The Total Synthesis of *Pseudomonocardia* sp. Quinolone Natural Products

*and*

Studies Towards the Total Synthesis of 1β-Hydroxyalantolactone

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Supervised by Prof. David R. Spring

This dissertation is submitted for the degree of **Doctor of Philosophy**
Declaration

This dissertation is submitted in fulfilment of the requirements for the degree of Doctor of Philosophy. It describes work carried out in the Department of Chemistry, University of Cambridge, between October 2014 and March 2018 under the supervision of Prof. David Spring. Unless otherwise indicated, the research described is my own and not the product of collaboration. The work presented in this dissertation has not been submitted for any other degree. It does not exceed the prescribed word limit for the Physics and Chemistry Degree Committee.

Signed: Date:

Stephen Michael Geddis
Corpus Christi College
Abstract

Natural products have long been known for their broad range of useful therapeutic properties, and have been widely utilised in the field of medicine. This dissertation describes work towards the total synthesis of natural products possessing biological activity in two important areas.

The first section concerns the total synthesis of six 4-quinolone natural products, four of which had never been synthesised before. These compounds were originally isolated from a soil bacterium of the genus *Pseudonocardia*, and bear intriguing structural resemblance to the *Pseudomonas* Quinolone Signal. This signalling molecule is vital to the quorum sensing activity of the human pathogen *Pseudomonas aeruginosa*, which is a phenomenon by which it regulates many of its virulence factors. These natural products possess the potential to disrupt this system, hence attenuating the pathogenicity of the bacteria. The routes that were developed are highly divergent, efficiently giving access to multiple natural products from mutual late stage intermediates. Key steps included regioselective epoxidation, palladium-catalysed heterocylisation and acid catalysed 1,3-transposition of an allylic alcohol.

In the second section, attention turns towards the total synthesis of the complex sesquiterpene lactone 1β-Hydroxyalantolactone. The compound possesses five stereogenic centres, one of which is quaternary, alongside a challenging tricyclic core scaffold. Previous biological studies have revealed a range of intriguing properties, including anti-inflammatory and anti-tumour activity. The chosen route utilises as its key step the catalytic desymmetrisation of a diene which was itself accessed by Birch reduction chemistry. Whilst the synthesis is as yet incomplete, access was granted to a key intermediate encompassing around half of the stereocentres present in the natural product.
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Thanks to Janine, for keeping me going when things have been tough, for all of our adventures, and for teaching me the true meaning of patience by means of the X5 bus. Finally, thanks to my parents, Barbara and Michael, and my brother James, who have always been there for me. Through their constant love and support, all of this was made possible.
Abbreviations

AD-mix-β (Sharpless) Asymmetric dihydroxylation mix-β
br Broad
BRSM Based on recovered starting material
Bu Butyl
CBS Corey-Bakshi-Shibata
CF Cystic fibrosis
CHP Cumene hydroperoxide
COD 1,5-Cyclooctadiene
COS Costunolide synthase
Cp Cyclopentadiene
d Doublet
d.r. Diastereomeric ratio
dba Dibenzylideneacetone
DCC N,N'-Dicyclohexycarbodiimide
DCE 1,2-Dichloroethene
DIBAL Diisobutylaluminum hydride
DIPEA N,N-Diisopropylethylamine
DMAD Dimethyl acetylenedicarboxylate
DMAP 4-Dimethylaminopyridine
DMAPP Dimethylallyl disphosphate
DMEDA N,N'-Dimethylethlenediamine
DMF N,N-Dimethylformamide
DMP Dess–Martin periodinane
DMPU N,N'-Dimethylpropylene urea
DMSO Dimethyl sulfoxide
DNA Deoxyribonucleic acid
dppf 1,1'-Bis(diphenylphosphino)ferrocene
e.e. Enantiomeric excess
E1cB Elimination unimolecular conjugate base
en Ethylenediamine
FCC Flash column chromatography
FGI Functional group interconversion
FPP Farnesyl diphosphate
GAA (+)-Germacrene A acid
GAO Germacrene A oxidase
GAS Germacrene A synthase
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Full Form</th>
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<tbody>
<tr>
<td>GPP</td>
<td>Geranyl diphosphate</td>
</tr>
<tr>
<td>h</td>
<td>Hours</td>
</tr>
<tr>
<td>HHQ</td>
<td>2-heptyl-4(1H)-quinolone</td>
</tr>
<tr>
<td>HMBC</td>
<td>Heteronuclear multiple bond correlation</td>
</tr>
<tr>
<td>HPLC</td>
<td>High-performance liquid chromatography</td>
</tr>
<tr>
<td>HSQC</td>
<td>Heteronuclear single-quantum correlation</td>
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<td>HWE</td>
<td>Horner-Wadsworth-Emmons</td>
</tr>
<tr>
<td>i</td>
<td>iso</td>
</tr>
<tr>
<td>IC₅₀</td>
<td>Half maximal inhibitory concentration</td>
</tr>
<tr>
<td>IJ-5</td>
<td>1β-Hydroxyalantolactone</td>
</tr>
<tr>
<td>IpC</td>
<td>iso-Pinocamphyl</td>
</tr>
<tr>
<td>IPP</td>
<td>Isopentenyl diphosphate</td>
</tr>
<tr>
<td>IR</td>
<td>Infra-red</td>
</tr>
<tr>
<td>JapA</td>
<td>Japonicone A</td>
</tr>
<tr>
<td>Kₒ</td>
<td>Dissociation constant</td>
</tr>
<tr>
<td>LCMS</td>
<td>Liquid chromatography-mass spectrometry</td>
</tr>
<tr>
<td>LPS</td>
<td>Lipopolysaccharides</td>
</tr>
<tr>
<td>m</td>
<td>meta</td>
</tr>
<tr>
<td>M</td>
<td>Molar</td>
</tr>
<tr>
<td>m</td>
<td>Multiplet/medium</td>
</tr>
<tr>
<td>m.p.</td>
<td>Melting point</td>
</tr>
<tr>
<td>m-CPBA</td>
<td>meta-Chloroperoxybenzoic acid</td>
</tr>
<tr>
<td>MDM2</td>
<td>Mouse double minute 2 homolog</td>
</tr>
<tr>
<td>Me</td>
<td>Methyl</td>
</tr>
<tr>
<td>MIC</td>
<td>Minimum inhibitory concentration</td>
</tr>
<tr>
<td>min</td>
<td>Minutes</td>
</tr>
<tr>
<td>mol. sieves</td>
<td>Molecular sieves</td>
</tr>
<tr>
<td>MOM</td>
<td>Methoxymethyl</td>
</tr>
<tr>
<td>mRNA</td>
<td>Messenger ribonucleic acid</td>
</tr>
<tr>
<td>MRSA</td>
<td>Methicillin-resistant <em>Staphylococcus aureus</em></td>
</tr>
<tr>
<td>MTMCl</td>
<td>Chloromethyl methyl sulfide</td>
</tr>
<tr>
<td>MvfR</td>
<td>Multiple virulence factor regulator</td>
</tr>
<tr>
<td>N</td>
<td>Normality</td>
</tr>
<tr>
<td>NBS</td>
<td>N-Bromosuccinimide</td>
</tr>
<tr>
<td>NF-κB</td>
<td>Nuclear factor κB</td>
</tr>
<tr>
<td>NMO</td>
<td>N-Methylmorpholine N-oxide</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear magnetic resonance</td>
</tr>
<tr>
<td>NO</td>
<td>Nitric oxide</td>
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</table>
NOESY  Nuclear Overhauser effect spectroscopy
NR     No reaction
Nu     Nucleophile
o      ortho
OD_{XXX} Optical density at wavelength XXX nm
p      para
p53    Tumor protein p53
PADA   Potassium azodicarboxylic acid
PCC    Pyridinium chlorochromate
PDC    Pyridinium dichromate
Pin    Pinacol
PMB    para-Methoxybenzyl
PPA    Polyphosphoric acid
ppm    Parts per million
PPTS   Pyridinium p-toluenesulfonate
PQS    *Pseudomonas* quinolone signal (2-heptyl-3-hydroxy-4(1H)-quinolone)
PqsR   *Pseudomonas* quinolone signal receptor
Pr     Propyl
PTSA   p-Toluenesulfonic acid
Pv     Pivalate
py     Pyridine
q      Quartet
QS     Quorum-sensing
quant. Quantitative
r.t.   Room temperature
Rf     Retention factor
Rhl    Rhamnolipid
s      Singlet/Strong
SAD    Sharpless asymmetric dihydroxylation
SAR    Structure-activity relationship
S_{N1} Substitution nucleophilic unimolecular
S_{N2} Substitution nucleophilic bimolecular
sp.    Species
STL    Sesquiterpene lactone
t      tert
t      Triplet
TBA    Tetra-n-butylammonium
<table>
<thead>
<tr>
<th>Acronym</th>
<th>Definition</th>
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</thead>
<tbody>
<tr>
<td>TBDMS</td>
<td><em>tert</em>-Butyldimethylsilyl</td>
</tr>
<tr>
<td>TBDPS</td>
<td><em>tert</em>-Butyldiphenylsilyl</td>
</tr>
<tr>
<td>TEA</td>
<td>Triethylamine</td>
</tr>
<tr>
<td>Tf</td>
<td>Triflate</td>
</tr>
<tr>
<td>TFA</td>
<td>Trifluoroacetic acid</td>
</tr>
<tr>
<td>THF</td>
<td>Tetrahydrofuran</td>
</tr>
<tr>
<td>TIPS</td>
<td>Triisopropylsilyl</td>
</tr>
<tr>
<td>TLC</td>
<td>Thin-layer chromatography</td>
</tr>
<tr>
<td>TMS</td>
<td>Trimethylsilyl</td>
</tr>
<tr>
<td>TNFα</td>
<td>Tumour necrosis factor α</td>
</tr>
<tr>
<td>TPAP</td>
<td>Tetrapropylammonium perruthenate</td>
</tr>
<tr>
<td>ℓ₉</td>
<td>Retention time</td>
</tr>
<tr>
<td>UV</td>
<td>Ultra-violet</td>
</tr>
<tr>
<td>w</td>
<td>Weak</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organisation</td>
</tr>
<tr>
<td>WT</td>
<td>Wild-type</td>
</tr>
<tr>
<td>δ</td>
<td>Chemical shift</td>
</tr>
<tr>
<td>ν</td>
<td>Wavenumber</td>
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The Total Synthesis of Natural Products

Since its advent in 1828 with Wöhler’s rational synthesis of urea, the chemical synthesis of natural products has acted as a constant source of fascination to the scientific community, and of considerable benefit to society as a whole. The pursuit of the efficient assembly of complex architectures from simple building blocks has led to many elegant solutions which clearly demonstrate the creativity demanded of those working in this field.

Upon the conclusion of a successful synthetic campaign, the organic chemist is rewarded with compounds which often possess very powerful biological activity. Indeed, many natural products have seen widespread utilisation in medicine; some representative examples are shown in Figure 1. Galantamine, obtained from the bulbs and flowers of Galanthus caucasicus has been shown to increase the concentration of acetylcholine in certain parts of the brain, and has been used in the treatment of Alzheimer’s Disease. Meanwhile, morphine, isolated from the opium poppy Papaver somniferum, is used to treat acute pain by interacting with the µ-δ receptors of the brain. Finally, Taxol, initially isolated from the bark of the Pacific Yew tree Taxus brevifolia is used in the treatment of several forms of cancer through modulation of microtubule dynamics.

![Figure 1: Examples of natural products which have seen widespread use in medicine.](image-url)

This biological richness of natural products is also evident upon consideration of approved drugs in recent years; for example 47% of the cancer drugs approved in the 25 years preceding 2007 were natural products or were directly derived therefrom. However, these molecules are often obtainable only in miniscule amounts from natural sources (such as extraction from essential plant oils or bacterial fermentation broths). This necessitates their chemical synthesis to access the quantities required for biological screening, clinical trials and eventual large-scale industrial drug manufacture.

Despite their high levels of biological activity, natural products often fall foul of limitations which render them unsuitable for use as drugs. They may possess unsuitable pharmacokinetic properties, insufficient stability under physiological conditions, or poor
selectivity for their desired target. Fortunately, the tools of organic chemistry can once again demonstrate their utility in facing these challenges by allowing access to optimised analogues of the natural compounds. For example, epothilone B 1 (Scheme 1), first isolated from Sorangium cellulosum, possesses potent anti-cancer activity by virtue of its ability to stabilise microtubules. However, its macrolactone ring was observed to undergo rapid esterase mediated hydrolysis, precluding in vivo application. Through switching this to a macrolactam, ixabepilone 2 was discovered, which possesses much improved metabolic stability and is now approved for the treatment of chemotherapy-resistant metastatic breast cancers.

Scheme 1: Replacement of the macrolactone in epothilone B with the macrolactam in ixabepilone results in much improved metabolic stability.

An additional output of total synthesis is the new methodology which must be developed to contend with the challenges associated with novel structural motifs present in the target compound. For example, Sheehan’s 1955 synthesis of penicillin V required the formation of a β-lactam ring as the final step. Whilst the high strain of this four-membered ring confers penicillin with its potent antimicrobial properties, this also makes its formation very challenging. Sheehan solved this problem by implementation of his methodology for amide coupling using N,N'-dicyclohexylcarbodiimide (DCC) to form the final four-membered lactam ring (Scheme 2). The use of DCC and related carbodiimide compounds is now a staple in coupling chemistry.

Scheme 2: Sheehan’s DCC coupling in the final step of his penicillin V synthesis.

The merits of total synthesis are clear. This dissertation describes the development of synthetic routes towards natural products possessing biological activity in two important areas. The first section is concerned with six 4-quinolone natural products, which were first isolated from a soil bacterium of the genus Pseudonocardia. These compounds possess interesting antimicrobial properties, and bear intriguing resemblance to an important
signalling molecule used by the pathogen *Pseudomonas aeruginosa*. In the second section, attention turns to the attempted development of a route towards the sesquiterpene lactone 1β-hydroxyalantolactone (also known as IJ-5), found in the aerial part of *Inula japonica*. This compound possesses anti-inflammatory and antitumour properties.
Section I

The Total Synthesis of *Pseudonocardia* sp. Quinolone Natural Products
1.0 Introduction

1.1 Natural Product Antibiotics

One area in which natural products have had a particularly profound impact is that of antibiotic discovery. An antibiotic is an agent which inhibits bacterial growth or kills bacteria. Their development was arguably one of the greatest advances in 20th century medicine. Many of the medical interventions which society relies upon to maintain the quality of life to which we have become accustomed require robust control of infection, such as chemotherapy, cardiac surgery and organ transplantation.

Natural products have played a key role throughout the history of antibiotics, repeatedly demonstrating themselves to be well suited for this application. Since Fleming’s discovery of penicillin in a culture of *Penicillium rubens*, molecules produced by microbes have been a major source of new classes of antibiotics. A selection of examples of core structures are shown in Figure 2. The penicillins (and other β-lactams) acylate the enzyme responsible for the formation of cross-links in the cell walls of bacterial cells, hence inhibiting cell wall growth. Aminoglycosides bind to the bacterial ribosome, leading to incorrect mRNA translation, causing the production of faulty proteins. Similarly, most tetracyclines inhibit protein elongation through binding to the ribosome, although some types act by disruption of the cell membrane. Macrolides also inhibit protein synthesis, in this case by causing premature dissociation of the growing protein from the ribosome.

![Core structures of representative classes of natural product antibiotics.](image)

Figure 2: Core structures of representative classes of natural product antibiotics.

It has been estimated that around 16,500 antibiotic natural products were known by 2002. These molecules are the product of millennia of evolutionary selection for evasion of the
mechanisms of defence developed by bacteria (of which synthetic antibiotic candidates often fall foul). However, this wealth of potential antibiotic drugs has been underutilised. This is due to a number of difficulties, not least of which are the low supply of the natural products, issues with their purification and their often poor pharmacological profiles. This once again highlights the need for the tools of organic synthesis, both in supplying enough of the natural product for biological screening, and in the production of analogues of the molecules themselves with superior pharmacokinetics.

This need for the development of new antibiotics is growing ever more urgent with the emergence of antibacterial resistance. The use of antibiotics creates an evolutionary pressure on bacteria, selecting for strains which are resistant to the drug. Recent years have seen the alarming appearance of methicillin-resistant Staphylococci aureus (MRSA), Pneumococci resistant to both penicillin and macrolides, vancomycin-resistant Enterococci and multidrug-resistant strains of Mycobacterium tuberculosis. It is estimated that in the US, around 2,000,000 are infected with resistant pathogens yearly, resulting in 23,000 deaths. Furthermore, resistance has been quick to emerge for many of the more recently developed antibiotic agents.

Bacteria may become resistant to antibiotics via a number of mechanisms. They may develop efflux systems to remove the drug from the cell, preventing it from reaching its target. This target may also become altered in resistant strains in such a way that the drug can no longer act on it. More directly, the bacteria may synthesise enzymes which destroy the antibiotic, or develop new metabolic pathways which bypass the action of the drug.

We risk entering a new era in which we will be unable to treat the most basic of infections. The only way to avoid this is by the development of novel antibacterial classes utilising new targets. Natural products are a prime source of candidates, as demonstrated by the recent discovery of teixobactin (Figure 3). Teixobactin was extracted from previously uncultured soil bacteria using newly developed methods, and is thought to act by binding to lipids in the cell wall.
1.2 4-quinolones

The 4-quinolones (Figure 4), both natural and synthetic, are molecules which display a plethora of biological activity, including roles in quorum sensing systems, iron chelation, anticancer properties, as well antibiotic behaviour. The relevance of these properties to antibiotic activity will be examined in this section, demonstrating the viability of 4-quinolone natural products as antibiotic agents.

1.2.1 Quorum Sensing Activity

Quorum sensing (QS) refers to a mechanism by which many bacteria are able to modulate their gene expression in a population density dependant manner, using small signalling molecules known as autoinducers. Each bacterium synthesises and releases the autoinducer into its environment. The extracellular concentration of the autoinducer rises with increasing population density, and once this concentration reaches a threshold value (and the population has become “quorate”), the autoinducer is able to bind to its receptor within the cell (Figure 5). The resultant complex then binds to its promoter region in DNA, leading to expression of the genes under regulation. This includes those responsible for the generation of the signalling molecules, leading to a positive-feedback loop which is proposed to ensure simultaneous activation across the entire population. This feedback phenomenon is also what gives rise to the term "autoinducer".
A broad range of bacterial phenotypes have been reported to operate under QS regulation. Much early work in the field concerned the marine bacterium *Vibrio fischeri*, which uses this system to control its bioluminescence.\textsuperscript{38-40} It has since been discovered that a variety of human pathogens use QS to regulate production of many of their virulence factors, including *Staphylococcus aureus, Pseudomonas aeruginosa, Vibrio cholerae* and *Bacillus cereus*.\textsuperscript{37} By only expressing these factors at high population densities, there is an increased probability of overwhelming the host’s defences.\textsuperscript{41} This implies an intriguing therapeutic application: by disrupting any part of the QS scheme, it could be possible to prevent the bacteria from carrying out this harmful behaviour.\textsuperscript{42} Indeed, mutants which are unable to carry out QS have been reported to display attenuated virulence.\textsuperscript{43,44} Critically, such an approach would not put the bacteria under a “life-or-death” selection, and it has been proposed that this should prevent the emergence of resistance mechanisms.\textsuperscript{31,45} Whilst recent research has cast doubt on this hypothesis,\textsuperscript{46} it is thought that the development of resistance will be slow, which would still represent a marked advantage when compared to the rapid resistance which has emerged for recently developed traditional antibiotics.\textsuperscript{31}

The World Health Organisation (WHO) recently designated the development of new antibiotics targeting *P. aeruginosa* as a critical priority.\textsuperscript{47} This opportunistic, Gram-negative bacterium has a high capacity for survival in a range of conditions, and is responsible for 9% of all hospital-acquired infections in the US.\textsuperscript{48} It poses a particular risk to those suffering from cystic fibrosis (CF), and is the major pathogen in infections of the CF lung, where it forms highly persistent biofilms.\textsuperscript{49,50}

*P. aeruginosa* is known to employ a complex hierarchal QS network.\textsuperscript{51} Among the autoinducers used is the *Pseudomonas* quinolone signal (PQS, 9), which binds to the transcriptional regulator PqsR (also known as MvfR), leading to the expression of virulence factors such as elastase, the toxins pyocyanin and hydrogen cyanide, and the adhesive protein lectin (Figure 6).\textsuperscript{52-55} PQS’s immediate precursor, 2-heptyl-4(1H)-quinolone (HHQ,
10), has also been shown to play a signalling role by binding to PqsR. Furthermore, recent studies suggest that these molecules may be able to bind to additional protein targets, including MexG (an efflux pump), PhzD1 (involved in phenazine biosynthesis), and RhlR (another QS regulator). These findings further underline the importance of 4-quinolones in *P. aeruginosa*.

![Figure 6](image)

**Figure 6**: 4-quinolone autoinducers are responsible for the QS regulation of multiple virulence factors in *P. aeruginosa*.

A range of small molecules have been shown to antagonise the activity of PQS, a selection of which are shown in Figure 7. By introducing a nitro group to the 6-position of HHQ, a compound 11 was discovered, which could inhibit the stimulation of PqsR by HHQ itself. Other electron-withdrawing groups in the same position gave similar results. However, it was noticed that *P. aeruginosa* was carrying out a 3-hydroxylation of 11 in a manner analogous to the conversion of HHQ to PQS to give a product which exhibited agonistic activity. Through blocking this position with a carboxamide moiety, a compound 12 was discovered which maintained its antagonism. Inhibitors which deviate slightly from the 4-quinolone core have also been reported, for example 4-N-alkylaminoquinoline 13 and quinazoline 14. Many natural products have also been reported to disrupt QS systems, including farnesol 15. This compound causes reduced PqsR promoted transcription in *P. aeruginosa*, leading to attenuated production of PQS and the toxin pyocyanin.

![Figure 7](image)

**Figure 7**: Selected small-molecule inhibitors of PQS mediated signalling.

Pyocyanin (16, Figure 8) is capable of disrupting many important biochemical processes. This leads to a numerous deleterious effects on human cells, including inhibited respiration and ciliary action. These effects allow pyocyanin to play a critical role in infection; indeed,
mutant *P. aeruginosa* strains which were unable to produce the toxin were unsuccessful in infecting the lungs of mice.\(^6^7\) Being able to prevent the production of pyocyanin could therefore be of great therapeutic benefit, and it may be that nature has already employed this strategy; the fungus *Candida albicans* is able to generate farnesol at sufficient concentration to lower *P. aeruginosa*’s production of the toxin, thus affording the fungus protection from attack.\(^6^4\)

![Figure 8: The structure of pyocyanin, and important toxin produced by *P. aeruginosa*.](image)

### 1.2.2 Synthetic quinolone antibiotics

The synthetic quinolone antibiotics are a key example of the antibiotic activity which 4-quinolones may possess. Characterised by the presence of a carboxylic acid moiety in the 3-position (Figure 9), they have been used to treat more than 800 million patients worldwide.\(^6^8\) Their clinical indications include infections of the upper and lower respiratory system, urinary tract, gastrointestinal tract, skin, bone and soft tissues, as well as prostatitis, and gynaecologic and sexually transmitted infections.\(^6^9\) These antibiotics act by inhibiting DNA gyrase and topoisomerase IV, which are enzymes essential for the relaxation of strain introduced to DNA during the replication process.\(^7^0\) The antibiotics hence result in chromosome fragmentation and cell death. However, their widespread use has led to the emergence of resistance, with its incidence doubling during the 1990s.\(^7^1\)

![Figure 9: Examples of synthetic quinolone antibiotics.](image)

### 1.2.3 Antibiotic 4-quinolone natural products

As well as utilising the 4-quinolone scaffold in the form of PQS in its quorum sensing system, *P. aeruginosa* also employs a number of 4-quinolone natural products as antibiotic agents active against other microbes. Named the Pyo compounds, they are strongly active against Gram-positive bacteria; the most potent being Pyo II, which was later found to be a mixture of 2-alkyl-4-hydroxyquinoline N-oxides (tautomeric with 4-quinolone) (Figure 10).\(^7^2,7^3\) This
activity may explain how \textit{P. aeruginosa} is able to outcompete \textit{Staphylococcus aureus} in cystic fibrosis lung infections.\textsuperscript{74} The compounds are believed to act via inhibition of the electron transport chain through the bc\textsubscript{1} segment; however, their high toxicity to mitochondrial respiration has prevented their use as therapeutic antibiotics.\textsuperscript{75,76}

\begin{center}
\includegraphics[width=0.5\textwidth]{antibiotic4-hydroxyquinolinesN-oxides.png}
\end{center}

\textbf{Figure 10:} Antibiotic 4-hydroxyquinolines N-oxides produced by \textit{P. Aeruginosa}. \(R = C_7H_{15}, C_9H_{19}, C_{11}H_{23}\).

A group of naturally occurring 4-quinolones have been isolated from the Chinese herbal remedy \textit{Evodia ruteacarpa} \textsuperscript{19-24, Figure 11} and were shown to be active against \textit{Helicobacter pylori}, which is responsible for the pathogenesis of chronic gastritis, peptic ulcers and gastric cancers.\textsuperscript{77,78} The molecules were shown to be selective, leaving other intestinal microbes unaffected, suggesting they could be used to eradicate \textit{H. pylori} whilst maintaining other healthy intestinal flora.

\begin{center}
\includegraphics[width=1\textwidth]{4-quinolone-compounds.png}
\end{center}

\textbf{Figure 11:} 4-quinolone compounds 19-24 extracted from \textit{Evodia ruteacarpa} with activity against \textit{Helicobacter pylori}.

\subsection*{1.2.4 \textit{Pseudonocardia} sp. CL38489 Natural Products}

Another group of 4-quinolones active against \textit{H. pylori} have been isolated from eight litres of the fermentation broth of the actinomycete \textit{Pseudonocardia} sp. CL38489, which itself was isolated from a soil sample from India. \textsuperscript{25-32, Figure 12}.\textsuperscript{16} Whilst the members of the set containing stereocentres were shown to be optically active, the absolute stereochemistry was
not assigned. The most active of these is the epoxide 32, which has a bactericidal minimum inhibitory concentration (MIC) of 10 ng/mL and a bacteriostatic MIC of 0.1 ng/mL. Meanwhile, no activity against *Bacillus stearothermophilus, Micrococcus luteus, Staphylococcus aureus* or *Pasteurella haemolytica* was found, implying a degree of selectivity. Incidentally, other antibiotic natural products have been isolated from alternative members of the *Pseudonocardia* genus which are associated with fungus-growing ants, where it is presumed they act symbiotically. Additionally, 25-32 bear intriguing similarity to the menaquinones, which are involved in the electron transport cycles of many bacteria.

![Chemical structures](image)

**Figure 12**: 4-quinolones 25-32 extracted from eight litres of the fermentation broth of *Pseudonocardia* sp. with activity against *H. pylori*, along with the mass of each which was obtained.

However, these molecules had yet to be screened against *P. aeruginosa*, which could bear interesting results, as their structural similarity to PQS suggests the possibility of modulation of quorum sensing processes. The barrier to such an investigation is the miniscule amounts in which they are available from natural sources (Figure 12), and so they represent an attractive target for total synthesis. Compound 25 had already been utilised as an intermediate in the production of the anti-tumour intervenolin 36 (Scheme 3); however the
other compounds were yet to be synthesised. With this in mind, the Spring Group embarked on the total synthesis of 25-32, with a particular interest in their potential quorum sensing activity. If shown to be active against P. aeruginosa, the design and construction of analogues to these would then further knowledge of the structure-activity relationship (SAR). Additionally, these analogues could then be used to further elucidate the antimicrobial properties of the compounds.

![Scheme 3: Synthesis of Intervenolin 36, utilising natural product 25 as a key intermediate.](image)

**1.3 Previous Routes Employed by the Spring Group**

The group’s synthesis of Natural Products 25-28 was carried out by Flavia Salvaggio and James Hodgkinson. They employed a disconnection by means of an sp\(^3\)-sp\(^2\) Suzuki reaction (Scheme 4). Suzuki reactions involving an sp\(^3\) partner are often challenging due to the formation of side products by means of β-H elimination from the alkyl-palladium intermediate; however the substrate used in this route lacks β-hydrogens, so this issue is avoided.

![Scheme 4: Disconnection of natural products 25-28 using an sp\(^3\)-sp\(^2\) Suzuki Coupling. X = halide or pseudo-halide](image)

The synthesis of the boronate ester coupling partner 38 utilised Negishi’s zirconium catalysed carboalumination on alkyne 40 as its key step. This was followed by Miyaura borylation to give 38 in modest yield. (Scheme 5)
With this in hand, it remained to synthesise the quinolone coupling partners. Formation of the partner required for 25 was achieved by a Conrad-Limpach reaction, a widely used method for synthesising the 4-quinolone scaffold, to give 42. Subsequent reduction with LiAlH₄ and halogenation gave the bromo- and chloro- (by Appel reaction and reaction with thionyl chloride respectively) coupling partners (44 & 45, Scheme 6).

The formation of the coupling partners required for the synthesis of 26 and 28 followed an analogous approach. However, the lack of a substituent at the 3-position allowed direct condensation of anilines 46 & 47 with dimethylacetylene dicarboxylate (DMAD) to afford the substrate for the Conrad-Limpach condensation. Cyclisation, reduction (using NaBH₄ in this case, with the relatively electron-deficient quinolone substituent perhaps increasing the electrophilicity of the normally unreactive ester group) followed by halogenation yielded the desired species 54-57 (Scheme 7).
Scheme 7: Synthesis of the coupling partners necessary for Suzuki coupling to give 26 & 28.

Synthesis of the coupling partner for 27 was attempted using the methodology developed for 25, 26 & 28. However, the Conrad-Limpach cyclisation step proceeded with very poor yield, so an alternative strategy was developed. Alcohol 43 (utilised in the synthesis of 25) served as the starting point, which was sequentially protected, methylated, deprotected and chlorinated to give the desired chloro compound 61 (Scheme 8).

Scheme 8: Alternative route developed for the synthesis of the coupling partner for 27.

These coupling partners (of general form 37) were then used in a Suzuki coupling with boronate ester 38 to give the natural products 25-28 in moderate yields (Scheme 9). The chloride compounds generally resulted in higher yields.
With natural products 25-28 in hand, their potential to modulate PqsR mediated QS was assessed using the heterologous E. coli system first used by Cugini et al. Unfortunately no agonistic or antagonistic transcriptional effects could be detected (data not shown). Next, the effect of the compounds on the growth of several bacterial species was investigated (Figure 13). No effect on the growth of P. aeruginosa was observed; however, the compounds appeared to induce an extended lag phase in the growth of S. aureus and E. coli (with the exception of 27 which was inactive in E. coli). The structurally related PQS has been reported to induce similar effects in a range of Gram-negative and Gram-positive species. Given the compounds’ similarity to menaquinone (vide supra), this may perhaps be due to interference with the bacteria’s electron transport chains.
Figure 13: The growth of bacterial species in the presence of 200 μM of natural products 25-28, alongside the results of DMSO negative control and gentamicin positive control experiments. Error bars represent standard deviation of three experiments. Figure adapted from Salvaggio et al.82

Synthesis of the remainder of the natural products was first attempted by Laura Carro (Spring Group). Carro’s attempts to synthesise 29 focused on addition of a Grignard reagent 63 (derived from iodo-compound 41, used in the synthesis of the boronate ester from the previous routes) to a suitable aldehyde (readily oxidised using Dess-Martin Periodinane from alcohol 53, used in the synthesis of 28).93 However, this was unsuccessful, which was attributed to difficulties in producing iodo-compound 41 in high purity, which stemmed from its volatility.

Scheme 10: Unsuccessful strategy to synthesise 29 utilising addition of a Grignard reagent 63 to aldehyde 62.
A Horner-Wadsworth-Emmons approach was attempted towards 30 (Scheme 11). The route coupled aldehyde 62 from the attempted synthesis of 29 (vide supra) with phosphonate 66, synthesised by Arbuzov reaction with α-bromo species 65 derived from commercially available 6-methyl-5-hepten-2-one 64. This was followed by addition of methylmagnesium bromide to give 30 (although with insufficient purity for full characterisation). However, the route suffered from poor yields in both the HWE and addition steps, and the intermediates used to give phosphonate 66 rapidly decomposed, leading to irreproducible yields. This route was therefore in need of optimisation or replacement.

Scheme 11: Attempted HWE synthesis of 30.

1.4 Project Outline

The work in this section describes the total synthesis of the remaining natural products 29-32. Given the structural similarities between the compounds, it was hoped to design a more divergent strategy, whereby multiple natural products could be derived from mutual late-stage intermediates. In particular, the allylic alcohols 29 and 30 are regioisomeric, and so perhaps could be interconverted using some manner of rearrangement chemistry. In this way, two natural products could be accessed using only one synthetic scheme. Similarly, 27, 31 & 32 all contain the core structure encompassed in natural product 25. This compound could perhaps be decorated to give the remaining targets 31 and 32 directly, alongside offering an improved synthesis of 27, which required a lengthy seven-step synthesis for its quinolone-chloride coupling partner (61, vide supra). Following successful conclusion of this strategy, further biological studies could be carried out, and analogues of the natural products constructed. This would facilitate investigation of the structure-activity relationship of any activity identified.
Scheme 12: Proposed divergent strategy for the synthesis of 29-32.
2.0 Results and Discussion

2.1 Total Synthesis of 30 and Attempted Rearrangement to 29

In order to validate the proposed divergent synthesis strategy, the first target was the allylic alcohol natural product 30 (Scheme 13). It was hoped that once this was accessed, chemistry could be found which would induce a rearrangement to the regioisomeric natural product 29, thus efficiently giving access to two of the target compounds from only one synthetic scheme. This section summarises a number of approaches that were attempted for the synthesis of 30, and, following the development of an efficient route, the attempted conversion to 29.

![Scheme 13: Divergent strategy for accessing 29 by rearrangement of its regioisomer 30.](image)

2.1.1 Approach 1: Aldol Condensation

The first strategy attempted for the synthesis of 30 is outlined in Scheme 14. It was envisaged that compound 67 could be constructed by the means of an aldol condensation between aldehyde 62 and commercially available ketone 64. Addition of a methyl group to the enone carbonyl would then yield target compound 30. Choice of a hard methyl anion equivalent should disfavour conjugate addition to the enone, and attack at the quinolone carbonyl would result in disruption of the extended conjugated system, and so it was anticipated that this reaction would proceed with acceptable selectivity. Preliminary studies utilising inorganic bases (such as NaOH) to drive the aldol condensation to form 67 proved unsuccessful (work carried out by L. Carro, Spring Group, data not shown). Attention then turned towards the application of the aldol protocol developed by Wang et al., who utilised a catalytic amount of proline and triethylamine (TEA) to couple various aromatic aldehydes with ketones under very mild conditions (Scheme 15).97

![Scheme 14: Aldol disconnection strategy for the formation of 30.](image)
Scheme 15: Procedure for coupling of aromatic aldehydes with ketone C-glycosides using catalytic amounts of proline and TEA.\(^\text{97}\)

The initial steps towards aldehyde coupling partner 62 followed the same methodology as was developed in the Spring Group synthesis of 28,\(^\text{83}\) although slightly higher yields were noted (Scheme 16).\(^\text{98}\)

Scheme 16: Initial steps towards aldehyde 62.

In order to convert ester 51 to alcohol 53, 23 equivalents of NaBH\(_4\) were used in the previous route.\(^\text{83}\) This resulted in practical difficulties; as the alcohol is too polar for an aqueous workup to be used, large amounts of inorganic salts had to be removed by flash chromatography on silica gel. In an effort to solve this problem, a number of reduction conditions were trialled (Table 1). Firstly, reduction using diisobutyl aluminium hydride (DIBAL) was attempted. This stability of the tetrahedral intermediate resulting from the initial attack is known to give aldehydes directly on occasion,\(^\text{99}\) which would shorten the synthetic scheme. However, despite multiple attempts with differing equivalents of DIBAL and reaction temperatures, \(^1\)H NMR spectroscopic and LCMS analysis of the crude reaction mixture revealed a complex mixture of products in each case, and neither the aldehyde nor alcohol product were ever observed (Table 1, Entries 1-3). Next, it was observed that reaction with LiAlH\(_4\) also resulted in a mixture of products, including one for which \(^1\)H NMR spectroscopic analysis showed that the singlet corresponding to H-3 had been replaced by additional alkyl peaks, and the \(^{13}\)C NMR spectrum showed a peak at \(\delta = 193.9\) ppm, tentatively consistent with partial reduction to give 68 (Table 1, see appendix for spectra). However, an analytically pure sample of this species could not be obtained, and no other products could be identified. The survival of the ketone implies that initial attack of the hydride takes place \(\beta\)- to the quinolone carbonyl (at the 2-position of the quinolone ring system), with protonation at the 3-position only occurring upon aqueous workup (Scheme 17). This behaviour contrasts to that
observed during the synthesis of 25, where the heterocycle tolerates LiAlH₄ (Scheme 6). The heterocycle in that case possesses an acidic proton in place of the methyl involved here, which is presumably removed under the reaction conditions, rendering the heterocycle anionic and inert to further attack by LiAlH₄. Finally, treatment with a lower number of equivalents of NaBH₄ than used previously yielded the desired alcohol 53 in good yield (Table 1, Entry 5).

**Table 1**: Screening of conditions for the reduction of alcohol 51, alongside tentative structure of side product 68.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>DIBAL (4.0 equiv.), THF, 0 °C, 1 h</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>DIBAL (2.5 equiv.), THF, 0 °C, 40 min</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3</td>
<td>DIBAL (1.0 equiv.), THF, -78 °C, 1 h</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4</td>
<td>LiAlH₄ (2.0 equiv.), THF, 0 °C, 45 min</td>
<td>Complex mixture,&lt;sup&gt;a&lt;/sup&gt; trace of impure 68&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>5</td>
<td>NaBH₄ (5.0 equiv.), CH₂Cl₂/Methanol, 0 °C, 3 h</td>
<td>78% 53&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>As determined by <sup>1</sup>H NMR spectroscopic and LCMS analysis of the crude product. <sup>b</sup>Evidence for presence after flash column chromatography by LCMS, <sup>1</sup>H and <sup>13</sup>C NMR spectroscopy, but analytically pure sample not obtained. <sup>c</sup>Isolated yield.

**Scheme 17**: Attack of LiAlH₄ β- to the carbonyl of 53 explains the survival of the ketone moiety.

With 53 in hand, it was then necessary to bring about oxidation to the desired aldehyde 62. Dess-Martin periodinane had been used previously by L. Carro to achieve this (data not shown), however, given the relative expense of this reagent, alternative conditions were sought to provide sufficient material for thorough investigation of the aldol step. Oxidation using pyridinium chlorochromate (PCC) was considered; however, the alcohol was too polar to be soluble in the CH₂Cl₂ solvent typically used in such a reaction. Instead, a Swern oxidation was carried out, although around 50 equiv. of DMSO had to be used to bring about sufficient solubility (Scheme 18). The reaction proceeded in moderate yield (due to
solubility and purification problems) to give an inseparable mixture of the desired aldehyde 62 and its hydrate 70.

Scheme 18: Swern oxidation of ester 53 to the desired aldehyde 62 and hydrate 70 (5:1 ratio resepctively). *Approximate combined yield of 62 and 70 following flash column chromatography, calculated using M_r for 62.

This mixture was then subjected to proline-TEA catalysed aldol conditions with commercially available ketone 64 (Scheme 19). After four days, LCMS and ¹H NMR spectroscopic analysis of the reaction mixture implied the presence of the desired α,β-unsaturated ketone product 67, alongside a slightly larger amount of the uneliminated product 71. Impure samples of what were tentatively assigned as 67 and 71 could be isolated, with both contaminated with unknown minor species. In their original report, Wang et al. noted the reluctance of electron-deficient aldehydes to eliminate, and so forced elimination of the impure sample of intermediate 71 was attempted by heating with para-toluenesulfonic acid (PTSA), but this resulted in a complex mixture of unidentifiable products.

Scheme 19: Initial attempt of proline-TEA catalysed aldol condensation of a mixture of 62 and 70 with ketone 64, and subsequent attempt to bring about elimination of the β-OH intermediate 71.

Driving the elimination of water from the intermediate 71 was identified as the major issue in the optimisation of this aldol reaction. An additional benefit of performing the reaction under dehydrating conditions would be the conversion of hydrate 71 to the reactive aldehyde 62, perhaps increasing the rate of the reaction. To this end, the effect of adding 3Å mol. sieves (which selectively sequester water in preference to MeOH and other small organic molecules) to the reaction mixture was assessed. The reaction was carried in presence of the mol. sieves and the relative amounts of the starting materials 62 and 70, β-OH intermediate 71 and desired (α,β)-unsaturated product 67 were monitored with time using
LCMS analysis. A control experiment, with no mol. sieves, was carried out in parallel. The data are summarised in Graph 1; shown on the vertical axis is the peak weight for each species found in the UV trace of the LCMS. Whilst not quantitative, this gives some indication of the relative concentrations of the species.

Graph 1: Relative LCMS peak size corresponding to starting materials (62 and 70 combined), intermediate 71 and product 67 in the aldol reaction of 62 and 70 with 64, measured over the course of 44 hours in presence of 3Å mol sieves; shown alongside results of a control experiment under identical conditions but with no mol. sieves.

The data shown in Graph 1 suggests that the addition of mol. sieves leads to a faster and more complete depletion of starting materials 62 and 70. Meanwhile, whilst the build-up of the intermediate 71 was initially faster, the relative amount tended to the same value as seen at late time in the control experiment. This may be explained by its faster generation being balanced by faster elimination to the final product 67; indeed, faster and more complete production of 67 is observed. However, the desired product 67 never becomes the most abundant species, and even after very long times, residual starting materials were still observed. However, this data did provide evidence of promotion of the reaction by mol. sieves, so a scale-up of the reaction was carried out. Once the amount of residual starting materials was low as determined by LCMS, the mixture was acidified to pH 1 using conc. H₂SO₄ and heated to reflux for 24 h in an attempt to induce elimination of the intermediate alcohol 71. LCMS analysis of the reaction mixture at this point showed the desired product to be the major compound present. Unfortunately, following work-up, a complex mixture of unidentifiable products was observed, indicating decomposition had most likely occurred.

Whilst the above experiments showed that the planned aldol reaction is theoretically feasible, the difficulty in achieving complete conversion implied that the yield would be limited. In
combination with the apparent tendency for decomposition, this was taken as reason to abandon this strategy.

2.1.2 Approach 2: C-H Activation

An area of synthetic chemistry which has seen significant growth in recent years is that of C-H activation. The opportunity to functionalise such a seemingly inert bond offers opportunities for novel disconnections in situations where installation of activating groups such as halides would be challenging, alongside much improved atom economy.

Nickel has emerged as a promising catalyst in this area, proving itself to be particularly proficient at the insertion of unsaturated moieties into C-H bonds. In particular, C-H activation of the 4-quinolone scaffold at the 2-position has been reported (Scheme 20). Additionally, similar reactions have been carried out with alkynes to give alkyne products (Scheme 21). Taken together, these results imply an efficient disconnection of 30 to simple quinolone (72) and alkyne (74) starting materials, readily synthesised by methylation of 4-hydroxyquinoline 75 and addition of an ethynyl-metal species to 6-methyl-5-hepten-2-one 64 respectively (Scheme 22). A concern with making this disconnection is whether the terminal C-H bond of the alkyne is also susceptible to C-H activation by nickel. However, in a study where this activation was desired, it was observed to only take place upon addition of relatively strong base (Scheme 23). It was therefore expected that this bond should remain inert under the Lewis acidic conditions employed in Ni-catalysed alkyne insertion reactions.

![Scheme 20: Ni-catalysed C-H activation of 4-quinolone scaffold.](image)
Scheme 21: Ni-catalysed insertion of alkynes into C-H bonds. X, Y, Z = CH, N, C=O; R¹, R², R³ = alkyl, aryl.

Scheme 22: Proposed disconnection of natural product 30 utilising Ni-catalysed C-H activation.

Scheme 23: Terminal alkyne C-H activation requires addition of relatively strong base.

Synthesis of the required 4-quinolone coupling partner 72 was achieved in modest yield (purification by column chromatography was challenging, with much of the product obtained contaminated by an unidentified impurity) by methylation of 4-hydroxyquinoline 75 using iodomethane with sodium hydride as a base (Scheme 24). The O-Me regioisomer could not be isolated.
Addition of ethynylmagnesium bromide to 6-methyl-5-hepten-2-one (64) proceeded in good yield according to a literature procedure to give alcohol 74 (Scheme 25).\textsuperscript{110} Due to the highly sensitive nature of Ni\textsuperscript{(0)}, the decision was made to protect this alcohol. Attempted silylation with tert-butylidemethylsilyl chloride (TBDMSCI) was unsatisfactorily sluggish, with residual starting material still present after 24 hours, presumably due to the sterically hindered nature of this tertiary alcohol.\textsuperscript{111} Silylation reactions using silyl chlorides are known to be accelerated by the addition of iodine;\textsuperscript{112} however, in the present case this resulted in formation of what was tentatively assigned as the iodo-etherication product 79. Although an analytically pure sample could not be obtained, \textsuperscript{1}H NMR spectroscopy revealed a quartet of doublets at $\delta = 1.99$ ppm, with coupling constants $J = 13.5$ and 3.4 Hz respectively. The larger coupling is consistent with the values often reported between trans-diaxial and geminal protons in cyclohexanes, consistent with the formation of 79. Other multiplets in agreement with this conclusion were present (see spectrum in appendix), but insufficient data were obtained to deduce the relative stereochemistry. This product was not anticipated, as results in the original publication displayed tolerance of olefins, but perhaps formation of the six-membered ring in this case is particularly favourable. Finally, silylation using TBDMS-triflate did yield the desired alkyne 80\textsuperscript{108}, although product material isolated after column chromatography on silica was contaminated by TBDMS alcohol (in a ratio of 25\% as judged by \textsuperscript{1}H NMR spectroscopy).
Scheme 25: Synthesis of alkyne 80, which was contaminated with TBDMS alcohol. A side-product obtained during attempted protection of 74 was tentatively assigned as iodo-etherication product 79 on the basis of $^1$H NMR spectroscopy, but the relative stereochemistry could not be deduced. $^a$Calculated using integrals obtained from $^1$H NMR spectroscopy of the impure product. $^b$Determined by $^1$H NMR spectroscopy.

With the coupling partners in hand, the proposed Ni catalysed C-H activation was attempted (Scheme 26). Due to the highly air and moisture sensitive nature of the Ni(0) catalyst and phosphine ligand, these reagents had to be weighed out in a dry box under argon, and the reaction was carried out in a Schlenk tube. Unfortunately, only starting material was recovered. The absence of any product and the practical challenges involved caused this route to be abandoned after this attempt.

Scheme 26: Attempted Ni catalysed C-H activation.

2.1.3 Approach 3: Pd Coupling

The use of palladium-catalysed cross coupling had already proven to be effective in the assembly of natural products 25-28 (vide supra). Additionally, this chemistry was also exploited in the form of a Suzuki coupling to afford 35 during the synthesis of the anti-tumour agent intervenolin (36) (vide supra, Scheme 3).$^{81}$

Following disconnection of the N-methyl group from 30, disconnection using a Suzuki coupling in a manner analogous to that used in the synthesis of intervenolin (36) yields a 4-quinoline triflate 83 and a boronate ester 84, which, it was envisaged, could be derived by hydroboration of the alkyne 74 (already synthesised during the C-H activation approach, Scheme 25).$^{84}$ Alternatively, propargylic alcohols are known to undergo reduction to allylic alcohols in the presence of LiAlH$_4$.$^{113}$ Application of this functional group interconversion
(FGI) yields an internal alkyne 85 which can be disconnected by means of a Sonogashira coupling to once again give the alkyne 74.\textsuperscript{114} Additionally, triflate 83 could also be employed in a cross metathesis strategy as outlined in Scheme 28. Disconnection of the methyl from 30 gives 82 as before, and subsequent disconnection of the allylic alcohol alkene gives vinyl quinolone 87 and linalool 86. As a tertiary unprotected allylic alcohol, linalool falls into Class II when using Grubbs second generation catalyst, whilst 87 is sterically similar to styrene, which is in Class I, so some selectivity for the cross-product might be anticipated.\textsuperscript{115} Finally, it was anticipated that 87 could be readily synthesised through Suzuki coupling of triflate 83 with a simple vinyl boronate ester. Carrying out this coupling would also provide a model study for the proposed reaction of 83 with 84 as proposed in Scheme 27.

![Scheme 27: Retrosynthetic analysis for 30 using disconnections analogous to those used in the synthesis of intervenolin 36. PG = protecting group.](image)

In order to synthesise the 4-quinolone triflate 83 needed for the above strategies, the methodology used in the intervenolin synthesis was trialled initially. Whilst protection using the same TBDMS protecting group was successful (Scheme 29), a pure sample of 89 collected by recrystallization from MeOH was observed to rapidly decompose back to starting...
material, precluding full characterisation. This difference in stability could be explained by the lack of allylic strain with the methyl group at the 3-position allowing more extensive conjugation of the ether oxygen with the quinolone system, rendering the silyl group more labile, or perhaps residual MeOH from the recrystallization was inducing deprotection. To avoid this issue, a methoxymethylene (MOM) ether protecting group was attached in good yield under Finkelstein conditions to give $90^{116,117}$. The selectivity was confirmed by HMBC and NOESY NMR spectroscopy, and is presumably due to the 2-amide tautomer being dominant under the reaction conditions.$^{34}$ Subsequent reaction with triflic anhydride also proceeded in good yield to give the triflate coupling partner $91^{81}$

Next, triflate $91$ was subjected to Suzuki coupling with vinyl boronic acid pinacol ester $92$ to give the vinyl quinolone $93$ needed for the proposed cross metathesis approach. Pleasingly, the reaction proceeded with good yield, clearly demonstrating the suitability of $91$ as a coupling partner (Scheme 30).$^{81}$

Meanwhile, linalool ($86$) was synthesised from 6-methyl-5-hepten-2-one $64$ in an analogous manner to alkyne $74$, with the low yield attributed to decomposition during storage of the Grignard reagent. $86$ was then subjected to alkene metathesis conditions with olefin $93$ (Scheme 31). Unfortunately, LCMS analysis of the crude reaction mixture revealed that most of olefin $93$ remained unreacted, alongside masses consistent with small amounts of desired
product, the homodimer and also the cross-dimer formed by reaction with the more hindered trisubstituted olefin of linalool. The absence of linalool by TLC implied that it was forming mainly homodimers, or cyclising intramolecularly. These observations revealed a lack of selectivity for cross metathesis between these substrates and a low reactivity of olefin 93, perhaps due to the electron withdrawing properties of the quinoline ring or as a result of coordination of the quinolone nitrogen to the ruthenium catalyst.\textsuperscript{118}

![Scheme 31](image)

**Scheme 31**: Synthesis of linalool 86, and subsequent attempted cross metathesis of olefin 93 with linalool 86.

With triflate 91 in hand and its suitability as a Suzuki coupling partner demonstrated, the formation of an (E)-vinylboronate ester from alkyne 74 was required. Firstly, hydroboration with pinacolborane in the presence of a catalytic amount of the Schwartz reagent was attempted (Scheme 32).\textsuperscript{119} However, whilst \textsuperscript{1}H NMR spectroscopic analysis of the crude reaction mixture did suggest the production of some of the desired product 95, the main product seemed to possess a terminal vinyl group, suggesting protodeboronation of the product was taking place under the reaction conditions, perhaps due to the unprotected alcohol moiety in 74.\textsuperscript{120}

![Scheme 32](image)

**Scheme 32**: Attempted hydroboration of alkyne 74.

Next, hydroiodination of 74 was attempted; if this proved successful, it was anticipated that Miyaura borylation could then be used to convert the iodoalkene 96 to the desired boronate ester 95.\textsuperscript{87} However, stoichiometric hydrozirconation and hydroalumination with the Schwartz reagent (1.2 equivalents) and DIBAL (1.1 equivalents) respectively, followed by quenching with iodine, both returned starting material.\textsuperscript{121,122}
Table 2: Attempted hydroiodination of alkyne 74.

<table>
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<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
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<tbody>
<tr>
<td>1</td>
<td>i) Cp₂ZrHCl, ii) I₂, CH₂Cl₂, 0 °C → r.t., 30 min</td>
<td>NR&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>i) DIBAL, ii) I₂, CH₂Cl₂, r.t., 4 h</td>
<td>NR&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

NR = No reaction. <sup>a</sup>As determined by <sup>1</sup>H NMR spectroscopic analysis of the crude product.

With success in the efforts to make the coupling partner needed for the Suzuki approach not forthcoming, attention was turned to the Sonogashira approach, whose coupling partner, alkyne 74, was already in hand. Pleasingly, this was found to undergo facile coupling in excellent yield with triflate 91 under Sonogashira conditions to give intermediate 97, which possesses all but one of the carbon atoms present in natural product 30 (Scheme 33).<sup>123</sup>

Reduction of the propargylic alcohol 97 with LiAlH₄ was then attempted (Scheme 34), and LCMS analysis of the crude product indicated that the addition of two hydrogens had proceeded as desired.<sup>113</sup> However, the <sup>1</sup>H NMR spectrum lacked the expected trans-olefin peaks, with alterations of the aromatic signals instead, consistent with reduction of the heterocycle. This is in line with the reactivity which was observed during the attempted reduction of ester 51. However, as mentioned in that case, the previous routes suggest that the quinolone system tolerates these harsh reductive conditions if the nitrogen has a proton attached, as presumably this is deprotonated under the reaction conditions, rendering the ring anionic and unreactive. In order to bring about this functionality, intermediate 97 was deprotected to form 98 with excellent yield under acidic conditions. Pleasingly, 98 was indeed observed to undergo reduction to (E)-allylic alcohol 100 in presence of LiAlH₄ with good yield, and survival of the 4-quinolone ring. The reaction is presumed to proceed via a cyclic organoaluminium species 99, which gives the desired alkene upon quenching with water.
Scheme 34: Direct reduction of 97 with LiAlH₄ resulted in reaction of the quinolone core, however deprotected 99 reacted as desired to give 100, which was then regioselectively methylated to supply natural product 30.

It then just remained to attach a methyl group to the nitrogen of the quinolone ring of 100 (Scheme 34). However, this was known to not be a trivial matter, as the steric demand of a 2-substituent often results in poor yields, whilst selection for N- vs. O- alkylation can also be problematic. Nonetheless, methylation using iodomethane and LiOt-Bu (the same base as was used for the analogous step in the synthesis of intervenolin) resulted in generation of natural product 30 with moderate yield. The Li⁺ counterion may coordinate to the quinolone oxygen, blocking its methylation, and indeed no O-methyl product was observed. By ¹H NMR spectroscopy, 30 was the dominant species in the crude product mixture, and so it may be that the low yield is due to loss into the aqueous phase during workup. Optimisation of this aspect of the procedure could likely improve the yield, but given that 30 was ultimately obtained more efficiently using an alternative method (see below), this was not attempted.

The identity of the product was confirmed by comparison of its ¹H NMR spectrum with that reported in the original natural product isolation paper.¹⁶

With the first natural product 30 in hand, there was an opportunity to validate the proposed divergent strategy, whereby multiple natural products could be obtained from mutual late stage intermediates. In this case, it was desired to convert 30 into its regioisomeric partner
It was proposed that treatment with sources of Cr(VI) could prove productive to this end, as this is known to be able to bring about 1,3-transpositions of allylic alcohols in a transformation known as the Babler-Dauben oxidation (Scheme 35). Only the rearranged form 101 would be able undergo further oxidation under the reaction conditions, which would render the transformation irreversible. Then, the resultant enone 101 could undergo Luche reduction to give the rearranged natural product 29.

Scheme 35: Attempted conversion of 30 to 29 by means of Babler-Dauben oxidation followed by Luche reduction. The allylic transposition is proposed to take place via two possible pathways: Pathway A (concerted and sigmatropic) and Pathway B (stepwise and dissociative).

Putting this into practice, 30 was treated with pyridinium chlorochromate (PCC); however, this resulted in recovery of starting material. It is thought that the allylic transposition can take place via two possible mechanisms as shown in Scheme 36: either in a concerted, sigmatropic (Pathway A) or stepwise, dissociative fashion (Pathway B). Given the strongly acidic conditions associated with PCC, it seems rational that the dissociative pathway would dominate; however, this would generate a carbocation in conjugation with the relatively electron deficient quinolone system in 103, and so perhaps this could account for the lack of reactivity. Given that the alternative Cr(VI) reagent pyridinium dichromate (PDC) is less acidic, and so could perhaps favour the concerted pathway, exposure to PDC was attempted. Unfortunately, this once again resulted in the recovery of starting material. Furthermore, the later observation of acid-mediated equilibration between 29 and 30 (vide infra) implies that carbocation 103 is in fact accessible, implying that factors other than electronics were disfavouring the oxidative rearrangement in the present case.
Given these discouraging results, an alternative approach was sought. It was proposed that rearrangement between the natural products in the reverse direction would prove more fruitful, as the additional conjugation between the olefin and quinolone core in 30 could provide a thermodynamic drive for the rearrangement. Such a transformation could simply occur under acidic conditions, or alternatively, rhenium catalysed conditions are known which can bring about 1,3-transpositions of allylic alcohols. To this end, an alternative route to natural product 29 was designed, which will be discussed in the following section.

Scheme 36: Proposed equilibration of 29 with 30 under acidic conditions, favouring 30 due to the additional conjugation of the olefin with the quinolone core.

2.2 Divergent Synthesis of 29 and 30

In the synthetic route towards 29, it was desired to utilise intermediates which had already been used in the synthesis of 30 discussed in the previous section, enabling further implementation of the chemistry which had already been developed. It was anticipated that a substrate 106 analogous to 91, with the pseudohalide triflate replaced by a true halide, could undergo lithium-halogen exchange to give a heteroaryl-metal species 107 which could then engage geranial 108 to give natural product 29 in MOM-protected form (Scheme 37).

Scheme 37: Proposed lithium-halogen exchange route towards natural product 29.

In order to access geranial 108 with the desired double bond geometry at the enal moiety, it was necessary to synthesise this from geraniol 110 (although the compound is also readily commercially available as a mixture with its isomer as citral). Oxidation proceeded according to a literature procedure utilising MnO₂ with good yield (Scheme 38). An extended reaction
time was required; however, the rate of oxidation by MnO₂ has been reported to vary depending on the source of the reagent itself, and additionally, the large amount of MnO₂ required made the maintenance of a homogenous reaction mixture technically challenging. Additionally, the MnO₂ was not activated by heating prior to use.

Scheme 38: Oxidation of geraniol 110 to geranial 108.

With the desired electrophile in hand, attention was turned to the generation of a suitable halide. Bromination of 90, which had already been accessed during the previous synthesis, was attempted (Table 3). A literature procedure utilising CBr₄ and PPh₃ was trialed (Table 3, Entry 1), and LCMS analysis of the crude product showed masses tentatively consistent with a mixture of the desired product 111 and a di-bromo analogue 112 being generated. Column chromatography gave 111 and 112 with approximate yields of 10% and 23% respectively, although neither compound was analytically pure, with the only evidence obtained for their structures being the mass obtained by LCMS analysis. Alternative conditions using N-bromosuccinimide (NBS) and PPh₃ were also trialled (Table 3, Entry 2), but with a similar outcome, and even more 112 being produced, and only trace amounts of 111 being detected. These results imply that the MOM protecting group was intolerant of the reaction conditions, allowing the second bromination event to occur. In an attempt to mitigate this, the CBr₄ conditions were trialled at room temperature (Table 3, Entry 3), but this approach suffered from poor solubility and returned the starting materials. Alternatively, as acid would be released as a side product during these reactions, which could be causing deprotection, TEA was added in order to maintain a higher pH (Table 3, Entry 4). However, this was observed to result in complete inhibition of the reaction. Finally, application of the CBr₄ mediated reaction at intermediate temperature still resulted in a mixture of both products, but a small sample of impure 111 was obtained with a yield of around 17% following column chromatography.
Table 3: Attempted bromination of 90 to 111, alongside generation of the undesired di-bromo compound 112.

```
Entry | Conditions | Result
--- | --- | ---
1 | CBr₄, PPh₃, toluene, 110 °C | Mixture of 111 and 112 (approx. 10%² and 23%² respectively)
2 | NBS, PPh₃, dioxane, 100 °C | Mainly 112, trace 111ᵇ
3 | CBr₄, PPh₃, toluene, r.t. | Poor solubility, NRᵇ
4 | CBr₄, PPh₃, TEA, toluene, 110 °C | NRᵇ
5 | CBr₄, PPh₃, toluene, 80 °C | ~17%ᵃ impure 111
```

NR = no reaction. ²Isolated yield. ᵇDetermined by LCMS analysis of crude product.

In an alternative approach, conversion of the triflate 91 (already accessible as part of the previous synthesis, see Scheme 29) to the bromide was also attempted. This utilised tetrabutylammonium bromide (TBAB) as a bromide source (Scheme 39), but resulted in a complex mixture of unidentifiable species, as determined by LCMS analysis of the crude product. Finally, the impure sample of 111 (Table 3, Entry 5) was used to assess the feasibility of the lithium-halogen exchange sequence shown in Scheme 37. Thus, 111 was treated sequentially with one equivalent of n-BuLi and then geranial 108; however, no product was observed, and a significant amount of starting material remained. Given this discouraging result, and the difficulties which had been encountered in accessing the bromide, this approach was abandoned.

In devising an alternative synthesis of the natural product 29, attention came to a report by Bernini et al. in which 4-quinolones 117 were constructed in a short sequence consisting of Michael addition of amines to ynones 115, followed by a copper-catalysed heterocyclisation (Scheme 40A). ¹³⁶ The ynones themselves could be easily constructed through Sonogashira coupling of simple alkynes 114 with a commercially available acid chloride 113. Other similar approaches also exist in the literature. ¹³⁷,¹³⁸ Application of this chemistry to a retrosynthetic
analysis of the target natural product 29 led to three simple precursors: the acid chloride 113, methylamine and a simple alkyne 118, which itself could be easily accessed from geranial 108 (Scheme 40B). The modularity of this approach would grant the synthesis great efficiency, whilst also expediting generation of analogues of the natural products. Moreover, this would represent to our knowledge the first synthesis of 1,2-dialkylquinolones by such a method. Indeed, in the work by Bernini et al. (Scheme 40A), cyclisation of compounds with $R^1 = \text{alkyl}$ required more forcing conditions and gave lower yields, whereas those with $R^2 = \text{aryl}$ failed to give any product. It was anticipated that through optimisation these limitations could be overcome.

Scheme 40A: Modular approach to 4-quinolones by Bernini et al., utilising sequential Sonogashira, Michael addition and copper catalysed heterocyclisation reactions. $R^1 = \text{alkyl, aryl}$; $R^2 = \text{aryl}$.

The forward synthesis commenced with the construction of the requisite alkyne 118, and reaction of geranial 108 with ethynylmagnesium bromide proceeded with good yield (Scheme 41), and then Sonogashira coupling using the previously published conditions was attempted. LCMS analysis of the crude product following workup detected a mass corresponding to 119, which implied that the desired reaction was indeed occurring, but that the unprotected alcohol present in the product was also engaging in an esterification reaction. Nonetheless, the result was encouraging, and it was decided to protect the alcohol in both MOM form 121 and TIPS form 120, in order to preclude the competing esterification.
Scheme 41: Synthesis of alkyne 118 and subsequent attempted Sonogashira coupling with acid chloride 118, and protection as TIPS ether 120 and MOM ether 121.

These protected propargylic alcohols were then submitted to the Sonogashira coupling conditions.\textsuperscript{136} TIPS ether 120 showed no reaction, perhaps as a result of the large steric bulk associated with the silyl protecting group. However, the MOM protected analogue 121 was able to successfully take part in the desired coupling with moderate yield (LCMS analysis of the crude product indicated some residual starting material was still present). Upon heating the resultant product 123 in a sealed tube with methylamine, Michael addition was observed, providing enamine 124 in a quantitative yield. The double bond geometry was assigned on the basis of NOESY NMR data, and is likely preferred both by steric grounds (avoiding the clash of the alkyl side chain with the carbonyl) and due to the presence of an intramolecular hydrogen bond between the N-H and carbonyl in the conformation shown.\textsuperscript{136}

Scheme 42: Sonogashira coupling of protected propargylic alcohols 120 and 121, and subsequent Michael addition with methylamine. Key NOESY correlations supporting the olefin geometry of 124 are shown in red. NR = no reaction.
The stage was then set for the key heterocyclisation event. Initially, 124 was exposed to the copper catalysed conditions mentioned above\(^\text{136}\) however, LCMS analysis of the crude product showed that the desired product was not forming, with a mass corresponding to an unidentified dimer detected instead (Table 4, Entry 1). This was unsurprising, given the lack of tolerance for this class of substrate which was noted in the original publication. Next, Buchwald-Hartwig conditions were attempted\(^\text{140}\). With the catalyst Pd\(_2\)dba\(_3\) and the ligand P(\(\alpha\)-tolyl)\(_3\), no reaction was observed (Table 4, Entry 2); however, upon switching the ligand to P(2-furyl)\(_3\), 125 could be isolated with quantitative yield. It is likely that the decreased bulk of the ligand is responsible for this marked increase in reactivity.

Table 4: Heterocyclisation of 124 to give MOM protected natural product 125.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>CuI (5 mol%), DMEDA (5 mol%), (t)-BuONa, DMSO, 80 °C</td>
<td>Unidentified dimer(^a)</td>
</tr>
<tr>
<td>2</td>
<td>Pd(_2)dba(_3) (5 mol%), P((\alpha)-tolyl)(_3) (20 mol%), K(_2)CO(_3), toluene, 100 °C</td>
<td>NR(^a)</td>
</tr>
<tr>
<td>3</td>
<td>Pd(_2)dba(_3) (5 mol%), P(2-furyl)(_3) (20 mol%), K(_2)CO(_3), toluene, 100 °C, 24 h</td>
<td>Quantitative 125(^b)</td>
</tr>
</tbody>
</table>

NR = no reaction. \(^a\)Determined by LCMS analysis of crude product. \(^b\)Isolated yield.

With the natural product in hand in protected form, removal of the MOM group was required. Upon room temperature treatment with equal amounts of methanol and aqueous HCl, no reaction was observed (Table 5, Entry 1), whereas addition of excess acid resulted in decomposition, with LCMS analysis of the crude product showing formation of a complex mixture of unidentifiable products (Table 1, Entry 2). Treatment with acidified MeOH at elevated temperature gave a more controlled reaction, and a 31% yield of natural product 29 was obtained (Table 5, Entry 3). However, LCMS analysis of the crude product following workup also identified the presence of a species with a mass corresponding to 126, indicating that under the acidic reaction conditions, addition of methanol across a double bond was occurring. It was therefore proposed that a change in solvent might result in an improved yield. Gratifyingly, treatment with pyridinium \(p\)-toluenesulfonate (PPTS) in \(t\)-BuOH, followed by workup and flash column chromatography, resulted in what was revealed by \(^1\)H NMR spectroscopic analysis to be a 4:1 mixture of 29 and 30 respectively, with a combined
yield of 72% (Table 5, Entry 4). The pair were separable by preparative HPLC, and so both natural products were accessible in a single synthetic step, in a clear demonstration of the efficiency of the chosen divergent strategy. It seems probable that the allylic alcohol transposition proceeds via a protonation of the alcohol favouring elimination of water to give a conjugated carbocation, which may recombine with water to give either 29 or 30 depending on the position of attack.

**Table 5:** Deprotection of 125 to give natural product 29, with concomitant rearrangement to give natural product 30.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1:1 MeOH / 1.5N HCl, r.t.</td>
<td>NR</td>
</tr>
<tr>
<td>2</td>
<td>1:1 MeOH / excess conc. HCl, r.t.</td>
<td>Decomposition&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3</td>
<td>MeOH/ few drops conc. HCl, 50 ºC, 16 h</td>
<td>31% 29&lt;sup&gt;b&lt;/sup&gt;, 126 detected&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4</td>
<td>PPTS, t-BuOH, 80 ºC, 48 h</td>
<td>4:1 29 : 30, 76% total yield&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

NR = no reaction. <sup>a</sup> Determined by LCMS analysis. <sup>b</sup> Isolated yield. <sup>c</sup> Combined yield of mixture containing 29 and 30 only.

**2.2.1 Attempted Asymmetric Synthesis of 29 and 30**

With access to these natural products now granted in racemic form, an extension of the strategy was sought whereby they could be accessed in enantioenriched form. In the original isolation paper, the compounds were observed to be optically active, but their absolute stereochemistry was not defined. Through comparison of the sense of optical rotation of enantioenriched synthetic samples with those previously reported, this ambiguity could be resolved. It was anticipated that racemic propargylic alcohol 118 could be prepared enantioselectively through oxidation to 127 followed by a Corey-Bakshi-Shibata (CBS) reduction or similar (Scheme 43). It was anticipated that the alkynyl substituent would be sufficiently small to allow good distinction between the prochiral faces of the ketone. The resultant enantioenriched 118<sup>*</sup> could then be advanced as previously to give 29<sup>*</sup>, also enantioselectively (assuming that racemisation could be avoided in the intervening steps). As the existing rearrangement to 30 is proposed to occur via a planar carbocation intermediate,
it is likely that the enantioenrichment would be lost should this method be attempted to synthesis 30*. However, rhenium-catalysed conditions exist which are known to bring about 1,3-equilibration of allylic alcohols with partial retention of stereochemistry.145

To investigate this strategy, a number of conditions for the oxidation of 118 were explored (Table 6). Exposure to PCC in the presence of 4Å molecular sieves resulted in a complex mixture of unidentifiable products, as determined by TLC analysis (Table 6, Entry 1). Next, the MnO₂ mediated conditions, which had proved successful previously for geraniol 108 (Table 6, Entry 2), were trialed. This also resulted in extensive decomposition, although column chromatography yielded a trace of a compound for which LCMS analysis showed a mass matching that of structure 128. Further evidence for this tentative structural assignment was provided by ¹H NMR spectroscopy, which showed signals of equal integral at δ = 8.64, 6.85 and 5.17 ppm, alongside the expected aliphatic resonances (although these were observed to overlap with impurity peaks, see spectrum in appendix for details). The resonance at 8.64 ppm is particularly suggestive of a highly symmetrical, electron deficient aromatic system. Similar [2+2+2] cyclotrimerisations are well known for transition metals, however the employment of manganese has thus far been limited, although a related transformation using Mn(I) has been reported.146,147 However, the sample of 128 which was obtained was not sufficiently pure for full characterisation, and was present only as a trace, and so this intriguing result was not pursued further. Finally, treatment with DMP also resulted in decomposition at both room temperature and 0 °C as determined by TLC analysis. Given this apparent tendency to undergo decomposition under even mild conditions, it seems likely that the desired product is highly unstable. As such, this route was abandoned, as the racemic material already obtained would be adequate for initial biological screening.

Scheme 43: Proposed enantioselective route to natural products 29 and 30. Absolute stereochemistries shown for 118* etc. is arbitrary and for illustrative purposes only.
Table 6: Attempted oxidation of propargylic alcohol 118 to 127, alongside tentatively-identified side-product 128.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>PCC, 4Å mol. sieves, CH₂Cl₂, 0 °C → r.t.</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>MnO₂, CH₂Cl₂, r.t.</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;, putative trace of 128 detected&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>3</td>
<td>DMP, CH₂Cl₂, r.t.</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4</td>
<td>DMP, CH₂Cl₂, 0 °C</td>
<td>Complex mixture&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Determined by TLC analysis. <sup>b</sup>Impure sample obtained by column chromatography and analysed by <sup>1</sup>H NMR spectroscopy and LCMS, giving tentative assignment.

2.3 Total Synthesis of 25, 27, 31 & 32

With access to six of the eight natural products (25-30) now granted, attention returned to the final two. In the proposed divergent route towards these (Scheme 12), natural product 25, which represents the core structure found in three of the other natural products, was to be used as a key late-stage intermediate in their synthesis. In order to access 25, it was decided to employ the route which had been developed previously by the Spring Group, which used an sp<sup>2</sup>-sp<sup>3</sup> Suzuki coupling to unite the prenyl side chain with the quinolone core.<sup>82</sup>

To facilitate this, the necessary boronate ester coupling partner was first constructed (Scheme 44). Treatment of cyclopropyl methyl ketone 129 with methylmagnesium iodide followed by acid brought about a rearrangement to iodide 130 with excellent yield (the previous route used methylmagnesium bromide, giving the analogous bromide product).<sup>148</sup> This then underwent an S<sub>N</sub>2 reaction with lithium acetylide to give alkyne 40, which was not isolated.<sup>149</sup> Subsequent zirconium-catalysed carboalumination followed by quenching with iodine gave vinyl iodide 41.<sup>150</sup> The poor isolated yield is thought to be as a result of the volatility of alkyne intermediate 40. Finally, Miyaura borylation gave the desired boronate ester coupling partner 38, with an improved yield noted over the previously reported 40%.
Meanwhile, the quinolone coupling partner was constructed using the Conrad-Limpach method. Thus, aniline underwent condensation with 131 to give 132,\(^{151}\) with an improvement over the previous reported yield of 61%. Treatment with polyphosphoric acid then induced cyclisation to quinolone 42. The low yield was in agreement with that reported previously using the same conditions (50%), and is perhaps due to the harsh conditions necessary. The ester moiety was then reduced with lithium aluminium hydride to supply alcohol 43, with an improved yield again noted.

At this juncture, some new methodology was attempted. Regioselective direct \(N\)-alkylation of these 4-quinolones had already been identified as challenging, with low conversions and generation of the \(O\)-alkylated isomer being noted. It was hypothesised that the alcohol moiety could act as a tether, helping to favour the desired regioselectivity. Condensation of 43 with a formaldehyde equivalent could reasonably be expected to give cyclised 133 as the thermodynamic product (Table 7). Then, treatment with acid could generate a transient iminium ion 134, which could undergo reduction \textit{in situ} to the desired \(N\)-methyl product 60 with high regioselectivity. Similar transformations have been applied in amino acid synthesis.\(^{152}\)
To this end, a number of approaches were attempted to bring about condensation of 43 with formaldehyde (Table 7), with the reaction monitored by TLC analysis in each case. Firstly, exposure to paraformaldehyde under aqueous conditions at a range of temperatures gave no conversion in each case (Table 7, Entries 1-3). Switching to non-aqueous conditions and the introduction of PTSA as a catalyst offered no improvement (Table 7, Entry 4). In an effort to shift the equilibrium of the reaction towards the products, dehydrating conditions were trialled. However, with the introduction of 4Å molecular sieves (Entry 5) and the employment of Dean-Stark conditions (Table 7, Entry 6), no reaction was observed. Finally, reaction was attempted with dibromomethane under phase-transfer catalysed conditions,\textsuperscript{153} which did show consumption of starting material; however, LCMS analysis suggested the formation of an unidentified dimer rather than the desired product. The apparent difficulty of this reaction may be due to the low nucleophilicity of the nitrogen in 43, due to the involvement of its lone pair in the extended conjugation of the quinolone core. As a result, this approach was abandoned, and the previously reported synthesis was resumed.

\textbf{Table 7}: Attempted route for regioselective methylation of 43, consisting of condensation with formaldehyde to give 133, followed by reduction of imine 134 under acidic conditions.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), H\textsubscript{2}O, r.t.</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>2</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), H\textsubscript{2}O, 65 °C</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>3</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), H\textsubscript{2}O, r.t., 100 °C</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>4</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), p-TSA, toluene / THF, reflux</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>5</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), p-TSA, toluene, 4Å mol. sieves, reflux</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>6</td>
<td>(CH\textsubscript{2}O\textsubscript{n}), p-TSA, toluene, Dean-Stark</td>
<td>NR\textsuperscript{a}</td>
</tr>
<tr>
<td>7</td>
<td>TBAB, CH\textsubscript{2}Br\textsubscript{2}, 50% NaOH\textsubscript{aq}, reflux</td>
<td>Dimer detected\textsuperscript{b}</td>
</tr>
</tbody>
</table>

NR = no reaction. \textsuperscript{a}Determined by TLC analysis. \textsuperscript{b}Mass observed in LCMS analysis

Alcohol 43 underwent chlorination with thionyl chloride to give 45 with moderate yield, perhaps due to the presence of some residual starting material. This was then successfully united with its boronate ester coupling partner under Suzuki conditions to give natural product 25, with a moderate yield comparable to that reported previously.\textsuperscript{82} Whilst such sp\textsuperscript{2}-
sp³ coupling reactions are traditionally considered challenging due to competing β-hydride elimination, this issue is avoided in the present case as 45 lacks any hydrogens in the β-position. Access to this compound allowed evaluation of the proposed divergent approach to give the remaining natural products. In the event, treatment of 25 with LiOtf-Bu (previously noted to confer good N- vs. O-selectivity³¹) followed by iodomethane provided natural product 27 with moderate yield, due to the difficulty in achieving full conversion, with multiple base-iodomethane treatment sequences required. Whilst this had been synthesised previously,⁸² the prior strategy used a quinolone coupling partner for the Suzuki step which already had the 1-Me in place, which itself required a seven-step synthesis including protection and deprotection (Scheme 8). The current route therefore offers a markedly more efficient synthesis. Next, exposure to m-CPBA in the presence of sodium bicarbonate brought about epoxidation to give natural product 32. ¹H NMR spectroscopic analysis of the crude product following workup showed the presence of additional peaks which could correspond to the regioisomeric product resulting from epoxidation at the alternative olefin. However, this was present only in a ratio of 15% (as determined by peak integration), and could not be isolated. This may account in part for the relatively low yield, alongside the small scale at which the reaction was carried out (~20 mg). Presumably, the reaction is rendered regioselective by the steric bulk of the quinolone system adjacent to the alternative olefin. This scheme therefore granted access to three natural products in direct succession, a clear demonstration of the efficiency of the divergent strategy.

**Scheme 46:** Conversion of alcohol 43 to natural product 25 by chlorination followed by Suzuki coupling, and subsequent derivatisation to natural products to 27 and 32.

Unfortunately, methylthiomethylation to give the final natural product 31 proved more challenging, despite the screening of a broad range of conditions (Table 8). The reactions
were monitored by taking small aliquots which were partitioned between water and EtOAc, followed by TLC analysis, visualising using both UV irradiation and KMnO$_4$ staining. Exposure to methylthiomethylene chloride (MTMCl) analogously to the successful methylation of 25 to 27 (Scheme 46) gave a very poor isolated yield (6%) of the desired natural product 31, alongside an isolated 37% of recovered starting material (Table 8, Entry 1). Variation of the number of equivalents of each reagent used offered no improvement, and demonstrated irreproducibility in the generation of 31 by this method (Table 8, Entries 2-3). To try to induce conversion, the reaction was heated to reflux (Table 8, Entry 4), but analysis showed the presence of starting material alone. It was then hypothesised that the product may be unstable under the reaction conditions, with potential elimination of volatile methane thiol, which would then be lost from the reaction mixture. In an attempt to disfavour this, the reaction was carried out in a sealed tube, but this offered no improvement (Table 8, Entry 5). Next lithium hydride was trialled as a base, but despite different temperatures and solvents being explored, no reaction resulted (Table 8, Entries 6-8). Then, NaI was added as a nucleophilic catalyst in a set of conditions which had proved successful in the literature on an amide substrate,$^{154}$ and in this case some consumption of starting material was observed (Table 8, Entry 9). However, no identifiable product was obtained in this case. In an effort to prevent any potential dimerization, the reaction was repeated at higher dilution, but this resulted in no reaction as determined by LCMS analysis of the crude product (Entry 10). Finally, $n$-BuLi was trialled as a base, but again to no avail (Entry 11).
**Table 8:** Attempted optimisation of methylthiomethylation of 25 to 31. MTMCl = methylthiomethylene chloride.

![Chemical structures](image)

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>LiO\textsubscript{t}-Bu (1.1 eq), MTMCl (3.0 eq), THF, 0 °C → r.t.</td>
<td>6%\textsuperscript{a}, 37% RSM\textsuperscript{b}</td>
</tr>
<tr>
<td>2</td>
<td>LiO\textsubscript{t}-Bu (2.0 eq), MTMCl (4.0 eq), THF, 0 °C → r.t.</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>3</td>
<td>LiO\textsubscript{t}-Bu (1.1 eq), MTMCl (6.0 eq), THF, 0 °C → r.t.</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>4</td>
<td>LiO\textsubscript{t}-Bu (1.1 eq), MTMCl (3.0 eq), THF, 0 °C → reflux</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>5</td>
<td>LiO\textsubscript{t}-Bu (1.1 eq), MTMCl (6.0 eq), THF, 0 °C → 70 °C, sealed tube</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>6</td>
<td>LiH (1.1 eq), MTMCl (6.0 eq), THF, 0 °C → r.t.</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>7</td>
<td>LiH (1.1 eq), MTMCl (6.0 eq), THF, 0 °C → 70 °C, sealed tube</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>8</td>
<td>LiH (1.1 eq), MTMCl (6.0 eq), DMF, 0 °C → 70 °C, sealed tube</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>9</td>
<td>LiO\textsubscript{t}-Bu (2.2 eq), MTMCl (4.0 eq), NaI, THF, 0 °C → r.t.</td>
<td>Low conversion\textsuperscript{c}, no identifiable product</td>
</tr>
<tr>
<td>10</td>
<td>As Entry 9, but with higher dilution</td>
<td>NR\textsuperscript{c}</td>
</tr>
<tr>
<td>11</td>
<td>(n)-BuLi (1.05 eq), MTMCl (1.2), THF/DMSO, 0 °C → r.t.</td>
<td>NR\textsuperscript{c}</td>
</tr>
</tbody>
</table>

NR = No reaction. \textsuperscript{a}Isolated yield. \textsuperscript{b}Starting material was isolated. \textsuperscript{c}As determined by LCMS analysis of the crude product following work up.

Nonetheless, a sufficient quantity of the desired natural product 31 had been collected to facilitate some preliminary biological testing (as discussed below), and this therefore completed the synthesis of all eight *Pseudonocardia* natural products by the Spring Group.
3.0 Conclusions and Future Work

Two synthetic schemes were developed, giving access to a total of six natural products (25, 27, 29-32), four of which (29-32) had never before been synthesised. The power of the proposed divergent synthesis was demonstrated, with multiple natural products efficiently accessed from late stage intermediates. The synthesis was reported in the literature in a 2016 publication.\(^{155}\)

In particular, a 4:1 separable mixture of 29 and 30 was accessible in only six steps with an overall yield of 30% (Scheme 47). In addition, an alternative approach giving 30 directly was also developed (Section 2.1), which could find utility in the construction of analogues of the natural products with structures unsuitable for synthesis by the method depicted in Scheme 47.

Scheme 47: Synthetic route for the synthesis of 29 and 30.

The route developed for the synthesis of 25, 27, 31 & 32 is shown in Scheme 48. This commenced with the construction of natural product 25 by the method developed previously in the Spring Group.\(^{82}\) This core scaffold was then derivatised to give directly the remainder of the natural products, with regioselective methylation and then epoxidation giving 27 and 32 directly. Methylthiomethylation to give 31 proved more challenging, but nonetheless, a
A sufficient quantity of material was obtained to facilitate some preliminary biological experiments.

**Scheme 48:** Synthetic route for synthesis of 25, 27, 31 & 32.

With these natural products now in hand, attention turned to the generation of structural analogues. By elucidating the change in biological activity resulting from small alterations of the compounds, details of the structure activity relationship (SAR) may be elucidated, enabling the design of more potent analogues. Of all the chemistry developed towards the natural products, the modularity of the route towards allylic alcohols 29 and 30 rendered it most suitable for this task. As outlined in Scheme 49, derivatised acid chlorides 140 could undergo Sonogashira coupling with a range of terminal alkynes 114. The resultant yrones...
141 could then undergo Michael addition with a selection of amines 142, which following heterocyclisation would give the desired analogues 144.

Scheme 49: Application of the methodology developed for the synthesis of allylic alcohols 29 and 30 to the synthesis of structural analogues.

This strategy formed the basis an M.Sci. project which was carried out by Teodora Coroama (Spring Group). The synthesis proceeded largely as expected, although some optimisation of the final cyclisation step was required (data not shown). A total of ten analogues were accessed, as shown in Figure 14. These may be split into two series with varying substituents on the nitrogen: one in which the side chain has been replaced by linear saturated analogue of similar length (A), and one in which it has been truncated (B).
Figure 14A: Analogues of the natural products containing a saturated side chain. B: Analogues of the natural products containing a truncated side chain. Synthesis carried out by Teodora Coroama.

Some preliminary screening of the entire library of natural products and analogues has been carried out by Suzie Forrest (Welch Group, Department of Biochemistry, University of Cambridge). As a convenient phenotypic assay to gauge whether the compounds are able to modulate quorum sensing in *P. aeruginosa*, the production of pyocyanin was measured. As discussed in the introduction, this toxin is known to be under regulation by PQS quorum sensing, and is responsible for cell damage in the lungs of those suffering from infection.\textsuperscript{156}

Cultures of wild type *P. aeruginosa* were grown in the presence of each of the compounds at a concentration of 200 μM, and after 8 hours, pyocyanin was extracted from each colony. The OD\textsubscript{520} was then measured, as pyocyanin absorbs light at this wavelength, and the value was normalised by the population of each culture, as measured by OD\textsubscript{600}. The resultant values are shown in Figure 15, alongside the values obtained for an untreated colony and one treated with a DMSO blank. None of the compounds were observed to significantly affect
the growth of *P. aeruginosa* (data not shown), and so a value lower than that for the DMSO blank on Figure 15 indicates reduced production of pyocyanin; indeed, this was observed for a number of the compounds. Natural product 27 showed a small attenuation; however, the strongest effect was seen for the saturated series of analogues 145-149. This is perhaps not surprising, as the side chain for these compounds bears close similarity to the heptyl chain present in the native ligand PQS. Whilst these preliminary results are intriguing, further repetition of these experiments will be carried out, along with exploration of further assays to bolster the hypothesis that these compounds are directly modulating quorum sensing. In particular, the use of an *E. coli* reporter strain to measure transcription of the pqsA promoter region has been reported,\(^6^4\) which could give important insights in this regard.

![Figure 15](image)

*Figure 15*: OD\(_{520}\) (absorption corresponding to pyocyanin) normalised by the culture population (measured by OD\(_{600}\)) for cultures grown in the presence of concentrations of 200 μM of natural products 25, 27, 29-32 and analogues 145-154 after 8 hours. WT = Wild type, no treatment added. DMSO = Treated with DMSO blank. Experiments carried out by Suzie Forrest. The experiment for 31 was not performed due to insufficient material.

Given the Spring Group's earlier observation that natural products 25-28 are able to slow the growth of *E. coli* and *S. aureus*,\(^8^2\) it was wished to explore whether the new compounds exhibited similar effects. These experiments were also carried out by Suzie Forrest, in which the colonies of the species under investigation were grown in the presence of a 200 μM concentration of each compound, and the population was monitored over time, as measured by OD\(_{600}\). As a negative control, a colony was treated with a DMSO blank, and as a positive control, a colony was treated with the antibiotic gentamicin. The results are shown in Figures 16-18, with a curve lying below that for the negative control indicating slowed growth of the bacterium under investigation.

The results for the natural products are shown in Figure 16. The effects associated with 25 and 27 were consistent with that reported previously, although the increase in population at later times was less marked in this case (it was noted that in the report by Salvaggio *et al.*,\(^8^2\)...
the biological data for 27 and 28 were erroneously switched. This will be corrected in a forthcoming publication). The other natural products resulted in slowed growth to varying degrees, but in none of the cases was bacteriostatic behaviour observed. Intriguingly, the compounds which were most active appeared to vary between the bacteria, indicating some selectivity in the mechanism of action which is at work.
Figure 16A: Growth of E. coli in the presence of 200 μM of natural products 25, 27, 29-32. B: Growth of S. aureus in the presence of 200 μM of natural products 25, 27, 29-32. Neg = Negative control (DMSO blank), Pos = Positive Control (Gentamicin). Experiments carried out in triplicate, with the exception of 31, for which there was insufficient material for repeats. Error bars refer to standard deviation. Experiments carried out by Suzie Forrest.

Moving on to the shortened analogues 145-149, these showed no effect on the growth of E. coli (Figure 17). However, they were observed to exhibit a profound effect on S. aureus, with some of the compounds showing comparable activity to the positive control. Considering the structures of the most potent compounds 145-147, these all possess small alkyl nitrogen substituents, with larger aromatic substituents resulting in reduced activity (148 and 149). This SAR insight should prove valuable in the future design of more potent compounds.
Figure 17A: Growth of *E. coli* in the presence of 200 μM of saturated analogues 145-149. B: Growth of *S. aureus* in the presence of 200 μM of natural products 145-149. Neg = Negative control (DMSO blank), Pos = Positive Control (Gentamicin). Experiments carried out in triplicate. Error bars refer to standard deviation. Experiments carried out by Suzie Forrest.

Finally, considering the data for the truncated analogues 150, 152 & 153 (Figure 18), the effects are much diminished, with only slightly slowed growth in the case of *S. aureus* (due to insufficient material, the experiments could not be performed for 151 & 154). In particular, 153 has an identical quinolone core to natural product 28, and comparison to the data for this compound further highlights the deleterious effects of the shortened side chain.83
These preliminary data suggest that the class of compounds under investigation possess some intriguing biological activities. Reduced production of the toxin pyocyanin in \( P. \) \textit{aeruginosa} was identified, hinting at modulation of quorum sensing, and secondly, slowed growth of \( E. \) \textit{coli} and \( S. \) \textit{aureus} was shown. In both cases, the saturated analogues 145-149 showed the most promising activity, with their effect on the growth of \( S. \) \textit{aureus} being comparable to the positive control. Following further characterisation of these effects, the chemistry discussed above could be used to produce a second generation of analogues, which will add to knowledge of the SAR and facilitate optimisation of the potency of these compounds.
Section II

Studies Towards the Total Synthesis of 1β-Hydroxyalantolactone
4.0 Introduction

4.1 Terpenes

Terpene natural products have long commanded the attention of synthetic chemists. Their seemingly simple biosynthetic origins belie their resulting complexity; as first noted by Ruzicka, the core scaffolds of terpenes are constructed of varying numbers of isoprene (155) units. Nature uses two building blocks to assemble these units together: dimethylallyl diphosphate (DMAPP, 156) and isopentenyl diphosphate (IPP, 158) (Scheme 50). This synthesis commences with elimination of pyrophosphate from DMAPP to give a stabilised carbocation 157. This then engages IPP to give another carbocation, which loses a proton to give geranyl diphosphate (GPP, 159). This process may be repeated multiple times to give linear chains of increasing length, starting with farnesyl diphosphate (FPP, 160). These compounds may then undergo a broad range of cation-induced cyclisation reactions catalysed by terpene cyclase enzymes. The resultant products are classified according their linear precursor, with GPP giving monoterpenes, FPP giving sesquiterpenes, etc.

It is the diversity in the manner with which these linear precursors may cyclise which gives rise to the great variety of scaffolds present in natural products. Subsequent rearrangement and oxidation reactions add an additional layer of complexity. Selected examples of the structures that may arise are shown in Figure 19, which also demonstrate the biological activity that these compounds may possess. Ingenol (161) was first isolated from *Euphorbia ingens*, and exhibits both anti-tumour and anti-HIV behaviour. Unusual structural features particular to an individual terpene lead to unique challenges in their total synthesis, and in the case of ingenol, the strained *in,out*-4,4,1 bicyclocundecane core poses a distinct challenge. Nonetheless, a number of synthetic routes have been reported. Secondly, ryanodine 162 binds with high affinity to an important family of ion-channels. Despite its complex, highly oxygenated architecture, its total synthesis has been achieved. Finally,
gibberellic acid (163) is an important plant growth hormone. Its total synthesis by Corey in 1978 represented an early landmark in the field.

In contrast to the more complex members of the class, many of the simpler terpenes are readily commercially available, often as both enantiomers (Figure 20). This has lent them great value as building blocks in the chiral pool, serving as the starting materials for the synthesis of more complex products. For example, Corey’s synthesis of picrotoxinin (168) commences with (−)-carvone (165). The chirality already present in the starting material may be used to induce the desired stereochemistry in the product, thus obviating the need for enantioselective reactions. However, terpenes have also seen use in this field, for example as chiral auxiliaries. Reaction of either enantiomer of α-pinene (166 or 167) with borane results in double hydroboration to give diisopinocampheylborane (Ipc₂BH, 169). This (and other closely related compounds) may then be used to bring about asymmetric reactions including hydroboration, reduction, allylation and ring-opening of epoxides.

Figure 19: Selected examples of terpene natural products.

Figure 20: Selected commercially available terpenes and their applications.
4.2 Sesquiterpene Lactones

Derived from three isoprene units, the sesquiterpene lactones (STLs) represent a large class of secondary metabolite, with over 5000 structures isolated to date.\textsuperscript{174} STLs are mainly associated with plants of the family Asteraceae, but can also be isolated from a broad range of other angiosperms, as well as some liverworts.\textsuperscript{175,176} The compounds have been reported to play a range of roles in these organisms, including protection from herbivores and microbial attack, and modulation of plant growth.\textsuperscript{177}

Whilst knowledge of the biosynthesis of STLs remains incomplete, the route outlined in Scheme 51 has been proposed.\textsuperscript{178} This commences with FPP (160), which is itself derived as described above in Scheme 50. In the presence of the enzyme germacrene A synthase (GAS), this undergoes elimination of diphosphate and subsequent cyclisation as shown to give (+)-germacrene A (170). The cytochrome P450 germacrene A oxidase (GAO) then carries out three sequential oxidations of the pendant C-12 methyl to supply (+)-germacrene A acid (GAA, 171). Costunolide synthase (COS) then carries out hydroxylation to 172, which may then spontaneously lactonise to give (+)-costunolide 173. Although in the case shown, hydroxylation occurs at C-6, reaction at C-8 may also occur, and with multiple possible stereochemistries. Following a broad range of subsequent cyclisations and oxidations, the diverse range of STLs result.

![Scheme 51: Proposed biosynthesis of STLs.\textsuperscript{178}](image)

Although roughly 30 distinct classes of STL skeletons are known,\textsuperscript{174} the most common are shown in Figure 21. The eudesmanolides possess two fused six-membered rings, the germacranolides a 10-membered macrocycle, and the guaianolides seven- and five-membered rings. Within these classes, variation exists with respect to stereochemistry, oxidation and double bond isomerism or hydrogenation. However, as shown, the vast majority of STLs possess an α-methylene-γ-lactone ring (although examples exist in which
the methylene is reduced, such as artemisinin and santonin,\textsuperscript{179} or is endocyclic, as in the hirsutinolides\textsuperscript{180}).

\begin{figure}
\centering
\includegraphics[width=\textwidth]{figure21.png}
\caption{The most common classes of STL.}
\end{figure}

It is the α-methylene-γ-lactone ring which is proposed to be responsible for the majority of the biological activity which is associated with STLs. This reactive moiety may engage nucleophiles present in biomolecules (such as cysteine residues) in Michael additions, thus alkylating the targets and interfering with their function.\textsuperscript{181} Whilst this inhibition can suffer from low specificity, leading to toxicity and limited therapeutic applications,\textsuperscript{182} selective examples do exist, demonstrating the potential utility of STL in drug development.\textsuperscript{183} Moreover, the clinical approval of Michael-acceptor containing drugs such as afatinib and ibrutinib has validated the covalent modification of proteins as a therapeutic strategy.\textsuperscript{184}

Many STLs have been demonstrated to possess anti-inflammatory and tumour-inhibitory properties.\textsuperscript{185,186} It has been shown that much of this activity may be due to inhibition of the nuclear factor κB (NF-κB), an important transcription factor which regulates the expression of many genes associated with immunity and cell proliferation.\textsuperscript{187} Prior to activation, NF-κB is bound to proteins of the IκB family, which inhibit its transcriptional activity by securing it in the cytoplasm. In response to a range of stimuli, including the cytokine tumour necrosis factor α (TNFα), viral infection and UV radiation, the IκBs become sequentially phosphorylated and ubiquitylated.\textsuperscript{188} This leads to degradation of IκB by the proteasome, releasing NF-κB, which then moves to the nucleus where it carries out its regulatory activity. Multiple mechanisms may operate for the inhibition of NF-κB by STLs: a study focusing on the action of parthenolide suggested the STL inhibited the degradation of IκB, thus preventing the release of NF-κB.\textsuperscript{189} Meanwhile, it has also been suggested that STLs may alkylate NF-κB itself, preventing it from binding to DNA.\textsuperscript{185}

Some STLs display marked biological activity without possessing a reactive α-methylene-γ-lactone. Perhaps the most notable example is the antimalarial artemisinin \textbf{174}, shown in
Figure 22. As shown in blue, the α-methylene of the lactone (a δ-lactone in this case) has been reduced, but nonetheless, the compound is active against strains of the parasite *Plasmodium falciparum* resistant to traditional treatments such as chloroquine. The activity is thought to be a result of the unusual trioxane ring which artemisinin possesses, which is proposed to undergo cleavage in the presence of iron (abundant within the parasite) to give toxic carbon centred radicals such as 175. Artemisinin (and its synthetic derivatives) have exhibited particular medicinal therapeutic value when used in combination with other therapies, and its endoperoxide pharmacophore has inspired the design of further antimalarials such as arterolane 176. Artemisinin therefore stands a clear example of the potential of STLs in shaping drug development.

Figure 22: Structure of artemisinin (174) and its proposed mode of action, alongside the analogue arterolane (176).

4.3 1β-hydroxyalantolactone (IJ-5)

The STL 1β-hydroxyalantolactone (177, also known as IJ-5, Figure 23), was first isolated in 1978 from *Inula helenium*, and has also been detected in other *Inula* species including *I. bratianica* and *I. japonica*. The latter two species have long been utilised in the traditional Chinese medicine “Xuanfuhua” for the treatment of inflammation (particularly respiratory) and digestive disorders. With its two fused six-membered rings, 1β-hydroxyalantolactone belongs to the STL class of eudesmanolides, and also possesses the α-methylene-γ-lactone moiety responsible for much of the biological activity associated with these compounds.

Figure 23: Structure of 1β-hydroxyalantolactone (IJ-5).

A number of studies have demonstrated the anti-inflammatory properties of IJ-5. In 2010, Qin *et al.* performed a phenotypic study measuring the effect of a range of STLs on the immune response of macrophages. On exposure to lipopolysaccharides (LPS), which are found in
bacterial membranes, macrophages normally release nitric oxide (NO) which can damage the bacteria, alongside playing a range of other immunity-related roles.\textsuperscript{199} However, IJ-5 was observed to attenuate NO production with IC\textsubscript{50} = 5.1 μM. This potency was higher than for other structurally related STLs, which was proposed to be due to improved membrane permeability. Furthermore, no toxicity was observed towards the cells at the concentrations under investigation.

As is common for STLs, IJ-5 is proposed to exert its anti-inflammatory effects through disruption of NF-κB signalling. Tumour necrosis factor alpha (TNF-α) is one of the many possible triggers of NF-κB mediated transcription. As discussed above, activation of this pathway occurs by phosphorylation, ubiquitylation and degradation of NF-κB’s inhibitor, IκB. As part of this scheme, TNF-α recruits the ubiquitin-conjugating enzyme UbcH5; however, a 2014 study by Liu \textit{et al.} provided evidence that IJ-5 alkylates an active site cysteine in UbcH5, thus inhibiting its activity.\textsuperscript{200} The researchers also demonstrated the therapeutic benefit of IJ-5 in murine models of hepatitis and arthritis, without the observation of any toxic effects. A later study demonstrated the protective benefits of treatment with IJ-5 against inflammation in a murine model of atopic dermatitis, as well as providing further evidence of NF-κB inhibition.\textsuperscript{18}

In addition to its anti-inflammatory effects, reports have also been made regarding the cytotoxicity of IJ-5 towards numerous cancer cell lines.\textsuperscript{201–203} Given NF-κB’s known role in oncogenesis,\textsuperscript{204} it is likely that IJ-5 acts through a similar mechanism in this case. Inspired by this range of highly promising activity, recent attempts have been made at the optimisation of the properties of IJ-5 through the synthesis of analogues. A pair of closely related studies generated a number of structures through direct derivatisation of IJ-5 itself.\textsuperscript{205,206} It was shown that any modification of the α-methylene-γ-lactone functionality, as in 179, 180, 182 & 183, resulted in complete abolishment of anti-inflammatory and anti-tumour activity, demonstrating the importance of this group. Acylation of the alcohol (178) resulted in slight reductions in both forms of activity, which stands in contrast to the SAR reported for another STL.\textsuperscript{207} Finally, oxidation to ketone 181 resulted in a reduction in anti-inflammatory activity, but increased cytotoxicity, which was proposed on the basis of docking studies to be due to hydrogen bonding with an active site serine. Finally, evidence was provided of low selectivity of the active compounds between normal and cancerous cells, implying that further optimisation will be necessary for successful application in this area.
A 2017 study by Chen et al. sought to optimise on the UbcH5 inhibitory effect which had been discovered for IJ-5. However, given the relatively low availability of IJ-5 from natural sources (reported isolation yields are highly variable, ranging between 3-500 mg/kg of dried *I. japonica*), it was chosen to form derivatives of the structurally related and more readily available α-santonin (184, Figure 25). A total of 73 analogues were accessed, the most potent of which was 185, which highly selectively inhibited UbcH5 with $K_D = 0.283 \, \mu\text{M}$.

IJ-5 is also indirectly associated with another intriguing biological effect through its appearance in the substructure of the related natural product japonicone A (JapA, 186). Examining the structure of JapA reveals it to be a Diels-Alder heterodimer of two STLs: the guaianolide 187 and IJ-5. A recent study showed JapA to be capable of inhibiting the important oncoprotein MDM2 through a previously unknown mode of action. MDM2 is best known in its role as a negative regulator of the tumour suppressing protein p53, which mediates a range of cancer-preventing activities in response to stress signals, including cell-cycle arrest, apoptosis and DNA repair. MDM2 binds to p53 and promotes its degradation through its E3 ubiquitin ligase activity, thus limiting p53 activity in healthy cells. However,
in many cancers, MDM2 is overexpressed, thus enabling the tumour to evade suppression by p53. Furthermore, MDM2 has also been implicated in a number of oncogenic roles independently from its p53 inhibiting role.\textsuperscript{214} Most small molecule inhibitors have targeted the p53-MDM2 interaction,\textsuperscript{215} meaning they will be ineffective against the large proportion of cancers which harbour mutant p53.\textsuperscript{216} However, JapA was shown to induce a range of anticancer effects in an MDM2-dependant manner, regardless of the p53 status of the tumour.\textsuperscript{19} JapA therefore represents an exciting new class of MDM2 inhibitor, and so investigation of the SAR of this compound would be of great value. This could be achieved through the construction of analogues of IJ-5, which following Diels-Alder reaction would give analogues of JapA.

\begin{equation}
\text{JapA, 186} \quad \implies \quad \text{IJ-5, 177} + \text{187}
\end{equation}

**Scheme 52:** MDM2 inhibiting natural product JapA is a Diels-Alder heterodimer of IJ-5 and another STL.

Despite the wealth of biological activity associated with IJ-5, to date no total synthesis of the compound has been reported. Development of such a route would allow access to IJ-5 without the need for unreliable extraction from natural sources, thus facilitating future biological studies. Additionally, the chemistry developed could be used for the construction of analogues inaccessible through direct derivatisation of IJ-5 itself, which could be used to further knowledge of the SAR for IJ-5’s anti-inflammatory and anti-tumour properties. These compounds could also be advanced into JapA analogues, enabling the design of improved MDM2 inhibitors. The development the first total synthesis of IJ-5 was therefore sought, which will be the subject of the following section.
5.0 Results and Discussion

This section discusses work towards the synthesis of IJ-5 (177), which, as outlined in the introduction, possesses promising anti-inflammatory and anti-tumour properties. The section begins with an outline of the proposed retrosynthesis, followed by details of efforts to put this into practice.

5.1 Retrosynthetic analysis of IJ-5

The retrosynthesis of IJ-5 is outlined in Scheme 53. It was thought that the (α,β)-unsaturated carbonyl present would likely be a highly potent Michael-acceptor, and so represented a potential liability in the forward synthesis. It was envisaged that this could be mitigated by leaving its installation until the final step, and so disconnection of the pendant methylene moiety at C-2 led back to the simplified lactone 188. Such a strategy is well preceded in the total synthesis of similar α-methylene-γ-butyrolactone natural products,\textsuperscript{217–219} and in the forward sense is normally achieved by treatment with Eschenmoser's salt.\textsuperscript{220} It was also thought prudent to have the alcohol moiety present protected until the end of the synthesis. Additionally, through choosing a UV-active protecting group (such as p-methoxybenzyl, PMB), there would be the added benefit of simplified analysis of the synthetic intermediates.
It was thought that 188 could be generated from diene 189. Iodolactonisation is a commonly employed method for bringing about similar transformations, although the product of performing this reaction on 189 would possess an unwanted iodine atom at C-11. Whilst this could likely be removed under radical conditions, this would introduce an extra step into the synthesis, and would represent poor atom economy. However, silver(I) triflate catalysed conditions have been reported by Yang et al. which would bring about the desired transformation in a traceless fashion. In a closely related example from Yang et al., formed in excellent yield, with a clear preference for 5-membered ring formation and a syn-orientation of substituents about the ring junction (Scheme 54). This selectivity could be due to kinetic considerations, and should ensure formation of the desired product 188 in the present case. The proposed substrate 189 is complicated by the presence of an additional olefin at Δ⁴,⁵; however it was proposed that the presence of the extra steric bulk of the methyl at C-6 should disfavour reaction at this position.
It was anticipated that the olefin at $\Delta^{11,12}$ could be generated from nitro compound 190 through a Nef reaction, reduction and elimination sequence. The stereochemistry at C-11 would be lost during these reactions, and so is of no consequence at this point. Then, the presence of this electron-withdrawing group enables a Diels-Alder disconnection back to 191. In the forward sense, if the alcohol protecting group at C-9 is sufficiently bulky, nitroethylene 192 should approach the diene from the opposite side to this substituent, ensuring the correct stereochemistry at C-10. Meanwhile, a cis-geometry at $\Delta^{3,4}$ would lead to that required at C-3. Hyperconjugative electron donation from two alkyl substituents at C-5 (versus one substituent at C-3) should lead to an increased electron density at C-3, which should engage the electron deficient $\beta$-carbon of nitroethylene 192, leaving the nitro group at C-11. However, the regioisomeric 12-nitro product could also be converted to the same eliminated product 189, perhaps with the use of a bulky base to disfavour elimination with the hydrogen at C-3.

The cis-$\Delta^{3,4}$ double bond of 191 could then be constructed using Wittig chemistry (Scheme 55A). This would imply a phosphonium ylide of the form 202, in which the ylide lacks direct stabilisation by the carbonyl, and so might reasonably be expected to favour generation of the desired cis-product. However, there is little literature precedent for this exact transformation, so an alternative strategy was also proposed (Scheme 55B). Firstly, aldehyde 193 could undergo homologation to terminal alkyne 203 by either the Corey-Fuchs method or by treatment with the Bestmann-Ohira reagent. Then, this could undergo alkylation by diazoacetate 204 in a mild, copper-catalysed procedure reported in the literature. Partial hydrogenation with Lindlar’s catalyst could then establish the required olefin stereochemistry.

Disconnection of the alcohol protecting group then enables a reconnection onto C-10 to give epoxide 194. In the synthetic direction, E1cB \( \beta \)- to the aldehyde should be very favourable, as this would release the strain from the epoxide. The aldehyde itself could be accessed by oxidation of alcohol 195. At this point, it was hoped that hydrogenation could be used to set up the stereochemistry required for the methyl at C-6, as the olefin precursor 196 would be expected to approach a heterogeneous hydrogenation catalyst with its less hindered face. Whilst the reactive epoxide could prove a liability during this reaction, the use of poisoned catalysts has been reported to allow the reduction of olefins selectively in the presence of epoxides.\(^{230}\)

We then arrive at a key step of the retrosynthesis. Disconnection of the epoxide gives the symmetrical diene 197, which could itself be readily accessed via Birch reduction of commercially available benzoic acid 199.\(^{231}\) It was anticipated that asymmetric epoxidation conditions could be found which could distinguish between the two olefins, whilst simultaneously directing the reaction to the same face of the substrate as the pendant hydroxyl group. Whilst 1,4-cyclohexadienes have been subjected to catalytic desymmetrisations before,\(^{231-233}\) this strategy would represent, to our knowledge, the first implementation of a hydroxyl-directed epoxidation.

A number of approaches have been reported for the asymmetric epoxidation of homoallylic alcohols. Chiral hydroxamic acid ligands have demonstrated themselves to be particularly adept at mediating this transformation, and have been deployed alongside vanadium, molybdenum, zirconium, hafnium and tungsten catalysts.\(^{234,235}\) One such system utilised a hydroxamic acid 207 (itself derived from \( \text{tert} \)-leucine) alongside VO(O\( \text{OiPr} \))\(_3\), with cumene hydroperoxide as a stoichiometric co-oxidant (Scheme 56).\(^{236}\) The methodology was applied to 206 where, like the desired substrate 197, the reaction takes place on a trisubstituted
olefin contained within a six-membered ring. Whilst the yield and enantiomeric excess were moderate, it was proposed that this system would provide a suitable starting point for the optimisation of the desired transformation.

![Scheme 56: Hydroxamic acid ligand based system for asymmetric epoxidation of homoallylic alcohols.](image)

### 5.2 Studies Towards the Total Synthesis of IJ-5

In order to evaluate the proposed desymmetrisation step, it was necessary to access the substrate for this reaction, alcohol 197. The synthesis of 197 commenced from the commercially available benzoic acid 199, which was exposed to Birch reduction conditions of O’Mahoney et al. (Scheme 57). In this strategy, a measured 3.7 equivalents of sodium metal was used as the reducing agent (with two equivalents consumed by the Birch reduction mechanism, and one equivalent consumed though reaction with the carboxylic acid moiety). However, this resulted in a product which \(^1\)H NMR spectroscopic analysis following workup revealed to be a 1:1.14 mixture of starting material 199 and product 198 respectively. The presence of the carboxylic moiety precluded separation by flash column chromatography (even when AcOH was added to the eluent) and attempts at recrystallisation in a range of solvents also proved unsuccessful. Nonetheless, it was decided to continue with this mixture in the hope that separation would be possible at a later stage.

![Scheme 57: Initial Birch reduction attempt.](image)

To this end, direct reduction of the mixture of 198 and 199 was attempted using LiAlH\(_4\) (Scheme 58A). Interestingly, this resulted in the isolation of a sample of 197, contaminated by only small amounts of the aromatic analogue. This observation implies that, under the reaction conditions, only 198 undergoes reduction, whereas 199 remains untouched, before being removed during the basic wash of the workup procedure. A possible explanation for this is shown in Scheme 58B. In the case of 199, the electrophilic end of the carbonyl is presumably held in the same plane as the two methyl substituents as result of the sp\(^2\)
hybridisation of the benzene ring carbons, which sterically hinder the approach of nucleophiles. Meanwhile, in 198, reduction of the aromatic ring pushes the carbon out of the plane of the methyl groups, allowing much better access by nucleophiles. However, the sample obtained of 197 was observed by $^1$H NMR spectroscopy to undergo decomposition overnight to an unidentifiable mixture of products, limiting the utility of this approach. This behaviour stands in contrast to the observed stability of samples of 197 obtained by the alternative approach discussed below, and implies that the decomposition may be mediated by trace impurities present only in this case.

![Scheme 58A](image)

**Scheme 58A:** Reduction of a mixture of 199 and 198 resulted in generation of almost pure 197. B: Proposed rationale for the difference in reactivity between 199 and 198. $^a$Approximate yield based on the calculated amount of 198 in the starting material mixture.

Esterification of the mixture of 199 and 198 was then investigated (Table 9). This was initially attempted through the intermediacy of a trifluoroacetyl mixed anhydride (Table 9, Entry 1). However, $^1$H NMR spectroscopic analysis following workup revealed the presence of 209 only. Next, the transformation was attempted via an S$_{N}$2 reaction with methyl iodide, which gratifyingly resulted in esterification of both species in the mixture, and the products 209 and 210 could be separated using flash column chromatography.
Table 9: Esterification of a mixture of 198 and 199.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>TFA anhydride, MeOH, r.t., 2 h</td>
<td>209 only&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>MeI, K₂CO₃, acetone, reflux, 2 h</td>
<td>Isolated 209 (51%)&lt;sup&gt;b&lt;/sup&gt; and 210 (52%)&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Based on ¹H NMR spectrum following workup. <sup>b</sup>Yield based on calculated amount of relevant precursor in starting material mixture.

However, given the inefficiency of this method for the generation of 210, attention was turned to the optimisation of the Birch reduction protocol. Pleasingly, it was found that treatment of 199 with an excess of sodium metal (rather than the measured 3.7 equivalents used earlier), it was possible to bring about complete conversion to 198, removing the need for further purification (Scheme 59). Additionally, after a minor change of the workup conditions (see experimental section for details), esterification to 210 proceeded with excellent yield on a scale of 8.4 g.

Scheme 59: Optimised synthesis of 210.

With access to pure ester 210 now achieved, its reduction to the desired alcohol 197 was explored (Table 10). An initial trial using LiAlH₄ was promising, and gave 197 with low isolated yield (Table 10, Entry 1). It was suspected that volatility of the product may be contributing to loss of material, and indeed repeating the procedure with an extended reaction time and more cautious concentration under vacuum resulted in a much-improved yield (Table 10, Entry 2). Finally, increasing the number of equivalents of the reducing agent gave the product with excellent yield, even on a relatively large scale (Table 10, Entry 3).

Table 10: Reduction of ester 210 to alcohol 197.
### Entry | Conditions | Isolated yield of 197
--- | --- | ---
1 | LiAlH₄ (1.2 equiv.), Et₂O, 0 °C, 2 h | 39%
2 | LiAlH₄ (1.2 equiv.), Et₂O, 0 °C → r.t., 16 h, cautious evaporation | 73%
3 | LiAlH₄ (1.5 equiv.), Et₂O, 0 °C → r.t., 16 h, 7.4 g scale | 87%

With the substrate 197 for the proposed catalytic desymmetrising epoxidation reaction now in hand, it was necessary to synthesise the required hydroxamic acid ligand 207, which was attempted using the procedure from the publication in which the compound was first described (outlined in Scheme 60). This commenced with protection of tert-leucine 211 using phthalic anhydride 216 (Table 11). When 4Å mol. sieves were used as the dehydrating agent, only a poor yield of product was obtained (Table 11, Entry 1). However, the employment of Dean-Stark conditions gave a more satisfactory result (Table 11, Entry 2).

![Scheme 60](image)

**Scheme 60:** Synthetic route for the synthesis of hydroxamic acid ligand 207.

**Table 11:** Protection of tert-leucine 211

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Isolated yield of 212</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>216, TEA, toluene, 4Å mol. sieves, reflux, 2 h</td>
<td>23%</td>
</tr>
<tr>
<td>2</td>
<td>216, TEA, toluene, Dean-Stark conditions, 2 h</td>
<td>73%</td>
</tr>
</tbody>
</table>
Next, it was necessary to synthesise the hydroxylamine 215 which would undergo coupling with 212 to give the required ligand 207. This commenced with condensation of hydroxylamine with benzophenone 213 to give oxime 214 with excellent yield (Table 12). For the reduction of oximes to hydroxylamines, exposure to sodium cyanoborohydride under acidic conditions is commonly employed. In an effort to avoid the potentially hazardous nature of such a procedure, alternative conditions were sought. Firstly, treatment with sodium borohydride in acetic acid was trialled; however, this resulted in recovery of starting material (Table 4, Entry 1). Then, reduction using pyridine-borane complex was attempted (Table 12, Entry 2). Whilst this did generate the desired product, the conversion was poor, with a large amount of starting material recovered. Given these disappointing results, it was decided to cautiously carry out the sodium cyanoborohydride reduction, which gave the desired hydroxylamine product 215 with excellent yield (Table 12, Entry 3).

Table 12: Synthesis of hydroxylamine 215.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>NaBH₄, AcOH, 0 °C → r.t., 16 h</td>
<td>NRᵃ</td>
</tr>
<tr>
<td>2</td>
<td>Py·BH₃, HCl_{aq}, EtOH, 0 °C, 30 min</td>
<td>30%ᵇ (54% BRSMᵇ)</td>
</tr>
<tr>
<td>3</td>
<td>NaBH₃CN, methyl orange, HCl_{aq}, MeOH, 0 °C, 4 h</td>
<td>81%ᵇ</td>
</tr>
</tbody>
</table>

NR = no reaction.ᵃAs determined by ¹H NMR spectroscopic analysis following workup.ᵇIsolated yield.

With both building blocks of ligand 207 in hand, their combination was then attempted (Table 13). In the original synthesis, this was achieved through activation of 212 as an acid chloride 217 which was treated directly with 215 without isolation. This was first attempted using the reported conditions, which utilised PCl₅ to generate 217 (Table 13, Entry 1). However, only starting material could be isolated, even when the reaction time was extended (Table 13, Entry 2). Given these disappointing results, and that the yield quoted in the original synthesis was a relatively low 34%, more efficacious conditions were sought. By changing the chlorinating agent to oxalyl chloride (Table 13, Entry 3), traces of what were putatively assigned as the desired product and its O-acyl regioisomer were observed. Both compounds showed the expected mass in the LCMS spectrum, and the putative sample of 207 showed ¹H NMR spectrum peaks roughly consistent with those reported in the literature (see appendix), and stained red with FeCl₃ on a TLC plate, indicative of the hydroxamic acid functionality. Meanwhile, the proposed O-acyl regioisomer produced an ¹H NMR spectrum with significant deviations from the literature data (see appendix), and did not stain red with
FeCl₃. However, neither compound was obtained with sufficient purity for full characterisation. Finally, introduction of TEA as a base in the coupling step resulted in the generation of a complex mixture of unidentifiable products, as determined by ¹H NMR spectroscopic and LCMS analysis of the crude product (Table 13, Entry 4).

Table 13: Attempted union of protected amino acid 212 and hydroxylamine 215 to give ligand 207.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions A</th>
<th>Conditions B</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>PCl₅, Et₂O, r.t., 2 h</td>
<td>CH₂Cl₂, -10 °C → r.t., 2 h</td>
<td>NR⁶</td>
</tr>
<tr>
<td>2</td>
<td>PCl₅, Et₂O, r.t., 2 h</td>
<td>CH₂Cl₂, -10 °C → r.t., 16 h</td>
<td>NR⁶</td>
</tr>
<tr>
<td>3</td>
<td>Oxalyl chloride, CH₂Cl₂/DMF, r.t., 5 h</td>
<td>CH₂Cl₂, -10 °C → r.t., 16 h</td>
<td>Putative trace 207 and O-acyl regioisomer⁶</td>
</tr>
<tr>
<td>4</td>
<td>Oxalyl chloride, CH₂Cl₂/DMF, r.t., 5 h</td>
<td>CH₂Cl₂, TEA, -10 °C → r.t., 16 h</td>
<td>Complex mixture⁶</td>
</tr>
</tbody>
</table>

NR = no reaction. ⁶As determined by ¹H NMR and LCMS spectroscopic analysis of the crude product.

It was hypothesised that this apparent difficulty in the formation of 207 may be due to the steric bulk adjacent to the nitrogen which was to be acylated, alongside the presence of a neighbouring less hindered oxygen. The probable generation of the O-acyl isomer above (Table 13, Entry 3) provides evidence for this explanation. A strategy has been reported for the synthesis of hydroxamic acids which utilises transient O-protection to ensure high regioselectivity and yield.²⁴⁹ Applying this methodology to the present case, acid 212 was activated as a mixed anhydride 218 through exposure to iso-butylchloroformate (Scheme 61). Meanwhile, 215 was treated with TMSCl, followed by 218 without isolation of either species. However, ¹H NMR spectroscopic and LCMS analysis following workup revealed a complex mixture from which no product could be obtained.

Scheme 61: Attempted strategy for the synthesis of 207 utilising a transient O-silylation.²⁴⁹
With these difficulties with the synthesis of the ligand 207 in mind, it was decided to continue with the synthetic plan in a racemic fashion, and return to optimisation of the asymmetric step at a later point. Hence, replacement of chiral hydroxamic acid ligand 207 with the simple achiral urea DMPU resulted in epoxidation to give 196 with moderate yield and excellent diastereoselectivity. A NOESY interaction was observed between the protons at positions 2 and 8, which is consistent with the expected stereochemistry. However, given that the perturbation of the orientation of Me-8 is likely to be small upon epoxidation, this NOESY correlation is not definitive, and so the stereochemical data discussed below for derivatives of 196 is vital to provide further evidence for the sense of diastereoselectivity of this reaction.

The hydrogenation of 196 was then explored (Table 14). It was hoped that the steric influence of the neighbouring alcohol substituent would result in hydrogenation of the olefin from the less hindered face to give the desired product 195 as shown. Initially, hydrogenation over palladium on carbon in MeOH was attempted; however, $^1$H NMR analysis following workup showed a complex mixture of products (Table 14, Entry 1). Whilst none of the products obtained were pure enough for full characterisation, on the basis of NMR spectral data it can be tentatively proposed that compounds 220 and 221, which lack the epoxide moiety, were amongst the mixture (see appendix for spectra). In particular, the $^1$H NMR spectrum for 220 appeared to show three peaks corresponding to methyl groups: two singlets at δ = 1.65 and 1.60 ppm, and a doublet at δ = 1.02 ppm. Additionally, only one peak consistent with an O-C-H proton was observed (at δ = 3.90 ppm), and so perhaps hydrogenation of a carbon oxygen bond could account for the extra methyl peak observed. In the $^{13}$C NMR spectrum, quaternary carbons were observed at δ = 128.5 and 125.0 ppm, consistent with both the olefin position shown and the lack of peaks seen in the alkene region of the $^1$H NMR spectrum. Meanwhile, the NMR spectra for 221 showed closer resemblance to 196, with the main differences being the appearance of a singlet integrating to 3H at δ = 3.28 ppm in the $^1$H spectrum, and a resonance at δ = 49.1 ppm in the $^{13}$C NMR spectrum, consistent with methanolation of the epoxide. HMBC between H-10 and C-3 provided evidence for the regiochemistry shown. To avoid formation of these side-products, a range of alternative solvents were screened, none of which offered any improvement (Table 14, Entries 2-4). Next, the use of Pearlman’s catalyst was attempted, and whilst this also

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**Scheme 62**: Hydroxyl-directed epoxidation of homoallylic alcohol 197. An observed NOESY correlation is shown in red.
resulted in a complex mixture of products, there appeared to be a slight preference for the formation of 220.

Table 14: Attempted hydrogenation of epoxide 196, alongside putative side products 220 and 221.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Pd/C, H&lt;sub&gt;2&lt;/sub&gt; (1 atm), MeOH, r.t., 16 h</td>
<td>Complex mixture, consistent with presence of 220 and 221</td>
</tr>
<tr>
<td>2</td>
<td>Pd/C, H&lt;sub&gt;2&lt;/sub&gt; (1 atm), EtOAc, r.t., 16 h</td>
<td>Complex mixture</td>
</tr>
<tr>
<td>3</td>
<td>Pd/C, H&lt;sub&gt;2&lt;/sub&gt; (1 atm), acetone, r.t., 16 h</td>
<td>Complex mixture</td>
</tr>
<tr>
<td>4</td>
<td>Pd/C, H&lt;sub&gt;2&lt;/sub&gt; (1 atm), pet. ether 40-60, r.t., 16 h</td>
<td>Complex mixture</td>
</tr>
<tr>
<td>5</td>
<td>Pd(OH)&lt;sub&gt;2&lt;/sub&gt;/C, H&lt;sub&gt;2&lt;/sub&gt; (1 atm), THF, r.t., 16 h</td>
<td>Complex mixture, 220 major component</td>
</tr>
<tr>
<td>6</td>
<td>Pd/C(en), H&lt;sub&gt;2&lt;/sub&gt; (1 atm), r.t., 24 h</td>
<td>NR</td>
</tr>
<tr>
<td>7</td>
<td>Pd/C(en), H&lt;sub&gt;2&lt;/sub&gt; (5 atm), r.t., 16 h</td>
<td>NR</td>
</tr>
<tr>
<td>8</td>
<td>PADA, AcOH, THF, r.t., 16 h</td>
<td>NR</td>
</tr>
<tr>
<td>9</td>
<td>222&lt;sup&gt;b&lt;/sup&gt;, H&lt;sub&gt;2&lt;/sub&gt;NNH&lt;sub&gt;2&lt;/sub&gt;, MeCN, 0 °C → r.t., 16 h</td>
<td>NR</td>
</tr>
</tbody>
</table>

NR = no reaction. <sup>a</sup>Reactions were monitored by TLC, and <sup>1</sup>H NMR spectroscopy performed after workup. <sup>b</sup>222 = 2-nitrobenzenesulfonyl chloride.

It seemed likely that the reactive epoxide moiety was not compatible with standard hydrogenation conditions, and so a less forcing procedure was sought. The hydrogenation of a number of olefins in the presence of an epoxide has been reported using a palladium on carbon catalyst which had been poisoned with ethylene diamine.<sup>230</sup> This catalyst was synthesised according to the literature procedure (Scheme 63),<sup>250</sup> and its applicability to substrate 196 assessed (Table 14, Entry 6). Unfortunately, only starting material was recovered from the reaction mixture. In the original publication, higher pressures were necessary to ensure good conversion for a trisubstituted olefin example; however, this offered no improvement in the present case (Table 14, Entry 7).

<sup>250</sup>Scheme 63: Synthesis of ethylenediamine poisoned catalyst.
Next, transfer hydrogenation from diimide was attempted. Firstly, the diimide precursor potassium azodicarboxylic acid (PADA) was synthesised according to a literature procedure (Scheme 64).\(^{251}\) This was then used to treat epoxide 196 in combination with AcOH (Table 14, Entry 8);\(^{252}\) however, this once again provoked no reaction. Finally, a one-pot procedure for the generation of diimide from hydrazine was attempted (Table 14, Entry 9),\(^{253}\) but this too was unsuccessful. This lack of reactivity may be due to steric hindrance about the olefin; the reduction of trisubstituted double bonds by diimide is often noted as being sluggish.\(^{254}\)

![Scheme 64: Synthesis of the diimide precursor PADA.\(^{251}\)](image)

It was becoming clear that, in 196, the epoxide was too reactive and the olefin too inert for the desired hydrogenation to take place. As it was desired to open the epoxide later on in the synthesis anyway, this was attempted at this point in order to remove this liability from the molecule. This could be achieved by oxidation of the alcohol in 196 to aldehyde 223, followed by an ElcB type reaction to open the epoxide to give 224 (Table 15). Then, hydrogenation from the more accessible side of the substrate could give 225, with the much more hindered and electron deficient tetrasubstituted olefin remaining intact. In the event, attempted Swern oxidation of 196 gave a complex mixture of products, but treatment with Dess-Martin periodinane gave a crude product whose \(^1\)H NMR spectrum was consistent with formation of 223. However, this was observed to undergo rapid decomposition, and so was exposed to the subsequent elimination conditions without isolation. However, treatment with NaOH (which proved successful on a related substrate in the literature, although this substrate lacked a β,γ-alkene),\(^{255}\) resulted in a complex mixture of products (Table 15, Entry 1). Similarly, exposure to a catalytic quantity of pyrrolidine also resulted in decomposition (Table 15, Entry 2). Clearly, neither 223 nor 224 were sufficiently stable for the desired transformation to be possible.
Table 15: Attempted oxidation of alcohol 196 to aldehyde 223 followed by in situ ring opening to give diene 224, which could undergo selective hydrogenation to 225.

As an alternative strategy, it was proposed to reversibly mask the epoxide as a functionality more stable with respect to the hydrogenation conditions. A potential means of achieving this is outlined in Scheme 65. Treatment of 196 with a Brønsted acid could result in the opening of the epoxide through attack of the conjugate base to give 226 (although the alternative regioisomer resulting from attack at the other end of the epoxide would be equally applicable to this strategy; indeed, a mixture of these regioisomers would be likely). The desired hydrogenation to 227 could then take place. Though the introduction of a bulky halide or pseudohalide substituent X on the bottom face of the ring could lower the diastereoselectivity of the reaction, it was hoped that the influence of the closer pendant alcohol-A would dominate. Then, treatment with base could result in deprotonation of alcohol-B, which could then eliminate the halide or pseudohalide to reform the epoxide moiety, giving the desired product 195.

Scheme 65: Strategy for the reversible masking of the reactive epoxide functionality of 196. X = halide or pseudohalide.

Firstly, a number of conditions were attempted for opening the epoxide with acetic acid to give 228 or its regioisomer 229 (Table 16). The crude product after treatment with acetic acid was revealed by 1H NMR spectroscopy to be a complex mixture (Table 16, Entry 1).
Interestingly, flash column chromatography gave an impure trace of a product whose $^1$H NMR spectrum showed a doublet at $\delta = 1.20$ ppm, likely corresponding to a methyl group, and a single likely alkene multiplet at $\delta = 5.69$ ppm. Furthermore, $^{13}$C NMR spectroscopy showed the presence of a signal at $\delta = 211.1$ ppm, which could be due to the presence of a carbonyl. This could be explained by the acid inducing a Meinwald rearrangement of the epoxide to give 230. Additional evidence for this structure was provided by DEPT, which identified a quaternary alkene carbon at $\delta = 135.3$ ppm and an alkene carbon with a single proton attached at $\delta = 121.8$ ppm, which implied that, intriguingly, the double bond did not move into conjugation with the carbonyl (see appendix for spectra). Carrying out the reaction at a lower temperature with dilution of the acetic acid with CH$_2$Cl$_2$ or Et$_2$O failed to give any improvement (Table 16, Entries 2 & 3). Addition of sodium acetate gave more promising results, and appeared to result in the generation of only one major, unidentified product (Table 16, Entry 4). However, scale up of this procedure once again resulted in the generation of a complex mixture of unidentifiable products (Table 16, Entry 5).

**Table 16: Unsuccessful attempted opening of epoxide 196 with AcOH, alongside putative side-product 230.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>AcOH, r.t. → 50 °C, 3 h</td>
<td>Complex mixture$^a$, possible trace 230$^b$</td>
</tr>
<tr>
<td>2</td>
<td>AcOH / CH$_2$Cl$_2$, r.t., 16 h</td>
<td>Complex mixture$^a$</td>
</tr>
<tr>
<td>3</td>
<td>AcOH / Et$_2$O, r.t., 16 h</td>
<td>Complex mixture$^a$</td>
</tr>
<tr>
<td>4</td>
<td>AcOH, NaOAc, 50 °C, 16 h</td>
<td>Majority single, unidentified species$^a$</td>
</tr>
<tr>
<td>5</td>
<td>Scale-up of Entry 4</td>
<td>Complex mixture$^a$</td>
</tr>
</tbody>
</table>

$^a$As determined by TLC analysis of the reaction mixture, and $^1$H NMR spectroscopy following workup. $^b$As evidenced by $^1$H and $^{13}$C NMR spectroscopic data following flash column chromatography (see main text for details).

Following these difficulties, opening of the epoxide with simple hydrogen halides was attempted. Treatment with HBr in AcOH resulted in a complex mixture of products, as determined by $^1$H NMR spectroscopic analysis following workup (Table 17, Entry 1); however, exposure to HCl allowed isolation of desired product 231 with low yield (alongside a range of uncharacterised side products, Table 17, Entry 2). By lowering the temperature, the yield of 231 was improved, with a significant amount of starting material also isolated (Table 17, Entry 3). The epoxide opens with attack of chloride at the more hindered end of
the epoxide, which is in line with the general trend for such reactions under acidic conditions. This offers greater stabilisation of the positive charge in the transition state by hyperconjugation. In the $^1$H NMR spectrum of the crude product, additional peaks were present which could have been due to the alternative regioisomer resulting from attack at the opposite end of the epoxide (with peak integrals in ratio of 0.25 relative to the product), although this could not be isolated. Key NOESY interactions supporting the relative stereochemistry are shown inset in red in Table 17; in particular, correlation of protons 8 & 4 to only one each of the geminal protons at position 5 provide evidence for trans-diaxial opening of the epoxide. Evidence for the regiochemistry of this reaction was provided by a $^1$H NMR spectrum in d$_6$-DMSO (see appendix), in which the OH peaks could be resolved as a doublet ($J = 5.8$ Hz, indicating coupling to the single proton at the 4-position) at $\delta = 5.41$ ppm, and a triplet ($J = 4.8$ Hz) at 4.80 ppm. Furthermore, treatment of 196 with an excess of acetic anhydride gave a product which, although not purified further following workup, possessed $^1$H NMR spectrum peaks which appeared tentatively to correspond to the structure 231-acetate (see appendix for spectrum). In addition to the expected alkene peak, further downfield resonances were observed at $\delta = 5.18$, 4.49 and 4.42 ppm. These are proposed to correspond to the protons at positions 1 & 4, and provide further evidence that 231 possesses a primary and a secondary alcohol, rather than a primary and a tertiary alcohol, which would have been the case if the epoxide opened with the opposite regioselectivity.

Table 17: Treatment of epoxide 196 with simple hydrogen halides. Key selected NOESY interactions supporting the relative stereochemistry are shown inset in red. The structure of tentatively assigned derivative 231-acetate is also shown, obtained without isolation after treatment of 231 (X = Cl) with acetic anhydride (6.0 equiv.), TEA (6.0 equiv.), DMAP (catalytic), CH$_2$Cl$_2$, r.t., 3 h.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>HBr, AcOH, 10 °C, 30 min</td>
<td>Complex Mixture$^a$</td>
</tr>
<tr>
<td>2</td>
<td>HCl, Et$_2$O/CHCl$_3$, 10 °C, 30 min</td>
<td>X = Cl$^b$, 25%</td>
</tr>
<tr>
<td>3</td>
<td>HCl, Et$_2$O/CHCl$_3$, -15 °C, 3 h</td>
<td>X = Cl$^b$, 54% (78% BRSM$^b$)</td>
</tr>
</tbody>
</table>

$^a$As determined by $^1$H NMR spectroscopic analysis of the crude product. $^b$Isolated yield.

With 231 in hand, its hydrogenation over palladium on carbon was attempted (Scheme 66). However, the only product which could be isolated was the undesired stereoisomer 232, alongside a complex mixture of unidentifiable side products. Selected key NOESY
correlations supporting the relative stereochemistry of 232 are shown inset in Scheme 66.

Interactions between the axial protons at positions 2, 4 & 6 established the syn-configuration of the substituents at positions 2 & 4, supporting the proposed diastereoselectivity in the initial epoxidation to 196. The coupling constants for the peak corresponding to the axial proton at the 6-position (shown inset in Scheme 66) provided evidence for the diastereoselectivity of the hydrogenation. Whilst the multiplet is complicated by slight inequalities in coupling constant and potential overlap with trace impurities, it roughly follows the form of a quartet of doublets with a larger coupling constant of around 13.6 Hz. Such a large value must be due to coupling to the other geminal proton at position 6, and two trans-diaxial protons at positions 5 and 7, which implies the equatorial configuration shown for methyl-9. It therefore appeared that the pendant primary alcohol was not sufficiently bulky to shield the undesired face of the olefin from the heterogeneous catalyst. It was proposed that by derivatising this alcohol with a sterically demanding protecting group, this difficulty could be overcome. Additionally, protection might have the added benefit of reducing the number of side reactions which could take place.

S4eme 66: Hydrogenation of 231. Selected key NOESY correlations supporting the relative stereochemistry of 232, alongside the multiplet observed for the axial proton in position 6.

Protection of the alcohol as a pivalate (Pv) ester 233 seemed an ideal choice, as this would incorporate a bulky t-butyl group. This was attempted using several conditions (Table 18). Treatment with pivaloyl chloride gave a complex mixture, as determined by $^1$H NMR spectroscopic analysis of the crude product (Table 18, Entry 1). Attempted purification resulted in isolation of a sample whose $^1$H NMR spectrum seemed consistent with the desired product, but with the presence of an extra smaller peak beside each peak. It was thought that this could be due to the product existing as a pair of rotamers as a result of the steric influence of the methyl groups; such behaviour has been reported for other bulky esters. However, high temperature NMR spectroscopy revealed that this was unlikely to be the case, as the pairs of peaks failed to coalesce, revealing the sample to be a simple mixture. Attempts to separate the mixture proved unsuccessful, and so alternative conditions were sought. However, addition of triethylamine or pyridine seemed to result in inhibition of the reaction, with recovery of a large amount of starting material (Table 18, Entries 2 & 3).
Table 18: Attempted protection of 196 as a pivalate ester 233.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>PvCl, CH₂Cl₂, 0 °C → r.t., 4 h</td>
<td>Complex mixture(^a)</td>
</tr>
<tr>
<td>2</td>
<td>PvCl, TEA, CH₂Cl₂, 0 °C → r.t., 16 h</td>
<td>Low conversion(^b)</td>
</tr>
<tr>
<td>3</td>
<td>PvCl, pyridine, 0 °C → r.t., 16 h</td>
<td>Low conversion(^b)</td>
</tr>
</tbody>
</table>

\(^a\)As determined by \(^1\)H NMR spectroscopic analysis of the crude product. \(^b\)\(^1\)H NMR spectrum of the crude product showed starting material as the main species, with only trace amounts of other unidentified products.

Next, protection as a tert-butyldimethylsilyl (TBDMS) ether was attempted (Table 19). Gratifyingly, treatment with TBDMSCl and imidazole gave the desired product 234, albeit with only moderate yield. Next, opening of the epoxide with HCl was explored, as this had proved most successful with the unprotected substrate 196. However, this was observed to be more challenging in this case, with \(^1\)H NMR spectroscopy of the crude product showing a significant quantity of 196. (Table 19, Entry 1). TLC analysis indicated that this deprotection was occurring during workup, and so a range of different solutions for quenching the reaction were explored. A pH 7.4 phosphate buffer proved to be optimal, and by also reducing the number of equivalents of HCl, a moderate yield of 235 was achieved (Table 19, Entry 2). Once again, there was a preference for opening of the epoxide at the more substituted end (as evidenced by the high degree of similarity of the \(^1\)H and \(^13\)C NMR spectra between 232 and 235).
Table 19: Protection of 196 as TBDMS ether 234, and subsequent treatment with HCl.

![Diagram](image)

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>HCl (1.2 equiv.), Et₂O/CHCl₃, 0 °C, 1 h</td>
<td>Extensive deprotection</td>
</tr>
<tr>
<td>2</td>
<td>HCl (1.05 equiv.), Et₂O/CHCl₃, 0 °C, 1 h</td>
<td>46% 235</td>
</tr>
</tbody>
</table>

The hydrogenation of 235 was then attempted. The literature suggested that using methanol as the solvent (as had been employed for the hydrogenation of 231 above) would result in deprotection, and so MeCN was used instead. However, the reaction was observed to be very sluggish, with ¹H NMR spectroscopy showing extensive amounts of starting material after workup. Whilst some other unidentified species could also be observed, these were present only in trace amounts, and could not be isolated.

Scheme 67: Attempted hydrogenation of silyl ether 235.

![Diagram](image)

Given this discouraging result, and the difficulties which had been encountered during the synthesis of 235, it was decided to explore the use of the tert-butyldiphenylsilyl (TBDPS) protecting group instead. This is well known to be more robust with respect to acidic conditions than the TBDMS group, which should lead to simplifications in the opening of the epoxide. In the event, protection of 196 using TBDPSCl proceeded with good yield to give 237 (Scheme 68). This was then exposed to the conditions which had proved optimal for the same reaction of the TBDMS ether 234. In this case, alongside the expected major product 238, it was also possible to isolate the minor regiosomer 239, which resulted from opening of the epoxide at the less hindered end. Evidence for the relative stereochemistry of 239 was provided by NOESY correlation, shown inset in red in Scheme 68. Whilst overlap of peaks in the ¹H NMR spectrum precluded full NOESY analysis of 238, the ¹H and ¹³C NMR spectra still closely resemble those of 231, which supports the regio- and stereochemistry shown. Given that 238 and 239 accounted for the vast majority of peaks in the ¹H NMR spectrum of the crude product, the moderate combined yield may be due to losses during purification.
Scheme 68: Protection of 196 as a TBDPS ether 237, and subsequent reaction with HCl to give 238 and 239. Shown inset in red are selected key NOESY correlations providing evidence for the relative stereochemistry of 239.

In exploring the hydrogenation of these species, it was decided to not pursue the reaction of 239. This decision was made as it possesses both a methyl and a chloride substituent on the side of the ring on which reaction was desired, which could lead to poor diastereoselectivity. However, when 238 was treated with the same conditions which had elicited reaction for 231 above, conversion in this case was much slower, with a significant amount of starting material recovered (Table 20, Entry 1). If Pearlman’s catalyst was employed instead, a complex mixture of products resulted (Table 20, Entry 2).

**Table 20: Attempted hydrogenation of 238.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>H₂ (1 atm), Pd/C, THF, r.t., 16 h</td>
<td>Recovered SM</td>
</tr>
<tr>
<td>2</td>
<td>H₂ (1 atm), Pd(OH)₂/C, THF, r.t., 16 h</td>
<td>Complex mixture</td>
</tr>
</tbody>
</table>

Following these discouraging results in the hydrogenation of substrates of the form of 238, and the accompanying difficulty in forming these compounds, it was becoming evident that the strategy outlined in Scheme 65 was not an efficient solution to the problems encountered in the hydrogenation of epoxide 196. As an alternative, it was decided to explore dihydroxylated compounds of the form 241 (Scheme 69). These would lack the strain which was causing problems in the epoxide 196, but the highlighted hydroxyl could still undergo elimination as desired later in the synthesis.
As a proof-of-principal for this strategy, it was first attempted to access 241 in racemic form. This would allow evaluation of the subsequent hydrogenation step, and would also be needed as a standard for determining the e.e. of any future enantioenriched samples of the compound by chiral HPLC analysis. Woodward’s modification of the Prévost reaction is a well-known method for cis-dihydroxylation of olefins (Scheme 70). In contrast to the analogous oxidation with OsO₄, the reaction is observed to occur on the more hindered side of the substrate. This is thought to be a result of a mechanism in which iodine first reacts with the olefin on the more sterically accessible side to give iodonium 243. Both sides of this reactive intermediate are sequentially attacked by acetate to give oxonium 245, with accompanying inversion at each centre. Hydrolysis then yields the product 246.

In order to augment this facial selectivity, and reduce the number of side reactions that could occur during the dihydroxylation, it was decided to first derivatise alcohol 197 with a suitably bulky protecting group. Thus, 197 underwent silylation with TBDPSCI in the presence of imidazole and a catalytic amount of DMAP to give 247 (Scheme 71). This derivative has the extra advantage of being UV-active, which would simplify later analysis by chiral HPLC. However, when exposed to Woodward’s conditions, a complex mixture of products resulted. In particular, some peaks in the aromatic region could be observed by ¹H NMR spectroscopy following workup. Such a species could result from double elimination of water from 248, and so it would appear that the product was not sufficiently stable with respect to the reaction conditions, or could instead perhaps originate from the TBDPS protecting group by iododesilylation.

Scheme 69: Proposed synthesis of an alternative substrate for hydrogenation
Next, dihydroxylation using OsO$_4$ was considered, with the pendant hydroxyl group of 197 acting as a directing group, leading to product 241 (Scheme 69). However, whilst this type of strategy has seen success in the reaction of allylic alcohols, stoichiometric quantities of toxic OsO$_4$ are required. Additionally, extension to homoallylic alcohol substrates (as in the present case) often has deleterious effects on the diastereoselectivity. However, an alternative strategy has been reported which carries out this transformation, consisting of hydroxyl-directed diboration followed by oxidation to give the desired products. As enantioselective diboration conditions have also been reported, it was thought that this strategy could be extended to an enantioselective synthesis of 241. However, in the event, treatment of 197 with the published conditions resulted in the recovery of starting material (Table 21, Entry 1). Whilst elevated temperature and prolonged reaction time did result in the consumption of the starting material, a complex mixture resulted, from which no product could be isolated (Table 21, Entry 2).

**Table 21: Attempted diboration-oxidation of alcohol 197.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B$_2$Pin$_2$, Cs$_2$CO$_3$, MeOH/THF, 70 °C, 3 h, then NaOH, H$_2$O$_2$, H$_2$O, r.t., 4 h</td>
<td>NR$^a$</td>
</tr>
<tr>
<td>2</td>
<td>B$_2$Pin$_2$, Cs$_2$CO$_3$, MeOH/THF, 90 °C (sealed tube) 48 h, then NaOH, H$_2$O$_2$, H$_2$O, r.t., 4 h</td>
<td>Complex mixture$^b$</td>
</tr>
</tbody>
</table>

NR = no reaction. $^a$H NMR spectroscopy following workup showed starting materials. $^b$As determined by 1H NMR spectroscopy following workup.

As the above results revealed difficulties in achieving dihydroxylation on the same face of the substrate as the pendant primary alcohol, reaction on the opposite side of the ring was next explored (Scheme 72). Given that steric effects alone should ensure this diastereoselectivity, the ester 210 already synthesised above could act as a suitable substrate. Whilst the product 249 has the ester substituent blocking the desired face of the olefin for hydrogen, it was predicted that this could be mitigated by reducing this to an alcohol in 250, where it could
then act as a directing group for a homogeneous hydrogenation catalyst. Subsequent oxidation and elimination would then supply the desired aldehyde 253.

Thus, the dihydroxylation of 210 was investigated. It was proposed that Sharpless Asymmetric Dihydroxylation (SAD) would be an ideal method for bringing about the desired transformation in an enantioselective manner. In order to elucidate the e.e. of this reaction by chiral HPLC, it would first be necessary to form a UV active derivative of the product. A racemic standard of such a derivative would also be required, and so 210 was treated with Upjohn dihydroxylation conditions (Scheme 73). The resultant diol 249 was treated directly with benzaldehyde dimethyl acetal without isolation to give the epimers 254 & 255. Whilst a mixture of diastereomers was obtained with respect to the configuration of the phenyl-bearing carbon atom, the d.r. resulting from the dihydroxylation step exceeded 20:1, based on $^1$H NMR spectroscopic analysis of the crude product.

With this racemic sample in hand, the analogous enantioselective reaction was attempted. Applying the mnemonic for the prediction of enantioselectivity, it was concluded that the so-called “β-Mix” of Sharpless asymmetric dihydroxylation reagents (incorporating a dihydroquinidine-derived ligand) should give the desired stereochemistry (Figure 26). Reaction of the desired olefin is predicted to proceed from the less hindered face of the substrate (blue arrow). The remaining olefin should prefer to react by the opposite face, however this would proceed past the pendant ester moiety and so should be disfavoured (red arrow).
Figure 26: Predicted enantioselectivity for dihydroxylation of each olefin in 210 with AD Mix-β. The expected approach on the more distant double bond (red arrow) would be hindered by the pendant ester moiety.

210 was therefore treated with AD-Mix-β, and the resultant diol 249 protected in situ as benzaldehyde acetals 254 and 255 (Scheme 74). It should be noted that the major enantiomer was not determined, and so the absolute configuration shown in the following schemes is intended only to demonstrate relative stereochemistry. In this case, the diastereomers were separated, and it was discovered that 254 was the major product. The relative stereochemistry of these compounds was assigned on the basis of NOESY data, shown in red on Scheme 74, which supported the conclusion that the dihydroxylation had occurred on the opposite side of the ring to the ester functionality on 210. Comparison with the racemic mixture obtained earlier (Scheme 73) indicated that 254 was also the favoured product in that case. This is perhaps due to the avoidance of steric interactions of the phenyl group with the methyl attached to the ring junction.

Scheme 74: Enantioselective dihydroxylation of 210 followed by derivatisation as a UV-active benzaldehyde acetal. Selected key NOESY correlations providing evidence for the relative stereochemistry of 254 and 255 are shown in red.

These samples were then analysed using chiral HPLC (See Experimental for details). Unfortunately, this indicated a disappointing e.e. of only 20%. Whilst poor enantioselectivity has been noted for similar substrates in the literature, it is hoped that with optimisation, this value may be improved through variation of the ligand and temperature of the reaction, and fine-tuning of the substrate structure. In particular, ligands possessing an anthraquinone linker (as opposed to the phthalazine based ligands used in the commercial AD-mixes) have been reported to give superior e.e. when applied to substrates lacking aromatic...
functionality. Alternatively, given that aryl-aryl stacking has been proposed to play a role in positioning the substrate in the catalyst binding pocket, introducing aryl functionality in an analogue of 210 (for example an aryl ester) could also lead to an improved e.e. Nonetheless, it was decided to continue with the proposed synthesis using this slightly enantioenriched material (of which the major enantiomer was not determined) in order to check the feasibility of the later steps.

It was decided to protect the diol 249 as an acetonide, as this would introduce increased steric bulk on top face of the ring, and therefore could improve the diastereoselectivity in the later hydrogenation step. Additionally, the symmetry of the acetal in this case reduces the number of possible diastereomers (cf. 254 & 255 above), simplifying the scheme. Therefore, 210 was dihydroxylated analogously to before, and protected in situ as acetonide 256 with a moderate yield over two steps.

![Scheme 75: Enantioselective dihydroxylation of 210 followed by protection as acetonide 256. Selected key NOESY correlations supporting the relative stereochemistry of 256 are shown in red.](image)

This product was then reduced using LiAlH₄ to give 257 in an excellent yield, allowing exploration of the proposed hydroxyl directed hydrogenation (Table 22). Gratifyingly, hydrogenation in the presence of 20 mol% of Crabtree’s catalyst brought about the desired transformation with moderate yield and selectivity (Entry 1). Whilst extensive peak overlap in the ¹H NMR spectrum precluded full NOESY analysis of 258, the diastereoselectivity may be inferred based on evidence for the stereochemistry obtained for its later derivatives 259 and 260 (see below). Additionally, NOESY data in support for the minor product of this reaction (258-diast) was obtained, which is shown inset in Table 22. Presumably, the pendant primary hydroxyl of 257 is able to coordinate to the electrophilic iridium centre of the catalyst and guide it to the correct face of the substrate. Intriguingly, whilst reducing the mol% of the catalyst necessitated extended reaction time, an improved d.r. was observed. This trend is in accordance with that noted in the literature, and is thought to be due to the existence of catalytically active polynuclear iridium species when the reaction is carried out with a higher mol%. This reactive species is proposed to be able to react without prior coordination by the hydroxyl group, hence lowering the selectivity.
Table 22: Reduction of ester 256 and subsequent hydroxyl-directed hydrogenation with Crabtree’s catalyst. The d.r. refers to the epimer at the asterisked carbon, with the diastereomer shown being the major product. Shown inset in red are selected key NOESY correlations in support of the stereochemistry of the minor product of the hydrogenation reaction. 258-diast.

![Diagram](image)

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Yield &amp; d.r. of 258</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Crabtree’s Catalyst (20 mol%), H₂ (1 atm), CH₂Cl₂, r.t., 3 h</td>
<td>58%&lt;sup&gt;a&lt;/sup&gt;, d.r. = 67:33&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>Crabtree’s Catalyst (1.5 mol%), H₂ (1 atm), CH₂Cl₂, r.t., 16 h</td>
<td>70%&lt;sup&gt;a&lt;/sup&gt;, d.r. = 92:8&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Combined isolated yield for the separated diastereomers. <sup>b</sup>Determined by <sup>1</sup>H NMR spectroscopy of crude product.

With the desired configuration of this key stereocentre now established, attention was turned to the oxidation of the primary alcohol and subsequent deprotection and β-hydroxyl elimination (Table 23). Oxidation using Dess-Martin conditions followed by treatment with acid without isolation resulted in the generation of 260 with low yield (Table 23, Entry 1), in which surprisingly β-OH elimination had not occurred. This may perhaps be due to the unfavourable steric strain which would be present in the resultant tetrasubstituted olefin product. Through isolation of the intermediate 259, a minor improvement in yield was achieved (Table 23, Entry 2). However, by switching to Ley oxidation conditions, a much-improved yield of aldehyde 260 was obtained (Table 23, Entry 3).<sup>271</sup> The relative stereochemistry of 259 and 260 was assigned on the basis of NOESY correlation, shown inset in red on Table 23.
Table 23: Sequential oxidation and deprotection of acetonide 258. Selected key NOESY correlations providing evidence for the relative stereochemistry of 259 and 260 are shown inset in red.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Conditions</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>DMP, NaHCO&lt;sub&gt;3&lt;/sub&gt;, CH&lt;sub&gt;2&lt;/sub&gt;Cl&lt;sub&gt;2&lt;/sub&gt;, 0 °C, 3 h</td>
<td>25% over two steps</td>
</tr>
<tr>
<td>2</td>
<td>DMP, NaHCO&lt;sub&gt;3&lt;/sub&gt;, CH&lt;sub&gt;2&lt;/sub&gt;Cl&lt;sub&gt;2&lt;/sub&gt;, 0 °C, 2 h</td>
<td>46% 259, 76% 260 (35% over two steps)</td>
</tr>
<tr>
<td>3</td>
<td>TPAP (5 mol%), NMO, 4Å mol. sieves, CH&lt;sub&gt;2&lt;/sub&gt;Cl&lt;sub&gt;2&lt;/sub&gt;, r.t., 16 h</td>
<td>76% 259, 81% 260 (62% over two steps)</td>
</tr>
</tbody>
</table>

<sup>a</sup> Performed without isolation of intermediate 259.

In an attempt to induce β-OH elimination of 260 under more forcing conditions, treatment with phosphoric acid was explored (Scheme 76). However, this instead resulted in a complex mixture of products, perhaps resulting from further elimination.

Scheme 76: Attempted β-OH elimination of 260 with H<sub>3</sub>PO<sub>4</sub>

In order to make this challenging reaction more favourable, a strategy for activating the substrate with respect to elimination was sought. Following a literature procedure,<sup>273</sup> diol 260 was converted into carbonate 262 by treatment with triphosgene under basic conditions (Scheme 77). β-elimination of this product would release an equivalent of carbon dioxide gas, making the desired transformation more entropically favourable. This was attempted directly, without isolation of 262, by treatment with triethylamine. The reaction appeared to proceed more slowly than noted in the original publication, presumably due to the increased strain of the product 261 in this case. An impure sample of 261 was obtained; however, this was contaminated with an unidentified impurity which could not be removed by column
chromatography on SiO$_2$. Evidence for the elimination taking place was the appearance of a singlet aldehyde peak at $\delta = 10.15$ ppm, whereas the aldehyde of 260 appeared as a doublet ($J = 3.1$ Hz) at $\delta = 9.79$ ppm (see appendix for spectrum). Further efforts to purify 261 are ongoing.

Scheme 77: $\beta$-elimination of 260 via activated carbonate intermediate 262.
6.0 Conclusions and Future Work

The current synthetic route towards 1β-hydroxyalantolactone 177 is summarised in Scheme 78. The most advanced isolated intermediate, diol 260, encompasses two out of the five required stereocentres, and was accessible in eight steps from commercially available benzoic acid 199, with an overall yield of 20%. Whilst the enantioselectivity in the key desymmetrisation of diene 210 is low, it is hoped that fine-tuning of the substrate structure and reaction conditions will enable improvement of this. Conversely, the diastereoselectivity for this step was high, as was also the case in the hydroxyl-directed hydrogenation of 257.

Following the successful purification of 261, the remainder of the synthetic strategy will be explored in accordance with the retrosynthetic analysis outlined in Scheme 53. Additionally, further optimisation of the synthetic route so far will be attempted. In particular, given that it is likely that carbonate formation will be used to favour β-elimination to give 261, this functionality could be installed earlier. Instead of protecting diol 249 as acetonide 256, carbonate formation at this stage could remove the need for two synthetic steps, giving much improved efficiency.
Scheme 78: Summary of the current synthetic route towards 1β-hydroxyalantolactone 177.
7.0 Experimental

7.1 General Experimental

All non-aqueous reactions were performed under argon using glassware that had been oven-dried overnight. Standard practices were employed when handling moisture- and air-sensitive materials.

Room temperature (r.t.) refers to ambient temperature. All temperatures below 0 °C are that of the external bath. Temperatures of 0 °C were maintained using an ice-water bath. Temperatures of -78 °C were maintained using an acetone-cardice bath. Temperatures of reactions performed in sealed tubes refer to the temperature of the external silicone oil bath.

All reagents and solvents were used as received unless otherwise stated. CH₂Cl₂, EtOAc, MeOH, MeCN and toluene were distilled from CaH₂. Tetrahydrofuran (THF) was dried over Na wire and distilled from a mixture of LiAlH₄ and CaH₂ with triphenylmethane as the indicator. Et₂O was distilled from a mixture of LiAlH₄ and CaH₂. Petroleum ether was distilled before use, with pet. ether 40-60 referring to the fraction between 40-60 °C, and pet. ether 30-40 referring to the fraction between 30-40 °C. The ratios of all solvent mixtures are expressed as volume concentrations (v/v). n-Butyllithium (n-BuLi) in hexanes was titrated with N-benzylbenzamide by the method of Chong et al. before use.²⁷⁴

Yields refer to chromatographically and spectroscopically pure compounds unless otherwise stated. Where possible, reactions were monitored by thin layer chromatography (TLC) and/or liquid chromatography-mass spectrometry (LCMS). TLC analysis was performed on commercially prepared glass plates pre-coated with Merck silica gel F₂₅₄. Visualisation was by the quenching of ultraviolet (UV) fluorescence (λ_max = 254 nm) or by staining with potassium permanganate or vanillin. Retention factors (Rf) are quoted to the nearest 0.01. LCMS analysis was performed on a Waters ACQUITY H-Class UPLC with an ESCi Multi-Mode Ionisation Waters SQ Detector 2 spectrometer using MassLynx 4.1 software; LC system: solvent A: 2 mM NH₄OAc in H₂O/MeCN (95:5); solvent B: MeCN; solvent C: 2% HCO₂H; gradient: A/B/C, 90:5:5-0:95:5 over 1 min at a flow rate of 0.6 mL.min⁻¹.

The naming of compounds and numbering of atoms does not follow IUPAC conventions. With the exception of 2¹², the R/S assignment of stereocentres describes the relative stereochemistry only, and does not indicate absolute configuration. Consistency in the numbering of related structures was sought, and so the numbering shown may not match that used in the compound naming.

Flash column chromatography (FCC) was carried out using either slurry-packed Merck 938 Keiselgel 60 SiO₂ (230-400 mesh) under a positive pressure of dry nitrogen.
Analytical high performance liquid chromatography (HPLC) was run on an Agilent 1260 Infinity machine, using a Supelcosil ABZ+PLUS column (150 mm × 4.6 mm, 3 µm) with a linear gradient system (solvent A: 0.05% (v/v) TFA/H₂O, solvent B: 0.05% (v/v) TFA/MeCN) over 15 min at a flow rate of 1 mL.min⁻¹, and UV detection at 185 (λmax = 220 nm and 254 nm). Retention times (tR) are reported to the nearest 0.01 min. Enantiomeric excess was determined using a chiral HPLC with a Phenomenex Amylose 1 chiral column.

Preparative HPLC was run on an Agilent 1260 Infinity machine, using a Supelcosil ABZ+PLUS column (250 mm × 21.2 mm, 5 µm) with a linear gradient system (solvent A: 0.1% (v/v) TFA/H₂O, solvent B: 0.05% (v/v) TFA/MeCN) over 20 min at a flow rate of 20 mL.min⁻¹, visualised by UV absorbance (λmax = 254 nm).

Infrared (IR) spectra were recorded neat on a Perkin-Elmer Spectrum One (FT-IR) spectrometer with internal referencing. Selected absorption maxima (νmax) are reported in wavenumbers (cm⁻¹) with the following abbreviations: w, weak; m, medium; s, strong.

Melting points (m.p.) were obtained on a Büchi B-545 melting point apparatus and are uncorrected.

Magnetic resonance spectra were processed using ACD/NMR Processor Academic Edition v. 12.01 or TopSpin v. 3.5 (Bruker). An aryl, quaternary, or two or more possible assignments were given when signals could not be distinguished by any means.

Proton magnetic resonance spectra were recorded using an internal deuterium lock (at 298 K unless stated otherwise) on Bruker DPX (400 MHz; 1H-13C DUL probe), Bruker Avance III HD (400 MHz; Smart probe), Bruker Avance III HD (500 MHz; Smart probe) and Bruker Avance III HD (500 MHz; DCH Cryoprobe) spectrometers. Proton assignments are supported by 1H-1H COSY, 1H-13C HSQC or 1H-13C HMBC spectra, or by analogy. Chemical shifts (δ) are quoted in ppm to the nearest 0.01 ppm and are referenced to the residual non-deuterated solvent peak. Discernible coupling constants for coupled protons are reported as measured values in Hertz, rounded to the nearest 0.1 Hz. Data are reported as: chemical shift, number of nuclei, multiplicity (br, broad; s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; or a combination thereof), coupling constants and assignment. Diastereotopic protons are assigned as X and X', where X' designates the lower-field proton.

Carbon magnetic resonance spectra were recorded using an internal deuterium lock (at 298 K unless stated otherwise) on Bruker DPX (101 MHz), Bruker Avance III HD (101 MHz) and Bruker Avance III HD (126 MHz) spectrometers with broadband proton decoupling. Carbon spectra assignments are supported by DEPT editing, 1H-13C HSQC or 1H-13C HMBC spectra, or by analogy. Chemical shifts (δ) are quoted in ppm to the nearest 0.1 ppm and are
referenced to the deuterated solvent peak. Data are reported as: chemical shift, multiplicity (if not a singlet), coupling constants and assignment.

High resolution mass spectrometry (HRMS) measurements were recorded on a Micromass QTOF mass spectrometer or a Waters LCT Premier Time of Flight mass spectrometer. Mass values are quoted within the error limits of ± 5 ppm mass units. ESI refers to the electrospray ionisation technique.

Microanalysis was performed by the University of Cambridge Microanalytical Laboratory in the Department of Chemistry, and is quoted to the nearest 0.1% for all elements except hydrogen, which is quoted to the nearest 0.01%. Reported atomic percentages are within the error limits of ± 0.40%

7.2 Section I Experimental

\((E)-2-(3,7\text{-dimethylocta-2,6-dien-1-yl})-3\text{-methylquinolin-4(1}H\text{-)}\text{-one (25)}\)

![Structural formula of compound 25](image)

To a solution of 45 (263 mg, 1.27 mmol) and 38 (264 mg, 1.06 mmol) in 1,4-dioxane (5 mL) was added a solution of sodium carbonate (2M, 5 mL) and \(\text{Pd(PPh}_3\text{)}_4\) (139 g, 0.12 mmol). The reaction vessel was sealed followed by heating at 120 °C for 2 h. The mixture was filtered through Celite, washing with EtOAc. The organic phase was washed with brine (2 x 10 mL), dried (\(\text{MgSO}_4\)) and concentrated \textit{in vacuo}. The crude material was purified by flash column chromatography (SiO\(_2\), 50% EtOAc) to obtain the desired compound as an off-white solid (141 mg, 0.48 mmol, 45%).

\(\text{R}_f(50\%\text{ EtOAc/pet. ether 40-60}) = 0.17;\) \(^1\text{H NMR (400 MHz, CDCl}_3\): \(\delta = 9.82\) (1H, br s, N-H), 8.36 (1H, d, \(J = 7.8\) Hz, H-5), 7.50 (1H, t, \(J = 7.7\) Hz, H-7), 7.40 (1H, d, \(J = 8.2\) Hz, H-8), 7.25 (1H, t, \(J = 7.5\) Hz, H-6), 5.29 (1H, t, \(J = 7.0\) Hz, H-2'), 5.10-5.04 (1H, m, H-6'), 3.50 (2H, \(d, J = 7.2\) Hz, H-1'), 2.15 (3H, s, H-11), 2.14-2.08 (4H, m, H-4' & H-5'), 1.67 (6H, s, H-8' & H-9'), 1.59 (3H, s, H-10'); \(^{13}\text{C NMR (100 MHz, CDCl}_3\): \(\delta = 177.9\) (C-4), 147.7 (C-2), 141.6 (C-10), 138.8 (C-3'), 132.2 (C-7'), 131.0 (C-7), 126.1 (C-5), 123.8 (C-9), 123.6 (C-6'), 123.1 (C-6), 117.4 (C-8), 117.3 (C-2'), 115.5 (C-3), 39.6 (C-4'), 31.2 (C-1'), 26.4 (C-5'), 25.8 (C-8'), 17.8 (C-10'), 16.4 (C-9'), 10.5 (C-11); \(\nu\) (cm\(^{-1}\)) (neat) = 1637 (m, C=O quinolone), 1607 (w, C=C Ar), 1589 (m, C=C Ar), 1549 (m, C=C Ar), 1492 (s, C=C Ar), 1472 (s, C=C Ar), 1444 (m, C=C Ar), 1357 (s, C=C Ar); \(\text{mp (EtOAc) = 188-189 °C (lit. value: 199-200 °C).}\)
Analytical data consistent with that previously published (slight changes in the NMR data were observed on a sample by sample basis, likely due to minute variations in the amount of water present in the samples). \(^{16}\)

\((E)\)-2-(3,7-dimethylocta-2,6-dien-1-yl)-1,3-dimethylquinolin-4(1\(H\))-one (27)

To a suspension of 25 (25 mg, 0.085 mmol) in THF (0.5 mL) was added LiOt-Bu (14 mg, 0.17 mmol), followed by stirring at r.t. for 20 min. Iodomethane (21 \(\mu\)L, 0.34 mmol) was added at 0 ºC followed by stirring at r.t. for 3 h, at which point LCMS indicated incomplete reaction so a further portion of iodomethane (21 \(\mu\)L, 0.34 mmol) followed by stirring at r.t. for 12 h. Sat. aq. NaHCO\(_3\) (2 mL) was added, followed by extraction with EtOAc (3 x 2 mL). The combined organic extracts were dried (MgSO\(_4\)) and concentrated \textit{in vacuo}. The crude product was purified using flash column chromatography (SiO\(_2\), 40% EtOAc/pet. ether 40-60) to give the desired compound as an off-white semi-solid (16 mg, 0.051 mmol, 60%).

\(R\)\(_f\) (50% EtOAc/pet. ether 40-60) = 0.23; \(^1\)H NMR (500 MHz, CDCl\(_3\)): \(\delta = 8.48\) (1H, dd, \(J = 8.2, 1.8\) Hz, H-5), 7.61 (1H, ddd, \(J = 8.9\) Hz, 7.0, 1.8 Hz, H-7), 7.46 (1H, d, \(J = 8.5\) Hz, H-8) 7.33 (1H, ddd, \(J = 7.9, 6.7, 0.6\) Hz, H-6), 5.08-5.01 (1H, m, H-2'), 3.72 (3H, s, H-11), 3.56 (2H, dd, \(J = 6.1, 0.9\) Hz, H-1'), 2.22 (3H, s, H-12), 2.13-2.04 (4H, m, H-4' & H-5'), 1.78 (3H, d, \(J = 1.2\) Hz, H-9'), 1.65 (3H, d, \(J = 0.9\) Hz, H-8'), 1.58 (3H, s, 10'); \(^{13}\)C NMR (125 MHz, CDCl\(_3\)): \(\delta = 177.1\) (C-4), 150.6 (C-2), 141.1 (C-10), 139.0 (C-3'), 131.9 (C-7'), 131.5 (C-7), 127.0 (C-5), 124.9 (C-9), 123.5 (C-6'), 122.7 (C-6), 118.3 (C-2'), 117.6 (C-3), 115.0 (C-8), 39.4 (C-4'), 34.8 (C-12), 31.0 (C-1'), 26.5 (C-5'), 25.7 (C-8'), 17.7 (C-10'), 16.5 (C-9'), 11.6 (C-11); \(\nu\) (cm\(^{-1}\)) (neat) = 1614 (m, C=O quinolone), 1593 (s, C=C Ar), 1540 (s, C=C Ar), 1499 (m, C=C Ar), 1471 (m, C=C Ar), 1436 (m, C=C Ar).

Analytical data consistent with that previously published. \(^{16}\)

\((E)\)-2-(3-hydroxy-3,7-dimethylocta-1,6-dien-1-yl)-1-methylquinolin-4(1\(H\))-one (30) (First Route)
To a stirred solution of 100 (500 mg, 1.68 mmol), in THF (10 mL) was added LiO-t-Bu (88 mg, 1.10 mmol) followed by stirring at r.t. for 20 min. The mixture was then cooled to 0 °C and iodomethane (0.41 mL, 6.59 mmol) was added followed by stirring at r.t. for 16 h. H$_2$O (10 mL) was added, followed by extraction with EtOAc (3 x 20 mL). The combined organic extracts were dried (MgSO$_4$) and concentrated in vacuo. Purification by flash column chromatography (SiO$_2$, 3% MeOH/CH$_2$Cl$_2$) gave the desired product as a yellow solid (325 mg, 1.05 mmol, 62%).

R$_f$ (3% MeOH/CH$_2$Cl$_2$) = 0.13; $^1$H NMR (400 MHz, CDCl$_3$): δ = 8.42 (1H, dd, J = 8.0, 1.6 Hz, H-5), 7.64 (1H, ddd, J = 8.4, 6.8, 1.6 Hz, H-7), 7.44 (1H, d, J = 8.8 Hz, H-8), 6.72 (1H, d, J = 15.6 Hz, H-12), 6.35 (1H, d, J = 14.8 Hz, H-13), 6.34 (1H, s, H-3), 5.12 (1H, m, H-17), 3.69 (3H, s, H-11), 2.69 (1H, br s, O-H), 2.09 (2H, m, H-16), 1.70 (2H, m, H-15), 1.68 (3H, s, H-21), 1.60 (3H, s, H-19), 1.39 (3H, s, H-20); $^{13}$C NMR (100 MHz, CDCl$_3$): δ = 177.9 (C-4), 152.3 (C-2), 147.0 (C-13), 141.5 (C-9), 132.6 (C-10), 132.2 (C-7), 126.8 (C-18), 126.6 (C-5), 123.9 (C-17), 123.5 (C-6), 121.7 (C-12), 115.4 (C-8), 109.8 (C-3), 73.5 (C-14), 42.1 (C-15), 35.5 (C-11), 28.4 (C-20), 25.7 (C-21), 22.9 (C-16), 17.8 (C-19); ν (cm$^{-1}$) (neat) = 3242 (br, O-H), 1655 (w, C=C), 1618 (s, quinoline ring), 1552 (s, quinoline ring); HRMS: m/z (ES) calculated for C$_{20}$H$_{25}$O$_2$N [M+H]$^+$ : 312.1958, found 312.1951; mp (CH$_2$Cl$_2$, MeOH) = 121-123 °C.

Analytical data consistent with the literature.$^{16}$

(E)-2-(1-hydroxy-3,7-dimethylocta-2,6-dien-1-yl)-1-methylquinolin-4(1H)-one (29) & (E)-2-(3-hydroxy-3,7-dimethylocta-1,6-dien-1-yl)-1-methylquinolin-4(1H)-one (30) (Second Route)

A mixture of 125 (62 mg, 0.17 mmol) and pyridinium tosylate (437 mg, 1.74 mmol) in t-BuOH (3 mL) was stirred at reflux for 48 h. Following cooling to r.t., the mixture was diluted with sat. aq. NaHCO$_3$ (3 mL) and extracted with EtOAc (3 x 2 mL). The combined organics were dried (MgSO$_4$) and concentrated in vacuo. Purification by flash column chromatography (SiO$_2$, 40-100% EtOAc/Pet ether 40-60) gave 29 and 30 as 4:1 mixture respectively (based on $^1$H NMR analysis) (39 mg, 0.13 mmol, 76%). The products were separated by semi-preparative HPLC (5-95% B) to provide 5 and 6 as off-white semi-solids.
Data for (E)-2-(1-hydroxy-3,7-dimethylocta-2,6-dien-1-yl)-1-methylquinolin-4(1H)-one (29):

**HPLC** $t_r = 11.04$ mins (5-95% B); $^1$H NMR (500 MHz, CDCl$_3$): $\delta = 8.27$ (1H, dd, $J = 7.9, 1.5$ Hz, H-5), 7.50 (1H, ddd, $J = 8.9, 7.0, 1.8$ Hz, H-7) 7.33 (1H, ddd, $J = 7.6, 7.0, 0.6$ Hz, H-6), 7.19 (1H, d, $J = 8.6$ Hz, H-8), 6.45 (1H, s, H-3), 5.48-5.42 (2H, m, H-1' & H-2'), 5.06-5.02 (1H, m, H-6'), 3.70 (3H, s, H-11), 2.12-2.05 (4H, m, H-4' & H-5'), 1.72 (3H, d, $J = 0.9$ Hz, H-9'), 1.65 (3H, s, H-8'), 1.56 (3H, s, H-10'); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta = 178.2$ (C-4), 156.0 (C-2), 141.9 (C-10 or C-3'), 141.8 (C-3' or C-10'), 132.1 (C-7), 132.0 (C-7'), 126.0 (C-5), 125.5 (C-9), 123.6 (C-6'), 123.3 (C-2'), 123.2 (C-6), 115.3 (C-8), 110.7 (C-3), 69.8 (C-1'), 39.6 (C-4'), 35.0 (C-11), 26.2 (C-5'), 25.7 (C-8'), 17.7 (C-10'), 17.0 (C-9'); $\nu$ (cm$^{-1}$) (neat) = 3240 (br, O-H), 1618 (C=O), 1597 (C=C Ar), 1499 (m, C=C Ar), 1443 (m, C=C Ar).

Analytical data consistent with that previously published (slight changes in the NMR data were observed on a sample by sample basis, likely due to minute variations in the amount of water present in the samples. Note that H-8', H-9' and H-10' are reassigned relative to the original isolation paper, on the basis of the carbons to which they were observed to undergo HMBC correlation).

Data for (E)-2-(3-hydroxy-3,7-dimethylocta-1,6-dien-1-yl)-1-methylquinolin-4(1H)-one (30):

**HPLC** $t_r = 10.11$ mins (5-95% B); $^1$H NMR (400 MHz, CDCl$_3$): $\delta = 8.42$ (1H, dd, $J = 8.0, 1.6$ Hz, H-5), 7.64 (1H, ddd, $J = 8.4, 6.8, 1.6$ Hz, H-7), 7.44 (1H, d, $J = 8.8$ Hz, H-8), 7.36 (1H, t, $J = 7.4$ Hz, H-6), 6.72 (1H, d, $J = 15.6$ Hz, H-1'), 6.35 (1H, d, $J = 14.8$ Hz, H-2'), 6.34 (1H, s, H-3), 5.12 (1H, m, H-6'), 3.69 (3H, s, H-11), 2.09 (2H, m, H-5'), 1.70 (2H, m, H-4'), 1.68 (3H, s, H-8'), 1.60 (3H, s, H-10'), 1.39 (3H, s, H-9'); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta = 177.9$ (C-4), 152.3 (C-2), 147.0 (C-2'), 141.5 (C-9), 132.5 (C-10), 132.2 (C-7), 126.7 (C-7'), 126.6 (C-5), 123.9 (C-6'), 123.4 (C-6), 121.6 (C-1'), 115.4 (C-8), 109.7 (C-3), 73.4 (C-3'), 42.1 (C-4'), 35.5 (C-11), 28.3 (C-9'), 25.7 (C-10'), 22.9 (C-5'), 17.8 (C-8'); $\nu$ (cm$^{-1}$) (neat) = 3242 (br, O-H), 1655 (w, C=C), 1618 (C=O), 1596 (s, quinolone ring), 1552 (s, quinolone ring).

Analytical data consistent with that previously published.
(E)-2-(3,7-dimethylocta-2,6-dien-1-yl)-3-methyl-1-((methylthio)methyl)quinolin-4(1H)-one (31)

To a suspension of 25 (52 mg, 0.18 mmol) in THF (0.5 mL) was added LiO-t-Bu (1M solution in THF, 0.2 mL, 0.2 mmol). The resulting solution was stirred for 20 min at r.t., followed by addition of chloromethyl methyl sulfide (55 µL, 0.65 mmol) at 0 °C. The mixture was warmed to r.t., followed by stirring for 12 h. H₂O (2 mL) was added, followed by extraction with EtOAc (3 x 1 mL). The combined organic phases were dried (Na₂SO₄) and concentrated in vacuo. The crude mixture was separated using flash column chromatography (SiO₂, 30-50% EtOAc/pet. ether 40-60) to give recovered starting material (20 mg, 0.068 mmol, 37%) and the desired compound as a yellow semi-solid (3.8 mg, 0.011 mmol, 6%).

Rᶠ (40% EtOAc/pet. ether 40-60) = 0.25; ¹H NMR (500 MHz, CDCl₃): δ = 8.47 (1H, dd, J = 7.9, 1.5 Hz, H-5), 7.67-7.60 (2H, m, H-7 & H-8), 7.35 (1H, ddd, J = 7.9, 6.4, 1.5 Hz, H-6), 5.14 (2H, s, H-12), 5.12-5.07 (1H, m, H-2'), 5.07-5.02 (1H, m, H-6'), 3.70 (2H, d, J = 6.1 Hz, H-1'), 2.22 (6H, m, H-11 & H-13), 2.14-2.07 (4H, m, H-4' & H-5'), 1.83 (3H, d, J = 0.9 Hz, H-8'), 1.67 (3H, s, H-10'), 1.60 (3H, s, H-9' [overlaps with water peak]); ¹³C NMR (125 MHz, CDCl₃): δ = 177.4 (C-4), 149.9 (C-2), 140.3 (C-10), 139.5 (C-3'), 131.9 (C-7'), 131.6 (C-7), 127.2 (C-5), 124.9 (C-9), 123.6 (C-6), 123.2 (C-6'), 118.6 (C-2'), 118.2 (C-3), 115.6 (C-8), 49.6 (C-12), 39.4 (C-4'), 30.1 (C-1'), 26.3 (C-5'), 25.7 (C-8'), 17.7 (C-10'), 16.5 (C-9'), 14.5 (C-13), 11.5 (C-11); ν (cm⁻¹) (neat) = 1615 (m, C=O quinolone), 1597 (s, C=C Ar), 1545 (m, C=C Ar). Analytical data consistent with that previously published.¹⁶

To a solution of 27 (23 mg, 0.074 mmol) in CH₂Cl₂ (0.5 mL) was added NaHCO₃ (8 mg, 0.096 mmol). The mixture was cooled to 0 °C and m-CPBA (77% purity, 18 mg, 0.081 mmol) was added, followed by stirring at r.t. for 30 min. H₂O (1 mL) was added and the phases separated. The organic phase was washed with sat. aq. NaHCO₃ (0.5 mL), and then the
combined aqueous phases were extracted with EtOAc (2 x 2 mL). The combined organic phases were dried (Na$_2$SO$_4$) and concentrated in vacuo. The crude product was purified using flash column chromatography (SiO$_2$, 70-80% EtOAc/pet. ether 40-60) to give the desired compound as a colourless semi-solid (10 mg, 0.032 mmol, 43%).

$R_f$ (70% EtOAc/pet. ether 40-60) = 0.21; $^1$H NMR (500 MHz, CDCl$_3$): $\delta$ = 8.48 (1H, dd, $J$ = 7.9, 1.5 Hz, H-5), 7.62 (1H, ddd, $J$ = 8.9, 7.0, 1.8 Hz, H-7), 7.47 (1H, d, $J$ = 8.5 Hz, H-8) 7.33 (1H, ddd, $J$ = 7.9, 7.0, 0.9 Hz, H-6), 5.15-5.11 (1H, m, H-2'), 3.73 (3H, s, H-12), 3.59 (2H, d, $J$ = 6.1 Hz, H-1'), 2.67 (1H, dd, $J$ = 7.0, 5.2 Hz, H-6'), 2.29-2.14 (2H, m, H-4'), 2.22 (3H, s, H-11), 1.82 (3H, d, $J$ = 1.2 Hz, H-9'), 1.72-1.60 (2H, m, H-5'), 1.28 (3H, s, H-10'); $^13$C NMR (125 MHz, CDCl$_3$): $\delta$ = 177.1 (C-4), 150.3 (C-2), 141.1 (C-10), 138.4 (C-3'), 131.5 (C-7), 127.0 (C-5), 124.9 (C-9), 122.8 (C-6), 118.7 (C-2'), 117.4 (C-3'), 115.0 (C-8), 63.9 (C-6'), 58.2 (C-7'), 36.4 (C-4'), 35.0 (C-12), 30.6 (C-1'), 27.4 (C-5'), 24.8 (C-8'), 18.8 (C-10'), 16.6 (C-9'), 11.7 (C-11); v (cm$^{-1}$) (neat) = 1614 (m, C=O quinolone), 1571 (m, C=C Ar), 1571 (m, C=C Ar), 1540 (s, C=C Ar), 1500 (m, C=C Ar), 1471 (m, C=C Ar).

Analytical data consistent with that previously published.$^{16}$

$(E)$-2-(2,6-dimethylhepta-1,5-dien-1-yl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (38)

$^41$ (500 mg, 2.00 mmol), bis(pinacolato)diboron (1.50 g, 5.91 mmol), PdCl$_2$(dpff) (73 mg, 0.10 mmol) and KOAc (392 mg, 4.00 mmol) were added to degassed DMSO (28 mL), and stirred at 50 °C for 12 h. H$_2$O (20 mL) was added, followed by extraction with Et$_2$O (3 x 20 mL). The combined organic phases were dried (MgSO$_4$) and concentrated in vacuo. Purification by flash column chromatography (SiO$_2$, 0-2% Et$_2$O/hexanes) gave the desired product as a colourless oil (334 mg, 1.33 mmol, 67%).

$R_f$ (3% Et$_2$O/pet. ether 40-60) = 0.29; $^1$H NMR (500 MHz, CDCl$_3$): $\delta$ = 5.14-5.09 (2H, m, H-4 & H-9), 2.16-2.08 (4H, m, H-5 & H-6), 1.99 (3H, d, $J$ = 0.7 Hz, H-8), 1.68 (3H, s, H-1), 1.60 (3H, s, H-3), 1.27 (12H, s, H-10'); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ = 162.8 (C-7), 131.7 (C-2), 124.0 (C-4), 82.6 (C-9), 77.2 (C-11 coincides with solvent signal), 42.1 (C-6), 26.4 (C-5), 25.7 (C-1), 24.9 (C-10), 21.3 (C-8), 17.6 (C-3); v (cm$^{-1}$) (neat) = 1638 (m, C=C).

Analytical data consistent with that previously reported.$^{82}$
(E)-1-iodo-2,6-dimethylhepta-1,5-diene (41)

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To a suspension of lithium acetylide ethylene diamine complex (90%, 2.3 g, 22.4 mmol) in DMSO (15 mL) was added dropwise at 10 °C 130 (4.50 g, 21.4 mmol), followed by stirring at r.t. for 2 h. The reaction was then quenched by dropwise addition of H₂O (7.5 mL) at 10 °C. The aqueous phase was extracted with hexanes (4 x 10 mL), and the combined organics were dried (MgSO₄) and cautiously concentrated in vacuo to give crude 40 (2.1 g). A sample of this was submitted directly to the next step (1.58 g, 16.1 mmol assuming previous step quant.)

To a solution of Cl₂ZrCp₂ (4.70 g, 16.4 mmol) in 1,2-dichloroethane (38 mL) was added AlMe₃ (2.0 M in heptane, 16.0 mL, 32.0 mmol). A homogeneous solution resulted after stirring at r.t. for 10 min, at which point crude 40 was added dropwise, followed by stirring at r.t. for 4 h. The mixture was cooled to 0 °C, and then a solution of iodine (4.89 g, 19.8 mmol) in THF (23 mL) was added dropwise, followed by stirring at r.t. for 20 min. The reaction was quenched by dropwise addition of water (30 mL) at 0 °C. The aqueous phase was extracted with Et₂O (3 x 30 mL), and the combined organic phases were washed with 1.0 M aq. sodium thiosulfate (50 mL), water (50 mL), dried (MgSO₄) and concentrated in vacuo. The residue was then suspended in pet. ether 40-60 (50 mL) and eluted through a silica plug, then concentrated in vacuo to yield the desired product as a yellow oil (922 mg, 3.68 mmol, 17%).

R_f (3% Et₂O/pet. ether 40-60) = 0.81; ¹H NMR (500 MHz, CDCl₃): \( \delta = 5.87 \) (1H, m, H-9), 5.06 (1H, m, H-4), 2.23-2.09 (4H, m, H-5 & H-6), 1.84 (3H, d, \( J = 1.1 \) Hz, H-8), 1.69 (3H, d, \( J = 1.1 \) Hz, H-1), 1.60 (3H, s, H-3); ¹³C NMR (125 MHz, CDCl₃): \( \delta = 147.9 \) (C-7), 132.4 (C-2), 123.1 (C-4), 74.8 (C-9), 39.5 (C-6), 26.4 (C-5), 25.7 (C-1), 24.0 (C-8), 17.7 (C-3); ν (cm⁻¹) (neat) = 1617 (w, C=C).

Analytical data consistent with that previously reported.⁸²

Ethyl 3-methyl-4-oxo-1,4-dihydroquinoline-2-carboxylate (42)

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\text{O} \\
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132 (12.2 g, 44.0 mmol) and polyphosphoric acid were stirred at 130 °C for 2 h. The mixture was allowed to cool for 10 min, then poured onto a mixture of ice and sat. aq. NaHCO₃ (100
mL). The resultant precipitate was collected by filtration and rinsed with Et₂O (10 mL) and H₂O (10 mL), to give the desired product as a white solid (4.51 g, 19.5 mmol, 44%).

\[ \text{R} \text{r (10\% MeOH/CH}_2\text{Cl}_2) = 0.58; \text{ } ^1\text{H NMR (500 MHz, CDCl}_3) : \delta = 9.11 (1\text{H, br s, N-H}), 8.35 (1\text{H, dt, } J = 8.1, 0.6 \text{ Hz, H-4}), 7.62 (1\text{H, ddd, } J = 8.4, 7.0, 1.4 \text{ Hz, H-2}), 7.36 (1\text{H, d, } J = 8.8 \text{ Hz, H-1}), 7.32 (1\text{H, ddd, } J = 8.2, 7.1, 1.1 \text{ Hz, H-3}), 4.52 (2\text{H, q, } J = 7.1 \text{ Hz, H-9}), 2.49 (3\text{H, s, H-13}), 3.27 (3\text{H, t, } J = 7.1 \text{ Hz, H-12}); ^{13}\text{C NMR (125 MHz, CDCl}_3) : \delta = 179.8 (\text{C-5}), 164.4 (\text{C-8}), 138.2 (\text{C-7}), 132.7 (\text{C-2}), 126.6 (\text{C-4}), 123.8 (\text{C-3}), 123.7 (\text{C-1}), 122.8 (\text{C-11}), 117.4 (\text{C-6}), 63.3 (\text{C-9}), 14.2 (\text{C-12}), 11.6 (\text{C-13}) (\text{C-10 not visible as quaternary}); \nu (\text{cm}^{-1}) \text{ (neat) = 1717 (s, C=O), 1604 (m, C=O), 1564 \text{ (C=C), 1526 (C=C); mp (H}_2\text{O) = 158-162 } \text{°C).}} \]

Analytical data consistent with that previously published (slight changes in the NMR data were observed on a sample by sample basis, likely due to minute variations in the amount of water present in the samples). 82

2-(hydroxymethyl)-3-methylquinolin-4(1H)-one (43)

\[
\text{To a solution of 42 (650 mg, 2.81 mmol) in THF (50 mL) at 0 } \text{ °C was added LiAlH}_4 (427 mg, 11.3 \text{ mmol) portionwise. The mixture was allowed to warm to r.t. and stirred for 2 h. The reaction was quenched by slow sequential addition at 0 } \text{ °C of EtOAc (10 mL) then MeOH (5 mL), then concentrated in vacuo and dry loaded onto silica. Purification by flash column chromatography (SiO}_2, 10\% \text{ MeOH/CH}_2\text{Cl}_2) \text{ gave the desired product as a white solid (407 mg, 2.15 mmol, 77\%).}} \]

\[ \text{R} \text{r (10\% MeOH/CH}_2\text{Cl}_2) = 0.51; \text{ } ^1\text{H NMR (400 MHz, d}_6\text{-DMSO): } \delta = 11.26 (1\text{H, br s, N-H}), 8.07 (1\text{H, dd, } J = 8.1, 1.5 \text{ Hz, H-4}), 7.84 (1\text{H, d, } J = 8.1 \text{ Hz, H-1}), 7.56 (1\text{H, ddd, } J = 6.9, 8.4, 1.5 \text{ Hz, H-2}), 7.25 (1\text{H, ddd, } J = 7.0, 8.1, 1.1 \text{ Hz, H-3}) 5.95 (1\text{H, t, } J = 5.7 \text{ Hz, O-H}), 4.62 (2\text{H, d, } J = 5.6 \text{ Hz, H-8}), 1.93 (3\text{H, s, H-9}); ^{13}\text{C NMR (100 MHz, d}_6\text{-DMSO): } \delta = 176.3 (\text{C-5}), 148.7 (\text{C-7}), 139.4 (\text{C-10}), 131.2 (\text{C-2}), 125.3 (\text{C-11}), 123.7 (\text{C-4}), 122.9 (\text{C-3}), 118.9 (\text{C-1}), 112.3 (\text{C-6}), 58.9 (\text{C-8}), 9.5 (\text{C-9}); \nu (\text{cm}^{-1}) \text{ (neat) = 3378 (br s, O-H), 1610 (m, C=O), 1564 (m, C=C), 1526 (m, C=C); mp (MeOH) = 252-258 } \text{°C (literature value = 158-159 } \text{°C).}} \]

Analytical data consistent with that previously reported. 82
2-(chloromethyl)-3-methylquinolin-4(1H)-one (45)

To a suspension of 43 (1.28 g, 6.77 mmol) in CH$_2$Cl$_2$ (50 mL) at 0 °C was added dropwise thionyl chloride (5.38 mL, 73.8 mmol). The mixture was stirred at 0 °C for 30 min and concentrated in vacuo. The crude sample was dry loaded onto silica gel and purified by flash column chromatography (SiO$_2$, 1-5% MeOH/CH$_2$Cl$_2$) to give the desired product as a white solid (824 mg, 3.97 mmol, 59%).

$R_f$ (10% MeOH/CH$_2$Cl$_2$) = 0.69; $^1$H NMR (500 MHz, d$_6$-DMSO): $\delta$ = 11.81 (1H, br s, N-H), 8.06 (1H, dd, $J$ = 8.1, 1.2 Hz, H-4), 7.62 (1H, ddd, $J$ = 8.4, 6.8, 1.6 Hz, H-2), 7.52 (1H, d, $J$ = 8.1 Hz, H-2), 7.28 (1H, ddd, $J$ = 8.0, 6.9, 1.1 Hz, H-2), 4.76 (2H, s, H-8), 2.06 (3H, s, H-9); $^{13}$C NMR (125 MHz, d$_6$-DMSO): $\delta$ = 177.1 (C-5), 144.2 (C-7), 139.6 (C-10), 132.2 (C-2), 125.5 (C-4), 123.4 (C-3), 123.4 (C-4), 118.4 (C-11), 116.4 (C-6), 41.7 (C-8), 10.3 (C-9); ν (cm$^{-1}$) (neat) = 1634 (m, C=O), 1553 (s, C=C), 1506 (s, C=C); mp (MeOH) = 234-236 °C (literature value = 277-278 °C).$^{82}$

Analytical data consistent with that previously reported.$^{82}$

Dimethyl 2-(methyl(phenyl)amino)maleate (49)

To a stirred solution of dimethyl acetylenedicarboxylate (6.90 mL, 56.0 mmol) in methanol (10 mL) was added N-methylaniline (5.05 mL, 46.7 mmol) dropwise. The mixture was refluxed at 70 °C for 24 h, cooled, then the solvent was removed in vacuo. The residue was redissolved in CH$_2$Cl$_2$ (100 mL) then washed with sat. aq. NH$_4$Cl (2 x 100mL) and water (2 x 100 mL), dried (MgSO$_4$) and concentrated in vacuo. Purification by flash column chromatography (SiO$_2$, 15% EtOAc/pet. ether 40-60) gave the desired product as an orange semi-solid (10.22 g, 41.0 mmol, 88%).

$R_f$ (20% EtOAc/pet ether 40-60) = 0.39; $^1$H NMR (400 MHz, CDCl$_3$): $\delta$ = 7.33 (2H, t, $J$ = 8 Hz, H-10), 7.24 (1H, t, $J$ = 5 Hz, H-11), 7.17 (2H, d, $J$ = 6 Hz, H-9), 4.77 (1H, s, H-3), 3.66 (3H, s, H-5/6), 3.61 (3H, s, H-5/6), 3.19 (3H, s, H-7); $^{13}$C NMR (100 MHz, CDCl$_3$): $\delta$ = 167.8 (C-1), 165.3 (C-4), 154.2 (C-8), 144.5 (C-2), 129.5 (C-10), 127.4 (C-11), 126.5 (C-9), 88.1 (C-3),
52.6 (C-5), 50.9 (C-6), 40.8 (C-7); ν (cm⁻¹) (neat) = 1725 (s, C=O), 1695 (s, C=O), 1571 (s, phenyl ring).

Analytical data consistent with the literature.²⁷⁵

**Methyl 1-methyl-4-oxo-1,4-dihydroquinoline-2-carboxylate (51)**

![Chemical structure of Methyl 1-methyl-4-oxo-1,4-dihydroquinoline-2-carboxylate (51)]

A mixture of 49 (8.86 g, 36.0 mmol) and polyphosphoric acid (22.5 g) was heated at 130 °C for 3 h, cooled and poured onto iced sat. NaHCO₃ (300 mL). The pH was adjusted to neutral by addition of further sat. NaHCO₃, and the mixture was extracted with EtOAc (3 x 200 mL). The combined organics were washed with brine (300 mL), dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 4% MeOH/CH₂Cl₂) gave the desired product as pink needle crystals (5.68 g, 26.0 mmol, 72%).

Rᵣ(4% MeOH/CH₂Cl₂) = 0.30; ¹H NMR (400 MHz, CDCl₃): δ = 8.49 (1H, dd, J = 7.6, 1.6 Hz, H-5), 7.71 (1H, ddd, J = 8.8, 7.2, 1.6 Hz, H-7), 7.52 (1H, d, J = 8.8 Hz, H-8), 7.39 (1H, t, J = 7.6 Hz, H-6), 6.63 (1H, s, H-3), 3.97 (3H, s, H-13), 3.81 (3H, s, H-11); ¹³C NMR (100 MHz, CDCl₃): δ = 178.1 (C-12), 164.2 (C-4), 143.7 (C-2), 142.0 (C-9), 133.1 (C-7), 127.2 (C-10), 126.7 (C-5), 124.2 (C-6), 116.1 (C-8), 112.6 (C-3), 53.6 (C-13), 37.3 (C-11); ν (cm⁻¹) (neat) = 1730 (s, ester C=O), 1620 (s, quinolone C=O), 1601 (s, quinolone ring), 1573 (s, quinolone ring), 1504 (s, quinolone ring); mp (CH₂Cl₂) = 143-149 °C (Lit. value = 132-134 °C).²⁷⁶

Analytical data consistent with the literature.²⁷⁶

**2-(hydroxymethyl)-1-methylquinolin-4(1H)-one (53)**

![Chemical structure of 2-(hydroxymethyl)-1-methylquinolin-4(1H)-one (53)]

Sodium borohydride (4.83 g, 129 mmol) was added portionwise to a solution of 51 (5.60 g, 25.8 mmol) in 1:1 CH₂Cl₂/MeOH (250 mL) at 0 °C. The mixture was stirred at 0 °C for 3 h, then EtOAc (100 mL) and MeOH (100 mL) were added followed by stirring at r.t. for 30 m. The solvent was removed in vacuo, and purification by flash column chromatography (SiO₂, 5-10% MeOH/CH₂Cl₂) gave the desired product as white needle-like crystals (3.80 g, 20.0 mmol, 78%).
$R_f (10\% \text{ MeOH/CH}_2\text{Cl}_2) = 0.43$; $^1\text{H NMR}$ (400 MHz, $d_6$-DMSO): $\delta = 8.17 \ (1\text{H}, \text{dd}, \text{J} = 8.0, 1.6 \text{ Hz, H-5}), \ 7.78 \ (1\text{H}, \text{d}, \text{J} = 8.0 \text{ Hz, H-8}), \ 7.73 \ (1\text{H}, \text{ddd}, \text{J} = 8.4, 6.8, 1.6 \text{ Hz, H-7}), \ 7.37 \ (1\text{H}, \text{ddd}, \text{J} = 8.0, 6.8, 1.2 \text{ Hz, H-6}), \ 6.23 \ (1\text{H}, \text{s, H-3}), \ 5.76 \ (1\text{H}, \text{t}, \text{J} = 5.8 \text{ Hz, O-H}), \ 4.59 \ (2\text{H}, \text{d}, \text{J} = 5.6 \text{ Hz, H-12}), \ 3.72 \ (3\text{H}, \text{s, H-11})$; $^{13}\text{C NMR}$ (100 MHz, $d_6$-DMSO): $\delta = 176.8 \ (\text{C-4}), \ 154.7 \ (\text{C-2}), \ 142.3 \ (\text{C-9}), \ 132.6 \ (\text{C-8}), \ 126.5 \ (\text{C-10}), \ 125.8 \ (\text{C-5}), \ 123.4 \ (\text{C-6}), \ 117.0 \ (\text{C-7}), \ 108.9 \ (\text{C-3}), \ 61.4 \ (\text{C-12}), \ 34.2 \ (\text{C-11}); \nu (\text{cm}^{-1}) \ (\text{neat}) = 3207 \ (\text{br, O-H}), \ 1623 \ (\text{s, C=O}), \ 1598 \ (\text{s, quinolone ring}), \ 1558 \ (\text{s, quinolone ring}), \ 1504 \ (\text{s, quinolone ring}); \text{HRMS}: \text{m/z} \ (\text{ES}) \ \text{calculated for C}_{11}H_{11}O_2NNa [\text{M+Na}^+] \ : \ 212.0682, \ \text{found} \ 212.0677; \ \text{mp} \ (\text{CH}_2\text{Cl}_2/\text{MeOH}) = 211-212 \degree \text{C} \ (\text{Lit. value} \ 190-192 \degree \text{C}).$\textsuperscript{277}

Analytical data consistent with the literature.\textsuperscript{82}

2-(formyl)-1-methylquinolin-4(1H)-one (62) & 2-(dihydroxymethyl)-1-methylquinolin-4(1H)-one (70)

To a solution of oxalyl chloride (0.049 mL, 0.58 mmol) in CH\textsubscript{2}Cl\textsubscript{2} (3 mL) at -78\degree \text{C was added DMSO (0.3 mL) followed by stirring at 20 m at -78\degree \text{C. A suspension of 53 (100 mg, 0.53 mmol) in 1:1 DMSO/CH}_2\text{Cl}_2 (3mL) was then added, followed by stirring at -78\degree \text{C for 1 h. TEA was then added (0.36 mL, 2.58 mmol) followed by stirring for 30 m. The mixture was then warmed to r.t., followed by removal of the volatile solvents in vacuo and DMSO by subjecting to stream of N\textsubscript{2}. Purification by flash column chromatography (SiO\textsubscript{2}, 4\% MeOH/CH\textsubscript{2}Cl\textsubscript{2} gave a mixture of the aldehyde and its hydrate in a 5:1 ratio respectively as an off-white solid (52.9 mg, 0.28 mmol, ~48\%). Approximate combined yield of 62 and 70 following flash column chromatography, calculated using M\textsubscript{r} for 62 (aldehyde).}

$R_f (5\% \text{ MeOH/CH}_2\text{Cl}_2) = 0.38$;

$^1\text{H NMR}$ (400 MHz, $d_6$-DMSO)

Aldehyde: $\delta = 9.93 \ (1\text{H}, \text{s, H-12}), \ 8.20 \ (1\text{H}, \text{d}, \text{J} = 8.0 \text{ Hz, H-5}), \ 7.89 \ (2\text{H}, \text{m, H-7&8}), \ 7.48 \ (1\text{H}, \text{t}, \text{J} = 6.8 \text{ Hz, H-6}), \ 6.73 \ (1\text{H}, \text{s, H-3}), \ 4.00 \ (3\text{H}, \text{s, H-11})$

Hydrate: $\delta = 8.17 \ (1\text{H}, \text{dd}, \text{J} = 7.8, 1.5, \text{H-5}), \ 7.77 \ (2\text{H}, \text{m, H-7&8}), \ 7.39 \ (1\text{H}, \text{ddd}, \text{J} = 7.8, 6.6, 1.2, \text{H-6}), \ 6.97 \ (2\text{H}, \text{d}, \text{J} = 5.8 \text{ Hz, O-H}), \ 6.36 \ (1\text{H}, \text{s, H-3}), \ 5.90 \ (1\text{H}, \text{t}, \text{J} = 5.8 \text{ Hz, H-12}), \ 3.87 \ (3\text{H}, \text{s, H-11})$

$^{13}\text{C NMR}$ (100 MHz, $d_6$-DMSO)

\textsuperscript{277}
Aldehyde: $\delta = 190.5$ (C-12), 177.4 (C-4), 145.7 (C-2), 142.1 (C-9), 133.6 (C-7), 126.9 (C-10), 125.5 (C-5), 124.3 (C-6), 117.6 (C-8), 116.6 (C-3), 34.9 (C-11).

Hydrate: Data could not be obtained.

$v$ (cm$^{-1}$) (neat) = 3271 (br s, O-H), 1727 (m, aldehyde C=O), 1623 (s, quinolone C=O), 1560 (s, quinoline ring), 1541 (s, quinoline ring), 1501 (s, quinoline ring).

HRMS:  
Aldehyde: m/z (ESI) calculated for C$_{11}$H$_{10}$O$_2$N [M+H]$^+$: 188.0733, found 188.0700  
Hydrate: m/z (ESI) calculated for C$_{11}$H$_{12}$O$_3$N [M+H]$^+$: 206.0812, found 206.0804  

$mp = 192$-$196 \, ^\circ C$

1-methylquinolin-4(1H)-one (72)

To a stirred solution of 4-hydroxyquinoline (500 mg, 3.44 mmol) in DMF (10 mL) at 0 °C was added sodium hydride (60% in mineral oil, 165 mg, 4.13 mmol) portionwise. The mixture was stirred at 0 °C for 15 min, then iodomethane (0.27 mL, 4.33 mmol) was added dropwise, followed by stirring at r.t. for 16 h. H$_2$O (10 mL) was then added, followed by extraction with CH$_2$Cl$_2$ (4 x 10 mL). The combined organic extracts were dried (MgSO$_4$) then concentrated $in$ vacuo. Purification by flash column chromatography (SiO$_2$, 3% MeOH/CH$_2$Cl$_2$) gave the desired product as yellow block-shaped crystals (273 mg, 1.72 mmol, 50%).

$R_f(10\% \text{ MeOH/CH}_2\text{Cl}_2) = 0.45$; $^1$H NMR (500 MHz, CDCl$_3$): $\delta = 8.44$ (1H, dd, $J = 7.5, 1.0$ Hz, H-5), 7.67 (1H, ddd, $J = 8.0, 7.0, 1.0$ Hz, -H7), 7.49 (1H, d, $J = 7.5$ Hz, H-3), 7.38 (2H, m, H-6&8), 6.24 (1H, dd, $J = 7.5, 3.0$ Hz, H-2), 3.78 (3H, d, $J = 1.5$ Hz, H-11); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta = 178.2$ (C-4), 143.6 (C-3), 140.6 (C-9), 132.2 (C-7), 127.0 (C-10), 126.9 (C-5), 123.7 (C-6), 115.3 (C-8), 110.0 (C-2), 40.6 (C-11); $v$ (cm$^{-1}$) (neat) = 1623 (s, C=O), 1575 (s, quinolone ring), 1504 (m, quinolone ring); $mp$ (CH$_2$Cl$_2$) = 150-158 °C (lit. value = 152-153 °C).

Analytical data consistent with the literature.
3,7-dimethyloct-6-en-1-yn-3-ol (74)

To a stirred solution of 6-methyl-5-hepten-2-one (2.00 mL, 13.6 mmol) in THF (40 mL) at -78 °C was added dropwise ethynylmagnesium bromide (0.5 M in THF, 40 mL, 20 mmol). The mixture was warmed to r.t. and stirred for 2 h, then quenched with sat. aq. KHSO₄ (20 mL) and extracted with TBME (4 x 30 mL). The combined organic layers were washed with brine (60 mL), dried (Na₂SO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 10% Et₂O/pet. ether 40-60) gave the desired product as a pale-yellow oil (1.59 g, 10.4 mmol, 76%).

Rᶠ(10% Et₂O/pet ether 40-60) = 0.25; ¹H NMR (500 MHz, CDCl₃) δ = 5.17 (1H, m, H-6), 2.46 (1H, s, H-1), 2.20 (2H, m, H-5), 1.72 (2H, m, H-4), 1.72 (3H, s, H-8), 1.61 (3H, s, H-10), 1.50 (3H, s, H-9); ¹³C NMR (125 MHz, CDCl₃) δ = 132.6 (C-7), 123.6 (C-6), 87.5 (C-2), 71.4 (C-1), 68.3 (C-3), 43.1 (C-4), 29.8 (C-9), 25.7 (C-8), 23.6 (C-5), 17.7 (C-10); v (cm⁻¹) (neat) = 3435 (br, O-H), 3309 (m, C≡C-H).

Analytical data consistent with the literature.¹¹⁰

3,7-dimethylocta-1,6-dien-3-ol (linalool) (86)

To a stirred solution of 6-methyl-5-hepten-2-one (0.50 mL, 3.39 mmol) in THF (10 mL) at -78 °C was added vinylmagnesium bromide (1.0 M in THF, 5.00 mL, 5.00 mmol) dropwise. The mixture was allowed to warm to r.t. then stirred for 1 h. The mixture was then quenched with sat. aq. NH₄Cl (10 mL) and extracted with Et₂O (3 x 10 mL). The combined organic extracts were washed with brine (20 mL), dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 10% Et₂O/pet. ether 40-60) gave the desired product as a pale-yellow oil (170 mg, 1.10 mmol, 32%).

Rᶠ(10% Et₂O/pet ether 40-60) = 0.27; ¹H NMR (500 MHz, CDCl₃) δ = 5.91 (1H, dd, J = 14.0, 11.0 Hz, H-2), 5.21 (1H, dd, J = 14.0, 1.0 Hz, H-1b), 5.12 (1H, m, H-6), 5.05 (1H, dd, J = 11.0, 1.0 Hz, H-1a), 2.02 (2H, m, H-5), 1.68 (3H, s, H-8), 1.60 (3H, s, H-10), 1.57 (2H, m, H-4), 1.28 (3H, s, H-9); ¹³C NMR (125 MHz, CDCl₃) δ = 145.0 (C-2), 132.0 (C-7), 124.3 (C-6), 111.7 (C-1), 73.5 (C-3), 42.0 (C-4), 27.9 (C-9), 25.7 (C-8), 22.8 (C-5), 17.7 (C-10); v (cm⁻¹) (neat) = 3045 (br, O-H), 1639 (w, C≡C-H).
Analytical data consistent with the literature.279

4-(methoxymethoxy)quinolin-2(1H)-one (90)

2,4-quinolinediol (2.00 g, 12.4 mmol) and TBAI (0.44 g, 1.19 mmol) were dissolved in DMF (40 mL). The mixture was cooled to 0 °C, and NaH (60 wt % in mineral oil, 0.52 g, 13.6 mmol) was added portonwise, followed by stirring at r.t. for 10 min. The mixture was returned to 0 °C and chloromethyl methyl ether (1.12 mL, 14.9 mmol) was added dropwise, followed by stirring at r.t. for 16 h. The mixture was diluted with water (200 mL) and extracted with EtOAc (4x 400 mL). The combined organics were washed with sat. aq. NaHCO₃, dried (Na₂SO₄) and concentrated in vacuo to yield the product as yellow needle crystals (2.16 g, 10.5 mmol, 85%).

Rf (5% MeOH/CH₂Cl₂) = 0.32; ¹H NMR (400 MHz, d₆-DMSO): δ = 11.41 (1H, s, N-H), 7.79 (1H, dd, J = 7.6, 1.5 Hz, H-5), 7.51 (1H, ddd, J = 8.5, 7.0, 1.5 Hz, H-7), 7.27 (1H, dd, J = 8.5, 0.5 Hz, H-8), 7.14 (1H, ddd, J = 8.0, 7.0, 1.0 Hz, H-6), 5.93 (1H, s, H-3), 5.38 (2H, s, H-11), 3.44 (3H, s, H-12); ¹³C NMR (100 MHz, d₆-DMSO): δ = 163.5 (C-2), 160.8 (C-4), 139.2 (C-9), 131.5 (C-7), 122.7 (C-5), 121.9 (C-6), 115.7 (C-8), 115.0 (C-10), 99.6 (C-3), 94.7 (C-11), 57.0 (C-12); ν (cm⁻¹) (neat) = 1651 (C=O), 1607 (s, quinoline ring), 1503 (m, quinoline ring);

HRMS: m/z (ESI) calculated for C₁₁H₁₂O₃N [M+H]+: 206.0812, found 206.0805; mp (EtOAc) = 170-173 °C.

4-(methoxymethoxy)quinolin-2-yl trifluoromethanesulfonate (91)

To a solution of 90 (2.02 g, 9.84 mmol) in CH₂Cl₂ (40 mL) was added 2,6-lutidene (2.30 mL, 19.7 mmol). The mixture was cooled to 0 °C and trifluoromethanesulfonic anhydride (2.50 mL, 14.8 mmol) was added dropwise, followed by stirring at r.t. for 1 h. The reaction was then quenched by the addition of 0.1 N aq. HCl (40 mL) and extracted with CH₂Cl₂ (3 x 30 mL). The combined organics were washed with sat. aq. NaHCO₃ (50 mL) and brine (50 mL), dried (Na₂SO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 10% Et₂O/pet. ether 40-60) gave the desired product as a white solid (2.35 g, 6.97 mmol, 71%).
R_f (10% EtO/pet ether 40-60) = 0.30; ^1H NMR (400 MHz, CDCl_3) δ = 8.22 (1H, dd, J = 8.4, 1.2 Hz, H-5), 7.98 (1H, d, J = 8.4 Hz, H-8), 7.79 (1H, ddd, J = 8.4, 7.8, 1.6 Hz, H-6), 7.60 (1H, ddd, J = 8.0, 6.8, 1.2 Hz, H-7), 6.84 (1H, s, H-3), 5.50 (2H, s, H-11), 3.59 (3H, s, H-12); ^13C NMR (100 MHz, CDCl_3): δ = 163.6 (C-4), 154.8 (C-2), 146.2 (C-9), 131.4 (C-6), 128.7 (C-8), 126.8 (C-7), 122.1 (C-5), 121.3 (C-10), 118.7 (q, J_{C:F} = 321 Hz, C-13), 95.1 (C-11/C-3), 95.1 (C-3/C-11), 57.0 (C-12); ν (cm\(^{-1}\)) (neat) = 1586 (s, quinoline ring), 1516 (m, quinoline ring); HRMS: m/z (ES) calculated for C_{12}H_{11}O_5NF_3S [M+H]^+: 338.0305, found 338.0299; mp (CH_2Cl_2) = 62-64 °C.

4-(methoxymethoxy)-2-vinylquinoline (93)

90 (500 mg, 1.48 mmol), vinyl boronic acid pinacol ester (0.25 mL, 1.48 mmol) and Pd(PPh_3)_4 (100 mg, 0.086 mmol) were suspended in 1,4-dioxane (5 mL) and 2M aq. Na_2CO_3 (5 mL). The mixture was refluxed at 100 °C for 16 h, then cooled to r.t. and diluted with EtOAc (30 mL) then filtered through Celite. The filtrate was washed with H_2O, dried (Na_2SO_4) and concentrated _in vacuo_. Purification by flash column chromatography (SiO_2, 30% EtO/pet. ether 40-60) gave the desired product as a pale-yellow oil (246 mg, 1.14 mmol, 77%).

R_f (30% EtO/pet ether 40-60) = 0.28; ^1H NMR (400 MHz, CDCl_3): δ = 8.14 (1H, dd, J = 8.0, 1.2 Hz, H-5), 7.98 (1H, d, J = 8.4, H-8), 7.66 (1H, ddd, J = 8.4, 6.8, 1.6 Hz, H-7), 7.45 (1H, ddd, J = 8.0, 6.8, 1.2 Hz, H-6), 7.19 (1H, s, H-3), 6.96 (1H, dd, J = 17.6, 10.8, H-13), 6.22 (1H, dd, J = 17.6, 0.8 Hz, H-14\(^a\)), 5.61 (1H, dd, J = 10.8, 0.8 Hz, H-14\(^b\)), 5.44 (2H, s, H-11), 3.53 (3H, s, H-12); ^13C NMR (100 MHz, CDCl_3): δ = 160.1 (C-4), 157.2 (C-2), 149.1 (C-9), 138.3 (C-13), 123.0 (C-7), 129.0 (C-8), 125.6 (C-6), 121.6 (C-5), 120.9 (C-10), 119.7 (C-14), 100.0 (C-3), 94.3 (C-11), 56.7 (C-12); ν (cm\(^{-1}\)) (neat) = 1616 (w, C=C), 1590 (s, quinoline ring), 1557 (m, quinoline ring), 1505 (s, quinoline ring); HRMS: m/z (ES) calculated for C_{13}H_{14}O_2N [M+H]^+: 216.1019, found 216.1015.

4-(methoxymethoxy)-2-(3-hydroxy-3,7-dimethyloct-6-en-1-yn-1-yl)quinoline (97)
To a stirred suspension of 91 (664 mg, 1.97 mmol), copper (I) iodide (19 mg, 0.099 mmol), Pd(PPh₃)₄ (114 mg, 0.099 mmol) and potassium carbonate (681 mg, 4.93 mmol) in H₂O (3.5 mL) and 1,2-dimethoxyethane (3.5 mL) was added 74 (750 mg, 4.93 mmol). The mixture was heated to 80 °C and stirred for 16 h, then cooled, diluted with EtOAc (30 mL) and filtered through Celite. H₂O (30 mL) was added and the aqueous layer was extracted with EtOAc (3 x 30 mL). The combined organic extracts were washed with brine (100 mL), dried (Na₂SO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 30% EtOAc/pet. ether 40-60) gave the desired product as an orange oil (595 mg, 1.75 mmol, 89%).

Rᵣ (30% EtOAc/pet ether 40-60) = 0.24; ¹H NMR (500 MHz, CDCl₃): δ = 8.16 (1H, m, H-5), 8.03 (1H, d, J = 8.5, Hz, H-8), 7.69 (1H, ddd, J = 8.5, 7.0, 1.5 Hz, H-7), 7.50 (1H, ddd, J = 8.0, 7.0, 1.0 Hz, H-6), 7.10 (1H, s, H-3), 5.42 (2H, s, H-11), 5.20 (1H, m, H-18), 3.54 (3H, s, H-12), 2.67 (1H, br s, O-H), 2.35 (2H, m, H-17), 1.86 (2H, m, H-16), 1.69 (3H, d, J = 0.5 Hz, H-20), 1.66 (3H, s, H-22), 1.64 (3H, s, H-21); ¹³C NMR (125 MHz, CDCl₃): δ = 159.6 (C-4), 149.1 (C-9), 143.9 (C-2), 132.5 (C-19), 130.3 (C-7), 128.9 (C-8), 126.3 (C-6), 123.8 (C-18), 121.6 (C-5), 120.7 (C-10), 106.1 (C-3), 94.4 (C-11), 93.1 (C-14), 83.8 (C-13), 68.6 (C-15), 56.7 (C-12), 43.2 (C-16), 29.7 (C-21), 25.7 (C-20), 23.7 (C-17), 17.8 (C-22); ν (cm⁻¹) (neat) = 3230 (br, O-H) 2232 (w, C≡C), 1587 (s, quinoline ring), 1556 (m, quinoline ring) 1503 (m, quinoline ring); HRMS: m/z (ES) calculated for C₂₁H₂₆O₃N [M+H]+ : 340.1907, found 340.1902.

2-(3-hydroxy-3,7-dimethyloct-6-en-1-yn-1-yl)quinolin-4(1H)-one (98)

A solution of 97 (100 mg, 0.30 mmol) was stirred in MeOH (10 mL) and 1.5 N aq. HCl (10 mL) for 3.5 h. The mixture was then extracted with Et₂O (4 x 20 mL) and the combined organics were washed with brine (60 mL), dried (MgSO₄) and concentrated in vacuo to give the product as yellow block crystals (81 mg, 0.28 mmol, 93%).

Rᵣ (2% MeOH/CH₂Cl₂) = 0.40; ¹H NMR (500 MHz, d₆-DMSO): δ = 8.09 (1H, dd, J = 8.1, 1.0 Hz, H-5), 7.73 (1H, ddd, J = 8.3, 6.9, 1.4 Hz, H-7), 7.67 (1H, d, J = 7.8 Hz, H-8), 7.40 (1H, ddd, J = 8.1, 7.1, 1.0 Hz, H-6), 6.29 (1H, s, H-3), 5.14 (1H, m, H-16), 2.19 (2H, m, H-15), 1.69 (2H, m, H-14), 1.66 (3H, s, H-18), 1.61 (3H, s, H-20), 1.51 (3H, s, H-19); ¹³C NMR (125 MHz, d₆-DMSO): δ = 176.8 (C-4), 140.7 (C-9), 133.0 (C-2), 132.6 (C-7), 131.6 (C-17), 125.7
(C-5), 125.2 (C-6), 124.3 (C-10/16), 124.0 (C-10/16), 118.6 (C-8), 112.9 (C-3), 100.7 (C-12), 
76.3 (C-11), 67.1 (C-13), 43.7 (C-14), 29.7 (C-19), 25.9 (C-18), 23.5 (C-15), 18.0 (C-20); v (cm⁻¹) (neat) = 3134 (br O-H), 2787 (br, N-H), 2233 (w, C≡C), 1626 (s, C=O), 1585 (s, 
quinoine ring), 1501 (m, quinoline ring);

HRMS: m/z (ES) calculated for C₁₉H₂₂O₂N [M+H]^+: 296.1645, found 296.1639; mp (CH₂Cl₂/MeOH) = 73-79 °C.

(E)-2-(3-hydroxy-3,7-dimethylocta-1,6-dien-1-yl)quinolin-4(1H)-one (100)

To a stirred solution of 98 (270 mg, 0.91 mmol) in THF (40 mL) was added LiAlH₄ (86 mg, 
2.27 mmol) portionwise. The mixture was stirred at r.t. for 16 h, then sat. aq. sodium 
potassium tartrate (40 mL) was added followed by stirring for 20 min. The mixture was 
extracted with EtOAc (3 x 50 mL) and the combined organic extracts were washed with brine 
(100 mL), dried (MgSO₄) and concentrated in vacuo to give the desired product as a yellow 
	solid (230 mg, 0.70 mmol, 77%).

Rf (5% MeOH/CH₂Cl₂) = 0.11; ¹H NMR (400 MHz, DMSO): δ = 11.40 (1H, s, N-H), 8.02 (1H, d, J = 8.0 Hz, H-5), 7.62 (2H, m, H-7&8), 7.28 (1H, m, H-6), 6.83 (1H, d, J = 16.0 Hz, H-12), 
6.44 (1H, d, J = 16.0 Hz, H-11), 6.19 (1H, s, H-3), 5.09 (1H, m, H-16), 4.90 (1H, s, O-H), 1.98 
(2H, m, H-15), 1.62 (3H, s, H-18), 1.55 (2H, m, H-14), 1.54 (3H, s, H-20), 1.12 (3H, s, H-19); 
¹³C NMR (100 MHz, DMSO): δ = 177.4 (C-4), 147.6 (C-2), 146.0 (C-12), 140.6 (C-9), 132.2 
(C-7), 131.0 (C-17), 125.5 (C-10), 125.3 (C-5/16), 125.2 (C-5/16), 123.4 (C-6), 120.9 (C-11), 
118.6 (C-8), 106.8 (C-3), 72.2 (C-13), 42.7 (C-14), 28.5 (C-19), 25.9 (C-18), 22.8 (C-15), 
18.0 (C-20); v (cm⁻¹) (neat) = 3257 (br, O-H), 2921 (br, N-H), 2921 (br, N-H), 1628 (m, C=O), 1586 (s, 
quinoine ring), 1539 (m, quinoline ring); HRMS: m/z (ES) calculated for C₁₉H₂₂O₂N [M+H]^+: 
298.1802, found 298.1796; mp (CH₂Cl₂/MeOH) = 69-76 °C.

(E)-3,7-dimethylocta-2,6-dienal (108)

Following the procedure of Lautens et al.¹³² A slurry of geraniol (3.00 mL, 17.1 mmol) and 
MnO₂ (85% purity, 17.5 g, 171 mmol) in CH₂Cl₂ (15 mL) was stirred at r.t. for 4 days, then 
filtered through Celite, washing with CH₂Cl₂. The filtrate was dried (MgSO₄) and the solvent
was removed in vacuo. The crude product was purified by flash column chromatography (SiO$_2$, 10% Et$_2$O/pet. ether 40-60) to give 5-iodo-2-methylpent-2-ene as a colourless oil (1.89 g, 12.4 mmol, 73%).

$R_f$(20% EtOAc/pet. ether 40-60) = 0.71; $^1$H NMR (500 MHz, CDCl$_3$): $\delta$ = 10.00 (1H, d, $J$ = 8.2 Hz, H-1), 5.90-5.87 (1H, m, H-2), 5.09-5.05 (1H, m, H-6), 2.26-2.19 (4H, m, H-4 & H-5), 2.17 (3H, s, H-9), 1.69 (3H, s, H-8), 1.61 (3H, s, H-10); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ = 191.3 (C-1), 163.9 (C-3), 132.9 (C-7), 127.4 (C-2), 122.5 (C-6), 40.6 (C-4), 25.7 (C-8 or C-5), 25.6 (C-8 or C-5), 17.7 (C-9 or C-10), 17.6 (C-9 or C-10); $\nu$ (cm$^{-1}$) (neat) = 1671 (s, C=O aldehyde).

Analytical data consistent with that previously published.$^{132}$

(E)-5,9-dimethyldeca-4,8-dien-1-yn-3-ol (118)

To a solution of 108 (1.23 g, 8.08 mmol) in THF (40 mL) at -78 °C was added ethynylmagnesium bromide (0.5 M in THF, 32.4 mL, 16.2 mmol) dropwise. The mixture was allowed to return to r.t., then stirred for 2 h. Sat. aq. NH$_4$Cl (20 mL) was added dropwise, followed by extraction with EtOAc (3 x 20 mL). The combined organic extracts were washed with brine (30 mL), dried (MgSO$_4$), then concentrated in vacuo. The crude product was purified by flash column chromatography (SiO$_2$, 20% Et$_2$O/pet. ether 40-60) to give the desired compound as a yellow oil (1.18 g, 6.30 mmol, 78%).

$R_f$(10% Et$_2$O/pet. ether 40-60) = 0.11; $^1$H NMR (500 MHz, CDCl$_3$): $\delta$ = 5.42-5.39 (1H, m, H-4), 5.13-5.09 (2H, m, H-3 & H-8), 2.52 (1H, d, $J$ = 2.1 Hz, H-1), 2.17-2.11 (2H, m, H-7), 2.08-2.05 (2H, m, H-6), 1.08 (1H, d, $J$ = 5.2 Hz, O-H), 1.71 (3H, d, $J$ = 1.2 Hz, H-11), 1.63 (3H, s, H-10), 1.60 (3H, s, H-12); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ = 141.1 (C-5), 132.0 (C-9), 124.0 (C-4), 123.6 (C-8), 84.5 (C-2), 72.5 (C-1), 59.0 (C-3), 39.3 (C-6), 26.2 (C-7), 25.7 (C-10), 17.7 (C-12), 16.6 (C-11); $\nu$ (cm$^{-1}$) (neat) = 3300 (m br, O-H), 3286 (m, C≡C-H), 3171 (s, C≡C-H); HRMS: m/z (ESI) calcd for C$_{12}$H$_{19}$O [M+H]$^+$: 179.1430, found 179.1426.

(E)-(5,9-dimethyldeca-4,8-dien-1-yn-3-yl)oxy)triisopropylsilane (120)
To a solution of 118 (100 mg, 0.56 mmol) and DMAP (129 mg, 1.06 mmol) in CH₂Cl₂ (3 mL) was added TIPSCI (204 mg, 1.06 mmol), followed by stirring for 4 days. Sat. aq. NaHCO₃ (3 mL) was added, the phases separated, and the aqueous phase extracted with Et₂O (3 x 2 mL). The combined organic phases were washed with (3 mL), brine (3 mL), dried (Na₂SO₄) and concentrated in vacuo. The crude product was purified by flash column chromatography (SiO₂, 5% Et₂O/pet. ether 40-60) to give the desired compound as a colourless oil (69.2 mg, 0.21 mmol, 38%).

\[ \text{Rf (10 % Et₂O/pet. ether 40-60) = 0.87; } ^1\text{H NMR (500 MHz, CDCl₃): } \delta = 5.39-5.36 (1H, m, H-4), 5.15 (1H, dd, J = 7.9, 2.1 Hz, H-3), 5.13-5.09 (1H, m, H-8), 2.45 (1H, d, J = 2.4 Hz, H-1), 2.16-2.10 (2H, m, H-7), 2.07-2.03 (2H, m, H-6), 1.70-1.69 (6H, m, H-10 & H-11), 1.62 (3H, s, H-12), 1.16-1.07 (21H, m, H-13 & H-14); ^1³\text{C NMR (125 MHz, CDCl₃): } \delta = 136.7 (C-5), 131.7 (C-9), 126.1 (C-4), 123.8 (C-8), 85.1 (C-2), 71.4 (C-1), 39.1 (C-6), 26.1 (C-7), 25.7 (C-10), 17.9 (C-12 or C-14), 17.8 (C-12 or C-14), 16.6 (C-11), 12.2 (C-13); \nu (cm⁻¹) (neat) = 3311 (w, C≡C-H), 2361 (w, C≡C); HRMS: m/z (ASAP) calcd for C₂₁H₂₉O₃Si [M+H⁺]: 335.2770, found 335.2766. \]

(E)-3-(methoxymethoxy)-5,9-dimethyldeca-4,8-dien-1-yne (121)

To a solution of 118 (1.00 g, 5.61 mmol) and DIPEA (2.9 mL, 16.8 mmol) in CH₂Cl₂ (30 mL) was added MOMCl (0.85 mL, 11.2 mmol), followed by stirring at r.t. for 48 h. Sat. aq. NaHCO₃ (10 mL) was added and the aqueous phase was extracted with Et₂O (3 x 20 mL). The combined organic phases were washed with brine (30 mL), dried (Na₂SO₄) then concentrated in vacuo. The crude product was purified by flash column chromatography (4% Et₂O/pet. ether 40-60) to give the desired compound as a colourless oil (1.07 g, 4.81 mmol, 86%).

\[ \text{Rf (10% Et₂O/pet. ether 40-60) = 0.50; } ^1\text{H NMR (500 MHz, CDCl₃): } \delta = 5.34-5.31 (1H, m, H-4), 5.11-5.07 (2H, m, H-3 & H-8), 4.85 (1H, d, J = 7.0 Hz, H-13°), 4.63 (1H, d, J = 6.7 Hz, H-13°), 3.41 (3H, s, H-14), 2.47 (1H, d, J = 2.1 Hz, H-1), 2.17-2.11 (2H, m, H-6), 2.09-2.05 (2H, m H-7), 1.74 (3H, s, H-14), 1.69 (3H, s, H-10), 1.61 (3H, s, H-12); ^1³\text{C NMR (125 MHz, CDCl₃): } \delta = 141.8 (C-5), 131.9 (C-9), 123.6 (C-8), 121.5 (C-4), 93.1 (C-13), 82.2 (C-2), 73.1 (C-1), 61.5 (C-3), 55.6 (C-14), 39.3 (C-6), 26.2 (C-7), 25.7 (C-10), 17.7 (C-12), 16.6 (C-11); \nu (cm⁻¹) (neat) = 3285 (w, C≡C-H), 2361 (w, C≡C); HRMS: data could not be obtained. \]
A mixture of 2-bromobenzyl chloride (0.15 mL, 1.15 mmol), PdCl$_2$(PPh$_3$)$_2$ (16 mg, 0.022 mmol) and Et$_3$N (0.15 mL, 1.08 mmol) and THF (6 mL) was stirred at r.t. for 10 min. Cul (7 mg, 0.033 mmol) was added, followed by a further 10 min of stirring at r.t. 121 (0.21 mL, 0.92 mmol) was added, followed by stirring for 1 h 45 min. The mixture was then diluted with EtOAc (15 mL), washed with sat. aq. NH$_4$Cl (10 mL), dried (Na$_2$SO$_4$) and concentrated *in vacuo*. The crude product was purified by flash column chromatography (SiO$_2$, 40-60% CH$_2$Cl$_2$/pet. ether 40-60) to give the desired compound as an orange oil (233 mg, 0.57 mmol, 62%).

$\text{Rf (40 \% CH}_2\text{Cl}_2$/pet. ether 40-60) = 0.14; $^1$H NMR (500 MHz, CDCl$_3$): $\delta = 8.03$ (1H, dd, $J = 7.6, 1.8$ Hz, H-6$'$), 7.70 (1H, dd, $J = 7.9, 1.2$ Hz, H-3$'$), 7.45 (1H, td, $J = 7.6, 1.2$ Hz, H-5$'$), 7.39 Hz (1H, td, $J = 7.6, 1.8$ Hz, H-4$'$), 5.39-5.36 (1H, m, H-5), 5.32 (1H, d, $J = 8.9$ Hz, H-4), 5.12-5.08 (1H, m, H-9), 4.87 (1H, d, $J = 7.0$ Hz, H-14$^a$), 4.69 (1H, d, $J = 6.7$ Hz, H-14$^b$), 3.43 (3H, s, H-15), 2.18-2.09 (4H, m, H-7 & H-8), 1.79 (3H, s, H-12), 1.69 (3H, s, H-11), 1.62 (3H, s, H-13); $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta = 176.9$ (C-1), 143.3 (C-6), 136.9 (C-1$'$), 135.0 (C-3$'$), 133.4 (C-4$'$), 133.1 (C-6$'$), 132.1 (C-10), 127.3 (C-5$'$), 123.4 (C-9), 121.2 (C-2$'$), 119.7 (C-5), 93.6 (C-14), 93.2 (C-3), 83.4 (C-2), 61.9 (C-4), 55.8 (C-15), 39.4 (C-7), 26.1 (C-8), 25.7 (C-11), 17.7 (C-13), 16.8 (C-12); $\nu$ (cm$^{-1}$) (neat) = 2206 (C=C), 1655 (s, C=O ketone), 1586 (w, C=C Ar), 1563 (w, C=C Ar); HRMS: $m/z$ (ASAP) calcd for C$_{21}$H$_{26}$O$_3$Br [M+H]$^+$: 405.1065, found 405.1054.

(2Z,5E)-1-(2-bromophenyl)-4-(methoxymethoxy)-6,10-dimethyl-3-(methylamino)undeca-2,5,9-trien-1-one (124)
To a solution 123 (233 mg, 0.57 mmol) in MeOH (1.8 mL) was added MeNH₂ (2.0 M in MeOH, 0.57 mL, 1.15 mmol). The reaction vessel was sealed, followed by heating and stirring at 50 °C for 3 h. The solution was cooled and concentrated in vacuo to give the desired compound as an orange oil (248 mg, 0.57 mmol, 100%).

Rf (50% EtOAc/pet. ether 40-60) = 0.52; ¹H NMR (500 MHz, CDCl₃): δ = 11.05 (1H, d, J = 4.8 Hz, N-H), 7.57 (1H, dd, J = 7.9, 0.9 Hz, H-6'), 7.44 (1H, dd, J = 7.6, 1.5 Hz, H-3'), 7.31 (1H, td, J = 7.5, 1.2 Hz, H-4'), 7.20-7.17 (1H, m, H-5'), 5.65 (1H, s, H-2), 5.24-5.21 (1H, m, H-5), 5.10 (1H, d, J = 9.16 Hz, H-4), 5.08-5.05 (1H, m, H-9), 4.65 (2H, s, H-16), 3.40 (3H, s, H-17), 3.03 (3H, d, J = 5.5 Hz, H-15), 2.17-2.11 (4H, m, H-7 & H-8), 1.82 (3H, d, J = 1.5 Hz, H-13), 1.68 (3H, s, H-11), 1.61 (3H, s, H-14); ¹³C NMR (125 MHz, CDCl₃): δ = 190.7 (C-1), 167.6 (C-3), 143.9 (C-6), 143.4 (C-1'), 133.2 (C-6'), 132.1 (C-10), 129.8 (C-5'), 129.0 (C-3'), 127.0 (C-4'), 123.4 (C-9), 120.4 (C-5), 119.4 (C-2'), 93.4 (C-16), 92.6 (C-2), 69.2 (C-4), 55.7 (C-17), 39.5 (C-7), 29.8 (C-15), 26.1 (C-8), 25.6 (C-11), 17.7 (C-14), 16.7 (C-13); ν (cm⁻¹) (neat) = 2910 (m, N-H), 1605 (s, C=O ketone), 1570 (s, C=C Ar);

HRMS: m/z (ES) calcd for C₂₂H₃₁NO₃[M+H]+: 436.1487, found 436.1491.

(E)-2-(1-(methoxymethoxy)-3,7-dimethylocta-2,6-dien-1-yl)-1-methylquinolin-4(1H)-one (125)

Using the conditions of Wolfe et al. A mixture of 124 (100 mg, 0.23 mmol), Cs₂CO₃ (150 mg, 0.45 mmol), Pd₂dba (10 mg, 0.011 mmol), P(2-furyl)₃ (10 mg, 0.045 mmol) and toluene (2.5 mL) was heated and stirred in a sealed tube at 100 °C for 24 h. The mixture was filtered through Celite, washing with EtOAc (10 mL), and concentrated in vacuo. The crude product was purified using flash column chromatography (SiO₂, 60-70% EtOAc/pet. ether 40-60) to give the desired compound as a brown oil (80 mg, 0.23 mmol, 100%).

Rf (70% EtOAc/pet. ether 40-60) = 0.25; ¹H NMR (400 MHz, CDCl₃): δ = 8.48 (1H, dd, J = 8.2, 1.7 Hz, H-5), 7.71 (1H, ddd, J = 8.5, 7.2, 1.7 Hz, H-7), 7.55 (1H, d, J = 8.5 Hz, H-8), 7.41 (1H, br t, J = 7.5 Hz, H-6), 6.57 (1H, s, H-3), 5.46 (1H, d, J = 8.5 Hz, H-1'), 5.37 (1H, br d, J = 8.9 Hz, H-2'), 5.07-5.02 (1H, m, H-6'), 4.75 (1H, d, J = 6.8 Hz, H-11b), 4.68 (1H, d, J = 6.8 Hz, H-11b), 3.81 (3H, s, H-11), 3.43 (3H, s, H-12'), 2.16-2.12 (4H, m, H-4' & H-5'), 1.83 (3H, s, H-9'), 1.67-1.65 (6H, m, H-8' & H-10'); ¹³C NMR (100 MHz, CDCl₃): δ = 178.4 (C-4), 152.9 (C-2), 143.8 (C-3'), 142.4 (C-10), 132.3 (C-7 or C-7'), 132.2 (C-7 or C-7'), 126.7 (C-5), 126.6 (C-9), 123.5 (C-6 or C-6'), 123.4 (C-6 or C-6'), 121.3 (C-2'), 115.4 (C-8), 110.6 (C-3), 93.8
To a solution of cyclopropyl methyl ketone (10 mL, 100 mmol) in Et₂O (50 mL) was added dropwise methylmagnesium iodide (3.0 M in Et₂O, 50 mL, 150 mmol). The mixture was heated to reflux for 2 h, then cooled and added dropwise to an ice cooled solution of conc. sulfuric acid in water (1:2, 45 mL). The resultant mixture was stirred at r.t. for 30 min, and then the organic phase was collected. The aqueous phase was extracted with Et₂O (2 x 150 mL), and then the combined organic phases were washed with 1.0 M aq. sodium thiosulfate (100 mL), sat. aq. NaHCO₃ (100 mmol), and brine (100 mL), dried (MgSO₄), then concentrated in vacuo to give the desired product as an orange oil (19.3 g, 91.8 mmol, 92%).

5-iodo-2-methylpent-2-ene (130)

![Chemical structure of 5-iodo-2-methylpent-2-ene (130)](image)

Analytical data consistent with that previously reported.²⁸⁰

Diethyl 2-methyl-3-(phenylamino)maleate (132)

![Chemical structure of Diethyl 2-methyl-3-(phenylamino)maleate (132)](image)

A mixture of aniline (5.80 mL, 63.5 mmol), diethyl oxalproprionate (10.0 mL, 53.0 mmol) and glacial acetic acid (26 mL) was stirred at 50 °C for 48 h. Water (50 mL) was then added, and the aqueous phase was extracted with CH₂Cl₂ (3 x 30 mL). The combined organic phases were dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 20-50% CH₂Cl₂/pet. ether 40-60) gave the desired compound as a yellow oil (12.2 g, 43.9 mmol, 83%).

Rf (20% EtOAc/pet. ether 40-60) = 0.56; ¹H NMR (500 MHz, CDCl₃): δ = 10.17 (1H, br s, N-H) 7.22-7.27 (2H, m, H-2), 7.05 (1H, t, J = 7.8 Hz, H-1), 6.99 (2H, d, J = 7.9 Hz, H-3), 4.22
(2H, q, J = 7.3 Hz, H-9), 4.17 (2H, q, J = 7.3 Hz, H-10), 1.85 (3H, s, H-11), 1.32 (3H, t, J = 7.2 Hz, H-12), 1.10 (3H, t, J = 7.2 Hz, H-13); \(^{13}\text{C NMR} \) (125 MHz, CDCl\(_3\)): \(\delta\) = 170.7 (C-8), 164.9 (C-6), 147.2 (C-5), 140.5 (C-4), 129.1 (C-2), 123.9 (C-1), 121.0 (C-3), 96.0 (C-7), 61.8 (C-9), 60.2 (C-10), 14.4 (C-12), 13.6 (C-13), 13.1 (C-11); \(\nu\) (cm\(^{-1}\)) (neat) = 3245 (w, N-H), 1736 (s, C=O), 1659 (s, C=O), 1595 (s, C=C), 1500 (s, C=C).

Analytical data consistent with the previously reported.\(^{82}\)
7.3 Section II Experimental

\((1S,2R,6R)-1,3\text{-dimethyl}-7\text{-oxabicyclo}\{4.1.0\}\text{hept-3-en-2-yl)}\text{methanol (196)}

\[
\begin{array}{c}
\text{O} \\
1 \quad 2 \quad 3 \\
4 \quad 5 \\
6 \quad 7 \\
8 \quad 9
\end{array}
\]

A solution of VO(O-iPr)\(_3\) (9 \(\mu\)L, 0.038 mmol) and DMPU (14 \(\mu\)L, 0.12 mmol) in toluene (8 mL) was stirred at r.t. for 1 h, then cooled to 0 °C. Cumene hydroperoxide (0.64 mL, 4.34 mmol), and 197 (400 mg, 2.89 mmol) were added, and the mixture was allowed to return to r.t. and stirred for 16 h. Sat. aq. sodium sulfite (10 mL) was added, and the phases separated. The aqueous phase was extracted with EtOAc (3 x 10 mL), and the combined organics were dried (MgSO\(_4\)) and concentrated in vacuo. Purification by flash column chromatography (SiO\(_2\), 10-20% EtOAc/pet. ether 40-60) gave the desired product as a colourless oil (368 mg, 2.39 mmol, 83%).

\(R_f (50\% \text{EtOAc/pet. ether 40-60}) = 0.51; ^1H NMR (400 MHz, CDCl\(_3\)): \delta = 5.33 (1H, s, H-6), 4.18 (1H, dd, \(J = 11.4, 0.9\) Hz, H-1), 3.95-3.86 (1H, m, H-1'), 2.99 (1H, t, \(J = 1.6\) Hz, H-4), 2.67 (1H, d, \(J = 8.9\) Hz, O-H), 2.53-2.38 (2H, m, H-5), 2.23 (1H, s, H-2), 1.72 (3H, s, H-9), 1.45 (3H, s, H-8); \(^{13}C NMR (100 MHz, CDCl_3): \delta = 129.4 (C-7), 119.4 (C-6), 60.6 (C-1), 60.1 (C-3), 57.4 (C-4), 43.2 (C-2), 25.9 (C-5), 22.2 (C-8), 21.0 (C-9); ν (cm\(^{-1}\)) (neat) = 3449 (br m, O-H); HRMS: m/z (ESI) calculated for C\(_9\)H\(_{15}\)O\(_2\) [M+H\(^+\)]: 155.1072, found 155.1077.

\((2,6\text{-dimethylcyclohexa-2,5-dien-1-yl)}\text{methanol (197)}

\[
\begin{array}{c}
\text{O} \\
1 \quad 2 \quad 3 \\
4 \quad 5 \\
6
\end{array}
\]

To a suspension of LiAlH\(_4\) (2.54 g, 67.0 mmol) in Et\(_2\)O (100 mL) at 0 °C was added dropwise a solution of 210 (7.40 g, 45.0 mmol) in Et\(_2\)O (50 mL). The mixture was allowed to warm to r.t. and stirred for 12 h before being cooled to 0 °C again. Sat. aq. Rochelle’s salt (50 mL) was added, followed by stirring at r.t. for 3 h. The phases were separated, and the aqueous phase extracted with Et\(_2\)O (3 x 50 mL). The combined organics were washed with brine (100 mL), dried (MgSO\(_4\)) and concentrated in vacuo. Purification by flash column chromatography (SiO\(_2\), 20% EtO/pet. ether 40-60) gave the desired product as a colourless oil (5.37 g, 39.0 mmol, 87%).

\(R_f (20\% \text{EtO/pet. ether 40-60}) = 0.27; ^1H NMR (400 MHz, CDCl\(_3\)): \delta = 5.72 (2H, s, H-4), 3.77 (2H, s, H-1), 2.77-2.58 (3H, m, H-5 & H-2), 1.78 (6H, s, H-6); ^{13}C NMR (100 MHz,
CDCl₃: δ = 131.2 (C-3), 123.2 (C-4), 60.5 (C-1), 47.4 (C-2), 27.7 (C-5). 21.3 (C-6); ν (cm⁻¹) (neat) = 3375 (br s, O-H).

2,6-dimethylcyclohexa-2,5-diene-1-carboxylic acid (198)

A dried 250 mL three-necked round-bottomed flask was charged with 2,6-dimethylbenzoic acid (5.00 g, 33.3 mmol) and EtOH (25 mL). The flask was cooled to –78 °C and fitted with a dry ice condenser, and NH₃ (~100 mL) was condensed into the reaction mixture. Excess sodium metal was added piecewise until a dense precipitate formed and a blue colour persisted for longer than 10 min. The mixture was stirred at -78 °C for a further 2 h, followed by addition of NH₄Cl (6.37 g, 120 mmol). After stirring for a final 1 h at – 78 °C, the condenser and dry ice bath were removed, and the NH₃ was allowed to evaporate overnight. The mixture was then poured onto ice (200 mL) and acidified to pH 1.5 with 3 N aq. HCl, then extracted with Et₂O (3 x 100 mL). The combined organics were dried (MgSO₄) and concentrated in vacuo to give the desired product as a white solid (5.00 g, 33.3 mmol, quant.).

Rf (10% Et₂O/pet. ether 40-60) = 0.13; ¹H NMR (400 MHz, CDCl₃): δ = 5.70 (2H, s, H-4), 3.50 (1H, t, J = 6.3 Hz, H-2), 2.87-2.63 (2H, m, H-5), 1.80 (6H, s, H-6); ¹³C NMR (100 MHz, CDCl₃): δ = 178.7 (C-1), 128.4 (C-3), 122.4 (C-4), 52.1 (C-2), 27.5 (C-5), 21.8 (C-6); mp (H₂O) = 92-97 °C; ν (cm⁻¹) (neat) = 1702 (s, C=O), Microanalysis: calcd. For C₉H₁₂O₂: C 71.0%, H 7.95%, N 0.0%, found C 70.9%, H 8.03%, N 0.0%.

Methyl 2,6-dimethylcyclohexa-2,5-diene-1-carboxylate (210)

A mixture of 198 (8.41 g, 55.0 mmol), K₂CO₃ (15.6 g, 113 mmol) and iodomethane (6.9 mL, 111 mmol) where refluxed in acetone (150 mL) for 12 h. The solvent was removed in vacuo, and water (100 mL) was added, followed by extraction with Et₂O (3 x 100 mL). The combined organics were washed with brine (100 mL), dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 5% Et₂O/pet. ether 40-60) gave the desired compound as colourless oil (8.40 g, 51.0 mmol, 93%).
(S)-2-(1,3-dioxoisouindolin-2-yl)-3,3-dimethylbutanoic acid (212)

L-tert-leucine (1.31 g, 10.0 mmol), phthalic anhydride (1.48 g, 10.0 mmol) and TEA (1.3 mL, 9.32 mmol) were refluxed in toluene (20 mL) under Dean-Stark conditions for 16 h. The mixture cooled to r.t., then water (20 mL) and conc. HCl (0.2 mL), followed by stirring until a precipitate formed. This was collected by filtration, washed with cold water (5 mL) and dried in vacuo to give the desired product as a white solid (1.91 g, 7.3 mmol, 73%).

\[
\begin{align*}
\text{1H NMR (400 MHz, d}_6\text{-DMSO):} & \quad \delta = 7.95-7.89 \ (4H, m, H-7 \ & H-8), \ 4.47 \ (1H, s, H-2), \ 1.09 \ (9H, s, H-4); \\
\text{13C NMR (100 MHz, d}_6\text{-DMSO):} \ & \delta = 169.2 \ (C-1), \ 168.2 \ (C-5), \ 135.4 \ (C-7), \ 131.4 \ (C-6), \ 123.9 \ (C-8), \ 59.6 \ (C-2), \ 35.4 \ (C-3), \ 28.2 \ (C-4); \ v \ (\text{cm}^{-1}) \ (\text{neat}) = 3233 \ (\text{br m, O-H}), \ 1754 \ (s, C=O), \ 1701 \ (s, C=O); \ mp \ (H_2O) = 135-139 \ °C \ (\text{literature value = 154 °C}); ^{283}[\alpha]_D^{20} \ (c = 0.37, \text{MeOH}) = -35.4 \ °.
\end{align*}
\]

Diphenylmethanone oxime (214)

A solution of benzophenone (2.00 g, 11.0 mmol), H\textsubscript{2}NOH.HCl (3.06 g, 44.0 mmol) and pyridine (4.40 mL, 54.4 mmol) in MeOH (40 mL) was stirred at r.t. for 16 h. 1:1 pet. ether 40-60/EtOAc (10 mL) was added, followed by washing with 1 N aq. HCl (30 mL), water (30 mL) and brine (30 mL). The organics were dried (MgSO\textsubscript{4}) and then concentrated in vacuo. Purification by flash column chromatography (SiO\textsubscript{2}, 10% EtOAc/pet. ether 40-60) gave the desired product as a white solid (1.92 g, 9.74 mmol, 88%).

\[
\begin{align*}
\text{Rf (15% EtOAc/pet. ether 40-60) = 0.43; 1H NMR (500 MHz, CDCl}_{3}) : \ & \delta = 8.56 \ (1H, br s, O-H), \ 7.50-7.31 \ (10H, m, H-3 to H-5, H-7 to H-9); \ & \text{13C NMR (125 MHz, CDCl}_{3}) : \ & \delta = 158.1 \ (C-1),
\end{align*}
\]
136.2 (C-2 or C-6), 132.6 (C-2 or C-6), 129.5 (C-5 or C-9), 129.2 (C-3, C-4, C-7 or C-8), 129.1 (C-5 or C-9), 128.3 (C-3, C-4, C-7 or C-8), 128.2 (C-3, C-4, C-7 or C-8), 127.8 (C-3, C-4, C-7 or C-8); ν (cm⁻¹) (neat) = 3251 (br w, O-H), 1693 (m, C=N); mp (EtOAc) = 132-134 °C (literature value 138-142 °C).²⁸¹

Analytical data consistent with that previously published.²⁸¹

N-benzhydrylhydroxylamine (215)

![N-benzhydrylhydroxylamine](image)

To a solution of 214 (500 mg, 2.55 mmol) in MeOH (10 mL) was added a small amount of methyl orange, followed by cooling to 0 °C. 3 N aq. HCl was added dropwise until the solution turned red. NaBH₃CN (1.0 M in THF, 10.2 mL, 10.2 mmol) was added dropwise, accompanied by dropwise addition of 3 N aq. HCl as necessary to maintain the red colour. The solution was then allowed to warm to r.t., followed by stirring for 4 h whilst maintaining the acidity of the solution. The reaction was quenched by the addition of 3 N aq. HCl (3 mL), then basified with sat. aq. NaHCO₃. The mixture was then extracted with CH₂Cl₂ (3 x 15 mL), and the combined organics were dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 10-15% EtOAc/pet. ether 40-60) gave the desired compound as an off-white solid (414 mg, 2.08 mmol, 81%).

Rₐ (15% EtOAc/pet. ether 40-60) = 0.23; ¹H NMR (500 MHz, CDCl₃): δ = 7.40 (2H, d, J = 7.8 Hz, H-3), 7.33 (2H, t, J = 7.4 Hz, H-4), 7.26 (1H, t, J = 7.3 Hz, H-5), 5.23 (1H, s, H-1); ¹³C NMR (125 MHz, CDCl₃): δ = 140.7 (C-2), 128.7 (C-4), 127.7 (C-3), 127.6 (C-5), 70.7 (C-1); ν (cm⁻¹) (neat) = 3230 (br w, O-H), 2971 (m, N-H); mp (EtOAc) = 69-70 °C (literature value = 66-77 °C).²⁸²

Analytical data consistent with that previously reported.²⁸²

Ethylendiamine poisoned palladium on carbon

Pd/C.(en)

Prepared according to the reported procedure by Sajiki et al.²⁵⁰ Palladium on carbon (10% loading, 500 mg, 0.47 mmol) and ethylenediamine (2.2 mL, 33 mmol) were stirred under argon in MeOH (20 mL) at r.t. for 48 h. The product was collected by filtration, washed with MeOH (10 mL) and Et₂O (10 mL) and dried in vacuo to give the desired compound as a black solid.
Potassium azodicarboxylic acid

Prepared according to the reported procedure by Beruben et al.\textsuperscript{251} To a 40% aqueous solution of KOH (6 mL) in a 10 °C acetone-ice bath was added azodicarbonamide (2.00 g, 17.2 mmol) portionwise at a rate which maintained the temperature at 10 °C. After the addition, stirring was continued at the same temperature for a further 1 h, at which point the resulting solid was collected by filtration and washed with cold MeOH (5 mL) to give the desired product as a yellow solid (3.2 g, 15.9 mmol, 92%).

(1\textit{R},5\textit{R},6\textit{R})-6-chloro-5-(hydroxymethyl)-4,6-dimethylcyclohex-3-en-1-ol (231)

To HCl (2.0 M in Et\textsubscript{2}O, 0.50 mL) and was added 196 (100 mg, 0.65 mmol) in chloroform (1.50 mL) at -15 °C, followed by stirring for 3 h at the same temperature. 10% w/w aq. NaHCO\textsubscript{3} (1 mL) was added, and the phases separated. The aqueous phase extracted with CH\textsubscript{2}Cl\textsubscript{2} (3 x 2 mL), dried (MgSO\textsubscript{4}) and concentrated \textit{in vacuo}. Purification by flash column chromatography (SiO\textsubscript{2}, 10-20% EtOAc/pet. ether 40-60) gave the desired compound as an off-white solid (66.6 mg, 0.35 mmol, 54%) alongside residual starting material 196 (30.4 mg, 0.20 mmol, 31%)

\textbf{\textit{R}}\textsubscript{f} (30% Et\textsubscript{2}O/pet. ether 40-60) = 0.21; \textit{\textsuperscript{1}}H NMR (500 MHz, CDCl\textsubscript{3}): \textit{\delta} = 5.53-5.50 (1H, m, H-6), 3.94 (1H, dd, \textit{\textit{J}} = 11.9, 4.0 Hz, H-1), 3.90 (1H, dd, \textit{\textit{J}} = 11.7, 2.6 Hz, H-1'), 3.85 (1H, t, \textit{\textit{J}} = 5.0 Hz, H-4), 3.15 (1H, br s, O-H), 2.69-2.62 (1H, m, H-5), 2.57 (1H, s, H-2), 2.16-2.09 (1H, m, H-5'), 1.79 (3H, s, H-9), 1.67 (3H, s, H-8); \textit{\textsuperscript{13}}C NMR (125 MHz, CDCl\textsubscript{3}): \textit{\delta} = 130.4 (C-7), 121.3 (C-6), 74.4 (C-3), 71.7 (C-4), 59.4 (C-1), 53.8 (C-2), 31.7 (C-5), 24.0 (C-8), 21.8 (C-9); \textit{\nu} (cm\textsuperscript{-1}) (neat) = 3232 (br s, O-H); \textbf{HRMS:} m/z (ESI) calculated for C\textsubscript{9}H\textsubscript{16}O\textsubscript{3}Cl [M+H]\textsuperscript{+}: 191.0833, found 191.0827; \textbf{mp} (EtOAc) = 76-77 °C.

(1\textit{R},2\textit{R},3\textit{R},4\textit{R})-2-chloro-3-(hydroxymethyl)-2,4-dimethylcyclohexan-1-ol (232)
A mixture of 231 (66.6 mg, 0.35 mmol) and palladium on carbon (10%, 6.6 mg) in MeOH (3 mL) were stirred under H₂ (balloon) for 12 h. The mixture was filtered through Celite and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 10-40% Et₂O/pet. ether 40-60) gave the desired compound as colourless needle-like crystals (7.4 mg, 0.038 mmol, 11%).

Rf (80% Et₂O/pet. ether 40-60) = 0.28; ¹H NMR (400 MHz, CDCl₃): δ = 4.03 (2H, s, H-1), 3.75 (1H, dd, J = 11.4, 4.1 Hz, H-4), 2.43 (1H, d, J = 1.7 Hz, O-H), 1.89 (1H, dtd, J = 13.2, 4.3, 2.9 Hz, H-5eq), 1.75 (1H, dq, J = 13.2, 3.6 Hz, H-6eq), 1.70-1.59 (1H, m, H-7), 1.53 (3H, s, H-8), 1.52-1.38 (2H, m, H-5ax and H-2). 1.25-1.12 (1H, m, H-6ax), 1.05 (3H, d, J = 6.4 Hz, H-9); ¹³C NMR (100 MHz, CDCl₃): δ = 83.2 (C-3), 78.8 (C-4), 61.7 (C-1), 56.4 (C-2), 32.4 (C-6), 31.7 (C-7), 29.5 (C-5), 20.0 (C-9), 18.0 (C-8); ν (cm⁻¹) (neat) = 3394 (br s, O-H); HRMS: m/z (ESI) calculated for C₉H₁₈O₂Si [M+H]+ : 193.0995, found 19.0998; mp (pet. ether 40-60) = 100-103 °C.

tert-butyl(((1S,2R,6R)-1,3-dimethyl-7-oxabicyclo[4.1.0]hept-3-en-2-yl)methoxy)dimethylsilane (234)

A solution of 196 (50 mg, 0.32 mmol), TBDMSCl (51 mg, 0.34 mmol) and imidazole (23 mg, 0.34 mmol) in DMF (0.7 mL) was stirred at r.t. for 12 h. H₂O (3 mL) was added, followed by extraction with EtOAc (2 x 3 mL). The combined organic phases were washed with water (5 x 2 mL) and brine (2 x 2 mL), then dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 3% Et₂O/pet. ether 40-60) gave the desired product as a colourless oil (44.2 mg, 0.16 mmol, 51%).

Rf (10% Et₂O/pet. ether 40-60) = 0.55; ¹H NMR (500 MHz, CDCl₃): δ = 5.21-5.18 (1H, m, H-6), 3.81 (2H, d, J = 5.8 Hz, H-1), 3.00-2.98 (1H, m, H-4), 2.44-2.37 (3H, m, H-2, H-5), 1.72-1.70 (3H, m, H-9), 1.50 (3H, s, H-8), 0.91 (9H, s, H-12), 0.90 (3H, s, H-10), 0.90 (3H, s, H-10'); ¹³C NMR (125 MHz, CDCl₃): δ = 130.4 (C-7), 118.1 (C-6), 63.3 (C-1), 58.8 (C-3), 58.5 (C-4), 45.6 (C-2), 26.2 (C-5), 26.0 (C-12), 23.7 (C-8), 22.2 (C-9), 18.2 (C-11), -5.4 (C-10), -5.5 (C-10'); ν (cm⁻¹) (neat) = No characteristic absorptions; HRMS: m/z (ESI) calculated for C₁₅H₂₉O₂Si [M+H]+ : 269.1937, found 269.1924.
(1R,5R,6R)-5-(((tert-butyldimethylsilyl)oxy)methyl)-6-chloro-4,6-dimethylcyclohex-3-en-1-ol (235)

A solution of 234 (39 mg, 0.15 mmol) in CHCl₃ (1 mL) was cooled to 0 °C and HCl (2.0 M in Et₂O, 76 μL, 0.15 mmol) was added dropwise, followed by stirring at 0 °C for 1 h. Phosphate buffer (0.01 M, pH 7.4, 1 mL) was added, and the phases separated. The aqueous phase was extracted with EtOAc (3 x 2 mL), and the combined organics were dried (Na₂SO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 2-3% Et₂O/pet ether 40-60) gave the desired product as a colourless oil (21 mg, 0.069 mmol, 46%).

Rf (10% Et₂O/pet. ether 40-60) = 0.33; ¹H NMR (400 MHz, CDCl₃): δ = 5.47-5.43 (1H, m, H-6), 3.97 (1H, br d, J = 8.5 Hz, O-H), 3.92 (1H, dd, J = 10.8, 3.7 Hz, H-1), 3.87 (1H, dd, J = 10.8, 2.8 Hz, H-1'), 3.78 (1H, dt, J = 8.1, 5.4 Hz, H-4), 2.64-2.55 (1H, m, H-5), 2.51 (1H, s, H-2), 2.15-2.07 (1H, m, H-5'), 1.75-1.73 (3H, m, H-9), 1.64 (3H, s, H-8), 0.87 (9H, s, H-12), 0.08 (3H, s, H-10), 0.08 (3H, s, H-10'); ¹³C NMR (100 MHz, CDCl₃): δ = 130.7 (C-7), 120.8 (C-6), 75.4 (C-3), 72.1 (C-4), 60.1 (C-1), 53.9 (C-2), 31.7 (C-5), 25.7 (C-12), 23.8 (C-8), 21.8 (C-9), 18.1 (C-11), -5.71 (C-10), -5.74 (C-10'); ν (cm⁻¹) (neat) = No characteristic absorptions;

HRMS: m/z (ESI) calculated for C₁₅H₃₀O₂Si₃⁵Cl [M+H]⁺ : 305.1704, found 305.1690.

tert-butyl(((1S,2R,6R)-1,3-dimethyl-7-oxabicyclo[4.1.0]hept-3-en-2-yl)methoxy)diphenylsilane (237)

To a solution of 196 (100 mg, 0.65 mmol), imidazole (89 mg, 1.31 mmol) and DMAP (cat.) in CH₂Cl₂ (1 mL) at 0 °C was added TBDPSCl (0.19 mL, 0.72 mL). The mixture was allowed to warm to r.t. and stirred for 1 h. Water (1 mL) was added, and the phases separated. The aqueous phase was extracted with EtOAc (3 x 1 mL), and the combined organics were washed with brine (2 mL), dried (Na₂SO₄) and concentrated in vacuo. Purification by flash
column chromatography (SiO₂, 5% Et₂O/pet. ether 40-60) gave the desired product as a white solid (200 mg, 0.51 mmol, 79%).

Rᵣ (20% EtOAc/pet. ether 40-60) = 0.64; ¹H NMR (500 MHz, CDCl₃): δ = 7.75-7.71 (4H, m, H-12 & H-12’), 7.45-7.37 (6H, m, H-11, H-11’, H-13 & H-13’), 5.19-5.16 (1H, m H-6), 3.93 (1H, dd, J = 10.3, 6.2 Hz, H-1), 3.82 (1H, dd, J = 10.3, 4.8 Hz, H-1’), 3.01-2.99 (1H, m, H-4), 2.46-2.40 (3H, m, H-5 & H-2), 1.63-1.61 (3H, m, H-9), 1.47 (3H, s, H-8), 1.08 (9H, s, H-15); ¹³C NMR (125 MHz, CDCl₃): δ = 135.7 (C-12), 135.7 (C-12’), 133.6 (C-10), 133.6 (C-10’), 130.4 (C-7), 129.6 (C-13 & C-13’), 127.7 (C-11 & C-11’), 118.0 (C-6), 64.1 (C-1), 58.8 (C-3), 58.6 (C-4), 45.6 (C-2), 26.8 (C-15), 26.2 (C-5), 23.5 (C-8), 22.2 (C-9), 19.1 (C-14); ν (cm⁻¹) (neat) = 1588 (w, C=C); HRMS: m/z (ESI) calculated for C₂₅H₃₃O₂Si [M+H]⁺: 393.2244, found 393.2226; mp (pet. ether 40-60) = 66-67 °C.

(1R,5R,6R)-5-(((tert-butyldiphenylsilyl)oxy)methyl)-6-chloro-4,6-dimethylcyclohex-3-en-1-ol (238) & (1S,2R,6S)-2-(((tert-butyldiphenylsilyl)oxy)methyl)-6-chloro-1,3-dimethylcyclohex-3-en-1-ol (239)

To solution of 237 (189 mg, 0.48 mmol) in CHCl₃ (1 mL) at 0 °C was added HCl (2.0 M in Et₂O, 0.25 mL, 0.50 mmol). The mixture was stirred for 1 h at 0 °C, then allowed to warm to r.t. and stirred for a further 3 h. H₂O (1 mL) was added and the phases separated. The aqueous layer was extracted with EtOAc (3 x 1 mL), and the combined organics were washed with brine (2 mL), dried (Na₂SO₄) and concentrated in vacuo. Purification by flash column chromatography (2% Et₂O/pet. ether 40-60) gave the title compounds: 238 as a colourless semi-solid (74 mg, 0.18 mmol, 37%) and 239 as a white solid (30 mg, 0.070 mmol, 15%)

Data for (1R,5R,6R)-5-(((tert-butyldiphenylsilyl)oxy)methyl)-6-chloro-4,6-dimethylcyclohex-3-en-1-ol (238)

Rᵣ (10% Et₂O/pet. ether 40-60) = 0.18; ¹H NMR (400 MHz, CDCl₃): δ = 7.69-7.63 (4H, m, H-11 & H-11’), 7.47-7.36 (6H, m, H-12, H-12’, H-13 & H-13’), 5.47 (1H, s, H-6), 3.92-3.83 (4H, m, H-1, H-4, O-H), 2.64-2.55 (1H, m, H-5 or H-5’), 2.52 (1H, s, H-2), 2.20-2.10 (1H, m, H-5 or H-5’), 1.63-1.60 (6H, m, H-8 & H-9), 1.05 (9H, s, H-15); ¹³C NMR (100 MHz, CDCl₃): δ = 135.9 (C-11), 135.7 (C-11’), 132.7 (C-10),132.3 (C-10’), 131.6 (C-7), 130.0 (C-13), 130.0 (C-
13'), 127.8 (C-12), 127.7 (C-12'), 120.7 (C-6), 75.9 (C-3), 72.5 (C-4), 61.4 (C-1), 53.9 (C-2), 31.7 (C-5), 26.8 (C-15), 22.7 (C-8), 21.9 (C-9), 19.2 (C-14); ν (cm⁻¹) (neat) = 3405 (br w, O-H), 1589 (w, C=C); HRMS: m/z (ESI) calculated for C_{25}H_{34}O_{2}Si_{35}Cl [M+H]^+ : 429.2017, found 429.1997.

Data for (1S,2R,6S)-2-(((tert-butylidiphenylsilyl)oxy)methyl)-6-chloro-1,3-dimethylcyclohex-3-en-1-ol (239)

R_f (10% Et_2O/pet. ether 40-60) = 0.32; ^1H NMR (500 MHz, CDCl_3): δ = 7.72-7.69 (2H, m, H-11), 7.68-7.65 (2H, m, H-11'), 7.47-7.37 (6H, m, H-12, H-12', H-13, H-13'), 5.37-5.33 (1H, m, H-6), 4.66 (1H, dd, J = 9.5, 6.3 Hz, H-4), 4.04 (1H, dd, J = 11.0, 2.6 Hz, H-1), 3.77 (1H, dd, J = 11.0, 5.6 Hz, H-1'), 3.74 (1H, s, O-H), 2.73-2.66 (1H, m, H-5'), 2.33-2.24 (1H, m, H-5), 2.23 (1H, d, J = 4.33 Hz, H-2), 1.52-1.50 (3H, m, H-9), 1.29 (3H, s, H-8), 1.05 (9H, s, H-15); ^13C NMR (125 MHz, CDCl_3): δ = 135.7 (C-11 & C-11'), 132.8 (C-10), 132.7 (C-10'), 132.6 (C-7), 130.0 (C-13 & C-13'), 127.8 (C-12), 127.8 (C-12'), 121.4 (C-6), 73.3 (C-3), 64.3 (C-4), 61.9 (C-1), 52.8 (C-2), 35.0 (C-5), 26.8 (C-15), 22.8 (C-8), 21.9 (C-9), 19.1 (C-14); ν (cm⁻¹) (neat) = 3675 (w, O-H), 1588 (w, C=C); HRMS: m/z (ESI) calculated for C_{25}H_{34}O_{2}Si_{35}Cl [M+H]^+ : 429.2017, found 429.2028; mp (pet. ether 40-60) = 125-129 °C.

tert-butyl((2,6-dimethylcyclohexa-2,5-dien-1-yl)methoxy)diphenylsilane (247)

To a solution of 196 (500 mg, 3.62 mmol), imidazole (493 mg, 7.24 mmol) and DMAP (cat.) in CH_2Cl_2 (5 mL) at 0 °C was added TBDPSCI (1.03 mL, 3.98 mmol). The mixture was allowed to warm to r.t. and stirred for 1 h. Water (5 mL) was added, and the phases separated. The aqueous phase was extracted with CH_2Cl_2 (3 x 5 mL), and the combined organics were washed with brine (5 mL), dried (MgSO_4) and concentrated in vacuo. Purification by flash column chromatography (SiO_2, pet. ether 30-40) gave the desired product as a white solid (1.00 g, 2.66 mmol, 73%).

R_f (10% Et_2O/pet. ether 40-60) = 0.72; ^1H NMR (400 MHz, CDCl_3): δ = 7.68-7.65 (4H, m, H-8), 7.44-7.35 (6H, m, H-9 & H-10), 5.59 (2H, s, H-4), 3.72 (2H, d, J = 3.8 Hz, H-1), 2.73-2.50 (3H, m, H-5 & H-2), 1.71 (6H, s, H-6), 1.53 (9H, s, H-12); ^13C NMR (100 MHz, CDCl_3): δ = 135.7 (C-8), 133.9 (C-3), 133.3 (C-7), 129.5 (C-10), 127.6 (C-9), 121.5 (C-4), 64.0 (C-1), 47.9 (C-2), 27.8 (C-5), 26.8 (C-12), 22.1 (C-6), 19.3 (C-11); ν (cm⁻¹) (neat) = 1588 (w, C=C), 136
1427 (w, C=C aromatic); HRMS: m/z (ESI) calculated for C_{25}H_{33}OSi [M+H]^+ : 377.2301, found 377.2285; mp (CH_2Cl_2) = 58-60 °C.

**Methyl (2R,3aS,4R,7aR)-3a,5-dimethyl-2-phenyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxole-4-carboxylate (254) & Methyl (2S,3aS,4R,7aR)-3a,5-dimethyl-2-phenyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxole-4-carboxylate (255)**

To a mixture of NMO (112 mg, 0.96 mmol), OsO_4 (2.5 wt.% in t-BuOH, 0.35 mL, 0.027 mmol), acetone (1 mL) and H_2O (2 mL) was added 210 (150 mg, 0.90 mmol) at r.t., followed by stirring at r.t. for 16 h. A slurry of sodium hydrosulfite (100 mg) and florisil (excess) in H_2O (5 mL) was added, stirred at r.t. for 10 min then filtered. The filtrate was neutralised to pH 7 with 3N HCl, and the acetone was removed in vacuo. The residue was acidified to pH 2 with 3N HCl, saturated with NaCl and extracted with EtOAc (3 x 3 mL), dried (MgSO_4) and concentrated in vacuo. The residue was redissolved in CH_2Cl_2 (2 mL), then benzaldehyde dimethyl acetal (0.45 mL, 2.70 mmol) and p-TsOH (cat.) were added, followed by stirring at r.t. for 16 h. Sat. aq. NaHCO_3 (2 mL) was added and the phases separated. The aqueous phase was extracted with CH_2Cl_2 (3 x 2 mL) and then the combined organic phases were washed with brine (3 mL), dried (MgSO_4) and concentrated in vacuo. Purification by flash column chromatography (SiO_2, 2-80% Et_2O/pet. ether 40-60) gave a 1:2 mixture (based on ^1H NMR analysis, using the spectra of the pure enantioenriched compounds [vide infra] as references) of 255 and 254 respectively (131 mg, 0.45 mmol, 50%).

254 and 255 were prepared in enantioenriched form using an identical protection procedure starting from a crude sample of 250 (150 mg, 0.75 mmol [maximum, assuming pure]) prepared using the Sharpless Asymmetric Dihydroxylation procedure described above, yielding 255 as a colourless oil (21 mg, 0.073 mmol, 10%) and 254 as a colourless oil (41 mg, 0.14 mmol, 19%). Enantiomeric excess was determined using chiral HPLC with a Phenomenex Amylose 1 chiral column, eluting with 50:50 Water/MeCN and a flow rate of 1.5 mL/min. The t_H for each enantiomer is listed, followed by the integral ratio for the enantioenriched case (the ratio for the racemic sample was 50:50 as expected).
Data for Methyl (2R,3aS,4R,7aR)-3a,5-dimethyl-2-phenyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxole-4-carboxylate (254)

\[
\begin{align*}
R_f (10\% \text{ Et}_2\text{O/pet. ether } 40-60) & = 0.25; \quad \text{HPLC } t_R = 7.64, 8.35 \text{ min (40:60)}; \quad \text{\textsuperscript{1}H NMR} \quad (400 \text{ MHz, CDCl}_3): \quad \delta = 7.45-7.41 (2H, m, H-14), 7.37-7.33 (3H, m, H-13 & H-15), 5.76-5.71 (2H, m, H-6 & H-11), 4.32 (1H, d, \textit{J} = 4.1 \text{ Hz, H-4}), 3.69 (3H, s, H-10), 3.33 (1H, s, H-2), 2.69-2.60 (1H, m, H-5'), 2.39 (1H, dd, \textit{J} = 16.6, 6.8 \text{ Hz, H-5}), 1.90 (3H, s, H-9), 1.53 (3H, s, H-8); \quad \text{\textsuperscript{13}C NMR} \quad (100 \text{ MHz, CDCl}_3): \quad \delta = 171.1 (C-1), 137.1 (C-12), 132.1 (C-7), 129.6 (C-15), 128.4 (C-13), 127.1 (C-14), 122.0 (C-6), 100.9 (C-11), 82.0 (C-3), 80.4 (C-4), 55.3 (C-10), 51.8 (C-2), 29.2 (C-5), 23.2 (C-9), 22.8 (C-8); \quad \text{\textnu} (\text{cm}^{-1}) (\text{neat}) = 1731 (s, C=O), 1450 (w, C=C), 1433 (w, C=C); \quad \left[\alpha\right]^D_{20} \quad (c = 0.67, \text{CH}_2\text{Cl}_2) = -27.6 ^\circ.
\end{align*}
\]

Data for Methyl (2S,3aS,4R,7aR)-3a,5-dimethyl-2-phenyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxole-4-carboxylate (255)

\[
\begin{align*}
R_f (10\% \text{ Et}_2\text{O/pet. ether } 40-60) & = 0.25; \quad \text{HPLC } t_R = 9.54, 10.7 \text{ min (39:61)}; \quad \text{\textsuperscript{1}H NMR} \quad (400 \text{ MHz, CDCl}_3): \quad \delta = 7.47-7.42 (2H, m, H-14), 7.38-7.33 (3H, m, H-13 & H-15), 5.87 (1H, s, H-11), 5.75 (1H, d, \textit{J} = 6.4 \text{ Hz, H-6}), 4.52 (1H, d, \textit{J} = 6.4 \text{ Hz, H-4}), 3.69 (3H, s, H-10), 3.42 (1H, s, H-2), 2.81-2.70 (1H, m, H-5'), 2.37 (1H, dd, \textit{J} = 17.9, 6.6 \text{ Hz, H-5}), 1.85-1.83 (3H, m, H-9), 1.52 (3H, s, H-8); \quad \text{\textsuperscript{13}C NMR} \quad (100 \text{ MHz, CDCl}_3): \quad \delta = 171.4 (C-1), 138.1 (C-12), 131.5 (C-7), 129.2 (C-15), 128.4 (C-13), 126.7 (C-14), 122.6 (C-6), 104.3 (C-11), 81.6 (C-3), 79.5 (C-4), 54.9 (C-2), 51.9 (C-10), 30.2 (C-5), 25.6 (C-8), 23.1 (C-9); \quad \text{\textnu} (\text{cm}^{-1}) (\text{neat}) = 1730 (s, C=O), 1450 (w, C=C), 1402 (w, C=C); \quad \left[\alpha\right]^D_{20} \quad (c = 0.48, \text{CH}_2\text{Cl}_2) = -12.5 ^\circ.
\end{align*}
\]
Methyl (3aS,4R,7aR)-2,2,3a,5-tetramethyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxole-4-carboxylate (256)

Sharpless asymmetric hydroxylation mix-β (40 g) and methane sulfonamide (4.16 g, 43.7 mmol) were stirred in H₂O (150 mL) and t-BuOH (150 mL) at r.t. for 5 min. The mixture was cooled to 0 °C and 210 (6.00 g, 36.1 mmol) was added dropwise, then allowed to warm to r.t. and stirred for 16 h. Sodium sulfite (65 g) was then added, followed by stirring at r.t. for 1 h. The mixture was then extracted with CH₂Cl₂ (3 x 150 mL) and EtOAc (2 x 150 mL), and the combined organics were washed with 10% aq. NaOH (200 mL), dried (MgSO₄) and concentrated in vacuo. The residue was redissolved in acetone (50 mL) and 2,2-dimethoxypropane (30 mL) and a catalytic amount of p-TsOH was added, followed by stirring at r.t. for 48 h. The solvent was then removed in vacuo and sat. aq. Na₂CO₃ (50 mL) was added, followed by extraction with Et₂O (3 x 50 mL). The combined organics were dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 5% Et₂O/pet. ether 40-60) gave the desired product as a colourless oil (5.26 g, 21.8 mmol, 60%).

\( R_f \) (10% Et₂O/pet. ether 40-60) = 0.45; ¹H NMR (400 MHz, CDCl₃): \( \delta = 5.55 \) (1H, s H-6), 4.20 (1H, dd, J = 5.7, 1.4 Hz, H-4), 3.65 (3H, s, H-10), 3.22 (1H, s, H-2), 2.57 (1H, m, H-5'), 2.29 (1H, dd, J = 17.9, 5.9 Hz, H-5), 1.74 (3H, s, H-9), 1.38 (3H, s, H-8), 1.36 (3H, s, H-13), 1.33 (3H, s, H-12); ¹³C NMR (100 MHz, CDCl₃): \( \delta = 171.9 \) (C-1), 131.5 (C-7), 121.4 (C-6), 108.4 (C-11), 81.8 (C-3), 79.1 (C-4), 55.6 (C-2), 51.7 (C-10), 29.0 (C-5), 27.7 (C-12), 27.6 (C-13), 24.0 (C-8), 22.4 (C-9); ν (cm⁻¹) (neat) = 1733 (s, C=O); HRMS: m/z (ESI) calculated for C₁₃H₂₁O₄ [M+H]⁺: 241.1434, found 241.1425; [α]₂⁰° (c = 0.08, CH₂Cl₂) = -172.3 °.

((3aS,4S,7aR)-2,2,3a,5-tetramethyl-3a,4,7,7a-tetrahydrobenzo[d][1,3]dioxol-4-yl) methanol (257)

To a suspension of LiAlH₄ (446 mg, 11.7 mmol) in Et₂O (12 mL) at 0 °C was added dropwise a solution of 256 (1.13 g, 4.70 mmol) in Et₂O (6 mL). The reaction was allowed to warm to r.t.
and stirred for 16 h. Sat. aq. potassium sodium tartrate (20 mL) was added dropwise at 0 °C, followed by stirring at r.t. for 1 h. The phases were separated, and the organic phases extracted with Et₂O (5 x 15 mL). The combined organic phases were washed with brine (30 mL), dried (MgSO₄) and concentrated in vacuo. Purification by flash column chromatography (SiO₂, 30% Et₂O/pet. ether 40-60) gave the desired product as a colourless oil (844 mg, 3.98 mmol, 85%).

Rᵣ (30% Et₂O/pet. ether 40-60) = 0.14; †H NMR (400 MHz, d₆-DMSO): δ = 5.38 (1H, s, H-6), 4.44 (1H, dd, J = 5.7, 4.3 Hz, H-4), 3.92 (1H, dd, J = 6.7, 2.8 Hz, O-H), 3.64 (1H, dt, J = 10.6, 4.0 Hz, H-1), 3.54 (1H, ddd, J = 10.6, 7.3, 5.6 Hz, H-1'), 2.48-2.40 (1H, m, H-5 or H-5'), 2.14 (1H, s, H-2), 2.08-2.01 (1H, m, H-5' or H-5), 1.77 (3H, s H-9), 1.29 (6H, s, H-11 & H-12), 1.13 (3H, s, H-8); †C NMR (100 MHz, d₆-DMSO): δ = 136.9 (C-7), 119.3 (C-6), 107.5 (C-10), 83.0 (C-3), 79.6 (C-4), 59.1 (C-1), 49.8 (C-2), 29.1 (C-5), 28.7 (C-11 or C-12), 28.0 (C-12 or C-11), 23.1 (C-8), 21.8 (C-9); ν (cm⁻¹) (neat) = 3488 (br w, O-H); HRMS: m/z (ESI) calculated for C₁₂H₂₁O₃[M+H]⁺: 213.1485, found 213.1467; [α]D²⁰ (c = 0.10, CH₂Cl₂) = -6.3 °.

((3aS,4S,5S,7aR)-2,2,3a,5-tetramethylhexahydrobenzo[d][1,3]dioxol-4-yl)methanol (258) & ((3aS,4S,5R,7aR)-2,2,3a,5-tetramethylhexahydrobenzo[d][1,3]dioxol-4-yl)methanol (258-diast)

To a solution of Crabtree’s catalyst (48 mg, 0.060 mmol) in CH₂Cl₂ (30 mL) was added a solution of 257 (840 mg, 3.96 mmol) in CH₂Cl₂ (5 mL). The mixture was then stirred at r.t. under H₂ (balloon) for 16 h. The solution was then passed through Celite and concentrated in vacuo. †H NMR analysis of the crude product revealed a ratio of 92:8 of the desired to undesired diastereomer. Purification by flash column chromatography (SiO₂, 20-30% Et₂O/pet. ether 40-60) gave 258 as a colourless oil (585 mg, 2.73 mmol, 69%) and 258-diast as a colourless oil (6.5 mg, 0.030 mmol, 1%).

Data for ((3aS,4S,5S,7aR)-2,2,3a,5-tetramethylhexahydrobenzo[d][1,3]dioxol-4-yl)methanol (258)

Rᵣ (50% Et₂O/pet. ether 40-60) = 0.30; †H NMR (400 MHz, d₆-DMSO): δ = 4.09 (1H, dd, J = 5.2, 3.8 Hz O-H), 3.70 (1H, m, H-4), 3.64 (1H, dt, J = 10.8, 3.6 Hz, H-1), 3.55-3.49 (1H, m, H-1'), 1.95-1.88 (1H, m, H-5), 1.69-1.58 (1H, m, H-5'), 1.41-1.16 (10H, m, H-2, H-6, H-6', H-7, H-11 & H-12), 1.13 (3H, s, H-8), 0.93 (3H, d, J = 5.6 Hz, H-9); †C NMR (100 MHz, d₆-
DMSO): \( \delta = 106.5 \) (C-10), \( 81.8 \) (C-3), \( 79.9 \) (C-4), \( 58.9 \) (C-1), \( 51.1 \) (C-2), \( 31.5 \) (C-7), \( 30.7 \) (C-6), \( 29.1 \) (C-11), \( 27.6 \) (C-12), \( 25.5 \) (C-5), \( 20.3 \) (C-9), \( 19.8 \) (C-8); \( \nu \) (cm\(^{-1}\)) (neat) = 3946 (br w, O-H); HRMS: m/z (ESI) calculated for C\(_{12}H_{23}O_3\) [M+H]\(^+\) : 215.1642, found 215.1640; \([\alpha]_D^{20}\) (c = 0.24, CH\(_2\)Cl\(_2\)) = -8.3 \(^\circ\).

Data for \((3aS,4S,5R,7aR)-2,2,3a,5-tetramethylhexahydrobenzo[d][1,3]dioxol-4-yl)methanol (258-diaint)

\( R_f \) (50% EtOAc/pet. ether 40-60) = 0.18; \(^1\)H NMR (500 MHz, d\(_6\)-DMSO): \( \delta = 4.20 \) (1H, dd, \( J = 6.1, 4.2 \) Hz, O-H), 3.72 (1H, t, \( J = 2.9 \) Hz, H-4), 3.58 (1H, dt, \( J = 10.2, 3.9 \) Hz, H-1), 3.35-3.27 (1H, m, H-1'), 2.15-2.08 (1H, m, H-7), 1.80-1.76 (2H, m, H-5 & H-5'), 1.72-1.63 (1H, m H-6), 1.35 (3H, s, H-11), 1.29-1.23 (1H, m, H-6'), 1.23 (3H, s, H-12), 1.02 (3H, s, H-8), 0.79 (3H, d, \( J = 7.4 \) Hz, H-9); \(^{13}\)C NMR (125 MHz, d\(_6\)-DMSO): \( \delta = 106.1 \) (C-10), \( 79.2 \) (C-3), \( 79.2 \) (C-4), \( 58.0 \) (C-1), \( 47.6 \) (C-2), 28.5 (C-11), 27.2 (C-12), 26.2 (C-7), 25.9 (C-6), 21.7 (C-8), 20.8 (C-5), 12.5 (C-9); \( \nu \) (cm\(^{-1}\)) (neat) = 3445 (br w, O-H); HRMS: m/z (ASAP+) calculated for C\(_{12}H_{22}O_3\) [M]\(^+\) : 214.1569, found 214.1560; \([\alpha]_D^{20}\) (c = 0.12, CH\(_2\)Cl\(_2\)) = -3.5 \(^\circ\).

\((3aS,4R,5S,7aR)-2,2,3a,5-tetramethylhexahydrobenzo[d][1,3]dioxole-4-carbaldehyde (259)

A mixture of 258 (1.00 g, 4.60 mmol), NMO (820 mg, 7.00 mmol) and activated 4 Å molecular sieves in CH\(_2\)Cl\(_2\) (50 mL) was stirred at r.t. for 10 min. TPAP (83 mg, 0.24 mmol) was added, followed by MeCN (10 mL), followed by stirring at r.t. for 16 h. The solution was then eluted through a silica gel pad and concentrated \textit{in vacuo} to give the desired product as a white solid (750 mg, 3.53 mmol, 76%).

\( R_f \) (50% EtOAc/pet. ether 40-60) = 0.54; \(^1\)H NMR (400 MHz, CDCl\(_3\)): \( \delta = 9.85 \) (1H, d, \( J = 2.7 \) Hz, H-1), 3.82 (1H, t, \( J = 3.1 \) Hz, H-4), 2.47 (1H, dd, \( J = 11.5, 2.7 \) Hz, H-2), 2.12-2.05 (1H, m, H-5'), 1.75-1.62 (2H, m, H-5 & H-7), 1.59 (3H, s, H-12), 1.55-1.47 (1H, m, H-6'), 1.39-1.27 (4H, m, H-6 & H-11), 1.24 (3H, s, H-8), 0.91 (3H, d, \( J = 6.4 \) Hz, H-9); \(^{13}\)C NMR (100 MHz, CDCl\(_3\)): \( \delta = 205.4 \) (C-1), 108.3 (C-10), 80.3 (C-3), 80.0 (C-4), 63.4 (C-2), 29.4 (C-7), 28.7 (C-12), 28.6 (C-6), 27.2 (C-11), 25.3 (C-5), 20.1 (C-8), 20.0 (C-9); \( \nu \) (cm\(^{-1}\)) (neat) = 1725 (s, C=O); HRMS: Data could not be obtained; mp (CH\(_2\)Cl\(_2\)) = 82-88 \(^\circ\)C; \([\alpha]_D^{20}\) (c = 0.10, CH\(_2\)Cl\(_2\)) = -7.0 \(^\circ\).
(1R,2S,3R,6S)-2,3-dihydroxy-2,6-dimethylcyclohexane-1-carbaldehyde (260)

259 (750 mg, 3.53 mmol) was stirred in a mixture of THF (10 mL) and 3N HCl (10 mL) at r.t. for 16 h. Half of the solvent was removed *in vacuo* and sat. aq. NaHCO₃ (10 mL) was added, followed by extraction with EtOAc (3 x 10 mL). The combined organic phases were dried (MgSO₄) and concentrated *in vacuo* to give the desired product as a colourless oil (491 mg, 2.85 mmol, 81%).

Rᵣ (50% Et₂O/pet. ether 40-60) = 0.59; ¹H NMR (500 MHz, d₆-DMSO): δ = 9.79 (1H, d, J = 3.1 Hz, H-1), 4.62 (1H, d, J = 3.2 Hz, OH-4), 4.25 (1H, s, OH-3), 3.33-3.30 (1H, m, H-2), 2.40 (1H, dd, J = 11.2, 3.2 Hz, H-4), 1.82-1.72 (1H, m H-7), 1.60-1.49 (2H, m, H-5 & H-5'), 1.36-1.21 (2H, m, H-6 & H-6'), 1.07 (3H, s, H-8), 0.75 (3H, d, J = 6.3 Hz, H-9); ¹³C NMR (125 MHz, d₆-DMSO): δ = 207.6 (C-1), 73.7 (C-4), 73.7 (C-3), 61.1 (C-2), 29.1 (C-7), 29.0 (C-5), 27.5 (C-6), 21.8 (C-8), 20.7 (C-9); ν (cm⁻¹) (neat) = 3425 (br s, O-H), 1704 (s, C=O); HRMS: m/z (ESI-) calculated for C₁₂H₂₂O₅ [M-H]: 171.1027, found 171.1027; [α]₂⁰°(c = 0.39, MeOH) = -17.9 °.
Bibliography


Appendix: NMR Spectra (Including data for tentatively assigned structures)
29
30 (First Route)
30 (Second Route)
MeOH 74
196
In CDCl₃:

In d₆-DMSO:
231-acetate
258-diast