Synthetic studies directed toward azaspirocyclic alkaloids using spironitrone intermediates

Michael Foley

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Michael Foley

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Abstract

HTX **1** is a 6,6-azaspirocyclic alkaloid found to naturally accumulate in the skin of certain frog species and has shown to exhibit activity as a non-competitive inhibitor of neuromuscular and central neuronal nicotinic acetylcholine receptors. A structurally similar, 6,5-azaspirocyclic system, is seen in the marine anti-inflammatory alkaloids pinnaic acid **2** and halichlorine **3**. Despite both halichlorine and pinnaic acid having different mechanisms of action, both exhibit anti-inflammatory properties of interest to the academic community. It is due to these properties of interest and the scarcity of this natural product analogue's availability from their natural sources that have led to synthetic studies being undertaken.

This study aimed to develop a common synthetic route to the core structures of all three natural products. The use of this approach towards a formal/total synthesis of all three and libraries of analogues was also investigated. The strategy centred on the use of a 6,5 or 6,6-spitonitrones **105** and **232** respectively. It was planned to access these nitrones by oxidative ring opening of the corresponding isoxazolidines **22** and **23**. While the preparation of multigram quantities of nitrone **105** was achieved a similar oxidation of isoxazolidine **23** was unsuccessful. Nevertheless, isoxazolidine **23** was used to access 6,6-azaspirocycles such as **222** and **223**. A similar library of 6,5-azaspirocycles was also accessed from the corresponding isoxazolidine **22**.

Nitrone **105** was used to access the core structures of both pinnaic acid **2** and halichlorine **3** via preparation of the allylated spirocycle **161** accessed via Grignard addition to nitrone **105**. Furthermore, addition of a range of Grignard reagents to this nitrone was used to prepare a small library of pinnaic acid analogues. The core structure of pinnaic acid **2** was prepared from allyl-derivative **170** via oxidative cleavage and subsequent Wittig homologation. The quinolizidine core structure **181** of halichlorine was accessed via intramolecular RCM from diene **180** - also prepared from key allylated spirocycle **170**. This thesis also investigated the further functionalisation of spirocyclic core **195** towards a formal synthesis of pinnaic acid. While homologation at C11 was achieved the formation of a rigid tricycle **192,** to allow stereoselective introduction of the C12 methyl group of pinnaic acid was unsuccessful.

Acknowledgements

First and most of all, I would like to thank my director of studies Dr. Vittorio Caprio, for his continued support and encouragement – and also for putting his faith in me all those years ago when I first started out. Without his expertise, guidance, and patience none of this would have been possible. I hope this thesis has done him proud.

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Abbreviations

3-D 3-Dimensional

9-BBN 9-Borabicyclo[3.3.1]nonane

Ac Acetyl

Aq. Aqueous

Bn Benzyl

BMT 2,6-di-tert-butyl-4-methylphenol

BOC *tert*-Butyloxycarbonyl

CITES Convention on International Trade in Endangered Species

cPLA2 Cytosolic phospholipase A2

Cat. Catalyst

COSY Correlated Spectroscopy

DBU 1,8-Diazabicyclo[5.4.0]undec-7-ene

DCC *N, N'*-Dicyclohexylcarbodiimide

DIBAL Diisobutylaluminium hydride

DMAP 4-Dimethylaminopyridine

DMF Dimethylformamide

DMSO Dimethyl sulfoxide

DNA Deoxyribonucleic acid

equiv. Equivalent

FBDD Fragment based drug discovery

FMO Frontier molecular orbital

FT-IR Fourier transform infrared spectroscopy

h Hours

H Hydrogen

HMQC Heteronuclear multiple quantum coherence

HOMO Highest occupied molecular orbital

HTX Histrionicotoxin

ICAM-1 Intracellular adhesion molecule-1

LDA Lithium diisopropylamide

LiDBB 4,4'-Di-*tert*-butylbiphenylide

LiHMDS Lithium bis(trimethylsilyl)amide

LUMO Lowest unoccupied molecular orbital

mCPBA *meta*-Chloroperoxybenzoic acid

Me Methyl

MeOH Methanol

MHz Megahertz

MW Microwave

Nf-kβ Nuclear factor kappa beta

NMO *N*-Methylmorpholine *N*-oxide

NMR Nuclear magnetic resonance

NOESY Nuclear overhauser effect spectroscopy

Ph Phenyl

ppm Parts per million

PPTS Pyridinium *p*-toluenesulfonate

quant. Quantitative

RCM Ring closing metathesis

r.t. Room temperature

sat. Saturated

TBDMS tert-Butyldimethylsilyl

TBDPS *tert*-Butyl(chloro)diphenylsilyl

TEA Triethylamine

Temp. Temperature

TCA Trichloroacetic acid

TFA Trifluoroacetyl group

TFAA Trifluoroacetic anhydride

THF Tetrahydrofuran

TLC Thin layer chromatography

TMSCl Trimethylsilyl chloride

TPAP Tetrapropylammonium perruthenate

TREAT•HF Triethylamine trihydrofluoride

Ts Tosylate

VCAM-1 Vascular cell adhesion molecule-1

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1. Introduction

1.1. Medicinal chemistry of azaspirocycles

Azaspirocycles are a key motif in a range of alkaloids with diverse biological activity. An azaspirocycle consists of two ring systems connected *via* a single carbon, while incorporating a nitrogen atom. Spirocycles have a complex, sp^3-rich defined 3-D geometry. Sp^3-rich, 3-D structures have been identified as scaffolds of interest as they occupy unexplored regions of chemical space which do not infringe on areas already considered heavily explored intellectual property.[1] Compounds of this nature have shown increases in clinical success rate, which is attributed to the greater occupation of target space.[2, 3] While studies looking to exploit the sp^3-rich nature of spirocycles have taken place,[4, 5] these are currently narrow in scope.[5]

These scaffolds' spirocyclic structural features, if bearing functional groups, also provides access to more complex 3-D structures, which bind more readily to biological targets.[2, 6] Spirocycles are important scaffolds in bioactive natural products and FDA-approved drugs including spironolactone[7] and irbesartan[8] (Figure 1).

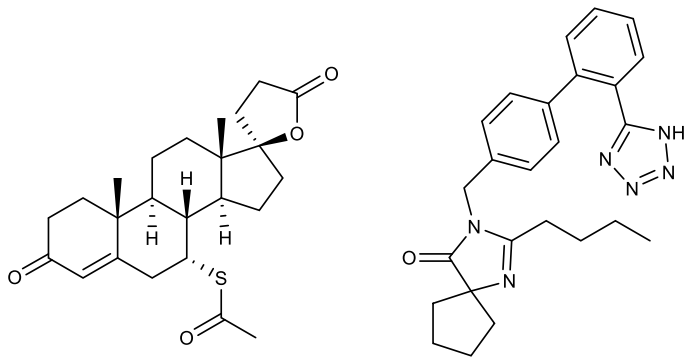


Figure 1:- FDA-approved spirocycle scaffold-containing drug compounds

Spirocycles with functionaliseable exit vectors have also been identified as useful fragments in fragment-based drug discovery (FBDD), a drug discovery screening technique utilising smaller fragments (MW of 150-250) in order to investigate potential leads.[9]

The azaspiro[4.5]decane spirocyclic ring system (), and other such similar motifs, has been of particular interest to synthetic chemists in recent years.[10]

With regards to naming - the prefix spiro is used in front of the name denoting the number of carbons. Thus, the scaffold below (Figure 2) is a spirodecane. Brackets incorporating the number of carbons in each ring is inserted between the spiro and alkane name with the numbers in order of the smallest ring. Each ring is numbered from the spiro atom with numbering starting on the smaller ring. Finally, an azo prefix is included to denote the fact that one ring incorporates a nitrogen atom. Thus, the scaffold below is a 6-azaspiro[4,5]decane

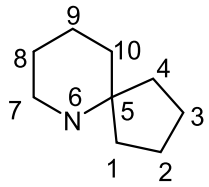


Figure 2:-Key azaspiro[4.5]decane spirocyclic ring system, a core facet of different natural products of synthetic interest.

Synthesising these motifs are not without challenges. The production of the quaternary, spiro-carbon is sterically challenging and this is formed along with the creation of one or two rings. Spirocyclisation introduces a new chiral centre and, as invariably there will be substitution patterns on the spiro scaffold, control of relative, and often, absolute stereochemistry is required to avoid the formation of complex product mixtures necessitating the development of a stereoselective synthesis.[11]

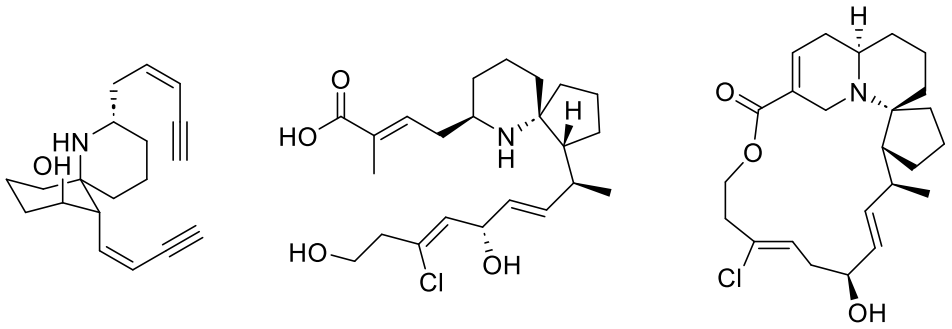
Different approaches have been developed by different research groups regarding the formation of the ring atoms. Broadly, the approaches deviate on whether the the spiro scaffold is formed in one step, via formation of two rings, simultaneously from an acyclic precursors or formation is step-wise by functionalisation of a preformed ring [10] Different examples of these different approaches are covered later in this thesis.

1.2. Natural Products

Histrionicotoxin (HTX) **1** (Figure 3), is a spirocyclic alkaloid with two *cis*-enyne side chains and is part of a group of 6,6-azaspirocyclic alkaloids found in certain frog species throughout South America. This group of alkaloids act as non-competitive inhibitors of neuromuscular and central neuronal nicotinic acetylcholine receptors.[12] As such, HTX **1** is seen as a valuable pharmacological probe molecule. These neurophysiological properties, and the low natural abundance of HTX (no more than 180 μg per frog is usually obtained per extraction)[13,14] has led to the development of many synthetic studies towards this molecule.[13] It has been hypothesised that HTX-based compounds accumulate on the surface of the skin of frog species and are obtained from particular ants and mites in its natural diet. This diet cannot be replicated in captivity[15] and extracting HTX alkaloids is no longer considered an environmentally sustainable option, underlined by the protection of certain species under the Convention on International Trade in Endangered Species (CITES) agreement.[13] Thus, detailed studies of this alkaloids requires the development of a flexible and efficient laboratory-based synthetic route.

A related 6,5-azaspirocyclic system is seen in the marine anti-inflammatory alkaloids pinnaic acid **2** and halichlorine **3**. (Figure 3). In similar fashion to histrionicotoxin, only minute quantities are available naturally, with just 200 mg of crystalline halichlorine extracted from 200 kg of the black marine sponge *Halichondria okadai.*[16] Similarly, just 1 mg of pinnaic acid was extracted from 10 kg of the Okinawan bivalve, *Pinna muricata*.[17]

While halichlorine and pinnaic acid exhibiting different mechanisms of action, both display a significant structural similarity-sharing an azaspiro[4.5]decane core with identical stereochemistry. Although it is thought that they bind to different biological targets both exhibit interesting anti-inflammatory properties, and have been investigated as potential non-steroidal anti-inflammatory drugs (NSAIDs).[18]



Histrionicotoxin (HTX) **Pinnaic Acid** **Halichlorine**

Figure 3:- Key azaspirocyclic natural products

1.3. Pharmacology of natural products

1.3.1. HTX-mode of action

The HTX **1** family of alkaloids act as non-competitive inhibitors of neuromuscular, ganglionic and central neuronal nicotinic acetylcholine receptors (nAChR) and as a result, have been utilised in neurophysiological research.[19] Non-competitive antagonists of this variety block cholinergic nerve impulse transmission *via* direct interaction with the receptor's ion channel or gating mechanism.[20] The interaction between HTX, as the non-competitive inhibitor, and the relevant active site that recognises such actions of the non-competitive antagonist, leads to inhibition of Na^+ channel influx of charged ions into pheochromocytoma (PC12) cells. These PC12 cells are a widely utilised cell culture model group for ganglionic nicotinic acetylcholine receptor-channel investigations as well as other Ca^{2+} ion channel investigations in general.[21, 22] In addition to this, HTX's have been found to interact with both sodium and potassium-gated ion channels and reduce conductance across the channel.[23] The interaction between HTX and the channel prevents charge conductance rendering the channel inactive.[24]

The precise mechanism of action between the non-competitive antagonist and the nAChR is currently unknown, however ultraviolet light-induced covalent labelling, which utilises the photoaffinity labelling of a specific site on a molecule with a ligand with a photolabile group, allows identification of any irradiated sites once deprived of all but 5 % of any light source. This allows identification of the active site, which is believed to be located directly in the ion channel at the centre of the nAChR.[25]

1.3.2. Pinnaic acid and halichlorine biological activity

Pinnaic acid **2** displays activity against expressed cytosolic phospholipase A2 (cPLA2).[18] This enzyme is a Ca^{2+} -sensitive enzyme which catalyses the cleavage of arachidonic acid from membrane phospholipids for conversion into pro-inflammatory prostaglandins.[17] The subsequent prostaglandins act to regulate cell inflammation. This inhibitory mechanism has led to pinnaic acid being identified as a potential NSAID.[26]

It has been found that cPLA2 becomes active in the presence of 0.2-20 μM of Ca^{2+} ions.[26] Studies investigating the effectiveness of pinnaic acid as an anti-inflammatory agent reveal an IC_{50} value of 0.2 mM.[27]

Halichlorine **3** has shown expressed activity against the formation of vascular cell adhesion molecule (VCAM) 1, a peptide involved in the procurement and transfer of leukocytes to the sites of tissue trauma. [18] As such, halichlorine has been postulated as a potential therapeutic treatment for a number of diseases such as atherosclerosis, coronary artery diseases, angina, and cardiovascular inflammatory diseases caused by extreme inflammatory responses.[28]

Specifically, halichlorine inhibits lipopolysaccharide-induced nuclear factor-kappa beta (NF-κb), a key transcription factor in the regulation of pro-inflammation expression in cells such as VCAM-1 and also intercellular adhesion molecule-1 (ICAM-1).[29] As a result, many research groups have investigated the synthesis of halichlorine due to its anti-inflammatory properties.[30]

While it appears that the 6,6- and 6,5-azaspirocyclic motifs exhibit quite different biological activities with the former exhibiting nicotinic action and the latter displaying anti-inflammatory properties, it has been reported that simple 6,5-azaspirocyclic systems do show nicotinic activity.[31] The basic 6,5-azaspirocyclic scaffolds **4-7** (Figure 4), show promising results in blocking nAChR activity by direct interaction with the Na^{+} ion channel. The activity of such simple azaspirocycles has received little attention but warrants further investigation especially given the structural similarity of pinnaic acid/halichlorine and histrionicotoxin and the discovery of their biological effects. These results indicate that simplified analogues of pinnaic acid/halichlorine (and even the natural products themselves) may also

exhibit nicotinic properties and that there may be some undiscovered similarities in bioactivity between pinnaic acid/halichlorine and the histrionicotoxins.

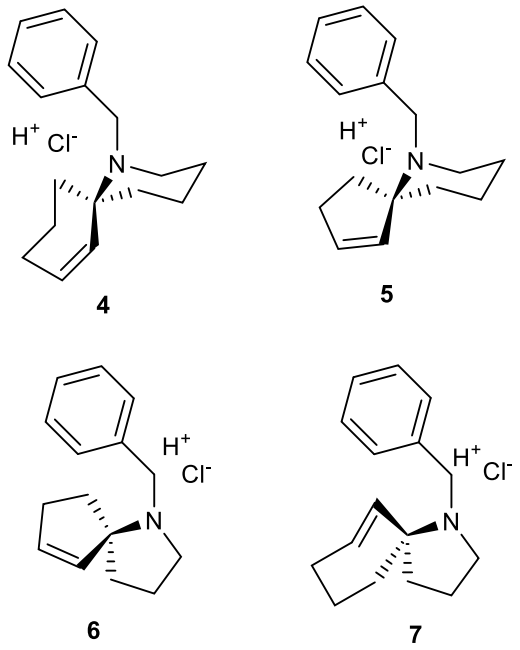
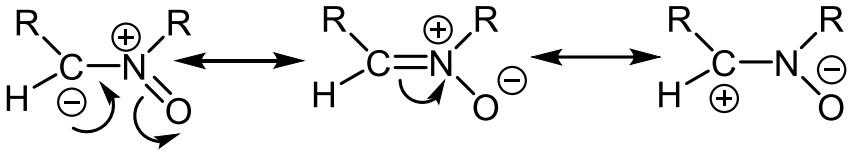


Figure 4:- Structures of simple 6,5- and 6,6-azaspirocycles with nAChR antagonist activity.

1.4. Chemistry of the nitrone functional group

1.4.1. Background

This project will exploit the diverse reactivity profile of nitrones as a strategy towards synthesising the core structures of alkaloids and their derivatives. Furthermore, the research described herein will centre on the application of this functional group to the development of synthetic routes to azaspirocycles and libraries of analogues.



Scheme 1:-1,3 dipole structure of a nitrone

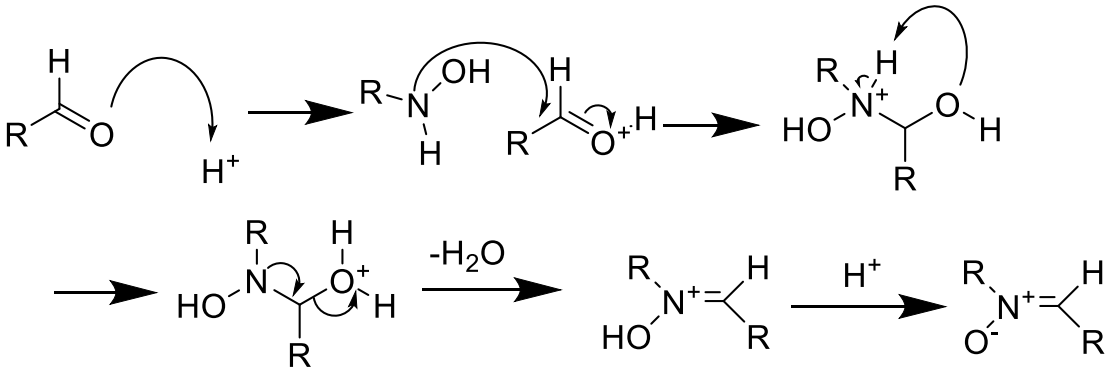
Nitrones are an allyl 1,3-dipole *N*-oxide and can be considered a *N*-oxide of an imine with the general formula X-CH=NO-Y.[32] The system consists of four π elections (two elections from the double bond and two electrons from the oxygen lone pair) across the three (C, N and O) atoms. (Scheme 1). Spectroscopic analyses of nitrones identifies several key characteristics. A sharp peak at around 1600 cm^{-1} is detected when nitrones are analysed using IR spectroscopy. The exact nature of this peak, after initial confusion, has been theorised to be due to the C=N bond stretch. A further peak in the 1170-1280cm^{-1} range can also be observed arising from the N^{+}-O^{-} bond stretch.[33, 34]

1.4.2 Synthesis of nitrones

A number of investigations have been carried out towards the synthesis of both cyclic and acyclic nitrones.[33-36] These approaches have been generally based on imine-enamine-type condensations of hydroxylamines or oximes, nucleophilic additions of hydroxylamines and direct oxidation of *N*-hydroxylamines or amines.

1.4.2.1. Condensation *of N*-monosubstituted hydroxylamines

A widely utilised strategy towards nitrones involves the condensation of aldehydes and ketones with *N*,*N*-disubstituted hydroxylamines proceeding *via* a mechanism akin to conventional imine formation – the nitrone corresponding to the iminium intermediate formed in such reactions. (Scheme 2)

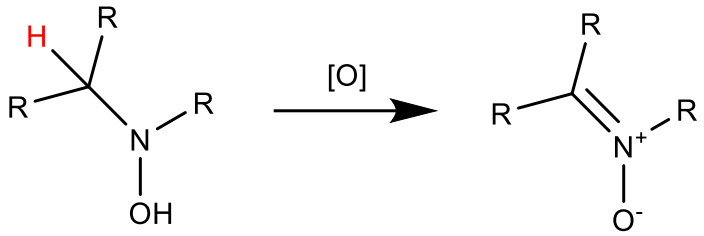


Scheme 2- Formation of nitrones via condensation of a secondary hydroxylamine with an aldehyde/ketone.

The condensation method generally proceeds more favourably using aldehydes rather than ketones. Although some aldehydes undergo polymerisation under basic reaction conditions.[33] Also, while this method is commonly used, there are limitations associated with steric hindrance arising from the presence of bulky R-groups surrounding the ketone which may prevent formation of the nitrone.[33, 34]

1.4.2.2. Oxidation of *N,N* -disubstituted hydroxylamines

One of the most widely used and effective methods of synthesising both acyclic and cyclic nitrones is the oxidation of *N,N*-disubstituted hydroxylamines. Studies have been carried out using a diverse range of common oxidising reagents under mild conditions.[33, 35, 36] One limitation is the carbon atom attached to the nitrogen in the hydroxylamine must carry a removable proton. (Scheme 3)



Scheme 3:- Synthesis of nitrone from *N,N*-disubstituted hydroxylamine

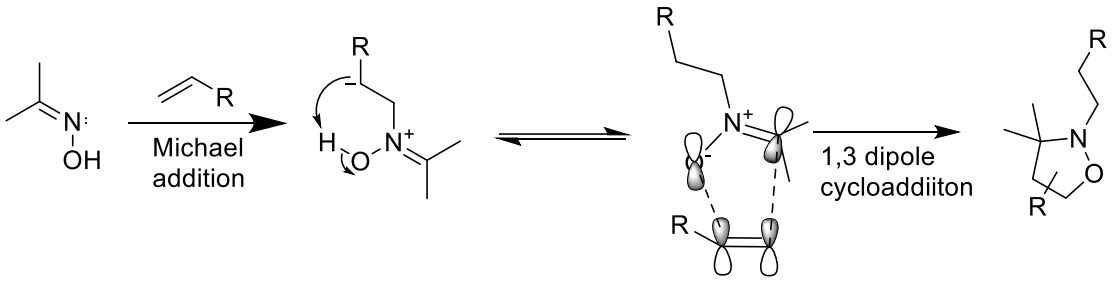
A wide array of oxidants are effective including mercuric oxide,[37] hypervalent iodine reagents,[38] Dess-Martin periodinane, activated manganese oxide,[39] hydrogen peroxide,[40] sodium hypochlorite,[41] and *t*-butyl hydroperoxide.[42] In fact it has also been reported that simply bubbling oxygen through a hydroxylamine solution in the presence of a suitable catalyst is sufficient to convert a hydroxylamine to the target nitrone.[43] Indeed autoxidation can occur, without need for a catalyst, in the presence of O_2.[44] These oxidation methods ideally proceed under relatively mild conditions with relatively few by-products. [38]

The use of yellow mercury(II) oxide, in particular, has been reported as effective with a diverse range of hydroxylamines under mild conditions and good yields. Moreover, high regioselectivity with unsymmetrical substrates are often observed.[35] Although effective, the use of mercuric oxide is being phased out due to toxic oxidant by-products.[45] The less toxic manganese oxide has been found to be an effective, less hazardous, alternative oxidant capable of oxidation to nitrones in good to excellent yields.[36]

1.4.2.3. Intramolecular cyclisation of Oximes

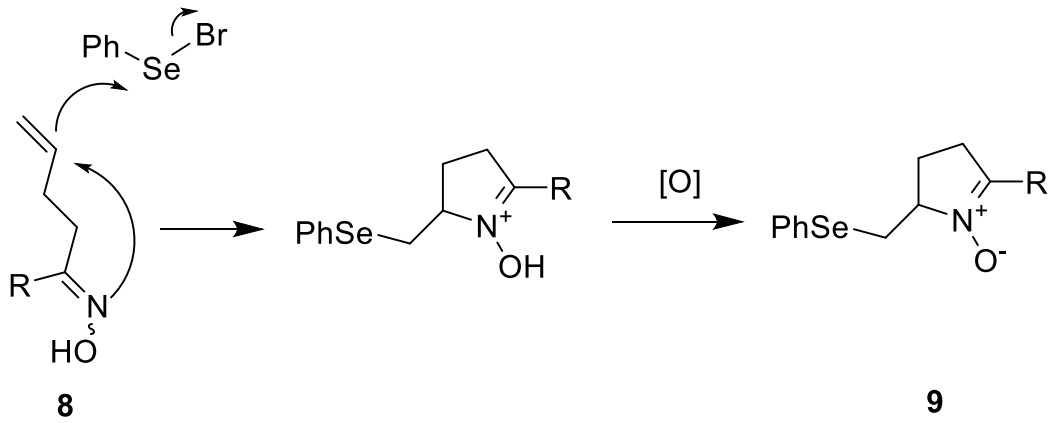
Oximes have been observed to form nitrones via *N*-alkylation reactions under certain conditions. Oximes are of particular interest in the synthesis of nitrones due to the relative ease of preparation and the diverse range of oximes that can be utilised.[46] However, the use of oximes has its drawbacks, such as competing production of oxime-*O*-ethers as well as nitrone products.[33] Nucleophilic addition can occur onto both small and larger alkyl bromides, with the smaller alkyl bromides favouring formation of nitrone products over oxime-*O*-ethers, due to the more favourable steric interactions with the less bulky substituents.[33, 47]

Grigg *et al.* released a series of studies investigating the tandem formation of nitrones from oximes *via* Michael addition followed by a 1,3 dipolar cycloaddition to afford a heterocycle system. The presence of two adjacent lone pair-containing atoms makes these prime candidates for a Michael addition in a 1,2-hydrogen shift procedure.[54,] (Scheme 4) However this route is complicated due to low regio- and stereospecificity leading to different products depending on if the Michael addition and subsequent cycloaddition occur intra- or intermolecularly.[48, 49]



Scheme 4:- General cascade synthesis of heterocycles *via* a 1,2-hydrogen shift formation of nitrone.

Dondas *et al.* reported using phenylselenyl halides in an impressive example of this nitrone formation-cyclisation tandem reaction to form an isoxazolidine *via* a nitrone intermediate.[46] The electrophile, in this case phenylselenyl bromide, initiates the reaction by activating the terminal alkene on oxime **8**. This, in turn, leads to nucleophilic attack from the oxime providing access to cyclic nitrone **9**. (Scheme 5) [46]

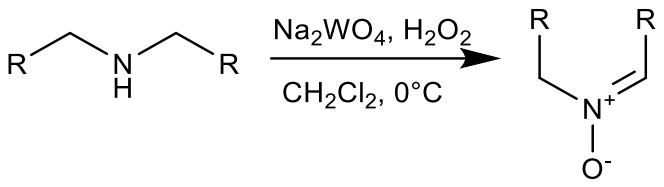


8 **9**

Scheme 5:- Formation of a cyclic nitrone from an oxime

1.4.2.4. Oxidation of Secondary Amines

A wide variety of oxidising agents can oxidise secondary amines, directly, to nitrones.[35, 36] Conversion of amines to nitrones have been investigated due to the potential similarity between this process and the biological process of dehydrogenation of amines by mitochondrial monoamine oxidases.[50] Addition of hydrogen peroxide in the presence of a variety of metal catalysts was first carried out by Murahashi *et al.*, with sodium tungstate dihydrate ($H_4Na_2O_6W$) found to be the most effective catalyst.[50] (Scheme *6*) The active catalyst present is a peroxytungstate species, formed from the reaction between the sodium tungstate dihydrate and hydrogen peroxide.[50]



Scheme 6:- Oxidation of a secondary amine to give a cyclic nitrone

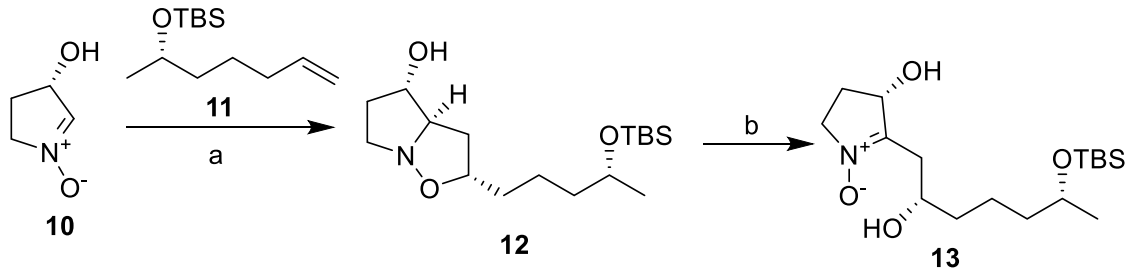
A range of other metal-based reagents, in conjunction with hydrogen peroxide, have been utilised including selenium dioxide,[51] MTO,[52] titanium(IV)[53] and platinum(II) complexes.[54] A metal-free alternative has been investigated by Gella *et al.*, utilising Oxone as the sole oxidising agent in the reaction.[55] Employment of oxidants such as hydrogen peroxide are of particular interest to synthetic chemists - with H_2O being the only additional by-product this approach represents a relatively "green" oxidation method.[54] However problems arise with this method when synthesising water-soluble nitrone targets.[35]

1.4.2.4. Oxidative ring opening of isoxazolidines

Synthetic methods to access nitrones *via* isoxazolidines have been of particular interest to synthetic chemists, as this strategy allows the development of an iterative nitrone formation/oxidation approach as isoxazolidines are the products of nitrone-based cycloadditions.[35, 36] The N-O bond present in the molecule is particularly labile and can be cleaved under relatively mild conditions.[56]

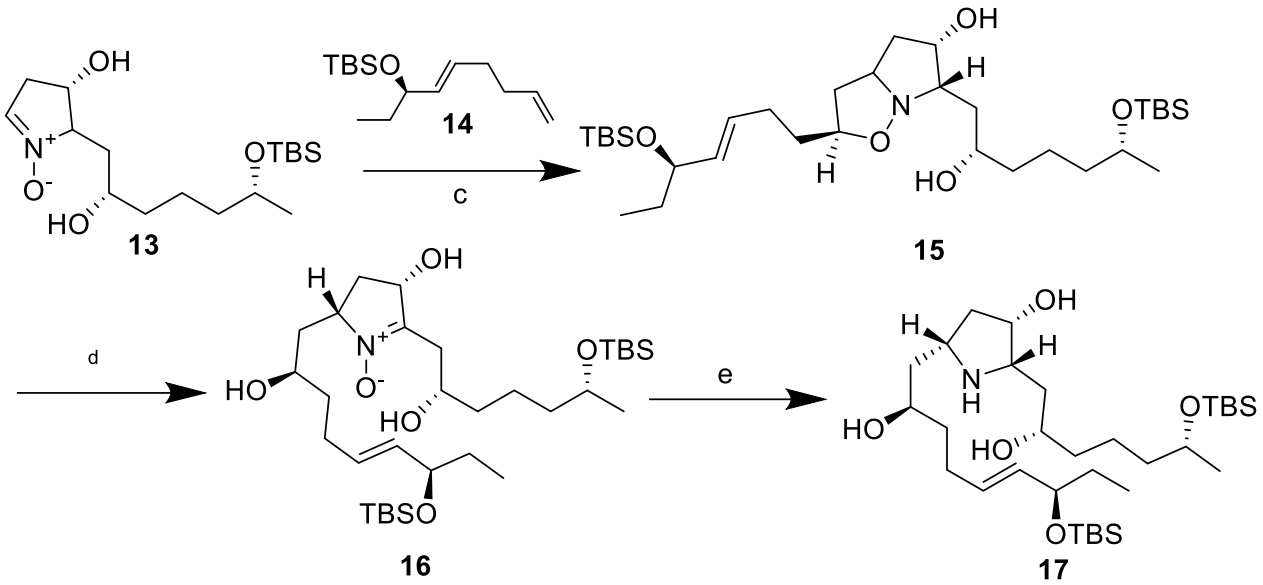
A common strategy towards isoxazolidines is *via* a 1,3-dipolar cycloaddition between a nitrone and a suitable alkene. A range of studies into the use of this strategy have been undertaken by synthesising isoxazolidines, with new stereocentres, in a stereospecific/stereoselective manner from a nitrone, followed by oxidative ring opening to yield a second nitrone for further functionalisation.[56]

Nagasawa *et al*., developed an excellent application of this strategy towards the total synthesis of crambescidin. 1,3-Dipolar cycloaddition onto a chiral, five-membered nitrone **10** with olefin **11**, followed by oxidative ring opening of the subsequent isoxazolidine **12** using mCPBA, gives nitrone intermediate **13** as a single diastereomer (Scheme 7).[57]



Scheme **7**:- *Reagents and conditions*: a) **11**, toluene, 110 ^{o}C, 67 % b) (i) PhOCSCl, Pyridine (ii) Bu_3SnH, AlBN, 52 % c) mCPBA , CH_2Cl_2, 0 ^{o}C

Following synthesis of nitrone **13**, a regioselective intermolecular dipolar cycloaddition with olefin **14** was undertaken, reintroducing the isoxazolidine group to afford **15**. A second oxidative ring opening could then be attempted on this more complex isoxazolidine **15**, with the same conditions using mCPBA to deliver nitrone **16**. A tandem reduction method converted the nitrone moiety to pyrrolidine **17**



Scheme 8:- *Reagents and c*onditions: c) **14,** toluene, 110 ^{o}C, 65 % d) mCPBA , CH_2Cl_2, 0 ^{o}C e) (i) $NaBH_4$, EtOH, 0 ^{o}C (ii) Separation 3. $Mo(CO)_6$, CH_3CN, 42 %

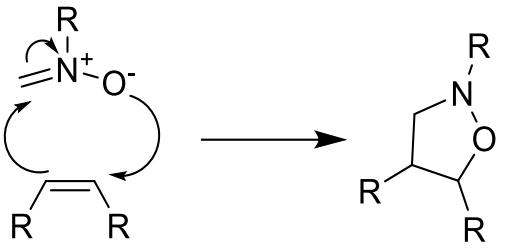
1.4.3. Reactions of nitrones

Nitrones have a diverse reactivity profile making them an attractive proposition for utilisation by organic chemists. These species undergo cycloadditions, nucleophilic attack by organometallic species and, recently have also been shown to participate in additions to electrophiles at the alpha carbon *via* umpolung-based methodologies.[18, 58] Nitrones readily undergo 1,3-dipole cycloaddition reactions with a wide array of carbon-containing dipolarophiles, both electron rich and electron poor, in contrast to the more well know Diels-Alder reaction,[18] to form isoxazolidines.[59]

One of the nitrone resonance contributors reveals the sp^2-carbon to be electron deficient and nitrones are often utilised in a number of nucleophilic addition reactions with organometallic reagents such as organolithium[60] and Grignard reagents [61].

1.4.3.1. 1,3 Dipolar Cycloaddition

Nitrones have been commonly utilised in 1,3-dipole cycloaddition reactions with both electron deficient and electron rich alkenes to provide 5-membered isoxazolidines.[18] The 1,3-dipolar cycloaddition can proceed in inter- and intramolecular forms giving access to a wide variety of isoxazolidines. (Scheme 9)



Scheme 9- Mechanism of a 1,3-dipole cycloaddition

Mechanistic studies, conducted by Huisgen *et al.,* found that the two σ-bonds are formed simultaneously in the transition state *via* the interaction between dipole and dipolaraphile, although the rate of formation of these two σ bonds can differ.[62]

1.4.3.1.1. Stereochemistry

Stereoselectivity of the cycloaddition can be readily predicted by consideration of the stereochemistry of the dipolarophile and alkene as well as *endo* and *exo* effects that determine the approach of the dipolaraphile and dipole with respect to each other. When considering the Diels-Alder reaction, for example, the stereospecificity is locked by the geometry of the dienophile i.e., a *cis*-dienophile will provide a *cis*-substituted product.

The stereoselectivity of the reaction arises from whether the substrate has an *endo* or *exo* approach, with an *endo* product being preferred over an *exo*-product in a process known as the Alder endo rule. The *endo* product is favoured, despite being the less stable transition state owing to steric effects, as there are favourable π-bonding secondary orbital interactions between the carbonyl group on the dienophile and the π-bonds of the diene in the *endo* TS. (Figure 5) This *endo* effect controls the relative stereochemistry between the substituents on the diene and those on the dienophile.

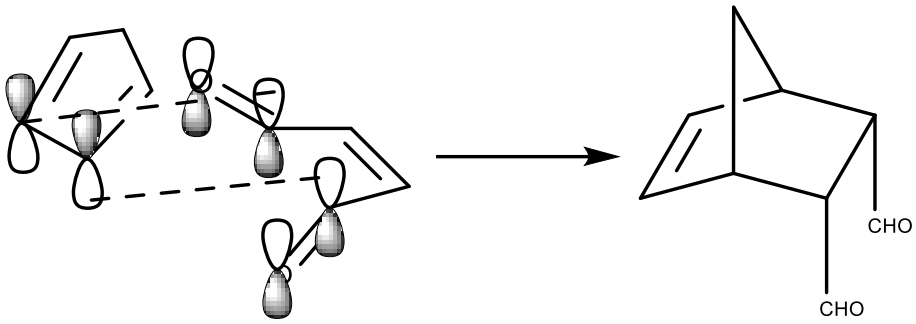


Figure 5:- Stereoselective Diels-alder reaction, giving rise to the favoured *endo* product.

A 1,3-dipolar cycloaddition of a nitrone with a dipolaraphile produces an isoxazolidine in which the stereochemistry of the cycloadduct can be determined in the same manner as for the Diels-Alder reaction. Thus, a *cis*-alkene will provide a *cis*-substituted product for instance. However, there is an absence of secondary orbital interactions (in contrast to the Diels-Alder reaction) which results in the *exo*-product predominating as steric interactions are now the most powerful force influencing the approach of the two reactants.

1.4.3.1.2. Regioselectivity

The regioselectivity of a cycloaddition can be determined by examining interactions of the FMO's of the 1,3-dipole and dipolarophiles. A principle of maximum overlap means the preferred regioisomers in the cycloaddition reaction arise by interaction of the orbitals on the dipole and dipolaraphile with largest coefficients. The atom with the largest coefficient on the HOMO will overlap with that with the largest coefficient on the LUMO to provide the product.[63]

For a 1,3-dipolar cycloaddition to occur between a nitrone and a dipolaraphiles, 4 π electrons from the dipole and 2 π electrons from the dipolaraphiles interact in a pericyclic [4+2] cycloaddition similar to a Diels-Alder cycloaddition.[63]

As nitrones can act as both an electrophile and nucleophile, the HOMO or the LUMO can occur on the nitrone or the dipolaraphile depending on whether the dipolaraphile is electron-rich or electron-deficient. Depending on the relative energy of FMO's the most important interactions can be $HOMO_{nitrone}$ – $LUMO_{dipolarophile}$ or *vice versa*. Thus, regioselectivity can only be predicted by prior determination of which are the interacting FMO's followed by determining the relative size of coefficients on these. (Figure 6)

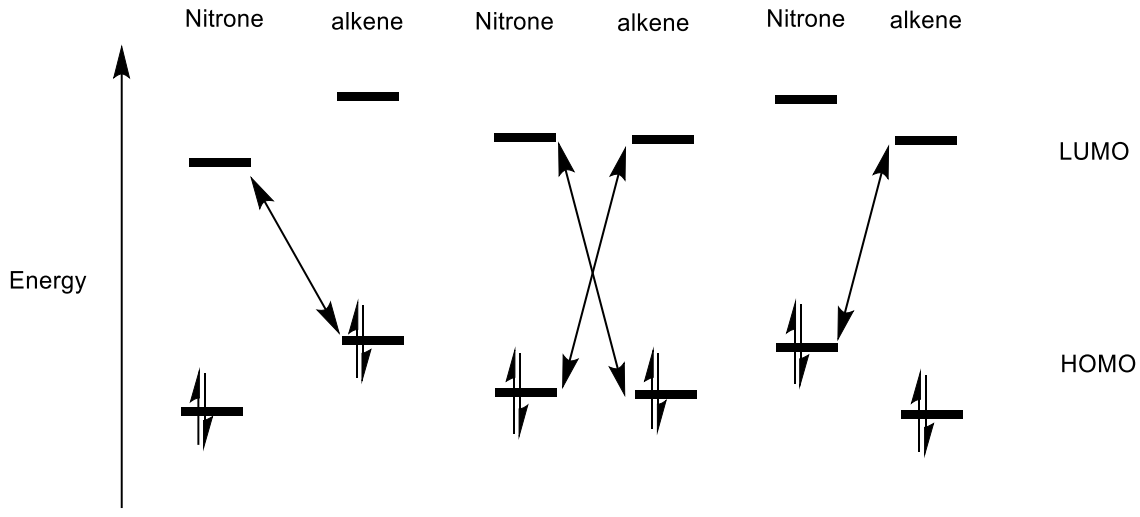


Figure 6:- Frontier molecular orbital diagram showing the different possible interactions between FMO's on nitrone and dipolaraphile

In situations where the cycloaddition is dominated by $HOMO_{nitrone}$ – $LUMO_{dipolarophile}$ interactions Lewis acids can be deployed to lower the energy of the $LUMO_{dipolaraphile}$,

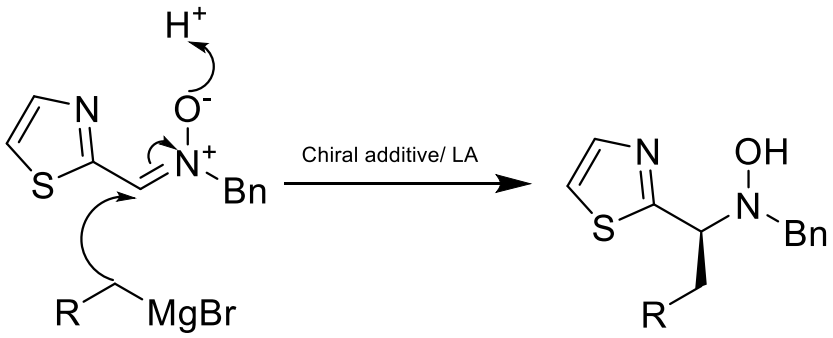
thus decreasing the energy gap between HOMO and LUMO and accelerating the reaction.

1.4.3.2. Nucleophilic addition

Nitrones react readily with nucleophiles such as organolithium and Grignard reagents to give substituted hydroxylamines. While organolithium/magnesium species have been the subject of most interest , studies have also been conducted into the use of silylketene acetals,[64] hydrogen cyanide,[65] and phosphorous-containing nucleophiles.[66]

The oxygen atom on the nitrone acts as an additional chelation centre, which subsequently allows the formation of a 5-membered transition state during the nucleophilic addition and this leads to an increase in electrophilicity of the C=N double bond.[67]

An enantioselective nucleophilic addition is desirable when pursuing natural product targets with a key stereocentre. For example, Dondoni *et al.* investigated a series of stereoselective Grignard additions utilising nitrones in conjunction with chiral ligand additives and Lewis acids (Scheme 10). The use of D-glucose diacetonide and $ZnBr_2$ led to cycloaddition with up to 74% ee and in good yield to give substituted *N*,*N*-disubstituted hydroxylamine.[68]



Scheme 10: Enantioselective nucleophilic addition mechanism of an organometallic reagent to a nitrone.

2. Previous synthesises of natural products

2.1. Previous synthetic studies towards HTX

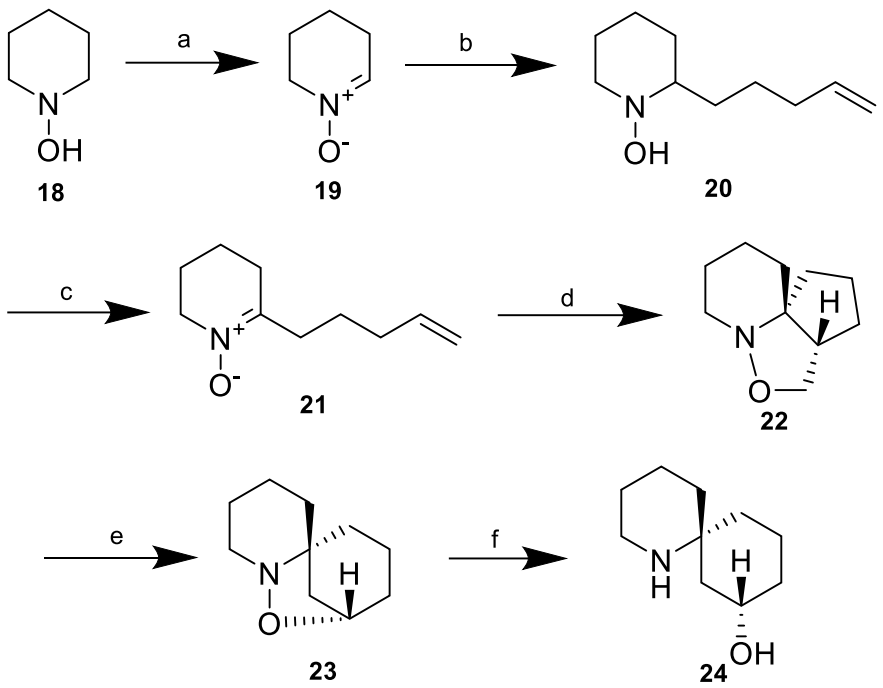
A wide range of approaches to the histrionicotoxin family of alkaloids have been investigated by numerous research groups, stretching back over 50 years.[13, 69] In this section only those involving nitrone cycloadditions as key steps, which are of relevance to the work in this thesis, will be discussed. A range of different strategies will be discussed here from those based on functionalising a pre-formed spiro-scaffold to approaches based on cyclising a heavily functionalised, acyclic precursor. The advantage of the former is the flexibility in easily accessing a range of analogues by altering functionalisation strategies.

However, the former depends on the use of scaffolds with a diverse reactivity profile and with good diastereofacial bias to enable high levels of stereselectivity during functionalisation. While this may be easier when adopting an early functionalisation strategy, such an approach requires an almost entirely new synthetic route to access each new analogue and the survival of different types of functionality throughout the synthesis. Furthermore, different functionality may adversely effect key cyclisation reactions later in the synthesis.

2.1.1. Gossinger group

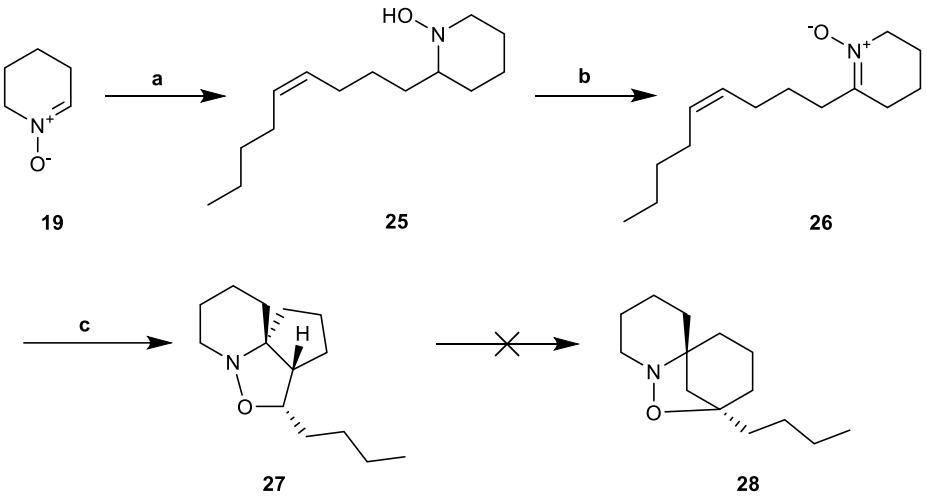
Nitrone chemistry has been utilised in an attempt to access (±)-2-depentenyl-7-debutyl pHTX by Gossinger *et al.* in 1975.[37] Although pHTX **24** was successfully synthesised, (±)-2-depentenyl pHTX **28** could not be successfully accessed, due to steric impairment preventing the desired retrocyclisation occurring to form the desired 6,6,5-cycloadduct. This is theorised to be a result of steric hindrance arising from the presence of the butyl side chain.[13] Tufariello *et al.* encountered similar issues with this step due to potential steric hindrance, preventing the intramolecular cycloaddition. [70]

In the synthetic studies directed towards (±)-2-depentyl-7-debutyl-pHTX **24** (Scheme 11), 1,5-dibromopentane underwent a double S_N2 addition reaction, in the presence of a base, to give piperidinol **18**. This hydroxylamine underwent an oxidation to cyclic nitrone **19** before a Grignard addition of pentenylmagnesium bromide gave hydroxylamine **20**. This hydroxylamine was again oxidised to nitrone **21**, which, when heated in toluene, underwent an intramolecular 1,3-dipole cycloaddition under kinetic conditions to give access to a 6,5,5-azaspirocycle adduct **22** as the major product. Further heating of this spirocyclic adduct in a sealed tube, under thermodynamic control, led to the formation of the more thermodynamically stable cycloadduct - the 6,6,5-azaspirocycle **23**. Following a Raney-nickel hydrogenation to cleave the N-O bond (±)-2-depentyl-7-debutyl-pHTX **24**, was successfully synthesised.[13, 37]



Scheme 11:- *Reagents and conditions:* (a) HgO, CH_2Cl_2 (b) $BrMg(CH_2)_3CH=CH_2$, THF, r.t (c) HgO, CH_2Cl_2 (d) toluene, reflux; (e) Toluene, 240 $^{\circ}C$ (f) H_2, Raney Ni, EtOH.

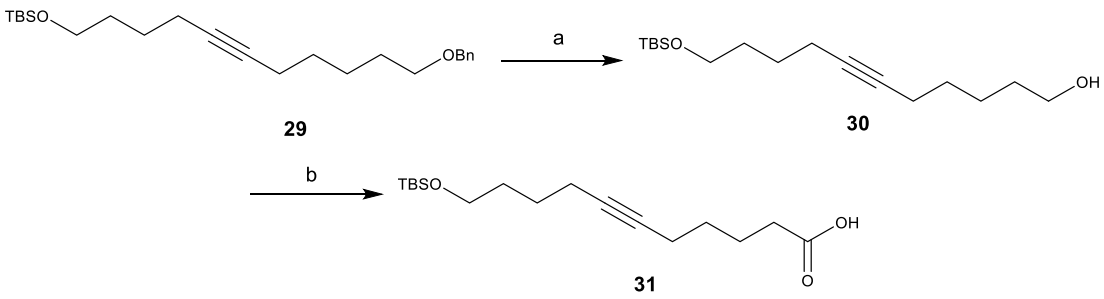
Following the successful synthesis of pHTX **24**, the Gossinger group attempted the synthesis of (±)-2-depentenyl pHTX adduct (Scheme 12). Cyclic nitrone **19** was synthesised using the same route as before using a Grignard addition to generate the olefin side chain in hydroxylamine **25** before oxidising to nitrone **26**. Again, this nitrone was cyclised, to give the 6,5,5-adduct **27**. Finally, the goal was to induce the [6,5,5]-structure of **27** to re-arrange to the [6,6,5], thermodynamically favoured, tricyclic product **28** bearing a pentyl side chain through exposure forcing, thermodynamic conditions under high heat/pressure. However, access to (±)-2-depentenyl pHTX **28** was unsuccessful due to the postulated steric hindrance arising from the presence of the butyl side chain as discussed above.[13, 37]



Scheme 12:- *Reagents and conditions:* a) $BrMg(CH_2)_3CH=CH(CH_2)_3Me$, Et_2O, reflux b) HgO, CH_2Cl_2 c) toluene, reflux d) toluene, 195 ℃

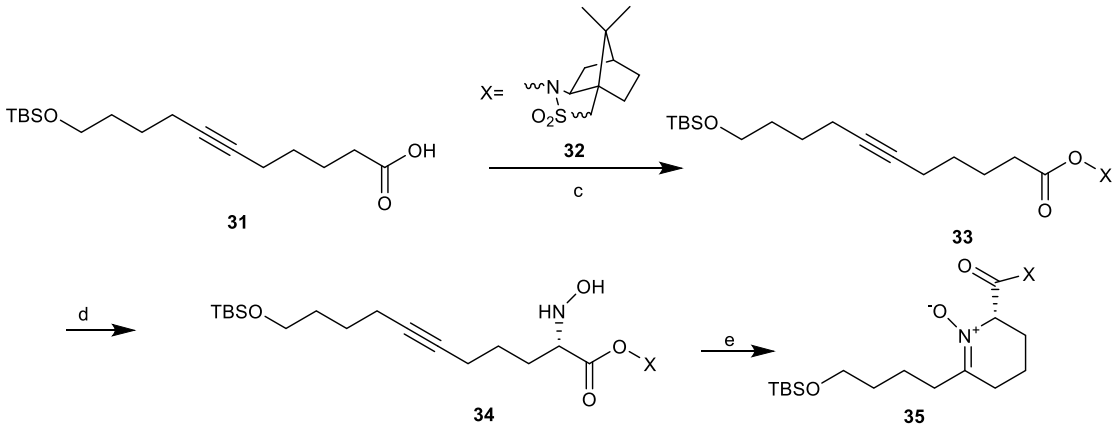
2.1.2. Holmes group

In 1999, the Holmes group published their enantioselective total synthesis towards HTX analogue HTX 283A **50**, which utilised a key [3+2] nitrone cycloaddition.[71] The procedure began with commercially available, differentially protected, dihydroxyacetylene **29**, which underwent debenzylation to give **30** before a Jones oxidation to give carboxylic acid **31**. (Scheme *13*)



Scheme 13:- *Reagents and conditions:* a) BCl_3·DMS, CH_2Cl_2 (b) Jones' reagent, acetone.

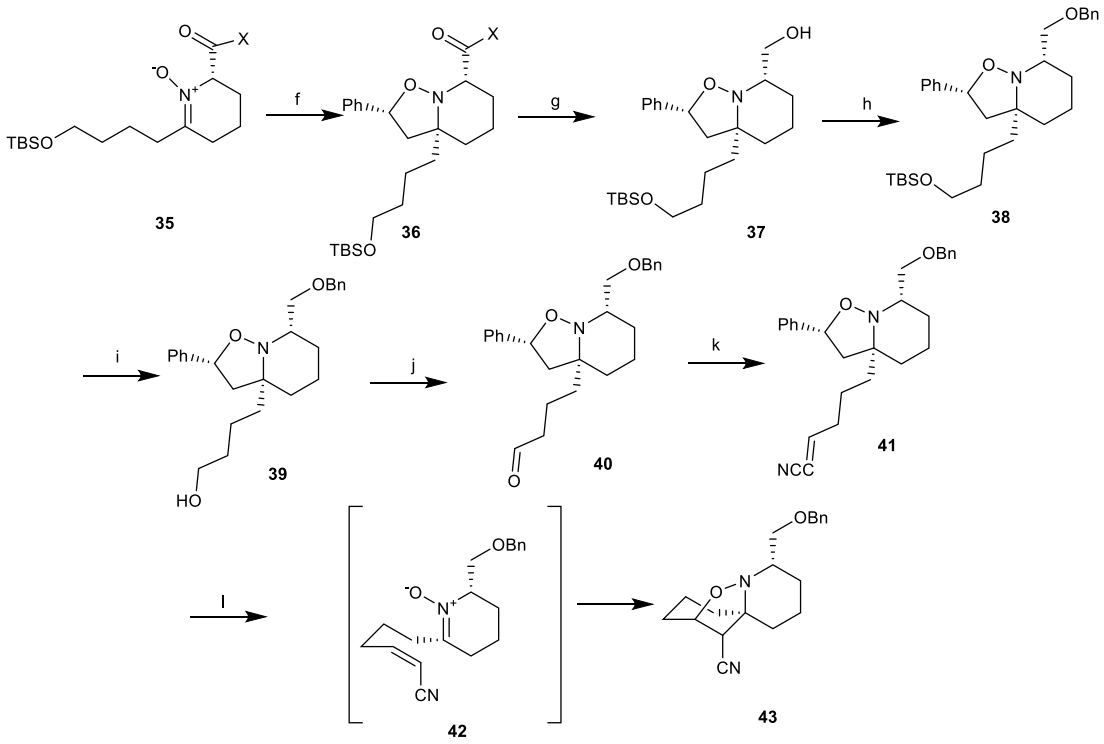
Next, (1*R*)-(+)-2,10-camphorsultam **32** was incorporated as a chiral auxiliary to give ester**33** as the single (*S*)-enantiomer. Hydroxylamine **34** was synthesised, diastereoselectively, in the next step under acidic conditions *via* acid hydrolysis. This was followed by formation of nitrone **35** *via* a hydroxylamine-alkyne cyclisation.[13, 71] (Scheme 14)



Scheme 14*:*- *Reagents and conditions:* c) Et_3N, pivaloyl chloride; (1R)-(+)-2,10-camphorsultam **32**, n-BuLi, THF, −78 ˚C d) $NaN(TMS)_2$,THF; 1-chloro-1-nitrosocyclohexane, THF, HCl e) toluene, 80 ˚C

The next series of steps focussed on protecting the nitrone group and elaborating the four-carbon side chain towards the goal of initiating an intramolecular dipolar

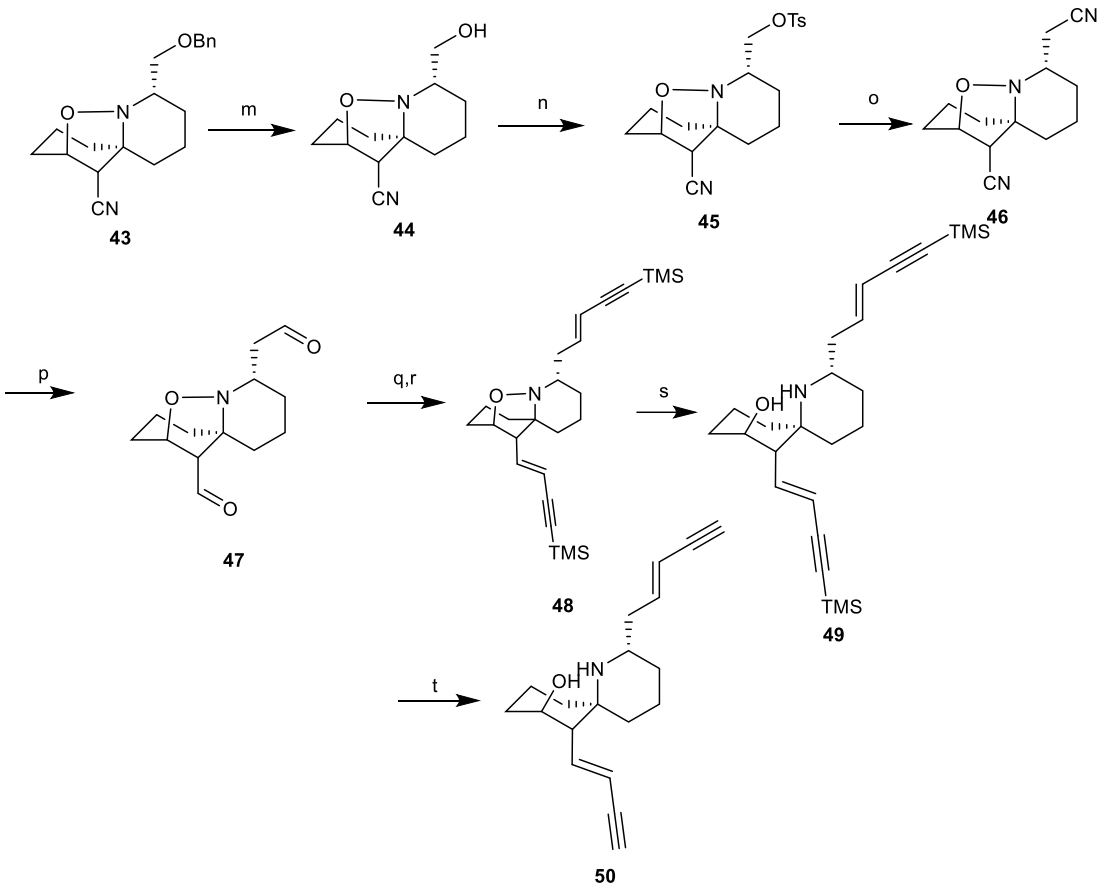
cycloaddition to access the spirocyclic core of HTX. The nitrone was protected as a styrene cycloadduct, as an unusual approach to mask the nitrone functional group. Protected isoxazolidine **36** was synthesised regio- and stereoselectively following an intermolecular 1,3-dipole cycloaddition with styrene. Several steps were next undertaken to remove the now redundant, chiral auxiliary to provide alcohol **37**. Selective protection of the alcohol with a benzyl group gave **38** before de-silylation to **39** and subsequent oxidation gave aldehyde **40**. A Peterson olefination followed to give access to *cis*-α,β-unsaturated nitrile **41**. Heating in a sealed tube at a high temperature released the styrene moiety in a retro [3+2] intermolecular cycloaddition to unmask the nitrone group, affording nitrone **42**. Another cycloaddition followed, this time in a intramolecular fashion, allowing access to the preferred thermodynamically favoured cycloadduct **43.** [13, 71] Fortunately, the nitrile group did not hinder the cycloaddition, as observed by research groups in previous studies using bulkier alkyl groups when attempted a similar cycloaddition.[13] (Scheme 15).



Scheme 15:- *Reagents and conditions:* f) Styrene, 75 °C; g) $LiAlH_4$, THF, 0 °C; h) NaH, BnBr, THF; i) HF, MeCN; j) TPAP, NMO; k) Me_3SiCH_2CN, n-BuLi, THF, $B(O^iPr)_3$,–78 °C; l) toluene, 190°C

The final series of steps were directed towards elaboration of the pendant side chains. The OBn group was removed accessing alcohol **44**, before conversion to

tosylate **45**. The tosyl group was converted to the dinitrile **46** which, upon treatment with DIBAL, gave dialdehyde **47**. A Stork-Wittig reaction followed by a Sonogashira coupling gave the *bis*-enyne **48** which was converted to HTX 283A **50** by reductive ring opening and subsequent deprotection. This total synthesis proceeds in an impressive overall yield of 16 % over 20 steps (Scheme 16).[13, 71]

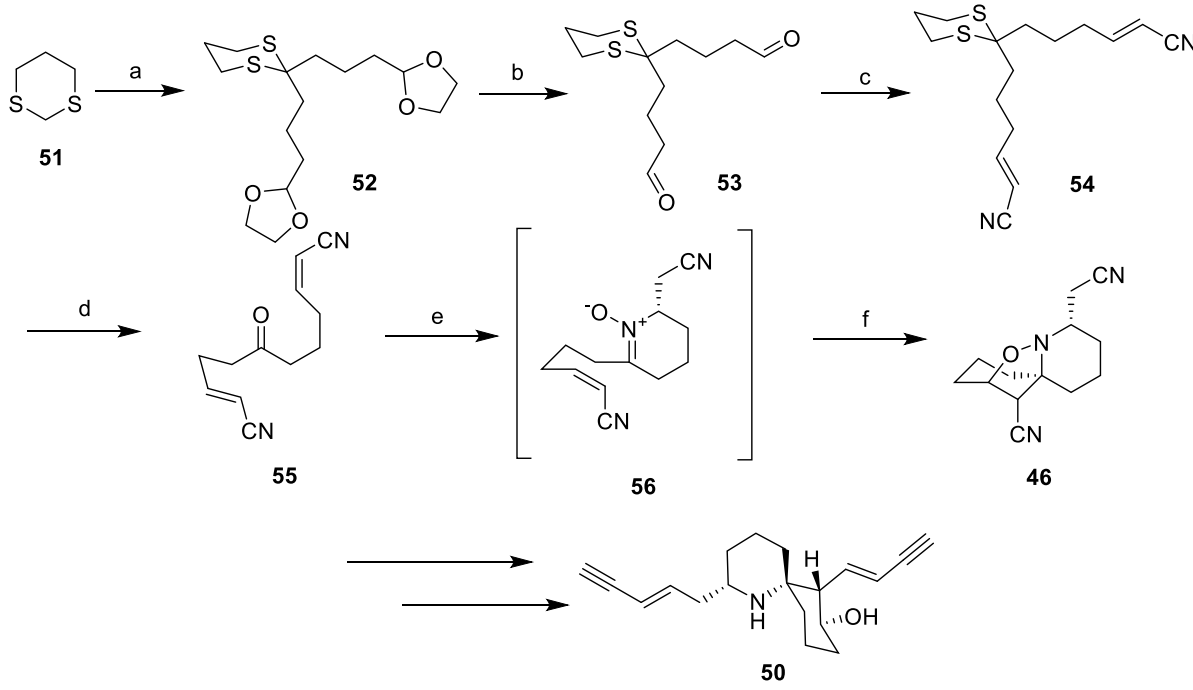


Scheme 16:- *Reagents and conditions:* m) $BCl_3{\cdot}Me_2S$, CH_2Cl_2; n) MsCl, Et_3N, DMAP, CH_2Cl_2; o) NaCN, DMSO, 55 °C; p) DIBAL, toluene,−78 °C; q) $KN(TMS)_2$, $[Ph_3P^+CH_2II^-]$, THF, −78 °C; r) $Pd(PPh_3)_4$,CuI,Et_2NH, $Me_3Si–C{\equiv}CH$ s) Zn, acetic acid t) K_2CO_3, MeOH.

2.1.3. Stockman group

Following the successful total synthesis of HTX 283A **50** by the Holmes group, Stockman *et al.* devised an alternative route towards the same target by developing a novel, bidirectional, route towards dinitrile **46** *via* a spironitrone intermediate.[72] The procedure began with commercially available 1,3-dithiane (**51**) which underwent a double alkylation *via* addition of *n*-butyllithium followed by 2-(3′-chloropropyl)-1,3-dioxolane to access symmetrical diacetal **52**. A simple deprotection followed to dialdehyde **53**, before a double Peterson olefination afforded dinitrile **54**.

Silver nitrate and NCS were used to remove the dithiane group and subsequent conversion to the oxime led to a spontaneous Michael addition to access the key nitrone **56** as an intermediate. A subsequent thermodynamic cycloaddition followed, completing the formal synthesis giving access to the dinitrile-functionalised 6,6,5-azaspirocycle intermediate **46** previously synthesised by the Holmes group (Scheme 17).[13, 72]



Scheme 17:- *Reagents and conditions: a)* ^{n}BuLi, THF, HMPA,−78 °C; 2-(3-chloropropyl)-1,3-dioxolane b) HCl, THF, H_2O; c) Me_3SiCH_2CN, THF, ^{n}BuLi,−78 °C; d) NCS, $AgNO_3$, MeCN; e) $NH_2OH{\cdot}HCl$, NaOAc, MeOH f) toluene, 160 °C, sealed tube.

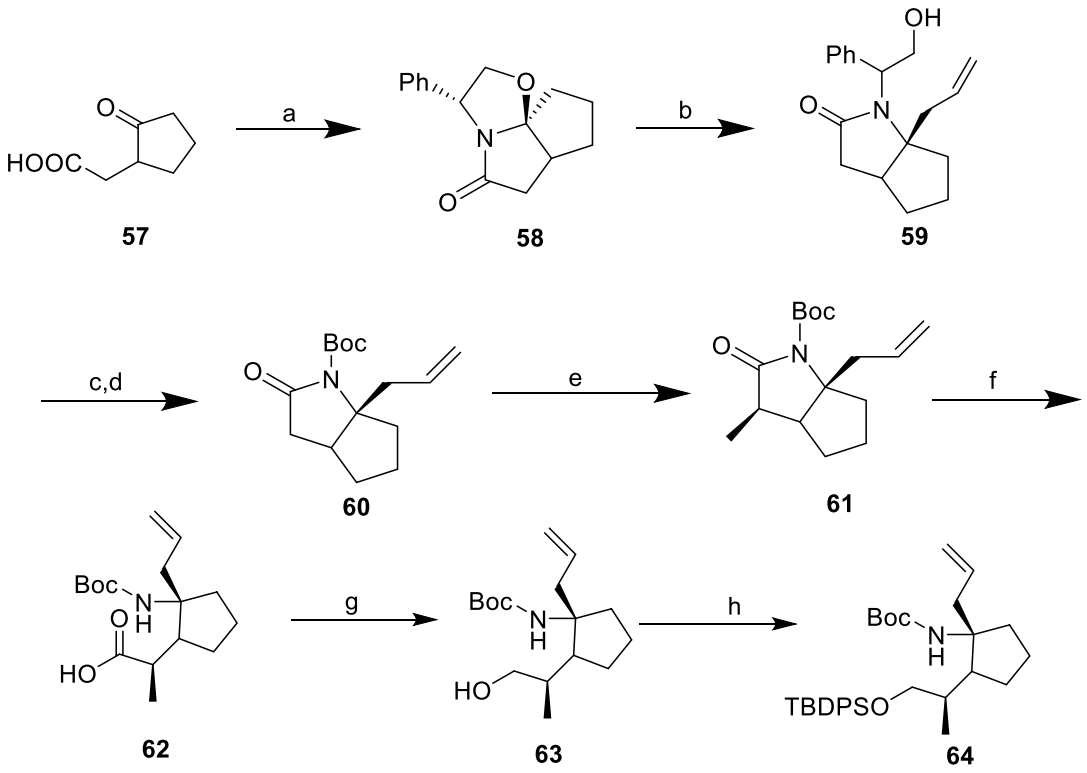
2.2 Previous synthesis towards pinnaic acid

As is the case with the histrionicotoxins, the unique azaspirocyclic core of pinnaic acid and halichlorine have stimulated a range of approaches to the synthesis of core structures and target molecules.[30, 73-75] This section will highlight those methods which utilise nitrone-based chemistry as key steps and will also cover the seminal first total syntheses by Danishefsky group where the chemistry and absolute stereochemistry of pinnaic acid/halichlorine was established.[30]

2.2.1. Danishefsky group

The Danishefsky research group engaged in synthetic studies towards the natural product target pinnaic acid **2** and were the first to achieve total synthesis.[76] Danishefsky *et al.* were interested in both this compound and halichlorine **3** due to the structurally similar azaspirocyclic backbone of the two natural product targets.[30]

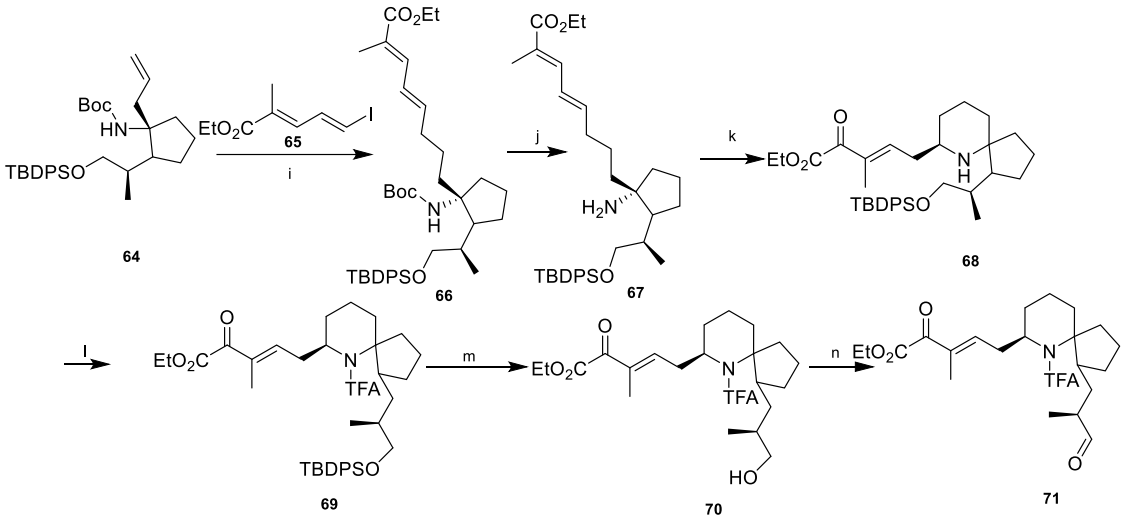
Total synthesis of pinnaic acid **2** began by preparing the key chiral, non-racemic starting material – "Meyer's lactam" **58** - by exposure of D-phenylglycinol to keto acid **57**. This lactam was allylated, in the presence of a Lewis acid, to access a bicyclic lactam **59**. De-alkylation and subsequent protection of the now free secondary amine afforded protected amine **60**. A key stereoselective methylation followed to establish the desired absolute stereochemistry at the C14 position in pinnaic acid. Thus, methylated bicyclic lactam **61** was successfully synthesised using lithium *bis*(trimethylsilyl)amide as a base and treatment with methyl iodide. Next, hydrolytic ring cleavage gave carboxylic acid **62** which was then converted to alcohol **63** before subsequent *O*-silylation to TBDPS-protected alcohol **64** (Scheme 18).[76]



Scheme 18:- *Reagents and conditions:* a) D-phenylglycinol, toluene, heat, 95 % b) Allyltrimethylsilane, $TiCl_4$, CH_2Cl_2, r.t., 99 % c) Na, NH_3 , THF, EtOH, −78 °C, 99 % d)

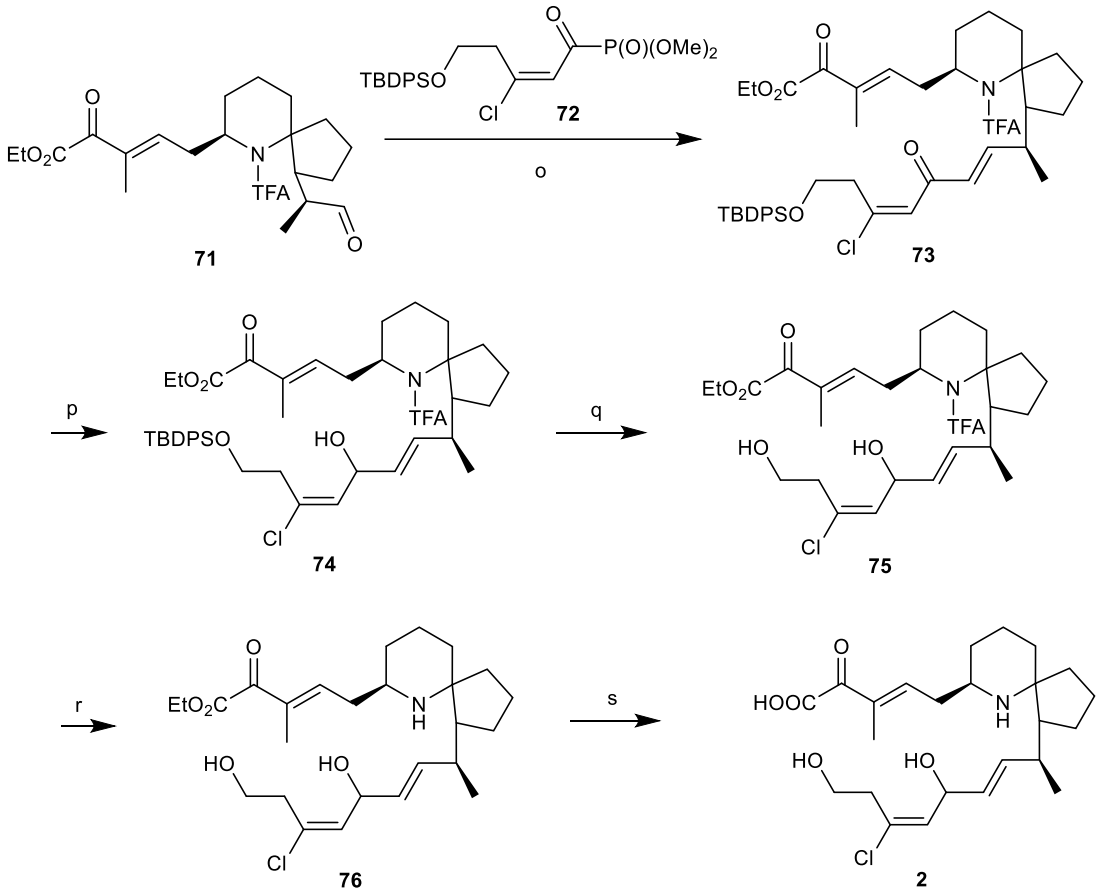
Boc_2O, DMAP, THF , 96% e) LiHMDS, MeI, THF ,−78 ˚C to 0 °C, 90% f) LiOH, THF, H_2O, −78 ˚C, 89% g) ClCOOEt, Et_3N, THF, $NaBH_4$, MeOH 82% h) TBDPSCl, Et_3N, DMAP, CH_2Cl_2, 95%

Hydroboration of **64** with 9-BBN followed by cross-coupling of the resulting organoborane with vinyl iodide **65** gave diene **66**. Following a difficult deprotonation of the nitrogen atom, treatment with DBU effected a stereocontrolled cyclisation providing spirocyclic (*E*)-alkene **68**. The nitrogen atom was re-protected with a TFA group giving access to compound **69**, before deprotection of the TBDPS group produced primary alcohol **70**. Subsequent oxidation gave aldehyde **71** (Scheme 19).



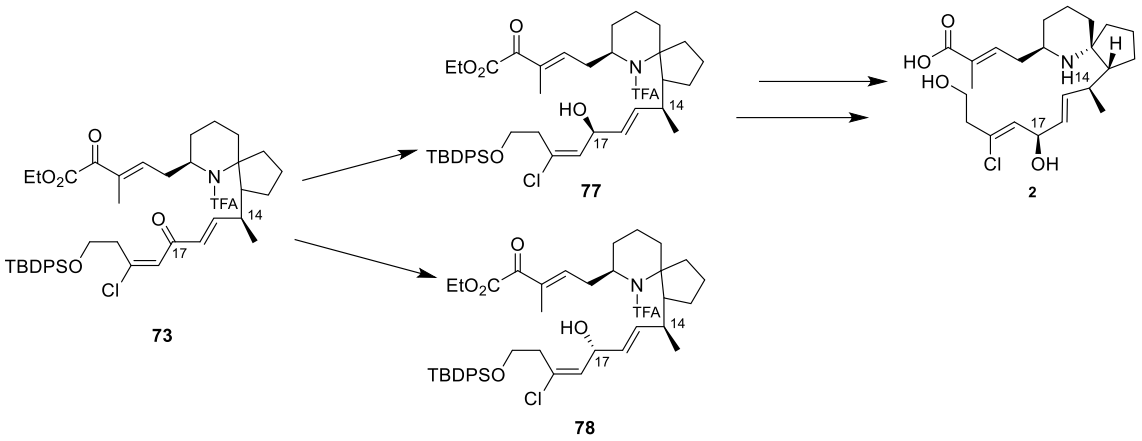
Scheme 19:-. *Reagents and conditions:* i) **65**, 9-BBN, THF; Pd(dppf)Cl_2, CH_2Cl_2, Ph_3As, Cs_2CO_3, DMF-Water, 75 % j) TFA, CH_2Cl_2 k) DBU, 81 % (over 2 steps) l) $(CF_3CO)_2O$, $ClCH_2CH_2Cl$, 0 ˚C, 88 % m) HF.Pyr, THF, pyridine, r.t, 91 % n) $^{i}Pr_4NRuO_4$, NMO, MeCN, 84 %

Unsaturated ketone **73** was accessed following a base-mediated coupling of phosphonate **72** with aldehyde **71**. Reduction of ketone **73** in the presence of (*S*)-alpine hydride gave alcohol **74** as a diastereomeric mixture. This mixture was subjected to desilylation, deprotection of the nitrogen (**76**) and ester hydrolysis to give natural product **2** as the final product in 19 steps.[30,76]



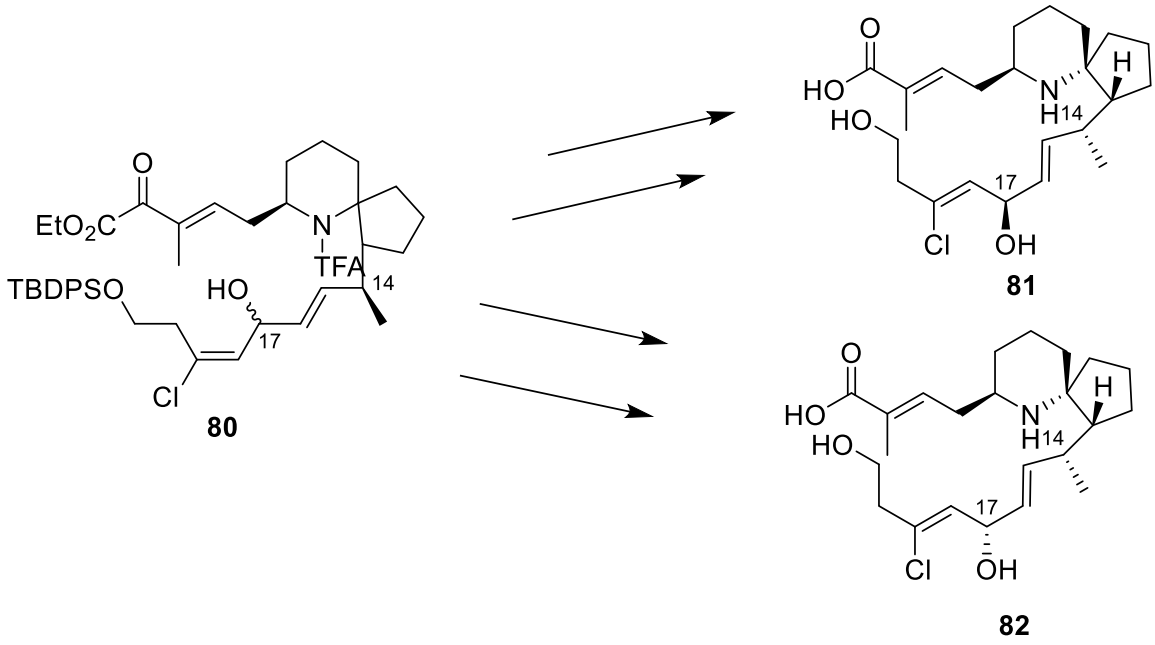
Scheme 20:- *Reagents and conditions:* o) LHMDS, THF, -78 ^{o}C p) (R) or (S)-alpine hydride, 30 % q) HF-Pyridine, THF, Pyridine, 0 ^{o}C, 95 % r) $NaBH_4$, EtOH, r.t, 93 % s) LiOH, THF/MeOH, H_2O, r.t, 90 %

However, while Danishefsky and his cohort correctly confirmed they had synthesised pinnaic acid **2** with correct relative stereochemistry, there was no reference sample on hand for comparison. Further experiments were undertaken to confirm the absolute stereochemistry of C14 and C17. Once ketone **73** was obtained, it was alternatively reduced using $CeCl_3$ and $NaBH_4$ to give alcohol **77**, with inverted (*S*)-stereochemistry at the C17 position, before an identical conclusion to the synthetic procedure gave pinnaic acid isomer **78** with inverted stereochemistry at the C17 position.



Scheme 21:- Investigations to determine the correct stereochemistry at the C17 position of synthesised pinnaic acid **2**

An alternative olefination on aldehyde was undertaken to investigate the stereocentre at the C14 position. Lactam **61** was deprotonated and reprotonated, at what would become the eventual C14 position, to afford epimeric lactam **79** with inverted (*S*)-stereochemistry at C14. Steps continued as with the other isomer, directed towards the ketone product with inverted stereochemistry at the C14 position as opposed to ketone **73.** The next step was again to reduce the ketone group using (*R*)- or (*S*)- alpine hydride affording a diastereomixture of separable alcohols **80**. These could then be individually taken forward, as previously outlined, to afford the final targets **81** and **82**, both epimers at the C17 position.



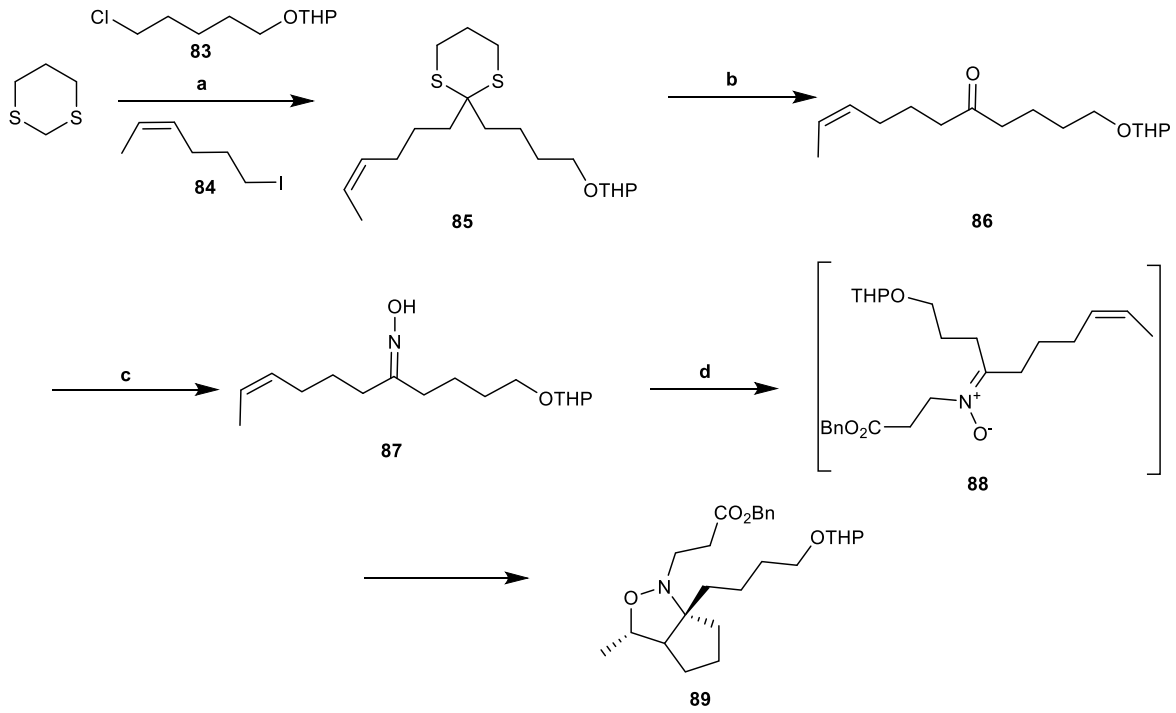
Scheme 22:- Investigations by the Danishefsky group to determine diastereo-assignments of C14 and C17 positions in pinnaic acid **2**

With all combinations of stereoisomers in hand, analysis could confirm the absolute stereochemistry at the C14 centre as(*R*) and the C17 centre as (*R*) by reference to the NMR spectra of naturally occurring pinnaic acid, confirming successful total synthesis of pinnaic acid **2**.[30]

2.2.2. Zhao group

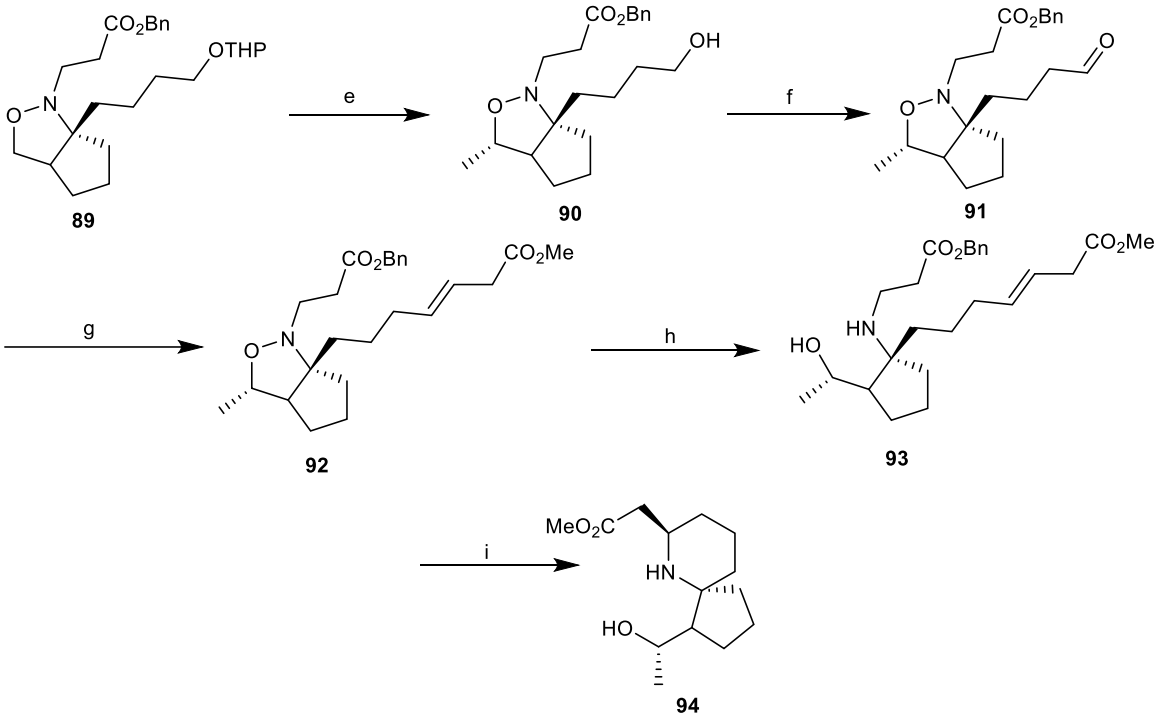
The Zhao research group investigated the synthesis of the pinnaic acid spirocyclic core *via* an a tandem [3+2] cycloaddition of a nitrone formed by intermolecular Michael reaction of an oxime.[77]

Commercially available dithiane was double alkylated with alkyl halides **83** and **84** to give disubstituted dithiane **85**. Hydrolysis gave ketone **86**, which was then converted to oxime **87**. A Michael addition with benzylacrylate was undertaken to access product **89** *via* nitrone intermediate **88** (Scheme 23).[30]



Scheme 23:-. *Reagents and conditions:* a) (i) BuLi, **83**, THF (ii) BuLi, 85 %, **84**, HMPA, THF, 97 % b) NCS, $AgNO_3$, MeCN-water, 97 % c) $H_2NOH.HCl$, AcONa, MeCN, 99 % d) CH_2CHCO_2Bn, xylene, 140 C, 24 hr, 92 %

A deprotection followed by a Swern oxidation gave aldehyde **91** which was then homologated. The isoxazolidine N-O bond on alkene **92** was cleaved using Zn and acetic acid to access amine **93** and a final intramolecular Michael reaction at a high temperature followed to access the desired alcohol **94,** core structure of pinnaic acid **2** (Scheme 24).[30, 77, 78]



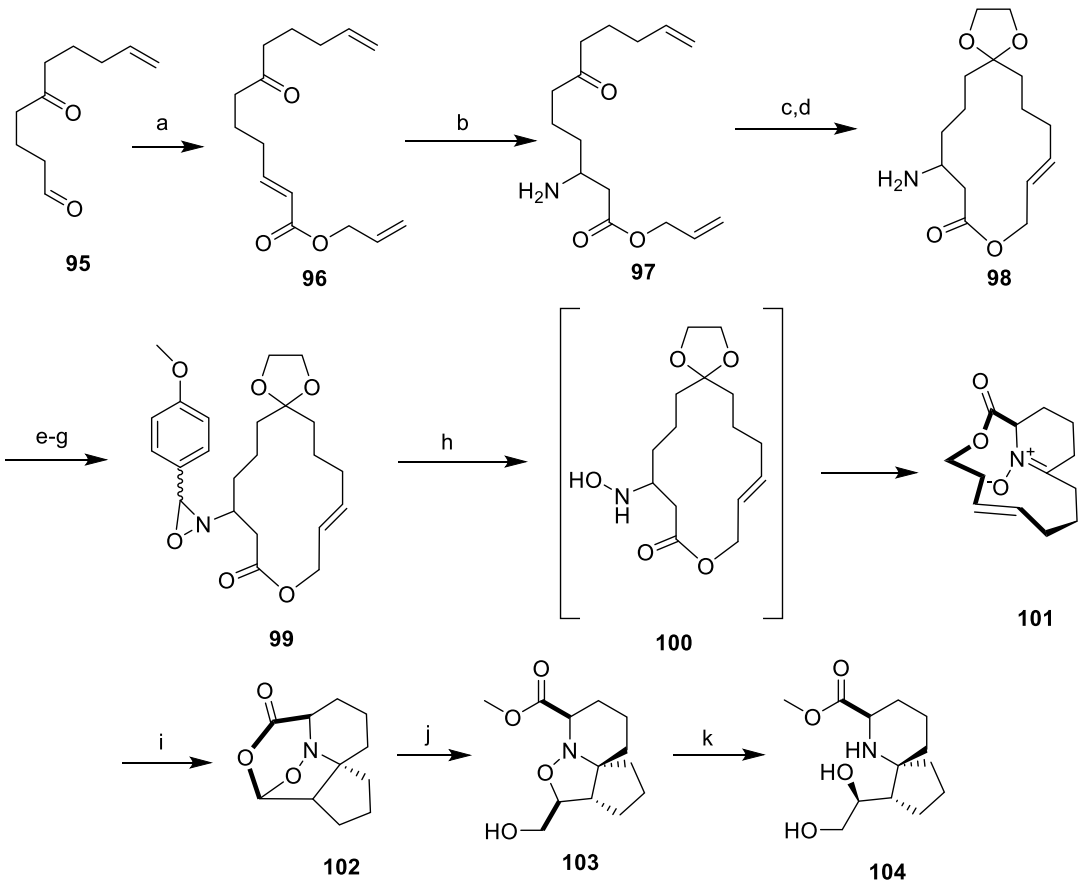
Scheme 24:- *Reagents and conditions:* e) TsOH, MeOH, 93 % f) Oxalyl chloride, DMSO, Et_3N, 97 % g) $PhP_3{=}CHCO_2Me$, CH_2Cl_2, 93 % h) Zn, AcOH, H_2O, 55 ^oC, 94 % i) $Cl_2C_6H_4$, reflux, 24 h, 84 %

2.2.3. White group

White *et al.* pursued the core structure of pinnaic acid **2** utilising an interesting approach based on the transannular addition of a nitrone to a double bond. This allowed for greater stereocontrol in accessing the desired stereochemistry in the target natural product.[30, 79]

The approach began with conversion of ketoaldehyde **95** into a *α,*β-unsaturated ester **96** *via* a Wittig reaction. Ester **96** was treated with an azide in the presence of a base to give β-azido ester **97**. Protection of the ketone followed by cyclisation in the presence of Grubbs' 2nd generation catalyst generated lactone **98**.[79]

The synthesis of the lactone presented problems, however. The reaction was reported to proceed with a poor yield of 21 %, in addition to difficulty separating the (*E))* and (*Z)* stereoisomers. Nevertheless, studies focussed on a subsequent three-step procedure to convert the lactone into a diastereoisomeric mixture of oxaziridines **99**. Hydrolysis of this mixture gave ketohydroxylamine **100**, which underwent spontaneous conversion to nitrone **101**. Upon heating in toluene under reflux, the nitrone underwent cycloaddition to give tetracycle **102** as a crystalline compound. This step is key for stereocontrol of the target molecule, as nitrone **101** is too small to allow the oxygen to pass through the ring, therefore the reaction must occur from the desired face to afford the preferred stereochemistry of the target molecule. Methanolysis followed to access isoxazolidine **103** before a final reductive ring opening, using SmI_2, afforded the target pinnaic acid core structure **104** in 11 steps (Scheme 25).[79]



Scheme 25:- *Reagents and conditions:* a) allyl (triphenylphosphoranylidene)acetate, CH_2Cl_2, 85 % b) HN_3, Et_3N, PhH, heat, 85% c) $(CH_2OSiMe_3)_2$, Me_3SiOTf, CH_2Cl_2, -78 °C, 99 % d) Grubbs I Gen, CH_2Cl_2, 21 % e) PhP f) p-$MeOC_6H_4CHO$, heat g) mCPBA, r.t, 79 % (over 3 steps) h) TsOH, Aq-MeOH i) PhMe, reflux, 64 % j) K_2CO_3, MeOH, heat, 88 % k) SmI_2, THF, 64 %

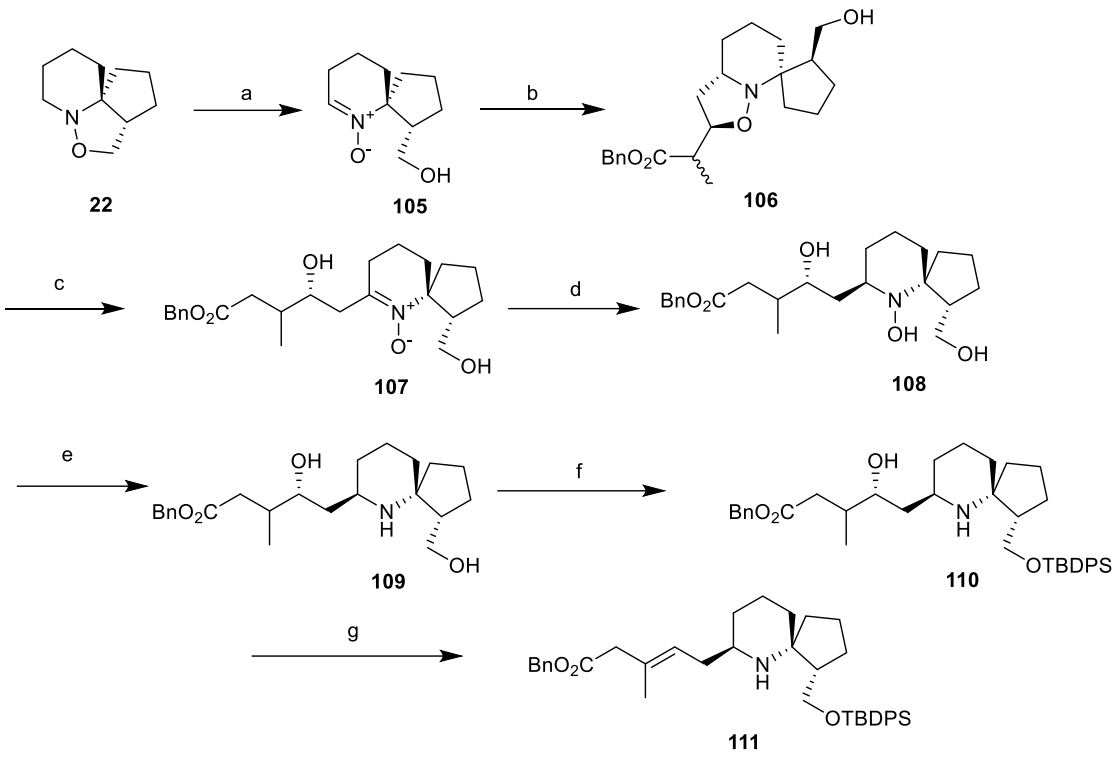
2.2.4. Caprio group

The Caprio research group has focused their work on accessing core spiro analogue structure **111** of natural product targets pinnaic acid **2** and halichlorine **3**. This has been accomplished by synthesising a key 6,5-spironitrone intermediate **105** in multi-gram quantities. [18] (Scheme *26*). This approach looks to takes advantage of the proficiency of a nitrone to undergo 1,3-dipole cycloadditions with a wide variety of dipolarophiles to provide access to a range of analogues from a common nitrone-functionalised platform. The disadvantage of this strategy is the fairly lengthy conversions of cycloadducts into spirocyclic core structures and it was envisioned that shorter routes could be developed via nucleophilic attack of Grignard species onto nitrone **105**.

Spironitrone **105** had to be accessed by first using the four-step method developed by Gossinger *et al.* to synthesise 6,5,5-azaspirocycle **22** (Scheme 11). Following this, an oxidative ring opening was achieved by slow dropwise addition of mCPBA to give key spironitrone intermediate **105** in multi-gram quantities which was enough to engage in further synthetic studies.

Synthesis then proceeded towards the pinnaic acid core structure (**2**) by first completing a cycloaddition with benzyl methacrylate to access isoxazolidine **106**. Cleavage of the isoxazolidine bond using mCPBA afforded nitrone **107** in good yield before reduction to hydroxylamine **108**. This useful tandem oxidation/reduction mechanism allowed an inversion at the C7-stereocentre to establish the correct relative stereochemistry seen in the natural pinnaic acid core structure.

A further reduction to amine **109** was achieved using zinc and a catalytic amount of indium metal. Protection of the hydroxyl group with a TBS-group afforded protected product **110**. Elimination of the remaining secondary alcohol then proceeded to give (*E*)-alkene **111** as a single diastereoisomer.(Scheme 26)[18]



Scheme 26:- . *Reagents and conditions:* a) mCPBA, CH_2Cl_2 ,0˚C to rt, 89%. b) Benzyl methacrylate, PhMe, 165 ˚C, 0.8 h , MW, 85%, c) mCPBA, CH_2Cl_2, 0 ˚C to rt, 91 %, d) $NaBH_4$, MeOH, 0 ˚C, 0.5 hrs, 68% e) cat. In, Zn, EtOH–NH_4Cl(aq) (2:1), reflux, rt, 4 hrs, 100% f) TBDPSCl, DMAP, Et_3N, 0 ˚C to rt, 89% g) MsCl, Et_3N, CH_2Cl_2, 0 ˚C to rt, 24 h (ii) NaH, THF, rt, 85% over 2 steps.

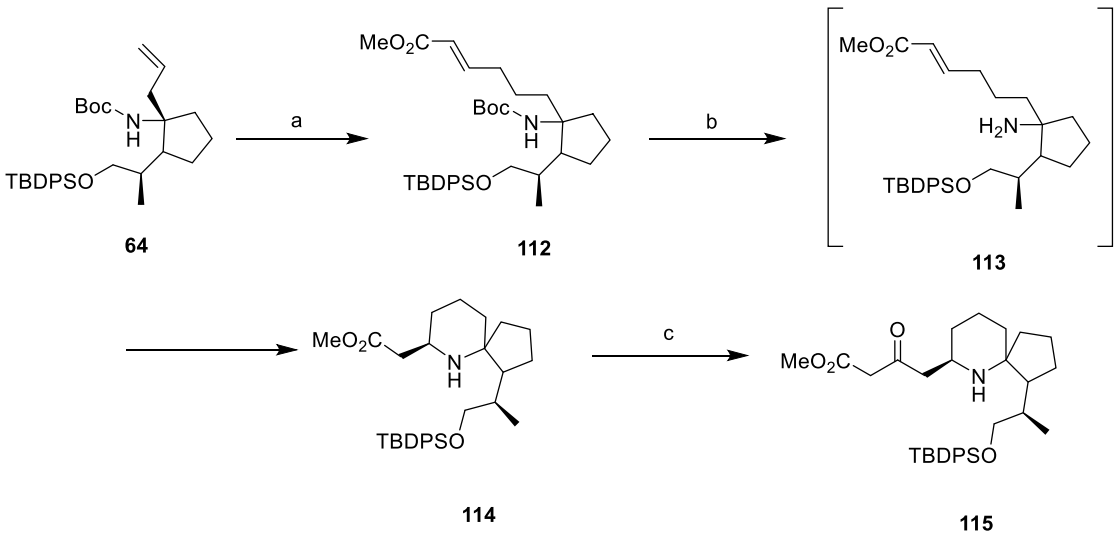
2.3 Previous synthesis towards Halichlorine

Halichlorine, in similar fashion to pinnaic acid, has been the subject of significant investigations by a number of research groups.[30, 80-82] As previously mentioned (Chapters 2.1 & 2.2) on the discussion of previous syntheses directed towards halichlorine will focus on those that utilise nitrone chemistry.

2.3.1 Danishefsky group

The first total synthesis of halichlorine **3** was reported by the Danishefsky research group (Scheme 27). Fortunately, due to the structural similarities between halichlorine **3** and pinnaic acid **2**, method development of the initial steps had occurred during the development of the total synthesis towards the latter.[30, 83, 84]

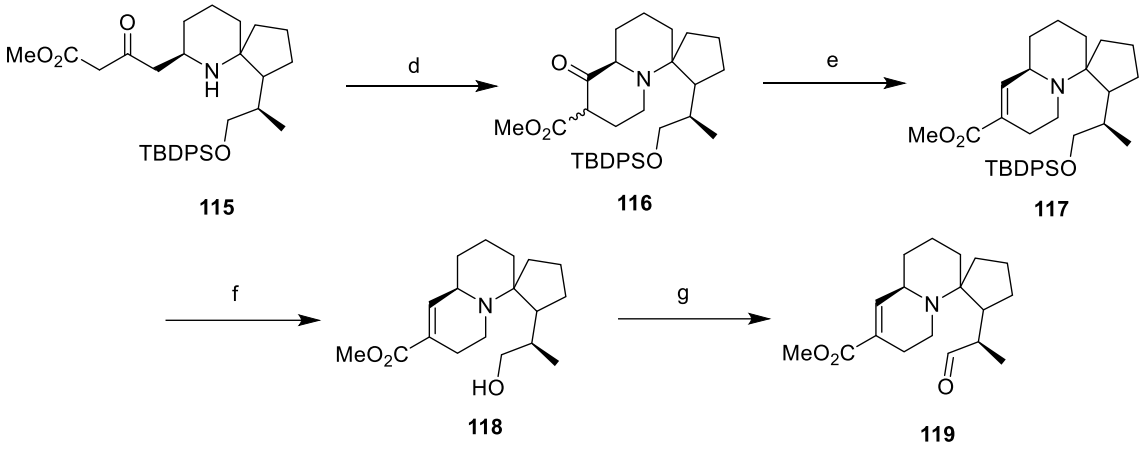
The procedure began with silyl ether **64** which was utilised in the total synthesis of pinnaic acid **2**. This time the allyl side chain was homologated *via* hydroboration followed by a Suzuki coupling with (*Z*)-iodoacrylate as the coupling partner to access intermediate **112**. A deprotection of the BOC group followed to afford amine **113** which triggered a spontaneous intramolecular Michael addition to afford tricycle **114** in good yield. With spirocycle **114** in hand, the synthetic study towards halichlorine could continue. A Claisen condensation with *t*-BuOCOMe produced β-keto ester **115**.



64 **112** **113** **114** **115**

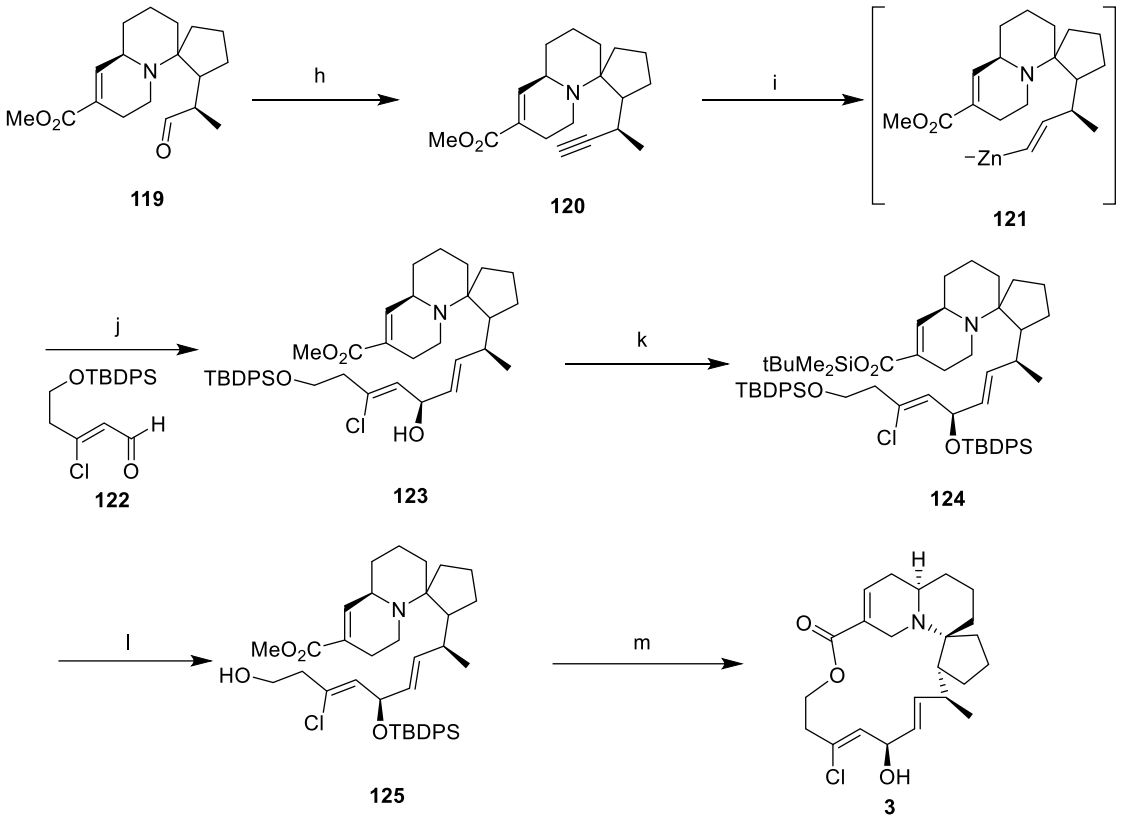
Scheme 27:- *Reagents and conditions:* a) 9-BBN, $Pd(dppf)Cl_2$, (*Z*)-iodoacrylate, Ph_3As, Cs_2CO_3, THF, DMF,H_2O b) CF_3CO_2H, CH_2Cl_2, 77 % over 3 steps c) $^tBuOCOMe$, $(Me_3Si)_2NLi$, THF, r.t, 86 %

A ring closure was then achieved *via* a Mannich reaction in the presence of formaldehyde affording the desired cyclic product **116** as a diastereoisomeric mixture. Deoxygenation with $Cp_2Zr(H)Cl$ led to formation of unsaturated ester **117**. Deprotection and subsequent oxidation of alcohol **118** afforded aldehyde **119** which proceeded with difficulty, due to the presence of the nitrogen atom, was finally accomplished using TPAP and NMO.[89,84]



Scheme 28:- *Reagents and conditions:* d) CH_2O, EtOH, 73 % e) $(Me_3Si)_2NLi$, THF, 0 °C, $Cp_2Zr(H)Cl$, r.t, 91 % f) HF.pyr, THF, 94 % g) TPAP, NMO, MeCN

As aldehyde **119** was sensitive to epimerisation during flash column chromatography, it was used in the next step without purification, where homologation with Gilbert reagent gave alkyne **120**. This alkyne substituent underwent hydrozirconation, and subsequent transmetallation with Me_2Zn, to zinc species **121**. This zinc species intermediate easily coupled with aldehyde **122,** which was prepared from Weinreb amide, and an optically pure amino alcohol **123** was formed. A selective *O*-Protection/deprotection followed protecting the alcohol group to afford silyl ether **124** before the second deprotection which afforded alcohol **125.** A subsequent macrolactonisation before a final desilylation gave halichlorine **3** in 21 steps.[83]

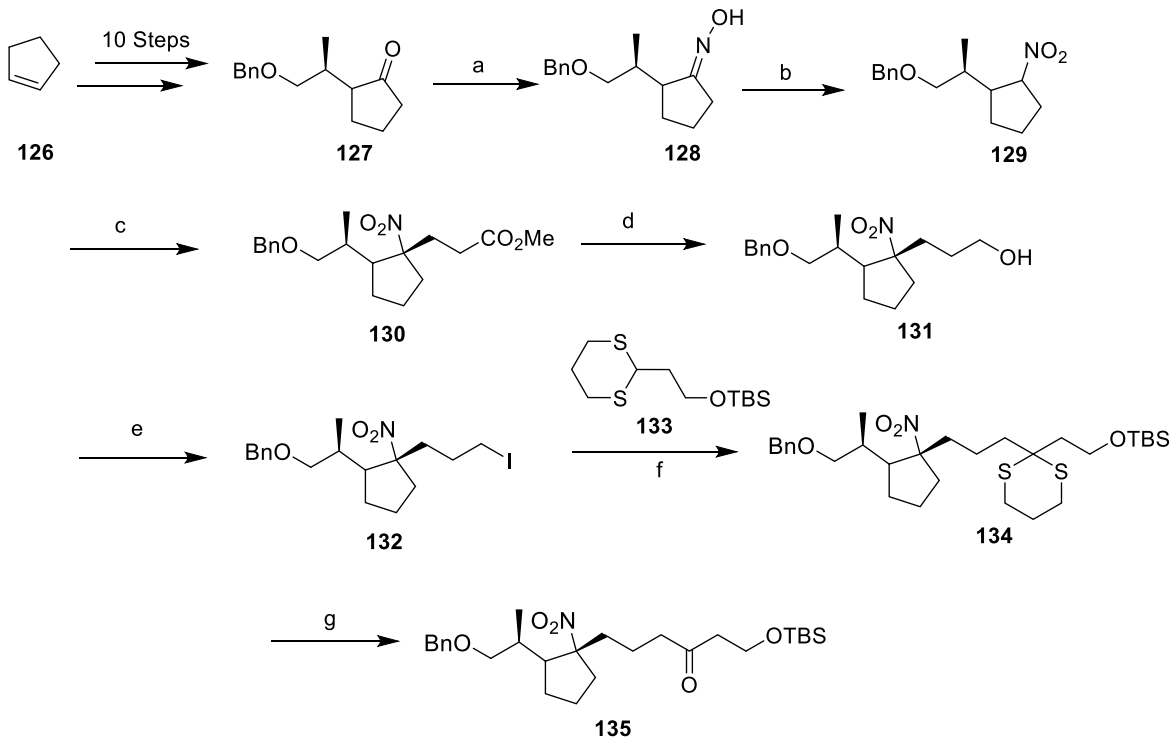


Scheme 29*:. Reagents and conditions:* h) $(MeO)_2P(O)CHN_2$, t-BuOK, THF, -78 °C, 57 % over 2 steps (i) $Cp_2Zr(H)Cl$, CH_2Cl_2, Me_2Zn, heptane, -65 °C (ii) (S)-diphenyl(1-methylpyrrolidin-2-yl)methanol, -30 °C j) **122**, DIBAH, PhMe, CH_2Cl_2, -78 °C k) *t*-$BuMe_2SiOTf$, 2,6-lutidine, CH_2Cl, -78 °C l) NH_4F, MeOH, H_2O, 66 % over 3 steps m) (i) EDCl

2.3.2. Wu group

The Wu research group devised a formal synthesis of halichlorine **3** using an interesting tandem oxidation/reduction to access the correct stereochemistry at the C7 position (Scheme *30*).[74]

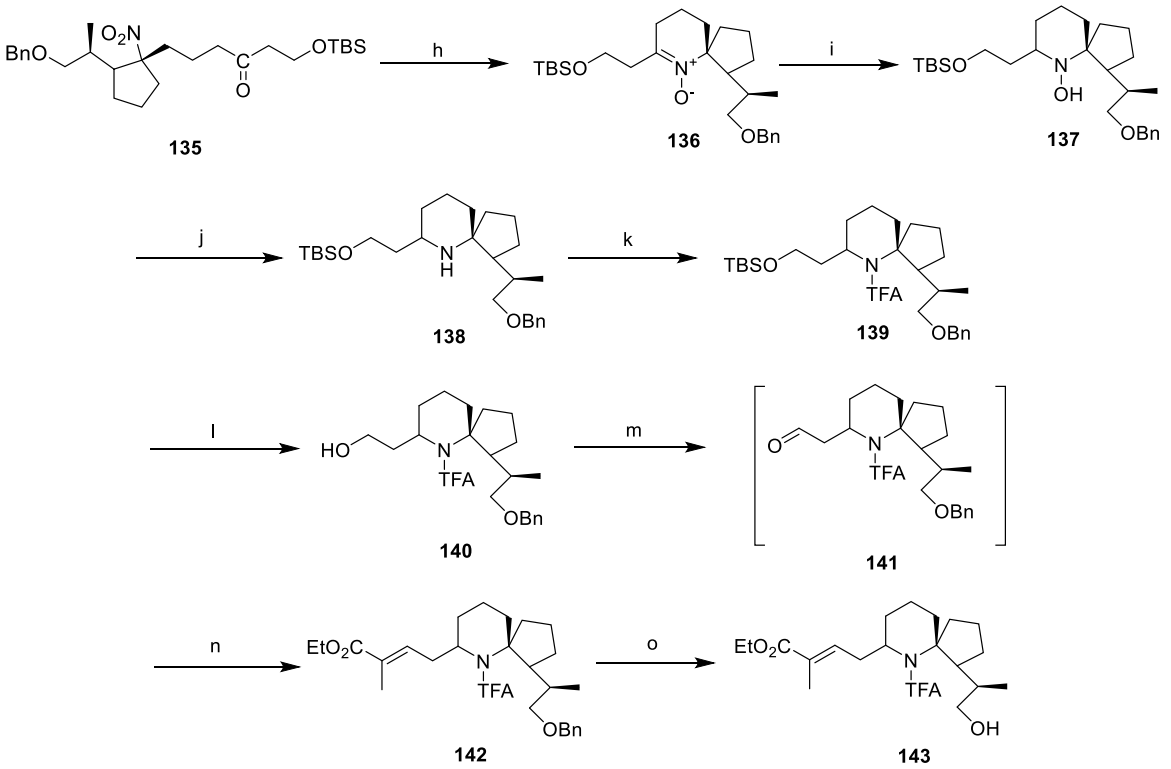
Synthesis began with the cyclopentanone **127**, synthesised from commercially available cyclopentene **126** in 10 steps. The ketone was converted to oxime **128** before subsequent transformation to nitrocyclopentane **129**. Nitroester **130** was subsequently formed *via* Michael addition to form the quaternary carbon with the desired stereochemistry. Reduction to nitro alcohol **131** using $NaBH_4$ followed forming the desired product in excellent yield. A tandem mesylation/iodination followed giving access to halogenated compound **132**. Metalation of dithiaane substrate **133** and subsequent alkylation with iodide **132** afforded **134**. The dithiane was finally removed to afford ketone **135**, a key intermediate in this formal synthesis (Scheme 30).



Scheme 30*:- Reaganets and conditions:*- a) $NH_2OH.HCl$, K_2CO_3, MeOH, r.t, 4 h, 97% b) mCPBA, Na_2HPO_4, crushed urea, MeCN, 80 °C, 76 % c) methyl acrylate, Triton B, *t*-BuOH, THF, r.t., 48 h, 95 % d) $NaBH_4$, dioxane/H_2O 1:1, r.t., 24 h, 84% e) (i) MsCl, Et_3N, DMAP, CH_2Cl_2, 0 °C, 2 h (ii) NaI, $NaHCO_3$, acetone, r.t., 24 h, 77% over two steps

f) **133**, *t*-BuLi,. HMPA, -78 °C, 40 min, 99% g) MeI, $CaCO_3$, MeCN/H_2O 4:1, r.t., 24 h, 91%

Following successful synthesis of ketone **135** in good overall yield, the nitrone was generated by reduction of the tertiary nitro group using Ni_2B and hydrazine hydrate. Reduction of the subsequent cyclic nitrone **136** with $NaBH_4$ gave access to hydroxylamine **137** with the desired stereochemistry. Enantioselectivity arises due to the hydride attacking from the less sterically hindered face, fortunately resulting in a high yield and good diastereoselectivity. Reduction to amine **138** followed using $TiCl_3$ before necessary protection of the amine with a TFA group to afford **139**. Deprotection of the protected hydroxyl group gave access to alcohol **140**. Olefination of the same side chain followed by first oxidising the alcohol to give the desired aldehyde intermediate **141**, before a subsequent Horner-Wadsworth-Emmons elongation of the carbon chain gave ethyl ester **142**. Final deprotection to give **143** completed the formal synthesis (Scheme 31).[74]



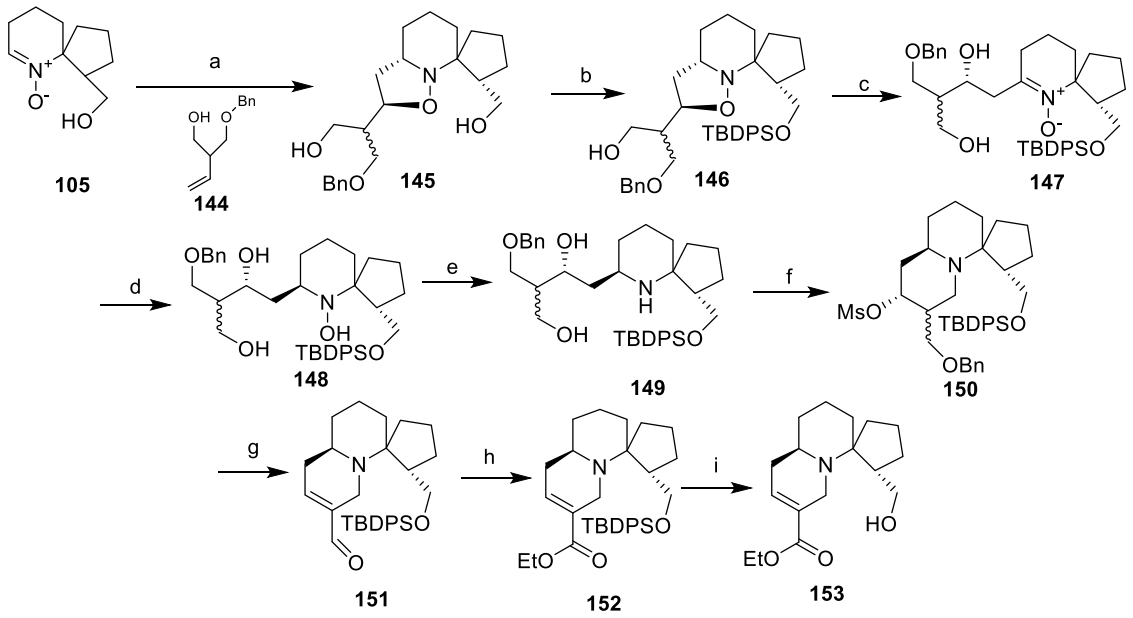
Scheme 31:- *Reagents and conditions:* h) Ni_2B, NH_2NH_2, H_2O, EtOH, reflux, 2 h, 72% i) $NaBH_4$, MeOH, 0 °C, 96% j) $TiCl_3$, NaOAc, MeOH, H_2O, r.t., 1.5 h, 79 % k) $(CF_3CO)_2O$, $(iPr)_2NEt$, CH_2ClCH_2Cl, 0 °C, 40 min, 84% (l) HF, Py, THF, r.t., 24 h, 88% m) PCC, Celite, CH_2Cl_2, r.t., 93 % n) triethyl-2-phosphonopropionate, NaH, THF, 0 °C, 90 % o) TMSI, CH_2Cl_2, r.t., 83 %

2.3.3. Caprio group

Following successful synthesis of the pinnaic acid **2** core structure, the same approach was applied towards synthesising the halichlorine core structure .[73]

Once again, spironitrone **105** was utilised in this approach. Cycloaddition with alternate dipolaraphile **144** proceeded stereoselectively to yield isoxazolidine **145** as a 1:1 mixture of diastereoisomers. The diastereomeric mixture was used in the next step without separation. Protection of the primary alcohol group with a TBDPS group afforded protected alcohol **146**.

Subsequently, a tandem oxidation/reduction sequence using mCPBA to access spironitrone **147,** followed by diastereoselective reduction using sodium borohydride gave hydroxylamine **148** with the desired relative stereochemistry. Further reduction of the hydroxylamine with zinc and catalytic amounts of indium afforded amine **149**. Cyclisation could now be attempted, using mesyl chloride in the presence of a base to give access to cyclised product **150** as a diastereoisomeric mixture. Deprotection of the OBn group was undertaken next in the presence of LiDBB. This primary alcohol was then oxidised to an aldehyde using Dess-Martin periodinane to afford the desired α,β-unsaturated aldehyde **151**. Further oxidation to the carboxylic acid using a Pinnick oxidation followed by esterification gave access to ester **152**. All that remained was to deprotect the hydroxyl group to synthesise the halichlorine core structure **153**.[73]



Scheme 32*:- Reagents and conditions:* a) PhMe, **144**, 210 °C, MW, 2 h, 78% b) TBDPSCl, DMAP, Et_3N, CH_2Cl_2, 0 °C, 1 h, 93% c) mCPBA, CH_2Cl_2, 0 °C to r.t., 1 h, 93% d) $NaBH_4$, MeOH, 0 °C, 30 min, 89%, e) In, Zn, EtOH–NH_4Cl(aq), (2 : 1), reflux, 4 h, 100% f) MsCl, Et_3N, CH_2Cl_2, 0 °C to reflux, 6 h, 99%, g) LiDBB, THF, 0 °C, 89% h) Dess–Martin periodinane, 0 °C, 1 h, 80% i) 1. $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, t-BuOH, 0 °C, 24 h, 74% 2. DCC, EtOH, DMAP, CH_2Cl_2, 0 °C, 24 h, 65%. j) TREAT·HF, NEt_3, MeCN, reflux, 4 h, 94%

2.4. Project aims

The overall aims of this project were to develop a general synthetic strategy that can be applied to prepare the core structures, and possibly a formal synthesis of HTX **1**, pinnaic acid **2** and halichlorine **3**. It was also planned to probe the feasibility of these strategies to provide facile access to a range of derivatives of the three natural product targets. As some previous literature has indicated that 6,5-azaspirodecanes as seen in halichlorine/pinnaic acid may exhibit similar, nicotinic, activity to HTX, it was also planned to utilise the chemistry developed towards these marine alkaloids to prepare a range of simple, functionalised 6,5-systems, with a view to bioevaluation for such neuroactive effects.

Initially, a route towards key spironitrone intermediates **105** had to be completed in an efficient, repeatable, and economical fashion. Multi-gram quantities of this key intermediate would provide sufficient material towards a range of more complex targets. As well as this spironitrone it is planned to access allylated derivatives of this spironitrone, of correct relative stereochemistry, as downstream key intermediates to access a range of analogues with the substitution patterns of the natural products – this also unlocks a potential formal synthesis of larger more complex targets such as analogues **156**-**158** (Figure 7).

Synthetic approaches towards alkyl esters such as **156** have been outlined previously in this thesis (see chapter 2.), It is hoped these methods can be applied in this project *via* an olefin-cross metathesis of an allylated spirocycle with a suitable alkene coupling reagent in the presence of a RCM catalyst. Use of a related, intramolecular RCM would also be the desired avenue of approach for the successful synthesis of halichlorine target **157.** (Figure 7)

The hydroxy group on allylspirocycle **155** also provides the opportunity to introduce the C11 side chain via an homologation using the Wittig reaction following the prerequisite oxidation of the hydroxy group. Indeed, methylated compound **158** is a key compound in previous total syntheses thus a strategy towards this target represents a formal synthesis of pinnaic acid.

Furthermore, total synthesis routes had to be sufficiently robust to be reproducible and efficiently afford quantities of material for not only further steps but also multiple forms of analysis in latter steps and potential bio-evaluative studies. Finally, harnessing the diverse

reactivity profile of nitrones and the presence of the hydroxy group in nitrone **105** provides an opportunity to access a wide range of spirocyclic compound libraries.

As outlined previously in this thesis (See chapter 2) multiple synthetic approaches have been attempted over the years directed towards spirocyclic bioactive compounds of interest similar to those of interest in this thesis. This approach, hoping to take advantage of the diverse reactivity profile of spironitrone **105** as well as the fact it contains the key spirocyclic framework of these target molecules, affords the added benefit of allowing the exploration of multiple avenues of investigation towards different target molecules, exploiting different techniques and approaches simultaneously.

Further avenues of interest also included the synthesis of truncated analogues of halichlorine.

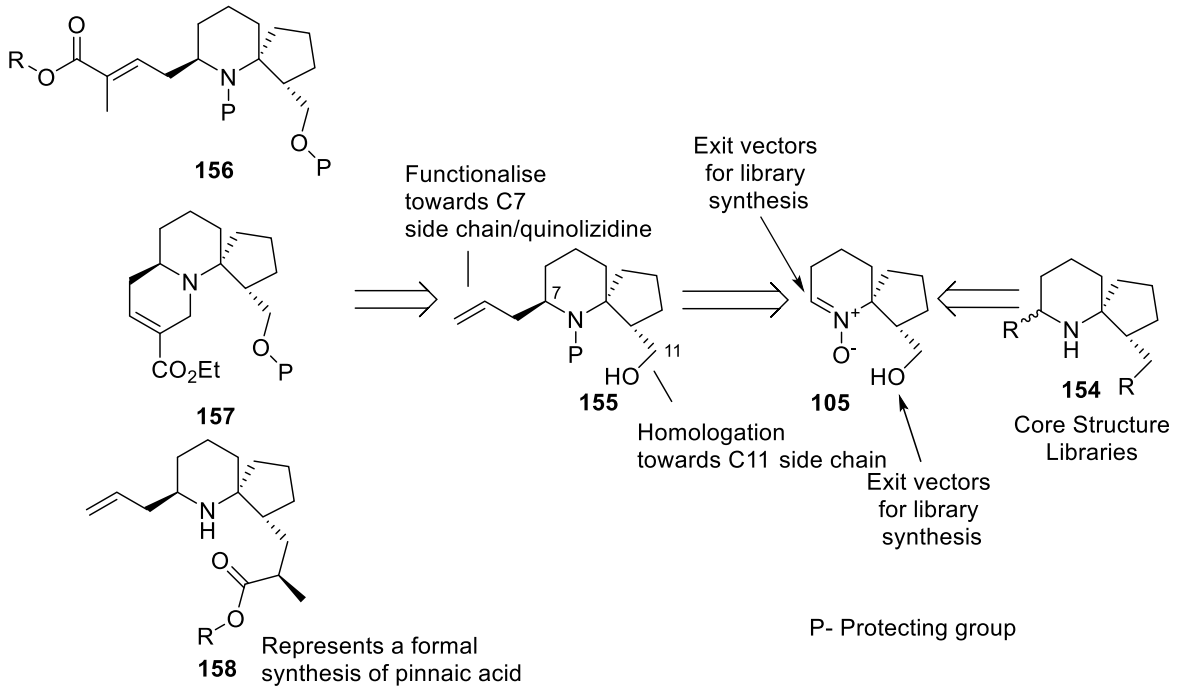


Figure 7: Retrosynthesis analysis of potential synthetic targets to which is the subject of this thesis

To further supplement the library of relatively complex molecules targeted in this project, a range of more simple azaspirocycles in the mould of **154,** which map onto structures **4**-**7**, will also be investigated in an effort to take advantage of the discovery that simple *N*-substituted azaspirocycles of this nature may exhibit nicotinic activity.

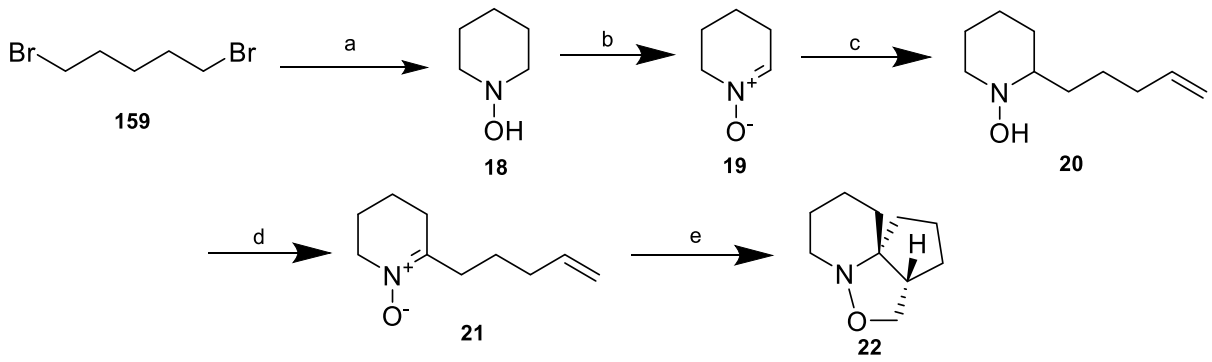
3. Synthesis of pinnaic acid core structures

The success of this research project hinged on successful synthesis of multi-gram quantities of spironitrone **105**. Synthesis of many of the targets of this thesis necessitated sufficient amounts of material to ensure efficient and economical access to suitable amounts of analogues for further structural elaboration and /or future biological evaluation.

Spirocyclic nitrone (**105**) was identified as a key intermediate by the Caprio group previously, due to its diverse reactivity profile (See Chapter 3.4) and its relative stability over protracted periods of time in comparison to the general stability of nitrones. Gossinger *et al.* devised a facile 5-step synthetic route to [6,5,5]-spirocyclic isoxazolidine **22** [37] (See Chapter 2.1), which also functions as useful precursor to compound libraries as it has the potential undergo *N*-alkylation to provide a range of novel, sp^3-rich, spirotricyclic ammonium salts and *N*-alkylated products.

3.1. Synthesis of Spironitrone

Commercially available 1,5-dibromopentane **159** underwent an S_N2 nucleophilic addition reaction with hydroxylamine hydrochloride to afford piperdin-1-ol **18** as a colourless oil. This hydroxylamine readily underwent oxidation, in the presence of mercuric oxide, to provide nitrone **19**. A subsequent Grignard addition with 4-penten-1-ylmagnesium bromide gave pentenyl-substituted adduct **20**, possessing the necessary olefinic side chain for the upcoming cycloaddition. Before this could take place however, a further, regioselective, oxidation of this hydroxylamine was required to give nitrone **21** which, again, proceeded smoothly. Finally, nitrone **21** was heated in toluene under vigorous reflux to effect a kinetic, 1,3 intramolecular dipole cycloaddition affording tricycle **22** in good yield. (Scheme 33)



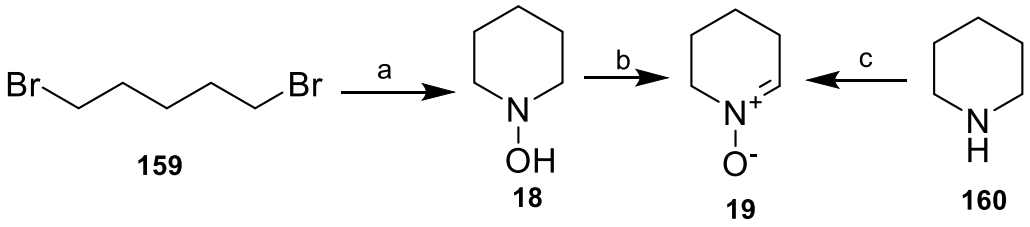
Scheme 33:- Reagents and conditions: a) $NH_2OH.HCl$, Et_3N, reflux, 65 % b) HgO, CH_2Cl_2, 0 °C, 100 % c) $BrMg(CH_2)_3CH{=}CH_2$, THF, r.t, 47 % d) HgO, CH_2Cl_2, 0 °C, 100 % e) toluene, reflux, 52 %

Optimisation of these steps had to be considered early in the project. Since large amounts of tricycle **22** had to be readily available, in consequence, large amounts of toxic mercuric oxide reagent were required, accompanied by the production of excessive amounts of toxic mercury-containing waste. Several approaches were considered to provide a safer, greener alternative.

Direct oxidation of commercially available piperidine **160** to nitrone **19** was attempted using sodium tungstate dihydrate and hydrogen peroxide as previously deployed to effect oxidation of similar derivatives by Murahasi *et al*. [50,85-87] This group have utilised this method to prepare (See chapter 1.4.3.4.) an extensive range of cyclic and acyclic nitrones.

This process is theoretically scalable to provide sufficient material for further steps.[50] However, the reaction provided a product mixture of regioisomers in our hands when used to directly oxidise piperidinol **18** which proved difficult to purify *via* column chromatography. This result is not unexpected considering literature reports of a yield of just 40 % published for this transformation.[86] Considering this was still the first step, the time and material constraints meant that while this method may have been more environmentally sustainable, it was not a feasible route if such problems persisted so early in our devised strategy.

Activated manganese (IV) oxide has been considered as an alternative to mercuric oxide-mediated oxidation of hydroxylamines,[39] so the use of this reagent was also attempted. Using MnO_2 as oxidant provided nitrone **19** smoothly and in good yield and could be deployed on a large scale to provide enough target scaffolds for future elaboration. (Scheme 34)



Scheme 34:- *Reagents and conditions:* a) $NH_2OH.HCl$, Et_3N, 4 h, reflux, 65 % b) MnO_2, CH_2Cl_2, 3 h, 0 °C, 100 % c) Na_2WO_4, 30 % aq. H_2O_2, 12 h, -5 °C, 22 %

The Caprio group has previously investigated the addition of diverse range of Grignard reagents onto nitrone **19** in pursuit of substituted spirocycles **105**.[88]

With sufficient quantities of nitrone **21** in hand, a 1,3-dipolar intramolecular cycloaddition was undertaken to synthesise multigram quantities of isoxazolidine **22**. The reaction proceeded, under kinetic conditions, in toluene under reflux to give the [6,5,5]-adduct (Figure 8). Identification of this adduct was confirmed *via* use of ^1H proton-NMR and 2-D COSY and HMQC spectra. Confirmation of the locations of the C5, C7 and C11 protons and their splitting of the CH_2 geminal protons allowed positive identification of isoxazolidine **22**.

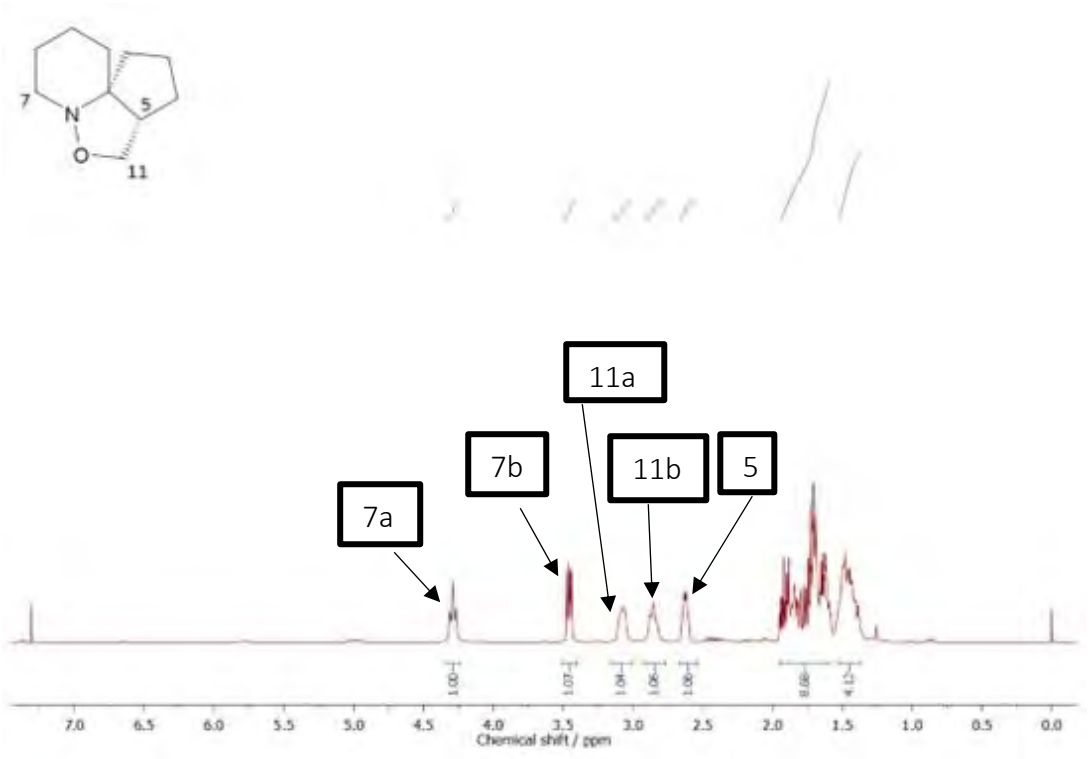
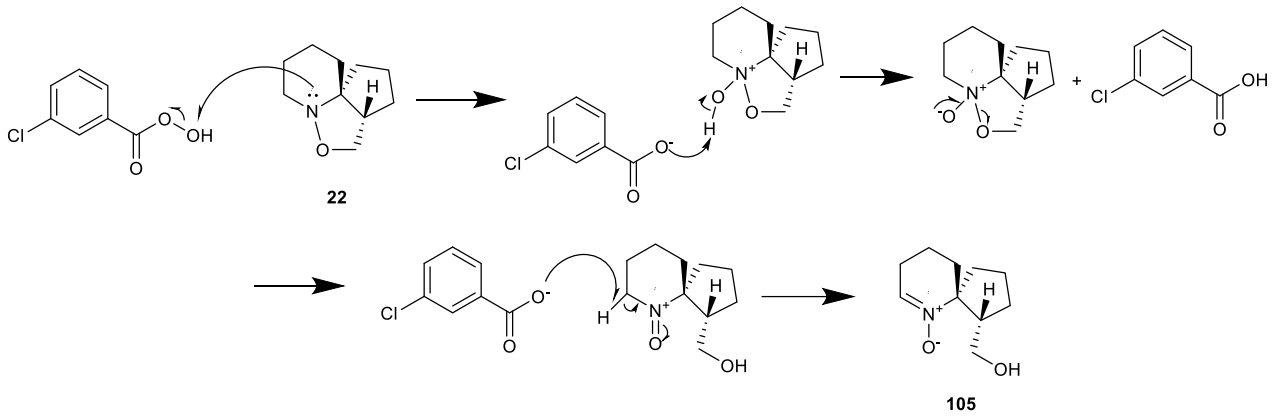


Figure 8:- ^{1}H NMR spectrum of key isoxazolidine **22** in $CDCl_3$

Investigations into accessing the thermodynamically favoured [6,6,5]-tricycle **23** were carried out later in the project as part of a synthetic strategy towards accessing the core structure of the histrionicotoxin family of alkaloids. Due to the similarities between these two isoxazolidines, it was imperative to be able to easily distinguish between the [6,5,5] and the [6,6,5] system. This is discussed in more detail in later in this thesis (See Chapter 6.1).

Tricycle **22** was then converted into key spironitrone intermediate **105** by an oxidative ring opening reaction (Scheme 35). The reaction proceeded with dropwise addition of a solution of mCPBA (ca.77%) in dichloromethane over 7 hours under inert conditions. Initial difficulties were encountered however, when the reaction mixture was not kept at a low temperature and when concentrations of benzoic acid reached high levels arising from higher rates of addition of mCPBA. Moreover, the reaction proceeded most smoothly when undertaken at larger scales of >3 g and regularly provided sufficient key intermediate to enable further, lengthy synthetic studies. The proposed mechanism of this key oxidative ring opening is shown below. (Scheme 35)



Scheme 35:- Mechanism of oxidation of isoxazolidine **22** to spironitrone **105**

Nitrone **105** could easily by identified by analysis of the ^{1}H-NMR spectrum (Figure 9), due to the triplet peak at 7.33 ppm, assigned to the C7 carbon α-to the nitrogen atom. As can be seen by comparison with the ^{1}H NMR spectrum of isoxazolidine **22** (Figure 8) there is a radical shift in several peaks following cleavage of the N-O bond and formation of the nitrone functional group.

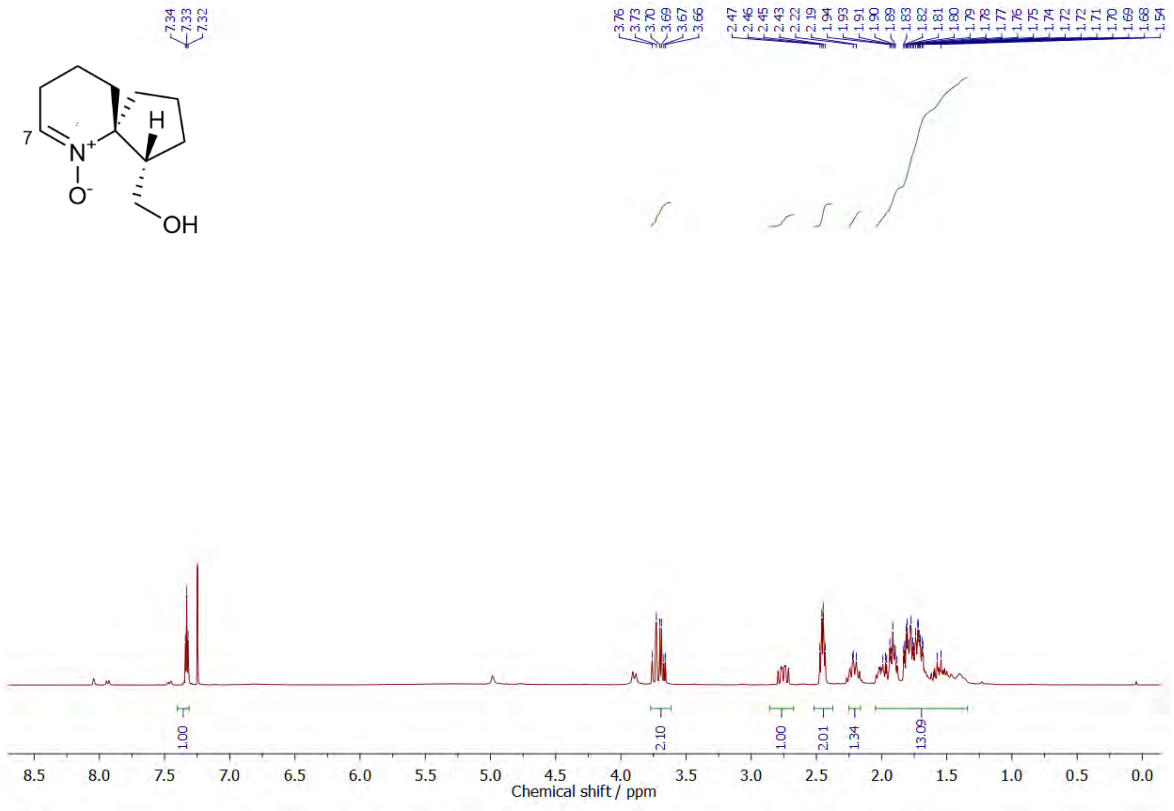
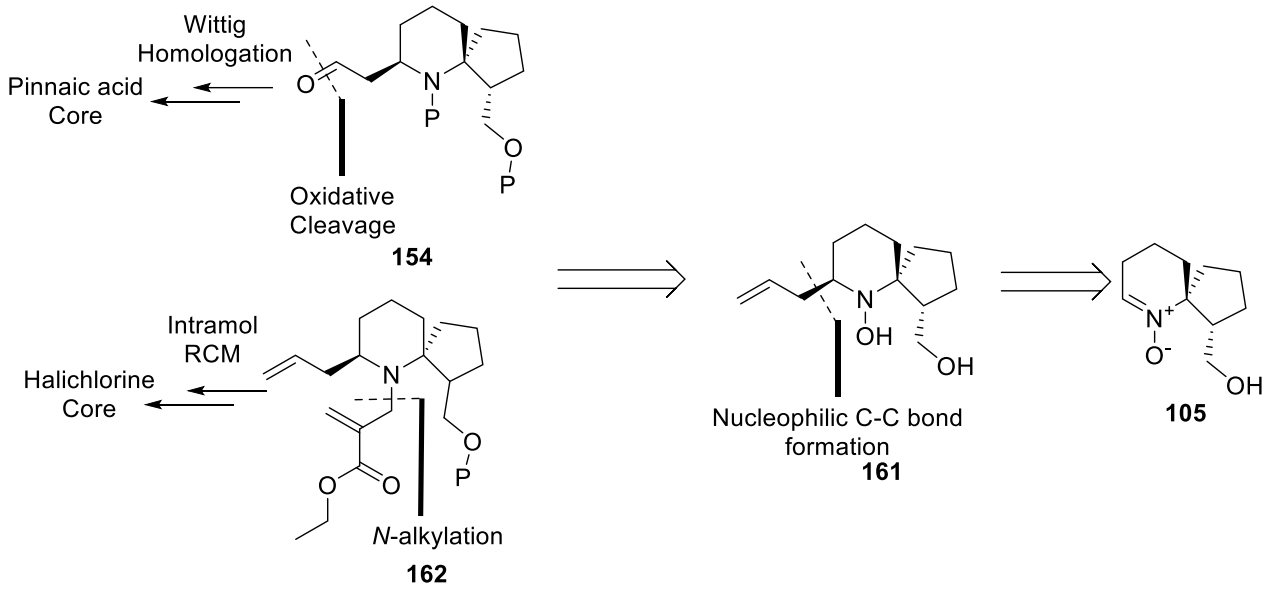


Figure 9:- ^{1}H NMR spectrum of key spironitrone intermediate **105** in $CDCl_3$

Nitrone **105** was easily purified *via* column chromatography. Presumably due to the bulkier, sterically hindered nature of this nitrone decomposition on silica gel is prevented/slowed. Securing a reproducible and reliable synthetic route to this key immediate, in multi-gram

quantities, allows investigations towards to a range of azaspirocycle-based targets and embarkation on possible formal syntheses of pinnaic acid/halichlorine.

Following the establishment of a reliable synthetic route to good quantities of key spironitrone attention shifted to investigate the feasibility of accessing the core structures of both pinnaic acid and halichlorine from this pivotal compound. It was envisioned that introduction of an allyl group α-to the nitrogen would provide a second key intermediate **161** capable of transformation to the core structures of both targets. For example, oxidative cleavage of the allyl group followed by Wittig-type homologation would lead to the core structure of pinnaic acid (**154**) while introduction of a suitably functionalised alkene-based moiety to nitrogen followed by an ring-closing metathesis (RCM) would provide facile access to the spiroquinolizidine system (**162**) found in halichlorine. (Scheme 36)



Scheme 36:- Retrosynthetic analysis of allylation of key spironitrone **105** towards core structures of both pinnaic acid and halichlorine.

3.2. Allylation of Spirocyclic Nitrone

The synthetic strategy of this project hinged on developing a similar synthetic route to both the core structures of pinnaic acid **2** and halichlorine **3** and analogues from spironitrone **105**.

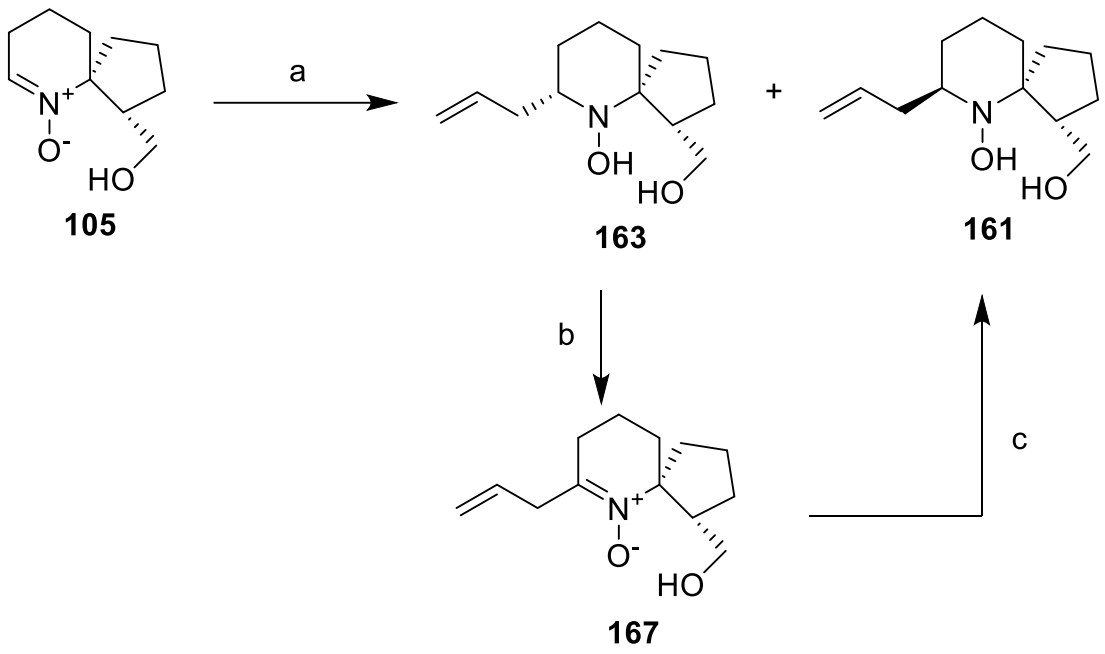
First, however, a Grignard addition at the alpha position to spironitrone **105** had to be undertaken. Previous studies by the Caprio group had revealed that a diastereomeric mixture of products would be obtained from this addition, enriched in the undesired isomer.[88] This observation necessitates inversion of the stereochemistry at the C7 position. Another potential problem was considered due to previous issues arising from isolation of water soluble, polar hydroxylamines following Grignard addition. Blackshaw *et al.* discovered the best recourse was to immediately convert the crude hydroxylamine product to the corresponding spiroamine using zinc in the presence of catalytic amounts of indium.[88]

As expected, following addition of freshly prepared allylmagnesium bromide to spironitrone **105** in anhydrous Et_2O under inert atmosphere afforded a diastereomeric mixture of hydroxylamines in an approximately 7:1 ratio (Scheme 37). As the desired stereoisomer with the same relative stereochemistry observed in pinnaic acid **2** and halichlorine **3** was the minor product, a tandem oxidation/reduction approach was devised to invert the stereochemistry of the major stereoisomer. The diastereofacial bias of nitrone **167** results in addition from the less sterically hindered face of this compound, providing the "undesired" diastereoisomer for synthesis of targets pinnaic acid/halichlorine.

It was reasoned that conversion of the diastereomeric mixture of allylated compounds (the 'desired' **161** & the 'undesired' **163**) to the corresponding nitrone followed by reduction from the 'undesired face' would invert stereochemistry. This strategy has been previously employed by the Caprio group to successfully invert stereochemistry of related structures towards the core scaffolds of pinnaic acid/halichlorine.[18]

It was envisaged that the mixture of hydroxylamines would be oxidised to the corresponding nitrone in the presence of mercuric oxide. Subsequent reduction using $NaBH_4$, would afford the desired hydroxylamine with the correct stereochemistry at the C7 position.

Oxidation with HgO followed by immediate reduction of crude material with sodium borohydride proceeded to completion affording the desired hydroxylamine **161** as a single diastereoisomer the correct stereochemistry in good yield (Scheme 37).



Scheme 37:- *Reagents and conditions:* a) $BrMgCH_2CH=CH_2$, Et_2O, r.t., 48 % b) HgO, CH_2Cl_2, r.t., 100 % c) $NaBH_4$, MeOH, r.t. 54 %

The relative stereochemistry of the hydroxylamine was confirmed by ^{1}H-NMR and 2-D NOESY NMR spectroscopy. Separation of the major hydroxylamine **163** and minor hydroxylamine **161** afforded two distinct ^{1}H NMR spectra (Figure 10 and Figure 11). In the spectrum of the major product, the peak at 3.1 ppm was assigned to 7-H at the key stereocentre using 2-D COSY and HMQC analysis.

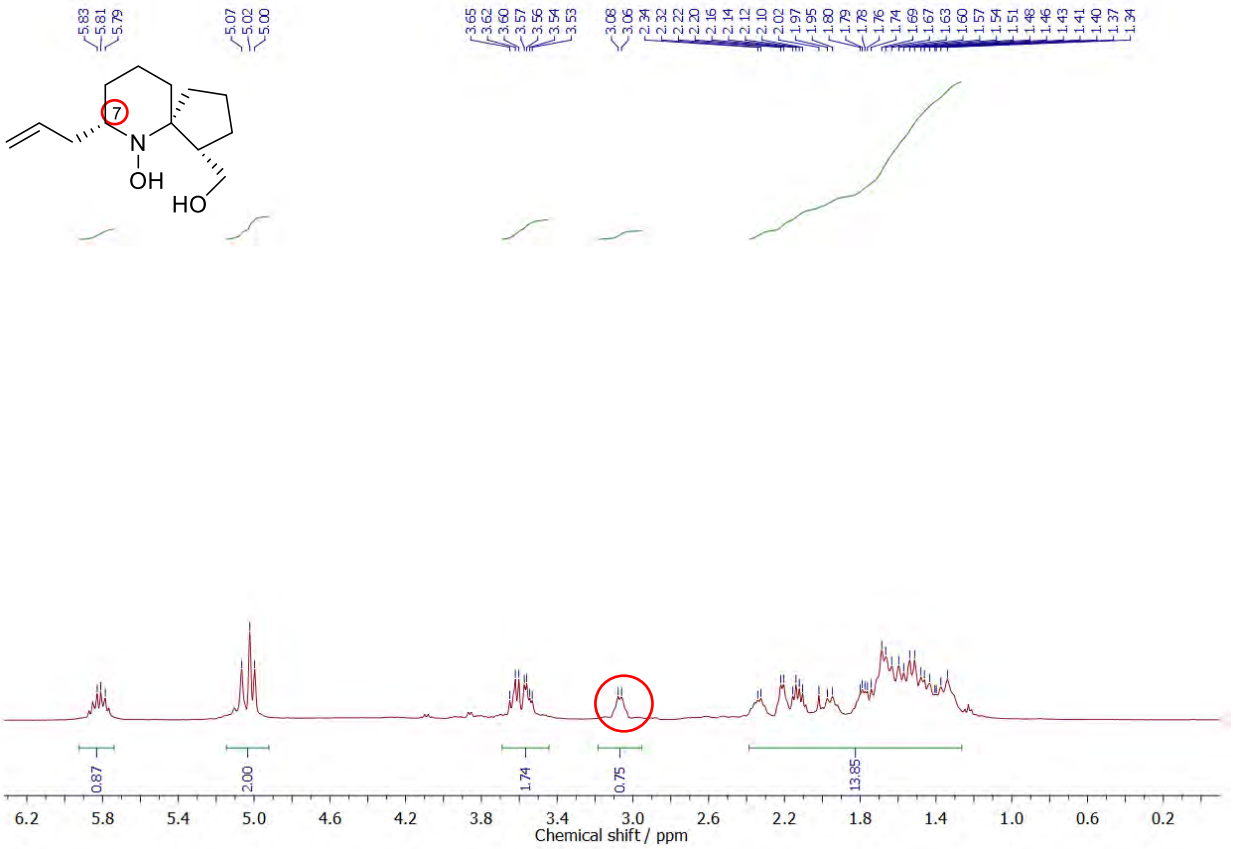


Figure 10:- ^{1}H-NMR spectrum of hydroxylamine **163** in $CDCl_3$.

The proton at the 7-H stereocentre in the minor hydroxylamine **161** was assigned to the peak at 2.6 ppm, again confirmed by COSY and HMQC analysis.

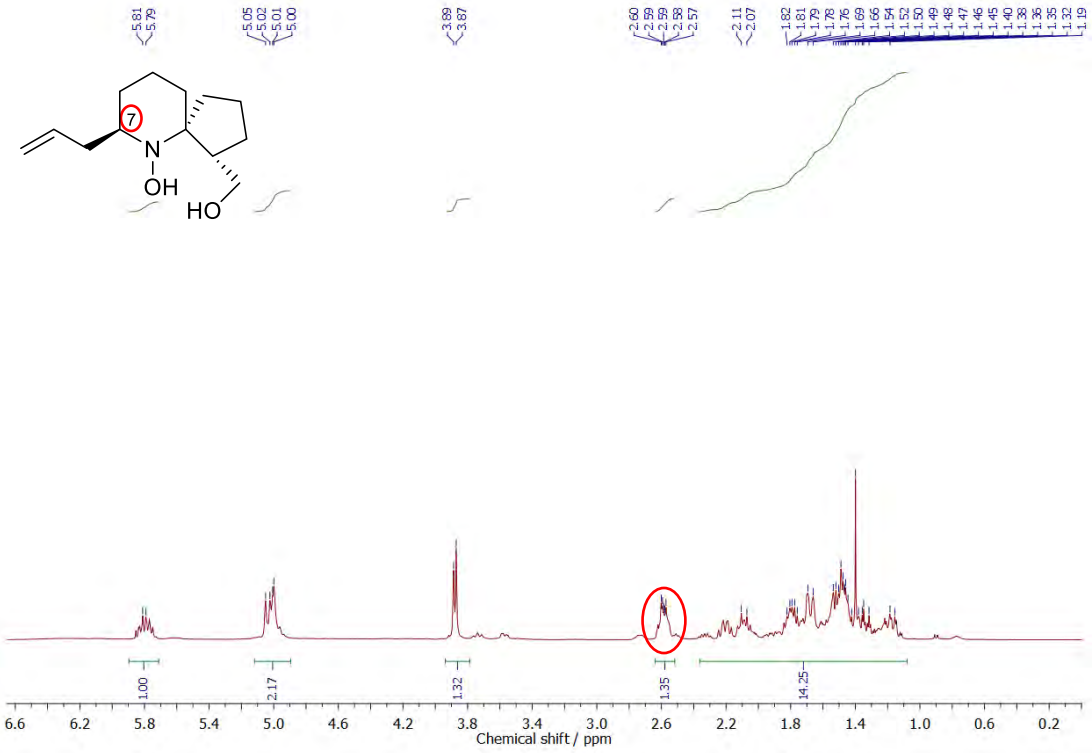


Figure 11:- ^{1}H-NMR spectrum of hydroxylamine **161** in $CDCl_3$.

Key proton assignments for the two hydroxylamines were confirmed by using 2-D NOESY spectra to correctly assign the proton at C7 in diastereoisomers **161** and **163**. (Figure 12) A coupling was expecting to be observed between the peaks at the C7 and C2 positions in line with the topological structure of **161**.

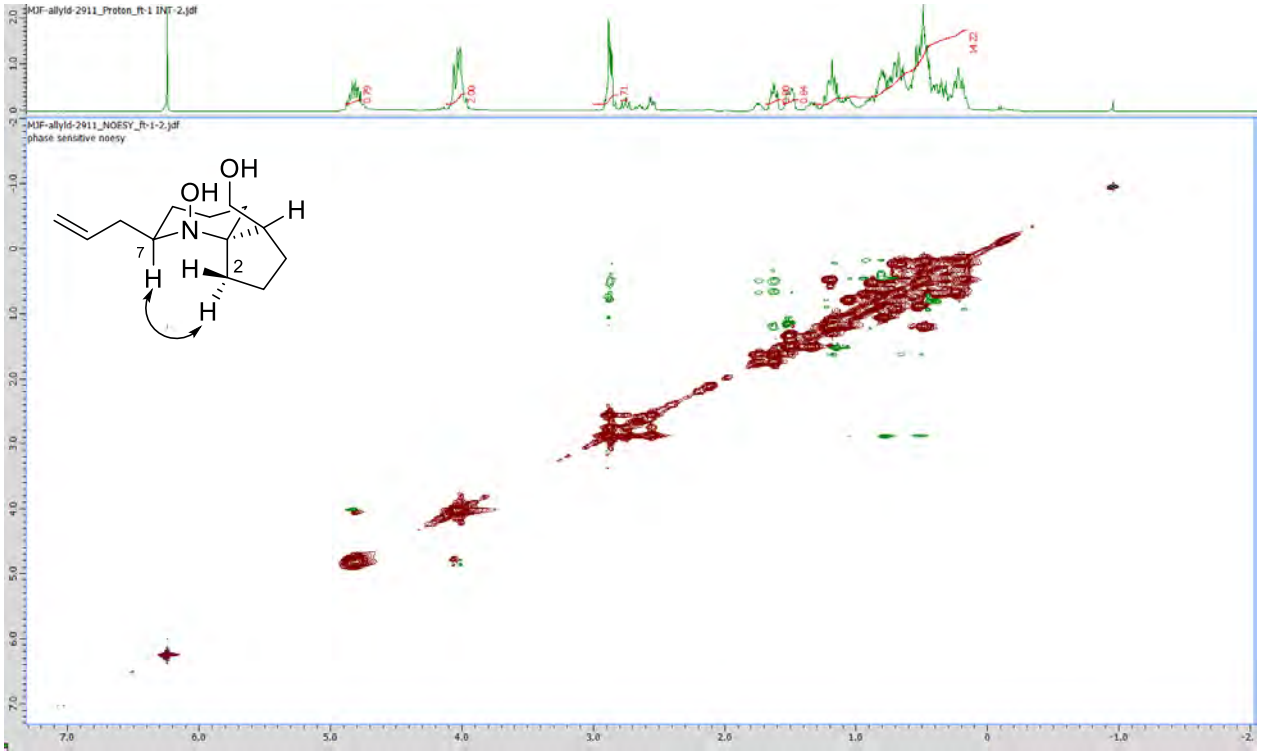


Figure 12:- 2-D NOESY spectra of hydroxylamine **161** in $CDCl_3$.

3.3. Investigation of the use of N,O-protecting Groups

With sufficient quantities of the allyl analogue **161**/**163** in hand, the next goal was to reduce the hydroxylamine group before subsequent protection of the secondary amine formed. This set of transformations would then be followed by protection of the hydroxy group at the C11 position. Techniques were first investigated using the undesired, major hydroxylamine **163** as part of method development, before applying successful methodology to the minor hydroxylamine **161** owing to the relatively larger amounts of the undesired stereoisomer to hand. As both routes proceeded similarly, both will be reported simultaneously in this section.

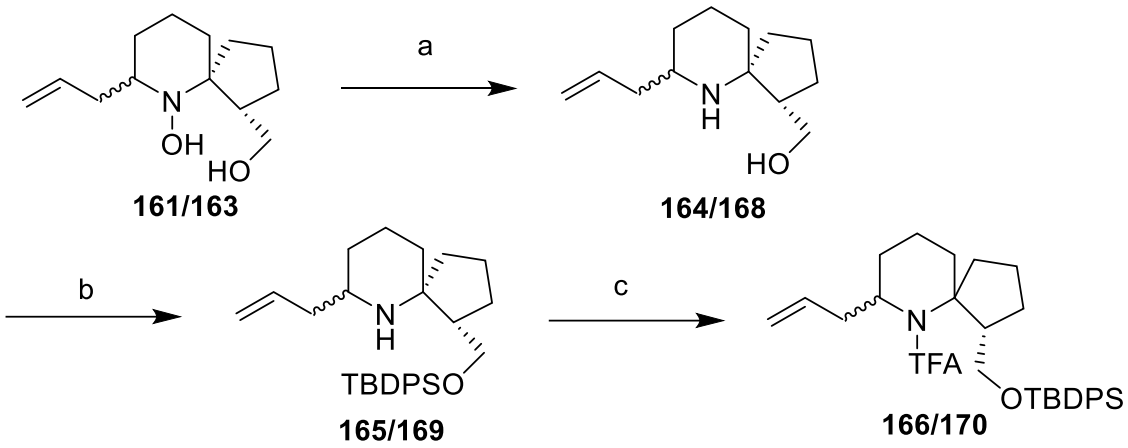
The next step was to reduce the hydroxylamine group to an amine. Zinc metal in acetic acid has been used numerous times in the pursuit of HTX **1**, pinnaic acid **2** and halichlorine **3** when cleaving N-O bonds found in isoxazolidines.[13, 30] However it is less common for this technique to be utilised in the conversion of hydroxylamine to amine, however this technique has been utilised to great effect by this research group.[18, 88]

A reduction of the hydroxylamine **161**/**163** occurred readily in aq. acetic acid using zinc metal as reductant. Reaction monitoring *via* TLC analysis confirmed the disappearance of hydroxylamine **161**/**163** to secondary amine **164**/**168,** with the amine identified as a much more polar spot, with very low Rf meaning this particular step was easy to monitor. Fortunately, ^{1}H-NMR spectral analysis revealed that a pure product formed without the need for column chromatography, avoiding the potentially difficult task of exposing the incredibly polar secondary amine to silica gel potentially leading to product loss.

It was next planned to protect the hydroxy group of **164**/**168** as a TBDPS-ether prior to *N*-protection. This protecting group is sufficiently robust to withstand a wide range of conditions yet readily removed and has been used to good effect by a number of research groups when pursuing derivatives of natural product alkaloid derivatives such as during the studies by the Caprio and Danishefsky groups.[18, 88, 89] (Scheme 18 & Scheme 26) This reaction proceeded without incident providing silyl ether in **165**/**169** good yield.

Danishefsky *et al.* had previously discussed the use of appropriate protecting groups for the secondary amine of the azaspirodecane framework. While BOC-protection would seem an obvious choice, this research group, reported difficulty in protecting with this moiety possibly due to unforeseen steric encumbrance around the nitrogen atom in azaspirocycles

of this type. Consequently, success was achieved using a trifluouroacetyl group.[89] Using trifluoroacetic anhydride and Hunig's base, acetylated amine **166/170** was successfully synthesised. (Scheme 38). It proved difficult to use ^{1}H-NMR spectral analysis to determine if the synthesis was successful as there were no additional protons present in the product compared to starting material.



Scheme 38:- *Reagents and conditions:* a) Zn, 33 % Aq. AcOH, 70 °C, 87 % b) TBDPSCl, DMAP, Et_3N, 0 °C, 88 % c) TFAA, Hunig's base, 0 °C, 45 %

However, TLC analysis showed a far less polar product was present compared to silyl ether starting material. Notably the C7 proton shifted considerably downfield to 3.75 ppm, due no doubt to the introduction of the trifluouroactyl group to the neighbouring N atom, creating a deshielding effect on the C7 proton (Figure 13).

In addition to this, a peak in ^{19}F-NMR spectrum was present as well as additional peaks in the ^{13}C-NMR spectrum indicating the presence of a carbonyl and CF_3 groups.

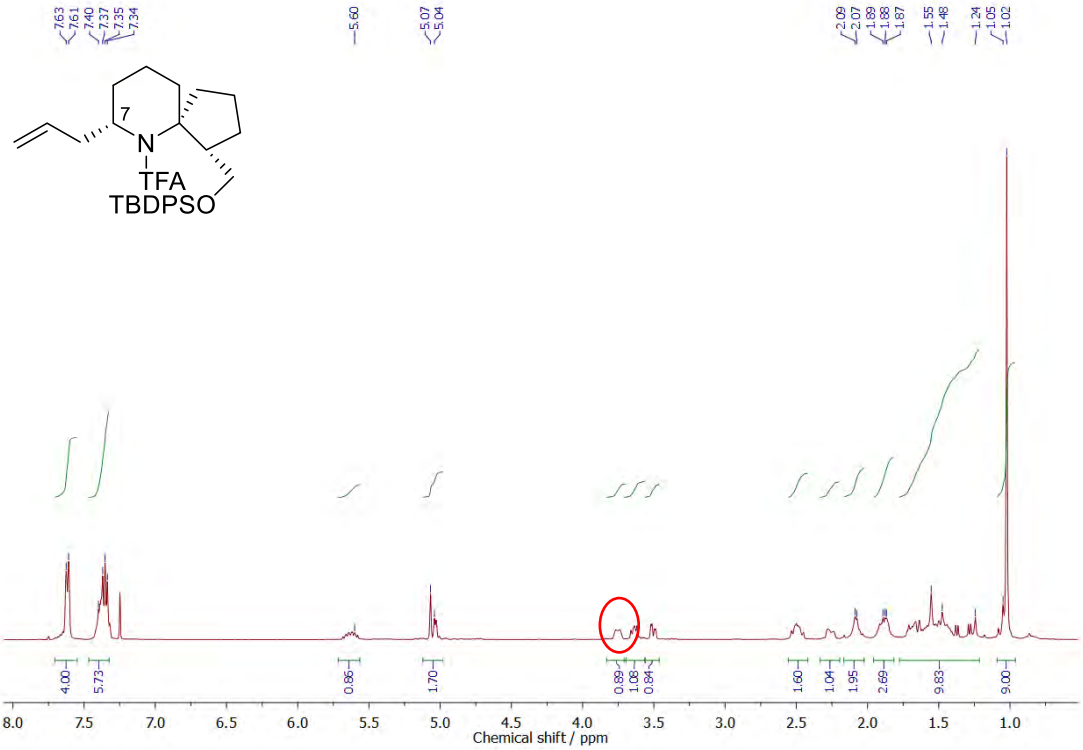
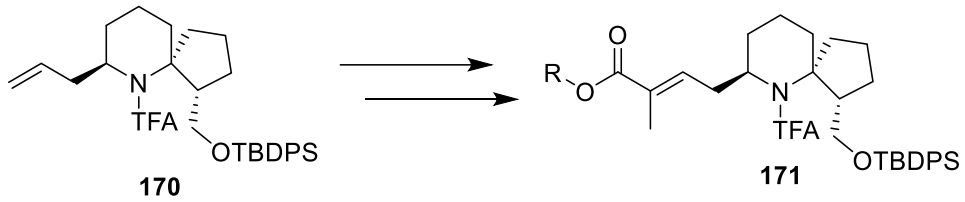


Figure 13:- ^{1}H NMR spectrum of tertiary amine **166** in $CDCl_3$.

3.4. Elaboration Towards the Core Structure of Pinnaic Acid

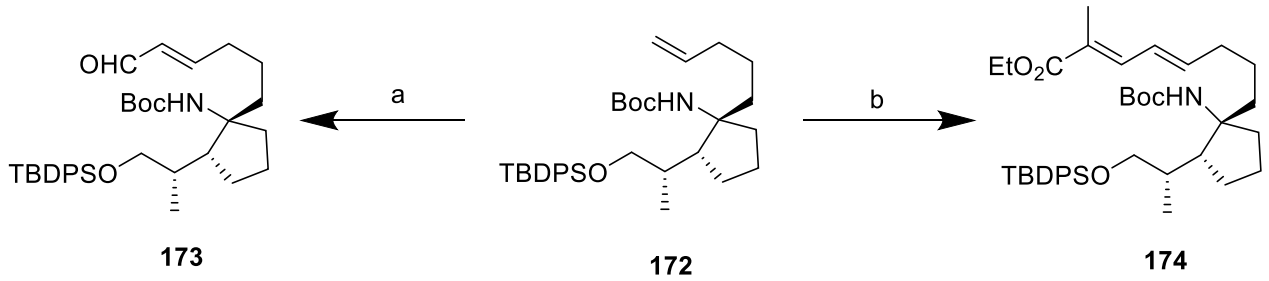
Following successful synthesis of tertiary amine (**170**), the final steps towards the pinnaic acid **2** core structure could be undertaken utilising the allyl side chain on acetylated amine as a handle for further elaboration (**171**) (Scheme 39).



Scheme 39:- General scheme towards target pinnaic acid general core structure target.

3.4.1. Olefin cross-metathesis-based approach to the pinnaic acid side chain

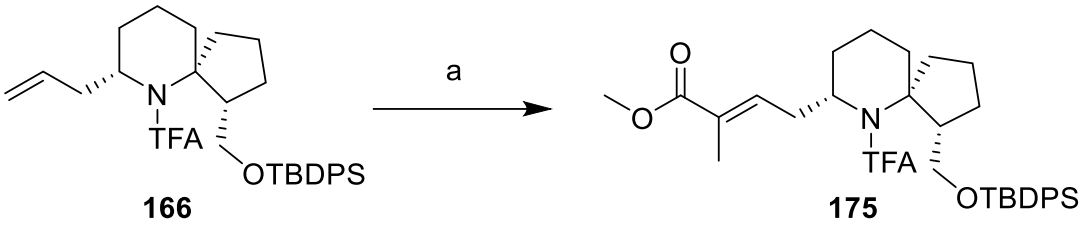
The first approach attempted to introduce the pinnaic acid enoate side chain by a simple, one-step cross metathesis with Grubbs' generation-(II) catalyst and methyl acrylate. Olefin metathesis in the presence of ruthenium containing catalytic reagents have been very well studied as a in carbon-carbon bond forming strategy.[90] Furthermore, Martin *et al.* reported great success in the use of olefin cross-metatheses using Grubbs (II) catalyst during their approaches towards formal syntheses of pinnaic acid and halichlorine, by utilising protected allyl motif **172** they successfully elongated the allyl chain to access spiroaldehyde **173** and ethyl ester **174**.[91]



Scheme 40:- Synthesis of analogues of pinnaic acid utilising a cross-methathesis approach by Martin *et al*. *Reagents and conditions:* a) (*E*)-But-2-enal, *Grubbs* (II) gen, CH_2Cl_2, 89 % b) ethyl (2*E*,4*E*)-2-methylhexa-2,4-dienoate, *Grubbs* (II) gen., CH_2Cl_2, (dr =10:1)

Initial investigations were carried out using the rather larger quantities of undesired isomer **166** available, to optimise reaction conditions before synthesis of core structure with desired stereochemistry was attempted.

Tertiary amine **166** was heated under reflux in the presence of Grubbs' catalyst and methyl acrylate to initiate an olefin cross-metathesis. Methyl ester **175** was synthesised on 25 mg scale in 65 % yield (Scheme 41).



Scheme 41:- Reagents and conditions: a) Methyl Acrylate, Grubbs' 2nd gen, toluene, reflux, 64 %

This initial success led to optimism that this route could be adapted towards more topologically advanced core structures – including a total/formal synthesis of pinnaic acid **2**.

However, subsequent attempts to scale-up the reaction to levels required to enable a multi-step synthesis did not yield successful results, with yields of ester **175** plummeting to less than 10 % on a 250 mg scale. It is believed this was due to the methyl acrylate, in much high concentrations, beginning to polymerise when heated under reflux. An increase in methyl acrylate added was necessary, due to the reaction not proceeding as readily as observed at lower-scales. This would lead to an increase in molar concentration which may have also played a factor in this reaction not proceeding correctly at this new scale.

This hypothesis was supported by the formation of a layer of plastic polymer on the reaction vessel. Repeated attempts to circumvent this unfortunate occurrence failed. Attempts at using less methyl acrylate reagent resulted in the reaction not proceeding to completion despite extended reaction time at reflux temperatures. Further investigation will be required to investigate how dilution factors effects the outcome of this synthesis, leading to a more successful outcome at smaller scales. It may be that more highly dilute reaction mixtures would mitigate the self-polymerisation of acrylate. This synthetic route was deemed not sustainable due to the high loss of material when attempted on larger scales. Further study would be beneficial to investigate if any alternative catalysts such as Hoveyda-Grubbs catalyst would prove beneficial.

3.4.2. Introduction of the pinnaic acid side chain *via* Wittig homologation

An alternative synthetic route was investigated to introduce the unsaturated side chain of pinnaic acid **2**. As sufficient amounts of amide **170** with the correct stereochemistry had been synthesised with the procedure optimised, studies could be carried out using the correct stereochemistry.

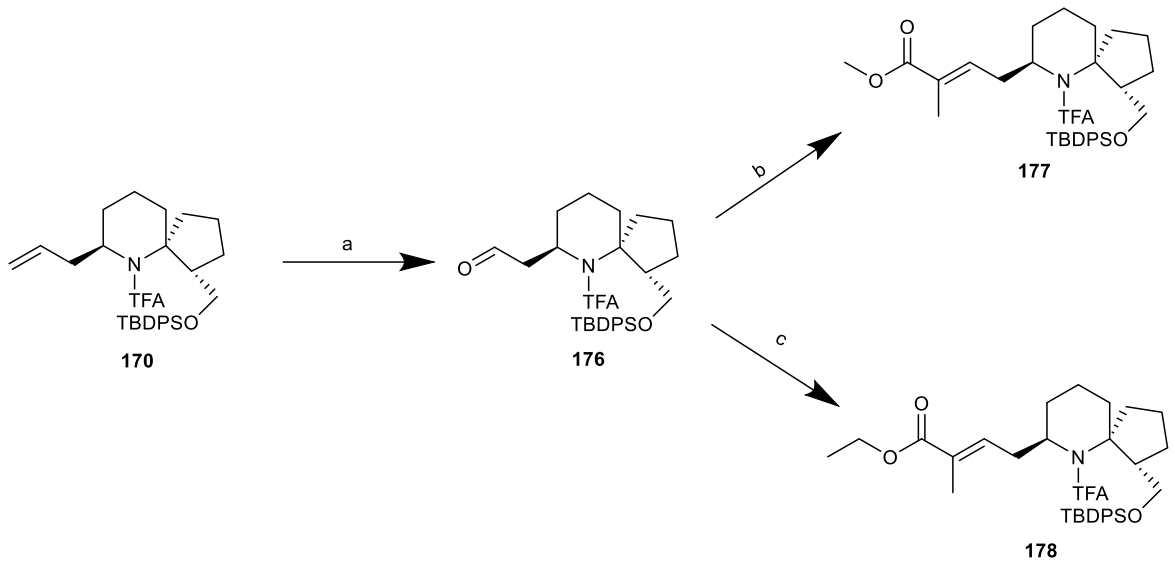
The goal was to access an aldehyde group in the place of the olefin of the allyl side chain *via* an oxidative cleavage. Stockman *et al.* reported using this oxidation method in their pursuit of pinnaic acid and halichlorine core structures.[92] This key intermediate could then undergo reactions with various phosphonate carbanions.

First, a two-step, tandem Lemieux-Johnson oxidation was attempted to synthesise the corresponding diol using OsO_4 as oxidant. This diol would then undergo cleavage in the presence of sodium periodate to form the desired aldehyde **176**. The two-step oxidation proceeded in good yield (74 %).

In addition to the good yield observed, aldehyde intermediate **176** demonstrated remarkable stability when stored at 0 ^{o}C, especially when considering the general instability of aldehydes in general. Analysis of a sample of following many months in storage using ^{1}H NMR spectroscopy and LC-MS revealed minimal degradation.

A Horner-Wadsworth-Emmons reaction with aldehyde **176** was completed with methyl 2-triphenylphosphonoanylidene propanoate to synthesise methyl ester **177**.

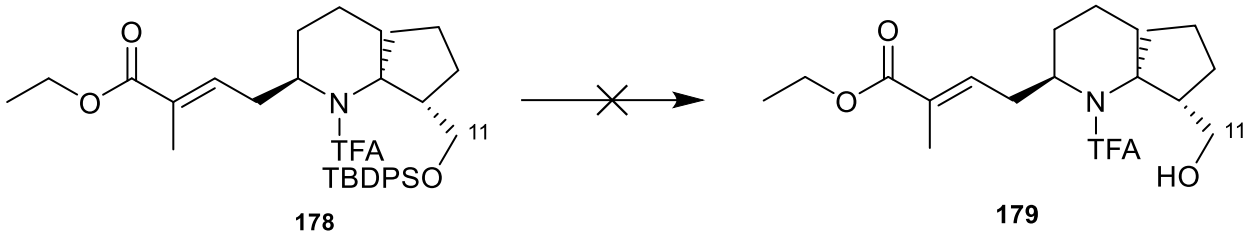
The ethyl ester (**178**) was next targeted, again, starting from aldehyde **176**. A Wittig reaction was undertaken with triethyl 2-phosphonopropanonate, in the presence of sodium hydride, to afford ethyl ester **178,** again, accessed in good yield (Scheme 42).



Scheme 42:- *Reagents and conditions:* a) (i) OsO_4, NMO, $(CH_3)_2CO$, r.t. (ii) $NaIO_4$, MeOH, 0 °C, 74 % over two steps b) Methyl 2-triphenylphosphoranylidene propanoate, CH_2Cl_2, r.t., 75 % c) NaH, triethyl 2-phosphonopropanoate, r.t., 90 %

The use of two separate techniques to access alkyl esters, both successful in achieving the desired target molecule, demonstrated not only the versatility of the aldehyde intermediate **176** but also the immense potential in synthesising this intermediate in multi-gram quantities for future studies. This was in addition to the stability of the molecule when stored at 0 ^{o}C.

With quantities of ethyl ester **178** in hand, further elaboration towards pinnaic acid **2** could now be investigated. A formal/total synthesis requires manipulation of the hydroxy group at C11. This required deprotection of the silyl ether at this position. Despite this step initially being viewed as relatively standard, a complex mixture of products, all running incredibly close to the baseline, when analysed by TLC was observed (Scheme 43), with an Rf much lower than anticipated for the target alcohol **179**. Initially, the lack of success of this step could not be discerned and an answer would not be obtained until further investigations later in the project (See section 4.2.2.).



Scheme 43:- Attempted deprotection of C11 hydroxyl group. *Reagents and conditions:- a)* TREAT.HF, TEA, acetonitrile, reflux.

Overall in this chapter, the main objective was to not only synthesise spironitrone **105** in multi-gram quantities, but to also bring it through in a timely manner and with an economical use of materials. This key intermediate was synthesised in multi-gram quantities in improved yields than those obtained previously in the research group, while also using a more ecologically sustainable synthetic route that by-passes any need for highly toxic mercuric (II) oxide.

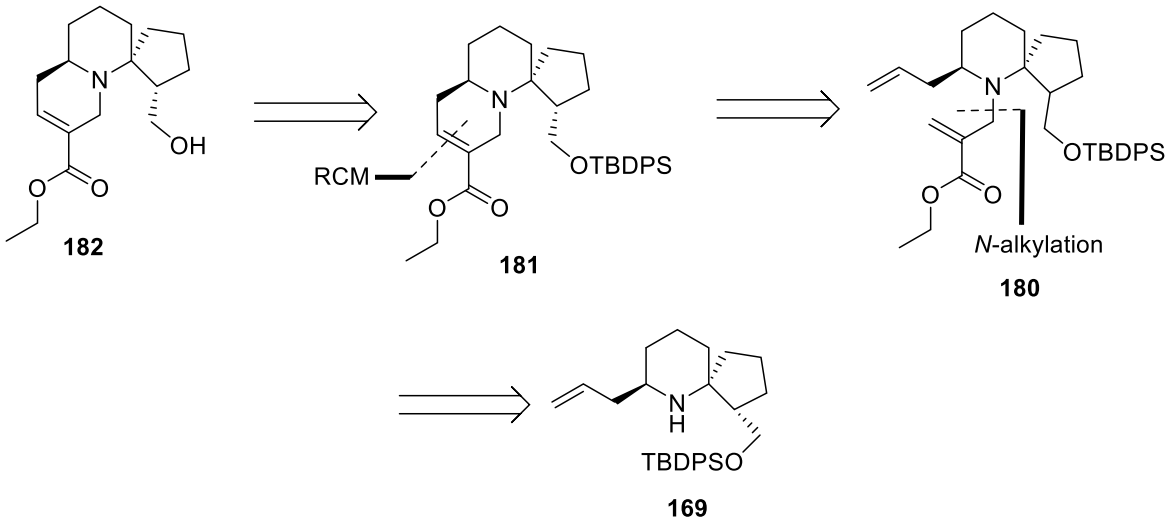
With significant quantities of spironitrone **105** in hand, synthesis continued towards target molecule, ethyl ester **178** in a diastereoselective manner. Previous studies by this group were unable to deliver hydroxylamine **161** in sufficient quantities for further investigations towards larger more complex analogues of pinnaic acid **2**. With this diastereoselective approach, not only could synthesis continue towards ethyl ester **178** but also provide a framework for further investigations not only for later chapters in this thesis but also for future investigations into these natural products of interest.

With the objective of synthesising the ethyl ester, the initial target outlined in section 2.4, successfully, sights were set even higher towards building on this framework. While this objective could not continue in this project due to time constraints, with the only method attempted being outline above (Scheme 43), a foundation has been set for further investigations towards even more complex molecules of interest.

4. Synthesis of the tricyclic core structure of halichlorine

Attention now turned to developing a synthetic route to the tricyclic core structure of halichlorine. Synthetic investigations in this chapter fortunately were aided by the structural similarities previously discussed in this thesis between pinnaic acid **2** and halichlorine **3**, resulting in synthetic routes towards each target being conducted simultaneously.

The goal was to synthesise a suitable diene to enable formation of the spiroquinolizidine core structure of halichlorine *via* ring-closing metathesis as previously reported by Kibayashi *et al*. in their formal synthesis of halichlorine.[93] In a similar fashion to the pinnaic acid core, the synthetic procedure towards halichlorine utilised silyl ether **169** as a key intermediate. (Scheme 38) Following successful synthesis of the silyl ether in multi-gram quantities, an alkylation of the free secondary amine could be attempted using ethyl 2-(bromomethyl)propenoate, in the presence of potassium carbonate, followed by ring closing-metathesis in the presence of Grubbs (II) catalyst. (Scheme 44)

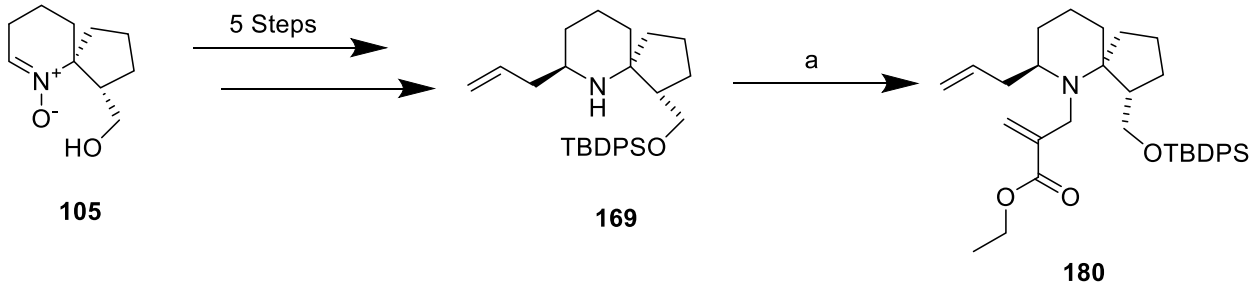


Scheme 44:- Retrosynthesis of halichlorine core structure target motif

Fortunately there was literature precedent for the use of ring-closing metathesis in the pursuit of spirocyclic natural product, with White *et al.* reporting success as previously discussed in this thesis.[79] (Scheme 25)

4.1. Synthesis of halichlorine core structure

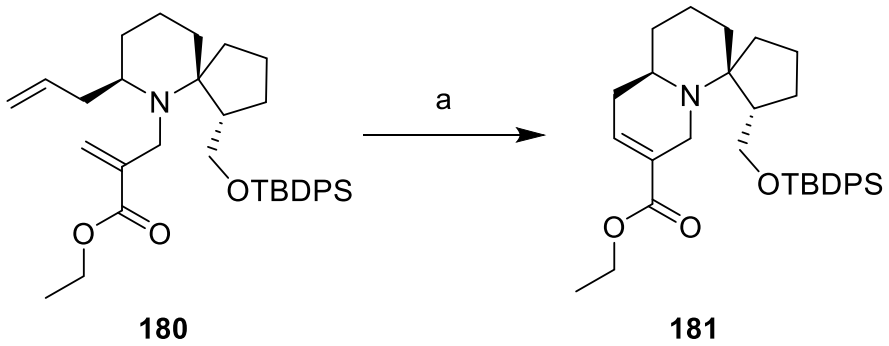
Attempting the *N*-alkylation with ethyl 2-(bromomethyl) propenoate to provide a substrate for planned intramolecular RCM under reflux conditions in acetonitrile produced only limited results. Attempts were, however, far more successful after an extended irradiation in a microwave reactor to give tertiary amine **180** in good yield. (Scheme 45)



Scheme 45:- Synthetic scheme directed towards tertirary amine **180.** *Reagents and conditions*: a) Ethyl 2-(bromomethyl) propenoate, K_2CO_3, acetonitrile, MW, 110 ^oC, 67 %

Ring closing metathesis in the presence of Grubbs catalyst was next attempted on tertiary amine **180**. Previous studies directed towards halichlorine core structure targets have utilised Grubbs (II) catalysts to effect RCM reaction with unsaturated esters to great effect.[80, 94]

Grubbs II generation catalyst was added, in catalytic amounts, to the diene **180** and the mixture heated under reflux conditions in dichloromethane. (Scheme 46)



Scheme 46:- *Reagents and conditions*: a) Grubbs 2nd Generation catalyst, CH_2Cl_2, reflux, 52 %

With synthesis of initial target **181** in hundreds of milligram quantities, sights were set towards accessing even more topologically advanced analogues of halichlorine which were

now within reach. ^{1}H NMR spectrum easily identified halichlorine target **181** due to the disappearance of multiple olefin peaks seen in substrate amine **180** leaving a single peak at 6.84 ppm indicating the C13 olefin proton had been generated during the ring closure. (Figure 14)

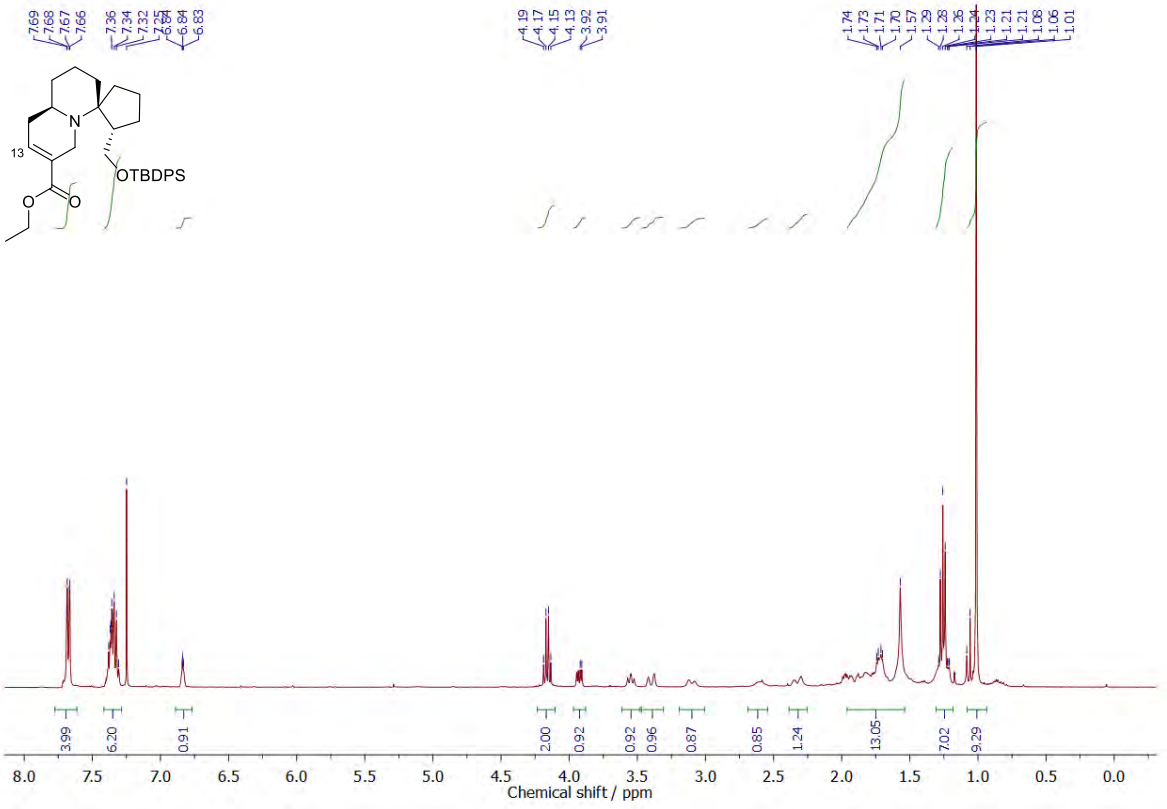
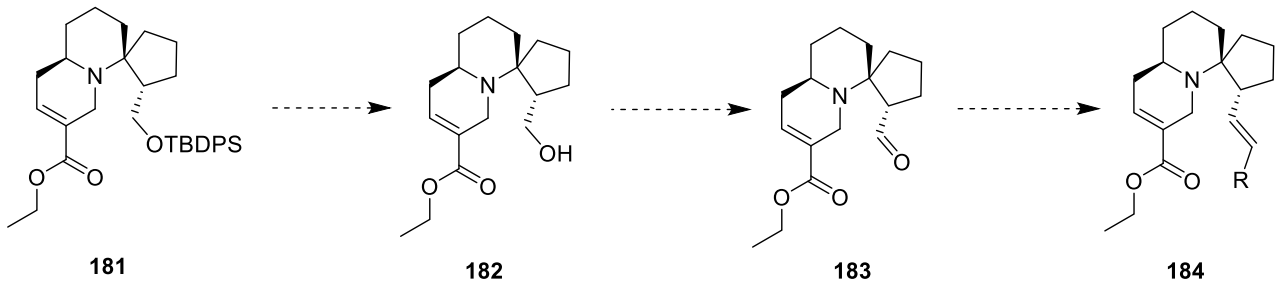


Figure 14:- ^{1}H-NMR spectrum of halichlorine core structure **181** in $CDCl_3$

A new synthetic route was envisioned to access more topologically advance motifs, potentially even directed towards total synthesis of halichlorine. Following successful deprotection of the TBDPS group, an oxidation from the subsequent hydroxy to the aldehyde would provide a handle which could be further functionalised.

Initially it was envisioned that the TBDPS group would be readily removed from the formal synthesis target **181** before an oxidation to an aldehyde motif. The aldehyde would unlock a series of homologation reactions towards more complex analogues and allow introduction of the C11 side chain present in halichlorine. (Scheme 47)



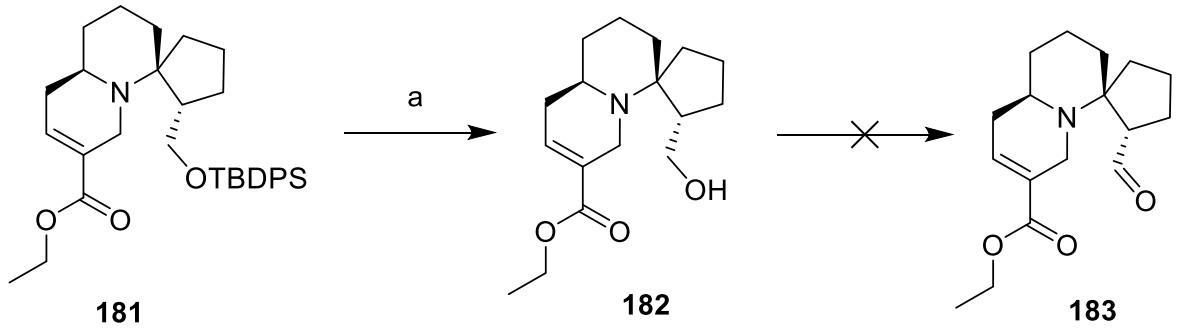
Scheme 47:- Proposed route to halichlorine compound library via Wittig homologation of aldehyde **183**

The first obstacle to progress was the removal of the protecting group on the C11 position. Cleaving the silyl ether and restoring the hydroxy group means losing significant molecular mass and therefore actual mass, leaving less material to use in further synthesis even if these proceed in excellent yield. Nevertheless, the deprotection of silyl ether to secondary alcohol **182** went smoothly in the presence of trimethylamine trihydroflouride and triethylamine.

Difficulty was envisioned in the next step which involved oxidation of the unmasked primary alcohol group to the corresponding aldehyde **183**. The lone pair present on the tertiary amine has the potential to interfere in the oxidation process negating the use of common oxidising reagents such as Dess-Martin periodinane.

Nonetheless, the oxidation was attempted and as expected the reaction did not proceed to completion. Danishefsky reported a similar oxidation (Scheme *28*) in the presence of a lone pair-containing N-atom during a Ley oxidation using *N*-methylmorpholine *N*-oxide (NMO) in the presence of catalytic amounts of tetrapropylammonium perruthenate (TPAP).[89]

Despite the success achieved by the group of Danishefsky *et al.* reaction again did not proceed to completion, unfortunately affording a complex mixture instead (Scheme 48).



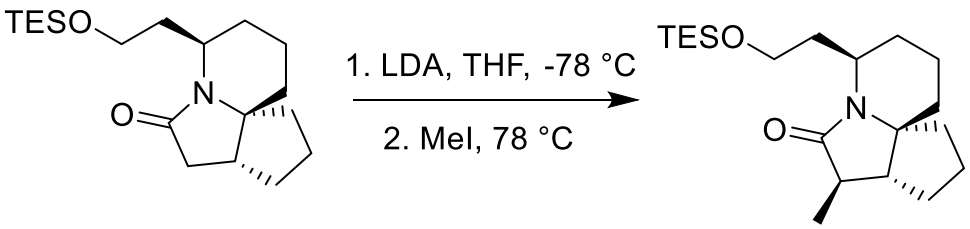
Scheme 48:- Attempted synthesis directed towards aldehyde **183**. *Reagents and conditions:* a) TREAT.HF, Et_3N, acetonitrile, 70 °C, 44 % b) Dess-Martin periodinane, Pyr, 0 °C

4.2. Formal synthesis of pinnaic acid and halichlorine core structure target

As key aldehyde intermediate **119** (See chapter 2.3.1) was prepared by the group of Danishefsky during the total synthesis of pinnaic acid **2** and halichlorine **3**,[84] a number of formal syntheses centre on accessing this target.

During the synthetic routes developed by Danishefsky *et al.* the C11 methyl group is introduced stereoselectively early on. The main problem that complicates the synthetic route discussed here is stereoselective installation of the C11 methyl group at a later stage, subsequent to creation of the spirocyclic scaffold onto a linear, conformationally flexible side chain where it would be difficult to introduce an alkyl group in a predictably stereoselective fashion. Most research in this area has solved this problem by locking this linear chain into a tricyclic, rigid manifold which then displays excellent diastereofacial bias.[30, 76, 93, 95, 96]

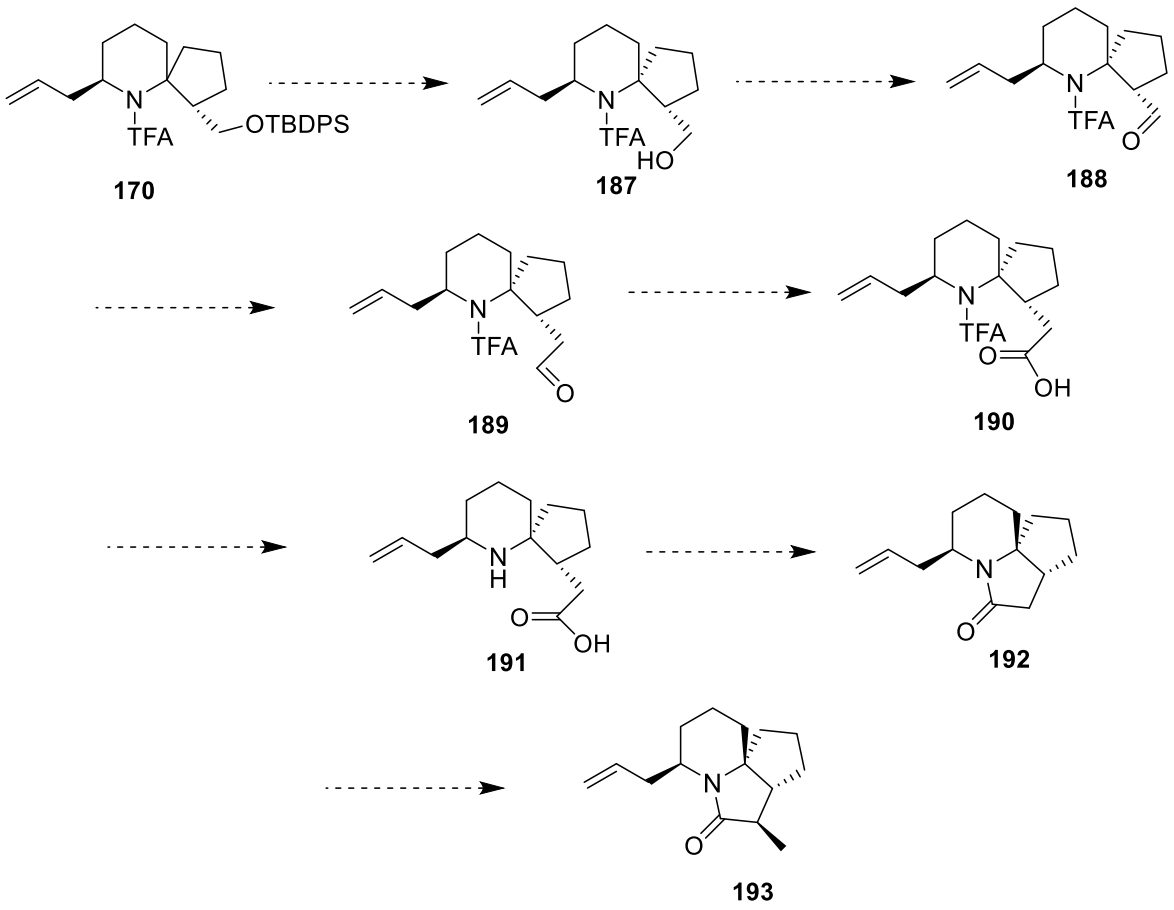
As a representative example, by Ihara *et al.* ,in a formal synthesis of pinnaic acid **2**, focussed on preparation of lactam **185**, which, on conversion to the enolate, displayed excellent stereoselectivity towards methylation, which occurred selectively from the convex β-face and was isolated as a sole diastereoisomer in 85 % yield.[95] (Scheme 49)



185 **186**

Scheme 49:- Ihara *Et al.* late-stage methylation of tricyclic lactam in their formal synthesis of pinnaic acid **2**

Thus, it was decided to investigate the feasibility of converting key aldehydes **188** and **189** to lactams of this type. The tricyclic structure of halichlorine manifold **181** precludes this strategy but it was considered theoretically feasible to convert pinnaic acid core structure **170** to the lactam **193**. (Scheme 50) If this approach can be adapted towards lactam **193**, this paves the way towards a total synthesis if the lactam ring can be cleaved post-methylation

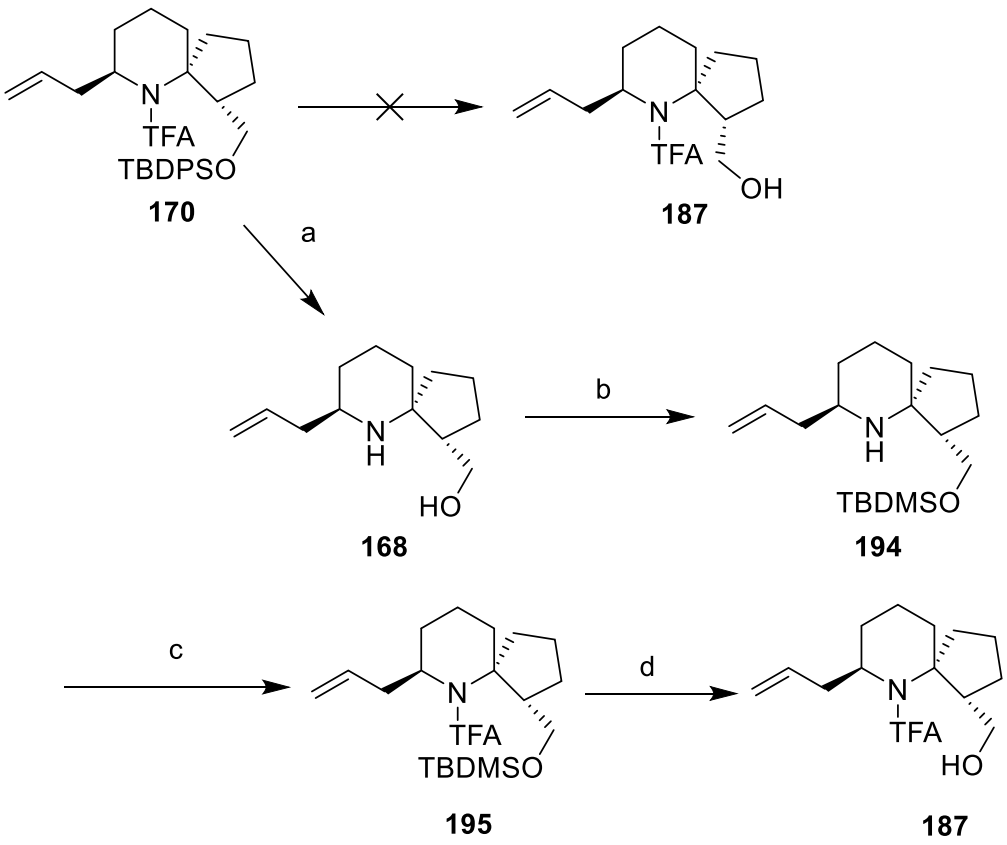


Scheme 50:- Proposed synthetic scheme directed towards formal synthesis target.

4.2.1. Investigation of alternative protecting groups.

Initial attempts to pursue this synthetic route met with difficulty almost immediately. Following the successful synthesis of TFA-protected amine **170** in multi-gram quantities, cleaving of the silyl ether to give access to the alcohol using TREAT.HF was attempted. However, TLC analysis revealed a complex mixture of compounds had formed, with far higher polarity than anticipated. ^{1}H-NMR spectral analysis was also inconclusive. Column chromatography led to isolation of spiroamine **168**. In actuality, the conditions used in the attempt to deprotect the C11 alcohol group had in fact led to cleavage of both *N,O*-protecting groups.

The immediate goal was to devise a strategy to enable selective cleavage of the *O*-protecting group whilst leaving the *N*-protecting group intact. Thus, the alcohol was protected as the more labile TBDMS-ether to afford silyl ether **194** and a trifluouroactyl group was again added to protect the free amine affording acetylated amine **195**. The deprotection was now attempted using mild pyridium *para*-toluenesulfonic acid in CH_2Cl_2. PPTS has been reported as a milder alternative deprotection method utilised in selective cleavage of smaller silyl ethers in the presence of other acid-sensitive protecting groups.[97] It was hypothesised that the use of relatively mild PPTS, would not affect the trifluouroactyl group as the much harsher fluoride containing method had previously. Fortunately, these conditions successfully led to selective cleavage of the silyl ether affording the desired alcohol **187** in good yield. (Scheme 51)



Scheme 51:- Alternative synthetic route to *N*-protected spiroalcohols. *Reagents and conditions:* a) TREAT.HF, TEA, acetonitrile, reflux, 100 % b) TBDMSCl, DMAP, TEA, CH_2Cl_2, 0 ^oC, 47 % c) TFAA, Hunig's base, CH_2Cl_2, 0 ^oC, 65 % d) PPTS, Acetone, r.t. 66 %

The synthesis continued with Dess-Martin periodinane (DMP)-mediated oxidation of the alcohol group to aldehyde **188**. A homologation procedure was then applied using a Horner-Wadsworth-Emmons reaction with (methoxymethyl)triphenylphosphonium chloride to introduce the C12 carbon atom. The methyl ether of the initially formed enol ether **196** spontaneously hydrolysed during aqueous work up affording homologated aldehyde **189** in good yield.

Synthesis of homologated aldehyde was an important milestone in this particular synthesis. Developing a synthetic route towards an aldehyde at the C12 position provides an intermediate that can provide access to a range of analogues via Wittig reactions, aldol reactions etc. This key intermediate was best analysed in CD_3OD due to solubility issues. The peak attributed to the aldehyde peak was found at 9.93 ppm in the proton spectrum (Figure 15). Oxidation under Pinnick conditions converted the aldehyde moiety to acid **190**.

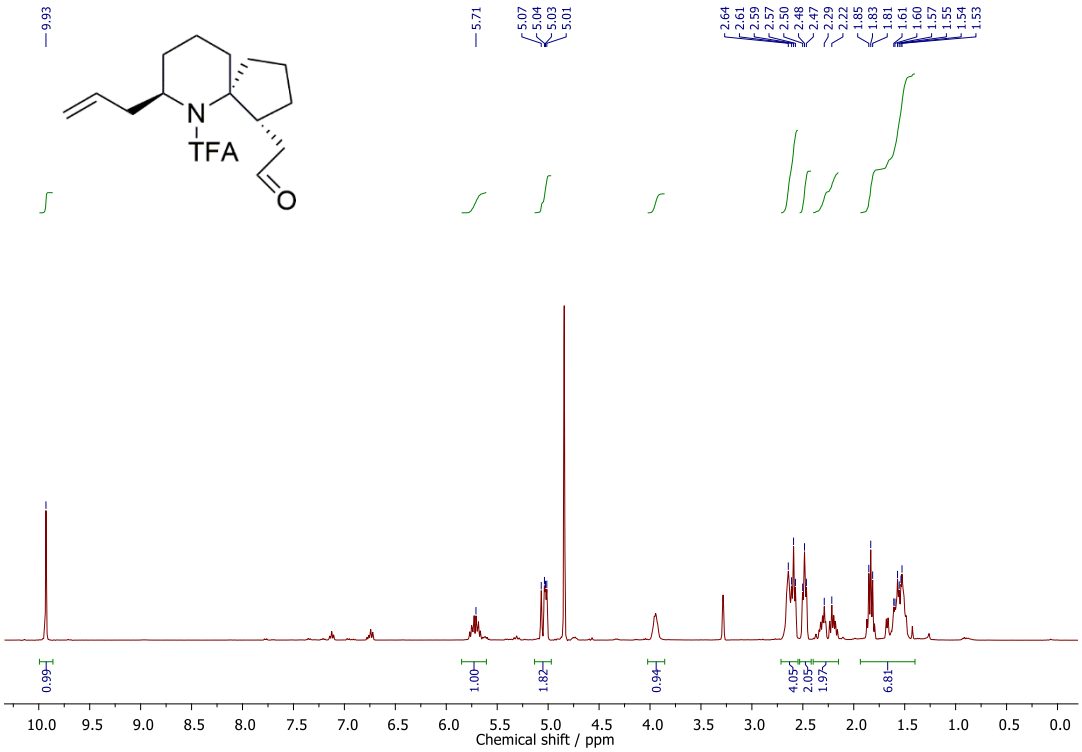
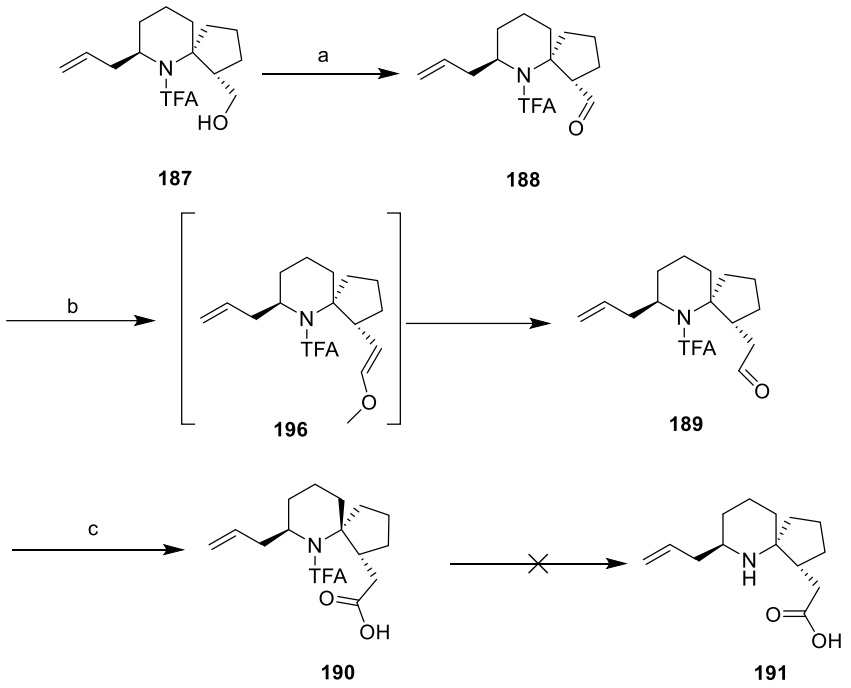


Figure 15:- ^{1}H NMR spectrum of homologated aldehyde intermediate **189** in CD_3OD

The next step in the proposed scheme was deprotection of the tertiary amine followed by lactamisation. It was envisioned that cyclisation could be achieved by conversion of the acid to the mixed anhydride *via* an *N*-acylation reaction with isobutyl chloroformate previously utilised by this research group. [98]

However, this two-step reaction did not proceed to completion due to the low amount of material available after significant steps already being undertaken. The removal of the TFA group was attempted on the small amount of material available. However, there was great difficulty in purification and subsequent analysis of the product of this relatively simple deprotection, due to the significant decrease in molar mass and subsequently actual mass. Also, the incredibly polar product made purification using column chromatography difficult on such a small scale. As a result, even a very high yield would lead to a reduction in already meagre masses of material to hand.

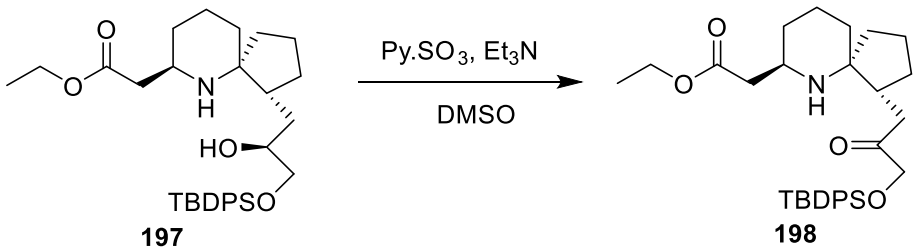


Scheme 52:- Synthetic route towards carboxylic acid 191. *Reagents and conditions*: a) Dess-Martin Periodinane, Pyr, CH_2Cl_2, 0 ^oC, 4 h 70 % b) (methoxymethyl)triphenylphosphonium chloride, NaH, THF, r.t., 12 h, 59 % c) Sodium chlorite, 2-Methyl-2-butene, acetonitrile, r.t, 23 %, d) $NaBH_4$, MeOH, 0 ^oC

4.2.2. Alternative routes to formal synthesis target

Due to the ever-increasing number of steps towards the formal synthesis target, alternative routes were investigated in order to more efficiently use material and time available.

Initially, an attempt was made to circumvent the protection-deprotection steps prior to oxidation of the primary alcohol **168** to the corresponding aldehyde. Complications in oxidising the oxygen atom in the presence of a free N atom could be overcome by using a Parikh-Doering oxidation method as previously reported in a successful approach to the total synthesis of pinnaic acid by Stockman *et al.*[81] (Scheme 53) In this synthesis, alcohol **197** was oxidised to ketone **198** which was subsequently taken forward towards a total synthesis of pinnaic acid **2** and halichlorine **3**.



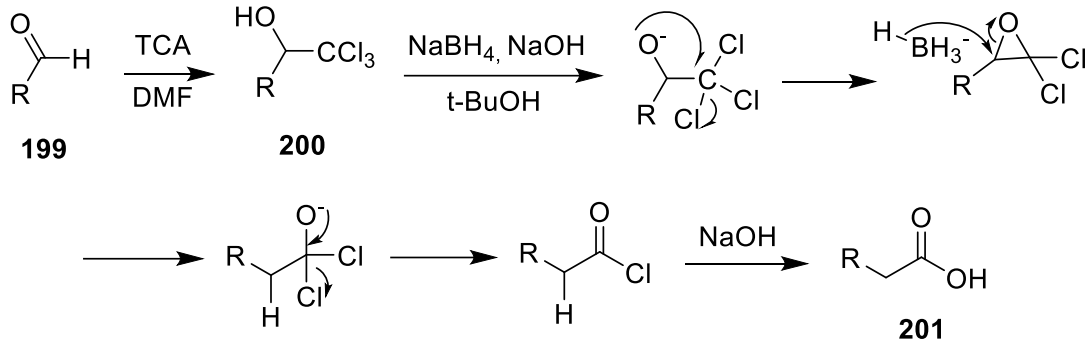
Scheme 53:- Stockman's successful oxidation of an azaspirocyclic ketone **198** by Parikh-Doering oxidation.

Although the reaction proceeded to completion in our hands, to generate aldehyde **202**, difficulties were encountered during the purification of the spiroaldehyde. Attempts to purify *via* column chromatography were complicated due to the polarity of the target compound and the relatively low stability of the aldehyde on silica gel. Due to these problems this strategy was abandoned.

Another alternative route is to attempt was to attempt direct conversion of the aldehyde to the homologated acid. This can be achieved *via* conversion of aldehyde **188** to the corresponding trichlorocarbinol under Corey-Link conditions. Before addition of $NaBH_4$ in the presence of NaOH base should then afford the target homologated carboxylic acid.

Such a synthetic route has been reported by Cafiero and Snowden.[99] They report a successful conversion of a wide range of aldehydes **199** to trichloromethylcarbinols **200,** via a Corey-Link reaction, in the presence of trichloroacetic acid in DMF. This is

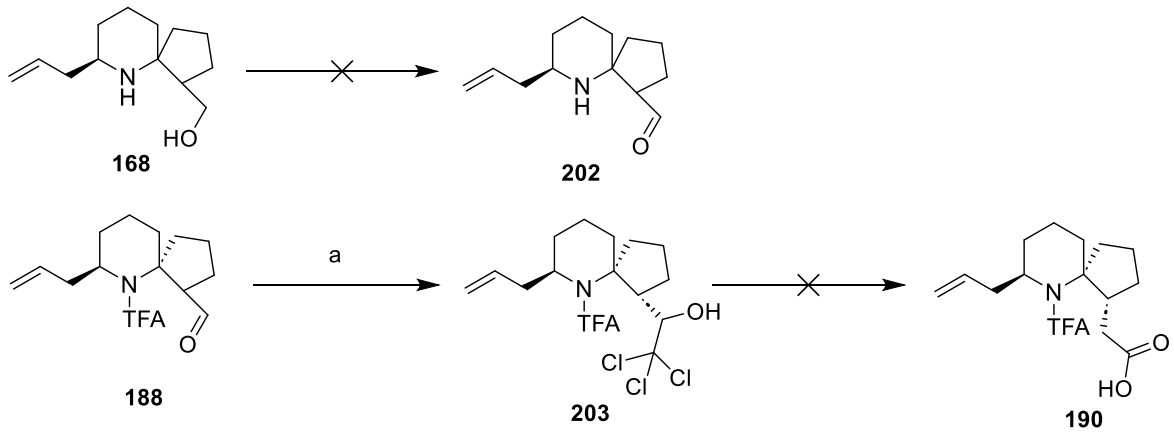
subsequently followed by the addition of $NaBH_4$, in the presence of a strong base, to synthesise the homologated acid in a Jocic-like reaction to access a single-carbon homologated carboxylic acid **201.** (Scheme 54)



Scheme 54:- Approach to homologated one carbon homologated acids **201** from aldehydes

This approach seemed ideal as it would avoid a number of steps in required in the original approach to lactam **192**.

Initial success in synthesising the trichlorocarbinol **203** in good yield was encouraging, however, the second homologation step proved to be a stumbling block. Purification of the crude product of acid **190** seemed to afford a complex mixture which was much more polar than anticipated. It became apparent that this method was not viable as the use of $NaBH_4$ is one of the more common methods of removing TFA groups from nitrogen and it may be that concomitant deprotection of the TFA group occurred during this process. (Scheme 55)



Scheme 55:- Alternative synthetic attempts towards formal synthesis target. *Reagants and conditions:-* a) DMSO, SO_3•pyridine, Et_3N, r.t b)DMF, Cl_3CCO_2H, sodium trichloroacetate, Et_2O, r.t., 31 % c) $NaBH_4$, NaOH, *t*-BuOH, 55 ^oC

Following a successful diastereoselective approach towards hydroxylamine **161** bearing the correct stereochemistry at the C7 position, as seen in spirocyclic natural products **2** and **3**, the stage was set for the attempt at a synthetic approach to halichlorine-based motifs **157** & **158**.

Tricyclic target **181** was synthesised in good yield. The next step to further elaborate this particular analogue will be synthesising the key aldehyde handle at the C11 position, which when achieved will offer a route to a number of more complex functionalised motifs of halichlorine **3**. The diastereoselective approach to **189** is a major hurdle that has been overcome as outlined in this chapter.

Furthermore, investigating the issues with the *O*-protecting groups which was preventing further progress in this and previously attempted synthesis (See chapter 3) not only allowed further investigations towards key homologated analogue **189** but also unlocks future research with the knowledge of which protecting groups to use already obtained. The next goal will be to circumvent the use of these protecting groups entirely if possible.

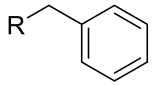
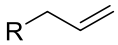
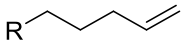
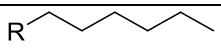
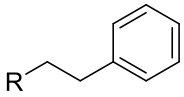
The method testing in the approach to halichlorine has been invaluable in not only leading towards the successfully synthesis of target molecule **181** but also in building on successfully synthesised alcohol **182** and synthesising a more complex library of analogues of halichlorine **3**.

5. Synthesis of library of smaller analogues of spirocyclic alkaloids

It has been shown that relatively simple 6,5-azaspirocycles of similar structure to the pinnaic acid/halichlorine display nicotinic activity (E.g. Compounds **4**-**7** , see chapter 1.3). This does raise the possibility that related structures, easily generated during the developed approaches to pinnaic acid and halichlorine might also display such properties. Reported bioactivities of pinnaic acid/halichlorine have all centred-on investigations of anti-inflammatory effects rather than neuroactive properties. It was envisaged that the strategy developed in this thesis towards the core structures of these natural products and histrionicotoxin **1** could be readily applied to develop a library of 6,5/6,6-adducts to enable future evaluation of bioactivity towards discovering new compounds with nicotinic activity. Blackshaw *et al* performed a series of inflammatory bioassays to determine if a series of smaller halichlorine analogues exhibited bioactivity.[88]

Investigations in this chapter focused on the addition of alkyl groups synonymous with natural products **1**-**3** and their derivatives. However, it is important to consider physicochemical factors and how these parameters might affect pharmacokinetic and pharmacodynamic properties of analogues and their application in bio-evaluative studies. While bio-evaluation was not undertaken in this thesis, computationally-generated predicted values for different physicochemical properties would have to be considered in potential future application as a drug molecule, where molecular weight, lipophilicity and membrane permeability amongst other factors would have to be considered. Log P as an indicator will also have to be considered to ensure any potential drug molecule can be suitably absorbed and distributed in the body and is able to penetrate vital membranes.

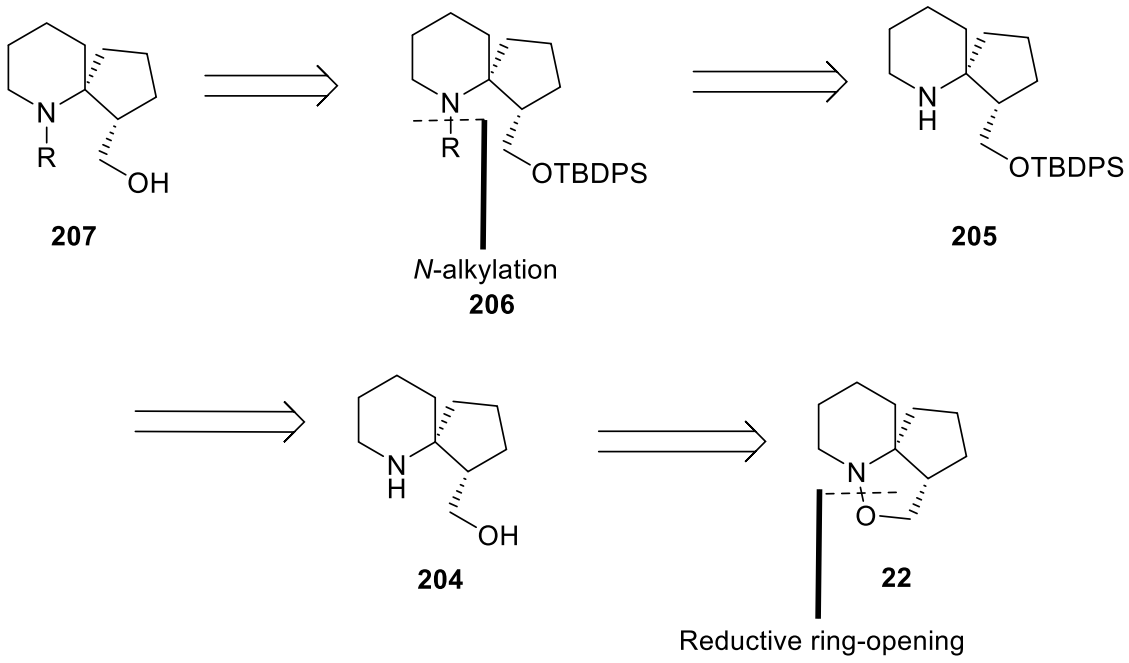
Table 1:-Physicochemical properties of alkyl groups of interest in this chapter calculated using 'chemdraw professional' software

Alkyl group	Chemical Formula	M/W	Boiling Point (°C)	LogP
	C_7H_7	91.13	113	2.52
	C_3H_5	41.07	-10	1.48
	C_5H_9	69.13	38	2.32
	C_6H_{13}	85.17	64	3.00
	C_8H_9	105.16	136	2.94

5.1. Synthesis of *N*-substituted spirocyclic adducts

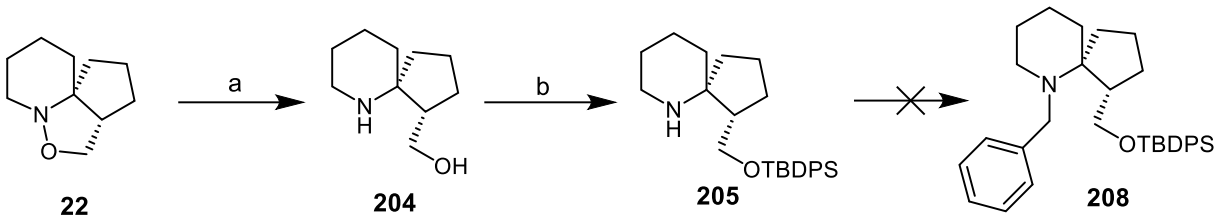
Initially, a reaction sequence was explored involving reductive ring opening of isoxazolidine **22** to give spiroamine **204** using zinc in aqueous acetic acid. No difficulties were foreseen as cleavage of a N-O isoxazolidine bond is well reported both in the literature and by this research group.[88, 100]

Following this, protection of the hydroxy group would provide silyl ether **205**. Alkylation of the amine group with a series of different alkyl bromides would provide silyl ether target **206** followed by deprotection of the TBDPS-ether would deliver a small library of *N*-substituted 6,5-motifs **207**. (Scheme 56) It was later planned to access a range of 6,6-spirocyclic analogues in similar fashion using isoxazolidine **23.**



Scheme 56:- Proposed synthetic route towards library of simple 6,5-spirocycles **207**

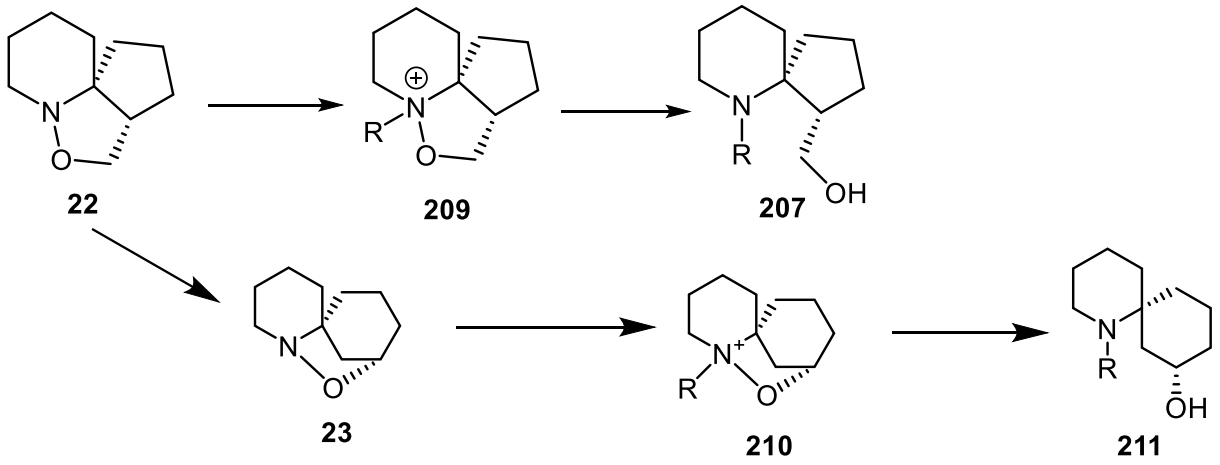
However, it quickly became apparent following synthesis of silyl ether **205** that the above strategy, involving successive protection/deprotection strategies, would not be a sustainable method for a sufficient library of analogues due to low efficiency of this linear approach and insufficient overall yields. Significant loss of yield after the first two steps, the reductive cleavage of the N-O bond and the subsequent protection of the hydroxy group was observed along with difficulty in purification of the amine **204** due to its high polarity. (Scheme 57)



Scheme 57:- Initial attempted synthesis of *N*-substituted benzyl spiroamine **208.** *Reagents and conditions:* a) Zn, Aq. acetic acid 70 °C, 72 % b) TBDPSiCl, DMAP, Et_3N, CH_2Cl_2, r.t, 83 %

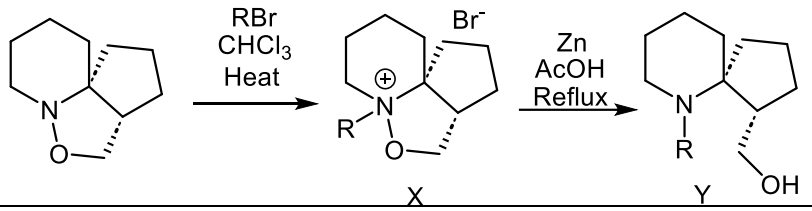
Consequently, a new synthetic pathway towards these *N*-substituted adducts was attempted, with good success, starting from spirocycle **22**. Direct nucleophilic addition of a series of alkyl bromides to this isoxazolidine gave access to *N*-substituted ammonium salts which also function as useful, charged, sp^3-rich scaffolds. (Scheme *58*) Reductive cleavage of the N-O bond then afforded the target *N*-alkylated tertiary amines in good yields (Table 1). The success achieved with this route gave assurance that a similar strategy could be implemented towards the 6,6-spirocyclic targets. Synthesis of 6,6-motifs was also successful using the same technique as outlined above with isoxazolidine **23** as starting material (Table 2).

It is clear that a library of azaspirocyclic analogues is readily available via this route for bio-evaluation studies.



Scheme 58:- Refined synthetic scheme towards library of *N*-substituted analogues. Reagents and conditions shown in table 1 & 2 below.

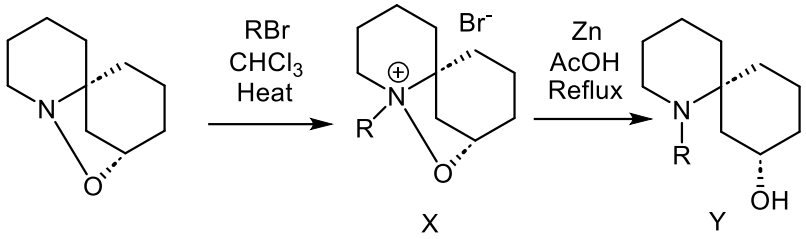
Table 2: Alkylation of 6,5,5-spirocyclic isoxazolidines and reduction of resulting alkyl ammonium salts



X Y

Entry	R	Product	Temp/°C	Yield X/ (%)	Yield Y/ (%)
1	Bn	212/217	r.t.	85	48
2	CH_2CH_2Ph	216	130	52	-
3	Allyl	213/218	r.t.	88	55
4	5-Pentenyl	214/219	100	75	45
5	n-hexyl	215	130	58	-

Table 3 Alkylation of 6,6,5-spiroisoxazolidines and reduction of resulting alkylammonium salts



X Y

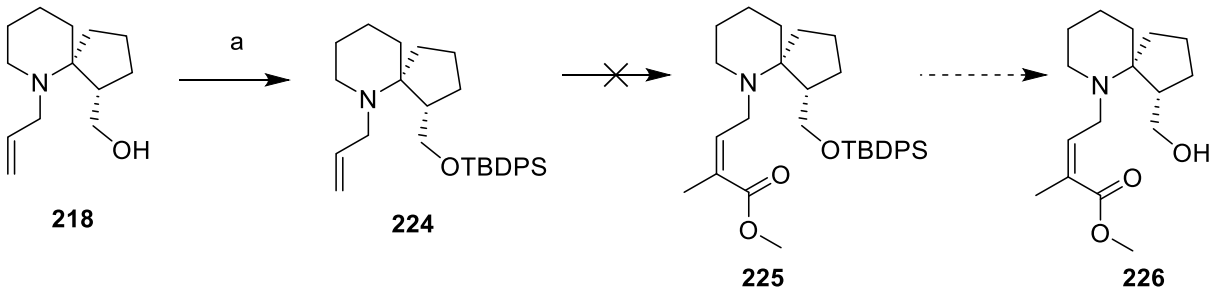
Entry	R	Product	Temp/°C	Yield X/ (%)	Yield Y/ (%)
1	Bn	220/222	r.t.	79	54
2	Allyl	221/223	r.t.	84	55

The reductive ring opening, by cleavage of the N-O bond, proceeded with difficulty in the cases of attempted reductions of ammonium salts **215** and **216**. This appears to be linked to the size of the *N*-alkyl chain with ammonium salts bearing large *N*-substituents being more resistant to the reductive ring opening, while reduction of analogues with smaller *N*-substituents proceeded more readily and with more impressive yields. Additionally, reaction monitoring was difficult. As the starting material was a salt, use of TLC to follow the reaction was not possible.

Use of zinc, in the presence of indium metal, as used by this research group previously to effect cleavage of N-O bonds in azaspirocyclic isoxazolidines[88] was also attempted but did not deliver improved results. Nevertheless, a range of tricyclic ammonium salts and

N-substituted spirocyclic amines were synthesised. It should be noted that HTX (in common with nicotine) binds as the protonated ammonium salt and the series of alkylated ammonium salts prepared also represent novel, multicyclic, sp^3-rich, water-soluble scaffolds for bioevaluation.

To further expand the library of analogues available to us, allyl adduct **218** was utilised in an attempt to synthesise more topologically advanced molecules of potential interest. Initially an *N*-substituted α, β -unsaturated ester motif structurally similar to the side chain of pinnaic acid **2** was targeted. The route involved protection of the free alcohol group with a TBDPS group, before attempting a cross-metathesis with methyl acrylate, in the presence of Grubbs' (II) catalyst. (Scheme *59*) Previous success with both these techniques (See sections 3.2 & 3.4.1) indicated that this would be a relatively simple route to a more complex target.



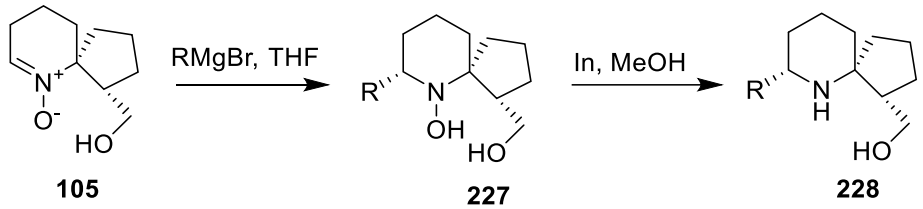
Scheme 59:- Attempted synthesis of more complex target structure. *Reagents and conditions*: a) TBDPSiCl, DMAP, TEA, CH_2Cl_2, 0 ^oC, 58 % b) Ethyl 2-(bromomethyl) propenoate, K_2CO_3, acetonitrile, MW 110 ^oC

While initial protection afforded silyl ether **224** in good yield, the attempted cross-metathesis did not proceed to completion.

5.2. Grignard additions to 6,5 spironitrone

It should be possible to access a range of azaspirocyclic analogues of pinnaic acid by Grignard addition to key spirocyclic nitrone **105**.

Previously, this research group had investigated the addition of a series of Grignard reagents to the 6,5-spironitrone, synthesising a small library of analogues.[88] The subsequent reduction of the hydroxylamine adducts, using the standard procedure of zinc in acetic acid, was problematic however - affording acetylated adducts in some cases. Limited success was achieved using catalytic amounts of indium metal in MeOH to give spiroamine targets. (Scheme *60*)

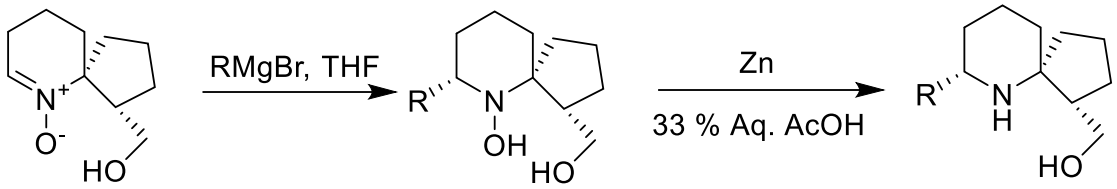


Scheme 60:- General scheme towards spiroamine initial utilised previously by the Caprio research group.

However, alternative methods have been investigated as part of this project to generate a synthetic procedure with increased yields and potentially negating the use of expensive metal catalysts.

Grignard additions proceeded according to those previously reported by this research group. Reduction was attempted using new optimised methods, discussed in previous chapters (Chapter 3.2) with good results achieved by the use of zinc metal in the presence of a weaker acid. Hydroxylamines proved difficult to purify so crude mixtures were instead reduced immediately following work-up. Spiroamine targets were successfully synthesised in good yield with no formation of acetylated products as previously observed.[88]

Table 4 Grignard addition of alkyl magnesium bromide reagents to spironitrone



Entry	R	Product	Yield (Over 2 steps) (%)
1	Phenyl	229	60
2	Benzyl	230	35
3	Pentyl	231	46

The relative stereochemistry of addition was assigned using 2-D NOESY NMR spectroscopy as applied to the allyl adducts discussed previously in this thesis (Chapter 3.2). The peak at 4.6 ppm (Figure 16) was assigned to 7-H, with cross peaks observed between this peak and with the peaks assigned to the 5-H peak at 2.8ppm (Figure 17).

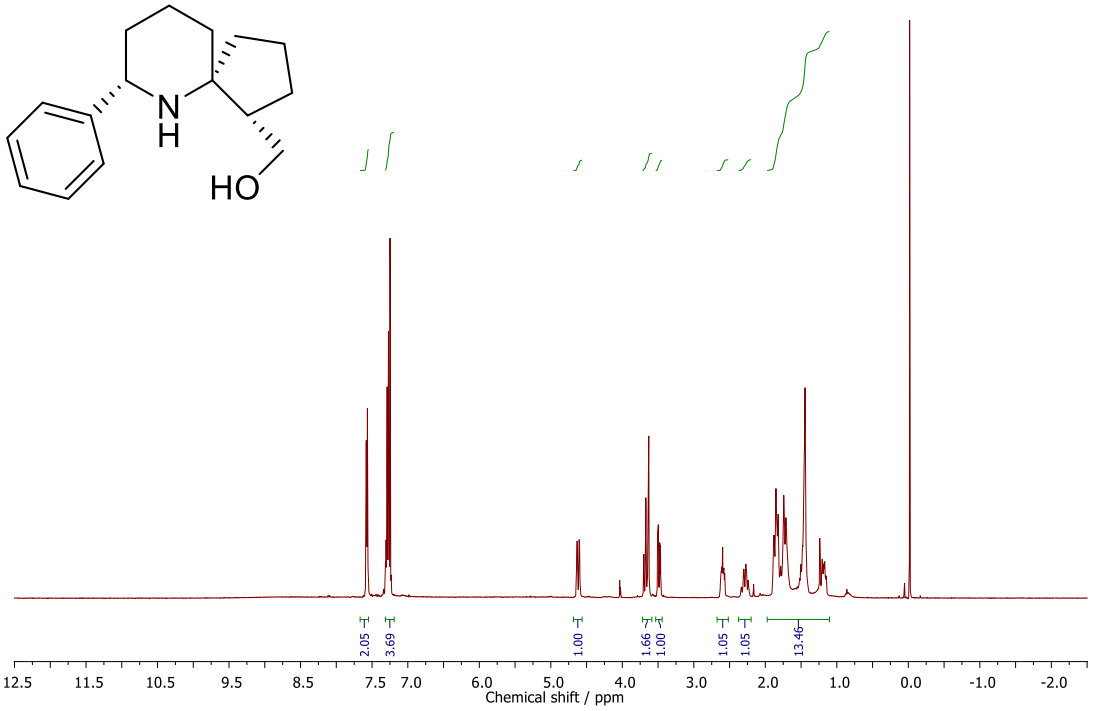


Figure 16:- ^{1}H NMR of spiroamine **229** in $CDCl_3$.

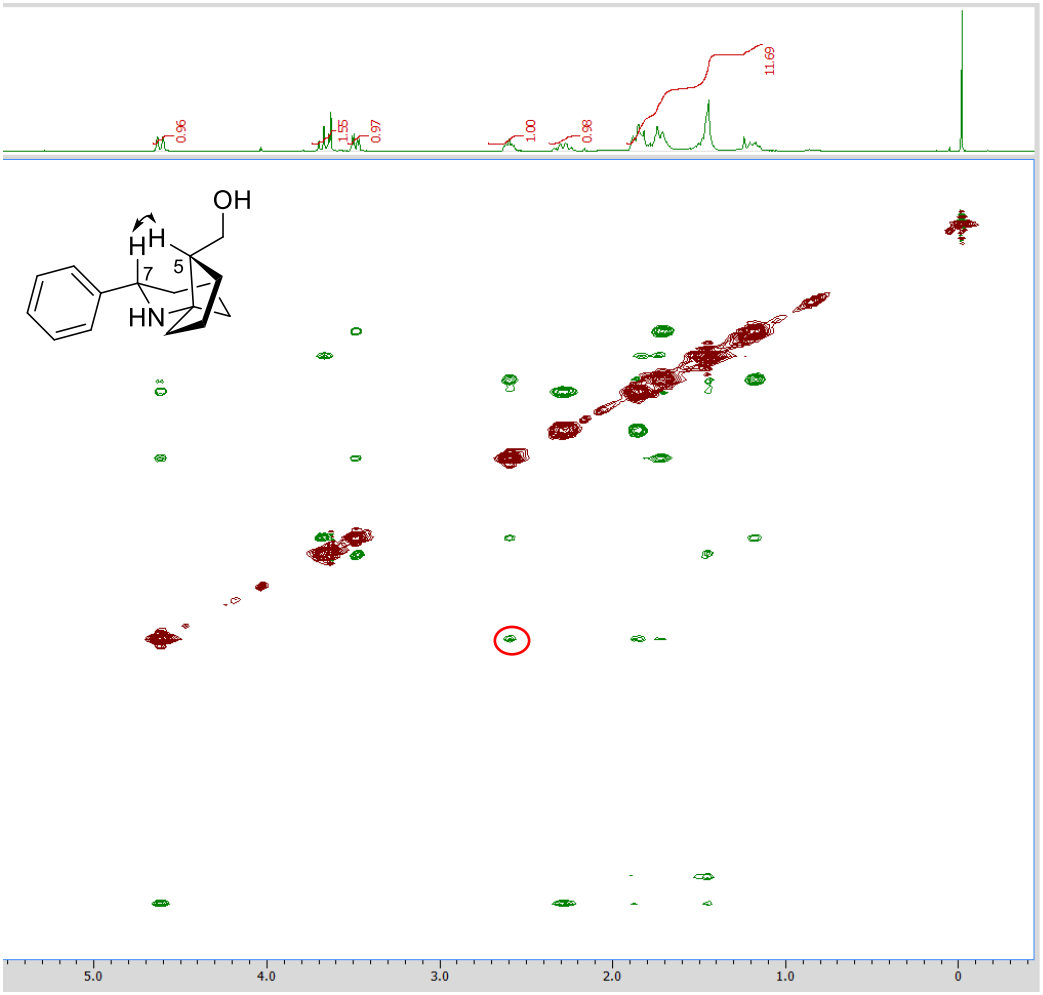


Figure 17:- 2-D NOESY of spiroamine **229** in $CDCl_3$

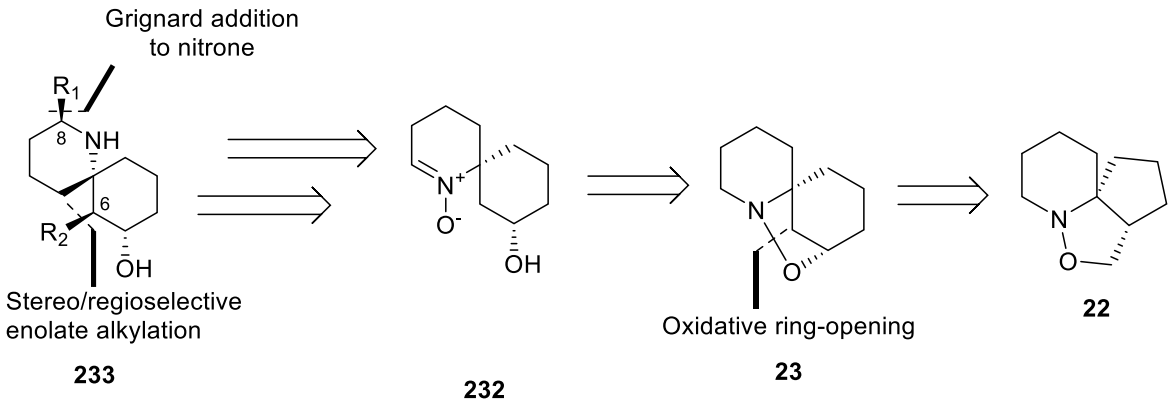
Outlined in this chapter is an approach towards a small library of novel spirocyclic analogues of the natural product targets. Development of these routes enabling alkylation of the N atom and the C7 position allows entry into a large library of analogues through varying the alkyl halides and Grignard reagents/organometallic nucleophiles used. Further bio-evaluative studies of these compounds will yield invaluable data into the neurological, inflammatory and nicotinic activity of these compounds.

A further benefit to this synthetic route is the ease of application to the 6,6,5 isoxazolidine allowing entry into a library of simple 6,6-spirocyclic motifs of HTX 1, which have proved challenging for this research group to access and there are very few reports of HTX analogues in the literature.

6. Attempted synthesis of 6,6 analogues of histrionicotoxin

Attention then turned to focus on adapting the methodology developed towards 6,5-azaspirocycles to the 6,6-core structures of histrionicotoxin. While simple analogues were synthesised previously (See chapter 5.1), sights were now set on more topologically advanced structures. Due to the structural similarities between the [6,5,5] **22** and [6,6,5] **23** core structures of natural products **1** and **2**/**3**, it was envisioned that similar synthetic routes could be investigated adapting previously successful routes towards 6,5-targets, in particular by exploiting nitrone chemistry by synthesising a 6,6 spironitrone **232** and using to great effect as with 6,5-spironitrone **105**.

Target HTX motifs that were to be pursued utilising nitrone chemistry to access both butyl and pentyl-containing 6,6,5 system **233**. In a similar fashion to the pursuit of pinnaic acid and halichlorine core structures, it was hypothesised that an alkylation of spironitrone **232** would introduce the side-chain at the C8 position. In addition, it was planned to explore a regioselective enolate alkylation to introduce the C6 position. (Scheme 61)



Scheme 61:- Retrosynthesis directed towards potential 6,6,5 target motifs

6.1. Synthesis of core structure

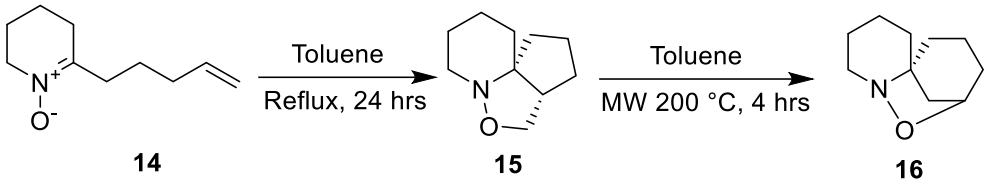
To pursue the 6,6-analogues of HTX, a key azaspirocyclic isoxazolidine intermediate needed to be synthesised in multi-gram quantities. This intermediate had previously been synthesised by converting the kinetic, 6,5,5-cycloadduct **22** to the more favourable 6,6,5-isoxazolidine **23** under thermodynamic conditions by heating under high temp and pressure under microwave conditions.

Synthesising isoxazolidine was relatively straight forward, however significant difficulty was encountered in scaling up operations to achieve multi-gram quantities needed for further synthesis. Reaction conditions needed to be harsh enough to force the product through to the thermodynamic favoured compound, but not so extreme that starting material would degrade beyond any usefulness.

Alternative methods were attempted including pressurised ACE reaction vessels that could be attempted using a stirred hotplate and a sand bath, however this approach only afforded miniscule quantities of 6,6-products. Additionally, 6,5-pentenyl nitrone **21** was heated under extreme conditions in an attempt to form the 6,6,5 adduct directly, however this only resulted in synthesis of the 6,5,5 system.

Fortunately, the 6,5,5 adduct could be recycled during the vast majority of attempts as unreacted starting material.

Table 5:- Reaction conditions investigated in synthesis of isoxazolidine.



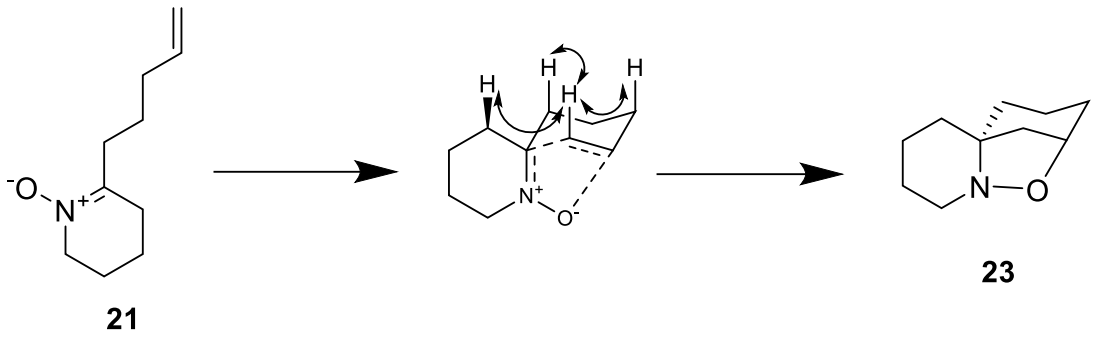
14 **15** **16**

Entry	Starting Material	Concentration (g/mL)	Reaction Vessel	Temperature (^{o}C)	Reaction time (h)	Yield (%)
1	21	0.25	Pressure tube	Reflux	24	0*
2	21	0.25	MW vessel	150	4	0*
3	22	0.0625	MW vessel	150	4	6
4	22	0.0625	MW vessel	220	4	0
5	22	0.25	Pressure Tube	Reflux	24	0*
6	22	0.25	MW vessel	220	4	41
7	22	0.25	MW vessel	250	2	0

*6,5,5 adduct **22** was instead collected

Following optimisations of concentration of starting material, temperature, and reaction duration, 6,6-isoxazolidine **23** was successfully synthesised in multi-gram quantities. Logistical problems also arose with equipment available in the lab, only 2 g scale reactions could be placed on at a time meaning large scale synthesis would be extremely slow. While a steady supply of 6,6 isoxazolidine could be obtained, the maximum scale that could be put on at once was 2 g at 200 ^{o}C, significantly impeding efficiency of this process

Gossinger *et al.* reported a relatively high activation energy is required to force the formation of the [6,6,5] adduct - caused by the 1,3-diaxial interactions found in the transition state (Scheme 62).



Scheme 62:- Cycloaddition and transition state en route to isooxazolidine **23**.

The conversion of the 6,5,5-adduct to the 6,6,5-isomer can be readily determined by analysis of the proton spectrum. The C5 carbon with a lone proton neighbours the O atom in the [6,6,5] adduct while there is a methylene at this position in the 6,5,5-adduct. (Figure 8) Indeed a peak at 4.5ppm does integrate as one proton rather than 2 (Figure 18), and HMQC analysis confirms this is a methine rather than a methylene.

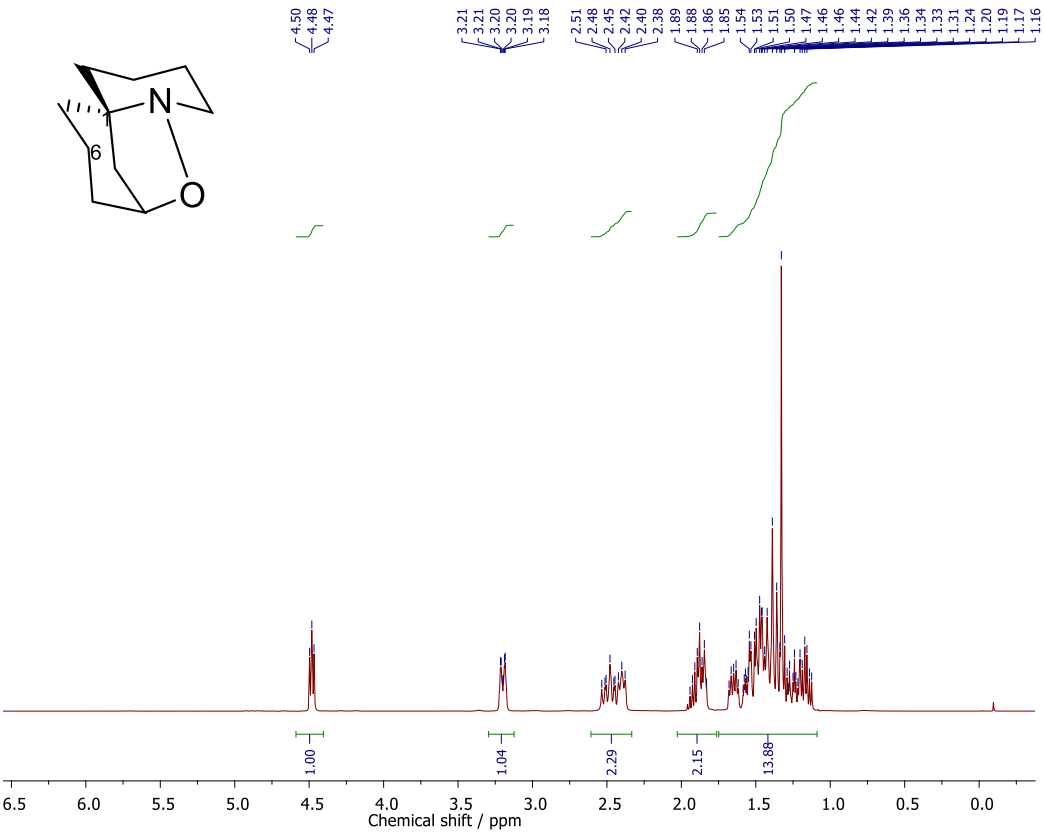
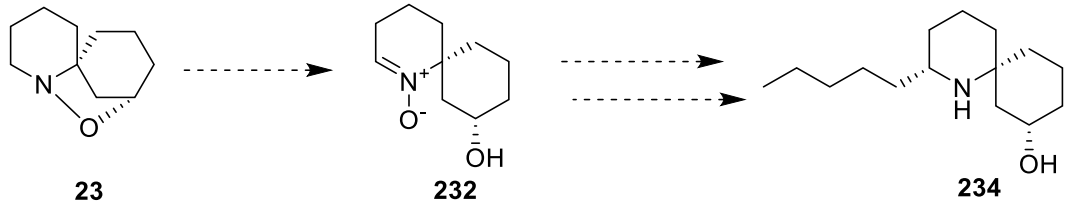


Figure 18:- ^{1}H NMR of 6,6 isoxazolidine **23** in $CDCl_3$.

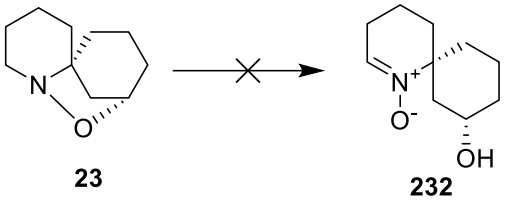
6.2.Attempted synthesis of alkylated HTX analogues

With multigram quantities of 6,6-isoxazolidine in hand, the next step was to attempt synthesis of key spironitrone **232**. It was then envisaged to introduce the pentenyl side chain of perhydrohistrionicotoxin via Grignard addition. (Scheme *63*)



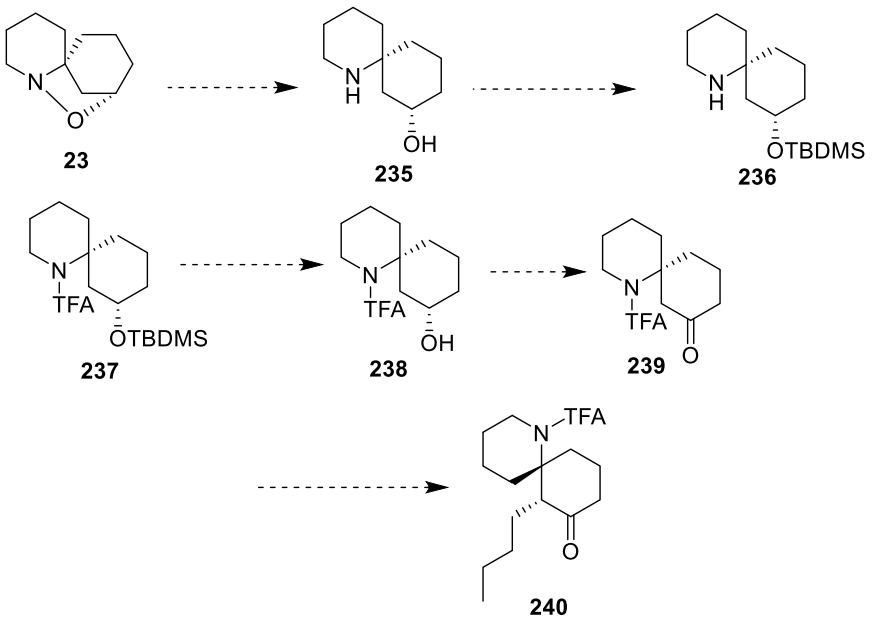
Scheme 63:- Proposed synthetic route towards pentyl spiroamine target.

As with the 6,5 adduct, a small-scale attempt at oxidative cleavage was unsuccessful. Therefore, 3g of isooxalidine **23** was targeted in the belief that a larger scale reaction would be more successful as observed during work towards the 6,5-spironitrone **105**. However, despite repeated attempts at synthesising the 6,6 spironitrone on a large scale using a slow rate of addition of mCPBA, reaction did not proceed to completion, instead affording a complex mixture of adducts. (Scheme *64*)



Scheme 64:- Failed synthesis of 6,6-spironitrone **232** Reagents and conditions:- mCPBA, CH_2Cl_2, 0 °C, 7 h, 85 % b) mCPBA, CH_2Cl_2, r.t, 7 h

An alternative synthetic target **240** was investigated which would provide analogues of potential interest for bio-evaluative studies and would allow exploration of the potential for a regioselective enolate alkylation to deliver the C6 side chain of HTX. (Scheme *65*)

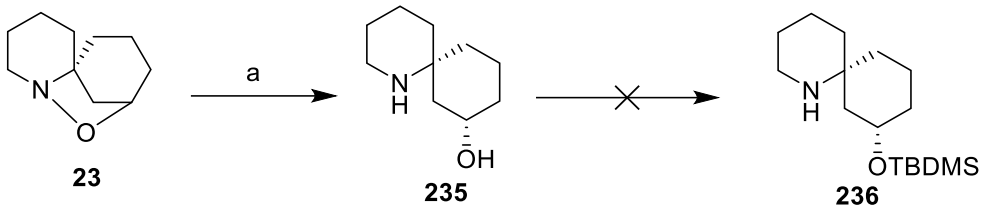


Scheme 65:- Proposed synthetic route towards butyl spiroamine target.

Lessons had been learned from previous similar synthesis from this project. As this route had protection/deprotection steps as seen previously in this thesis (See chapter 4.2.2.) it was known from the beginning that TBDPSCl could not be used as the protection group and that the use of TBDMSCl was required in the synthesis of silyl ether **236**.

A key difference in this approach than seen previously is the presence of a secondary alcohol on 6,6 spiroamine **235, i**nstead of the primary alcohol group present with spiroamine **168**. A reduction of the ketone group on **239** would preclude the C-C bond formation α to the ketone to afford the target **240** with the desired butyl side-chain at the C6 position.

Initial attempts towards this target began smoothly, with synthesis of the 6,6-spiroamine in good yield without the need for purification. Initial attempts at protection of the free alcohol with a TBDMS group failed, potentially due to the alcohol being in a secondary rather than primary position as in compound **194**. (Scheme *66*). Notably, there is no previous example of this protecting group being used on a similar secondary alcohol during previous work towards the HTX family of alkaloids.



Scheme 66:- Failed silylation of secondary alcohol. *Reagents and conditions:* a) Zn, 33 % aq. AcOH, 70 °C b)) TBDMSCl, DMAP, TEA, CH_2Cl_2, 0 ^oC

While the synthesis of target molecules **234** and **240** was unsucessful, invaluable data has still been acquired during the synthesis of isoxazolidine **23**, which has proven difficult to isolate in sufficient yield to enable long-term, complex multi-step synthetic routes.

Isoxazolidine **23** can still be isolated in sufficient yield for smaller scale investigations in the future (see chapter 5) but as of yet not in sufficient quantities for investigation into the key oxidative ring opening step (which must be performed at scale for success) that would afford key spironitrone intermediate **232**. If this step were to be mastered then several different routes could be explored, as has been demonstrated by the versatility of 6,5-spironitrone **105**. With sufficient time this particular avenue can no doubt be explored even further.

7. Summary & Future work

7.1 Summary

The overall aims of this project were partially achieved with successful synthesis of several scaffolds initially targeted. Not only was a new, stereoselective strategy towards the core structures of pinnaic acid and halichlorine designed and successfully implemented but this approach can be applied to access the azaspirodecane core of both molecules in multi-gram quantities. Moreover, this strategy enables efficient access to target scaffolds with relative stereochemistry matching that of the natural products and is sufficiently flexible to enable facile synthesis of a range of core structure analogues of both targets for future bioevaluation.

The synthesis was successful in accessing both targets with the desired side chains/polycyclic systems including methyl ester derivatives **175**-**177** of the pinnaic acid core (Chapter 3.41 & 3.4.2) and the spiroquinolizidine core **181** of halichlorine (Chapter 4.1).

As such,sights were set even higher towards more topologically advanced motifs - even a potential total/formal synthesis. While a formal synthesis of pinnaic acid was not completely achieved good progress was made towards introducing the final step of introducing stereospecific methyl group at the C12 position. Developments were also made including the synthesis of a key aldehyde intermediate **189** that could be potentially used to introduce this final side chain. Efforts in this direction were complicated by the necessity of introducing the C12 methyl group in a stereoselective sense – an obstacle which has lengthened previous synthetic approaches in this area.

A small library of *N*-alkylated azaspiro/isoxazolidine derivatives were accessed, with facility, from key isoxazolidine **22** and **23**. The biological activity of these sp^3-rich compounds remains to be determined, although, based on previous reports, it is not unlikely that these compounds may exhibit nicotinic activity. Moreover, the strategy

developed in this thesis will be applicable to the synthesis of a wide range of such compounds as outlined in this thesis (Chapter 5.1).

While a 6,6,5-azaspirocycle **23**, of much potential use towards histrionicotoxin was prepared – unfortunately, this could not be converted to the target natural product itself owing to problems during oxidative ring opening of this cycloadduct to give the key 6,6,5-spironitrone **232**, exacerbated by lack of resources available to bring through large quantities of isoxazolidine **23**. However the successful application of spironitrone **105** in the synthesis of larger more complex natural product targets to great effect in this thesis (Figure 19), demonstrates that if these issues can be overcome, as in the initial attempts in the synthesis of spironitrone **105**, it is likely that the great degree of success in utilising spironitrone **105** can also be applied towards synthetic analogues of HTX **1**.

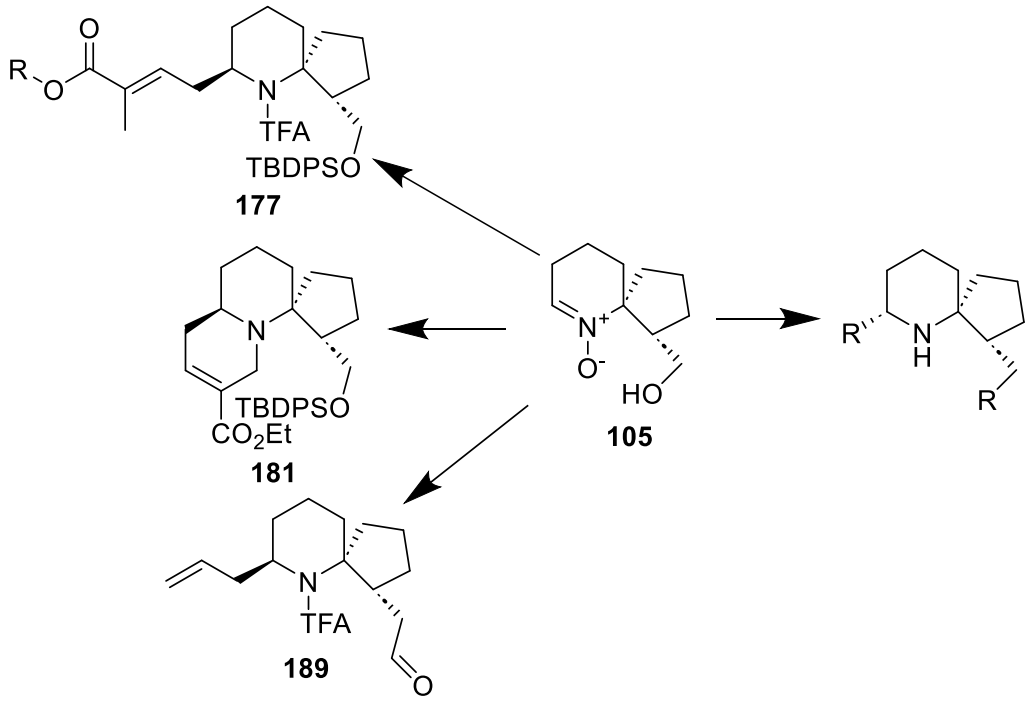


Figure 19:- Overview of key synthetic products synthesised in this project

7.2 Future Work

As previously discussed, there is potential for further investigations in previous chapters discussed in this thesis. Methyl ester **177** and tertiary amine **181** can be considered the core structures of both pinnaic acid and halichlorine and once they can be reliable and economically synthesised in good amounts in future, the focus on utilising these molecules toward more advanced formal syntheses of both targets can begin. A sound synthetic strategy is in place towards a formal synthesis (Chapter 4.2) of pinnaic acid, and with more time invested, there is no reason to doubt that a formal synthesis cannot be established. Furthermore, a key aldehyde **189** successfully synthesised in the attempt of this strategy has been prepared in quantities that enables the preparation of a wide range of C12-based analogues of pinnaic acid, for instance via a series of Wittig reactions.

The similarity in nicotinic activity shared by both 6,5- and 6,6-azaspiro scaffolds discussed previously (Chapter 1.2) indicates that there is strong rationale for investigating the synthesis and bioevaluation of hybrid pinnaic acid/histrionicotoxin structures.

As this project was successful in accessing 6,5-scaffolds but rather less so in approaching the 6,6-core of HTX these molecules should be based on the 6,5-azaspirodecane core of pinnaic acid but bearing side chains derived from HTX such as **241** & **242**. (Figure 20) It will be important to try and ensure that these side chains are orientated in the correct space that maps onto the 3D structure of HTX so that the same pharmacophore is reproduced. If a more reliable and sturdy route to 6,6-scaffolds avails itself, there is also potential for hybrid compounds incorporating the azaspirodecane core of HTX bearing the side chains found in pinnaic acid analogues such as **243** & **244**. (Figure 19)

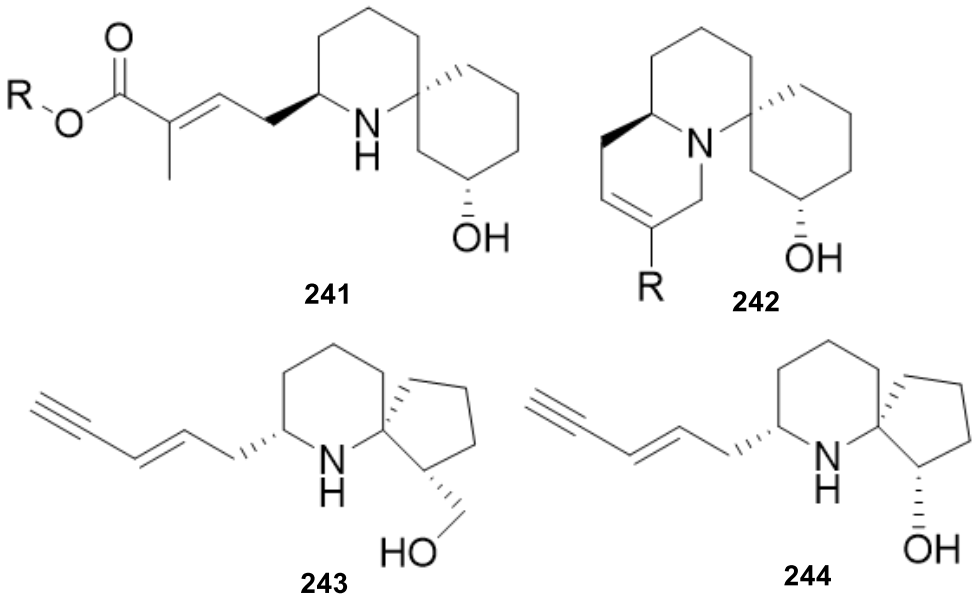
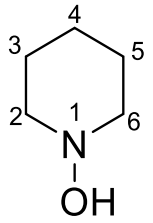


Figure 20:- Potential "hybrid" HTX/pinnaic acid/halichlorine-based compound as targets for further study.

8. Experimental

All reactions were performed using oven dried glassware. Reactions that were air-sensitive were performed using a Schlenk line under Ar. Dry solvents were collected from an Innovative Technology Pure Solv solvent drying system. All chemicals were purchased from either Sigma Aldrich, Alfa Aesar or Fluorochem. Thin layer chromatography was performed using Sigma Aldrich silica gel 60A F254 plates, and a basic potassium permanganate solution was used as a visualisation stain. Flash column chromatography was performed using Sigma Aldrich silica gel pore size 60, 230-400 mesh as the solid support with indicated eluent. NMR spectra were recorded using a Jeol ECS 400 NMR spectrometer, operating at 400 MHz for ^{1}H nuclei and 100 MHz for ^{13}C nuclei. Chemical shifts are reported relative to TMS (^{1}H 0.00 ppm) and $CDCl_3$ (^{1}H 7.26 ppm, ^{13}C 77.0 ppm) and are reported in parts per million (ppm) on the δ scale. When reporting NMR, a and b protons refer to different geminal protons. Infrared spectra were recorded using a Thermo Nicolet 380 FT-IR. Mass spectra and accurate mass data were obtained on an Agilent Technologies 6540 UHD Accurate-Mass Q-TOF LC/MS.

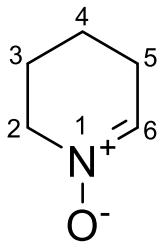
Piperdin-1-ol (18)



A solution of 1,5-dibromopentane (30g, 130 mmol) in triethylamine (100 mL) was added to a suspension of hydroxylamine hydrochloride (22.84g 0.325 mol,) in trimethylamine (130 mL) at r.t. The mixture was stirred under reflux for 4 h, cooled to r.t and diluted with diethyl ether (50 mL). The solution was filtered through a pad of Celite and the filtrate concentrated under reduced pressure to give a pale yellow oil. Purification by flash column chromatography on silica gel using CH_2Cl_2: MeOH (9.5:0.5) as the eluent gave *the title compound* (8.57 g, 65%) as a colourless oil.

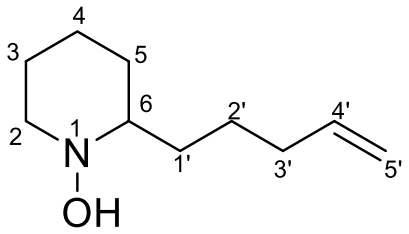
^{1}H NMR (400 MHz, $CDCl_3$) δ 3.37-3.19 (d, *J* = 9.9 Hz, 2H, 2a-H, 6a-H), 2.42-2.37 (ddd, *J* = 15.0, 9.9, 3.9 Hz, 2H, 2b-H, 6b-H), 1.75 – 1.61 (m, 2H, 3a-H, 5a-H), 1.54-1.48 (ddd, *J* = 16.5, 10.0, 3.4 Hz, 3H, 3b-H, 4a-H, 5b-H), 1.16 – 1.00 (m, 1H, 4b-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 58.98 (C2, C6), 25.34 (C3, C5), 23.03 (C4) **IR/cm^{-1}** 3248 (O-H) 2938 (C-H) 2834 (C-H) 1444 (N-O) **HRMS calcd for $C_5H_{11}NO$ [M+H]:** 102.0919 **found [M+H]**: 102.0915

2,3,4,5-Tetrahydropyridine 1-oxide (19)



Mercuric oxide (45.75 g, 212 mmol) in CH_2Cl_2 (100 mL) was added to a solution of **18** (8.57 g, 84.7 mmol) in CH_2Cl_2 (40 mL) at 0 °C. The mixture was allowed to warm to r.t. and stirred for 3h. The slurry was filtered through sodium sulphate and a pad of Celite. The filtrate was concentrated under reduced pressure to give *the title compound* as an orange oil. The crude product was used in the next step without purification.

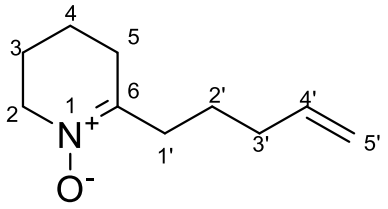
6-(pent-4’-en-5’-yl)piperidinol (20)



The synthesis was carried out according to the literature procedure.[88] A solution of freshly prepared pent-4-enylmagnesium bromide (101.64 mmol) in THF (60 mL) was added, via cannula addition, dropwise, over five minutes, to a solution of stirred nitrone **19** (8.57 g, 84.7 mmol) in THF (50 mL), at 0 °C. The solution was allowed to warm to r.t and left to stir for 20 h. Water (20 mL) was added and the mixture was concentrated under reduced pressure to give an orange residue. Sat aq. NH_4Cl (120 mL) was added and the residue extracted with diethyl ether (3 x 150 mL). The combined organic extracts were dried with anhydrous magnesium sulphate and concentrated under reduced pressure to give an orange oil. Purification by flash column chromatography on silica gel using diethyl ether: hexane (1:1) as the eluent gave *the title compound* (4.02 g, 47%) as a colourless oil.

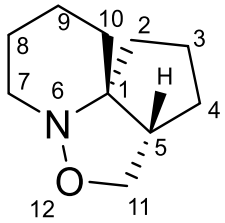
^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.90 – 5.73 (m, 1H, 4'-H), 4.98 (dd, 2H, 5'-H), 3.35 – 3.25 (m, 1H, 2a-H), 2.58 – 2.43 (m, 1H, 2b-H), 2.25 (tt, *J* = 13.8 & 6.8 Hz, 1H, 6-H), 1.91 – 1.82 (m, 1H,3a-H), 1.79 – 1.05 (m, 9H, 3b-H, 4-H, 5-H, 2'-H, 3'H). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 138.94 (C4'), 114.46 (C5'), 67.64 (C6), 34.23 (C2), 32.83 (C3'), 31.10 (C3), 26.98 (C4), 25.88 (C5), 25.37 (C2'), 23.83 (C1'). **IR/cm^{-1}:** 3168 (O-H), 2929 (C-H), 2836 (C-H), 1640 (C=C) **HRMS calcd for $C_{10}H_{20}NO$ [M+H]** 170.1544: **found [M+H]**: 170.1539.

6-(pent-4'-en-yl)-2,3,4,5-tetrahydropyridine 1-oxide (21)



A solution of **20** (4.02 g, 23.75 mmol) in dichloromethane (20 mL) was added to a stirred suspension of mercuric oxide (11.93g, 59.37 mmol) in CH_2Cl_2 (60 mL), at 0 °C. The mixture was allowed to warm to r.t. and stirred for 3h. The slurry was filtered through a pad of Celite/sodium sulphate and the filtrate was concentrated under reduced pressure to give *the title compound* as an orange oil. The crude product was used in the next step without purification.

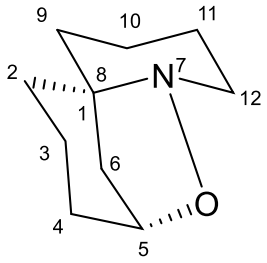
(1S*,5S*)-octahydro-1*H*-cyclopenta[3,4][1,2]oxazolo[6,12-*a*]pyridine (22)



A solution of nitrone **21** (4.02 g, 24.04 mmol) in toluene (40 mL) was stirred under reflux for 48 h. The solution was cooled to r.t and concentrated under reduced pressure to give a brown oil. Purification by flash column chromatography on silica gel using diethyl ether: hexane (1:1) as the eluent gave *the title compound* (2.09g, 52%) as a pale yellow oil.

^{1}H NMR (400 MHz, $CDCl_3$) δ: 4.30 (t, *J* = 8.5 Hz, 1H, 7a-H), 3.47 (dd, *J* = 8.3 & 5.0 Hz, 1H, 7b-H), 3.17 – 3.03 (m, 1H, 11a-H), 2.87 (m, 1H, 11b-H), 2.70 – 2.55 (m, 1H, 5-H), 2.02 – 1.58 (m, 8H, 8-H, 9-H, 3-H, 4-H), 1.57 – 1.37 (m, 4H, 2-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 75.43 (C1), 72.36 (C7), 50.58 (C11), 50.00 (C5), 39.39 (C8), 36.83 (C4), 33.19 (C9), 31.41 (C3), 24.15 (C10), 23.03 (C2) **IR/cm^{-1}:** 2935 (C-H), 2885 (C-H), 1460 (C-H), 1256 (C-N), 1042 (C-O). **HRMS calcd for $C_{10}H_{18}NO$ [M+H]:** 168.1388 **found [M+H]:** 168.1385

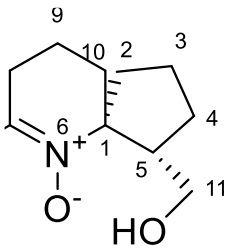
(1S*,5S*)-7-Oxa-6-azatricyclo[6.3.1.0^{1,6}]dodecane (23)



A solution of isoxazolidine **22** (1.67 g, 9.99 mmol) in toluene (8 mL) in a G30 microwave reaction vial was heated in a microwave reactor for 2.5 h at 220 °C . The solution was concentrated under reduced pressure to give a brown oil. Purification by flash column chromatography using diethyl ether: hexane (1:1) as the eluent gave *the title compound* as a pale orange oil (0.68g, 41%).

^{1}H NMR (400 MHz, $CDCl_3$) δ: 4.61 – 4.50 (m, 1H, 5-H), 3.27 (ddd, *J* = 10.5, 4.8, 3.0 Hz, 1H, 7a-H), 2.54 (ddd, *J* = 25.8, 15.8, 6.0 Hz, 2H, 4a-H, 7b-H), 2.06 – 1.90 (m, 2H, 5b-H, 6a-H), 1.79 – 1.19 (m, 11H, 2-H, 3-H, 4-H, 6b-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 75.47 (C5), 62.46 (C1), 55.39 (C7), 40.07 (C4), 38.44 (C6), 33.27 (C8), 31.50 (C2), 24.83 (C10), 19.70 (C3), 18.84 (C9). **IR/cm^{-1}** 2932 (C-H) 2853 (C-H) 1740 (C-H) 1310 (C-N) 1095 (C-O) **HRMS calcd for $C_{10}H_{18}NO$ [M+H]:** 168.1388 **found [M+H]:** 168.1376

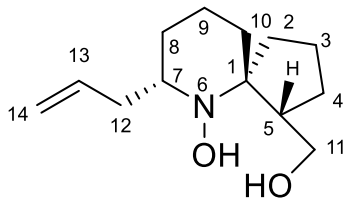
(1S*,5S*)-(6-oxido-6-azaspiro[4.5]dec-6-en-1-yl)methanol (105)



A solution of mCPBA (2.78, 14.89 mmol) in CH_2Cl_2 (70 mL) was added dropwise, over 7 h, to a solution of isoxazolidine **22** (1.86g, 11.12 mmol) in CH_2Cl_2 (50 mL), at 0 °C. The solution was stirred for 20 h at r.t. and then quenched with sat. aq. sodium bicarbonate: sodium thiosulphate (1:1) (80 mL) and stirred for 30 minutes. The solution was extracted with CH_2Cl_2 (3x40 mL) and the combined organic phases were dried with anhydrous magnesium sulphate and concentrated under reduced pressure to give an orange oil. Purification by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent gave *the title compound* (0.91g, 49%) as an orange solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.33 (t, *J* = 4.0 Hz, 1H, 7-H), 3.84 – 3.54 (m, 2H, 11-H), 2.74 (dt, *J* = 9.8, 5.6 Hz, 1H, 5-H), 2.52 – 2.39 (m, 2H, 8-H), 2.28 – 1.40 (m, 10H, 2-H 3-H, 4-H, 9-H 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 142.61(C7), 76.89(C1), 61.28(C11), 52.85(C5), 38.66(C8), 37.18(C4), 28.57(C10), 26.51(C2), 24.17(C9), 15.56(C3). **IR/cm^{-1}:** 3347 (O-H) 2941 (C-H) 2869 (C-H) 1709 (C=N). **HRMS calcd for $C_{10}H_{18}NO_2$** 184.1337 **found [M+H]:** 184.1332

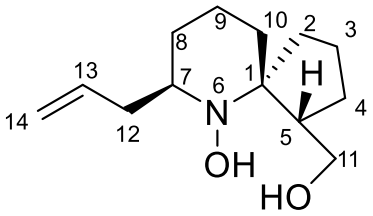
(1S*,5S*,7S*)-11-(hydroxymethyl)-7-allyl-6-azaspiro[4.5]decan-11-ol (163+161)



The synthesis was carried out according to the literature procedure.[88] A freshly prepared solution of allylmagnesium bromide (39.96 mmol) in diethyl ether (100 mL) was added dropwise, over 5 minutes, via cannula, to a solution of spironitrone 105 (2.4 g, 13.10 mmol) in anhydrous diethyl ether (40 mL) at 0 °C. The solution was stirred for 30 mins. at r.t and then quenched with sat. aq. ammonium chloride (25 mL). The mixture was extracted using CH_2Cl_2 (3 x 15 mL) and the combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a diastereomix of **161** and **163** as an orange oil (1.2 g, 48%). Flash column chromatography using EtOAc:cyclohexane (7:3) as the eluent afforded diastereoisomer **163** as a pale yellow oil. (0.67 g, 25 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.92 – 5.69 (m, 1H, 13-H), 5.22 – 4.92 (m, 2H, 14-H), 3.73 – 3.48 (m, 2H, 11-H), 3.17 – 2.99 (m, 1H, 7-H), 2.42 – 1.13 (m, 15H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H, 12-H). ^{13}C NMR (101 MHz, CDCl3) **δ ^{13}C NMR (101 MHz, $CDCl_3$) δ** 136.3 (C13), 116.7 (C14), 73.0 (C1), 65.5 (C11), 61.2 (C7), 43.3 (C5), 38.7 (C12), 37.5 (C8), 28.8 (C10), 27.7 (C2), 23.9 (C3), 20.4 (C4), 20.0 (C9). **IR/cm^{-1}** 3243 (O-H) 2936 (C-H) 2878 (C-H) 1443 (N-O) **HRMS calcd for $C_{13}H_{22}NO_2$ [M-H]** 224.1651**: found [M-H]** 224.1656

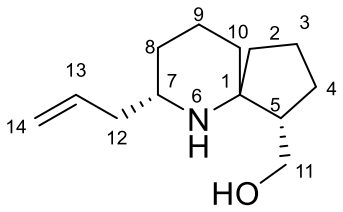
(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (161)



Sodium borohydride (4.4 g, 116.4 mmol) was added, portionwise, over 5 min to a solution of nitrone **147** (2.6 g, 11.64 mmol) in MeOH (180 mL) at 0 °C. The solution was stirred for 1 h at r.t then concentrated under reduced pressure. Purification by flash column chromatography using Et_2O:hexane (1:1) as the eluent gave *the title compound* as a yellow oil (1.4g, 54 %).

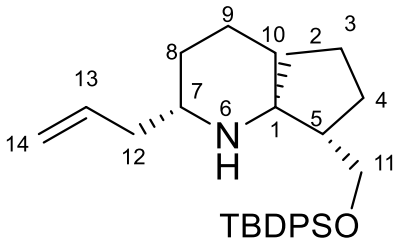
^{1}H NMR (400 MHz, $CDCl_3$) δ 5.80 – 5.64 (m, 1H, 13-H), 5.02 – 4.84 (m, 2H, 14-H), 3.80 (qd, *J* = 11.0, 6.5 Hz, 2H, 11-H), 2.59 – 2.45 (m, 1H, 7-H), 2.21 – 2.07 (m, 1H, 5-H), 2.05 – 1.84 (m, 2H, 12-H), 1.82 – 1.34 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H). ^{13}C NMR (101 MHz, **$CDCl_3$**) δ 136.08 (C13), 116.69 (C14), 72.50 (C1), 65.93 (C11), 63.84 (C7), 61.76 (C12), 59.95 (C5), 52.95 (C8), 49.18 (C4), 39.88 (C9), 38.28 (C3), 28.36 (10), 24.87 (C2) . **IR/cm^{-1}** 3275 (O-H) 2984 (C-H) 2845 (C-H) 1498 (N-O) **HRMS calcd for $C_{13}H_{22}NO_2$ found [M-H]** 224.1651**: found [M-H]** 224.1655

(1*S**,5*S**,7*R**)-11-(hydroxymethyl)-7-allyl-6-azaspiro[4.5]decane (164)



Zinc powder (0.84 g, 12.90 mmol) was added to a solution of hydroxylamine **163** (0.69, 3.07 mmol) in AcOH aq. (1:2) (18 mL), at r.t. The mixture was heated to 70 °C and stirred for 3 hours. The mixture was then quenched via addition of ammonium hydroxide (6 mL) and extracted with ethyl acetate (3 x 25 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give *the title compound* as a colourless oil (0.57 g, 83 %) The crude produce was used in the next step without purification.

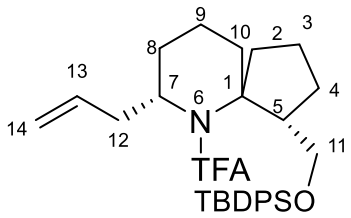
(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (165)



TBDPSCl (1.4 g, 5.4mmol) was added to a stirred solution of spiroamine **164** (0.56 g, 2.70 mmol), triethylamine (1.35 g, 13.50 mmol) and DMAP (0.01 g, 0.08 mmol) in CH_2Cl_2 (55 mL) at 0 °C. The solution was allowed to warm to r.t and stirred for 48 h. The solution was quenched by the addition of water (85 mL) and extracted with CH_2Cl_2 (3 x 45 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a pink oil. Purification by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent gave *the title compound* as a colourless oil (0.68 g, 56 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.79 – 7.60 (m, 4H, Ar), 7.56 – 7.28 (m, 6H, Ar), 5.29 (dddd, *J* = 14.8, 10.0, 8.8, 5.9 Hz, 1H, 13-H), 5.01 – 4.79 (m, 2H, 14-H), 3.56 (dd, *J* = 3.7 Hz, 1H, 11a-H), 3.45 (t, 1H, 11b-H), 2.31 – 1.31 (m, 15H, 2-H, 3-H, 4-H, 5-H, 7-H, 8-H, 9-H, 10-H, 12-H), 1.19 – 0.97 (m, 9H, tBu). **^{13}C NMR (101 MHz, $CDCl_3$) δ**: 135.87(C13), 135.79(Ar), 135.30(Ar), 133.82(Ar), 129.73(Ar), 127.74(Ar), 127.69(Ar), 117.65(C14), 63.86(C1), 63.75(C11), 51.98(C7), 41.81(C5), 40.70(C12), 35.82 (C8), 32.36(C10), 27.04(tBu), 26.17(C2), 21.51(C9), 19.29(C3), 19.09(C4). **IR/cm^{-1}** 3050 (N-H) 2930 (C-H) 2856 (C-H) 1823 (C=C Aromatic) 1110 (C-O) **HRMS calcd for $C_{29}H_{42}NOSi$ [M+H]** 448.3036**: found [M+H]** 448.3040

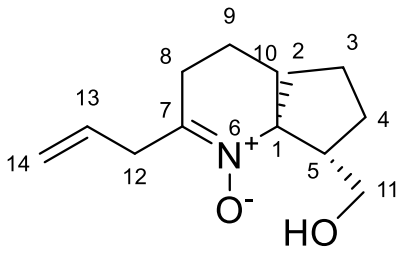
(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane-2,2,2-trifluoroactyl (166)



TFAA (2.2 g, 10.70 mmol) was cautiously added, dropwise, over 5 minutes to a solution of silyl ether **165** (0.48 g, 1.07 mmol) and Hunig's base (1.4 g, 10.70 mmol) in anhydrous CH_2Cl_2 (50 mL) at 0 °C. The solution was stirred for 4 h at r.t before being quenched with sat aq. $NaHCO_3$ (30 mL) and extracted using CH_2Cl_2 (3 x 25 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.40 g, 69 %).

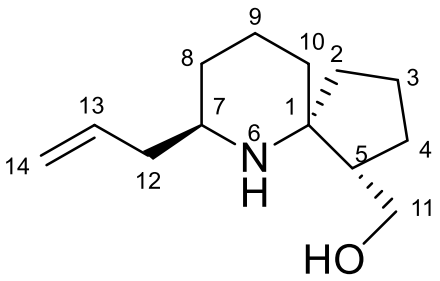
^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.70 – 7.59 (m, 4H, Ar), 7.46 – 7.29 (m, 6H, Ar), 5.71 – 5.51 (m, 1H, 13-H), 5.09 – 4.94 (m, 2H, 14-H), 3.83 – 3.71 (m, 1H, 7-H), 3.69 – 3.46 (m, 2H, 11-H), 2.59 – 2.43 (m, 1H, 5-H), 2.34 – 1.38 (m, 14H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H), 1.11 – 0.93 (s, 9H, tBu). **^{13}C NMR (101 MHz, $CDCl_3$) δ: ^{13}C NMR (101 MHz, $CDCl_3$) δ**: 158.00 (C=O), 135.72 (Ar), 135.63 (C13), 134.36 (Ar), 134.11 (Ar), 133.88 (Ar), 129.62 (Ar), 127.66 (Ar), 117.90 (C14), 115.38 (C-F_3), 68.11 (C1), 64.71 (C11), 54.81 (C5), 53.20 (C7), 41.93 (C12), 39.36 (C8), 37.95 (C10), 26.97(tBu), 24.53(C2), 22.26(C9), 19.31(C3), 13.57 (C4). **IR/cm^{-1}** 2934 (C-H) 2859 (C-H) 1780 (C=C Aromatic) 1680 (C=O) 1140 (C-O) **HRMS calcd for $C_{31}H_{39}F_3NO_2Si$ [M-H]** 542.2702**: found [M-H]** 542.2761

(1S*,5S*)-7-allyl-1-(hydroxymethyl)-6-azaspiro[4.5]dec-6-ene 6-oxide (167)



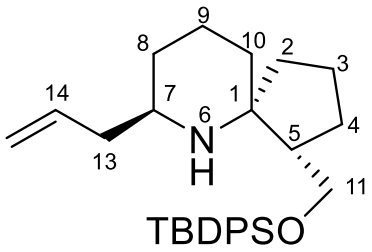
Mercuric oxide (13.00 g, 60.18) was added to a solution of a diastereomeric mixture of allylated spirocycles **163** + **161** (2.7g, 12.04 mmol) in CH_2Cl_2 at 0 °C and the mixture was stirred at r.t for 4 h then filtered through a pad of Celite. The filtrate was dried using anhydrous sodium sulfate and concentrated under reduced pressure to give *the title compound* as an orange oil. The product was used in the next step without further purification.

(1*S**,5*S**,7*R**)-1-(hydroxymethyl)-7-allyl-6-azaspiro[4.5]decane (168)



Zinc powder (0.86 g, 13.11 mmol) was added to a stirred solution of hydroxylamine **161** (0.7 g, 3.12mmol) in AcOH aq. (1:2) (18 mL), at r.t. The mixture was stirred for 3 h at 70 ^{o}C then quenched by addition of 14 M aqueous ammonium hydroxide (5 mL) and extracted using ethyl acetate (3 x 25 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give *the title compound* as a colourless oil (0.68 g, 99%) The crude product was used in the next step without further purification.

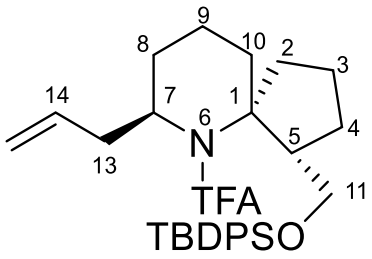
(1*S**,5*S**,7*R**)-5-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (169)



Triethylamine (2.66 g, 26.29 mmol), TBDPSCl (2.62 g, 9.55 mmol) and DMAP (0.001 g, 0.004 mmol) were added to a solution of spiroamine **168** (1.00 g, 4.78 mmol),) in CH_2Cl_2 (80 mL) at 0 °C. The solution was allowed to warm to r.t and stirred for 48 h. The solution was quenched by the addition of water (60 mL) and extracted using CH_2Cl_2 (3 x 45 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a pink oil. Purification by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent gave *the title compound* as a colourless oil (1.99 g, 88 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.76 – 7.62 (m, 4H, Ar), 7.48 – 7.33 (m, 6H, Ar), 5.81 – 5.68 (m, 1H, 13-H), 4.99 (dd, *J* = 24.5, 13.6 Hz, 2H, 14-H), 3.77 (dt, *J* = 10.2, 5.2 Hz, 2H, 11-H), 2.75 – 2.60 (m, 1H, 5-H), 2.14 – 1.98 (m, 2H, 12-H), 1.82 – 1.32 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H), 1.05 (s, 9H, tBu). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 136.14 (Ar), 135.80 (Ar), 134.98 (Ar), 133.89 (Ar), 129.62 (C13), 116.82 (C14), 64.43 (C1), 63.03 (C7), 52.26 (C12), 51.34 (C11), 42.27 (C5), 37.04 (C8), 35.69 (C10), 33.15 (C2), 28.03 (C4), 27.31 (tBu), 22.46 (C9), 19.40 (C3). **IR/cm^{-1}** 2930 (C-H) 2858 (C-H) 1428 (C=C Aromatic) 1112 (C-O) **HRMS calcd for $C_{29}H_{42}NOSi$** 448.3036: **found [M+H]** 448.3030

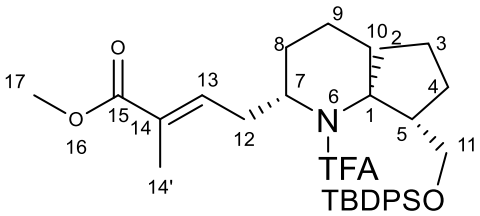
(1*S**,5*S**,7*R**)-5-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane-2,2,2-trifluoroactyl (170)



TFAA (8.9 g, 42.5 mmol) was added, dropwise, over 5 minutes to a stirred solution of silyl ether **169** (1.9 g, 4.25 mmol) and Hunig's base (5.4 g, 42.5 mmol) in CH_2Cl_2 (300 mL), at 0 °C. The solution was stirred for 4 h at r.t then quenched by the addition of sat aq. $NaHCO_3$ (35 mL) and extracted using CH_2Cl_2 (3 x 25 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.95 g, 45

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.69 – 7.56 (m, 4H, Ar), 7.47 – 7.30 (m, 6H, Ar), 5.72 – 5.51 (m, 1H, 13-H), 5.07 (dd, 2H, 14-H), 3.78 (br s, 1H, 7-H), 3.69 – 3.48 (m, 2H, 11-H), 2.57 – 2.42 (m, 1H, 12a-H), 2.37 – 1.37 (m, 14H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H, 11b-H), 1.04 (s, 9H, ^{t}Bu). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 157.20 (C=O), 135.87 (Ar), 134.53 (Ar), 129.43 (Ar), 127.71 (Ar), 126.37 (C13), 117.91 (C14), 115.80 (CF_3), 68.55 (C1) , 64.45 (C11), 57.32 (C5), 54.68 (C7) 49.03 (C12), 41.32 (C8), 35.28 (C4), 26.11 (tBu), 24.77 (C10), 23.30 (C2), 19.54 (C3), 13.96 (C9). **IR/cm^{-1}** 2942 (C-H) 2860 (C-H) 1583 (C=O) 1188 (C-O) **HRMS calcd for $C_{31}H_{40}F_3NO_2SiNa$**: 566.2679 **found [M+Na]** 566.2685

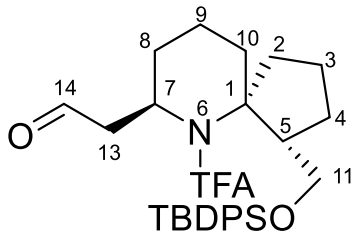
(E)-methyl-14'-((1S,5S,7S)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-13-methylbut-14-enoate (175)



Methyl acrylate (0.02 g, 0.23 mmol) was added to a solution of protected amide **166** (0.023 g, 0.05 mmol) and Grubbs' 2nd Gen. catalyst (0.005 g, 0.005 mmol) in toluene (1 mL) at r.t. The solution was stirred under reflux for 3 h, cooled to r.t and concentrated under reduced pressure to give the crude product as a brown oil. Purification by flash column chromatography using Et_2O:hexane (8:2) as the eluent gave *the title compound* as a colourless oil (0.02 g, 64 %).

^{1}H NMR (400 MHz,) δ 7.70 – 7.56 (m, 6H, Ar), 7.49 – 7.29 (m, 4H, Ar), 6.55 (t, *J* = 6.9 Hz, 1H, 13-H), 3.87 (d, *J* = 10.7 Hz, 1H, 7-H), 3.73 (s, 3H, 17-H), 3.64 (dd, *J* = 10.2, 7.3 Hz, 1H, 11a-H), 3.50 (dd, *J* = 10.3, 4.1 Hz, 1H, 11b-H), 2.83 – 2.68 (m, 1H, 12a-H), 2.59 – 2.45 (m, 1H,8a-H), 2.29 (d, *J* = 12.6 Hz, 1H, 12b-H), 2.16 – 2.01 (m, 2H, 5-H 8b-H), 1.99 – 1.35 (m, 13H, 2-H, 3-H, 4-H, 9-H, 10-H, 14'-H), 1.09 – 0.97 (s, 9H, ^{t}Bu). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 168.25 (C=O), 168.20 (C=O), 137.00 (Ar), 135.61 (C13), 134.03 (Ar), 133.81 (Ar), 130.02 (Ar), 129.67 (C14), 127.69 (CF_3), 68.15 (C11), 64.66 (C1), 56.80 (C5), 52.85 (C7), 52.03 (C17), 39.42 (C8), 36.46 (C4), 35.53 (C14'), 26.96 (C12), 24.55 (^{t}Bu), 19.29 (C10), 14.20 (C2), 13.78 (C3), 12.85 (C9). **IR/cm^{-1}** 2952 (C-H) 2859 (C-H) 1719 (C=O) 1687 (C-O) 1472 (C-O) **HRMS calcd for $C_{34}H_{41}F_3NO_2Si$ [M+H]** 616.3058 **found [M+H]** 616.3069

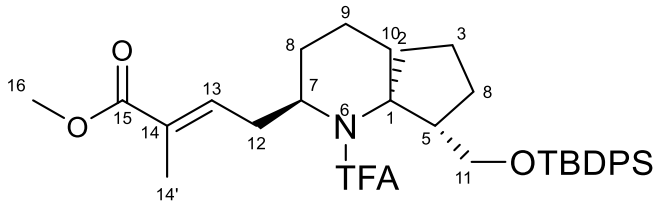
2-((1S,5S,7R)-1-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)acetaldehyde (176)



Osmium(IV) tetraoxide, 4 % w.t/v *t*BuOH (0.001 g, 0.054 mmol) was added dropwise over 5 minutes to a solution of protected amine **170** and *N*-methylmorpholine *N*-oxide (0.126 g, 1.08 mmol) in acetone (1.5 mL) at r.t and stirred for 3 hr. Sat. aq. sodium sulfite (5 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 5 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give a yellow oil. The crude product was dissolved in MeOH (2 mL) and sodium periodate (0.3 g, 1.4 mmol) was added at r.t and the mixture stirred for 2 h. The solution was diluted with water (10 mL) and extracted with CH_2Cl_2 (3 x 5 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give *the title compound* as a pale yellow oil (0.06 g, 75 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 9.38 (s, 1H, 13-H), 7.62 (ddd, *J* = 7.9, 4.2, 1.5 Hz, 4H, Ar), 7.47 – 7.31 (m, 6H, Ar), 4.52 (dd, *J* = 4.2 Hz, 1H, 7-H), 3.68 (dd, *J* = 10.7, 6.0 Hz, 1H, 11a-H), 3.56 (dd, 1H, 11b-H), 2.68 – 2.65 (m, 2H, 12-H), 2.22 (td, *J* = 11.7, 8.2 Hz, 5H), 2.10 – 1.52 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H), 1.05 (s, 9H, ^{t}Bu).). **^{13}C NMR (101 MHz, $CDCl_3$) δ**^{13}C NMR (101 MHz,) δ 198.29 (C13), 157.56 (C=O), 135.66 (Ar), 134.18(Ar), 133.56(Ar), 129.85(Ar), 127.85(Ar), 118.23 (CF_3), 68.24 (C11), 64.64 (C1), 56.85 (C5), 50.19 (C12), 46.71 (C7) 34.94 (C4), 30.80 (C8), 29.80, (C2) 27.18(^{t}Bu), 24.68 (C10), 19.35 (C3), 13.79 (C9). **IR/cm^{-1}** 2955 (C-H) 2859 (C-H) 1687 (C=O) 1428 (C-O) **HRMS calcd for $C_{30}H_{37}F_3NO_3Si$** 544.2495 **found [M-H]** 544.2492

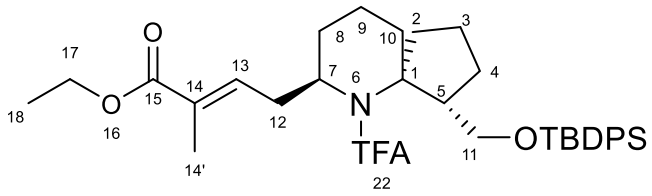
(E)-methyl-14-((1S,5S,7R)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-2-methylbut-2-enoate (177)



Methyl 2-triphenylphosphoranylidene propanoate (0.08 g, 0.24 mmol) was added, dropwise, to a solution of aldehyde **176** (0.09 g, 0.16 mmol) in CH_2Cl_2 at r.t and the mixture stirred for 8h. The solution was concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.10 g, 75 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65 – 7.58 (m, 4H, Ar), 7.44 – 7.28 (m, 6H, Ar), 6.61 – 6.43 (m, 1H, 13-H), 3.91 – 3.78 (m, 1H, 7-H), 3.75 – 3.72 (m, 3H, 15-H), 3.52 (ddd, *J* = 19.8, 10.1, 4.6 Hz, 2H, 11-H), 2.86 – 2.69 (m, 1H, 13a-H), 2.53 (dt, *J* = 14.4, 9.1 Hz, 1H, 13b-H), 2.36 – 2.03 (m, 3H, 5-H, 8-H), 1.96 – 1.47 (m, 13H, 2-H, 3-H, 4-H, 9-H, 10-H, 16-H), 1.03 (s, 9H, ^{t}Bu). ^{13}C NMR (101 MHz,) δ 168.26 (C=O), 141.37 (Ar), 137.01 (C13), 135.62 (Ar), 133.66 (Ar), 129.68 (Ar), 127.70 (C14), 118.10 (CF_3), 68.03 (C11), 64.46 (C1), 56.80 (C7), 54.87 (C20), 52.83 (C5), 39.42 (C12), 36.46 (C8), 30.84 (C21), 26.97 (^{t}Bu), 24.56 (C2), 23.32 (C10), 19.30 (C9), 14.08 (C3) **IR/cm^{-1}** 2955 (C-H) 2859 (C-H) 1687 (C=O) 1428 (C-O) **HRMS calcd for $C_{34}H_{41}F_3NO_2Si$ [M+H]** 616.3492 **found [M+H]** 616.3452

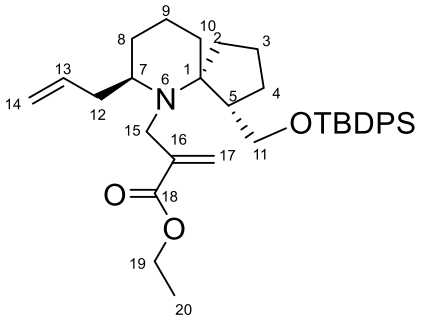
(E)-ethyl-5-((1S,5S,7R)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-2-methylbut-2-enoate(178)



Triethyl 2-phosphono propanionate (0.14g, 0.48 mmol) was added dropwise to a slurry of sodium hydride (0.02 mg, 0.96 mmol) in THF (3 mL) and the mixture left to stir for 1 hr at r.t. A solution of aldehyde **176** (0.09 g, 0.16 mmol) in THF (0.5 mL) was added, via canula, and the mixture stirred at r.t for 48 h. The reaction was quenched *via* addition of sat. aq. ammonium chloride (3 mL) and brine (3 mL) and extracted with EtOAc (3 x 5 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.10 g, 90 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.56 (dt, *J* = 8.0, 1.4 Hz, 4H, Ar), 7.41 – 7.23 (m, 6H, Ar), 6.42 (dd, *J* = 11.3, 4.1 Hz, 1H, 13-H), 4.24 – 4.07 (m, 2H, 16-H) 3.80 (d, *J* = 11.3 Hz, 1H, 7-H), 3.55 (ddd, *J* = 16.1, 10.5, 6.3 Hz, 2H, 11-H), 2.56 – 2.38 (m, 1H, 12a-H) 2.25 – 1.17 (m, 20H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H, 12b-H, 15-H, 17-H), 0.96 (s, 9H, ^{t}Bu). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 167.84 (C=O), 156.92 (C=O), 136.61 (Ar), 135.64 (C13), 135.57 (Ar), 134.02 (Ar), 133.66 (Ar), 130.23 (Ar), 129.70 (Ar), 127.73 (C14), 118.34(CF_3), 68.33 (C11), 64.45 (C1), 60.86 (C15), 56.80 (C7), 52.71 (C5), 35.52 (C4), 35.28 (C8), 30.85 , 27.06 (^{t}Bu), 24.56 (C15), 23.31 (C2), 19.25 (C17), 14.35 (C10), 14.20 (C9), 12.71 (C3). **IR/cm^{-1}** 2931 (C-H) 2857 (C-H) 1689 (C=O) 1428 (C-O) **HRMS calcd for $C_{35}H_{47}F_3NO_4Si$[M+H]** 630.3226 **found [M+H]** 630.3227

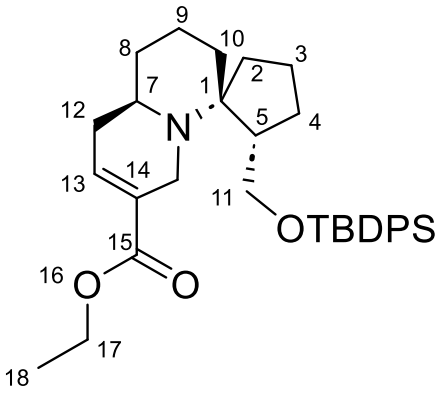
ethyl-2-(((1S,5S,7R)-7-allyl-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-azaspiro[4.5]decan-6-yl)methyl)acrylate (180)



A solution of silyl ether **169** (0.23 g, 0.51 mmol) and K_2CO_3 (0.36 g, 2.57 mmol) in acetonitrile (2.5 mL) in a G30 microwave reaction vial was heated in a microwave reactor for 10 h at 60 °C. The mixture was diluted with toluene (20 mL) and concentrated under reduced pressure to give a dark yellow oil. Purification by flash column chromatography using Et_2O:hexane (9.5:0.5) as the eluent gave *the title compound* as a pale yellow oil (0.20 g, 67 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.71 – 7.54 (m, 4H, Ar), 7.51 – 7.31 (m, 6H, Ar), 5.92 (d, *J* = 1.9 Hz, 1H, 17a-H), 5.75 (d, *J* = 2.0 Hz, 1H, 17b-H), 5.61 – 5.41 (m, 1H, 13-H), 4.95 – 4.77 (m, 1H, 14-H), 4.13 (q, 1H, 19-H), 3.82 (m, 1H, 11a-H), 3.30-3.38 (m, 2H, 11b-H, 15a-H) 3.11-3.06 (m, 1H, 15b-H) 2.63 (br s, 1H, 7-H), 2.08-1.13 (m, 18H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H, 12-H, 20-H), 1.02 (s, 9H, tBu). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 166.97 (C=O), 139.89 (Ar), 137.30 (Ar), 135.68 (C16), 135.62 (Ar), 134.34 (Ar), 134.30 (Ar), 129.43 (C17), 127.57 (C13), 115.98 (C14), 69.59 (C1), 63.96 (C11), 60.35 (C19), 53.53 (C7), 52.77 (C12), 39.95 (C15), 32.26 (C5), 29.80(C8), 29.52 (C20) 27.03 (tBu), 20.86 (C10), 20.60 (C2), 19.32 (C9), 14.30 (C3) **IR/cm^{-1}** 2931 (C-H) 2857 (C-H) 1709 (C=O) 1639 (C-O) **HRMS calcd for $C_{35}H_{50}NO_3Si$[M+H]** 560.3559 **found [M+H]** 560.3554

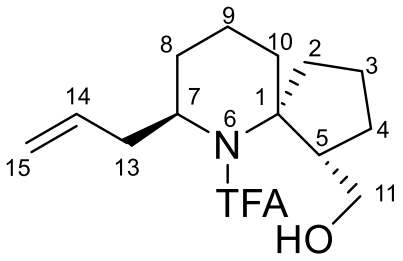
(1S,5S,7R)-ethyl-11-(((tert-butyldiphenylsilyl)oxy)methyl)-1',2',3',6',9',9a'-hexahydrospiro[cyclopentane-1,4'-quinolizine]-7'-carboxylate (181)



Grubbs' 2nd Generation catalyst (0.005 g, 0.005 mmol) was added to a solution of alkylated tertiary amine **180** (0.19 g, 0.34 mmol) in anhydrous CH_2Cl_2 (30 mL) at r.t. The solution was stirred under reflux for 2 h. then cooled to r.t and concentrated under reduced pressure to give a brown oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.13 g, 52 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.75 – 7.58 (m, 4H, Ar), 7.50 – 7.27 (m, 6H, Ar), 6.84 (m, 1H, 13-H), 4.16 (q, *J* = 7.1 Hz, 2H, 17-H), 3.93 (dd, *J* = 10.4, 4.6 Hz, 1H, 11a-H), 3.55 (t, *J* = 9.9 Hz, 1H, 11b-H), 3.40 (d, *J* = 17.0 Hz, 1H, 15a-H), 3.10 (d, *J* = 17.0 Hz, 1H, 15b-H), 2.67 – 2.53 (m, 1H, 7-H), 2.40 – 2.25 (m, 1H, 12a-H), 2.08 – 1.19 (m, 17H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H, 12b-H, 18-H), 1.01 (s, 9H, tBu). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 166.06 (C=O), 136.07 (Ar), 135.75 (Ar), 134.21 (C13), 129.43 (Ar), 128.55 (Ar), 127.61 (C14), 67.62 (C1), 63.68 (C11), 60.24 (C17), 53.21 (C5), 51.06 (C7), 44.13 (C15), 34.16 (C12), 31.69 (C10), 30.55 (C2), 28.89 (C8), 26.95 (tBu), 21.42 (C18), 19.29 (C9), 14.37 (C3) **IR/cm^{-1}** 2921 (C-H) 2853 (C-H) 1716 (C=O) 1456 (C-O) **HRMS calcd for $C_{33}H_{46}NO_3Si$[M+H** 532.3246 **found [M+H]** 532.3253

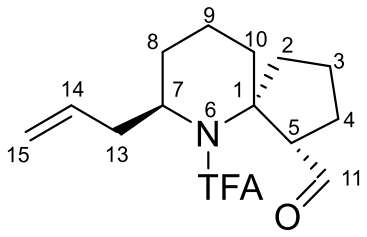
1-((1S,5S,7R)-7-allyl-1-(hydroxymethyl)-6-azaspiro[4.5]decan-6-yl)-2,2,2-trifluoroethanone (187)



PPTS (4.22 g, 16.04 mmol) was added to a solution of silyl ether **195** (0.25 g, 0.60 mmol) in methanol (48 mL) at r.t. The solution was left to stir for 3 h at r.t. then quenched by addition of sat. aq. $NaHCO_3$ (100 mL) and extracted with CH_2Cl_2 (3 x 75 mL). The combined organic phases were dried using anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (1:1) as the eluent gave *the title compound* as a pale yellow oil (0.12 g, 66 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.80 – 5.54 (m, 1H, 14-H), 5.09 (dd, *J* = 13.5, 7.2 Hz, 2H, 15-H), 3.91 (d, *J* = 10.9 Hz, 1H, 7-H), 3.69 – 3.45 (m, 2H, 11-H), 2.66 – 2.45 (m, 2H, 13-H), 2.39 – 1.34 (m, 13H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 157.62 (C=O), 134.29 (C14), 118.30 (C15), 115.50 (CF_3), 68.96 (C1), 63.70 (C11), 56.18 (C5), 53.51 (C7), 41.33 (C13), 36.38 (C4), 34.59 (C8), 30.53 (C10), 24.69 (C2), 23.01 (C3), 14.49 (C9). **IR/cm^{-1}** 3431.2 (O-H) 2933.2 (C-H) 2859 (C-H) 1685 (C=O) 1137 (C-O) **HRMS calcd for $C_{15}H_{23}F_3NO_2$ [M+H]** 306.1680**: found [M+H]** 306.1682

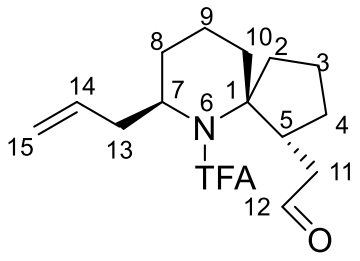
(1S,5S,7R)-7-allyl-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decane-1-carbaldehyde (188)



Dess-Martin periodinane (0.2 g, 0.26 mmol) was added to a solution of spiroalcohol **187** (0.04 g, 0.13 mmol) and pyridine (0.07 g, 0.78 mmol) at 0 °C in CH_2Cl_2. The solution was left to stir for 2 h then quenched by the addition of sat aq. $NaHCO_3$ (20 mL) and sat aq. sodium bisulfite (5 mL). The mixture was extracted using CH_2Cl_2 (3 x 15 mL) and the combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (8:2) as the eluent gave *the title compound* as a colourless oil (0.03 g, 70 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 9.35 (s, 1H, 11-H), 5.87 – 5.62 (m, 1H, 14-H), 5.10 (dd, *J* = 11.3, 9.7, 0.8 Hz, 2H, 15-H), 3.93 (d, *J* = 8.2, 4.3 Hz, 1H, 7-H), 2.80 – 2.39 (m, 3H, 5-H, 11-H), 2.21 (dd, *J* = 10.9, 7.7 Hz, 2H, 4-H), 2.07 – 1.37 (m, 10H, 2-H, 3-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 199.60 (C11), 156.22(C=O), 134.47 (C14), 118.06 (C15), 115.25 (CF_3), 68.67 (C1), 62.57 (C7), 53.71 (C5), 40.49 (C13), 35.32 (C4), 34.90 (C8) 27.45 (C10), 25.10 (C2), 22.28 (C9), 14.17 (C3) **IR/cm^{-1}** 2954 (C-H) 1711 (C=O) 1667 (C=O) **HRMS calcd for $C_{15}H_{19}F_3NO_2$ [M-H]** 302.1367**: found [M-H]** 302.1371

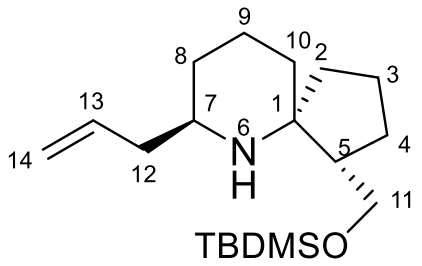
2-((1R,5S,7R)-7-allyl-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-1-yl)acetaldehyde (189)



A solution of (methoxymethyl)triphenylphosphonium chloride (0.08 g, 0.23 mmol) in THF (5 mL) was added to potassium tert-butoxide (0.02 g, 0.21 mmol) at 0 °C and the mixture stirred for 40 mins at r.t. A solution of aldehyde **188** (0.05 g, 0.16 mmol) in acetonitrile (2 mL) was added at 0 °C and the mixture stirred for 24 h at r.t. The reaction mixture was quenched with water (10 mL) and extracted with Et_2O (3 x 10 mL). The combined organic phases were dried with anhydrous sodium sulfate to give the crude product as an orange oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a colourless oil (0.03 g, 60 %).

^{1}H NMR (400 MHz, CD_3OD) δ 9.93 (s, 1H, 12-H), 5.86 – 5.56 (m, 1H, 14-H), 5.04 (dd, *J* = 13.4, 8.1 Hz, 2H, 15-H), 3.95 (d, , *J* = 5.4, 1H, 7-H), 2.72 – 2.45 (m, 6H, 4-H, 3a-H, 5-H, 11-H), 2.42 – 2.11 (m, 2H, 13-H), 1.97 – 1.17 (m, 9H, 2-H, 3b-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz , CD_3OD) δ** 188.05 (C12), 165.26 (C=O), 138.93 (C14), 132.67 (C15), 119.36 (CF_3), 49.33 (C7), 38.75 (C11), 38.23 (C5), 37.58(C4), 35.01(C3), 33.90 (C13), 30.38(C8), 28.05 (C10), 24.58 (C2), 21.39(C9). **IR/cm^{-1}** 2923 (C-H) 2859 (C-H) 1711 (C=O) 1667 (C=O) 1440 (C=C) **HRMS calcd for $C_{16}H_{23}F_3NO_2$[M+H]** 318.1680**: found [M+H]** 318.1669

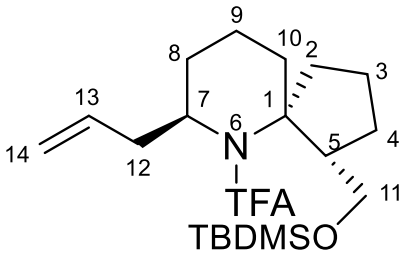
(1S,5S,7R)-7-allyl-1-(((tert-butyldimethylsilyl)oxy)methyl)-6-azaspiro[4.5]decane (194)



A solution of TBDMSCl (1.2 g, 7.88 mmol) in CH_2Cl_2 (10 mL) was added to a solution of spiroamine **168** (0.55 g, 2.63 mmol), triethylamine (1.60 g, 15.78 mmol) and DMAP (0.01 g, 0.004 mmol) in CH_2Cl_2 (60 mL) at 0 °C. The solution was allowed to warm to r.t and stirred for 48 hours. The mixture was quenched by the addition of water (85 mL) and extracted with CH_2Cl_2 (3 x 45 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a pink oil. Purification by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent gave *the title compound* as a colourless oil (0.4 g, 47 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.80 – 5.59 (m, 1H, 13-H), 5.40 - 5.21 (dd, *J* = 61.2, 13.5 Hz, 2H, 14-H), 4.22 – 4.00 (m, 2H, 11-H), 3.22 – 3.09 (m, 1H, 7-H), 2.88 – 2.71 (m, 1H, 12a-H), 2.68 – 2.53 (m, 1H, 12b-H), 2.29 – 1.35 (m, 13H, 2-H, 3-H, 4-H, 5-H, 8-H, 9-H, 10-H), 0.96 – 0.85 (s, 9H, tBu), 0.10 (s, 6H, $SiMe_2$). **^{13}C NMR (101 MHz, $CDCl_3$) δ:** 130.86 (C13), 121.98 (C14), 70.67 (C1), 67.74 (C11), 63.66 (C7), 53.36 (C5), 51.32(C12), 37.40 (C10) , 35.80 (C2) , 33.85 (C8), 28.17 (C4), 26.11 (tBu), 24.56 (C9), 20.52 (C3), -5.30 ($SiMe_2$). **IR/cm^{-1}** 2928 (C-H) 2857 (C-H) 1250 (C-O) **HRMS calcd for $C_{19}H_{38}NOSi$[M+H]** 324.2722**: found [M+H]** 324.2730

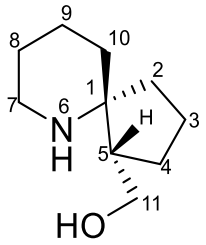
1-((1S,5S,7R)-7-allyl-1-(((tert-butyldimethylsilyl)oxy)methyl)-6-azaspiro[4.5]decan-6-yl)-2,2,2-trifluoroethanone (195)



TFAA (1.60 g, 7.70 mmol) was added, dropwise, over 5 minutes to a stirred solution of silyl ether **194** (0.25 g, 0.77 mmol) and Hunig's base (0.99 g, 7.70 mmol) in CH_2Cl_2 (35 mL), at 0 °C. The solution was stirred for 4 h at r.t then quenched by the addition of sat aq. $NaHCO_3$ (40 mL) and extracted using CH_2Cl_2 (3 x 25 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a yellow oil. Purification by flash column chromatography using EtOAc:hexane (9:1) as the eluent gave *the title compound* as a pale yellow oil (0.21 g, 65 %).

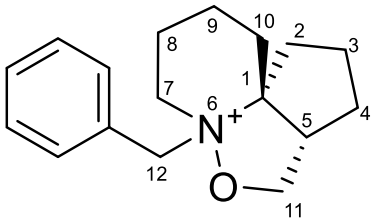
^{1}H NMR (400 MHz, $CDCl_3$) δ 5.79 – 5.62 (m, 1H, 14-H), 5.15 – 5.01 (m, 2H, 15-H), 3.93 – 3.83 (m, 1H, 7-H), 3.55 (d, *J* = 6.3 Hz, 2H, 11-H), 2.53 (t, *J* = 8.9 Hz, 2H, 13-H), 2.34 – 2.01 (m, 3H, 5-H, 8-H), 1.96 – 1.05 (m, 12H, 2-H, 3-H, 4-H, 9-H, 10-H), 0.87 (s, 9H, ^{t}Bu), 0.02 (s, 6H, $SiMe_2$). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 156.99 (C=O), 134.61 (C14), 117.87 (C15), 115.66 (CF_3), 68.32 (C1), 63.60 (C5), 56.96 (C7), 53.41 (C13), 40.99 (C8), 35.29 (C10), 30.60 (C2), 26.10 (^{t}Bu), 24.63(C4), 22.77 (C3), 18.48 (C9), -5.20 ($SiMe_2$). **IR/cm^{-1}** 2953 (C-H) 2859 (C-H) 1686 (C=O) 1137 (C-O) **HRMS calcd for [M+H]** **$C_{21}H_{37}F_3NO_2Si$** 420.2545**: found [M+H]** 420.2546

(1S*,5S*)-6-azaspiro[4.5]dec-1-ylmethanol (204)



Zinc powder (2.95 g, 45.12 mmol) was added to a solution of isoxazolidine **22** (0.71 g, 4.25 mm) in acetic acid: water (1:2) (50 mL), at r.t. The solution was stirred under reflux for 4 h then cooled to r.t. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give *the title compound* (0.51 g, 72 %) as a white solid. The product was used in the next step without purification.

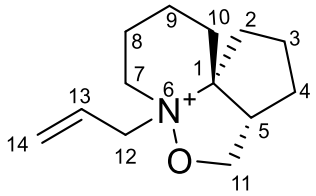
(1S*,5S*)6-benzyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (212)



Benzyl bromide (0.18 g, 4.8 mmol) was added, dropwise, over 1 minute, to a stirred solution of isoxazolidine **22** (0.2 g, 1.20 mmol) in $CHCl_3$ (14 mL) at r.t. The solution was stirred for 24 hours at r.t. then toluene (90 mL) was added and the mixture concentrated under reduced pressure to give an orange oil. Addition of hexane (50 mL) at 0 °C followed by filtration of the resulting solid gave *the title compound* as a light brown solid (0.26 g, 85 %). The product was used in the next step without further purification.

^{1}H NMR (400 MHz, CD_3OD-d_4) δ 7.65 – 7.42 (m, 5H, Ar), 5.08 (m, 1H, 12a-H), 4.89 – 4.80 (m, 1H, 12b-H), 4.59 (t, , *J* = 8.4 Hz, 1H, 11a-H), 4.24 (m, 1H, 11b-H), 3.56 – 3.40 (m, 2H, 5-H, 7a-H), 3.36 – 3.19 (m, 1H, 7b-H), 2.68 – 2.51 (m, 1H, 2a-H), 2.35 – 1.73 (m, 11H, 2b-H, 3-H, 4-H, 8-H, 9-H, 10-H). ^{13}C NMR (101 MHz,) δ 132.12 (Ar), 130.39 (Ar), 128.95 (Ar), 89.34 (C12), 75.68 (C11), 61.53 (C1), 57.38 (C7), 44.98 (C5), 34.17 (C8), 28.76 (C2), 27.38 (C10), 21.80 (C4), 21.53 (C9), 17.85 (C3). **IR/cm^{-1}** IR 2943 (C-H) 2872 (C-H) 1455 (C-O) **HRMS calcd for $C_{17}H_{24}NO^+$** 258.1858: **found [M]** 258.1870

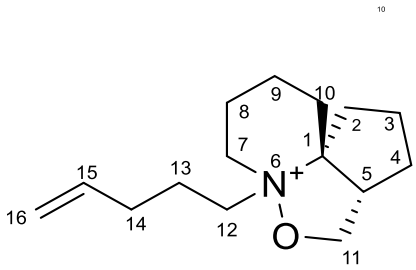
(1S*,5S*)-6-allyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (213)



Allyl bromide (0.15 g, 1.2 mmol) was added, dropwise, over 1 minute, to a solution of isoxazolidine **22** (0.05 g, 0.30 mmol) in $CHCl_3$ (14 mL), The solution was stirred for 48 hours at r.t. before toluene (50 mL) was added. The mixture was concentrated under reduced pressure to give an orange oil. Addition of hexane (50 mL) followed by filtration of the resulting solid gave *the title compound* as a light orange solid (0.06, 88 %). The product was used in the next step without further purification.

^{1}H NMR (400 MHz, CD_3OD-d_4) δ 5.93 – 5.79 (m, 2H, 14a-H, 13-H), 5.61 (m, 1H, 14b-H), 5.06 – 4.87 (m, 2H, 7a-H, 11a-H), 4.16 – 3.92 (m, 3H, 7b-H, 11b-H, 12a-H), 3.83 – 3.65 (m, 2H, 5-H, 12b-H), 2.50 – 2.36 (m, 1H, 8a-H), 2.28 (dd, *J* = 19.6, 4.5 Hz 1H, 4a-H), 2.19 – 1.67 (m, 10H, 2-H, 3-H, 4b-H, 8b-H, 9-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 128.47 (C14), 124.45 (C13), 88.94 (C11), 76.60 (C7), 60.87(C1), 58.81 (C12), 44.88 (C5), 35.09 (C4), 29.37 (C8), 28.66 (C3), 22.47(C9), 21.72(C2), 18.02 (C10). **IR/cm^{-1}** 2939 (C-H) 2876 (C-H) 1456 (C=C) **HRMS calcd for $C_{13}H_{22}NO^+$** 208.1701: **found [M]** 208.1703

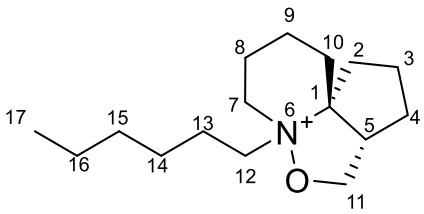
(1S*,5S*)-6-(pent-4-en-1-yl)octahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (214)



A solution of isoxazolidine **22** (0.02 g, 0.12 mmol) and 5-bromopent-1-ene (0.14 g, 0.48 mmol) in $CHCl_3$ (9 mL) in a G30 microwave reaction vial was heated in the microwave reactor for 3 hours at 110 °C (4.5 bars). Toluene (90 mL) was added and the mixture concentrated under reduced pressure to give a black oil. Addition of hexane (60 mL) followed by filtration of the resulting solid gave a brown solid which was purified by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent to give *the title compound* as a light brown solid (0.015 g, 55 %)

^{1}H NMR (400 MHz, CD_3OD-d_4) δ 5.94 – 5.74 (m, 1H, 15-H), 5.17 – 4.97 (m, 2H, 16-H), 4.68 – 4.56 (m, 1H, 11a-H), 4.19 – 4.09 (m, 1H, 11b-H), 4.01 – 3.83 (m, 2H, 7a-H, 12a-H), 3.67 – 3.40 (m, 3H, 5-H, 7b-h, 12b-H), 2.43 – 1.63 (m, 16H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H, 13-H, 14-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 136.37 (C15), 115.49 (C16), 89.26 (C1), 75.43 (C11), 57.76 (C12), 57.09 (C5), 44.87 (C7), 33.98 (C4), 29.92 (C8), 28.66 (C13), 27.26 (C14), 22.14 (C10), 21.86 (C2), 21.36 (C3), 17.87 (C9). **IR/cm^{-1}** 2933 (C-H) 2833 (C-H) 1444 (C-O) **HRMS calcd for $C_{15}H_{26}NO^+$** 236.2009: **found [M]** 236.2014

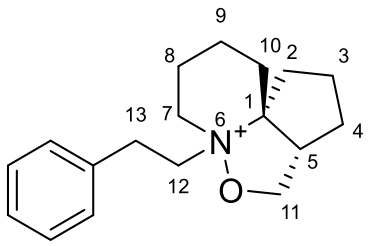
(1S*,5S*)-6-hexyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (215)



A solution of isoxazolidine **22** (0.02 g, 0.12 mmol) and 1-bromohexane (0.17 g, 0.48 mmol) in $CHCl_3$ (9 mL) in a G30 microwave reaction vial was heated via the microwave reactor for 3 hours at 130 °C (4.5 bars) Toluene (90 mL) was added and the mixture concentrated under reduced pressure to give a black oil. Addition of hexane (60 mL) followed by filtration of the resulting solid gave a brown solid which was purified by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent to give *the title compound* as a light brown solid (0.013 g, 47 %)

^{1}H NMR (400 MHz, CD_3OD-d_4) δ δ 2.99 (dt, *J* = 23.7, 8.0 Hz, 2H, 11-H), 2.76 – 2.46 (m, 3H, 5-H 7a-H, 12a-H), 2.02 – 1.26 (m, 21H, 2-H, 3-H, 4-H, 7b-H, 8-H, 9-H, 10-H, 12b-H, 13-H, 14-H, 15-H, 16-H), 0.91 (td, *J* = 6.7, 2.7 Hz, 3H. 17-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 59.53 (C1), 52.37 (C11), 41.41 (C5), 35.80 (C12), 35.72 (C7), 31.71 (C4), 31.08(C13), 27.67 (C8), 27.34 (C10), 25.93 (C2), 25.73 (C14), 24.63 (C15), 22.15(C9), 21.50 (C3), 20.04 (C16), 12.97 (C17). **IR/cm^{-1}** 2932 (C-H) 2859 (C-H) 1656 (C-O) **HRMS calcd for $C_{16}H_{30}NO^+$** 252.2322: **found [M]** 252.2327

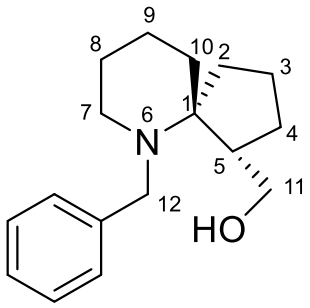
(1S*,5S*)-6-phenethyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (216)



A solution of isoxazolidine **22** (0.02 g, 0.12 mmol) and 2-bromoethylbenzene (0.19 g, 0.48 mmol) in $CHCl_3$ (9 mL) in a G30 microwave reaction vial was heated via the microwave reactor for 6 hours at 130 °C (4.5 bars) Toluene (90 mL) was added and the mixture concentrated under reduced pressure to give a black oil. Addition of hexane (60 mL) followed by filtration of the resulting solid gave a brown solid which was purified by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent to give *the title compound* as a light brown solid (0.012 g, 44 %)

^{1}H NMR (400 MHz, CD_3OD-d_4) δ 7.46 – 7.13 (m, 5H, Ar), 4.72 – 4.55 (m, 1H, 13a-H), 4.08 – 3.89 (m, 1H, 13b-H), 3.60 – 3.47 (m, 2H, 7a-H, 12a-H), 3.25 – 2.91 (m, 3H, 5-H, 7b-H, 12b-H), 2.09 – 1.58 (m, 14H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 128.79 (Ar), 128.47 (Ar), 127.17 (Ar), 89.62 (C13), 75.60 (C1), 58.53 (C11), 44.91 (C7), 33.98 (C12), 28.97 (C5), 28.57 (C4), 27.21 (C8), 21.92 (C2), 21.36 (C10), 18.68 (C3), 17.83 (C9). **IR/cm^{-1}** 2930 (C-H) 2845 (C-H) 1631 (C-O) 1455 (C=C) **HRMS calcd for $C_{18}H_{26}NO^+$** 272.2009 **found [M]** 272.2008

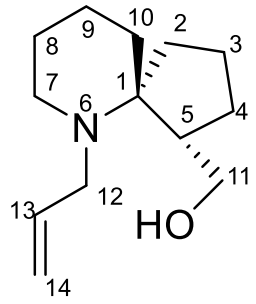
(1S*,5S*) -(6-benzyl-6-azaspiro[4.5]decan-1-yl)methanol (217)



Zinc powder (0.75 g, 11.61 mmol) was added to a solution of ammonium salt 212 (0.2 g, 0.77 mmol) in in acetic acid: water (1:2) (20 mL) at r.t. The solution was stirred under reflux for 4 h then cooled to r.t. A solution of aq. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a colourless oil. (0.1 g, 48 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.35 – 7.10 (m, 5H, Ar), 3.96 – 3.81 (m, 2H, 7a-H, 12a-H), 3.67 (dd, *J* = 11.5, 6.2 Hz, 1H, 7b-H), 3.56 (d, *J* = 13.6 Hz, 1H, 12b-H), 2.93 – 2.69 (m, 1H, 11a-H), 2.46 – 2.28 (m, 2H, 8a-H, 11b-H), 2.09 – 1.91 (m, 1H, 4a-H), 1.87 – 1.45 (m, 9H, 2-H, 3a-H, 4b-H, 5-H, 8b-H, 9a-H, 10-H), 1.33 – 1.14 (m, 2H, 3b-H, 9b-H) **^{13}C NMR (101 MHz, $CDCl_3$) δ** 139.72 (Ar), 128.57 (Ar), 127.04 (Ar), 68.95 (C7), 65.72 (C1), 60.49 (C12), 54.39 (C11), 47.45 (C5), 45.45 (C8), 33.31 (C4), 27.87 (C2), 21.62 (C10), 21.15 (C9), 14.31 (C3). **IR/cm^{-1}** 3345 (O-H) 2932 (C-H) 2861 (C-H) 1494 (C=C) 1443 (C-O) **HRMS calcd for $C_{17}H_{26}NO$ [M+H]** 260.2014: **found [M+H]** 260.2009

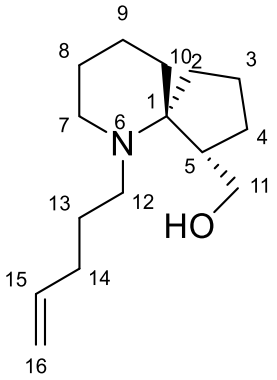
(1S*,5S*)-(6-allyl-6-azaspiro[4.5]decan-1-yl)methanol (218)



Zinc powder (2.95 g, 45.12 mmol) was added to a solution of ammonium salt 213 (0.2 g, 0.77 mmol) in acetic acid: water (1:2) (20 mL) at r.t. The solution was stirred under reflux for 4 h then cooled to r.t.. A solution of aq. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a colourless oil. (0.11 g, 55 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.86 – 5.60 (m, 1H, 13-H), 5.17 – 4.99 (m, 2H, 14-H), 3.76 – 3.55 (m, 2H, 11-H), 3.34 – 3.16 (m, 1H, 12a-H), 2.97 – 2.77 (m, 2H, 7a-H, 12b-H), 2.36 – 2.12 (m, 2H, 5-H, 7b-H), 1.80 – 1.04 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H). **^{13}C NMR (101 MHz, $CDCl_3$) δ** 139.56 (C13), 116.83 (C14), 68.60 (C1), 65.60 (C11), 60.44 (C12), 54.69 (C7), 48.57 (C5), 36.54 (C8), 31.27 (C4), 28.90 (C2), 22.82 (C10), 21.85 (C9), 14.25 (C3). **IR/cm^{-1}** 3343 (O-H) 2931 (C-H) 2862 (C-H) 1641 (C=C) 1442 (C-O) **HRMS calcd for $C_{13}H_{23}NO$ [M+H]** 210.1858: **found [M+H]** 210.1869

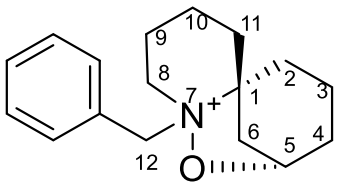
(1S*,5S*)-(6-(pent-4-en-1-yl)-6-azaspiro[4.5]decan-1-yl)methanol (219)



Zinc powder (0.83 g, 14.4 mmol) was added to a solution of ammonium salt **214** (0.2 g, 0.84 mmol) in acetic acid: water (1:2) (20 mL) at r.t. The solution was stirred under reflux for 4 h then cooled to r.t.. A solution of aq. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a colourless oil. (0.09, 45 %)

^{1}H NMR (400 MHz, CD_3OD -d_4) 5.84 (ddt, *J* = 16.9, 10.2, 6.7 Hz, 1H, 15-H), 5.18 – 5.00 (m, 2H, 16-H), 4.65 – 4.56 (m, 1H, 11a-H), 4.13 (dd, *J* = 8.2, 6.8 Hz, 1H, 11b-H), 4.00 – 3.78 (m, 2H, 7a-H, 12a-H), 3.66 – 3.39 (m, 2H, 5-H, 7b-H), 2.40 – 1.71 (m, 16H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H, 12b-H, 13-H, 14-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 138.29 (C15), 113.95 (C16), 64.23 (C11), 61.26 (C1), 58.91 (C7), 50.52 (C5), 41.69 (C12), 35.66 (C8), 34.11 (C4), 27.07 (C10), 24.93 (C2), 24.26(C14), 22.82 (C13), 21.54 (C9), 20.78 (C3). **IR/cm^{-1}** 3288 (O-H) 2929 (C-H) 2860 (C-H) 1697 (C=C) 1444 (C-O) **HRMS calcd for $C_{15}H_{28}NO$ [M+H]:** 238.2171 **found [M+H]** 238.2172

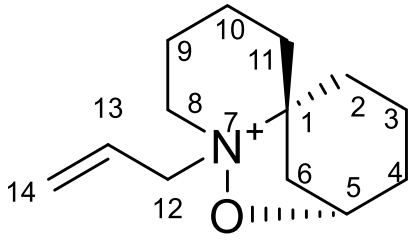
(1S*,5S*)-7-benzyloctahydro-6H-2,5a-methanopyrido[1,2-b][1,2]oxazepin-10-ium bromide (220)



Benzyl bromide (0.18 g, 4.8 mmol) was added, dropwise, over 1 minute, to a stirred solution of isoxazolidine **23** (0.06 g, 0.35 mmol) in $CHCl_3$ (14 mL), at r.t. The solution was stirred for 24 hours at r.t. then toluene (90 mL) was added and the mixture concentrated under reduced pressure to give an orange oil. Addition of hexane (50 mL) at 0 °C gave the *title compound* as a light brown solid (0.26 g, 79 %). The product was used in the next step without the need for further purification.

^{1}H NMR (400 MHz, CD_3OD-d_4) δ 7.68 – 7.57 (m, 2H, Ar), 7.54 – 7.44 (m, 3H, Ar), 5.17 – 5.05 (m, 1H, 5-H), 5.06 – 4.96 (m, 2H, 12-H), 3.52 (t, *J* = 13.5 Hz, 1H, 6a-H), 3.42 – 3.31 (m, 2H, 6b-H, 8a-H), 2.38 – 1.93 (m, 9H, 2-H, 4-H, 8b-H, 10-H, 11-H), 1.82 (ddd, *J* = 17.5, 13.1, 5.1 Hz, 4H, 3-H, 9-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 132.28 (Ar), 130.29 (Ar), 128.95 (Ar), 128.48 (Ar), 128.20 (Ar), 82.88 (C12), 80.06 (C1), 59.81 (C8), 58.02 (C5), 38.66 (C6), 32.99 (C2), 30.11 (C11), 28.98 (C10), 22.31 (C4), 17.85 (C9), 16.98 (C3). **IR/cm^{-1}** 2944 (C-H) 2872 (C-H) 1456 (C-O) **HRMS calcd for $C_{17}H_{24}NO^+$** 258.1852 **found [M]** 258.1854

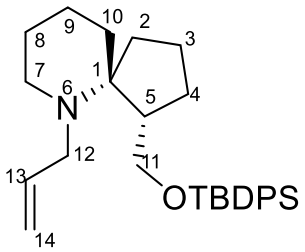
(1S*,5S*)-7-allyloctahydro-5H-2,5a-methanopyrido[1,2-b][1,2]oxazepin-12-ium bromide (221)



Allyl bromide (0.15 g, 1.4 mmol) was added, dropwise, over 1 minute, to a solution of isoxazolidine **23** (0.08 g, 0.45 mmol) in $CHCl_3$. The solution was stirred for 48 hours at r.t. before toluene (50 mL) was added. The mixture was concentrated under reduced pressure to give an orange oil. Addition of hexane (50 mL) followed by filtration of the resulting solid gave *the title compound* as a light orange solid (0.06, 84 %). The product was used in the next step without the need for purification.

^{1}H NMR (400 MHz, CD_3OD-d_4) 6.16 – 6.01 (m, 1H, 13-H), 5.70 (dd, *J* = 26.6, 13.6 Hz, 2H, 14-H), 5.12 – 5.02 (m, 1H, 5-H), 4.44 (tdd, *J* = 14.1, 8.8, 5.4 Hz, 2H, 12-H), 3.91 (d, *J* = 13.1 Hz, 1H, 8a-H), 3.72 – 3.58 (m, 1H, 8b-H), 3.35 (dd, *J* = 10.1, 7.2 Hz, 1H, 6a-H), 2.29 – 1.61 (m, 13H, 2-H, 3-H, 4-H, 6b-H, 9-H, 10-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 126.32 (C13), 125.71 (C14), 82.69 (C1), 79.27 (C5), 60.46 (C12), 57.05 (C8), 38.59 (C6), 33.04 (C11), 29.95 (C2), 28.86 (C10), 21.92 (C3), 17.49 (C9), 16.83 (C4). **IR/cm^{-1}** 2946 (C-H) 2846 (C-H) 1464 (C-O) **HRMS calcd for $C_{13}H_{22}NO^+$** 208.1696 **found [M]:** 208.1701

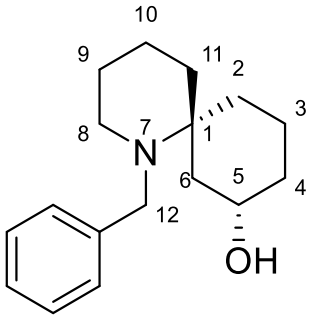
(1S*,5S*)-6-allyl-1-(((tert-butyldiphenylsilyl)oxy)methyl)-6-azaspiro[4.5]decane (224)



TBDPSCl (1.3 g, 4.78 mmol) was added to a stirred solution of spiroamine 218 (0.25 g, 1.19 mmol), triethylamine (0.66g, 6.54 mmol) and DMAP (0.01 g, 0.08 mmol) in CH_2Cl_2 (25 mL) at 0 °C. The solution was allowed to warm to r.t and stirred for 48 hours. The solution was quenched by the addition of water (mL) and extracted with CH_2Cl_2 (3 x 45 mL). The combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure to give the crude product as a pink oil. Purification by flash column chromatography using CH_2Cl_2: MeOH (9:1) as the eluent gave *the title compound* as a colourless oil (0.26g, 58 %).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.69 (ddd, *J* = 9.2, 6.5, 1.5 Hz, 4H, Ar), 7.42 – 7.29 (m, 6H, Ar), 5.51 (dddd, *J* = 17.1, 10.2, 7.1, 4.7 Hz, 1H, 13-H), 5.03 – 4.86 (m, 2H, 14-H), 3.89 (dd, *J* = 10.5, 4.2 Hz, 1H, 11a-H), 3.52 (t, *J* = 10.3 Hz, 1H, 11b-H), 3.07 – 2.95 (m, 1H, 12a-H), 2.74 (dd, *J* = 14.4, 7.0 Hz, 1H, 12b-H), 2.48 – 2.35 (m, 1H, 7a-H), 2.24 – 2.11 (m, 1H, 7b-H), 2.08 – 1.98 (m, 1H, 5-H), 1.87 – 1.24 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H), 1.10 – 1.00 (m, 9H, ^{t}Bu). **(101 MHz, $CDCl_3$) δ** 137.56 (Ar), 135.82 (Ar), 135.78 (C13), 134.88 (Ar), 129.48 (Ar), 127.81 (Ar), 115.35 (C14), 66.82 (C1), 64.02 (C11), 53.14(C5), 46.97 (C7), 28.34 (C8), 27.07 (C2), 26.58 (C10), 21.80 (C4), 21.73 (C3), 19.36 (C9). **IR/cm^{-1}** 2930 (C-H) 2856 (C-H) 1427 (C-O) **HRMS calcd for $C_{29}H_{42}NOSi$ [M+H]:** 448.3035 **found [M+H]** 448.3039

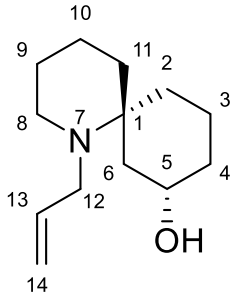
(1S*,5S*)-7-benzyl-1-azaspiro[5.5]undecan-8-ol (222)



Zinc powder (0.75 g, 11.61 mmol) was added to a solution of ammonium salt **220** (0.2 g, 0.75 mmol) in acetic acid: water (1:2) (20 mL) at r.t. The solution was stirred under reflux for 4 h then cooled to r.t.. A solution of aq. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a colourless oil.. (0.11 g, 54 %)

^{1}H NMR (400 MHz, $CHCl_3$-*d*) δ 7.36 – 7.16 (m, 5H, Ar), 4.06 (d, *J* = 14.6 Hz, 1H, 5-H), 3.95 (d, *J* = 12.9 Hz, 1H, 12a-H), 3.69 – 3.59 (m, 1H, 12b-H), 2.95 (t, *J* = 12.0 Hz, 1H, 8a-H), 2.64 (d, *J* = 12.5 Hz, 1H, 6a-H), 2.48 (d, *J* = 14.7 Hz, 1H, 8b-H), 1.99 (ddd, *J* = 15.3, 11.2, 4.7 Hz, 2H, 4a-H, 9a-H), 1.82 (d, *J* = 9.8 Hz, 1H, 4b-H), 1.73 – 1.41 (m, 7H, 2-H, 6b-H, 9-H, 11-H), 1.36 – 1.10 (m, 4H, 3-H, 10-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 139.86 (Ar), 128.67 (Ar), 128.64 (Ar), 127.04 (Ar), 67.88 (C1), 56.70 (C5), 49.40 (C12), 42.69 (C8), 36.31 (C6), 35.94 (C4) , 34.39 (C9), 32.66 (C2), 20.10 (C11), 19.08 (C3), 16.35 (C10). **IR/cm^{-1}** 3366 (O-H) 2926 (C-H) 2858 (C-H) 1687 (C=C Aromatic) **HRMS calcd for $C_{17}H_{26}NO$[M+H]:** 260.2014 **found [M+H]** 260.2021

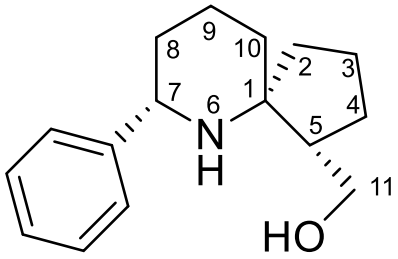
(1S*,5S*)-7-allyl-1-azaspiro[5.5]undecan-8-ol (223)



Zinc powder (2.95 g, 45.12 mmol) was added to a solution of ammonium salt **221** (0.2 g, 0.96 mmol) in acetic acid: water (1:2) (20 mL) at r.t. The solution was stirred under reflux for 4 h then cooled to r.t.. A solution of aq. 1M NaOH (75 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 40 mL). The combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. Purification by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a colourless oil. (0.11 g, 55 %)

^{1}H NMR (400 MHz, $CHCl_3$-*d*) 5.75 (dddd, *J* = 17.3, 10.0, 8.5, 4.5 Hz, 1H, 13-H), 5.10 (dd, *J* = 25.0, 13.5 Hz, 2H, 14-H), 3.97 (br s, 1H, 5-H), 3.38 – 3.29 (m, 1H, 12a-H), 3.14 (dd, *J* = 13.6, 8.4 Hz, 1H, 12b-H), 3.03 (t, *J* = 13.0 Hz, 1H, 8a-H), 2.76 (d, *J* = 13.6 Hz, 1H, 8b-H), 2.56 (d, *J* = 11.7 Hz, 1H, 6a-H), 1.90 – 1.34 (m, 9H, 2-H, 4-H, 6b-H, 9-H, 10-H), 1.27 – 1.03 (m, 4H, 3-H, 9-H). **^{13}C NMR (101 MHz, CD_3OD-d_4) δ** 137.41 (C13), 116.76 (C14), 67.75 (C1), 60.49 (C5), 56.39 (C12), 48.51 (C8), 43.23 (C6), 35.95 (C2), 34.37 (C11), 32.78 (C4), 19.92 (C9), 16.27 (C3), 14.27 (C10). **IR/cm^{-1}** 3321 (O-H) 2928 (C-H) 2863 (C-H) 1451 (C-O)) **HRMS calcd for $C_{13}H_{24}NO$ [M+H]:** 210.1858 **found [M+H]** 210.1862

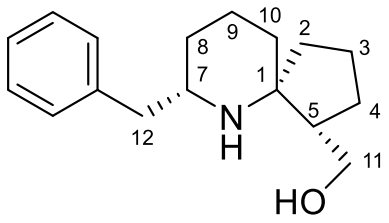
(1S*,5S*,7S*)-7-phenyl-6-azaspiro[4.5]decan-1-yl)methanol (229)



The synthesis was carried out according to the literature procedure.[88] A freshly prepared solution of phenylmagnesium bromide (1.2 mmol) in diethyl ether (40 mL) was added dropwise, over 5 minutes, via cannula addition, to a solution of spironitrone **105** (0.15 g, 0.82 mmol) in anhydrous diethyl ether (20 mL) at 0 °C. The solution was stirred for 30 mins. at r.t and then quenched with sat. aq. ammonium chloride (40 mL). The mixture was extracted using CH_2Cl_2 (3 x 15 mL) and the combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give an orange oil. The crude residue was dissolved in acetic acid: water (1:2) (15 mL) and zinc powder (0.32g, 4.82 mmol) was added at r.t. The reaction was stirred at 70 °C for 6 h then cooled to r.t and quenched using ammonium hydroxide (5 mL). The mixture was extracted using EtOAc (3 x 15 mL) and the combined organic phases were dried using anhydrous sodium sulfate and concentrated under reduced pressure. Purification of the residue by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a pale yellow oil. (0.12 g, 60 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.56 (dd, *J* = 18.8, 12.4 Hz, 2H, Ar), 7.39 – 7.20 (m, 3H, Ar), 4.62 (dd, *J* = 13.1, 2.4 Hz, 1H, 7-H), 3.66 (dd, *J* = 19.8, 8.1 Hz, 1H, 11a-H), 3.47 (dt, *J* = 20.6, 10.3 Hz, 1H, 11b-H), 2.60 (ddd, *J* = 11.4, 7.1, 4.1 Hz, 1H, 5-H), 2.41 – 2.21 (m, 1H, 8a-H), 1.95 – 1.12 (m, 13H, 2-H, 3-H, 4-H, 6-H, 8b-H, 9-H, 10-H), **(101 MHz, $CDCl_3$) δ**: 136.64 (Ar), 128.95 (Ar), 128.76 (Ar), 128.71 (Ar), 68.94 (C11), 63.35 (C1), 59.57 (C7), 40.40(C5), 33.96 (C8), 32.04 (C2), 28.78 (C10), 25.02 (C4), 20.03 (C3), 18.49 (C9) **IR/cm^{-1}** 3318 (O-H) 2936 (C-H) 2861 (C-H) 1431 (C=C Aromatic) **HRMS calcd for $C_{16}H_{24}NO$[M+H] :** 246.1857 **found [M+H]** 246.1856

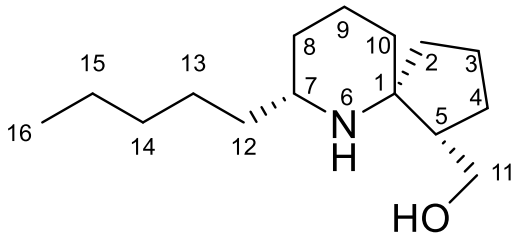
(1S*,5S*,7S*)-7-benzyl-6-azaspiro[4.5]decan-1-yl)methanol (230)



The synthesis was carried out according to the literature procedure.[88] A freshly prepared solution of benzylmagnesium bromide (4 mmol) in diethyl ether (40 mL) was added dropwise, over 5 minutes, via cannula, to a solution of spironitrone 105(0.1 g, 0.55 mmol) in anhydrous diethyl ether (10 mL) at 0 °C. The solution was stirred for 30 mins. at r.t, and then quenched with sat. aq. ammonium chloride (30 mL). The mixture was extracted with CH_2Cl_2 (3 x 15 mL) and the combined organic phases were dried with anhydrous magnesium sulphate and concentrated under reduced pressure to give an orange oil. The crude residue was dissolved in acetic acid: water (1:2) and zinc powder (0.23 g, 3.53 mmol) was added at r.t. The reaction was stirred at 70 ^{o}C for 6 h then cooled to r.t and quenched using ammonium hydroxide (2 mL). The mixture was extracted using EtOAc (3 x 15 mL) and the combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure. Purification of the residue by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a pale yellow oil. (0.05 g, 35 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37 – 7.11 (m, 5H, Ar), 4.00 – 3.87 (m, 1H, 12a-H), 3.81 – 3.67 (m, 1H, 12b-H), 3.51 – 3.28 (m, 1H, 7-H), 3.01 (t, *J* = 8.0 Hz, 2H, 5-H 11a-H), 2.85 (dd, *J* = 15.6, 9.1 Hz, 1H, 11b-H), 2.05 – 1.17 (m, 12H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H). **(101 MHz, $CDCl_3$) δ**: 129.46 (Ar), 129.26 (Ar), 128.85 (Ar), 128.78 (Ar), 126.88 (Ar), 126.72 (Ar), 66.48 (C12), 62.40 (C1), 54.72 (C7), 50.09 (C5), 41.91 (C11), 35.23 (C8), 34.23 (C4), 30.06 (C2), 26.82 (C10), 22.62 (C3), 21.19 (C9). **IR/cm^{-1}** 3290 (O-H) 2930 (C-H) 2862 (C-H) 1603 (C-O) 1453 (C=C) **HRMS calcd for $C_{17}H_{26}NO$[M+H]** 260.2014 **found [M+H]** 260.2021

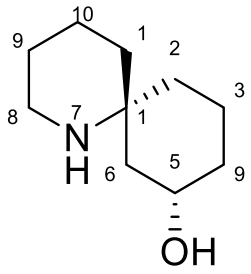
(1S*,5S*,7S*)-7-pentyl-6-azaspiro[4.5]decan-1-yl)methanol (231)



The synthesis was carried out according to the literature procedure.[88] A freshly prepared solution of pentylmagnesium bromide (5.5 mmol) in diethyl ether (40 mL) was added dropwise, over 5 minutes, via cannula, to a solution of spironitrone 105 (0.1 g, 0.55 mmol) in anhydrous diethyl ether (10 mL) at 0 °C. The solution was stirred for 30 mins. at r.t. and then quenched with sat. aq. ammonium chloride (20 mL). The mixture was extracted using CH_2Cl_2 (3 x 15 mL) and the combined organic phases were dried using anhydrous magnesium sulphate and concentrated under reduced pressure to give a yellow oil. The crude residue was dissolved in acetic acid: water (1:2) (5 mL) and zinc powder (0.23 g, 3.53 mmol) was added at r.t. The reaction was stirred at 70 °C for 6 h then cooled to r.t and quenched with ammonia hydroxide (2 mL). The mixture was extracted with EtOAc (3 x 15 mL) and the combined organic phases were dried with anhydrous sodium sulfate and concentrated under reduced pressure. Purification of the residue by flash column chromatography using CH_2Cl_2:MeOH (9:1) as the eluent gave *the title compound* as a pale yellow oil. (0.06 g, 46 %)

^{1}H NMR (400 MHz, $CDCl_3$) δ: ^{1}H 3.61 (dd, *J* = 16.2, 10.5 Hz 2H, 11-H), 2.77 – 2.62 (m, 1H, 7-H), 2.16 – 2.04 (m, 1H, 5-H), 1.91 – 1.12 (m, 19H, 2-H, 3-H, 4-H, 8-H, 9-H, 10-H, 12-H, 13-H, 14-H, 15-H), 0.88 – 0.80 (m, 3H, 16-H). **(101 MHz, $CDCl_3$) δ**: 65.80 (C11) 64.08 (C1), 53.39 (C7), 41.64 (C5), 41.20 (C8), 37.98 (C4), 37.45 (C12), 33.08 (C10), 27.83 (C2), 25.62 (C13), 22.68 (C14), 22.64 (C15), 21.05 (C3), 20.48 (C9), 14.16 (C16). **IR/cm^{-1}** 3373 (O-H) 2926 (C-H) 2858 (C-H) 1458 (C-H) **HRMS calcd for $C_{15}H_{30}NO$[M+H]** 240.2327 **found [M+H]** 240.2325

(1S*,5S*)-1-azaspiro[5.5]undecan-8-ol (235)



Zinc powder (0.51 g, 7.50 mmol) was added to a solution of isoxazolidine **23** (0.30 g, 1.79 mmol) in acetic acid: water 1:2 (9 mL), at r.t. The solution was stirred under reflux for 4 h then cooled to r.t. and aq. ammonium hydroxide (3 mL) added. The mixture was extracted with CH_2Cl_2 (3 x 40 mL) and the combined organic phases were dried with anhydrous magnesium sulphate and concentrated under reduced pressure to give *the title compound* as a white solid (0.25 g, 83 %)

9. References

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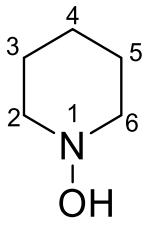
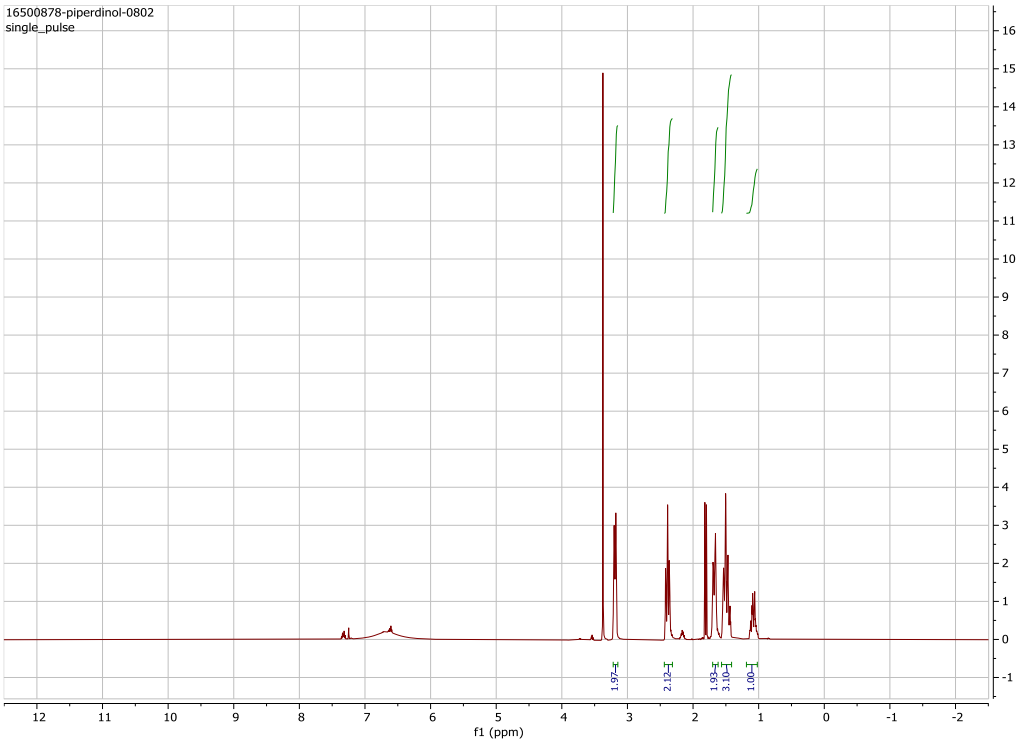
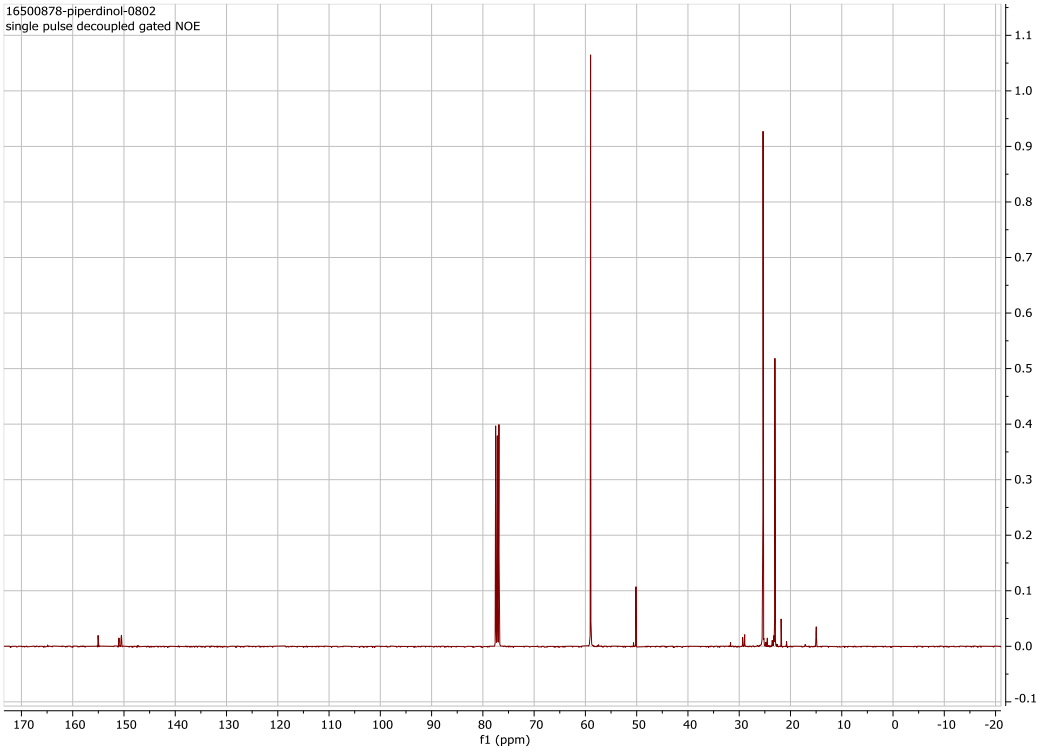
10. Appendix

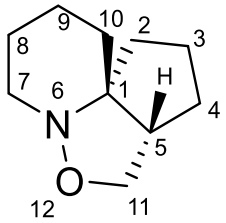
Piperdin-1-ol (18)

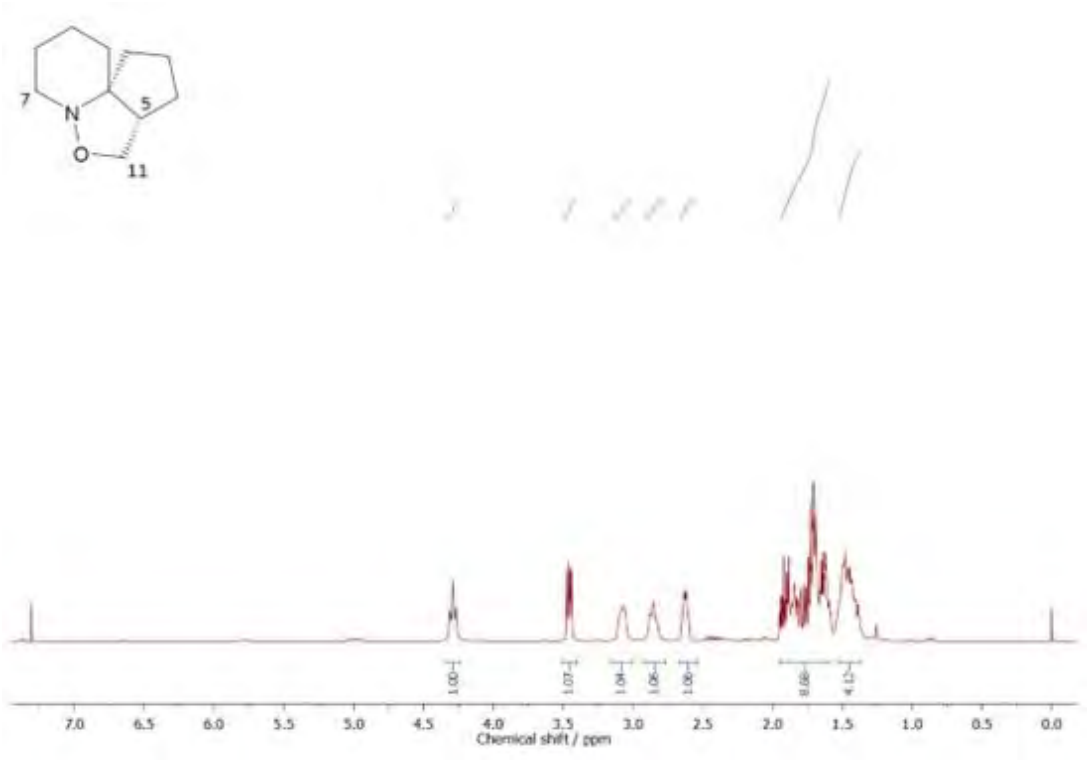




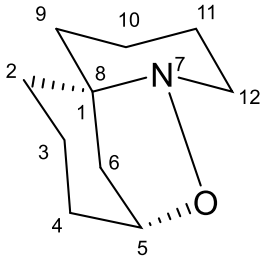

16500878-piperdinol-0802
single pulse decoupled gated NOE
1.1
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
-0.1
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
-20
f1 (ppm)

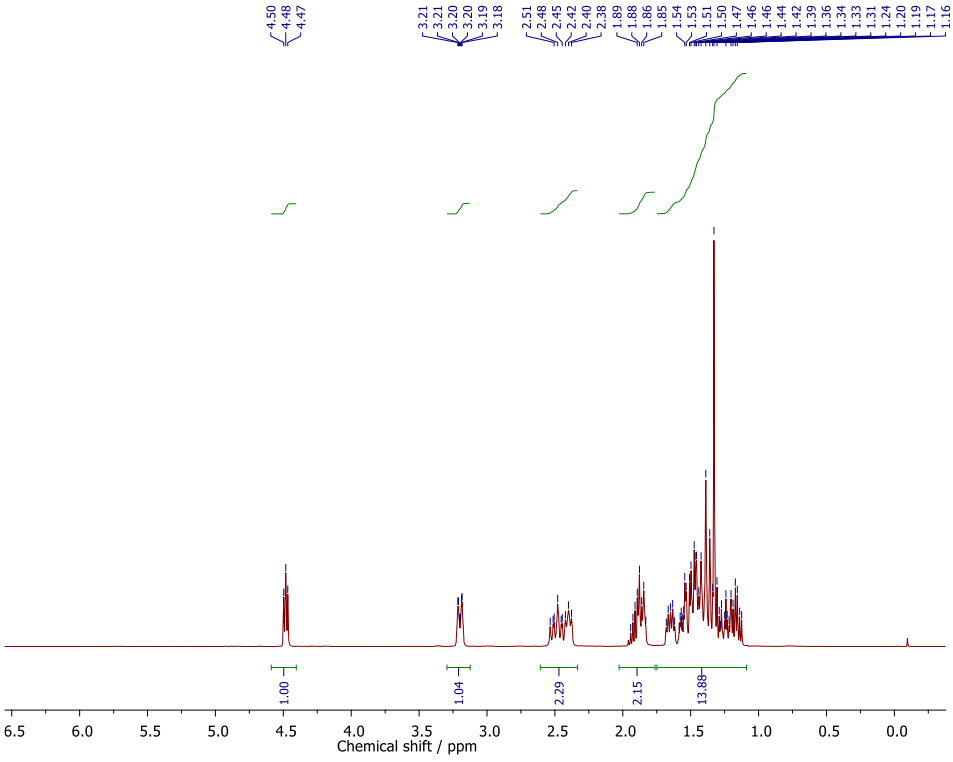
(1S*,5S*)-octahydro-1*H*-cyclopenta[3,4][1,2]oxazolo[6,12-*a*]pyridine (22)



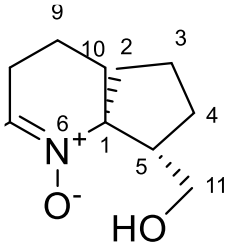


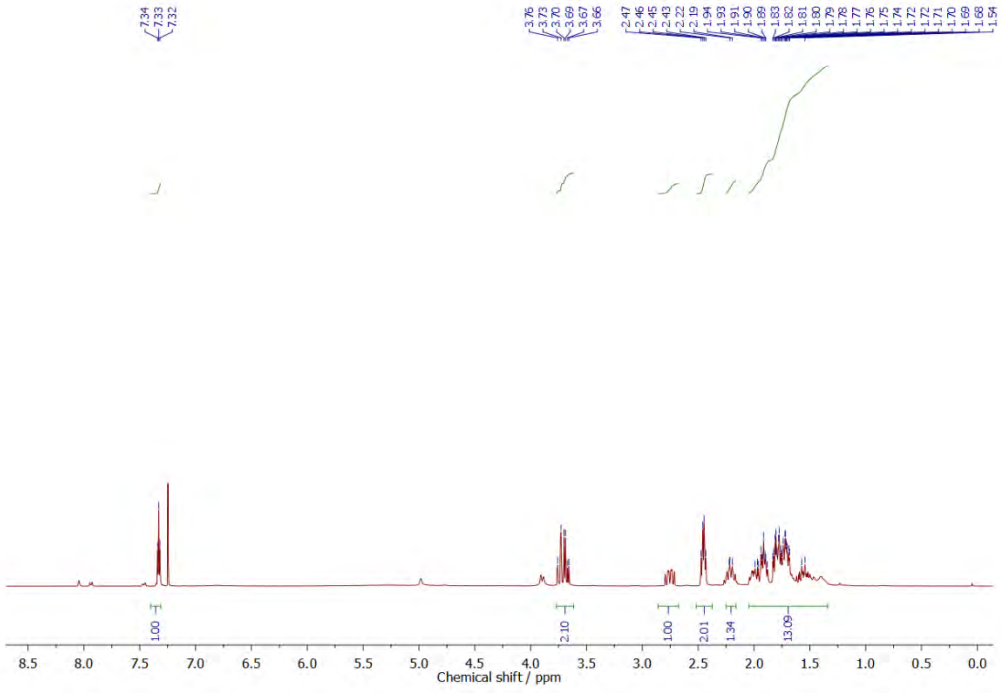
(1S*,5S*)-7-Oxa-6-azatricyclo[6.3.1.0^{1,6}]dodecane (23)



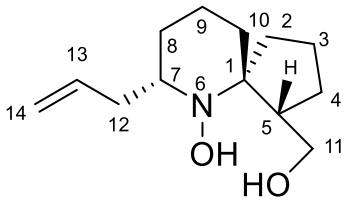


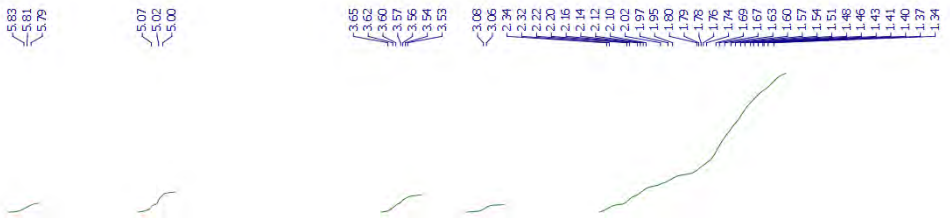
(1S*,5S*)-(6-oxido-6-azaspiro[4.5]dec-6-en-1-yl)methanol (105)

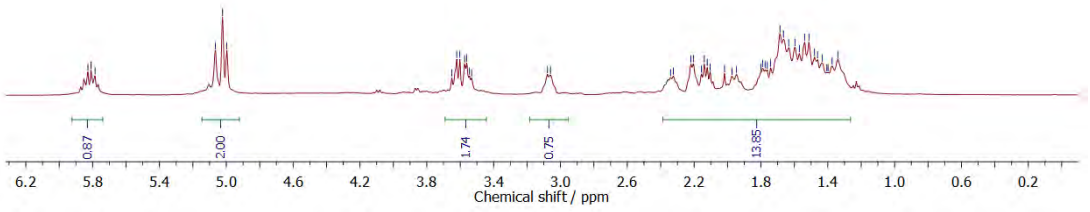


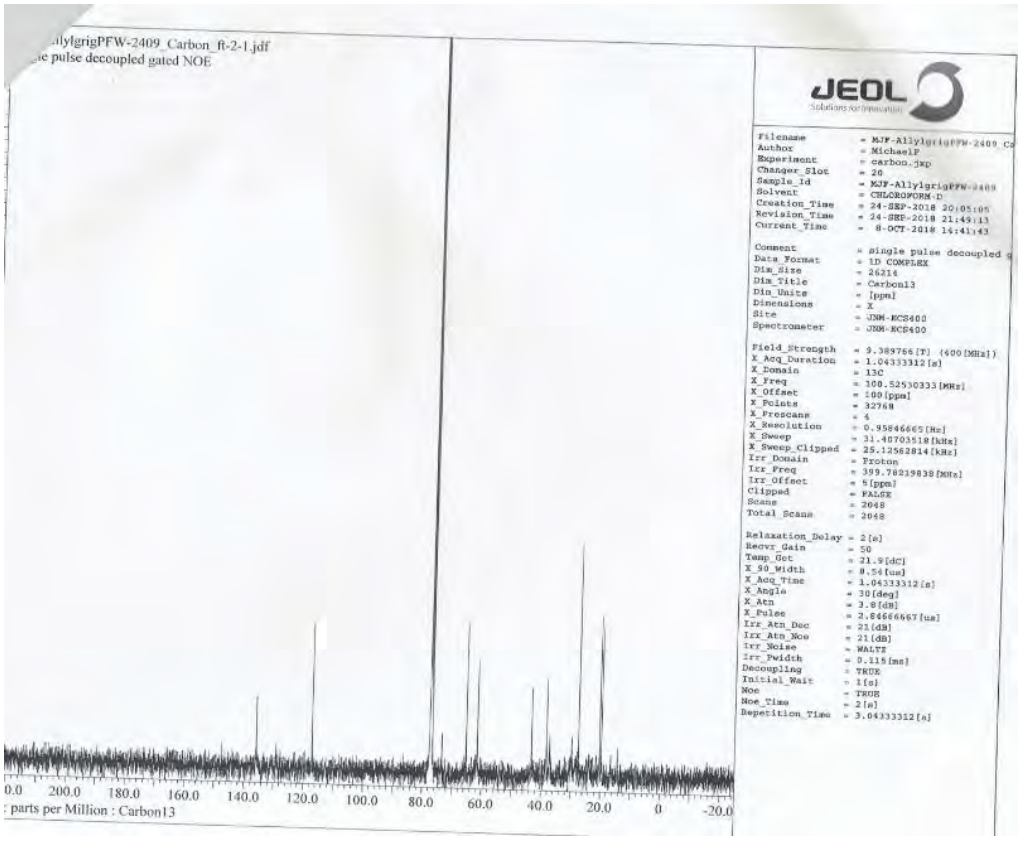


(1S*,5S*,7S*)-11-(hydroxymethyl)-7-allyl-6-azaspiro[4.5]decan-11-ol (163+161)

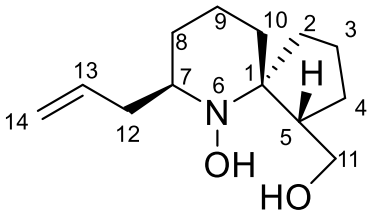


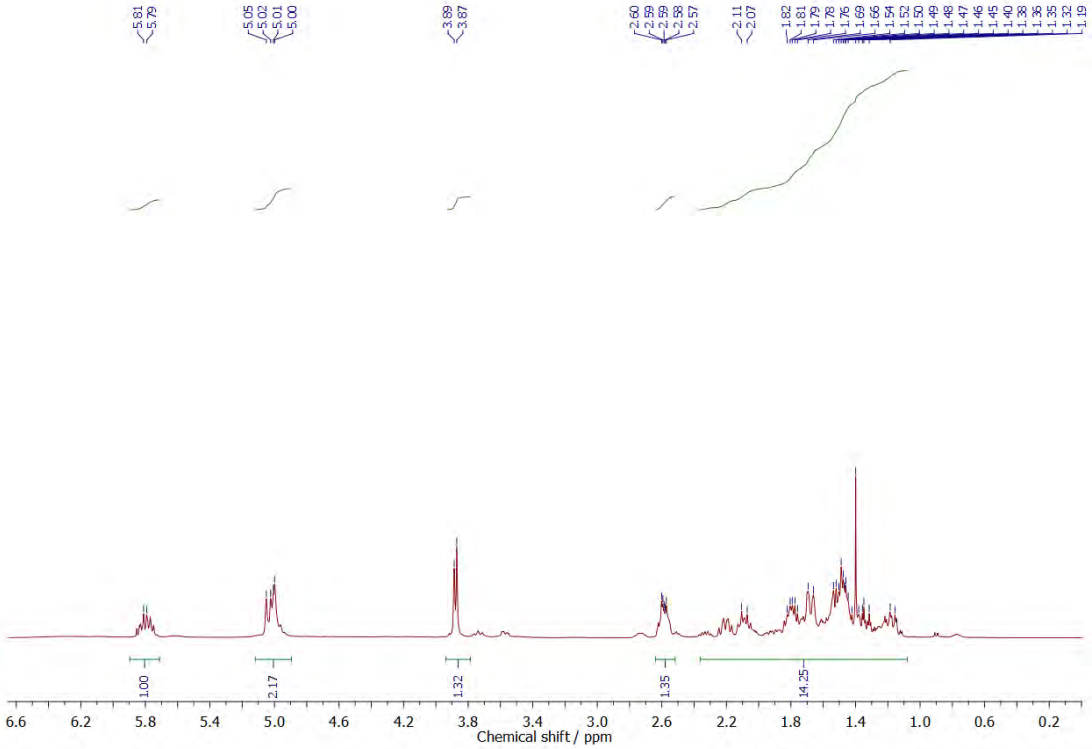


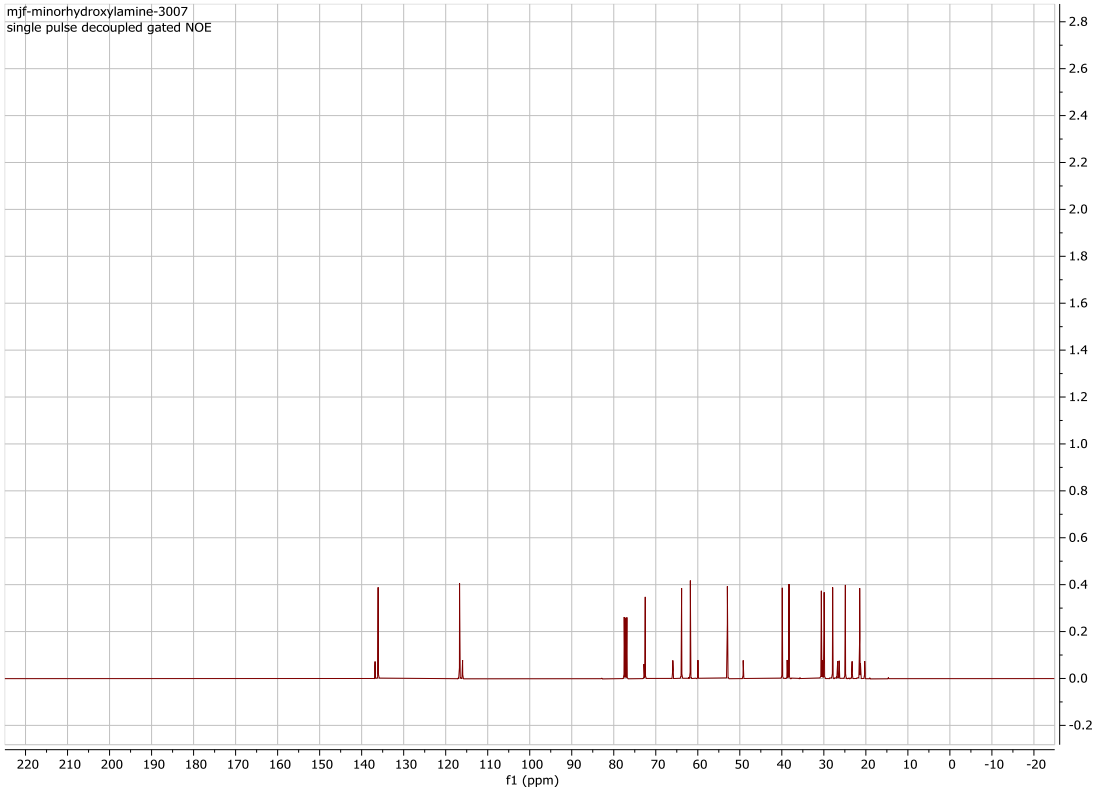



AllylgrigPFW-2409_Carbon_ft-2-1.jdf
se pulse decoupled gated NOE
JEOL
Solutions for Innovation
Filename = MJF-AllylgrigPFW-2409_Ca
Author = MichaelF
Experiment = carbon.jxp
Changer_Slot = 20
Sample_Id = MJF-AllylgrigPFW-2409
Solvent = CHLOROFORM-D
Creation_Time = 24-SEP-2018 20:05:05
Revision_Time = 24-SEP-2018 21:49:13
Current_Time = 8-OCT-2018 16:41:43
Comment = Single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECZ400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 2[s]
Recvr_Gain = 50
Temp_Get = 21.9[dC]
X_90_Width = 8.56[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.0[dB]
X_Pulse = 2.56666667[us]
Irr_Atn_Dec = 21[dB]
Irr_Atn_Noe = 21[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE
Initial_Wait = 1[s]
Noe = TRUE
Noe_Time = 2[s]
Repetition_Time = 3.04333312[s]
0.0 200.0 180.0 160.0 140.0 120.0 100.0 80.0 60.0 40.0 20.0 0 -20.0
parts per Million : Carbon13

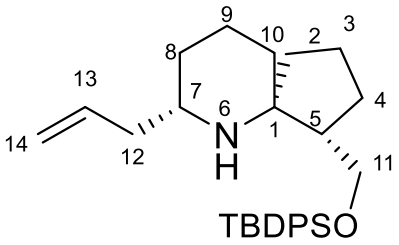
(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (161)

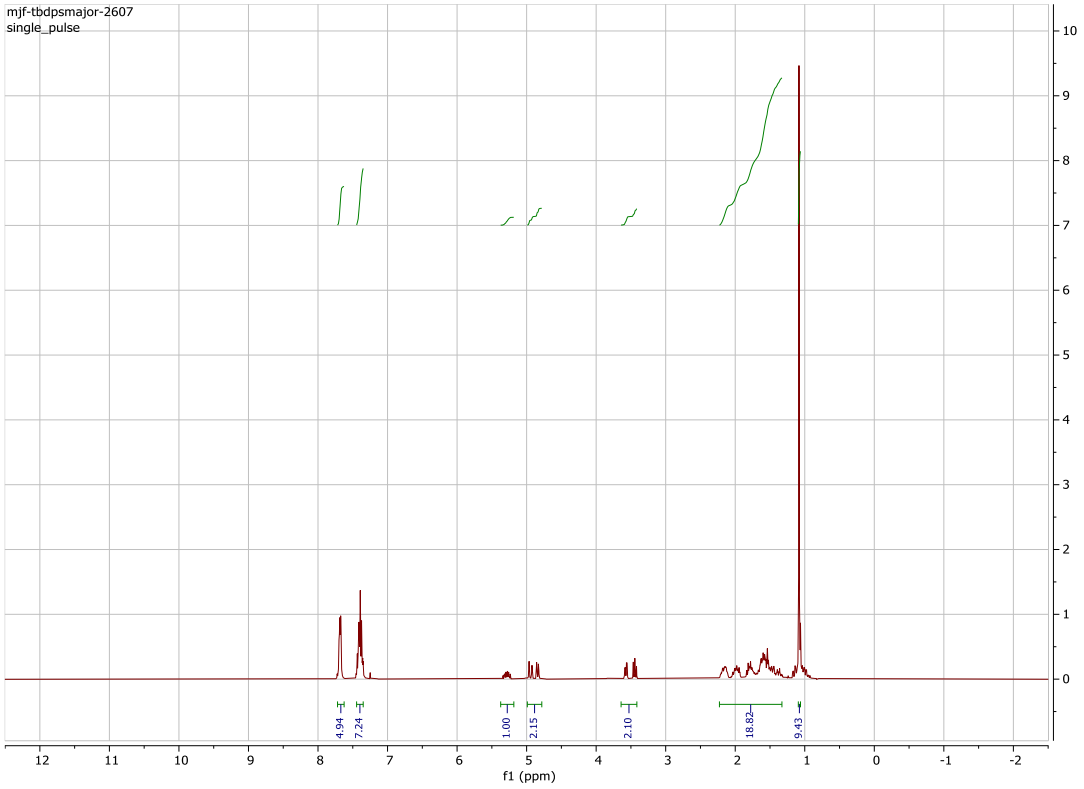


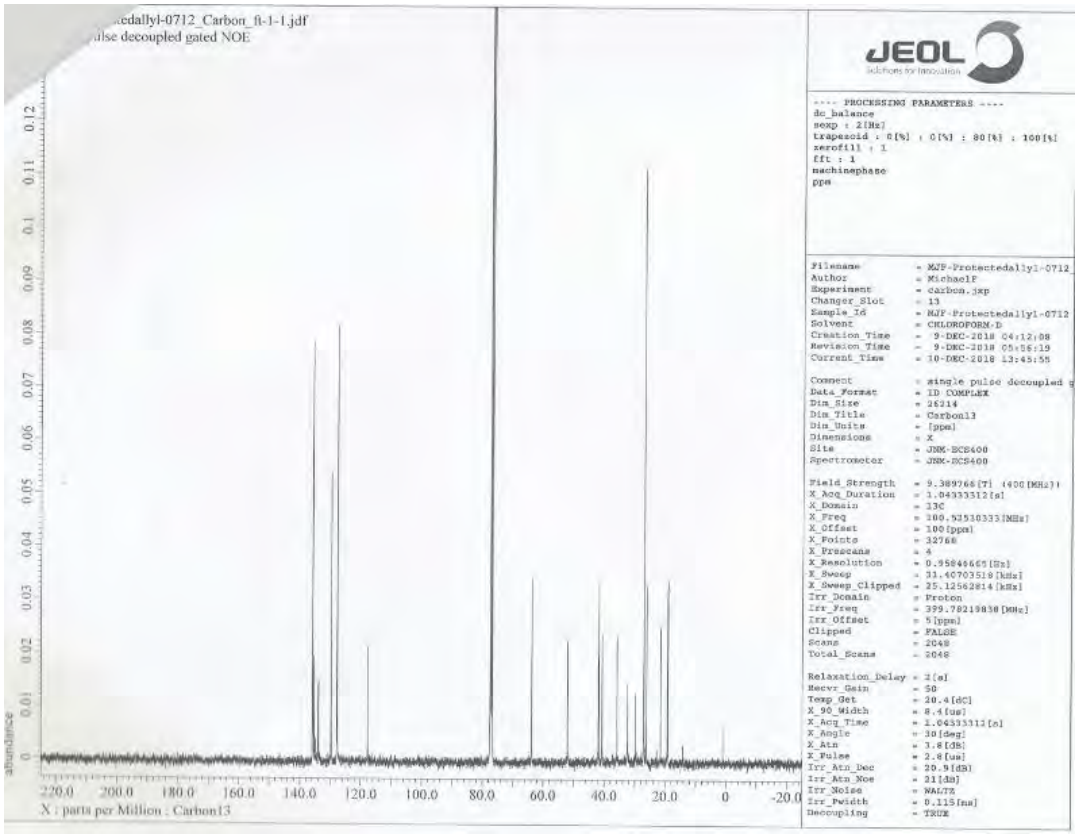



mjf-minorhydroxylamine-3007
single pulse decoupled gated NOE
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (165)






cedallyl-0712_Carbon_ft-1-1.jdf
ulse decoupled gated NOE
abundance
0.12
0.11
0.1
0.09
0.08
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0
X : parts per Million : Carbon13
JEOL
solutions for innovation
---- PROCESSING PARAMETERS ----
dc_balance
nexp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = MJF-Protectedallyl-0712
Author = MichaelF
Experiment = carbon.jxp
Changer_Slot = 13
Sample_Id = MJF-Protectedallyl-0712
Solvent = CHLOROFORM-D
Creation_Time = 9-DEC-2018 04:12:08
Revision_Time = 9-DEC-2018 05:56:19
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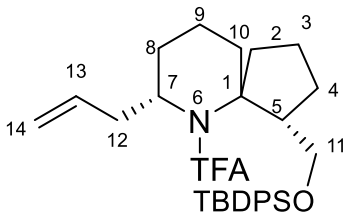
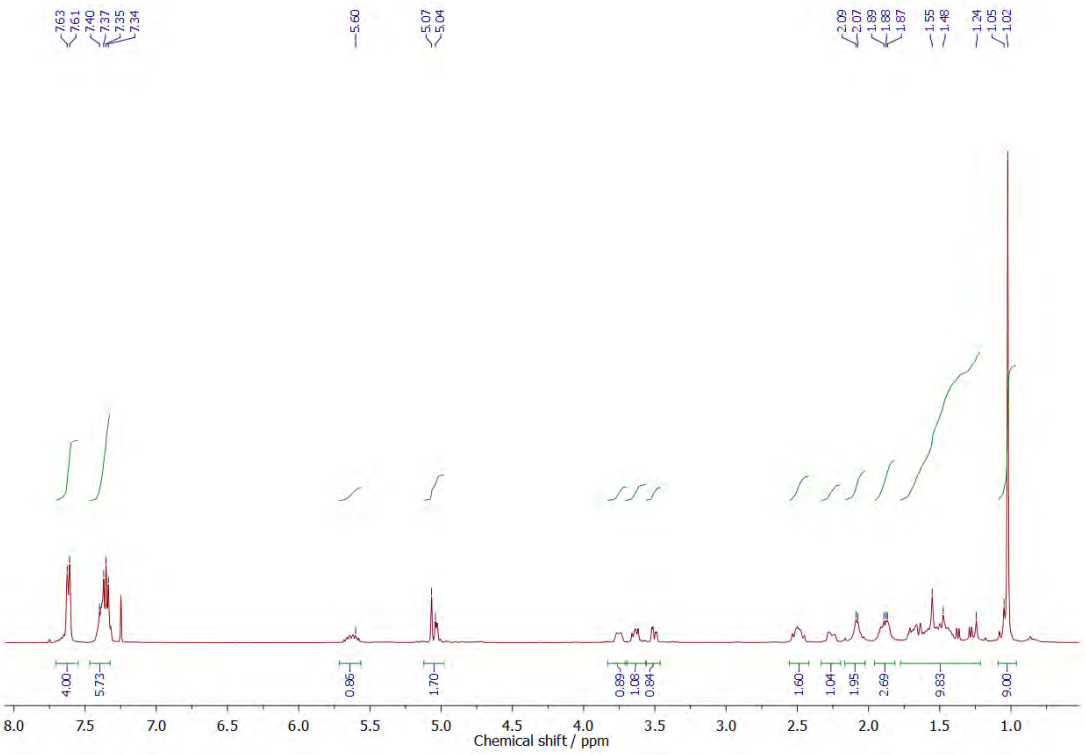
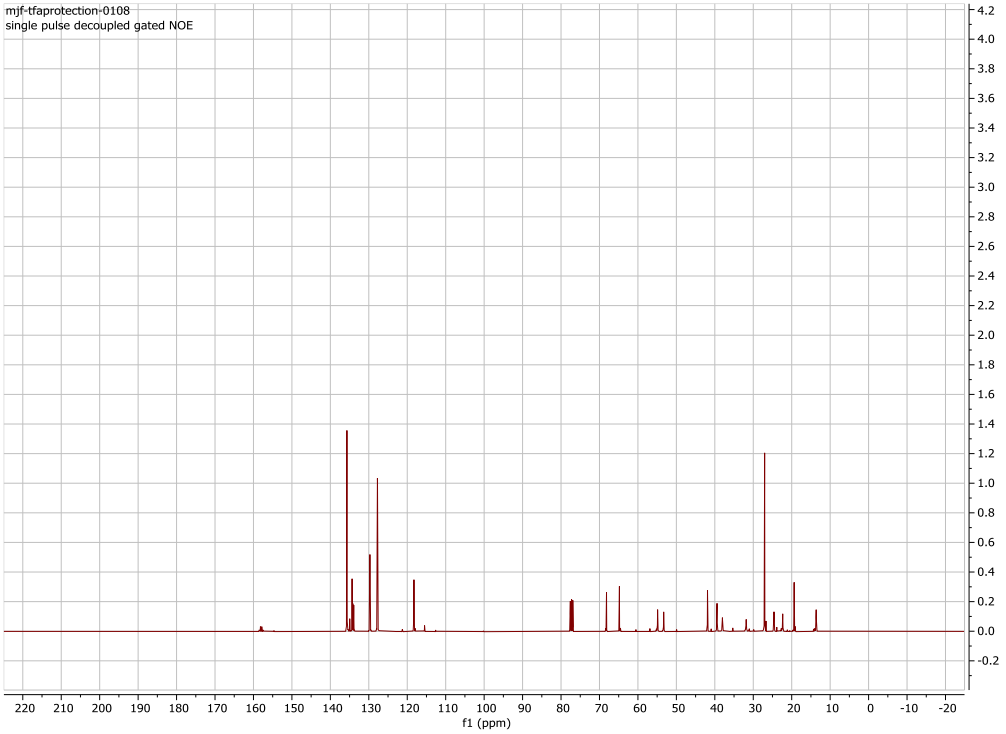
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 2[s]
Recvr_Gain = 50
Temp_Get = 20.4[dC]
X_90_Width = 8.4[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 1.8[dB]
X_Pulse = 2.8[us]
Irr_Atn_Dec = 20.9[dB]
Irr_Atn_Noe = 21[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE

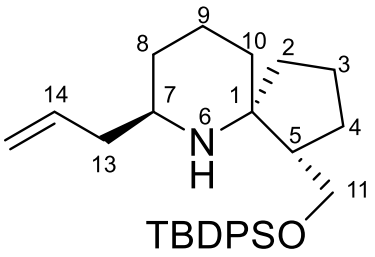
(1*S**,5*S**,7*R**)-11-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane-2,2,2-trifluoroactyl (166)

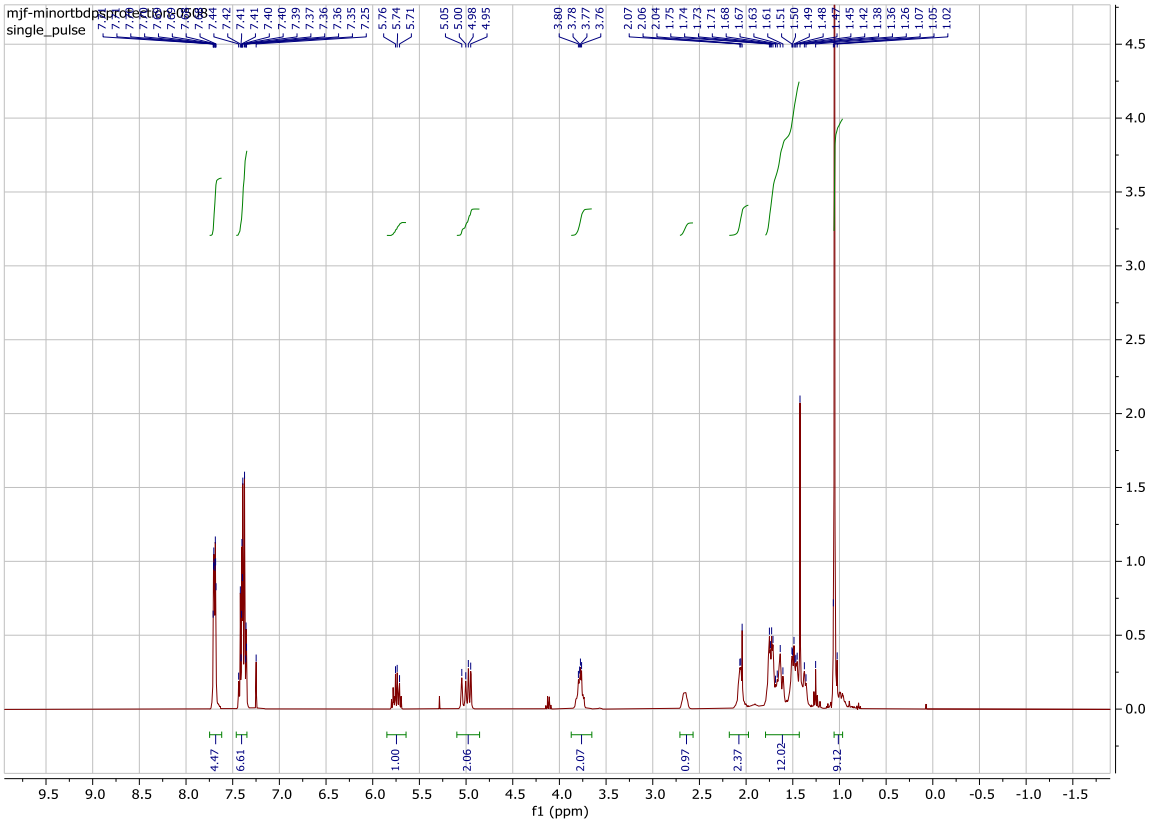


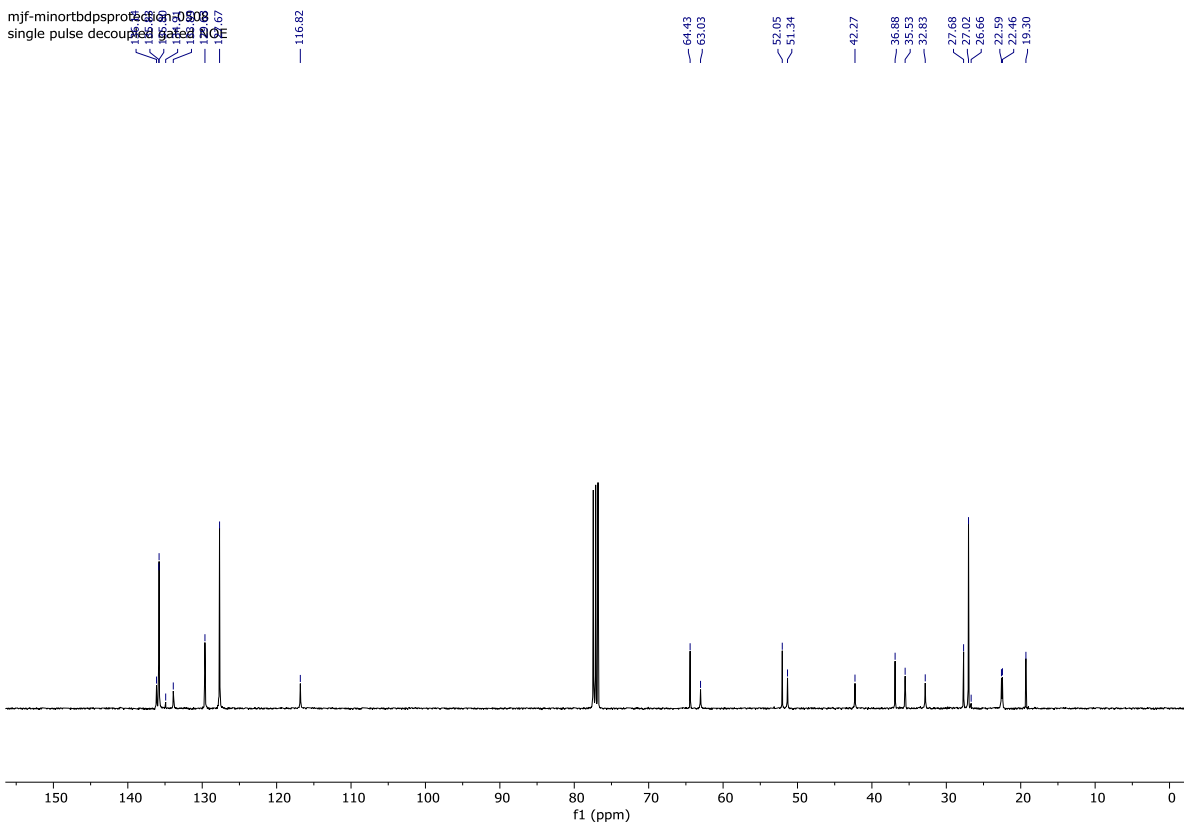



mjf-tfaprotection-0108
single pulse decoupled gated NOE
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

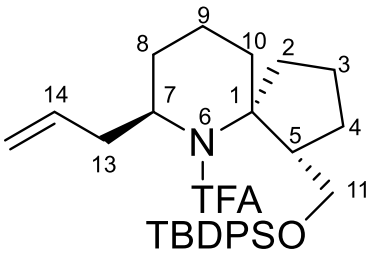
(1*S**,5*S**,7*R**)-5-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane (169)

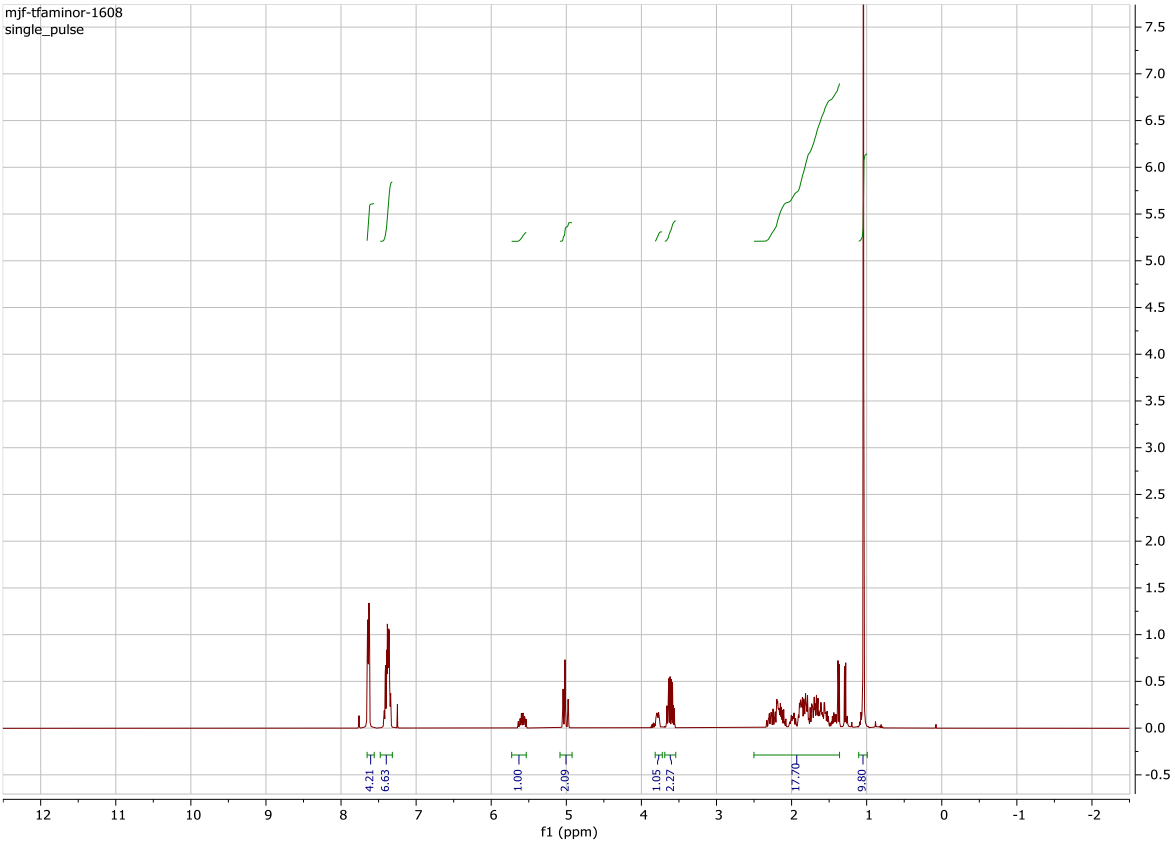


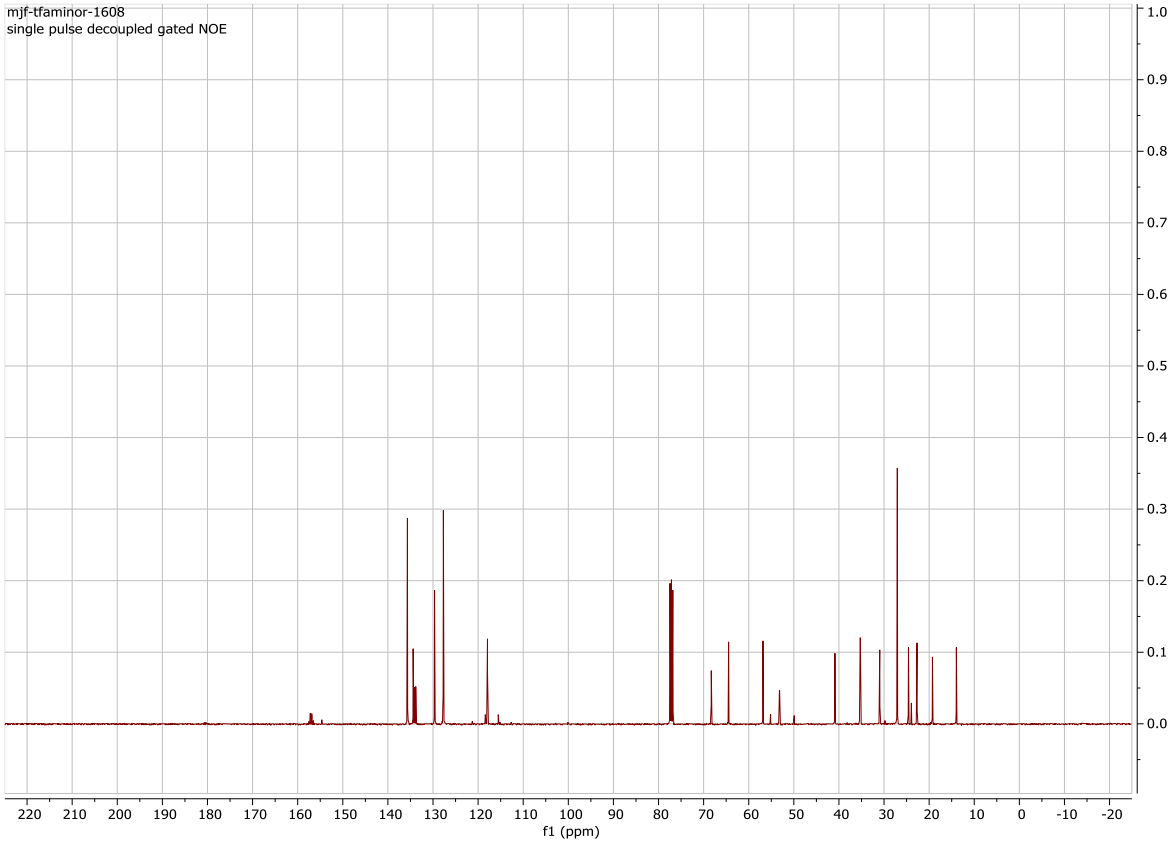



mjf-minortbdpsprotec13cpd-0508
single pulse decoupled gated NOE
116.82
64.43
63.03
52.05
51.34
42.27
36.88
35.53
32.83
27.68
27.02
26.66
22.59
22.46
19.30
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

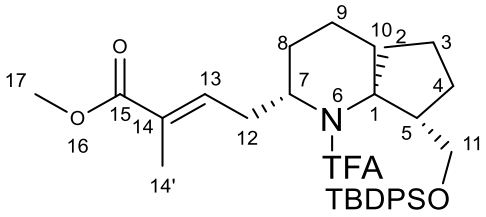
(1*S**,5*S**,7*R**)-5-(tert-butyldiphenylsilyloxymethyl)-7-allyl-6-azaspiro[4.5]decane-2,2,2-trifluoroactyl (170)

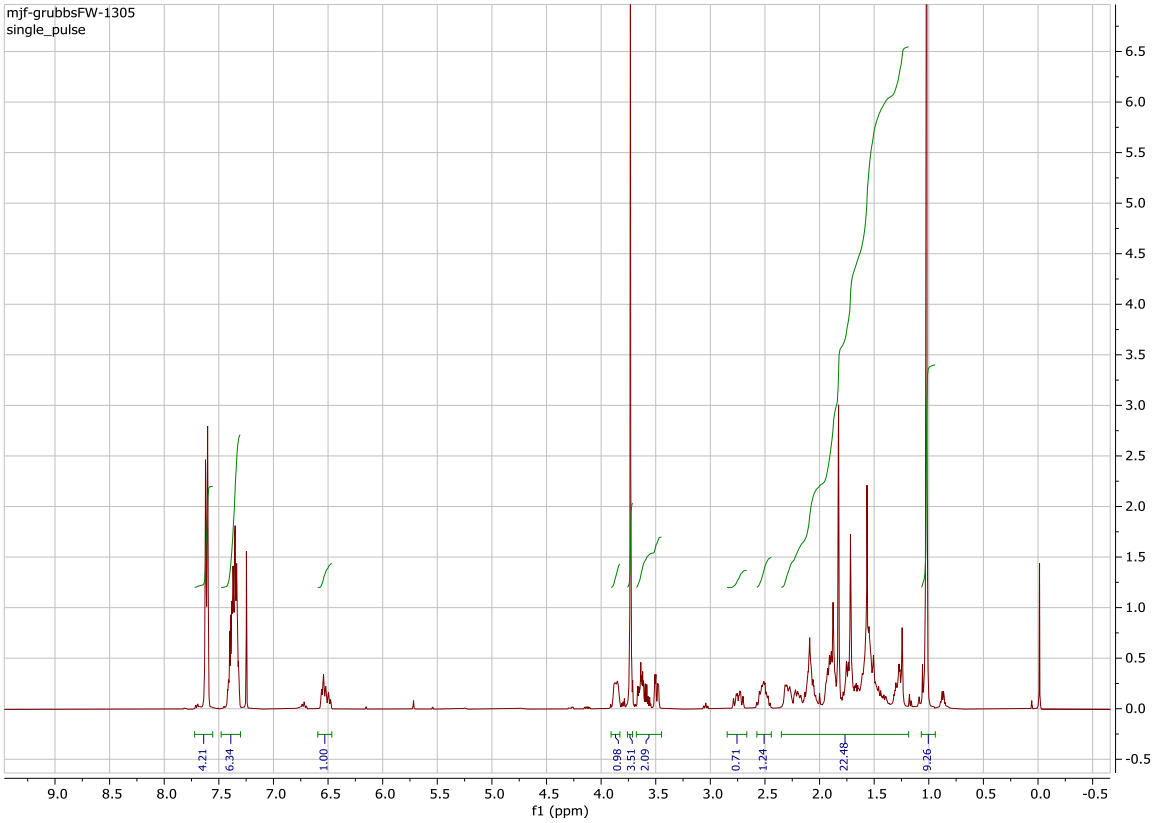


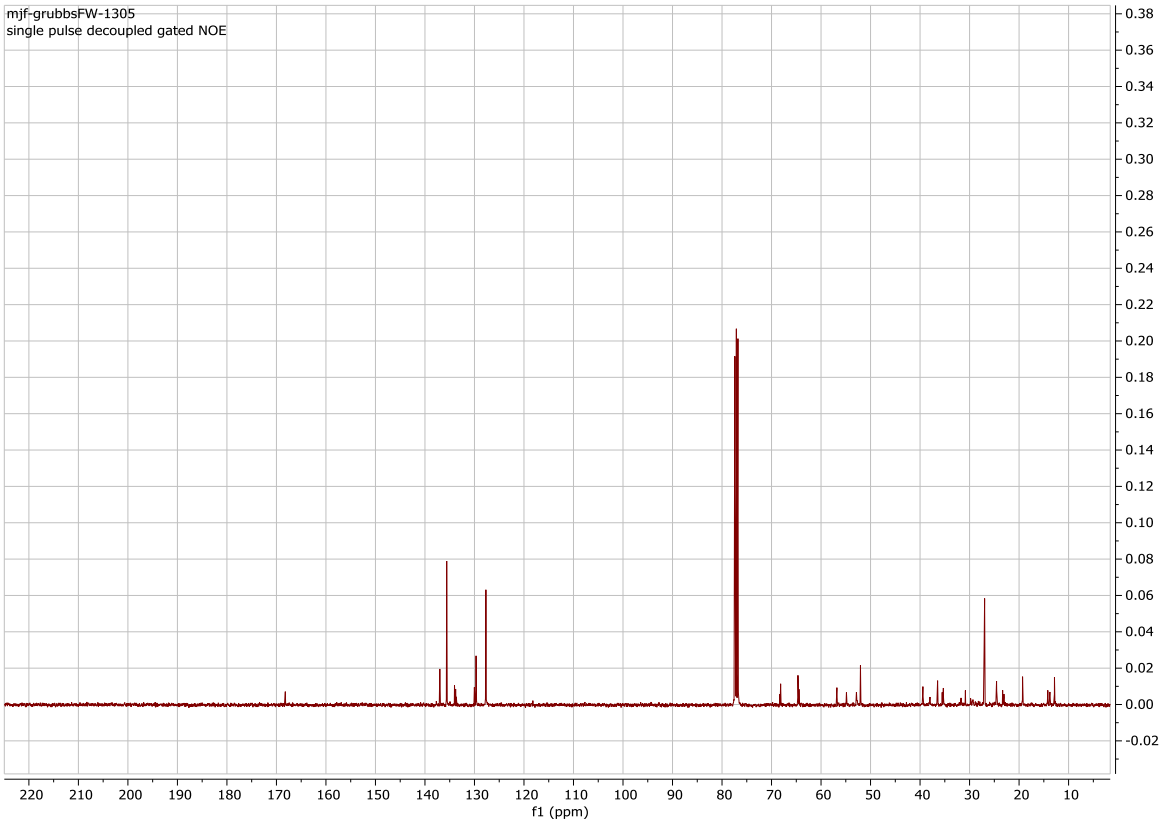



mjf-tfaminor-1608
single pulse decoupled gated NOE
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

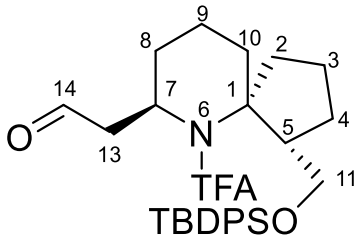
(E)-methyl-14’-((1S,5S,7S)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-13-methylbut-14-enoate (175)

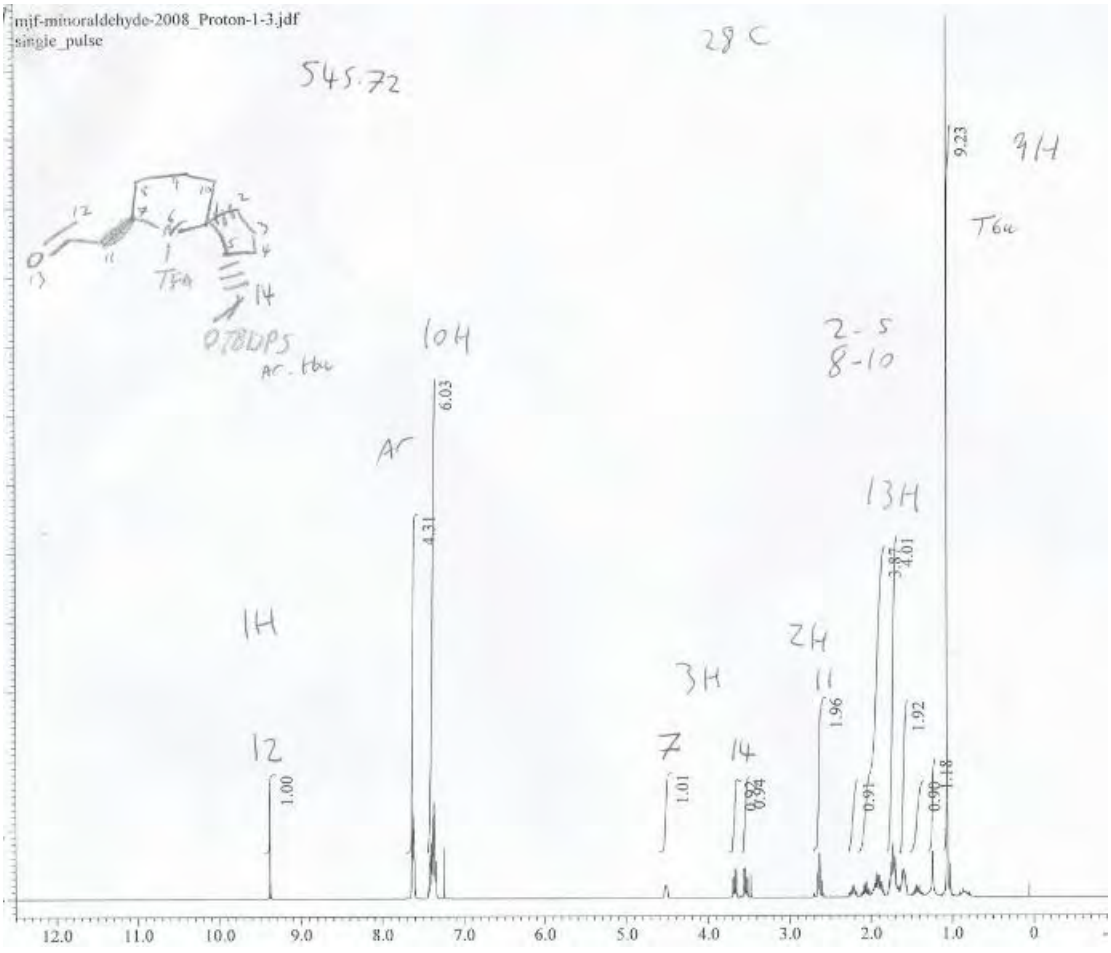


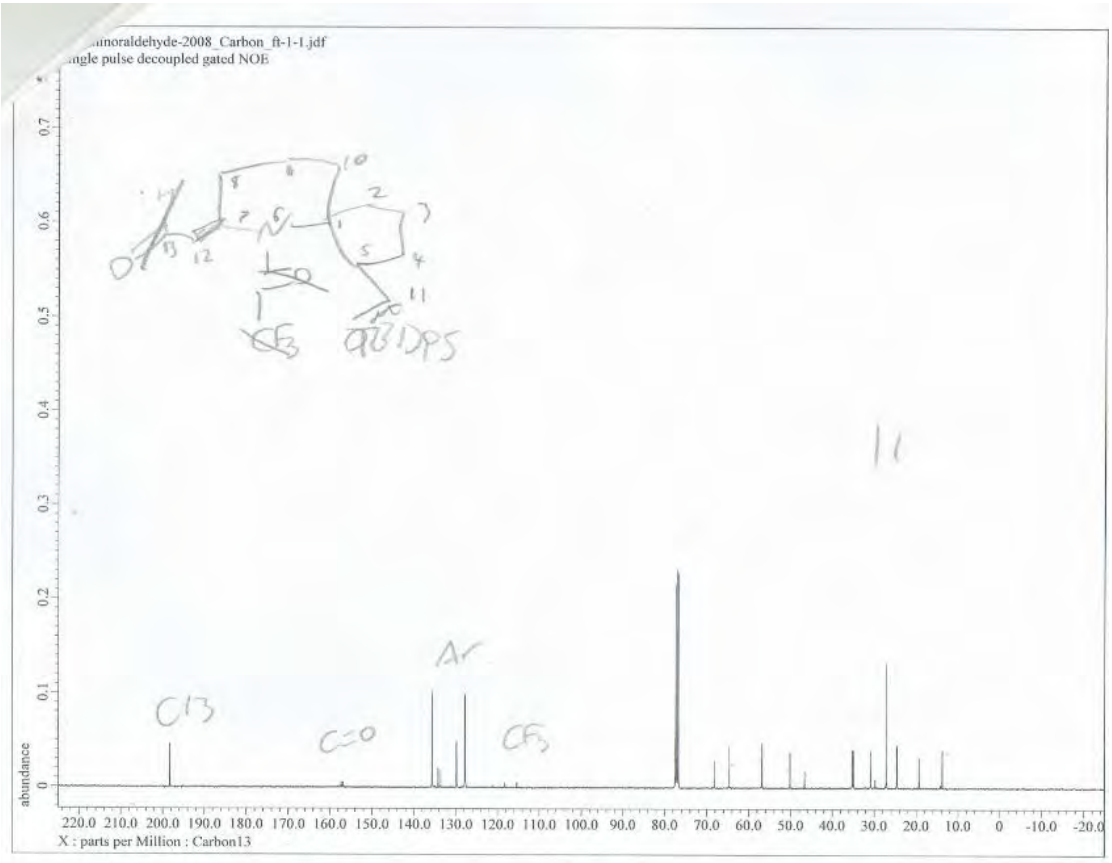



mjf-grubbsFW-1305
single pulse decoupled gated NOE
0.38
0.36
0.34
0.32
0.30
0.28
0.26
0.24
0.22
0.20
0.18
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
-0.02
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

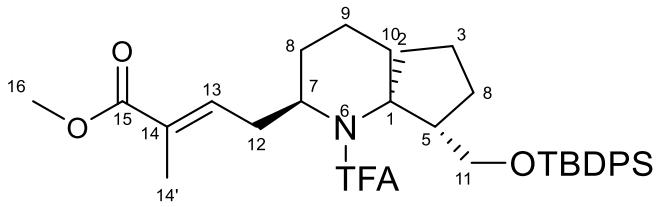
2-((1S,5S,7R)-1-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)acetaldehyde (176)

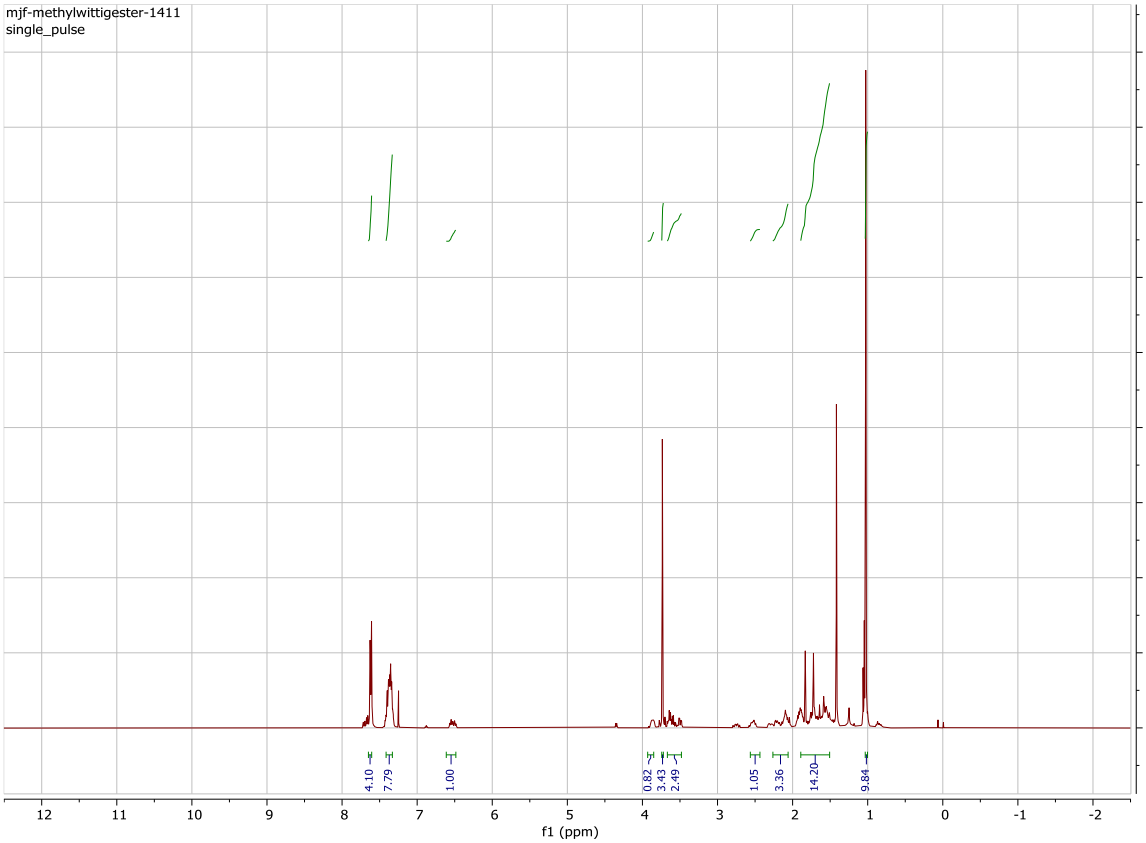


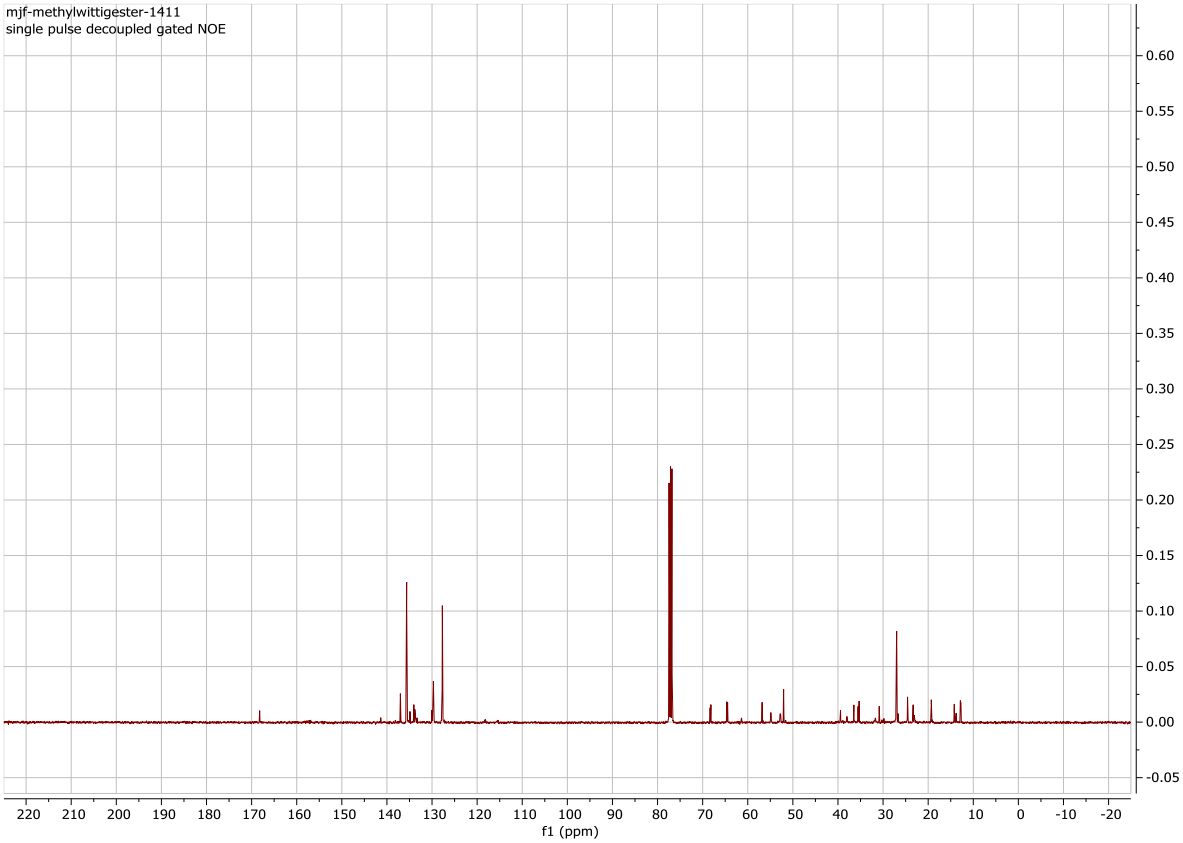



...inoraldehyde-2008_Carbon_ft-1-1.jdf
...ngle pulse decoupled gated NOE
abundance
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
C13
C=O
Ar
CF3
220.0 210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0 -10.0 -20.0
X : parts per Million : Carbon13

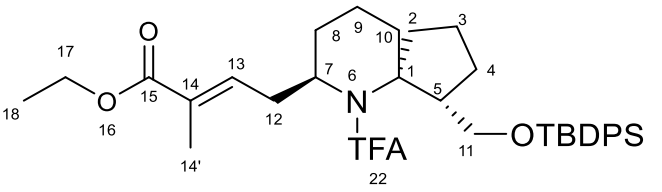
(E)-methyl-14-((1S,5S,7R)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-2-methylbut-2-enoate (177)

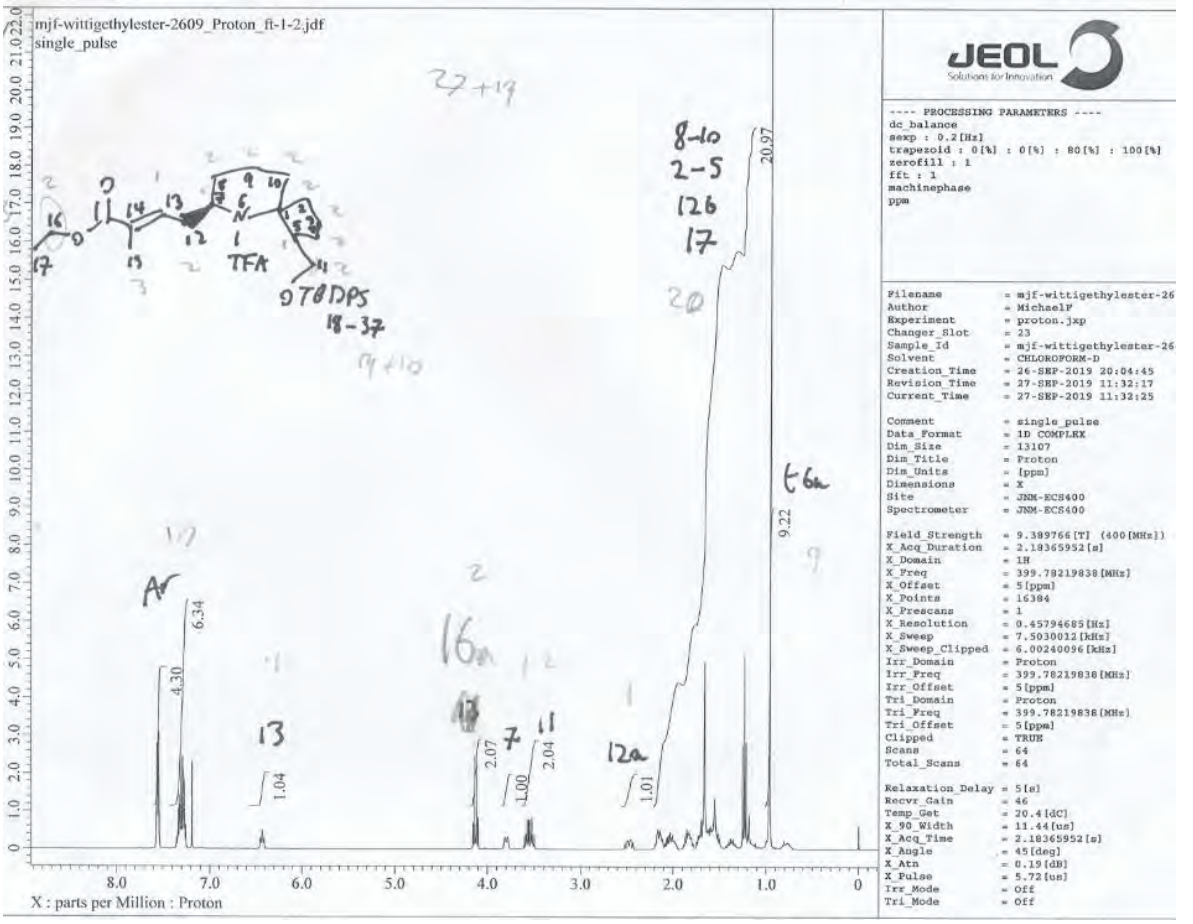


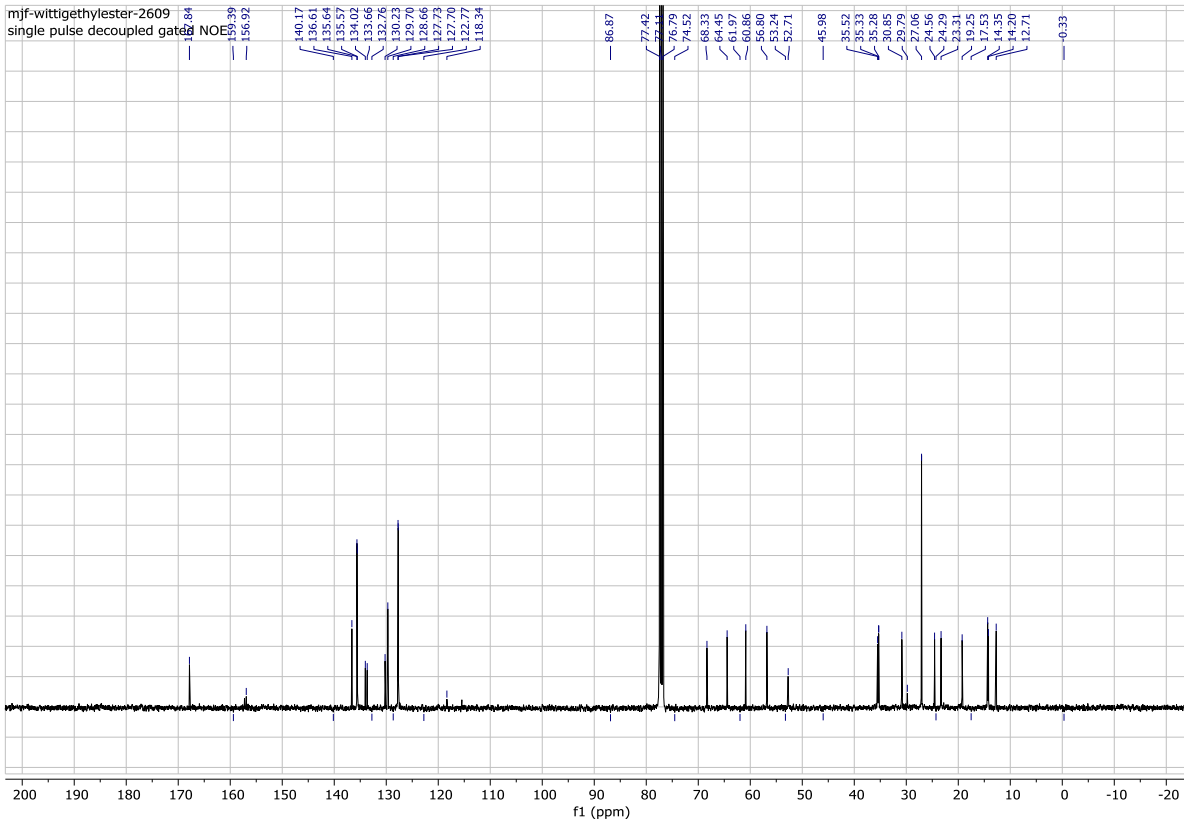



mjf-methylwittigester-1411
single pulse decoupled gated NOE
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

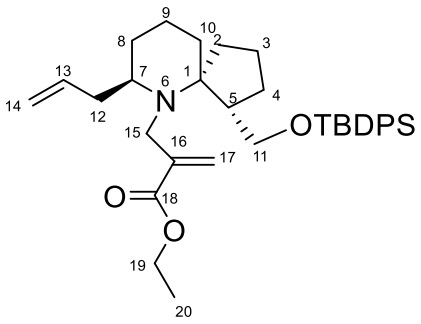
(E)-ethyl-5-((1S,5S,7R)-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-7-yl)-2-methylbut-2-enoate(178)

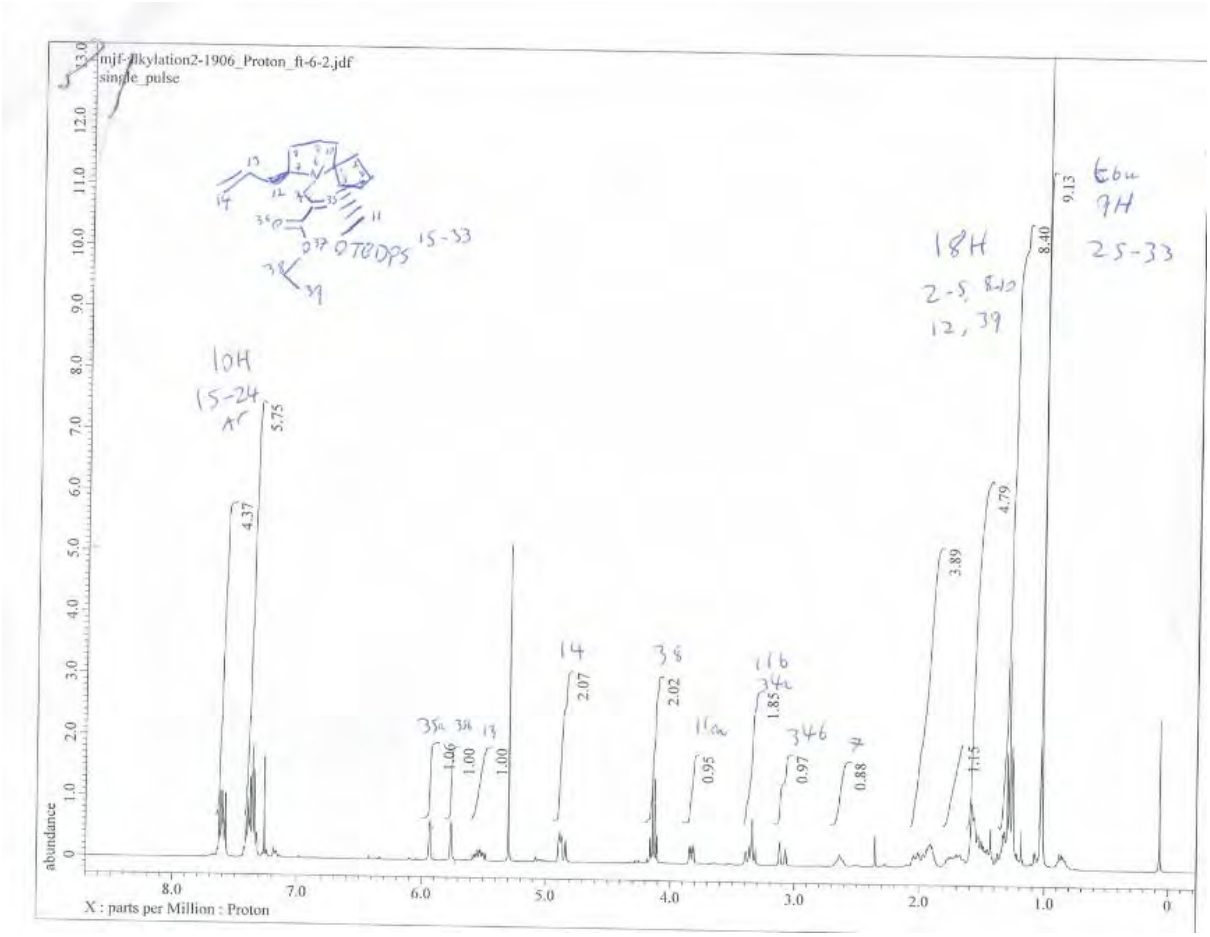


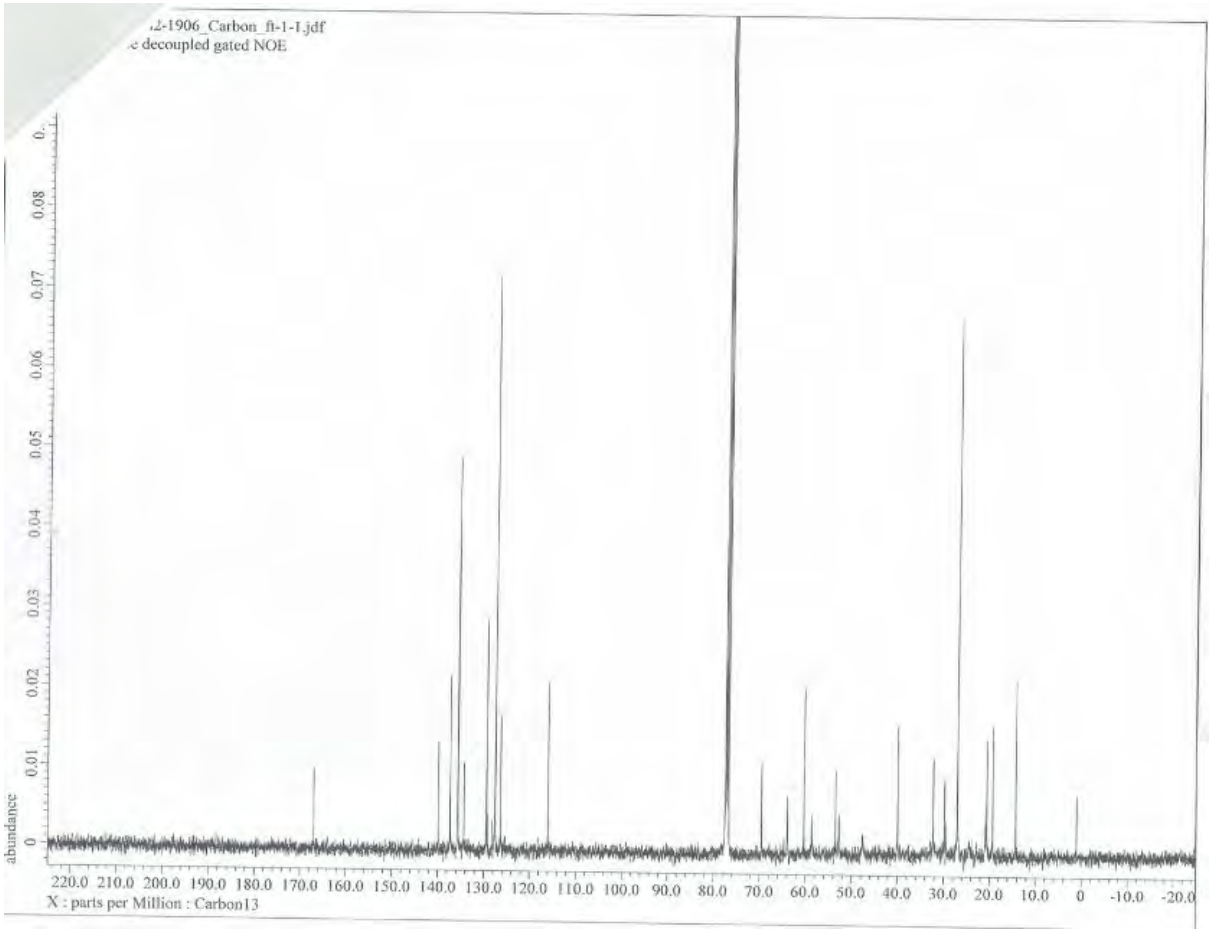



mjf-wittigethylester-2609
single pulse decoupled gated NOE
172.84
159.39
156.92
140.17
136.61
135.64
135.57
134.02
133.66
132.76
130.23
129.70
128.66
127.73
127.70
122.77
118.34
86.87
77.42
77.11
76.79
74.52
68.33
64.45
61.97
60.86
55.80
53.24
52.71
45.98
35.52
35.33
35.28
30.85
29.79
27.06
24.56
24.29
23.31
19.25
17.53
14.35
14.20
12.71
0.33
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

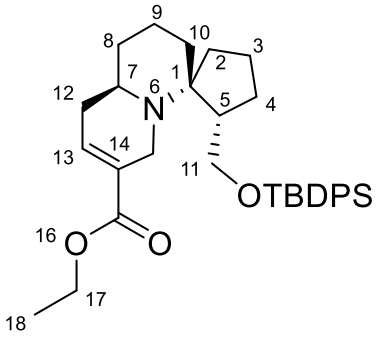
ethyl-2-(((1S,5S,7R)-7-allyl-11-(((tert-butyldiphenylsilyl)oxy)methyl)-6-azaspiro[4.5]decan-6-yl)methyl)acrylate (180)

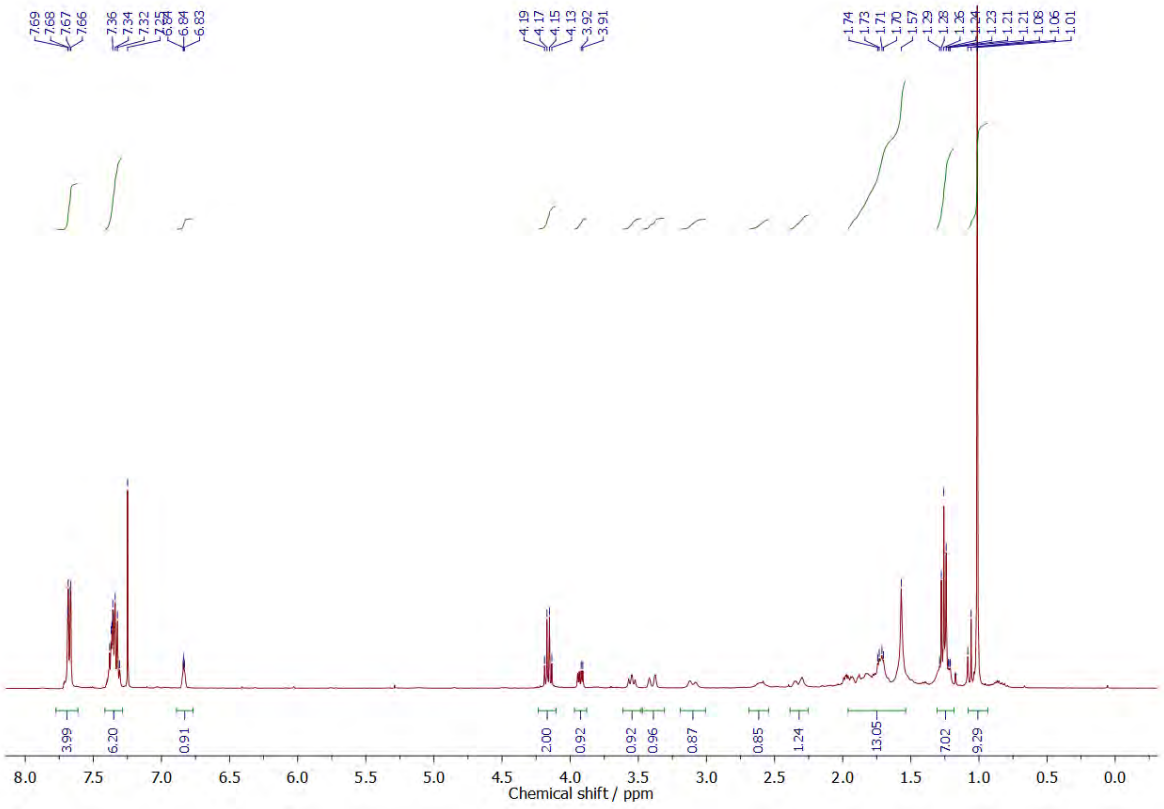


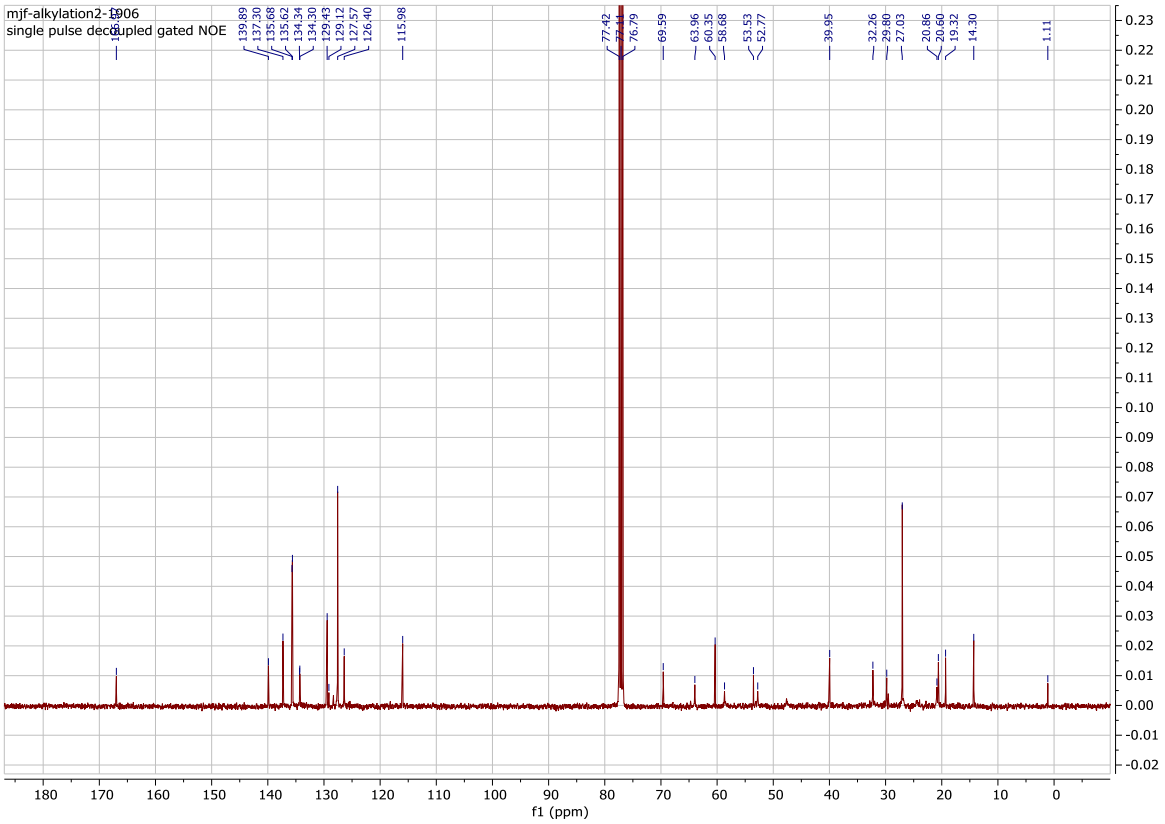



2-1906_Carbon_ft-1-1.jdf
e decoupled gated NOE
abundance
0
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
220.0 210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0 -10.0 -20.0
X : parts per Million : Carbon13

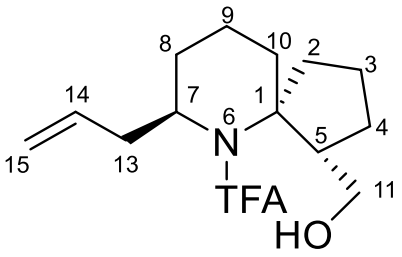
(1S,5S,7R)-ethyl-11-(((tert-butyldiphenylsilyl)oxy)methyl)-1',2',3',6',9',9a'-hexahydrospiro[cyclopentane-1,4'-quinolizine]-7'-carboxylate (181)

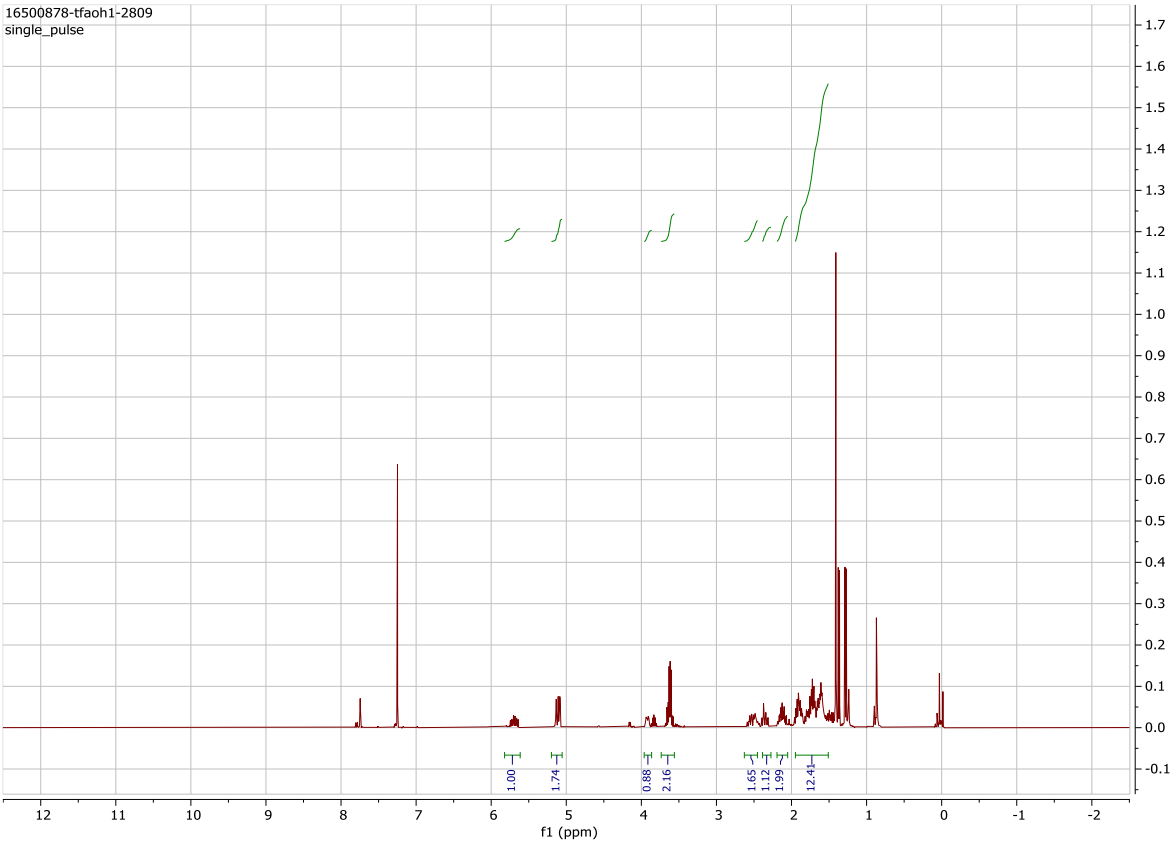


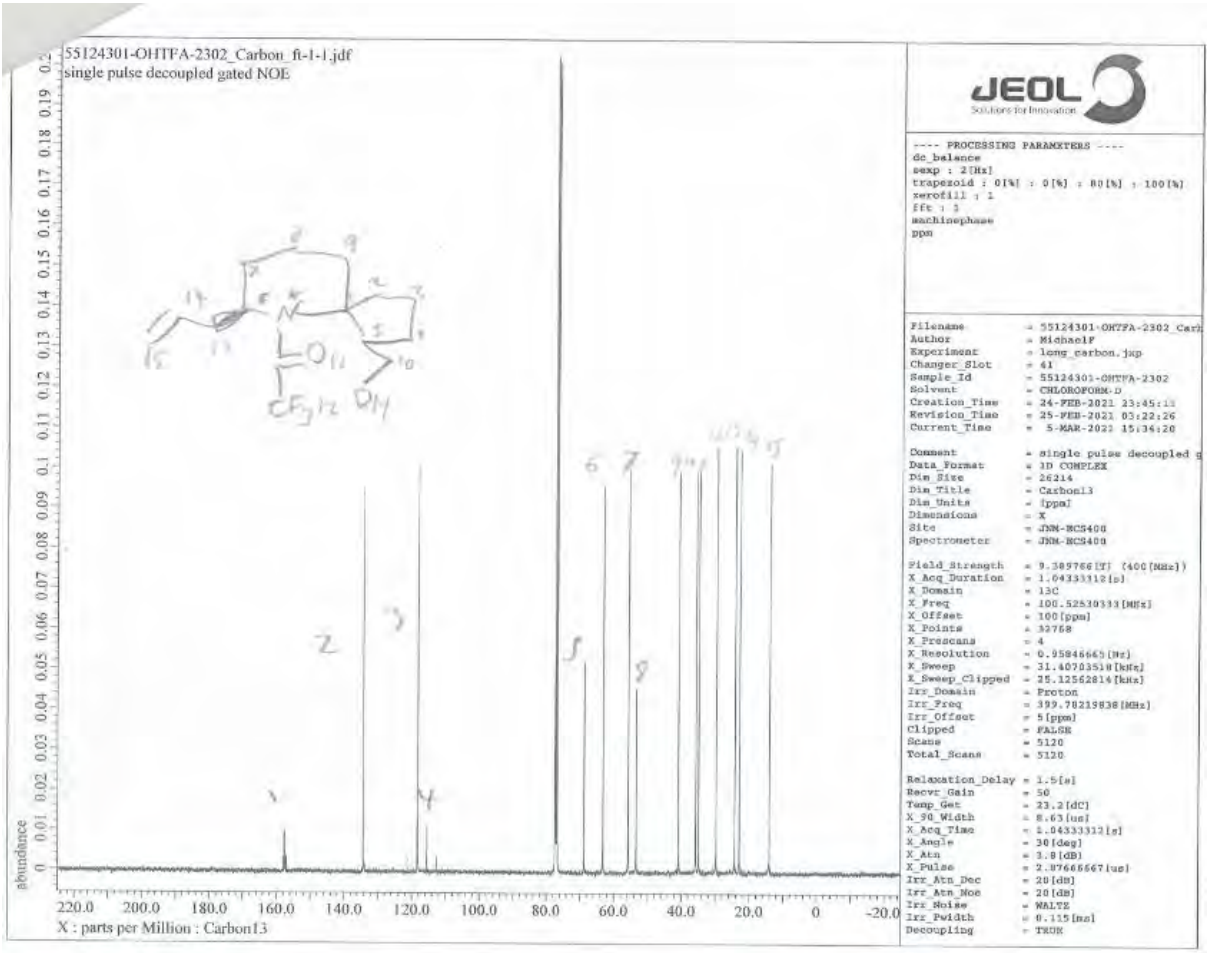



mjf-alkylation2-1906
single pulse decoupled gated NOE
168.18
139.89
137.30
135.68
135.62
134.34
134.30
129.43
129.12
127.57
126.40
115.98
77.42
77.41
76.79
69.59
63.96
60.35
58.68
53.53
52.77
39.95
32.26
29.80
27.03
20.86
20.60
19.32
14.30
1.11
f1 (ppm)

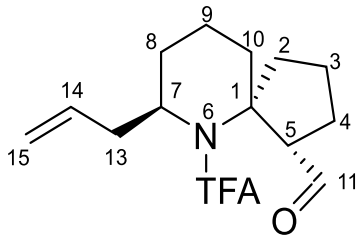
1-((1S,5S,7R)-7-allyl-1-(hydroxymethyl)-6-azaspiro[4.5]decan-6-yl)-2,2,2-trifluoroethanone (187)

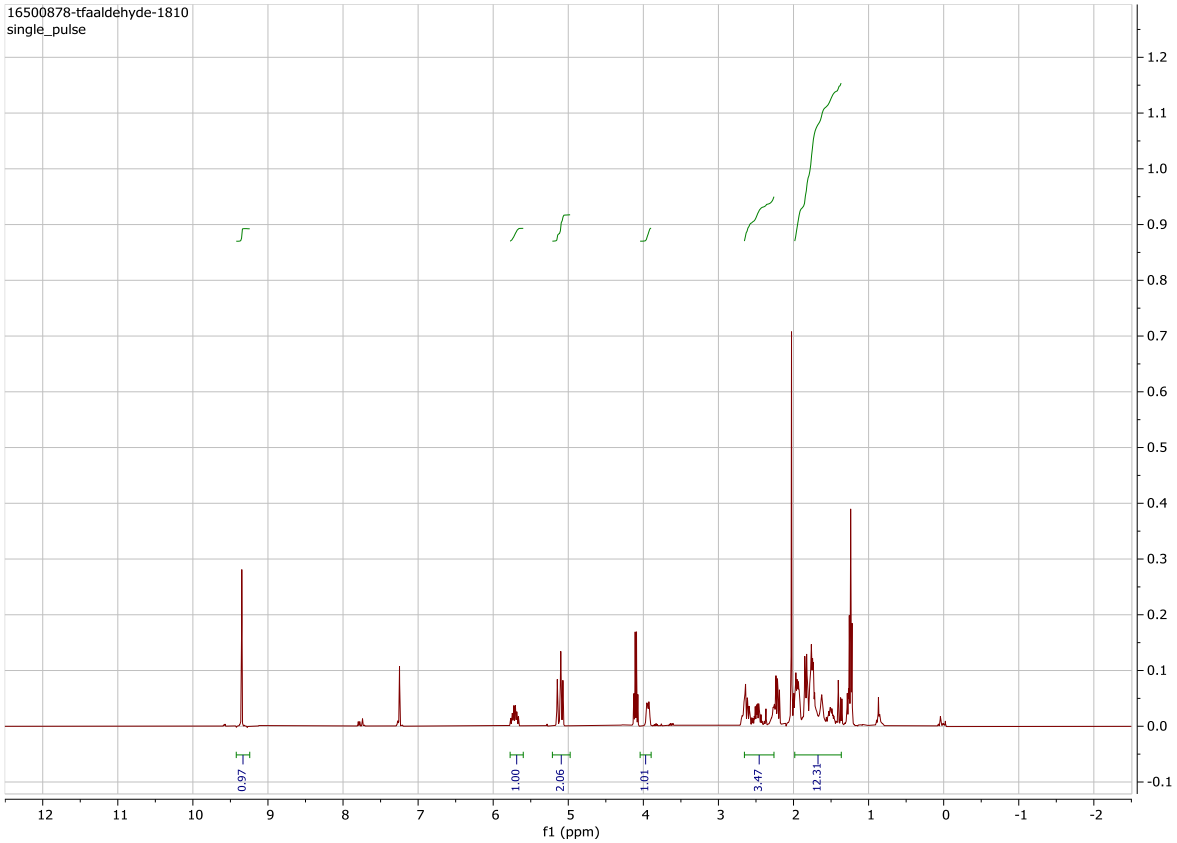


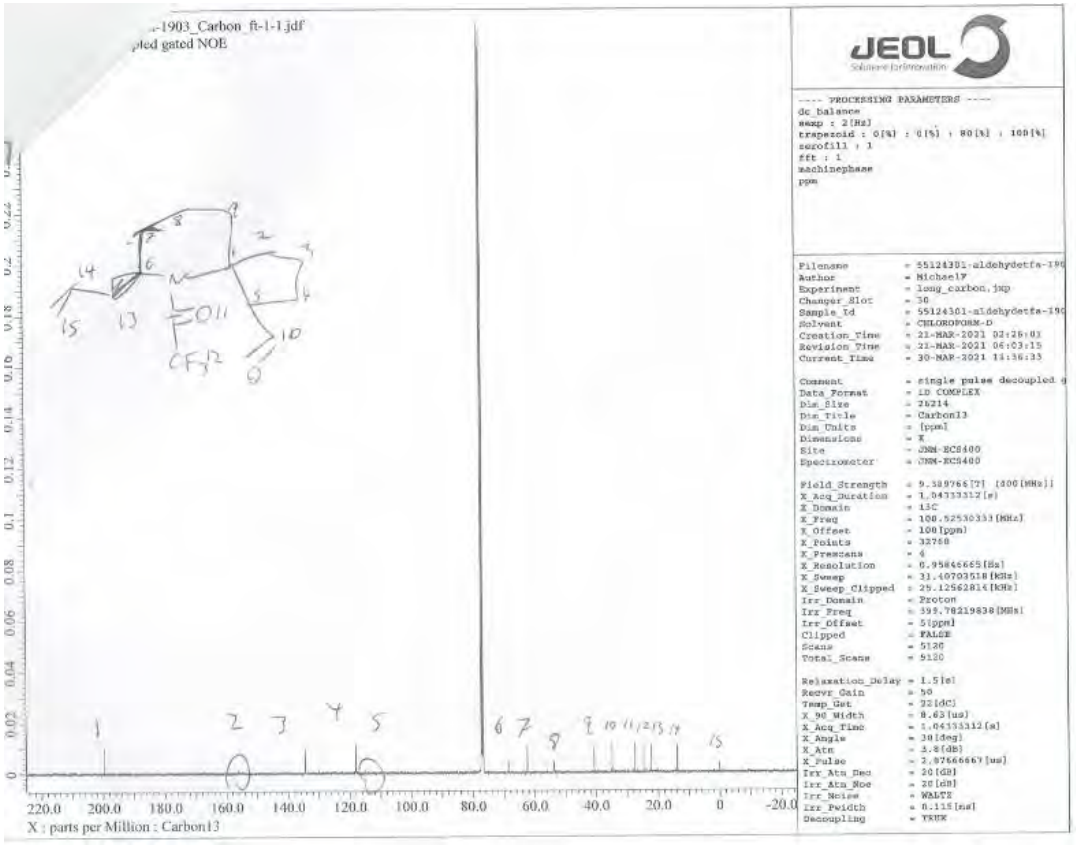



55124301-OHTFA-2302_Carbon_ft-1-1.jdf
single pulse decoupled gated NOE
abundance
X : parts per Million : Carbon13
---- PROCESSING PARAMETERS ----
dc_balance
sexp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-OHTFA-2302_Carb
Author = MichaelF
Experiment = 1dmg_carbon.jxp
Changer_Slot = 41
Sample_Id = 55124301-OHTFA-2302
Solvent = CHLOROFORM-D
Creation_Time = 24-FEB-2021 23:45:11
Revision_Time = 25-FEB-2021 03:22:26
Current_Time = 5-MAR-2021 15:36:20
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 5120
Total_Scans = 5120
Relaxation_Delay = 1.5[s]
Recvr_Gain = 50
Temp_Get = 23.2[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.0[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE

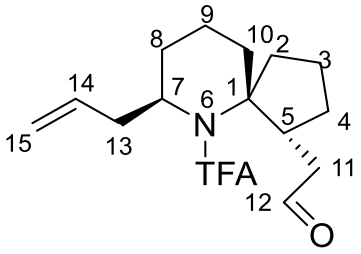
(1S,5S,7R)-7-allyl-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decane-1-carbaldehyde (188)

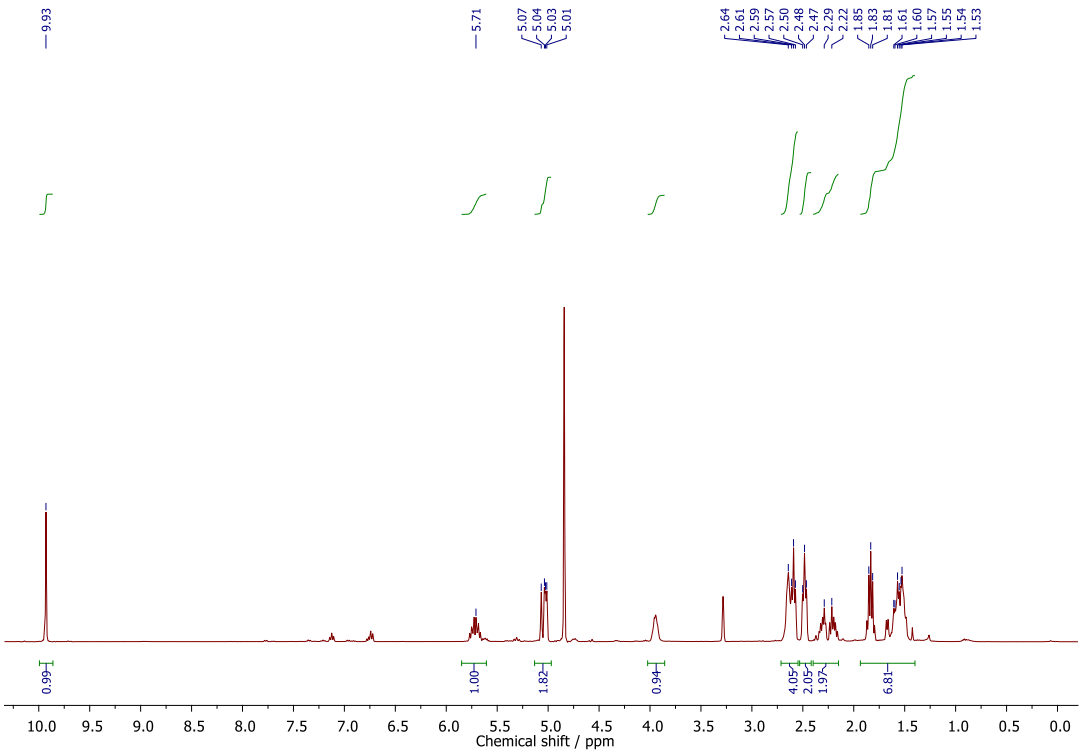


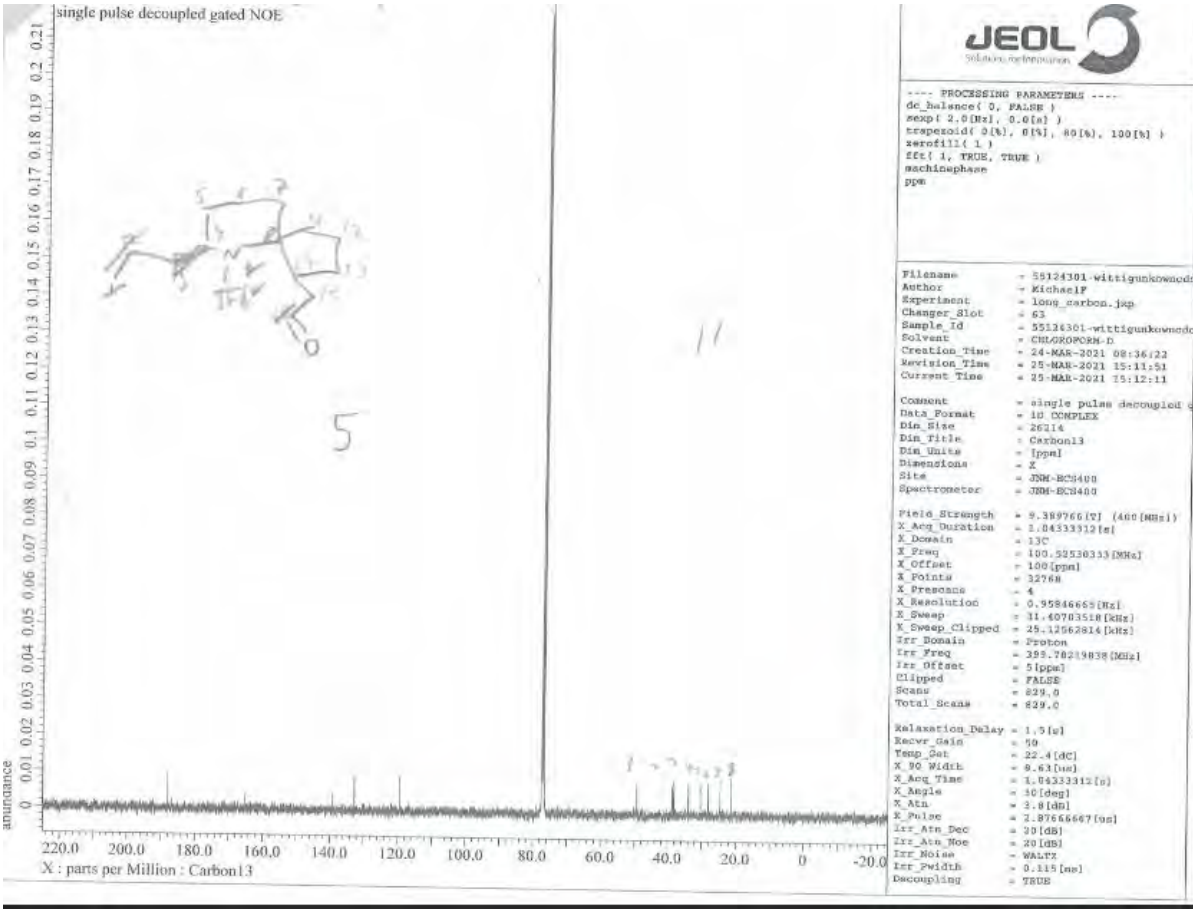



-1903_Carbon_ft-1-1.jdf
pled gated NOE
JEOL
Solutions for Innovation
---- PROCESSING PARAMETERS ----
dc_balance
exmp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-aldehydetfa-190
Author = MichaelF
Experiment = long_carbon.jxp
Changer_Slot = 30
Sample_Id = 55124301-aldehydetfa-190
Solvent = CHLOROFORM-D
Creation_Time = 21-MAR-2021 02:26:03
Revision_Time = 21-MAR-2021 06:03:15
Current_Time = 30-MAR-2021 14:36:33
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 5120
Total_Scans = 5120
Relaxation_Delay = 1.5[s]
Recvr_Gain = 50
Temp_Get = 22[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.8[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 8.115[ms]
Decoupling = YNNN
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15
220.0 200.0 180.0 160.0 140.0 120.0 100.0 80.0 60.0 40.0 20.0 0 -20.0
X : parts per Million : Carbon13

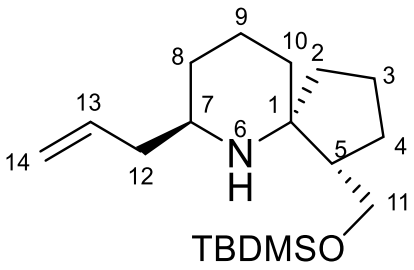
2-((1R,5S,7R)-7-allyl-6-(2,2,2-trifluoroacetyl)-6-azaspiro[4.5]decan-1-yl)acetaldehyde (189)

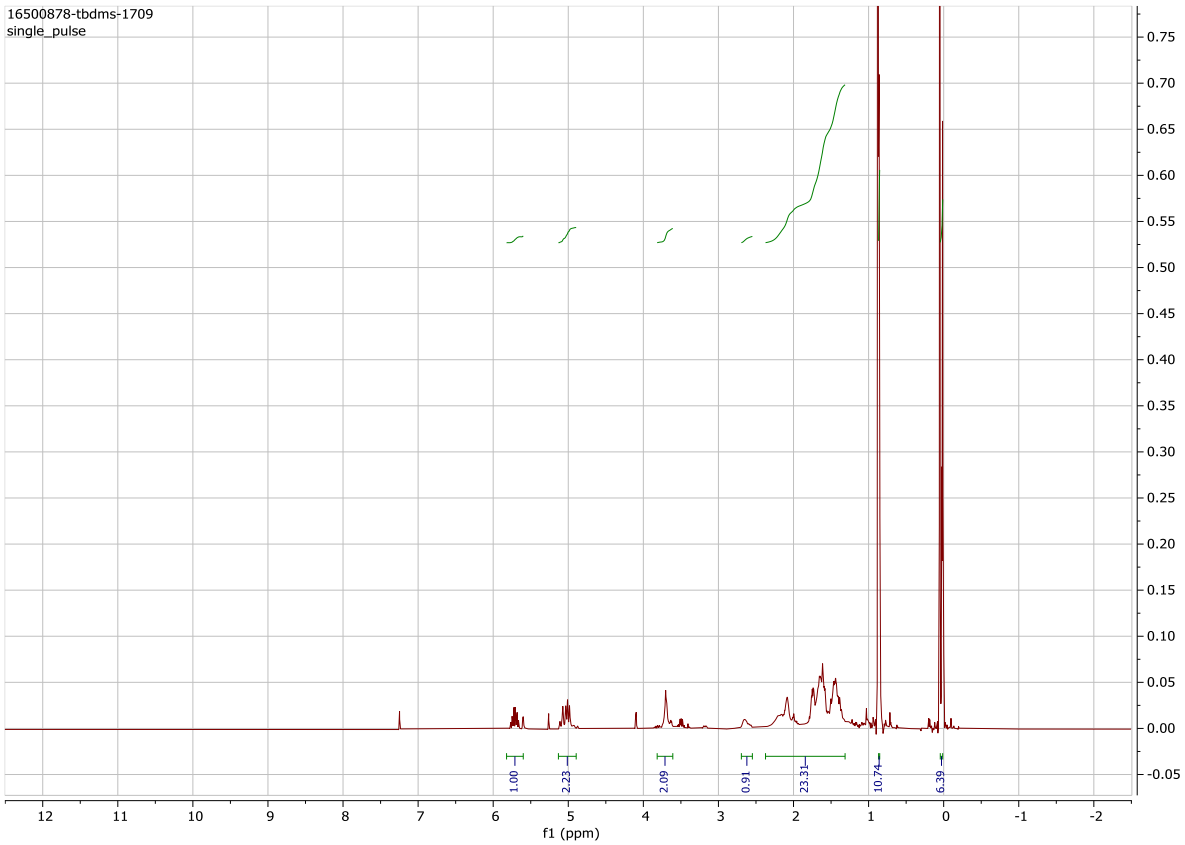


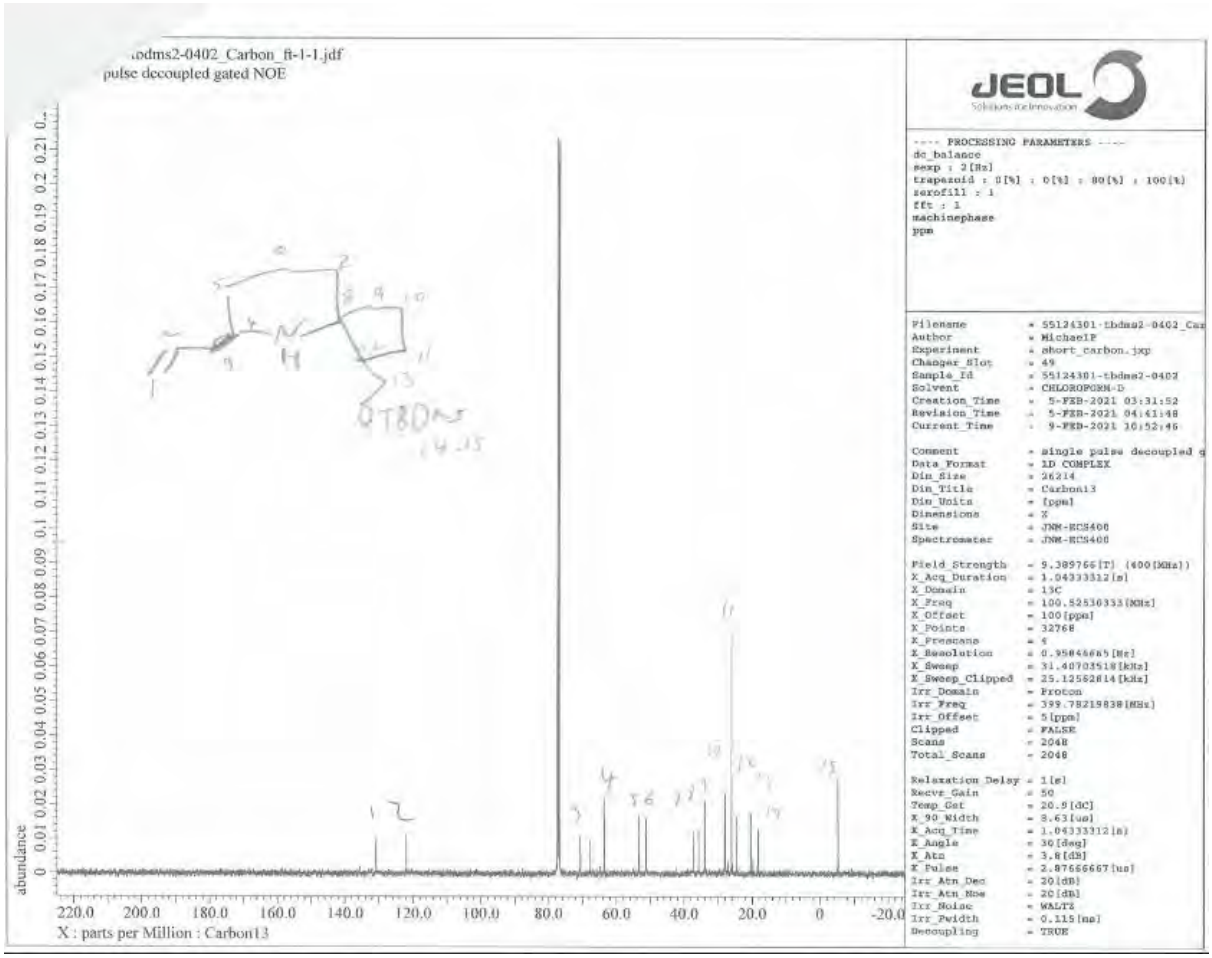



single pulse decoupled gated NOE
JEOL
---- PROCESSING PARAMETERS ----
dc_balance(0, FALSE)
expf(2.0[Hz], 0.0[s])
trapezoid(0[%], 0[%], 80[%], 100[%])
zerofill(1)
fft(1, TRUE, TRUE)
machinephase
ppm
Filename = 55124301-wittigunknowncdc
Author = Michael F
Experiment = long_carbon.jxp
Changer_Slot = 63
Sample_Id = 55124301-wittigunknowncdc
Solvent = CHLOROFORM-D
Creation_Time = 24-MAR-2021 08:36:22
Revision_Time = 25-MAR-2021 15:11:51
Current_Time = 25-MAR-2021 15:12:11
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 829.0
Total_Scans = 829.0
Relaxation_Delay = 1.5[s]
Recvr_Gain = 50
Temp_Set = 22.4[dC]
X_90_Width = 9.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.0[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 39[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE
anundance
X : parts per Million : Carbon13

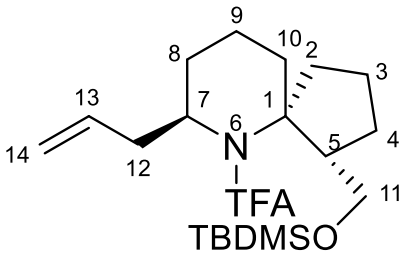
(1S,5S,7R)-7-allyl-1-(((tert-butyldimethylsilyl)oxy)methyl)-6-azaspiro[4.5]decane (194)

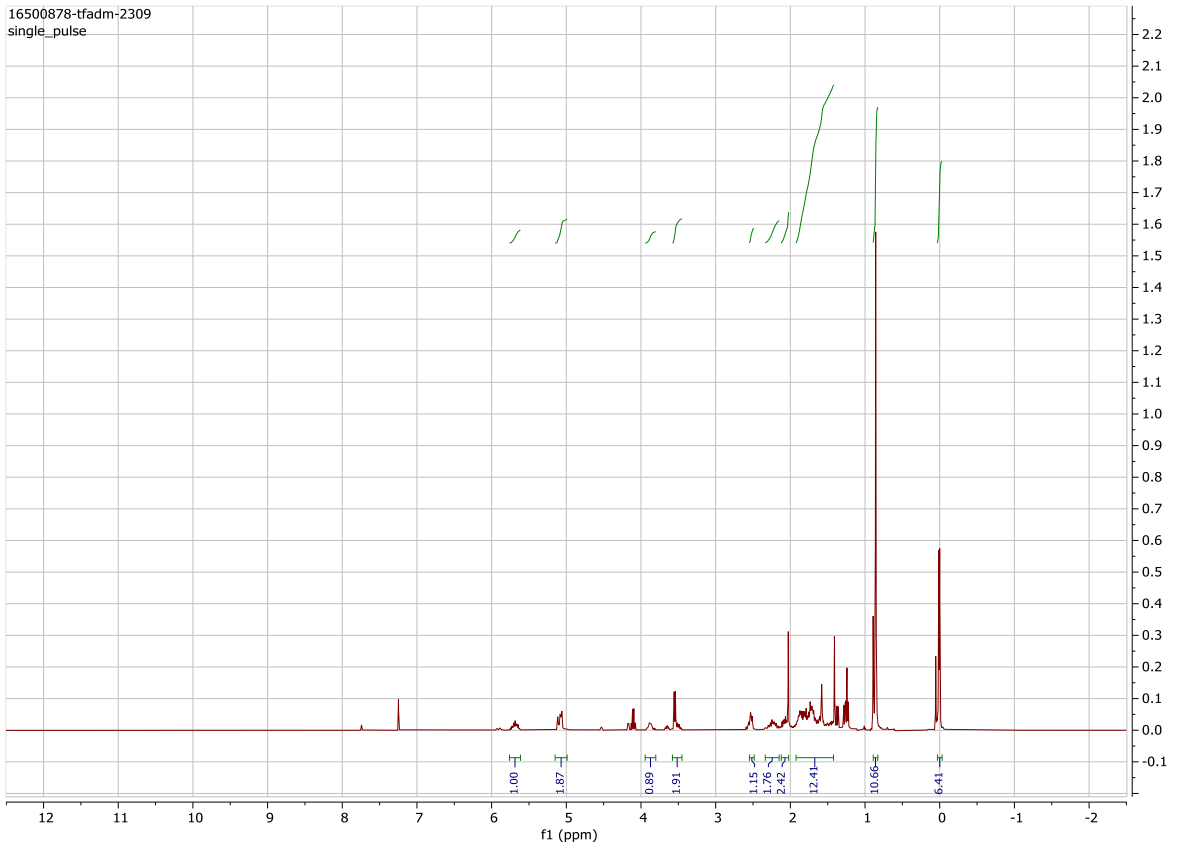


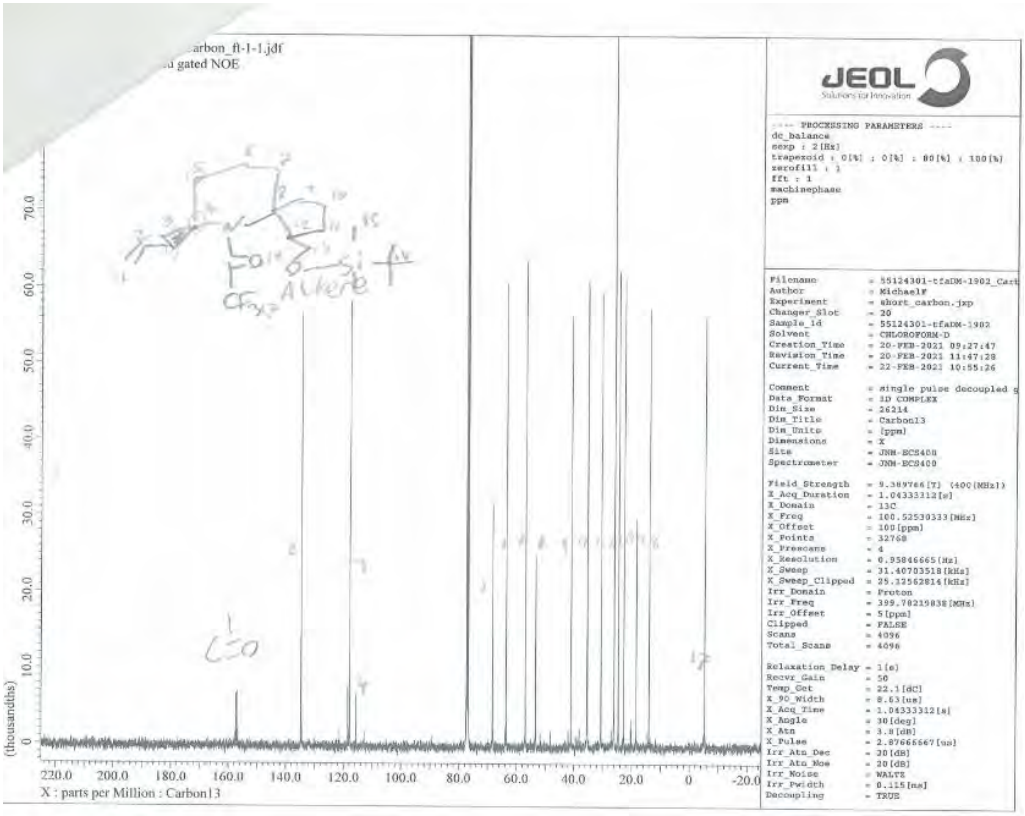



.bdms2-0402_Carbon_fi-1-1.jdf
pulse decoupled gated NOE
JEOL
---- PROCESSING PARAMETERS ----
dc_balance
exp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-tbdms2-0402_Car
Author = MichaelF
Experiment = short_carbon.jxp
Changer_Slot = 49
Sample_Id = 55124301-tbdms2-0402
Solvent = CHLOROFORM-D
Creation_Time = 5-FEB-2021 03:31:52
Revision_Time = 5-FEB-2021 04:41:48
Current_Time = 9-FEB-2021 10:52:46
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846685[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 1[s]
Recvr_Gain = 50
Temp_Get = 20.9[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.6[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE
abundance
X : parts per Million : Carbon13
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0

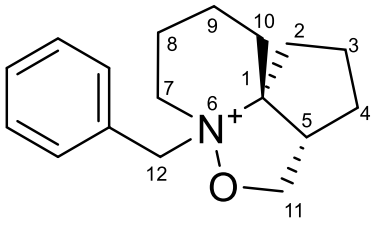
1-((1S,5S,7R)-7-allyl-1-(((tert-butyldimethylsilyl)oxy)methyl)-6-azaspiro[4.5]decan-6-yl)-2,2,2-trifluoroethanone (195)

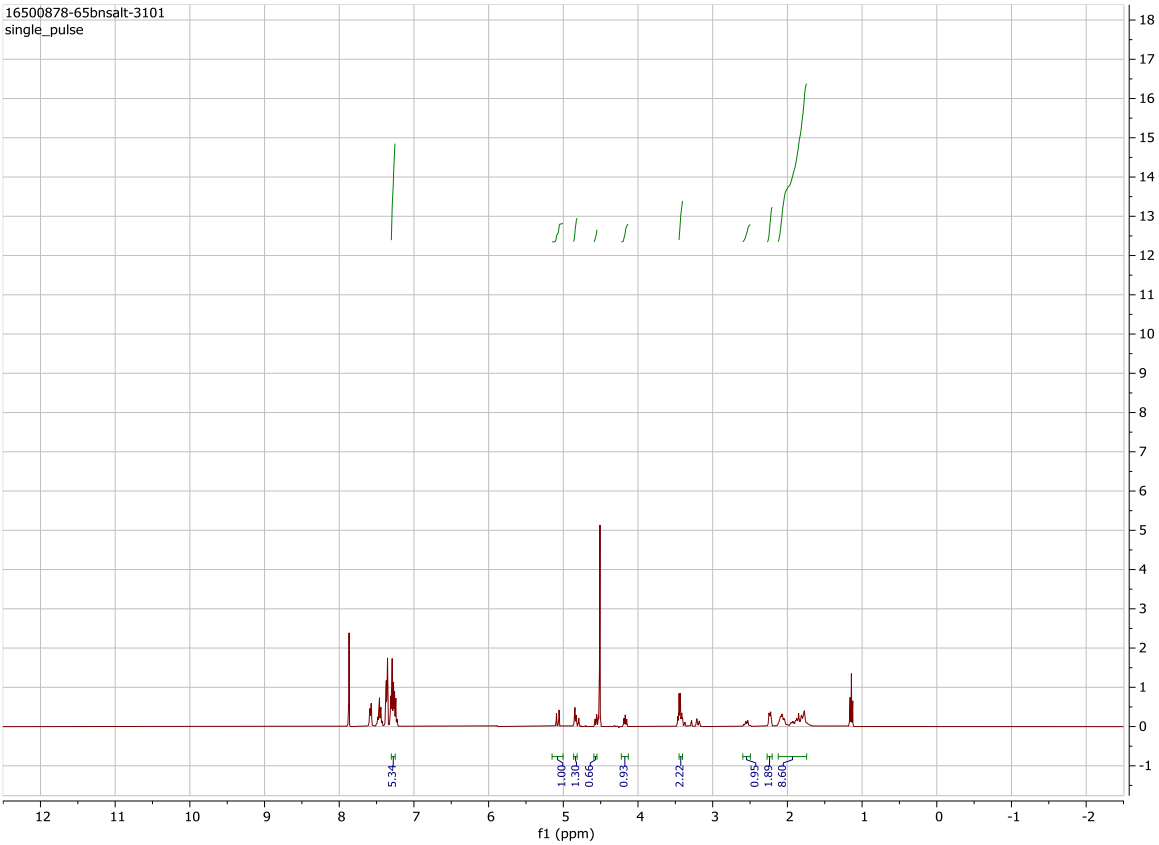


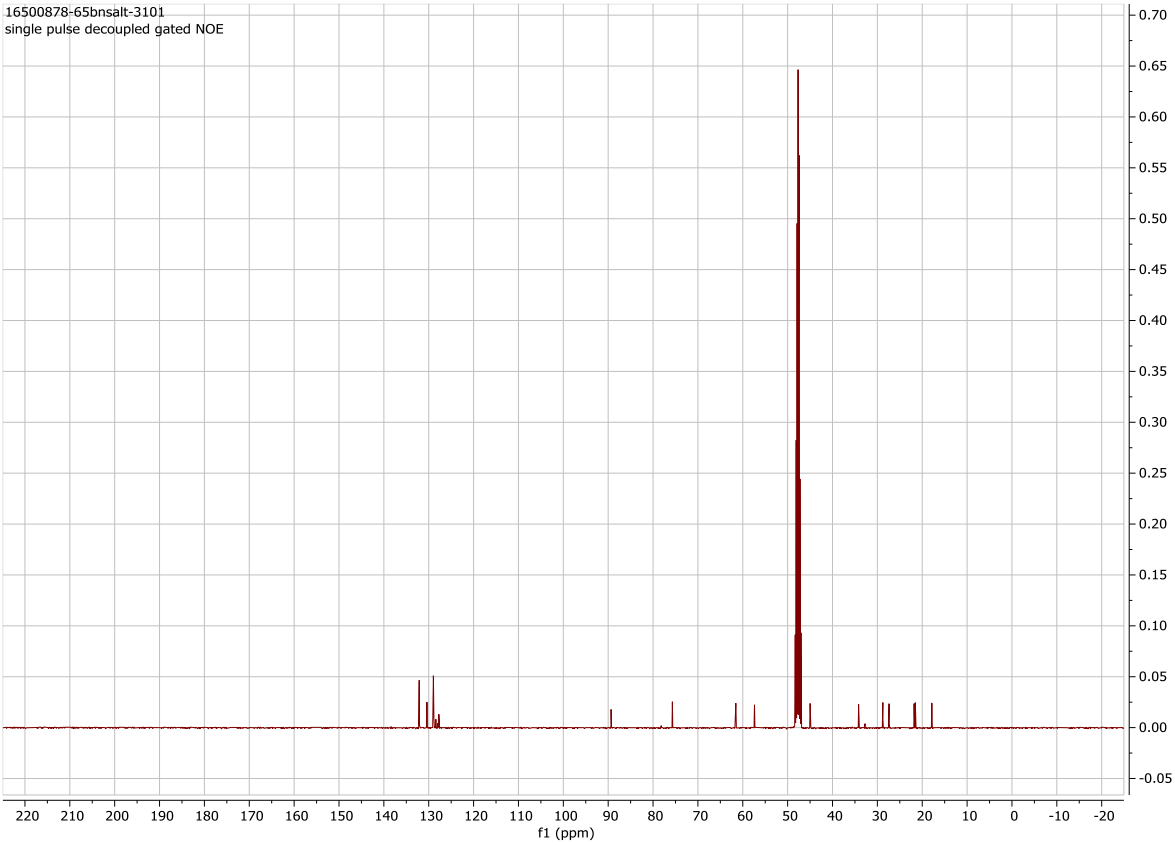



arbon_ft-1-1.jdf
gated NOE
JEOL
Solutions for Innovation
---- PROCESSING PARAMETERS ----
dc_balance
sexp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-tfaNM-1902_Carb
Author = MichaelF
Experiment = short_carbon.jxp
Changer_Slot = 20
Sample_Id = 55124301-tfaNM-1902
Solvent = CHLOROFORM-D
Creation_Time = 20-FEB-2021 09:27:47
Revision_Time = 20-FEB-2021 11:47:28
Current_Time = 22-FEB-2021 10:55:26
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78215838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 4096
Total_Scans = 4096
Relaxation_Delay = 1[s]
Recvr_Gain = 50
Temp_Get = 22.1[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.8[dB]
X_Pulse = 2.87666567[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 9.115[ms]
Decoupling = TRUE
(thousandths)
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0
X : parts per Million : Carbon13

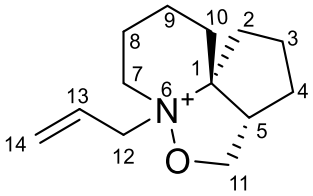
(1S*,5S*)6-benzyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (212)

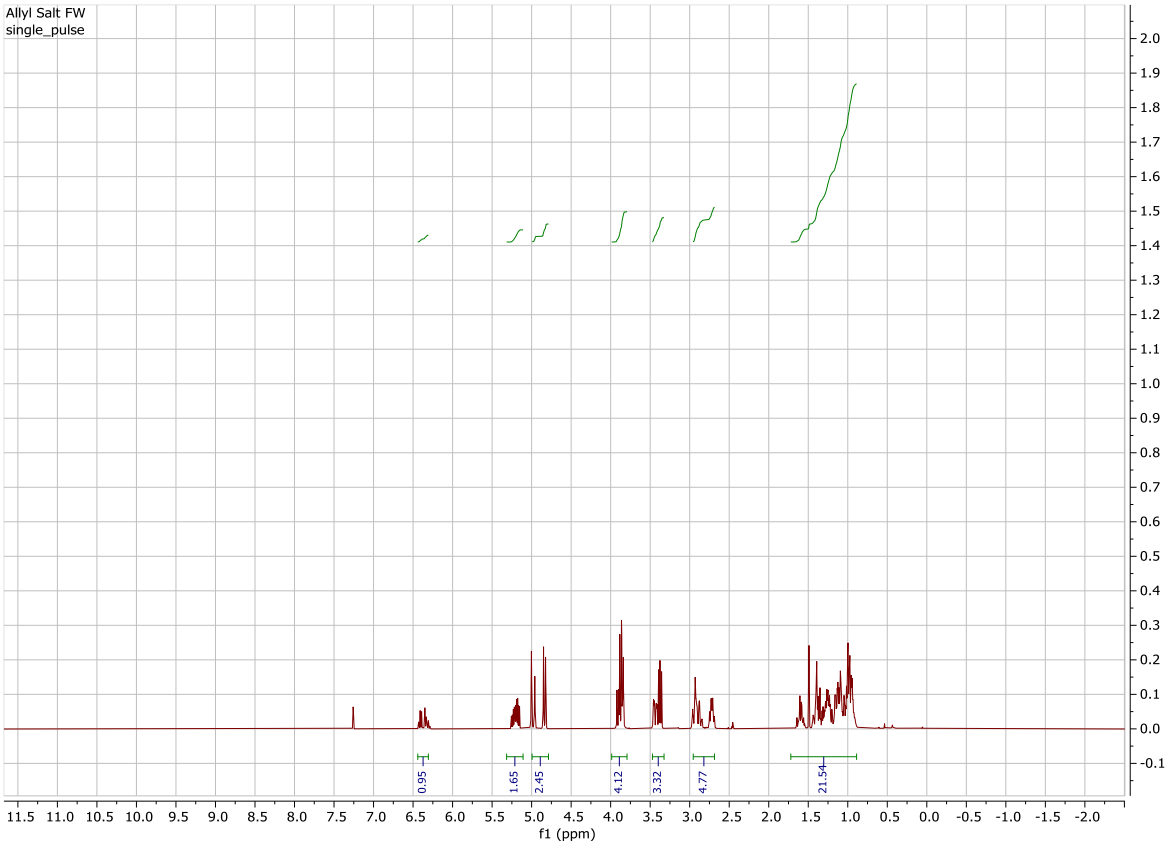


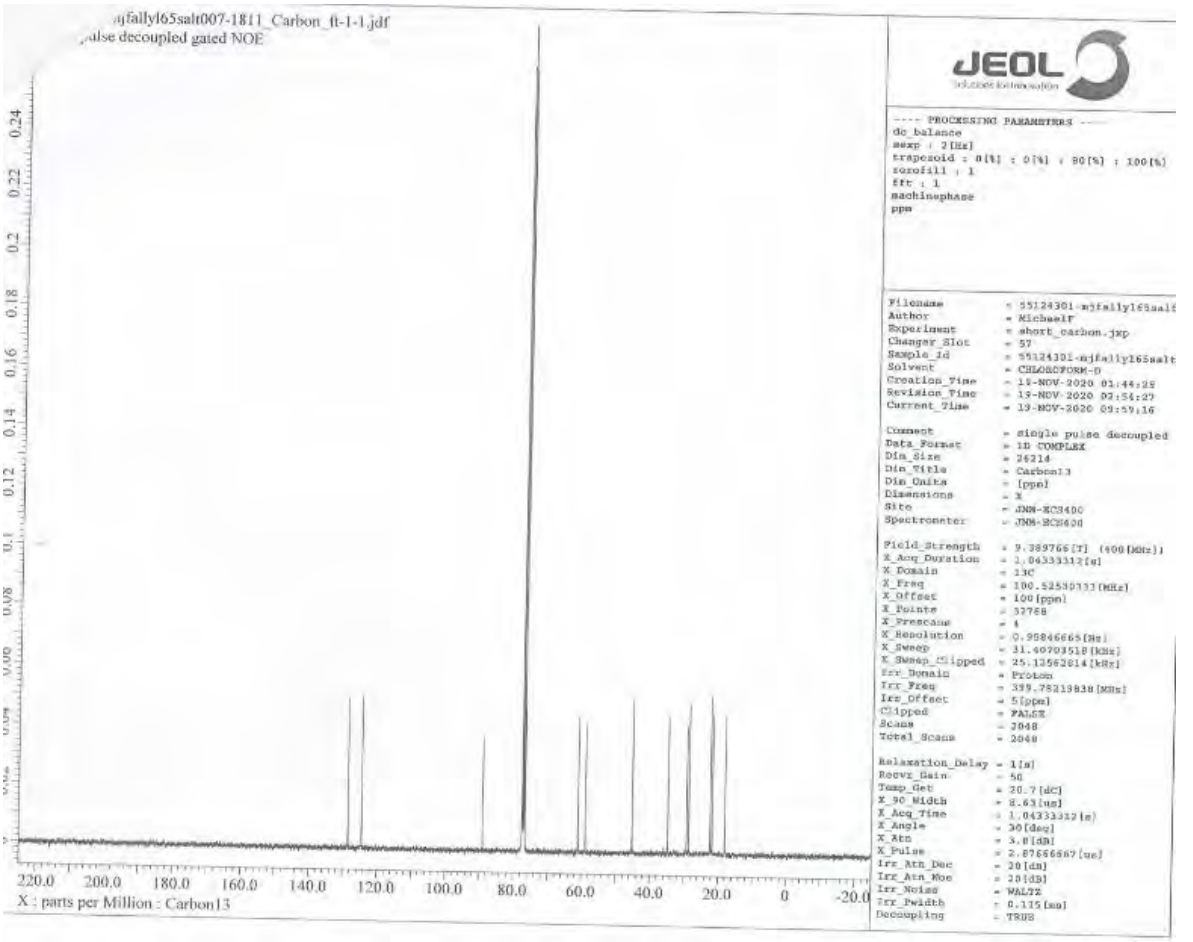



16500878-65bnsalt-3101
single pulse decoupled gated NOE
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

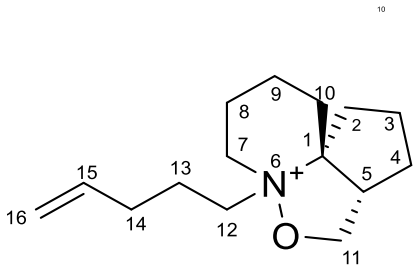
(1S*,5S*)-6-allyloctahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (213)

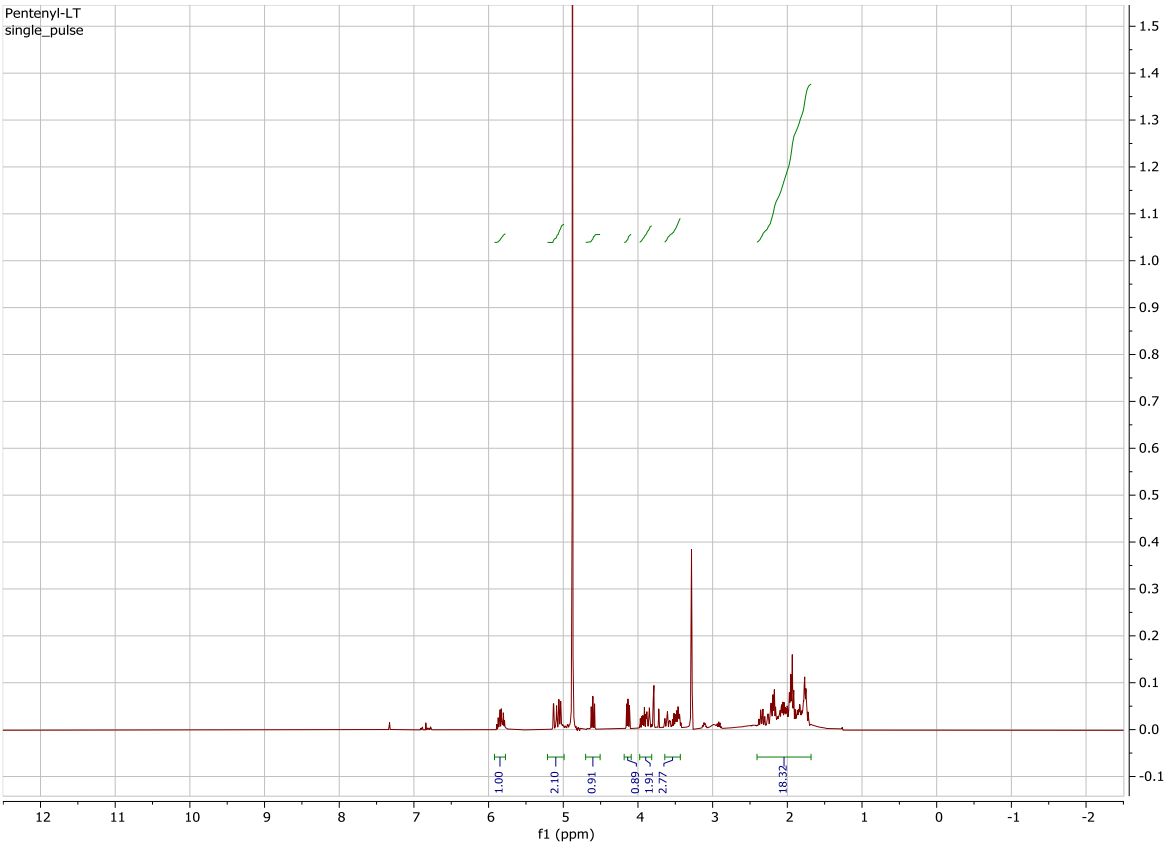


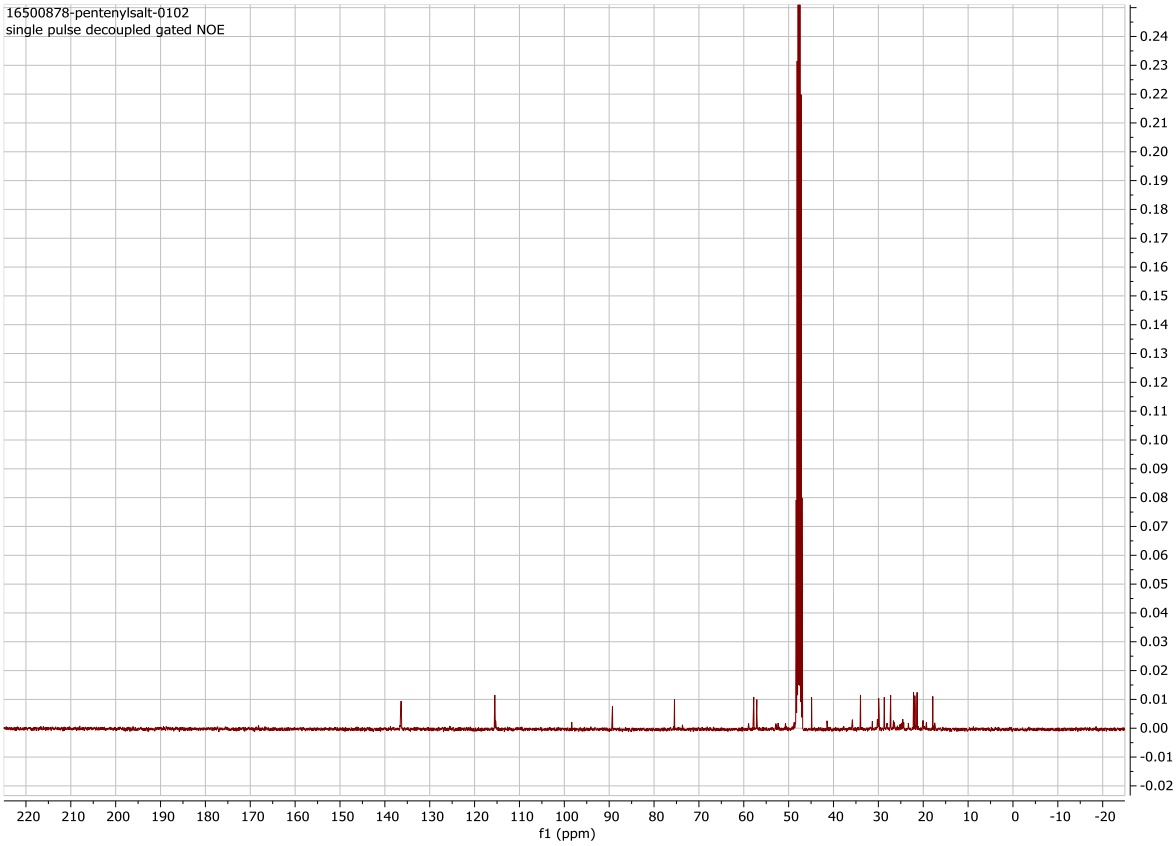



mjfally165salt007-1811_Carbon_ft-1-1.jdf
ulse decoupled gated NOE
0.24
0.22
0.2
0.18
0.16
0.14
0.12
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0
X : parts per Million : Carbon13
JEOL
---- PROCESSING PARAMETERS ----
dc_balance
exp : 2[Hz]
trapezoid : 0[%] : 0[%] : 90[%] : 100[%]
zerofill : 1
fft : 1
manualphase
ppm
Filename = 55124301-mjfally165salt
Author = Michaelf
Experiment = short_carbon.jxp
Changer_Slot = 57
Sample_Id = 55124301-mjfally165salt
Solvent = CHLOROFORM-D
Creation_Time = 19-NOV-2020 01:44:28
Revision_Time = 19-NOV-2020 02:56:27
Current_Time = 19-NOV-2020 09:59:16
Comment = single pulse decoupled
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-EC3400
Spectrometer = JNM-EC3400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78213838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 1[s]
Recvr_Gain = 50
Temp_Get = 20.7[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.0[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Mod = 28[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE

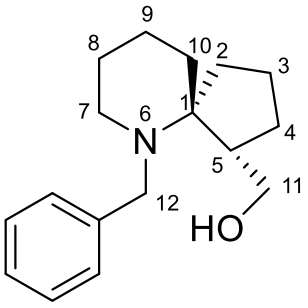
(1S*,5S*)-6-(pent-4-en-1-yl)octahydro-1H,8H-cyclopenta[3,4]isoxazolo[2,3-a]pyridin-6-ium bromide (214)

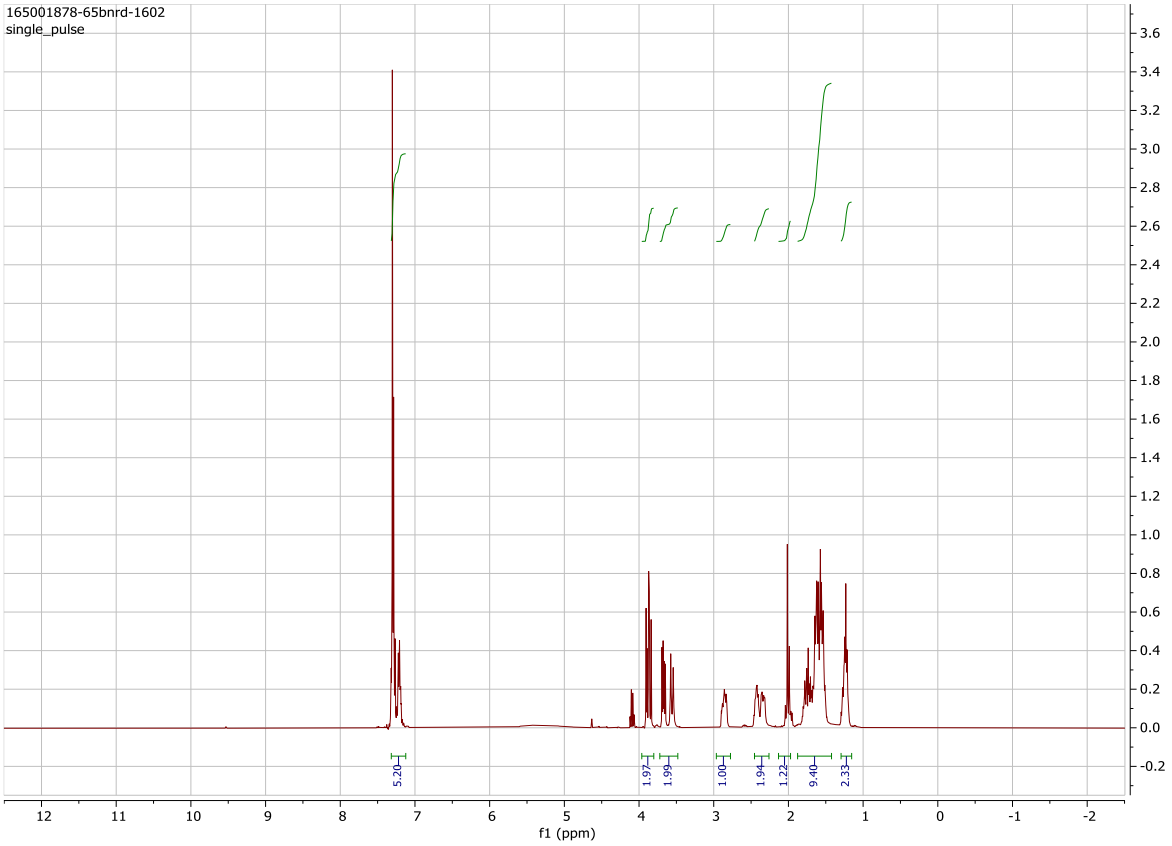


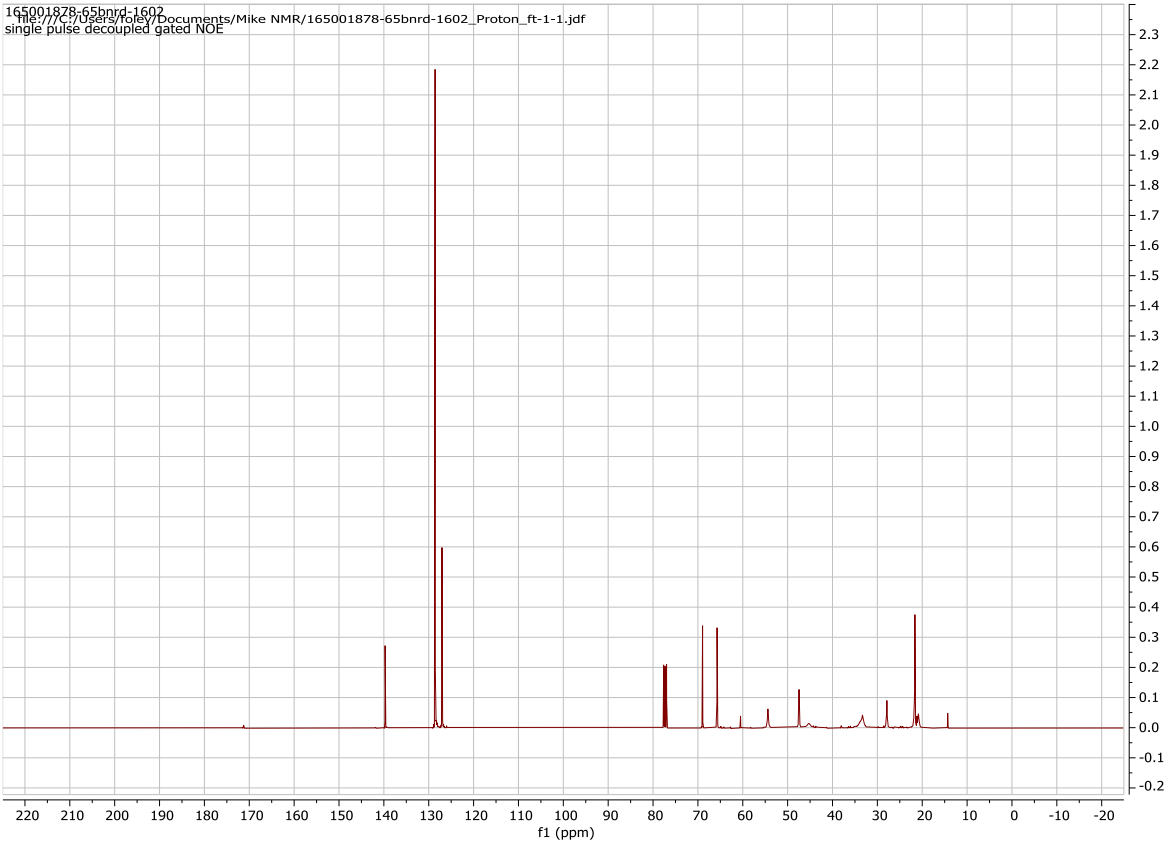



16500878-pentenylsalt-0102
single pulse decoupled gated NOE
0.24
0.23
0.22
0.21
0.20
0.19
0.18
0.17
0.16
0.15
0.14
0.13
0.12
0.11
0.10
0.09
0.08
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0.00
-0.01
-0.02
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

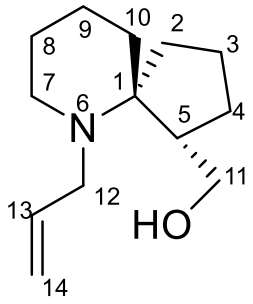
(1S*,5S*) -(6-benzyl-6-azaspiro[4.5]decan-1-yl)methanol (217)

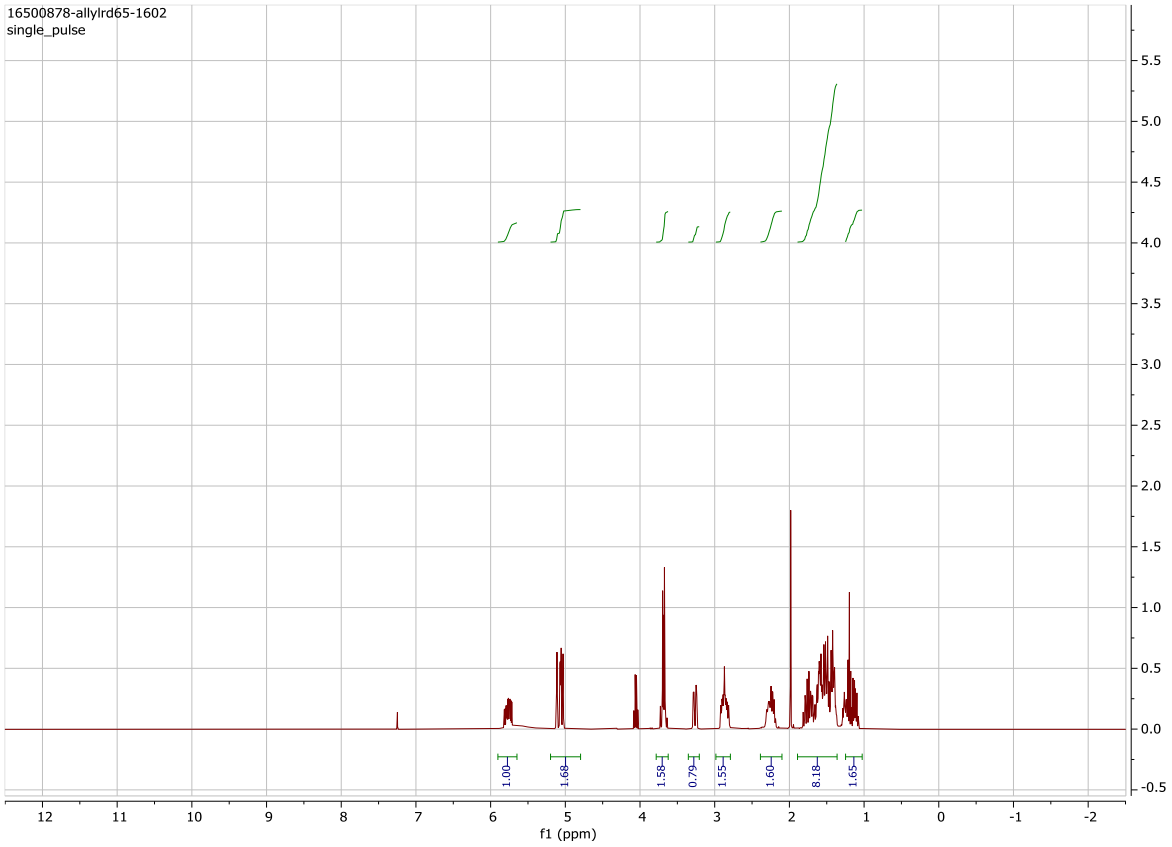


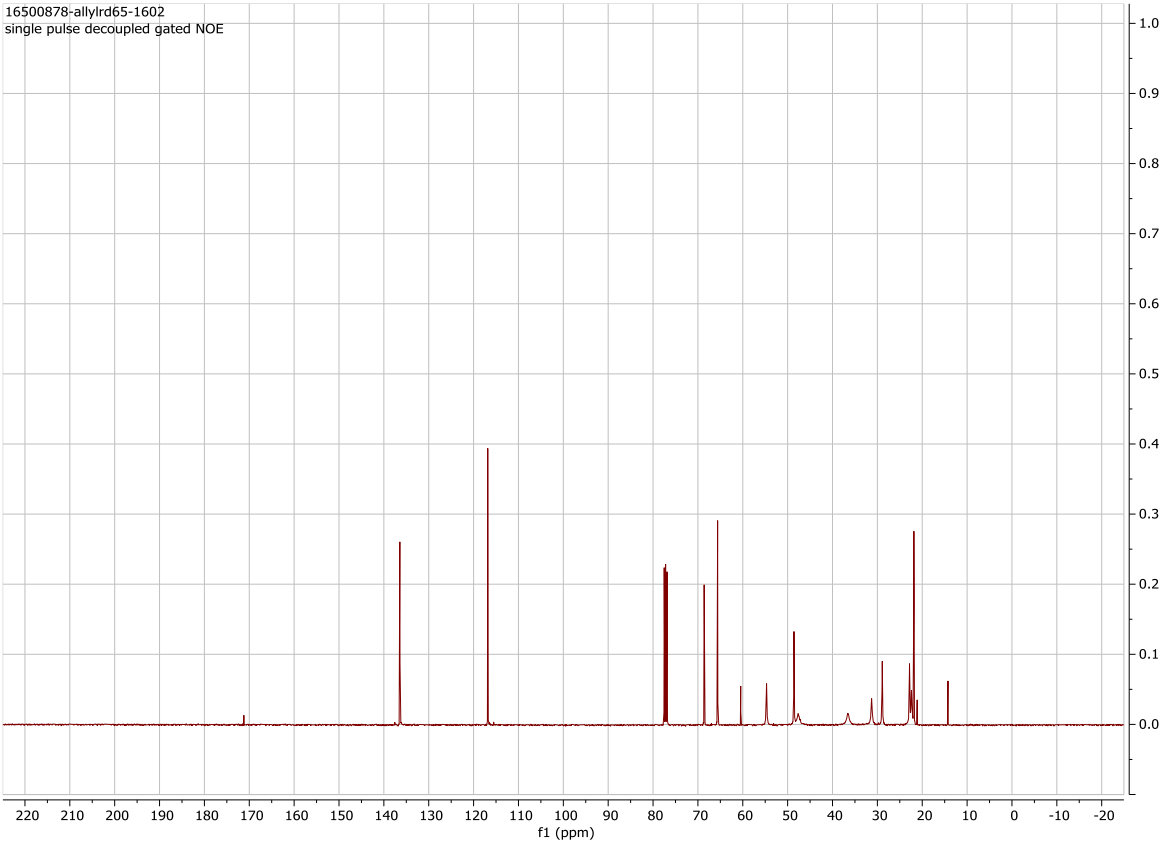



165001878-65bnrd-1602
file:///C:/Users/foley/Documents/Mike NMR/165001878-65bnrd-1602_Proton_ft-1-1.jdf
single pulse decoupled gated NOE
f1 (ppm)

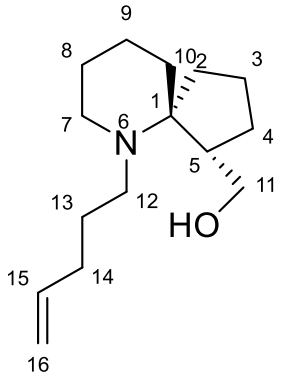
(1S*,5S*)-(6-allyl-6-azaspiro[4.5]decan-1-yl)methanol (218)

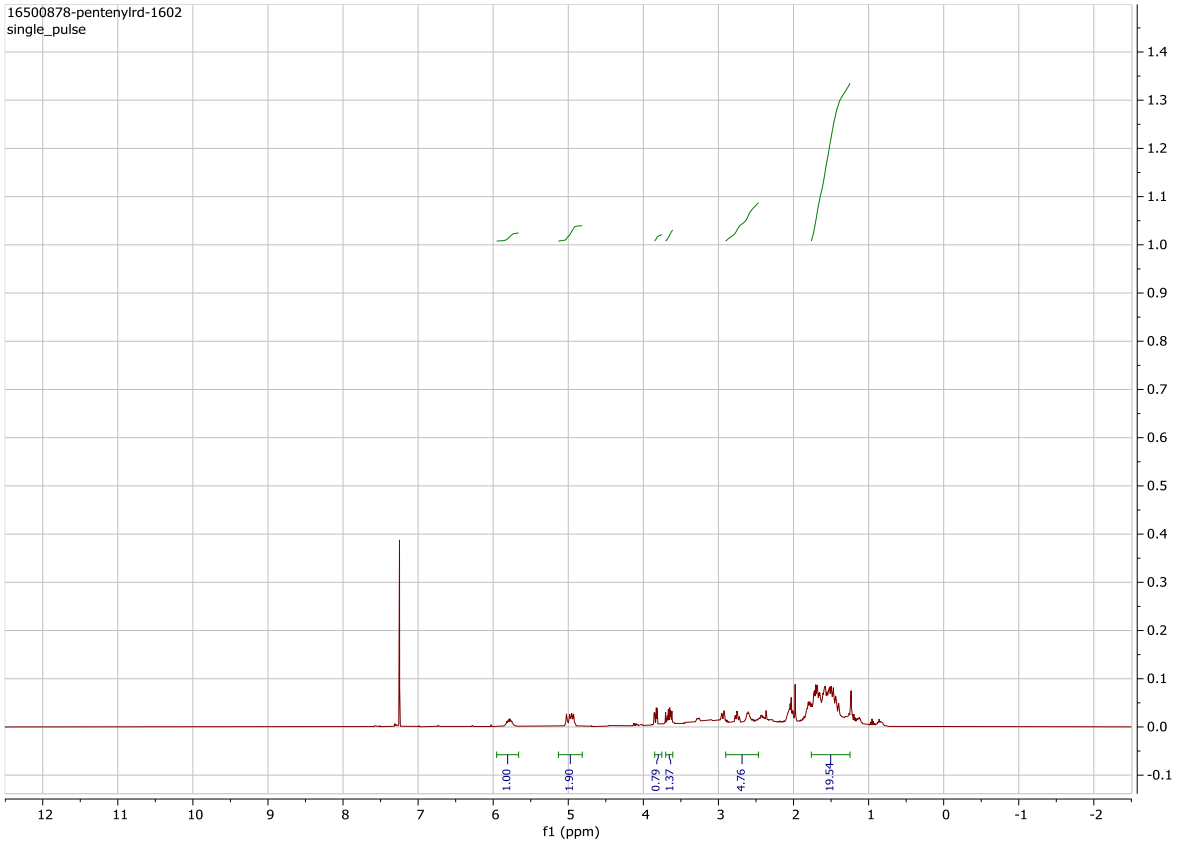


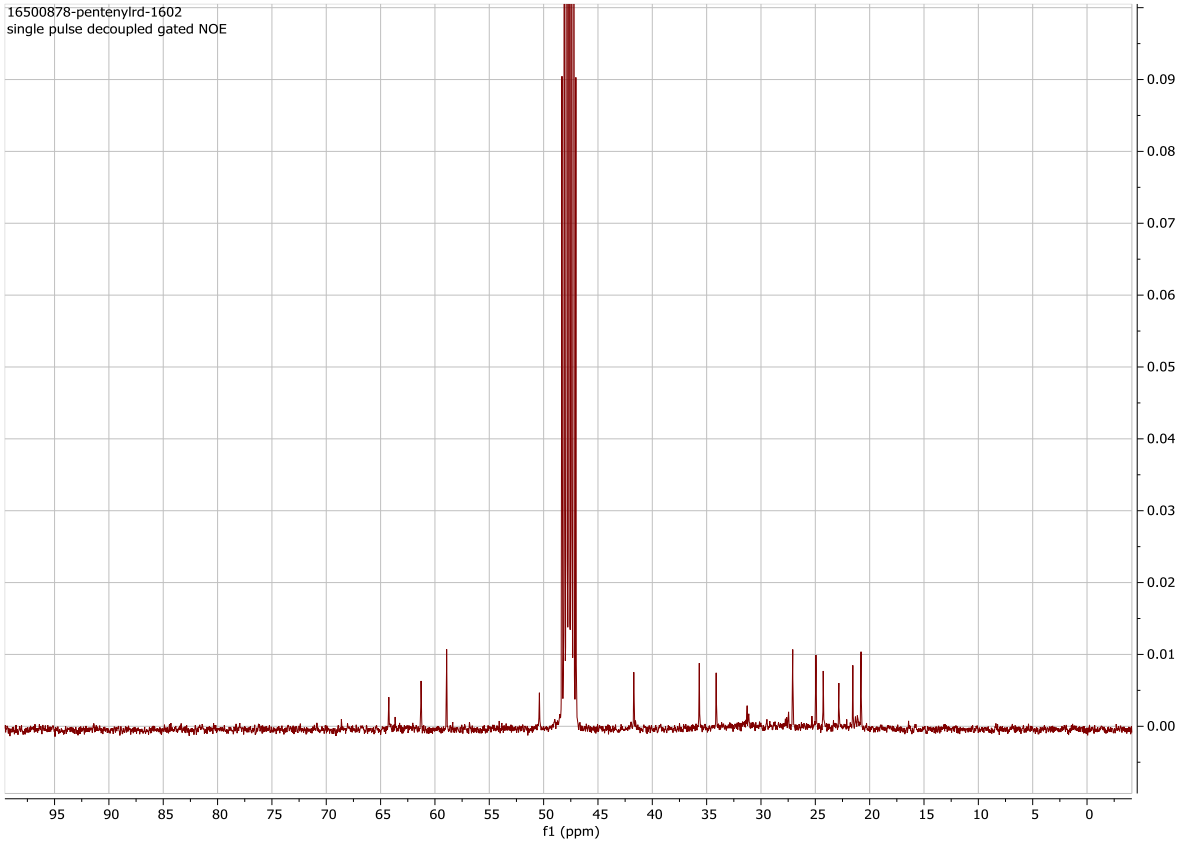



16500878-allylrd65-1602
single pulse decoupled gated NOE
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

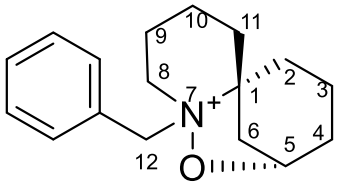
(1S*,5S*)-(6-(pent-4-en-1-yl)-6-azaspiro[4.5]decan-1-yl)methanol (219)

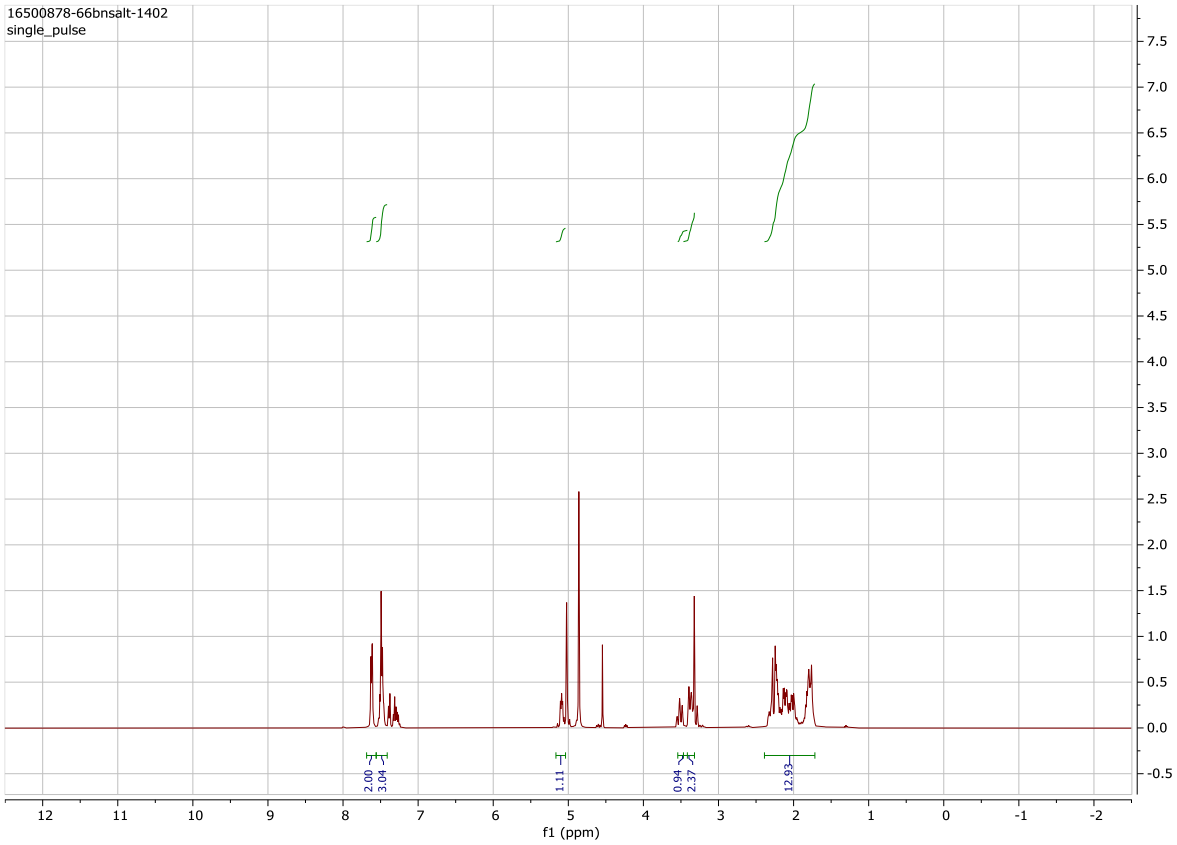


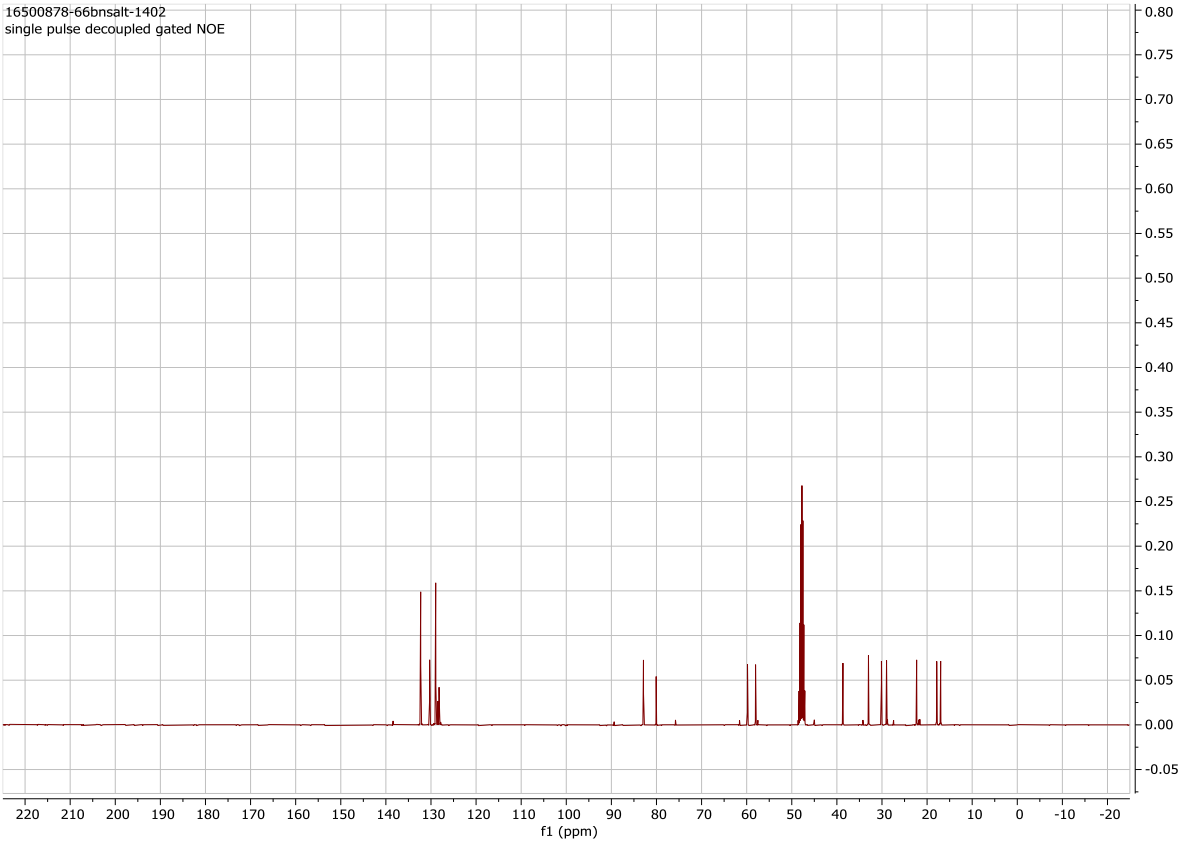



16500878-pentenylrd-1602
single pulse decoupled gated NOE
0.09
0.08
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0.00
95
90
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
f1 (ppm)

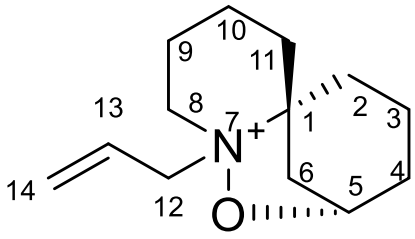
(1S*,5S*)-7-benzyloctahydro-6H-2,5a-methanopyrido[1,2-b][1,2]oxazepin-10-ium bromide (220)

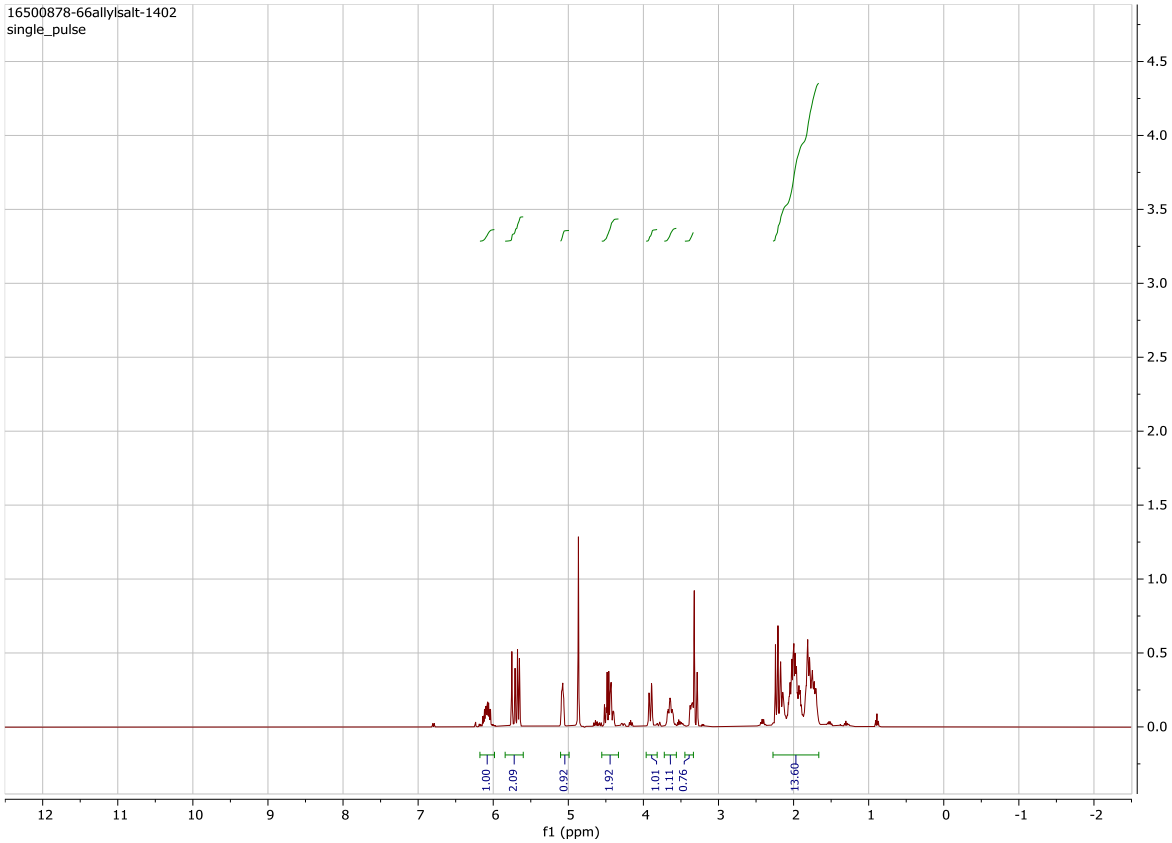


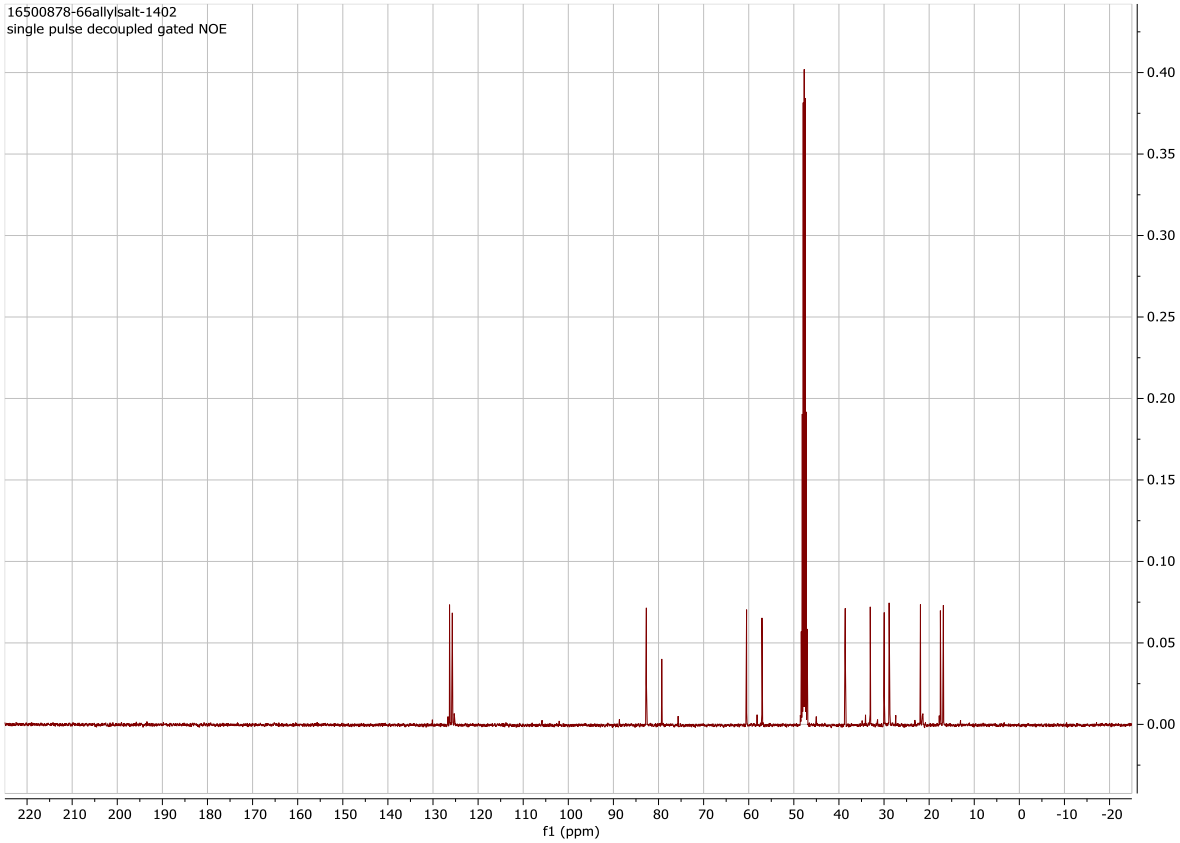



16500878-66bnsalt-1402
single pulse decoupled gated NOE
0.80
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

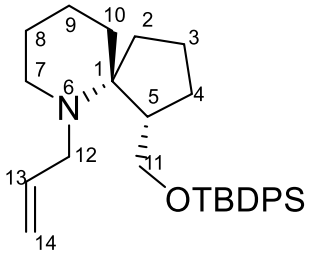
(1S*,5S*)-7-allyloctahydro-5H-2,5a-methanopyrido[1,2-b][1,2]oxazepin-12-ium bromide (221)

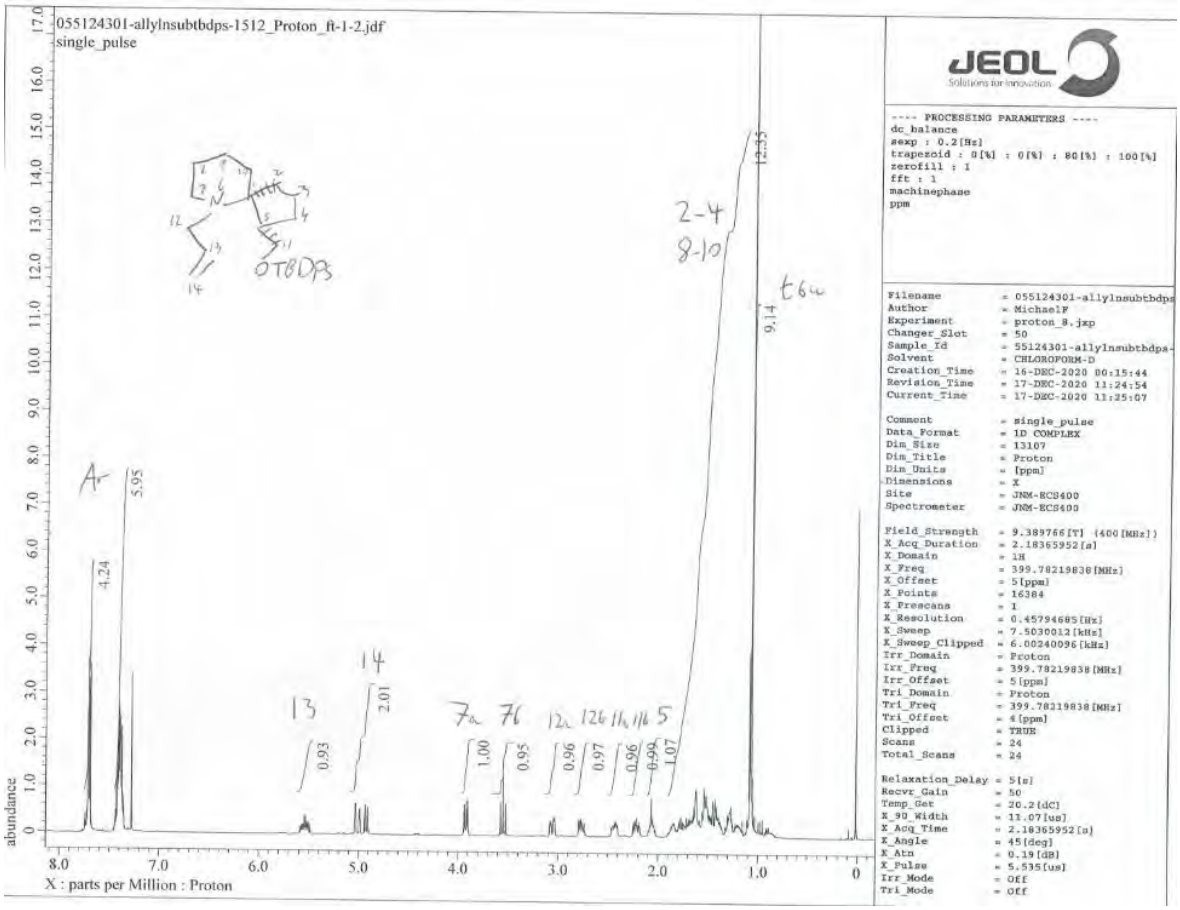


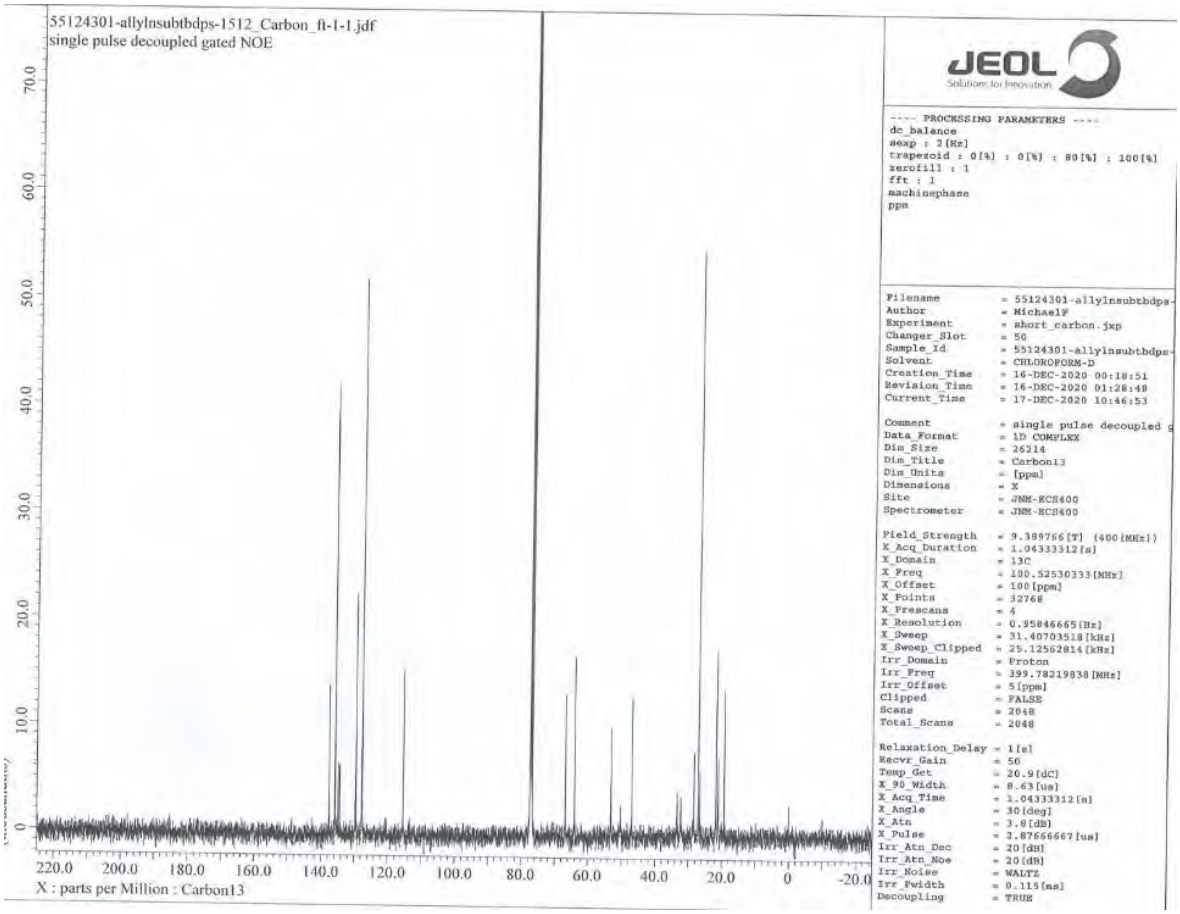



16500878-66allylsalt-1402
single pulse decoupled gated NOE
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

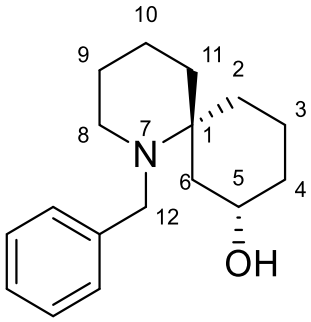
(1S*,5S*)-6-allyl-1-(((tert-butyldiphenylsilyl)oxy)methyl)-6-azaspiro[4.5]decane (224)

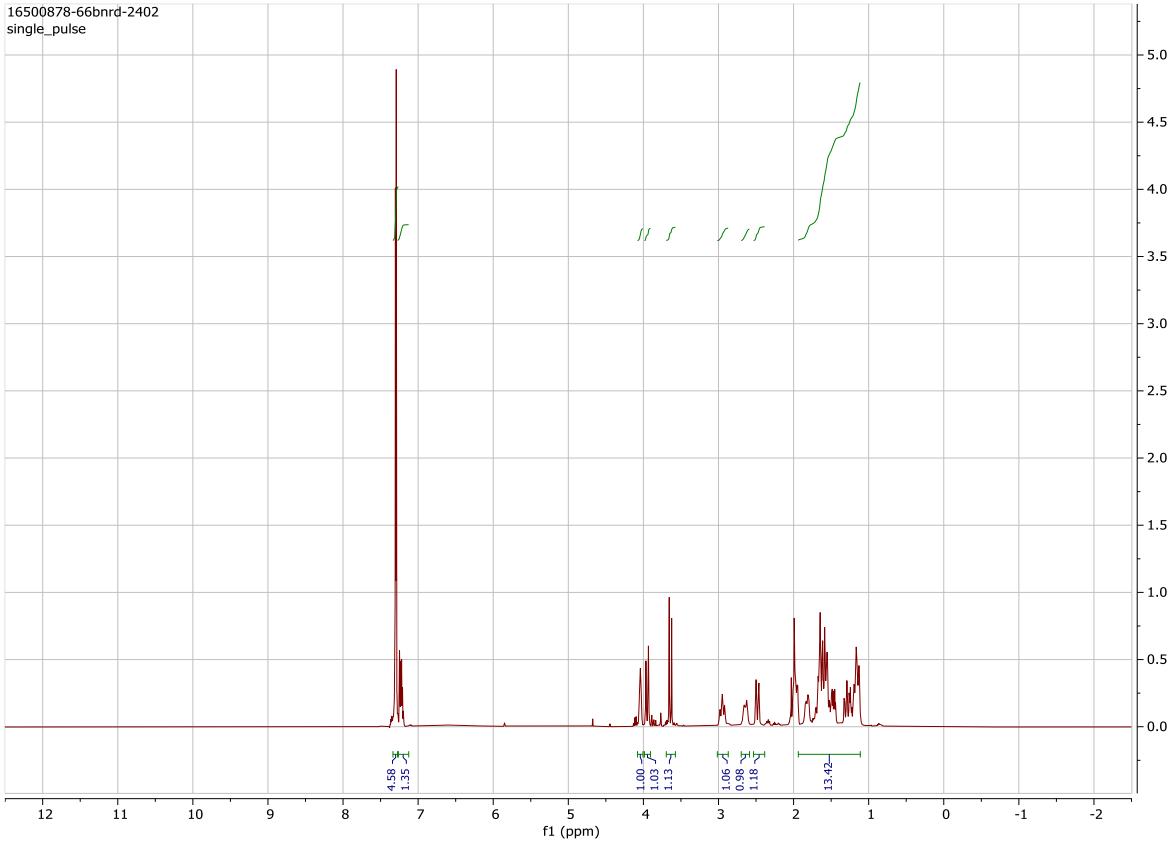


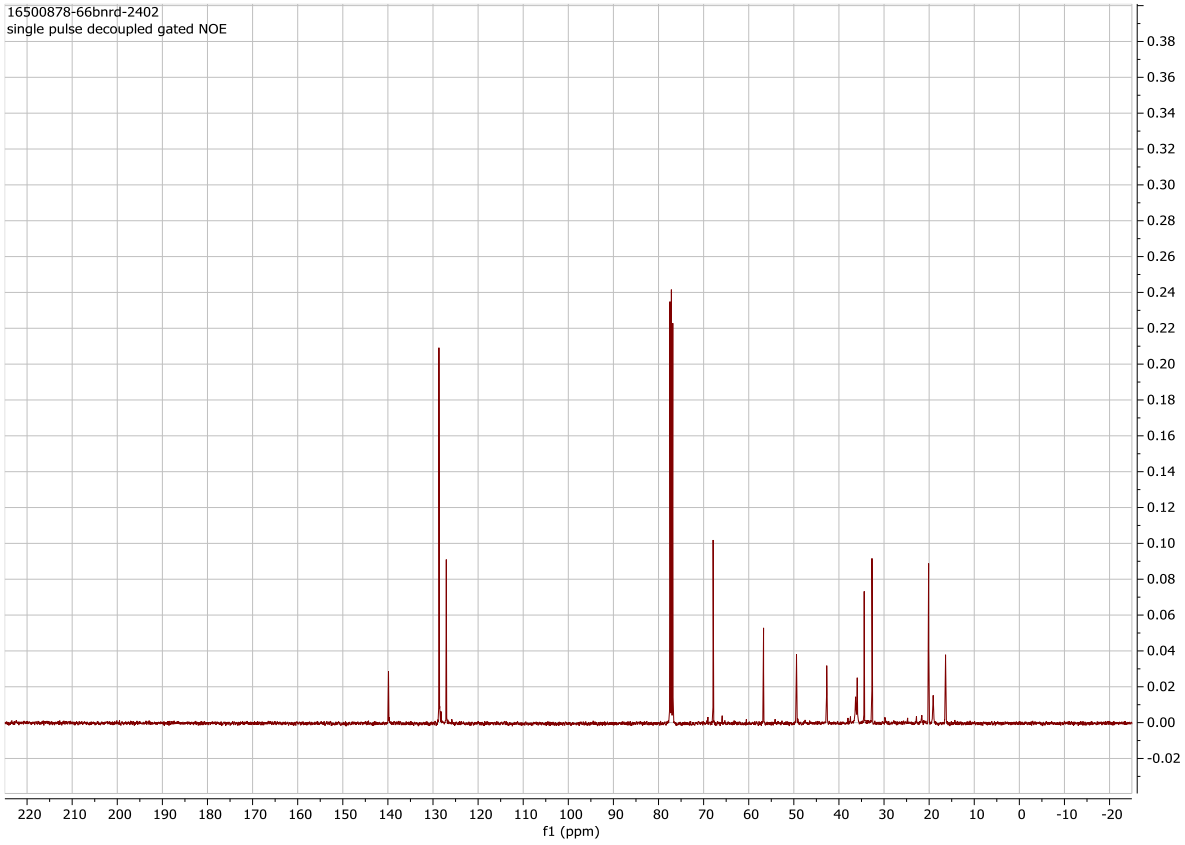



55124301-allylnsubtbdps-1512_Carbon_ft-1-1.jdf
single pulse decoupled gated NOE
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0
X : parts per Million : Carbon13
JEOL
Solutions for Innovation
---- PROCESSING PARAMETERS ----
dc_balance
exp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-allylnsubtbdps-
Author = MichaelF
Experiment = short_carbon.jxp
Changer_Slot = 50
Sample_Id = 55124301-allylnsubtbdps-
Solvent = CHLOROFORM-D
Creation_Time = 16-DEC-2020 00:18:51
Revision_Time = 16-DEC-2020 01:28:48
Current_Time = 17-DEC-2020 10:46:53
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 1[s]
Recvr_Gain = 56
Temp_Get = 20.9[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.8[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE

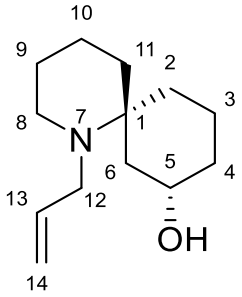
(1S*,5S*)-7-benzyl-1-azaspiro[5.5]undecan-8-ol (222)

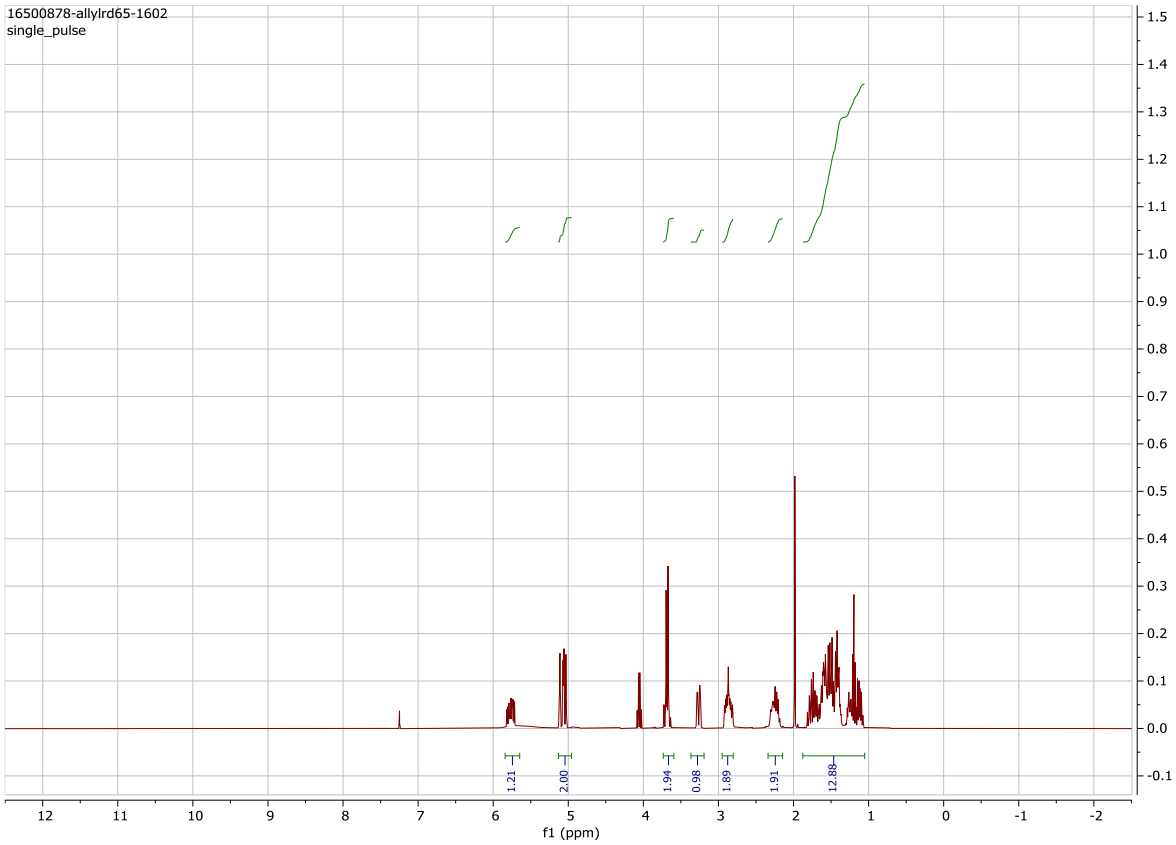


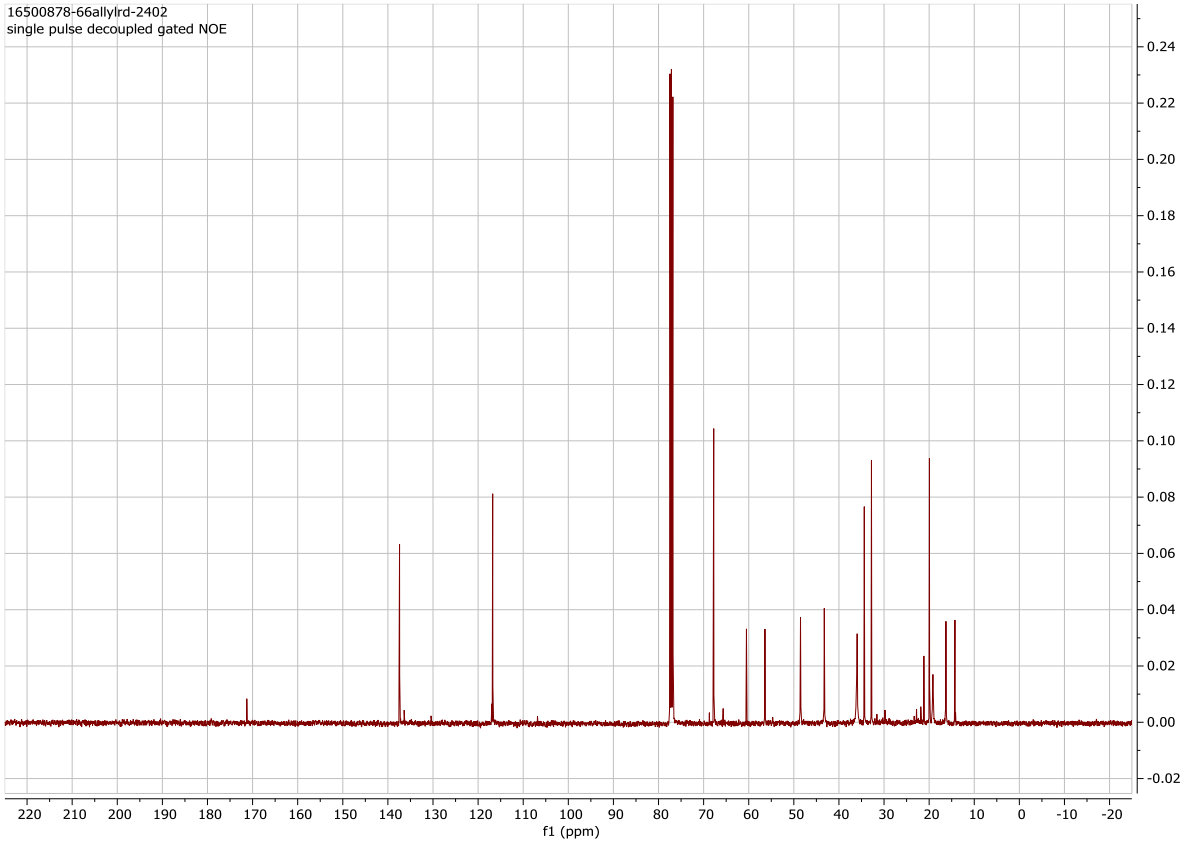



16500878-66bnrd-2402
single pulse decoupled gated NOE
0.38
0.36
0.34
0.32
0.30
0.28
0.26
0.24
0.22
0.20
0.18
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
-0.02
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

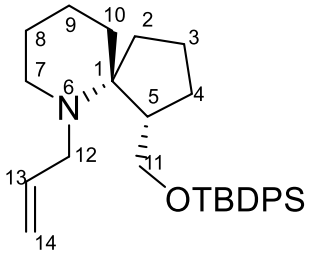
(1S*,5S*)-7-allyl-1-azaspiro[5.5]undecan-8-ol (223)

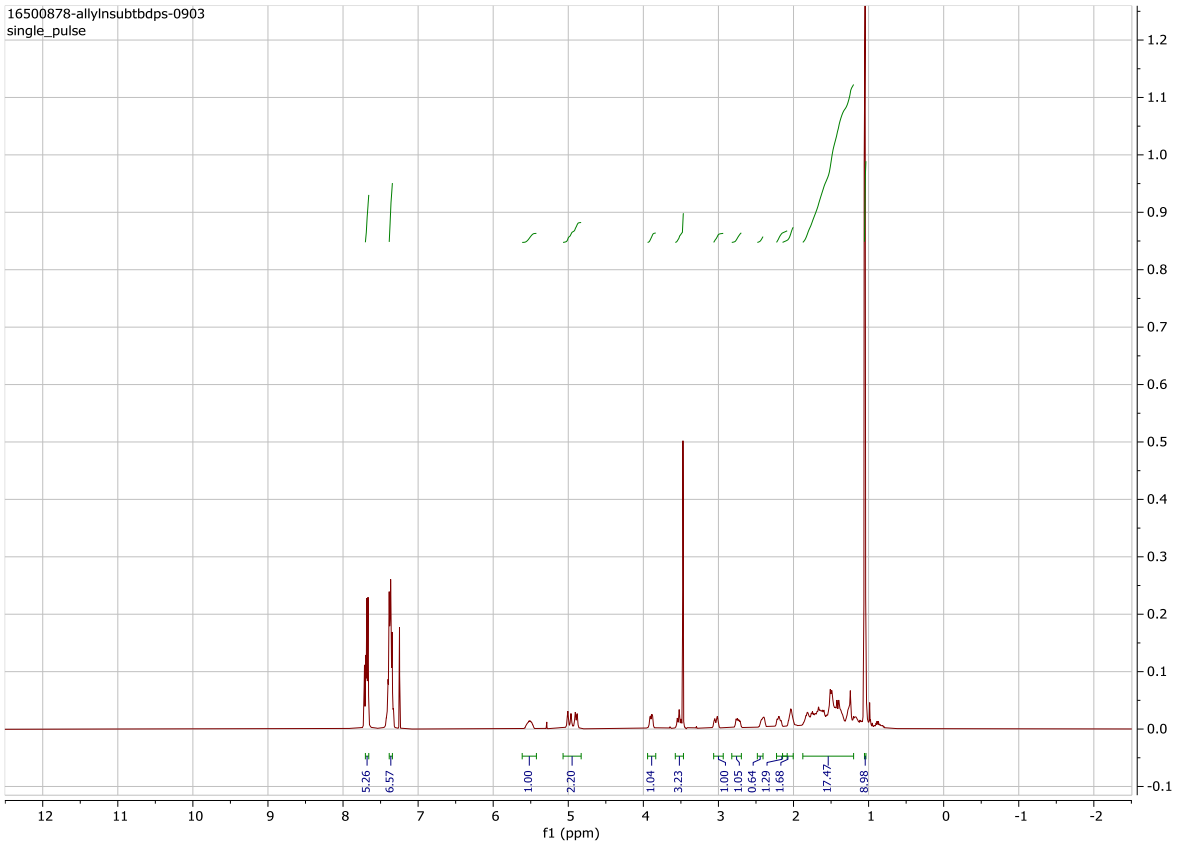


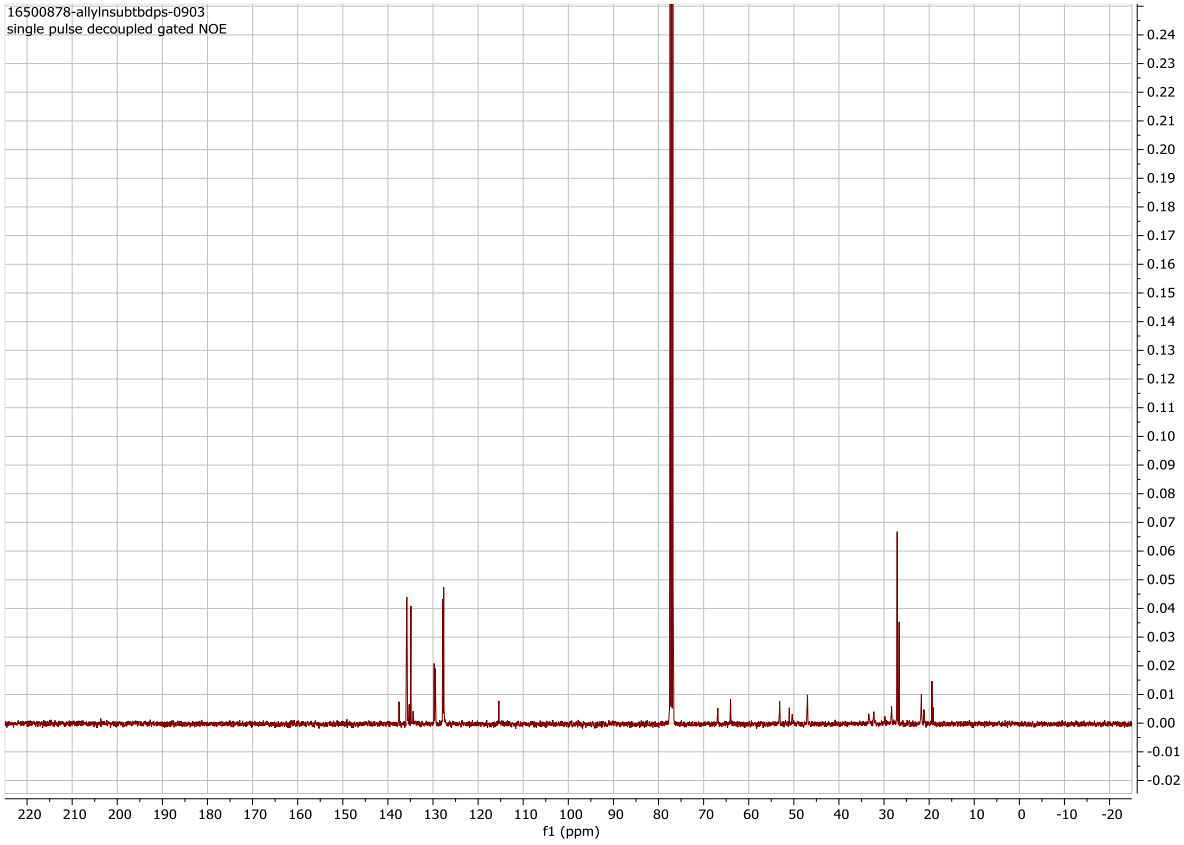



16500878-66allylrd-2402
single pulse decoupled gated NOE
0.24
0.22
0.20
0.18
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
-0.02
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

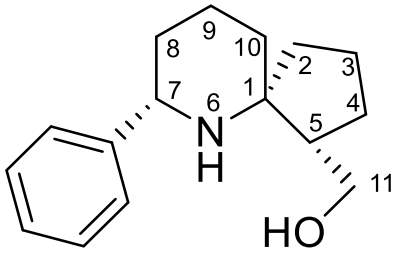
(1S*,5S*)-6-allyl-1-(((tert-butyldiphenylsilyl)oxy)methyl)-6-azaspiro[4.5]decane (224)

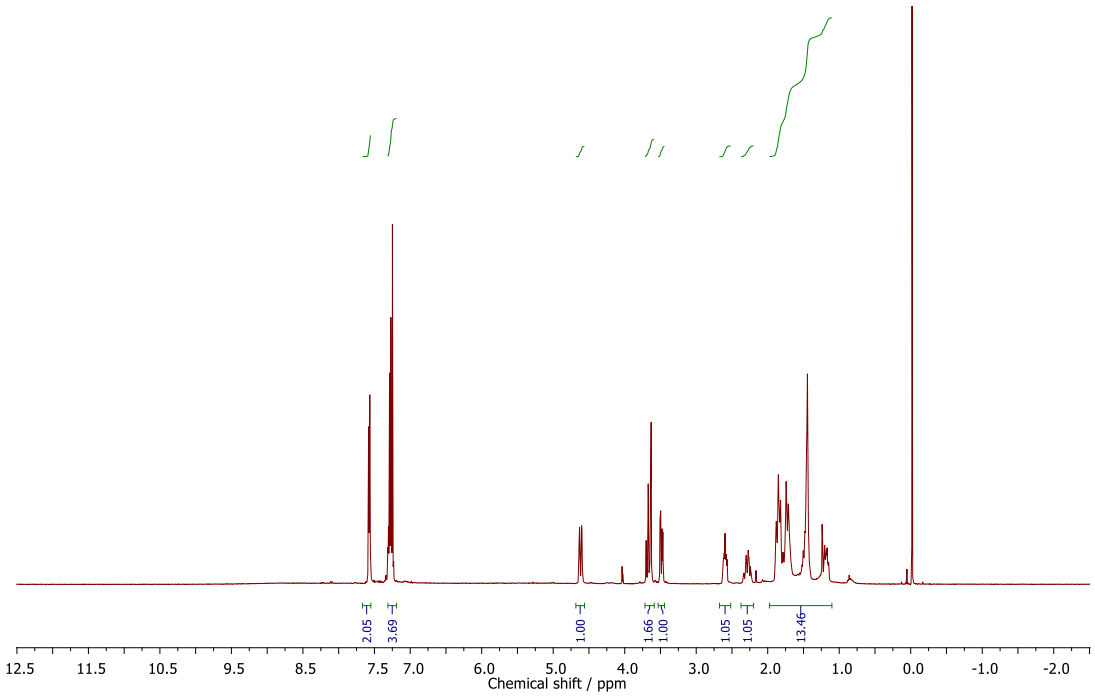


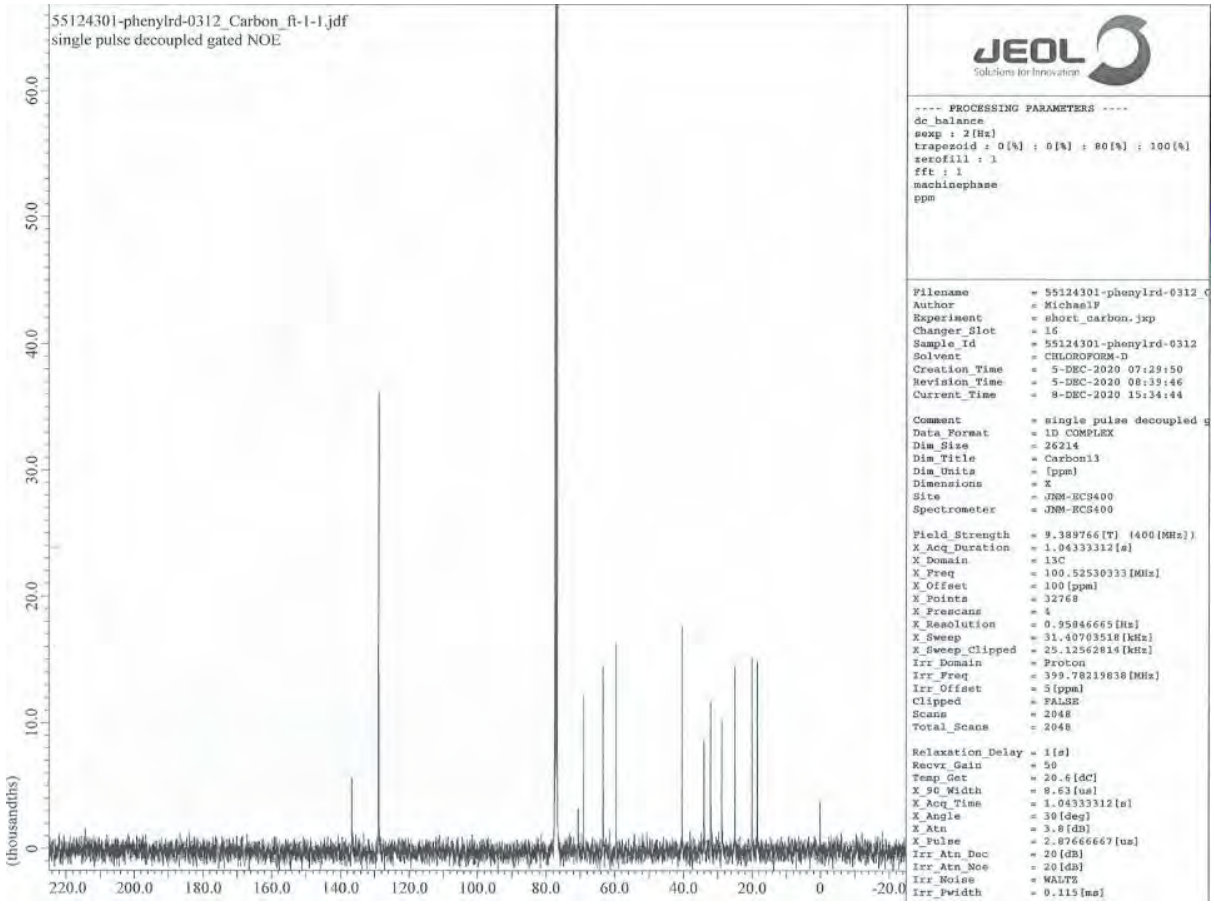



16500878-allylnsubtbdps-0903
single pulse decoupled gated NOE
f1 (ppm)

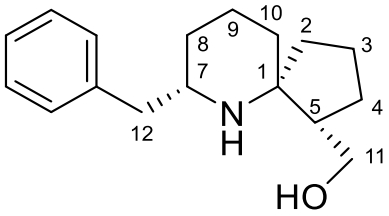
(1S*,5S*,7S*)-7-phenyl-6-azaspiro[4.5]decan-1-yl)methanol (229)

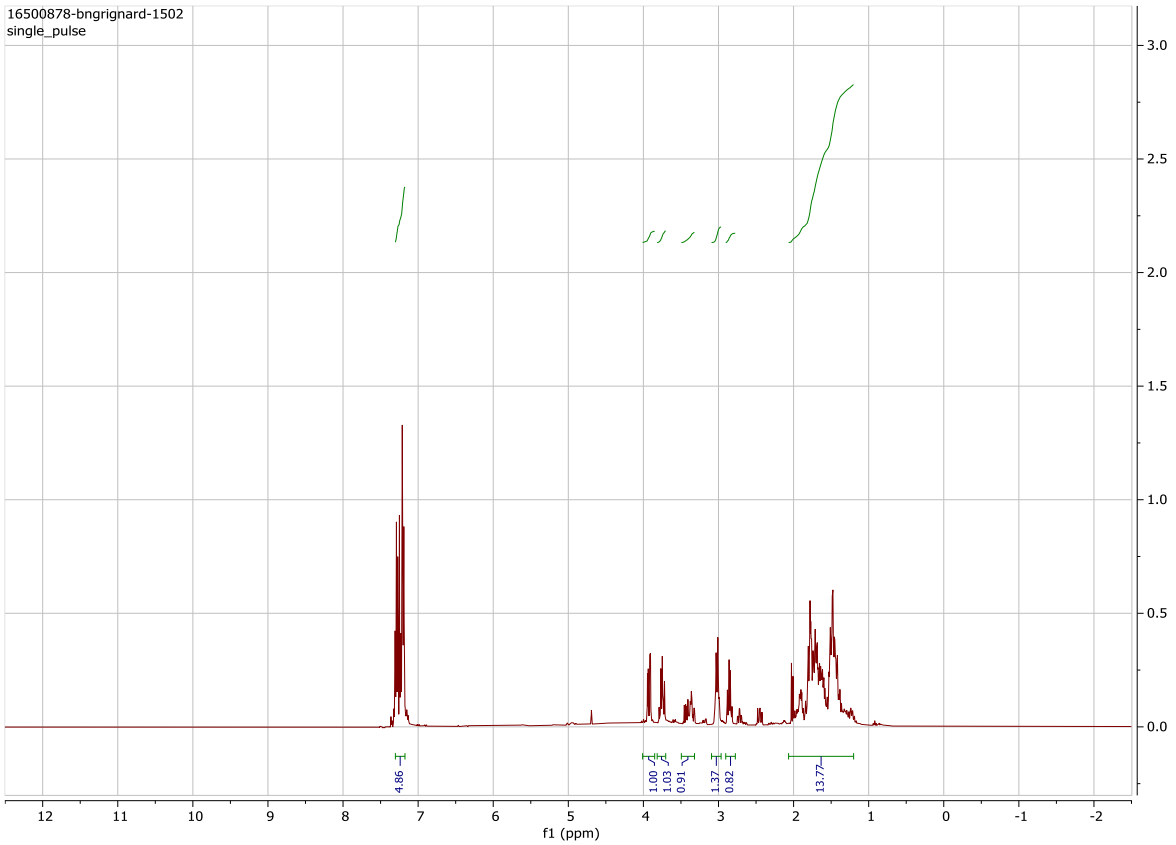


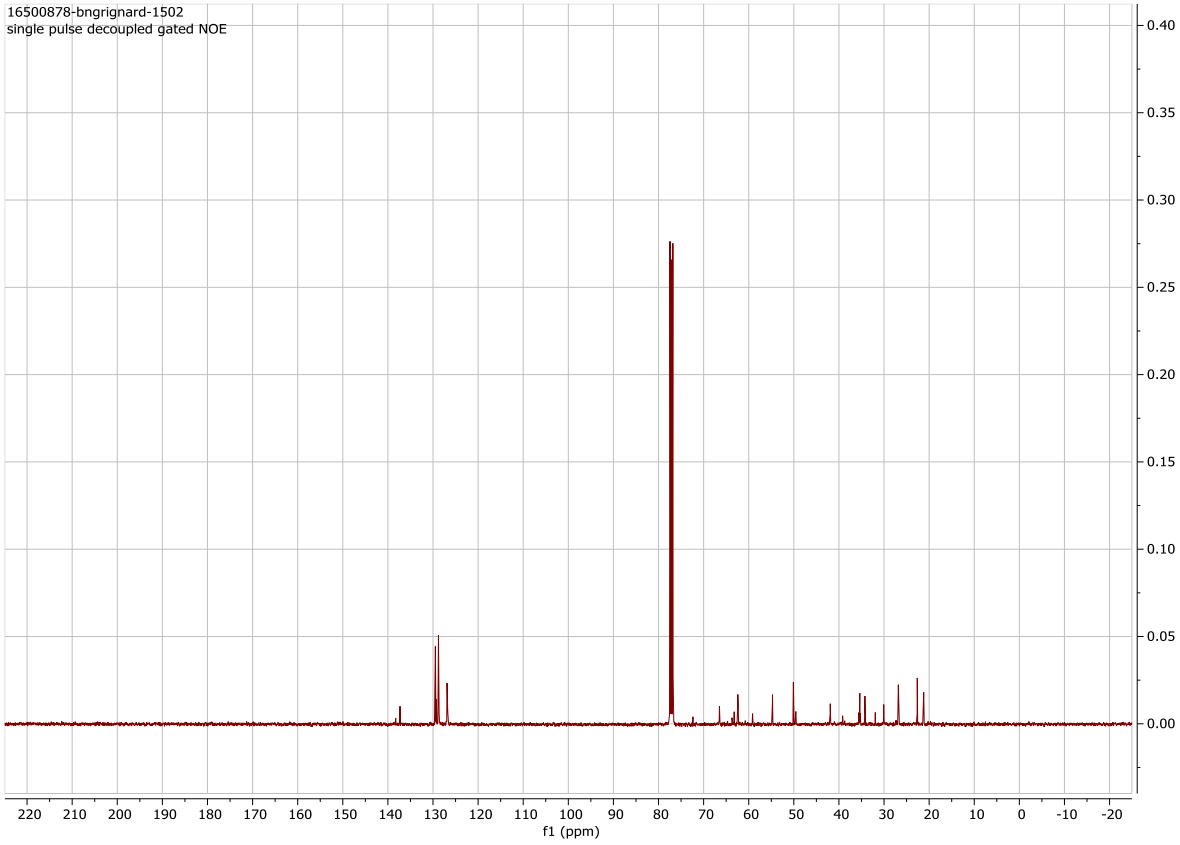



55124301-phenylrd-0312_Carbon_ft-1-1.jdf
single pulse decoupled gated NOE
JEOL
Solutions for Innovation
---- PROCESSING PARAMETERS ----
dc_balance
sexp : 2[Hz]
trapezoid : 0[%] : 0[%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-phenylrd-0312_C
Author = MichaelP
Experiment = short_carbon.jxp
Changer_Slot = 16
Sample_Id = 55124301-phenylrd-0312
Solvent = CHLOROFORM-D
Creation_Time = 5-DEC-2020 07:29:50
Revision_Time = 5-DEC-2020 08:39:46
Current_Time = 8-DEC-2020 15:34:44
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 1[s]
Recvr_Gain = 50
Temp_Get = 20.6[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.8[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
(thousandths)
60.0
50.0
40.0
30.0
20.0
10.0
0
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0

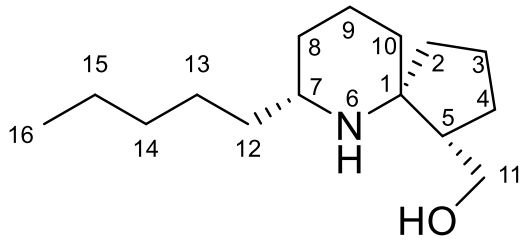
(1S*,5S*,7S*)-7-benzyl-6-azaspiro[4.5]decan-1-yl)methanol (230)

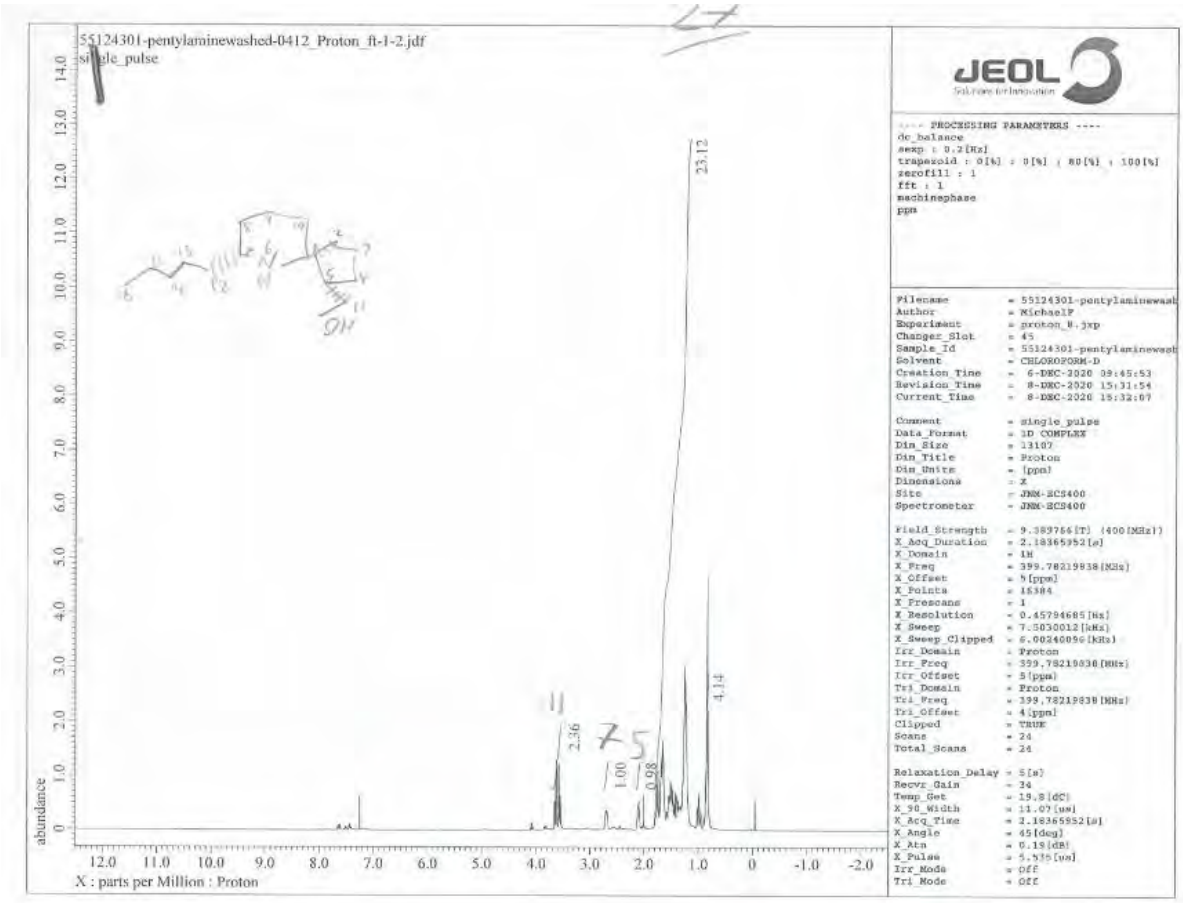


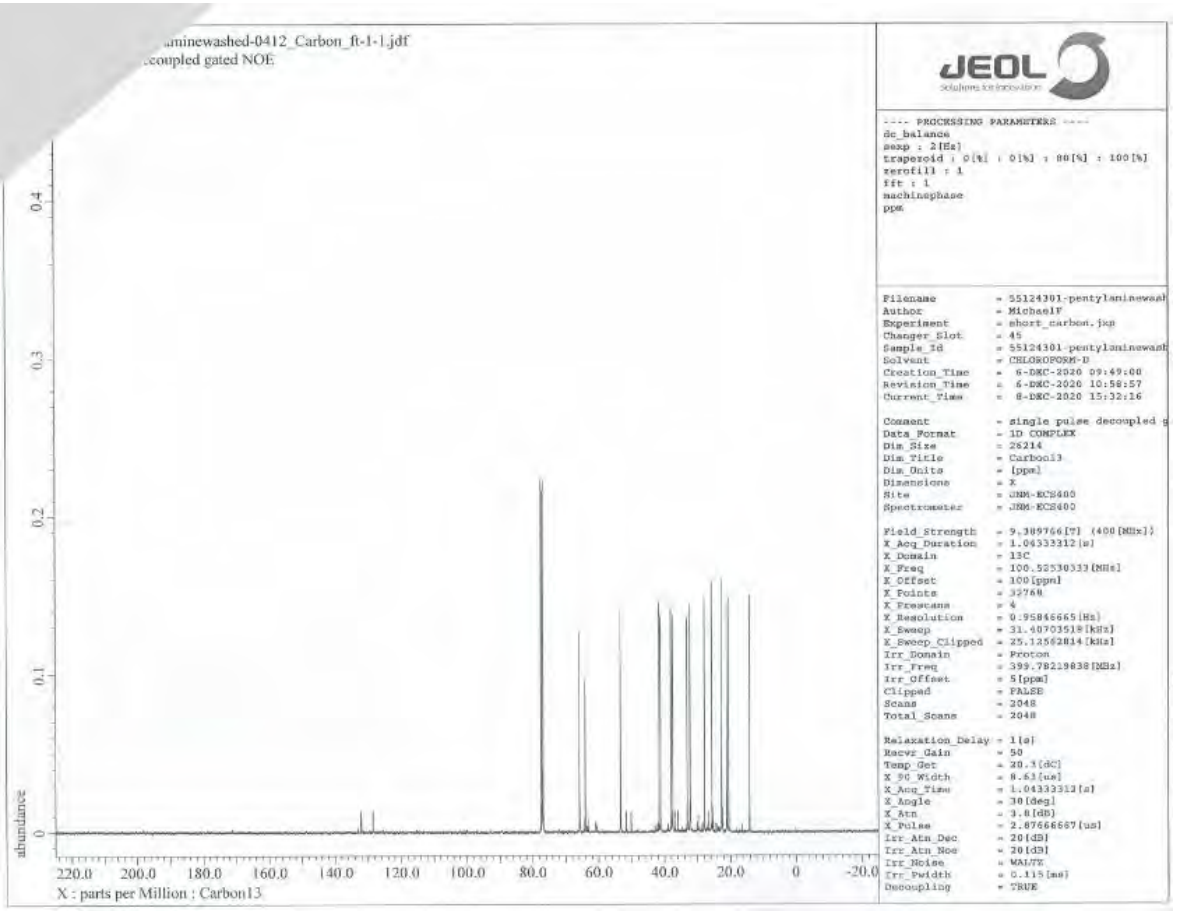



16500878-bngrignard-1502
single pulse decoupled gated NOE
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

(1S*,5S*,7S*)-7-pentyl-6-azaspiro[4.5]decan-1-yl)methanol (231)






aminewashed-0412_Carbon_ft-1-1.jdf
coupled gated NOE
JEOL
---- PROCESSING PARAMETERS ----
dc_balance
sexp : 2[Hz]
trapezoid : 0[%] : 01%] : 80[%] : 100[%]
zerofill : 1
fft : 1
machinephase
ppm
Filename = 55124301-pentylaminewash
Author = MichaelF
Experiment = short_carbon.jxp
Changer_Slot = 45
Sample_Id = 55124301-pentylaminewash
Solvent = CHLOROFORM-D
Creation_Time = 6-DEC-2020 09:49:00
Revision_Time = 6-DEC-2020 10:58:57
Current_Time = 8-DEC-2020 15:32:16
Comment = single pulse decoupled g
Data_Format = 1D COMPLEX
Dim_Size = 26214
Dim_Title = Carbon13
Dim_Units = [ppm]
Dimensions = X
Site = JNM-ECS400
Spectrometer = JNM-ECS400
Field_Strength = 9.389766[T] (400[MHz])
X_Acq_Duration = 1.04333312[s]
X_Domain = 13C
X_Freq = 100.52530333[MHz]
X_Offset = 100[ppm]
X_Points = 32768
X_Prescans = 4
X_Resolution = 0.95846665[Hz]
X_Sweep = 31.40703518[kHz]
X_Sweep_Clipped = 25.12562814[kHz]
Irr_Domain = Proton
Irr_Freq = 399.78219838[MHz]
Irr_Offset = 5[ppm]
Clipped = FALSE
Scans = 2048
Total_Scans = 2048
Relaxation_Delay = 1[s]
Recvr_Gain = 50
Temp_Get = 20.3[dC]
X_90_Width = 8.63[us]
X_Acq_Time = 1.04333312[s]
X_Angle = 30[deg]
X_Atn = 3.8[dB]
X_Pulse = 2.87666667[us]
Irr_Atn_Dec = 20[dB]
Irr_Atn_Noe = 20[dB]
Irr_Noise = WALTZ
Irr_Pwidth = 0.115[ms]
Decoupling = TRUE
abundance
0
0.1
0.2
0.3
0.4
220.0
200.0
180.0
160.0
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0
-20.0
X : parts per Million : Carbon13