TOWARDS THE TOTAL SYNTHESIS OF THE HERQULINES

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The candidate confirms that the work submitted is her own and that appropriate credit has been given where reference has been made to the work of others.

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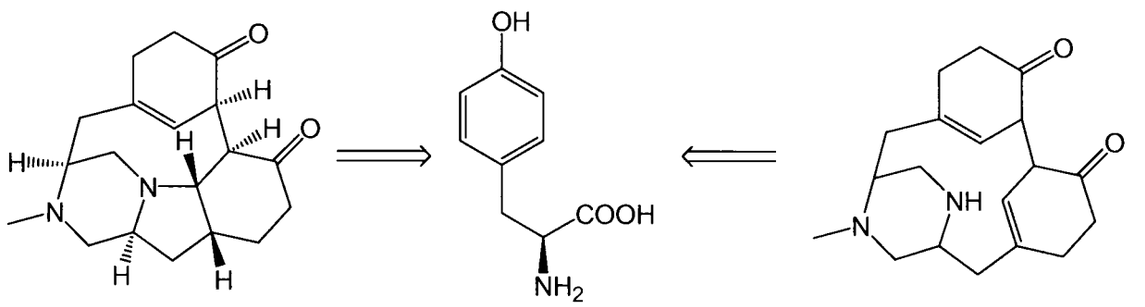
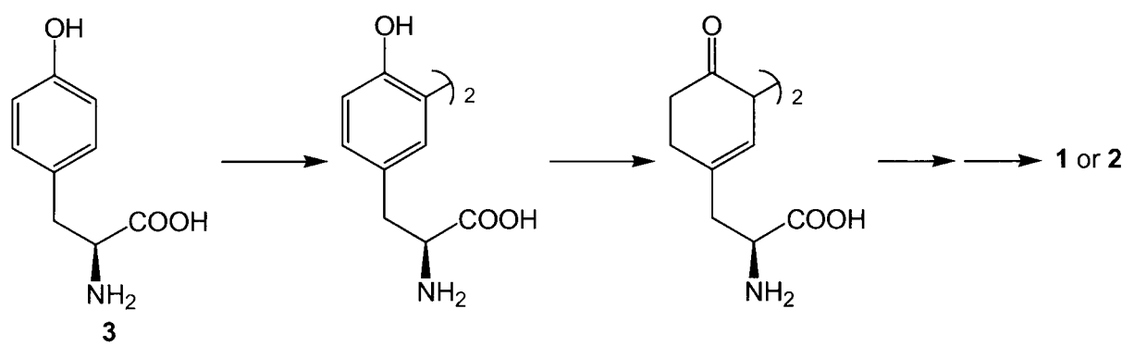
ABSTRACT

Herqulines A and B (**1** and **2** respectively) were isolated from the culture broth of a soil-derived *Penicillium* species in 1979 and 1996 respectively. They were both found to inhibit platelet aggregation, with herquline B (whose stereochemistry has not been elucidated) being the stronger inhibitor. The herqulines are thought to be biosynthesised from tyrosine **3**, but very little work relating to their total synthesis has been reported.



1 **3** **2**

This thesis describes work towards the total synthesis of herqulines A and B, focussing on two key reactions: the linking of two tyrosine molecules *via* the aromatic rings, and the reduction of the aromatic rings to give the required pattern of unsaturation.



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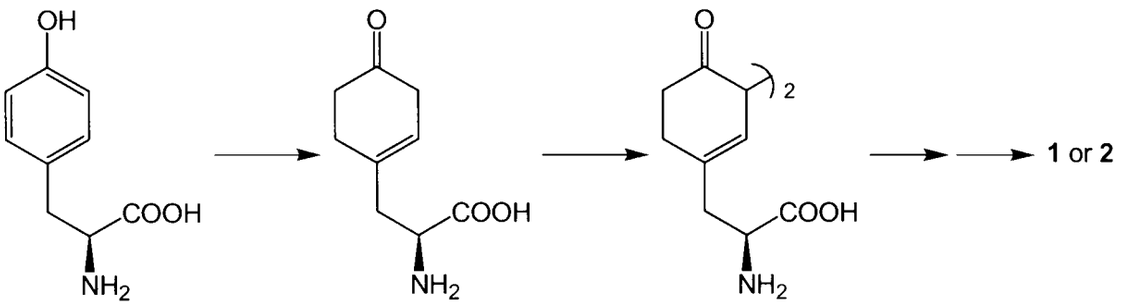
However, aryl-aryl bond formation followed by the symmetrical reduction of the biphenyl system was not successful – the required regiochemistry of the reduction posed a particular problem.

Although the target molecules **1** and **2** were not generated, strategies for their total synthesis were investigated. The most recently examined approach, which avoided the regiochemical problem mentioned above and involved the reduction of the aromatic ring of tyrosine followed by the linking of two reduced tyrosine moieties, appears promising.



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CONTENTS

ABBREVIATIONS

Ac	Acetyl
ACCN	1,1'-Azobis(cyclohexanecarbonitrile)
ADME	Absorption, distribution, metabolism, excretion
ADP	Adenosine diphosphate
AIBN	1,1'-Azobis(isobutyronitrile)
Ar	Aryl
aq.	Aqueous
ATP	Adenosine triphosphate
BINOL	1,1'-Bi-2-naphthol
Bn	Benzyl
Boc	*t*-Butoxycarbonyl
BSE	Bovine spongiform encephalopathy
Bu	Butyl
Cbz	Carbobenzyloxy
D	Aspartic acid
DBP	Dibenzoyl peroxide
DCC	Dicyclohexylcarbodiimide
DCM	Dichloromethane
DCNB	1,3-Dicyanobenzene
DDQ	Dichlorodicyanoquinone
dec.	Decomposed
DMA	*N,N*-Dimethylacetamide
DMAP	*N,N*-Dimethyl-4-aminopyridine
DME	Dimethoxyethane
DMF	*N,N*-Dimethyl formamide
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid
E	Energy
eq.	Equivalent(s)
Et	Ethyl

Ether	Diethyl ether
FR	Free radical
h	Hours
H	Histidine
HMDS	Hexamethyldisilazide
HMPA	Hexamethylphosphoramide
HOBt	1-Hydroxybenzotriazole
HRMS	High resolution mass spectrometry
Hplc	High performance liquid chromatography
i	iso
IC_{50}	The concentration at which 50 % of the test units are inhibited
IR	Infra red
LDA	Lithium diisopropylamide
m	*meta*
MAD	Bis-(2,6-di-*t*-butyl-4-methylphenoxy)methylaluminium
Me	Methyl
MRSA	Methicillin-resistant *Staphylococcus areus*
n	Normal
NBS	*N*-Bromosuccinimide
NMR	Nuclear magnetic resonance
NOC	Non-oxidative coupling
nOe	Nuclear Overhauser effect
NR	Non-radical
o	*ortho*
p	*para*
PAF	Platelet activating factor
Ph	Phenyl
Phe	Phenylalanine
PIDA	Phenyl iodine(III) diacetate
PIFA	Phenyl iodine(III) bis(trifluoroacetate)
PMB	*p*-Methoxybenzyl
ppm	Parts per million

Pr	Propyl
PSDIB	Polymer-supported diacetoxyiodo(III) benzene
Q	Glutamine
RNA	Ribonucleic acid
RT	Room temperature
s	Secondary
t	Tertiary
TBAI	Tetrabutylammonium iodide
TBDMS	*t*-Butyldimethylsilyl
TEMPO	2,2,6,6-Tetramethylpiperidinooxy
TFA	Trifluoroacetic acid
TFAA	Trifluoroacetic acid anhydride
THF	Tetrahydrofuran
TIPS	Triisopropylsilyl
tlc	Thin layer chromatography
TMEDA	*N,N,N',N'*-Tetramethylethylenediamine
TMS	Trimethylsilyl *or* tetramethylsilane
Tol	Tolyl
tRNA	Transfer ribonucleic acid
Ts	Toluenesulfonyl
Tyr	Tyrosine
UV	Ultra violet
Y	Tyrosine

CHAPTER 1
INTRODUCTION

1.1 The Herqulines

1.1.1 Isolation

Herquline A **1** is a naturally-occurring pentacyclic alkaloid containing a piperazine ring and two ketone moieties. It also contains a central nine-membered ring. Herquline A (Figure 1) was first discovered in 1979 by a group searching for novel alkaloids produced by micro-organisms.[1] It was isolated by solvent extraction and recrystallisation from a culture broth of the fungal strain *Penicillium herquei* Fg-372, which had been obtained from a soil sample collected at Saitama Prefecture, Japan. Herquline A has also been isolated from a *Penicillium* species derived from the marine bivalve mollusc *Mytilus coruscus.*[2]

The molecular formula and physical properties of herquline A were reported initially; the structure, determined by X-ray crystallography, was reported separately.[3]

Figure 1 Herquline A

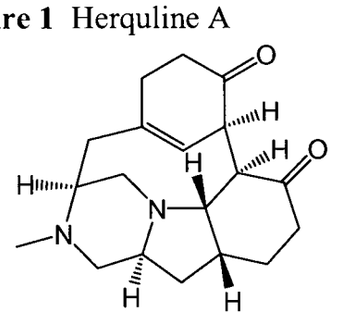

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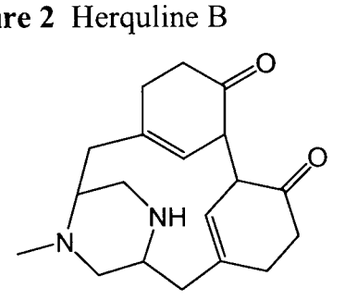
Figure 2 Herquline B



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Herquline B **2** (Figure 2) has also been isolated from *P. herquei* Fg-372.[4] It contains the same key structural elements as herquline A but is tetracyclic, containing a central twelve-membered ring. The stereochemistry of herquline B has not been elucidated; the two herqulines share a high degree of structural similarity and it seems likely that some, if not all, of the chiral centres would have the same configuration as herquline A.

1.1.2 Biological Activity

The herqulines do not exhibit any antibacterial properties but have been shown to inhibit platelet aggregation.[1, 4] During tests with platelet rich plasma from rabbit blood in which platelet aggregation had been induced using either platelet activating factor (PAF) or adenosine diphosphate (ADP), both herqulines A and B inhibited platelet aggregation. Herquline A showed only weak inhibition, but herquline B showed an IC_{50} value comparable to currently available 'anti-platelet' drugs used to prevent the formation of blood clots within the cardiovascular system (see Section 1.2.4).[4]

Table 1 Anti-platelet activities of the herqulines

Herquline	IC_{50} (μM)	
	PAF-induced aggregation	ADP-induced aggregation
A	240	180
B	5.0	1.6

1.2 Platelet Aggregation

1.2.1 Introduction

Platelet aggregation is the initial step involved in the formation of a blood clot. A soft platelet plug forms at the site of an injury and, following a complex cascade sequence, fibrinogen is converted to fibrin, which forms a mesh of fibres. This mesh traps platelets and other blood cells, resulting in a hard clot.[5] However, platelets can also adhere to damaged endothelial cells inside arteries, leading to spontaneous formation of a clot attached to the vascular wall (a thrombus). There are then two possible outcomes:

1) Separation of the thrombus from the endothelium can lead to heart attack, stroke or peripheral vascular disease if it then becomes lodged in the blood vessels of the heart, brain or other area of the body respectively. It has been shown that following a heart attack, patients remain hypersensitive to platelet activating agents such as ADP for a period of two months to two years, thereby increasing the likelihood of a further thrombotic event.[6]

2) The thrombus may become incorporated into the vascular wall. The resulting rough surface can contribute to the growth of an atherosclerotic plaque,* leading to narrowing of the arteries.[7] The flow of blood is reduced; consequently the muscles of the heart do not receive enough oxygen. This is known as myocardial ischaemia, and causes the chest pains characteristic of angina.

1.2.2 The Role of Platelet Activating Factor in Platelet Aggregation

PAF is an analogue of phosphatidyl choline† and is found throughout the body. It acts as a general messenger within biological pathways and is therefore generated by, and known to cause responses in, a wide range of cells.[8] PAF activates cells by increasing their functional and metabolic activities; consequently it is involved in many different biological pathways including inflammation responses, asthma, arthritis and platelet aggregation.[9] PAF initiates reversible platelet aggregation which may then be propagated by other substances (see Section 1.2.3).

1.2.3 The Role of Adenosine Diphosphate in Platelet Aggregation

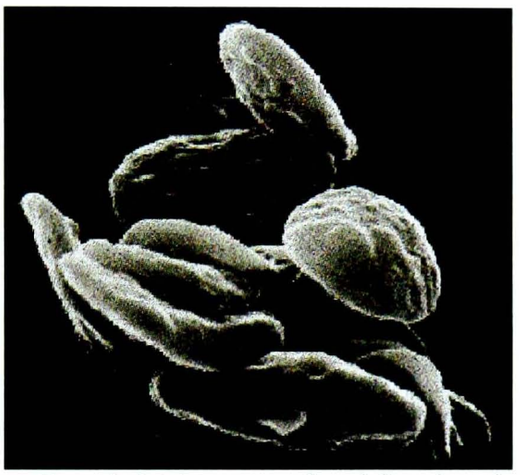
Platelets have been shown to possess three types of ADP receptor on the surface membrane – $P2Y_1$, $P2Y_{12}$ and $P2X_1$.[10] The $P2Y_1$ and $P2Y_{12}$ receptors are G-protein coupled receptors which, when activated, initiate complex signal sequences within the platelet. The $P2Y_1$ receptor is coupled to the Gq pathway which, when activated by ADP, results in the release of Ca^{2+} from internal stores leading to an increase in the cytosolic concentration of Ca^{2+}. $P2Y_1$ also causes morphological changes of the platelet resulting in reversible platelet adhesion (Figure 3).[11, 12] The $P2Y_{12}$ receptor is coupled to the Gi pathway which, when activated by ADP, results in amplification and completion of platelet aggregation. It has been shown that activation of both $P2Y_1$ and $P2Y_{12}$ is necessary for normal ADP-induced platelet aggregation, as inhibition of either receptor is sufficient to prevent aggregation.

The $P2X_1$ receptor is a gated ion channel that, when activated by ATP or ADP, allows the influx of Ca^{2+} into the platelets thus reducing the calcium ion concentration of the plasma and further increasing the cytosolic Ca^{2+} concentration. This increase in

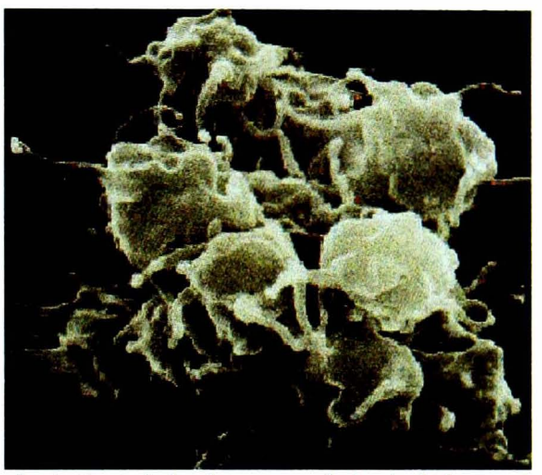
* A build-up of fatty substances and cellular waste products.
† The most abundant phospholipid found in plants and animals, and a key component of lipid bilayers.

cytosolic Ca^{2+} concentration results in the secretion of the contents of dense storage granules, including ADP, ATP and fibrinogen.[13] Thus ADP has an autocatalytic role in platelet aggregation, and can act as a potentiator for many other weak platelet activators. Platelet-platelet interactions are mediated by the binding of fibrinogen to a receptor on the platelet membrane, resulting in stronger platelet aggregation. Ca^{2+} is essential for the binding of fibrinogen to this receptor.[14]

Figure 3 The structure of platelets[a]



a. Normal platelets – smooth and disc-shaped.



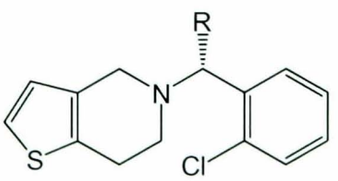
b. Activated platelets – morphological changes have made the platelets 'sticky', initiating clotting.

[a] Reproduced from *The Platelet Homepage*.[15]

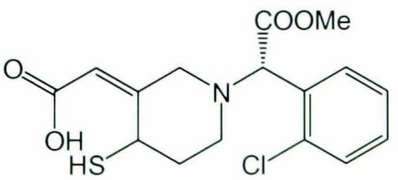
1.2.4 Inhibition of ADP-Induced Platelet Aggregation

Inhibitors of ADP-induced platelet aggregation are effective antithrombotic drugs. The thienopyridine drugs ticlopidine **4** and clopidogrel **5** (Figure 4) are irreversible inhibitors of the $P2Y_{12}$ receptor.[10] They are in fact prodrugs – inactive *in vitro* and metabolised in the liver to give their active derivatives. The active metabolite **6** of clopidogrel displayed an IC_{50} value of 1.8 μM when tested against ADP induced platelet aggregation.[16]

Figure 4 Current antithrombotic drugs



Ticlopidine 4 R = H
Clopidogrel 5 R = COOMe



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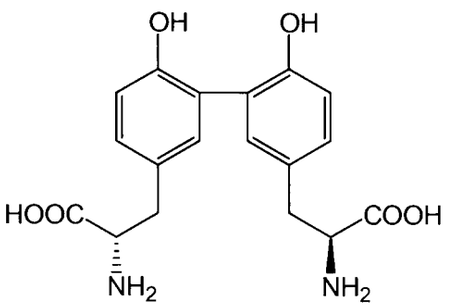
It is not known whether the herqulines inhibit platelet aggregation *via* the same mechanism as ticlopidine and clopidogrel, or whether a separate pathway is involved.

1.3 The Dityrosine Linkage

1.3.1 Introduction

Dityrosine **7** (Figure 5) was first observed in 1959 by Gross and Sizer[17] as a product of the oxidation of L-tyrosine by horseradish peroxidase and hydrogen peroxide *in vitro*.

Figure 5 Dityrosine



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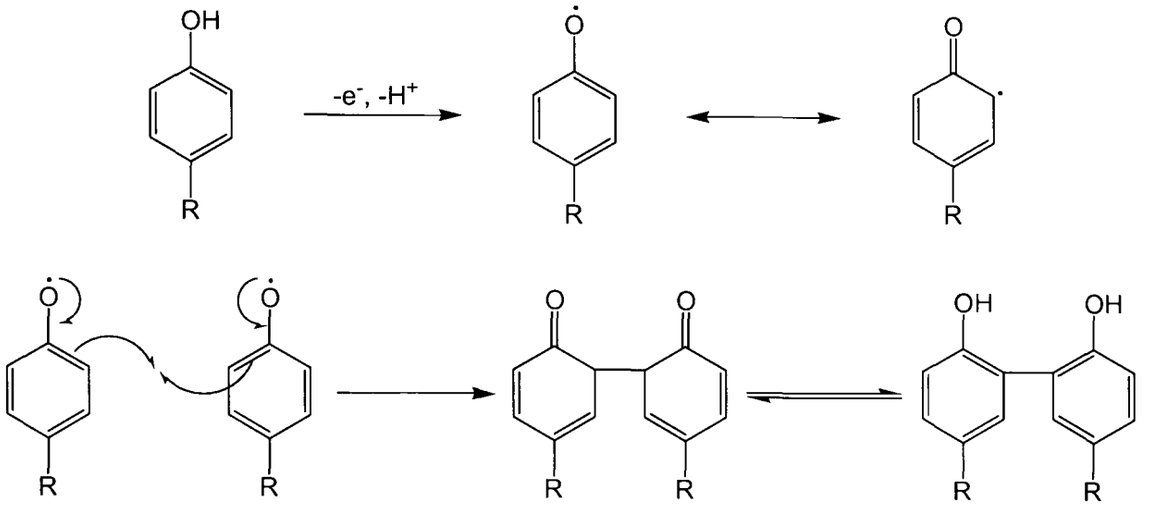
Dityrosine linkages have since been found to occur naturally in structural proteins such as keratin, elastin, collagen, silk proteins, dental enamel matrix, skin proteins of arthropods, proteins of cataractous lenses and cell walls of the yeasts *Saccharomyces cerevisae* and *Candida albicans*.[18, 19] The dityrosine cross-links are thought to confer increased structural stability due to restricted conformational flexibility; the strained conformation may also result in reduced lability towards proteases.[20]

1.3.2 Dityrosine and Disease

It has been suggested that these naturally occurring dityrosine bonds are synthesised *via* photochemical reactions or reactions involving oxygen radical species (Scheme 1). Consequently the presence of dityrosine within proteins is widely considered an indicator of oxidative stress.[21]

Scheme 1 Oxidative phenol coupling



The formation of dityrosine linkages has been associated with the development of Parkinson's disease. An amyloid disease, Parkinson's is caused by the misfolding of globular proteins, resulting in the formation of insoluble protein fibrils which then accumulate in the affected area.* Aggregation of the protein α-synuclein and its deposition as fibrils is involved in the pathogenesis of Parkinson's disease. It has been shown that formation of a dityrosine-linked 'pre-nucleus' is the critical rate-limiting step in the nucleation of α-synuclein fibrils.[22]

A study of the effect of dityrosine formation on the conformations, structural stabilities and biological activities of several proteins has been carried out by Balasubramanian and Kanwar.[23] Ribonuclease A,† α-crystallin, γB-crystallin‡ and calmodulin§ were studied. Tyrosine residues on the surface of the protein were found to form dityrosine bonds (both inter- and intramolecular linkages) most readily. Protein conformation was not found to be significantly altered by dityrosine formation, but the tyrosine-linked dimeric proteins were more easily denatured. The biological activities of the dityrosine-containing proteins were reduced, though not abolished. Intramolecular dityrosine formation appeared to give rise to more severe structural and functional consequences; this was presumably due to the more restricted conformation adopted as a result of the dityrosine bond.

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* Other amyloid diseases include Alzheimer's disease and BSE.

† Ribonuclease A is a digestive enzyme catalysing the hydrolysis of RNA.

‡ The crystallins are structural proteins found in the eye lens. Aggregation of these proteins is thought to contibute to the formation of cataracts.

§ Calmodulin is a key enzyme in signal transduction pathways.

1.3.3 Dityrosine in Natural Products

The dityrosine subunit is an important structural element in a variety of biologically significant molecules including the natural products RP 66453 (Figure 6) which binds selectively to neurotensin receptors[24, 25] and bastadin 3* (Figure 7) which shows moderate antibacterial activity.[26]

Figure 6 RP 66453

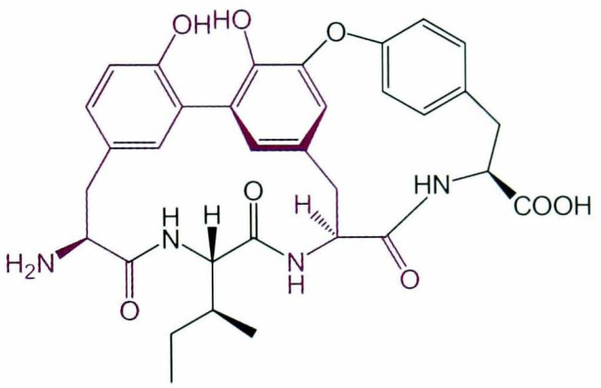

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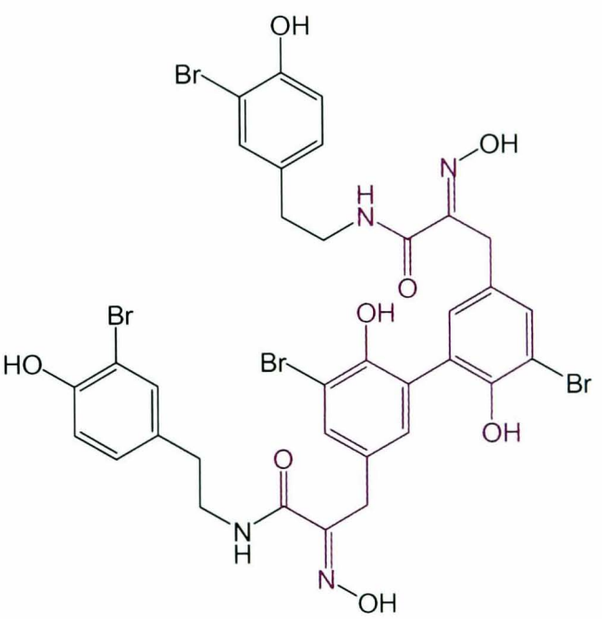
Figure 7 Bastadin 3



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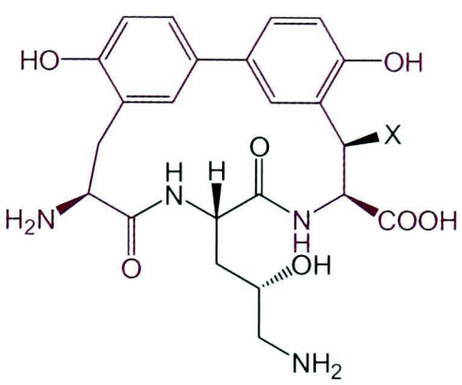
* Bastadin 3 is one of a large family of tyrosine-derived natural products isolated from the marine sponge *Ianthella basta*. Other bastadins have shown anti-cancer and anti-inflammatory activities.

The antibiotics biphenomycin A and B (**10a** and **10b** respectively) and vancomycin **11a** (Table 2) also contain similar structural motifs, with the biphenomycins containing a *para-para*-linked biphenol unit and vancomycin containing an *ortho-ortho*-linked di(phenylglycine) unit.

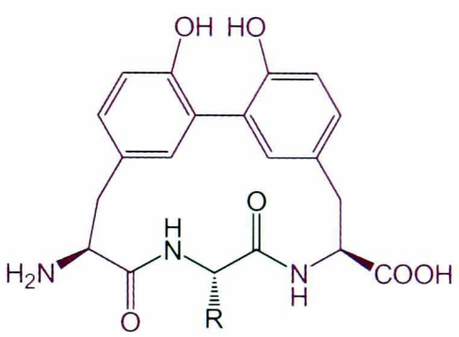
The biphenomycins **10a** and **10b** (Figure 8) are naturally occurring antibacterial agents first reported in 1985.[27] The biphenomycin analogues **12a** and **12b** (Figure 9) have been synthesised but have shown little antibacterial activity, indicating that the positions of the hydroxyl groups may be important.[28]

Figure 8



10a X = OH
10b X = H

Figure 9

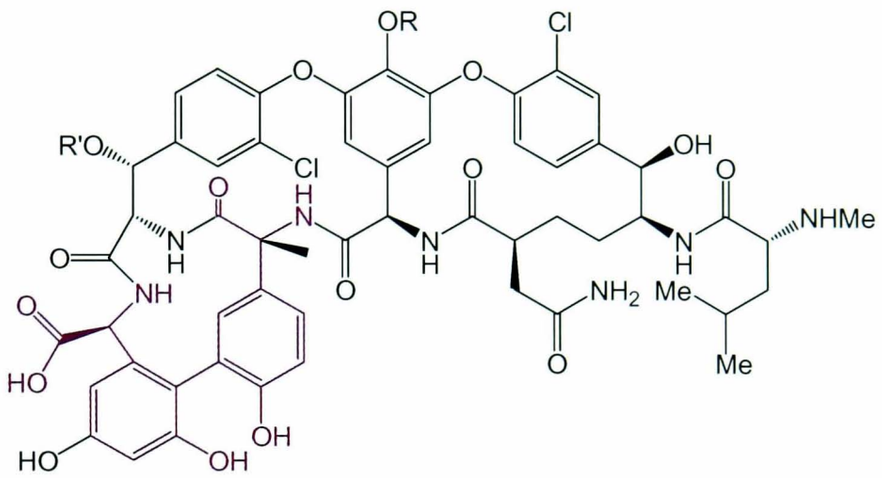


12a R = Me
12b R = $CH_2CH_2CH_2NH_2$

Vancomycin **11a** is a glycopeptide antibiotic and binds very strongly to the dipeptide D-alanine-D-alanine found within the structure of bacterial cell walls. In this way, vancomycin prevents cross-linking of the peptidoglycan chains of the cell wall. Consequently the structural integrity of the cell wall is compromised such that the cell bursts due to its high internal osmotic pressure.[29] Due to its high potency and possible serious side effects, vancomycin has become the antibiotic of last resort against Gram-positive bacterial infections including methicillin-resistant *Staphylococcus aureus* (MRSA). However, resistance to vancomycin, particularly among *Enterococcus* species, has become a serious problem in recent years.[30] In the continuing search for new antibacterial agents, many derivatives of vancomycin have been developed (Table 2). The antibiotic balhimycin **11b**, isolated in 1994, is a naturally occurring glycopeptide showing similar biological activity to vancomycin.[31] It does however show greater activity against anaerobic organisms such as *Clostridium* species.[32] The vancomycin derivatives **11c** and **11d** were synthesised from vancomycin in order to produce a superior antibacterial agent with favourable ADME properties. The disulfide

11c showed greater activity than vancomycin against MRSA; disulfide **11d** showed good activity against vancomycin-resistant *Enterococcus*.[33]

Table 2 Vancomycin **11a** and selected derivatives



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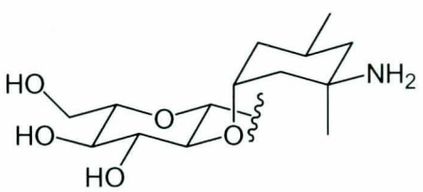
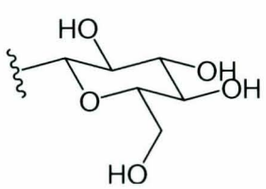
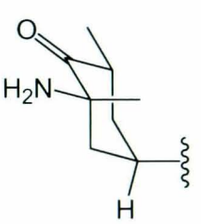
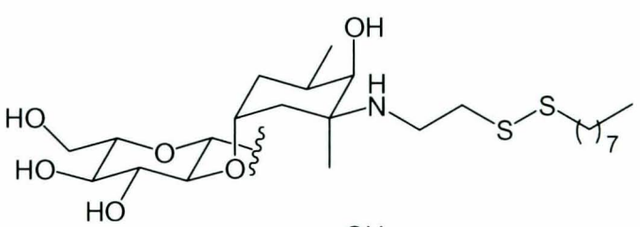
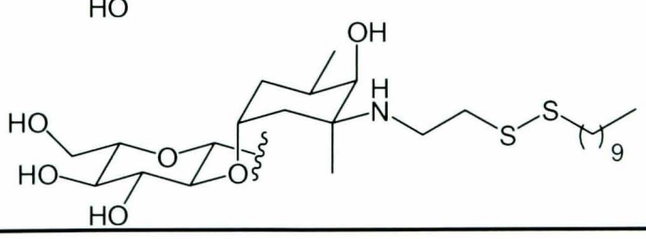
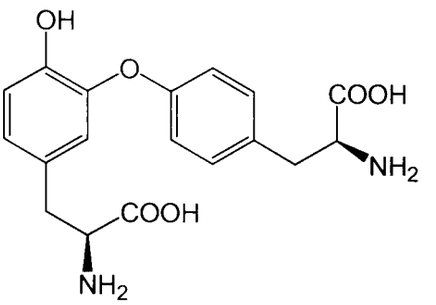
	R	R'
a		H
b		
c		H
d		H

Vancomycin **11a** and bastadin 3 **9** also contain an isodityrosine unit. Isodityrosine **13** (Figure 10) is another structural element found frequently in natural products, and is also a common by-product arising from many syntheses of dityrosine.[20] Its generation

can be explained by consideration of the oxidative coupling pathway shown in Scheme 1 – the coupling of two tyrosine radicals where one residue reacts at carbon and the other at oxygen would result in the formation of isodityrosine.

Figure 10 Isodityrosine



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1.4 Biosynthesis of the Herqulines

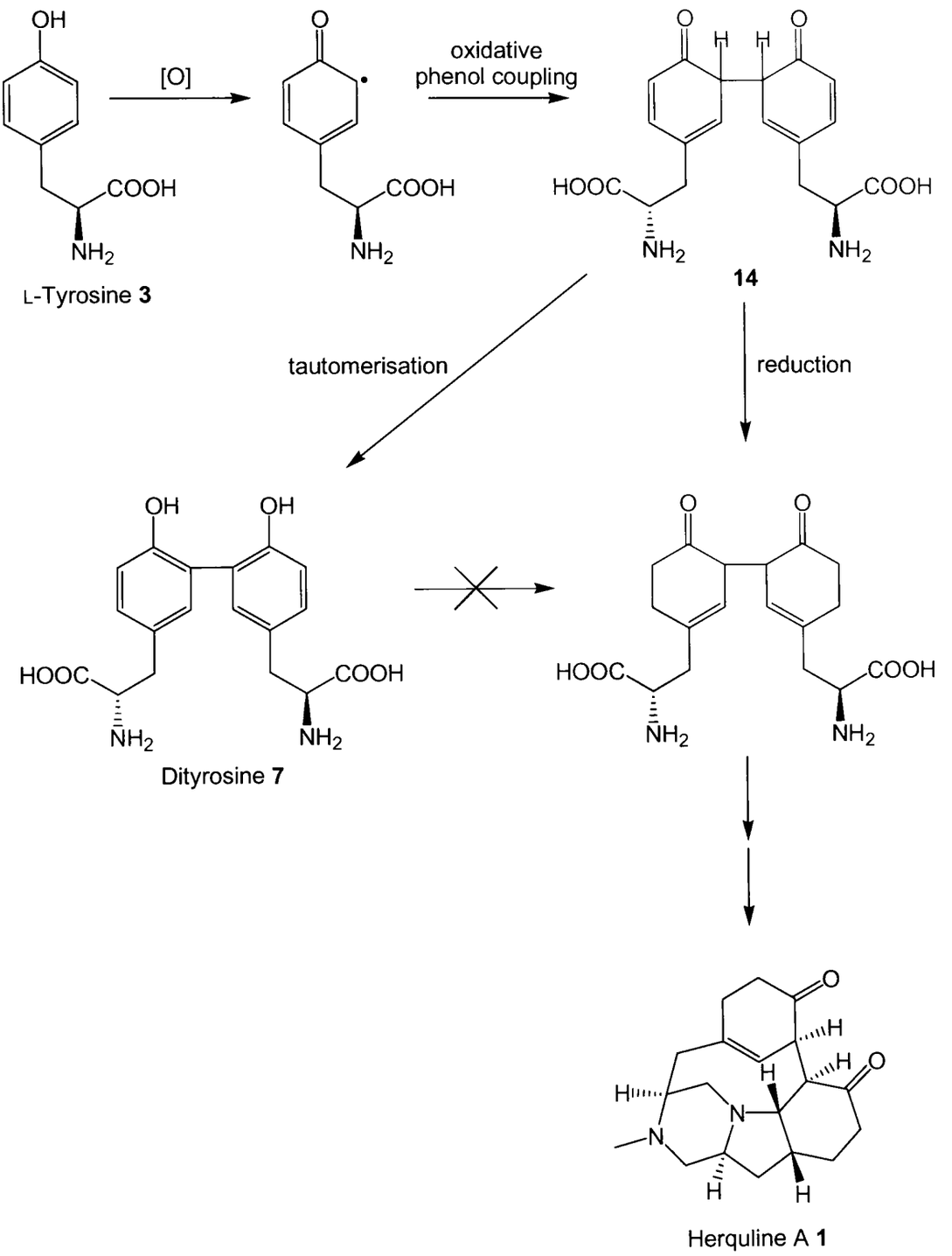
Consideration of the structure of the herqulines led to the observation that they probably arise biosynthetically from two molecules of L-tyrosine **3**. Indeed, it has been shown that the production of herquline A from a culture of *P. herquei* increases when L-tyrosine is added.[4] A possible route for the biosynthesis of the herqulines is shown in Scheme 2.

Reduction of the aromatic rings of the tyrosine molecules is clearly necessary in order to produce the herqulines. The classical method for performing this type of reduction in synthetic chemistry is the Birch reduction.* However, since there is no known equivalent of the Birch reduction in nature, it is assumed that the reduction of the aromatic rings must occur on the 'keto-tautomer' **14** of the biphenol. One example of a biochemical Birch-type reduction has been reported: it has been proposed that the partial reduction of a benzoyl moiety by benzoyl-CoA reductase proceeds *via* alternating single-electron addition and proton transfer steps.[34] However, this reduction results in the formation of a conjugated cyclohexadiene, rather than the 1,4-cyclohexadiene normally produced by a Birch reduction. The presence of an electron-withdrawing substituent on the aromatic ring (a thioester moiety) is also significant,

* Treatment of aromatic rings with a solution of sodium or lithium in liquid ammonia, usually in the presence of an alcohol, results in the formation of 1,4-cyclohexadienes.

since such groups are not present in the herqulines and greatly facilitate electron addition.

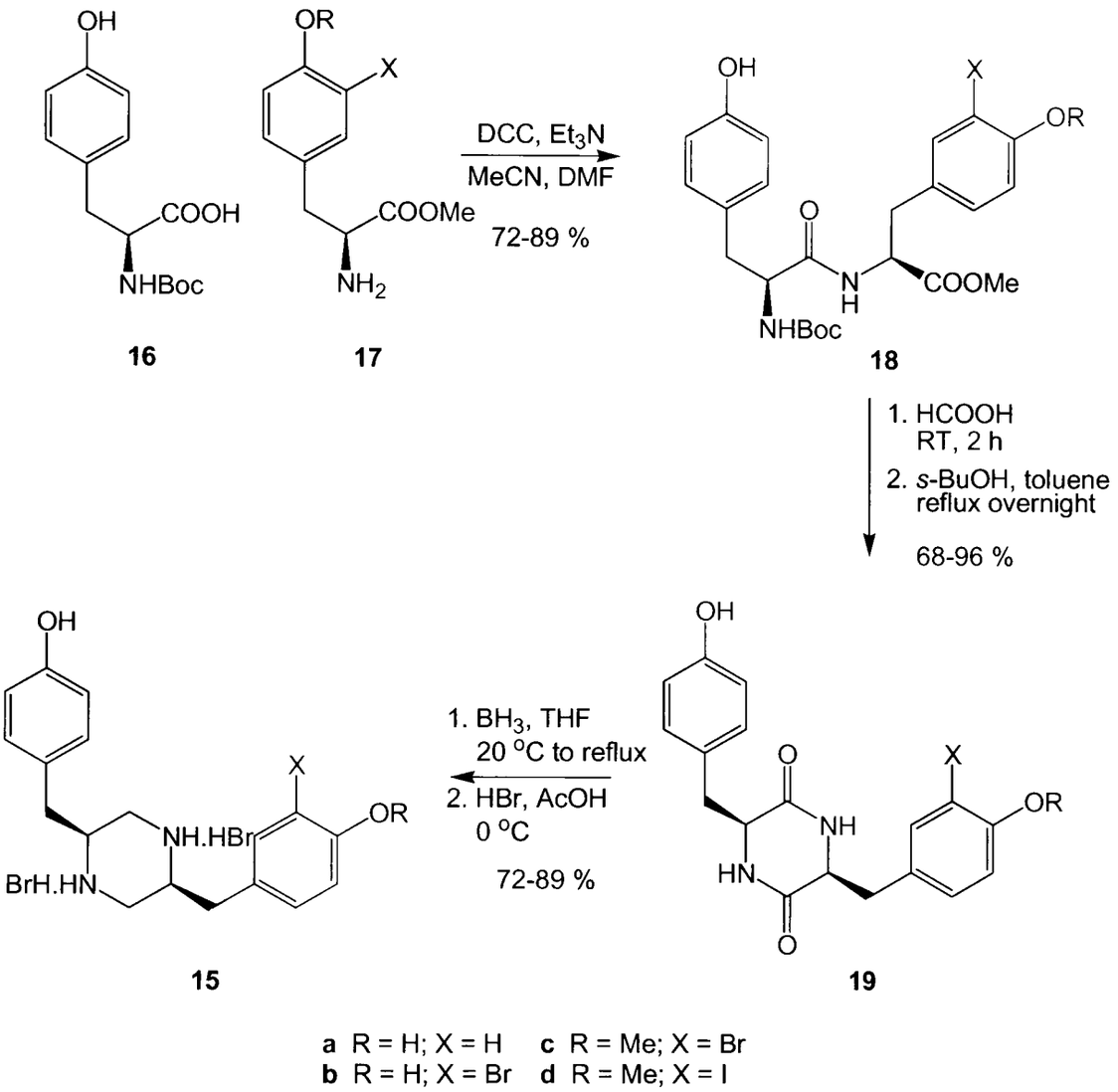
Scheme 2 Possible biosynthesis of Herquline A



1.5 Previous Synthesis Towards the Herqulines

The total synthesis of the herqulines has not been reported to date. In 1985 Jung reported the synthesis of a series of tyrosine-derived piperazines **15a-d** (Scheme 3) intended as intermediates in a synthesis of the herqulines.[35] No other work relating to the herqulines has been reported.

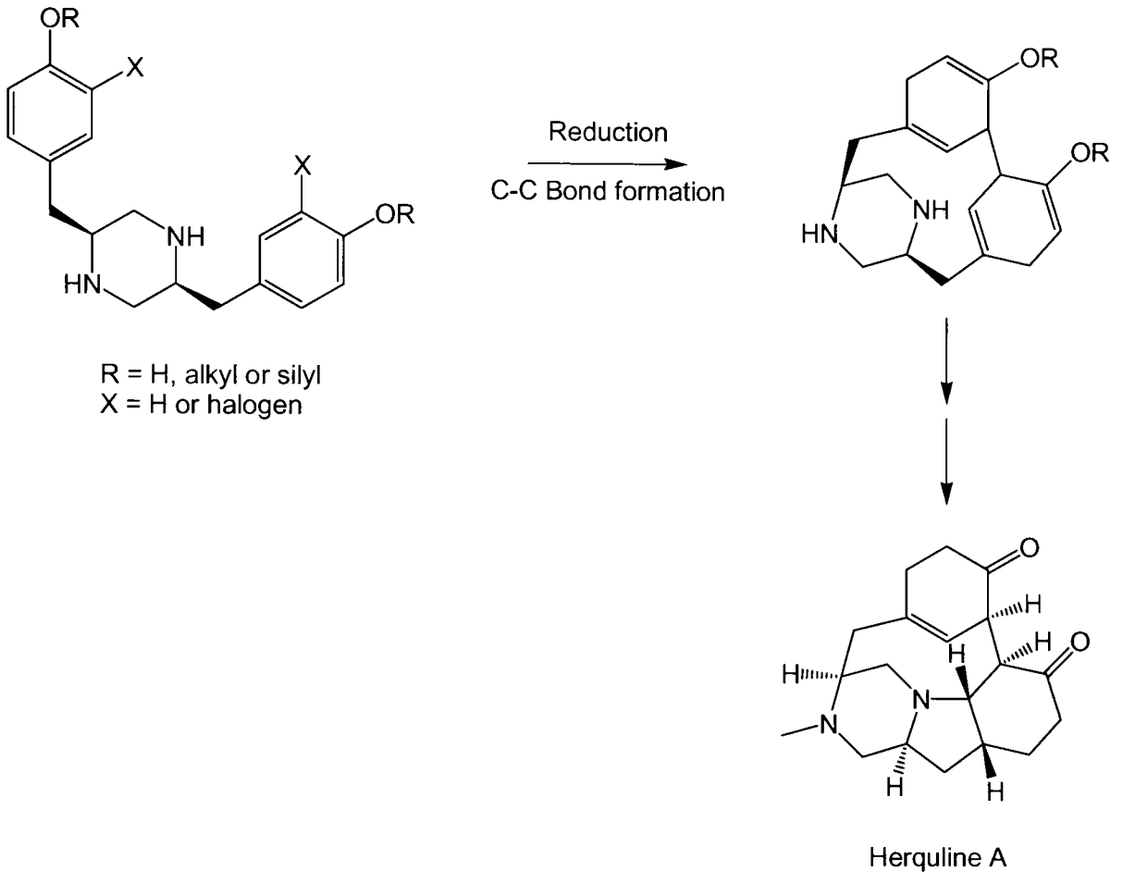
Scheme 3 Synthesis of tyrosine-derived piperazines[35]



a R = H; X = H **c** R = Me; X = Br
b R = H; X = Br **d** R = Me; X = I

Jung proposed that performing a carbon-carbon bond forming reaction on reduced derivatives of this kind of piperazine would give an intermediate in the synthesis of herquline A (Scheme 4).

Scheme 4 Potential use of piperazine intermediates in the synthesis of herquline A

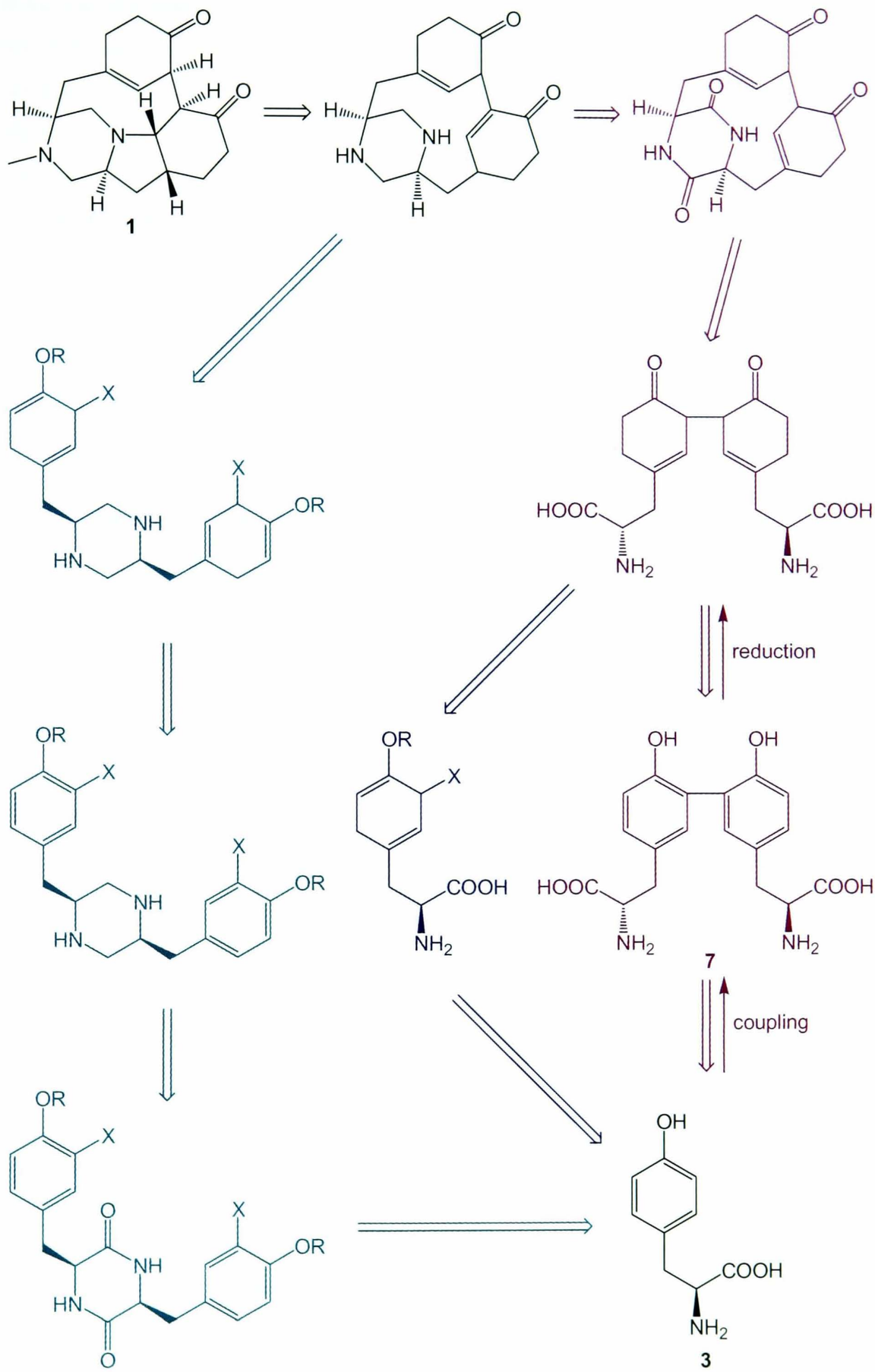


1.6 Proposed Total Synthesis of the Herqulines

1.6.1 Retrosynthetic Analysis

Working from the proposed biosynthetic precursor L-tyrosine, retrosynthetic analysis was performed on herquline A **1** to provide a starting point for the project (Scheme 5). Herquline A **1** contains six chiral centres of which, in the suggested synthesis, two would be provided by using L-tyrosine as the starting material. It was assumed that herqulines A and B are low energy stereoisomers since epimerisation of chiral centres may occur readily *in vivo* by deprotonation-protonation reactions. It was therefore hoped that the use of reaction conditions favouring equilibration would allow generation of the desired isomers.

Scheme 5 Retrosynthetic analysis



Route 1 (shown in green) illustrates the type of route proposed by Jung, involving the reduction of a piperazine intermediate. Route 2 (shown in purple) was the starting point for this thesis and involves the reduction of dityrosine. A variation on this route involving the reversal of the two key steps (coupling and reduction) is shown in blue. Route 2 provides not only a promising pathway toward the herqulines, but the opportunity to investigate an interesting reduction of a biphenyl system (variations on the Birch reduction are of particular interest to our group).

The two key steps in this retrosynthetic plan are the generation of dityrosine and the reduction of the aromatic rings with appropriate regiochemistry. These steps have been the focus of much of the work involved in this project.

1.6.2 Formation of Dityrosine

The formation of dityrosine is discussed in detail in Chapter 2. This chapter contains background information on a range of aryl-aryl coupling techniques including oxidative couplings, palladium-catalysed reactions and nickel-mediated couplings. Details of their application to the synthesis of the herqulines are also given.

1.6.3 Reduction of Dityrosine

Work towards the reduction of dityrosine is discussed in detail in Chapter 3. This chapter includes background information on the Birch reduction and its potential application to the synthesis of the herqulines. Also described is work towards on the synthesis and subsequent reduction of tyrosine-derived spirolactones.

1.6.4 Synthesis & Reactions of Birch-Reduced Tyrosine Systems

Chapter 4 details background information relating to the Birch reduction and subsequent coupling of tyrosine. The application of these techniques to a both a model system and tyrosine is reported. Investigation into stereochemical aspects of the synthesis of herquline A is also described.

CHAPTER 2
FORMATION OF DITYROSINE

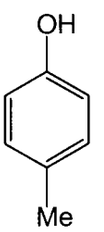
2.1 Introduction

As discussed in Section 1.6, the homocoupling of L-tyrosine **3** to form dityrosine **7** was considered a key step in the synthesis of herqulines A and B. This chapter contains background information relating to a range of different coupling techniques as well as discussion of results obtained during the course of this project.

A wide variety of techniques has been used to generate aryl-aryl bonds similar to those found in dityrosine units. There are also a number of methods that have been used specifically to generate dityrosine-base molecules. For this project, a homocoupling technique rather than one involving the preparation of a separate organometallic intermediate was sought in order to utilise the type of key symmetrical intermediates identified previously (Section 1.6.1). Another important consideration was the yield of reactions generating biaryl compounds containing electron-donating substituents *ortho* to the aryl-aryl bond; both *ortho*-substitution and the presence of electron-donating groups have been found to reduce the efficiency of reactions involving the homocoupling of aryl halides.[36]

It was decided that the initial use of model compounds would be beneficial, allowing the testing of a number of different reaction conditions without the additional complications of labile protons or protecting groups. Consequently *p*-cresol **20** (Figure 11) – a readily available aromatic molecule with the same substitution pattern as tyrosine – was chosen to test many of the reactions under consideration.

Figure 11 *p*-Cresol

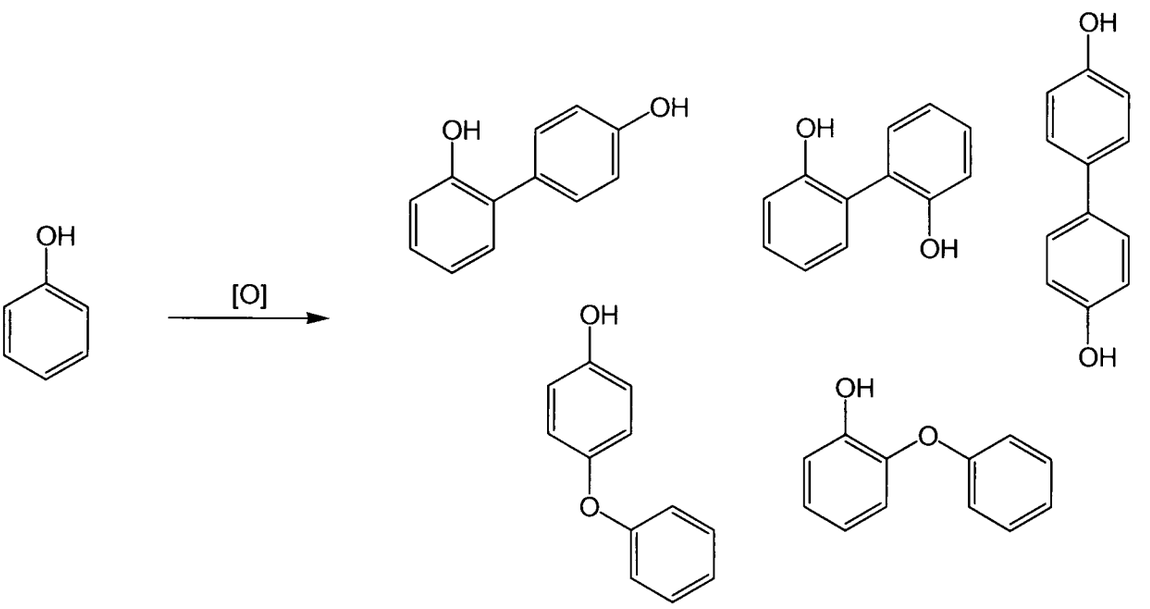


20

2.2 Oxidative Coupling Methods

In the presence of a variety of oxidising agents, phenolic molecules can combine to form a range of products arising from carbon-carbon or carbon-oxygen coupling. Carbon atoms *ortho* or *para* to the aromatic hydroxyl group are able to undergo coupling; the range of possible products is shown in Figure 12. Polymerisation products are also possible in many cases.

Figure 12 Possible products arising from the oxidation of phenol

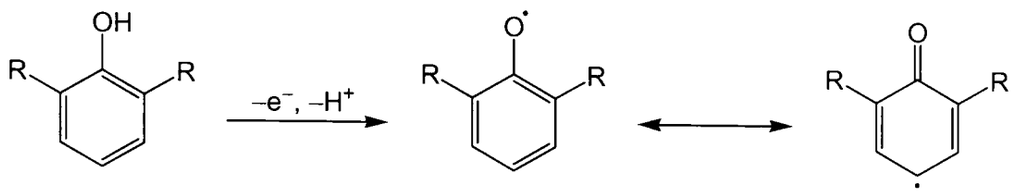


2.2.1 Mechanisms of Oxidative Phenol Coupling

The involvement of a radical dimerisation mechanism in oxidative phenol coupling is widely accepted and was briefly discussed in Section 1.3. However, there are a number of variants of this mechanism that must be fully considered.[37] The mechanisms described below are shown with reference to the *para-para* coupling of a 2,6-disubstituted phenol.

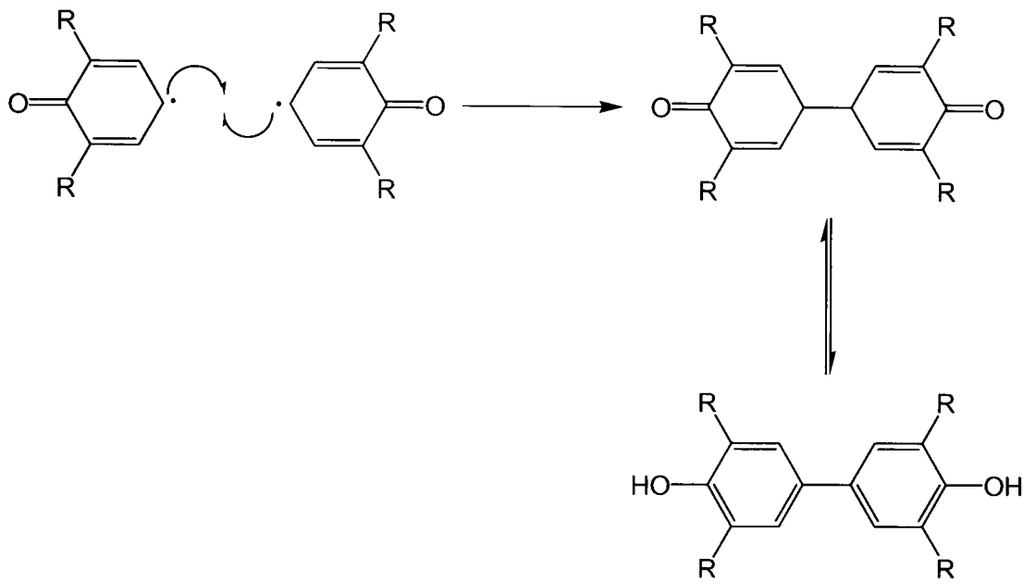
The initial step in any radical-based mechanism must be the generation of the phenoxy radical (Scheme 6). The radical can then react *via* the oxygen or at unsubstituted *ortho* or *para* carbon centres. Only reaction *via* carbon will be considered here.

Scheme 6 Generation of the phenoxy radical



The most widely accepted mechanism for oxidative phenol coupling is the direct combination of two phenoxy radicals (FR1 mechanism; Scheme 7).

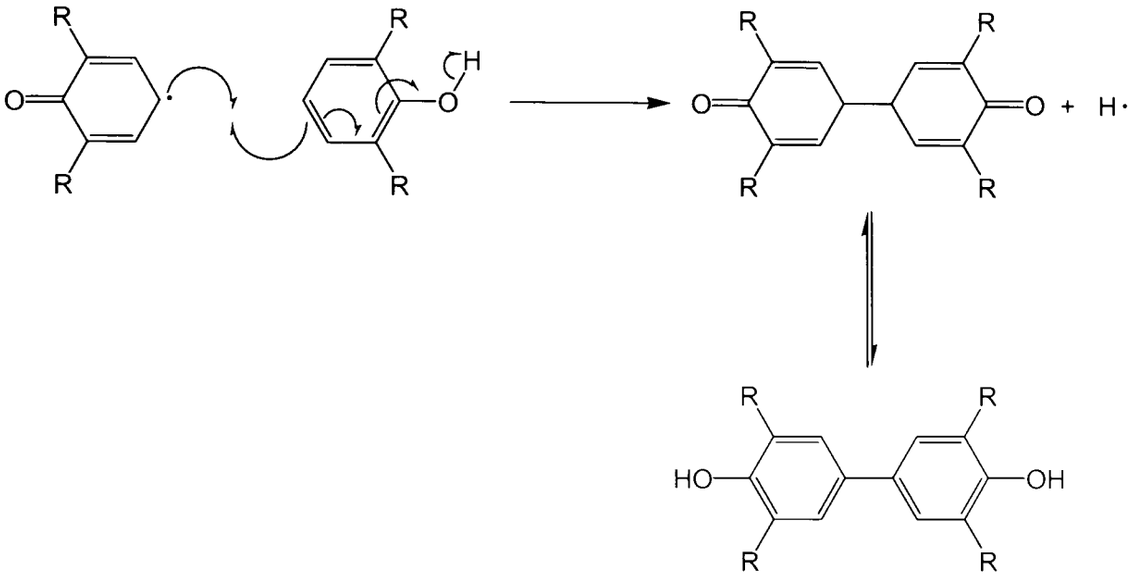
Scheme 7 FR1 Mechanism – the direct coupling of two phenoxy radicals



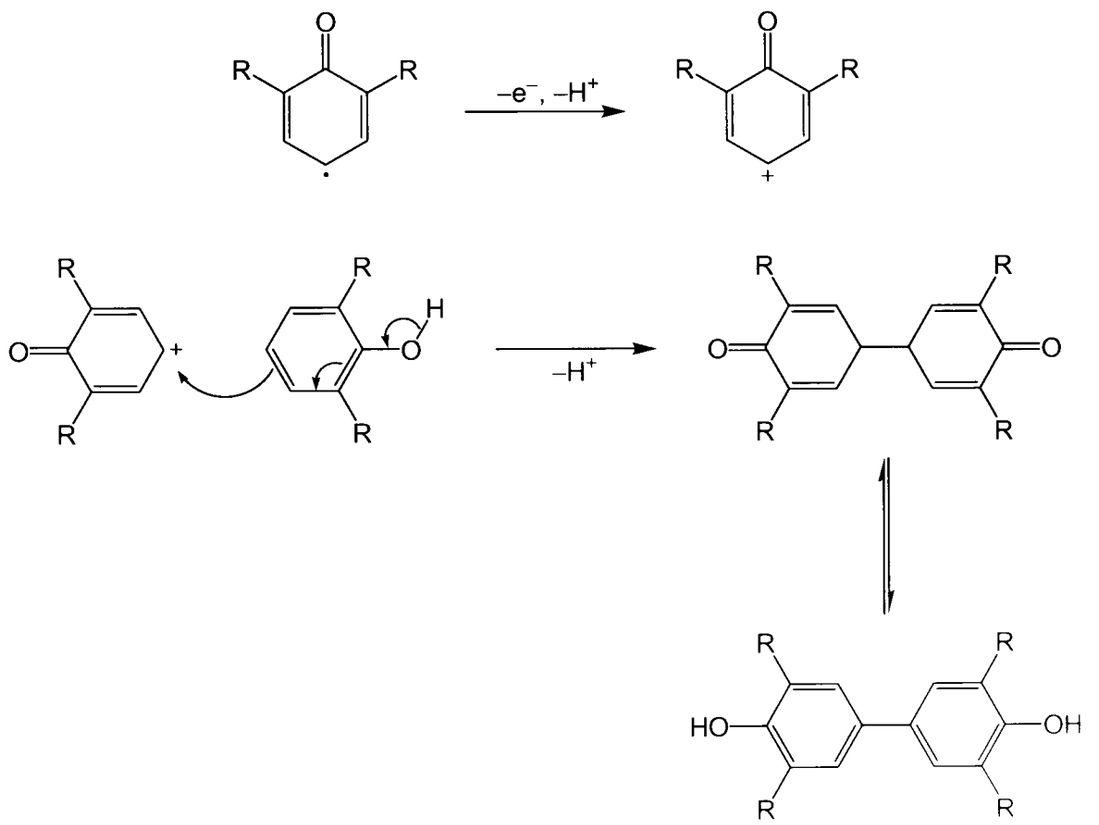
However on consideration of the general pathway of radical reactions (initiation, propagation and termination) it seems likely that a more 'traditional' propagation step may occur, in which the phenoxy radical reacts with a phenol to give the coupled product and another radical species (FR2 mechanism; Scheme 8).

Another possibility is that the phenoxy radical is further oxidised to give a phenoxy cation, which then reacts with a phenol in a non-radical manner (FR3 mechanism; Scheme 9). This is the least favourable of the three mechanisms discussed thus far, owing to the large amount of energy required to oxidise a phenoxy radical to a phenoxy cation.

Scheme 8 FR2 Mechanism – reaction of a phenoxy radical with a phenol

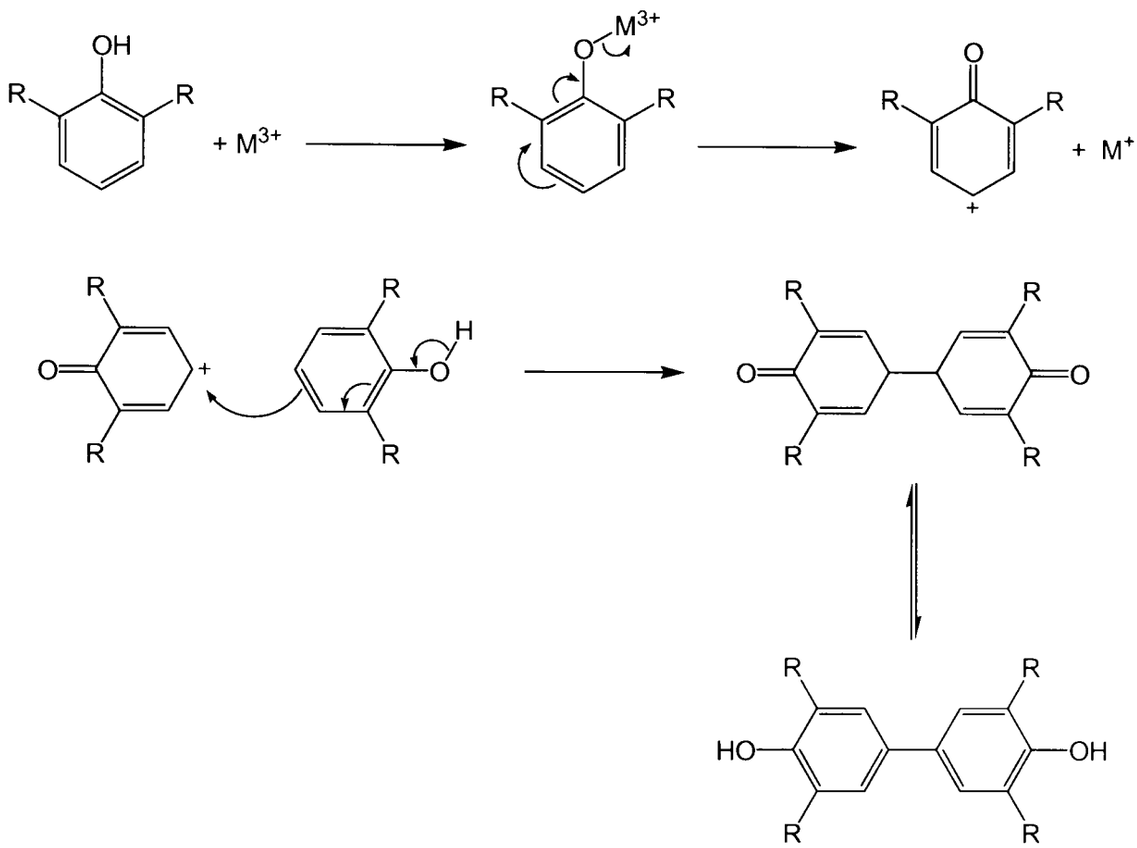


Scheme 9 FR3 Mechanism – two consecutive one-electron oxidations resulting in a phenoxy cation, which then reacts with a phenol



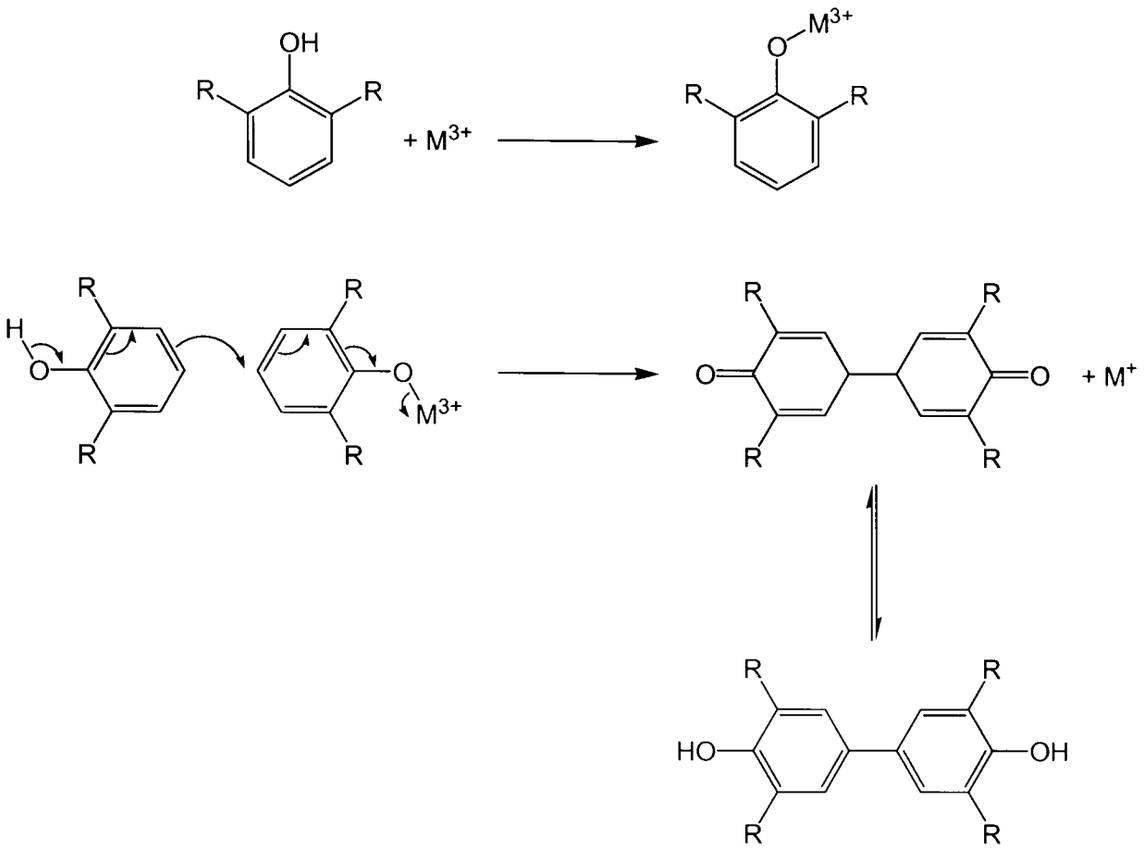
Alternatively, oxidative phenol coupling may occur *via* a mechanism in which a phenoxy radical is never generated. A phenol may undergo a two-electron oxidation producing a phenoxy cation, which then reacts with a phenol as described above (NR1 mechanism; Scheme 10).

Scheme 10 NR1 Mechanism – two-electron oxidation resulting in a phenoxy cation, which then reacts with a phenol



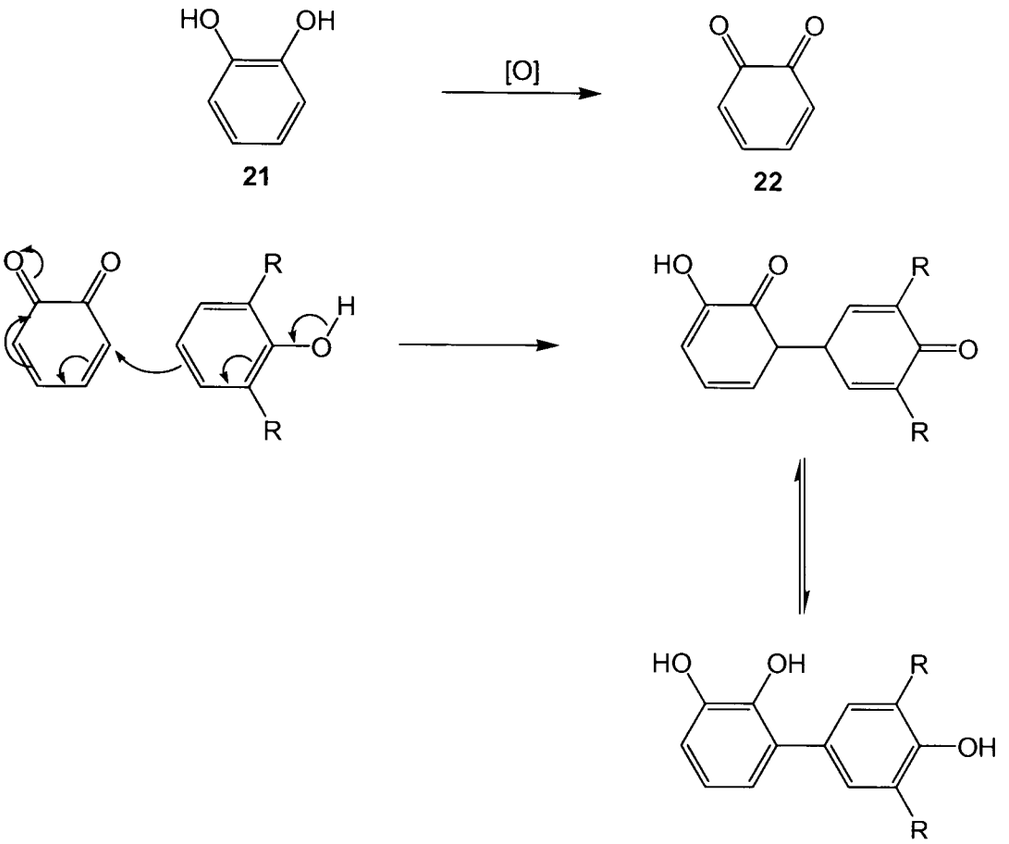
An alternative pathway avoids the formation of both the phenoxy radical and the phenoxy cation (both high-energy reactive intermediates). A phenoxy-metal complex reacts with a phenol in a concerted redox-coupling step (NR2 mechanism; Scheme 11).

Scheme 11 NR2 Mechanism – concerted coupling and electron transfer mechanism



In the interest of completeness, one final mechanism should be considered. Non-oxidative coupling may occur when one of the coupling moieties is reduced during coupling; consequently no external oxidant is required. It is feasible that the treatment of a mixture of catechol **21** and a 2,6-disubstituted phenol with an oxidising agent may result in the oxidation of catechol to give *ortho*-quinone **22**. The quinone is then able to act as an internal oxidant in the coupling reaction (Scheme 12).

Scheme 12 NOC Mechanism – one of the coupling moieties acts as an internal oxidant



2.2.2 Enzyme Catalysed Oxidation

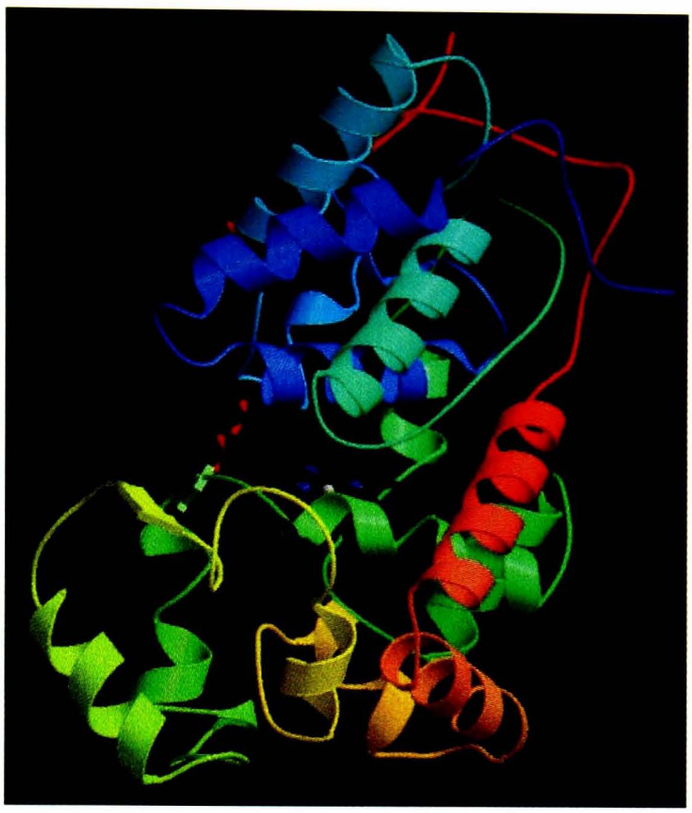
2.2.2.1 Introduction

Dityrosine itself can be generated *in vitro* by the action of horseradish peroxidase and hydrogen peroxide. Horseradish peroxidase and hydrogen peroxide have also been used to generate dityrosine cross-links *in vitro* in fibroin,* insulin, ribonuclease and chymotrypsin.†[19]

Horseradish peroxidase is one member of a large group of plant peroxidase enzymes and has been widely studied.[38-40] The horseradish peroxidase/hydrogen peroxide system is thought to produce dityrosine *via* a one-electron oxidation of tyrosine to give a phenoxy radical.[41, 42] Therefore the enzyme-catalysed oxidation probably proceeds *via* an FR1 or FR2 mechanism.

* Fibroin is a structural protein, and the major component of silk fibres.
† Insulin, ribonuclease and chymotrypsin are all globular rather than structural proteins.

Figure 13 Horseradish peroxidase[a]



[a] Reproduced using data from the Research Collaboratory for Structural Bioinformatics Protein Data Bank.[40, 43]

Amadò *et al.* reported the horseradish peroxidase-catalysed synthesis of dityrosine in 17 % yield in 1994; the analogous synthesis of di-(*N*-acetyl-L-tyrosine) proceeded in 11 % yield.[19] Anderson has since reported the synthesis of dityrosine in 27 % yield using a similar procedure.[18]

More recently, Rieker has reported a systematic study of chemical and enzymatic oxidative coupling techniques used to facilitate the formation of dityrosines and isodityrosines.[20] The group reported enzymatic syntheses of dityrosines resulting in moderate to good yields (shown in Table 3). However, the study was carried out using very small quantities of protected tyrosines (30-32 μmol) and higher yields were only obtained using tyrosine amides. The use of amides may not be appropriate for a total synthesis, as amide bonds can be particularly difficult to cleave.

Table 3 Yield of horseradish peroxidase-catalysed dityrosine formations[20]

Substrate	% Yield
N-Acetyl-L-tyrosine	53
N-Cbz-L-tyrosine	60
N-Acetyl-L-tyrosine amide	90

2.2.2.2 Application to the Synthesis of the Herqulines

Enzyme-catalysed formation of dityrosine was not investigated due to a number of factors:

1) Most routes resulted in low to moderate yields.

2) Higher yields were only achieved using amide protecting groups, which may have led to problems later in the synthesis. Amides are extremely stable and hydrolysis often requires heating in strongly acidic or basic solution.

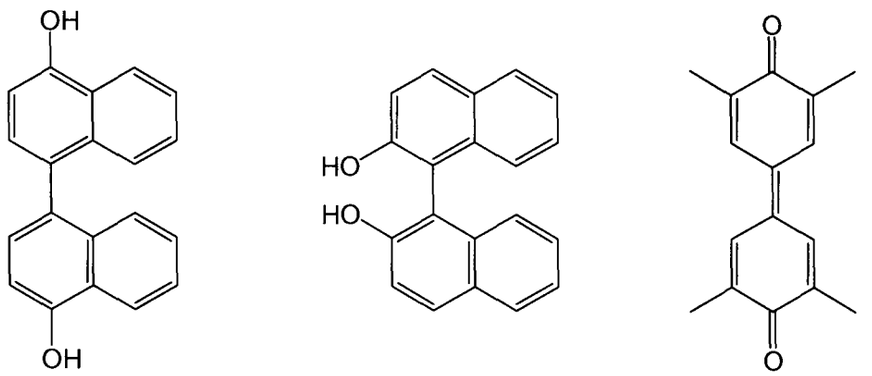
3) The enzymatic syntheses of dityrosines involved complex and lengthy preparation, extraction and purification techniques.

2.2.3 Oxidation Using Vanadium

2.2.3.1 Introduction

Oxidative coupling of phenols using vanadium tetrachloride or vanadium oxytrichloride was first reported by Carrick *et al.* in 1969.[44] Vanadium oxytrichloride affected the oxidative coupling of phenol itself in very low yield, but use of more reactive substrates led to the isolation of the dimers **23-25** in 38 to 40 % yield (Figure 14).

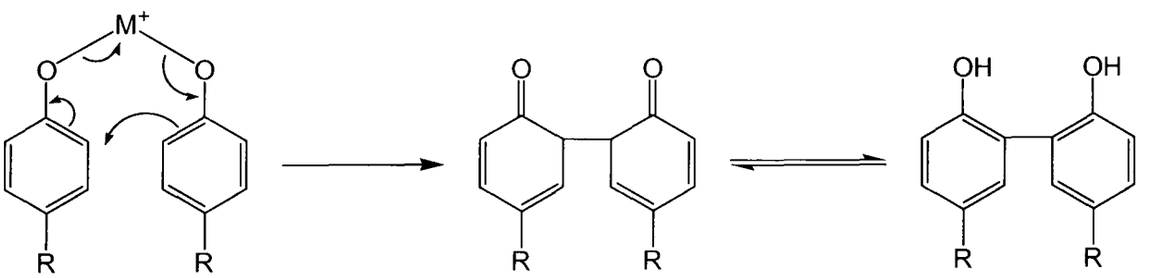
Figure 14



23 **24** **25**

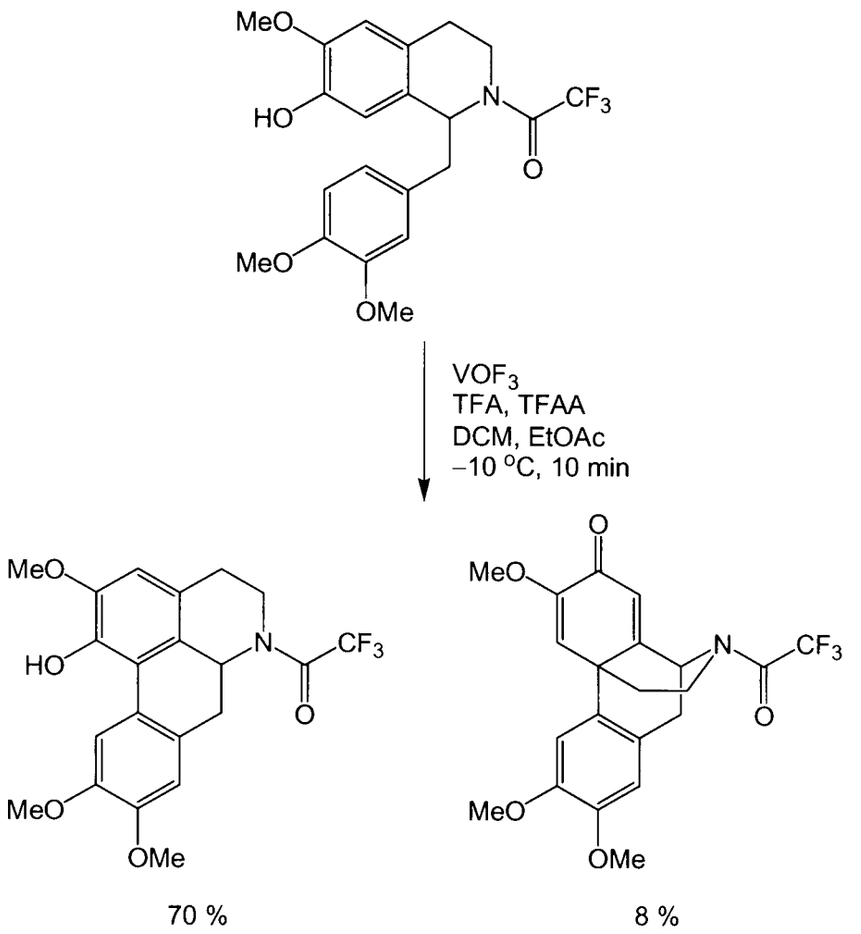
Subsequent mechanistic investigations indicated that vanadium phenoxides were formed during the reaction. This led the group to propose that coupling occurred not *via* a radical mechanism, but by a rearrangement of electrons within a complex containing at least two phenoxide residues and at least one metal centre. In their 1973 review of the subject, McDonald and Hamilton[37] also concluded that the mechanism of oxidation by vanadium (V) compounds is complex and may proceed *via* a two-electron, non-radical, mechanism. These deductions imply that in the case of *ortho-ortho* coupling a concerted cyclic mechanism such as that shown in Scheme 13 may be involved.

Scheme 13 Proposed cyclic NR2 mechanism for *ortho-ortho* coupling



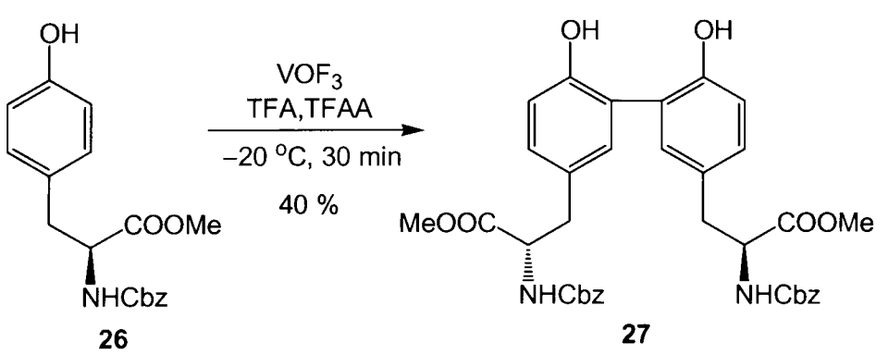
Intramolecular monophenol couplings facilitated by vanadium oxytrifluoride have been reported as part of the syntheses of a variety of alkaloids.[45] An example is shown in Scheme 14; although the reaction shown here is slightly different from the type of dimerisation under consideration, the key step (oxidation of the phenol) must proceed in essentially the same manner.

Scheme 14



Subsequently, researchers from SmithKline Beecham used vanadium oxytrifluoride to synthesise a protected dityrosine **27** in moderate yield as part of the synthesis of some analogues of biphenomycin (Scheme 15; see Section 1.3.2 for the biphenomycin analogues).[28] A yield of 40 % was achieved; when the reaction was performed using vanadium oxytrichloride only 20 % of the desired product was obtained.

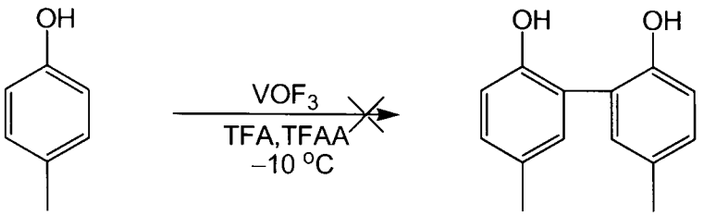
Scheme 15



2.2.3.2 Application to the Synthesis of the Herqulines

An attempt was made to couple *p*-cresol **20** using vanadium oxytrifluoride (Scheme 16). However, the only products isolated appeared to be vanadium salts or complexes.*

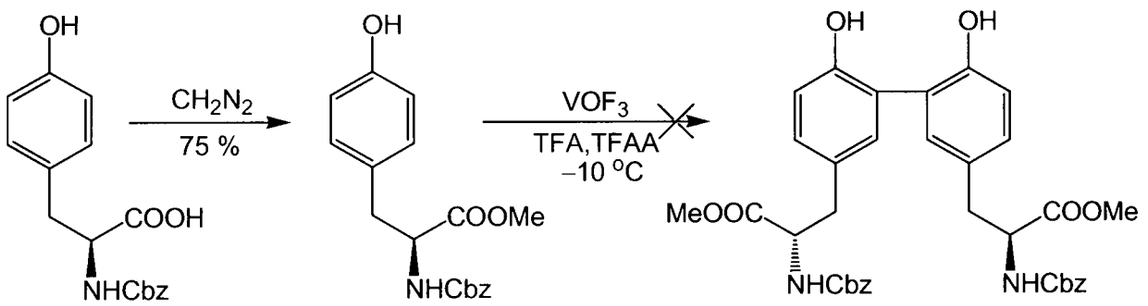
Scheme 16



20 **28**

An attempt was made to reproduce the oxidative coupling of *N*-Cbz-L-tyrosine methyl ester. Commercially available *N*-Cbz-L-tyrosine was treated with a freshly prepared solution of diazomethane to give the protected tyrosine **26**. However, treatment with vanadium oxytrifluoride and TFA failed to afford the desired dityrosine **27** (Scheme 17).

Scheme 17



29 **26** **27**

These results would appear to indicate that hydrolysis of vanadium-phenoxide complexes was not successful under the conditions employed, and a separate hydrolysis step following the oxidation may be necessary.

Following the failure of vanadium oxytrifluoride-mediated coupling, other methods of oxidative coupling were examined.

* Carrick *et al.* reported that they were unable to isolate cresol dimers following the oxidation of cresols using vanadium oxytrichloride.

2.2.4 Oxidation Using DDQ

2.2.4.1 Introduction

The use of DDQ (Figure 15) and aluminium chloride to facilitate the oxidative coupling of phenols has also been reported.[37] DDQ is a two-electron oxidant which is known to abstract hydride from hydroaromatic compounds, such as dihydropyridines, to form cationic intermediates. The oxidative coupling of phenols using DDQ may therefore proceed *via* an NR1 mechanism. Aluminium chloride is thought to play a role in preventing the formation of trimers and higher order oligomers *via* the formation of an aluminium complex (Figure 16).[46]

Figure 15 2,3-Dichloro-5,6-dicyano-1,4-benzoquinone (DDQ)

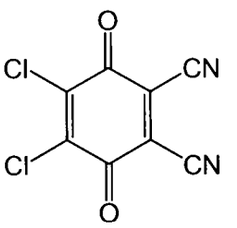
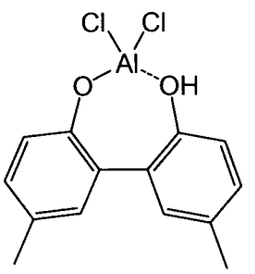
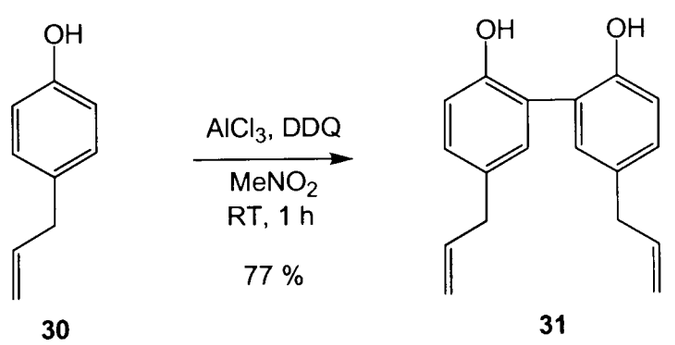


Figure 16



Gu *et al.* have used DDQ and aluminium chloride to oxidatively couple 4-allyl phenol **30**, producing the dimer **31** in 77 % yield (Scheme 18).[47]

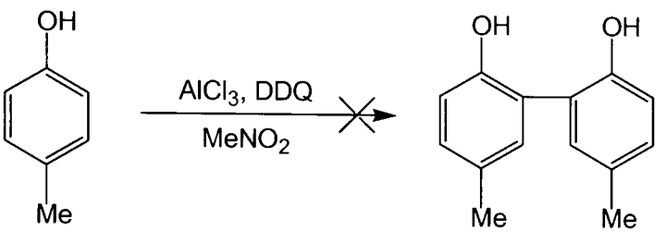
Scheme 18



2.2.4.2 Application to the Synthesis of the Herqulines

Attempts to couple *p*-cresol **20** using this method were unsuccessful – the ^{1}H-NMR of the crude product indicated that starting material was still present and no evidence of dimer formation was observed (Scheme 19).

Scheme 19



20 **28**

Following several unsuccessful attempts at DDQ-mediated coupling, other conditions facilitating oxidative coupling were sought.

2.2.5 Oxidation Using Ferric Chloride

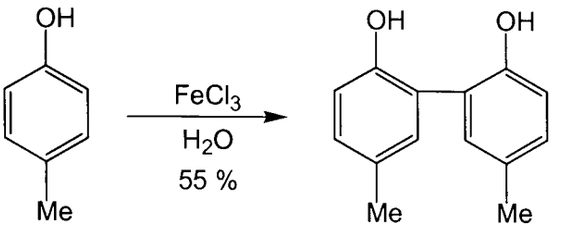
2.2.5.1 Introduction

Ferric chloride is a mild single-electron oxidant, with electron transfer thought to occur *via* an iron-oxygen bond.[48] When used in phenolic oxidative coupling reactions, ferric chloride results mostly in carbon-carbon rather than carbon-oxygen coupling; the formation of a stable iron-oxygen bond may account for this.

A study of the oxidative coupling of *p*-cresol **20** using an aqueous solution of ferric chloride carried out by Asakura *et al.* led to the generation of the *ortho-ortho* dimer **28** (Scheme 20).[49] No polymerisation products were observed; this was ascribed to the fact that *p*-cresol was soluble in the reaction medium but the product was precipitated out of solution as oil droplets and thus removed from the oxidative system. Similarly, the formation of Pummerer's ketone* **32** (Figure 17) was not observed.

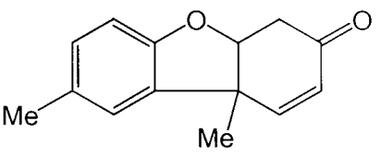
* Pummerer's ketone is a product arising from the *ortho-para* coupling of *p*-cresol and is a common by-product of *p*-cresol oxidation

Scheme 20



20 **28**

Figure 17 Pummerer's ketone

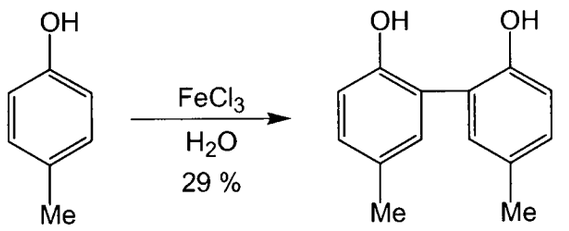


32

2.2.5.2 Application to the Model System

Using the method of Asakura *et al.* the dimer **28** was generated in 29 % yield using aqueous ferric chloride (Scheme 21).[49]

Scheme 21



20 **28**

Although the yield of dimer **28** was only moderate, this method had advantages:

1) As well as the ease of isolation of the crude product (which was found to precipitate out as a sticky solid), the reaction was a straightforward one, performed at room temperature with no observed susceptibility to other conditions.

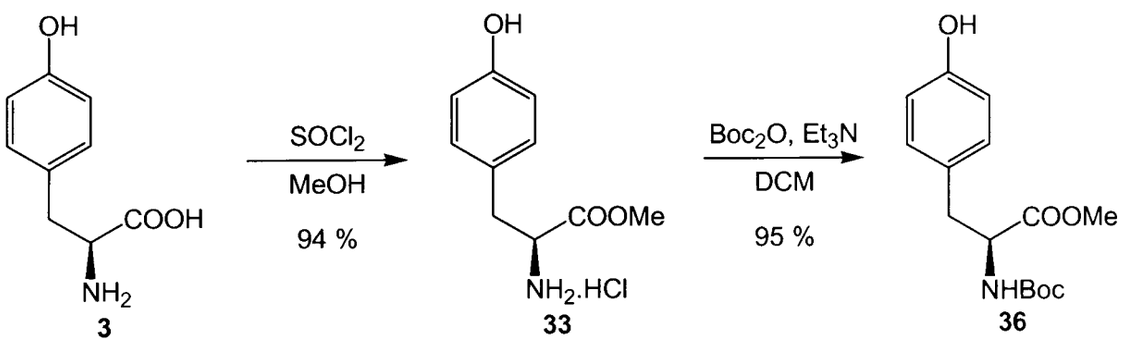
2) *p*-Cresol and ferric chloride are readily available at low cost.

3) The reaction was easily performed on a large scale.

2.2.5.3 Application to the Synthesis of the Herqulines

In order to apply this oxidative coupling reaction to the synthesis of the herqulines, a protected tyrosine molecule was synthesised. L-Tyrosine **3** was treated with thionyl chloride in methanol to give L-tyrosine methyl ester hydrochloride **33** (Scheme 22).[50] Since this method of esterification can result in racemisation of amino acids in some cases, D,L-tyrosine methyl ester **34** was synthesised utilising the same method (Scheme 23). Comparison of tyrosine **33** and the racemate **35** by chiral hplc (Figure 18) confirmed that no racemisation had occurred during esterification of L-tyrosine. The ester **33** was then treated with Boc anhydride and triethylamine to give *N*-Boc-L-tyrosine methyl ester **36** (Scheme 22).[51]

Scheme 22



Scheme 23

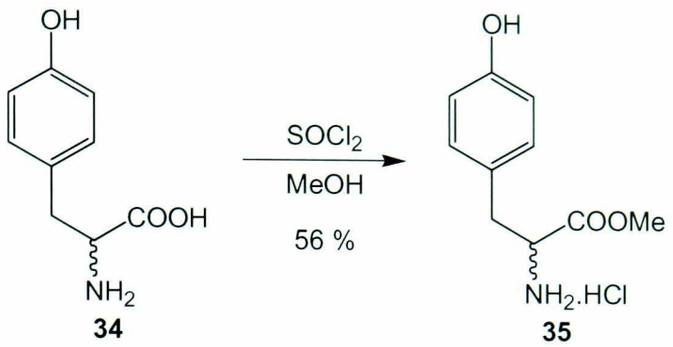
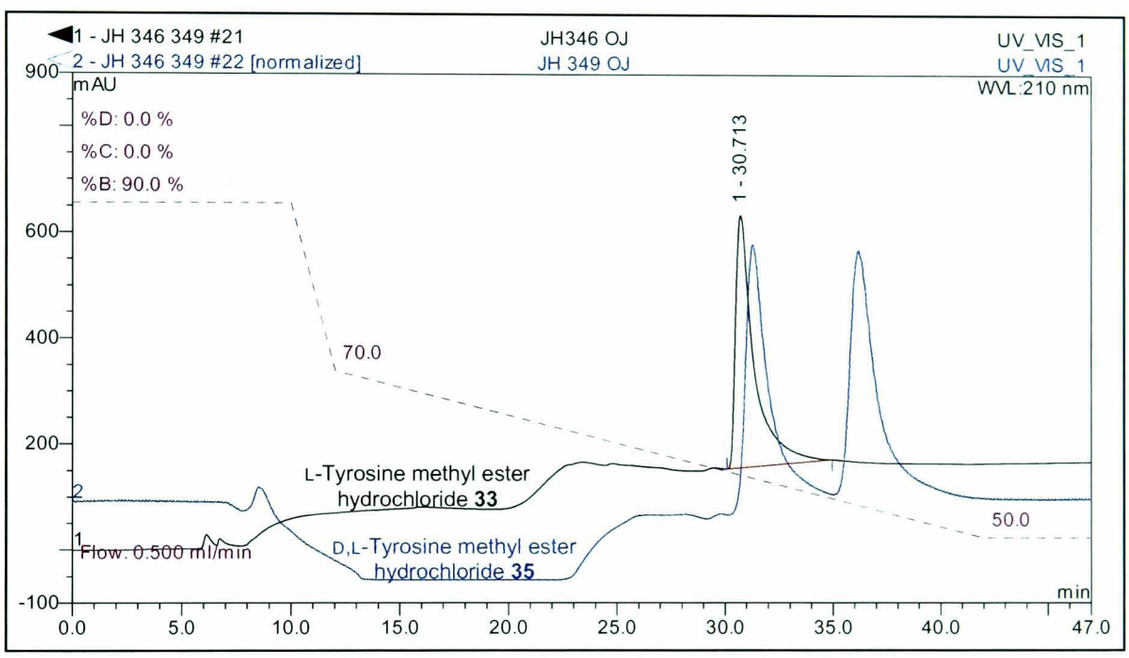


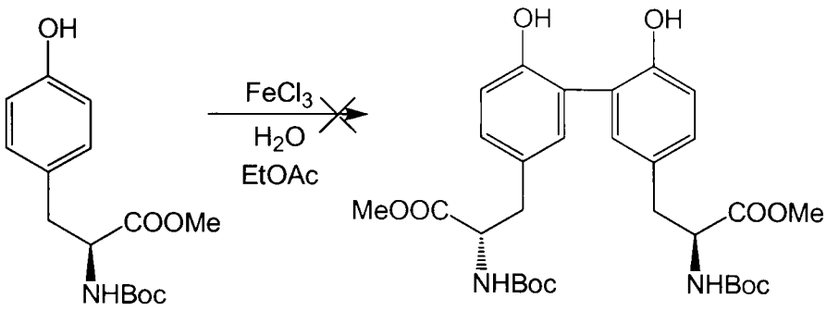
Figure 18



However, when the protected tyrosine **36** was treated with aqueous ferric chloride no reaction occurred (Scheme 24);* significantly, the colour change of yellow to dark blue observed during the oxidation of *p*-cresol – thought to be due to the formation of an iron-phenoxide complex[52] – did not occur.

* Asakura *et al.* found that addition of a small amount of ethyl acetate assisted the dimerisation of *p*-cresol. Ethyl acetate was used to facilitate dissolution of the starting material in this case.

Scheme 24



36 **37**

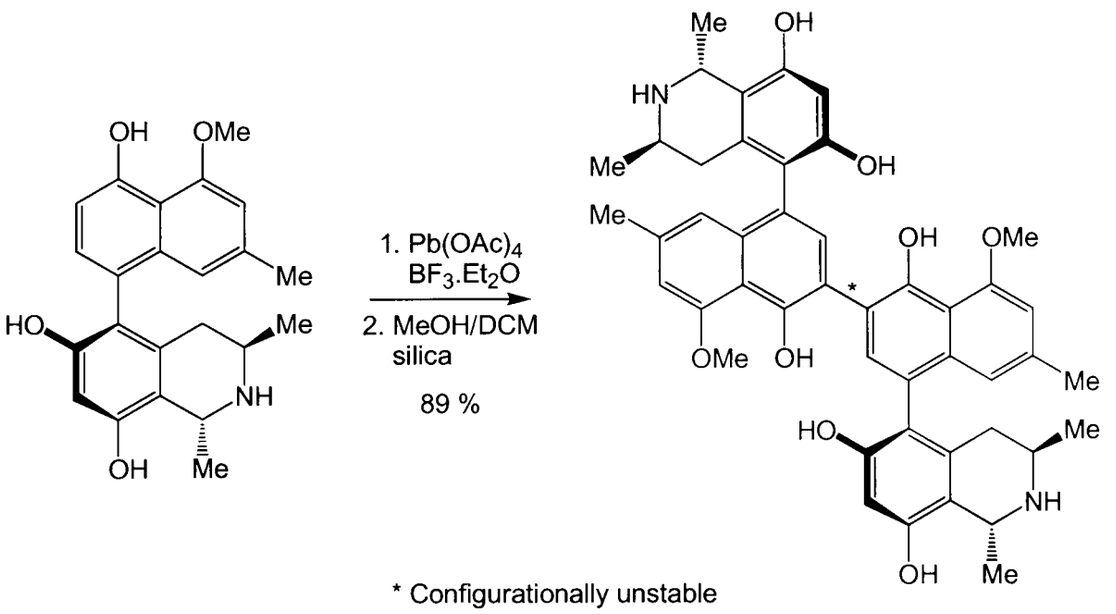
It is possible that the presence of other heteroatoms caused problems and prevented the formation of iron-phenoxide complexes.

2.2.6 Oxidation Using Lead Tetraacetate

2.2.6.1 Introduction

Lead tetraacetate is a well-known oxidising agent and has been widely used in the oxidative cleavage of vicinal diols,[53] oxidative cyclisation reactions[54, 55] and conversion of carboxylic acids to acid chlorides (the Kochi reaction).[56] It has also been used to perform oxidative phenol coupling reactions (Scheme 25).[57]

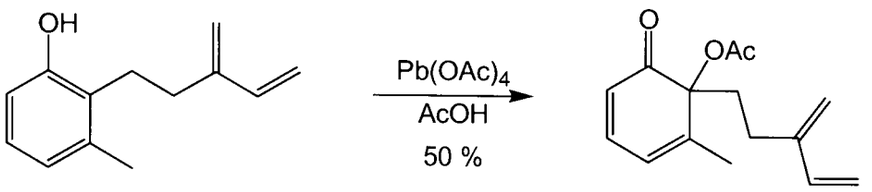
Scheme 25



* Configurationally unstable

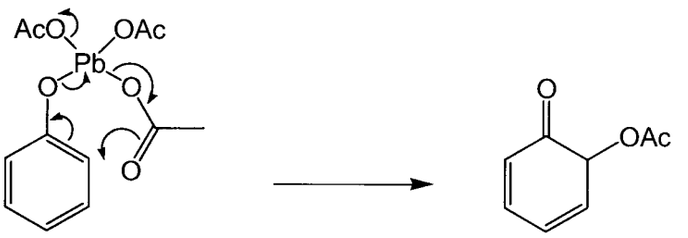
However, the treatment of phenols with lead tetraacetate in acetic acid often results in oxidation with addition of an acetate group, known as Wessely oxidation, rather than oxidative coupling (Scheme 26).[58]

Scheme 26 Wessely oxidation



Studies of the Wessely oxidation have indicated the existence of an intermediate lead-phenol complex which undergoes heterolytic decomposition (and therefore an NR1- or NR2-type mechanism). However, the treatment of phenols with lead tetraacetate in methanol also results in an acetoxy-substituted major product, suggesting an intramolecular mechanism (Scheme 27). This is analogous to the cyclic NR2 mechanism proposed earlier (Scheme 13, Section 2.2.1).[37, 59]

Scheme 27 Proposed intramolecular mechanism of the Wessely oxidation



2.2.6.2 Application to the Synthesis of the Herqulines

Lead tetraacetate was not utilised in attempts to generate the *p*-cresol dimer or dityrosine, but was used later in attempts at intramolecular coupling of tyrosine-based systems (see Section 2.2.7.2).

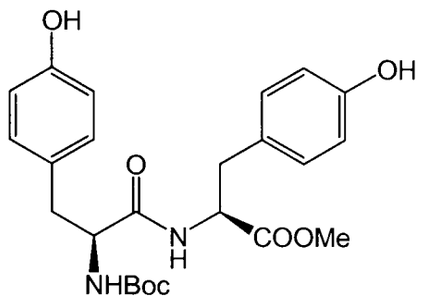
2.2.7 Intramolecular Coupling

2.2.7.1 Synthesis of a Tyrosine Dipeptide and Diketopiperazine

Achieving the oxidative phenol coupling of *p*-cresol proved useful for synthesising subsequent model compounds (see Sections 2.4.2, 3.2.6.2 and 3.4.2.2), but a method for

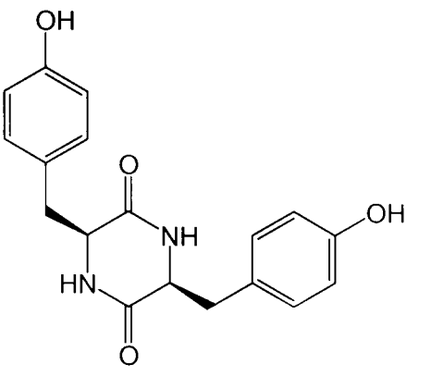
the synthesis of a dityrosine was still required. At this point, linking the lower part of the molecule by forming an amide bond (Figure 19a) or a diketopiperazine ring (Figure 19b) was considered. It was hoped that making the oxidative coupling reaction an intramolecular process would make it more favourable due to the smaller entropy of activation required.

Figure 19a



18a

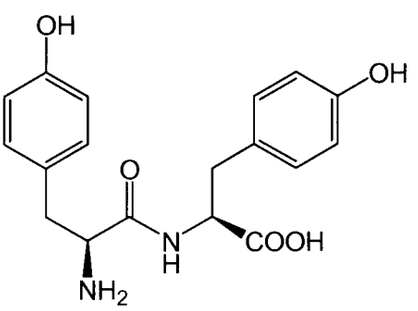
Figure 19b



19a

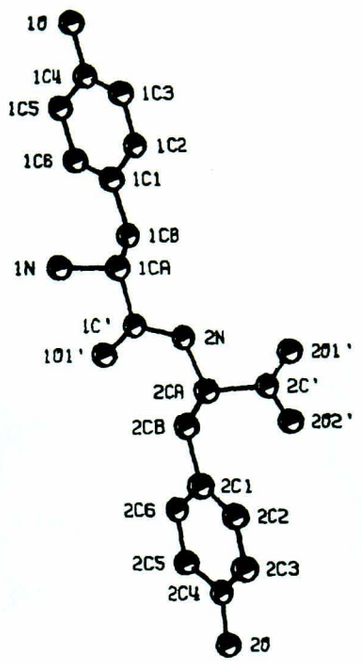
However, X-ray crystallography of (L-tyrosine)-(L-tyrosine) **38** (Figure 20) has shown that such dipeptides tend to adopt conformations in which the two aromatic rings are distant from each other. Figure 21 shows a crystal-structure conformation of (L-tyrosine)-(L-tyrosine);[60] Figure 22 shows the conformation of (L-tyrosine)-(L-tyrosine) in the active site of an enzyme.[61]

Figure 20



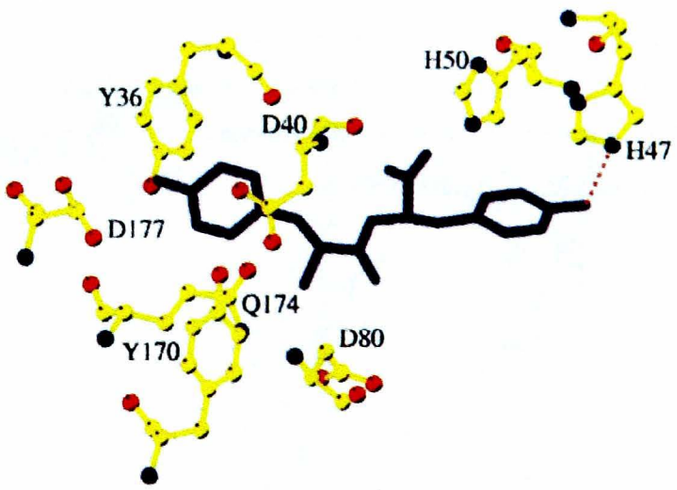
38

Figure 21 X-ray structure of (L-tyrosine)-(L-tyrosine) **38** (obtained from recrystallisation from water)[a]



[a] Reproduced from *International Journal of Peptide and Protein Research.*[60]

Figure 22 X-ray structure of (L-tyrosine)-(L-tyrosine) **38** in the active site of *Staphylococcus aureus* tyrosyl tRNA synthetase[a]



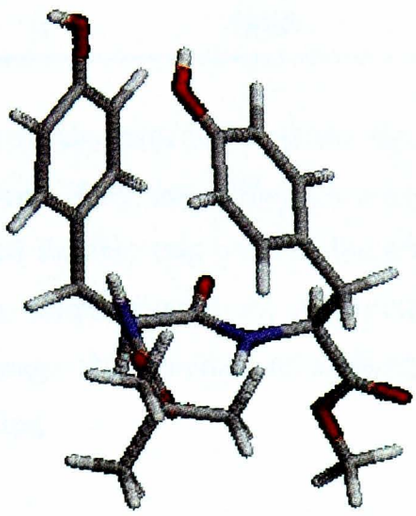
[a] Reproduced from *Bioorganic and Medicinal Chemistry Letters.*[61]

Since both the aqueous environment and the active site of an enzyme were likely to minimise any intramolecular interactions, molecular mechanics calculations were used to perform conformational searches in order to examine the low energy conformations of the dipeptide and diketopiperazine under consideration.* The dipeptide **18a** was

* Molecular mechanics calculations were routinely carried out using gas phase parameters; the dipeptide **18a** was also examined by conformational searching using aqueous parameters, with very similar results.

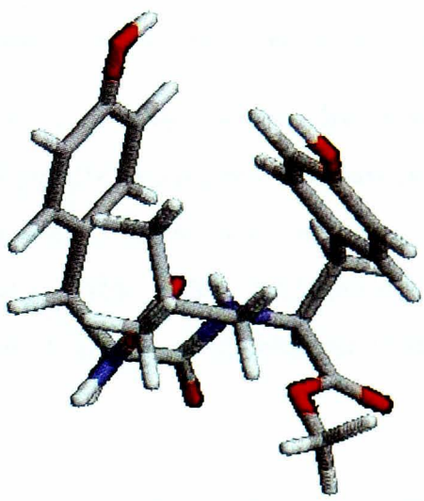
found to have three low-energy conformations (Figure 23a-c). The relative population of each conformation was then calculated using Boltzmann factors (Table 4).[62]

Figure 23a



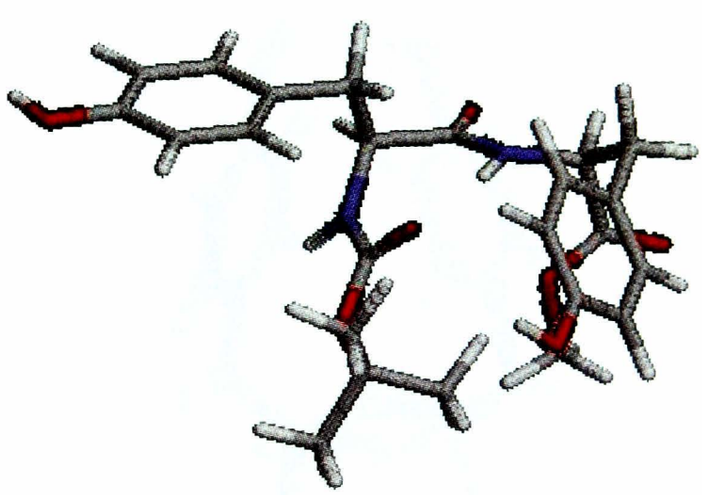
The aromatic rings are close together, with the potential for a π-stacking interaction and a possible weak hydrogen bond between the phenol OH groups.

Figure 23b



The aromatic rings are slightly further apart, with the potential for an edge-face interaction.

Figure 23c



The aromatic rings are far apart, so there are no π-interactions and no intramolecular hydrogen bonds.

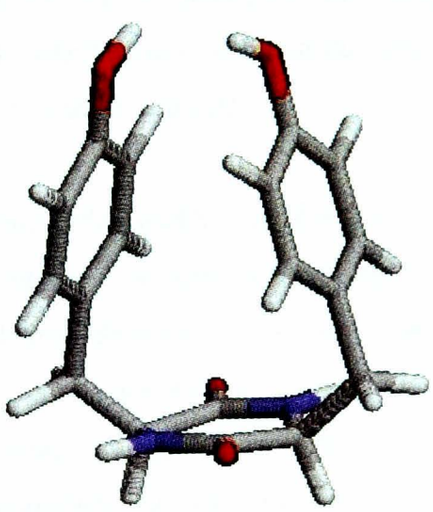
Table 4 Relative populations of the low-energy conformations of the dipeptide **18a**

Conformation	E (kJ mol^{-1})*	Boltzmann Factor† (at 25 °C)	Relative Population
a	178.3	5.54×10^{-32}	2.5
b	180.4	2.39×10^{-32}	1
c	180.6	2.90×10^{-32}	1

These results indicated that the dipeptide might be a suitable substrate for oxidative coupling. With little difference in energy between the three identified conformations, it seemed feasible that even in the presence of a highly polar solvent such as water – which would disrupt any intramolecular hydrogen bonds – the π-interactions may encourage the adoption of conformations able to undergo intramolecular oxidative coupling.

The diketopiperazine **19a** was found to have two low energy conformations (Figure 24a-b). The relative population of each conformation was calculated using Boltzmann factors (Table 4).

Figure 24a

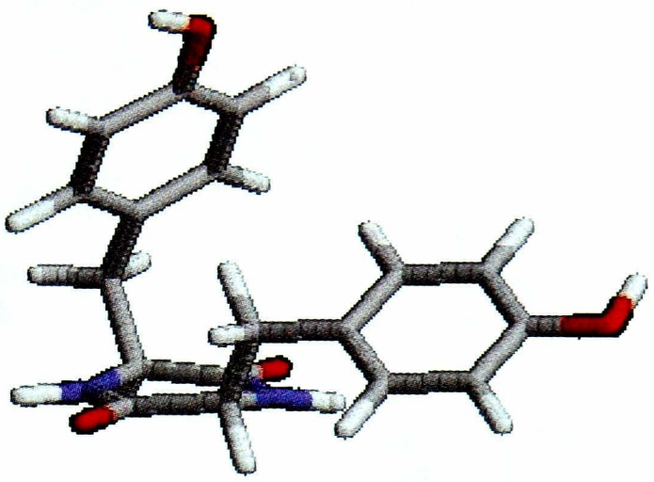


The aromatic rings are close together, with the potential for a π-stacking interaction and a possible weak hydrogen bond between the phenol OH groups.

* E refers to the 'molecular mechanics energy', which approximates to the internal energy (U) of a molecule. Molecular mechanics calculations take no account of entropic factors.

† Boltzmann factor: exp (–E/RT).

Figure 24b



One aromatic ring is above the diketopiperazine ring, while the other is twisted away from the rest of the molecule.

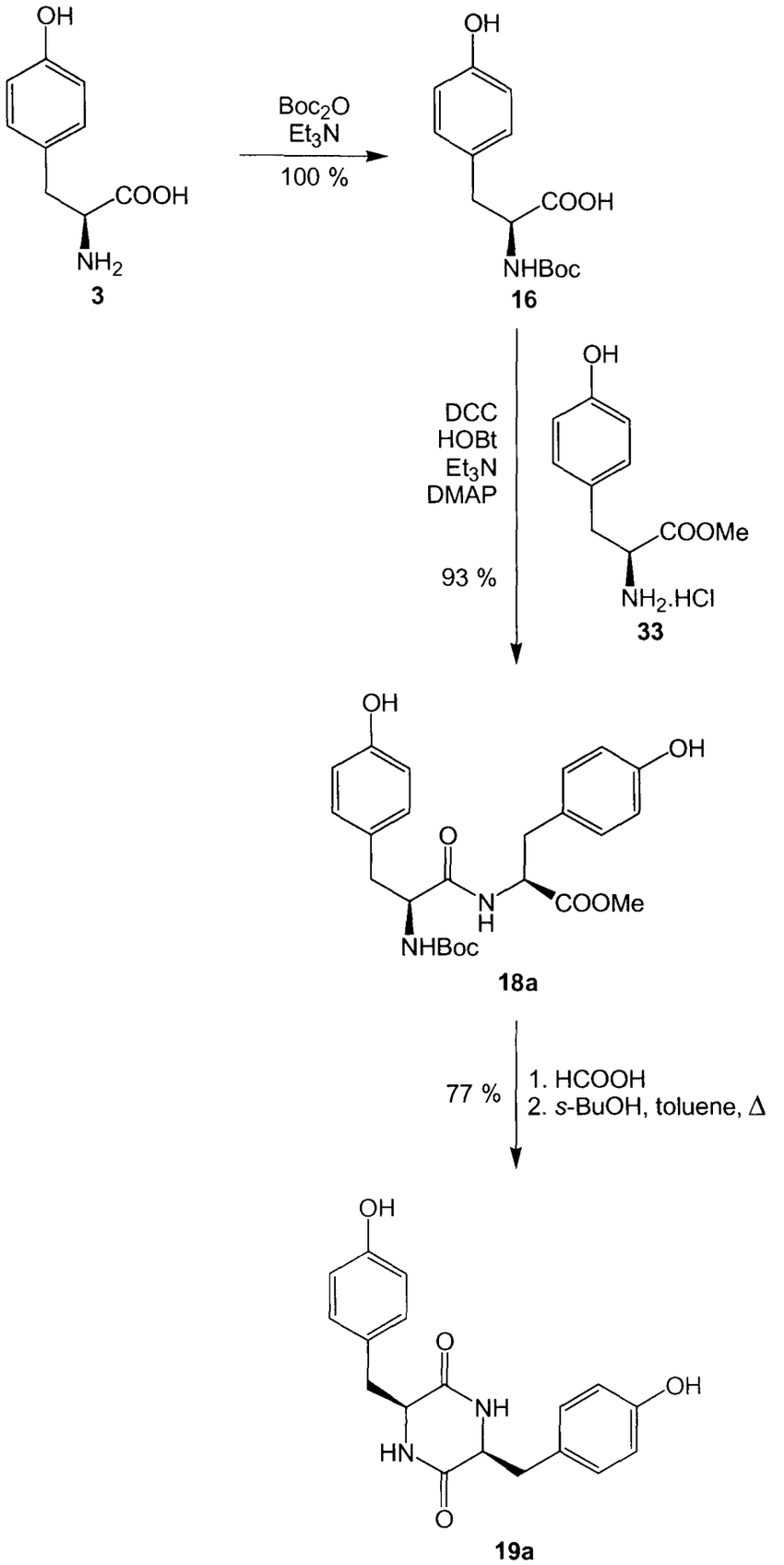
Table 5

Conformation	E (kJ mol^{-1})	Boltzmann Factor (at 25 °C)	Relative Population
a	97.5	8.11×10^{-18}	81
b	108.4	9.97×10^{-20}	1

These results confirmed that the diketopiperazine **19a** should be a suitable substrate for oxidative coupling, with the conformation in which the aromatic rings are close together (Figure 24a) being by far the most favourable.

In order to synthesise the molecules under consideration, L-tyrosine **3** was treated with Boc anhydride and triethylamine[51] to give *N*-Boc-L-tyrosine **16** in quantitative yield (Scheme 28). The Boc-protected tyrosine **16** was then treated with tyrosine methyl ester **33** and standard peptide coupling reagents to produce the protected dipeptide **18a** in excellent yield.[63, 64] Following the method of Nitecki, the dipeptide **18a** was treated with formic acid in order to remove the Boc protecting group and then heated at reflux in a mixture of *s*-butanol and toluene to give the corresponding diketopiperazine **19a** in 77 % yield.[65] Diketopiperazine formation was confirmed by the loss of signals corresponding to the methyl and *t*-butyl groups from the ^{1}H-NMR spectrum, as well as by simplification of the spectrum due to formation of a symmetrical molecule. The diketopiperazine **19a** was synthesised in 67 % overall yield from L-tyrosine.

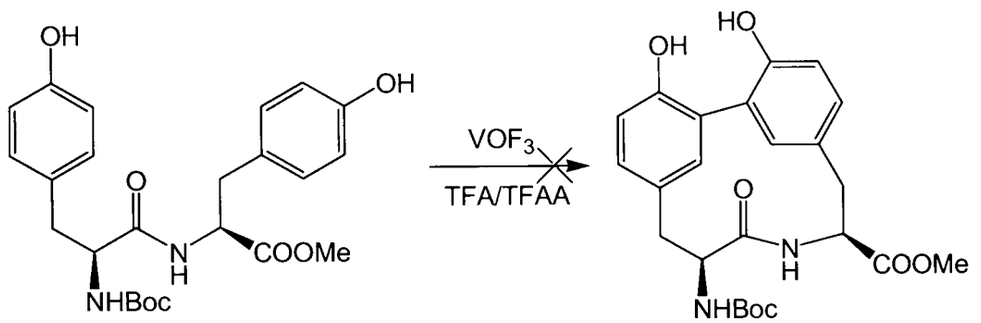
Scheme 28 Synthesis of cyclo(L-tyrosine)-(L-tyrosine) **19a**



2.2.7.2 Attempts at Intramolecular Oxidative Coupling

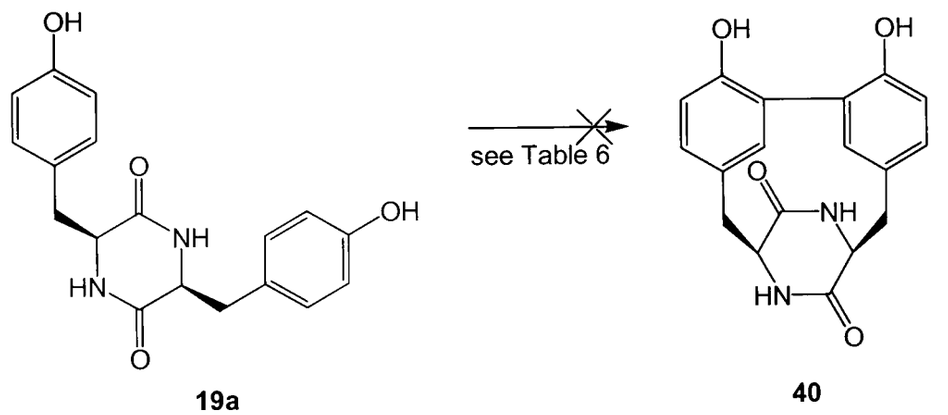
Having synthesised the dipeptide **18a** and the diketopiperazine **19a**, intramolecular oxidative coupling reactions were attempted. Treatment of the dipeptide **18a** (Scheme 29) and the diketopiperazine **19a** (Scheme 30) with vanadium oxytrifluoride resulted only in the isolation of vanadium salts or complexes. Application of the ferric chloride method to the diketopiperazine gave no reaction and the starting material was recovered. Again, the colour change of yellow to blue normally observed during the oxidation of *p*-cresol did not occur in this case.

Scheme 29



18a **39**

Scheme 30



19a **40**

Table 6 Reagents used in attempts to oxidise the diketopiperazine **19a**

Reagents	Outcome
VOF_3, TFA/TFAA	No product observed
$FeCl_3$	No reaction
$Pb(OAc)_4$, AcOH	No reaction

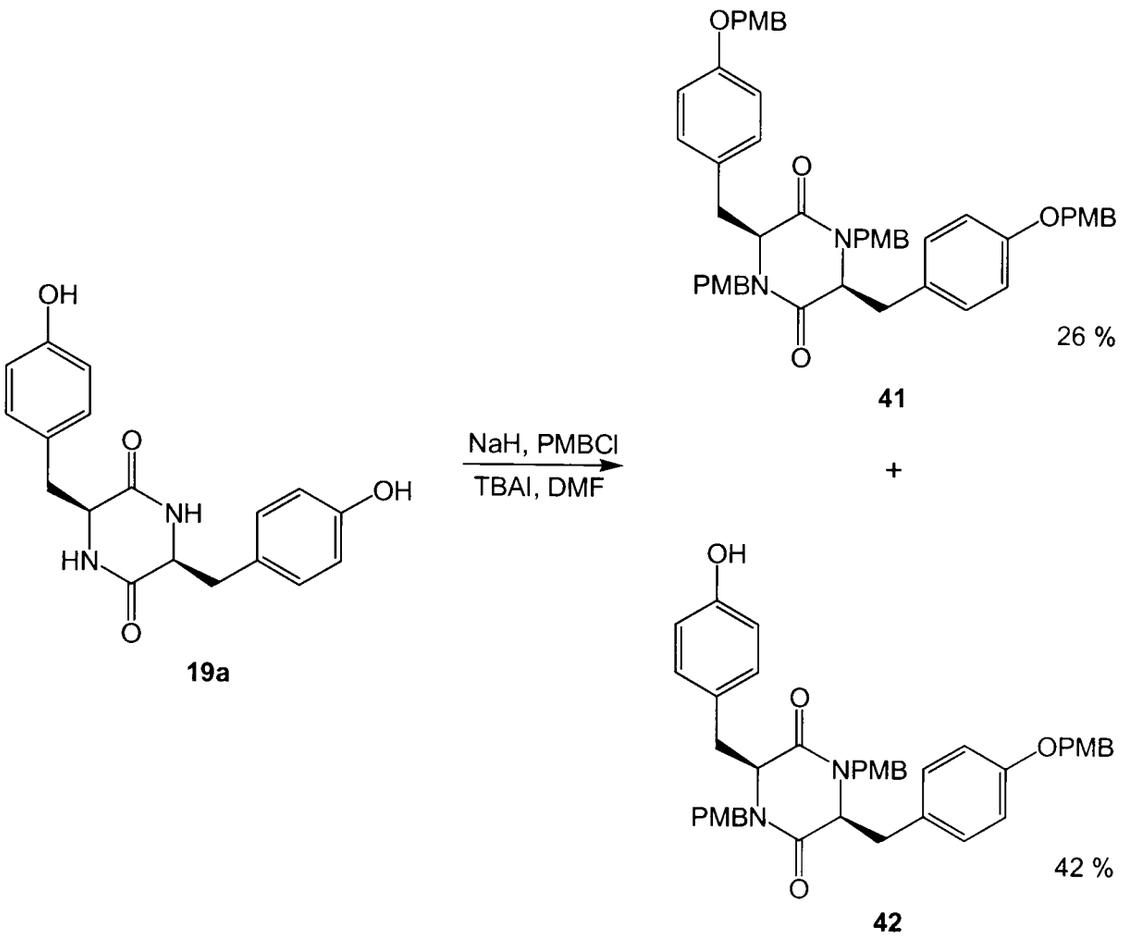
Working with diketopiperazines presented a significant problem since the compounds synthesised during the course of this project were found to be extremely polar and were soluble only in highly polar solvents such as DMF and DMSO. They were insoluble in most common solvents such as methanol, ethyl acetate, DCM, acetone and water. This made performing reactions and the subsequent isolation of products extremely difficult, and may go some way towards accounting for the failure of these oxidative coupling reactions. In an effort to overcome this problem, an oxidative coupling using lead tetraacetate was attempted, since the solvent commonly employed is acetic acid (in which diketopiperazine **19a** was soluble).[66] However, no reaction was observed and the starting material was recovered.

2.2.7.3 Protection of the Diketopiperazine

In an attempt to reduce the polarity of the diketopiperazine **19a** and make it easier to work with, suitable protection was sought. Protection of the amide NH groups while leaving the (more nucleophilic) phenol groups free to facilitate oxidative coupling was desired. The *p*-methoxybenzyl protecting group was chosen since this group has been used to protect amide groups. It is also possible to remove *O*-PMB groups in the presence of *N*-PMB groups using DDQ.[67] Consequently, a solution of the diketopiperazine **19a** in DMF was treated with sodium hydride, *p*-methoxybenzylchloride and TBAI to obtain the fully protected diketopiperazine **41** in 26 % yield (Scheme 31), confirmed by the presence of two doublets at 5.28 and 3.64 ppm corresponding to the NCH_2Ar protons and a singlet integrating for two protons at 4.98 ppm corresponding to the OCH_2Ar protons.[68]

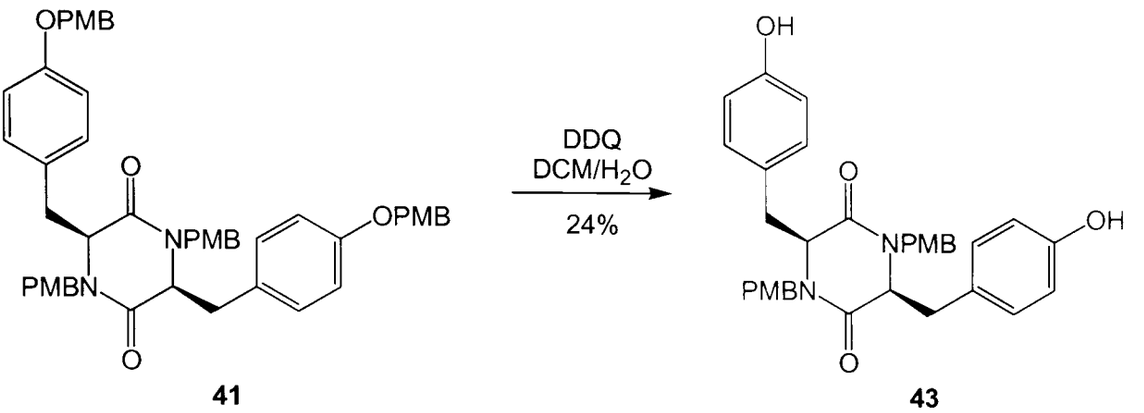
A tris-protected diketopiperazine **42** was also isolated in 42 % yield. This compound showed doublets at 5.27 and 3.37 ppm and a singlet at 3.75 ppm; all three signals exhibited equal integrations, indicating that the amide groups were protected but that only one of the phenol groups had undergone protection. Other signals in the ^{1}H-NMR spectrum appeared distorted when compared to that of the fully protected molecule **41**, indicating a loss of symmetry.

Scheme 31



The fully protected diketopiperazine **41** was then treated with DDQ in order to remove the *O*-PMB groups (Scheme 32).[69]. Flash chromatography of the crude product gave a small amount of the *N*-protected diketopiperazine **43**. This was confirmed by loss of the singlet at 4.98 ppm corresponding to the OCH_2Ar protons; the presence of doublets at 5.28 and 3.64 ppm indicated that the *N*-PMB groups had remained intact. Unfortunately, the low yields of both the protection and deprotection reactions meant that the amount of the *N*-protected diketopiperazine **43** obtained was not sufficient to attempt the oxidative coupling reaction.

Scheme 32



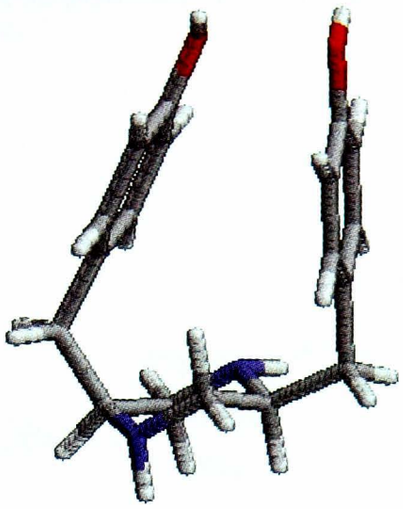
41 **43**

2.2.7.4 Reduction of the Diketopiperazine

A second approach towards making these compounds easier to handle was to reduce the diketopiperazine ring to a piperazine. This step would be required for the synthesis of herqulines A and B, and reduction should facilitate selective protection of the NH moieties.

Molecular mechanics calculations were used to examine low-energy conformations of the piperazine **15a**, in order to assess the feasibility of an intramolecular oxidative coupling reaction on the piperazine **15a** rather than the dipeptide **18a** or the diketopiperazine **19a**. The piperazine **15a** was found to have two low energy conformations (Figure 25a-b). The relative population of each conformation was calculated using Boltzmann factors (Table 7).

Figure 25a



The aromatic rings are close together, with the potential for a π-stacking interaction and a possible weak hydrogen bond between the phenol OH groups.

Figure 25b

The aromatic rings are far apart in order to minimise steric interactions.

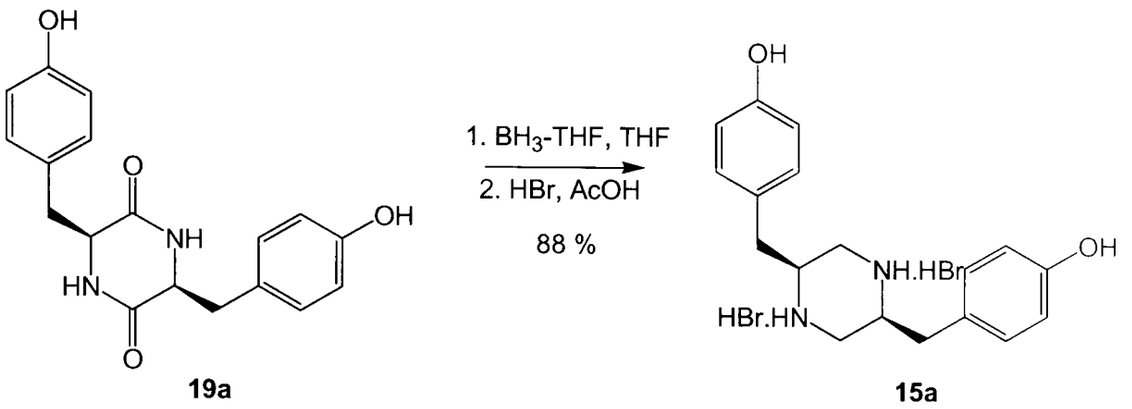
Table 7

Conformation	E (kJ mol^{-1})	Boltzmann Factor (at 25 °C)	Relative Population
a	54.9	2.38×10^{-10}	3
b	57.6	8.00×10^{-11}	1

These results indicate that the π-stacking interaction between the two aromatic rings in the piperazine **15a** is strong enough to make the conformation in which the two aromatic rings are close together (Figure 25a) the global energy minimum, despite steric factors.

Following the method of Jung, the diketopiperazine **19a** was reduced using borane-THF complex and the corresponding piperazine was isolated as its dihydrobromide salt **15a** in good yield (Scheme 33).[35]

Scheme 33

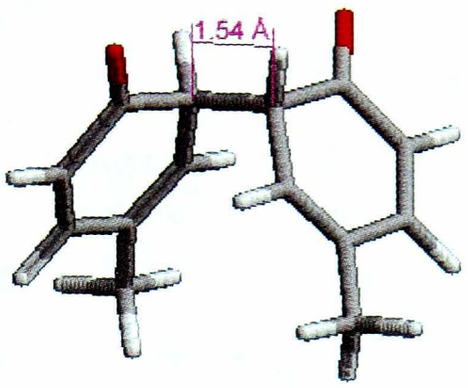


Having generated the piperazine **15a**, selective protection of the amines should have been straightforward. However, due to time constraints, this work was not completed.

2.2.8 Synthesis of Oxidative Coupling Transition State Mimics

As the oxidative coupling of the dipeptide **18a** and the diketopiperazine **19a** had been unsuccessful thus far, it was decided to test the theory behind the intramolecular coupling of the dipeptide **18a**. In order to allow oxidative coupling to occur, the two aromatic rings must be in close proximity. The bond length in the initially formed 'keto-tautomer' has been measured as 1.54 Å using conformational searching (the global minimum energy conformation of the 'keto-tautomer' of the *p*-cresol dimer **28** is shown in Figure 26).

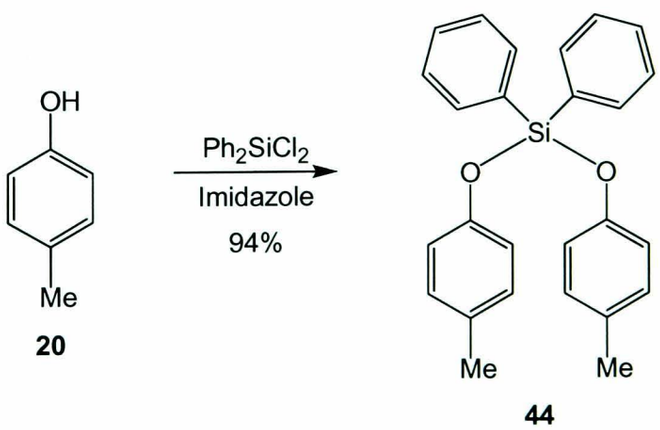
Figure 26



In order to confirm the molecular modelling results discussed earlier (Section 2.2.7.1) – which suggested that an intramolecular π-π-interaction could bring the aromatic rings into close proximity – the synthesis of a silicon-tethered dipeptide, mimicking the strained transition-state of an intramolecular oxidative coupling reaction, was attempted. This type of structure is also a mimic for the key intermediate in the cyclic NR2 mechanism (Scheme 13) thought to be involved in vanadium oxytrifluoride and lead tetraacetate oxidations.

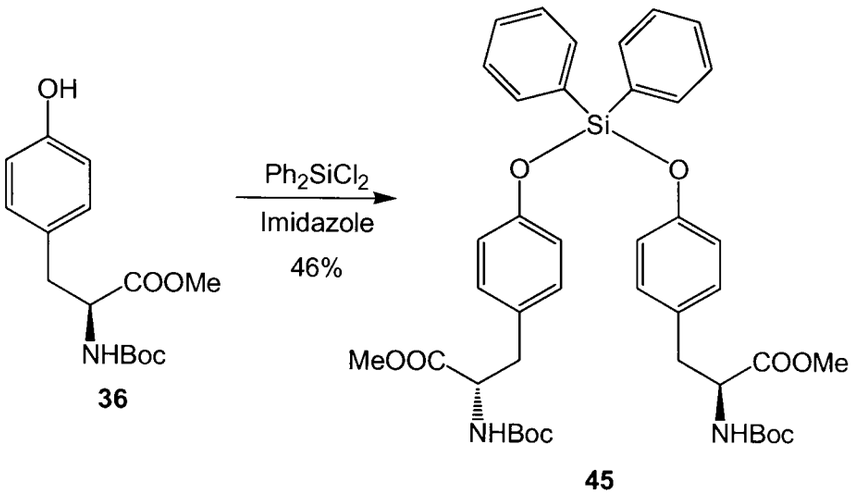
Intermolecular coupling was examined initially in order to validate the chemistry. Consequently, a bis(*p*-cresol)-substituted silane **44** was readily synthesised using *p*-cresol and dichlorodiphenylsilane (Scheme 34).[70]

Scheme 34



However using the same conditions, the corresponding silane **45** containing the protected tyrosine moiety was generated in a more moderate yield (Scheme 35), presumably due to the greater steric demand of the substituents.

Scheme 35



When the dipeptide **18a** was treated with dichlorodiphenylsilane, a mixture of products was formed (Scheme 36). The components were found to be inseparable by flash chromatography but were separated by reverse-phase hplc. However, hplc resulted in a significant loss of mass and only traces of products were isolated. A small amount of the desired silane **46** was obtained; a by-product – identified as cyclic siloxane **48** – was also isolated. This siloxane was thought to arise from bis-silylation of the amide **18a** and subsequent hydrolysis of the silicon-chlorine bonds during work-up and hplc (Figure 27). The formation of this bis-silylated amide **47** indicated that the formation of the cyclic silane **46** was retarded due to the ring strain involved. However, the clean hydrolysis of the amide **47** to give the siloxane **48** indicated that an increase in ring size from 17 to 19 may lead to a decrease in ring strain.

Scheme 36

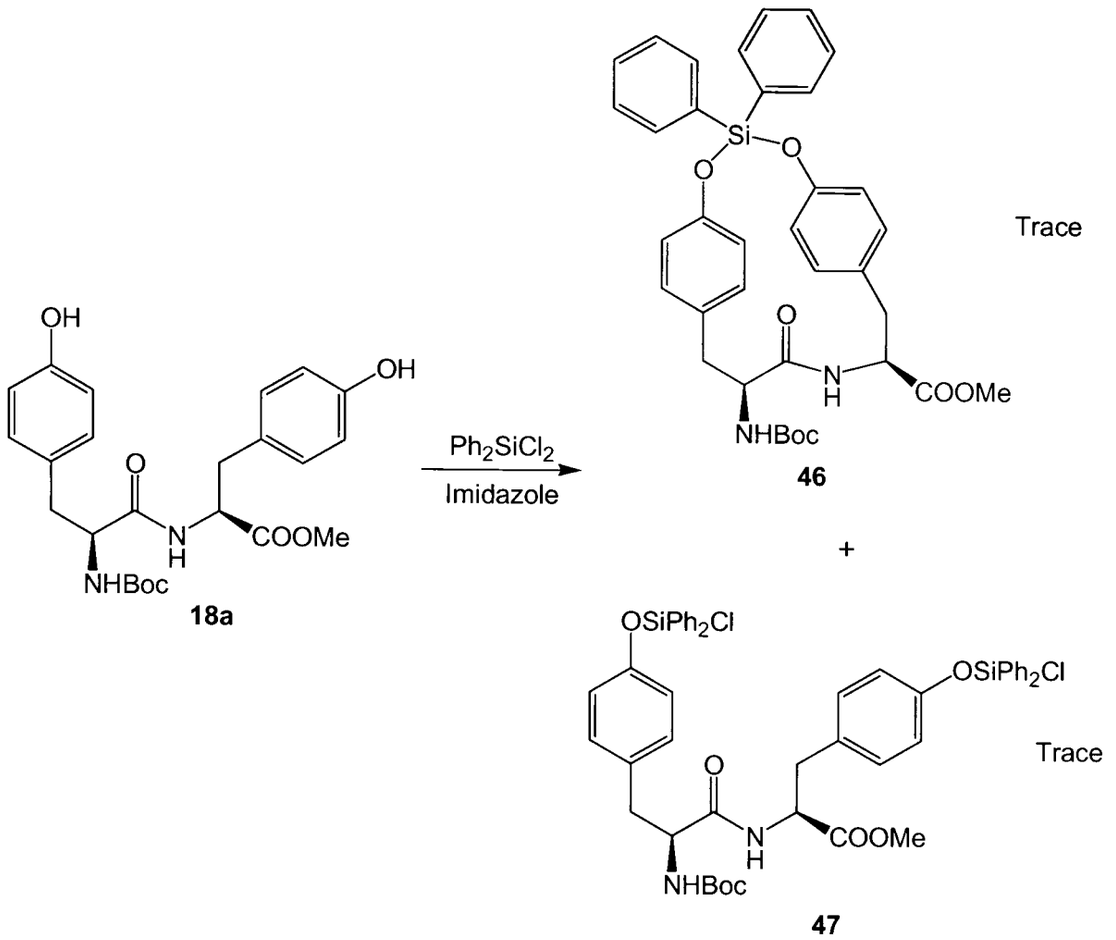
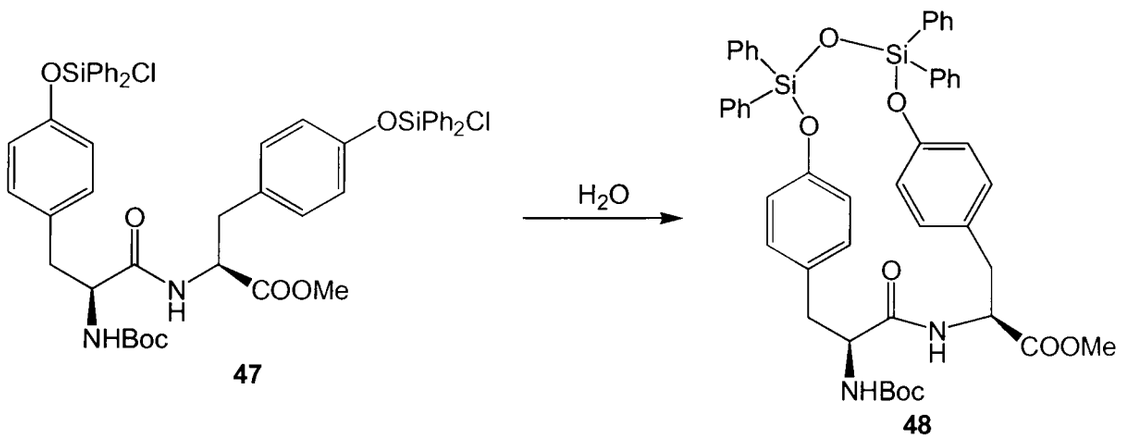


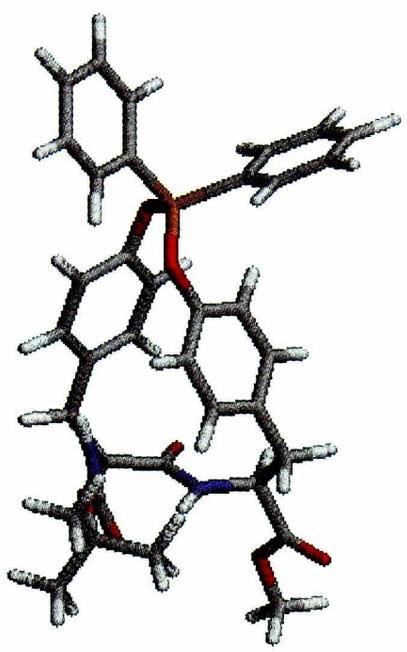
Figure 27



This work indicated that, despite the molecular modelling results, the amide **18a** may not be a suitable substrate for oxidative coupling as the formation of the required intermediate/transition state was likely to be hampered by ring strain. However, purification problems may also have played a role in the low yield of this reaction.

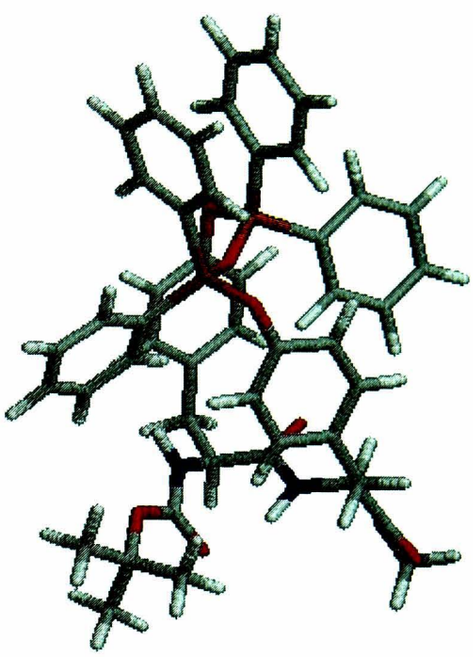
Molecular mechanics calculations were used to find the global energy minimum conformations of the siloxanes **46** and **48** (Figure 28a-b), and to compare the energies of the two molecules (Table 8).

Figure 28a



46

Figure 28b



48

Table 8

Siloxane	Diphenylsilyl units	Ring size	E (kJ mol^{-1})	Strain E (kJ mol^{-1})*
46	1	17	218	141
48	2	19	145	109
		ΔE	73	32

These results show that despite containing a greater number of atoms, siloxane **48** (containing two diphenylsilyl groups and a nineteen-membered ring) exhibited a lower energy value for both total and strain energies than siloxane **46** (containing one diphenylsilyl group and a seventeen-membered ring). This may account for the formation of the siloxane **48** on hydrolysis of the bis(chlorosilyl) compound **47**, rather than a bis(hydroxysilyl) compound.

* The strain energy given is the sum of terms relating to bond length, bond angles, dihedral angles and van der Waals interactions.

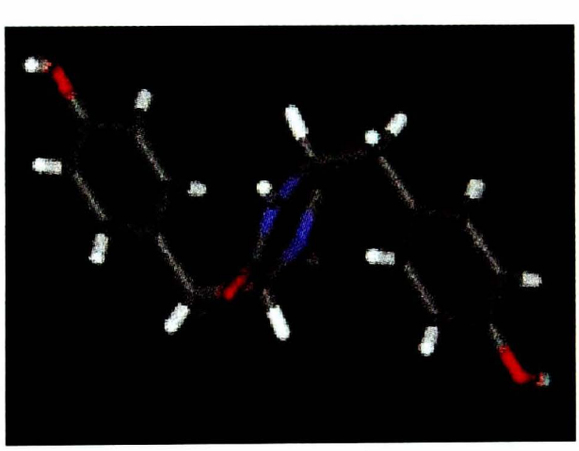
Attempts to synthesise a silicon-linked diketopiperazine failed, presumably due to solubility problems. However, information relating to the conformation of the diketopiperazine was still desired.

2.2.9 The Conformation of the Tyrosine Diketopiperazine

2.2.9.1 Introduction

The tyrosine diketopiperazine **19a** was obtained as a powder and attempts at recrystallisation failed. Therefore, a crystalline diketopiperazine derivative was sought in order to try to obtain an X-ray structure. The only set of X-ray data[71] reported for a tyrosine diketopiperazine to date related to cyclo(L-tyrosine)-(D-tyrosine) and was not therefore applicable to work relating to the herqulines. The stereochemistry of the molecule meant that the two aromatic rings were located on opposite sides of the almost planar diketopiperazine ring (Figure 29). However, it was interesting to note that both aromatic rings were orientated towards the diketopiperazine ring rather than away from it, indicating that there may be a favourable π-π interaction between the aromatic ring and the partially delocalised π-system of the diketopiperazine ring.

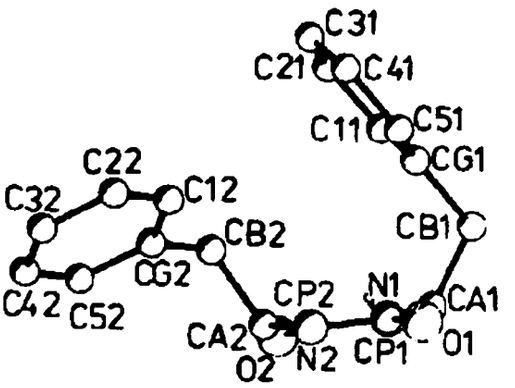
Figure 29[a]



[a] Reproduced using data from *Acta Crystallographica Section C.*[71]

The crystal structure of *cyclo*(L-phenylalanine)-(L-phenylalanine), in which both aromatic rings are located on the same side of the diketopiperazine ring, also showed one of the aromatic rings orientated towards the diketopiperazine ring (Figure 30).[72]

Figure 30[a]

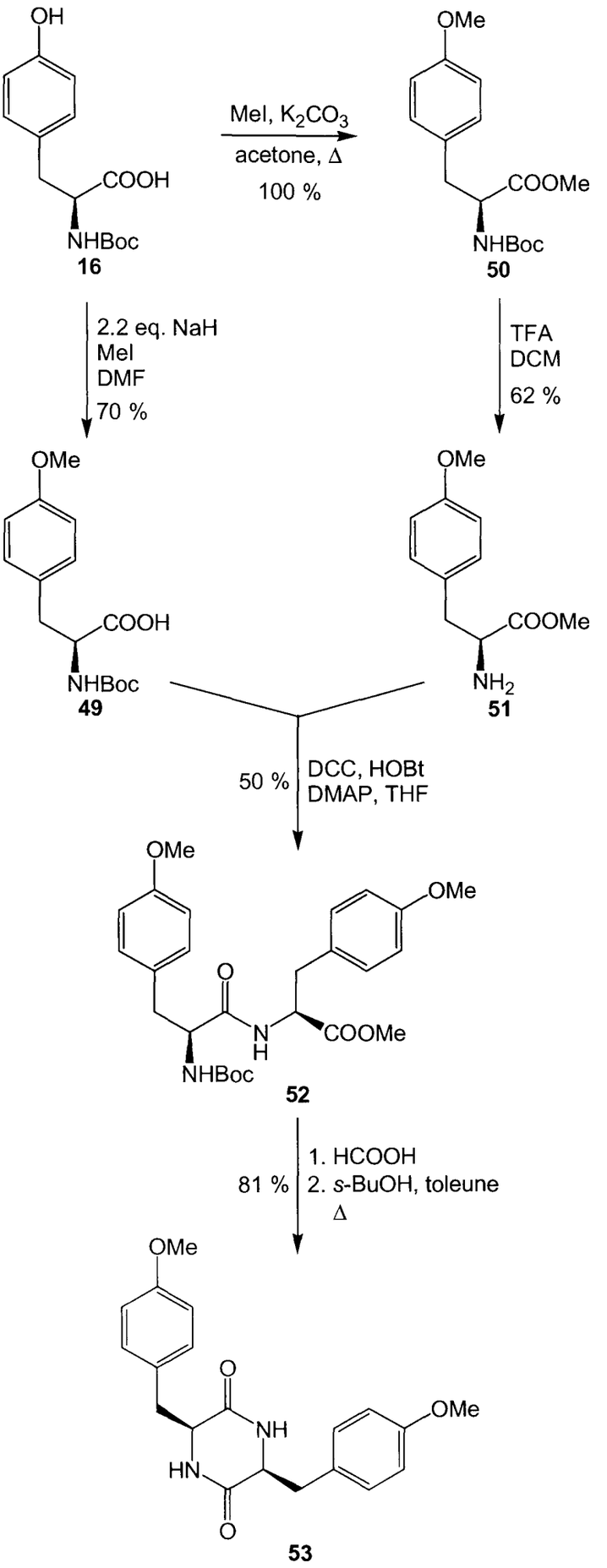


[a] Reproduced from *Acta Crystallographica Section C.*[72]

2.2.9.2 Synthesis of Diketopiperazine Derivatives

N-Boc-L-tyrosine **16** was treated with sodium hydride and one equivalent of methyl iodide in order to selectively protect the phenol group and generate the protected acid **49** (Scheme 37). The corresponding amine was synthesised by treatment of *N*-Boc-L-tyrosine with potassium carbonate and excess methyl iodide in order to generate the fully protected tyrosine **50**. The Boc group was then removed using TFA to afford the protected amine **51** in 62 % yield. The acid **49** and amine **51** were coupled using DCC and HOBt to give the methyl-protected dipeptide **52** in 50 % yield. Treatment with formic acid followed by heating at reflux in *s*-butanol and toluene to gave the corresponding protected diketopiperazine **53** in 81 % yield.

Scheme 37

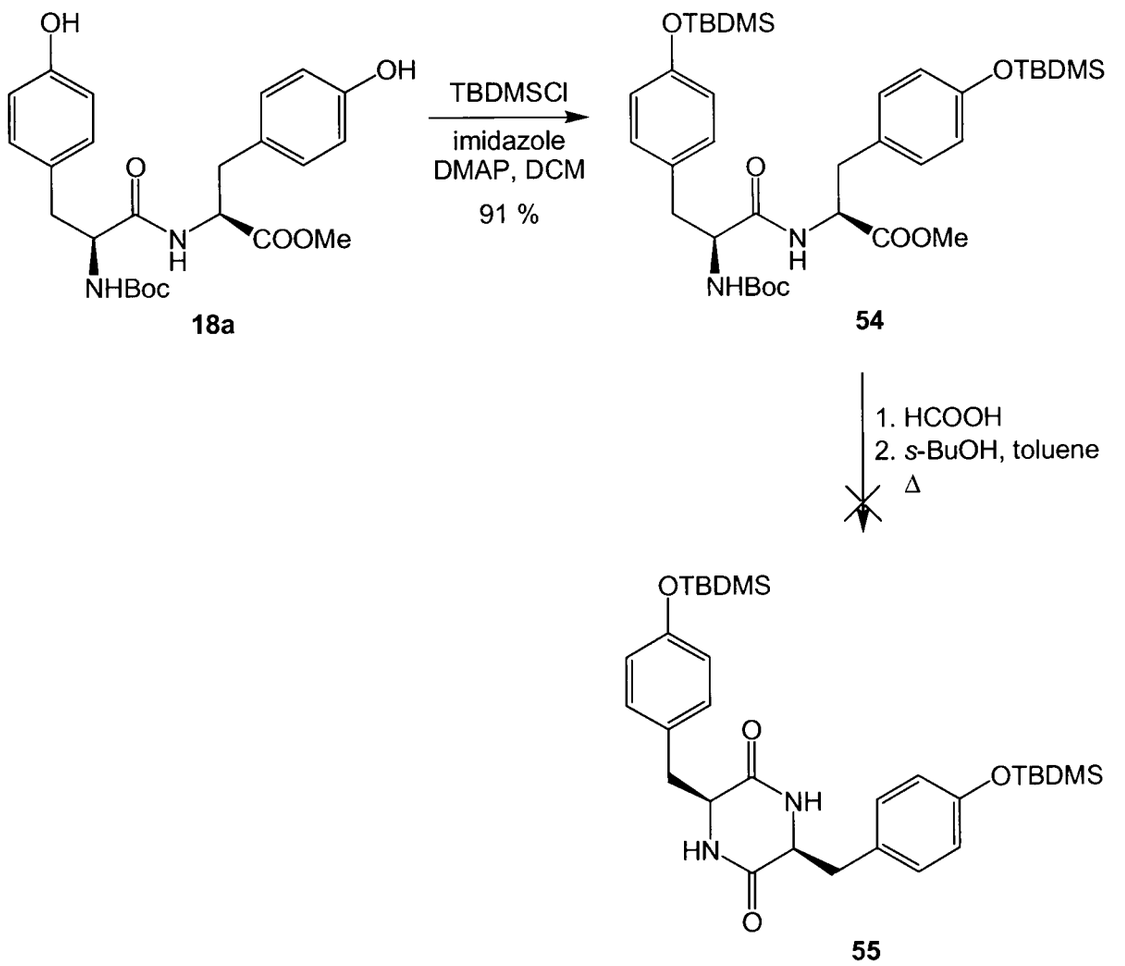

OH
COOH
NHBoc
16
MeI, K_2CO_3
acetone, Δ
100 %
OMe
COOMe
NHBoc
50
2.2 eq. NaH
MeI
DMF
70 %
TFA
DCM
62 %
OMe
COOH
NHBoc
49
OMe
COOMe
NH_2
51
50 %
DCC, HOBt
DMAP, THF
OMe
OMe
O
N
H
COOMe
NHBoc
52
81 %
1. HCOOH
2. s-BuOH, toluene
Δ
OMe
O
NH
HN
O
OMe
53

Unfortunately, the methyl-protected diketopiperazine **53** was also non-crystalline and was again soluble only in highly polar solvents such as DMF or DMSO.

Preparation of a silicon-protected derivative was then attempted since silicon-based protection usually confers a significant decrease in overall polarity of a molecule; it was hoped that this would facilitate recrystallisation.

Synthesis of the TBDMS-protected diketopiperazine **55** was attempted: the dipeptide **18a** was treated with TBDMS chloride and imidazole to give the protected dipeptide **54**. This was then treated with formic acid followed by heating at reflux in *s*-butanol and toluene to affect ring formation. However, desilylation occurred under these conditions and none of the desired diketopiperazine **55** was obtained.

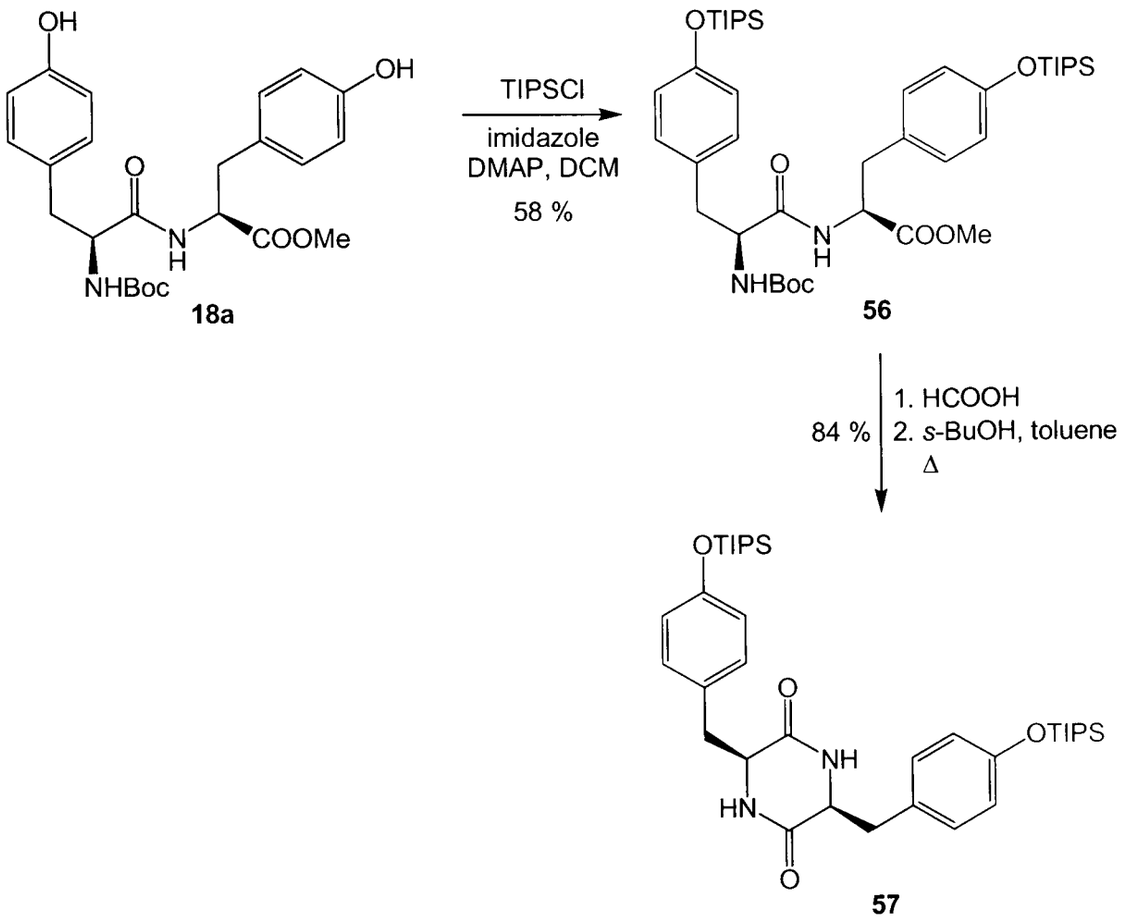
Scheme 38



Consequently, use of the TIPS protecting group was considered, since *O*-TIPS groups are known to be more stable under acid conditions than *O*-TBDMS groups.[73] The

dipeptide **18a** was treated with TIPS chloride and imidazole, and the corresponding protected dipeptide **56** was generated in 58 % yield. On treatment with formic acid followed by heating at reflux in *s*-butanol and toluene, the dipeptide formed the corresponding diketopiperazine **57** in 84 % yield.

Scheme 39



Having obtained the TIPS-protected diketopiperazine **57** (found to be soluble in DCM), recrystallisation was performed. Unfortunately, the crystals formed were not of a quality suitable for X-ray crystallography.

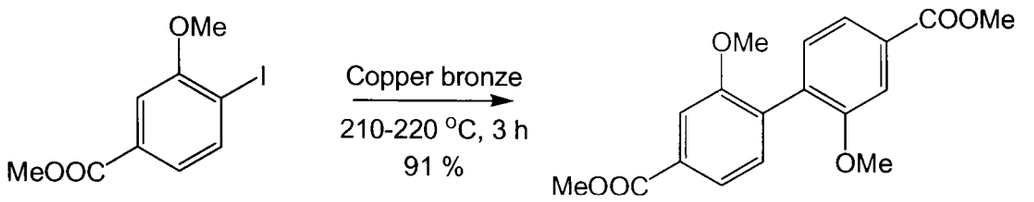
2.3 Ullmann Coupling

2.3.1 Introduction

A classical method for the homocoupling of aryl halides is the Ullmann reaction (Scheme 40).[74, 75] Ullmann coupling involves the heating of aryl halides (usually iodides) with copper, often at temperatures in excess of 200 °C. It is used to prepare

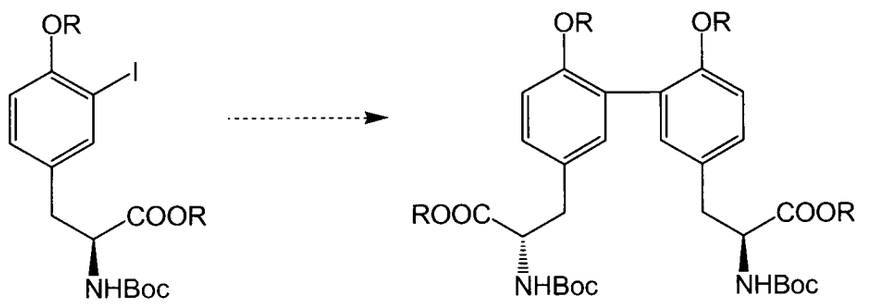
symmetrical biaryls and has a broad scope, including the coupling of *ortho*-substituted aryl halides.[75-78]

Scheme 40



However, despite the wide scope of the Ullmann reaction, the high temperature involved can present problems. Consequently, a number of other routes to these symmetrical products have been developed. A number of palladium-catalysed Ullmann reactions have been reported in the literature[79-81] as well as indium-mediated reactions[82] and nickel-catalysed systems.[83, 84] However, these types of coupling reaction are known to be hindered by bulky *ortho*-substituents and by electron donating substituents. Since the tyrosine system under consideration contained protected hydroxyl groups *ortho* to the aryl-aryl bond (Scheme 41), it was necessary to find a method that had been shown to be successful in the presence of such substituents.

Scheme 41



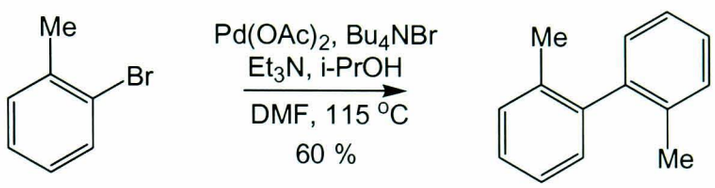
2.3.2 Palladium-Catalysed Ullmann Coupling

2.3.2.1 Introduction

Lemaire has performed the palladium-catalysed homocoupling of a number of substituted aryl halides using palladium acetate and tetrabutylammonium bromide in DMF.[36, 85] The method has also been applied to the coupling of halopyridines and haloquinolines. However, the group noted the detrimental effect of both electron-

donating substituents and steric hindrance caused by *ortho*-substitution. Consequently, the homocoupling of 2-bromotoluene **58** (Scheme 42) proceeded in moderate yield.

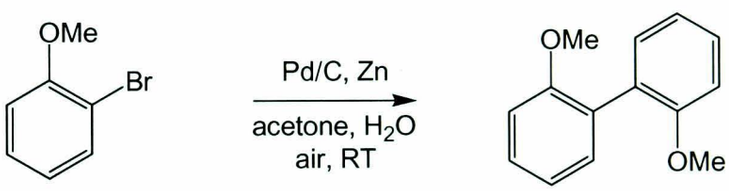
Scheme 42



58 **59**

A similar reaction catalysed by palladium on charcoal has been reported by Li (Scheme 43).[81] The reaction is unusual in that it occurs readily in the presence of both air and water; indeed, the presence of water has been shown to be necessary for catalytic activity of the palladium.

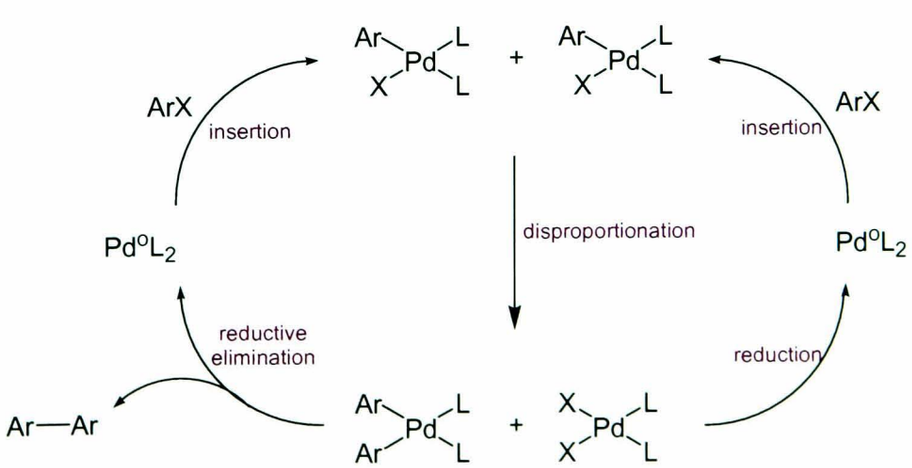
Scheme 43



60 **61**

These palladium-catalysed Ullmann reactions are thought to proceed *via* a catalytic cycle involving insertion into the C-X bond, disproportionation, reductive elimination and a reduction step (Scheme 44).[80]

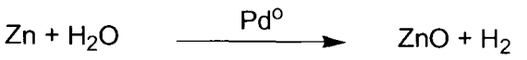
Scheme 44



The reducing agent in the reaction described by Lemaire (Scheme 42) has not been fully elucidated, but is thought to be the DMF used as a solvent.[86]

Li has proposed that zinc acts as the reducing agent in the aqueous reaction shown in Scheme 43, with electron transfer from zinc to palladium(II) occurring either directly or *via* the solvent.[80] However Mudkhopadhyay *et al.* have suggested that a reaction between zinc and water leads to the formation of hydrogen gas, which then acts as the reducing agent.[87]

Scheme 45



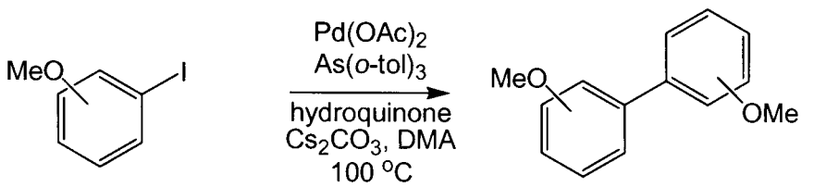
This theory would account for the failure of the reaction in the absence of water. However, following the survival of a benzylic ether under the reaction conditions – such functional groups are normally susceptible to hydrogenation in the presence of hydrogen and palladium on charcoal – Li has concluded that hydrogen gas participation is unlikely.[80]

However, no explanation has yet been found (relating to reduction either *via* zinc or hydrogen gas) for the stability of organopalladium species under the reaction conditions. Organopalladium species and the mechanisms relating to their formation and reaction are generally considered highly sensitive to the presence of water and oxygen, yet in this reaction they are essential and tolerated respectively. Li has studied aqueous reactions catalysed by palladium, rhodium and ruthenium and has observed that the reactions are much less air-sensitive that the corresponding reactions in organic solvents. He has suggested that the hard and soft acid/base theory may have a role in the stability of carbon-metal bonds in water: water molecules are hard and therefore interfere little with the soft late-transition metals.[88] It is however unclear how this relates to the decreased air-sensitivity of organometallic species in water.

A further example of a palladium-catalysed Ullmann reaction has been reported by Rawal (Scheme 46, Table 9);[79] a range of simple substituted aryl iodides and bromides were homocoupled successfully. Homocoupling was most efficient when performed on aryl iodides, but aryl bromides gave moderate to good yields depending on other

substituents. Similarly, the reaction occurred most rapidly when tri-*o*-tolylarsine was used as a ligand, but tri-*o*-tolylphosphine also gave moderate to good yields when longer reaction times were employed. A number of different bases were tolerated, but the use of hydroquinone as the reducing agent was found to be essential for successful homocoupling.

Scheme 46



It was interesting to note the effect that the position of the methoxy group had on the efficiency of the reaction (Table 9).

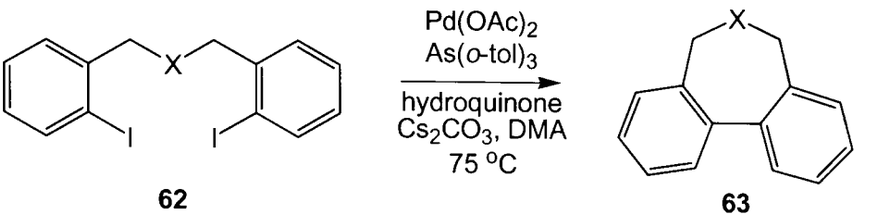
Table 9 The effect of methoxy-substitution on homocoupling (Scheme 46)

Position	Temperature (°C)	Catalyst loading (%)	Reaction time (h)	Yield (%)
para	75	2	3	95
meta	75	2	5.5	96
ortho	100	4	48	82

Clearly the presence of an *ortho*-methoxy substituent retarded the reaction, but use of a longer reaction time, greater catalyst loading and a higher temperature meant that a good yield of the desired product was still obtained.

Rawal also used this method to facilitate intramolecular couplings (Scheme 47, Table 10). Seven-membered heterocyclic and carbocyclic rings were formed in yields of 61 to 82 % despite the *ortho*-substitution that reduced yields in the intermolecular reaction. The smaller activation entropy required appeared to compensate for the steric hinderance conferred by *ortho*-substitution.

Scheme 47



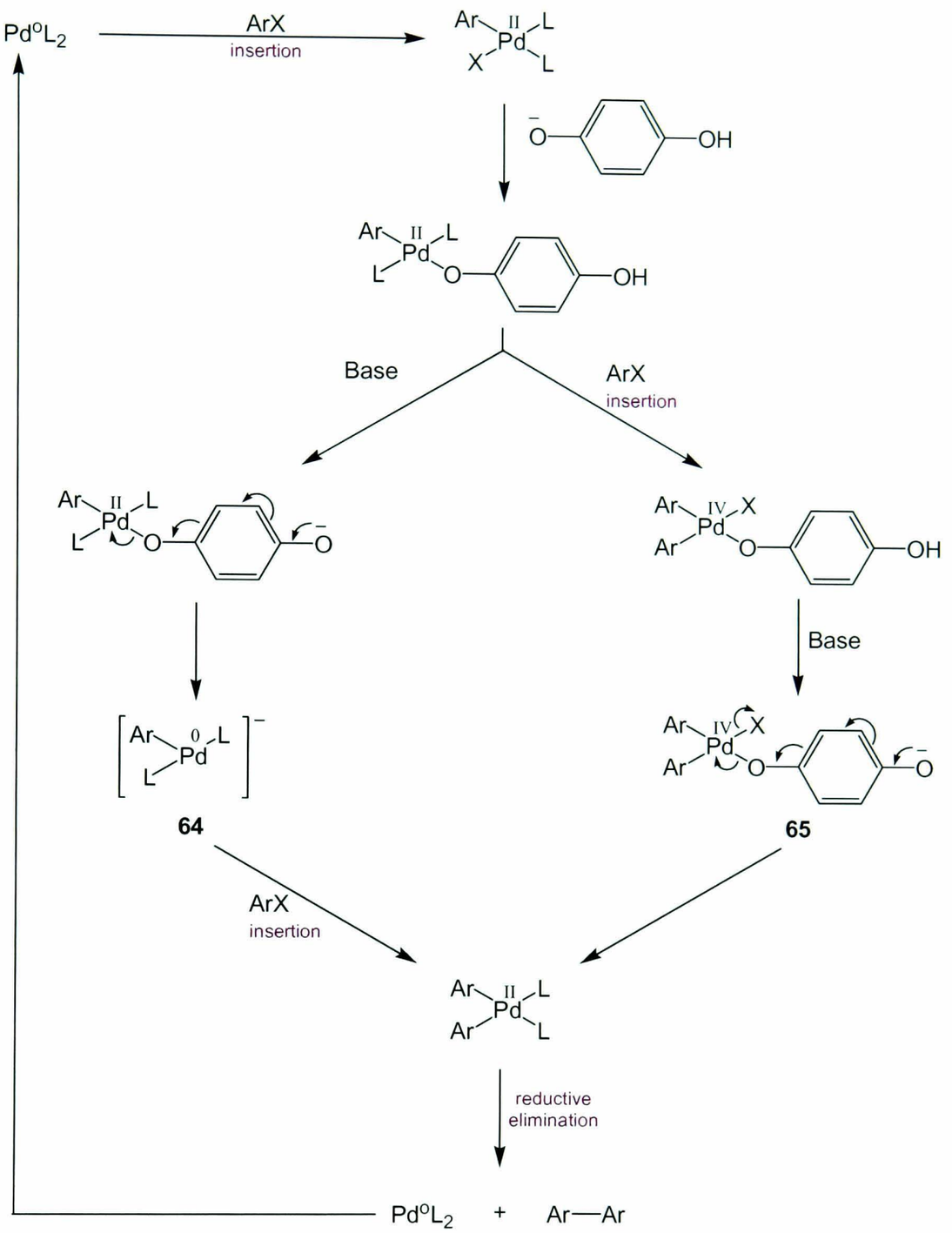
62 **63**

Table 10 Yields of intramolecular palladium-catalysed Ullmann couplings (Scheme 47)

X	% Yield
NTs	82
O	61
$C(COOEt)_2$	65

The authors proposed a mechanism accounting for the essential nature of hydroquinone in the reaction, involving either an anionic arylpalladium species[89] **64** or a palladium(IV) intermediate **65**. Consequently, this mechanism does not involve a disproportionation step.

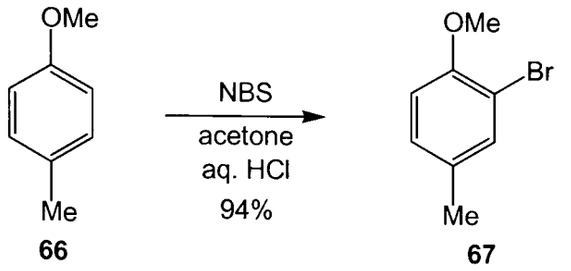
Scheme 48



2.3.2.2 Application to a Model System

In order to test the application of these reactions to the system under consideration, a model compound was used initially. Commercially available 2-bromo-4-methyl anisole **67** was used to test the reaction conditions since it has the same substitution pattern as a halogenated tyrosine. When larger quantities were required, it was synthesised in excellent yield from 4-methylanisole **66** (Scheme 49).[90]

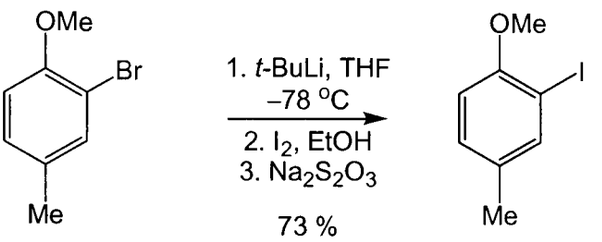
Scheme 49



66 **67**

The use of an aryl iodide instead of an aryl bromide to compensate for decreased reactivity due to *ortho*-substitution was considered. 2-Iodo-4-methyl anisole **68** was therefore synthesised from 2-bromo-4-methyl anisole **67** as shown in Scheme 50.[91, 92]

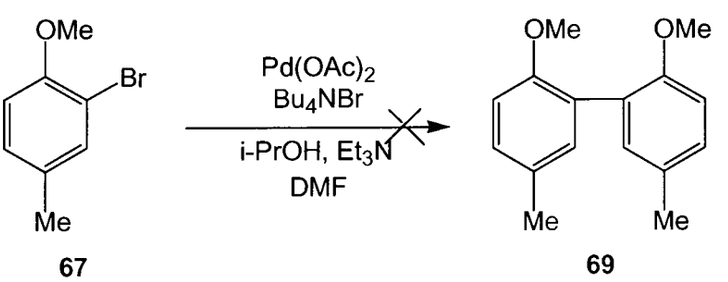
Scheme 50



67 **68**

When the conditions reported by Lemaire were applied to the model aryl bromide **67** no reaction occurred and the starting material was recovered; the presence of two electron-donating substituents on the ring appeared to be responsible for a significant decrease in reactivity.

Scheme 51

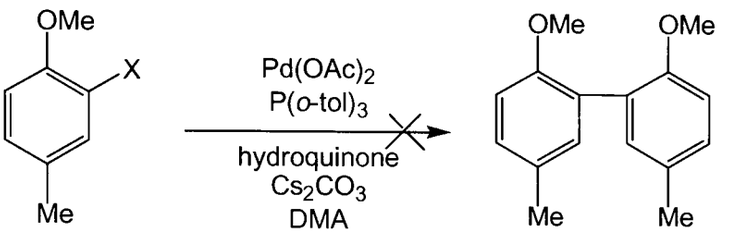


67 **69**

The palladium acetate-catalysed reaction reported by Rawal[79] was also attempted using the model aryl bromide **67** (Scheme 52); again no reaction was observed and the starting

material was recovered. Similarly when the reaction was repeated using the model iodide **68**, none of the dimer **69** was formed.

Scheme 52

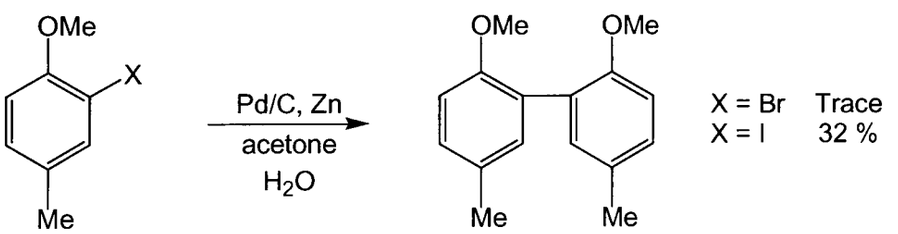


67 X = Br
68 X = I

69

The model aryl bromide **67** was also used to test the aqueous reaction conditions reported by Li (Scheme 53). ^{1}H-NMR spectroscopy of the crude product confirmed the presence of the desired product through an upfield-shift of the aromatic proton signals; however, the dimer **69** was isolated in less than 1 % yield. The reaction was more successful when repeated using the model iodide **68**; however ^{1}H-NMR spectroscopy indicated that a significant amount of reductive dehalogenation was also occurring, with the desired dimer **69** being obtained in only 32 % yield and the reduction product 4-methylanisole **66** in 27 % yield.

Scheme 53



67 X = Br
68 X = I

69

2.3.3 Nickel-Mediated Ullmann Coupling

2.3.3.1 Introduction

The nickel-catalysed homocoupling of aryl halides has been the subject of much work due to the relatively mild conditions employed, and the tolerance of many functional groups.[86]

Rieke has used metallic nickel to facilitate these reactions,[93] and good yields have been observed for unsubstituted and *para*-substituted aryl iodides (the corresponding aryl bromides gave only moderate yields of the desired products). However, the use of *ortho*-substituted aryl iodides resulted in less than 1 % yield of the desired product, with the major product arising from reductive dehalogenation. This illustrates the significant effect that *ortho*-substituents can have on these reactions (Scheme 54, Table 11).

Scheme 54

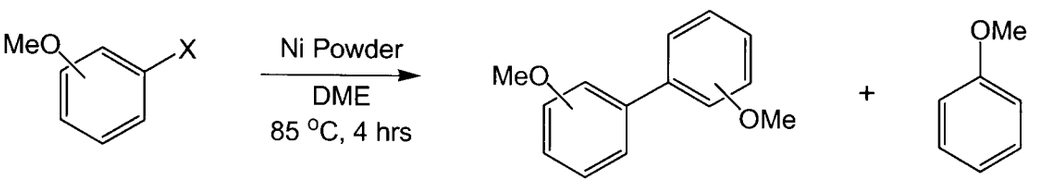


Table 11 Effect of methoxy-substitution on nickel-mediated homocoupling (Scheme 54)

Substitution	X	Reaction time (hours)	% Dimer	% Anisole **70**
para	Br	18	57	43
para	I	2	85	15
ortho	I	4	<1	85

The reductive dehalogenation process is in competition with homocoupling. These results indicate that in this case, the presence of an *ortho*-substituent has hindered the homocoupling reaction so much that it has become much slower than dehalogenation; hence the major product is anisole **70**.

Iyoda reported the reductive homocoupling of aryl halides using bis(triphenylphosphine)nickel(II) dibromide (Scheme 55, Table 12).[84] *Ortho*-substituted molecules have been coupled in good yield with the use of a higher catalyst loading and longer reaction time than needed for *meta*- or *para*-substituted aryl halides.

Scheme 55

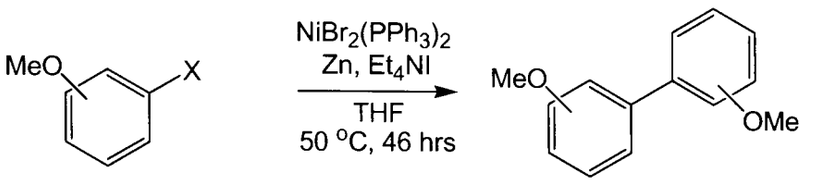
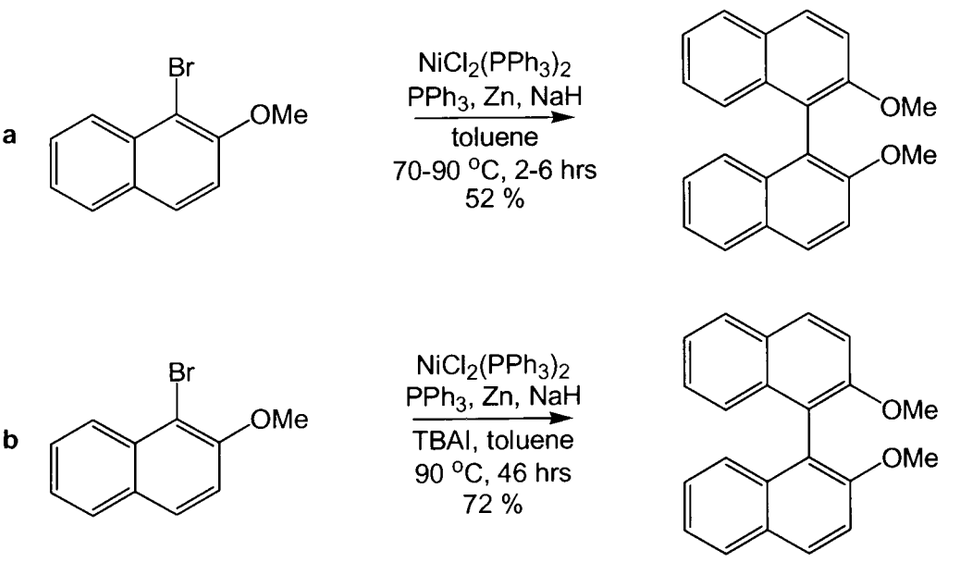


Table 12 Effect of methoxy substitution on bis(triphenylphosphine)nickel(II) dibromide-mediated homocoupling (Scheme 55)

Substitution	X	Reaction time (hours)	% Catalyst loading	% Dimer
para	Cl	20	10	67
para	Br	4	10	72
ortho	Br	46	50	81

A variation of this method employing sodium hydride was reported by Lin,[83] and substrates with *ortho*-substituents were coupled in moderate yields (Scheme 56a). A later publication revealed that the addition of tetra-butyl ammonium iodide to the system facilitated the coupling of *ortho*-substituted aryl halides, and allowed the coupling of a range of bis-*ortho*-substituted molecules in high yield (Scheme 56b).[94]

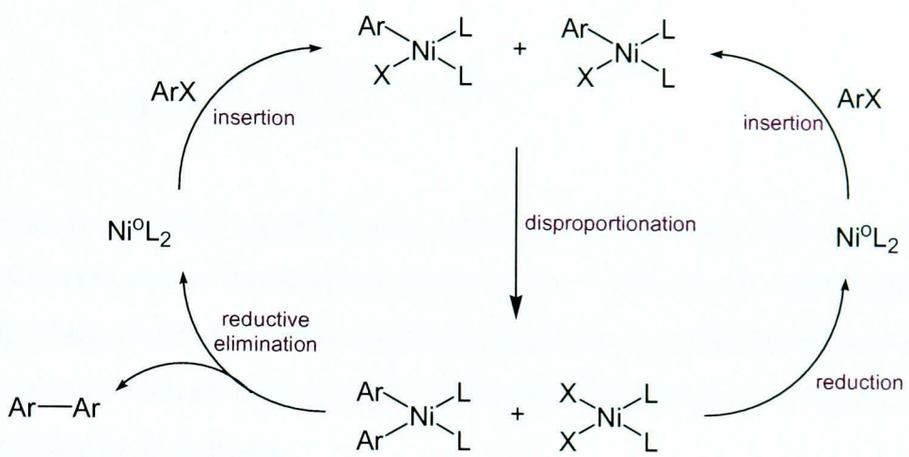
Scheme 56



The exact mechanism of nickel-mediated coupling has not been elucidated and remains controversial.[86] Two mechanisms are postulated:[89]

1) The nickel mediated coupling of aryl halides may proceed in a manner analogous to that described for palladium-catalysed coupling (Scheme 57).

Scheme 57



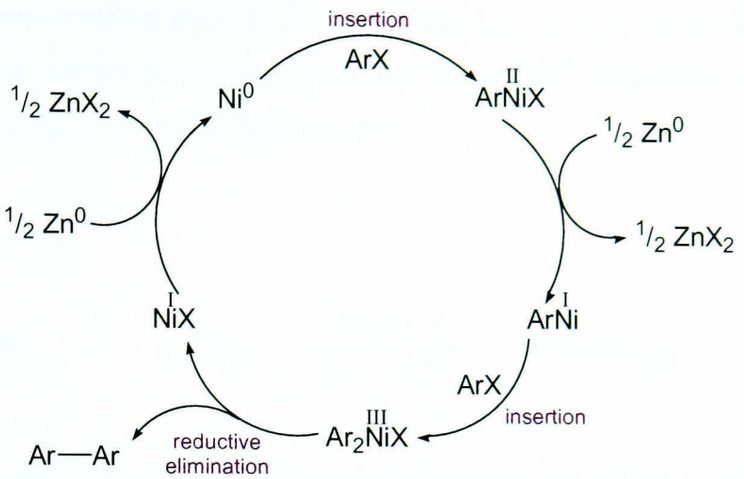
Several mechanistic studies have supported this type of mechanism:

a) Semmelhack *et al.* reported that bis(triphenylphosphine)phenylnickel(II) bromide (a key intermediate) underwent decomposition to give biphenyl at 25 °C in DMF, and that after 12 hours at 50 °C biphenyl was obtained in 99 % yield.[95]

b) Rieke has isolated the postulated nickel(II) intermediates bis(triethylphosphine)(pentafluorophenyl)nickel(II) iodide and bis-(triethylphosphine)bis(pentafluorophenyl)nickel(II) from reaction mixtures by trapping with triethylphosphine (refer to Scheme 54).[93]

2) An extensive mechanistic study by Tsou and Kochi proposed the involvement of nickel(I) and nickel(III) intermediates as key intermediates in the catalytic cycle (Scheme 58).[96]

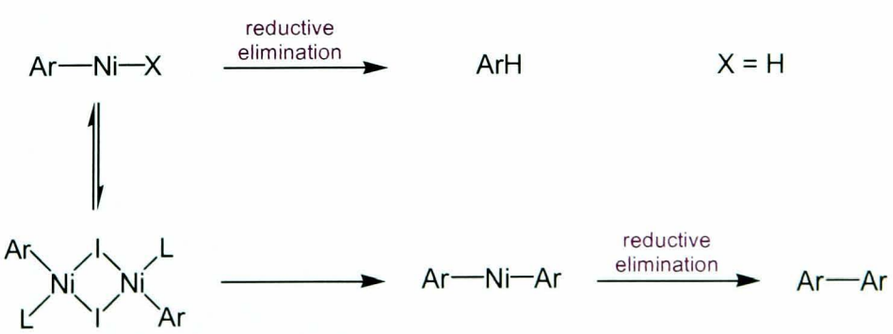
Scheme 58



However, it should be noted that this work involved reactions performed in non-polar solvents such as benzene and cyclohexane. Consequently it has been noted that, as many nickel mediated couplings require the use of moderately or highly polar solvents such as THF, DMF or DME, their conclusions may not be validly extrapolated to all systems.

The beneficial effect of iodide ions on the yield of nickel-mediated homocoupling reactions has been noted by several groups. However, the exact role of iodide is unclear. Zembayashi *et al.* have suggested that iodide acts as a polarisable bridging ion between the nickel and zinc centres in the electron transfer process, thus facilitating reduction of nickel.[97] Lin noted that the addition of tetrabutylammonium iodide to the nickel system (refer to Scheme 55) resulted in a significant increase in homocoupling over dehalogenation. The linking of two arylnickel moieties *via* two iodide ions facilitating coupling (Scheme 59) was proposed to account for this.[94]

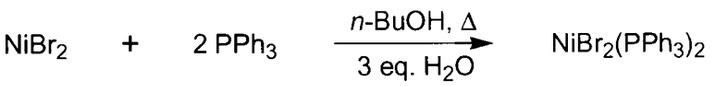
Scheme 59



2.3.3.2 Application to the Model System

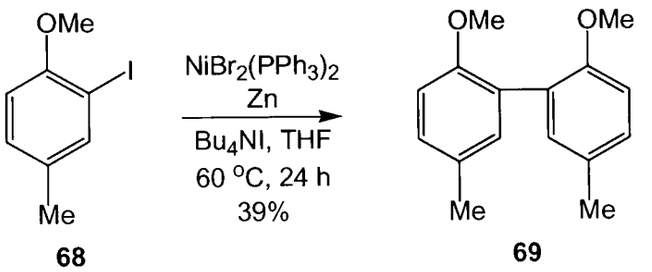
Nickel-mediated homocoupling was investigated, again using a model system initially. The nickel-catalyst precursor bis(triphenylphosphine)nickel(II) dibromide was prepared according to the method of Venanzi (Scheme 60).[98]

Scheme 60



Coupling of the model iodide **68** (see Scheme 50 for preparation) was then performed (Scheme 61). The first attempt provided a 39 % yield of the desired product **69**.

Scheme 61



68 **69**

2.3.3.3 Optimisation of the Reaction Conditions

An optimisation study was undertaken using the model system in order to improve the yield of this reaction. Solvent effects and the effects of other additives were investigated (Table 13).

Table 13 Optimisation of the nickel-mediated coupling of aryl iodide **68**

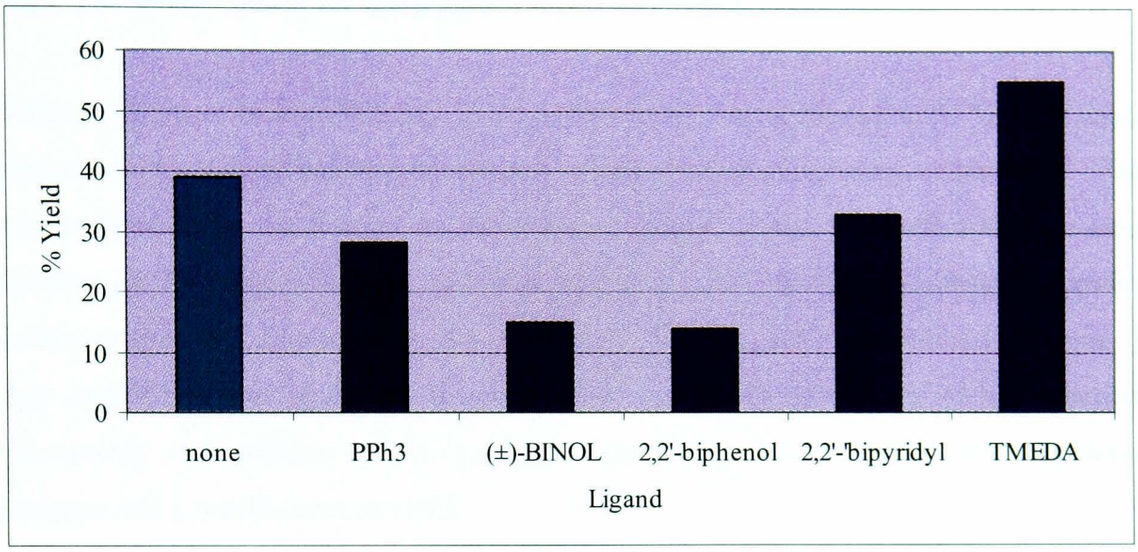
Entry	Conditions[a]	Solvent	% Yield
1		THF	39
2		DMF	16
3		DME	0
4	0.1 eq. TBAI	THF	20
5	– TBAI	THF	16
6	– $NiBr_2(PPh_3)_2$	THF	0
7	+ PPh_3	THF	28
8	+ BINOL	THF	15
9	+ 2,2'-biphenol	THF	14
10	+ 2,2'-bipyridyl	THF	33
11	+ TMEDA	THF	55
12	**68** added at start	THF	26

[a] Unless otherwise stated, all reactions were carried using 0.5 equivalents of $NiBr_2(PPh_3)_2$, 1 equivalent of TBAI and 1.5 equivalents of zinc powder. The aryl iodide **68** was added to the reaction mixture after 30 minutes at room temperature. The reaction mixture was then heated at 60 °C for 24 hours.

These results showed that use of THF as the solvent produced the best yield (Table 13, entries 1-3) and that use of one equivalent of TBAI was essential for a good yield of the desired dimer **69** (Table 13, entries 1, 4 & 5). These results are consistent with those reported by Iyoda.[84]

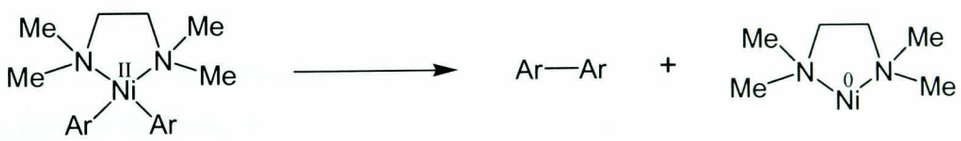
Co-ordinating ligands have been used in a number of palladium and nickel-catalysed coupling reactions to increase yields and selectivities.[99, 100] Since the catalyst and ligand used in a reaction can have a significant effect on the efficiency of the reaction, and since the optimum catalytic system for a given reaction may be highly specific,[101] a range of readily available ligands was investigated. A number of bidentate phosphine ligands are in regular use,[101] but bidentate nitrogen ligands have also been used.[100, 102] The results of these experiments are shown in Table 13, entries 7-11, and in Figure 31.

Figure 31 Effect of additives on yield of model dimer **69**



The results of this study show that the addition of triphenylphosphine reduced rather than improved the yield (again consistent with work by Iyoda). The oxygen-based ligands (±)-BINOL and biphenol resulted in much reduced yields, but the bidentate nitrogen ligands bipyridyl and *N,N,N',N'*-tetramethylethylenediamine (TMEDA) appeared more promising: bipyridyl resulted in a yield similar to that obtained in the absence of a ligand; TMEDA significantly improved the yield. Bidentate ligands such as TMEDA are thought to improve the yield of coupling reactions by changing the electronic character of the metal centre and by forcing the two substrate molecules take up the *cis*-arrangement required for reductive elimination (Figure 32).

Figure 32



2.3.3.4 Application to the Synthesis of the Herqulines

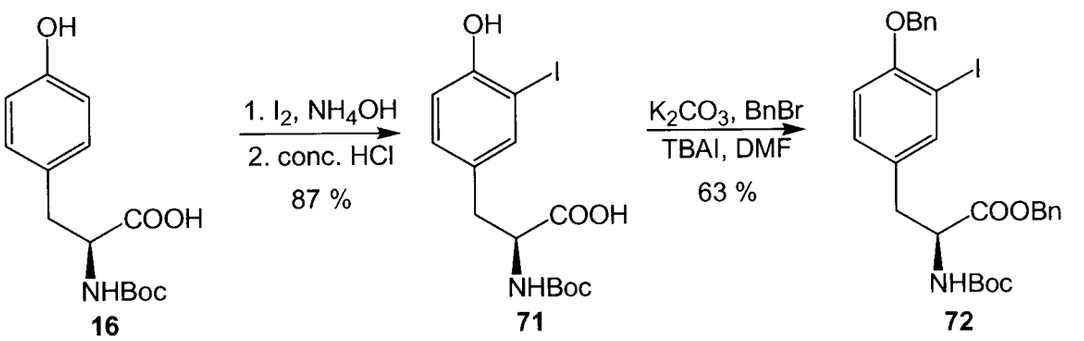
In order to attempt the synthesis of a dityrosine moiety, it was first necessary to generate an appropriately protected halogenated tyrosine unit (Scheme 62). At this stage of the project, reduction of the aromatic rings *via* spirolactones was under consideration (see Chapter 3) and spirolactone formation required the free carboxylic acids and phenols. Therefore, an important consideration when choosing protecting groups was the

capacity to protect and deprotect both the carboxylic acid and phenol groups in a single step. The group chosen for this purpose was the benzyl group.[73]

N-Boc-L-tyrosine **16** (see Scheme 28 for preparation) was iodinated in 87 % yield using ammonium hydroxide and one equivalent of iodine to give the mono-iodide **71**.[103] This was confirmed by the change in the aromatic region of the ^{1}H-NMR spectrum from doublets at 7.04 and 6.70 ppm to a characteristic 1,2,4-trisubstituted aromatic pattern with a doublet at 7.58 ppm, a double doublet at 7.07 ppm and a doublet at 6.78 ppm. It was established that great care must be taken during the quenching of this reaction: excessively fast addition of hydrochloric acid resulted in a significant temperature increase and a much-reduced yield.

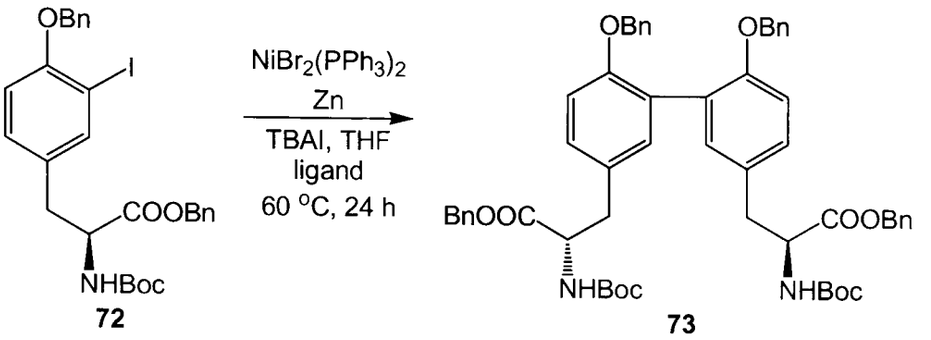
The iodotyrosine **71** was then benzylated,[104] with the desired protected tyrosine **72** being obtained in 63 % yield. The protected iodotyrosine was therefore synthesised from L-tyrosine in three steps and 55 % overall yield.

Scheme 62



The original nickel-mediated coupling reaction conditions were applied to the protected iodotyrosine **72** (Scheme 63) but resulted in a poor yield of only 6 %.

Scheme 63



The best two results from the optimisation study (addition of TMEDA or bipyridyl) were then applied to the protected iodotyrosine (Table 14).

Table 14 Effect of ligands on yield of tyrosine dimer **73**

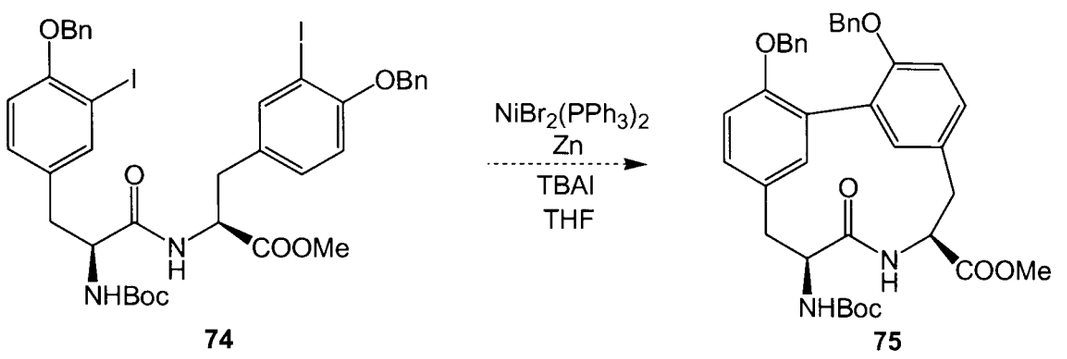
Added ligand	% Yield
none	6
bipyridyl	12
TMEDA	12

Although the results of the optimisation study carried out on the model compound appeared promising, when applied to the tyrosine system the yields were disappointing. This may be because the substrate, with its benzyl protecting groups, is much bulkier than the model system; alternatively, the slightly acidic NH group may have interfered with the nickel system in some way.

2.3.3.5 Intramolecular Coupling

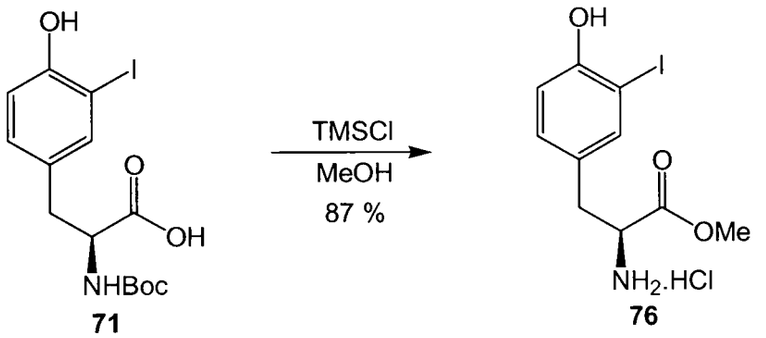
It was again hoped that an intramolecular version of the reaction (Scheme 64) might be more successful, due to the more favourable entropy of activation.

Scheme 64



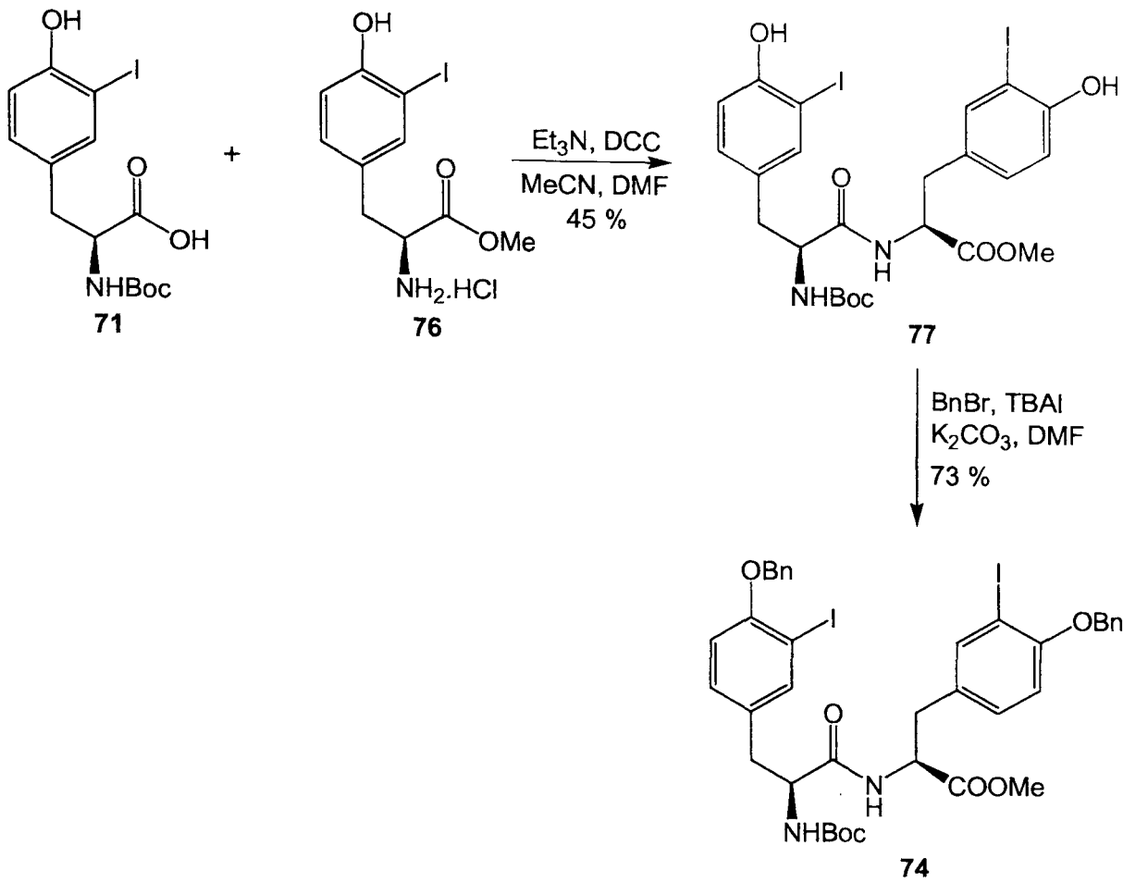
In order to prepare the required diiododipeptide **74**, the iodotyrosine methyl ester **76** was needed. Iodination of L-tyrosine itself resulted in a messy crude product and poor mass recovery, presumably due to an isolation problem. The required ester **76** was therefore synthesised from *N*-Boc-3-iodotyrosine **71** in 87 % yield using TMS chloride (Scheme 65).[105]

Scheme 65



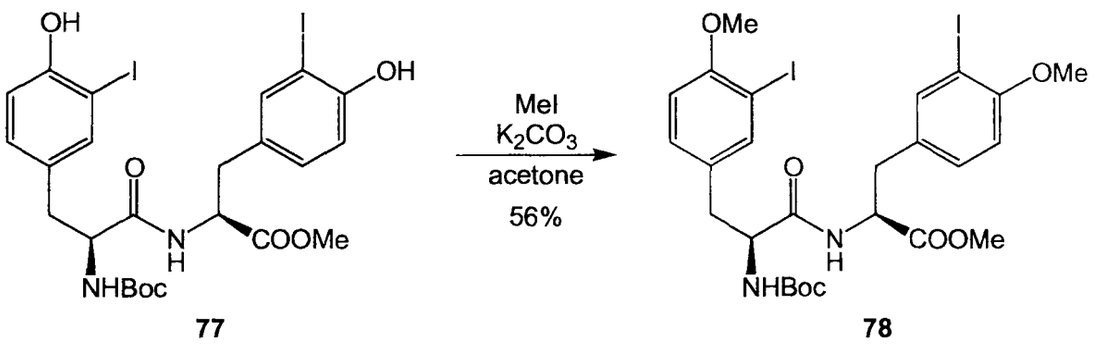
The protected dipeptide **77** was then formed in 45 % yield using standard peptide coupling reagents[35] and benzyl protected[104] in 73 % yield (Scheme 66).

Scheme 66



The corresponding methyl-protected dipeptide **78** was also synthesised in order to try to reduce the steric bulk of the *ortho*-substituents (Scheme 67).[106]

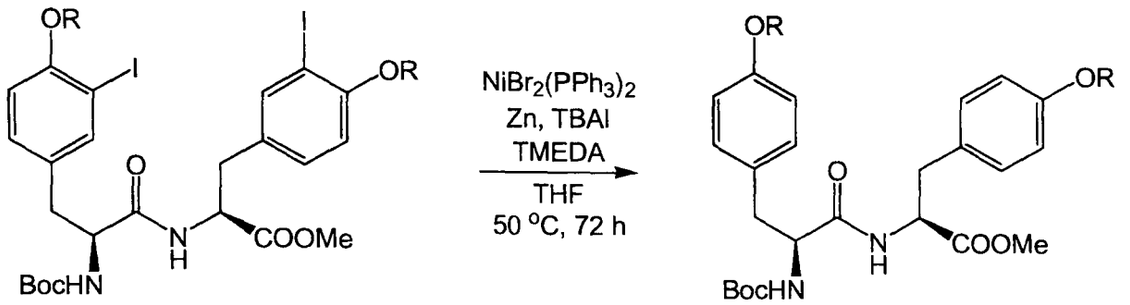
Scheme 67



However, when the nickel-based coupling conditions were applied to both the benzyl- and methyl-protected dipeptides, no biphenyl products were observed in either case. The major products were the dehalogenated dipeptides **79** or **52** (Scheme 68), confirmed by the presence of four doublets in the aromatic region of the ^{1}H-NMR spectra. This

indicated that dehalogenation was occurring more rapidly than any coupling processes, which may be due to the conformation of the dipeptides.

Scheme 68



74 R = Bn
78 R = Me

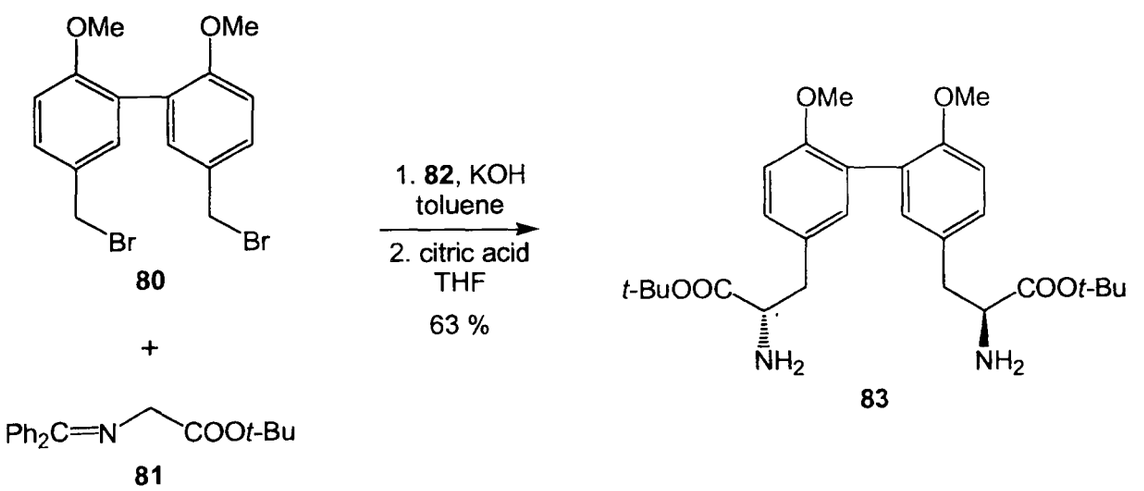
79 R = Bn
52 R = Me

2.4 Asymmetric Phase-Transfer Method

2.4.1 Introduction

Lygo has reported the synthesis of a protected dityrosine *via* a double asymmetric alkylation reaction (Scheme 69); the reaction was catalysed by a cinchonidine-derived chiral quaternary ammonium salt **82** acting as a phase-transfer reagent.[107, 108] This route is significantly different from the others discussed in that it does not involve the coupling of tyrosines. Consequently, the first two chiral centres formed in the synthesis of the herqulines would originate not from tyrosine, but from this asymmetric alkylation reaction.

Scheme 69



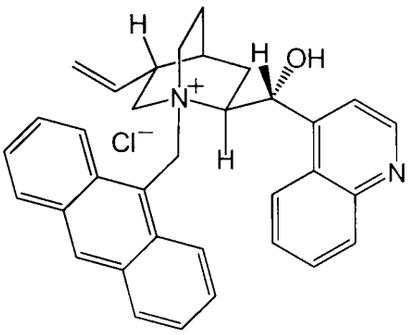
80

+

81

83

Figure 33 *N*-9-Anthracenylmethylcinchonidinium chloride

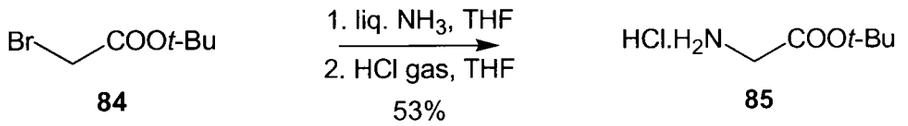


82

2.4.2 Application to the Synthesis of the Herqulines

In accordance with Lygo's work, *t*-butyl *N*-(diphenylmethylene)glycinate **81** was synthesised from *t*-butyl bromoacetate. Glycine *t*-butyl ester hydrochloride **85** was first generated in 53 % yield by the action of liquid ammonia followed by hydrogen chloride gas on *t*-butyl bromoacetate **84** (Scheme 70).[109] Formation of the intermediate free amine was confirmed by the change in chemical shift of the methylene signal in the ^{1}H-NMR spectrum from 3.76 ppm to 3.33 ppm. Formation of the hydrochloride salt also resulted in a subtle change in the chemical shift of this signal, and was accompanied by a change in physical state from an oil to a crystalline solid.

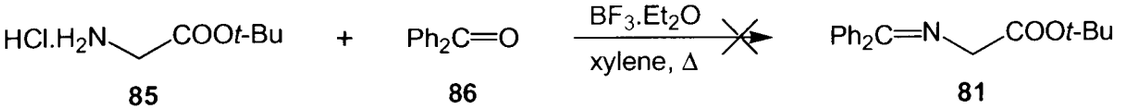
Scheme 70



84 **85**

Subsequent treatment of the amino ester **85** with benzophenone **86** and boron trifluoride-diethyl ether complex in refluxing xylene using a Dean-Stark apparatus for azeotropic removal of water[110] resulted in no reaction (Scheme 71).

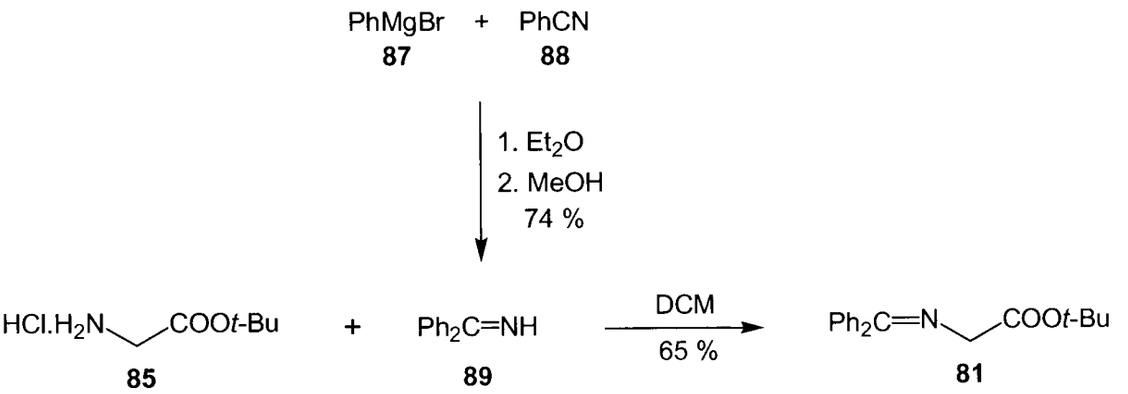
Scheme 71



85 **86** **81**

Benzophenone imine **89** was therefore synthesised from phenylmagnesium bromide*[111] **87** and benzonitrile **88** in 74 % yield according to the method of Pickard and Tolbert (Scheme 72).[112] Formation of the imine was confirmed by observation of a broad singlet at 9.70 ppm in the ^{1}H-NMR spectrum corresponding the imine proton, and of a peak at 1659 cm^{-1} in the IR spectrum corresponding to the C=N stretch. Stirring the protected amino acid **85** and benzophenone imine **89** in DCM at room temperature then produced the desired imine **81** in 65 % yield. Observation of the molecular ion by mass spectrometry and of peaks at 1735 and 1623 cm^{-1} (corresponding to the ester and imine respectively) in the IR spectrum confirmed the synthesis of the imine **81**.

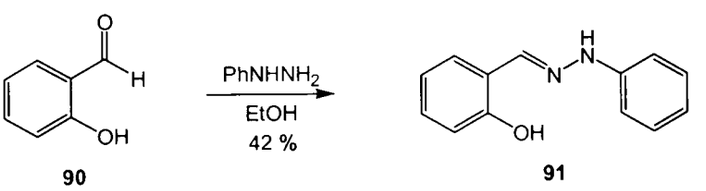
Scheme 72



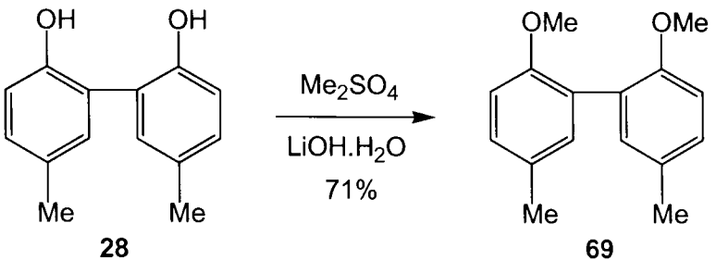
However, problems with this route began with the synthesis of the dibromide **80**.

The methyl anisole dimer **69** was synthesised in good yield from the previously generated *p*-cresol dimer **28** (Scheme 73; see section 2.2.5.2 for preparation).

* Phenylmagnesium bromide was titrated against salicylaldehyde phenylhydrazone **91** before use for accurate determination of molarity. Salicylaldehyde phenylhydrazone **91** was generated from salicylaldehyde **90** and phenylhydrazine.

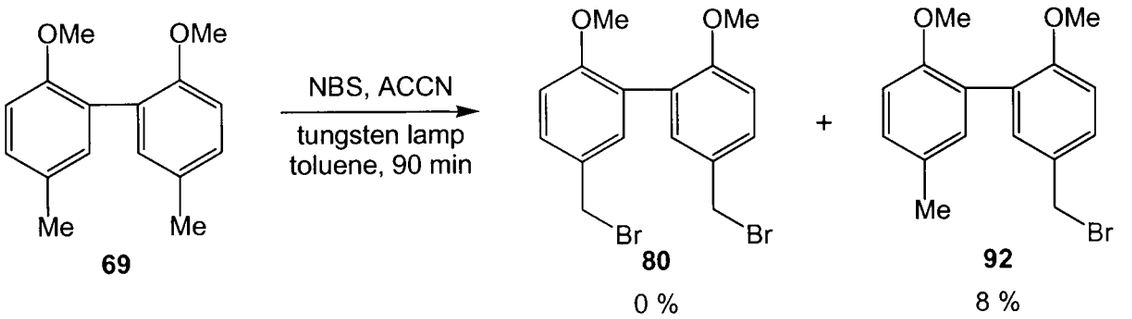


Scheme 73



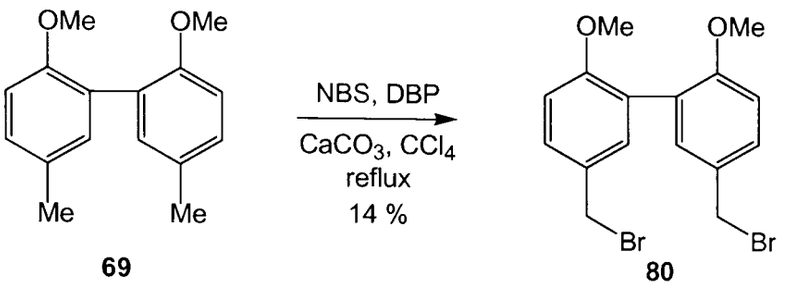
Lygo had reported the synthesis of the dibromide **80** in 93 % yield from the methyl anisole dimer **69**. The radical bromination reaction reportedly required the use of both AIBN and a 100 W sunlight lamp. However, due to availability, 1,1'-azobis(cyclohexanecarbonitrile) (ACCN) was used instead of AIBN and a standard light bulb instead of a sunlight lamp (Scheme 74). Analysis of the crude reaction mixture by tlc indicated a mixture of seven products. Flash chromatography of the crude product resulted in 21 % recovery of the starting material **69** and 8 % yield of the monobromide **92**, but none of the desired dibromide **80** was isolated.

Scheme 74



An alternative procedure involved the use of dibenzoyl peroxide as the radical initiator, and this was applied to the dimer **69** (Scheme 75).[113] Again the crude product was a complex mixture, but flash chromatography led to isolation of the desired dibromide **80** in 14 % yield.

Scheme 75



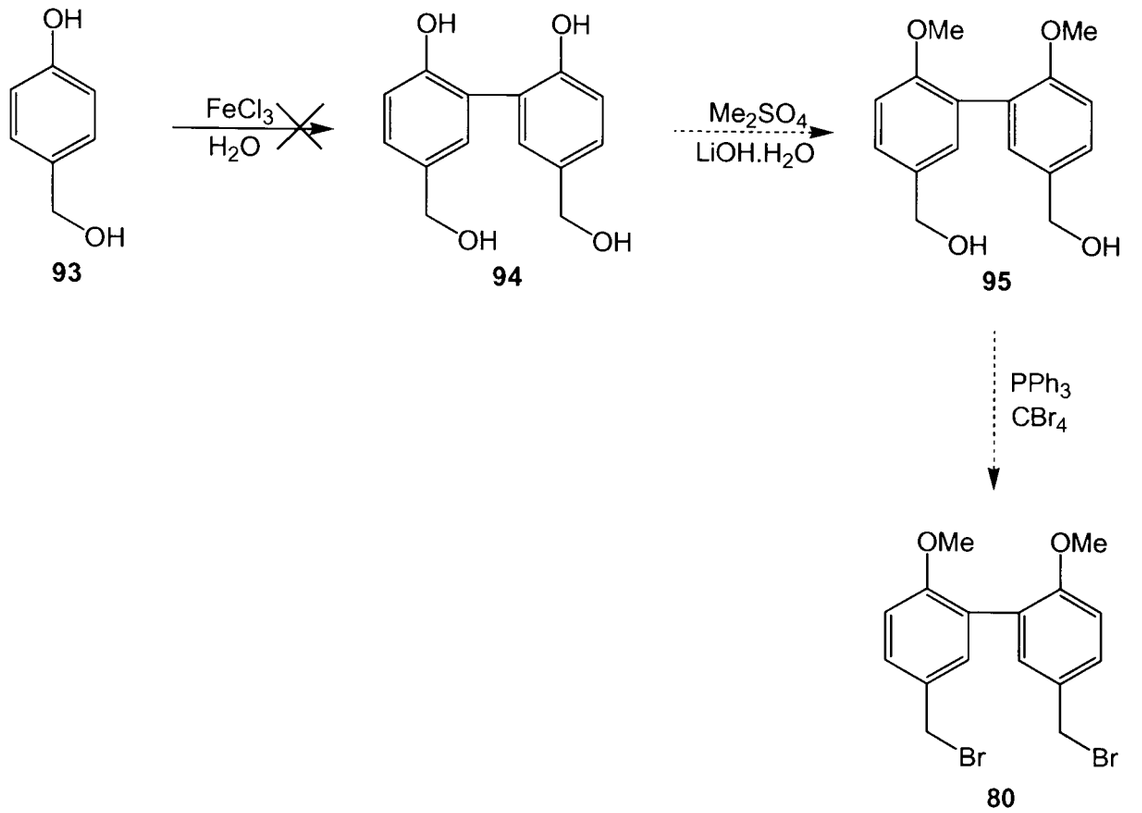
69 **80**

The yield could not be improved upon because of the somewhat erratic nature of the reaction. The reaction time was found to be crucial, with initial formation of the monobromide being followed by dibromide formation. However, too long a reaction time seemed to result in decomposition and a mixture of unidentifiable products. Additionally, the time required appeared to vary; consequently, regular monitoring by tlc was necessary throughout the course of the reaction.

An alternative was examined: oxidative coupling with ferric chloride was used previously to couple *p*-cresol **20** (Section 2.2.5.2).[49] The possibility of utilising this technique in the synthesis of dibromide **80** was considered. If commercially available 4-hydroxybenzyl alcohol **93** could be oxidatively coupled in the same way, the dimer could be methylated at the phenolic OH group and subsequently treated with brominating conditions such as triphenylphosphine and carbon tetrabromide (Scheme 76).

Unfortunately, 4-hydroxybenzyl alcohol **93** did not undergo dimerisation in the presence of ferric chloride and only the starting material was recovered. This may have been due to complex formation at the aliphatic hydroxy group instead of at the phenoxy group.

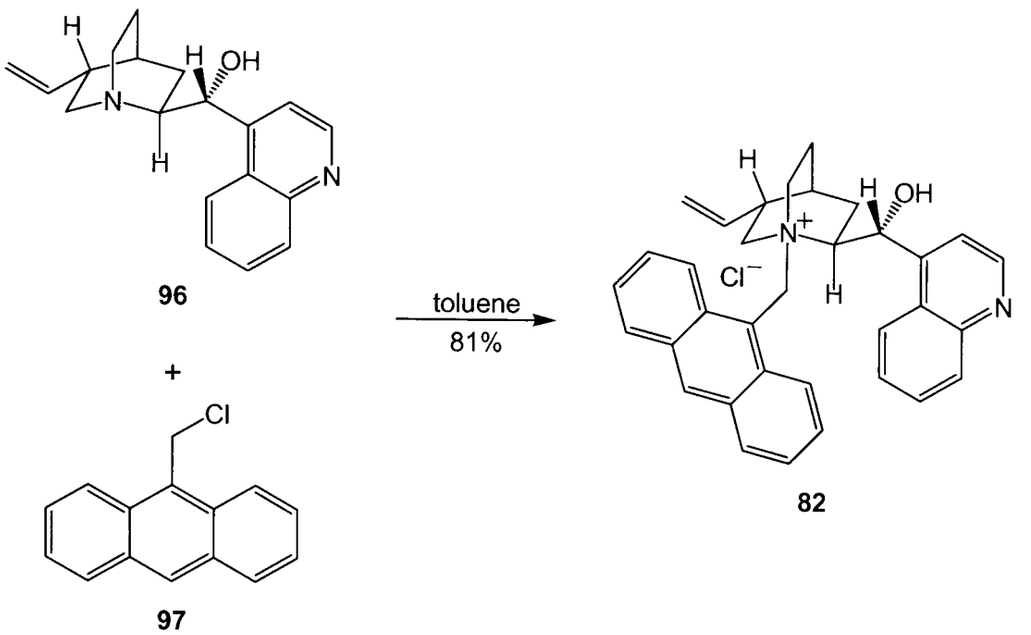
Scheme 76



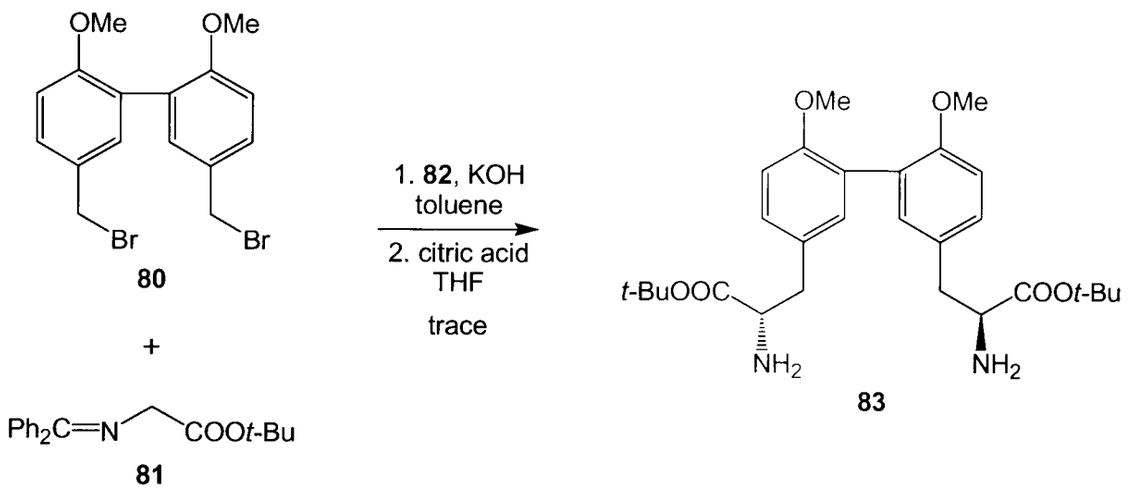
The phase-transfer catalyst (*N*-9-anthracenylmethyl)-(−)-cinchonidinium chloride) **82** was synthesised in 81 % yield from (−)-cinchonidine **96** and 9-(chloromethyl)anthracene **97** as described by Corey (Scheme 77).[114] Formation of the catalyst was confirmed by observation of the molecular ion by mass spectroscopy and by comparison of the ^{1}H-NMR spectrum with data reported by Corey.

The chiral catalyst **82** was then used to facilitate the alkylation reaction (Scheme 78). The ^{1}H-NMR spectrum of the crude product contained double doublets at 3.59 ppm (corresponding to the α-protons) and at 3.02 and 2.79 ppm (corresponding to the methylene protons) confirming the formation of the protected dityrosine **83**. No signals below 8 ppm were observed, indicating the absence of an imine system. However, the reaction was carried out on a very small scale, and the dityrosine **83** could not be isolated.

Scheme 77



Scheme 78



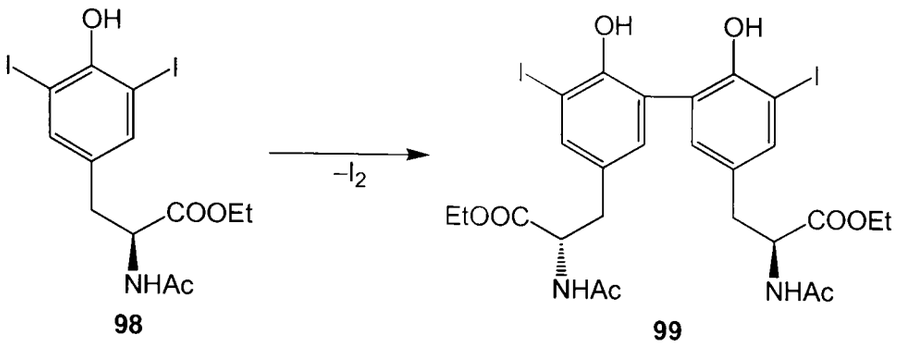
It was decided that the number of steps involved in this route, combined with the problematic radical reaction, made the asymmetric alkylation reaction unsuitable for the synthesis of the herqulines.

2.5 Bowman Coupling

2.5.1 Introduction

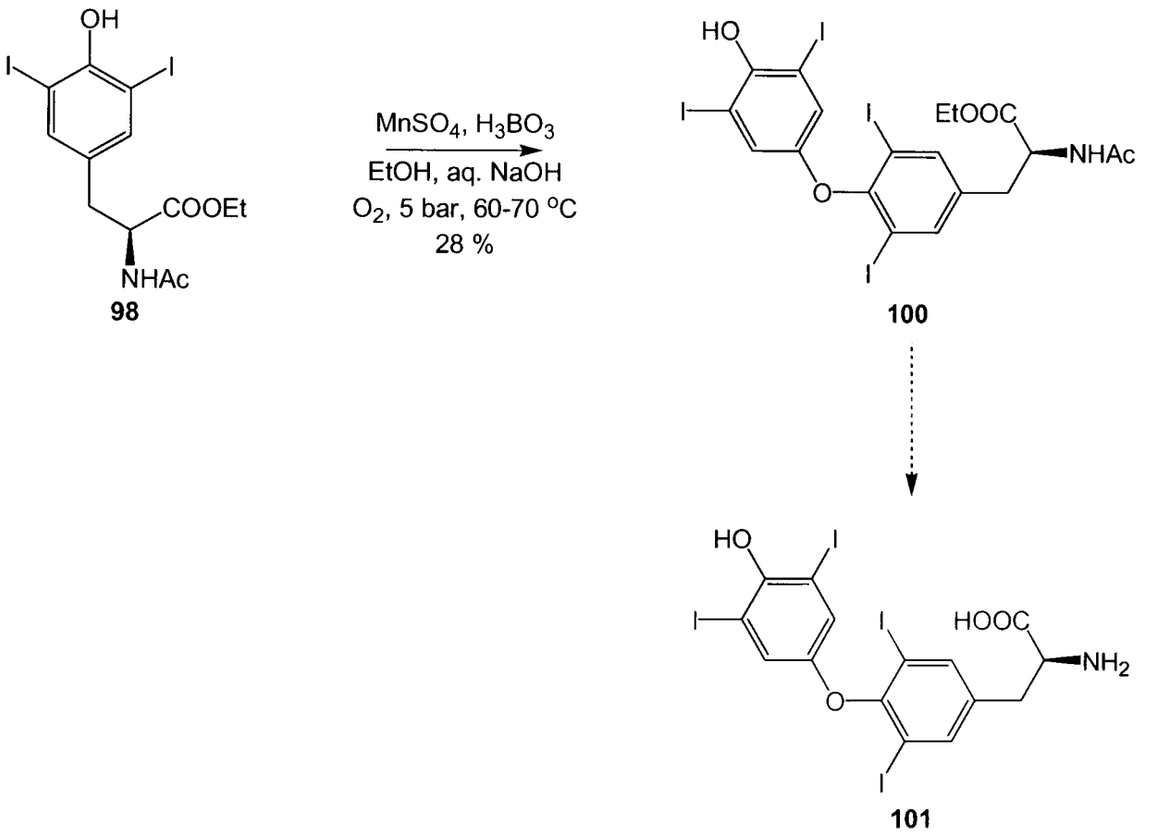
The unusual homocoupling of a protected diiodotyrosine resulting in a diiododityrosine has been reported by Bowman (Scheme 79).[115]

Scheme 79



The reaction was discovered during attempts to synthesise a derivative of thyroxine* **101** (Scheme 80).[116]

Scheme 80

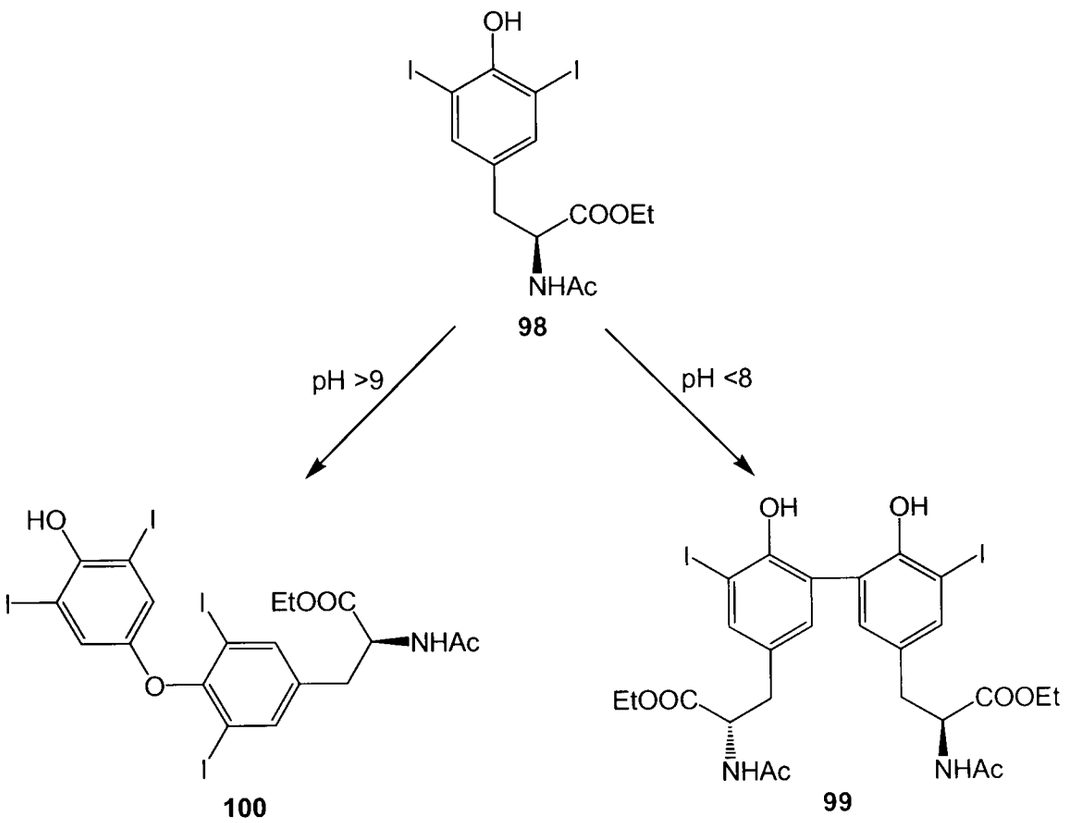


However, the formation of a dityrosine by-product **99** was observed. Dityrosine formation was found to occur concurrently with a drop in the pH of the reaction mixture

* The major hormone secreted by the thyroid gland. Thyroxine binds to DNA to increase the number and activity of mitochondria, thereby increasing the metabolic rate; it is essential for growth and development in mammals.

(Figure 34) but it was not clear whether the drop in pH was caused by the formation of the dimer or *vice versa.*

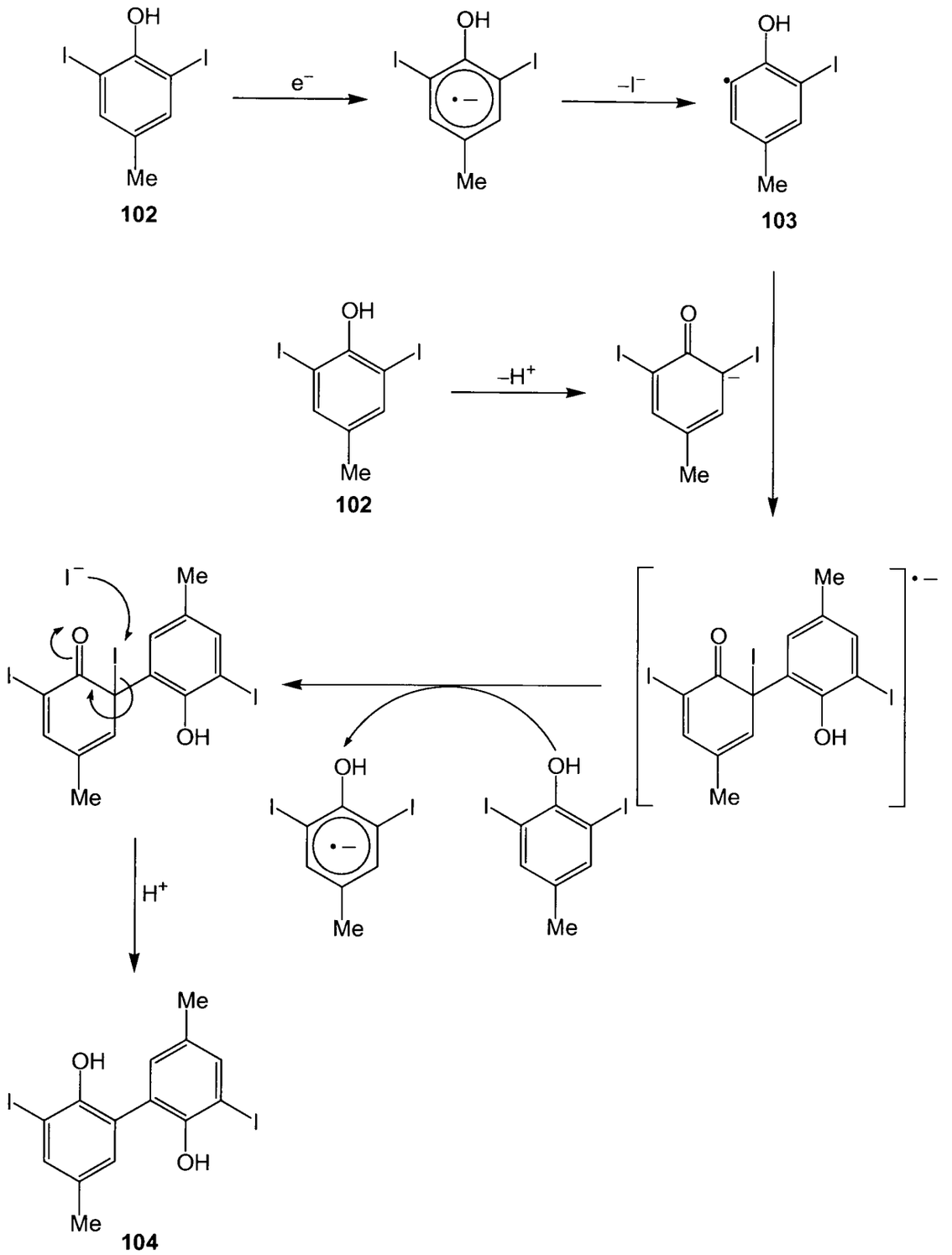
Figure 34 Effect of pH on the reaction of *N*-acetyl-3,5-diiodo-L-tyrosine ethyl ester **98**



Reaction conditions: $MnSO_4$, H_3BO_3, EtOH, aq. NaOH, O_2, 5 bar, 60-70 °C.

Since the *in vivo* formation of thyroxines is widely believed to occur *via* phenoxy radicals,[117] the group first considered a radical mechanism in order to account for the formation of the dityrosine by-product **99** (Scheme 81).

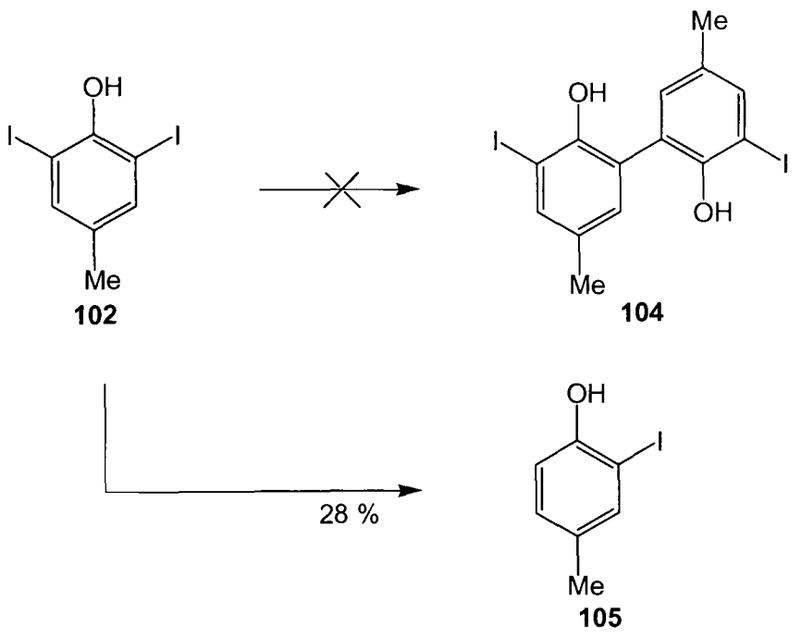
Scheme 81 Possible radical mechanism for the formation of diiododityrosines proposed by Bell and Bowman[116]



However, Bell and Bowman found that the treatment of diiodo-*p*-cresol **102** with base in a range of solvents (exposed to combinations of heat, light and manganese salts) gave none of the expected product **104**; only the starting material was recovered in each case.

Exposure of diiodo-*p*-cresol **102** to hexabutylditin – a reagent known to favour radical formation[118] – also failed to generate the desired dimer **104** (Scheme 82). These conditions did however cause dehalogenation and iodo-*p*-cresol **105** was isolated in 28 % yield. This indicated that the radical intermediate **103** had been formed, but did not subsequently undergo the coupling reaction described above (Scheme 81). It was therefore concluded that the dimerisation of diiodophenols did not occur *via* a radical mechanism.

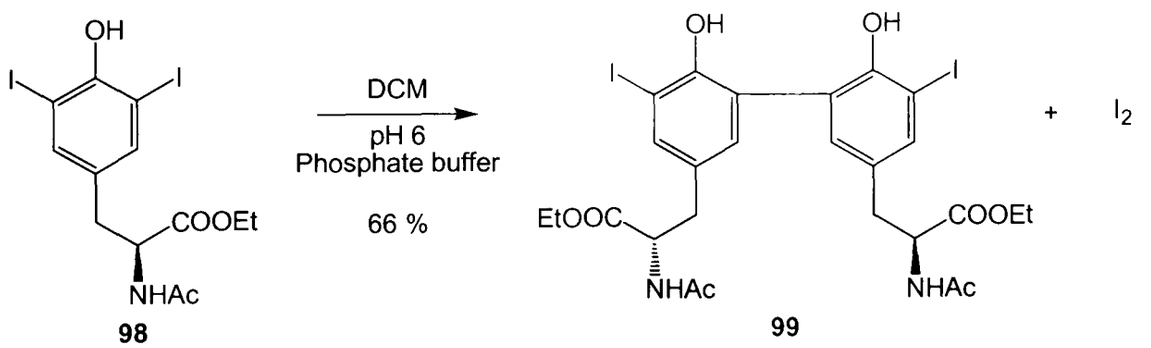
Scheme 82



Reaction conditions: Sn_2Bu_6, toluene, Δ, *hν*.

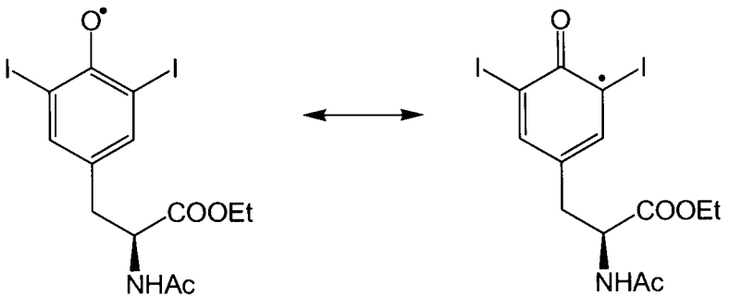
Further development of the coupling reaction led to the identification of milder reaction conditions, using a biphasic mixture of DCM and aqueous borate or phosphate buffer (Scheme 83). Again, control of pH appeared crucial and the optimum pH was found to be between 5.0 and 7.2.

Scheme 83



The success of this reaction in an aqueous medium, together with the fact that no formation of isodityrosine was observed, implied a polar reaction mechanism. As discussed previously (Section 1.3.2) when a phenoxyl radical is formed, it may react *via* carbon or oxygen (Figure 35) resulting in the formation of a dityrosine or an isodityrosine respectively. Consequently, isodityrosine is a common by-product arising from syntheses of dityrosine. Since none of the corresponding isodityrosine was observed following the DCM/phosphate buffer reaction, the involvement of a phenoxyl radical was considered unlikely.

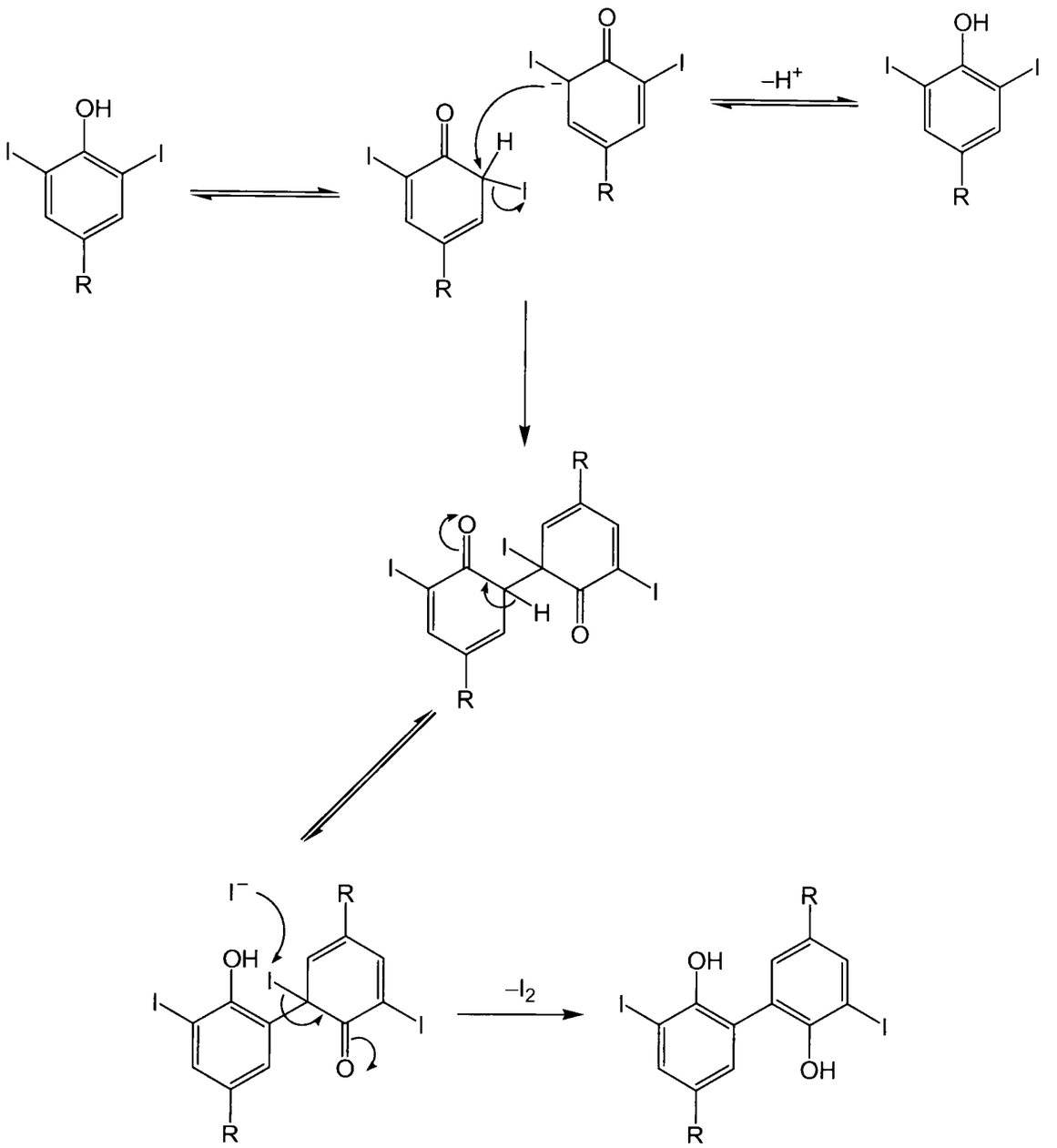
Figure 35 Canonical structures of the diiodophenoxy radical



Consequently, Bowman proposed an S_N2 mechanism accounting for the formation of diiodotyrosines (Scheme 84).

This S_N2 mechanism accounts for the pH-dependency of the reaction since the mechanism requires the participation of both the phenol and the phenoxide. This would only be possible if the pH of the reaction mixture was maintained at a value close to the pK_a of the substrate. The pK_a of a diiodophenol is approximately 6.5,[116] therefore maintenance of the pH of the reaction mixture at around 6 would allow an equilibrium between the phenol and the phenoxide. This mechanism also accounts for the formation of iodine observed during the reaction.

Scheme 84



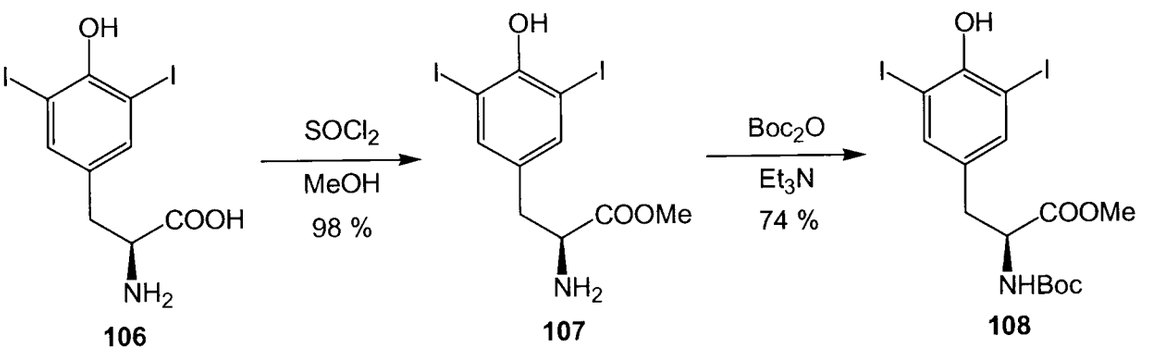
Further work by Bell and Bowman established that there was no reduction in yield when the reaction was performed in the presence of a radical inhibitor (TEMPO). A small drop in yield was observed when the reaction was performed in the absence of light; a 50 % yield of the expected product was nevertheless obtained, indicating that light was not a critical factor in the reaction mechanism. These results served to reinforce the proposed S_N2 mechanism.

2.5.2 Application to the Synthesis of the Herqulines

In order to make use of the Bowman coupling reaction as part of the synthesis of the herqulines, an appropriate diiodotyrosine was synthesised. It was decided that the carboxylate group would be protected as a methyl ester as this straightforward reaction had been used previously; this would also serve to simplify the ^{1}H-NMR spectra of subsequent compounds. The Boc group was chosen to protect the amino moiety. The procedure was again a straightforward one, but more importantly provided a robust, yet easily cleaved, protecting group. Another key consideration was the fact that carbamates are stable under metal-ammonia reduction, which was to be utilised later in the synthesis.

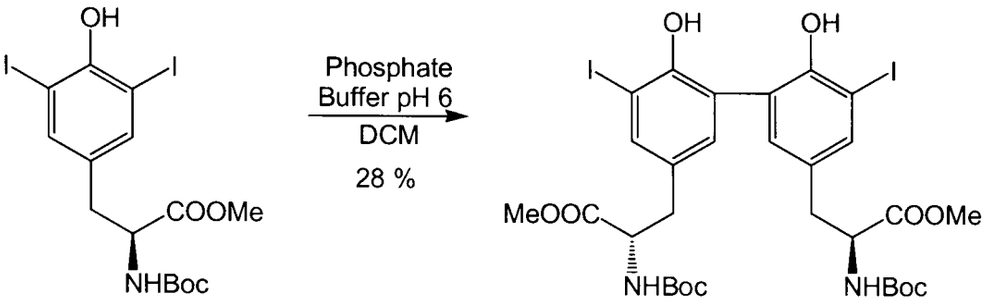
Commercially available 3,5-diiodo-L-tyrosine dihydrate **106** was protected[119] by methyl esterification using thionyl chloride and methanol[50] in 98 % yield and Boc-protection using Boc anhydride and triethylamine[51] in 74 % yield (Scheme 85). Following Bowman's procedure, the protected tyrosine **108** was then stirred in phosphate buffer and DCM for four days; the corresponding dityrosine **109** was obtained in 28 % yield. The formation of the dimer was confirmed by the presence of two separate signals in the aromatic region of the ^{1}H-NMR spectrum (7.50 ppm and 7.15 ppm) and by mass spectrometry.

Scheme 85



106 **107** **108**

Scheme 86



108 **109**

2.5.3 Optimisation of the Reaction Conditions

An optimisation study was carried out in order to improve the yield of the coupling reaction, and experiments were undertaken in order to determine the optimum concentration of the reaction mixture, the optimum reaction time and the effect of other reagents (Table 15). Unless otherwise stated, all experiments were carried out at ambient temperature and without an inert atmosphere. In order to clearly illustrate the key conclusions reached as a result of this work, some of the data are illustrated graphically below.*

* All graphs show series of reactions in which a single factor was varied.

Table 15 Optimisation of Bowman Coupling

Entry	Time (days)	Conc. (mg/ml)[a]	Other Conditions[b]	% Yield of 109[c]
1	4	67		28
2	0.5	20		2
3	1	30		55
4	2	30		45
5	3	30		35
6	4	30		21
7	5	30		15
8	3	100		4
9	3	50		12
10	3	20		30
11	3	10		5
12	3	40		28
13	1	30	acetate buffer pH 4.6	0
14	3	20	sunlight	25
15	3	20	dark	35
16	3	20	dark, 35 °C	37
17	3	20	0.3 eq. I_2	28
18	3	40	excess $Na_2S_2O_3$	14
19	3	20	$Na_2S_2O_3$ dropwise	53
20	1	30	Ethyl vinyl ether dropwise	15
21	1	30	1 eq. NaI	27
22	1	30	1 eq. Bu_4NI	17
23	1	30	bipyridyl	14
24	1	30	bipyridyl & $SnCl_2$ dropwise	0
25	1	30	$AgNO_3$ dropwise	28
26	1	30	Ag_2SO_4 dropwise	30
27	1	30	2 eq. I_2	30
28	1	30	cat. I_2 & Ag_2SO_4 dropwise	34

[a] Concentration of starting material in DCM. [b] Reactions were carried out using equal volumes of DCM & pH 6 phosphate buffer, vigorous magnetic stirring and dropwise addition of 0.05 M NaOH as necessary to maintain pH 6. [c] Yield determined by quantitative reverse-phase hplc.

Using the method described by Bowman, a yield of 28 % of the protected dityrosine **109** had been achieved (Table 15; entry 1). In order to improve this, the effect of both reaction time and concentration on yield were investigated. The results are illustrated in Figure 36 and Figure 37 respectively.

Figure 36 Effect of time on yield of dityrosine **109**

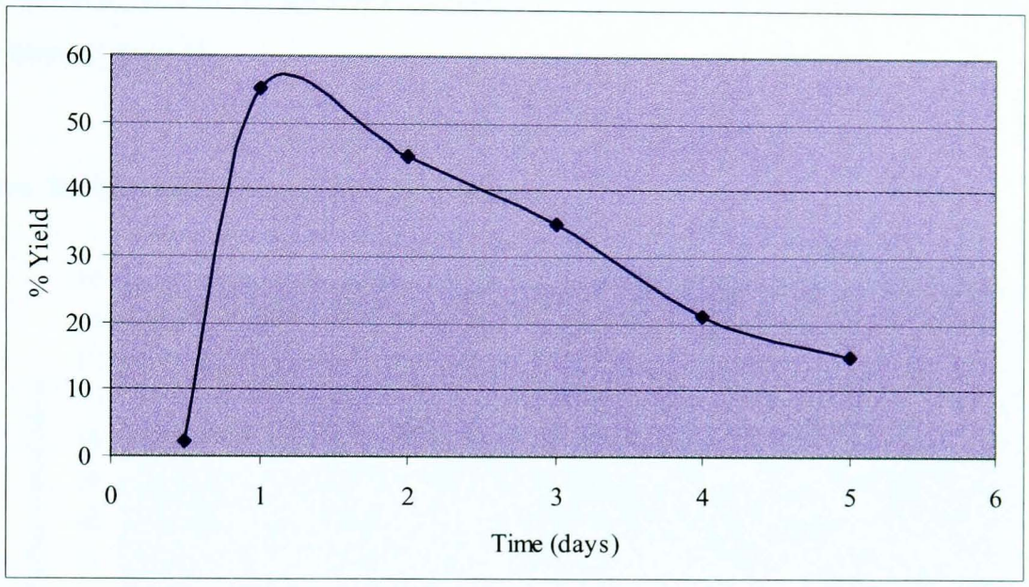
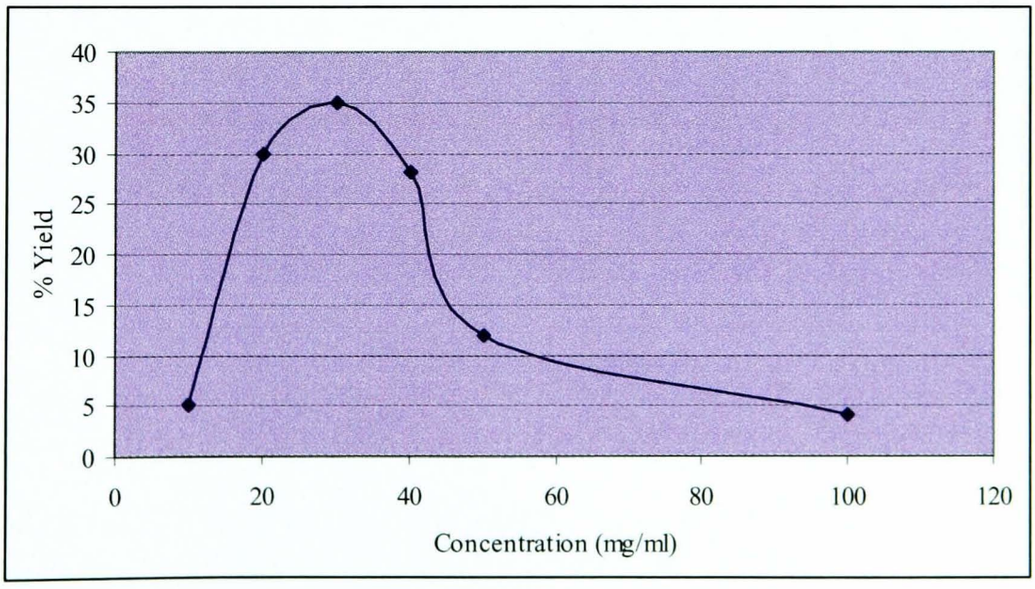


Figure 36 shows that, following an initial period during which the reaction became established, the yield decreased with time. An optimum reaction time of 24 hours was identified from this set of experiments.

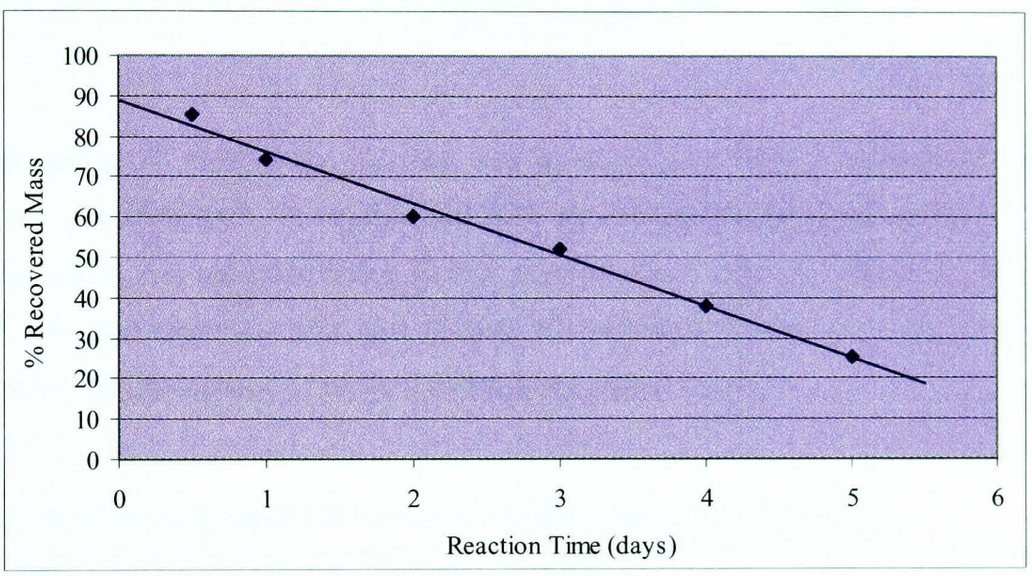
Figure 37 Effect of concentration on yield of dityrosine **109**



Investigation of the effect of the concentration of the reaction mixture on yield of dityrosine **109** also provided a clear result, and indicated an optimum concentration of 30 mg/ml.

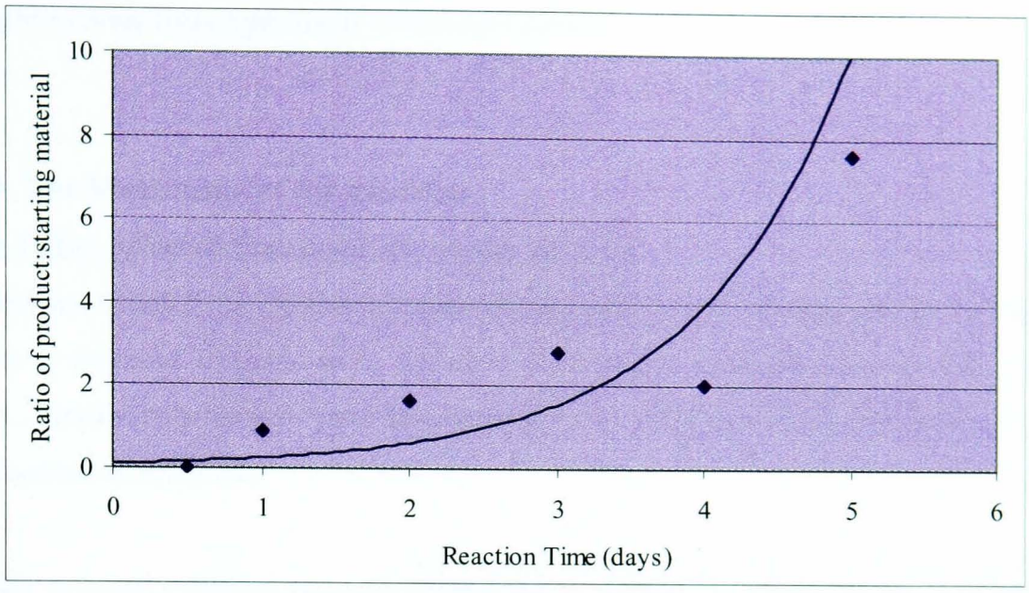
Also examined were the crude yield (percentage recovered mass) of each reaction and the ratio of product to starting material. The effect of time on crude yield followed the same general pattern as observed for the yield of the dimer: the crude yield decreased with time (Figure 38).

Figure 38 Effect of time on recovered mass



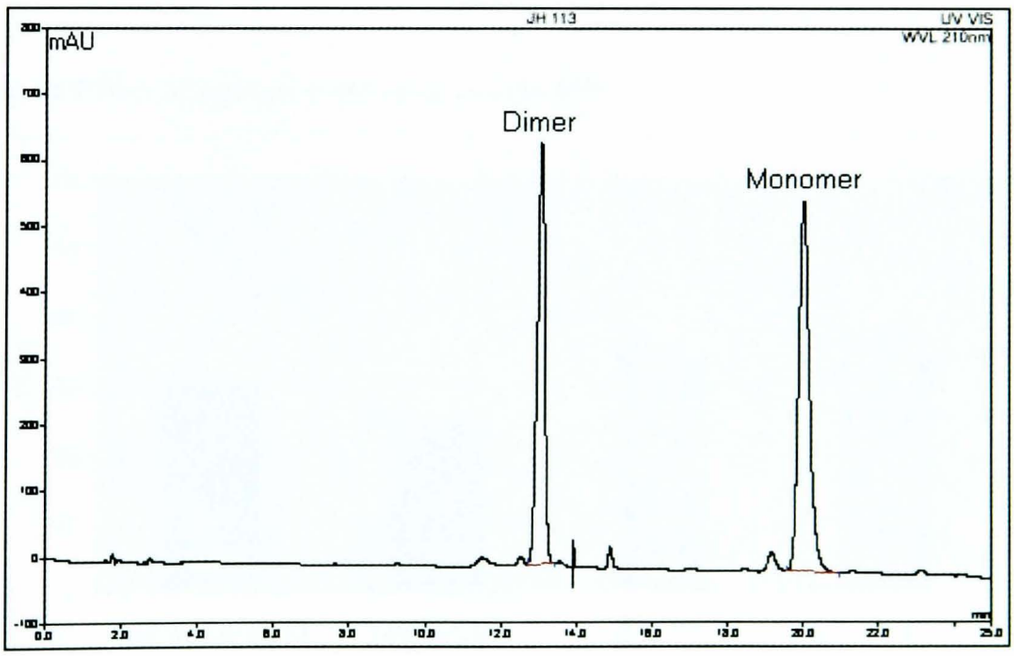
The ratio of product to starting material (determined by hplc analysis of the crude product) was also affected by the length of time of the reaction (Figure 39).

Figure 39 Effect of time on ratio of product **109**:starting material **108**



As the length of time of the reaction was increased, the ratio of product to starting material also increased. In conjunction with the decrease in both recovered mass and product yield, this indicated that a further reaction was occurring. However, both ^{1}H-NMR data and analytical hplc data (Figure 40) indicated that the crude products were almost entirely composed of product and starting material only.

Figure 40 Hplc analysis of the Bowman coupling reaction (Scheme 86)



One explanation for these observations was that a further unknown reaction was occurring, resulting in a water-soluble product. Subsequent acidification and re-

extraction of an aqueous fraction of the reaction mixture gave traces of compounds thought to arise from hydrolysis of the ester groups.

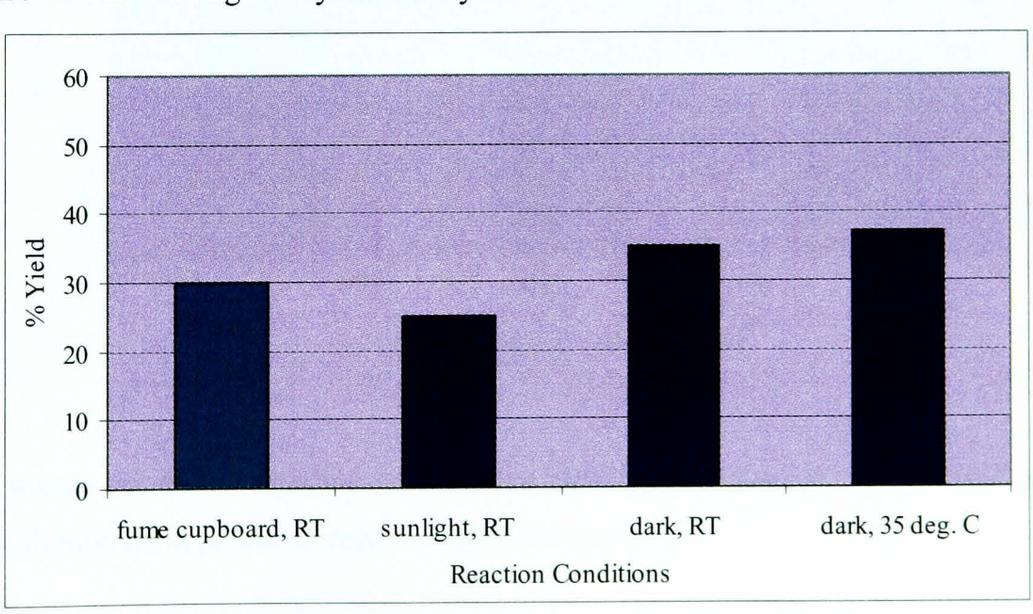
2.5.4 The Mechanism of the Reaction

2.5.4.1 The Effect of Heat and Light on the Reaction

Having established the optimum concentration and length of time of the reaction, a range of different reagents were added to the reaction mixture to try to improve the yield. Several experiments were first carried out in order to confirm the polar nature of the reaction mechanism.

As this unusual coupling reaction progressed, a deep pink-purple colour developed in the organic layer due to the accumulation of iodine. Additionally, it was noted that samples of the diiododityrosine starting material **108** in chlorinated solvents turned pink after several days on the bench. Samples from which light was excluded remained colourless. Consequently, reactions were carried out in which the biphasic mixture was exposed to sunlight (placed on a sunny windowsill) and in which light was excluded. However, the presence or absence of sunlight appeared to have little effect on the yield of the product (Figure 41). The temperature at which the reaction was performed also appeared to have little effect on the yield of the product.

Figure 41 Effect of light on yield of dityrosine **109**



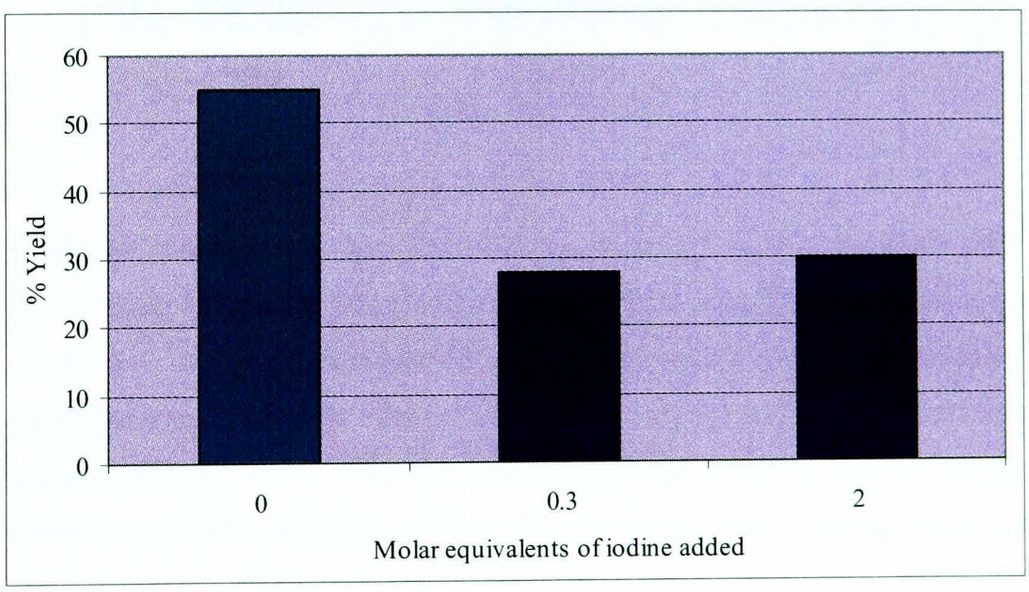
These results confirmed those reported by Bowman. Heat and UV light are often used to increase the rate of homolytic cleavage and therefore the rate of initiation of radical processes. Since the reaction conditions tested appeared to have little effect on the yield of the reaction, the involvement of a radical mechanism appeared unlikely.

2.5.4.2 The Role of Iodine

Attention was then turned to the role of iodine in the reaction. During the course of the reaction, the pink-purple colour deepened with time; consideration was therefore given to the fact that accumulation of iodine may have limited the yield of the reaction. Reduction of the concentration of iodine in the reaction mixture may therefore have allowed any equilibrium processes to be pushed further towards completion.*

Initially, the effect of the addition of extra iodine to the reaction mixture was examined in order to confirm that the accumulation of iodine had an effect on the reaction (Figure 42).

Figure 42 The effect of the addition of extra iodine on yield of dityrosine **109**

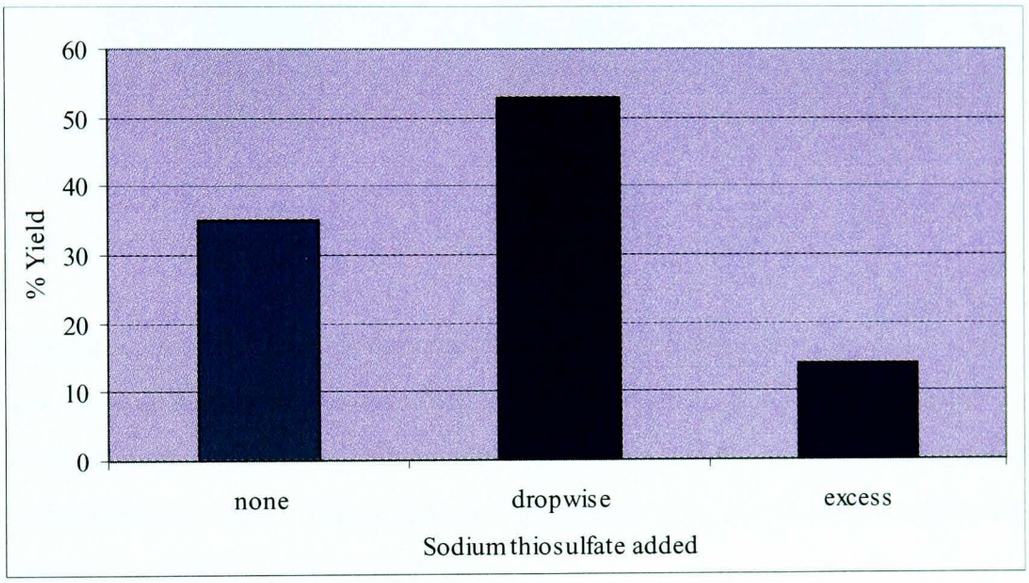


The results of these experiments indicated that the addition of extra iodine to the reaction mixture appeared to retard or limit the reaction.

* Le Chatelier's principle states: "any change in one of the variables that determines the state of a system in equilibrium causes a shift in the position of equilibrium in a direction that tends to counteract the change in the variable under consideration".

A number of methods for removing iodine from the reaction mixture were then tested, starting with the addition of sodium thiosulfate. However the addition of excess sodium thiosulfate, resulting in complete decolourisation of the reaction mixture, led to a significantly reduced yield (Figure 43). The emergence of a general relationship between yield and time elapsed before decolorisation led to the hypothesis that decolorisation resulted in permanent quenching of the reaction. A similar phenomenon had been noted by Bell.[116] Consequently, sodium thiosulfate was added to the reaction mixture in a dropwise manner, so as to maintain a pale pink colour.

Figure 43 Effect of sodium thiosulfate on yield of dityrosine **109**



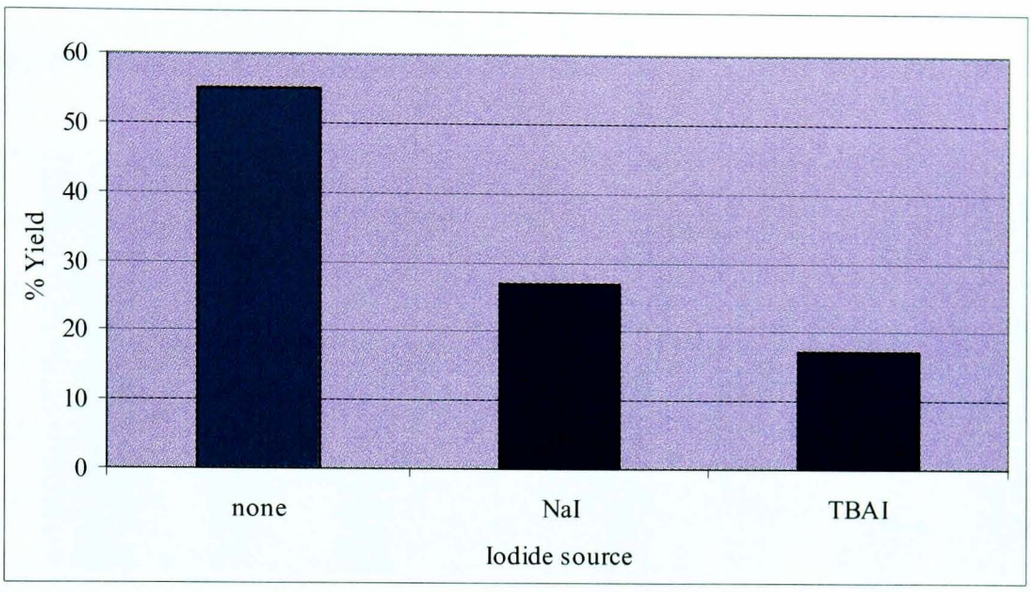
This technique resulted in a significant improvement in the yield of dimer **109**. The amount of sodium thiosulfate added had to be carefully controlled, to prevent quenching of the reaction. A 0.05 M aqueous solution of sodium thiosulfate was added to the reaction mixture in a dropwise manner at regular intervals in order to maintain a pale pink colour. These results indicated that iodine might have a role in the reaction mechanism, but that this mechanism is best facilitated by a low concentration of iodine.

Since sodium thiosulfate converts iodine to iodide ions (Scheme 87) the effect of iodide on the reaction was also considered; silver iodide and TBAI were used to investigate this theory (Figure 44).

Scheme 87

$$I_2 + 2\ S_2O_3^{2-} \longrightarrow 2\ I^- + S_4O_6^{2-}$$

Figure 44 Effect of iodide on yield of dityrosine **109**



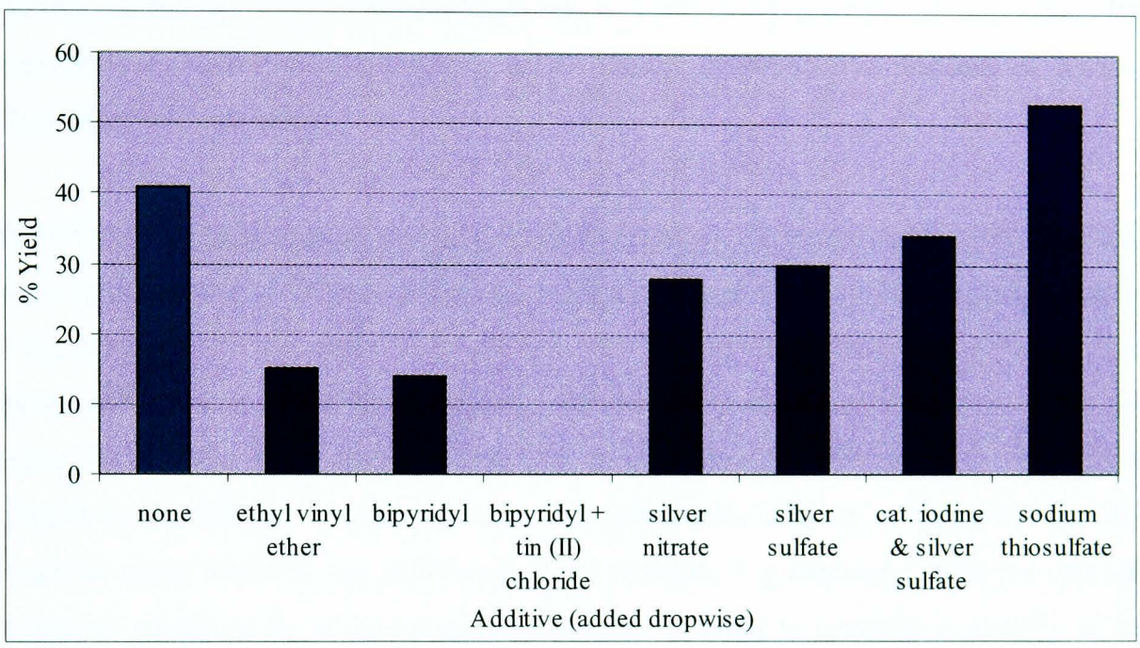
These reactions clearly showed that the presence of iodide ions reduced the yield of the reaction. It was interesting to note that while the addition of one equivalent of sodium iodide significantly reduced the yield, the use of the more lipophilic TBAI resulted in a further reduction in yield. This suggested that iodide ions might play an inhibitory role in a reaction occurring in the organic layer.

Following these results, a method for the removal of iodine from the reaction mixture without the formation of iodide ions was sought (Figure 45). The use of an alkene was considered since alkenes reversibly bind iodine; simple alkenes would also reside largely in the organic layer rather than the aqueous layer, which may lead to more efficient iodine sequestration. A range of alkenes was screened against solutions of iodine in DCM for their decolourising ability; ethyl vinyl ether appeared to be the most successful, and was added dropwise to the reaction mixture as a 0.05 M solution in DCM. The use of tin (II) chloride and bipyridyl was tested since there is evidence that these two molecules form a complex which can also bind iodine;[120] bipyridyl was added alone as a control.

Since the pH of a mixture of iodine and water was found to steadily decrease over several days, there must be an equilibrium between iodine and iodide in water (resulting

in the formation of hydroiodic acid, hence the decrease in pH). The addition of silver salts to the reaction mixture was therefore also investigated, since silver halides are readily formed and are insoluble; this would irreversibly remove iodide ions and consequently iodine from the reaction mixture.

Figure 45 Effect of other iodine-sequestering additives on yield of dityrosine **109**



However, none of these alternative iodine-sequestering reagents resulted in a better yield than the original reaction, so addition of sodium thiosulfate remained the optimum technique for the removal of iodine from the reaction mixture.

2.5.4.3 Conclusions

1) Light and temperature were found not to play a significant role in the reaction mechanism, implying a polar rather than a radical mechanism. These results confirm those reported by Bell and Bowman.[115]

2) Iodine appeared to play a role in the mechanism, with the presence of both molecular iodine and iodide ions found to retard or limit the coupling reaction. However, the presence of a small amount of iodine was necessary for the reaction to proceed.

3) The optimum method for limiting the accumulation of iodine was dropwise addition of a 0.05 M aqueous solution of sodium thiosulfate in order to maintain a pale pink colour.

2.5.5 Scaling Up the Reaction

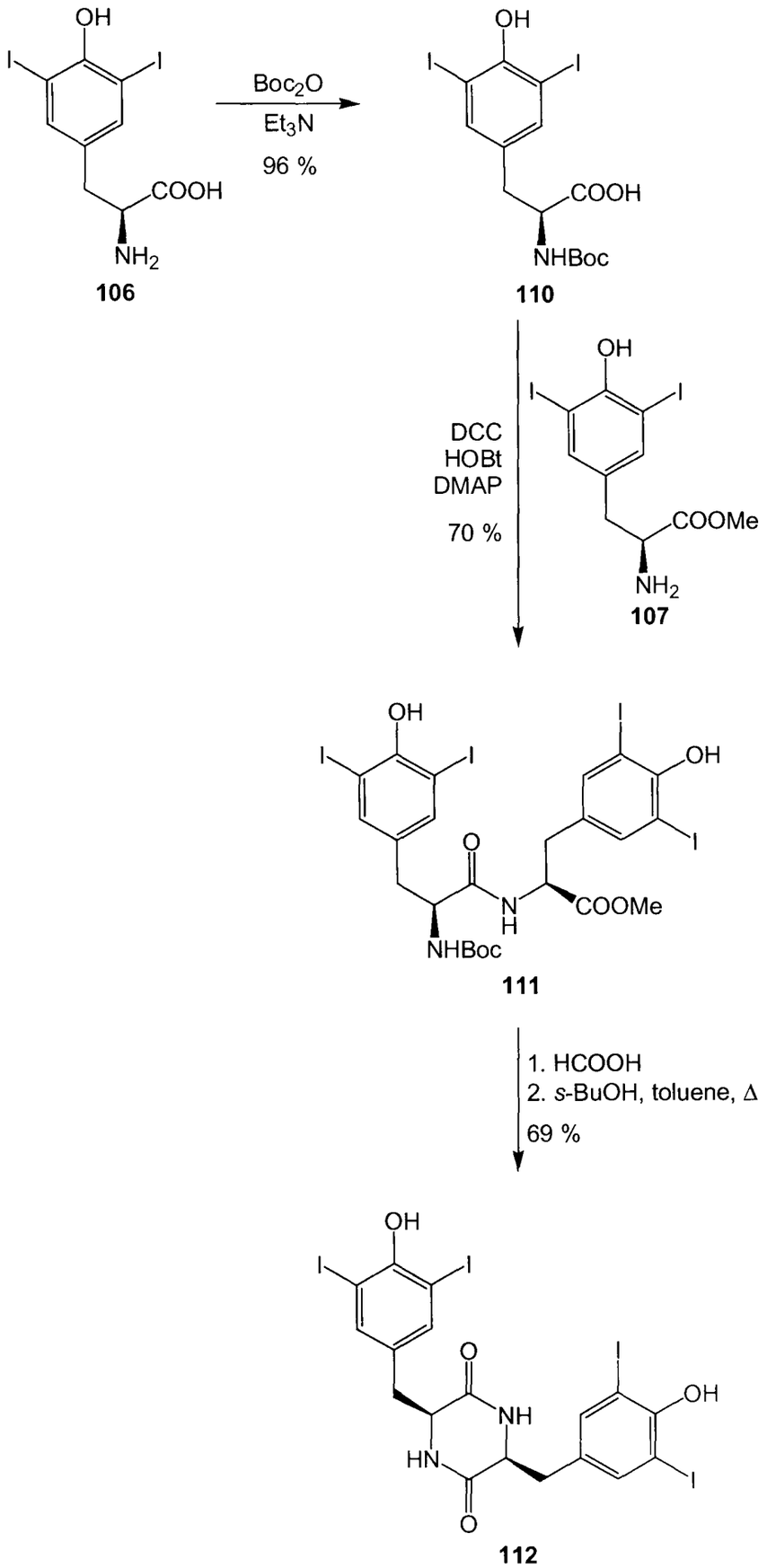
Having performed much optimisation work, a yield of 53 % of the desired dimer **109** was obtained using a concentration of 30 mg/ml and dropwise addition of sodium thiosulfate over 24 hours.

However, the optimisation work was carried out on a small scale, with reactions performed on 100 to 200 mg of starting material. When the reaction was performed on a 1 g scale, the yield dropped to an average of 21 %. At larger scales only traces of the dimer **109** were observed. Quantities of solvents were scaled up correspondingly, but since the reaction was a biphasic one, it is possible that the efficiency of mixing was altered by the change in scale and that this reduced the success of the reaction. The reaction could therefore be performed using multiple 1 g reactions, with the reaction mixtures combined for work-up and purification, in order to generate a quantity of the diiodotyrosine **109**. However, this method was inconvenient, and the desire to generate the heterocyclic rings of the herqulines led to the consideration of an intramolecular reaction.

2.5.6 Intramolecular Bowman Coupling

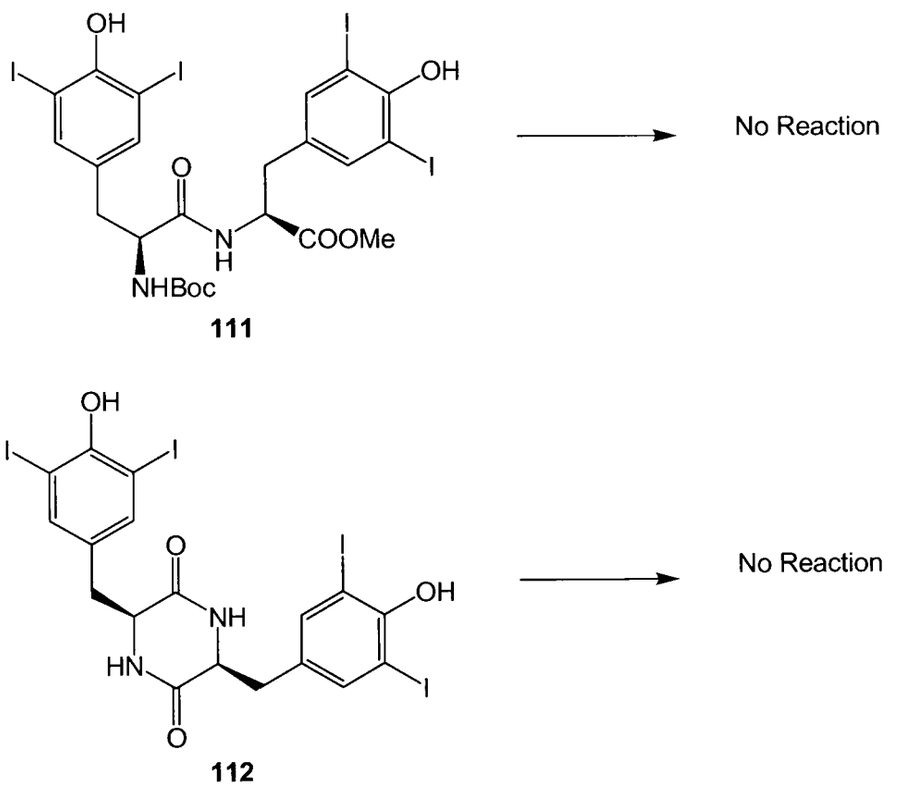
In an attempt to improve the efficiency of the Bowman coupling reaction, the possibility of an intramolecular reaction was examined; no attempts at such reactions had been previously reported. *N*-Boc-diiodo-L-tyrosine **110** was synthesised in excellent yield from diiodo-L-tyrosine dihydrate using Boc anhydride and triethylamine (Scheme 88). The acid **110** was then treated with the previously generated amine **107** (see Scheme 85 for preparation) and standard peptide coupling reagents to give the tetraiododipeptide **111**. The corresponding diketopiperazine **112** was also generated, using the conditions of Nitecki discussed previously (Section 2.2.7.1).[65]

Scheme 88


OH
I
I
COOH
NH2
106
Boc2O
Et3N
96 %
NHBoc
110
DCC
HOBt
DMAP
70 %
COOMe
107
O
N
H
111
1. HCOOH
2. s-BuOH, toluene, Δ
69 %
NH
HN
112

Unfortunately, when the dipeptide **111** and the diketopiperazine **112** were treated with the Bowman coupling conditions (reactions were carried out in duplicate, using either sodium thiosulfate or a catalytic amount of iodine followed by silver sulfate) no coupling was observed (Scheme 89). No pink colour was observed without addition of iodine in either case; the diketopiperazine **112** also suffered from the solubility problems discussed previously (Section 2.2.7.2) and did not dissolve in the biphasic solvent system.

Scheme 89



Reaction conditions: DCM, pH 6 phosphate buffer, $Na_2S_2O_3$ *or* cat. I_2/Ag_2SO_4.

As the intramolecular couplings had been unsuccessful, the route was not pursued further.

2.6 Conclusions

A model biphenol was synthesised *via* oxidative coupling using ferric chloride, but this and other oxidative coupling techniques failed to generate a dityrosine molecule.

The nickel-mediated Ullmann coupling of 2-iodo-4-methyl anisole was used to generate a model biphenyl, but the reaction was not successful when applied to a tyrosine system, presumably because of the increased steric demand of the protected amino acid.

The Bowman coupling reaction provided a diiododityrosine, and extensive optimisation work led to a moderate yield. However, at larger scales the reaction was not successful, presumably due to changes in the efficiency of mixing.

CHAPTER 3

REDUCTION OF A DITYROSINE SYSTEM

3.1 Introduction

The reduction of a dityrosine system was the second key step in the total synthesis of the herqulines as discussed previously (Section 1.6). This chapter contains background information relating to techniques for the reduction of aromatic rings as well as a discussion of results obtained during the course of this project.

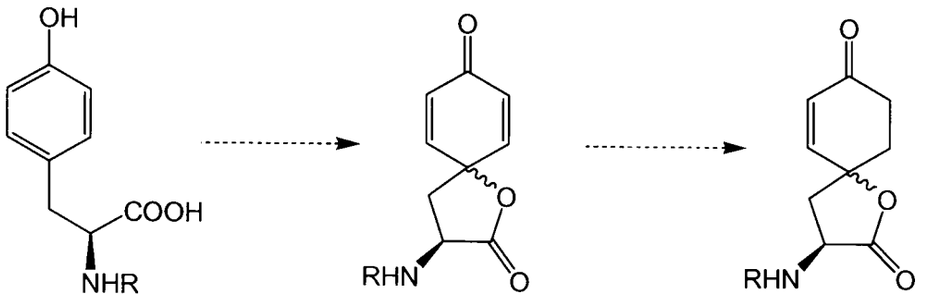
The classical method for the reduction of aromatic rings is metal-ammonia reduction, often described as the Birch reduction. The treatment of benzenoid compounds with a solution of an alkali metal in liquid ammonia, in the presence of an alcohol, leads to the formation of 1,4-cyclohexadienes. The Birch reduction was developed during the 1940s as part of the synthesis of the steroid hormone 19-nortestosterone (nandrolone). The reaction's efficiency and potential synthetic utility have meant that it has remained in constant use ever since.[121]

Metal-ammonia reduction is unique in that it is the only set of reducing conditions able to reduce aromatic rings to unsaturated but non-aromatic ones.* The Birch reduction and variations on the reaction were examined in relation to the synthesis of herqulines A and B. Model compounds (often based on *p*-cresol **20**) were used in order to simplify the process (see Sections 3.2-3.4).

One entirely different method of ring reduction was also considered: formation of a spirolactone-dienone system (Scheme 90). The dienone may then be partially reduced in order to generate a more saturated, non-aromatic system (see Section 3.5).

* Forcing conditions can be used to facilitate catalytic hydrogenation of aromatic rings but, since alkenes undergo hydrogenation more readily than arenes, the result is usually a fully saturated system.

Scheme 90

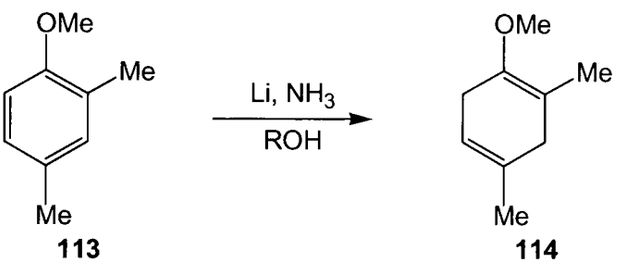


3.2 The Birch Reduction

3.2.1 Introduction

The Birch reduction produces non-conjugated cyclohexadienes from phenyl rings, with regioselectivity favouring the location of electron-donating substituents at olefinic positions.[122] Scheme 91 shows the expected outcome of the Birch reduction of a simple aromatic molecule with the same substitution pattern as dityrosine.[123]

Scheme 91

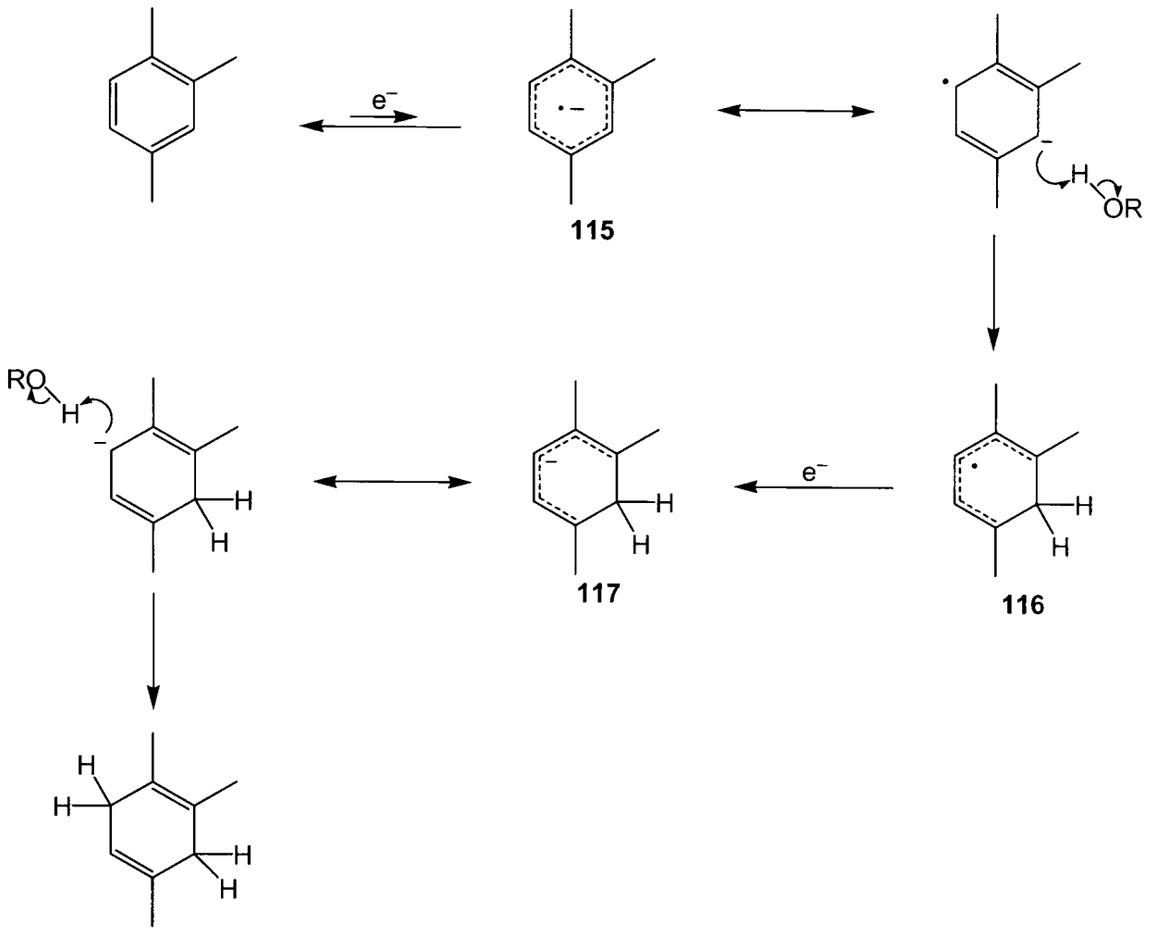


113 **114**

3.2.2 Mechanism of the Birch Reduction

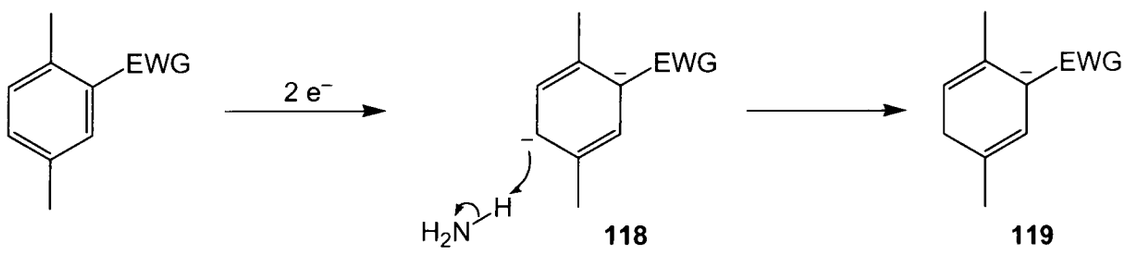
Alkali metals dissolve in liquid ammonia to produce solutions that behave as if they contain metal cations and solvated electrons; these solutions are characteristically deep blue in colour. Aromatic substrates can accept an electron from the solution, resulting in the formation of a radical anion **115** (Scheme 92).[124] This radical anion is then protonated by the alcohol present, and the resulting radical **116** is able to accept another electron from the reaction medium. The anion **117** formed may then be protonated by any excess alcohol present, or remain as the anion until quenched by another protonating (or alkylating)[125, 126] agent.

Scheme 92



When more activated substrates (such as polyaromatic compounds or aryl carboxylic acids) are reduced, addition of a second electron to the radical anion may occur, resulting in the formation of a dianion **118** (Scheme 93). This dianion is often sufficiently basic to be protonated by ammonia, so that the addition of an alcohol to the reaction mixture is unnecessary and often detrimental to the yield.[122] The stabilised anion **119** then survives until it is quenched by a protonating (or alkylating) agent.

Scheme 93



3.2.3 The Formation of 1,4-Cyclohexadienes

A number of explanations have been put forward for the formation of 1,4-cyclohexadienes rather than conjugated 1,3-cyclohexadienes:

1) The addition of one electron initially results in the formation of a delocalised radical anion species **115**. Protonation of the radical anion then takes place, with the site of protonation governed by a) reaction at a position of high electron-density and b) generation of a stable radical species.

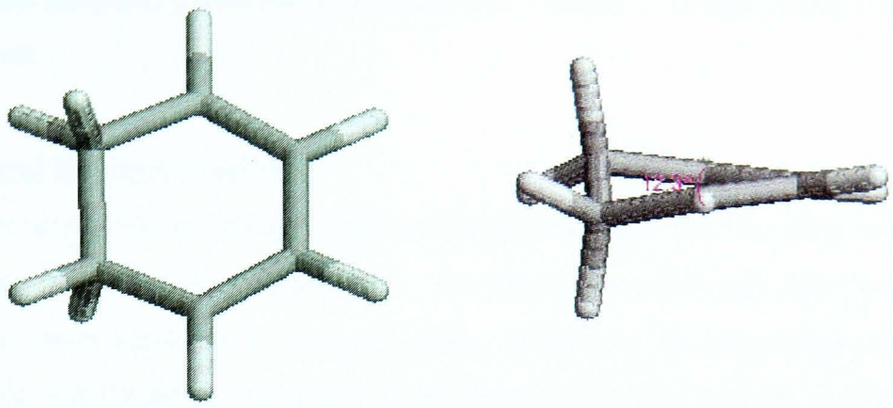
2) It has been proposed that the non-conjugated product is formed as a result of protonation of the monoanion **117** at the position of highest electron density, but theoretical methods have produced differing results when calculating the electron densities of the monoanion **117**.[127]

3) Although its validity has been widely questioned, the 'principle of least motion'* has also been used to explain the formation of 1,4-cyclohexadienes.[53] When average bond orders of the monoanion are considered, protonation at the position '*para*' to the first protonation would result in the smallest change to the electron distribution.

4) The Birch reduction is considered a good example of a kinetically-controlled reaction, as the product is a non-conjugated cyclohexadiene. The conjugated diene is thermodynamically more stable, although the energy difference is actually very small (1.1 kJ mol^{-1}).[128] This is because the 1,3-cyclohexadiene system does not exist in a planar conformation, and therefore cannot be fully conjugated. The non-planar conformation is thought to be a result of ring strain at the saturated carbons and a steric interaction between the methylene hydrogens.[128]

* The principle of least motion states: "those elementary reactions will be favoured that involve the least change in atomic position and electronic configuration".

Figure 46 1,3-Cyclohexadiene



Once formed, a 1,4-cyclohexadiene can undergo alkene isomerisation *via* deprotonation by a metal amide (MNH_2). However lithium amide does not react with cyclohexadienes; consequently lithium is the metal most widely used in Birch reductions.[129] Therefore protonation of the monoanion is effectively irreversible and the 1,4- and 1,3-cyclohexadienes are formed in a ratio reflecting the relative rates of protonation of the respective monoanions. Sodium amide and potassium amide are able to deprotonate cyclohexadienes, and therefore the use of sodium or potassium in Birch reductions results in formation of a greater proportion of the conjugated cyclohexadiene. Sodium and potassium also form the amide more readily than lithium, especially in the presence of transition-metal salts.

3.2.4 Regiochemistry of the Birch Reduction

In most cases the Birch reduction proceeds with reliable and predictable regiochemistry. Electron-donating substituents are found at olefinic positions of the product (Scheme 91), and electron-withdrawing substituents at sp^3 centres (Scheme 93) as far as possible. The reason for this is clear from the mechanism: carbanions are most stable when formed adjacent to an electron-withdrawing group (or distant from an electron-donating one). Protonation then occurs, resulting in the formation of sp^3 centres at the sites of the carbanions.

3.2.5 Application to Model Systems

In order to gain familiarity with the Birch reduction, a series of simple model reductions was carried out.

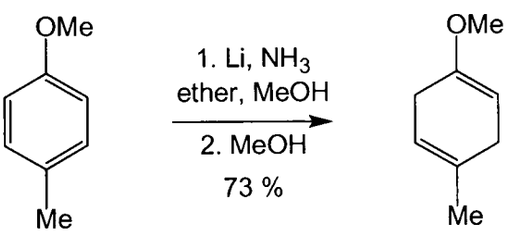
3.2.5.1 General Reaction Conditions

Lithium is generally the preferred metal for Birch reductions as it is more soluble in ammonia than sodium and potassium.[130] As discussed previously (Section 3.2.3) lithium has a lower tendency to form its amide, which can facilitate other reactions. Lithium amide also results in less double bond isomerisation than sodium amide; double bond isomerisation could lead to conjugated dienes and therefore possibly to over-reduction. Consequently, lithium was chosen as the metal to be employed. Ammonia was condensed and distilled from sodium to remove moisture and traces of iron salts, and equipment was flame-dried and flushed with argon before use.*

3.2.5.2 Birch Reductions

The Birch reduction of 4-methyl anisole **66** was performed (Scheme 94);[131] the compound was chosen in order to model a protected tyrosine system. The expected cyclohexadiene **120** was isolated in good yield.

Scheme 94

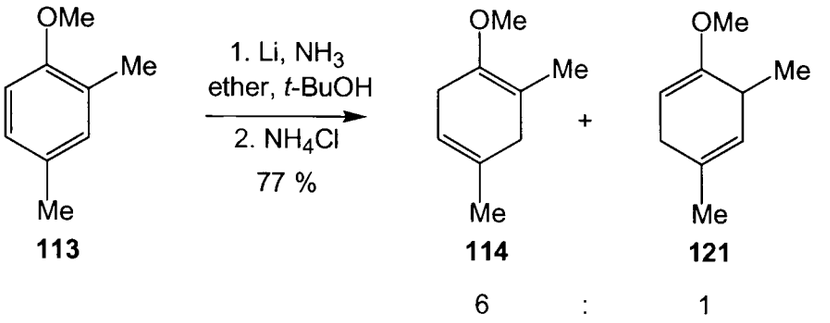


66 **120**

Similarly, 2,4-dimethyl anisole **113** was chosen to model a protected dityrosine system. The anisole **113** was subjected to Birch conditions[123, 132] and a mixture of the expected cyclohexadiene **114** and a small amount of the regioisomer **121** was isolated (Scheme 95).

* Lithium and nitrogen readily react to form lithium nitride.

Scheme 95

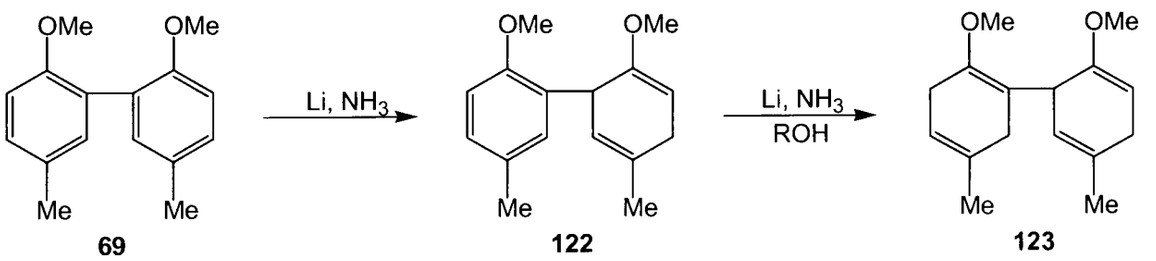


3.2.6 Birch Reductions of Biphenyl Systems

3.2.6.1 Introduction

The reduction of a biphenyl system follows the regiochemical guidelines described previously (Section 3.2.4) but sequential reduction of the two aromatic rings leads to an interesting regiochemical result. The reduction of one aromatic ring proceeds in the absence of an alcohol and with again predictable regiochemistry – the link to the second aromatic ring is always at a saturated position (Scheme 96).[133] This regiochemistry arises as a result of formation of the highly resonance-stabilised benzylic anion. The reduction of the second aromatic ring requires the presence of an alcohol and results in the standard expected regiochemistry, that is with electron-donating substituents located at olefinic positions.[133] Consequently, the symmetrical biphenyl **69** is reduced to give the unsymmetrical bis(cyclohexadiene) system **123**.

Scheme 96

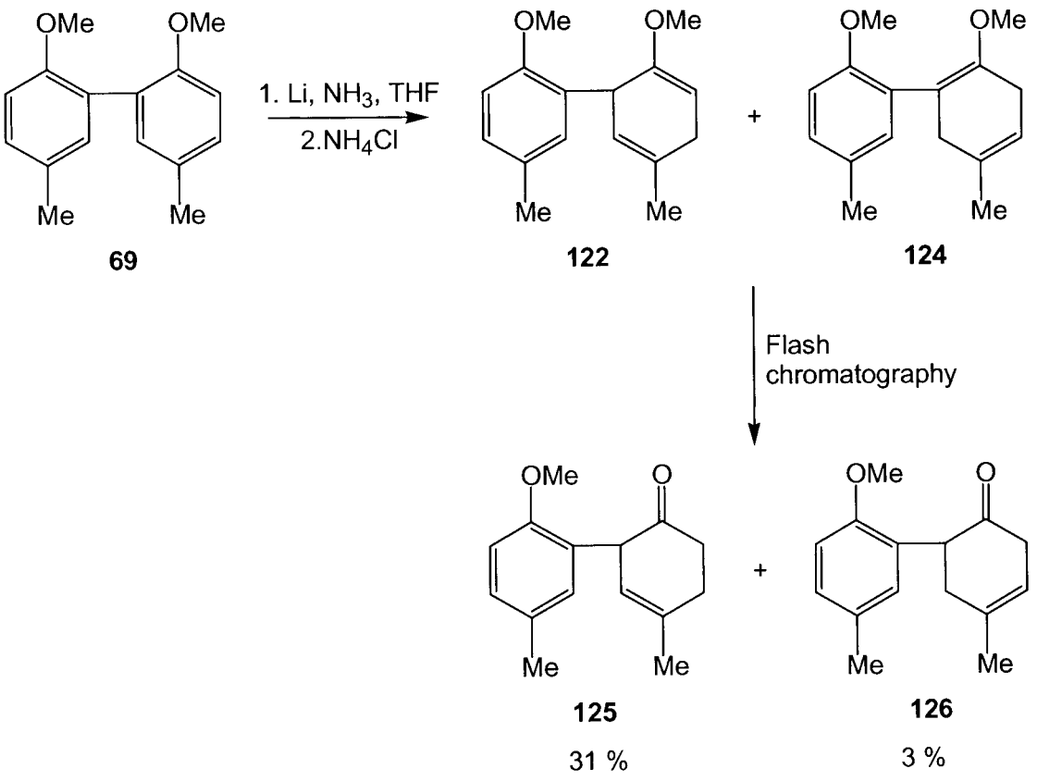


3.2.6.2 Application to a Model System

In order to gain further experience with the Birch reduction, the methyl anisole dimer **69** (see Section 2.4.2 for preparation) was chosen to model a protected dityrosine system.

The dimer **69** was treated with lithium and liquid ammonia. In order to reduce just one ring and produce the aryl cyclohexadiene **122**, alcohol was omitted and the reaction was quenched with ammonium chloride (Scheme 97).[133] This reaction was initially performed by adding lithium to a solution of model dimer **69**; it was later noted that addition of model dimer **69** to a solution of lithium gave rise to fewer by-products. The Birch reduction was repeated at –78 °C instead of at reflux (–33 °C), but after the same period of time ^{1}H-NMR spectroscopy showed that some starting material remained. This indicated that the reaction had been slowed, but no improvement in selectivity was observed.

Scheme 97



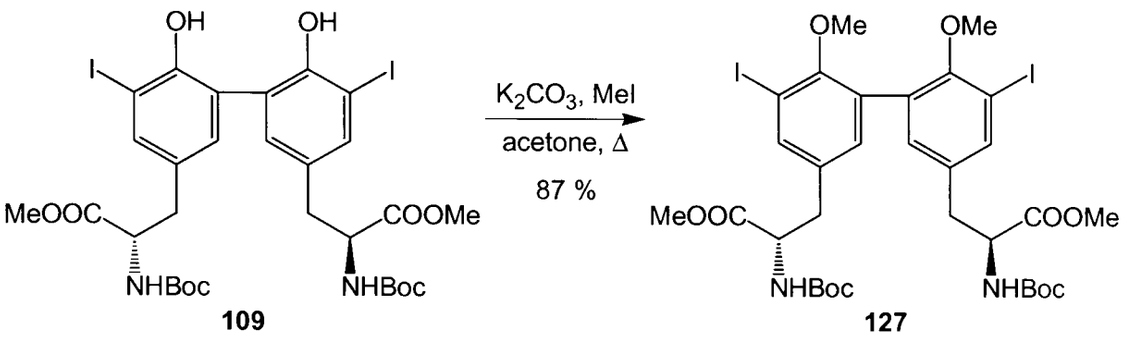
In all cases the expected cyclohexadiene **122** was the major one, with a small amount of the other regioisomer **124** also observed; hydrolysis to the corresponding β,γ-enones **125** and **126** was observed as a result of flash chromatography.

3.2.6.3 Application to the Dityrosine System

Having synthesised a diiododityrosine **109** *via* the Bowman coupling reaction (Section 2.5.2-3) the Birch reduction of this system was considered. Since the Birch reduction of

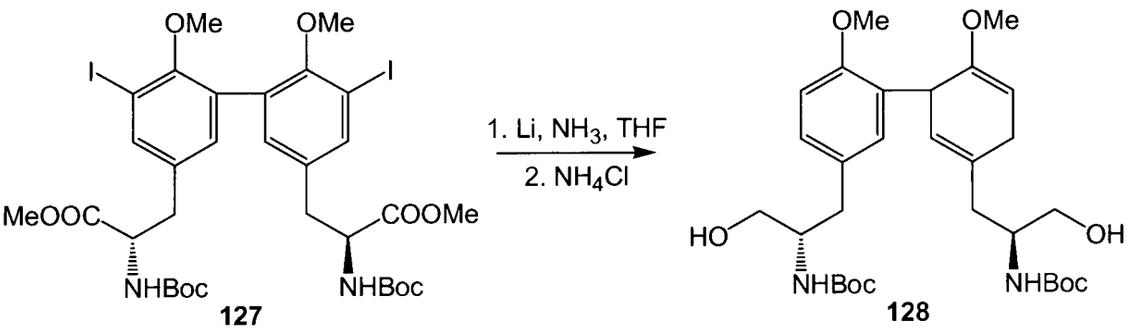
aryl halides was unreported, investigation of the system was required. As phenols do not undergo Birch reduction, the phenol hydroxyl groups were protected to give the bis(methyl ether) **127** in good yield (Scheme 98).

Scheme 98



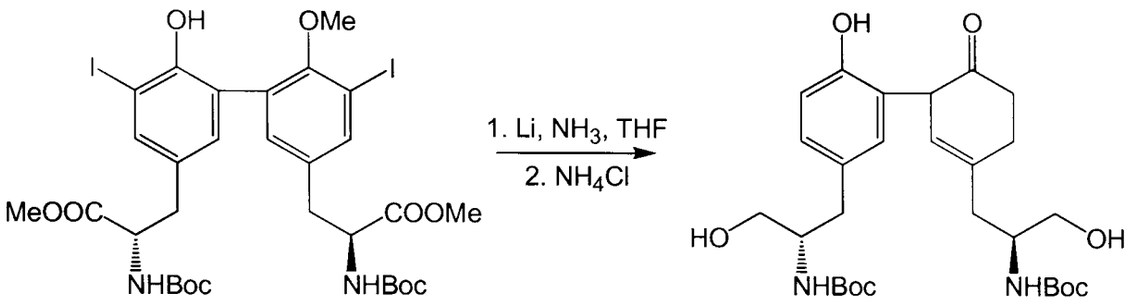
The fully protected diiododityrosine **127** was then treated with lithium and liquid ammonia and the reaction quenched with ammonium chloride (Scheme 99). The presence of peaks at 5.6-4.6 ppm in the ^{1}H-NMR spectrum of the crude product indicated that some ring reduction had occurred but, as expected, some aromatic signals remained. LCMS was used to determine that deiodination had occurred – no material retaining iodine atoms was observed. The major product of the reduction was identified as the aryl cyclohexadiene **128**, which had undergone deiodination and reduction of the ester groups.

Scheme 99



Also identified was the phenolic enone **130**, thought to arise from the Birch reduction of **129** (Scheme 100) – a minor impurity generated by partial methyl protection in the previous step (Scheme 98).

Scheme 100

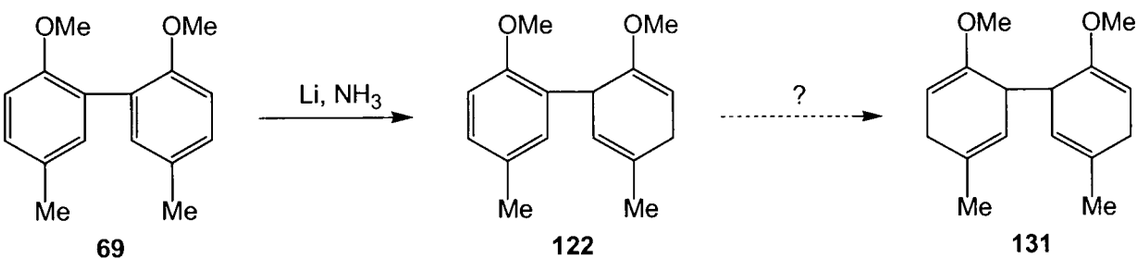


129 **130**

3.2.6.4 The Regiochemical Problem

In order to generate the pattern of unsaturation required by herqulines A and B the reduction of the first aromatic ring might proceed using the standard Birch reduction in the absence of alcohol. However, a second Birch reduction would provide the wrong regiochemistry, and so the reaction had to be altered, or the step replaced with an entirely different one (Scheme 101).

Scheme 101



69 **122** **131**

Several strategies for producing the desired regiochemistry in the reduction of the second ring were considered:

1) The 'photo-Birch' reduction (see Section 3.3).

2) A directed Birch reduction with intramolecular protonation (see Section 3.4).

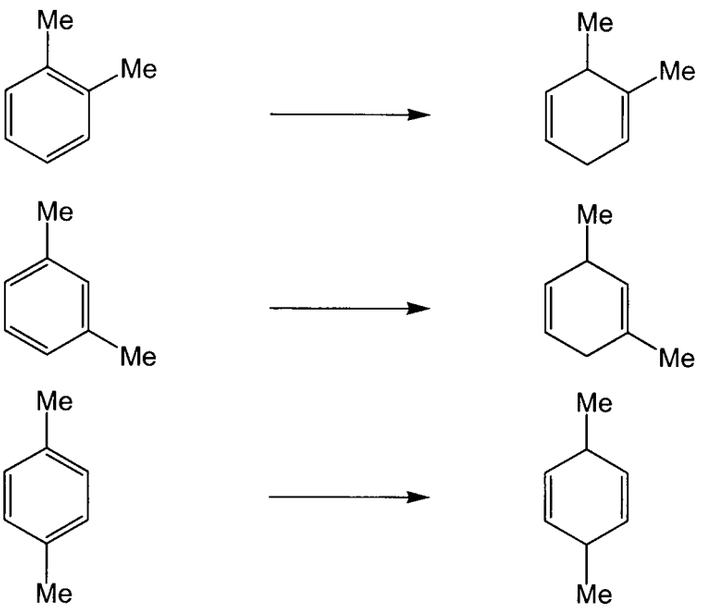
3) Oxidation of the aromatic ring to form a spirolactone, which could then be reduced (see Section 3.5).

3.3 Photo-Birch Reduction

3.3.1 Introduction

The photoreduction of polyaromatic compounds such as naphthalene, anthracene and phenanthrene has been reported using sodium borohydride and a dicyanobenzene.[134, 135] The photoreduction of xylenes to give 1,4-cyclohexadienes under similar conditions was reported by Epling and Florio in 1986.[136] Interestingly, the reaction exhibited regiochemistry opposite to that expected from a Birch reduction (Scheme 102).

Scheme 102 The photo-Birch reduction of xylenes

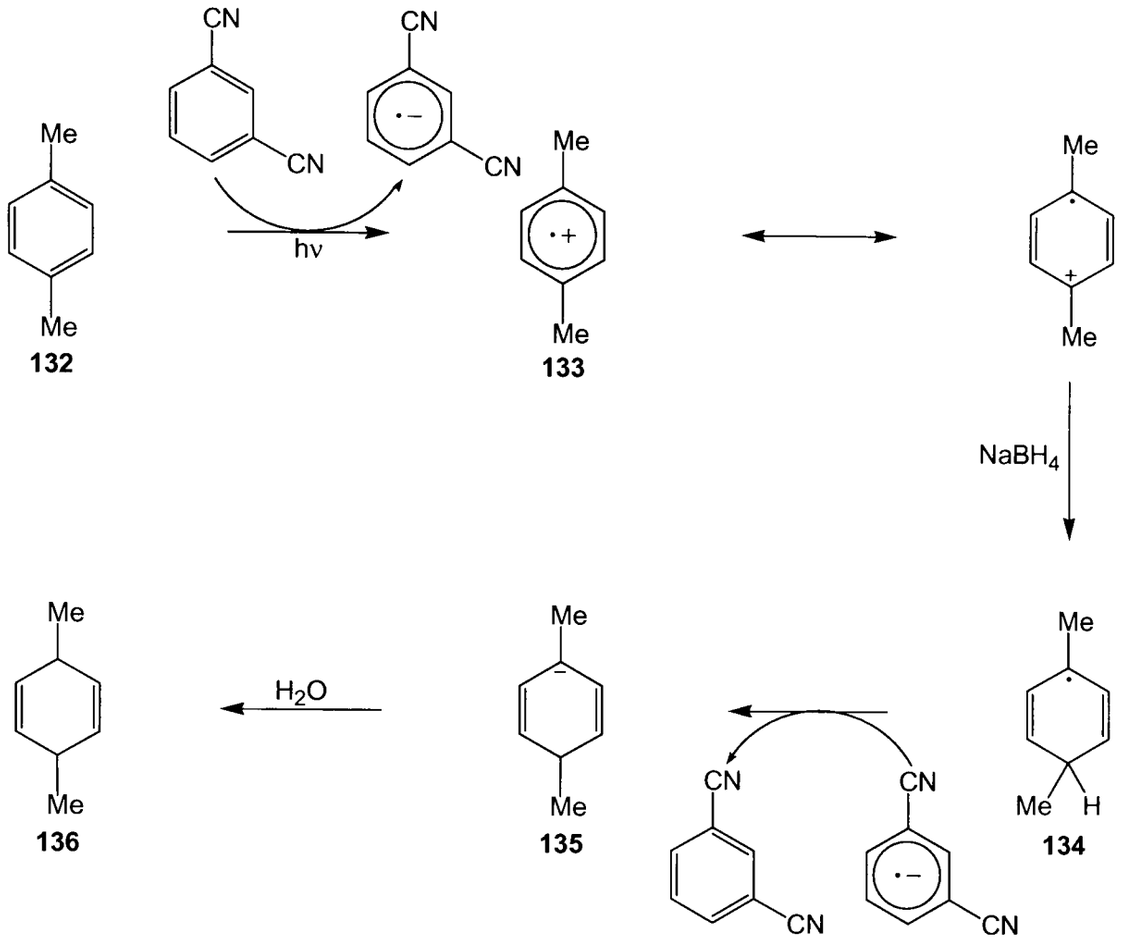


Reaction conditions: $NaBH_4$, 1,3-dicyanobenzene, hv, MeCN, H_2O.

Epling and Florio proposed that the reaction proceeded *via* the formation of a xylyl radical cation **133** from a charge transfer reaction with dicyanobenzene (Scheme 103). Hydride transfer from sodium borohydride then resulted in the formation of a xylyl radical **134** and a second charge transfer process generated a xylyl anion **135** and regenerated the dicyanobenzene. Protonation by water then resulted in the formation of the observed 1,4-cyclohexadiene **136**.

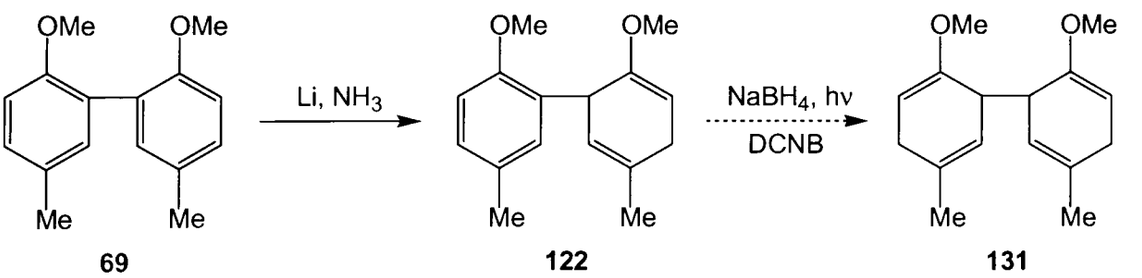
This mechanism accounts for the regiochemical outcome of the reaction since addition of the first hydrogen atom arises from addition of hydride to a radical cation **133**, rather than from addition of a proton to a radical anion, as in the Birch reduction. The position at which the first hydrogen atom is added, is therefore the one at which a positive charge (rather than a negative one) is most stabilised.

Scheme 103 Mechanism of the photo-Birch reduction



The photo-Birch reduction could potentially be used to reduce the second aromatic ring of dityrosine (Scheme 104). However, there are no reports of this type of reaction having been applied to substrates other than xylenes and polyaromatic hydrocarbons. It is also important to recognise that this reaction is based on an initial charge transfer from the substrate to dicyanobenzene. Therefore, any potential substrate must be sufficiently electron-rich to allow this process to occur.

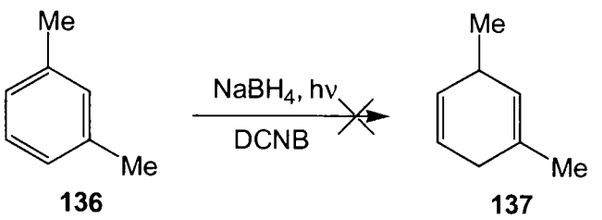
Scheme 104



3.3.2 Application to a Model System

Since the photo-Birch reduction may have been able to provide the solution to the regiochemical problem associated with the synthesis of the herqulines, the reaction was investigated. As there had only been one report of this type of reduction it was decided to repeat the reduction of *m*-xylene described by Epling and Florio (Scheme 105).[136] However, despite a number of attempts, the photochemical reduction could not be repeated. No material was recovered, which may have been due to volatility of the product or to problems experienced extracting a small amount of material from a highly dilute solution.

Scheme 105



136 **137**

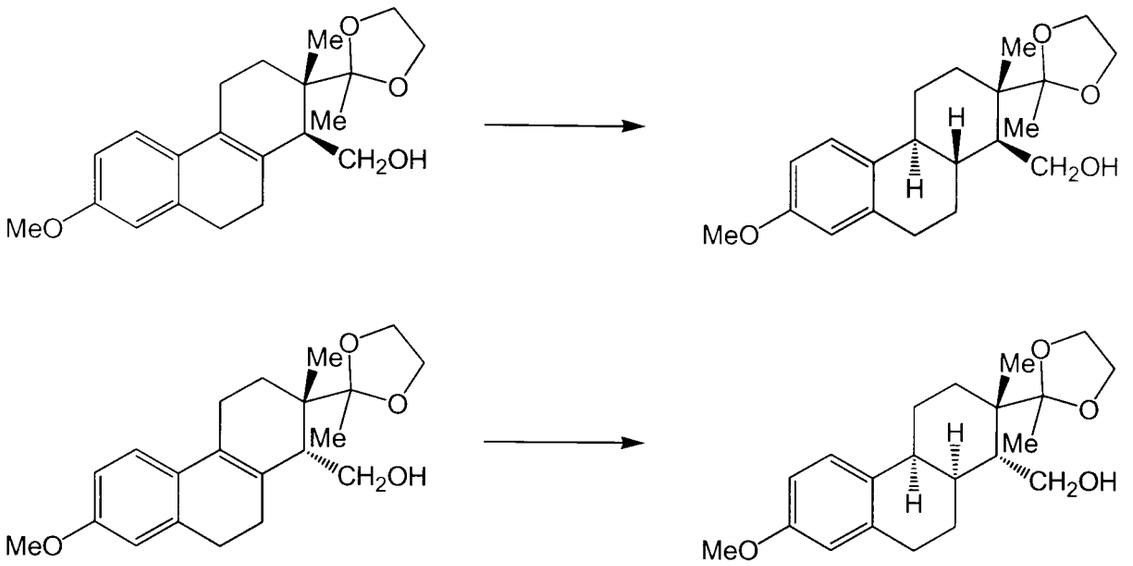
As a result of the failure of this reaction and a lack of familiarity with photochemical techniques, this route was not pursued further.

3.4 Directed Birch Reduction

3.4.1 Introduction

A number of groups have shown that by conducting a Birch reduction without an external alcohol and instead using an intramolecular proton source, the outcome of the reaction can be modified. The presence of a neighbouring hydroxyl group has been shown to enhance the rate of Birch reductions, facilitate over-reduction of the ring[137] and to allow a proton to be 'delivered' to a reduction site, thus controlling the stereochemistry of the product. This strategy has been used to control the stereochemistry of the reduction of styrene double bonds (Scheme 106).[138]

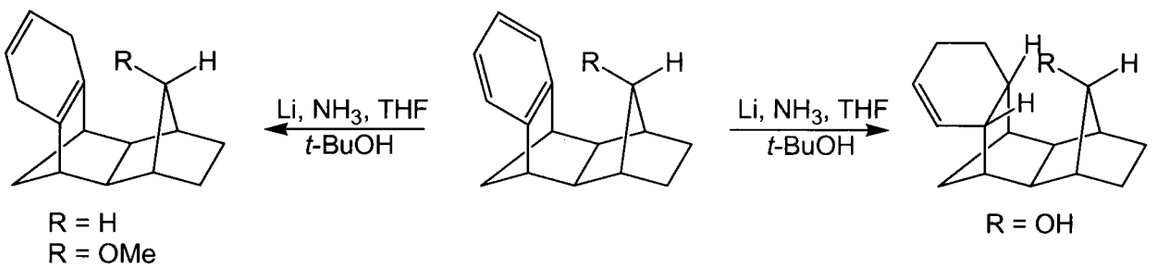
Scheme 106



Reaction conditions: 1. Na, liq. NH_3, THF; 2. NH_4Cl.

A neighbouring hydroxyl group has also been used to modify the course of the Birch reduction of a phenyl ring within a conformationally-restricted molecule (Scheme 107).[139, 140]

Scheme 107



The presence of a hydroxyl group in close proximity to the aromatic ring not only resulted in over-reduction of the ring, but also altered the position of the double bond in the product.

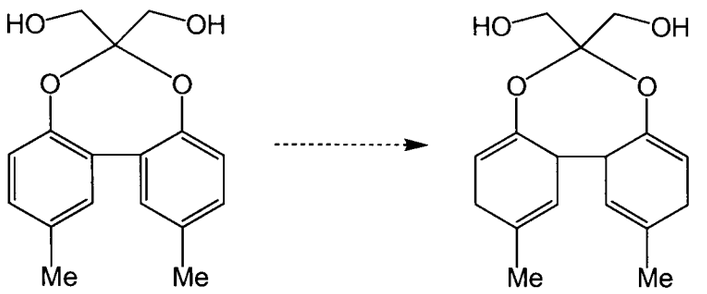
3.4.2 Application to a Model System

3.4.2.1 Introduction

Addition of hydroxyl groups to the model biphenyl system **69** (based on *p*-cresol) was therefore considered. It was hoped that a proximal hydroxyl group could influence the

regiochemistry of the second ring reduction and facilitate the generation of a symmetrical intermediate (Scheme 108).

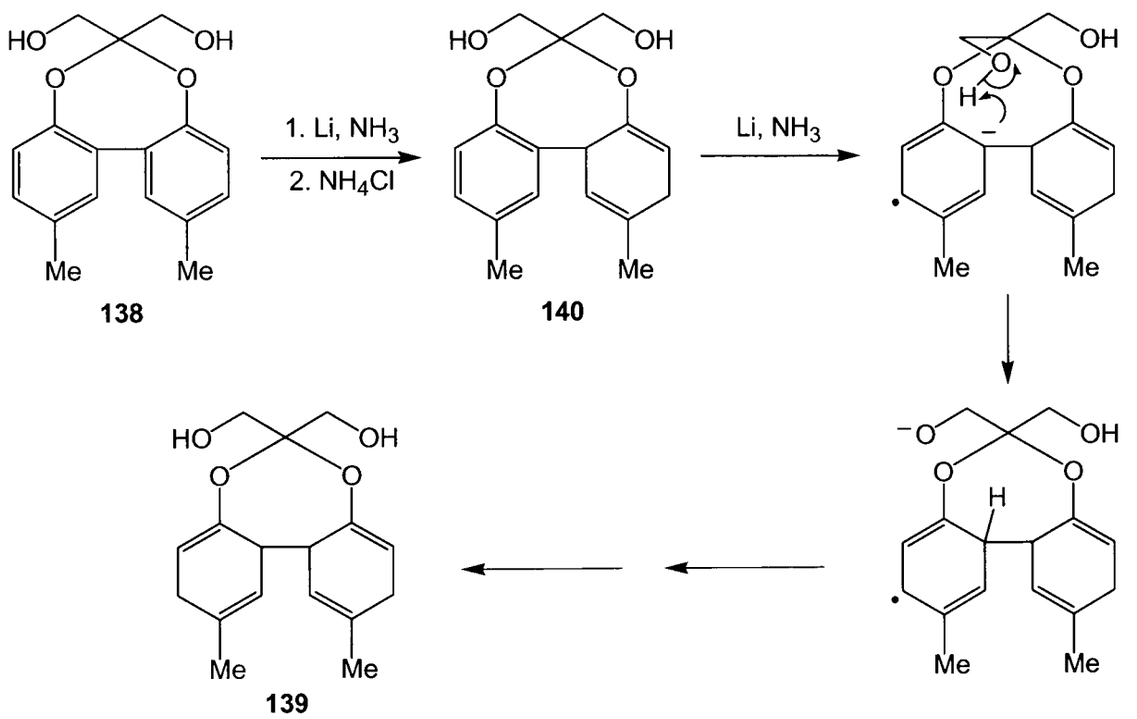
Scheme 108



138 **139**

The reduction of the first ring would proceed as normal. It was hoped that the proximity of the hydroxyl groups would then influence the course of the second ring reduction and deliver a proton to the desired site (Scheme 109).

Scheme 109

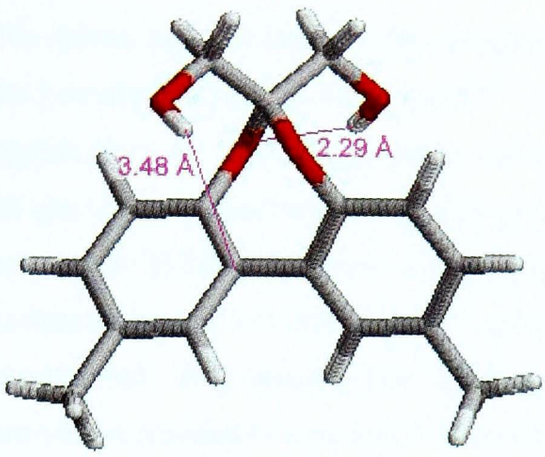


138 **140** **139**

Molecular mechanics calculations were used to perform a conformational search in order to examine low-energy conformations of the proposed model diol **138**. The global energy minimum conformation (Figure 47a) indicated the presence of two intramolecular hydrogen bonds between the hydroxyl groups and the acetal oxygen

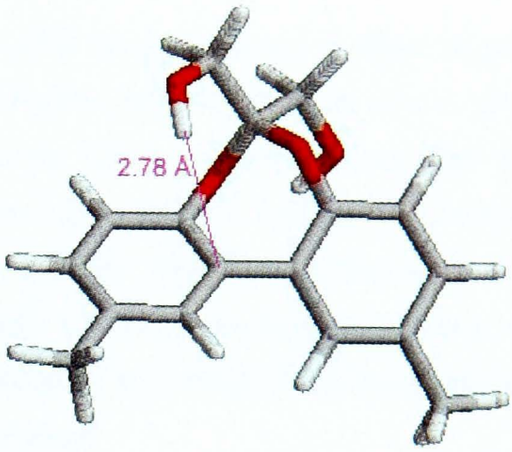
atoms. This caused the hydroxyl groups to take up a position in which the protons were close to the aromatic rings (a distance of 3.48 Å).

Figure 47a



Global energy minimum conformation

Figure 47b



Optimum conformation for proton transfer

It was apparent that a small rotation around the C–O bond would lessen the distance between the proton and the proposed site of protonation. The C–O bond was therefore rotated, and the new distance between the proton and the site of protonation found to be 2.78 Å (Figure 47b). The relative populations of the two conformations at –33 °C were calculated (Table 16), indicating that although the global energy minimum was the favoured conformation, a small proportion of the optimum conformer should also be present, allowing the intramolecular protonation to occur.

Table 16

Conformation	E (kJ mol^{-1})	Boltzmann Factor (–33 °C)	Relative Population
Global minimum	–23.4	1.24×10^5	20
Optimum	–17.4	6.13×10^3	1

3.4.2.2 Synthesis of the Model Compounds

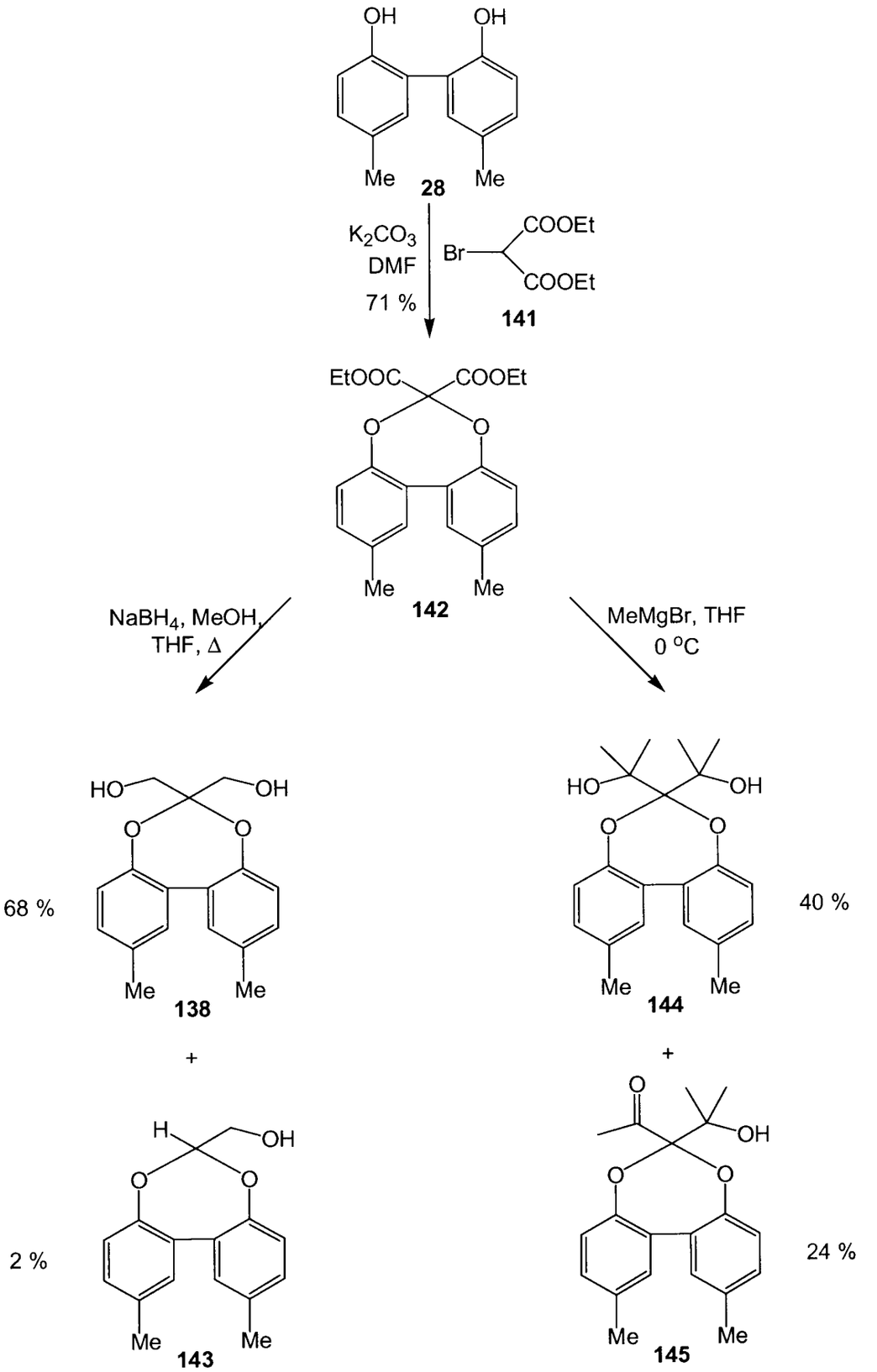
Before the directed Birch reduction could be attempted, a suitable model substrate was required. The diester **142** was synthesised from the *p*-cresol dimer **28** and diethyl bromomalonate **141** (Scheme 110);[141] after much optimisation work, the dropwise addition of diethyl bromomalonate **141** to a solution of the dimer **28** over 8 hours led to

a 71 % yield. The absence of a singlet at 4.84 ppm in the ^{1}H-NMR spectrum (corresponding to the α-proton of diethyl bromomalonate) and observation of a peak at 1770 cm^{-1} (corresponding to the ester carbonyl absorption) in the IR spectrum confirmed the formation of the diester.

The diester **142** was treated with a mixture of sodium borohydride and methanol to give the primary diol **138** in 68 % yield.[142] The reduction was confirmed by loss of ethyl signals from the ^{1}H-NMR spectrum and loss of the carbonyl absorption peak from the IR spectrum. A small amount of a by-product was also obtained: a triplet signal at 5.60 ppm in the ^{1}H-NMR spectrum corresponding to a C*H*CH_2 fragment and observation of a molecular ion at *m/z* 256 by mass spectrometry led to the structural assignment of alcohol **143**. It is possible that this by-product arose as a result of an intramolecular retro-aldol reaction of a partially reduced aldol system.

In order to produce a second model compound, the diester **142** was treated with methyl magnesiumbromide to give the tertiary diol **144** in 40 % yield.[143] The loss of the carbonyl absorption from the IR spectrum and loss of the ethyl fragment from the ^{1}H-NMR spectrum together with the presence of a singlet at 1.46 ppm integrating for twelve protons confirmed the structure of the product. A by-product was also isolated from this reaction: the presence of a carbonyl absorption peak at 1717 cm^{-1} in the IR spectrum and an extra singlet in the ^{1}H-NMR spectrum at 2.16 ppm led to the structural assignment of ketol **145**. The by-product was consistently obtained in 24 % yield despite the addition of excess methyl magnesiumbromide. It is therefore possible that a stable magnesium complex was formed, preventing further addition of the Grignard reagent and limiting the formation of the tertiary diol **144**.

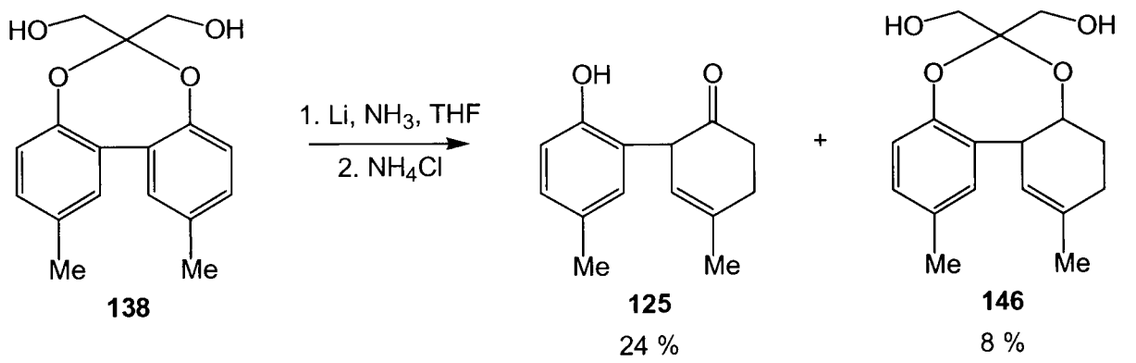
Scheme 110


OH
OH
Me
Me
28
K_2CO_3
DMF
71 %
Br
COOEt
COOEt
141
EtOOC
COOEt
O
O
Me
Me
142
$NaBH_4$, MeOH,
THF, Δ
MeMgBr, THF
0 °C
HO
OH
O
O
Me
Me
138
68 %
+
H
OH
143
2 %
HO
OH
144
40 %
+
O
OH
145
24 %

3.4.2.3 Application to the Model System

The primary diol **138** was subjected to Birch conditions (with no other alcohol present) but only one ring was reduced, as would be expected of any biphenyl under those conditions (Scheme 111). The major product was the β,γ-enone **125**, confirmed by the absence of the CH_2 signal at 3.94 ppm and the presence of a single olefinic signal at 5.55 ppm in the ^{1}H-NMR spectrum of the product. A minor product was also isolated, and was identified as the reduced diol **146** following observation of a singlet at 5.35 ppm (corresponding to a single olefinic proton) and a doublet at 3.65 ppm (corresponding to the methylene groups) in the ^{1}H-NMR spectrum. The coupling constant between the methine protons was measured as 6.3 Hz, and did not therefore provide a useful indication of the stereochemistry of the over-reduced product **146**. No evidence of a non-aromatic product was observed.

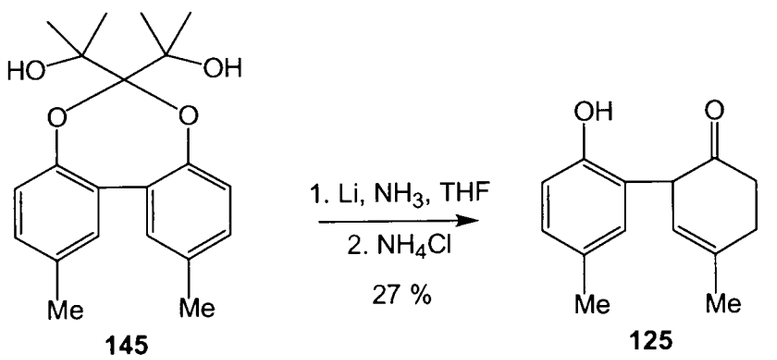
Scheme 111



138 | **125** 24 % | **146** 8 %

Reduction of the tertiary diol **145** also gave the β,γ-enone **125**, although this time as the only isolated product (Scheme 112). Again, there was no evidence of a non-aromatic product.

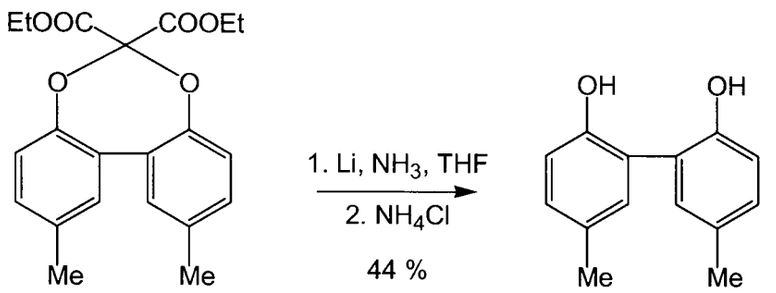
Scheme 112



145 | **125**

The diester **142** was also subjected to Birch conditions in order to establish if the diol could be generated *in situ*, thus reducing the number of steps in the synthesis (Scheme 113). However, this only resulted in hydrolysis of the acetal moiety, regenerating the *p*-cresol dimer **28**. Phenols are stable to Birch conditions,* and consequently no ring reduction was observed.

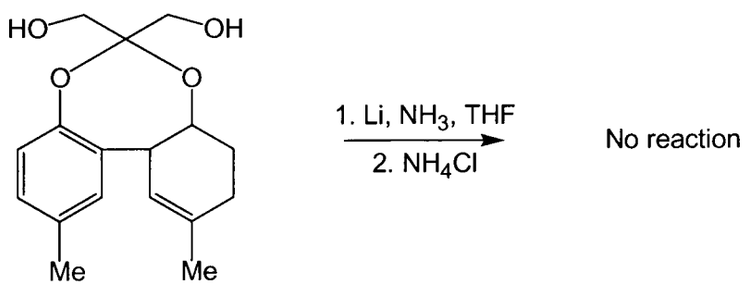
Scheme 113



142 **28**

A further reduction of the aryl cyclohexadiene **146** (Scheme 114) was attempted in order to rule out the possibility that the hydroxyl groups had become co-ordinated to lithium ions during the reaction, rendering them unable to deliver protons. Having worked up the reaction and regenerated the free hydroxyl groups it was hoped that the second aromatic ring may have been reduced. However, no reaction occurred and the starting material was recovered.

Scheme 114



146

Although the reduction of both aromatic rings of the biphenyl systems examined did not proceed as had been hoped, the isolation of an over-reduced product indicated that the presence of proximal hydroxyl groups had influenced the course of the reaction.

* The phenoxide anion is formed; the negative charge then prevents the molecule from accepting electrons from the reaction medium.

Having tried a number of different variations and conditions in order to facilitate the directed Birch reduction without success, it was decided to move on and investigate another method of achieving the unsaturation required for the synthesis of the herqulines – reduction *via* spirolactones.

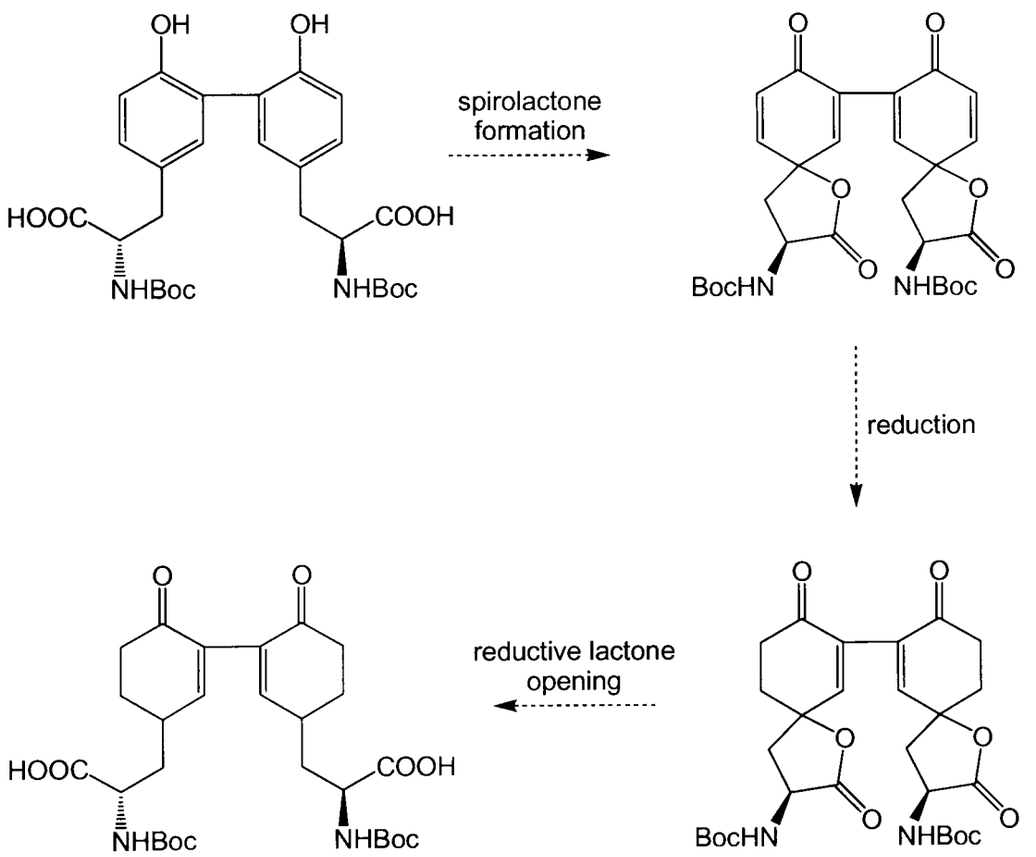
3.5 Reduction *via* Spirolactone Formation

3.5.1 Introduction

3.5.1.1 Strategy

The formation of a dityrosine dispirolactone moiety would result in the formation of two double dienone systems. It was hoped that the least hindered double bonds could be selectively reduced; reductive opening of the spirolactones would then generate a reduced dityrosine molecule (Scheme 115).

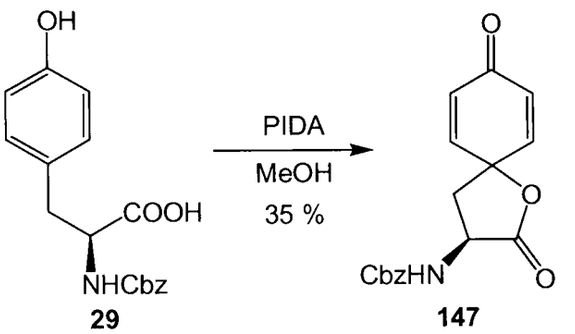
Scheme 115



3.5.1.2 Formation of Tyrosine Spirolactones

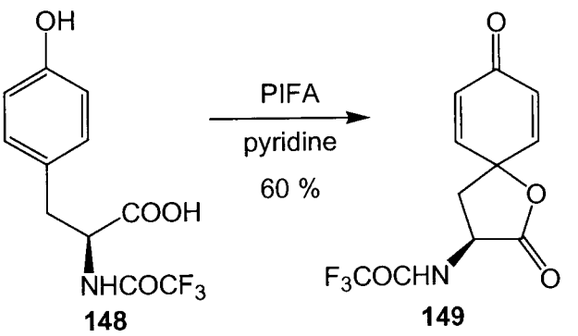
The formation of tyrosine spirolactones has been facilitated by a range of mild oxidising agents including hypervalent iodine reagents. Wipf and Kim used phenyl iodine(III) diacetate (PIDA) to generate a tyrosine spirolactone **147** as part of work towards the total synthesis of *Stemona* alkaloids (Scheme 116).[144, 145]

Scheme 116

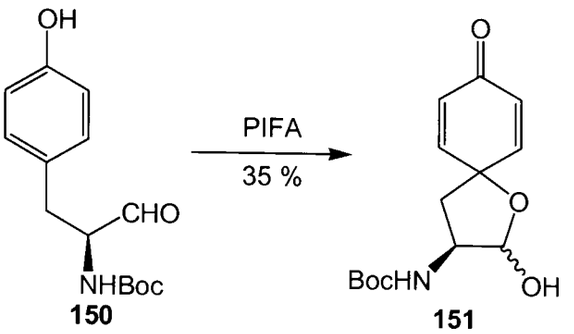


Phenyl iodine(III) bis(trifluoroacetate) (PIFA) has also been used to generate tyrosine spirolactones such as **149** (Scheme 117). [146] Interestingly, PIFA has also been used to synthesise a spirolactol **151** from a protected tyrosinal **150**, affecting oxidation of the phenyl ring without oxidation of the aldehyde moiety (Scheme 118).[147]

Scheme 117

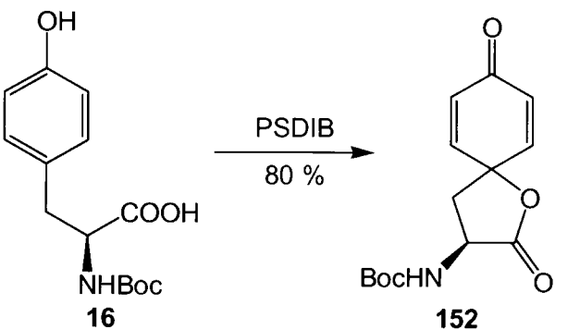


Scheme 118



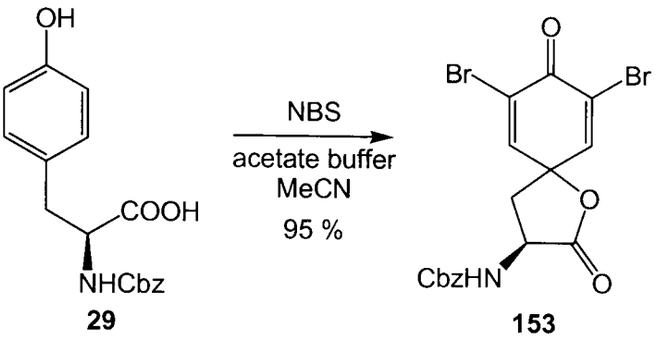
A polymer-supported version of PIDA, polymer-supported diacetoxyiodo(III) benzene (PSDIB), has been used to generate similar spirolactones such as **152** (Scheme 119). The method provided consistently higher yields of the desired product than either PIDA or PIFA.[148] However, the technique does require the preparation of the resin-bound reagent.

Scheme 119



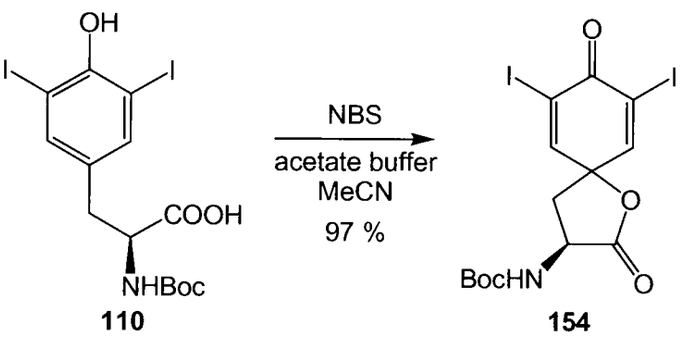
The use of *N*-bromosuccinimide (NBS) in the synthesis of tyrosine spirolactones was first reported by Schmir in 1959 (Scheme 120)[149] and has been used more recently by Reiker as part of an investigation of spirolactone formation techniques.[150] The method generates spirolactones in excellent yields, but has one drawback: unsubstituted tyrosines undergo bromination at the positions *ortho* to the hydroxyl group. However, if these positions are already substituted, no bromination occurs (Scheme 121).[149]

Scheme 120



29 **153**

Scheme 121

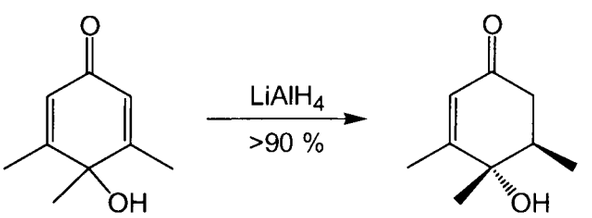


110 **154**

3.5.1.3 Reduction of the Dienone System

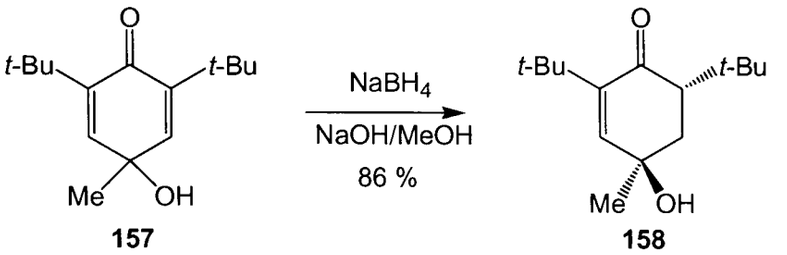
Following spirolactone formation, reduction of the least hindered double bonds of the dienones was considered. Selective reduction of one side of an hydroxylated dienone system has been achieved using lithium aluminium hydride (Scheme 122)[151] or sodium borohydride (Scheme 123).[152] Both reactions gave good yields of the desired enones and both showed a high degree of stereocontrol, with addition of hydride from the same face as the hydroxyl group, presumably arising from co-ordination of the hydride reagent to the hydroxyl group.

Scheme 122



155 **156**

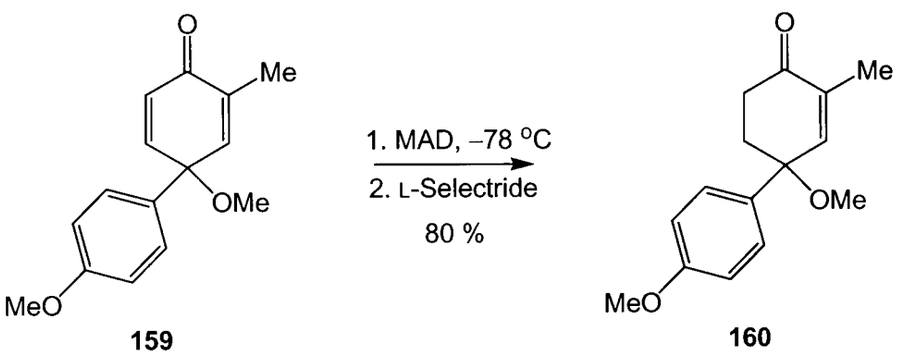
Scheme 123



157 **158**

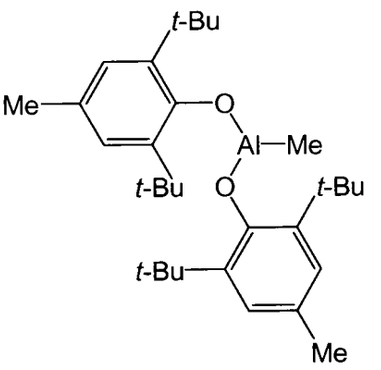
One example of the reduction of the least substituted double bond of a dienone system has been reported by Doty and Morrow (Scheme 124).[153] The use of the bulky Lewis acid bis-(2,6-di-*t*-butyl-4-methylphenoxy)methylaluminium (MAD; Figure 48)[154] followed by addition of L-Selectride gave regioselective reduction of the least hindered double bond in good yield.

Scheme 124



159 **160**

Figure 48 Bis-(2,6-di-t-butyl-4-methylphenoxy)methylaluminium (MAD)



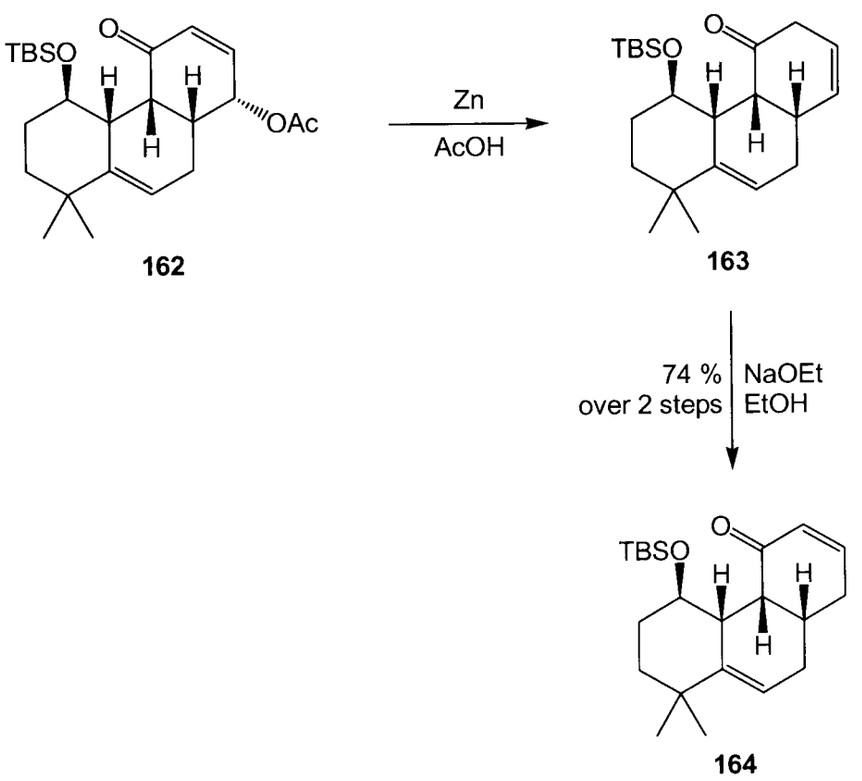
161

3.5.1.4 Reductive Lactone Opening

In order for the reduction of the aromatic rings *via* spirolactones to be successful, the spirolactones must be opened after reduction in order to allow formation of the heterocyclic rings of herqulines A and B. No examples of the reductive opening of spirolactones have been reported, but several groups have detailed the reductive cleavage of allylic esters.

The reductive cleavage of allylic acetates using zinc and acetic acid has been reported as part of the synthesis of analogues of forskolin (Scheme 125).[155] The reduction of allylic acetate **162** resulted in the formation of the β,γ-enone **163**; the reduction was followed by an isomerisation step in order to generate the α,β-enone **164**, and both steps proceeded in good yield.

Scheme 125

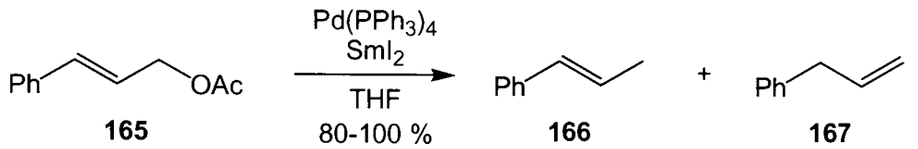


162 **163** **164**

The reduction of an allylic ester **165** has also been achieved in good yield using tetrakis(triphenylphosphine)palladium(0) and samarium iodide in catalytic amounts (Scheme 126).[156] Interestingly, the site of protonation was heavily influenced by the choice of protonating agent. The bulky *t*-butanol gave mostly γ-protonation resulting in

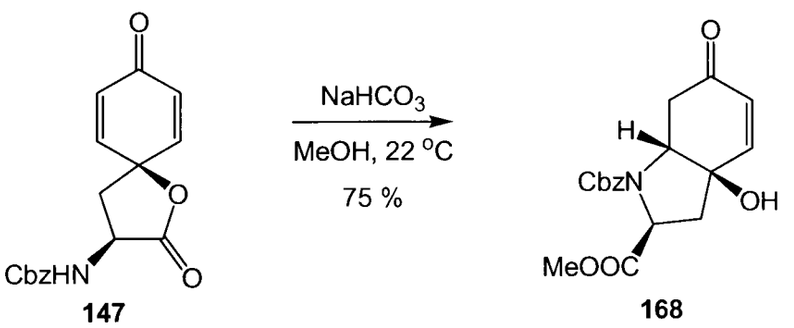
the α,β-enone **166**, while water gave mostly α-protonation resulting in the β,γ-enone **167**.

Scheme 126



As part of their work towards the total synthesis of *Stemona* alkaloids, Wipf and Kim treated a tyrosine spirolactone **147** (see Scheme 116 for preparation) with sodium bicarbonate in methanol (Scheme 127). The result was methanolysis of the lactone followed by Michael addition of the carbamate to the enone moiety. Bicycle **168** was the only diastereoisomer observed, indicating a high degree of stereocontrol during spirolactone formation.

Scheme 127



3.5.2 Application to the Product of the Nickel-Mediated Reaction

3.5.2.1 Introduction

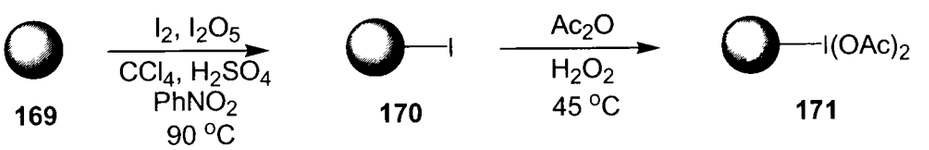
Spirolactone formation was first planned during work towards the nickel-mediated homocoupling of an iodotyrosine (Section 2.3.2). This was the reason for the double benzyl protection of the iodotyrosine starting material – one deprotection step would then provide the substrate for spirolactonisation.

The ideal reagent for facilitating spirolactone formation would be NBS since it provides almost quantitative yields in many cases, and is cheap and readily available. However as the dityrosine product of nickel-mediated coupling would have two unsubstituted positions *ortho* to the phenol group, these positions would be brominated by NBS. The used of PIDA or PIFA does not result in halogenation, but provides spirolactones in moderate yields at best.[146, 147] The use of PSDIB was considered, since Ley has reported good yields and high levels of product purity for the spirolactonisation of tyrosine derivatives.[148]

3.5.2.2 Synthesis of PSDIB

Poly(iodostyrene) **170** was prepared from commercially available polystyrene **169**, iodine, iodine pentoxide and sulfuric acid according to the method of Wang and Chen (Scheme 128).[157] Microanalysis indicated an iodine loading of 2.32 mmol/g. Poly(iodostyrene) **170** was then treated with peracetic acid, freshly prepared from acetic acid and hydrogen peroxide, to give PSDIB **171**. Microanalysis indicated a diacetoxyiodine loading of 1.85 mmol/g.

Scheme 128



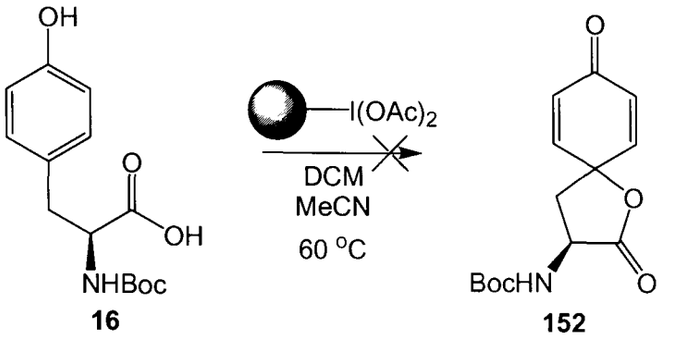
169 **170** **171**

3.5.2.3 Synthesis of a Tyrosine Spirolactone

In order to validate the chemistry, the formation of a tyrosine spirolactone was attempted before work on the spirolactonisation of tyrosine was considered.

N-Boc-L-tyrosine **16** was treated with PSDIB according to the method of Ley, but none of the desired spirolactone **152** was isolated (Scheme 129).[148] It is possible that the failure of the reaction originated from a lack of experience in working with solid-supported reagents, such as a problem with the preparation or isolation of the PSDIB, or separation of the spirolactone from the PSDIB.

Scheme 129



16 152

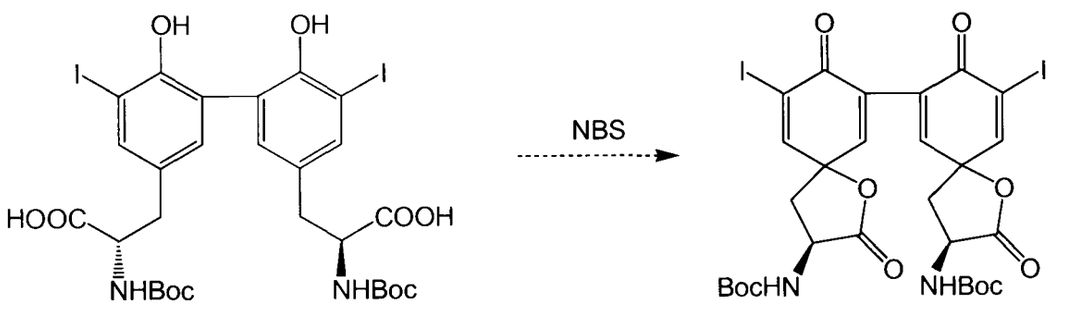
The reaction was not investigated further since the nickel-mediated coupling reaction failed to provide a good yield of the desired dityrosine.

3.5.3 Application to the Product of the Bowman Reaction

3.5.3.1 Introduction

With work on the formation of the dityrosine bond having moved on to the unusual coupling reaction reported by Bowman (Section 2.5) consideration was given to the formation of a dispirolactone from the diiododityrosine produced by the Bowman coupling reaction. Since the positions *ortho* to the phenol groups were all substituted, NBS could be used to affect spirolactonisation (Scheme 130).

Scheme 130



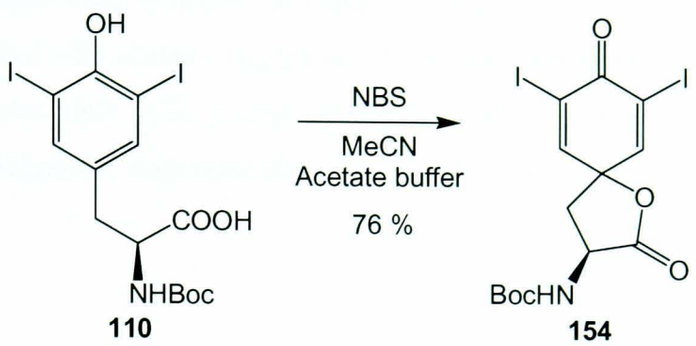
172 173

3.5.3.2 Synthesis of a Tyrosine Spirolactone

Again, in order to validate the chemistry, the formation of a tyrosine spirolactone was attempted before work on the spirolactonisation of tyrosine was considered.

N-Boc-3,5-Diiodo-L-tyrosine **110** (Section 2.5.6 for preparation) was treated with NBS in acetonitrile and pH 4.6 acetate buffer according to the method of Schmir (Scheme 131). The spirolactone **154** was isolated in 76 % yield.[149, 150]

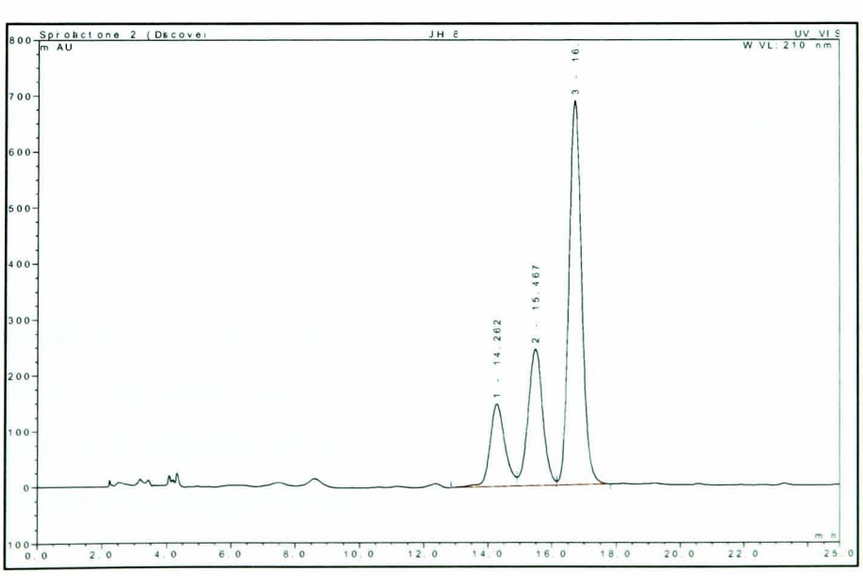
Scheme 131



110 **154**

The ^{1}H-NMR spectrum of the product confirmed spirolactone formation with the presence of two signals at 7.64 ppm and 7.31 ppm corresponding to the diastereotopic vinyl protons. The IR spectrum was also helpful, with three absorptions being observed in the carbonyl region (1803, 1694 and 1676 cm^{-1}). However, the ^{1}H-NMR spectrum indicated a mixture of at least two products, although this was not evident from tlc. Not surprisingly, the components could not be separated by flash chromatography; preparative reverse-phase hplc was used to purify the mixture, which consisted of one major product and two minor products (Figure 49).

Figure 49

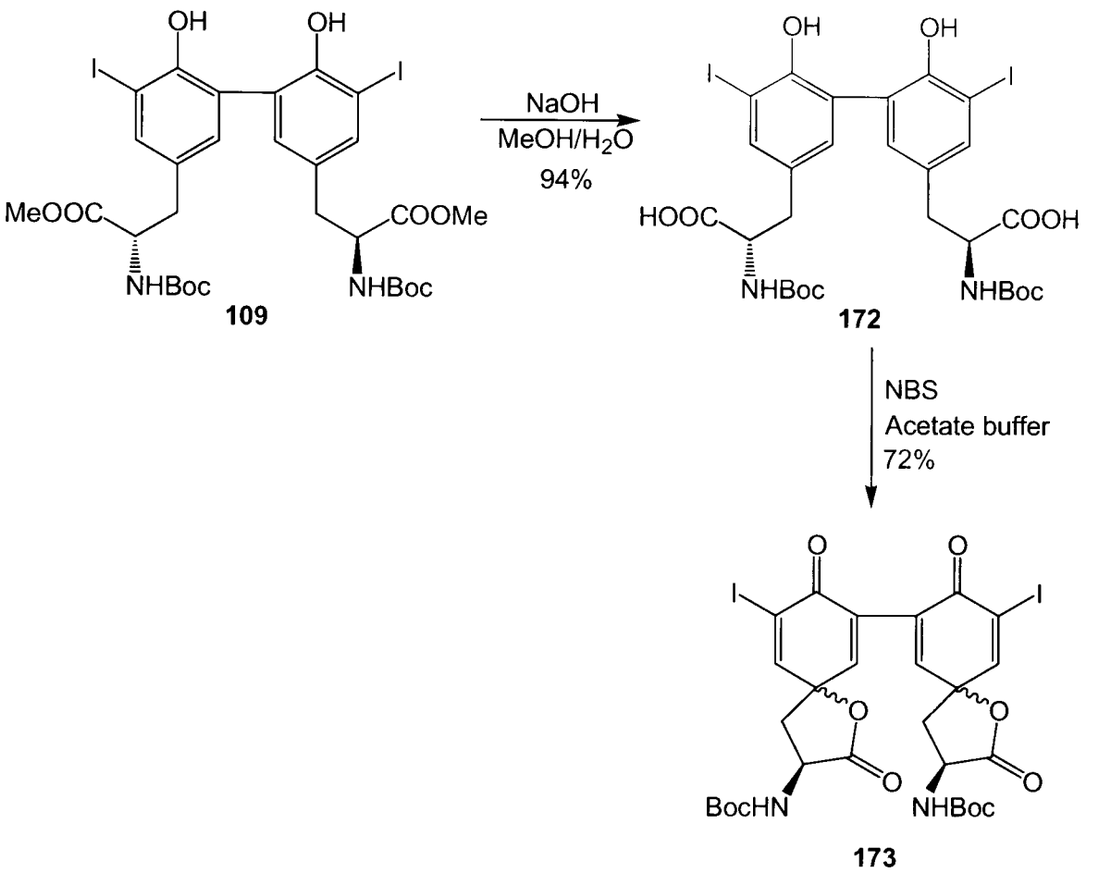


The major product (peak 3) was identified as the spirolactone **154**, and this was recovered in 51 %, resulting in a 39 % overall yield from tyrosine **110**.

3.5.3.3 Synthesis of the Dispirolactone

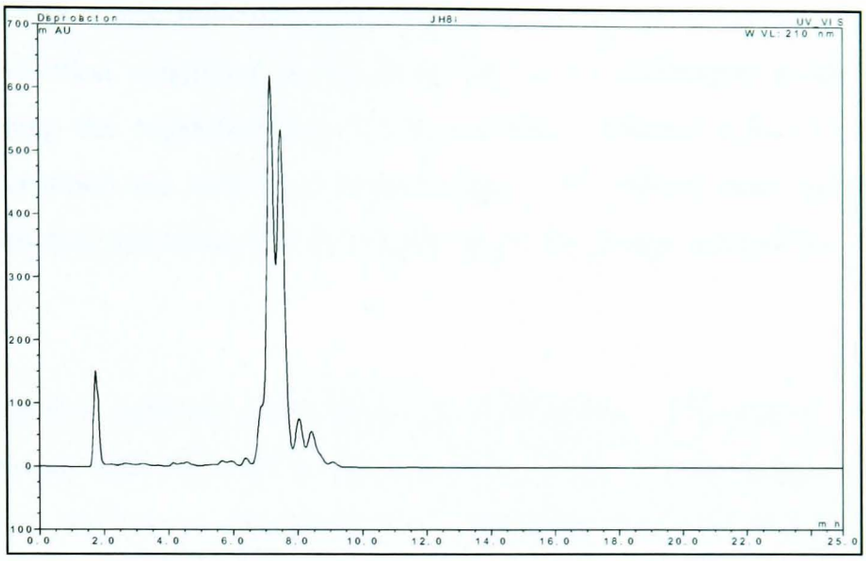
The diiododityrosine **109**, synthesised using the Bowman coupling reaction (Section 2.5.2-3), was treated with sodium hydroxide to give the Boc-protected dityrosine **172** in 94 % yield (Scheme 132). The double spirolactonisation reaction was then performed using NBS, generating the dispirolactone **173** in 72 % yield.

Scheme 132



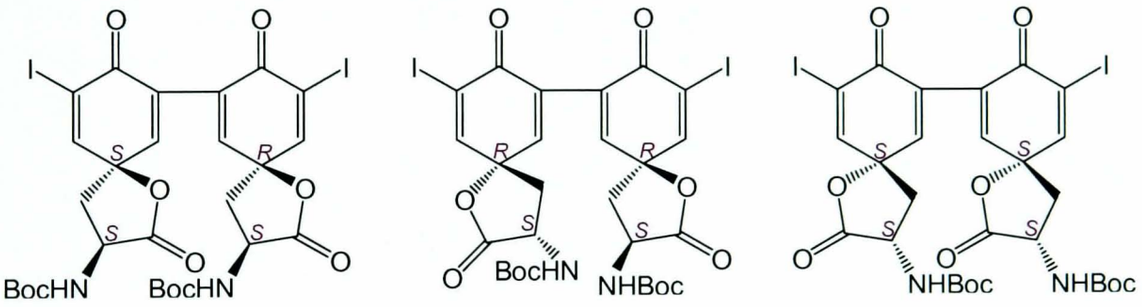
^{1}H-NMR spectroscopy of the crude product indicated that the product appeared to be a mixture of two compounds; none of the dityrosine remained, and all the signals were consistent with those expected of the dispirolactone **173**. Analytical hplc showed that the crude product was an approximate 1:1 mixture of two compounds.

Figure 50



Since ^{1}H-NMR spectroscopy indicated a mixture of two dispirolactones, the possible products of the reaction were considered. The symmetry of the starting material meant that there were three possible diastereoisomeric products of the dispirolactonisation reaction (Figure 51).

Figure 51



Although this reaction appeared promising, the routine purification of large amounts of material by hplc was not desirable. This fact, combined with the problems experienced when scaling-up the Bowman coupling reaction (Section 2.5.5), meant that the reduction of the dispirolactone was not pursued.

3.6 Conclusions

Several small molecules including a biphenyl system were subjected to Birch reduction conditions in order to gain experience with the technique.

Model biphenyl compounds bearing free hydroxyl groups were prepared and subjected to Birch reduction conditions in the hope that an intramolecular protonation would occur, altering the regiochemistry of the reaction. Unfortunately, all the isolated products contained one unreduced aromatic ring. An over-reduced cyclohexane was however isolated, indicating that the nearby hydroxyl groups altered the course of the reaction.

NBS was used to generate a diiodotyrosine spirolactone. Subsequent treatment of a diiododityrosine with NBS led to the formation of the corresponding dispirolactone, obtained as a mixture of diastereoisomers. However, the reaction pathway was not pursued.

Following these unsatisfactory attempts at reducing a biphenyl system with regiochemistry different from that provided by a Birch reduction, a change in strategy was sought.

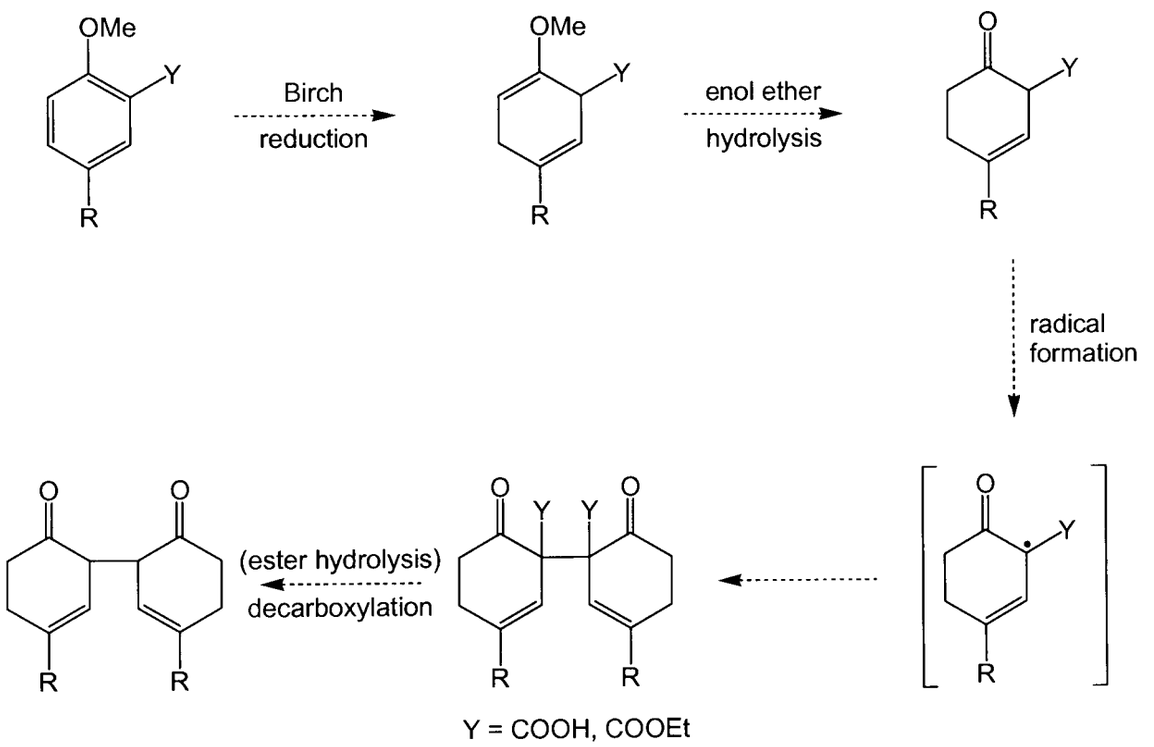
CHAPTER 4

SYNTHESIS & REACTIONS OF BIRCH-REDUCED TYROSINE SYSTEMS

4.1 Introduction

The Birch reduction of a tyrosine system and subsequent coupling was considered as an alternative to the reduction of a dityrosine system. This approach required the reduction of an aromatic system, followed by enol ether hydrolysis to generate a cyclohexenone. It was hoped that the introduction of a carboxylic acid or ester substituent adjacent to the carbonyl group would facilitate the generation of a radical, allowing subsequent homodimerisation of the reduced system (Scheme 133). The carboxylate group could then be removed, furnishing the required dienone system.

Scheme 133



This strategy involved the investigation of a number of different reactions: generation of a functionalised aromatic compound and its subsequent Birch reduction (see Section 4.2), enol ether hydrolysis (see Section 4.3) and radical coupling (see Section 4.4). Also considered was the synthesis of a fused-6,5-bicyclic system, analogous to that found in herquline A (see Section 4.5).

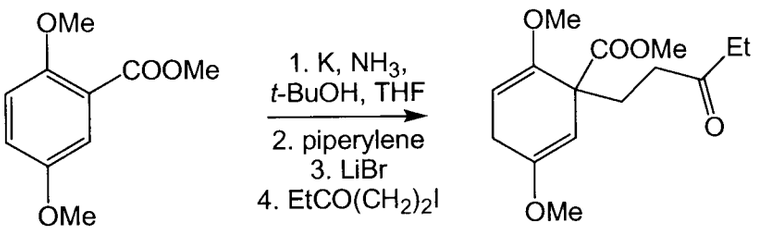
4.2 Birch Reduction of a Functionalised System

4.2.1 Introduction

The Birch reduction of aromatic carboxylic acids under standard conditions is straightforward. Carboxylic acids are not reduced, since the carboxylate anion is formed. Electrostatic repulsion then prevents the addition of an electron to the carboxylate system; reduction of the aromatic ring proceeds unhindered.

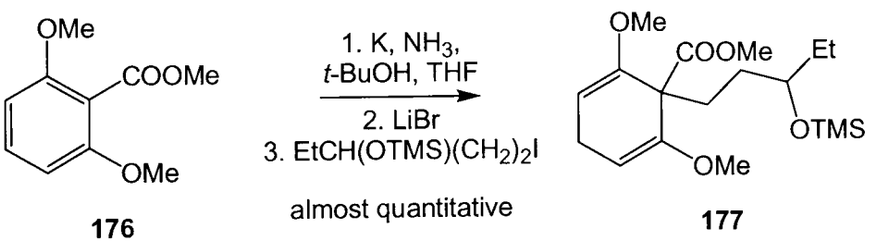
Esters are normally reduced to alcohols under Birch conditions.[122] However, aromatic carboxylic esters can be reduced to cyclohexadienes with retention of the ester group using conditions detailed by Mander.[158-160] The use of potassium instead of lithium, the addition of 1.5 equivalents of *t*-butanol and a reaction temperature of −78 °C have been shown to allow the survival of carboxylic esters during Birch reduction. Mander used these conditions to achieve the reductive alkylation of a range of benzoic esters (Scheme 134-6).

Scheme 134



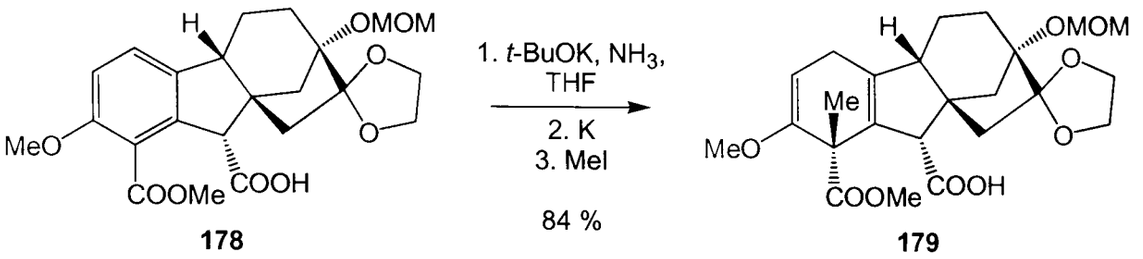
174 83 % **175**

Scheme 135



176 almost quantitative **177**

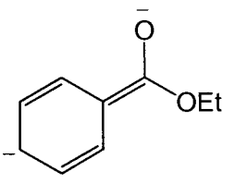
Scheme 136



178 179

The potassium *t*-butoxide formed neutralises any ammonium ions present (formed by deprotonation of acidic functional groups). Aromatic esters remain intact under these conditions since the addition of two electrons to the ester or the aromatic ring results in a dianion with a delocalised π-enolate system (Figure 52). This system is then preferentially protonated at the position *para* to the ester group. The enolate monoanion is not basic enough to be protonated by ammonia, and so exists until the reaction mixture is quenched.

Figure 52



Under normal Birch conditions, with an excess of an alcohol present, sequential electron-addition and protonation steps would result in the reduction of both the aromatic ring and the ester group.

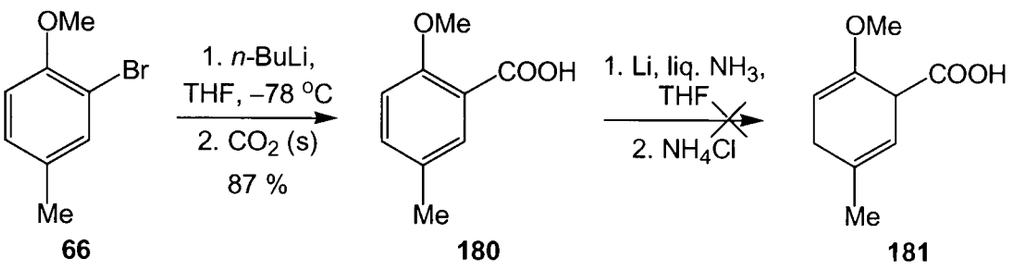
4.2.2 Synthesis and Birch Reduction of Aryl Carboxylic Acids

As discussed later (Section 4.4.1.1), there is good precedent for the coupling of carboxylic acids using a strong base and iodine; consequently, the use of carboxylic acids as a functionalising group was investigated. As with previous work, model compounds were investigated initially in order to avoid the complications of labile protons and protecting groups.

4.2.2.1 Application to a Model System

2-Bromo-4-methyl anisole **67** was treated with *n*-butyllithium in order to generate the aryllithium, which was then quenched by the addition of solid carbon dioxide to give the corresponding benzoic acid **180** in good yield (Scheme 137).[161, 162] The benzoic acid **180** was then treated with lithium in liquid ammonia; as discussed previously (Section 3.2.2), no alcohol was required.[163] However, ^{1}H-NMR spectroscopy indicated that the crude product* was a complex mixture, containing some of the benzoic acid **180** and several olefinic products which had undergone enol ether hydrolysis.

Scheme 137

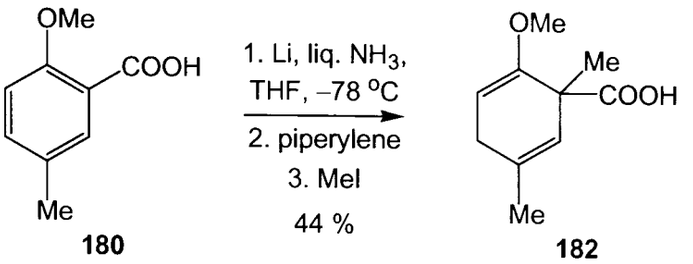


In an attempt to make the cyclohexadiene easier to handle, a Birch reduction-alkylation reaction was performed. The method involved a standard Birch reduction in which the ammonium chloride quench was replaced with piperylene to react with excess electrons,† and methyl iodide to quench the surviving cyclohexadienyl anion.[126] ^{1}H-NMR spectroscopy of the crude product showed signals at 5.12 and 4.87 ppm corresponding to olefinic protons, and at 1.64 and 1.24 ppm corresponding to two methyl groups, confirming the formation of the expected cyclohexadiene **182** (Scheme 138). Without the addition of piperylene, the crude product of the reaction was a complex mixture and the individual products could not be identified.

* Cyclohexadiene enol ethers such as **182** were found to be unstable, particularly to flash chromatography. Consequently, many of the cyclohexadienes synthesised were not purified and had to be stored at –18 °C.

† Piperylene is a mixture of 1,3-pentadienes. Excess electrons are readily added to conjugated dienes. Piperylene is used to prevent any reaction between the electrons and the alkylating agent used to quench the anion.

Scheme 138



180 **182**

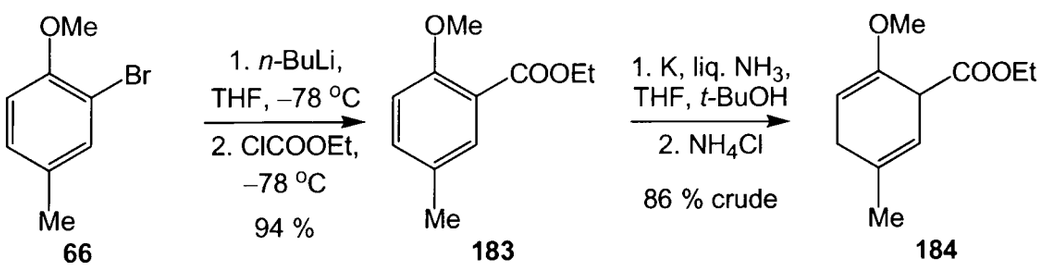
4.2.3 Synthesis and Birch Reduction of Aryl Esters

4.2.3.1 Application to a Model System

Following the unsuccessful attempt at Birch reduction of a carboxylic acid, use of an ester group was investigated. It was hoped that using an ester rather than a carboxylic acid would facilitate handling and purification of the products.

The aryl bromide **66** was again treated with *n*-butyllithium; the resulting organolithium was inversely quenched with excess ethyl chloroformate at –78 °C in order to avoid the formation of a diarylketone. This afforded the desired benzoic ester **183** in excellent yield (Scheme 139).[164] The aryl ester **183** was then treated with potassium and *t*-butanol in liquid ammonia, and the crude cyclohexadiene was obtained in good yield. Purification by flash chromatography was not attempted since the product appeared to decompose when analysed by tlc. Formation of the cyclohexadiene **184** was confirmed by loss of significant peaks from the aromatic region of the ^{1}H-NMR spectrum, and by the presence of signals at 5.37 and 4.82 ppm corresponding to the olefinic protons.

Scheme 139

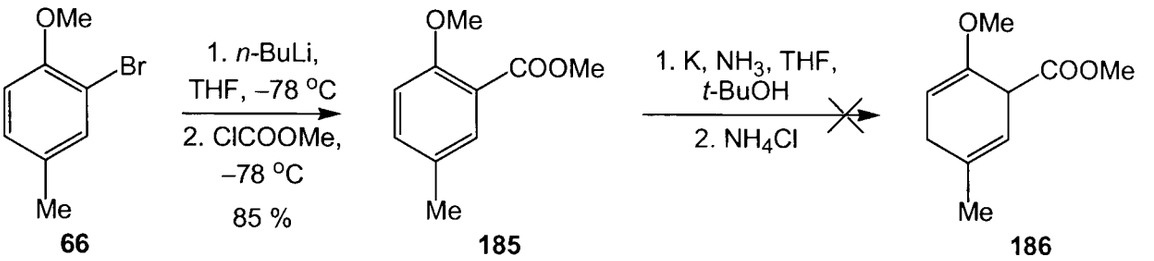


66 **183** **184**

The synthesis and reduction of the corresponding methyl ester was attempted (Scheme 140). The methyl benzoate **185** was synthesised from the aryl bromide **66** using conditions analogous to those described previously. However, when the ester was

treated with potassium and *t*-butanol in liquid ammonia, although ^{1}H-NMR spectroscopy indicated that some reduction of the aromatic ring had occurred, the spectrum was extremely complex; the crude product appeared to contain a number of compounds which could not be identified.

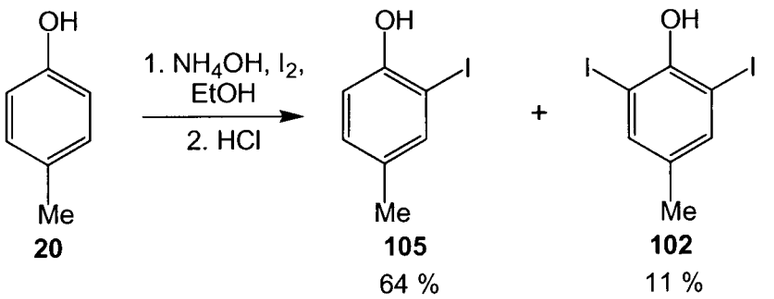
Scheme 140



Problems experienced during attempts to hydrolyse the methyl enol ether **184** (see Section 4.3.2) led to investigation of the TBDMS protecting group, which is stable to Birch conditions and can be readily removed with fluoride.[73]

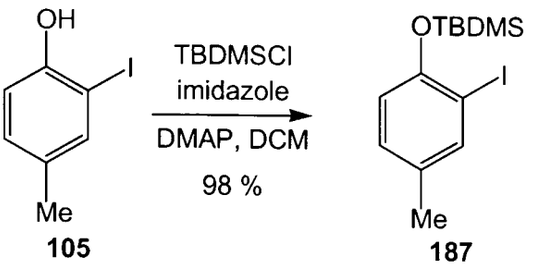
In order to generate the corresponding functionalised aryl siloxanes, *p*-cresol **20** was treated with ammonium hydroxide and iodine to give the iodocresol **105** in 64 % yield (Scheme 141).[103] Also isolated was a small amount of the diiodocresol **102**.

Scheme 141



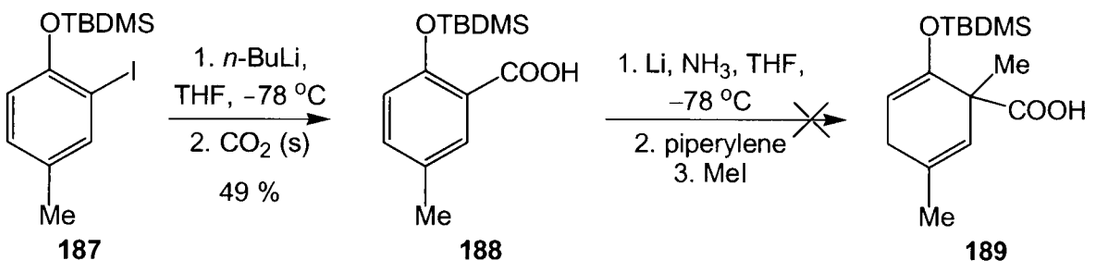
The iodocresol **105** was then treated with TBDMS chloride and imidazole to give the protected cresol **187** in almost quantitative yield (Scheme 142).

Scheme 142



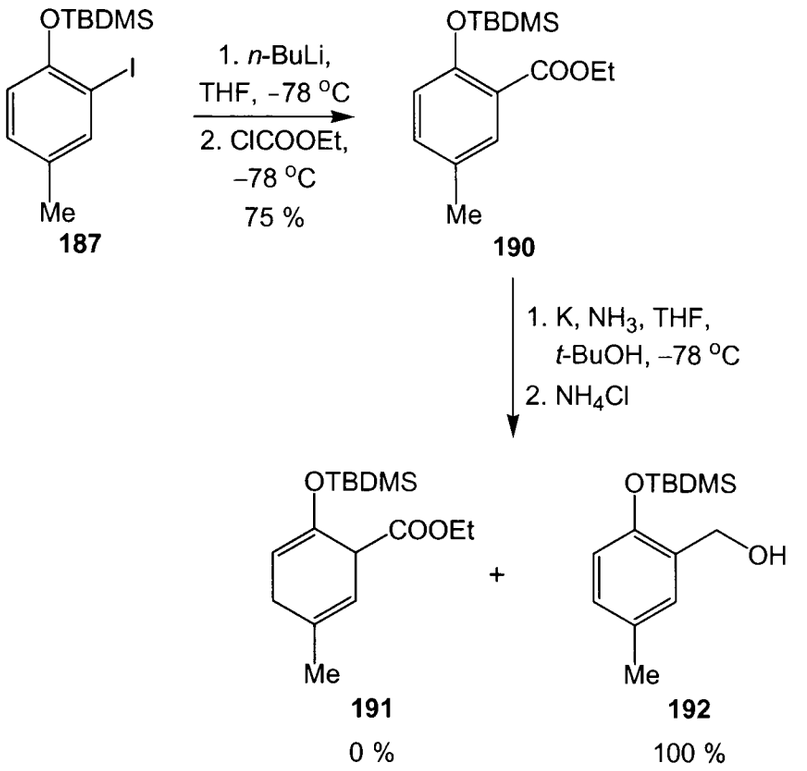
In reactions analogous to those detailed previously, the aryl iodide **187** was treated with *n*-butyllithium and solid carbon dioxide to give the benzoic acid **188** (Scheme 143).[161, 162] The acid was then treated with the same Birch reduction-alkylation sequence that was used to treat the methyl-protected cresol (refer to Scheme 138). However, no reduction of the aromatic ring was observed and the starting material was recovered along with the corresponding methyl ester.

Scheme 143



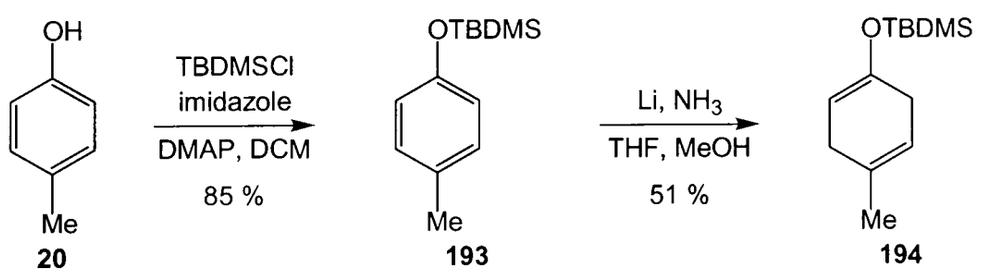
Treatment of the aryl iodide **187** with *n*-butyllithium and ethyl chloroformate resulted in the synthesis of the aryl ester **190** in 75 % yield (Scheme 144).[164] However, when the ester was treated with potassium and *t*-butanol in liquid ammonia, no reduction of the aromatic ring was observed. The ^{1}H-NMR spectrum of the crude product contained no signals corresponding to the ethyl group, indicating that the ester group had been reduced to give the benzylic alcohol **192** in quantitative yield.

Scheme 144



The results of these reactions implied that the protecting group was hindering the Birch reduction; presumably the sterically demanding TBDMS group was preventing protonation of the dianion species. Consequently a less substituted system was chosen in order to investigate the general applicability of TBDMS-protected phenols to Birch reductions. Therefore *p*-cresol **20** was treated with TBDMS chloride and imidazole to give the protected cresol **193** in good yield (Scheme 145). Treatment of the TBDMS-protected cresol **193** with lithium in liquid ammonia in the presence of methanol resulted in isolation of the expected cyclohexadiene **194** in moderate yield.

Scheme 145

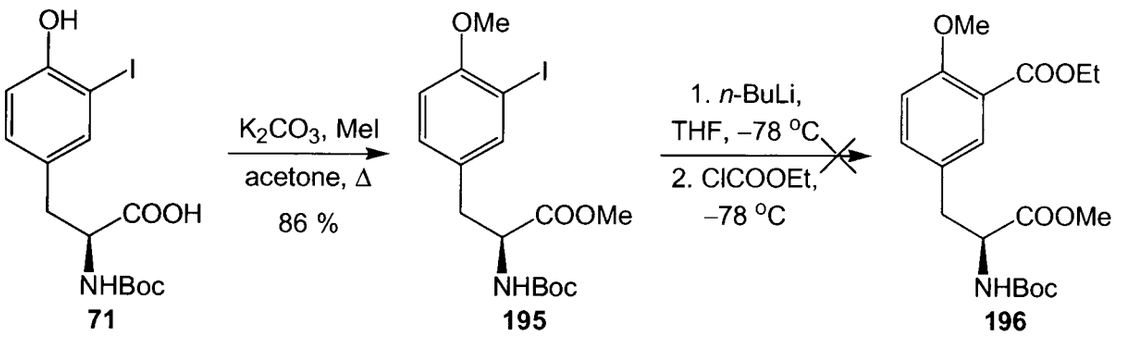


This reaction confirmed that the TBDMS protecting group is compatible with the Birch reduction but, perhaps because of its steric bulk, the group appeared to hinder the reduction of aromatic rings with substituents *ortho* to the silyloxy group.

4.2.3.2 Application to a Tyrosine System

Having achieved the reduction of the ethyl ester **183**, the reduction conditions were applied to tyrosine. The phenol hydroxyl group had to be protected since phenols do not undergo Birch reduction. It was decided that protection of the carboxylic acid as an ester would be beneficial. This would result in reduction of the carboxylate group during the Birch reduction, thus reducing the acidity of the proton adjacent to the carbamate group. Having an acidic proton at this position could potentially cause problems during the coupling step since some of the radical-generation techniques involve the use of a strong base. *N*-Boc-iodo-L-tyrosine **71** (see Section 2.3.3.4 for preparation) was therefore treated with potassium carbonate and methyl iodide to give the fully protected molecule **195** in 86 % yield (Scheme 146).

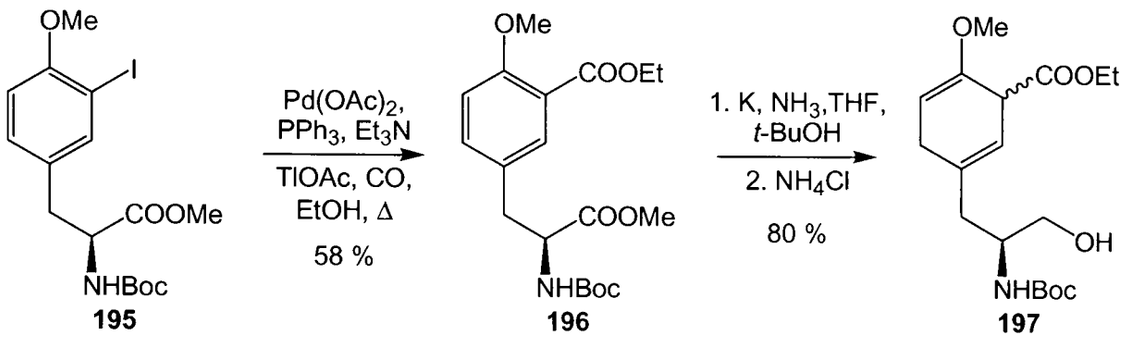
Scheme 146



Ethoxycarbonylation of iodotyrosine **195** with *n*-butyllithium and ethyl chloroformate was not successful, presumably due to the presence of the slightly acidic carbamate NH proton. Consequently, the iodotyrosine **195** was treated with palladium(II) acetate, triphenylphosphine, triethylamine, carbon monoxide and ethanol in order to affect a palladium-catalysed carbonylation reaction, quenched *in-situ* with ethanol.[165, 166] The corresponding ester **196** was produced in only 28 % yield. The addition of a stoichiometric amount of thallium(I) acetate as described by Grigg resulted in an improved yield of 58 % (Scheme 147).[167, 168]

Reduction of the diester **196** with potassium and *t*-butanol in liquid ammonia led to the formation of the cyclohexadiene **197**. The structure was confirmed by ^{1}H-NMR spectroscopy: signals at 5.47 and 4.80 ppm confirmed reduction of the aromatic ring, the absence of one singlet from around 3.6 ppm confirmed reduction of the aliphatic ester and the presence of signals corresponding to the ethyl fragment confirmed that the ethyl ester had survived the Birch reduction.

Scheme 147

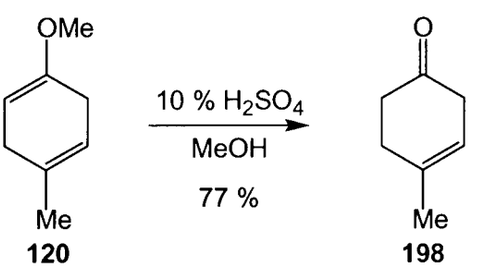


4.3 Enol Ether Hydrolysis

4.3.1 Introduction

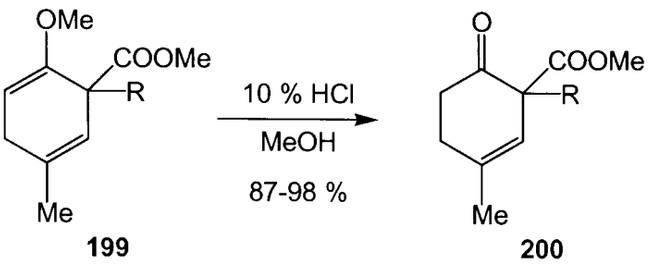
The hydrolysis of methyl enol ethers is reportedly a straightforward reaction, often performed using a dilute mineral acid.[169] Corey reported the hydrolysis of 1-methoxy-4-methylcyclohexa-1,4-diene **120** in 77 % yield by shaking a solution of the substrate with dilute sulphuric acid.[170]

Scheme 148



A similar hydrolysis reaction performed using dilute hydrochloric acid has been reported by Schultz (Scheme 149).

Scheme 149



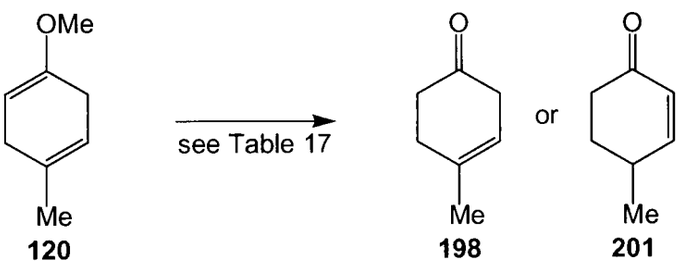
199 **200**

a R = Me
b R = Et
c R = $(CH_2)_3Cl$

4.3.2 Application to a Model System

Hydrolysis of the model enol ether **120** (see Section 3.2.5.2 for preparation) was initially attempted using dilute sulfuric acid (Scheme 150).[170] Unfortunately, this method failed to affect hydrolysis of the enol ether moiety, and led instead to a complex mixture of products. A range of reagents was therefore screened in an attempt to generate the desired enone **198** (Scheme 150, Table 17).*

Scheme 150



120 **198** **201**

* When present, the desired product could be easily identified using ^{1}H-NMR spectroscopy by comparison with data reported by Corey. The α,β-enone showed two peaks in the olefinic region rather than the single signal arising from the β,γ-enone. For the purposes of radical generation, a mixture of α,β- and β,γ-enones would be acceptable since they would be in equilibrium. However, a mixture of enones resulted in more complex NMR spectra, making it more difficult to determine levels of purity and to monitor subsequent reactions.

Table 17 Reagents tested for enol ether hydrolysis

Entry	Acid	Solvent	Time (h)	Result
1	Dilute H_2SO_4	MeOH	2	Complex mixture. No product
2	Dilute H_2SO_4	Et_2O	wash	Complex mixture. Some re-aromatisation.
3	Aqueous citric acid	Et_2O	wash	Starting material only.
4	Silica	MeOH	96	Starting material only.
5	Phosphate buffer/H_3PO_4 (pH 2)	EtOAc	3	Mixture of α,β- & β,γ-enones.
6	Phosphate buffer/H_3PO_4 (pH 2)	MeOH	2	Mixture of α,β- & β,γ-enones. Low yield.
7	Phosphate buffer/H_3PO_4 (pH 2)	Et_2O	72	β,γ-Enone and re-aromatisation only.
8	TMSCl, NaI, 60 °C; NaCl	MeCN	1.5	Starting material and re-aromatisation.
9	TMSCl, NaI, 60 °C; TBAF	MeCN	1.5	Complex mixture. Some re-aromatisation.
10	TMSCl, NaI, 60 °C; K_2CO_3	MeCN	1.5	Starting material and re-aromatisation.
11	TMSCl, NaI, 60 °C; acetic acid	MeCN	1.5	Unable to isolate material.
12	Boron tribromide, –78 °C	DCM	1	Complex mixture.
13	Aqueous oxalic acid	MeOH	0.5	Pure β,γ-enone.

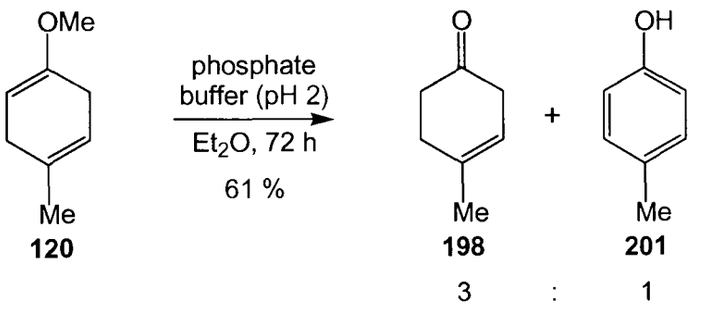
Following the unsatisfactory results of the sulfuric acid reactions (Table 17, entries 1 and 2), a milder acid was sought.

Enol ether hydrolysis using citric acid was attempted (Table 17, entry 3), but no reaction occurred. Since the hydrolysis of similar compounds had previously been observed during flash chromatography (Section 3.4.2.3), the cyclohexadiene **120** was stirred with flash silica (Table 17, entry 4), but this method also failed to accomplish hydrolysis of the enol ether moiety.

In an attempt to generate a moderately acidic system, aqueous phosphate buffer was adjusted to pH 2 using concentrated phosphoric acid (Table 17, entries 5-7). Treatment

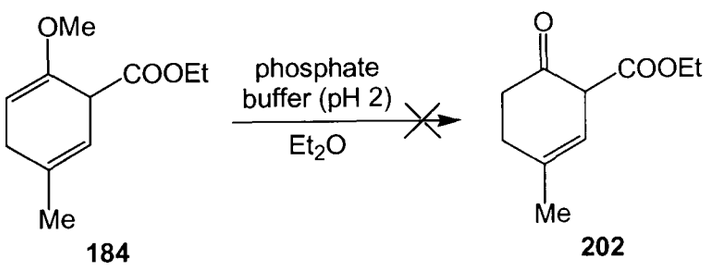
of a solution of the cyclohexadiene **120** with the buffer led to the formation of the expected enones **198** and **201**. The choice of solvent had a significant effect on the reaction, with the ether system producing the best result (Scheme 151). This system did however require a longer reaction time, presumably because of the reduced interaction between the substrate and the acid in the biphasic system.

Scheme 151



The next step was to hydrolyse the ester-substituted enol ether using these conditions. However, when the ester **184** was treated with a biphasic mixture of pH 2 phosphate buffer and ether, no enone formation was observed and only the starting material was recovered (Scheme 152).

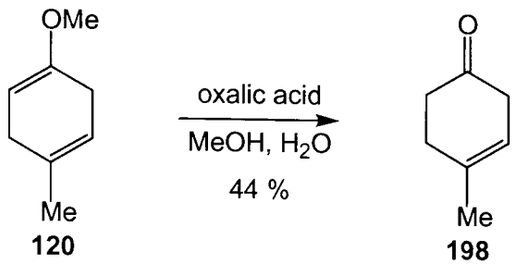
Scheme 152



A range of further reagents including *in-situ* formation of TMS iodide followed by a range of quenches,[171, 172] boron tribromide[173] and oxalic acid[174] (Table 17, entries 8-13) were then tested on the cyclohexadiene **120** used previously.

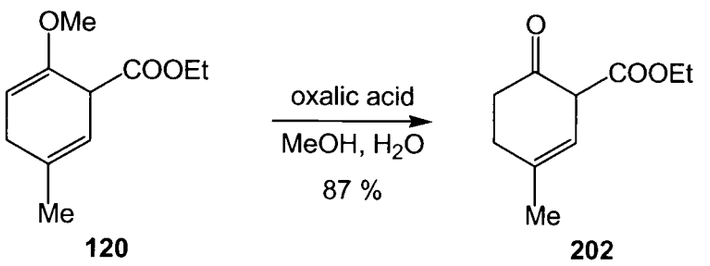
These experiments provided only one set of successful conditions – aqueous oxalic acid (Scheme 153).

Scheme 153



The ester **184** was then subjected to the same conditions, and the corresponding β,γ-enone **202** was generated in good yield (Scheme 154).*

Scheme 154



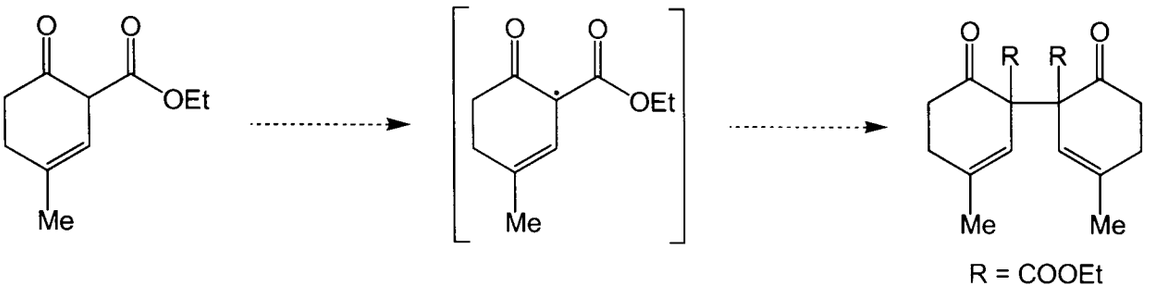
4.4 Radical Coupling

4.4.1 Introduction

Following the synthesis of the unsaturated β-ketoester **202**, it was hoped that a radical could be generated at the α-position. Homocoupling of the radical species would then result in the formation of the type of bicyclic system found in herqulines A and B.

* The yield given for formation of the β-ketoester **202** is a crude yield since the compound was unstable to flash chromatography.

Scheme 155

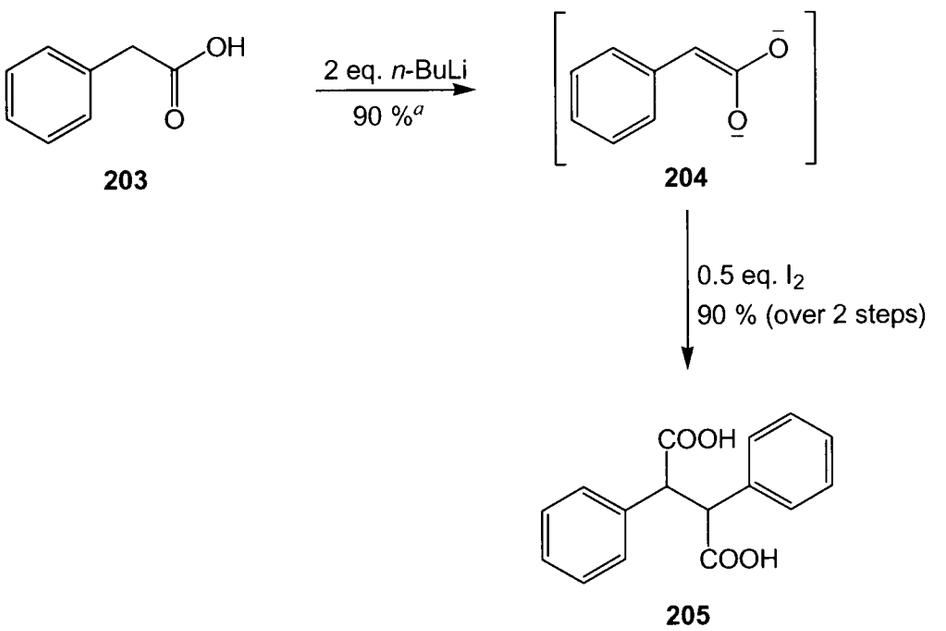


4.4.1.1 Iodine as a Radical Initiator

Introduction

An iodine-mediated oxidative coupling reaction has been reported by Belletire (Scheme 156).[175] It was noted that generation of the enolate **204** was the key step in the reaction and appeared to proceed differently for different carboxylic acids; consequently the authors recommended optimisation of the deprotonation step using a simple alkylation reaction as a model for the coupling reaction.[176]

Scheme 156

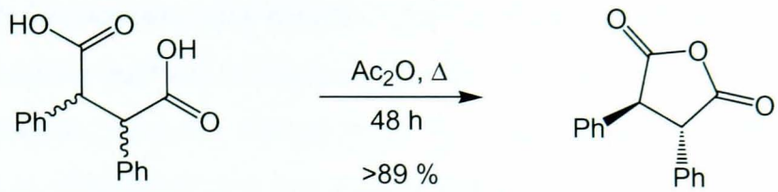


203 **204** **205**

[a] Yield estimated by trapping with BnBr

A preference for formation of the racemic mixture of *anti* diastereoisomers rather than the meso *syn* product has also been reported. The mixture of products **205** could be converted to the racemic anhydride **206** using acetic anyhdride (Scheme 157).[177]

Scheme 157



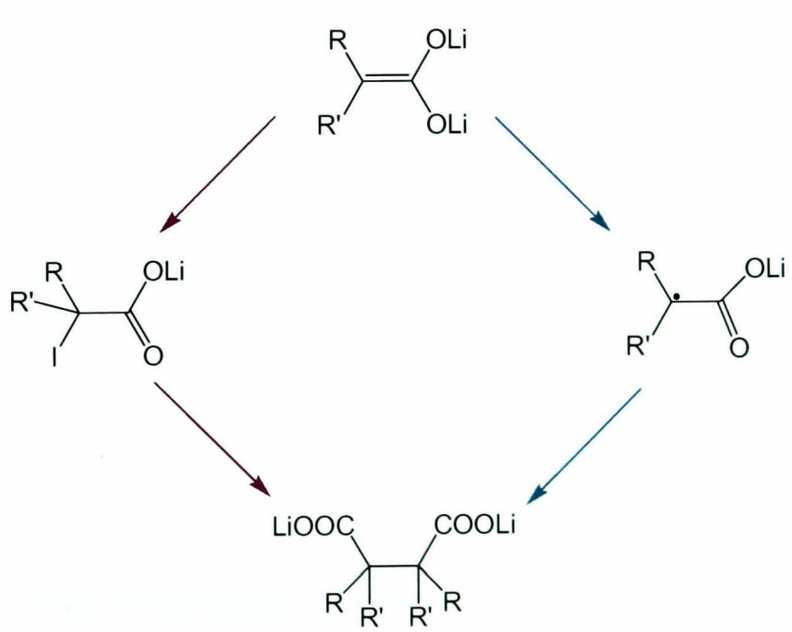
205 **(±)-206**

The choice of base in the reaction is significant – results have shown that the presence of amines (including diisopropylamine formed by protonation of LDA) adversely affected the coupling reaction. This was thought to be the result of the formation of a charge-transfer complex between diisopropylamine and iodine.* This undesirable side-reaction could be avoided either by isolating the dianion salt[175] or using a base such as lithium HMDS whose conjugate acid has a reduced tendency to form the charge-transfer complex.[178]

The Mechanism of the Reaction

Belletire *et al.* suggested that this type of reaction proceeds *via* an intermediate alkyl iodide, which then undergoes nucleophilic substitution (Scheme 158, shown in purple). However the group also considered the possibility of an electron transfer process, resulting in the formation and subsequent dimerisation of radical anions (Scheme 158, shown in green).

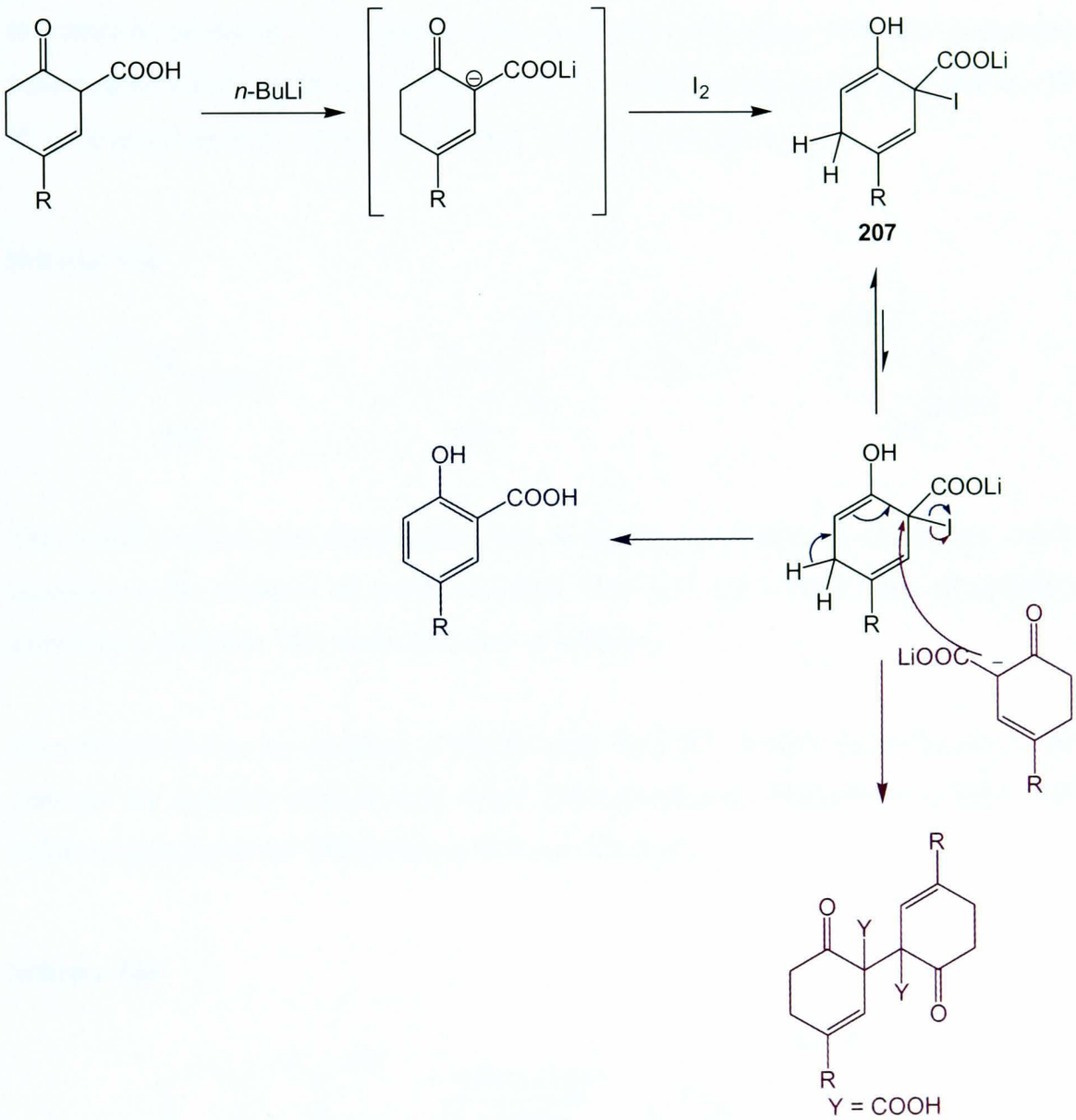
Scheme 158 Possible mechanisms of iodine-mediated coupling



* A deep green colour is observed on formation of the complex in which an electron is transferred from diisopropylamine to iodine. The complex then decomposes, liberating protons.

Since the reaction has been performed on trisubstituted systems (R,R' ≠ H) the electron transfer mechanism appears more feasible – S_N2 reactions are highly unlikely at tertiary centres. The precise pathway is important for the coupling of unsaturated β-ketoesters under consideration, since the formation of an intermediate iodide **207** would almost certainly lead to elimination and rearomatisation (Scheme 159, shown in blue) rather than the nucleophilic substitution reaction (Scheme 159, shown in purple).

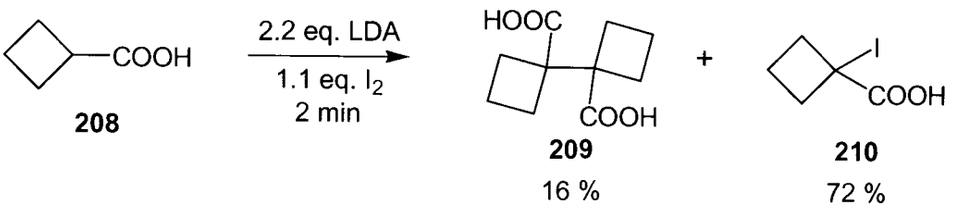
Scheme 159 Impact of an intermediate alkyl halide on coupling of an unsaturated β-ketoester



A mechanistic study of the radical coupling of cyclobutanecarboxylic acid **208** under similar conditions has been reported by Renaud and Fox.[178] The treatment of

cyclobutanecarboxylic acid with LDA and iodine at −78 °C gave 16 % of the expected dimer **209** in 2 minutes (Scheme 160).

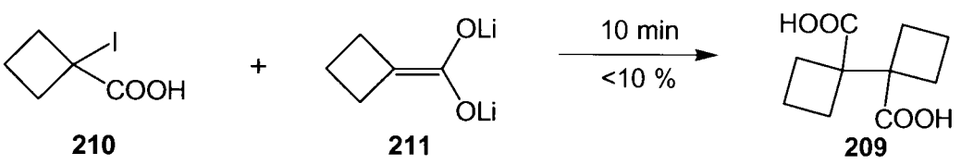
Scheme 160



208 **209** 16 % **210** 72 %

In contrast, treatment of lithium 1-iodocyclobutanecarboxylate **210** (the postulated intermediate iodide) with the di-lithium salt of cyclobutanecarboxylic acid **211** at −78 °C gave less than 10 % of the dimer **209** in 10 minutes (Scheme 161).

Scheme 161

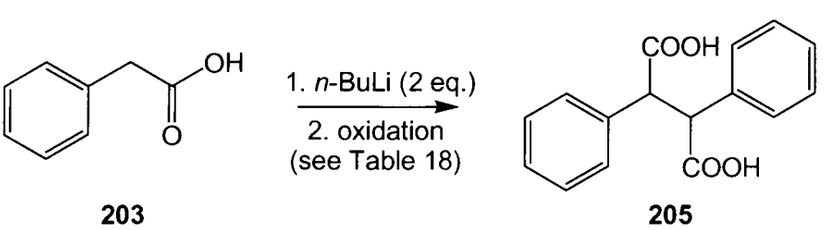


210 **211** **209**

This result implied that dimerisation was occurring *via* a radical mechanism, since coupling in the presence of iodine (Scheme 160) was much faster than nucleophilic substitution (Scheme 161) under the same conditions.

Also examined was the coupling of phenylacetic acid **203**. Following generation of the dianion, the reaction mixture was either electrochemically oxidised or treated with iodine to give the dimer **205** (Scheme 162 and Table 18).

Scheme 162



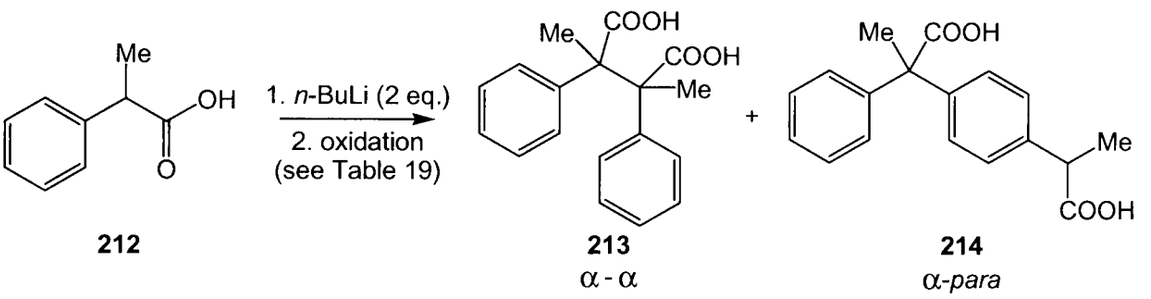
203 **205**

Table 18 Effect of oxidant on dimerisation of phenylacetic acid **203**

Oxidant	Yield of **205** (%)	*dl*/meso
Electrochemical	30-40	2:1
Iodine	90	11:1

However, electrochemical oxidation and treatment with iodine gave significantly different yields and selectivities. The authors attributed the low yield of the electrochemical oxidation to a competing process in which hydrogen was abstracted from the solvent (THF). In an attempt to clarify the situation, a more hindered substrate was chosen in order to limit the hydrogen abstraction pathway (Scheme 163 and Table 19). This resulted in the formation not only of the expected α,α-dimer **213**, but the α-*para* coupled product **214** as well.

Scheme 163



212

213 α - α

214 α-*para*

Table 19 Effect of oxidant on dimerisation of methylphenylacetic acid

Method	Oxidant	α-α dimer **213** (%)	α-*para* dimer **214** (%)
a	Electrochemical	16	40
b	Iodine	23	38

In this case, electrochemical oxidation and treatment with iodine resulted in similar yields and product distributions, providing more weight to the proposed radical mechanism. The observation of α-*para* coupling further implied a radical mechanism, since oxidative phenol coupling (which is known to often involve radical species) frequently gives rise to products of this type (Section 2.2).

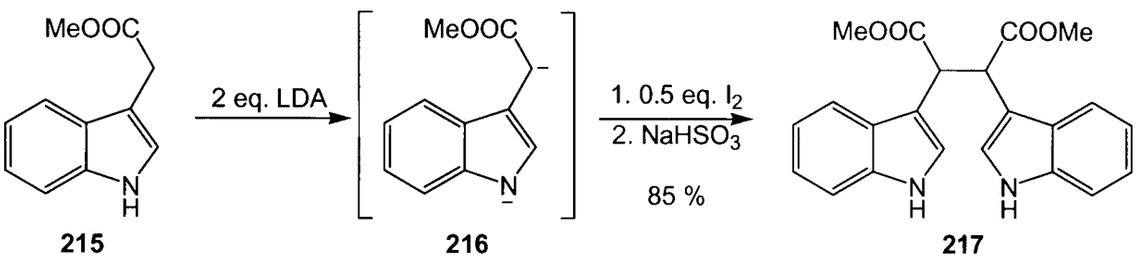
In summary, Renaud and Fox have shown distinct similarities between the results of electrochemical and iodine-mediated oxidative couplings. The observation of α-*para*

coupling also pointed to a radical pathway. No evidence was found for the involvement of intermediate iodides, and an attempt at dimer formation involving the postulated intermediate iodide was unsuccessful.

Application of Iodine-Mediated Coupling to Esters

An analogous reaction involving the radical coupling of carboxylic esters has been described as part of the synthesis of an indolocarbazole alkaloid (Scheme 164). A previous attempt to couple the corresponding carboxylic acids had proceeded in poor yield (<38 %).[179]

Scheme 164

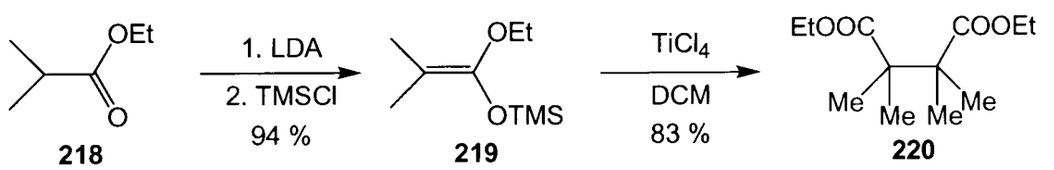


4.4.1.2 Other Coupling Techniques

A number of other methods have been used to facilitate the coupling of carboxylic esters, presumably also *via* generation of a radical at the position adjacent to the carbonyl group.

1) A dimerisation reaction resulting in the formation of quaternary centres has been performed by treating the silyl enol ether **219** of an ester with titanium tetrachloride (Scheme 165).[180]

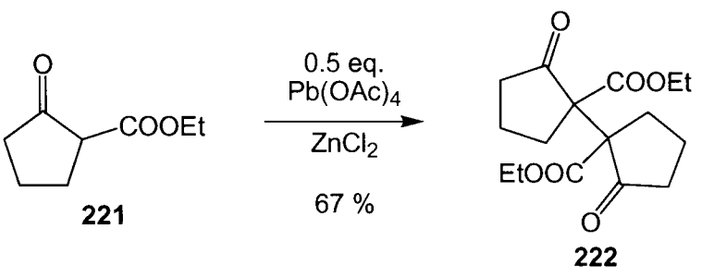
Scheme 165



2) Lead tetraacetate and zinc chloride have been used to facilitate the coupling of a cyclic β-ketoester **221** (Scheme 166).[181] The product **222** was obtained as a

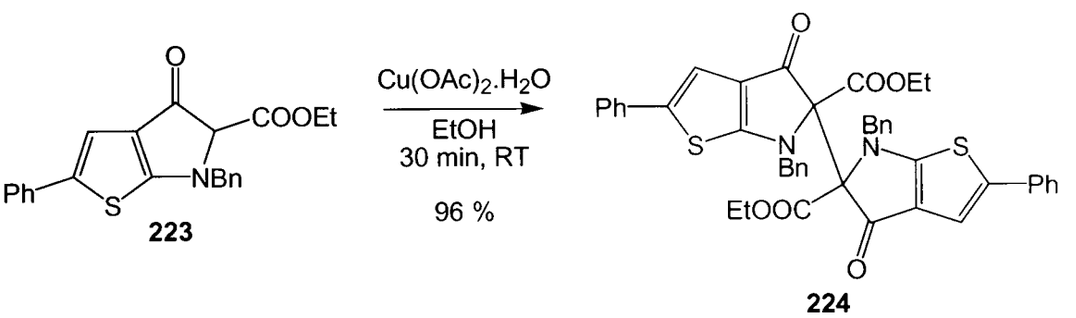
single diastereoisomer (although the isomer was not identified), thought to arise from a radical coupling of two molecules complexed to a zinc ion.

Scheme 166



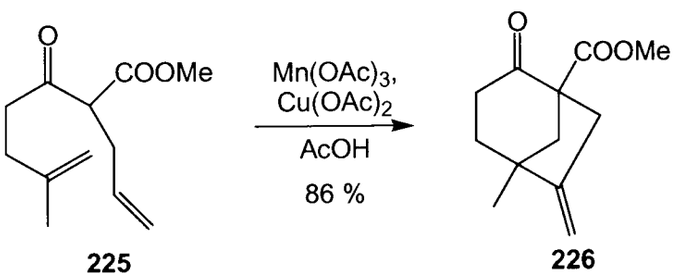
3) Another example of β-ketoester coupling, using the single electron oxidant copper(II) acetate, has been reported by Lee *et al.* (Scheme 167).[182]

Scheme 167



4) A combination of copper(II) acetate and manganese(III) acetate has also been used to generate radicals at the α-carbon of β-ketoesters in order to affect radical cyclisation reactions.[183]

Scheme 168

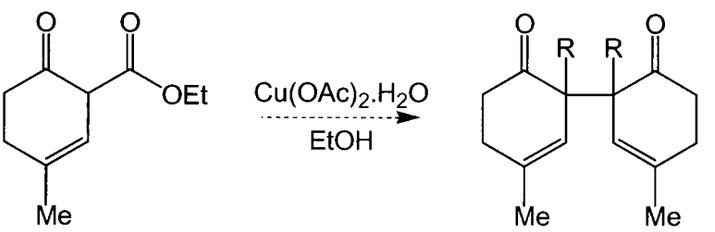


4.4.2 Application to a Model System

Having generated the model β-ketoester **202** (Section 4.3.2), radical coupling facilitated by copper(II) acetate was attempted. As the β-ketoester **202** was found to be unstable to flash chromatography, a small amount was purified by reduced pressure distillation. Unfortunately, this process resulted in some rearomatisation, but ^{1}H-NMR spectroscopy allowed easy identification of the β-ketoester and the benzoic ester. The signals were well resolved and sufficiently separated to allow monitoring of the dimerisation reaction under consideration.

The radical coupling reaction was attempted using copper(II) acetate (Scheme 169) according to the method of Lee described above (Section 4.4.1.2).

Scheme 169



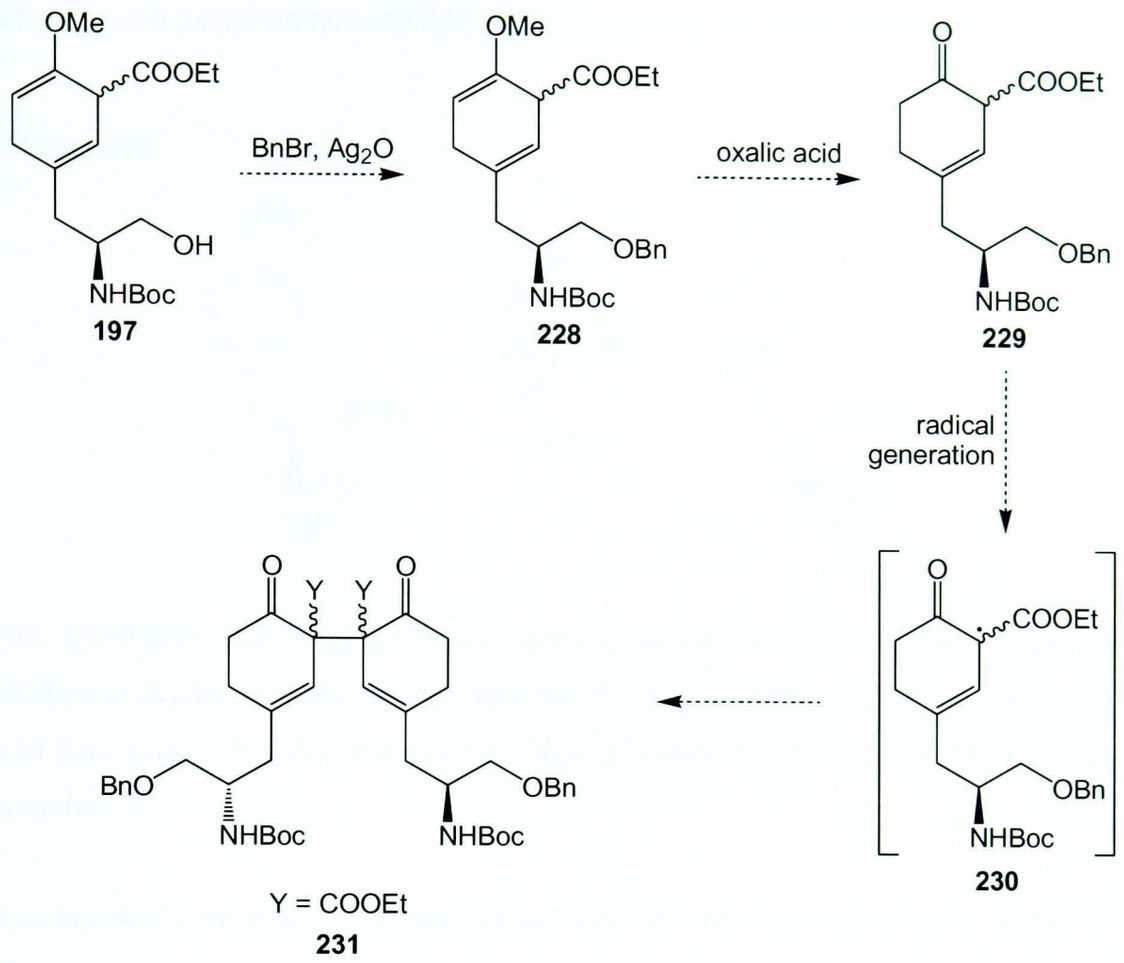
202 R = COOEt **227**

It was encouraging to find that, although the ^{1}H-NMR spectrum of the crude product indicated that some rearomatisation had occurred, some olefinic material remained. The absence of the singlet at 3.95 ppm corresponding to the α-proton indicated that some of the dimer **227** may have been generated. Unfortunately the molecular ion was not observed following analysis by mass spectrometry, which would have been helpful in confirming the formation of the dimer **227**.

4.4.3 Application to the Synthesis of the Herqulines

Unfortunately, time constraints prevented the further development of the radical coupling reaction and its application to a tyrosine system. Benzyl protection and enol ether hydrolysis of the Birch-reduced tyrosine **197** (see Section 4.2.3.2 for preparation) would have generated the substrate required in order to attempt the radical coupling reaction on a tyrosine-based system (Scheme 170).

Scheme 170

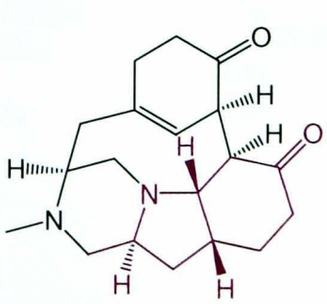


4.5 Synthesis of the 6,5-Fused Ring System

4.5.1 Introduction

Since progress towards the synthesis of herqulines A and B had been slow, it was decided to investigate formation of the 6,5-ring system of herquline A **1** (Figure 53), including any stereochemical considerations arising from the formation of such a bicyclic system.

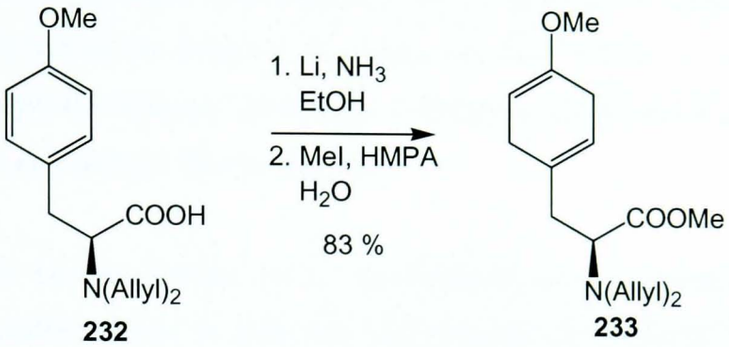
Figure 53



1

Two Birch reductions of tyrosine systems have been reported in the literature. The reduction of a protected tyrosine **232** has been reported by Ganem (Scheme 171).[184]

Scheme 171

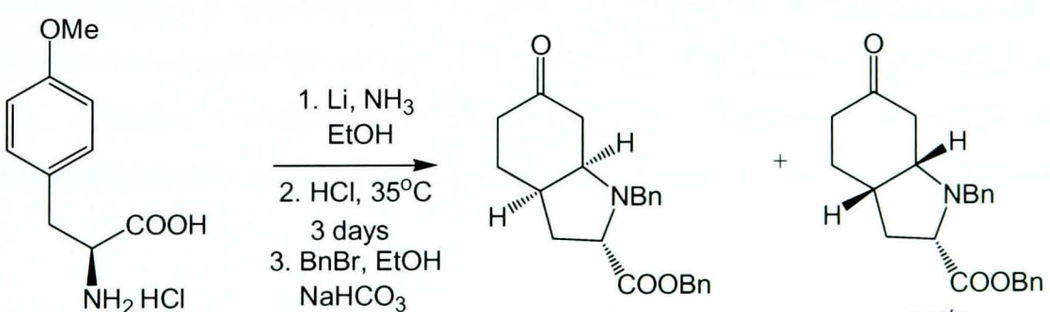


232 **233**

The *N,N*-diallyl protecting group was used to prevent the facile intramolecular 1,4-addition of the amine group to the enone moiety after hydrolysis of the enol ether. This is of note as the 1,4-addition would result in a 6,5-fused ring system analogous to that of herquline A.

Another Birch reduction of a protected tyrosine **234** has been reported by Bonjoch.[185] The group reported that, following the Birch reduction, acidic hydrolysis of the enol ether led to a cyclised product as a mixture of the *exo* and *endo* isomers (Scheme 172). The benzyl protected products **235** and **236** were isolated in 44 % yield over three steps, but no information was reported regarding the ratio of isomers observed.

Scheme 172



234 44 % *exo* **235** *endo* **236**

4.5.2 Application to the Tyrosine System

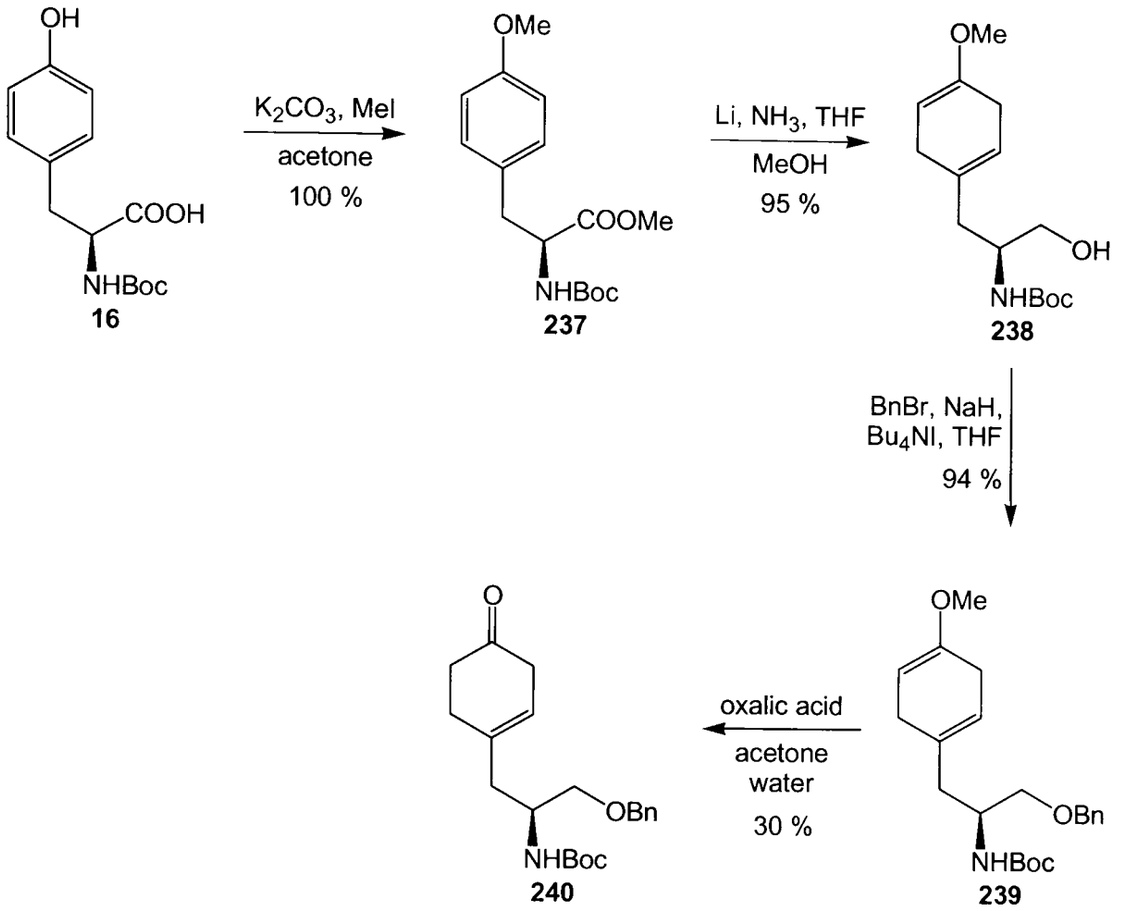
In order to attempt the Birch reduction of the tyrosine system, the functional groups of tyrosine had to be protected appropriately. Boc-protected amines are stable to Birch reduction,[73] so this group was chosen. Although carboxylic acids are stable under Birch conditions, previous problems with handling and isolation of cyclohexadienes bearing acid substituents led to the decision to protect the acid moiety as a methyl ester as previously. The phenol hydroxyl group was simultaneously protected as a methyl ether since phenols do not undergo Birch reduction.

N-Boc-L-tyrosine **16** (see Section 2.2.7.1 for preparation) was treated with potassium carbonate and methyl iodide to give the fully protected tyrosine **237** (Scheme 173). Treatment of the protected tyrosine **237** with lithium in liquid ammonia in the presence of methanol then led to reduction of the aromatic ring and of the ester to give the expected cyclohexadiene **238** in excellent yield. The newly generated alcohol group was then protected as the corresponding benzyl ether **239** in good yield. Hydrolysis of the enol ether using aqueous oxalic acid then provided the β,γ-enone **240** in moderate yield.*

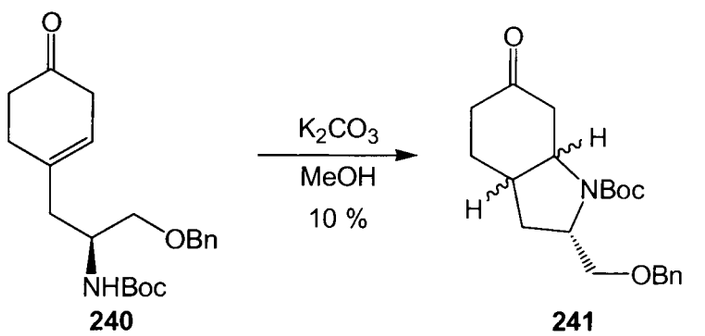
Treatment of the β,γ-enone **240** with potassium carbonate in methanol (Scheme 174) followed by purification by reverse phase hplc led to isolation of the expected bicyclic system **241** as a mixture of diastereoisomers – presumably the *endo* and *exo* isomers, with *cis*-ring-junctions in both cases. Formation of the bicyclic system was confirmed by ^{1}H-NMR spectroscopy: the absence of a signal from the olefinic region and the presence of two signals at around 4 ppm corresponding to the two protons adjacent to the carbamate group indicated that 1,4 addition had occurred. Examination of the ^{13}C-NMR spectrum revealed that all expected signals were 'doubled up' despite purification by hplc, indicating a mixture of two diastereoisomers or rotamers. The sample could not be purified further or the stereochemistry of the product(s) conclusively determined.

* The cyclohexadienes **238** and **239** could not be purified by flash chromatography; the yields for the Birch reduction and benzyl protection steps are therefore yields of crude products. Consequently, the yield for the hydrolysis step includes purification after three steps.

Scheme 173

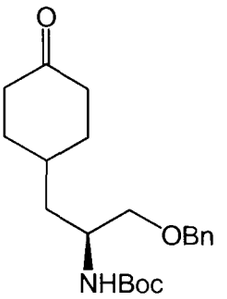


Scheme 174



Also isolated was the cyclohexanone **242** (Figure 54), thought to arise from over-reduction during the Birch reduction step. The presence of a broad doublet at 4.75 ppm in the ^{1}H-NMR spectrum, corresponding to the NH group, indicated that no ring formation had occurred.

Figure 54



242

4.6 Conclusions

A model benzoic ester was synthesised, and Birch reduction led to formation of a cyclohexadiene without reduction of the ester group. Addition of an ester substituent and Birch reduction were then carried out on the tyrosine system.

Following problems with dilute mineral acids, enol ether hydrolysis was achieved using aqueous oxalic acid.

The radical coupling of a model β-ketoester was attempted using copper(II) acetate. It was pleasing to find that the substrate did not immediately rearomatise under these conditions, and that some of the desired dimer may have been formed.

A reduced oxindole system analogous to that found in herquline A was synthesised. Unfortunately the mixture of diastereoisomers was inseparable by hplc.

CHAPTER 5
CONCLUSIONS

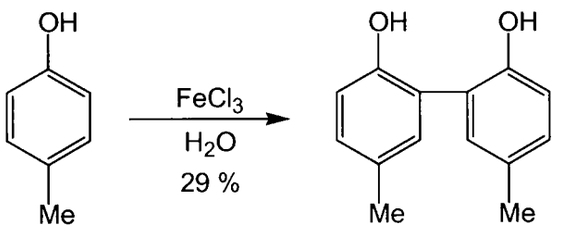
5.1 Formation of Dityrosine

A number of different techniques were examined in attempts to synthesise a dityrosine derivative:

5.1.1 Oxidative Coupling Techniques

A number of different reagents were used in attempts to affect the oxidative coupling of the model compound *p*-cresol **20**. The *p*-cresol dimer **28** was eventually generated using ferric chloride (Scheme 175); the straightforward nature of the reaction and the readily available starting materials helped to compensate for the modest yield.

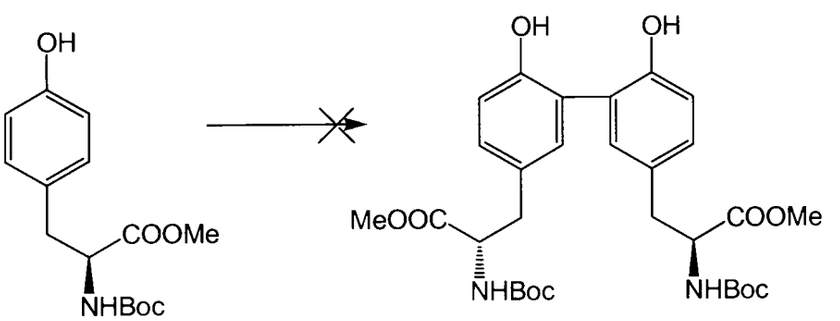
Scheme 175



20 **28**

However, the oxidative coupling of tyrosine derivatives could not be achieved using any of the methods investigated (Scheme 176).

Scheme 176

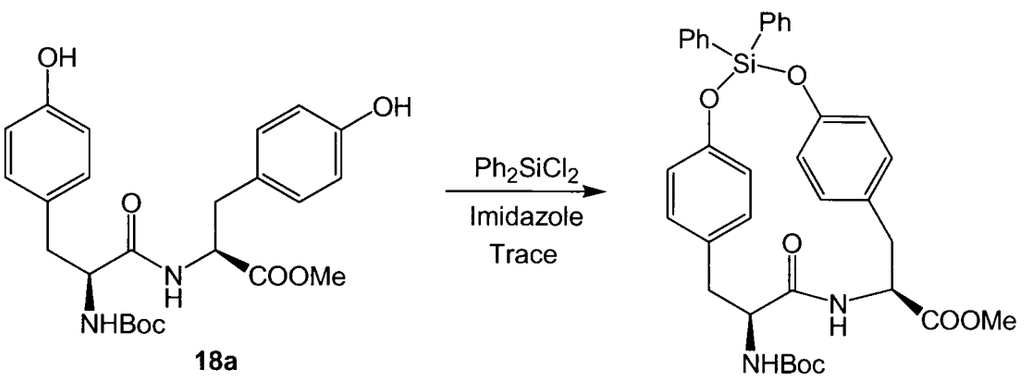


36 **37**

A tyrosine dipeptide and diketopiperazine were synthesised, but intramolecular oxidative coupling was not successful.

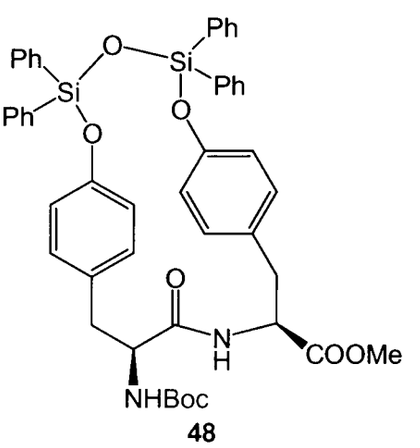
The linking of the two phenol oxygen atoms of a dipeptide *via* a silicon group was attempted in order to mimic the transition state involved in oxidative coupling (Scheme 177).

Scheme 177



The siloxane **46** was obtained in very low yield, and molecular modelling confirmed that the seventeen-membered ring was highly strained. The isolation of a cyclic siloxane **48** containing a nineteen-membered (Figure 55) ring along with molecular modelling results indicated that the nineteen-membered ring was significantly less strained that the seventeen-membered ring. These results implied that the transition state involved in an intramolecular oxidative coupling reaction would be highly strained; this may account for the failure of such reactions on the dipeptide **18a**.

Figure 55

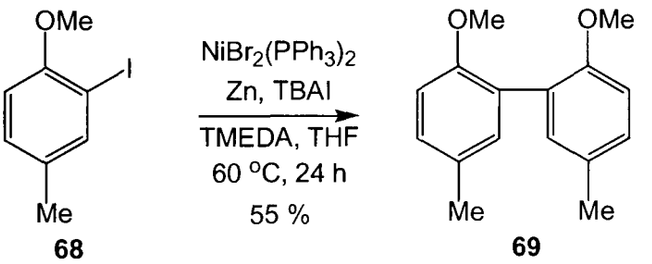


Attempts were made to synthesise a crystalline diketopiperazine derivative in order to obtain more information on the conformation of tyrosine diketopiperazines, but the derivatives synthesised could not be recrystallised in a satisfactory manner.

5.1.2 Ullmann-Type Coupling

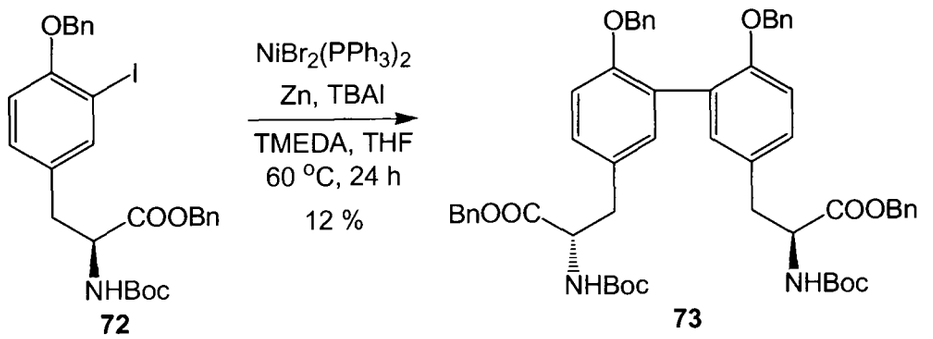
The nickel-mediated Ullmann coupling of a model aryl halide **68** was achieved and the methyl anisole dimer **69** generated. Optimisation work was carried out, and the yield improved by the addition of the bidentate ligand TMEDA (Scheme 178).

Scheme 178



However, the reaction proceeded in very low yield when applied to the tyrosine system **72** (Scheme 179), possibly due to the increased steric bulk around the centres to be coupled or to the slightly acidic NH group.

Scheme 179

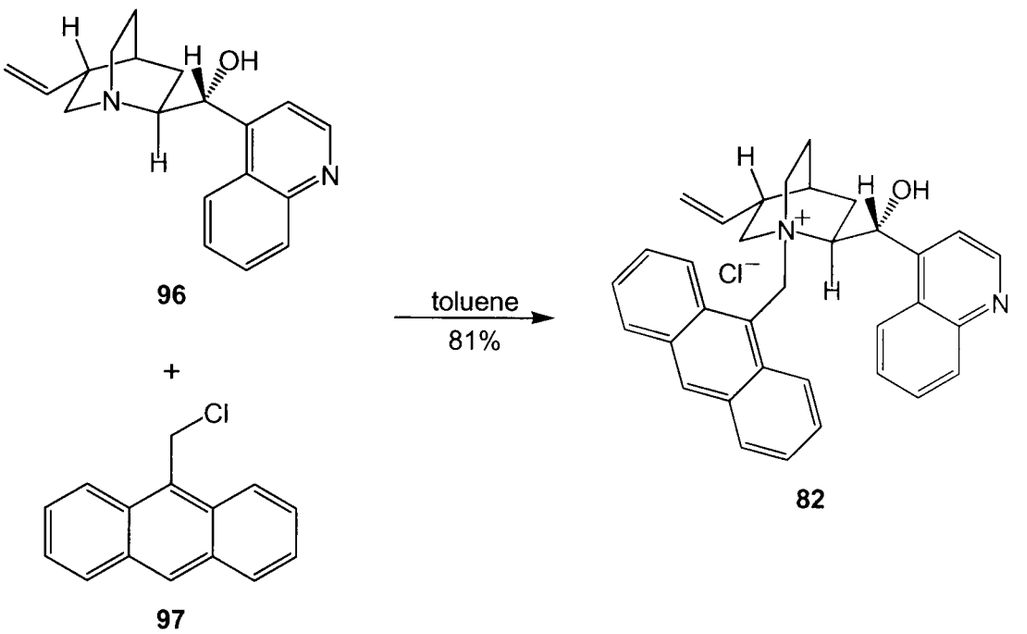


Iodinated tyrosine dipeptides **78** and **79** were synthesised, but the nickel-mediated reaction did not work on an intramolecular basis and dehalogenation was observed.

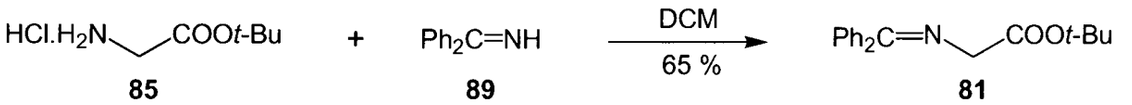
5.1.3 Asymmetric Alkylation

The required phase transfer catalyst **82** (Scheme 180) and *t*-butyl *N*-(diphenylmethylene)glycinate **81** (Scheme 181) were successfully synthesised.

Scheme 180

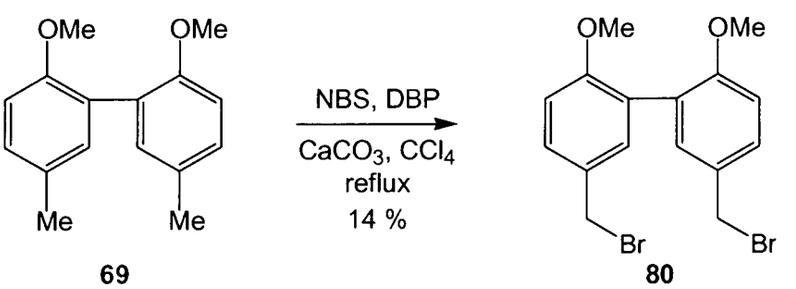


Scheme 181



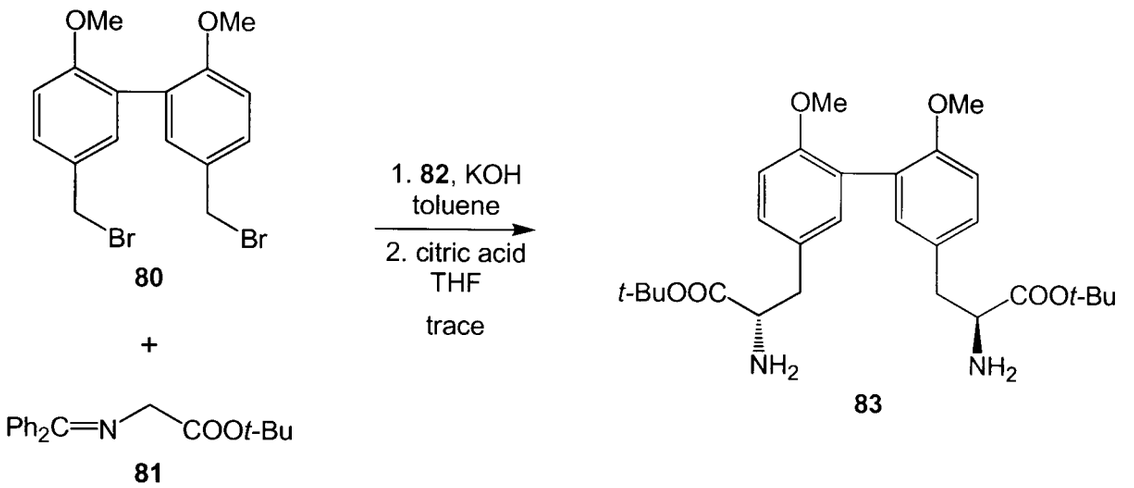
The required dibromobiphenyl compound **69** was prepared, but the inconsistent nature of the reaction and a difficult separation by flash chromatography resulted in a low yield of the dibromide **80** (Scheme 182).

Scheme 182



The asymmetric alkylation step was attempted and a small amount of the protected dityrosine **83** synthesised (Scheme 183).

Scheme 183

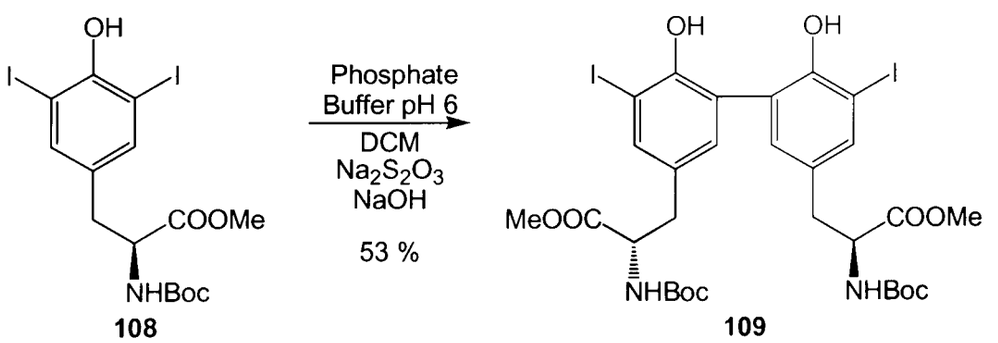


However, the route was not pursued due to the number of steps involved and the problematic radical bromination reaction.

5.1.4 Bowman Coupling

A protected diiodotyrosine derivative **108** was prepared, and the corresponding diiododityrosine **109** synthesised. Optimisation work was carried out, and an optimum reaction time and concentration identified. The yield of the reaction was also improved by the dropwise addition of a solution of sodium thiosulfate in order to maintain a low concentration of iodine in the reaction mixture (Scheme 184).

Scheme 184



Although the optimisation work improved the yield of the reaction, the process was not successful when scaled up, possibly due to a change in the efficiency of mixing. An iodinated dipeptide **111** and diketopiperazine **112** were prepared, but the Bowman coupling was not successful on an intramolecular basis.

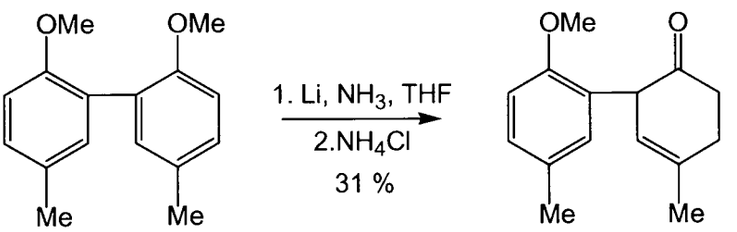
5.2 Reduction of a Dityrosine System

A number of different techniques were examined in work towards the reduction of a dityrosine system:

5.2.1 The Birch Reduction

Experience in performing Birch reductions was gained by carrying out reductions on small model compounds. These initial reactions helped to establish general reaction conditions and to confirm the high degree of regioselectivity of the reaction. The Birch reduction of a model biphenyl system **69** confirmed that reduction of one aromatic ring proceeded as expected, and provided the expected (and desired) regioselectivity (Scheme 185).

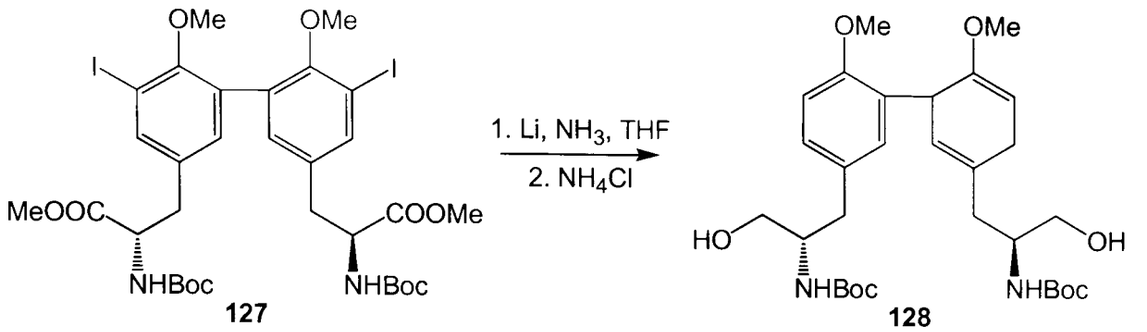
Scheme 185



69 **125**

The Birch reduction of a protected diiododityrosine system established that aryl iodides undergo reductive dehalogenation and standard Birch reduction under reducing metal conditions (Scheme 186).

Scheme 186

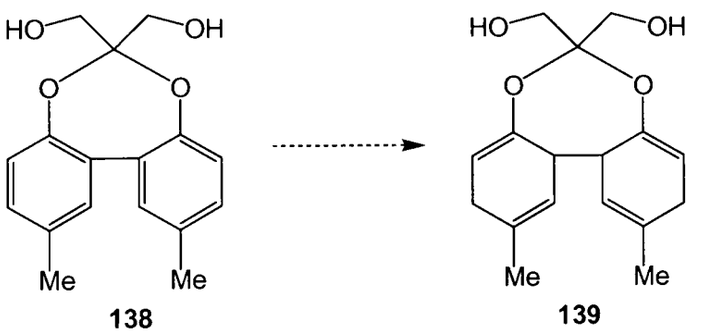


This confirmed that the diiododityrosine produced by the Bowman coupling reaction may be a suitable substrate for further studies.

5.2.2 The Directed Birch Reduction

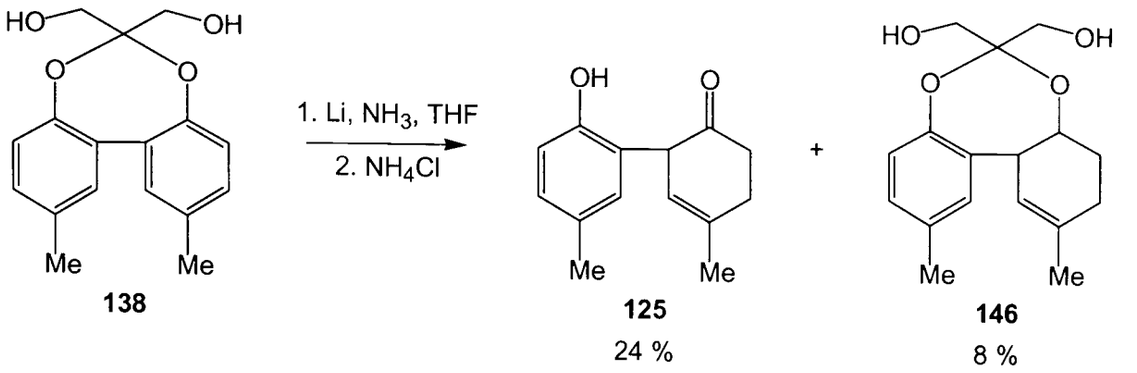
Two model biphenyl compounds containing hydroxyl groups in close proximity to the biphenyl system were prepared in the hope that the hydroxyl groups would act as an intramolecular proton source, and so alter the regiochemistry of the Birch reduction (Scheme 187).

Scheme 187



However under Birch reduction conditions, the hydroxyl groups failed to facilitate reduction of the second aromatic ring, and the major products were the hydrolysis products of aryl cyclohexadienes (Scheme 188). A small amount of an over-reduced product **146** was isolated, indicating that the proximal hydroxyl groups had influenced the course of the Birch reduction.

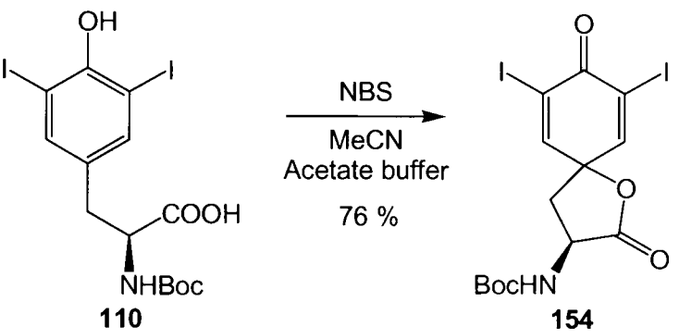
Scheme 188



5.2.3 Reduction *via* Spirolactone Formation

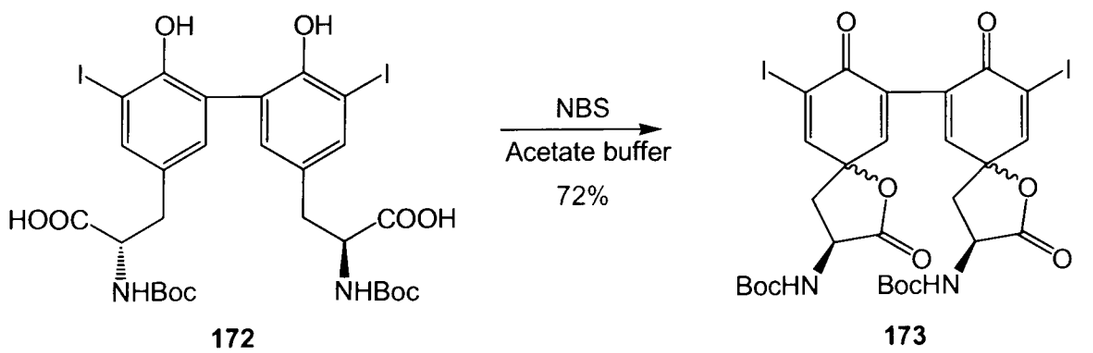
NBS was used to generate a diiodotyrosine spirolactone (Scheme 189).

Scheme 189



Subsequently, NBS was used to generate a dispirolactone **173** from a diiododityrosine **172** (Scheme 190), with two of the three possible diastereoisomers being formed.

Scheme 190



Reduction of the dispirolactone was not pursued since an identification of an efficient method for dityrosine formation had not been successful; a new route for the synthesis of herqulines A and B was sought.

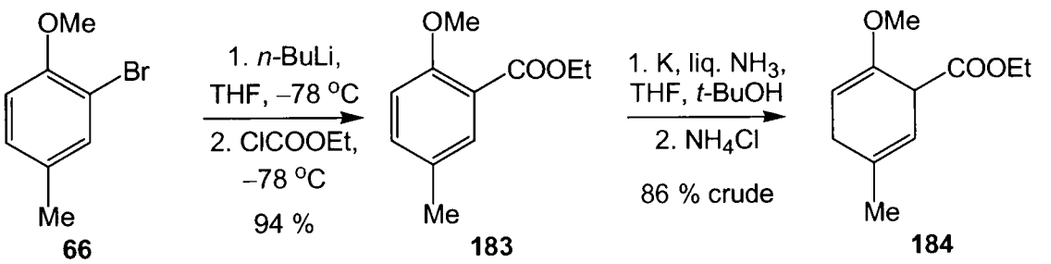
5.3 Synthesis & Reactions of Birch-Reduced Tyrosine Systems

The reduction of a tyrosine system and subsequent coupling was investigated. This strategy removed the regiochemical problem associated with the Birch reduction of a biphenyl system.

5.3.1 Birch Reduction of a Functionalised System

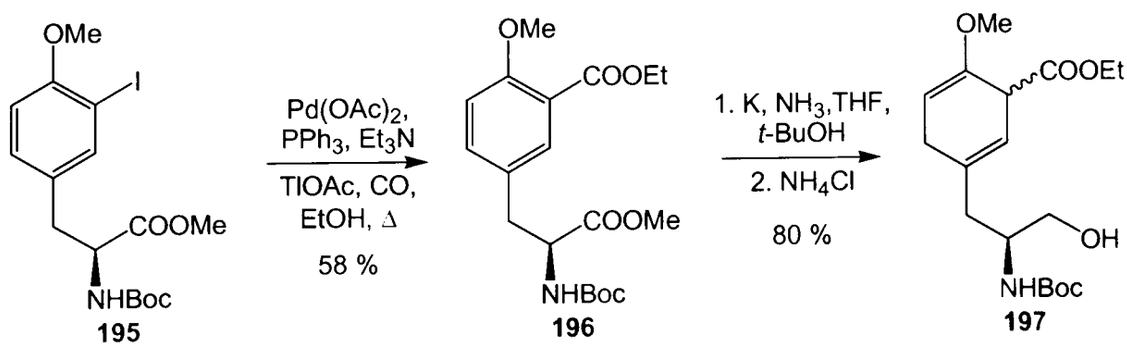
A model benzoic ester **183** was generated from an aryl bromide **66** (Scheme 191). Birch reduction of the ester using potassium and *t*-butanol resulted in the formation of the corresponding cyclohexadiene **184**, without reduction of the carboxylic ester.

Scheme 191



An ester group was then introduced to the aromatic ring of tyrosine using a protected iodotyrosine **195** and palladium-catalysed carbonylation (Scheme 192). Birch reduction using potassium and *t*-butanol again provided a cyclohexadiene **197** without reduction of the aromatic ester. The aliphatic ester was reduced as expected.

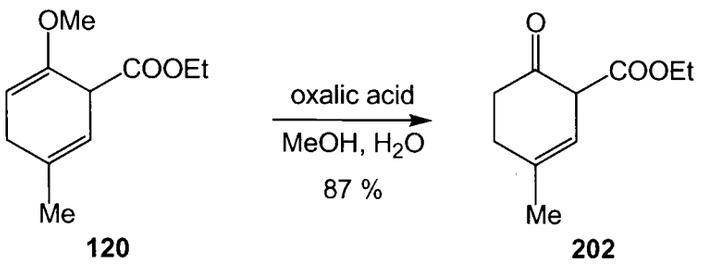
Scheme 192



5.3.2 Enol ether hydrolysis

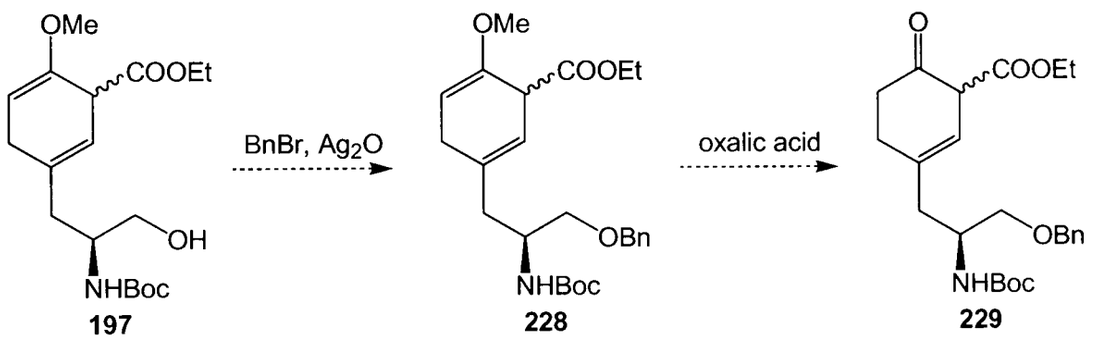
Following the testing of a number of different reagents, aqueous oxalic acid was found to be the reagent of choice, providing only the β,γ-enone **202** (Scheme 193).

Scheme 193



Unfortunately, time constraints prevented the application of this technique to the tyrosine system **197** (Scheme 194).

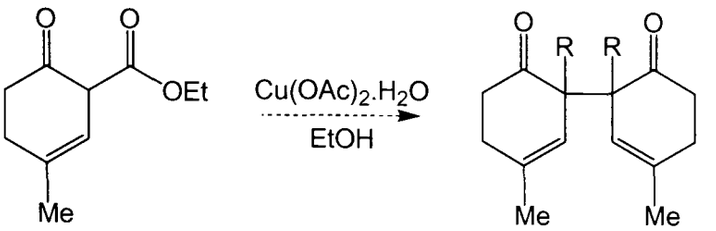
Scheme 194



5.3.3 Radical Coupling

Radical coupling of an unsaturated β-ketoester **202** was attempted using the single-electron oxidant copper (II) acetate (Scheme 195).

Scheme 195



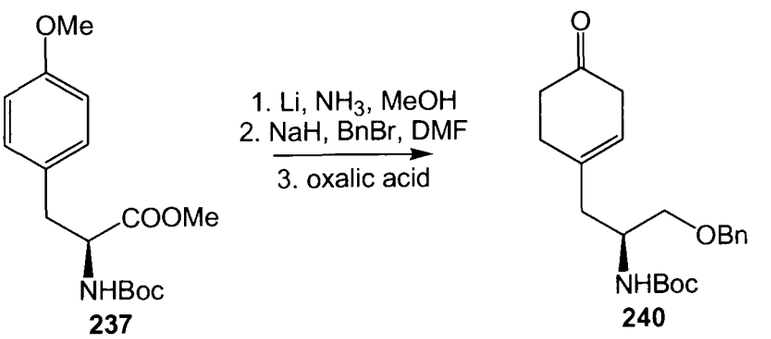
202 **227**

It was encouraging to find that the system did not undergo complete rearomatisation on treatment with a single-electron oxidant. The presence of new signals in the ^{1}H-NMR spectrum indicated that some of the dimer may have been formed; however, problems experienced during purification of the β-ketoester **202** meant that formation of the dimer could not be conclusively established.

5.3.4 Synthesis of the 6,5-Ring System

A appropriately protected tyrosine moiety **237** was synthesised and the system treated with Birch reduction conditions; the corresponding cyclohexadiene was generated with reduction of the carboxylic ester (Scheme 196). Benzyl protection of the alcohol followed by enol ether hydrolysis provided the β,γ-enone **240**.

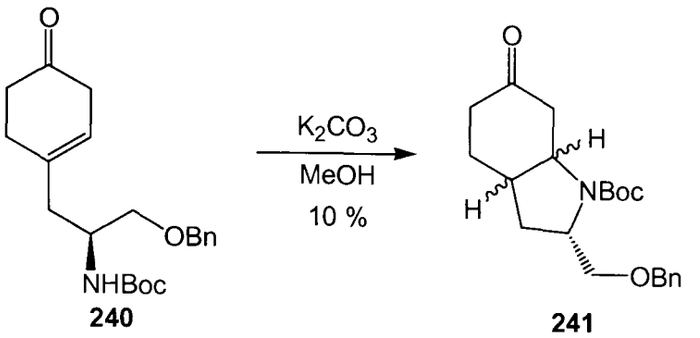
Scheme 196



237 **240**

Treatment of the β,γ-enone **240** with potassium carbonate in methanol then led to the formation of the desired bicyclic system **241** (Scheme 197); unfortunately the resulting mixture of diastereoisomers could not be separated.

Scheme 197



5.4 Summary

Since no significant work toward the synthesis of the herqulines had been previously reported, this project has been focussed on the strategy for achieving the two key steps in the synthesis. It has established that while an intramolecular oxidative phenol coupling reaction may afford a molecule with a carbon skeleton very similar to that of the herqulines, the problem of reduction of the aromatic rings at this stage is not a trivial one.

Whilst the intramolecular coupling reactions examined in Chapter 2 were not successful, an intramolecular coupling strategy provides an attractive solution to the formation of the heterocyclic rings of the herqulines. The formation of the piperazine ring at a later stage of the synthesis would require the use of orthogonal protecting groups; a hetero-coupling reaction would then be required to synthesise an unsymmetrical dityrosine-derived compound.

The radical coupling of a reduced, functionalised tyrosine derivative appears to be the most promising strategy to date. The power of such a strategy may be increased by combination with the intramolecular coupling strategy discussed above.

5.5 Future Work

The completion of a small number of experiments would help to conclude this project and clarify several points under investigation:

1) Acquisition of a crystalline tyrosine-diketopiperazine and an X-ray crystal structure in order to obtain more information on the conformation of diketopiperazines.

2) Investigation of the oxidative coupling of the *N*-PMB-protected tyrosine diketopiperazine and a protected tyrosine piperazine.

3) Investigation of the nickel-mediated Ullmann reaction on a methyl-protected rather than a benzyl-protected tyrosine.

4) Investigation of the effect of mechanical rather than magnetic stirring on the Bowman coupling reaction.

Finally, this work could be continued by further investigation into the radical coupling of β-ketoesters and the application of this reaction to tyrosine-derived systems. The use of an intramolecular radical coupling strategy may result in further progress towards the total synthesis of herqulines A and B.

CHAPTER 6
EXPERIMENTAL

6.1 General Experimental

Ether refers to diethyl ether and petrol refers to petroleum spirit (b.p. 40-60 °C) unless otherwise stated. Ethyl acetate and petrol were distilled before use. THF and ether were freshly distilled from sodium/benzophenone and DCM was freshly distilled from calcium hydride. Methanol and ethanol were both distilled from magnesium/iodine and stored over 4 Å molecular sieves, and triethylamine was distilled from calcium hydride and stored over potassium hydroxide. Toluene was freshly distilled from calcium hydride or dried over 3 batches of 4 Å molecular sieves; DMF was dried over 3 batches of 4 Å molecular sieves. Benzonitrile was stirred over phosphorus pentoxide overnight and distilled under reduced pressure and piperylene was dried over 3 batches of 4 Å molecular sieves. *t*-Butanol was distilled from calcium hydride and stored over 4 Å molecular sieves. Solvents were removed under reduced pressure using a Büchi rotary evaporator at diaphragm pump pressure; DMF was removed under reduced pressure using a Büchi rotary evaporator at oil pump pressure. Samples were routinely dried under high vacuum overnight before analysis.

Aqueous reactions were carried out using distilled water and aqueous solutions were saturated unless stated otherwise. All non-aqueous reactions were carried out under nitrogen using flame-dried glassware. Birch reductions were carried out using liquid ammonia freshly distilled from sodium and argon-flushed flame-dried equipment; dryness was maintained using a soda lime drying tube. Grignard reagents were titrated against salicylaldehyde phenylhydrazone according to the method of Love and Jones[111] before use. *n*-Butyllithium and *t*-butyllithium were titrated against diphenylacetic acid or salicylaldehyde phenylhydrazone before use. Hydrogen chloride gas was produced by the addition of concentrated sulfuric acid to calcium chloride; the gas was dried by passage through 4 Å molecular sieves. Diazomethane solutions were prepared using Diazald® and potassium hydroxide using standard procedures.[186] Zinc powder was activated by washing with aqueous hydrochloric acid (2 M) and acetone; the powder was then dried under high vacuum.

Flash column chromatography was carried out using silica (35-70 μm particles) according to the method of Still, Kahn and Mitra.[187] Thin layer chromatography was carried out on commercially available pre-coated plates (Merck silica Kieselgel $60F_{254}$). Analytical hplc was carried out with acetonitrile in water or i-propanol in hexane on a Dionex/Gynkotek system using full-spectrum UV detection. Preparative hplc was carried out with acetonitrile and water on a Gilson preparatory system using single-wavelength UV detection.

Proton and carbon NMR spectra were recorded on a Bruker DPX 300, DRX 500 or Avance 500 Fourier transform spectrometer using an internal deuterium lock. Chemical shifts are quoted in parts per million downfield of tetramethylsilane; spectra recorded in chloroform-*d* contained tetramethylsilane as an internal standard while spectra recorded in other solvents were calibrated such that residual solvent peaks matched published values. Coupling constants (*J*) are given in Hz and refer to vicinal H-H couplings unless indicated otherwise. Carbon NMR spectra were recorded with broad band proton decoupling. COSY, TOCSY, DEPT, HMQC, HMBC and nOe spectra were used in the assignment of proton and carbon NMR spectra where necessary.

Mass spectra were recorded on a VG AutoSpec mass spectrometer, operating at 70eV, using electron impact ionisation (EI) or fast atom bombardment (FAB). Electrospray (ES) spectra were recorded using positive ionisation on a Micromass LCT TOF spectrometer or a Waters-Micromass ZMD spectrometer. Accurate molecular weights were recorded by the staff of the Department of Chemistry using the instruments listed above by peak matching with perfluorokerosene or reserpine used as a standard, or by staff of the EPSRC National Mass Spectrometry Service using positive ES ionisation on a Finnegan MAT 900 XLT or a Finnegan MAT 95XP spectrometer and polyethylenimine as a standard. LCMS was carried out using a Waters-Micromass ZQ system. Microanalyses were carried out by the staff of the Department of Chemistry using a Carlo Erba 1108 automatic analyser and a Perkin Elmer AAnalyser 100 atomic absorption spectrometer. Infrared spectra were recorded on a Nicolet Avatar 360 FT-IR infrared spectrophotometer or a Perkin-Elmer Spectrum One FT-IR infrared spectrophotometer using sodium chloride plates with signals referenced to the polystyrene 1601 cm^{-1} absorption. Solid phase spectra were recorded using diamond plates. Melting points were determined on a Reichert hot stage apparatus and are

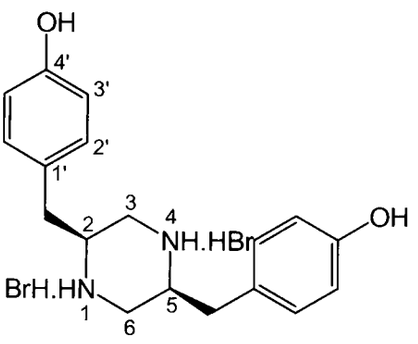
uncorrected. Optical rotations were recorded on an Optical Activity A-1000 polarimeter (using the sodium D line; 589 nm) and $[\alpha]_D^{20}$ are given in units of 10^{-1} deg cm^2 g^{-1}.

6.2 Experimental Details

Bis(triphenylphosphine)nickel(II) dibromide

A solution of triphenylphosphine (10.51 g, 0.04 mol) in boiling *n*-butanol (100 ml) was added to a solution of nickel(II) bromide (anhydrous; 4.44 g, 0.02 mol) and water (1.1 ml, 0.06 mol) in boiling *n*-butanol (100 ml). The reaction mixture was allowed to cool slowly, and the resulting precipitate isolated by reduced pressure filtration to give the nickel complex (9.511 g, 64 %) as dark green needles, m.p. 229 °C (from *n*-butanol; lit.[188] 228-231 °C); (Found C, 57.9; H, 3.8; Br, 21.4; Ni, 7.8; P, 8.1; $C_{36}H_{30}Br_2NiP_2$ requires C, 58.2; H, 4.1; Br, 21.5; Ni, 7.9; P, 8.3 %); ν_{max}/cm^{-1} (nujol mull)* 526, 494, 336, 264 (Ni-Br), 226 and 220.

(2*S*,5*S*)-2,5-Bis-(*p*-hydroxybenzyl)piperazine dihydrobromide 15a

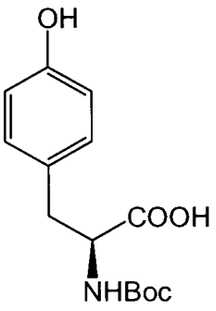


BH_3-THF complex (1 M solution in THF; 5 ml, 5.00 mmol) was added to a stirred solution/suspension of the diketopiperazine **19a** (201 mg, 0.62 mmol) in anhydrous THF (3 ml). The reaction mixture was heated at reflux, but after 14 hours some solid material still remained. A further portion of BH_3-THF complex (5 ml, 5.00 mmol) was added and the reaction mixture heated at reflux for a further 24 hours. The mixture was allowed to cool to room temperature then cooled in ice. Hydrogen bromide (45 % solution in acetic acid; 3 ml) was added dropwise and the reaction mixture stirred at 0 °C for 2 hours. The resulting precipitate was isolated by reduced-pressure filtration; the filtrate was diluted with hexane (5 ml) and stored at −18 °C for 24 hours in order to obtain a second crop. The two crops were combined to give the piperazine **15a** (248 mg, 88 %) as a colourless powder, m.p. >290 °C; R_f 0.19 (methanol); $[\alpha]_D$ −7.6 (*c* 1 in water); ν_{max}/cm^{-1} (solid phase) 3211br (NH_3^+ & OH), 1610 (Ar), 1513 (Ar), 1452 (Ar) and 1185 (ArOH); δ_H (300 MHz; water-d_2) 7.22 (4 H, d, *J* 8.5, 4 × 2'-H), 6.87 (2 H, *J* 8.5, 4

* Recorded on a Philips PU 9522 infrared spectrometer using polystyrene plates (600 – 200 cm^{-1}).

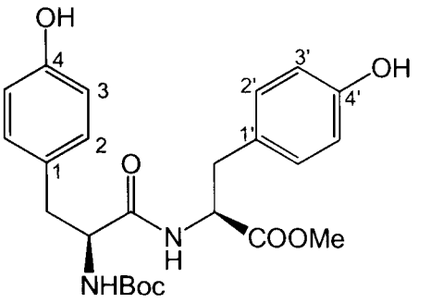
× 3'-H), 3.96 (2 H, app. quintet, *J* 5.4, 2 × C*H*NH), 3.48-3.45 (4 H, m, 2 × CH_2NH) and 3.10 (4 H, d, *J* 7.9, 2 × CH_2Ar); δ_C (75 MHz; water-d_2) 155.6 (C4'), 131.2 (C2'), 125.5 (C1), 116.5 (C3'), 52.9 (CHNH), 41.4 (CH_2NH) and 33.5 (CH_2Ar); *m/z* (ES) 597 (5 %, M_2H^+), 299 (100, MH^+) and 193 ($MH_2^+ - CH_2C_6H_4OH$).

N-*t*-Butoxycarbonyl-L-tyrosine 16



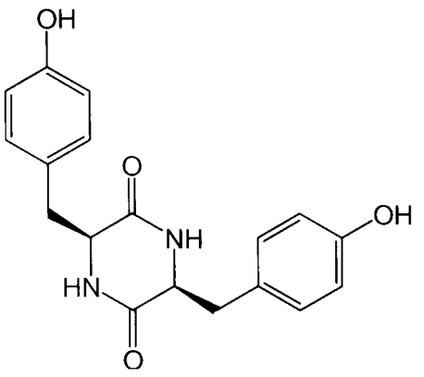
Triethylamine (11.6 ml, 83.4 mmol) was added to a stirred solution/suspension of tyrosine **3** (10.0 g, 55.4 mmol) in dioxane (100 ml) and water (100 ml). The reaction mixture was cooled to 0 °C and di-*t*-butyl dicarbonate (14.0 ml, 61.0 mmol) added. The mixture was stirred for 1 hour at 0 °C, then for 20 hours at room temperature. The dioxane was removed *in vacuo* and the residue acidified using aqueous hydrochloric acid (2 M), then extracted with ethyl acetate (3 × 40 ml). The combined organic extracts were washed with aqueous HCl (2 M; 40 ml) and brine (40 ml), dried ($MgSO_4$) and concentrated to give the carbamate **16** (15.69 g, 100%) as a colourless foam, m.p. 52-54 °C (lit.[189] 136-138 °C); R_f 0.46 (50 % ethyl acetate & 2 % acetic acid in petrol); $[\alpha]_D$ +16 (*c* 0.5 in methanol); ν_{max}/cm^{-1} (nujol mull) 3337 (COOH & OH), 1720 (acid C=O), 1638 (carbamate C=O), 1615 (Ar), 1516 (NH/CN) and 1160 (PhOH); δ_H (300 MHz; methanol-d_4) 7.04 (2 H, d, *J* 8.4, 2 × 3-H), 6.70 (2 H, d, *J* 8.4, 2 × 2-H), 4.27 (1 H, dd, *J* 8.7 and 5.1, CH), 3.05 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 5.1, CH_AH_B), 2.81 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 8.7, CH_AH_B) and 1.38 (9 H, s, *t*-Bu); δ_C (75 MHz; methanol-d_4) 176.1 (COOH), 158.2 (NHCO), 157.7 (C1), 131.7 (C2), 129.7 (C4), 116.5 (C3), 80.9 (CMe_3), 57.0 (CH), 38.4 (CH_2) and 29.1 (CMe_3); *m/z* (EI) 281 (<1 %, M^+), 245 (1), 164 (29, $M^+ - NH_2Boc$), 136 (10, MH^+ – COOH, – Boc), 107 (100, $HOC_6H_4CH_2^+$), 91 (57, $C_7H_7^+$) and 57 (47, *t*-Bu^+).

(*N*-*t*-butoxycarbonyl-L-tyrosine)-(L-tyrosine methyl ester) 18a



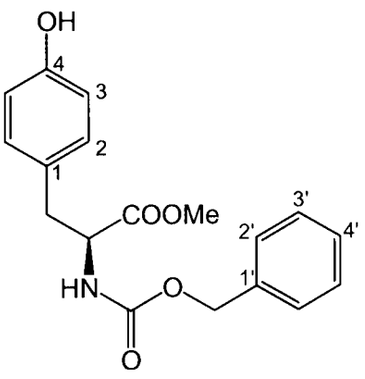
The carboxylic acid **16** (3.009 g, 10.71 mmol), the amine **33** (2.476 g, 10.70 mmol), HOBt (1.451 g, 10.75 mmol), triethylamine (2.2 ml, 15.8 mmol) and a catalytic quantity of DMAP were added to a stirred solution of DCC (2.240 g, 10.87 mmol) in anhydrous THF (100 ml). The reaction mixture was stirred at room temperature for 24 hours, then kept at −18 °C for 16 hours. The mixture was filtered while cold and the filtrate concentrated. The residue was partitioned between ethyl acetate (50 ml) and water (30 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 20 ml). The combined organic extracts were washed with water (15 ml), aqueous hydrochloric acid (2 M; 15 ml), water (15 ml), sodium bicarbonate (10 %; 15 ml) and brine (15 ml), dried ($MgSO_4$) and concentrated to give a crude product. The residue was dissolved in the minimum amount of THF and kept at −18 °C for 48 hours. The mixture was then filtered under reduced pressure while still cold and the filtrate concentrated to give the dipeptide **18a** (4.553 g, 93 %) as a colourless solid, m.p. 82-84 °C (lit.[35] 87-90 °C); R_f 0.33 (50 % ethyl acetate in petrol); $[\alpha]_D$ −4.0 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3305 (OH & NH), 1727 (ester C=O), 1686 (carbamate C=O), 1660 (amide C=O), 1614 (Ar), 1596 (Ar), 1515 (Ar), 1242 and 1161 (ArOH); δ_H (300 MHz; methanol-d_4) 6.93 (2 H, d, *J* 8.3, 2 × 2-H or 2'-H), 6.90 (2 H, d, *J* 8.3, 2 × 2-H or 2'-H) 6.61 (2 H, d, *J* 8.3, 2 × 3-H or 3'-H), 6.60 (2 H, d, *J* 8.3, 2 × 3-H or 3'-H), 4.51 (1 H, app. t, *J* 7.2, C*H*NHBoc), 4.13 (1 H, dd, *J* 8.7 and 5.6, C*H*COOMe), 3.56 (3 H, s, COOMe), 2.93 (1 H, dd, $^2J_{HH}$ 14.3 and *J* 7.2, CH_AH$_B$CHNHBoc), 2.84 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 5.6, CH_AH$_B$COOMe), 2.81 (1 H, dd, $^2J_{HH}$ 14.3 and *J* 7.2, CH$_A$$H_B$CHNHBoc), 2.58 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 8.7, CH$_A$$H_B$COOMe), and 1.20 (9 H, s, *t*-Bu); δ_C (75 MHz; methanol-d_4) 174.5 & 173.5 (*C*OOMe & CH*C*ONH), 157.8 & 157.5 (NH*C*OO, C4 & C4'), 131.6 (C2 & C2'), 129.4 & 128.7 (C1 & C1'), 116.6 & 116.4 (C3 & C3'), 81.0 (*C*Me$_3$), 57.8 (*C*HCOOMe), 55.6 (*C*HNHBoc), 52.9 (COO*Me*), 38.7 & 38.1 (2 × CH_2) and 29.0 (C*Me*$_3$); *m/z* (FAB) 917 (8 %, M_2H^+), 459 (53, MH^+), 402 (40, MH^+ − *t*-Bu), 359 (46, MH^+ − Boc) and 136 (100, $HOC_6H_4CH_2CHNH_2^+$).

Cyclo(L-tyrosine)-(L-tyrosine) 19a



A solution of the dipeptide **18a** (2.504 g, 5.462 mmol) in formic acid (125 ml) was stirred at room temperature for 2 hours. The mixture was concentrated and the residue dissolved in *s*-butanol (125 ml) and toluene (54 ml). The solution was heated at reflux for 14 hours, then cooled to room temperature and concentrated. The residue was dispersed in toluene (60 ml) and the product was isolated by filtration and washed with ether (2 × 10 ml) to give the diketopiperazine **19a** (1.364 g, 77 %) as a colourless powder, m.p. 270-272 °C (dec.; lit.[35] 277-279 °C); R_f 0.00 (methanol); $[\alpha]_D$ −127.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3312-2929 (OH & NH), 1651 (C=O), 1615 (Ar), 1598 (Ar), 1517 (Ar), 1338 (ArOH), 1247, 1173 (ArOH), 1102, 1012 and 827; δ_H (300 MHz; DMSO-d_6) 9.23 (2 H, br s, 2 × OH), 7.79 (2 H, br s, 2 × NH), 6.84 (4 H, d, *J* 8.3, 4 × 2-H), 6.68 (4 H, d, *J* 8.3, 4 × 3-H), 3.85 (2 H, br s, 2 × CH), 2.52 (2 H, dd, $^2J_{HH}$ 13.3 and *J* 4.6, 2 × CH_AH_B) and 2.10 (2 H, dd, $^2J_{HH}$ 13.3 and *J* 6.5, 2 × CH_AH_B); δ_C (75 MHz; DMSO-d_6) 166.6 (C=O), 156.4 (C4), 131.1 (C2), 126.9 (C1), 115.4 (C3), 56.1 (CH) and 39.2 (CH_2); *m/z* (FAB) 327 (67 %, MH^+), 286 (14), 256 (13), 220 (23, MH^+ – $CH_2C_6H_4OH$) and 133 (100).

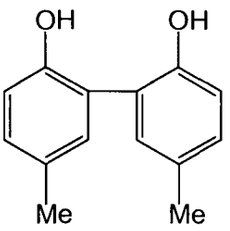
***N*-Benzyloxycarbonyl-L-tyrosine methyl ester 26**



A freshly prepared solution of diazomethane was added to a stirred solution of *N*-Cbz-L-tyrosine **29** (315 mg, 1.00 mmol) in ethanol (8 ml) at 0 °C until a pale yellow colour persisted. Nitrogen was bubbled through the reaction mixture until the yellow colour was displaced. The mixture was then allowed to warm to room temperature and concentrated to give the ester **26** (248 mg, 75 %) as a colourless solid, m.p. 92-93 °C (lit.[190] 91-92 °C); R_f 0.44 (50 % ethyl acetate in petrol); $[\alpha]_D$ −5.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3340 (OH), 2485, 2071, 1699 (C=O), 1516 (Ar), 1353 (ArOH) and 1174 (ArOH); δ_H (300 MHz; methanol-d_4) 7.04-6.90 (5 H, m, Ph), 6.71 (2 H, d, *J* 8.5, 2 × 2-H), 6.43 (2 H, d, *J* 8.5, 2 × 3-H), 4.73 (2 H, s, OCH_2Ph), 4.12 (1 H, dd, *J* 8.9 and 5.6, $CHCH_2$), 3.35 (3 H, s, Me),

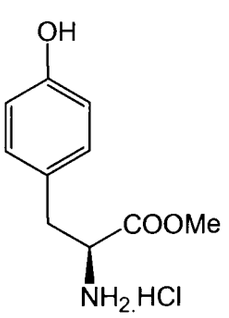
2.74 (1 H, dd, $^2J_{HH}$ 13.8, J 5.6, CHCH_AH$_B$) and 2.54 (1 H, dd, $^2J_{HH}$ 13.8, J 8.9, CHCH$_A$$H_B$); δ_C (75 MHz; methanol-d_4) 174.3 (*C*OOMe), 158.5 (NH*C*OOBn), 157.5 (C4), 138.2 (C1'), 131.5 (C2), 129.6 (C3'), 129.1 & 128.9 (C1 & C4'), 128.8 (C2'), 116.5 (C3), 67.7 (*C*H$_2$Ph), 57.4 (*C*HCH$_2$), 52.9 (Me) and 38.0 (CH*C*H$_2$); *m/z* (EI) 329 (2 %, M^+), 181 (100), 107 (67, $HOC_6H_4CH_2^+$), and 91 (71, $C_7H_7^+$).

5,5'-Dimethyl biphenyl-2,2'-diol 28



Ferric chloride (15.0 g, 92.6 mmol) was added to a stirred solution of *p*-cresol **20** (10.0 g, 92.6 mmol) in water (610 ml). The reaction mixture was stirred at room temperature and the resulting brown precipitate collected by reduced pressure filtration after 12 days. Second and third crops were collected after 24 and 31 days respectively. The crude product was purified by flash chromatography (eluent: 15 % ethyl acetate in petrol) to give the biphenol **28** (2.81 g, 29 %) as a cream-coloured solid, m.p. 163-164 °C (lit.[48] 153-154 °C); R_f 0.40 (20 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3549 (OH), 1498 (Ar), 1382 (ArOH) and 1185 (ArOH); δ_H (300 MHz; chloroform-*d*) 7.10 (2H, dd, J 8.1 and $^4J_{HH}$ 1.9, 4-H & 4'-H), 7.07 (2H, d, $^4J_{HH}$ 1.9, 6-H & 6'-H), 6.91 (2H, d, J 8.1, 3-H & 3'-H), 5.66 (2H, br s, OH) and 2.32 (6H, s, 2 × Me); δ_C (75 MHz; chloroform-*d*) 151.0 (C2 & C2'), 132.0 (C4 & C4'), 131.2 (C5 & C5'), 130.7 (C6 & C6'), 124.0 (C3 & C3'), 116.9 (C1 & C1') and 20.9 (Me); *m/z* (EI) 214 (100 %, M^+), 199 (33, M^+ – Me) and 171 (51).

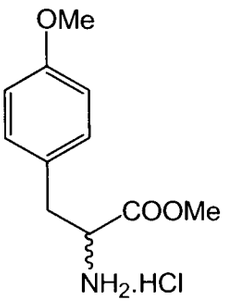
L-Tyrosine methyl ester hydrochloride 33



Thionyl chloride (40.0 ml, 0.55 mol) was added dropwise to a stirred solution/suspension of L-tyrosine dihydrate **3** (10.013 g, 55.32 mmol) in anhydrous methanol (100 ml) at –40 °C. The reaction mixture was allowed to warm to room temperature over 90 minutes, then heated at reflux for 17 hours. The mixture was concentrated to give a crude product, which was recrystallised from methanol/ether to give the methyl ester **33** (12.02 g, 94 %) as an off-white solid, m.p. 133-135 °C (lit.[191] 179-192 °C); R_f 0.65 (methanol); $[\alpha]_D$ +23.0 (*c* 1.0 in methanol); ν_{max} cm^{-1} (nujol mull) 3355 (NH_3^+), 3300 (NH_3^+), 1744 (C=O), 1597 (Ar), 1515 (Ar), 1258, 1179, 1020 and 839; δ_H

(300 MHz; DMSO-d_6) 9.52 (1 H, br s, OH), 8.68 (3 H, br s, NH_3^+), 7.00 (2 H, d, J 8.2, 2 × 2-H), 6.73 (2 H, d, J 8.2, 2 × 3-H), 4.14 (1 H, app. t, J 6.1, CH), 3.65 (3 H, s, Me), 3.09 (1 H, dd, $^2J_{HH}$ 14.1 and J 6.1, CH_AH_B) and 2.66 (1 H, dd, $^2J_{HH}$ 14.1 and J 6.1, CH_AH_B); δ_C (75 MHz; DMSO-d_6) 175.8 (C=O), 156.2 (C4), 130.4 (C2), 128.1 (C1), 115.3 (C3), 56.3 (CH), 51.6 (Me) and 40.5 (CH_2); *m/z* (EI) 195 (39 %, M^+), 136 (43, M^+ – COOMe), 107 (100, $HOC_6H_4CH_2^+$) and 88 (55, $MeOOCCHNH_2^+$).

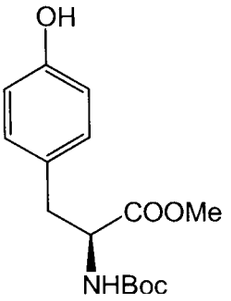
D,L-Tyrosine methyl ester hydrochloride 35



Thionyl chloride (2.05 ml, 28.10 mmol) was added dropwise to a stirred solution/suspension of D,L-tyrosine **34** (0.506g, 2.80 mmol) in anhydrous methanol (5 ml) at –40 °C. The reaction mixture was allowed to warm to room temperature over 2 hours, then heated at reflux for 17 hours. The reaction mixture was concentrated and the residue dissolved in water (20 ml). The solution was basified using aqueous sodium hydroxide (2 M) and extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give the racemic ester **35** (308 mg, 56 %) as a cream-coloured solid, m.p. 109 °C (lit.[192] 120-122 °C); R_f 0.68 (methanol); ν_{max}/cm^{-1} (nujol mull) 3353 (NH_2), 3299 (NH_2), 1743 (C=O), 1596 (NH & Ar), 1515 (Ar), 1257 (ArOH), 1171, 1019 (ArOH) and 839; δ_H (300 MHz; DMSO-d_6) 6.95 (2 H, d, J 8.4, 2 × 2-H), 6.66 (2 H, d, J 8.4, 2 × 3-H), 3.56 (3 H, s, Me), 3.48 (1 H, app. t, J 6.6, CH), 2.74 (1 H, dd, $^2J_{HH}$ 13.4 and J 6.6, CH_AH_B) and 2.66 (1 H, dd, $^2J_{HH}$ 13.4 and J 6.6, CH_AH_B); δ_C (75 MHz; DMSO-d_6) 175.8 (C=O), 156.2 (C4), 130.4 (C2), 128.1 (C1), 115.3 (C3), 56.26 (CH), 51.6 (Me) and 40.8 (CH_2); *m/z* (ES) 391 (10 %, $[M - HCl]_2H^+$), 218 (15, $MNa^+ - HCl$), 196 (100, $MH^+ - HCl$), 179 (20, $MH^+ - HCl, - OH$).

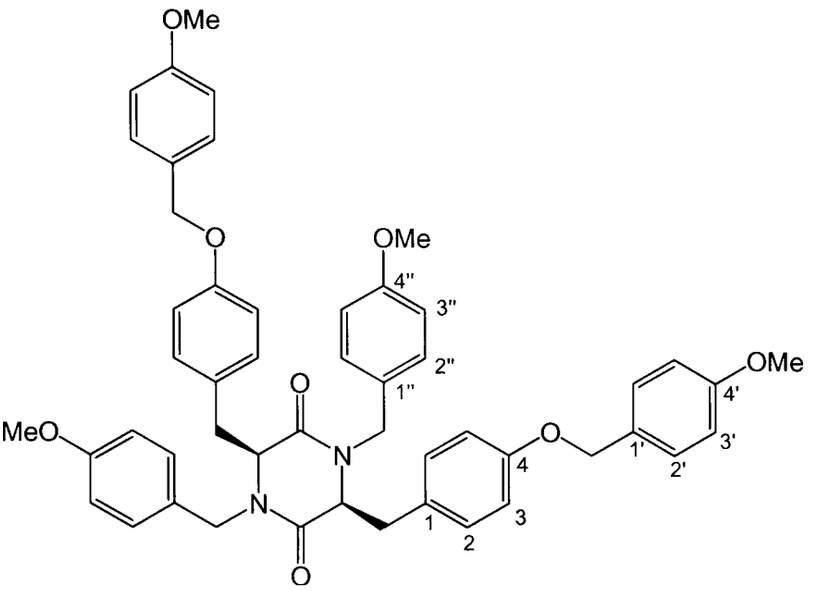
Chiral hplc (gradient elution: 10 % i-propanol for 10 minutes 10 to 30 % i-propanol over 2 minutes, then 30 to 50 % i-propanol in hexane over 30 minutes; eluting 0.5 ml/minute from a Chiracel OJ 250 × 4.6 mm cellulose derivative silica gel column) determined that the ester **33** had been synthesised in >98 % ee.

_N_-_t_-Butoxycarbonyl-L-tyrosine methyl ester 36



Triethylamine (7.5 ml, 53.9 mmol) was added to a stirred solution/suspension of the methyl ester **33** (4.993 g, 21.57 mmol) in anhydrous DCM (100 ml). The mixture was cooled to 0 °C and di-*t*-butyl dicarbonate (5.5 ml, 24.0 mmol) was added. The reaction mixture was then stirred for 30 minutes at 0 °C, and for 24 hours at room temperature. The reaction mixture was poured into water (50 ml), the layers were separated and the aqueous fraction extracted with DCM (3 × 20 ml). The combined organic extracts were washed with water (30 ml), aqueous hydrochloric acid (2 M; 30 ml) and brine (30 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 25 to 30 % ethyl acetate in petrol) to give the protected tyrosine **36** (6.076 g, 95 %) as a colourless solid, m.p. 102-104 °C (lit.[193] 101-105 °C); R_f 0.39 (30 % ethyl acetate in petrol); $[\alpha]_D$ +7.9 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3380 (OH & NH), 1714 (ester C=O), 1690 (carbamate C=O), 1616 (Ar), 1597 (Ar), 1519 (Ar), 1275 and 1158; δ_H (300 MHz; chloroform-*d*) 6.95 (2 H, d, *J* 8.4, 2 × 3-H), 6.90 (1 H, br s, OH), 6.74 (2 H, d, *J* 8.4, 2 × 2-H), 5.10 (1 H, d, *J* 8.2, NH), 4.54 (1 H, d app. t, *J* 8.2 and 5.8, CH), 3.70 (3 H, s, Me), 3.03 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 5.8, CH_AH$_B$), 2.96 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 5.8, CH$_A$$H_B$) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 173.2 (*C*OOMe), 155.8 & 155.8 (C4 & NHCO), 130.7 (C3), 127.3 (C1), 116.0 (C2), 80.8 (*C*Me$_3$), 55.1 (CH), 52.8 (COO*Me*), 37.9 (CH_2) and 28.7 (C*Me*$_3$); *m/z* (EI) 239 (20 %, MH^+ – *t*-Bu), 222 (58, MH^+ – *t*-Bu, – OH), 212 (30), 195 (13, MH^+ – Boc), 178 (40, MH^+ – Boc, – OH) and 107 (100).

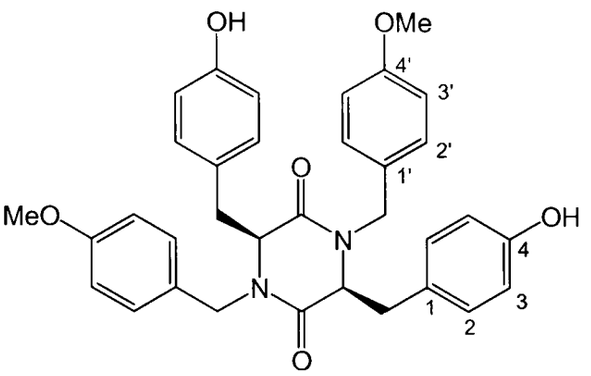
_O,O',N,N'_-Tetra-(_p_-methoxybenzyl)-cyclo(L-tyrosine)-(L-tyrosine) 41



Sodium hydride (60 % dispersion in mineral oil; 136 mg, 3.40 mmol), TBAI (57 mg, 0.15 mmol) and PMB chloride (470 µl, 3.45 mmol) were added to a stirred solution of the diketopiperazine **19a** (249 mg, 0.76 mmol) in anhydrous DMF (12 ml) and the reaction mixture was stirred at room temperature for 19 hours. The reaction mixture was then concentrated (oil pump pressure) and the residue partitioned between ethyl acetate (10 ml) and water (10ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 40 % ethyl acetate in petrol) to give the *protected diketopiperazine* **41** (160 mg, 26 %) as a colourless solid, m.p. 43-45 °C; R_f 0.26 (40 % ethyl acetate in petrol); $[\alpha]_D$ –99.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (solid phase) 3034-2836 (CH), 1661 (C=O), 1610 (Ar), 1585 (Ar), 1510 (Ar), 1546 (Ar), 1241 (ArOR), 1035 and 823; δ_H (300 MHz; chloroform-*d*) 7.32 (4 H, d, *J* 8.7, 4 × 2'-H), 7.03 (4 H, d, *J* 8.7, 4 × 2-H), 6.95 (4 H, d, *J* 8.7 4 × 3-H), 6.87 (4 H, d, *J* 8.5, 4 × 2"-H), 6.86 (4 H, d, *J* 8.7, 4 × 3'-H), 6.77 (4 H, d, *J* 8.5, 4 × 3"-H), 5.28 (2 H, d, $^2J_{HH}$ 14.5, 2 × NCH_AH$_B$Ar), 4.98 (4 H, s, 2 × OCH_2Ar), 4.05 (2 H, dd, *J* 6.8 and 4.4, 2 × CH), 3.79 (6 H, s, 2 × OMe), 3.78 (6 H, s, 2 × OMe), 3.64 (2 H, d, $^2J_{HH}$ 14.5, 2 × NCH$_A$$H_B$Ar), 2.87 (2 H, dd, $^2J_{HH}$ 14.3 and *J* 4.4, CHCH_AH$_B$) and 2.38 (2 H, dd, $^2J_{HH}$ 14.3 and *J* 6.8, CHCH$_A$$H_B$); δ_C (75 MHz; chloroform-*d*) 166.2 (C=O), 159.4 & 159.3 (C4' & C4"), 158.1 (C4), 130.6 (C2), 129.5 (C2"), 129.2 (C1), 129.1 (C2'), 128.8 (C1'), 127.4 (C1"), 115.4 (C3), 114.1 & 114.0 (C3' & C3"), 69.8 (OCH_2), 60.3 (CH), 55.3 (OMe), 55.3 (OMe), 46.6 (NCH_2) and 38.2 ($CHCH_2$); *m/z* (EI) 806 (4 %, M^+), 700 (14, MH^+ – C_6H_4OMe), 580 (51, MH_2^+ – C_6H_4OMe, – $CH_2C_6H_4OMe$), 460 (54, MH_3^+ – C_6H_4OMe, – 2($CH_2C_6H_4OMe$)), 353 (18, MH_4^+ – 2(C_6H_4OMe), – 2($CH_2C_6H_4OMe$)),

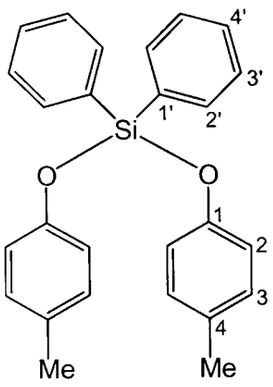
227 (31) and 121 (100, $MeOC_6H_4CH_2^+$); HRMS (ES) found: 807.3621; $C_{50}H_{51}N_2O_8$ requires MH^+ 807.3645.

_N,N'_-Bis(_p_-methoxybenzyl)-cyclo(_O_-methyl-L-tyrosine)-(_O_-methyl-L-tyrosine) 43



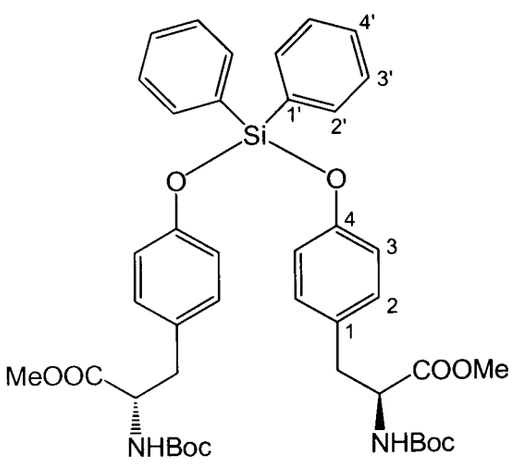
DDQ (65 mg, 0.29 mmol) was added to a stirred solution of the tetrakis-protected diketopiperazine **41** (100 mg, 0.12 mmol) in DCM (1.3 ml) and water (0.1 ml). The reaction mixture was stirred at room temperature for 6 hours, after which time a further portion of DDQ (62.mg, 0.027). The reaction mixture was then stirred for a further 16 hours and aqueous sodium bicarbonate (5 ml) was added to the reaction mixture. The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 2 ml). The combined organic extracts were washed with aqueous sodium bicarbonate (2 ml) and brine (2 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 50 % ethyl acetate in petrol) to give the *bis-protected diketopiperazine* **43** (31 mg, 45 %) as colourless prisms, m.p. 88-90 °C; R_f 0.13 (50 % ethyl acetate in petrol); $[\alpha]_D$ 8.0 (*c* 0.1 in methanol); ν_{max}/cm^{-1} (solid phase) 3310(OH), 2931 (CH), 1645 (C=O), 1611 (Ar), 1511 (Ar), 1463 (Ar), 1340 (ArOH), 1243 (ArOR), 1174 (ArOH), 1023 and 832; δ_H (300 MHz; chloroform-*d*) 6.95 (4 H, d, *J* 8.2, 4 × 2-H or 2'-H), 4.88 (4 H, d, *J* 8.7, 4 × 2-H or 2'-H), 6.81 (4 H, d, *J* 7.7, 4 × 3-H or 3'-H), 6.78 (4 H, d, *J* 8.2, 4 × 3-H or 3'-H), 5.31 (2 H, d, $^2J_{HH}$ 14.9, 2 × NCH_AH_BAr), 4.08 (2 H, dd, *J* 7.2 and 3.6, 2 × $CHCH_2$), 3.36 (2 H, $^2J_{HH}$ 14.9, 2 × NCH_AH_BAr), 2.93 (2 H, dd, $^2J_{HH}$ 14.3 and *J* 3.6, 2 × $CHCH_AH_B$) and 2.27 (2 H, dd, $^2J_{HH}$ 14.3 and 7.2, 2 × $CHCH_AH_B$); δ_C (75 MHz; chloroform-*d*) 164.3 & 163.0 (C=O & C4), 160.2 (C4'), 133.4 & 130.7 (C2 & C2'), 126.6 & 126.1 (C1 & C1'), 116.2 & 114.8 (C3 & C3'), 66.3 (CH), 55.8 (OMe), 49.3 (NCH_2) and 30.1 ($CHCH_2$); *m/z* (ES) 687 (40 %, M^+ + PMB), 589 (15, MNa^+), 567 (100, MH^+), 459 (20, $M^+ - MeOC_6H_4$) and 338 (40, $M^+ - MeOC_6H_4$, $- MeOC_6H_4CH_2$); HRMS (ES) found: M^+ 567.2473; $C_{34}H_{34}N_2O_6$ requires MH^+ 567.2495.

Bis(phenyl) bis(*p*-tolyloxy)silane 44



Imidazole (345 mg, 5.07 mmol) was added to a stirred solution of *p*-cresol **20** (506 mg, 4.69 mmol) in anhydrous DCM (10 ml). The mixture was cooled to 0 °C and dichlorodiphenylsilane (490 µl, 2.33 mmol) was added. The reaction mixture was allowed to warm to 15 °C over 90 minutes, poured into water (10 ml), the layers separated and the aqueous layer extracted with DCM (2 × 10 ml). The combined organic extracts were washed with water (5 ml) and brine (5 ml), dried ($MgSO_4$) and concentrated to give the siloxane **44** (866 mg, 94 %) as a yellow oil; R_f 0.56 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3071-2861 (CH), 1611 (Ar), 1592 (Ar), 1508 (Ar), 1429, 1263 (ArOR), 1127, 1061 (SiO), 945 and 821; δ_H (300 MHz; chloroform-*d*) 7.77-7.74 (4 H, m, 4 × 2'-H), 7.46-7.33 (6 H, m, 4 × 3'-H & 2 × 4'-H), 6.95 (4 H, d, *J* 8.6, 4 × 3-H), 6.86 (4 H, d, *J* 8.6, 4 × 2-H), and 2.22 (6 H, s, 2 × Me); δ_C (75 MHz; chloroform-*d*) 152.2 (C1), 135.4 (C2'), 132.0 (C1'), 131.7 (C4), 131.2 (C4'), 130.4 (C3), 128.4 (C3'), 119.9 (C2) and 21.0 (Me); *m/z* (EI) 396 (14 %, M^+), 289 (100, $M^+ - OC_6H_4Me$) and 91 (47, $C_7H_7^+$).

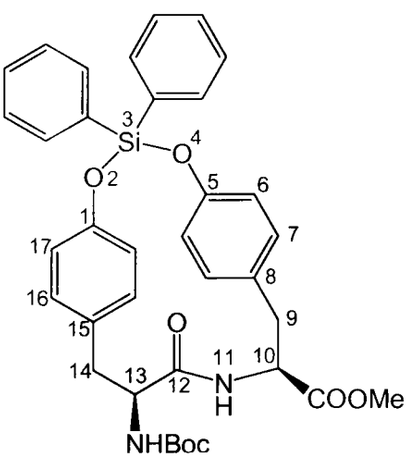
Diphenyl bis(*O*-[*N*-*t*-butoxycarbonyl]-L-tyrosinyl methyl ester)-silane 45



Imidazole (127 mg, 1.86 mmol), dichlorodiphenylsilane (180 µl, 0.85 mmol) and a catalytic quantity of DMAP were added to a stirred solution of the protected tyrosine **36** (509 mg, 1.72 mmol), in anhydrous DCM (10 ml). The reaction mixture was stirred for 17 hours at room temperature, then poured into water (10 ml). The layers were separated and the aqueous fraction extracted with DCM (3 × 5 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by reverse phase hplc (gradient elution: 10 to 90 % acetonitrile in water over 40 minutes; eluting at 20 ml/minute from a Supelco Discovery C18 250 × 21.2 mm 5 µm column) to give the *silane* **45** (366 mg, 28 %) as a colourless oil; R_f 0.25 (30 % ethyl acetate in petrol); $[\alpha]_D$ +3.2 (*c* 1.0 in methanol); ν_{max} cm^{-1} (nujol mull) 3357 (NH), 1746 (ester

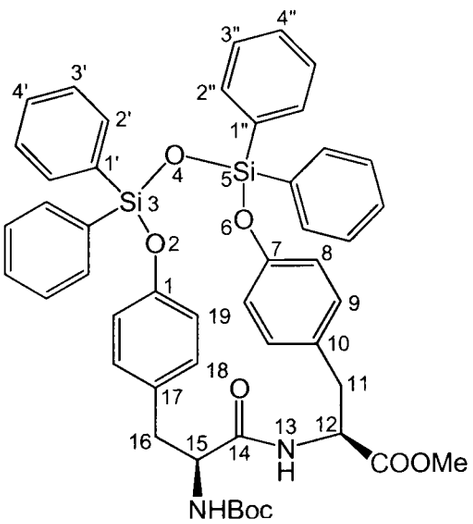
C=O), 1712 (carbamate C=O), 1609 (Ar), 1505 (Ar), 1264 (ArOR), 1160, 1056 (SiO) and 919; δ_H (300 MHz; chloroform-*d*) 7.73 (4 H, d, *J* 7.6, 4 × 2'-H), 7.43 (2 H, t, *J* 7.6, 2 × 4'-H), 7.36 (4 H, app t, *J* 7.6, 4 × 3'-H), 6.92 (4 H, d, *J* 8.6, 4 × 2-H), 6.87 (4 H, d, *J* 8.6, 4 × 3-H), 4.99 (2 H, d, *J* 7.0, 2 × NH), 4.50 (2 H, app q, *J* 7.0, 1 × CH), 3.60 (6 H, s, 2 × Me), 2.98 (2 H, dd, $^2J_{HH}$ 13.9 and *J* 7.0 , 2 × CH_AH_B), 2.93 (2 H, dd, $^2J_{HH}$ 13.9 and *J* 7.0 , 2 × CH_AH_B) and 1.40 (18 H, s, 2 × *t*-Bu); δ_C (75 MHz; chloroform-*d*) 172.8 (*C*OOMe), 155.6 (NHCO), 153.4 (C4), 135.4 (C2'), 131.6 (C1), 131.4 (C4'), 130.8 (C2), 130.0 (C1'), 128.5 (C3'), 120.3 (C3), 80.4 (*C*Me$_3$), 54.9 (CH), 52.6 (Me), 38.0 (CH_2) and 28.7 (C*Me*$_3$); *m/z* (ES) 793 (35 %, MNa^+), 771 (45, MH^+); *m/z* (EI) 670 (<1 %, MH^+ – Boc), 597 (11 %, MH_2^+ – NHBoc, – COOMe), 508 (100, MH^+ – NHBoc, – COOMe, – OMe, – *t*-Bu), 395 (100 MH^+ – 2(BocNHCHCOOMe)), 289 (100, MH^+ – BocNHCHCOOMe, – $OC_6H_4CH_2CH(NHBoc)COOMe$), 213 (77, $PhSiOC_6H_4CH_3^+$) and 57 (53, *t*-Bu); HRMS (ES) found: 793.3131; $C_{42}H_{50}N_2O_{10}Si$ requires MNa^+ 793.3132.

(10*S*,13*S*)-*N*-(*t*-Butoxycarbonyl)-13-amino-12-oxo-3,3-diphenyl-2,4-dioxa-11-aza-3-sila-tricyclo [13.2.2.2^{5,8}] henicosa-1(18), 5(21), 6, 8(20), 15(19), 16-hexaene-10-carboxylic acid methyl ester 46 and N-(*t*-Butoxycarbonyl)-15-amino-14-oxo-3,3,5,5-tetraphenyl-2,4,6-trioxa-13-aza-3,5-disila-tricyclo [15.2.2.2^{7,10}] tricosa-1(20), 7(23), 8, 10(22), 17(21), 18-hexaene-12-carboxylic acid methyl ester 48



Imidazole (169 mg, 2.48 mmol) and a catalytic quantity of DMAP were added to a stirred solution of the dipeptide **18a** (508 mg, 1.11 mmol) in anhydrous DCM (20 ml). Dichlorodiphenylsilane (230 µl, 1.10 mmol) was added dropwise to the solution, and the reaction mixture was stirred at room temperature for 24 hours. The reaction mixture was poured into water (20 ml), the layers were separated and the aqueous fraction extracted with ethyl acetate (2 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by reverse-phase hplc (gradient elution: 40 to 90 % acetonitrile in water over 40 minutes; eluting at 20 ml/minute from a Supelco Discovery C18 250 × 21.2 mm 5 µm column) to give the

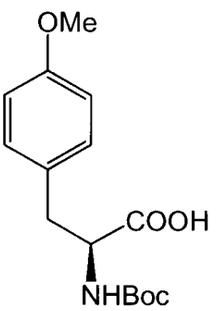
cyclic siloxane **46** (18 mg, 3 %) as a colourless solid, m.p. 52-54 °C; R_f 0.40 (30 % ethyl acetate in petrol); $[\alpha]_D$ +17.2 (*c* 1.0 in DCM); ν_{max}/cm^{-1} (solid phase) 3389br (NH), 3071-2855 (CH), 1733 (ester C=O), 1666br (carbamate C=O & amide C=O), 1613 (Ar), 1594 (Ar), 1513 (Ar), 1443 (Ar), 1368, 1258 (ArOR), 1117, 1053 (SiO), 920, 843 (Ar), 733 and 512; δ_H (500 MHz; chloroform-*d*) 7.71-7.16 (10 H, m, 2 × Ph), 6.94 (2 H, br s, 2 × 7-H or 16-H), 6.78 (2 H, d, *J* 7.8, 2 × 7-H or 16-H), 6.67 (2 H, d, *J* 7.8, 2 × 6-H or 17-H), 6.66 (2 H, d, *J* 7.8, 2 × 6-H or 17-H), 6.29 (1 H, br s, NH), 5.18 (1 H, br s, NH), 4.72 (1 H, br s, CH), 4.27 (1 H, br s, CH), 3.67 (3 H, s, Me), 2.98 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 4.7, CH_AH$_B$), 2.94 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 6.0, CH_AH$_B$), 2.91 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 6.4, CH$_A$$H_B$), 2.85 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 6.9, CH$_A$$H_B$) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.6 & 171.3 (*C*OOMe & CH*C*ONHCH), 155.6 & 155.2 (C1, C5 & CHNH*C*OO*t*-Bu), 134.9, 134.6, 134.4 & 134.3 (Ph), 130.7, 130.4, 130.3 & 130.1 (C7, C8, C15 & C16), 127.9, 127.8, 127.7 & 127.6 (Ph), 115.7 (C6 & C17), one peak missing (*C*Me$_3$), 53.4 (2 × CH), 55.4 (Me), 36.9 (2 × CH$_2$) and 28.3 (C*Me$_3$*); *m/z* (ES) 661 (10 %, MNa$^+$), 639 (7, MH$^+$), 583 (37, MH$_2^+$ – *t*-Bu), 539 (100, MH$_2^+$ – Boc) and 359 (80, MH$_4^+$ – Boc, – SiPh$_2$); HRMS (ES) found: 539.1998 and 661.2361; $C_{36}H_{38}N_2O_7Si$ requires *MH$_2^+$* – *Boc* 539.2002 and *MNa$^+$* 661.2346.



Also isolated was the *cyclic bis(siloxane)* **48** (6 g, 1 %) as a colourless solid, m.p. 53-55 °C; R_f 0.40 (30 % ethyl acetate in petrol); $[\alpha]_D$ +10.8 (*c* 0.5 in DCM); ν_{max}/cm^{-1} (solid phase) 3404 (NH), 3335 (NH), 3071-2850 (CH), 1740 (ester C=O), 1714 (carbamate C=O), 1677 (amide C=O), 1610 (Ar), 1592 (Ar), 1512 (Ar), 1247 (ArOR), 1079 (SiO), 937, 720 & 699; δ_H (500 MHz; chloroform-*d*) 7.54-7.42 (8 H, m, 2'-H & 2"-H), 7.31-7.26 (4 H, m, 4'-H & 4"-H), 7.22-7.14 (8 H, m, 3'-H & 3"-H), 6.77 (2 H, br s, 2 × 18-H), 6.65 (2 H, d, *J* 8.1, 2 × 8-H), 6.58 (2 H, d, *J* 8.1, 2 × 19-H), 6.53 (2 H, br s, 2 × 9-H), 5.76 (1 H, br s, N*H*CHCOOMe), 4.97 (1 H, br s, N*H*COO*t*-Bu), 4.67 (1 H, d app t, *J* 8.6 and 6.0, C*H*COOMe), 4.26 (1 H, m, C*H*NHCOO*t*-Bu), 3.56 (3 H, s, Me), 3.18 (1 H, d, $^2J_{HH}$ 13.9, CH_AH$_B$CHNHCOO*t*-Bu), 2.83 (2 H, m, CH_2CHCOOMe), 2.48 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 8.6 CH$_A$$H_B$CHNHCOO*t*-Bu) and 1.36 (9 H, s, *t*-Bu) ; δ_C (125 MHz; chloroform-*d*) 171.5 (*C*OOMe), 171.1

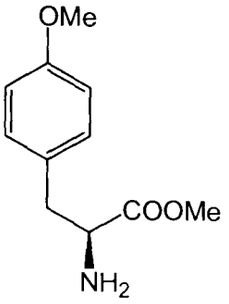
(CHCONHCH), 155.3 (NHCOO*t*-Bu), 153.1 (C1 & C7), 134.7 (C2' & C2"), 132.6 & 132.4 (C1' & C1"), 130.7 & 130.6 (C4' & C4"), 130.3 (C18), 129.7 (C9), 128.5 (C10 & C17), 128.0 & 127.9 (C3' & C3"), 120.0 & 119.9 (C8 & C19), 80.2 (*C*Me$_3$), 55.3 (*C*HNHCOO*t*-Bu), 52.5 (*C*HCOOMe), 52.3 (Me), 33.4 (*C*H$_2$CHNHCOO*t*-Bu), 31.9 (*C*H$_2$CHCOOMe) and 28.4 (C*Me$_3$*); *m/z* (ES) 860 (5 %, MNa$^+$), 854 (20, MH$_2$O$^+$) and 837 (53, MH$^+$); HRMS (ES) found: M$^+$ 737.2514; $C_{48}H_{48}N_2O_8Si_2$ requires *MH$_2^+$ – Boc* 737.2503.

N-t-Butoxycarbonyl-*O*-methyl-L-tyrosine 49



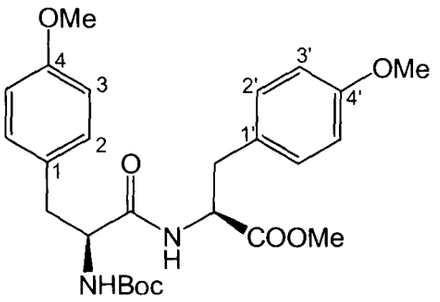
Sodium hydride (60 % dispersion in mineral oil; 655 mg, 16.38 mmol) was added to a stirred solution of the carbamate **16** (2.013 g, 7.16 mmol) in anhydrous DMF (30 ml) at 0 °C. The reaction mixture was stirred for 1 hour at 0-5 °C, and methyl iodide (445 µl, 7.15 mmol) was added. The reaction mixture was stirred for a further 4 hours at 0-5 °C, then diluted with ice-water (60 ml) and ethyl acetate (40 ml). The layers were separated and the organic fraction discarded. The aqueous fraction was acidified using aqueous hydrochloric acid (2 M) and extracted with ethyl acetate (4 × 40 ml). The combined organic extracts were washed with aqueous hydrochloric acid (2 M; 2 × 30 ml), and brine (30 ml), dried ($MgSO_4$) and concentrated to give the methyl ether **49** (1.480 g, 70 %) as a colourless solid, m.p. 90-92 °C (lit.[194] 49-50 °C); R_f 0.32 (30 % ethyl acetate & 2 % acetic acid in petrol); $[\alpha]_D$ +24 (*c* 0.4 in methanol); Found: C, 61.0; H, 7.15; N, 4.45; $C_{15}H_{21}NO_5$ requires C, 61.0; H, 7.17; N, 4.74 %; ν_{max}/cm^{-1} (nujol mull) 3380 (NH), 2854 (COOH), 1721 (acid C=O), 1684 (carbamate C=O), 1613 (Ar), 1516 (NH/CN) and 1251 (ArOR); δ_H (300 MHz; methanol-d_4) 6.97 (2 H, d, *J* 8.6, 2 × 2-H), 6.66 (2 H, d, *J* 8.6, 2 × 3-H), 4.12 (1 H, dd, *J* 8.9 and 5.1, CH), 3.58 (3 H, s, Me), 2.91 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 5.1, C*H$_A$*H$_B$), 2.67 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 8.9, CH$_A$*H$_B$*) and 1.21 (9 H, s, *t*-Bu); δ_C (75 MHz; methanol-d_4) 175.9 (COOH), 160.4 (C4), 158.2 (NHCO), 131.7 (C2), 130.9 (C3), 80.9 (*C*Me$_3$), 56.9 (CH), 56.0 (Me), 38.3 (CH$_2$) and 29.1 (C*Me$_3$*); *m/z* (EI) 295 (1 %, M$^+$), 239 (3, MH$^+$ – *t*-Bu), 193, (1, M$^+$ – BocH), 178 (6, M$^+$ – NH$_2$Boc), 149 (6, M$^+$ – COOH, – Boc), 121 (100, MeOC$_6$H$_4$CH$_2^+$) and 77 (7, C$_6$H$_5^+$).

O-Methyl-L-tyrosine methyl ester 51



TFA (5 ml) was added to a stirred solution of the protected tyrosine **50** (3.051 g, 9.86 mmol) in DCM (20 ml). The reaction mixture was stirred for 4 hours at room temperature, then concentrated under reduced pressure. The residue was dissolved in DCM (30 ml) and washed with aqueous hydrochloric acid (2 M; 10 ml), aqueous sodium bicarbonate (10 %; 10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give the amine **51** (1.279 g, 62 %) as a yellow powder, m.p. 190-192 °C; R_f 0.86 (methanol); $[\alpha]_D$ −26.4 (*c* 0.5 in DMSO); ν_{max}/cm^{-1} (nujol mull) 3593-2316 (NH_2), 1743 (C=O), 1659 (NH_2), 1612 (Ar), 1583 (Ar), 1513 (Ar), 1248 (ArOR), 1033 and 835; δ_H (300 MHz; methanol-d_4) 7.03 (2 H, d, *J* 8.7, 2 × 2-H), 6.79 (2 H, d, *J* 8.7, 2 × 3-H), 3.75 (1 H, app. t, *J* 6.6, CH), 3.68 (3 H, s, ArO*Me*), 3.62 (3 H, s, COOMe), 2.93 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 6.6, CH_AH_B) and 2.84 (1 H, dd, $^2J_{HH}$ 13.9 and *J* 6.6, CH_AH_B); δ_C (75 MHz; methanol-d_4) 175.0 (C=O), 160.7 (C4), 131.7 (C2), 129.5 (C1), 115.4 (C3), 56.6 (CH), 56.0 (ArO*Me*), 53.0 (COO*Me*) and 39.9 (CH_2); *m/z* (ES) 419 (7 %, M_2H^+), 387 (42, M_2^+ − OMe), 288 (25), 251 (38), 210 (100, MH^+) and 193 (19, $M^+ - NH_2$).

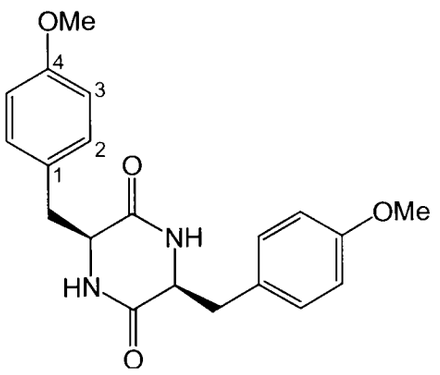
(*N-t*-Butoxycarbonyl-*O*-methyl-L-tyrosine)-(*O*-methyl-L-tyrosine methyl ester) 52



The carboxylic acid **49** (1.002 g, 3.40 mmol), HOBt (459 mg, 3.40 mmol) and a catalytic quantity of DMAP were added to a stirred solution/suspension of DCC (697 mg, 3.38 mmol) and the amine **50** (713 mg, 3.41 mmol) in anhydrous THF (32 ml). The reaction mixture was stirred for 24 hours at room temperature and then kept without stirring for 18 hours at −18 °C. The mixture was filtered under reduced pressure and the filtrate concentrated. The residue was partitioned between ethyl acetate (20 ml) and water (10 ml), the layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with water (10 ml), aqueous hydrochloric acid (2 M; 10 ml), water (10 ml), aqueous sodium bicarbonate (10 %; 10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 30 to 35 % ethyl acetate in petrol) to give the *dipeptide* **52** (832 mg,

50 %) as a colourless powder, m.p. 117-119 °C; R_f 0.16 (30 % ethyl acetate in petrol); $[\alpha]_D$ –0.4 (*c* 1.0 in methanol); Found: C, 64.2; H, 6.95; N, 6.0; $C_{26}H_{34}N_2O_5$ requires C, 64.2; H, 7.04; N, 5.8 %; ν_{max}/cm^{-1} (nujol mull) 3323 (NH), 1746 (ester C=O), 1688 (carbamate C=O), 1659 (amide C=O), 1614 (Ar), 1584 (Ar), 1515 (Ar), 1240 (Ar), 1176 and 1033; δ_H (300 MHz; chloroform-*d*) 7.11 (2 H, d, *J* 8.6, 2 × 2-H or 2'-H), 6.90 (2 H, d, *J* 8.7, 2 × 2-H or 2'-H), 6.82 (2 H, d, *J* 8.6, 2 × 3-H or 3'-H), 6.77 (2 H, d, *J* 8.7, 2 × 3-H or 3'-H), 6.28 (1 H, d, *J* 6.7, NH), 5.00 (1 H, br d, *J* 6.2, NH), 4.74 (1 H, d app. t, *J* 6.7 and 5.6, CH), 4.29 (1 H, br d, *J* 6.2, CH), 3.77 (3 H, s ArO*Me*), 3.77 (3 H, s, ArO*Me*), 3.67 (3 H, s, COOMe), 3.00-2.96 (4 H, m, 2 × CH_2), and 1.41 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.9 & 171.3 (*C*OOMe & CH*C*ONHCH), 159.1 (C4 & C4'), 156.7 (CH*C*OO*t*-Bu), 130.8 & 130.6 (C2 & C2'), 128.8 & 127.9 (C1 & C1'), 114.5 & 114.3 (C3 & C3'), 80.6 (*C*Me$_3$), 56.2 (CH), 55.6 (ArO*Me*), 55.6 (ArO*Me*), 53.8 (CH), 52.7 (COO*Me*), 37.8 (CH_2), 37.5 (CH_2) and 28.7 (C*Me$_3$*); *m/z* (ES) 995 (50 %, M_2Na^+), 509 (95, MNa^+), 487 (43, MH^+), 431 (97, MH_2^+ – *t*-Bu), 387 (100, MH_2^+ – Boc) and 225 (45).

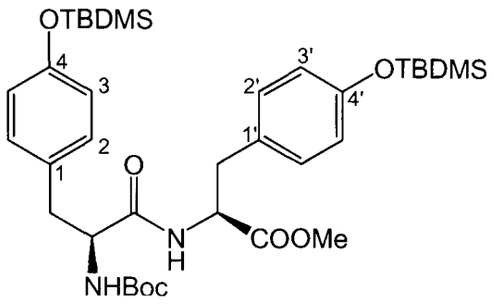
Cyclo(*O*-methyl-L-tyrosine)-(*O*-methyl-L-tyrosine) 53



A solution of the dipeptide **52** (733 mg, 1.651 mmol), in formic acid (38 ml) was stirred at room temperature for 2 hours. The reaction mixture was concentrated, the residue dissolved in *s*-butanol (38 ml) and toluene (16 ml) and heated at reflux for 22 hours. The mixture was concentrated and the residue dispersed in toluene (25 ml). The suspension was filtered to give the *diketopiperazine* **53** (0.471 g, 81 %) as a colourless solid, m.p. >290 °C; R_f 0.75 (methanol); $[\alpha]_D$ –88.0 (*c* 0.1 in DMSO); Found: C, 67.9; H, 6.2; N, 7.6; $C_{20}H_{22}N_2O_4$ requires C, 67.8; H, 6.3; N, 7.9 %; ν_{max}/cm^{-1} (nujol mull) 3213 (NH), 1674, 1660 (C=O), 1612 (Ar), 1583 (NHCN), 1513 (Ar), 1246 (ArOR) & 1036; δ_H (300 MHz; DMSO-d_6) 7.82 (2 H, d, *J* 1.9, 2 × NH), 6.89 (4 H, d, *J* 8.6, 4 × 2-H), 6.79 (4 H, d, *J* 8.6, 4 × 3-H), 3.86 (2 H, app td, *J* 5.5 and 1.9, 2 × CH), 3.64 (6 H, s, 2 × OMe), 2.49 (2 H, dd, $^2J_{HH}$ 13.8 and *J* 5.5, 2 × CH_AH$_B$) and 2.14 (2 H, dd, $^2J_{HH}$ 13.8 and *J* 5.5, 2 × CH$_A$$H_B$); δ_C (75 MHz; DMSO-d_6) 166.6 (NHCO),

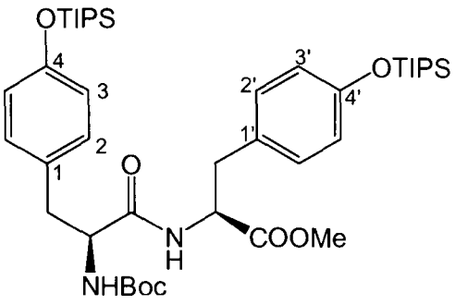
158.3 (C4), 131.2 (C2), 128.7 (C1), 114.0 (C3), 55.9 (CH), 55.3 (OMe) and 38.8 (CH_2); *m/z* (EI) 354 (17 %, M^+) and 121 (100, $MeOC_6H_4CH_2^+$).

(*N*-*t*-Butoxycarbonyl-*O*-*t*-butyldimthylsilyl-L-tyrosine)-(*O*-*t*-butyldimethylsilyl-L-tyrosine methyl ester) 54



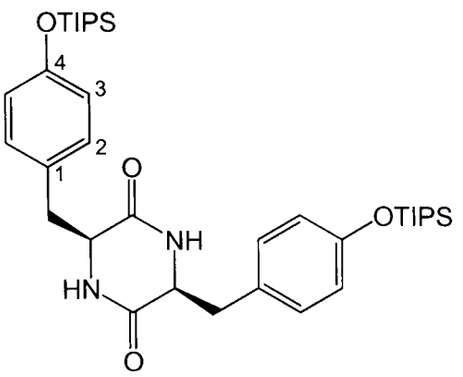
TBDMS chloride (728 mg, 4.82 mmol), imidazole (377 mg, 5.54 mmol) and a catalytic quantity of DMAP were added to a stirred solution of the dipeptide **18a** (993 mg, 2.17 mmol) in anhydrous DCM (30 ml). The reaction mixture was stirred at room temperature for 48 hours, then poured into water (20 ml). The layers were separated and the aqueous fraction extracted with DCM (3 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10ml), dried ($MgSO_4$) and concentrated to give the *bis(siloxane)* **54** (1.348 g, 91 %) as a colourless oil; R_f 0.58 (30 % ethyl acetate in petrol); $[\alpha]_D$ –2.6 (*c* 2.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3306 (NH), 1747 (ester C=O), 1683 (carbamate C=O), 1658 (amide C=O), 1610 (Ar), 1511 (Ar), 1255 (ArOR), 1171 and 917; δ_H (300 MHz; benzene-d_6) 6.92 (2 H, d, *J* 8.5, 2 × 2-H or 2'-H), 6.86 (2 H, d, *J* 8.5, 2 × 2-H or 2'-H), 6.70 (3 H, d, *J* 8.5, 2 × 3-H or 3'-H & NH), 6.66 (2 H, d, *J* 8.5, 2 × 3-H or 3'-H), 5.26 (1 H, d, *J* 7.2, NH), 4.82 (1 H, d app. t, *J* 6.9 and 5.8, CH), 4.43 (1 H, br d, *J* 7.2, CH), 3.13 (3 H, s, Me), 2.96 (2 H, dd, $^2J_{HH}$ 13.6 and *J* 6.4, 2 × CH_AH$_B$), 2.84 (2 H, dd, $^2J_{HH}$ 13.6 and *J* 5.8, 2 × CH$_A$$H_B$), 1.28 (9 H, s, NHCOO*t*-Bu), 0.88 (9 H, s, Si*t*-Bu), 0.88 (9 H, s, Si*t*-Bu), 0.00 (6 H, s, $SiMe_2$) and –0.01 (6 H, s, $SiMe_2$); δ_C (75 MHz; benzene-d_6) 172.0 (*C*OOMe), 171.7 (CH*C*ONHCH), 156.0 (NH*C*OO*t*-Bu), 155.6 & 155.1 (C4 & C4'), 131.1 (C2 & C2'), 130.4 & 129.7 (C1 & C1'), 120.7 (C3 & C3'), 79.9 (O*C*Me$_3$), 56.6 (CH), 54.3 (CH), 51.9 (Me), 38.1 (CH_2), 37.9 (CH_2), 26.3 (SiC*Me$_3$*), 26.1 (SiC*Me$_3$*), 18.6 (Si*C*Me$_3$), 18.6, (Si*C*Me$_3$), –3.0 ($SiMe_2$) and –4.1 ($SiMe_2$); *m/z* (EI) 687 (<1 %, M^+), 556 (1, M^+ – OTBDMS), 292 (33), 262 (17, $TBDMSOC_6H_4CH_2CHCO^+$), 221 (82, $TBDMSOC_6H_4CH_2^+$), 192 (12, $TBDMSOC_6H_5^+$), 147 (44), 110 (32) and 75 (100); *m/z* (ES) 687 (62 %, MH^+) and 101 (100); HRMS (ES) found: 687.3854; $C_{36}H_{58}N_2O_7Si_2$ requires *MH*$^+$ 687.3861.

(*N*-*t*-Butoxycarbonyl-*O*-triisopropylsilyl-L-tyrosine)-(*O*-triisopropylsilyl-L-tyrosine methyl ester) 56



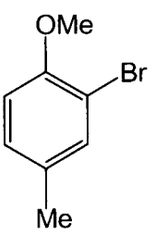
TIPS chloride (1.1 ml, 5.14 mmol) was added to a stirred solution of the dipeptide **18a** (499 mg, 1.09 mmol), imidazole (1.87 mg, 2.74 mmol) and a catalytic quantity of DMAP in anhydrous DCM (15 ml). The reaction mixture was stirred at room temperature for 19 hours, then poured into water and the layers separated. The aqueous fraction was extracted with DCM (2 × 10 ml) and the combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was then purified by flash chromatography (eluent: 15 % ethyl acetate in petrol) to give the *bis(siloxane)* **56** (489 mg, 58 %) as a colourless powder, m.p. 114-116 °C; R_f 0.19 (15 % ethyl acetate in petrol); $[\alpha]_D$ –2.0 (*c* 1 in methanol); Found: C, 65.6; H, 9.05; N, 3.7; $C_{42}H_{70}N_2O_7Si_2$ requires C, 65.41; H, 9.15; N, 3.63 %; ν_{max}/cm^{-1} (nujol mull) 3394 (NH), 3284 (NH), 1748 (ester C=O), 1710 (carbamate C=O), 1653 (amide C=O), 1609 (Ar), 1570 (NH/CN), 1510 (Ar), 1267 (ArOR), 1174, 1057 (SiO) & 917; δ_H (500 MHz; chloroform-*d*) 7.03 (2 H, d, *J* 8.5, 2 × 2-H or 2'-H), 6.85 (2 H, d, *J* 8.5, 2 × 2-H or 2'-H), 6.80 (2 H, d, *J* 8.5, 2 × 3-H or 3'-H), 6.75 (2 H, d, *J* 8.5, 2 × 3-H or 3'-H), 6.28 (1 H, d, *J* 7.5, NH), 4.88 (1 H, br s, NH), 4.71 (1 H, d app. t, *J* 7.5 and 6.1, CH), 4.25 (1 H, m, CH), 3.63 (3 H, s, Me), 2.97 (2 H, dd, $^2J_{HH}$ 15.9 and *J* 7.1, 2 × CH_AH_B), 2.94 (2 H, dd, $^2J_{HH}$ 15.9 and *J* 6.1, 2 × CH_AH_B), 1.40 (9 H, s, *t*-Bu), 1.23 (3 H, septet, *J* 5.8, 3 × $CH(CH_3)_2$), 1.23 (3 H, septet, *J* 5.8, 3 × $CH(CH_3)_2$), 1.08 (18 H, s, 3 × $CH(CH_3)_2$) and 1.07 (18 H, s, 3 × $CH(CH_3)_2$); δ_C (125 MHz; chloroform-*d*) 171.5 (*C*OOMe), 170.8 (CH*C*ONHCH), 155.2 (NH*C*OO*t*-Bu, C4 & C4'), 130.3 & 130.2 (C2 & C2'), 128.0 (C1 & C1'), 120.1 & 120.0 (C3 & C3'), 80.1 (CMe_3), 53.5 (2 × CH), 52.2 (Me), 37.4 (2 × CH_2), 28.3 (CMe_3), 17.9 & 17.9 (6 × $CH(CH_3)_2$) and 12.7 (6 × SiCH); *m/z* (ES) 794 (5 %, MNa^+), 772 (60, MH^+), 715 (75, MH^+ – *t*-Bu), and 671 (100, MH_2^+ – Boc).

Cyclo(*O*-triisopropylsilyl-L-tyrosine)-(*O*-triisopropylsilyl-L-tyrosine) 57



A solution of the silylated dipeptide **56** (196 mg, 0.254 mmol), in formic acid (7 ml) was stirred at room temperature for 2 hours. The reaction mixture was then concentrated, the residue dissolved in *s*-butanol (7 ml) and toluene (3 ml) and heated at reflux for 18 hours. The mixture was concentrated and the residue partitioned between ethyl acetate (25 ml) and water (25 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (2 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give the *diketopiperazine* **57** (137 mg, 84 %) as a colourless solid, m.p. 212-214 °C; R_f 0.18 (ethyl acetate); $[\alpha]_D$ −680 (*c* 1.0 in DCM); ν_{max}/cm^{-1} (nujol mull) 3186 (NH), 1671 (C=O), 1608 (Ar), 1509 (Ar), 1266 (ArOR) and 919; δ_H (300 MHz; chloroform-*d*) 7.00 (4 H, d, *J* 8.4, 4 × 2-H), 6.86 (4 H, d, *J* 8.4, 4 × 3-H), 5.94 (2 H, d, *J* 2.3, 2 × NH), 4.10 (2 H, dt, *J* 8.5 and 2.3, 2 × CH), 3.03 (2 H, dd, $^2J_{HH}$ 13.8 and *J* 2.3, 2 × CH_AH_B), 2.33 (2 H, dd, $^2J_{HH}$ 13.8 and *J* 8.5, 2 × CH_AH_B), 1.23 (6 H, septet, *J* 7.7, 6 × $CH(CH_3)_2$) and 1.06 (36 H, d, *J* 7.7, 6 × $CH(CH_3)_2$); δ_C (75 MHz; chloroform-*d*) 166.9 (C=O), 156.0 (C4), 131.2 (C2), 127.7 (C1), 120.7 (C3), 56.9 (CH), 40.0 (CH_2), 18.3 ($CH(CH_3)_2$) and 13.1 ($CH(CH_3)_2$); *m/z* (ES) 639 (100 %, MH^+), 483 (13, MH^+ − TIPS) and 352 (5); HRMS (ES) found: 639.4000; $C_{36}H_{58}N_2O_4Si_2$ requires MH^+ 639.4013.

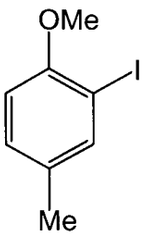
2-Bromo-4-methyl anisole 67



NBS (14.646 g, 82.28 mmol) was added to a stirred solution of 4-methyl anisole **66** (10.5 ml, 83.4 mmol) in acetone (164 ml). The mixture was stirred at room temperature for 45 minutes and aqueous hydrochloric acid (1 M; 820 µl) was added. The reaction mixture was then stirred for a further 18 hours at room temperature and concentrated. The residue was dispersed in petrol and filtered to remove succinimide. The filtrate was concentrated to give a crude product, which was then purified by flash chromatography (eluent: 3 % ethyl acetate in petrol) to give the aryl bromide **67** (15.788 g, 94 %) as a colourless oil; R_f 0.76 (20 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3004-2837 (CH), 1603 (Ar), 1497 (Ar), 1459 (Ar), 1286, 1254 (ArOR), 1055, 1023 & 804; δ_H (300 MHz;

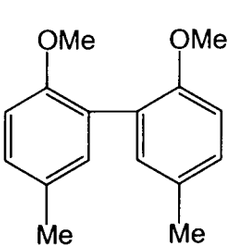
chloroform-*d*) 7.36 (1 H, d, $^4J_{HH}$ 1.9, 3-H), 7.05 (1 H, dd, *J* 8.3 and $^4J_{HH}$ 1.9, 5-H), 6.79 (1 H, d, *J* 8.3, 6-H), 3.86 (3 H, s, OMe) and 2.27 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 154.1 (C1), 134.2 (C3), 131.9 (C4), 129.3 (C5), 112.2 (C6), 111.7 (C2), 56.7 (OMe) and 20.6 (Me); *m/z* (EI) 200/202 (100 %, M^+), 185/187 (42, M^+ – Me), 121 (27, M^+ – Br) and 78 (46).

2-Iodo-4-methyl anisole 68



t-Butyllithium (1.05 M; 11.3 ml, 11.87 mmol) was added dropwise to a stirred solution of the aryl bromide **67** (720 µl, 5.42 mmol) in anhydrous THF (20 ml) at –78 °C. The reaction mixture was stirred for 1 hour at –78 °C, then a solution of iodine (4.25 g, 16.73 mmol) in anhydrous THF (30 ml) was added dropwise until an orange-brown colour persisted (~ 23 ml). The reaction mixture was stirred for a further hour at –78 °C. Excess *t*-butyllithium was quenched by addition of ethanol (10 ml), and aqueous sodium thiosulfate (2 M; 50 ml) was added to remove excess iodine. The mixture was extracted with DCM (3 × 20 ml) and the combined organic extracts washed with water (30 ml) and brine (30 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 5 % toluene in petrol) to give the aryl iodide **68** (983 mg, 73 %) as a yellow oil, R_f 0.63 (10 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2937 (C–H), 1601 (Ar), 1276, 1250 (ArOR) and 1050; δ_H (300 MHz; chloroform-*d*) 7.60 (1 H, d, $^4J_{HH}$ 1.5, 3-H), 7.09 (1 H, dd, *J* 8.5 and $^4J_{HH}$ 1.5, 5-H), 6.71 (1 H, d, *J* 8.5, 6-H), 3.84 (3 H, s, OMe) and 2.25 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 156.4 (C1), 140.2 (C3), 132.4 (C4), 130.4 (C5), 111.1 (C6), 86.1 (C2), 56.8 (OMe) and 20.4 (Me); *m/z* (EI) 248 (100 %, M^+), 233 (37, M^+ – Me), 199 (31), 127 (35), 106 (28, M^+ – Me, – I), 91 (33, M^+ – 2Me, – I) and 78 (37).

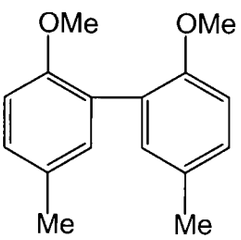
2,2'-Dimethoxy-5,5'-dimethyl biphenyl 69



Lithium hydroxide monohydrate (354 mg, 8.44 mmol) was added to a stirred solution of the biphenol **28** (892 mg, 4.17 mmol) in anhydrous THF (5 ml) and the reaction mixture stirred for 10 minutes at room temperature. Dimethyl sulfate (790 µl, 8.36 mmol) was added and the reaction mixture stirred for 90 minutes at room

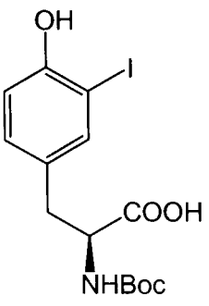
temperature. A further portion of dimethyl sulfate (200 μl, 2.12 mmol) was added and the reaction mixture stirred for a further 64 hours at room temperature. Aqueous sodium hydroxide (2 M; 10 ml) was added and the mixture stirred vigorously for 2 hours. The layers were separated and the aqueous fraction extracted with ether (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give the bis(methyl ether) **69** (700 mg, 69 %) as a cream-coloured solid, m.p. 56 °C (lit.[195] 63-64 °C); R_f 0.50 (10 % ethyl acetate in petrol); ν_{max} cm^{-1} (nujol mull) 1507 (Ar), 1377, 1237 (ArOR) and 807; δ_H (300 MHz; chloroform-*d*) 7.11 (2 H, dd, *J* 8.3 and $^4J_{HH}$ 2.2, 4-H & 4'-H), 7.03 (2 H, d, $^4J_{HH}$ 2.2, 6-H & 6'-H), 6.86 (2 H, d, *J* 8.3, 3-H & 3'-H), 3.74 (6 H, s, 2 × OMe) and 2.31 (6 H, s, 2 × Me); δ_C (75 MHz; chloroform-*d*) 155.4 (C2 & C2'), 132.4 (C6 & C6'), 129.9 (C5 & C5'), 129.3 (C4 & C4'), 128.2 (C1 & C1'), 111.5 (C3 & C3'), 56.34 (OMe) and 20.9 (Me); *m/z* (EI) 242 (100 %, M^+), 227 (45, M^+ – Me), 212 (77, M^+ – 2Me) and 195 (47, M^+ – Me, – MeOH).

2,2'-Dimethoxy-5,5'-dimethyl biphenyl 69



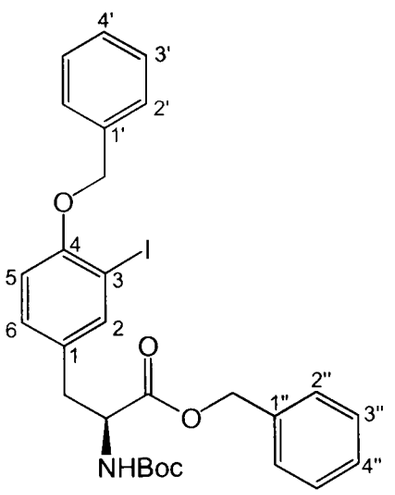
Anhydrous THF (1 ml) was added to a mixture of bis(triphenylphosphine)nickel(II) dibromide (153 mg, 0.21 mmol), zinc powder (43 mg, 0.66 mmol) and TBAI (148 mg, 0.40 mmol) and the mixture stirred for 30 minutes at room temperature. A solution of the aryl iodide **68** (107 mg, 0.43 mmol) in anhydrous THF (0.7 ml) was added, and the reaction mixture stirred at 50 °C for 24 hours. The reaction mixture was filtered through celite twice, and the filtrate poured into water and extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with water (15 ml) and brine (15 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 5 % ethyl acetate in petrol) to give the biphenyl **69** (29 mg, 56 %) as a colourless solid, spectroscopically identical to that obtained previously.

N-*t*-Butoxycarbonyl-3-iodo-L-tyrosine 71



A solution of iodine (905 mg, 3.56 mmol) in ethanol (15 ml) was added dropwise to a stirred solution of the carbamate **16** (1.006 g, 3.58 mmol) in aqueous ammonia (35 %; 65 ml) at 0 °C. The reaction mixture was stirred for a further 30 minutes at 0 °C, then slowly acidified using aqueous hydrochloric acid (conc.) with the temperature maintained at 0 °C. The resulting suspension was extracted with ethyl acetate (3 × 30 ml). The combined organic extracts were washed with aqueous sodium thiosulfate (2 M; 30 ml) and brine (30 ml), dried ($MgSO_4$) and concentrated to give the iodotyrosine **71** (1.260 g, 87 %) as a cream solid, m.p. 51-54 °C; R_f 0.49 (50 % ethyl acetate & 2 % acetic acid in petrol); $[\alpha]_D$ +15.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3318 (COOH, OH & NH), 1714 (acid C=O), 1684 (carbamate C=O), 1504 (NH/CN), 1413 (Ar) and 1159 (ArOH); δ_H (300 MHz; methanol-d_4) 7.58 (1 H, d, $^4J_{HH}$ 2.1, 2-H), 7.07 (1 H, dd, *J* 8.3 and $^4J_{HH}$ 2.1, 6-H), 6.78 (1 H, d, *J* 8.3, 5-H), 4.29 (1 H, dd, *J* 9.0 and 4.9, CH), 3.07 (1 H, dd, $^2J_{HH}$ 14.0 and *J* 4.9, CH_AH_B), 2.80 (1 H, dd, $^2J_{HH}$ 14.0 and *J* 9.0, CH_AH_B) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; methanol-d_4) 175.7 (COOH), 158.2 (NHCO), 157.3 (C4), 141.9 (C1), 141.5 (C2), 131.8 (C6), 116.0 (C5), 84.8 (C3), 81.0 (CMe_3), 56.7 (CH), 37.7 (CH_2) and 29.1 (CMe_3); *m/z* (EI) (<1 %, M^+), 359 (11), 290 (21, $M^+ - NH_2Boc$), 233 (100, $HOC_6H_3ICH_2^+$) and 57 (34, *t*-Bu^+).

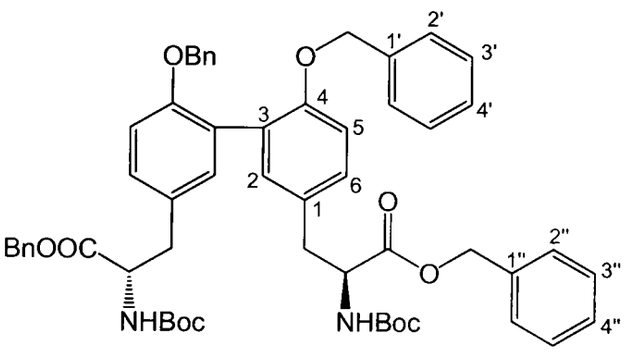
N-*t*-Butoxycarbonyl-*O*-benzyl-3-iodo-L-tyrosine benzyl ester 72



Potassium carbonate (891 mg, 6.55 mmol), TBAI (50 mg, 0.14 mmol) and benzyl bromide (800 µl, 6.73 mmol) were added to a stirred solution of the iodotyrosine **71** (1.055 g, 2.59 mmol) in anhydrous DMF (10 ml). The reaction mixture was stirred for 20 hours at room temperature, poured into water (15 ml) and extracted with ethyl acetate (3 × 15 ml). The combined organic extracts were washed with aqueous sodium bicarbonate (10 %; 20 ml), water (2 × 15 ml) and brine (15 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 10 to 15 % ethyl acetate in petrol) to give the *protected tyrosine* **72** (758 mg, 63 %) as a pale yellow oil; R_f 0.69 (20 % ethyl acetate in petrol);

$[\alpha]_D$ −2.9 (*c* 0.7 in methanol); Found: C, 57.6; H, 5.3; I, 21.4; N, 2.55; $C_{28}H_{30}INO_5$ requires C, 57.3; H, 5.15; I, 21.6; N, 2.3 %; ν_{max}/cm^{-1} (nujol mull) 3360 (NH), 1732 (ester C=O), 1681 (carbamate C=O), 1599 (Ar), 1521 (Ar), 1251 (ArOH), 1165, 1050 and 734; δ_H (500 MHz; chloroform-*d*) 7.54-7.29 (11 H, m, $ArOCH_2Ph$, $COOCH_2Ph$ & 2-H), 6.92 (1 H, d, *J* 8.1, 6-H), 6.68 (1 H, d, *J* 8.1, 5-H), 5.15 (1 H, d, $^2J_{HH}$ 12.2, $COOCH_AH_BPh$), 5.10 (1 H, d, $^2J_{HH}$ 12.2, $COOCH_AH_BPh$), 5.08 (2 H, s, $ArOCH_2Ph$), 5.01 (1 H, d, *J* 5.9, NH), 4.56 (1 H, d app. t, *J* 5.9 and 5.6, CH), 3.02 (1 H, dd, $^2J_{HH}$ 14.7 and *J* 5.6, CH_AH_B), 2.95 (1 H, dd, $^2J_{HH}$ 14.7 and *J* 5.6, CH_AH_B) and 1.43 (9 H, s, *t*-Bu); δ_C (125 MHz; chloroform-*d*) 171.9 (*C*OOBn), 156.7 (C4), 155.4 (NHCO), 140.7 (C2), 136.9 (C1'), 135.4 (C1"), 130.8 (C1), 130.7 (C6), 129.0 (C3' & C3"), 128.3 (C4' & C4"), 127.4 (C2' & C2"), 112.9 (C5), 87.2 (C3), 80.5 (*C*Me$_3$), 71.3 ($ArOCH_2Ph$), 67.7 ($COOCH_2Ph$), 54.9 (CH), 37.3 (CH_2) and 28.7 (C*Me$_3$*); *m/z* (EI) 470 (13 %, M^+ – NH_2Boc), 323 (17, $BnOC_6H_3ICH_2^+$), 179 (7) and 91 (100, $C_7H_7^+$);

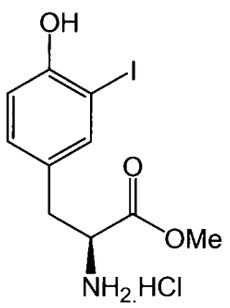
Di-(*N*-*t*-butoxycarbonyl-*O*-benzyl-L-tyrosine benzyl ester) 73



Anhydrous THF (12 ml) was added to a mixture of bis(triphenylphosphine)-nickel(II) bromide (1.259 g, 1.70 mmol), zinc powder (0.333 g, 5.12 mmol), TBAI (1.260 g, 3.42 mmol) and TMEDA (520 µl, 3.45 mmol) and the mixture stirred at room temperature for 40 minutes. A solution of the iodotyrosine **72** (1.992 g, 3.39 mmol) in anhydrous THF (8 ml), was added to the mixture, which was then stirred for 3 days at 50 °C. The reaction mixture was filtered through celite and washed through with ethyl acetate. The filtrate was poured into water (10 ml), the layers separated, and the aqueous fraction extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 20 % ethyl acetate in petrol) to give the *dityrosine* **73** (184 mg, 12 %) as a colourless solid, m.p. 47-49 °C; R_f 0.22 (20 % ethyl acetate in petrol); $[\alpha]_D$ +1.6 (*c* 0.5 in methanol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3437 (NH), 1738 (ester C=O), 1709 (carbamate C=O), 1499, 1244 (ArOR) and 1166; δ_H (300 MHz; chloroform-*d*) 7.33-

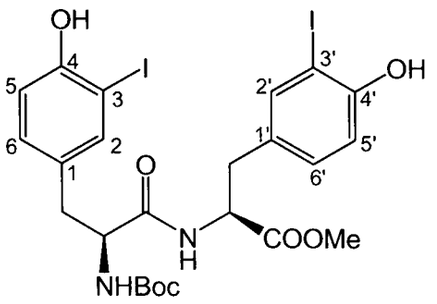
7.13 (20 H, m, 4 × CH_2Ph), 7.00 (2 H, d, $^4J_{HH}$ 2.1, 2 × 2-H), 6.95 (2 H, dd, J 8.2 and $^4J_{HH}$ 2.1, 2 × 6-H), 6.81 (2 H, d, J 8.2, 2 × 5-H), 5.13 (2 H, d, $^2J_{HH}$ 12.2, 2 × $COOCH_AH_BPh$), 5.05 (2 H, d, $^2J_{HH}$ 12.2, 2 × $COOCH_AH_BPh$), 5.02 (2 H, d, J 7.7, 2 × NH), 4.95 (4 H, s, 2 × $ArOCH_2Ph$), 4.59 (2 H, d app. t, J 7.7 and 6.2, 2 × CH), 3.04 (4 H, d, J 6.8, 2 × CH_2CH) and 1.37 (18 H, s, 2 × t-Bu); δ_C (75 MHz; chloroform-d) 172.3 ($COOBn$), 155.7 (C4), 155.6 (NHCO), 137.9 (C1'), 135.7 (C1"), 133.0 (C2) 129.7, 128.9, 128.9, 128.8, 127.6 & 127.0 (C1, C2', C2", C3', C3", C4', C4" & C6), 128.3 (C3), 113.5 (C5), 80.3 (CMe_3), 70.7 ($ArOCH_2Ph$), 67.4 ($COOCH_2Ph$), 55.0 (CH), 37.8 (CH_2CH) and 28.7 (CMe_3); *m/z* (ES) 945 (35 %, $MNaH^+$), 944 (58, MNa^+), 922 (54), 866 (40), 767 (52) and 766 (100); HRMS (ES) found: 921.4326; $C_{56}H_{61}N_2O_{10}$ requires MH^+ 921.435.

3-Iodo-L-tyrosine methyl ester hydrochloride 76



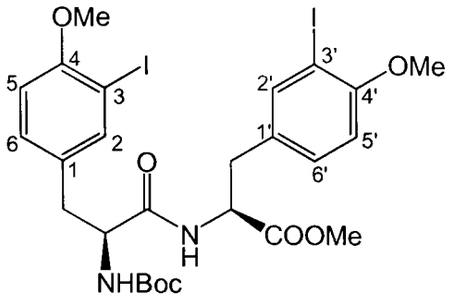
TMSCl (7.80 ml, 61.26 mmol) was added to a stirred solution of the carbamate **71** (4.918 g, 12.08 mmol) in anhydrous methanol (36 ml) and the reaction mixture stirred for 21 hours at room temperature. Methanol was removed *in vacuo* and the residue dispersed in ether (70 ml). The suspension was stirred for 30 minutes, then filtered to give the methyl ester **76** (3.776 g, 87 %) as a colourless solid, m.p. 191-193 °C (dec.; lit.[196] 194.5-197 °C); R_f 0.69 (50 % methanol in DCM); $[\alpha]_D$ +0.9 (*c* 3.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3584 (OH), 3271 (NH_2), 3221 (NH_2), 1746 (C=O), 1505, 1416 (PhOH), 1247 and 1218 (PhOH); δ_H (300 MHz; methanol-d_4) 7.63 (1 H, d, $^4J_{HH}$ 2.2, 2-H), 7.11 (1 H, dd, J 8.3 and $^4J_{HH}$ 2.2, 6-H), 6.86 (1 H, d, J 8.3, 5-H), 4.28 (1 H, dd, J 7.4 and 6.0, CH), 3.84 (3 H, s, Me), 3.17 (1 H, dd, $^2J_{HH}$ 14.3 and J 6.0, CH_AH_B) and 3.07 (1 H, dd, $^2J_{HH}$ 14.3 and J 7.4, CH_AH_B); δ_C (75 MHz; methanol-d_4) 170.8 ($COOMe$), 158.4 (C4), 141.6 (C2), 132.0 (C6), 128.2 (C1), 116.5 (C5), 85.6 (C3), 55.6 (CH), 54.0 (Me) and 36.4 (CH_2); *m/z* (EI) 321 (30 %, M^+ – HCl), 262 (62, M^+ – HCl – COOMe), 233 (100, $HOC_6H_3ICH_2^+$) and 88 (70, $H_2NCHCOOMe^+$).

(*N*-*t*-Butoxycarbonyl-3-iodo-L-tyrosine)-(3-iodo-L-tyrosine methyl ester) 77



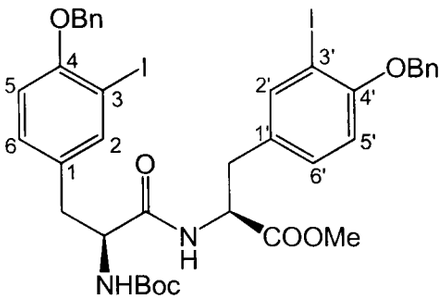
Anhydrous triethylamine (1.2 ml, 8.63 mmol) and DCC (1.9 ml, 8.52 mmol) were added to a stirred solution of the carboxylic acid **71** (3.423 g, 8.41 mmol) and the amine hydrochloride **76** (3.001 mg, 8.40 mmol) in anhydrous DMF (8 ml) and anhydrous methanol (30 ml). The reaction mixture was stirred for 2 hours at 0 °C, then kept at –18 °C (without stirring) for 18 hours. The mixture was filtered and the residue washed with ice-cold ethyl acetate (4 × 1 ml). The filtrate was concentrated (oil pump pressure) and the residue poured into water (20 ml) and ethyl acetate (30 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 15 ml). The combined organic extracts were washed with water (3 × 20 ml), aqueous hydrochloric acid (2 M; 10 ml), water (20 ml), aqueous sodium bicarbonate (10 %; 10 ml) and brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 50 % ethyl acetate in petrol) to give the *dipeptide* **77** (0.284 g, 40 %) as a colourless solid, m.p. 88-90 °C; R_f 0.52 (50 % ethyl acetate in petrol); $[\alpha]_D$ +27.2 (*c* 2.0 in DCM); ν_{max}/cm^{-1} (nujol mull) 3584 (OH), 3282 (NH), 1743 (ester C=O), 1655br (amide & carbamate C=O), 1415 (ArOH) and 1159 (ArOH); δ_H (300 MHz; chloroform-*d*) 7.49 (1 H, d, $^2J_{HH}$ 2.0, 2-H or 2'-H), 7.34 (1 H, d, $^2J_{HH}$ 1.5, 2-H or 2'-H), 7.01 (1 H, d, *J* 7.2, 5-H or 5'-H), 6.85 (1 H, dd, *J* 8.2 and $^2J_{HH}$ 1.5, 6-H or 6'-H), 6.78 (2 H, m, 5-H or 5'-H and 6-H or 6'-H), 6.58 (1 H, m, NH), 5.17 (1 H, m, NH), 4.75 (1 H, d, *m*, CH), 4.29 (1 H, m, CH), 3.71 (3 H, s, Me), 2.98-2.90 (4 H, m, 2 × CH_2) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.3 & 171.1 (*C*OOMe & CH*C*ONH), 155.6 (NH*C*OO*t*-Bu), 154.5 & 154.4 (C4 & C4'), 139.1 (C2 & C2'), 130.8 & 130.7 (C6 & C6'), 130.0 & 129.2 (C1 & C1'), 115.2 (C5 & C5'), 85.3 & 85.1 (C3 & C3'), 80.8 (*C*Me_3), 53.6 (2 × CH), 52.7 (Me), 36.9 & 36.5 (2 × CH_2) and 28.3 (C*Me*$_3$); *m/z* (ES) 1443 (10 %, M_2Na^+), 733 (65, MNa^+), 655 (537, MH^+ – CMe_2CH_2), 611 (100, MNa^+ – O*t*-Bu, – 2 OH, – Me), 485 (27), 262 (30) and 225 (32); HRMS (ES) found: 732.9872; $C_{24}H_{28}I_2N_2O_7$ requires *MNa*$^+$ 732.9884.

(*O*-Methyl-*N*-*t*-butoxycarbonyl-3-iodo-L-tyrosine)-(*O*-methyl-3'-iodo-L-tyrosine methyl ester) 78



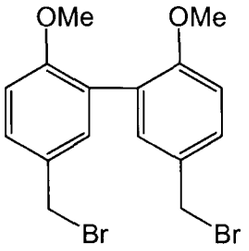
Potassium carbonate (981 mg, 7.11 mmol) and methyl iodide (270 µl, 4.34 mmol) were added to a stirred solution of the dipeptide **77** (1.000 g, 1.41 mmol) in acetone (15 ml). The reaction mixture was heated at reflux for 48 hours, then concentrated and the residue partitioned between ethyl acetate (20 ml) and water (20 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 15 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product, which was purified by flash chromatography (eluent: 40 % ethyl acetate in petrol) to give the *dimethylated dipeptide* **78** (582 mg, 56 %) as a colourless solid, m.p. 85-87 °C; R_f 0.37 (40 % ethyl acetate in petrol); $[\alpha]_D$ −1.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (solid phase) 3313 (NH), 2933 (CH), 1737 (ester C=O), 1688 (carbamate C=O), 1653 (amide C=O), 1599 (Ar), 1491 (Ar), 1250 (ArOR), 1166, 1047 and 667; δ_H (300 MHz; chloroform-*d*) 7.62 (1 H, d, $^4J_{HH}$ 1.9, 2-H or 2'-H), 7.40 (1 H, d, $^4J_{HH}$ 2.0, 2-H or 2'-H), 7.16 (1 H, dd, *J* 8.4 and $^4J_{HH}$ 1.9, 6-H or 6'-H), 6.93 (1 H, dd, *J* 8.4 and $^4J_{HH}$ 2.0, 6-H or 6'-H), 6.74 (1 H, d, *J* 8.4, 5-H or 5'-H), 6.69 (1 H, d, *J* 8.4, 5-H or 5'-H), 6.34 (1 H, d, *J* 6.4, N*H*Boc), 4.95 (1 H, br s, N*H*CHCOOMe), 4.74 (1 H, d app. t, *J* 6.4 and 6.0, C*H*NHBoc), 4.27 (1 H, app. d, *J* 6.0, C*H*COOMe), 3.85 (6 H, s, 2 × OMe), 3.71 (3 H, s, COOMe), 2.97 (4 H, m, 2 × CH_2) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.2 (ester C=O), 170.5 (amide C=O), 157.2 (C4 & C4'), 155.3 (carbamate C=O), 140.2 & 140.1 (C2 & C2'), 130.6 & 129.7 (C1 & C1'), 130.4 & 130.3 (C6 & C6'), 111.0 & 110.8 (C5 & C5'), 85.8 (C3 & C3'), 80.4 (*C*Me$_3$), 56.4 & 56.3 (2 × OMe), 55.8 (*C*HCOOMe), 53.3 (*C*HNHBoc), 52.5 (COO*Me*), 36.5 (2 × CH_2) and 28.3 (C*Me*$_3$); *m/z* (FAB) 1476 (5 %, M_2^+), 739 (22, MH^+), 683 (44, MH_2^+ − *t*-Bu), 638 (63, MH_2^+ − Boc), 318 (28, $MeOC_6H_3ICH_2CHCOOMe^+$ + H), 276 (71, $MeOC_6H_3ICH_2CHNH^+$) and 247 (34, $MeOC_6H_3ICH_2^+$); HRMS (ES) found: 739.0372; $C_{26}H_{32}I_2N_2O_7$ requires MH^+ 739.0372.

(*N*-*t*-Butoxycarbonyl-*O*-benzyl-3-iodo-L-tyrosine)-(*O*-benzyl-3-iodo-L-tyrosine methyl ester) 79



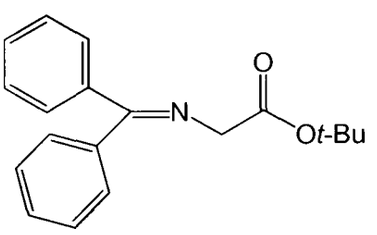
Potassium carbonate (487 mg, 3.53 mmol), TBAI (26 mg, 0.07 mmol) and benzyl bromide (420 µl, 3.53 mmol) were added to a stirred solution of the dipeptide **77** (1.016 g, 1.43 mmol) in anhydrous DMF (6 ml) and the reaction mixture stirred for 18 hours at room temperature. The reaction mixture was poured into water (10 ml) and extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with water (3 × 10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 20 to 50 % ethyl acetate in petrol) to give the *benzylated dipeptide* **79** (927 mg, 73 %) as a colourless solid, m.p. 143-145 °C; R_f 0.62 (50 % ethyl acetate in petrol); $[\alpha]_D$ +21.6 (*c* 2.0 in DCM); ν_{max}/cm^{-1} (nujol mull) 3326 (NH), 1747 (ester C=O), 1688 (carbamate C=O), 1651 (amide C=O), 1598 (Ar), 1517 (NH/CN), 1255 (ArOR); δ_H (300 MHz; chloroform-*d*) 7.64 (1 H, d, $^4J_{HH}$ 2.4, 2-H or 2'-H), 7.47 (4 H, d, *J* 7.7, from 2 × CH_2Ph), 7.43 (1 H, d, $^4J_{HH}$ 2.1, 2-H or 2'-H), 7.40-7.30 (6 H, m, from 2 × CH_2Ph), 7.12 (1 H, dd, *J* 8.7, $^4J_{HH}$ 2.4, 6-H or 6'-H), 6.88 (1 H, dd, *J* 8.5, $^4J_{HH}$ 2.1, 6-H or 6'-H), 6.77 (1 H, d, *J* 8.7, 5-H or 5'-H), 6.72 (1 H, d, *J* 8.5, 5-H or 5'-H), 6.35 (1 H, d, *J* 7.3, NH), 5.11 (2 H, s, CH_2Ph), 5.10 (2 H, s, CH_2Ph), 4.97 (1 H, d, *J* 7.0, NH), 4.74 (1 H, d app. t, *J* 7.3 and 5.7, CH), 4.28 (1 H, d app. t, *J* 7.0 and 5.7, CH), 3.69 (3 H, s, Me), 2.99 (2 H, d, $^2J_{HH}$ 15.4 and *J* 5.7, 2 × CH_AH_B), 2.94 (2 H, d, $^2J_{HH}$ 15.4 and *J* 5.7, 2 × CH_AH_B) and 1.41 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.6 & 171.0 (*C*OOMe & CH*C*ONH), 156.8 (NH*C*OO*t*-Bu), 140.6 (C2 & C2'), 136.8 (CH_2Ph), 131.3, 130.8, 130.6 & 130.5 (C1, C1', C4, C4', C6 & C6'), 129.0 (CH_2Ph), 128.3 (CH_2Ph), 127.4 (CH_2Ph), 113.1 & 112.9 (C5 & C5'), 87.4 & 87.1 (C3 & C3'), one peak missing (*C*Me$_3$), 71.3 (2 × CH_2Ph), 53.7 (2 × CH), 52.9 (Me), 37.0 (2 × CH_2CH) and 28.7 (CMe_3); *m/z* (FAB) 891 (5 %, MH^+), 791 (20, MH^+ – Boc), 352 (21) and 91 (100, $C_7H_7^+$); HRMS (ES) found: 891.0987; $C_{38}H_{40}I_2N_2O_7$ requires *MH* 891.0998.

5,5'-Bis-bromomethyl-2,2'-dimethoxy biphenyl 80



NBS (220 mg, 1.24 mmol) was added portion-wise to a stirred solution/suspension of the biphenyl **69** (151 mg, 0.62 mmol), dibenzoyl peroxide (DBP; 70 %; 3 mg, 0.012 mmol) and calcium carbonate (125 mg, 1.25 mmol) in carbon tetrachloride (5 ml) at reflux. A further portion of DBP (70 %; 3 mg, 0.012 mmol) was added, and the reaction mixture heated at reflux for 38 hours, with two further additions of DBP (70 %; 3 mg, 0.012 mmol). The reaction mixture was cooled to room temperature, washed with water (3 × 2 ml) and brine (3 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 10 % ethyl acetate in petrol) to give the *dibromide* **80** (35 mg, 14 %) as an colourless powder, m.p. 115-117 °C; R_f 0.31 (10 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solid phase) 3002-2834 (CH), 1604 (Ar), 1493 (Ar), 1446 (Ar), 1296 (CH_2Br), 1236 (ArOR), 1024, 814 and 509; δ_H (300 MHz; chloroform-*d*) 7.37 (2 H, dd, *J* 8.5 and $^2J_{HH}$ 2.4, 4-H & 4'-H), 7.28 (2 H, d, $^2J_{HH}$ 2.4, 6-H & 6'-H), 6.93 (2 H, d, *J* 8.5, 3-H & 3'-H), 4.53 (2 H, s, 2 × CH_2) and 3.77 (3 H, s, 2 × OMe); δ_C (75 MHz; chloroform-*d*) 157.5 C2), 132.7 (C4), 130.2 (C6), 130.0 (C5), 127.9 (C1), 111.6 (C3), 56.2 (OMe) and 34.4 (CH_2); *m/z* (EI)* 400 (10 %, M^+), 321 (100, M^+ – Br), 225 (65, M^+ – 2 Br, – Me), 194 (20, M^+ – 2 Br, – Me, – OMe) and 120 (15); HRMS (EI)* found: 399.9495; $C_{16}H_{16}{}^{79}Br^{81}BrO_2$ requires M^+ 399.9497.

***t*-Butyl *N*-(diphenylmethylene)glycinate 81**

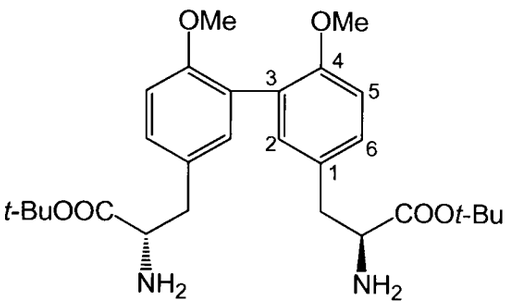


Benzophenone imine **89** (200 µl, 1.19 mmol) was added to a stirred solution/suspension of glycine *t*-butyl ester hydrochloride **85** (196 mg, 1.16 mmol) in DCM (4 ml) and the reaction mixture was stirred at room temperature for 22 hours. The reaction mixture was filtered to remove ammonium chloride, and the filtrate concentrated. The residue was dissolved in ether (5 ml), washed with water (5 ml) and brine (5 ml), dried and concentrated to give a colourless solid which was recrystallised from ether/petrol to give the glycinate **81** (222 mg, 65 %) as colourless plates, m.p. 111-113 °C (lit.[197] 111-112 °C); dec. on silica; ν_{max}/cm^{-1} (nujol mull) 1735 (C=O) 1623 (C=N), 1577 (Ar), 1491 (Ar), 1219, 1150 and

* Recorded on a Waters GCT TOF spectrometer.

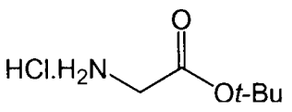
694; δ_H (300 MHz; chloroform-*d*) 7.68-7.17 (10 H, m, 2 × Ph), 4.12 (2 H, s, CH_2) and 1.46 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 171.9 (C=N), 170.3 (C=O), 139.8 (Ar; C), 136.6 (Ar; C), 130.8 (Ar; CH), 129.2 (2 × Ar; CH), 129.0 (Ar; CH), 128.4 (Ar; CH), 128.1 (Ar; CH), 81.5 (CMe_3), 56.7 (CH_2) and 28.5 (CMe_3); *m/z* (ES) 296 (100 %, MH^+), 240 (30, $MH^+ - CMe_2CH_2$) and 182 (48, $MH_2^+ - CH_2COOt\text{-Bu}$).

Di-(*O*-methyl-L-tyrosine *t*-butyl ester) 83



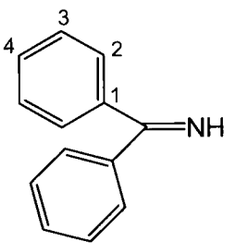
The glycinate **81** (52 mg, 0.18 mmol), *N*-(9-anthracenylmethyl)-cinchonidinium chloride **82** (2 mg, 0.005 mmol) and aqueous potassium hydroxide (50 %; 2 ml) were added to a stirred solution of the dibromide **80** (35 mg, 0.088 mmol) in anhydrous toluene (2 ml) and the reaction mixture was stirred for 42 hours at room temperature. The mixture was concentrated and ethyl acetate (5 ml) added to the residue. The aqueous layer was removed and the ethyl acetate evaporated. THF (2 ml) and aqueous citric acid (15 %; 2 ml) were added and the mixture stirred for 3 hours at room temperature. The THF was evaporated and the aqueous layer basified with aqueous sodium carbonate (2 M) and extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated. The residue was dissolved in ethyl acetate (5 ml) and extracted with citric acid (2 M; 5 ml). The aqueous fraction was basified with aqueous sodium hydroxide (2 M) and re-extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give a yellow oil (6 mg) from which the desired dityrosine **83** could be identified: ν_{max}/cm^{-1} (solution in chloroform-*d*) 3422br (NH), 2980 (CH), 1727 (C=O), 1656 (NH/CN), 1247 (ArOR), 1153, 910 and 731; δ_H (300 MHz; chloroform-*d*) 7.16 (2 H, dd, *J* 8.5 and $^4J_{HH}$ 2.3, 2 × 6-H), 7.08 (2 H, d, $^4J_{HH}$ 2.3, 2 × 2-H), 6.90 (2 H, d, *J* 8.5, 2 × 5-H), 3.74 (6 H, s, 2 × OMe), 3.59 (2 H, dd, *J* 7.8 and 5.1, 2 × CH), 3.02 (2 H, dd, $^2J_{HH}$ 13.6 and *J* 5.1, 2 × CH_AH_B), 2.79 (2 H, dd, $^2J_{HH}$ 13.6 and *J* 7.8, 2 × CH_AH_B) and 1.44 (18 H, s, 2 × *t*-Bu); *m/z* (ES) 629 (60 %), 550 (65), 529 (50), 389 (100, MH_3^+ – 2 *t*-Bu), 359 (80) and 275 (55); HRMS (ES) found: 389.1698; $C_{28}H_{40}N_2O_6$ requires $MH_3^+ - 2$ t-*Bu* 389.1713.

Glycine *t*-butyl ester hydrochloride 85



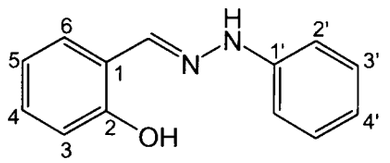
t-Butyl bromoacetate **84** (7.6 ml, 51.5 mmol) was added to a mixture of anhydrous THF (20 ml) and anhydrous liquid ammonia (50 ml) at –40 °C, and the reaction mixture stirred for 2 hours at –40 °C. The reaction mixture was allowed to warm to room temperature overnight and filtered to remove ammonium bromide. The filtrate was concentrated to give the amino ester as a pale yellow oil (5.194 g, 77 %). Hydrogen chloride gas was bubbled through a solution of the ester (4.760 g, 36.33 mmol) in anhydrous THF (20 ml) until precipitation appeared complete. The reaction mixture was filtered to give the hydrochloride salt **85** (4.171 g, 53 % over two steps) as colourless prisms, m.p. 140-141 °C (lit.[198] 137-139 °C); R_f 0.66 (50 % methanol in DCM); ν_{max}/cm^{-1} (nujol mull) 3186 (NH_3^+), 1750 (C=O), 1528, 1241 (C–O) and 1152 (C–O); δ_H (300 MHz; chloroform-*d*) 8.51 (3 H, br s, NH_3^+), 3.88 (2 H, s, CH_2) and 1.48 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 166.9 (C=O), 84.2 (CMe_3), 41.4 (CH_2) and 28.5 (CMe_3); *m/z* (ES) 173 (100 %), 132 (80, $M^+ - Cl^-$) and 117 (20).

Benzophenone imine 89



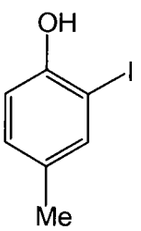
A solution of anhydrous benzonitrile **88** (10.2 ml, 0.10 mol) in anhydrous ether (20 ml) was added dropwise to a mechanically stirred solution of phenylmagnesium bromide **87** (2.94 M in ether; 36 ml, 0.106 mol) at such a rate as to maintain a gentle reflux. The reaction mixture was stirred for 5 hours at reflux, then allowed to cool to room temperature. Anhydrous methanol (24 ml) was added, and the reaction mixture stirred for a further 30 minutes at room temperature. The mixture was filtered and the filtrate concentrated to give a crude product which was purified by reduced pressure distillation (121 °C/4 mm Hg; lit.[199] 135-138 °C/4 mm Hg) to give the imine **89** (12.571 g, 74 %) as a colourless oil; dec. on silica; ν_{max}/cm^{-1} (film) 3256 (NH), 3058 (CH), 1659 (C=N), 1599 (Ar), 1568 (Ar), 1489 (Ar), 1477 (Ar), 1363, 1277, 1196, 892 and 696; δ_H (300 MHz; chloroform-*d*) 9.70 (1 H, br s, NH), 7.82-7.38 (10 H, m, Ar); δ_C (75 MHz; chloroform-*d*) 178.8 (C=N), 130.7 & 130.5 (C1 & C4) and 128.8 & 128.7 (C2 & C3); *m/z* (ES) 182 (100 %, MH^+).

Salicylaldehyde phenylhydrazone 91



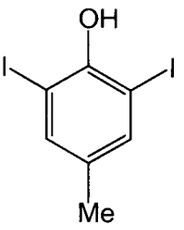
A solution of salicylaldehyde **90** (2.9 ml, 27.2 mmol) in ethanol (15 ml) was added to a stirred solution of phenyl hydrazine (2.7 ml, 27.5 mmol) in ethanol (10 ml) and the reaction mixture stirred for 30 minutes at room temperature. The reaction mixture was cooled to −15 °C, then filtered under reduced pressure. The precipitate was washed with ice-cold ethanol, methanol and ether, and dried to give the hydrazone **91** (2.430 g, 42 %) as colourless needles, m.p. 140-142 °C (from ethanol; lit.[200] 141-142 °C); R_f 0.58 (30 % ethyl acetate in petrol); ν_{max}/cm^{-1} (nujol mull) 3315 (NH), 1619 (C=N), 1602 (NH), 1587 (Ar), 1524 (Ar), 1357 (PhOH) and 1206 (PhOH); δ_H (300 MHz; chloroform-*d*) 10.86 (1 H, s, OH or NH), 7.86 (1 H, s, CHN), 7.48 (1 H, s, OH or NH), 7.46-7.17 (3 H, m, 3 × Ar), 7.14 (1 H, dd, *J* 7.7 and $^4J_{HH}$ 1.6, Ar), 7.02-6.87 (5 H, m, 5 × Ar); δ_C (75 MHz; chloroform-*d*) 157.4 (C2), 143.8 (C1'), 141.6 (C=N), 130.5 (C4 or C6), 130.0 (C3'), 129.8 (C4 or C6), 121.3 (C5), 119.9 (C4'), 118.9 (C1), 117.0 (C3) and 113.0 (C2'); *m/z* (ES) 213 (62 %, MH^+), 130 (13) and 105 (100).

2-Iodo-4-methyl phenol 105 and 2,6-diiodo-4-methyl phenol 102



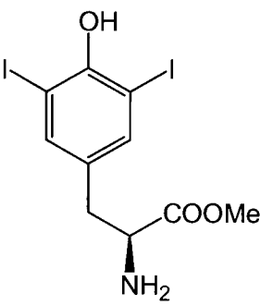
A solution of iodine (4.720 g, 18.58 mmol) in ethanol (75 ml) was added dropwise over 30 minutes to a stirred solution of *p*-cresol **20** (1.999 g, 18.51 mmol) in aqueous ammonia (35 %; 100 ml) at 0 °C. The reaction mixture was stirred for a further 30 minutes at 0 °C. Aqueous hydrochloric acid (conc.; 120 ml) was then added dropwise *via* an addition funnel, with the temperature maintained at 0 °C. The resulting mixture was filtered, the filtrate concentrated and the residue extracted with ethyl acetate (3 × 50 ml). The combined organic extracts were washed with water (20 ml), aqueous sodium thiosulfate (2 M; 20 ml) and brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 5 to 10 % ethyl acetate in petrol) to give the aryl iodide **105** (2.761 g, 64 %) as a pale yellow oil; R_f 0.19 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3480 (OH), 2920 (C-H), 1601 (Ar), 1576 (Ar), 1486 (Ar), 1454 (Ar), 1399 (ArOH), 1281, 1179 (ArOH) and 1034; δ_H (300 MHz; chloroform-*d*) 7.47 (1 H, d, $^4J_{HH}$ 1.7, 3-H), 7.03 (1 H, dd, *J* 8.2 and $^4J_{HH}$ 1.7, 5-H), 6.88 (1 H, d, *J* 8.2, 6-H), 5.17 (1 H, s, OH) and 2.25 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 153.0 (C1), 138.7

(C3), 132.4 (C4), 131.3 (C5), 115.1 (C6), 85.8 (C2) and 20.4 (Me); *m/z* (EI) 234 (100 %, M^+), 127 (18, I^+), 107 (48, $M^+ - I$) and 77 (35).



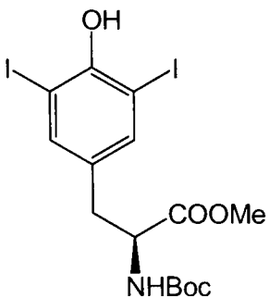
Also isolated was the diiodide **102** (719 mg, 11 %) as colourless needles, m.p. 59-61 °C (lit.[201] 55-58 °C); R_f 0.38 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solid phase) 3449 (OH), 2919 (CH), 1735, 1586 (Ar), 1544 (Ar), 1458 (Ar), 1386 (ArOH), 1308, 1266, 1229, 1149 (ArOH), 853, 703 and 555; δ_H (300 MHz; chloroform-*d*) 7.50 (2 H, s, 2 × 3-H), 5.57 (1 H, s, OH) and 2.21 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 151.8 (C1), 140.0 (C3), 134.3 (C4), 82.4 (C2) and 19.9 (Me); *m/z* (EI) 360 (100 %, M^+), 233 (57, $M^+ - I$), 127 (36, I^+), 106 (47, $M^+ - 2I$) and 78 (54).

3,5-Diiodo-L-tyrosine methyl ester 107



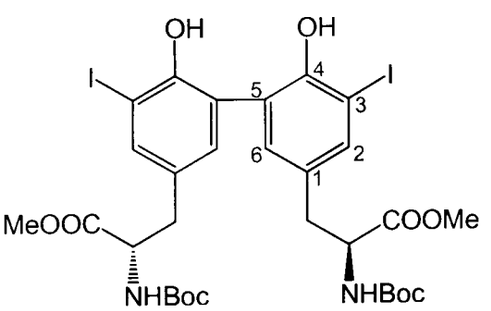
Thionyl chloride (3.2 ml, 43.9 mmol) was added dropwise to a stirred solution/suspension of 3,5-diiodo-L-tyrosine dihydrate **106** (2.000 g, 4.26 mmol) in anhydrous methanol (20 ml) at –25 °C. The resulting clear solution was allowed to warm to room temperature, then heated at reflux for 15 hours. The solvent was removed under reduced pressure and the residue dissolved in water (125 ml), then neutralised with aqueous sodium hydroxide (2 M). The resulting suspension was centrifuged in portions (3000 r.p.m.; 5 minutes), the supernatant decanted off and the residue dried under vacuum to give the ester **107** (1.868 g, 98 %) as a cream-coloured solid, m.p. 177 °C (dec.); R_f 0.86 (49 % methanol and 2 % triethylamine in DCM); $[\alpha]_D$ +14.6 (*c* 2.0 in DMSO); Found: C, 26.7; H, 2.45; I, 56.6; N, 2.9; $C_{10}H_{11}I_2NO_3$ requires C, 26.9; H, 2.50; I, 56.8; N, 3.1 %; ν_{max}/cm^{-1} (nujol mull) 3437 (OH), 3243 (NH_2), 1733 (C=O) and 1580 (NH); δ_H (300 MHz; DMSO-d_6) 7.53 (2 H, s, 2-H & 6-H), 3.83 (2 H, br s, NH_2), 3.58 (3 H, s, Me), 3.50 (1 H, dd, *J* 7.5 and 5.9, CH), 2.73 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 5.9, CH_AH_B) and 2.62 (1 H, dd, $^2J_{HH}$ 13.5 and *J* 7.5, CH_AH_B); δ_C (75 MHz; DMSO-d_6) 175.4 (C=O), 154.7 (C4), 140.0 (C2 & C6), 133.7 (C1), 87.5 (C3 & C5), 55.8 (CH), 51.8 (Me) and 33.7 (CH_2); *m/z* (ES) 470 (100 %, MNa^+) and 448 (37, MH^+).

_N_-_t_-Butoxycarbonyl-3,5-diiodo-L-tyrosine methyl ester 108



Triethylamine (2.2 ml, 15.8 mmol), was added to a stirred solution/suspension of the ester **107** (4.778 g, 10.69 mmol) in anhydrous DCM (50 ml). The resulting clear solution was cooled to 0 °C and di-_t_-butyl dicarbonate (2.682 g, 12.30 mmol) was added in one portion. The reaction mixture was stirred at 0 °C for 30 minutes, then at room temperature for 17 hours. The reaction mixture was poured into water (50 ml), the layers separated and the aqueous fraction extracted with DCM (3 × 20 ml). The combined organic extracts were washed with brine (2 × 40 ml), dried (Na_2SO_4) and concentrated to give a crude product which was purified by flash chromatography (eluent: 20 % ethyl acetate in petrol) to give the protected diiodotyrosine **108** (4.346 g, 74 %) as a cream-coloured solid, m.p. 127-129 °C; R_f 0.43 (20 % ethyl acetate in petrol); $[\alpha]_D$ +5.6 (_c_ 2.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3455 (OH), 3360 (NH), 1728 (ester C=O), 1685 (carbamate C=O), 1523 (NH/CN), 1369 (ArOH) and 1165 (ArOH); δ_H (300 MHz; chloroform-_d_) 7.44 (2 H, s, 2-H & 6-H), 5.72 (1 H, br s, OH), 5.03 (1 H, br d, _J_ 7.8, NH), 4.49 (1 H, d app. t, _J_ 7.8 and 5.7, CH), 3.75 (3 H, s, COOMe), 3.03 (1 H, dd, $^2J_{HH}$ 13.8 and _J_ 5.7, CH_AH_B), 2.90 (1 H, dd, $^2J_{HH}$ 13.8 and _J_ 5.7, CH_AH_B) and 1.45 (9 H, s, _t_-Bu); δ_C (75 MHz; chloroform-_d_) 172.3 (_C_OOMe), 155.3 (NHCO), 153.1 (C4), 140.4 (C2 & C6), 132.6 (C1), 82.6 (C3 & C5), 80.6 (_C_Me$_3$), 54.8 (CH), 52.9 (COO_Me_), 36.7 (CH_2) and 82.7 (C_Me_$_3$); _m/z_ (EI) 491 (5 %, MH^+ – _t_-Bu), 474 (3, M^+ – O_t_-Bu), 430 (62, MH^+ – Boc, – OH), 359 (60, $HOC_6H_2I_2CH_2^+$), 323 (10, $HOC_6H_2ICH_2^+$), 88 (59) and 57 (100, _t_-Bu).

Di-(_N_-_t_-butoxycarbonyl-3-iodo-L-tyrosine methyl ester) 109

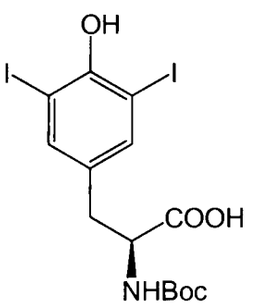


Aqueous phosphate buffer (pH 6; 5 × 33 ml) was added to 5 separate stirred solutions of the protected diiodotyrosine **108** (5 × 1.001 g, 1.83 mmol) in DCM (5 × 33 ml). The reaction mixtures were stirred for 26 hours at room temperature, during which time the pH was maintained at 6 by addition of aqueous sodium hydroxide (0.05 M) as necessary and the colour was maintained at pale pink by addition of aqueous sodium thiosulfate (0.05 M). The reaction mixtures were combined and the layers separated. The aqueous fraction

was then extracted with DCM (3 × 50 ml). The combined organic extracts were washed with aqueous sodium thiosulfate (2 M; 50 ml) and brine (50 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (20 % ethyl acetate in petrol) to give the *diiododityrosine* **109** (598 mg, 16 %) as a cream-coloured solid, m.p. 83-85 °C; R_f 0.12 (20 % ethyl acetate in petrol); $[\alpha]_D$ +0.32 (*c* 5.0 in methanol); Found: C, 43.2; H, 4.65; I, 30.1; N, 3.5; $C_{30}H_{38}I_2N_2O_{10}$ requires C, 42.9; H, 4.60; I, 30.2; N, 3.3 %; ν_{max}/cm^{-1} (nujol mull) 3491-3165 (OH & NH), 1744 (ester C=O), 1694 (carbamate C=O), 1509 (NH/CN), 1367 (ArOH) and 1163 (ArOH); δ_H (300 MHz; chloroform-*d*) 7.50 (2 H, d, $^4J_{HH}$ 1.8, 2 × 6-H), 7.15 (2 H, d, $^4J_{HH}$ 1.8, 2 × 2-H), 5.11 (2 H, d, *J* 6.8, 2 × NH), 4.61 (2 H, d app. t, *J* 8.1 and 6.8, 2 × CH), 3.77 (6 H, s, 2 × Me), 3.20 (2 H, dd, $^2J_{HH}$ 13.5 and *J* 8.1, 2 × CH_AH_B), 2.61 (2 H, dd, $^2J_{HH}$ 13.5 and *J* 8.1, 2 × CH_AH_B) and 1.29 (18 H, s, 2 × *t*-Bu); δ_C (75 MHz; chloroform-*d*) 172.3 (*C*OOMe), 155.4 (NHCO), 152.8 (C4), 141.3 (C1), 132.6 (C5), 130.1 (C2), 122.8 (C6), 85.8 (C3), 80.8 (*C*Me$_3$), 54.8 (CH), 53.0 (COO*Me*), 39.0 (CH_2) and 28.6 (C*Me*$_3$); *m/z* (ES) 863 (17 %, MNa^+), 862 (100, $MNa^+ - H$) and 805 (28, $MNa^+ - H$, – *t*-Bu).

Optimisation reactions were routinely analysed by quantitative reverse-phase analytical hplc. Purified samples of the diiodotyrosine **108** and the diiododityrosine **109** were used to calibrate the integration of the UV chromatogram at 218 nm; crude reaction products were then analysed using the calibrated system (gradient elution: 20 to 80 % acetonitrile in water over 25 minutes; eluting 1 ml/minute from an Alphasil 5 ODS 250 × 4.6 mm column).

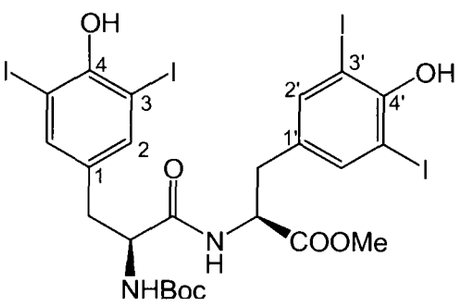
N-Butoxycarbonyl-3,5-diiodo-L-tyrosine 110



Triethylamine (2.2 ml, 15.8 mmol) was added to a stirred solution/suspension of 3,5-diiodo-L-tyrosine dihydrate **106** (4.999 g, 10.66 mmol) in THF (25 ml) and water (25 ml). The resulting clear solution was cooled to 0 °C and di-*t*-butyl dicarbonate (2.70 ml, 11.77 mmol) added. The reaction mixture was stirred at 0 °C for 30 minutes, then at room temperature for 14 hours. The THF was removed under reduced pressure and the residue poured into water (50 ml), acidified with aqueous hydrochloric acid (2 M) and extracted with ethyl acetate (3 × 40 ml). The combined organic extracts were washed with brine (50 ml), dried ($MgSO_4$)

and concentrated to give the carbamate **110** (5.421 g, 95 %) as a pale pink solid, m.p. 184 °C (dec.; lit.[202] 187-189 °C); R_f 0.56 (50 % ethyl acetate in petrol); $[\alpha]_D$ +16.4 (*c* 2.0 in methanol); Found: C, 31.8; H, 3.25; I, 47.3; N, 2.4; $C_{14}H_{17}I_2NO_5$ requires C, 31.5; H, 3.20; I, 47.6; N, 2.6 %; ν_{max}/cm^{-1} (nujol mull) 3500-2500 (COOH), 3489 (OH), 3345 (NH), 1712 (acid C=O), 1692 (carbamate C=O), 1256 (NH/CN), 1366 (ArOH) and 1157 (ArOH); δ_H (300 MHz; acetone-d_6) 7.57 (2 H, s, 2-H & 6-H), 6.07 (1 H, s, *J* 8.8, NH), 4.23 (1 H, d app. t, *J* 8.8 and 6.2, CH), 3.13 (2 H, br s, COOH & OH), 3.00 (1 H, dd, $^2J_{HH}$ 14.0 and *J* 6.2, CH_AH_B), 2.76 (1 H, dd, $^2J_{HH}$ 14.0 and *J* 6.2, CH_AH_B) and 1.24 (9 H, s, *t*-Bu); δ_C (75 MHz; acetone-d_6) 173.6 (COOH), 156.6 (NHCO), 155.1 (C4), 141.7 (C2 & C6), 135.1 (C1), 84.6 (C3 & C5), 79.8 (CMe_3), 55.8 (CH), 36.4 (CH_2) and 29.0 (CMe_3); *m/z* (EI) 533 (<1 %, M^+), 518 (<1, M^+ – Me), 505 (<1), 477 (2, M^+ – *t*-Bu), 459 (3), 433 (2, MH^+ – Boc), 416 (5, MH^+ – Boc, – OH), 387 (5), 358 (100, $HOC_6H_2I_2CH_2^+$) and 233 (80, $HOC_6H_2ICH_2^+$).

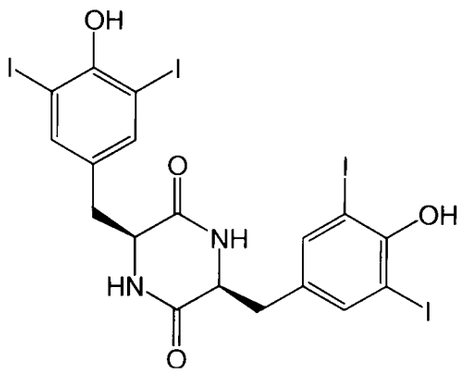
(*N*-*t*-Butoxycarbonyl-3,5-diiodo-L-tyrosine)-(3,5-diiodo-L-tyrosine methyl ester) 111



HOBt (55 mg, 0.40 mmol) and a catalytic quantity of DMAP were added to a stirred solution/suspension of the carboxylic acid **110** (210 mg, 0.39 mmol) and DCC (85 mg, 0.41 mmol) in anhydrous THF (10 ml). A solution/suspension of the amine **107** (170 mg, 0.36 mmol) in anhydrous THF (10 ml) was added and the reaction mixture stirred at room temperature for 24 hours. The mixture was then kept at −18 °C for 18 hours and filtered under reduced pressure. The filtrate was evaporated and the residue partitioned between ethyl acetate (10 ml) and water (10 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (2 × 10 ml). The combined organic extracts were washed with water (10 ml), aqueous hydrochloric acid (10 ml), water (10 ml), aqueous sodium bicarbonate (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 30 % ethyl acetate in petrol) to give the *dipeptide* **111** (170 mg, 50 %) as a pale yellow solid, m.p. 192 °C (dec.); R_f 0.23 (30 % ethyl acetate in petrol); $[\alpha]_D$ −16.4 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3453 (NH or OH), 3316 (NH or OH), 1743

(ester C=O), 1689 (carbamate C=O), 1652 (amide C-O), 1520 (Ar, NH CN), 1365 (ArOH) and 1163 (ArOH); δ_H (300 MHz; chloroform-*d*) 7.54 (2 H, s, Ar), 7.35 (2 H, s, Ar), 6.64 (1 H, d, *J* 7.4, NH), 5.93 (2 H, br s, 2 × OH), 5.04 (1 H, d, *J* 7.5, NH), 4.75 (1 H, d app. t, *J* 7.4 and 5.6, CH), 4.32 (1 H, d app. t, *J* 7.5 and 6.2, CH), 3.74 (3 H, s, Me), 2.97-2.92 (4 H, m, 2 × CH_2) and 1.43 (9 H, s, *t*Bu); δ_C (75 MHz; chloroform-*d*) 171.3 & 170.8 (*C*OOMe & CH*C*ONHCH), one peak missing (NH*C*OO*t*Bu), 153.2 & 153.2 (C4 & C4'), 140.4 & 140.3 (C2 & C2'), 132.9 & 132.1 (C1 & C1'), 83.0 (C3 & C3'), 82.7 (*C*Me$_3$), 53.6 & 53.5 (2 × CH), 53.1 (Me), 36.5 (2 × CH_2) and 28.7 (C*Me$_3$*); *m/z* (ES) 985 (55 %, MNa^+), 963 (15, MH^+), 907 (45, MH^+ – *t*Bu), 863 (75, MH_2^+ – Boc), 225 (100) & 100 (50); HRMS (ES) found: 984.7807; $C_{24}H_{26}I_4N_2O_7$ requires *MNa*$^+$ 984.7811.

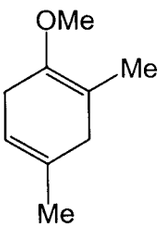
Cyclo-(3,5-diiodo-L-tyrosine)-(3,5-diiodo-L-tyrosine) 112



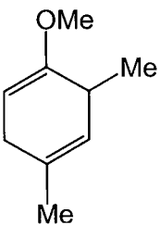
A solution of the dipeptide **111** (596 mg, 0.62 mmol) in formic acid (50 ml) was stirred at room temperature for 2 hours. The mixture was concentrated and the residue dissolved in *s*-butanol (50 ml) and toluene (20 ml). The solution was heated at reflux for 18 hours, then cooled to room temperature and concentrated. The residue was dispersed in toluene (20 ml) and the product was isolated by filtration and washed with ether (5 ml) to give the *diketopiperazine* **112** (355 mg, 69 %) as an off-white solid, m.p. 198-200 °C (dec.); R_f 0.10 (ethyl acetate); $[\alpha]_D$ –248 (*c* 1.0 in DMSO); ν_{max}/cm^{-1} (nujol mull) 1667 (C=O), 1539 (Ar), 1313 (ArOH), 1236 (ArOH) and 1151; δ_H (300 MHz; DMSO-d_6) 9.43 (2 H, br s, 2 × OH), 8.14 (2 H, s, 2 × NH), 7.44 (4 H, s, Ar), 3.91 (2 H, app. t, *J* 5.1, 2 × CH), 2.45 (2 H, dd, $^2J_{HH}$ 13.6, *J* 5.1, 2 × CH_AH$_B$) and 2.20 (2 H, dd, $^2J_{HH}$ 13.6, *J* 5.1, 2 × CH$_A$*H$_B$*); δ_C (75 MHz; DMSO-d_6) 166.5 (C=O), 154.5 (C4), 140.5 (C2 & C6), 133.1 (C1), 87.2 (C3 & C5), 55.8 (CH), and 38.0 (CH_2); *m/z* (FAB) 984 (21 %), 831 (100, MH^+), 705 (14, MH_2^+ – I), 472 (27, MH^+ – $CH_2C_6H_2I_2OH$), 359 (28, $HOC_6H_2I_2CH_2^+$) and 233 ($HOC_6H_3ICH_2^+$); HRSM (ES) found: 847.7478; $C_{18}H_{14}I_4N_2O_4$ requires *MNH$_4$*$^+$ 847.7471.

1-Methoxy-2,4-dimethylcyclohexa-1,4-diene 114 & 4-methoxy-1,3-dimethylcyclohexa-1,4-diene 121

Lithium (sufficient to maintain the blue colour) was added to a stirred solution of 2,4-dimethyl anisole (1.02 ml, 7.35 mmol) in anhydrous liquid ammonia (30 ml), *t*-butanol (4 ml) and ether (2.5 ml) and the reaction mixture stirred at reflux for 2 hours. Aqueous ammonium chloride (sufficient to discharge the blue colour) was added and the ammonia allowed to evaporate. Ice was added to the residue, and the mixture extracted with ether (3 × 15 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried (Na_2SO_4) and concentrated to give a mixture of the cyclohexadienes **114** and **121** as a colourless oil (779 mg, 77 %).



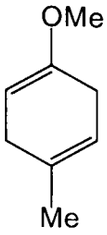
Cyclohexadiene **114**: δ_H (300 MHz; chloroform-*d*) 5.37 (1 H, app. octet, *J* 1.5, 5-H), 3.52 (3 H, s, OMe), 2.82-2.73 (2 H, m, 6-CH_2), 2.60 (1 H, d, $^2J_{HH}$ 8.2, 3-CH_AH$_B$), 2.58 (1 H, d, $^2J_{HH}$ 8.2, 3-CH$_A$H$_B$), 1.69-1.66 (3 H, m, 4-Me) and 1.63 (3 H, t, *J* 1.0, 2-Me); δ_C (175 MHz; chloroform-*d*) 145.5 (C1), 131.6 (C4), 118.0 (C5), 111.1 (C2), 56.2 (OMe), 37.6 (C3), 26.3 (C6), 22.6 (4-Me), and 14.8 (2-Me).



Cyclohexadiene **121**: δ_H (300 MHz; chloroform-*d*) 5.29 (1 H, d, *J* 1.6, 2-H), 4.58 (1 H, app. t, *J* 3.6, 5-H), 3.54 (3 H, s, OMe), 3.48 (1 H, s, 3-H), 2.71-2.65 (2 H, m, 6-CH_2), 1.61 (3 H, s, 1-Me) and 1.12 (3 H, d, *J* 6.9, 3-Me); δ_C (175 MHz; chloroform-*d*) 157.4 (C4), 130.4 (C1), 124.9 (C2), 89.8 (C5), 54.0 (OMe), 33.7 (C3), 31.3 (C6), 22.6 (1-Me), and 20.4 (3-Me).

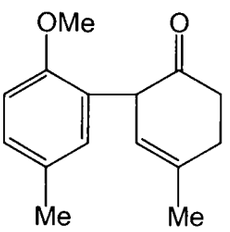
Combined data: R_f 0.83 & 0.54 (30 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2967-2826 (CH), 1712 (C=C), 1697 (C=C), 1679 (C=C), 1663 (C=C), 1447, 1353, 1231 (ROR), 1203, 1175, 1144 and 1010;

1-Methoxy-4-methyl cyclohexa-1,4-diene 120



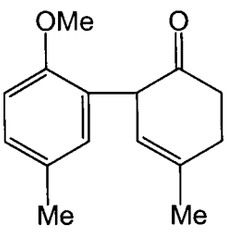
Lithium (sufficient to maintain the blue colour) was added to a stirred solution of 4-methyl anisole **66** (10.3 ml, 81.81 mmol) in anhydrous liquid ammonia (60 ml) and ether (20 ml) and the reaction mixture stirred at reflux for 30 minutes. Methanol (20 ml) was added dropwise with caution (with more lithium added as necessary) and the reaction mixture stirred for a further 30 minutes at reflux. Methanol (sufficient to displace the blue colour) was added, the reaction quenched with water (30 ml) and the ammonia allowed to evaporate. The residue was filtered under reduced pressure and the filtrate extracted with ether (3 × 20 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried (Na_2SO_4) and concentrated to give the cyclohexadiene **120** (7.500 g, 73 %) as a colourless oil; R_f 0.74 (10 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2828 (C–H), 1699 (C=C), 1448, 1387 and 1217 (C–O); δ_H (300 MHz; chloroform-*d*) 5.36 (1 H, s, 5-H), 4.61 (1 H, s, 2-H), 3.54 (3 H, s, OMe), 2.70 (4 H, s, 2 × CH_2) and 1.69 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 153.5 (C1), 132.1 (C4), 118.1 (C6), 90.8 (C3), 54.3 (OMe), 31.5 (C2), 29.6 (C5) and 23.1 (Me); *m/z* (EI) 124 (91 %, M^+), 109 (100, M^+ – Me), 191 (37, $C_7H_7^+$) and 77 (42).

85. 2-(2'-Methoxy-5'-methyl-phenyl)-4-methyl-cyclohex-3-enone 125

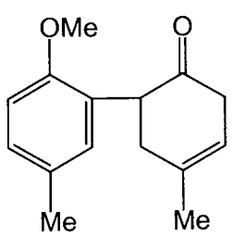


A solution of the diol **144** (101 mg, 0.29 mmol) in anhydrous THF (10 ml) was added to a stirred solution of lithium (sufficient to maintain the blue colour) in anhydrous liquid ammonia (15 ml) and the reaction mixture stirred at reflux for 18 hours. The reaction was quenched with solid ammonium chloride (sufficient to displace the blue colour) and the ammonia allowed to evaporate. The residue was dispersed in water (sufficient to dissolve excess ammonium chloride) and extracted with ether (3 × 10 ml). The combined organic extracts were dried (Na_2SO_4) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 3 - 20 % ethyl acetate in petrol) to give the crude *aryl cyclohexenone* **125** (27.0 mg, 27 %) as a colourless oil; spectroscopically identical to that obtained previously.

4-Methyl-2-(2'-methoxy-5'-methyl phenyl)-cyclohex-3-enone 125 and 4-methyl-2-(2'-methoxy-5'-methyl phenyl)-cyclohex-4-enone 126

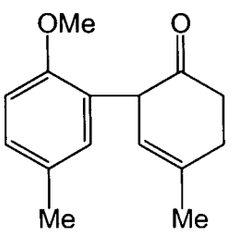


Lithium (sufficient to maintain the blue colour) was added to a stirred solution of the methyl anisole dimer **69** (98 mg, 0.405 mmol) in anhydrous liquid ammonia (15 ml) and THF (10 ml) and the reaction mixture stirred at reflux for 2 hours. The reaction was quenched with solid ammonium chloride (sufficient to discharge the blue colour) and the ammonia allowed to evaporate. The mixture was dispersed in water (sufficient to dissolve excess ammonium chloride) and extracted with ether (3 × 15 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried (Na_2SO_4) and concentrated to give the *cyclohexadienes* **122** and **124** as a colourless oil which was purified by flash chromatography (eluent: 5 % toluene in petrol) to give the *aryl cyclohexenone* **125** (27.0 mg, 27 %) as a colourless oil; R_f 0.53 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3417, 2925-2838 (CH), 1715 (C=O), 1688 (C=C), 1609 (Ar), 1587 (Ar), 1504 (Ar), 1463 (Ar), 1242 (ArOR), 1033 and 807; δ_H(300 MHz; chloroform-*d*) 7.01 (1 H, dd, *J* 8.4 and $^4J_{HH}$ 2.2, 4'-H), 6.92 (1 H, d, $^4J_{HH}$ 2.2, 6'-H), 6.78 (1 H, d, *J* 8.4, 3'-H), 5.40 (1 H, dq, *J* 3.0 and $^4J_{HH}$ 1.3, 3-H), 4.34 (1 H, dq, *J* 3.0 and $^4J_{HH}$ 2.0, 2-H), 3.76 (3 H, s, ArO*Me*), 2.70-2.46 (4 H, m, 5-CH_2 and 6-CH_2), 2.27 (3 H, s, Ar*Me*) and 1.87 (3 H, s, 4-Me); δ_C (75 MHz; chloroform-*d*) 210.6 (C=O), 155.2 (C2'), 135.1 (C4), 130.7 (C6'), 130.3 (C5), 129.1 (C4'), 128.6 (C1'), 123.7 (C3), 111.6 (C3'), 56.2 (OMe), 49.9 (C2), 38.3 (C6), 31.2 (C5), 23.6 (4-Me) and 21.0 (Ar*Me*); *m/z* (ES) 248 (40 %) and 231 (100, MH^+); HRMS (ES) found: 231.1388; $C_{15}H_{18}O_2$ requires MH^+ 231.1385.

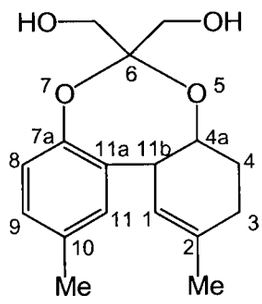


Also isolated was the *aryl cyclohexenone* **126** (3 mg, 3 %) as a yellow oil, R_f 0.53 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solution in DCM) 1643br (C=O, C=C & Ar), 1499 (Ar), 1422 and 896; δ_H (300 MHz; chloroform-*d*) 6.98-6.95 (2 H, m, 4'-H & 6'-H), 6.76 (1 H, d, *J* 8.6, 3'-H), 5.36 (1 H, app. sextet, $^4J_{HH}$ 1.5, 5-H), 3.80 (3 H, s, OMe), 3.79-3.73 (1 H, m, 2-H), 2.28 (3 H, s, Ar*Me*), 2.02-1.88 (3 H, m, 3-CH_AH$_B$ & 6-CH_2), 1.75 (3 H, m, 4-Me) and 1.46-1.35 (1 H, m, 3-C$H_A H_B$); δ_C (75 MHz; chloroform-*d*) one peak missing (C=O), 155.2 (C2'), 135.6 & 135.3 (C4 & C1'), 129.8 (C5'), 129.5 (C4'), 127.4 (C6'), 124.9 (C5), 110.6 (C3'), 560. (OMe), 34.8 (C2), 30.5 & 30.4 (C3 & C6), 24.5 (4-Me) and 21.9 (Ar*Me*); *m/z* (ES) 297 (75 %), 231 (100, MH^+) and 215 (70, M^+ – Me); HRMS (ES) found: 231.1394; $C_{15}H_{18}O_2$ requires MH^+ 231.1385.

2-(2'-Methoxy-5'-methyl-phenyl)-4-methyl-cyclohex-3-enone 125 & 3,4,4a,11b-Tetrahydro-6,6-Dihydroxymethyl-2,10-dimethyl-6*H*-dibenzo[*d*,*f*][1,3]dioxepin 146

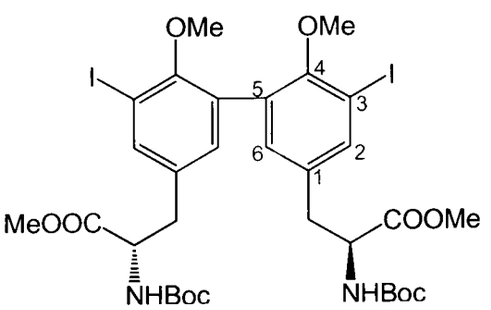


A solution of the diol **138** (101 mg, 0.35 mmol) in anhydrous THF (10 ml) was added to a stirred solution of lithium (sufficient to maintain a blue colour) in anhydrous liquid ammonia (~15 ml) and the reaction mixture was stirred at reflux for 40 minutes. The reaction was quenched with ammonium chloride (sufficient to displace the blue colour) and the ammonia allowed to evaporate. The residue was dissolved in water and the mixture extracted with ether (3 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($Na_2S_2O_4$) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 10 to 40 % ethyl acetate in petrol) to give the aryl enone **125** (25 mg, 33 %) as a colourless oil spectroscopically identical to that obtained previously.



Also isolated was the *arylcyclohexene* **146** (8 mg, 8 %) as a yellow oil; R_f 0.42 (50 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solution in DCM) 3401 (OH), 1643 (C=C), 1452 and 1068 (OH); δ_H (300 MHz; chloroform-*d*) 7.01 (1 H, dd, *J* 8.5 and $^4J_{HH}$ 2.0, 9-H), 6.91 (1 H, d, $^4J_{HH}$ 2.0, 11-H), 6.80 (1 H, d, *J* 8.5, 8-H), 5.36 (1 H, br d, *J* 5.4, 1-H), 4.30 (1 H, ddd, *J* 11.3, 6.3 and 3.8, 4a-H), 4.16-4.09 (2 H, m, 2 × CH_AH_BOH), 3.97 (1 H, br d, *J* 6.3, 11b-H), 3.78-3.62 (2 H, m, 2 × CH_AH_BOH), 2.32 (3 H, s, Ar*Me*), 2.15-2.01 (2 H, m, 4-CH_2), 1.84 (2 H, br s, 3-CH_2) and 1.60 (3 H, s, 2-Me); *m/z* (ES) 313 (100 %, MNa^+).

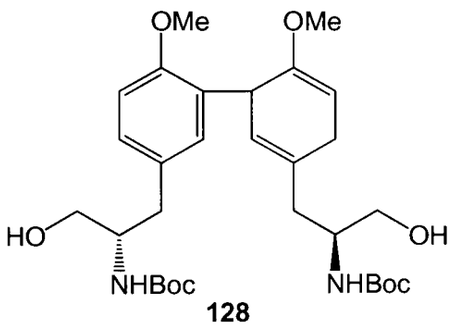
Di-(*O*-methyl-*N*-*t*-butoxycarbonyl-3-iodo-L-tyrosine) 127



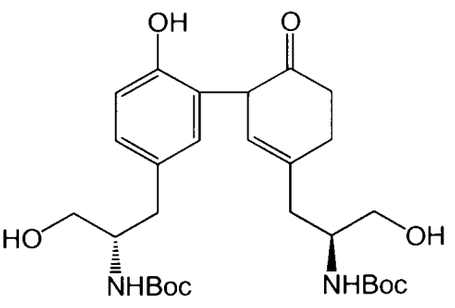
Methyl iodide (80 µl, 1.29 mmol) was added to a stirred solution/suspension of the diiododityrosine **109** (427 mg, 0.508 mmol) and potassium carbonate (213 mg, 1.54 mmol) in acetone (7 ml) at room temperature. The reaction mixture was heated at reflux for 16 hours, then concentrated. The residue was partitioned between ethyl acetate (5 ml) and water (10 ml) and the layers separated. The aqueous fraction was extracted with ethyl acetate (3 × 5 ml) and the combined organic extracts washed with water (5 ml) and

brine (5 ml), dried ($MgSO_4$) and concentrated to give the *protected diiododityrosine* **127** (382 mg, 87 %) as a pale yellow solid, m.p. 68-70 °C; R_f 0.32 (30 % ethyl acetate in petrol); $[\alpha]_D$ +50.0 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3368 (NH), 1744 (ester C=O), 1714 (carbamate C=O), 1500 (Ar), 1245 (ArOR) and 1166; δ_H (300 MHz; chloroform-*d*) 7.55 (2 H, d, $^4J_{HH}$ 1.8, 2 × 2-H), 7.15 (2 H, d, $^4J_{HH}$, 1.8, 2 × 6-H), 5.04 (2 H, d, *J* 7.7, 2 × NH), 4.55 (2 H, d app. t, *J* 7.7 and 5.8, 2 × CH), 3.76 (6 H, s, 2 × COO*Me*), 3.45 (6 H, s, 2 × ArO*Me*), 3.11 (2 H, dd, $^2J_{HH}$ 14.0 and *J* 5.8, 2 × CH_AH_B), 2.97, 2 H, dd, $^2J_{HH}$ 14.0 and *J* 5.8, 2 × CH_AH_B) and 1.44 (18 H, s, 2 × *t*-Bu); δ_C (75 MHz; chloroform-*d*) 172.3 (*C*OOMe), 156.3 (C4), 155.3 (NHCO), 140.4 (C2), 134.1 (C1), 133.6 (C6), 131.4 (C5), 93.0 (C3), 80.6 (*C*Me$_3$), 60.7 (ArO*Me*), 54.8 (CH), 52.9 (COO*Me*), 37.3 (CH_2) and 28.7 (C*Me*$_3$); *m/z* (ES) 891 (30 %, MNa^+), 713 (95, MH_3^+ – *t*-Bu, – Boc), 669 (100, MH_4^+ – 2 Boc) and 609 (45); HRMS (ES) found: 891.0839; $C_{32}H_{42}I_2N_2O_{10}$ requires MNa^+ 891.0827.

Birch Reduction of Di-(*O*-methyl-*N*-*t*-butoxycarbonyl-3-iodo-L-tyrosine) 127



128

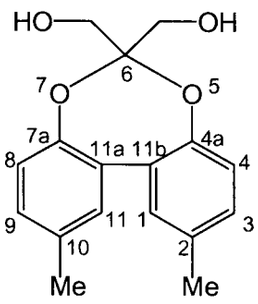


130

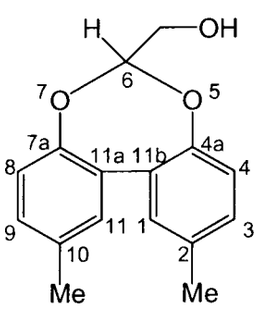
A solution of the diiododityrosine **127** (195 mg, 0.23 mmol) in anhydrous THF (5 ml) was added dropwise to a solution of lithium (sufficient to maintain a blue colour) in anhydrous liquid ammonia (~15 ml). The reaction mixture was stirred at reflux for 1 hour, then the reaction was quenched with ammonium chloride (sufficient to displace the blue colour) and the ammonia allowed to evaporate. The residue was dissolved in water (20 ml) and the mixture extracted with ether (3 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give a crude product which was analysed by LCMS (gradient elution: 20 to 95 % acetonitrile over 5 minutes, then 95 % acetonitrile in water for 3 minutes; eluting 1 ml/minute from using a Waters XTerra Prep MS C_{18} 5 μm 19 × 50 mm column); *m/z* (ES) 5.5 minutes (**128**): 563 (75 %, MH^+) and 463 (100, MH_2^+ – Boc); 6.0 minutes (**130**): 545 (80 %, MH^+), 435 (100,

MH_2^+ – Boc) and 379 (20, MH_3^+ – Boc, – *t*-Bu); 6.2 minutes (**130**): 535 (60 %, MH^+) and 435 (100, MH_2^+ – Boc); 8.0 minutes (**128**): 639 (100 %, MNa^+).

6,6-Dihydroxymethyl-2,10-dimethyl-6*H*-dibenzo[*d*,*f*][1,3]dioxepin 138 and 6-hydroxymethyl-2,10-dimethyl-6*H*-dibenzo[*d*,*f*][1,3]dioxepin 143



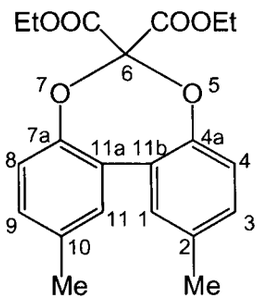
Anhydrous methanol (3 ml) was added dropwise over 1 hour to a refluxing solution of the diester **142** (702 mg, 1.90 mmol) and sodium borohydride (161 mg, 4.23 mmol) in anhydrous THF (7 ml) and the mixture refluxed for a further 3 hours. The reaction mixture was allowed to cool and poured in to water (20 ml). The layers were separated and the aqueous fraction extracted with DCM (6 × 10 ml). The combined organic extracts were washed with brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (40 % ethyl acetate in petrol) to give the *diol* **138** (371 mg, 68 %) as colourless plates, m.p. 106-108 °C; R_f 0.43 (50 % ethyl acetate in petrol); ν_{max}/cm^{-1} (nujol mull) 3440 (OH) and 1247 (ArOR); δ_H (300 MHz; chloroform-*d*) 7.28 (2 H, d, $^4J_{HH}$ 1.8, 1-H & 11-H), 7.11 (2 H, dd, *J* 8.1 $^4J_{HH}$ 1.8, 3-H & 9-H), 7.03 (2 H, d, *J* 8.1, 4-H & 8-H), 3.94 (4 H, d, *J* 6.5, 2 × CH_2), 2.64 (2 H, t, *J* 6.5, 2 × OH) and 2.39 (6 H, s, 2 × Me); δ_C (75 MHz; chloroform-*d*) 148.8 (C4a & C7a), 135.6 (C2 & C10), 132.5 (C11a & C11b), 129.9 (C3 & C9), 129.4 (C1 & C11), 123.1 (C4 & C8), 114.6 (C6), 62.7 (CH_2) and 21.4 (Me); *m/z* (ES) 309 (100 %, MNa^+); HRMS (ES) found: 309.1079; $C_{17}H_{18}O_4$ requires MNa^+ 309.1103.



Also isolated was the *alcohol* **143** (9.1 mg, 2 %) as a colourless oil; R_f 0.64 (50 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3601 (OH), 2926 (CH), 1254 (ArOR) and 1045 (OH); δ_H (300 MHz; chloroform-*d*) 7.34 (2 H, d, $^4J_{HH}$ 1.9, 1-H & 11-H), 7.08 (2 H, dd, *J* 8.2 and $^4J_{HH}$ 1.9, 3-H & 9-H), 7.00 (2 H, d, *J* 8.2, 4-H & 8-H), 5.61 (1 H, t, *J* 5.7, 6-H), 3.86 (2 H, app. t, *J* 5.7, CH_2), 2.37 (6 H, s, Me) and 2.32 (1 H, t, *J* 5.7, OH); δ_C (75 MHz; chloroform-*d*) 151.3 (C4a & C7a), 134.9 (C2 & C10), 130.8 (C11a & C11b), 130.0 (C3 & C9), 129.4 (C1 & C11), 121.7 (C4 & C8), 109.1 (C6), 63.5 (CH_2) and 21.4 (Me); *m/z* (EI) 256 (100 %, M^+), 225 (56, M^+ – CH_2OH), 209 (51, M^+ – 2Me, – OH), 195 (65, MH^+ –

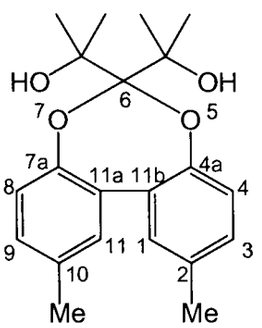
$OCHCH_2OH$), 182 (31, $M^+ - 2 \times Me, - OCH[O]CH_2OH$) and 152 (32); HRMS (EI) found: 256.1096; $C_{16}H_{16}O_3$ requires M^+ 256.1099.

6,6-Diethoxycarbonyl-2,10-dimethyl-6*H*-dibenzo[*d*,*f*][1,3]dioxepin 142

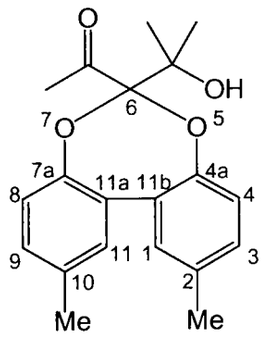


Diethyl bromomalonate (5.0 ml, 29.33 mmol) was added dropwise over 8 hours to a stirred solution/suspension of the biphenol **28** (2.44 g, 11.40 mmol) and potassium carbonate (1.94 g, 14.06 mmol) in anhydrous DMF (10 ml) and the reaction mixture stirred for a further 18 hours at room temperature. The reaction mixture was poured into water (30 ml) and extracted with ethyl acetate (3 × 30 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried (Na_2SO_4) and concentrated to give a crude product from which the solid material was collected by filtration, washed with petrol and purified by recrystallisation (from DCM/petrol) to give the *dioxepin* **142** (2.99 g, 71 %) as colourless needles, m.p. 93-94 °C (from DCM/petrol); R_f 0.47 (20 % ethyl acetate in petrol); Found C, 68.1; H. 6.15; $C_{21}H_{22}O_6$ requires C, 68.1; H, 6.00 %; ν_{max}/cm^{-1} (nujol mull) 1770 (C=O), 1495 (Ar), 1248 (ArOR), 1122, 1096 and 1045; δ_H (300 MHz; chloroform-*d*) 7.28 (2 H, d, $^4J_{HH}$ 1.7, 1-H & 11-H), 7.18 (2 H, d, *J* 8.2, 4-H & 8-H), 7.12 (2 H, dd, *J* 8.2 and $^4J_{HH}$ 1.7, 3-H & 9-H), 4.36 (4 H, q, *J* 7.1, 2 × CH_2CH_3), 2.39 (6 H, s, 2 × Ar*Me*) and 1.34 (6 H, t, *J* 7.1, 2 × CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 164.7 (*C*OOEt), 148.8 (C4a & C7a), 136.4 (C2 & C10), 132.1 (C11a & C11b), 129.9 (C3 & C9), 129.2 (C1 & C11), 122.9 (C4 & C8), 107.8 (C6), 63.2 (CH_2CH_3), 21.5 (Ar*Me*) and 14.5 (CH_2CH_3); *m/z* (FAB) 393 (9 %, MNa^+), 370 (42, M^+), 297 (80, M^+ – COOEt) and 69 (83).

6,6-Di(1-hydroxy-1-methylethyl)-2,10-dimethyl-6*H*-dibenzo [*d,f*] [1,3] dioxepin 144 and 6-acetyl-6-(1-hydroxy-1-methylethyl)-2,10-dimthyl-6*H*-dibenzo [*d,f*] [1,3] dioxepin 145



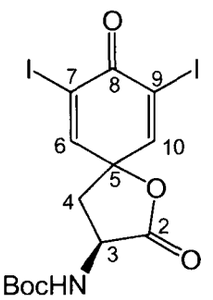
Methylmagnesium bromide (3 M solution in ether; 5.4 ml, 16.2 mmol) was added dropwise to a stirred solution of the diester **142** (992 mg, 2.68 mmol) in anhydrous THF (12 ml) at 0 °C and the reaction mixture stirred for 30 minutes at 0 °C. A further portion of methylmagnesium bromide (2.7 ml, 8.1 mmol) was added and the mixture stirred for a further hour at 0 °C. The reaction mixture was quenched with aqueous ammonium chloride (10 ml) and poured into water (30 ml). The layers were separated and the aqueous fraction extracted with ether (3 × 20 ml). The combined organic extracts were washed with brine (20 ml), dried (Na_2SO_4) and concentrated to give a crude product which was purified by flash chromatography (eluent: 20 % ethyl acetate in petrol) to give the *diol* **144** (348 mg, 40 %) as a colourless solid, m.p. 163-165 °C; R_f 0.33 (20 % ethyl acetate in petrol); Found: C, 73.4; H, 7.55; $C_{21}H_{26}O_4$ requires C, 73.7; H, 7.65 %; ν_{max}/cm^{-1} (nujol mull) 3440-3328 (OH), 1401 (OH), 1226 (ArOR) and 1120 (OH); δ_H (300 MHz; chloroform-*d*) 7.24 (2 H, s, 1-H & 11-H), 7.08 (4 H, s, 3-H, 4-H, 8-H & 9-H), 2.65 (2 H, s, 2 × OH), 2.37 (6 H, s, 2 × Ar*Me*) and 1.46 (12 H, s, 2 × C*Me*$_2$OH); δ_C (75 MHz, chloroform-*d*) 152.5 (C4a & C7a), 134.1 (C2 & C10), 129.9 (C3 & C9), 129.8 (C1 & C11), 129.6 (C11a & C11b), 122.9 (C4 & C8), 121.6 (C6), 79.0 (*C*Me$_2$OH), 28.8 (C*Me*$_2$OH) and 21.3 (Ar*Me*); *m/z* (EI) 266 (100 %, M – CMe$_2$OH, – OH); *m/z* (ES) 366 (12 %, $MNaH^+$) and 365 (100, MNa^+).



Also isolated was the *ketol* **145** (207 mg, 24 %) as a colourless solid, m.p. 109-112 °C; R_f 0.35 (20 % ethyl acetate in petrol); Found: C, 73.4; H, 6.65; $C_{20}H_{22}O_4$ requires C, 73.6; H, 6.80 %; ν_{max}/cm^{-1} (nujol mull) 3477 (OH), 1717 (C=O), 355 (OH), 1257 (ArOR) and 1121 (OH); δ_H (300 MHz; chloroform-*d*) 7.26 (2 H, s, 1-H and 11-H), 7.09 (4 H, s 3-H, 4-H, 8-H and 9-H), 2.81 (1 H, s, OH), 2.37 (6 H, s, Ar*Me*), 2.16 (3 H, s, COMe) and 1.40 (6 H, s, C*Me*$_2$OH); δ_C (75 MHz; chloroform-*d*) 207.3 (C=O), 150.8 (C-4a & C-7a), 135.0 (C-2 & C-10), 130.7 (C-11a & C-11b), 130.0 (C-3 and C-9), 129.2 (C-1 & C-11), 123.8 (C-4 and C-8), 117.3

(C-6), 76.6 (*C*Me$_2$OH), 30.5 (CO*Me*), 25.7 (C*Me*$_2$OH) and 21.4 (Ar*Me*); *m/z* (ES) 350 (10 %, MNaH$^+$) and 349 (100, MNa$^+$).

(*S*)-*N*-*t*-Butoxycarbonyl-3-amino-7,9-diiodo-1-oxa-spiro[4.5]deca-6,9-diene-2,8-dione 154



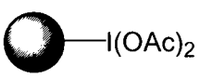
A solution of NBS (2.01 g, 11.29 mmol) and acetonitrile (27 ml) in acetate buffer (0.2 M, pH 4; 108 ml) was added to a stirred solution/suspension of the carbamate **110** (2.01 g, 3.77 mmol) and acetonitrile (96 ml) in acetate buffer (384 ml) and the reaction mixture stirred at room temperature for 90 minutes. The resulting suspension was filtered under reduced pressure and concentrated to give a crude product (1.52 g, 76 %) which was purified by hplc (isocratic elution: 45 % acetonitrile in water over 40 minutes; eluting 20 ml/minute from a Supelco Discovery C_{18} 5 μm 21.2 × 250 mm reverse phase column; peaks observed at 210 nm) to give the *spirolactone* **154** as yellow needles, m.p. 163-165 °C (from ethyl acetate/petrol); R_f 0.38 (30 % ethyl acetate in petrol); $[\alpha]_D$ −142 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (nujol mull) 3341 (NH), 1803 (lactone C=O), 1694 (carbamate C=O), 1676 (dienone C=O), 1592 (C=C), 1526 (NH/CN), 1267 (C–O) and 1168 (C–O); δ_H (500 MHz; DMSO-d_6) 8.31 (1 H, d, $^4J_{HH}$ 2.6, 6-H or 10-H), 7.83 (1 H, d, $^4J_{HH}$ 2.6, 6-H or 10-H), 7.58 (1 H, d, *J* 8.3, NH), 4.75 (1 H, app. td, *J* 10.1 and 8.3, 3-H), 2.68 (1 H, dd, $^2J_{HH}$ 13.2 and *J* 10.1, C*H*$_A$H$_B$), 2.40 (1 H, dd, $^2J_{HH}$ 13.2 and *J* 10.1, CH$_A$*H*$_B$) and 1.43 (9 H, s, *t*-Bu); δ_C (125 MHz; DMSO-d_6) 174.4 & 174.3 (C2 & C8), 156.7 & 156.3 (C6 & C10), 155.8 (NHCO), 101.2 & 99.9 (C7 & C9), 80.8 (C5), 80.0 (*C*Me$_3$), 49.8 (CHNH), 35.9 (CH$_2$) and 29.0 (C*Me*$_3$); *m/z* (ES) 554 (100 %, MNa$^+$), 498 (10, MNaH$^+$ – *t*-Bu), 432 (53, MH$_2^+$ – Boc), 415 (40, MH$^+$ – BocNH) and 388 (37); HRMS (ES) found: 553.8957; $C_{14}H_{15}I_2NO_5$ requires *MNa*$^+$ 553.8937.

Poly(iodostyrene) 170

Carbon tetrachloride (4 ml), aqueous sulfuric acid (50 %; 3 ml) and nitrobenzene (20 ml) were added to a mixture of polystyrene **169** (1 % divinyl benzene, 100-200 mesh; 1.600 g) and iodine pentoxide (609 mg, 1.82 mmol). The reaction mixture was stirred very slowly at 90 °C for 96 hours, then allowed to cool and

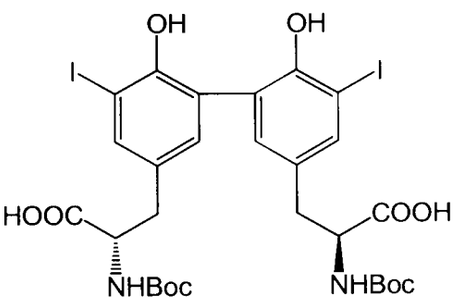
filtered. The residue was washed with chloroform (3 × 10 ml), THF 3 × 10 ml) and methanol (3 × 10 ml), then dried under high-vacuum to give the polymer **170** (1.950 g, loading 2.32 mmol/g) as light brown granules (Found C, 60.9; H, 5.05; I, 29.5 %).

Polymer-supported diacetoxyiodosobenzene (PSDIB) 171



Aqueous hydrogen peroxide (30 %; 5 ml, 52.06 mmol) was added dropwise to acetic anhydride (18 ml, 190.94 mmol) at 0 °C. The reaction mixture was allowed to warm slowly to room temperature and was then stirred for 24 hours at room temperature. Poly(iodostyrene) **170** (997 mg) was added to the fresh peracetic acid and the reaction mixture stirred very slowly at 45 °C for 20 hours. The mixture was allowed to cool, then filtered. The residue was washed with chloroform (3 × 2 ml), THF (3 × 2 ml) and methanol (3 × 2 ml) and dried under high-vacuum to give the polymer **171** (375 mg, loading 1.85 mmol/g) as light brown granules (Found: C, 55.2; H, 4.65; I, 28.3 %).

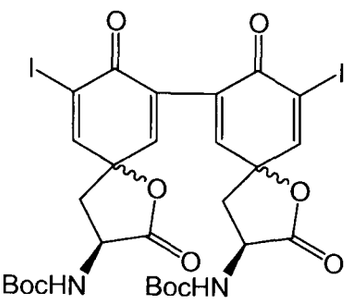
Di-(*N*-*t*-butoxycarbonyl-3-iodo-L-tyrosine) 172



Aqueous sodium hydroxide (1 M; 10 ml) was added to a stirred solution of the protected diiododityrosine **109** (206 mg, 0.25 mmol) in methanol (10 ml) and the reaction mixture stirred for 2.5 hours at room temperature. The mixture was concentrated and the residue extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give the *diacid* **172** (0.188 g, 94 %) as pale yellow plates m.p. 115-116 °C; R_f 0.08 (30 % ethyl acetate and 2 % AcOH in petrol); $[\alpha]_D$ +23.2 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3674-3103 (COOH), 3391 (NH), 1732 (acid C=O) and 1700 (carbamate C=O); δ_H (300 MHz; methanol-d_4) 7.65 (2 H, d, $^4J_{HH}$ 1.9, 2 × 2-H), 7.08 (2 H, d, $^4J_{HH}$ 1.9, 2 × 6-H), 4.36 (2 H, dd, *J* 8.6 and 4.9, 2 × CH), 3.14 (2 H, dd, $^2J_{HH}$ 13.9, *J* 4.9, 2 × CH_AH_B), 2.88 (2 H, dd, $^2J_{HH}$ 13.9, *J* 8.6, 2 × CH_AH_B) and 1.42 (18 H, s, 2 × *t*-Bu); δ_C (75 MHz; methanol-d_4) 175.4 (COOH), 158.2 (NHCO), 154.1 (C4), 141.6 (C2), 134.3 (C6), 132.7 (C1), 127.2 (C5), 87.8 (C3), 81.1 (CMe_3), 56.6 (CH), 37.9 (CH_2) and 29.2 (CMe_3); *m/z* (ES)

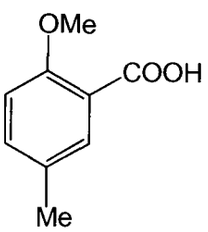
835 (73 %), 813 (84, MH^+), 757 (41, MH^+ – Boc), 701 (36) and 657 (30); HRMS (ES) found: 921.4326; $C_{56}H_{61}N_2O_{10}$ requires MH^+ 921.435.

(3*S*, 3'*S*, 5*R/S*, 5'*R/S*)-*N,N'*-Di(*t*-butoxycarbonyl)-3,3'-diamino-9,9'-diiodo-[7,7']-bi-[1-oxa-spiro[4.5]decyl]-6,6',9,9'-tetraene-2,2',8,8'-tetraone 173



A solution of NBS (62 mg, 0.347 mmol) in acetonitrile (0.9 ml) and acetate buffer (0.2 M, pH 4; 3.9 ml) was added to a stirred solution of the diacid **172** (50 mg, 0.062 mmol) in acetonitrile (1.6 ml) and acetate buffer (6.4 ml). The reaction mixture was stirred at room temperature for 90 minutes, then the acetonitrile was evaporated and the residue extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with brine (5 ml), dried ($MgSO_4$) and concentrated to give the crude *dispirolactone* **173** as a mixture of diastereoisomers (36 mg, 72 %) as a yellow solid; R_f 0.30 and 0.60 (30 % ethyl acetate in petrol); δ_H (300 MHz; chloroform-*d*) 7.64 (1 H, br s, olefinic), 7.36 (1 H, br s, olefinic), 7.06 – 6.94 (2 H, m, 2 × olefinic), 5.52 and 5.40 (2 H, 2 × br s, 2 × NH), 4.52 (2 H, m, 2 × CH), 2.76 – 2.70 (2 H, m, 2 × CH_AH_B), 2.55 (2 H, app q, *J* 11.7, 2 × CH_AH_B), 1.39 (18 H, s, 2 × *t*-Bu).

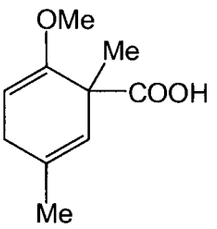
2-Methoxy-5-methyl benzoic acid 180



n-Butyllithium (1.14 M solution in hexanes; 1.67 ml, 1.90 mmol) was added dropwise to a stirred solution of the aryl bromide **67** (240 µl, 1.81 mmol) in anhydrous THF (7 ml) at –78 °C. The mixture was stirred at –78 °C for 10 mins; freshly crushed solid carbon dioxide was then added and the reaction mixture allowed to warm slowly to room temperature over 17 hours. The mixture was concentrated and the residue partitioned between ether (5 ml) and aqueous sodium hydroxide (2 M; 5 ml). The organic fraction was removed and discarded. The aqueous fraction was then acidified using aqueous hydrochloric (conc.) and extracted with ethyl acetate (3 × 5ml). The combined organic extracts were washed with brine (5 ml), dried ($MgSO_4$) and concentrated to give the benzoic acid **180** (262 mg, 87 %) as a pale yellow solid, m.p. 64-66 °C (lit.[203] 68-69 °C); R_f 0.57 (50 % ethyl acetate & 2 % AcOH in petrol); ν_{max} cm^{-1} (nujol mull) 3420-

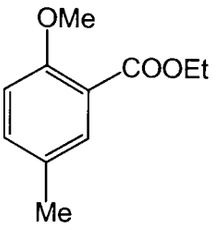
1806 (COOH), 1712 (C=O), 1617 (Ar), 1584 (Ar), 1503 (Ar), 1249 (ArOR), 1021 and 755; δ_H (300 MHz; methanol-d_4) 7.66 (1 H, d, $^4J_{HH}$ 1.5, 6-H), 7.35 (1 H, dd, J 8.5 and $^4J_{HH}$ 1.5, 4-H), 7.03 (1 H, d, J 8.5, 3-H), 3.90 (3 H, s, OMe) and 2.31 (3 H, s, Me); δ_C (75 MHz; methanol-d_4) 169.9 (C=O), 158.8 (C2), 136.1 (C4), 133.6 (C6), 131.6 (C5), 120.6 (C1), 113.8 (C3), 57.0 (OMe), 20.7 (Me); *m/z* (EI) 166 (98 %, M^+), 149 (77, M^+ – OH), 119 (100, M^+ – 2Me, – OH), 93 (84), 91 (96, M^+ – 2Me, – COOH), 72 (95) and 51 (74).

(±)-2-Methoxy-1,5-dimethyl cyclohexa-1,4-diene carboxylic acid 182



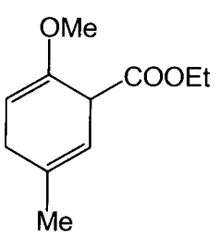
Anhydrous ammonia (~15 ml) was distilled onto a solution of the benzoic acid **180** (110 mg, 0.66 mmol) in anhydrous THF (5 ml). Lithium granules (19 mg, 2.71 mmol) were added and the mixture stirred at –78 °C for 30 minutes. The blue colour was displaced by addition of piperylene (0.1 ml) and methyl iodide (75 µl, 1.20 mmol) was added. The reaction mixture was then stirred at –78 °C for a further 30 minutes and the ammonia removed in a stream of nitrogen. The residue was dissolved in water (10 ml) and extracted with ether (5 ml). The layers were separated and the organic fraction discarded. The aqueous fraction was acidified using pH 5 phosphate buffer and aqueous phosphoric acid (conc.) and extracted with ethyl acetate (3 ×5 ml). The combined organic extracts were washed with brine (5 ml), dried ($MgSO_4$) and concentrated to give the *cyclohexadiene* **182** (184 mg, 44 %) as a yellow oil; R_f 0.56 (50 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3306-2628 (OH), 2935 (CH), 1707 (C=O), 1666 (C=C), 1614 (C=C), 1530, 1450, 1254 (ROR), 1168 and 1035; δ_H (300 MHz; methanol-d_4) 5.12 (1 H, app. sextet, $^4J_{HH}$ 1.3, 6-H), 4.87 (1 H, app. t, J 4.0, 3-H), 3.44 (3 H, s, OMe), 2.67-2.64 (2 H, m, CH_2), 1.64 (3 H, d, $^4J_{HH}$ 1.3, 5-Me), and 1.24 (3 H, s, 1-Me); δ_C (75 MHz; methanol-d_4) one peak missing (C=O), 156.7 (C2), 136.0 (C5), 152.3 (C6), 93.0 (C3), 55.1 (OMe), C1 obscured by solvent peak, 32.4 (C4), 24.3 (5-Me) and 22.9 (1-Me); *m/z* (EI) 182 (14 %, M^+), 137 (100, M^+ – COOH), 122 (44, M^+ – COOH, – Me), 105 (15, M^+ – COOH, – 2 Me) and 91 (35, $C_7H_7^+$).

2-Methoxy-5-methyl benzoic acid ethyl ester 183



n-Butyllithium (1.14 M solution in hexanes; 10.5 ml, 12.0 mmol) was added to a stirred solution of the aryl bromide **67** (1.45 ml, 10.91 mmol) in anhydrous THF (25 ml) at –78 °C. The mixture was stirred at –78 °C for 30 minutes, then cannulated into a stirred solution of ethyl chloroformate (4.15 ml, 43.41 mmol) in anhydrous THF (50 ml) also at –78 °C over 10 minutes. The reaction mixture was then stirred at –78 °C for 1 hour and at room temperature for 22 hours. The mixture was concentrated and the residue partitioned between ethyl acetate (30 ml) and aqueous sodium bicarbonate (10 %; 30 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 15 ml). The combined organic extracts were washed with brine (20 ml), dried ($MgSO_4$) and concentrated to give the *benzoic ester* **183** (1.999 g, 94 %) as a yellow oil, R_f 0.55 (20 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2958 (C-H), 1728 (C=O), 1614 (Ar), 1582 (Ar), 1503 (Ar), 1464 (Ar), 1303, 1259 (ArOR), 1201, 1080, 1026 and 812 (Ar); δ_H (300 MHz; chloroform-*d*) 7.59 (1 H, d, $^2J_{HH}$ 2.4, 6-H), 2.23 (1 H, dd, *J* 8.2 and $^2J_{HH}$ 2.4, 4-H), 6.88 (1 H, d, *J* 8.2, 3-H), 4.36 (2 H, q, *J* 7.2, CH_2CH_3), 3.87 (3 H, s, OMe), 2.31 (3 H, s, Ar*Me*), and 1.38 (3 H, t, *J* 7.2, CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 166.4 (C=O), 157.1 (C2), 133.8 (C4), 131.8 (C6), 129.4 (C5), 120.1 (C1), 112.1 (C3), 60.8 (CH_2), 56.2 (OMe), 20.3 (Ar*Me*), and 14.3 (CH_2CH_3); *m/z* (EI) 194 (22 %, M^+), 149 (100, M^+ – OEt), 119 (15, M^+ – OEt, – 2Me) and 91 (40, $C_7H_7^+$); HRMS (EI) found: 194.0943; $C_{11}H_{14}O_3$ requires M^+ 194.0943.

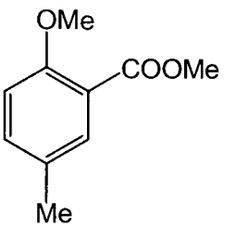
(±)-2-Methoxy-5-methyl cyclohexa-2,5-diene carboxylic acid ethyl ester 184



Anhydrous ammonia (~30 ml) was distilled onto a solution of the benzoic ester **183** (1.492 g, 7.69 mmol) and anhydrous *t*-butanol (1.1 ml, 11.5 mmol) in anhydrous THF (10 ml). Potassium (sufficient to maintain a blue colour) was added and the reaction mixture stirred at –78 °C for 2 hours. Ammonium chloride (sufficient to displace the blue colour) was added and the ammonia removed in a stream of nitrogen. The residue was partitioned between ether (20 ml) and water (30 ml). The layers were separated and the aqueous layer extracted with ether (2 × 20 ml). The combined organic extracts were washed with brine (20 ml), dried ($MgSO_4$) and concentrated to give the *cyclohexadiene* **184** (1.287 g, 85 %) as a yellow oil; dec. on silica; ν_{max}/cm^{-1} (film) 3463, 2934 (C-H),

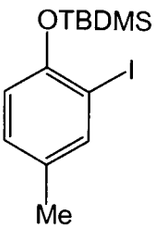
1738 (C=O), 1698 (C=C), 1669 (C=C), 1501, 1447, 1370, 1158 and 1032; δ_H (300 MHz; chloroform-*d*) 5.37 (1 H, d sextet, *J* 3.6 and $^4J_{HH}$ 1.4, 6-H) 4.82 (1 H, app. t, *J* 4.1, 3-H), 4.19 (1 H, dq, $^2J_{HH}$ 10.8 and *J* 7.2, $CH_AH_BCH_3$), 4.14 (1 H, dq, $^2J_{HH}$ 10.8 and *J* 7.2, $CH_AH_BCH_3$), 3.79-3.70 (1 H, m, 1-H), 3.56 (3 H, s, OMe), 2.84 (1 H, dm, $^2J_{HH}$ 22.0, 4-CH_AH_B), 2.66 (1 H, dddd, $^2J_{HH}$ 22.0, $^5J_{HH}$ 6.1, *J* 4.1 and $^4J_{HH}$ 1.4, 4-CH_AH_B), 1.74 (3 H, d, $^4J_{HH}$ 1.4, 5-Me) and 1.26 (3 H, app. t, *J* 7.2 CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 172.6 (C=O), 151.1 (C2), 135.3 (C5), 116.6 (C6), 93.5 (C3), 61.3 (CH_2CH_3), 54.8 (OMe), 47.3 (C1), 31.4 (C4), 23.0 (Me) and 14.4 (CH_2CH_3); *m/z* (EI) 196 (25 %, M^+), 123 (100, M^+ – COOEt), 108 (23, M^+ – COOEt, – Me) and 91 (21, $C_7C_7^+$); HRMS (EI) found: 197.1174; $C_{11}H_{16}O_3$ requires MH^+ 197.1178.

2-Methoxy-5-methyl benzoic acid methyl ester 185



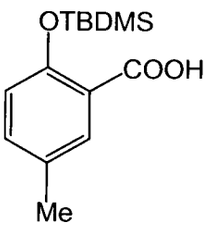
n-Butyllithium (1.27 M solution in hexanes; 14.0 ml, 17.8 mmol) was added dropwise to a stirred solution of the aryl bromide **67** (2.1 ml, 15.80 mmol) in anhydrous THF (35 ml) at –78 °C. The mixture was stirred at –78 °C for 30 minutes, then cannulated dropwise into a stirred solution of methyl chloroformate (5.1 ml, 65.7 mmol) in anhydrous THF (75 ml) also at –78 °C. The reaction mixture was then allowed to warm slowly to room temperature over 19 hours. The reaction mixture was concentrated and the residue partitioned between ethyl acetate (30 ml) and aqueous sodium bicarbonate (30 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 15 ml). The combined organic extracts were washed with brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (gradient elution: 10 to 20 % ethyl acetate in petrol) to give the benzoic ester **185** (2.635 g, 83 %) as a yellow oil; R_f 0.88 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 3001-2838 (CH), 1730 (C=O), 1614 (Ar), 1586 (Ar), 1503 (Ar), 1463 (Ar), 1255 (ArOR), 1083 and 812; δ_H (300 MHz; chloroform-*d*) 7.60 (1 H, d, $^4J_{HH}$ 1.8, 6-H), 7.25 (1 H, dd, *J* 8.5 and $^4J_{HH}$ 1.8, 4-H), 6.87 (1 H, d, *J* 8.5, 3-H), 3.88 (3 H, s, OMe), 3.86 (3 H, s, COOMe) and 2.29 (3 H, s, Ar*Me*); δ_C (75 MHz; chloroform-*d*) 167.2 (C=O), 157.5 (C2), 134.4 (C4), 132.4 (C6), 129.8 (C5), 120.1 (C1), 112.5 (C3), 56.5 (OMe), 52.4 (COO*Me*) and 20.6 (Ar*Me*); *m/z* (FAB) 181 (100 %, MH^+) and 149 (57, M^+ – OMe).

_O_-_t_-Butyldimethylsilyl-2-iodo-4-methyl phenol 187



TBDMS chloride (2.395 g, 15.86 mmol), imidazole (1.075 g, 15.81 mmol) and a catalytic quantity of DMAP were added to a stirred solution of the phenol **105** (2.45 g, 10.47 mmol) in anhydrous DCM (60 ml). The reaction mixture was stirred at room temperature for 19 hours, then poured into water (30 ml). The layers were separated and the aqueous fraction extracted with ether (2 × 10 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried (Na_2SO_4) and concentrated to give the *siloxane* **187** (3.556 g, 98 %) as a pale yellow oil, R_f 0.84 (5 % ethyl acetate in petrol); Found: C, 44.85; H, 5.95; I, 36.45; $C_{13}H_{21}IOSi$ requires C, 44.86; H, 6.08; I, 36.44 %; ν_{max}/cm^{-1} (film) 2885 (C-H), 1597 (Ar), 1486 (Ar), 1283 (Si-Me), 1255 (ArOR), 1038 (Si-O), 920 and 839 (Si-Me); δ_H (300 MHz; chloroform-*d*) 7.58 (1 H, d, $^4J_{HH}$ 1.8, 3-H), 6.97 (1 H, dd, *J* 8.2 and $^4J_{HH}$ 1.8, 5-H), 6.71 (1 H, d, *J* 8.2, 6-H), 2.24 (3 H, s, 4-Me), 1.08 (9 H, s, *t*-Bu) and 0.26 (6 H, s, $SiMe_2$); δ_C (75 MHz; chloroform-*d*) 153.4 (C1), 140.2 (C3), 132.7 (C4), 130.2 (C5), 118.5 (C6), 90.6 (C2), 26.3 (CMe_3), 20.4 (4-Me), 18.8 (CMe_3) and −3.6, ($SiMe_2$); *m/z* (EI) 348 (11 %, M^+), 291 (100, M^+ − *t*-Bu), 206 (12, M^+ − I, − Me), 185 (66), 164 (88, M^+ − *t*-Bu, − I), 149 (62, M^+ − *t*-Bu, − I, − Me), 128 (18), 105 (19) and 73 (25).

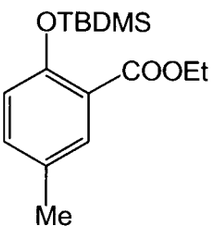
_O_-(_t_-Butlydimethylsilyl)-2-hydroxy-5-methylbenzoic acid 188



n-Butyllithium (1.27 M solution in hexanes; 9.0 ml, 11.4 mmol), was added dropwise to a stirred solution of the aryl iodide **187** (3.559 g, 10.22 mmol) in anhydrous THF (20 ml) at −78 °C. The mixture was stirred at −78 °C for 10 minutes and freshly crushed solid CO_2 (~ 150 ml) was added. The reaction mixture was allowed to warm slowly to room temperature over 18 hours. The reaction mixture was then concentrated and the residue partitioned between ether (30 ml) and aqueous sodium hydroxide (2 M; 30 ml). The layers were separated and the ether layer dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 5 % ethyl acetate in petrol) to give the *benzoic acid* **188** (1.322 mg, 49 %) as a colourless solid, m.p. 28-29 °C; R_f 0.43 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solid phase) 3325 (COOH), 3022-2854 (CH), 1655 (C=O), 1600 (Ar), 1497 (Ar), 1461 (Ar), 1389, 1271 (SiMe), 1255 (ArOR), 1145, 1073 (SiO) and 888 (SiO); δ_H (300 MHz; chloroform-*d*) 7.15 (1 H, d,

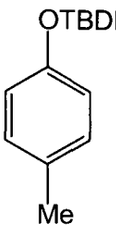
$^4J_{HH}$ 2.2, 6-H), 7.04 (1 H, dd, *J* 8.1 and $^4J_{HH}$ 2.2, 4-H), 6.62 (1 H, d, *J* 8.1, 3-H), 4.92 (1 H, s, COOH), 2.29 (3 H, s, ArMe), 0.93 (9 H, s, *t*-Bu) and 0.34 (6 H, s, 2 × SiMe); δ_C (75 MHz; chloroform-*d*) one peak missing (C=O), 158.6 (C2), 137.1 (C6), 131.3 (C4), 129.0 (C5), 122.5 (C1), 114.8 (C3), 27.0 (C*Me*$_3$), 20.7 (Ar*Me*), 171.7 (*C*Me$_3$) and –4.6 (SiMe); *m/z* (EI) 223 (31 %, MH^+ – COOH), 207 (48, MH^+ – COOH, – Me), 191 (21, M^+ – COOH, –2Me), 165 (100, MH^+ – COOH, – *t*-Bu), 147 (82), 137 (69), 119 (37), 105 (88) and 91 ($C_7H_7^+$); HRMS (EI) found: 223.1516; $C_{14}H_{22}O_3Si$ requires *MH*$^+$ – *CO*$_2$ 223.1518.

2-(*t*-Butyldimethylsilyloxy)-5-methyl benzoic acid ethyl ester 190



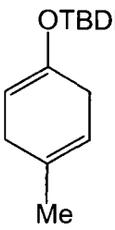
n-Butyllithium (1.14 M solution in hexanes; 0.55 ml, 0.63 mmol) was added dropwise to a stirred solution of the aryl iodide **187** (207 mg, 0.60 mmol) in anhydrous THF (5 ml) at –78 °C. The mixture was stirred at –78 °C for 30 minutes, then cannulated into a stirred solution of ethyl chloroformate (220 µl, 2.30 mmol) in anhydrous THF (10 ml), also at –78 °C. The reaction mixture was stirred at –78 °C for 30 minutes, then at room temperature for 4 hours. The mixture was concentrated and the residue partitioned between ethyl acetate (10 ml) and aqueous sodium bicarbonate (10 %; 10 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 5ml). The combined organic extracts were washed with brine (10 ml), dried (Na_2SO_4) and concentrated to give the *siloxane* **190** (132 mg, 75 %) as a yellow oil; R_f 0.49 (5 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2928-2857 (CH), 1760 (C=O), 1583 (Ar), 1509 (Ar), 1471, 1240 (ester CO), 1051 (SiO) and 837 (SiMe); δ_H (300 MHz; chloroform-*d*) 7.23 (1 H, d, $^4J_{HH}$ 1.5, 6-H), 7.19 (1 H, dd, *J* 8.2 and $^4J_{HH}$ 1.5, 4-H), 7.03 (1 H, d, *J* 8.2, 3-H), 4.29 (2 H, q, *J* 7.0, *CH*$_2$CH$_3$), 2.34 (3 H, s, Ar*Me*), 1.37 (3 H, t, *J* 7.0, CH$_2$*CH*$_3$), 0.88 (9 H, s, *t*-Bu) and 0.29 (6 H, s, SiMe$_2$); δ_C (75 MHz; chloroform-*d*) 154.6 (C=O), 154.2 (C2), 137.2 (C6), 135.0 (C5), 131.5 (C4), 128.9 (C1), 121.8 (C3), 64.9 (CH$_2$), 27.0 (C*Me*$_3$), 21.4 (Ar*Me*), 17.8 (*C*Me$_3$), 14.8 (CH$_2$*CH*$_3$) and –4.6 (SiMe$_2$); *m/z* (EI) 291 (27 %, M^+ – 3H), 237 (81, M^+ – *t*-Bu), 209 (100), 149 (71, M^+ – TBDMS, – 2Me) and 105 (37); *m/z* (ES) 312 (60 %, MOH^+) and 295 (100, MH^+); HRMS (EI) found: 237.0959; $C_{16}H_{26}O_3Si$ requires M^+ – *t*-Bu 237.0947.

_O_-_t_-Butyldimethylsilyl-4-methyl phenol 193



TBDMS chloride (1.063 g, 7.04 mmol), imidazole (473 mg, 6.96 mmol) and a catalytic quantity of DMAP were added to a stirred solution of *p*-cresol **20** (1.027 g, 9.51 mmol) in anhydrous DCM. The reaction mixture was stirred at room temperature for 20 hours, then poured into water. The layers were separated and the aqueous fraction extracted with ether (3 × 10 ml). The combined organic extracts were washed with water (10 ml), aqueous hydrochloric acid (2 M; 10 ml), water (10 ml), aqueous sodium hydroxide (2 M; 10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give the siloxane **193** (1.335 g, 85 %) as a pale yellow oil; R_f 0.55 (5 % toluene in petrol); ν_{max}/cm^{-1} (film) 3028-2859 (CH), 1612 (Ar), 1582 (Ar), 1510 (Ar), 1255 (SiMe) and 915; δ_H (300 MHz; chloroform-*d*) 7.03 (2 H, d, *J* 8.2, 2 × 3-H), 6.74 (2 H, d, *J* 8.2, 2 × 2-H), 2.28 (3 H, s, Me), 0.99 (9 H, s, *t*-Bu) and 0.19 (6 H, s, $SiMe_2$); δ_C (75 MHz; chloroform-*d*) 153.4 (C1), 130.6 (C4), 130.0 (C3), 119.9 (C2), 25.8 (C*Me*$_3$), 20.7 (Ar*Me*), 18.3 (*C*Me_3) and –4.3 ($SiMe_2$); *m/z* (EI) 222.5 (15 %, M^+), 165 (100, $M^+ - t$-Bu) and 91 (17 ($C_7H_7^+$).

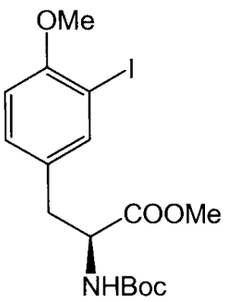
_O_-(_t_-Butyldimethylsilyl)-4-methyl cyclohexa-1,4-dienol 194



Lithium (sufficient to maintain a blue colour) was added to a stirred solution of the silyl-protected phenol **193** (500 mg, 2.25 mmol) in anhydrous THF (2 ml) and anhydrous ammonia (~15 ml). The reaction mixture was stirred at reflux for 30 minutes, then anhydrous methanol (0.5 ml) was added dropwise, with more lithium added as necessary to maintain the blue colour. The reaction mixture was stirred at reflux for a further 30 minutes. Anhydrous methanol (sufficient to displace the blue colour) was added and the ammonia allowed to evaporate. The residue was partitioned between ether (10 ml) and water (15 ml). The layers were separated and the aqueous fraction extracted with ether (2 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried (Na_2SO_4) and concentrated to give the cyclohexadiene **194** (255 mg, 51 %) as a yellow oil; R_f 0.39 (20 % ethyl acetate in petrol); ν_{max}/cm^{-1} (film) 2956-2857 (CH), 1701 (C=C), 1668 (C=C), 1510, 1254 (ROR), 1203, 883 (CH) and 838 (CH); δ_H (300 MHz; chloroform-*d*) 5.33 (1 H, m, 5-H), 4.83 (1 H, m, 2-H), 2.64 (4 H, br s, 2 × CH_2), 1.67 (3 H, br s, Me), 0.92 (9 H, s, *t*-Bu) and 0.14 (6 H, s, $SiMe_2$); δ_C (75 MHz; chloroform-*d*) 148.4 (C1), 131.5 (C4), 118.3 (C5), 101.1 (C2), 31.7 & 31.4 (C3 & C6),

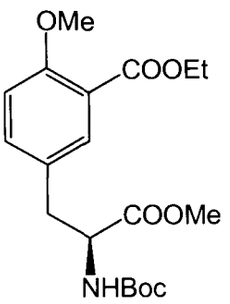
25.8 (C*Me*$_3$), 22.9 (Me), 18.1 (*C*Me$_3$) and –4.3 (SiMe$_2$); *m/z* (EI) 224 (10 %, M$^+$), 165 (100), 97 (17) and 75 (49).

O-Methyl-*N*-*t*-butoxycarbonyl-3-iodo-L-tyrosine methyl ester 195



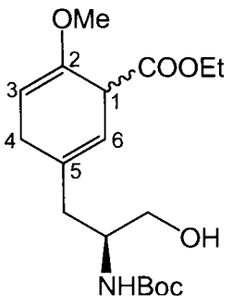
Potassium carbonate (11.93 g, 86.45 mmol) and methyl iodide (3.2 ml, 51.4 mmol) were added to a stirred solution of the carbamate **71** (7.04 g, 17.30 mmol) in acetone (150 ml). The reaction mixture was then heated at reflux for 44 hours and concentrated. The residue was partitioned between ethyl actetate (100 ml) and water (100 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 20 ml). The combined organic extracts were washed with aqueous sodium hydroxide (2 M; 50 ml), aqueous sodium thiosulfate (2 M; 50 ml) and brine (50 ml), dried ($MgSO_4$) and concentrated to give the protected iodotyrosine **195** (6.536 g, 86 %) as a yellow oil; R_f 0.30 (20 % ethyl acetate in petrol); $[\alpha]_D$ +7.6 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (film) 3427 (NH), 2977 (CH), 1740 (ester C=O), 1713 (carbamate C=O), 1599 (Ar), 1491 (Ar), 1366, 1254 (ArOR), 1165, 1049 and 1018; δ_H (300 MHz; chloroform-*d*) 7.53 (1 H, d, $^4J_{HH}$ 1.9, 2-H), 7.07 (1 H, dd, *J* 8.4 and $^4J_{HH}$ 1.9, 6-H), 6.75 (1 H, d, *J* 8.4, 5-H), 5.00 (1 H, br d, *J* 8.0, NH), 4.53 (1 H, d app. t, *J* 8.0 and 5.6, CH), 3.85 (3 H, s, OMe), 3.73 (3 H, s, COOMe), 3.05 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 5.6 C*H*$_A$H$_B$), 2.95 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 5.6, CH$_A$*H*$_B$) and 1.44 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 172.5 (*C*OOMe), 157.6 (C4), 155.4 (NHCO), 140.7 (C2), 130.7 & 130.6 (C1 & C6), 111.2 (C5), 86.3 (C3), 80.5 (*C*Me$_3$), 56.8 (COO*Me*), 54.9 (CH), 52.7 (OMe), 37.3 (CH_2) and 28.7 (C*Me*$_3$); *m/z* (FAB) 436 (17 %, MH$^+$), 380 (90, MH$^+$ – *t*-Bu), 336 (100, MH_2^+ – Boc), 276 (24, MH$^+$ – Boc, – COOMe) and 247 (41, $MeOC_6H_3ICH_2^+$).

_O_-Methyl-_N_-_t_-butoxycarbonyl-3-ethoxycarbonyl-L-tyrosine methyl ester 196



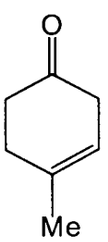
Palladium acetate (12 mg, 0.05 mmol), thallium acetate (138 mg, 0.52 mmol), triphenylphosphine (29 mg, 0.11 mmol) and triethylamine (80 µl, 0.58 mmol) were added to a stirred solution of protected iodotyrosine **195** (204 mg, 0.47 mmol) in anhydrous degassed ethanol (5.5 ml). The reaction mixture was stirred under an atmosphere of carbon monoxide at 60 °C for 30 hours. The reaction mixture was then cooled to room temperature, filtered through celite and washed through with methanol. The filtrate was concentrated and the residue partitioned between ethyl acetate (10 ml) and water (10 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with water (5 ml) and brine (5 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by flash chromatography (eluent: 30 % ethyl acetate in petrol) to give the *ester* **196** (105 mg, 58 %) as a yellow oil; R_f 0.37 (30 % ethyl acetate in petrol); $[\alpha]_D$ +6.4 (*c* 1.5 in methanol); ν_{max}/cm^{-1} (film) 3366 (NH), 2978 (CH), 1711br (C=O), 1614 (Ar), 1582 (Ar), 1503 (Ar), 1261 (ArOR) and 1081; δ_H (300 MHz; chloroform-*d*) 7.54 (1 H, d, $^4J_{HH}$ 1.9, 2-H), 7.23 (1 H, dd, *J* 8.5 and $^4J_{HH}$ 1.9, 6-H), 6.92 (1 H, d, *J* 8.5, 5-H), 5.01 (1 H, br d, *J* 7.8, NH), 4.56 (1 H, d app. t, *J* 7.8 and 5.9, CH), 4.35 (2 H, q, *J* 7.2, CH_2CH_3), 3.89 (3 H, s, OMe), 3.73 (3 H, s, COOMe), 3.10 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 5.9, CH_AH_BCH), 3.01 (1 H, dd, $^2J_{HH}$ 13.8 and *J* 5.9, CH_AH_BCH), 1.42 (9 H, s, *t*-Bu) and 1.38 (3 H, t, *J* 7.2, CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 172.6 (*C*OOMe), 166.3 (*C*OOEt), 158.7 (C4), 155.4 (NHCO), 134.5 (C6), 132.7 (C2), 128.0 (C1), 120.7 (C3), 112.7 (C5), 80.4 (CMe_3), 61.3 (CH_2CH_3), 56.5 (OMe), 54.8 (CH), 52.7 (COO*Me*), 37.7 (CH_2CH), 28.7 (CMe_3) and 17.7 (CH_2CH_3); *m/z* (ES) 404 (74 %, MNa^+), 348 (72, $MNaH^+$ – *t*-Bu), 280 (67, M^+ – Boc) and 236 (100); HRMS (ES) found: 404.1657; $C_{19}H_{27}NO_7$ requires MNa^+ 404.4685.

(1*R/S*)(2'*S*)-*N*-(*t*-Butoxycarbonyl)-5-(2'-amino-3'-hydroxypropyl)-2-methoxy-cyclohexa-2,5-dienecarboxylic acid ethyl ester 197



Ammonia (~60 ml) was freshly distilled onto a solution of the benzoic ester **196** (1.267 g, 3.32 mmol) and *t*-butanol (480 µl, 5.03 mmol) in anhydrous THF (10 ml). Potassium (sufficient to maintain a blue colour) was added and the reaction mixture stirred at −78 °C for 2 hours. The reaction was quenched by the addition of ammonium chloride (sufficient to displace the blue colour) and the ammonia removed in a stream of nitrogen. The residue was partitioned between ether (20 ml) and water (20 ml). The layers were separated and the aqueous fraction extracted with ether (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give the *cyclohexadiene* **197** (949 mg, 80 %) as a yellow solid, m.p. 30-32 °C; R_f 0.18 (40 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solid phase) 3371 (OH), 2976 (CH), 1701br (ester & carbamate C=O), 1615 (C=C), 1581 (C=C), 1509, 1366, 1247 (ROR), 1171 and 1030 (OH); δ_H (500 MHz; chloroform-*d*) 5.47 (1 H, d, *J* 10.7, 6-H), 4.80 (1 H, app. q, *J* 3.9, 3-H), 4.21-4.12 (2 H, m, CH_2CH_3), 3.90-3.88 (1 H, m, $CHCH_2$), 3.84-3.62 (3 H, m, 1-H & CH_2OH), 3.67 (3 H, s, OMe), 3.04-2.62 (2 H, m, 4-CH_2), 2.31 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 6.8, $CH_AH_BCHCH_2OH$), 2.22 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 7.7, $CH_AH_BCHCH_2OH$), 1.44 & 1.43 (9 H, 2 × s, *t*-Bu) and 1.27 (3 H, s, CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 172.3 & 172.2 (*C*OOEt), 166.6 (NH*C*OO*t*-Bu), 156.6 & 156.5 (C2), 139.5 (C5), 119.7 & 119.6 (C6), 93.4 & 93.3 (C3), 80.2 (CMe_3), 65.7 (CH_2OH), 61.5 & 61.3 (CH_2CH_3), 56.6 & 56.5 ($CHCH_2$), 54.9 (OMe), 47.8 & 47.2 (C1), 38.9 (CH_2CH), 29.8 & 29.5 (C4), 28.7 (CMe_3) and 14.7 & 14.6 (CH_2CH_3); *m/z* (ES) 709 (5 %, $M_2^+ - H$), 356 (10, MH^+), 256 and (100, MH_2^+ − Boc); HRMS (ES) found: 356.2067; $C_{18}H_{29}NO_6$ requires MH^+ 356.2068.

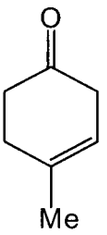
4-Methyl cyclohex-3-enone 198



Aqueous phosphate buffer (pH 6; 30 ml), was added to a stirred solution of the enol ether **120** (1.005 g, 8.24 mmol) in ether (20 ml). The mixture was acidified to pH 2 using aqueous phosphoric acid (conc.) and stirred at room temperature for 3 days. The reaction mixture was poured into water (10 ml), separated and the aqueous fraction extracted with ether (3 × 10 ml). The

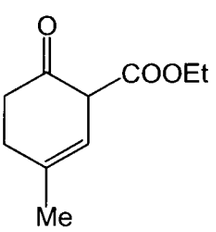
combined organic extracts were washed with water (15 ml) and brine (15 ml), dried ($MgSO_4$) and concentrated to give the crude enone **198** (3:1 enone:aromatic; 754 mg, 61%) as a pale yellow oil. A small amount was purified by flash chromatography (eluent: 10 % ether in petrol) for analytical purposes to give the β,γ-enone **198** as a colourless oil; R_f 0.40 (10 % ethyl acetate in petrol); ν_{max}/cm^{-1} (solution in chloroform-*d*) 3412, 2970, 2929, 1717 (C=O), 1442 and 1196; δ_H (300 MHz; chloroform-*d*) 5.44 (1 H, t sextet, *J* 4.1 and $^4J_{HH}$ 1.6, 3-H), 2.84-2.83 (2 H, m, 2-CH_2), 2.52-2.38 (4 H, m, 5-CH_2 & 6-CH_2) and 1.78 (3 H, s, Me); δ_C (75 MHz; chloroform-*d*) 211.7 (C1), 135.1 (C4), 118.6 (C3), 40.0 (C2), 38.9 (C6), 30.7 (C5) and 23.5 (Me); *m/z* (EI) 111 (53 %, MH^+), 97 (79, MH^+ – Me), 71 (82) and 57 (100).

4-Methyl cyclohex-3-enone 198



A solution of oxalic acid dihydrate (55 mg, 0.44 mmol) in water (2 ml) was added to a solution of the enol ether **120** (52 mg, 0.42 mmol) in methanol (2 ml). The reaction mixture was stirred at room temperature for 45 minutes, then concentrated. The aqueous residue was extracted with ethyl acetate (3 × 5 ml) and the combined organic extracts washed with water (5 ml), aqueous sodium bicarbonate (10 %; 5 ml) and brine (5 ml), dried ($MgSO_4$) and concentrated to give the β,γ-enone **198** (20 mg, 44 %) as a pale yellow oil, spectroscopically identical to that obtained previously.

3-Methyl-6-oxo-cyclohex-2-enecarboxylic acid ethyl ester 202



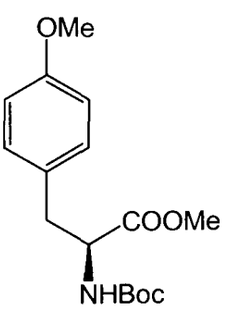
A solution of oxalic acid dihydrate (880 mg, 6.99 mmol) in water (50 ml) was added to a stirred solution of enol ether **184** (1.234 g, 6.34 mmol) in methanol (50 ml). The reaction mixture was stirred at room temperature for 27 hours, after which time the methanol was evaporated and the residue extracted with ether (4 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml), dried ($MgSO_4$) and concentrated to give the crude *β,γ-enone* **202** as a yellow oil (584 mg, 51 %). A small amount of the crude product was then purified by reduced pressure distillation using a Kugelröhr (b.p. 150 °C/4 mm Hg) to give the *β,γ-enone* (obtained as a 1:1 mixture with 2-hydroxy-4-methyl benzoic acid ethyl ester due to rearomatisation) as yellow oil; R_f 0.24 (30 % ethyl acetate in petrol); ν_{max} cm^{-1} (film) 1738 (ester C=O),

1731 (ketone C=O), 1645 (C=C), 1369, 1260 and 1023; δ_H (500 MHz; chloroform-*d*) 5.52 (1 H, app. sextet, *J* 1.3 2-H), 4.24 (1 H, q, *J* 7.2, $CH_AH_BCH_3$), 4.23 (1 H, q, *J* 7.2, $CH_AH_BCH_3$), 3.96 (1 H, s, 1-H), 3.00 (1 H, d app. t, $^2J_{HH}$ 14.5 and *J* 8.1, 5-CH_AH_B), 2.68-2.59 (2 H, m, 5-CH_AH_B & 4-CH_AH_B), 2.49 (1 H, d app. t, $^2J_{HH}$ 17.5 and *J* 8.6, 4-CH_AH_B), 1.84 (3 H, s, 3-Me) and 1.29 (3 H, t, *J* 7.2, CH_2CH_3); δ_C (75 MHz; chloroform-*d*) 206.6 (C6), 170.6 (*C*OOEt), 141.3 (C3), 122.7 (C2), 63.1 (CH_2CH_3), 54.5 (C1), 35.5 (C5), 31.7 (C4), 23.5 (Me) and 14.4 (CH_2CH_3); *m/z* (ES) 196 (20 %), 183 (100, MH^+) and 155 (MH_2^+ – Et).

Radical Coupling of 3-Methyl-6-oxo-cyclohex-2-enecarboxylic acid ethyl ester 202

Copper(II) acetate monohydrate (69 mg, 0.34 mmol) was added to a stirred solution of the β-ketoester x (48 mg, 0.27 mmol) in dry ethanol (5ml). The reaction mixture was stirred at room temperature for 2 hours then concentrated. The residue was partitioned between ethyl acetate (5 ml) and water (5 ml) and the layers separated. The aqueous fraction was extracted with ethyl acetate (2 × 2 ml) and the combined organic extracts were washed with water (2 ml) and brine (2 ml), dried ($MgSO_4$) and evaporated to give a yellow oil (57 mg).

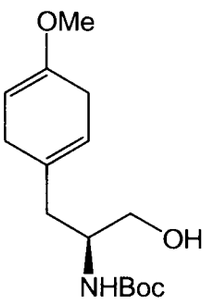
***O*-Methyl-*N*-*t*-butoxycarbonyl-L-tyrosine methyl ester 237**



Potassium carbonate (7.354 g, 53.29 mmol) and methyl iodide (2.0 ml, 32.1 mmol) were added to a stirred solution of the carbamate **16** (2.984 g, 10.62 mmol) in acetone (100 ml). The reaction mixture was heated at reflux for 21 hours, then cooled to room temperature and filtered. The filtrate was concentrated and the residue partitioned between ethyl acetate (40 ml) and water (60 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 10 ml). The combined organic extracts were washed with water (20 ml) and brine (20 ml), dried ($MgSO_4$) and concentrated to give the protected tyrosine **237** (3.292 g, 100 %) as a yellow oil; R_f 0.58 (30 % ethyl acetate in petrol); $[\alpha]_D$ +4.6 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (film) 3370 (NH), 2977 (C-H), 1747 (ester C=O), 1715 (carbamate C=O), 1613 (Ar), 1584 (Ar), 1442 (Ar), 1366, 1249 (ArOR), 1166 and 1034; δ_H (300 MHz; chloroform-*d*) 7.04 (2 H, d, *J* 8.7, 2 × 2-H), 6.83 (2 H, d, *J* 8.7, 2 × 3-H), 4.99 (1 H, d, *J*

7.9, NH), 4.54 (1 H, d app. t, *J* 7.9 and 6.1, CH), 3.78 (3 H, s, OMe), 3.71 (3 H, s, COOMe), 3.06 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 6.1, CH_AH$_B$), 2.99 (1 H, dd, $^2J_{HH}$ 14.1 and *J* 6.1, CH$_A$$H_B$) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 172.9 (C=O), 159.0 (C4), 155.4 (NHCO), 130.7 (C2), 128.3 (C1), 114.4 (C3), 80.3 (*C*Me$_3$), 55.6 (OMe), 54.9 (CH), 52.6 (COO*Me*), 37.9 (CH_2) and 28.7 C*Me*$_3$); *m/z* (FAB) 619 (30 %, M_2H^+), 310 (18, MH^+), 254 (MH^+ – *t*-Bu), 210 (100, MH_2^+ – Boc), 194 (70, MH^+ – Boc, – Me), 150 (32, MH^+ – Boc, – COOMe) and 121 (97, $MeOC_6H_4CH_2^+$).

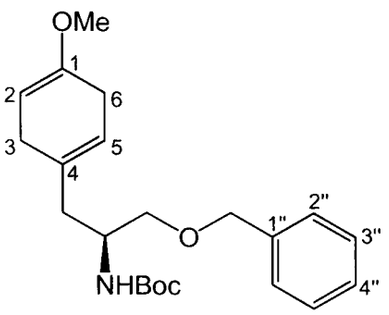
_N_-(*t*-Butoxycarbonyl)-2-amino-3-(4'-methoxycyclohexa-1',4'-dienyl)-propan-1-ol 238



Ammonia (~100 ml) was distilled onto a solution of the protected tyrosine **237** (3.292 g, 10.64 mmol) in anhydrous THF (20 ml). Lithium (sufficient to maintain a blue colour) was added and the reaction mixture stirred at reflux for 30 minutes. Anhydrous methanol (10.6 ml) was added slowly to the mixture, with more lithium added as necessary to maintain the blue colour. The reaction mixture was then stirred at reflux for a further 60 minutes. Anhydrous methanol (sufficient to displace the blue colour) was then added and the ammonia was removed in a stream of nitrogen. The residue was partitioned between ether (40 ml) and water (60 ml). The layers were separated and the aqueous fraction extracted with ether (3 × 10 ml). The combined organic extracts were washed with water (10 ml) and brine (10 ml) dried ($MgSO_4$) and concentrated to give the *cyclohexadiene* **238** (2.857 g, 95 %) as a yellow oil; R_f 0.38 (30 % ethyl acetate in petrol); $[\alpha]_D$ –1.9 (*c* 1.1 in methanol); ν_{max}/cm^{-1} (film) 3398 (OH), 3932 (CH), 1695 (C=O), 1513 (NH/CN), 1366, 1251 (ROR), 1217, 1172 and 1056 (CH_2OH); δ_H (500 MHz; methanol-d_4) 5.47 (1 H, br s, 5'-H), 4.68 (1 H, br s, 2'-H), 3.74-3.68 (1 H, m, C*H*CH$_2$OH), 3.54 (3 H, s, OMe), 3.51 (1 H, dd, $^2J_{HH}$ 11.6 and *J* 5.6, CH_AH$_B$OH), 3.48 (1 H, dd, $^2J_{HH}$ 11.6 and *J* 6.0, CH$_A$$H_B$OH), 2.90-2.81 (1 H, m, 3'-CH_AH$_B$), 2.74 (1 H, ddd, $^2J_{HH}$ 29.1, *J* 7.7 and $^4J_{HH}$ 2.6, 3'-CH$_A$$H_B$) 2.70 (2 H, br s, 6'-$CH_2$), 2.27 (1 H, dd, $^2J_{HH}$ 13.7 and *J* 5.1, CH_AH$_B$CHCH$_2$OH), 2.08 (1 H, dd, $^2J_{HH}$ 13.7 and *J* 9.4, CH$_A$$H_B$CHCH$_2$OH) and 1.44 (9 H, s, *t*-Bu); δ_C (125 MHz; methanol-d_4) 158.5 (NHCO), 154.3 (C1'), 133.9 (C4'), 121.5 (C5'), 91.7 (C2'), 80.2 (*C*Me$_3$), 65.5 (*C*H$_2$OH), 54.5 (OMe), 52.3 (*C*HCH$_2$OH), 40.2 (*C*H$_2$CHCH$_2$OH), 30.5 (C3' & C6') and 29.1 (C*Me*$_3$); *m/z* (ES) 588 (78 %, M_2Na^+ – H), 347 (60), 306 (65, MNa^+), 228 (34) and

184 (100, $MHNa^+$ – O*t*-Bu, – OMe, – OH); HRMS (ES) found: 306.1693; $C_{15}H_{25}NO_4$ requires MNa^+ 306.1681.

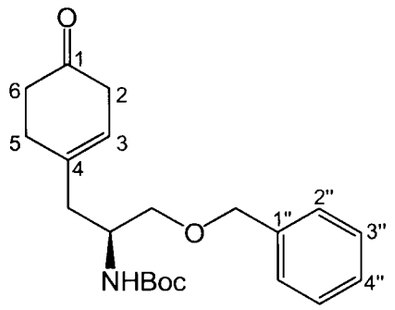
(*S*)-*O*-Benzyl-*N*-(*t*-butoxycarbonyl)-4-(2'-amino-3'-hydroxypropyl)-1-methoxycyclohexa-1,4-diene 239



TBAI (575 mg, 1.56 mmol) and sodium hydride (60 % dispersion in mineral oil; 1.386 g, 34.65 mmol) were added to a stirred solution of the alcohol **238** (8.767 g, 30.92 mmol) in anhydrous THF (100 ml) at 0 °C. The mixture was allowed to warm to room temperature over 45 minutes, then stirred at room temperature for 15 minutes. The mixture was cooled to 0 °C and benzyl bromide (4.1 ml, 34.48 mmol) was added dropwise. The reaction mixture was allowed to warm slowly to room temperature and stirred for 23 hours. The reaction was quenched by addition of water (20 ml), the mixture concentrated and the residue partitioned between ether (30 ml) and water (30 ml). The layers were separated and the aqueous fraction extracted with ether (3 × 10 ml). The combined organic extracts were washed with brine (10 ml), dried ($MgSO_4$) and concentrated to give the crude ether **239** (9.9807 g, 86 %) as a yellow oil. A small amount was purified by flash chromatography (eluent: 10 % ethyl acetate in petrol) for analytical purposes to give the *benzyl ether* **239** as a colourless oil; R_f 0.13 (10 % ethyl acetate in petrol); $[\alpha]_D$ −12.8 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (film) 3360 (NH), 2930 (OH), 1712 (C=O), 1610 (C=C), 1496, 1365, 1216 and 1171; δ_H (500 MHz; chloroform-*d*) 7.36-7.29 (5 H, m, Ph), 5.39 (1 H, s, 2-H), 4.75 (1 H, br s, NH), 4.60 (1 H, s, 5-H), 4.54 (1 H, d, $^2J_{HH}$ 12.0, CH_AH$_B$Ph), 4.47 (1 H, d, $^2J_{HH}$ 12.0, CH$_A$$H_B$Ph), 3.87 (1 H, m, C*H*NH), 3.54 (3 H, s, OMe), 3.49 (1 H, s, CH_AH$_B$OBn), 3.48-3.46 (1 H, m, CH$_A$$H_B$OBn), 2.78-2.76 (4 H, m, 3-CH$_2$ & 6-CH$_2$), 2.30 (1 H, dd, $^2J_{HH}$ 13.7 and *J* 7.7, CH_AH$_B$CHCH$_2$OBn), 2.20 (1 H, dd, $^2J_{HH}$ 13.7 and *J* 7.2, CH$_A$$H_B$CHCH$_2$OBn) and 1.45 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 155.9 (NHCO), 153.1 (C4), 138.6 (C1"), 132.5 (C1), 130.8 (C4"), 128.8 (C3"), 128.0 (C2"), 121.0 (C2), 90.9 (C5), 79.6 (*C*Me$_3$), 73.6 (*C*H$_2$Ph), 71.6 (*C*H$_2$OBn), 54.3 (OMe), 48.9 (CHCH$_2$OBn), 39.8 (*C*H$_2$CHCH$_2$OBn), 29.6 (C3 & C6) and 28.8 (C*Me$_3$*); *m/z* (ES) 396 (70 %, MNa^+), 340 (28, $MNaH^+$ – *t*-Bu), 274 (100, MH_2^+ – Boc), 242 (48, MH^+ – Boc, – OMe) and 202

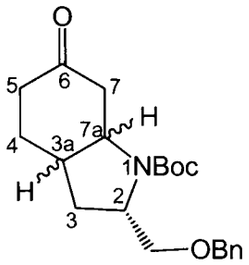
($MNaH^+$ – *t*-Bu, – OMe, – OBn); HRMS (ES) found: 396.2145; $C_{22}H_{31}NO_4$ requires MNa^+ 396.2151.

(*S*)-*O*-Benzyl-*N*-(*t*-Butoxycarbonyl)-4-(2'-amino-3'-benzyloxy-propyl)cyclohex-3-enone 240



A solution of oxalic acid dihydrate (3.520 g, 27.94 mmol) in water (56 ml) was added to a stirred solution of the enol ether **239** (9.469 g, 26.096 mmol) in acetone (56 ml). The reaction mixture was stirred for 18 hours at room temperature, then concentrated and the residue extracted with ether (3 × 30 ml). The combined organic extracts were washed with brine (20 ml), dried ($MgSO_4$) and concentrated to give a crude product which was then purified by flash chromatography (eluent: 20 % ethyl acetate in petrol) to give the *β,γ-enone* **240** (2.810 g, 30 %) as a yellow oil; R_f 0.46 (30 % ethyl acetate in petrol); $[\alpha]_D$ –2.8 (*c* 1.0 in methanol); ν_{max}/cm^{-1} (film) 3367 (NH), 2978 (CH), 1738 (carbamate C=O), 1713 (ketone C=O), 1498 (Ar), 1454 (Ar), 1367, 1243, 1171 and 1047; δ_H (300 MHz; chloroform-*d*) 7.38-7.30 (5 H, m, Ph), 5.46 (1 H, app. t, *J* 3.6, 3-H), 4.81 (1 H, d, *J* 8.2, NH), 4.56 (1 H, d, $^2J_{HH}$ 11.9, CH_AH_BPh), 4.49 (1 H, d, $^2J_{HH}$ 11.9, CH_AH_BPh), 3.90 (1 H, m, C*H*NH), 3.49 (2 H, d, *J* 3.5, CH_2OBn), 2.81 (2 H, m, 2-CH_2), 2.55-2.23 (6 H, m, 5-CH_2, 6-CH_2 & 1'-CH_2) and 1.42 (9 H, s, *t*-Bu); δ_C (75 MHz; chloroform-*d*) 210.8 (C1), 155.5 (NHCO), 138.0 (C1"), 135.7 (C4), 128.4 (C3"), 127.8 (C4"), 127.7 (C2"), 120.9 (C3), 79.3 (CMe_3), 73.3 (CH_2Ph), 71.5 (CH_2OBn), 40.2 & 38.7 (CH_2CHCH_2OBn, C5 & C6), 39.7 (C2) and 28.4 (CMe_3); *m/z* (ES) 773 (20 %), 741 (5, M_2Na^+), 687 (15), 649 (25), 444 (65), 412 (35), 382 (100, MNa^+), 367 (40, MNa^+ – Me), 326 (85, $MNaH^+$ – *t*-Bu) and 282 ($MNaH^+$ – Boc); HRMS (ES) found: 382.2004; $C_{21}H_{29}NO_4$ requires MNa^+ 382.1994.

(4a*R/S*)(7a*R/S*)(2*S*)-*O*-Benzyl-*N*-(*t*-butoxycarbonyl)-2-hydroxymethyl-octahyro-indol-6-one 241



Potassium carbonate (293 mg, 2.13 mmol) was added to a stirred solution of the β,γ-enone **240** (505 mg, 1.41 mmol) in anhydrous methanol (25 ml). The reaction mixture was stirred at room temperature for 18 hours, then concentrated and the residue partitioned between ethyl acetate (10 ml) and water (10 ml). The layers were separated and the aqueous fraction extracted with ethyl acetate (3 × 5 ml). The combined organic extracts were washed with water (5 ml) and brine (5 ml), dried ($MgSO_4$) and concentrated to give a crude product which was purified by hplc (gradient elution: 30 to 70 % acetontitrile in water over 30 minutes; eluting 20 ml/minute from a ThermoHypersil 250 × 21.2 mm 8 μm Hyperprep HS C18 column; peaks observed at 200 nm) to give the *bicyclic ketone* **241** as a colourless solid (42 mg, 10 %) as a mixture of diastereoisomers: R_f 0.23 (30 % ethyl acetate in petrol); δ_H (500 MHz; chloroform-*d*) 7.34-7.27 (5 H, m, Ph), 4.55 1 H, d, $^2J_{HH}$ 13.1, CH_AH$_B$Ph), 4.50 (1 H, d, $^2J_{HH}$ 13.1, CH$_A$$H_B$Ph), 4.17-4.10 (1 H, m, 2-H or 7a-H), 4.05-3.98 (1 H, m, 2-H or 7a-H), 3.68-3.40 (2 H, m, CH_2OBn), 3.03 & 2.88 (1 H, 2 × dd, $^2J_{HH}$ 15.2 and *J* 5.2, 7-CH_AH$_B$), 2.76-2.74 & 2.58 (1 H, m & dd, $^2J_{HH}$ 14.1 and *J* 10.7, 7-CH$_A$$H_B$), 2.45-2.38 (1 H, m, 5-CH_AH$_B$), 2.28 (1 H, app. q, *J* 12.0, 5-CH$_A$$H_B$), 2.20-2.09 (3 H, m, 3-CH_AH$_B$ & 4-CH$_2$), 2.07-1.96 (1 H, m, 3a-H), 1.81 & 1.76 (1 H, 2 × app. dd, $^2J_{HH}$ 13.7 and *J* 7.3, 3-CH$_A$$H_B$) and 1.46 & 1.43 (9 H, 2 × s, *t*-Bu); ;δ_C (125 MHz; chloroform-*d*) 210.7 & 210.6 (C6), 154.0 & 153.6 (NHCO), 138.5 & 138.3 (C1'), 128.4 & 128.4 (C3'), 127.6 & 127.5 (C4'), 127.5 & 127.5 (C2'), 80.0 & 79.8 (*C*Me$_3$), 73.4 & 73.7 (*C*H$_2$Ph), 70.9 & 70.3 (*C*H$_2$OBn), 58.0, 57.0, 56.9 & 56.6 (C2 & C7a), 43.5 & 42.0 (C7), 36.7 & 36.6 (C5), 34.6 & 33.5 (C3a), 28.5 & 28.4 (C*Me$_3$*) and 24.8, 24.6 & 24.4 (C3 & C4); *m/z* (ES) 741 (10 %, M_2Na^+), 360 (45, MH^+), 304 (100, MH_2^+ – *t*-Bu) and 260 (MH_2^+ – Boc); HRMS (ES) found: 382.2003; $C_{21}H_{29}NO_4$ requires MNa^+ 382.1994.

6.3 Computational Details

Molecular Modelling was carried out using MacroModel v. 8.0 within Maestro v. 5.0.019 on a Silicon Graphics workstation.[204, 205] The MM2* forcefield was used and the Monte Carlo method was used for conformational searching.[206] Calculations were

carried using gas-phase parameters, PRCG minimisation converging on gradient and distance-dependent dielectrics.

REFERENCES

1 S. Ömura, S. Hirano, Y. Iwai and R. Masuma, *J. Antibiotics*, 1979, **32**, 768.

2 T. Kagata, H. Shigemori, Y. Mikami and J. Kobayashi, *J. Nat. Prod.*, 2000, **63**, 886.

3 A. Furusaki, T. Matsumoto, H. Ogura, H. Takayanagi, A. Hirano and S. Ömura, *J. Chem. Soc., Chem. Commun.*, 1980, 698.

4 Y. Enomoto, K. Shiomi, M. Hayashi, R. Masuma, T. Kawakubo, K. Tomosawa, Y. Iwai and S. Ömura, *J. Antibiotics*, 1996, **49**, 50.

5 *Textbook of Biochemistry with Clinical Correlations*, 5th edition, ed. T. M. Devlin, Wiley-Liss, New York, 2002.

6 F. Dreyfuss and J. Zahavi, *Atherosclerosis*, 1973, **17**, 107.

7 J. Y. T. Lam, J.-G. Latour, J. Lespérance and D. Waters, *Am. J. Cardiology*, 1994, **73**, 333.

8 M. Koltai, D. Hosford, P. Guinot, A. Esanu and P. Braquet, *Drugs*, 1991, **42**, 9.

9 L. Stryer, *Biochemistry*, 4th edition, W. H. Freeman and Company, New York, 1995.

10 C. Gachet, *Thromb. Haemost.*, 2001, **86**, 222.

11 D. L. Bhatt and E. J. Topol, *Nature Reviews Drug Discovery*, 2003, **2**, 15.

12 S. P. Kunapuli, *Trends Pharmacol. Sci.*, 1998, **19**, 391.

13 M. A. Packham, *Can. J. Physiol. Pharmacol.*, 1994, **72**, 278.

14 R. N. Puri, *Biochem. Pharmacol.*, 1999, **57**, 851.

15 M.-R. Mueller, A. Salat, S. Pulaki, D. Boehm and P. Sangl, Centre for Biomedical Research, University of Vienna, The Platelet Homepage, www.akh-wien.ac.at/biomed-research/htx/platweb1.htm, 1996.

16 P. Savi, J. M. Pereillo, M. F. Uzabiaga, J. Combalbert, C. Picard, J. P. Maffrand, M. Pascal and J. M. Herbert, *Thromb. Haemost.*, 2000, **84**, 891.

17 A. J. Gross and I. W. Sizer, *J. Biol. Chem.*, 1959, **234**, 1611.

18 S. A. Malencik, J. F. Sprouse, C. A. Swanson and S. R. Anderson, *Anal. Biochem.*, 1996, **242**, 202.

19 R. Amadò, R. Aeschbach and H. Neukom, *Methods Enzymol.*, 1994, **107**, 377.

20 H. Eickhoff, G. Jung and A. Rieker, *Tetrahedron*, 2001, **57**, 353.

21 I. L. Shamovsky, R. J. Rioplelle and G. M. Ross, *J. Phys. Chem.*, 2001, **105**, 1061.

22 S. Krishnan, E. Y. Chi, S. J. Wood, B. S. Kendrick, L. C, W. Gazon-Rodriguez, J. Wypych, T. W. Randolph, L. O. Narhi, A. L. Biere, M. Citron and J. F. Carpenter, *Biochemistry*, 2003, **42**, 829.

23 D. Balasubramanian and R. Kanwar, *Mol. Cell. Biochem.*, 2002, **234/235**, 27.

24 G. Helynck, C. Dubertret, D. Frechet and J. Leboul, *J. Antibiotics*, 1998, **51**, 512.

25 M. Bois-Choussey, P. Cristau and J. Zhu, *Angew. Chem. Int. Ed.*, 2003, **42**, 4238.

26 Z. Guo, K. Machiya, G. M. Salamonczyk and C. J. Sih, *J. Org. Chem.*, 1998, **63**, 4269.

27 M. Ezaki, M. Iwami, M. Yamashita, S. Hashimoto, T. Komori, K. Umehara, Y. Mine, M. Kohsaka, H. Aoki and H. Imanaka, *J. Antibiotics*, 1985, **38**, 1453.

28 A. G. Brown and P. D. Edwards, *Tetrahedron Lett.*, 1990, **31**, 6581.

29 G. L. Patrick, *An Introduction to Medicinal Chemistry*, 2nd edition, Oxford University Press, Oxford, 2001.

30 H. P. Endtz, N. van den Braak, H. A. Verbrugh and A. van Belkum, *Eur. J. Clin. Microbiol. Infect. Dis.*, 1999, **18**, 683.

31 S. Chatterjee, E. K. S. Vijayakumar, S. R. Nadkarni, M. V. Patel, J. Blumbach and B. N. Ganguli, *J. Org. Chem.*, 1994, **59**, 3480.

32 S. R. Nadkarni, M. V. Patel, Sugata Chatterjee, E. K. S. Vijayakumar, K. R. Desikan, J. Blumbach and B. N. Ganguli, *J. Antibiotics*, 1994, **47**, 334.

33 Y. Mu, M. Nodwell, J. L. Pace, J.-S. Shaw and J. K. Judice, *Bioorg. Med. Chem. Lett.*, 2004, **14**, 735.

34 H. Möbitz and M. Boll, *Biochemistry*, 2002, **41**, 1752.

35 M. E. Jung and J. C. Rohloff, *J. Org. Chem.*, 1985, **50**, 4909.

36 J. Hassan, V. Penalva, L. Lavenot, C. Gozzi and M. Lemaire, *Tetrahedron*, 1998, **54**, 13793.

37 P. D. McDonald and G. A. Hamilton, in *Mechanisms of Phenolic Oxidative Coupling Reactions*, vol. 5-B, ed. W. S. Trahanovsky, New York, 1973.

38 M. Gajhede, D. J. Schuller, A. Henriksen, A. T. Smith and T. L. Poulos, *Nature Struct. Biol.*, 1997, **4**, 1032.

39 W. Wang, S. Noël, M. Desmadril, J. Guéguen and T. Michon, *Biochem. J.*, 1999, **340**, 329.

40 G. I. Berglund, G. H. Carlsson, A. T. Smith, H. Szöke, A. Henriksen and J. Hajdu, *Nature*, 2002, **417**, 463.

41 A. Henriksen, A. T. Smith and M. Gajhede, *J. Biol. Chem.*, 1999, **274**, 35005.

42 M. Gajhede, *Biochem. Soc. Trans.*, 2001, **29**, 91.

43 Research Collaboratory for Structural Bioinformatics, Protein Data Bank, www.rcsb.org/pdb, 2004.

44 W. L. Carrick, G. L. Karapinka and G. T. Kwiatkowski, *J. Org. Chem.*, 1969, **34**, 2388.

45 S. M. Kupchan, O. P. Dhingra and C.-K. Kim, *J. Org. Chem.*, 1978, **43**, 4076.

46 G. Sartori, R. Maggi, F. Bigi and M. Grandi, *J. Org. Chem.*, 1993, **58**, 7271.

47 W. Gu, X. She, X. Pan and T.-K. Yang, *Tetrahedron: Asymmetry*, 1998, **9**, 1377.

48 G. Sartori, R. Maggi, F. Bigi, A. Arienti and G. Casnati, *Tetrahedron*, 1992, **48**, 9483.

49 K. Asakura, E. Honda and S. Osanai, *Chem. Lett.*, 1995, 583.

50 D. Ma, W. Tang, A. P. Kozikowski, N. E. Lewin and P. M. Blumberg, *J. Org. Chem.*, 1999, **64**, 6366.

51 M. E. Jung and T. L. Lazarova, *J. Org. Chem.*, 1999, **64**, 2976.

52 S. V. Jovanovich, M. G. Simic, S. Steenken and Y. Hara, *J. Chem. Soc., Perkin Trans. 2*, 1998, 2365.

53 M. B. Smith and J. March, *March's Advanced Organic Chemistry*, 5th edition, Wiley, New York, 2001.

54 M. L. Mihailovic and Z. Cekovic, *Synthesis*, 1970, 209.

55 J. Warkentin, *Synthesis*, 1970, 279.

56 J. K. Kochi, *J. Am. Chem. Soc.*, 1965, **87**, 2500.

57 G. Bringmann and S. Tasler, *Tetrahedron*, 2001, **57**, 331.

58 N. K. Bhamare, T. Granger, C. R. John and P. Yates, *Tetrahedron Lett.*, 1991, **32**, 4439.

59 D. G. Hewitt, *J. Chem. Soc. (C)*, 1971.

60 E. Subramanian, V. Lalitha and J. Bordner, *Int. J. Pept. Protein Res.*, 1984, **24**, 55.

61 R. L. Jarvest, J. M. Berge, C. S. V. Houge-Frydrych, C. Janson, L. M. Mensah, P. T. O'Hanlon, A. Pope, A. Saldanha and X. Qui, *Bioorg. Med. Chem. Lett.*, 1999, **9**, 2859.

62 J. M. Goodman, *Chemical Applications of Molecular Modelling*, The Royal Society of Chemistry, Cambridge, 1998.

63 M. Ramírez Osuna, G. Aguirre, R. Somanathan and E. Molins, *Tetrahedron: Asymmetry*, 2002, **13**, 2261.

64 R. Beugelmans, A. Bigot and J. Zhu, *Tetrahedron Lett.*, 1994, **35**, 7391.

65 D. E. Nitecki, B. Halpern and J. W. Westley, *J. Org. Chem.*, 1968, **33**, 864.

66 B. B. Snider and Z. Shi, *J. Am. Chem. Soc.*, 1992, **114**, 1790.

67 P. J. Kocienski, *Protecting Groups*, Thieme, Stuttgart, 1994.

68 M. Yamamura, T. Nakayama, H. Hashimoto, C.-G. Shin and J. Yoshimura, *J. Org. Chem.*, 1988, **53**, 6035.

69 S. Nakamura, J. Onagaki, T. Sugimoto, Y. Ura and S. Hashimoto, *Tetrahedron*, 2002, **58**, 10375.

70 A. J. Pearson and J. B. Kim, *Org. Lett.*, 2002, **4**, 2837.

71 I. A. Razak, S. S. Raj, H.-K. Fun, Z.-F. Chen, J. Zhang, R.-G. Xiong and X.-Z. You, *Acta Cryst.*, 2000, **C56**, e341.

72 M. Gdaniec, *Acta Cryst.*, 1986, **C42**, 1345.

73 T. W. Greene and P. G. M. Wuts, *Protective Groups in Organic Chemistry*, 3rd edition, Wiley-Interscience, New York, 1999.

74 P. E. Fanta, *Synthesis*, 1974, 9.

75 M. A. Rizzacasa and M. V. Sargent, *Aust. J. Chem.*, 1988, **41**, 1087.

76 T. Nelson and A. I. Meyers, *Tetrahedron Lett.*, 1994, **35**, 3259.

77 G.-Q. Lin and M. Zhong, *Tetrahedron Lett.*, 1996, **37**, 3015.

78 F. M. Hauser and P. J. F. Gauuan, *Org. Lett.*, 1999, **1**, 671.

79 D. D. Hennings, T. Iwama and V. H. Rawal, *Org. Lett.*, 1999, **1**, 1205.

80 S. Venkatraman and C.-J. Li, *Tetrahedron Lett.*, 2000, **41**, 4831.

81 S. Venkatraman and C.-J. Li, *Org. Lett.*, 1999, **1**, 1133.

82 B. C. Ranu, P. Dutta and A. Sarkar, *Tetrahedron Lett.*, 1998, **39**, 9557.

83 G.-Q. Lin and R. Hong, *J. Org. Chem.*, 2001, **66**, 2877.

84 M. Iyoda, H. Otsuka, K. Sato, N. Nisato and M. Oda, *Bull. Chem. Soc. Jpn.*, 1990, **63**, 80.

85 V. Penalva, J. Hassan, L. Lavenot, C. Gozzi and M. Lemaire, *Tetrahedron Lett.*, 1998, **39**, 2559.

86 J. Hassan, M. Sévignon, C. Gozzi, E. Schulz and M. Lemaire, *Chem. Rev.*, 2002, **102**, 1359.

87 S. Mukhopadhyay, G. Rothenberg, D. Gitis and Y. Sasson, *Org. Lett.*, 2000, **2**, 211.

88 C.-J. Li, *Acc. Chem. Res.*, 2002, **35**, 533.

89 C. Amatore and A. Jutand, *Organometallics*, 1988, **7**, 2208.

90 B. Andersh, D. L. Murphy and R. J. Olson, *Synth. Commun.*, 2000, **30**, 2091.

91 O. Mongin, C. Papamicaël, N. Hoyler and A. Gossauer. *J. Org. Chem.*, 1998, **63**, 5568.

92 W. F. Bailey and M. W. Carson, *J. Org. Chem.*, 1998, **63**, 9960.

93 H. Matsumoto, S. Inaba and R. D. Rieke, *J. Org. Chem.*, 1983, **48**, 840.

94 R. Hong, R. Hoen, J. Zhang and G.-Q. Lin, *Synlett*, 2001, 1527.

95 M. F. Semmelhack, P. Helquist, L. D. Jones, L. Keller, L. Mendelson, L. Speltz Ryono, J. Gorzynski Smith and R. D. Stauffer, *J. Am. Chem. Soc.*, 1981, **103**, 6460.

96 T. T. Tsou and J. K. Kochi, *J. Am. Chem. Soc.*, 1979, **101**, 7547.

97 M. Zembayashi, K. Tamao, J. Yoshida and M. Kumada, *Tetrahedron Lett.*, 1977, **18**, 4089.

98 L. M. Venanzi, *J. Chem. Soc.*, 1958, 719.

99 I. Yamada, N. Yamazaki, M. Yamaguchi and T. Yamagishi, *J. Mol. Catal. A: Chem.*, 1997, **120**, L13.

100 S. Yamaguchi, S. Ohno and K. Tamao, *Synlett*, 1997, 1199.

101 T. Kamikawa and T. Hayashi, *Synlett*, 1997, 163.

102 M. Kuroboshi, Y. Waki and H. Tanaka, *Synlett*, 2002, 637.

103 J. Chiarello and M. M. Joullié, *Synth. Commun.*, 1988, **18**, 2211.

104 C. Chen, Y.-F. Zhu and K. Wilcoxen, *J. Org. Chem.*, 2000, **65**, 2574.

105 B.-C. Chen, A. P. Skoumbourdis, P. Guo, M. S. Bednarz, O. R. Kocy, J. E. Sundeen and G. D. Vite, *J. Org. Chem.*, 1999, **64**, 9294.

106 S. Chandrasekhar, T. Ramachandar and M. Venkat Reddy, *Synthesis*, 2002, 1867.

107 B. Lygo, *Tetrahedron Lett.*, 1999, **40**, 1389.

108 B. Lygo and P. G. Wainwright, *Tetrahedron Lett.*, 1997, **39**, 8595.

109 F. Cavelier, M. Rolland and J. Verducci, *Org. Prep. Procedures Intl. Oppi Briefs*, 1994, **26**, 608.

110 K.-J. Fasth, G. Antoni and B. Långström, *J. Chem. Soc. Perkin Trans. 1*, 1988, 3081.

111 B. E. Love and E. G. Jones, *J. Org. Chem.*, 1999, **64**, 3755.

112 P. L. Pickard and T. L. Tolbert, *Organic Syntheses*, 1973, V, 520.

113 H. Fretz, *Tetrahedron*, 1998, **54**, 4849.

114 E. J. Corey, F. Xu and M. C. Noe, *J. Am. Chem. Soc.*, 1997, **119**, 12414.

115 N. V. Bell, W. R. Bowman, P. F. Coe, A. T. Turner and D. Whybrow, *Tetrahedron Lett.*, 1997, **38**, 2581.

116 N. V. Bell, PhD Thesis, Loughborough University, 1997 and references cited therein.

117 Y.-A. Ma, C. J. Sih and A. Harms, *J. Am. Chem. Soc.*, 1999, **121**, 8967.

118 M. Kizil and J. A. Murphy, *Tetrahedron*, 1997, **53**, 16847.

119 N. R. Curtis, J. J. Kulagowski, P. D. Leeson, I. M. Mawer, M. P. Ridgill, M. Rowley, S. Grimwood and G. R. Marshall, *Bioorg. Med. Chem. Lett.*, 1996, **6**, 1145.

120 K. Dehnicke and P. Ruschke, *Zeitschrift fuer Anorganische und Allgemeine Chimie*, 1978, **444**, 54.

121 G. S. R. Subba Rao, *Pure Appl. Chem.*, 2003, **75**, 1443.

122 P. W. Rabideau and Z. Marcinow, *Org. React.*, 1992, **42**, 1.

123 G. A. Schiehser and J. D. White, *J. Org. Chem*, 1980, **45**, 1864.

124 H. E. Zimmerman and P. A. Wang, *J. Am. Chem. Soc.*, 1993, **115**, 2205.

125 A. G. Schultz and N. J. Green, *J. Am. Chem. Soc.*, 1991, **113**, 4931.

126 T. J. Donohoe, R. R. Harji and R. P. C. Cousins, *Chem. Commun.*, 1999, 141.

127 P. W. Rabideau, *Tetrahedron*, 1989, **45**, 1579.

128 A. J. Birch, A. L. Hinde and L. Radom, *J. Am. Chem. Soc.*, 1981, **103**, 284.

129 P. W. Rabideau and D. L. Huser, *J. Org. Chem.*, 1983, **48**, 4266.

130 R. G. Harvey, *Synthesis.*, 1970, 161.

131 S. V. Ley, N. J. Anthony, A. Armstrong, M. G. Brasca, T. Clarke, D. Culshaw, C. Greck, P. Grice, A. B. Jones, B. Lygo, A. Madin, R. N. Sheppard, A. M. Z. Slawin and D. J. Williams, *Tetrahedron*, 1989, **45**, 7161.

132 H. Curtis, B. F. G. Johnson and G. R. Stephenson, *J. Chem. Soc., Dalton Trans.*, 1985, 1723.

133 A. J. Birch and G. Nadamuni, *J. Chem. Soc., Perkin Trans. 1*, 1974, 545.

134 K. Mizuno, H. Okamoto, C. Pac and H. Sakurai, *J. Chem. Soc., Chem. Commun.*, 1975, 839.

135 M. Yasuda, C. Pac and H. Sakurai, *J. Org. Chem.*, 1981, **46**, 788.

136 G. A. Epling and E. Florio, *Tetrahedron Lett.*, 1986, **27**, 1469.

137 E. Fujita, M. Shibuya, S. Nakamura, Y. Okada and T. Fujita, *J. Chem. Soc., Perkin Trans. 1*, 1974, 165.

138 Z. Lin, J. Chen and Z. Valenta, *Tetrahedron Lett.*, 1997, **38**, 3863.

139 E. Cotsaris and M. N. Paddon-Row, *J. Chem. Soc., Perkin Trans. 1*, 1984, 1487.

140 T. J. Donohoe, R. Garg and C. A. Stevenson, *Tetrahedron: Asymmetry*, 1996, **7**, 317.

141 R. E. Johnson and E. R. Bacon, *Tetrahedron Lett.*, 1994, **35**, 9327.

142 K. Soai, H. Oyamada, M. Takase and A. Ookawa, *Bull. Chem. Soc. Jpn.*, 1984, **57**, 1948.

143 A.-C. Guevel and D. J. Hart, *J. Org. Chem.*, 1996, **61**, 473.

144 H. Hara, T. Inoue, H. Nakamura, M. Endoh and O. Hoshino, *Tetrahedron Lett.*, 1992, **33**, 6491.

145 P. Wipf and Y. Kim, *Tetrahedron Lett.*, 1992, **33**, 5477.

146 A. V. Rama Rao, M. K. Gurjar and P. A. Sharma, *Tetrahedron Lett.*, 1991, **32**, 6613.

147 A. McKillop, L. McLaren, R. J. Watson, R. J. K. Taylor and N. Lewis, *Tetrahedron Lett.*, 1993, **34**, 5519.

148 S. V. Ley, A. W. Thomas and H. Finch, *J. Chem. Soc., Perkin Trans. 1*, 1999, 669.

149 G. L. Schmir, L. A. Cohen and B. Witkop, *J. Am. Chem. Soc.*, 1959, **81**, 2228.

150 A. Hutinec, A. Ziogas, M. El-Mobayed and A. Rieker, *J. Chem. Soc., Perkin Trans. 1*, 1998, 2201.

151 M. Solomon, W. C. L. Jamison, M. McCormick, D. Liotta, D. A. Cherry, J. E. Mills, R. D. Shah, J. D. Rodgers and C. A. Maryanoff, *J. Am. Chem. Soc.*, 1988, **110**, 3702.

152 A. Nishinaga, S. Kojima, T. Mashino and K. Maruyama, *Chem. Lett.*, 1994, 961.

153 B. J. Doty and G. W. Morrow, *Tetrahedron Lett.*, 1990, **31**, 6125.

154 T. Ooi and K. Maruoka, *Rev. Heteroatom Chem.*, 1998, **18**, 61.

155 J. I. Levin, *Tetrahedron Lett.*, 1996, **37**, 3079.

156 A. Yoshida, T. Hanamoto, J. Inanaga and K. Mikami, *Tetrahedron Lett.*, 1998, **39**, 1777.

157 G.-P. Wang and Z.-C. Chen, *Synth. Commun.*, 1999, **29**, 2859.

158 L. N. Mander and R. J. Hamilton, *Tetrahedron Lett.*, 1981, **22**, 4115.

159 J. M. Hook, L. N. Mander and R. Urech, *J. Org. Chem.*, 1984, **49**, 3250.

160 R. J. Hamilton, L. N. Mander and S. P. Sethi, *Tetrahedron*, 1986, **42**, 2881.

161 W. Dmowski and K. Piasecka-Maciejewska, *Tetrahedron*, 1998, **54**, 6781.

162 R. C. Helgeson, B. P. Czech, E. Chapoteau, C. R. Gebauer, A. Kumar and D. J. Cram, *J. Am. Chem. Soc.*, 1989, **111**, 6339.

163 M. E. McEntee and A. R. Pinder, *J. Chem. Soc.*, 1957, 4419.

164 M. L. Hammond, I. E. Kopha, R. A. Zambias, C. G. Caldwell, J. Boger, F. Baker, T. Bach, S. Luell and D. E. MacIntyre, *J. Med. Chem.*, 1989, **32**, 1006.

165 S. Lindman, G. Lindberg, F. Nyberg, A. Karlén and A. Hallberg, *Bioorg. Med. Chem. Lett.*, 2000, **8**, 2375.

166 D. Ma, C. Xia, J. Jiang, J. Zhang and W. Tang, *J. Org. Chem.*, 2003, **68**, 442.

167 R. Grigg, P. Kennewell and A. J. Teasdale, *Tetrahedron Lett.*, 1992, **33**, 7789.

168 R. Grigg, C. Loganathan, S. Sukirthalingam and V. Sridharan, *Tetrahedron Lett.*, 1990, **31**, 4573.

169 A. G. Schultz, *Chem. Commun.*, 1999, 1263.

170 E. J. Corey and D. S. Watt, *J. Am. Chem. Soc.*, 1973, **95**, 2303.

171 M. E. Jung and M. A. Lyster, *J. Org. Chem.*, 1977, **42**, 3761.

172 P. Allevi, P. Ciuffred, A. Longo and M. Anastasia, *Tetrahedron: Asymmetry*, 1998, **1**.

173 M. Demuynck, P. J. De Clercq and M. Vandewalle, *J. Org. Chem.*, 1979, **44**, 4863.

174 K. Mori and M. Mastsu, *Tetrahedron*, 1968, **24**, 3127.

175 J. L. Belletire, E. G. Spletzer and A. R. Pinhas, *Tetrahedron Lett.*, 1984, **25**, 5969.

176 J. L. Belletire and D. F. Fry, *J. Org. Chem.*, 1987, **52**, 2549.

177 J. L. Belletire and S. L. Fremont, *Tetrahedron Lett.*, 1986, **27**, 127.

178 P. Renaud and M. A. Fox, *J. Org. Chem.*, 1988, **53**, 3745.

179 J. Bergman, E. Koch and B. Pelcman, *J. Chem. Soc., Perkin Trans. 1*, 2000, 2609.

180 E. Juaristi and J. S. Cruz-Sánchez, *J. Org. Chem.*, 1988, **53**, 3334.

181 C. J. Parkinson, T. W. Hambley and J. T. Pinhey, *J. Chem. Soc., Perkin Trans. 1*, 1997, 1465.

182 D. J. Lee, K. Kim and Y. J. Park, *Org. Lett.*, 2002, **4**, 873.

183 M. A. Dombroski, S. A. Kates and B. B. Snider, *J. Am. Chem. Soc.*, 1990, **112**, 2759.

184 B. C. Laguzza and B. Ganem, *Tetrahedron Lett.*, 1981, **22**, 1483.

185 J. Bonjoch, J. Catena, E. Isábel, M. López-Canet and N. Valls, *Tetrahedron: Asymmetry*, 1996, **7**, 1899.

186 J. Leonard, B. Lygo and G. Procter, *Advanced Practical Organic Chemistry*, 2nd edition, Stanley Thornes, Cheltenham, 1998.

187 W. C. Still, M. Kahn and A. Mitra, *J. Org. Chem.*, 1978, **43**, 2923.

188 G. N. la Mar, W. de W. Horrocks and L. C. Allen, *J. Chem. Phys.*, 1964, **41**, 2126.

189 G. W. Anderson and A. C. McGregor, *J. Am. Chem. Soc.*, 1957, **79**, 6180.

190 J. B. West and C.-H. Wong, *J. Org. Chem.*, 1986, **51**, 2728.

191 Wuensch, *Chem. Ber.*, 1958, 543.

192 J. W. Wood and T. D. Fontaine, *J. Org. Chem.*, 1952, **17**, 891.

193 M. M. Hann, P. G. Sammes, P. D. Kennewell and J. B. Taylor, *J. Chem. Soc. Perkin Trans. 1*, 1982, 307.

194 V. Dourtoglou, B. Gross, V. Lambropoulou and C. Zioudrou, *Synthesis*, 1984, 572.

195 K. E. Koenig, G. M. Lein, P. Stuckler, T. Kaneda and D. J. Cram, *J. Am. Chem. Soc.*, 1979, **101**, 3553.

196 B. Rzeszotarska, B. Nadolska and J. Tarnawski, *Liebigs Ann. Chem.*, 1981, 1294.

197 M. J. O'Donnell and R. L. Polt, *J. Org. Chem.*, 1982, **47**, 2663.

198 Y. Wu and D. H. Busch, *J. Am. Chem. Soc.*, 1970, **92**, 3326.

199 C. J. Thoman and I. M. Hunsberger, *J. Org. Chem.*, 1968, **33**, 2852.

200 B. Y. Dakova, M. J. Evers, L. R. Christiaens and M. R. Guillaume, *Bull. Soc. Chim. Belg.*, 1987, **96**, 219.

201 N. V. Bell, W. R. Bowman, P. F. Coe, A. T. Turner and D. Whybrow, *Can. J. Chem.*, 1997, **75**, 873.

202 S. Lemaire, D. Yamashiro, C. Behrens and C. H. Li, *J. Am. Chem. Soc.*, 1977, **199**, 1577.

203 M. Schlosser, P. Maccaroni and E. Marzi, *Tetrahedron*, 1998, **54**, 2763.

204 MacroModel v. 8.0, Schrödinger, 2002.

205 F. Mahamadi, N. G. J. Richards, W. C. Guida, R. Liskamp, M. Lipton, C. Caufield, G. Chang, T. Hendrickson and W. C. Still, *J. Comput. Chem.*, 1990, **11**, 440.

206 N. L. Allinger, *J. Am. Chem. Soc.*, 1977, **99**, 8127.