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FACULTY OF ENGINEERING, SCIENCE AND MATHEMATICS

School of Chemistry

Total Synthesis of Ulocladol A and Analogues, Alternariol 9-Methyl Ether and Alternariol

by

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Thesis for the degree of Doctor of Philosophy

UNIVERSITY OF SOUTHAMPTON

ABSTRACT

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TOTAL SYNTHESIS OF ULOCLADOL A AND ANALOGUES, ALTERNARIOL 9-METHYL
ETHER AND ALTERNARIOL

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Ulocladol A is a natural product which has been shown to exhibit tyrosine kinase
inhibitory activity against the enzyme Lck. It was isolated from the marine sponge
Callyspongia vaginalis in 1999. Prior to the commencement of this project there had been
three reported syntheses of ulocladol A, all of which started from intermediates prepared
in the synthesis of graphislactones C and D. The routes were relatively long and did not
provide much scope for analogous compounds.

We proposed it would be possible to prepare ulocladol A via a route which would allow for
the synthesis of analogues. Any analogues prepared would be tested against five tyrosine
kinases to determine whether any SAR could be elucidated.

This report details the use of Suzuki coupling methodology to prepare ulocladol A and
eighteen analogues. Several different catalyst systems are reported due to the steric and
electronic effects of the substrates used in the Suzuki reaction; also included in the report
are the results of the biological testing. Two of the analogues, **2.47** and **2.60**, exhibit
greater inhibitory activity than ulocladol A and show good activity against four other
tyrosine kinases.

In addition to the synthesis of ulocladol A and analogues, the report also details the
synthesis of alternariol 9-methyl ether and alternariol in the shortest convergent synthesis
to date.

Contents

Declaration of Authorship

I, Lauren Jayne Sudlow, declare that the thesis entitled "*Total synthesis of ulocladol A and analogues, alternariol 9-methyl ether and alternariol*" and the work presented in the thesis are both my own, and have been generated by me as the result of my own original research. I confirm that:

- this work was done wholly or mainly while in candidature for a research degree at this University;
- where any part of this thesis has previously been submitted for a degree or any other qualification at this university or any other institution, this has clearly been stated;
- where I have consulted the published work of others, this is always clearly attributed;
- where I have quoted from the work of others, the source is always given. With the exception of such quotations, this thesis is entirely my own work;
- I have acknowledged all main sources of help;
- where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself;
- none of this work has been published before submission.

Signed: ....

Date: ....

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Abbreviations

The following abbreviations have been used in the text.

Ac	Acetyl
ADP	Adenosine diphosphate
aq.	Aqueous
ATP	Adenosine triphosphate
Boc	*tert*-Butoxycarbonyl
Bn	Benzyl
br	Broad
Btk	Bruton's tyrosine protein kinase
CoA	Coenzyme A
conc	Concentrated
d	Doublet
Decomp.	Decomposed
DIAD	Diisopropylazodicarboxylate
DIBAL-H	Diisobutylaluminium hydride
DIPEA	Diisopropylethylamine
DMAP	4-Dimethylaminopyridine
DMF	*N,N'*-Dimethylformamide
DMSO	Dimethyl sulfoxide
DPPP	1,3-Bis(diphenylphosphino)propane
EDC	1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide
Equiv	Equivalents
ES-	Negative electrospray
ES+	Positive electrospray
FTIR	Fourier transform infrared
Fyn	Proto-oncogene tyrosine protein kinase
h	Hour
HOBt	1-Hydroxybenzotriazole
HRMS	High resolution mass spectrum
IR	Infrared
Lck	Lymphocyte-specific tyrosine protein kinase
LDA	Lithium diisopropylamide

LRMS	Low resolution mass spectrum
Lyn	V-yes-1 Yamaguchi sarcoma viral related oncogene homolog tyrosine protein kinase
m	Multiplet
min	Minute
M.P	Melting point
MS	Mass spectrum
mw	Microwave
NBS	*N*-Bromosuccinimide
NMR	Nuclear magnetic resonance
ppm	Parts per million
PTSA	*p*-Toluenesulfonic acid
q	Quartet
quant.	Quantitative
s	Singlet
SAR	Structure-Activity Relationship
Src	Sarcoma tyrosine protein kinase
rt	Room temperature
t	Triplet
TBDMS	*tert*-Butyldimethylsilyl
TFA	Trifluoroacetic acid
TFAA	Trifluoroacetic anhydride
Tf	Trifluoromethanesulfonyl
THF	Tetrahydrofuran
TLC	Thin layer chromatography
TMS	Trimethylsilane
UV	Ultraviolet
VLC	Vacuum-liquid chromatography

1. Introduction

1.1   Natural Products

A compound which is derived from a source such as a plant, animal or microorganism is defined as a natural product.[1] Although a large number of natural products are biologically inactive or have no known biological activity, there are some which exhibit interesting activity. Many of these biologically active natural products have been used medicinally for thousands of years. The first written record, dating back to 2600 BC, contained approximately 1000 substances from plant sources which were used to treat illnesses from simple colds to parasitic infections.[2] Other early records include the Egyptian *Ebers Papyrus* dating from 1500 BC and the Chinese *Materia Medica* dating from 1100 BC. The records also included formulae of how to administer the crude natural products, such as gargles, poultices and infusions. However, it wasn't until the start of the 19th century that Friedrich Sertürner isolated morphine, the first pharmacologically active, pure compound from a plant.[3] This isolation led to huge interest in the extraction and purification of drugs from plants, as well as the vast screening of microorganisms following the discovery of penicillin by Alexander Fleming in 1928. Towards the end of the 20th century natural products or natural product inspired compounds accounted for approximately 80% of drugs, including morphine, lovastatin and paclitaxel (Figure 1).[3]

Morphine   Lovastatin   Paclitaxel
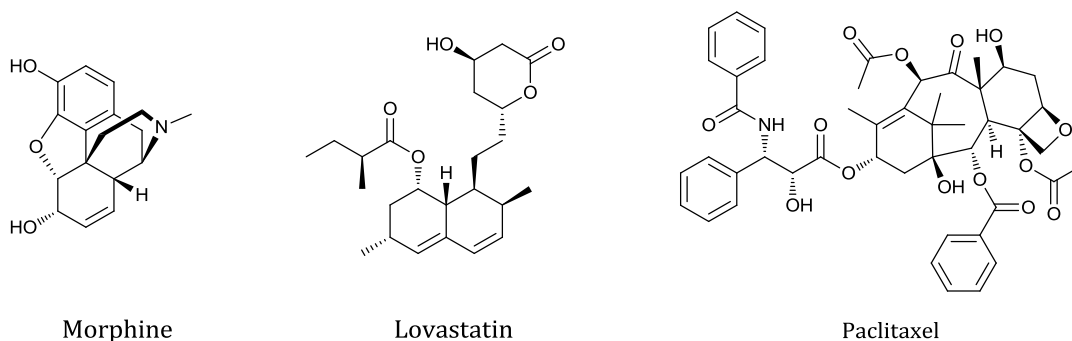
Figure 1 – Natural products

The three natural products were isolated from different natural sources: morphine, an analgesic, was isolated from the opium poppy plant; lovastatin, used to lower cholesterol, was isolated from the fungus *Aspergillus terreus* and paclitaxel, an anti-cancer agent, was isolated from the Pacific yew tree. They all have very different structures which are highly oxygenated with three or more rings and multiple stereogenic centres.

Despite the quantity of drugs derived from natural products and the huge variation in structure, the arrival of high throughput screening and combinatorial chemistry towards the end of the 20th century caused the interest in natural products to decline. Pharmaceutical companies placed great emphasis on the ability to prepare and screen large libraries of synthetic compounds quickly and consequently vastly reduced or terminated their research in the more time consuming area of natural products.[4] They began screening large libraries of compounds, assuming that if they screened enough compounds a lead would be found. The early libraries often contained peptides and nucleotides which were easy to synthesise through combinatorial chemistry and could undergo functional group interconversions to prepare large numbers of compounds for screening. However, the compounds were often limited in the variety of functional groups, chiral centres and ring systems usually observed in natural products and small drug molecules, resulting in disappointment in the pharmaceutical industry when the large quantities of new drugs anticipated failed to arise.

Although the process of combinatorial chemistry and screening of synthetic libraries has been refined and emphasis is now placed on quality, not quantity of compounds, a revitalisation in the area of natural products has taken place. There are several reasons for this, one of which is the competition created by the refinement of synthesis and screening of synthetic compounds. This refinement has meant that the process of extraction, isolation and testing of natural products has had to evolve in order to remain viable, and automation is increasingly being used in the extraction and isolation stages.

A second reason is the chemical diversity seen within natural products. Natural products occupy a more diverse area of chemical space when compared to that of synthetic compounds, with less aromaticity and nitrogen atoms but a greater number of chiral centres, ring systems and oxygen atoms. A comparison between the chemical space occupied by natural products, drugs and a selection of combinatorial compounds demonstrates this diversity (Figure 2).[4]
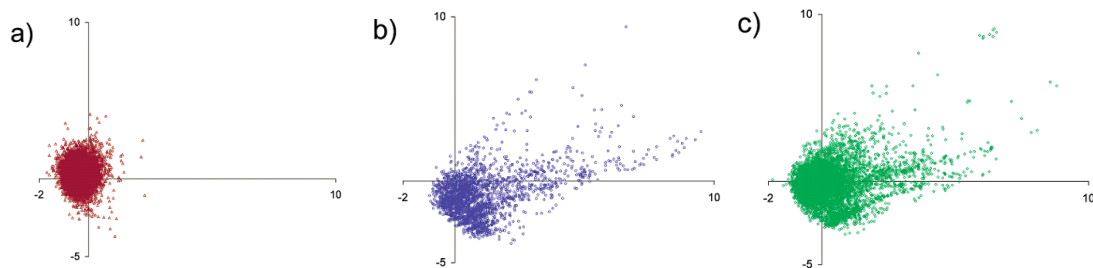
The plot of the first two principal components, obtained from a database containing a) a random selection of combinatorial compounds (n 13 506), b) natural products (n 3287), and c) drugs (n 10 968). For clarity, the data points from the three databases are plotted separately but on the same axes. The first two components explain about 54% of the variance. The figure shows that combinatorial compounds cover a well-defined area in the diversity space given by these principal components. In contrast, natural products and drugs cover almost all of this space as well as a much larger additional volume. Drugs and natural products have approximately the same coverage of this space.

Figure 2 – Occupation of chemical space

The graphs show that combinatorial compounds **a)** occupy a relatively localised area of chemical space. Natural products **b)** and drugs **c)** cover a much more diverse area as well as the same area as combinatorial compounds. This diversity has been demonstrated in the period between 2005 and 2007, with approval being given for the introduction of 13 natural product based drugs, of which, five were members of new classes (Figure 3).[5]
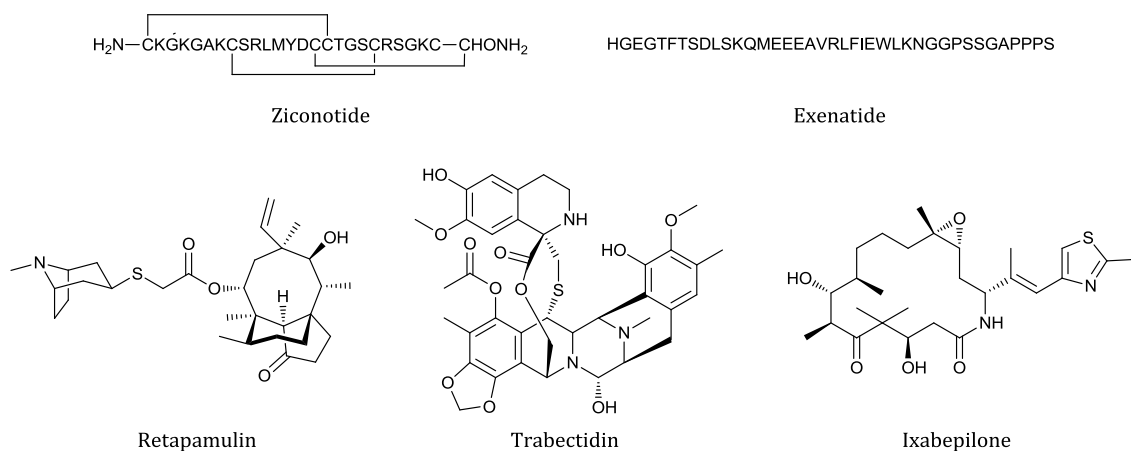
Figure 3 – Approved natural product based drugs

Ziconotide is derived from a toxin produced by the cone snail *Conus magus*. It is a member of the conotoxin class of drugs and is used in the treatment of chronic pain.[6] Exenatide, derived from a hormone found in the saliva of the Gila monster, is an incretin mimetic used to improve control of blood sugar levels in type II diabetics by enhancing glucose-dependent insulin secretion, slowing gastric emptying and suppressing elevated glucagon secretion.[7] Retapamulin is a topical antibiotic derived from the natural product

pleuromutilin which is produced by the mushroom *Pleurotus mutilus*. It is the first of a new class of drugs, named the pleuromutilin antibiotics, which have been shown to be effective against some Gram-positive bacteria, including MRSA.[8] Trabectidin, isolated from the sea squirt *Ecteinascidia turbinate*, and ixabepilone, an epothilone secreted by the bacterium *Sorangium cellulosum*, are both members of new classes of drugs aimed at the treatment of cancer. Trabectidin is a tetrahydroisoquinoline alkaloid which has been approved for the treatment of advanced soft tissue sarcoma, given orphan drug status in the treatment of relapsed ovarian cancer and is currently undergoing clinical trials for the treatment of breast and prostate cancer. Trabectidin is thought to bind to the minor groove of DNA and cause inhibition of cell proliferation by interrupting the cell cycle. Ixabepilone acts against tumour cells in a different way, by binding to microtubule β-subunits and stabilising them. This blocks the cells in the mitotic phase causing apoptosis; the same mechanism of action employed by Taxol. An advantage of Ixabepilone is its ability to remain active against cancers which are resistant to Taxol.

In addition to Trabectidin and Ixabepilone, nearly 40% of more than 200 natural product inspired compounds, undergoing preclinical and clinical trials in 2008, were aimed at cancer targets.[9] As the overall age-standardised incidence rate for cancer has increased by 25% in the period between 1977 and 2006, cancer therapies are becoming increasingly important.[10] Recent breakthroughs in the understanding of cell communication have encouraged research into specific targets, with focus on what causes cancer cells to replicate allowing tumours to grow. One of the areas being focused on is protein kinases.

1.2 Protein Kinases

Protein kinases are enzymes which catalyse the modification of protein substrates through phosphorylation of specific amino acid residues. This phosphorylation takes place in the enzyme active site and leads to a conformational change in the protein substrate. The conformational change results from several factors: the addition of the phosphate group to the amino acid residue disrupts hydrogen bonding on the phosphorylated functional group; and the introduction of the two negative charges, from the phosphate group, enables the formation of ionic bonds with cationic species leading to a change in the tertiary structure of the protein substrate. This conformational change is the means by which many cellular processes are regulated; these include metabolism, transcription, differentiation, apoptosis, ion transport and hormone responses.[11]

Prior to catalysing the phosphorylation of a protein substrate, the protein kinase must itself be activated. Protein kinases have two conformations: one active, where phosphorylation of the protein substrate can take place, and one inactive, where no phosphorylation occurs.

The protein kinase domain consists of two lobes, the N-lobe and the C-lobe, which sandwich the enzyme active site. The N-lobe is the smaller of the two and consists of a five-stranded β-sheet and one α-helix; the C-lobe is larger, predominantly consisting of α-helices (Figure 4).
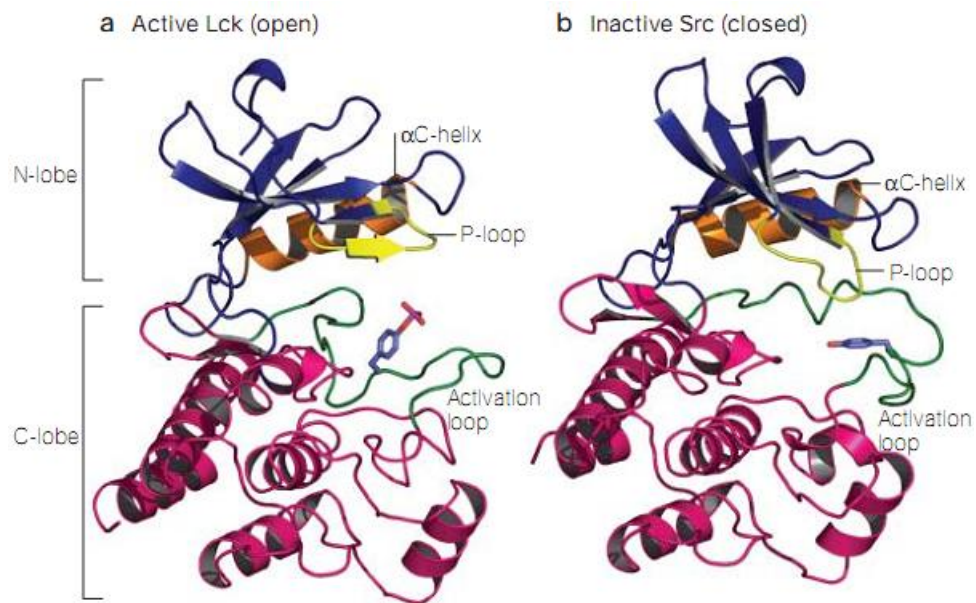
**Figure 4 - The active and inactive forms of two protein kinase domains[12]**

Figure 4 shows the active conformation of the protein kinase Lck, and the inactive conformation of the protein kinase Src. When in the inactive conformation an activation loop, a chain of 20-30 amino acids, is folded into the enzyme active site preventing access through steric hindrance. In the active conformation the activation loop has been extended away from the entrance to the active site which allows access. Activation of the protein kinase occurs through phosphorylation of the activation loop, which stabilises the resulting active conformation. In addition to activating the protein kinase, the activation loop also functions as a binding platform for the protein substrate. Once in the active conformation the protein kinase plays a role in three aspects of the phosphorylation process:-

- Firstly, it must bind a phosphate donor, known as the cofactor or coenzyme.
- Secondly, it must bind a protein substrate.
- Thirdly, it must enable the transfer of a phosphate group between the cofactor and the protein substrate.

The protein kinase active site has two binding sites: one for the cofactor and one for the protein substrate. In order for phosphorylation of the protein substrate to occur, the enzyme must firstly bind a cofactor; without this cofactor phosphorylation would not occur. In the case of protein kinases the cofactor is usually adenosine triphosphate (ATP).

The way in which ATP binds to the enzyme active site of the epidermal growth factor receptor is illustrative of the way that ATP binds to all other kinase active sites (Figure 5).
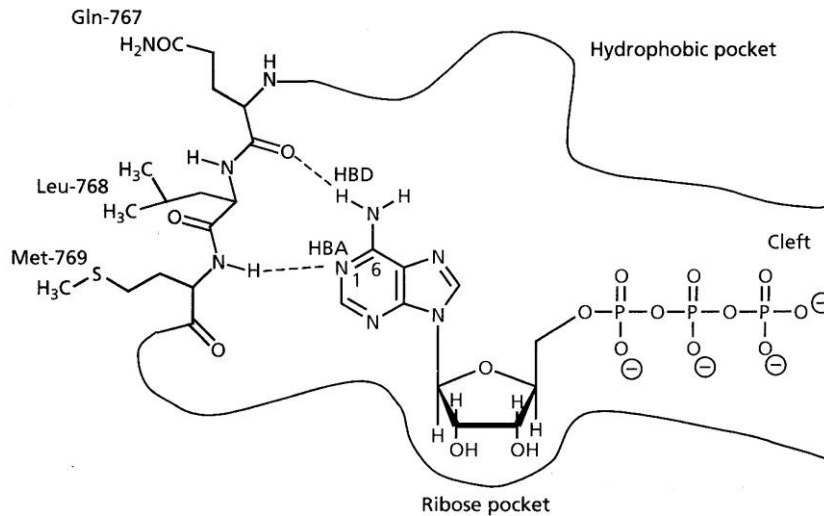
Figure 5 - Binding of ATP to the kinase active site of the epidermal growth factor receptor[11]

The figure demonstrates how the enzyme active site binds ATP in several ways. Towards the back of the active site there are two hydrogen bonds between the amino acid residues of the protein backbone and the adenine functionality of ATP, whilst the ribose sugar rests in a pocket at the base of the active site. The phosphate chain extends out towards the entrance of the enzyme active site, along the cleft, forming a complex with a divalent cation, usually Mg^{2+} or Mn^{2+}, and several hydrogen bonds with amino acid residues. The divalent cation reduces the likelihood of dissociation of the cofactor from the enzyme active site.

Figure 5 also shows a hydrophobic pocket towards the top of the active site opposite the ribose pocket. This pocket is guarded by an amino acid residue known as the "gatekeeper residue."[13] The size of the gatekeeper residue varies between different protein kinases allowing selectivity between different protein substrates. In addition to the selectivity inferred by the gatekeeper, the hydrophobic pocket itself is lined with amino acid residues. These amino acids differ depending on the protein kinase, allowing specific protein substrates to be recognised and bound within the hydrophobic pocket.

The third role the protein kinase has is enabling the transfer of a phosphate group between the cofactor and the protein substrate. The active site of the protein kinase facilitates the transfer by bringing the phosphate chain of the cofactor and the amino acid residue of the protein substrate into close proximity with each other.

There are two main categories of protein kinases which are grouped according to the amino acid side chain, on the protein substrate, that is phosphorylated:-

- The serine-threonine kinases
- The tyrosine kinases

1.2.1 The Serine-Threonine Kinases

The serine-threonine kinases are a class of protein kinase which catalyse the phosphorylation of the hydroxyl group of serine and threonine amino acid residues (Figure 6).
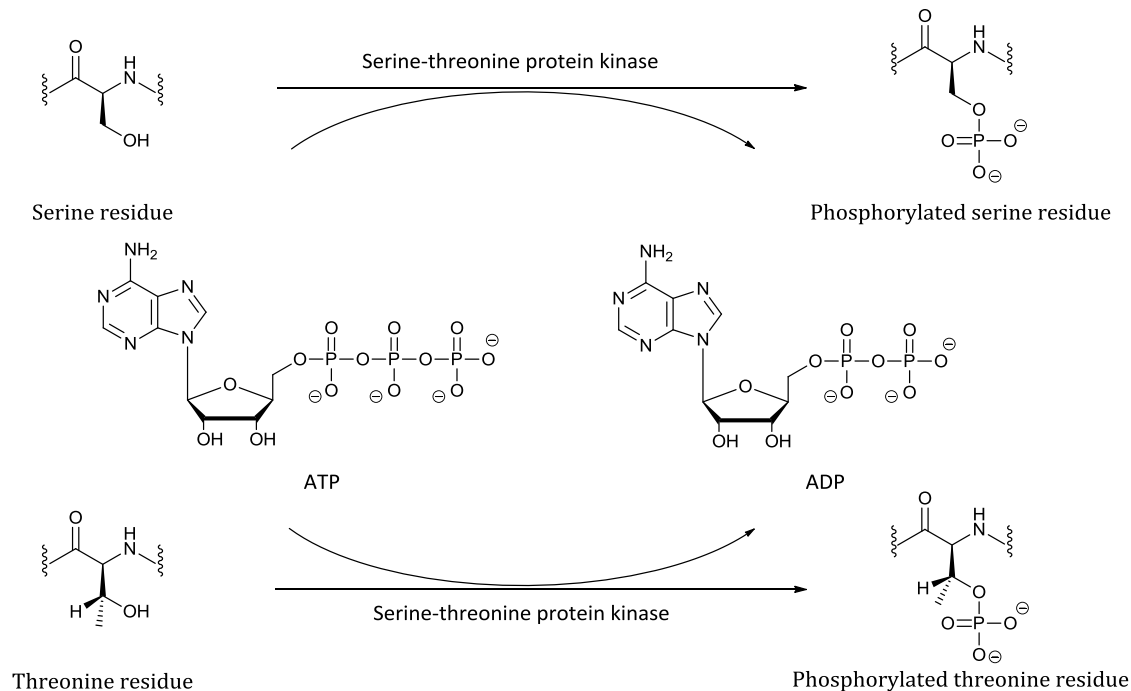
Figure 6 - Phosphorylation of serine and threonine residues

Following the reaction the phosphorylated amino acid residue is now a charged species and has increased in size due to the phosphate group. This leads to a conformational change in the protein substrate which then goes on to regulate a cellular process. In the process of phosphorylating the protein substrate, the cofactor, ATP, loses a phosphate group becoming adenosine diphosphate, ADP. ATP can be regenerated through metabolic processes and also by an enzyme called phosphatase. Phosphatase has the opposite effect to a protein kinase as it removes phosphate groups from amino acid residues, acting as an "off switch" to the protein kinases "on switch."

1.2.2 The Tyrosine Kinases

The tyrosine kinases phosphorylate tyrosine amino acid residues on protein substrates (Figure 7).
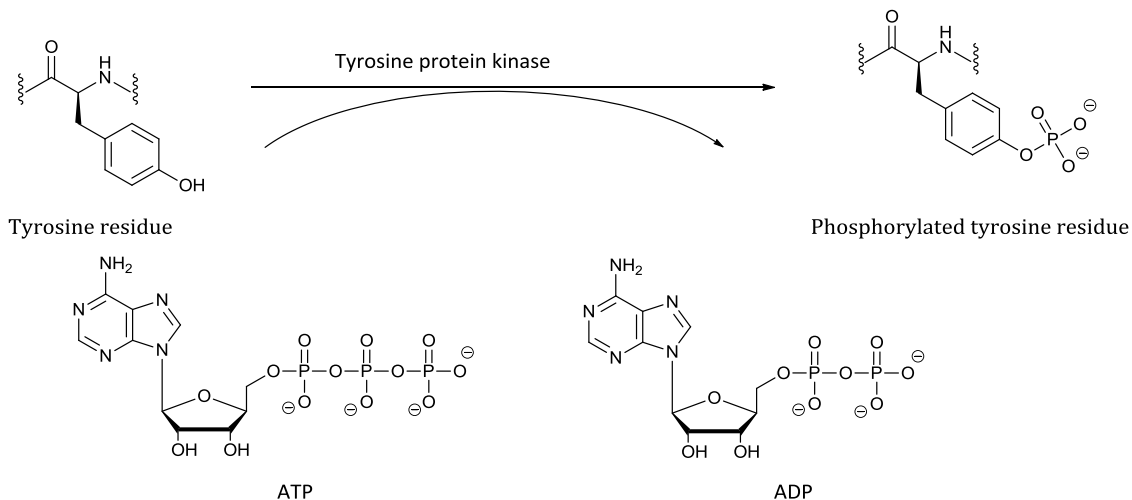
Figure 7 - Phosphorylation of tyrosine residue

This phosphorylation causes an effect on cellular processes within the body. Mutation or deregulation of a protein kinase can have serious consequences within the body, leading to diseases such as: endocrine disorders, cardiovascular diseases, immunodeficiencies and cancer. For this reason there is great interest in the area of protein kinases for the treatment of disease.

One of the areas of interest for research is cancer and there are currently several drugs on the market which act on protein kinases. The first drug developed to specifically inhibit one enzyme was Imatinib, marketed as Glivec (UK) and Gleevec (USA), which is a tyrosine kinase inhibitor (Figure 8).
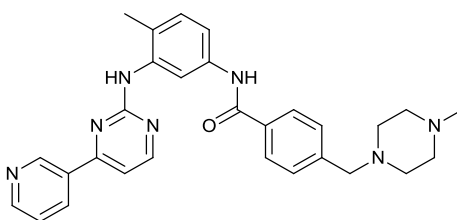

Imatinib

Figure 8 - Imatinib

Imatinib was developed through rational drug design by Nicholas Lydon, Brian Druker and Charles Sawyers at the end of the 20th century. It is used in the treatment of chronic myelogenous leukaemia, a relatively rare cancer affecting approximately 600 people per year in the UK. Chronic myelogenous leukaemia is caused by the Philadelphia

chromosome, an abnormal gene formed by the translocation of chromosomes 9 and 22. This translocation fuses the Bcr (breakpoint cluster region) and Abl (Abelson leukaemia virus) genes, creating the fusion protein Bcr-Abl which produces an abnormal tyrosine kinase. The tyrosine kinase is permanently active causing excessive production of abnormal white blood cells through increased mitosis and prevention of apoptosis. Imatinib acts as an ATP mimic, competitively binding to the ATP binding site on the Bcr-Abl protein thereby inhibiting enzyme activity and preventing the transduction of energy signals necessary for cell growth and death (Figure 9).
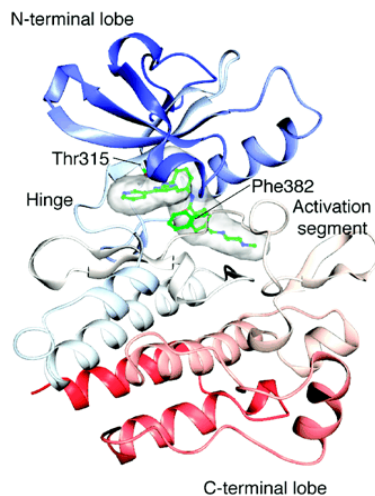

The structure of the catalytic domain of cAbl in complex with Gleevec. The N-terminal lobe consists of a ß sheet and one conserved {alpha} helix (helix C). The C-terminal lobe is largely helical and contains a segment, the activation segment, which includes residue(s) that in many kinases are phosphorylated for activity. The hinge region connects the two lobes. The protein structure is color ramped so that residues close to the N terminus are blue, and those close to the C terminus are red. Gleevec is shown bound to the ATP-binding site, from which it extends under the C helix. Thr315, the "gatekeeper" residue, and Phe382, the conserved phenylalanine that marks the beginning of the activation segment, are labeled.

Figure 9 - Binding of Gleevec[14]

Treatment of early stage chronic myeloid leukaemia with Imatinib has resulted in remission for nearly 100% of cases.

Although none of the approved drugs targeting protein kinases are natural product derived compounds, there are currently several in clinical trials. These include the isoflavone genistein, alvocidib and enzastaurin (Figure 10).

10
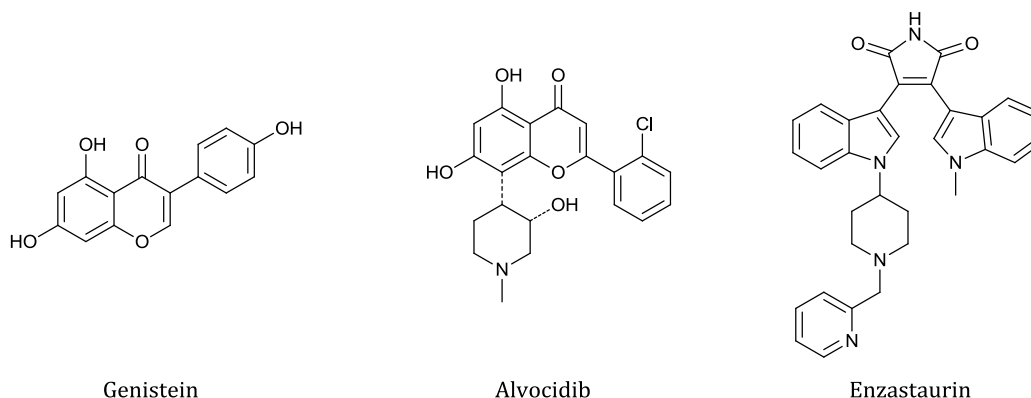
Figure 10 – Kinase targeted drugs developed from natural products

Genistein is a tyrosine kinase inhibitor which is currently undergoing clinical trials for breast, prostrate, brain and cervical cancer. Alvovidib is derived from the natural product rohitukine, an alkaloid isolated from the stem bark of *Dysoxylum binectarifum*. It is a cyclin-dependent kinase inhibitor which is currently in clinical trials for the treatment of chronic lymphocytic leukaemia. Enzastaurin was developed from the natural product staurosporine, an alkaloid isolated from the bacteria *Streptomyces staurosporeus*. It is a serine-threonine kinase inhibitor and is currently undergoing phase II and III clinical trials for the treatment of cancer.

Three other natural products which have shown anticancer activity are ulocladol A (**1.1**), alternariol 9-methyl ether (**1.2**) and alternariol (**1.3**). Ulocladol A has shown tyrosine kinase inhibitory activity and alternariol and alternariol 9-methyl ether have both shown activity against KB and KBv200 cell lines (Figure 11).

**1.1**

1.2 R = OMe
1.3 R = OH
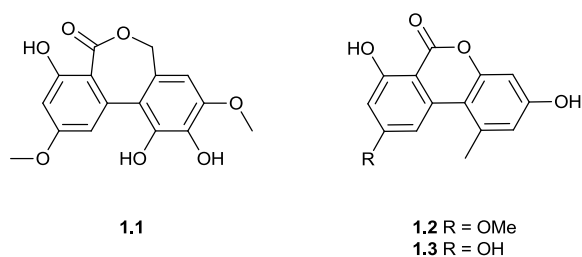
Figure 11 – Structures of ulocladol A, alternariol 9-methyl ether and alternariol

1.3 Ulocladol A

1.3.1 Isolation and Biological Activity

In 1999 an ethyl acetate extract of the fungal culture of *Ulocladium botrytis*, isolated from the marine sponge *Callyspongia vaginalis*, showed tyrosine kinase inhibitory activity and antimicrobial activity.[15] Bio-assay guided work up by VLC and HPLC led to the isolation of ulocladol A and the anti-fungal metabolite **1.4** (Figure 12).
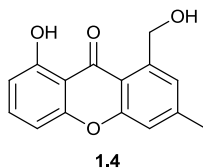
Figure 12 – 1-Hydroxy-6-methyl-8-(hydroxymethyl)xanthone

The anti-fungal metabolite **1.4** was identified as 1-hydroxy-6-methyl-8-(hydroxymethyl) xanthone, a compound previously isolated by Ayer and Taylor in 1976 and Hein *et al.* in 1998.[16, 17] Ulocladol A was found to exhibit tyrosine kinase inhibitory activity against p56[lck] with a reduction in enzyme activity to 7% at 0.02 μg/μL.

Ulocladol A also adheres to Lipinski's 'Rule of Five,' which states that poor oral absorption or permeation is more likely when:-

- There are more than 5 H-bond donors
- The molecular weight is over 500
- The Log P is over 5
- There are more than 10 H-bond acceptors

Ulocladol A was found to have a biaryl core linked by a seven membered lactone ring, as shown in Figure 11. It has three hydrogen bond donor groups and seven hydrogen bond acceptor groups; a molecular weight of 318 Daltons and a Log P which is predicted to be 1.81. This suggests that ulocladol A would be a good lead for further investigation.

It is likely that ulocladol A is subject to axial chirality, due to the restricted rotation about the aryl-aryl bond, resulting in the formation of atropisomers. Atropisomerism is typically found in *ortho* substituted biaryl systems where steric congestion restricts free rotation about the carbon-carbon biaryl bond.[18] Extensive studies of atropisomers have found that it is possible to resolve the different isomers if the biaryls are tetra-*ortho*- substituted,

although this is less likely if two of the substituents are small. Tri-*ortho-* substituted biaryls can be interconverted, although not usually at room temperature and finally, di-*ortho* substituted biaryls can usually only be resolved if the substituents are large. In the case of ulocladol A the *ortho* substituents are not particularly large, but they are linked through the lactone ring and it is unknown how this will affect the stereochemistry of the compound.

The exact conformation of the natural product has not been determined and it is uncertain whether it is racemic or one specific atropisomer. In a laboratory synthesis of compounds of this nature, it is possible to control stereochemistry depending on the reagents and route involved. If a palladium-mediated coupling reaction was applied, axial chirality can be controlled through the use of chiral catalysts and ligands. However, due to the unknown stereochemistry of ulocladol A, we will not initially focus on preparing a specific atropisomer and instead will use a racemic route.

1.3.2 Predicted Interaction of Ulocladol A with Enzyme Active Site

The industrial sponsor of the project provided a model of ulocladol A docked into the active site of the enzyme Lck.[19] This model shows some interesting interactions (Figure 13).

a   b
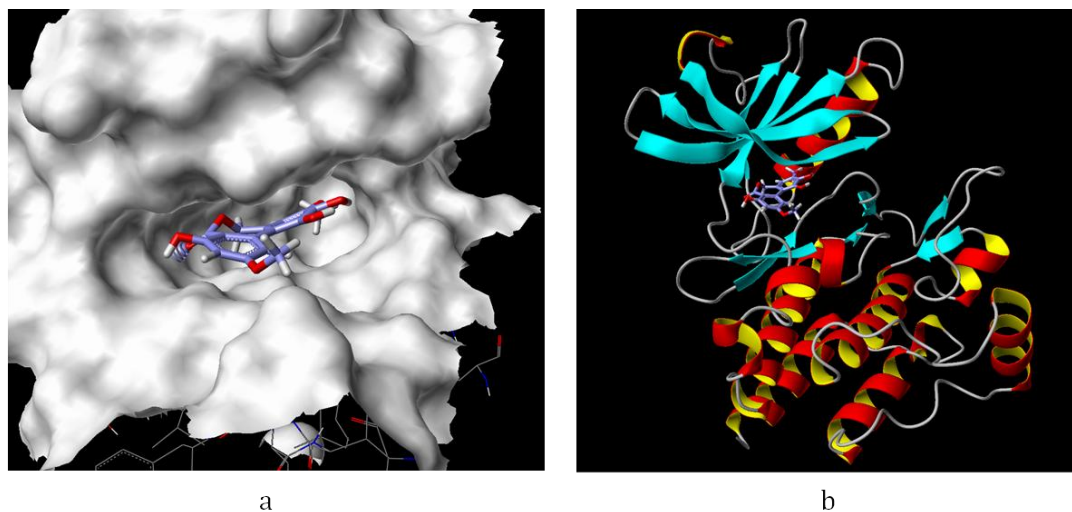
Figure 13 – Predicted interaction of ulocladol A with Lck binding site

Figure 13a shows the spatial orientation of ulocladol A in the binding pocket of the Lck active site. The right hand ring of ulocladol A with the dihydroxymethoxy phenyl moiety points into the back of the pocket, presumably due to interactions with hydrogen bond
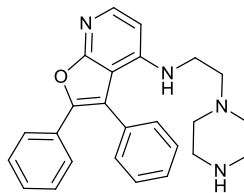
acceptor sites. The carbonyl of the lactone ring forms a hydrogen bond with the "hinge" of the active site whilst the left hand ring is towards the periphery of the binding pocket. Figure 13b shows a cartoon representation of the Lck enzyme. The active site can be observed towards the top of the model, located in the space between the hinged β-sheet and the main body of the enzyme made up of α-helices. It is also possible to see the carbonyl of the lactone ring pointing towards the inside hinge of the enzyme. The predicted interactions can be seen more clearly in an expanded view of the active site (Figure 14).[19]
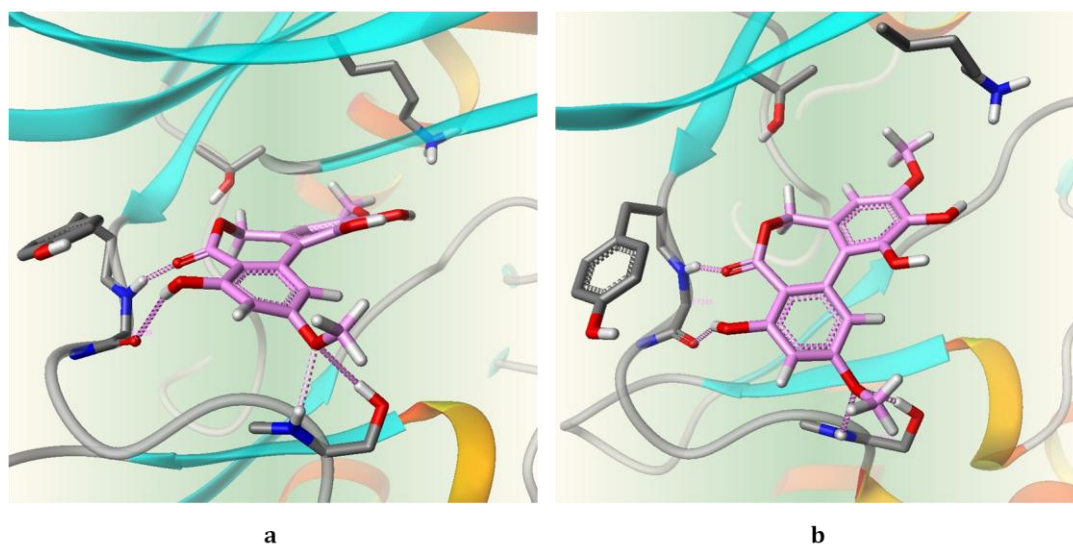
Figure 14 – The front view (a) and top view (b) of the predicted interactions of ulocladol A within the active site of Lck

Figure 14 shows the expected hydrogen bonding interaction between the lactone carbonyl and the hydrogen atom on the nitrogen of the tyrosine amino acid residue. It also shows three additional predicted hydrogen bonding interactions, between the phenolic and methoxy moieties on the left hand ring and the enzyme active site, further stabilising the binding of ulocladol A. With this is mind, it was proposed that the substitution of the right hand ring would provide further scope for improving the activity of any analogues of ulocladol A, whilst substitution of the left hand ring would be of less significance due to the predicted interactions already in place.

A paper by Martin *et al*. details the synthesis of a novel series of furanopyrimidines which were also found to be inhibitors of Lck.[20] One of the best compounds was 2,3-diphenyl-N-(2-(piperazin-1-yl)ethyl)furo[2,3-b]pyridin-4-amine (**1.5**) (Figure 15).

1.5

Inhibitor **1.5** was found to be a potent inhibitor of Lck with an IC_{50} of 0.21 µM. A comparison of an X-ray crystal structure of **1.5** bound to the active site of Lck with the X-ray structure of ulocladol A bound to the active site of Lck, shows that the two molecules appear to sit in the same pocket (Figure 16).[21]
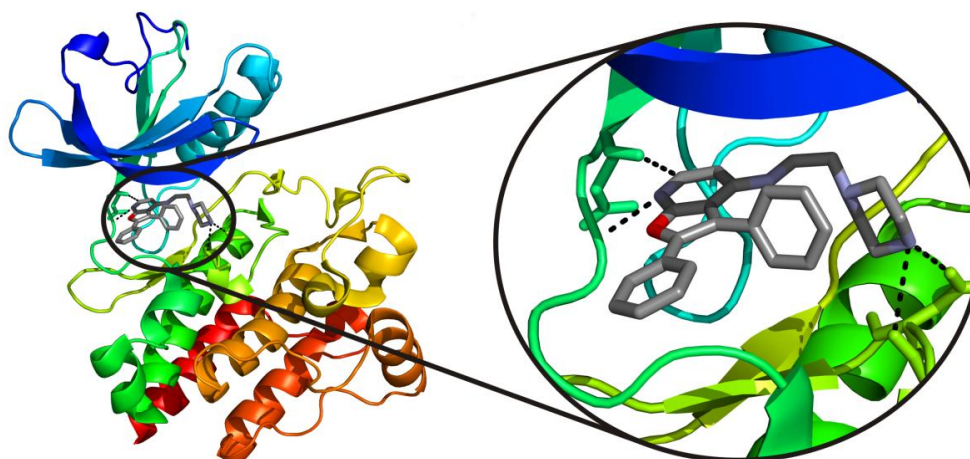
Figure 16 – Predicted X-ray structure of 1.5 bound to Lck binding site

The compound sits in the hinge of the active site in the same orientation as ulocladol A. The nitrogen in the pyridine ring points into the left hand side of the pocket and appears to interact with the same site as the ester in the lactone ring of ulocladol A. In addition, the piperizine ring points to the front right of the pocket in the same direction that the phenolic groups on the right hand ring of ulocladol A point.

1.3.3 Synthetic Approaches to Ulocladol A

There have been four total syntheses of ulocladol A, three of which were reported by Abe *et al.* in a paper published in 2006.[22] Abe described three routes to ulocladol A: one involved the demethylation of a hydroxyl group on another, previously synthesised natural product; one required a benzyl deprotection and the third a ring opening and closing

procedure. The three syntheses required the use of intermediates reported in an earlier synthesis of a group of natural products called graphislactones.

1.3.3.1 Synthesis of Graphislactone D Followed by Selective Demethylation

In 2005 Abe *et al.* reported the 12 step synthesis of graphislactone D (**1.19**) (Scheme 1).[23]
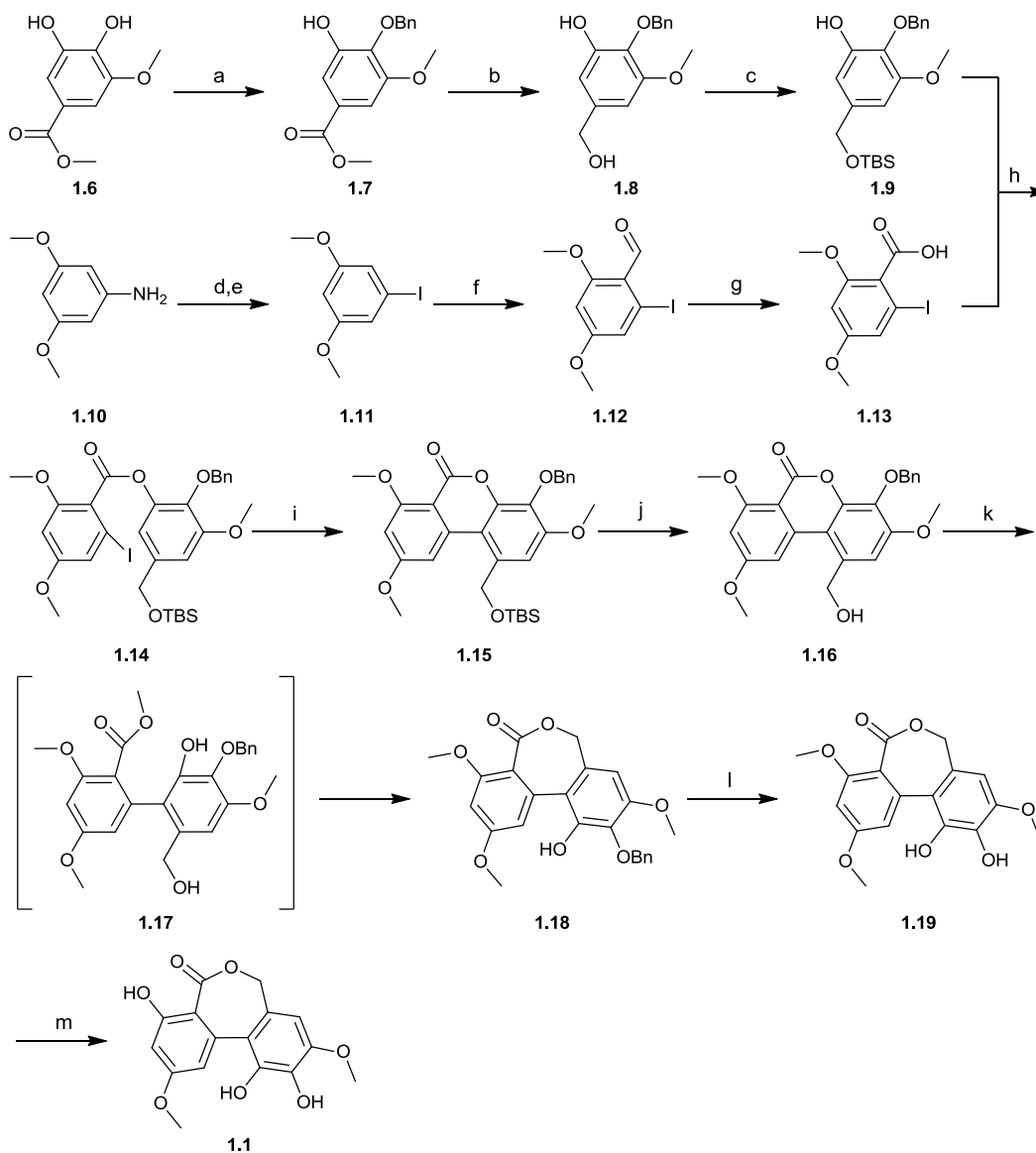
Reagents and Conditions: a) BnBr, K_2CO_3, DMF, 70%; b) LiAlH$_4$, THF, 91%; c) TBSCl, imidazole, CH_2Cl_2, 71%; d) NaNO$_2$, conc HCl, H_2O; e) KI, H_2O, 55%; f) POCl$_3$, DMF, 64%; g) 30% H_2O_2, 80% NaClO$_2$, NaH$_2$PO$_4$, CH$_3$CN, H_2O, 98%; h) EDC, DMAP, CH_2Cl_2, 61%; i) Pd(OAc)$_2$, *n*-Bu$_3$P, K_2CO_3, DMA, 85%; j) TBAF, THF, 90%; k) K_2CO_3, MeOH, 85%; l) H_2, 10% Pd/C, EtOAc, 58%; m) BBr$_3$, CHCl$_2$, 21-73%
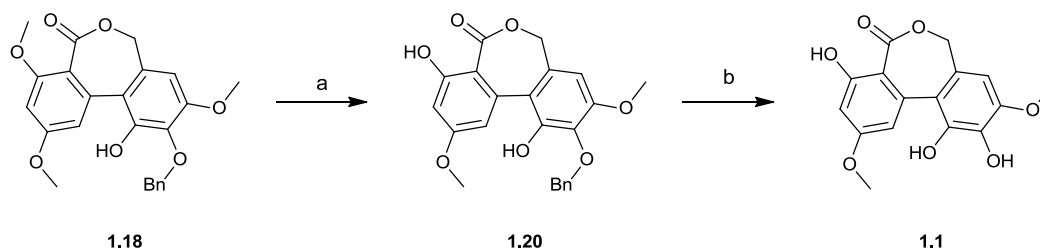
Scheme 1

Commercially available methyl 3,4-dihydroxy-5-methoxybenzoate (**1.6**) was selectively benzyl protected and then reduced to prepare benzyl alcohol **1.8**, which was then protected with a silyl protecting group to afford **1.9**.

Aromatic substitution of 3,5-dimethoxyaniline (**1.10**) followed by the introduction of an aldehyde using the Vilsmeier reaction afforded **1.12**. Oxidation using hydrogen peroxide yielded **1.13** which then underwent esterification with **1.9** to furnish ester **1.14**. Palladium-mediated, intramolecular, biaryl coupling of **1.14** yielded the six-membered lactone **1.15**. Silyl deprotection of **1.15** followed by treatment with methanolic potassium carbonate resulted in the ring reconstruction of **1.16**, via a ring opened intermediate **1.17**, to yield the seven-membered lactone **1.18**. Benzyl deprotection of **1.18** afforded graphislactone D (**1.19**) in 4% overall yield.

The following year Abe reported the selective demethylation of graphislactone D (**1.19**) with boron tribromide to afford ulocladol A in 21-73% yield. Despite best efforts, Abe was unable to reproduce the high yields in repeat reactions.

1.3.3.2   Synthesis by Selective Demethylation and Deprotection of Intermediate 1.18

Due to the difficulties observed following the demethylation of **1.19** to afford ulocladol A, Abe reversed the order of demethylation and debenzylation (Scheme 2).
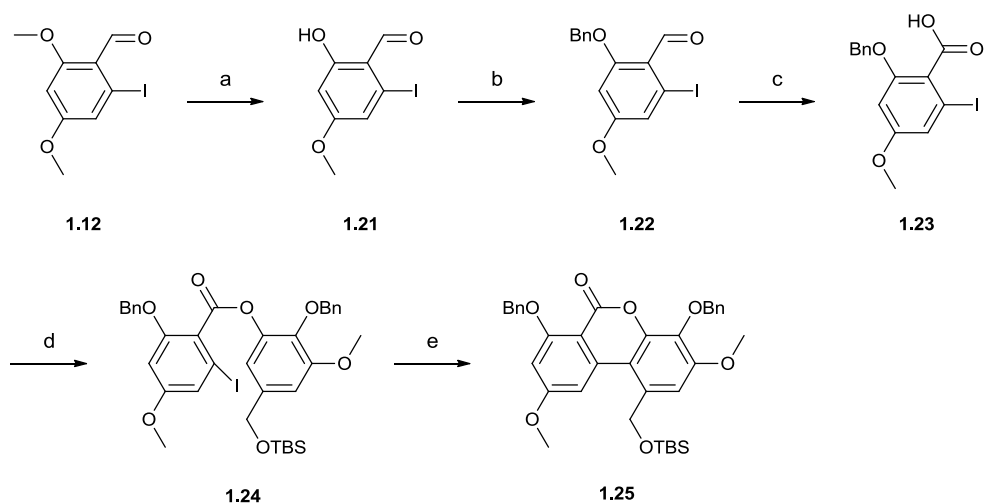

Reagents and Conditions: a) AlCl$_3$, NaI, CH$_2$Cl$_2$-CH$_3$CN, 83%; b) H$_2$, Pd/C, EtOAc, 60%

Scheme 2

Using the Lewis acid aluminium chloride, Abe selectively demethylated **1.18** in 83% yield to afford benzyl protected **1.20**. Standard hydrogenolysis protocol yielded ulocladol A (**1.1**) in 60% yield.

### 1.3.3.3   Synthesis From 2-Iodo-4,6-dimethoxybenzalydehyde

The third method reported by Abe *et al.* involved intermediate **1.26**, which was prepared by the group in a previously reported synthesis of graphislactone C (Scheme 3).[23]

Reagents and Conditions: a) AlCl$_3$, NaI, CH$_3$CN, CH$_2$Cl$_2$, 60%; b) BnBr, K$_2$CO$_3$, DMF, 87%; c) 30% H$_2$O$_2$, 80% NaClO$_2$, NaH$_2$PO$_4$, CH$_3$CN, H$_2$O, 98%; d) **1.9**, EDC, DMAP, CH$_2$Cl$_2$, 65%; e) Pd(OAc)$_2$, *n*-Bu$_3$P, K$_2$CO$_3$, DMA, 90%

Scheme 3

Aldehyde **1.12**, prepared as an intermediate in the synthesis of graphislactone D, underwent selective cleavage of one of the methoxy groups *ortho* to the aldehyde, with AlCl$_3$ in 60% yield, to liberate a hydroxyl group which was then protected with a benzyl protecting group, **1.22**, in 87% yield. The aldehyde was then oxidised to the carboxylic acid **1.23** in 98% yield. Condensation of carboxylic acid **1.23** and phenol **1.9** led to the formation of ester **1.24** in 65% yield. Palladium mediated biaryl coupling gave **1.25** in 90% yield. Although not reported in either of the papers associated with the synthesis of graphislactone C or ulocladol A published by Abe *et al.*, intermediate **1.25** underwent deprotection of the silyl protecting group to yield **1.26**. Intermediate **1.26** required a reconstruction of the six-membered lactone ring (Scheme 4).

18
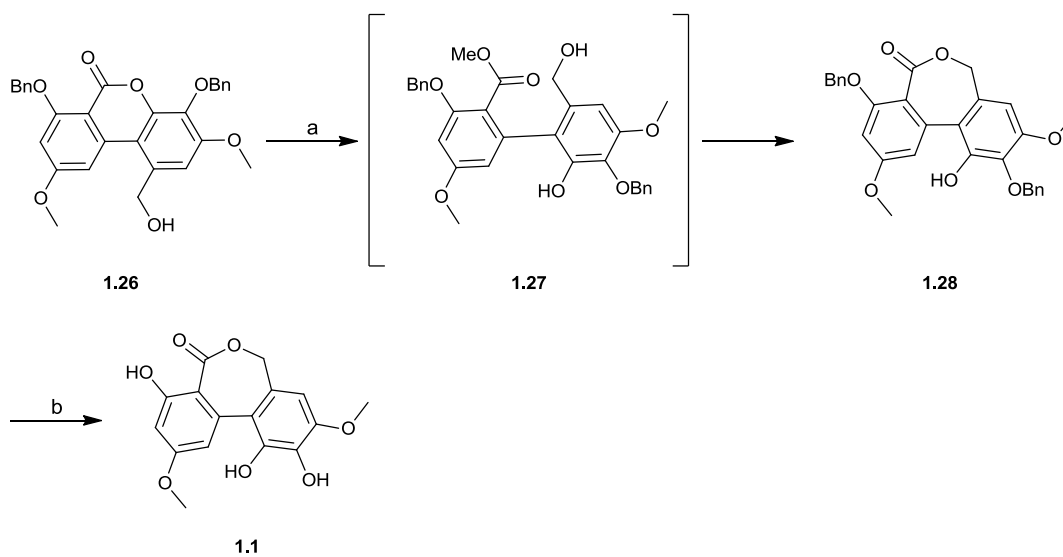
Reagents and Conditions: a) K_2CO_3, MeOH, 82%; b) H_2, Pd/C, EtOAc, 63%

Scheme 4

Reconstruction of the six-membered lactone ring **1.26**, using potassium carbonate in methanol, resulted in the formation of a seven-membered lactone ring **1.28** via the intermediate **1.27** in 82% yield. Standard hydrogenolysis of the benzyl protecting groups furnished ulocladol A in 63% yield.

1.3.3.4 Synthesis From 2,5-Dihydroxy-2,2-dimethyl-benzo[1,3]dioxin-4-one

In the final six months of this project, Podlech *et al.* reported a total synthesis of ulocladol A.[24] The reported synthesis detailed much of the work that was currently being undertaken and the effect of this will be discussed in greater detail in subsequent chapters. However, the reported work only details the synthesis of ulocladol A, with no reference to the potential for synthesis of analogues (Scheme 5).
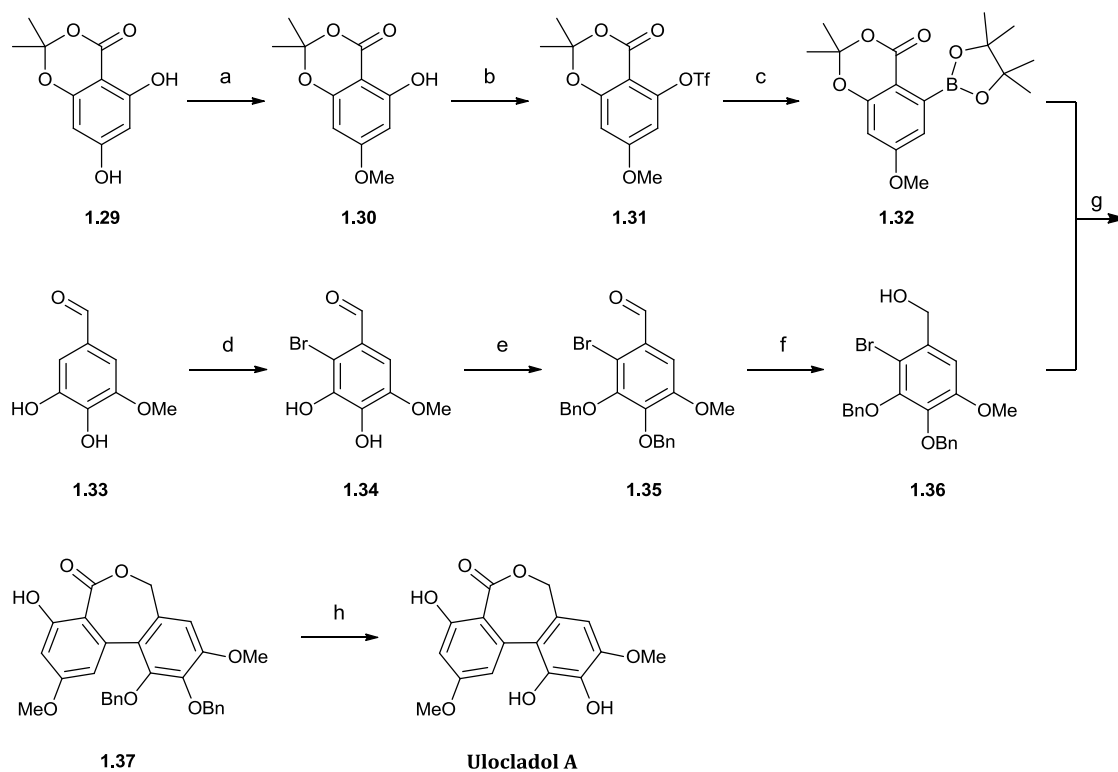
Reagents and conditions: a) MeOH, PPh$_3$, DIAD, THF, 0 °C, 2 h, 78%; b) Tf$_2$O, pyridine, 0 °C, 12 h, 95%; c) Et$_3$N, Pd(PPh$_3$)$_4$, pinacolborane, dioxane, 80 °C, 2 h, 75%; d) Br$_2$, AcOH, rt, 1 h, 75%; e) NaH, BnBr, DMF, -78 °C, 15 min, 50%; f) DIBAL-H, THF, -78 °C, 2 h, 99%; g) Pd(OAc)$_2$, SPhos, Cs$_2$CO$_3$, dioxane/H$_2$O (6:1), 80 °C, 2 h, 65%; h) Pd/C (10-15 mol %), H$_2$, MeOH, rt, 2 h, 60%

Scheme 5

Due to the reported work being published at a time when very similar work was underway in our laboratory, the published route will be discussed in context with the work carried out in this project.

1.4 Alternariol and Alternariol 9-Methyl Ether

1.4.1 Isolation and Biological Activity

Alternariol (**1.3**) and alternariol 9-methyl ether (**1.2**) are secondary metabolites of the *Alternaria* fungi, first isolated by Raistrick *et al*. in 1953.[25] Raistrick *et al.* successfully determined the structure of alternariol, but were unable to conclusively determine the position of the methyl ether in alternariol 9-methyl ether. It wasn't until 1961 that the structure of alternariol 9-methyl ether was deduced by Thomas through a series of analytical and exploratory experiments.[26]

The two natural products have caused significant problems agriculturally through crop fouling. In addition, both alternariol and alternariol 9-methyl ether have shown anti-cancer potential, inhibiting KB and KBv200 cell lines (Table 1).

	KB (IC_{50}, µg/mL)	KBv200 (IC_{50}, µg/mL)
Alternariol 9-methyl ether	4.82	4.94
Alternariol	3.17	3.12

Table 1 – Biological activity of alternariol 9-methyl ether and alternariol

1.4.2 Synthetic Approaches

Since their isolation in 1953 there have been several total syntheses of alternariol and alternariol 9-methyl ether. In total, alternariol has been prepared four times and alternariol 9-methyl ether has been prepared twice.[27-30]

#### 1.4.2.1 Synthesis From Methyl Orsellinate

The first synthesis of alternariol was reported in 1977 by Harris *et al*. and involved the use of a biomimetic route.[27] A biomimetic route is a synthetic route to a molecule which aims to imitate a reaction pathway found in nature. Harris *et al.* utilise a synthetic route based on the acetate pathway used in nature to synthesise polyketides and fatty acids; these are essential precursors in the formation of many compounds, including prostaglandins and anthraquinones.[31] In nature, the first step of the acetate pathway is the conversion of acetyl-CoA to the more reactive malonyl-CoA, through a carboxylation reaction (Scheme 6).
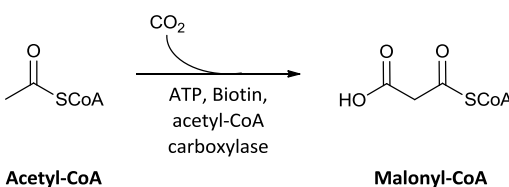
Scheme 6

The reaction is catalysed by the enzyme acetyl-CoA carboxylase and requires the presence of ATP, CO_2, in the form of HCO_3, and biotin, the coenzyme. Malonyl-CoA is a more reactive nucleophile than acetyl-CoA due to the increased acidity of the α-methylene protons, enabling the acetate pathway to occur (Scheme 7).
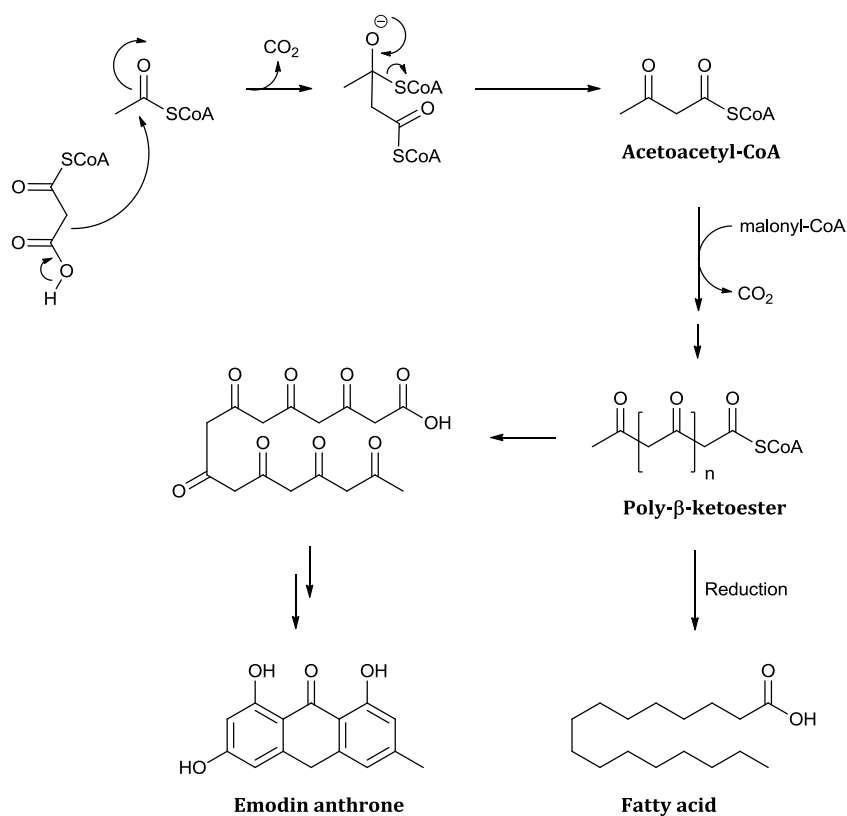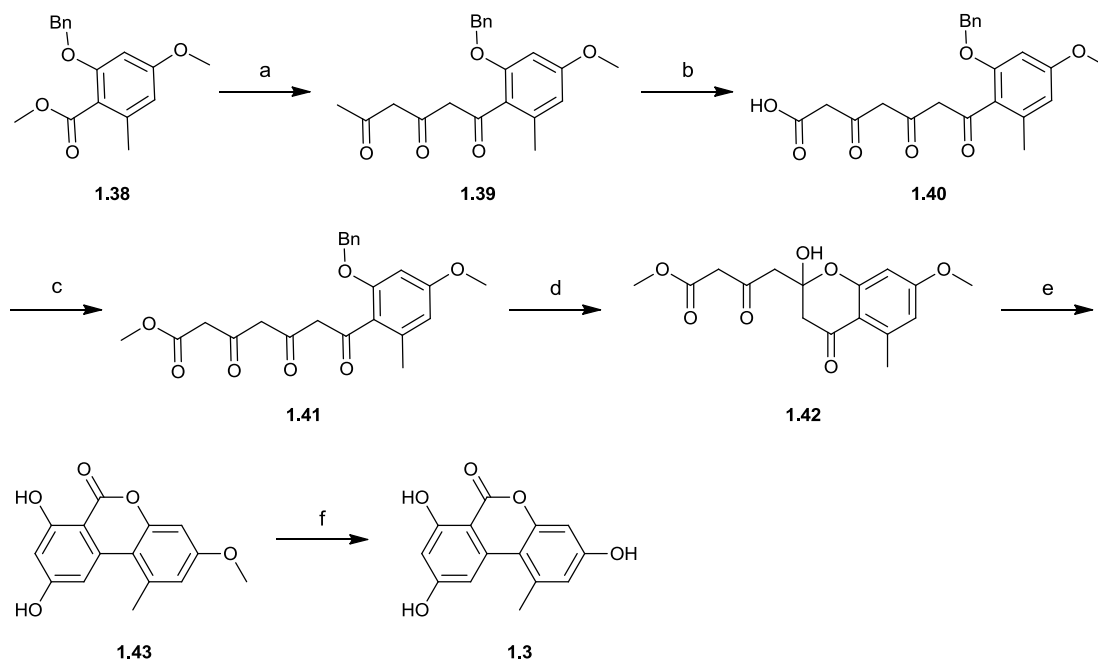
Scheme 7

Following the synthesis of malonyl-CoA, the second step is a Claisen type condensation reaction with acetyl-CoA, furnishing acetoacetyl-CoA. Whilst this reaction is thermodynamically unfavourable, it is pushed to completion by the release of CO_2. Repetition of this condensation reaction results in the formation of poly-β-ketoesters, which can undergo further reaction leading to fatty acids and polycyclic compounds such as emodin anthrone.[32]

Harris *et al.* were able to successfully apply a biomimetic route, inspired by the acetate pathway, to the synthesis of alternariol, isolating the product in six steps (Scheme 8).[27]

Reagents and Conditions: a) Dilithioacetylacetone, THF, 88%; b) Lithium diisopropylamide, THF, CO_2, HCl, 19%; c) CH_2N_2, Et_2O, 95%; d) 10 % Pd/C, EtOH, H_2, 86%; e) 1 M methanolic NaOAc, 64%; f) HI, 90%

Scheme 8

Starting from methyl orsellinate (**1.38**), Harris *et al*. used dilithioacetylacetone to extend the ester to a carbonyl chain with six carbon atoms. This chain was extended again and oxidised to form carboxylic acid **1.40**. Esterification followed by hydrogenation of a methoxy group led to **1.42** in 86% yield. Reaction of **1.42** with a 1 M methanolic sodium acetate solution yielded the precursor to alternariol **1.43** in 64% yield. Demethylation of **1.43** with HI furnished the desired product alternariol (**1.3**) in 90% yield.

1.4.2.2 Synthesis From 2,4-Dinitroaniline

A second synthesis published in 1977 by Sóti *et al.* reported the use of the Hurtley reaction to couple the left and right hand rings (Scheme 9).[28]
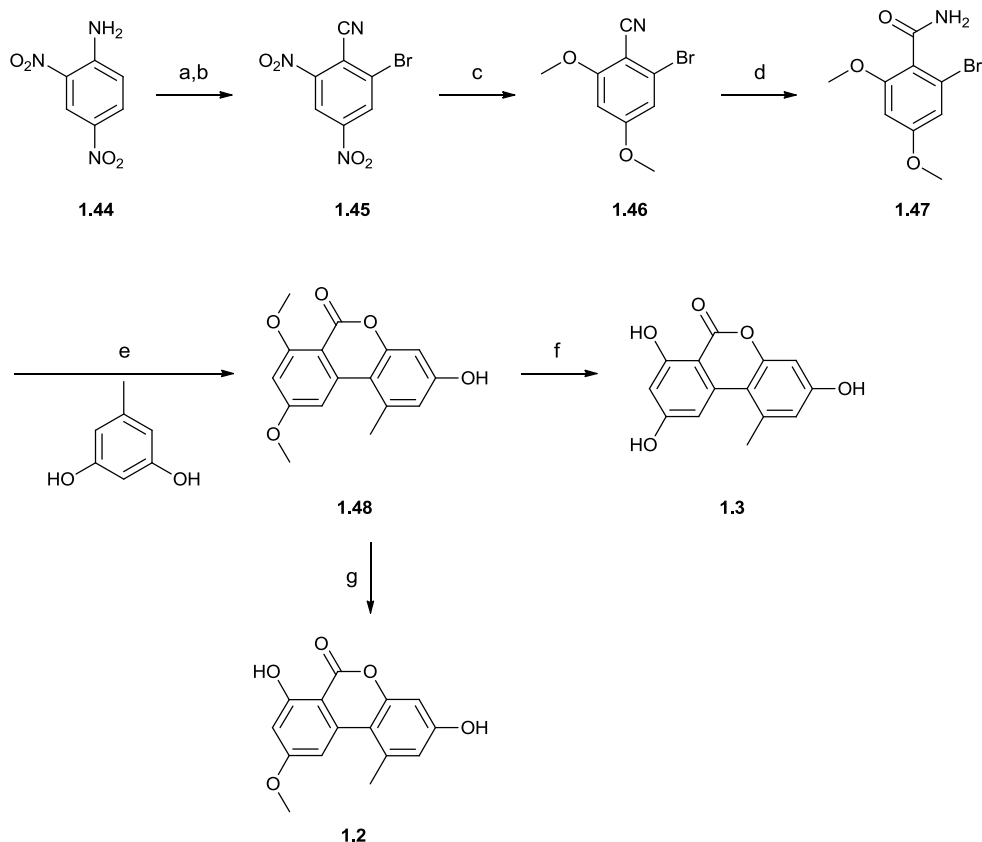
Reagents and Conditions: a) Br$_2$, 95% acetic acid, rt; b) HNO$_2$/K$_2$[Cu(CN)$_4$], <20 °C, 30 min, 13% over two steps; c) Na, MeOH, Δ, 16 h, 95%; d) KOH, H$_2$O:EtOH, 125 °C in an autoclave, 18 h, 82%; e) 1M NaOH, 1% CuSO$_4$ solution, Δ, 4 h followed by 2M HCl, Δ, 30 min, 25%; f) HI, Δ, 4 h, 81%; f) HBr, acetic acid, Δ, 10 min, 73%

Scheme 9

From 2,4-dinitroaniline (**1.44**), Sóti *et al.* used the Sandmeyer reaction to prepare the brominated benzonitrile compound **1.45**. Sodium methoxide was used to furnish the desired dimethoxy product **1.46** which was then hydrolysed to the amide **1.47** using a solution of KOH in 1:1 water: ethanol. The precursor to alternariol 9-methyl ether (**1.2**) and alternariol (**1.3**), **1.48**, was prepared by the Hurtley reaction, coupling the amide **1.47** with orcinol, although in poor yield. Complete demethylation using HI furnished alternariol (**1.3**) in 81% yield, whilst incomplete demethylation using HBr furnished alternariol 9-methyl ether in 73%. This linear route led to the isolation of alternariol 9-methyl ether and alternariol, each in five steps and approximately 2% overall yield.

1.4.2.3 Synthesis From 2,4-Dimethyoxy-6-methylbenzaldehyde

The third synthesis of alternariol (**1.3**) was reported by Rao *et al.* in 1990 (Scheme 10).[29]
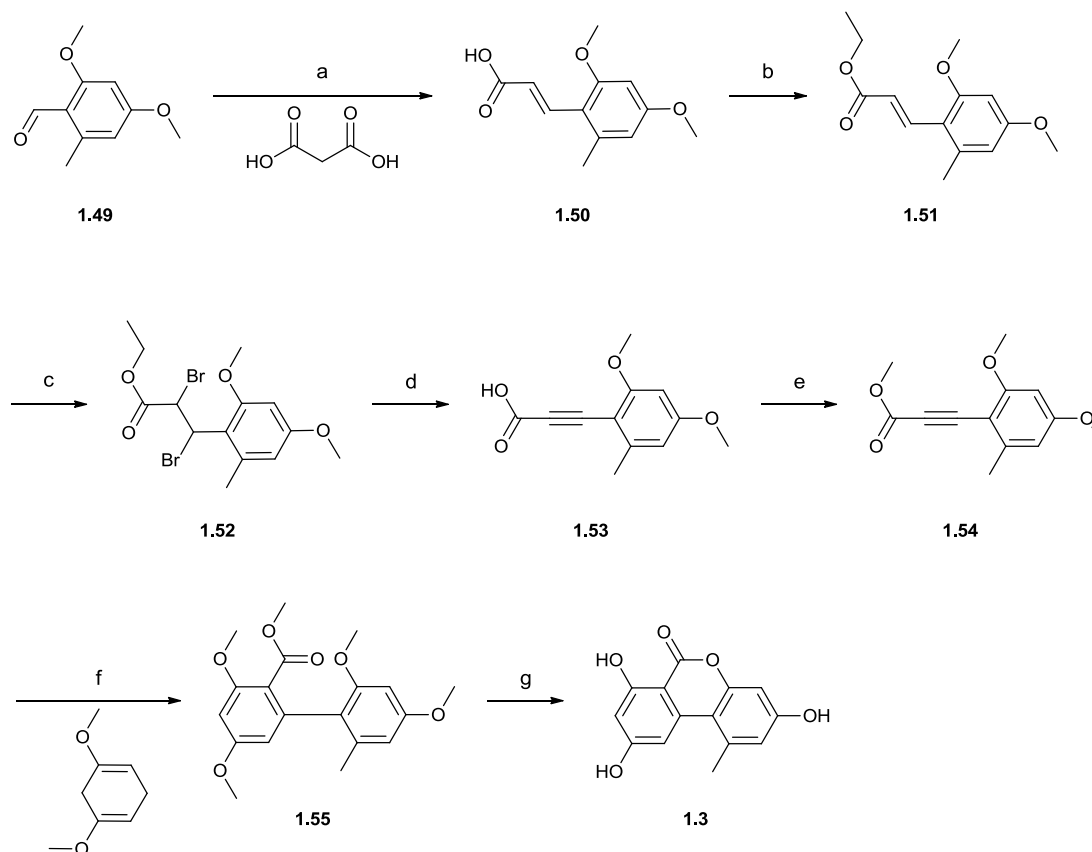
Reagents and Conditions: a) pyridine, piperidine, 100 °C, 15 h, 70%; b) EtOH, conc. H_2SO_4, Δ, 6 h, 80%; c) Br_2, CCl_4, hv, 0 °C, 3 h, 80%; d) KOH, H_2O, dioxane, EtOH, 100 °C, 7 h, 80%; e) MeOH, NaOH, Me_2SO_4, rt, 1 h, 60%; f) 180 °C, 30 h then 220 °C, 4 h, 35%; g) BBr_3, DCM, rt, 10 h, 67%

Scheme 10

Rao *et al.* began the synthesis with the commercially available 2,4-dimethyoxy-6-methylbenzaldehyde (**1.49**), converting to the cinnamic acid **1.50** by refluxing with malonic acid and pyridine. Esterification of the acid, followed by dibromination of the alkene afforded **1.52** in 64% over the two steps. Subsequent dehydrobromination using aqueous KOH furnished the alkyne **1.53** in 80% yield. Esterification of the free acid **1.53** followed by a cycloaddition reaction with 1,4-dimethoxycyclohexa-1,5-diene afforded the precursor to alternariol, **1.55**. Demethylation using BBr_3 afforded the desired product, alternariol, in seven steps and 5% overall.

1.4.2.4 Synthesis From Orcinol

The most recent synthesis of alternariol 9-methyl ether and alternariol was a seven step convergent synthesis reported by Podlech *et al.* in 2005 (Scheme 11).[30]
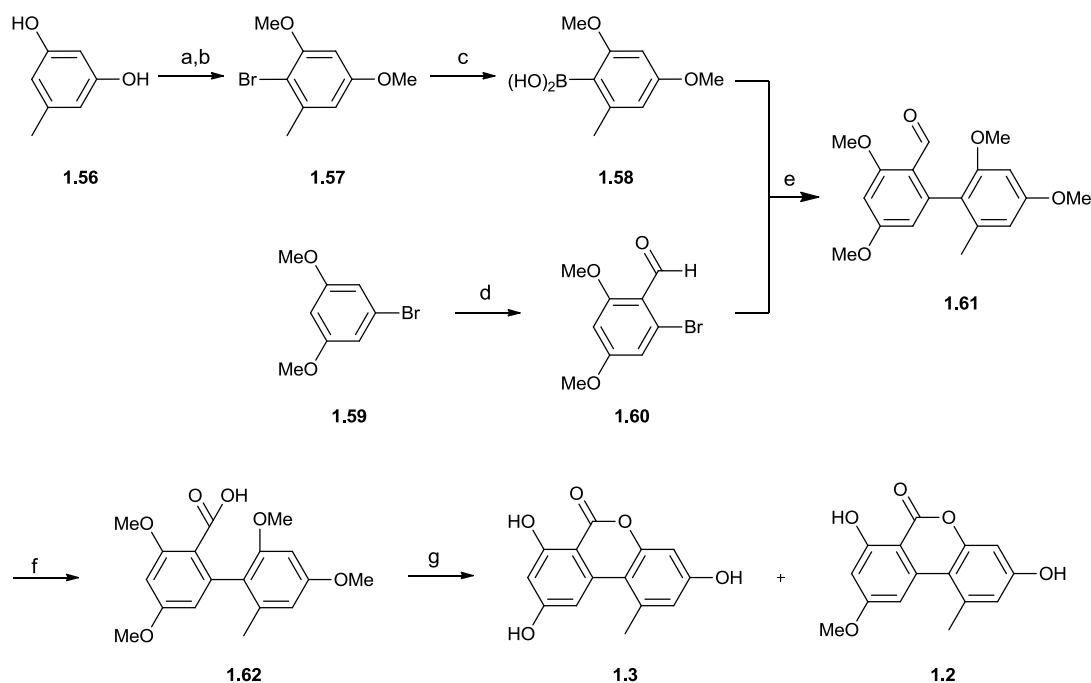
Reagents and Conditions: a) Me_2SO_4, K_2CO_3, acetone, 99%; b) NBS, $CHCl_3$, 99%; c) n-BuLi, $B(Oi\text{-Pr})_3$, THF, 99%; d) $POCl_3$, DMF, 78%; e) K_2CO_3, $Pd(PPh_3)_4$, DMF, 78%; f) NaH_2PO_4, $NaClO_2$, 2-methyl-2-butene, t-BuOH/H_2O, 85%; g) BBr_3, CH_2Cl_2, 73 % of **1.3**, ~20% of **1.2**

Scheme 11

Commercially available orcinol (**1.56**) was first methylated and then brominated to yield **1.57** in 98% over two steps. Lithiation using *n*-butyllithium followed by trapping with triisopropyl borate resulted in the formation of boronic acid **1.58**. The boronic acid **1.58** was then immediately reacted with aldehyde **1.60** using Suzuki methodology to yield the biaryl system **1.61**, which was subsequently oxidised to the carboxylic acid **1.62**. Demethylation using the Lewis acid boron tribromide resulted in a mixture of alternariol and alternariol 9-methyl ether in 73% and 20% yield respectively.

1.5 Project Aims

1.5.1 Ulocladol A and Analogues

We intend to design a synthetic route to ulocladol A which will allow the synthesis of analogues. Any analogues prepared will be tested for biological activity and the results compared with those of the natural product. We intend to determine the SAR of ulocladol A so that this knowledge can be used to prepare a more active, drug-like compound.

### 1.5.2 Alternariol and Alternariol 9-Methyl Ether

Due to the similarities in structure between ulocladol A, alternariol and alternariol 9-methyl ether we hope to adapt the synthetic route used to prepare ulocladol A in order to prepare alternariol and alternariol 9-methyl ether. Due to the extensive work already undertaken in the preparation of these compounds, we do not intend to prepare analogues of these compounds.

2   Synthesis of Ulocladol A and Analogues

2.1   C-H Activated Coupling of Esters

The three reported syntheses of ulocladol A by Abe *et al*. all involved rearrangement of the lactone ring.[22] In each case the first step was the formation of an ester bond, which was then followed by palladium mediated coupling of the biaryl rings resulting in the formation of a six membered lactone ring. This six membered lactone ring then underwent rearrangement to form the desired seven membered ring. We proposed that it would be possible to prepare the protected benzyl benzoate ester **2.3**, from a substituted benzoyl chloride **2.1** and a substituted benzyl alcohol **2.2**, which would then undergo palladium mediated biaryl coupling. This would yield the desired seven membered lactone ring **2.4** without need for further rearrangement (Scheme 12).
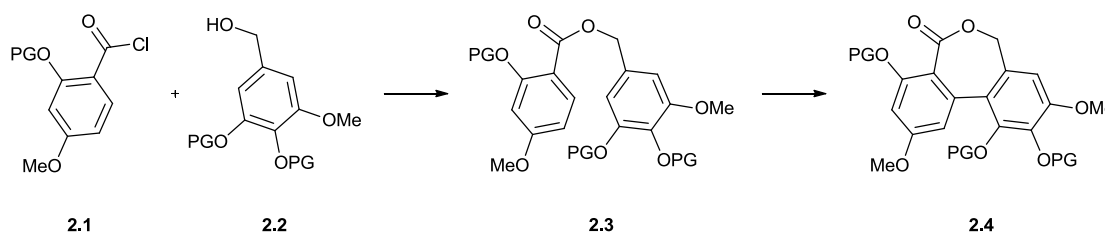
Scheme 12

Formation of other functionalised esters would also allow for the synthesis of analogues of ulocladol, with potential for substitution on both left and right hand rings.

In 2006 Gaunt *et al*. published their work on C-H activated Heck coupling (Scheme 13).[33]
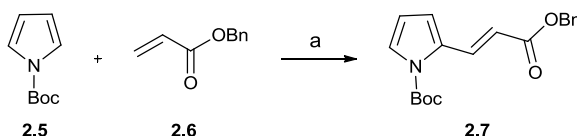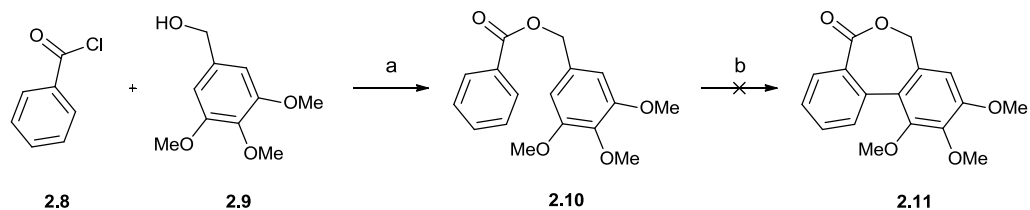

Reagents and Conditions: a) 10 mol% Pd(OAc)$_2$, *t*-BuOOBz, AcOH:Dioxane:DMSO, 35 °C, 73%

Scheme 13

They selectively coupled a pyrrole ring **2.5** and an alkene **2.6**, using Pd(OAc)$_2$ as a catalyst and *t*-BuOOBz as an oxidant, forming the carbon-carbon bond of **2.7** in good yield. We decided to apply these reaction conditions to simple benzyl benzoate esters in order to prepare a biaryl compound with a central seven membered lactone ring.

We decided to use a simple ester with limited functionalisation to test the reaction conditions. We prepared the ester 3,4,5-trimethoxybenzyl benzoate (**2.10**) from benzoyl chloride (**2.8**) and 3,4,5-trimethoxybenzyl alcohol (**2.9**), using standard esterification methodology, in excellent yield (Scheme 14).


2.8   **2.9**   **2.10**   **2.11**

Reagents and Conditions: a) Et$_3$N, MeCN, rt, 3 h, 98%; b) Pd(OAc)$_2$, t-BuOOBz, AcOH:Dioxane:DMSO, 35 °C, 96-120 h

Scheme 14

We then attempted to intramolecularly couple the two aromatic rings of **2.10** using the conditions reported by Gaunt *et al.* However, we were unsuccessful and the desired product **2.11** was not prepared. ^{1}H NMR and mass spectrometry suggested that the ester bond had broken and the only compounds isolated at the end of the reaction were starting material, benzoic acid and 3,4,5-trimethoxybenzyl alcohol. We repeated the reaction, increasing the temperature to 70 °C and extending the time to 120 h, but were again unsuccessful.

Further background reading into C-H activated, intramolecular coupling led us to a paper by Harayama *et al.*[34] Harayama reported his research into aryl-aryl coupling of two rings linked by an amide with a halide functional group on one of the rings (Scheme 15).


2.12   **2.13**
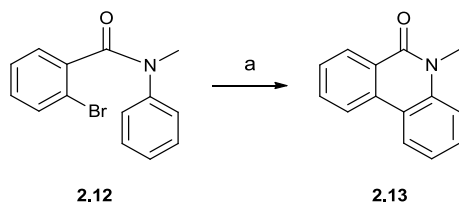
Reagents and Conditions: Pd(OAc)$_2$ (1 eq), DPPP (1 eq), n-Bu$_3$P (1 eq), Ag$_2$CO$_3$ (2 eq), DMF, Δ, 20 min, 93%

Scheme 15

2-Bromo-*N*-methyl-*N*-phenylbenzamide (**2.12**) was successfully cyclised using Pd(OAc)$_2$, DPPP and n-Bu$_3$P to prepare 5-methylphenanthridin-6(5H)-one (**2.13**).

30

We therefore decided to prepare an ester with a halide present on one of the rings. We successfully mono-brominated 3,4,5-trimethoxybenzyl alcohol (**2.9**), using *N*-bromosuccinimide to prepare 2-bromo-3,4,5-trimethoxybenzyl alcohol (**2.14**) in good yield (Scheme 16).
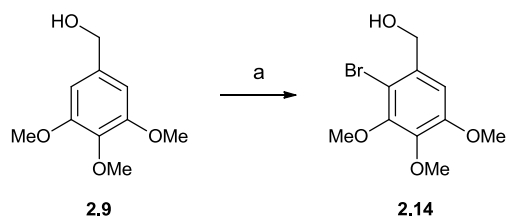
Reagents and Conditions: a) NBS, CHCl$_3$, Δ, 3 h, 85%
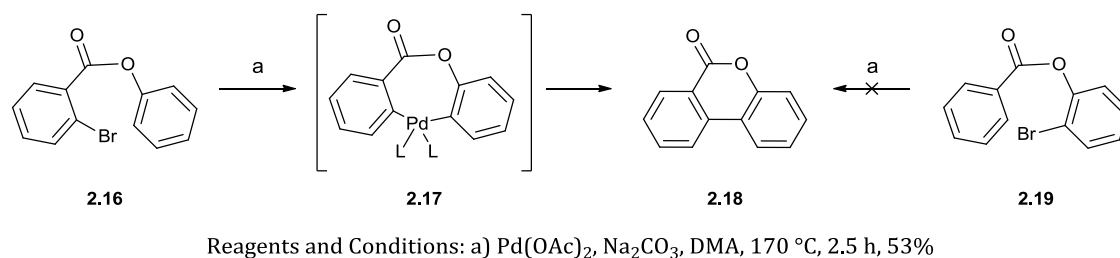
Scheme 16

The ester 2-bromo-3,4,5-trimethoxybenzyl benzoate (**2.15**) was prepared by reacting benzoyl chloride (**2.8**) and **2.14**, in the presence of triethylamine, in good yield. We then attempted to prepare the cyclised product **2.11**, by intramolecularly coupling ester **2.15** in the presence of Pd(OAc)$_2$, *n*-Bu$_3$P, DPPP, DMF and K$_2$CO$_3$. However, we were unsuccessful (Scheme 17).
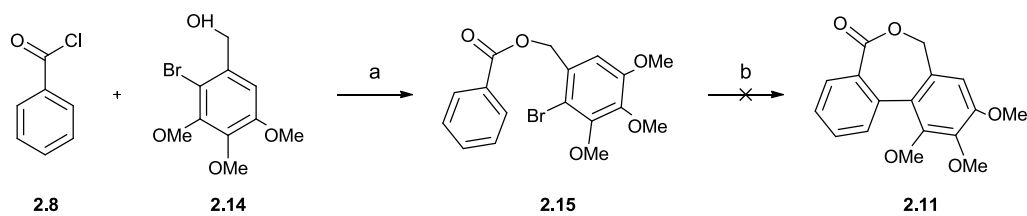
Reagents and Conditions: a) MeCN, Et$_3$N, rt, 3 h, 94%; b) Pd(OAc)$_2$, *n*-Bu$_3$P, DPPP, K$_2$CO$_3$, Δ, 24 h
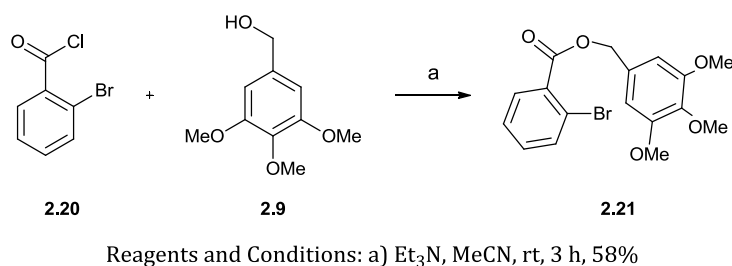
Scheme 17

An earlier paper by Ames and Opalko, published in 1984, reported their findings on palladium-mediated cyclisation of bromo substituted phenyl benzoates.[35] They determined that the location of the bromine functional group was important in the C-H activated coupling reaction (Scheme 18).

Reagents and Conditions: a) Pd(OAc)$_2$, Na$_2$CO$_3$, DMA, 170 °C, 2.5 h, 53%

Scheme 18

When bromine was situated on the left hand ring, **2.16**, the cyclisation occurred as expected, proceeding via a seven membered intermediate **2.17**. The intermediate then underwent elimination of palladium to yield benzocoumarin (**2.18**), the desired cyclised product. If bromine was situated on the right hand ring **2.19**, the desired reaction did not take place and instead, **2.19** underwent hydrolysis of the ester and no cyclised product or starting material was isolated.

As we had undertaken the reaction with the bromine on the right hand ring, we decided to repeat the reaction using an ester with bromine situated on the right hand ring. We prepared the ester 3,4,5-trimethoxybenzyl-2-bromobenzoate (**2.21**) through reaction of 2-bromobenzoyl chloride (**2.20**) and 3,4,5-trimethoxybenzyl alcohol (**2.9**) in reasonable yield (Scheme 19).


Reagents and Conditions: a) Et$_3$N, MeCN, rt, 3 h, 58%

Scheme 19

We then attempted to intramolecularly couple 3,4,5-trimethoxybenzyl-2-bromobenzoate in the presence of Pd(OAc)$_2$, n-Bu$_3$P, DPPP, DMF and K$_2$CO$_3$. However, the reaction was again unsuccessful and we were only able to isolate starting material and decomposed starting material from the crude reaction mixture. We repeated the reaction with different catalysts, ligands and bases but were still unsuccessful.

Further research implied that in some cases bromine was not reactive enough for a C-H activated reaction to take place, and a more reactive halide was required. A paper by Harayama *et al.* reported his use of C-H activation to intramolecularly couple iodinated esters in the synthesis of the natural product arnottin I.[36]

Harayama *et al.* used phenyl *o*-iodobenzoate as a test substrate for optimisation of the reaction conditions (Scheme 20).

2.22   **2.18**
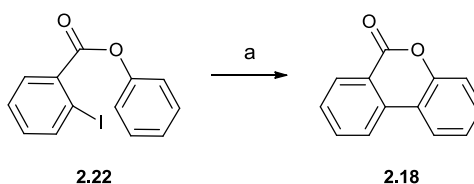
Reagents and Conditions: a) catalyst (0.1 eq), ligand (0.2 eq), base (2 eq), DMF, Δ

Scheme 20

They successfully prepared benzocoumarin (**2.18**) from phenyl *o*-iodobenzoate (**2.22**) using a range of conditions (Table 2).

Run	Catalyst	Ligand	Base	Time (h)	Temp	Yield (%) of 2.18
1	Pd(OAc)$_2$	None	NaOAc	1	Reflux	84
2	Pd(OAc)$_2$	DPPP	NaOAc	2	Reflux	75
3	Pd(OAc)$_2$	PPh$_3$	NaOAc	2	130 °C	73
4	Pd(acac)$_2$	PPh$_3$	NaOAc	2.5	130 °C	57
5	Pd(OAc)$_2$	PPh$_3$	Ag$_2$CO$_3$	24	Reflux	38

Table 2-Reported Conditions and Yields in the Synthesis of Benzocoumarin

They determined that the best results were obtained using Pd(OAc)$_2$ as a catalyst, sodium acetate as a base and DMF as the solvent. They reported varying results regarding the use of a ligand, with the best result being obtained when no ligand was present in the reaction, as shown in entry 1. Changing the base from sodium acetate to silver carbonate had a detrimental effect on the yield, reducing it by 35%, as shown in entry 5.

Harayama *et al.* also reported their efforts to couple the regioisomer of **2.22**, where the iodo group was on the right hand ring **2.23** (Scheme 21).

2.23   **2.18**
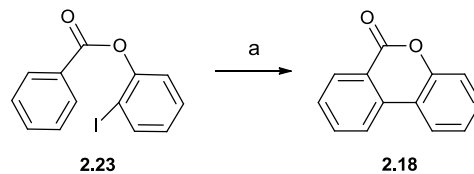
Reagents and Conditions: a) Pd(OAc)$_2$ (0.1 eq), PPh$_3$ (0.2 eq), NaOAc (2 eq), DMF, Δ, 2 h, 6%

Scheme 21

They successfully prepared benzocoumarin (**2.18**) from *o*-iodophenyl benzoate (**2.23**) using Pd(OAc)$_2$, PPh$_3$ and NaOAc. However, at 6%, the yield was very low, suggesting that despite the more reactive halogen the location is still very important.

We decided to try the reagents reported by Harayama *et al.* for the C-H activated coupling of an iodo-substituted ester. Prior to this we had to iodinate the required ester and we proposed to do this through halogen exchange of **2.21**. We tried to exchange the bromine atom for an iodine atom using catalytic copper iodide, sodium iodide and *N,N'*-dimethylethylene diamine (Scheme 22).

2.21   **2.24**
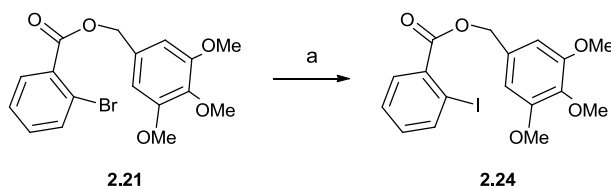
Reagents and Conditions: a) CuI, NaI, *N,N'*-dimethylethylene diamine, dioxane, 110 °C, 23 h
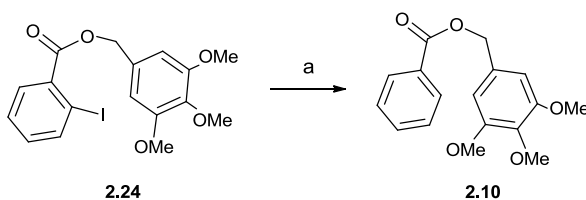
Scheme 22

The reaction mixture was heated at 110 °C for 23 h, after which TLC showed starting material remained. We added additional amounts of CuI, NaI and *N,N'*-dimethylethylene diamine and then heated for an additional 48 h at 110 °C. After a total reaction time of 71 h we stopped the reaction. Crude ^{1}H NMR suggested that we had a mixture of starting material **2.21** and product **2.24**. We tried to separate the mixture using column chromatography but were unsuccessful as the product and starting material ran at the same Rf. The presence of iodinated product **2.24** was confirmed by mass spectrometry with the presence of a peak at 451 m/z corresponding to a [M+Na]$^+$ molecular ion. ^{1}H NMR suggested that there was a 2:1 ratio of starting material:desired product. Despite this, we decided to proceed with the coupling reaction using the mixture of brominated and iodinated esters (Scheme 23).
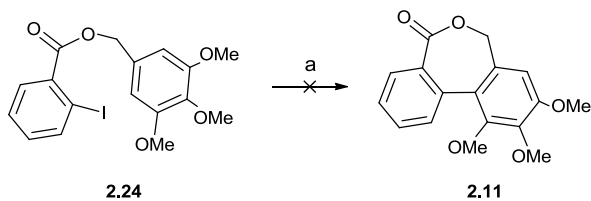
Reagents and Conditions: a) Pd(OAc)$_2$, PPh$_3$, NaOAc, 130 °C, 24 h

Scheme 23

We initially tried the reaction in the absence of the ligand. After 4 h there was no change in TLC from starting material, so 2 equivalents of the ligand was added and the reaction stirred at 130 °C for a further 20 h. After a total of 24 h, TLC indicated a change with the appearance of a second spot on the TLC plate. ^{1}H NMR and mass spectrometry of the crude product suggested that the reaction had not been successful. Instead of the expected product, the palladium had inserted into the C-I bond and then eliminated to form a C-H bond rather than cyclising to form the desired seven-membered ring (Scheme 24).

Reagents and Conditions: a) Pd(OAc)$_2$, PPh$_3$, NaOAc, 130 °C, 24 h

Scheme 24

This resulted in the formation of **2.10**, which was confirmed by mass spectrometry and comparison of the ^{1}H NMR spectrum with the spectrum of the previously prepared unhalogenated ester. A mixture of the brominated and iodinated starting material was also isolated.

Despite functionalising the ester with a more reactive halogen as suggested by Harayama *et al.* the aryl-aryl bond forming reaction was still unsuccessful. When comparing the prepared ester **2.24** with the esters reported by Harayama there are several differences. The methoxy groups present on the right hand ring of ester **2.24** may have more than one detrimental effect on the reaction; not only are they sterically bulky groups, but they are electron donating groups which could have an electronic effect on the reaction. In addition to the effects of the methoxy groups, we are trying to form a seven membered lactone ring.

In order to form the seven membered ring the intermediate palladacycle is an eight membered ring. This ring size is much more difficult to form than the seven membered palladacycle intermediate prepared by Harayama. It is possible that these differences caused the C-H activation reaction to be unsuccessful. Therefore, we decided to move away from this route to ulocladol A.

2.2 Ullmann Coupling

A paper published by Tsuboi *et al.* in 2007 reported their work on the total synthesis of 3,3',4'-tri-*o*-methylellagic acid.[37] They successfully used intramolecular Ullmann coupling to prepare a sterically hindered, biaryl system, linked by a 6-membered lactone ring **2.26** (Scheme 25).

Reagents and Conditions: a) Cu, DMF, 110 °C 3h, then 180 °C 16 h, 59%
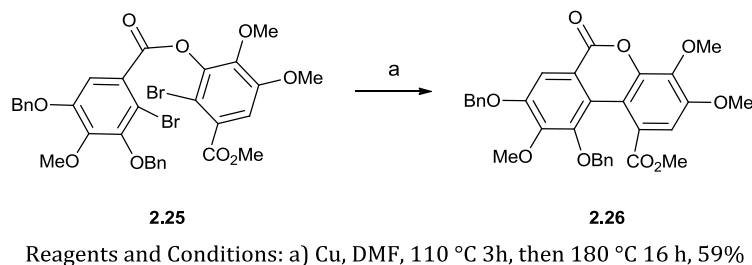
Scheme 25

The system was cyclised using one equivalent of copper in 59% yield. This ester had several bulky electron donating and withdrawing groups, which suggested that, in this system, the electronic and steric nature of nearby groups did not affect the reaction. We decided to try this method on our test model which would still cyclise to yield a seven-membered lactone ring. We successfully reacted 2-bromobenzoyl chloride (**2.20**) and 2-bromo-3,4,5-trimethoxybenzyl alcohol (**2.14**), in the presence of triethylamine, to prepare the dihalogenated 2-bromo-3,4,5-trimethoxybenzyl 2-bromobenzoate (**2.27**) in quantitative yield (Scheme 26).

2.20   **2.14**   **2.27**
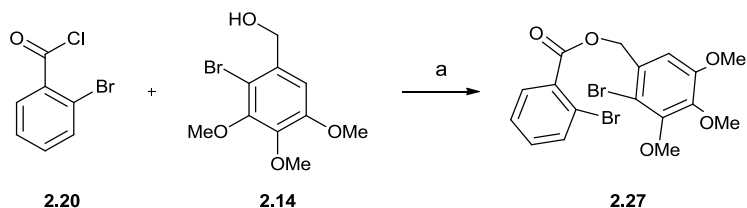
Reagents and Conditions: a) Et₃N, MeCN, rt, 3 h, quant.

Scheme 26

Having successfully prepared the ester, the next step was to couple the two aryl rings under the conditions reported by Tsuboi *et al.* (Scheme 27).


2.27   **2.28**
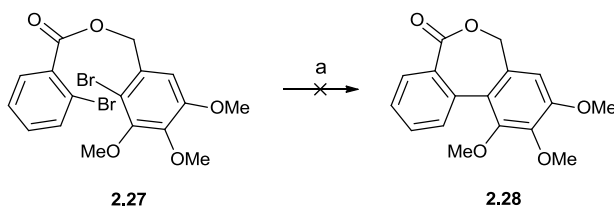
Reagents and Conditions: a) Cu, DMF, 110 °C, 3h, then 180 °C, 18 h

Scheme 27

Unfortunately we were not successful, with both ^{1}H NMR and mass spectrometry suggesting that copper insertion into one of the C-Br bonds had occurred but cyclisation had not taken place. Instead, when the copper eliminated, a C-H bond formed, which was confirmed by ^{1}H NMR with the addition of an extra proton signal in the aromatic region and the presence of an isotope pattern in the mass spectrum indicative of only one bromine atom in the molecule. We repeated the reaction, extending the reaction time to 48 h and increasing the quantity of copper to two equivalents, but were still unsuccessful.

Another paper by Kanth *et al.* reported their research on the homo-coupling of aryl magnesium bromides catalysed by zinc bromide.[38] They successfully coupled a range of aryl magnesium bromides at room temperature in good yield. Although Kanth *et al.* only reported work on homo-coupling we were hopeful that the conditions would be applicable to our system, as the two rings would be held in close proximity due to the linking ester bond. Therefore we tried to couple the two rings using the reported conditions (Scheme 28).

2.27   **2.28**
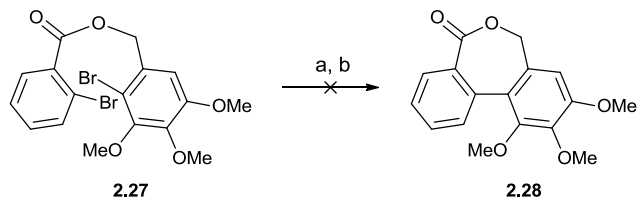
Reagents and Conditions: a) Mg, Et$_2$O, 0 °C → rt, 1 h; b) ZnBr$_2$ (5 mol%), rt, 6 h

Scheme 28

The first step was the formation of a Grignard reagent which was then added to a mixture of zinc bromide in ether and then stirred for 6 h at rt. After 6 h no change was observed in the TLC. We extended the reaction time by 24 h and warmed to 35 °C but the reaction failed and no insertion into either of the C-Br bonds was observed.

A third procedure by Yuan *et al.* reported their research on the homo-coupling of various aryl halide compounds catalysed by manganese (II) chloride.[39] We decided to use the conditions reported by Yuan *et al.* to try and couple the ester **2.27** (Scheme 29).

2.27   **2.28**
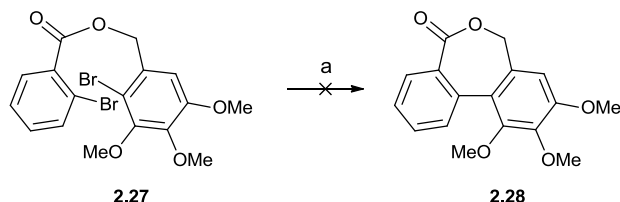
Reagents and Conditions: a) Mg, MnCl$_2$ (10 mol%), THF, rt, 24 h

Scheme 29

After 24 h no change was observed in TLC. Crude ^{1}H NMR and mass spectrometry suggested that there was no insertion into either C-Br bond. Literature searches revealed that it was possible to form a biaryl carbon-carbon bond with a linking seven-membered ring and this was demonstrated in a paper by Fagnou *et al* (Scheme 30).[40]

2.29a X = Cl, R = CO₂Me   **2.30** R = CO₂Me (73%)
2.29b X = Br, R = H   **2.31** R = H (69%)
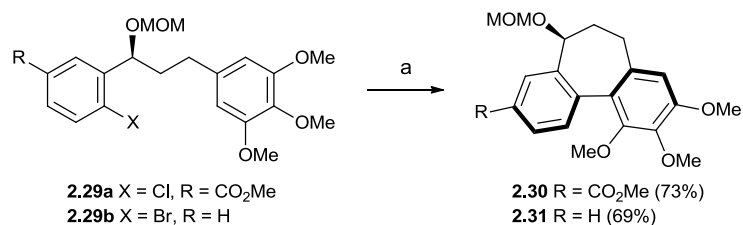
Reagents and Conditions: a) Pd(OAc)$_2$ (10 mol%), DavePhos (10 mol%), K$_2$CO$_3$ (2 eq), DMA, 145 °C

Scheme 30

Fagnou *et al.* reported the successful formation of a carbon-carbon bond, with a linking seven-membered ring, through C-H activation. The paper reported the use of both chloride and bromide with good yields observed in each. From this paper we were able to determine that the methoxy groups did not impose steric and electronic effects on the reaction to a degree that would prevent the coupling from occurring. This implied that the problems we observed in our reactions could be caused by a different part of the molecule. The major difference between the compounds we prepared and those reported by Fagnou *et al.* is the presence of an ester linking bond. We proposed that the lack of success observed in both the C-H activated coupling and Ullmann coupling could be due to the inability of the two reactive sites to come into close enough proximity for the reaction to occur. The ester bond is relatively rigid and less likely to rotate round into a conformation which would allow the two desired sites to react. In addition to this, the CH$_2$ would allow for rotation of the right hand ring away from the left hand ring in order to reduce steric hindrance, reducing the possibility of interaction of the two rings. This would explain why, in some cases, we observed palladium insertion into a C-X bond, but no carbon-carbon bond formation.

This implies that, in the case of our system, the ester bond is an obstacle in the coupling reaction. Therefore, we proposed that if we were to replace the ester with an ether we would be able to successfully couple the two aryl rings and then reintroduce the carbonyl at a later stage. The next step was to prepare an ether system which we could use to test the arylation coupling conditions. Fagnou *et al.* reported success coupling ethers which were mono-brominated on the left hand ring. Unfortunately, despite attempting to prepare ethers which were mono-brominated on either the left hand or right hand ring, we were only able to prepare the dibrominated ether **2.33** (Scheme 31).
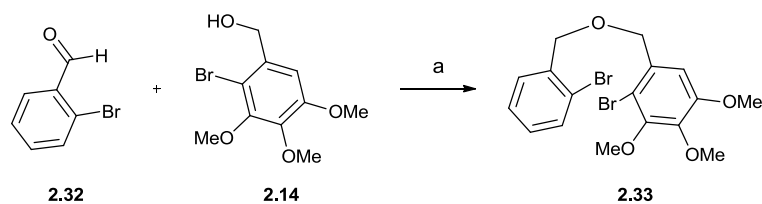
Reagents and Conditions: a) Et$_3$SiH, TFA, DCM, 0 °C → rt, 16 h, 24%

Scheme 31

Ether **2.33** was prepared by reacting 2-bromobenzaldehyde with 2-bromo-3,4,5-trimethoxybenzyl alcohol in 24% yield. We then tried to form a carbon-carbon bond between the aryl rings of ether **2.33** following the conditions reported by Fagnou *et al.*, although with SPhos in place of DavePhos (Scheme 32).
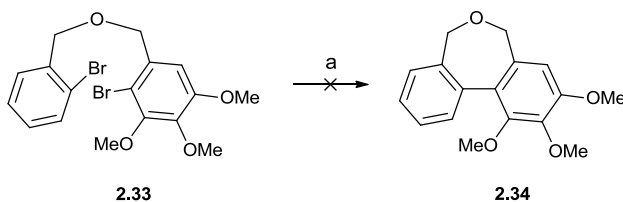
Reagents and Conditions: a) Pd(OAc)$_2$ (10 mol%), SPhos (10 mol%), K$_2$CO$_3$, DMA, 145 °C

Scheme 32

However, ^{1}H NMR and mass spectrometry suggested that the reaction had been unsuccessful with no product observed.

Due to the lack of success in C-H activated coupling and Ullmann coupling, we decided to change the proposed route to ulocladol and concentrate on cross coupling. Instead of forming the carbon-carbon bond through intramolecular coupling once the ester bond was in place, we proposed to firstly form the carbon-carbon bond and then cyclise to prepare the seven-membered lactone.

2.3 Suzuki Coupling

There are several different methods of forming a carbon-carbon bond between two aryl rings. Some of the better known methods include Stille, Negishi and Suzuki coupling.[41-46] All three of these reactions require an organometallic reagent and a catalyst. The reactions are usually mediated by palladium and involve the coupling of an aryl halide or triflate

with an organostannane in the Stille reaction, an organozinc in the Negishi or an organoboron in the Suzuki reaction. We proposed that it would be possible to prepare ulocladol A through the Suzuki reaction using an aryl triflate and an aryl boronate (Figure 17).


1) Oxidative addition; 2) Halogen-base exchange; 3)Transmetallation;
4) Reductive elimination
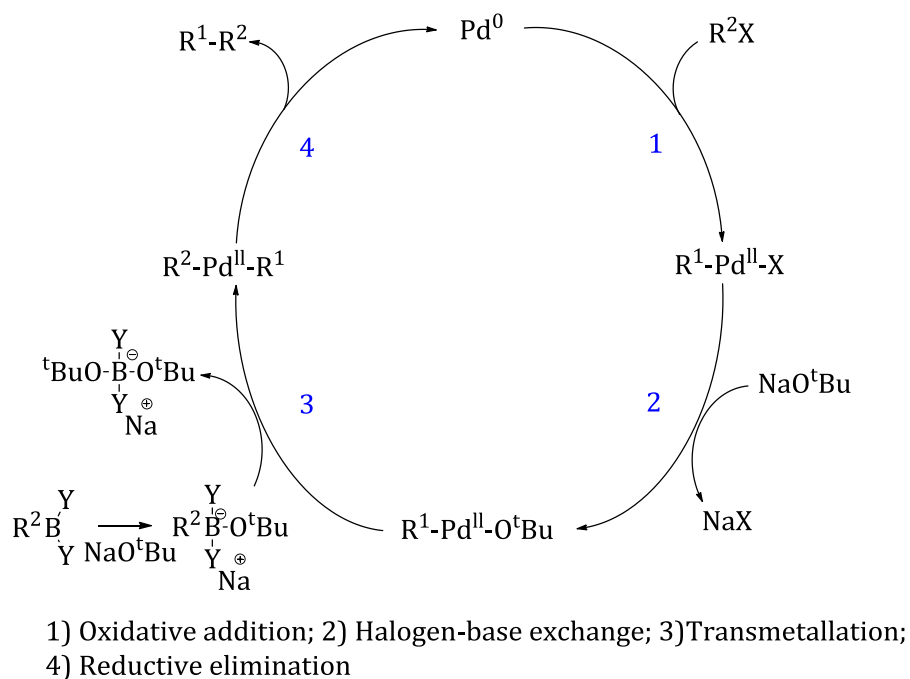
Figure 17 – Suzuki coupling reaction

The first step involves oxidative addition of the aryl triflate to a zerovalent palladium source yielding a palladium (II) complex. Transmetallation between the palladium complex and the organoboron compound followed by reductive elimination yields the desired cross coupled product with regeneration of the palladium zerovalent catalyst.

Retrosynthesis of ulocladol A using Suzuki coupling as the key step in this synthesis provided us with two fragments (Scheme 33).
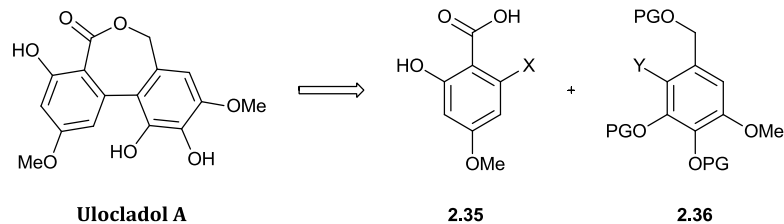
Scheme 33

Splitting ulocladol A across the carbon-carbon bond and the ester group gave fragments **2.35** and **2.36**, both of which would need to carry a group in the X and Y positions that would be suitable for cross coupling. Fragment **2.35** is similar to a known compound **2.37** which was used in Suzuki couplings by Takahashi *et al.* in the total synthesis of dehydroaltenusin (Scheme 34).[47]
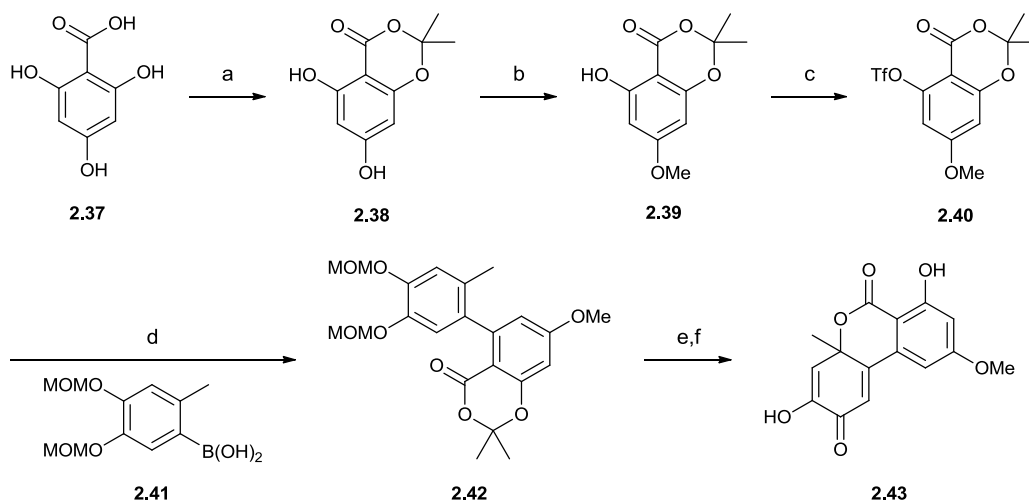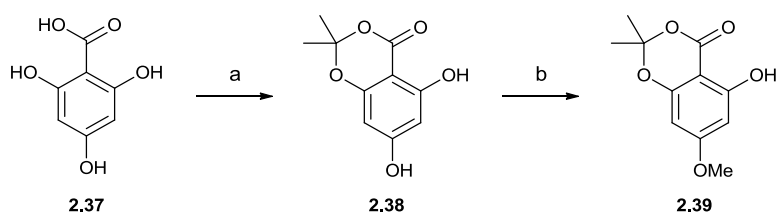

Reagents and Conditions: a) acetone, SOCl$_2$, DMAP, DME, rt, 56%; b) DIAD, Ph$_3$P, MeOH, THF, rt, 89%; c) Tf$_2$O, pyridine, 0 °C, 94%; d) Pd(PPh$_3$)$_4$, K$_3$PO$_4$, KBr, dioxane, 100 °C, 93%; e) BCl$_3$, CH$_2$Cl$_2$, 0 °C → rt; f) FeCl$_3$, aq. EtOH, rt, 52% over 2 steps

Scheme 34

We proposed ulocladol A could be prepared by coupling the triflate **2.40** with the corresponding boronic acid.

The first step was to protect the carboxylic acid as an acetonide. Takahashi *et al.* reported their use of thionyl chloride, acetone and catalytic DMAP to prepare acetonide **2.38**. However, this method proved unsuccessful and we were unable to obtain the desired product. Further investigation yielded an alternative method reported by Danishefsky *et al.*, to protect the carboxylic acid as an acetonide.[48] The commercially available 2,4,6-

trihydroxybenzoic acid monohydrate (**2.37**) was successfully protected using TFA, TFAA and acetone in 61% yield (Scheme 35).


Reagents and Conditions: a) TFA, TFAA, acetone, rt, 24 h, 61%; b) MeOH, Ph$_3$P, DIAD, THF, 0 °C → rt, 2 h, 87%

Scheme 35

Compared to the literature yields this was an increase of 5% and 27% for the thionyl chloride and TFA methods respectively.

The phenol *para* to the protected carboxylic acid was then selectively alkylated using a standard Mitsunobu protocol to prepare **2.38** as a white solid in 87% yield. We proposed that the selectivity observed in this reaction could be due to several factors. Following the first protecting step, the two phenols have different signals in the ^{1}H NMR spectrum. The phenol *ortho* to the carbonyl corresponds to a sharp singlet at 10.45 ppm, whereas the phenol *para* to the carbonyl is a broad singlet at 9.66 ppm. This suggests the presence of a hydrogen bond between the hydrogen of the *ortho* phenol and the carbonyl oxygen atom. This hydrogen bond lowers the acidity of the proton therefore reducing the likelihood of this phenol reacting in the Mitsunobu reaction. In addition to this, not only is the proton on the phenol *para* to the carbonyl likely to be more acidic, and therefore more reactive, than the phenol *ortho* to the carbonyl, it is also not sterically hindered by neighbouring groups.

The next step in the synthesis was to convert the remaining phenol into a triflate group which could then be used in a Suzuki coupling reaction. The procedure reported by both Takahashi *et al.*, and Danishefsky *et al.* described the use of trifluoromethanesulfonic anhydride in the presence of pyridine to prepare the triflate **2.40** (Scheme 36).
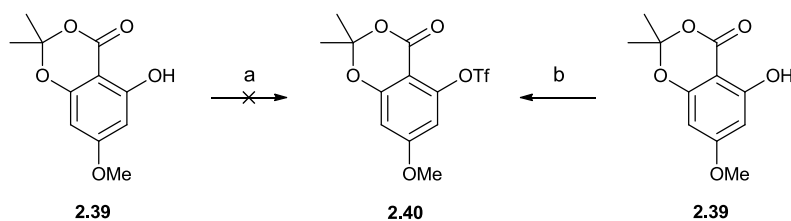
Reagents and Conditions: a) Tf$_2$O (1.1 eq), pyridine, 0 °C, 12 h; b) Tf$_2$O, 3 M LiOH (aq). toluene, 0 °C, 3 h, 81%

Scheme 36

However, the reaction was unsuccessful under these conditions and only starting material was isolated. Following an alternative procedure reported by Frantz *et al.*, we successfully prepared **2.40** in 81%, by adding trifluoromethanesulfonic anhydride to a biphasic solution of aqueous LiOH and toluene.[49] The product was confirmed by ^{1}H NMR with the loss of the phenolic OH signal at 10.43 ppm and mass spectrometry with a molecular ion peak at 357 m/z.

Having successfully prepared the left hand side of ulocladol A we decided to test the stability of the triflate to Suzuki coupling conditions with a commercially available boronic acid **2.44** (Figure 18).
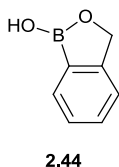
Figure 18 – 2-(Hydroxymethyl)benzeneboronic acid hemiester

We chose **2.44** as the test boronic acid for two reasons. Firstly, there were no additional substituents on the benzene ring which could adversely affect the Suzuki reaction. Secondly, the boronic acid was trapped in a five-membered ring with the benzyl alcohol required for the formation of the lactone ring. This meant that it was not necessary to protect the alcohol during the Suzuki reaction and then add an additional deprotection step once the reaction was complete.

A 1990 paper by Suzuki *et al.* reported their research on palladium-mediated cross coupling.[50] Suzuki *et al.* successfully coupled a range of aryl and vinyl triflates with

organoboron compounds using 2.5 mol% of PdCl$_2$(dppf) and excess K$_3$PO$_4$ in THF. We applied these conditions to the triflate **2.40** and the boronic acid **2.44** (Scheme 37).

Reagents and Conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h, 34%
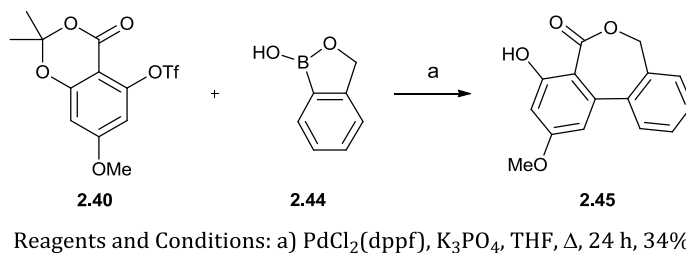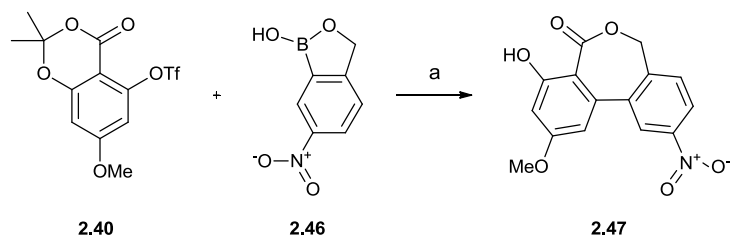
Scheme 37

The reaction was successful, and we obtained the cyclised product **2.45** in 34% yield following purification. We were surprised to discover that the lactone ring had cyclised in situ and proposed that this was due to the basic nature of the benzyl alcohol anion which can intramolecularly attack the carbonyl causing loss of the acetonide protecting group. This resulted in the preparation of an analogue of ulocladol A in four steps.

The structure of the product was confirmed by ^{1}H NMR and mass spectrometry. Several key signals were observed in the ^{1}H NMR spectrum. There was a sharp peak at 10.64 ppm, indicating the presence of a hydrogen bonded phenol; two doublets at 6.61 and 6.63 ppm indicating the two aromatic protons on the left hand ring; two broad singlets at 5.00 and 5.17 ppm assigned to the two protons on the CH$_2$ of the lactone ring; and a singlet at 3.89 indicating the presence of the methoxy group on the left hand ring. A multiplet at 7.35 – 7.69 ppm, which integrated at four protons, indicated the presence of the four aromatic protons on the right hand ring. The presence of a peak at 255 m/z confirmed the presence of a product with a [M-H]⁻ molecular ion. The analogue **2.45** was tested for biological activity against a variety of tyrosine kinase inhibitors and was found to have limited activity. This implied that the substituents on the right hand ring were important in the activity of ulocladol A against tyrosine kinase.

It was proposed that the Structure-Activity Relationships (SAR) of ulocladol A could be investigated, by coupling the prepared triflate **2.40** with substituted organoboron compounds. The activity of these analogues could then be compared to the activity of **2.45**, allowing us to establish the effect of right hand ring substituents on biological activity.

It was possible to purchase one cyclised benzene boronic acid with a substituent. This was 2-hydroxymethyl-5-nitrophenylboronic acid, **(2.46)** (Scheme 38).

Reagents and Conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h, 32%

Scheme 38

2.46 was cross coupled with triflate **2.40** to prepare **2.47** in 32% yield. The product was confirmed by mass spectrometry with a peak at m/z 300 for the [M-H]⁻ mass ion. In addition, all the major signals in the positive electrospray mass spectrum could be attributed to ions relating to the product mass ion. However, the ^{1}H NMR suggested the presence of 2 compounds, with double the expected number of signals in the spectrum apart from the hydrogen bonded phenol. The signal for the phenol at 10.61 ppm is the only peak that is not duplicated and does not integrate to two protons (Figure 19).
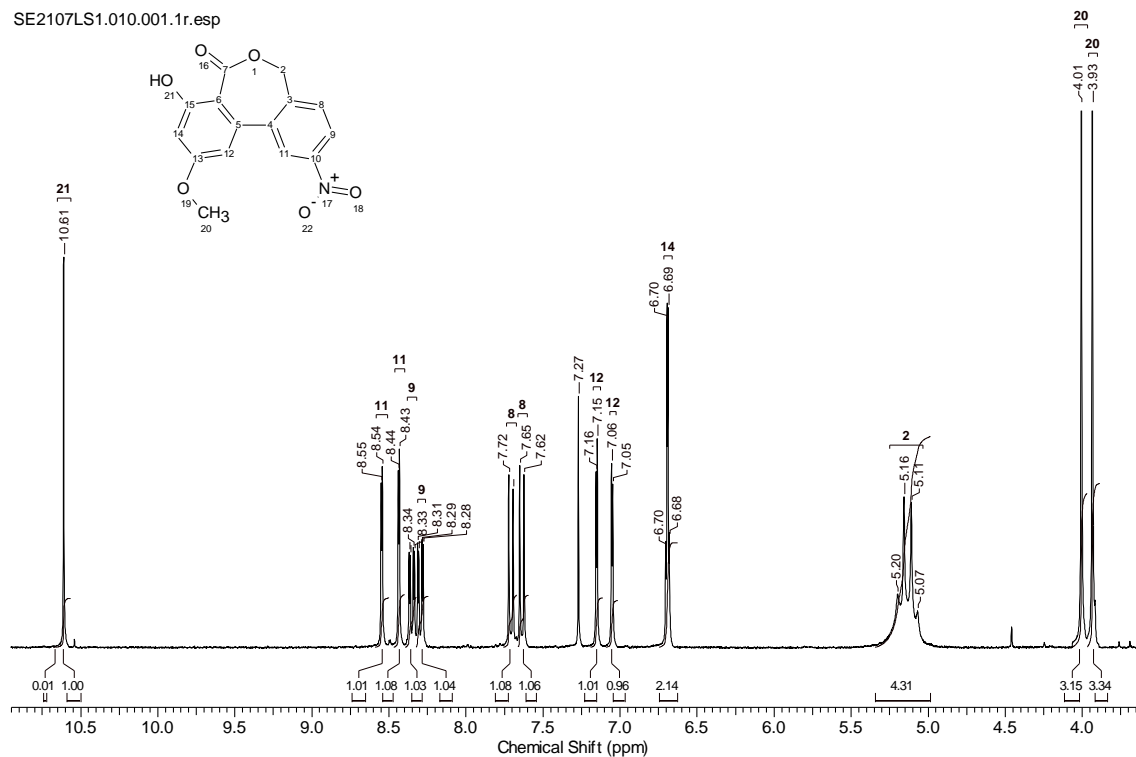
Figure 19 – ¹H NMR spectrum of 2.47

TLC and mass spectrometry are both consistent with only one compound being present, whereas NMR suggests two, very similar compounds. Closer inspection of the aromatic region shows that each peak appears to be doubled, although the distance between each set of corresponding peaks is variable (Figure 20).
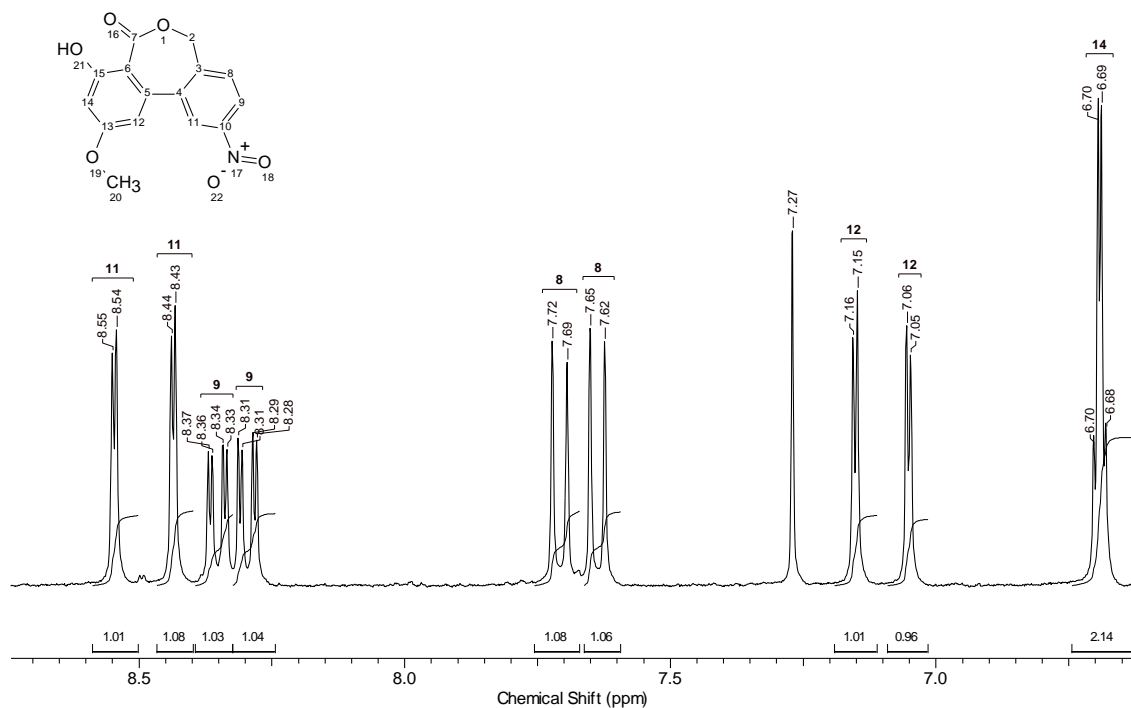
Figure 20 – Aromatic region of ¹H NMR spectrum of 2.47

The protons closest to the aryl-aryl bond appear to experience the greatest distance between corresponding peaks, whilst those furthest away are only slightly shifted. This can be observed with the signals on the right hand ring. Proton 11, the proton *ortho* to the aryl-aryl bond, appears to correspond to the two signals at 8.44 ppm and 8.55 ppm. The difference between the two shifts is 0.1 ppm. The signals corresponding to proton 9, the proton *para* to the aryl-aryl bond on the right hand ring, at 8.30 ppm and 8.35 ppm are only separated by 0.05 ppm, half the distance of the protons closest to the carbon-carbon bond. The peaks at 7.64 ppm and 7.71 ppm, corresponding to proton 8, which is the only proton *meta* to the aryl-aryl bond, are separated by 0.07 ppm. This appears to suggest that the further the distance from the aryl-aryl bond the proton is the smaller the difference in the shifts between corresponding peaks.

This is not observed in the same way for the protons on the right hand ring. Closer inspection of the signals at 7.05 ppm, 7.15 ppm, 6.70 ppm and 6.69 ppm suggests that the two protons at 7.05 ppm and 7.15 ppm couple to each other, as do the two protons at 6.70 ppm and 6.69 ppm. In the case of these four protons, not only are the distances between the coupled signals different, but the shifts of the peaks are also quite different. We are

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unsure why this occurs and also why ^{1}H NMR suggests that there are two compounds present.

The presence of two compounds could also be observed with the ^{13}C NMR spectrum Instead of the expected 15 signals, 24 were observed. 13 of the peaks could be attributed to the aromatic C-H, CH_2 and OCH_3 carbons with 2 signals for each of the CH_2 and OCH_3 groups and 9 for the 5 aromatic C-H carbons. The remaining 11 signals correspond to the remaining 8 quaternary carbons.

Due to the similarities in shifts of the corresponding peaks and the presence of only one mass ion peak we believe that the two compounds must be very closely related. One possibility is the presence of regioisomers. Whilst we can be certain from NMR experiments that the triflate starting material was one compound, we are unsure about the commercially bought 2-hydroxymethyl-5-nitrophenylboronic acid (**2.46**). Unfortunately, all of the starting material was consumed in the reaction so we were unable to determine whether more than one compound was present. If we were able to prepare regioisomers of analogue **2.47** we would be able to compare the ^{1}H NMR spectra. This would allow us to determine whether the explanation for the doubling of the peaks was due to the presence of more than one regioisomer.

As no additional cyclised benzene boronic acids were commercially available, it was decided to prepare them synthetically. Zhdankin and co-workers published a route to the unsubstituted benzene boronic acid **2.44** starting from 2-bromobenzyl alcohol (Scheme 39). [51]
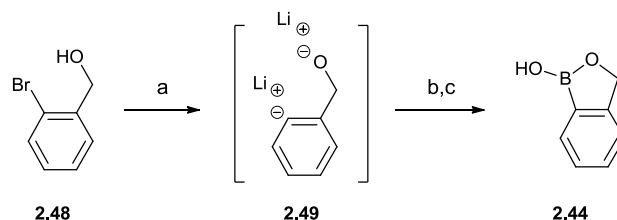
Reagents and Conditions: a) *n*-BuLi, Et$_2$O, 0 °C, 2 h; b) B(O*i*-Pr)$_3$, Et$_2$O, -78 °C, 2 h then rt 16 h; c) HCl, 86% overall

Scheme 39

Zhdankin suggested that the reaction went through a dianion intermediate **2.49** which was trapped with triisopropyl borate and then hydrolysed to form the desired boronic acid. We

proposed to use this method to prepare substituted benzene boronic acids which could then be coupled with the triflate.

As the unsubstituted analogue of ulocladol A had already been successfully prepared it was decided to synthesise a benzene boronic acid with one substituent which would not affect the lithiation and trapping reaction. With this in mind we decided to prepare 2-bromo-4-methylbenzyl alcohol (Scheme 40).
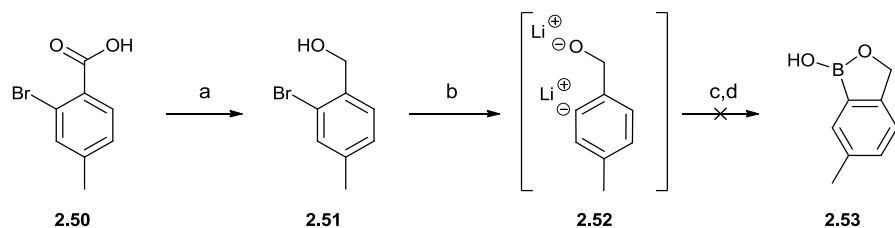
Reagents and Conditions: a) NaBH$_4$, ZrCl$_4$, THF, rt, 16 h, 90%; b) n-BuLi, Et$_2$O, 0 °C, 2 h; c) B(Oi-Pr)$_3$, Et$_2$O, -78 °C, 2 h then rt 16 h; d) HCl

Scheme 40

We successfully reduced carboxylic acid **2.50** to benzyl alcohol **2.51**, using sodium borohydride and zirconium chloride, in 90% yield.[52] The next step was to prepare the cyclised boronic acid. Following the procedure reported by Zhdankin and co-workers, we attempted to prepare the dianion species **2.52** and then trap with triisopropyl borate to yield the desired product **2.53**. However, the reaction failed. We changed the reaction conditions experimenting with time, temperature and equivalents of n-BuLi and B(Oi-Pr)$_3$. In addition the borate was also changed and fresh triisopropyl borate, trimethyl borate and triethyl borate were all tried but to no avail and we were only able to isolate 4-methylbenzyl alcohol from the crude reaction. This led us to the conclusion that it was the trapping step which was causing the problem. Treatment of 2-bromo-4-methylbenzyl alcohol with butyllithium leads to the formation of a dianion species **2.52** which should then trap a borate molecule. If a borate has not been successfully trapped, addition of hydrochloric acid will cause protonation of the dianion species forming 4-methylbenzyl alcohol as a side product of the reaction.

As we were unable to prepare substituted benzene boronic acids in this way we decided to consider another route to ulocladol A and its analogues. A common solution to problems with Suzuki cross coupling is to exchange the location of the functional groups involved in

the reaction. In this case, if we were to exchange the triflate group on the left hand ring for a boronic acid, we would have a lot more options for groups on the right hand ring. As well as coupling the boronic acid with a triflate group, it would also be possible to use halides. As these are much more readily available it would allow a more in depth investigation into the SAR of the right hand ring.

Following a procedure reported by Altemöller *et al.* we were able to substitute the triflate group with a boronic ester (Scheme 41).[53]

2.40   **2.54**   **2.55**   **2.56**

Reagents and Conditions: a) Pd(PPh$_3$)$_4$, Et$_3$N, THF, Δ, 4 h, 83%
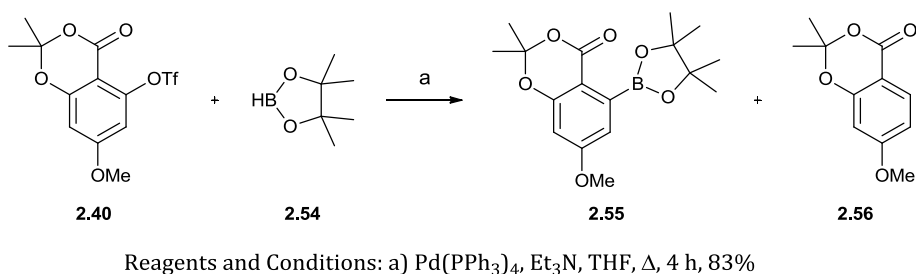
Scheme 41

However, it was necessary to adapt the conditions reported by Altemöller *et al.* as we observed varying yields (Table 3).

Entry	Triflate (mmol)	Pinacol Borane (eq)	Pd(PPh$_3$)$_4$ (eq)	Et$_3$N (eq)	Solvent	Time (h)	Yield (%)
Reported Conditions	0.50	16.84[a]	0.05	3	Dioxane	4	54
1	0.25	16.84[a]	0.05	3	Dioxane	4.75	0
2	0.84	16.84[a]	0.06	3	THF	12	26
3	1.40	15[b]	0.06	3	THF	4	49
4	1.25	10[b]	0.06	3	THF	4	59
5	1.00	5[b]	0.06	3	THF	4	83
6	4.00	5[b]	0.06	3	THF	4	92
7	0.84	2.5[b]	0.06	3	THF	6	36

a: 1-2% Triethylamine stabilised solution   b: 1M solution in THF

Table 3-Optimisation of Conditions for the Formation of the Boronic Ester

Altemöller *et al.* reported a yield of 54% of the desired product **2.55** and 22% yield of the side product **2.56**. When we repeated the reaction using the conditions reported by Altemöller *et al.* we were unable to isolate any product, and the only identifiable products

of the reaction were starting material, the side product **2.56** and an alkyl residue. We found that if dioxane was replaced with THF, catalyst loading increased to 6 mol% and the reaction time extended to 12 h the yield increased to 26%. However this was not an acceptable yield for an intermediate in the synthesis. In addition to the poor yield we also observed an impurity in the product which could not be removed by column chromatography, recrystallisation or acid and base washes. We were unable to determine the identity of the impurity with absolute certainty but proposed that it was related to the structure of pinacol borane. We hoped that if we were able to reduce the quantity of pinacol borane required in the reaction, without adversely affecting the yield, we would be able to reduce the quantity of impurity in the product.

We had observed that the yield was adversely affected if triethylamine and tetrahydrofuran were not freshly distilled for the reaction. As Altemöller *et al.* had used a 1-2% triethylamine stabilised solution as their source of pinacol borane, we considered the possibility that this was adversely affecting the reaction. If the triethylamine used to stabilise the pinacol borane was wet or impure it could cause problems in the reaction. Therefore it was decided to try using a 1 M solution of pinacol borane in THF. We found that 15 equivalents of a 1 M solution increased the yield of the reaction by 23%. The reaction time was also affected, reducing it back down to 4 h. However, a large quantity of impurity was still observed in the product. In an attempt to reduce the quantity of the impurity, we reduced the equivalents of pinacol borane to 10 (entry 4). We were surprised to discover that this did not have a detrimental effect on the yield, and actually caused an increase to 59% whilst retaining the shorter reaction time. We repeated the reaction reducing the equivalents of pinacol borane to 5 and were able to increase the yield to 83%. We found that this also caused a decrease in the quantity of impurity observed in the product, as well as being a reproducible reaction on a larger scale. Further reduction of pinacol borane to 2 equivalents caused a decrease in yield to 36%.

Having successfully prepared the boronic ester intermediate the next step was to couple the boronic ester with a bromobenzyl alcohol. Due to the previously attempted C-H activation route we had a stock of 2-bromo-4-methylbenzyl alcohol (**2.51**) and 2-bromo-3,4,5-trimethoxybenzyl alcohol (**2.14**) readily available. We proposed to test stability of the boronic ester to the Suzuki reaction conditions by coupling with the methoxy substituted bromobenzyl alcohol. We decided not to protect the benzyl alcohol because, if

the reaction was successful, this would negate the need to add an additional two steps to the synthesis. Using the Suzuki conditions applied to the synthesis of analogues **2.45** and **2.47** we successfully coupled the boronic ester with 2-bromo-4-methylbenzyl alcohol (Scheme 42).
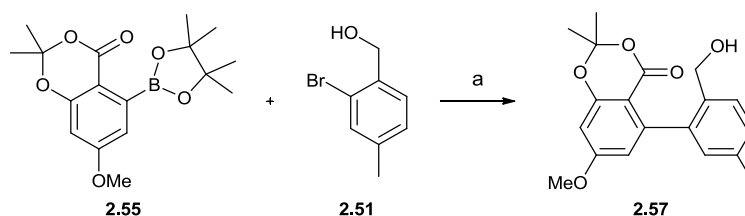
Reagents and Conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h, 56%

Scheme 42

The reaction proceeded in moderate yield and the uncyclised, protected product **2.57** was isolated following column chromatography. The uncyclised product was confirmed by ^{1}H NMR through the absence of a signal at approximately 10.5 ppm for the hydrogen bonded phenol on the left hand ring, and the addition of two singlets at 1.78 ppm and 1.74 ppm indicating the presence of the additional methyl groups on the acetonide protecting group. There were five signals in the aromatic region as expected, as well as a singlet at 3.85 ppm for the methyl group on the right hand ring and a multiplet between 4.34-4.42 ppm indicating the benzyl CH$_2$. We then proceeded to cyclise the lactone ring through addition of base. We hoped the base would deprotonate the benzyl alcohol which would then attack the acetonide carbonyl as proposed in the Suzuki coupling reactions to prepare **2.45** and **2.58** (Scheme 43).
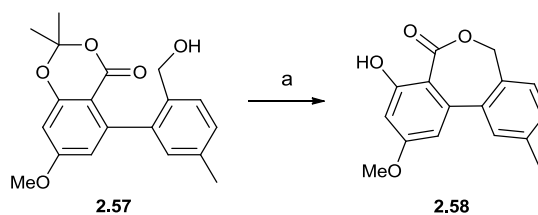
Reagents and conditions: a) NaH, THF, rt, 5 h, 50%

Scheme 43

The uncyclised compound **2.57** was stirred in a suspension of NaH in THF for 5 h, after which aqueous workup and column chromatography yielded 50% of the desired product

2.58 as an off white solid. Only one compound was observed in the ¹H NMR spectrum, with only 5 signals for the 5 aromatic protons.

One of the aims of the project was to try to gather information about the SAR of ulocladol A. Having established that both the boronic ester starting material and the unprotected bromobenzyl alcohol were stable and reactive under Suzuki coupling conditions, we began to target analogues based on commercially available starting materials, ease of synthesis of starting materials and location and functionality of substituents. From the earlier attempted synthesis of the bromo substituted esters we had a supply of 2-bromo-3,4,5-trimethoxybenzyl alcohol, which could be coupled with the boronic ester to yield an analogue with substituents in the same location as ulocladol A. The boronic ester was coupled with 2-bromo-3,4,5-trimethoxybenzyl alcohol under Suzuki coupling conditions (Scheme 44).
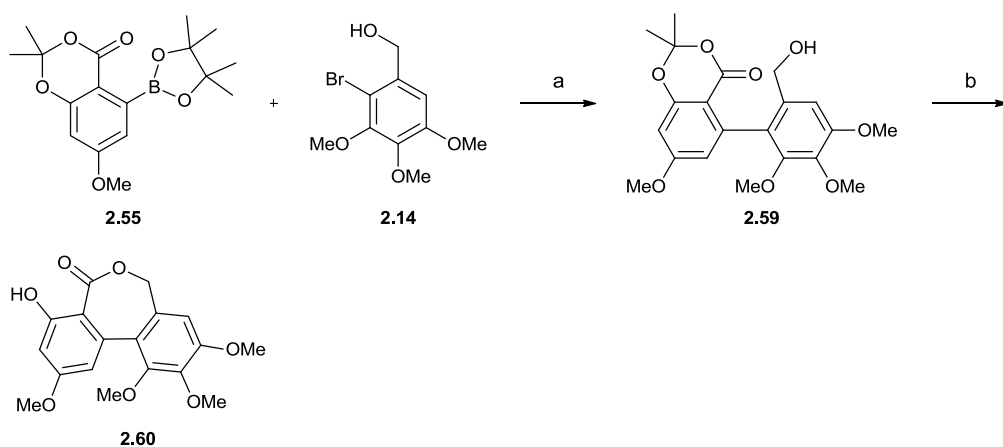
Reagents and conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h; b) NaH, THF, rt, 5 h, 22% over 2 steps

Scheme 44

Coupling of **2.55** with **2.14** produced a mixture of uncyclised product **2.59**, cyclised product **2.60**, the impurity observed in the boronic ester starting material and unreacted **2.14**. As the mixture could not be separated by column chromatography it was further reacted with NaH in the hope that this would yield only cyclised product **2.60**. The reaction successfully transformed the uncyclised product **2.59** to cyclised product **2.60** in 22% yield over two steps and removed the impurity from the boronic ester starting material. However, the unreacted starting material was not removed. The impurity could

not be removed with acid and base washes or through recrystallisation. The product was confirmed by mass spectrometry with a signal at 369 m/z for the [M+Na]$^+$ mass ion.

Having successfully prepared the trimethoxy analogue, we proposed that it would be possible to prepare ulocladol A by selectively removing methoxy groups from the right hand ring. A literature procedure by Reeder *et al.* reported the use of boron tribromide to demethylate a methoxy group on a hexasubstituted benzene ring (Scheme 45).[54]

Reagents and Conditions: a) BBr$_3$, CH$_2$Cl$_2$, -78 °C, 70%
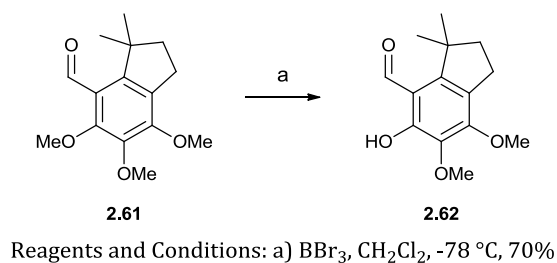
Scheme 45

Following this procedure we attempted to selectively demethylate two of the methoxy groups of the trimethoxy analogue **2.60**, in the 4 and 5 positions of the right hand ring using boron tribromide (Scheme 46).

Reagents and conditions: a) BBr$_3$, DCM, -78 → 0 °C, 0.5 to 16 h
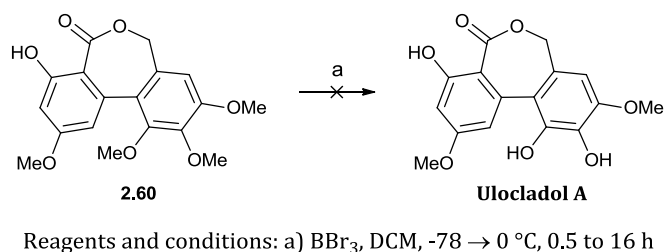
Scheme 46

However the reaction was unsuccessful and no product was isolated. We repeated the reaction, extending the reaction time and increasing the temperature of the reaction, but no demethylation of any of the methoxy groups was observed and only starting material was isolated.

As the demethylation reaction had been unsuccessful, we decided to concentrate on the SAR of ulocladol A by continuing to prepare analogues with comparable functional groups.

We were able to purchase 2-bromo-5-methoxybenzoic acid (**2.63**) which we successfully reduced to the benzyl alcohol **2.64** in 87% yield (Scheme 47).
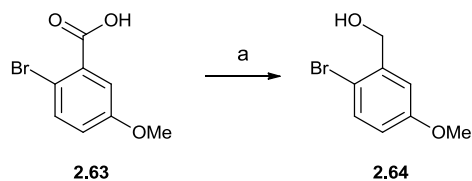
Reagents and Conditions: a) NaBH$_4$, ZrCl$_4$, THF, rt, 16 h, 87%

Scheme 47

We chose this benzyl alcohol because, when coupled with the boronic ester, it would yield an analogue with a methoxy group in the same position on the right hand ring as the natural product. When comparing the biological activity of this analogue with ulocladol A we will be able to determine whether the absent phenols on the right hand ring play a part in the biological activity of the natural product. The next step in the synthesis was to couple 2-bromo-5-methoxybenzyl alcohol (**2.64**) with the boronic ester intermediate **2.55** (Scheme 48).
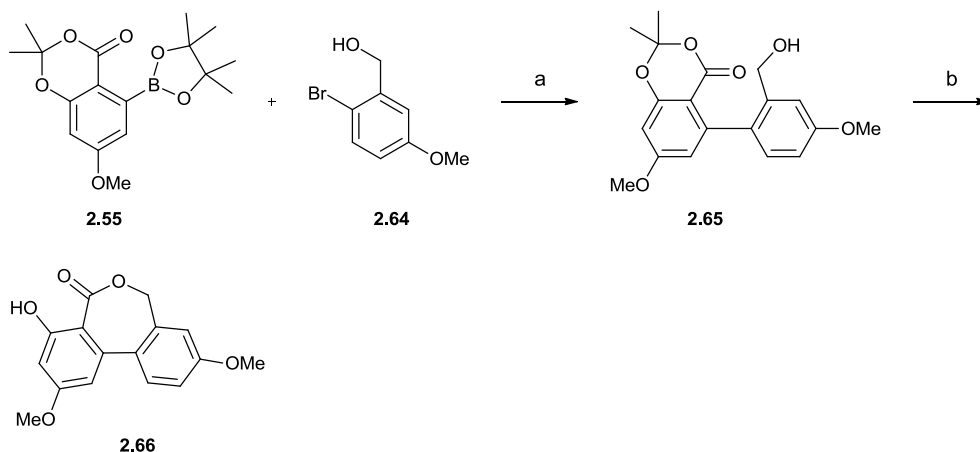
Reagents and conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h, 21%; b) NaH, THF, rt, 5 h, 62%

Scheme 48

The uncyclised compound **2.65** was isolated in 21% yield, although with impurities observed in the ^{1}H NMR. We decided to cyclise the lactone ring and purify the product at a later stage. The lactone was formed using sodium hydride to deprotect and cyclise furnishing the desired product **2.66** in 62% yield. The product was confirmed by mass

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spectrometry, with a peak at 287 m/z for the [M+H]+ mass ion, and ^{1}H NMR, with 5 signals in the aromatic region, a phenol at 10.59 ppm, 2 methoxy signals and 2 broad singlets at 5.13 ppm and 4.78 ppm for the CH_2.

With five analogues of ulocladol A prepared, it was necessary to prepare the natural product itself in order to have a fair comparison of biological activity. Literature searches revealed a route to a compound which could be used to prepare the natural product. The four-step synthesis yielded the desired product **2.71** in 17% overall yield (Scheme 49).[55-57]
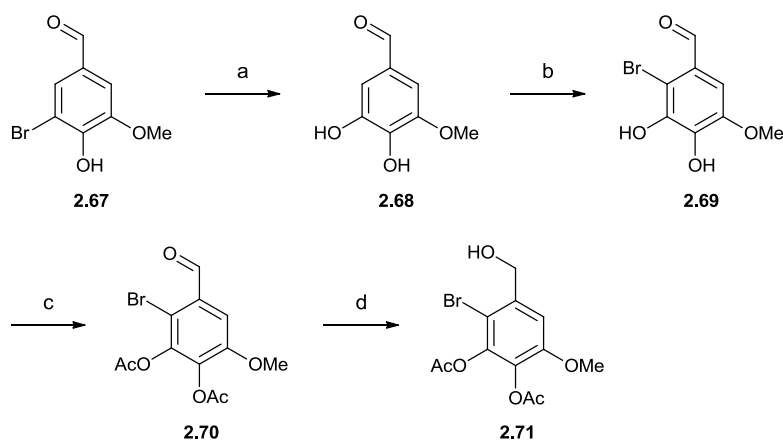
Reagents and conditions: a) NaOH, Cu powder, H_2O, Δ, 24 h, 54%; b) Br_2, acetic acid, rt, 5 min, 41%; c) Ac_2O, Et_3N, DMAP, DCM, rt, 1.5 h, 78%; d) $NaBH_4$, MeOH, 0 °C, 0.5 h, 96%

Scheme 49

The commercially available starting material, 5-bromovanillin (**2.67**), was hydrolytically dehalogenated using catalytic copper powder and aqueous sodium hydroxide to furnish the aldehyde **2.68**. The product was recrystallised from toluene and the structure confirmed by mass spectrometry and ^{1}H NMR. Selective aromatic bromination followed by acetyl protection of the phenolic groups led to the isolation of the protected aldehyde **2.70**. We then successfully reduced the aldehyde to the benzyl alcohol using sodium borohydride in excellent yield. The next step was to couple the bromobenzyl alcohol **2.71** with the boronic ester intermediate using the Suzuki conditions already established (Scheme 50).

Reagents and conditions: a) PdCl$_2$(dppf), K$_3$PO$_4$, THF, Δ, 24 h
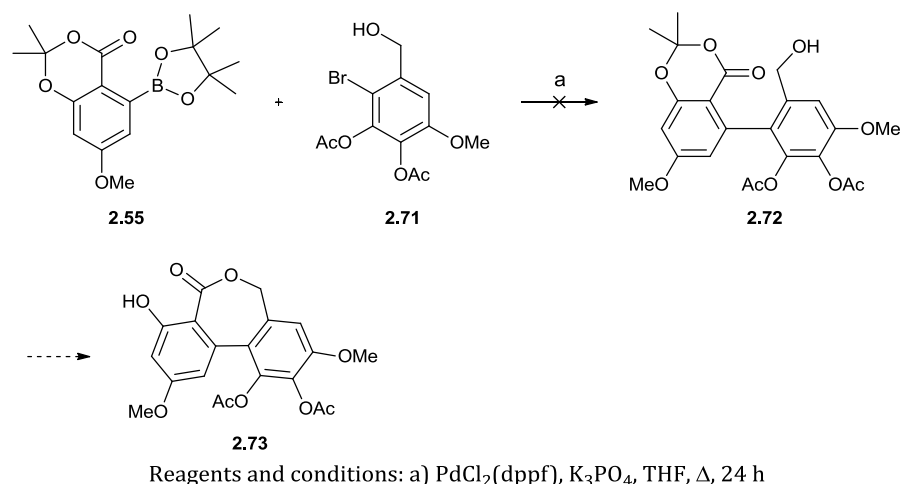
Scheme 50

The reaction was unsuccessful and no product, cyclised or uncyclised, was isolated from the reaction. The only compounds isolated from the reaction were boronic ester starting material and the phenol **2.39** as a by-product of the reaction. We proposed that the failure of the reaction could have several explanations. Firstly, it is possible that the acetyl protecting groups are not stable to the basic conditions of the Suzuki reaction. If this were the case and the acetyl groups were removed prior to the reaction, the free phenolic groups could interfere with the reaction. Secondly, the bulkiness of substituent groups can have an effect on the reaction and. whilst we have previously coupled a penta-substituted bromobenzyl alcohol, **2.14**, under these Suzuki conditions, the yield was very low. In this case, the acetyl groups are bulky groups and this could have had an adverse steric effect on the reaction. Finally, the electronic nature of the acetyl groups may have an effect on the reaction. Unlike the trimethoxy substituted bromobenzyl alcohol, the acetyl groups are electron withdrawing groups resulting in an electron deficient ring. This electronic effect may have adversely affected the reaction.

In order to determine whether the reaction failed because of the sensitivity of the protecting groups to the basic reaction conditions or because of steric hindrance around the ring, we decided to repeat the reaction using different catalyst systems which were more accepting of steric bulk. If the reactions still failed, it would imply that the acetyl protecting groups were not stable under Suzuki coupling conditions. In addition to this we would also be able to test a different catalyst system which could lead to higher yields of the desired product.

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A paper by Buchwald *et al.* reported their research on Suzuki coupling reactions between a boronic acid and a sterically hindered bromide(Scheme 51).[58]
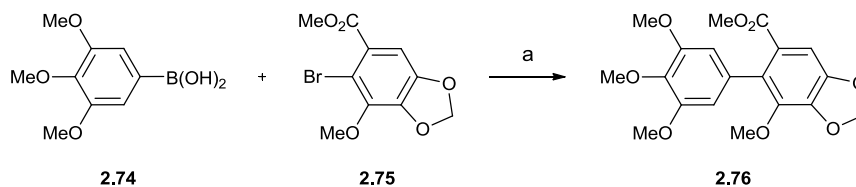
Reagents and Conditions: a) $Pd_2(dba)_3$ (0.0025 mol%), SPhos (0.02 mol%), K_3PO_4 (2 equiv), THF, 80 °C, 20 h, 93%

Scheme 51

SPhos is often used as a ligand in Suzuki coupling reactions containing bulky, sterically hindered reagents. As shown in Scheme 51, Buchwald *et al.* successfully cross-coupled a boronic acid **2.74** with a hindered aryl bromide **2.75**, furnishing the desired biaryl product **2.76** in 93%. The aryl bromide **2.75** bears some resemblance to **2.71**, the aryl bromide required to prepare ulocladol A, with both bromides being penta-substituted with four out of the five functional groups containing oxygen atoms. The location of functional groups, with respect to the bromide, is also the same in both compounds. Due to these similarities we decided to apply the Suzuki conditions to our substrates (Scheme 52).
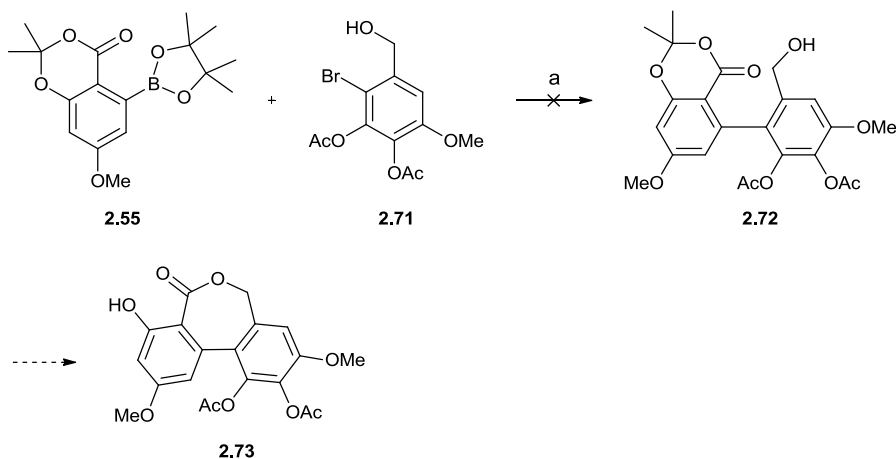
Reagents and conditions: a) $Pd(OAc)_2$, SPhos, K_3PO_4, THF, Δ, 6 h

Scheme 52

We attempted to couple the boronic ester **2.55** with the aryl bromide **2.71** but were unsuccessful. Mass spectrometry of the crude reaction mixture showed no mass ion peaks for either the cyclised or uncylised products and the only recognisable peak in the spectrum was that of the boronic ester starting material. Crude ^{1}H NMR confirmed this

with no peaks between 4 and 6 ppm which is the region where the protons from the CH_2, either cyclised into the lactone ring or as the benzyl alcohol, are usually observed. In addition to the lack of desired product, there was also no indication of the presence of the bromobenzyl alcohol starting material **2.71** in the crude reaction mixture. This implied that the acetyl protecting groups were problematic in the Suzuki coupling reaction and it would be necessary to determine which protecting groups could be used successfully.

Due to the similarities between our substrates and those used by Buchwald *et al.*, we decided to test the reported reaction conditions on other bromobenzyl alcohols in the hope that the reaction yields could be improved. Commercially available 2-bromo-4,5-dimethoxybenzyl alcohol (**2.77**) was coupled with the boronic ester **2.55** using the reported conditions (Scheme 53).

Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_3PO_4, THF, Δ, 21 h, 34%
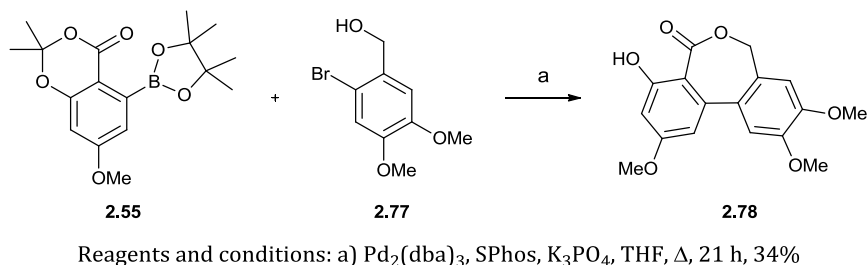
Scheme 53

Analysis of the crude material, with the presence of characteristic signals in the ^{1}H NMR spectrum for the phenol and benzylic CH_2 at 10.69 ppm and 5.02 ppm respectively, suggested that the lactone ring had cyclised *in situ*. Purification by column chromatography yielded the desired product **2.78** with an impurity which ran at the same Rf. We were unable to separate this impurity by column chromatography, and instead used acid and base washes. The impurity was determined to be unreacted bromobenzyl alcohol. The clean product was confirmed by mass spectrometry with a mass ion peak at m/z 339 [M+Na][+]. ^{1}H NMR of the pure compound showed four signals in the aromatic region, integrating as four protons, and three singlets between 3.90 – 3.97 ppm for the three methoxy groups, in addition to the signals at 10.69 and 5.02 ppm.

Although the yield of this reaction was still low, the cyclisation step occurred *in situ*, negating the need for an additional step in the reaction. For this reason we decided to continue with the Buchwald conditions for other bromobenzyl alcohols.

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We were able to purchase a precursor for a regioisomer of the bromobenzyl alcohol **2.77**. The ester, methyl-3,5-dimethoxybenzoate (**2.79**), was transformed into the desired bromobenzyl alcohol in two steps (Scheme 54).[59]
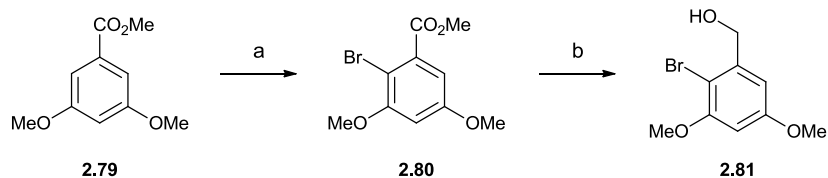
Reagents and Conditions: a) NBS, MeCN, 0 °C, 7 h, 80%; b) DIBAL, THF, 0 °C, 0.5 h, 85%

Scheme 54

Methyl-3,5-dimethoxybenzoate (**2.79**) was selectively brominated using *N*-bromosuccinimide to yield methyl-2-bromo-3,5-dimethoxybenzoate (**2.80**) in 80%. The ester was then reduced using DIBAL to furnish 2-bromo-3,5-dimethoxybenzyl alcohol (**2.81**) in 70% yield. The structures of both products were confirmed by mass spectrometry and NMR spectroscopy. We then went on to couple the bromobenzyl alcohol **2.81** with the boronic ester **2.55** (Scheme 55).
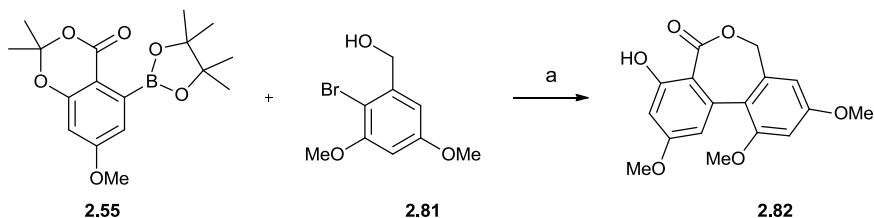
Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_3PO_4, THF, Δ, 21 h, 8%

Scheme 55

The desired, cyclised product **2.82** was isolated in 8% yield after purification by column chromatography and acid and base washes to remove additional impurities. The molecular weight was confirmed by mass spectrometry with a peak at m/z 339 for the [M+Na]⁺ molecular ion. In addition, NMR spectrometry confirmed the structure with characteristic peaks of: a singlet at 10.18 ppm for the phenolic OH group on the left hand ring; four doublets between 6.54 – 6.82 ppm for the four aromatic protons; two broad doublets at 5.11 ppm and 4.82 ppm for the CH_2 and three singlets between 3.80 – 3.88 ppm for the three methoxy groups.

As well as determining the SAR of ulocladol A, one of the aims of the project was to prepare an analogue which was not only more biologically active but also more bioavailable. With this in mind, we looked into purchasing bromobenzyl alcohols which could increase biological activity, change the Log P or give us further insight into SAR. We were able to purchase four bromobenzyl alcohols which could be coupled with the boronic ester using the Suzuki coupling conditions reported by Buchwald *et al*. The first coupling was between the boronic ester and a *para*, nitrile substituted bromobenzyl alcohol. The nitrile group occupies the same position as one of the phenol groups on the right hand ring of ulocladol A. Whilst it is not a hydrogen bond donor it could accept hydrogen bonds and will give an idea as to whether it is necessary to have a hydrogen bond donor group in this position (Scheme 56).
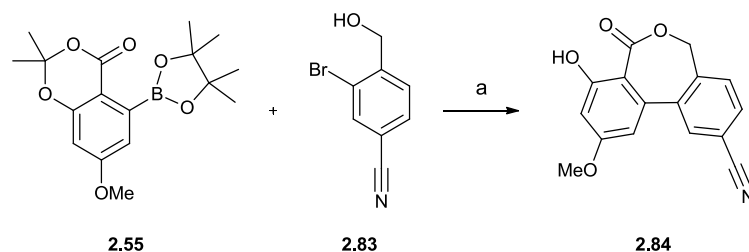
Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_3PO_4, THF, Δ, 21 h, 6%

Scheme 56

The reaction was successful and the desired cyclised product **2.84** was isolated in 6% yield. The molecular weight of the product was confirmed by mass spectrometry with a mass ion signal corresponding to a $[M+Na]^+$ molecular ion of m/z 304. The structure was confirmed by ^1H NMR with the presence of characteristic signals at: 10.56 ppm for the phenolic OH; four doublets and one doublet of doublets in the aromatic region corresponding to the five aromatic protons and two broad signals at 5.16 ppm and 5.02 ppm for the CH_2 protons.

The second and third bromobenzyl alcohols to be coupled with the boronic ester were both fluorine derivatives: one with a trifluoromethyl substituent **2.85b** and the other with a fluoro substituent **2.85a** (Scheme 57).

Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_3PO_4, THF, Δ, 21 h
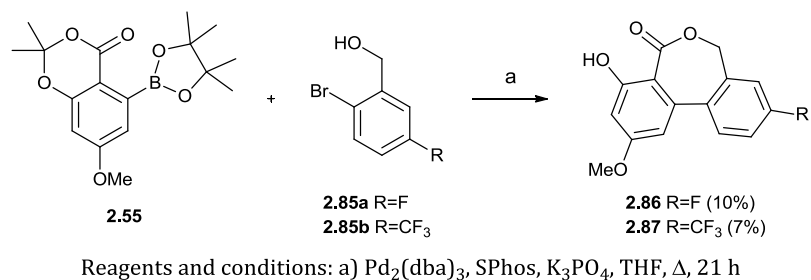
Scheme 57

The two analogues were successfully prepared using the conditions reported by Buchwald *et al*. The correct structures were confirmed by mass spectrometry and ^{1}H NMR with the expected characteristic peaks in the ^{1}H NMR spectra and the corresponding molecular ion signals in the mass spectra. ^{13}C NMR spectrometry for both compounds showed splitting due to fluorine half spin. The greatest effect was seen in the ^{13}C NMR spectrum for the fluoro substituted analogue **2.86**, with splitting all around the right hand ring. The ipso carbon was split into a doublet with a coupling constant of 250.5 Hz; the *ortho* carbons were split into doublets with coupling constants of 21.4 Hz and 22.1 Hz; the *meta* carbons were split into doublets with coupling constants of 8.3 Hz and 7.8 Hz and the *para* carbon was split into a doublet with a coupling constant of 3.4 Hz.

The effect of the fluorine half spin was less apparent in the ^{13}C NMR spectrum of the trifluoromethyl substituted analogue **2.87** with the splitting only apparent on the *ipso* and *ortho* carbons. The signals were split into quartets with a coupling constant of 33.0 Hz for the *ipso* carbon and coupling constants of 3.6 Hz and 3.7 Hz for the *ortho* carbons. No signal was observed for the carbon of the trifluoromethyl group itself.

The fourth bromobenzyl alcohol purchased was (6-bromo-1,3-benzodioxol-5-yl)methanol (**2.88**) which was successfully coupled with the boronic ester (Scheme 58).
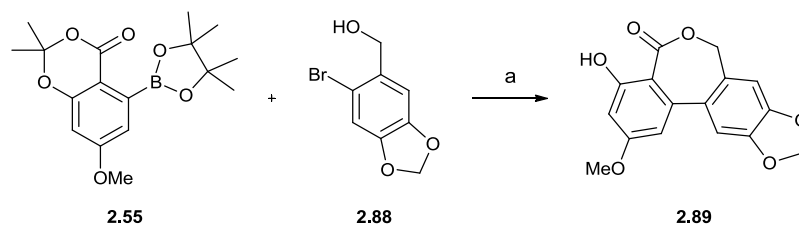
Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_3PO_4, THF, Δ, 21 h, 5%

Scheme 58

The product was confirmed by mass spectrometry, with a peak at m/z 323 corresponding to the $[M+Na]^+$ molecular ion, and ^1H NMR, with characteristic peaks at: 10.59 for the phenolic OH; two doublets and two singlets for the four aromatic protons; a singlet at 6.06 ppm for the oxazole CH_2; two doublets at 5.10 ppm and 4.83 ppm, with coupling constants of 11.7 Hz and 11.5 Hz respectively, for the CH_2 and a singlet at 3.89 ppm for the methoxy group on the left hand ring.

Having established that it was possible to use the conditions reported by Buchwald *et al.* to couple the pinacol ester with bromobenzyl alcohols, we decided to further investigate a coupling which would furnish ulocladol A. We have proposed that the acetyl protected fragment **2.71** was not stable to the basic conditions of the Suzuki reaction so a base stable protecting group would be required. However, prior to investigating protecting groups, we decided to try and couple the pinacol ester with the unprotected bromobenzyl alcohol. We attempted to reduce the aldehyde to the benzyl alcohol **2.90** using sodium borohydride in methanol (Scheme 59).
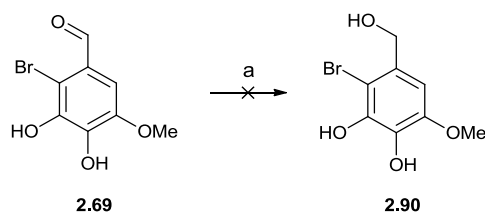
Reagents and conditions: a) $NaBH_4$, MeOH, 0 °C, 0.5 h

Scheme 59

Unfortunately the reaction was unsuccessful and no product could be seen by mass spectrometry or NMR spectroscopy. As TLC suggested that the crude product did not move from the baseline in a 20% methanol in DCM solvent system, the reaction was abandoned.

Instead we decided to try and couple the unprotected aldehyde **2.69** with the pinacol ester (Scheme 60).

Reagents and conditions: a) Pd$_2$(dba)$_3$, SPhos, K$_3$PO$_4$, THF, Δ, 21 h
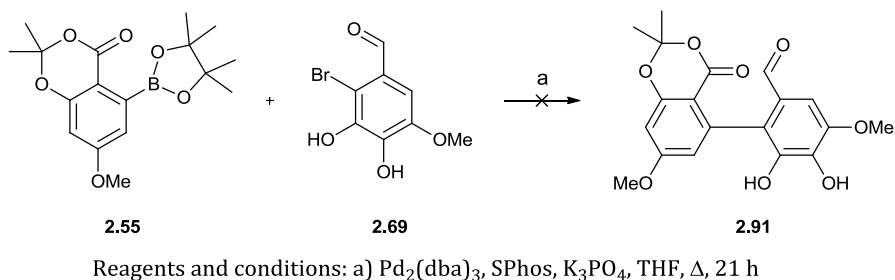
Scheme 60

However the reaction was unsuccessful and neither product **2.91** nor starting material were isolated. Having established that we were unable to either couple the unprotected aldehyde with the pinacol ester or reduce the aldehyde to the unprotected benzyl alcohol, we decided to protect the catechol with a protecting group which would neither be sterically hindering nor base labile. With these considerations in mind, we decided to try and protect the catechol with a linked protecting group which would reduce steric bulk and allow the Suzuki coupling to take place. A group which fits these criteria is the acetonide group. We hoped to use dimethoxypropane to form the protected catechol **2.92**. Several different sets of reaction conditions were attempted and these are shown below (Table 4).
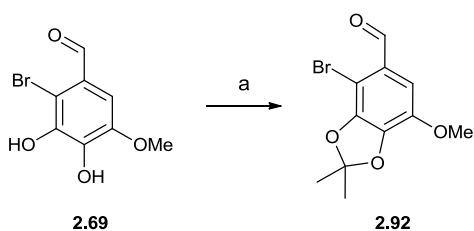
Reagents and conditions: a) See below

Entry	Reagents	Solvent	Conditions	Method	Result
1	DMP, P$_2$O$_5$[60]	Toluene	Reflux, 14 h	Soxhlet extractor	No product
2	DMP, p-TsOH.H$_2$O, acetone[61]	Toluene	Reflux, 86 h	Dean-Stark	No product

3	Acetone, P_2O_5[62]	Toluene	75 °C, 3 h	Thermal	No product
4	DMP, p-TsOH.H_2O, acetone[63]	Toluene	Reflux, 16 h	Soxhlet extractor	No product
5	Acetone, p-TsOH.H_2O[64]	Toluene	Reflux, 24 h	Dean-Stark	No product
6	DMP, p-TsOH.H_2O, acetone[63]	Benzene	Reflux, 16 h	Soxhlet extractor	No product
7	Acetone, p-TsOH.H_2O[64]	Benzene	Reflux, 24 h	Dean-Stark	No product

Table 4 – Acetonide protecting group conditions

Unfortunately, all of the reaction conditions attempted using toluene as a solvent were unsuccessful, entry 1-5. Several of the reported methods for protecting catechols with acetonides use benzene as a solvent instead of toluene. This is because benzene is a better azeotrope for water than toluene so when Dean-Stark or Soxhlet extractor apparatus are used, water is removed more effectively from the reaction vessel. For this reason we decided to retry two sets of conditions, entries 6 and 7, using benzene as a solvent. However, the reactions were still unsuccessful resulting in the investigation into different protecting groups.

A paper by Mayor *et al.* reported investigations into a variety of different protecting groups for catechols.[65] Following the literature procedures we attempted to protect the catechol with two different protecting groups (Scheme 61).
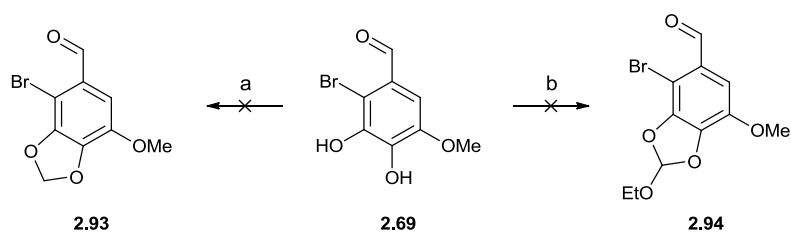
Reagents and Conditions: a) NaOH, dry DCM, DMSO, 120 °C, 20 h; b) Triethylformate, amberlyst 15, toluene, Δ, 18 h

Scheme 61

Despite literature precedence, the reactions were again unsuccessful, and neither the protected catechol **2.93** nor **2.94** were isolated.

A literature procedure reported the use of a silyl group to protect a catechol functionality.[66] Following the reported reaction conditions we attempted to protect the catechol **2.69** using a diisopropylsilyl protecting group **2.95** (Scheme 62).
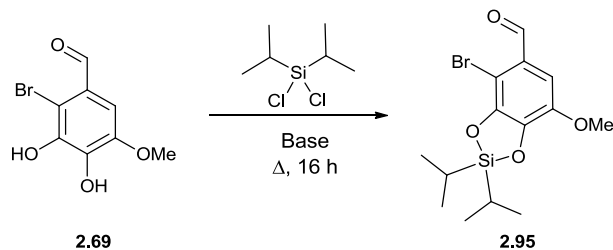
Scheme 62

We tried the reaction with both triethylamine and imidazole as the base, as well as with and without the additive HOBt.[66, 67] Unfortunately, we were unable to isolate any product so investigation into a silyl protecting group was discontinued. Instead we focused on protecting the catechol with either a boronate or a benzylidene protecting group (Scheme 63).[64, 68]
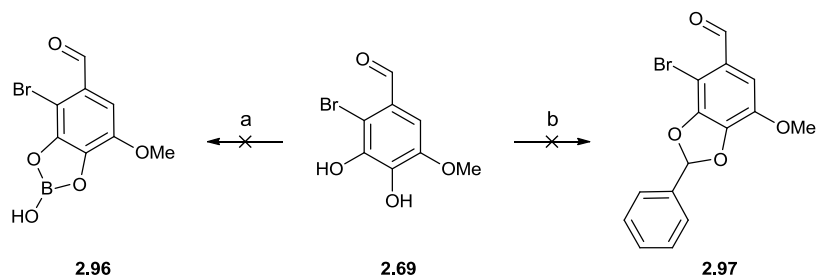
Reagents and Conditions: a) B(OH)$_3$, toluene, Dean-Stark, Δ; b) Benzaldehyde, p-TsOH, benzene, Δ

Scheme 63

However both methods were unsuccessful and we were unable to protect the catechol with either the boronate **2.96** or benzylidene **2.97** protecting group.

Due to the disappointing lack of success in protecting the catechol with a linked protecting group, we decided to revise our attempts to protect the catechol with individual protecting groups.

The Boc protecting group is a well-known protecting group and is widely used in the protection of both amines and alcohols. As the Boc group is base stable, due to it being a carbonate and sterically hindered, it should not be susceptible to the basic conditions of

the Suzuki reaction. Using a standard Boc protection procedure we successfully protected the catechol in 95% (Scheme 64).[69]
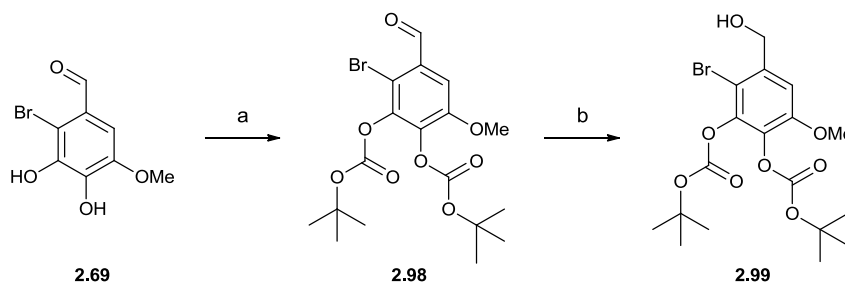
Reagents and conditions: a) (Boc)$_2$O, DMAP, DCM, rt, 2 h, 95%; b) NaBH$_4$, EtOH, -50 °C, 0.5 h, 99%

Scheme 64

The benzaldehyde **2.98** was then reduced to the benzyl alcohol **2.99** using sodium borohydride at -50 °C. The low temperature was required to prevent deprotection of the catechol which was observed at higher temperatures. With the benzyl alcohol in hand we then went on to react with the boronic ester (Scheme 65).
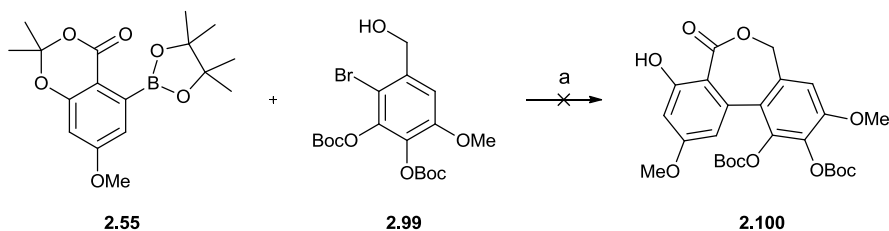
Reagents and conditions: a) Pd$_2$(dba)$_3$, SPhos, K$_3$PO$_4$, THF, Δ, 21 h

Scheme 65

However, no product or precursor of the product was isolated from the reaction mixture. Mass spectrometry also suggested that neither of the starting materials were present. We proposed that the Boc protecting groups were not stable to the Suzuki coupling conditions resulting in no product forming.

It was decided to investigate a more stable protecting group for the catechol. A literature procedure reported the use of iodoacetonitrile to alkylate a hydroxyl group. We proposed that we could protect the catechol with nitrile groups which could then be removed following the Suzuki coupling reaction. Following the literature procedure by Broka *et al.* we successfully alkylated the catechol to yield the desired product **2.101** (Scheme 66).[70]

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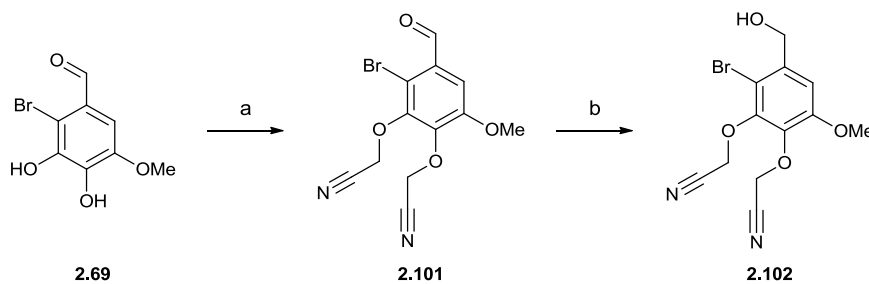
Reagents and Conditions: a) ICH$_2$CN, K$_2$CO$_3$, DMF, 60 °C, 16 h, 32%; b) NaBH$_4$, EtOH, 0 °C, 0.5 h, 92%

Scheme 66

Although the alkylation step was low yielding we continued with the reduction step preparing the desired bromobenzyl alcohol **2.102** in 92%. We repeated the Suzuki reaction under the same conditions as attempted with the Boc protected bromobenzyl alcohol, unfortunately this reaction was also unsuccessful.

We were unsure whether the lack of success with the Suzuki coupling reaction was due to the protecting group, the steric hindrance or the Suzuki conditions. Therefore, we decided to try the coupling reaction with a less sterically hindered bromobenzyl alcohol. We were able to purchase 4-formyl-2-methoxyphenyl acetate (**2.103**) which could be brominated and protected to furnish a similar bromobenzyl alcohol to that required to prepare ulocladol A, although without the phenol *ortho* to the bromine (Scheme 67).
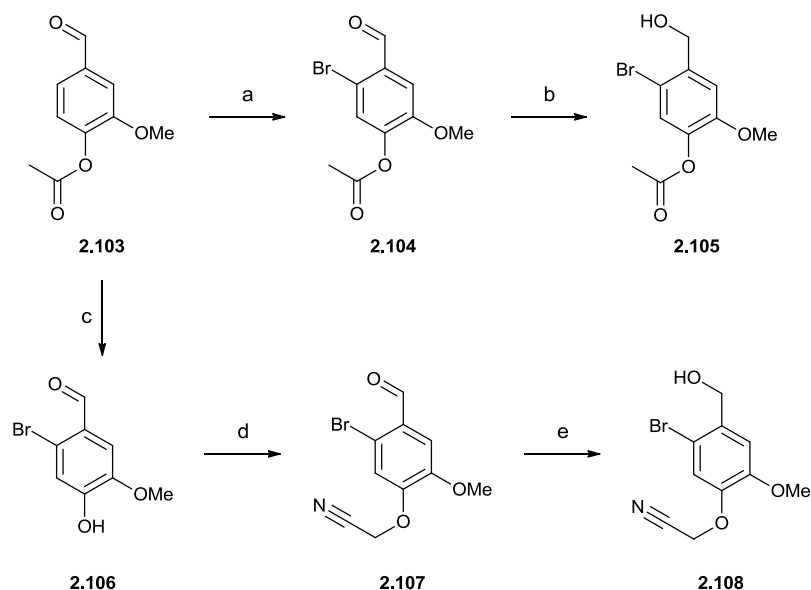
Reagents and conditions: a) KBr, Br$_2$, H$_2$O, rt, 10 h, 94%; b) NaBH$_4$, EtOH, -70 °C, 0.5 h, 82%; c) KBr, Br$_2$, H$_2$O, rt, 16 h, then 6N HCl, 90 °C, 9 h, 59% over 2 steps; d) ICH$_2$CN, K$_2$CO$_3$, DMF, 60 °C, 16 h, 57%; e) NaBH$_4$, EtOH, 0 °C, 0.5 h, 59%

Scheme 67

We selectively brominated **2.103** *para* to the methoxy group to yield the brominated benzaldehyde **2.104** in 94%. The benzaldehyde was then reduced with sodium borohydride to yield the acetyl protected phenol **2.105** in 82%. We also prepared the cyanomethyl protected phenol **2.108** in 20% over four steps.

The next step was to couple the two bromides with the pinacol ester **2.55**, using the Suzuki coupling conditions reported by Buchwald (Scheme 68).
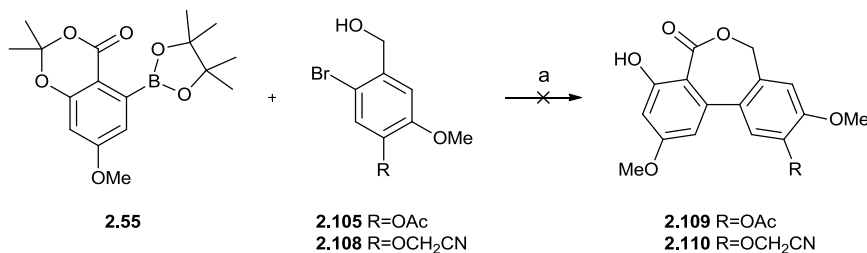
Reagents and conditions: a) Pd$_2$(dba)$_3$, SPhos, K$_3$PO$_4$, THF, Δ, 21 h

Scheme 68

Unfortunately the Suzuki coupling reactions were both unsuccessful and we were unable to prepare either the acetyl protected product **2.109** or the nitrile protected product **2.110**. As a cyanomethyl protected phenol would not have been susceptible to the basic

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conditions of the Suzuki reaction, we considered that the problems observed in the Suzuki coupling reaction were not caused by the protecting group.

Despite successfully preparing six analogues of ulocladol A using the conditions reported by Buchwald *et al*., we had observed several problems. Firstly, the Suzuki coupling reaction was very messy and required extensive purification in order to obtain the desired product. As well as purifying by column chromatography multiple times, it was necessary to undertake acid and base washes and extractions in order to isolate a small quantity of product. Secondly, the reactions were very low yielding, with five out of the six analogues being prepared in equal to or less than 10%. Finally, the conditions were still not applicable to all substrates and we have not been able to prepare ulocladol A using these conditions. Therefore we decided to investigate a catalyst system which could be applicable to a much broader range of bromobenzyl alcohols and enable us to prepare ulocladol A. We chose to use the boronic ester intermediate **2.55** and the mono substituted bromobenzyl alcohol, 2-bromo-4-methylbenzyl alcohol **2.51**, as our test substrate system (Scheme 69).
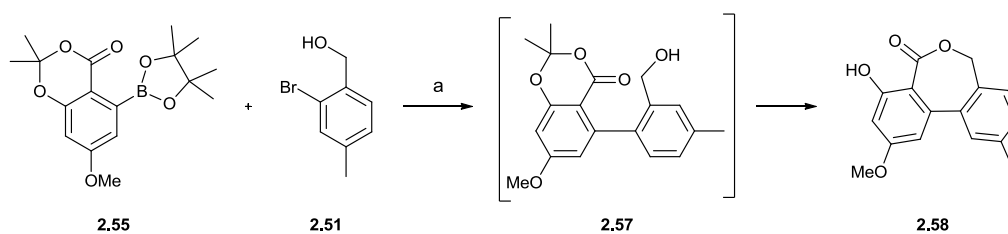
Reagents and Conditions: a) See Table 5 for conditions and yields

Scheme 69

We decided to use this substrate system because the methyl group is not strongly electron withdrawing or donating, bulky or sterically hindering in the coupling reaction. In addition to this, it is inexpensive and easily prepared. We tested a variety of catalyst systems which are summarized in the table below (Table 5).

Entry	Catalyst	Base	Solvent	Conditions			Result
				Temp	Time	Method	
1	Pd(PPh$_3$)$_4$ (5 mol %)	NaHCO$_3$ (2M)	Dioxane/H$_2$O	130 °C	10 min	Microwave	**2.58** (70%)
2	PdCl$_2$(dppf) (5 mol %)	K$_2$CO$_3$ (2M)	Acetonitrile/H$_2$O	130 °C	10 min	Microwave	**2.57** (25%)
3	Pd(PPh$_3$)$_4$ (5 Mol %)	NaHCO$_3$ (2M)	Dioxane/H$_2$O	100 °C	16 h	Sealed tube	**2.58** (36%)
4	PdCl$_2$(dppf) (5 mol %)	K$_2$CO$_3$ (2M)	Acetonitrile/H$_2$O	100 °C	16 h	Sealed tube	**2.58** (5%)
5	PEPPSI-*i*Pr (5 mol %)	Cs$_2$CO$_3$ (2M)	Dioxane/H$_2$O	100 °C	16 h	Sealed tube	**2.58** (32%)
6	PEPPSI-*i*Pr (2 mol %)	K$_3$PO$_4$ (2M)	Dioxane/H$_2$O	rt	16 h + 1 h 60 °C	Thermal	Recovered starting material
7	PEPPSI-*i*Pr (2 mol %)	Cs$_2$CO$_3$	DMF	100 °C	16 h	Thermal	No product
8	PEPPSI-*i*Pr (6 mol %)	CsF (2M)	Dioxane/H$_2$O	100 °C	16 h	Thermal	No product
9	PdCl$_2$(dppf) (5 mol %)	Cs$_2$CO$_3$	DMF	100 °C	16 h	Thermal	No product
10	PdCl$_2$(dppf) (5 mol %)	K$_2$CO$_3$ (2M)	Acetonitrile/H$_2$O	75 °C	16 h	Thermal	No product

Table 5 – Testing of catalyst conditions for Suzuki reaction

The catalyst testing showed some interesting results and demonstrated that not only was the catalyst system important, but also the mode of reaction. If we compare entries 2, 4 and 10 we can see that all three use $PdCl_2(dppf)$ as the catalyst, an aqueous 2M K_2CO_3 solution as the base, and acetonitrile as the solvent. However, the mode of reaction, time and temperature are different. Entry 2 was undertaken in the microwave at 130 °C for 10 min and, of the three, reported the highest yield albeit of the uncyclised product **2.57**. Although at a lower temperature and extended reaction time, entry 4 was undertaken in a sealed tube so could exceed the boiling point of acetonitrile. The cyclised product **2.58** was isolated in 5% yield and confirmed to be the desired product by both mass spectrometry and ^{1}H NMR.

The third entry for comparison, 10, was performed under thermal conditions at 75 °C for 16 h, but yielded no product, cyclised or uncyclised. Whilst the lower temperature is likely to adversely affect the yield, it is unlikely to be so detrimental as to yield no product at all. Instead these results implied that the mode of reaction is very important and the outcome is not solely dependent on the catalyst system and base as originally suspected.

The most successful set of conditions were obtained when using $Pd(PPh_3)_4$ as the catalyst, with an aqueous 2M $NaHCO_3$ solution of base, dioxane as the solvent and heating in the microwave. Using this set of conditions, cyclised product **2.58** was obtained in 70% yield, the best result for any Suzuki reaction we had undertaken. We repeated the reaction using the same reagents but heating in a sealed tube at 100 °C for 16 h. Mass spectrometry suggested the presence of cyclised product **2.58**, with 36% product isolated following purification.

In addition to using the microwave, we were interested in testing catalysts which could be used at room temperature. PEPPSI-*i*Pr is a relatively novel catalyst which has had success in Suzuki coupling reactions for a variety of substrates at room temperature.[71, 72] We tested this catalyst with a variety of bases, solvents and conditions but with limited success, entries 6-8. The only successful conditions with this catalyst were those reported in entry 5 where a sealed tube was used. Mass spectrometry showed the presence of cyclised product **2.58** however ^{1}H NMR spectroscopy suggested that this was in minimal yield and the crude was not purified.

Due to the success shown in the microwave, we decided to move on and apply the microwave conditions used in entry 1 to other substrates.

The first novel substrate we applied the conditions to was 2-bromo-4-(trifluoromethyl)benzyl alcohol (**2.111**) which, when coupled with the pinacol ester, would yield a regioisomer of analogue **2.87** (Scheme 70).

Reagents and conditions: a) Pd(PPh$_3$)$_4$, 2M NaHCO$_3$, dioxane, mw, 130 °C, 10 min, 32%
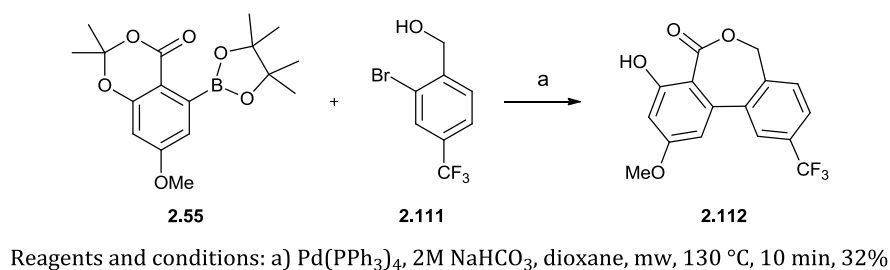
Scheme 70

The product **2.112** was isolated in 32% yield following column chromatography and acid and base washes. Although the acid and base washes were still required to remove bromobenzyl alcohol starting material, the separation of other impurities by column chromatography was improved with fewer co-running spots. The product was confirmed by ^{1}H NMR spectroscopy with the expected broad singlets at 5.21 ppm and 5.05 ppm for the CH$_2$ protons, the phenolic OH signal at 10.64 ppm and 5 aromatic protons between 6.65-7.81 ppm. In addition to this, the ^{13}C NMR showed splitting on the carbons *ipso* and *ortho* to the CF$_3$ group on the right hand ring due to the half spin of fluorine. A ^{19}F NMR also confirmed the presence of fluorine in the molecule with a peak at -62.78 ppm.

Another analogue we were interested in with respect to exploring SAR was the regioisomer of the nitro substituted analogue **2.47**, where the nitro group is substituted *meta* to the carbon-carbon bond rather than *para*. We were able to purchase 2-bromo-5-nitrobenzoic acid (**2.113**) which could be reduced to the benzyl alcohol and then coupled with the boronic ester to result in an analogue substituted *para* to the aryl-aryl bond. The first step was to reduce the carboxylic acid to the benzyl alcohol (Scheme 71).[73]
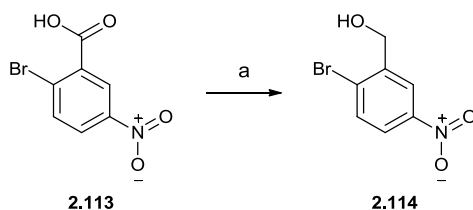
Reagents and conditions: a) NaBH$_4$, BF$_3$.OEt$_2$, THF 0 °C→rt, 16 h, quant.

Scheme 71

2-Bromo-5-nitrobenzyl alcohol (**2.114**) was successfully prepared in quantitative yield as a yellow solid. The product was confirmed by ^{1}H NMR with the presence of three aromatic protons and a singlet at 4.83 ppm for the CH$_2$. The next step was to couple the bromobenzyl alcohol with the pinacol ester using the reported microwave conditions (Scheme 72).
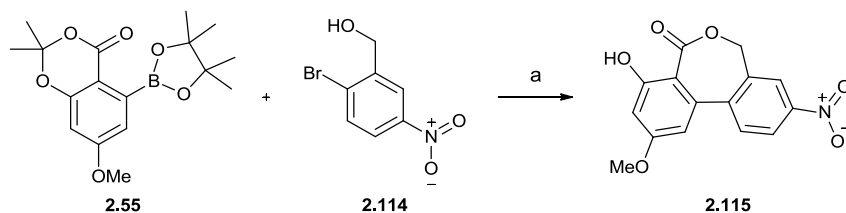
Reagents and conditions: a) Pd(PPh$_3$)$_4$, 2M NaHCO$_3$, dioxane, mw, 130 °C, 10 min, 31%

Scheme 72

The product **2.115** was successfully prepared in 31% yield as a colourless crystalline solid. The structure of the product was confirmed by NMR spectroscopy with five peaks for the aromatic protons between 6.50 ppm and 9.00 ppm, two broad, overlapping singlets at 5.35 ppm for the CH$_2$ and a singlet at 4.10 ppm for the methoxy group on the left hand ring. The corresponding peaks and aromatic protons were identified by COSY NMR data. Unlike analogue **2.47** where the nitro group is substituted *meta* to the aryl-aryl bond, the ^{1}H NMR spectrum suggests that **2.115** is a single compound due to the presence of only one set of signals for the aromatic protons. The molecular weight of the product was confirmed by mass spectrometry with a peak at m/z 356 for the [M+Na+MeOH]+ ion. An X-ray crystal structure also confirmed the correct product structure (Figure 21).
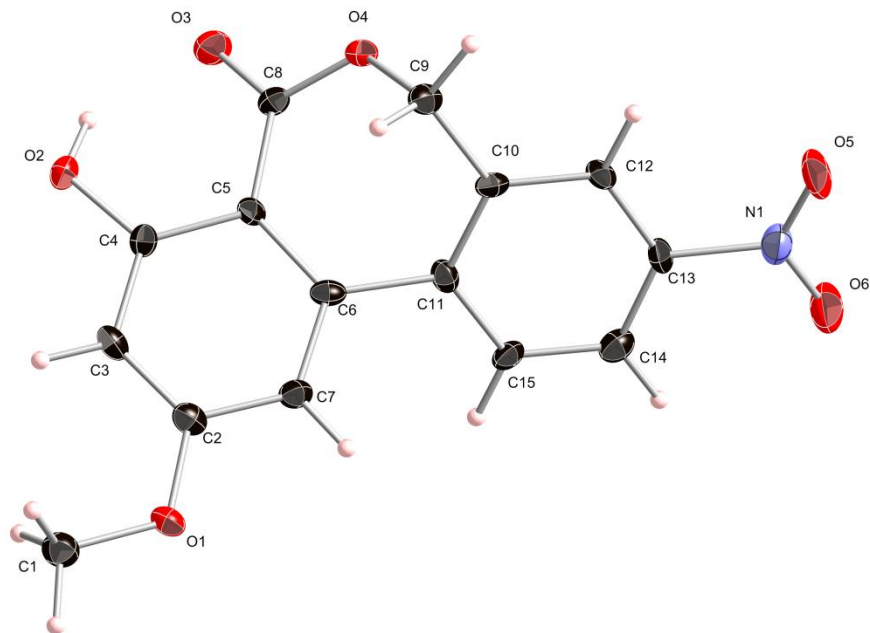
Figure 21 – X-ray crystal structure of 2.115

The nitro group can clearly be seen on the right hand ring, *para* to the carbon-carbon bond. In addition, on the left hand ring, there is an intramolecular hydrogen bond between O(2)–H(2)···O(3). The distance between O(3) and H(2), the hydrogen acceptor bond, is 1.88 Å which is typical for a hydrogen bond of this type. The presence of this hydrogen bond also explains why a sharp singlet is observed in the ^{1}H NMR for the phenolic group.

If we view the structure side-on, the two aryl rings are twisted out of plane with respect to one another (Figure 22).
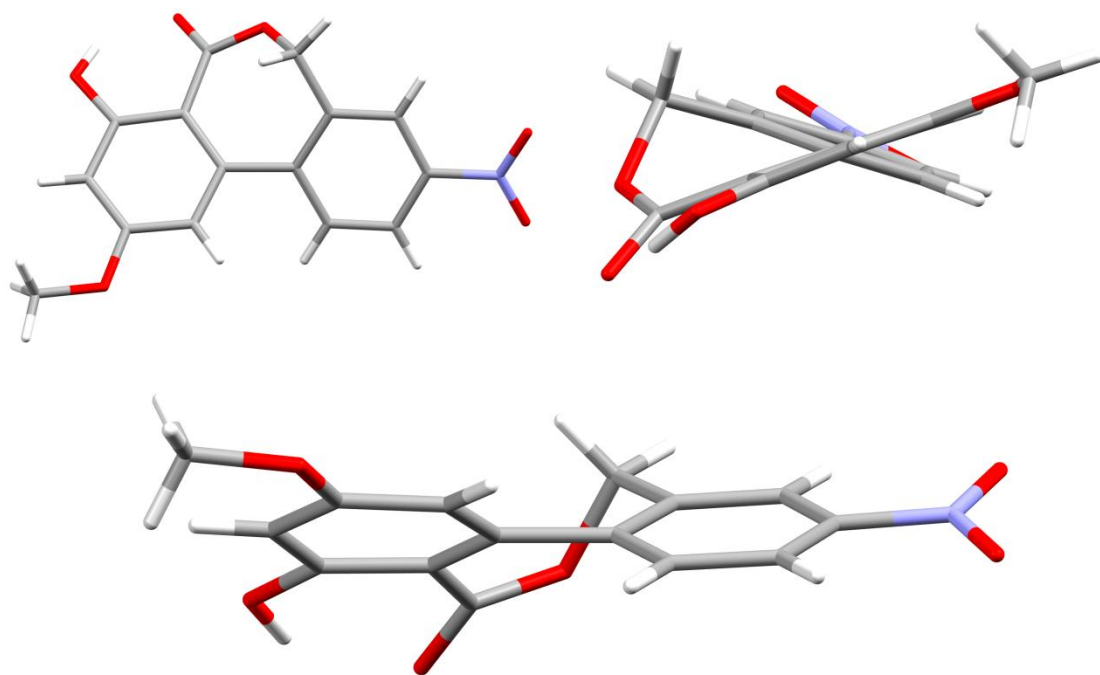
Figure 22 – Rotated views of X-ray crystal structure[74]

The three views of the X-ray crystal structure demonstrate the twist in the molecule about the aryl-aryl bond, with the two aromatic rings being out of plane to one another. This is best seen in the top right structure where we are looking from the left hand side of the molecule back towards the nitro group on the right hand ring. The torsion angle of the internal ring was measured between C(5)-C(6)-C(11)-C(10) and found to be 38.6 °. This rotation would appear to be influenced by the formation of the intra-molecular hydrogen bond, which dictates the arrangement of the kinked seven membered lactone ring and in turn the twist of the two aryl rings.

The ^{1}H NMR spectrum of **2.115** suggests that only one compound is present, unlike the ^{1}H NMR spectrum of the regioisomer **2.47**. One possible explanation for this doubling up of peaks in the ^{1}H NMR spectrum of **2.47** was the presence of a second regioisomer. We suggested that one way of determining if this was the case was to overlay the ^{1}H NMR spectrum of **2.47** with that of a regioisomer (Figure 23).
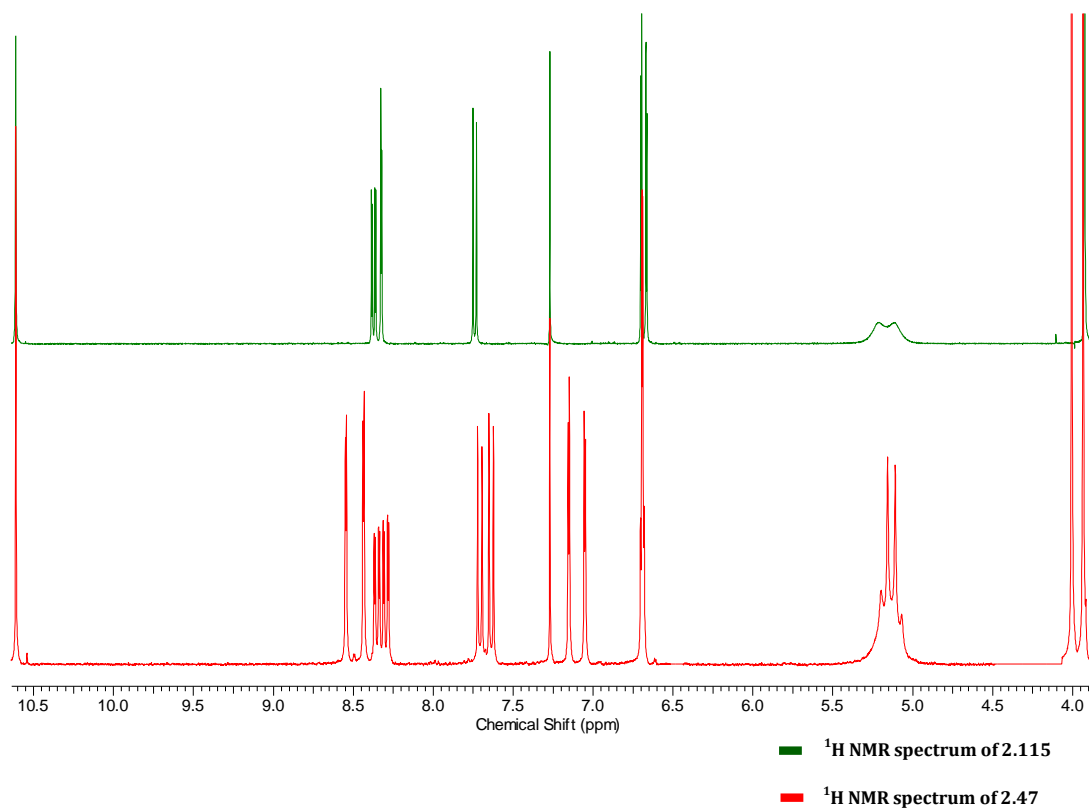
Figure 23 - Overlay of ¹H NMR spectra of 2.47 and 2.115

From the overlaid spectra we can see that the peaks for the phenol overlap exactly. The peak for the methoxy group on the right hand ring of **2.115** also overlaps with one of the peaks associated with a methoxy group in the spectrum of **2.47**. If we look at the aromatic regions of the two spectra, we can see that the peaks for the *para* substituted analogue **2.115** are very similar to the additional set of peaks seen in the spectrum of **2.47**, however the peaks do not exactly overlap. The slight differences seen in the positions of the peaks may be the consequence of two factors: the difference in field strength between the two spectra and secondly, the difference in concentration of the two samples. Unfortunately, due to lack of material, it has not been possible to reanalyse the two compounds under identical conditions.

As nitro functional groups are rarely found in medicinal compounds, due to issues with toxicity, we decided it would be advantageous to prepare a compound which would have similar biological effects without the toxicity generally attributed to a nitro group. One of the possibilities for this is a pyridine ring.

We were able to purchase 3-bromoisonicotinaldehyde (**2.116**) which we successfully reduced to the benzyl alcohol in 90% yield (Scheme 73).

2.116   2.117
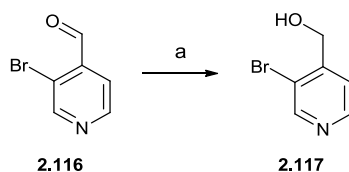
Reagents and conditions: a) NaBH$_4$, EtOH, rt, 1 h, 90%

Scheme 73

We then went on to couple (3-bromopyridin-4-yl)methanol (**2.117**) with the pinacol ester **2.55** using microwave conditions (Scheme 74).

2.55   2.117   2.118
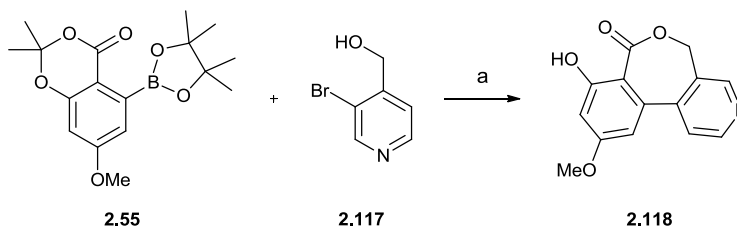
Reagents and conditions: a) Pd(PPh$_3$)$_4$, 2M NaHCO$_3$, dioxane, mw, 130 °C, 10 min, 10%

Scheme 74

The pyridine substituted analogue **2.118** was prepared in 10% yield. The product was confirmed by mass spectrometry, with a signal at 280 m/z for the [M+Na]$^+$ molecular ion, and NMR with the presence of 5 distinct signals for the 5 aromatic protons and a broad CH$_2$ signal at 5.08 ppm.

We were also able to purchase 2-bromo-4-fluorobenzyl alcohol (**2.119**), which we successfully coupled with the pinacol ester **2.55** to furnish **2.120,** a regioisomer of analogue **2.86** (Scheme 75).

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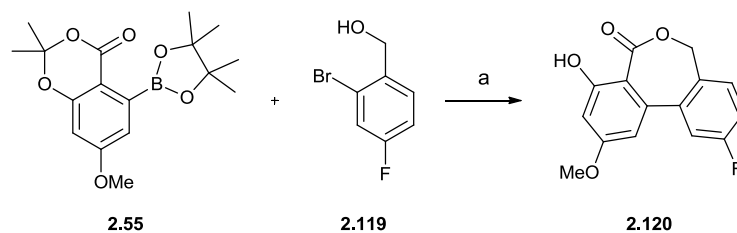
Reagents and conditions: a) Pd(PPh$_3$)$_4$, 2M NaHCO$_3$, dioxane, mw, 130 °C, 10 min, 10%

Scheme 75

Having successfully prepared seventeen analogues of ulocladol A, using a variety of different routes, it was necessary to prepare ulocladol A in order to not only confirm the structure of the natural product, but to also have a sample of the natural product with which to compare the biological activity of the analogues. Therefore this again became the focus of the project.

Unfortunately, prior to commencing with further testing of catechol protecting groups and conditions for the Suzuki coupling reaction, a paper was published by Podlech *et al.* which reported the synthesis of ulocladol A using the same key intermediate **2.55** we have used (Scheme 76).[24]
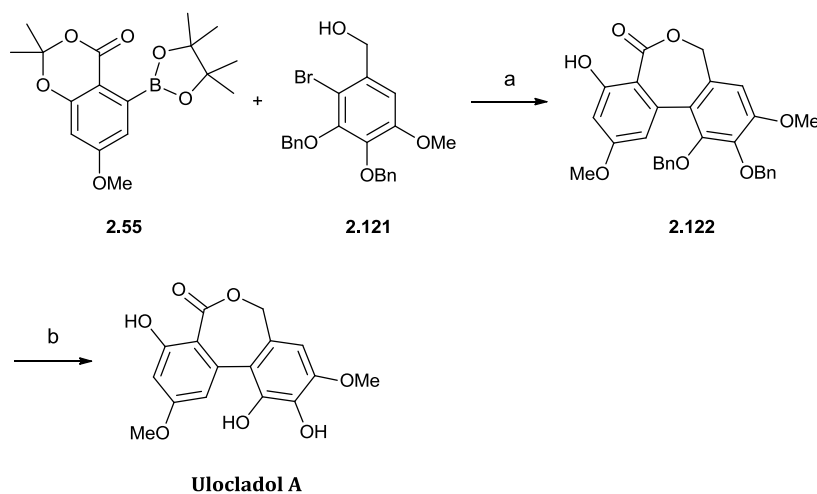
Reagents and conditions: a) Pd(OAc)$_2$, SPhos, Cs$_2$CO$_3$, dioxane/H$_2$O (6:1), 80 °C, 2 h, 65%; b) Pd/C (10-15 mol%), H$_2$, MeOH, rt, 2 h, 60%

Scheme 76

Benzyl protected ulocladol A **2.122** was prepared by reacting the pinacol ester intermediate **2.55** with the benzyl protected bromobenzyl alcohol **2.121** using Suzuki

coupling methodology. Podlech *et al.* used Pd(OAc)$_2$ as the palladium source, SPhos as the ligand, Cs$_2$CO$_3$ as the base and a mixture of dioxane/H$_2$O as the solvent. The reaction was heated at 80 °C for 2 h before being purified by column chromatography to furnish the desired product **2.122** in 65% yield. These conditions are not too dissimilar from those that we used prior to switching to the microwave with one of the major differences being the protecting group of the bromobenzyl alcohol. In this case, Podlech *et al.* used standard hydrogenolysis conditions, reacting benzyl protected **2.122** with hydrogen over Pd/C for 2 h which yielded the desired product in 60%.

This led us to believe that the problems that we faced had been due to the nature of the catechol protecting group. We decided to repeat the reactions reported by Podlech *et al.* in order to prepare ulocladol A. The first step was to prepare the benzyl protected bromobenzyl alcohol (Scheme 77).
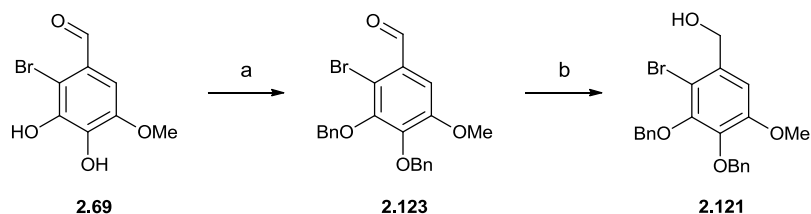
Reagents and conditions: a) NaH, BnBr, DMF, -78 °C, 15 min, 59%; b) DIBAL-H, THF, -78 °C, 2 h, quant.

Scheme 77

2-Bromo-3,4-dihydroxy-5-methoxybenzaldehyde (**2.69**) was protected by reacting with benzyl bromide at -78 °C. The desired product **2.123** was furnished in 59% yield and the correct structure confirmed by comparison of the ^{1}H NMR spectrum with the data reported by Podlech *et al.* Having successfully protected the catechol we then reduced the aldehyde to the corresponding alcohol in order to prepare the desired product **2.121** in quantitative yield.

With the protected bromobenzyl alcohol **2.121** in hand, the next step was to couple with the pinacol ester using the Suzuki conditions reported by Podlech *et al.* (Scheme 78).
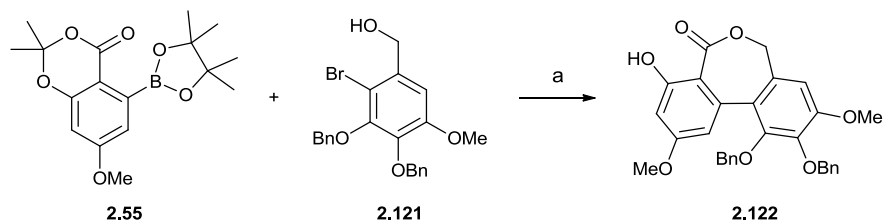
Reagents and conditions: a) Pd(OAc)$_2$, SPhos, Cs$_2$CO$_3$, dioxane/H$_2$O (6:1), 80 °C, 2 h, 54%

Scheme 78

The reaction proceeded in good yield to furnish the desired product **2.122**. The structure was confirmed by comparison of the data with that of the literature.

Having successfully prepared the precursor to ulocladol A we then attempted to deprotect the catechol in order to prepare the natural product. Following the conditions reported by Podlech *et al*. we attempted to remove the benzyl protecting groups using a standard hydrogenation protocol. After 1 h TLC suggested that the reaction had gone to completion due to the absence of starting material. The reaction was stopped and the crude product submitted for ¹H NMR and mass spectrometry. Unfortunately, both ¹H NMR and mass spectrometry suggested that, whilst the desired deprotection had taken place, the reaction had proceeded too far and the lactone ring had broken (Scheme 79).
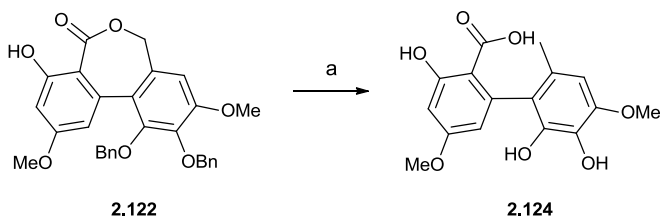
Reagents and conditions: a) H$_2$, Pd/C, MeOH, 1 h

Scheme 79

The mass spectrometry showed no signals which corresponded to either starting material or product, instead showing a mass ion peak of m/z 343 for the [M+Na]+ molecular ion of the reductively cleaved lactone ring.

A signal in the ¹H NMR spectrum at 1.97 ppm, which integrated for three protons, suggested the presence of a methyl group. In addition to this, there was no signal at approximately 5 ppm for the CH$_2$ protons in the lactone ring. This suggested that the

lactone ring had split between the oxygen atom and the CH_2 yielding a carboxylic acid and a methyl group **2.124**.

 While it might be possible to oxidise **2.124** back to a benzylic alcohol and attempt lactone formation, this would add more steps to the synthesis and be inelegant. The reaction was repeated; however it was decided to monitor the reaction every five minutes by TLC to ensure that the lactone ring did not break for a second time. After five minutes the reaction was stopped as TLC suggested that all of the starting material had reacted. ^{1}H NMR and mass spectrometry of the crude product suggested that the lactone ring was still intact and both benzyl groups had been removed (Scheme 80).

Reagents and conditions: a) H_2, Pd/C, MeOH, rt, 5 min, 14%
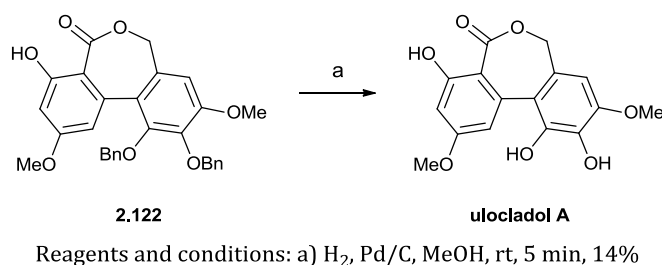
Scheme 80

Ulocladol A was prepared in 14% yield and the structure confirmed by comparison of the ^{1}H and ^{13}C NMR spectra with that of the of the natural product (Table 6).

ulocladol A
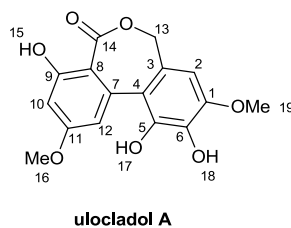
Atom	^{1}H NMR (ppm)				^{13}C NMR (ppm)	
	Found	J (Hz)	Reported[24]	J (Hz)	Found	Reported[24]
1	-	-	-	-	146.3	146.3
2	6.58 (s)	-	6.57 (s)	-	103.1	103.1
3	-	-	-	-	127.1	127.1
4	-	-	-	-	118.8	118.8
5	-	-	-	-	135.9	135.9

6	-	-	-	-	133.8	133.8
7	-	-	-	-	142.0	142.1
8	-	-	-	-	106.9	106.9
9	-	-	-	-	163.0	163.0
10	6.56 (d)	2.5	6.55 (d)	2.5	100.8	100.8
11	-	-	-	-	163.2	163.2
12	6.92 (d)	2.5	6.92 (d)	2.5	110.2	110.2
13	4.80 (d)	12	4.79 (d)	11.7	70.3	70.3
	5.12 (d)	12	5.11 (d)	11.7		
14	-	-	-	-	172.5	172.5
15	10.31 (s)	-	10.29 (s)	-	-	-
16	3.87 (s)	-	3.87 (s)	-	55.5	55.5
17	5.78 (br. s)	-	5.77 (br. s)	-	-	-
18	5.69 (br. s)	-	5.68 (br. s)	-	-	-
19	3.95 (s)	-	3.95 (s)	-	56.3	56.3

Table 6 – Reported and collected ^{1}H NMR and ^{13}C NMR data for ulocladol A

Having successfully prepared ulocladol A using protecting groups which were stable to the Suzuki reaction conditions, we decided to try and change the functional groups on the right hand ring. We were interested in determining whether the phenol and methoxy groups had any effect on the SAR. Previous work in the group had led to the synthesis of the phenol **2.125**, which has no methoxy group in the position *ortho* to the phenol group. From this starting material we successfully prepared the triflate **2.126** in 51% followed by formation of the boronic ester in 14% yield (Scheme 81).

Reagents and conditions: a) Tf$_2$O, LiOH (aq), toluene, rt, 3 h, 51%; b) Pd(PPh$_3$)$_4$, Et$_3$N, pinacol borane, THF, Δ, 4 h, 14%
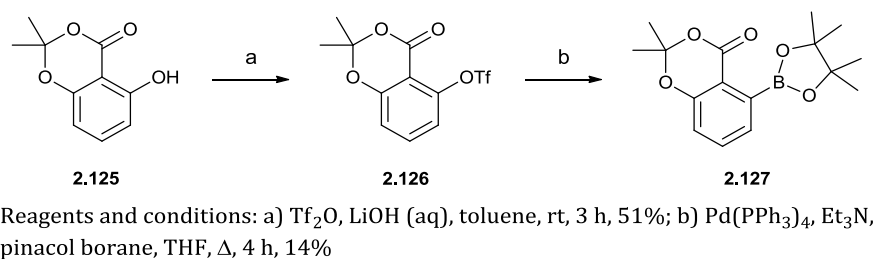
Scheme 81

Boronate ester **2.127** was confirmed to be the desired product by ^{1}H NMR with three aromatic protons at 6.93 ppm, 7.14 ppm and 7.47-7.52 ppm, as well as two singlets at 1.73

84

ppm and 1.42 ppm for the acetonide methyl groups and the pinacol ester methyl groups. The product was found to have the correct mass with a peak at 359 for the [M+Na+MeOH]$^+$ molecular ion in the mass spectrum. The next step was to couple **2.127** with the benzyl protected right hand fragment **2.121** using the conditions reported by Podlech *et al.* (Scheme 82).

| **2.127** | **2.121** | **2.128** | **2.129** |
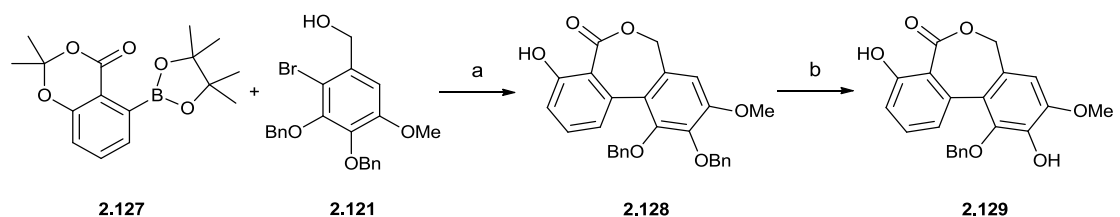
Reagents and conditions: a) Pd(OAc)$_2$, SPhos, Cs$_2$CO$_3$, dioxane/H$_2$O, 80 °C, 4 h, 51%; b) H$_2$, Pd/C, MeOH, rt, 2 min, 48%

Scheme 82

The benzyl protected analogue **2.128** was prepared in 51% yield. The structure was confirmed by ^{1}H NMR with characteristic signals at 9.63 ppm for the phenol, 3.85 ppm for the methoxy CH$_3$ and 6 doublets for the three CH$_2$ groups. The set of doublets for the lactone ring bound CH$_2$ were at 4.73 ppm and 4.94 ppm with a coupling constant of 12.0 Hz, with the other four doublets at 4.55 ppm, 4.77 ppm, 5.00 ppm and 5.12 ppm which all had coupling constants of 11 Hz. Each CH$_2$ corresponds to two distinct doublets in the ^{1}H NMR spectrum. This is because the two protons on each CH$_2$ are diastereotopic and therefore not chemically equivalent (Figure 24).
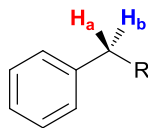
Figure 24 - Diastereotopic protons

If a molecule is achiral the diastereotopic protons usually appear as a singlet in the ^{1}H NMR spectrum. In the case of ulocladol A and its analogues, each compound has two possible conformational isomers due to the planar axis of chirality, which is caused by the two aromatic rings being twisted out of plane to each other and unable to freely rotate. The two diastereoisomers created by this planar axis of chirality are atropisomers. The planar axis of chirality results in the diastereotopic protons of some of the analogues appearing as two

doublets in the ^{1}H NMR spectra. This is because H_a is split into a doublet by H_b and H_b is split into a doublet by H_a.

The X-ray crystal structure of the nitro analogue, **2.115**, has shown the planar axis of chirality between the two aryl rings. It is likely that this occurs in order to accommodate both the hydrogen bond, between the phenol and the carbonyl oxygen, and the seven membered lactone ring. This twist in the lactone ring explains why the methylene group adjacent to the lactone is diastereotopic. Whilst methylene groups on benzyl ethers are not usually diastereotopic, in the case of **2.128** it is likely that the two benzyl ether groups are too bulky to freely rotate resulting in the methylene groups being diastereotopic.

In addition to the signals seen for the three CH_2 groups, there was a singlet at 6.70 ppm for the aromatic proton on the right hand ring as well as another 6 multiplets integrating to 13 protons for the remaining aromatic protons. The ^{13}C NMR and DEPT spectra showed 3 CH_2 signals, 1 CH_3 signal, 10 CH signals and 11 quaternary signals which all confirm the structure of the product. With the benzyl protected analogue in hand we then attempted to deprotect the phenolic groups using a standard hydrogenation protocol. However ^{1}H NMR suggested that only partial deprotection had occurred with the loss of only one of the benzyl protecting groups. The ^{1}H NMR spectrum also suggested that the deprotection had been selective with only one regioisomer present and not a mixture of the two. Using COSY, HMQC and HMBC NMR studies we were able to determine that the correct regioisomer had been deprotected on the phenol *meta* to the carbon-carbon bond **2.129**. The assignment was mainly based on the correlations in the HMBC spectrum (Figure 25).
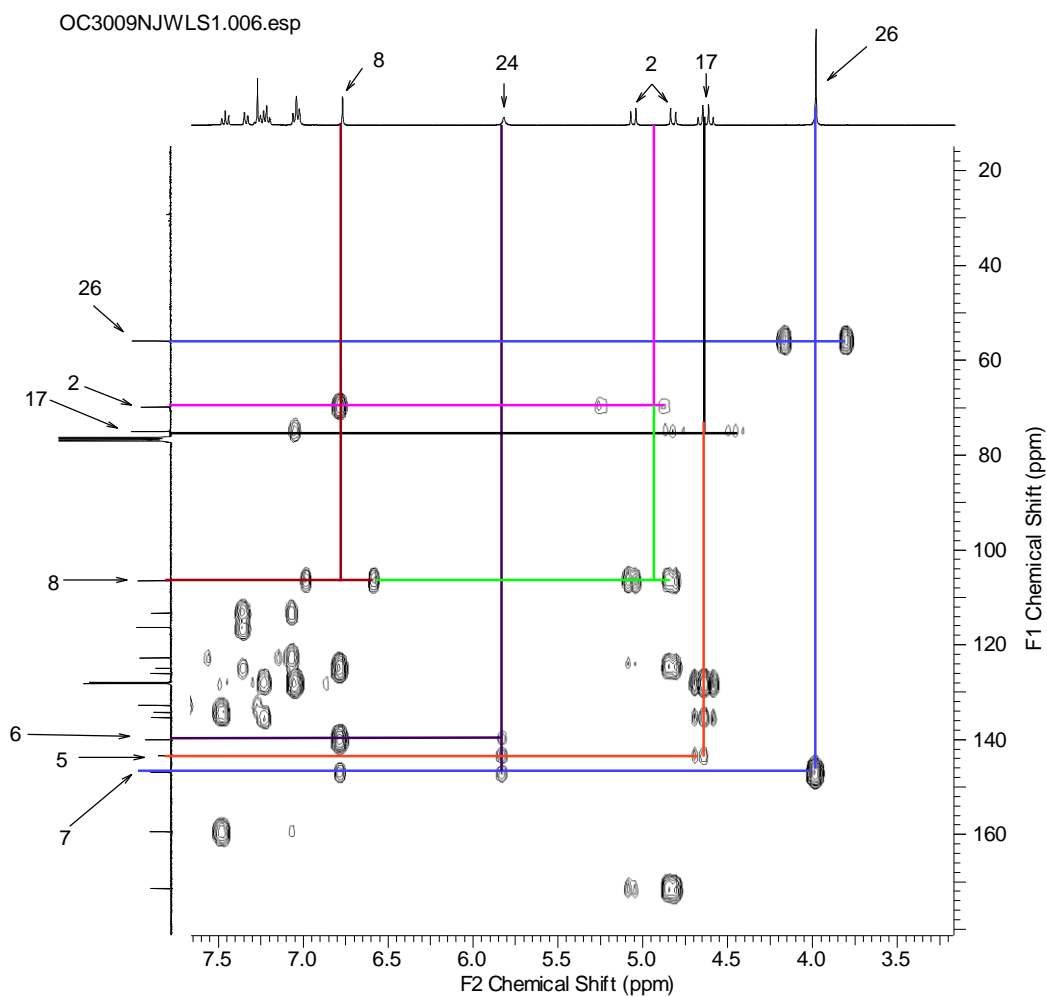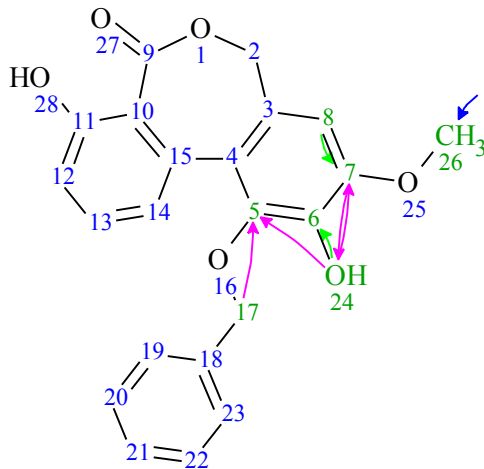
Figure 25 – HMBC spectrum of 2.129

From the spectrum it is possible to assign which phenolic group has been deprotected. If we look at the methoxy group on the right hand ring, from the [superscript]1H NMR spectrum we can

determine that the protons on the CH_3 correspond to the signal at 3.98 ppm. From this signal we can deduce that the corresponding peak in the ^{13}C NMR is at 56.4 ppm. Long range coupling then suggests that C7, the carbon the OMe group is connected to, is at 147.3 ppm. The next step is to determine which other protons C7 couples to long range. If we look along the horizontal blue line, we can see that there is a long range coupling to H24 at 5.82 ppm and H8 at 6.77 ppm. H8 is the aromatic proton next to Ar-OMe, and H24 is the deprotected phenol on C6. From H24 and H8 we can determine that the peaks in the ^{13}C NMR at 106.9 ppm and 140.4 ppm correspond to C6 and C8 respectively. We can also determine which CH_2 is part of the lactone and therefore which CH_2 is part of the benzyl protecting group.

From the HMBC spectrum we can see that H8 experiences long range coupling to a signal in the ^{13}C NMR at 70.3 ppm. This carbon correlates to the CH_2 protons in the ^1H NMR at 5.06 ppm and 4.83 ppm and can only correspond to C2 as the distance between H8 and C17, the benzyl CH_2, is too great to see long range coupling. The selective deprotection of the benzyl group in the middle is also consistent with reactions of 3,4,5-trifunctionalized phenols as the central group suffers from the most steric clashes.

Having determined which regioisomer was present the next step was to fully deprotect the benzyl groups in order to prepare an analogue of ulocladol A without a methoxy group on the left hand ring (Scheme 83).

Reagents and conditions: a) H_2, Pd/C, MeOH, rt, 2 min
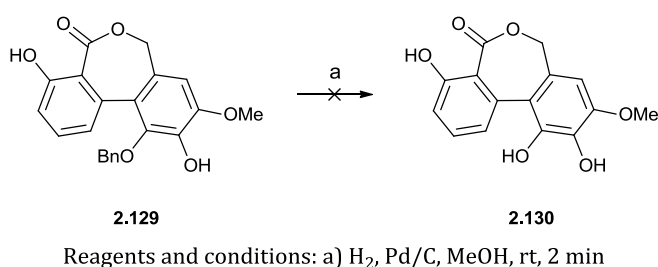
Scheme 83

Unfortunately the reaction was unsuccessful and, in addition to deprotecting the remaining phenolic group, the lactone ring was broken. Due to lack of material and time constraints we were unable to repeat this reaction.

2.3.1 Variable Temperature NMR

The ¹H NMR spectra of the 18 analogues prepared have all had two distinguishing features in common: the methylene group in the seven membered lactone ring and the phenol group on the left hand ring. However, whilst the shape of the signal in the ¹H NMR spectrum for the phenol on each of the analogues has remained relatively consistent, the methylene signal has varied from a broad singlet, seen in **2.118** (Figure 26), to two sharp doublets, seen in **2.82** (Figure 27).
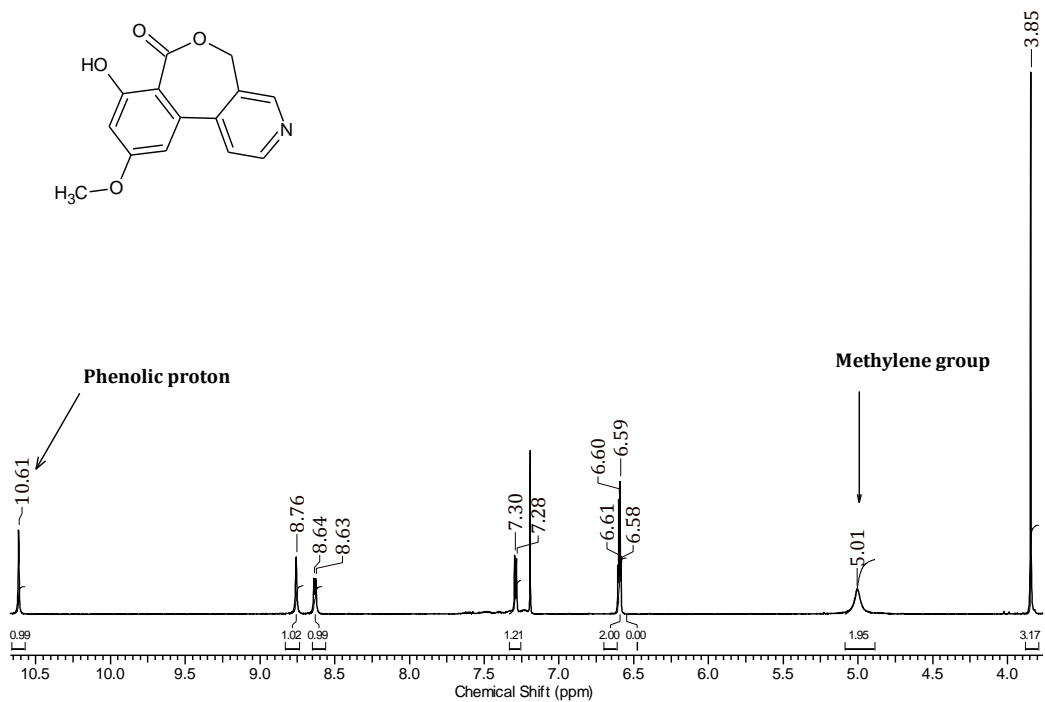
2.118

Figure 26 - ¹H NMR spectrum of 2.118
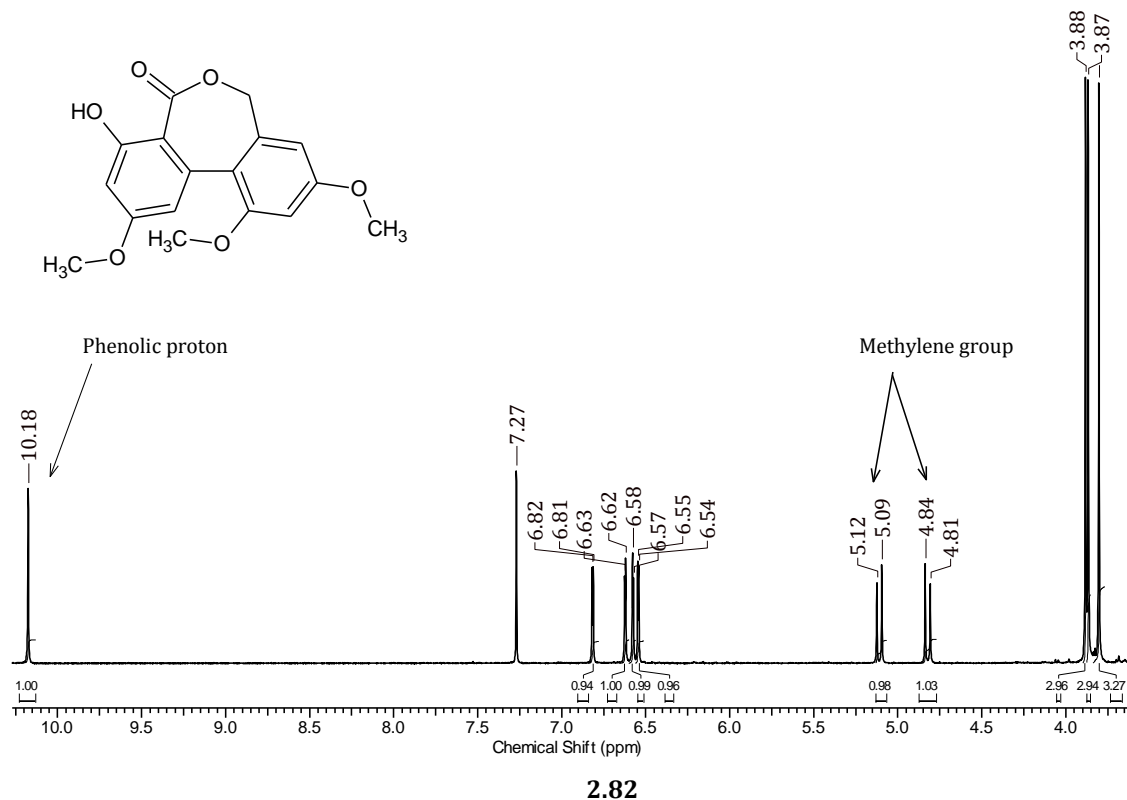
Figure 27 - ¹H NMR spectrum of 2.82

The figures above show the differences between the peak shapes of the methylene group in the two different analogues. It is possible that the distortion of the pyridine ring in **2.118** results in the lactone ring adopting a more planar arrangement, which in turn reduces the energy barrier for movement of the methylene group. Consequently the methylene protons are not diastereotopic and result in a singlet in the ¹H NMR spectrum.

In the case of **2.82** the aryl-aryl bond has three *ortho* substituents. The greater the number of substituents around an aryl-aryl bond, the greater the potential for steric clashes, which forces the aryl rings to adopt a more staggered conformation. This could explain why the methylene protons are diastereotopic and therefore very sharp doublets in the ¹H NMR spectrum.

Use of Variable Temperature NMR would further our understanding of this phenomenon. If the observed signals for the methylene group in the ¹H NMR spectrum coalesce into one signal as the sample is heated, it would indicate that the aryl-aryl bond overcomes the energy barrier of rotation on heating.

Due to constraints in quantity of material, we chose to subject the *meta* substituted fluoro analogue **2.120** to Variable Temperature NMR. This material exhibited two broad singlets in ^{1}H NMR using CDCl$_3$ as the solvent. In order to carry out the Variable Temperature NMR over the desired temperature range a change in solvent to *deutero*-DMSO was required (Figure 28).
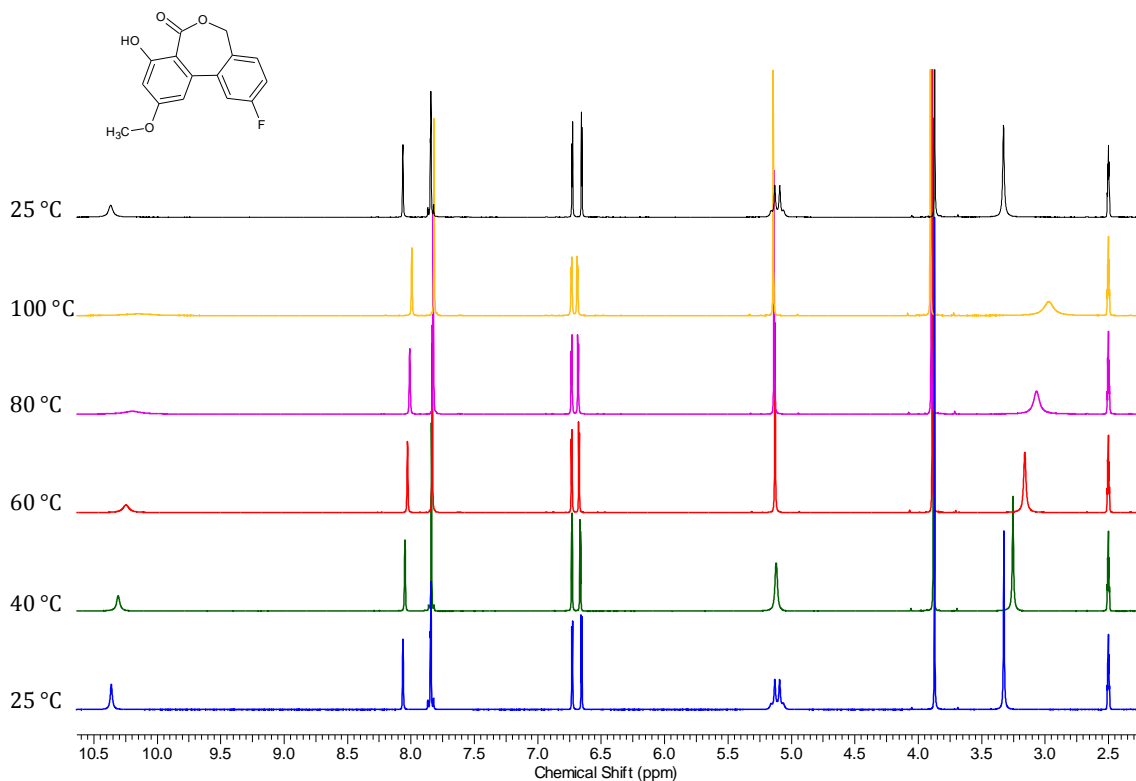
Figure 28 - Variable Temperature NMR of 2.120

The first detail to note is that the change in solvent system resulted in a change in the position of the ^{1}H NMR signals. In addition to this the signal for the methylene group has changed from two broad singlets to two doublets, which appear to be close to coalescence. Upon heating it is clear that full coalescence is achieved by 60 °C; additionally the phenolic signal is seen to broaden whilst shifting upfield. The broadening and upfield shift of the phenolic signal can be explained by hydrogen bonding and exchangeable protons. At lower temperatures the phenolic hydrogen undergoes hydrogen bonding with the carbonyl oxygen; as the temperature increases the hydrogen bond dissociates leading to the broadening of the phenolic signal in the ^{1}H NMR. This phenolic signal undergoes further broadening as the loss of the hydrogen bond results in the phenolic proton becoming exchangeable. These changes are consistent with the proposed explanation for the

diastereotopic methylene protons and indicate a general increase in energy within the molecule. A final ^{1}H NMR was taken at 25 °C to establish whether the sample had decomposed on heating. The spectrum shows that the molecule did not decompose on heating and instead returned to its original conformation.

3   Synthesis of Alternariol and Alternariol 9-Methyl Ether

3.1   Total Synthesis of Alternariol and Alternariol 9-Methyl Ether

Alternariol and alternariol 9-methyl ether have been reported as cell growth inhibitors although the mechanism is unknown.[75] Due to the resemblances in structure of alternariol and alternariol 9-methyl ether to ulocladol A, we thought it would be interesting to test the two natural products against kinases (Figure 29).

Ulocladol A (1.1)   **Alternariol 9-methyl ether (1.2)**   **Alternariol (1.3)**
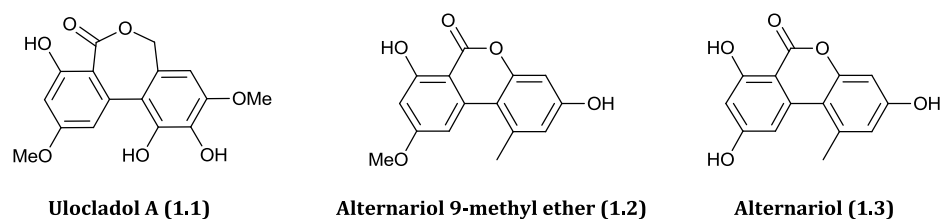
Figure 29 – Ulocladol A, alternariol 9-methyl ether and alternariol

All three natural products have a biaryl system linked by a lactone ring. The left hand rings are substituted in the same positions, with a phenol group *ortho* to the ester of the lactone ring and either a phenol or methoxy group *para* to the ester of the lactone ring. This suggests that the left hand rings for each of the three natural products could come from the same starting material. This starting material could then be coupled to the corresponding fragments for the right hand ring using Suzuki coupling methodology. With this in mind we proposed that we could prepare alternariol (**1.3**) and alternariol 9-methyl ether (**1.2**) using the same boronic ester used to prepare ulocladol A (**1.1**) (Scheme 84).
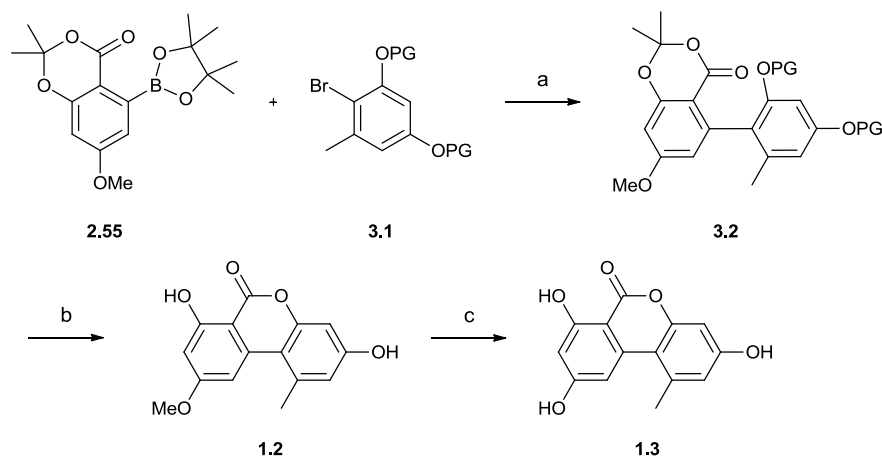
Reagents and conditions: a) Suzuki coupling methodology; b) deprotection and cyclisation of lactone ring; c) BBr$_3$

Scheme 84

We proposed that **2.55**, the pinacol ester used in the synthesis of ulocladol A (**1.1**), could be coupled with the corresponding protected brominated phenol **3.1** to yield the uncyclised compound **3.2**. Deprotection and cyclisation of **3.2** would furnish alternariol 9-methyl ether (**1.2**). The final step would be removal of the methoxy group with BBr$_3$, a procedure reported in the literature by Podlech *et al*[30]. We hoped that this would furnish alternariol 9-methyl ether (**1.2**) and alternariol (**1.3**) in eight and nine steps respectively.

3.2 Synthesis of Alternariol and Alternariol 9-Methyl Ether

With the left hand ring fragment in hand, we began to synthesise the right hand ring fragment (Scheme 84).
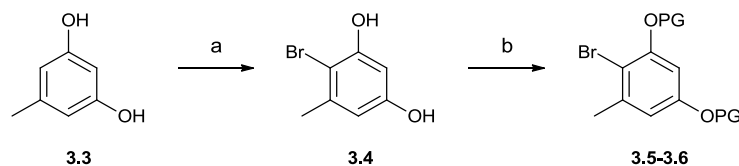
Reagents and condtitions: a) Tetrabutylammonium tribromide, MeOH, DCM, 0 °C→rt, 16 h, 45%; b) See table 1 for details

Scheme 85

We successfully brominated commercially available orcinol (**3.3**) in 45% yield. The bromine was confirmed to be *ortho* to the methyl group through ^{1}H NMR spectrometry. Two distinct doublets could be seen for the aromatic protons which indicated that the aromatic protons were in different environments. If the bromine was located between the

two phenolic groups the resulting symmetry in the molecule would lead to only one signal in the aromatic region.

Due to the sensitive nature of the Suzuki reaction the next step was to protect the hydroxyl groups. We protected the phenolic groups with acetyl and cyanomethyl ether protecting groups (Table 7).

Entry	Protecting Group	Conditions	Product	Yield
1	Acetyl	Ac_2O, DMAP, Et_3N, DCM, rt, 16 h		**3.5** - 84%
2	Cyanomethyl ether	ICH_2CN, K_2CO_3, DMF, Δ, 16 h		**3.6** - 34%

Table 7 – Phenol protecting groups

We attempted to couple **3.5** and **3.6** with the pinacol ester intermediate **2.55** as well as the unprotected diol **3.4** using the conditions reported by Buchwald *et al.* (Scheme 86).[58]

Reagents and conditions: a) $Pd_2(dba)_3$, SPhos, K_2PO_4, THF, Δ, 16 h
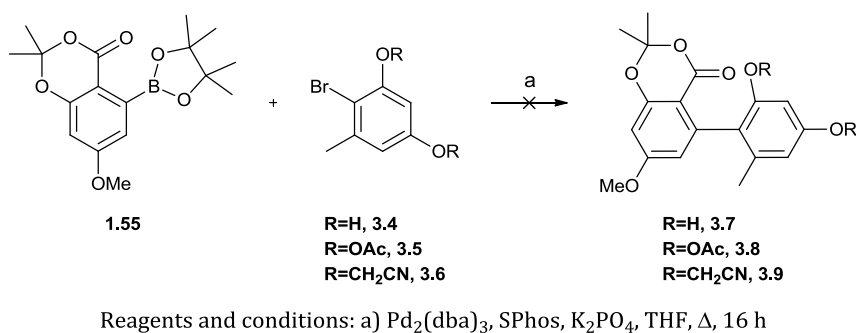
Scheme 86

However, we were unsuccessful each time and no product was isolated from any of the reactions.

We suggested that this was due to the protecting group as found in the case of ulocladol A. From investigation into protecting groups for the right hand fragment of ulocladol A, we determined that acetyl and Boc protecting groups were unstable to Suzuki coupling methodology. Attempts to prepare ulocladol A by coupling the pinacol ester **2.55** with the unprotected catechol had also been unsuccessful, however we were unsure whether this was due to the presence of the free catechol or the aldehyde. We had found during the

synthesis of ulocladol A and its analogues that a free benzyl alcohol could be tolerated in the Suzuki reaction. In addition to this, a more recent set of conditions for the Suzuki reaction, reported by Podlech *et al.*, seemed to be more tolerant of different functional groups.[24] Therefore we decided to try and couple the unprotected diol **3.4** with the pinacol ester intermediate **2.55** using the conditions reported by Podlech *et al.* (Scheme 87).

Reagents and conditions: a) Pd(OAc)$_2$, SPhos, Cs$_2$CO$_3$, dioxane/H$_2$O, Δ, 5 h, 44%
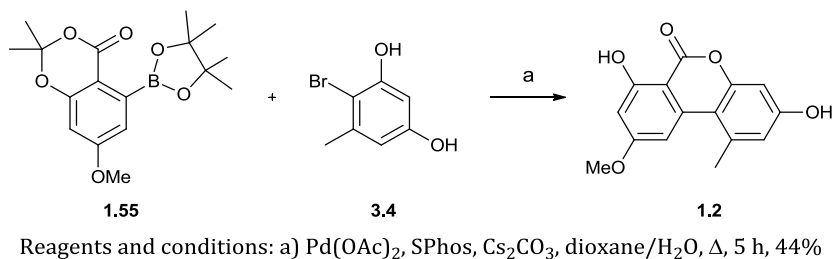
Scheme 87

Recrystallisation of the crude product from methanol afforded alternariol 9-methyl ether (**1.2**) in 44% as a purple crystalline solid. The structure of the product was confirmed by ^{1}H NMR spectroscopy with four signals at 7.21 ppm, 6.72 ppm, 6.64 ppm and 6.61 ppm for the aromatic protons and two signals at 2.73 ppm and 3.91 ppm for the methyl and methoxy groups. Two singlets, one broad at 11.82 ppm and one sharp at 10.36 ppm were observed for the two phenolic groups. An X-ray crystal structure also confirmed the structure of the product (Figure 30).

96
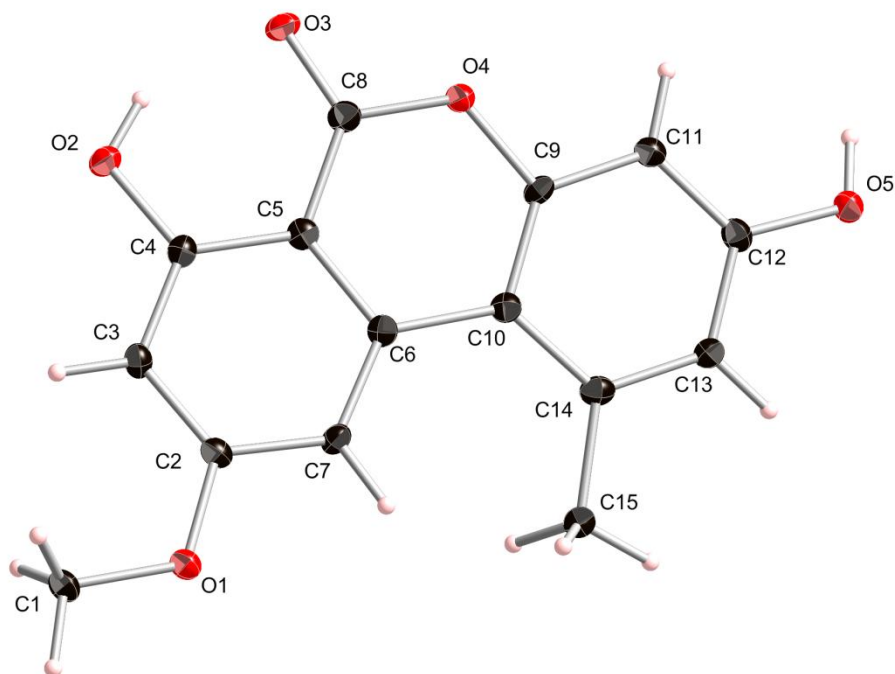
Figure 30 – X-ray crystal structure of alternariol 9-methyl ether

The torsion angle of the lactone ring between C(5)-C(6)-C(10)-C(9) is -6.28°. This is significantly different from the larger torsion angle of 38.55° for the nitro analogue of ulocladol A which was measured through the same atoms. A comparison of the two structures side by side demonstrates how alternariol 9-methyl ether has a more planar conformation than the nitro analogue (Figure 31).[74]
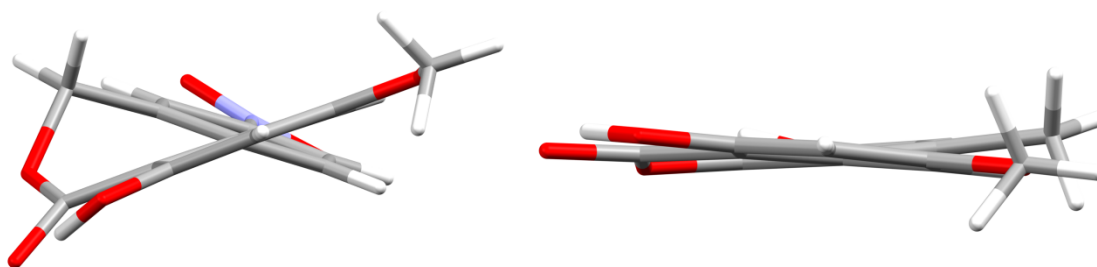
Figure 31 – Nitro analogue 2.115 (L); Alternariol 9-methyl ether (1.2) (R)

The figure shows the nitro analogue **2.115** on the left and alternariol 9-methyl ether (**1.2**) on the right. The twist in the nitro analogue can clearly be seen whilst alternariol 9-methyl ether is almost linear. This suggests that the reason the nitro analogue is twisted is due to the seven-membered lactone ring which is too hindered to sit in the plane.

With alternariol 9-methyl ether (**1.2**) in hand, the final step was to remove the methoxy group in order to furnish alternariol. Following a literature procedure by Podlech *et al.* we successfully removed the methoxy group using boron tribromide (Scheme 88).

Reagents and conditions: a) BBr$_3$, DCM, rt, 16 h, 57%
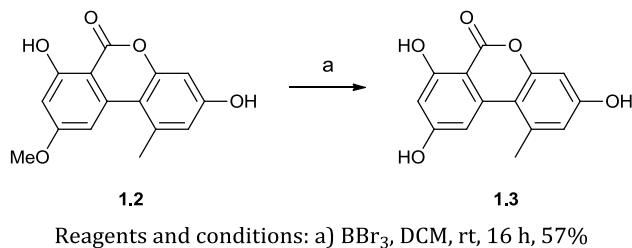
Scheme 88

The reaction was successful and the product was isolated in 57% yield following purification. The structure was confirmed by comparison of ^{1}H NMR collected data with those reported in the literature (Table 8).

Alternariol
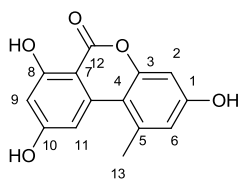
Atom	^{1}H NMR (MeOD)			
	Found	*J* (Hz)	**Reported**[30]	*J* (Hz)
1	-	-	-	-
2	6.58 (d)	2.5	6.62 (d)	2.6
3	-	-	-	-
4	-	-	-	-
5	-	-	-	-
6	6.67 (d)	2.5	6.71 (d)	2.6
7	-	-	-	-
8	-	-	-	-
9	6.35 (d)	2.0	6.38 (d)	1.9
10	-	-	-	-
11	7.22 (d)	2.0	7.27 (d)	2.0

12	-	-	-	-
13	2.73 (s)	-	2.77 (s)	-

Table 8 – Reported and collected ¹H NMR data for alternariol

The product was also confirmed by mass spectrometry with a peak at 313 m/z for the [M+Na+MeOH]$^+$ mass ion.

We successfully prepared the natural products alternariol 9-methyl ether (**1.2**), in 18% overall, and alternariol (**1.3**), in 10% overall, in six and seven steps respectively. These syntheses are the shortest convergent syntheses of alternariol 9-methyl ether (**1.2**) and alternariol (**1.3**) to date.

4 Biological Results

4.1 Biological Results for Ulocladol A and Analogues

4.1.1 Background

The original isolation of ulocladol A reports it to be an inhibitor of the tyrosine kinase lck. We tested ulocladol A and analogues against five different tyrosine kinases. These were Lck, Lyn, Btk, Src and Fyn. All five kinases belong to the same branch of the human kinome (Figure 32).
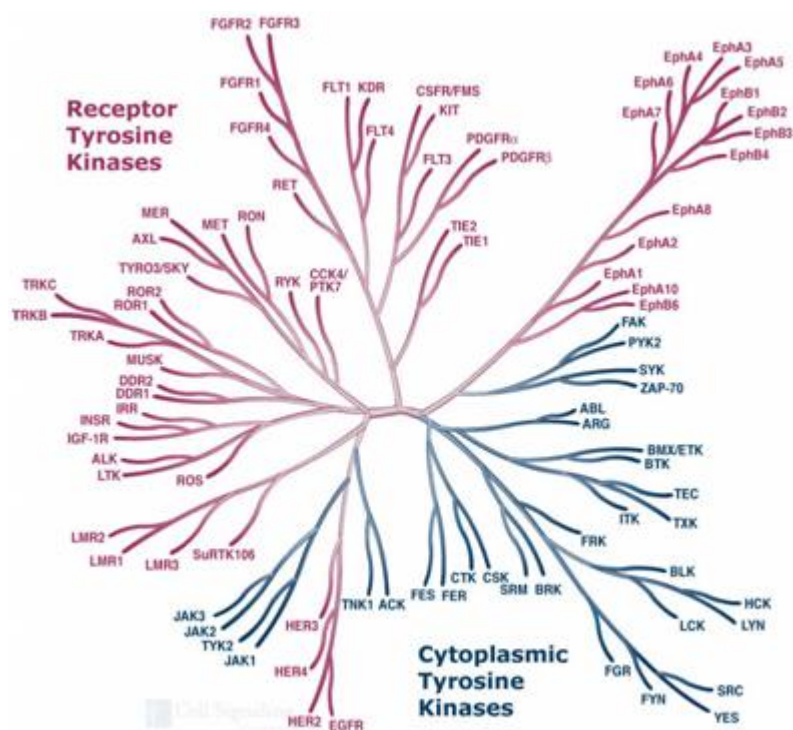
Figure 32 – Tyrosine kinase branch of the human kinome[76]

4.1.1.1 Src Protein Tyrosine Kinase Family

Lck, Lyn, Src and Fyn all belong to the Src family of tyrosine kinases.[77] Src and Fyn are expressed in most tissues, whereas Lck and Lyn are expressed primarily in hematopoietic cells. Lck and Lyn have also both been found in neurons suggesting they may have functions in other cells[78]. The Src family of protein kinases have many different functions within the cells from cell activation to cell growth and cell migration. Deregulation of these kinases has been implicated in several different types of cancer including prostate, colon
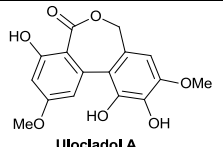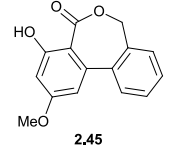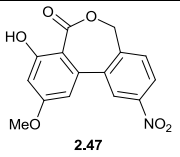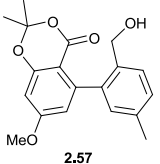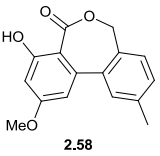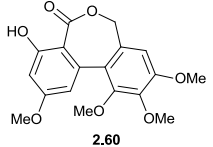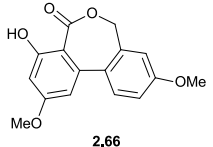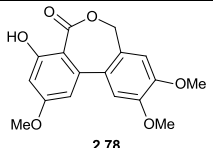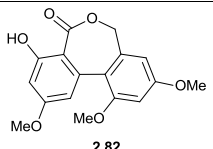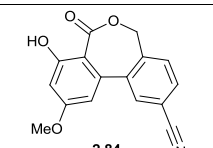
and breast. Btk plays a crucial role in B cell maturation. Mutation of the Btk gene was found to be the cause of the immunodeficiency disease, X-lined agammaglobulinemia.
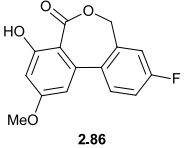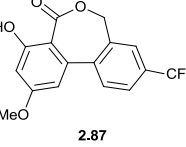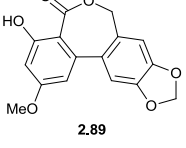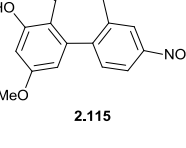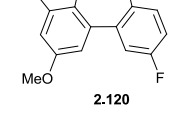
Inhibition of any of these kinases would be beneficial in the treatment of cancer and other diseases.
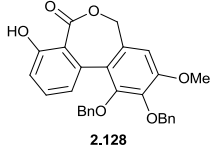
4.1.2 Results

The results of the biological activity tests against 15 of the analogues and the natural product are shown in the tables below. Ulocladol A is shown at the top for ease of comparison between analogues and the natural product (Table 9).

Analogue	MW	cLogP[79]	Kinase Inhibitor[a]														
			Lck			Lyn			Btk			Src			Fyn		
			pIC$_{50}$	IC$_{50}$	%Inh	pIC$_{50}$	IC$_{50}$	%Inh	pIC$_{50}$	IC$_{50}$	%Inh	pIC$_{50}$	IC$_{50}$	%Inh	pIC$_{50}$	IC$_{50}$	%Inh
Ulocladol A	318	1.3	5.6	2.51 E^{-6}	93			74	5.85	1.41 E^{-6}	95			36			38
2.45	256	2.6			<50			<50			<50			<50			<50
2.47	301	2.4	6.91	1.23 E^{-7}	97	6.84	1.45 E^{-7}	100	5.92	1.20 E^{-6}	88	6.22	6.03 E^{-7}	97	5.87	1.35 E^{-6}	95
2.57	328	4			26			22			6			8			3
2.58	270	3.1			63			78			18			27			8

Analogue	MW	cLogP	Kinase Inhibitor[a]														
			Lck			Lyn			Btk			Src			Fyn		
			pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh
2.60	346	1.9	6.19	6.46 E^{-7}	97	6.54	2.88 E^{-7}	98	5.84	1.45 E^{-6}	88	6.04	9.12 E^{-7}	95	5.72	1.91 E^{-6}	93
2.66	286	2.5			81	5.54	2.86 E^{-6}	89			55	5.28	5.25 E^{-6}	66			33
2.78	316	2.3			26			4			4			0			5
2.82	316	2.6	5.59	2.57 E^{-6}	81			13			-2			8			0
2.84	281	2.1			3			13			7			-7			4

| Analogue | MW | cLogP | Kinase Inhibitor[a] | | | | | | | | | | | | | | |
| | | | Lck | | | Lyn | | | Btk | | | Src | | | Fyn | | |
			pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh
2.86	274	2.8			30			36			2			19			8
2.87	324	3.5			38			18			6			11			4
2.89	300	2.6			20			12			9			-6			11
2.115	301	2.4			46			19			39			10			13
2.120	274	2.8			35			14			54			11			17

Analogue	MW	cLogP	Kinase Inhibitor[a]														
			Lck			Lyn			Btk			Src			Fyn		
			pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh	pIC_{50}	IC_{50}	%Inh
2.128	468	5.4			52			14			18			5			6

a) % inhibition refers to the degree of inhibition observed when compounds were tested at 10^{-5}M; Dose-response curves were performed to calculate pIC50 and IC50.

Table 9 – Inhibitory activity of analogues against Lck, Lyn, Btk, Src and Fyn

The biological data is summarised in the graph below (Figure 33).
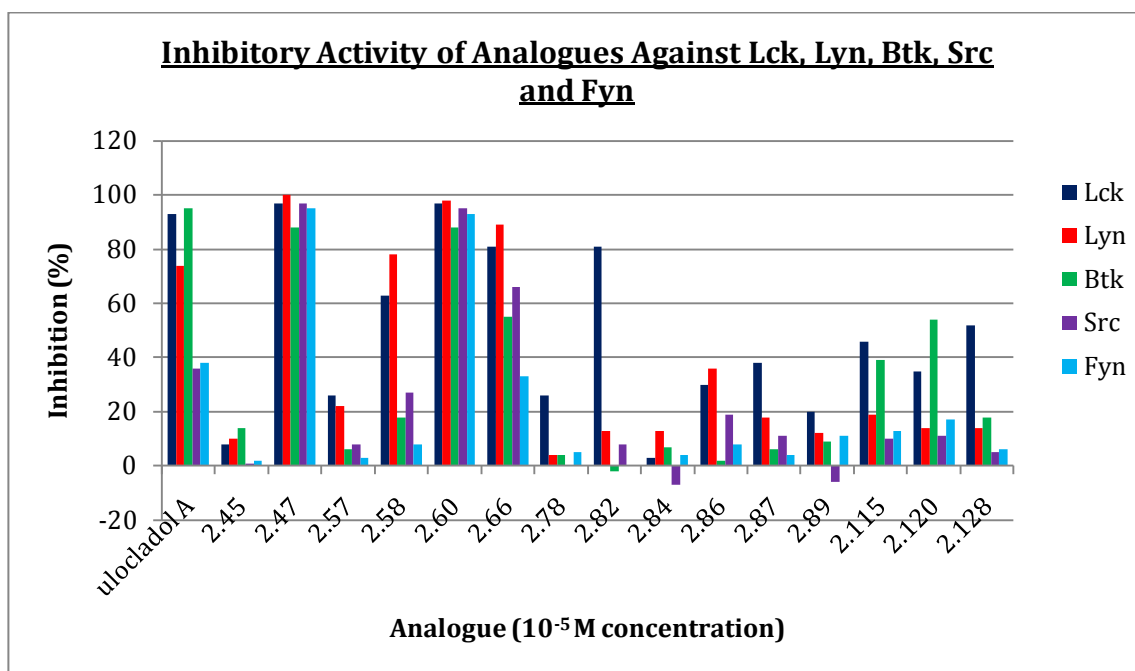
Figure 33 – Inhibitory activity of analogues

The biological activity data show some interesting results. The data suggests that both the position and type of functional group on the right hand ring is important in relation to activity. The majority of the analogues exhibit low enzyme inhibitory activity with 10 out of 15 analogues inhibiting all five tyrosine kinases in less than 60% at a concentration of 10^{-5} M. The most promising analogues are **2.47** with a nitro group *meta* to the aryl-aryl bond, **2.60** with three methoxy groups *ortho*, *meta* and *para* to the carbon-carbon bond and **2.66** with a methoxy group *para* to the carbon-carbon bond. These three analogues all inhibit Lck enzyme activity above 80%, as well as showing moderate to good inhibition of the other four tyrosine kinases. One other notable analogue is **2.82** with methoxy groups *ortho* and *para* to the carbon-carbon bond. This analogue selectively inhibits 81% of Lck enzyme activity but is inactive against the other four tyrosine kinase inhibitors.

Whilst we are unable to definitively elucidate SAR activity from the biological data collected, due to the incomplete set of analogues prepared, we have split the results into groups in the hope that some comparisons can be made.

The first group are the analogues which are solely substituted in the *meta* position to the aryl-aryl bond (Figure 34).
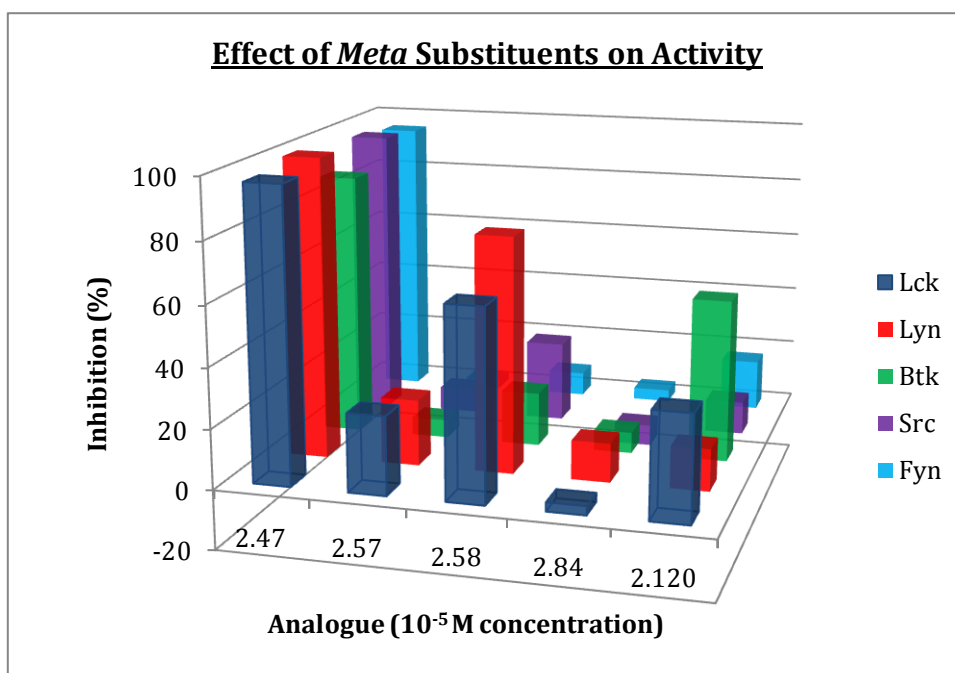
Figure 34 – Inhibitory activity of *meta* substituted analogues

From the graph we can see that the nitro substituted analogue **2.47** is the only *meta* substituted analogue which shows good inhibitory activity against all five kinases, although we cannot be certain that this is due to the effect of a single component or a synergistic effect resulting from the mixture of compounds.

The ring closed, methyl substituted analogue **2.58** shows average inhibitory activity against Lck and Lyn but poor activity against Btk, Src and Fyn. The ring opened methyl substituted analogue **2.57**, the nitrile substituted analogue **2.84** and the fluoro substituted analogue **2.120** all show poor inhibitory activity against all five kinases. This suggests that a group which can form hydrogen bonds is necessary in the *meta* position of the right hand ring.

The second group for comparison are the *para* substituted analogues (Figure 35).

108
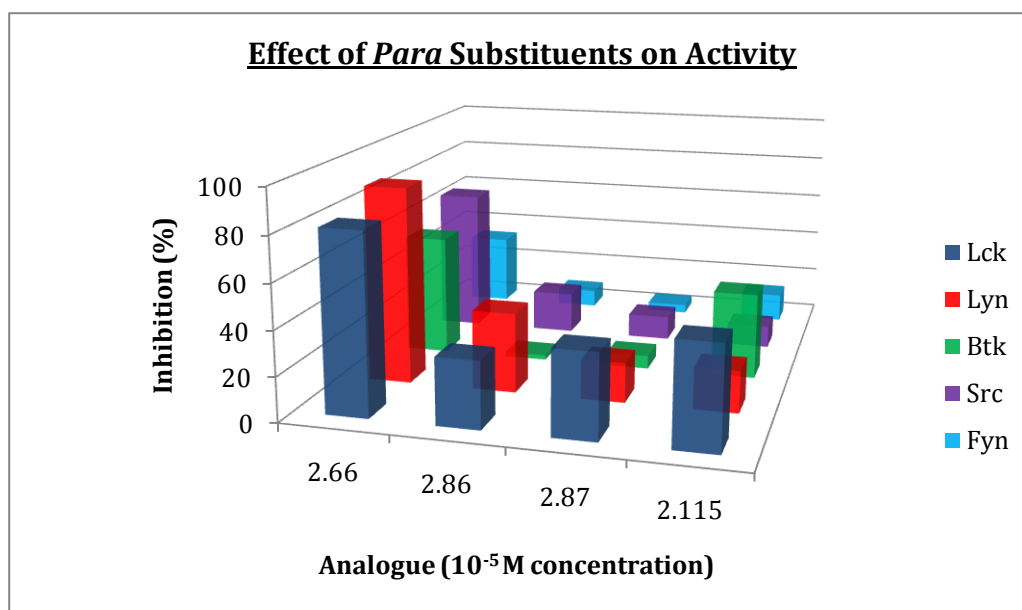
Figure 35 – Effect of *para* substituents on activity

In general, the substituents in the *para* position on the right hand ring have poor inhibitory activity, with the methoxy substituted analogue **2.66** being the only one to inhibit Lck above 50%. Surprisingly the nitro analogue **2.115** does not inhibit any of the kinases above 50% unlike its regioisomer **2.47**, which inhibits all the kinases above 85%.

The graph below looks at two pairs of regioisomers which are substituted in the *meta* and *para* positions with either a fluoro or nitro group (Figure 36).
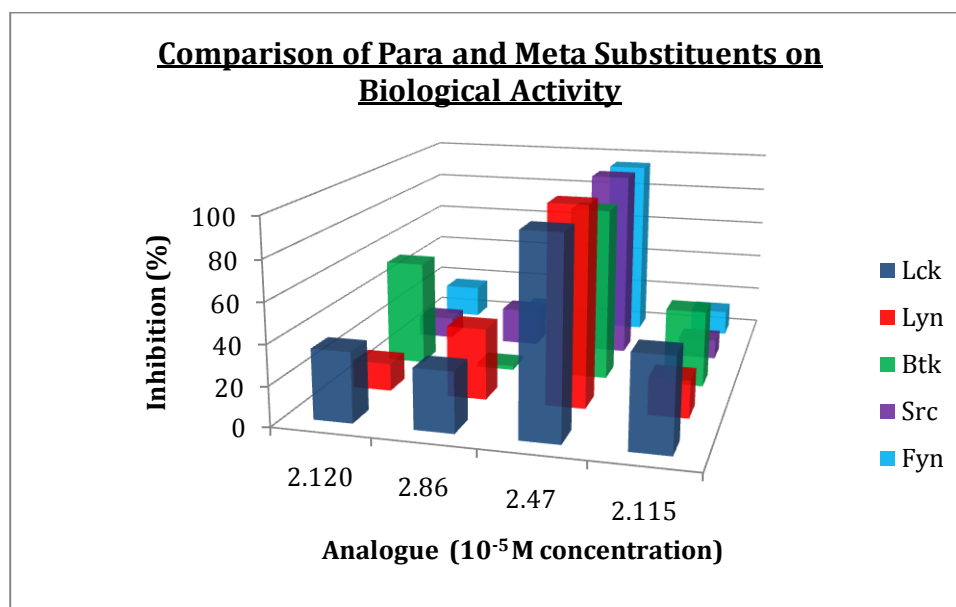
Figure 36 – Comparison of *para* and *meta* substituents effect on activity

The graph shows that the *meta* substituted nitro analogue **2.47** is the most active of the four analogues shown. It exhibits greater than 95% inhibition of the Lck, Lyn, Src and Fyn tyrosine kinases as well as inhibiting the Btk tyrosine kinase at 88%. If we look at the *meta* substituted fluoro analogue **2.120** we can see that the activity is much lower and, in general, exhibits less than 50% inhibition of all the kinases.

The *para* substituted groups all exhibit enzyme inhibitory activity of less than 50%. This includes the *para* substituted nitro analogue. Despite the *meta* substituted analogue being the most active analogue, the *para* substituted analogue does not exhibit the same inhibitory activity, implying that the *meta* position is important in the biological activity. The *para* substituted fluoro analogue is also inactive against the five tyrosine kinases with inhibition of less than 40%.

The next graph shows the biological activity of the methoxy substituted analogues (Figure 37).
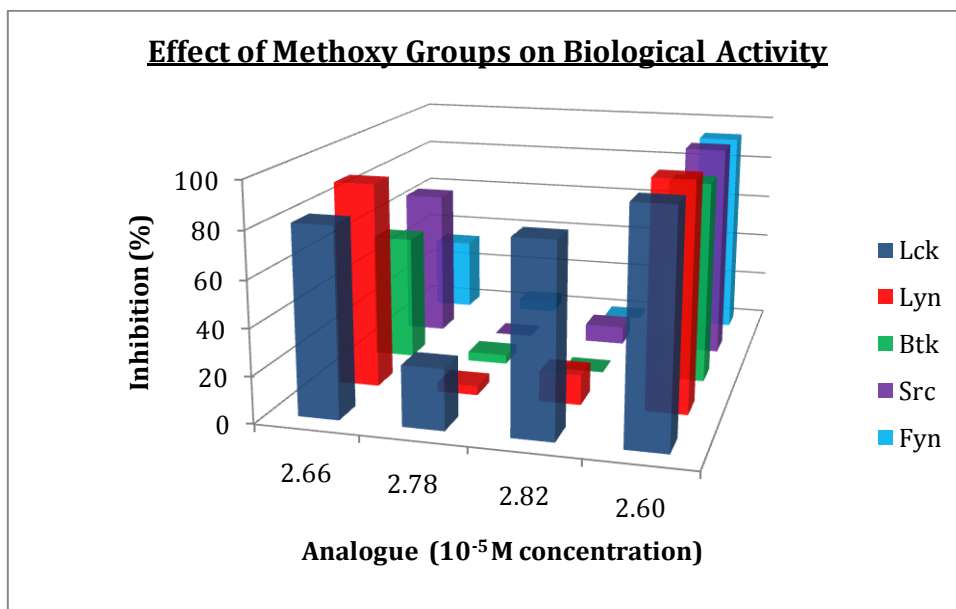
Figure 37 – Effect of methoxy groups on activity

From the graph we can see that **2.60**, the trisubstituted *ortho*, *meta* and *para* methoxy analogue is the most active compound. The *para* substituted analogue **2.66** shows good inhibitory activity against Lck and Lyn but is less effective against Btk, Src and Fyn. The *meta* and *para* disubstituted compound **2.78** is inactive against all the tyrosine kinases, whereas the *ortho* and *para* disubstituted compound **2.82** shows activity against Lck but is inactive against the other four kinases.

The model structure of ulocladol A bound to the Lck active site showed the right hand ring of ulocladol A pointing towards the back of the enzyme active site. This suggested that the phenolic groups on the right hand ring were interacting with hydrogen bond acceptor sites. The trimethoxy substituted analogue **2.60** has no phenolic groups on the right hand ring with which it can form hydrogen bonds to an acceptor; however, the trimethoxy groups can act as hydrogen bond acceptors, implying that there are hydrogen bond donor sites within the pocket.

Unfortunately we are unable to determine whether changing the groups on the left hand ring would have a large effect on biological activity. We can tell from the unsubstituted analogue **2.45** that the groups on the right hand ring play a large role in activity with the highest inhibition of all the kinases being 14%. We were unsuccessful in our attempts to deprotect the benzyl protected analogue **2.128** to yield an analogue which would tell us what effect, if any, the methoxy group on the left hand ring had on the activity.

4.2   Biological Results for Alternariol 9-Methyl Ether and Alternariol

4.2.1   Background

Alternariol 9-methyl ether and alternariol were both tested for activity against the same five tyrosine kinases used for ulocladol A and analogues.

4.2.2   Results

The results for the biological activity of alternariol 9-methyl ether and alternariol are shown below (Table 10).
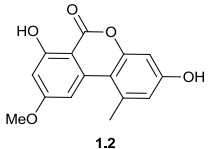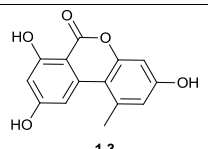
Compound	MW	cLogP	Tyrosine Kinase (% Inhibition at 10^{-5} M)				
			Lck	Lyn	Btk	Src	Fyn
1.2	272	2.5	24	15	17	8	12
1.3	258	1.9	85	58	32	34	44

Table 10 – Inhibitory activity of alternariol 9-methyl ether and alternariol

The data show that alternariol 9-methyl ether (**1.2**) exhibits minimal inhibitory activity against all of the tyrosine kinases, with the greatest inhibition being against Lck with an

inhibition of 24%. Alternariol (**1.3**) shows better activity, with an inhibition of Lck of 85% at 10^{-5} M concentrations.

The X-ray crystal structure of alternariol 9-methyl ether (**1.2**) suggested that the molecule adopts a more planar conformation than the ulocladol analogues (Figure 38).
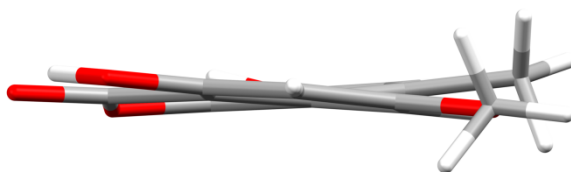
Figure 38 - Side on view of alternariol 9-methyl ether (1.2) looking from the left hand ring back towards the right hand ring

Due to the change in conformation it is difficult to predict whether alternariol (**1.3**) and alternariol 9-methyl ether (**1.2**) bind to a tyrosine kinase active site in the same way as ulocladol A and its analogues. From the biological data collected for these two natural products, we can only deduce that when the methoxy group on the left hand ring of alternariol 9-methyl ether (**1.2**) is removed to furnish alternariol (**1.3**) there is an increase in the inhibitory activity against the five tyrosine kinases.

In order to elucidate full SAR activity for ulocladol A (**1.1**), alternariol 9-methyl ether (**1.2**) and alternariol (**1.3**) a much larger range of analogues for all three would be required.

5   Conclusion

5.1   Ulocladol A and Analogues

We have successfully prepared ulocladol A in 0.4% over ten steps in a convergent synthesis. The route is applicable to the synthesis of analogues of ulocladol A and has been used to prepare eighteen different analogues with a variety of functional groups on the right hand ring. Fifteen of these analogues have been tested for biological activity against five tyrosine kinases.

Two analogues, the *meta* substituted nitro analogue **2.47** and the trimethoxy substituted analogue **2.60**, have been found to exhibit greater inhibition of Lck than ulocladol A, and show very good inhibitory activity against the other four kinases.

Due to time constraints and lack of material, we were unable to investigate the effect the substituents on the left hand ring have on biological activity beyond the lack of activity observed when there are no substituents on the right hand ring.

5.2   Alternariol 9-Methyl Ether and Alternariol

We have successfully prepared alternariol 9-methyl ether and alternariol in a convergent route which did not require protection of the phenolic groups on the right hand ring. Alternariol 9-methyl ether was prepared in five steps in 18% overall and alternariol was prepared in six steps in 10% overall. This is the shortest convergent synthesis of alternariol 9-methyl ether and alternariol to date. Alternariol 9-methyl ether and alternariol were both tested for biological activity against the five tyrosine kinases, with alternariol found to selectively inhibit Lck at 85%.

5.3   Chemistry

Over the course of the project we have used several different catalyst systems on different substrates. This has mainly been due to the difficulties found in coupling substrates which have differing steric and electronic effects on the Suzuki reaction. We also found that switching the position of the boronate from the right hand ring to the left hand ring was more practical due to the increased number and variety of substrates available to couple (Figure 39).
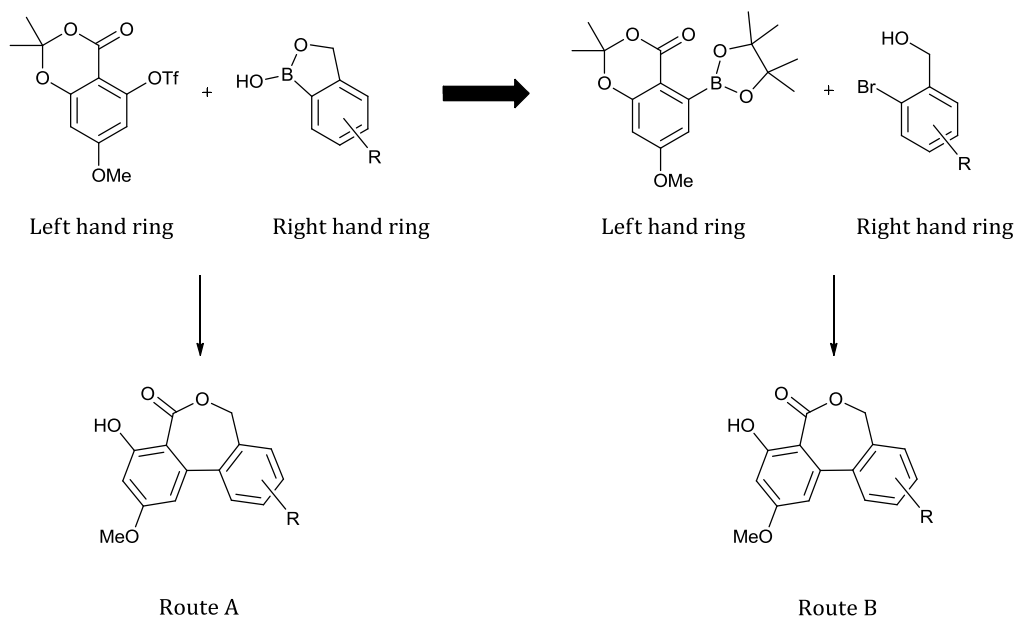
Figure 39 - Switching the boronate group from the left hand ring (A) to the right (B) was more practical

The initial catalyst system, a thermal reaction using PdCl$_2$(dppf), proved to be low yielding and often resulted in a mixture of cyclised and uncyclised product which required either further reaction or extensive purification. A change to the conditions reported by Buchwald *et al.* allowed us to prepare a further six analogues of ulocladol A, however the problems with low yields and extensive purification were not solved.[46]

We then began testing different catalyst systems under both thermal and microwave conditions. This resulted in the use of Pd(PPh$_3$)$_4$ and 2M NaHCO$_3$ in dioxane to prepare a further four analogues and, although the yields did not improve by a great amount, the final compounds required less extensive purification.

The final catalyst system, reported by Atemöller *et al.* in their synthesis of ulocladol A, was successfully used to prepare two analogues of ulocladol A, one of which was the precursor to ulocladol A, and ulocladol A itself.[24] In addition to this we also applied this method to the successful synthesis of alternariol 9-methyl ether, which then enabled us to prepare alternariol. We found that the method reported by Atemöller *et al.* resulted in higher yielding reactions with the resulting compounds being easier to purify.

6 Future Work

Due to time limits and availability of starting materials we have been unable to establish a complete picture of SAR relating to ulocladol A. If it were possible to undertake further work in this area it would be interesting to build a complete picture of SAR and determine which of the functional groups on ulocladol A were involved in the inhibition of the Lck enzymes active site. If possible it would also be beneficial to obtain protein X-ray crystal structures of the analogues bound to the Lck enzyme active site to determine the true bonding motif.

In addition to this it would be interesting to resynthesise the nitro analogue **2.47** ensuring that only one compound was present. The compound could then be retested against the tyrosine kinases to determine whether the results of the first biological assay were due to the nitro analogue **2.47**, or whether a synergistic effect, caused by the mix of two compounds, occurred.

Furthermore, given the apparent role of the lactone ester in binding to the enzyme active site, it would be of interest to examine the effect on inhibitory action if the lactone ester was replaced with either a thioester or an amide.

Whilst only limited work was carried out in the area of alternariol 9-methyl ether and alternariol, it would be interesting to determine how the two molecules bind to the enzyme active site and if the more planar conformation of the molecules, when compared to ulocladol A, has any effect on inhibitory activity. It would also be of interest to exploit the synthetic methodology discovered to produce additional analogues of alternariol with the aim of formulating a SAR for this molecular core.

7   Experimental

7.1   General Experimental

All reagents were used directly as obtained from commercial suppliers unless otherwise stated. When dry solvents were required, THF was freshly distilled from sodium benzophenone ketyl under argon; CH_2Cl_2 was freshly distilled from calcium hydride under argon and toluene was freshly distilled from sodium metal under argon.

Reactions were monitored by thin layer chromatography using pre-coated aluminium backed sheets of silica and visualised with UV light and a cerium molybdate stain. Column chromatography was carried out using MN Kieselgel 60, 0.04-0.063 mm 230-400 mesh ASTM silica gel.

Melting points were determined using an Electrothermal melting point apparatus and are uncorrected.

Infrared spectra were recorded using a Thermo Nicolet 380 FT-IR spectrometer with a Smart Orbit Golden Gate attachment and processed using the Omnic software suite. Absorptions are reported in wavenumbers (cm^{-1}).

NMR spectra were recorded on either a Bruker AV300 spectrometer or a Bruker DPX400 spectrometer operating at 300 or 400 MHz respectively for ^1H experiments, and 75 MHz or 100 MHz respectively for ^{13}C and Dept-135 experiments. Proton and carbon NMR were taken in $CDCl_3$, unless stated otherwise, using tetramethylsilane as an internal standard. Spectral data was processed using ACD Labs software. Chemical shift, δ, is reported in parts per million with multiplicities described using the abbreviations s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; and br, broad.

Electrospray mass spectra were recorded using a Thermoquest Trace MS and Micromass Platform II single quadrapole mass spectrometer.

High resolution mass spectra were collected by the School of Chemistry Mass Spectrometry Service using a Bruker Apex III FT-ICR mass spectrometer fitted with an Apollo electrospray ionisation source.

7.2 Kinase Inhibition Assays

Inhibition of kinase activity was measured using the Immobilized Metal Assay for Phosphochemicals (IMAP) assay. IMAP is a homogeneous fluorescence polarization (FP) assay based on affinity capture of phosphorylated peptide substrates. IMAP uses fluorescein-labeled peptide substrates that, upon phosphorylation by a protein kinase, bind to so-called IMAP nanoparticles, which are derivatized with trivalent metal complexes. Binding causes a change in the rate of the molecular motion of the peptide, and results in an increase in the FP value observed for the fluorescein label attached to the substrate peptide.

Ten point serial dilutions using a $\sqrt{10}$ dilution factor were used for dose response testing of compounds. Starting concentration was 10^{-6} M for Lck and 10^{-5} M for Src, FynT, Btk and Lyn IMAP assays. Dose-response curves were run as two experiments (N=1) on duplicate plates (n=2). All data was normalized to percentage effect based on maximum (Max) and minimum (Min) control values. On every 384 assay plate 16 wells were used as minimum wells (wells with ATP, 0% effect) and 16 wells were used as maximum wells (wells without ATP, 100% effect). 16 wells were used for measuring the background signal, obtained from a kinase reaction containing all constituents except the labelled peptide substrate. Percentage effect was plotted against log dilution concentration of compound to obtain sigmoid dose response curves. pIC_{50} values were calculated using ActivityBase.

All biochemical assays were run at $K_{M,ATP}$ of the enzyme using non-saturated conditions, meaning that during the incubation time it was assured that the signal increase was linear with time. For all biochemical assays a reference standard was used on each plate. Newly purchased enzyme batches were tested in serial dilutions with the reference standard to assure that comparable compound pIC_{50}s were obtained in all assays run over time using different enzyme batches.

7.2.1 Lck IMAP Assay

Enzyme used was N-terminal His6-tagged recombinant full-length human Lck from Millipore. Phosphorylation substrate was a fluorescein-labeled peptide (5FAM-KVEKIGEGTYGVV-NH$_2$) derived from p34cdc2 from Molecular Devices. Enzymes, substrate and ATP were diluted in Kinase Reaction buffer (10 mM Tris-HCl, 10 mM MgCl$_2$, 0.01%

Tween-20, 0.05% NaN$_3$ pH 7.2, 1 mM DTT). The final volume at the kinase reaction step of the assay in the 384-well plate was 20 μL. Final amount of enzyme in the reaction was 0.1 U/mL. Enzyme was pre-incubated with compounds diluted in 1% DMSO for 60 minutes at room temperature in the dark. Subsequently, peptide substrate to a final concentration of 100 nM, and ATP to a final concentration of 6 μM were added and the mix was incubated for 120 minutes at room temperature in the dark. IMAP progressive binding buffer (75% 1x buffer A, 25% 1x buffer B with 1:600 Progressive Binding Reagent; Molecular Devices) was added, followed by an incubation step of 60 minutes at room temperature in the dark. Finally, the FP signal was read on an Envision Multilabel reader (Perkin Elmer).

7.2.2   Src IMAP Assay

Enzyme used was N-terminal His6-tagged recombinant full-length human Src from Millipore. Peptide substrate was fluorescein labelled p34cdc2 peptide substrate (5FAM-KVEKIGEGTYGVV-NH$_2$) from Molecular Devices. Enzymes, substrate and ATP were diluted in Kinase Reaction buffer (10 mM Tris-HCl, 10 mM MgCl$_2$, 0.01% Tween-20, 0.05% NaN$_3$ pH 7.2, 1 mM DTT). The final volume at the kinase reaction step of the assay in the 384-well plate was 20 μL. Final amount of enzyme in the reaction was 0.2 U/mL. Enzyme was pre-incubated with compounds diluted in 1% DMSO for 60 minutes at room temperature in the dark. Subsequently, peptide substrate to a final concentration of 100 nM, and ATP to a final concentration of 4 μM were added and the mix was incubated for 120 minutes at room temperature in the dark. IMAP progressive binding buffer (75% 1x buffer A, 25% 1x buffer B with 1:600 Progressive Binding Reagent; Molecular Devices) was added, followed by an incubation step of 60 minutes at room temperature in the dark. Finally, the FP signal was read on an Envision Multilabel reader (Perkin Elmer).

7.2.3   FynT IMAP Assay

Recombinant full-length human FynT was expressed and purified at Schering-Plough. Thrombin cleavage was used to remove a His6-tag that was used for purification. The final preparation of the enzyme was over 90% pure. Peptide substrate was fluorescein labelled p34cdc2 peptide substrate (5FAM-KVEKIGEGTYGVV-NH$_2$) from Molecular Devices. Enzymes, substrate and ATP were diluted in Kinase Reaction buffer (10 mM Tris-HCl, 10

mM MgCl$_2$, 0.01% Tween-20, 0.05% NaN$_3$ pH 7.2, 1 mM DTT). The final volume at the kinase reaction step of the assay in the 384-well plate was 20 μL. Final concentration of the enzyme in the reaction was 125 ng/mL. Enzyme was pre-incubated with compounds diluted in 1% DMSO for 60 minutes at room temperature in the dark. Subsequently, peptide substrate to a final concentration of 100 nM, and ATP to a final concentration of 0.2 μM were added and the mix was incubated for 120 minutes at room temperature in the dark. IMAP progressive binding buffer (75% 1x buffer A, 25% 1x buffer B with 1:600 Progressive Binding Reagent; Molecular Devices) was added, followed by an incubation step of 60 minutes at room temperature in the dark. Finally, the FP signal was read on an Envision Multilabel reader (Perkin Elmer).

7.2.4 Btk IMAP Assay

Enzyme used was N-terminal His6-tagged recombinant full-length human Btk from Millipore. Peptide substrate was fluorescein labelled Blk/Lyntide peptide substrate (5FAM-EFPIYDFLPAKKK-NH$_2$) from Molecular Devices. Enzymes, substrate and ATP were diluted in Kinase Reaction buffer (10 mM Tris-HCl, 10 mM MgCl$_2$, 0.01% Tween-20, 0.05% NaN$_3$ pH 7.2, 1 mM DTT). The final volume at the kinase reaction step of the assay in the 384-well plate was 20 μL. Final amount of enzyme in the reaction was 0.1 U/mL. Enzyme was pre-incubated with compounds diluted in 1% DMSO for 60 minutes at room temperature in the dark. Subsequently, peptide substrate to a final concentration of 50 nM, and ATP to a final concentration of 5 μM were added and the mix was incubated for 120 minutes at room temperature in the dark. IMAP progressive binding buffer (75% 1x buffer A, 25% 1x buffer B with 1:600 Progressive Binding Reagent; Molecular Devices) was added, followed by an incubation step of 60 minutes at room temperature in the dark. Finally, the FP signal was read on an Envision Multilabel reader (Perkin Elmer).

7.2.5 Lyn IMAP Assay

Enzyme used was N-terminal His6-tagged recombinant full-length human Lyn from Millipore. Peptide substrate was fluorescein labelled Blk/Lyntide peptide substrate (5FAM-EFPIYDFLPAKKK-NH$_2$) from Molecular Devices. Enzymes, substrate and ATP were diluted in Kinase Reaction buffer (10 mM Tris-HCl, 10 mM MgCl$_2$, 0.01% Tween-20, 0.05%

NaN$_3$ pH 7.2, 1 mM DTT). The final volume at the kinase reaction step of the assay in the 384-well plate was 20 µL. Final amount of enzyme in the reaction was 62.5 mU/mL. Enzyme was pre-incubated with compounds diluted in 1% DMSO for 60 minutes at room temperature in the dark. Subsequently, peptide substrate to a final concentration of 100 nM, and ATP to a final concentration of 2 µM were added and the mix was incubated for 120 minutes at room temperature in the dark. IMAP progressive binding buffer (75% 1x buffer A, 25% 1x buffer B with 1:600 Progressive Binding Reagent; Molecular Devices) was added, followed by an incubation step of 60 minutes at room temperature in the dark. Finally, the FP signal was read on an Envision Multilabel reader (Perkin Elmer).

7.3   Experimental

3,4,5-Trimethoxybenzyl benzoate (2.10).
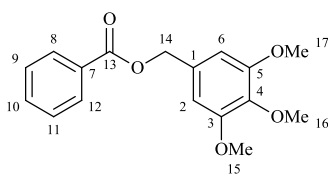
To a solution of 3,4,5-trimethoxybenzyl alcohol (0.5 mL, 0.003 mol) and benzoyl chloride (0.843 g, 0.006 mol) in acetonitrile (15 mL) under argon was added triethylamine (0.4 mL, 0.003 mol) dropwise. The reaction mixture was stirred at RT for 3 h and then poured into ice cold water (15 mL). The product was extracted with DCM (3 x 15 mL), dried over MgSO$_4$ and concentrated *in vacuo*. Purification by column chromatography (SiO$_2$, 0-25% EtOAc in hexane) afforded **2.10** as a white solid (0.933 g, 98%).

M.P: 45 – 47 °C

IR υ_{max}/cm^{-1}: 2945, 1720, 1595

^{1}H NMR (300 MHz, CDCl$_3$): δ 8.09 (2 H, dd, *J*=8.4 Hz, H8, H12), 7.54 – 7.62 (1 H, m, H10), 7.41 – 7.49 (2 H, m, H9, H11), 6.69 (2 H, s, H2, H6), 5.30 (2 H, s, H14), 3.89 (6 H, s, H15, H17), 3.86 (3 H, s, H16) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 166.4 (C13), 153.3 (C3, C5), 138.0 (C7), 133.1 (C10), 131.6 (C4), 130.1 (C1), 129.7 (C9, C11), 128.4 (C8, C12), 105.5 (C2, C6), 67.0 (C14), 60.8 (C16), 56.1 (C15, C17) ppm

LRMS ES$^+$ *m/z* (%): 325 ([M+Na]$^+$, 48), 627 ([2M+Na]$^+$, 100).

2-Bromo-3,4,5-trimethoxybenzyl alcohol (2.14).
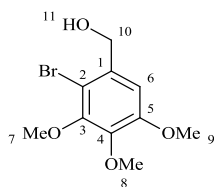
To a solution of 3,4,5-trimethoxybenzyl alcohol (1.181 g, 5.958 mmol) in CHCl$_3$ (40 mL) was added N-bromosuccinimide (1.110 g, 6.260 mmol). The reaction mixture was stirred at reflux for 3 h. The organic phase was washed with water (3 x 25 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 35% EtOAc in hexane) afforded **2.14** as a white solid (1.402 g, 85%).

M.P: 52 – 54 °C reported M.P : 54.5 – 55.5 °C[80]

IR υ$_{max}$/cm^{-1}: 3432, 2954, 1572, 1481, 1448, 1426, 1396

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.90 (1H, s, H6), 4.72 (2H, s, H10), 3.91 (3H, s, H8), 3.89 (6H, s, H7, H9), 1.95 (1H, br s, H11) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 153.0 (C), 150.9 (C), 135.3 (C), 118.4 (C), 108.4 (C), 107.7 (C6), 65.1 (C10), 61.1 (C7), 61.0 (C8), 56.2 (C9) ppm

LRMS ES$^+$ *m/z* (%): 331, 333 ([M+Na+MeOH]$^+$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[80]

2-Bromo-3,4,5-trimethoxybenzyl benzoate (2.15).
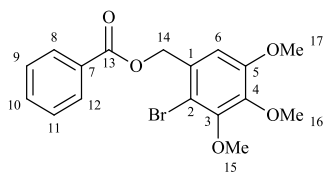
To a solution of 2-bromo-3,4,5-trimethoxybenzyl alcohol **2.14** (0.831 g, 0.003 mol) and benzoyl chloride (0.843 g, 0.006 mol) in acetonitrile (15 mL) under argon was added triethylamine (0.4 mL, 0.003 mol) dropwise. The reaction mixture was stirred at rt for 3 h and then poured into ice cold water (15 mL). The product was extracted with DCM (3 x 15 mL), dried over MgSO$_4$ and concentrated *in vacuo*. Purification by column chromatography (SiO$_2$, 0-25% EtOAc in hexane) afforded **2.15** as a colourless oil (1.070 g, 94%).

IR υ_{max}/cm^{-1}: 2933, 1724, 1576

^{1}H NMR (300 MHz, CDCl$_3$): δ 8.02 (2 H, d, *J* = 8.1 Hz, H8, H12), 7.42 – 7.57 (1 H, m, H10), 7.28 – 7.42 (2 H, m, H9, H11), 6.85 (1 H, s, H6), 5.34 (2 H, s, H14), 3.84 (3H, s, H17), 3.83 (3 H, s, H16), 3.78 (3 H, s, H15) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 170.2 (C13), 165.6 (C), 152.4 (C), 150.7 (C), 142.8 (C), 132.7 (C10), 130.4 (C), 129.2 (C19, C23), 128.0 (C8, C20), 109.8 (C), 109.0 (C6), 66.0 (C14), 60.6 (C15), 60.5 (C16), 55.7 (C17) ppm

LRMS ES$^+$ *m/z* (%): 403, 405 ([M+Na]$^+$, 100)

HRMS (ES): C$_{17}$H$_{17}$O$_5$Br [M+Na]$^+$ calculated 403.0152, found 403.0154.

3,4,5-Trimethoxybenzyl 2-bromobenzoate (2.21).
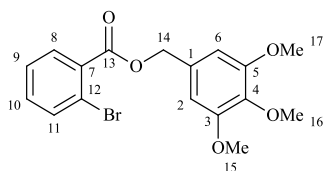
To a solution of 3,4,5-trimethoxybenzyl alcohol (0.83 mL, 0.005 mol) and 2-bromobenzoyl chloride (2.195 g, 0.010 mol) in acetonitrile (20 mL) under argon was added triethylamine

(0.7 mL, 0.005 mol) dropwise. The reaction mixture was stirred at rt for 3 h and then poured into ice cold water (20 mL). The product was extracted with DCM (3 x 20 mL), dried over MgSO$_4$ and concentrated *in vacuo*. Purification by column chromatography (SiO$_2$, 0-25% EtOAc in hexane) afforded **2.21** as a colourless oil (2.203 g, 58%).

IR υ$_{max}$/cm^{-1}: 2941, 1720, 1595, 1504

^{1}H NMR (300 MHz, (CD$_3$)$_2$CO): δ 7.77 – 7.87 (1 H, m, H8), 7.67 – 7.76 (1 H, m, H11), 7.41 – 7.53 (2 H, m, H9, H10), 6.83 (2 H, s, H2, H6), 5.30 (2 H, s, H14), 3.83 (6 H, s, H15, H17), 3.72 (3 H, s, H16) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ δ 165.3 (C13), 152.8 (C), 137.5 (C), 133.7 (CH), 132.2 (C), 131.6 (C), 130.8 (C), 130.6 (CH), 126.7 (CH), 121.0 (C12), 105.1 (C2, C6), 67.0 (C14), 60.2 (C16), 55.6 (C15, C17) ppm

LRMS ES$^+$ *m/z* (%): 403, 405 ([M+Na]$^+$, 100).

3,4,5-Trimethoxybenzyl-2-iodobenzoate (2.24).
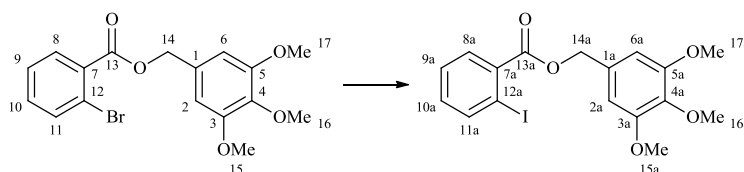
A mixture of copper iodide (0.014 g, 0.075 mmol) and sodium iodide (0.450 g, 3.00 mmol) was purged with argon for 20 min. To this mixture was added dioxane (2 mL), N,N'-dimethylethylene diamine (0.02 mL, 0.15 mmol) and a solution of 3,4,5-trimethoxybenzyl-2-bromobenzoate **2.21** (0.572 g, 1.50 mmol) in dioxane (1 mL). The solution was stirred at 110 °C for 23 h after which it was cooled to rt and diluted with 30% aqueous ammonia solution (7 mL). The product was extracted with DCM (3 x 25 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. The resulting crude material was purified by column chromatography (10% EtOAc in hexane) to yield a pale yellow oil (0.613 g).

^{1}H NMR spectroscopy and mass spectroscopy suggested the product was a mixture of starting material and iodinated product in a 2:1 ratio. The two compounds could not be separated by column chromatography or crystallisation.

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¹H NMR (400 MHz, CDCl₃): δ 7.99 (1 H, d, *J*=8.0 Hz, H8a), 7.80 (3 H, dd, *J*=7.5, 2.0 Hz, H11a, H8), 7.63 – 7.70 (2 H, m, H11), 7.31 – 7.42 (5 H, m, H9, H10, H9a), 7.15 (1 H, td, *J*=7.7, 1.8 Hz, H10a), 6.69-6.72 (6 H, m, H2, H6, H2a, H6a), 5.31 (6 H, s, H14, H14a), 3.88 (18 H, s, H17, H15, H17a, H15a), 3.86 (9 H, s, H16, H16a) ppm

LRMS ES⁺ *m/z* (%): 403, 405 ([M+Na]⁺, 100), 451 ([M+Na]⁺, 89).

2-Bromo-3,4,5-trimethoxybenzyl-2-bromobenzoate (2.27).
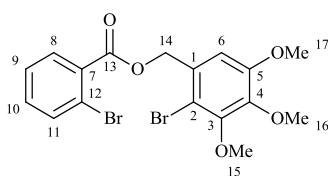
To a solution of 2-bromobenzyl chloride (1.750 g, 0.008 mol) and 2-bromo-3,4,5-trimethoxybenzyl alcohol **2.14** (1.108 g, 0.004 mol) in acetonitrile (16 mL) under argon, was added triethylamine (0.56 mL, 0.004 mol) dropwise. The reaction mixture was stirred at rt for 3 h and then poured into ice cold water (15 mL). The product was extracted with DCM (3 x 15 mL), dried over MgSO₄ and the solvent removed *in vacuo*. Column chromatography (15% EtOAc in hexane) afforded **2.27** as a yellow solid (1.836 g, quant.).

M.P: 42 – 44 °C

IR υ$_{max}$/cm⁻¹: 2937, 1727, 1591, 1565

¹H NMR (400 MHz, CDCl₃): δ 7.83 (1H, dd, *J*=7.0, 2.0 Hz, H8), 7.63 (1 H, dd, *J*=7.0, 1.0 Hz, H11), 7.28 – 7.37 (2 H, m, H9, H10), 6.93 (1 H, s, H6), 5.40 (2 H, s, H14), 3.89 (3 H, s, H17), 3.88 (3 H, s, H15), 3.85 (3 H, s, H16) ppm

¹³C NMR (101 MHz, CDCl₃): δ 165.3 (C13), 152.6 (C), 150.9 (C), 143.1 (C) 134.2 (C10), 132.6 (C8), 131.7 (C), 131.4 (C11) 130.1 (C), 127.1 (C9), 121.5 (C12), 110.0 (C2), 109.3 (C6), 67.0 (C14), 60.9 (C15), 60.85 (C16), 56.1 (C17) ppm

LRMS ES⁺ *m/z* (%): 513, 515, 517 ([M+Na+MeOH]⁺, 100)

HRMS (ES): C₁₇H₁₆O₅Br₂Na [M+Na]⁺ calculated 480.9257, found 480.9263.

2-Bromo-1-(((2-bromobenzyl)oxy)methyl)-3,4,5-trimethoxybenzene (2.33).
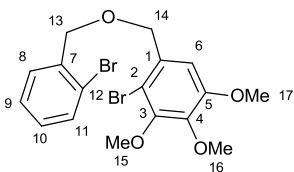
To a stirred solution of 2-bromo-3,4,5-trimethoxybenzylalcohol **2.14** (0.420 g, 1.5 mmol), 2-bromobenzaldehyde (0.12 mL, 1 mmol) and triethylsilane (0.24 mL, 1.5 mmol) in DCM (5 mL) at 0°C was added TFA (0.22 mL, 3 mmol) dropwise. The colourless solution was gradually warmed to rt. After 16 h the reaction mixture was poured into water (5 mL) and the organic layer separated, dried over $MgSO_4$ and the solvent removed *in vacuo*. The crude was purified by column chromatography (10% EtOAc in hexane) to yield the desired product **2.33** as a colourless oil (0.105 g, 24%).

IR υ_{max}/cm^{-1}: 2936, 1568, 1482, 1393

^{1}H NMR (400 MHz, CDCl$_3$): δ 7.51 – 7.58 (2 H, m, H8, H11), 7.33 (1 H, td, *J*=7.5, 7.5, 1.0 Hz, H9), 7.17 (1 H, td, *J*=7.8, 7.8, 2.0 Hz, H10), 6.97 (1 H, s, H6), 4.72 (2 H, s, H14), 4.67 (2 H, s, H13), 3.91 (3 H, s, H17), 3.89 (3 H, s, H15), 3.87 (3 H, s, H16) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 152.8 (C), 150.7 (C), 142.3 (C), 137.3 (C), 132.6 (CH), 129.3 (CH), 129.1 (CH), 127.4 (CH), 122.8 (C), 109.5 (C), 108.7 (C), 107.8 (C6), 72.1 (C14), 72.0 (C13), 61.0 (C15), 60.9 (C16), 56.1 (C17) ppm

LRMS ES$^+$ *m/z* (%): 467, 469, 471 ([M+Na]$^+$, 100)

HRMS (ES): C$_{17}$H$_{18}$O$_4$Br$_2$Na [M+Na]$^+$ calculated 466.9464, found 466.9463.

5,7-Dihydroxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (2.38).
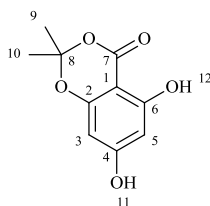
To a stirred suspension of 2,4,6-trihydroxybenzoic acid monohydrate (5.000 g, 27mmol) in TFA (40 mL, 537 mmol) at 0 °C was added TFAA (25 mL, 180 mmol) and acetone (5 mL, 68 mmol). The reaction mixture was slowly warmed to rt and left for 12 h. NaHCO$_3$ was added until no further effervescence occurred. The product was extracted with EtOAc (3 x 50 mL), dried over MgSO$_4$ and concentrated *in vacuo*. Purification by column chromatography (SiO$_2$, 35% EtOAc in hexane) afforded **2.38** as a white solid (3.418 g, 61 %).

M.P: 194 – 196 °C reported M.P: 203 – 206 °C[48]

IR υ_{max}/cm^{-1}: 3236, 1637, 1591, 1482

^{1}H NMR (300 MHz, (CD$_3$)$_2$CO): δ 10.45 (1 H, s, H12), 9.66 (1 H, br. s, H11), 6.08 (1H, d, J = 1.8 Hz, H5), 6.00 (1H, d, J = 2.2 Hz, H3), 1.72 (6 H, s, H9, H10) ppm

^{13}C NMR (75 MHz, (CD$_3$)$_2$CO): δ 167.3 (C3), 165.9 (C), 164.2 (C), 158.3 (C), 107.8 (C), 98.1 (C5), 96.3 (C3), 93.2 (C8), 25.7 (C9, C10) ppm

LRMS ES$^+$ *m/z* (%):209 ([M-H]$^-$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[47]

5-Hydroxy-7-methoxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (2.39).
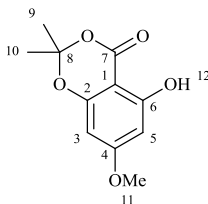
To a solution of 5,7-dihydroxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one **2.38** (4.767 g, 22.7 mmol) and methanol (0.96 mL, 23.8 mmol) in THF (60 mL) at 0 °C under argon, was added triphenylphosphine (6.247 g, 23.8 mmol) and DIAD (4.7 mL, 23.8 mmol). The reaction mixture was warmed to rt and stirred for 2 h. The product was extracted with EtOAc (3 x 30 mL), washed with water (3 x 20 mL) and brine (3 x 20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 10% EtOAc in hexane) afforded **2.39** as a white solid (4.403 g, 87%).

M.P: 104 – 107 °C reported M.P: 108 – 109 °C[47]

IR υ_{max}/cm^{-1}: 3183, 1690, 1629

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.43 (1 H, s, H12), 6.14 (1H, d, J = 2.3 Hz, H5), 6.00 (1H, d, J = 2.3 Hz, H3), 3.81 (3 H, s, H11) 1.72 (6 H, s, H9, H10) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 167.7 (C7), 165.2 (C), 163.1 (C), 156.9 (C), 106.9 (C), 95.7 (C5), 94.6 (C3), 93.1 (C8), 55.7 (C11), 25.6 (C9, C10) ppm

LRMS ES$^+$ m/z (%): 247 ([M+Na]$^+$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[47]

7-Methoxy-2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate (2.40).
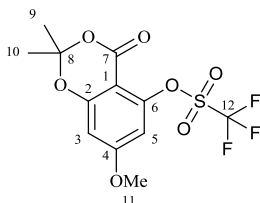
To a biphasic solution of 5-hydroxy-7-methoxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one **2.39** (3.834 g, 17.1 mmol) in toluene (80 mL) and aqueous LiOH (1.229 g, 51.3 mmol in 25 mL H$_2$O) at 0 °C, was added trifluoromethanesulfonic anhydride (6 mL, 35.4 mmol) dropwise. The reaction mixture was stirred at 0 °C for 3 h after which EtOAc (50 mL) was added. The organic phase was washed with sat. NaHCO$_3$ solution (3 x 30 mL) and water (3 x 25 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 10% EtOAc in hexane) afforded **2.40** as a white solid (5.293 g, 87%).

M.P: 63 – 66 °C reported M.P: 58 – 59 °C[47]

IR υ_{max}/cm^{-1}: 1743, 1629, 1576

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.53 (1 H, d, J = 2.3 Hz, H5), 6.48 (1 H, d, J = 2.3 Hz, H3), 3.87 (3 H, s, H11), 1.73 (6 H, s, H9, H10) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 165.5 (C7), 158.8 (C4), 157.0 (C), 149.9 (C), 118.7 (C12, d, *J*=321.8 Hz), 106.6 (C), 105.3 (C5), 101.1 (C3), 101.0 (C), 56.3 (C11), 25.5 (C9, C10) ppm

LRMS ES$^+$ *m/z* (%): 357 ([M+H]$^+$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[47]

4-Hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.45).
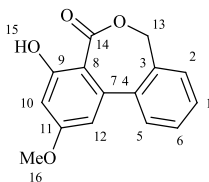
To a mixture of 7-methoxy-2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate **2.40** (0.100 g, 0.281 mmol), K$_3$PO$_4$ (0.089 g, 0.421 mmol), PdCl$_2$(dppf) (0.010g, 0.014 mmol) and 2-(hydroxymethyl)benzene boronic acid hemi-ester (0.041 g, 0.309 mmol) was added THF (5 mL) under argon. The reaction mixture was stirred at reflux for 5 h after which a further 0.75 eq. of 2-(hydroxymethyl)benzene boronic acid hemi-ester (0.028 g, 0.211 mmol) was added. The reaction mixture was stirred at reflux for a further 16 h and then cooled to rt. 3 M NaOAc solution (0.5 mL) and 30% H$_2$O$_2$ (0.5 mL) were added and the reaction mixture stirred at rt for 1 h. The product was extracted with EtOAc (10 mL), filtered through celite and washed with water (10 mL) and brine (10 mL). The organic phase was dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 35% EtOAc in hexane) afforded **2.45** as a white solid (0.024 g, 34%).

M.P: 98 – 100 °C

IR υ$_{max}$/cm^{-1}: 3044, 1654, 1612, 1568

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.65 (1 H, s, H15), 7.46 – 7.57 (2 H, m, H5, H6), 7.40 – 7.45 (2 H, m, H1, H2), 6.65 (1 H, d, *J*=2.6 Hz, H12), 6.62 (1 H, d, *J*=2.6 Hz, H10), 5.17 (1 H, br s, H13), 5.00 (1 H, br s, H13), 3.90 (3 H, s, H16) ppm

¹³C NMR (75 MHz, CDCl$_3$): δ 172.6 (C14), 164.0 (C), 163.8 (C), 141.1 (C), 139.4 (C), 134.2 (C), 130.2 (CH), 129.3 (CH), 128.9 (CH), 128.0 (CH), 109.2 (CH), 106.3 (C), 101.2 (CH), 69.8 (C13), 55.6 (C16) ppm

LRMS ES⁺ *m/z* (%): 255 ([M-H]⁻, 94)

HRMS (ES): C$_{15}$H$_{12}$O$_4$Na [M+Na]⁺ calculated 279.0628, found 279.0637.

4-Hydroxy-2-methoxy-10-nitrodibenzo[c,e]oxepin-5(7H)-one (2.47).
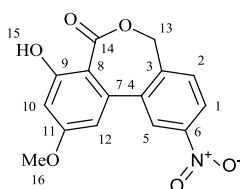
To a mixture of 7-methoxy-2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate **2.40** (0.178 g, 0.500 mmol), K$_3$PO$_4$ (0.151 g, 0.750 mmol), PdCl$_2$(dppf) (0.092g, 0.013 mmol) and 2-(hydroxymethyl)-4-nitrobenzene boronic acid hemi-ester (0.098 g, 0.550 mmol) was added THF (10 mL) under argon. The reaction mixture was stirred at reflux for 24 h and then cooled to rt. 3 M NaOAc solution (0.5 mL) and 30% H$_2$O$_2$ (0.5 mL) were added and the reaction mixture stirred at rt for 1 h. The product was extracted with EtOAc (10 mL), filtered through celite and washed with water (10 mL) and brine (10 mL). The organic phase was dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 10% EtOAc in hexane) afforded the desired product **2.47** as a yellow solid (0.056 g, 32%).

M.P: 186 – 188 °C

IR υ$_{max}$/cm^{-1}: 3023, 1729, 1663, 1613, 1528, 1423, 1347

¹H NMR (300 MHz, CDCl$_3$): δ 10.61 (1 H, s, H15), 8.55 (1 H, d, *J*=2.2 Hz, H5), 8.44 (1 H, d, *J*=2.2 Hz, H5), 8.35 (1 H, dd, *J*=8.2, 2.4 Hz, H1), 8.30 (1 H, dd, *J*=8.4 Hz, H1), 7.71 (1 H, d, *J*=8.4 Hz, H2), 7.64 (1 H, d, *J*=8.4 Hz, H2), 7.15 (1 H, d, *J*=2.6 Hz, H12), 7.05 (1 H, d, *J*=2.2 Hz, H12), 6.69 (2 H, q, *J*=2.6 Hz, H14), 5.04 – 5.25 (4 H, m, H13), 4.01 (3 H, s, H16), 3.93 (3 H, s, H16) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 164.4 (C), 164.3 (C), 162.6 (C), 149.4 (C), 149.1 (C), 141.1 (C), 141.0 (C), 140.2 (C), 138.9 (C), 138.5 (C), 138.2 (C), 130.3 (CH), 129.3 (CH), 124.4 (CH), 124.3 (CH), 123.6 (CH), 123.6 (CH), 113.9 (CH), 110.1 (CH), 110.0 (CH), 102.2 (CH), 68.5 (C13), 67.5 (C13), 56.4 (C16), 55.8 (C16) ppm

LRMS ES *m/z* (%): 300 ([M - H]$^-$, 100).

(2-Bromo-4-methylphenyl)methanol (2.51).
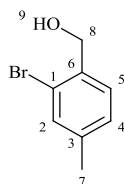
To a solution of ZrCl$_4$ (0.682 g, 2.93 mmol) in THF (17 mL) was added NaBH$_4$ (0.443 g, 11.70 mmol) at 0 °C under argon. The reaction mixture was stirred for 10 min after which a solution of 2-bromo-4-methylbenzoic acid (0.500 g, 2.34 mmol) in THF (4 mL) was added dropwise. After stirring at rt for 16 h the product was extracted with EtOAc (20 mL), washed with water (2 x 20 mL) and brine (2 x 20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 35% EtOAc in hexane) afforded **2.51** as a white solid (0.420 g, 90%).

M.P: 42 – 44 °C reported M.P: 57 °C[81]

IR υ$_{max}$/cm^{-1}: 3330, 1606, 1490, 1446

^{1}H NMR (300 MHz, CDCl$_3$): δ 7.39 (1 H, d, *J*=1.5 Hz, H2), 7.34 (1 H, d, *J*=7.7 Hz, H5), 7.13 (1 H, dd, *J*=7.7, 1.5 Hz, H4), 4.72 (2 H, s, H8), 2.34 (3 H, s, H7), 1.95 (1 H, br s, H9) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 139.4 (C6), 136.7 (C7), 133.1 (C2), 128.9 (C5), 128.4 (C4), 122.6 (C1), 65.0 (C8), 20.7 (C7) ppm

HRMS (ES): C$_8$H$_9$OBrNa [M+Na]$^+$ calculated 222.9729, found 222.9728.

The ^{1}H NMR spectra corresponded to the reported data.[81]

7-Methoxy-2,2-dimethyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-4H-benzo[d][1,3]dioxin-4-one (2.55).
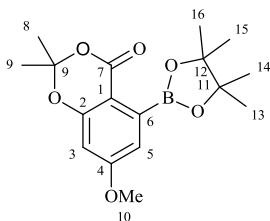
A solution of 7-methoxy-2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate **2.40** (1.425 g, 4.2 mmol), Pd(PPh$_3$)$_4$ (0.277 g, 0.2 mmol) and freshly distilled triethylamine (1.7 mL, 12.6 mmol) in THF (20 mL) was degassed under argon in a ultrasonication bath for 10 min. A 1M solution of 4,4,5,5-Tetramethyl-1,3,2-dioxaborolane in THF (21 mL, 21 mmol) was added dropwise over 5 min and the reaction mixture heated to reflux for 4 h. The solvent was removed *in vacuo* to yield the crude product. Column chromatography (SiO$_2$, 10% EtOAc in hexane) afforded **2.55** as a white solid (1.230 g, 92%).

M.P: 88 – 90 °C reported M.P: 108 – 110 °C[53]

IR υ_{max}/cm^{-1}: 1720, 1603, 1580, 1444

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.65 (1 H, d, *J*=2.4 Hz, H5), 6.38 (1 H, d, *J*=2.4 Hz, H3), 3.82 (3 H, s, H10), 1.70 (6 H, s, H8, H9), 1.40 (12 H, s, H13 – H16) ppm

LRMS ES$^+$ *m/z* 357 ([M+Na]$^+$, 100)

HRMS (ES): C$_{17}$H$_{23}$BO$_6$Na [M+Na]$^+$ calculated 357.1480, found 357.1486.

The ^{1}H NMR spectra corresponded to the reported data.[53]

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5-(2-(Hydroxymethyl)-5-methylphenyl)-7-methoxy-2,3-dimethyl-4H-benzo[d][1,3] dioxin-4-one (2.57).
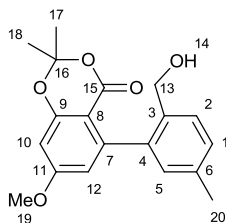
To a stirred solution of the pinacol ester **2.55** (0.050 g, 0.150 mmol) in THF (5 mL), under argon, was added (2-bromo-4-methylphenyl)methanol **2.51** (0.045 g, 0.224 mmol), $PdCl_2(dppf)$ (0.006 g, 0.0078 mmol) and K_3PO_4 (0.048 g, 0.244 mmol). The reaction mixture was stirred at reflux for 24 h and then cooled to rt. 3 M NaOAc solution (0.5 mL) and 30% H_2O_2 (0.5 mL) were added and the reaction mixture stirred at rt for 1 h. EtOAc (10 mL) was added and the solution filtered through celite. The resulting filtrate was washed with water (3 x 10 mL) and brine (3 x 10 mL), dried over $MgSO_4$ and the solvent removed *in vacuo*. Column chromatography (SiO_2, 10% EtOAc in hexane) afforded **2.57** as a thick yellow paste (0.026 g, 56%).

M.P: 39 – 41 °C

IR υ_{max}/cm^{-1}: 3463, 2941, 1735, 1610, 1580, 1289

^{1}H NMR (400 MHz, CDCl$_3$): δ 7.41 (1 H, d, *J*=7.7 Hz, H1), 7.22 (1 H, d, *J*=7.5 Hz, H2), 6.92 (1 H, s, H5), 6.47 – 6.50 (1 H, m, H12), 6.44 – 6.47 (1 H, m, H10), 4.41 (1 H, d, *J*=12.0 Hz, H13), 4.37 (1 H, d, *J*=12.3 Hz, H13), 3.86 (3 H, s, H19), 2.37 (3 H, s, H20), 1.78 (3 H, s, H18), 1.74 (3 H, s, H17) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 164.6 (C15), 160.5 (C), 158.6 (C), 146.1 (C), 139.8 (C1), 137.3 (C), 134.8 (C2), 129.0 (C), 128.9 (C5), 113.3 (C12), 105.7 (C), 105.5 (C), 100.9 (C10), 63.4 (C13), 55.8 (C19), 26.0 (C18), 25.3 (C17), 21.2 (C20) ppm

LRMS ES *m/z* (%): 351 ([M+Na]$^+$, 100)

HRMS (ES): $C_{19}H_{20}O_5Na$ [M+Na]$^+$ calculated 351.1203, found 351.1209.

4-Hydroxy-2-methoxy-10-methyldibenzo[c,e]oxepin-5(7H)-one (2.58).
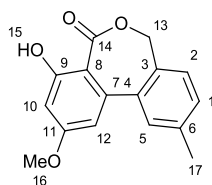
To a stirred solution of **2.57** (0.026 g, 0.083 mmol) in THF (2 mL), under argon, was added NaH (0.004 g, 0.166 mmol) at rt. After 5 hours the reaction was diluted with EtOAc (10 mL) and the organic solution washed with brine (3 x 5 mL) and NH_4Cl (3 x 5 mL). The solution was dried over magnesium sulphate and the solvent removed *in vacuo* to yield the desired product **2.58** without further purification as a white solid (0.019 g, 50%).

M.P: 99 – 101 °C

^{1}H NMR (400 MHz, $CDCl_3$): δ 10.66 (1 H, s, H15), 7.37 (1 H, s, H5), 7.31 (1 H, d, *J*=8.0 Hz, H2), 7.24 (1 H, d, *J*=8.0 Hz, H1), 6.65 (1 H, d, *J*=2.5 Hz, H12), 6.61 (1 H, d, *J*=2.5 Hz, H10), 5.16 (1 H, br s, H13), 4.94 (1 H, br s, H13), 3.90 (3 H, s, H17), 2.44 (3 H, s, H17) ppm

^{13}C NMR (101 MHz, $CDCl_3$): δ 180.8 (C14), 172.7 (C11), 164.0 (C9), 163.7 (C), 140.2 (C), 139.3 (C), 131.4 (C), 129.9 (C2), 129.6 (C1), 127.9 (C5), 109.0 (C12), 106.4 (C), 101.1 (C10), 69.6 (C13), 55.6 (C16), 21.4 (C17) ppm

LRMS ES *m/z* (%): 293 ([M+Na]$^+$, 100)

HRMS (ES): $C_{16}H_{14}O_4Na$ [M+Na]$^+$ calculated 293.0784, found 293.0782.

4-Hydroxy-2,9,10,11-tetramethoxydibenzo[c,e]oxepin-5(7H)-one (2.60).
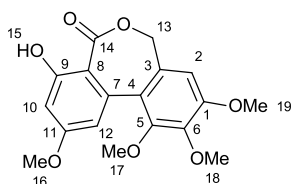
To a stirred solution of the pinacol ester **2.55** (0.235 g, 0.703 mmol) in THF (15 mL), under argon, was added 2-bromo-3,4,5-trimethoxybenzylalcohol **2.14** (0.312 g, 1.13 mmol), $PdCl_2$(dppf) (0.031 g, 0.042 mmol) and K_3PO_4 (0.299 g, 1.41 mmol). The reaction mixture

was stirred at reflux for 24 h and then cooled to rt. 3 M NaOAc solution (3 mL) and 30% H_2O_2 (3 mL) were added and the reaction mixture stirred at rt for 1 h. EtOAc (30 mL) was added and the solution was filtered through celite. The organic filtrate was washed with water (3 x 15 mL) and brine (3 x 15 mL), dried over $MgSO_4$ and the solvent removed *in vacuo*. Column chromatography (SiO_2, 10% EtOAc in hexane) afforded a mixture of cyclised and uncyclised product (0.248 g, 0.61 mmol).

The crude was taken up in THF (5 mL) and cooled to 0 °C. NaH (0.022 g, 0.92 mmol) was added and the reaction mixture at 0 °C for 6 h. EtOAc (10 mL) was added and the organic solution washed with water (3 x 10 mL) and brine (3 x 10 mL), dried over $MgSO_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO_2, 10% EtOAc in hexane) afforded the desired product as a white solid. However, the product contained an unknown impurity which could not be removed with acid and base washes, recrystallisation or further column chromatography. The impure product was isolated as an off-white solid (0.053 g, 22%).

^{1}H NMR (300 MHz, $CDCl_3$): δ 10.19 (1 H, s, H15), 6.86 (1 H, d, *J*=2.6 Hz, H12), 6.75 (1 H, s, H2), 6.58 (1 H, d, *J*2.6 Hz, H10), 5.10 (1 H, d, *J*=12.1 Hz, H13), 4.82 (1 H, d, *J*=12.1 Hz, H13), 3.93 (3 H, s, CH_3), 3.90 (3 H, s, CH_3), 3.88 (3 H, s, CH_3), 3.62 (3 H, s, H16) ppm

LRMS ES *m/z* (%): 369 ([M+Na]$^+$, 100)

Attempts to obtain ^{13}C and DEPT NMR spectra were hampered by the degradation of the material over time.

(2-Bromo-5-methoxyphenyl)methanol (2.64).
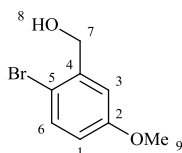
To a solution of $ZrCl_4$ (0.629 g, 2.7 mmol) in THF (16 mL) was added $NaBH_4$ (0.409 g, 10.8 mmol) under argon. After stirring for 10 min, a solution of 2-bromo-5-methoxylbenzoic acid (0.500 g, 2.16 mmol) in THF (4 mL) was added, and the reaction mixture stirred for a further 16 h. The product was extracted with EtOAc (20 mL), washed with water (2 x 20

mL) and brine (2 x 20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (35% EtOAc in hexane) afforded **2.64** as a white solid (0.416 g, 87%).

M.P: 33 – 35 °C reported M.P: 46.5 – 48.5 °C[82]

IR υ_{max}/cm^{-1}: 3319, 2911, 2823, 1467, 1013

^{1}H NMR (300 MHz, CDCl$_3$): δ 7.42 (1H, d, *J*=8.7 Hz, H6), 7.07 (1H, d, *J*=3.1 Hz, H3), 6.72 (1H, dd, *J*=8.7, 3.1 Hz, H1), 4.71 (2H, s, H7), 3.81 (3H, s, H9), 2.02 (1H, br. s, H8) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 159.3 (C2), 140.7 (C4), 133.1 (C6), 114.8 (C1), 114.3 (C3), 112.5 (C5), 65.0 (C7), 55.5 (C9) ppm

HRMS (ES): C$_8$H$_9$O$_2$BrNa [M+Na]$^+$ calculated 238.9678, found 238.9677.

The ^{1}H NMR spectra corresponded to the reported data.[82]

4-Hydroxy-2,9-dimethoxydibenzo[c,e]oxepin-5(7H)-one (2.66).
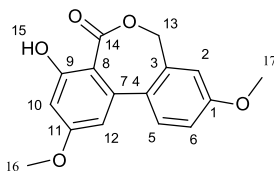
To a stirred solution of **2.65** (0.020 g, 0.058 mmol) in THF (2 mL) under argon was added NaH (0.001 g, 0.070 mmol) at 0 °C. The colourless solution was warmed to rt over 3 hour after which EtOAc (10 mL) was added. The organic solution was washed with brine (3 x 10 mL) and NH$_4$Cl (3 x 10 mL), dried over MgSO$_4$ and the solvent removed *in vacuo* to yield the desired product **2.66** as an off-white solid (0.010 g, 62%).

M.P: 166 – 168 °C

IR υ_{max}/cm^{-1}: 3089, 3023, 1729, 1663, 1613

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.59 (1 H, s, H15), 7.41 (1 H, d, *J*=8.4 Hz, H5), 6.95 (1 H, dd, *J*=8.6, 2.7 Hz, H6), 6.86 (1 H, d, *J*=2.9 Hz, H2), 6.50 (2 H, q, *J*=2.6 Hz, H10, H12), 5.13 (1 H, br. s, H13), 4.78 (1 H, br. s, H13), 3.81 (3 H, s, H16), 3.80 (3 H, s, H17) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.8 (C14), 164.1 (C11), 164.0 (C9), 160.1 (C1), 141.1 (C7), 135.5 (C3), 131.8 (C4), 130.7 (C5), 115.6 (C6), 113.4 (C2), 108.8 (C10), 106.0 (C8), 100.7 (C12), 69.9 (C13), 55.6 (C16), 55.6 (C17) ppm

LRMS ES$^+$ *m/z* (%): 287 ([M+H]$^+$, 100)

HRMS (ES): C$_{16}$H$_{14}$O$_5$Na [M+Na]$^+$ calculated 309.0733, found 309.0735.

3,4-Dihydroxy-5-methoxybenzaldehyde (2.68).
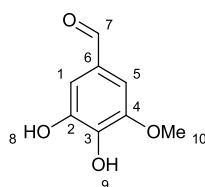
A stirred slurry of 5-bromovanillin (10.000 g, 0.044 mol), sodium hydroxide (12.120 g, 0.302 mol) and copper powder (0.056 g, 0.001 mmol) in water (150 mL) was heated to reflux. After 24 h Na$_2$HPO$_4$ (0.246 g, 0.002 mol) was added and the reaction mixture heated for a further 30 min. The mixture was cooled to rt and filtered to remove a whitish green precipitate. The filtrate was acidified to pH 4 using 1 N HCl and the product then extracted with EtOAc (3 x 300 mL). The filtrate was stirred in activated carbon, filtered and the organic solution washed with saturated EDTA solution (3 x 100 mL) and brine (3 x 100 mL). The combined organic extracts were dried over MgSO$_4$ and the solvent removed *in vacuo*. Recrystallisation of the resulting crude from toluene furnished the desired product **2.68** as an off white solid (3.936 g, 54%).

 M.P: 108 – 110 °C reported M.P: 132 – 133 °C[55]

IR υ$_{max}$/cm^{-1}: 3296, 1675, 1588, 1508, 1459

^{1}H NMR (300 MHz, (CD$_3$)$_2$SO): δ 10.76 (1 H, br. s, H7), 9.77 (1 H, s, H8, H9), 7.73 (1 H, s, H1), 7.41 (1 H, s, H5), 3.91 (3 H, s, H10) ppm

^{13}C NMR (75 MHz, (CD$_3$)$_2$SO): δ 190.7 (C7), 150.1 (C), 148.9 (C), 129.2 (C), 129.1 (C), 109.8 (C), 109.5 (C), 56.6 (C10) ppm

LRMS ES- *m/z* (%): 167 ([M-H]-, 100).

2-Bromo-3,4-dihydroxy-5-methoxybenzaldehyde (2.69).
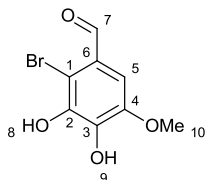
To a stirred solution of 3,4-dihydroxy-5-methoxybenzaldehyde **2.68** (0.505 g, 3.0 mmol) in acetic acid (1 mL) was added bromine (0.16 mL, 3.15 mmol) in acetic acid (1 mL) at rt. A yellow precipitate formed on addition of the bromine which was filtered off and washed with saturated sodium thiosulphate solution (10 mL) and water (10 mL). Recrystallisation of the resulting crude from ethanol furnished the desired product **2.69** as a yellow crystalline solid (0.300 g, 41%).

M.P: 150 – 152 °C reported M.P: 171 – 172 °C[57]

IR υ_{max}/cm^{-1}: 3100, 1656, 1572, 1486, 1459

^{1}H NMR (400 MHz, (CD$_3$)$_2$CO): δ 10.22 (1 H, s, H7), 8.73 (2 H, br. s, H8, H9), 7.12 (1 H, s, H5), 3.91 (1 H, s, H10) ppm

LRMS ES- *m/z* (%): 245, 247 ([M-H]-, 100).

The ^{1}H NMR spectra corresponded to the reported data.[57]

3-Bromo-4-formyl-6-methoxy-1,2-phenylene diacetate (2.70).
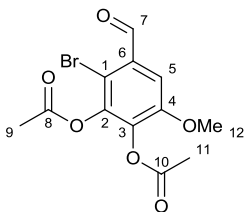
To a solution of 2-bromo-3,4-dihydroxy-5-methoxybenzylalcohol **2.69** (1.20 g, 5.1 mmol), triethylamine (1.8 mL, 12.7 mmol) and DMAP (0.025 g, 0.2 mmol) in DCM (20 mL), was added acetic anhydride (1.2 mL, 12.7 mmol) under argon. The reaction mixture was stirred at rt for 90 min after which the organic solution was washed with saturated Na_2CO_3 solution (3 x 10 mL) and water (3 x 10 mL), dried over $MgSO_4$ and the solvent removed *in vacuo*. Column chromatography (SiO_2, 40% EtOAc in hexane) afforded the desired product **2.70** as a white crystalline solid (1.251 g, 78%).

M.P: 112 – 114 °C reported M.P: 125 °C[56]

IR υ_{max}/cm^{-1}: 1782, 1694, 1384, 1371

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.19 (1 H, s, H7), 7.38 (1 H, s, H5), 3.80 (3 H, s, H12), 2.29 (3 H, s, H11), 2.22 (3 H, s, H9) ppm

LRMS ES$^+$ *m/z* (%): 385, 387 ([M+Na+MeOH]$^+$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[56]

3,4-Diacetoxy-2-bromo-5-methoxybenzyl alcohol (2.71).
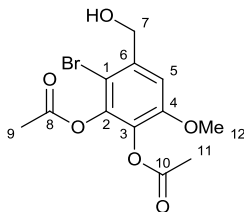
To a solution of 3-bromo-4-formyl-6-methoxy-1,2-phenylene diacetate **2.70** (0.320 g, 0.97 mmol) in MeOH (10 mL) at 0 °C, was added NaBH$_4$ (0.018 g, 0.48 mmol). The reaction mixture was stirred at 0 °C for 30 min after which water (10 mL) was added and the solution acidified with 1 M H_2SO_4. The product was extracted with DCM (3 x 10 mL), the combined organic extractions dried over $MgSO_4$ and the solvent removed *in vacuo*. The desired product **2.71** was isolated as a purple solid with no further purification required (0.308 g, 96%).

M.P: 114 – 116 °C reported M.P: 138 °C[56]

IR υ_{max}/cm^{-1}: 3501, 1777, 1493

^{1}H NMR (300 MHz, (CD$_3$)$_2$CO): δ 7.31 (1 H, s, H5), 4.67 (2 H, s, H7), 3.87 (3 H, s, H12), 2.33 (3 H, s, H11), 2.26 (3 H, s, H9) ppm

^{13}C NMR (75 MHz, (CD$_3$)$_2$CO): δ 168.1 (C8), 167.9 (C10), 152.7 (C), 142.4 (C), 140.7 (C6), 132.9 (C4), 109.6 (C5), 106.6 (C1), 64.2 (C7), 56.8 (C12), 20.3 (C9), 20.1 (C11) ppm.

The ^{1}H NMR spectra corresponded to the reported data.[56]

4-Hydroxy-2,9,10-trimethoxydibenzo[c,e]oxepin-5(7H)-one (2.78).
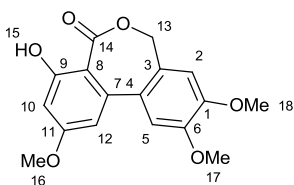
An oven dried two-necked flask was cooled under vacuum and backfilled with argon. Pinacol ester **2.55** (0.200 g, 0.600 mmol), 2-bromo-4,5-dimethoxybenzylalcohol (0.148 g, 0.600 mmol) and potassium phosphate (0.256 g, 1.2 mmol) were added and the flask evacuated and backfilled with argon three times. A second oven dried two-necked flask was cooled under vacuum and backfilled with argon. Tris(dibenzylideneacetone)palladium (0.005 g, 0.006 mmol) and SPhos (0.010 g, 0.024 mmol) were added and the flask was evacuated and backfilled with argon three times. THF (2 x 1 mL) was added to each flask, the palladium complex transferred to the reaction flask with additional THF (3 mL) and the reaction mixture stirred at reflux for 21 h. The reaction mixture was cooled, diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid. The product was washed with ether (5 mL) and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL), extracted with EtOAc (2 x 5 mL) and the organic layer dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL) and the organic layer dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The product **2.78** was isolated from the acidic extraction as a white solid (0.064 g, 34%).

M.P: 192 – 194 °C

IR υ_{max}/cm^{-1}: 2938, 1652, 1607, 1567, 1520

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.69 (1 H, s, H15), 7.03 (1 H, s, H2), 6.92 (1 H, s, H5), 6.61 (1 H, d, J=2.6 Hz, H12), 6.59 (1 H, d, J=2.6 Hz, H10), 5.15 (1 H, br. s, H13), 4.90 (1 H, br. s, H13), 3.96 (3 H, s), 3.95 (3 H, s), 3.91 (3 H, s) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 172.7 (C14), 163.9 (C11), 150.2 (C), 150.0 (C), 149.4 (C), 132.0 (C), 126.8 (C), 112.1 (C2), 110.8 (C5), 108.9 (C12), 104.6 (C), 104.2 (C), 100.4 (C10), 69.5 (C13), 56.2 (C16, 18), 55.6 (C17) ppm

LRMS ES$^+$ m/z (%): 339 ([M+Na]$^+$, 100)

HRMS (ES): C$_{17}$H$_{16}$O$_6$Na [M+Na]$^+$ calculated 339.0839, found 339.0836.

Methyl-2-bromo-3,5-dimethoxybenzoate (2.80).
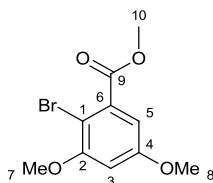
To a stirred solution of methyl-3,5-dimethoxybenzoate (1.000 g, 5.1 mmol) in acetonitrile (8 mL) at 0°C was added *N*-bromosuccinimide (0.910 g, 5.1 mmol). The solution was stirred for 7 h after which the product was extracted with EtOAc (2 x 25 mL), washed with brine (1 x 25 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 20% EtOAc in hexane) afforded **2.80** as a white solid (1.133 g, 80%).

M.P: 54 – 57 °C reported M.P: 55 – 57 °C[59]

IR υ_{max}/cm^{-1}: 1720, 1572, 1433

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.78 (1 H, d, J=2.6 Hz, H5), 6.56 (1 H, d, J=2.6 Hz, H3), 3.91 (3 H, s, H10), 3.86 (3 H, s, H7), 3.80 (3 H, s, H8) ppm

LRMS ES+ *m/z* (%): 329, 331 ([M+Na+MeOH]+, 100).

The ^{1}H NMR spectra corresponded to the reported data.[59]

2-Bromo-3,5-dimethoxybenzylalcohol (2.81).
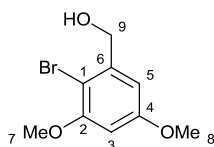
To a stirred solution of methyl 2-bromo-3,5-dimethoxybenzoate **2.80** (1.000 g, 3.6 mmol) in THF (10 mL) at 0°C was added a 1N solution of DIBAL in hexane (9.1 mL, 9.1 mmol) dropwise. After 30 min a 1M solution of potassium phosphate (15 mL) was added and the product extracted with ether (3 x 10 mL). The organic solution was washed with brine (15 mL), dried over MgSO₄ and the solvent removed *in vacuo*. Purification by column chromatography (SiO₂, 30% EtOAc in hexane) afforded the desired product **2.81** as a white solid (0.764 g, 85%)

M.P: 90 – 93 °C reported M.P: 94 – 95.2 °C[59]

IR υ$_{max}$/cm^{-1}: 3277, 1588, 1455, 1417

^{1}H NMR (400 MHz, CDCl₃): δ 6.71 (1 H, d, *J*=2.8 Hz, H5), 6.45 (1 H, d, *J*=2.8 Hz, H3), 4.74 (2 H, s, H9), 3.88 (3 H, s, H7), 3.83 (3 H, s, H8) ppm

LRMS ES *m/z* (%): 269, 271 [M + Na]+, 100).

The ^{1}H NMR spectra corresponded to the reported data.[59]

4-Hydroxy-2,9,11-trimethoxydibenzo[c,e]oxepin-5(7H)-one (2.82).
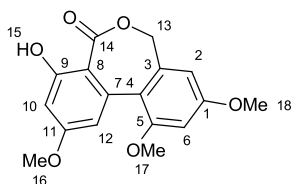
This product was synthesised by Joanna Loh, an undergraduate student on a 6 month placement from Singapore.

An oven dried two-necked flask was cooled under vacuum and backfilled with argon. Pinacol ester **2.55** (0.250 g, 0.75 mmol), 2-bromo-3,5-dimethoxybenzylalcohol **2.81** (0.124 g, 0.50 mmol) and potassium phosphate (0.212 g, 1.00 mmol) were added and the flask evacuated and backfilled with argon three times. A second oven dried two-necked flask was cooled under vacuum and backfilled with argon. Tris(dibenzylideneacetone)palladium (0.005 g, 0.01 mmol) and SPhos (0.008 g, 0.02 mmol) were added to the flask which was then evacuated and backfilled with argon three times. THF (2 x 0.75 mL) was added to each flask after which the Palladium complex was transferred to the reaction flask with additional THF (2 mL) for washing. The reaction mixture was stirred at reflux for 21 h. The reaction mixture was cooled, diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid. The solid was washed with ether (5 mL) and the supernatant liquid decanted off. The remaining solid was dissolved in 5M NaOH solution (5 mL), extracted with EtOAc (2 x 5 mL) and the organic layer dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL) and the organic solution dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The desired product **2.82** was isolated from the acidic extraction as a white solid (0.012 g, 8%).

M.P: 190 – 193 °C

IR υ_{max}/cm^{-1}: 3338, 1656, 1610

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.18 (1 H, s, H15), 6.82 (1 H, d, *J*=2.5 Hz, H2), 6.62 (1 H, d, *J*=2.5 Hz, H6), 6.58 (1 H, d, *J*=2.5 Hz, H12), 6.54 (1 H, d, *J*=2.5 Hz, H10), 5.11 (1 H, d, *J*=12.0 Hz, H13), 4.82 (1 H, d, *J*=12.0 Hz, H13), 3.88 (3 H, s), 3.87 (3 H, s), 3.80 (3 H, s) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.5 (C14), 162.9 (C16), 162.5 (C), 160.7 (C), 158.3 (C), 137.2 (C), 136.0 (C), 130.9 (C), 111.2 (C12), 106.9 (C), 104.7 (C2), 100.4 (C10), 100.3 (C6), 96.4 (C), 70.3 (C13), 56.0 (C17), 55.6 (C16), 55.5 (C18) ppm

LRMS ES$^+$ *m/z* (%): 339 ([M+Na]$^+$, 100)

HRMS (ES): C$_{17}$H$_{16}$O$_6$Na [M+Na]$^+$ calculated 339.0839, found 339.0848.

8-Hydroxy-10-methoxy-7-oxo-5,7-dihydrobenzo[c,e]ozepine-2-carbonitrile (2.84).
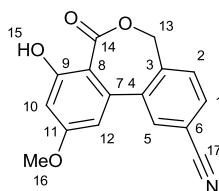
Two flasks were oven dried, cooled under vacuum and backfilled with argon. To one flask, pinacol ester **2.55** (0.150 g, 0.45 mmol), 2-bromo-4-cyanobenzylalcohol (0.095 g, 0.45 mmol) and potassium phosphate (0.191 g, 0.9 mmol) were added and the flask evacuated and backfilled with argon three times. Tris(dibenzylideneacetone)palladium (0.004 g, 0.0045 mmol) and SPhos (0.007 g, 0.018 mmol) were added to the second flask which was then evacuated and backfilled with argon three times. THF (2 x 1 mL) was added to each flask after which the Palladium complex was transferred to the reaction flask with additional THF (2 mL) for washing. The reaction mixture was stirred at reflux for 16 h. The reaction mixture was cooled, diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid. Ether (5 mL) was added and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL) and extracted with EtOAc (2 x 5 mL). The organic layer was dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL). The organic layer was dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The desired product **2.84** was isolated from the EtOAc extraction of the acidic solution as a white solid (0.008 g, 6%).

M.P: 221 – 223 °C

IR υ_{max}/cm^{-1}: 1659, 1618, 1569, 1380, 1225, 1164

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.55 (1 H, br. s, H15), 7.83 (1 H, d, *J*=1.1 Hz, H5), 7.72 (1 H, dd, *J*=7.7, 1.5 Hz, H1), 7.55 (1 H, d, *J*=7.7 Hz, H2), 6.64 (1 H, d, *J*=2.2 Hz, H12), 6.59 (1 H, d, *J*=2.2 Hz, H10), 5.16 (1 H, br. s, H13), 5.02 (1 H, br. s, H13), 3.90 (3 H, s, H16) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 164.3 (C14), 163.8 (C16), 145.7 (C), 140.5 (C), 138.6 (C), 138.5 (C), 132.8 (C5), 132.2 (C1), 128.9 (C2), 117.8 (C), 114.2 (C), 109.6 (C), 105.6 (C12), 102.0 (C10), 68.8 (C13), 55.8 (C16) ppm

LRMS ES+ *m/z* (%): 585 ([2M+Na]+, 100)

HRMS (ES): $C_{17}H_{15}NO_5Na$ [M+Na+MeOH]+ calculated 336.0842, found 336.0846.

9-Fluoro-4-hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.86).
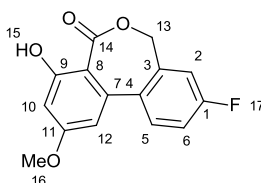
Two flasks were oven dried, cooled under vacuum and backfilled with argon. To one flask, pinacol ester **2.55** (0.150 g, 0.45 mmol), 2-bromo-5-fluorobenzylalcohol (0.092 g, 0.45 mmol) and potassium phosphate (0.191 g, 0.9 mmol) were added and the flask evacuated and backfilled with argon three times. Tris(dibenzylideneacetone)palladium (0.004 g, 0.0045 mmol) and SPhos (0.007 g, 0.018 mmol) were added to the second flask which was then evacuated and backfilled with argon three times. THF (2 x 1 mL) was added to each flask after which the Palladium complex was transferred to the reaction flask with additional THF (2 mL) for washing. The reaction mixture was stirred at reflux for 16 h after which it was diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO_2, 10% EtOAc in hexane) yielded an off white solid. Ether (5 mL) was added and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL), extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over $MgSO_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over $MgSO_4$, filtered and the solvent removed *in vacuo*. The product **2.86** was isolated from the EtOAc extraction of the acidic solution as a white solid (0.012 g, 10%).

M.P: 167 – 169 °C

IR υ_{max}/cm-1: 1663, 1607, 1569, 1504, 1274

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.61 (1 H, s, H15), 7.54 (1 H, dd, *J*=8.8, 5.3 Hz, H5), 7.21 (1 H, td, *J*=8.5, 3.0 Hz, H6), 7.15 (1 H, dd, *J*=8.0, 2.5 Hz, H2), 6.61 (1 H, d, *J*=2.5 Hz, H12), 6.59 (1 H, d, *J*=2.5 Hz, H10), 5.15 (1 H, br. s, H13), 4.93 (1 H, br. s, H13), 3.90 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.4 (C14), 164.1 (C11), 164.0 (C), 163.9 (C1, d, *J*=250.8), 140.0 (C), 136.0 (C3, d, *J*=7.8 Hz), 135.4 (C4, d, *J*=2.9 Hz), 131.3 (C5, d, *J*=8.7 Hz), 117.2 (C6, d, *J*=21.4 Hz), 115.0 (C2, d, *J*=22.4 Hz), 109.3 (C12), 105.9 (C), 101.1 (C10), 69.1 (C13), 55.7 (C16) ppm

^{19}F NMR (282 MHz, CDCl$_3$): δ -112.17 (1 F, s, F17) ppm

LRMS ES$^+$ *m/z* (%): 329 ([M+Na+MeOH]$^+$, 96)

HRMS (ES): C$_{15}$H$_{11}$O$_4$FNa [M+Na]$^+$ calculated 297.0534, found 297.0539.

4-Hydroxy-2-methoxy-9-(trifluoromethyl)dibenzo[c,e]oxepin-5(7H)-one (2.87).
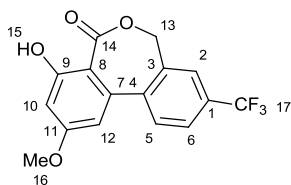
Two flasks were oven dried, cooled under vacuum and backfilled with argon. To one flask, pinacol ester **2.55** (0.150 g, 0.45 mmol), 2-bromo-5-(trifluoromethyl)benzylalcohol (0.115 g, 0.45 mmol) and potassium phosphate (0.191 g, 0.9 mmol) were added and the flask evacuated and backfilled with argon three times. Tris(dibenzylideneacetone)palladium (0.004 g, 0.0045 mmol) and SPhos (0.007 g, 0.018 mmol) were added to the second flask which was then evacuated and backfilled with argon three times. THF (2 x 1 mL) was added to each flask after which the Palladium complex was transferred to the reaction flask with additional THF (2 mL) for washing. The reaction mixture was stirred at reflux for 16 h after which it was diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid. Ether (5 mL) was added and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL), extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic

layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over $MgSO_4$, filtered and the solvent removed *in vacuo*. The product **2.87** was isolated from the EtOAc extraction of the acidic solution as a white solid (0.010 g, 7%).

M.P: 134 – 136 °C

IR υ_{max}/cm^{-1}: 1653, 1614, 1569, 1335

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.63 (1 H, s, H15), 7.78 (1 H, dd, *J*=8.0, 1.0 Hz, H6), 7.66 – 7.72 (2 H, m, H2, H5), 6.67 (1 H, d, *J*=2.5 Hz, H12), 6.65 (1 H, d, *J*=2.5 Hz, H10), 5.22 (1 H, br. s, H13), 5.05 (1 H, br. s, H13), 3.92 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.1 (C14), 164.2 (C), 164.1 (C), 142.8 (C), 139.4 (C), 134.8 (C), 131.0 (C, q, *J*=33.0 Hz), 129.9 (C), 126.9 (C, q, *J*=3.7 Hz), 125.0 (C, q, *J*=3.6 Hz), 109.8 (C), 106.0 (C), 101.9 (C), 69.1 (C13), 55.7 (C16) ppm

LRMS ES$^+$ *m/z* (%): 379 ([M+Na+MeOH]$^+$, 100)

HRMS (ES): $C_{16}H_{11}O_4F_3Na$ [M+Na]$^+$ calculated 347.0502, found 347.0499.

4-Hydroxy-2-methoxy-[1,3]dioxolo[4',5':4,5]benzo[1,2-c]benzo[e]oxepin-5(7H)-one (2.89).
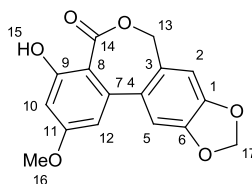
Two flasks were oven dried, cooled under vacuum and backfilled with argon. To one flask, pinacol ester **2.55** (0.150 g, 0.45 mmol), (6-bromo-1,3-benzoldioxol-5-yl)methanol (0.104 g, 0.45 mmol)and potassium phosphate (0.191 g, 0.90 mmol) were added and the flask evacuated and backfilled with argon three times. Tris(dibenzylideneacetone)palladium (0.004 g, 0.0045 mmol) and SPhos (0.007 g, 0.018 mmol) were added to the second flask which was then evacuated and backfilled with argon three times. THF (2 x 1 mL) was added to each flask after which the Palladium complex was transferred to the reaction

flask with additional THF (2 mL) for washing. The reaction mixture was stirred at reflux for 16 h after which it was diluted with EtOAc (10 mL) and filtered through a pad of silica gel. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid. Ether (5 mL) was added and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL), extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl, re-extracted with EtOAc (2 x 5 mL) and the combined organic extracts dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The product **2.89** was isolated from the EtOAc extraction of the acidic solution as a white solid (0.007 g, 5%).

M.P: 212 – 214 °C

IR υ_{max}/cm^{-1}: 2926, 1663, 1610, 1497, 1482, 1255

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.59 (1 H, s, H15), 7.01 (1 H, s, H5), 6.89 (1 H, s, H2), 6.58 (1 H, d, *J*=2.5 Hz, H12), 6.56 (1 H, d, *J*=2.5 Hz, H10), 6.06 (2 H, s, H17), 5.10 (1 H, d, *J*=12.0 Hz, H13), 4.83 (1 H, d, *J*=12.0 Hz, H13), 3.89 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.6 (C14), 163.9 (C), 163.6 (C), 149.2 (C), 148.1 (C), 141.0 (C), 133.7 (C), 128.2 (C), 109.5 (C6), 108.8 (C5), 108.2 (C12), 106.2 (C), 101.8 (C17), 100.9 (C10), 69.4 (C13), 55.6 (C16) ppm

LRMS ES$^+$ *m/z* (%): 623 ([2M+Na]$^+$, 100)

HRMS (ES): C$_{16}$H$_{12}$NaO$_6$ [M+Na]$^+$ calculated 323.0526, found 323.0529.

2-Bromo-3,4-di-*tert*-butoxycarbonyloxy-5-methoxybenzaldehyde (2.98).
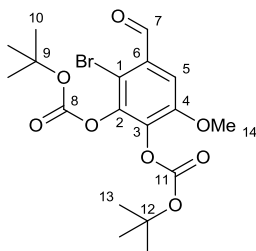
To a stirred solution of di-*tert*-butyldicarbonate (0.524 g, 2.4 mmol) in DCM (10 mL), under argon, was added 2-bromo-3,4-dihydroxy-5-methoxybenzaldehyde **2.69** (0.296 g, 1.2 mmol) and 4-dimethylaminopyridine (0.015 g, 0.12 mmol). After 2 h the solvent was removed *in vacuo* to yield the desired product **2.98** as a white solid without further purification (1.017 g, 95%).

M.P: 106 – 108 °C

IR υ_{max}/cm^{-1}: 1773, 1686, 1595, 1482, 1376, 1327

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.29 (1 H, s, H7), 7.44 (1 H, s, H5), 3.90 (3 H, s, H14), 1.54 (9 H, s, H13), 1.52 (9 H, s, H10) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 190.0 (C7), 152.1 (1 C, Ar), 149.2 (C11), 149.0 (C8), 142.3 (C), 138.8 (C), 130.9 (C), 113.8 (C),109.4 (C5), 84.9 (C12), 84.6 (C9) 56.5 (C14), 27.4 (C13), 27.3 (C10) ppm

LRMS ES$^+$ *m/z* (%): 501, 503 ([M+Na+MeOH]$^+$, 100)

HRMS (ES): C$_{19}$H$_{27}$O$_9$BrNa [M+Na+MeOH]$^+$ calculated 501.0731, found 501.0739.

2-Bromo-3,4-di-*tert*butoxycarbonyloxy-5-methoxybenzyl alcohol (2.99).
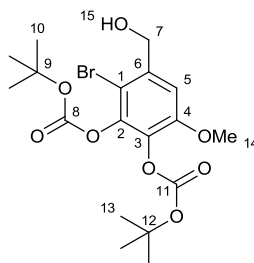
To a stirred solution of 2-bromo-3,4-di-*tert*-butoxycarbonyloxy-5-methoxybenzaldehyde **2.98** (0.537 g, 1.2 mmol) in ethanol (10 mL) at -50 °C was added sodium borohydride (0.023 g, 0.6 mmol). After 30 min water (20 mL) was added and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were washed with brine (15 mL), dried over MgSO$_4$ and the solvent removed *in vacuo* to yield the desired product **2.99** as a yellow solid without further purification (0.537 g, 99 %).

M.P: 92 – 94°C

IR υ_{max}/cm^{-1}: 2983, 1780, 1255

^{1}H NMR (400 MHz, CDCl$_3$): δ 7.11 (1H, s, H5), 4.72 (2 H, s, H7), 3.88 (3 H, 2, H14), 1.56 (9 H, s, H13), 1.54 (9 H, s, H10) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 151.8 (C4), 150.0 (C11), 149.7 (C8), 141.6 (C), 138.3 (C), 132.9 (C), 109.2 (C7), 106.8 (C), 84.5 (C9), 84.0 (C12), 64.6 (C7), 56.4 (C14), 27.6 (C10, C13) ppm

LRMS ES$^+$ *m/z* (%): 919, 921, 923 ([2M+Na]$^+$, 100)

HRMS (ES): C$_{18}$H$_{25}$O$_8$BrNa [M+Na]$^+$ calculated 471.0625, found 471.0634.

2-Bromo-3,4-diacetonitrile-5-methoxybenzaldehyde (2.101).
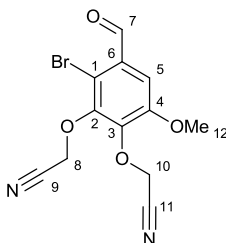
To a stirred solution of 2-bromo-3,4-dihydroxy-5-methoxybenzaldehyde **2.69** (0.988 g, 4 mmol) and K$_2$CO$_3$ (1.658 g, 12 mmol) in DMF (20 mL) was added iodoacetonitrile (0.98 mL, 12 mmol). The reaction mixture was heated to 60 °C for 16 h after which it was cooled to rt. Water (50 mL) was added and the product extracted with 1:1 toluene:EtOAc (3x25 mL). The combined organic extracts were washed with water (25 mL) and brine (25 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 40% EtOAc in hexane) afforded the desired product **2.101** as a white solid (0.410 g, 32%).

M.P: 129 – 131 °C

IR υ_{max}/cm^{-1}: 2986, 2937, 1682, 1588, 1478, 1436, 1387, 1372, 1334, 1285, 1168, 1123

150

¹H NMR (300 MHz, CDCl₃): δ 10.31 (1 H, s, H7), 7.45 (1 H, s, H5), 4.98 (2 H, s, H10), 4.89 (2 H, s, H8), 3.99 (3 H, s, H12) ppm

¹³C NMR (101 MHz, CDCl₃): δ 190.0 (C7), 152.4 (C4), 147.8 (C), 144.5 (C), 130.6 (C), 114.6 (C), 109.5 (C5), 58.2 (C8), 57.8 (C10), 56.6 (C12) ppm

LRMS ES⁺ *m/z* (%): 379, 381 ([M+Na+MeOH]⁺, 100)

HRMS (ES): C₁₂H₉O₄BrN₂Na [M+Na]⁺ calculated 346.9638, found 346.9644.

2-Bromo-3,4-diacetonitrile-5-methoxybenzyl alcohol (2.102).
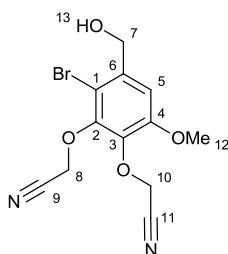
To an ice-cold, stirred solution of 2-bromo-3,4-diacetonitrile-5-methoxybenzaldehyde **2.101** (0.446 g, 1.37 mmol) in ethanol (10 mL) was added sodium borohydride (0.052 g, 1.37 mmol). After 30 min water (20 mL) was added to the flask and the product was extracted with EtOAc (2 x 20 mL). The combined organic extracts were washed with brine (20 mL), dried over MgSO₄ and the solvent removed *in vacuo*. The desired product **2.102** was isolated without further purification as a white solid (0.412 g, 92%).

M.P: 111 – 113 °C

IR υ_max/cm⁻¹: 3436, 3376, 1470, 1448, 1417, 1323

¹H NMR (300 MHz, CDCl₃): δ 7.04 (1 H, s, H5), 4.76 (2 H, s, H10), 4.75 (2 H, s, H8), 4.55 (2 H, s, H7), 3.82 (3 H, s, H12) ppm

¹³C NMR (75 MHz, CDCl₃): δ 152.2 (C4), 147.2 (C), 138.7 (C), 138.0 (C), 115.0 (C11), 114.9 (C9), 108.3 (C5), 106.5 (C), 63.3 (C7), 57.8 (C10), 57.6 (C8), 56.0 (C12) ppm

LRMS ES⁺ *m/z* (%): 349, 351 ([M+Na]⁺, 100)

HRMS (ES): $C_{12}H_{11}O_4BrN_2Na$ [M+Na]$^+$ calculated 348.9794, found 348.9799.

5-Bromo-4-formyl-2-methoxyphenyl acetate (2.104).
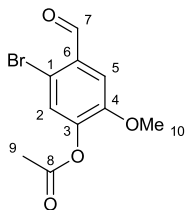
To a stirred suspension of 4-acetoxy-3-methoxybenzaldehyde (0.583 g, 3.0 mmol) and potassium bromide (1.214 g, 10.2 mmol) in water (22 mL) was added bromine (0.17 mL, 3.3 mmol) dropwise. The suspension was stirred at rt for 10 h. The resulting orange precipitate was collected by filtration and washed with water (3 x 10 mL). The precipitate was recrystallised from chloroform and the desired product **2.104** isolated as an orange crystalline solid (0.772 g, 94%).

M.P: 96 – 98 °C reported M.P: 109 – 110 °C[83]

IR υ_{max}/cm^{-1}: 3407, 1763, 1492, 1463

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.27 (1 H, s, H7), 7.51 (1 H, s, H2), 7.36 (1 H, s, H5), 3.88 (3 H, s, H10), 2.34 (3 H, s, H9) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 190.8 (C7), 167.8 (C8), 151.2 (C), 144.9 (C), 131.6 (C), 127.9 (C2), 117.8 (C), 112.1 (C5), 56.3 (C10), 20.5 (C9) ppm.

5-Bromo-4-(hydroxymethyl)-2-methoxyphenyl acetate (2.105).
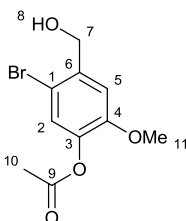
To an ice-cold, stirred solution of 5-bromo-4-formyl-2-methoxyphenyl acetate **2.104** (0.308 g, 1.13 mmol) in ethanol (10 mL) was added sodium borohydride (0.021 g, 0.56 mmol). After 0.5 h, the reaction mixture was warmed to rt and the water (20 mL) added. The product was extracted with EtOAc (3 x 15 mL) and the combined organic extracts dried over $MgSO_4$ and concentrated *in vacuo*. The crude was purified by column chromatography (SiO_2, 25% EtOAc in hexane) to yield the desired product **2.105** as a white solid (0.093 g, 30%).

M.P: 80 – 82 °C

IR υ_{max}/cm^{-1}: 3440, 1758, 1595, 1486, 1463

^{1}H NMR (300 MHz, CDCl$_3$): δ 7.22 (1 H, s, H2), 7.15 (1 H, s, H5), 4.68 (2 H, d, *J*=5.9 Hz, H7), 3.83 (3 H, s, H11), 2.31 (3 H, s, H10) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 168.8 (C9), 150.7 (C), 139.2 (C), 138.3 (C), 126.6 (C5), 112.3 (C2), 111.2 (C), 64.6 (C7), 56.1 (C11), 20.5 (C10) ppm.

The ^{1}H NMR spectra corresponded to the reported data.[84]

2-Bromo-4-hydroxy-5-methoxybenzaldehyde (2.106).
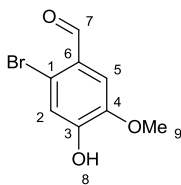
To a stirred solution of 4-acetoxy-3-methoxybenzaldehyde (1.165 g, 6.0 mmol) and KBr (2.428 g, 20.4 mmol) in water (44 mL) was added bromine (0.34 mL, 6.6 mmol) dropwise. The orange solution was stirred at rt overnight. The resulting orange precipitate was collected by filtration and washed with water. The crude material was suspended in 6M HCl (30 mL) and heated to 90 °C with stirring for 9 h. The resulting off-white precipitate was collected by filtration and dissolved in EtOAc (30 mL). The organic solution was washed with saturated sodium hydrogen carbonate solution (3 x 20 mL), dried over $MgSO_4$ and the solvent removed *in vacuo*. The desired product **2.106** was isolated without further purification as an off-white solid (0.815 g, 59% over two steps).

M.P: 142 – 144 °C reported M.P: 174 – 174 °C[85]

IR υ_{max}/cm^{-1}: 3107, 1663, 1595, 1557, 1508, 1417

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.04 (1 H, s, H7), 9.29 (1 H, br s, H8), 7.30 (1 H, s, H2), 7.05 (1 H, s, H5), 3.82 (3 H, s, H9) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 190.5 (C7), 153.1 (C), 147.4 (C), 125.3 (C), 120.3 (C), 119.4 (C2), 110.6 (C5), 55.9 (C9) ppm

LRMS ES$^-$ m/z (%): 229, 231 ([M-H]$^-$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[85]

2-(5-bromo-4-formyl-2-methoxyphenoxy)acetonitrile (2.107).
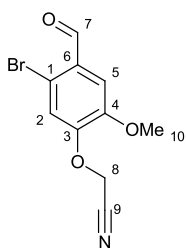
To a stirred solution of **2.106** (0.578 g, 2.5 mmol) and K$_2$CO$_3$ (0.691 g, 5.0 mmol) in DMF (10 mL) was added iodoacetonitrile (0.41 mL, 5.0 mmol). The reaction mixture was heated to 60 °C for 16 h after which it was cooled to rt. Water (10 mL) was added and the product extracted with 1:1 toluene:EtOAc (3 x 20 mL). The combined organic extracts were washed with water (20 mL) and brine (20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 30% EtOAc in hexane) afforded the desired product **2.107** as a white solid (0.386 g, 57 %)

M.P: 84 – 86 °C

IR υ_{max}/cm^{-1}: 1663, 1426, 1380, 1281

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.24 (1 H, s, H7), 7.50 (1 H, s, H2), 7.23 (1 H, s, H5), 4.92 (2 H, s, H8), 3.94 (3 H, s, H10) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 190.5 (C7), 150.6 (C), 149.9 (C), 129.3 (C), 120.0 (C2), 119.1 (C9), 114.1 (C), 111.9 (C5), 56.3 (C10), 54.9 (C8) ppm

LRMS ES$^+$ *m/z* (%): 324, 326 ([M+Na+MeOH]$^+$, 100)

HRMS (ES): C$_{11}$H$_{12}$O$_4$BrNNa [M+Na+MeOH]$^+$ calculated 323.9842, found 323.9850.

2-(5-Bromo-4-(hydroxymethyl)-2-methoxyphenoxy)acetonitrile (2.108).
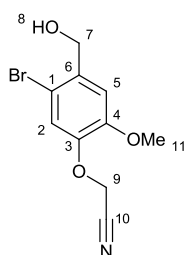
To an ice-cold, stirred solution of **2.107** (0.270 g, 1.0 mmol) in ethanol (10 mL) was added sodium borohydride (0.038 g, 1.0 mmol). After 0.5 h, the reaction mixture was warmed to rt and the water (10 mL) added. The product was extracted with EtOAc (3 x 15 mL) and the combined organic extracts dried over MgSO$_4$ and concentrated *in vacuo*. The crude was purified by column chromatography (SiO$_2$, 30% EtOAc in hexane) to yield the desired product **2.108** as a white solid (0.160 g, 59%).

M.P: 107 – 109 °C

IR υ$_{max}$/cm^{-1}: 3402, 2937, 1750, 1497, 1467, 1383

^{1}H NMR (400 MHz, CDCl$_3$): δ 7.10 (1 H, s, H2), 7.08 (1 H, s, H5), 4.73 (2 H, s, H9), 4.53 (2 H, s, H7), 3.79 (3 H, s, H11) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 154.5 (C), 149.2 (C), 141.2 (C), 125.9 (C), 119.5 (C), 116.5 (C), 115.3 (C), 67.8 (C), 60.3 (C), 60.2 (C) ppm

LRMS ES$^+$ *m/z* (%): 294, 296 ([M+K]$^+$, 99).

4-Hydroxy-2-methoxy-10-(trifluoromethyl)dibenzo[c,e]oxepin-5(7H)-one (2.112).
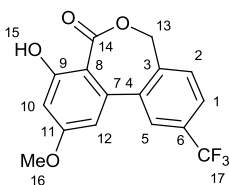
A solution of pinacol ester **2.55** (0.067 g, 0.2 mmol), 2-bromo-4-(trifluoromethyl)benzyl alcohol (0.051 g, 0.2 mmol), Pd(PPh$_3$)$_4$ (0.012 g, 0.01 mmol) and 2 M NaHCO$_3$ (0.3 mL, 0.6 mmol) in 1,4-dioxane (2 mL) was degassed for 10 min. The solution was then heated in a microwave to 130 °C for 10 min. The product was then diluted with EtOAc (10 mL), washed with brine (2 x 10 mL) and water (2 x 10 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded an off white solid which was confirmed as impure product. The product was washed with ether (5 mL) and the supernatant liquid decanted off. The remaining solid was dissolved in 5 M NaOH solution (5 mL). The basic solution was extracted with EtOAc (2 x 5 mL) and the organic layer dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The basic layer was acidified to pH 5 using 2 M HCl and re-extracted with EtOAc (2 x 5 mL). The organic layer was dried over MgSO$_4$, filtered and the solvent removed *in vacuo*. The desired product **2.112** was isolated from EtOAc extraction of the acidic layer as a white solid (0.020 g, 32%).

M.P: 163 – 165 °C

IR υ_{max}/cm^{-1}: 1664, 1612, 1570, 1376

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.64 (1 H, s, H15), 7.81 (1 H, s, H5), 7.71 (1 H, dd, *J*=7.8, 1.3 Hz, H1), 7.57 (1 H, d, *J*=8.0 Hz, H2), 6.65-6.67 (2 H, m, H10, H12), 5.21 (1 H, br. s, H13), 5.05 (1 H, br. s, H13), 3.92 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 173.3 (C14), 172.1 (C), 164.2 (C6, d, *J*=12.6 Hz), 140.2 (C), 139.4 (C), 137.5 (C), 132.7 (C17, t, *J*=33.0 Hz), 128.6 (C2), 126.2 (C5, q, *J*=3.9 Hz), 125.6 (C1, q, *J*=3.9 Hz), 122.3 (C), 109.7 (C12), 105.9 (C), 101.7 (C10), 69.0 (C13), 55.8 (C16) ppm

^{19}F {H} NMR (282 MHz, CDCl$_3$): δ -62.78 (3 F, s, F17) ppm

LRMS ES- *m/z* (%): 323 ([M-H]$^+$, 20)

HRMS (ES): $C_{16}H_{11}O_4F_3Na$ [M+Na]$^+$ calculated 347.0502, found 347.0501.

(2-Bromo-5-nitrophenyl)methanol (2.114).
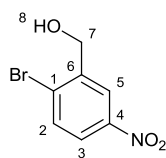
A stirred solution of 2-bromo-5-nitrobenzoic acid (0.246 g, 1 mmol) and sodium borohydride (0.113 g, 3 mmol) in tetrahydrofuran (5 mL) was cooled to 0 °C. Boron trifluoride diethyletherate (0.38 mL, 3 mmol) was added dropwise, the solution warmed to rt and stirred for 16 h. The reaction mixture was quenched with 1 M sodium hydroxide solution (10 mL) and stirred for 3 h. The solution was diluted with water (10 mL) and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were dried over MgSO$_4$ and the solvent removed *in vacuo* to yield the desired product **2.114** as a yellow solid without further purification (0.235 g, quant.).

M.P: Decomp. 75 °C

IR υ_{max}/cm^{-1}: 3232, 1535, 1342, 1021

^{1}H NMR (300 MHz, CDCl$_3$): δ 8.43 (1 H, d, *J*=2.7 Hz, H5), 8.02 (1 H, dd, *J*=8.7, 2.7 Hz, H3), 7.72 (1 H, d, *J*=8.7 Hz, H2), 4.83 (2 H, s, H7) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 147.5 (C), 142.0 (C), 133.3 (C), 128.7 (C), 123.3 (C), 122.9 (C), 64.0 (C7) ppm

No mass ion peak was detected in either negative or positive LRMS.

4-Hydroxy-2-methoxy-9-nitrodibenzo[c,e]oxepin-5(7H)-one (2.115).
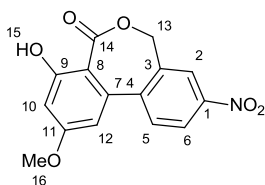
A solution of the pinacol ester **2.55** (0.067 g, 0.2 mmol), 2-bromo-5-nitrobenzyl alcohol **2.114** (0.139 g, 0.6 mmol), Pd(PPh$_3$)$_4$ (0.012 g, 0.01 mmol), 2M NaHCO$_3$ (0.3 mL, 0.6 mmol) and 1,4-dioxane (2 mL) was degassed for 10 min. The solution was heated in a microwave to 130 °C for 10 min after which it was diluted with EtOAc (10 mL). The organic solution was washed with brine (2 x 10 mL), water (2 x 10 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded the desired product **2.115** as a white solid (0.019 g, 31%).

M.P: 203 – 205 °C

IR υ_{max}/cm^{-1}: 3081, 1662, 1605, 1570, 1523

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.61 (1 H, s, H15), 8.37 (1 H, dd, *J*=8.5, 2.5 Hz, H6), 8.32 (1 H, d, *J*=2.5 Hz, H2), 7.74 (1 H, d, *J*=8.5 Hz, H5), 6.70 (1 H, d, *J*=2.5 Hz, H12), 6.67 (1 H, d, *J*=2.5 Hz, H10), 5.22 (1 H, br. s, H13), 5.11 (1 H, br. s, H13), 3.93 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 171.8 (C14), 164.3 (C11), 164.3 (C9), 147.6 (C), 145.6 (C), 138.5 (C), 135.5 (C), 130.5 (C5), 125.0 (C6), 123.1 (C2), 110.3 (C12), 105.8 (C), 102.4 (C10), 68.7 (C13), 55.8 (C16) ppm

LRMS ES$^+$ *m/z* (%): 356 ([M+Na+MeOH]$^+$, 100)

HRMS (ES): C$_{17}$H$_{16}$O$_5$Br$_2$Na [M+Na+MeOH]$^+$ calculated 356.0741, found 356.0742.

(3-Bromopyridin-4-yl)methanol (2.117).
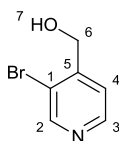
To a stirred solution of 3-bromo-4-pyridinecarboxaldehyde (0.186 g, 1 mmol) in ethanol (10 mL) was added sodium borohydride (0.038 g, 1 mmol). After 1 h water (10 mL) and EtOAc (30 mL) was added. The organic layer was separated, dried over $MgSO_4$ and the solvent removed *in vacuo*. The desired product **2.117** was isolated as a white solid without further purification (0.169 g, 90%).

^{1}H NMR (400 MHz, CDCl$_3$): δ 8.54 (1 H, s, H2), 8.44 (1 H, d, *J*=5.0 Hz, H3), 7.47 (1 H, d, *J*=5.0 Hz, H4), 4.69 (2 H, s, H6), 2.86 (1 H, br s, H7) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 151.4 (C2), 149.7 (C5), 148.8 (C3), 122.8 (C4), 120.2 (C1), 63.8 (C6) ppm

LRMS ES$^+$ *m/z* (%): 188, 190 ([M+H]$^+$, 88).

8-Hydroxy-10-methoxybenzo[5,6]oxepino[4,3-c]pyridin-7(5H)-one (2.118).
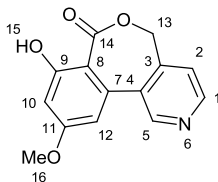
A solution of the pinacol ester **2.55** (0.067 g, 0.2 mmol), (3-bromopyridin-4-yl)methanol **2.117** (0.038 g, 0.2 mmol), Pd(PPh$_3$)$_4$ (0.012 g, 0.01 mmol) and 2 M NaHCO$_3$ (0.15 mL, 0.6 mmol) in 1,4-dioxane (2 mL) was degassed for 10 min. The reaction mixture was heated to 130°C for 10 min in a microwave after which it was poured into water (10 mL) and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were dried over MgSO$_4$ and the solvent removed *in vacuo*. The desired product **2.118** was isolated after column chromatography (SiO$_2$, 10% EtOAc in hexane) as a white solid (0.005 g, 10%).

M.P: 146 – 148 °C

IR υ$_{max}$/cm^{-1}: 3014, 1655, 1614, 1567

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.69 (1 H, s, H15), 8.83 (1 H, s, H5), 8.71 (1 H, d, *J*=5.0 Hz, H1), 7.37 (1 H, d, *J*=4.5 Hz, H2), 6.68 (1 H, d, *J*=2.5 Hz, H12), 6.67 (1 H, d, *J*=2.5 Hz, H10), 5.08 (2 H, br s, H13), 3.92 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.0 (C14), 164.6 (C), 164.3 (C), 150.1 (C5), 149.6 (C1), 141.8 (C), 134.7 (C), 130.8 (C), 121.7 (C2), 109.3 (C12), 106.0 (C), 101.9 (C10), 68.2 (C13), 55.8 (C16) ppm

LRMS ES$^+$ *m/z* 302 (%): ([M+Na+MeOH]$^+$, 13)

HRMS (ES): C$_{14}$H$_{11}$NO$_4$Na [M+Na]$^+$ calculated 280.0580, found 280.0579.

10-Fluoro-4-hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.120).
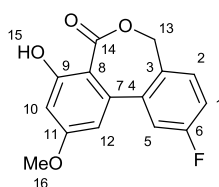
A solution of the pinacol ester **2.55** (0.067 g, 0.2 mmol), 2-bromo-4-fluorobenzyl alcohol (0.041 g, 0.2 mmol), Pd(PPh$_3$)$_4$ (0.012 g, 0.01 mmol) and 2 M NaHCO$_3$ (0.15 mL, 0.6 mmol) in 1,4-dioxane (2 mL) was degassed for 10 min. The reaction mixture was heated to 130°C for 10 min in a microwave after which it was poured into water (10 mL) and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were dried over MgSO$_4$ and the solvent removed *in vacuo*. The desired product **2.120** was isolated after column chromatography (SiO$_2$, 10% EtOAc in hexane) as a white solid (0.005 g, 10%).

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.63 (1 H, s, H15), 7.42 (1 H, dd, *J*=8.0, 5.5 Hz, H2), 7.27 (1 H, dd, *J*=9.5, 2.5 Hz, H5), 7.13 (1 H, td, *J*=8.3, 2.5 Hz, H1), 6.64 (1 H, d, *J*=2.5 Hz, H12), 6.62 (1 H, d, *J*=2.5 Hz, H10), 5.14 (1 H, br. s, H13), 4.97 (1 H, br. s, H13), 3.91 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.4 (C14), 164.1 (C), 163.9 (C), 163.5 (C6, d, *J*=248.8), 141.6 (C4, d, *J*=7.8 Hz), 139.8 (C), 130.2 (C3, d, *J*=2.9 Hz), 129.9 (C2, d, *J*=8.7 Hz), 116.3 (C5, d, *J*=23.3 Hz), 115.7 (C1, d, *J*=22.4), 109.3 (C12), 106.1 (C), 101.6 (C10), 68.9 (C13), 55.7 (C16) ppm

LRMS ES$^+$ *m/z* (%): 297 ([M+Na]$^+$, 100)

HRMS (ES): C$_{15}$H$_{11}$FO$_4$Na [M+Na]$^+$ calculated 297.0534, found 297.0538.

3,4-Bis(benzyloxy)-2-bromo-5-methoxybenzaldehyde (2.123).
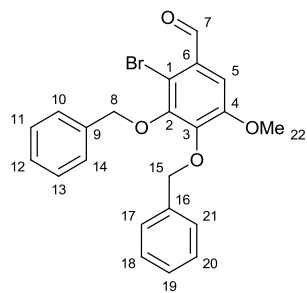
To a stirred suspension of sodium hydride (0.400 g, 10 mmol) in DMF (5 mL) at -78 °C was added 2-bromo-3,4-dihydroxy-5-methoxybenzaldehyde **2.69** (0.988 g, 4 mmol) in DMF (5 mL) and benzyl bromide (1.4 mL, 12 mmol). After 2 h the solution was poured into water (10 mL) and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were dried over $MgSO_4$ and the solvent removed *in vacuo*. The crude product was purified by column chromatography (SiO_2, 10% EtOAc in hexane) to yield the desired product **2.123** as a white solid (1.003 g, 59%).

M.P: 76 – 79 °C reported M.P: 82 – 86 °C[24]

IR υ_{max}/cm[-1]: 3064, 3032, 2943, 2866, 1688, 1576, 1451

^{1}H NMR (300 MHz, $CDCl_3$): δ 10.32 (1 H, s, H7), 7.31-7.52 (11 H, m), 5.16 (2 H, s, H8), 5.07 (2 H, s, H15), 3.92 (3 H, s, H22) ppm

HRMS (ES): $C_{22}H_{19}BrO_4Na$ [M+Na]+ calculated 449.0359, found 449.0356.

The ^{1}H NMR spectra corresponded to the reported data.[24]

(3,4-Bis(benzyloxy)-2-bromo-5-methoxyphenyl)methanol (2.121).
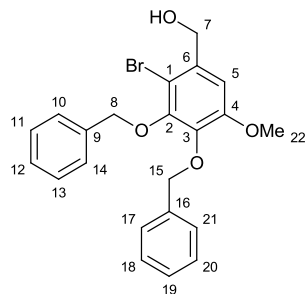
To a stirred solution of 3,4-bis(benzyloxy)-2-bromo-5-methoxybenzaldehyde **2.123** (0.855 g, 2 mmol) in THF (12 mL) at -78 °C under argon was added DIBAL-H (3.4 mL, 4 mmol) dropwise. After 2 h, 2M HCl (2 mL) was added dropwise and the resulting solution poured into water (20 mL). The product was extracted with EtOAc (3 x 30 mL), the combined organic extracts dried over $MgSO_4$ and the solvent removed *in vacuo*. The desired product **2.121** was isolated in quantitative yield as a white solid with no further purification required.

M.P: 72 – 74 °C reported M.P: 74 – 76 °C[24]

IR υ_{max}/cm^{-1}: 3397, 2936, 1569, 1454, 1415

^{1}H NMR (300 MHz, CDCl$_3$): δ 7.43-7.53 (4 H, m, H10,14,17,21), 7.31-7.40 (6 H, m, H11-13,18-20), 6.94 (1 H, s, H5), 5.06 (2 H, s, H8), 5.04 (2 H, s, H15), 4.75 (2 H, d, *J*=6.0 Hz, H7), 3.89 (3 H, s, H22) ppm

LRMS ES$^+$ *m/z* (%): 451, 453 ([M+Na]$^+$, 100).

The ^{1}H NMR spectra corresponded to the reported data.[24]

10,11-Bis(benzyloxy)-4-hydroxy-2,9-dimethoxydibenzo[c,e]oxepin-5(7H)-one (2.122).
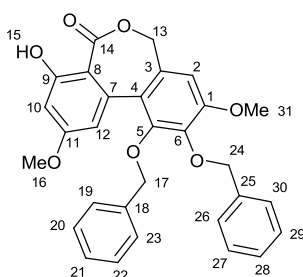
A mixture of dioxane (6 mL) and water (1 mL) was degassed for 10 min before being added to a flask containing pinacol ester **2.55** (0.434 g, 1.3 mmol), (3,4-Bis(benzyloxy)-2-bromo-5-methoxyphenyl)methanol **2.121** (0.429 g, 1 mmol), palladium acetate (0.007 g, 0.03 mmol), SPhos (0.025 g, 0.06 mmol) and caesium carbonate (0.977 g, 3 mmol). The solution was heated to 80 °C for 2 h. The reaction mixture was cooled to rt, NH$_4$Cl (2 mL) added and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts

were dried over $MgSO_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 10% EtOAc in hexane) yielded the desired product **2.122** as a white solid (0.251 g, 54%).

M.P: 112 – 114 °C reported M.P: 180 – 182 °C[24]

IR υ_{max}/cm^{-1}: 2941, 1734, 1657, 1611, 1565

^{1}H NMR (300 MHz, CDCl$_3$): δ 10.26 (1 H, s, H15), 7.50 (2 H, dd, *J*=7.7, 2.1 Hz, Ph), 7.32-7.41 (3 H, m, Ph), 7.12-7.25 (3 H, m, Ph), 6.96 (2 H, d, *J*=7.5 Hz, Ph), 6.84 (1 H, d, *J*=2.6 Hz, H12), 6.77 (1 H, s, H2), 6.54 (1 H, d, *J*=2.6 Hz, H10), 5.20 (1 H, d, *J*=10.5 Hz, CH$_2$), 5.02-5.11 (2 H, m, CH$_2$), 4.85 (1 H, d, *J*=10.9 Hz, CH$_2$), 4.80 (1 H, d, *J*=12.1 Hz, CH$_2$), 4.59 (1 H, d, *J*=10.9 Hz, CH$_2$), 3.94 (3 H, s, H16), 3.79 (3 H, s, H31) ppm

LRMS ES$^+$ *m/z* (%): 521 ([M+Na]$^+$, 100)

HRMS (ES): $C_{30}H_{26}O_7Na$ [M+Na]$^+$ calculated 521.1571, found 521.1570.

The ^{1}H NMR spectra corresponded to the reported data.[24]

Ulocladol A (1.1)
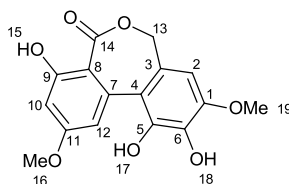
An oven-dried flask was charged with protected ulocladol A **2.122** (0.199 g, 0.4 mmol), Pd/C (10–15 mol%) (0.006 g, 0.06 mmol) and methanol (8 mL). The flask was evacuated and back-filled with hydrogen. The reaction mixture was stirred under a hydrogen atmosphere for 2 min after which it was filtered through a pad of celite and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 1% MeOH in DCM) failed to remove all impurities. A second attempt at purification through column chromatography (SiO$_2$, 10% EtOAc in hexane) also failed to remove all impurities. Recrystallisation of the product from CDCl$_3$ furnished the desired product as a white solid (0.018 g, 14%).

M.P: Decomposed at 170 °C reported M.P: 180 – 182 °C[24]

IR υ_{max}/cm[-1]: 3412, 1650, 1614, 1567, 1514

^{1}H NMR (400 MHz, CDCl$_3$): δ 10.31 (1 H, s, H15), 6.92 (1 H, d, *J*=2.5 Hz, H12), 6.58 (1 H, s, H2), 6.56 (1 H, d, *J*=2.5 Hz, H10), 5.78 (1 H, br s, H17), 5.69 (1 H, br s, H18), 5.12 (1 H, d, *J*=12.0 Hz, H13), 4.80 (1 H, d, *J*=12.0 Hz, H13), 3.95 (3 H, s, H16), 3.87 (3 H, s, H19) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 172.5 (C14), 163.2 (C11), 163.0 (C9), 146.3 (C2), 142.0 (C7), 135.9 (C5), 133.8 (C6), 127.1 (C3), 118.8 (C4), 110.2 (C12), 106.9 (C8), 103.1 (C1), 100.8 (C10), 70.3 (C13), 56.3 (C19), 55.5 (C16) ppm

LRMS ES+ *m/z* (%): 341 ([M+Na]+,100)

HRMS (ES): C$_{17}$H$_{18}$O$_8$Na [M+Na+MeOH]+ calculated 373.0530, found 373.0523.

The ^{1}H NMR spectra corresponded to the reported data.[24]

2,2-Dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate (2.126).
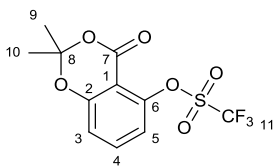
To a biphasic solution of 5-Hydroxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (1.503 g, 7.7 mmol) in toluene (36 mL) and aqueous LiOH (0.556 g, 23.2 mmol in 11 mL H$_2$O) at 0 °C, was added trifluoromethanesulfonic anhydride (6 mL, 35.4 mmol) dropwise. The reaction mixture was stirred at 0 °C for 3 h after which water (30 mL) was added. The product was extracted with EtOAc (3 x 20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Column chromatography (SiO$_2$, 20% EtOAc in hexane) afforded **2.126** as a white, crystalline solid (1.286 g, 51%).

M.P: 102 – 104 °C reported M.P: 114 – 115 °C[86]

^{1}H NMR (300 MHz, CDCl$_3$): δ 7.61 (1 H, t, *J*=8.3 Hz, H4), 7.06 (1 H, dd, *J*=8.7, 1.1 Hz, H5), 7.01 (1 H, d, *J*=7.9 Hz, H3), 1.77 (6 H, s, H9, 10) ppm

Rep. Data: ^{1}H NMR[87, 88] (400 MHz, CDCl3): δ 7.60 (1 H, dd, *J*=8.4, 8.4 Hz), 7.05 (1 H, d, *J*=8.4 Hz), 7.00 (1 H, d, *J*=8.4 Hz), 1.76 (6 H, s) ppm.

The ^{1}H NMR spectra corresponded to the reported data.[87, 88]

2,2-Dimethyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-4H-benzo[d][1,3]dioxin-4-one (2.127).
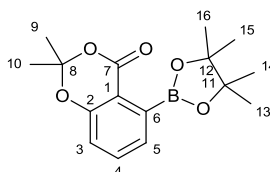
A solution of 2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate **2.126** (1.280 g, 3.9 mmol), Pd(PPh$_3$)$_4$ (0.225 g, 0.2 mmol) and freshly distilled triethylamine (1.6 mL, 11.7 mmol) in THF (10 mL) was degassed under argon in a ultrasonication bath for 10 min. A 1M solution of 4,4,5,5-Tetramethyl-1,3,2-dioxaborolane in THF (11.7 mL, 11.7 mmol) was added dropwise over 5 min and the reaction mixture then heated to reflux for 4 h. The solvent was removed *in vacuo* to yield the crude product. Column chromatography (SiO$_2$, 15% EtOAc in hexane) afforded **2.127** as a white solid (0.167 g, 14%).

M.P: 103 – 105 °C

^{1}H NMR (400 MHz, CDCl$_3$): δ 7.50 (1H, dd, *J*=7.0, 7.0 Hz, H4), 7.14 (1 H, dd, *J*=7.5, 1.0 Hz, H5), 6.93 (1 H, dd, *J*=8.5, 1.0 Hz, H3), 1.73 (6 H, s, H9, 10), 1.42 (12 H, s, H13-16) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 162.0 (C7), 155.6 (C2), 135.5 (C4), 126.3 (C5), 117.6 (C3), 116.1 (C1), 106.2 (C8), 84.4 (C11, 12), 25.8 (C9, 10), 24.7 (C13-15) ppm.

10,11-Bis(benzyloxy)-4-hydroxy-9-methoxydibenzo[c,e]oxepin-5(7H)-one (2.128).
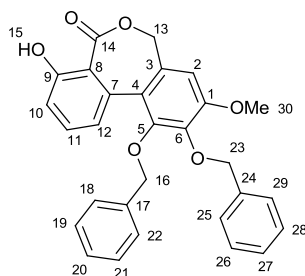
A mixture of dioxane (3 mL) and water (0.5 mL) was degassed for 10 min before being added to a flask containing pinacol ester **2.127** (0.115 g, 0.34 mmol), (3,4-Bis(benzyloxy)-2-bromo-5-methoxyphenyl)methanol **2.121** (0.124 g, 0.29 mmol), palladium acetate (0.002 g, 0.01 mmol), SPhos (0.007 g, 0.02 mmol) and caesium carbonate (0.283 g, 0.87 mmol). The solution was heated to 80 °C for 2 h. The reaction mixture was cooled to rt, NH_4Cl (1 mL) added and the product extracted with EtOAc (3 x 5 mL). The combined organic extracts were dried over $MgSO_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO_2, 10% EtOAc in hexane) yielded the desired product **2.128** as a white solid (0.070 g, 51%).

^{1}H NMR (400 MHz, CDCl[3]): δ 9.63 (1 H, s, H15), 7.43 (2 H, dd, *J*=8.0, 1.5 Hz, Ph), 7.22-7.37 (4 H, m, Ph), 7.11-7.21 (2 H, m, Ph), 7.06 (2 H, t, *J*=7.3 Hz, H11), 6.95 (1 H, dd, *J*=8.0 Hz, H10), 6.78-6.87 (2 H, m, H12, Ph), 6.70 (1 H, s, H2), 5.12 (1 H, d, *J*=11.0 Hz, H16), 5.00 (1 H, d, *J*=11.0 Hz, H16), 4.94 (1 H, d, *J*=12.0 Hz, H13), 4.78 (1 H, d, *J*=11.0 Hz, H23), 4.73 (1 H, d, *J*=12.0 Hz, H13), 4.55 (1 H, d, *J*=11.0 Hz, H23), 3.85 (3 H, s, H30) ppm

^{13}C NMR (101 MHz, CDCl[3]): δ 171.7 (C14), 159.7 (C9), 153.7 (C1), 150.8 (C), 143.1 (C6), 137.2 (C), 135.9 (C), 134.7 (C), 132.8 (C11), 130.7 (C), 128.6 (CH), 128.4 (CH), 128.3 (CH), 128.04 (CH), 128.02 (CH), 128.00 (CH), 126.1 (C3), 123.7 (C12), 116.4 (C10), 113.6 (C), 107.7 (C2), 75.8 (C23), 75.4 (C16), 70.1 (C13), 56.1 (C30) ppm

LRMS ES+ *m/z* (%): 491 ([M+Na]+, 100)

HRMS (ES): $C_{29}H_{24}O_6Na$ [M+Na]+ calculated 491.1465, found 491.1473.

11-(benzyloxy)-4,10-dihydroxy-9-methoxydibenzo[c,e]oxepin-5(7H)-one (2.129).
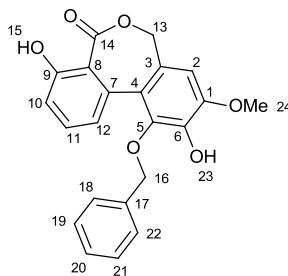
An oven-dried flask was charged with 11-(benzyloxy)-4,10-dihydroxy-9-methoxydibenzo[c,e]oxepin-5(7H)-one **2.128** (0.030 g, 0.06 mmol), Pd/C (10–15 mol%) (0.001 g, 0.01 mmol) and ethanol (3 mL). The flask was evacuated and back-filled with hydrogen. The reaction mixture was stirred under a hydrogen atmosphere for 2 min after which it was filtered through a pad of celite and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 10% EtOAc in hexane) furnished the desired product as a white solid (0.011 g, 48%).

M.P: 104 – 106 °C

IR υ_{max}/cm^{-1}: 3413, 2936, 1666, 1607, 1567, 1500

^{1}H NMR (400 MHz, CDCl$_3$): δ 9.70 (1 H, s, H15), 7.46 (1 H, t, *J*=7.8 Hz, H11), 7.34 (1 H, dd, *J*=8.1, 1.5 Hz, H12), 7.26-7.30 (1 H, m, H20), 7.18-7.26 (2 H, m, H19, 21), 7.01-7.07 (3 H, m, H10, 18, 22), 6.77 (1 H, s, H2), 5.82 (1 H, s, H23), 5.06 (1 H, d, *J*=12.1 Hz, H13), 4.82 (1 H, d, *J*=12.1 Hz, H13), 4.66 (1 H, d, *J*=11.1 Hz, H16), 4.60 (1 H, d, *J*=11.1 Hz, H16), 3.98 (3 H, s, H24) ppm

^{13}C NMR (101 MHz, CDCl$_3$): δ 171.9 (C14), 159.8 (C9), 147.3 (C1), 143.8 (C5), 140.4 (C6), 135.8 (C17), 134.7 (C4), 133.2 (C11), 128.6 (C19, 21), 128.4 (C20), 128.35 (C18, 22), 126.5 (C7), 125.3 (C3), 123.2 (C12), 116.8 (C10), 113.8 (C8), 106.9 (C2), 75.4 (C16), 70.3 (C13), 56.4 (C24) ppm

HRMS (ES): C$_{22}$H$_{18}$O$_6$Na [M+Na]$^+$ calculated 401.0996, found 401.1004.

4-Bromo-5-methylbenzene-1,3-diol (3.4).
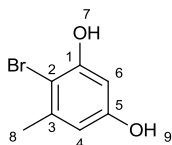
To a stirred solution of orcinol (0.993 g, 8.0 mmol) in DCM (30 mL) and MeOH (20 mL) was added a solution of tetrabutyl ammoniu tribromide (3.857 g, 8.0 mmol) in DCM (30 mL) and MeOH (20 mL) dropwise. The solution was stirred at rt for 16 h. The reaction mixture was concentrated and poured into water (20 mL). The product was extracted with DCM (2 x 20 mL), the combined organic extracts dried over $MgSO_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO_2, 20% EtOAc in hexane) yielded the desired product **3.4** as a white solid (0.724 g, 45%).

M.P: 134 – 136 °C reported M.P: 132 – 135 °C[89]

IR υ_{max}/cm^{-1}: 3353, 1586, 1459, 1323

^{1}H NMR (300 MHz, $(CD_3)_2CO$): δ 8.32 (2 H, br. s, H7, 9), 6.39 (1 H, d, *J*=2.9 Hz, H6), 6.36 (1 H, d, *J*=2.9 Hz, H4), 2.27 (3 H, s, H8) ppm

^{13}C NMR (75 MHz, $(CD_3)_2CO$): δ 158.1 (C5), 155.6 (C1), 140.2 (C3), 110.3 (C4), 102.8 (C2), 102.0 (C6), 23.4 (C8) ppm

LRMS ES$^-$ *m/z* (%): 201, 203 ([M-H]$^-$, 100).

4-Bromo-5-methyl-1,3-phenylene diacetate (3.5).
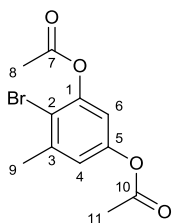
To a stirred solution of 4-bromo-5-methylbenzene-1,3-diol **3.4** (0.203 g, 1.0 mmol), acetic anhydride (0.24 mL, 2.5 mmol) and 4-dimethylaminopyridine (0.005 g, 0.04 mmol) in DCM (5 mL) was added triethylamine (0.35 mL, 2.5 mmol). The reaction mixture was stirred at

rt for 16 h, after which it was diluted with DCM (10 mL). The organic solution was washed with saturated sodium carbonate solution (10 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Purification by column chromatography (SiO$_2$, 10% EtOAc in hexane) furnished the desired product **3.5** as a white solid (0.242 g, 84%).

M.P: 52 – 54 °C

IR υ_{max}/cm^{-1}: 1774, 1369, 1189, 1136

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.93 (1 H, d, *J*=3.0 Hz, H4), 6.82 (1 H, d, *J*=3.0 Hz, H6), 2.42 (3 H, s, H9), 2.33 (3 H, s, H11), 2.25 (3 H, s, H8) ppm

^{13}C NMR (75 MHz, (CDCl$_3$): δ 168.8 (C10), 168.3 (C7), 149.5 (C), 148.6 (C), 140.5 (C), 121.3 (C), 115.6 (C6), 114.7 (C4), 23.3 (C9), 21.2 (C11), 20.8 (C8) ppm

LRMS ES$^+$ *m/z* (%): 309, 311 ([M+Na]$^+$, 79)

HRMS (ES): C$_{11}$H$_{11}$O$_4$BrNa [M+Na]$^+$ calculated 308.9733, found 310.9724.

2,2'-((4-Bromo-5-methyl-1,3-phenylene)bis(oxy))diacetonitrile (3.6).
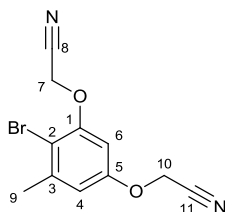
To a stirred solution of 4-bromo-5-methyl-1,3-phenylene diacetate **3.5** (0.506 g, 2.5 mmol) and K$_2$CO$_3$ (1.037 g, 7.5 mmol) in DMF (10 mL) was added iodoacetonitrile (0.61 mL, 7.5 mmol). The reaction mixture was heated to 60 °C for 16 h after which it was cooled to rt. Water (10 mL) was added and the product extracted with 1:1 toluene:EtOAc (3 x 20 mL). The combined organic extracts were washed with water (20 mL) and brine (20 mL), dried over MgSO$_4$ and the solvent removed *in vacuo*. Purification by column chromatography (40% EtOAc in hexane) afforded the desired product **3.6** as a white solid (0.240 g, 34 %)

M.P: 104 – 106 °C

IR υ_{max}/cm^{-1}: 2929, 1582, 1470, 1446

^{1}H NMR (300 MHz, CDCl$_3$): δ 6.60 (1 H, d, J=3.0 Hz, H6), 6.51 (1 H, d, J=3.0 Hz, H4), 4.79 (2 H, s, H10), 4.74 (2 H, s, H7), 2.35 (3 H, s, H9) ppm

^{13}C NMR (75 MHz, CDCl$_3$): δ 155.9 (C), 153.9 (C), 141.5 (C), 114.6 (C8), 114.5 (C11), 111.0 (C4), 109.0 (C), 101.3 (C6), 54.9 (C7), 53.8 (C11), 23.6 (C9) ppm

LRMS ES$^+$ m/z (%): 303, 305 ([M+Na]$^+$, 100)

HRMS (ES): C$_{11}$H$_9$N$_2$O$_2$BrNa [M+Na]$^+$ calculated 302.9740, found 302.9736.

Alternariol-9-methyl ether (1.2).
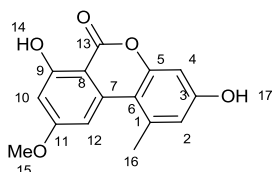
A degassed solution of 1,4-dioxane (2.4 mL) and water (0.4 mL) was added to a mixture of the pinacol ester **2.55** (0.174 g, 0.52 mmol), phenol **3.4** (0.081 g, 0.4 mmol), Pd (OAc)$_2$ (0.003 g, 0.012 mmol), SPhos (0.010 g, 0.024 mmol) and Cs$_2$CO$_3$ (0.391 g, 1.2 mmol) under argon. The stirred solution was heated in a microwave at 130 °C for 5 min. The reaction mixture was diluted with NH$_4$Cl (10 mL) and the product extracted with EtOAc (3 x 10 mL). The combined organic extracts were dried over MgSO$_4$ and the solvent removed *in vacuo*. Recrystallisation of the crude material from methanol yielded the desired product **1.2** as a purple crystalline solid (0.048 g, 44%).

M.P: >246 °C reported M.P: 277 – 279 °C[75]

IR υ_{max}/cm^{-1}: 3375, 1651, 1616, 1592, 1568

^{1}H NMR (400 MHz, (CD$_3$)$_2$SO): δ 11.82 (1 H, br. s, H17), 10.36 (1 H, s, H14), 7.21 (1 H, d, J=2.4 Hz, H12), 6.72 (1 H, d, J=2.0 Hz, H2), 6.64 (1 H, d, J=2.4 Hz, H10), 6.61 (1 H, d, J=2.0 Hz, H4), 3.91 (3 H, s, H15), 2.73 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, (CD$_3$)$_2$SO): δ 166.2 (C), 164.7 (C), 164.1 (C), 158.6 (C), 152.6 (C), 138.4 (C), 137.8 (C), 117.6 (CH), 108.8 (C), 103.4 (CH), 101.6 (CH) 99.2 (CH), 98.5 (C), 55.8 (C15), 25.0 (C16) ppm

LRMS ES$^+$ *m/z* (%): 327 ([M+Na+MeOH]$^+$, 100)

HRMS (ES): C$_{15}$H$_{12}$O$_5$Na [M+Na]$^+$ calculated 295.0577, found 295.0582.

The ^{1}H NMR spectra corresponded to the reported data.[75]

Alternariol (1.3).
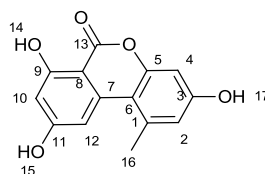
To a stirred solution of alternariol 9-methylether **1.2** (0.030 g, 0.1 mmol) in DCM (3 mL), under argon, was added a 1 M solution of BBr$_3$ in DCM (0.44 mL, 4.4 mmol). The solution was stirred at rt for 16 h after which MeOH (15 mL) was added dropwise. The solvent was removed *in vacuo* and the resulting crude material purified by column chromatography (2% MeOH in DCM) to yield the desired product **1.3** as a white solid (0.016 g, 0.063 mmol).

IR υ$_{max}$/cm^{-1}: 3438, 3162, 1659, 1608, 1416

^{1}H NMR (400 MHz, CD$_3$OD): δ 7.22 (1H, d, *J*=2.0 Hz, H12), 6.67 (1 H, d, *J*=2.5 Hz, H2), 6.58 (1 H, d, *J*=2.5 Hz, H4), 6.35 (1 H, d, *J*=2.0 Hz, H10), 2.73 (3 H, s, H16) ppm

^{13}C NMR (101 MHz, CD$_3$OD): δ 167.1 (C), 167.0 (C), 166 (C), 160.0 (C), 154.6 (C), 140.2 (C), 140.0 (C), 118.7 (C2), 111.1 (C), 105.7 (C12), 102.9 (C4), 102.1 (C10), 99.2 (C), 26.0 (C16) ppm

LRMS ES$^+$ *m/z* (%): 313 ([M+Na+MeOH]$^+$, 60%)

HRMS (ES): C$_{14}$H$_{11}$O$_5$ [M+H]$^+$ calculated 259.0601, found 259.0597.

The ^{1}H NMR spectra corresponded to the reported data.[30]

8 Appendix

8.1 X-Ray Crystal Structure Data for 2.115
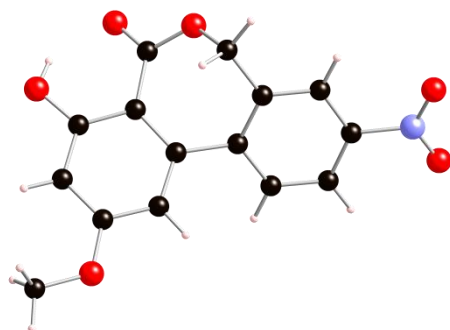
Table 1. Crystal data and structure refinement details.

Identification code	**2009sot1058 (LS5529/90)**
Empirical formula	$C_{15}H_{11}NO_6$
Formula weight	301.25
Temperature	120(2) K
Wavelength	0.71073 Å
Crystal system	Monoclinic
Space group	$P2_1/c$
Unit cell dimensions	a = 4.0312(3) Å
	b = 41.493(4) Å β = 103.619(4)°
	c = 7.9266(8) Å
Volume	1288.6(2) Å^3
Z	4
Density (calculated)	1.553 Mg / m^3
Absorption coefficient	0.122 mm^{-1}
$F(000)$	624
Crystal	Lath; Colourless
Crystal size	$0.10 \times 0.06 \times 0.01$ mm^3
θ range for data collection	2.95 – 25.03°
Index ranges	$-4 \le h \le 4, -49 \le k \le 49, -9 \le l \le 9$
Reflections collected	7987
Independent reflections	2254 [R_{int} = 0.0816]
Completeness to θ = 25.03°	98.9 %
Absorption correction	Semi−empirical from equivalents
Max. and min. transmission	0.9988 and 0.9879
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	2254 / 0 / 201
Goodness-of-fit on F^2	1.088
Final R indices [$F^2 > 2\sigma(F^2)$]	$R1$ = 0.0974, $wR2$ = 0.1687
R indices (all data)	$R1$ = 0.1588, $wR2$ = 0.2011
Largest diff. peak and hole	0.361 and −0.353 e Å^{-3}

8.2 X-Ray Crystal Structure Data for 1.2
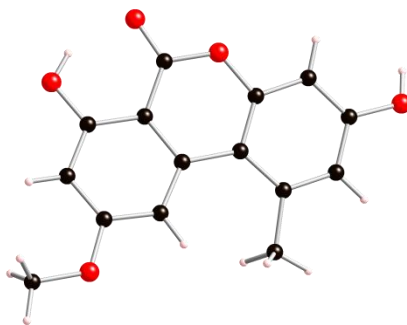
Table 1. Crystal data and structure refinement details.

Identification code	**2009sot1168 (LS5671/8)**
Empirical formula	$C_{15}H_{12}O_5$
Formula weight	272.25
Temperature	120(2) K
Wavelength	0.71073 Å
Crystal system	Orthorhombic
Space group	*Fdd*2
Unit cell dimensions	a = 31.7929(10) Å
	b = 39.3485(10) Å
	c = 3.76590(10) Å
Volume	4711.2(2) Å^3
Z	16
Density (calculated)	1.535 Mg / m^3
Absorption coefficient	0.116 mm^{-1}
F(000)	2272
Crystal	Rod; Colourless
Crystal size	$0.15 \times 0.03 \times 0.02$ mm^3
θ range for data collection	$3.30 - 27.48°$
Index ranges	$-39 \leq h \leq 40, -50 \leq k \leq 50, -4 \leq l \leq 4$
Reflections collected	15064
Independent reflections	1542 [R_{int} = 0.0745]
Completeness to θ = 27.48°	99.7 %
Absorption correction	Semi−empirical from equivalents
Max. and min. transmission	0.9977 and 0.9828
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	1542 / 1 / 185
Goodness-of-fit on F^2	1.090
Final *R* indices [$F^2 > 2\sigma(F^2)$]	*R1* = 0.0488, *wR2* = 0.0992
R indices (all data)	*R1* = 0.0615, *wR2* = 0.1055
Absolute structure parameter	Not reliably determined
Largest diff. peak and hole	0.243 and -0.259 e Å^{-3}

3,4,5-Trimethoxybenzyl benzoate (2.10)

Comment	LS4998/74	Date	02 Jun 2008 13:05:20
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.4951	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2008\JU0208LS1\10\PDATA\1\1r

JU0208LS1.010.001.1r.esp
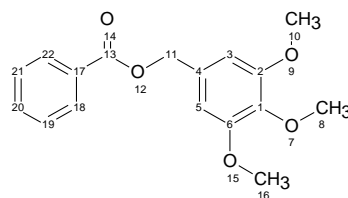
Formula $C_{17}H_{18}O_5$ **FW** 302.3218
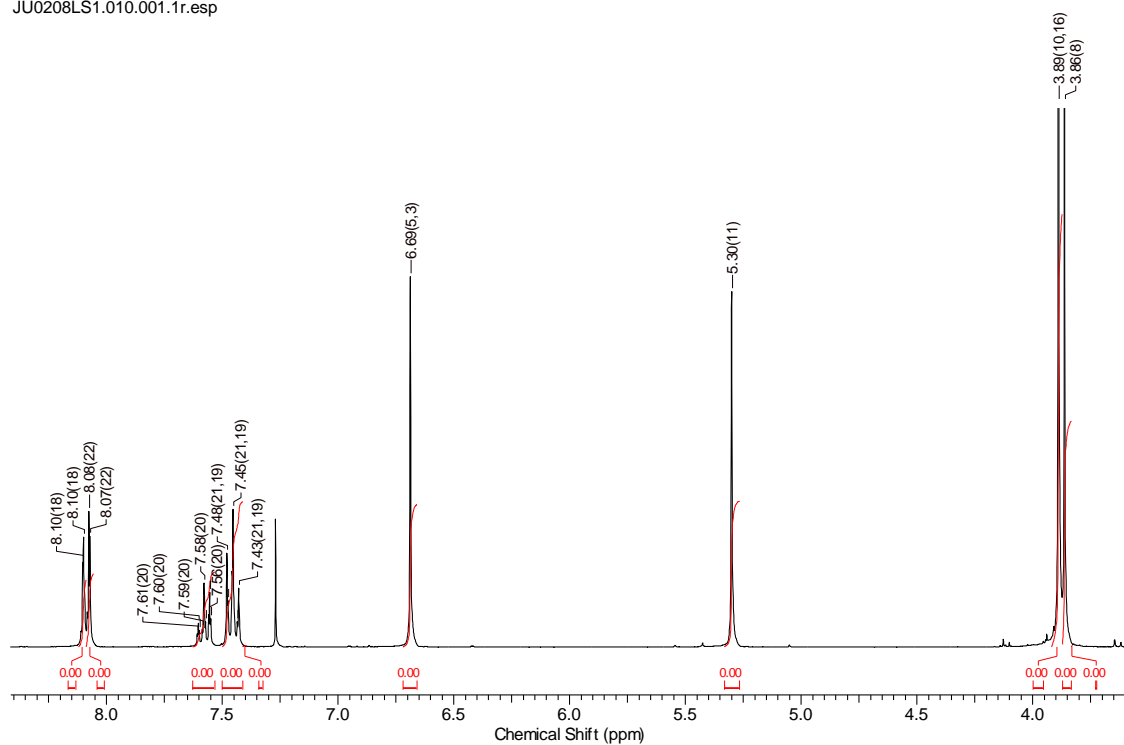
¹H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.86 (3 H, s) 3.89 (6 H, s) 5.30 (2 H, s) 6.69 (2 H, s) 7.41 - 7.49 (2 H, m) 7.54 - 7.62 (1 H, m) 8.09 (2 H, dd, *J*=8.4, 1.0 Hz)

2-Bromo-3,4,5-trimethoxybenzyl alcohol (2.14)

Comment	LS/4781/37 columned		
Date	14 Feb 2007 14:04:48		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8612	Temperature (degree C)	27.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2007\FE1407LS1\10\PDATA\1\1r

FE1407LS1.010.001.1r.esp
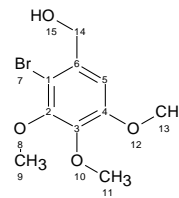
Formula $C_{10}H_{13}BrO_4$ FW   277.1118
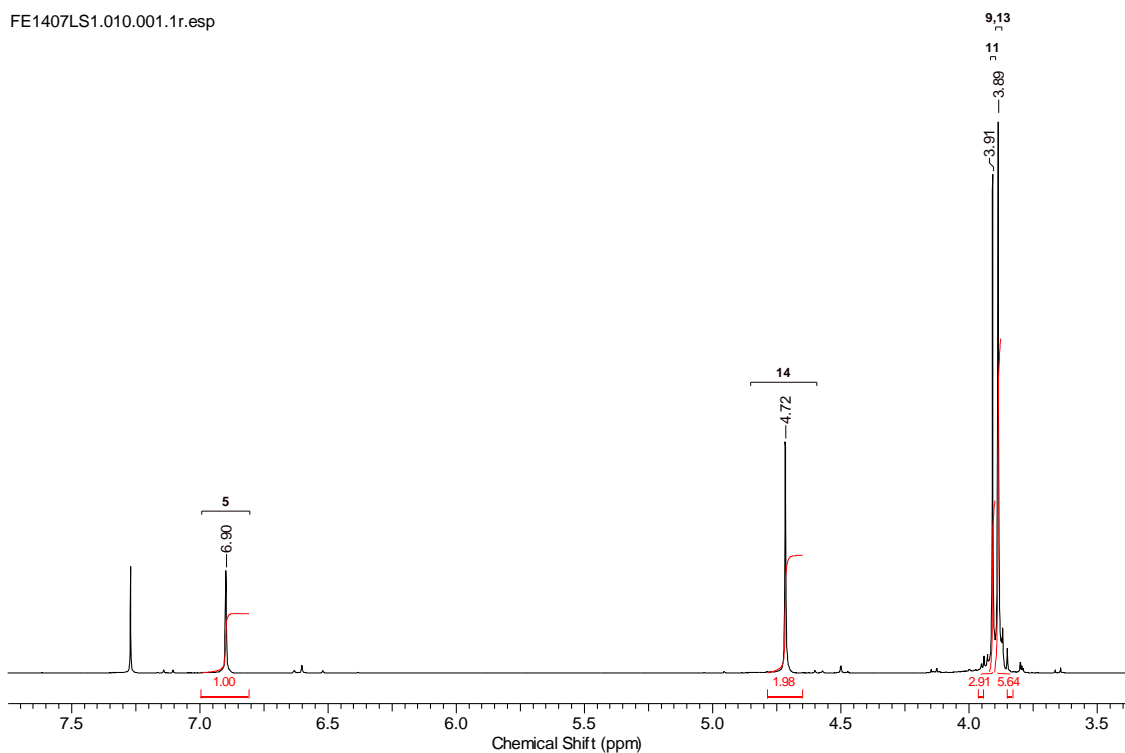
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.89 (6 H, s) 3.91 (3 H, s) 4.72 (2 H, s) 6.90 (1 H, s)

2-Bromo-3,4,5-trimethoxybenzyl benzoate (2.15)

Comment	LS4998/80	Date	12 Jun 2008 16:13:04
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1500.0565	Temperature (degree C)	25.160
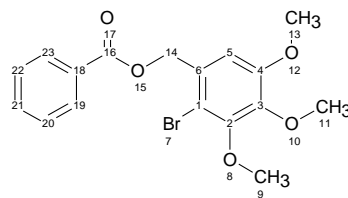
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300 1\2008\JU1208LS1\10\PDATA\1\1r

JU1208LS1.010.001.1r.esp

Formula $C_{17}H_{17}BrO_5$ **FW**   381.2179

¹H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.78 (3 H, s) 3.83 (3 H, s) 3.84 (3 H, s) 5.34 (2 H, s) 6.85 (1 H, s) 7.29 - 7.42 (2 H, m) 7.42 - 7.54 (1 H, m) 8.02 (2 H, d, *J*=8.1 Hz)

176

3,4,5-Trimethoxybenzyl 2-bromobenzoate (2.21)

Comment	LS5291/23 F94-119		
Date	24 Sep 2008 14:11:28		
Frequency (MHz)	300.13	**Nucleus**	1H
Number of Transients	8	**Solvent**	Acetone
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	1846.7201	**Temperature (degree C)**	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2008\SE2408LS3\10\PDATA\1\1r

Formula $C_{17}H_{17}BrO_5$ **FW**   381.2179

SE2408LS3.010.001.1r.esp
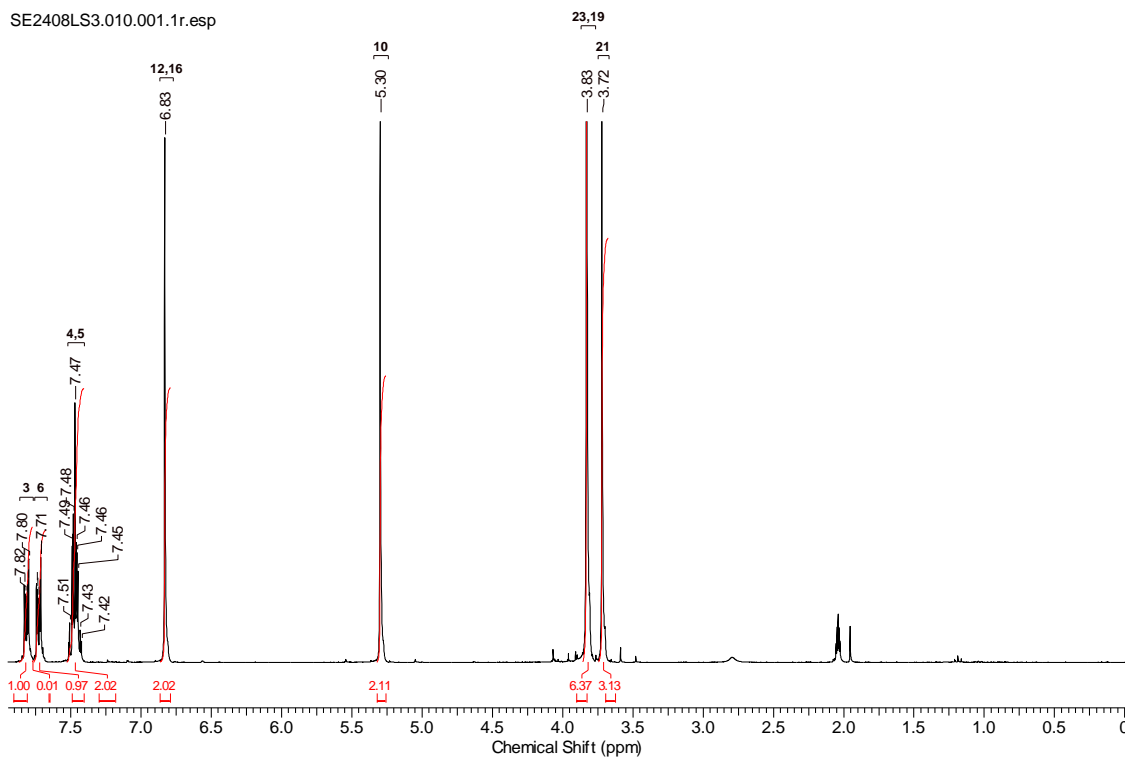
^{1}H NMR (300 MHz, *Acetone*) δ ppm 3.72 (3 H, s) 3.83 (6 H, s) 5.30 (2 H, s) 6.83 (2 H, s) 7.41 - 7.53 (2 H, m) 7.67 - 7.76 (1 H, m) 7.77 - 7.87 (1 H, m)

3,4,5-Trimethoxybenzyl-2-iodobenzoate (2.24)

Comment	LS5291/28	Date	23 Oct 2008 10:10:08
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9382	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\OC2308LS1\10\FID
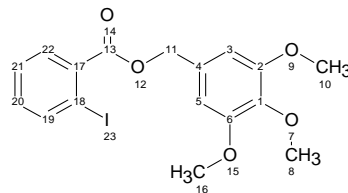
OC2308LS1.010.esp

Formula $C_{17}H_{17}IO_5$ **FW** 428.2183
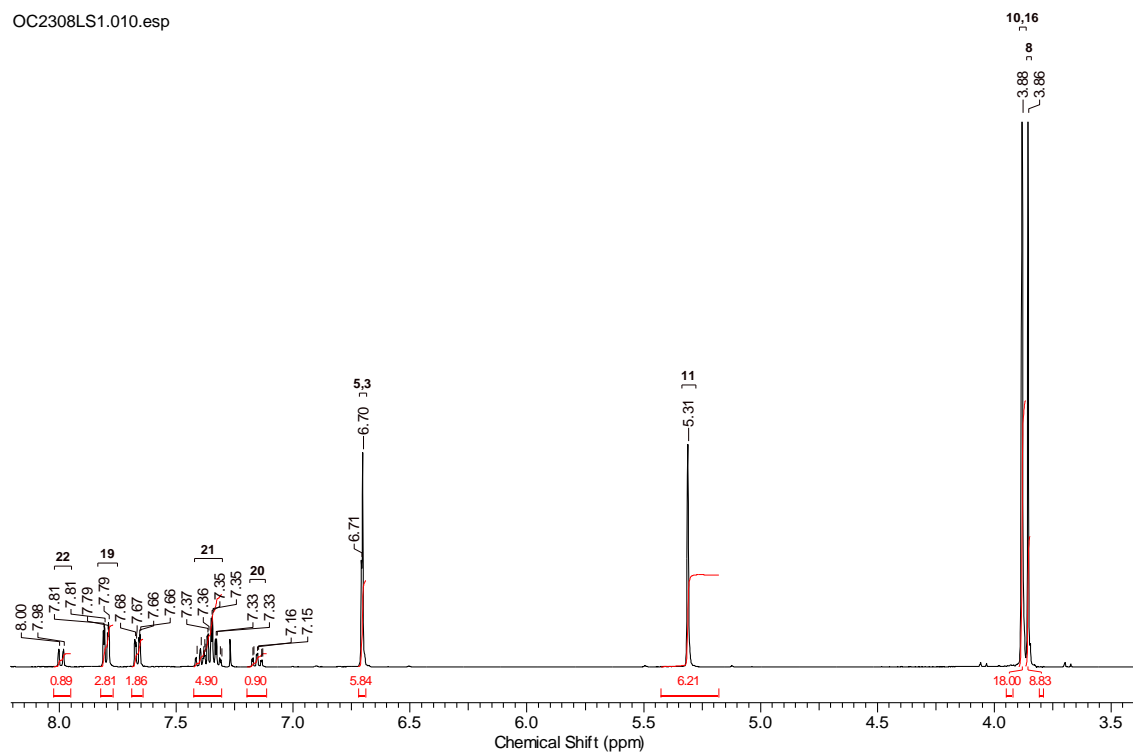
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.86 (9 H, s) 3.88 (18 H, s) 5.31 (6 H, s) 6.69 - 6.72 (6 H, m) 7.15 (1 H, td, *J*=7.7, 1.8 Hz) 7.31 - 7.42 (5 H, m) 7.63 - 7.70 (2 H, m) 7.80 (3 H, dd, *J*=7.5, 2.0 Hz) 7.99 (1 H, d, *J*=8.0 Hz)

2-Bromo-3,4,5-trimethoxybenzyl-2-bromobenzoate (2.27)

Comment	LS5291/39	Date	24 Nov 2008 10:35:44
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2453.9421	Temperature (degree C)	27.000
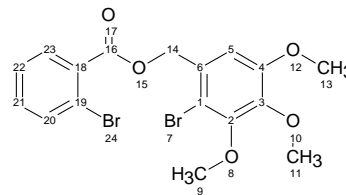
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400 1\NV2408LS1\10\FID

NV2408LS1.010.esp

Formula $C_{17}H_{16}Br_2O_5$
FW 460.1139
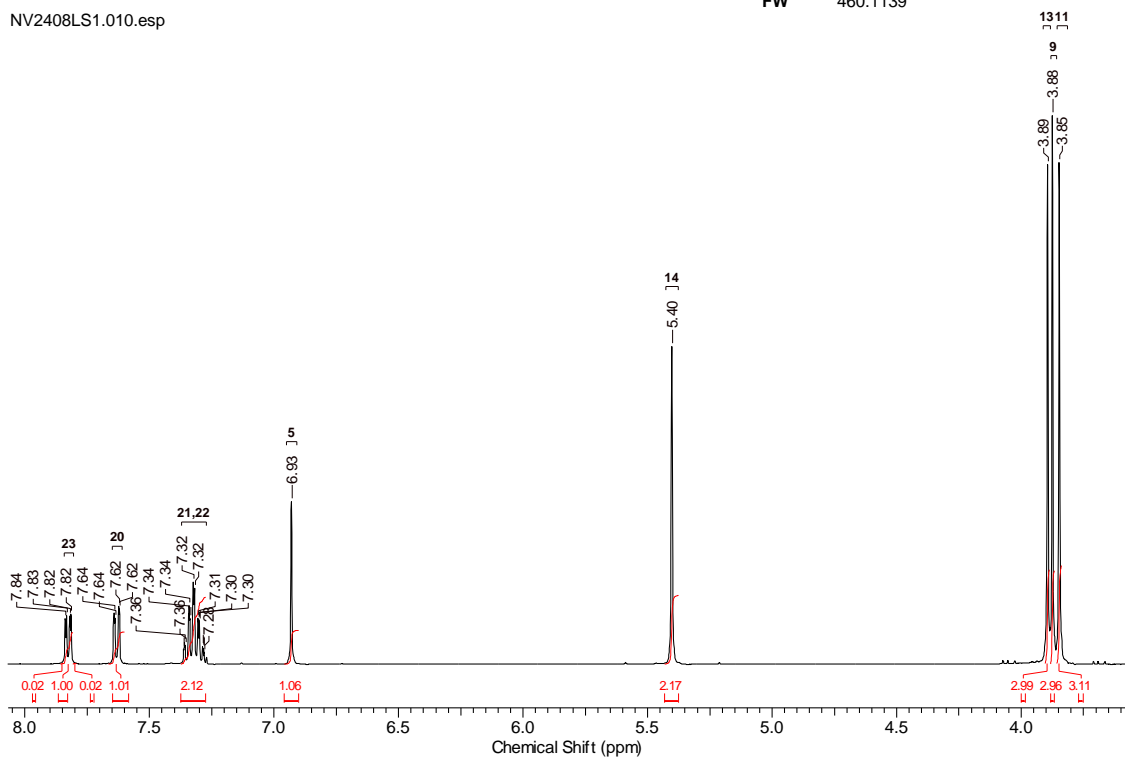
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.85 (3 H, s) 3.88 (3 H, s) 3.89 (3 H, s) 5.40 (2 H, s) 6.93 (1 H, s) 7.28 - 7.37 (2 H, m) 7.63 (1 H, dd, *J*=7.0, 1.0 Hz) 7.83 (1 H, dd, *J*=7.0, 2.0 Hz)

2-Bromo-1-(((2-bromobenzyl)oxy)methyl)-3,4,5-trimethoxybenzene (2.33)

Comment	LS5529/54 F7-12		
Date	20 Jul 2009 13:30:40		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9387	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\JY2009LS3\10\FID

JY2009LS3.010.esp
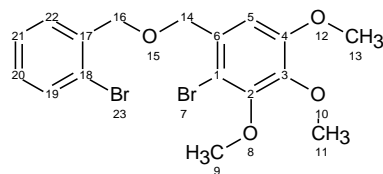
Formula $C_{17}H_{18}Br_2O_4$
FW   446.1304
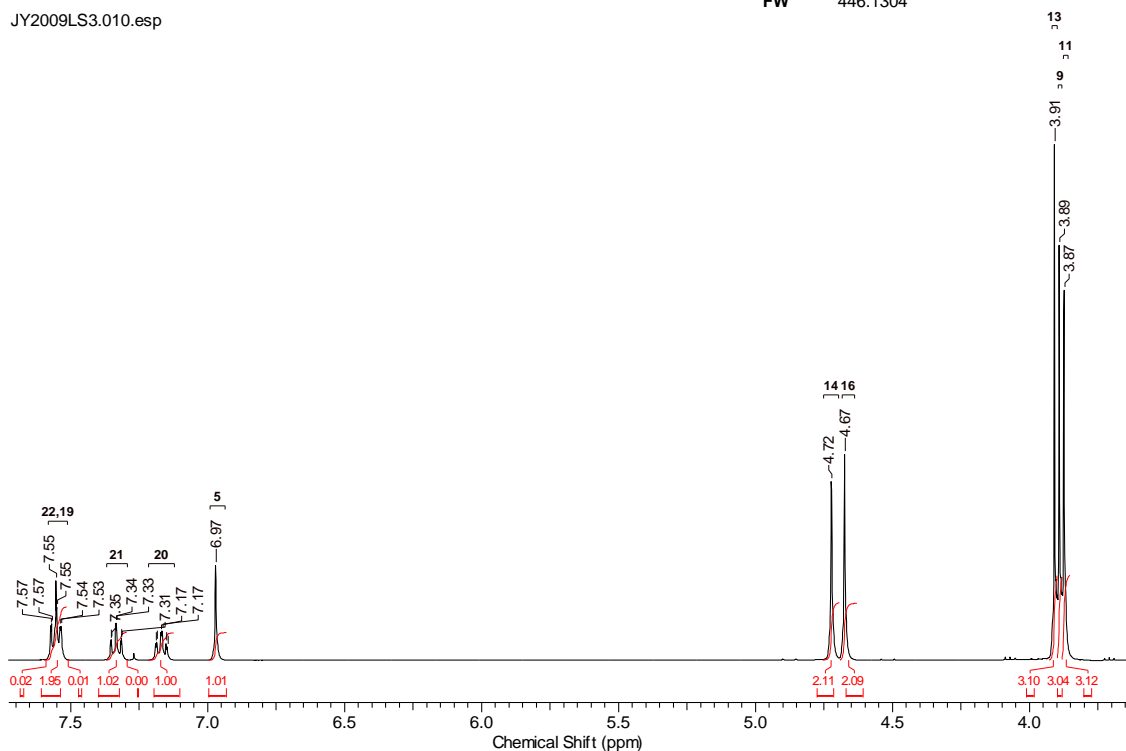
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.87 (3 H, s) 3.89 (3 H, s) 3.91 (3 H, s) 4.67 (2 H, s) 4.72 (2 H, s) 6.97 (1 H, s) 7.17 (1 H, td, *J*=7.8 x (2), 2.0 Hz) 7.33 (1 H, td, *J*=7.5 x (2), 1.0 Hz) 7.51 - 7.58 (2 H, m)

180

5,7-Dihydroxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (2.38)

Comment	LS/4781/26 A			
Date	21 Dec 2006 10:48:32			
Frequency (MHz)	300.13	**Nucleus**	1H	
Number of Transients	16	**Solvent**	Acetone	
Original Points Count	16384	**Points Count**	16384	
Spectrum Offset (Hz)	1496.9465			

Temperature (degree C) 23.700
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2006\DC2106LS1\10\PDATA\1\1r

DC2106LS1.010.001.1r.esp
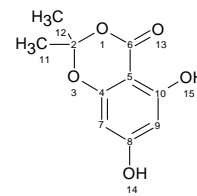
Formula $C_{10}H_{10}O_5$ **FW**   210.1834
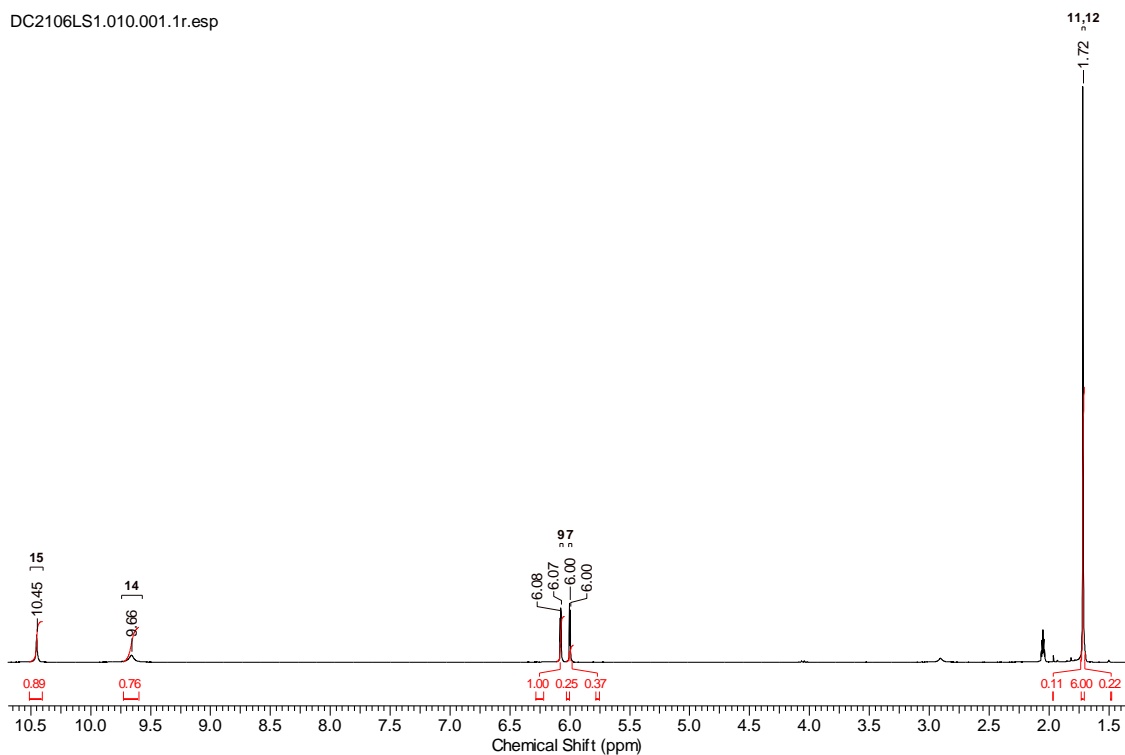
^{1}H NMR (300 MHz, *Acetone*) δ ppm 1.72 (6 H, s) 6.00 (1 H, d, *J*=2.2 Hz) 6.08 (1 H, d, *J*=1.8 Hz) 9.66 (1 H, br. s.) 10.45 (1 H, s)

5-Hydroxy-7-methoxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (2.39)

^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.72 (6 H, s) 3.81 (3 H, s) 6.00 (1 H, d, *J*=2.2 Hz) 6.14 (1 H, d, *J*=2.2 Hz) 10.43 (1 H, s)

7-Methoxy-2,2-dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate (2.40)

Comment	LS/4781/23 columned B		
Date	30 Nov 2006 18:12:16		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8612	Temperature (degree C)	27.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2006\NV3006LS2\10\PDATA\1\1r

NV3006LS2.010.001.1r.esp
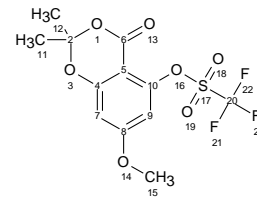
Formula C$_{12}$H$_{11}$F$_3$O$_7$S
FW 356.2717
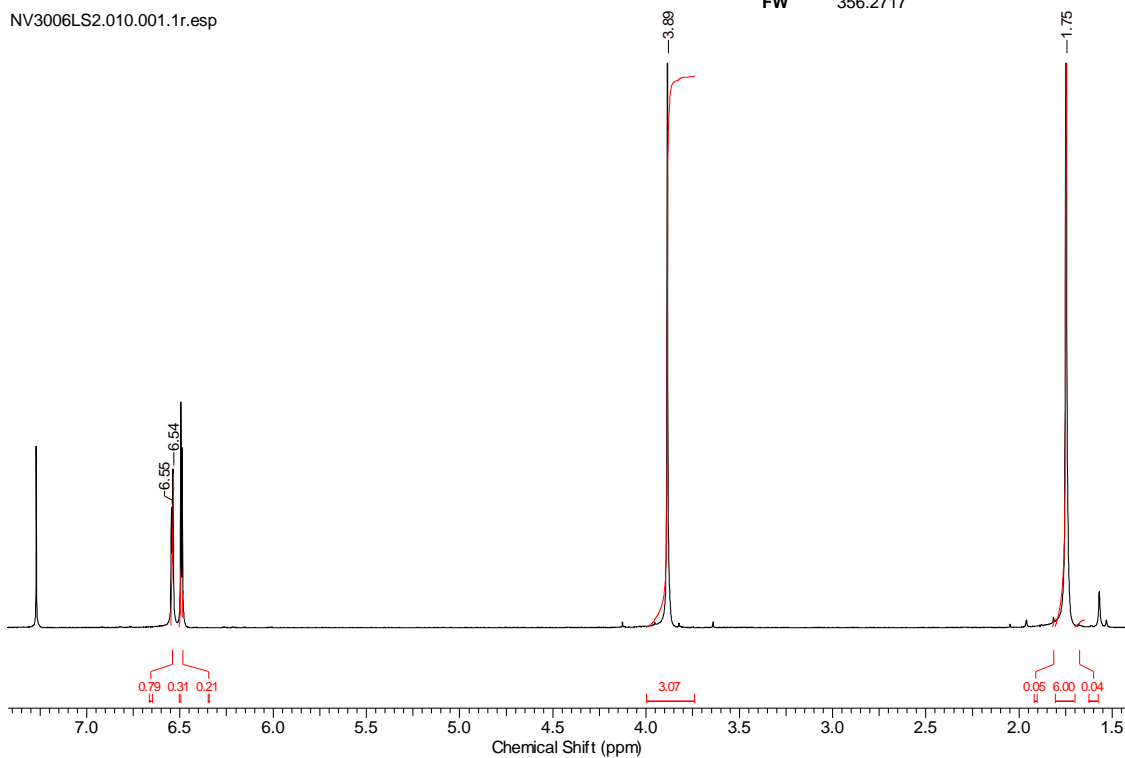
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.75 (6 H, s) 3.89 (3 H, s) 6.49 (1 H, d, *J*=2.2 Hz) 6.54 (1 H, d, *J*=2.2 Hz)

4-Hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.45)
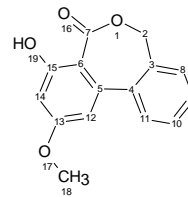
Comment	LS/4781/28 Fn 7		
Date	18 Dec 2006 15:57:52		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8611	Temperature (degree C)	27.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2006\DC1806LS1\10\PDATA\1\1r

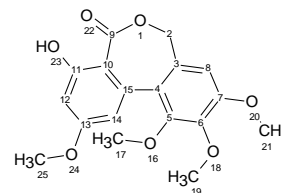DC1806LS1.010.001.1r.esp

Formula $C_{15}H_{12}O_4$ FW   256.2534
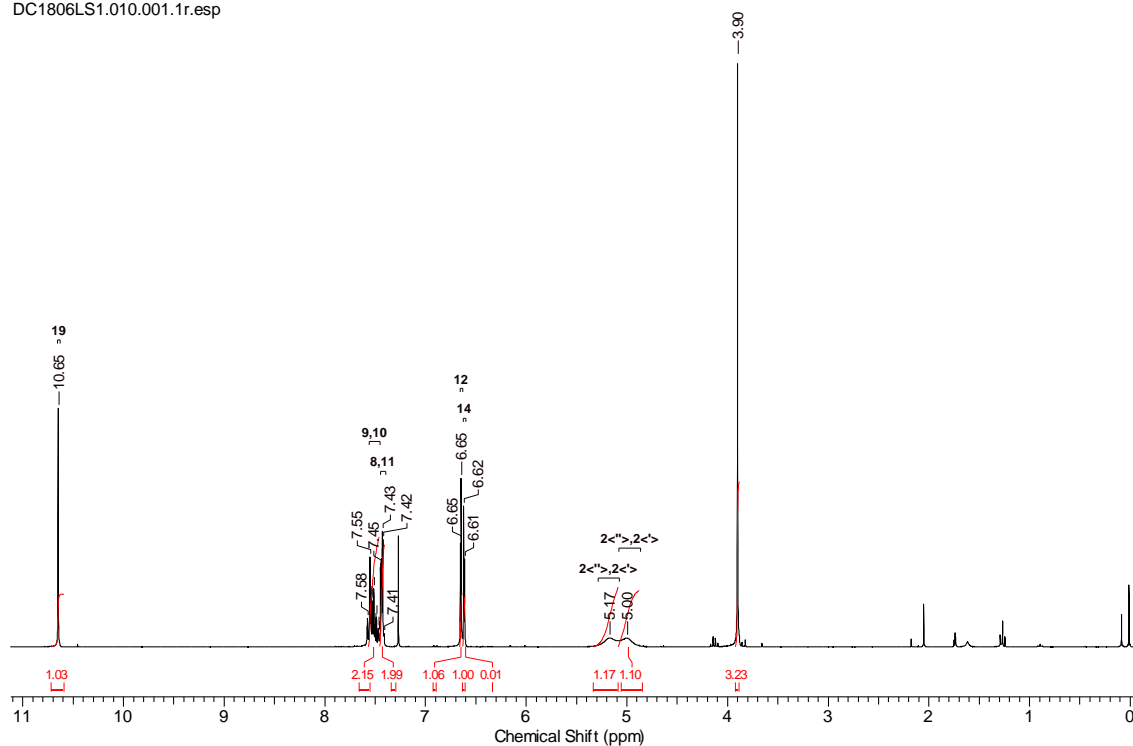
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.90 (3 H, s) 5.00 (1 H, br. s.) 5.17 (1 H, br. s.) 6.62 (1 H, d, *J*=2.6 Hz) 6.65 (1 H, d, *J*=2.6 Hz) 7.40 - 7.45 (2 H, m) 7.46 - 7.57 (2 H, m) 10.65 (1 H, s)

4-Hydroxy-2-methoxy-10-nitrodibenzo[c,e]oxepin-5(7H)-one (2.47)

Comment	LS4781/85b1		
Date	21 Sep 2007 14:41:20		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8612	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2007\SE2107LS1\10\PDATA\1\1r

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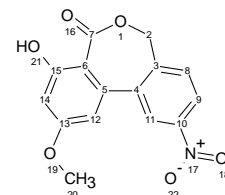
Formula $C_{15}H_{11}NO_6$ FW   301.2509
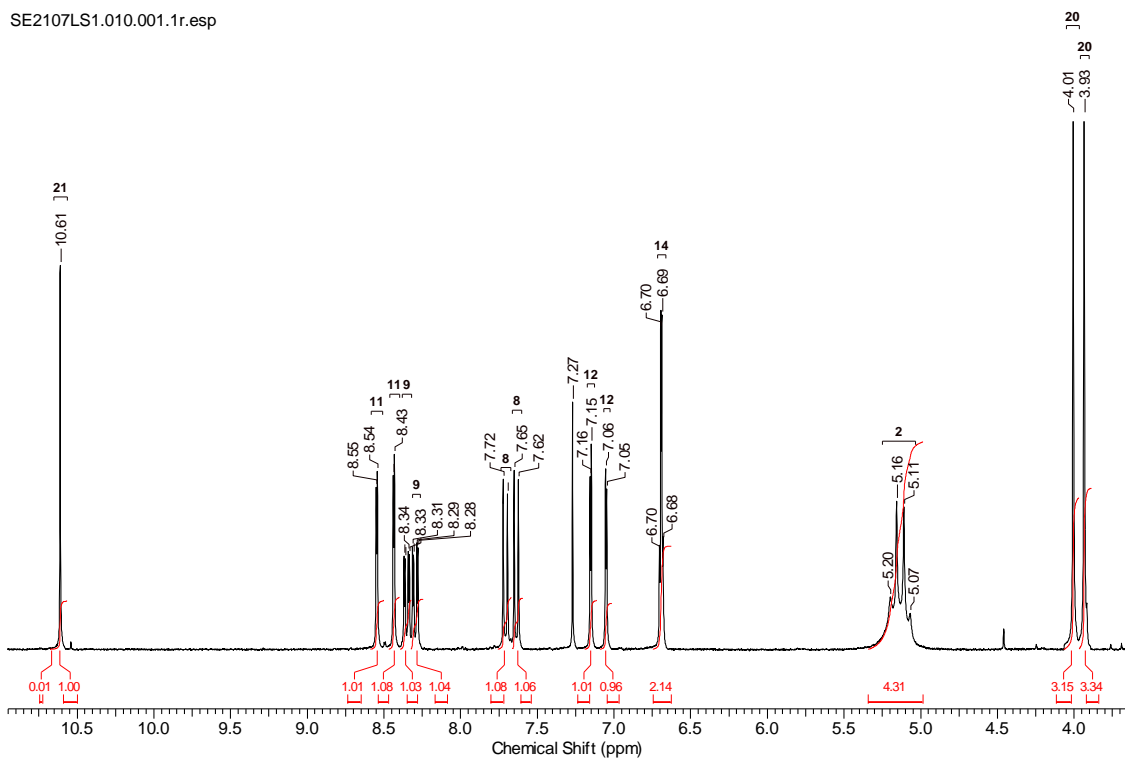
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.93 (3 H, s) 4.01 (3 H, s) 5.04 - 5.25 (4 H, m) 6.69 (2 H, q, *J*=2.6 Hz) 7.05 (1 H, d, *J*=2.2 Hz) 7.15 (1 H, d, *J*=2.6 Hz) 7.64 (1 H, d, *J*=8.4 Hz) 7.71 (1 H, d, *J*=8.4 Hz) 8.30 (1 H, dd, *J*=8.2, 2.4 Hz) 8.35 (1 H, dd, *J*=8.2, 2.4 Hz) 8.44 (1 H, d, *J*=2.2 Hz) 8.55 (1 H, d, *J*=2.2 Hz) 10.61 (1 H, s)

(2-Bromo-4-methylphenyl)methanol (2.51)

Comment	LS/4781/46 columned		
Date	13 Mar 2007 09:55:12		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8610	Temperature (degree C)	27.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2007\MA1307LS1\10\PDATA\1\1r

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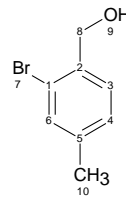
Formula C_8H_9BrO FW   201.0605
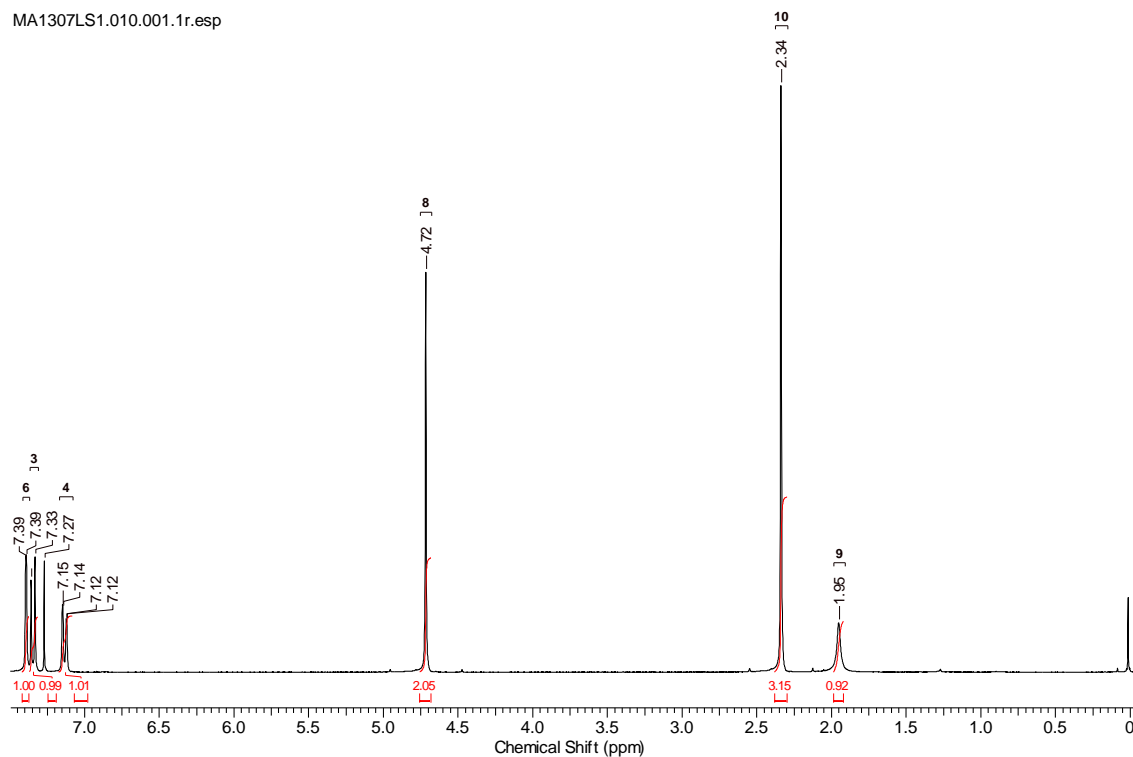
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.95 (1 H, br. s.) 2.34 (3 H, s) 4.72 (2 H, s) 7.13 (1 H, dd, *J*=7.7, 1.5 Hz) 7.34 (1 H, d, *J*=7.7 Hz) 7.39 (1 H, d, *J*=1.5 Hz)

7-Methoxy-2,2-dimethyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-4H-benzo[d][1,3]dioxin-4-one (2.55)

Comment	LS5529/32 F56-112		
Date	05 Jun 2009 11:44:16		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.9613	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2009\JU0509LS1\10\PDATA\1\1r

JU0509LS1.010.001.1r.esp
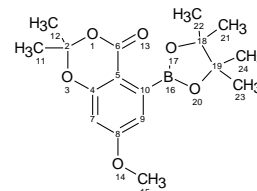
Formula $C_{17}H_{23}BO_6$ FW   334.1719
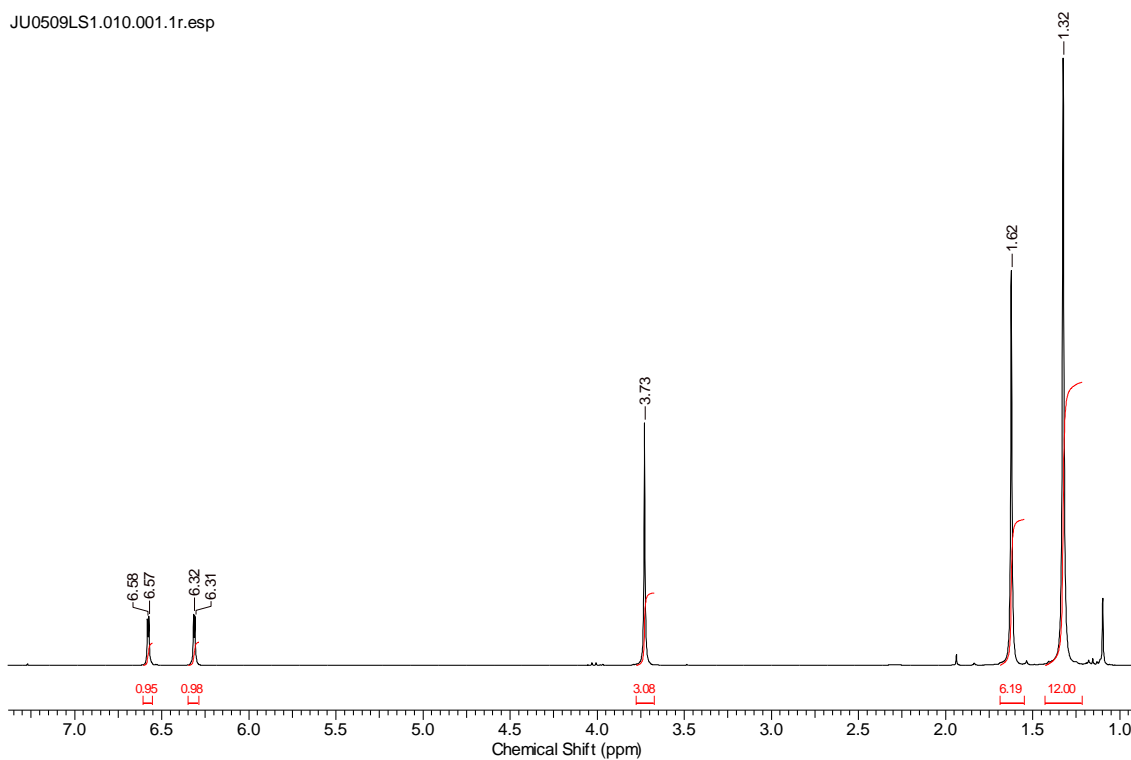
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.32 (12 H, s) 1.62 (6 H, s) 3.73 (3 H, s) 6.31 (1 H, d, *J*=2.6 Hz) 6.58 (1 H, d, *J*=2.6 Hz)

5-(2-(Hydroxymethyl)-5-methylphenyl)-7-methoxy-2,3-dimethyl-4H-benzo[d][1,3] dioxin-4-one (2.57)

Comment	LS5529/29 F11-20		
Date	02 Jun 2009 13:20:00		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000
File Name	C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400 1\JU0209LS1\10\FID		

JU0209LS1.010.esp
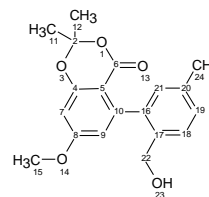
Formula $C_{19}H_{20}O_5$ FW   328.3591
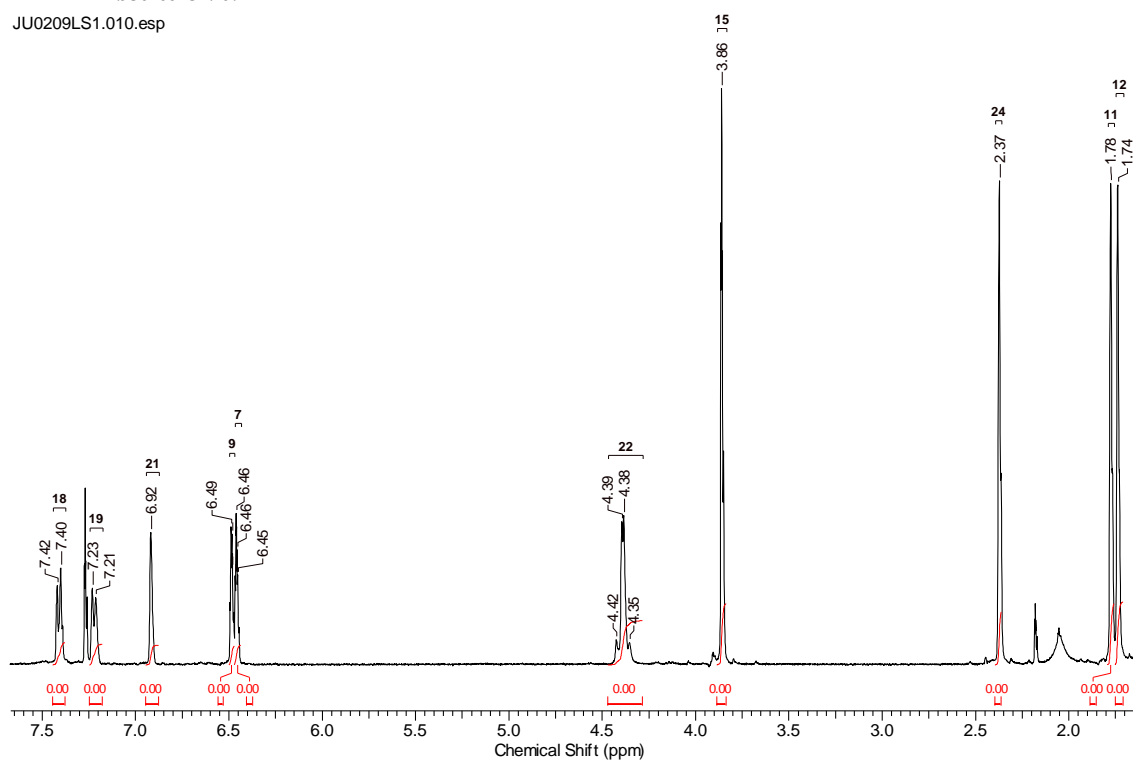
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 1.74 (3 H, s) 1.78 (3 H, s) 2.37 (3 H, s) 3.86 (3 H, s) 4.28 - 4.47 (2 H, m) 6.44 - 6.47 (1 H, m) 6.47 - 6.50 (1 H, m) 6.92 (1 H, s) 7.22 (1 H, d, *J*=7.5 Hz) 7.41 (1 H, d, *J*=8.0 Hz)

4-Hydroxy-2-methoxy-10-methyldibenzo[c,e]oxepin-5(7H)-one (2.58)

Comment	LS/4781/79.2a		
Date	25 Jun 2007 15:40:48		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2454.4441	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400 1\JU2507LS1\1\FID

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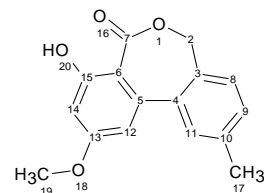
Formula $C_{16}H_{14}O_4$ FW   270.2800
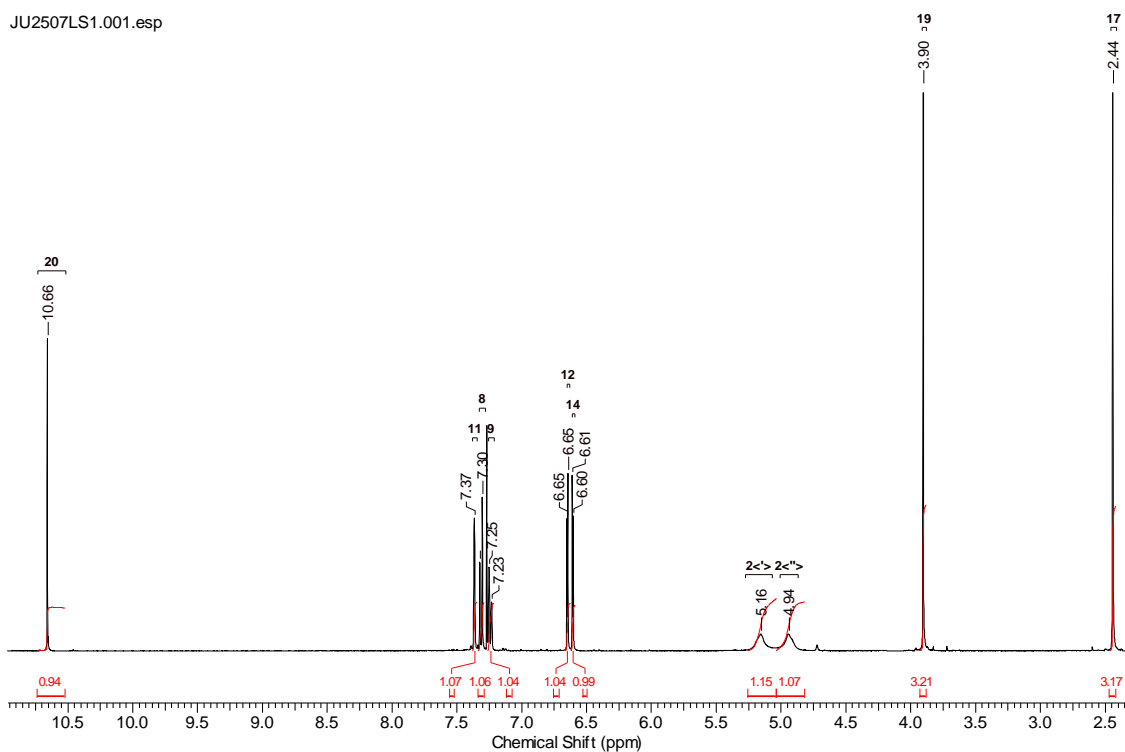
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 2.44 (3 H, s) 3.90 (3 H, s) 4.94 (1 H, br. s.) 5.16 (1 H, br. s.) 6.61 (1 H, d, *J*=2.5 Hz) 6.65 (1 H, d, *J*=2.5 Hz) 7.24 (1 H, d, *J*=8.0 Hz) 7.31 (1 H, d, *J*=8.0 Hz) 7.37 (1 H, s) 10.66 (1 H, s)

4-Hydroxy-2,9,10,11-tetramethoxydibenzo[c,e]oxepin-5(7H)-one (2.60)

Comment	LS4998/32	Date	04 Jan 2008 18:44:16
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.4954	Temperature (degree C)	25.160

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1\2008\JA0408LS1\10\PDATA\1\1r

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Formula $C_{18}H_{18}O_7$ **FW** 346.3313
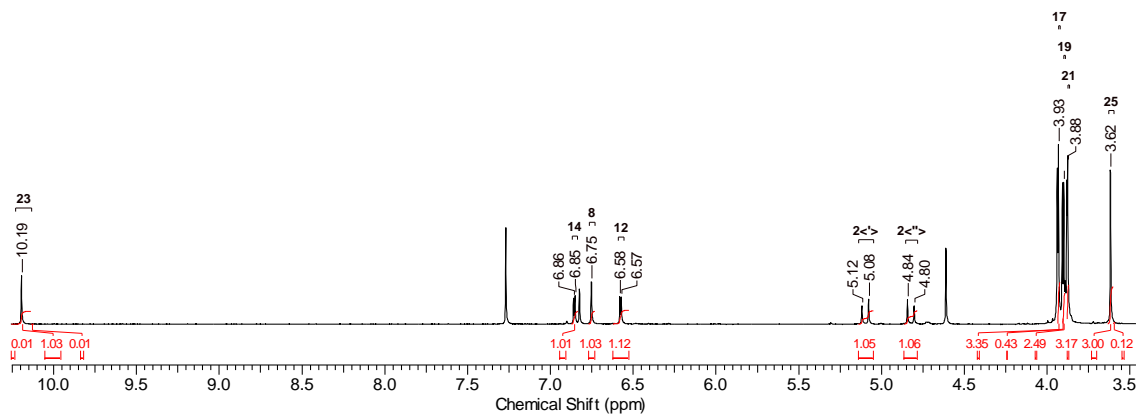
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.62 (3 H, s) 3.88 (3 H, s) 3.90 (3 H, s) 3.93 (3 H, s) 4.82 (1 H, d, *J*=12.1 Hz) 5.10 (1 H, d, *J*=12.1 Hz) 6.58 (1 H, d, *J*=2.6 Hz) 6.75 (1 H, s) 6.86 (1 H, d, *J*=2.6 Hz) 10.19 (1 H, s)

(2-Bromo-5-methoxyphenyl)methanol (2.64)

Comment	LS/4781/47 columned		
Date	13 Mar 2007 10:31:28		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8611	Temperature (degree C)	27.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300 1\2007\MA1307LS2\10\PDATA\1\1r

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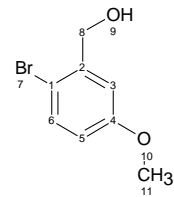
Formula $C_8H_9BrO_2$ FW   217.0599
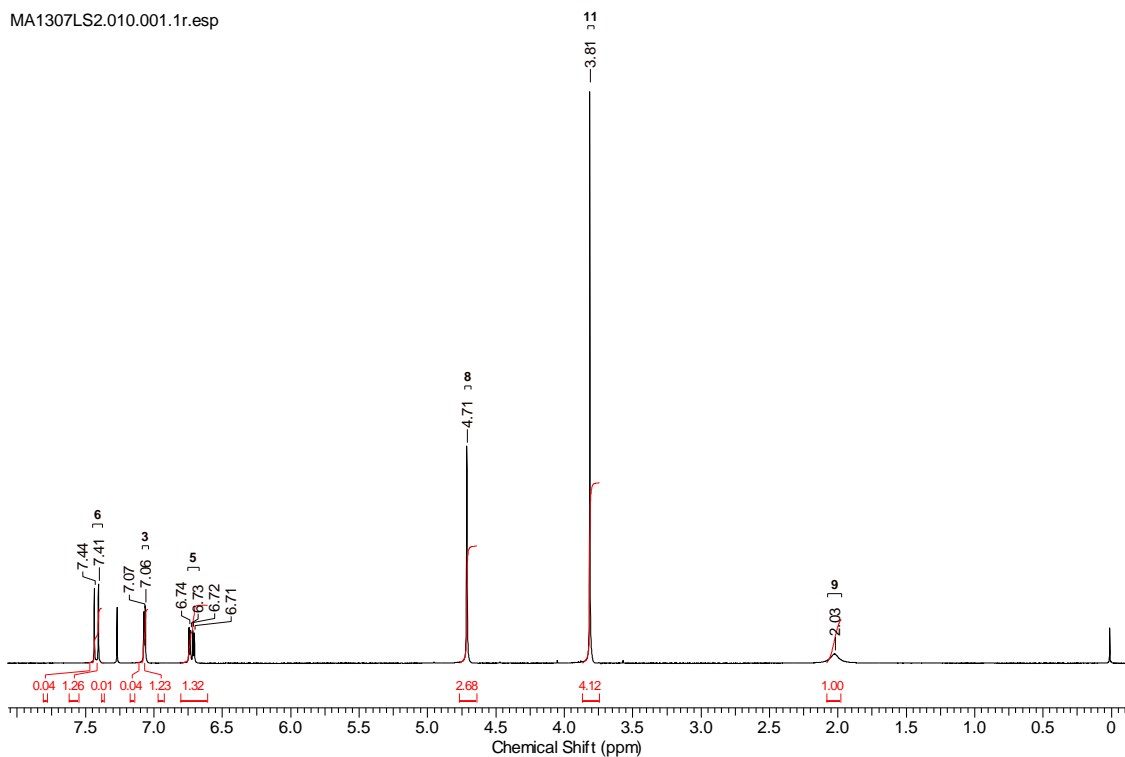
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.03 (1 H, br. s.) 3.81 (3 H, s) 4.71 (2 H, s) 6.72 (1 H, dd, *J*=8.6, 3.1 Hz) 7.07 (1 H, d, *J*=2.9 Hz) 7.42 (1 H, d, *J*=8.8 Hz)

4-Hydroxy-2,9-dimethoxydibenzo[c,e]oxepin-5(7H)-one (2.66)

Comment	LS/4781/83.2		
Date	27 Jun 2007 18:59:28		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1473.2133	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
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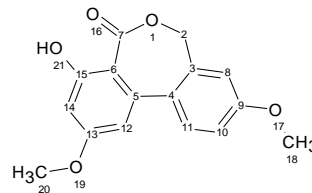
Formula $C_{16}H_{14}O_5$ **FW** 286.2794
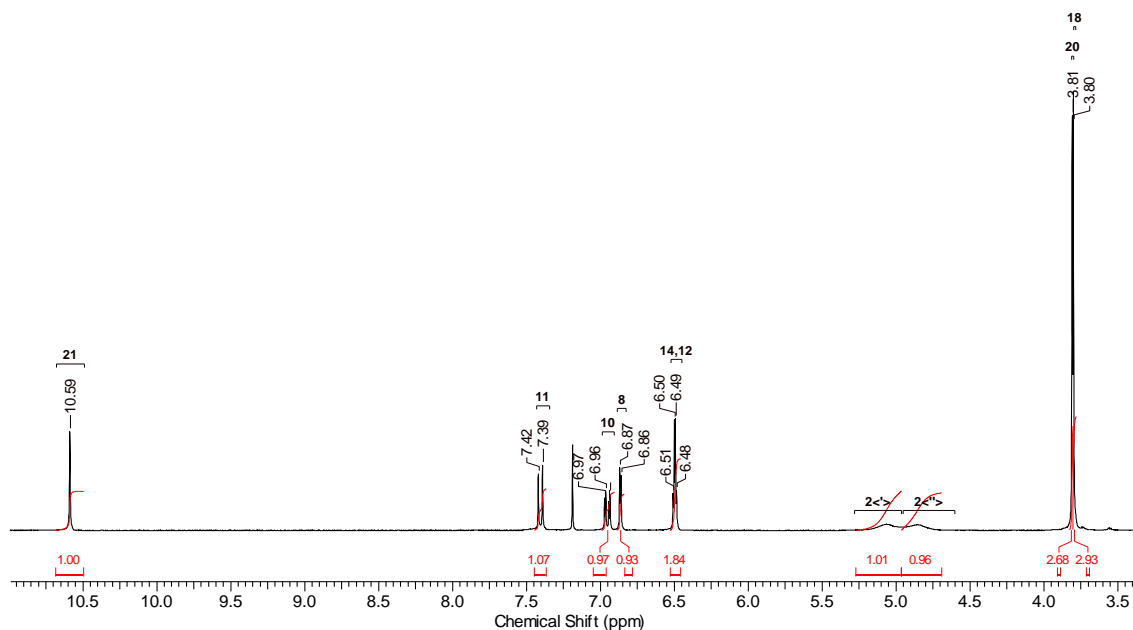
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.80 (3 H, s) 3.81 (3 H, s) 4.78 (1 H, br. s) 5.13 (1 H, br. s) 6.50 (2 H, q, *J*=2.6 Hz) 6.86 (1 H, d, *J*=2.9 Hz) 6.95 (1 H, dd, *J*=8.6, 2.7 Hz) 7.41 (1 H, d, *J*=8.4 Hz) 10.59 (1 H, s)

3,4-Dihydroxy-5-methoxybenzaldehyde (2.68)

Comment	LS5529/6 Batch 3			
Date	03 Apr 2009 16:15:12			
Frequency (MHz)	300.13	**Nucleus**	1H	
Number of Transients	8	**Solvent**	Acetone	
Original Points Count	16384	**Points Count**	16384	
Spectrum Offset (Hz)	1849.7213			

Temperature (degrees C) 25.000

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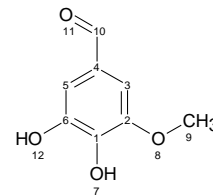
Formula $C_8H_8O_4$ **FW** 168.1467
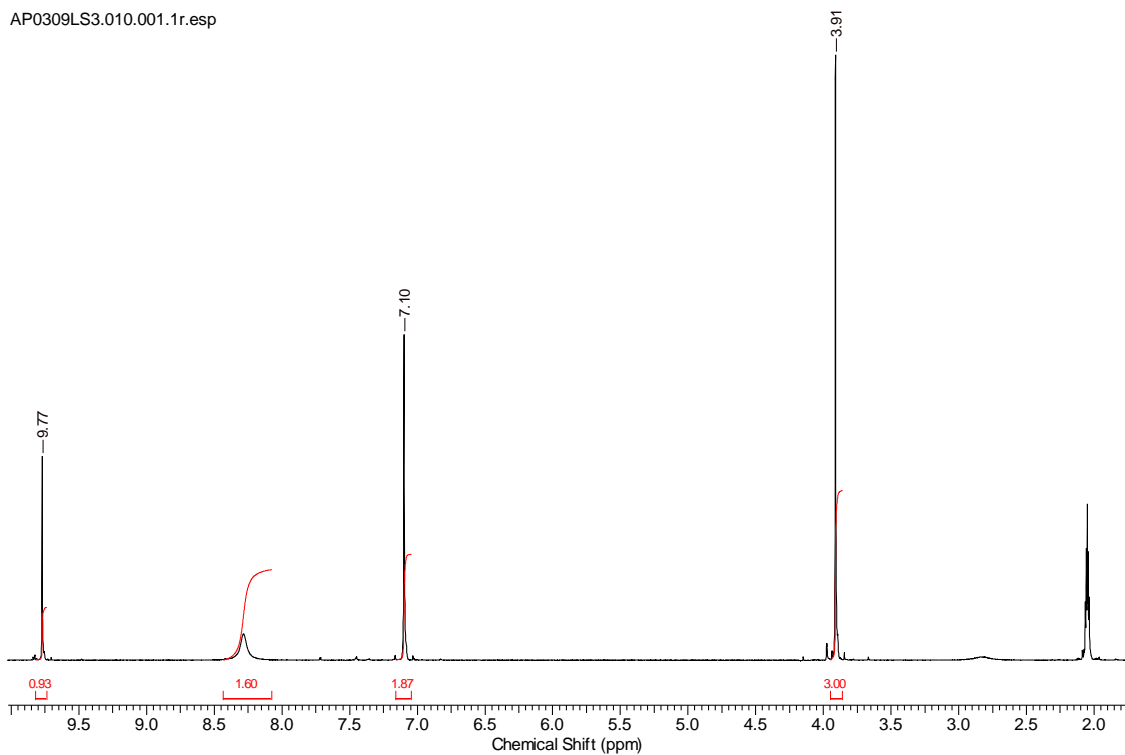
^{1}H NMR (300 MHz, *Acetone*) δ ppm 3.91 (3 H, s) 7.10 (2 H, s) 8.25 (2 H, br. s) 9.77 (1 H, s)

2-Bromo-3,4-dihydroxy-5-methoxybenzaldehyde (2.69)

Comment	LS4998/42B1		
Date	26 Feb 2008 20:09:36		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	Aceton
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2464.3511		

Temperature (degree) C\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\FE2608LS1\10\FID

FE2608LS1.010.esp
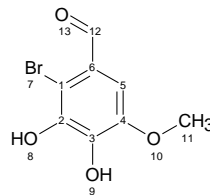
Formula $C_8H_7BrO_4$ FW   247.0428
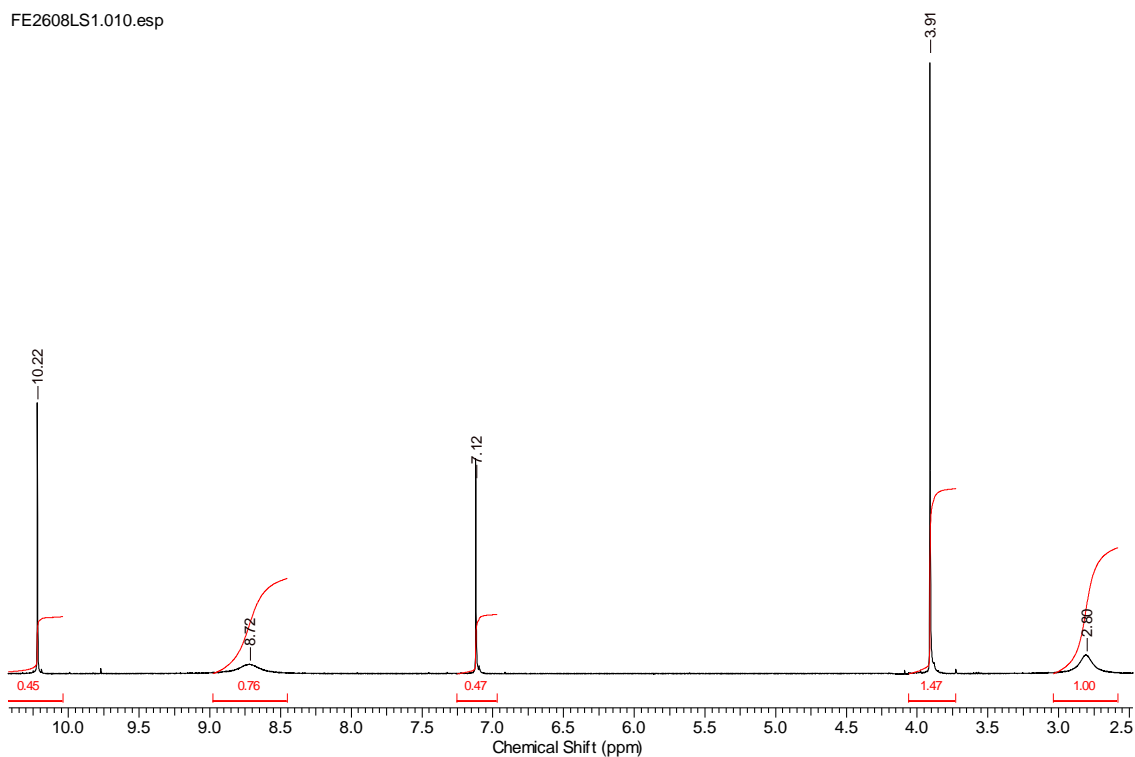
^{1}H NMR (400 MHz, *Aceton*) δ ppm 2.81 (1 H, br. s) 3.91 (3 H, s) 7.12 (1 H, s) 8.72 (1 H, br. s) 10.28 (7 H, s)

194

3-Bromo-4-formyl-6-methoxy-1,2-phenylene diacetate (2.70)

Comment	LS4998/55B		
Date	14 Apr 2008 23:13:20		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1491.6406	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
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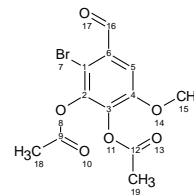
Formula $C_{12}H_{13}BrO_6$ FW   333.1320
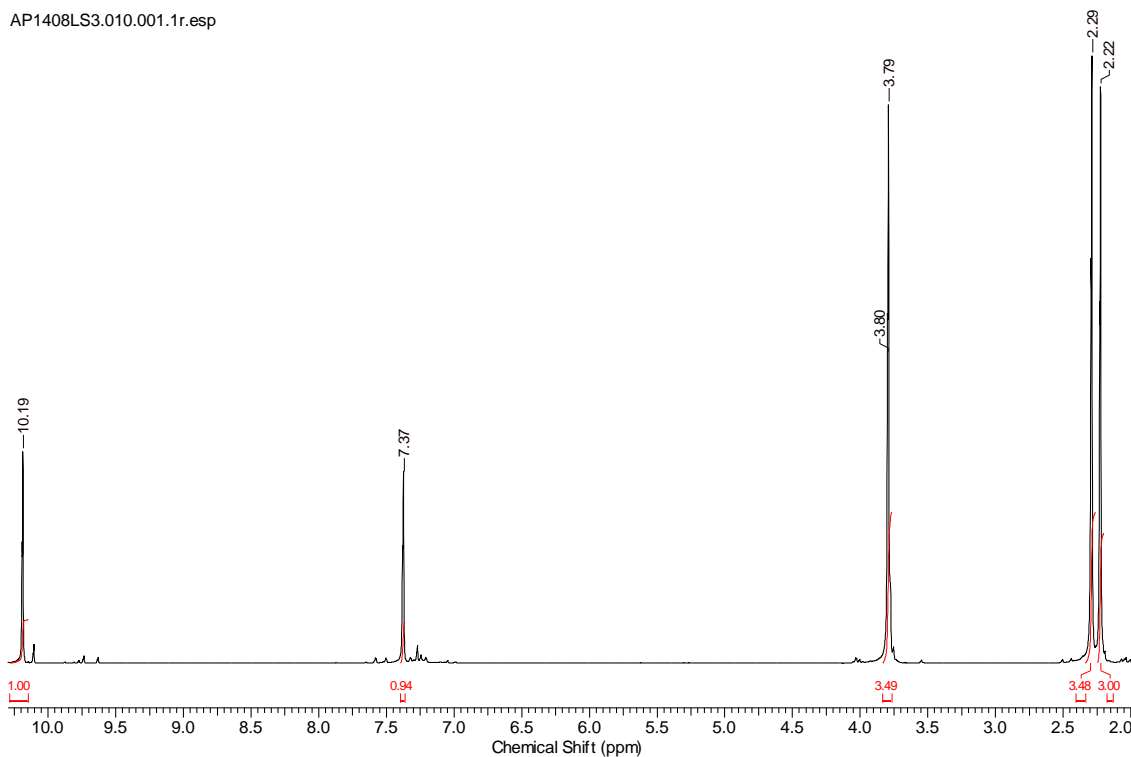
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.22 (3 H, s) 2.29 (3 H, s) 3.77 - 3.83 (3 H, m) 7.37 (1 H, s) 10.19 (1 H, s)

3,4-Diacetoxy-2-bromo-5-methoxybenzyl alcohol (2.71)

Comment	LS4998/47		
Date	29 Feb 2008 17:31:44		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	Acetone
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1496.5806		
Temperature (degree C) 28.700			

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300 1\2008\FE2908LS1\10\PDATA\1\1r

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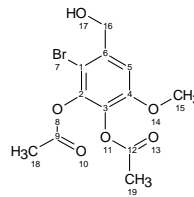
Formula $C_{12}H_{13}BrO_6$ FW   333.1320
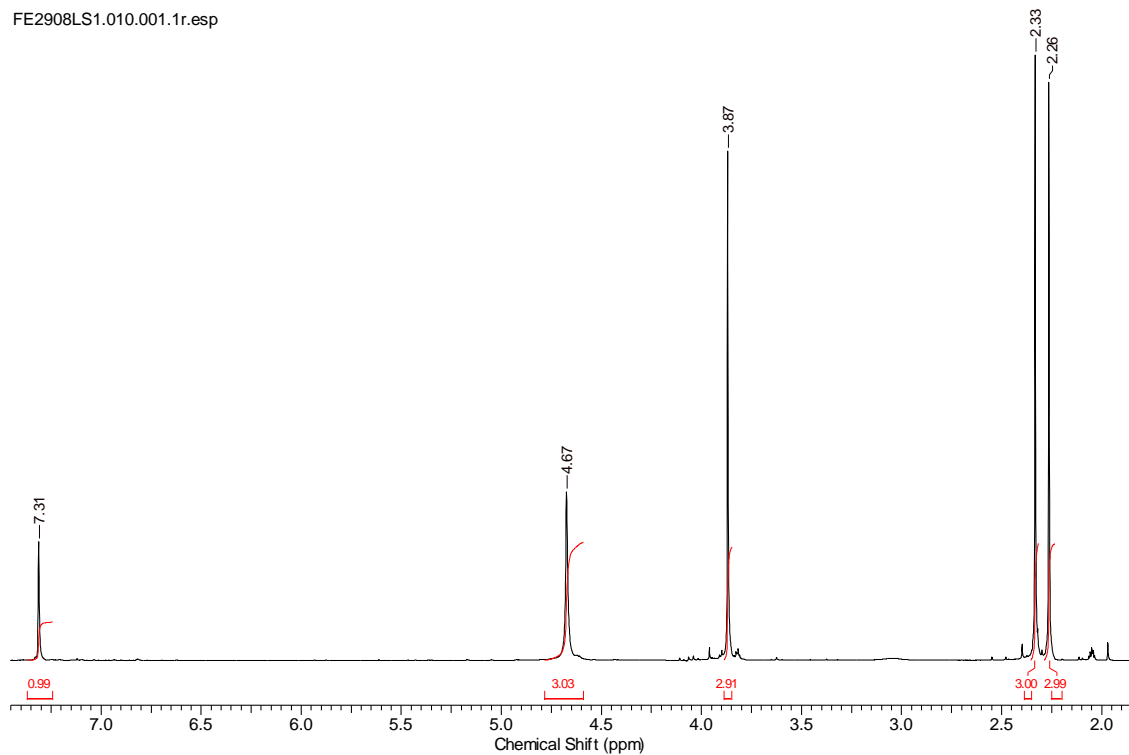
^{1}H NMR (300 MHz, *Acetone*) δ ppm 2.26 (3 H, s) 2.33 (3 H, s) 3.87 (3 H, s) 4.67 (2 H, s) 7.31 (1 H, s)

4-Hydroxy-2,9,10-trimethoxydibenzo[c,e]oxepin-5(7H)-one (2.78)

Comment	LS5291/36 EA acid ext		
Date	13 Jan 2009 22:30:24		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	32	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.4952	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2009\JA1309LS2\10\PDATA\1\1r

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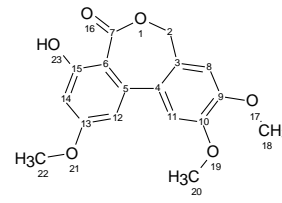
Formula C$_{17}$H$_{16}$O$_6$ FW   316.3053
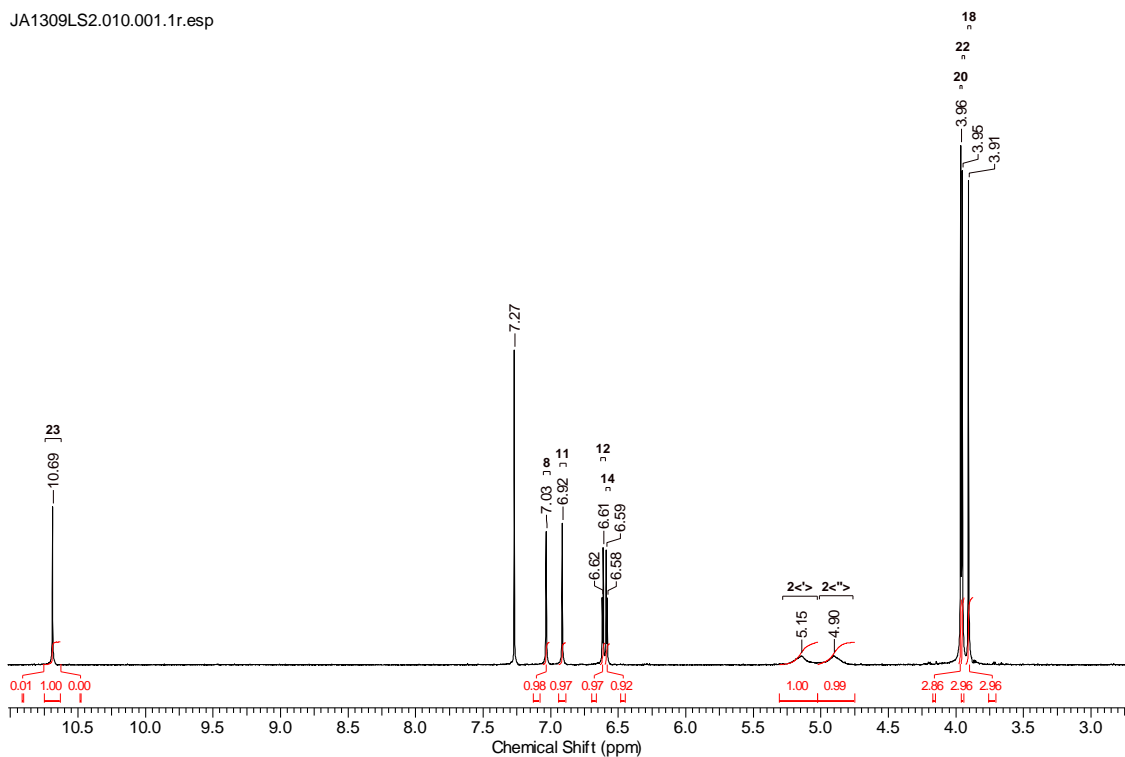
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.91 (3 H, s) 3.95 (3 H, s) 3.96 (3 H, s) 4.90 (1 H, br. s.) 5.15 (1 H, br. s.) 6.59 (1 H, d, *J*=2.6 Hz) 6.61 (1 H, d, *J*=2.6 Hz) 6.92 (1 H, s) 7.03 (1 H, s) 10.69 (1 H, s)

Methyl-2-bromo-3,5-dimethoxybenzoate (2.80)

Comment	LS4998/94	Date	31 Jul 2008 11:35:44
Frequency (MHz)	300.13	**Nucleus**	1H
Number of Transients	8	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	1850.9613	**Temperature (degree C)**	25.000
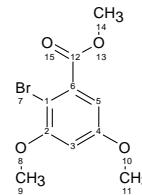
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2\2008\JY3108LS1\10\PDATA\1\1r

JY3108LS1.010.001.1r.esp

Formula $C_{10}H_{11}BrO_4$ **FW** 275.0959
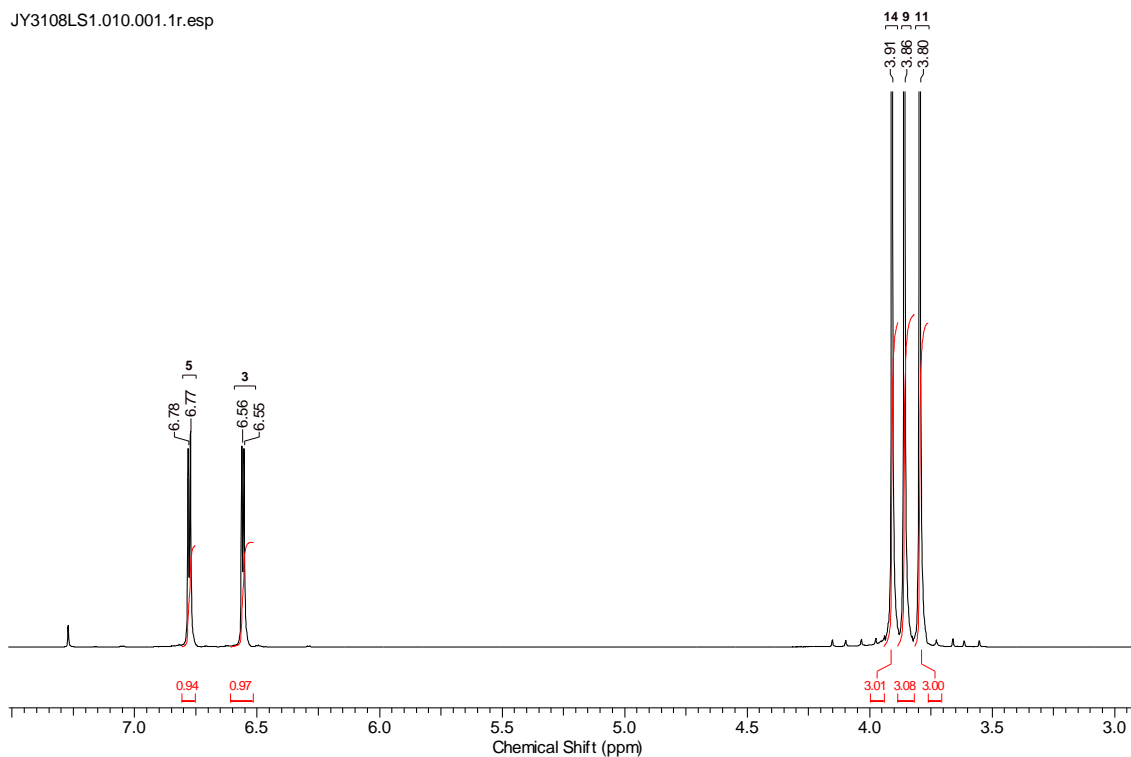
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.80 (3 H, s) 3.86 (3 H, s) 3.91 (3 H, s) 6.56 (1 H, d, *J*=2.6 Hz) 6.78 (1 H, d, *J*=2.6 Hz)

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2-Bromo-3,5-dimethoxybenzylalcohol (2.81)

Comment	LS5291/6	Date	08 Aug 2008 16:34:08
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000
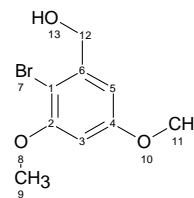
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\AU0808LS2\10\FID

AU0808LS2.010.esp

Formula $C_9H_{11}BrO_3$ **FW** 247.0858
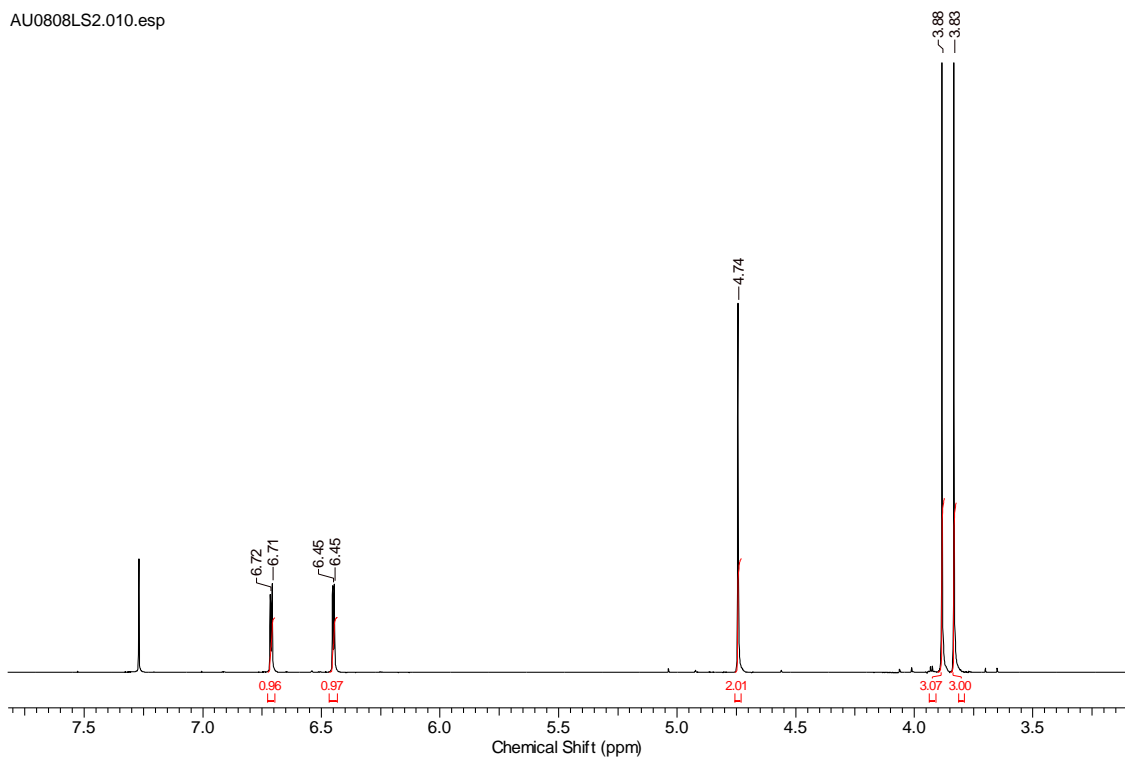
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.83 (3 H, s) 3.88 (3 H, s) 4.74 (2 H, s) 6.45 (1 H, d, *J*=2.8 Hz) 6.71 (1 H, d, *J*=2.8 Hz)

4-Hydroxy-2,9,11-trimethoxydibenzo[c,e]oxepin-5(7H)-one (2.82)

Comment	JL61 org 2 (EtOAc)		
Date	02 Dec 2008 14:58:08		
Frequency (MHz)	400.13	**Nucleus**	1H
Number of Transients	16	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	2452.9385	**Temperature (degree C)**	27.000

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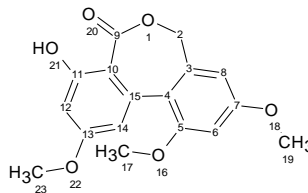
Formula $C_{17}H_{16}O_6$ **FW** 316.3053
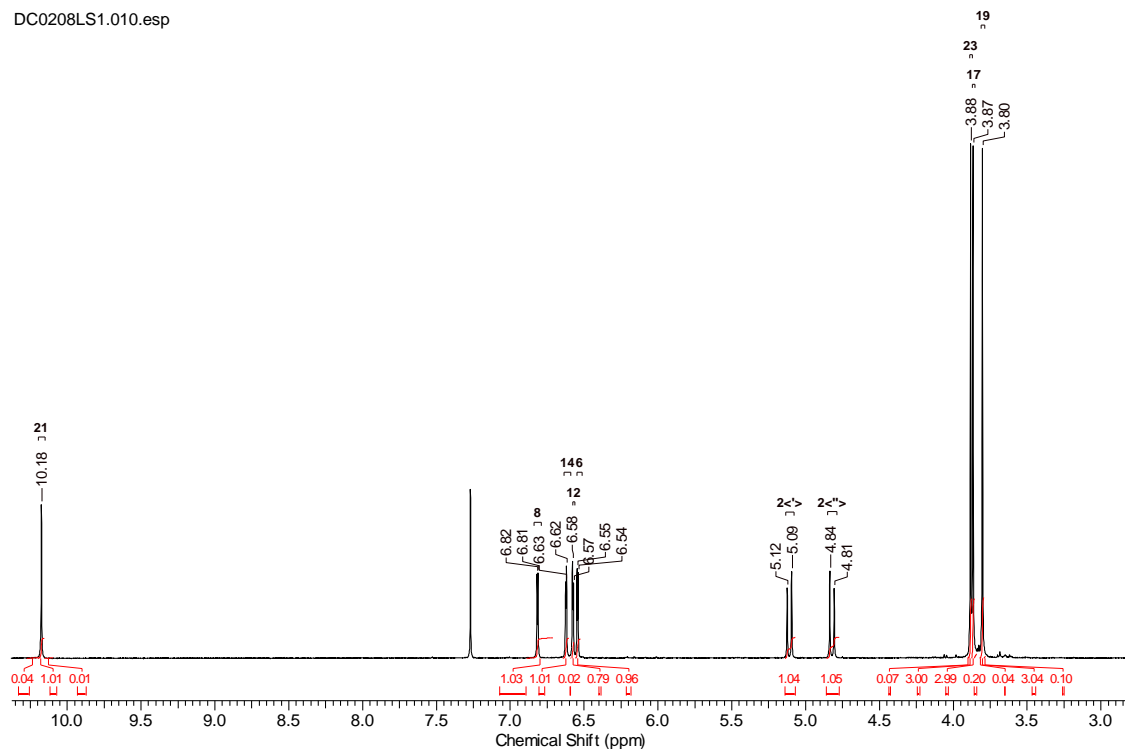
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.80 (3 H, s) 3.87 (3 H, s) 3.88 (3 H, s) 4.82 (1 H, d, *J*=12.0 Hz) 5.11 (1 H, d, *J*=12.0 Hz) 6.54 (1 H, d, *J*=2.5 Hz) 6.58 (1 H, d, *J*=2.5 Hz) 6.62 (1 H, d, *J*=2.5 Hz) 6.82 (1 H, d, *J*=2.5 Hz) 10.18 (1 H, s)

200

8-Hydroxy-10-methoxy-7-oxo-5,7-dihydrobenzo[c,e]ozepine-2-carbonitrile (2.84)

Comment	LS5529/22 F165-184 B		
Date	19 May 2009 17:36:16		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.4951	Temperature (degree C)	24.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300 1\2009\MY1909LS1\10\PDATA\1\1r

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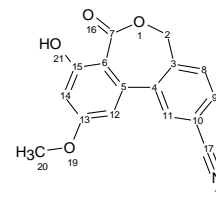
Formula $C_{16}H_{11}NO_4$ FW 281.2628
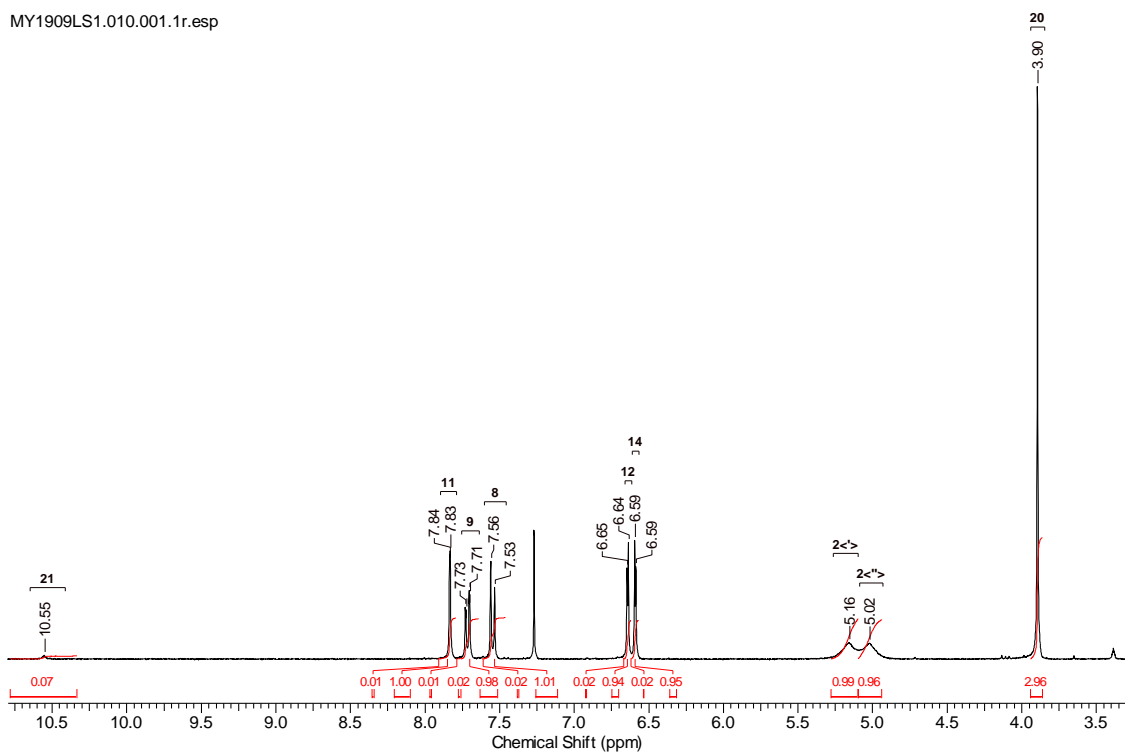
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.90 (3 H, s) 5.02 (1 H, br. s.) 5.16 (1 H, br. s.) 6.59 (1 H, d, *J*=2.2 Hz) 6.64 (1 H, d, *J*=2.2 Hz) 7.55 (1 H, d, *J*=7.7 Hz) 7.72 (1 H, dd, *J*=7.7, 1.5 Hz) 7.83 (1 H, d, *J*=1.1 Hz) 10.55 (1 H, br. s.)

9-Fluoro-4-hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.86)

Comment	LS5529/26 EA extract of base		
Date	27 May 2009 19:58:56		
Frequency (MHz)	400.13	**Nucleus**	1H
Number of Transients	32	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	2452.9387	**Temperature (degree C)**	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
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MY2709LS1.010.esp
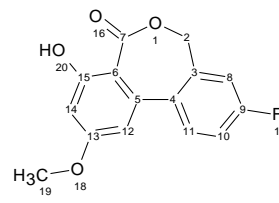
Formula $C_{15}H_{11}FO_4$ **FW** 274.2438
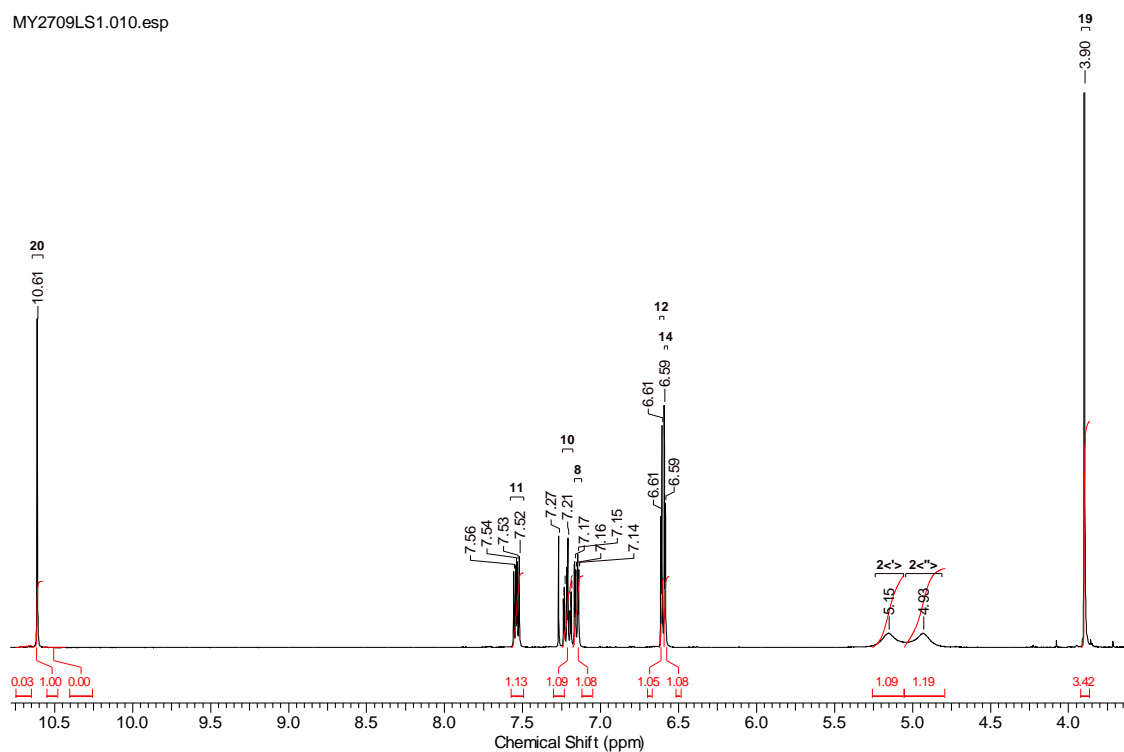
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.90 (3 H, s) 4.93 (1 H, br. s.) 5.15 (1 H, br. s.) 6.59 (1 H, d, *J*=2.5 Hz) 6.61 (1 H, d, *J*=2.5 Hz) 7.15 (1 H, dd, *J*=8.0, 2.5 Hz) 7.21 (1 H, td, *J*=8.5, 3.0 Hz) 7.54 (1 H, dd, *J*=8.8, 5.3 Hz) 10.61 (1 H, s)

202

4-Hydroxy-2-methoxy-9-(trifluoromethyl)dibenzo[c,e]oxepin-5(7H)-one (2.87)

Comment	LS5529/27 EA extract of base		
Date	19 Jun 2009 04:18:08		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	32	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2453.4404	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
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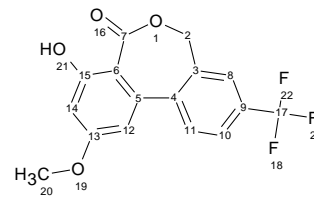
Formula $C_{16}H_{11}F_3O_4$ FW   324.2513
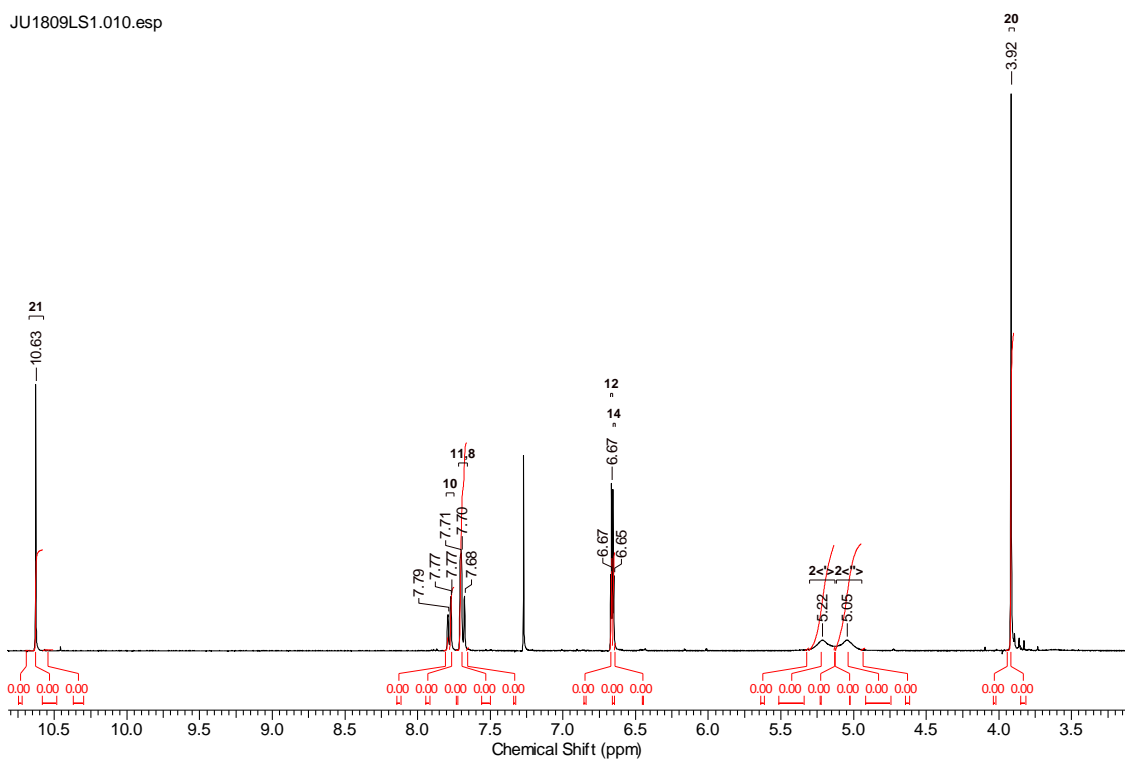
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.92 (3 H, s) 5.05 (1 H, br. s.) 5.22 (1 H, br. s.) 6.65 (1 H, d, *J*=2.5 Hz) 6.67 (1 H, d, *J*=2.5 Hz) 7.66 - 7.72 (2 H, m) 7.78 (1 H, dd, *J*=8.0, 1.0 Hz) 10.63 (1 H, s)

4-Hydroxy-2-methoxy-[1,3]dioxolo[4',5':4,5]benzo[1,2-c]benzo[e]oxepin-5(7H)-one (2.89)

Comment	LS5529/21.1 acid EA extract		
Date	22 May 2009 19:29:04		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	64	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000
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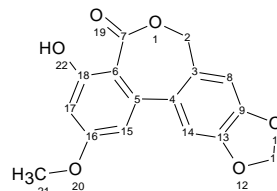
Formula $C_{16}H_{12}O_6$ FW   300.2629
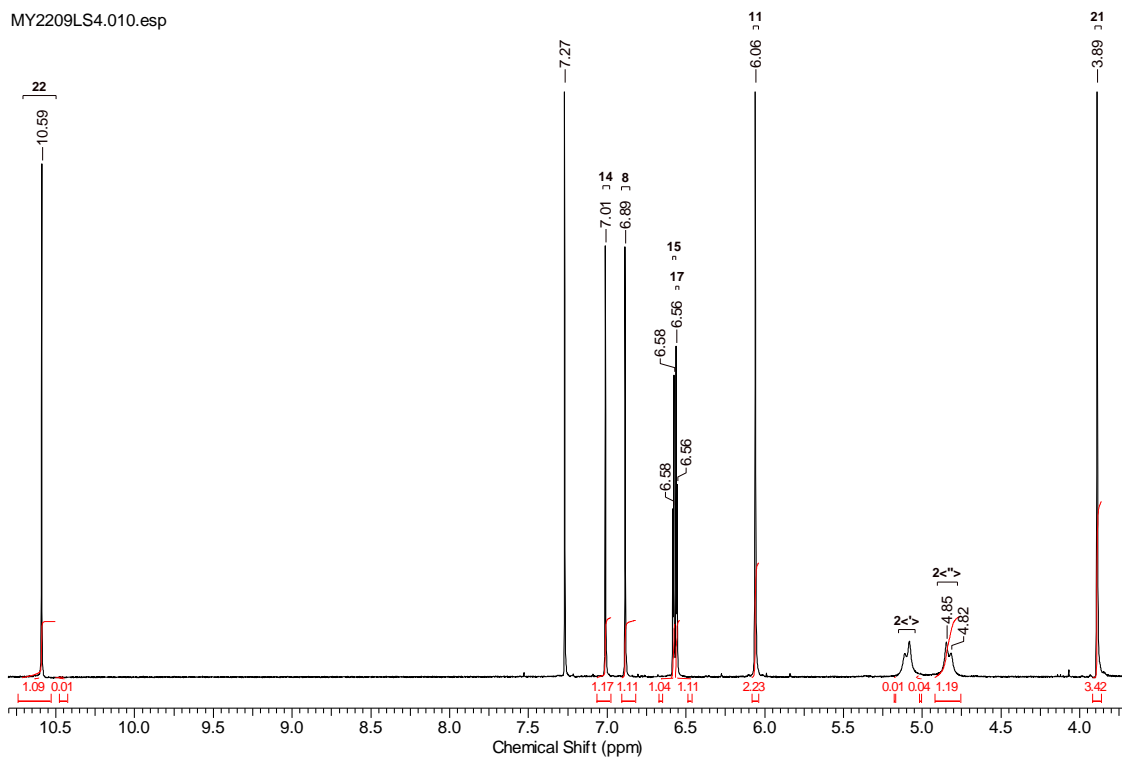
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.89 (3 H, s) 4.83 (1 H, d, *J*=12.0 Hz) 5.10 (1 H, d, *J*=12.0 Hz) 6.06 (2 H, s) 6.56 (1 H, d, *J*=2.5 Hz) 6.58 (1 H, d, *J*=2.5 Hz) 6.89 (1 H, s) 7.01 (1 H, s) 10.59 (1 H, s)

# 2-Bromo-3,4-di-*tert*-butoxycarbonyloxy-5-methoxybenzaldehyde (2.98)

Comment	LS5529/9 F5		
Date	06 Apr 2009 16:40:48		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.8613	Temperature (degree C)	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
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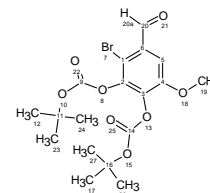
Formula $C_{18}H_{23}BrO_8$ FW   447.2744
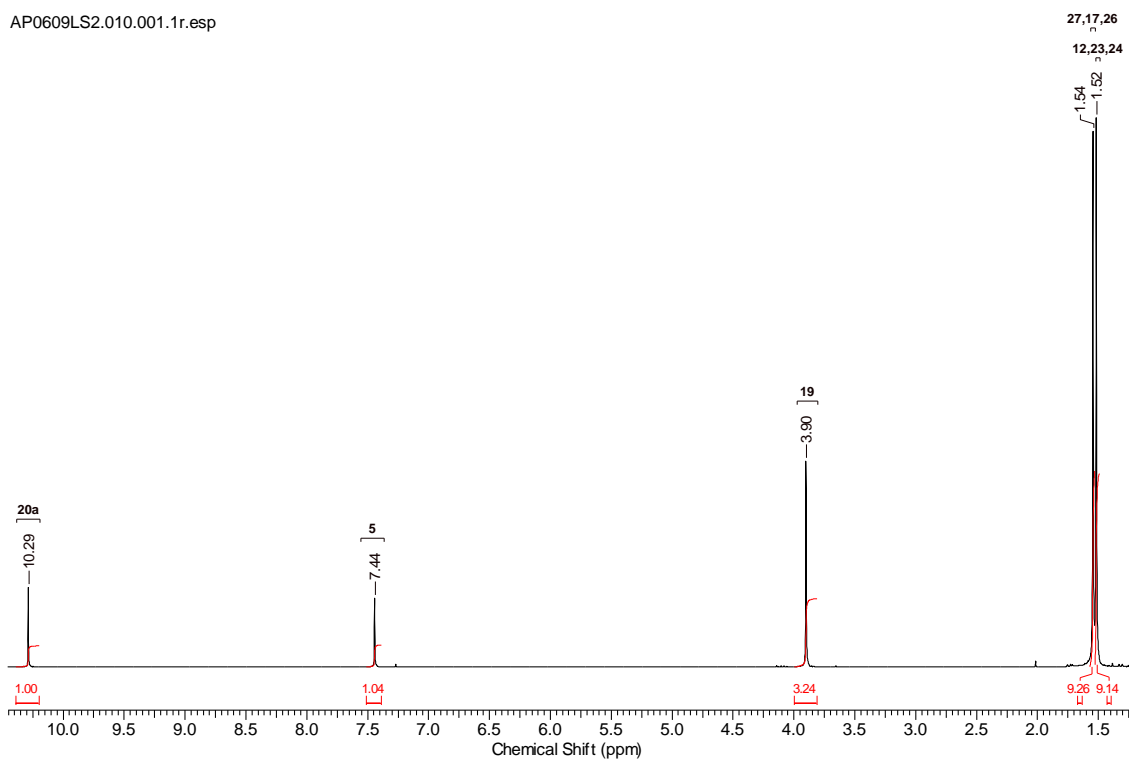
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.52 (9 H, s) 1.54 (9 H, s) 3.90 (3 H, s) 7.44 (1 H, s) 10.29 (1 H, s)

2-Bromo-3,4-di-*tert*butoxycarbonyloxy-5-methoxybenzyl alcohol (2.99)

Comment	LS5529/46 F48-66			
Date	20 Jun 2009 04:09:36			
Frequency (MHz)	400.13	**Nucleus**	1H	
Number of Transients	16	**Solvent**	CHLOROFORM-d	
Original Points Count	16384	**Points Count**	16384	
Spectrum Offset (Hz)	2453.4404	**Temperature (degree C)**	27.000	

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\JU1909LS3\10\FID

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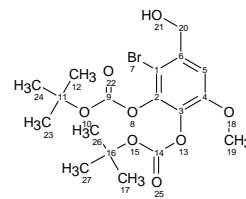
Formula $C_{18}H_{25}BrO_8$ **FW** 449.2903
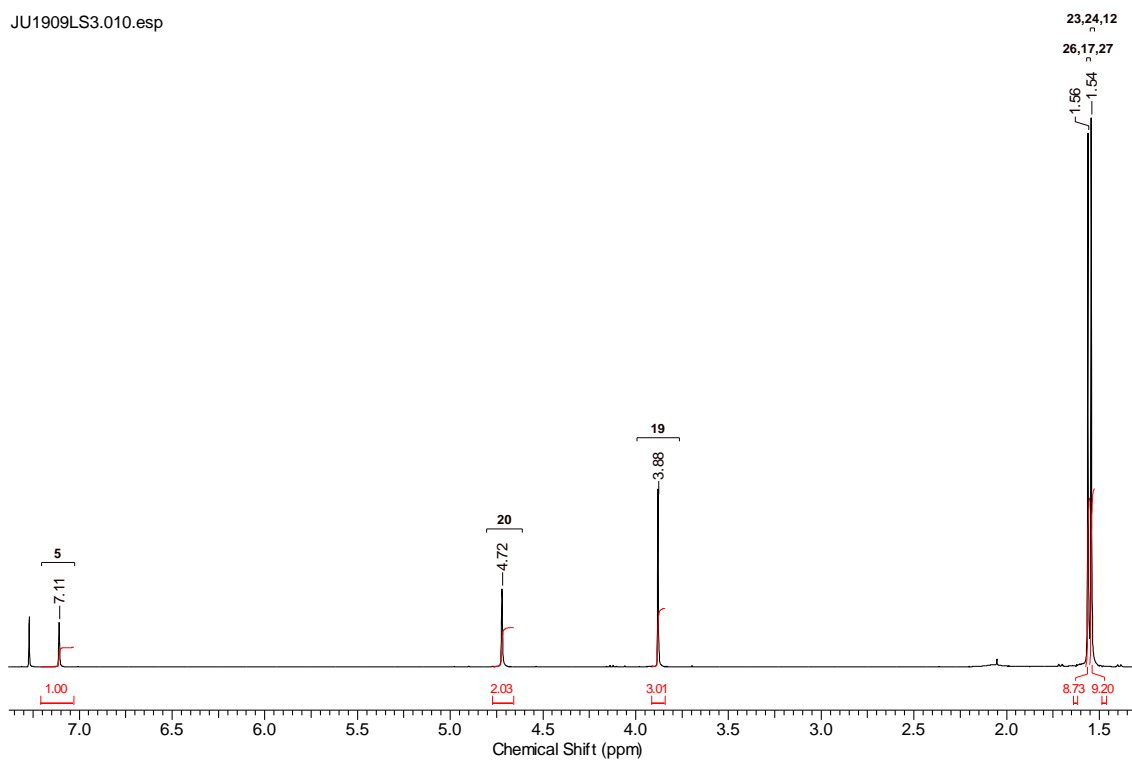
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 1.54 (9 H, s) 1.56 (9 H, s) 3.88 (3 H, s) 4.72 (2 H, s) 7.11 (1 H, s)

206

2-Bromo-3,4-diacetonitrile-5-methoxybenzaldehyde (2.101)

Comment	LS5529/1 F36-79		
Date	26 Mar 2009 16:42:40		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1849.8312	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2009\MA2609LS2\10\PDATA\1\1r

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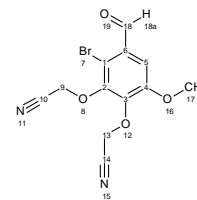
Formula $C_{12}H_9BrN_2O_4$
FW 325.1149
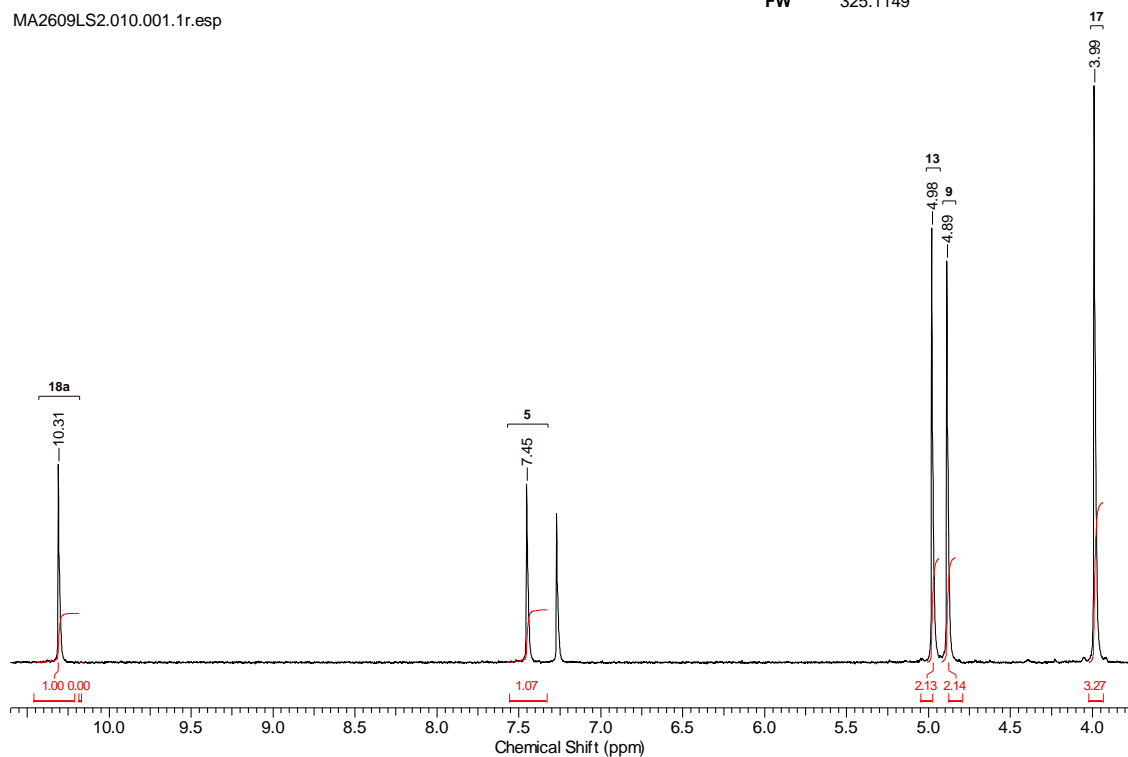
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.99 (3 H, s) 4.89 (2 H, s) 4.98 (2 H, s) 7.45 (1 H, s) 10.31 (1 H, s)

2-Bromo-3,4-diacetonitrile-5-methoxybenzyl alcohol (2.102)

Comment	LS5529/3	Date	27 Mar 2009 13:49:52
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	848.2556	Temperature (degree C)	27.000
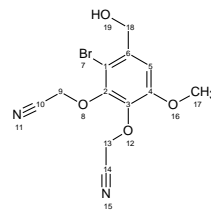
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MA2709LS1.010.esp

Formula $C_{12}H_{11}BrN_2O_4$
FW 327.1307
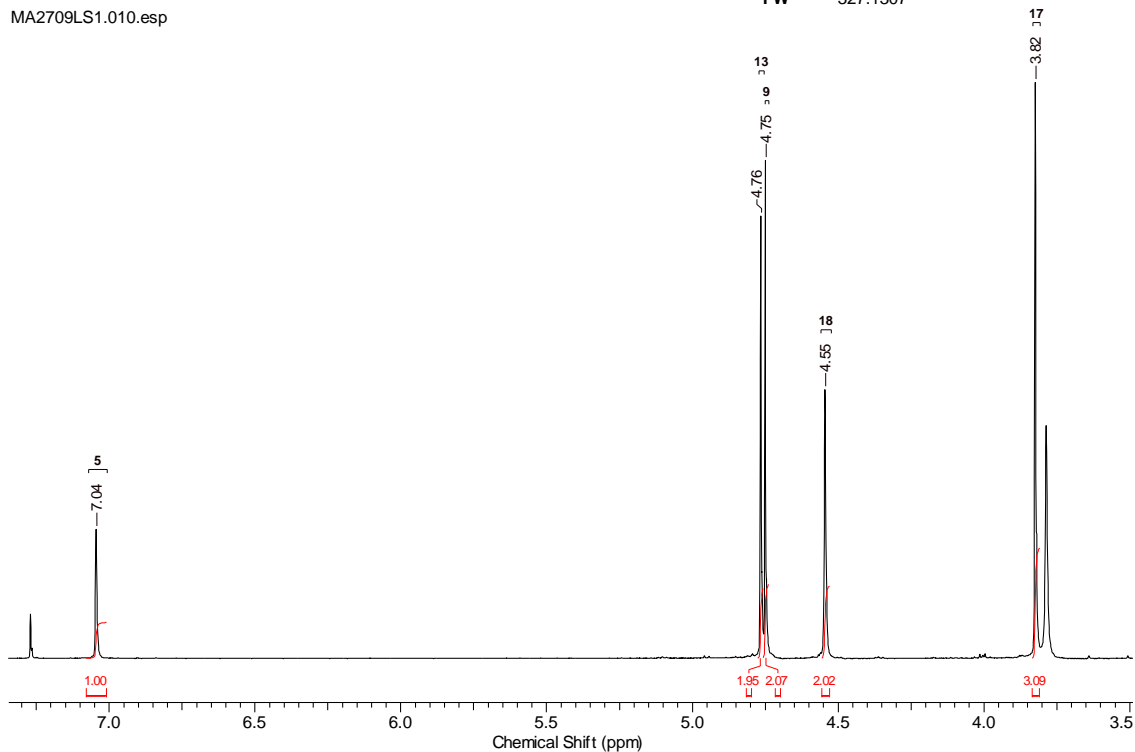
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.82 (3 H, s) 4.55 (2 H, s) 4.75 (2 H, s) 4.76 (2 H, s) 7.04 (1 H, s)

208

5-Bromo-4-formyl-2-methoxyphenyl acetate (2.104)

Comment	LS5291/58	Date	16 Dec 2008 19:31:12
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1497.4955	Temperature (degree C)	25.160
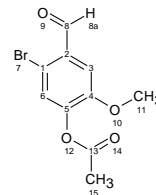
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Formula $C_{10}H_9BrO_4$ **FW** 273.0801
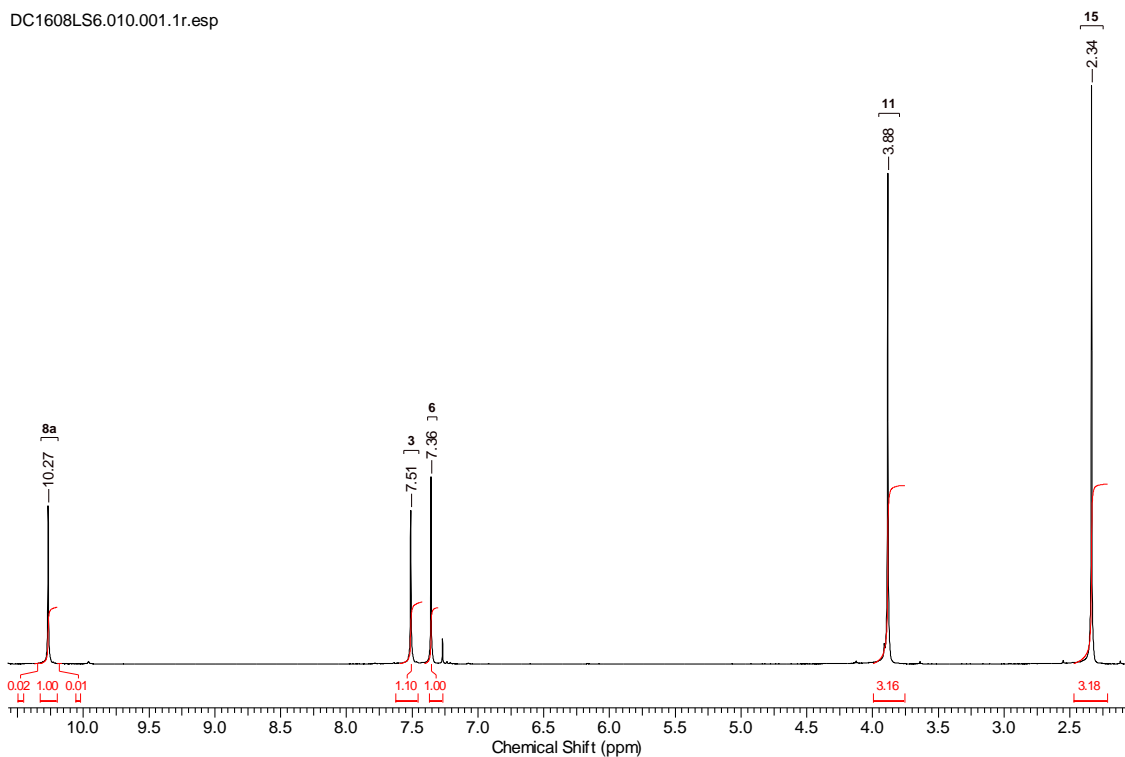
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.34 (3 H, s) 3.88 (3 H, s) 7.36 (1 H, s) 7.51 (1 H, s) 10.27 (1 H, s)

5-Bromo-4-(hydroxymethyl)-2-methoxyphenyl acetate (2.105)

Comment	LS5291/59 F10-15		
Date	19 Dec 2008 10:08:00		
Frequency (MHz)	300.13	**Nucleus**	1H
Number of Transients	16	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	1497.4954	**Temperature (degree C)**	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300 1\2008\DC1908LS1\10\PDATA\1\1r

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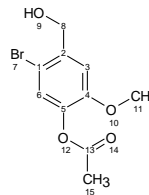
Formula $C_{10}H_{11}BrO_4$ **FW** 275.0959
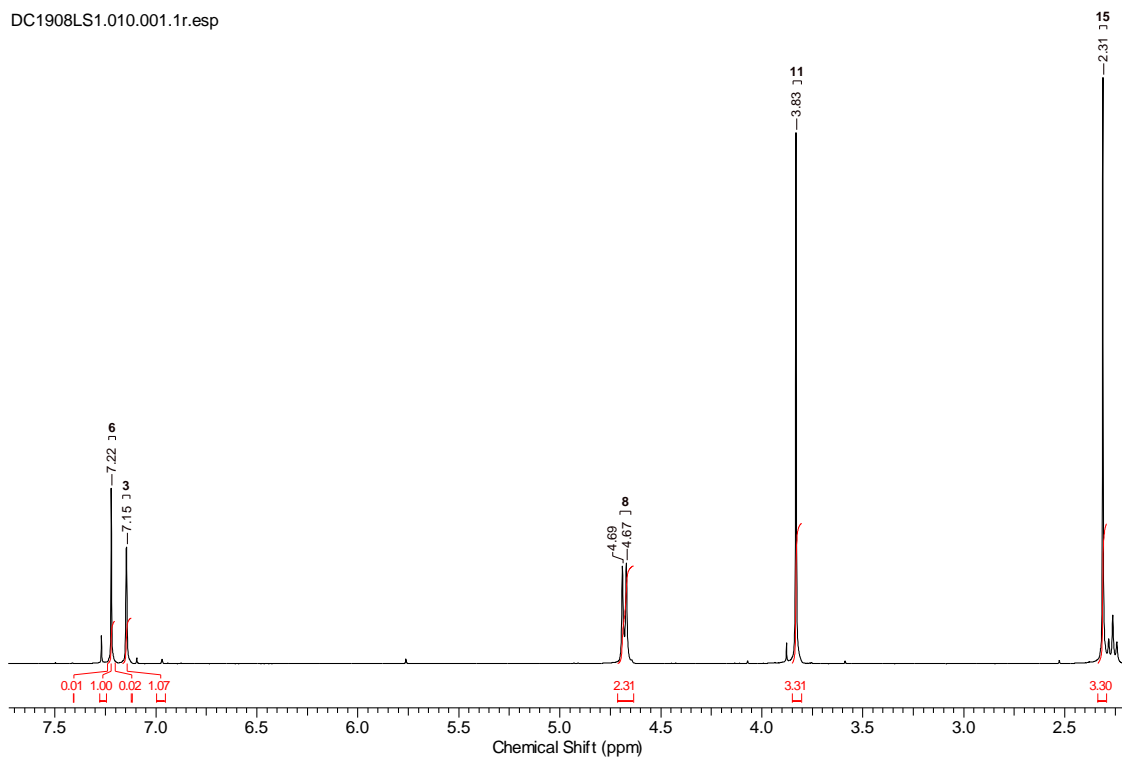
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.31 (3 H, s) 3.83 (3 H, s) 4.68 (2 H, d, *J*=5.9 Hz) 7.15 (1 H, s) 7.22 (1 H, s)
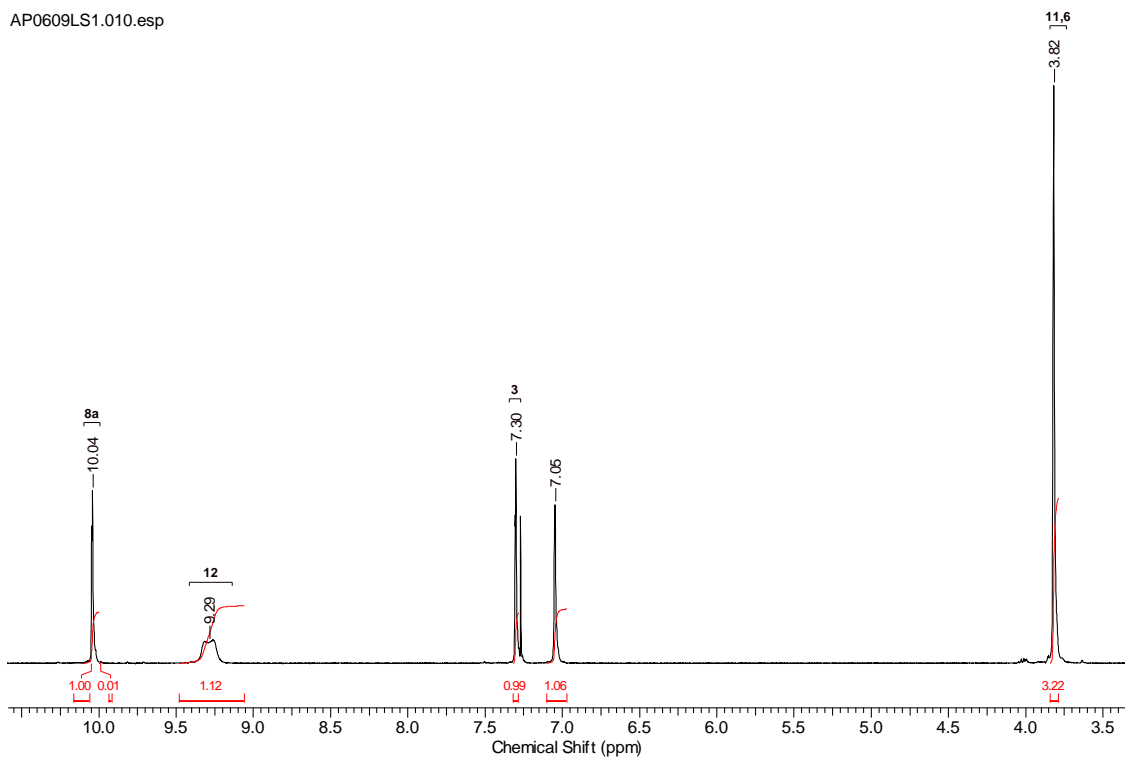
2-Bromo-4-hydroxy-5-methoxybenzaldehyde (2.106)

Comment	LS5529/13	Date	06 Apr 2009 20:13:52
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9382	Temperature (degree C)	27.000
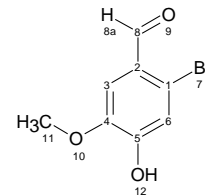
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
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AP0609LS1.010.esp

Formula $C_8H_7BrO_3$ FW   231.0434

^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.82 (3 H, s) 7.05 (1 H, s) 7.30 (1 H, s) 9.29 (1 H, br. s.) 10.04 (1 H, s)

2-(5-Bromo-4-formyl-2-methoxyphenoxy)acetonitrile (2.107)

Comment	LS5529/14 F8-12		
Date	07 Apr 2009 10:44:16		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
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AP0709LS1.010.esp
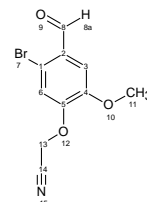
Formula $C_{10}H_8BrNO_3$
**FW** 270.0794
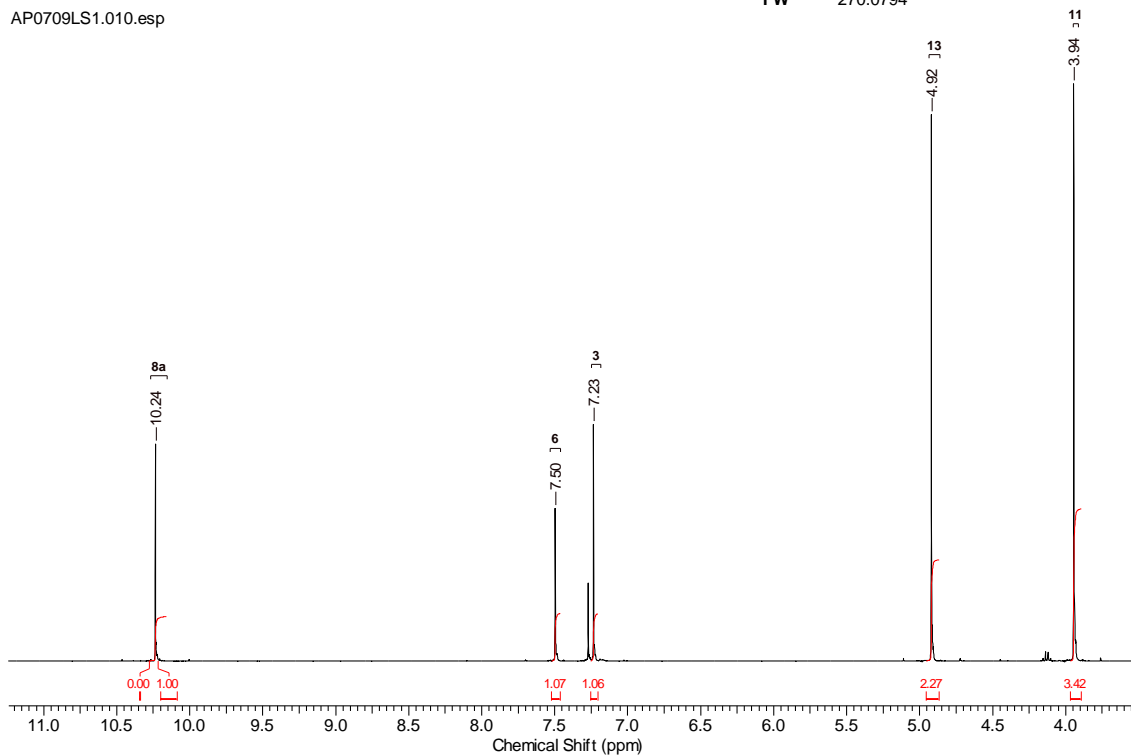
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.94 (3 H, s) 4.92 (2 H, s) 7.23 (1 H, s) 7.50 (1 H, s) 10.24 (1 H, s)

2-(5-Bromo-4-(hydroxymethyl)-2-methoxyphenoxy)acetonitrile (2.108)

Comment	LS5529/16 F44-60		
Date	23 Apr 2009 09:46:56		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.9613	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
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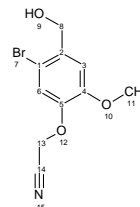
Formula C$_{10}$H$_{10}$BrNO$_3$
FW 272.0953
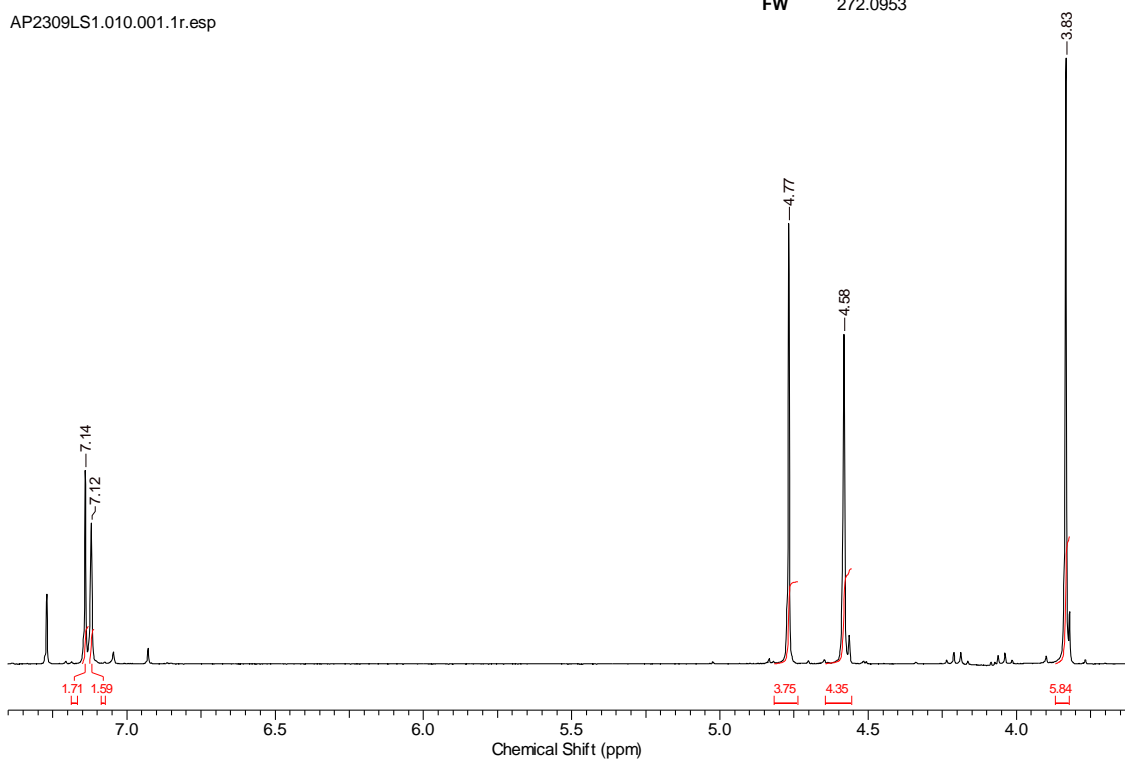
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.83 (3 H, s) 4.58 (2 H, s) 4.77 (2 H, s) 7.12 (1 H, s) 7.14 (1 H, s)

# 4-Hydroxy-2-methoxy-10-(trifluoromethyl)dibenzo[c,e]oxepin-5(7H)-one (2.112)

Comment	LS5529/79	Date	03 Sep 2009 19:41:52
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	64	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000
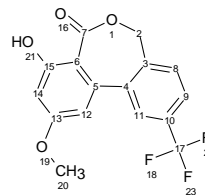
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SE0309LS6.010.esp

Formula $C_{16}H_{11}F_3O_4$ **FW**   324.2513
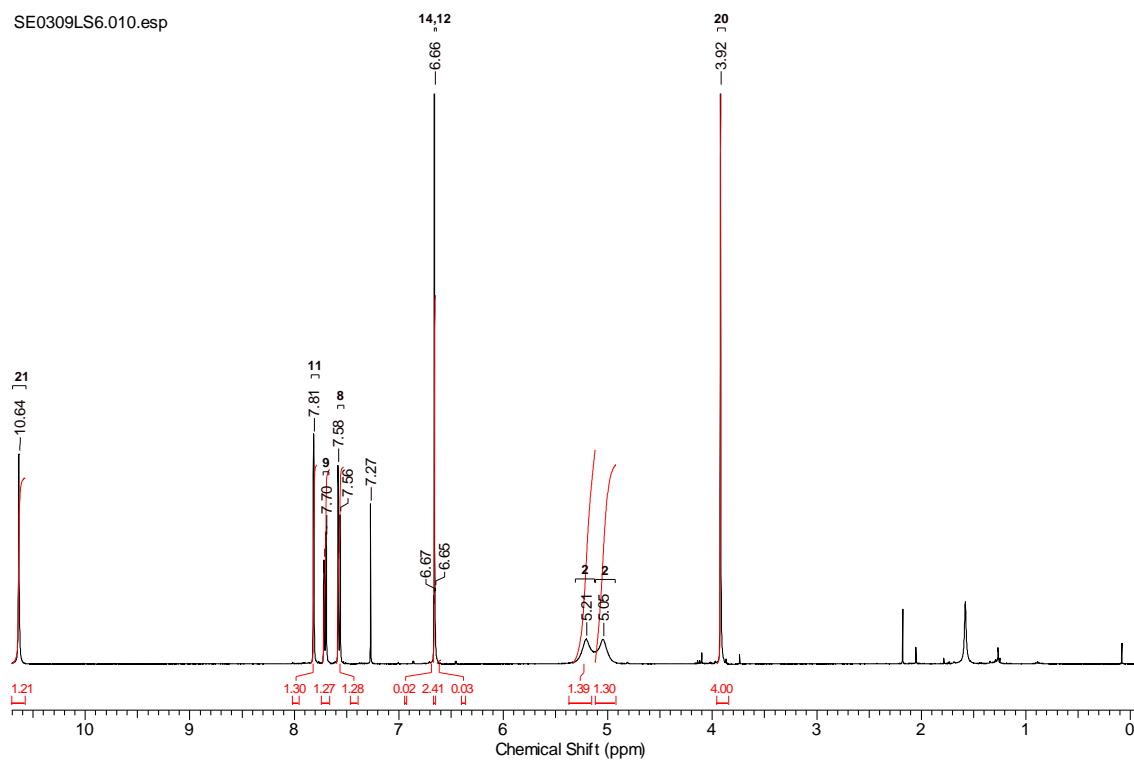

^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.92 (3 H, s) 5.05 (1 H, br. s.) 5.21 (1 H, br. s.) 6.65 - 6.67 (2 H, m) 7.57 (1 H, d, *J*=8.0 Hz) 7.71 (1 H, dd, *J*=7.8, 1.3 Hz) 7.81 (1 H, s) 10.64 (1 H, s)
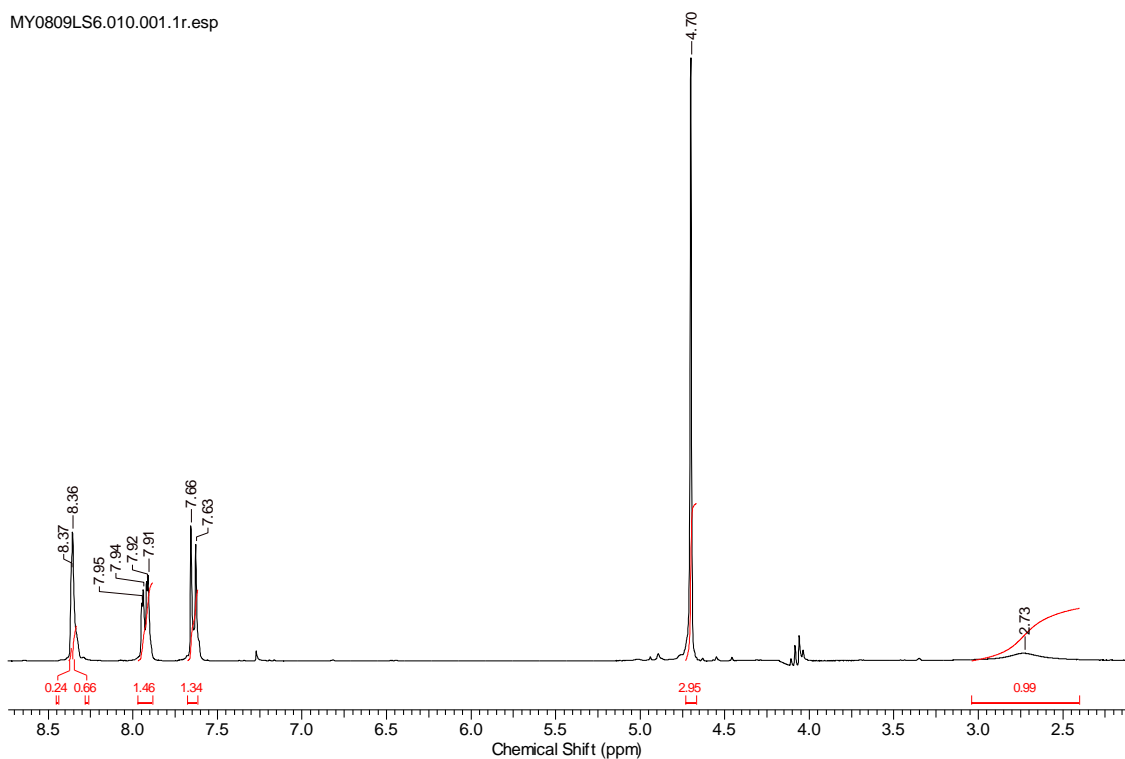
(2-Bromo-5-nitrophenyl)methanol (2.114)

Comment	LS5529/24	Date	08 May 2009 18:23:12
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.2078	Temperature (degree C)	25.100
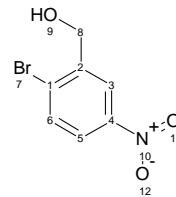
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2009\MY0809LS6\10\PDATA\1\1r

MY0809LS6.010.001.1r.esp

Formula $C_7H_6BrNO_3$ **FW**   232.0314

^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.73 (1 H, br. s.) 4.70 (2 H, s) 7.64 (1 H, d, *J*=8.7 Hz) 7.93 (1 H, dd, *J*=8.7, 2.6 Hz) 8.36 (1 H, d, *J*=2.6 Hz)

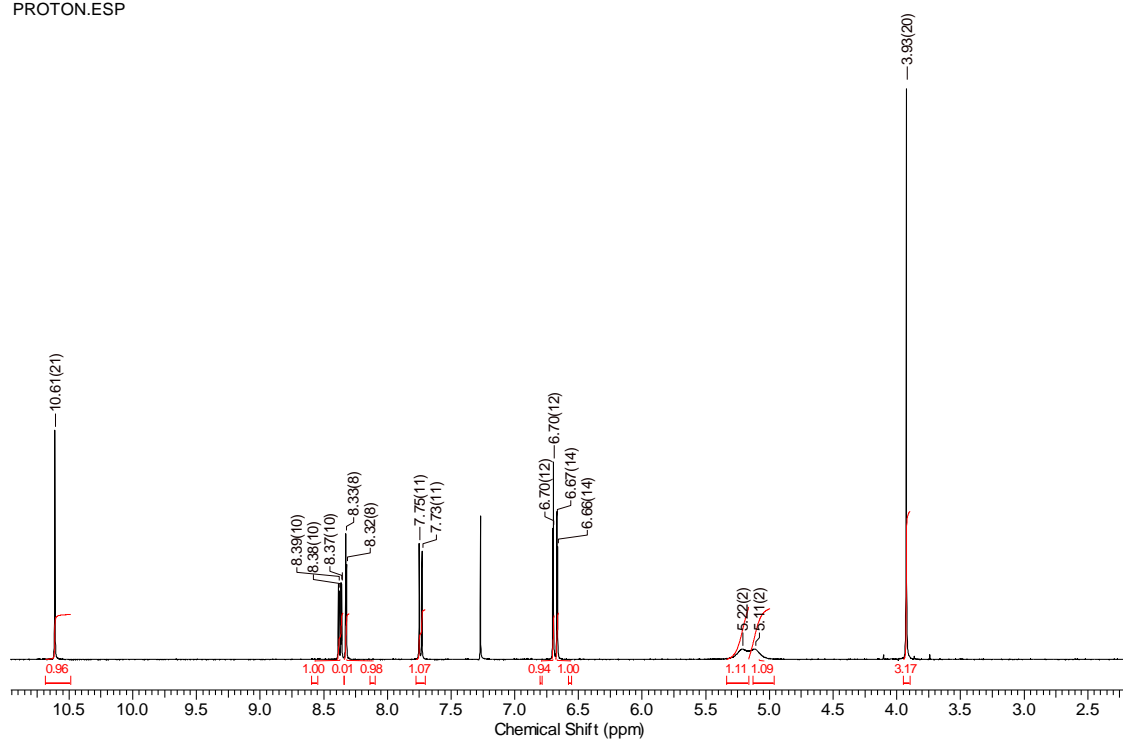# 4-Hydroxy-2-methoxy-9-nitrodibenzo[c,e]oxepin-5(7H)-one (2.115)

Comment	LS5529/90	Date	23 Sep 2009 16:17:04
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2453.4404	Temperature (degree C)	27.000
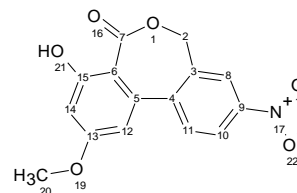
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400 1\SE2309LS1\10\FID

PROTON.ESP

Formula $C_{15}H_{11}NO_6$ FW   301.2509

^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.93 (3 H, s) 5.11 (1 H, br. s.) 5.22 (1 H, br. s.) 6.67 (1 H, d, *J*=2.5 Hz) 6.70 (1 H, d, *J*=2.5 Hz) 7.74 (1 H, d, *J*=8.5 Hz) 8.32 (1 H, d, *J*=2.5 Hz) 8.37 (1 H, dd, *J*=8.5, 2.5 Hz) 10.61 (1 H, s)
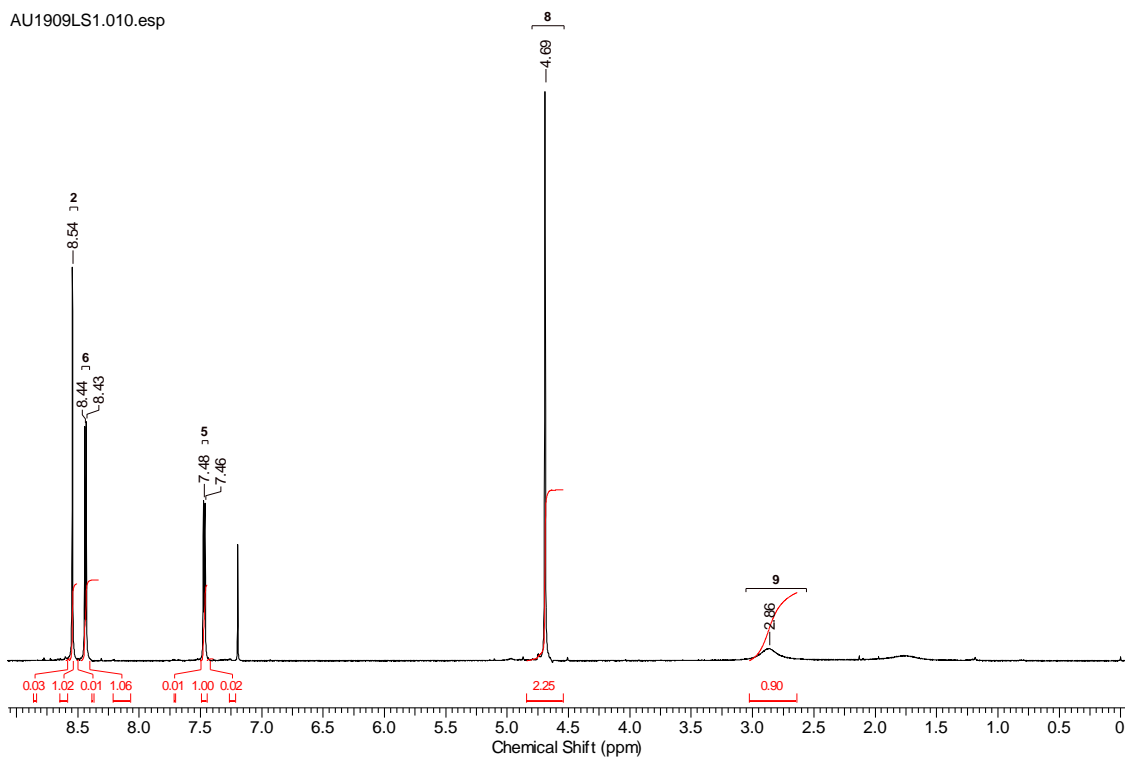
(3-Bromopyridin-4-yl)methanol (2.117)

Comment	LS5529/76	Date	19 Aug 2009 10:57:04
Frequency (MHz)	400.13	**Nucleus**	1H
Number of Transients	16	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	2423.5579	**Temperature (degree C)**	27.000
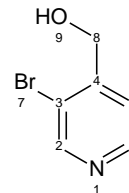
File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\AU1909LS1\10\FID

AU1909LS1.010.esp

Formula C_6H_6BrNO **FW** 188.0219

^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 2.86 (1 H, br. s.) 4.69 (2 H, s) 7.47 (1 H, d, *J*=5.0 Hz) 8.44 (1 H, d, *J*=5.0 Hz) 8.54 (1 H, s)

# 8-Hydroxy-10-methoxybenzo[5,6]oxepino[4,3-c]pyridin-7(5H)-one (2.118)

Comment	LS5529/81 F105+106		
Date	30 Oct 2009 15:25:52		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9387	Temperature (degree C)	27.000
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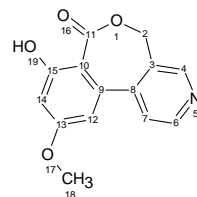
Formula $C_{14}H_{11}NO_4$ FW   257.2414

OC3009LS1.010.esp
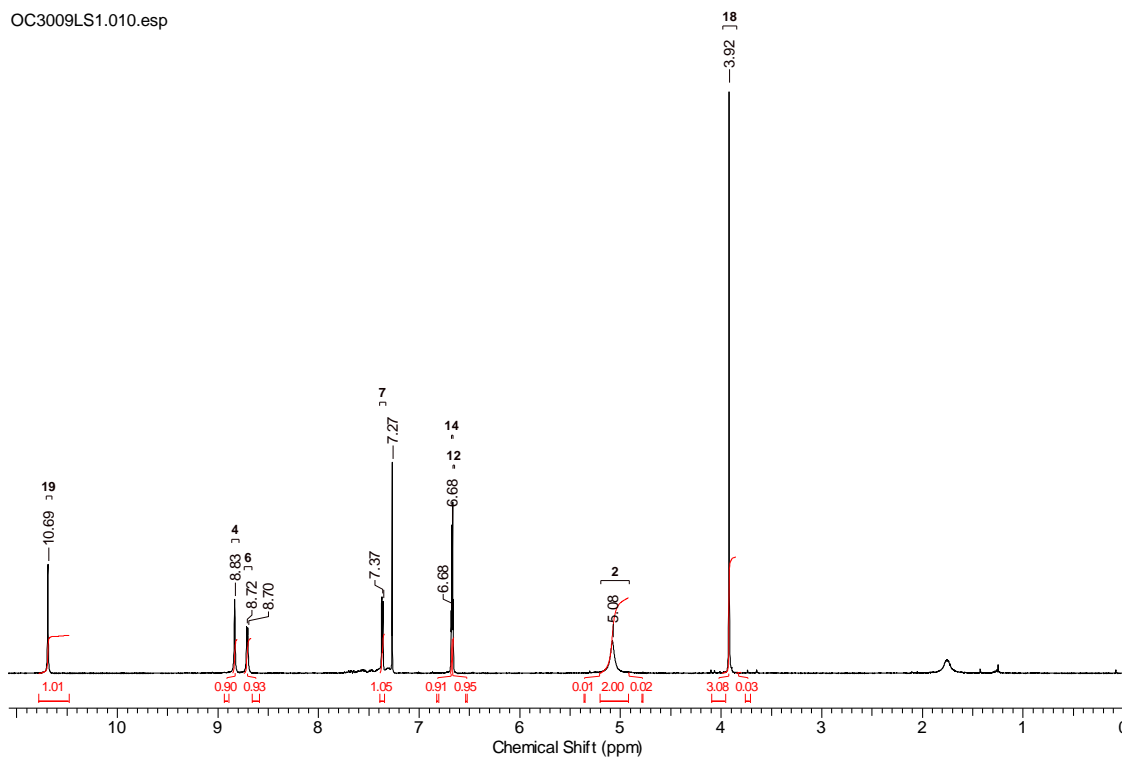
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.92 (3 H, s) 5.08 (2 H, br. s.) 6.67 (1 H, d, *J*=2.5 Hz) 6.68 (1 H, d, *J*=2.5 Hz) 7.37 (1 H, d, *J*=4.5 Hz) 8.71 (1 H, d, *J*=5.0 Hz) 8.83 (1 H, s) 10.69 (1 H, s)

10-Fluoro-4-hydroxy-2-methoxydibenzo[c,e]oxepin-5(7H)-one (2.120)

Comment	LS5529/78	Date	03 Sep 2009 23:15:12
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	128	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000

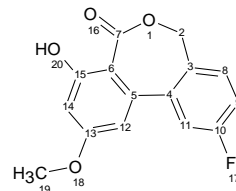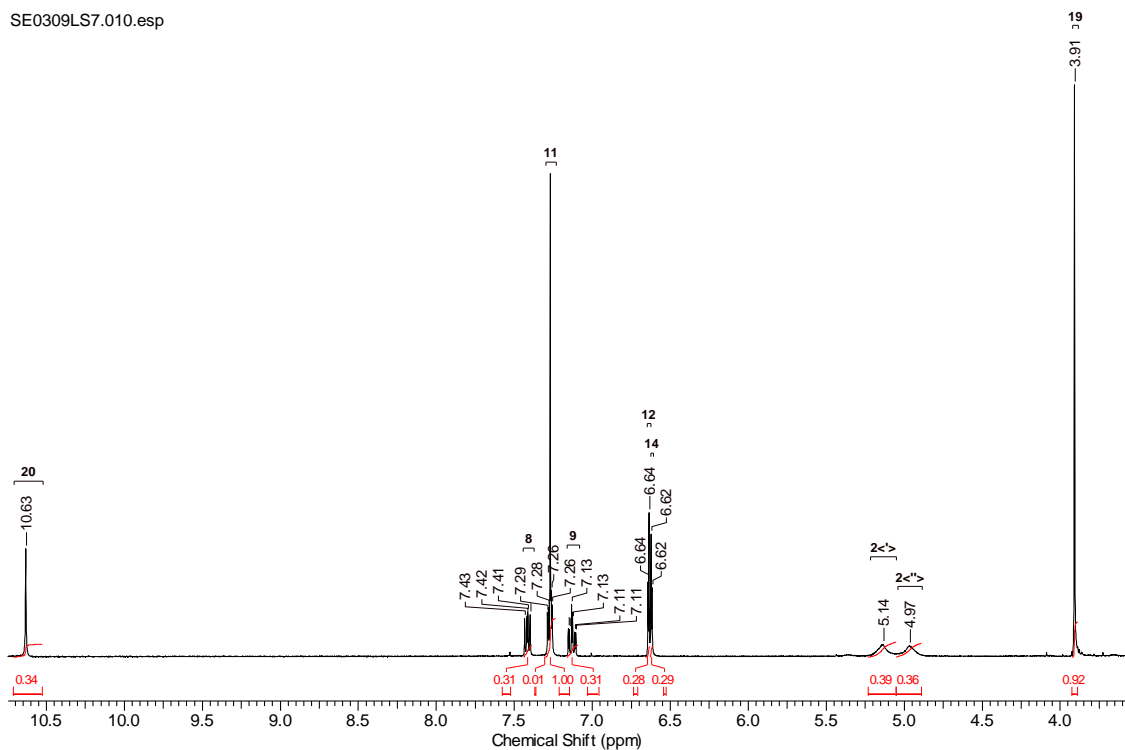File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\SE0309LS7\10\FID

SE0309LS7.010.esp

Formula $C_{15}H_{11}FO_4$ FW   274.2438

^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.91 (3 H, s) 4.97 (1 H, br. s.) 5.14 (1 H, br. s.) 6.62 (1 H, d, *J*=2.5 Hz) 6.64 (1 H, d, *J*=2.5 Hz) 7.13 (1 H, ddd, *J*=8.3, 2.5 Hz) 7.27 (1 H, dd, *J*=9.5, 2.5 Hz) 7.42 (1 H, dd, *J*=8.0, 5.5 Hz) 10.63 (1 H, s)
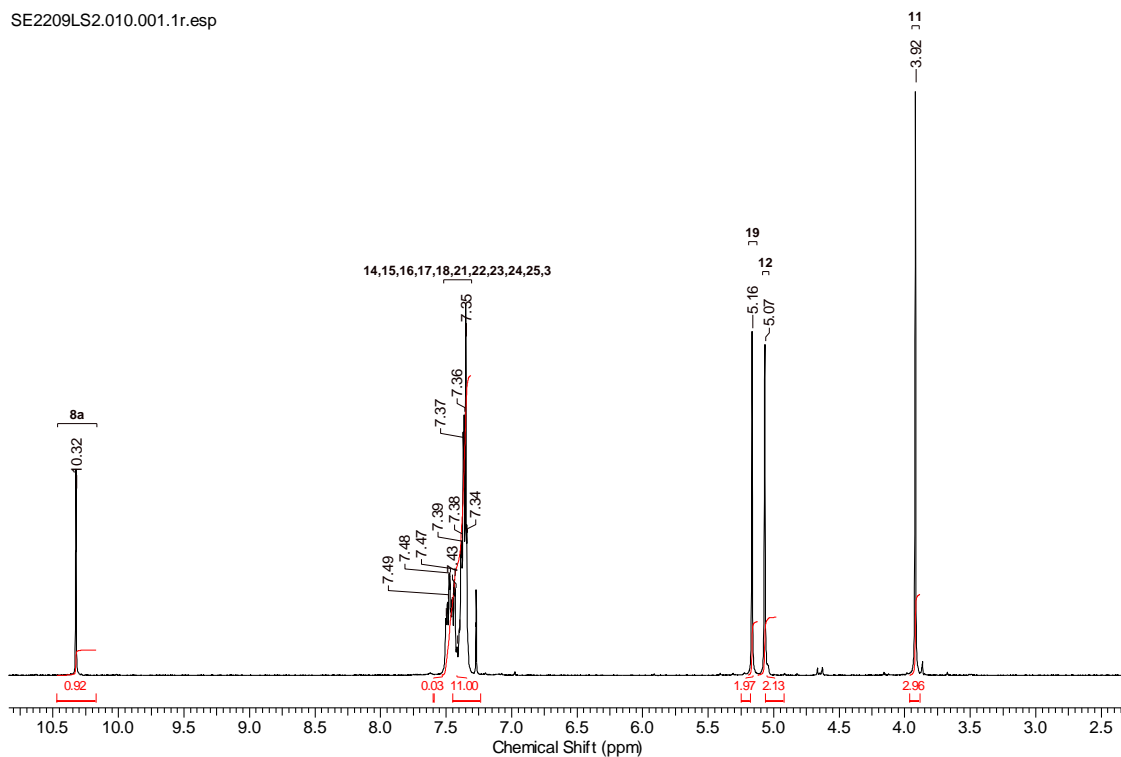
3,4-Bis(benzyloxy)-2-bromo-5-methoxybenzaldehyde (2.123)

Comment	LS5671/3 F19-36		
Date	22 Sep 2009 17:19:12		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.5845	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
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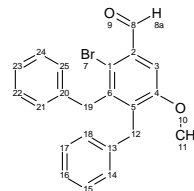
Formula $C_{22}H_{19}BrO_2$ FW 395.2891

^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.92 (3 H, s) 5.07 (2 H, s) 5.16 (2 H, s) 7.31 - 7.52 (11 H, m) 10.32 (1 H, s)

# (3,-Bis(benzyloxy)-2-bromo-5-methoxyphenyl)methanol (2.121)

Comment	LS5671/15	Date	06 Oct 2009 09:32:00
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.5846	Temperature (degree C)	25.000
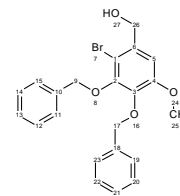
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2\2009\OC0609LS1\10\PDATA\1\1r

OC0609LS1.010.001.1r.esp

Formula $C_{22}H_{21}BrO_4$ **FW**   429.3037
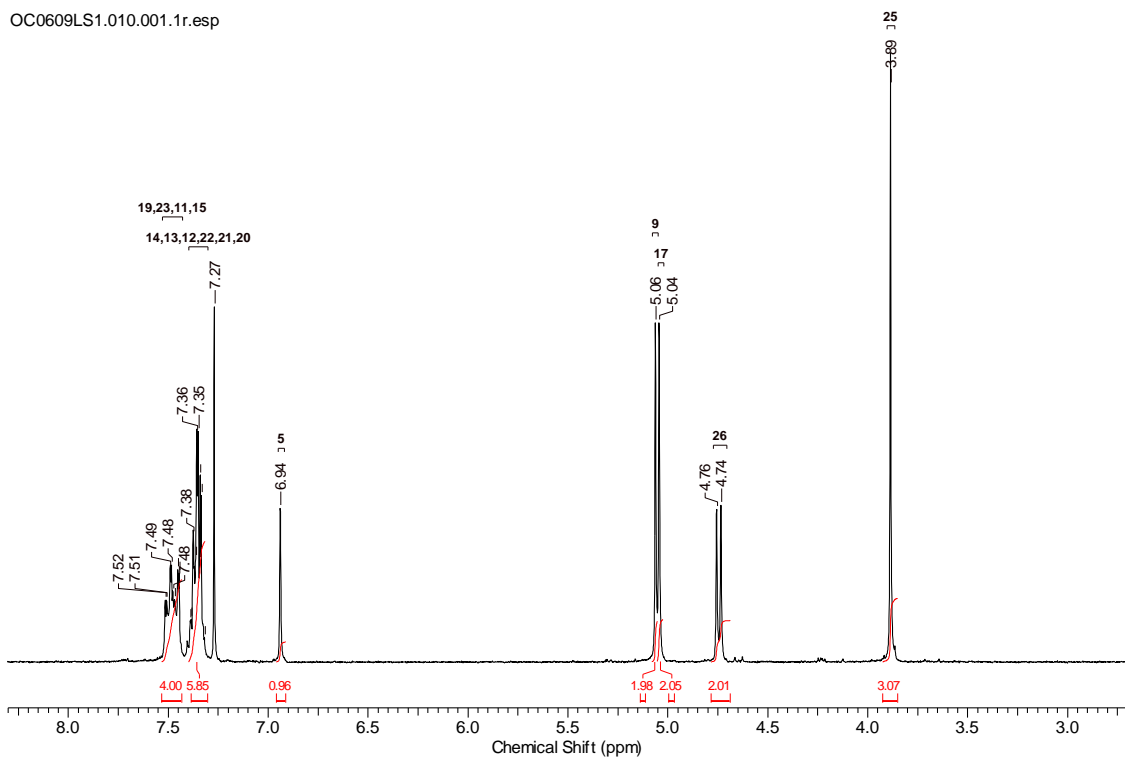
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.89 (3 H, s) 4.75 (2 H, d, *J*=6.0 Hz) 5.04 (2 H, s) 5.06 (2 H, s) 6.94 (1 H, s) 7.31 - 7.40 (6 H, m) 7.43 - 7.53 (4 H, m)

10,11-Bis(benzyloxy)-4-hydroxy-2,9-dimethoxydibenzo[c,e]oxepin-5(7H)-one (2.122)
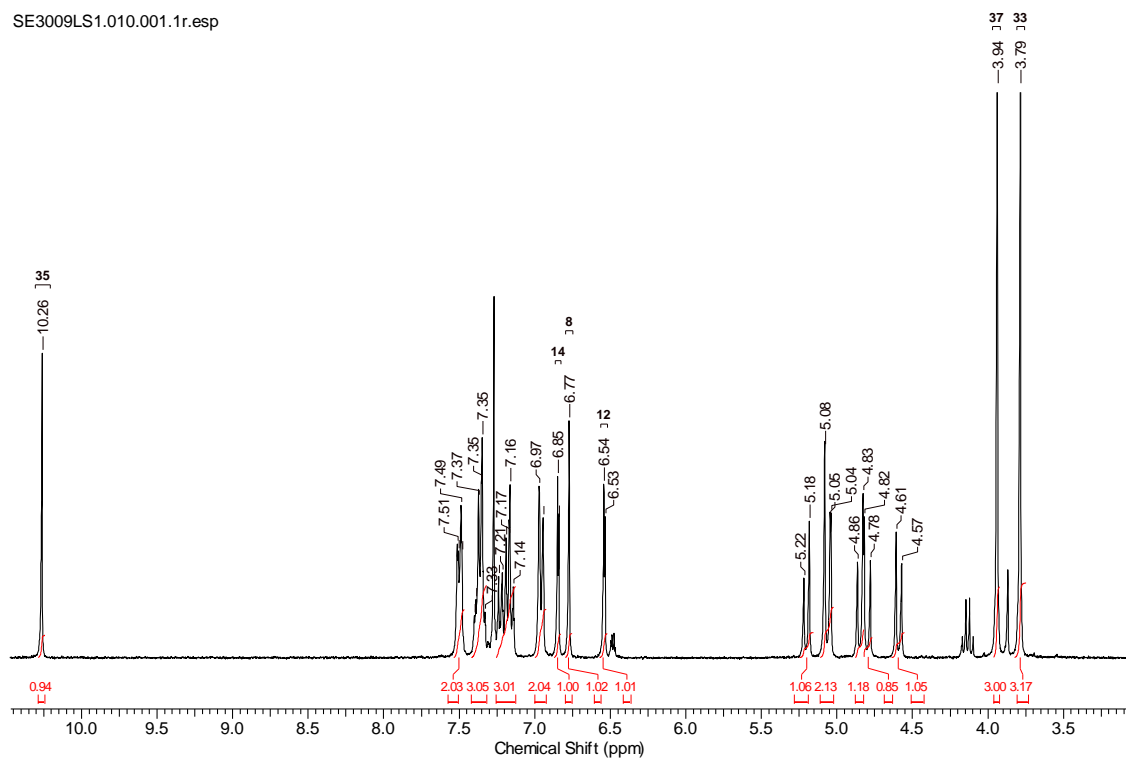
Comment	LS5671/10 F86-102		
Date	30 Sep 2009 14:07:12		
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.9615	Temperature (degree C)	25.000
File Name	C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300 2\2009\SE3009LS1\10\PDATA\1\1r		

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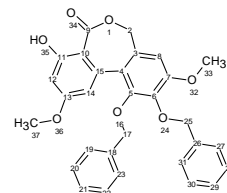
Formula $C_{30}H_{26}O_7$ **FW** 498.5232

^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 3.79 (3 H, s) 3.94 (3 H, s) 4.59 (1 H, d, *J*=10.9 Hz) 4.80 (1 H, d, *J*=12.1 Hz) 4.85 (1 H, d, *J*=10.9 Hz) 5.02 - 5.11 (2 H, m) 5.20 (1 H, d, *J*=10.5 Hz) 6.54 (1 H, d, *J*=2.6 Hz) 6.77 (1 H, s) 6.84 (1 H, d, *J*=2.6 Hz) 6.96 (2 H, d, *J*=7.5 Hz) 7.12 - 7.25 (3 H, m) 7.32 - 7.41 (3 H, m) 7.50 (2 H, dd, *J*=7.7, 2.1 Hz) 10.26 (1 H, s)

222

Ulocladol A (1.1)

Comment	LS5671/13	Date	03 Oct 2009 00:08:32
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\OC0209LS1\10\FID
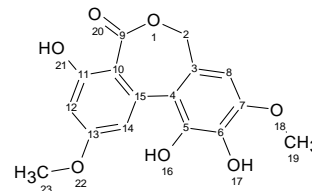
OC0209LS1.010.esp

Formula $C_{16}H_{14}O_7$ **FW**   318.2782
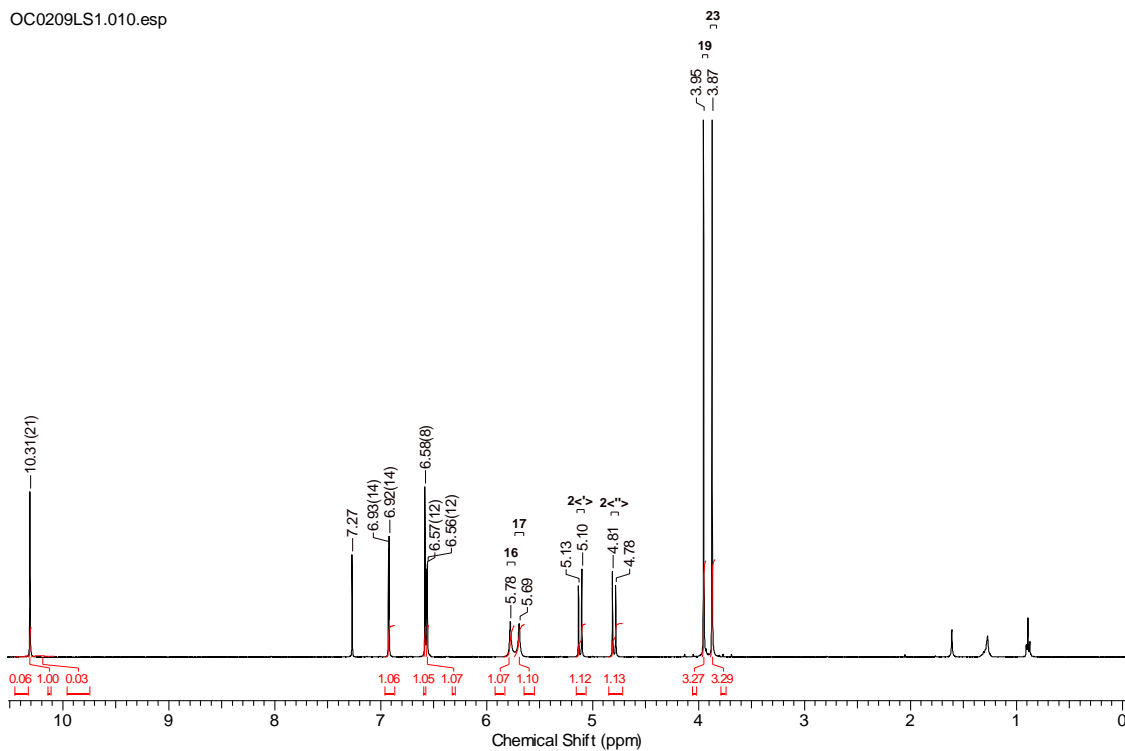
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.87 (3 H, s) 3.95 (3 H, s) 4.80 (1 H, d, *J*=12.0 Hz) 5.12 (1 H, d, *J*=12.0 Hz) 5.69 (1 H, br. s.) 5.78 (1 H, br. s.) 6.56 (1 H, d, *J*=3.0 Hz) 6.58 (1 H, s) 6.92 (1 H, d, *J*=2.5 Hz) 10.31 (1 H, s)

2,2-Dimethyl-4-oxo-4H-benzo[d][1,3]dioxin-5-yl trifluoromethanesulfonate (2.126)
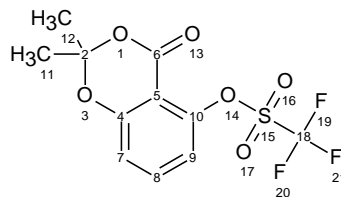

Comment	LS5671/16	Date	06 Oct 2009 09:36:16
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1849.8312	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2009\OC0609LS2\10\PDATA\1\1r

OC0609LS2.010.001.1r.esp

Formula $C_{11}H_9F_3O_6S$ **FW** 326.2458
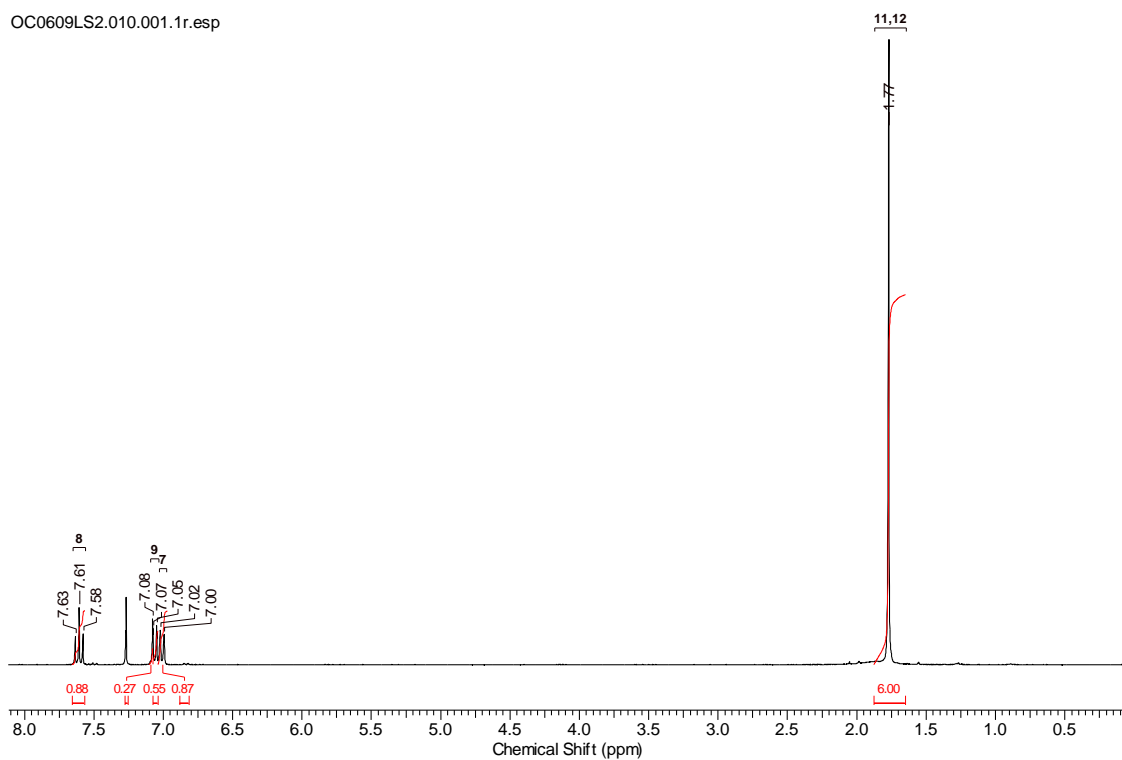
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 1.77 (6 H, s) 7.01 (1 H, d, *J*=7.9 Hz) 7.06 (1 H, dd, *J*=8.7, 1.1 Hz) 7.61 (1 H, t, *J*=8.3 Hz)

224

2,2-Dimethyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-4H-benzo[d][1,3]dioxin-4-one (2.127)

Comment	LS5671/17 F66-78		
Date	22 Oct 2009 00:04:16		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2452.9385	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\OC2109LS3\10\FID

OC2109LS3.010.esp
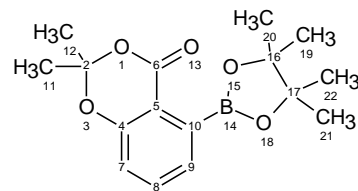
Formula $C_{16}H_{21}BO_5$ FW   304.1459
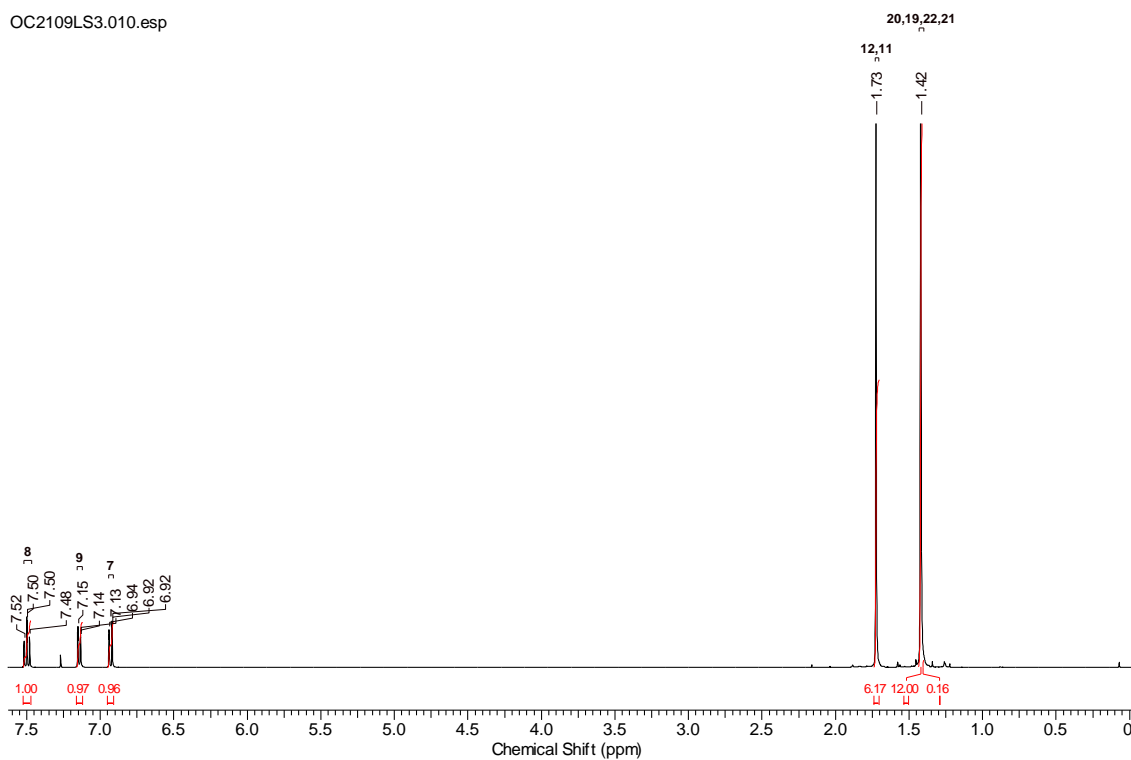
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 1.42 (12 H, s) 1.73 (6 H, s) 6.93 (1 H, dd, *J*=8.5, 1.0 Hz) 7.14 (1 H, dd, *J*=7.5, 1.0 Hz) 7.50 (1 H, dd, *J*=7.0 Hz)

10,11-Bis(benzyloxy)-4-hydroxy-9-methoxydibenzo[c,e]oxepin-5(7H)-one (2.128)

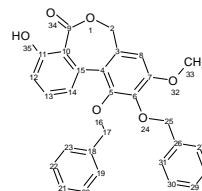| Comment	LS5671/19 F15-40		
Date	21 Oct 2009 21:34:56		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2414.7917	Temperature (degree C)	27.000
File Name	C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400 1\OC2109LS2\10\FID		

Formula C$_{29}$H$_{24}$O$_6$ FW   468.4973

OC2109LS2.010.esp
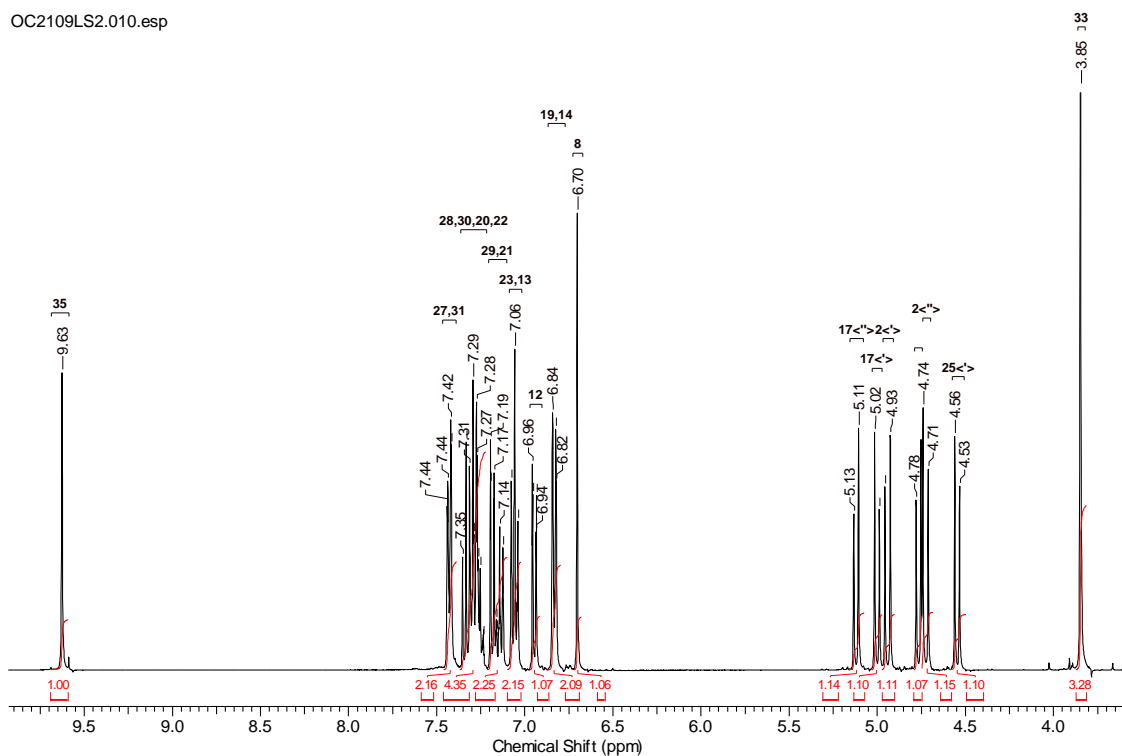
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.85 (3 H, s) 4.55 (1 H, d, *J*=11.0 Hz) 4.73 (1 H, d, *J*=12.0 Hz) 4.78 (1 H, d, *J*=11.0 Hz) 4.94 (1 H, d, *J*=12.0 Hz) 5.00 (1 H, d, *J*=11.0 Hz) 5.12 (1 H, d, *J*=11.0 Hz) 6.70 (1 H, s) 6.78 - 6.87 (2 H, m) 6.95 (1 H, dd, *J*=8.0, 1.0 Hz) 7.06 (2 H, t, *J*=7.3 Hz) 7.11 - 7.21 (2 H, m) 7.22 - 7.37 (4 H, m) 7.43 (2 H, dd, *J*=8.0, 1.5 Hz) 9.63 (1 H, s)

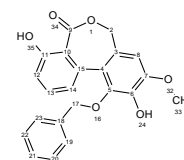# 11-(Benzyloxy)-4,10-dihydroxy-9-methoxydibenzo[c,e]oxepin-5(7H)-one (2.129)

Comment	LS5671/25 F15-28		
Date	30 Oct 2009 14:41:04		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	64	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2465.0759	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\OC3009NJWLS1\1\FID

Formula $C_{29}H_{24}O_6$ FW   468.4973

OC3009NJWLS1.001.esp
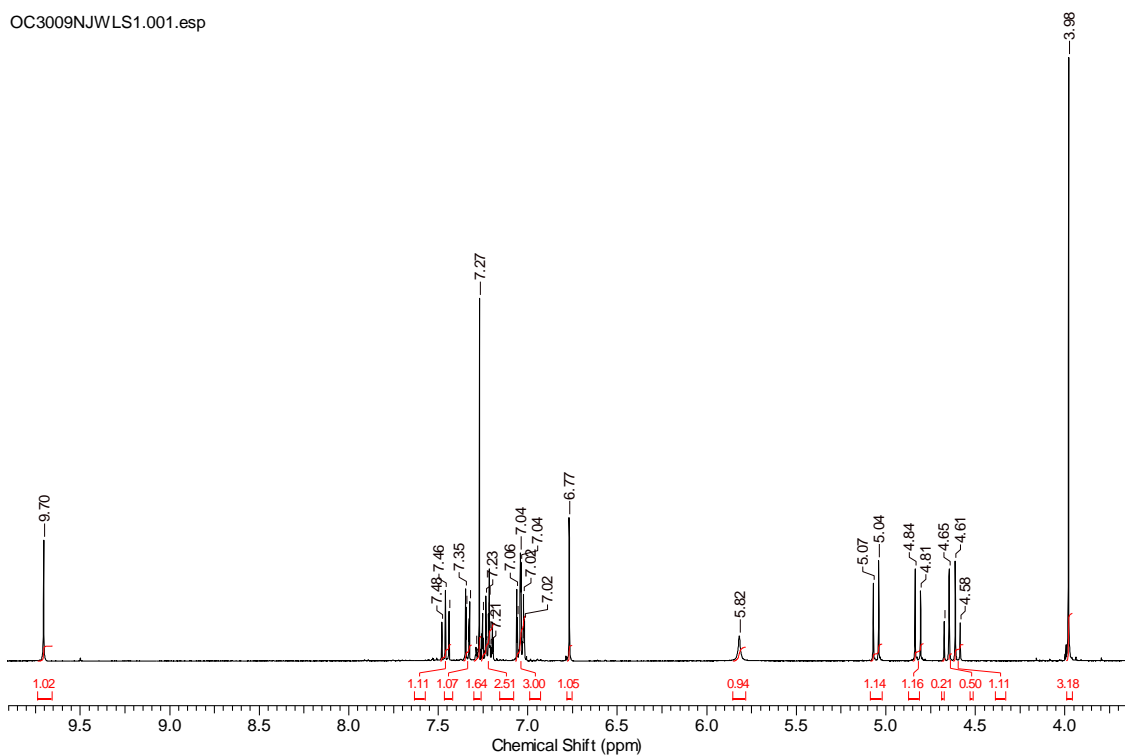
^{1}H NMR (400 MHz, *CHLOROFORM-d*) δ ppm 3.98 (3 H, s) 4.60 (1 H, d, *J*=11.1 Hz) 4.66 (1 H, d, *J*=11.1 Hz) 4.82 (1 H, d, *J*=12.1 Hz) 5.06 (1 H, d, *J*=12.1 Hz) 5.82 (1 H, s) 6.77 (1 H, s) 7.01 - 7.07 (3 H, m) 7.18 - 7.26 (2 H, m) 7.26 - 7.30 (1 H, m) 7.34 (1 H, dd, *J*=8.1, 1.5 Hz) 7.46 (1 H, t, *J*=7.8 Hz) 9.70 (1 H, s)

4-Bromo-5-methylbenzene-1,3-diol (3.4)

Comment	LS5291/18 F25-48 recrys		
Date	08 Sep 2008 16:53:36		
Frequency (MHz)	300.13	**Nucleus**	1H
Number of Transients	16	**Solvent**	Acetone
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	1496.5804	**Temperature (degree C)**	25.160

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300.1\300
1\2008\SE0808LS1\10\PDATA\1\1r

SE0808LS1.010.001.1r.esp
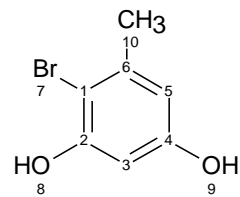
Formula $C_7H_7BrO_2$ **FW**   203.0333
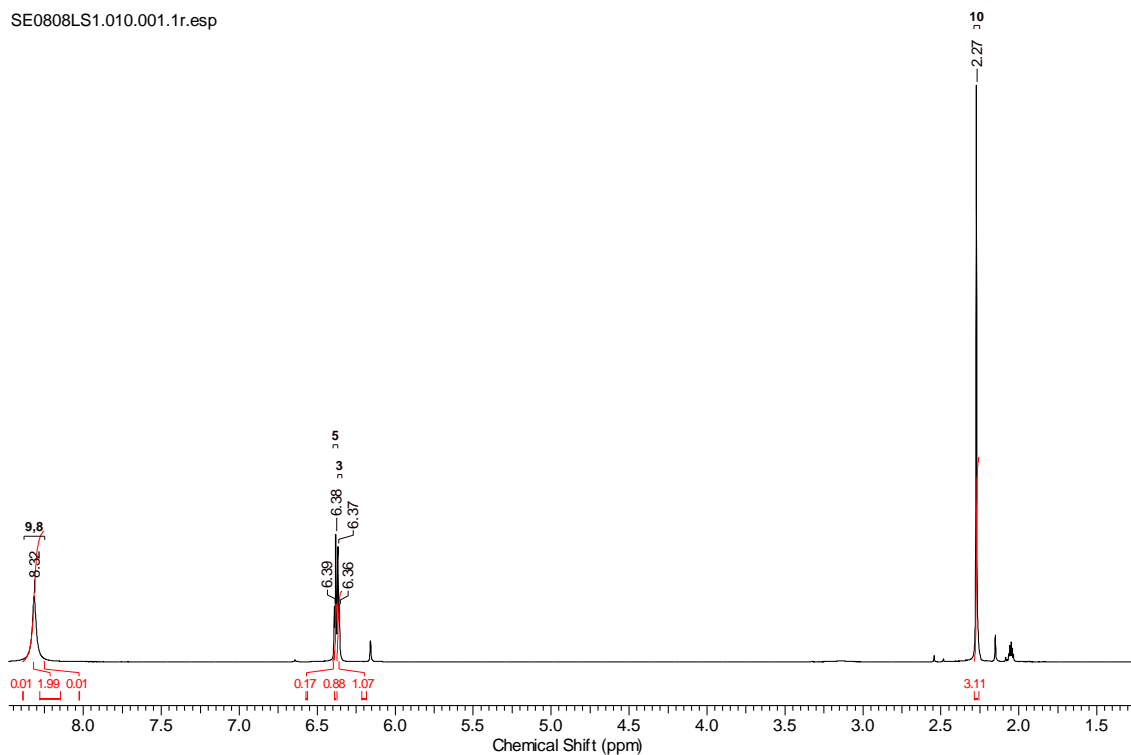
^{1}H NMR (300 MHz, *Acetone*) δ ppm 2.27 (3 H, s) 6.36 (1 H, d, *J*=2.9 Hz) 6.39 (1 H, d, *J*=2.9 Hz) 8.32 (2 H, br. s.)

228

4-Bromo-5-methyl-1,3-phenylene diacetate (3.5)

Comment	LS5529/86 F7-11		
Date	07 Sep 2009 15:13:20		
Frequency (MHz)	300.13	**Nucleus**	1H
Number of Transients	8	**Solvent**	CHLOROFORM-d
Original Points Count	16384	**Points Count**	16384
Spectrum Offset (Hz)	1850.5846	**Temperature (degree C)**	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300 2\2009\SE0709LS3\10\PDATA\1\1r

SE0709LS3.010.001.1r.esp

Formula $C_{11}H_{11}BrO_4$ **FW**   287.1066
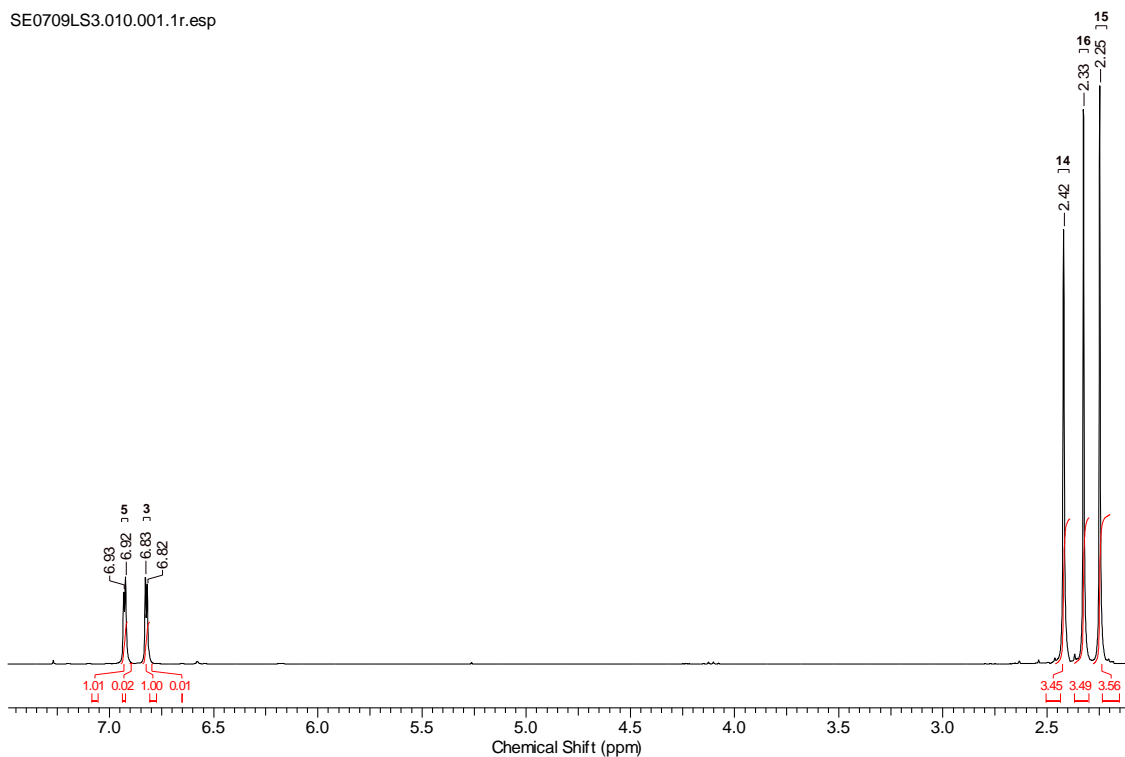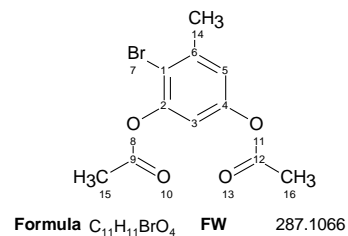
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.25 (3 H, s) 2.33 (3 H, s) 2.42 (3 H, s) 6.82 (1 H, d, *J*=3.0 Hz) 6.93 (1 H, d, *J*=3.0 Hz)

2,2'-((4-Bromo-5-methyl-1,3-phenylene)bis(oxy))diacetonitrile (3.6)

Comment	LS5529/18	Date	05 May 2009 17:29:52
Frequency (MHz)	300.13	Nucleus	1H
Number of Transients	8	Solvent	CHLOROFORM-d
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	1850.5844	Temperature (degree C)	25.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\300
2\2009\MY0509LS4\10\PDATA\1\1r

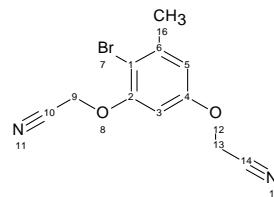MY0509LS4.010.001.1r.esp

Formula $C_{11}H_9BrN_2O_2$
FW 281.1054
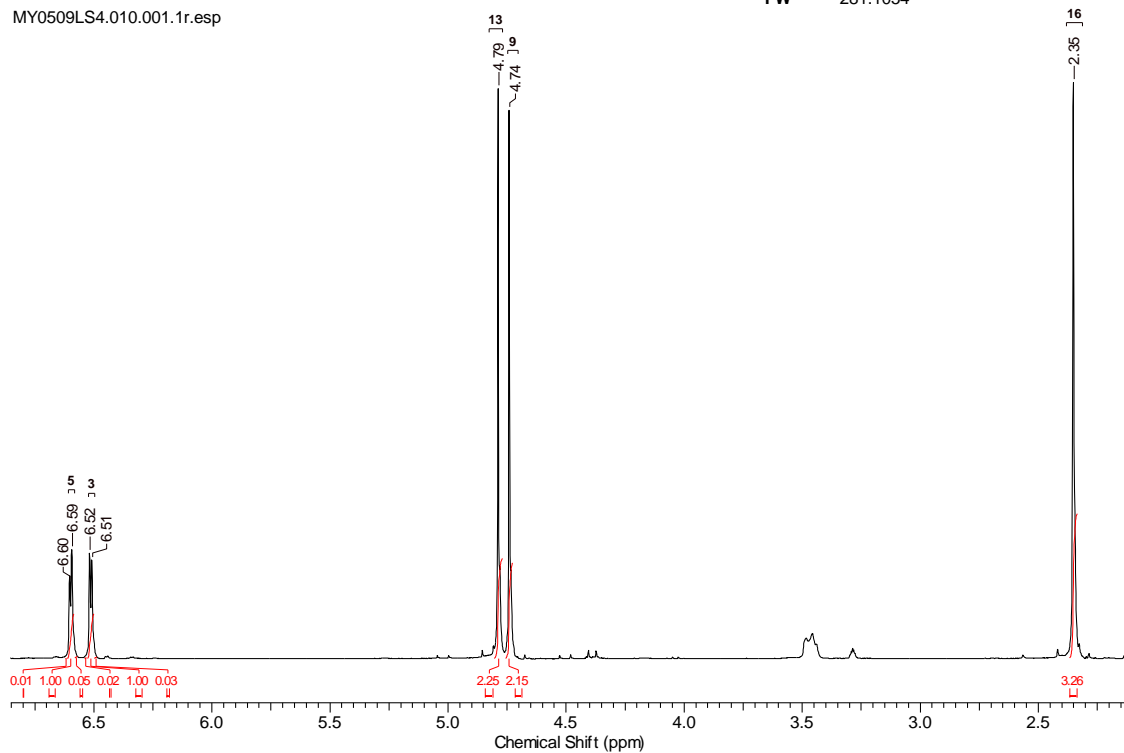
^{1}H NMR (300 MHz, *CHLOROFORM-d*) δ ppm 2.35 (3 H, s) 4.74 (2 H, s) 4.79 (2 H, s) 6.51 (1 H, d, *J*=3.0 Hz) 6.60 (1 H, d, *J*=3.0 Hz)

Alternariol-9-methyl ether (1.2)

Comment	LS 5671/8		
Date	02 Oct 2009 16:14:56		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	128	Solvent	DMSO-d6
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2442.7415		
Filename	C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\OC0209NJWLS1\1\FID		
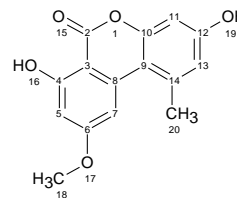
Formula $C_{15}H_{12}O_5$ **FW**   272.2528

OC0209NJWLS1.001.esp
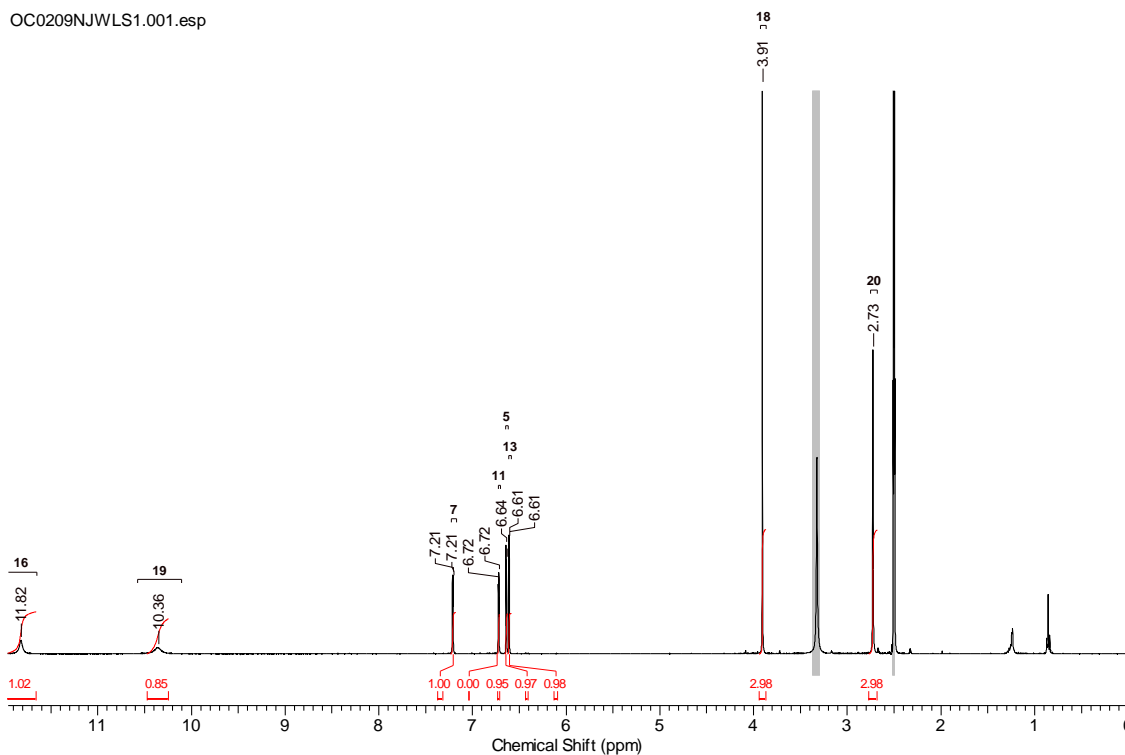
^{1}H NMR (400 MHz, *DMSO-d$_6$*) δ ppm 2.73 (3 H, s) 3.91 (3 H, s) 6.61 (1 H, d, *J*=2.0 Hz) 6.64 (1 H, d, *J*=2.4 Hz) 6.72 (1 H, d, *J*=2.0 Hz) 7.21 (1 H, d, *J*=2.4 Hz) 10.36 (1 H, br. s.) 11.82 (1 H, br. s.)

231

Alternariol (1.3)

Comment	LS5671/24 F27-34		
Date	21 Oct 2009 18:44:16		
Frequency (MHz)	400.13	Nucleus	1H
Number of Transients	16	Solvent	MeOH
Original Points Count	16384	Points Count	16384
Spectrum Offset (Hz)	2463.0674	Temperature (degree C)	27.000

File Name C:\DOCUMENTS AND SETTINGS\LAUREN\DESKTOP\NMR\400
1\OC2109LS1\10\FID

OC2109LS1.010.esp
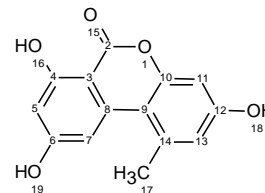
Formula $C_{14}H_{10}O_5$ **FW** 258.2262
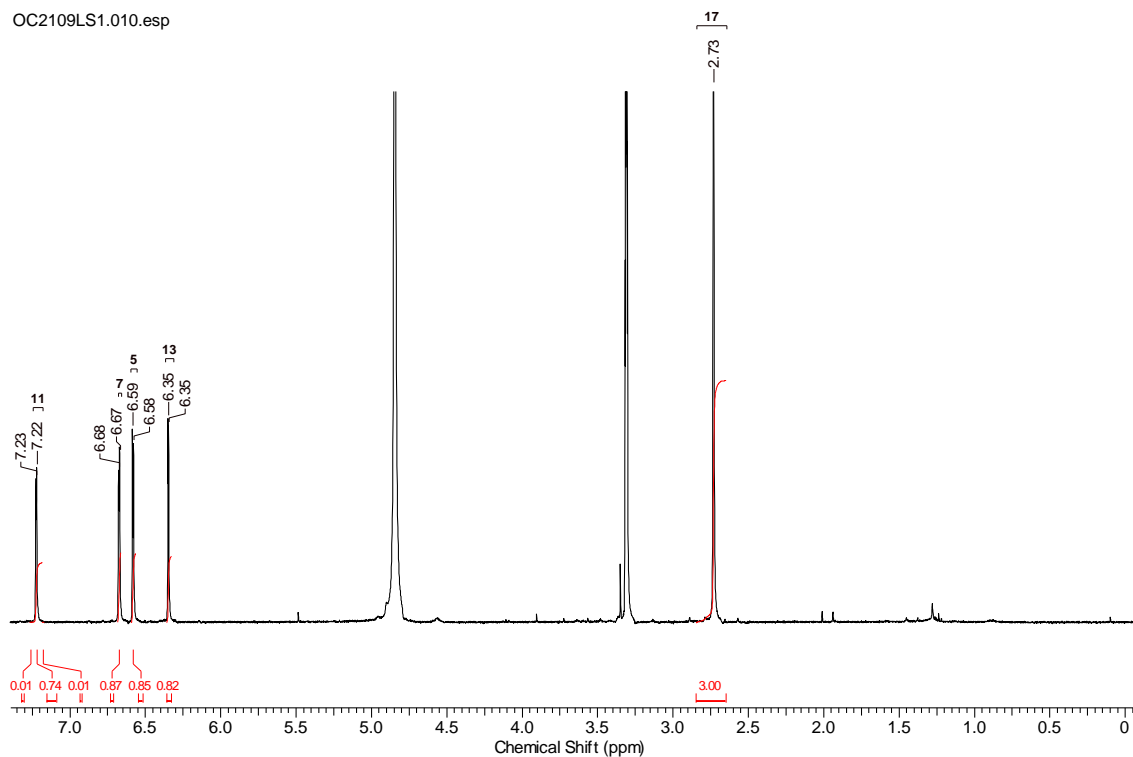
^{1}H NMR (400 MHz, *MeOH*) δ ppm 2.73 (3 H, s) 6.35 (1 H, d, *J*=2.0 Hz) 6.58 (1 H, d, *J*=2.5 Hz) 6.67 (1 H, d, *J*=2.5 Hz) 7.22 (1 H, d, *J*=2.0 Hz)

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9 References

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