mechanisms by bacteria are also observed.\textsuperscript{20}

Bacterial infections continue to pose a substantial threat throughout the world because of the emergence of multidrug resistance (MDR) of common pathogens and the rapid emergence of new infections. With the approval of a large number of antimicrobials within classes and the lack of new drug classes, antibiotic drug development has reached a saturation point.\textsuperscript{21} For these reasons there is a pressing need for the development of antibiotics with novel modes of action. Because of the failure of alternative drug discovery methods to deliver many lead compounds, a new wave of interest in natural products has recently emerged.

\textbf{2.2.3 Marine Natural Products}

Among the many fronts being explored to identify novel scaffolds for medicinal study, the marine environment has proven a fertile source of bioactive and chemically diverse molecules. Diverse habitats of the oceans range from warm tropical shallow water coral reefs to areas of sub-zero, deep trenches with high pressure and no light. Differences in the ecosystems of the marine environment result in the production of structurally novel, biologically active secondary metabolites.\textsuperscript{22} Evolution of these metabolites has been driven by various challenges encountered by marine creatures. It has been determined that marine plants and animals are constantly exposed to high concentrations of pathogenic bacteria,
approximately one million cells per milliliter, that readily attach to an organism’s surface. In addition, competition for space, bio-fouling of surfaces, predation and successful reproduction over distances provoked marine invertebrates and animals to develop advanced defense mechanisms.23

In the period from 1965 to 2005 more than 15,000 marine natural products have been isolated.24 Marine derived natural products have a broad spectrum of biological activity, but special attention is given to their anticancer, antimicrobial, antiviral and neuro-modulating properties.25 The importance of marine natural products in drug development is rapidly growing. With the development of new isolation techniques and screening methods more novel therapeutic entities are being supplied to investigate their potential in the treatment of human diseases. An interesting example of a marine natural product with strong cytotoxic activity is trabectedin (2-24, Figure 2.5), a tetrahydroisoquinoline alkaloid which was isolated from Ecteinascidia turbinata, a tunicate found in the Caribbean and Mediterranean seas.26 It is the first marine anticancer agent approved in the European Union for patients with soft tissue sarcoma.27 Currently, trabectedin is undergoing Phase III anticancer clinical trials in the US.

DMXBA, 3-(2,4-dimethoxybenzylidene)-anabaseine (2-23), is a derivative of anabaseine alkaloids produced by some marine worms. Clinical trials of 2-23 have demonstrated significant improvements in cognition of healthy young males and schizophrenics.28 The γ-lactam containing marine natural product salinosporamide A (2-25) is currently in the development stage for anticancer therapy. 2-25 showed an IC$_{50}$ < 2 ng/mL against HCT-116 cells, and was highly selective tumor cell growth inhibitor of 60 cell line
panel at the National Cancer Institute. Salinosporamide A was discovered in the marine bacterium *Salinispora tropica.* Chlorinated alkyl functional group of 2-25 is a key moiety responsible for attraction to a binding site, which enables lactone ring opening and irreversible modification of the proteasome resulting in potent inhibition of growth of cancer cells.

Figure 2.5: Bioactive natural products isolated from various marine species.

Largely unexplored, the marine world is presumably the most biodiverse habitat. Marine environment has a huge potential to be the most vast resource for the discovery of medicinally valuable structures with novel modes of action and structural features covering a biologically relevant area of chemical space.
2.2.3.1 Isolation and Biological Activity of Synoxazolidinones A-C

Recently, bioassay-guided fractionation of Synoicum pulmonaria (a sub-arctic ascidian collected off the Norwegian Coast) led to isolation of a novel family of natural products named synoxazolidinones A, B and C (Figure 2.6). Tadesse et al. reported promising antimicrobial and anticancer activity for the synoxazolidinone family of natural products.

**Figure 2.6:** Synoxazolidinone family of natural products and their biological activity.
In general, all three members exhibit antimicrobial activity against gram-positive bacteria (including Methicillin-resistant *Staphylococcus aureus*). Synoxazolidinones A (2-26) and C (2-28) also moderately inhibit growth of some gram-negative bacteria. In addition, synoxazolidinone C displayed cytotoxic properties towards eukaryotic cells.

All three natural products are guanidine containing molecules which also incorporate brominated aromatics attached to a unique 5-alkylidene-2-alkyl-oxazolidin-4-one heterocycle. The alkylidene fragment of the 3,5-dibromo-4-methoxybenzyl group was determined to have the (Z)-alkene configuration. The simplest structural analogue in the synoxazolidinone family of natural products is synoxazolidinone B (2-27). Unlike the other synoxazolidinone family members, 2-27 lacks a chlorine on the alkyl chain next to the 4-oxazolidinone heterocycle. Lack of the chlorine substituent in synoxazolidinone B may correlate with its diminished MIC values against MRSA (30 µg/mL) comparing to other members of the family. Introduction of the chlorine atom results in a significant enhancement of antibacterial activity for synoxazolidinone A. 2-26 has MIC values against *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* (MRSA) of 10 µg/mL. Additionally, synoxazolidinone A (2-26) inhibits visible growth of the *Corynebacterium glutamicum* at the concentration 6.25 µg/mL and 12.5 µg/mL for the fungi *Saccharomyces cerevisiae*.34

Synoxazolidinone C (2-28) is a bicyclic member of the synoxazolidinone family, and similarly to synoxazolidinone A, it contains a chlorine atom on the alkyl chain next to the aminal center which is now incorporated into the pyrrolidine ring. 2-28 is the most active amongst the synoxazolidinones and inhibits growth of the *Staphylococcus aureus* and MRSA at a concentration of 10 µg/mL. It also shows a MIC of 30 µg/mL against gram-negative
*Escherichia coli* and 20 μg/mL against another common gram-positive pathogen *Enterococcus faecalis*. Synoxazolidinone C is cytotoxic to human cancer cell lines A2058 (melanoma), MCF-7 (breast adenocarcinoma) and HT-29 (colon carcinoma) with an IC_{50} value of 30.5 μM (Figure 2.6). Normal lung fibroblast cells (MRC-5) were killed in the presence of 2-28 at the same concentration. The presence of a pyrrolidine ring in synoxazolidinone C seems to increase its overall cytotoxicity, although the limited data collected to date on this family of natural products does not allow for a complete understanding of their relative activity and toxicity. More recently, one study demonstrated that synoxazolidinones A and C were active in micro-nanomolar range of concentration and inhibited growth of several marine microorganisms at as low as 20 nM having an effect on micro- and macrofouling.

According to the biosynthetic route proposed by Tadesse *et al.*, the initial step in the biosynthesis of synoxazolidinones A and B (Scheme 2.2) is an amide bond formation which occurs between brominated tyrosine derived metabolite 2-28 and an agmatine derivative 2-29.

---

21
Amidation resulting in the formation of 2-30 is then followed by the construction of 4-oxazolidinone ring. The stage of the introduction of the chlorine atom in biosynthesis of 2-31 remains unclear. Synoxazolidinone C can be viewed as cyclic analogue of synoxazolidinone A, and although Tadesse et al. have not carried out biosynthetic studies it was proposed that 2-26 can be a precursor of 2-28 through an oxidation/cyclization event.\textsuperscript{36}
2.2.3.2 Isolation and Biological Activity of Lipoxazolidinones A-C

The only other report on naturally occurring oxazolidin-4-ones describes the isolation of the antibacterial lipoxazolidinones A-C. These natural products were also found in marine environment of actinomycete strain of the genus *Marinspora*, collected from a Guam marine sediment (Figure 2.7).

![Lipoxazolidinones A-C](Ref. (41))

*Marinspora*
(Guam marine sediment)

Staphylococcus aureus: 1 µg/mL
Staphylococcus aureus (MRSA): 1 µg/mL
Enterococcus faecalis: 1 µg/mL
Escherichia coli: >32 µg/mL

Staphylococcus aureus: 1.5 µg/mL
Staphylococcus aureus (MRSA): 3 µg/mL
Enterococcus faecalis: 3 µg/mL
Escherichia coli: >32 µg/mL

Staphylococcus aureus: 3 µg/mL
Staphylococcus aureus (MRSA): 2 µg/mL
Enterococcus faecalis: 2 µg/mL
Escherichia coli: >32 µg/mL

**Figure 2.7:** Family of lipoxazolidinone natural products and their biological activity.

In their structure, the lipoxazolidinones incorporate an unsaturated alkylidene substituent at C2 between oxygen and nitrogen atoms, the alkyl chain is at C5 of the 4-oxazolidinone ring (opposite to the substitution pattern seen in synoxazolidinones, where aromatic benzylidene is at C5 and C2 is saturated). Hydrolysis studies of lipoxazolidinone A at different pH values (Scheme 2.3) revealed that 2-33 converts to 2-36 in neutral media (pH
7.4). The structure of an unusual intermediate 2-36 was confirmed by NMR spectroscopy, whereas treatment of lipoxazolidinone A (2-33) with strong acid led to rapid decomposition with formation of primary amide 2-37 and hydroxy-acid 2-38.

**Scheme 2.3**: Degradation pathways for lipoxazolidinone A.

Lipoxazolidinones A-C (2-33, 2-34, 2-35) and hydrolysis product 2-36 were screened against a panel of gram-positive and gram-negative bacteria. Hydrolysis product 2-36 did not show biological activity, which highlights the importance of the oxazolidinone ring system for antimicrobial efficacy. Antibacterial potency of the lipoxazolidinones is similar to that of the clinically used 2-oxazolidinone antibiotic linezolid (2-21). 2-33, 2-34 and 2-35 showed MIC values of 1-3 µg/mL against MRSA and *Enterococcus faecalis* (Figure 2.7). There have been no reports describing the synthesis of lipoxazolidinone family of the marine natural products.

### 2.3 Conclusion and Outlook

Even though oxazolidinones are rarely seen in nature, they are an established class of compounds in medicinal chemistry (particularly 2-oxazolidinones). In fact, linezolid (2-21) is an antimicrobial agent that belongs to the 2-oxazolidinone class of drugs. While the
oxazolidinone heterocycle is a common scaffold in the structure of the synoxazolidinones, lipoxazolidinones and linezolid, the compounds are clearly distinguished as 4- and 2-oxazolidinones, respectively and each representative has a unique substitution pattern. The isolation of antimicrobial agents with novel core structures (the lipoxazolidinones and the synoxazolidinones) provides new templates for SAR investigations, which may result in leads with new mechanisms of antibacterial action or potency against intractable infections. From an organic synthesis point of view, variously substituted 4-oxazolidinone heterocyclic scaffolds present a challenge of developing a functional group tolerant and flexible synthetic methodology for preparation of this family of natural products and their unnatural analogs.

Our interest in the synoxazolidinone natural products resulted from their promising biological activity, unknown mechanism of action and largely unknown selectivity profile in addition to a unique heterocyclic scaffold. Total synthesis of the synoxazolidinones presents opportunities for reaction development. Intrigued by how the biological activity is varied with subtle modifications around the synoxazolidinone scaffold we became curious to explore the impact of other substituents on the biological potential of these molecules. To this end, we are interested in the synthesis of derivatives of the synoxazolidinones with biological activity superior or complementary to what are reported for these natural products; additionally, we aim to identify sites that tolerate derivitization to allow for future chemical biology studies.
2.4 References


(3) http://www.wildsouthflorida.com/images2014/periwinkle%201200.jpg

(4) http://chinesemedicinenews.com/wp-content/uploads/2008/03/happytree.jpg

(5) http://www.naturalmedicinalherbs.net/thumbnails/wendys/papaver-somniferum=opium-poppy.jpg

(6) http://www.worldbotanical.com/Taxus%20images/Taxus-brevifolia-15802e.jpg


http://www.nature.com/nchembio/journal/v3/n9/fig_tab/nchembio.2007.24_F1.html


http://www.nature.com/nrmicro/journal/v12/n1/fig_tab/nrmicro3155_F5.html


(31) http://www.boldsystems.org/pics/_w300/OPQCS/bn2010-024%2B1289217344.jpg

(32) http://www.snorkelstj.com/picture_library/creatures/tunicates/mangrove_tunicate_8453.JPG

(33) http://bioweb.uwlax.edu/bio203/2011/kolb_kimb/Salinispora%20spore.jpg


(35) http://www.habitas.org.uk/marinelife/tunicata/synpuls.jpg


CHAPTER 3

Previous Syntheses and Applications of Oxazolidine-4-ones
3.1 Abstract

Existing synthetic approaches to variously substituted oxazolidine-4-ones are described. Due to the useful asymmetric applications of 2,5-dialkyl-4-oxazolidinones numerous approaches were developed to access these heterocycles. Methods for synthesis of 2,5-dialkyl-4-oxazolidinones are summarized and important examples highlighted (Section 3.2.2). In contrast, members of the 5-alkylidene-2-alkyl-4-oxazolidinone subclass, found in the synoxazolidinones, are represented by very few specific literature examples provided in Section 3.2.3; therefore, a general approach of their synthesis is needed. With the exception of two isolated analogues described in Section 3.2.4, compounds containing 2-alkylidene-5-alkyloxazolidin-4-one, also found in the lipoxazolidinones, are to a large extent unexplored.

3.2 Introduction and Background

3.2.1 Classification and Nomenclature of Oxazolidinones

An oxazolidine is a 5-membered heterocyclic compound which consists of one nitrogen and an oxygen separated by one and two carbon atoms (Scheme 3.1). Oxazolidine-4-ones (also 4-oxazolidinones) are a sub-class of oxazolidines with a carbonyl group between oxygen and nitrogen of the heterocyclic ring at C4. Depending on the saturation of the ring at positions two and five, 4-oxazolidinones can be formally divided into two groups: dialkyloxazolidin-4-ones (I) and alkylidene-alkyloxazolidinones regioisomers (II and III) (Scheme 3.1).
Scheme 3.1: Structures and nomenclature of oxazolidine-4-ones.

Substitution at C5 and C2 is not limited to monoalkyl and aryl substituents, 4-oxazolidinone can be found in fusion with other azapolycycles also containing ether, amide moieties or quaternary centers. 2,5-dialkyl-oxazolidin-4-ones (I) have found application in asymmetric synthesis of enantiopure 1,2-diols and α-hydroxy carboxylic acids (Scheme 3.2). ¹

Scheme 3.2: Synthesis of optically active α-hydroxy acids and diols from oxazolin-4-one derivatives.

The use of saturated 4-oxazolidinones of type I was reported for high yielding construction of 7- and 8-membered lactams (Scheme 3.3). ²
Scheme 3.3: Synthesis of 4-oxazolidinones from O-acyl-mandelamides.

Thus, the 4-oxazolidinone ring of 3-4 provides an excellent structural bias for the ring-closing metathesis cyclization leading to medium-sized heterocycles that are usually hard to form.

Additionally, there are many examples of compounds with general structure I having substitution on nitrogen as well (Scheme 3.3). A novel asymmetric synthetic scheme to various N-fused polycyclic indolizidines, such as tashiromine (3-9) was developed utilizing ring opening properties of 4-oxazolidinones.  

Scheme 3.4: Synthetic application of the diastereoselective 4-oxazolidinone ring-opening towards synthesis of (+)-tashiromine.
3.2.2 Previous Syntheses of 2,5-dialkyl oxazolidin-4-ones of Type I

Retrosynthetic analysis of approaches available for the synthesis of 2,5-dialkyl oxazolidin-4-ones is summarized in Scheme 3.5 and these methods are described in more detail below.

Scheme 3.5: Retrosynthetic disconnections 2,5-dialkyl-4-oxazolidinones and common starting materials for their synthesis.

The very first report regarding formation of 4-oxazolidinones can be found in works of Emil Fischer in 1896. Fischer’s well known synthesis of oxazole 3-11 (Scheme 3.4) via condensation of equimolar amounts of acetaldehyde and cyanohydrin 3-10 in dry ethereal hydrogen chloride was regularly accompanied by the formation of compound with structure 3-12. He also noticed, that amides of lactic acid 3-15 and mandelic acid 3-16 undergo condensation with acetone in presence of hydrogen chloride to give 4-oxazolidinone products 3-13 and 3-14 which he named “amide-acetones”.

In 1949, Cornforth extended this method to other aldehydes and ketones and explored the use of various acids such as p-toluenesulfonic and glacial acetic acid to obtained 3-17 and
Later, in 1977 Pilgram and Pollard successfully repeated procedures described by Fischer and Cornforth and were able to further expand the product scope.\(^6\) \(p\text{TsOH}\) in refluxing toluene was used to form 2,5-dialkyl-4-oxazolidinones in good to moderate yields. Stereochemistry and ratios of the 4-oxazolidinone products were elucidated by NMR. Significant preference for the cis product formation was observed. Trans product dominated when R had more electron-withdrawing character (Scheme 3.7).

**Scheme 3.6:** Fischer’s oxazole synthesis and its extension.

\[
\begin{align*}
\text{OH} & \xrightarrow{\text{MeCHO, HCl}\ (\text{g}, \text{Et}_2\text{O})} \text{Me} \xrightarrow{\text{N-Ph}} \text{Me} \xrightarrow{\text{O-Ph}} \text{Me} \\
\text{O} & \xrightarrow{\text{R-OH}} \text{O} \xrightarrow{\text{PhCHO, pTsOH, PhMe, } \Delta} \text{R} \xrightarrow{\text{POCl}_3, 80-85\,^\circ\text{C}} \text{N-O} \xrightarrow{\text{PhMe, } \Delta} \text{R}
\end{align*}
\]

Ref. (4, 5)

**Scheme 3.7:** Synthesis and relative configuration of oxazolidine-4-ones

\[
\begin{align*}
\text{Me}_n & \xrightarrow{\text{RCHO, pTsOH, PhMe, } \Delta} \text{Me}_{n-1} \xrightarrow{\text{R-OH}} \text{Me}_{n-1} \xrightarrow{\text{O-R}} \text{Me}_{n-1} \xrightarrow{\text{N-R}} \text{R}
\end{align*}
\]

Ref. (6)
obtained via condensation of lactamide 3-15 with various aldehydes.

Similarly to Fischer’s oxazole synthesis conditions (Scheme 3.6), cyclocondensation of cyanohydrins (3-22) with aldehydes and ketones (3-23) employing a mixture of sulfuric and acetic acids in the presence of acetic anhydride instead of ethereal hydrogen chloride was also reported. These conditions allowed for the preparation of a small library of dialkyloxazolidinones (3-24) and their Cl-containing analogues (3-25, Scheme 3.8). Lower temperatures and shortened reaction times employed in this protocol resulted in greater yields of products relative to previously reported methods.\(^7\)

\[
\begin{align*}
\text{Scheme 3.8: Cyclocondensation of cyanohydrins with aldehydes and ketones.}
\end{align*}
\]

As mentioned above, preparation of 2,5-dialkyl substituted oxazolidine-4-ones is commonly achieved via acid mediated dehydration of aldehydes with \(\alpha\)-hydroxy amides\(^6\) such as 3-15, 3-16, 3-27. Their \(N\)-alkyl derivatives\(^8\) also undergo cyclization to the corresponding oxazolidinones in strongly acidic media. For example, a key intermediate 3-28 in the synthesis of anti-glaucoma compound 3-29 was prepared by condensation of glycolamide 3-27 with aldehyde 3-26 in the presence of \(p\)TsOH in very low 9% yield (Scheme 3.9).\(^9\)
Scheme 3.9: Example of pTsOH mediated dehydration of α-hydroxy amides with aldehydes for preparation of 4-oxazolinones.

The use of Lewis acids in the condensation of α-hydroxy amides with simple aldehydes has also been employed. Reaction of glycolic amide (3-27) with racemic 3,5,5-trimethylhexanal (3-30) in THF in the presence of boron trifluoride diethyl etherate (Scheme 3.10) was reported for preparation of 2-(2,4,4-trimethylpentyl)oxazolidin-4-one (3-31) in poor 11% yield as 1:2 mixture of diastereomers.10

Scheme 3.10: Example of Lewis acid mediated dehydration of α-hydroxy amides with aldehydes for preparation of 4-oxazolinones.

In addition to aldehydes and ketones, their ether precursors can be used for this type of cyclization. As an example, reaction of α-chloroether 3-33 with a homologue of mandelamide 3-32 was used to prepare 3-34 (Scheme 3.11) in 54% yield as 4:1 diastereomeric mixture. Interestingly, both diastereomers showed in vitro cytotoxicity against P388 (murine leukemia cell line) with IC50 values of 57 µg/mL for the major and 37 µg/mL for the minor isomer.11 The yield and selectivity reported for 3-34 are similar to what was
observed by Pilgram (Scheme 3.7). An obvious drawback of this protocol is that gaseous hydrogen chloride most likely will not be tolerated if acid sensitive functionalities are present in the molecule.

![Chemical Structure](image)

**Scheme 3.11:** Example of acid mediated dehydration of α-hydroxy amides with α-chloroethers for preparation of 4-oxazolidinones.

An alternative method was employed for the synthesis of 4-oxazolidinone containing polycycles 3-37 and 3-40 (Scheme 3.12). It involves reaction of TMS protected acyl chlorides of hydroxy acids 3-35 and 3-36 with 3,4-dihydro-β-carboline (3-41). An N-acyliminium ion is formed *in situ* and then trapped by a proximal hydroxyl group released upon cleavage of TMS protecting group. Moderate yields and a preference for cis-isomers of these heterocycles were observed. This method was used to prepare a N-phenyl protected analogue of 3-18 (Scheme 3.6) in 45% yield.

![Chemical Structure](image)

**Scheme 3.12:** One pot preparation of 4-oxazolidinones via addition to an N-acyliminium ion.

Many variations of this multistep protocol involving hydroxyl acid activation
followed by cyclization with imines are employed in preparation of various $N$-containing fused heterocycles.$^{14,15}$ As an example, the one-pot, three-step, conversion of nitro-vinyl substituted cyclohexane 3-42, which proceeded in high yield and diastereoselectivity to give functionalized 4-oxazolidinone 3-43 (Scheme 3.13).

![Scheme 3.13: Synthesis of 4-oxazolidinones from nitro-substituted cyclohexane.](image)

An interesting disconnection for an enantioselective route to $N$-tosyl protected 2,5-dialkyl-4-oxazolidinones with general structure 3-47 very recently was established (Scheme 3.14). It can be described as a formal Lewis base catalyzed [3+2] cycloaddition of ketenes and oxaziridines.$^1$ $N$-heterocyclic carbene catalyst 3-46 (generated in situ from the triazolium salt in the presence of cesium carbonate) catalyzes the cycloaddition of ketene 3-45 and racemic oxaziridine 3-44. Chen and Ye propose a catalytic cycle involving a zwitterionic enolate intermediate which is formed upon addition of a chiral Lewis base catalyst such as for example 3-46 to ketene precursor 3-45. Two transient species (imine and epoxide) react with each other to produce 4-oxazolidinones of general structure 3-49.
**Scheme 3.14:** Substrate scope and proposed mechanism of enantioselective synthesis of 4-oxazolidinones via [3+2] cycloaddition.

Significant preference for cis isomers is observed and yields vary from 41 to 78% for the simple alkyl and aryl derivatives. Application of this method is limited since substrates bearing Lewis basic functional groups may interfere with the chiral catalyst. The resultant oxazolidinones 3-49 can be saponified to important synthetic building blocks such as α-hydroxy carboxylic acids 3-2 or reduced to 1,2-diols 3-1 with almost no erosion of enantiopurity (Scheme 3.2).

Another useful protocol was reported very recently by Stoltz and coworkers for the preparation of tetrasubstituted 4-oxazolidinones (3-53, 3-54) in an asymmetric fashion via palladium (II) catalyzed decarboxylative allylic alkylation procedure (Scheme 3.15).
**Scheme 3.15**: The enantioselective synthesis of α-disubstituted 4-oxazolidinones via palladium-catalyzed allylic alkylation.

The racemic oxazolidinone precursors of 3-51 and 3-52 for asymmetric resolution via Pd catalysis was prepared using mentioned earlier pTsOH mediated condensation between hydroxy amide 3-15 and acetone (Scheme 3.6).

Coupling of (S)-mandelic acid (3-55) with amines followed by coupling with another acid using EDCI affords O-acyl-mandelamides 3-57 in good yields (Scheme 3.16). Upon treatment with TBSOTf amide derivatives 3-57 can be converted to oxazolidinones 3-58. The proposed coordination of the TBS group to the ester carbonyl activates it for the intramolecular nucleophilic attack by the secondary amide. The bulkiness of the TBS group makes one of the intermediates very disfavored which accounts for the observed high levels of diastereoselectivity.
Scheme 3.16: Diastereoselective synthesis of 4-oxazolidinones from O-acyl-mandelamides.

This method allowed preparation of oxazolidinone 3-4 which was transformed into bicyclic intermediate 3-5 using Grubbs catalyst in excellent yield. 3-5, upon reduction followed by basic hydrolysis, was converted into lactam 3-6 in good yield (Scheme 3.3). In comparison, neither of the acyclic amides 3-59 or 3-60 gave satisfactory results in the formation of lactones 3-61 and 3-62 (Scheme 3.17).

Scheme 3.17: Challenge in ring closing metathesis of acyclic amides.

Oxidations and light promoted transformations of α-keto-amides were also reported to furnish oxazolidinone heterocycles. Treatment of 2-azetidinone 3-63 with CAN resulted in formation of tetrasubstituted 4-oxazolidinone 3-64 in 53% yield (Scheme 3.18).
Scheme 3.18: Oxidative cleavage of 2-azetidinones.

Treatment of α-keto-amide 3-65 with potassium bicarbonate in a mixture of water and acetonitrile under photo-irradiation conditions gave bicyclic oxazolidinone 3-66 which upon extended reaction time gave imide of phenylacetic acid 3-67 in 64% yield (Scheme 3.19).\(^{19}\) Photo-irradiation of the optically pure atropisomers of 3-68 leads to formation of a 2:1 mixture of cis/trans diastereomers of 3-69 in good yield and high enantioselectivity (Scheme 3.19).\(^{20}\)

Scheme 3.19: Light promoted transformations of secondary α-keto- amides leading to 4-oxazolidinones.

As 4-oxazolidinones are a sub-class of oxazolidines, it is not surprising that several methods of modification of related unsaturated oxazoline heterocycles (3-72, 3-74) were
explored (Scheme 3.20). Oxazoline 3-72 was obtained via [3+2] cycloaddition of difluorinated azomethine ylide generated in situ form diphenyl imine 3-70 and electron-deficient ketone 3-71. Treatment of fluorooxazoline 3-72 with equimolar amount of KOH in DMSO leads to dialkyloxazolidinone 3-73 in 91% yield. In a similar fashion, oxazoline precursors 3-74 were transformed to tetrasubstituted 4-oxazolidinones 3-78 in moderate to good yields (Scheme 3.20).

Scheme 3.20: Access to 4-oxazolidinones via ring modification of oxazoles.

Acylation of heterocyclic imine 3-74 with phenylacetyl chloride resulted in formation of unstable N-acyliminium ion 3-75, which may exist in the equilibrium with chlorinated derivative 3-76. Subsequent treatment with sodium hydroxide followed by oxidation using PCC provided alkyl and aryl substituted N-acyl oxazolidinones 3-78 as racemic mixtures of diastereomers.

In addition to the aforementioned multistep/multicomponent transformations,
oxazolidine-4-ones were also prepared via intramolecular additions within acyclic amide precursor 3-79 (Scheme 3.21). Amide 3-79 undergoes cyclization and TFE promoted hydrolysis by water, which comes from commercial wet m-CPBA, giving rise to observed spiro-oxazolidinone 3-80 product.\textsuperscript{23} C-N bond formation is catalyzed by a hypervalent iodine (III) species generated from iodoarene and m-CPBA.

![Scheme 3.21: Intramolecular cyclizations leading to 4-oxazolidinones.](image)

Felix \textit{et al.} showed that \textit{O}-tosyl derivative 3-82 can serve as a useful precursor for the preparation of 4-oxazolidinone containing carbohydrate 3-84 which is a product of intramolecular amide bond formation in 3-83.\textsuperscript{24} Sugars, incorporating a dialkyl oxazolidinone moiety in their structure (3-87) very often are components of natural products, and also serve as synthetic precursors for iminosugars. Catalytic hydrogenation of azide 3-86 provided the
amine which undergoes spontaneous cyclization to oxazolidinone derivative 3-87 (Scheme 3.21).

3.2.3 Previous Syntheses of 5-alkylidene-2-alkyloxazolidin-4-ones of Type II

Few literature approaches to the synthesis of 5-alkylidene-2-alkyloxazolidin-4-ones can be found within a context of preparation of specific target compounds. Described below are methods that were optimized to access particular derivatives. General methods to access 4-oxazolidinones are not well established.

Retrosynthetic analysis of existing approaches is shown in Scheme 3.22.

Scheme 3.22: Unsaturated 5-alkylidene-2-alkyl-4-oxazolidinones and common precursors for their synthesis.

A related class of heterocycles, namely the oxazolones, was exploited as precursors to 4-oxazolidinones. Laxmi et al. reported that hydrogenation of 3-88 over palladium on charcoal resulted in reduction of its azomethine carbon to give oxazolidin-4-one 3-89 (Scheme 3.23).

Scheme 3.23: Synthesis of 4-oxazolidinone 3-89 via reduction of oxazolone 3-88.
The authors also noted that 3-89 is readily oxidized back to 3-88 when its methanolic solution was exposed to air.26

Although the reduction of 3-88 is an attractive entry to 4-oxazolidinones, the preparation of starting material 5-benzylidene-2-phenyl-4,5-dihydrooxazol-4-one (3-88) is quite tedious and requires multiple linear steps (Scheme 3.24). The synthesis begins with acylation of deprotonated benzamide 3-90 with 3-phenylpropanoyl chloride to form imide 3-91. Subsequent dibromination and dehydrobromination provides N-benzoxy-2-bromo-3-phenylpropenimide 3-92 which upon reflux with sodium hydride in DME gives 3-88 with 21% yield over 4 steps.

![Scheme 3.24: Synthesis of (Z)-5-benzylidene-2-phenyloxazol-4-(5H)-one (3-88).](image)

The unsaturation at C5 in the ring of 5-alkylidene-2-alkyl-4-oxazolidinones creates the possibility of entry to these heterocycles via substituted 2,5-dialkyl-4-oxazolidinones, which were described in the previous section. In one example Roush and coworkers, utilized the widely employed dehydration of α-hydroxy amides to form 2,5-dialkyl oxazolidin-4-one 3-95 in 86% yield as 2:1 mixture of diastereomers (Scheme 3.25). One-pot N-acylation of 3-95 and oxidation with 1 equiv. of mCPBA followed by elimination of sulfoxide upon treatment of the reaction mixture with triethoxyphosphite in refluxing benzene furnished product 3-96 containing exocyclic double bond in 68% yield.27
Scheme 3.25: Synthesis of 5-alkylidene-2-alkyl-4-oxazolidineone 3-96 and Diels-Alder reaction leading to formation of 2,5-dialkyloxazolidin-4-ones 3-98 and 3-99.

Oxazolidin-4-one 3-96 served as a highly diastereoselective dienophile in Diels-Alder reactions with cyclohexadiene (3-97). Tetra-substituted 2,5-dialkyl oxazolidinones 3-98 and 3-99 were obtained as a 5:1 diastereomeric mixture in 57% yield. Significant preference of 3-96 for exo- cycloaddition was surprising because most α-oxygenated dienophiles generally display modest preference for either endo- or exo- cyclo adducts (Scheme 3.25).²⁷

An interesting result of rather unexpected 5-alkylideneoxazolidin-4-one 3-107 formation occurred during photooxygenation of the tricyclic 4-pyrrolin-3-one 3-102, obtained from cyclocondensation of DBU (3-100) with DMAD (3-101) (Scheme 3-26).²⁸ When treated with singlet oxygen 4-pyrrolin-3-one 3-102 forms dioxetane 3-103 which presumably collapses to tricycle 3-104. Hydrolysis of 3-104 results in opening to macrocycle 3-105. Intramolecular conjugate addition of the enol tautomer 3-106 to the α,β-unsaturated ester delivers 3-107 in 85% yield.²⁹
Scheme 3.26: Photo-oxygenation of the 4-pyrrolin-3-one resulting in 4-oxazolidinone formation.

Photolysis of secondary α-keto amide 3-108 proceeds with loss of phenol 3-110 and formation of 2-methylidene-4-oxazolidinone 3-109 in 79% yield (Scheme 3.27); similarly, this method was employed for synthesis of 2,5-dialkyl oxazolidinones (Scheme 3.17).

Scheme 3.27: Photolysis of secondary α-ketoamides leading to 4 oxazolidinones.

Steinmetz and coworkers also determined that electronic nature of para-substituted phenolic leaving group in 3-108 impacts the yield of 4-oxazolidinone 3-109.

3.2.4 Previous Syntheses of 2-alkylidene-5-alkyloxazolidin-4-ones of Type III

Examples of oxazolidine-4-ones incorporating an alkene fragment at C2 are limited to the lipoxazolidinone natural products and few derivatives from work of Albert Padwa on
Rh(II) promoted cycloaddition cascades.\textsuperscript{31,32} When treated with Rh(II) catalyst diazoimides 3-112 and 3-115 form Rh-carbenoids (Scheme 3.28) that subsequently form dipoles 3-113 and 3-116 with the neighboring carbonyl group of corresponding pyrrolidone (R\textsubscript{1} = Me or CO\textsubscript{2}Me).\textsuperscript{31} Treatment of 3-113 (R\textsubscript{1} = Me) with N-phenyl maleimide resulted in a dipolar cycloaddition reaction with the formation of the expected single exo/anti diastereomer 3-117 containing 2,5-dialkyl-4-oxazolidinone moiety in 90% yield; however when isomunchnone 3-116 was subjected to the same conditions, unexpected fused 2-alkylidene-4-oxazolidinone 3-118 formed in 77% yield via proton abstraction at the carbon with R\textsubscript{1} = CO\textsubscript{2}Me.

\begin{equation}
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-111 \\
3-114
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-112 \\
3-115
\end{array}
\rightarrow
\begin{array}{c}
\text{R}1: \text{Me} \\
\text{R}1: \text{CO}_2\text{Me}
\end{array} \begin{array}{c}
3-113 \\
3-116
\end{array}
\rightarrow
\begin{equation}
\text{Scheme 3.28: Synthesis of fused oxazolidin-4-ones via dipolar cycloaddition reactions.}
\end{equation}

Diazo indoline 3-119 underwent similar Rh(II) promoted transformation.\textsuperscript{32} Heating of 3-119 in benzene at reflux led to isolation of 3-120 in 56% yield.
3.3 Conclusions and Outlook

From an organic synthesis point of view, variously substituted 4-oxazolidinone heterocyclic scaffolds, especially those with ring unsaturation (type II and III) represent a challenge of developing a functional group tolerant and flexible synthetic method for their preparation. The described routes to 4-oxazolidinones of type II in particular are moderately yielding and the conditions used involve strong bases and oxidizing agents. Since 4-oxazolidinones are medicinally relevant heterocycles, general methods for their synthesis are still needed to increase the diversity of their structural analogues. The isolation of antimicrobial natural products with novel core structures (the lipoxazolidinones and the synoxazolidinones) provide new templates for SAR investigations. These molecules have the potential to become leads with new mechanisms of antibacterial action or potency against currently difficult to treat infections. Initially we became interested in 5-alkylidene-2-alkyl-4-oxazolidinones in the context of total synthesis of the synoxazolidinones since, their structural features, such as presence of a guanidine unit, substituted aromatics and α-Cl next to the aminal center represented more complex synthetic challenge and allowed for more sites of derivatization going forward.
3.4 References


(4) Fischer, E. *Ber.* 1896, 29, 205.


CHAPTER 4

Our Approaches to 5-alkylidene-2-alkyl-4-oxazolidinones
4.1 Abstract

We have developed a one step protocol for the preparation of 4-oxazolidinone heterocycles from α-keto amides and aldehydes. During the course of this work we have also prepared primary α-keto amides bearing enolizable protons via a 2-step amine coupling/deprotection approach. These synthetic efforts have led to a series of 4-oxazolidinone products that allowed for systematic evaluation of the aromatic ring’s impact in the antimicrobial activity of the 4-oxazolidinones.

4.2 Introduction and Background

4.2.1 Oxazolidin-4-one Containing Natural Products

The continued emergence of multidrug-resistant bacteria highlights the pressing need for the development of novel antimicrobial agents. Compounds that display antimicrobial activity and bear novel structural features are compelling targets for synthesis and serve as a platform for antibiotic development. Synoxazolidinones A (2-26) and B (2-27) are recently discovered natural products isolated from the subarctic ascidian Synoicum pulmonaria, collected off the Norwegian coast (Figure 2.6). The synoxazolidinones 2-26 and 2-27 contain an unusual 4-oxazolidinone heterocycle bearing an exocyclic conjugated aromatic moiety (Figure 4.1). To our knowledge, an alkene containing 4-oxazolidinone is present in only one other class of natural products (the lipoxazolidinones 2-33 and 2-34, Figure 4.1), albeit with an alternate substitution pattern.
Figure 4.1: Natural products containing a 4-oxazolidinone heterocycle with unsaturation at C2 or C5.

Interestingly, although rare in natural products, oxazolidinones (particularly 2-oxazolidinones) have a rich history in medicinal chemistry as highlighted by successful synthetic drugs such as linezolid (2-21). A synthetic approach to these bioactive scaffolds would thus not only serve the synthesis of this class of natural products but also enable the synthesis of heterocyclic libraries.

Inspired by the proposed biosynthetic route, we envisioned an $N$-acyliminium ion formation/cyclization cascade that would construct the core 4-oxazolidinone ring while stereoselectively controlling the alkene geometry and the aminal center in the process (Scheme 4.1). In situ generation of iminium ions and their subsequent trapping with appended nucleophiles has provided rapid access to a variety of heterocyclic scaffolds.
Scheme 4.1: Proposed transformation to prepare 4-oxazolidinones.

N-acyliminium ion cyclizations are widely employed in the synthesis of nitrogen containing heterocycles, especially in the asymmetric synthesis of complex molecules where they often represent a key transformation in the sequence.\textsuperscript{10,11} We planned to utilize the $\alpha$-substituent (R, Scheme 4.1) to influence stereochemical outcome of the proposed ring closure.

4.2.2 Synthesis and Properties of N-acyliminium Ions

By their nature, N-acyliminium ions are very reactive intermediates, which can be intercepted by a variety of nucleophiles. Due to their reactive nature N-acyliminium ions are very often generated \textit{in situ} in the presence of nucleophilic species they further react with. Some N-acyliminium ions can be isolated and characterized as their salts, but generally they are sensitive compounds and are typically prepared directly prior to use with no purification.

N-acyliminiums are readily available through several well-developed methods (Scheme 4.2). Condensation of amides 4-5 and aldehydes 4-6 is widely employed; however, the lack of electron density on the amide nitrogen sometimes makes the reaction problematic. Higher temperatures and acid catalysis may be invoked to facilitate formation of reactive intermediates.\textsuperscript{11} Condensations involving more electron-rich amides (4-5, R= electron-donating group) tend to be more efficient. Another method that is frequently used for
preparation of \( N \)-acyliminium ions is \( \alpha \)-fragmentation of tertiary \( N \)-acyl amides 4-3. The equilibrium between fragmented \( N \)-acyliminium species and its precursor depends on the nature of the leaving group (LG, Scheme 4.2). \(^{13}\)

![Scheme 4.2: Common precursors to preparation of \( N \)-acyliminium ions in situ.]

In some cases very reactive \( N \)-acyliminiums can be pre-formed by the isomerization of enamides 4-4 using metal catalysis. \(^{14}\) Very often they are subsequently trapped \textit{in situ} by a nucleophile in an intramolecular fashion or stabilized \textit{in situ} by heteroatoms of the solvent. Straightforward acylation of imines 4-8 with activated acid intermediates 4-7 is perhaps one of the most frequently used methods of generating \( N \)-acyliminium ions. \(^{15}\) The literature contains various approaches to prepare iminium ions, with less common methods involving \( \alpha \)-decarboxylation of \( N \)-acyl aminoacids \(^{16}\) and \( \alpha \)-deprotonation-elimination of \( N \)-oxides \(^{17}\) also described.

\( N \)-Acyliminium ions usually react with nucleophiles in a fast and irreversible manner. Even though acylimines are considered to be highly electrophilic also, they are still less electrophilic than their corresponding aldehydes (Scheme 4.3). Their cationic character is also dependent on their \( \alpha \)-substituents \( R_2 \) and \( R_3 \). \(^{18}\)
Scheme 4.3: Relative electrophilicity of various iminium ions.

The regio- and stereochemical outcome of the reactions of $\pi$-nucleophiles with $N$-acyliminium ions is governed by many factors. Baldwin’s rules based on Bürgi-Dunitz trajectory of nucleophilic attack onto $\pi$-systems have been suggested to account for many cyclizations.\textsuperscript{19} According to these rules 5-endo-trig cyclization is a disfavored process.\textsuperscript{20-22} Nevertheless, there are numerous examples of 5-endo-trig cyclizations of iminium ions that occur readily (Scheme 4.4). Heteroatoms (O, S, N) can act as nucleophiles in the intramolecular addition to iminium ions, giving 5-membered oxazolidines, thiazolidines, or imidazolidines respectively. Generally, the attack of the heteroatom nucleophile is occurring through an endo-mode.\textsuperscript{23}

Scheme 4.4: Example of intramolecular addition to an iminium ion.

A chair-like conformation of the $\pi$-complex 4-13 was used to explain the highly diastereoselective formation of pirolizidine heterocycle 4-14, a precursor to isoretonecanol.
(4-15) (Scheme 4.4).\textsuperscript{24} This chair-like type of transition state is widely employed for the prediction of the stereochemical outcome of intramolecular nucleophilic additions to \textit{N}-acyliminiums leading to five- and six-membered rings.\textsuperscript{25}

### 4.2.3 Our Approaches to 4-oxazolidinones

Methods to prepare 4-oxazolidinones bearing exocyclic alkenes are limited and rely on multi-step protocols that are intolerant of diverse functional groups. As part of our program directed toward the discovery of novel scaffolds with antimicrobial activity we have explored several approaches for the preparation of 4-oxazolidinone heterocycles (Scheme 4.5).

\textbf{Scheme 4.5:} Approaches to 4-oxazolidinones developed in our laboratory.

According to our synthetic plan which utilizes reactive \textit{N}-acyliminium ion intermediates (Scheme 4.1), 4-oxazolidinones 4-2 can be accessed via two distinct cascade pathways. First, a dehydration/cyclization approach (Scheme 4.5, left) requires amides 4-16 and aldehydes 4-17 as starting materials. In the second acylation/cyclization approach (Scheme 4.5, right), a reactive \textit{N}-acyliminium ion will arise during acylation of the aldimine 4-19 with an enolizable keto-acid intermediate 4-18 and subsequently undergo intramolecular cyclization with an appended enol nucleophile. The proposed synthetic pathways require preparation of enolizable \textit{\alpha}-keto carbonyl derivatives 4-18 and 4-16. Enolizable aldimines 4-
represent a major challenge in 4-oxazolidinone synthesis as these molecules can readily epimerize and are not stable to harsh reaction conditions.

The following section will describe synthesis of the required starting amides 4-16 to probe our proposed dehydration/cyclization approach.

4.3 Results and Discussion

4.3.1 Synthesis of Arylpyruvic Acids and their Amides

Due to the frequent appearance of the $\alpha$-keto amide moiety in natural products many methods for their preparation were developed.\textsuperscript{26,27} $\alpha$-Keto amides also serve as useful precursors in the elaboration of important heterocycles.\textsuperscript{28-30} Although numerous efficient strategies (standard coupling protocols and nitrile hydrolysis) for the synthesis of various $\alpha$-keto amides were described, methods for the preparation of the primary amides of pyruvic acids were absent in literature. To the best of our knowledge, preparation of primary phenylpyruvamides with an unsubstituted enolizable methylene unit have not been reported previously.

We desired an approach that converted phenylpyruvic acids to their primary amide derivatives in one step, and therefore explored the activation of enolizable $\alpha$-keto acids and coupling with ammonia or ammonia surrogates. Amidation of $\alpha$-keto acids with ammonia appeared to be the most straightforward entry to phenylpyruvamides.\textsuperscript{31} With this goal in mind, we first attempted the activation of phenylpyruvic acid 4-10 and coupling with ammonia and/or its equivalents (Table 4.1).
Table 4.1: Attempts at synthesis of 2-oxo-3-phenylpropanamide.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>T3P, DIPEA, DCM, 0 °C, Mg3N2, H2O</td>
<td>NR</td>
</tr>
<tr>
<td>2</td>
<td>pyBop, DIPEA, DCM, 0 °C, Mg3N2, H2O</td>
<td>NR</td>
</tr>
<tr>
<td>3</td>
<td>(COCl)2, DMF (cat.), THF, 0 °C, NH4OH 28% aq., 0 °C</td>
<td>19*</td>
</tr>
<tr>
<td>4</td>
<td>(COCl)2, DMF (cat.), THF, 0 °C, NH3, g., 0 °C</td>
<td>36</td>
</tr>
<tr>
<td>5</td>
<td>(COCl)2, DMF (cat.), THF, 0 °C, NH3 (g), 0 °C</td>
<td>11*</td>
</tr>
<tr>
<td>6</td>
<td>1,3-Imidazole, CO(NH)2, 300 MW, 3 min., 210 °C</td>
<td>NR</td>
</tr>
</tbody>
</table>

* - impurities.

We first explored traditional synthetic approaches to primary amides relying on the treatment of acyl halides, acid anhydrides, esters or acids with ammonia.\textsuperscript{32} Formation of phenylpyruvic acid chloride using oxalyl chloride followed by treatment of the reaction mixture with an aqueous ammonia solution provided the corresponding amide 4-11 in 19\% yield (Entry 3, Table 4.1). Suspecting that low yields may be due to competitive hydrolysis of the acyl chloride we used dry ammonia gas. On the first attempt, an improved 36\% yield was observed, but unfortunately this result was not reproducible and the next batch of amide 4-11 was prepared in only 11\% yield (Entries 4 and 5, Table 4.1). Aryl pyruvic acids themselves are prone to decomposition when kept at room temperature, and the presence of an enolizable α-keto group makes their corresponding acyl chlorides very reactive and unstable. Despite good yields reported for the synthesis of β,γ-unsaturated primary α-keto amides using the magnesium nitride/water system as an ammonia source, neither T3P nor pyBop coupling agents gave desired phenylpyruvamide 4-11 (Entries 1 and 2, Table 4.1).\textsuperscript{31}
Disappointingly, all attempts to prepare 4-11 via this strategy provided unacceptably low yields due to the reactivity of the enol tautomer of the activated acid predominating under the reaction conditions, leading to dimeric and polymeric material and low yields of desired amide.

It became apparent that a more nucleophilic amine (and an easily revealed primary amide synthon) was required for the desired coupling reaction. N-alkylamides, easily prepared by coupling carboxylic acid derivatives with electron-rich alkylamines, are often used as precursors to primary amides. Subsequent cleavage of substituents like trityl- or methoxybenzyl- is then performed in acidic reaction media.\textsuperscript{33} For example, Kan \textit{et al.} had successfully debenzylated various 2,4-dimethoxy-N-benzylamides with four equivalents of \( p \)-toluenesulfonic acid (\( p \)TsOH) in refluxing toluene;\textsuperscript{34} therefore, we set out to evaluate a two-step protocol for the synthesis of 4-11.

Coupling of acid 4-10 with 2,4-dimethoxybenzylamine (2,4-DMBA) employing DCC/HOBt in THF provided the secondary amide 4-12 in 72\% yield, whereas DCC/DMAP in DCM yielded only 54\% of dimethoxybenzylamide 4-12 (Table 4.2). With 4-12 in hand we conducted acid-promoted cleavage of the DMB group and demonstrated that TFA at elevated temperature (70 °C, 1 h) provided efficient conversion to the targeted primary \( \alpha \)-keto amide 4-11 (Entry 7, Table 4.2). We found other nucleophilic amines, as well as other acids such as \( p \)TsOH (Entry 5, Table 4.2), to also function well in the coupling reaction.
Table 4.2: Acid promoted debenzylation of 4-12 obtained via coupling of phenylpyruvic acid with 2,4-dimethoxybenzylamine.

![Chemical structure diagram]

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>TFA-DCM (or THF), 1:1, rt</td>
<td>NR</td>
</tr>
<tr>
<td>2</td>
<td>pTsOH, PhMe, rt</td>
<td>NR</td>
</tr>
<tr>
<td>3</td>
<td>CAN, MeCN-H₂O, 9:1, rt</td>
<td>NR</td>
</tr>
<tr>
<td>4</td>
<td>BF₃·Et₂O or BF₃·AcOH, DCM</td>
<td>NR</td>
</tr>
<tr>
<td>5</td>
<td>pTsOH, PhMe, 85 °C</td>
<td>43</td>
</tr>
<tr>
<td>6</td>
<td>TFA-THF, 2:1, 70 °C</td>
<td>57</td>
</tr>
<tr>
<td>7</td>
<td>TFA-THF, 1:1, 70 °C</td>
<td>65</td>
</tr>
</tbody>
</table>

A two-step protocol employing TFA afforded the most consistent yields on scale and we were able to apply this approach to a series of substituted aromatic derivatives bearing both electron-rich and electron-poor rings (Table 4.3). Via this route variously substituted pyruvic acids 4-13a-j were successfully converted into their primary amides 4-15a-j.
Table 4.3: Substrate scope for amide coupling/DMB cleavage protocol.

<table>
<thead>
<tr>
<th></th>
<th>DCC, HOBt, 2,4-DMBA</th>
<th>DMB</th>
<th>TFA-THF, 70 ºC</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-13a-j</td>
<td>THF, 0 ºC - rt</td>
<td></td>
<td>27-92%</td>
</tr>
<tr>
<td>4-14a-j</td>
<td></td>
<td></td>
<td>48-76%</td>
</tr>
<tr>
<td>4-15a-j</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

With this scalable and rapid access to a wide range of substituted amides\(^\text{38}\) in hand we were able to proceed to the exploration of the acid catalyzed dehydration/cyclization approach to prepare 4-oxazolidinones. At the outset of this work it was unclear whether such reaction conditions would provide 4-oxazolidinones via the desired \(\text{O}-\)addition to acyliminium ion.

4.3.2 Acid Promoted Cyclization/Dehydration Cascade of Primary Keto Amides and Aldehydes

4.3.2.1 Cyclization/Dehydration of Unsubstituted Aliphatic Aldehydes

One of the typical entries to iminium ions is the condensation of amines with aldehydes.\(^\text{35}\) Amines bearing electron-withdrawing substituents are usually less reactive in these condensation reactions. To compensate for low nucleophilicity of nitrogen bearing
electron-withdrawing substituents, activation of the carbonyl group of the reacting partner by a Brønsted or Lewis acid is employed. With this in mind we selected to focus our efforts on trifluoroacetic acid for the acid promoter since it was readily removable from our reaction products and proved compatible with the amide starting materials (Table 4.4). Reaction conditions were optimized on model substrates amide 4-11 and hexanal 4-16.

**Table 4.4:** Acid concentration screen for 4-oxazolidinone formation via dehydration/cyclization approach.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Yielda (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>TFA (1 equiv.), hexanal (2 equiv.)</td>
<td>&lt; 10b</td>
</tr>
<tr>
<td>2</td>
<td>TFA (4 equiv.), hexanal (2 equiv.)</td>
<td>&lt; 10b</td>
</tr>
<tr>
<td>3</td>
<td>TFA (4 equiv.), hexanal (10 equiv.)</td>
<td>&lt; 10b</td>
</tr>
<tr>
<td>4</td>
<td>TFA (12 equiv.), hexanal (2 equiv.)</td>
<td>&lt; 10b,c</td>
</tr>
<tr>
<td>5</td>
<td>TFA (50%, v/v), hexanal (1 equiv.)</td>
<td>&lt; 10b</td>
</tr>
<tr>
<td>6</td>
<td>TFA (50%, v/v), hexanal (2 equiv.)</td>
<td>29bc</td>
</tr>
<tr>
<td>7</td>
<td>TFA (50%, v/v), hexanal (10 equiv.)</td>
<td>&lt; 10b,c</td>
</tr>
<tr>
<td>8</td>
<td>TFA (50%, v/v), hexanal (2 equiv.)</td>
<td>48bd</td>
</tr>
<tr>
<td>9</td>
<td>TFA (65%, v/v), hexanal (1 equiv.)</td>
<td>&lt; 10be</td>
</tr>
<tr>
<td>10</td>
<td>TFA (65%, v/v), hexanal (2 equiv.)</td>
<td>57b</td>
</tr>
</tbody>
</table>

Ref. (38)

*a* - isolated yields; *b* - incomplete conversion of 4-11; *c* - significant impurities present; *d* - 0.4 mmol scale; *e* - slow conversion.

Screening of acid concentration and reactant ratios revealed that 2:1 TFA/THF (v/v) and 2 equiv. of aldehyde provided the most efficient conversion, yielding 57% of 4-17 after purification on silica gel (Entry 10, Table 4.4). We found that there was an ideal range for the
acid/reactant ratio and reactions that were too dilute or too concentrated were significantly messier, likely due to the slow initial dehydration reaction and additional decomposition pathways of the starting material or 4-oxazolidinone product if the reaction is allowed to proceed for extended periods of time. It is important to note that the mass recovery observed in these reactions is high (> 80%) and a significant loss of material arises during the purification process (SiO₂). We then applied the acid promoted dehydration/cyclization reaction to the previously prepared keto amides (4-15a–i, Table 4.3) and hexanal (4-16) to define the functional group tolerance of the transformation as well as to prepare a series of compounds to explore the antimicrobial SAR studies of the left hand fragment of the synoxazolidinones. Utilizing the optimized conditions (Entry 10, Table 4.4) a number of electron-rich and electron-poor arylpyruvic amides were converted to their corresponding 4-oxazolidinone products in moderate to good yields (Table 4.5). In all cases the 4-oxazolidinone was the only significant product observed in the crude reaction mixtures.

**Table 4.5:** Substrate scope for dehydration/cyclization reaction.

<table>
<thead>
<tr>
<th>R</th>
<th>Product</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Me</td>
<td>4-18</td>
<td>47%</td>
</tr>
<tr>
<td>tBu</td>
<td>4-19</td>
<td>47%</td>
</tr>
<tr>
<td>F</td>
<td>4-20</td>
<td>57%</td>
</tr>
<tr>
<td>Cl</td>
<td>4-21</td>
<td>63%</td>
</tr>
<tr>
<td>F₃C</td>
<td>4-22</td>
<td>32%</td>
</tr>
<tr>
<td>NO₂</td>
<td>4-23</td>
<td>39%</td>
</tr>
<tr>
<td>Br</td>
<td>4-24</td>
<td>39%</td>
</tr>
<tr>
<td>MeO</td>
<td>4-25</td>
<td>67%</td>
</tr>
<tr>
<td>Br</td>
<td>4-26</td>
<td>37%</td>
</tr>
</tbody>
</table>

Ref. (38)
4.3.2.2 Cyclization/Dehydration of Substituted Aliphatic Aldehydes

In order to proceeded to exploration of the influence of an α-Cl substituent on reaction yield and stereoselectivity, 4-16 was converted into racemic α-Cl-hexanal 4-28 using an electrophilic chlorination protocol described by Jorgensen. Treating a mixture of α-Cl-hexanal 4-28 and amide 4-11 in TFA under optimized conditions resulted in very poor 8% yield of the desired α-Cl-oxazolidinone product, isolated as a 1:1 diastereomeric mixture (Entry 3, Table 4.6).

Table 4.6: Dehydration of 2-oxo-3-phenylpropanamide 4-11 with enolizable aldehydes 4-28, 4-16.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Comment</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4-16, T3P, DMF, 80 °C</td>
<td>complex mixture&lt;sup&gt;a&lt;/sup&gt; containing 4-17</td>
<td>ND*</td>
</tr>
<tr>
<td>2</td>
<td>4-16, TFA, DCM (1:1), rt</td>
<td>4-17</td>
<td>48</td>
</tr>
<tr>
<td>3</td>
<td>4-28, TFA, DCM (1:1), rt</td>
<td>4-27, 1:1 mixture of diastereomers&lt;sup&gt;b&lt;/sup&gt;</td>
<td>8</td>
</tr>
<tr>
<td>4</td>
<td>4-28, BF&lt;sub&gt;3&lt;/sub&gt;·Et&lt;sub&gt;2&lt;/sub&gt;O, THF, rt</td>
<td>4-27, 1:1 mixture of diastereomers&lt;sup&gt;b&lt;/sup&gt;</td>
<td>22</td>
</tr>
</tbody>
</table>

* - initial exploration of these conditions on a simple model system was performed by Sebastian Guevera.  
<sup>a</sup> - determined by HPLC-MS analysis, <sup>b</sup> - purified by reverse phase flash chromatography.

We next explored propylphosphonic anhydride (T3P), a recently developed coupling agent also shown to act as a water scavenger. Heating a mixture of amide 4-11 and hexanal (4-16) with T3P at 80 °C in DMF resulted in a complex reaction mixture which contained
only trace amounts of desired product 4-17 (Entry 1, Table 4.6). Trachsel et al. previously reported diastereoselective dehydration between aldehyde 3-30 and glycolamide 3-27 for preparation of fragrant oxazolidin-4-one 3-31 employing boron trifluoride etherate in THF (Scheme 3.10). We expected that a strong Lewis acid such as boron trifluoride etherate will better activate the carbonyl for condensation with an amide and possibly influence the diastereoselectivity of the ring closure. When we utilized these conditions with our substrates phenypyruvamide 4-11 and aldehyde 4-28 target 4-oxazolidinone 4-27 was obtained in a slightly higher 22% yield, but the diastereomeric ratio was still 1:1 (Entry 4, Table 4.6). Despite poor yield for the synthesis of Cl-containing 4-oxazolidinones we proceeded forward and attempted to access more substituted scaffolds relevant for the synoxazolidinones.

With a route to the core heterocycle in hand, we turned our attention to the incorporation of the functional groups required for the synoxazolidinones. To account for the presence of an acyclic guanidine functional group in synoxazolidinone A (2-26) and B (2-27) our approach required synthesis of Boc-protected guanidine aldehydes 4-30 and 4-31. For preparation of 4-31 we followed a known literature procedure (Scheme 4.6), and 4-31 was obtained in two steps with 88% overall yield starting from commercially available starting material.\(^{39,40}\) Reaction of S-methylthioisourea 4-29 with 1,4-aminobutanol in THF at 50 °C for 2 hours provided 4-28 quantitatively.\(^{39}\) Swern oxidation of guanidine containing alcohol 4-28 produced desired aldehyde substrate 4-31.\(^{40}\) Proline mediated α-chlorination of 4-31 using N-chlorosuccinimide (NCS) was accomplished in 50% yield.\(^{36}\) In our hands neat α-Cl aldehyde 4-30 appeared to be unstable, but could be stored well in solution. (Scheme 4.6).
We spent a significant amount of effort trying to optimize the dehydration/enol addition reaction using Boc-protected guanidine aldehydes 4-30 and 4-31. Unfortunately, previously developed conditions which involve acids did not result in formation of products 4-32 and 4-33. Higher temperatures and change of the solvent polarity allowed the observation of traces of 4-33, but significant amount of decomposition was also noticed (Entry 1, Table 4.7). We observed loss of the Boc- protecting groups and further decomposition of the guanidine containing aldehydes seem to occur faster than the desired cyclization cascade.

Scheme 4.6: Preparation of $N^2, N^3$-Bis(tert-butoxycarbonyl)-$N^4$-(3-chloro-4-oxybutyl)guanidine (4-31).
Table 4.7: Acid mediated dehydration/cyclization of 2-oxo-3-phenylpropanamide 4-11 with guanidine containing aldehydes 4-30 and 4-31.

![Chemical Structure](image)

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Comment</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4-31, TFA</td>
<td>SM</td>
<td>NR</td>
</tr>
<tr>
<td></td>
<td>solvents:</td>
<td>drying agent: CuSO₄, MgSO₄, 4 Å MS</td>
<td>traces of 4-33 decomposition of SM</td>
</tr>
<tr>
<td></td>
<td>DCM, CCl₄, PhH, PhMe, THF, DMSO</td>
<td>temperature: rt, 50 – 90 °C</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>4-31, Ti(OMe)₄, THF, 70 °C</td>
<td>complex mixture⁵</td>
<td>ND</td>
</tr>
<tr>
<td>3</td>
<td>4-31, BF₃·Et₂O, THF, rt</td>
<td>traces of 4-33bung</td>
<td>ND</td>
</tr>
<tr>
<td>4</td>
<td>4-31, BF₃·Et₂O, THF, rt, then TFA, DCM (1:1), rt</td>
<td>4-33bung, SM decomposition</td>
<td>23⁵</td>
</tr>
<tr>
<td>5</td>
<td>4-30, BF₃·Et₂O, THF, rt, then TFA, DCM (1:1)</td>
<td>traces of 4-32, SM decomposition</td>
<td>ND</td>
</tr>
</tbody>
</table>

* - impurities; a - determined by HPLC-MS analysis; b - purified by reverse phase flash chromatography;

Since Dean-Stark conditions for azeotropic water removal seemed impractical on a small reaction scale we also explored several water scavengers. Combination of TFA with 4 Å molecular sieves (4 Å MS) in various solvents did not produce significant quantities of product. Addition of other mild dehydration agents such as magnesium and copper sulfates in DCM also failed; however, when 4-31 was subjected to boron trifluoride diethyletherate conditions the cyclization occurred. After subsequent removal of the Boc-groups by 50% v/v solution of TFA in DCM, 4-oxazolidinone 4-33 was isolated in 23% yield over 2 steps (Entry 4, Table 7.4). Unfortunately, α-Cl-aldehyde 4-30 did not cyclize with phenylpyruvamide 4-
11 under BF$_3$·Et$_2$O or TFA conditions to provide chloro-oxazolidinone 4-32 (Entry 5, Table 7.4).

It is worth mentioning that in all of the cases where we observed formation of the products (Tables 4.6 and 4.7) starting material amide 4-11 was never completely consumed. Cases where there was no amide left after the reaction contained unknown decomposition byproducts or involved hydrolysis of 2-oxo-3-phenylpropanamide 4-11 to the parent α-keto acid 4-10. Excess of aldehyde and dehydrating agent, extended reaction times and higher temperatures never resulted in full conversion without decomposition or hydrolysis of amide 4-11.

Anticipating increased reactivity from a more electron-rich amide, such as 4-15g which we prepared from 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanoic acid (4-13g) using conditions optimized in our lab for the synthesis of 4-11 (Table 4.2). The majority of acids 4-13 as well as acid 4-13g was not commercially available so they had to be prepared via Erlenmeyer–Plöchl synthesis of an azlactone intermediate (Scheme 4.7).$^{41,42}$ Condensation of N-acetylglycine with 3,5-dibromo-4-methoxy benzaldehyde (4-34) in acetic anhydride followed by hydrolysis of 4-benzylideneoxazol-5(4H)-one (4-35) with 3 N HCl provided acid 4-13g in 68% yield.$^{43,44}$ 4-13g was then coupled with 2,4-dimethoxy-N-benzylamine to provide amide 4-14g which was successfully de-benzylated to yield primary amide 4-15g (Scheme 4.7).$^{38}$
Scheme 4.7: Synthesis of 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanoic acid (4-13g) and its primary amide 4-15g.

At this point we had all the required precursors for the synthesis of both synoxazolidinones A and B according to our dehydration/cyclization approach. Despite our moderate success in the synthesis of simple chlorinated 4-oxazolidinones (Table 4.6), we still evaluated these conditions to access synoxazolidinones (Table 4.8).

With both substrates 4-15g and 4-31, TFA and BF₃·Et₂O conditions failed to provide significant product formation. Heating over 50 °C was required for cyclization to be observed (Entries 1,6 and 2-4, Table 4.8), other strong acids such as methanesulfonic acid and hydrochloric acid gave complex reaction mixtures containing only traces of the natural products. Finally, p-toluenesulfonic acid was employed and the reaction was heated in toluene (Entry 7, Table 4.8). With some optimization using five equivalents of pTsOH at 85 °C in toluene we were able to isolate synoxazolidinone A as 1:1 mixture of diastereomers and synoxazolidinone B with 22% and 23% yields respectfully. The presence of the polar guanidine group added a challenge to purification.
**Table 4.8:** Dehydration of 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanamide (4-15g) with aldehydes 4-30 and 4-31.

\[
\text{Conditions: L-Pro, NCS, DCM, 0 }^\circ\text{C}
\]

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Comment</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4-31, TFA</td>
<td>SM</td>
<td>NR</td>
</tr>
<tr>
<td></td>
<td>DCM (1:1), 50 °C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>4-30 or 4-31, TFA</td>
<td>traces of 2-26 or 2-27, decomposition of SM</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>PhMe or Dioxane (1:1), 85 °C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>4-30 or 4-31, 4M HCl</td>
<td>complex mixture(^a)</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>Dioxane, 85 °C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>4-30, MSA, PhMe, 85 °C</td>
<td>complex mixture(^a)</td>
<td>ND</td>
</tr>
<tr>
<td>5</td>
<td>4-30, BF(_3)·Et(_2)O, PhMe, 85 °C</td>
<td>decomposition of SM</td>
<td>NR</td>
</tr>
<tr>
<td>6</td>
<td>4-30 or 4-31, pTsOH</td>
<td>SM</td>
<td>NR</td>
</tr>
<tr>
<td></td>
<td>DCM, 50 °C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>4-30 or 4-31, pTsOH</td>
<td>2-26 and 2-27</td>
<td>ND(^a)</td>
</tr>
<tr>
<td></td>
<td>PhMe, 80 °C, 10 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>4-31, pTsOH</td>
<td>2-27(^b)</td>
<td>23(^*)</td>
</tr>
<tr>
<td></td>
<td>PhMe, 80 °C, 30 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>4-30, pTsOH</td>
<td>decomposition of SM</td>
<td>NR</td>
</tr>
<tr>
<td></td>
<td>PhMe, 85 °C, 40 min, 75 MW</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>4-30, pTsOH</td>
<td>2-26, 1:1 mixture of diastereomers(^b)</td>
<td>22(^*)</td>
</tr>
<tr>
<td></td>
<td>PhMe, 85 °C, 30 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>4-30, pTsOH</td>
<td>2-26, 1:1 mixture of diastereomers(^b)</td>
<td>6(^*)</td>
</tr>
<tr>
<td></td>
<td>PhMe, 85 °C, 4 Å MS, 30 min</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\(^a\) – impurities; \(^b\) – determined by HPLC-MS analysis; \(^\ast\) – purified by reverse phase flash chromatography.

We used reverse phase column chromatography purification on C18-silica with a water-acetonitrile gradient and TFA as an additive. Despite our best efforts, isolation was accompanied with impurities of unknown nature in both natural products and therefore we
could not confidently confirm their structure at this stage. Unfortunately, amide 4-15g did not prove more reactive in the dehydration/enol addition sequence. Instead, it was hydrolyzing back into corresponding acid 4-13g even more readily than 4-11 was converting to 4-10. Attempts to remove water during the reaction using 4 Å MS did not result in an improvement of yield of the reaction or purity of the product (Entry 11, Table 4.8). In addition, scaling up the reaction to get larger quantities of the natural products (in order to achieve successful purification) failed several times.

Our attempts to achieve cyclization of substituted aldehydes and primary α-keto amides with enolizable methylene unit show that the energetic barrier of the dehydration reaction between α-aldehyde and primary α-keto amide is most likely high. Increasing of the temperature and/or acid strength facilitated 4-oxazolidinone formation, but it was usually much slower than decomposition of the starting material. The main decomposition pathways involved loss of Boc-protecting group of the guanidine aldehyde and hydrolysis of primary amides to their parent acids.

**4.4 Conclusions and outlook.**

In summary, the acid promoted dehydration/cyclization method is straightforward for the preparation of 4-oxazolidiones derived from unsubstituted aliphatic aldehydes; however, it is not compatible with more sterically hindered aldehydes such as α-chloro aldehydes, or functionalized aldehydes such as those bearing guanidine groups. Although these limitations have not allowed this approach to be extended to the synoxazolidinone family of natural
products, this acid catalyzed method provides an attractive one-step synthesis of analog structures.

4.5 Experimental

THF was purified using an alumina filtration system. Aldehydes were purchased from a commercial chemical company and used as received. Reactions were monitored by TLC analysis (pre-coated silica gel 60 F254 plates, 500 μm layer thickness) and visualization was accomplished with a 254 nm UV light and by staining with a KMnO₄ solution (1.5 g of KMnO₄, 10 g of K₂CO₃, and 1.25 mL of a 10% NaOH solution in 200 mL of water). Reactions were also monitored by LC-MS (2.6 mm C18 50 x 2.10 mm column). Flash chromatography on SiO₂ was used to purify the crude reaction mixtures and performed on a flash system utilizing pre-packed cartridges and linear gradients. Melting points were determined using a capillary melting point apparatus. Infrared spectra were determined on a FT/IR spectrometer. ¹H, ¹³C and ¹⁹F NMR spectra were obtained on a 400 MHz instrument in CDCl₃ unless otherwise noted. Chemical shifts were reported in parts per million with the residual solvent peak used as an internal standard (CDCl₃ = 7.26 ppm for ¹H and 77.16 ppm for ¹³C). ¹H NMR spectra were run at 300 or 400 MHz and are tabulated as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, p = pentet, m = multiplet, bs = broad singlet, dt = doublet of triplet, tt = triplet of triplet), number of protons, and coupling constant(s). ¹³C NMR spectra were run at 100 MHz using a proton-decoupled pulse sequence with a d1 of 0 second unless otherwise noted, and are tabulated by observed peak. High-resolution mass spectra were obtained on an ion trap mass spectrometer using heated
electrospray ionization (HESI). All aryl-2-oxopropanoic acids were prepared according to general procedure previously described in literature.\textsuperscript{43,45}

**General Procedure for Preparation of \(N\)-(2,4-dimethoxybenzyl)-3-aryl-2-oxopropanamides. General Protocol A:**

All 2,4-dimethoxybenzyl amides were prepared via DCC coupling of the corresponding aryl-2-oxopropanoic acid with 2,4-dimethoxybenzyl amine.\textsuperscript{46} Typically, to a 0 \(^\circ\)C solution of the acid (1 equiv.), \(N\)-hydroxybenzotriazole (1.2 equiv.) and \(N,N'\)-dicyclohexylcarbodiimide (1.2 equiv.) in anhydrous THF (0.2 M) was slowly added 2,4-dimethoxybenzyl amine (1 equiv.). The resulting mixture was warmed to ambient temperature and stirred until full consumption of starting material (3-12 h, monitored by LC-MS). The reaction mixture then was filtered, the resulting white solid was washed with THF, the organic layers were combined, concentrated \textit{in vacuo} and the crude oily residue purified by flash chromatography on SiO\textsubscript{2} (2-55% EtOAc/hexanes).

\[
\text{N-(2,4-Dimethoxybenzyl)-2-oxo-3-phenylpropanamide (4-12).}
\]

According to general protocol A, \textbf{4-12} (0.51 g, 72%) was obtained as a yellow solid: \(^1\)H NMR (300 MHz, CDCl\textsubscript{3}) \(\delta\) 7.54 – 6.96 (m, 6H), 6.56 – 6.15 (m, 2H), 4.39 (d, 2H, \(J = 6.1\) Hz), 4.22 (s, 2H), 3.81 (s, 3H), 3.79 (s, 3H). \(^1^3\)C NMR (75 MHz, CDCl\textsubscript{3}) \(\delta\) 196.3, 160.9, 159.6, 158.7, 132.9, 130.7, 129.9, 128.7, 127.2, 117.6, 104.0, 98.7, 55.5, 55.4, 43.3, 39.2. IR
(film): 3403, 2937, 1684, 1508, 1209, 1036, 700 cm⁻¹. HRMS (ESI) m/z calcd for C₁₈H₁₉NO₄ [M+H]⁺ 314.1369, found 314.1384; mp = 61-63 °C.

![Chemical structure](image)

**N-(2,4-Dimethoxybenzyl)-2-oxo-3-(p-tolyl)propanamide (4-14a).**

According to general protocol A, 4-14a (1.92 g, 67%) was obtained as a yellow oil: 

¹H NMR (300 MHz, CDCl₃) δ 7.37 (s, 1H), 7.12 (s, 4H), 6.59 – 6.14 (m, 2H), 4.39 (d, 2H, J = 6.1 Hz), 4.17 (s, 2H), 3.81 (s, 3H), 3.79 (s, 3H), 2.32 (s, 3H). ¹³C NMR (101 MHz, CDCl₃) δ 196.4, 160.9, 159.6, 158.7, 136.9, 130.7, 129.8, 129.4, 117.6, 104.0, 98.7, 55.5, 55.4, 42.9, 39.2, 21.2. IR (film): 3414, 2961, 1683, 1509, 1208, 1036, 795 cm⁻¹. HRMS (ESI) m/z calcd for C₁₉H₂₁NO₄ [M+Na]⁺ 350.1363, found 350.1359.

![Chemical structure](image)

**3-(4-(Tert-butyl)phenyl)-N-(2,4-dimethoxybenzyl)-2-oxopropanamide (4-14b).**

According to general protocol A, 4-14b (0.19 g, 55%) was as a clear oil: ¹H NMR (300 MHz, CDCl₃) δ 7.36 (s, 1H), 7.39 – 7.29 (m, 2H), 7.25 – 7.12 (m, 3H), 6.67 – 6.22 (m, 2H), 4.39 (d, 2H, J = 6.1 Hz), 4.18 (s, 2H), 3.80 (s, 3H), 3.79 (s, 3H), 1.30 (s, 9H). ¹³C NMR (75 MHz, CDCl₃) δ 196.5, 160.9, 159.7, 158.8, 150.1, 130.7, 129.8, 129.6, 125.7, 117.6, 104.0, 98.7, 55.5, 55.5, 42.8, 39.2, 34.6, 31.4. IR (film): 3414, 2961, 1683, 1508, 1208, 1037, 835 cm⁻¹. HRMS (ESI) m/z calcd for C₂₂H₂₇NO₄ [M+H]⁺ 370.2013, found 370.2005.
N-(2,4-Dimethoxybenzyl)-3-(4-fluorophenyl)-2-oxopropanamide (4-14c).

According to general protocol A, 4-14c (1.27 g, 70%) was as a yellow solid: \(^1\text{H NMR} \ (300 \text{ MHz, CDCl}_3) \ \delta 7.37 \ (s, 1\text{H}), 7.24 - 7.10 \ (m, 3\text{H}), 7.07 - 6.90 \ (m, 2\text{H}), 6.56 - 6.31 \ (m, 2\text{H}), 4.39 \ (d, 2\text{H}, \ J = 6.1 \text{ Hz}), 4.18 \ (s, 2\text{H}), 3.81 \ (s, 3\text{H}), 3.79 \ (s, 3\text{H}). \ ^{13}\text{C NMR} \ (101 \text{ MHz, CDCl}_3) \ \delta 196.1, 163.4, 161.0, 160.9, 159.5, 158.8, 131.5, 131.5, 130.7, 128.6, 128.6, 117.5, 115.7, 115.5, 104.0, 98.8, 55.5, 55.5, 42.5, 39.3. \ IR \ (film): \ 3413, \ 2938, \ 1683, \ 1509, \ 1209, \ 1035, \ 825 \text{ cm}^{-1}. \ \text{HRMS (ESI) } m/z \ \text{calcd for C}_{18}\text{H}_{18}\text{FNO}_4 [\text{M+H}]^+ 354.1112, \ \text{found 354.1114}; \ \text{mp} = 90-93 ^\circ\text{C}.

3-(4-Chlorophenyl)-N-(2,4-dimethoxybenzyl)-2-oxopropanamide (4-14d).

According to general protocol A, 4-14d (1.0 g, 57%) was as a yellow solid: \(^1\text{H NMR} \ (300 \text{ MHz, CDCl}_3) \ \delta 7.39 \ (s, 1\text{H}), 7.31 - 7.26 \ (m, 2\text{H}), 7.21 - 7.11 \ (m, 3\text{H}), 6.59 - 6.24 \ (m, 2\text{H}), 4.39 \ (d, 2\text{H}, \ J = 6.1 \text{ Hz}), 4.18 \ (s, 2\text{H}), 3.81 \ (s, 3\text{H}), 3.79 \ (s, 3\text{H}). \ ^{13}\text{C NMR} \ (75 \text{ MHz, CDCl}_3) \ \delta 195.9, 161.0, 159.4, 158.7, 133.2, 131.4, 131.3, 130.7, 128.9, 117.5, 104.0, 98.7, 55.5, 55.5, 42.7, 39.3. \ IR \ (film): \ 3406, \ 2937, \ 1684, \ 1508, \ 1208, \ 1036, \ 805 \text{ cm}^{-1}. \ \text{HRMS (ESI) } m/z \ \text{calcd for C}_{18}\text{H}_{18}\text{ClNO}_4 [\text{M+H}]^+ 348.0997, \ \text{found 348.0987}; \ \text{mp} = 90-92 ^\circ\text{C}.
\[N-(2,4\text{-dimethoxybenzyl})-2\text{-oxo}-3-(4-(trifluoromethyl)phenyl)propanamide (4-14e).\]

According to general protocol A, 4-14e (98.5 mg, 24\%) was obtained as a white solid: \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.57 (d, \(J = 8.1\) Hz, 2H), 7.41 (s, 1H), 7.35 (d, 2H, \(J = 8.1\) Hz), 7.16 (d, 1H, \(J = 8.2\) Hz), 6.55 – 6.32 (m, 2H), 4.40 (d, 2H, \(J = 6.1\) Hz), 4.28 (s, 2H), 3.80 (s, 3H), 3.79 (s, 3H). \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 195.5, 161.0, 159.3, 158.7, 137.1, 130.3, 130.3, 125.6, 117.4, 104.1, 98.8, 55.5, 55.4, 43.1, 39.3. IR (film): 3407, 3324, 2927, 1675, 1507, 1327, 1106, 830, 593 cm\(^{-1}\). HRMS (ESI) \(m/z\) calcd for C\(_{19}\)H\(_{18}\)F\(_3\)NO\(_4\) [M+H]\(^+\) 382.1261, found 382.1257. Rf = 0.69 (40\% EtOAc/Hexanes); mp = 120-121°C.

\[N-(2,4\text{-Dimethoxybenzyl})-3-(2\text{-nitrophenyl})-2\text{-oxopropanamide (4-14f).}\]

According to general protocol A, 4-14f (0.72 g, 84\%) was obtained as a slightly yellow solid: \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 8.13 (dd, 1H, \(J = 8.2, 1.2\) Hz), 7.67 – 7.41 (m, 2H), 7.35 (s, 1H), 7.32 – 7.11 (m, 2H), 6.44 – 6.41 (m, 2H), 4.66 (s, 2H), 4.43 (d, 2H, \(J = 6.1\) Hz), 3.81 (s, 3H), 3.79 (s, 3H). \(^{13}\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 194.5, 160.9, 159.3, 158.7, 148.7, 133.8, 133.7, 130.7, 129.8, 128.8, 125.5, 117.5, 104.0, 98.7, 55.5, 55.4, 42.9, 39.2. IR (film): 3407, 2936, 1684, 1523, 1345, 1036, 725 cm\(^{-1}\). HRMS (ESI) \(m/z\) calcd for C\(_{18}\)H\(_{18}\)N\(_2\)O\(_6\) [M+H]\(^+\) 359.1238, found 359.1236. Rf = 0.53 (50\% EtOAc/Hexanes); mp = 79-81°C.
3-(3,5-Dibromo-4-methoxyphenyl)-N-(2,4-dimethoxybenzyl)-2-oxopropanamide (4-14g).

According to general protocol A, 4-14g (76.0 mg, 62%) was obtained as a white gum:

$^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.39 (s, 1H), 7.35 (s, 2H), 7.14 (d, 1H, $J = 8.1$ Hz), 6.55 – 6.29 (m, 2H), 4.38 (d, 1H, $J = 6.1$ Hz), 4.11 (s, 2H), 3.84 (s, 3H), 3.80 (s, 3H), 3.77 (s, 3H).

$^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 195.2, 160.9, 159.1, 158.7, 153.3, 134.0, 131.6, 130.7, 118.1, 117.3, 104.0, 98.7, 60.6, 55.5, 55.4, 41.7, 39.3. IR (film): 3645, 2932, 1683, 1509, 1208, 993, 738 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{19}$H$_{19}$Br$_2$NO$_5$ [M+H]$^+$ 499.9703, found 499.970. Rf = 0.23 (20% EtOAc/Hexanes).

N-(2,4-Dimethoxybenzyl)-3-(4-methoxyphenyl)-2-oxopropanamide (4-14h).

According to general protocol A, 4-14h (0.56 g, 32%) was obtained as an orange oil:

$^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.37 (s, 1H), 7.18 – 7.13 (m, 3H), 6.94 – 6.76 (m, 2H), 6.53 – 6.30 (m, 2H), 4.38 (d, 2H, $J = 6.1$ Hz), 4.15 (s, 2H), 3.81 (s, 3H), 3.79 (s, 3H), 3.78 (s, 3H).

$^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 196.5, 160.9, 159.7, 158.8, 158.7, 131.0, 130.7, 124.8, 117.6, 114.2, 104.0, 98.7, 55.5, 55.4, 42.4, 39.2. IR (film): 3409, 2935, 1683, 1509, 1249, 1034, 824 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{19}$H$_{21}$NO$_5$ [M+H]$^+$ 344.1493, found 344.1490.
According to general protocol A, 4-14i (0.10 g, 27%) was obtained as a yellow oil: \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 8.15 (d, 2H, \(J = 8.8\) Hz), 7.38 (d, 2H, \(J = 8.8\) Hz), 7.38 (s, 1H), 7.14 (d, 1H, \(J = 8.2\) Hz), 6.50 – 6.17 (m, 2H), 4.39 (d, 2H, \(J = 6.1\) Hz), 4.32 (s, 2H), 3.80 (s, 3H), 3.77 (s, 3H). \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 195.0, 161.0, 159.1, 158.7, 147.2, 140.6, 130.8, 117.3, 104.1, 98.7, 55.5, 55.4, 43.1, 39.3. IR (film): 3404, 2938, 1682, 1518, 1346, 1036, 728 cm\(^{-1}\). HRMS (ESI) \(m/z\) calcd for C\(_{18}\)H\(_{18}\)N\(_2\)O\(_6\) [M+H]\(^+\) 359.1238, found 359.1229. Rf = 0.49 (40% EtOAc/Hexanes).

**General deprotection procedure for preparation of 2-oxo-3-arylpropanamides.**

**General protocol B:**

A solution of the corresponding \(N\)-(2,4-dimethoxybenzyl)-3-aryl-2-oxopropanamide (0.03M) in TFA/THF (2:1, v/v) was heated at 70 °C for 1 h. The reaction mixture then was allowed to warm to room temperature, quenched with a saturated solution of NaHCO\(_3\), and diluted with EtOAc. The organic layer was separated and the aqueous layer was washed with EtOAc (3x). The combined organic layers were concentrated *in vacuo* and the crude oily residue purified by flash chromatography on SiO\(_2\) (5-65% EtOAc/hexanes).
2-Oxo-3-phenylpropanamide (4-11).

According to general protocol B, 4-11 (58.9 mg, 57%) was obtained as a white solid: 
$^1$H NMR (300 MHz, DMSO-$d_6$) δ 7.98 (s, 1H), 7.72 (s, 1H), 7.61 – 6.85 (m, 5H), 4.14 (s, 2H). $^{13}$C NMR (101 MHz, DMSO-$d_6$) δ 196.8, 163.2, 133.9, 129.9, 128.3, 126.7, 42.9. IR (film): 3403, 1731, 1658, 1061, 695 cm$^{-1}$. HRMS (ESI) m/z calcd for C$_9$H$_9$NO$_2$ [M+Na]$^+$ 186.0525, found 186.0525; mp = 99-101 °C.

2-Oxo-3-(p-tolyl)propanamide (4-15a).

According to general protocol B, 4-15a (68.9 mg (64%) was obtained as a yellow solid, $^1$H NMR (300 MHz, CDCl$_3$) δ 7.14 (s, 4H), 6.87 (s, 1H), 6.09 (s, 1H), 4.17 (s, 2H), 2.34 (s, 1H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 195.8, 162.2, 137.1, 129.8, 129.5, 129.4, 42.6, 21.2. IR (film): 3409, 1732, 1654, 1335, 790 cm$^{-1}$. HRMS (ESI) m/z calcd for C$_{10}$H$_{11}$NO$_2$ [M+Na]$^+$ 200.0682, found 200.0682; mp = 107-109 °C.
3-[(4-(Tert-butyl)phenyl)-2-oxopropanamide (4-15b).

According to general protocol B, 4-15b (67.1 mg, 63%) was obtained as a slightly pink solid: $^1$H NMR (300 MHz, CDCl$_3$) δ 7.74 (d, 2H, $J = 8.3$ Hz), 7.42 (d, 2H, $J = 8.6$ Hz), 6.69 (s, 1H), 6.22 (s, 1H), 1.33 (s, 9H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 195.7, 162.2, 150.3, 129.6, 129.4, 125.8, 42.5, 34.6, 31.4. IR (film): 3407, 2962, 1679, 1473, 1205, 812, 630 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{13}$H$_{17}$NO$_4$ [M+H]$^+$ 220.1332, found 220.1328; mp = 91-93 °C.

3-[(4-Fluorophenyl)-2-oxopropanamide (4-15c).

According to general protocol B, 4-15c (53.5 mg, 49%) was obtained as a yellow solid: $^1$H NMR (300 MHz, CDCl$_3$) δ 7.36 – 7.11 (m, 2H), 7.14 – 6.83 (m, 2H), 6.80 (s, 1H), 5.53 (s, 1H), 4.19 (s, 2H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 195.4, 163.9, 161.9, 160.6, 131.6, 131.5, 128.2, 115.9, 115.6, 42.2. IR (film): 3403, 1718, 1667, 1228, 806, 608 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_9$H$_8$FNO$_2$ [M+H]$^+$ 182.0612, found 182.0612; mp = 109-111 °C.
According to general protocol B, **4-15d** (82.2 mg, 72%) was obtained as a slightly pink solid: $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.38 – 7.13 (m, 4H), 6.80 (s, 1H), 5.49 (s, 1H), 4.19 (s, 2H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 194.1, 160.8, 132.5, 130.3, 130.0, 128.0, 41.4. IR (film): 3391, 1789, 1651, 1326, 796, 656 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_9$H$_8$ClNO$_2$ [M+H]$^+$ 198.0316, found 198.0316; mp = 107-110 °C.

According to general protocol B, **4-15e** (98.5 mg, 70%) was obtained as an off-white solid: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.60 (d, 2H, $J = 8.1$ Hz), 7.37 (d, 2H, $J = 8.0$ Hz), 6.82 (s, 1H), 5.71 (s, 1H), 4.28 (s, 2H). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 194.8, 161.5, 136.7, 130.4, 125.8, 42.8. IR (film): 3391, 3178, 1729, 1661, 1333, 1105, 808, 678 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{10}$H$_8$F$_3$NO$_2$ [M+H]$^+$ 232.0579, found 232.0578. Rf = 0.45 (50% EtOAc/Hexanes); sublimes at 150 °C.
3-(2-Nitrophenyl)-2-oxopropanamide (4-15f).

According to general protocol B, 4-15f (62.5 mg (54%) was obtained as a yellow solid: $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 8.12 (dd, 1H, $J = 8.2, 1.3$ Hz), 8.05 (s, 1H), 7.81 (s, 1H), 7.74 (m, 1H), 7.56 (m, 2H), 4.60 (s, 2H). $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ 195.3, 162.4, 148.4, 134.1, 134.0, 130.3, 128.7, 124.9, 42.0. IR (film): 3382, 1667, 1520, 1341, 1033, 700 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_9$H$_8$N$_2$O$_4$ [M+H]$^+$ 209.0557, found 209.0555. Rf = 0.32 (50% EtOAc/Hexanes); mp = 148-151 °C.

3-(3,5-Dibromo-4-methoxyphenyl)-2-oxopropanamide (4-15g).

According to general protocol B, 4-15g (30.0 mg, 61%) was obtained as a clear gum: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.39 (s, 2H), 6.85 (s, 1H), 5.88 (s, 1H), 4.12 (s, 2H), 3.87 (s, 3H). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 194.6, 161.4, 153.5, 134.0, 131.2, 118.3, 60.8, 41.5. IR (film): 3454, 3340, 1697, 1473, 1260, 991, 740 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{10}$H$_9$Br$_2$NO$_3$ [M+H]$^+$ 349.9022, found 349.9019. Rf = 0.33 (40% EtOAc/Hexanes).
3-(4-Methoxyphenyl)-2-oxopropanamide (4-15h).

According to general protocol B, 4-15h (69.4 mg, 62%) was obtained as a yellow solid: $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.17 (d, 2H, $J$ = 8.3 Hz), 6.87 (d, 2H, $J$ = 8.6 Hz), 6.79 (s, 1H), 5.50 (s, 1H), 4.15 (s, 2H), 3.79 (s, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 195.7, 162.1, 158.9, 131.0, 124.4, 114.3, 55.4, 42.1. IR (film): 3401, 1716, 1666, 1029, 805 cm$^{-1}$. HRMS (ESI) m/z calcld for C$_{10}$H$_{11}$NO$_3$ [M+H]$^+$ 184.0812, found 184.0813; mp = 110-113 °C.

3-(4-Nitrophenyl)-2-oxopropanamide (4-15i).

According to general protocol B, 4-15i (34.9 mg, 57%) was obtained as a yellow gum: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.20 (d, 2H, $J$ = 8.8 Hz), 7.42 (d, 2H, $J$ = 8.7 Hz), 6.82 (s, 1H), 5.64 (s, 1H), 4.34 (s, 2H). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 194.3, 161.2, 140.1, 131.0, 124.0, 42.8. IR (film): 3426, 1672, 1519, 1348, 1109, 801 cm$^{-1}$. HRMS (ESI) m/z calcld for C$_9$H$_8$N$_2$O$_4$ [M+H]$^+$ 209.0557, found 209.0557. Rf = 0.22 (50% EtOAc/Hexanes).
General procedure for preparation of 4-oxazolidinines via dehydration of 2-oxo-3-arylpropanamides. General protocol C:

To a solution of the corresponding 2-oxo-3-arylpropanamide (1 equiv., 0.1 M) in TFA/DCM (1:1, v/v) under an inert atmosphere was added hexanal (2 equiv.) and the reaction was stirred at room temperature for 17–24 h. The reaction mixture was concentrated \textit{in vacuo} and the resultant crude oil was purified by flash chromatography on SiO$_2$ (1-60%, EtOAc/hexanes).

\[
\text{(Z)-5-Benzylidene-2-pentyloxazolidin-4-one (4-17).}
\]

According to general protocol C, 4-17 (49.6 mg, 48\%) was obtained as an off-white film: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 9.34 (s, 1H), 7.82 – 7.56 (m, 2H), 7.48 – 7.08 (m, 3H), 6.22 (s, 1H), 5.65 (t, 1H, $J = 5.2$ Hz), 2.03 – 1.71 (m, 2H), 1.63 – 1.10 (m, 7H), 0.92 (t, 3H, $J = 7.0$ Hz). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 166.0, 144.0, 133.9, 129.2, 128.6, 127.6, 103.2, 89.2, 36.6, 31.5, 22.8, 22.6, 14.1. IR (film): 3208, 2930, 1713, 1365, 1179, 762, 693 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{19}$NO$_2$ [M+H]$^+$ 244.1343, found 244.1334
(Z)-5-(4-Methylbenzylidene)-2-pentyloxazolidin-4-one (4-18).

According to general protocol C, 4-18 (44.4 mg, 47%) was obtained as a yellow oil: $^1$H NMR (300 MHz, CDCl$_3$) δ 8.08 (s, 1H), 7.57 (d, 2H, $J = 8.1$ Hz), 7.18 (d, 2H, $J = 8.2$ Hz), 6.23 (s, 1H), 5.64 (t, 1H, $J = 5.2$ Hz), 2.36 (s, 3H), 2.05 – 1.74 (m, 2H), 1.64 – 1.17 (m, 9H), 0.91 (t, 3H, $J = 7.0$ Hz). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 166.3, 143.3, 137.7, 130.9, 129.4, 129.2, 103.6, 89.2, 36.6, 31.5, 22.8, 22.6, 21.5, 14.1. IR (film): 3186, 2929, 1715, 1365, 1182, 811 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{16}$H$_{21}$NO$_2$ [M+H]$^+$ 260.1645, found 260.1644.

(Z)-5-(4-Methylbenzylidene)-2-pentyloxazolidin-4-one (4-19).

According to general protocol C, 4-19 (40.5 mg, 47%) was obtained as a clear oil: $^1$H NMR (300 MHz, CDCl$_3$) δ 8.02 (s, 1H), 7.62 (d, 2H, $J = 8.5$ Hz), 7.40 (d, 2H, $J = 8.4$ Hz), 6.24 (s, 1H), 5.63 (t, 1H, $J = 5.1$ Hz), 1.95 – 1.70 (m, 2H), 1.62 – 1.02 (m, 22H), 0.91 (t, 3H, $J = 7.0$ Hz). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 166.3, 150.9, 143.4, 131.0, 129.0, 125.6, 103.4, 89.2, 36.6, 34.8, 31.5, 31.4, 22.8, 22.6, 14.1. IR (film): 3192, 2959, 1715, 1365, 1182, 861, 570 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{19}$H$_{27}$NO$_2$ [M+H]$^+$ 302.2115, found 302.2110.
(Z)-5-(4-Fluorobenzylidene)-2-pentyloxazolidin-4-one (4-20).

According to general protocol C, 4-20 (41.2 mg, 57%) was obtained as a yellow solid: $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 8.34 (s, 1H), 7.77 – 7.48 (m, 2H), 7.05 (t, 2H, $J = 8.8$ Hz), 6.19 (s, 1H), 5.63 (t, 1H, $J = 5.2$ Hz), 2.12 – 1.70 (m, 2H), 1.67 – 1.20 (m, 7H), 0.91 (t, $J = 6.9$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 165.8, 163.7, 160.4, 143.6, 130.9, 130.8, 130.1, 115.8, 115.5, 102.1, 89.2, 36.6, 31.5, 22.8, 22.6, 14.1. IR (film): 3160, 2857, 1727, 1508, 1375, 1223, 808, 500 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$FNO$_2$ [M+H]$^+$ 264.1397, found 264.1392; mp = 114-116 °C.

(Z)-5-(4-Chlorobenzylidene)-2-pentyloxazolidin-4-one (4-21).

According to general protocol C, 4-21 (44.4 mg, 63%) was obtained as white solid: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.53 (s, 1H), 7.66 – 7.47 (m, 2H), 7.41 – 7.29 (m, 2H), 6.17 (s, 1H), 5.64 (t, 1H, $J = 5.2$ Hz), 1.98 – 1.73 (m, 2H), 1.62 – 1.22 (m, 7H), 0.91 (t, 3H, $J = 7.0$ Hz). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 165.7, 144.3, 133.2, 132.4, 130.4, 128.8, 102.0, 89.3,
36.6, 31.5, 22.8, 22.6, 14.1. IR (film): 3192, 2930, 1715, 1489, 1369, 1092, 857 cm\(^{-1}\). HRMS (ESI) \(m/z\) calcd for \(\text{C}_{15}\text{H}_{18}\text{ClNO}_2 [\text{M+H}]^+\) 280.1099, found 280.1107; mp = 103–105 °C.

\[
\text{(Z)-2-Pentyl-5-(4-(trifluoromethyl)benzylidene)oxazolidin-4-one (4-22).}
\]

According to general protocol C, 4-22 (8.7 mg, 32%) was as a brown oil: \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 8.92 (s, 1H), 7.76 (d, 2H, \(J = 8.0\) Hz), 7.61 (d, 2H, \(J = 8.1\) Hz), 6.26 (s, 1H), 5.71 (br s, 1H), 1.88 (s, 2H), 1.71 – 1.29 (m, 7H), 0.91 (t, 3H, \(J = 6.6\) Hz). \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 165.8, 145.3, 137.1, 129.3, 125.6, 122.9, 102.5, 89.7, 36.4, 31.5, 22.8, 22.6, 14.0. IR (film): 3213, 2930, 1714, 1324, 1117, 861 cm\(^{-1}\). HRMS (ESI) \(m/z\) calcd for \(\text{C}_{16}\text{H}_{18}\text{F}_3\text{NO}_2 [\text{M+H}]^+\) 312.1217, found 312.1214. Rf = 0.27 (40% EtOAc/hexanes).

\[
\text{(Z)-5-(2-Nitrobenzylidene)-2-pentyloxazolidin-4-one (4-23).}
\]

According to general protocol C, 4-23 (26.1 mg (30%) as a slightly yellow film: \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 8.75 (s, 1H), 8.12 (dd, 1H, \(J = 8.0, 1.3\) Hz), 7.90 (dd, 1H, \(J = 8.2, 1.3\) Hz), 7.66 – 7.47 (m, 1H), 7.44 – 7.30 (m, 1H), 6.68 (s, 1H), 5.65 (t, , -1H \(J = 5.3\) Hz), 2.00 – 1.76 (m, 2H), 1.62 – 1.20 (m, 7H), 0.92 (t, 3H, \(J = 7.0\) Hz). \(^{13}\)C NMR (101 MHz,
CDCl$_3$) $\delta$ 164.5, 148.5, 146.7, 132.6, 130.8, 128.3, 127.8, 124.8, 96.4, 89.5, 36.5, 31.5, 22.8, 22.6, 14.1. IR (film): 3163, 2928, 1727, 1693, 1521, 1344, 1110, 751 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$N$_2$O$_4$ [M+H]$^+$ 291.1339, found 291.1332. Rf = 0.2 (30% EtOAc/hexanes).

![Chemical Structure](image)

(Z)-5-(3,5-Dibromo-4-methoxybenzylidene)-2-pentyloxazolidin-4-one (4-24).

According to general protocol C, 4-24 (50 mg, 50%) was obtained as a white film: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.98 (s, 1H), 7.79 (s, 2H), 6.02 (s, 1H), 5.66 (br s, 1H), 3.89 (s, 3H), 1.89 – 1.74 (m, 1H), 1.43 (m, 7H), 0.91 (t, 3H, $J$ = 6.8 Hz). $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 165.1, 153.2, 144.9, 134.1, 132.9, 132.7, 118.2, 100.2, 89.6, 60.9, 36.5, 31.4, 22.8, 22.6, 14.0. IR (film): 3156, 2928, 1720, 1670, 1472, 1373, 1003, 740 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$Br$_2$O$_3$ [M+H]$^+$ 431.9804, found 431.9801.

![Chemical Structure](image)

(Z)-5-(4-Methoxybenzylidene)-2-pentyloxazolidin-4-one (4-25).

According to general protocol C, 4-25 (26.9 mg, 67%) was obtained as a clear oil: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 9.02 (s, 1H), 7.73 – 7.49 (m, 2H), 7.11 – 6.73 (m, 2H), 6.18 (s, 1H), 5.62 (t, 1H, $J$ = 5.2 Hz), 3.83 (s, 3H), 1.93 – 1.75 (m, 2H), 1.61 – 1.19 (m, 7H), 0.91 (t, 3H, $J$ = 6.8 Hz). HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$O$_4$ [M+H]$^+$ 268.1096, found 268.1093.
3H, $J = 7.0$ Hz). $^{13}$C NMR (101 MHz, CDCl$_3$) δ 166.1, 159.1, 142.5, 130.6, 126.7, 114.1, 103.1, 103.0, 88.9, 55.4, 36.7, 31.6, 22.8, 22.6, 14.1. IR (film): 3189, 2931, 1709, 1511, 1363, 1251, 1177, 857 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{16}$H$_{21}$NO$_3$ [M+H]$^+$ 276.1594, found 276.1591. Rf = 0.44 (50% EtOAc/hexanes).

![Chemical Structure](image)

(Z)-5-(4-Nitrobenzylidene)-2-pentyloxazolidin-4-one (4-26).

According to general protocol C, 4-26 (17.6 mg, 37%) was obtained as yellow film:

$^1$H NMR (400 MHz, CDCl$_3$) δ 8.97 (s, 1H), 8.30 – 8.00 (m, 2H), 7.90 – 7.70 (m, 2H), 6.26 (s, 1H), 5.72 (t, 1H, $J = 5.3$ Hz), 1.89 (m, 2H), 1.73 – 1.19 (m, 7H), 0.92 (t, 3H, $J = 7.0$ Hz).

$^{13}$C NMR (101 MHz, CDCl$_3$) δ 164.7, 146.8, 146.3, 140.6, 129.5, 124.0, 100.9, 100.9, 89.8, 36.5, 31.5, 22.8, 22.6, 14.1. IR (film): 3157, 2858, 1722, 1591, 1513, 1336, 862 cm$^{-1}$. HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$N$_2$O$_4$ [M+Na]$^+$ 313.1159, found 313.1157. Rf = 0.2 (40% EtOAc/hexanes).
4.6 References


(41) Plöchl, J. Ber. 1884, 17, 1616.


CHAPTER 5

First Total Synthesis of Synoxazolidinones A and B
5.1 Abstract

A five-step total synthesis of the marine natural product synoxazolidinone A was achieved through a diastereoselective imine acylation/cyclization cascade. Description of the efforts in the search for a suitable imine and activated acid intermediates are provided. Synoxazolidinones A and B along with additional series of 4-oxazolidinone analogues were also prepared to explore the potential of these 4-oxazolidinones as antimicrobial agents.

5.2 Introduction and Background

In order to overcome the limitation of our previous dehydration/cyclization method, that is being unable to access 4-oxazolidinones containing substitution (α-Cl or terminal guanidine) on the alkyl chain attached to C2 of the heterocycle (Table 4.6, Table 4.7) in satisfactory yields and purity, we explored other conditions to generate the desired \(N\)-acyliminium ion (Scheme 5.1).

\[
\begin{align*}
\text{5-3} & \quad + \quad \text{5-4} \quad \xrightarrow{R_1, R_2 = ?} \quad \text{4-1} \\
\text{5-1} & \quad \xrightarrow{R_1, R_3 = \text{H}} \quad \text{5-2}
\end{align*}
\]

**Scheme 5.1:** Potential addition pathways for acyliminium intermediate 4-1.
One of the alternative ways to form acyliminiums in situ is direct acylation of imines (Chapter 4, Section 4.2.2). A variety of nitrogen-containing heterocyclic scaffolds have been prepared through subsequent intramolecular trapping of the N-acyliminium ion with appended S- and O-nucleophiles, including oxazolines, thiazolines and some oxazolidinones.\(^1,2\) Therefore we next proposed a different disconnection which requires the formation of an activated enolizable α-keto carboxylic acid (5-3) and subsequent reaction with an enolizable α-chloroimine (5-4, \(R_3 = -\text{Cl}\)). Analogous to the previous approach, we believe that the enol tautomer of the intermediate undergoes cyclization via addition of the \(O\)-atom to form the oxazolidin-4-one (4-2) ring in 5-endo fashion (Scheme 5.1). At the outset of our explorations of this cyclization pathway we also considered the possibility of the formation of alternative cycloadduct (5-2) arising from C-addition to the reactive iminium species. Indeed, intermolecular addition of keto esters\(^3\) to imines has been reported to yield medicinally relevant 3-hydroxy-1,5-dihydro-2H-pyrrol-2-one heterocycles such as 5-8 (Scheme 5.2) through an addition/cyclization/dehydration sequence.\(^4\)

![Scheme 5.2: Synthesis of 3-hydroxy-1,5-dihydro-2H-pyrrol-2-one 5-8.](image)

In addition to poor yields for \(\alpha\)-Cl substituted aldehydes, another drawback in our previous dehydration/cyclization approach is absence of diastereoselectivity (Scheme 5.3).
Scheme 5.3: Possible origin of lack of the diastereoselectivity and low reaction yields for α-substituted aldehydes in our previous approach described in Chapter 4.

The lack of the activation and steric crowding of the iminium nitrogen (5-9, 5-10) further supported our next generation proposal to use N-substituted imines (5-4). We envisioned that a more sterically demanding and/or chiral substituent present on the nitrogen of 5-11 at the moment of cyclization would influence the stereochemical outcome as well as improve the yield of our target Cl-containing 4-oxazolidinones.

5.3 Results and Discussion

5.3.1 Acylation of N-substituted Aldimines with Activated α-keto-acid Derivatives

As an initial test of the proposed reaction, phenylpyruvic acid chloride was treated with variously N-substituted imines to explore the feasibility and addition chemoselectivity of this process.
5.3.1.1 Acylation of Diisobutylaluminum- and Trimethylsilyl-imines

A major concern in reactions involving aldimines is their diminished reactivity towards nucleophiles in comparison with corresponding aldehydes. Various methods have been developed to increase the electrophilic properties of imine derivatives. One of the ways to activate imines for the addition of nucleophiles involves placing strongly electron-withdrawing groups (such as for example tosyl-) on the nitrogen atom; however, subsequent cleavage is necessary to obtain free amines or amides. In most cases removal of the nitrogen substituent is not facile and often strong bases or oxidizing agents are employed.\(^5\) Due to the harsh conditions required for the removal of electron-withdrawing \(N\)-protecting groups the use of \(N\)-metallo\(^6\) and \(N\)-silylimines\(^7\) have been explored.

Treatment of aldehydes with one equivalent of lithium hexamethyldisilylamine (LiHMDS) at low temperatures allows formation of silyl imines in situ. Highly reactive \(N\)-silyl imines can be subsequently trapped by a variety of external nucleophiles at the azomethine carbon (Scheme 5.4).\(^8\)

![Scheme 5.4: Synthesis of \(N\)-silylimines.](image)

The in situ formation of the silyl imine of hexanal (4-16) and treatment with phenylpyruvic acid chloride (5-17) in THF at low temperatures successfully provided 4-oxazolidinone compound 4-17 in 25% yield (Scheme 5.5). Unfortunately, no product formation was observed when we used guanidine containing aldehyde 4-31 as a substrate.
Deprotonation of 4-31 and its decomposition under these basic reaction conditions was observed. Application of N-trimethylsilyl imines is generally limited to non-enolizable aldehydes. Silylimines of carbonyl compounds containing acidic methylene protons upon reaction with nucleophiles usually give poor yields of the addition products, due to their competitive deprotonation, which may account for our failure to obtain 4-33 and 4-27.

Scheme 5.5: Acylation of N-silylimines with phenylpyruvic acid chloride 5-17.

N-diisobutylaluminium imines can also be used as an alternative for highly reactive imine derivatives. Additionally, they are prepared under somewhat milder reductive conditions. In our hands partial reduction of benzonitrile 5-18 with DIBAL-H in THF at -70 °C furnished an N-aluminoimine species (Scheme 5.6). In situ acylation with 5-17 most likely proceeded with formation of an adduct 5-19, which upon warming up to room temperature did not provide the cyclization product 5-20. Complexation of the internal O-nucleophile with oxophilic aluminum may prevent the cyclization event.
Scheme 5.6: Acylation of N-diisobutylaluminium imine with phenylpyruvic acid chloride.

5.3.1.2 Acylation of Chiral N-tert-butanesulfinimines

*N*-tert-butanesulfinyl is widely employed as a chiral directing group which also activates imines for the addition of many different classes of nucleophiles.\(^{10,11}\) An attractive feature of this group is that it can be easily cleaved under mild acidic conditions after nucleophilic addition is complete. A high-yielding method for the preparation of various tert-butanesulfinyl aldimines has been developed by Ellman *et al.*\(^{12}\) *N*-tert-butanesulfinimines can be prepared via condensation of aldehydes with *tert*-butanesulfinamide in presence of copper (II) sulfate which serves as a Lewis acid catalyst and dehydrating agent. Importantly, this class of imines is stable to silica purification and can be stored for long periods without decomposition.

Model sulfinyl aldimine 5-23 was obtained in excellent yield according to a literature procedure starting from hexanal (4-16) via dehydration with chiral 5-22 (Scheme 5.7). A one pot α-chlorination of hexanal (4-16) followed by condensation with 5-22 afforded α-Cl *tert*-butanesulfinyl aldimine 5-24 in 86% yield over two steps as a 1:1 mixture of diastereomers.
Scheme 5.7: Preparation of *N*-tert-butanesulfinimine 5-23 and 5-24.

Excited to explore reactivity of these stable imines in our acylation/cyclization cascade we began with their acylation with the acid chloride derived from 4-10. We observed significant formation of 4-oxazolidinone 4-17 after several hours of stirring of the reaction mixture at room temperature. Unfortunately, we were not able to easily isolate product after aqueous work-up followed by purification on regular phase silica gel. Only small amounts of product could be recovered from an oven dried PTLC plate after direct loading of the reaction mixture and developing the plate in the gradient of EtOAc-Hex, with racemic 4-17 obtained in 15% yield (Entries 1 and 2, Table 5.1). The corresponding 4-oxazolidinones 5-26 and 5-25 bearing a chiral auxiliary on the nitrogen atom were not observed (according to HPLC-MS of the reaction mixture). Initially, we reasoned that the auxiliary was cleaved off during cyclization due to the acidic reaction media as the acid chloride formation via Vilsmeier-Haack type intermediate (employing oxalyl chloride and DMF) proceeds with liberation of hydrochloric acid. In order to prevent undesired cleavage of the chiral auxiliary we next explored different additives and acid scavengers.
Basic additives such as TEA or DABCO and proton scavengers such as DTBMP, NaHCO₃ or proton-sponge did not promote formation of chiral 4-oxazolidinone bearing the auxiliary and the material isolated was still racemic (Entry 4, Table 5.1).

**Table 5.1**: Acylation of N-tert-butanesulfinimines 5-23 and 5-24 with phenylpyruvic acid chloride 5-17.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Comment</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5-23, THF/DCM, 0 °C – rt, 3 h</td>
<td>traces⁹ of 4-17</td>
<td>ND</td>
</tr>
<tr>
<td>2</td>
<td>5-23, 0.1 M, THF/DCM, 0 °C – rt, 1:1, 8 h</td>
<td>racemic 4-17</td>
<td>15⁹</td>
</tr>
<tr>
<td>3</td>
<td>5-23, THF, rt, 1:2, 6 h</td>
<td>racemic 4-17</td>
<td>46⁹</td>
</tr>
<tr>
<td>4</td>
<td>5-23, 0.1 M, THF, rt, 1:2, additive:</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>a) TEA</td>
<td>traces⁹ of 4-17, decomposition of SM</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>b) DABCO</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>c) Pyridine</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>d) Proton-sponge</td>
<td>complex mixture⁹ containing 4-17 and SM</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>e) TBACl</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>f) DTBMP</td>
<td>racemic 4-17</td>
<td>45⁹</td>
</tr>
<tr>
<td></td>
<td>g) NaHCO₃</td>
<td></td>
<td>9⁹</td>
</tr>
<tr>
<td>5</td>
<td>5-23, 0.2 M, THF/DCM, rt, 1:1, 3 h</td>
<td>racemic 4-17</td>
<td>60⁹</td>
</tr>
<tr>
<td>6</td>
<td>5-24, THF/DCM, rt, 1:1, 6 h</td>
<td>complex mixture⁹ containing 4-27 and SM</td>
<td>ND</td>
</tr>
</tbody>
</table>

⁹ – determined by HPLC-MS analysis, b – purified by reverse phase flash chromatography, c – purified preparative thin layer chromatography.
Employing two equivalents of acid chloride and reverse phase purification resulted in isolation of racemic 4-oxazolidinone 4-17 in 46% yield (Entry 3, Table 5.1). The yield of 4-17 appeared higher in more concentrated reaction mixtures with no additives (Entry 5, Table 5.1).

The fact that material isolated from the reaction was always racemic may imply that the auxiliary is absent at the moment of ring closure and can be cleaved either before or after the acylation event. Therefore, we explored other methods of activation of the phenylpyruvic acid for nucleophilic attack by imines, anticipating that acid-free activation of 4-10 will help to preserve the chiral auxiliary in our cyclization step and achieve stereoselectivity.

We next explored most of the conventional methods of activation of the phenylpyruvic acid such as acylimidazole,\textsuperscript{13} \textit{O}-acylisourea,\textsuperscript{14,15} and mixed phosphoric\textsuperscript{16} and carbonic\textsuperscript{17} anhydrides. Activation of 4-10 followed by treatment with imine 5-23 failed to provide 4-oxazolidinone products 4-17 or 5-25 (Entries 1,3 and 5-7, Table 5.2). Conditions involving higher temperatures or acyl halide formed using thionyl chloride\textsuperscript{18} or cyanuryl chloride\textsuperscript{19} led to decomposition of starting material (Entries 2, 4, 8, and 9, Table 5.2).

It is possible that less electrophilic (compared to 5-17) activated acyl intermediates of 4-10 do not react with the electron-poor nitrogen of the imine 5-23 and only when the sulfinyl auxiliary is cleaved during \textit{N}-acylation under the acidic conditions employed in the acid chloride reaction can take place. If the acidic media is actually required for the reaction to occur, then the mechanism shown in Scheme 5.8 may explain the lack of asymmetric induction and failure of non-acidic coupling conditions to yield 5-25.
Table 5.2: Reaction of \(N\text{-}\text{tert}\)-butanesulfinimine 5-23 with various phenylpyruvic acid activated species.

\[
\begin{array}{|c|c|c|c|}
\hline
\text{Entry} & \text{Conditions} & \text{Comment} & \text{Yield (\%)} \\
\hline
1 & \text{CDI, THF, DMAP, rt} & \text{SM} & \text{NR} \\
2 & \text{CDI, THF, DMAP, } \Delta & \text{Decomposition of SM} & \text{NR} \\
3 & \text{DCC, DMAP, DCM, 0 °C – rt} & \text{SM} & \text{NR} \\
4 & \text{DCC, DMAP, DCM, } \Delta & \text{Decomposition of SM} & \text{NR} \\
5 & \text{EDC, TEA, DCM, 0 °C – rt} & \text{SM} & \text{NR} \\
6 & \text{T3P, Py, EtOAc/DCM, - 40 °C} & \text{SM} & \text{NR} \\
7 & \text{MeO(CO)Cl, NMM, DCM, - 25 °C} & \text{SM} & \text{NR} \\
8 & (\text{NCCl})_3, \text{NMM, MeCN, rt – 55 °C} & \text{Decomposition of SM} & \text{NR} \\
9 & \text{SOCl}_2, \text{DCM, } \Delta & \text{Decomposition of SM} & \text{NR} \\
10 & (\text{COCl})_2, \text{DMF}_{\text{(cat.)}}, \text{THF, 0 °C} & 4-17 & 60 \\
11 & \text{Ghosez’s reagent, THF, 0 °C} & \text{Side product 5-28} & 36 \\
\hline
\end{array}
\]

One would expect that similarly to \(t\text{BuCO}_2\)- (Boc-) groups, \(t\text{BuSO}\)- auxiliary would decompose to innocuous isobutylene and SO gas; however, while Boc- readily liberates CO\(_2\) in acidic media, SO is very unstable and does not form that easily.\(^\text{20}\) Upon reaction with HCl, 5-23 most likely converts to 5-31 with release of 5-33. The unstable and reactive N-H imine 5-31 subsequently undergoes acylation with 5-17 which leads to the formation of racemic product 4-17.
Scheme 5.8: Proposed mechanism of loss of the auxiliary during the acylation of chiral tert-butanesulfinyl aldimines.

Although 4-17 formed in good yield (Entry 5, Table 5.1), attempts to acylate α-chlorinated substrate 4-24 failed multiple times (Entry 7, Table 5.1), most likely because α-chloro substitution of 5-31 is further destabilizing.

Interestingly, when we attempted to prepare the acid chloride under conditions that do not generate HCl by use of α-chloroenamines\(^{21}\) (Ghosez et al.) oxazolidin-4-one 4-17 did not form (Entry 11, Table 5.2). Instead, when phenylpyruvic acid 4-10 was treated with tetramethyl-α-chloroenamine (5-27) an unexpected white precipitate formed at 0 °C in THF, the structure of which was determined to be 5-28. (Scheme 5.9).

Scheme 5.9: Reaction of phenylpyruvic acid with Ghosez’s reagent (5-27).
Compound **5-28** has not been reported to date, though a very similar structural analogue was discovered very recently (*guignaric acid, Scheme 5.8*). **5-29** was isolated from *Guignardia bidwellii* (grape black rot fungus) and appears to have phytotoxic properties. We believe that **5-28** forms when the Vilsmeier-Haack type intermediate of phenylpyruvic acid is intercepted by the internal enol nucleophile via intramolecular attack of an iminium ion, in analogy to the mechanism we are proposing for the synthesis of our desired 4-oxazolidinone (Scheme 5.1).

To explore extension of this approach to the natural product targets, conditions optimized for model substrates (Entry 5, Table 5.1 and Entry 10, Table 5.2) were applied to **5-35** acid chloride of 3,5-dibromo-4-methoxyphenylpyruvic acid (**4-13g**) and guanidine containing *N*-tert-butanesulfinimine **5-34** (Scheme 5.10). **5-34** was prepared from previously synthesized aldehyde **4-31** via copper (II) mediated condensation in 78\% yield.

![Scheme 5.10: Synthesis of synoxazolidinone B via acylation of guanidine containing *N*-tert-butanesulfinimine **5-34**.](image)

One-pot acylation of imine **5-34** followed by Boc-deprotection of guanidine using 50\% trifluoroacetic acid solution in DCM gave natural product synoxazolidinone B (**2-27**) which was isolated after purification on reversed phase silica in low 22\% yield but high
purity. $^1$H NMR, $^{13}$C NMR and HRMS spectra of our synthetic synoxazolidinone B appeared identical to those reported in the literature.$^{24}$

Despite a moderate 17% yield over 3 steps starting from aldehyde 4-31 for synoxazolidinone B, we were excited to move forward and see if we could transform $\alpha$-chloro aldehyde 4-30 into synoxazolidinone A (2-26). Employing copper (II) sulfate as the dehydrating agent was not successful in the condensation of 4-30 with tert-butanesulfinamide 5-22. An alternative method, employing reflux with titanium (IV) ethoxide in THF, provided unstable sulfanyl aldimine 5-36 in 51% yield (Scheme 5.11).$^{25}$ Unfortunately, acylation of 5-36 with 3,5-dibromo-4-methoxyphenylpyruvic acid chloride (5-35) derived from 4-13g provided only traces of synoxazolidinone A. This result is in accordance with the failure to prepare even simplified Cl-oxazolidinone 4-27 using our imine acylation approach (Entry 6, Table 5.1).

**Scheme 5.11:** Attempts at the synthesis of synoxazolidinone A via acylation of guanidine containing $N$-tert-butanesulfinimine 5-36.
In summary, we were unable to control the stereoselectivity in the acylation of the \(N\)-\textit{tert}-butanesulfinimines, and thus isolated racemic synoxazolidinone B in moderate yield. Unfortunately, only traces of Cl-containing oxazolidine-4-ones were observed; milder method or more stable intermediates, addressing the substitution pattern of natural products, were needed.

5.3.1.3 Acylation of \(N\)-alkylimines

Learning from our experience with various protected imines (\(N\)-TMS, \(N\)-Al(iBu)\(_2\), \(N\)-S(O)\(i\)Bu) we came to the conclusion that \(N\)-alkyl aldimes may be more suitable precursors in our acylation/cyclization cascade. The presence of the electron-donating alkyl group on the nitrogen of aldimes makes them reactive towards electrophiles. In contrast to \(N\)-TMS and \(N\)-Al(iBu)\(_2\) imines, \(N\)-alkyl imines are usually formed under mild, anhydrous and base-free conditions and are widely utilized in the synthesis of important \(N\)-containing heterocycles. For example, Evans \textit{et al.} used \(N\)-benzylaldimine 5-39, formed by condensation of the aldehyde 5-38 with amine 5-37 \textit{in situ}, for preparation of \(\beta\)-lactam 5-40 (Scheme 5.12).\(^{26}\)

\textbf{Scheme 5.12:} Synthesis of \(\beta\)-lactams via [2+2] cycloaddition of benzylaldimines and ketenes derived from acid chlorides
Reaction of the ketene electrophile (generated by deprotonation of the acyl chloride 5-41) with aldimine 5-39 at -78 °C in DCM proceeded to form the cis β-lactam with a high level of diastereocntrol and excellent yield.

To explore the feasibility and addition chemoselectivity of the proposed acylation/cyclization cascade, 2,4-dimethoxybenzyl imine 5-44 was treated with phenylpyruvic acid (4-10) activated by various activating agents. Coupling agents such as DCC or cyanuric chloride provided the undesired carbon addition product 5-43 exclusively in up to 57% yield. We cannot rule out an intermolecular imine addition process analogous to that reported; however, that mechanism appears unlikely under the conditions employed. By contrast, the acid chloride generated from oxalyl chloride/DMF provided the desired 4-oxazolidinone 5-42 as the major product from the reaction mixture (24–40% yield of isolated product). In addition to the acid chloride, pyBOP/Hunig’s base also proved effective for the preparation of the 4-oxazolidinone products in 20–30% yield (Scheme 5.13).

Scheme 5.13: Chemoselectivity of N-alkyl imine acylation/enol addition.

The complete switch in selectivity is striking and is possibly accounted for by an electrocyclization mechanism (rather than a 5-endo enol addition) in which the equilibrium of keto/enol tautomers, influenced by pH value and other factors, would dictate reaction selectivity. The mass balance in both the acid chloride and pyBOP reactions is high, with
the remainder of the material being lost through activated acid dimerization. Although the yields for this process are modest, the readily available starting materials, straightforward isolation, and one-step protocol allow ample material to be prepared through this approach. Furthermore, the carbon addition products, 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones (5-43) are commonly found in nature and possess a wide range of biological activities.30 N-alkyl imine acylation/enol addition provides a mild method to generate these heterocycles and provides compatibility with alkyl substituents that is not possible with current methods.3

With a route to the core heterocycle in hand, we turned our attention to the incorporation of the functional groups required for the synoxazolidinones (α-Cl or terminal guanidine). Our efforts were primarily centered around Cl-containing substrates as their formation suffered in all of our previous approaches. Additionally, the Cl-containing compounds are of greater interest from the perspective of their biological properties.

Condensation of benzylamines 5-48a-c with aldehydes 4-28 and 4-16 using magnesium sulfate as the dehydrating agent provided crude imines with purity sufficient for the next step. Subsequent acylation with phenylpyruvic acid chloride 5-17 resulted in successful formation of oxazolidin-4-ones 5-45a-c and 5-46a,b. In the case of α-chlorinated substrates, different diastereomeric ratios were observed depending on the substitution of the benzyl group (Table 5.3). Performing the cyclization at 0 °C in dichloromethane provided 5-45a as 2:1 mixture of diastereomers. When the temperature was lowered to -78 °C the diastereomeric ratio increased to 3.5:1 for the same substrate (Entry 1 and 2, Table 5.3). Interestingly, the presence of a chiral center on the benzylic methyl group (Me-, 5-48b) did
not change the diastereoselectivity of the reaction at 0 °C (2:1 \( dr \)) (Entries 3 and 4, Table 5.3), but increased it slightly at -78 °C (5-45b, 4:1 \( dr \)).

**Table 5.3**: Diastereoselectivity of the \( N \)-alkyl imine acylation/enol addition.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Product</th>
<th>Imine</th>
<th>( T, ^\circ C )</th>
<th>( dr^c )</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5-45a</td>
<td>5-48a</td>
<td>-78</td>
<td>3.5:1</td>
<td>22</td>
</tr>
<tr>
<td>2</td>
<td>5-45b</td>
<td>5-48b</td>
<td>0</td>
<td>2:1</td>
<td>5( d )</td>
</tr>
<tr>
<td>3</td>
<td>5-45c</td>
<td>5-48c</td>
<td>-78</td>
<td>4:1</td>
<td>36</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td></td>
<td>0</td>
<td>2:1</td>
<td>41</td>
</tr>
<tr>
<td>5</td>
<td>5-45c</td>
<td>5-47</td>
<td>-78</td>
<td>9:1</td>
<td>22</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td></td>
<td>0</td>
<td>3.5:1</td>
<td>25</td>
</tr>
<tr>
<td>7</td>
<td>5-46b</td>
<td></td>
<td>-78</td>
<td>4:1</td>
<td>17</td>
</tr>
<tr>
<td>8</td>
<td></td>
<td></td>
<td>0</td>
<td>3:1</td>
<td>32</td>
</tr>
<tr>
<td>9</td>
<td>5-46a</td>
<td></td>
<td>0( a )</td>
<td></td>
<td>38</td>
</tr>
<tr>
<td>10</td>
<td></td>
<td></td>
<td>0( b )</td>
<td></td>
<td>42</td>
</tr>
</tbody>
</table>

\( a – 1:2 \) acid chloride : imine; \( b – 2:1 \) acid chloride : imine; \( c – \) diastereomeric ratio was determined after purification, crude reaction mixtures contained impurities obstructing characteristic proton resonance frequencies; \( d – \) isolation was performed after three hours.
A far stronger impact on diastereoselectivity was observed when an ortho- substituent was introduced in the aromatic ring of the benzyl group. A 4:1 diastereomeric ratio was observed at 0 °C when R₁ was changed from H to –OMe (Entry 6, Table 5.3) and a 9:1 mixture of diastereomers was obtained when reaction was performed at -78 °C in DCM (Entry 5 and 6 Table 5.3). It is worth mentioning that the diastereomeric ratios were determined after isolation as the characteristic signals in ¹H NMR spectra of crude reaction mixtures were obstructed with peaks from impurities. Our yields were moderate, ranging from 17-53%. Although consistent diastereomeric ratios were observed, at this point we were not able to prove that the major diastereomer we were obtaining had the same relative configuration as the natural product synoxazolidinone A (2-26).

We propose that in the transition state 5-56 Cl is more sterically demanding than the alkyl chain and the hydrogen should be placed in pseudo axial position so that minimal 1,3-diaxial interactions with bulky N-benzyl occur (Scheme 5.14). The enol is expected to attack from the side opposite to the larger, Cl- substituent to give major diastereomer 5-45c.

**Scheme 5.14:** Proposed transition state of N-acyliminium ion intermediate leading to the formation of the major diastereomer of 4-oxazolidinone 5-54c.
Removal of the dimethoxybenzyl group was achieved with no significant erosion of the \( dr \), but minor decomposition was observed. Employing unoptimized acidic conditions we successfully converted a single diastereomer of 5-45 into 4-oxazolidinone 4-27 in 49% yield (Scheme 5.15).

\[
\text{Scheme 5.15: Deprotection of 2,4-DMB oxazolidin-4-one 5-45c.}
\]

Very minor amounts of another diastereoisomer of 4-27 were detected (4% yield) which was either due to its presence in the starting material or possible epimerization during the DMB-removal step.

### 5.3.2 The First Total Synthesis of Synoxazolidinones A and B

Being the simplest member of the family, synoxazolidinone B (2-27) was initially targeted for synthesis (Scheme 5.16). The treatment of 4-aminobutanol (5-51) with \( N,N' \)-bis(tert-butoxycarbonyl)-S-methylisothiourea (4-29) provided a Boc-protected guanidine alcohol 4-28 that was oxidized under Swern conditions to yield aldehyde 4-31 in 88% yield over the two steps.
Conversion of the aldehyde to the requisite benzylimine was accomplished by treatment with 2,4-dimethoxybenzyl amine (5-48c) and MgSO₄, and the crude imine was directly subjected to the aforementioned acylation/cyclization cascade with acid chloride 5-35 to provide the Boc-functionalized synoxazolidinone B scaffold 5-52 in 30% yield over two steps. Subsequent deprotection provided compound 2-27 in 77% yield after preparative reverse-phase chromatography and the ¹H and ¹³C NMR spectroscopy data are in agreement with the isolation report. The first total synthesis of synoxazolidinone B was accomplished in 20% yield over four steps.²⁷

In parallel to the efforts to construct synoxazolidinone B, we explored asymmetric installation of the secondary chloride substituent present in synoxazolidinone A (2-26). To determine the potential of employing an imine acylation/cyclization cascade analogous to that employed for the deschloro natural product 2-27, we required access to enantiopure α-chloro aldehyde 4-30 (Scheme 5.17).³¹ Although the chlorination of aldehydes is a well established reaction, with several elegant catalytic asymmetric approaches,³² it became
apparent that a protected guanidine moiety in close proximity to the reacting aldehyde was significantly complicating this reaction. All attempts at catalytic asymmetric chlorination of 4-31 proved unsuccessful, with no reactivity observed. To overcome these difficulties, we employed stoichiometric proline to promote efficient chlorination of aldehyde 4-31, thereby producing compound 4-30 in 60% yield.

Scheme 5.17: The first total synthesis of synoxazolidinone A.

The most successful asymmetric chlorination we have achieved provides aldehyde 4-30 in 56% ee (2 equiv. proline amide, 2 equiv. NCS, dioxane, 0 – 8 °C); however, the crude product can not be successfully carried forward and all attempts at 4-30 aldehyde purification furnished racemate in low yield. New catalyst development, a multistep work-around, or a substrate lacking the guanidine functionality would likely be required to overcome these obstacles.
With aldehyde 4-30 in hand, we carried out the imine formation/acylation/cyclization cascade to provide compound 5-55 in 40% yield and 4:1 dr favoring the desired diastereomer (Scheme 5.17). Significant efforts were made to improve this diastereoselectivity; however, no conditions provided a higher quantity of the desired product upon isolation. As we earlier demonstrated on the simple substrates (Table 5.3), the presence of the dimethoxybenzyl group was essential to obtaining diastereoselectivity, with many substrates and conditions yielding a 1:1 mixture of diastereomers. The diastereomeric ratio ranged from 5:1 to 2.5:1 depending on the batch of acid chloride and conditions employed; 4:1 is the most commonly observed diastereoselectivity in the case of the natural products. The $^1$H NMR spectra for the two diastereomers are significantly different (chemical shifts and coupling constants) for both the protected 5-55 and deprotected 2-26 synoxazolidinones, thus allowing straightforward assignment of the isolated diastereomers.

With the natural product skeleton complete, removal of both the dimethoxybenzyl and Boc protecting groups was achieved with TFA (40 °C, 48 h) to yield synoxazolidinone A (2-26) as a 4:1 mixture of diastereomers in 88% yield. No erosion of the diastereomeric ratio during the deprotection reactions was detected, even upon prolonged heating. The diastereomers of the fully deprotected synoxazolidinone A natural product proved difficult to separate without the use of HPLC, but fortunately, the partially deprotected product 5-55 (TFA/DCM, rt, 2 h) was readily separable by reverse phase chromatography. As with synoxazolidinone B, the NMR data for 2-26 are in full agreement with the reported data. The first total synthesis of synoxazolidinone A was accomplished in 19% yield over five steps.
In addition to two natural products, we prepared a small panel of compounds (5-57 – 5-64) (Scheme 5.18).

![Scheme 5.18: Small library of 4-oxazolidinones containing α-Cl and terminal guanidine substitution.](Image)

We explored the impact of deleting the guanidine moiety, making the aryl ring electron-deficient (5-60, 5-61, 5-63, 5-64), or installing an additional chlorine substituent (5-58, 5-59, 5-60) in order to define an initial structure-activity relationship (SAR) for this class of compounds.

All of the analogues were produced without modification to our method, thereby highlighting the straightforward nature of this approach for generating an array of 4-
oxazolidinone products. It should be noted that the dichloro- compounds (5-58, 5-59, 5-60) arise from chlorination of the crude imine\textsuperscript{33} (excess NCS) since purified mono- and dichloro aldehydes do not undergo our dehydration/acylation/cyclization cascade reaction (Chapter 4).

5.3.3 Studies Towards Total Synthesis of Synoxazolidinone C

Synoxazolidinone C (2-28) is a bicyclic member of the synoxazolidinone family of natural products.\textsuperscript{34} 2-28 has a broader spectrum of biologically activity compared to synoxazolidinones A and B. In addition to promising antimicrobial activity synoxazolidinone C is cytotoxic towards human cancer cells (melanoma, breast adenocarcinoma and colon carcinoma). The anti-cancer and antimicrobial activity of synoxazolidinone C, coupled with its unique bicyclic structure presents new opportunities for biological discoveries and challenges for 4-oxazolidinone ring synthesis.

Inspired by our previous work on the total synthesis of synoxazolidinones A and B via imine acylation/cyclization cascade we propose a chlorolactonization as key transformation our approach to the synthesis of synoxazolidinone C (2-28). It is expected that upon reaction with electrophilic chlorine, enamide 5-74 will form a chloronium ion which will be attacked by the enol nucleophile in a stereoselective fashion to form bicyclic intermediate 5-72 (Scheme 5.19).

The thermal decomposition of carboxylic azides to form an isocyanate is referred to as the Curtius rearrangement. Isocyanate intermediates can be isolated or converted to addition products of interest. We further propose Curtius rearrangement of the azide derived
from 5-72 to a primary amine precursor 5-71, which will be guanidinated to provide synoxazolidinone C 5-70.

Desired enamide 5-74 will arise via alkene isomerization of 5-75 formed during ring closing metathesis reaction of diallylamide derivative 5-76. RCM precursor 5-76 will be obtained by coupling of diallyl amine derivative 5-77 with previously obtained acid 4-13g (Scheme 5.19).

Scheme 5.19. Retrosynthetic analysis of synoxazolidinone C.
During the pursuit toward acyclic synoxazolidinones A and B we took a few steps forward in the exploration of the proposed route to synoxazolidinone C. At the beginning of our studies towards synoxazolidinone C, we employed a simplified model and used phenylpyruvic acid 4-10 and diallyl amine 5-78 as starting materials. Diallylamide 5-79 was readily prepared by acylation of 5-78 with the acyl chloride of 4-10 in 64% yield. 5-79 then smoothly underwent ring closing metathesis employing Grubbs 1st generation catalyst to provide 5-81 in 88% yield (Scheme 5.20).

Scheme 5.20: Synthesis of cyclic amides 5-81 and 5-82 as isomerization precursors.

The synthesis of a different dibromomethoxy-containing substrate 5-82 was accomplished through the same sequence starting from the corresponding acid 4-13g. With starting materials 5-81 and 5-82 in hand we began screening for a suitable isomerization catalyst.

Numerous examples of transition metal catalyzed reactions leading to double bond migration have been reported in the literature.\textsuperscript{35-39} Generally, acyclic alkenes isomerize more readily than cyclic ones; strain in small and medium sized rings is an important factor to consider when predicting isomerization outcome. It has been reported that 2-pyrrolines, obtained from \(N,N\)-bisallylamides by ring-closing metathesis reaction can undergo subsequent isomerization promoted by ruthenium hydride complexes.\textsuperscript{40} In our hands substrate 5-81 failed to isomerize under various conditions that generate Ru-hydride species.
(Entries 1-3, Table 5.4), and reduction of the pyrroline to pyrrolidine ring was observed when hydrogen gas was employed as a source of hydride (Entry 3, Table 5.4).

**Table 5.4:** Attempts at isomerization of cyclic amides 5-81 and 5-82 to the corresponding enamides 5-83 and 5-84.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Enamide (%)</th>
<th>SM (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5-81, Grubbs II (7.5 mol%) NaH, PhMe, Δ</td>
<td>0</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>5-81, Grubbs I (5 mol %) NaOH/iPrOH, PhMe, Δ</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>5-81, Grubbs I (5 mol%) H₂, DCM, RT</td>
<td>10*</td>
<td>61</td>
</tr>
<tr>
<td>4</td>
<td>5-81, [Fe₂(CO)₉], THF, Δ</td>
<td>0</td>
<td>90</td>
</tr>
<tr>
<td>5</td>
<td>5-81, [Fe(CO)₅], neat, Δ</td>
<td>0</td>
<td>84</td>
</tr>
<tr>
<td>6</td>
<td>5-81, [Rh(PPh₃)₃Cl] (30 mol%) PhMe</td>
<td>0</td>
<td>55</td>
</tr>
<tr>
<td>7</td>
<td>5-81, DPPE [Pd(O(CO)CF₃)₂] DIPEA, TFA, PhMe, Δ</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>8</td>
<td>5-81, [Ir(COD)Cl]₂ (5 mol%) C₅H₅NO (5 mol%), P(CyHex)₃ (10 mol%) PhMe, Δ</td>
<td>40</td>
<td>25</td>
</tr>
<tr>
<td>9</td>
<td>5-81, [Ir(COD)Cl]₂ (10 mol%) C₅H₅NO (10 mol%), P(CyHex)₃ (20 mol%) PhMe, Δ</td>
<td>9</td>
<td>5</td>
</tr>
<tr>
<td>10</td>
<td>5-82, [Ir(COD)Cl]₂ (5 mol%) C₅H₅NO (5 mol%), P(CyHex)₃ (10 mol%) PhMe, Δ</td>
<td>30</td>
<td>15</td>
</tr>
</tbody>
</table>

* - yield for the compound with reduced double bond.
Stille and Becker previously reported that iron carbonyls can promote isomerization of simple N-acyl pyrrolines and acyclic N-allylamides to their corresponding enamides.\textsuperscript{41} Unfortunately, these conditions did not yield isomerization product 5-83 and starting material 5-81 was almost completely recovered (Entries 4 and 5, Table 5.4). Finally, conditions employing Iridium complex\textsuperscript{35} provided 5-83 in 40% yield with 25% of starting material 5-81 recovered (Entry 8, Table 5.4). Increasing of the catalytic loading of iridium-COD catalyst and phosphine ligands led to lower yields associated with decomposition (Entry 9, Table 5.4). Conditions employing iridium-COD dimer with P(CyHex)\textsubscript{3} as ligand in presence of Cs\textsubscript{2}CO\textsubscript{3} in refluxing toluene also provided dibromomethoxy product 5-84 which was isolated in 30% yield with 15% of starting material 5-82 recovered (Entry 10, Table 5.4).

Halolactonization is a cyclization process in which an oxygen nucleophile adds to a halonium ion to form lactone ring. Mechanistically, the reaction involves a positively charged halonium ion in a substrate which also contains a carboxylic acid or alcohol functional group. The hydroxyl group, acting as nucleophile, attacks the halonium ion and intramolecular ring closure proceeds to produce lactone. The reaction is usually performed under mildly basic conditions in order to facilitate nucleophilic attack. The regioselectivity of halolactonization can be predicted according to Baldwin’s rules according to which 5-exo-tet ring closures are allowed while 6-endo-tet ring closures are disallowed. Successful synthesis of 5-83 and 5-84 allowed us to proceed and probe our key halolactonization transformation to construct the bicyclic 4-oxazolidinone and install secondary chlorine moiety in one step. When 5-83 was treated with NBS in presence of sodium bicarbonate we observed formation of two Br-containing products in approximately 1:1 ratio (Scheme 5.21).
Compound **5-86** is the desired product of anti addition across the double bond formed via **5-exo-tet** ring closure. We believe that another constituent of this racemic mixture is product **5-87** formed via **6-endo-tet** cyclization of the enol **5-85**, which according to Baldwin’s classification of ring closures for enolates\(^\text{42}\) is also allowed. Further investigation of product structures is required to confirm relative stereochemistry of racemates **5-86** and **5-87**.

**5.4 Conclusions and Outlook**

In conclusion, we have developed a rapid and diastereoselective synthesis for the synoxazolidinone family of natural products. Through these efforts, we have gained insight into the chemistry of these unusual heterocycles and provided new lead compounds for antimicrobial development. Efforts to increase the potency and selectivity of this class of compounds are underway, as well as studies aimed at identifying their intracellular targets.
5.5 Experimental

General Information

THF was purified using an alumina filtration system. Aldehydes were purchased from a commercial chemical company and used as received. Reactions were monitored by TLC analysis (pre-coated silica gel 60 F_{254} plates, 250 mm layer thickness) and visualization was accomplished with a 254 nm UV light and by staining with a KMnO_{4} solution (1.5 g of KMnO_{4}, 10 g of K_{2}CO_{3}, and 1.25 mL of a 10% NaOH solution in 200 mL of water). Reactions were also monitored by LC-MS (2.6 mm C18 50 x 2.10 mm column). Flash chromatography on SiO_{2} was used to purify the crude reaction mixtures and performed on a flash system utilizing pre-packed cartridges and linear gradients.

Melting points were determined using a capillary melting point apparatus. Infrared spectra were determined on a FT/IR spectrometer. ^1H, ^13C and ^19F NMR spectra were obtained on a 400 MHz instrument in CDCl_{3} unless otherwise noted. Chemical shifts were reported in parts per million with the residual solvent peak used as an internal standard (CDCl_{3} = 7.26 ppm for ^1H and 77.16 ppm for ^13C). ^1H NMR spectra were run at 300 or 400 MHz and are tabulated as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, p = pentet, m = multiplet, bs = broad singlet, dt = doublet of triplet, tt = triplet of triplet), number of protons, and coupling constant(s). ^13C NMR spectra were run at 100 MHz using a proton-decoupled pulse sequence with a d_{1} of 0 second unless otherwise noted, and are tabulated by observed peak. High-resolution mass spectra were obtained on an ion trap mass spectrometer using heated electrospray ionization (HESI).
To a solution of cyanuric chloride (21 mg, 0.11 mmol) in dichloromethane (1.5 mL) at 0 °C was added phenylpyruvic acid (55 mg, 0.33 mmol) and the reaction was stirred for 2 h. NaCl (25 mg, 0.43 mmol) was then added and reaction mixture stirred for 1 h. A solution of crude imine (0.33 mL, 0.33 mmol, see compd 5-42 for prep) in dichloromethane was added at 0 °C and the reaction was allowed to warm to rt over 20 h. The reaction mixture was filtered through Celite®, the filter cake washed with dichloromethane. The filtrate was concentrated in vacuo and the crude residue purified by flash chromatography on SiO₂ (EtOAc/hexanes, 5 to 40%) to provide 16.8 mg (39%) of 5-43 as a light yellow oil: Rf = 0.48 (40% EtOAc/hexanes); ¹H NMR: (300 MHz, CDCl₃) δ 7.62 – 7.56 (m, 2 H), 7.40 – 7.32 (m, 2 H), 7.28 – 7.16 (m, 2 H), 6.47 – 6.41 (m, 2 H), 5.04 (d, J = 15.0 Hz, 1 H), 4.38 (t, J = 3.5 Hz, 1 H), 4.29 (d, J = 15.0 Hz, 1 H), 3.84 (s, 3 H), 3.80 (s, 3 H), 2.02 – 1.89 (m, 1 H), 1.85 – 1.65 (m, 1 H), 1.19 – 0.99 (m, 4 H), 0.96 – 0.74 (m, 2 H), 0.75 (t, J = 7.0 Hz, 3 H); ¹³C NMR (75 MHz, CDCl₃) δ 167.6, 160.7, 158.4, 142.3, 131.9, 131.0, 129.6, 128.6, 127.4, 127.4, 121.5, 117.5, 104.4, 98.6, 56.9, 55.6, 55.5, 38.4, 31.7, 28.6, 22.6, 20.8, 14.1; IR (film): 3160, 2932, 1665, 1455, 1209, 1035, 764 cm⁻¹; HRMS (ESI) m/z calcd for C₂₄H₃₀NO₄, [M+H]⁺ 396.2169, found 396.2173.
(Z)-5-Benzylidene-3-(2,4-dimethoxybenzyl)-2-pentyloxazolidin-4-one (5-42).

To a solution of hexanal (0.12 mL, 0.98 mmol) and magnesium sulfate (540 mg, 4.53 mmol) in dichloromethane (1 mL) was slowly added 2,4-dimethoxybenzylamine (160 µL, 1.06 mmol) and the slurry was stirred at room temperature under an inert atmosphere for 40 minutes. The crude reaction mixture was filtered, washed with tetrahydrofuran (1 mL) and the solution of crude imine was used in the next step. The solution of imine was cooled to 0 °C and of N,N-diisopropylethylamine (90 µL, 0.53 mmol) was added followed by addition of PyBOP (0.28 g, 0.53 mmol) and phenyl pyruvic acid (80 mg, 0.49 mmol). The resultant mixture was allowed to warm to room temperature and stirred for 18 h. The reaction mixture was filtered through Celite®, filter cake was washed with dichloromethane, solvent concentrated in vacuo and the crude residue purified by flash chromatography on SiO₂ (EtOAc/hexanes 5 to 40%) to provide 46.2 mg (24%) of 5-42 as a light yellow oil: Rf = 0.58 (40% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 7.65 (d, J = 7.8 Hz, 2 H), 7.34 (t, J = 7.7 Hz, 2 H), 7.29 – 7.19 (m, 2 H), 6.47 – 6.42 (m, 2 H), 6.25 (s, 1 H), 5.35 (dd, J = 6.3, 2.1 Hz, 1 H), 4.94 (d, J = 14.7 Hz, 1 H), 4.24 (d, J = 14.8 Hz, 1 H), 3.82 (s, 3 H), 3.80 (s, 3 H), 2.06 – 1.96 (m, 1 H), 1.75 –1.63 (m, 1 H), 1.48 – 1.15 (m, 6 H), 0.89 (t, J = 7.4 Hz, 3 H); ¹³C NMR (101 MHz, CDCl₃) δ 162.4, 161.2, 158.5, 144.7, 134.4, 131.8, 129.2, 128.7, 127.3,
116.1, 104.7, 102.7, 98.7, 91.1, 55.6, 38.6, 33.4, 31.7, 22.7, 22.0, 14.2; IR (film): 2928, 2856, 1709, 1613, 1508, 1416, 1209, 1034, 694 cm\(^{-1}\).

\[\text{N}^2, \text{N}\text{3}-\text{Bis(tert-butoxycarbonyl)}-\text{N}^1-(4\text{-hydroxybutyl})\text{guanidine}^{23} (4\text{-}31).\]

\(\text{N},\text{N}'\text{-bis(tert-butoxycarbonyl)}\text{-S-methylisothiourea} (4.01 \text{ g}, 13.6 \text{ mmol}) \) was dissolved in tetrahydrofuran (40 mL) and a solution of 4-amino-1-butanol (2.44 g, 2.51 mL, 27.2 mmol) in tetrahydrofuran (80 mL) was added drop wise over 10 minutes. The reaction was heated at 50 °C for 2 h and then cooled to room temperature. Water (120 mL) and dichloromethane (200 mL) were added, the organic layer separated and the aqueous layer was extracted with DCM (2 \(\times\) 120 mL), the combined organic layers were washed with water (360 mL) and brine (360 mL) then dried (MgSO\(_4\)), filtered, and concentrated \textit{in vacuo} to give 4.33 g (100%) of \(\text{N}^2, \text{N}\text{3}-\text{bis(tert-butoxycarbonyl)}\text{-N}^1-(4\text{-hydroxybutyl})\text{guanidine}^{43}\) as a white solid that was used without further purification.

Oxalyl chloride (0.67 mL, 7.8 mmol) was added to a solution of dry dimethylsulfoxide (2.45 mL, 15.6 mmol) in dichloromethane (150 mL) at -78 °C and stirred for 40 minutes under an inert atmosphere. A solution of \(\text{N}^2, \text{N}\text{3}-\text{bis(tert-butoxycarbonyl)}\text{-N}^1-(4\text{-hydroxybutyl})\text{guanidine} (6.48 \text{ mmol}, 2.15 \text{ g})\) in dichloromethane (150 mL) was slowly added and reaction stirred for 40 minutes. Triethylamine (4.52 mL, 32.4 mmol) was slowly added and the reaction stirred for 40 minutes then warmed to room temperature, quenched
with water (150 mL), extracted with DCM (3 x 250 mL), combined organic extracts dried (MgSO₄), filtered and concentrated in vacuo. The crude residue was purified by flash chromatography on SiO₂ (EtOAc/hexanes, 40 to 20% EtOAc) to provide 1.87 g (88%) of 4-31 as a colorless oil: ¹H NMR (400 MHz, CDCl₃) δ 11.44 (s, 1 H), 9.74 (t, J = 1.2 Hz, 1 H), 8.31 (s, 1 H), 3.65 – 3.16 (m, 2 H), 2.48 (t, J = 7.4 Hz, 2 H), 1.86 (p, J = 7.3 Hz, 2 H), 1.44 (s, 18 H).

\[
\begin{align*}
\text{N}^2, \text{N}^3-\text{Bis(tert-butoxycarbonyl)}-\text{N}^1-(4\text{-oxy}-3\text{-chloro}-\text{butyl})\text{guanidine}^{36} (4\text{-30}).
\end{align*}
\]

To a solution of 4-31 (0.33 g, 1.0 mmol) in DCM (2 mL) at 0 °C was added L-proline (0.13 g, 1.1 mmol) under an inert atmosphere and the reaction was stirred for 30 minutes. NCS (0.18 g, 1.3 mmol) was added and the reaction was allowed to warm up to room temperature over 2 h at which time hexanes (1 mL) was added and a white precipitate formed. The slurry was dry loaded and purified by flash chromatography on SiO₂ (Et₂O/hexanes, 20 to 100%) to yield 218 mg (60%) of racemic semi-pure 4-30 as a white foam (keep air free and in solution): ¹H NMR (400 MHz, CDCl₃) δ 11.46 (br s, 1 H), 9.54 (d, J = 1.5 Hz, 1 H), 8.46 (br s, 1 H), 4.31 (ddd, J = 9.0, 5.0, 1.7 Hz, 1 H), 3.73 – 3.52 (m, 2 H), 2.47 – 2.33 (m, 1 H), 2.09 – 1.95 (m, 1 H), 1.49 (s, 18 H); ¹³C NMR (75 MHz, CDCl₃) δ 194.2, 163.5, 156.4, 153.3, 83.5, 79.5, 61.3, 37.3, 31.5, 28.3, 28.1.

130
3-(3,5-Dibromo-4-methoxyphenyl)-2-oxopropanoic acid (4-13g)\textsuperscript{44,45}

A slurry of 2,5-dibromo-4-methoxy-benzaldehyde (4.12 g, 13.9 mmol), sodium acetate (1.15 g, 13.9 mmol) and N-acetylglycine (1.64 g, 13.9 mmol) in acetic anhydride (30 mL) was heated at 100 °C for 4 h. The reaction mixture was allowed to cool to room temperature over 2 h and the resulting yellow precipitate removed by filtration. The filtrate was diluted with DCM (100 mL), washed with distilled water (100 mL), dried (MgSO\textsubscript{4}) and concentrated in vacuo to afford azlactone, which was used without further purification in the next step. Azlactone was suspended in HCl (100 mL of 3 M aqueous solution), and heated at reflux for 19 h. The resulting precipitate was filtered and dried in a vacuum desiccator over potassium hydroxide for 20 h to provide 2.75 g (56%) of acid as an off-white solid: \textsuperscript{1}H NMR (400 MHz, DMSO-\textit{d}\textsubscript{6}) δ 9.79 (br s, 1 H), 8.06 (s, 2 H), 6.35 (s, 1 H), 3.79 (s, 3 H).

3-(3,5-Dibromo-4-methoxyphenyl)-2-oxopropanoyl chloride (5-35).

To a solution of 3-(3,5-dibromo-4-methoxyphenyl)-2-oxopropanoic acid (88 mg, 0.25 mmol) in THF (0.5 mL) at 0 °C was added DMF (3 drops) followed by slow addition of oxalyl chloride (38 µL, 0.45 mmol). The reaction was allowed to warm to room temperature
over 90 minutes and concentrated in vacuo to provide crude 5-35 that was used without further purification.

\[
\text{\begin{figure}[h]
\centering
\includegraphics[width=0.5\textwidth]{structure}
\caption{Structure of 2-Oxo-3-(4-(trifluoromethyl)phenyl)propanoic acid (4-13e).}
\end{figure}}
\]

2-Oxo-3-(4-(trifluoromethyl)phenyl)propanoic acid (4-13e).\(^{46}\)

A slurry of 4-(trifluoromethyl) benzaldehyde (2.02 g, 11.6 mmol), sodium acetate (1.03 g, 12.6 mmol) and N-acetylglycine (1.48 g, 12.6 mmol) in acetic anhydride (20 mL) was heated at 110 °C for 3 h. The reaction mixture was allowed to cool to room temperature over 2 h, the resulting yellow precipitate removed by filtration, the filtrate washed with cold distilled water (60 mL), dried (MgSO\(_4\)) and concentrated in vacuo to afford azlactone, which was used without further purification. Azlactone was suspended in HCl (40 mL of 3 M aqueous solution) and heated at 85 °C for 17 h and cooled in an ice bath to facilitate crystallization. The resulting crystals were isolated by filtration and dried in a vacuum desiccator over potassium hydroxide to provide 1.38 g (52\%) of acid as a yellow solid: \(^1\)H NMR (400 MHz, DMSO-\(d_6\)) \(\delta\) 13.49 (br s, 1 H), 9.80 (br s, 1 H), 7.95 (d, \(J = 8.2\) Hz, 2 H), 7.68 (d, \(J = 8.3\) Hz, 2 H), 6.46 (s, 1 H). \(^{13}\)C NMR (101 MHz, DMSO-\(d_6\)) \(\delta\) 166.0, 144.1, 139.3, 129.6, CF\(_3\) (127.4, 127.1, 126.8, 126.4), 125.8, 125.2, 125.1, 123.1, 107.6.
2-Oxo-3-(4-(trifluoromethyl)phenyl)propanoyl chloride (5-87).

To a solution of 2-oxo-3-(4-(trifluoromethyl)phenyl)propanoic acid (58 mg, 0.25 mmol) in THF (0.5 mL) at 0 °C was added DMF (3 drops) followed by slow addition of oxalyl chloride (38 µL, 0.45 mmol). The reaction mixture was allowed to warm to room temperature over 90 minutes and concentrated in vacuo to provide crude acid chloride that was used without further purification.

\[ \text{N}^2, \text{N}^3 - \text{Bis(tert-butoxycarbonyl)-N}^1-(3-(5-((Z)-3,5-dibromo-4-methoxybenzylidene)-3-(2,4-dimethoxybenzyl)-4-oxooxazolidin-2-yl)propyl)guanidine (5-52).} \]

To a solution of 4-31 (79 mg, 0.22 mmol) and magnesium sulfate (0.11 g, 0.93 mmol) in dichloromethane (1 mL) was added 2,4-dimethoxybenzylamine (36 µL, 0.23 mmol) at room temperature and the reaction mixture was stirred for 30 minutes until starting material disappeared. The reaction was cooled to 0 °C, crude 5-35 (0.25 mmol) was added in
dichloromethane (0.5 mL) via syringe and the reaction mixture was allowed to warm to room temperature and stirred for 18 h. The reaction mixture was concentrated \textit{in vacuo} and the oily residue purified by flash column chromatography on SiO$_2$ (Et$_2$O/hexanes, 10 to 100% Et$_2$O) to provide 52 mg (30%) of 5-52 as a colorless oil: $^1$H NMR (400 MHz, CDCl$_3$) \( \delta \) 11.50 (br s, 1 H), 8.42 (br s, 1 H), 7.74 (s, 2 H), 7.27 (d, \( J = 7.3 \) Hz, 1 H), 6.47-6.43 (m, 2 H), 6.06 (s, 1 H), 5.41 (dd, \( J = 4.7, 1.7 \) Hz, 1 H), 4.93 (d, \( J = 14.6 \) Hz, 1 H), 4.22 (d, \( J = 14.7 \) Hz, 1 H), 3.87 (s, 3 H), 3.82 (s, 3 H), 3.80 (s, 3 H), 3.48 (br s, 2 H) 2.14 – 2.00 (m, 1 H), 1.84 – 1.63 (m, 1 H), 1.65 – 1.56 (m, 2 H), 1.49 (s, 9 H), 1.48 (s, 9 H); $^{13}$C NMR (101 MHz, CDCl$_3$) \( \delta \) 161.5, 161.3, 158.5, 156.1, 153.3, 152.9, 145.4, 133.0, 132.8, 131.9, 118.2, 115.6, 104.7, 99.7, 98.6, 90.7, 60.8, 55.6, 38.8, 30.4, 28.4, 28.2, 21.9; HRMS (ESI) \( m/z \) calcd for C$_{34}$H$_{43}$Br$_2$N$_4$O$_9$, [M+H]$^+$ 811.1569, found 811.1548.

\[
(Z)-1-(3-(5-\text{ (3,5-Dibromo-4-methoxybenzylidene)-4-oxooxazolidin-2-yl}) \text{ propyl}) \text{ guanidine (2-27).}
\]

A solution of 5-52 (24.9 mg, 30.6 \( \mu \)mol) in trifluoroacetic acid (2.5 mL) was sealed in a vial and heated at 40 °C for 48 h. When the deprotection was complete (the mixture acquired deep purple color) the solution was concentrated \textit{in vacuo} and the residue was purified by reverse phase column chromatography on C18 SiO$_2$ (H$_2$O/CH$_3$CN, 10 to 70%
CH₃CN, 0.1% TFA additive) to provide 10.7 mg (77%) of **2-27** as a colorless oil: ¹H NMR (300 MHz, MeOD) δ 7.87 (s, 2 H), 6.03 (s, 1 H), 5.77 (t, J = 4.3 Hz, 1 H), 3.85 (s, 3 H), 3.29 – 3.24 (m, 2 H), 2.08 – 1.91 (m, 1 H), 1.92 – 1.78 (m, 1 H), 1.77 – 1.61 (m, 2 H); ¹³C NMR (126 MHz, MeOD) δ 165.3, 158.6, 154.3, 146.9, 134.4, 133.9, 119.0, 100.1, 90.1, 61.2, 42.0, 34.3, 23.4; HRMS (ESI) m/z calcd for C₁₅H₁₉Br₂N₄O₃, [M+H]⁺ 460.9818, found 460.9820.

---

**N², N³-Bis (tert-butoxycarbonyl) –N¹-(3-chloro-5- ((Z)-3,5-dibromo-4-methoxy benzylidene)-3-(2,4-dimethoxybenzyl)-4-oxooxazolidin-2-yl)propyl)guanidine (5-54).**

To a solution of aldehyde **4-30** (95.9 mg, 0.26 mmol) and magnesium sulfate (142 mg, 1.18 mmol) in chloroform (1 mL) was slowly added 2,4-dimethoxybenzylamine (32 µL, 0.21 mmol) and the reaction mixture was stirred for 20 minutes until starting material disappeared. The reaction mixture was then cooled to 0 °C, crude **5-35** (0.25 mmol) in chloroform (0.5 mL) was added via syringe, the reaction mixture was allowed to warm up to room temperature and stirred for 16 h concentrated *in vacuo* and the oily residue purified by flash column chromatography on SiO₂ (Et₂O/hexanes, 20 to 100% Et₂O) to provide 79 mg (40%) of **5-54** as a light yellow oil (4.4:1 diastereomeric mixture): Characteristic signals of
major diastereomer: $^1$H (400 MHz, CDCl$_3$) 7.72 (s, 2 H), 6.08 (s, 1 H), 5.45 (s, 1 H), 5.00 (d, $J = 14.9$ Hz, 1 H), 4.45 (m, $J = 4.46$, 2.8 Hz, 1 H), 4.17 (d, $J = 14.7$ Hz, 1 H); HRMS (ESI) $m/z$ calcd for C$_{34}$H$_{42}$Br$_2$ClN$_4$O$_9$, [M-H] 843.1012, found 843.1029;

![Chemical Structure](image)

1-(3-Chloro-3-(5-((Z)-3,5-dibromo-4-methoxybenzylidene)-3-(2,4-dimethoxybenzyl)-4-oxooxazolidin-2-yl)propyl)guanidine (5-55).

A solution of 5-54 (79 mg, 93 µmol) in dichloromethane (1.85 mL) and trifluoroacetic acid (1.85 mL) was stirred at room temperature until disappearance of starting material (~2 h), concentrated in vacuo and the crude residue purified by reverse phase column chromatography on C18 SiO$_2$ (H$_2$O/CH$_3$CN, 10 to 50% CH$_3$CN, 0.1% TFA additive) to provide 44.7 mg (93%) of 5-55 (single diastereomer, calculated from major diastereomer) as a colorless oil: $^1$H NMR (400 MHz, MeOD) δ 7.84 (s, 2 H), 7.26 (d, $J = 8.4$ Hz, 1 H), 6.58 (d, $J = 2.4$ Hz, 1 H), 6.53 (dd, $J = 8.3$, 2.4 Hz, 1 H), 6.10 (s, 1 H), 5.67 (s, 1 H), 4.95-4.87 (d, 1 H, obscured by H$_2$O), 4.62 – 4.57 (m, 1 H), 4.29 (d, $J = 14.7$ Hz, 1 H), 3.85 (s, 3 H), 3.84 (s, 3 H), 3.79 (s, 3 H), 3.63 – 3.41 (m, 2 H), 2.40 – 2.25 (m, 1 H), 2.22 – 2.11 (m, 1 H); $^{13}$C NMR (101 MHz, MeOD) δ 163.1, 160.0, 158.8, 154.5, 146.7, 134.0, 134.0, 132.6, 119.0,
118.7, 115.8, 106.1, 100.9, 99.5, 95.9, 92.4, 61.1, 59.9, 55.8, 40.1, 39.7, 33.5; HRMS (ESI) 
m/z calcd for C_{24}H_{28}Br_{2}ClN_{4}O_{5}, [M+H]^+ 645.0110, found 645.0108.

![Chemical structure](image)

1-((S)-3-Chloro-3-((S)-5-((Z)-3,5-dibromo-4-methoxybenzylidene)-4-oxooxazolidin-2-yl)propyl)guanidine (2-26).

A solution of 5-55 (16.2 mg, 25.1 µmol) in trifluoroacetic acid (2 mL) was heated at 40 °C for 48 hours, concentrated *in vacuo* and the crude residue purified by reverse phase column chromatography on SiO₂ (H₂O/CH₃CN, 10 to 60% CH₃CN, 0.1% TFA additive) to provide 10.9 mg (88%) of 2-26 as a colorless oil: 

**¹H NMR (400 MHz, MeOD)** δ 7.89 (s, 2 H), 6.08 (s, 1 H), 5.91 (d, J = 2.4 Hz, 1 H), 4.30 (dt, J = 11.0, 2.6 Hz, 1 H), 3.86 (s, 3 H), 3.56 – 3.35 (m, 2 H), 2.32 – 2.22 (m, 1 H), 2.10 – 1.99 (m, 1 H); 

**¹³C NMR (101 MHz, MeOD)** δ 165.6, 158.8, 154.5, 146.5, 134.1, 119.1, 100.6, 90.3, 61.7, 61.2, 39.5, 32.3. HRMS (ESI) m/z calcd for C_{12}H_{16}Br_{2}ClN_{4}O_{3} [M-H] 492.9283, found 492.9290.

**Procedure for the preparation of chlorinated analogues. General procedure A:**

To a solution of hexanal (0.51 mmol, 0.065 mL) in dichloromethane (1.0 mL) at 0 °C was added L-proline (0.05 mmol, 10 mol%, 7 mg) followed by the addition of N-chlorosuccinimide (90 mg, 0.67 mmol, 1.3 equiv.). The reaction mixture was allowed to
warm to room temperature over 4 hours then MgSO₄ (0.32 g, 2.6 mmol) and 4-dimethoxybenzylamine (0.079 mL, 0.51 mmol) were added and the resultant mixture was stirred at room temperature for 30 min. The reaction was cooled to 0 °C and crude acid chloride was added (1 equiv., freshly prepared according to general procedure) in dichloromethane (1 mL) over 5 min, warmed to room temperature and stirred for 20 h. The reaction mixture was filtered through Celite®, filter cake was washed with dichloromethane, concentrated in vacuo and the crude orange residue purified by flash chromatography on SiO₂ (EtOAc/hexanes, 3-30% EtOAc) to provide both mono- and di-chlorinated products.

(Z)-2-(1-Chloropentyl)-5-(3,5-dibromo-4-methoxybenzylidene)oxazolidin-4-one (5-57).

17.5 mg (5.6%) obtained (according to general procedure A) in as a colorless oil: single diastereomer, rf = 0.36 (20% EtOAc/hexanes): \(^{1}\)H NMR (400 MHz, CDCl₃) \(\delta\) 7.80 (s, 2 H), 7.29 – 7.24 (m, 1 H), 6.49 – 6.44 (m, 2 H), 6.10 (s, 1 H), 5.56 (d, \(J = 2.1\) Hz, 1 H), 4.88 (d, \(J = 14.7\) Hz, 1 H), 4.33 (d, \(J = 14.7\) Hz, 1 H), 4.18 – 4.13 (m, \(J = 1\) H), 3.88 (s, 3 H), 3.85 (s, 3 H), 3.80 (s, 3 H), 1.59 – 1.49 (m, 2 H), 1.34 – 1.18 (m, 4 H), 0.86 (t, \(J = 7.1\) Hz, 3 H); \(^{13}\)C NMR (75 MHz, CDCl₃) \(\delta\) 162.0, 161.5, 158.4, 153.2, 144.6, 133.1, 132.6, 132.1, 118.3, 115.2, 104.9, 100.7, 98.7, 92.4, 61.1, 60.9, 55.6, 39.6, 28.8, 28.5, 22.2, 14.1; IR (film): 3002,
2930, 2361, 1716, 1112, 618 cm⁻¹; HRMS (ESI) m/z calcd for C₂₅H₂₉Br₂ClNO₅, [M+H]⁺ 616.0096, found 616.0098.

A solution of protected compound (14.4 mg, 23.3 µmol) in trifluoroacetic acid (2 mL), was heated at 40 °C until complete disappearance of starting material (17 h), concentrated in vacuo and purified by flash chromatography on SiO₂ (EtOAc/hexanes, 5 to 30% EtOAc) to provide 9.8 mg (90%) of 5-57 as a colorless oil: Rf = 0.31 (30% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 7.81 (br s, 1H), 7.78 (s, 2 H), 6.11 (s, 1 H), 5.58 (d, J = 6.6 Hz, 1 H), 3.89 (s, 3 H), 3.89 (m, 1 H)*, 2.10 – 2.00 (m, 1 H), 1.74 – 1.84 (m, 1 H), 1.74 – 1.53 (m, 1 H), 1.52 – 1.38 (m, 3 H), 0.97 (t, J = 7.2 Hz, 3 H); * COSY correlation with proton at δ 2.05. ¹³C NMR (101 MHz, CDCl₃) δ 163.8, 153.5, 143.9, 133.2, 132.1, 118.4, 101.5, 101.3, 89.4, 64.2, 61.0, 32.5, 27.8, 22.3, 14.0; IR (film): 3150, 3058, 2925, 2361, 1725, 1362, 1268, 1002, 741 cm⁻¹; HRMS (ESI) m/z calcd for C₁₆H₁₉Br₂ClNO₃, [M+H]⁺ 467.9417, found 467.9399.

![Chemical Structure](image)

(Z)-5-(3,5-Dibromo-4-methoxybenzylidene)-2-(1,1-dichloropentyl)oxazolidin-4-one (5-58).

38 mg (11.6%) obtained (according to general procedure A) as a colorless oil: Rf = 0.40 (20% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 7.77 (s, 2 H), 7.20 (d, J = 8.6 Hz,
1 H), 6.47 – 6.43 (m, 2 H), 6.16 (s, 1 H), 5.53 (s, 1 H), 5.25 (d, J = 14.8 Hz, 1 H), 4.60 (d, J = 14.9 Hz, 1 H), 3.88 (s, 3 H), 3.80 (s, 3 H), 3.76 (s, 3 H), 2.32 – 2.16 (m, 2 H), 1.72 – 1.64 (m, 2 H), 1.47 – 1.39 (m, 2 H), 0.97 (t, J = 7.3 Hz, 3 H); \(^{13}\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 163.3, 161.3, 158.9, 153.4, 144.0, 133.1, 132.3, 131.8, 118.3, 115.0, 104.2, 101.1, 98.8, 94.1, 92.6, 60.9, 55.5, 43.2, 43.0, 26.3, 22.4, 14.0; IR (film): 3068, 2959, 2836, 1731, 1440, 997, 734 cm\(^{-1}\); HRMS (ESI) calcd for C\(_{25}\)H\(_{28}\)Br\(_2\)Cl\(_2\)NO\(_5\), [M+H]+ 649.97058, found m/z 649.97070.

A solution of protected compound (16.7 mg, 25.6 µmol) in trifluoroacetic acid (2 mL) was heated at 40 °C until complete disappearance of starting material (17 h), concentrated in vacuo and purified by flash chromatography on SiO\(_2\) (EtOAc/hexanes, 5 to 30% EtOAc) to provide 10 mg (78%) of 5-58 as a white film: Rf = 0.38 (30% EtOAc/hexanes); \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 8.22 (br s, 1 H), 7.78 (s, 2 H), 6.16 (s, 1 H), 5.77 (s, 1 H), 3.90 (s, 3 H), 2.41 – 2.19 (m, 2 H), 1.82 – 1.72 (m, 2 H), 1.54 – 1.40 (m, 2 H), 1.01 (t, J = 7.3 Hz, 3 H); \(^{13}\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 164.4, 153.8, 143.6, 133.4, 131.8, 119.6, 118.4, 102.1, 93.6, 90.9, 76.3, 60.9, 41.9, 26.4, 22.4, 14.0; IR (film): 3148, 3054, 2939, 2358, 1701, 1471, 1105, 551 cm\(^{-1}\); HRMS (ESI) m/z calcd for C\(_{16}\)H\(_{18}\)Br\(_2\)Cl\(_2\)NO\(_3\), [M+H]+ 499.9025, found 499.9028.
(Z)-2-(1-Chloropentyl)-5-(4-(trifluoromethyl)benzylidene) oxazolidin-4-one (61).

13 mg (5.2%) obtained (according to general procedure A) as a colorless oil: rf = 0.33 (20% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.76 (d, $J = 8.3$ Hz, 2 H), 7.60 (d, $J = 8.5$ Hz, 2 H), 7.30 – 7.26 (m, 1 H), 6.50 – 6.47 (m, 2 H), 6.29 (s, 1 H), 5.58 (d, $J = 1.9$ Hz, 1 H), 4.89 (d, $J = 14.6$ Hz, 1 H), 4.34 (d, $J = 14.7$ Hz, 1 H), 4.24 – 4.16 (m, 1 H), 3.86 (s, 3 H), 3.81 (s, 3 H), 1.58 – 1.52 (m, 2 H), 1.27 – 1.17 (m, 4 H), 0.85 (t, $J = 7.2$ Hz, 3 H); $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 162.2, 161.5, 158.4, 145.2, 137.3, 132.2, 129.3, 125.6, 125.6, 115.2, 105.0, 102.4, 100.0, 98.7, 92.6, 61.0, 55.65, 55.62, 39.4, 28.5, 28.5, 22.2, 14.0; IR (film): 3003, 2934, 2841, 1717, 1409, 1323, 1123, 863 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{25}$H$_{28}$ClF$_3$NO$_4$, [M+H]$^+$ 498.1654, found 498.1655.

A solution of protected compound (10.8 mg, 21.7 µmol) in trifluoroacetic acid (2 mL) was heated at 40 °C until complete disappearance of starting material (17 h), concentrated in vacuo and purified by flash chromatography on SiO$_2$ (EtOAc/hexanes, 5 to 30% EtOAc) to provide 6.2 mg (82%) of 5-61 as a white film: Rf = 0.26 (30% EtOAc/hexanes); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.97 (br s, 1 H), 7.73 (d, $J = 8.4$ Hz, 2 H), 7.62 (d, $J = 8.1$ Hz, 2 H), 6.31 (s, 1 H), 5.60 (d, $J = 6.8$ Hz, 1 H), 3.96 – 3.88 (m, 1 H), 2.15 – 1.99 (m, 1 H), 1.86 – 1.60 (m, 2 H), 1.1.58 – 1.46 (m, 3 H), 0.96 (t, $J = 7.2$ Hz, 3 H); $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 164.2, 144.4, 136.8, 129.8, 129.4, 126.0, 125.7, 125.64, 125.59, 103.1, 89.5, 64.1, 32.3,
27.9, 22.2, 14.0; IR (film): 3162, 2960, 2360, 1723, 1325, 1117, 862 cm⁻¹; HRMS (ESI) m/z calcd for C₁₆H₁₈ClF₃NO₂, [M+H]⁺ 348.0973, found 348.0972.

(Z)-2-(1,1-Dichloropentyl)-5-(4-(trifluoromethyl)benzylidene) oxazolidin-4-one (5-60).

36.5 mg (13.6%) obtained (according to general procedure A) as a colorless oil: Rf = 0.43 (20% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 7.72 (d, J = 8.3 Hz, 2 H), 7.59 (d, J = 8.3 Hz, 2 H), 7.22 (d, J = 8.2 Hz, 1 H), 6.49 – 6.44 (m, 2 H), 6.36 (s, 1 H), 5.54 (s, 1 H), 5.27 (d, J = 14.9 Hz, 1 H), 4.62 (d, J = 14.9 Hz, 1 H), 3.81 (s, 3 H), 3.76 (s, 3 H), 2.32 – 2.12 (m, 2 H), 1.77 – 1.67 (m, 2 H), 1.45 – 1.40 (m, 2 H), 0.96 (t, J = 7.3 Hz, 3 H); ¹³C NMR (75 MHz, CDCl₃) δ 163.6, 161.3, 158.9, 144.4, 137.0, 131.8, 129.3, 125.6, 115.0, 104.3, 102.8, 98.8, 94.3, 92.8, 55.54, 55.51, 43.4, 42.3, 26.3, 22.4, 14.0; IR (film): 3002, 2960, 2838, 1726, 1615, 1403, 1323, 866 cm⁻¹; HRMS (ESI) m/z calcd for C₂₅H₂₇Cl₂F₃NO₄, [M+H]⁺ 532.1264, found m/z 532.1283.

A solution of protected compound (27.1 mg, 50.9 µmol) trifluoroacetic acid (2 mL) was heated at 40 °C until complete disappearance of starting material (17 h), concentrated in vacuo and purified by flash chromatography on SiO₂ (EtOAc/hexanes (5 to 30% EtOAc) to provide 13.2 mg (68%) of 5-60 as a white film: Rf = 0.35 (30% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 8.73 (br s, 1 H), 7.74 (d, J = 8.3 Hz, 2 H), 7.63 (d, J = 8.4 Hz, 2 H),
6.37 (s, 1 H), 5.80 (s, 1 H), 2.33 (m, 1 H), 2.40 – 2.26 (m, 1 H), 2.24 – 2.12 (m, 1 H), 1.84 – 1.72 (m, 2 H), 1.52 – 1.40 (m, 2 H), 0.99 (t, J = 7.3 Hz, 3 H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 165.3, 144.1, 136.3, 129.7, 129.6, 125.7, 122.8, 104.1, 104.0, 93.4, 91.4, 91.3, 41.5, 26.5, 22.4, 14.0; IR (film): 3179, 2961, 2874, 2357, 1720, 1323, 1119, 866 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{16}\)H\(_{17}\)Cl\(_2\)F\(_3\)NO\(_2\), [M+H]\(^+\) 382.0583, found 382.0584.

(Z)-1-(3-(4-Oxo-5-(4-(trifluoromethyl)benzylidene)oxazolidin-2-yl)propyl)guanidine

(23).

To a solution of aldehyde 4-31 (84 mg, 0.25 mmol) and magnesium sulfate (150 mg, 1.25 mmol) in dichloromethane (1 mL) was added 2,4-dimethoxybenzylamine (38 \(\mu\)L, 0.25 mmol) and the resulting reaction mixture was stirred for 30 minutes until starting material disappeared, cooled to 0 °C and crude 2-Oxo-3-(4-(trifluoromethyl)phenyl)propanoyl chloride (0.25 mmol) 5-87 was added in dichloromethane (0.5 mL) via syringe. The reaction mixture was allowed to warm to room temperature over 17 h, concentrated \textit{in vacuo} and the oily residue purified by flash column chromatography on SiO\(_2\) (EtOAc/hexanes (5 to 40% EtOAc) to provide 35.3 mg (20%) of \(N^2, N^3\)-Bis(tert-butoxycarbonyl)-\(N^1\)-(3-(5-(Z)-4-(trifluoromethyl)benzylidene)-3-(2,4-dimethoxybenzyl)-4-oxooxazolidin-2-yl) propyl)guanidine as a light yellow oil: \(R_f = 0.29\) (30% EtOAc/hexanes); \(^1\)H NMR (400 MHz,
CDCl$_3$ $\delta$ 11.50 (br s, 1 H), 8.47 (br s, 1 H), 7.71 (d, $J = 8.2$ Hz, 2 H), 7.57 (d, $J = 8.3$ Hz, 2 H), 7.30 – 7.26 (m, 1 H), 6.49 – 6.43 (m, 2 H), 6.26 (s, 1 H), 5.41 (dd, $J = 5.6$, 2.0 Hz, 1 H), 4.94 (d, $J = 14.7$ Hz, 1 H), 4.25 (d, $J = 14.7$ Hz, 1 H), 3.83 (s, 3 H), 3.79 (s, 3 H), 3.59 – 3.39 (m, 2 H), 2.16 – 2.02 (m, 1 H), 1.85 – 1.73 (m, 1 H), 1.70 – 1.63 (m, 2 H), 1.48 (s, 18 H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 161.7, 161.3, 158.4, 156.1, 153.3, 145.8, 137.6, 131.9, 129.1, 128.9, 128.6, 125.5, 115.5, 104.7, 101.5, 101.4, 98.65, 98.58, 90.75, 90.68, 55.59, 55.51, 40.6, 38.8, 30.4, 28.3, 28.1, 22.0; IR(film): 3329, 3132, 2978, 1716, 1616, 1323, 1123, 733 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{34}$H$_{44}$F$_{3}$N$_{4}$O$_{8}$, [M+H]$^+$ 693.3106, found 693.3092.

A solution of protected compound (33.3 mg, 48.1 µmol) in trifluoroacetic acid (2.5 mL) was heated at 40 °C for 48 h, concentrated in vacuo and the residue purified by reverse phase column chromatography on C18 SiO$_2$ (H$_2$O/CH$_3$CN, 10 to 60% CH$_3$CN, 0.1% TFA additive) to provide 13.1 mg (79%) of 5-64 as a colorless oil: $^1$H NMR (400 MHz, MeOD) $\delta$ 7.83 (d, $J = 8.2$ Hz, 2 H), 7.63 (d, $J = 8.2$ Hz, 2 H), 6.20 (s, 1 H), 5.78 (t, $J = 4.7$ Hz, 1 H), 3.27 (t, $J = 7.0$ Hz, 2 H), 2.02 – 1.87 (m, 1 H), 1.85 – 1.69 (m, 3 H); $^{13}$C NMR (101 MHz, MeOD) $\delta$ 165.5, 158.6, 147.5, 139.2, 130.2, CF$_3$ (130.2, 129.9, 129.6, 129.9), 126.4, 101.7, 90.0, 41.9, 34.3, 23.3; HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{18}$F$_{3}$N$_{4}$O$_{2}$, [M+H]$^+$ 343.1376, found 343.1373.
1-((S)-3-Chloro-3-((S)-4-oxo-5-((Z)-4-(trifluoromethyl)benzylidene)oxazolidin-2-yl)propyl)guanidine (5-63).

To a solution of aldehyde 4-30 (83.6 mg, 22.9 µmol) and magnesium sulfate (140 mg, 1.17 mmol) in chloroform (1 mL) was added 2,4-dimethoxybenzylamine (35 µL, 0.23 mmol) at room temperature and the reaction mixture was stirred for 30 minutes until starting material disappeared. The reaction was cooled to 0 °C, crude 2-oxo-3-(4-(trifluoromethyl)phenyl)propanoyl chloride (0.25 mmol) in chloroform (0.7 mL) was added via syringe and stirred overnight. The reaction mixture was concentrated in vacuo and oily residue purified by flash column chromatography on SiO$_2$ (Et$_2$O/hexanes, 10-75% Et$_2$O) to provide 14.5 mg (8.7%) of protected product as a light yellow oil (inseparable mixture of diastereomers ~3:1): Rf = 0.44 (60% Et$_2$O/hexanes); Characteristic signals of major diastereomer: $^1$H (400 MHz, CDCl$_3$) 5.44 (s, 1 H), 5.02 (d, $J$ = 14.8 Hz, 1 H)

A solution of the semi-purified protected compound (11 mg, 0.015 mmol) in dichloromethane (1 mL) and trifluoroacetic acid (1 mL) was stirred at room temperature until disappearance of starting material (~2 h), concentrated in vacuo and purified by reverse phase column chromatography on C18 SiO$_2$ (H$_2$O/CH$_3$CN, 10 to 50% CH$_3$CN, 0.1% TFA additive) to provide 5.1 mg (86%, calculated for major diastereomer) of deprotected
A solution of deprotected guanidine (5.0 mg, 9.5 µmol) in trifluoroacetic acid (1 mL) was heated at 40 °C for 48 hours, concentrated in vacuo and purified by reverse phase column chromatography on C18 SiO2 (H2O/CH3CN, 10-60% CH3CN, 0.1% TFA additive) to provide 3.1 mg (91%) of 5-63 as a colorless oil: 1H NMR (500 MHz, MeOD) δ 7.84 (d, J = 8.2 Hz, 2 H), 7.65 (d, J = 8.3 Hz, 2 H), 6.25 (s, 1 H), 5.92 (d, J = 2.3 Hz, 1 H), 4.30 (dt, J = 11.1, 2.5 Hz, 1 H), 3.57 – 3.41 (m, 2 H), 2.30 – 2.20 (m, 1 H), 2.15 – 2.01 (m, 1 H); 13C NMR (126 MHz, MeOD) δ 165.7, 158.7, 147.1, 145.9, 138.8, 130.4, 126.4, 102.1, 90.2, 61.7, 39.6, 32.2; HRMS (ESI) m/z calcd for C15H17ClF3N4O2 [M+H]+ 377.0987, found 377.0993.
(Z)-1-(3,3-Dichloro-3-(5-(3,5-dibromo-4-methoxybenzylidene)-4-oxooxazolidin-2-yl)propyl)guanidine (5-59).

To a solution of 16 (80 mg, 0.24 mmol) and magnesium sulfate (0.1 g, 0.8 mmol) in dichloromethane (1 mL) was added 2,4-dimethoxybenzylamine (35 µL, 0.23 mmol) and the resulting slurry stirred for 5 min. N-chlorosuccinimide (28 mg, 0.21 mmol) was added in one portion and the reaction mixture was stirred at room temperature for 1 h. To this was added a solution of crude acid chloride in dichloromethane (0.5 mL) at 0 °C and reaction mixture was allowed to warm to room temperature and stirred for 6 hours, diluted with dichloromethane and filtered through a plug of Celite®, filter cake was washed with dichloromethane, solvent concentrated in vacuo and the brown oily residue purified by flash chromatography on SiO$_2$ (EtOAc/hexanes, 10 to 50% EtOAc) to provide 26.6 mg (30%) of protected product as a light yellow oil that was carried on to the next step.

A solution of protected product (24.1 mg, 27.3 µmol) in trifluoroacetic acid (3 mL) was heated at 40 °C for 96 hours, concentrated in vacuo and the residue purified by reverse phase column chromatography on C18 SiO$_2$ (H$_2$O/CH$_3$CN, 10-60% CH$_3$CN, 0.1% TFA additive) to provide 7.9 mg (55%) of 5-59 as a colorless oil: $^1$H NMR (400 MHz, MeOD) $\delta$ 7.90 (s, 2 H), 6.16 (s, 1 H), 6.03 (s, 1 H), 3.87 (s, 3 H), 3.75 (t, $J = 7.1$ Hz, 2 H), 2.69 – 2.51
(m, 2 H); $^{13}$C NMR (101 MHz, MeOD) $\delta$ 165.7, 158.7, 154.8, 145.8, 134.3, 133.6, 119.2, 101.5, 95.9, 92.5, 61.2, 41.3, 38.4; IR (film): 3347, 3189, 1678, 1472, 1353, 1202, 994 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{15}$H$_{17}$Br$_2$Cl$_2$N$_4$O$_3$ [M+H]$^+$ 528.9039, found 528.9044.
5.6 References


(40) Bressy, C.; Menant, C.; Piva, O. Synlett. 2005, 577.


CHAPTER 6

Antimicrobial Activity of 4-oxazolidinones
6.1 Abstract

Antimicrobial evaluation of novel 4-oxazolidinones is presented herein. A series of the analogues of synoxazolidinones A and B were tested against several strains of clinically relevant bacteria. Efforts to increase the potency and selectivity of this class of compounds are described, as well as studies aimed at identifying their intracellular targets. Our antimicrobial SAR studies confirmed the importance of the chlorine substituent for antimicrobial activity and revealed simplified dichloro- derivatives that are equally potent against MRSA.

6.2 Introduction and Background

The world-wide spread of antibiotic resistant bacteria causes life-threatening and untreatable infections that have been more frequently acquired in the community and in hospitals. The problem is the proliferation of so-called “ESKAPE” microorganisms (Enterococcus spp., Staphylococcus aureus, Klebsiella spp., Acinetobacter baumannii, Pseudomonas aeruginosa and Enterobacter spp.) the treatment of which is becoming difficult, or in some cases impossible. Antibacterial drug resistance continues to pose a substantial threat to public health and modern medicine world-wide resulting in enormous economic and human cost. The numbers are alarming across the globe: out of 400,000 instances of infections caused by resistant pathogens 25,000 were lethal (Europe, 2007); excess health care costs associated with antibiotic resistance in the US health system reached
20 billion dollars per year; 54% of isolated bacterial strains of *Staphylococcus aureus* (~23,000) from Ukrainian hospitals in 2010 were resistant to antimicrobial treatments.

The issue of resistance is amplified by the lack of new therapeutic options in the antibiotic development pipeline. Typically, two approaches are undertaken towards the discovery of more potent antimicrobials against pathogens. These include investigation/targeting resistance pathways in bacteria and searching for antibacterial compounds with novel modes of action. The first approach is aiming to conserve the efficacy of existing antimicrobials by replacing ineffective ones by their chemical scaffold analogues with improved activity. For the second approach identification of novel molecular scaffolds is required.

Understanding how antimicrobial natural products and their structural derivatives kill pathogenic bacteria is essential for the discovery of new pathways to combat resistance. Knowledge of their mechanism of action may provide scientists with insights on how to overcome pathogenicity in microbes and restore/enhance effectiveness of current treatments. In addition, influencing bacterial metabolism and mutagenesis with biologically active small molecules such as natural products may uncover new targets giving rise to the next generation of antibiotics.

Having synoxazolidinone natural products in hand as well as substantial amounts of their 4-oxazolidinone analogues (Scheme 6.1), we set out for the systematic evaluation of their antimicrobial properties.
**Scheme 6.1**: Sites of modification in our 4-oxazolidinone library.

### 6.3 Results and Discussion

#### 6.3.1 MIC Values of 4-oxazolidinones Against Pathogenic Bacteria

The first step in the evaluation of antimicrobial properties of our 4-oxazolidinone library was determination of the Minimum Inhibitory Concentration (MIC) for each analogue. MIC is defined as a concentration of the compound at which visible bacterial growth is inhibited. The MIC was determined according to CLSI guidelines using microdilution protocol (Table 6.1 - 6.4). Screening was performed against several clinically relevant bacterial strains and standard antibiotics (linezolid, vancomycin and tetracycline, Table 6.2) were used as control.

At the outset of our efforts there was concern regarding the stability of the 4-oxazolidinone heterocycles for use as chemical probes and medicinal lead structures; however, the 4-oxazolidinone products have proven bench stable for months and can be purified by reverse phase chromatography (0.1% TFA). Further, no significant decomposition was observed in biologically relevant aqueous buffers for extended periods of time.
With synoxazolidinone A (2-26) and B (2-27) and their analogues in hand, we explored the antimicrobial activity of these compounds against several bacterial strains. The activities of compounds 2-26 and 2-27 are in line with those reported for the isolated natural products against both sensitive and resistant gram-positive bacteria *Staphylococcus aureus* (Entries 1 and 2, Table 6.1). Interestingly, a 2:1 mixture of diastereomers (Entry 3, Table 6.1) of synoxazolidinone A (5-62) appeared slightly less active than single major diastereomer (2-26).

**Table 6.1: Antimicrobial activity of guanidine-containing 4-oxazolidinones.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Compound**</th>
<th>MIC against various bacteria, µg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>Staphylococcus aureus</em></td>
</tr>
<tr>
<td>1</td>
<td>2-27</td>
<td>23.1</td>
</tr>
<tr>
<td>2</td>
<td>2-26</td>
<td>6.2</td>
</tr>
<tr>
<td>3</td>
<td>5-62</td>
<td>12.4</td>
</tr>
<tr>
<td>4</td>
<td>5-59</td>
<td>12.5</td>
</tr>
<tr>
<td>5</td>
<td>5-64</td>
<td>60</td>
</tr>
<tr>
<td>6</td>
<td>5-63</td>
<td>&gt;100</td>
</tr>
<tr>
<td>7</td>
<td>6-1</td>
<td>&gt;128</td>
</tr>
<tr>
<td>8</td>
<td>6-2</td>
<td>&gt;128</td>
</tr>
</tbody>
</table>

- - not tested; >"number" – number indicates maximum concentration tested; * - MIC values of compounds which were tested against E. coli were also the same (inactive) against *Yersinia pseudotuberculosis*, *Vibrio cholerae*, *Enterobacter aerogenes*, *Ochrobactrum anthropi* and *Providencia alcalifaciens* (not shown in the tables). ** - see Scheme 6.2 for structures of compounds.

Antimicrobial activity of 4-oxazolidinones against other bacteria other than *Staphylococcus aureus* and MRSA was tested by the Linigton lab as a part of their HTS antibiotic screening.²

*Staphylococcus aureus* – ATCC 29213; MRSA – ATCC 33591 – methicillin-resistant *Staphylococcus aureus*; *Acinetobacter baumannii* – ATCC 19606

156
Scheme 6.2: Antimicrobial activity of guanidine-containing 4-oxazolidinones (structures of compounds from Table 6.1).

Table 6.2: Antimicrobial activity of commercially available antibiotics.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Compound</th>
<th>MIC against various bacteria, µg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Staphylococcus aureus</td>
</tr>
<tr>
<td>1</td>
<td>Vancomycin</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>Tetracycline</td>
<td>0.25</td>
</tr>
<tr>
<td>3</td>
<td>Linezolid</td>
<td>0.5</td>
</tr>
</tbody>
</table>
Table 6.3: Antimicrobial activity of aliphatic 4-oxazolidinones containing varied aromatic substituent.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Compound**</th>
<th>MIC against various bacteria, µg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>Staphylococcus aureus</strong></td>
<td><strong>Staphylococcus aureus (MRSA)</strong></td>
</tr>
<tr>
<td>1</td>
<td>4-24</td>
<td>10.8</td>
</tr>
<tr>
<td>2</td>
<td>5-57</td>
<td>&gt;50</td>
</tr>
<tr>
<td>3</td>
<td>5-57a</td>
<td>23.4</td>
</tr>
<tr>
<td>4</td>
<td>5-58</td>
<td>50</td>
</tr>
<tr>
<td>5</td>
<td>4-22</td>
<td>16</td>
</tr>
<tr>
<td>6</td>
<td>5-61</td>
<td>&gt;100</td>
</tr>
<tr>
<td>7</td>
<td>5-60</td>
<td>10</td>
</tr>
<tr>
<td>8</td>
<td>4-26</td>
<td>14.5</td>
</tr>
<tr>
<td>9</td>
<td>4-23</td>
<td>&gt;128</td>
</tr>
<tr>
<td>10</td>
<td>4-25</td>
<td>128</td>
</tr>
<tr>
<td>11</td>
<td>4-18</td>
<td>25.9</td>
</tr>
<tr>
<td>12</td>
<td>4-19</td>
<td>30.1</td>
</tr>
<tr>
<td>13</td>
<td>2-26</td>
<td>&gt;32</td>
</tr>
<tr>
<td>14</td>
<td>2-27</td>
<td>&gt;32</td>
</tr>
<tr>
<td>15</td>
<td>6-7</td>
<td>&gt;37</td>
</tr>
<tr>
<td>16</td>
<td>4-17</td>
<td>64</td>
</tr>
<tr>
<td>17</td>
<td>4-17a</td>
<td>64</td>
</tr>
<tr>
<td>18</td>
<td>4-17b</td>
<td>64</td>
</tr>
</tbody>
</table>

- - not tested; >“number” – number indicates maximum concentration tested; * - MIC values of compounds which were tested against E. coli were also the same (inactive) against Yersinia pseudotuberculosis, Vibrio cholerae, Enterobacter aerogenes, Ochrobactrum anthropi and Providencia alcalifaciens (not shown in the tables). ** - see Scheme 6.3 for structures of compounds.

Antimicrobial activity of 4-oxazolidinones against other bacteria other than Staphylococcus aureus and MRSA was tested by the Lingston lab as a part of their HTS antibiotic screening.

Staphylococcus aureus – ATCC 29213; MRSA – ATCC 33591 – methicillin-resistant Staphylococcus aureus; Acinetobacter baumannii – ATCC 19606
Scheme 6.3: Antimicrobial activity of aliphatic 4-oxazolidinones containing varied aromatic substituent (structures of compounds from Table 6.3).
The synthesized compounds shown in Table 6.3 were designed to probe the impact of the benzylidene fragment of the synoxazolidinones on antimicrobial activity. Generally, deletion of the guanidine moiety on dichloro (5-59 vs 5-58) and deschloro (2-27 vs 4-24) derivatives did not affect their antimicrobial activity (Table 6.1, Table 6.3) but was detrimental to the antimicrobial activity of monochloro derivatives (5-63 vs 5-61); the installation of an additional chloride or an electron-withdrawing group on the aryl ring was generally beneficial. In particular, compounds 5-59 and 5-60 displayed comparable activity to that of 2-26 and increased activity against *A. baumannii.*

**Table 6.4:** Antimicrobial activity of miscellaneous 4-oxazolidinones.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Compound**</th>
<th>MIC against various bacteria, µg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>Staphylococcus aureus</em></td>
</tr>
<tr>
<td>1</td>
<td>6-6</td>
<td>23.1</td>
</tr>
<tr>
<td>2</td>
<td>6-9</td>
<td>43.0</td>
</tr>
<tr>
<td>3</td>
<td>6-10</td>
<td>&gt;20</td>
</tr>
<tr>
<td>4</td>
<td>6-3</td>
<td>&gt;128</td>
</tr>
<tr>
<td>5</td>
<td>6-4</td>
<td>&gt;128</td>
</tr>
<tr>
<td>6</td>
<td>6-5</td>
<td>&gt;20</td>
</tr>
<tr>
<td>7</td>
<td>6-11</td>
<td>&gt;32</td>
</tr>
<tr>
<td>8</td>
<td>5-61</td>
<td>&gt;37</td>
</tr>
<tr>
<td>9</td>
<td>6-8</td>
<td>29.2</td>
</tr>
</tbody>
</table>

- - not tested; >"number" – number indicates maximum concentration tested; *- MIC values of compounds which were tested against *E. coli* were also the same (inactive) against *Yersinia pseudotuberculosis, Vibrio cholerae, Enterobacter aerogenes,* *Ochrobactrum anthropi* and *Providencia alcalifaciens* (not shown in the tables). **- see Scheme 6.4 for structures of compounds.

Antimicrobial activity of 4-oxazolidinones against other bacteria other than *Staphylococcus aureus* and MRSA was tested by the Linigton lab as a part of their HTS antibiotic screening.2

*Staphylococcus aureus* – ATCC 29213; MRSA – ATCC 33591 – methicillin-resistant *Staphylococcus aureus; Acinetobacter baumannii* – ATCC 19606

160
Overall, analogues containing electron-rich aryl groups (4-25, 4-18 and 4-19) possessed low antimicrobial activity against Gram-positive *Staphylococcus aureus* and MRSA and no activity at the concentrations tested against Gram negative *Acinetobacter baumannii*. Conversely, electron-deficient aromatic rings (4-26, 4-22) were significantly more potent with the *p*-CF$_3$ series (4-22, 5-60, 5-61, 5-64) displaying the lowest MIC across the various species of bacteria. Trifluoromethyl-substituted oxazolidinones have improved antimicrobial activity, are more easily accessible and subsequently inspired natural product analogue (5-59).
6.3.2 Biofilm Inhibition Properties of 4-oxazolidinones

Bacterial biofilms are a part of the bacterial lifestyle in which single-cell organisms adopt multicellular “group behavior” which facilitates survival in adverse environments (dehydration, pH and temperature changes, antibiotic treatment). Such a bacterial community is embedded in an extracellular polymeric substance (EPS) – a slimy matrix consisting of polysaccharides, proteins, nucleic acids and lipids. EPS is very important for bacteria, as it mediates their initial attachment to different surfaces, causing for example colonization of medical implants. Bacteria surrounded by EPS compared to the planktonic state are more virulent and insensitive to antibiotics; biofilm also creates a physical barrier for antibiotic penetration.

Very often biofilms contain compartments deficient in oxygen and nutrients. These conditions within biofilms force bacterial cells to go into a dormant metabolic state, rendering processes such as cell-wall biosynthesis and translation (typical antibiotic targets) inactive. Such cells are called persisters and are virtually inaccessible by common antibiotics, especially when they are within EPS. As a result, biofilm-associated infections have a high percentage of relapse and are several orders of magnitude more resistant to antibiotic treatment. For example, showed that slowly growing colonies of *S. aureus* are more resistant to cell-wall-active antibiotics, because cell-wall division is slowed down which reduces the efficacy of β-lactam antibiotics.

Given the significant role of the biofilm-associated communities in survival of pathogenic bacteria, there is a strong need in the development of small molecules which inhibit or disrupt bacterial biofilms. One of the requirements for biofilm inhibitors is that
they do not directly influence bacterial survival of bacteria, but rather disrupt formation of EPS, enabling other antibiotics to exert their action.\textsuperscript{10,13} It is for this reason that it is expected that resistance to anti-biofilm compounds will not develop rapidly. Many biofilm-disrupting small molecules have been discovered from diverse natural sources, such as 2-aminoimidazoles,\textsuperscript{14} brominated furanones,\textsuperscript{15} triterpenes,\textsuperscript{16} corosolic and asiatic acids.\textsuperscript{17} Inspired by the natural product oroidin (6-12), Melander and coworkers have developed several 2-aminoimidazole based analogues as effective non-microbicidal agents that can modulate bacterial biofilms (Scheme 6.5).

![Scheme 6.5: Natural (6-12) and synthetic (6-13) biofilm inhibitors and methicillin.](image)

Melander \textit{et al.} have also demonstrated that 2-aminoimidazole derivative 6-13, when used as an adjuvant (45 \textmu M) in combination with methicillin (25 \textmu M), resulted in 90\% reduction in MRSA growth.\textsuperscript{9} Significantly, a four-fold reduction of MIC value of methicillin (6-14) was observed. In these studies, other resistant bacteria (multidrug-resistant \textit{Acinetobacter baumannii} and \textit{E. coli}) was also resensitized by compound 6-13 to the action of conventional antibiotics.
Table 6.5: Effects of synoxazolidinone A (2-26) on adhesion and growth of fouling marine microorganisms.

<table>
<thead>
<tr>
<th>organism</th>
<th>Ad(^a)</th>
<th>Gr(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marine Bacteria</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Halomonas aquamarina</td>
<td>20</td>
<td>-</td>
</tr>
<tr>
<td>Polaribacter erogenii</td>
<td>-</td>
<td>20</td>
</tr>
<tr>
<td>Pseudoalteromonas elyakovii</td>
<td>-</td>
<td>0.02</td>
</tr>
<tr>
<td>Roseobacter litoralis</td>
<td>-</td>
<td>0.02</td>
</tr>
<tr>
<td>Shewanella putrefaciens</td>
<td>-</td>
<td>0.2</td>
</tr>
<tr>
<td>Vibrio aestuarianus</td>
<td>-</td>
<td>0.02</td>
</tr>
<tr>
<td>Vibrio carteri</td>
<td>-</td>
<td>2</td>
</tr>
<tr>
<td>Vibrio harveyi</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Vibrio natriegens</td>
<td>-</td>
<td>0.02</td>
</tr>
<tr>
<td>Vibrio proteolyticus</td>
<td>-</td>
<td>0.02</td>
</tr>
<tr>
<td>Microalgae</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cylindrotheca closterium</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Exantherachyris gairaliensis</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Halamphora coffeeaformis</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Pleurochrysis roseoffensis</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Porphyridium purpureum</td>
<td>-</td>
<td>20</td>
</tr>
<tr>
<td>Crustacean Settlement</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Balanus improvisus (IC(_{50}))</td>
<td>15</td>
<td></td>
</tr>
</tbody>
</table>

Ref. (4)

\(^a\) adhesion inhibition; \(^b\) growth inhibition;
\(^c\) compound inactive at concentrations ≥10 µg/mL.

In recent studies by Trepos et al. exploration of biofilm inhibition properties of marine natural products including synoxazolidinone A and C was carried out against 16 marine bacterial species.\(^4\) The synoxazolidinones displayed MIC values (Table 6.5) in the micro- and nanomolar range, with the growth of several strains being inhibited at as low as 20 nM, and were shown to affect both the micro and macrofouling processes. As can be seen (Table 6.5), the MIC values of the synoxazolidinone A (2-26) against marine species are, significantly lower than MIC values we obtained against terrestrial pathogens (Table 6.1). This difference shows increased antibacterial effect of synoxazolidinone A towards marine environment and implies that the synoxazolidinones have been evolved by the producing

164
organism to target marine bacteria and that this high antibiotic potency is not directly translated to kill terrestrial bacteria.\textsuperscript{4}

These findings prompted our curiosity to explore biofilm inhibition properties of synoxazolidinone A and analogues (Figure 6.1 - 6.7). MRSA (BAA-44) was used as representative gram-positive bacterial strain for all tested compounds in our biofilm inhibition studies. IC\textsubscript{50} is defined as the concentration of compound required to inhibit 50\% of biofilm growth, compared to an untreated control. The ability of each compound to inhibit biofilm formation was assessed via crystal violet reporter assay.\textsuperscript{11} We found that synoxazolidinone A derivative 5-59 (Figure 6.2) with additional Cl- atom had slightly better IC\textsubscript{50} value than natural product itself (Figure 6.1). For both 2-26 and 5-59 \textasciitilde 80\% of MRSA biofilm was inhibited at 50 \textmu M which is above MIC values of these compounds. Interestingly, higher concentrations of 2-26 and 5-59 did not cause complete inhibition of biofilm due to anticipated killing of the bacteria, instead biofilms were produced in increased amounts rendering 2-26 and 5-59 inactive at high concentrations.

An interesting effect of deletion of guanidine group in pCF\textsubscript{3}- series was observed: deschlorinated derivative 4-22 (Figure 6.3) was more potent in inhibiting MRSA biofilm compared to 5-64 (Figure 6.4) and the monochlorinated pCF\textsubscript{3}- analogue lacking guanidine 5-61 (Figure 6.5) was rather inactive compared to 5-63 (Figure 6.6). The most potent 4-oxazolidinone biofilm inhibitor we discovered was 5-60 (Figure 6.7), which had the lowest IC\textsubscript{50} value of \textasciitilde 1.2 \textmu M in the series (Table 6.6).
Figure 6.1: Inhibition of MRSA biofilm formation by 2-26.

Figure 6.2: Inhibition of MRSA biofilm formation by 5-59.
**Figure 6.3:** Inhibition of MRSA biofilm formation by 4-22.

**Figure 6.4:** Inhibition of MRSA biofilm formation by 5-64.
Figure 6.5: Inhibition of MRSA biofilm formation by 5-61.

Figure 6.6: Inhibition of MRSA biofilm formation by 5-63.
**Figure 6.7:** Inhibition of MRSA biofilm formation by 5-60.

**Table 6.6:** MIC and IC\textsubscript{50} values of 4-oxazolidinones against MRSA.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Compound</th>
<th>MIC (MRSA)\textsuperscript{a} (\mu\text{g/mL})</th>
<th>MIC (MRSA)\textsuperscript{a} (\mu\text{M})</th>
<th>IC\textsubscript{50} (MRSA)\textsuperscript{b} (\mu\text{M})</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2-26</td>
<td>12.4</td>
<td>25</td>
<td>47.58</td>
</tr>
<tr>
<td>2</td>
<td>5-59</td>
<td>20.0</td>
<td>38</td>
<td>30.99</td>
</tr>
<tr>
<td>3</td>
<td>4-22</td>
<td>16.0</td>
<td>51</td>
<td>52.00</td>
</tr>
<tr>
<td>4</td>
<td>5-64</td>
<td>40.0</td>
<td>117</td>
<td>170.06</td>
</tr>
<tr>
<td>5</td>
<td>5-61</td>
<td>&gt;100</td>
<td>&gt;288</td>
<td>&gt;200</td>
</tr>
<tr>
<td>6</td>
<td>5-63</td>
<td>&gt;100</td>
<td>&gt;266</td>
<td>80.18</td>
</tr>
<tr>
<td>7</td>
<td>5-60</td>
<td>18.0\textsuperscript{b} (20.0\textsuperscript{a})</td>
<td>47\textsuperscript{b} (52\textsuperscript{a})</td>
<td>1.26</td>
</tr>
</tbody>
</table>

\textsuperscript{a} – ATCC 33591, \textsuperscript{b} – BAA-44. MIC values for BAA-44 appeared similar to those found for ATCC 33591.
Bacteria are often exposed to non-lethal concentrations of antibiotics, which was shown to contribute to the evolution of resistance. Continued exposure to sub-MIC concentrations of antibiotics which act on bacterial cell-wall or inhibit protein synthesis was shown to stimulate biofilm formation. Bacteria can also up-regulate production of biofilms in response to high levels of cellular damage caused by high concentration of antimicrobial agents. Figures 6.1 and 6.2 show that high concentrations (~4 x MIC) of 2-26 and 5-59 result in overproduction of biofilm (as indicated by low % inhibition at above IC<sub>50</sub> concentrations) not observed for other compounds tested.

For CF<sub>3</sub>-containing analogues 4-22, 5-63 and 5-60, the amount of biofilm inhibition remained constant at high concentrations (exceeding their IC<sub>50</sub> and MIC values). Interestingly, 5-63 is not toxic to MRSA, but successfully inhibits biofilm formation (Figure 6.6, Table 6.6).

With lead compound 5-60 in hand we carried out additional antimicrobial studies such as MRSA growth curve analysis (Figure 6.8) and resensitization of MRSA towards the action of methicillin (Figure 6.9, Table 6.7).
By performing growth curve analysis we further demonstrated that compound 5-60 is not exerting microbicidal action on MRSA at concentrations above the IC$_{50}$, up to 8 µM (Figure 6.8). Significant killing is observed at concentrations above 16 µM (which roughly corresponds to ~30% of MIC value for 5-60). Therefore, we continued exploration of
antimicrobial properties of this potent analogue and performed preliminary resensitization studies in which methicillin was used at different concentrations in presence of sub-MIC levels of our lead compound 5-60.

To probe suppression of antibiotic resistance, the MIC of methicillin was determined in the presence of 21%, 42% and 85% the MIC of 5-60. TSBG media was inoculated with MRSA (BAA-44), and methicillin was serially diluted in both the inoculated media alone to serve as a control, as well as inoculated media supplemented with 5-60. The MICs of the methicillin in the presence of 5-60 were recorded and compared with that of MIC of methicillin alone. Fold-reduction was calculated by dividing the MIC of methicillin by the MIC of methicillin with 5-60 (Figure 6.9, Table 6.7).

![MRSA resensitization to methicillin](image)

**Figure 6.9:** Resensitization of MRSA to methicillin by 5-60.

A 4-fold reduction in MIC of methicillin was observed in presence of 10 µM of 5-60 when the resensitization assay was performed in TSBG media (Figure 6.9, blue); however, when
the media was changed to MHB, 5-60 had no significant influence in improving MIC of methicillin (Figure 6.9, red).

Table 6.7: Effect of media on MIC of methicillin in presence of varied amounts of 5-60.

<table>
<thead>
<tr>
<th>Concentration 5-60, µM</th>
<th>MIC methicillin µg/mL (TSBG)</th>
<th>Resensitization (TSBG)</th>
<th>MIC methicillin µg/mL (MHB)</th>
<th>Resensitization (MHB)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>128</td>
<td>-</td>
<td>128</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>32</td>
<td>4-fold</td>
<td>128</td>
<td>-</td>
</tr>
<tr>
<td>20</td>
<td>8</td>
<td>16-fold</td>
<td>128</td>
<td>-</td>
</tr>
<tr>
<td>40</td>
<td>2</td>
<td>64-fold</td>
<td>64</td>
<td>2-fold</td>
</tr>
</tbody>
</table>

We also studied effects of various antibiotics, including 4-oxazolidinones on bacterial growth and translation in an *in vitro* prokaryotic protein expression system (S30 *E. coli* extract). The protein expression assay was performed using *E. coli* Extract System for Circular DNA (Promega, product L1020) according to manufacturer guidelines and protocol.12
Initially we tested synoxazolidinone B (2-27) and few simple analogues (4-24 and 4-26) and used chloramphenicol and erythromycin as references for complete protein inhibition. We determined that 4-oxazolidinones 4-24, 4-26 and 2-27 do not inhibit protein synthesis.

Figure 6.10: Effects 4-oxazolidinones on protein synthesis.
6.4 Conclusions and Outlook

Despite common misconceptions, it is often possible to achieve improved antimicrobial activity for the natural products by introducing modifications around natural product’s core structures. In our SAR studies of synoxazolidinones we explored various structural modification strategies, such as decorating (introduction of additional chlorine atom), degrading (removal of the guanidine group) and substituting (dibromomethoxy to $p$CF$_3$-containing aromatic). We demonstrated that improved antimicrobial activity against MRSA can be achieved with simplified analogues. The “chemical post-evolution” of the synoxazolidinones explored in our lab provided important learning cycles for future structure optimization aimed at improvement antimicrobial and anti-biofilm potency of these compounds as well as search for their biological targets.
6.5 Experimental

**Broth microdilution protocol for minimum inhibition concentration (MIC) determination**

Fresh Mueller–Hinton broth (MHB) – 3 ml, was inoculated (5 × 10^5 CFU mL^-1 ) with MRSA (ATCC 33591 or BAA-44) methicillin-resistant *Staphylococcus aureus*. The resulting bacterial suspension was aliquoted (1 mL) into culture tubes and test compound (10 mM DMSO stock solution) was added to reach the final concentration of the interest. Bacteria not treated with the antimicrobial compound served as the control. Rows 2–12 of a 96-well microtiter plate were filled with remaining bacterial subcultures. The samples containing test compound were aliquoted (200 µL) into the first row wells of the microtiter plate (2 wells per compound). Row 1 wells were mixed 6 – 8 times and 100 µL was transferred down to row 2. Row 2 wells were mixed 6 – 8 times, followed by a 100 µL transfer from row 2 down to row 3. The rest of the rows of the plate were serially diluted in similar fashion. The plate was covered with sealing tape for 96-well plates (ThermoFisher) and incubated under stationary conditions at 37 °C. Duplicate of the plate was prepared. The lowest concentration of test compound at which no visible growth of bacteria occurred – minimum inhibitory concentration (MIC) values were recorded after 16 h. ³

**Inhibition of MRSA biofilm formation**

MRSA (BAA-44) was subculture overnight to an OD600 of 0.01 into tryptic soy broth with 5% glucose (TSBG). Solutions of desired concentrations (sub-MIC) of the test compound were prepared in the resulting bacterial suspension and 100 µL was aliquoted into
the wells of a PVC 96-well plate (and duplicate). Bacterial suspension to which no antimicrobial compounds was added served as control. Plates were covered with with sealing tape for 96-well plates (ThermoFisher) and incubated under stationary condition at 37 °C for 24 h. After which plates washed thoroughly with water discarding media and planktonic bacterial cells. Each well of washed plates was stained with 110 µL 0.1% solution of crystal violet (CV) at room temperature for 30 min. After which plates were thoroughly washed with water again to remove excess of CV. The remaining on the well wall stain was dissolved in 200 µL 95% ethanol, and 100 µL was transferred to the corresponding wells of a polystyrene microtiter dish for OD reading. Biofilm inhibition was quantified by plate reader set at OD$_{540}$ of each well. Blank wells were used as background and subtracted out. Percent of inhibition for compound was calculated relative to the untreated control wells.$^{3}$

**Broth microdilution method for antibiotic resensitization**

Fresh Mueller-Hinton Broth (MHB) or tryptic soy broth with 5% glucose (TSBG) was inoculated (5x10$^5$ CFU mL$^{-1}$) with MRSA. Aliquots (5 mL) of the resulting bacterial suspension were distributed to culture tubes, and compound from a 10 mM stock solution was added to give the final concentration of interest. Bacteria not treated with antimicrobial compound served as a control. After resting for 30 minutes at room temperature, 1 mL of each sample was transferred to a culture tube, and methicillin sodium salt was added from a 128 mg mL$^{-1}$ water stock solution to give a final concentration of 128 µg mL$^{-1}$. Rows 2-12 of a 96-well plate were filled (100 µL per well) from the remaining 4 mL bacterial subculture. After standing for 10 minutes, aliquots (200 µL) of the samples containing antibiotic were
distributed across the corresponding first row wells of the microtiter plate. First row was mixed 6-8 times, and 100 µL was transferred from row 1 to down to row 2. This procedure was then repeated to dilute the rest of the wells. Bacterial growth in the presence of compound alone to which no methicillin was added – row 1. Row 11 – methicillin. The plate was then covered and sealed with sealing tape for 96-well plates (ThermoFisher) and incubated under stationary conditions at 37 °C. The lowest concentration of test mixture (methicillin and test compound) at which no visible growth of bacteria occurred – minimum inhibitory concentration (MIC) values were recorded after 16 h. Fold reduction was determined by comparison with control lane.

**Growth curve analysis**

Growth curve analysis was performed by subculturing MRSA at an OD₆₀₀ of 0.01 into the MHB media from an overnight culture in duplicates. 3 mL aliquots of subcultured MRSA was grown at subMIC concentrations in presence or absence of the test compound in an incubator at 37 °C with shaker set to 200 rpm. Optical densities were taken at 2, 4, 6, 8 and 24 hours. 100 µL were withdrawn and diluted 10-fold. Bacteria in absence of any compound served as control. OD₆₀₀ measurements were plotted against time.
6.6 References


(12) https://www.promega.com/~media/files/resources/protcards/e%20coli%20s30%20extract%20system%20for%20circular%20dna%20quick%20protocol.pdf


CHAPTER 7

Synthesis and Application of 1,5-dihydro-2H-pyrrol-2-ones as Precursors for Novel Highly Functionalized \( N \)-containing heterocycles and \( \beta \)-amino acids
7.1 Abstract

Our endeavors during total the synthesis of the synoxazolidinone family of natural products also uncovered another type of interesting heterocyclic scaffold: 1,5-dihydro-2H-pyrrol-2-ones. Conditions were identified that can turnover the reaction selectivity to C-addition and lead to functionalized pyrrolidinones. Current methods for their synthesis involve heating in the presence of Brønsted or Lewis acids and suffer from enolizable aldehydes producing low yields. We successfully optimized mild conditions to obtain alkyl substituted dihydro-pyrrol-2-ones in high yields. Investigation into the reactivity of these heterocycles provided novel pyrrolidine-2,3-dione scaffolds and the inspiration for an approach to anticancer natural product pretazettine. Progress towards the total synthesis of pretazettine is described. Finally, we utilized novel pyrrolidine-2,3-diones to prepare unprecedented β2,2,3-amino acids.

7.2 Introduction and Background

During our efforts directed at the total synthesis of the synoxazolidinones via an imine acylation/cyclization cascade (Chapter 5) we screened variously N-substituted imines (for example tBuSO-, DMB-) for reactivity with a number of phenyl pyruvic acid activated intermediates (acid chloride, anhydride, acyl imidazole, esters etc.) 7-6 (Scheme 7.1). Finally, acidic conditions involving acylation of preformed N-dimethoxybenzyl imines with phenylpyruvic acid chloride reproducibly gave satisfactory yields of 4-oxazolidinone products (Scheme 5.18). Interestingly, some coupling reagents tended to give mixtures of 4-oxazolidinone (O-addition product) and regioisomeric cyclization product dihydro-pyrrol-2-
one (C-addition product) upon activation of phenylpyruvic acid for the reaction with \( N \)-dimethoxybenzyl imines (Scheme 7.1). It should be noted that in all cases of our alternative acid promoted dehydration/cyclization approach (Chapter 4) the 4-oxazolidinone was the only significant product observed in the crude reaction mixtures and no trace of the C-addition regioisomer was identified.

**Scheme 7.1:** Potential equilibrium in addition pathways of \( N \)-acyliminium intermediates 7-1 and 7-2 leading to various ratios of regioisomeric cyclization (C- vs O-) products 7-4 and 7-3.

As we mentioned previously, a complete switch in the reaction selectivity was possible with some activating agents (Scheme 5.13). Most often, oxazolidinone to
dihydropyrrolone ratio varied (Scheme 7.1) depending on the type of activation employed and small amounts of 7-9 were formed when imine 7-7 was treated with phenylpyruvic acid (4-10) only.

Ryabukhin et al. reported the reaction of methyl esters of pyruvic acids with aromatic aldehydes and amines leading to combinatorial library of aryl substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones (Scheme 7.2-A).¹

Conditions involved addition of TMSCl or AcOH to a mixture of ester, aldehyde and amine in refluxing DMF. More than 100 substrates were obtained (11-93% yields). Another similar condensation of sodium ethyl oxaloacetate and various amines with mostly non-enolizable aldehydes was conducted in refluxing ethanol giving rise to esters in 18-77% yields (Scheme 7.2-B).² In a recent medicinal chemistry study on protein-protein interaction inhibitors, a library of variously substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones was synthesized employing an analogous multicomponent approach.³ These milder conditions employed esters which were added to preformed imines in dioxane at 0 °C; cyclization products were isolated in 5-60% yields after overnight stirring at room temperature (Scheme 7.2-C). A great diversity of substitution was accessed for dihydropyrrolones, although condensations of alkyl aldehydes suffered from low yields (Scheme 7.2-C, 7-17).
Scheme 7.2: Common condensation approaches to of 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones via condensation of oxo-esters.
Commonly found in nature, alkyl substituted dihydro-pyrrol-2-one possess a wide range of biological activities such as antimicrobial (7-11), antiviral, anti-tumor (7-10), nootropic and antineoplastic properties (Figure 7.1).

![7-10 tenuazonic acid anti-tumor, antiviral](image1)

![7-11 ikarugamycin antimicrobial](image2)

**Figure 7.1:** Examples of naturally occurring biologically active dihydropyrrolones.

7.3 Results and Discussion

7.3.1 Optimization of the Synthesis of Alkyl Substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones

Given the importance of dihydropyrrolones in medicinal chemistry, efficient access to alkyl analogues would be a useful extension for the diversification of these heterocycles. With some promising preliminary results for the synthesis of 7-8, we set out to optimize conditions and expand the substrate scope. We first observed formation of 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones during attempts to optimize the synthesis of 4-oxazolidinones via an imine acylation cascade. When cyanuric chloride was employed to activate phenyl pyruvic acid for the reaction with N-dimethoxybenzyl imines, various ratios of both cyclization products were isolated (Entries 4-7, Table 7.1). Upon changing solvents and exploring basic additives no obvious trend in cyclization preference could be observed. Yields were rather a reflection of the stability of the products after purification on the silica gel and never exceeded 50%
independently of the conditions we employed (equiv. of (CNCl)$_3$, work-up etc.).

**Table 7.1**: Formal [3+2] cycloaddition between pyruvates and imines.

<table>
<thead>
<tr>
<th>Entry</th>
<th>R</th>
<th>X</th>
<th>Conditions</th>
<th>Yield (%)</th>
<th>7-21</th>
<th>7-22</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>PhB(OH)$_2$ (5 mol%), DCM, rt</td>
<td>a, d</td>
<td></td>
<td>33</td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
<td>PhB(OH)$_2$ (20 mol%), DCM, rt</td>
<td>-</td>
<td>53</td>
<td>16</td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td>PhBO(CH$_2$)$_2$NH$_2$ (20 mol%), DCM, rt</td>
<td>-</td>
<td>56</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>-OH</td>
<td>(CNCl)$_3$, TEA, MeCN, 0 °C</td>
<td>(CNCl)$_3$, TEA, THF, 0 °C</td>
<td>7</td>
<td>40</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td></td>
<td>(CNCl)$_3$, TEA, THF, 0 °C</td>
<td>(CNCl)$_3$, TEA, DCM, 0 °C</td>
<td>3</td>
<td>26</td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
<td>(CNCl)$_3$, NaCl, DCM, 0 °C</td>
<td>(CNCl)$_3$, NaCl, DCM, 0 °C</td>
<td>48</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td></td>
<td>BNPPA (20 mol%), MeCN, rt</td>
<td>Cu(CF$_3$CO)$_2$, DCM, rt</td>
<td>-</td>
<td>23</td>
</tr>
<tr>
<td>7</td>
<td></td>
<td></td>
<td>Cu(CF$_3$CO)$_2$, DCM, rt</td>
<td>-</td>
<td>-</td>
<td>a, d</td>
</tr>
<tr>
<td>8</td>
<td></td>
<td>-C$_2$H$_5$</td>
<td>PyBOP, DIPEA, THF/DCM, 0 °C</td>
<td>-</td>
<td>24</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td></td>
<td></td>
<td>(COCI)$_2$, DMF, THF/DCM, 0 °C</td>
<td>-</td>
<td>17-40</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td></td>
<td>-OH</td>
<td>MeCN, rt</td>
<td>-</td>
<td>10</td>
<td>-</td>
</tr>
<tr>
<td>11</td>
<td></td>
<td>-OMe</td>
<td>BNPPA 20 mol%, MeCN, rt</td>
<td>-</td>
<td>10$^{a,d}$</td>
<td>-</td>
</tr>
<tr>
<td>12</td>
<td></td>
<td>-OMe</td>
<td>BNPPA 20 mol%, MeCN, rt</td>
<td>-</td>
<td>10$^{a,d}$</td>
<td>-</td>
</tr>
<tr>
<td>13</td>
<td>-C$_2$H$_5$</td>
<td>MeCN, rt</td>
<td>-</td>
<td>20$^{a,d}$</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>-OMe</td>
<td>MeCN, rt</td>
<td>-</td>
<td>25-60$^{a,d}$</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>-OMe</td>
<td>MeCN, 0 °C</td>
<td>-</td>
<td>83$^{b,f}$</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>16</td>
<td>-OMe</td>
<td>DCM, 0 °C</td>
<td>-</td>
<td>92$^{c,f}$</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>-OMe</td>
<td>MeCN, 0 °C</td>
<td>-</td>
<td>78$^{d,c}$</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>18</td>
<td>-OMe</td>
<td>MeCN, 0 °C</td>
<td>-</td>
<td>45$^{d,c}$</td>
<td>71$^{d,c}$</td>
<td></td>
</tr>
</tbody>
</table>

*a - after purification on silica gel; b- crystallization yield; c- yield of O-TBDMS protected pyrrolone; d- imine was preformed; e- excess of imine; f- imine formed in presence of 7-6; - - not formed.

Boronic esters and boronic acids were previously described to promote condensation reactions of phenyl pyruvic acids.$^5$ In our hands, up to 56% of simple 4-oxazolidinone could
be isolated employing (2-aminoethoxy)diphenylborane and (Entry 3, Table 7.1) low yields or mixtures were observed for C-addition product (Entries 1 and 2, Table 7.1).

Interestingly, when preformed imine was mixed with phenyl pyruvic acid only, a background reaction occurred providing up to 10% of C-addition product (Entry 12, Table 7.1). Addition of Lewis and Brønsted acids further elevated yield of 7-21 (Entries 8 and 9, Table 7.1) in the reaction of phenyl pyruvic acid, but not the its ester (Entry 13, Table 7.1).

Full conversion was observed when ester (7-27) was treated with preformed imine, but varied yield of product could be isolated (Entries 14 and 15, Table 7.1), again pointing to the instability of the product to SiO₂-column purification. Similarly, Guo et al. observed instability of N-tosyl 3-hydroxy-1,5-dihydro-2H-pyrrol-2-one derivatives to purification. Fortunately, some products precipitated (Entries 16, 18 and 19, Table 7.1) when the reaction was carried out in acetonitrile and therefore could be isolated in high yields. The best conditions involved sequential mixing of ester, 1.5 equiv. of aldehyde and 1.1 equiv. of 2,4-DMBA in MeCN at 0 °C.

In our hands protection of the 3-hydroxy group of dihydropyrrolone as TBDMS- or allyl ethers as well as carbonates led to high yields of stable products 7-60, 7-63 and 7-64. Similar to 4-oxazolidinones, removal of the DMB group was successfully achieved employing TFA in DCM, allowing for isolation of dihydropyrrolones 7-66 and 7-65 in high yields (Scheme 7.3).
Scheme 7.3: O-protection and N-deprotection of dihydropyrrolones.

The substrate scope was then further extended to other aliphatic aldehydes in addition to various aromatic and heteroaromatic aldehydes. We also demonstrated flexibility in the ester and amine substitution. When the two-step formation/O-alkylation procedure was performed in one pot, stable allyl ethers were isolated in good to high yields (Scheme 7.4).

Scheme 7.4: Substrate scope of O-allyl-1,5-dihydro-2H-pyrrol-2-ones.
In contrast to reported reaction protocols, (Scheme 7.4) our multi-component method provides significantly higher yields of alkyl substituted dihydropyrrolone derivatives, with electron-deficient esters proving the most reactive and yields for aromatic aldehydes slightly lowered. It should be noted that methyl esters with alkyl substituents failed to provide products 7-50, most likely due to very low abundance of the enol tautomer under standard reaction conditions (Scheme 7.5). Sterically hindered imines (of pivaldehyde and perfluoro butanal) also did not undergo cyclization to form the corresponding derivative of 7-50.

Scheme 7.5: Limitations of our methodology: 1,5-dihydro-2H-pyrrol-2-ones inaccessible via standard conditions (Table 7.1)

In summary, via this multi-component approach, three reagents are combined to produce a single C-addition product with structural components from the ester, amine and aldehyde building blocks, providing a high degree of diversity for target 1,5-dihydro-2H-pyrrol-2-one scaffolds (including enolizable aldehydes) while the number of synthetic operations is minimized. We envisioned utilizing O-alkylated 1,5-dihydro-2H-pyrrol-2-ones derived from enolizable aldehydes in the synthesis of the densely functionalized natural product pretazettine (7-51).
7.3.2 An Entry to Pretazettine via Functionalized 1,5-dihydro-2H-pyrrol-2-ones

*Amaryllidaceae* alkaloids exhibit potent anticancer activities, including cytostatic and inhibitory effects. Pretazettine is the most potent anti-cancer natural product in this family (Figure 7.2). The therapeutic potency of pretazettine has been demonstrated in animal models (mice) of Lewis lung carcinoma (LLC), a tumor known to be resistant to chemotherapy. Due to its superior biological activity pretazettine has received significant attention as a synthetic target.

![Figure 7.2](image)

**Figure 7.2:** Representative members of the crinine class of *Amaryllidaceae* alkaloids.

Pretazettine is the most complex representative of the crinine class of *Amaryllidaceae* alkaloids (Figure 7.2) recognized by the presence of *cis*-3a-arylhydroindole system. The first asymmetric approach to pretazettine (10 steps, \( \sim 22\% \) overall yield) was reported in 1998; just a year later, Baldwin and coworkers published total syntheses of optically pure haemanthidine, pretazettine (24 steps, \( \sim 3\% \) overall yield), and tazettine employing D-mannose as the starting material. It is known that haemanthidine, undergoes conversion to pretazettine under mild conditions and pretazettine can be further converted to tazettine in basic media (Scheme 7.6).

Despite the availability of many synthetic entries to pretazettine and related alkaloids
of *Amaryllidaceae* family, the development of more rapid approaches would enable diversification of the core of the arylhydroindole system. Based on our preliminary explorations of the reactivity of *O*-alkylated 1,5-dihydro-2*H*-pyrrol-2-ones we propose an approach, in which a reductive Heck reaction will be used to install the quaternary center and relative stereochemical configuration in the pyrrolidine ring of pretazettine’s core (7-55, Scheme 7.7).

![Scheme 7.6: Interconversion between 7-54, 7-51 and 7-52.](image)

Retrosynthetically, advanced intermediate 7-55 will be converted to pretazettine (7-51) upon ring-closing metathesis reaction followed by benzylic oxidation of the pyrane ring (Scheme 7.7). Key alkyl pyrrolone 7-56 will arise from a multi-component reaction between 7-57, 7-58 and 7-59 performed under conditions we have developed previously (Table 7.1).

![Scheme 7.7: Retrosynthetic analysis of our approach to pretazettine (7-51).](image)

To date we have demonstrated feasibility of the proposed key transformations in
model systems containing modifications around the pyrrolone core. We first demonstrated that a diene moiety as in 7-56 for a future ring-closing metathesis reaction can be incorporated. Ester 7-66 was successfully converted to 7-68 in 44% unoptimized yield (Scheme 7.8). Subsequently, the benzyl halide moiety, required for the Pd-catalyzed reductive Heck reaction, was installed in a model substrate 7-67 providing ether 7-69 in 94% yield.

Scheme 7.8: Synthesis of model substrates bearing key functional moieties required for synthesis of pretazettine.

The intramolecular Heck reaction\textsuperscript{12} is a powerful transformation that allows for coupling of an aryl or alkenyl halide with a nucleophile within the same molecule. The reaction is widely employed for the synthesis of carbocyclic and heterocyclic ring systems of various sizes. Mechanistically, the reaction proceeds via a typical palladium catalytic cycle: an aryl-Pd (II) species undergo addition to an alkene followed by β-hydride elimination (7-78) resulting in reduction of Pd(II) to Pd(0) and recycling of the catalyst (Table 7.2). In fact, the Heck reaction was already utilized by Overman and co-workers in probing the migratory insertion transition state during synthesis of pretazettine. A preference for an eclipsed
orientation of the Pd-C and alkene bonds was observed in the key intramolecular insertion step leading to the formation 6a-epipretazettine structure (7-86) of which was confirmed by X-ray analysis (Scheme 7.9).

![Scheme 7.9](image)

**Scheme 7.9:** Overman’s synthesis of 6a-epipretazettine via an intramolecular Heck reaction.

If no β-hydrogens are available to eliminate, the alkyl palladium species can participate in further tandem reactions (trapping by nucleophiles and migration to other double bonds), allowing for the rapid generation of the molecular complexity.13 β-Hydride elimination results in the turnover of the Pd-catalyst and can be also achieved by reducing the alkyl palladium intermediate. Heck couplings relying on external hydride sources to regenerate Pd(0) from Pd(II) are often related to as reductive Heck reactions.14 We envisioned the use of a reductive Heck reaction in the construction of the fused bicyclic core of pretazettine will provide the desired relative stereochemistry around pyrrolidine ring of 7-51, in contrast to previous methods to this scaffold.
With substrate 7-69 in hand we explored the utility of our proposed key reductive Heck transformation in the synthesis of model substrate 7-79 (Table 7.2). Reduction of a palladium enolate was performed by Gao et al. to access trans-hydroazulene core of englerin A. This Pd-enolate was accessed via reductive Heck transformation\textsuperscript{15} via employment of palladium acetate in the presence of sodium formate salt (hydride source) and quaternary ammonium salt in DMF at ambient temperature.

**Table 7.2:** Reductive Heck reaction of \textit{O}-benzyl-1,5-dihydro-2\textit{H}-pyrrol-2-one 7-69 for the synthesis of the model \{6.5\} ring system of pretazettine.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Conversion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>20 mol% PdCl\textsubscript{2}-2MeCN, HCOONa, (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, rt, 9 h</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>20 mol% Pd(OAc)\textsubscript{2}, HCOONa, (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, rt, 26 h</td>
<td>20</td>
</tr>
<tr>
<td>3</td>
<td>20 mol% Pd(OAc)\textsubscript{2}, HCOONa, (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, rt, \textasciitilde6 d</td>
<td>30</td>
</tr>
<tr>
<td>4</td>
<td>20 mol% Pd(OAc)\textsubscript{2}, HCOONa, (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, 65 °C, 11 h</td>
<td>50</td>
</tr>
<tr>
<td>5</td>
<td>20 mol% Pd(OAc)\textsubscript{2}, HCOONa (1.4 equiv.), (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, 65 °C, 14h</td>
<td>95</td>
</tr>
<tr>
<td>6</td>
<td>20 mol% Pd(OAc)\textsubscript{2}, HCOONa (3.2 equiv.), (C\textsubscript{7}H\textsubscript{13})\textsubscript{4}NCl, DMF, 65 °C, 14h</td>
<td>100</td>
</tr>
</tbody>
</table>

* - isolated yield; conversion was determined by integration of UV intensities of product and starting material.
We applied similar conditions to our substrate 7-69 which resulted in slow reaction with only ~30% conversion after 6 days (Entries 2 and 3, Table 7.2). Heating the reaction mixture and increasing the amount of sodium formate significantly accelerated the reaction and improved conversion (Entries 4 and 5, Table 7.2). Full conversion to the product with the desired molecular weight was finally achieved, but only 48% of this product was isolated on 19 mg reaction scale. Upon inspection of the $^1$H NMR spectrum of the isolated product we could not identify the proton at C3 of the pyrrolidone ring in 7-79, possibly due to obstruction of its singlet by other signals. The DMB group was removed to simplify the spectrum, but the desired signal was still absent, suggesting that arene reduction product 7-80 could form instead (Scheme 7.10) or 7-79 exists as tautomer 7-81.

Scheme 7.10: Possible products in our reductive Heck approach (7-79 desired, 7-80, 7-81 undesired).

Further investigations of the product structure, outcome of the reaction and screen for alternative conditions are ongoing in our lab.
7.3.3 Attempts at Asymmetric Synthesis of Alkyl Substituted 3-hydroxy-1,5-dihydro-2H-pyrrol-2-ones

Optically pure $N$-containing compounds, in particular heterocycles, have received significant research attention because of their frequent appearance in natural products. Additionally, possessing a wide-range of biological and pharmaceutical activities, $N$-containing heterocycles often serve as platforms for the discovery of new drug candidates. Among synthetic approaches to heterocycles, the use of multicomponent reactions (MCRs) is a particularly powerful tool to access diversity of substitution in relatively short time. Although MCRs have been widely utilized for synthesis of heterocycles, application of asymmetric approaches in multi-component reactions is still not fully explored. In particular, reactions that proceed via imine intermediates generated during the condensation of carbonyl compounds and amines lack approaches providing high levels of optical purity. The subsequent generation of water after the condensation reaction can for example disable some Lewis acid catalysts, resulting in low enantiomeric ratios of the product or stalled reaction. The challenge for such multi-component reactions is that, the chiral catalyst must be compatible with water. In addition, depending on the conditions, and sometimes even the order of addition of components, MCRs can undergo divergent pathways, as for example demonstrated by our oxazolidinone to dihydropyrrolone ratios (Table 7.1).

During optimization of the synthesis of alkyl containing 1,5-dihydro-2$H$-pyrrol-2-ones we noticed that yields of desired product were improved when imine formation took place in the presence of the ester. Noticeably, attempts to remove water (dehydrating reagents) during the reaction led to lower yields and other byproducts (including 4-
oxazolidinones). Another interesting aspect of the reactivity of enolizable aldehydes with benzyl amine in presence of an α-keto ester is that even at 0 °C, reaction proceeded quickly; and in presence of boronic acids (Entries, 1 – 3, Table 7.1) a significant exotherm was observed and the reaction was complete in less than 10 minutes. By performing NMR studies in CD$_3$CN and CDCl$_3$ we determined that the imine is rapidly formed and by itself is stable in the solution for several hours at room temperature and no side-reactions take place upon mixing the ester with aldehyde (Scheme 7.11). However, when amine is added to a solution of ester in absence of aldehyde formation of a byproduct can be observed. Therefore, to prevent cross condensation of the amine with ester, we added amine to a solution of both aldehyde and ester. We determined that slight excess of aldehyde was beneficial and ensured instant conversion of amine to aldimine which then reacted with ester to give 1,5-dihydro-2H-pyrrol-2-one. The net result of this MCR is formation of a C-C bond and an amide bond with the release of water and methanol under mild conditions (no catalysts or heating).

Scheme 7.11: Observed reactivity during condensation of 2,4-DMBA, enolizable aldehydes and phenyl pyruvic ester.

Condensation between the aldehyde and nucleophilic benzylamine proceeds with the formation of aldimine and release of an equivalent of water. When the resultant Schiff base undergoes reaction with the ester two possible pathways can be proposed to account for the
formation of $C$-addition product (Scheme 7.12). If the first step is the addition of the basic imine nitrogen to the carbonyl of the ester, then a short-lived tetrahedral intermediate can be assumed; loss of methoxide results in the formation of an $N$-acyliminium ion intermediate which in principle can give rise to both 4-oxazolidinone of dihydropyrrolone products (Scheme 7.1). Deprotonation of the enol by basic methoxide may explain the preference for $C$-addition under these conditions and the absence of pyrrolidinones in the acid mediated transformations we utilized for the synthesis of 4-oxazolidinones (Table 5.3). Additionally, the presence of the acid in reaction of the ester lowered the yield of pyrrolidinone products (Entry 13, Table 7.1).

Upon intramolecular C-C bond formation between $N$-acyliminium ion and the appended enol nucleophile cyclic, keto-amide and methanol are formed (Scheme 7.12-A). The presence of water and/or methanol most likely facilitates tautomerization to the corresponding 1,5-dihydro-2$H$-pyrrol-2-one. A chair-like transition state may account for stereochemistry of the cyclization and can be achieved via coordination to the enol or azomethine carbon of the iminium ion. Based on this mechanistic hypothesis, chiral bases or chiral cation binding catalysts may be beneficial in achieving enantioselectivity in the ring closure in this case.

Methyl esters are one of the least electrophilic carbonyl derivatives and are certainly less reactive than acid chlorides. Conversion of esters to amides usually requires extended reaction times and high temperatures associated with aminolysis, although catalytic methods have been developed.$^{16}$ If the first step in our process is intermolecular $C$-addition of the enol to an imine facilitated by hydrogen bonding in a chair-like six-membered transition state,
then amide bond forms in an intramolecular fashion. We believe that intramolecular amidation in this neutral pathway proceeds through a tetrahedral intermediate which after proton transfer rapidly loses methanol giving rise to the product (Scheme 7.12-A). Therefore, hydrogen bonding catalyst should have an impact on the enantiomeric distribution of the product if the reaction proceeds through this pathway.

**Scheme 7.12**: Proposed mechanisms for the formation 1,5-dihydro-2H-pyrrol-2-one.

Without significant proof for either of our proposed pathways leading to 1,5-dihydro-2H-pyrrol-2-one, we proceed to the exploration of various catalysts that could promote
stereoselectivity in the reaction and provide insights into the reaction mechanism. Previous work by Guo et al. describes preparation of N-tosyl aryl pyrrolones 7-26 via a catalytic asymmetric approach (Scheme 7.13), though electronic nature of the starting material 7-25 is significantly different from ours.\(^6\)

![Scheme 7.13: Asymmetric entry to N-tosyl aryl-containing pyrrolones 7-26.](image)

In this report quinuclidine based catalyst 7-23 is acting as chiral base to generate enolate of 7-24 which undergoes cycloaddition reaction with electron-deficient tosyl imines of aromatic aldehydes.

In order to obtain enantiomerically enriched products during a chemical transformation the source of chirality has to be proximal to the key bond formation. In theory, an element of chirality (axis, point or plane) can be installed on any of the reacting components: starting materials, catalyst or reaction media; in practice, the majority of efficient asymmetric reactions involve chiral catalysts or chiral substrates. Our efforts towards the asymmetric preparation of 1,5-dihydro-2H-pyrrol-2-one were mostly centered around chiral catalysts (Scheme 7.14) as well as introduction of chiral moieties on both ester (Scheme 7.15) and amine (Scheme 7.16).

Iron(II) chloride and copper(II) trifluoromethane sulfonate were explored as the Lewis acidic component in complexes with chiral bis-oxazoline ligand (7-28, 7-29).\(^{27}\) These
BOX-based Lewis acids catalyze reactions of substrates capable of two point binding (α-keto esters or amides, carbamates) in aldol (carbonyls and enolates) reactions as well as Mannich-type (iminiums and enols) reactions and aza-Nazarov cyclization (N-acyliminium ions).\(^\text{18}\) Unfortunately, no enantioselectivity was observed when these catalysts were used in model multicomponent reactions (Scheme 7.14), with incomplete conversion and byproducts formed.

Jacobsen and co-workers successfully employed thiourea-based catalysts in enantioselective addition of silyl ketene acetals to N-Boc aldimines to obtain enatio pure β-aryl-β-amino acids.\(^\text{19}\) Chiral urea-based catalysts (7-30, 7-32) did not affect stereochemistry of our cyclization and seemed to slow down the reaction. A different hydrogen bonding TADDOL-based catalyst can accelerate the cycloaddition reaction between aminosiloxo-containing dienes and dienophiles.\(^\text{20}\) Despite the full conversion of starting material, 5-43 was obtained in racemic form when 7-36 was employed as catalyst.

Organo-phosphoric acids can catalyze a range of asymmetric processes among which nucleophilic addition to imines proved to be the most controllable by BINOL-based catalysts.\(^\text{21}\) The bifunctional nature of the phosphoric acid (Lewis basic P=O and imine activating Brønsted acidic P-O-H) is well suited for the catalysis of imine-addition processes such as Streker (imines and nitrile) and aza-Henry reaction (imines and nitro-alkanes). In our hands chiral Vapol hydrogen phosphate 7-33 also failed to provide optically pure 5-43.
Scheme 7.14: Conditions and chiral reagents explored when attempting asymmetric synthesis of 5-43.

Johnston and coworkers previously demonstrated that Brønsted acid salt (1:1) of 7-38 was effective in the proton-catalyzed enantioselective aza-Henry reaction of Boc-aldimines
and nitro-alkanes. Although conversion to 5-43 in presence of this exact catalyst was slow, we did not see enantioselectivity in our multi-component process (Scheme 7.14). An efficient enantioselective Ag-catalyzed protocol for Mannich reaction of TMS-enol ethers and alkyl imines was reported by Hoveyda et al. After synthesizing phosphine ligand 7-35 and treating our in situ formed N-dimethoxybenzyl imine with the ester 7-27 in the presence of silver (I) acetate and 7-35 resulted in incomplete conversion and absence of enantioselectivity.

In search of alternative ways to impart stereoselectivity, we turned our attention to chiral solvents and ionic liquids. Aza-Baylis–Hillman reaction between activated alkenes and imines in chiral ionic liquid composed of methyltrioctylammonium dimalatoborate (7-39) was reported to yield β-amino ketones in up to 84% enantiomeric excess. Disappointingly, 5-43 did not form under these conditions. Attempts to remove water (3 Å MS, 4 Å MS, MgSO₄, Na₂SO₄) led to lower yields of racemic 1,5-dihydro-2H-pyrrol-2-one product due to formation of byproducts (including 4-oxazolidinone).

Chiral ester or amide (Scheme 7.15) auxiliaries appeared rather unstable (7-42, 7-45, or 7-46) or unreactive (7-40 and 7-41) and no asymmetric induction was observed for both 7-43 and 7-44. These results are possibly due to chiral group being displaced by the imine in the reaction prior to formation of chiral center.
Scheme 7.15: Chiral phenylpyruvic derivatives explored in attempts towards the asymmetric synthesis of 5-43.

The diastereoselective addition of various nucleophiles to the C-N double bond of chiral imines, hydrazones or sulfinimines is a convenient route to obtain optically pure nitrogen-containing compounds after removal of the chiral auxiliaries. For example, chiral imines derived from enantiopure 1-phenylalkyllamines are inexpensive, readily available chiral auxiliaries and have been widely utilized in organic synthesis, particularly in the construction of quaternary carbon stereocenters. When we employed chiral benzyl amines instead of 2,4-DMBA we observed significant reduction in reaction rate at 0 °C and low diastereomeric ratio (7-85 - 7-88, Scheme 7.16). Attempts to lower the temperature to achieve better diastereoselectivity yielded no product and reaction seemed to stall at temperatures below -20 °C. When chiral amino alcohols were employed (7-89 – 7-93) little to no product was formed. Interestingly, a 1:5 ratio of diastereomers of the product 7-48 formed when trans-aminocyclohexanol 7-97 was employed and diastereoselectivity was
lowered (1:2) when similar aromatic analogue 7-99 was used for the reaction. Furthermore, OTBS-protected amine 7-98 failed to provide 7-48 (Scheme 7.16).

![Chemical Reaction](image)

**Scheme 7.16:** Chiral amine derivatives explored in attempts at diastereoselective preparation of 7-48.

Altogether, these observations may suggest that steric crowding of the imine (derived from bulky secondary amine) is associated with the slow reaction. In addition, no reaction was observed with electron-deficient imines derived from 7-64 and 7-95, again pointing to the importance of the lone pair of Schiff-basic nitrogen in this reaction (Scheme 7.3).

As can be observed from our screening studies, the enantioselective MCRs remain a
challenge, in particular for the synthesis of 1,5-dihydro-2\(H\)-pyrrol-2-ones. Further exploration into reaction mechanism and modes of catalytic activation is undoubtedly required to access these medicinally relevant nitrogen-containing heterocycles in chiral fashion. The need for their asymmetric preparation is further signified by the important synthetic transformation of 1,5-dihydro-2\(H\)-pyrrol-2-ones discovered in our group (Pd-catalyzed alkylations, Claisen rearrangement). The reactivity and useful synthetic applications of these heterocycles are discussed next.

7.3.4 Synthesis of Novel 4-allyl-pyrrolidine-2,3-diones

7.3.4.1 Claisen Rearrangement of 3-(allyloxy)-1,5-dihydro-2\(H\)-pyrrol-2-ones

The Claisen rearrangement is a [3,3]-sigmatropic rearrangement of allyl vinyl ethers to \(\gamma,\delta\)-unsaturated carbonyl compounds under thermal conditions.\(^{46}\) The majority of Claisen rearrangements require high temperatures (>100 °C), but many examples of catalytic variants have been described.\(^{27}\) A six-membered transition state with delocalized electronic structure is the characteristic of any [3,3]-sigmatropic rearrangement. (Scheme 7.17). The Claisen rearrangement is frequently employed in organic synthesis, including the synthesis of terpene natural products.\(^{28}\)

Recognizing the allyl vinyl moiety in 3-(allyloxy)-1,5-dihydro-2\(H\)-pyrrol-2-ones (Scheme 7.4) we decided to probe them as substrates for Claisen rearrangement. When heated overnight in refluxing toluene 7-64b converted into novel 4-allyl-pyrrolidine-2,3-dione 7-71b (Scheme 7.17). Importantly, the reaction proceeded in a diastereospecific
fashion as only one diastereomer was obtained. The crystal structure was obtained for 7-71b confirming preference of the allyl to attack from the least hindered face, opposite to the substituent at C5 (cyclopropyl in 7-51).

Scheme 7.17: Claisen rearrangement of 3-(allyloxy)-1,5-dihydro-2H-pyrrol-2-ones.

HPLC-MS analysis of the reaction mixture at different time points revealed that rearrangement was slow and rather prolonged heating was required (12 - 24 hours) for full conversion (Scheme 7.18). Despite the high temperature and long reaction times no decomposition of the starting material 7-64 was observed for the majority of the substrates and 4-allyl-pyrrolidine-2,3-diones 7-71 formed cleanly and quantitatively. Other O-allyl-1,5-dihydro-2H-pyrrol-2-ones (Scheme 7.4) were successfully converted into the corresponding 4-allyl-pyrrolidine-2,3-diones demonstrating wide functional group tolerance of our method.
Scheme 7.18: Novel 4-allyl-pyrrolidine-2,3-diones via Claisen rearrangement of O-allyl-1,5-dihydro-2H-pyrrol-2-ones.
7.3.4.2 Tsuji-Trost Allylation of 3-(hydroxy)-1,5-dihydro-2H-pyrrol-2-ones

It is worth mentioning that during the Claisen rearrangement of two substrates 7-64j (Scheme 7.4) deep green coloration of the reaction mixture was observed and purification afforded varied yields of product (10 - 18%) which reflects significant decomposition. In addition, we also observed formation of 7-71j (up to 15%) as side product during synthesis of the corresponding parent allyl ether 7-64j employing potassium carbonate and excess of allyl bromide in acetone (0 °C - rt). Most likely C-alkylation 7-70j took place in these conditions. This observation prompted us to explore more rapid access to 7-71 via direct C-allylation of the enolates of 7-70. Substrate 7-70h was chosen as a model substrate to optimize reaction conditions due to the straightforward analysis of its 1H NMR spectrum.

The Tsuji-Trost reaction is the Pd-catalyzed allylation of nucleophiles, such enolates (Table 7.3) with allyl containing compounds (allyl acetate or allyl bromide). This transformation is very powerful for the generation of quaternary carbon stereocenters from simple synthetic precursors. The asymmetric version of this reaction (using chiral phosphine ligands) allows for enantioselective carbon-carbon, carbon-nitrogen, and carbon-oxygen bond formation under mild conditions, which makes it extremely important for the synthesis of complex molecules. Usually soft enolate nucleophiles will directly add to the Pd-allyl species at the carbon atom.
Table 7.3: C-alkylation of 1,5-dihydro-2H-pyrrol-2-ones via Tsuji-Trost reaction.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Conversion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5 mol% Pd, 10 mol% L, AllylOAc, PhMe, rt</td>
<td>NR</td>
</tr>
<tr>
<td>2</td>
<td>5 mol% Pd, 10 mol% L, tBuOK, AllylOAc, PhMe, rt</td>
<td>100</td>
</tr>
<tr>
<td>3</td>
<td>tBuOK, AllylOAc, PhMe, rt</td>
<td>NR</td>
</tr>
<tr>
<td>4</td>
<td>5 mol% Pd, 10 mol% L, tBuOK, AllylOAc, PhMe, 0 °C - rt</td>
<td>95*</td>
</tr>
</tbody>
</table>

*Pd: \([\text{Pd}_{\eta_3}C_3H_5CI]_2\); L: R(+)BINAP; * - isolated yield.

Standard conditions of Tsuji-Trost allylation (employing Pd-allyl dimer catalyst and BINAP as ligand) applied towards 7-70h rapidly provided 7-71h with full conversion observed in 30 minutes (Entry 2, Table 7.3). We found that allylation did not proceed if Pd-catalyst or base were omitted (Entries 1 and 3, Table 7.3). Cooling the reaction down to 0 °C cleanly provided desired product in 95% yield with the same relative stereochemistry observed for the Claisen rearrangement product (confirmed by crude NMR). Substrate 7-71j was also prepared according to this method in 93% yield and full conversion observed or alkyl containing substrates (7-64d).

We also demonstrated that the dimethoxybenzyl substituent can be removed to reveal the free nitrogen of novel pyrrolidinediones. Prolonged refluxing of the Claisen
rearrangement product 7-71h in TFA was required to achieve deprotection, and corresponding cyclic keto-amide product was isolated in 54% yield with incomplete conversion (62% BRSM) (Scheme 7.19).

![Scheme 7.19: Removal of DMB-protecting group.](image)

As we described previously, the DMB-protecting group could be cleaved more easily from precursor allyl ethers (Scheme 7.4). Treatment of the 7-64h with a 1:1 mixture of TFA and DCM at room temperature gave 7-66h within four hours in 70% yield after isolation from silica gel. Claisen rearrangement of 7-66h to 7-100 also proceeded with the formation of a single diastereomer.
7.3.5 Properties of Novel 4-allyl-pyrrolidine-2,3-diones

7.3.5.1 Synthesis of Novel $\beta^{2,2,3}$-Amino Acids Containing Quaternary Stereocenter

Amino acids serve as the building blocks for proteins and are critical for cell survival. They are also starting points for the synthesis of many important molecules (natural products, vitamins and nucleotides) in all living cells. Although less abundant than $\alpha$-amino acids, $\beta$-amino acids (Scheme 7.20) can be found in nature both in free and peptide bound forms. Naturally occurring compounds containing $\beta$-amino acid moieties (for example, cytotoxic activity is seen for the potent antitumor agent paclitaxel) are often highly biologically active. They were shown to possess antimicrobial and antifungal properties both in peptidic and $\beta$-lactam forms.\(^{31}\) The $\beta$-peptides (oligomers of $\beta$-amino acids) have been shown to form interesting secondary structures and superior proteolytic stability to proteases. Due to their stability to degradation by enzymes, $\beta$-amino acid containing peptides found extensive application in peptidic drugs.\(^{32}\) Substituted $\beta$-amino acids, such as $\beta^{2,2}$- amino acids (particularly $\alpha,\alpha$-dialkyl-) and $\beta^{2,2,3}$-amino acids (mainly $\alpha,\alpha,\beta$-trialkyl-), were shown to give rise to restricted conformations of $\beta$-peptides which has inspired some research groups to utilize them as synthetic biopolymers for the design of spatially defined nanoscopic patterns.\(^{33}\)
Scheme 7.20: Designation of amino acids.

Given the considerable biological potential of β-amino acids, a tremendous amount of effort has been dedicated to the development of their synthesis. A multitude of methods exist for the synthesis of monosubstituted at the β² or β³ positions β-amino acids; however, only few approaches describe the preparation of disubstituted β²,2- or β³,3-derivatives as this substitution pattern requires synthetically challenging formation of a quaternary stereocenter. In most cases, limitations in the synthesis of starting material lead to a lack of versatility in the achievable substitution pattern.

We envisioned utilizing 4-allyl-pyrrolidine-2,3-diones to prepare unprecedented β²,2,3-amino acids via oxidation-ring opening transformation (Scheme 7.21). This approach would allow installation of diverse aromatic substitutients at the all-carbon quaternary stereocenter. Current methods for the synthesis of β-amino acids with quaternary stereocenter are limited to alkyl substituents at this position.

Scheme 7.21: Our approach to novel β²,2,3-amino acids.
Relating structural similarity of 7-100 to isatins\textsuperscript{38} we proposed that oxidative basic conditions may be utilized for the conversion to 7-101.\textsuperscript{39} Isatins 7-102 can be converted to the corresponding anthranilic acids 7-103 by treatment with aqueous sodium hydroxide and 30\% hydrogen peroxide (Scheme 7.21). Regardless of the nature of the substitution of the aromatic ring, the reaction proceeds rapidly at room temperature (15 min), whereas isatins containing substituent on the nitrogen atom require slightly longer reaction times.\textsuperscript{40} After relatively few optimization efforts we were able to convert a set of representative 4-allylpyrrolidine-2,3-diones 7-104 into amino acids 7-105 in good yields employing similar conditions (Scheme 7.22).

![Scheme 7.22](image)

\textbf{Scheme 7.22}: Synthesis of novel $\beta^{2,2,3}$-amino acids via ring opening of 4-allylpyrrolidine-2,3-diones.

Going forward, our method proved very scalable and we were able to streamline the procedure and prepare gram quantities of $\beta^{2,2,3}$-amino acid 7-105a. Starting from three component condensation reaction of a methyl ester 7-27 hexanal and 2,4-dimethoxybenzylamine (DMBA) 1,5-dihydro-2H-pyrrol-2-one 5-43 was isolated by filtration (Scheme 7.23).
Scheme 7.23: Gram-scale synthesis of $\beta^{2,3}$-amino acid 7-105a.

One-pot allylation of 5-43 with allyl bromide followed by removal of dimethoxybenzyl substituent using TFA in DCM (1:1 v/v) furnished allyl vinyl ether 7-66 in 77% yield. 1.5 grams of 7-66 underwent a smooth Claisen rearrangement in refluxing toluene to give pure 4-allyl-pyrrolidine-2,3-dione 7-104a quantitatively. Product 7-104a was treated with 1M NaOH and 30% hydrogen peroxide to obtain $\beta$-amino acid 7-105a which could be isolated by filtration after acidic work up or easily purified by flash chromatography on a reversed phase silica column. We were able to obtain a crystal of 7-105a and confirm the structure by X-Ray analysis (Scheme 7.23).

7.3.5.2 Synthesis of Highly Substituted Polycyclic $N$-heterocycles

Containing Quaternary Stereocenter

The biological activity of a small molecule inherently depends on its three-dimensional structure. Usually, the more complex a molecule is, the more selective and specific are its interactions with a biological target. Structural diversity is one of the main
reasons why natural products and their core-scaffold analogues have higher hit rates in drug
discovery compared to libraries of synthetically derived/modified planar heterocycles in
typical HTS pharmaceutical studies. It has been demonstrated that the identification of
biologically active small molecules was more frequent when more structurally and
functionally diverse libraries of compounds were screened.\textsuperscript{41} 

Given this growing interest in non-planar and diverse heterocyclic scaffolds we also
explored ring modifications of the pyrrolidine-2,3-diones. The presence of a keto-amide
functionality in the ring of pyrrolidine-2,3-dione provides a straightforward entry for
nucleophiles for building structural complexity around their core. In addition, olefin
functionality of 4-allyl-pyrrolidine-2,3-diones can serve as a handle for further modifications
via Wittig and ring-closing metathesis reactions. For example, alkene containing derivative
\textbf{7-71a}, when treated with Grubbs 1\textsuperscript{st} generation catalyst undergoes ring-closing metathesis
with the formation of \textbf{7-75} possessing a [7.5] bicyclic system (Scheme 7.24).

\begin{center}
\includegraphics[width=\textwidth]{Scheme_7.24}
\end{center}

\textbf{Scheme 7.24}: Ring-closing metathesis reaction of 4-allyl-pyrrolidine-2,3-diones
for the synthesis of \textit{N}-containing polycyclic compounds.
We had previously planned to utilize a RCM reaction for the construction of similar fused [6.5] core of pretazettine; successful formation of 7-75 further supports our approach (Scheme 7.7).

Similarly, the reaction of 7-104a at the ketone carbonyl with allyl magnesium chloride provided a 1:1 mixture of diastereomers of 7-107 which after work up was treated with Grubbs ruthenium catalyst to provide 7-106 in 67% unoptimized yield. The diastereomers were readily separable via silica-gel chromatography.42

7.4 Conclusions and Outlook

A variety of interesting transformations (cycloadditions, reduction to amino alcohols etc.) can be envisioned to build in complexity around 4-allyl-pyrrolidine-2,3-diones. Explorations directed towards the discovery of other synthetically and biologically useful modes of reactivity of these heterocycles, as well as attempts to render their synthesis asymmetric are ongoing in our laboratory.
7.5 Experimental

**General Information**

THF was purified using an alumina filtration system. Aldehydes were purchased from a commercial chemical company and used as received. Reactions were monitored by TLC analysis (pre-coated silica gel 60 F254 plates, 500 µm layer thickness) and visualization was accomplished with a 254 nm UV light and by staining with a KMnO₄ solution (1.5 g of KMnO₄, 10 g of K₂CO₃, and 1.25 mL of a 10% NaOH solution in 200 mL of water). Reactions were also monitored by LC-MS (2.6 mm C18 50 x 2.10 mm column). Flash chromatography on SiO₂ was used to purify the crude reaction mixtures and performed on a flash system utilizing pre-packed cartridges and linear gradients. Melting points were determined using a capillary melting point apparatus. Infrared spectra were determined on a FT/IR spectrometer. ¹H and ¹³C NMR spectra were obtained on a 300 or 400 MHz instrument in CDCl₃ unless otherwise noted. Chemical shifts were reported in parts per million with the residual solvent peak used as an internal standard (CDCl₃ = 7.26 ppm for ¹H and 77.16 ppm for ¹³C). ¹H NMR spectra were run at 300 or 400 MHz and are tabulated as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, p = pentet, m = multiplet, bs = broad singlet, dt = doublet of triplet, tt = triplet of triplet), number of protons, and coupling constant(s). ¹³C NMR spectra were run at 100 MHz using a proton-decoupled pulse sequence with a d1 of 0 second unless otherwise noted, and are tabulated by observed peak. High-resolution mass spectra were obtained on an ion trap mass spectrometer using heated electrospray ionization (HESI). All aryl-2-oxopropanoic acids and their methyl esters were prepared according to general procedures previously described in literature.⁴³, ⁴⁴
General protocol for synthesis of methyl esters

![Chemical structure](image)

To a stirred under an inert atmosphere 0 °C solution of a corresponding arylpyruvic acid (1 equiv.) in dry DMF (0.2M) was added 1.01 equiv. of DBU and MeI (5.11 equiv.). Stirring at 0 °C continued for 2.5 h. Then poured into separatory funnel with 4:1 mixture of diethyl ether and ~0.4M HCl (prepared by dilution of 1 volume of 3M HCl with 6 volumes of deionized water). Organic layer was washed twice with water, once with saturated NaCl then dried over MgSO₄. Filtered, diethyl ether was concentrated in vacuo. Solid obtained, was used in the next step without further purification.

Methyl (Z)-2-hydroxy-3-phenylacrylate (7-27)⁴⁴.

Starting from phenylpyruvic acid (0.50 g, 2.98 mmol) following general protocol, 7-27 (0.48 g, 90%) was obtained as a white solid, spectral data closely matched with literature reported: $^1$H NMR (300 MHz, CDCl₃) δ 7.95 – 7.58 (m, 2H), 7.47 – 7.08 (m, 3H), 6.54 (s, 1H), 3.92 (s, 3H); $^{13}$C NMR (75 MHz, CDCl₃) δ = 166.9, 139.2, 134.2, 130.2, 130.0, 128.6, 128.2, 111.3, 53.4.
Methyl (Z)-2-hydroxy-3-(4-(trifluoromethyl)phenyl)acrylate (7-27e).

Starting from 2-oxo-3-(4-(trifluoromethyl)phenyl)propanoic acid (0.12 g, 0.51 mmol) following general protocol, 7-27e (0.11 g, 87%) was obtained as a yellow solid: \(^1\)H NMR (300 MHz, DMSO-\(d_6\)) \(\delta\) 10.06 (s, 1H), 7.97 (d, \(J = 8.1\) Hz, 2H), 7.70 (d, \(J = 8.4\) Hz, 2H), 6.49 (s, 2H), 3.82 (s, 2H); \(^13\)C NMR (75 MHz, DMSO-\(d_6\)) \(\delta\) 164.7, 143.2, 138.8, 129.7, 129.5, 126.9, 125.2, 125.1, 108.7, 52.8; IR (film): 3465, 3404, 1696, 1443, 1321, 1243, 1065 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{11}\)H\(_9\)F\(_3\)O\(_3\) [M-H] \(-245.0420\), found 245.0428. Mp = 109-110 °C.

Methyl (2Z,4E)-2-hydroxy-5-phenylpenta-2,4-dienoate (7-66).

Starting from acid 7-6j (100 mg, 0.51 mmol) following described above general protocol, 7-66 (92.9 mg, 88%) was obtained as an orange solid: \(^1\)H NMR (400 MHz, DMSO-\(d_6\)) \(\delta\) 9.32 (s, 1H), 7.90 – 6.98 (m, 7H), 6.78 (d, \(J = 16.0\) Hz, 1H), 6.36 (d, \(J = 11.3\) Hz, 1H), 3.76 (s, 3H); \(^13\)C NMR (101 MHz, DMSO-\(d_6\)) \(\delta\) 164.5, 141.3, 137.0, 133.4, 128.8, 127.9, 126.4, 126.3, 122.1, 113.1, 52.2; IR (film): 3478, 3412, 1688, 1442, 1383, 1240 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{12}\)H\(_{12}\)O\(_3\) [M+H]\(^+\) 205.0859, found 205.0855. Mp = 110-111 °C.
(2Z,4E)-2-hydroxy-5-phenylpenta-2,4-dienoic acid (7-6j).

A slurry of trans-cinnamaldehyde 1.99 g (11.3 mmol), sodium acetate (1.03 g, 12.5 mmol) and N-acetylglycine (1.47 g, 12.5 mmol) in acetic anhydride (10 mL) was heated at 110 °C for 2 h. After which heating was discontinued and reaction mixture allowed to reach room temperature, then placed on ice bath and cooled to 0-4 °C, filtered while cold and the solid washed with small amounts of ice-cold water. Resultant gummy solid was suspended in 50 ml of 3M HCl in a flask equipped with condenser and heated at reflux for 24 h. Reaction mixture was allowed to reach room temperature and further cooled to 0-4 °C, filtered while cold and solid washed with small amounts of ice-cold water. Dried in vacuum desiccator over KOH for 24h. 0.85 g of 7-6j as a light brown solid was obtained in 39% yield: $^1$H NMR (400 MHz, DMSO-d$_6$) δ 12.97 (s, 1H), 9.02 (s, 1H), 7.55 – 7.43 (m, 2H), 7.34 (t, $J$ = 7.6 Hz, 2H), 7.29 – 7.10 (m, 2H), 6.73 (d, $J$ = 15.9 Hz, 1H), 6.30 (d, $J$ = 11.3 Hz, 1H); $^{13}$C NMR (101 MHz, DMSO-d$_6$) δ 165.7, 142.1, 137.1, 132.7, 128.8, 127.8, 126.4, 122.5, 112.0; IR (film): 3362, 2937, 1687, 1508, 1209, 920 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{11}$H$_{10}$O$_3$ [M-H]$^-$ 189.0557, found 189.0556. Mp = 183-184 °C.
Methyl (Z)-2-hydroxy-4-oxo-4-phenylbut-2-enoate (7-27o).³

Following the literature procedure: 184 mg of sodium (8 mmol) was dissolved in 4 ml of dry methanol at 0 °C under an inert atmosphere to produce a 2.00 M solution of sodium methoxide. Dimethyl oxalate (0.59 g, 5.0 mmol) was added to this solution and reaction mixture was stirred for 15 min. 0.59 mL of acetophenone (5.0 mmol) was then added and stirring continued for additional 15 min. A microwave assisted reaction was then carried out for 5 min at 250 W, and 30 °C. After 5 min of the microwave reaction, precipitation was observed. The precipitate was collected by filtration and was washed with diethyl ether (15 ml). Resulting solid was dissolved in water (30 ml) and solution was acidified by addition of glacial AcOH (3 ml) to pH 3-4 at 0 °C. Precipitate formed was filtered and dried in vacuo overnight. 254.6 mg of 7-27o as a white creamy solid was obtained in 25% yield, spectral data closely matched with literature reported: ¹H NMR (300 MHz, CDCl₃) δ 8.12 – 7.93 (m, 2H), 7.62 (m, 2H), 7.55 – 7.44 (m, 2H), 7.10 (s, 2H), 3.95 (s, 3H).
General protocol for synthesis of 1,5-dihydro-2H-pyrrol-2-ones (I)

Solution of a corresponding methyl ester (1.0 equiv.) in acetonitrile (0.35 M) was placed under an inert atmosphere and cooled to 0 °C on ice bath. Aldehyde (2.05 equiv.) was added dropwise, followed by slow addition of 2,4-dimethoxybenzylamine (1.1 equiv.). Stirring continued until full consumption of starting material (~2 h). Solid formed was isolated by filtration, washed with small amount of acetonitrile and dried under high vacuum overnight.

In cases when product did not precipitate out of the solution, acetonitrile was removed in vacuo and residue either directly used in the next step (alkylation, dimethoxybenzyl- removal) or purified by column chromatography. Yield reduction after purification was attributed to instability of some substrates to silica gel due to presence of the unprotected enol- moiety, confirmed by resubjecting product to column chromatography.
According to general protocol I which employs ester 7-27 (1.41 g, 7.52 mmol), hexanal (1.93 ml, 15.4 mmol) and 2,4-dimethoxybenzylamine (1.27 ml, 8.27 mmol), 5-43 (2.48 g, 83%) was obtained as a white solid, spectral data closely matched with literature reported: Rf = 0.48 (40% EtOAc/hexanes); $^1$H NMR: (300 MHz, CDCl$_3$) $\delta$ 7.62 – 7.56 (m, 2H), 7.40 – 7.32 (m, 2H), 7.28 – 7.16 (m, 2H), 6.47 – 6.41 (m, 2H), 5.04 (d, $J$ = 15.0 Hz, 1H), 4.38 (t, $J$ = 3.5 Hz, 1H), 4.29 (d, $J$ = 15.0 Hz, 1H), 3.84 (s, 3H), 3.80 (s, 3H), 2.02 – 1.89 (m, 1H), 1.85 – 1.65 (m, 1H), 1.19 – 0.99 (m, 4H), 0.96 – 0.74 (m, 2H), 0.75 (t, $J$ = 7.0 Hz, 3H); mp = 140 – 141 ºC.

According to general protocol I which employs ester 7-27 (0.47 g, 2.51 mmol), isobutyaldehyde and 2,4-dimethoxybenzylamine (0.42 ml, 2.76 mmol) (0.48 ml, 5.14
mmol), 7-70c (0.78 g, 84%) was obtained as a white solid: $^1$H NMR (400 MHz, CDCl$_3$) δ 7.65 – 7.48 (m, 2H), 7.35 (m, 2H), 7.31 – 7.12 (m, 1H), 7.11 (m, 1H), 6.62 – 6.27 (m, 1H), 5.13 (d, $J$ = 15.3 Hz, 1H), 4.65 – 4.17 (m, 2H), 3.81 (s, 3H), 3.80 (s, 3H), 2.53 – 2.13 (m, 1H), 0.96 (d, $J$ = 7.1 Hz, 3H), 0.69 (d, $J$ = 7.0 Hz, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) δ 160.5, 158.4, 132.6, 130.6, 128.4, 128.2, 127.4, 117.6, 104.3, 98.6, 62.1, 55.5, 39.9, 29.8, 18.5, 17.0; IR (film): 3172, 2962, 1663, 1508, 1455, 1386, 1209, 1035 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{22}$H$_{25}$NO$_4$ [M+H]$^+$ 368.1856, found 368.1853. Mp = 190-191 °C; 5-cyclopropyl-1-(2,4-dimethoxybenzyl)-3-hydroxy-4-phenyl-1,5-dihydro-$2H$-pyrrol-2-one (70-7b).

According to general protocol I which employs ester 7-27 (1.15 g, 6.13 mmol), cyclopropanecarboxaldehyde (0.96 ml, 12.6 mmol) and 2,4-dimethoxybenzylamine (1.03 ml, 6.74 mmol), 70-7b (1.75 g, 78%) was obtained as a white solid: Rf = 0.28 (30% EtOAc/hexanes); $^1$H NMR (300 MHz, CDCl$_3$) δ 7.58 (d, $J$ = 7.8 Hz, 2H), 7.37 (d, $J$ = 7.3 Hz, 2H), 7.30 – 7.18 (m, 1H), 7.13 (d, $J$ = 8.1 Hz, 1H), 6.67 – 6.16 (m, 2H), 5.04 (d, $J$ = 15.4 Hz, 1H), 4.63 (d, $J$ = 15.4 Hz, 1H), 3.94 (d, $J$ = 8.6 Hz, 1H), 3.83 (s, 3H), 3.79 (s, 3H), 0.81 (m, 1H), 0.69 – 0.46 (m, 1H), 0.46 – 0.30 (m, 2H), 0.29 – 0.10 (m, 1H); $^{13}$C NMR (75 MHz, CDCl$_3$) δ 167.1, 160.4, 158.3, 142.3, 132.5, 130.3, 128.2, 128.2, 127.3, 122.3, 117.9, 104.4, 98.6, 60.5, 55.5, 38.8, 13.5, 4.2, 1.7; IR (film): 3174, 2938, 1663 1456, 1209, 1034, 697 cm$^{-1}$.
HRMS (ESI) m/z calcd for C_{22}H_{23}NO_4 [M+H]^+ 366.1699, found 366.1700. Mp = 193 – 194 °C.

1-(2,4-dimethoxybenzyl)-3-hydroxy-4,5-diphenyl-1,5-dihydro-2H-pyrrol-2-one (7-70h).

According to general protocol I which employs ester 7-27 (1.25 g, 6.66 mmol), benzaldehyde (1.36 ml, 13.3 mmol) and 2,4-dimethoxybenzylamine (1.12 ml, 7.33 mmol), 7-70h (1.89 g, 71%) was obtained as a white solid: \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 7.77 – 7.46 (m, 2H), 7.35 – 7.26 (m, 3H), 7.24 – 7.16 (m, 4H), 7.16 – 7.03 (m, 2H), 6.46 (m, 2H), 5.17 (s, 1H), 4.91 (d, \(J = 14.9\) Hz, 1H), 3.83 (d, \(J = 15.0\) Hz, 1H), 3.81 (s, 3H), 3.82 (s, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 167.3, 160.7, 158.6, 143.0, 136.5, 131.8, 131.3, 128.9, 128.6, 128.5, 128.3, 127.3, 127.2, 121.7, 117.4, 104.2, 98.6, 62.3, 55.5, 55.4, 39.0; IR (film): 3168, 2936, 1665, 1455, 1209, 1035, 702 cm\(^{-1}\); HRMS (ESI) m/z calcd for C\(_{25}\)H\(_{23}\)NO\(_4\) [M+H]^+ 402.1699, found 366.1689. Mp = 181-182 °C.
(E)-1-(2,4-dimethoxybenzyl)-3-hydroxy-5-pentyl-4-styrlyl-1,5-dihydro-2H-pyrrol-2-one (7-68).

According to general protocol I which employs 7-66 (129 mg, 0.60 mmol), hexanal (0.15 ml, 1.23 mmol) and 2,4-dimethoxybenzylamine (0.10 ml, 0.66 mmol), 7-68 (112 mg, 44%) was obtained as a white solid: $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.44 (d, $J$ = 7.1 Hz, 2H), 7.34 – 7.30 (m, 2H), 7.26 – 7.09 (m, 2H), 7.00 (d, $J$ = 16.5 Hz, 1H), 6.75 (d, $J$ = 16.5 Hz, 1H), 6.56 – 6.26 (m, 2H), 5.02 (d, $J$ = 14.7 Hz, 1H), 4.26 (d, $J$ = 15.0 Hz, 1H), 4.14 (br s, 1H), 3.84 (s, 3H), 3.80 (s, 3H), 2.08 – 1.89 (m, 1H), 1.91 – 1.70 (m, 1H), 1.31 – 1.08 (m, 4H), 1.02 – 0.90 (m, $J$ = 18.7 Hz, 2H), 0.83 (t, $J$ = 6.8 Hz, 3H); $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 167.4, 160.6, 158.4, 143.6, 137.5, 131.0, 130.5, 128.8, 127.8, 126.5, 121.5, 118.0, 117.6, 104.4, 98.6, 56.8, 55.6, 38.3, 31.9, 29.0, 22.7, 20.9, 14.2; IR (film): 3131, 2931, 1654, 1453, 1208, 1037, 692 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{26}$H$_{31}$NO$_4$ [M+H]$^+$ 422.2326, found 422.2317. Mp = 122 – 123 ºC.
General protocol for allylation of 1,5-dihydro-2H-pyrrol-2-ones (II)

To a solution of 1,5-dihydro-2H-pyrrol-2-ones (1 equiv.) in acetone (0.1M) at 0 °C was added to potassium carbonate (1.4 equiv.), followed by addition of allyl bromide (2.8 equiv.). Reaction was allowed to warm up to room temperature and stirred under an inert atmosphere until full consumption of starting material (10-18 h). Reaction mixture then was diluted with acetone, filtered and solvent evaporated in vacuo. Crude residue was purified by flash chromatography on SiO$_2$ (EtOAc/hexanes, 2 to 32%).

General protocol for two-step synthesis of 3-(allyloxy)-1,5-dihydro-2H-pyrrol-2-ones (III)

To a stirred under inert atmosphere solution of corresponding ester (1 equiv.) in DCM (0.025M) at 0 °C was added to aldehyde (1.6 equiv.), followed by slow addition of 0.025M DCM solution of 2,4-dimethoxybenzylamine (1.1 equiv.). Reaction was allowed to warm up to room temperature and stirred until full consumption of starting material (2-20 h). Then DCM was evaporated and crude oily residue kept under high vacuum for 30 min. Resultant
oil was dissolved in dry acetone (0.1M) under an inert atmosphere and potassium carbonate (2.8 equiv.) was added in one portion followed by addition of allyl bromide (5.6 equiv.). Stirring at room temperature continued until alkylation was complete (4-24 h). Reaction mixture then was diluted with acetone, filtered and solvent evaporated in vacuo. Crude residue purified by flash chromatography on SiO₂ (EtOAc/hexanes, 2 to 32%).

3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-pentyl-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-64).

According to general two-step protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and hexanal (25.0 µl, 0.19 mmol), 7-64 (37.4 mg, 70%) was obtained as a clear oil: ¹H NMR (400 MHz, CDCl₃) δ 7.61 – 7.46 (m, 2H), 7.40 – 7.31 (m, 2H), 7.33 – 7.21 (m, 1H), 7.18 (m, 1H), 6.46 (m, 1H), 6.12 – 5.80 (m, 1H), 5.47 – 5.26 (m, 1H), 5.26 – 5.14 (m, 1H), 5.07 (m, 1H), 5.02 – 4.83 (m, 2H), 4.35 (t, J = 3.6 Hz, 1H), 4.23 (d, J = 15.0 Hz, 1H), 3.83 (s, 3H), 3.80 (s, 3H), 2.05 – 1.80 (m, 1H), 1.60 m, 1H), 1.16 – 0.95 (m, 4H), 0.86 (m, 3H), 0.74 (t, J = 7.1 Hz, 3H); ¹³C NMR (101 MHz, CDCl₃) δ 167.0, 160.5, 158.4, 144.6, 134.1, 131.6, 131.1, 131.0, 128.5, 128.1, 127.8, 118.0, 118.0, 104.4, 98.5, 71.3, 56.4, 55.5, 37.8, 31.6, 28.5, 22.5, 20.7, 14.0; IR (film): 2933, 2859, 1686, 1508, 1209, 1035 764 cm⁻¹; HRMS (ESI) m/z calcd for C₂₇H₃₃NO₄ [M+H]^+ 436.2482, found 436.2485.
Alternatively, 7-64 (203 mg, 0.47 mmol) was obtained in 96% yield after allylation of 5-43 (195 mg, 0.49 mmol) following general procedure II

\[
(Z)-3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-(hex-3-en-1-yl)-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-64a).
\]

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and cis-4-heptanal (27.2 µl, 0.19 mmol), 7-64a (35.4 mg, 64%) was obtained as a slightly yellow oil: Rf = 0.66 (20% EtOAc/hexanes); \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 7.52 (m, 2H), 7.39 – 7.33 (m, 2H), 7.33 – 7.25 (m, 1H), 7.20 (m, 1H), 6.52 – 6.37 (m, 2H), 6.18 – 5.71 (m, 1H), 5.35 (m, 1H), 5.29 – 5.15 (m, 2H), 5.13 – 4.81 (m, 4H), 4.47 – 4.32 (m, 1H), 4.25 (d, \(J = 15.0\) Hz, 1H), 3.83 (s, 3H), 3.80 (s, 3H), 2.17 – 1.87 (m, 1H), 1.86 – 1.46 (m, 5H), 0.79 (t, \(J = 7.5\) Hz, 3H); \(^13\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 167.0, 160.5, 158.4, 144.6, 134.1, 132.5, 131.5, 131.0, 130.9, 128.5, 128.2, 127.8, 127.7, 118.0, 117.9, 104.5, 98.6, 71.3, 56.2, 55.5, 37.9, 28.7, 20.3, 19.1, 14.3; IR (film): 3004, 2960, 1686, 1508, 1134, 1055, 751, 697 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{28}\)H\(_{33}\)NO\(_4\) [M+H]\(^+\) 448.2482, found 448.2489.
3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-isopropyl-4-phenyl-1,5-dihydro-2\textit{H}-pyrrol-2-one (7-64c).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and isobutyraldehyde (18.4 µl, 0.19 mmol), 7-64c (36.5 mg, 73%) was obtained as a slightly yellow oil: Rf = 0.35 (20% EtOAc/hexanes); \textsuperscript{1}H NMR (400 MHz, CDCl\textsubscript{3}) \(\delta\) 7.43 (m, 2H), 7.39 – 7.21 (m, 3H), 7.13 (d, \(J = 7.9\) Hz, 1H), 6.58 – 6.25 (m, 2H), 6.18 – 5.78 (m, 1H), 5.32 (m, 1H), 5.18 (m, 1H), 5.13 – 4.96 (m, 2H), 4.88 (m, 1H), 4.28 (d, \(J = 15.3\) Hz, 1H), 4.25 (d, \(J = 2.2\) Hz, 1H), 3.81 (s, 3H), 3.80 (s, 3H), 2.21 (ddd, \(J = 14.1, 7.1, 2.2\) Hz, 1H), 0.89 (d, \(J = 7.1\) Hz, 3H), 0.63 (d, \(J = 7.0\) Hz, 3H); \textsuperscript{13}C NMR (101 MHz, CDCl\textsubscript{3}) \(\delta\) 167.5, 160.4, 158.4, 144.6, 134.1, 132.4, 132.3, 130.6, 128.5, 128.3, 128.1, 118.0, 104.3, 98.5, 71.3, 61.8, 55.5, 39.2, 29.6, 17.9, 17.2; IR (film): 2960, 2837, 1686, 1509, 1209, 1035, 764 cm\textsuperscript{-1}; HRMS (ESI) \(m/z\) calcd for C\textsubscript{25}H\textsubscript{29}NO\textsubscript{4} [M+H]\textsuperscript{+} 408.2169, found 408.2171.
According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and cyclopropanecarboxaldehyde (15.1 µl, 0.19 mmol), 7-64b (29.5 mg, 59%) was obtained as a slightly yellow oil: Rf = 0.66 (20% EtOAc/hexanes); $^1$H NMR (300 MHz, CDCl$_3$) δ 7.58 – 7.41 (m, 2H), 7.40 – 7.18 (m, 3H), 7.12 (d, $J$ = 7.9 Hz, 1H), 6.55 – 6.30 (m, 2H), 5.97 (m, 1H), 5.30 (m, 1H), 5.21 – 5.09 (m, 1H), 5.06 – 4.71 (m, 3H), 4.60 (d, $J$ = 15.5 Hz, 1H), 3.83 (s, 3H), 3.80 (s, 3H), 3.82 (d, 15.4 Hz, 1H), 0.73 – 0.59 (m, 1H), 0.59 – 0.41 (m, 1H), 0.31 (m, 2H), 0.13 – 0.02 (m, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) δ 166.5, 160.3, 158.2, 144.2, 134.1, 132.3, 132.1, 130.2, 128.7, 128.2, 128.0, 118.4, 117.9, 104.3, 98.5, 71.4, 61.0, 61.0, 55.5, 55.4, 38.3, 13.7, 3.7, 1.6; IR (film): 3082, 2936, 1686, 1208, 1034, 764 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{25}$H$_{27}$NO$_4$ [M+H]$^+$ 406.2013, found 406.2012.

Alternatively, 7-64b (258 mg, 0.58 mmol) was obtained in 82% yield after allylation of 7-70b (260 mg, 0.71 mmol) following general procedure II.
3-(allyloxy)-5-(((tert-butyldimethylsilyl)oxy)methyl)-1-(2,4-dimethoxybenzyl)-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-64k).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and (tert-butyldimethylsilyloxy)acetaldehyde (41.8 µl, 0.19 mmol), 7-64k (29.5 mg, 47%) was obtained as a clear oil: Rf = 0.74 (20% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.62 – 7.47 (m, 2H), 7.42 – 7.30 (m, 3H), 7.29 – 7.21 (m, 1H), 7.16 (d, \(J = 8.4\) Hz, 1H), 6.45 (m, 2H), 6.18 – 5.88 (m, 1H), 5.35 (m, 1H), 5.19 (m, 1H), 5.07 – 4.87 (m, 2H), 4.44 (d, \(J = 15.4\) Hz, 1H), 4.30 (t, \(J = 3.2\) Hz, 1H), 3.97 (dd, \(J = 10.7, 2.7\) Hz, 1H), 3.82 (s, 3H), 3.80(s, 3H), 3.60 (dd, \(J = 10.7, 3.8\) Hz, 1H), 0.78 (s, 9H), -0.15 (s, 3H), -0.22 (s, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 167.2, 160.3, 158.3, 145.3, 134.2, 131.6, 130.5, 128.5, 128.3, 128.0, 127.8, 118.2, 117.7, 104.3, 98.5, 71.3, 61.6, 59.1, 55.5, 38.8, 25.8, 18.2, -5.7; IR (film): 2933, 2855, 1688, 1508, 1209, 1035, 837, 695 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for \(C_{29}H_{39}NO_5Si\) [M+H]\(^+\) 510.2670, found 510.2667.
3-(allyloxy)-1-(2,4-dimethoxybenzyl)-4,5-diphenyl-1,5-dihydro-2H-pyrrol-2-one (7-64h).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and benzaldehyde (20.2 µl, 0.19 mmol), 7-64h (31.8 mg, 58%) was obtained as a clear oil: Rf = 0.6 (15% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.55 – 7.43 (m, 2H), 7.33 – 7.23 (m, 3H), 7.23 – 7.12 (m, 4H), 7.11 – 7.06 (m, 1H), 6.46 – 6.43 (m, 2H), 6.22 – 5.96 (m, 1H), 5.49 – 5.35 (m, 1H), 5.29 – 5.23 (m, 1H), 5.22 – 5.16 (m, 1H), , 5.14 (s, 1H), 5.12 – 5.01 (m, 1H), 4.85 (d, \(J = 14.9\) Hz, 1H), 3.81 (s, 3H), 3.75 (s, 3H), 3.75 (d, \(J = 14.9\) Hz, 1H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 166.5, 160.6, 158.5, 145.0, 136.6, 134.1, 131.5, 131.3, 131.1, 128.8, 128.5, 128.3, 128.0, 127.7, 118.2, 117.8, 104.2, 98.5, 71.4, 61.9, 55.5, 55.4, 38.5; IR (film): 2934, 1687, 1507, 1209, 1034, 760, 694 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{28}\)H\(_{27}\)NO\(_4\) [M+H]\(^+\) 442.2012, found 442.2014.

Alternatively, 7-64h (248 mg, 0.56 mmol) was obtained in 79% yield after allylation of 7-70h (285 mg, 0.71 mmol) following general procedure II. It is worth mentioning that 7-71h (52 mg, 0.12 mmol) also formed in 17% yield.
3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-(4-methoxyphenyl)-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-64n).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and p-anisaldehyde (24.2 µl, 0.19 mmol), 7-64n (26 mg, 45%) was obtained as a clear oil: Rf = 0.48 (20% EtOAc/hexanes); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.59 – 7.41 (m, 2H), 7.25 – 7.04 (m, 5H), 6.91 – 6.71 (m, 2H), 6.55 – 6.35 (m, 2H), 6.23 – 5.91 (m, 1H), 5.50 – 5.35 (m, 1H), 5.29 – 5.23 (m, 1H), 5.22 – 5.15 (m, 1H), 5.11 (s, 1H), 5.10 – 5.01 (m, 1H), 4.84 (d, $J = 14.9$ Hz, 1H), 3.81 (s, 3H), 3.79 (s, 3H), 3.77 (d, $J = 14.9$ Hz, 1H), 3.76 (s, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 166.4, 160.6, 159.6, 158.5, 145.0, 134.2, 131.6, 131.2, 131.1, 129.5, 128.4, 128.3, 127.9, 127.8, 118.1, 118.0, 114.2, 104.3, 98.6, 71.4, 61.4, 55.5, 55.4, 55.3, 38.4; IR (film): 2935, 2835, 1687, 1611, 1209, 1033, 834, 693 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{29}$H$_{29}$NO$_5$ [M+H]$^+$ 472.2119, found 472.2119.
According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and 4-nitrobenzaldehyde (30.2 mg, 0.19 mmol), 7-64l (36.4 mg, 61%) was obtained as a yellow oil: Rf = 0.68 (30% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.27 – 7.94 (m, 2H), 7.49 – 7.40 (m, 2H), 7.39 – 7.28 (m, 2H), 7.23 – 7.13 (m, 3H), 7.08 (d, $J$ = 8.2 Hz, 1H), 6.57 – 6.23 (m, 2H), 6.23 – 5.85 (m, 1H), 5.45 – 5.38 (m, 1H), 5.31 – 5.25 (m, 1H), 5.24 (br s, 1H), 5.23 – 5.17 (m, 1H), 5.12 – 5.05 (m, 1H), 4.86 (d, $J$ = 14.9 Hz, 1H), 3.80 (d, $J$ = 14.9 Hz, 2H), 3.80 (s, 1H), 3.76 (s, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 166.4, 160.9, 158.4, 147.9, 145.2, 144.5, 133.9, 131.5, 130.8, 130.3, 129.1, 128.5, 128.4, 127.6, 124.0, 118.4, 117.1, 104.5, 98.5, 71.4, 61.0, 55.5, 55.4, 38.7; IR (film): 2939, 1692, 1522, 1348, 1209, 1035, 827, 694 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{28}$H$_{26}$N$_2$O$_6$ [M-H]$^-$ 485.1718, found 485.1711.
3-(allyloxy)-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(4-(trifluoromethyl)phenyl)-1,5-
dihydro-2H-pyrrol-2-one (7-64m).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and 4-(trifluoromethyl)benzaldehyde (27.5 µl, 0.19 mmol), 7-64m (40.3 mg, 64%) was obtained as a clear oil: Rf = 0.76 (30% EtOAc/hexanes); \( ^1 \)H NMR (400 MHz, CDCl\(_3\)) \( \delta \) 7.59 – 7.43 (m, 4H), 7.35 – 7.12 (m, 5H), 7.09 (d, \( J = 8.2 \) Hz, 1H), 6.52 – 6.36 (m, 1H), 6.19 – 6.02 (m, 1H), 5.53 – 5.34 (m, 1H), 5.30 – 5.22 (m, 1H), 5.20 (br s, 1H), 5.16 – 5.03 (m, 1H), 4.85 (d, \( J = 14.9 \) Hz, 1H), 3.80 (s, 3H), 3.80 (d, \( J = 15.0 \) Hz, 1H), 3.76 (s, 3H); \( ^{13} \)C NMR (101 MHz, CDCl\(_3\)) \( \delta \) 166.4, 160.8, 158.4, 145.1, 141.0, 134.0, 131.4, 131.0, 130.6, 128.6, 128.4, 128.2, 127.6, 125.8, 125.7, 118.4, 117.3, 104.4, 98.5, 71.4, 61.4, 55.5, 55.4, 38.6; IR (film): 2939, 1692, 1508, 1325, 1209, 763 cm\(^{-1}\); HRMS (ESI) \( m/z \) calcd for C\(_{29}\)H\(_{26}\)F\(_3\)NO\(_4\) [M+H]\(^+\) 510.1887, found 510.1894.
According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and nicotinaldehyde (18.9 µl, 0.19 mmol), 7-64j (28.2 mg, 52%) was obtained as a yellow oil: Rf = 0.31 (30% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 8.61 – 8.28 (m, 2H), 7.55 – 7.44 (m, 1H), 7.40 – 7.37 (m, 1H), 7.24 – 7.07 (m, 5H), 6.60 – 6.33 (m, 5H), 6.07 (m, 1H), 5.46 – 5.36 (m, 1H), 5.29 – 5.25 (m, 1H), 5.24 – 5.19. (m, 1H), 5.18 (br s, 1H), 5.10 – 5.05 (m, 1H), 4.84 (d, \(J = 14.8\) Hz, 1H), 3.82 (d, \(J = 15.4\) Hz, 1H), 3.80 (s, 3H), 3.78 (s, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 166.4, 160.8, 158.4, 150.5, 150.0, 145.2, 134.9, 134.0, 132.6, 131.5, 130.9, 130.4, 128.5, 128.3, 127.6, 124.0, 118.4, 117.3, 104.4, 98.5, 71.4, 59.3, 55.5, 55.4, 38.5; IR (film): 2937, 1689, 1508, 1209, 1034, 763 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{27}\)H\(_{26}\)N\(_2\)O\(_4\) [M+H]\(^+\) 443.1965, found 443.1969.

3-(allyloxy)-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(pyridin-3-yl)-1,5-dihydro-2\(H\)-pyrrol-2-one (7-64f).
3-(allyloxy)-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(thiazol-4-yl)-1,5-dihydro-2H-pyrrol-2-one (7-64j).

According to general protocol III which employs ester 7-27 (22.0 mg, 0.12 mmol) and thiazole-4-carboxaldehyde (22.7 mg, 0.19 mmol), 7-64j (34.9 mg, 63%) was obtained as a yellow oil: Rf = 0.42 (20% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.70 (d, $J = 2.0$ Hz, 1H), 7.86 – 7.49 (m, 2H), 7.35 – 6.85 (m, 5H), 6.59 – 6.29 (m, 2H), 6.13 – 6.03 (m, 1H), 5.58 (s, 1H), 5.43 – 5.38 (m, 1H), 5.30 – 5.21 (m, 1H), 5.20 – 5.08 (m, 2H), 4.89 (d, $J = 15.0$ Hz, 1H), 3.92 (d, $J = 15.0$ Hz, 1H), 3.79 (s, 3H), 3.77 (s, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 166.4, 160.5, 158.5, 153.6, 153.0, 145.0, 134.1, 131.3, 131.0, 129.5, 128.3, 128.0, 127.6, 118.0, 117.6, 117.3, 104.2, 98.5, 71.3, 57.7, 55.5, 38.9; IR (film): 3083, 2936, 1687, 1508, 1209, 1035, 763 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{25}$H$_{24}$N$_{2}$O$_4$S [M+H]$^+$ 449.1529, found 449.1531.
(E)-3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-pentyl-4-styryl-1,5-dihydro-2H-pyrrol-2-one (7-64o).

According to general protocol III which employs 7-66 (52.0 mg, 0.23 mmol) and hexanal (51 µl, 0.42 mmol), 7-64o (62.8 mg, 59%) was obtained as a yellow oil: Rf = 0.54 (30% EtOAc/hexanes); 1H NMR (300 MHz, CDCl₃) δ 7.52 – 7.40 (m, 2H), 7.36 – 7.31 (m, 2H), 7.30 – 7.20 (m, 1H), 7.16 (d, J = 9.1 Hz, 1H), 6.98 (d, J = 17.3 Hz, 1H), 6.68 (d, J = 16.7 Hz, 0H), 6.53 – 6.38 (m, 2H), 6.12 – 5.97 (m, 1H), 5.44 – 5.35(m, 1H), 5.29 – 5.19 (m, 1H), 5.15 – 4.87 (m, 3H), 4.19 (d, J = 15.1 Hz, 1H), 4.13 (t, J = 3.4 Hz, 1H), 3.83 (s, 3H), 3.80 (s, 3H), 1.96 (s, 1H), 1.79 (s, 1H), 1.37 – 1.05 (m, 4H), 1.01 – 0.88 (m, 2H), 0.82 (t, J = 6.9 Hz, 3H); 13C NMR (101 MHz, CDCl₃) δ 166.6, 160.6, 158.5, 145.6, 137.1, 134.2, 131.3, 131.0, 130.5, 128.8, 128.2, 126.7, 118.2, 118.0, 117.8, 104.5, 98.6, 71.4, 55.9, 55.6, 55.5, 37.7, 31.9, 29.1, 22.7, 20.9, 14.1; IR (film): 2932, 2859, 1683, 1611, 1508, 1209, 1034, 692 cm⁻¹; HRMS (ESI) m/z calcd for C₂₉H₃₅NO₄ [M+H]⁺ 462.2639, found 462.2634.

7-71j (10.4 mg, 0.023 mmol) also formed in 10% yield.

Alternatively, 7-64j (43.2 mg, 0.093 mmol) was obtained in 54% yield after allylation of 7-70j (73.1 mg, 0.17 mmol) according to general procedure II which employs 1.3 equiv. of potassium carbonate (30 mg, 0.22) and 2.5 equiv. of allyl bromide (38 µL, 0.43 mmol). It
is worth mentioning that 7-71j (12.4 mg, 0.027 mmol) also formed in 15% yield.

\[ \text{3-(allyloxy)-1-(2,4-dimethoxybenzyl)-5-pentyl-4-(4-(trifluoromethyl)phenyl)-1,5-dihydro-2H-pyrrol-2-one (7-64e).} \]

According to general protocol III which employs ester 2 (32.1 mg, 0.13 mmol) and hexanal (26.2 µl, 0.21 mmol), 7-64e (43.8 mg, 67%) was obtained as a yellow oil: Rf = 0.5 (20% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta 7.68 – 7.56 (m, 4H), 7.23 – 7.16 (m, 1H), 6.59 – 6.33 (m, 2H), 6.07 – 5.93 (m, 1H), 5.36 (d, \(J = 17.2\), 1H), 5.29 – 5.07 (m, 2H), 5.09 – 4.90 (m, 2H), 4.47 (t, \(J = 3.4\) Hz, 1H), 4.23 (d, \(J = 15.0\) Hz, 1H), 3.84 (s, 3H), 3.80 (s, 3H), 2.01 – 1.82 (m, 1H), 1.64 – 1.50 (m, 1H), 1.21 – 0.83 (m, 6H), 0.75 (t, \(J = 7.1\) Hz, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta 166.4, 160.7, 158.5, 146.0, 135.2, 133.9, 131.1, 128.7, 127.8, 125.5, 125.4, 118.3, 117.8, 104.6, 98.7, 77.5, 77.2, 76.8, 71.3, 56.3, 55.6, 55.5, 38.0, 31.6, 28.4, 22.5, 20.8, 14.0; IR (film): 2935, 2861, 1688, 1614, 1508, 1326, 1122, 836 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{28}\)H\(_{32}\)F\(_3\)NO\(_4\) [M+H]\(^+\) 504.2356, found 504.2353.
3-(allyloxy)-4-benzoyl-1-(2,4-dimethoxybenzyl)-5-pentyl-1,5-dihydro-2H-pyrrol-2-one (7-64o).

According to general protocol III which employs ester 7-27o (27.0 mg, 0.13 mmol) and hexanal (26.2 µl, 0.21 mmol), 7-64o (49.8 mg, 82%) was obtained as a yellow oil: Rf = 0.28 (15% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.90 – 7.69 (m, 2H), 7.66 – 7.49 (m, 1H), 7.51 – 7.36 (m, 2H), 7.24 – 7.12 (m, 2H), 6.47 – 6.42 (m, 2H), 5.80 – 5.61 (m, 1H), 5.09 – 4.80 (m, 5H), 4.52 – 4.37 (m, 1H), 4.21 (d, $J$ = 14.8 Hz, 1H), 3.81 (s, 3H), 3.79 (s, 3H), 2.18 – 1.56 (m, 1H), 1.34 – 1.07 (m, 4H), 1.07 – 0.94 (m, 2H), 0.80 (t, $J$ = 7.0 Hz, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 191.1, 165.3, 160.8, 158.4, 150.6, 138.2, 136.3, 133.0, 132.8, 131.3, 129.4, 128.3, 126.4, 117.9, 117.3, 104.5, 98.5, 71.5, 57.0, 55.5, 38.3, 31.8, 27.8, 22.6, 21.8, 14; IR (film): 2932, 2860, 1693, 1615, 1508, 1209, 1036, 833 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{28}$H$_{33}$NO$_5$ [M+H]$^+$ 464.2432, found 464.2427.
1-allyl-3-(allyloxy)-5-pentyl-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-64p).

According to general protocol III which employs ester 7-27 (44.0 mg, 0.25 mmol), hexanal (50.0 µl, 0.39 mmol) and allyl amine (21.0 µl, 0.28 mmol), 7-64p (46.1 mg, 57%) was obtained as a clear oil: Rf = 0.5 (20% EtOAc/hexanes); \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 7.59 – 7.54 (m, \(J = 8.4, 1.3\) Hz, 2H), 7.51 – 7.17 (m, 3H), 6.03 – 5.93 (m, 1H), 5.89 – 5.75 (m, 1H), 5.37 – 5.27 (m, 1H), 5.26 – 5.15 (m, 3), 5.09 – 4.98 (m, 1H), 4.95 – 4.84 (m, 1H), 4.71 – 4.47 (m, 2H), 3.58 (dd, \(J = 15.6, 7.5\) Hz, 1H), 1.91 – 1.73 (m, 1H), 1.72 – 1.58 (m, 1H), 1.17 – 0.99 (m, 4H), 0.95 – 0.77 (m, 2H), 0.74 (t, \(J = 6.9\) Hz, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 166.7, 144.6, 134.1, 133.5, 131.5, 131.3, 128.6, 128.3, 127.8, 118.0, 71.3, 56.5, 42.9, 31.6, 28.7, 22.5, 20.8, 14.0; IR (film): 3083, 2931, 2860, 1688, 988, 925, 765, 731 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{21}\)H\(_{27}\)NO\(_2\) [M+H]\(^+\) 326.2115, found 326.2114.

Synthesis of 4-allyl-pyrrolidine-2,3-diones via Pd(II) mediated allylation of 1,5-dihydro-2H-pyrrol-2-ones (IV)

Solution of dihydropyrrolone (1 equiv.) in degassed toluene (0.1M) was placed under an inert atmosphere on ice bath and cooled to 0 °C. Potassium tret-butoxide (1 equiv.) was added to this solution and stirring maintained, meanwhile in a separate vessel 5 mol% of
allylpalladium (II) chloride dimer was dissolved in degassed toluene (0.013M) under an inert atmosphere, 10 mol% of \( R(+)\)-BINAP and allyl acetate (2 equiv.) were added and mixture stirred at room temperature for 5 minutes. Resultant yellow suspension was then added to the above mixture of base and dihydropyrrolone and stirring at 0 °C continued. Upon full conversion of the starting material as indicated by HPLC-MS (~1 h) toluene was removed in vacuo on a heated to 40 °C bath. Crude NMR indicated single diastereomer. Crude yellow oil was purified by flash chromatography on SiO\(_2\) (EtOAc/hexanes, 2 to 82%).

**General protocol for Claisen rearrangement of 3-(allyloxy)-1,5-dihydro-2H-pyrrol-2-ones (V)**

A solution of corresponding allyl ether in toluene (0.04 M) is heated at reflux (110 °C) under an inert atmosphere for 16-24 h. Toluene is then evaporated in vacuo on a heated to 40 °C bath. Resultant keto-amide is obtained quantitatively in high purity as a single diastereomer.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-pentyl-4-phenylpyrrolidine-2,3-dione (7-71d).

According to general protocol V which employs 7-64, 7-71d (56.5 mg, quant.) was obtained as a yellow oil: $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.49 – 7.00 (m, 6H), 6.50 – 6.45 (m, 2H), 5.35 – 5.13 (m, 2H), 4.95 – 4.59 (m, 2H), 4.13 (d, $J$ = 14.3 Hz, 1H), 3.81 (s, 6H), 3.62 (dd, $J$ = 5.9, 2.3 Hz, 1H), 2.67 (dd, $J$ = 14.1, 6.7 Hz, 1H), 2.40 (dd, $J$ = 14.1, 7.7 Hz, 1H), 1.57 – 1.47 (m, 1H), 1.41 – 1.17 (m, 1H), 1.01 – 0.77 (m, 6H), 0.62 (t, $J$ = 7.2 Hz, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 199.9, 161.3, 159.3, 158.9, 136.2, 132.3, 130.9, 128.3, 128.0, 127.5, 119.9, 114.8, 104.5, 98.6, 62.3, 55.5, 55.4, 43.8, 40.7, 31.4, 29.5, 22.5, 22.0, 13.8; IR (film): 2930, 2954, 1758, 1712, 1509, 1210, 703 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{27}$H$_{33}$NO$_4$ [M+H]$^+$ 436.2482, found 436.2486.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-((Z)-hex-3-en-1-yl)-4-phenylpyrrolidine-2,3-dione (7-71a).

According to general protocol V which employs 7-64, 7-71a (29.9 mg, quant.) was obtained as a yellow oil: Rf = 0.6 (20% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.50 – 7.06 (m, 6H), 6.51 – 6.46 (m, 2H), 5.34 – 5.21 (m, 2H), 5.21 – 5.09 (m, 1H), 4.94 – 4.64 (m, 3H), 4.18 (d, \(J = 14.3\) Hz, 1H), 3.82 (s, 7H), 3.66 (dd, \(J = 6.1, 1.8\) Hz, 1H), 2.69 (dd, \(J = 14.1, 6.7\) Hz, 1H), 2.43 (dd, \(J = 14.1, 7.7\) Hz, 1H), 1.65 – 1.23 (m, 6H), 0.73 (t, \(J = 7.5\) Hz, 3H); \(^1\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 199.9, 161.4, 159.3, 158.9, 136.3, 132.9, 132.3, 130.9, 128.5, 128.1, 127.6, 126.5, 120.1, 114.8, 104.6, 98.7, 62.0, 55.5, 55.5, 43.8, 40.8, 29.8, 21.0, 20.2, 14.3; IR (film): 3075, 2960, 1758, 1713, 1509, 1209, 924, 704 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for \(C_{28}H_{33}NO_4\) [M+H]\(^+\) 448.2482, found 448.2473.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-isopropyl-4-phenylpyrrrolidine-2,3-dione (7-71c).

According to general protocol V which employs 7-64c, 7-71c (34.8 mg, quant.) was obtained as a yellow oil: Rf = 0.43 (40% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.48 – 6.45 (m, 2H), 7.39 – 7.06 (m, 4H), 6.53 – 6.49 (m, 2H), 5.44 (d, $J = 14.2$ Hz, 1H), 5.21 – 5.09 (m, 1H), 4.91 – 4.78 (m, 1H), 4.71 – 4.52 (m, 1H), 4.21 (d, $J = 14.2$ Hz, 1H), 3.83 (s, 6H), 3.67 (d, $J = 1.3$ Hz, 1H), 2.55 (dd, $J = 14.4$, 6.6 Hz, 1H), 2.26 (dd, $J = 14.4$, 7.8 Hz, 1H), 1.78 – 1.68 (m, 1H), 1.07 (d, $J = 7.3$ Hz, 3H), 0.45 (d, $J = 6.7$ Hz, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 198.2, 161.4, 160.4, 158.9, 136.4, 132.9, 131.1, 128.5, 128.0, 127.4, 119.6, 115.2, 104.4, 98.8, 68.5, 57.7, 55.6, 55.4, 43.6, 30.7, 22.1, 15.6; IR (film): 3076, 2964, 1758, 1712, 1508, 1210, 1035, 922, 705 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{25}$H$_{29}$NO$_4$ [M+H]$^+$ 408.2169, found 408.2167.
(rac)-4-allyl-5-cyclopropyl-1-(2,4-dimethoxybenzyl)-4-phenylpyrrolidine-2,3-dione (7-71b).

According to general protocol V which employs 7-64b, 7-71b (8.7 mg, quant.) was obtained as a white solid: Rf = 0.13 (20% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.39 – 7.21 (m, 3H), 7.20 – 7.04 (m, 3H), 6.46 – 6.42 (m, 2H), 5.60 – 5.33 (m, 1H), 5.18 (d, $J$ = 14.6 Hz, 1H), 5.08 – 4.79 (m, 2H), 4.59 (d, $J$ = 14.7 Hz, 1H), 3.81 (s, 3H), 3.79 (s, 3H), 3.02 – 2.77 (m, 2H), 2.71 – 2.41 (m, 1H), 0.52 – 0.48 (m, 1H), 0.34 – 0.29 (m, 1H), 0.22 – 0.08 (m, 1H), -0.05 – -0.31 (m, 2H); $^{13}$C NMR (176 MHz, CDCl$_3$) $\delta$ 202.8, 160.9, 159.5, 158.5, 138.1, 131.3, 131.0, 128.4, 128.3, 127.5, 120.5, 115.5, 104.2, 98.6, 67.4, 55.5, 55.3, 40.7, 13.8, 5.1, 2.2; IR (film): 3077, 2933, 1760, 1711, 1508, 1209, 1033, 702 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{25}$H$_{27}$NO$_4$ [M+H]$^+$ 408.2169, found 408.2167. Mp = 142-145 °C; X-Ray was obtained.
(rac)-4-allyl-5-(((tert-butyldimethylsilyl)oxy)methyl)-1-(2,4-dimethoxybenzyl)-4-phenylpyrrolidine-2,3-dione (7-71k).

According to general protocol V which employs 7-64k, 7-71k (26.9 mg, quant.) was obtained as a yellow oil: Rf = 0.55 (40% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) δ 7.60 – 7.38 (m, 2H), 7.39 – 7.04 (m, 4H), 6.59 – 6.38 (m, 2H), 5.36 – 5.25 (m, 1H), 5.17 (d, $J$ = 14.4 Hz, 1H), 4.85 (dd, $J$ = 10.2, 1.7 Hz, 1H), 4.70 (dd, $J$ = 17.0, 1.6 Hz, 1H), 4.12 (d, $J$ = 14.4 Hz, 1H), 3.83 (s, 6H), 3.67 – 3.41 (m, 2H), 3.20 (dd, $J$ = 10.8, 1.5 Hz, 1H), 2.56 (dd, $J$ = 14.2, 7.2 Hz, 1H), 2.43 (dd, $J$ = 14.2, 7.2 Hz, 1H), 0.71 (s, 9H), -0.24 (s, 3H), -0.36 (s, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) δ 197.0, 161.4, 160.3, 159.0, 136.5, 132.4, 131.4, 128.1, 127.9, 127.1, 119.5, 115.1, 104.8, 98.9, 63.9, 58.0, 55.6, 54.4, 43.1, 40.4, 25.7, 18.2, -6.1; IR (film): 3416, 2929, 1762, 1714, 1209, 924, 837 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{29}$H$_{39}$NO$_5$Si [M+H]$^+$ 510.2670, found 510.2666.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-4,5-diphenylpyrrolidine-2,3-dione (7-71h).

According to general protocol V which employs 7-64h, 7-71h (26.2 mg, quant.) was obtained as a yellow oil: \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta\) 7.10 – 6.81 (m, 9H), 6.71 (br s, 2H), 6.53 – 6.31 (m, 2H), 5.50 – 5.33 (m, 1H), 5.28 (d, \(J = 13.9\) Hz, 1H), 5.05 – 4.80 (m, 2H), 4.63 (s, 1H), 3.83 (s, 3H), 3.78 (d, \(J = 13.9\) Hz, 1H), 3.76 (s, 3H), 2.89 (dd, \(J = 14.1, 6.8\) Hz, 1H), 2.65 (dd, \(J = 14.0, 7.7\) Hz, 1H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 201.2, 161.4, 159.2, 159.0, 136.6, 136.2, 132.6, 131.0, 129.1, 128.7, 128.3, 128.2, 128.1, 127.9, 126.9, 125.4, 120.3, 114.6, 104.2, 98.5, 67.7, 57.8, 55.5, 55.3, 43.6, 41.7; IR (film): 3064, 2936, 1759, 1713, 1508, 1210, 925, 700 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{25}\)H\(_{23}\)NO\(_4\) [M+H]\(^+\) 442.2012, found 442.2013.

Alternatively, 7-71h (105 mg, 95%) was prepared from 7-70h (101 mg, 0.25 mmol) utilizing conditions described in protocol IV.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-(4-methoxyphenyl)-4-phenylpyrrolidine-2,3-dione (7-71n).

According to general protocol V which employs 7-64n, 7-71n (20.8 mg, quant.) was obtained as a yellow oil: \( R_f = 0.39 \) (40% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl$_3$) \( \delta \) 7.10 – 6.79 (m, 6H), 6.65 – 6.51 (m, 4H), 6.50 – 6.34 (m, 2H), 5.48 – 5.32 (m, 1H), 5.26 (d, \( J = 14.0 \) Hz, 1H), 5.00 – 4.81 (m, 2H), 4.57 (s, 1H), 3.83 (s, 3H), 3.76 (s, 3H), 3.74 (d, \( J = 14.0 \) Hz, 1H), 3.68 (s, 3H), 2.85 (dd, \( J = 14.1, 6.7 \) Hz, 1H), 2.62 (dd, \( J = 14.1, 7.7 \) Hz, 1H); \(^1\)C NMR (101 MHz, CDCl$_3$) \( \delta \) 201.3, 161.4, 159.3, 159.1, 136.9, 132.6, 131.2, 128.2, 127.9, 126.9, 120.2, 114.8, 113.7, 104.3, 98.6, 67.2, 57.8, 55.6, 55.4, 55.3, 43.8, 41.6; IR (film): 2935, 1759, 1712, 1511, 1209, 1033, 835, 700 cm$^{-1}$; HRMS (ESI) \( m/z \) calcd for C$_{29}$H$_{29}$NO$_5$ [M+H]$^+$ 472.2119, found 472.2194.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-(4-nitrophenyl)-4-phenylpyrroloidine-2,3-dione (7-711).

According to general protocol V which employs 7-641, 7-711 (30.3 mg, quant.) was obtained as a yellow oil: Rf = 0.14 (20% EtOAc/hexanes); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.90 (d, $J = 8.7$ Hz, 2H), 7.39 – 7.34 (m, 1H), 7.04 – 6.75 (m, 7H), 6.43 – 6.38 (m, 2H), 5.52 – 5.31 (m, 1H), 5.25 (d, $J = 14.2$ Hz, 1H), 5.08 – 4.86 (m, 2H), 4.76 (s, 1H), 3.84 (d, $J = 14.1$ Hz, 1H), 3.81 (s, 3H), 3.71 (s, 3H), 2.94 (dd, $J = 14.1$, 6.3 Hz, 1H), 2.70 (dd, $J = 14.3$, 7.9 Hz, 1H); $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 200.4, 161.7, 159.2, 158.9, 147.4, 144.1, 136.0, 132.6, 130.4, 128.4, 128.0, 127.6, 123.4, 120.9, 114.0, 104.5, 98.6, 66.9, 57.7, 55.6, 55.3, 43.4, 41.9; IR (film): 2938, 1761, 1715, 1522, 1348, 1210, 1033, 705 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{28}$H$_{26}$N$_2$O$_6$ [M+H]$^+$ 485.1718, found 485.1711.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(4-
(trifluoromethyl)phenyl)pyrrolidine-2,3-dione (7-71m).

According to general protocol V which employs 7-64m, 7-71m (32.6 mg, quant.) was obtained as a yellow oil: Rf = 0.2 (20% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.29 (d, $J$ = 8.4 Hz, 2H), 7.09 – 6.59 (m, 8H), 6.45 – 6.38 (m, 2H), 5.49 – 5.34 (m, 1H), 5.25 (d, $J$ = 14.1 Hz, 1H), 5.05 – 4.87 (m, 2H), 4.70 (s, 1H), 3.82 (d, $J$ = 14.1 Hz, 1H), 3.81 (s, 3H), 3.71 (s, 3H), 2.92 (dd, $J$ = 14.1, 6.6 Hz, 1H), 2.68 (dd, $J$ = 14.1, 7.8 Hz, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 200.8, 161.6, 159.3, 159.0, 140.7, 136.2, 132.6, 130.7, 128.2, 128.1, 127.3, 125.2, 120.7, 114.3, 104.4, 98.5, 67.2, 57.7, 55.6, 55.3, 43.4, 41.8; IR (film): 2939, 1761, 1715, 1509, 1116, 928, 701 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{29}$H$_{26}$F$_3$NO$_4$ [M+H]$^+$ 510.1887, found 510.1877.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(pyridin-3-yl)pyrrolidine-2,3-dione (7-71f).

According to general protocol V which employs 7-64f, 7-71f (21.1 mg, quant.) was obtained as a yellow oil: Rf = 0.32 (80% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.29 (dd, $J = 4.5, 1.9$ Hz, 1H), 8.05 (s, 1H), 7.08 – 6.74 (m, 8H), 6.60 – 6.31 (m, 2H), 5.54 – 5.30 (m, 1H), 5.22 (d, $J = 14.1$ Hz, 1H), 5.03 – 4.84 (m, 2H), 4.69 (s, 1H), 3.82 (d, $J = 14.1$ Hz, 1H), 3.81 (s, 3H), 3.72 (s, 3H), 2.91 (dd, $J = 14.0, 6.5$ Hz, 1H), 2.69 (dd, $J = 14.1, 7.8$ Hz, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 200.7, 161.6, 159.2, 158.9, 149.9, 149.5, 136.5, 132.6, 132.3, 130.6, 128.4, 128.1, 127.4, 123.1, 120.8, 114.4, 104.5, 98.6, 65.2, 57.4, 55.6, 55.3, 43.6, 41.6; IR (film): 2937, 1760, 1714, 1509, 1210, 926, 716 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{27}$H$_{26}$N$_2$O$_4$ [M+H]$^+$ 443.1965, found 443.1958.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-4-phenyl-5-(thiazol-4-yl)pyrrolidine-2,3-dione (7-71g).

According to general protocol V which employs 7-64g, 7-71g (32.8 mg, quant.) was obtained as a yellow oil: Rf = 0.34 (20% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 8.40 (d, $J = 1.9$ Hz, 1H), 7.14 – 6.91 (m, 6H), 6.83 (d, $J = 1.9$ Hz, 1H), 6.59 – 6.29 (m, 3H), 5.46 – 5.32 (m, 1H), 5.19 (d, $J = 14.2$ Hz, 1H), 4.95 – 3.77 (m, 3H), 3.83 (d, $J = 14.2$ Hz, 1H), 3.82 (s, 3H), 3.78 (s, 3H), 2.82 (dd, $J = 14.0$, 7.0 Hz, 1H), 2.66 (dd, $J = 14.1$, 7.5 Hz, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 199.4, 161.4, 159.4, 159.0, 153.2, 153.1, 136.3, 132.5, 131.0, 127.9, 127.8, 126.9, 120.2, 117.2, 114.7, 104.4, 98.6, 63.1, 57.3, 55.6, 55.4, 43.3, 41.5; IR (film): 3081, 2937, 1761, 1713, 1509, 1209, 1035, 925, 731 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{25}$H$_{24}$N$_2$O$_4$S [M+H]$^+$ 449.1529, found 449.1531.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-pentyl-4-((E)-styryl)pyrrolidine-2,3-dione (7-71j).

Attempts to prepare 7-71j via Claisen rearrangement (V) result in decomposition of starting material allyl ether 7-64j, and only up to 18% of product could be isolated with impurities after purification by flash chromatography on SiO₂.

According to protocol IV which employs 7-68 (22.0 mg, 0.05 mmol), 7-71j (22.3 mg, 93%) was obtained as a yellow oil: Rf = 0.65 (50% EtOAc/hexanes); ¹H NMR (400 MHz, CDCl₃) δ 7.57 – 7.08 (m, 6H), 6.72 (d, J = 16.5 Hz, 1H), 6.51 – 6.46 (m, 2H), 6.10 (d, J = 16.5 Hz, 1H), 5.58 – 5.39 (m, 1H), 5.22 (d, J = 14.3 Hz, 1H), 4.96 (d, J = 10.2 Hz, 1H), 4.76 (d, J = 16.3 Hz, 1H), 4.17 (d, J = 14.3 Hz, 1H), 3.83 (s, 6H), 3.58 (dd, J = 5.9, 3.1 Hz, 1H), 2.36 (dd, J = 13.6, 7.4 Hz, 1H), 2.26 (dd, J = 13.7, 7.0 Hz, 1H), 1.83 – 1.63 (m, 2H), 1.34 – 1.02 (m, 6H), 0.77 (t, J = 6.8 Hz, 3H); ¹³C NMR (101 MHz, CDCl₃) δ 158.9, 158.0, 136.8, 133.7, 133.1, 132.2, 131.5, 131.2, 128.8, 128.1, 126.4, 124.3, 120.3, 115.2, 104.7, 98.7, 60.4, 55.6, 55.5, 53.2, 42.8, 40.6, 31.8, 29.7, 23.6, 22.4, 14.0; IR (film): 2931, 1759, 1710, 1509, 1209, 1035, 926, 695 cm⁻¹; HRMS (ESI) m/z calcd for C₂₉H₃₅NO₄ [M+H]⁺ 462.2639, found 462.2634.
(rac)-4-allyl-1-(2,4-dimethoxybenzyl)-5-pentyl-4-(4-(trifluoromethyl)phenyl)pyrrolidine-2,3-dione (7-71m).

According to general protocol V which employs 7-64m, 7-71m (32.7 mg, 84%) was obtained after purification by flash chromatography on SiO₂ (EtOAc/hexanes, 2 to 42%), a yellow oil: Rf = 0.55 (40% EtOAc/hexanes); ¹H NMR (300 MHz, CDCl₃) δ 7.62 – 7.51 (m, J = 8.6 Hz, 4H), 7.24 (d, J = 4.5 Hz, 1H), 6.54 – 6.47 (m, 2H), 5.45 – 5.00 (m, 2H), 4.98 – 4.81 (m, 1H), 4.79 – 4.70 (m, 1H), 4.13 (d, J = 14.2 Hz, 1H), 3.83 (s, 3H), 3.82 (s, 3H), 3.68 (dd, J = 5.6, 2.4 Hz, 1H), 2.63 (dd, J = 14.1, 7.0 Hz, 1H), 2.43 (dd, J = 14.2, 7.5 Hz, 1H), 1.64 – 1.46 (m, 1H), 1.39 – 1.19 (m, 1H), 1.08 – 0.65 (m, 5H), 0.61 (t, J = 7.0 Hz, 3H), 0.52 (s, 1H); ¹³C NMR (75 MHz, CDCl₃) δ 199.1, 161.5, 158.9, 140.5, 132.4, 130.2, 128.6, 125.3, 125.2, 120.6, 114.6, 104.6, 98.7, 62.0, 55.6, 55.5, 55.3, 44.0, 40.9, 31.3, 29.5, 22.3, 21.9, 13.7; IR (film): 2933, 1759, 1714, 1509, 1328, 926, 837 cm⁻¹; HRMS (ESI) m/z calcld for C₂₈H₃₂F₃NO₄ [M+H]⁺ 504.2356, found 504.2353.
(rac)-4-allyl-4-benzoyl-1-(2,4-dimethoxybenzyl)-5-pentylpyrrolidine-2,3-dione (7-71o).

According to general protocol V which employs 7-64o, 7-71o (49.0 mg, quant.) was obtained as a yellow oil: Rf = 0.33 (30% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.61 – 7.46 (m, 3H), 7.45 – 7.32 (m, 2H), 7.23 – 7.20 (m, 1H), 6.56 – 6.29 (m, 2H), 5.41 – 5.28 (m, 1H), 5.24 (d, $J$ = 14.4 Hz, 1H), 4.97 – 4.65 (m, 2H), 4.37 (d, $J$ = 14.4 Hz, 1H), 3.80 (s, 3H), 3.79 (s, 3H), 3.67 (dd, $J$ = 10.0, 2.9 Hz, 1H), 2.74 (dd, $J$ = 13.8, 6.9 Hz, 1H), 2.63 (dd, $J$ = 13.8, 7.9 Hz, 1H), 1.94 – 1.84 (m, 1H), 1.69 – 1.36 (m, 1H), 1.22 – 0.86 (m, 6H), 0.74 (t, $J$ = 7.1 Hz, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 198.8, 195.9, 161.3, 159.0, 158.7, 136.5, 133.3, 132.1, 130.3, 129.3, 128.8, 121.4, 114.9, 104.6, 98.4, 61.9, 59.5, 55.5, 55.4, 42.2, 40.3, 31.7, 28.8, 25.1, 22.2, 13.9; IR (film): 2961, 1756, 1714, 1676, 1261, 1097, 1031, 799 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{28}$H$_{33}$NO$_5$ [M+H]$^+$ 464.2431, found 464.2421.
(rac)-1,4-diallyl-5-pentyl-4-phenylpyrrolidine-2,3-dione (7-71p).

According to general protocol V which employs 7-64p, 7-71p (19.7 mg, quant) was obtained as a yellow oil: Rf = 0.28 (20% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.46 – 7.05 (m, 5H), 5.86 – 5.78 (m, 1H), 5.53 – 5.30 (m, 3H), 5.18 – 4.87 (m, 2H), 4.77 – 4.45 (m, 1H), 3.93 (dd, \(J = 5.9, 2.7\) Hz, 1H), 3.65 (dd, \(J = 14.8, 8.3\) Hz, 1H), 2.92 (dd, \(J = 14.0, 6.4\) Hz, 1H), 2.65 (dd, \(J = 14.0, 8.0\) Hz, 1H), 1.56 – 1.32 (m, 2H), 0.99 – 0.82 (m, 3H), 0.80 – 0.66 (m, 3H), 0.64 (t, \(J = 7.1\) Hz, 3H); \(^13\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 199.7, 159.5, 136.5, 131.0, 130.8, 128.5, 128.0, 127.7, 120.8, 120.5, 62.6, 55.5, 45.2, 44.4, 31.4, 29.6, 22.7, 22.0, 13.8; IR (film): 2955, 2928, 1712, 1447, 928, 702 cm\(^{-1}\); HRMS (ESI) m/z calcd for C\(_{21}\)H\(_{27}\)NO\(_2\) [M+H]\(^+\) 326.2115, found 326.2112.

(rac)-4-allyl-5-pentyl-4-phenylpyrrolidine-2,3-dione (7-104a).

According to general protocol V which employs 0.13M PhMe solution of 7-66 (1.31 g, 4.59 mmol), 7-104a (1.1 g, 84%) was obtained after purification by flash chromatography on SiO\(_2\) (EtOAc/hexanes, 2 to 42%), an off white solid: Rf = 0.29 (40% EtOAc/hexanes); \(^1\)H
NMR (400 MHz, CDCl\textsubscript{3}) \(\delta\) 10.13 (s, 1H), 7.46 – 7.04 (m, 5H), 5.74 – 5.29 (m, 1H), 5.29 – 4.91 (m, 2H), 3.90 (dd, \(J = 8.5, 3.6\) Hz, 1H), 3.00 (dd, \(J = 14.1, 6.4\) Hz, 1H), 2.80 (dd, \(J = 14.1, 8.1\) Hz, 1H), 1.52 – 0.93 (m, 8H), 0.73 (t, \(J = 7.0\) Hz, 3H); \(^{13}\)C NMR (101 MHz, CDCl\textsubscript{3}) \(\delta\) 202.3, 162.3, 137.0, 131.2, 128.5, 127.8, 127.6, 120.6, 59.7, 56.5, 41.9, 34.5, 31.4, 25.1, 22.2, 13.9; IR (film): 3186, 3062, 2927, 1714, 1756, 1448, 1326, 929, 702 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcld for C\(_{18}\)H\(_{23}\)NO\(_2\) 286.1802 [M+H]\(^{+}\), found 286.1797. Mp = 89 – 90 °C.

\[(\text{rac})-4\text{-allyl-5-cyclopropyl-4-phenylpyrrolidine-2,3-dione (7-104b).}\]

According to general protocol V which employs 0.1M PhMe solution of 7-66b (150 mg, 0.59 mmol), 7-104b (126 mg, 84%) was obtained after purification by flash chromatography on SiO\(_2\) (EtOAc/hexanes, 2 to 42%), an off white solid: Rf = 0.19 (40% EtOAc/hexanes); \(^1\)H NMR (400 MHz, CDCl\textsubscript{3}) \(\delta\) 9.61 (s, 1H), 7.47 – 7.03 (m, 5H), 5.72 – 5.39 (m, 1H), 5.26 – 4.89 (m, 2H), 3.24 (d, \(J = 8.3\) Hz, 1H), 3.03 (dd, \(J = 14.0, 6.1\) Hz, 1H), 2.82 (dd, \(J = 14.1, 8.3\) Hz, 1H), 0.54 – -0.11 (m, 5H); \(^{13}\)C NMR (101 MHz, CDCl\textsubscript{3}) \(\delta\) 203.6, 162.0, 138.2, 131.4, 129.0, 128.5, 128.0, 127.7, 127.5, 120.8, 64.2, 57.1, 41.3, 14.3, 3.8, 2.8; IR (film): 3217, 3081, 1763, 1725, 1432, 1288, 921, 699 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcld for C\(_{16}\)H\(_{17}\)NO\(_2\) 278.1152 [M+Na]\(^{+}\), found 278.1145.
(rac)-4-allyl-4,5-diphenylpyrroolidine-2,3-dione (7-100).

According to general protocol V which employs 0.11M PhMe solution of 7-66b (100 mg, 0.34 mmol), 7-100 (90.5 mg, 91%) was obtained after purification by flash chromatography on SiO$_2$ (EtOAc/hexanes, 2 to 42%), an off white oily solid: Rf = 0.3 (40% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 9.30 (s, 1H), 7.15 – 6.95 (m, 6H), 6.91 – 6.61 (m, 4H), 5.75 – 5.60 (m, 1H), 5.43 – 4.81 (m, 3H), 3.09 (dd, $J$ = 14.2, 6.1 Hz, 1H), 2.93 (dd, $J$ = 14.2, 8.3 Hz, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 202.9, 162.1, 137.4, 136.9, 131.6, 128.3, 128.2, 128.0, 127.9, 127.3, 127.2, 121.0, 63.3, 59.0, 41.5; IR (film): 3114, 3064, 1761, 1727, 1456, 927, 757, 697 cm$^{-1}$; HRMS (ESI) $m/z$ calcd for C$_{19}$H$_{17}$NO$_2$ 292.1332 [M+H]$^+$, found 292.1329.

DMB-removal procedures

3-hydroxy-5-pentyl-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-65).

In a vial, protected pyrroolidinone 5-43 (21.0 mg, 0.05 mmol) was dissolved in 1 ml of DCM and 1 ml of TFA added; vial capped tight and the reaction stirred at room temperature for 1h when solution acquired deep purple color, full conversion was observed by HPLC-MS.
Solvent and TFA were evaporated and purple oily residue purified by flash chromatography eluting EtOAc/hexanes (2-62% EtOAc). 10.4 mg, an off-white solid, 80% yield: Rf = 0.29 (40% EtOAc/hexanes); $^1$H NMR (400 MHz, CDCl$_3$) δ 7.65 – 7.55 (m, 1H), 7.45 – 7.35 (m, 2H), 7.34 – 7-26 (m, 1H), 4.52 (d, $J$ = 5.7 Hz, 0H), 2.00 – 1.8 (m, 1H), 1.49 – 1.31 (m, 1H), 1.30 – 1.1 (m, 6H), 0.95 – 0.71 (m, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) δ 169.3, 141.5, 131.7, 128.8, 127.9, 127.5, 124.5, 54.8, 33.6, 31.7, 24.9, 22.6, 14.1; IR (film): 3213, 2927, 1699, 1298, 897, 696 cm$^{-1}$; HRMS (ESI) m/z calcd for C$_{15}$H$_{19}$NO$_2$ 246.1488 [M+H$^+$], found 246.1489. Mp = 160-161 °C.

![Image](image_url)

3-hydroxy-4,5-diphenyl-1,5-dihydro-2H-pyrrol-2-one (7-65h).

In a vial, protected pyrrolidinone 7-70h (0.20 g, 0.49 mmol) was dissolved in 1 ml of DCM in and 1 ml of TFA added dropwise; vial capped tight and reaction mixture stirred at room temperature for 1h when solution acquired deep purple color, full conversion was observed by HPLC-MS. Solvent and TFA were evaporated and purple oily residue purified by flash chromatography eluting EtOAc/hexanes (2-100% EtOAc). 0.12 g, an off-white solid, 97% yield: $^1$H NMR (300 MHz, DMSO-d$_6$) δ 10.29 (s, 1H), 8.90 (s, 1H), 7.58 (d, $J$ = 7.6 Hz, 2H), 7.37 – 6.95 (m, 8H), 5.55 (s, 1H); $^{13}$C NMR (101 MHz, DMSO) δ 168.0, 144.5, 139.6, 132.3, 128.6, 128.1, 127.8, 127.5, 126.9, 126.7, 121.8, 56.9; IR (film): 3330, 2920, 1694, 1434,
1205, 903, 692 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for \(\text{C}_{16}\text{H}_{13}\text{NO}_2\) 252.1019 [M+H]\(^+\), found 252.1013. \(\text{Mp} = 198 – 199 \degree \text{C}\)

![Chemical structure](image)

**3-(allyloxy)-5-pentyl-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-66).**

In a vial, protected O-allyl pyrrolidinone \(7\-64\) (23.0 mg, 0.05 mmol) was dissolved in 1 ml of DCM and 1 ml of TFA added dropwise; vial capped and the reaction stirred at room temperature for 1h when solution acquired deep purple color, full conversion was observed by HPLC-MS. Solvent and TFA were evaporated and purple oily residue purified by flash chromatography eluting EtOAc/hexanes (2-62% EtOAc). 13.6 mg, an off-white solid, 90% yield: \(\text{Rf} = 0.38\) (40% EtOAc/hexanes); \(^1\text{H NMR}\) (400 MHz, CDCl\(_3\)) \(\delta\) 12.47 (br s, 1H), 8.56 (s, 1H), 7.66 – 7.34 (m, 5H), 6.05 – 5.89 (m, 1H), 5.36 – 5.26 (m, 1H), 5.21 (d, \(J = 10.9\) Hz, 1H), 4.89 – 4.52 (m, 2H), 4.58 (dd, \(J = 7.9, 3.1\) Hz, 1H), 1.45 – 1.25 (m, 8H), 0.83 (t, \(J = 6.6\) Hz, 3H); HRMS (ESI) \(m/z\) calcd for \(\text{C}_{18}\text{H}_{23}\text{NO}_2\) 286.1802 [M+H]\(^+\), found 286.1797.
3-(allyloxy)-5-cyclopropyl-4-phenyl-1,5-dihydro-2H-pyrrol-2-one (7-66b).

In a round bottom flask, protected O-allyl pyrrolidinone 7-64b (0.26 g, 0.64 mmol) was dissolved in 2.15 ml of DCM and 2.15 ml of TFA added dropwise; reaction stirred at room temperature for 1.5 h when solution acquired deep purple color, full conversion was observed by HPLC-MS. Solvent and TFA were evaporated and purple oily residue purified by flash chromatography eluting EtOAc/hexanes (2-32% EtOAc). 0.15 g, an off-white solid, 92% yield;

3-(allyloxy)-4,5-diphenyl-1,5-dihydro-2H-pyrrol-2-one (7-66h).

In a round bottom flask, O-Allyl pyrrolidinone 7-64h (0.24 g, 0.54 mmol) was dissolved in 2 ml of DCM and 2 ml of TFA added dropwise; reaction stirred at room temperature for 4h when solution acquired deep purple color and full conversion was observed by HPLC-MS. Solvent and TFA were evaporated and purple oily residue purified by flash chromatography eluting EtOAc/hexanes (2-42% EtOAc). 0.11 g, an off-white solid, 70% yield: $^1$H NMR (400
MHz, CDCl$_3$) $\delta$ 8.27 (s, 1H), 7.56 – 7.50 (m, 2H), 7.40 – 6.99 (m, 8H), 6.06 – 5.96 (m, 1H), 5.50 (s, 1H), 5.46 – 5.19 (m, 2H), 5.05 – 4.90 (m, 1H), 4.90 – 4.77 (m, 1H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 136.2, 135.7, 133.2, 130.5, 129.4, 129.2, 129.1, 128.6, 128.2, 127.6, 119.2, 72.2, 60.5;

(rac)-4-allyl-4,5-diphenylpyrrolidine-2,3-dione (7-100).

In a vial, protected C-allyl dione 7-71h (38 mg, 0.11 mmol) was dissolved in 0.5 ml of TFA; vial was capped tight and heated at 80 ºC for 24 h. TFA was then evaporated and crude purple oil purified by flash chromatography eluting EtOAc/hexanes (2-100% EtOAc). 17.2 mg, an off-white film, 54% yiled (61% BRSM). Starting material (5.5 mg, 11%) was recovered. *For characterization see above.*

*It should be noted that increasing of the reaction times (30+ h), amount of TFA or temperature led to full conversion associated with decomposition of starting material and product.*
Synthesis of beta amino acids

(rac)-2-(amino(phenyl)methyl)-2-phenylpent-4-enoic acid (7-105c).

7-100 (69.8 mg, 0.24 mmol) was suspended in 1M aqueous solution of sodium hydroxide (0.95 ml, 4equiv.) then placed on the ice bath and cooled to 0 °C. 30% aqueous hydrogen peroxide (0.11 ml, 4 equiv.) was then slowly added; reaction mixture was allowed to warm up to room temperature and stirred until full conversion of starting material was observed by HPLC-MS (4 h). Reaction mixture was then concentrated in vacuo on a heated to 40 °C bath. Resultant crude material was purified by reverse phase column chromatography on C18 SiO₂ (H₂O/CH₃CN, 2-50% CH₃CN, 0.1% TFA additive) flash chromatography column. 47.8 mg, a slightly yellow oil, 71% yield: ¹H NMR (400 MHz, CD₃OD) δ 7.53 – 7.36 (m, 4H), 7.30 (t, J = 7.7 Hz, 2H), 7.18 – 7.02 (m, 2H), 6.87 (d, J = 7.4 Hz, 2H), 5.95 (m, 1H), 5.41 – 5.15 (m, 2H), 4.86 (s, 1H), 2.79 (m, 1H), 2.58 (dd, J = 14.3, 7.8 Hz, 1H); ¹³C NMR (101 MHz, CD₃OD) δ 177.3, 136.3, 133.9, 133.2, 130.8, 130.4, 130.0, 129.7, 129.4, 121.2, 59.9, 58.2, 40.0; IR (film): 3440, 1681, 1203, 1137, 704 cm⁻¹; HRMS (ESI) m/z calcd for C₁₈H₁₉NO₂ 282.1489 [M+H]⁺, found 282.1484.
(rac)-2-allyl-3-amino-2-phenyloctanoic acid (7-105a).

7-104a (68.1 mg, 0.24 mmol) was suspended in 1M aqueous solution of sodium hydroxide (0.95 ml, 4equiv.) then placed on the ice bath and cooled to 0 °C. 30% aqueous hydrogen peroxide (0.11 ml, 4 equiv.) was then slowly added; reaction mixture was allowed to warm up to room temperature and stirred until full conversion of starting material was observed by HPLC-MS (8 h). Reaction mixture was then directly purified by reverse phase column chromatography on C18 SiO₂ (H₂O/CH₃CN, 2-65% CH₃CN, 0.1% TFA additive) flash chromatography column. 68.3 mg, a clear glassy oil, 74% yield, TFA salt: ¹H NMR (400 MHz, CD₃OD) δ 7.56 – 7.19 (m, 5H), 5.92 – 5.63 (m, 1H), 5.41 – 5.18 (m, 2H), 3.82 – 3.44 (m, 1H), 3.23 (dd, J = 14.4, 8.7 Hz, 1H), 2.91 (dd, J = 14.3, 5.8 Hz, 1H), 2.00 – 1.72 (m, 1H), 1.41 – 1.16 (m, 8H), 0.90 (t, J = 7.0 Hz, 3H); ¹³C NMR (101 MHz, CD₃OD) δ 177.5, 136.9, 133.2, 129.9, 129.4, 129.3, 120.7, 58.0, 55.8, 39.6, 32.6, 29.0, 26.4, 23.4, 14.3; IR (film): 3147, 2960, 2873, 1670, 1498, 1197, 1145, 703 cm⁻¹; HRMS (ESI) m/z calcd for C₁₇H₂₅NO₂ 276.1958 [M+H]⁺, found 276.1953. X-Ray was obtained.
(rac)-2-(amino(cyclopropyl)methyl)-2-phenylpent-4-enoic acid (7-105b)

7-104b (0.23 g, 0.9 mmol) was suspended in 1M aqueous solution of sodium hydroxide (3.7 ml, 4equiv.) then placed on the ice bath and cooled to 0 °C. 30% aqueous hydrogen peroxide (0.42 ml, 4 equiv.) was then slowly added; reaction mixture was allowed to warm up to room temperature and stirred overnight (19 h). Reaction mixture was then concentrated in vacuo on a heated to 40 °C bath. Resultant crude material was purified by reverse phase column chromatography on C18 SiO₂ (H₂O/CH₃CN, 5-70% CH₃CN, 0.1% TFA additive) flash chromatography column. 95.5 mg, a clear oil, 43% yield: ¹H NMR (300 MHz, CD₃OD) δ 7.79 – 7.04 (m, 5H), 5.84 – 5.72 (m, 1H), 5.48 – 5.15 (m, 2H), 3.45 (dd, J = 14.4, 7.2 Hz, 1H), 3.02 (d, J = 10.3 Hz, 1H), 2.93 (dd, J = 14.4, 6.7 Hz, 1H), 1.05 – 0.21 (m, 5H); ¹³C NMR (75 MHz, CD₃OD) δ 177.2, 137.2, 133.7, 129.9, 129.6, 129.4, 128.7, 120.6, 60.3, 59.4, 39.8, 11.7, 11.4, 7.2, 2.5; IR (film): 3453, 3087, 1673, 1488, 1198, 706 cm⁻¹; HRMS (ESI) m/z calcd for C₁₅H₁₉NO₂ 246.1489 [M+H]⁺, found 246.1485.

Multiple byproducts were observed, starting material most likely participates in side-reactions with base/peroxide.
Diversification of 4-allyl-pyrroolidine-2,3-diones via Grignard/RCM Reactions

7-104a (0.14 g 0.5 mmol) was dissolved in 6.4 ml of dry THF under an inert atmosphere and cooled on ice bath to 0 °C. 2M THF solution of allylmagnesium chloride (0.6 ml, 2.3 equiv.) was added dropwise. Stirring at 0 °C was maintained for 3h. Reaction was then quenched with 9.0 ml of saturated ammonium chloride. Resultant suspension was transferred into separatory funnel and extracted with EtOAc 3 x 25 ml. Organic extracts were combined, washed with saturated ammonium chloride 1 x 25 ml and dried with Na₂SO₄. Filtered and solvent concentrated in vacuo. Crude oil obtained, was subjected to additional drying under high vacuum after which it was used in the next step without further purification; dissolved in 10 ml of dry DCM under an inert atmosphere and 10 mol% of Grubbs 1st generation catalyst (43.2 mg, 0.05 mmol) added. Reaction mixture was stirred at room temperature overnight (15 h). Next morning, another 5 mol% of Grubbs 1st generation catalyst (21.6 mg) was added and stirring maintained for additional 4h. Solvent was then evaporated and black oily residue was purified by flash chromatography eluting EtOAc/hexanes (9-65% EtOAc). 101 mg, a gray film (minor [Ru]-cat poison) 67% yield, 1:1 mixture of diastereomers which were separated:
(rac-syn)-7a-hydroxy-3-pentyl-3a-phenyl-2,3,3a,4,7,7a-hexahydro-1H-isoindol-1-one (7-106a).

t22: Rf = 0.34 (50% EtOAc/hexanes); \(^1^H\) NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.54 – 7.06 (m, 1H), 6.29 (s, 1H), 5.80 – 5.55 (m, 2H), 3.58 (dd, \(J = 10.6, 2.4\) Hz, 1H), 2.97 (s, 1H), 2.92 – 2.8 (m, 1H), 2.75 – 2.48 (m, 3H), 1.44 – 1.06 (m, 8H), 0.82 (t, \(J = 4.7\) Hz, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 179.5, 140.0, 128.5, 128.1, 126.8, 124.1, 123.8, 75.8, 61.3, 51.8, 34.9, 34.8, 31.7, 31.5, 27.2, 22.6, 14.1; IR (film): 3314, 3195, 2907, 1691, 1433, 1089, 664 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for C\(_{19}\)H\(_{25}\)NO\(_2\) 322.1778 [M+Na]\(^+\), found 322.1774.

(rac-anti)-7a-hydroxy-3-pentyl-3a-phenyl-2,3,3a,4,7,7a-hexahydro-1H-isoindol-1-one (7-106b).

t24: Rf = 0.23 (50% EtOAc/hexanes); \(^1^H\) NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.51 – 7.11 (m, 5H), 5.97 – 5.69 (m, 2H), 5.46 – 5.23 (m, 1H), 3.97 (dd, \(J = 8.6, 5.6\) Hz, 1H), 3.10 – 2.75 (m, 3H), 2.51 – 2.09 (m, 2H), 1.40 – 0.96 (m, 8H), 0.81 (t, \(J = 7.0\) Hz, 3H); \(^{13}\)C NMR (101 MHz, CDCl\(_3\)) \(\delta\) 177.7, 138.6, 128.0, 127.2, 126.7, 123.7, 78.0, 62.2, 52.9, 51.8, 31.1, 30.4, 29.0, 26.6, 22.5, 14.0; IR (film): 3348, 3243, 2924, 1691, 1195, 901, 701 cm\(^{-1}\); HRMS (ESI) \(m/z\)
calcd for C_{19}H_{25}NO_{2} 322.1778 [M+Na]^+, found 322.1774.

**RCM**

![RCM Reaction Scheme](image)

7-71a (37 mg, 0.08 mmol) was dissolved in 2.5 ml of dry DCM under an inert atmosphere and 10 mol % of Grubbs 1st generation catalyst (7.1 mg, 0.008 mmol) added, reaction mixture stirred at room temperature for 4h. Another 15 mol% of Grubbs 1st generation catalyst (11 mg, 0.012 mmol) added and stirring continued until the next day (24h). Reaction mixture was then concentrated *in vacuo* and black oil purified by flash chromatography eluting EtOAc/hexanes (2-62% EtOAc). 23.2 mg, a gray film (minor [Ru]-cat poison), 72% yield.

![Purified Product](image)

(rac)-1-(2,4-dimethoxybenzyl)-3a-phenyl-1,3a,4,7,8,8a-hexahydrocyclohepta[b]pyrrole-2,3-dione (7-107).

$^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.51 – 7.06 (m, 6H), 6.63 – 6.31 (m, 2H), 5.76 – 5.65 (m, 1H), 5.60 – 5.46 (m, 1H), 5.14 (d, $J = 14.6$ Hz, 1H), 4.54 (d, $J = 14.5$ Hz, 1H), 3.85 (dd, $J = 11.6$, 4.3 Hz, 1H), 3.82 (s, 3H), 3.81 (s, 3H), 3.35 (dd, $J = 17.5$, 5.6 Hz, 1H), 2.37 – 2.19 (m, 2H), 1.85 – 1.67 (m, 3H); $^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ 198.7, 188.5, 162.4, 161.0, 158.5, 135.7,
131.8, 129.2, 128.0, 127.8, 126.2, 115.6, 104.7, 98.7, 63.0, 55.6, 55.5, 54.1, 39.9, 32.2, 24.1, 23.2; IR (film): 3414, 2920, 1760, 1712, 1454, 1209, 1033, 700 cm\(^{-1}\); HRMS (ESI) \(m/z\) calcd for \(\text{C}_{24}\text{H}_{25}\text{NO}_{4}\) 392.1856 [M+Na]\(^+\), found 392.1856.
7.6 References


(8) Furusawa, E.; Fumsawa, S. Chemotherapy 1986, 32, 521.


275


