Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

Synthesis of substrate analogues and inhibitors for phosphoribosyl anthranilate isomerase and indole-3-glycerolphosphate synthase.

Benjamin Joseph Mulchin

2008

Synthesis of substrate analogues and inhibitors for phosphoribosyl anthranilate isomerase and indole-3-glycerolphosphate synthase.

A thesis presented in partial fulfillment of the requirements for the degree of

Doctor of Philosophy
In
Chemistry

At Massey University, Turitea, Palmerston North,
New Zealand.

Benjamin Joseph Mulchin
2008

Abstract

The general biosynthetic pathway for tryptophan is known. However, little information has been gathered on how substrates and enzymes interact when phosphoribosylanthranilate isomerase (PRAI) and indole-3-glycerolphosphate synthase (IPGS) convert a substituted phenyl ring, PRA, into an indole moiety, IGP, *via* 1-(*O*-carboxyphenylamino)-1-deoxyribulose-5-phosphate (CdRP). There has been no serious synthetic approach to develop methodology to produce a plethora of substrate and product analogues of CdRP. The studies described in this thesis cover methodology focusing on secondary aryl amine formation, using reductive amination, nucleophilic substitution and epoxide ring opening, leading to CdRP analogues. Reductive aminations with D-ribose failed to produce any aryl glycosylamine precursor, possibly due to the low nucleophilicity of aryl amines such as aniline. Removing the aromaticity and using cyclohexylamine produced secondary amines in moderate yield in the presence of benzylpentanal, and $NaBH_3CN$, at a pH of 5.5. This led to a successful reductive amination using anthranilate methyl ester. Secondary aryl amine synthesis *via* epoxide ring opening proved consistently reproducible. Using $LiNTf_2$ and high equivalents of cyclohexylamine or aniline in neat conditions opened protected epoxides. This has led to the formation of advanced secondary aryl amine synthons and the development of methodology leading to target compounds with functionality at the 1,2 and 5 positions. Nucleophilic substitution using caesium base, high equivalents aniline at room temperature, gave a moderate yield of secondary aryl amines from sulfonyl and bromide good leaving groups. Raising the reaction temperature improved yields using low equivalents of aniline, with the optimal temperature being 50 °C. Ultimately using both the high equivalents of aniline or anthranilate methyl ester and warming the reaction in DMF gave the highest yields of secondary aryl amines. No overalkylated tertiary amine was isolated when a caesium base was used. Boc *N*-protection of 1-phenylamino-4-pentene and asymmetric dihydroxylation gave the corresponding diol, which was phosphorylated giving the protected target 1,4,5 compound. The methodology leading to the protected target 1,4,5 compound synthesis provides a means to the synthesis additional of CdRP analogues.

Acknowledgements

I would like to thank my chief supervisor Emily Parker for all the guidance, advice and motivation she has given me throughout my thesis. Research and learning was enjoyable through Emily’s personal approach.

I would like to thank my co-supervisor Geoff Jameson for the help, brain teasing discussions and guidance throughout my Ph.D. I would especially like to thank the Marsden grant for funding me and Massey University and the IFS department for this opportunity.

I would like to thank Matthias Rost for the insightful discussions and helpfulness in the lab. Thank-you Linley Schofield (best chocolate cake) and Carol Taylor for streamlining my ideas and exposing me to different forms of science. To the Shikimate group ladies, Meek-yung, Amy, Celia and Jenness thanks for the latest gossip and fun times. To the Shikimate group gentleman, Aidan, Andy and Leonardo, thank-you for keeping the lab a fun place to learn. Thanks Scott for an avenue to bounce ideas off.

I am particularly grateful also to all my colleges at the Institute of Fundamental Sciences including: Patrick Edwards, Carl Otter, Bill Williams, Adrian Jull, David Harding, William Barker, Renaud Hardré, Pavel Krist, Julie Hardie, Eric Ainscough, David Lun, Fiona Cochrane, Bob Parsons, Kristin, Julia, Chantelle, Hemi, Simon, and Jeff .

Thank-you to Gareth, Keiran, Chetan and Jayesh who kept my interest in things other than chemistry alive when chemistry threatened to engulf everything. A humble thank-you to the girls who found my laptop on the bus and gave it back. Justine thank-you for your support and drive to better yourself and me. Thank-you Kat for still being with me for all those countless nights I’ve woken you and for those supportive pushes and proof readings.

To my Grandparents, thank-you for your encouragement and reassurance that everything will end up all right. Mum and Dad without your support this thesis would have been infinitely more difficult. Your unconditional love has provided many a soft landing after the chemistry has been hard and a simple thank-you is not enough.

Table of contents

Abbreviations **xi**
Index of figures **xv**
Index of schemes **xvi**
Index of tables **xxii**
Index of graphs **xxiii**

CHAPTER 1: Tryptophan biosynthesis. **Page**

1.1. Thesis introduction: **1**
    1.1.2. Roles of tryptophan and other aromatic amino acids: 1
1.2. Pathways: **2**
    1.2.1. Shikimate pathway: 2
    1.2.2. Biosynthesis of tryptophan: 4
1.3. Enzyme structure basics of PRAI and IGPS: **7**
    1.3.1. Introduction: 7
    1.3.2. Sources of PRAI and IGPS: 7
    1.3.3. PRAI and IGPS enzymes: 7
1.4. Biochemical details of PRAI: **8**
    1.4.1. Enzyme kinetics for PRAI: 8
    1.4.2. Proposed mechanism catalysed by PRAI – The Amadori rearrangement: 10
    1.4.3. X-ray structural analysis of product analogue rCdRP crystallized in *t*PRAI: 11
1.5. Biochemical details of IGPS: **12**
    1.5.1. Enzyme kinetics for IGPS: 12
    1.5.2. Proposed mechanism catalysed by IGPS – indole ring formation: 14
    1.5.3. Residue interactions and model studies of intermediates of indole ring formation: 16
    1.5.4. Reaction catalysed by IGPS – indole ring formation: 17

1.6. Previous synthetic research on PRA and CdRP: 18

1.6.1. PRA synthesis: 18

1.6.2. Synthesis of PRA derivatives: 19

1.6.3. CdRP synthesis: 20

1.6.4. Synthesis of CdRP derivatives: 21

1.6.5. rCdRP synthesis: 22

1.7. Target compounds: 23

1.7.1. PRA analogue targets for the investigation of PRAI reaction: 23

1.7.2. CdRP analogue targets for the investigation of PRAI and IGPS: 24

1.7.3. Target 1,2,5 compounds for the investigation of the PRAI and IGPS: 26

1.7.4. Target 1,4,5 compounds for the investigation of the PRAI and IGPS: 27

1.8. Synthetic ideas: 28

1.8.1 Synthetic goals: 28

1.8.2. Retrosynthesis of CdRP like compounds: 28

1.9. Formation of secondary amines: 29

1.9.1. Introduction: 29

1.9.2. Condensation: 30

1.9.3. *N*-alkylation *via* nucleophilic substitution: 31

1.9.4. Nucleophilic substitution with metal coordination: 34

1.9.5. Epoxide ring opening: 35

1.9.6. Imine formation: 36

1.9.7. Solid phase synthesis: 37

1.10. Summary of thesis aims: 39

CHAPTER 2: Reductive aminations.

2.1. The initial retrosynthetic plan: 40

2.1.2. Phosphorylation introduction: 41

2.1.3. Literature background on phosphorylations: 43
2.1.4. Literature deprotection of phosphate esters: 46
2.2. Phosphorylation of D-ribonolactone: 47
2.3. D-Ribonolactone modifications: 49
2.3.1. D-Ribonolactone protection literature background: 49
2.3.2. D-Ribonolactone protections: 49
2.3.3. Di-*iso*-butylaluminum hydride reductions: 52
2.3.4. Literature background of Dibal-H reductions and silyl ethers: 53
2.3.5. Alternative D-ribonolactone protections: 57
2.3.6. Benzylidene and *iso*-propylidene acetal D-ribonolactone protections: 62
2.4. D-Ribose modifications: 66
2.5. Esterification of anthranilic acid: 69
2.6. Introduction to reductive aminations: 71
2.6.1. Reductive amination using metal catalysis: 71
2.6.2 Reductive amination using hydride reducing agents: 73
2.7. Reductive aminations of lactols: 76
2.7.1. Introduction: 76
2.7.2. Literature background to the reductive aminations of lactols: 76
2.7.3. Reductive aminations of D-ribose with aryl amines: 78
2.8. Properties of aryl amines: 82
2.8.1. pK_a: 82
2.8.2. The influence on pK_a by substituents: 83
2.9. Model reductive amination chemistry: 85
2.9.1. Introduction: 85
2.9.2. Preparation of aldehyde: 86
2.9.3. Model reductive aminations with phosphate present: 87
2.9.4. Preparation of protected aldehyde: 89
2.9.5. Model reductive aminations with protected aldehyde: 90

2.9.6. Model reductive aminations with protected aldehyde and anthranilate methyl ester: 92

2.10. Conclusions: 93

CHAPTER 3: Nucleophilic substitution of good leaving groups.

3.1. Synthesis of rCdRP-like target compounds: 96

3.1.2. Synthesis of secondary rCdRP-like aryl amines: 97

3.1.3. Proposed synthesis of secondary rCdRP-like aryl amines: 102

3.2. Synthesis of sulfonyl good leaving groups: 104

3.2.1. Introduction: 104

3.2.2. Preparation of sulfonyl good leaving groups on pentan-1,5-diol: 105

3.2.3. Preparation of sulfonyl good leaving groups on 4-penten-1-ol: 109

3.3. *N*-nucleophilic substitution of sulfonyl good leaving groups: 110

3.3.1. Introduction to *N*-nucleophilic substitution: 110

3.3.2. Nucleophilic substitution of sulfonyl good leaving groups on pentan-1,5-diol by aniline: 112

3.3.3. Nucleophilic substitution of sulfonyl good leaving groups on 4-penten-1-ol by aniline: 114

3.4. *N*-nucleophilic substitution of bromide moieties: 115

3.4.1. Preparation of bromide moieties from pentan-1,5-diol and 4-penten-1-ol: 115

3.4.2. *N*-nucleophilic substitution of 5-*O*-*para*-methoxybenzyl bromopentanol and bromo-4-pentene: 118

3.4.3. Synthesis of a target 1,4,5 compound *N*-*tert*-butyloxycarbonyl-1-phenylamino-5-diethyl phosphate-4-pentanol: 121

3.5. Nucleophilic substitution of *O*- and *C*-nucleophiles: 127

3.5.1. Substitution of good leaving groups on

pentan-1,5-diol moieties by *O*- and *C*-nucleophiles: 127
3.5.2. Attempted synthesis of target 1,4,5 compound using a *C*-nucleophile: 130
3.6. Conclusions: 132

CHAPTER 4: Epoxide ring opening.

4.1. Introduction: 135
4.1.2. Retrosynthetic plan: 135
4.2. Literature on epoxide ring formation: 136
4.2.1. Good leaving groups: 136
4.2.2. Epoxidation from alkenes: 137
4.3. Formation of protected epoxide: 137
4.3.1. 1-*O*-*tert*-Butyldimethylsilyl-4,5-epoxypentanol: 139
4.3.2. 1-*O*-*tert*-Butyldiphenylsilyl-4,5-epoxypentanol: 141
4.4. Literature on epoxide ring opening: 143
4.5. Epoxide ring opening: 146
4.5.1. Ring opening of 1-*O*-*tert*-butyldimethylsilyl-4,5-epoxypentanol: 146
4.5.2. Ring opening of 1-*O*-*tert*-butyldiphenylsilyl-4,5-epoxypentanol: 152
4.6. Conclusions: 152

CHAPTER 5: Summary of thesis.

5.1. Overall conclusions: 154
5.1.2. Conclusions of reductive amination chemistry: 154
5.1.3. Conclusions of nucleophilic substitution of good leaving groups: 160
5.1.4. Conclusions of epoxide ring opening methodology: 169
5.2. Possible future experiments: 171

CHAPTER 6: Experimental.

6.1. General Methods: **173**

6.1.2. Reagents and solvents: 173

6.1.3. Synthetic methods: 175

6.1.4. Chromatography: 175

6.1.5. Characterisation: 176

6.2. Experiments described in Chapter Two: **177**

5-*O*-Triphenylmethyl-D-ribonolactone **2.09**: 177

2,3-Bis-*O*-*tert*-butyldimethylsilyl-5-*O*-triphenylmethyl-D-ribonolactone **2.12**: 178

2-*O*-*tert*-Butyldiphenylsilyl-5-*O*-triphenylmethyl-D-ribonolactone **2.28**: 179

2,3-*O*-Dibenzyl-5-*O*-triphenylmethyl-D-ribonolactone **2.35**: 180

2-*O*-Benzyl-5-*O*-triphenylmethyl-D-ribonolactone **2.36**: 181

2,3-*O*-*iso*-Propylidene-D-ribono-1,4-lactone **2.44**: 182

3,4-*O*-*iso*-Propylidene-D-ribono-1,5-lactone **2.45**: 182

Methyl-2,3-*O*-*iso*-propylidene-*β*-D-ribofuranoside **2.48**: 183

Methyl-2,3-*O*-*iso*-propylidene-5-*O*-benzyl-*β*-D-ribofuranoside **2.49**: 184

Methyl-2,3-*O*-*iso*-propylidene-5-*O*-diphenylphosphate-*β*-D-ribofuranoside **2.50**: 185

Anthranilate methyl ester **2.51**: 186

5-Diphenyl phosphate pentanol **2.72**: 187

5-Diphenyl phosphate pentanal **2.73**: 188

5-*O*-Benzylpentanol **2.76**: 189

5-*O*-Benzylpentanal **2.77**: 189

5-*O*-Benzylpentylcyclohexylamine **2.83**: 190

5-*O*-Benzylpentyl anthranilate methyl ester **2.85**: 191

6.3. Experiments described in Chapter Three: **192**

(2*S*,3*R*,4*R*)-3,4-Bis-*O*-*tert*-butyldimethylsilyl-1-*O*-triphenylmethyl tetrahydrofuran **3.06**: 192

3,4-Bis-*O*-*tert*-butyldimethylsilyl-2,5-bis-*O*-toluenesulfonyl-1-*O*-triphenylmethyl-D-ribitol **3.08**: 193
(2*S*,3*R*,4*S*)-*N*-5-Anthranilate methyl ester-3,4-bis-*O*-*tert*-butyldimethylsilyl-1-*O*-triphenylmethyl **3.10**: 194
5-*O*-*tert*-Butyldiphenylsilyl-D-ribonolactone **3.14**: 195
5-Tetrahydropyranyloxy pentanol **3.21**: 196
5-*O*-*tert*-Butyldimethylsilyl pentanol **3.23**: 197
5-*O*-*tert*-Butyldimethylsilyl-1-*O*-methanesulfonyl pentanol **3.24**: 197
5-*O*-*tert*-Butyldimethylsilyl-1-*O*-toluenesulfonyl pentanol **3.25**: 198
4-Pentenyl methanesulfonyl **3.26**: 199
4-Pentenyl toluenesulfonyl **3.27**: 199
5-*O*-*tert*-Butyldimethylsilyl-1-phenylamino-pentane **3.37**: 200
5-*O*-*tert*-Butyldimethylsilyl-1-*O*-trifluoro-methanesulfonyl pentanol **3.38**. 201
1-Phenylamino-4-pentene **3.39**: 202
Bromopentan-5-ol **3.40**: 203
5-*O*-*para*-Methoxybenzyl bromopentanol **3.42**: 203
Bromo-4-pentene **3.43**: 204
5-*O*-*para*-Methoxybenzyl-1-phenylamino-pentane **3.44**: 205
5-*O*-*para*-Methoxybenzyl pentylcyclohexylamine **3.46**: 206
Anthranilate methyl ester-4-pentene **3.47**: 207
N-9-Fluorenylmethoxycarbonyl-1-phenylamino-4-pentene **3.48**: 208
N-9-Fluorenylmethoxycarbonyl-1-phenylamino-4,5-pentan-diol **3.54**: 209
N-*tert*-Butyloxycarbonyl-1-phenylamino-4-pentene **3.55**: 210
N-*tert*-Butyloxycarbonyl-1-phenylamino-4,5-pentan-diol **3.56**: 211
N-*tert*-Butyloxycarbonyl-1-phenylamino-5-diethyl phosphate-4-pentanol **3.57**: 212
N-*Tert*-butyloxycarbonyl-1-phenylamino-4-diethyl phosphate-5-pentanol **3.58**: 212
5-*O*-*tert*-Butyldimethylsilyl-1-phenoxy-pentane **3.60**: 213
5-*O*-*para*-Methoxybenzyl-1-phenoxy-pentane **3.61**: 214
1-Phenoxy-pentanol **3.63**: 215

2-(5-Hydroxy-pentyl)-phenol **2.64**: 215
5-*O*-*tert*-Butyldimethylsilyl-1-phenyl-hexane **3.66**: 216
Hex-5-enylbenzene **3.67**: 217
5,6-Epoxyhexylbenzene **3.68**: 217
6.4. Experiments described in Chapter Four: 218
1-*O*-*tert*-Butyldimethylsilyl-4-penten-1-ol **4.11**: 218
1-*O*-*tert*-Butyldiphenylsilyl-4-penten-1-ol **4.17**: 219
1-*O*-*tert*-Butyldimethylsilyl-4,5-epoxypentanol **4.12**: 220
1-*O*-*tert*-Butyldiphenylsilyl-4,5-epoxypentanol **4.16**: 220
5-*O*-Benzylpentene **4.18**: 221
5-*O*-*tert*-Butyldimethylsilyl-1-phenylamino-pentan-2-ol **4.31**: 222
5-*O*-*tert*-Butyldimethylsilyl-1-cyclohexylamino-pentan-2-ol **4.33**: 223
5-*O*-*tert*-Butyldiphenylsilyl-1-phenylamino-pentan-2-ol **4.34**: 224

References: 225

Abbreviations

Ac	Acetyl
ACE-Cl	α-Chloroethyl chloroformate
AD-mix	Asymmetric dihydroxylation reagent
Aq	Aqueous
Arg	Arginine
Asn	Asparagine
Asp	Aspartic acid
BINAP	2,2’-Bis(diphenylphosphino)-1,1’-binaphthyl
bmim	1-Butyl-3-methylimidazolium
Bn	Benzyl
Boc	*tert*-Butyloxycarbonyl
BTP	1,3-Bis(tris(hydroxymethyl)methylamino)propane
Bz	Benzoyl

CdRP	1-(*O*-Carboxyphenylamino)-1-deoxyribulose-5-phosphate
$^1H/^1H$ COSY	Proton correlation spectroscopy
m-CPBA	*meta*-Chloroperoxybenzoic acid
CSA	Camphorsulfonic acid
Cys	Cysteine
DAHP	3-Deoxy-D-*arabino*-heptulosonate 7-phosphate
DAH7P	3-Deoxy-D-*arabino*-heptulosonate 7-phosphate
DCP	1,2-Dichloropropane
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
DEAE	Diethylamino ethanol
DHP	3,4-Dihydro-2*H*-pyran
DHQ	Dehydroquinate
$(DHQ)_2$-PHAL	Dihydroquinine phthalazine
DHS	Dehydroshikimate
Diab-H	Disiamylborane
Dibal-H	Di-*iso*-butylaluminum hydride
DIEA	Di-*iso*-propylethylamine
DMAP	4-Dimethylaminopyridine
DMF	*N*,*N*-Dimethylformamide
DMP	2,2-Dimethoxypropane
DMP	Dess-Martin periodinane
DMSO	Dimethyl sulfoxide
*e*IGPS	*Escherichia coli* IGPS
*e*PRAI	*Escherichia coli* PRAI
E4P	D-Erythrose 4-phosphate
EDG	Electron-donating groups
EDTA	Ethylenediamine tetra-acetic acid (di-sodium salt)
EPSP	5-*Enol*pyruvyl-shikimate 3-phosphate
Eq	Equivalent
EWG	Electron-withdrawing groups
Fmoc	9-Fluorenylmethoxycarbonyl

FT	Fourier transform
GL	Good leaving group
Glu	Glutamic acid
His	Histidine
HMPA	Hexamethylphosphoric triamide
HREIMS	High Resolution Electron Impact Mass Spectrometry
$^1H/^{13}C$ HMQC	Heteronuclear multiple quantum coherence
IEX	Ion exchange chromatography
IGPS	Indole-3-glycerolphosphate synthase
Ile	Isoleucine
im	Imidazole
k_{cat}	Catalytic rate/turnover number
k_{cat}/K_M	Catalytic efficiency
KDO8P	3-Deoxy-D-*manno*-octulosonate 8-phosphate
K_i	Enzyme inhibitor affinity
K_M	Michaelis constant
K_p	Enzyme product affinity
L. Pet.	Light petroleum
Leu	Leucine
Lys	Lysine
MEM	β-Methoxyethoxymethyl
4 Å M.S.	4 Å Molecular sieves
MW	Molecular weight
Ms	Mesyl, methanesulfonate
N. P.	No product
NMP	*N*-Methyl pyrrolidone
NMR	Nuclear magnetic resonance
Nu	Nucleophile
PB	Pyridine-borane
PEP	Phospho*enol*pyruvate

Ph	Phenyl
Phe	Phenylalanine
pK_a	Acid dissociation constant
PMB	*para*-Methoxybenzyl
PMHS	Polymethylhydrosiloxane
PPG	Primary protecting groups
ppm	Parts per million
Pro	Proline
PRAI	Phosphoribosyl anthranilate isomerase
Psi	Pounds per square inch
py	Pyridine
rCdRP	1-(*O*-Carboxyphenylamino)-1-deoxyribose-5-phosphate
R_f	Retardation factor/retention factor
R5P	D-Ribose 5-phosphate
RT	Room temperature
*s*IGPS	*Sulfolobus solfataricus* IGPS
Ser	Serine
*t*IGPS	*Thermotoga maritima* IGPS
*t*PRAI	*Thermotoga maritima* PRAI
TBAB	Tetrabutylammonium bromide
TBAF	Tetrabutylammonium fluoride
TBAI	Tetrabutylammonium iodide
TBDMS	*tert*-Butyldimethylsilyl
TBDPS	*tert*-Butyldiphenylsilyl
TBHP	*tert*-Butyl hydroperoxide
Tf	Trifluoromethane sulfonate
TFA	Trifluoroacetyl
THCP	Tributylammonium hydrobenzoin cyclic phosphate
THF	Tetrahydrofuran
THP	Tetrahydropyran
TLC	Thin layer chromatography

TMS	Tetramethylsilane
TMSBr	Trimethylsilyl bromide
Tr	Trityl, triphenyl methyl
Trp	Tryptophan
Ts	Tosyl, toluene-sulfonate
*p*TSA	*para*-Toluenesulfonic acid
Tyr	Tyrosine
UV	Ultraviolet
V_{max}	Maximum reaction velocity
*y*PRAI	*Saccharomyces cerevisiae* PRAI

Index of figures

Figure		**Page**
1.01	Aromatic amino acids.	2
1.02	Stereoview of substrate CdRP bound to the active site of *s*IGPS.	14
1.03	Stereoview of the bound catalytic intermediate 1 (a) and intermediate 2 (b) from molecular models of the active site in *s*IGPS.	16
1.04	Ring analogue target compounds for potential inhibition of PRAI.	24
1.05	PRA **1.05** carbohydrate analogues target compounds for potential inhibition of PRAI.	24
1.06	rCdRP carbohydrate-analogue target compounds for potential inhibition of PRAI and IGPS.	25
1.07	Ring analogue target compounds for product inhibition of PRAI.	26
1.08	Ring analogue target compounds for PRAI.	27
1.09	Target 1,4,5 compounds for inhibition of PRAI and IGPS.	28
2.01.	Mono-protected product.	47
2.02.	Resonance structures for aniline **2.65**, lowering pK_a.	83
2.03.	Effects of steric hindrance by groups on the phenyl ring.	84
2.04.	Primary and secondary amines.	85

2.05. Tertiary amine by-product **2.84**. 92
3.01. Selective removal of trityl groups in the presence of acetals. 102
3.02. Chiral ligand of AD-mix-α. 123
4.01. Target 1,2,5 compounds. 135
4.02. Jacobsen's catalyst[333-335] **4.13**, a cobalt-based salen ligand complex. 139
6.01. Chiral ligand of AD-mix-α. 175

Index of schemes

Scheme **Page**
1.01 The shikimate pathway. 3
1.02 Chorismate, the precursor to the aromatic amino acids and aromatic compounds. 4
1.03 Formation of anthranilate **1.02** *via* bicyclic derivative **1.03**. 4
1.04 Formation of anthranilate **1.02** *via* Michael-type addition and loss of pyruvate. 5
1.05 Tryptophan biosynthesis. 6
1.06 Plausible mechanism of the Amadori rearrangement forming CdRP **1.06**. 11
1.07 Indole ring formation showing substrate interactions with Lys 53, Lys 110, and Glu 159 from *s*IGPS. 15
1.08 First synthesis of PRA **1.05** by Creighton.[42] 18
1.09 Berger *et al*.[48] synthesis of ribosyl aniline **1.09**. 19
1.10 Synthesis of dephosphorylated PRA **1.10**. 20
1.11 Heat-induced synthesis of CdRP **1.06**. 20
1.12 Synthesis of 1-(*O*-carboxyphenylamino)-1-deoxyribulose **1.11**. 21
1.13 Inhibition of IGPS by 1-(phenylamino)-1-deoxyribulose 5'-phosphate **1.12**. 22
1.14 Successful reduction of CdRP **1.06** to rCrRP **1.13**. 23
1.15 Retrosynthesis of CdRP-like compounds. 29
1.16 Retrosynthesis of secondary amines. 30
1.17 Okahara *et al*.[74] *N*-alkylation to form aryl secondary amine **1.29**. 32

1.18 Example of Ullmann[269] and Goldberg[270] couplings by Buchwald *et al.*[77] 33
1.19 General scheme of palladium-catalysed secondary amine formation. 35
1.20 Glycosylamine formation *via* an epoxide. 36
1.21 Imine formation and subsequent formation of secondary amine. 37
1.22 General synthesis of secondary amine *via* resin **1.40**. 38
2.01 General synthetic strategy. 41
2.02 Phosphorylation route options. 42
2.03 Carbohydrate phosphate formation by use of diphenyl phosphorochloridate at room temperature (RT). 44
2.04 Phosphorylation by triethyl phosphate and iodine. 44
2.05 Example of electrophilic phosphorylation. 45
2.06 Phosphorylation of C5-hydroxyl using the phosphoramidite method. 46
2.07 Failed synthesis of D-ribonolactone-5-diphenyl phosphate **2.07**. 48
2.08 D-Ribonolactone modification from Taylor *et al.*[134] 49
2.09 Primary tritylation of D-ribonolactone. 50
2.10 Formation of intermediate **2.10** from catalytic DMAP. 50
2.11 Disilylation of trityl **2.09**. 51
2.12 Retrosynthesis for the reductive amination of lactol. 51
2.13 Dibal-H reduction of disilyl protected lactone **2.12**. 52
2.14 Desilylation by Dibal-H. 53
2.15 Dibal-H reduction by Ley *et al.*[144] 53
2.16 Dibal-H reduction by Ireland and Wilcox.[145] 53
2.17 Dibal-H ester reduction by Pamies and Backvall.[146] 54
2.18 Dibal-H acetate reduction by Yamada *et al.*[147] 54
2.19 Dibal-H acetal reduction by Iqbal *et al.*[148] 55
2.20 Dibal-H desilylation by Xu and Newcomb.[151] 55
2.21 Dibal-H reduction in the presence of TBDPS. 56
2.22 Dibal-H reduction for benzyl-protected lactone **2.26**. 56
2.23 Diab-H reduction for benzyl-protected lactone **2.26**. 57
2.24 Diab-H preparation. 57
2.25 TBDPSCl protection of diol **2.09**. 58

2.26 Lactone benzylation using BnBr and NaH. 58
2.27 *Cis* diol benzylation using BnBr and Ag_2O. 59
2.28 Reaction 5 - the best conditions for the formation of dibenzylated lactone **2.35**. 60
2.29 Attempts at tribenzylating D-ribonolactone **2.03**. 62
2.30 Potential benzylidene acetal products. 63
2.31 Unprotected C5 hydroxyl group leads to acid-catalysed rearrangement. 63
2.32 Dibal-H reduction of *iso*-propylidene acetal D-ribonolactone moieties. 64
2.33 Potential *iso*-propylidene acetal products 64
2.34 Retrosynthesis of protected lactol *via* D-ribose methyl glycoside. 66
2.35 One-step formation of 2,3-*O*-*iso*-propylidene-β-D-ribofuranoside **2.48** from D-ribose **2.47**. 67
2.36 Primary hydroxyl group benzylation. 67
2.37 Primary hydroxyl group phosphorylation. 68
2.38 Methyl esterification of anthranilic acid. 69
2.39 Methyl esterification of 4-aminobenzoic acid. 70
2.40 Esterification *via* isolated acid chloride. 71
2.41 Imine formation and subsequent formation of secondary amine. 72
2.42 Platinum oxide use in indirect reductive amination. 73
2.43 Retrosynthesis of potential inhibitors *via* D-ribose **2.47**. 76
2.44 Glycosylamine formation using D-ribose **2.47**. 77
2.45 Glycosylamine **2.64** formation using anthranilate ethyl ester. 77
2.46 Parameters used in reductive aminations involving D-ribose **2.47**. 78
2.47 Aldehyde equilibrium and formation of the imine **2.68** before reduction to the aryl glycosylamine precursor **2.66**. 81
2.48 Retrosynthetic pathway of 5-phosphonopentyl anthranilic acid **2.74**. 86
2.49 Synthesis of mono phosphorylated product **2.72**. 87
2.50 Synthesis of aldehyde **2.73**. 87
2.51 Attempted synthesis of 5-phosphonopentyl anthranilate methyl ester **2.75**. 88
2.52 Synthesis of benzyl-protected product **2.76**. 89
2.53 Synthesis of protected aldehyde **2.77**. 89

2.54 Unsuccessful synthesis of benzyl-protected pentylaminophenyl **2.78**. 90
2.55 Harrison's[279] attempts at synthesis of benzyl-protected hexylamino-acetic ethyl ester **2.81**. 91
2.56 Successful synthesis of benzyl-protected pentylcyclohexyl amine **2.83**. 92
2.57 Successful synthesis of benzyl-protected pentyl anthranilate methyl ester **2.85**. 93
2.58 Formation of benzyl-protected pentyl anthranilate methyl ester **2.85** over three steps. 95
3.01 Literature synthesis by Taylor *et al*.[134] 96
3.02 General retrosynthetic strategy for CdRP-like compounds. 97
3.03 Initial attempts at forming a primary-mono good-leaving group. 98
3.04 Synthesis of bis-mesyl **3.07** and bis-tosyl **3.08** compounds. 99
3.05 Lack of aryl amine reactivity at room temperature. 99
3.06 Nucleophilic substitution forming pyrrolidine derivative **3.10**. 100
3.07 Synthesis of precursor towards formation of a primary good leaving group. 101
3.08 Formic acid removal of trityl group by Taylor *et al*.[134] 101
3.09 Attempted removal of trityl group. 101
3.10 Synthesis of 5-*O*-*tert*-butyldiphenylsilyl-D-ribonolactone **3.14**. 103
3.11 Proposed alternative strategy to primary good leaving group **3.18**. 103
3.12 Retrosynthetic pathway for preparation of secondary aryl amines from pentan-1,5-diol **3.19**. 104
3.13 Retrosynthetic pathway for target 1,4,5 compounds from 4-penten-1-ol **3.20**. 105
3.14 Pentan-1,5-diol **3.19** protection with DHP in hexanes. 106
3.15 Silylation of pentan-1,5-diol **3.19** using NaH. 106
3.16 Mesylation of mono silyl **3.23**. 108
3.17 Tosylation of mono silyl **3.23**. 108
3.18 Formation of good leaving groups on 4-penten-1-ol **3.20**. 109
3.19 Protection/deprotection strategy leading to the synthesis of secondary amine **3.29**. 110

3.20 Chemoselectivity of secondary amine **3.34** by caesium base. 111
3.21 Use of caesium carbonate and low equivalents of aniline **3.36** in the synthesis of secondary aryl amine **3.37**. 112
3.22 Synthesis of 5-*O*-*tert*-butyldimethylsilyl-1-*O*-trifluoro-methanesulfonyl pentanol **3.38**. 113
3.23 Use of higher equivalents of aniline **3.36** in the synthesis of secondary aryl amine **3.37**. 113
3.24 Use of low equivalents of aniline **3.36** in the synthesis of secondary aryl amine **3.39**. 114
3.25 Neat aniline **3.36** in the synthesis of secondary aryl amine **3.39**. 115
3.26 Synthesis of 5-*O*-*para*-methoxybenzyl bromopentanol **3.42** from pentan-1,5-diol **3.19**. 116
3.27 Synthesis of bromo-4-pentene **3.43** by PBr_3 and pyridine. 117
3.28 Secondary amine **3.46** synthesis using low equivalents of cyclohexylamine **3.45**. 118
3.29 Synthesis of secondary aryl amine **3.44** *via* heating. 119
3.30 Synthesis of 1-phenylamino-4-pentene **3.39** using heat and high equivalents of aniline **3.36**. 120
3.31 Synthesis of anthranilate methyl ester-4-pentene **3.47** using heat and high equivalents of anthranilate methyl ester **3.09**. 121
3.32 Synthesis of *N*-9-fluorenylmethoxycarbonyl -1-phenylamino-4-pentene **3.48**. 122
3.33 Overview of asymmetric dihydroxylation using either AD-mix-α or β. 122
3.34 Catalytic cycle of the AD reaction with $K_3Fe(CN)_6$ as the cooxidant. 123
3.35 Synthesis of *N*-9-fluorenylmethoxycarbonyl-1-phenylamino -4,5-pentan-diol **3.54** using AD-mix-α. 124
3.36 Synthesis of *N*-*tert*-butyloxycarbonyl-1-phenylamino -4-pentene **3.55** using a heterogeneous system and NaOH. 125
3.37 Synthesis of *N*-*tert*-butyloxycarbonyl-1-phenylamino -4,5-pentan-diol **3.56** using AD-mix-α. 126
3.38 Synthesis of *N*-*tert*-butyloxycarbonyl-1-phenylamino

-5-diethyl phosphate-4-pentanol **3.57**. 126
3.39 Substitution of mesyl **3.24** by phenol **3.59** at room temperature. 127
3.40 Substitution of bromide **3.42** by phenol **3.59** at room temperature. 128
3.41 Mixture of products formed when phenol **3.59** substituted bromopentan-5-ol **3.40**. 129
3.42 Substitution of mesyl **3.24** with benzyl magnesium bromide **3.65**. 130
3.43 Synthesis of hex-5-enylbenzene **3.67** *via* substitution of 4-pentenyl methanesulfonyl **3.26** with benzyl magnesium bromide **3.65**. 131
3.44 Synthesis of 5,6-epoxyhexylbenzene **3.68** by *m*-CPBA. 131
3.45 Epoxide ring opening by inorganic phosphate described by Bolte *et al*.[325] 131
3.46 Attempted ring opening of 5,6-epoxyhexylbenzene **3.68** with inorganic phosphate. 132
3.47 Synthesis of a protected target 1,4,5 compound **3.57** over 5 steps. 134
4.01 General retrosyntheic strategy. 136
4.02 Epoxide formation from good leaving group. 136
4.03 4,5-Epoxypentan-1-ol formation from *m*-CPBA. 137
4.04 4,5-Epoxypentan-1-ol formation from oxaziridinium salt. 137
4.05 Protected epoxide **4.12** formation by Yang *et al*.[331] 138
4.06 Hydrolytic kinetic resolution of racemic epoxide **4.12**. 138
4.07 Synthesis of protected epoxide **4.12**. 140
4.08 Synthesis of 1-*O*-*tert*-butyldiphenylsilyl-4,5-epoxypentanol **4.16**. 142
4.09 Synthesis of 5-*O*-benzylpentene **4.18**. 142
4.10 Lewis acid catalytic cyclic for epoxide ring opening. 144
4.11 Parameters to form secondary aryl amine **4.26**. 145
4.12 Parameters to form secondary aryl amine **4.28.** 145
4.13 Epoxide ring opening by refluxing aniline. 146
4.14 Parameters for epoxide ring opening by aryl amines. 146
4.15 Low-yielding opening of TBDMS-protected epoxide **4.12** using aniline **4.24**, and $ZnCl_2$ in refluxing MeCN. 148
4.16 Cyclohexylamine **4.32** opening of the TBDMS-protected epoxide **4.12**. 149
4.17 Aniline **4.24** opening of TBDMS-protected epoxide **4.12**. 150

4.18 Aniline **4.24** opening of TBDPS-protected epoxide **4.16**. 152
4.19 Formation of secondary aryl amine synthon **4.31** over three steps. 153
5.01 Failed Dibal-H reduction of disilyl protected lactone **2.12**. 155
5.02 Parameters used in reductive aminations involving D-ribose **2.47**. 157
5.03 Hirota *et al.*[252] glycosylamine formation using D-ribose **2.47**. 158
5.04 Synthesis of 5-*O*-benzylpentyl anthranilate methyl ester **2.85**. 159
5.05 Formation of the furan derivative **3.06**, after attempting to synthesise a sulfonyl leaving group on diol **3.02** primary hydroxyl group. 161
5.06 Nucleophilic substitution forming pyrrolidine derivative **3.10**. 162
5.07 Use of caesium carbonate and low equivalents of aniline **3.36** in the synthesis of secondary aryl amine **3.37**. 163
5.08 Neat aniline **3.36** in the synthesis of secondary aryl amine **3.39**. 164
5.09 Synthesis of secondary aryl amine **3.44** *via* heating. 165
5.10 Synthesis of anthranilate methyl ester-4-pentene **3.47** using heat and high equivalents of anthranilate methyl ester **3.09**. 166
5.11 Synthesis of a protected target 1,4,5 compound **3.57**. 168
5.12 Substitution of bromide **3.42** by phenol **3.59** at room temperature. 168
5.13 Attempted ring opening of 5,6-epoxyhexylbenzene **3.68** with inorganic phosphate. 169
5.14 Formation of secondary aryl amine synthon **4.31** over three steps. 170
5.15 Proposed alternative strategy to primary good leaving group **3.18**. 172

Index of tables

Table **Page**
1.01 Enzyme kinetics of PRAI enzymes. 10
1.02 Enzyme kinetic parameters of IGPS enzymes. 13
2.01 Conditions used in benzylation reactions. 59
2.02 Conditions used in *iso*-propylidene acetal reactions. 65
2.03 Conditions used in esterification reactions. 70
2.04 Conditions used in lactol reductive amination reactions. 79
2.05 $\mathrm{p}K_\mathrm{a}$ of different potential nucleophiles at 25 °C. 82

3.01 Conditions used in silylation of pentan-1,5-diol **3.19** using NaH. 107
3.02 Conditions used in silylation of pentan-1,5-diol **3.19** using imidazole. 108
3.03 Conditions used in the synthesis of bromopentan-5-ol **3.40**. 116
3.04 Conditions used in the synthesis of secondary aryl amine **3.44**. 119
3.05 Conditions used in the synthesis of 1-phenylamino-4-pentene **3.39**. 120
4.01 Conditions used in silylation reactions. 140
4.02 Conditions used in epoxidation reactions. 141
4.03 Reactions using low equivalents of aryl amine at room temperature to open the TBDMS-protected epoxide **4.12**. 147
4.04 Reactions using low equivalents of aryl amine at reflux to open the protected epoxide **4.12**. 149
4.05 Reactions using high equivalents of neat aryl amine at room temperature to open the TBDMS-protected epoxide **4.12**. 151

Index of graphs

Graph **Page**

4.01 ^{1}H NMR spectrum of secondary aryl amine **4.31** between 3.0 and 7.15 ppm. 150