Copyright Statement

The digital copy of this thesis is protected by the Copyright Act 1994 (New Zealand).

This thesis may be consulted by you, provided you comply with the provisions of the Act and the following conditions of use:

- Any use you make of these documents or images must be for research or private study purposes only, and you may not make them available to any other person.
- Authors control the copyright of their thesis. You will recognise the author’s right to be identified as the author of this thesis, and due acknowledgement will be made to the author where appropriate.
- You will obtain the author's permission before publishing any material from their thesis.

To request permissions please use the Feedback form on our webpage. http://researchspace.auckland.ac.nz/feedback

General copyright and disclaimer

In addition to the above conditions, authors give their consent for the digital copy of their work to be used subject to the conditions specified on the Library Thesis Consent Form and Deposit Licence.

Note: Masters Theses

The digital copy of a masters thesis is as submitted for examination and contains no corrections. The print copy, usually available in the University Library, may contain corrections made by hand, which have been requested by the supervisor.
Synthetic Studies Towards Berkelic Acid

A thesis submitted in fulfilment of the
requirements for the degree of Doctor of Philosophy

by

Zoe Elizabeth Wilson

Department of Chemistry
University of Auckland
August 2010
Abstract

This thesis describes the synthetic endeavours towards berkelic acid, an extremophile derived bioactive natural product. *Penicillium* sp. Pitna 4 is a fungus isolated from Berkeley Pit Lake, a metal laden, pH 2.5 lake which formed when an abandoned copper mine in Butte, Montana filled with infiltrating ground water. Berkelic acid is one of several bioactive natural products isolated from this unlikely source and has been found to have desirable selective activity against the ovarian cancer cell line OVCAR-3, as well as inhibitory activity against caspase-1 and matrix metalloprotease-3. Synthetic access to this molecule is highly desirable due to its bioactivity and novel tetracyclic structure as well as the fact that the planned bioremediation of Berkeley Pit Lake may eliminate its natural source.

The synthetic studies undertaken have focused on developing a flexible strategy for the synthesis of berkelic acid that allows for future modification to structure to allow investigation of biological activity. The strategy is based on the use of a novel one-pot Horner-Wadsworth-Emmons/oxa-Michael cascade to couple two advanced intermediates – a phosphonate and a lactol. A final deprotection/spiroketalisation step then furnishes the spiroketal moiety. Careful functional group manipulations and key introduction of chirality were pivotal in the successful synthesis of a series of coupling partners which allowed the successful synthesis of a series of tricyclic analogues of berkelic acid as well as the entire tetracyclic core, with and without full substitution on the aromatic ring. The formal total synthesis of berkelic acid faltered at the penultimate step, but this project has none the less established a sound approach to this molecule which will build the foundations for a future total synthesis.
Preface

All the work described in this thesis was carried out by the author in the Department of Chemistry at the University of Auckland, except where due reference to the work of others has been made in the text.

Some parts of this work have been previously published:


- Featured as an Instant insight article in Chemical Biology (“Life at the extremes”, Chemical Biology, 2008, 3, B95)
Acknowledgements

Firstly I would like to take this opportunity to thank Professor Margaret Brimble for all her hard work and support. Your ability to get things done, and your passion for interesting science is inspiring and I will always be immensely grateful for all that you have done to help me through the years and the opportunities I have gained because of this.

Also thanks always to Jon for being such a great person to go and talk to, for being an excellent travel buddy, for putting up with my endless questions and visits to his office and for getting me to work early everyday no matter how much I want to sleep in…

I would like to thank the other members of the Brimble group, especially the wonderful Anoma for always being such a happy presence around the lab, no matter how many times I go ask her to help me sort out stuff and Janice for her amazing ability to sort out problems. Special mention must also go to Renata, Dom, Kris, Olivia, Jack and Amanda for being such nice people to have around. Thanks also to Jonathan for putting up with me taking over the whole fume hood on a much too regular basis...

On a more formal note thank you to the University of Auckland for the doctoral scholarship which has allowed me to be here and carry out this work in the first place.

Thank you to my friends for their support and for making sure I occasionally do something other than work, Meredith - I probably owe my continuing sanity in large part to those Meredith rated movie trips and the chats, Ellie, Mary, Rob, Troy, Megs, Chris and everyone else – thanks for all the fun times.

Thanks to Ralph for being so entertaining and giving the best hugs even if you do end up leaving white fur everywhere…

Thanks to the St Gallen Symposium and all the wonderful people I have met there for opening my eyes to the world around me.

Also a huge thanks goes to my ballet teachers past and present – Mrs M, Saori, Paola, Meika, Emma et al. for keeping me in shape, helping me focus and teaching me much more than ballet.

Finally I have been blessed with the best family a person could ever want:

My extended family for being interested in what I am doing, and understanding that its not personal when I disappear into the lab for extended periods of time.

Amelia, Jon and Griffin – thanks for always making me feel like your home is my home and for the endlessly fun Friday nights…

Mum and Dad, thank you so much for everything. Anything I have ever and will ever achieve is because of you and all your hard work and love. I only hope that one day I can repay the favour!

Finally thanks to Gabby – I am so lucky to have you! A best friend/soul mate/sister all wrapped into one. Life would be so lonely without you. Thanks for all the things little and big you have done to help me, especially these last few months when things have gotten so crazy.

Zoe Wilson

August 2010
Table of Contents

Abstract i.
Preface ii.
Acknowledgements iii.
Table of contents iv.
Abbreviations vii.

Chapter 1: Introduction

1.1. The importance of extremophiles 3
1.2. Berkelic acid – isolation and biology 3
   1.2.1. Berkeley Pit Lake 3
   1.2.2. Isolation of berkelic acid 4
   1.2.3. Biological activity 5
1.3. Related molecules 5
   1.3.1. Other secondary metabolites from Berkeley Pit Lake 5
   1.3.2. Other 6,5-benzannulated spiroketal containing natural products 7
1.4. Berkelic acid – Chemistry 8
   1.4.1. Zhou and Snider – Tetracyclic model 8
   1.4.2. Fürstner et al. – Berkelic acid methyl ester synthesis and structural revision 9
   1.4.3. Snider et al. – Total synthesis of berkelic acid 1b and assignment of absolute stereochemistry 12
   1.4.4. De Brabander – Biomimetic total synthesis 14
   1.4.5. Summary of synthetic endeavours to date 17

Chapter 2: Discussion

2.1. Overview 21
2.2. Synthesis of tricyclic analogues 22
2.2.1. Introduction 22

2.2.2. Retrosynthesis 22

2.2.3. Synthesis of phosphonate coupling partner 96 23

2.2.4. Synthesis of aldehyde coupling partners 112 - 118 28

2.2.5. Coupling and cyclisation 32

2.2.6. Late stage functionalisation of the spiroketal molecules 40

2.3. Synthesis of the tetracyclic core of berkelic acid 42

2.3.1. Introduction 42

2.3.2. Retrosynthesis 43

2.3.3. Synthesis of phosphonate 149 using TBDMS protecting groups 44

2.3.4. Synthesis of chiral lactone 175 45

2.3.5. Model Horner-Wadsworth-Emmons/oxa-Michael cascade 73

2.3.6. Coupling and completion 76

2.3.7. Summary of the synthesis of tetracyclic model spiroketal 94 88

2.4. Total synthesis of berkelic acid 1 90

2.4.1. Introduction 90

2.4.2. Retrosynthesis 90

2.4.3. Synthesis of fully substituted lactone 204 92

2.4.4. Attempted synthesis of phosphonate 92 for total synthesis of berkelic acid 115

2.4.5. Revision of synthetic strategy 130

2.4.6. Synthesis of phosphonate 268 for formal synthesis 131

2.4.7. Coupling of lactone 204 with phosphonates 96 and 268 and cyclisation to form spiroketals 132

2.5. Overall summary and conclusions 139

2.6. Future work 143
Chapter 3: Experimental Procedures

3.1. General details 149

3.2. Synthesis of model tricyclic analogues of berkelic acid 149
   3.2.1. Synthesis of phosphonate coupling partner 96 149
   3.2.2. Synthesis of aldehyde coupling partners 112 - 118 153
   3.2.3. Coupling and cyclisation 158
   3.2.4. Late stage functionalisation of the spiroketal molecules 171

3.3. Synthesis of the tetracyclic core of berkelic acid 171
   3.3.1. Synthesis of phosphonate 149 using TBDMS protecting groups 171
   3.3.2. Synthesis of chiral lactone 175 172
   3.3.3. Model Horner-Wadsworth-Emmons/oxa-Michael cascade 197
   3.3.4. Coupling and completion 199

3.4. Attempted total synthesis of berkelic acid 202
   3.4.1. Synthesis of fully substituted lactone 204 202
   3.4.2. Attempted synthesis of phosphonate 92 for total synthesis of berkelic acid 220
   3.4.3. Synthesis of phosphonate 268 for formal synthesis 231
   3.4.4. Attempted coupling and cyclisation to complete formal synthesis 232

References 239

Appendix

Index for Appendix 245
## Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>λ</td>
<td>wavelength</td>
</tr>
<tr>
<td>δ</td>
<td>chemical shift</td>
</tr>
<tr>
<td>µ</td>
<td>micro</td>
</tr>
<tr>
<td>°C</td>
<td>degrees celcius</td>
</tr>
<tr>
<td>1D</td>
<td>one dimensional</td>
</tr>
<tr>
<td>2D</td>
<td>two dimensional</td>
</tr>
<tr>
<td>Å</td>
<td>angstrom</td>
</tr>
<tr>
<td>Ac</td>
<td>acetyl</td>
</tr>
<tr>
<td>aq.</td>
<td>aqueous</td>
</tr>
<tr>
<td>atm</td>
<td>atmosphere(s)</td>
</tr>
<tr>
<td>b.p.</td>
<td>boiling point</td>
</tr>
<tr>
<td>Bn</td>
<td>benzyl</td>
</tr>
<tr>
<td>br</td>
<td>broad</td>
</tr>
<tr>
<td>cat.</td>
<td>catalytic</td>
</tr>
<tr>
<td>CBS</td>
<td>Corey-Bakshii-Shibata</td>
</tr>
<tr>
<td>CSA</td>
<td>camphorsulfonic acid</td>
</tr>
<tr>
<td>d</td>
<td>doublet</td>
</tr>
<tr>
<td>d.r.</td>
<td>diastereomeric ratio</td>
</tr>
<tr>
<td>DBU</td>
<td>1,8-diazabicyclo[5.4.0]undec-7-ene</td>
</tr>
<tr>
<td>DCC</td>
<td>N,N'-dicyclohexylcarbodiimide</td>
</tr>
<tr>
<td>dd</td>
<td>double of doublet</td>
</tr>
<tr>
<td>ddd</td>
<td>double of double of doublet</td>
</tr>
<tr>
<td>ddt</td>
<td>double of double of triplet</td>
</tr>
<tr>
<td>DEAD</td>
<td>diethyl azodicarboxylate</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Definition</td>
</tr>
<tr>
<td>--------------</td>
<td>------------</td>
</tr>
<tr>
<td>DIBAL-H</td>
<td>diisobutylaluminium hydride</td>
</tr>
<tr>
<td>4-DMAP</td>
<td>N,N-dimethyl-4-aminopyridine</td>
</tr>
<tr>
<td>DME</td>
<td>dimethoxyethane</td>
</tr>
<tr>
<td>DMF</td>
<td>N,N-dimethylformamide</td>
</tr>
<tr>
<td>DMP</td>
<td>Dess-Martin periodinane</td>
</tr>
<tr>
<td>DMSO</td>
<td>dimethyl sulfoxide</td>
</tr>
<tr>
<td>EDC</td>
<td>1-(3-dimethylaminopropyl)-3-ethylcarbodiimide</td>
</tr>
<tr>
<td>e.e.</td>
<td>enantiomeric excess</td>
</tr>
<tr>
<td>EI</td>
<td>electron impact</td>
</tr>
<tr>
<td>ent</td>
<td>enantiomer</td>
</tr>
<tr>
<td>epi</td>
<td>epimer</td>
</tr>
<tr>
<td>eq.</td>
<td>equivalent</td>
</tr>
<tr>
<td>ESI</td>
<td>electrospray ionisation</td>
</tr>
<tr>
<td>et al.</td>
<td>et alii (and others)</td>
</tr>
<tr>
<td>Et</td>
<td>ethyl</td>
</tr>
<tr>
<td>FAB</td>
<td>fast atom bombardment</td>
</tr>
<tr>
<td>g</td>
<td>gram(s)</td>
</tr>
<tr>
<td>h</td>
<td>hour(s)</td>
</tr>
<tr>
<td>HPLC</td>
<td>high pressure liquid chromatography</td>
</tr>
<tr>
<td>HRMS</td>
<td>high resolution mass spectroscopy</td>
</tr>
<tr>
<td>HSQC</td>
<td>heteronuclear single quantum correlation</td>
</tr>
<tr>
<td>HWE</td>
<td>Horner-Wadsworth-Emmons</td>
</tr>
<tr>
<td>HWE/oxa-M</td>
<td>Horner-Wadsworth-Emmons/oxa-Michael</td>
</tr>
<tr>
<td>Hz</td>
<td>hertz</td>
</tr>
<tr>
<td>i.d.</td>
<td>internal diameter</td>
</tr>
<tr>
<td>iPr</td>
<td>isopropyl</td>
</tr>
</tbody>
</table>
IR: infra-red

J: coupling constant

KHMD: potassium hexamethyldisilazide

LiHMDS: lithium hexamethyldisilazide

L: litre

IC₅₀: concentration to inhibit 50% of activity

LDA: lithium diisopropylamide

LTMP: lithium 2,2,6,6-tetramethylpiperidine

lit.: literature

m: meta

M: molar

m: multiplet

m.p.: melting point

m/z: mass to charge ratio

mCPBA: meta-chloroper oxybenzoic acid

Me: methyl

MHz: megahertz

min: minute(s)

mmHg: millimeters mercury

MMP: matrix metalloprotease

mmol: millimole(s)

mol: mole(s)

Ms: methanesulfonyl

MS: molecular sieves

MTBE: methyl tert-butyl ether

n: normal
NBS  \( N \)-bromosuccinimide
NIS  \( N \)-iodosuccinimide
NMO  \( N \)-methylmorpholine-\( N \)-oxide
NMR  nuclear magnetic resonance
nOe  nuclear Overhauser effect
NOESY  nuclear Overhauser effect spectroscopy
\( p \)  para
Ph  phenyl
PHAL  phthalazine
PIFA  bis(trifluoroacetoxy)iodobenzene
PMB  para-methoxybenzyl
ppm  parts per million
PPTS  pyridinium para-toluenesulfonate
q  quartet
quant.  quantitative
R  unspecified alkyl group
rt  room temperature
RCM  ring closing metathesis
\( R_f \)  retention factor
s  singlet
sat.  saturated
\( t \)  \( tert \) (tertiary)
t  triplet
TA  tricyclic analogue
TBAF  tetrabutylammonium fluoride
TBAI  tetrabutylammonium iodide
TBDMS  \textit{tert}-butyldimethylsilyl
TBDPS  \textit{tert}-butyldiphenylsilyl
TBHP  \textit{tert}-butylhydroperoxide
\textit{tBu}  \textit{tert}-butyl
TES  triethylsilyl
temp.  temperature
Tf  triflic
TFA  trifluoroacetic acid
THF  tetrahydrofuran
TLC  thin layer chromatography
TMEDA  \textit{N},\textit{N},\textit{N}',\textit{N}'-tetramethylethane-1,2-diamine
TMS  trimethylsilyl
Ts  toluenesulfonyl
\textit{v}  flow rate
\textit{v}  wavenumber (cm\textsuperscript{-1})
Val  Valine
vol  volume