UNIVERSITY OF SOUTHAMPTON

Faculty of Natural and Environmental Sciences

School of Chemistry

Towards the Synthesis of Marine Natural Products and their Analogues by Exploiting the Moore Rearrangement

by

Marc Geoffrey Radigois

Thesis for the degree of Doctor of Philosophy

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ABSTRACT

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Since its development in the 1980s, the Moore rearrangement has become a powerful and versatile tool in synthetic chemistry.[1] Mainly exploited in the synthesis of quinones, it can be used to access complex structures by careful manipulation of the reaction conditions. Herein is discussed its application in the synthesis of analogues of the natural product cribrostatin 6, as well as towards the first total synthesis of the pyrroloacridine alpkinidine.

Cribrostatin 6, isolated in 2003, has been shown to be active against seven cancer cell lines.[2] Following the procedure developed by Harrowven *et al.*,[3] cribrostatin 6 along with ten analogues were prepared for screening against the cancer cell line MCF-7. The results have shown the quinone unit to be amenable to change, with trends apparent in the cytotoxicity of these related compounds with respect to the nature of the substituents at position 8 and 9 on the cribrostatin 6 skeleton, their steric bulk and electronic properties. Further work is required to confirm these preliminary results in conjunction with exploring the effects of modifying the imidazopyridine unit.

With alpkinidine, synthetic work has looked to exploit this ring expansion in order to generate part of the fused pentacyclic ring system of this target natural product. Despite setbacks encountered along the way, the assembly of three of the five rings was achieved. Unfortunately, an inability to initiate the cascade of reactions required to furnish the last two rings of this acridine system halted progress towards alpkinidine. Although the target system could not be reached with the initial route developed, the Moore rearrangement still represents an interesting means of establishing part of this pyrroloacridine system. Should this prove not feasible, an alternative route based around an isoquinoline core has also been proposed.

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DECLARATION OF AUTHORSHIP

I, Marc Geoffrey Radigois, declare that this thesis and the work presented in it are my own and has been generated by me as the result of my own original research.

Towards the Synthesis of Marine Natural Products and their Analogues by Exploiting the Moore Rearrangement

I confirm that:

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2. 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 been clearly stated;
3. Where I have consulted the published work of others, this is always clearly attributed;
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Lastly, but by no means least, to my family and friends and, in particular, my Mum, my two sisters, Claire and Héloïse, thank you for your unwavering faith and support throughout the years. Words cannot describe how grateful I am for everything you have done for me. This PhD would definitely not have been possible without you!

Definitions and Abbreviations

Boc	*tert*-butoxycarbonyl
CAN	ceric ammonium nitrate
COSY	correlation spectroscopy
DCM	dichloromethane
DDQ	2,3-dichloro-5,6-dicyano-1,4-benzoquinone
DMAP	4-*N,N*-dimethylaminopyridine
DMF	*N,N*-dimethylformamide
DMSO	dimethylsulfoxide
DPPA	diphenylphosphoryl azide
EDC	1-ethyl-3-(3-dimethylaminopropyl)carbodiimide
EDG	electron donating group
EI	electron ionisation
equiv.	equivalent
ESI	electrospray ionisation
et al.	*et alii*
EWG	electron withdrawing group
FT	Fourier transform
g	grams
GI_{50}	concentration for 50% of maximal inhibition of cell proliferation
Hz	hertz
h	hours
HPLC	high-performance liquid chromatography
HRMS	high resolution mass spectrometry

IBX	2-iodoxybenzoic acid
IC_{50}	concentration causing 50% inhibition of the desired activity
IR	infrared
IPA	isopropyl alcohol
Kcal.	Kilocalorie
LHMDS	lithium hexamethyldisilazane
LRMS	low resolution mass spectrometry
mol	mole
MP	melting point
mL	millilitres
min	minutes
M	molar
MBC	minimum bactericidal concentration
MIC	minimum inhibitory concentration
MOM	methoxymethyl
NBS	*N*-bromosuccinimide
NMR	nuclear magnetic resonance
NOE	nuclear Overhauser effect
ROS	reactive oxygen species
RSM	recovered starting material
RT	room temperature
sat.	saturated (aqueous solution)
sp.	species
spp.	species pluralis
TBDMS	*tert*-butyldimethylsilyl

TFAA	trifluoroacetic anhydride
THF	tetrahydrofuran
TLC	thin phase chromatography
TMEDA	tetramethylethylenediamine
TMSCN	trimethylsilyl cyanide
UV	ultraviolet

Chapter 1: The Moore rearrangement

Cyclobutenone synthesis

Cyclobutenones are valuable synthons in organic chemistry, providing access to a variety of cyclic systems, a selection of which is depicted in Figure **1.1**.[1, 3-4] Consequently, their synthesis and chemistry has been the subject of much work. They are generally prepared from readily available squaric acid by converting it to a diester in yields ranging from 77-97% (Scheme **1.1**).[5] These are then reacted with organolithium reagents (alkynyl, aryl, heteroaryl and vinyl in nature) to generate the desired cyclobutenones **1.13**, **1.15** and **1.16** (Scheme **1.1**).

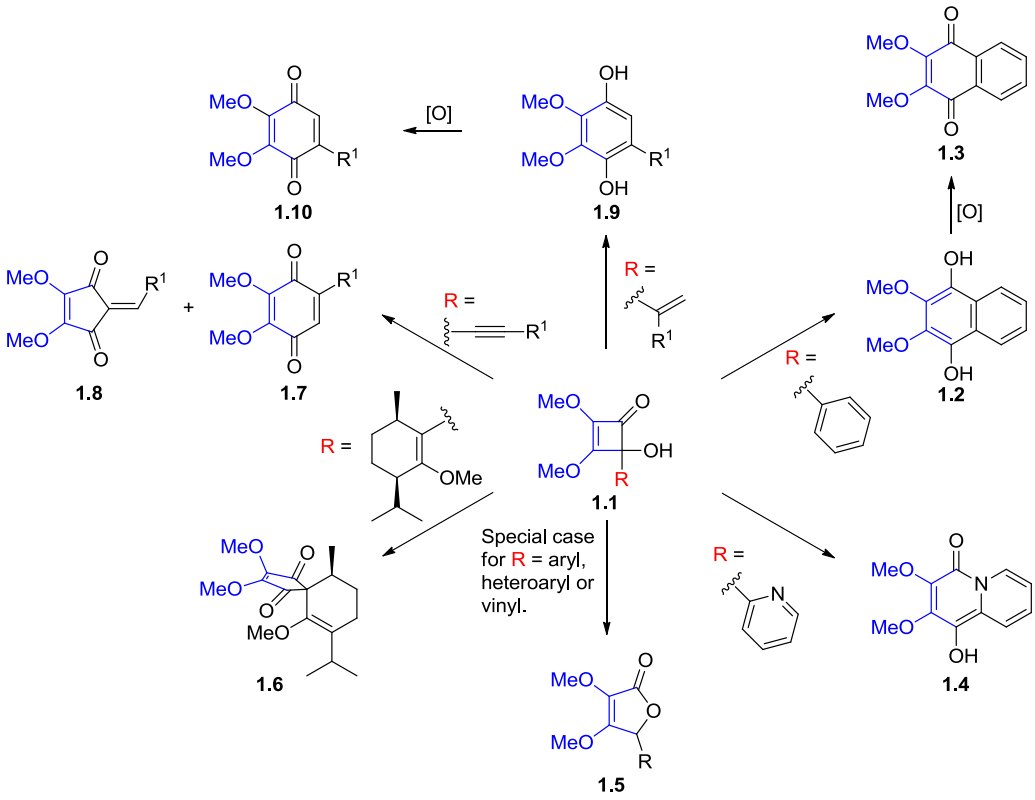
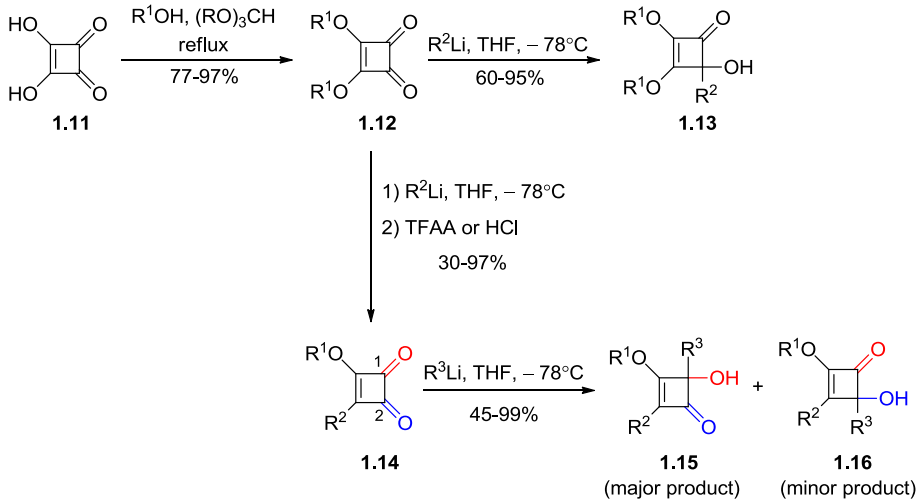


Figure 1.1: Array of products that can arise from the Moore rearrangement of cyclobutenones.[3-4, 4k, 4l]

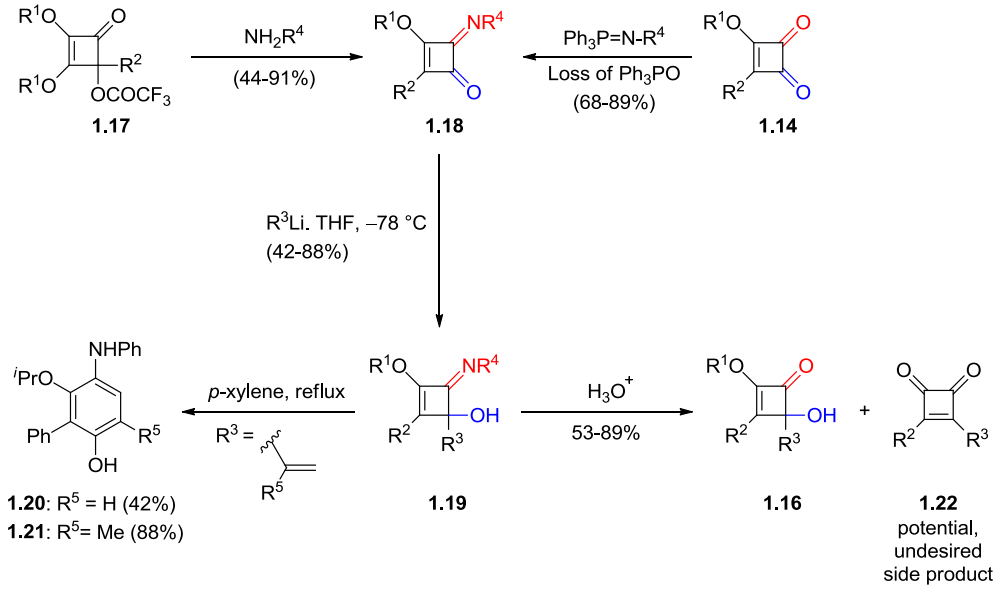
The control of substituent patterns afforded by the Moore rearrangement and the products it can access has made it an attractive area of research. The presence of two different carbonyl groups in the cyclobutenedione precursors could complicate the synthesis of cyclobutenones as, in theory, nucleophilic addition can occur to either carbonyl leading to a mixture of products **1.15** and **1.16**. In practice, for substrates such as **1.14** addition generally favours the ketonic carbonyl (C1 carbonyl in Scheme **1.1**) over the vinylogous

ester carbonyl (C2 carbonyl in Scheme **1.1**), with cyclobutenone **1.15** the major product isolated.[4c, 4d, 6]



Scheme 1.1: Synthesis of hydroxycyclobutenones from squaric acid.[5]

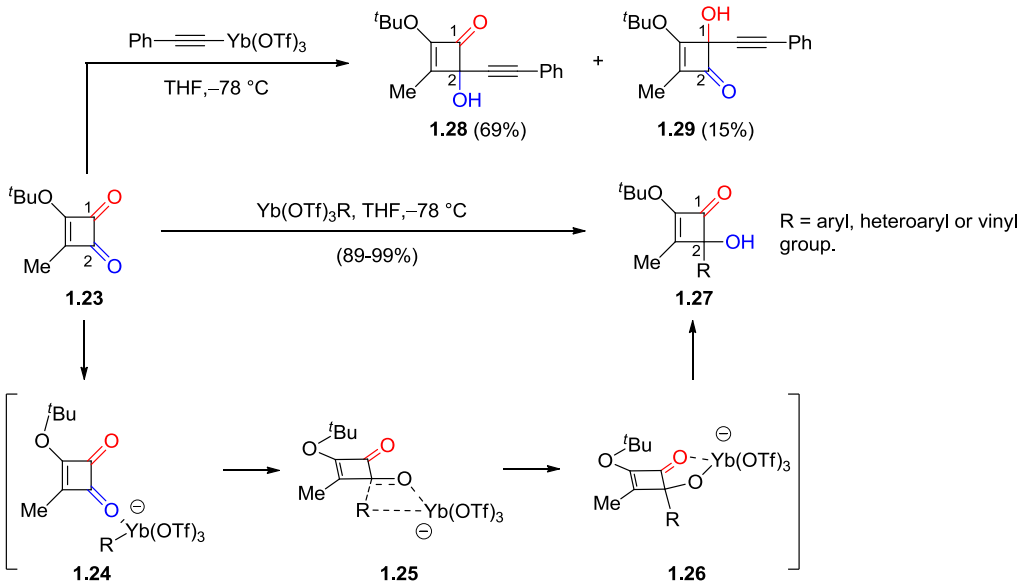
Access to cyclobutenone **1.16** is still possible with two strategies having been developed to achieve this.[6a, 6c, 7] The first of these masks the more reactive carbonyl as an imine, a step that gives yields in the range of 44–91% (Scheme **1.2**). Addition then proceeds to the vinylogous ester carbonyl and is followed by deprotection to generate the desired cyclobutenone **1.16** (Scheme **1.2**).



Scheme 1.2: Synthesis of cyclobutenimine.[4d, 6c, 7-8]

Indeed, it is the deprotection step that limits the effectiveness of this strategy as it uses acid to unmask the cyclobutenone. This is problematic as the newly generated tertiary alcohol in **1.16** is acid labile and readily gives cyclobutenedione **1.22** as by-product on treatment with mineral acid.

The alternative methodology employs ytterbium ate complexes to add to the less reactive carbonyl (Scheme **1.3**). This is achieved by firstly generating the organoytterbium species through reaction of the desired organolithium reagent with $Yb(OTf)_3$ at −78 °C, before adding cyclobutedione **1.23** to the reaction mixture. With this methodology, near quantitative yields (89-99%) of cyclobutenones **1.27** are obtained without the need for deprotection.[6a] One exception to the observed regioselectivity occurs with phenyl acetylene. Here, addition to the vinylogous ester carbonyl is still favoured with **1.28** formed in 69%, however addition to the ketonic carbonyl is also observed with **1.29** obtained in 15% yield (Scheme **1.3**).



Scheme 1.3: Addition to vinylogous ester carbonyl using $Yb(OTf)_3$.[6a]

Based on a computational study of the reaction, the ate complex $[Yb(OTf)_3R]^-$ is first believed to bind with the more Lewis basic C2 carbonyl in cyclobuteneone **1.23** (Scheme **1.3**).[6a] Complexation at C1 is possible but requires the ^{t}Bu group to rotate away from the ytterbium moiety at a penalty estimated to be 2.6 kcal.mol^{-1}. Indeed, this steric influence seems to be significant in determining the outcome of the addition as, when a methoxy group is used instead of a ^{t}butoxy group, a near 1 : 1 ratio of C1 and C2 adducts is formed. Thus, this steric requirement is necessary for this methodology to work effectively. As it is incorporated within a protecting group that can easily be removed or replaced by other functionalities, this is of little consequence.

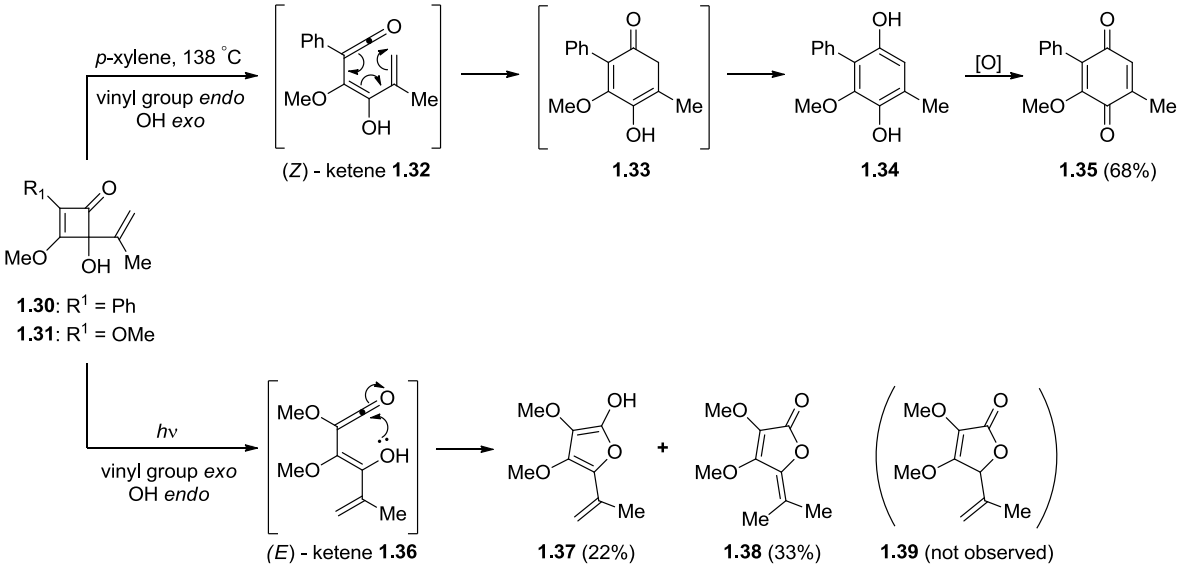
The Moore rearrangement

The ring expansion of cyclobutenones is a valuable tool for synthesising densely substituted cyclic systems in high yield, which would otherwise be difficult to achieve by conventional means.[3] Developed independently in the 1980s by both Moore and co-

workers and Liebeskind and co-worker, it involves an initial electrocyclic ring opening to form a reactive ketene.[1, 4a-c, 4f, 9] Ketene formation can be achieved either through the use of heat or via irradiation with light. The course of the reaction is then determined by the nature of the unsaturated substituent R present on the cyclobutenone ring (**1.1**) as demonstrated in Figure **1.1**.

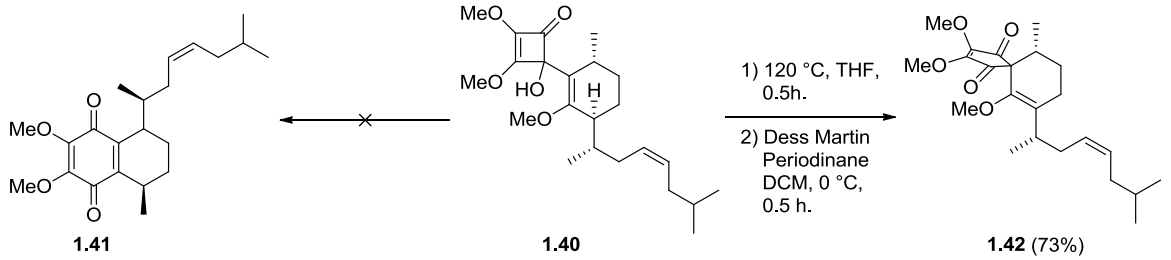
1. Rearrangement of vinylcyclobutenones

For vinyl systems, electrocyclic ring opening can result in either (*Z*)- or (*E*)-vinylketene formation. When the (*Z*)-ketene (**1.32**) is formed, a 6π-electrocyclisation follows leading to hydroquinone **1.34**, which can then be oxidised to quinone **1.35** (Scheme **1.4**). In contrast, when the (*E*)-ketene (**1.36**) is formed, nucleophilic trapping of the reactive ketene by the hydroxyl functionality occurs usually giving rise to a 2(5*H*)-furanone such as **1.39**. In the example shown in Scheme **1.4**, both hydroxyfuran **1.37** and furanone **1.38** are isolated.



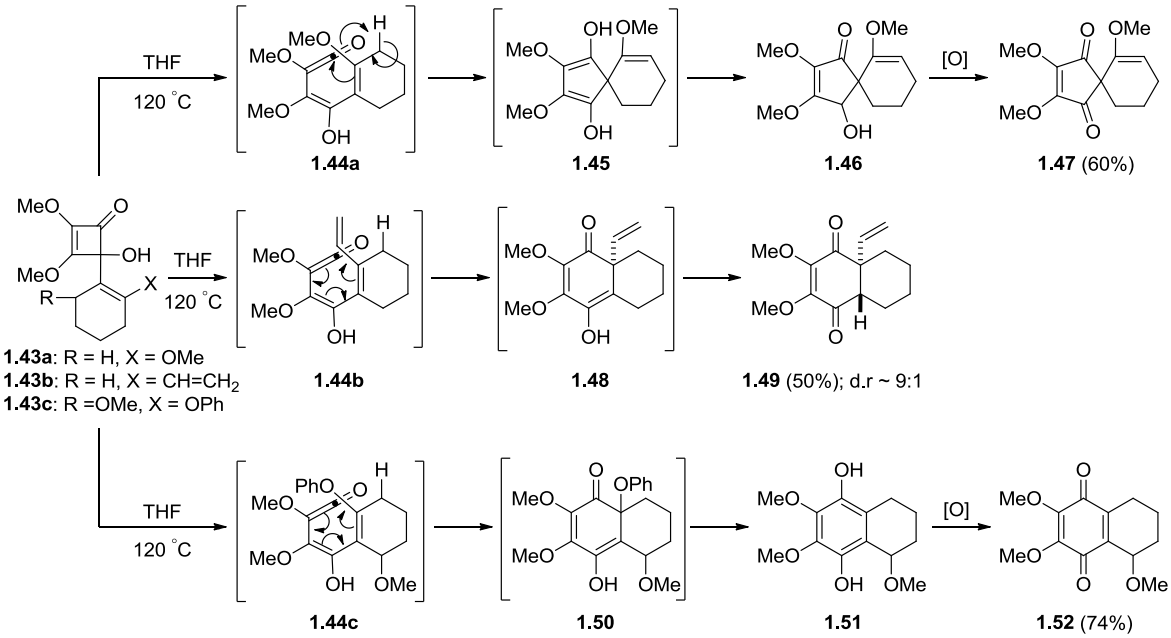
Scheme 1.4: The Moore rearrangement of vinylcyclobutenones.[4c, 9a]

The mechanism outlined in scheme **1.4** generally applies to most vinyl systems.[1, 4c, 9a] However, there are a few exceptions leading to spirocycles such as **1.6** in Figure **1.1**. Discovered during an attempted synthesis of colombiasin A by Harrowven *et al.*,[4k] vinylcyclobutenone **1.40** was found to give spirocycle **1.42** after oxidation rather than the expected quinone **1.41** (Scheme **1.5**).



Scheme 1.5: Unexpected spirocycle **1.42** from vinylcyclobutenone **1.40**.[4k]

In order to understand this unexpected result, thermolyses of substrates with a range of vinyl appendages were examined. It was found that electronic rather than steric factors governed the course of the reaction (Scheme **1.6**). More specifically, the nature of the substituent present on the distal carbon of the vinyl unit dictated whether vinylketene **1.44** gave a carbonyl-ene reaction in preference to the usual 6π-electrocyclisation. Thus, when the substituent was a powerful electron donor (*e.g.* OMe) a carbonyl-ene cyclisation occurred. In contrast, when electron density in the vinyl unit is reduced, 6π-electrocyclisation follows, leading to a cyclohexadienone (**1.48** or **1.50**) that can collapse to a cyclohexenedione (**1.49**) or a quinone (**1.52**) depending on the leaving group ability of vinyl appendage, X.

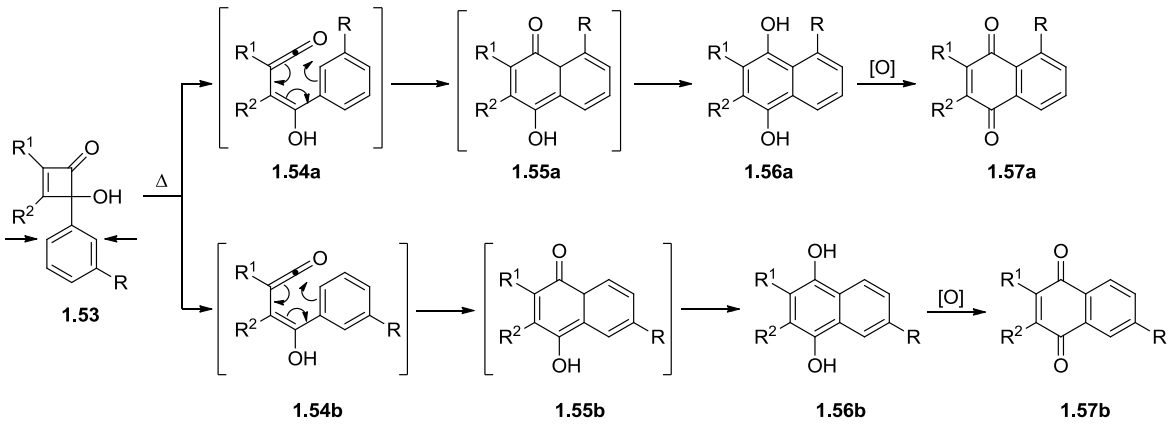


Scheme 1.6: Thermolysis of vinylcyclobutenone with distal substituent X.[4k]

2. Rearrangement of Aryl and heteroarylcyclobutenones

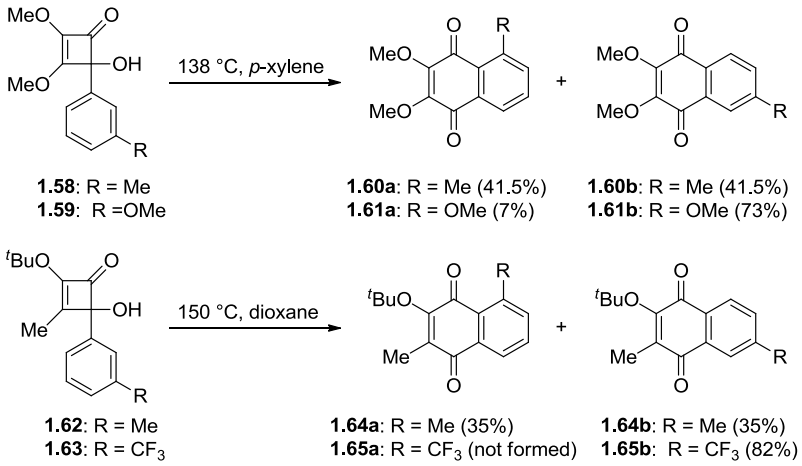
The ring expansion of aryl- and heteroarylcyclobutenones proceeds in much the same way as with vinylcyclobutenones (Scheme **1.4**), with either quinone or furanone products isolated in yields ranging from 73-99%.[1, 3-4, 4f, 4h, 4l, 9c, 10] It is worth mentioning that a regioselectivity issue arises for *meta*-substituted aryl systems. This stems from the fact that once the (*Z*)-ketene is formed, cyclisation can occur to two possible sites, potentially leading to two possible quinone products (**1.57a** and **1.57b**, Scheme **1.7**)

Initial results by Moore *et al*.[4a] found that both products were indeed formed, in a ratio that depended on the nature of substituent groups (Scheme **1.8**). When the *meta*-substituent was a methyl group, a 1 : 1 mixture of quinone products was observed. In contrast, when the *meta*-substituent was a methoxy group, a 10 : 1 ratio in favour of quinone **1.61b** was obtained. Similar product distributions were observed by Harrowven *et al*.[6a] (Scheme **1.8**).



Scheme 1.7: Moore rearrangement with *meta*-substituted aromatic rings.[4a]

Since these results displayed similar selectivities to those given in electrophilic aromatic substitution reactions (S_EAr), Moore suggested that the rearrangement could be viewed as an electrophilic attack of the ketene on the aromatic ring. The effect of electron donating groups on the electrocyclisation would then be expected to be greater, which would account for the difference in regioselectivity when switching from a methyl to a methoxy substituent.



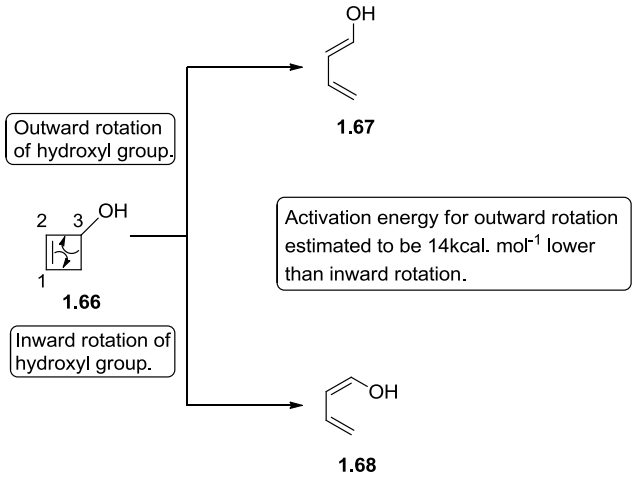
Scheme 1.8: Experimental results for the ring expansion of *meta*-substituted arylcyclobutenones.[4a, 6a]

A more complete picture of the reaction was determined by Harrowven *et al*.[3] They observed a correlation between the rate of the reaction and the inductive effect (σ_I) of a substituent, which included sizeable steric contribution. Their study indicated that 6π-electrocycisation was the rate determining step, which was confirmed by computational modelling of the reaction. Modelling revealed that in the transition state the angle between the aromatic ring and the forming sigma bond is 40° and reduces to 22.1° over the course of the electrocyclisation step. From this it was suggested that the rate of reaction is determined by the ease with which the developing sigma bond is formed rather than the initial interaction of the two ends of the extended π system.

Their model neatly explains why the reaction is under inductive control and has a significant steric component. Thus, for *meta*-substituted arenes (Scheme **1.7**), rotamer **1.54a** has its substituent closer to the newly formed bond than rotamer **1.54b**. Consequently, the rate of cyclisation via rotamer **1.54a** will be influenced to a greater extent than for rotamer **1.54b**. As a result, inductive electron withdrawing *meta*-substituents will cyclise preferentially via rotamer **1.54b** whereas inductive electron donating *meta*-substituents will favour reaction via rotamer **1.54a** (Scheme **1.7**). This was confirmed experimentally by the thermolysis of substrates such as **1.63**, which gave naphthoquinone **1.65b** as the sole product of the reaction as its inductively withdrawing substituent effectively biased cyclisation through rotamer **1.54b** (Scheme **1.7**).[6a]

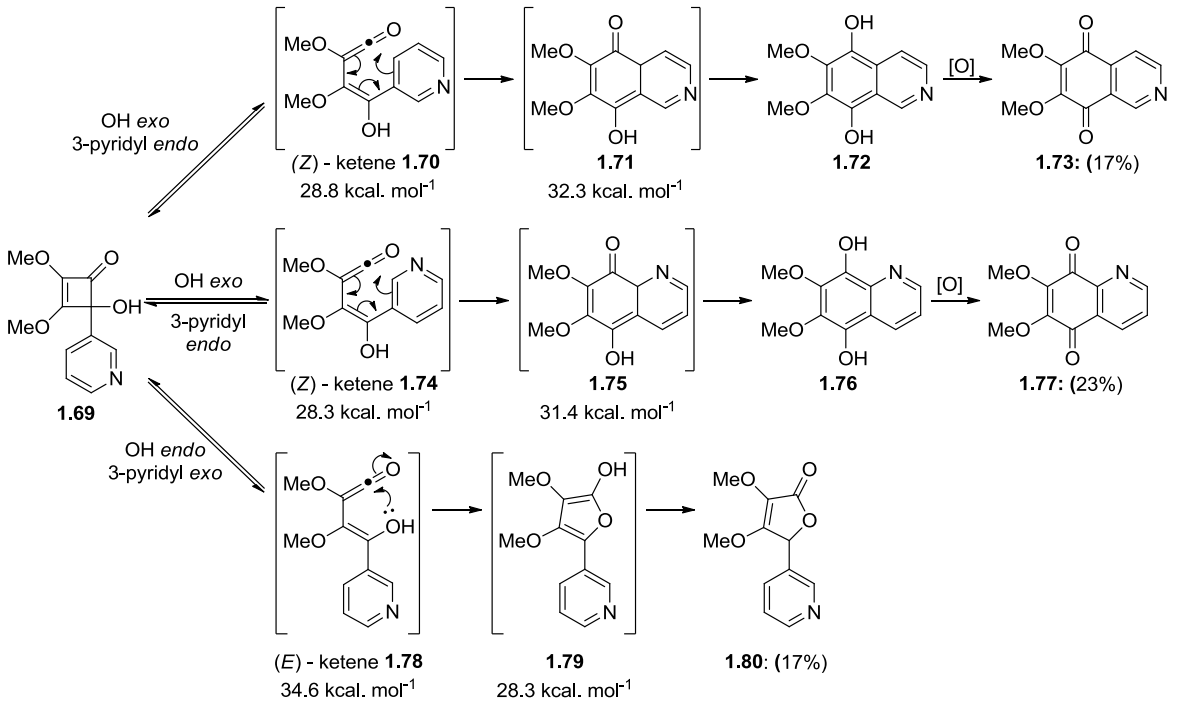
In a related study, Harrowven *et al.*[4l] also addressed the issue of torquoselectivity in the Moore rearrangement. Until their study, it had been assumed from experimental data that the electrocyclic ring-opening reaction was torquoselective, with the (*Z*)-ketene formed during thermolysis and the (*E*)-ketene formed on photolysis. Indeed, this had been rationalised based on the thermal expansion of cyclobutene ring systems (**1.66**), where Houk *et al*.[11] had shown these to favour outward rotation of a hydroxyl group by 14 kcal.

mol^{-1} over inward rotation (Scheme **1.9**). The reason for this stems from the fact that both HOMO and LUMO interactions with oxygen are favoured when the hydroxyl group undergoes an outward rather than inward rotation. Thus it came to be perceived that thermolytic ring expansions only yielded the (*Z*)-ketene, with the converse true for photolytic ring expansions.



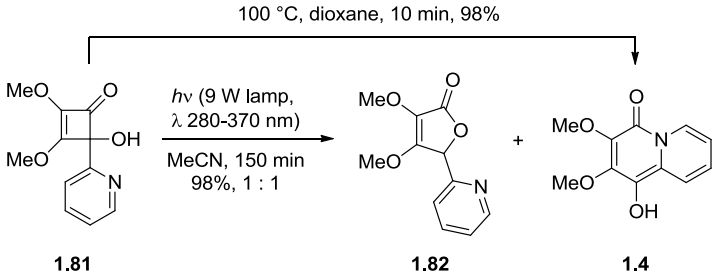
Scheme 1.9: Electrocyclic ring opening of cyclobutene systems.[11]

However, rearrangement of pyridylcyclobutenones **1.69** and **1.81** returned unexpected results (Schemes **1.10** and **1.11**).[4l] For 3-pyridylcyclobutenone **1.69**, thermolysis led to a mixture of furanone **1.80** and quinones **1.73** and **1.77** which was inconsistent with the reaction proceeding via a torquoselective ring opening (Scheme **1.10**). Interestingly, computational modelling of the reaction showed that electrocyclic ring opening was selective, with the formation of (*Z*)-ketenes **1.70** and **1.74** being favoured over (*E*)-ketene **1.78** by 6 kcal. mol^{-1}. However, importantly, for pathways leading to quinoline **1.77** and isoquinoline **1.73** the rate determining step of the reaction is the 6π-electrocyclisation step with an activation energy greater than 31 kcal. mol^{-1} (Scheme **1.10**). This is comparable to the electrocyclic ring opening of **1.69** to (*E*)-ketene **1.78**, explaining why a complex product mixture containing furanone **1.80** and quinones **1.73** and **1.77** is obtained in this reaction.

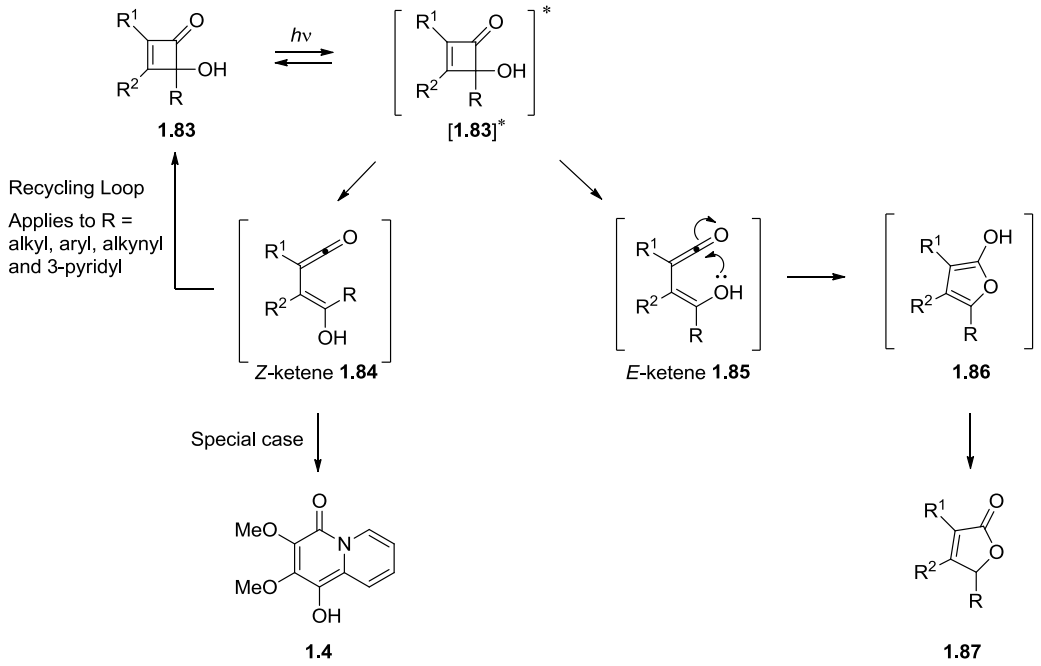


Scheme 1.10: Thermolysis of 3-pyridylcyclobutenone **1.69**.[4l]

As for 2-pyridylcyclobutenone **1.81**, unlike other cyclobutenones studied, photolysis generated both quinolizinone **1.4** and furanone **1.82** in similar yield (Scheme **1.11**).[4l] Again, this result casts doubt on the notion of torquoselective ring opening of the cyclobutenone in the Moore rearrangement. Once again, computational modelling of the reaction determined that thermal electrocyclic ring opening was torquoselective.[4l]



Scheme 1.11: Photolysis and thermolysis of 2-pyridylcyclobutenone **1.81**.[4l]

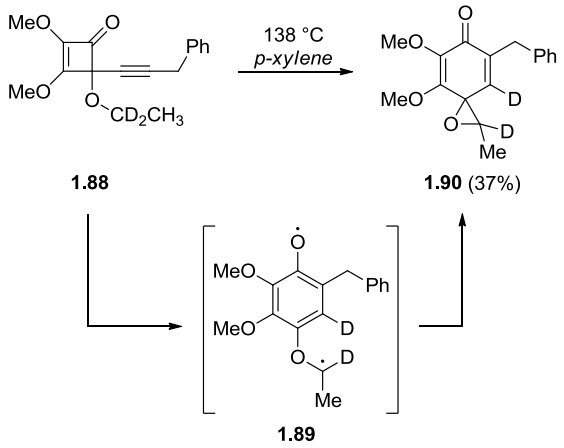


Scheme 1.12: Schematic representation of the selectivity of the Moore rearrangement.[4l]

Under photochemical conditions cyclobutenone **1.83** is promoted to an excited state [**1.83**]* with sufficient energy to collapse to either of the ketenes **1.84** or **1.85** (Scheme **1.12**). In most cases, the energy barrier for 6π-cyclisation is higher than 4π-electrocyclisation back to the starting material so the (*Z*)-ketene is recycled. By way of contrast, for (*E*)-ketene **1.85** ring closure to furan **1.86** is lower in energy than 4π-electrocyclisation so it proceeds to give the corresponding 2(5*H*)-furanone **1.87**. 2-pyridylcyclobutenone **1.81** represents a special case as the pyridine nitrogen can trap the (*Z*)-ketene in a low energy pathway (3.5 kcal. mol^{-1}) so quinolizinone **1.4** and 2(5*H*)-furanone **1.82** are formed in equal measure (Scheme **1.12**).

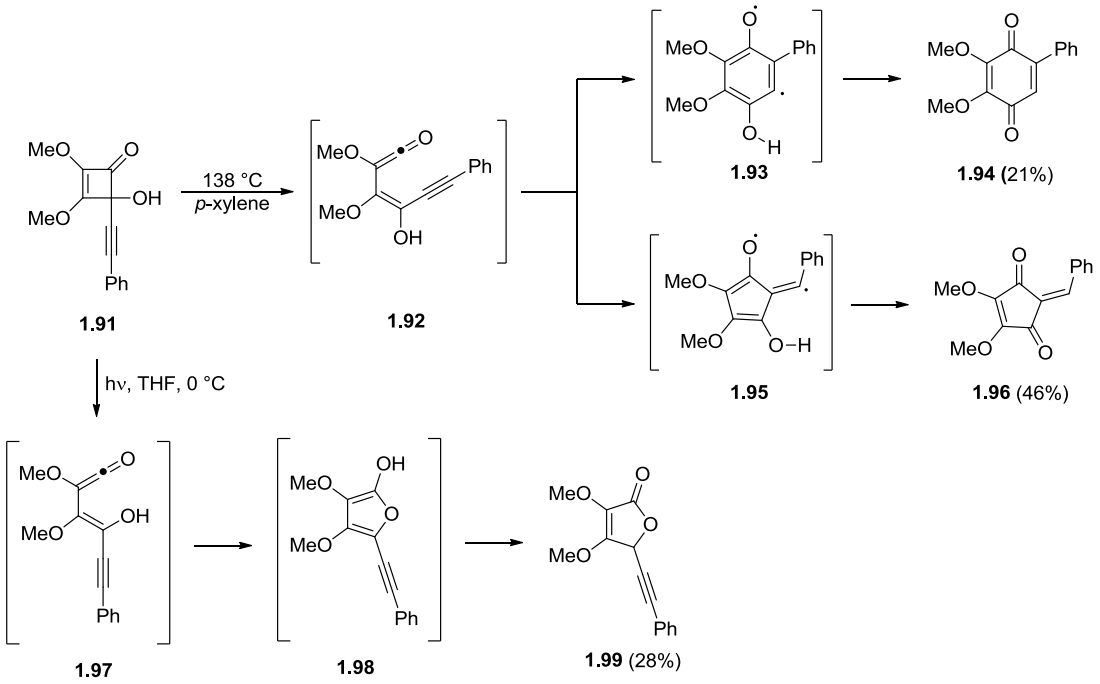
3. Moore rearrangement of alkynylcyclobutenones

The alkynyl variant of the Moore rearrangement is slightly different in that it proceeds via a diradical mechanism leading to quinones and cyclopentenediones directly. The reaction was pioneered by Moore *et al.* who proposed a mechanistic course based on computational studies and experimental results, including those attained with cyclobutenone **1.88**, where thermolysis led to spirocycle **1.90** exclusively (Scheme **1.13**).[4b, 4m, 11-12] From these studies he deduced that diradical species were intermediates in the reaction.

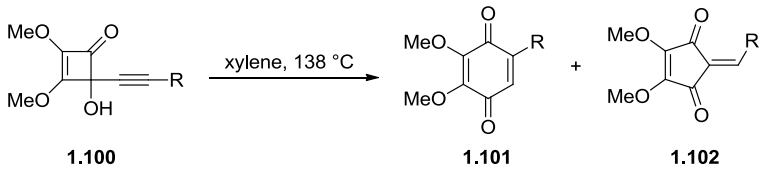


Scheme 1.13: Thermolysis of cyclobutenone **1.88** suggesting expansion occurs via radical mechanism.[4b]

The first step of the rearrangement operates in much the same way as the others, with electrocyclic ring opening of the cyclobutenone leading to either the (*Z*) or (*E*)-ketene **1.92** or **1.97** (Scheme **1.14**). With respect to the (*E*)-ketene **1.97**, the outcome of the reaction is the same as that described previously, with furanone **1.98** being obtained as final product. By contrast, reactions proceeding via the (*Z*)-ketene **1.92**, can form either or both of the biradical species **1.93** or **1.95**. From experimental findings, Moore concluded that the product distribution was determined by the ability of the alkyne residue, R, to stabilise the carbon-centred radical formed, as radical stabilising groups showed a preference for cyclopentenedione formation while alkyl residues favoured quinone production (Schemes **1.14** and **1.15**).[4b, 12]



Scheme 1.14: Moore rearrangement of 2,3-dimethoxy-4-hydroxy-4-phenylcyclobut-2-enone **1.91**.[4b]

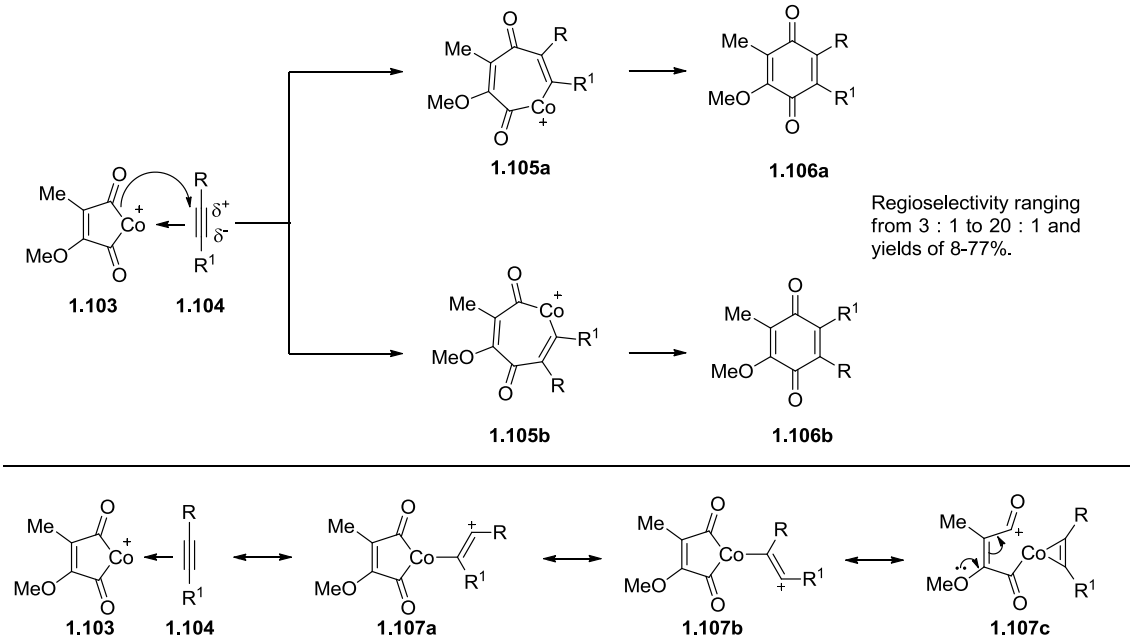


Entry	*R*	***1.101*** / *yield (%)*	***1.102*** / *yield (%)*
1	*$^{n}C_4H_9$*	78	-
2	*CH_2Ph*	71	-
3	*OEt*	25	50
4	*CO_2Et*	-	66
5	*$CH{=}CHOCH_3$*	-	49

Scheme 1.15: Product distribution for Moore rearrangement for selected alkynylcyclobutenediones.[4b]

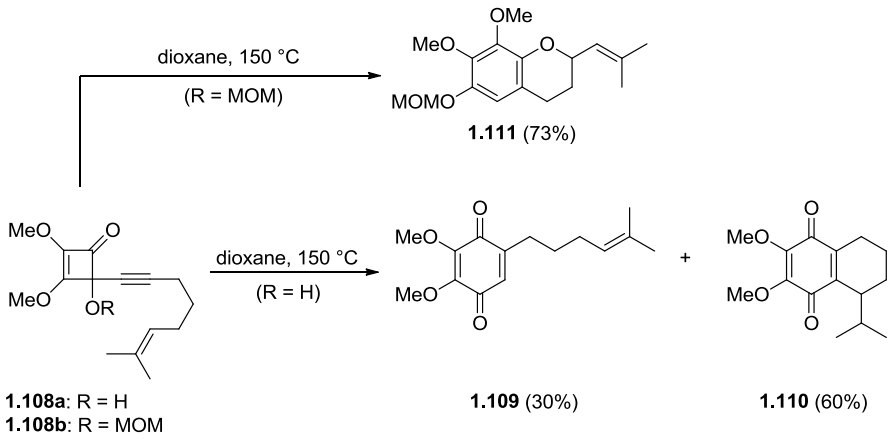
It is noteworthy that Liebeskind *et al.* developed an alternative strategy for the synthesis of quinones using cyclobutenediones and alkynes.[13] In this methodology, cobalt complexes of cyclobutenediones are reacted with alkynes to access the desired quinone as shown in scheme **1.16**. One drawback of this method is that mixtures of regioisomers are commonly obtained when unsymmetrical cyclobutenediones are employed. Modest selectivities were observed with terminal alkynes and those bearing electron withdrawing groups, while substrates with electron donating alkyne residues gave regioselectivites of

up to 20 : 1. Liebebskind rationalised the outcome in terms of an initial insertion of alkyne **1.104** into the cobalt complex followed by reductive elimination (Scheme **1.16**).



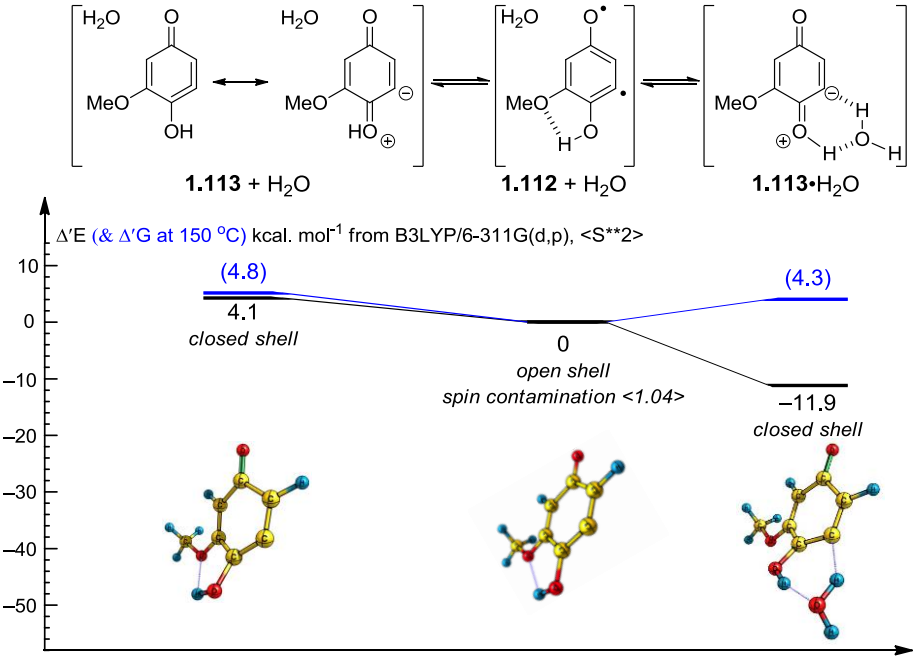
Scheme 1.16: Proposed mechanism for alkyne insertion into maleoylcobalt complexes of cyclobutenediones.[13]

Finally, Harrowven *et al.*[4m] discovered another interesting facet of alkynylcyclobutenone rearrangement during studies directed towards the synthesis of quinone **1.110** from cyclobutenone **1.108a**. They found that much of the mass balance of the reaction was lost to the formation of side product **1.109**, which stems from hydrogen atom abstraction rather than the desired 6-*exo*-trig cyclisation. In an attempt to improve the yield of **1.110**, they spiked the solvent with D_2O in the hope that this would slow the rate of hydrogen atom abstraction down without affecting the rate of cyclisation.



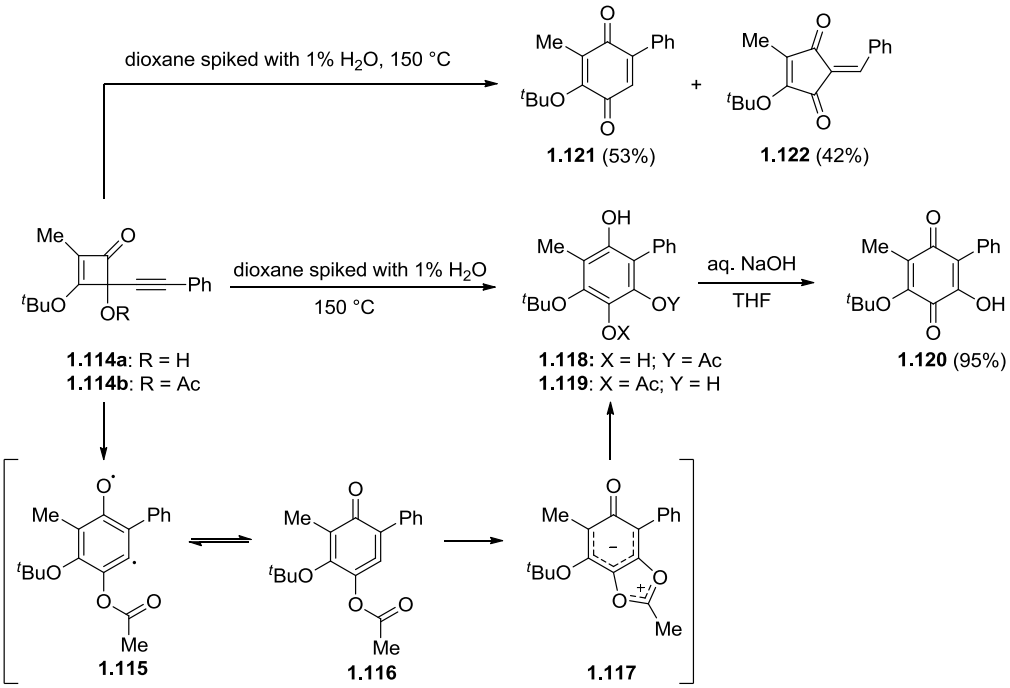
Scheme 1.17: Effect of protecting hydroxyl functionality on reaction outcome.[4m]

Instead, the presence of deuterium oxide had quite the opposite effect, with deuterated **1.109** being isolated in 95% yield! To make sense of this result, they proposed the intermediacy of a zwitterionic orbital isomer **1.113** as it could participate in a proton transfer reaction catalysed by D_2O (Scheme **1.18**). Modelling of the reaction pathway using computational methods revealed that the presence of water made the water-zwitterion complex **1.113**•H_2O more stable than biradical species **1.112** by 11.9 kcal. mol^{-1} (Scheme **1.18**). To test the importance of the hydroxyl functionality further, they went on to protect the hydroxyl functionality in **1.108a** and showed that this shut down the route to quinone **1.109**, giving benzopyran **1.111** in 73% yield via diradical intermediates (Scheme **1.17**).



Scheme 1.18: Computational modelling results for the orbital isomers involved in alkynylcyclobutenone rearrangements.[4m]

Furthermore, modelling revealed that the 6 membered ring adopted a puckered geometry upon complexation with water leading them to suggest the water complex had considerable cyclohexatriene character (**1.113**, Scheme **1.18**), which could also lead to novel reactivity. This was demonstrated by protecting the hydroxyl group as its acyl ester. Thermolysis now led to the formation of acetates **1.118** and **1.119**, via intermediate **1.117**, in 95% yield rather than the usual mixture of quinone **1.121** and cyclopentadione **1.122** (Scheme **1.19**).



Scheme 1.19: Acyl protection of alkycyclobutenedione.[4m]

Chapter 2: Cribrostatin 6

Introduction

In 2003, Pettit and co-workers reported the isolation of a natural product that was uniquely based on the imidazo[5,1-a]pyridine subunit.[2] It was isolated from the marine sponge *chribrochalina* Sp. together with a small family of related compounds (Figure **2.1**), and named cribrostatin 6 (**2.6**).[2, 14]

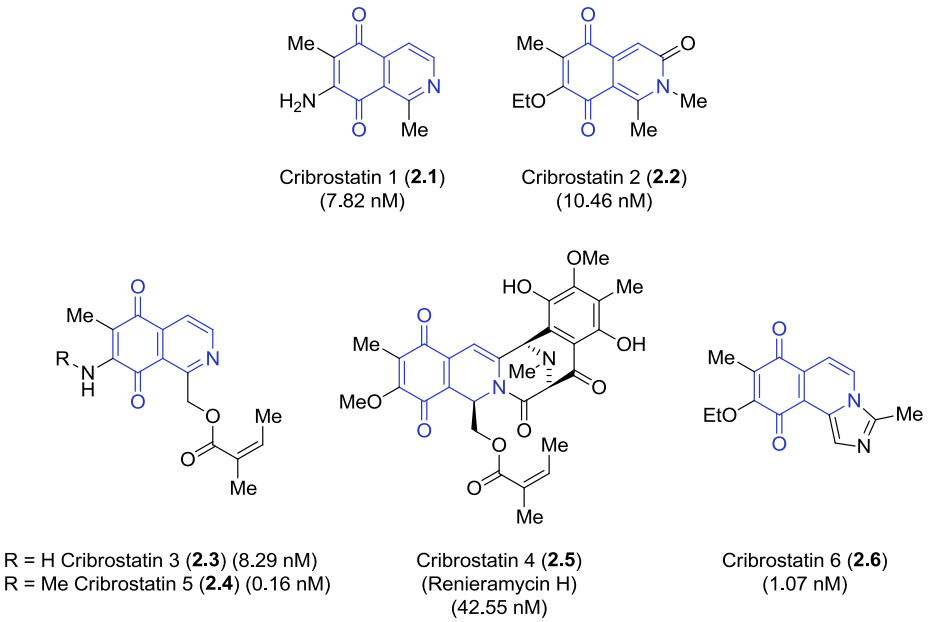


Figure 2.1: Cribrostatin family including GI_{50} values measured for P388 leukemia cell line.[2, 14]

Cribrostatin 6 has been screened against 111 strains of bacteria and seven cancer cell lines. With respect to bactericidal activity, cribrostatin 6 inhibited growth for all the strains tested, with a minimum inhibitory concentration ranging from 0.46 nM to 120 nM (Table **2.1**).[15] In terms of antineoplastic activity, cribrostatin 6 displayed GI_{50} values in the nanomolar region, with the natural product showing the highest affinity for the breast cancer cell line MCF-7 (Entry 2, Table **2.2**).

Entry	*Bacteria*	*MIC / nM*	*MBC / nM*
1	*Enterococcus* ssp. (vancomycin resistant)	120	> 240
2	*Staphylococcus aureus* (Methicillin resistant)*	7.40 - 59.23	> 240
3	*S. Pneumoniae* ATTCC 700673 (multidrug resistant)	0.46	0.92
4	*S. Pneumoniae* (Penicillin resistant)*	0.92 - 14.81	7.40 - 59.23

Table 2.1: Reported minimum bactericidal concentration (MBC) and minimum inhibitory concentration (MIC) for cribrostatin 6 against a selected range of bacteria.[15]

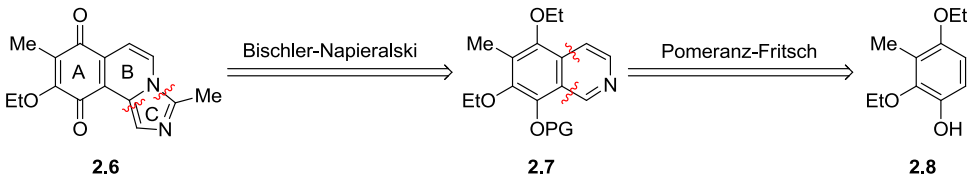
* Several strains of bacteria were tested.

Entry	*Cell Line*	*GI_{50} / nM*
1	Pancreas-adenocarcinoma (BXP-3)	> 3.70
2	Breast-adenocarcinoma (MCF-7)	0.78
3	CNS glioblastoma (SF-268)	0.89
4	Colon-adenocarcinoma (KM20L2)	> 3.70
5	Prostate (DU-145)	1.41

Table 2.2: Antineoplastic activity for cribrostatin 6 against selected cancer cell lines.[2]

Synthetic routes to cribrostatin 6

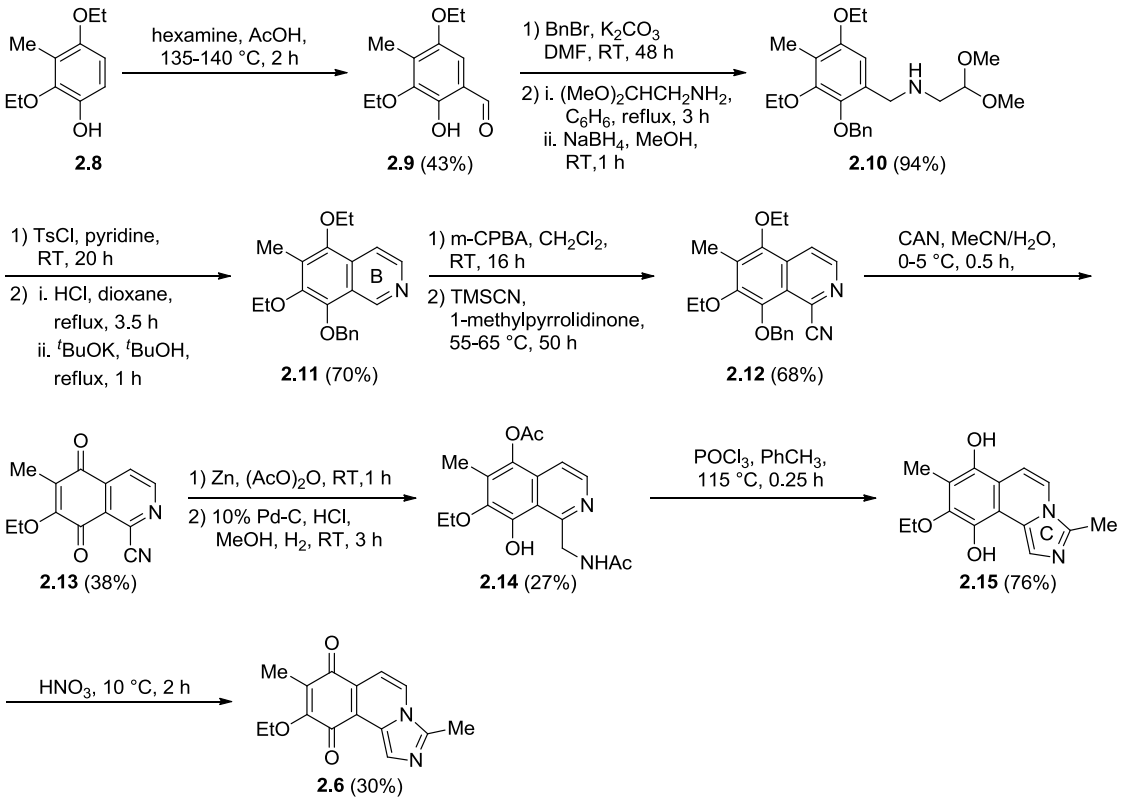
Nakahara *et al*.[16] were the first to develop a route to cribrostatin 6, having previously synthesised both cribrostatins 1 and 2.[17] Their strategy involved the successive instalment of the B and C ring of cribrostatin 6 using Pomeranz-Fritsch and Bischler-Napieralski reactions as key steps (Scheme **2.1**).



Scheme 2.1: Retrosynthetic analysis of cribrostatin 6 by Nakahara *et al*.[16a]

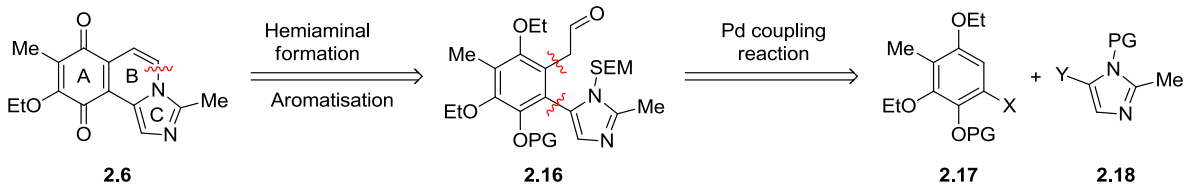
Construction of ring B required benzylamine **2.10**, which was prepared from commercially available 2,4-diethoxy-3-methylphenol **2.8** in 40% yield over 2 steps (Scheme **2.2**). A modified Pomeranz-Fritsch cyclisation of **2.10** then afforded isoquinoline **2.11**, the core of

cribrostatin 6, in 70% yield. It was next transformed into amide **2.14** over 5 steps, setting up a Bischler-Napieralski cyclisation to **2.15**, constituting the ABC framework of cribrostatin 6 in 76% yield. Finally, treatment with nitric acid furnished cribrostatin 6 (**2.6**) in 0.5% overall yield for a 12 step synthesis (Scheme **2.2**).



Scheme 2.2: Synthetic route developed by Nakahara *et al.*[16]

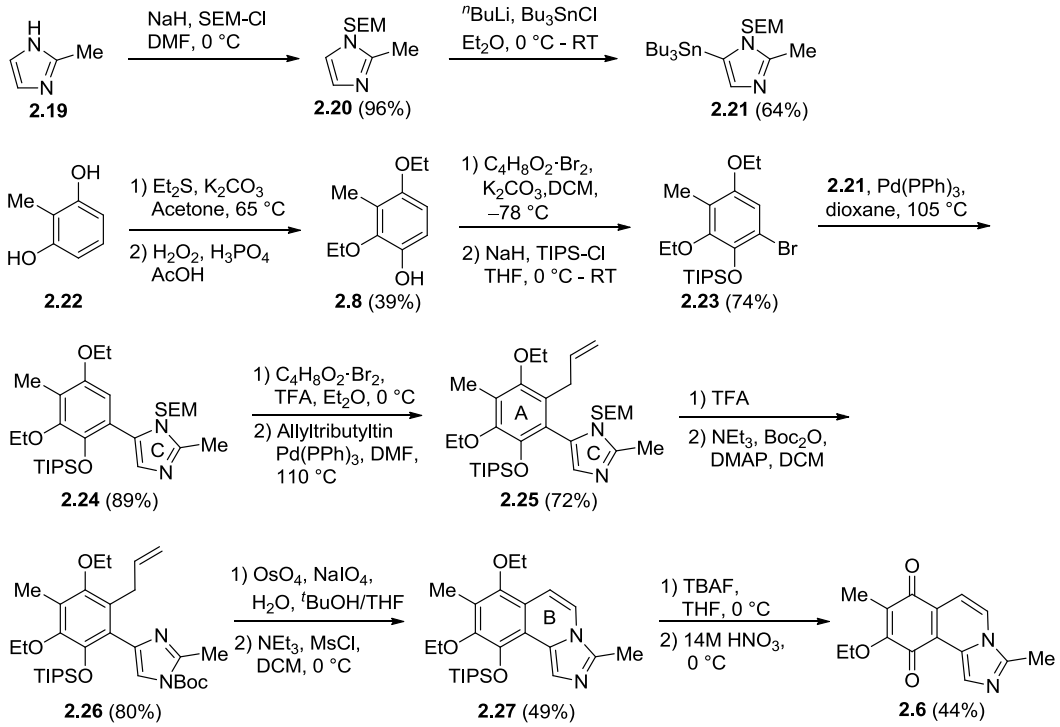
Shortly thereafter, Kelly *et al.* published an alternative synthesis of the natural product.[18] They proposed forming the AC ring system first by means of a palladium catalysed cross-coupling between simple precursors **2.17** and **2.18** (Scheme **2.3**). Ring B would then be constructed via an intramolecular cyclisation after installation of a pendant aldehyde functional group to the AC ring system (Scheme **2.3**).



Scheme 2.3: Retrosynthetic analysis of cribrostatin 6 by Kelly *et al.*[18]

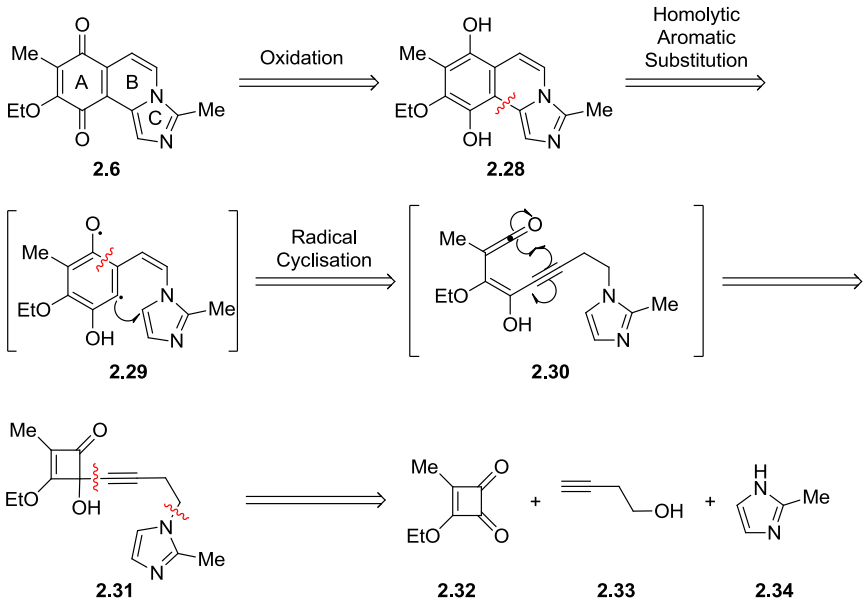
Desired biaryl **2.24** was formed in 89% yield through a Stille coupling of methylimidazole **2.21** with protected phenol **2.23** (Scheme **2.4**). A second Stille coupling followed, to install an allyl group on the A ring of biaryl **2.24**. Dihydroxylation of allyl **2.26**, oxidation and

concomitant cyclisation furnished the fused tricyclic system of cribrostatin 6 (**2.27**) in 49% after rearomatisation (Scheme **2.4**). Lastly, deprotection and oxidation gave cribrostatin 6 in 2% overall yield for the 13 steps from resorcinol **2.22** (Scheme **2.4**).



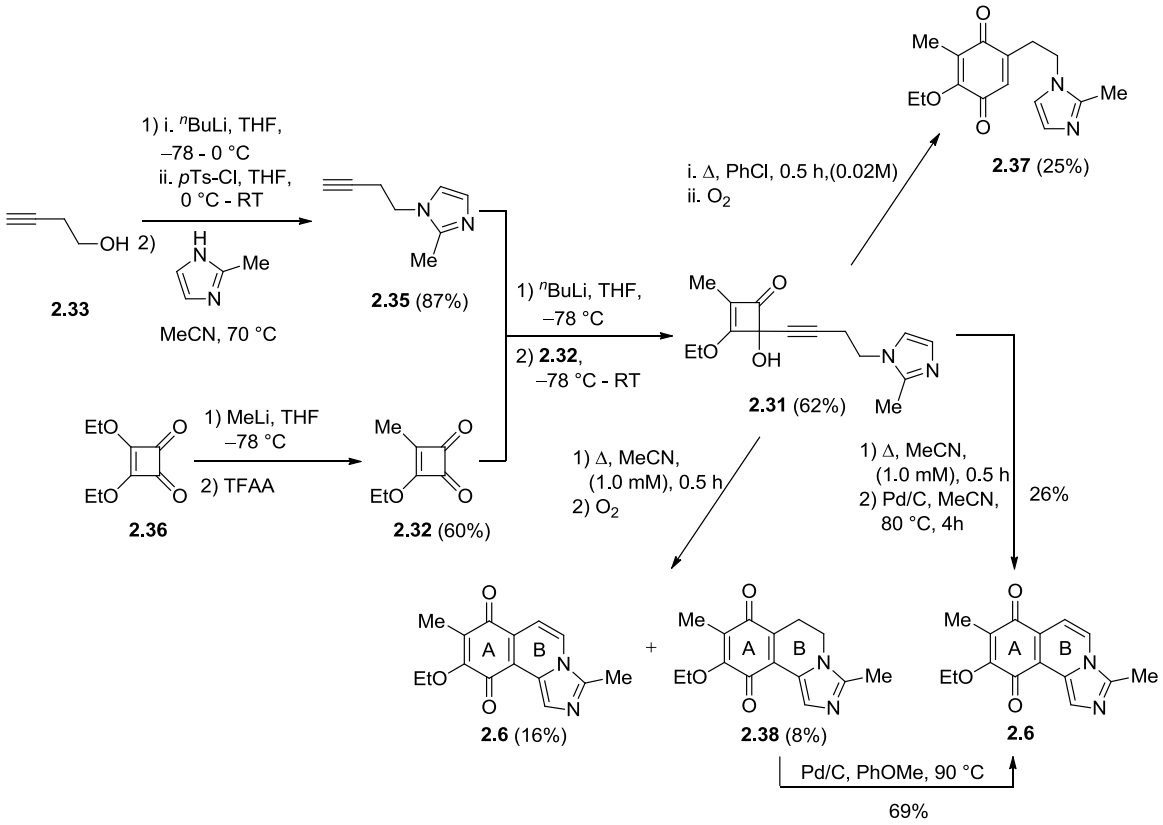
Scheme 2.4: Synthesis of cribrostatin 6 by Kelly *et al*.[18]

In contrast, Martin *et al.* envisioned forming the ABC ring system via a homolytic aromatic substitution reaction with an *in situ* formed biradical **2.29** (Scheme **2.5**).[19] This, they suggested, could be achieved in one step from cyclobutenone **2.31** using the Moore rearrangement (Scheme **2.5**), with **2.31** derived from simple precursors **2.32**, **2.33** and **2.34**.



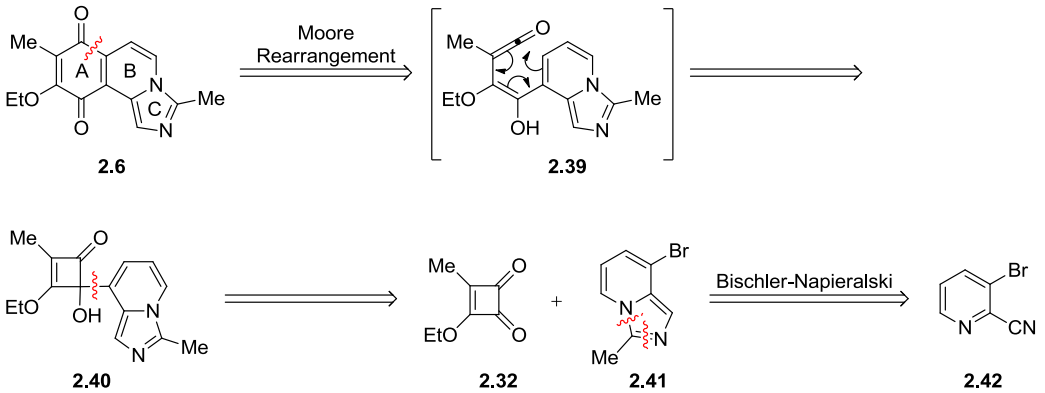
Scheme 2.5: Retrosynthetic analysis by Martin *et al*.[19]

Their synthesis of key cyclobutenone **2.31** commenced with an S_N2 reaction between tosyl protected 1-butynol (**2.33**) and 2-methyl-1*H*-imidazole (**2.34**), forming imidazoalkyne **2.35** in 87% yield (Scheme **2.6**). Its lithiation and addition to cyclobutenedione **2.32** next led to intermediate **2.31** (62%), which gave on thermolysis a 2 : 1 mixture of quinone **2.38** and cribrostatin 6 in 8% and 16% yield respectively. That the former could be converted to the latter by dehydrogenation using Pd/C (69%) was the only notable success in their attempts to improve the overall yield of the synthesis. Indeed, optimisation work appeared to show that hydrogen atom abstraction was favoured at higher concentrations, leading to benzoquinone **2.37** (Scheme **2.6**).[4m]

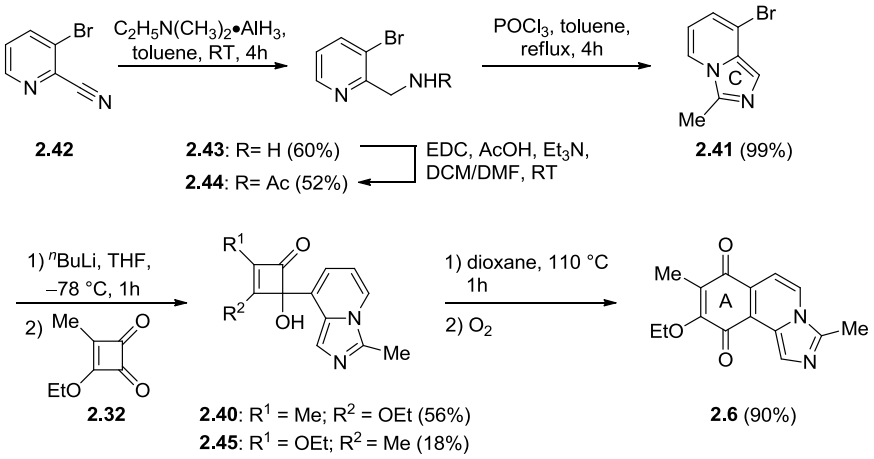


Scheme 2.6: Synthetic route developed by Martin *et al*.[19]

Finally, an analogous route was developed by Harrowven *et al.* Their strategy utilised the alkenyl variant of the Moore rearrangement to generate the A ring of cribrostatin 6 from cyclobutenone **2.40**, the BC ring having been synthesised first from commercially available 3-bromo-2-cyanopyridine (Scheme **2.7**).[3] Much like Nakahara *et al*.[16a], the BC ring system of cribrostatin 6 was formed through a Bischler-Napieralski cyclisation of amide **2.44** in 99% yield (Scheme **2.8**). Halogen-lithium exchange followed by addition to cyclobutenedione **2.32** then gave a 3 : 1 mixture of the regioisomeric adducts **2.40** and **2.45**. Finally, thermolysis of **2.40** afforded the natural product in 90% yield (Scheme **2.8**).



Scheme 2.7: Retrosynthetic analysis by Harrowven *et al.*[3]



Scheme 2.8: Synthetic route developed by Harrowven *et al.*[3]

Biological activity of cribrostatin 6 and its analogues.

Hergenrother *et al*.[20] conducted an in-depth analysis of cribrostatin 6's antineoplastic activity. From their study, they observed that this natural product was cytotoxic towards both quiescent and rapidly dividing cells, a valuable observation as most common antineoplastic agents are inactive towards quiescent cells.[20] It was also noted that cribrostatin 6 kills through apoptosis, with only 8% of cells being viable after 16 h of incubation.

The effects on DNA, topoisomerase I and II and cell cycle arrest were also assessed as part of the study. It was discovered that cribrostatin 6 had minimal interaction with DNA, unlike doxorubicin (a quinone antineoplastic agent); it does not inhibit topoisomerase I and II strongly *in vitro*, as do many approved cancer drugs, and does not induce cell cycle arrest prior to apoptosis.

Finally, a profiling experiment highlighted the activation of several oxidant transcripts, most notably HMOX1, which is controlled by a transcript factor that is activated when under oxidative stress.[20] This reinforced the aforementioned results, but also allowed them to compare cribrostatin 6 against known bioactive molecules. Interestingly, they found no profile correlation between cribrostatin 6 and a known library of 1300 small molecules.

Included in the aforementioned study (published as part of Hoyt's doctoral research thesis) were eighteen analogues of cribrostatin 6 designed to explore structure activity relationships (SAR) in this natural product.[21] Three lines of enquiry were established (Figure **2.2**).

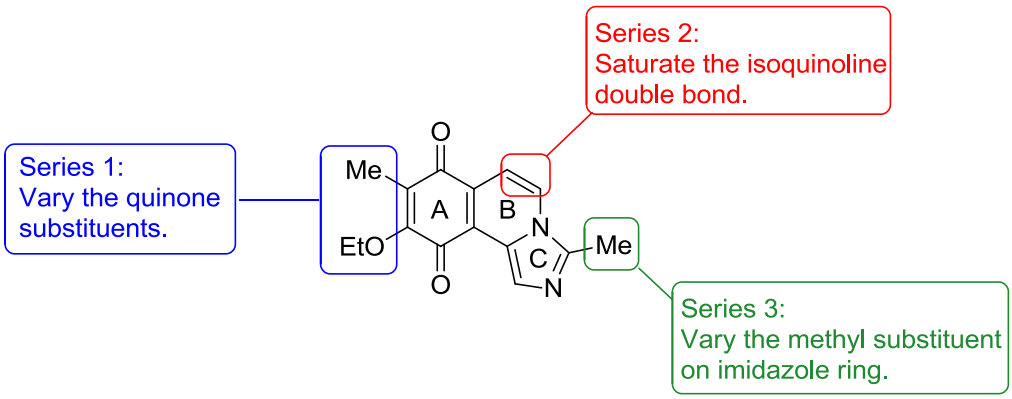
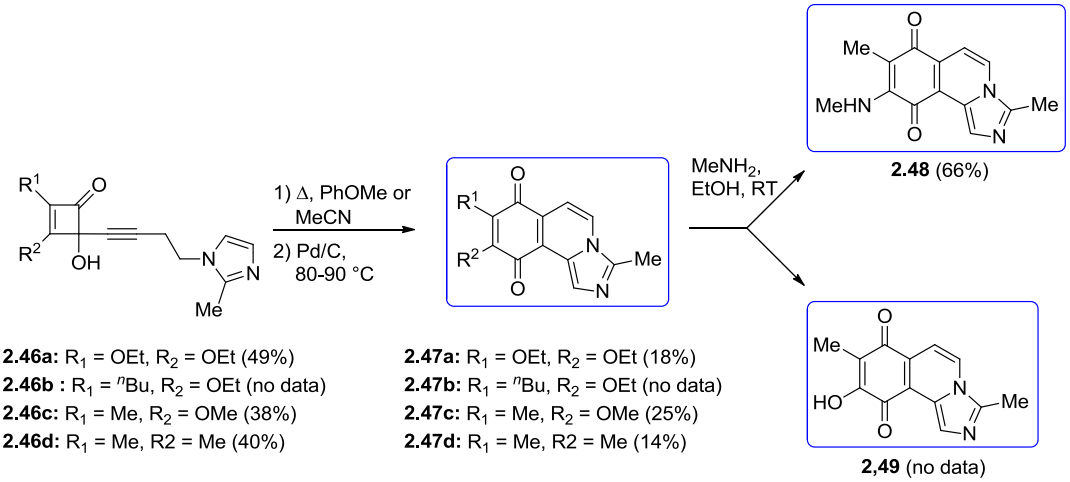


Figure 2.2: Modification envisaged by Hergenrother *et al*.[21]

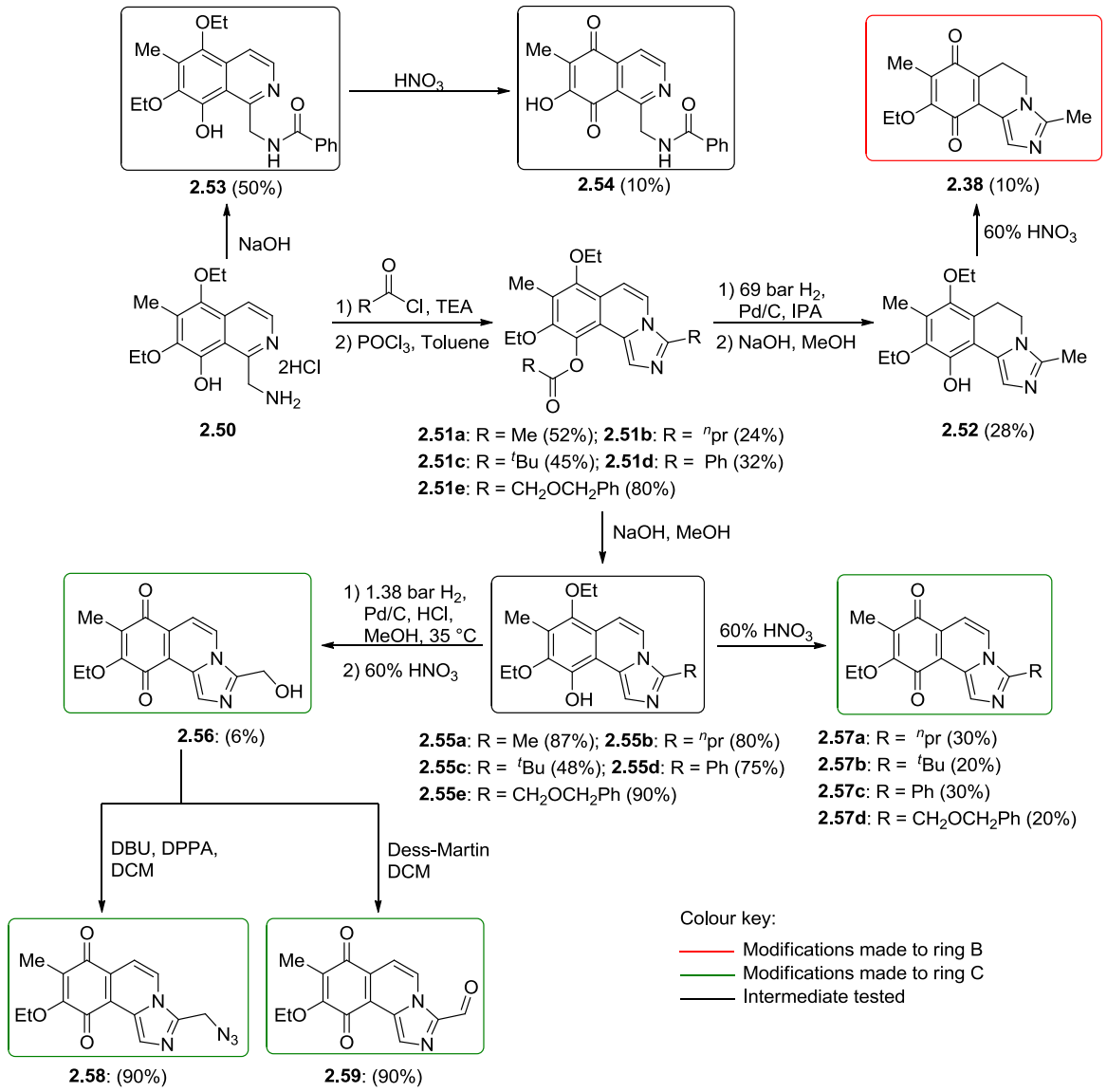
The first series focussed on the quinone unit. Initially, Hoyt tried to form an unsubstituted version of cribrostatin 6 by starting with 2,5-dimethoxybenzaldehyde and following a modified version of Nakahara *et al*.'s synthetic route.[16a] Unfortunately, the desired quinone could not be formed by that route and it was abandoned. The six analogues tested in series 1 were provided by Martin *et al.* (Scheme **2.9**),[22] and prepared following their strategy described earlier (Scheme **2.6**).

Series 2 looked at modifications to ring B of the isoquinoline unit (Figure **2.2**). To that end, the double bond of the isoquinoline ring was reduced to give **2.38** in 1% yield from common intermediate **2.50** (Scheme **2.10**). This compound proved to be unstable both in solid form and in solution, forming cribrostatin 6 over several hours.

Finally, series 3 centred on the imidazole ring (Figure **2.2**). Here, the methyl substituent of ring C was replaced with both polar and non-polar groups. These structural changes were introduced prior to the Bischler-Napieralski cyclisation using four different acyl chlorides (Scheme **2.10**).



Scheme 2.9: Analogues of cribrostatin 6 synthesised by Martin *et al*.[21-22]



Scheme 2.10: Analogues synthesised by Hoyt.[21]

These compounds were screened against U937 and HL-60 cancer cell lines. The results are given in Table **2.3**, with a summary of the general trends observed presented in Figure **2.3**. With respect to series 1, it is apparent that the quinone unit is amenable to change. IC_{50} values range from 1.9 to 6.3 μM in HL-60 cell line, displaying similar activity to the natural product (entries 1 to 7, Table **2.3**). One notable exception was **2.49** with an IC_{50} value greater than 100 μM (entry 7, Table **2.3**). These results were mirrored in the U-937 cell line, suggesting that the presence of a hydroxyl group at position 9 is not tolerated.

Entry	*Compound*	*U-937 (human lymphoma cancer cell line) / μM*	*HL-60 (human leukemia cell line) / μM*
1	Cribrostatin 6	5 ± 1	0.3 ± 0.05
2	**2.47a**	4 ± 1	3.3 ± 0.5
3	**2.47b**	12 ± 5	4.7 ± 0.3
4	**2.47c**	6 ± 2	1.9 ± 0.6
5	**2.47d**	9 ± 1	2.5 ± 0.8
6	**2.48**	12 ± 2	6.3 ± 0.5
7	**2.49**	63 ± 5	>100
8	**2.38**	14 ± 4	0.8 ± 0.2
9	**2.53**	18 ± 1	8 ± 2
10	**2.54**	>100	>100
11	**2.55a**	79 ± 3	91 ± 5
12	**2.56**	17 ± 7	3 ± 0.8
13	**2.57a**	17 ± 6	0.8 ± 0.2
14	**2.57b**	6 ± 3	0.2 ± 0.05
15	**2.57c**	>100	6 ± 1
16	**2.57d**	>100	>100
17	**2.58**	16 ± 3	1.3 ± 03
18	**2.59**	7 ± 2	0.9 ± 0.05

Table 2.3: IC_{50} values for cribrostatin 6 and its analogues, determined by Hoyt.[21]

For series 2, the instability of **2.38** leads to a high level of uncertainty surrounding the reported IC_{50} values (0.8 and 14 μM). Consequently, no conclusion could be drawn from these results. In terms of series 3, most substituent changes are tolerated with IC_{50} values measured within the 0.2 - 17 μM range across both cell lines (entries 12 to 18, Table **2.3**). Notably, analogue **2.57d** (entry 16, Table **2.3**), possessing a benzyloxymethyl group, has an IC_{50} greater than 100 μM in both cell lines tested. This is also true to some extent of analogue **2.57c** bearing a phenyl group. It had an IC_{50} greater than 100 μM in U937 cells compared to 6 μM in HL-60 cells (entry 15, Table **2.3**).

Also tested were intermediates **2.53**, **2.54** and **2.55a** (Scheme **2.10**, Table **2.3**). Both compounds **2.54** and **2.55a** have high IC_{50} values, in the 79 - 100 μM region when taking both cell lines into account (entries 10 and 11, Table **2.3**). According to Hoyt, this shows that the quinone core and the fused tricyclic unit are essential for the natural product's cyctotoxicity. The conclusion seems premature, however, in view of the fact that only three compounds were tested. Furthermore, although isoquinoline quinone **2.54** showed complete loss of activity, it possesses a hydroxyl group at position 7 on the isoquinoline unit, which had previously been shown not to be tolerated at this position (analogue **2.49**, entry 7, Table **2.3**). In addition, compound **2.53**, from which **2.54** is derived, has an IC_{50}

value ranging from 8 - 18 μM. This is comparable with analogues from series 1 and 3 even though it does not possess a quinone core or the fused tricyclic system of cribrostatin 6. These anomalies suggest that more analogues are required to demonstrate the necessity of the quinone core and tricyclic system (Table **2.3**).

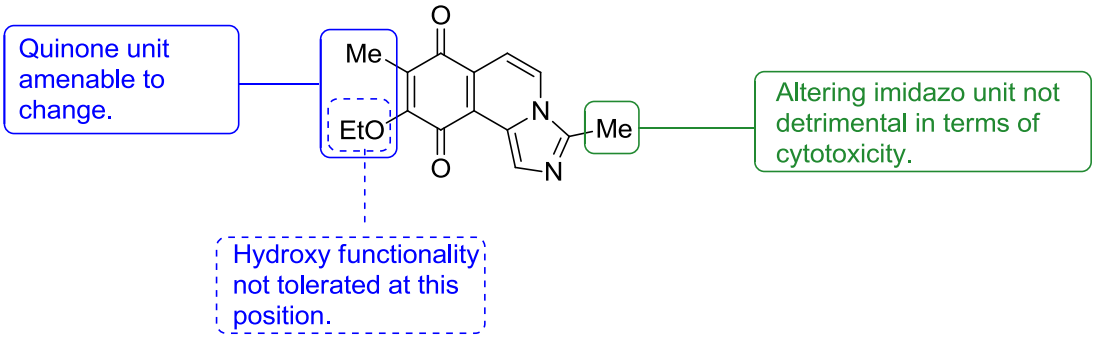
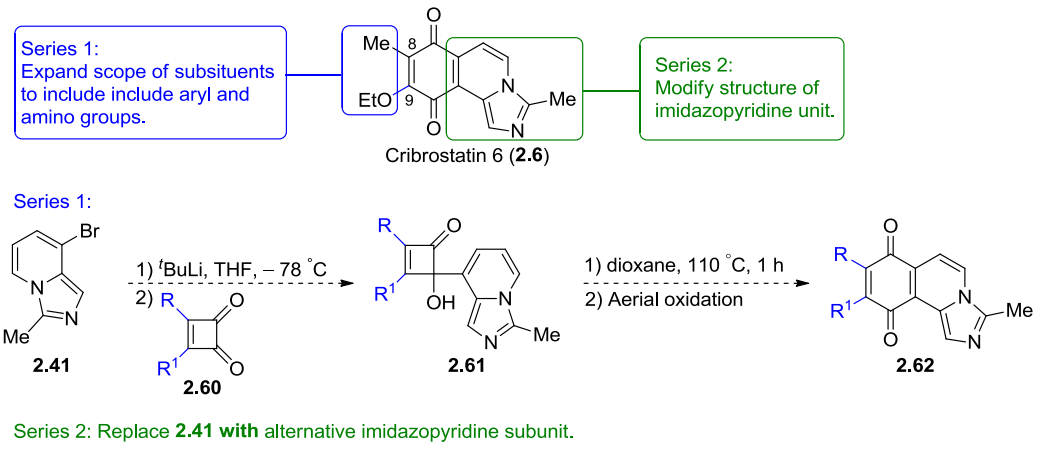


Figure 2.3: Structure activity observations for cribrostatin 6 based on results by Hoyt.

The work carried out by Hergenrother *et al*. highlights the potential of cribrostatin 6 as a promising anti-cancer agent. From the analogues synthesised to date,[21-23] changes to substituents at position 8 and 9 on the cribrostatin 6 skeleton have been limited (Scheme **2.11**). Our aim is to consolidate and expand on the data acquired in this series by preparing a small library of compounds with a view to incorporating both amino and aryl substituents at these positions. Synthesis of these analogues shall be achieved through the use of the Moore rearrangement by exploiting the chemistry of cyclobutenediones **2.60** and cyclobutenones **2.61**, as exemplified in Scheme **2.11**. In addition to this, synthetic work will try to explore the importance of the imidazo[5,1-a]pyridine subunit of cribrostatin 6 by replacing heterocycle **2.41** with an alternative imidazopyridine system. Herein is presented and discussed the synthesis and screening of a number of analogues of cribrostatin 6 as well as attempts to modify the imidazo[5,1-a]pyridine core of **2.6**.

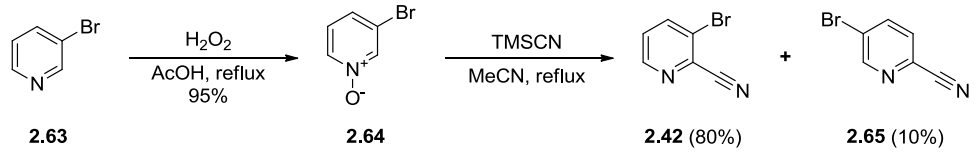


Scheme 2.11: Initial strategy for the synthesis of cribrostatin 6 analogues.

Synthesis of cribrostatin 6

As shown by Hoyt, the synthetic route developed by Nakahara *et al.*[16a] is best suited to the preparation of analogues with an altered imidazopyridine ring. Structural changes to the quinone unit are rather more difficult to achieve using this route as it would require the complete 12 step synthesis to be run through to form one analogue which is a costly and inefficient process. The same can be said for the synthetic route of Kelly *et al.*[18] In contrast, both routes employing the Moore rearrangement are short and convergent, making them attractive for the synthesis of analogues. Of the two routes, Harrowven *et al.*[3] is the more efficient with an overall yield of 16% as compared to 14% for roughly the same number of steps.

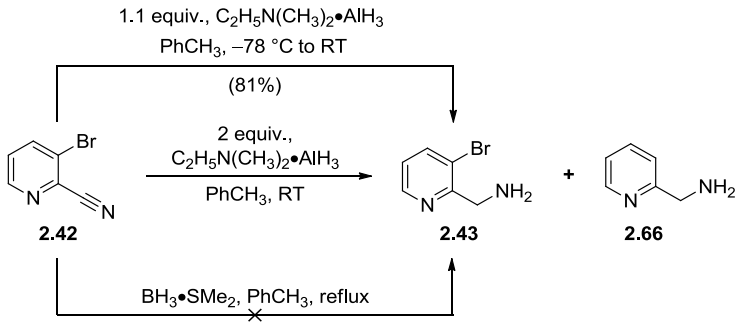
Thus, our synthesis started with the conversion of 3-bromopyridine **2.63** to its *N*-oxide **2.64** (Scheme **2.12**), to activate the pyridine ring towards nucleophilic attack. Treating a solution of **2.63** in acetic acid with hydrogen peroxide (large excess) produced *N*-oxide **2.64** in a 95% yield. This was then converted to the desired nitrile **2.42** using trimethylsilyl cyanide (TMSCN). Nucleophilic attack occurred at both the 2 and 6 positions of the pyridine ring, giving rise to the desired bromopicolinonitrile **2.42** and its regioisomer **2.65** (Scheme **2.12**). These could be separated readily by flash column chromatography.



2.63 **2.64** **2.42** (80%) **2.65** (10%)

Scheme 2.12: Reaction conditions for generating the nitrile adduct **2.42**.

Reduction of the nitrile functionality was next achieved using the commercially available *N,N*-dimethylethylamine alane complex. Employing the published conditions[3] gave considerable amounts of 2-pyrimidylmethylamine **2.66** (28 - 36% conversion based on crude ^{1}H NMRs), while isolation of **2.43** from the reaction mixture by flash column chromatography proved troublesome, with the product isolated in 25% yield (Scheme **2.13**).

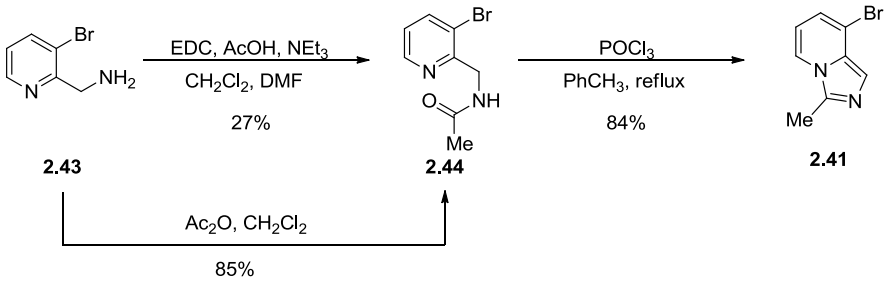


Scheme 2.13: Nitrile reduction using alane.

It was hoped that substitution of the alane complex with a borane reagent might limit over reduction of **2.42** to **2.66**. However, only starting material was observed when a mixture of **2.42** and BH_3•SMe_2 complex were left to stir at room temperature for an hour. Since reduction of nitriles with borane is known to be sluggish,[24] the reaction mixture was next heated at reflux causing a dramatic change in the colour of the solution and the formation of a myriad of products as evidenced by TLC. Consequently, this route was abandoned due to problems associated with the formation and purification of amine **2.43**.

It has been reported in literature that reduction of nitriles is best affected using alanes.[24] Hence, the original conditions were re-examined. Attempts to limit the amount of **2.66** formed by performing the reaction at low temperatures (–78 °C) were unsuccessful, with only the starting material recovered. However, reducing the amount of alane used to 1.1 equivalents and allowing the reaction mixture to warm to room temperature slowly proved fruitful, with **2.43** synthesised in 49% yield. In seeking to improve the yield further, the work-up was modified to remove the methanol quench prior to extraction. Pleasingly, this change enhanced the yield of amine **2.43** to 81%.

Amine **2.43** was then converted to amide **2.44** with the use of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC). Initially, a mixture of **2.43** and **2.66** was acetylated. It was thought the reaction had proceeded reasonably, with an estimated yield of 44% (based on ratio of products **2.43** and **2.66** respectively, as determined by NMR). However, when amine **2.43** was isolated, the EDC acetylation step was found to give only 27% yield of amide **2.44**. Various conditions were screened, with the best results obtained using acetic anhydride (Table **2.4** and Scheme **2.14**).

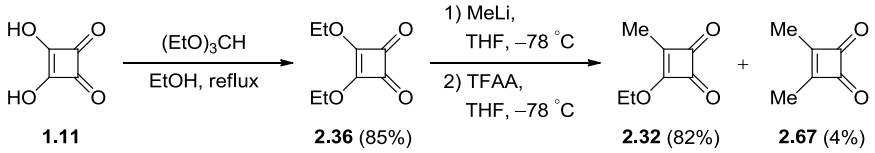


Scheme 2.14: Formation of imidazopyridine **2.41**.

Entry	*Acetylating Conditions*	*Yield / %*
1	EDC, AcOH, NEt_3	27
2	Acetic anhydride + DMAP	20
3	Acetic anhydride	85

Table 2.4: Acetylation conditions of **2.43**.

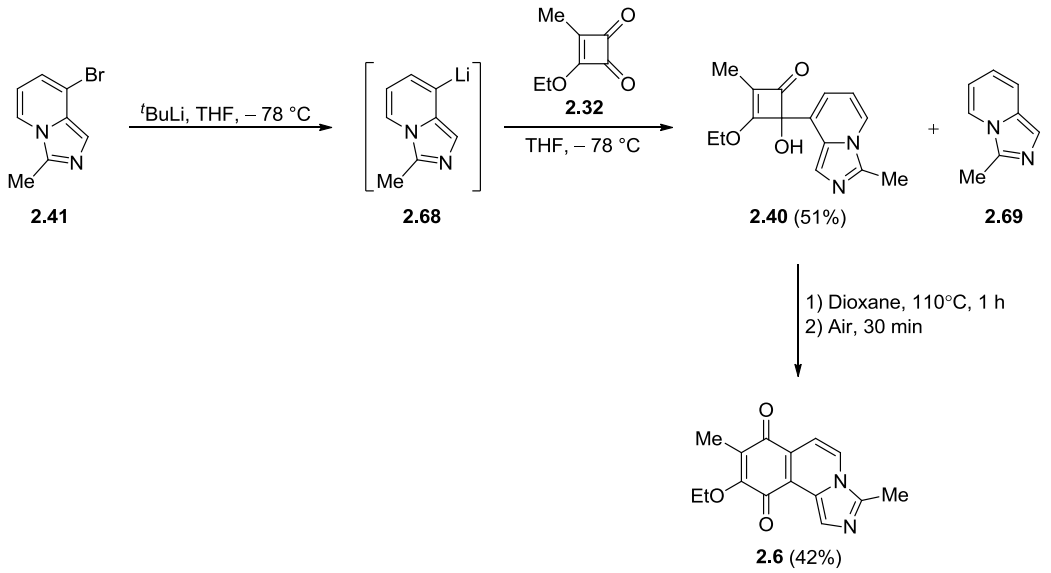
Bischler-Napieralski cyclisation of acetamide **2.44** using $POCl_3$ gave the desired heterocycle **2.41** cleanly and in good yield (89%) (Scheme **2.14**). With this intermediate in hand, our focus shifted towards the cyclobutenedione component required in the key addition step. This was prepared from squaric acid **1.11**, *via* diester **2.36**, according to the literature procedure (Scheme **2.15**).[5, 25]



Scheme 2.15: Synthesis of cyclobutenedione **2.32** from squaric acid.[5, 25]

Diethyl squarate **2.36** was isolated in 85% yield after flash column chromatography and was subsequently reacted with methyllithium (MeLi), yielding both the desired cyclobutenedione **2.32** and the unwanted adduct **2.67**. This side product stems from a second addition of MeLi to adduct **2.32**. By lowering the concentration of **2.36** and transferring the organolithium slowly at –78 °C, cyclobutenone **2.32** could be isolated (82%) with only trace contamination of the double addition product **2.67**.

Reaction of **2.32** with *in situ* generated organolithium **2.68** was then attempted (Scheme **2.16**). This did not yield the desired product. Instead, the material isolated was consistent with the protiodehalogenated imidazopyridine (**2.69**), which was thought to arise from reaction of the lithiated pyridine species with the butyl bromide formed during halogen-lithium exchange.



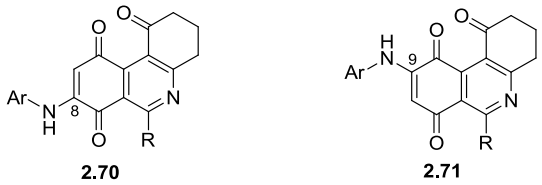
Scheme 2.16: Final steps in the synthesis of cribrostatin 6.

In order to circumvent this side reaction, ^{t}BuLi was used in place of ^{n}BuLi, then the cyclobutenedione was added to the resulting organolithium solution.[26] These conditions did yield the desired product in 51% yield, which was comparable to that published by Harrowven *et al.*[3] The other regioisomer **2.45** (Scheme **2.8**), derived from addition of the organolithium to the vinylogous ester carbonyl, was not isolated or visible in the NMR spectrum of the crude product mixture.

Finally, Cribrostatin 6 was prepared by thermolysis of cyclobutenone **2.40** in dioxane under flow conditions, followed by aerial oxidation. From NMR analysis, thermolysis of **2.40** had gone to completion yet the target molecule was isolated in 30% yield after flash chromatography. The low yield was thought to be due to solubility issues as a 0.1 M solution of **2.40** was used instead of the more commonly used 0.05 M.[3] The reaction was repeated in a round-bottomed flask but the yield could only be improved to 42%.

Cribrostatin 6 Analogues

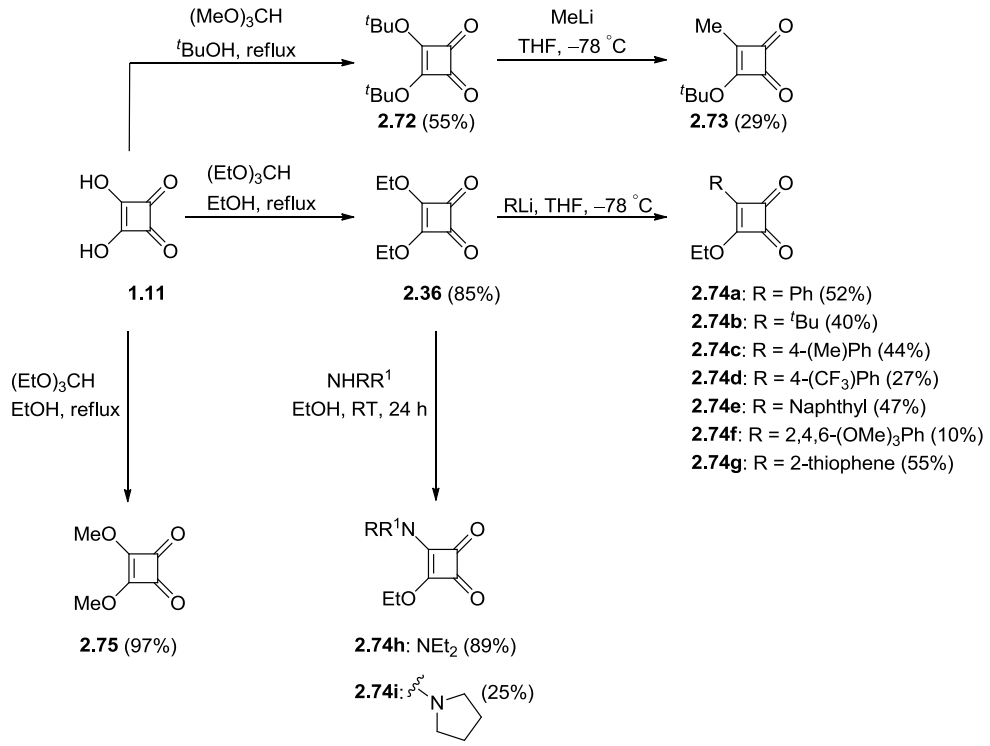
Following this, the synthesis of analogues was instigated. Emphasis was placed on installing aromatic as well as amino substituents as the latter had been shown by Valderrama *et al.*[27] to increase cytotoxicity when present at position 8 rather than 9 of isoquinoline quinone systems (Figure **2.4**). In addition, several natural products contain this structural motif, with notable examples in the cribrostatin family (Figure **2.1**).



2.70 **2.71**

Figure 2.4: 8 and 9- substituted phenanthridinequinone studied by Valderrama *et al.*[27]

Synthesis of the requisite cyclobutenediones was accomplished following established procedures (Scheme **2.17**).[5, 25] For the aromatic substituted cyclobutenediones, these were generally attained in yields ranging from 10 to 55% and were in line with published data.[25] Attempts to increase the yield of **2.74d** by using the two step procedure led to marginal improvements in the yield (23 to 27%) but the advantage was insufficient for us to abandon the "one pot" procedure. As for the monosquaramides, adduct **2.74h** was prepared in high yield, in stark contrast to the pyrrolidine analogue **2.74i** (Scheme **2.17**).

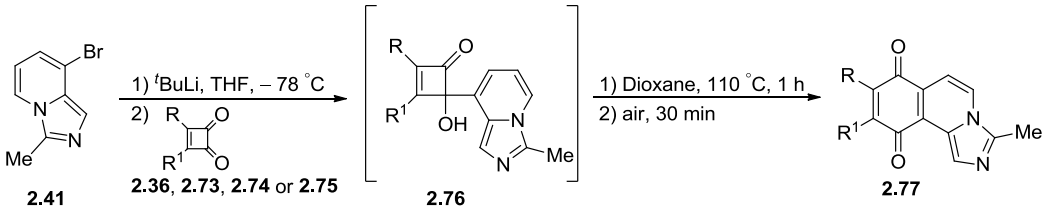


Scheme 2.17: Synthesis of various cyclobutenediones.

These cyclobutenediones were then reacted with the organolithium formed *in situ* from imidazopyridine **2.41** on treatment with ^{t}BuLi (Scheme **2.18**). In this way cyclobutenone adducts **2.76a** through **2.76l** were each prepared. Initially, due to the well-documented instability of cyclobutenones, the crude reaction mixtures were thermolysed directly (**2.76a**, **i** and **l**). However, it proved beneficial to carry out a quick chromatographic purification to remove impurities prior to the Moore rearrangement. In most cases, the cyclobutenone

adducts **2.76** were isolated in yields ranging from 16 to 63%. Thermolysis in dioxane at 110 °C under flow then resulted in the desired quinones **2.77**, albeit in low yield (2 - 32%, Scheme **2.18**). No attempt was made to optimise the reaction as only small quantities of material were required at this stage for biological evaluation.

Only two of the prepared adducts failed to furnish the corresponding quinone. For thienyl cyclobutenone **2.74g**, the crude product mixture could not be purified, while in the case of the *N,N*-diethylamino analogue **2.74h** no rearrangement had taken place after 23 h at 110 °C. Consequently, the thermolysis was repeated at 150 °C for 1 hour using the flow system. NMR analysis suggested that rearrangement may have taken place but only a trace amounts of what we believed to be the desired product **2.77i** was retrieved after purification by flash chromatography, and this could not be improved upon. Since these compounds may be attained from the quinone directly (as demonstrated by Martin *et al.*), we decided to address their synthesis at a later date should the biological evaluation return favourable activity in similar tested analogues.[22]



a: R = Ph, R^1 = OEt (18%); **b**: R = ^{t}Bu, R^1 = OEt (19%); **c**: R = 4-(Me)Ph, R^1 = OEt (20%); **d**: R = 4-(CF_3)Ph, R^1 = OEt (4%); **e**: R = Naphthyl, R^1 = OEt (28%); **f**: R = 2,4,6-$(OMe)_3$Ph, R^1 = OEt (32%); **g**: R = 2-thiophene, R^1 = OEt (0%); **h**: R = OEt; R^1 = NEt_2 (0%); **i**: R = Me, R^1 = O^{t}Bu (8%); **j**: R = O^{t}Bu, R^1 = Me (16%); **k:** R = R^1 = OEt (2%); **l**: R = R^1 = OMe (14%)

Scheme 2.18: Analogues of cribrostatin 6 prepared.

After HPLC purification, the compounds were screened against MCF-7 breast cancer cells (work performed by Dr Asby as part of collaboration with Professor Tavassoli). Pleasingly, all showed IC_{50} values in the nM to μM region (Table **2.5**), with two analogues found to be more potent than the parent natural product (**2.77a** and **2.77k** respectively, entries 2 and 10 in Table **2.5**). Although the pool of analogues prepared was small, some structure-activity trends can be inferred.

In terms of the quinone substituents, the presence of a ^{t}butoxy group at position 8 rather than 9 on the crisbrostatin 6 skeleton results in a decrease in cytotoxicity, although two ethoxy substituents increases cytoxicity (entries 8, 9, and 10, Table **2.5**). This suggests that it is favourable to have an alkoxy residue at position 9 rather than 8 on the cribrostatin 6 ring system, and that the synthesis of a regioisomeric series where the alkoxy group is at position 8 on the quinone unit is unlikely to deliver more potent analogues.

With respect to the aromatic series, the phenyl analogue (**2.77a**) proved to be the most active compound synthesised. From the analogues prepared, it is clear that adding complexity to the phenyl ring results in a loss in cytotoxicity, with IC_{50} values increasing from 428 nM to 0.99 - 2.02 μM region. In terms of the role electronic and steric effects have on the system, both factors appear to influence the biological properties of this molecule to some extent. Increasing the steric demand of the aromatic substituent results in a decrease in cytotoxicity, as exemplified by comparing analogues **2.77a** with **2.77c** or **2.77d** (entries 2, 4 and 5, Table **2.5**), or **2.77a** with either **2.77e** or **2.77f** (entries 2, 6 and 7, Table **2.5**). This is also apparent for non-aromatic substituents when one compares **2.6** with **2.77b** (entries 1 and 3, Table **2.5**). As for electronic effects, data seems to indicate that aryl substituents with an electron withdrawing group in the 4 position of the phenyl ring (**2.77d**) lowers activity to a greater extent than an electron donating group at the same position (**2.77c**), although more analogues need to be prepared to properly establish a correlation (entries 4 and 5, Table **2.5**).

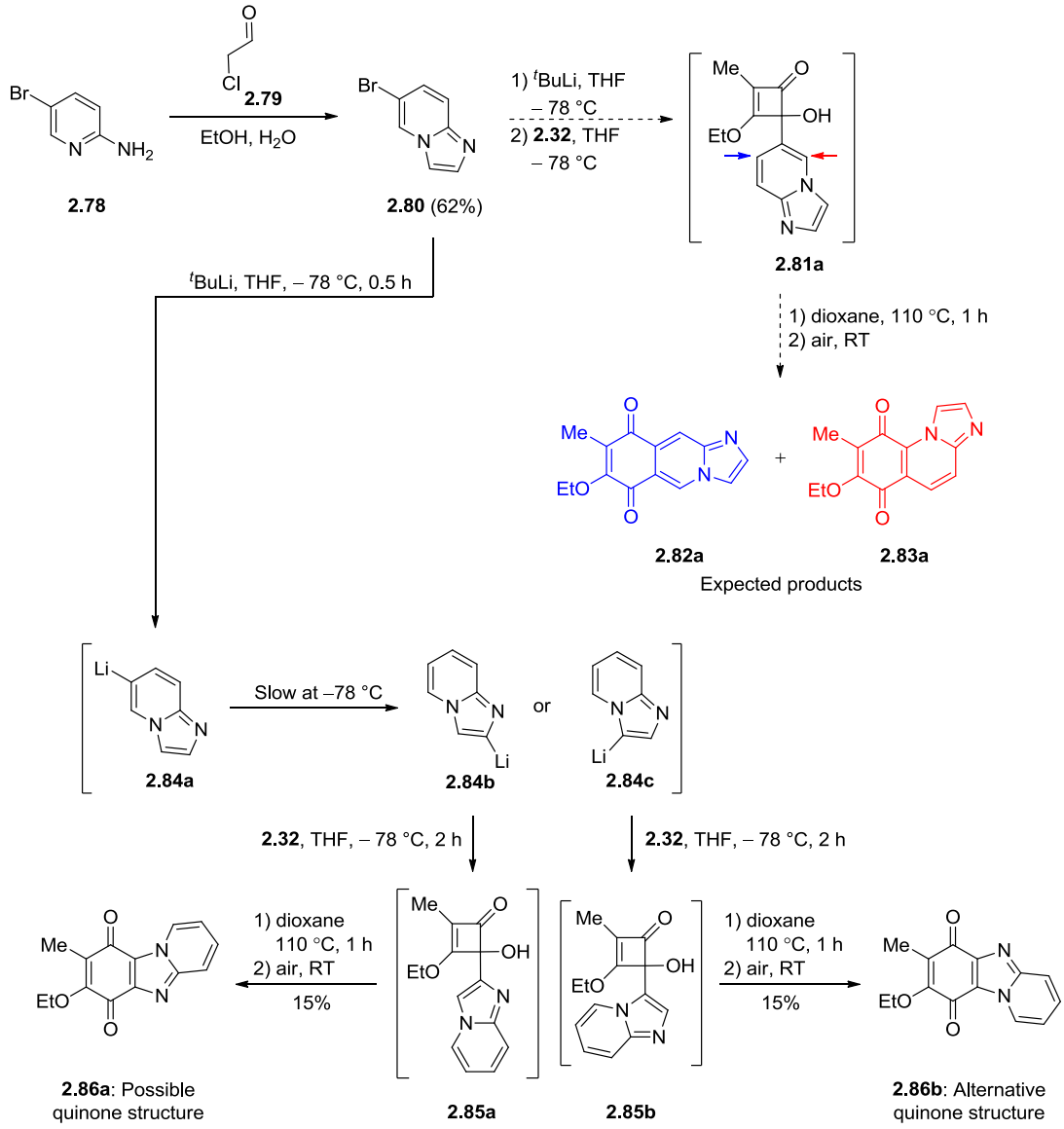
Entry	*Compound*	*IC_{50} (nM) against MCF-7 breast cancer cell line*
1	*Cribrostatin 6 (**2.6**)*	628 ± 67
2	***2.77a***	428 ± 40
3	***2.77b***	1671 ± 112
4	***2.77c***	993 ± 186
5	***2.77d***	1661 ± 143
6	***2.77e***	2024 ± 156
7	***2.77f***	1692 ± 160
8	***2.77i***	1546 ± 139
9	***2.77j***	2064 ± 153
10	***2.77k***	253 ± 43

Table 2.5: IC_{50} measured by Dr Asby for cribrostatin and its anologues.

Alternative core skeleton to cribrostatin 6

Having synthesised a small number of analogues of cribrostatin 6 based around the quinone core, focus shifted towards changing the heterocyclic partner used within the addition step. From previous work, it has been proposed that the imidazo subunit in cribrostatin 6 is essential.[21] Apart from replacing the methyl group present on ring C, experimental work has not explored the nature of the BC ring system. Thus, it was envisaged that heterocycle **2.80** would replace imidazopyridine **2.41** (Scheme **2.19**).

Imidazopyridine **2.80** was obtained in 62% yield by refluxing 5-bromo-2-aminopyridine **2.78** with acetaldehyde **2.79** in an aqueous ethanol solution.

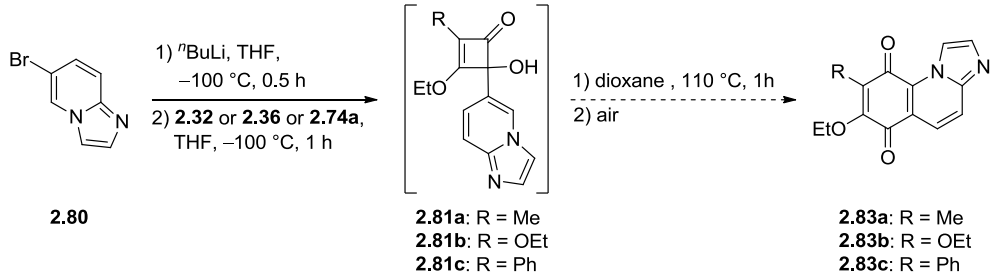


Scheme 2.19: Synthesis of 6-bromoimidazo[1,2-a]pyridine and subsequent reaction with cyclobutneone **2.32**.

Addition to cyclobutenedione **2.32** was then investigated. This was expected to give **2.81a** after addition of the *in situ* formed lithium species **2.84a**, which after thermolysis could potentially give both quinone **2.82a** and **2.83a** (Scheme **2.19**). In reality, NMR data acquired for the major product isolated after thermolysis was inconsistent with that expected for either compound **2.82a** or **2.83a**. Specifically, COSY NMR spectroscopy showed coupling between the aromatic protons suggesting the presence of a benzo[4,5]imidazo[1,2-a]pyridine-6,9-dione core, and that quinone **2.86a** or **2.86b** had been formed. Unfortunately, the exact structure of **2.86** could not be confirmed by X-ray

crystallography as the diffraction pattern obtained was too diffused and therefore some degree of uncertainty exists as to the exact structure of the product isolated.

Compound **2.86** could only arise if the initially formed organolithium species **2.84a** undergoes lithium migration generating either organolithiums **2.84b** or **2.84c** (Scheme **2.19**). This reaction was repeated with cyclobutenediones **2.32**, **2.36** and **2.74a** (Scheme **2.20**). It was found that by carrying out the reaction at –100 °C, lithium migration was not observed. Much like before, it proved beneficial to carry out a quick chromatographic purification to remove impurities prior to the Moore rearrangement, with yields of **2.81** ranging from 25 to 34%. Unfortunately, although the major product isolated from the Moore rearrangement of these cyclobutenones was most consistent with **2.83**, the presence of an impurity has hampered conclusive identification of the product isolated. Attempts to remove this impurity by HPLC (both reverse and normal phase) as well as preparative thin layer chromatography (TLC) were to no avail, with work from this section abandoned since no clean sample could be generated for biological evaluation.



Scheme 2.20: Attempted synthesis of quinone **2.83**.

Conclusions and future work

Cribrostatin 6 is an intriguing natural product both structurally and in terms of its biological properties. Four total syntheses have been described to date and a number of analogues have been prepared. Focussing primarily on aromatic analogues of this natural product, ten compounds were synthesised and screened against the breast cancer cell line MCF-7 (as part of collaborative work with Professor Tavassoli's group) with three analogues exhibiting submicromolar activity.

From the analogues prepared, steric factors seem to play an important role in respect of biological activity. Replacing the methyl group on the quinone unit in cribrostatin 6 with anything other than a phenyl or an ethoxy residue generally leads to a reduction in cytotoxicity. In addition, switching the position of a *t*butoxy group from 9 to 8 on the cribrostatin 6 skeleton results in a less potent analogue. While for the aryl analogues,

adding an electron donating group (EDG) at the 4 position of the phenyl ring reduces cytotoxicity, albeit to a lesser extent than adding an electron withdrawing group (EWG). It should be stressed that these observations are made by comparison within a relatively small sample group so may conceal a more complex trends.

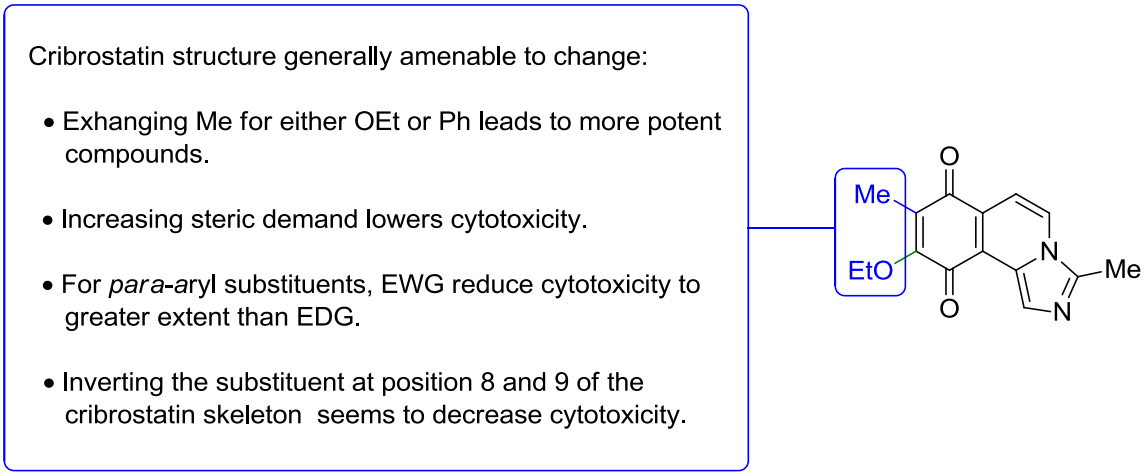
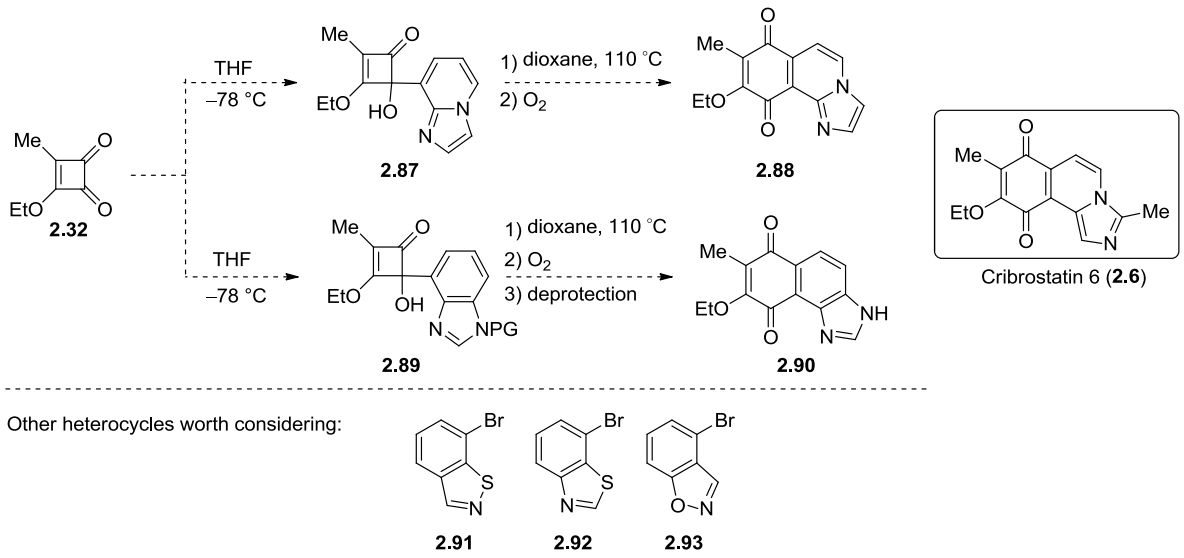


Figure 2.5: Observation from biological evaluation of cribrostatin 6 analogues

Further work is needed to consolidate and expand on the trends observed here. More analogues varying both the electronic and steric demands of the system are needed to confirm the tentative conclusions drawn from this preliminary study. This is also true in respect of changes to substituents 8 and 9 on the quinone unit. In addition, the introduction of an amino group on the quinone unit may be interesting to study, given the preliminary work carried out by Valderama *et al*.[27]

Finally, the least studied part of the cribrostatin 6 framework is the imidazopyridine unit and its importance regarding cytotoxicity towards cancer cells. This could take the form of replacing the imidazo[5,1-a]pyridine unit with heterocycles such as imidazo[1,2-a]pyridine (leading to quinone **2.88**), benzo[d]imidazole (giving quinone **2.90**), benzoisothiazole **2.91** and benzothiazole **2.92** or benzoisoxazole **2.93** to name but a few. In this way we could provide information on unique structures hitherto unprecedented in nature (Scheme **2.21**).



Scheme 2.21: Modification of the cribrostatin 6 framework.

Chapter 3: Alpkinidine

In 2002, Crews *et al.* extracted from the sponge *Xestospongia carbonaria* a novel marine natural product which they named alpkinidine (**3.6**).[28] It possesses a rare pyrroloacridine ring system, which is similar to that of the plakinidine family of natural products (Figure **3.1**). It has been shown by Crews *et al*. to be selective towards solid tumours cells although to lesser extent than other acridine natural products.[28] However, the small quantity of material isolated thus far has prevented a more in depth analysis of alpkinidine's biological properties.

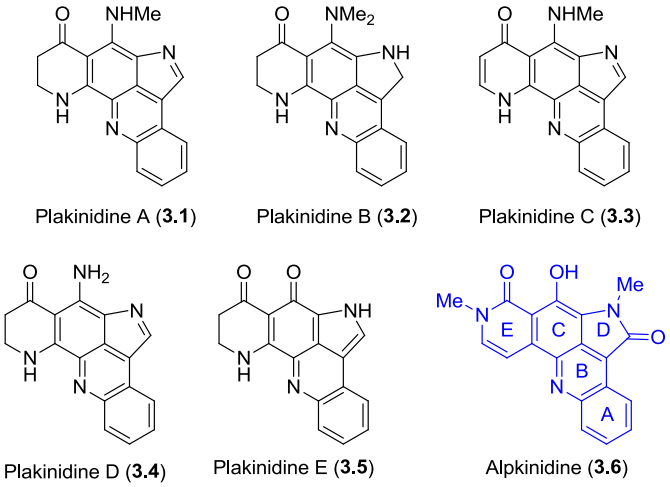


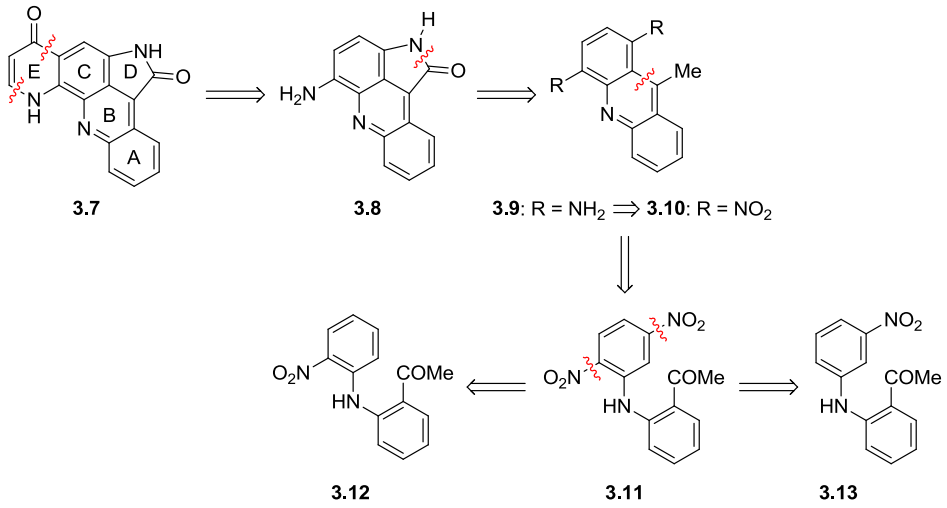
Figure 3.1: Pyrroloacridine natural products.[28]

Synthetic studies towards the plakinidine family and alpkinidine

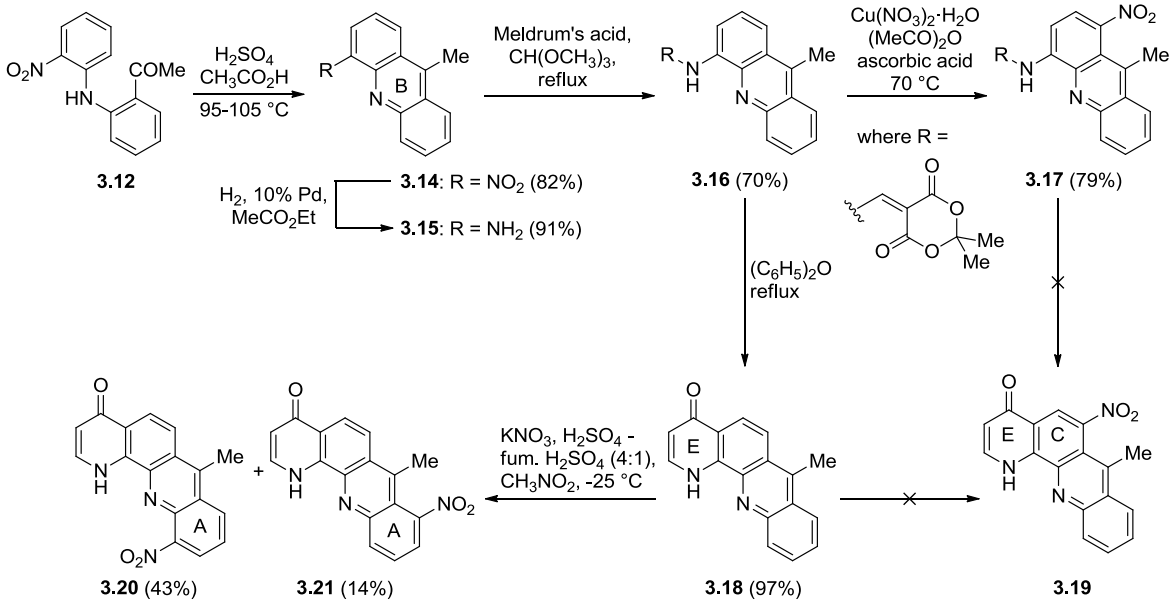
To date, there has been no reported total synthesis of alpkinidine or any of its congeners. That said, there have been several studies towards the pyrroloacridine cores of these six secondary metabolites. The first of these was developed by Kitahara *et al*. in 2004.[29] Their aim was to target the general pyrroloacridine core **3.7** (Scheme **3.1**). The strategy they employed lay in establishing the ABC ring system first via electrophilic aromatic substitution (S_EAr) from either **3.12** or **3.13**. Both the D and E rings would then be added to the core structure successively. With respect to the D ring, this would be achieved by oxidation of the methyl functionality using SeO_2. As for ring E, thermal cyclisation of Meldrum's acid derivative of **3.8** would generate target compound **3.7**.

Initially, **3.12** was preferred to assemble the ABC ring system (Scheme **3.2**). Acridine **3.14** was generated in 82% yield and subsequently converted to aminoacridine **3.15** in 91% yield. This was then reacted with Meldrum's acid and trimethyl orthoformate to form **3.16**

in 70%. From **3.16**, nitration to **3.17** proceeded in high yield, however cyclisation to install ring D proved unsuccessful (Scheme **3.2**). Installation of ring E was possible, e.g. **3.16** to **3.18**, but then nitration of **3.18** occurred in ring A rather than the desired ring C (Scheme **3.2**).



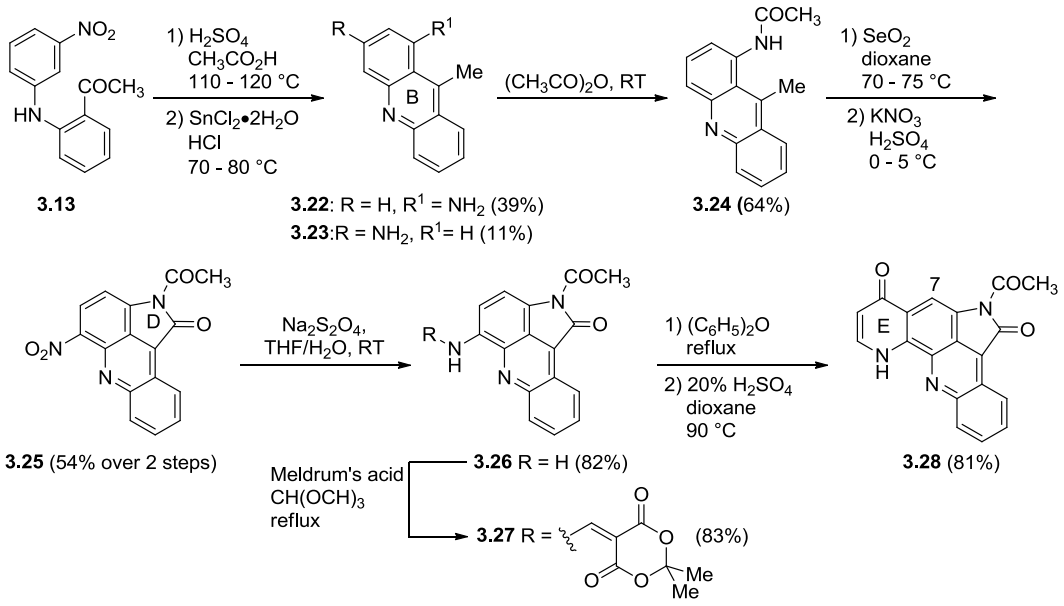
Scheme 3.1: Synthetic strategy to pyrroloacridine model core system **3.7** of the plakinidine family.[29]



Scheme 3.2: Synthetic route to plakinidine model target **3.7** starting from 1-(2-((2-nitrophenyl) amino) phenyl)ethanone **3.12**.[29]

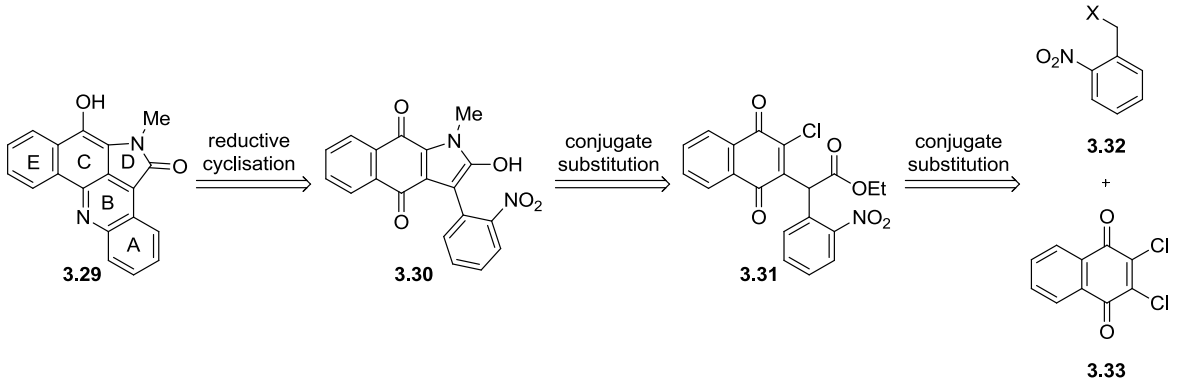
Since nitration of ring C could not be achieved via this route, Kitahara *et al*. opted to begin with nitrodiphenylamine **3.13** with the ring C nitro functionality already in place (Scheme **3.3**). From **3.13**, acridine **3.22** could be prepared, albeit in a much lower yield

than with the previous route (39% over 2 steps). The amino functionality on ring C was then acylated to give acridine **3.24** in 64%. Installation of ring D using SeO_2 followed by nitration of ring C, resulted in pyrroloacridine **3.25** in 54% yield over 2 steps. Reduction of the nitro group was carried out next giving aminoacridine **3.26**, ready to form ring E. This was achieved by reacting **3.26** with Meldrum's acid and trimethyl orthoformate, forming compound **3.27**, which was cyclised to generate **3.28** in 7.5% overall yield. Although Kitahara *et al*.[29] establish the ABD ring system of alpkinidine, ring C lacks the requisite hydroxyl group at C7 (Scheme **3.3**). Moreover, ring E is incorrect for alpkinidine. Lastly, the low yielding first step and the linear nature of the route make this approach less attractive.



Scheme 3.3: Synthetic route to pyrroloacridine target **3.28**.[29]

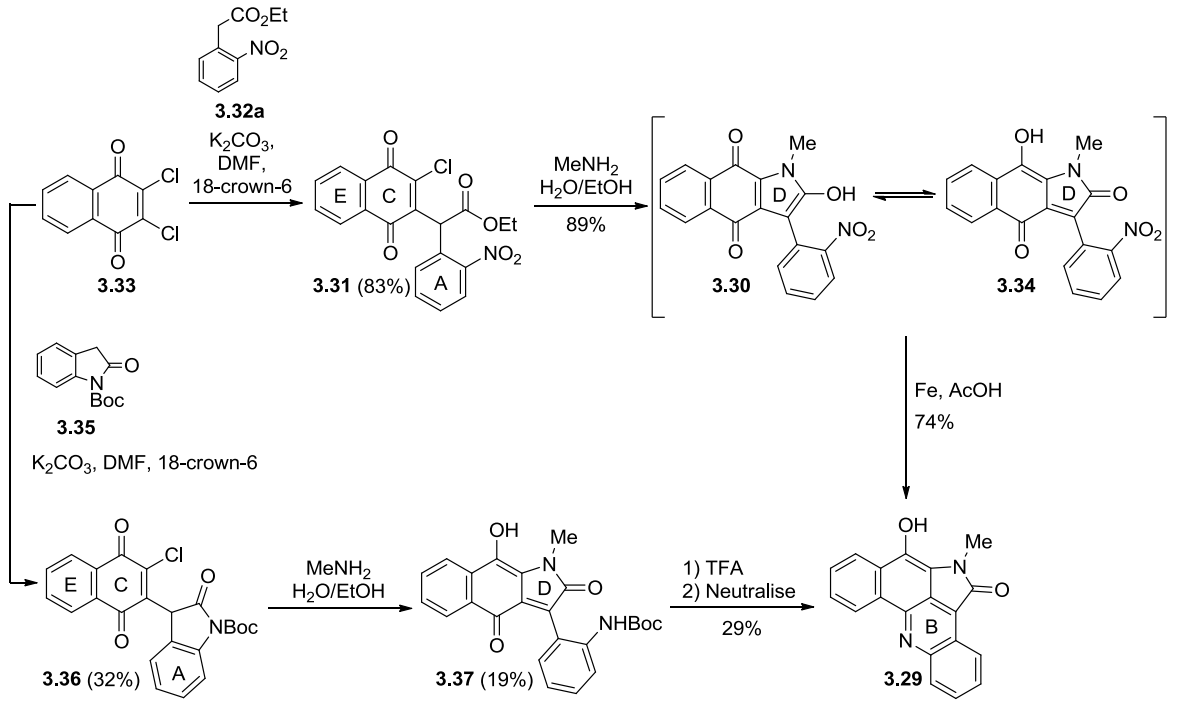
More recently, Piggott *et al*.[30] decided to prepare a model system of alpkinidine whereby the methylpyridone ring (ring E) was substituted for a benzene ring (Scheme **3.4**). Their approach was to start with the EC ring unit in place using naphthoquinone **3.33**, with subsequent conjugate substitutions used to install first ring A, then ring D (Scheme **3.4**). With the ACDE rings in place, only ring B remained and this could be attained through reduction of the nitro functionality.



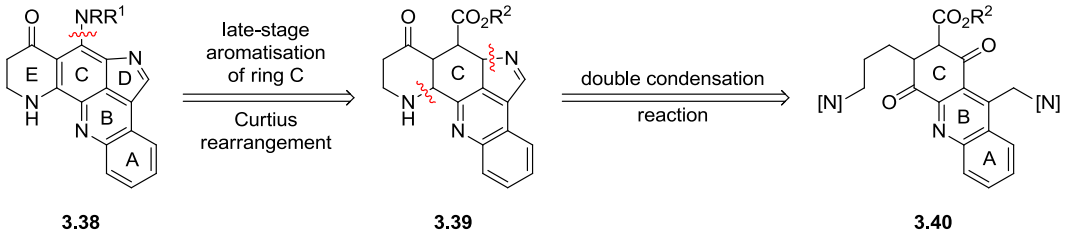
Scheme 3.4: Retrosynthetic analysis alpkinidine's model system by Piggott *et al.*[30]

To that end, starting from 2,3-dichloronaphthaquinone **3.33**, conjugate substitution with nitroester **3.32a** furnished the AEC ring system of the alpkinidine target in 83% (Scheme **3.5**). Treatment of **3.31** with $MeNH_2$ gave a mixture of tautomers **3.30** and **3.34** in approximately 4 : 1 ratio in favour of quinone **3.30**. Next, reduction of the nitro functionality afforded target **3.29** in 55% overall yield (Scheme **3.5**). The synthesis of the alpkinidine target was also achieved using oxindole **3.35**. Alas, the 2% overall yield obtained using this route was considerably lower than the previous route for the same number of steps (Scheme **3.5**).

In 2015, Fukuyama *et al.* proposed a route to the plakinidine family.[31] Their plan was to assemble the ABC core structure of the plakinidine family first (**3.40**), with both the E and D rings installed via condensation reactions (Scheme **3.6**).



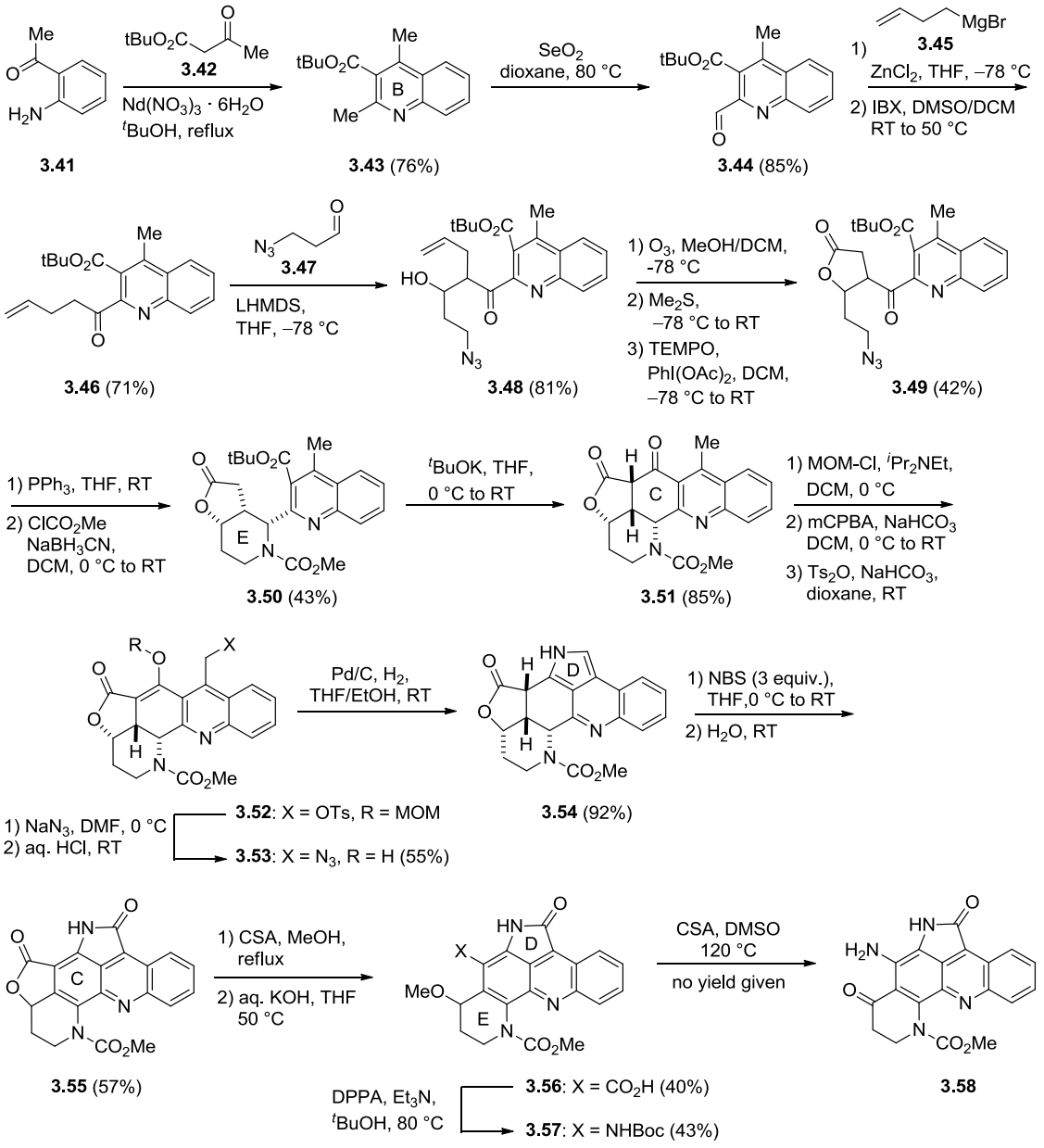
Scheme 3.5: Synthetic routes to alpkinidine developed by Piggott *et al*.[30]



3.38 **3.39** **3.40**

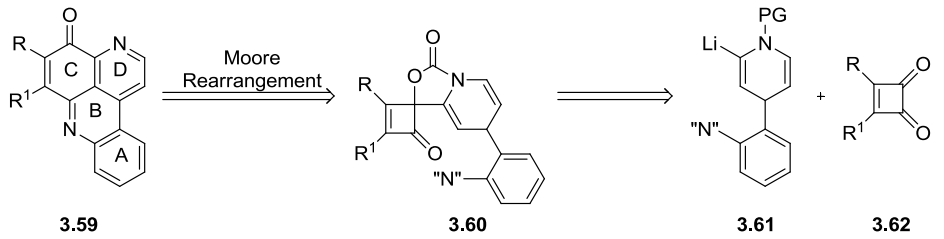
Scheme 3.6: Retrosynthetic analysis by Fukuyama *et al*.[31]

Their synthesis was started by generating the AB ring system **3.43** from 2-aminoacetophenone **3.41** using the Friedländer quinoline synthesis (Scheme **3.7**). Installation of an azide sidechain off ring B of **3.46**, allowed for ring E to be formed using the Staudinger reaction in 21% overall yield from quinoline **3.43**. Once this was achieved, a Dieckmann condensation of lactone **3.50** furnished ring C (**3.51**). Ring D was introduced by a second reduction of an azide functionality installed on ring B from **3.52** to **3.53** (Scheme **3.7**). Aromatisation of ring C followed, and was achieved in 57% yield through the oxidation of acridine **3.54**. Finally, ring opening of lactam **3.55** and subsequent Curtius rearrangement of **3.56** furnished the ABCDE ring system of the plakinidine family in 1% overall yield, with a carbonyl functionality in ring D and ring E still needing minor modifications. It should be noted that treatment of pyrroloacridine **3.57** with camphorsulfonic acid in DMSO resulted in the formation of **3.58** which closely resembles plakinidines A,B and D. However, the data provided for **3.58** is limited to ^{1}H NMR and LRMS, with no yield given (Scheme **3.7**).



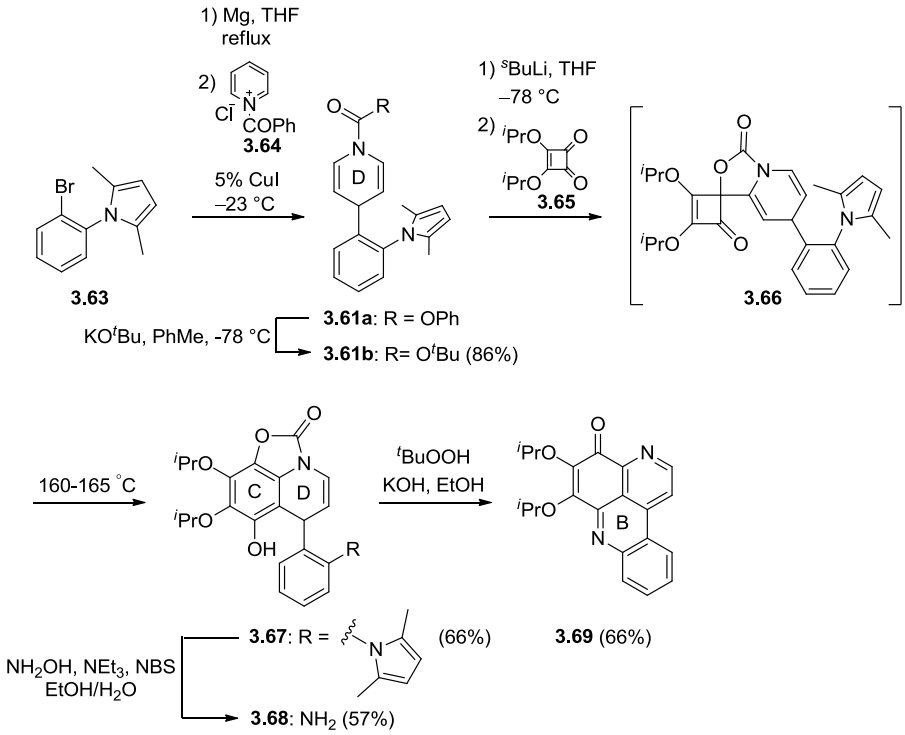
Scheme 3.7: Synthetic route to the core structure of the plakinidine family.[31]

To conclude, the synthesis of pyridoacridine **3.59** by Liebeskind *et al*. is worth mentioning since the motifs are related and natural products belonging to both families have been isolated from the same sponge (Scheme **3.8**).[28, 32] Their strategy employed the Moore rearrangement to generate the CD ring system. This could be achieved from cyclobutenone **3.60**, which in turn could be derived from cyclobutenedione **3.62** and dihydropyridine **3.61** (Scheme **3.8**).



Scheme 3.8: Retrosynthetic route to pyridoacridine system developed by Liebeskind *et al*.[32]

Thus protected 2-bromoanaline **3.63** was converted to the corresponding organocuprate reagent to induce its 1,4-addition to pyridinium salt **3.64** forming the required dihydropyridine **3.61b** in 86% (Scheme **3.9**). *Ortho*-lithiation of dihyropyridine **3.61b** using ^{s}BuLi, followed by treatment with diisopropyl squarate and subsequent thermolysis afforded the CD ring system in 66% yield. Their synthesis was completed by unmasking the latent amino group in oxazoloquinoline **3.67** to give pyridoacridine **3.69** in 21% overall yield after oxidation and condensation (Scheme **3.9**).



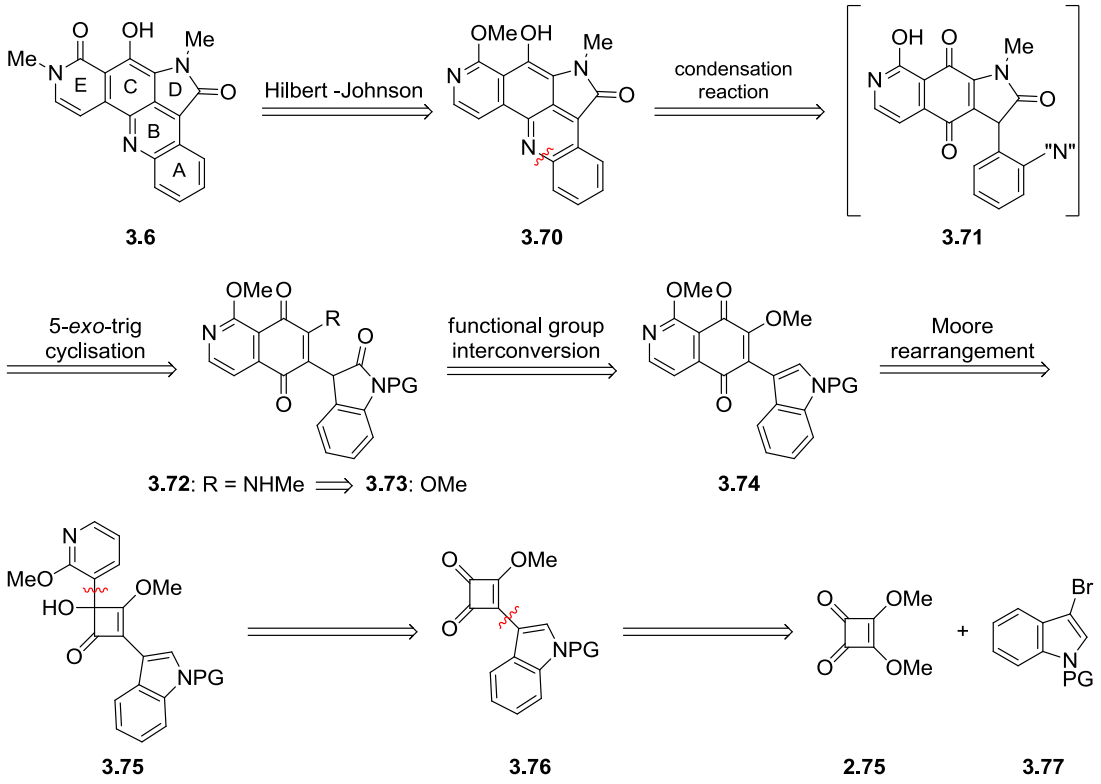
Scheme 3.9: Synthesis of pyridoacridine core using the Moore rearrangement.[32]

Design of a synthetic route to alpkinidine

Both Kitahara and Fukuyama's synthetic strategies could provide the desired ABCD ring systems for alpkinidine, however incorporation of the E ring would remain challenging. Moreover, both are linear and low yielding sequences, making them less attractive for use

in the first total synthesis of alpkinidine. As for the route developed by Liebeskind *et* al., it could ostensibly be adapted to form the ABCD ring system common to the plakinidine family. Yet again, the methylpyridone ring (E) present in alpkinidine complicates matters, with its inclusion by no means trivial.

Instead, taking inspiration from Piggot *et al.*'s succinct methodology, one could conceive constructing the EC ring system using the Moore rearrangement. This could be prepared from the appropriate protected 3-bromoindole (**3.77**) and dimethyl squarate (**2.75**) (Scheme **3.10**). With the EC ring system in place, the indole moiety could be oxidised to form oxindole **3.73**, ready to undergo 5-*exo*-trig cyclisation to the ABCDE pyrroloacridine core for alpkinidine. Finally, conversion of ring E into the requisite methylpyridone could be achieved using methodology developed by Hilbert-Johnson,[33] thus providing the natural product in 7 steps (Scheme **3.10**).

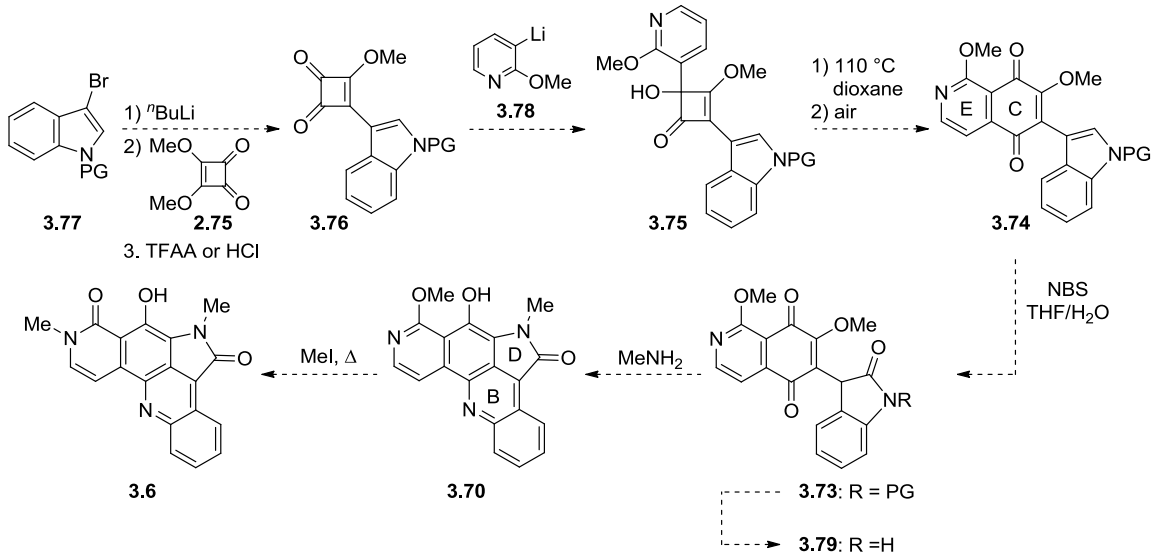


Scheme 3.10: Proposed route to alpkinidine.

Synthesis of alpkinidine

As proposed earlier, we plan to use the Moore rearrangement to establish the EC ring system of alpkinidine. In order to achieve this, indolylcyclobutenediones **3.76** had to be synthesised from dimethyl squarate (**2.75**) and the corresponding 3-bromoindole (**3.77**, Scheme **3.11**). Hence our strategy required the synthesis of a suitably protected 3-bromoindole, capable of withstanding treatment with 3-lithio-2-methoxypyridine as well

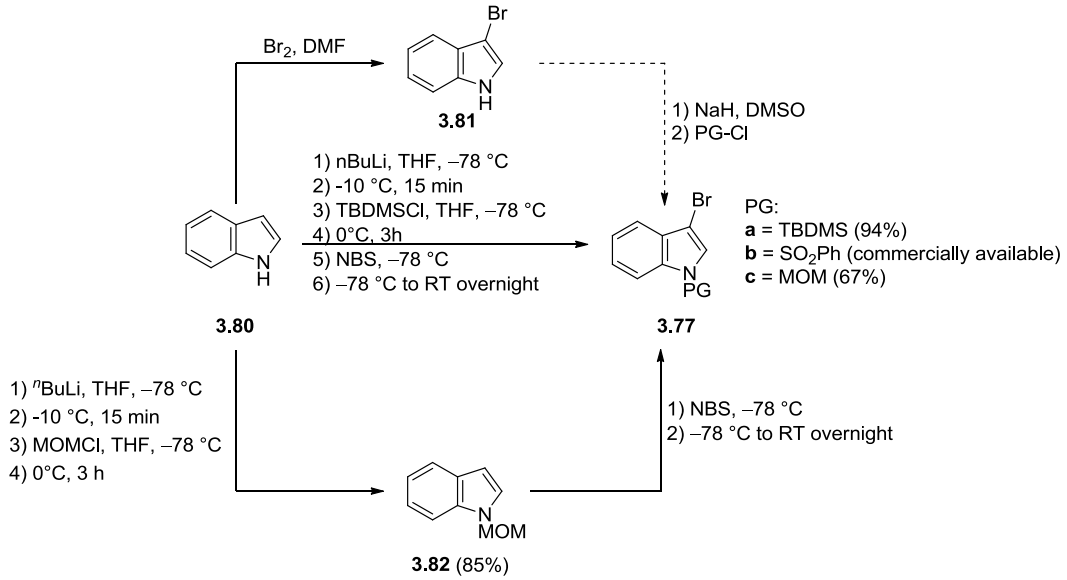
as the acidic conditions required to form cyclobutenedione **3.76**. Additionally, the protecting group must be easily removed in order to facilitate the formation of ring B.



Scheme 3.11: Proposed forward synthesis to Alpkinidine.

To this end a *t*butyldimethylsilyl (TBDMS) group was selected due to its capacity to stabilise the required 3-lithioindole needed to form cyclobutenedione **3.76**, even at 0°C.[34] It should also prove facile to remove in the latter stages of the proposed route. One concern with this protecting group is its potential incompatibility towards trifluoroacetic anhydride (TFAA) and acids as these are the reagents employed to generate cyclobutenedione **3.76**. As an alternative to this protecting group, the methoxymethyl (MOM) and phenylsulfonyl (SO_2Ph) groups seemed appropriate as these are likely to prove more robust than TBDMS.

Synthesis of the TBDMS protected-3-bromoindole was achieved by treating indole **3.80** with *n*BuLi (1.2 equivalent) followed by TBDMSCl and finally *N*-bromosuccinimide (NBS) according to a published procedure.[34-35] This gave direct access to the desired bromoindole **3.77a** in one step in 94% isolated yield (Scheme **3.12**). By contrast, the one pot synthesis of the MOM protected version of **3.77** proved problematic with the compound suffering violent decomposition on standing at room temperature.



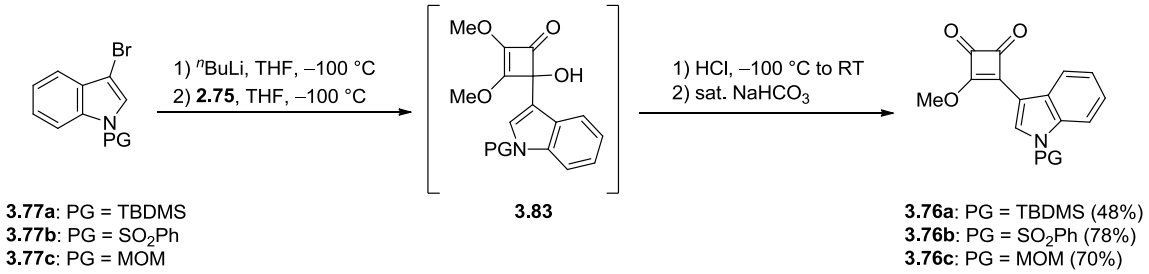
Scheme 3.12: Synthesis of 3-bromoindoles **3.77**.

The procedure used to prepare the TBDMS indole **3.77a** had stated that purification had to be carried out as soon as possible after it is synthesised as traces of 3-bromoindole **3.81** could catalyse decomposition of **3.77** (Scheme **3.12**).[34] To that end, the synthesis of MOM indole **3.77c** was conducted in a stepwise manner and proceeded smoothly, giving indole **3.82** in 85% yield after purification. It was then successfully brominated in 67% yield to give **3.77c** which was found to be stable for 5 days at RT in absence of oxygen. In contrast, unprotected 3-bromoindole **3.81** proved to be highly unstable, decomposing during acquisition of proton NMR. In parallel to this, the synthesis of dimethyl squarate **2.75** was achieved in 94% yield following the protocol developed by Moore *et al*.[5]

With both starting materials in hand, the synthesis of cyclobutenedione **3.76** was attempted. With respect to the TBDMS protected indole **3.77a**, this proved low yielding, with adduct **3.76a** isolated in 10% yield, and the main product observed in the crude NMR was the debrominated indole. A literature search revealed that Hu *et al*., in their total synthesis of asterredione, had carried out a similar reaction using BOC protected 3-bromoindole to form the BOC variant of indolycyclobutenedione **3.76** in 76% yield.[36] Following their protocol, the yield of **3.76a** was doubled to 20%.

In order to rule out quenching of the 3-lithioindole species before it had time to react with dimethyl squarate, the reaction time was extended to 6 h. The yield of **3.76a** was increased to 48%, suggesting we had indeed quenched the reaction too early. Unfortunately, maintaining the reaction at −78 ºC a further 16 h did not improve matters, with **3.76a** isolated in only 25% yield. Since then, yields of **3.76a** have been erratic, with 48% being the highest attained.

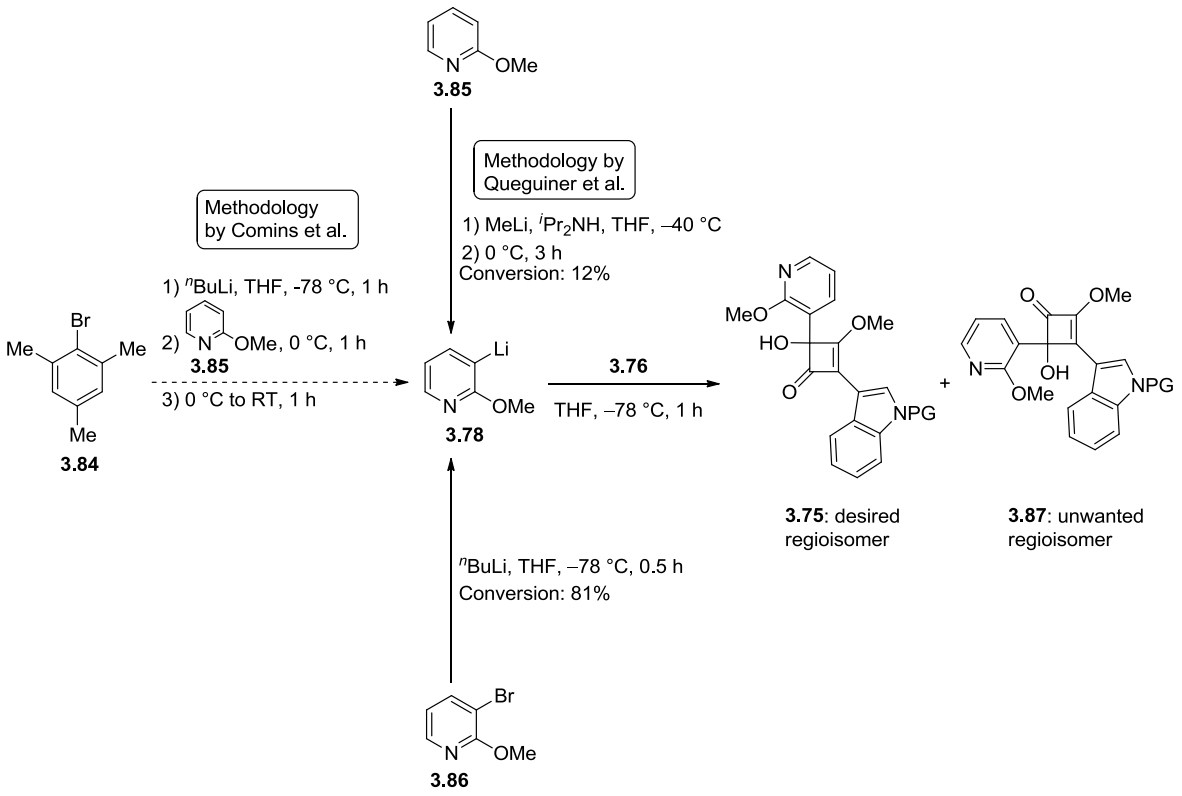
In terms of the commercially available SO_2Ph protected indole, initially only 22% of cyclobutenedione **3.76b** was formed. However, the ^1H NMR of the crude reaction mixture suggested incomplete conversion of the hydroxyl intermediate **3.83b** into cyclobutenedione **3.76b** (Scheme **3.13**). If this was indeed the case then altering the work-up procedure should translate in an improved yield of **3.76b**. It was found that using 5 equivalents of HCl gave adduct **3.76b** in 78% yield. Pleasingly, unlike with the TBDMS indole, the synthesis of **3.76b** proved robust, reproducible and in line with published results.[36] As for the MOM protected indole, forming indolylcyclobutenedione **3.76c** fared much better than with the previous two protecting groups, with **3.76c** isolated in 70% (Scheme **3.13**).



Scheme 3.13: Indolylcyclobutenedione synthesis.

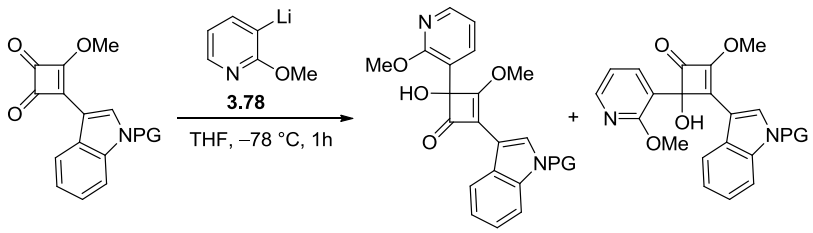
Next, addition of 3-lithio-2-methoxypyridine **3.78** to cyclobutenedione **3.76** was investigated. Since 2-methoxypyridine is commercially available, its addition was examined first. *Ortho*-lithiation of **3.85** has been reported by both Comins *et al*.[37] and Quéguigner *et al*.[38] The first of these methods consists of generating the organolithium species *in situ* by deprotonation with mesityllithium (Scheme **3.14**). This appeared quite convoluted compared to Quéguigner *et al*.'s method which relied on *in situ* generation of lithium diisopropylamine. When this was used to generate **3.78**, cyclobutenedione **3.76** was recovered in 80% yield. In order to establish the root cause for this failure, formation of the organolithium species was tested by quenching the reaction with deuterated methanol instead of cyclobutenedione **3.76**. An ^1H NMR of this test reaction revealed very little deuterium incorporation (12%) at position 3 on the pyridine ring, indicating poor efficiency in direct lithiation, and that an alternative strategy was required.

Halogen-lithium exchange should provide sufficient amounts of the lithium species **3.78** for the addition step. For this, 3-bromo-2-methoxypyridine was prepared as per a literature procedure from 3-bromo-2-chloropyridine through nucleophilic aromatic substitution with sodium methoxide.[39] Pleasingly, pyridine **3.86** was synthesised in 72% yield, comparable with published data.[39]



Scheme 3.14: Methodology to attain 3-lithio-2methoxypyridine.

In order to ensure good conversion to the desired 3-lithio-2-methoxypyridine, the test reaction was also carried out using **3.86**. This was found to be more efficient with 81% deuterium incorporation on quenching with deuterated methanol. Having confirmed the effectiveness in generating the lithiated species **3.78**, the addition reaction was tried with the SO_2Ph protected cyclobutenedione **3.76b**. In this instance, cyclobutenone **3.75b** was isolated in 16% yield (entry 4, Table **3.1**). In order to ensure that the reaction was not being quenched too early, it was repeated and left for 5 h. Unfortunately, this did not improve matters, with the amount of adduct **3.75b** formed estimated to be only 17%. This suggested that the SO_2Ph group may be interfering with the addition process by making the proton at C2 on the indole moiety too acidic.



3.76 | **3.75**: desired regioisomer | **3.87**: unwanted regioisomer

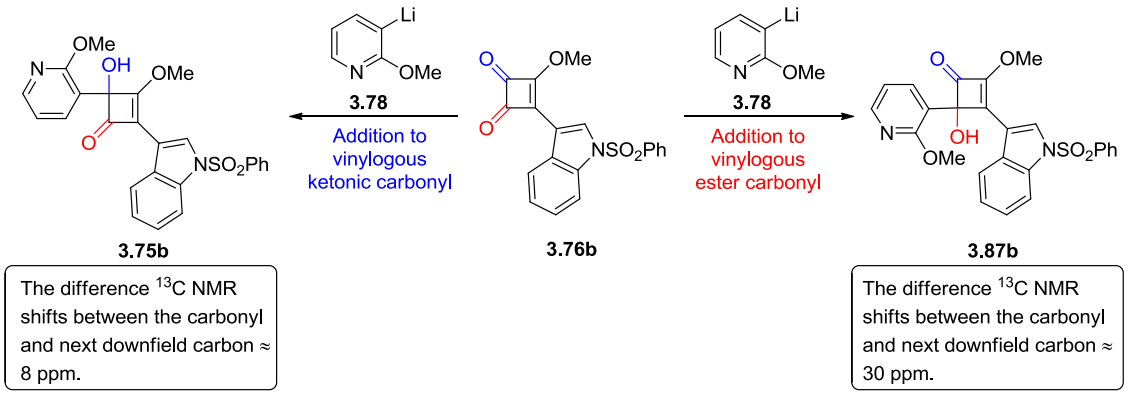
Entry	*PG*	*RLi 3.78 (eq.)*	*Time / (h)*	*% Composition in crude NMR of 3.76*	*% Composition in crude NMR of 3.75 (isolated yield)*	*% Composition in crude NMR of 3.87*
1	TBDMS	1	1	0	100 (75)	Not observed
2	MOM	1	1	40	60 (53)	Not observed
3	MOM	2	1	3	97 (77)	Not observed
4	SO_2Ph	1	1	75	25 (16)	Not observed
5	SO_2Ph	1	5h	83	17	Not observed
6	SO_2Ph	2	1	15	81 (40)	Not observed
7	SO_2Ph	1 (+TMEDA)	1	61	39 (20)	Not observed

Table 3.1: Addition of 3-lithio-2-methoxypyridine to cyclobutenedione **3.76**.

Increasing the amount of organolithium species used should negate this effect. Indeed, when 2 equivalents of organolithium were used, the yield to cyclobutenone **3.75b** increased to 40% (entry 6, Table **3.1**). For completeness, the reaction was carried out in the presence of TMEDA, however a considerable amount of starting material was observed in the crude ^{1}H NMR. Conversion was estimated at 39% whereas 81% conversion to cyclobutenedione **3.75b** was observed when using 2 equivalents of organlithium (Table **3.1**).

As mentioned in Chapter 1, when two different substituents are present in the cyclobutenedione, addition can occur at either carbonyl. From the addition reaction, two possible regioisomers can be formed as shown in Scheme **3.15**. An NOE experiment was conducted in the hope that we could establish which regioisomer had been formed. Unfortunately the identity of the regioisomer formed could not be determined by an NOE experiment. Since an oil had been isolated, no X-ray data could be generated to confirm the structure of the compound. Instead, focus shifted towards published data obtained for similar systems. Interestingly, these showed substantial differences in their ^{13}C NMR. For addition to the more reactive, ketonic carbonyl, a difference of approximately 8 ppm

between the carbonyl shift and the next downfield carbon is observed (Scheme **3.15**). In contrast, this difference increases to 30 ppm for regioisomers akin to **3.87**.[6a]

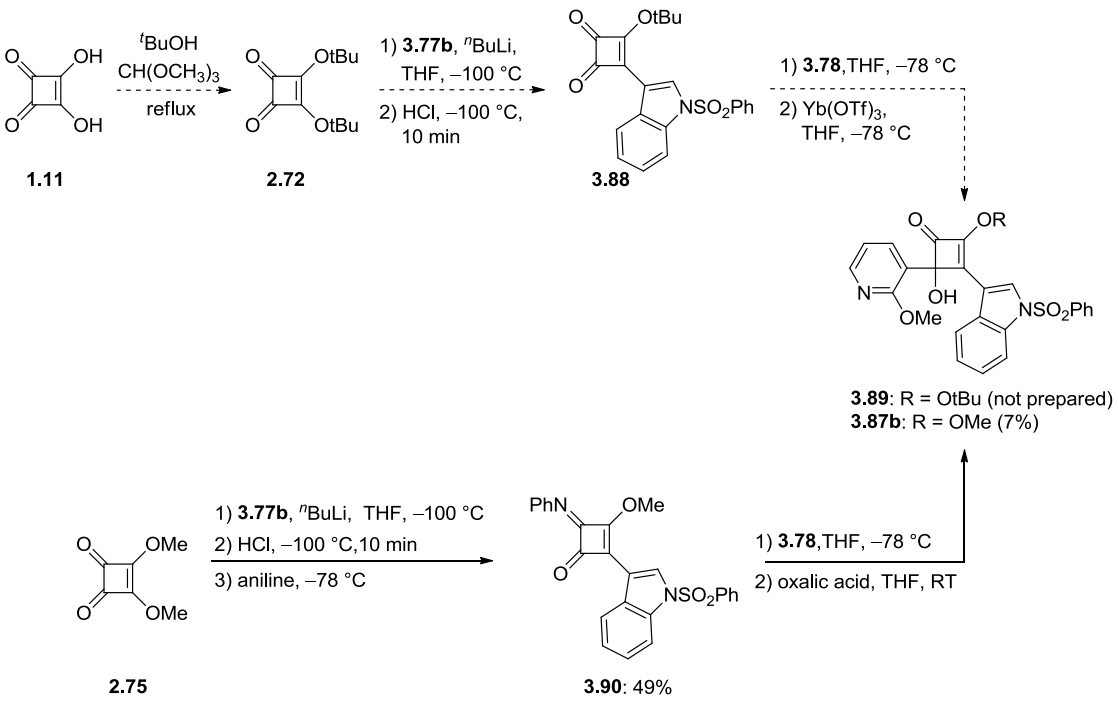


Scheme 3.15: Addition to cyclobutenedione **3.76b**.[6a]

With respect to the product isolated, the difference between the carbonyl and the next downfield carbon is 4.6 ppm, strongly suggesting addition had occurred at the more reactive carbonyl and the desired regioisomer had indeed been formed.

In order to conclusively prove the correct isomer had been formed, synthesis of undesired regioisomer **3.87** was attempted. As mentioned in Chapter 1, this can be achieved in two ways. The use of ytterbium triflate to gain access to cyclobutenone **3.87** would, in theory, be easier to achieve as no modification to the cyclobutenedione is required, nor is there any need for deprotection once addition of the lithium species has been carried out. One of the main drawbacks to this methodology is its current limitation to derivatives of di-^{t}butyl squarate. Hence, this would have to be synthesised. In addition to this, it is uncertain that the ^{t}butoxy group would survive the 5 equivalents of HCl used to form cyclobutenedione **3.88** (Scheme **3.16**).

Synthesis of cyclobutenimine **3.90** proved quite straightforward with **3.90** isolated in 49% yield (Scheme **3.16**).[6c] This was converted into unwanted isomer **3.87b** in 7% yield following a procedure developed by Trost *et al*.[8] ^{13}C NMR of said product differed from that of the previously isolated cyclobutenone product **3.75b**. It showed a difference between the carbonyl and next downfield carbon of 26.6 ppm. This result is consistent with published data, thus confirming the identity of the cyclobutenone product isolated under normal conditions as that arising from addition to the more reactive carbonyl.



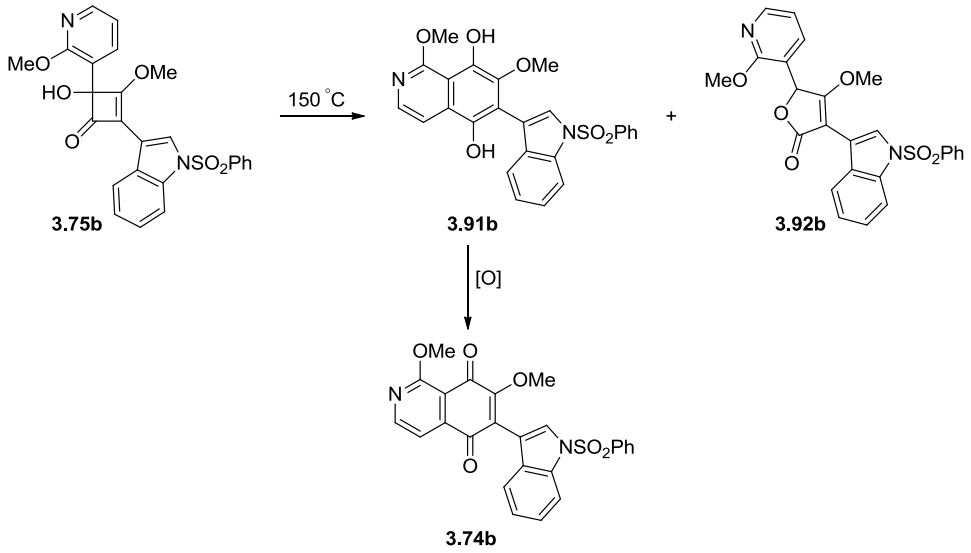
Scheme 3.16: Synthetic routes to unwanted regioisomer **3.87**.

Having established that the desired cyclobutenone **3.75b** was formed, addition was carried out using both TBDMS and MOM protected cyclobutenediones **3.76**. For the TBDMS variant of **3.76**, addition proceeded to give cyclcobutenone in 75% yield using only 1 equivalent of 3-lithio-2-methoxypyridine (entry 1, Table **3.1**). This result seemed to confirm the theory that **3.78** was being quenched by **3.76b**. Similar yields were obtained with MOM cyclobutendione **3.76c** (entries 2 and 3, Table **3.1**). Initially, **3.75c** was isolated in 53% yield, along with 31% recovered cyclobutenedione **3.76c**. By increasing the amount of organolithium used to 1.8 equivalents, the yield of **3.75c** was improved to 77%. The identity of the regioisomer formed for both cyclobutenone **3.75a** and **3.75c** was confirmed by ^{13}C NMR, which were consistent with those observed for the SO_2Ph variant **3.75b**. In addition, based on ^{1}H NMR of the crude reaction mixture, the undesired cyclobutenone **3.87** does not appear to be formed for any of the cyclobutenediones (Table **3.1**).

Having successfully synthesised the desired cyclobutenone **3.75** for each of the indole protecting groups, attention now turned towards thermolysis to generate the EC ring system of alpkinidine. Due to the low yield of cyclobutenedione **3.76a** and difficulties surrounding the synthesis of 3-bromo-1-methoxymethylindole **3.77c**, most of the experimental work on the Moore rearrangement was carried out with the SO_2Ph adduct.

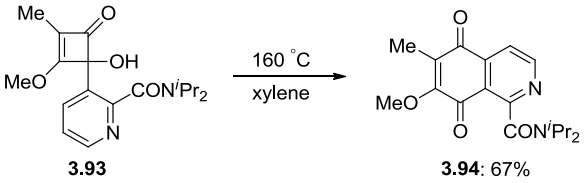
Initially, thermolysis was carried out in a sealed vessel at 110 ºC for 1 h. Surprisingly, only starting material was retrieved. Increasing the temperature to 150 ºC led to the formation of a new compound (major component in the reaction mixture). Characterisation of the

isolated product suggested that this may be hydroquinone **3.91b** (Scheme **3.17**) since 14 protons and 2 OMe signals were evidenced in the NMR spectrum and a molecular ion of 477 was observed by mass spectrometry. With NMR data clearly different from starting cyclobutenone **3.75b**, it was believed that the hydroquinone had been formed instead and that aerial oxidation may be sluggish with this compound. In order to try and force oxidation to the desired quinone, the isolated material was treated with various oxidants, including Ag_2O, CAN and DDQ to name but a few. Unfortunately, quinone **3.74b** was not formed. This raised doubts about the assigned identity of the main thermolysis product and spectral data was thus revisited. The singlet proton observed at 6.24 ppm had been assigned as one of the phenolic protons however, when a D_2O shake experiment was carried out, no loss of signal was observed suggesting this was not an exchangeable proton as first thought. Moreover, 2D NMR experiments showed that this proton was connected to the carbon at 73.7 ppm. Finally, the lack of an OH signal in the IR spectrum confirmed that hydroquinone **3.91b** had not been formed. Since the data was also inconsistent with that of quinone **3.74b**, the only logical outcome was that furanone **3.92b** had been formed. The identity of the major product was confirmed by comparing the acquired data with that of previously published furanones.[4l] Once this had been established, the identity of the minor component of the reaction mixture was identified as the desired quinone **3.74b**. This result was quite unexpected.



Scheme 3.17: Thermal ring expansion of **3.75b**.

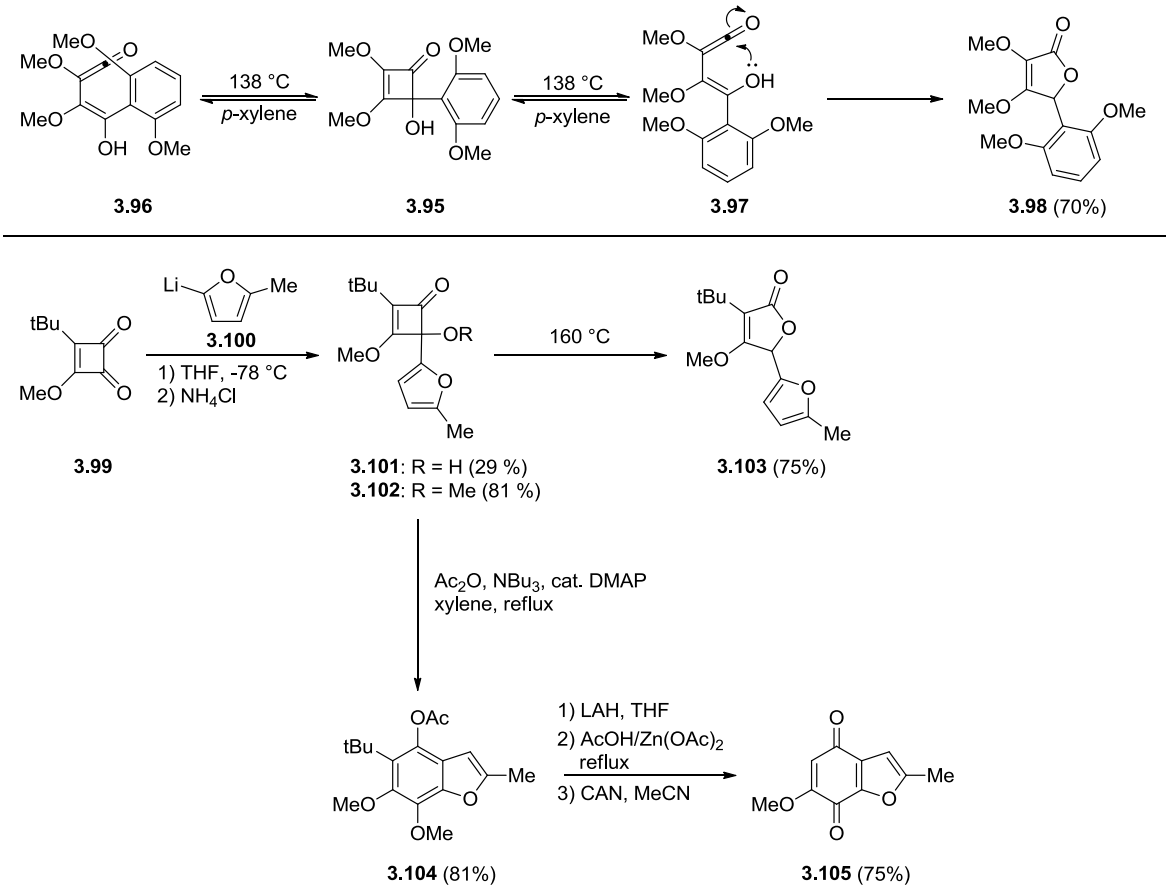
Although in the case of the 3-pyridyl example **1.69** in Chapter 1 a furanone was formed during thermolysis, the quinone products generally comprise the majority of the material isolated (Scheme **1.10**). Here, the furanone product constituted 88% of the material isolated! Moreover, Liebeskind *et al.* thermolysed a related system **3.93** and obtained only the quinone product (Scheme **3.18**).[4f]



3.93 **3.94**: 67%

Scheme 3.18: Thermolysis by Liebeskind *et al.* of related pyridylcyclobutenone **3.93**.[4f]

Few examples exist in the literature whereby the furanone is the only product isolated from thermal ring expansion of cyclobutenones (Scheme **3.19**).[4a, 4h] The first of these was uncovered by Moore *et al*. in 1988 during a study of arylcyclobutenones.[4a] In the case of cyclobutenone **3.95**, only furanone **3.98** was formed. This is presumably because cyclisation of ketene **3.96** is hampered by the presence of two *ortho* methoxy groups, allowing the alternate pathway via ketene **3.97** to outcompete it (Scheme **3.19**).



Scheme 3.19: Reported cyclobutenone thermolyses leading to furanone products.[4a, 4h]

The second example comes from Liebeskind *et al*. where 2-furylcyclobutenone **3.101** cyclises to give furanone **3.103** (Scheme **3.19**).[4h] What is more interesting is that the tendency was reversed by protecting the hydroxyl functionality and by employing Ac_2O and NBu_3 in the thermolysis reaction. When these conditions were applied, benzofuran **3.104** was formed in 81% yield, which could be converted to furanoquinone **3.105** in 75% over 3 steps (Scheme **3.19**). This is analogous to the work by Harrowven *et al*. discussed

in Chapter 1 whereby the outcome of the rearrangement was drastically altered by capping the hydroxyl functionality.[4m]

With this in mind, the Moore rearrangement of cyclobutenone **3.75b** was investigated. Firstly, we looked to establish whether better product ratios could be obtained using an alternative solvent than dioxane (Table **3.2**). Improved ratios were observed with aromatic solvents (toluene or chlorobenzene) although furanone **3.92b** was still the preferred product in all solvents employed. This was also found to be the case for the reaction temperature over the range studied, with furanone formation outpacing quinone formation above 130 °C (Table **3.2**).

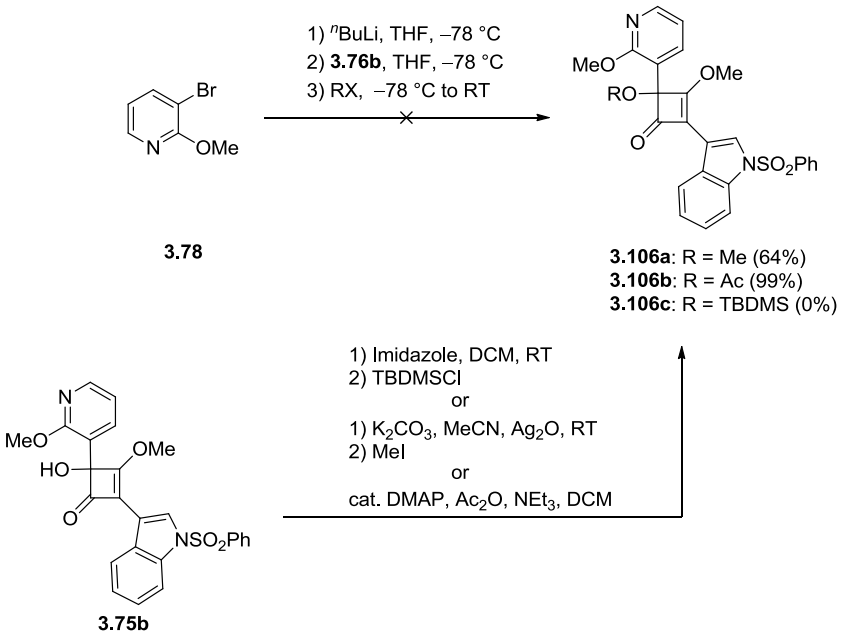
Enry	*Solvent*	*T / °C*	*t/ h*	*% Composition in crude NMR of 3.75b*	*% Composition in crude NMR 3.92b*	*% Composition in crude NMR of 3.74b (isolated yield)*
1	Dioxane	110	1	100	0	0
2	Dioxane	110	48	Composition could not be determined due to complex mixture.		
3	Dioxane	120	1	Composition could not be determined due to complex mixture.		
4	Dioxane	130	1	66	34	0
5	Dioxane	138	1	57	43	0
6	Dioxane	150	2	0	88	12
7	C_6H_5Cl	150	2	0	66	34
8	Toluene	150	3	0	69	31 (28)
9	Toluene	110	1	100	0	0
10	Toluene	120	1	Composition could not be determined due to complex mixture.		
11	Toluene	130	1	46	34	20
12	Toluene	170	0.5	0	69	31

Table 3.2: Modification of the reaction parameters for the Moore rearrangement of cyclobutenone **3.75b**.

Having exhausted all avenues with respect to the reaction parameters, focus shifted to cyclobutenone **3.75b** in the hope that conditions might be identified to alter the ratio of products in favour of the desired quinone as demonstrated by Harrowven *et al*. and Liebeskind *et al.*[4h, 4m] Taking inspiration from Liebeskind *et al*., replacing the hydrogen with

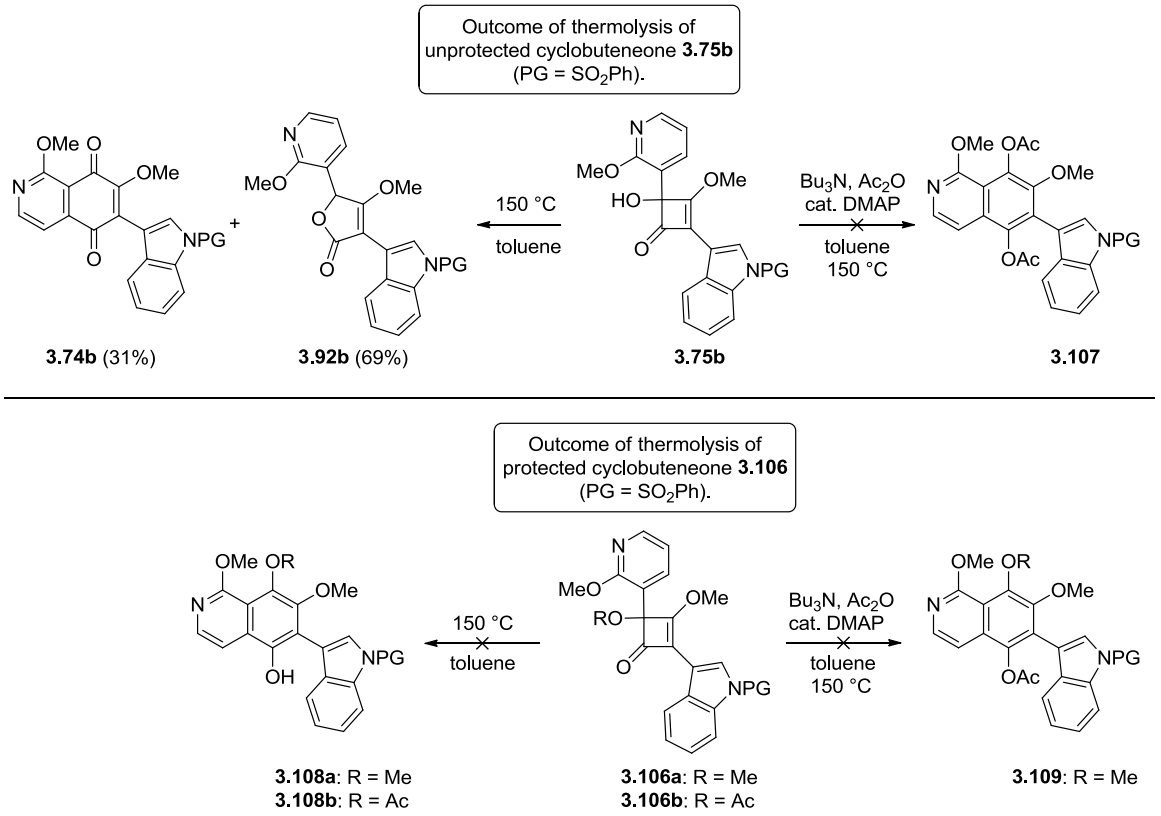
a methyl group was envisaged, along with TBDMS and an acyl group. The choice of the TBDMS group was based on the notion that steric bulk may raise the energy barrier for cyclisation leading to furanone **3.92**, allowing quinone formation to compete. As for the acyl cap, this should make the oxygen lone pair less available for cyclisation to furanone **3.92**.

Initially, attempts to form cyclobutenone **3.106** from **3.76b** directly by quenching the alkoxide formed during the addition step did not pay dividends (Scheme **3.20**). In all cases, cyclobutenone **3.75b** was isolated instead. Direct modification of cyclobutenone **3.75b** was then trialled (Scheme **3.20**). This was found to work for methyl and acyl capping (**3.106a** and **3.106b**) but the TBDMS variant **3.106c** could not be formed.



Scheme 3.20: Synthesis of modified cyclobutenone **3.106**.

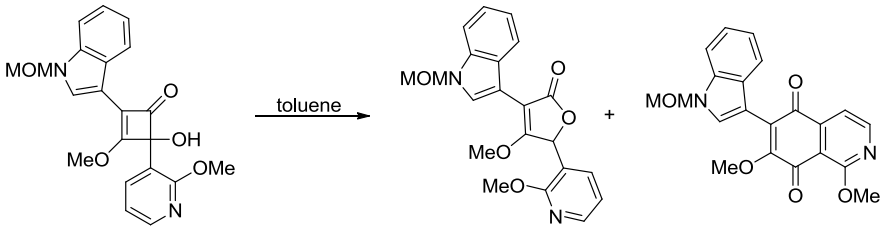
With two modified cyclobutenone **3.106** in hand, the Moore rearrangement was re-investigated (Scheme **3.21**). Initially, the conditions developed by Liebeskind *et al*. were tested on the modified cyclobutenone **3.106a**.[4h] For completeness, the same reaction was also carried out on cyclobutenone **3.75b**. In all cases, a complex product mixture was obtained with no discernible isoquinoline products **3.107** and **3.109** observed (Scheme **3.21**). Thermolysis of both the acyl and methyl capped cyclobutenones was also investigated under normal conditions (Scheme **3.21**). For the acyl derivative, this proved too labile with furanone **3.92b** the major species observed in the crude ^{1}H NMR. As for the methyl adduct, none of the desired isoquinoline **3.108a** was formed as the mixture deteriorated on heating.



Scheme 3.21: Investigation in the thermolysis of derivatives of cyclobutenones **3.75b** and **3.106**.

Once the MOM cyclobutenone was generated in sufficient quantity, it too was tested to see if it mimicked the results observed with the SO_2Ph system. Like its congeners, thermolysis at 110 ºC returned only starting material (Table **3.3**). The same was true at 130 ºC. However, at least four species were formed after heating for 1 h at 150 ºC. Further thermolysis for 2 h resulted in a simpler mixture, with quinone **3.74c** isolated in 29% (Table **3.3**). Interestingly, only traces of furanone product were formed as detected in the crude ^{1}H NMR, however this increased to approximately 50% when the reaction was thermolysed continuously for 3 h at 150 ºC in a sealed vial (Table **3.3**).

Since the isolated yield with the MOM protecting group of 29% is similar to that obtained with the SO_2Ph group (28%), the thermolysis for **3.75b** was revisited. When the Moore rearrangement was conducted in refluxing xylene, furanone formation was curbed to approximately 25% (based on crude ^{1}H NMR). Finally, TBDMS cylobutenone **3.75a** was also thermolysed but led to a very complex product mixture we presume was due to the lability of this group. As mentioned previously, the synthesis of TBDMS protected cyclobutenedione **3.76a** has proven quite erratic with yields ranging from 20 - 48%, therefore this route was abandoned.



3.75c **3.92c** **3.74c**

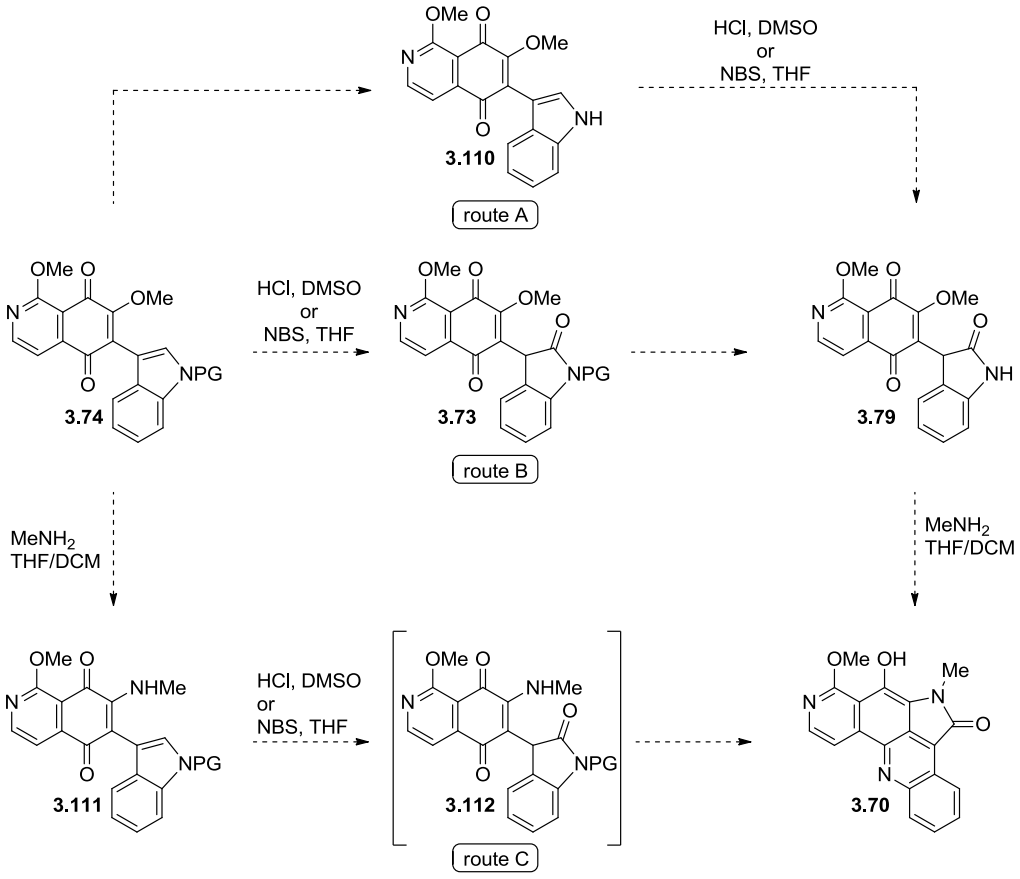
Entry	*T/ ºC*	*t / h*	*% Composition in crude NMR of 3.75c*	*% Composition in crude NMR of 3.74c* *(isolated yield)*	*% Composition in crude NMR 3.92c* *(isolated yield)*
1	110	1	100	0	0
2	130	1	100	0	0
3	150	3	0	(29%)	trace
4	150	3 continuously	0	(25%)	(29%)

Table 3.3: Thermolysis of MOM cyclobutenone **3.75c**.

With both the MOM and SO_2Ph protected quinones prepared, albeit in low yield, work moved towards establishing ring D of alpkinidine. This could be achieved by oxidation and deprotection of the indole ring to give **3.79** by way of either **3.73** or **3.110** (Scheme **3.22**, route A and B). Following this, conjugate substitution of methylamine on quinone **3.79** should initiate a cascade of reactions starting with a 5-*exo*-trig cyclisation to form ring D, followed by condensation to form ring B to **3.70** (routes A or B, Scheme **3.22**). Alternatively, the pentacyclic core **3.70** could be accessed via route C (Scheme **3.22**). Here, the sequence is reversed, with conjugate substitution occurring first to give **3.111** (Scheme **3.22**). Oxidation of **3.111** would then initiate the cascade of reactions leading ultimately to **3.70** after deprotection (Scheme **3.22**). However, the potential for unwanted side reactions was deemed greater with this pathway.

With this in mind, the oxidation of quinones **3.74b** and **3.74c** was initiated (Scheme **3.23** and **3.24** respectively). With the SO_2Ph protected quinone, oxidation to **3.73b** using HCl/DMSO resulted in the loss of the quinone bound OMe group (Scheme **3.23**).[40] As for oxidation using NBS, only starting material was recovered (Scheme **3.23**).[41] The lack of reactivity towards NBS may stem from the strong electron withdrawing nature of this protecting group. Hence, removal of the SO_2Ph group was attempted next. Deprotection of **3.74b** did not occur when treated with NaOH (Scheme **3.23**). Instead, loss of the quinone bound OMe group was observed. Although this is undesired, it does not hamper progression to the pentacyclic core of alpkinidine since it should prove just as facile to displace with $MeNH_2$ as a methoxy group. Attempts to force the reaction by heating the solution did not give any of the unprotected quinone **3.110** (Scheme **3.23**). Attention was then turned to reductive desulfonation. This was achieved using magnesium powder, with

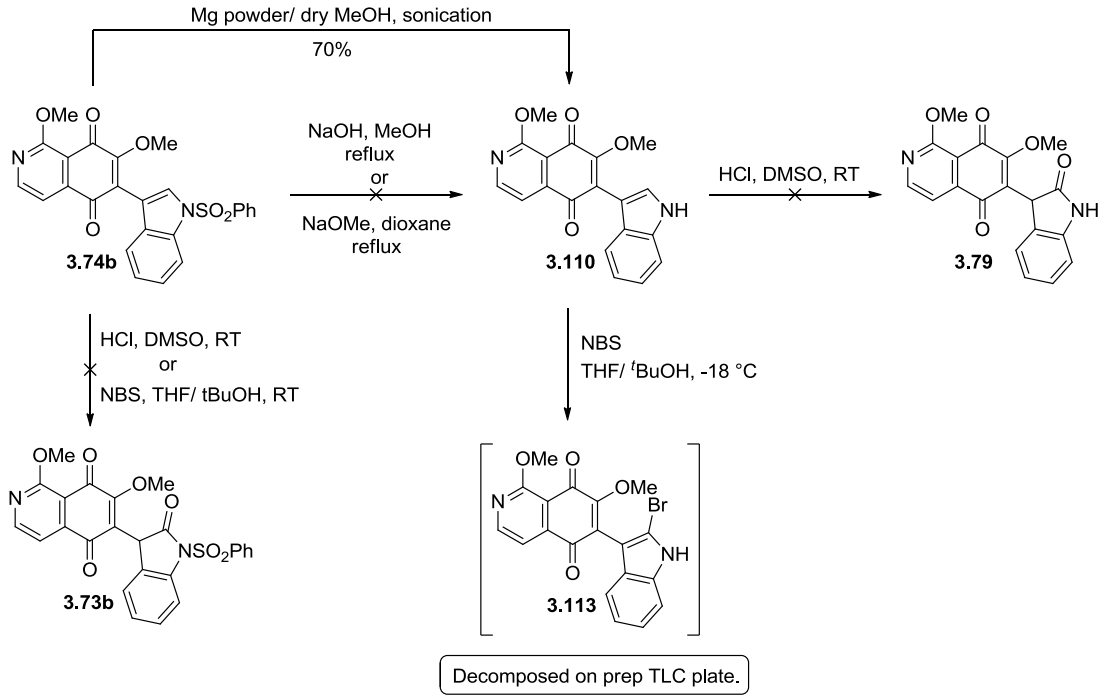
the aid of sonication, to give unprotected quinone **3.110** in 70% (Scheme **3.23**).[42] Once removed, oxidation of quinone **3.110** was explored. Using HCl/DMSO, the desired oxindole **3.79** was not formed, while the reaction of **3.110** with NBS gave the 2-bromoindole (**3.113**).[43] This proved unstable, decomposing readily during preparative TLC purification (Scheme **3.23**).



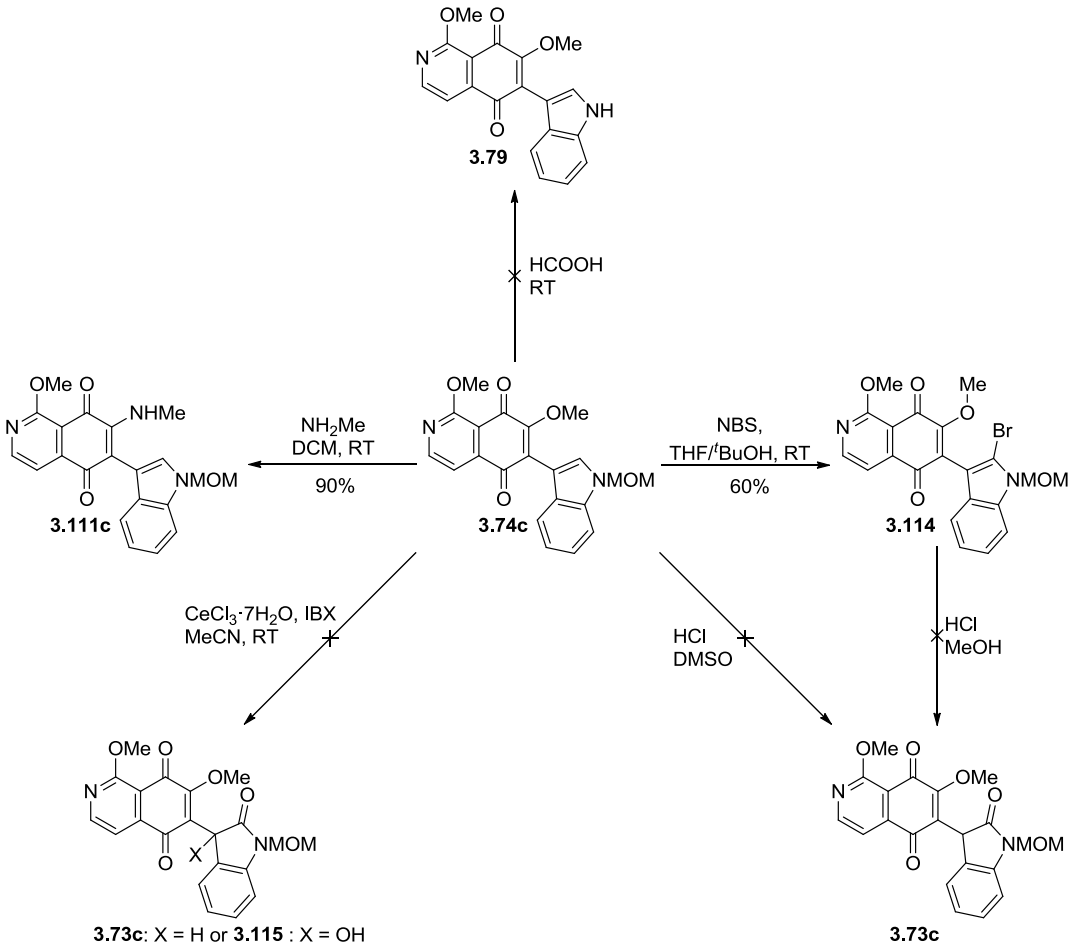
Scheme 3.22: Synthetic routes to alpkinidine precursor **3.70**.

In parallel to this work, the MOM protected quinone **3.74c** was also investigated (Scheme **3.24**). Initially, formation of oxindole **3.73c** using the conventional DMSO/HCl system and NBS was attempted. For the HCl/DMSO system, ^{1}H NMR suggested that a new species had formed after 56 equivalents of acid was used (new doublet observed adjacent to pyridine doublet for quinone **3.74c**). After 72h, only one species was believed to be present, however no product could be isolated from preparative TLC, with the product believed to have decomposed on the plate. Since the MOM group is known to be labile under strongly acidic conditions, deprotection was attempted (Scheme **3.24**). This proved more difficult than anticipated, with only partial deprotection observed by NMR analysis of the isolated product.[44]

Oxidation using NBS fared slightly better, with bromination occurring at the C2 position on the indole ring to give **3.114** (Scheme **3.24**).[43] Although not the desired product, bromoindoles are prone to form oxindoles when treated with acid.[45] Regrettably, when quinone **3.114** was treated with HCl, complete degradation of the material was observed by ^{1}H NMR (Scheme **3.24**). Neither could oxindole **3.73c** nor **3.115** be formed using $CeCl_3$/IBX oxidant system, as this resulted in complete cleavage of the quinone unit under these conditions (Scheme **3.24**).[46] With all attempts to oxidise the indole unit having met with failure, it was concluded that oxindole **3.73** was unlikely to be formed from quinone **3.74**.



Scheme 3.23: Attempts at forming ABCDE pentacycle.



Scheme 3.24: Attempts to generate pentacyclic alpkinidine core from MOM protected quinone **3.74c**.

Conclusion and future work

The pentacyclic structure found in pyrroloacridines has provided an interesting challenge for synthetic chemists, with currently no total synthesis reported for any of the six natural products known for this class of compound. With alpkinidine, work looked to take advantage of the Moore rearrangement to establish the EC ring system of this natural product. This required indolylcycbutenediones (**3.76**) to be synthesised and they were prepared in good to moderated yield depending on the protecting group. These were then converted to the corresponding cyclobutenones (**3.75**) through the addition of 3-lithio-2-methoxypyridine, again in good to moderate yield.

The ring expansion of the SO_2Ph protected cyclobutenone **3.75b** did not proceed as expected as it gave furanone **3.92b** as the major product. Although the outcome of the reaction could not be reversed, the ratio between furanone and quinone products was improved from 88 : 12 to 69 : 31 in favour of the furanone by switching solvent. With respect to MOM protected cyclobutenone **3.75c**, quinone **3.74c** was the major product

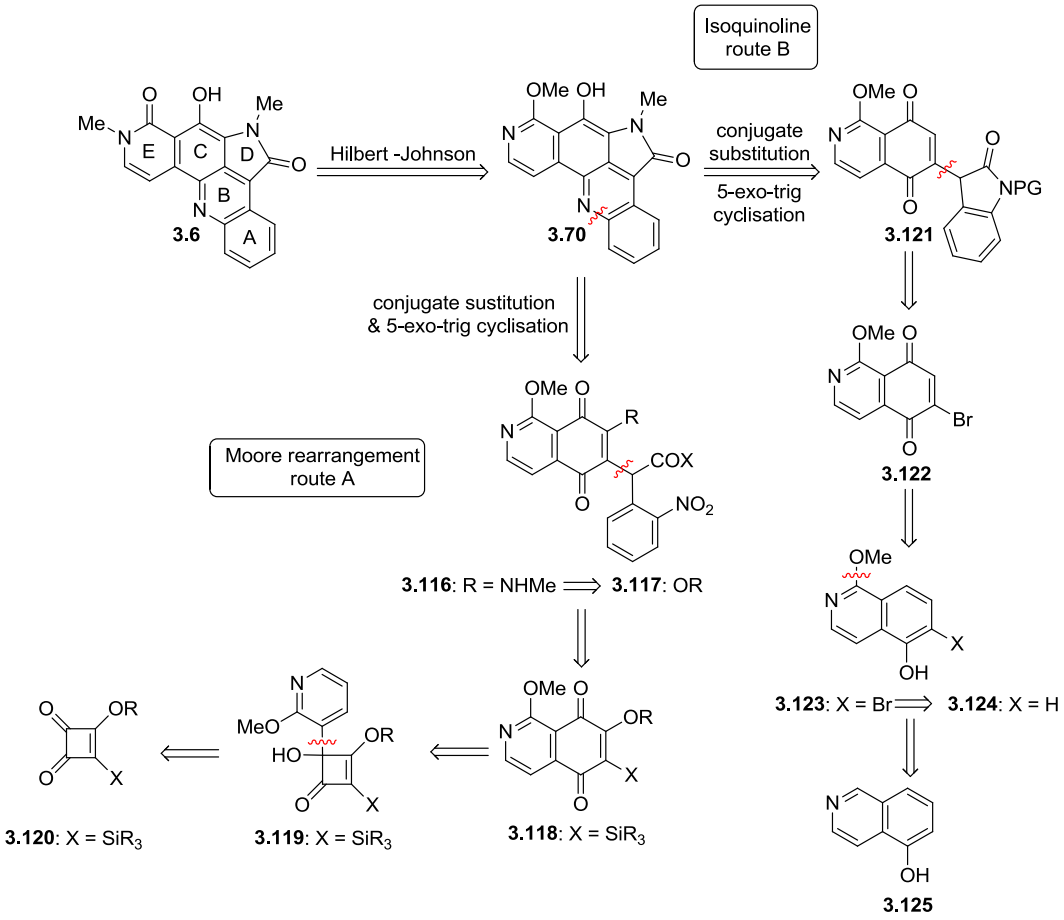
isolated from the Moore rearrangement, albeit in low yield. Furanone formation was observed when thermolysis was carried out continuously for 3 h in a sealed vial.

The main stumbling block towards the target natural product has been formation of ring D. In order to achieve this, the indole moiety must be oxidised at C2 to allow for the 5-*exo*-trig cyclisation. This transformation proved more difficult than anticipated both for the MOM and SO_2Ph protected indole **3.74**. In the case of the SO_2Ph protecting group, attempts to oxidise the indole unit were not successful whereas the MOM protected indole could be oxidised but not to the oxindole! Conversion of the resulting bromoindole **3.114** did not succeed. Nor did removal of the SO_2Ph protecting group affect the outcome, with the bromo adduct **3.113** formed decomposing readily.

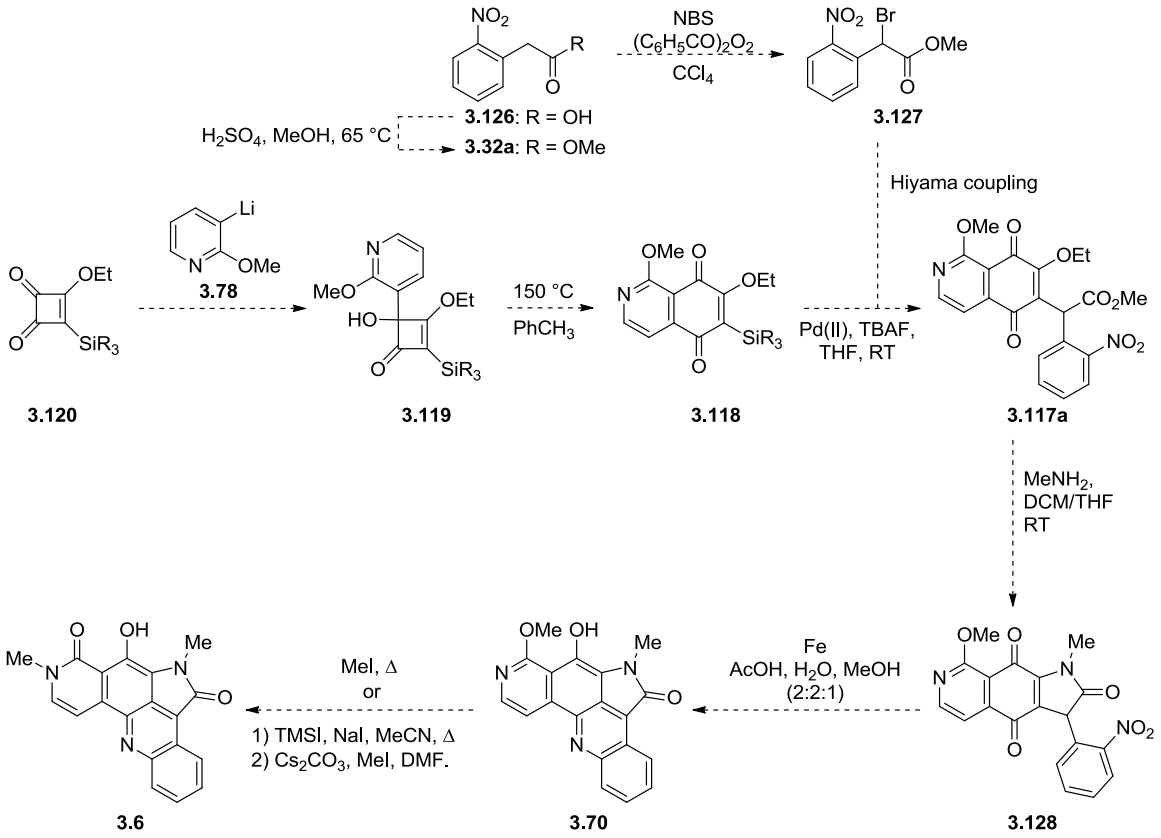
A slightly different approach is needed if alpkinidine is to be synthesised. Since ring D seems to be crucial to the formation of the pyrroloacridine core, building blocks for which no modifications are required to achieve the 5-*exo*-trig cyclisation should be exploited in the design of any future routes. Thus, looking back at the synthetic route of Piggot *et al*., either oxindole **3.35** or derivatives of the nitro ester **3.32a** seem the most convenient ways to furnish ring D (Scheme **3.5**).[30] With this in mind, one can envisage disconnections from alpkinidine along the lines shown in Scheme **3.25**.

The first of these strands, route **A** (Scheme **3.25**), looks to establish the core EC ring still via the Moore rearrangement. One difference is the use of the silylcyclobutenedione **3.120**. This should provide access to quinone **3.118** via thermolysis of cyclobutenone **3.119** (Scheme **3.26**). Once formed, quinone **3.118** may undergo a Hiyama cross-coupling reaction with nitroester **3.127**, resulting in quinone **3.117a**. From this point, the synthesis would bear resemblance to that of Piggot *et al*.[30] However, exploiting the Moore rearrangement to form the CE ring system, and affecting the insertion of the nitroester **3.127** by palladium catalysed chemistry rather than the conjugate substitution employed by Piggot *et al*. should provide enough novelty (Scheme **3.26**).

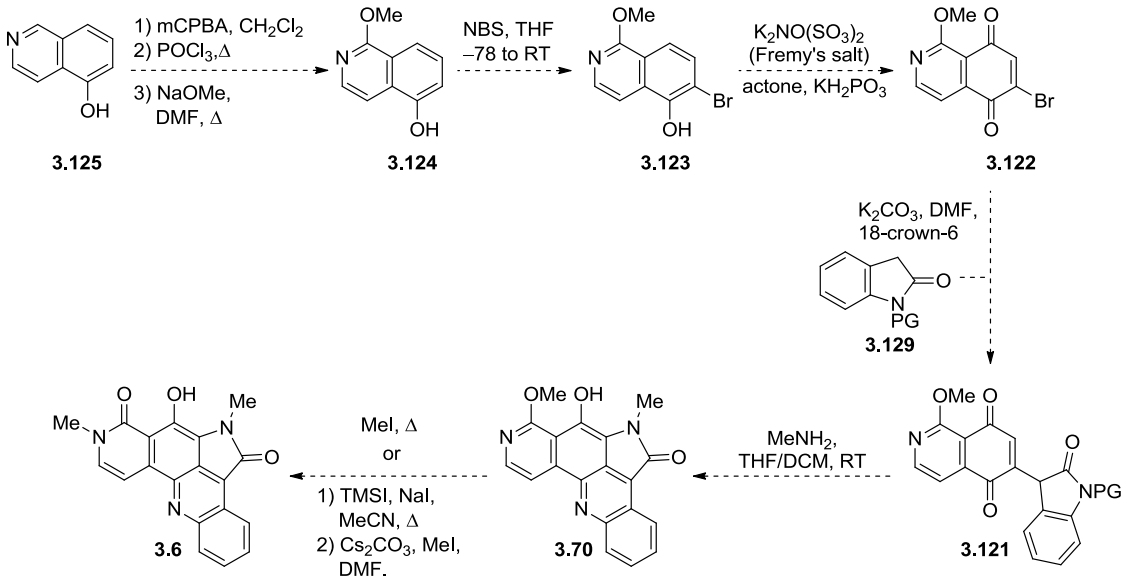
Alternatively, alpkinidine could be accessed through route **B** (Scheme **3.25**). Here, the EC ring would already be in place instead of being generated by the Moore rearrangement. Selective bromination of isoquinonline **3.124** followed by oxidation using Fremy's salt ($K_2NO(SO_3)_2$) should provide bromoquinone **3.122** (Scheme **3.27**). Double conjugate addition would give rise to alpkinidine precursor **3.70**. Finally, heating precursor **3.70** in the presence of methyl iodide should furnish the target natural product (Scheme **3.27**). The main drawback with this method is its similarity to Piggott *et al*.'s route. Nevertheless, it may prove the best means of achieving the target natural product.



Scheme 3.25: Possible new routes to alpkinidine.



Scheme 3.26: Proposed route to alpkinidne using Moore rearrangement.



Scheme 3.27: Proposed route to alpkinidine from 5-hydoxysoquinoline **3.12.**

Chapter 4: Experimental

General Information

Moisture sensitive reactions were carried out in oven-dried glassware cooled to RT under an inert atmosphere of argon. Thermolyses were carried out in stainless steel tubing (1mm internal diameter, capacity 10 mL) using a Vapourtec R4/R2+ continuous flow device unless otherwise stated.

Commercially available reagents purchased were used without further purification unless otherwise stated. Solvents were purchased from Fisher Scientific except 1,4-dioxane which was purchased from Rathburn. Dry solvents were prepared by distillation from sodium under argon (THF, toluene) or from CaH_2 under argon (DCM, MeCN) or using a Pure Solv 5-Mid solvent purification system (Innovative Technology Inc.).

TLC was performed using Merck DC-Alufolien 60 F254 0.2 mm aluminium-backed plates. Once run, plates were first visualised under a UV lamp then using a Hanessian stain (cerium molybdate). Column chromatography was conducted using silica (60 Å, 230-400 mesh size) purchased from Sigma-Aldrich or (60 Å, 400-630 μm) purchased from Merck and packed as a slurry unless otherwise stated. Preparative TLC was conducted using Merck F254 0.25 μm silica gel plates. HPLC purification was conducted on all test samples using a silica semi-preparative column connected to a refractive index detector.

Analytical data was acquired using the following equipment. Nuclear magnetic resonance spectroscopy was obtained using Bruker AV300 (300/75 MHz), AVII400, AVIIIHD400 or DRX-400 (400/100 MHz) NMR spectrometers with samples prepared in $CDCl_3$ unless stated otherwise. Chemical shift values are reported in parts per million (ppm) downfield of tetramethylsilane with residual solvent as the internal standard. Resonance signals are describe as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), dd (doublet of doublets), dt (doublet of triplets), ddd (doublet of doublets of doublets), br. (broad) and app. (apparent).

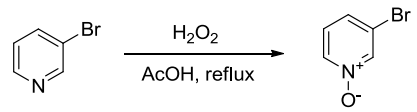
Infrared spectroscopy was performed using a Thermo Nicolet 380 FT-IR or a Perkin Elmer Spectrum One FT-IR spectrometers. Spectra were acquired neat or from an evaporated solution in $CDCl_3$ or DCM. Absorption maxima ($\nu_{max)}$ were reported as wavenumbers (cm^{-1}) and described as br-broad, s-strong, m-medium and w-weak.

Mass spectrometry analyses were performed using electron ionisation (EI) and positive mode electrospray ionisation (ESI+). EI data were generated from a Finnigan Trace 2000 series GC-MS using a Zebron ZB5-MS column, while ESI+ analyses were performed using a Waters HPLC system connected to a ZMD mass spectrometer using a C18 column. High resolution mass spectra were recorded on either a Bruker Apex TF-ICR mass spectrometer equipped with a 4.7 T actively shielded superconducting magnet and Apollo ESI ion source, or a Bruker maXis ESI-TOF (time of flight) spectrometer coupled to a Dionex Ultimate 3000 HPLC and Apollo ESI ion source, or an Agilent ESI-TOF mass spectrometer at 3500 V emitter voltage. High resolution mass spectra were recorded by Dr G. J. Langley or J. Herniman (Chemistry, Southampton) or by Jeffrey Ng (ICES, Singapore) and are reported to four decimal places.

Melting points were recorded using an Electrothermal 1A9100 series digital melting point apparatus or Büchi B-540 and are uncorrected.

Experimental Procedures

3-Bromopyridine *N*-oxide (2.64).



2.63 C_5H_4BrN (158.00)

2.64 C_5H_4BrNO (174.00)

Modified from a procedure established in a Bayer patent:[47]

To a solution of 3-bromopyridine **2.63** (2.40 mL, 25.0 mmol) in acetic acid (30 mL) was added an excess of H_2O_2 (19 mL, 164 mmol). The resultant mixture was heated at reflux for 48 h. Removal of acetic acid by azeotropic distillation with toluene, then chloroform, at reduced pressure, gave a crude mixture to which was added sat. K_2CO_3 (30 mL). The aqueous phase was separated and extracted with dichloromethane (3 x 30 mL), then the organic phases were combined, washed with sat. K_2CO_3 (30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to afford the title compound **2.64** (4.15 g, 23.9 mmol, 95%) as an orange oil.

FT-IR ν_{max} (neat, cm^{-1}) 3400 (br), 3059 (m), 3024 (m), 2739 (w), 2480 (w), 2122 (w), 1900 (w), 1592 (m), 1534 (m), 1464 (s), 1421 (s), 1295 (m), 1249 (s), 1163 (m), 1085 (m), 1007 (s), 886 (s), 798 (m), 775 (m), 665 (m), 547 (m), 490 (w), 437 (w).

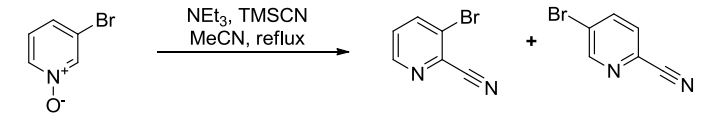
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.36 (1 H, s, Ar-**H**), 8.14 (1 H, d, *J* = 6.6 Hz, Ar-**H**), 7.40 (1 H, d, *J* = 8.4 Hz, Ar-**H**), 7.16 (1 H, dd, *J* = 8.2, 6.8 Hz, Ar-**H**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 140.9 (**C**H), 138.1 (**C**H), 128.5 (**C**H), 126.0 (**C**H), 120.5 (**C**).

LRMS (EI) $^m I_z$: 173 ($[M^{79}Br]^{+\bullet}$, 100%), 175 ($[M^{81}Br]^{+\bullet}$, 92%).

Spectroscopic data in agreement with that reported in literature.[48]

3-Bromopicolinonitrile (2.42) and 5-Bromopicolinonitrile (2.65).



2.64 C_5H_4BrNO (174.00)

2.42 $C_6H_3BrN_2$ (183.01)

2.65 $C_6H_3BrN_2$ (183.01)

Modified from a procedure established in a Bayer patent:[47]

To a solution of 3-bromopyridine-*N*-oxide **2.64** (0.85 g, 4.87 mmol) in dry acetonitrile (20 mL) were added triethylamine (0.70 mL, 5.02 mmol) and trimethylsilylcyanide (1.90 mL, 15.19 mmol) sequentially. The reaction mixture was then heated at reflux. After 16 h 6M NaOH (25 mL) was added. The aqueous phase was separated and extracted with ethyl acetate (3 x 30 mL). The organic phases were combined, then washed with 6M NaOH (2 x 25 mL) and brine (25 mL), dried with $MgSO_4$ and concentrated at reduced pressure. Purification by flash column chromatography (5-20% ethyl acetate in petroleum ether) gave firstly **2.65** (0.07 g, 0.39 mmol, 8%) then **2.42** (0.71 g, 3.88 mmol, 80%) both as colourless solids.

Data for **2.65**:

MP 127-130 °C (lit[48] 128-129 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2970 (w), 2934 (w), 2361 (w), 1796 (m), 1700 (m), 1590 (s), 1483 (m), 1426 (s), 1385 (w), 1350 (w),1307 (m), 1226 (m), 1169 (w), 1133 (w), 1077 (m), 1033 (m), 981 (m), 927 (w), 869 (w), 808 (w), 784 (w), 630 (w), 612 (w), 505 (w), 473 (w), 452 (w).

^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.78 (1H, d, *J* = 2.2 Hz, Ar-**H**), 8.00 (1H, dd, *J* = 8.2, 2.4 Hz, Ar-**H**), 7.60 (1H, d, *J* = 8.2 Hz, Ar-**H**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 152.5 (**C**H), 139.8 (**C**H), 132.0 (**C**), 129.2 (**C**H), 125.0 (**C**), 116.5 (**C**).

LRMS (EI) $^m/_z$: 182 ($[M^{79}Br]^{+\bullet}$, 93%), 184 ($[M^{81}Br]^{+\bullet}$, 89%), 103 (100%)

Spectroscopic data in agreement with that reported in literature.[49]

Data for **2.42**:

MP 97-99 °C (lit[49] 99 °C)

FT-IR ν_{max} (neat, cm^{-1}) 3748 (w), 3733 (w), 3063 (w), 2361 (s), 2340 (m), 2160 (s), 1554 (m), 1415 (s), 1249 (w), 1133 (w), 1058 (m), 1023 (s), 796 (s), 744 (m), 652 (m), 573 (w), 559 (m), 495 (m), 450 (w), 418 (w).

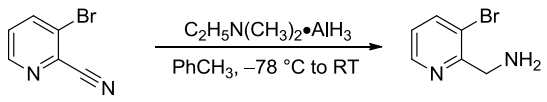
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.63 (1 H, dd, *J* = 4.6, 1.3 Hz, Ar-**H**), 8.04 (1 H, dd, *J* = 8.2, 1.3 Hz, Ar-**H**), 7.45 (1 H, dd, *J* = 8.4, 4.8 Hz, Ar-**H**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 149.0 (**C**H), 140.6 (**C**H), 134.8 (**C**), 127.7 (**C**H), 124.4 (**C**), 115.6 (**C**).

LRMS (EI) $^{m}I_z$: 182 ($[M^{79}Br]^{+\bullet}$, 84%), 184 ($[M^{81}Br]^{+\bullet}$, 79%), 75 (100%)

Spectroscopic data in agreement with that reported in literature. [50]

(3-Bromopyridin-2-yl)methanamine (2.43).



2.42
$C_6H_3BrN_2$
(183.01)

2.43
$C_6H_7BrN_2$
(187.04)

Modified from a procedure established by Harrowven et al.: [3]

To a solution of 3-bromopicolinonitrile **2.42** (1.0 g, 5.40 mmol) in dry toluene (30 mL) at –78 °C was added *N,N*-dimethylethylamine alane complex (12.0 mL, 6.0 mmol) dropwise over 15 min using a syringe. Once addition was complete, the solution was warmed to RT. After 1.75 h, the solution was cooled to –78 °C and methanol (55 mL) was added. On warming to RT, the solution was concentrated at reduced pressure then partitioned between sat. sodium potassium tartrate (30 ml) and chloroform (30 mL). The aqueous phase was separated and extracted with chloroform (2 x 30 mL). The organic phases were then combined, washed with sat. K_2CO_3 (2 x 30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to afford the title compound **2.43** (0.82 g, 4.37 mmol, 81%) as a brown oil.

FT-IR ν_{max} (neat, cm^{-1}) 3364 (w), 3282 (w), 3052 (w), 2928 (w), 2206 (w), 1671 (w), 1573 (w), 1424 (m), 1126 (w), 1067 (w), 1019 (m), 909 (m), 794 (m), 728 (s), 643 (w).

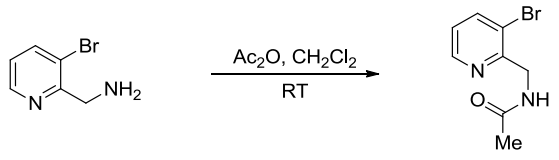
^{1}H NMR (300 MHz, $CDCl_3$) δ_H 8.48 (1H, d, *J* = 4.4 Hz, Ar-**H**), 7.79 (1H, dd, *J* = 7.9, 0.9 Hz, Ar-**H**), 7.04 (1H, app q, *J* = 7.9, 4.6 Hz, Ar-**H**), 4.04 (2H, s, C**H$_2$**), 2.07 (2H, br. s, N**H$_2$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C 159.3 (**C**), 147.5 (**C**H), 140.0 (**C**H), 123.0 (**C**H), 120.1 (**C**), 46.9 (**C**H_2).

LRMS (EI) $^m I_z$: 186 ([M^{79}Br]$^{+\bullet}$, 46%), 188 ([M^{81}Br]$^{+\bullet}$, 46%), 78 (100%).

Spectroscopic data in agreement with that reported in literature.[3]

_N_-((3-Bromopyridin-2-yl)methyl)acetamide (2.44).



2.43
$C_6H_7BrN_2$
(187.04)

2.44
$C_8H_9BrN_2O$
(229.07)

Modified from a procedure established by Harrowven et al.[3]

To a solution of methanamine **2.43** (0.53 g, 2.82 mmol) in dry dichloromethane (65 mL) was added acetic anhydride (1.35 mL, 14.28 mmol). After 16 h, 1M NaOH (40 mL) was added. The aqueous phase was separated and extracted with dichloromethane (3 x 40 mL). The organic phases were then combined, washed with sat. NaOH (40 mL) and brine (40 mL), dried over $MgSO_4$ and concentrated at reduced pressure to afford the title compound **2.44** (0.55 g, 2.40 mmol, 85%) as a light brown oil.

FT-IR ν_{max} (neat, cm^{-1}) 3294 (br), 3058 (w), 2921 (w), 2848 (w), 1654 (s), 1569 (m), 1553 (m), 1524 (m), 1423 (m), 1372 (w), 1290 (w), 1129 (w), 1067 (w), 1025 (w), 798 (w), 714 (w), 609 (w).

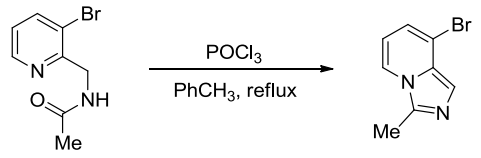
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.47 (1H, dd, *J* = 4.8, 1.1 Hz, Ar-**H**), 7.86 (1H, dd, *J* = 8.1, 1.5 Hz, Ar-**H**), 7.17 (1H, br. s., N**H**), 7.12 (1H, dd, *J* = 8.1, 4.8 Hz, Ar-**H**) 4.59 (2H, d, *J* = 4.4 Hz, C**H$_2$**), 2.11 (3H, s, C**H$_3$**).

^{13}C NMR (75 MHz ,$CDCl_3$) δ_C ppm 170.0 (**C**O), 153.6 (**C**), 146.8 (**C**H), 140.3 (**C**H), 123.5 (**C**H), 120.2 (**C**), 44.0 (**C**H_2), 23.2 (**C**H_3).

LRMS (EI) $^{m}I_{z}$: 228 ([$M^{79}Br$]$^{+\bullet}$, 100%); 230 ([$M^{81}Br$]$^{+\bullet}$, 60%).

Spectroscopic data in agreement with that reported in literature.[3]

8-Bromo-3-methylimidazo-[5,1-a]-pyridine (2.41).



2.44
$C_8H_9BrN_2O$
(229.07)

2.41
$C_8H_7BrN_2$
(211.06)

Following a procedure established by Harrowven et al.:[3]

To a solution of acetamide **2.44** (90.8 mg, 0.40 mmol) in dry toluene (20 mL) was added phosphorous oxychloride (0.15 mL, 1.60 mmol) and the mixture heated at reflux under argon for 1.75 h. After cooling to RT, sat. $NaHCO_3$ (30 mL) was added. The aqueous phase was separated and extracted with ethyl acetate (3 x 20 mL). The organic phases were combined, washed with water (2 x 20 ml) and brine (20 mL), dried over $MgSO_4$ and concentrated at reduced pressure to afford the title compound **2.41** (70.5 mg, 0.33 mmol, 84%) as a brown solid.

MP 59 – 64 °C (Lit[3] 62-64 °C)

FT-IR ν_{max} (neat, cm^{-1}) 3080 (br), 2923 (w), 1654 (m), 1629 (m), 1571 (w), 1490 (m), 1430 (m), 1358 (m), 1305 (m), 1088 (m), 1021 (m), 973 (m), 907 (m), 834 (m), 790 (m), 744 (s), 662 (m), 618 (w), 553 (m), 500 (w).

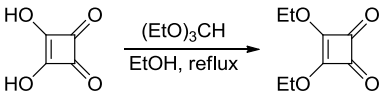
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 7.63 (1H, d, *J* = 7.0 Hz, Ar-**H**), 7.41 (1H, s, Ar-**H**), 6.86 (1H, d, *J* = 7.0 Hz, Ar-**H**), 6.42 (1H, t, *J* = 7.0 Hz, Ar-**H**), 2.63 (3H, s, C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 136.6 (**C**), 129.9 (**C**), 120.3 (**C**H), 120.2 (**C**H), 119.7 (**C**H), 112.5 (**C**H), 112.1 (**C**), 12.8 (**C**H_3)

LRMS (EI) $^{m}I_{z}$: 210 ([$M^{79}Br$]$^{+\bullet}$, 100%), 212 ([$M^{81}Br$]$^{+\bullet}$, 98%).

Spectroscopic data in agreement with that reported in literature.[3]

3,4-Diethoxy-3-cyclobutene-1,2-dione (2.36).



1.11
$C_4H_2O_4$
(114.06)

2.36
$C_8H_{10}O_4$
(170.16)

Modified from a procedure established by Moore et al.:[5]

To a solution of squaric acid **1.11** (13.74 g, 120.4 mmol) in ethanol (120 mL) was added triethyl orthoformate (50 mL, 300.6 mmol). The reaction mixture was heated at reflux (80 °C) for 7h. After 7h, the reaction mixture was concentrated using short path distillation, removing over 1/3 of the ethanol (36 mL). The reaction mixture was then left to stir overnight during which time the solution turned orange, and then concentrated at reduced pressure. Purification by flash chromatography (0 - 50% ethyl acetate in petroleum ether) afforded the title compound **2.36** (17.5 g, 103 mmol, 85%) as a clear oil.

FT-IR ν_{max} (neat, cm^{-1}) 2985 (w), 2360 (w), 1811 (m), 1729 (s), 1590 (s), 1480 (m), 1420 (s), 1381 (s), 1327 (s), 1225 (w), 1204 (w), 1153 (w), 1076 (s), 1022 (s), 984 (s), 854 (m), 799 (s), 667 (m), 595 (w), 454 (w).

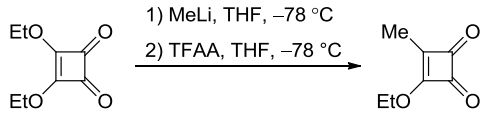
^{1}H NMR (300 MHz ,$CDCl_3$) δ_H ppm 4.74 (4H, q, *J* = 7.3 Hz, OC**H$_2$**), 1.48 (6H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz ,$CDCl_3$) δ_c ppm 189.2 (**C**O), 184.2 (**C**), 70.5 (O**C**H$_2$), 15.6 (OCH_2**C**H$_3$).

LRMS (EI) ^{m}***I*$_z$**: 170 ($[M]^{+\bullet}$, 100%).

Spectroscopic data in agreement with that reported in literature.[5]

3-Ethoxy-4-methylcyclobutene-1,2-dione (2.32).



2.36
$C_8H_{10}O_4$
(170.16)

2.32
$C_7H_8O_3$
(140.14)

Modified from a procedure established by Moore et al.:[25]

To a solution of diethyl squarate **2.36** (2.78 g, 16.3 mmol) in THF (140 mL) at –78 °C was added MeLi (2.42 M in diethyl ether, 7.00 mL, 16.9 mmol) in THF solution (30 mL) via cannula over 45 minutes. After 1 h, trifluoroacetic anhydride (2.7 mL, 19.4 mmol) was added over 1 min using a syringe. After 10 min, sat. $NaHCO_3$ (60 mL) was added and the resultant mixture warmed to RT and extracted with diethyl ether (3 x 60 mL). The organic phases were combined, washed with water (60 mL) and brine (60 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (30%-100% ethyl acetate in petroleum ether) to afford title compound **2.32** (1.95 g, 13.9 mmol, 82%) as a pale yellow oil.

FT-IR ν_{max} (neat, cm^{-1}) 2986 (w), 2361 (w), 1798 (s), 1748 (s), 1588 (s), 1474 (w), 1409 (m), 1384 (s), 1326 (s), 1162 (w), 1065 (s), 1011 (s), 977 (m), 857 (w), 840 (m), 736 (m), 664 (w), 588 (w), 475 (m).

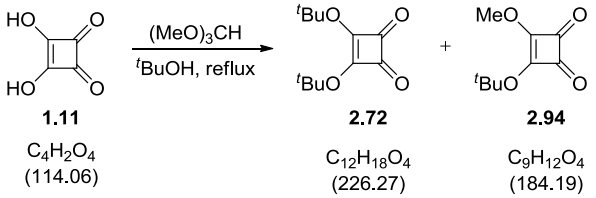
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 4.75 (2H, q, *J* = 7.1 Hz, OC**H$_2$**), 2.18 (3H, s,C**H$_3$**), 1.46 (3H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 198.7 (**C**O), 195.5 (**C**O), 193.8 (**C**), 180.2 (**C**), 70.5 (O**C**H$_2$), 15.4 (OCH_2**C**H$_3$), 9.5 (**C**H$_3$).

LRMS (EI) $^m I_z$: 140 ($[M]^{+\bullet}$, 3%), 83 (100%).

Spectroscopic data in agreement with that reported in literature.[25]

3,4-di-*tert*-Butoxy-3-cyclobutene-1,2-dione (2.72) and
3-(*tert*-Butoxy)-4-methoxycyclobut-3-ene-1,2-dione (2.94).



1.11 $C_4H_2O_4$ (114.06)

2.72 $C_{12}H_{18}O_4$ (226.27)

2.94 $C_9H_{12}O_4$ (184.19)

Modified from a procedure established by Moore et al.:[5]

To a flask charged with squaric acid **1.11** (2.53 g, 22.2 mmol) was added ^{t}BuOH (88.8 mL) and heated to reflux (85 °C). After 0.5 h, trimethyl orthoformate (35.0 mL, 330 mmol) was added over 0.5h. After addition was complete, the solution was concentrated using short path distillation, removing methanol (40 mL) and then left to reflux. After 16.5 h, the solution was concentrated at reduced pressure. Purification by flash chromatography (1 - 10% ethyl acetate in hexane) afforded first the title compound **2.72** (2.77 g, 12.4 mmol, 55%) as a colourless solid, followed by **2.94** (0.50 g, 2.71 mmol, 12%) as a colourless solid.

Data for **2.72**:

MP 99 – 101 °C (Lit[5] 103-104 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2979 (w), 2927 (w), 1804 (w), 1773 (w), 1725 (w), 1614 (w), 1574 (s), 1476 (w), 1461 (w), 1144 (s), 1085 (w), 1069 (m), 1041 (w), 1006 (w), 963 (w), 818 (w), 728 (w).

^{1}H NMR (300 MHz ,$CDCl_3$) δ_H 1.63 (18H, s, OC(C**H$_3$)$_3$**).

^{13}C NMR (75 MHz ,$CDCl_3$) δ_C 188.7 (**C**O), 186.3 (**C**), 87.0 (O**C**$(CH_3)_3$), 28.7 (OC(C**H$_3$)$_3$**).

LRMS (EI) $^m\boldsymbol{I}_z$: 226.0 ($[M]^{+\bullet}$, 1%), 41.5 (100%).

Data for **2.94**:

MP 96.5 - 96.9 °C (Lit[5] 89-90 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2957 (w), 2925 (w), 2855 (w), 1807 (w), 1721 (s), 1600 (w), 1585 (s), 1481 (w), 1457 (s), 1383 (s), 1374 (s), 1273 (w), 1249 (w), 1150 (s), 1092 (s), 1008 (m), 929 (w), 853 (w), 821 (s), 753 (s), 652 (w).

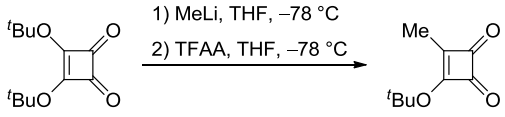
^{1}H NMR (300 MHz ,$CDCl_3$) δ_H 4.41 (3H, s, OC**H$_3$**), 1.61 (9H, s, OC(C**H$_3$)$_3$**).

^{13}C NMR (75 MHz ,$CDCl_3$) δ_C 189.5 (**C**O), 188.5 (**C**O), 185.7 (**C**), 184.8 (**C**), 87.4 (O**C**$(CH_3)_3$), 60.8 (O**C**H_3), 28.6 (OC(C**H$_3$**)$_3$).

LRMS (EI) $^m I_z$: 184([M]$^{+\bullet}$, 11%), 57 (100%).

Spectroscopic data in agreement with that reported in literature.[5]

3-*tert*-Butoxy-4-methylcyclobutene-1,2-dione (2.73).



2.72 $C_{12}H_{18}O_4$ (226.27)

2.73 $C_9H_{12}O_3$ (168.19)

Modified from a procedure established by Moore et al.:[25]

To a solution of di-*tert*-butyl squarate **2.72** (1.00 g, 4.42 mmol) in THF (40 mL) at –78 °C was added MeLi (1.84 M in diethyl ether, 2.60 mL, 4.84 mmol) in THF solution (2.5 mL) via cannula over 1 min. After 1 h, trifluoroacetic anhydride (0.74 mL, 5.30 mmol) was added over 1 min using a syringe. After 10 min, sat. $NaHCO_3$ (30 mL) was added and the resultant mixture warmed to RT. The aqueous phase was separated and further extracted with diethyl ether (3 x 30 mL). The organic phases were combined, washed with brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by flash chromatography (30%-100% ethyl acetate in cyclohexane) afforded the title compound **2.73** (0.22 g, 1.31 mmol, 30%) as a colourless solid.

MP 72-74 °C (Lit[6b] 72-73 °C)

FT-IR ν_{max} (neat, cm^{-1}) 1799 (s), 1748 (s), 1584 (s), 1393 (s), 1375 (s), 1348 (s), 1153 (s), 1059 (w), 1035 (w), 979 (w), 940 (w), 844 (w).

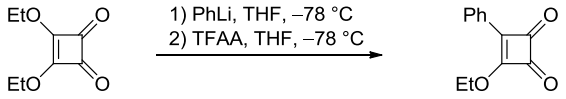
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 2.19 (3 H, s, C**H$_3$**), 1.62 (9 H, s, (C**H$_3$**)$_3$).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 199.9 (**C**O), 195.9 (**C**O), 192.8 (**C**), 182.7 (**C**), 87.5 (**C**), 28.7 (C(**C**H_3)$_3$), 9.3 (**C**H_3).

LRMS (EI) $^m I_z$: 168 ([M]$^{+\bullet}$, 8%), 57 (100%).

Spectroscopic data in agreement with that reported in literature.[25]

3-Ethoxy-4-phenylcyclobutene-1,2-dione (2.74a).



2.36
$C_8H_6O_4$
(170.16)

2.74a
$C_{12}H_{10}O_3$
(202.21)

Modified from a procedure established by Moore et al.:[25]

To a solution of diethyl squarate **2.36** (0.50g, 2.95 mmol) in THF (30 mL) at –78 °C was added a solution of PhLi (0.91 M in dibutyl ether, 3.5 mL, 3.2 mmol) in THF (10 mL) via cannula over 10 min. After 1 h, trifluoroacetic anhydride (0.5 mL, 3.5 mmol) was added over 1 min using a syringe. After 10 min, sat. $NaHCO_3$ (15 mL) was added. The resultant mixture was warmed to RT and the aqueous phase separated and extracted with diethyl ether (3 x 20 mL). The organic phases were combined, washed with sat. $NaHCO_3$ (20 mL) and brine (20 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (30-100% ethyl acetate in petroleum ether) to give **2.74a** (0.31g, 1.53 mmol, 52%) as a finely divided yellow solid.

MP 128.0 - 128.5 °C (Lit[51] 133-134 °C)

FT-IR ν_{max} (neat, cm^{-1}) 3058 (w), 2360 (w), 1778 (m), 1740 (m), 1583 (m), 1495 (w), 1469 (w), 1450 (w), 1384 (m), 1346 (m), 1319 (w), 1177 (w), 1103 (w), 1023 (m), 994 (m), 912 (w), 848 (w), 793 (w), 770 (m), 743 (m), 694 (m), 644 (w), 613 (w), 423 (w).

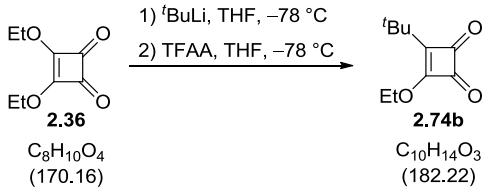
^{1}H NMR (300 MHz ,$CDCl_3$) δ_H ppm 8.03 (2H, dd, *J* = 7.9, 1.6 Hz, Ar-**H**), 7.56 - 7.44 (3H, m, Ar-**H**), 4.96 (2H, q, *J* = 7.3 Hz, OC**H$_2$**), 1.58 (3H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz ,$CDCl_3$) δ_C ppm 194.5 (**C**O), 192.7(**C**O), 192.3 (**C**), 173.7 (**C**), 132.6 (**C**H), 129.0 (**C**H), 127.7 (**C**), 127.6 (**C**H), 71.4 (O**C**H_2), 15.8 (OCH_2**C**H_3).

LRMS (EI) $^m\boldsymbol{I}_z$: 202 ($[M]^{+\bullet}$, 14%), 89 (100%).

Spectroscopic data in agreement with that reported in literature.[25]

3-(*tert*-Butyl)-4-ethoxycyclobut-3-ene-1,2-dione (2.74b).



Modified from a procedure established by Moore et al.:[25]

To a solution of ^{t}BuLi (1.81 mL, 3.63 mmol) in THF (10 mL) at –78 °C was added diethyl squarate **2.36** (0.56 g, 3.30 mmol) in THF solution (30 mL) via cannula over 35 min. After 1 h, trifluoroacetic anhydride (0.55 mL, 3.96 mmol) was added over 1 min using a syringe. After 10 min, sat. $NaHCO_3$ (30 mL) was added and the resultant mixture warmed to RT and extracted with diethyl ether (3 x 30 mL). The organic phases were combined, washed with brine (30 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (30%-100% ethyl acetate in petroleum ether) to afford title compound **2.74b** (0.24 g, 1.32 mmol, 40%) as a yellow oil.

FT-IR ν_{max} (neat, cm^{-1}) 2970 (w), 2871 (w), 2362 (w), 1791 (s), 1752 (s), 1586 (s), 1482 (w), 1407 (w), 1380 (m), 1344 (s), 1233 (w), 1045 (w), 1019 (m), 945 (w), 859 (w), 790 (m), 612 (w).

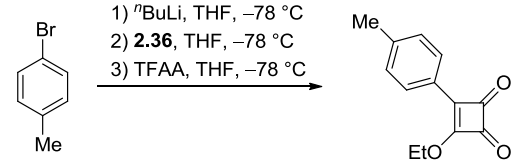
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 4.80 (2 H, q, *J*=7.2 Hz, OC**H$_2$**), 1.48 (3 H, t, *J*=7.1 Hz, OCH_2C**H$_3$**), 1.34 (9 H, s, C(C**H$_3$)$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 196.7 (**C**O), 194.8 (**C**O), 194.4 (**C**), 191.2 (**C**), 70.5 (O**C**H$_2$), 34.4 (**C**), 27.1 (C(**C**H$_3$)$_3$), 15.7 (OCH_2**C**H$_3$).

LRMS (ESI+) $^{m}/_{z}$: 183 ($[MH]^+$, 100%).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 183.1016; detected 183.1007.

3-Ethoxy-4-tolylcyclobutene-1,2-dione (2.74c).



2.95
C_7H_7Br
(171.03)

2.74c
$C_{13}H_{12}O_3$
(216.24)

Modified from a procedure established by Moore et al.:[25]

To a solution of 4-bromotoluene (0.26 g, 1.52 mmol) in THF (6 mL) at –78 °C was added ^{n}BuLi (0.78 mL, 2.33M , 1.82 mmol) over 1 min using a syringe. After 0.5 h, diethyl squarate **2.36** (0.22 g, 1.28 mmol) in THF (6 mL) was added via cannula over 30 min. After 1 h, trifluoroacetic anhydride (0.24 mL, 1.69 mmol) was added over 2 min using a syringe. After 10 min, sat. $NaHCO_3$ (15 mL) was added. The resultant mixture was warmed to RT and the aqueous phase separated and extracted with diethyl ether (3 x 20 mL). The organic phases were combined, washed with sat. $NaHCO_3$ (20 mL) and brine (20 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (30-100% ethyl acetate in petroleum ether) to give **2.74c** (0.12g, 0.56 mmol, 44%) as a yellow solid.

MP 123 - 129 °C (Lit[52] 140 - 142 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2361 (s), 2340 (m), 2159 (m), 2031 (w), 1974 (w), 1777 (s), 1736 (s), 1587 (s), 1510 (m), 1386 (m), 1350 (s), 1317 (w), 1028 (w), 997 (w), 853 (w), 830 (m), 798 (m), 594 (w), 462 (s).

^{1}H NMR (300 MHz, CDCl3) δ_H ppm 7.96 (2 H, d, *J* = 8.4 Hz, Ar-**H**), 7.32 (2 H, d, *J* = 8.1 Hz, Ar-**H**), 4.97 (2 H, q, *J* = 7.0 Hz, OC**H$_2$**), 2.44 (3 H, s, C**H$_3$**), 1.59 (4 H, t, *J*=7.1 Hz, OCH_2C**H$_3$**).

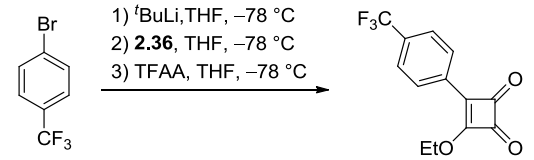
^{13}C NMR (101 MHz, CDCl3) δ_C ppm 193.9 (**C**O), 192.7(**C**O), 192.0 (**C**), 173.7 (**C**), 143.6 (**C**), 129.7 (**C**H), 127.6 (**C**H), 125.0 (**C**), 71.2 (O**C**H_2), 21.8 (**C**H_3), 15.7 (OCH_2**C**H_3).

LRMS (EI) $^{m}I_z$: 216 ($[M]^{+\bullet}$, 17%), 159 (100%).

HRMS (ESI+) $^{m}I_z$ $[MH]^+$ expected 217.0859; detected 217.0864; $^{m}I_z$ $[MNa]^+$ expected 239.0679; detected 239.0683.

Spectroscopic data in agreement with that reported in literature.[52]

3-Ethoxy-4-(4-(trifluoromethyl)phenyl)cyclobut-3-ene-1,2-dione (2.74d).



2.96

$C_7H_4BrF_3$
(225.01)

2.74d

$C_{13}H_9F_3O_3$
(270.21)

Modified from a procedure established by Moore et al.:[25]

To a solution of ^{t}BuLi (2.87 mL, 5.75 mmol) in THF (5 mL) at –78 °C was added 1-bromo-4-trifluoromethylbenzene (0.65g, 2.87 mmol) in THF (10 mL) via cannula. After 1.3 h, a solution of diethyl squarate **2.36** (0.48 g, 2.82 mmol) in THF (20 mL) was added via cannula over 12 minutes, during which time the solution turned dark red. After 1 h, trifluoroacetic anhydride (0.48 mL, 3.4 mmol) was added over 1 min using a syringe, during which time the solution became yellow. After 10 min, sat. $NaHCO_3$ (30 mL) was added. The resultant mixture was warmed to RT and the aqueous phase separated and extracted with diethyl ether (3 x 15 mL). The organic phases were combined, washed with brine (30 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-10% ethyl acetate in petroleum ether) to give **2.74d** (0.18 g, 0.65 mmol, 23%) as a fine yellow solid.

MP 116 - 120 °C

FT-IR ν_{max} (neat, cm^{-1}) 2992 (w), 2942 (w), 1786 (m), 1736 (m), 1598 (m), 1512 (w), 1409 (m), 1385 (m), 1384 (m), 1350 (m), 1317 (s), 1171 (m), 1118 (s), 1064 (s), 1018 (m), 990 (m), 899(w), 855 (m), 795 (w), 688 (w), 613 (w), 482 (w), 463(w).

^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.16 (2 H, dq, *J* = 0.8, 8.1 Hz, Ar-**H**), 7.77 (2 H, dq, *J* = 0.8, 8.1 Hz, Ar-**H**), 5.02 (2 H, q, *J* = 7.3 Hz, OC**H$_2$**), 1.62 (3 H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**).

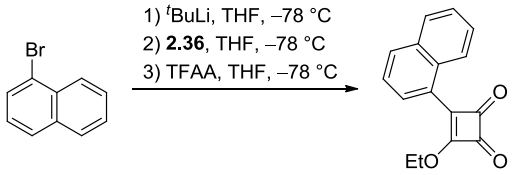
^{13}C NMR (101 MHz ,$CDCl_3$) δ_C ppm 195.4 (**C**O), 192.5 (**C**O), 192.0 (**C**), 171.7 (**C**), 133.6 (q, $^2J_{C\text{-}F}$ = 33.2 Hz, **C**), 130.6 (q, $^4J_{C\text{-}F}$ = 1.4 Hz, **C**), 127.7 (**C**H), 126.0 (q, $^3J_{C\text{-}F}$ = 4.2 Hz, **C**H), 123.5 (q, $^1J_{C\text{-}F}$ = 272.7 Hz, **C**F_3), 72.0 (O**C**H_2), 15.8 (OCH_2**C**H_3).

^{19}F NMR (282 MHz, $CDCl_3$) δ_F -63.29 (3 F, s, C**F$_3$**).

LRMS (EI) $^m\boldsymbol{I}_z$: 270 ($[M]^{+\bullet}$, 7%), 213 (100%).

HRMS (ESI +) $^{m}I_{z}$ $[MH]^+$ expected 271.0577; detected 271.0577; $^{m}I_{z}$ $[MNa]^+$ expected 293.0396; detected 293.0397.

3-Ethoxy-4-(naphthalen-1-yl)cyclobut-3-ene-1,2-dione (2.74e).



2.97
$C_{10}H_7Br$
(207.07)

2.74e
$C_{16}H_{12}O_3$
(252.26)

Modified from a procedure established by Moore et al.:[25]

To a solution of ^{t}BuLi (2.1 mL, 4.03 mmol) in THF (25 mL) at –78 °C, was added 1-bromonaphthalene (0.28 mL, 2.00 mmol) over 1 min using a syringe. No apparent colour change observed. After 0.75 h, diethyl squarate **2.36** (0.33g, 1.92 mmol) in THF (45 mL) was added via cannula over 10 min, during which time the solution became orange. After 1 h, trifluoroacetic anhydride (0.33 mL, 2.4 mmol) was added over 2 min using a syringe, during which time the solution became almost colourless. After 10 min, sat. $NaHCO_3$ (30 mL) was added. The resultant mixture was warmed to RT and the aqueous phase separated and extracted with diethyl ether (3 x 40 mL). The organic phases were combined, washed with brine (30 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-50% ethyl acetate in petroleum ether) to give **2.74e** (0.23g, 0.91 mmol, 47%) as a dark yellow (ochre) solid.

MP 113 - 117 °C

FT-IR ν_{max} (neat, cm^{-1}) 3048 (w), 2983 (w), 2933 (w), 1777 (s), 1741 (s), 1566 (s), 1510 (m), 1469 (w), 1414 (m), 1379 (m), 1335 (s), 1265 (w), 1108 (w), 1027 (w), 1008 (w), 905 (w), 857 (w), 812 (m), 796 (m), 743 (m), 774 (m), 642 (w), 588 (w).

^{1}H NMR (300 MHz ,$CDCl_3$) δ_H ppm 8.43 (1 H, d, *J* = 8.4 Hz, Ar-**H**), 8.18 (1 H, dd, *J* = 1.1, 7.3 Hz, Ar-**H**), 8.03 (1 H, d, *J* = 8.1 Hz, Ar-**H**), 7.91 (1 H, dd, J = 1.5, 7.7 Hz, Ar-**H**), 7.56 - 7.69 (3 H, m, Ar-**H**), 5.06 (2 H, q, *J*=7.0 Hz, OC**H$_2$**), 1.62 (3 H, t, *J*=7.1 Hz OCH_2C**H$_3$**).

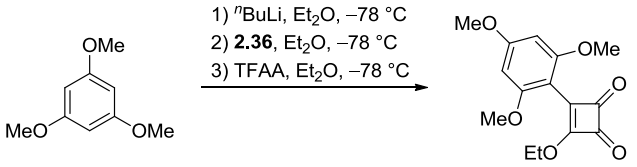
^{13}C NMR (75 MHz ,$CDCl_3$) δ_C ppm 194.4 (**C**O), 192.7 (**C**O), 192.4 (**C**), 176.4 (**C**), 133.9 (**C**), 133.3 (**C**H), 130.24 (**C**), 128.7 (**C**H), 128.20 (**C**H), 127.5 (**C**H), 126.7 (**C**H), 126.5 (**C**H), 125.6 (**C**), 125.2 (**C**H), 71.6 (O**C**H_2), 15.9 (OCH_2**C**H_3).

LRMS (EI) $^{m}I_z$: 252 ([M]$^{+\bullet}$, 13%), 139 (100%).

HRMS (ESI+) $^{m}I_z$ [MH]$^+$ expected 253.0859; detected 253.0854; $^{m}I_z$ [MNa]$^+$ expected 275.0679; detected 275.0676.

Spectroscopic data in agreement with that reported in the literature.[4b]

3-Ethoxy-4-(2,4,6-trimethoxyphenyl)cyclobut-3-ene-1,2-dione (2.74f).



2.98
$C_9H_{12}O_3$
(168.19)

2.74f
$C_{15}H_{16}O_6$
(292.28)

Modified from a procedure established by Moore et al.:[25]

To a solution of 1,3,5-trimethoxybenzene (0.44g, 2.61 mmol) in diethyl ether (26 mL) at –78 °C was added a solution of ^{n}BuLi (1.2 mL, 2.87 mmol), and the solution warmed to RT. After 4.5 h, the solution was transferred via cannula to a solution of diethyl squarate **2.36** (0.38 g, 2.22 mmol) in diethyl ether (35 mL) at –78 °C via cannula. After 2h, trifluoroacetic anhydride (0.36mL, 2.62 mmol) was added over 1 min using a syringe. After 10 min, the resultant mixture was warmed to RT and sat. $NaHCO_3$ (15 mL) was added. The aqueous phase was separated and extracted with diethyl ether (3 x 20 mL). The organic phases were combined, washed with brine (20 mL), dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (5-40% ethyl acetate in petroleum ether) to give **2.74f** (67.5 mg, 0.26 mmol, 10%) as a yellow solid.

MP 92 - 94 °C

FT-IR ν_{max} (neat, cm^{-1}) 2942 (w), 2844 (w), 2360 (w), 1783 (m), 1735 (m), 1604 (s), 1571 (s), 1494 (w), 1454 (m), 1400 (m), 1330 (s), 1230 (m), 1205 (s), 1133 (s), 1089 (m), 1025 (s), 948 (m), 921 (m), 814 (m), 797 (s), 675 (m), 629 (m), 583 (m), 558 (m), 467 (m).

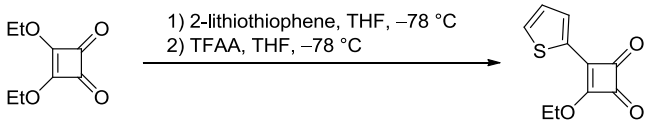
^{1}H NMR (400 MHz ,$CDCl_3$) δ_H ppm 6.13 (2 H, s, Ar-**H**), 4.82 (2 H, q, *J* = 7.1 Hz, OC**H$_2$**), 3.85 (3 H, s, OC**H$_3$**), 3.83 (6 H, s, OC**H$_3$**), 1.47 (3 H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**)

^{13}C NMR (101 MHz ,$CDCl_3$) δ_C ppm 194.8 (**C**O), 193.9 (**C**O), 192.6 (**C**), 174.7 (**C**), 164.9 (**C**), 159.0 (**C**), 99.2 (**C**), 90.6 (**C**H), 70.0 (O**C**H_2), 55.9 (O**C**H_3), 55.5 (O**C**H_3), 15.7 (OCH_2**C**H_3).

LRMS (EI) $^{m}I_z$: 292 ($[M]^{+\bullet}$, 40%), 207 (100%).

HRMS (ESI+) $^{m}I_z$ $[MH]^+$ expected 293.1020; detected 293.1019; $^{m}I_z$ $[MNa]^+$ expected 315.0839; detected 315.0838.

3-Ethoxy-4-(2-thiophenyl)cyclobutene-1,2-dione (2.74g).



2.36
$C_8H_6O_4$
(170.16)

2.74g
$C_{10}H_8O_3S$
(208.23)

Modified from a procedures established by Moore et al.:[25]

To a solution of commercially available 2-lithiothiophene (0.34 ml, 3.16 mmol) in THF (35 mL) at –78 °C was added a solution of diethyl squarate **2.36** (0.49 g, 2.88 mmol) in THF (35 mL) via cannula over 40 min. After 1 h, trifluoroacetic anhydride (0.48 mL, 3.46 mmol) was added dropwise over 1 min using a syringe, during which time the solution became bright yellow. After 10 min, sat. $NaHCO_3$ (30 mL) was added and the resultant mixture was warmed to RT. The aqueous phase was separated and extracted with diethyl ether (3 x 20 mL). The organic phases were then combined, washed with brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to an ochre solid. Purification by flash chromatography (5 - 10% ethyl acetate in cyclohexane) gave **2.74g** (0.31g, 1.49 mmol, 55%) as a dark yellow solid.

MP 140 – 142 ºC

FT-IR ν_{max} (neat, cm^{-1}) 3093 (w), 2991 (w), 2923 (w), 2851 (w), 1778 (m), 1738 (s), 1586 (s), 1501 (m), 1467 (m), 1414 (s), 1386 (s), 1350 (s), 1216 (m), 1186 (m), 1103 (m), 1027 (s), 995 (m), 852 (m),

824 (m), 794 (m), 736 (s), 650 (m), 628 (m), 595 (w), 552 (w), 450 (m), 416 (w).

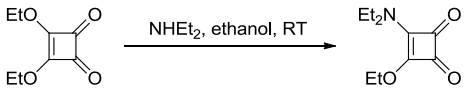
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 7.88 (1H, dd, *J* = 3.8, 0.9 Hz, Ar-**H**), 7.79 (1H, dd, *J* = 5.1, 0.7 Hz, Ar-**H**), 7.26 (1H, dd, *J* = 4.9, 3.8 Hz, Ar-**H**), 4.94 (2 H, q, *J* = 7.3 Hz, OC**H$_2$**), 1.57 (3 H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 191.0 (**C**O), 190.5 (**C**O), 190.2 (**C**), 168.2 (**C**), 133.9 (**C**H), 131.3(**C**H), 128.8(**C**H), 127.4(**C**), 71.4 (O**C**H_2), 15.8 (OCH_2**C**H_3).

LRMS (EI) $^{m}I_z$: 208 ($[M]^{+\bullet}$, 56%), 123 (100%).

HRMS (ESI+) $^{m}I_z$ $[MH]^+$ expected 209.0267; detected 209.0266.

3-Diethylamino-4-ethoxycyclobutene-1,2-dione (2.74h).



2.36 $C_8H_6O_4$ (170.16)

2.74h $C_{10}H_{15}NO_3$ (197.23)

Modified from a procedure established by Ivanovsky et al.:[53]

To a solution of 3,4-diethoxycyclobutene-1,2-dione **2.36** (0.98 g, 5.74 mmol) in ethanol (15 mL) was added *N,N*-diethylamine (0.58 mL, 5.61 mmol). After 21 h at RT, the solution was concentrated at reduced pressure to a colourless solid then dissolved in diethyl ether (30 mL), washed with sat. K_2CO_3 (2 x 30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by flash chromatography (50-100% ethyl acetate in petroleum ether) afforded the title compound **2.74h** (0.99 g, 5.02 mmol, 89%) as a pale yellow solid.

MP 45 - 47 °C (Lit[54] 40-41°C)

FT-IR ν_{max} (neat, cm^{-1}) 2970 (w), 2934 (w), 2361 (w), 1796 (m), 1700 (s), 1590 (s), 1483 (s), 1426 (s), 1385 (s), 1351 (m), 1307 (s), 1226 (s), 1169 (w), 1133 (w), 1077 (s), 1034 (s), 981 (s), 927 (w), 869 (m), 808 (m), 784 (m), 631 (w), 612 (w), 505 (w), 473 (w), 452 (w).

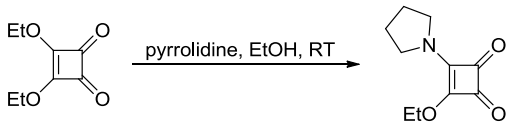
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 4.55 (2H, q, *J* = 7.2 Hz, OC**H$_2$**), 3.53 (2H, q, *J* = 7.3 Hz, NC**H$_2$**), 3.28 (2H, q, *J* = 7.0 Hz, NC**H$_2$**), 1.25 (3H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**), 1.05 (6H, t, *J* = 7.1 Hz, NCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 188.4 (**C**O), 181.6 (**C**O), 175.7 (**C**), 170.9 (**C**), 68.8 (OCH_2**C**H_3), 43.6 (N**C**H_2), 43.2 (N**C**H_2), 15.3 (OCH_2C**H$_3$**), 14.1 (NCH_2C**H$_3$**), 13.7 (NCH_2C**H$_3$**).

LRMS (ESI+) $^{m}I_z$: 261 ([M+Na+MeCN]$^+$, 100%).

Spectroscopic data in agreement with that reported in literature.[54]

3-Ethoxy-4-pyrrolidinocyclobutene-1,2-dione (2.74i).



2.36
$C_8H_6O_4$
(170.16)

2.74i
$C_{10}H_{13}NO_3$
(195.22)

Following a modified procedure established by Ivanovsky et al..[53]

To a solution of cyclobutenedione **2.36** (1.04g, 6.13 mmol) in ethanol (15 mL) was added pyrrolidine (0.62 mL, 7.40 mmol). After 25 h at RT, the solution was concentrated at reduced pressure, then partitioned between diethyl ether (30 mL) and sat. $NaHCO_3$ (30 mL). The aqueous phase was separated and extracted with diethyl ether (2 x 30 mL). The organic phases were combined, washed with sat. $NaHCO_3$ (30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to an orange solid. Recrystallisation from propan-2-ol afforded the title compound **2.74i** (0.30 g, 1.54 mmol, 25%) as an orange solid.

MP 87-89 °C (Lit[53, 55] 175-178 °C and 90.5 °C).

FT-IR ν_{max} (neat, cm^{-1}) 3083 (m), 1590 (m), 1532 (w), 1462 (s), 1418 (s), 1287 (m), 1250 (s), 1155 (m), 1083 (m), 1007 (s), 888 (s), 849 (m), 780(s), 746 (s), 663 (s), 546 (m), 489 (m), 437 (w).

^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 4.47 (2 H, q, *J* = 7.2 Hz, OC**H$_2$**), 3.55 (2 H, m, NC**H$_2$**), 3.40 (2 H, m, NC**H$_2$**),

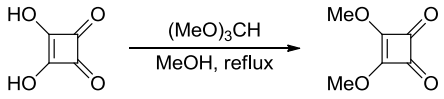
1.75 (4 H, m, $NCH_2C\mathbf{H_2}$),
1.20 (3 H, t, *J* = 7.0 Hz, $OCH_2C\mathbf{H_3}$).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 188.3 (**C**O), 181.6 (**C**O), 176.1 (**C**), 169.5 (**C**), 68.4 ($O\mathbf{C}H_2$), 47.9 ($N\mathbf{C}H_2$), 47.8 ($N\mathbf{C}H_2$), 24.4 ($NCH_2\mathbf{C}H_2$), 24.1 ($NCH_2\mathbf{C}H_2$), 15.1 ($OCH_2\mathbf{C}H_3$).

LRMS (EI) $^m\!/_z$: 195 ($[M]^{+\bullet}$, 87%), 82 (100%).

Spectroscopic data in agreement with that reported in literature.[53, 55]

3,4-Dimethoxy-3-cyclobutene-1,2-dione (2.75).



1.11 $C_4H_2O_4$ (114.06) **2.75** $C_6H_6O_4$ (142.11)

Following a modified procedure established by Moore et al.:[5]

To a solution of squaric acid **1.11** (8.54 g, 74.9 mmol) in methanol (85.0 mL) was added trimethyl orthoformate (17.5 mL, 160.0 mmol). The reaction mixture was heated at reflux (65 °C) for 16.5 h, during which time the solution became translucent, and then concentrated at reduced pressure. Purification by flash chromatography (25 - 50% ethyl acetate in petroleum ether) afforded the title compound **2.75** (10.3 g, 72.6 mmol, 97%) as an off-white solid.

MP 43-46 °C (Lit[5] 55-57 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2964 (w), 1813 (m), 1733 (s), 1604 (s), 1590 (s), 1486 (s), 1418 (s), 1368 (s), 1085 (m), 1039 (s), 926 (m), 831 (s), 747 (m), 644 (s), 630 (s).

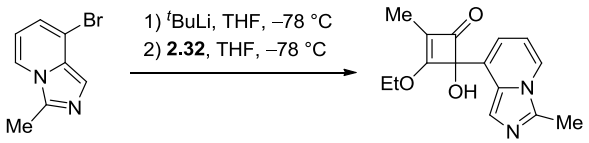
^{1}H NMR (400 MHz, $CDCl_3$) δ_H 4.38 (6 H, s, $OC\mathbf{H_3}$).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C 189.1 (**C**O), 184.4 (**C**), 60.9 ($O\mathbf{C}H_3$),

LRMS (ESI+) $^m\!/_z$:143 ($[MH]^+$, 5%), 506 (100%).

Spectroscopic data in agreement with that reported in literature.[5]

3-Ethoxy-4-hydroxy-2-methyl-4-(3-methylimidazo[1,5-a]pyridin-8-yl)cyclobut-2-enone (2.40).



2.41
$C_8H_7BrN_2$
(211.06)

2.40
$C_{15}H_{16}N_2O_3$
(272.30)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.49 M in pentane, 0.70 mL, 1.04 mmol) in THF (2 mL) at –78 °C was added imidazopyridine **2.41** (111 mg, 0.53 mmol) in THF (1 mL) dropwise over 1 min using a syringe, during which time the solution became brown. After 30 min, cyclobutenedione **2.32** (86.3 mg, 0.62 mmol) in THF (2 ml) was added dropwise via cannula over 1 min. After 1 h, sat. $NaHCO_3$ (10 mL) was added, the solution was warmed to RT and dichloromethane (15 mL) added. The aqueous phase was separated and extracted with dichloromethane (2 x 15 mL). The organic phases were then combined, washed with brine (2 x 15 mL), dried over $MgSO_4$ and concentrated under reduced pressure to a dark yellow oil. Purification by flash column chromatography (0-10% methanol in dichloromethane with 1% aq ammonia solution) gave **2.40** (73.1 mg, 27 mmol, 51%) as an orange oil.

FTIR ν_{max} (neat, cm^{-1}) 3311 (br. w), 2925 (m), 2854 (w), 1759 (w), 1618 (s), 1385 (s), 1325 (s), 1276 (w), 1258 (w), 1178 (m), 1144 (m), 1065 (m), 1014 (m), 938 (w), 888 (w), 764 (s), 750 (s), 663 (w).

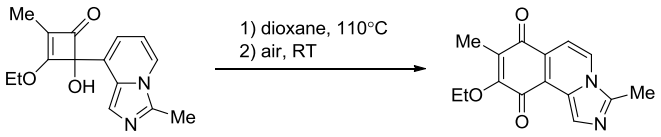
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 7.58 (1H, d, *J* = 7.1 Hz, Ar-**H**), 7.32 (1H, s, Ar-**H**), 6.97 (1H, d, *J* = 6.6 Hz, Ar-**H**), 6.55 (1H, app. t, *J* = 6.8 Hz, Ar-**H**), 4.44 (1H, dq, *J* = 10.1, 7.2 Hz, OC**H**H), 4.21 (1H, dq, *J* = 10.1, 7.2 Hz OCH**H**), 2.59 (3H, s, C**H$_3$**), 1.79 (3H, s, C**H$_3$**), 1.31 (3H, t, *J* = 7.1 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 190.6 (**C**O), 183.0 (**C**), 135.3 (**C**), 128.3 (**C**), 127.7 (**C**), 124.8 (**C**), 120.4 (**C**H), 118.4 (**C**H), 116.4 (**C**H), 112.0 (**C**H), 91.5 (**C**), 69.0 (O**C**H_2), 15.0 (**C**H_3), 12.4 (**C**H_3), 6.7 (OCH_2**C**H_3).

LRMS (ESI+) ^{m}***I*$_z$**: 273 ($[MH]^+$, 100%).

Spectroscopic data in agreement with that reported in literature.[3]

Cribrostatin 6 (2.6).



2.40
$C_{15}H_{16}N_2O_3$
(272.30)

2.6
$C_{15}H_{14}N_2O_3$
(270.28)

Following a procedure established by Harrowven et al.:[3]

Cyclobutenone **2.40** (48.7 mg, 0.18 mmol) in dioxane (2 mL) was heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resultant solution was then stirred in air for 30 min then concentrated at reduced pressure to afford a crude brown oily solid. Purification by flash chromatography (0-10% methanol in dichloromethane with 1% aq ammonia) gave the title compound **2.6** (14.8 mg, 0.05 mmol, 30%) as a blue solid.

FT-IR ν_{max} (neat, cm^{-1}) 2923 (w), 1661 (m), 1627 (m), 1526 (m), 1486 (w), 1443 (w), 1388(w), 1377 (w), 1316 (m), 1291 (m), 1171 (m), 1094 (w), 973 (w), 949 (w), 892 (w), 821 (w), 791 (w), 736 (w), 657 (w).

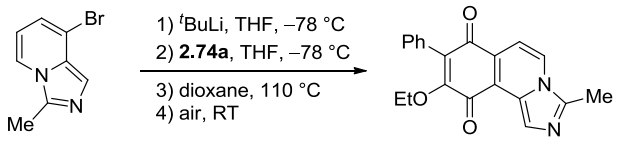
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.27 (1H, s, Ar-**H**), 7.88 (1H, d, *J* = 7.3 Hz, Ar-**H**), 7.22 (1H, d, *J* = 7.3 Hz, Ar-**H**), 4.41 (2H, q, *J* = 7.0 Hz, OC**H$_2$**), 2.71 (3H, s, C**H$_3$**), 2.08 (3H, s, C**H$_3$**), 1.43 (3H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 185.0 (**C**O), 180.7 (**C**O), 156.2 (**C**), 137.7 (**C**), 130.1 (**C**), 125.9 (**C**H), 125.0 (**C**), 124.7 (**C**H), 123.9 (**C**), 123.6 (**C**), 107.7 (**C**H), 69.7 (O**C**H_2), 16.0 (**C**H_3), 12.6 (**C**H_3), 9.2 (**C**H_3).

LRMS (EI) $^m\boldsymbol{I}_z$: 270 ($[M]^{+\bullet}$, 24%), 207 (100%).

Spectroscopic data in agreement with that reported in literature.[2]

9-Ethoxy-8-phenyl-3-methylimidazo[5,1-a]isoquinoline-7,10-dione (2.77a).



2.41
$C_8H_7BrN_2$
(211.06)

2.77a
$C_{20}H_{16}N_2O_3$
(332.35)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.55 M in dibutyl ether, 0.62 mL, 0.96 mmol) in THF (2 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (96.4 mg, 0.46 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.74a** (99.5 mg, 0.49 mmol) in THF (2 mL) was added via cannula dropwise over 1 min followed, after 1 h, by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) was added. The aqueous phase was separated and extracted with dichloromethane (2 × 15 mL) then the organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. The resulting brown oil (128 mg, 0.38 mmol) was dissolved in dioxane (7.6 mL) then heated at 110 °C under flow in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was stirred under air for 30 min, concentrated at reduced pressure and then purified by column chromatography (0-100% ethyl acetate in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77a** (28.0 mg, 0.08 mmol, 17%) as a burgundy red solid.

MP decomposes above 130 °C

FT-IR ν_{max} (neat, cm^{-1}) 2983 (w), 2922 (w), 2851(w), 2361 (w), 2333 (w), 1658 (s), 1615 (s), 1566 (m), 1493 (w), 1440 (m), 1369 (m), 1336 (w), 1273 (m), 1259 (m), 1242 (m), 1204 (m), 1171 (s), 1104 (w), 1057 (s), 1016 (s), 936 (w), 906 (w), 879 (w), 863 (w), 783 (s), 745 (m), 696 (w), 647 (w), 610 (w), 507 (m).

^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.34 (1 H, s, Ar**H**), 7.92 (1 H, d, *J* = 7.3 Hz, Ar**H**), 7.46–7.35 (5 H, m, 5 × Ar**H**), 7.26 (1 H, d, *J* = 7.3 Hz, Ar**H**), 4.15 (2 H, q, *J* = 7.2 Hz, OC**H$_2$**), 2.73 (3 H, s, C**H$_3$**) 1.25 (3 H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

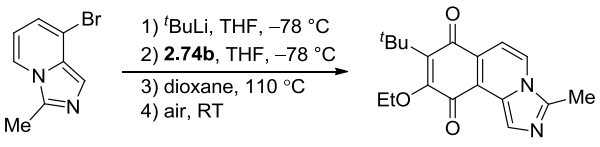
^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 184.0 (**C**O), 181.2 (**C**O), 155.5 (**C**), 137.9 (**C**H), 130.5 (**C**H), 130.4 (**C**), 130.2 (**C**), 128.6 (**C**H),

127.8 (**C**H), 126.2 (**C**), 125.1 (**C**H), 125.0 (**C**), 123.8 (**C**), 123.6 (**C**), 107.9 (**C**H), 70.2 (**C**H_2), 15.6 (**C**H_3), 12.7 (**C**H_3).

LRMS (EI) $^{m}/_{z}$**:** 332 ([M]$^{+\bullet}$, 43%), 334 (100%).

HRMS (ESI+) $^{m}/_{z}$ [MH]$^{+}$ expected 333.1234; detected 333.1238.

8-(*tert*-Butyl)-9-ethoxy-3-methylimidazo[5,1-a]isoquinoline-7,10-dione (2.77b).



2.41
$C_8H_7BrN_2$
(211.06)

2.77b
$C_{18}H_{20}N_2O_3$
(312.36)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.91 M in dibutyl ether, 0.55 mL, 1.05 mmol) in THF (1 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (106 mg, 0.50 mmol) in THF (2 mL) dropwise via cannula over 5 min. After 30 min, cyclobutenedione **2.74b** (88.5 mg, 0.49 mmol) in THF (3 mL) was added dropwise via cannula over 5 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) was added then the aqueous phase was separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave an orange/brown solid (97.9 mg, 0.31 mmol). A sample of this material (77.6 mg) was then dissolved in dioxane (6.6 mL) and heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was stirred in air for 30 min then concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77b** (22.3 mg, 0.07 mmol, 19%) as a burgundy red solid.

MP 128–130 °C

FT-IR ν_{max} (neat, cm^{-1}) 2957 (m), 2925 (m), 1665 (s), 1642 (s), 1620 (s), 1578 (w), 1538 (s), 1484 (w), 1458 (m), 1362 (m), 1324 (m), 1284 (s), 1224 (w), 1176 (s), 1104 (w), 1068 (s), 1036 (w),

1013 (w), 974 (w), 917 (w), 877 (m), 835 (m), 816 (m), 775 (w), 745 (m), 664 (m), 503 (w), 420 (w).

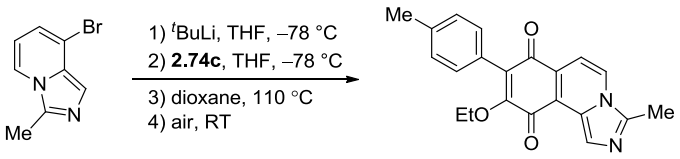
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.23 (1 H, s, Ar**H**), 7.88 (1 H, dd, *J* = 7.4, 0.7 Hz, Ar**H**), 7.17 (1 H, d, *J* = 7.3 Hz, Ar**H**), 4.24 (2 H, q, *J* = 7.1 Hz, OC**H$_2$**), 2.71 (3H, s, C**H$_3$**), 1.48 (3 H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**), 1.46 (9 H, s, $C(C$**H$_3$**$)_3$).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 185.9 (**C**O), 181.3 (**C**O), 156.7 (**C**), 140.5 (**C**), 137.6 (**C**), 126.9 (**C**), 125.6 (**C**H), 125.0 (**C**H), 123.4 (**C**), 122.7 (**C**), 108.0 (**C**H), 70.1 (**C**H_2), 36.3 (**C**), 31.2 ($C($**C**$H_3)_3$), 15.6 (**C**H_3), 12.7 (**C**H_3).

LRMS (ESI+) **$^{m}I_z$**: 313 ($[MH]^+$,100%).

HRMS (ESI+) **$^{m}I_z$** $[MH]^+$ expected 313.1547, detected 313.1543.

9-Ethoxy-3-methyl-8-(*p*-tolyl)imidazo[5,1-a]isoquinoline-7,10-dione (2.77c)



2.41
$C_8H_7BrN_2$
(211.06)

2.77c
$C_{21}H_{18}N_2O_3$
(346.38)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.92 M in dibutyl ether, 0.54 mL, 1.04 mmol) in THF (2 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (104 mg, 0.49 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.74c** (102 mg, 0.47 mmol) in THF (3 mL) was added dropwise via cannula over 4 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) was added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave a brown oil (101 mg, 0.29 mmol). The material was dissolved in dioxane (3.5 mL) then heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was then stirred in air for 30 min,

concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77c (**32.9 mg, 0.09 mmol, 20%) as a burgundy red solid.

MP 162–164 °C

FT-IR ν_{max} (neat, cm^{-1}) 2924 (w), 1664 (s), 1642 (m), 1616 (s), 1532 (w), 1512 (m), 1488 (w), 1458 (m), 1366 (w), 1329 (m), 1300 (s), 1176 (s), 1061 (s), 995 (w), 974 (w), 910 (w), 854 (w), 810 (m), 773 (w), 743 (m), 664 (w), 517 (w), 469 (w), 431 (w).

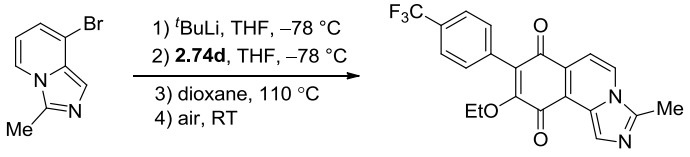
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.34 (1 H, s, Ar**H**), 7.92 (1 H, d, *J* = 7.3 Hz, Ar**H**), 7.31–7.23 (5 H, m, Ar**H**), 4.14 (2 H, q, *J* = 7.0 Hz, OC**H$_2$**), 2.74 (3 H, s, C**H$_3$**), 2.42 (3 H, s, C**H$_3$**), 1.26 (3 H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

^{13}C NMR (101 MHz , $CDCl_3$) δ_C ppm 184.2 (**C**O), 181.2 (**C**O), 155.4 (**C**), 138.6 (**C**), 137.8 (**C**), 130.5 (**C**H), 130.3 (**C**), 128.6 (**C**H), 127.4 (**C**), 126.2 (**C**H), 125.0 (**C**H), 125.0 (**C**), 123.8 (**C**), 123.6 (**C**), 107.9 (**C**H), 70.0 (O**C**H_2), 21.40 (**C**H_3), 15.6 (**C**H_3), 12.7 (**C**H_3).

LRMS (ESI+) $^{m}/_{z}$: 347 ($[MH]^+$, 100%).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 347.1390, detected 347.1398.

9-Ethoxy-3-methyl-8-(4-(trifluoromethyl)phenyl)imidazo[5,1-a]isoquinoline-7,10-dione (2.77d).



2.41
$C_8H_7BrN_2$
(211.06)

2.77d
$C_{21}H_{15}F_3N_2O_3$
(400.35)

Following a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.92 M in dibutyl ether, 0.52 mL, 1.00 mmol) in THF (1 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (100 mg, 0.48 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.74d** (121 mg, 0.45 mmol) in THF (3 mL) was added dropwise via cannula over 1 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and

dichloromethane (15 mL) was added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave a brown oil (67.7 mg, 0.17 mmol). A sample of this material (56 mg) was then dissolved in dioxane (3.5 mL) and heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was then stirred under air for 30 min, concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77d** (4.6 mg, 0.01 mmol, 4 %) as a burgundy red solid.

MP 144–149 °C

FT-IR ν_{max} (neat, cm^{-1}) 2926 (w), 2363 (w), 1667 (m), 1643 (m), 1617 (m), 1533 (w), 1489 (w), 1460 (w), 1408 (w), 1375 (w), 1325 (s), 1168 (s), 1123 (s), 1060 (s), 1020 (w), 994 (w), 911 (w), 858 (w), 827 (w), 789 (w), 760 (w), 744 (w), 664 (w), 599 (w), 387 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.34 (1 H, s, Ar**H**), 7.95 (1 H, d, *J* = 7.3 Hz, Ar**H**), 7.71 (2 H, d, *J* = 8.2 Hz, Ar**H**), 7.50 (2 H, d, *J* = 8.1 Hz, Ar**H**), 7.26 (1 H, d, *J* = 7.1 Hz, Ar-**H**), 4.29 (2 H, q, *J* = 7.1 Hz, OC**H$_2$**), 2.74 (3 H, s, C**H$_3$**), 1.27 (3 H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

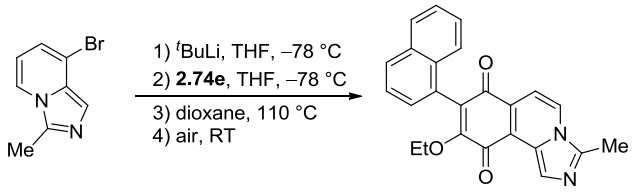
^{13}C NMR (101 MHz , $CDCl_3$) δ_C ppm 183.5 (**C**O), 180.9 (**C**O), 155.8 (**C**), 138.1 (**C**), 131.0 (**C**H), 126.4 (**C**H), 125.3 (**C**H), 124.9 (**C**), 124.7 (**C**H, q, J_{CF} = 3.9 Hz), 123.7 (**C**), 123.7 (**C**), 107.8 (**C**H), 70.5 (O**C**H_2), 15.7 (**C**H_3), 12.7 (**C**H_3); *Four quaternary carbons not observed.*

^{19}FNMR (376 MHz, $CDCl_3$) δ_F ppm –62.8 (CF_3)

LRMS (ESI+) $^{m}/_{z}$: 401 ($[MH]^+$, 100%).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 401.1108, detected 401.1115.

9-Ethoxy-3-methyl-8-(naphthalen-1-yl)imidazo[5,1-a]isoquinoline-7,10-dione (2.77e).



2.41
$C_8H_7BrN_2$
(211.06)

2.77e
$C_{24}H_{18}N_2O_3$
(382.41)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.92 M in dibutyl ether, 0.56 mL, 1.07 mmol) in THF (1 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (108 mg, 0.51 mmol) in THF (2 mL), dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.74e** (123 mg, 0.49 mmol) in THF (3mL) was added dropwise via cannula over 2 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) was added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) afforded an oil (112 mg, 0.29 mmol). Part of this material (73.2 mg, 0.19 mmol) was dissolved in dioxane (4.6 mL) then heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was then stirred under air for 30 min, concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77e** (34.1 mg, 0.09 mmol, 28%) as a dark grey solid.

MP decomposes to tar at 93–96 °C.

FT-IR ν_{max} (neat, cm^{-1}) 3049 (w), 2981 (w), 2926 (w), 1667 (m), 1640(m), 1618 (s), 1531 (m), 1509 (w), 1489 (w), 1459 (m), 1365 (w), 1298 (s), 1259 (w), 1233 (w), 1177 (s), 1076 (m), 1060 (m), 1029 (w), 1010 (w), 993 (w), 974 (w), 910 (m), 870 (w), 833 (w), 800 (m), 779 (m), 732 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.39 (1 H, d, *J* = 0.6 Hz, Ar**H**), 7.96–7.89 (3 H, m, Ar**H**), 7.66 (1 H, d, *J* = 8.1 Hz, Ar-**H**), 7.55 (1 H, dd, *J* = 8.1, 7.2 Hz, Ar**H**), 7.53–7.44 (2 H, m, Ar**H**), 7.39 (1 H, dd, *J* = 7.0, 1.0 Hz, Ar**H**), 7.25 (1 H, d, *J* = 7.3 Hz),

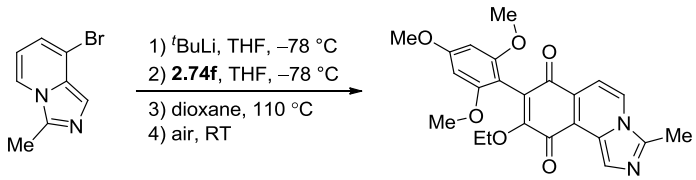
3.97 (1 H, dq, *J* = 9.9, 7.2 Hz, OC**H**H),
3.87 (1 H, dq, *J* = 9.9, 7.0 Hz, OCH**H**), 2.75 (3 H, s, C**H$_3$**),
1.05 (3 H, t, *J* = 7.0 Hz, OCH$_2$C**H$_3$**).

^{13}C NMR (101 MHz , $CDCl_3$) δ_C ppm 184.0 (**C**O), 180.8 (**C**O), 156.5 (**C**), 137.9 (**C**), 133.4 (**C**), 132.0 (**C**), 129.2 (**C**H), 129.0 (**C**), 128.5 (**C**H), 128.3 (**C**H), 127.4 (**C**), 126.5 (**C**H), 126.2 (**C**H), 126.0 (**C**H), 125.4 (**C**H), 125.2 (**C**H), 125.2 (**C**), 125.0 (**C**H), 123.8 (**C**), 123.7 (**C**), 107.9 (**C**H), 69.5 (O**C**H$_2$), 15.5 (**C**H$_3$), 12.8 (**C**H$_3$).

LRMS (EI) $^{m}I_z$: 383 ([MH]$^+$, 100%).

HRMS (ESI+) $^{m}I_z$ [MH]$^+$ expected 383.1390, detected 383.1387.

9-Ethoxy-3-methyl-8-(2,4,6-trimethoxyphenyl)imidazo[5,1-a]isoquinoline-7,10-dione (2.77f).



2.41
$C_8H_7BrN_2$
(211.06)

2.77f
$C_{23}H_{22}N_2O_6$
(422.43)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.92 M in dibutyl ether, 0.53 mL, 1.02 mmol) in THF (2 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (134 mg, 0.46 mmol) in THF (2 mL) dropwise via cannula over 3 min. After 45 min, cyclobutenedione **2.74f** (130 mg, 0.45 mmol) in THF (5 mL) was added dropwise via cannula over 5 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave a brown oil (154 mg, 0.36 mmol). A sample of this material (67.4 mg) was then dissolved in dioxane (3.7 mL) and heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was then stirred under air for 30 min, concentrated at reduced pressure and purified by column

chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77f** (26.0 mg, 0.06 mmol, 32%) as a dark grey solid.

MP 250–253 °C

FT-IR ν_{max} (neat, cm^{-1}) 2923 (m), 2850 (w), 1665 (m), 1610 (s), 1589 (s), 1533 (w), 1499 (w), 1460 (m), 1415 (w), 1367 (w), 1300 (s), 1228 (m), 1205 (m), 1178 (m), 1156 (s), 1127 (s), 1065 (m), 1035 (w), 995 (w), 947 (w), 920 (w), 812 (w), 730 (w).

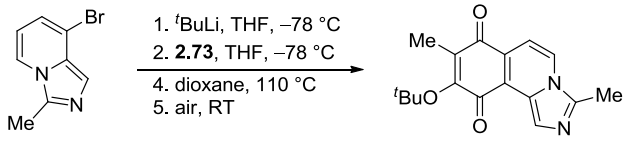
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.34 (1 H, s, Ar**H**), 7.90 (1 H, dd, *J* = 7.5, 0.7 Hz, Ar**H**), 7.27 (1 H, d, *J* = 7.5 Hz, Ar**H**), 6.20 (2 H, s, Ar**H**), 4.07 (2 H, q, *J* = 7.1 Hz, OC**H$_2$**), 3.87 (3 H, s, OC**H$_3$**), 3.75 (6 H, s, 2 × OC**H$_3$**), 2.74 (3H, s, C**H$_3$**), 1.23 (3 H, t, *J* = 7.0 Hz, C**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 183.4 (**C**O), 181.0 (**C**O), 162.2 (**C**), 158.8 (**C**), 156.8 (**C**), 137.5 (**C**), 125.7 (**C**H), 124.7 (**C**H), 124.1 (**C**), 123.7 (**C**), 122.8 (**C**), 108.3 (**C**H), 101.9 (**C**), 90.6 (**C**H), 68.5 (O**C**H_2), 55.8 (**C**H_3), 55.4 (**C**H_3), 15.6 (**C**H_3), 12.7 (**C**H_3); *one quaternary carbon not observed*.

LRMS (ESI+) $^m\!I_z$: 423 ($[MH]^+$,100%).

HRMS (ESI+) $^m\!I_z$ $[MH]^+$ expected 423.1551, detected 423.1553.

9-(*tert*-Butoxy)-3,8-dimethylimidazo[5,1-a]isoquinoline-7,10-dione (2.77i).



2.41
$C_8H_7BrN_2$
(211.06)

2.77i
$C_{17}H_{18}N_2O_3$
(298.34)

Modified from a procedure established by Harrowven et al.[3]

To a solution of ^{t}BuLi (1.55 M in dibutyl ether, 0.56 mL, 1.07 mmol) in THF (1 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (107 mg, 0.51 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.73** (83.1 mg, 0.49 mmol) in THF (3 mL) was added dropwise via cannula over 5 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and

dichloromethane (15 mL) was added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave a brown oil (24.7 mg, 0.08 mmol) which was dissolved in dioxane (1.6 mL) then heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was stirred under air for 30 min then concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77i** (11.6 mg, 0.04 mmol, 8%) as a dark blue solid.

MP 187–190 °C

FT-IR ν_{max} (neat, cm^{-1}) 2977 (w), 2927 (w), 1668 (m), 1641 (s), 1621 (s), 1603 (m), 1530 (m), 1487 (w), 1461 (m), 1392 (w), 1367 (s), 1332 (m), 1304 (s), 1289 (s), 1260 9w), 1241 (w), 1178 (w), 1143 (s), 1090 (s), 1025 (w), 986 (m), 974 (w), 949 (w), 915 (w), 863 (s), 833 (w), 820 (m), 755 (w), 732 (m), 714 (m).

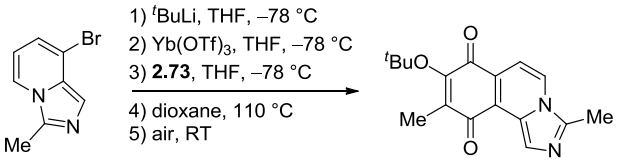
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.31 (1 H, d, *J* = 0.7 Hz, Ar-**H**), 7.88 (1 H, d, *J* = 7.3, 0.6 Hz, Ar-**H**), 7.23 (1 H, d, *J* = 7.3 Hz, Ar-**H**), 2.71 (3 H, s, C**H$_3$**), 2.11 (3 H, s, C**H$_3$**),1.47 (9 H, s, C(C**H$_3$**)$_3$).

^{13}C NMR (101 MHz , $CDCl_3$) δ_C ppm 185.4 (**C**O), 182.2 (**C**O), 155.6 (**C**), 137.7 (**C**), 135.1 (**C**), 126.3 (**C**H), 125.0 (**C**), 124.6 (**C**H), 124.2 (**C**), 123.9 (**C**), 107.7 (**C**H), 85.1 (**C**), 29.6 (C(**C**H_3)$_3$), 12.7 (**C**H_3), 10.9 (**C**H_3).

LRMS (ESI+) $^{m}/_{z}$: 299 ($[MH]^+$, 100%).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 299.1390, detected 299.1389.

8-(*tert*-Butoxy)-3,9-dimethylimidazo[5,1-a]isoquinoline-7,10-dione (2.77j)



2.41
$C_8H_7BrN_2$
(211.06)

2.77h
$C_{17}H_{18}N_2O_3$
(298.34)

Modified from a procedure established by Harrowven et al.[6a]

To a solution of ^{t}BuLi (1.55 M in dibutyl ether, 0.58 mL, 1.11 mmol) in THF (1 mL) at –78 °C was added 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (111 mg, 0.53 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, the solution was then added via cannula to a solution of ytterbium triflate (299 mg, 0.48 mmol) in THF (5 mL) at –78 °C over 4 min. After 1 h, cyclobutenedione **2.73** (77.9 mg, 0.46 mmol) in THF (3 mL) was added dropwise via cannula over 1 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) was added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with brine (2 × 15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) gave a brown oil (36.7 mg, 0.12 mmol). Part of this material (21.3 mg, 0.07 mmol) was dissolved in dioxane (1.7 mL) and heated at 110 °C in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was then stirred under air for 30 min, concentrated at reduced pressure and purified by column chromatography (0-10% MeOH in dichloromethane with 1% aq. ammonia solution) to give the title compound **2.77j** (12.5 mg, 0.04 mmol, 16%) as a dark blue solid.

MP 161–164 °C

FT-IR ν_{max} (neat, cm^{-1}) 2977 (w), 2927 (w), 1658 (s), 1619 (m), 1603 (s), 1530 (m), 1489 (w), 1458 (w), 1369 (s), 1326 (m), 1287 (s), 1260 (w), 1216 (m), 1149 (s), 1072 (m), 1013 (m), 974 (w), 948 (w), 840 (m), 793 (m), 727 (m), 677 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.34 (1 H, d, *J* = 0.9 Hz, Ar-**H**), 7.88 (1 H, dd, *J* = 7.3, 0.6 Hz, Ar-**H**), 7.20 (1 H, d, *J* = 7.3 Hz, Ar-**H**), 2.72 (3 H, s, C**H$_3$**), 2.12 (3 H, s, C**H$_3$**),1.46 (9 H, s, C(C**H$_3$**)$_3$).

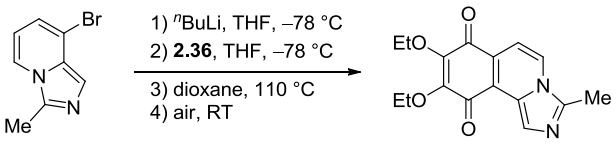
^{13}C NMR (101 MHz , $CDCl_3$) δ_C ppm 186.0 (**C**O), 181.7 (**C**O), 155.0 (**C**), 137.9 (**C**), 136.2 (**C**), 126.8 (**C**H), 124.8 (**C**), 124.3 (**C**H), 124.1 (**C**),

123.8 (**C**), 107.6 (**C**H), 85.0 (**C**), 29.6 (C(**C**H_3)$_3$), 12.7 (**C**H_3), 10.8 (**C**H_3).

LRMS (ESI+) $^{m}I_{z}$: 299 ($[MH]^+$, 100%).

HRMS (ESI+) $^{m}I_{z}$ $[MH]^+$ expected 299.1390, detected 299.1392.

8,9-Diethoxy-3-methylimidazo[5,1-a]isoquinoline-7,10-dione (2.77k)



2.41
$C_8H_7BrN_2$
(211.06)

2.77k
$C_{16}H_{16}N_2O_3$
(300.31)

Modified from a procedure established by Harrowven et al.[3]

To a solution of 8-bromo-3-methylimidazo[5,1-a]pyridine **2.41** (505 mg, 2.56 mmol) in THF (10 mL) cooled to –78 °C was added ^{n}BuLi (2.40 M in hexanes, 1.30 mL, 3.12 mmol) dropwise using a syringe over 1 min. After 45 min, cyclobutenedione **2.36** (0.42 g, 2.47 mmol) in THF (10 mL) was added dropwise via cannula over 4 min, followed after 1 h by sat. $NaHCO_3$ (10 mL). The resulting solution was warmed to RT and dichloromethane (15 mL) added. The aqueous phase was then separated and extracted with dichloromethane (2 × 15 mL). The organic phases were combined, washed with sat. $NaHCO_3$ (15 mL) and brine (15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) resulted in an oil (76.5 mg) which was dissolved in dioxane (5 mL) then heated at 110 °C under flow in stainless-steel tubing for a residence time of 1 h using a Vapourtec R4/R2+ device. The resulting solution was stirred under air at RT for 30 min then concentrated at reduced pressure. Purification by column chromatography (0-10% methanol in dichloromethane with 1% aq. ammonia) to give the title compound **2.77k** (18.4 mg, 0.06 mmol, 2 %) as a burgundy red solid.

MP 97–99 °C (Lit[22] 125-127 °C)

FT-IR ν_{max} (neat, cm^{-1}) 2985 (w), 2929 (w), 1667 (s), 1650 (s), 1620 (s), 1603 (s), 1528 (m), 1485 (w), 1457 (w), 1387 (w), 1334 (w), 1313 (w), 1271 (s), 1208 (m), 1174 (s), 1092 (w), 1049 (s), 1009 (w), 880 (w), 858 (w), 820 (w), 792 (w), 777 (w), 747 (m).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.32 (1 H, d, *J* = 0.7 Hz, Ar**H**), 7.87 (1 H, dd, *J* = 7.3, 0.9 Hz, Ar**H**), 7.19 (1 H d, *J* = 7.3 Hz, Ar**H**), 4.38 (3H, q, OC**H**$_2$), 4.37 (3 H, q, OC**H**$_2$), 2.71 (3 H, s, C**H**$_3$), 1.45 (3 H, t, *J* = 7.1 Hz, $OCH_2C\mathbf{H}_3$), 1.44 (3 H, t, *J* = 7.1 Hz, $OCH_2C\mathbf{H}_3$).

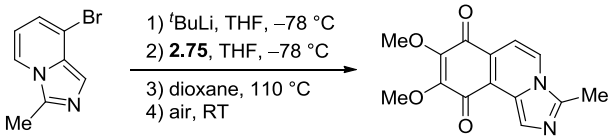
^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 181.7 (**C**O), 181.3 (**C**O), 145.9 (**C**), 145.5 (**C**), 137.9 (**C**), 126.7 (**C**H), 124.5 (**C**H), 124.0 (**C**), 123.5 (**C**), 123.3 (**C**), 107.4 (**C**H), 69.9 (O**C**H$_2$), 69.9 (O**C**H$_2$), 15.6 (2 x $OCH_2\mathbf{C}H_3$), 12.7 (**C**H$_3$).

LRMS (ESI) $^{m}/_{z}$: 301 ($[MH]^+$, 100%).

HRMS (ESI) $^{m}/_{z}$ $[MH]^+$ expected 301.1183; detected 301.1178.

Spectroscopic data in agreement with that reported in literature.[22]

8,9-Dimethoxy-3-methylimidazo[5,1-a]isoquinoline-7,10-dione (2.77l).



2.41 $C_8H_7BrN_2$ (211.06)

2.77l $C_{14}H_{12}N_2O_4$ (272.26)

Modified from a procedure established by Harrowven et al.:[3]

To a solution of ^{t}BuLi (1.55 M in pentane, 0.67 mL, 1.04 mmol) in THF (2 mL) at –78 °C was added imidazopyridine **2.41** (109.7 mg, 0.52 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.75** (70.1 mg, 0.49 mmol) in THF (2 mL) was added via cannula over 3 min. After 1 h, sat. $NaHCO_3$ (10 mL) was added and the solution was warmed to RT and dichloromethane (20 mL) added. The aqueous phase was then separated and extracted with dichloromethane (2 x 20 mL). The organic phases were combined, washed with sat. $NaHCO_3$ (30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to a red/brown oil. The crude material was dissolved in dioxane (11 mL) then heated at 110°C for 1 h. The resulting solution was then stirred in air for 30 min and concentrated at reduced pressure. Purification by flash chromatography (0-10% methanol in dichloromethane with 1% aq ammonia) gave the title compound **2.77l** (19.0 mg, 0.07 mmol, 14%) as a burgundy red solid.

MP 179 – 181 ºC

FT-IR ν_{max} (neat, cm^{-1}) 3081 (w), 2954 (w), 2852 (w), 1669 (m), 1648 (s), 1620 (s), 1603 (s), 1528 (m), 1442 (m), 1333 (m), 1311 (m), 1277 (s), 1204 (s), 1165 (s), 1088 (m), 1048 (s), 954 (m), 925 (s), 876 (m), 796 (m), 785 (m), 748 (s), 663 (w), 627 (w), 575 (w), 444 (w).

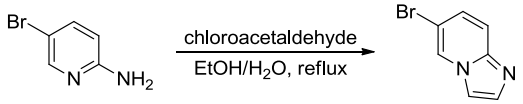
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.31 (1 H, s, Ar-**H**), 7.87 (1 H, d, *J* = 7.3 Hz, Ar-**H**), 7.19 (1 H d, *J* = 7.3 Hz, Ar-**H**), 4.11 (3H, s, OC**H**$_3$), 4.10 (3 H, s, OC**H**$_3$), 2.71 (3 H, s, C**H**$_3$).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 181.4 (**C**O), 180.9 (**C**O), 138.0 (**C**), 126.8 (**C**H), 124.6 (**C**H), 123.4 (**C**), 123.1 (**C**), 107.3 (**C**H), 61.5 (O**C**H$_3$), 61.4 (O**C**H$_3$), 12.7 (**C**H$_3$), 3 quaternary carbons not observed.

LRMS (EI) $^{m}/_{z}$: 272 ([M]$^{+\bullet}$, 100%).

HRMS (ESI+) $^{m}/_{z}$ [MH]$^{+}$ expected 273.0870; detected 273.0869.

6-Bromoimidazo[1,2-a]pyridine (2.80).



2.78
$C_5H_5BrN_2$
(173.01)

2.80
$C_7H_5BrN_2$
(197.03)

Following a modified procedure established in patent.[56]

To a solution of **2.78** (2.25 g, 13.0 mmol) in ethanol (35 mL) was added an aqueous solution of chloroacetaldehyde (2.80 mL, 44.1 mmoL) and the reaction set to reflux (85 °C). After 3 h, ethanol was removed by distillation at reduced pressure, then sat. $NaHCO_3$ (30 mL) and ethyl acetate (30 mL) were added. The aqueous phase was separated and extracted using ethyl acetate (2 x 30 mL). The organic phases were then combined, washed with sat. $NaHCO_3$ (30 mL) and brine (30 mL), dried over $MgSO_4$ and concentrated at reduced pressure to afford a brown solid. Purification by flash chromatography (0 - 10% methanol in dichloromethane) afforded **2.80** (1.59 g, 8.07 mmol, 62%) as a light brown solid.

FT-IR ν_{max} (neat, cm^{-1}) 3137 (w), 3090 (w), 1722 (w), 1502 (m), 1470 (w), 1422 (w), 1336 (w), 1304 (m), 1260 (w), 1221 (w), 1139 (w),

1122 (w), 1053 (w), 918 (w), 900 (w), 861 (w), 837 (w), 793 (m), 722 (m), 682 (m), 587 (w), 568 (w), 432 (w), 415 (m).

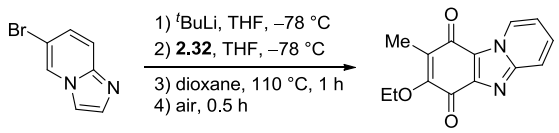
^{1}H NMR (300 MHz, $CDCl_3$) δ_H ppm 8.27 (1 H, dd, *J* = 2.0, 0.9 Hz, Ar-**H**), 7.62 (1 H, *J* = 1.3 Hz, Ar-**H**), 7.54 (1 H, dd, *J* = 0.6, 1.3 Hz, Ar-**H**), 7.50 (1 H, dt, *J* = 0.8, 9.5 Hz, Ar-**H**), 7.20 (1 H, dd, *J* = 9.5, 1.8, Ar-**H**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 142.8 (**C**), 133.3 (**C**H), 126.7 (**C**H), 125.0 (**C**H), 117.2 (**C**H), 111.9 (**C**H), 105.9 (**C**).

LRMS (EI) $^{m}I_z$: 196 ($[M]^{+\bullet}$, 100%).

Spectroscopic data in agreement with that reported in literature.[57]

7-Ethoxy-8-methylbenzo[4,5]imidazo[1,2-a]pyridine-6,9-dione (2.86).



2.80
$C_7H_5BrN_2$
(197.03)

2.86a
$C_{14}H_{12}N_2O_3$
(256.26)

Following a modified procedure established by Harrowven et al.:[4k]

To a solution of ^{t}BuLi (1.55 M in pentane, 0.75 mL, 1.16 mmol) in THF (2 mL) at –78 °C was added imidazopyridine **2.80** (115 mg, 0.58 mmol) in THF (2 mL) dropwise via cannula over 1 min. After 30 min, cyclobutenedione **2.32** (77.2 mg, 0.55 mmol) in THF (2 mL) was added dropwise via cannula over 1 min. After 2 h, sat. $NaHCO_3$ (10 mL) was added, the solution warmed to RT and dichloromehane (15 mL) added. The aqueous phase was separated and extracted with dichloromethane (2 x 15 mL). The organic phases were then combined, washed with brine (2 x 20 mL), dried over $MgSO_4$ and concentrated under reduced pressure to afford a brown/yellow oil (0.133 g, 0.51 mmol). The crude material was dissolved in dioxane (10.4 mL) and heated at 110 °C for 5 h. The resultant solution was stirred in air for 30 min then concentrated at reduced pressure. Purification by flash column chromatography (0-15% ethyl acetate in dichloromethane) gave the title compound **2.86** (21.4 mg, 0.08 mmol, 15%) as an orange solid.

MP 169-173 °C

FTIR υ_{max} (neat, cm^{-1}) 2923 (m), 2852 (w), 1735 (m), 1660 (s), 1599 (w), 1521 (w), 1499 (w), 1457 (w), 1411 (w) 1369 (w), 1306 (s), 1276 (s), 1261 (s), 1181 (m), 1099 (s), 1019 (s), 922 (w), 861 (w), 800 (s), 765 (s), 752 (s).

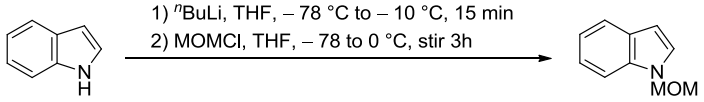
^{1}H NMR (300MHz, $CDCl_3$) δ_H ppm 9.14 (1 H, d, *J* = 6.6 Hz, Ar-**H**), 7.87 (1 H, d, *J* = 8.8 Hz, Ar-**H**), 7.55 (1 H, ddd, *J* = 9.0, 7.1, 1.5 Hz, Ar-**H**), 7.20 (1 H, app. td, *J* = 6.9, 0.9 Hz, Ar-**H**), 4.40 (2 H, q, *J* = 7.0 Hz, OC**H$_2$**), 2.09 (3 H, s, C**H$_3$**), 1.44 (3 H, t, *J* = 7.0 Hz, OCH_2C**H$_3$**).

^{13}C NMR (75 MHz, $CDCl_3$) δ_C ppm 182.8 (**C**O), 173.0 (**C**), 156.8 (**C**), 148.6 (**C**), 145.4 (**C**), 130.0 (**C**H), 129.8 (**C**), 127.7 (**C**H), 119.7 (**C**H), 116.9 (**C**H), 70.0 (O**C**H_2), 16.0 (OCH_2**C**H_3), 9.3 (**C**H_3). *One quaternary carbon missing.*

LRMS (EI) $^{m}I_z$: 256 ($[M]^{+\bullet}$, 52%), 78 (100%).

HRMS (ESI+) $^{m}I_z$ $[MH]^+$ expected 257.0921, detected 257.0919, $[MNa]^+$ expected 279.0740, detected 279.0740.

1-(Methoxymethyl)-1H-indole (3.82).



3.80
C_8H_7N
(117.15)

3.82
$C_{10}H_{11}NO$
(161.20)

Modified from a procedure established by Bosch et al.:[34]

To a solution of indole **3.80** (0.98, 8.38 mmol) in THF (13.4 mL) cooled to –78 °C was added ^{n}BuLi (2.5 M in hexanes, 9.5mmol, 3.8 mL) using a syringe. Once addition complete, the solution was warmed to –10 °C. After 15 min, the solution was cooled back down to –78 °C and MOMCl (0.78 mL, 10.3 mmol) was added using a syringe. Once addition complete, the reaction mixture was warmed to RT. After 3h, sat. $NaHCO_3$ (16 mL) was added and the organic phase separated. The aqueous phase was further extracted with diethyl ether (3 x 15 mL). The combined organic phases were rinsed with brine (15 mL), dried over $MgSO_4$ and concentrated at reduced pressure. Purification by flash chromatography using ethyl acetate inpentane as an eluent mixture (0-5% ethyl acetate in pentane) to afford title compound **3.82** (1.14 g, 7.07 mmol, 84.5%) colourless oil.

FTIR ν_{max} (neat, cm^{-1}) 2931 (w), 1613 (w), 1516 (w), 1462 (m), 1441 (w), 1397 (w), 1333 (w), 1311 (m), 1303 (m), 1231 (m), 1204 (w), 1184 (m), 1153 (w), 1134 (m), 1100 (s), 1085 (m), 1064 (m), 1014 (w), 960 (w), 913 (m), 883 (w), 739 (s), 625(w).

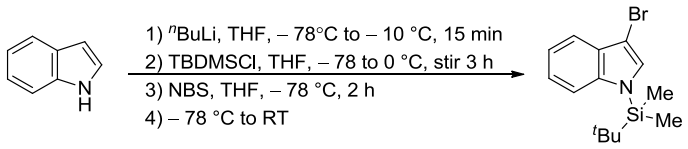
^{1}H NMR (400 MHz, $CO(CD_3)_2$) δ_H ppm 7.58 (1H, dt, *J* = 0.8 7.9 Hz, Ar-**H**), 7.54 (1H,app. dd, *J* = 0.8, 8.2 Hz, Ar-**H**), 7.37 (2H, d, *J* = 3.2 Hz, Ar-**H**), 7.18 (1H, ddd, *J* = 1.1, 7.6, 8.3 Hz, Ar-**H**), 7.08 (1H, ddd, *J* = 1.0, 7.2, 8.0 Hz Ar-**H**), 6.50 (1H, dd, *J* = 0.7, 3,2 Hz, Ar-**H**) 5.53 (2 H, s, NC**H$_2$**O), 3.21 (3 H, s, OC**H$_3$**).

^{13}C NMR (400 MHz, $CO(CD_3)_2$) δ_C ppm 137.6 (**C**), 130.3 (**C**), 129.6 (**C**H), 122.7 (**C**H), 121.6 (**C**H), 120.9 (**C**H), 111.1 (**C**H), 102.9 (**C**H), 78.0 (N**C**H_2), 56.0 (CH_2O**C**H_3).

HRMS (ESI+) $^m\!I_z$ $[MH]^+$ expected 162.0913, detected 162.0919.

Spectroscopic data in agreement with that reported in literature.[58]

3-Bromo-1-(*tert*-butyldimethylsilyl)-1H-indole (3.77a).



3.80
C_8H_7N
(117.15)

3.77a
$C_{14}H_{20}BrNSi$
(310.31)

Following procedure established by Bosch et al.:[34]

To a solution of indole **3.80** (0.75g, 6.43 mmol) in THF (20 mL) cooled to –78 °C was added ^{n}BuLi (2.4 M in hexanes, 7.72mmol, 3.22 mL) using a syringe. Once addition was complete, the solution was warmed to –10 °C. After 15 minutes, the solution was cooled back down to –78 °C and TBDMSCl (1.18 g, 7.86 mmol) in THF (6 mL) was added via cannula. Once addition was complete, the reaction mixture was warmed to 0 °C. After 3h, the solution was cooled back down to –78 °C and NBS (1.15 g, 6.43 mmol) added. The solution was left to stir at –78 °C for 2h before being allowed to warm to RT overnight. Hexane spiked with pyridine (1% v/v) was added and the resultant solution filtered through

Celite®. The concentrated crude mixture was purified by flash chromatography using neat hexane to afford title compound **3.76a** (1.88 g, 0.60 mmol, 94%) as an colourless solid.

N.B.: **3.76a** should be stored in the freezer as it decomposes over time at room temperature (turns green).

MP decomposes above 73 °C (Lit[59] 73.2-74,1 °C)

FTIR ν_{max} (neat, cm^{-1}) 2956 (w), 2929 (w), 2858 (w), 1608 (w), 1514 (w), 1449 (s), 1362 (w), 1286 (s), 1257 (s), 1198 (m), 1160 (m), 1142 (s), 1076 (w), 1018 (m), 985 (w), 932 (m), 839 (s), 819 (s), 809 (s), 792 (s), 739 (s), 692 (m), 669 (w), 634 (w).

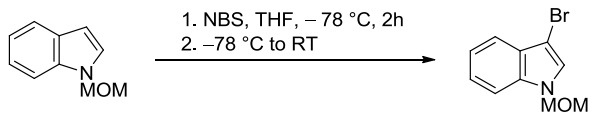
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 7.60 – 7.56 (1H, m, Ar-**H**), 7.51 – 7.48 (1H, m, Ar-**H**), 7.23 – 7.19 (2H, m, Ar-**H**), 7.18 (1H, s, Ar-**H**), 0.95 (9 H, s, C(C**H$_3$**)$_3$), 0.61 (6 H, s, SiC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 140.3 (**C**), 129.9 (**C**), 129.7 (**C**H), 122.5 (**C**H), 120.6 (**C**H), 119.2 (**C**H), 114.1 (**C**H), 93.7 (**C**), 26.2 (C(**C**H_3)$_3$), 19.4 (**C**), -4.0 (Si**C**H_3).

LRMS (ESI+) $^m/_z$: 310 ([M(^{79}Br)H]$^+$, 53%), 312 ([M(^{81}Br)H]$^+$, 61%), 232.1 (100%).

Spectroscopic data in agreement with that reported in literature.[34]

3-Bromo-1-(methoxymethyl)-1H-indole (3.77c).



3.82
$C_{10}H_{11}NO$
(161.20)

3.77c
$C_{10}H_{11}BrNO$
(240.10)

Following procedure established by Bosch et al.:[34]

To a solution of indole **3.82** (0.63g, 3.91 mmol) in THF (15 mL) cooled to –78 °C was added NBS (0.75g, 4.20 mmol). After 2h, the reaction mixture was allowed to warmed to RT overnight, and pentane (30 mL) spiked with pyridine (1% v/v) was added. The resultant solution was filtered through Celite®, concentrated at reduced

pressure and purified by flash chromatography using 10% diethyl ether in pentane to afford title compound **3.77c** as an orange oil (0.63g, 2.62 mmol, 67%).

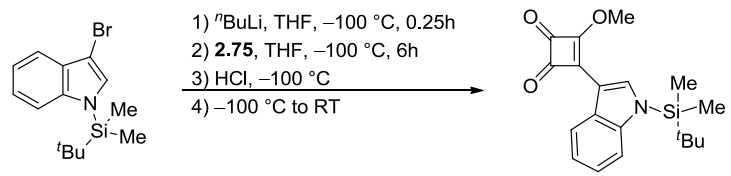
FTIR ν_{max} (neat, cm^{-1}) 2940 (w), 1746 (s), 1610 (s), 1483 (s), 1471 (s), 1361 (m), 1339 (s), 1293 (m), 1242 (m), 1185 (m), 1158 (w), 1121 (s), 1093 (s), 1075 (s), 1020 (w), 938 (w), 905 (m), 810 (m), 750 (s), 683 (s), 663 (m), 629 (s), 605 (s).

^{1}H NMR (400 MHz, $CO(CD_3)_2$) δ_H ppm 7.60 (1H, app. d, *J* = 8.2 Hz, Ar-**H**), 7.55 (1H, s, Ar-**H**), 7.50 (1H, dt, *J* = 1, 7.8 Hz, Ar-**H**), 7.29 (1H, ddd, *J* = 0.9, 7.2, 8.1 Hz, Ar-**H**), 7.22 (1H, ddd, *J* = 1.0, 7.0, 8.0 Hz, Ar-**H**), 5.56 (s, 2H, NC**H**$_2$O), 3.24 (s, 3H, OC**H**$_3$).

^{13}C NMR (101 MHz, $CO(CD_3)_2$) δ_C ppm 137.1(**C**), 128.8 (**C**H), 128.8 (**C**), 124.0 (**C**H), 121.9 (**C**H), 119.7 (**C**H), 111.7 (**C**H), 91.3 (**C**), 78.2 (N**C**H$_2$O), 56.2 (O**C**H$_3$).

HRMS (ESI+) Molecular ion *could not be detected by HRMS.*

3-(1-(*tert*-Butyldimethylsilyl)-1H-indol-3-yl)-4-methoxycyclobut-3-ene-1,2-dione (3.76a).



3.77a
$C_{14}H_{20}BrNSi$
(310.31)

3.76a
$C_{19}H_{23}NO_3Si$
(341.48)

Following a procedure established by Hu et al.:[36]

To a solution of indole **3.77a** (0.31 g, 1 mmol) in THF (6 mL) at –100 °C was added a solution of ^{n}BuLi (2.5 M in hexanes, 0.48 mL, 1.2 mmol) over 1 minute using a syringe. After 0.25 h, dimethyl squarate **2.75** (138.8 mg , 0.97 mmol) in THF (2 mL) was added via cannula. After 6h, 6M HCl (3 eq, 0.5 mL) was added and the solution allowed to warm to RT. After 30 minutes, the solution was basified with sat. $NaHCO_3$ (10 mL). The aqueous phase was separated and extracted with DCM (3 x 15 mL). The organic phases were combined, dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in pentane) to give **3.76a** (160.4 mg, 0.18 mmol, 48%) as a yellow solid.

MP 164 - 166 ºC

FTIR ν_{max} (neat, cm^{-1}) 2393 (w), 1784 (s), 1735 (s), 1616 (m), 1594 (s), 1508 (s), 1480 (m), 1458 (m), 1389 (s), 1363 (w), 1336 (w), 1289 (m), 1256 (m), 1228 (m), 1182 (s), 1157 (m), 1134 (w), 1105 (m), 1080 (w), 1035 (m), 968 (s), 931 (m), 841 (s), 814 (s), 792 (s), 749 (s), 688 (m), 615 (m).

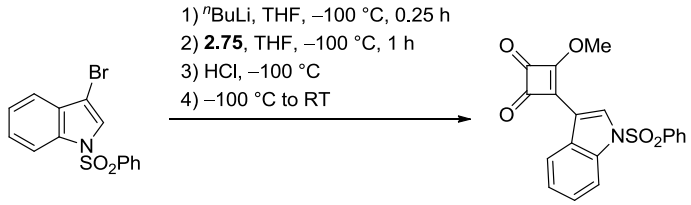
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.24 (1H, m Ar-**H**), 8.22 (1H, s, Ar-**H**), 7.55 (1H, m, Ar-**H**), 7.29 (2H, m, Ar-**H**), 4.64 (3H, s, OC**H$_3$**), 0.97 (9 H, s, C(C**H$_3$**)$_3$), 0.69 (6 H, s, SiC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 192.9 (**C**O), 190.2 (**C**O), 189.7 (**C**), 170.7 (**C**), 141.7 (**C**), 136.5 (**C**H), 127.9 (**C**), 123.5 (**C**H), 122.3 (**C**H), 122.2 (**C**H), 114.5 (**C**H), 109.1 (**C**), 61.1 (O**C**H$_3$), 26.2 (C(**C**H$_3$)$_3$), 19.2 (**C**(CH$_3$)$_3$), -4.0 (Si(**C**H$_3$)$_2$).

LRMS (ESI+) $^m\!/_z$: 342 ([MH]$^+$, 100%).

HRMS (ESI+) $^m\!/_z$ [MH]$^+$ expected 342.1520, detected 342.1533, [MNa]$^+$ expected 364.1339, detected 364.1351.

3-Methoxy-4-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-3-ene-1,2-dione (3.76b).



3.77b
$C_{14}H_{10}BrNO_2S$
(336.20)

3.76b
$C_{19}H_{13}NO_5S$
(367.38)

Following a procedure established by Hu et al.:[36]

To a solution of indole **3.77b** (481.4 g, 1.43 mmol) in THF (6 mL) at –100 °C was added a solution of ^{n}BuLi (2.5 M in hexanes, 0.69 mL, 1.73 mmol) over 1 minute using a syringe. After 0.25 h, dimethyl squarate **2.75** (198.2 mg , 1.39 mmol) in THF (5 mL) was added via cannula. After 1h, 6M HCl (5 eq, 1.19 mL) was added and the solution allowed to warm to RT. After 30 minutes, sat. $NaHCO_3$ (10 mL) was added and the organic phase was separated. The aqueous phase was then extracted with DCM (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated at reduced pressure and purified by

flash chromatography (0-25% ethyl acetate in pentane) to give **3.76b** (400.3 mg, 1.1 mmol, 78%) as a yellow solid.

MP Decomposes above 190 ºC (black tar formed 205 – 210 ºC)

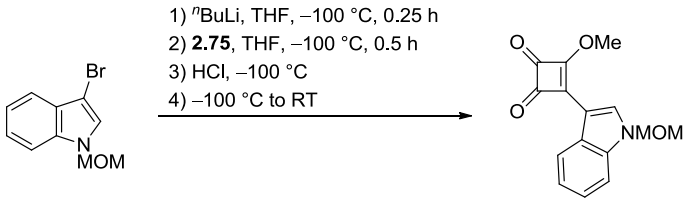
FTIR ν_{max} (neat, cm^{-1}) 3114 (w), 1790 (s), 1755 (s), 1620 (m), 1611 (s), 1513 (s), 1481 (w), 1451 (m), 1373 (s), 1320 (w), 1265 (w), 1221 (w), 1178 (s), 1137 (s), 1114 (s), 1089 (s), 1033 (s), 971 (s), 921 (w), 839 (w), 753 (s), 727 (s), 689 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.49 (1H, s, Ar-**H**), 8.26 – 8.21 (1H, m, Ar-**H**), 8.02 (1H, app. d, *J* = 8.2Hz, Ar-**H**), 8.00 – 7.97 (2H, m, Ar-**H**), 7.64 – 7.59 (1H, m, Ar-**H**), 7.54 – 7.49 (2H, m, Ar-**H**), 7.43 (1H, ddd, *J* = 1.3, 7.4, 8.3 Hz , Ar-**H**), 7.40 – 7.35 (1H, m, Ar-**H**), 4.66 (3H, s, OC**H**$_3$).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C 192.5 (**C**O), 191.5 (**C**O), 190.7(**C**), 169.1(**C**), 137.5 (**C**), 134.8 (**C**), 134.6 (**C**H), 129.7 (**C**H), 128.6 (**C**H), 127.2 (**C**H), 127.1 (**C**) 126.1 (**C**H), 124.7 (**C**H), 123.0 (**C**H), 113.5 (**C**H), 111.2 (**C**), 61.7 (**C**H).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 368.0587, detected 368.0589.

3-Methoxy-4-(1-(methoxymethyl)-1H-indol-3-yl)cyclobut-3-ene-1,2-dione (3.76c).



3.77c
$C_{10}H_{10}BrNO$
(240.10)

3.76c
$C_{15}H_{13}NO_4$
(271.27)

Following a procedure established by Hu et al.:[36]

To a solution of indole **3.77c** (0.76 g, 3.18 mmol) in THF (30 mL) at –100 °C was added a solution of ^{n}BuLi (1.60 M in hexanes, 2.40 mL, 3.84 mmol) over 1 min using a syringe. After 0.25 h, dimethyl squarate **2.75** (0.38 g, 2.67 mmol) in THF (27 mL) was added via cannula. After 1 h, 6M HCl (2 eq, 0.9 mL) was added and the solution allowed to warm to RT. After 10 minutes, the solution was basified with sat. $NaHCO_3$ (10 mL). The aqueous

phase was separated and extracted with ethyl acetate (3 x 30 mL). The organic phases were combined, dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in petrol) to give **3.76c** (0.51 g, 1.88 mmol, 70%) as a light yellow solid.

MP 155 - 157 ºC

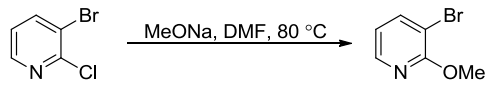
FTIR ν_{max} (neat, cm^{-1}) 1790 (m), 1734 (w), 1720 (m), 1600 (s), 1508 (s), 1468 (w), 1396 (s), 1307 (m), 1244 (m), 1191 (w), 1159 (w), 1130 (m), 1100 (s), 1055 (w), 1014 (w), 931 (w), 917 (w), 866 (w), 817 (w), 763 (w), 744 (s), 652 (w), 638 (w), 621 (m).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.24 (1H, s,Ar-**H**), 8.24 – 8.20 (1H, m, Ar-**H**), 7.58 - 7.54 (1H, m, Ar-**H**), 7.41 – 7.31 (m, 2H, Ar-**H**), 5.54 (s, 2H, NC**H**$_2$O), 4.65 (s, 3H, OC**H**$_3$), 3.31 (s, 3H, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 192.8 (**C**O), 190.4 (**C**O), 189.6 (**C**), 170.3 (**C**), 136.8 (**C**), 132.2 (**C**H), 126.1 (**C**), 124.3 (**C**H), 123.0 (**C**H), 122.4 (**C**H), 111.0 (**C**H), 106.4 (**C**), 78.4 (N**C**H$_2$O), 61.2 (O**C**H$_3$) 56.4 (CH$_2$O**C**H$_3$).

HRMS (ESI+) $^m/_z$ $[MH]^+$ 272.0907 expected , 272.0917detected.

3-Bromo-2-methoxypyridine (3.86).



3.130
C_5H_3BrClN
(192.44)

3.86
C_6H_6BrNO
(188.02)

Following a procedure established by Testaferri et al.:[39]

A solution of 3-bromo-2-chloropyridine **3.130** (0.57 g, 2.96 mmol) and NaOMe (0.84 g, 15.54 mmol) in dry DMF (12 mL) was heated to 80 °C under argon. Once complete consumption of starting material was observed by TLC, the mixture was allowed to cool to RT before being poured into water (15 mL). The aqueous phase was separated and further extracted with diethyl ether (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-50% diethyl ether in pentane) to give **3.86** (0.40 g, 2.12 mmol, 72%) as a pale yellow oil.

FTIR ν_{max} (neat, cm^{-1}) 1584 (s), 1468 (s), 1440 (w), 1403 (s), 1298 (m), 1255 (m), 1121 (w), 1070 (m), 1034 (s), 1010 (m), 785 (m), 752 (m), 676 (m).

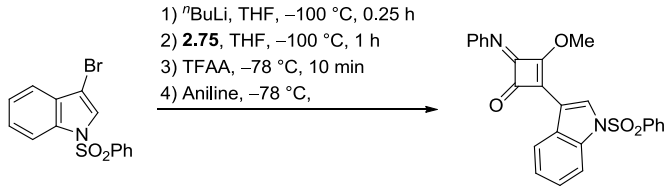
^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.11 (1H, dd, *J* = 1.7, 4.9 Hz, Ar-**H**), 7.81 (1H, dd, *J* = 1.7, 7.6 Hz, Ar-**H**), 6.78 (1H, dd, *J* = 4.9, 7.6 Hz, Ar-**H**), 4.03 (3H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 160.1 (**C**), 145.5 (**C**H), 141.6 (**C**H), 117.8 (**C**H), 107.1 (**C**), 54.4 (**C**H).

LRMS (ESI+) $^{m}/_{z}$: $[M(^{79}Br)H]^+$ 188, $[M(^{81}Br)H]^+$ 190, 507 (100%).

Spectroscopic data in agreement with that reported in literature.[39]

3-Methoxy-4-(phenylimino)-2-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-2-en-1-one (3.90).



3.77b
$C_{14}H_{10}BrNO_2S$
(336.20)

3.90
$C_{25}H_{18}N_2O_4S$
(442.49)

Modified from a procedure established by Hu et al.:[36]

To a solution of indole **3.77b** (441.4 g, 1.30 mmol) in THF (8 mL) at –100 °C was added a solution of ^{n}BuLi (2.36 M in hexanes, 0.68 mL, 1.60 mmol) over 1 min using a syringe. After 0.25 h, dimethyl squarate **2.75** (169.6 mg , 1.19 mmol) in THF (8 mL) was added via cannula. After 1 h, TFAA (1.22 mL, 1.58 mmol) was added using a syringe. After 10 min, the solution was warmed to –78 °C and aniline (147.5 mg, 1.58 mmol) was added. The solution was then warmed to RT. After 30 min, sat. $NaHCO_3$ (10 mL) was added and the organic phase was separated. The aqueous phase was extracted with DCM (3 x 15 mL). The combined organic phases were dried over $MgSO_4$, concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in pentane) to give **3.90** (253.2 mg, 0.57 mmol, 49%) as a yellow solid.

MP 166 - 167 ºC

FTIR ν_{max} (neat, cm^{-1}) 1775 (w), 1678 (m), 1597 (w), 1574 (m), 1539 (m), 1481 (w), 1447 (m), 1385 (s), 1329 (w), 1278 (w), 1180 (s), 1132 (s),

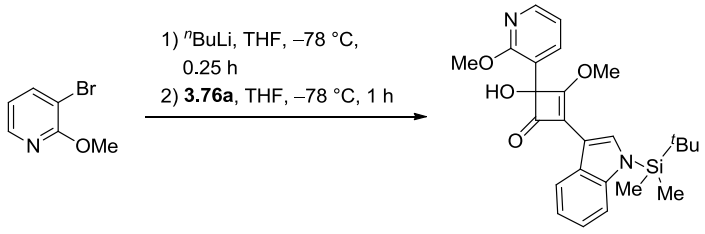
1101 (s), 1030 (m), 1021 (m), 969 (s), 835 (w), 802 (w), 753 (s), 727 (s), 686 (s).

^{1}H NMR (400 MHz, $(CD_3)_2CO$) δ_H ppm 8.30 - 8.26 (1H, m, Ar-**H**), 8.27 (1H, s, Ar-**H**), 8.11 - 8.08 (2H, m, Ar-**H**), 8.06 (1H, dt, *J* = 0.7, 8.3 Hz, Ar-**H**) 7.76 - 7.70 (1H, m, Ar-**H**), 7.66 - 7.61 (2H, m, Ar-**H**), 7.49 – 7.41 (3H, m, Ar-**H**), 7.41 – 7.34 (3H, m, Ar-**H**), 7.23 - 7.18 (1H, m, Ar-**H**), 4.86 (3H, s, OC**H**$_3$).

^{13}C NMR (101 MHz, $(CD_3)_2CO$) δ_C ppm 187.0 (**C**O), 185.4 (**C**), 168.2 (**C**), 149.5 (**C**), 147.4 (**C**), 138.6 (**C**), 136.0 (**C**H), 135.8 (**C**), 131.0 (**C**H), 129.7 (**C**H), 128.7 (**C**), 128.2 (**C**H), 127.1 (**C**H), 127.0 (**C**H), 127.0 (**C**H), 125.4 (**C**H), 124.7 (**C**H), 124.1 (**C**H), 114.5 (**C**H), 112.9 (**C**), 62.6 (O**C**H$_3$).

HRMS (ESI+) $^{m}I_z$ $[MH]^+$ expected 443.1060, detected 443.1073.

2-(1-(*tert*-Butyldimethylsilyl)-1H-indol-3-yl)-4-hydroxy-3-methoxy-4-(2-methoxypyridin-3-yl)cyclobut-2-enone (3.75a).



3.86
C_6H_6BrNO
(188.02)

3.75a
$C_{25}H_{30}N_2O_4Si$
(450.61)

Following a procedure established by Harrowven et al.:[3]

To a solution of 3-bromo-2-methoxypyridine **3.86** (49.6 mg, 0.26 mmol) in THF (2.5 mL) at –78 °C was added a solution of ^{n}BuLi (2.63 M in hexanes, 0.12 mL, 0.32 mmol) over 1 min using a syringe. After 0.5 h, cyclobutenedione **3.76a** (68.8 mg, 0.2 mmol) in THF (3 mL) was added via cannula. After 1 h, sat. $NaHCO_3$ (10 mL) was added and the solution warmed to RT. The aqueous phase was separated and extracted further with DCM (3 x 15 mL). The combined organic phases were dried over Na_2SO_4, concentrated at reduced pressure and purified by flash chromatography (0-10% MeOH in DCM) to give **3.75a** (48.9 mg, 0.11 mmol, 75%) as a yellow oil.

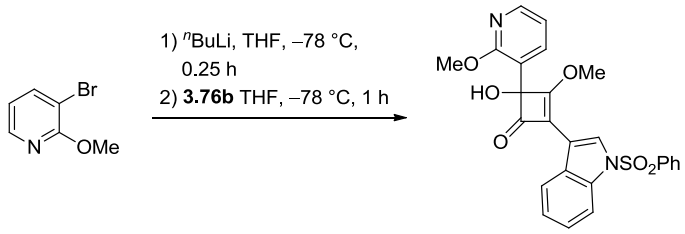
FTIR ν_{max} (neat, cm^{-1}) 3375 (m), 2954 (m), 2930 (m), 2857 (m), 1756 (s), 1637 (s), 1584 (s), 1522 (s), 1465 (s), 1454 (s), 1408 (s), 1378 (s), 1328 (m), 1299 (m), 1259 (s), 1149 (s), 1011 (s), 973 (w), 945 (w), 839 (m), 791 (m), 810 (m), 744 (m), 691 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.26 – 8.11 (2H, m, Ar-**H**), 7.75 (1H, d, *J* = 6.9 Hz, Ar-**H**), 7.69 (1H, s, Ar-**H**), 7.54 - 7.47 (1H, m, Ar-**H**), 7.24 - 7.14 (2H, m, Ar-**H**), 7.01 - 6.90 (1H, m, Ar-**H**), 4.78 (1H, s, O**H**), 4.18 (3H, s, OC**H$_3$**), 4.05 (3H, s, OC**H$_3$**), 0.95 (9H, s, C(C**H$_3$**)$_3$), 0.63 (6H, s, Si(C**H$_3$**)$_2$).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 186.0 (**C**O), 173.7 (**C**), 160.6 (**C**), 146.8 (**C**H), 140.9 (**C**), 136.7 (**C**H), 132.1 (**C**H), 128.7 (**C**H), 123.3 (**C**), 122.2 (**C**H), 121.6 (**C**H), 120.5 (**C**H), 119.9 (**C**), 117.4 (**C**H), 114.0 (**C**H), 107.4 (**C**), 91.7 (**C**), 59.9 (O**C**H_3), 53.9 (O**C**H_3),26.3 (SiC(**C**H_3)$_3$), 19.3 (**C**(CH_3)$_3$), −4.0 (Si(**C**H_3)$_2$).

HRMS(ESI+) $^{m}I_z$ $[MH]^+$ expected 451.2048, detected 451.2061, $[MNa]^+$ expected 473.1867, detected 473.1871.

4-Hydroxy-3-methoxy-4-(2-methoxypyridin-3-yl)-2-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-2-enone (3.75b).



3.86
C_6H_6BrNO
(188.02)

3.75b
$C_{25}H_{20}N_2O_6S$
(476.50)

Following a procedure established by Harrowven et al.:[3]

To a solution of 3-bromo-2-methoxypyridine (**3.86**) (0.24 g, 1.27 mmol) in THF (12 mL) at –78 °C was added a solution of ^{n}BuLi (2.55 M in hexanes, 0.60 mL, 1.53 mmol) over 1 min using a syringe. After 0.25 h, cyclobutenedione **3.76b** (237 mg, 0.64 mmol) in THF (8 mL) was added via cannula. After 1 h, sat. $NaHCO_3$ (10 mL) was added and the solution allowed to warm to RT. The aqueous phase was separated and extracted further with DCM (3 x 15 mL). The combined organic phases were dried over Na_2SO_4, concentrated at

reduced pressure and purified by flash chromatography (0-50% ethyl acetate in pentane) to give **3.75b** (123 mg, 0.26 mmol, 40%) as a yellow oil.

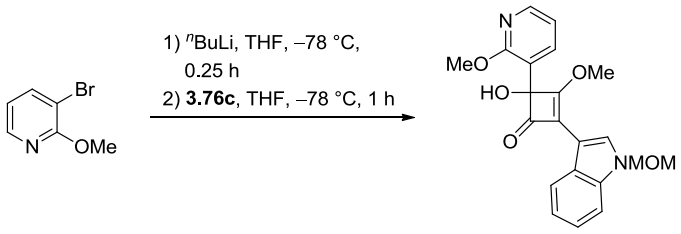
FTIR ν_{max} (neat, cm^{-1}) 3382 (br. w), 2955 (w), 1759 (m), 1643 (s), 1584 (s), 1532 (m), 1466 (s), 1448 (s), 1408 (s), 1372 (s), 1320 (m), 1265 (m), 1176 (s), 1133 (s), 1108 (s), 1091 (s), 1010 (s), 976 (w), 947 (m), 910 (w), 733 (s), 686 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.19 – 8.13 (2H ,m, Ar-**H**), 8.00 (1H, s, Ar-**H**), 7.98 (1H, app. d, *J* = 8.3 Hz, Ar-**H**), 7.93 – 7.89 (2H, m, Ar-**H**), 7.77 (1H, dd, *J* = 1.8, 7.5 Hz, Ar-**H**), 7.56 - 7.51 (1H, m, Ar-**H**), 7.47 - 7.41 (2H, m, Ar-**H**), 7.37 - 7.31 (1H, m, Ar-**H**), 7.30 - 7.23 (1H, m, Ar-**H**), 6.96 (1H, dd, *J* = 4.9, 7.5 Hz, Ar-**H**), 4.98 (1H, br. s, O**H**), 4.17 (3H, s, OC**H$_3$**), 3.98 (3H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 185.4 (**C**O), 177.5 (**C**), 160.4 (**C**), 147.0 (**C**H), 138.0 (**C**), 136.8 (**C**H), 134.5 (**C**), 134.0 (**C**H), 129.3 (**C**H), 128.4 (**C**), 126.9 (**C**H), 125.5 (**C**H), 125.2 (**C**H), 123.7 (**C**H), 122.6 (**C**H), 120.6 (**C**), 119.1 (**C**), 117.5 (**C**H), 113.3 (**C**H), 111.2 (**C**), 91.7 (**C**), 60.3 (O**C**H$_3$), 53.9 (O**C**H$_3$).

HRMS (ESI+) $^m/_z$ $[MH]^+$ 477.1115 expected , 477.1116 detected, $[MNa]^+$ 499.0934 expected , 499.0920 detected.

4-Hydroxy-3-methoxy-2-(1-(methoxymethyl)-1H-indol-3-yl)-4-(2-methoxypyridin-3-yl)cyclobut-2-enone (3.75c).



3.86
C_6H_6BrNO
(188.02)

3.75c
$C_{21}H_{20}N_2O_5$
(380.39)

Following a procedure established by Harrowven et al.:[3]

To a solution of 3-bromo-2-methoxypyridine **3.86** (0.35 g, 1.81 mmol) in THF (15 mL) at –78 °C was added a solution of ^{n}BuLi (1.55 M in hexanes, 1.40 mL, 2.17 mmol) over 1 min using a syringe. After 0.25 h, cyclobutenedione **3.76c** (0.27 g, 1.01 mmol) in THF (15 mL) was added via cannula. After 1 h, sat. $NaHCO_3$ (10 mL) was added and the solution

allowed to warm to RT. The aqueous phase was separated and extracted further with ethyl acetate (3 x 30 mL). The combined organic phases were dried over Na_2SO_4, concentrated at reduced pressure and purified by flash chromatography (0-50% ethyl acetate in pentane) to give **3.75c** (0.31 g, 0.80 mmol, 80%) as a yellow oil.

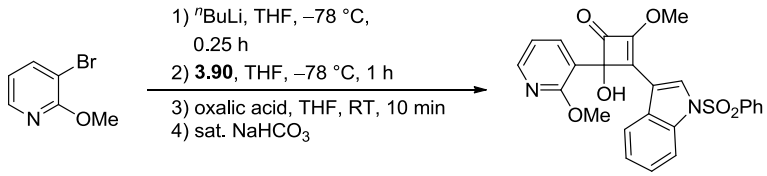
FTIR ν_{max} (neat, cm^{-1}) 3354 (w), 2953 (w), 1757 (m), 1642 (s), 1613 (w), 1584 (s), 1524 (s), 1466 (s), 1406 (s), 1379 (m), 1308 (s), 1262 (s), 1181 (w), 1160 (w), 1130 (m), 1102 (s), 1086 (s), 1018 (s), 955 (w), 913 (m), 832 (m), 747 (s), 703 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.19 (1H, app. d, *J* = 7.8 Hz, Ar-H), 8.15 (1H, dd, *J* = 1.9, 5.0 Hz, Ar-**H**), 7.84 (1H, dd, *J* = 1.9, 7.5 Hz, Ar-**H**), 7.67 (1H, s, Ar-**H**), 7.47 (1H, app. d, *J* = 8.1 Hz, Ar-**H**), 7.28 (1H, ddd, *J* = 1.1, 7.2, 8.2 Hz, Ar-**H**), 7.22 (1H, ddd, *J* = 1.0, 7.2, 8.0 Hz, Ar-**H**), 6.96 (1H, dd, *J* = 5.0, 7.5 Hz, Ar-**H**), 5.39 (1H, d, *J* = 11.0 Hz, NC**H**HO), 5.35 (1H, d, *J* = 11.1 Hz, NCH**H**O), 5.15 (1H, br. s, O**H**), 4.15 (3H, s, OC**H$_3$**), 3.99 (3H, s, OC**H$_3$**), 3,22 (3H, s, CH_2OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 186.3 (**C**O), 174.9 (**C**), 160.5 (**C**), 146.7 (**C**H), 136.9 (**C**H), 136.0 (**C**), 128.6 (**C**H), 126.6 (**C**), 122.9 (**C**H), 122.8 (**C**), 121.9 (**C**H), 120.9 (**C**H), 119.8 (**C**), 117.4 (**C**H), 110.0 (**C**H), 105.1 (**C**), 91.3 (**C**), 77.6 (N**C**H_2O), 59.8 (O**C**H_3), 55.9 (O**C**H_3), 53.8 (CH_2O**C**H_3).

HRMS(ESI+) $^m/_z$ $[MH]^+$ expected 381.1445, detected 381.1450, $[MNa]^+$ expected 403.1264, detected 403.1278.

4-Hydroxy-2-methoxy-4-(2-methoxypyridin-3-yl)-3-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-2-enone (3.87).



3.86
C_6H_6BrNO
(188.02)

3.87b
$C_{25}H_{20}N_2O_6S$
(476.50)

Following procedure established by Trost et al.:[8]

To a solution of 3-bromo-2-methoxypyridine **3.86** (80.2 mg, 0.42 mmol) in THF (4 mL) at –78 °C was added a solution of ^{n}BuLi (2.33 M in hexanes, 0.22 mL, 0.51 mmol) over 1 min using a syringe. After 0.25 h, cyclobutenedione **3.90** (151.6 mg , 0.34 mmol) in THF (3 mL) was added via cannula. After 1 h, sat. $NaHCO_3$ (10 mL) was added and the solution allowed to warm to RT. The aqueous phase was separated and extracted further with Et_2O (3 x 15 mL). The combined organic phases were dried over Na_2SO_4, concentrated at reduced pressure. The crude mixture was re-dissolved in THF (7 mL) and oxalic acid (90.7 mg) in water (4 mL). After 10 min., sat. $NaHCO_3$ (10 mL) was added to the reaction mixture. The aqueous phase was then separated and extracted further with Et_2O (3 x 15 mL). The combined organic phases were dried over Na_2SO_4, concentrated at reduced pressure purified by flash chromatography (0-50% ethyl acetate in pentane) to give **3.87b** (13.0 mg, 0.06 mmol, 7 %) as a yellow oil.

FTIR ν_{max} (neat, cm^{-1}) 3394 (br. w), 1758 (s), 1637 (m), 1583 (m), 1526 (m), 1465 (m), 1449 (s), 1407 (s), 1378 (s), 1319 (w), 1271 (w), 1175 (s), 1136 (s), 1099 (s), 1042 (m), 1011 (s), 982 (m), 858 (m), 789 (m), 750 (m), 727 (s), 685 (s).

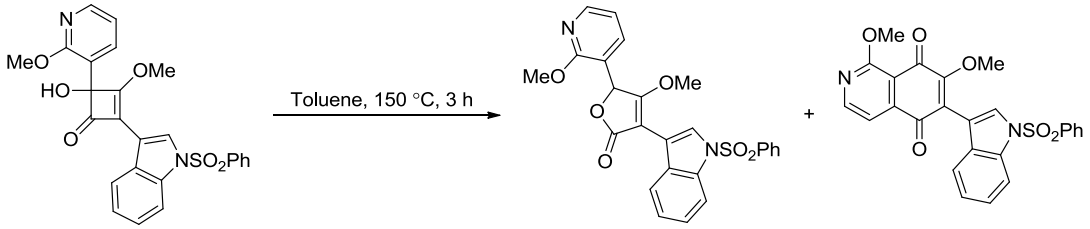
^{1}H NMR (400 MHz, $CO(CD_3)_2$) δ_H ppm 8.23 (1 H, dd, *J* = 1.9, 7.4 Hz, Ar-**H**), 8.12 (1 H, dd, *J* = 1.9, 4.9 Hz, Ar-**H**), 8.05 - 7.97 (2H, m, Ar-**H**), 7.89 - 7.84 (2 H, m), 7.83 (1 H, s, Ar-**H**), 7.71 - 7.65 (1 H, m, Ar-**H**), 7.59 - 7.53 (2 H, m, Ar-**H**), 7.42 - 7.36 (1 H, m, Ar-**H**), 7.31 - 7.25 (1 H, m, Ar-**H**), 7.11 (1 H, dd, *J* = 5.0, 7.5 Hz, Ar-**H**), 5.73 (1 H, br. s, O**H**), 4.29 (3 H, s, OC**H$_3$**), 3.74 (3 H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CO(CD_3)_2$) δ_C ppm 188.6 (**C**O), 162.1 (**C**), 154.1 (**C**), 147.7 (**C**H), 143.7 (**C**), 139.2 (**C**H), 138.7 (**C**), 136.3 (**C**), 136.1 (**C**H), 131.2 (**C**H), 129.6 (**C**), 128.2 (**C**H), 127.5 (**C**H), 127.1 (**C**H), 125.5 (**C**H),

124.1 (**C**H), 122.7 (**C**), 118.5 (**C**H), 115.3 (**C**), 114.8 (**C**H), 88.1 (**C**), 60.0 (O**C**H_3), 54.1 (O**C**H_3)

HRMS (ESI+) m/z $[MH]^+$ 477.1115 expected, 477.1120 detected; $[MNa]^+$ 499.0934 expected , 499.0948 detected.

1,7-Dimethoxy-6-(1-(phenylsulfonyl)-1H-indol-3-yl)isoquinoline-5,8-dione (3.74b) and 4-Methoxy-5-(2-methoxypyridin-3-yl)-3-(1-(phenylsulfonyl)-1H-indol-3-yl)furan-2(5H)-one (3.92b).



3.75b
$C_{25}H_{20}N_2O_6S$
(476.50)

3.92b
$C_{25}H_{20}N_2O_6S$
(476.50)

3.74b
$C_{25}H_{18}N_2O_6S$
(474.49)

Modified from a procedure established by Harrowven et al.:[3]

A solution of **3.75b** (24.2 mg, 0.05 mmol) in toluene (1.00 mL) was heated to 150 °C in a sealed vessel. After 3 h, the resultant mixture was concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in pentane) to give **3.74b** first (6.90 mg, 0.02 mmol, 28%) as an orange oil, followed by **3.92b** (12.8 mg, 0.03 mmol, 53%) as a yellow oil.

Data for **3.74b**:

FTIR ν_{max} (neat, cm^{-1}) 2923 (w), 1715 (w), 1672 (m), 1574 (m), 1469 (m), 1448 (s), 1391 (m), 1370 (m), 1306 (m), 1236 (w), 1215(w), 1175 (s), 1135 (m), 1118 (w), 1093 (m), 1014 (m), 970 (m), 930 (w), 792 (w), 755 (m), 726 (s), 686 (m), 635 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.59 (1H, d, *J* = 5.0 Hz, Ar-**H**), 8.04 - 8.01 (1H, m, Ar-**H**), 7.99 – 7.95 (2H, m, Ar-**H**), 7.89 (1H, s, Ar-**H**), 7.60 (1H, d, *J* = 5.1 Hz, Ar-**H**), 7.58 – 7.54 (1H, m, Ar-**H**), 7.51 – 7.45 (2H, m, Ar-**H**), 7.36 - 7.31 (2H, m, Ar-**H**), 7.26 - 7.22 (1H, m, Ar-**H**), 4.19 (3H, s, OC**H$_3$**), 3.99 (3H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 182.9 (**C**O), 179.3 (**C**O), 162.2 (**C**), 158.1 (**C**), 153.5 (**C**H), 141.7 (**C**), 138.1 (**C**), 134.6 (**C**), 134.0 (**C**H), 129.7 (**C**), 129.4 (**C**H), 128.6 (**C**H), 127.0 (**C**H), 124.8 (**C**H), 123.5 (**C**H), 121.9 (**C**), 121.6 (**C**H), 113.5 (**C**H), 113.1 (**C**H), 112.7 (**C**), 111.5 (**C**), 61.5 (O**C**H_3), 55.0 (O**C**H_3).

HRMS(ESI+) $^{m}/_{z}$ $[MH]^+$ expected 475.0958, detected 475.0969.

Data for **3.92**:

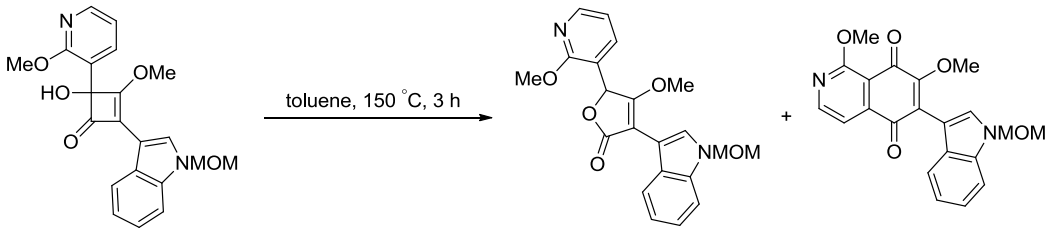
FTIR ν_{max} (neat, cm^{-1}) 2954 (w), 2924 (w), 2850 (w), 1755 (s), 1675 (s), 1587 (m), 1556 (w), 1469 (s), 1448 (s), 1416 (s), 1370 (s), 1328 (m), 1176 (s), 1124 (s), 1086 (m),1054 (s), 1019 (s), 993 (s), 941 (w), 910 (w), 873 (w), 860 (w), 755 (s), 728 (s), 686 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.26 (1H, dd, *J* = 1.9, 5.0 Hz, Ar-**H**), 8.0 (1H, d, *J* = 8.2 Hz, Ar-**H**), 7.98 – 7.91 (2H, m, Ar-**H**), 7.93 (1H, s, Ar-**H**), 7.67 (1H, d, *J* = 7.9 Hz, Ar-**H**), 7.58 (1H, app. d, *J* = 7.7 Hz, Ar-**H**), 7.54 (1H, d, *J* = 7.3 Hz, Ar-**H**), 7.49 - 7.42 (2H, m, Ar-**H**), 7.39 - 7.32 (1H, m, Ar-**H**), 7.31 - 7.25 (1H, m, Ar-**H**), 6.97 (1H,dd, *J* = 5.1, 6.9 Hz, Ar-**H**), 6.24 (1H, s, C**H**), 4.05 (3H, s, OC**H_3**), 3.75 (3H, s, OC**H_3**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 174.1(**C**O), 171.7 (**C**), 161.8 (**C**), 148.5 (**C**H), 138.0 (**C**), 137.2 (**C**H), 134.7 (**C**), 134.0 (**C**H), 130.0 (**C**), 129.4 (**C**H),126.9 (**C**H), 126.4 (**C**H), 125.1 (**C**H), 123.6 (**C**H), 121.4 (**C**H), 117.2 (**C**H), 116.5 (**C**), 113.5 (**C**H), 111.1 (**C**), 97.6 (**C**), 73.7 (**C**H), 59.2 (O**C**H_3), 53.9 (O**C**H_3).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ expected 477.1116, detected 477.1115, $[MNa]^+$ expected 499.0934, detected 499.0944.

1,7-Dimethoxy-6-(1-(methoxymethyl)-1H-indol-3-yl)isoquinoline-5,8-dione (3.74c) and 4-Methoxy-3-(1-(methoxymethyl)-1H-indol-3-yl)-5-(2-methoxypyridin-3-yl)furan-2(5H)-one (3.92c).



3.75c
$C_{21}H_{20}N_2O_5$
(380.4)

3.92c
$C_{21}H_{20}N_2O_5$
(380.4)

3.74c
$C_{21}H_{18}N_2O_5$
(378.38)

Modified from a procedure established by Harrowven et al.:[3]

A solution of **3.75c** (77.1 mg, 0.20 mmol) in toluene (4.2 mL) was heated to 150 °C in a sealed vessel for 1 h. After each hour, the reaction vessel was cooled to RT and the mixture left to stir under aerobic conditions for 10 mins. This process was repeated 3 times, when no more starting material could be observed by ^{1}H NMR. After 3 h, the crude mixture was concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in pentane) to give **3.74c** (21.9 mg, 0.06 mmol, 29%) as a red oil.

Data for **3.74c**:

FTIR ν_{max} (neat, cm^{-1}) 2951 (w), 1721 (w), 1666 (s), 1575 (s), 1524 (w), 1468 (s), 1450 (m), 1390 (s), 1367 (m), 1294 (s), 1246 (m), 1219(s0, 1185 (m), 1158 (w), 1131 (m), 1114 (s), 1094 (s), 1058 (w), 1027 (m), 997 (s), 797 (m), 745 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.57 (1H, d, *J* = 5.0 Hz, Ar-**H**), 7.63 (1H, s, Ar-**H**), 7.61 (1H, d, *J* = 5.1 Hz, Ar-**H**), 7.54 (1H, dt, *J* = 1.0, 8.1 Hz, Ar-**H**), 7.50 (1H, dt, *J* = 1.0, 8.0 Hz, Ar-**H**), 7.30(1H, ddd, *J* = 1.1, 7.2, 8.2 Hz, Ar-**H**), 7.21 (1H, ddd, *J* = 1.0, 7.2, 8.0 Hz, Ar-**H**), 5.54 (2H, s, NC**H$_2$**O), 4.20 (3H,s, OC**H$_3$**), 3.98 (3H, s, OC**H$_3$**), 3.34 (3H, s, NCH_2OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 184.1 (**C**O), 179.7 (**C**O), 162.1 (**C**), 156.7 (**C**), 153.1 (**C**H), 142.0 (**C**), 136.2 (**C**), 131.8 (**C**H), 127.8 (**C**), 124.5 (**C**), 122.7 (**C**H), 121.7 (**C**H), 121.0 (**C**H), 113.1 (**C**H), 112.8 (**C**), 110.3 (**C**H), 105.2 (**C**), 77.9 (N**C**H_2O), 61.0 (O**C**H_3), 56.1 (NCH_2O **C**H_3), 54.9 (O**C**H_3).

HRMS (ESI+) $^m/_z$ $[MH]^+$ 379.1288 expected, detected 379.1298.

A solution of **3.75c** (61.7 mg, 0.16 mmol) in toluene (4 mL) was heated to 150 °C in a sealed vessel. After 3 h, the reaction vessel was cooled to RT and the mixture left to stir under aerobic conditions for 10 min. The crude mixture was concentrated at reduced pressure and purified by flash chromatography (0-25% ethyl acetate in pentane) to give **3.74c** first (14.7 mg, 0.04 mmol, 25%) as a red oil, followed by **3.92c** (17.7 mg, 0.05, 29%) as a yellow oil.

Data for **3.92c**:

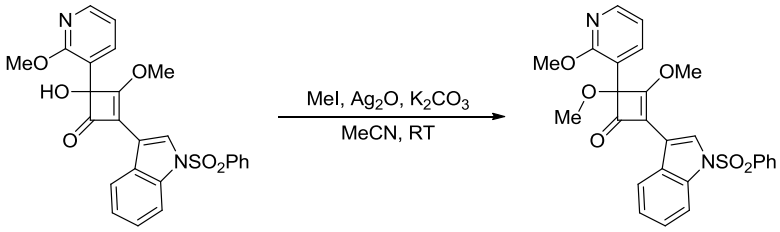
FTIR ν_{max} (neat, cm^{-1}) 2952 (w), 2927 (w), 1752 (s), 1671 (s), 1591 (m), 1540 (w), 1468 (s), 1415 (s), 1374 (m), 1328 (s), 1287 (m), 1265 (w), 1245 (w), 1181 (m), 1131 (m), 1088 (s), 1053 (s), 1017 (s), 941 (w), 909 (m), 874 (w), 859 (w), 782 (m), 745 (s), 701 (w), 649 (w).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.25 (1H, dd, *J* = 2.0, 5.0 Hz, Ar-**H**), 7.77 (1H, dt, *J* = 0.9, 8.1 Hz, Ar-**H**), 7.66 (1H, s, Ar-**H**), 7.63 (1H, dd, *J* = 1.9, 7.4 Hz, Ar-**H**), 7.52 (1H, dt, *J* = 0.7, 8.2 Hz, Ar-**H**), 7.29 (1H, ddd, *J* = 1.2, 7.2, 8.3 Hz, Ar-**H**), 7.21 (1H, ddd, *J* = 1.0, 7.2, 8.0 Hz, Ar-**H**), 6.97 (1H,dd, *J* = 5.0, 7.4 Hz, Ar-**H**), 6.27 (1H, s, C**H**), 5.50 (2H, s, NC**H_2**O), 4.06 (3H, s, OC**H_3**), 3.80 (3H, s, OC**H_3**), 3.30 (3H, s, NCH_2OC**H_3**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 172.7(**C**O), 171.8 (**C**), 162.0 (**C**), 148.3 (**C**H), 137.0 (**C**), 136.3 (**C**), 128.8 (**C**H), 128.3(**C**), 122.7 (**C**H), 121.0 (**C**H), 120.8 (**C**H), 117.2 (**C**H), 117.1 (**C**), 110.2 (**C**H), 104.6 (**C**), 99.7 (**C**), 77.7 (N**C**H_2), 73.3 (**C**H), 58.8 (O**C**H_3), 56.0 (NCH_2O**C**H_3), 53.9 (O**C**H_3).

HRMS (ESI+) $^{m}/_{z}$ $[MH]^+$ 381.1372 expected, detected 381.1447, $[MNa]^+$ 403.1270 expected, detected 403.1267.

3,4-Dimethoxy-4-(2-methoxypyridin-3-yl)-2-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-2-en-1-one (3.106a).



3.75b
$C_{25}H_{20}N_2O_6S$
(476.50)

3.106a
$C_{26}H_{22}N_2O_6S$
(490.53)

To a solution of **3.75b** (95.1 mg, 0.20 mmol) in acetonitrile (5.00 mL) was added Ag_2O (2eq, 93.7 mg) followed by K_2CO_3 (1.6 equiv., 45 mg). After 0.25 h, MeI (4 equiv., 113.6 mg, 0.05 mL) was added, and the mixture left to stir at RT. After 16 h, the solution was filtered and concentrated at reduced pressure. Purification by flash chromatography (0-50% ethyl acetate in pentane) afforded **3.106a** (63.0 mg, 0.13 mmol, 64.5%) as a yellow oil.

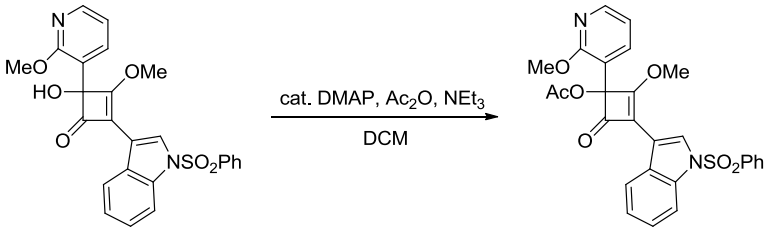
FT-IR ν_{max} (neat, cm^{-1}) 2953 (w), 1762 (m), 1647 (m), 1584 (s), 1542 (w), 1531 (w), 1466 (s), 1449 (s), 1407 (s), 1373 (s), 1304 (w), 1266 (s), 1176 (s), 1132 (s), 1108 (m), 1091 (w), 1080 (w), 1018 (m), 959 (m), 936 (s), 703 (s), 686 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.21 (1H, app d, *J* = 7.4 Hz, 1H, Ar-**H**), 8.16 (1H, dd, *J* = 1.7, 4.9 Hz, Ar-**H**), 8.07 (1H, dd, *J* = 1.7, 7.5 Hz, Ar-**H**), 8.05 (1H, s, Ar-**H**), 8.01 (1H, app. d, *J* = 8.2 Hz, Ar-**H**), 7.98 – 7.93 (2H, m, Ar-**H**), 7.59 – 7.53 (1H, m, Ar-**H**), 7.50 – 7.47 (2H, m, Ar-**H**), 7.37 (1H, ddd, *J* = 1.1, 7.4, 8.3 Hz, Ar-**H**), 7.31 (1H, m, Ar-**H**), 7.02 (1H, dd, *J* = 4.9, 7.5 Hz, Ar-**H**), 4.10 (3H, s, OC**H$_3$**), 3.84 (3H, s, OC**H$_3$**), 3.58 (3H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 184.7 (**C**O), 178.7 (**C**), 159.8 (**C**), 146.5 (**C**H), 138.3 (**C**H), 138.1 (**C**), 134.6 (**C**), 134.0 (**C**H), 129.3 (**C**H), 128.5 (**C**), 126.9 (**C**H), 125.4 (**C**H), 125.2 (**C**H), 123.7 (**C**H), 122.7 (**C**H), 122.0 (**C**), 118.9 (**C**), 117.4 (**C**H), 113.3 (**C**H), 111.1 (**C**), 95.4 (**C**), 59.9 (O**C**H_3), 54.0 (O**C**H_3), 52.7 (O**C**H_3).

HRMS (ESI+) $^m/_z$ $[MH]^+$ expected 491.1271, detected 491.1279.

2-Methoxy-1-(2-methoxypyridin-3-yl)-4-oxo-3-(1-(phenylsulfonyl)-1H-indol-3-yl)cyclobut-2-en-1-yl acetate (3.106b).



3.75b
$C_{25}H_{20}N_2O_6S$
(476.50)

3.106b
$C_{27}H_{22}N_2O_7S$
(518.54)

Modified from a procedure by Braddock et al.[60]

To a solution of **3.75b** (53.6 mg, 0.11 mmol) and DMAP (4.30 mg, 0.04 mmol) in DCM (5.00 mL) was added NEt_3 (0.04 mL, 0.29 mmol) followed by Ac_2O (0.02 mL, 0.21 mmol). After 4 h, MeOH (5 mL) was added and the resultant mixture concentrated. The crude material was dissolved in ethyl acetate (15 mL) and washed with 5% HCl solution (15 mL), followed by sat. $NaHCO_3$ until the pH was above 7. The organic phase was then dried with Na_2SO_4 and concentrated at reduced pressure to afford **3.106b** as a yellow oil (57 mg, 0.11 mmol, 99%).

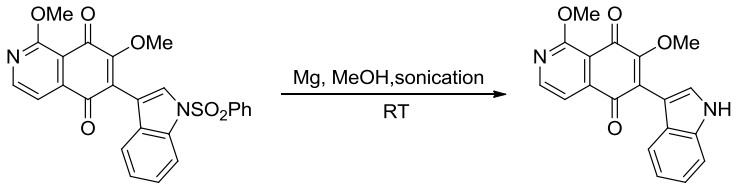
FTIR ν_{max} (neat, cm^{-1}) 2952 (w), 1751 (m), 1634 (m), 1583 (m), 1467 (s), 1448 (s), 1407, (m), 1371 (s), 1306 (w), 1260 (m), 1228 (m), 1176 (s), 1127 (s), 1103 (w), 1088 (m), 1020 (s), 750 (s), 727 (s), 687 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.18 (1H, dd, *J* = 1.8, 4.9 Hz, Ar-**H**), 8.16 – 8.12 (1H, m, Ar-**H**), 8.00 – 7.97 (1H, m, Ar-**H**), 7.99 (1H, s, Ar-**H**), 7.95 – 7.90 (2H, m, Ar-**H**), 7.82 (1H, dd, *J* = 1.9, 7.6 Hz, Ar-**H**), 7.58 – 7.52 (1H, m, Ar-**H**), 7.48 – 7.43 (2H, m, Ar-H), 7.35 (1H, ddd, *J* = 1.3, 7.4, 8.6 Hz, Ar-**H**), 7.33 – 7.29 (1H, m, Ar-**H**), 7.00 (1H, dd, *J* = 5.0, 7.6 Hz, Ar-**H**), 4.13 (s, 3H, C**H**$_3$), 3.90 (s, 3H, C**H**$_3$), 2.22 (s, 3H, C**H**$_3$).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 179.9 (**C**O), 176.0 (**C**O), 169.1 (**C**), 159.9 (**C**), 147.5 (**C**H), 138.0 (**C**), 137.2 (**C**H), 134.5 (**C**), 134.0 (**C**H), 129.4 (**C**H), 128.6 (**C**), 126.9 (**C**H), 125.6 (**C**H), 125.2 (**C**H), 123.7 (**C**H), 122.6 (**C**H), 122.0 (**C**), 117.2 (**C**H), 116.9 (**C**), 113.3 (**C**H), 111.0 (**C**), 93.9 (**C**), 60.5 (**C**H$_3$), 53.8 (**C**H$_3$), 21.5 (**C**H$_3$).

HRMS (ESI+) $^{m}I_{z}$ $[MH]^{+}$ expected 519.122, detected 519.1229.

6-(1H-Indol-3-yl)-1,7-dimethoxyisoquinoline-5,8-dione (3.110).



3.74b
$C_{25}H_{18}N_2O_6S$
(474.49)

3.110
$C_{19}H_{14}N_2O_4$
(334.33)

Following a procedure established by Nyasse et al.:[42]

To a mixture of **3.74b** (12.7 mg, 0.03 mmol) and Mg powder (14 mg, 0.58 mmol) was added dry MeOH (1.00 mL) and left to sonicate at RT. After 1h, the reaction was diluted with DCM (15 mL) and HCl (6 mL, 1M) added. The aqueous phase was separated and extracted with DCM (3 x 15 mL). The combined organic phases were rinsed with sat. $NaHCO_3$ (15 mL) followed by brine (15 mL), dried over Na_2SO_4 and concentrated at reduced pressure. Purification by preparative thin layer chromatography (TLC) (50% ethyl acetate in pentane) afforded **3.110** (7 mg, 0.02 mmol, 70%) as a purple oil.

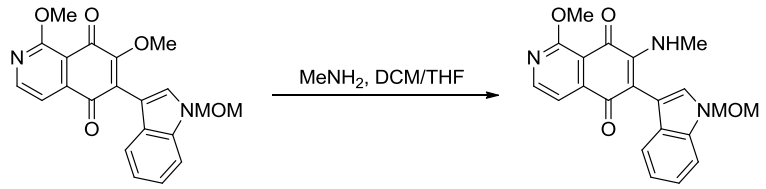
FTIR ν_{max} (neat, cm^{-1}) 3359 (br. w), 2924 (s), 2852 (m), 1738 (w), 1663 (s), 1597 (m), 1576 (m), 1518 (w), 1451 (s), 1392 (s), 1360 (w), 1292 (s), 1242 (m), 1210 (m), 1128 (w), 1098 (m), 1062 (w), 1009 (s), 932 (w), 859 (w), 789 (w), 745 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.67 (1H, br. s, N**H**), 8.57 (1H, d, *J* = 5.0, Ar-**H**), 7.63 (1H, d, *J* = 2.8 Hz, Ar-**H**),7.62 (1H, d, *J* = 5.0 Hz, Ar-**H**), 7.52 - 7.50 (1H, m, Ar-H), 7.43 (1H, dt, *J* = 1.4, 8.3 Hz, Ar-H), 7.25 (1H, ddd, *J* = 1.1, 7.2, 8.0 Hz, Ar-**H**), 7.18 (1H, ddd, *J* = 1.0, 7.0, 8.1 Hz, Ar-**H**), 4.20 (3H, s, OC**H$_3$**), 3.95 (3H, s, OC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 184.3 (**C**O), 179.7 (**C**O), 162.1 (**C**), 156.7 (**C**), 153.0 (**C**H), 142.1 (**C**), 135.7 (**C**), 128.1 (**C**H), 126.5 (**C**), 124.9 (**C**), 122.5 (**C**H), 121.4 (**C**H), 120.6 (**C**H), 113.2 (**C**H), 112.8 (**C**), 111.4 (**C**H), 105.4 (**C**), 61.0 (O**C**H_3), 54.9 (O**C**H_3)

HRMS (ESI+) $^{m}I_{z}$ $[MH]^{+}$ 335.1026 expected, detected 335.1029.

1-Methoxy-6-(1-(methoxymethyl)-1H-indol-3-yl)-7-(methylamino)isoquinoline-5,8-dione (3.111).



3.74c
$C_{21}H_{18}N_2O$
(378.38)

3.111
$C_{21}H_{19}N_3O_4$
(377.4)

Following a procedure established by Wang et al.: [61]

To a solution of **3.74c** (20.7 mg, 0.06 mmol) in DCM (1.50 mL) was added methylamine (2M in THF, 2.4 mL, 4.8 mmol) and the reaction left to stir at room temperature. After 24 h, the resultant mixture was concentrated at reduced pressure and purified by flash chromatography (neat $CHCl_3$) to give **3.111** (18.5 mg, 0.05 mmol, 90%) as a red oily solid.

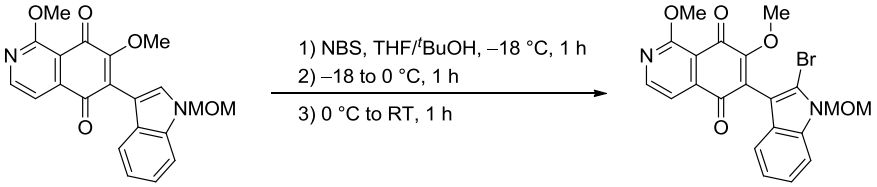
FTIR ν_{max} (neat, cm^{-1}) 3318 (m), 2929 (w), 1679 (s), 1599 (m), 1567 (s), 1538 (s), 1522 (w), 1454 (m), 1422 (w), 1393 (m), 1365 (m), 1340 (w), 1317 (w), 1290 (m), 1246 (s), 1212 (w), 1181 (w), 1151 (w), 1125 (w), 1094 (m), 1054 (w), 974 (m), 916 (w), 868 (w), 794 (w), 747 (s).

^{1}H NMR (400 MHz, $CDCl_3$) δ_H ppm 8.55 (1H, d, *J* = 5.0, Ar-**H**), 7.64 (1H, d, *J* = 5.1 Hz, Ar-**H**), 7.52 (1H, app. d, *J* = 8.2 Hz, Ar-**H**), 7.39 (1H, app. d, *J* = 1.0, 8.0 Hz, Ar-**H**), 7.33 (1H, s, Ar-**H**), 7.30 - 7.23 (1H, m, Ar-**H**), 7.19 - 7.12 (1H, m, Ar-**H**), 6.29 (1H, br. s, N**H**), 5.52 (2H, s, NC**H$_2$**O), 4.19 (3H, s, OC**H$_3$**), 3.30 (3H, s, NCH_2OC**H$_3$**), 2.53 (3H, d, *J* = 5.7 Hz, NHC**H$_3$**).

^{13}C NMR (101 MHz, $CDCl_3$) δ_C ppm 180.5 (**C**O), 180.4 (**C**O), 162.1 (**C**), 154.0 (**C**H), 147.4 (**C**), 144.0 (**C**), 135.8 (**C**), 130.1 (**C**), 129.6 (**C**H), 122.5 (**C**H), 120.8 (**C**H), 120.0 (**C**H), 113.8 (**C**H), 111.9 (**C**), 110.1 (**C**H) , 107.9 (**C**), 107.0 (**C**), 77.6 (N**C**H_2O), 55.9 (NCH_2OCH_3), 54.9 (O**C**H_3), 31.7 (N**C**H_3).

HRMS (ESI+) $^m/_z$ $[MH]^+$ 378.1448 expected, detected 378.1436, $[MNa]^+$ 400.1268 expected, detected 400.1256.

6-(2-Bromo-1-(methoxymethyl)-1H-indol-3-yl)-1,7-dimethoxyisoquinoline-5,8-dione (3.114).



3.74c
$C_{21}H_{18}N_2O_5$
(378.38)

3.114
$C_{21}H_{17}BrN_2O_5$
(457.28)

Modified from a procedure established by Wanner et al.:[43]

To a solution of **3.74c** (20.0 mg, 0.05 mmol) in THF/^{t}BuOH (1.00 mL, 1 : 1 mixture) at −18 ºC was added NBS (10.9 mg, 0.06) portionwise. After 1 h, the reaction was warmed to 0 ºC. After 1 h, the reaction was warmed to RT. The resultant mixture was concentrated at reduced pressure and purified by preparative TLC (50% ethyl acetate in pentane) to give **3.114** (14.5 mg, 0.03 mmol, 60%) as an orange solid.

FT-IR ν_{max} (neat, cm^{-1}) 2925 (s), 2854 (m), 1736 (w), 1673 (m), 1574 (m), 1525 (w), 1468 (s), 1453 (s), 1391 (s), 1357 (m), 1295 (s), 1242 (w), 1219 (m), 1157 (w), 1098 (s), 1058 (m), 995 (m), 914 (w), 815 (w), 744 (m).

^{1}H NMR (400 MHz, $(CD_3)_2CO$) δ_H 8.64 (1H, d, *J* = 5.0 Hz, Ar-**H**), 7.64 – 7.62 (1H, m, Ar-**H**), 7.56 (1H, d, *J* = 5.1 Hz, Ar-**H**), 7.40 – 7.38 (1H, m, Ar-**H**), 7.26 (1H, ddd, *J* = 1.2, 7.2, 8.3 Hz, Ar-**H**), 7.12 (1H, ddd, *J* = 1.0, 7.3, 8.0 Hz, Ar-**H**), 5.70 (2H, s, NC**H$_2$**O), 4.11 (3H, s, OC**H$_3$**), 3.93 (3H, s, OC**H$_3$**), 3.34 (3H, s, NCH_2OCH_3).

^{13}C NMR (101 MHz, $(CD_3)_2CO$) δ_C ppm 183.1 (**C**O), 179.6 (**C**O), 163.1 (**C**), 161.1(**C**), 154.2 (**C**H), 143.0 (**C**), 138.0 (**C**), 129.3 (**C**), 123.6 (**C**H), 122.2 (**C**), 122.0 (**C**H), 120.5 (**C**H), 116.3 (**C**), 114.2 (**C**), 113.7 (**C**H), 111.4 (**C**H), 109.5 (**C**), 76.2 (N**C**H_2O), 61.4 (O**C**H_3), 56.3 (N**C**H_2O**C**H_3), 55.0 (O**C**H_3).

LRMS (ESI+) $^m/_z$: 457 ([M^{79}BrH]$^+$, 18%), 459 ([M^{81}BrH]$^+$, 28%), 122 (100%).

HRMS (ESI+) $^m/_z$ [M^{79}BrH]$^+$ expected 457.0394, detected 457.0402.

Chapter 5: List of References

1. Moore, H. Y., Benjamin, *Chemtr:- Org. Chem.* **1992**, *5*, 273 - 313.

2. Pettit, G. R.; Collins, J. C.; Knight, J. C.; Herald, D. L.; Nieman, R. A.; Williams, M. D.; Pettit, R. K., *J. Nat. Prod.* **2003**, *66*, 544-547.

3. Mohamed, M.; Goncalves, T. P.; Whitby, R. J.; Sneddon, H. F.; Harrowven, D. C., *Chem.-Eur. J.* **2011**, *17*, 13698-13705.

4. (a) Moore, H. W.; Perri, S. T., *J. Org. Chem.* **1988**, *53*, 996-1003; (b) Foland, L. D.; Karlsson, J. O.; Perri, S. T.; Schwabe, R.; Xu, S. L.; Patil, S.; Moore, H. W., *J. Am. Chem. Soc.* **1989**, *111*, 975-989; (c) Perri, S. T.; Moore, H. W., *J. Am. Chem. Soc.* **1990**, *112*, 1897-1905; (d) Winters, M. P.; Stranberg, M.; Moore, H. W., *J. Org. Chem.* **1994**, *59*, 7572-7574; (e) Oppermann, G.; Stranberg, M.; Moore, H. W.; Schaumann, E.; Adiwidjaja, G., *Synthesis* **2010**, 2027-2038; (f) Liebeskind, L. S.; Iyer, S.; Jewell, C. F., Jr., *J. Org. Chem.* **1986**, *51*, 3065-3067; (g) Sun, L.; Liebeskind, L. S., *J. Org. Chem.* **1994**, *59*, 6856-6858; (h) Liu, F.; Liebeskind, L. S., *J. Org. Chem.* **1998**, *63*, 2835-2844; (i) Zhang, S.; Liebeskind, L. S., *J. Org. Chem.* **1999**, *64*, 4042-4049; (j) Mingo, P.; Zhang, S.; Liebeskind, L. S., *J. Org. Chem.* **1999**, *64*, 2145-2148; (k) Harrowven, D. C.; Pascoe, D. D.; Guy, I. L., *Angew. Chem., Int. Ed.* **2007**, *46*, 425-428; (l) Harrowven, D. C.; Mohamed, M.; Goncalves, T. P.; Whitby, R. J.; Bolien, D.; Sneddon, H. F., *Angew. Chem., Int. Ed.* **2012**, *51*, 4405-4408; (m) Goncalves, T. P.; Mohamed, M.; Whitby, R. J.; Sneddon, H. F.; Harrowven, D. C., *Angew. Chem., Int. Ed.* **2015**, *54*, 4531-4534.

5. Liu, H.; Tomooka, C. S.; Moore, H. W., *Synth. Commun.* **1997**, *27*, 2177-2180.

6. (a) Packard, E.; Pascoe, D. D.; Maddaluno, J.; Goncalves, T. P.; Harrowven, D. C., *Angew. Chem., Int. Ed.* **2013**, *52*, 13076-13079; (b) Enhsen, A.; Karabelas, K.; Heerding, J. M.; Moore, H. W., *J. Org. Chem.* **1990**, *55*, 1177-1185; (c) Schaumann, E.; Oppermann, G.; Stranberg, M.; Moore, H. W., *Aust. J. Chem.* **2010**, *63*, 1656-1664.

7. Schaumann, E.; Oppermann, G.; Stranberg, M.; Moore, H.; Adiwidjaja, G., *Synthesis* **2010**, 2027-2038.

8. Trost, B. M.; Thiel, O. R.; Tsui, H.-C., *J. Am. Chem. Soc.* **2003**, *125*, 13155-13164.

9. (a) Perri, S. T.; Foland, L. D.; Moore, H. W., *Tetrahedron Lett.* **1988**, *29*, 3529-32; (b) Liebeskind, L. S., *Tetrahedron* **1989**, *45*, 3053-3060; (c) Liebeskind, L. S.; Fengl, R. W.; Wirtz, K. R.; Shawe, T. T., *J. Org. Chem.* **1988**, *53*, 2482-2488.

10. (a) Lee, K. H.; Moore, H. W., *Tetrahedron Lett.* **1993,** *34* (2), 235-8; (b) Liebeskind, L. S.; Granberg, K. L.; Zhang, J., *J. Org. Chem.* **1992,** *57* (16), 4345-52.

11. Kirmse, W.; Rondan, N. G.; Houk, K. N., *J. Am. Chem. Soc.* **1984**, *106*, 7989-7991.

12. Musch, P. W.; Remenyi, C.; Helten, H.; Engels, B., *J. Am. Chem. Soc.* **2002**, *124*, 1823-1828.

13. Iyer, S.; Liebeskind, L. S., *J. Am. Chem. Soc.* **1987**, *109*, 2759-2770.

14. Pettit, G. R.; Knight, J. C.; Collins, J. C.; Herald, D. L.; Pettit, R. K.; Boyd, M. R.; Young, V. G., *J. Nat. Prod.* **2000**, *63*, 793-798.

15. Pettit, R. K.; Fakoury, B. R.; Knight, J. C.; Weber, C. A.; Pettit, G. R.; Cage, G. D.; Pon, S., *J. Med. Microbiol.* **2004**, *53*, 61-65.

16. (a) Nakahara, S.; Kubo, A., *Heterocycles* **2004**, *63*, 2355-2362; (b) Nakahara, S.; Kubo, A.; Mikami, Y.; Ito, J., *Heterocycles* **2006**, *68*, 515-520.

17. Nakahara, S.; Numata, R.; Tanaka, Y.; Kubo, A., *Heterocycles* **1995**, *41*, 651-654.

18. Markey, M. D.; Kelly, T. R., *J. Org. Chem.* **2008**, *73*, 7441-7443.

19. Knueppel, D.; Martin, S. F., *Angew. Chem., Int. Ed.* **2009**, *48*, 2569-2571.

20. Hoyt, M. T.; Palchaudhuri, R.; Hergenrother, P. J., *Invest. New Drugs* **2011**, *29*, 562-573.

21. Hoyt, M. T., PhD Thesis, University of Illinois Urbana-Champaign, 2010. Available at: https://www.ideals.illinois.edu/bitstream/handle/2142/15525/Hoyt_Mirth.pdf?sequence=1. [Accessed 10 June 2012]

22. Knueppel, D.; Martin, S. F., *Tetrahedron* **2011**, *67*, 9765-9770.

23. Nakahara, S.; Kubo, A., *Heterocycles* **2003**, *60*, 2717-2725.

24. M. Hudlicky, *Reductions in Organic Chemistry*, 1st Edition, Ellis Horwood Limited, Chichester, 1984; p 309.

25. Reed, M. W.; Pollart, D. J.; Perri, S. T.; Foland, L. D.; Moore, H. W., *J. Org. Chem.* **1988**, *53*, 2477-2482.

26. (a) Corey, E. J.; Beames, D. J., *J. Amer. Chem. Soc.* **1972**, *94*, 7210-7211; (b) Seebach, D.; Neumann, H., *Chem. Ber.* **1974**, *107*, 847-853.

27. Valderrama, J. A.; Ibacache, A.; Rodriguez, J. A.; Theoduloz, C.; Benites, J., *Eur. J. Med. Chem.* **2011**, *46*, 3398-3409.

28. Thale, Z.; Johnson, T.; Tenney, K.; Wenzel, P. J.; Lobkovsky, E.; Clardy, J.; Media, J.; Pietraszkiewicz, H.; Valeriote, F. A.; Crews, P., *J. Org. Chem.* **2002**, *67*, 9384-9391.

29. Kitahara, Y.; Mizuno, T.; Kubo, A., *Tetrahedron* **2004**, *60*, 4283-4288.

30. Buccini, M.; Jeow, S. Y.; Byrne, L.; Skelton, B. W.; Nguyen, T. M.; Chai, C. L. L.; Piggott, M. J., *Eur. J. Org. Chem.* **2013**, 3232-3240.

31. Komori, T.; Yokoshima, S.; Fukuyama, T., *Synlett* **2015,** *26*, 1537-1540.

32. Zhang, D.; Llorente, I.; Liebeskind, L. S., *J. Org. Chem.* **1997**, *62*, 4330-4338.

33. (a) Hilbert, G. E.; Johnson, T. B., *J. Am. Chem. Soc.* **1930**, *52*, 4489-4494; (b) Iida, H.; Suda, M.; Nakajima, E.; Hakamatsuka, H.; Nagashima, Y.; Joho, K.; Amemiya, K.; Moromizato, T.; Matsumoto, K.; Murakami, Y.; Hamana, H., *Heterocycles* **2010**, *81*, 2057-2062.

34. Amat, M.; Hadida, S.; Sathyanarayana, S.; Bosch, J., *Org. Synth.* **1997**, *74*, 248.

35. Amat, M.; Hadida, S.; Sathyanarayana, S.; Bosch, J., *J. Org. Chem.* **1994**, *59*, 10-11.

36. Gai, S.; Zhang, Q.; Hu, X., *J. Org. Chem.* **2014**, *79*, 2111-2114.

37. Comins, D. L.; LaMunyon, D. H., *Tetrahedron Lett.* **1988**, *29*, 773-776.

38. Trecourt, F.; Mallet, M.; Marsais, F.; Quéguiner, G., *J. Org. Chem.* **1988**, *53*, 1367-1371.

39. Testaferri, L.; Tiecco, M.; Tingoli, M.; Bartoli, D.; Massoli, A., *Tetrahedron* **1985**, *41*, 1373-1384.

40. Trost, B. M.; Xie, J.; Sieber, J. D., *J. Am. Chem. Soc.* **2011**, *133*, 20611-20622.

41. (a) Snell, R. H.; Woodward, R. L.; Willis, M. C., *Angew. Chem., Int. Ed.* **2011**, *50*, 9116-9119; (b) Snell, R. H.; Durbin, M. J.; Woodward, R. L.; Willis, M. C., *Chem. - Eur. J.* **2012**, *18*, 16754-16764.

42. Nyasse, B.; Grehn, L.; Ragnarsson, U., *Chem. Commun.* **1997**, 1017-1018.

43. Wanner, M. J.; Ingemann, S.; van Maarseveen, J. H.; Hiemstra, H., *Eur. J. Org. Chem.* **2013**, 1100-1106.

44. Neo, A. G.; Lopez, C.; Lopez, A.; Castedo, L.; Tojo, G., *Tetrahedron* **2013**, *69*, 11010-11016.

45. Bhat, V.; MacKay, J. A.; Rawal, V. H., *Org. Lett.* **2011**, *13*, 3214-3217.

46. Liu, Y.; Zhang, L.; Jia, Y., *Tetrahedron Lett.* **2012**, *53*, 684-687.

47. Bayer Aktengesellschaft, US Pat. US5354749 A1, 1994, p16.

48. Rong, D.; Phillips, V. A.; Rubio, R. S.; Ángeles Castro, M.; Wheelhouse, R. T., *Tetrahedron Lett.* **2008**, *49*, 6933-6935.

49. (a) Song, J. J.; Yee, N. K.; Tan, Z.; Xu, J.; Kapadia, S. R.; Senanayake, C. H., *Org. Lett.* **2004**, *6*, 4905-4907; (b) Ismail, M. A.; Brun, R.; Easterbrook, J. D.; Tanious, F. A.; Wilson, W. D.; Boykin, D. W., *J. Med. Chem.* **2003**, *46*, 4761-4769.

50. Friel, D. K.; Snapper, M. L.; Hoveyda, A. H., *J. Am. Chem. Soc.* **2008**, *130*, 9942-9951.

51. Verniest, G.; Colpaert, J.; Toernroos, K. W.; De Kimpe, N., *J. Org. Chem.* **2005**, *70*, 4549-4552.

52. Dehmlow, E. V. S., Hans G., *Chem. Ber.* **1980**, *113*, 1-8.

53. Ivanovsky, S. A.; Dorogov, M. V.; Kravchenko, D. V.; Ivachtchenko, A. V., *Synth. Commun.* **2007**, *37*, 2527-2542.

54. Thorpe, J., *J. Chem. Soc. B* **1968**, 435-436.

55. Roth, H. J.; Sporleder, H., *Arch. Pharm.* **1970**, *303*, 886-95.

56. Katoh, I.; Aoki, T.,; Suzuki, H.; Utsunomiya, I.; Kuroda, N.; Iwaki, T., US Pat. US2011/98471 A1, 2011, p57.

57. (a) Suzuki, H.; Utsunomiya, I.; Shudo, K.; Fujimura, T.; Tsuji, M.; Kato, I.; Aoki, T.; Ino, A.; Iwaki, T., *ACS Med. Chem. Lett.* **2013**, *4*, 1074-1078; (b) Enguehard, C.; Hervet, M.; Thery, I.; Renou, J.-L.; Fauvelle, F.; Gueiffier, A., *Helv. Chim. Acta* **2001**, *84*, 3610-3615.

58. Potavathri, S.; Pereira, K. C.; Gorelsky, S. I.; Pike, A.; LeBris, A. P.; DeBoef, B., *J. Am. Chem. Soc.* **2010**, *132*, 14676-14681.

59. Nirogi, R.; Dwarampudi, A.; Bhatta, V.; Kota, L.; Dubey, P. K., *Asian J. Chem.* **2013**, *25*, 9293-9298.

60. Braddock, D. C.; Millan, D. S.; Perez-Fuertes, Y.; Pouwer, R. H.; Sheppard, R. N.; Solanki, S.; White, A. J. P., *J. Org. Chem.* **2009**, *74*, 1835-1841.

61. Wang, S.; Kohn, H., *J. Org. Chem.* **1997**, *62*, 5404-5412.