Synthesis and Reactivity of Novel Pyrazolothiatriazines and Thiadiazines

A thesis submitted for the fulfilment of the degree of

Doctor of Philosophy

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Declaration

I hereby declare that this thesis was carried out at the School of Chemical and Physical Sciences at the Flinders University of South Australia. I certify that the thesis does not incorporate without acknowledgement any material previously submitted for a degree or diploma in any university. To the best of my knowledge the document does not contain any material previously published or written by another person except where acknowledgement by citation of the original publication is made in the text.

Rebecca Norman

19th January 2015

Acknowledgements

I would like to offer my sincerest gratitude to my supervisors: Assoc. Prof. Mike Perkins and Dr. Craig Francis. Mike, your Medicinal Chemistry degree was a fascinating course and I enjoyed my time as an undergraduate thoroughly. I was especially captivated by your unique style of teaching and your own interest in your field really shone through. During my postgraduate studies, your patience and immense knowledge were most valuable to me. I very much appreciate that you gave me room to work on my own ideas while still providing me with the direction I needed. Your genuine interest and engagement in my research was most encouraging, as you were always optimistic when I obtained some of those 'what the..?' results. Craig, you've been able to provide so much input into my postgraduate studies and it is hard for me to think of you as an external supervisor. Being just an e-mail or phone call away was as if you were here at Flinders. You were always keen to hear about my results and I appreciate the patience you showed when some of my progress reports came a little late. Furthermore, your guidance enabled me to stay on track and remain motivated through the more challenging stretches and you have been an excellent mentor. I attribute the level of my PhD research, publications, and experiences gained to the both of you. I have been so lucky to have had two knowledgeable, supportive and welcoming supervisors.

I wish to also extend my thanks to Dr. Andris 'Andy' Liepa who, to me, was one of my unofficial supervisors for this research with so many ideas and a high level of insight into this chemistry. Assoc. Prof. Martin Johnston, as well as acting as the 'go-to' person for the NMR machines which so many people use (and expects to work 24/7) and giving training and assistance, took the time to help me with many of my own NMR problems while also offering advice on some of my chemistry problems. Dr. Chris Sumby at Adelaide was responsible for training both Mike and myself in the use of the X-ray diffractometer at Adelaide University. Drs Jardine, Braybrook and Cosgriff have provided me with the mass spectrometry advice that has been vital to this project.

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I lastly want to take this opportunity to thank my friends and family who, despite being at the end of this section, were essential in holding me together and keeping me sane. Mr. Maynard, you got me through my undergraduate exam periods and somehow you've been able to put up with me during my crazy-thesis-writing-period as well. Thankyou for always being there for me.

Mum and Dad, stop asking when I'll get a real job.

Publications and Presentations

The following list represents publications that have resulted from research outlined in this thesis and presentations that were given at various conferences, symposia and meetings.

Publications

1. Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2013**, *66*, 1323. 'The First Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine Derivatives and their Unusual Reactions with Acylating Agents'

2. Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2015**, 68, *in press.* 'Substitution Reactions of Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-Dioxides.'

3. Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2015**, *in press.* 'Synthesis and Reactivity of Novel Pyrazolo[3,4-*e*][1,2,4]Thiadiazine Derivatives.'

4. Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2015**, *in CSIRO internal review.* 'Cleavage and Rearrangement of Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-Dioxides.'

Manuscripts in preparation:

1. Chee Ling Tong, Rebecca E. Norman, Michael V. Perkins, Kevin Jarrett, Craig E. Buckley, Xiaofei Duan, Robert N. Lamb, Colin L. Raston, to be titled: 'One-pot synthesis of PdO/SBA-15 under neutral conditions: synthesis, characterization, and catalytic properties.'

External Presentations

ICOS 20 – The RACI 20th International Conference on Organic Synthesis, ELTE convention centre (Budapest, Hungary), 29th June -4th July **2014**, 15 minute oral presentation.

The Southern Highlands Conference on Heterocyclic Chemistry, Gibraltar Hotel (Bowral, NSW), 25th-27th August **2013**, Received a Postgraduate Student Award for a 45 minute oral presentation.

The RACI Adelaide Synthetic Chemistry Symposium, Adelaide University (Adelaide, SA), 10th December **2012**, 25 minute oral presentation.

The 37th Annual Synthesis Symposium, BIO21 Institute, Melbourne University (Parkville, VIC), 7th December **2012**, Poster Presentation.

CSIRO - Materials Science and Engineering (Clayton, VIC), 30 minute oral presentations annually.

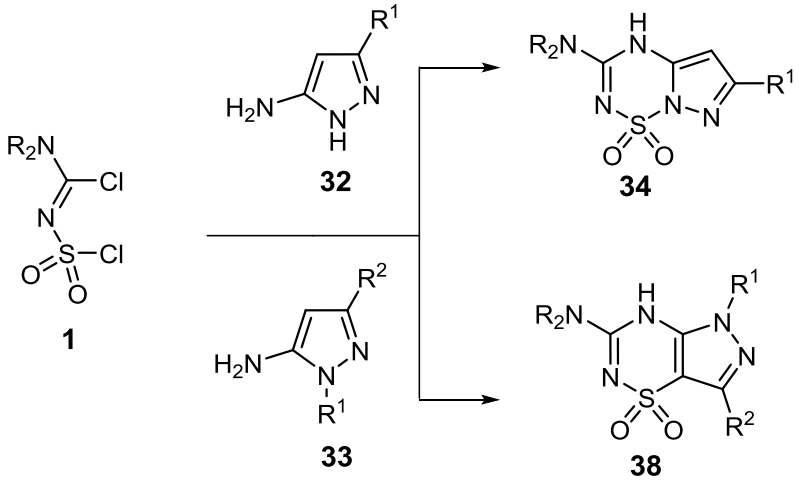
ICOS 19 – The RACI 19th International Conference on Organic Synthesis, Melbourne Convention centre (Melbourne, VIC), 1-6th July **2012**, awarded a student bursary for a poster presentation from the RACI.

CTx Annual Postgraduate Research Symposium, Monash Institute of Pharmaceutical Sciences (Parkville, VIC), 11th October **2013**, 19th October **2012**, 7th November **2011**, Poster presentations and short (5min) oral communications.

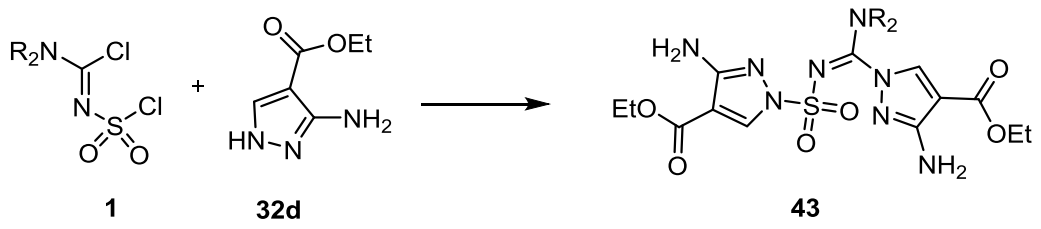
CTx Annual Retreat, Balgownie Estate (Yarra Valley, VIC), 5th December **2012**, 30th November **2011**, Poster presentations.

Abstract

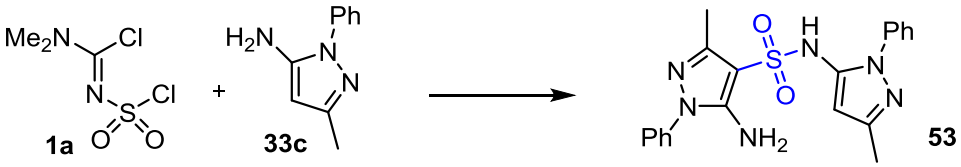
This research extends a body of work on the use of 1,3-dielectrophilic species: *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1** to generate novel, low molecular weight heterocyclic compounds. The versatility of the dichloride compounds **1** was demonstrated by a series of reactions with readily available 3-aminopyrazoles **32**. These selectively furnished representatives of the previously unreported pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system, compounds **34**. The dielectrophiles **1** were also condensed with 1-substituted 5-aminopyrazoles **33** to provide novel pyrazolo[3,4-*e*][1,2,4]thiadiazine dioxides **38** as the sole isolated products.



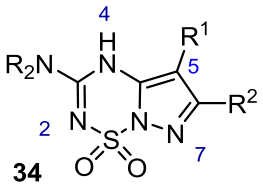
In some circumstances, the 4-ester substituted pyrazole **32d** gave rise to bis-adducts **43** (or intermediate chlorides) by reaction at the ring nitrogen N1 instead.

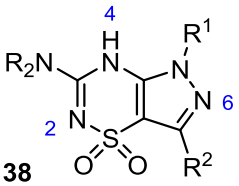


An unexpected sulfonamide product **53** was isolated in one instance from a reaction between pyrazole **33c** and dichloride compound **1a**, which appeared to have formed via rearrangement of an intermediate similar to bis-adducts **43**.

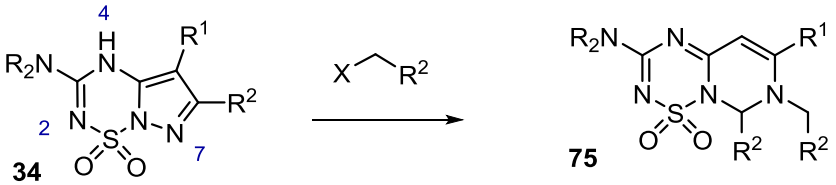


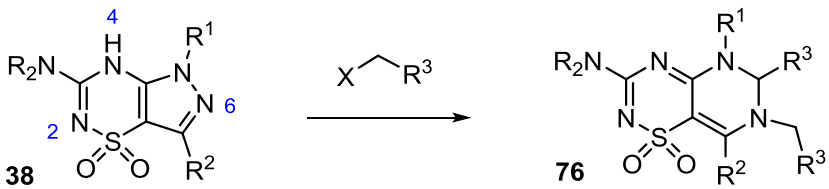
Fused pyrazole compounds **34** and **38** were shown to possess three nucleophilic NH sites which underwent a range of substitution reactions.



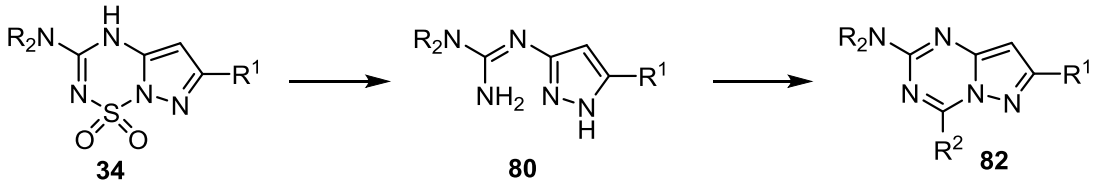


Methylation of representative substrates of compounds **34** occurred at both N4 and N7. Benzylation occurred preferentially at pyrazole nitrogen N7, but also at the pyrazole carbon C5 (when R^1=H). Alkylation with α-halo esters occurred at both N4 and N7, but the latter derivatives, under the reaction conditions, underwent a ring expansion to afford the first reported pyrimido[1,6-*b*][1,2,4,6]thiatriazine derivatives **75**. The tautomeric NH moieties of compounds **38** were reactive towards a selection of alkylating agents including benzylic halides, dimethyl sulfate and ethyl bromoacetate and alkylation occurred mostly at either thiadiazine ring nitrogens N2 or N4. A similar 'ring expansion' of the fused pyrazole ring gave the rare pyrimido[4,5-*e*][1,2,4]thiadiazine ring system **76**.

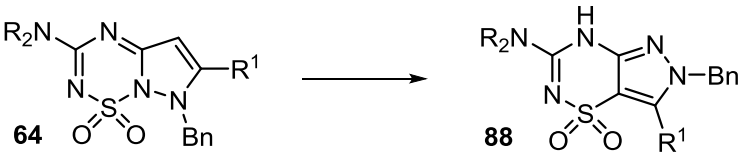




The tendency of some representative pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** to undergo ring cleavage at the sulfamide moiety was observed under a variety of conditions. Extrusion of the sulfur dioxide moiety was exploited for supplementary chemical transformations to produce pyrazolo[1,5-*b*][1,2,4]triazines **82** by the formation of guanidines **80** as intermediates. A range of cheap and commercially available electrophiles such as acid anhydrides and orthoesters were utilised to afford compounds **82**.



The pyrazolo[1,5-*b*][1,2,4,6]thaitraizine ring system **34** underwent a nucleophilic addition of C5 to *N*-acylpyridinium or *N*-acylpyridazinium species; revealed by the attempted acylation with pyridine as acyl-transfer agent. Sulfonylation, thiolation and bromination was also achieved with selective reaction at C5. Bromination or tosylation of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** afforded unstable 5-bromo or 5-tosyl derivatives, respectively, which were not synthetically useful due to insufficient stability. The susceptibility of the sulfamide moiety towards nucleophilic attack by a series of alcohols and amines was also established. An unusual rearrangement occurred upon heating fused pyrazolothiatriazine derivatives **64** affording isomeric thiadiazine dioxides **88**.



These results provided insight into the relative thermal and chemical stabilities of both systems **34** and **38**, and derivatives thereof, as well as general patterns of reactivity and selectivity of various substitution reactions.

Abbreviations

A number of common, non-standard abbreviations have been used throughout this thesis. Given here are the abbreviations followed by the standard name.

Ac_2O	Acetic anhydride
Ac	Acetyl
aq	Aqueous
Bn	Benzyl
Boc	*Tertiary*-butylcarbonyl
Boc_2O	Di-*tertiary*-butyl dicarbonate
Bu^n_4NBr	Tetra-*n*-butyl ammonium bromide
$Bu^n_4NHSO_4$	Tetra-*n*-butyl ammonium hydrogen sulfate
Bz_2O	Benzoic anhydride
CCDC	Cambridge Crystallographic Data Centre
$CDCl_3$	Deuterochloroform
CH_2Cl_2	Dichloromethane
$CHCl_3$	Chloroform
CSIRO	Commonwealth Scientific and Industrial Research Organisation
CTx	Cancer Therapeutics Cooperative Research Centre
DCC	N,N-dicyclohexyl carbodiimide
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
dec.	Decomposition point
DMAc	N,N-dimethyl acetamide
DMAP	N,N-dimethylamino pyridine
DME	Dimethoxy ethane
DMF	N,N-dimethyl formamide
DMPU	1,3-dimethyltetrahydropyrimidin-2(1*H*)-one
DMSO	Dimethylsulfoxide
DMSO-d_6	Deuterated dimethylsulfoxide
DNA	Deoxy-ribonucleic acid
EI	Electron Ionisation
ES	Electrospray
Et	Ethyl
Et_2O	Diethyl ether
Et_3N	Triethylamine
EtOAc	Ethyl acetate
EtOH	Ethanol
EWG	Electron Withdrawing Group
HCl	Hydrochloric acid
HETCOR	Heteronuclear Correlation Spectroscopy

HMBC	Heteronuclear Multiple-Bond Correlation Spectroscopy
HMQC	Heteronuclear Single-Quantum Correlation Spectroscopy
HPLC	High Pressure Liquid Chromatography
HRMS	High Resolution Mass Spectrometry
$^{i}Pr_2NEt$	N,N'-diisopropylethylamine
J	Coupling constant (Hz)
m.p.	Melting point
m/z	Mass to charge ratio
MeCN	Acetonitrile
MeOH	Methanol
MeOTs	Methyl p-toluenesulfonate
MS	Mass spectrometry
MsOH	Methane sulfonic acid
MWI	Microwave irradiation
NBS	N-bromosuccinimide
*n*BuLi	*n*Butyllithium
NH_4OH	Aqueous ammonia
NIS	N-Iodosuccinimide
NMR	Nuclear Magnetic Resonance
nOe	Nuclear Overhauser Effect
NOESY	Nuclear Overhauser Effect Correlation Spectroscopy
[O]	Oxidation
ORTEP	Oak-Ridge Thermal Ellipsoid Plot
PEPPSI-^{i}pr	1,3-Bis(2,6-Diisopropylphenyl)imidazol-2-ylidene](3-chloropyridyl)palladium(II) dichloride
Ph	Phenyl
PMB	Para-methoxybenzyl
PPh_3	Triphenylphosphine
ppm	Parts per million
p-TsOH	Para-toluenesulfonic acid
QSAR	Quantitative Structure Activity Relationship
R_F	Retardation factor
RNA	Ribonucleic acid
rt	Room temperature
t-BuOH	Tertiary-butanol
t-BuOK	Potassium tertiary-butoxide
TFA	Trifluroacetic acid
TFAA	Trifluroacetic anhydride
THF	Tetrahydrofuran
TLC	Thin Layer Chromatography
TsCl	p-toluene sulfonyl chloride
UV	Ultraviolet
X4	Mixed hexanes

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CHAPTER 1: GENERAL INTRODUCTION

1.1 The Importance of Heterocycles in Drug Discovery and Industry

The majority of pharmaceuticals and biologically active agrochemicals contain a heterocyclic ring; hence the vitality of heterocyclic chemistry is of vast practical and theoretical significance. The scientific discipline of medicinal chemistry has advanced to introduce various techniques in order to accelerate the drug discovery process; and these include combinatorial chemistry, microwave assisted synthesis, and high-throughput/parallel synthesis and purification (such as HPLC). The screening of chemical compound libraries for a desired biological activity plays an essential role in the discovery of new pharmaceuticals and represents a fundamental research method in medicinal chemistry to find and evaluate active compounds. The proportion of potential 'drug-like' molecules meeting Lipinski's rule of five[1] which have already been synthesized only represents a small fraction of the possible structures which meet these predicted physio-chemical properties. The most challenging aspect of drug discovery is arguably the selection of new molecules from this endless pool of possibilities which have the potential to be biologically active, but are able to be easily synthesised and modified. An important requirement is also that these compounds need to be free of patent competition, which encourages the exploration of new chemical space.

Chemists' historical exploration of chemical space has been arguably disproportionate and asymmetric. Approximately half of all known compounds are based on just 0.25% of the known molecular scaffolds (**Figure 1**).[2] Emphasis has been placed on the most synthetically accessible scaffolds and there appears to be a lack of synthetic exploration of less common candidates. It is for this reason that a systematic exploration of chemical space is needed to allow scaffolds of small molecules to be varied combinatorially. A review by Dow *et al*[2] outlined a "build couple pair approach," specifically with dinucleophilic pairing reactions. These involve condensations between two bifunctional building blocks where the initial reaction is chemoselective, followed

by a subsequent intramolecular reaction between the remaining functional groups to form a ring (**Figure 2**).

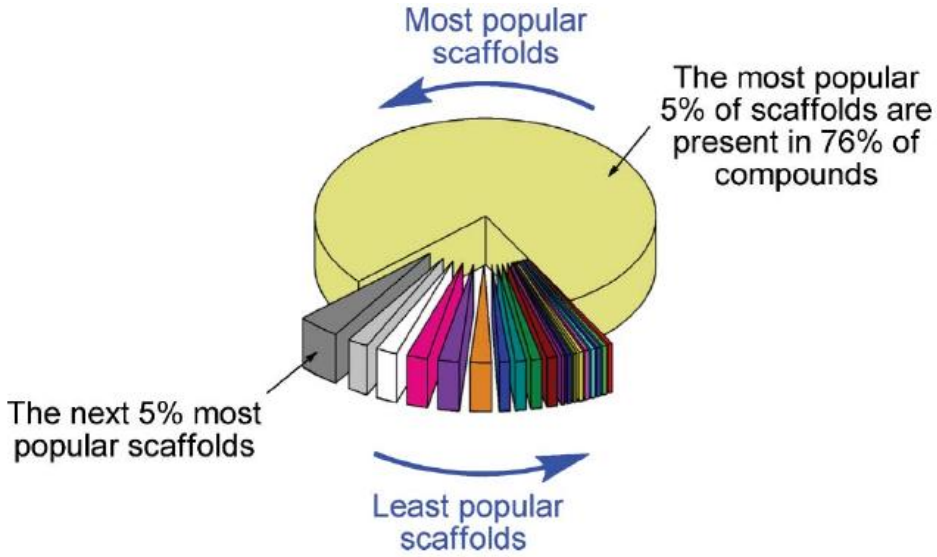

Fig 1. The 24,282,284 cyclic compounds known in the CAS registry in 2008 grouped from the most popular to the least popular scaffolds.[2,3]

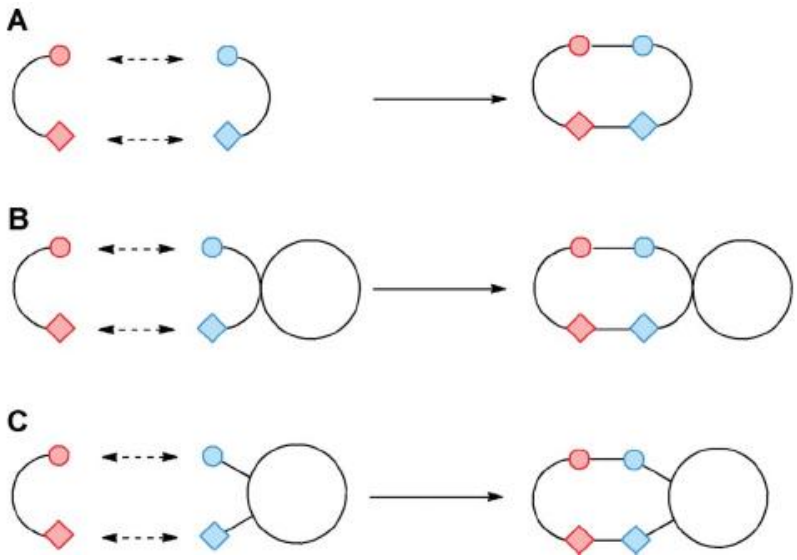

Fig 2. The "build-couple-pair" approach of drug discovery from bifunctional building blocks using bidentate pairing reactions. Panel A represents monocyclic scaffolds whereas panel B and panel C illustrate the use of cyclic building blocks to afford spirocyclic or fused scaffolds respectively.

The quest for applications of heterocycles as pharmaceutical and industrial compounds means considerable research effort continues to be directed towards new synthetic pathways for novel heterocyclic structures.[4] New ring structures supply scaffolds for the exploration of uncharted chemical space, and can enable identification of the chemical, biological, and physical behaviour of novel systems.[5] Low molecular weight compounds in particular have the amenability for fast and extensive class expansion. Identifying and optimising methods of synthesis, isolation and characterisation of novel heterocyclic systems is therefore an important area of synthetic organic chemistry.

Heteroaromatic rings also serve as bioisosteres of several structures, including phenyl rings and carboxylic acid derivatives for example, and sometimes deliver greater pharmacological activity to the resulting compounds. A commendable review by Dalvie *et al*[6] provides an examination of the biotransformation pathways of various heterocyclic systems, including fused pyrazoles which this thesis will encompass. Here it has been documented from Structure-Metabolism Relationship (SMR) studies that the incorporation of heteroatoms into an aromatic ring influences not only the biochemical and chemical activity, but also a drug candidate's metabolism.[6] A more recent review by St. Jean and Fotsch[7] summarises various approaches to improving the metabolic stability of the different classes of heterocycles.

The discovery of biologically or industrially significant molecules is most certainly shaped by their synthetic or biosynthetic accessibility. Synthetic organic chemistry therefore plays a key role in the exploration of chemical space which is vital to the discovery and application of novel cyclic compounds. The utilisation of ambiphile pairing reactions to form novel or uncommon heterocyclic ring systems is an attractive prospect due to the narrow range of previously explored heterocyclic scaffolds in drug-like molecules (**Figure 3**).

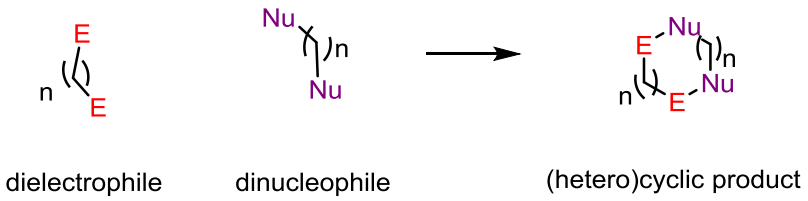

Fig 3. The synthesis of cyclic or heterocyclic molecules from reactive bidentate precursors.

To this end, the unusual dielectrophilic structures: *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1** have been investigated as bidentate electrophiles for the synthesis of a diversity of structurally unique heterocyclic ring systems (**Figure 4**).

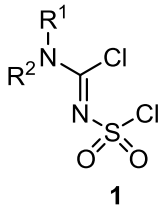

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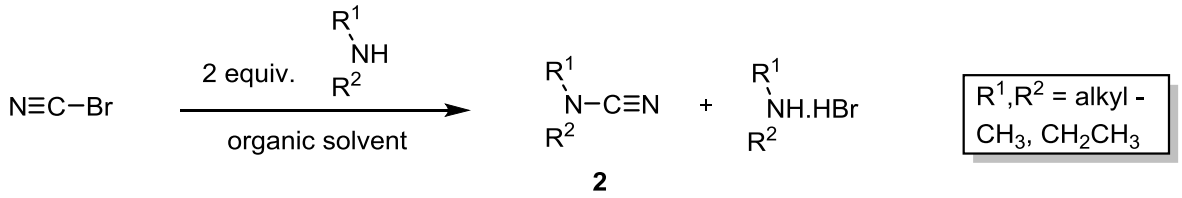
Fig 4. *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1**.

1.2 Synthesis of *N*,*N*-Dialkyl *N*'-Chlorosulfonyl Chloroformamidines

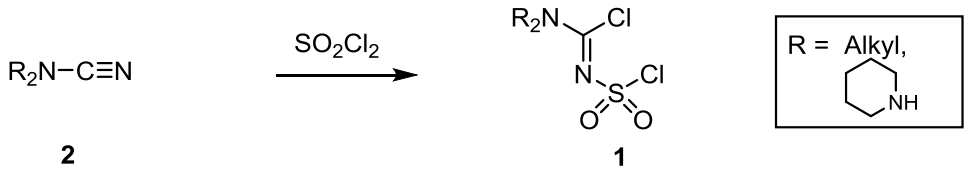
In 1953 Garbrecht and Herbst prepared dialkylcyanamides **2** in a single step from the reaction of cyanogen bromide with various secondary amines (**Scheme 1**).[8] A variety of alkyl substituents were provided by alternative symmetrical and asymmetrical amine starting materials.



Scheme 1: The reaction of cyanogen bromide with secondary amines.

Substituted cyanamides are very reactive compounds, and a number of their derivatives are synthetically valuable for research and industry alike. The most recent review of cyanamides was published in 2004 by D. Nekrasov which discusses the known methods for preparation, typical transformations, and some aspects of practical applications of these compounds in medicine, industry and agriculture.[9]

In 1973, Norbert Schindler reported the reactions of some symmetrical dialkylcyanamides **1** with various sulphur and phosphorous halides.[10] Markovskii and coworkers had also reported the reaction between dialkyl cyanamides and either sulfuryl chloride or thionyl chloride.[11] Treatment of dialkylcyanamides **2** with sulfuryl chloride furnished the *N*,*N*-dialkyl *N*'-chlorosulfonylchloroformamidines **1** in **Scheme 2**.



Scheme 2: The reaction of dialkylcyanamides **1** with sulfuryl chloride to afford dichlorides **2**.

These dichlorides **1** exhibited more signals than expected in the NMR in that two separate environments were reported for the dialkylamino substituents, although the R groups were the same.[10,12] Broadening of proton and carbon signals of α- and β-positions of the secondary amine entity of substituted amidines is often reported due to a magnetic non-equivalence of the two R groups.[13,14] The rate constant for the exchange of protons flanking these chains is reduced in this case by the double bond character the dialkyl amino substituent possesses, meaning that two signals are seen on the NMR

timescale. It is not surprising that the sulfonyl formamidine compounds **1** exhibit similar behaviour. Trisubstituted formamidines such as these could present two types of isomerism (**Figure 5**) and the hindered rotation about the C-NR_2 bond arises due to resonance effects.[15]

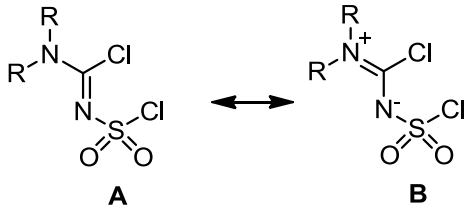


Fig 5. Resonance forms of *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **2**

The contribution of forms A and B means that the dialkylamino bond exhibits partial double bond character, and the formamide group is approximately planar.[15] A syn isomer has been proposed (**Figure 6**) which would affect dichlorides containing two different alkyl chains on the secondary amine; however this was considered unlikely in the case of sulfonyl formamidines **1** since space filling molecular models have shown the syn isomer to be sterically crowded.[14]

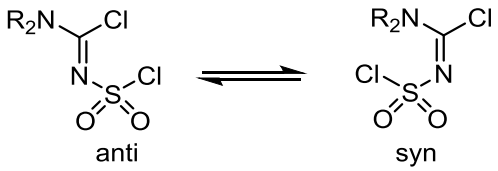
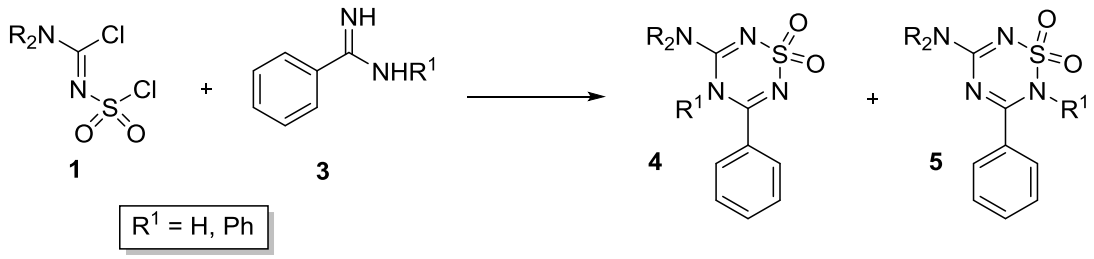


Fig 6. The syn and anti tautomerism of *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1**

Compounds **1** possess two reactive electrophilic sites, one at the amidinyl chloride and the other at the sulfamoyl chloride moiety. Early studies on the reactions of the dielectrophile **1** with reagents such as amines, water and alcohols validated their reactive 1,3-dielectrophilic nature.[12,16] These compounds could therefore function as building blocks for synthesis of various ring systems by the action of bidentate nucleophiles.

1.3 *N,N*-dialkyl *N'*-chlorosulfonyl chloroformamidines as ambident dielectrophilic reagents

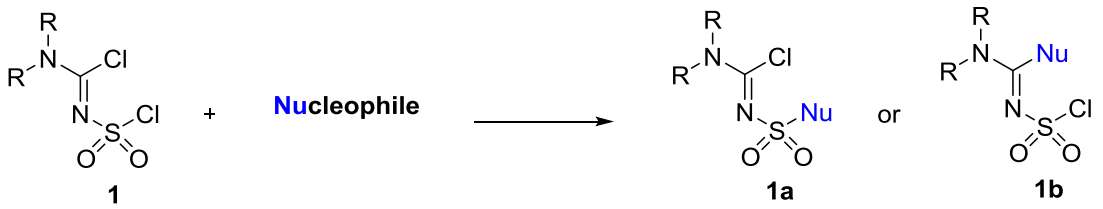
It was hypothesised that dichlorides **1** may react with bidentate nucleophiles to generate various ring structures. Markovskii *et al* demonstrated this via the reaction of compounds **1** with benzamidines **3** to afford novel [1,2,4,6]thiatriazines **4** and **5**.[12] These condensations took place readily under thermal effect with satisfactory yields, generating hydrochloric acid as a by-product which was removed in the presence of amine base (**Scheme 3**).



Scheme 3: The reaction of dichlorides **1** with benzamides **3** to provide thiatriazines **4** and **5**.

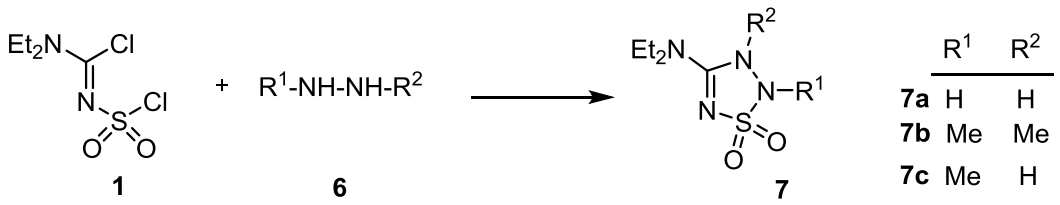
The favourable formation of compounds **4** over the tautomeric form **5** (when R=H) was proposed due to the acidic characteristics of the SO_2NH group. However when R was a bulky phenyl substituent, the group proposed the formation of isomer **5** based on the fact that phenylbenzamidine was known to acylate preferentially at the substituted nitrogen,[17] and the stronger electrophilic site of the acid chlorides **1** was believed to be at the sulfamoyl group.[12]

The relative reactivity of the two electrophilic sites in compounds **1** was disputed in early research. Some literature suggested (on the basis of infrared spectroscopy) a greater susceptibility of the sulfamoyl chloride moiety relative to the amidinyl chloride, towards nucleophilic attack by secondary amines such as morpholine and piperidine to generate sulfamides **1a** (**Scheme 4**).[12] On the other hand, an opposite regioselectivity in the reaction of **1** with secondary amines to furnish isomers **1b** has also been proposed from the analysis of fragmentation from mass spectrometry experiments.[18]



Scheme 4: The two possible products from reaction of one equivalent of nucleophile with dichlorides **1**.

A report by Knollmüller and Kosma[16] examined the reaction of *N*,*N*-diethyl *N*'-chlorosulfonyl chloroformamidines (**1**) with hydrazine, *N*-methylhydrazine and 1,2-dimethylhydrazine to furnish [1,2,3,5]thiatriazole derivatives **7** (**Scheme 5**).



Scheme 5: The reaction of a single dichloride **1** with a small set of substituted hydrazines **6**.

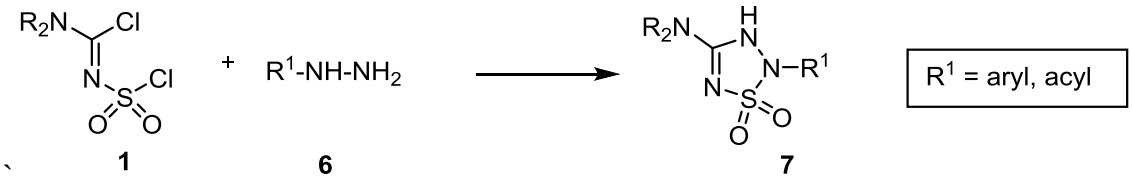
Unambiguous structural elucidation of isomers **7c** and **7d** proved difficult due to the similarities in NMR and IR spectra. The structures were assigned based on the literature precedent for chemical shifts of a sulfamide NH which were expected to be higher than the NH group further from the sulphur atom.[16] It was also suggested that the isomer **7c** was the more stable product, and that the more hindered nitrogen reacted first at the sulfamide group based on earlier evaluations from Markovskii and coworkers.[12]

Reports on the chemistry of the dichloro compounds **1** did not appear again in the literature for the greater part of two decades. The few studies that had been published in the area during this period did not include the preparation of derivatives from dinucleophiles. Furthermore, structural assignments were supported only by mass spectrometry, IR spectrometry and NMR spectrometry which were the most readily

available characterisation techniques at the time. It could not be assumed that the regioselectivity was correctly reported, especially given that the possible isomers formed lacked informative NMR and IR signals. It also appeared that the potential offered by such a versatile intermediate that would allow opportunities for the synthesis of a variety of new or uncommon heterocyclic ring systems had been overlooked.

1.4 The history of research on *N,N* – dialkyl *N'*-chlorosulfonyl chloroformamidines as synthons for novel ring systems

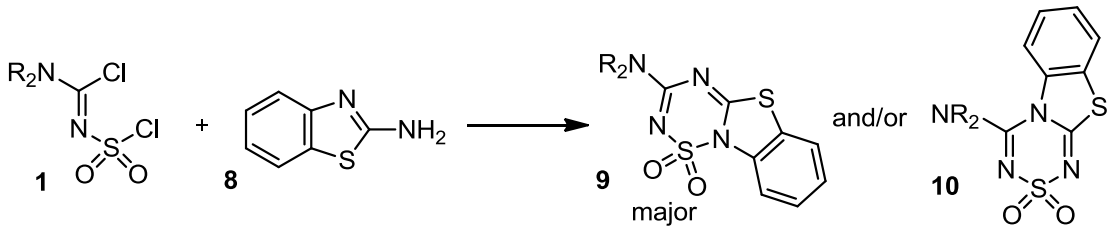
The production of novel, low molecular weight heterocycles from dichlorides **1** began as an area of research more than a decade ago at the Commonwealth Scientific and Industrial Research Organisation (CSIRO) in Australia. In an expansion on the work by Knollmüller and Kosma, reactions of a range of hydrazine derivatives **6** reacting as 1,2-dinucleophiles with compounds **1** in the presence of mild base was published by Liepa and colleagues to afford [1,2,3,5]thiatriazoles **7** regioselectively in high yields.[19] The structures of compounds **7** were unequivocally elucidated by X-ray crystallography of some representatives (**Scheme 6**).[19]



Scheme 6: Regioselective reaction of **1** with hydrazine derivatives **6** to selectively produce a single isomer **7**.

This research confirmed that the less hindered nitrogen atom of the hydrazines **6** reacted with the amidine carbon of **1**, and the other nitrogen atom cyclised at the sulfamoyl group. A higher electrophilicity of the amidinyl carbon in **1** was proposed initially[19] based on the fact that, in general, monosubstituted acyl and aryl hydrazines react with most electrophiles at the unsubstituted amino group.[20,21]

The dichlorides **1** were also treated with some 2-aminothiazole derivatives, for example 2-benzothiazoles **8,** as representative 1,3-dinucleophiles, to form the fused [1,2,4,6] thiatriazines **9** and **10** in **Scheme 7**.[22]



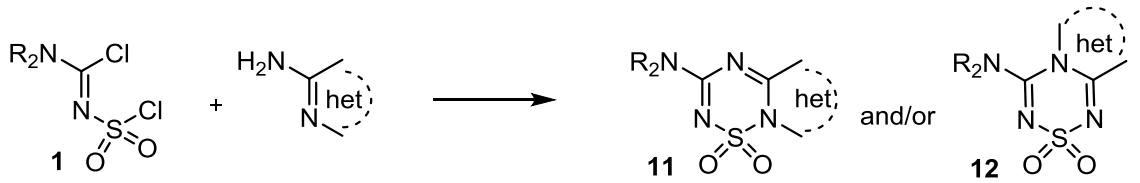
Scheme 7: The reaction of dichlorides **1** with 2-aminobenzothiazoles **8** to form the isomeric fused [1,2,4,6]thiatriazines **9** and **10**.

In these examples, the major product was the fused thiazolo[3,2-*b*][1,2,4,6]thiatriazine **9**. This outcome implied that the greater nucleophilicity of the exocyclic amine results in selective reaction at the amidinyl carbon first, and that the selectivity may also be influenced by steric interference from the dialkylamino group.[22]

The regioselectivity observed in these cases reinforced the hypothesis that the amidinyl carbon in **1** is the more reactive electrophile.[22] It is clear, however, that reactions with **1** are governed by steric factors as well as reactivity.

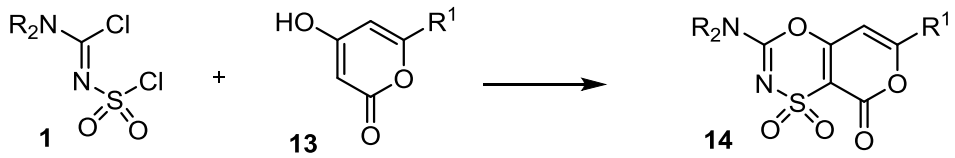
It became clear to the CSIRO scientists that the dichlorides **1** represented an unusual C-N-S building block which offered potential for reactions with a variety of readily available dinucleophiles to afford a suite of new or uncommon heterocyclic ring systems. A series of studies was commenced, aimed at synthesising a variety of novel systems derived from treatment of the dichloride **1** with diverse classes of dinucleophilic systems.

Some representative 1,3-dinucleophilic 2-amino-1-azaheterocycle ring systems reacted with dichloride **1** substrates to afford fused [1,2,4,6]thiatriazines **11** and **12** (**Scheme 8**).[23,24]



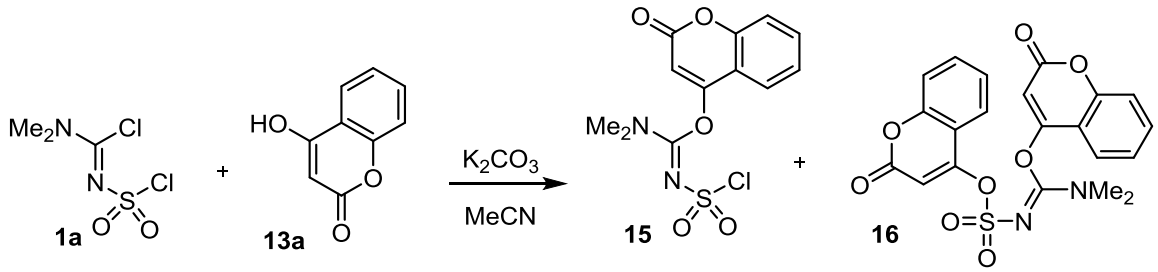
Scheme 8: Reactions of 2-amino-1-azaheterocycles as 1,3-N-C-N dinucleophiles with the dichloro compound **1**.

The group at CSIRO wished to expand the range of substrates away from nitrogen nucleophiles to further examine the versatility of these reactions. A series of 4-hydroxy-2-pyrones **13** reacted with examples of dichlorides **1** to afford a pyrano[3,4-*e*][1,4,3]oxathiazin-8-one derivatives **14** as the sole isolated products (**Scheme 9**).[25]



Scheme 9: Fused pyrano[3,4-*e*][1,4,3]oxathiazin-8-ones **14** obtained from reaction of 4-hydroxy-2-pyrones **13** with dichlorides **1**.

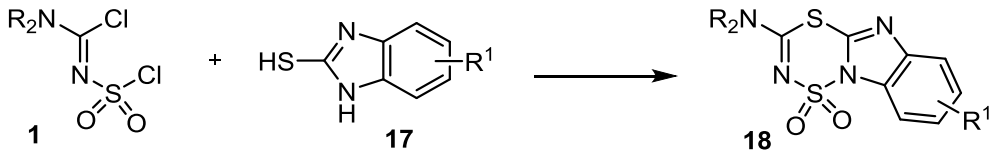
4-Hydroxycoumarin (**13a**) was treated with dichloride **1a** which lead to the formation of the coumarin-carbamimidate **15** and, in one instance, the bis-adduct **16** from nucleophilic attack of the hydroxyl group at both the sulfur and amidinyl carbon atoms of **1** (**Scheme 10**).



Scheme 10: The formation of carbamimidate **15** and bis-adduct **16** from hydroxycoumarin **13a**.

It appeared that the carbonate base promoted the nucleophilicity of the hydroxyl group to such an extent that a second coumarin reacts before the pyrone moiety cyclises. The particular sulfamoyl chloride **15** formed were apparently incapable of cyclising, or slow to cyclise, to the oxathiazine **14** material. The group suggested that the formation of compounds **14** was possible because these reactions proceeded through reaction of the C3 atom on the pyrone ring at the sulfur initially, to then ring close by means of the hydroxyl group.

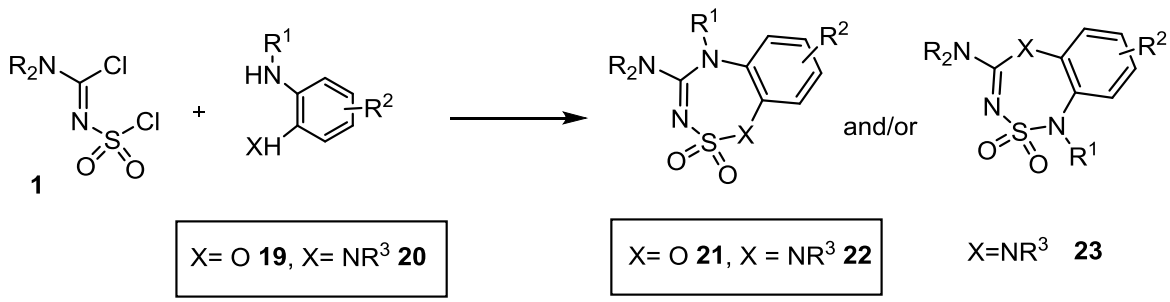
2-Mercapto-1*H*-azaheterocycles were next explored as examples of 1,3-SCN dinucleophilic systems. A set of 2-mercaptobenzimidazole substrates **17** reacted with the dichlorides **1** to yield fused [1,4,2,6]dithiadiazines **18** as the only isolated products (**Scheme 11**).[26]



Scheme 11: Reaction of 2-mercaptobenzimidazoles **17** with dichloro compounds **1**.

These results were presumed to be a consequence of the greater nucleophilicity of the SH moiety in comparison to the imidazole NH groups, and hence dominant reactivity of the S-nucleophile was observed at the amidinyl carbon. The formation of a $S(O_2)$-S bond from reaction of the SH group at the sulfamoyl chloride to furnish a dithiadiazine ring was not favoured, and there are currently no reports of the [1,2,3,5]-dithiadiazine ring system in the literature.

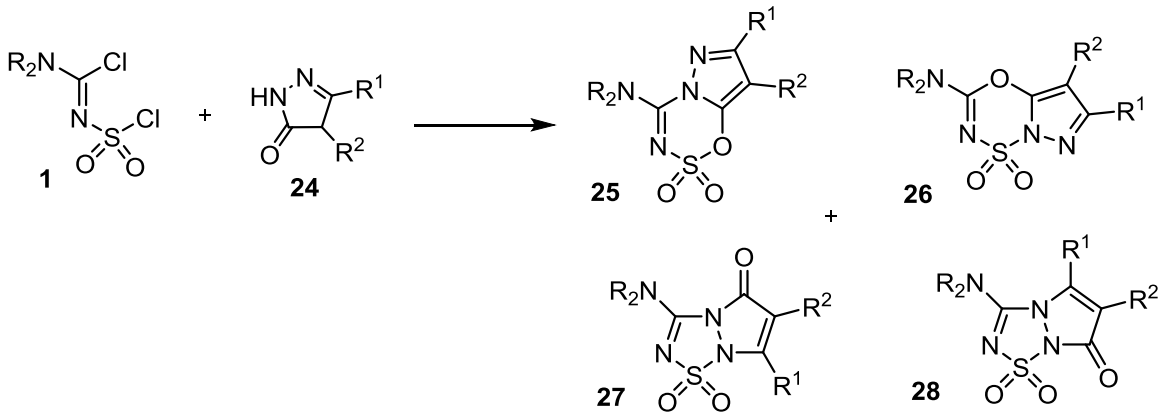
Seven-membered heterocycles were then pursued with the application of 1,4-dinucleophilic systems. The reaction of dichlorides **1** with 2-aminophenols **19** generated benzo[f][1,2,3,5]oxathiadiazepines **21**, and reaction with 1,2-diaminobenzenes **20** provided benzo[e][1,2,4,7]thiatriazepines **22** and occasionally benzo[e][1,2,4,7]thiatriazepines **23** (**Scheme 12**).[27]



Scheme 12: Seven membered ring systems formed via reaction of electrophile **1** with 2-aminophenols **19** and 1,2-diaminobenzenes **20**.

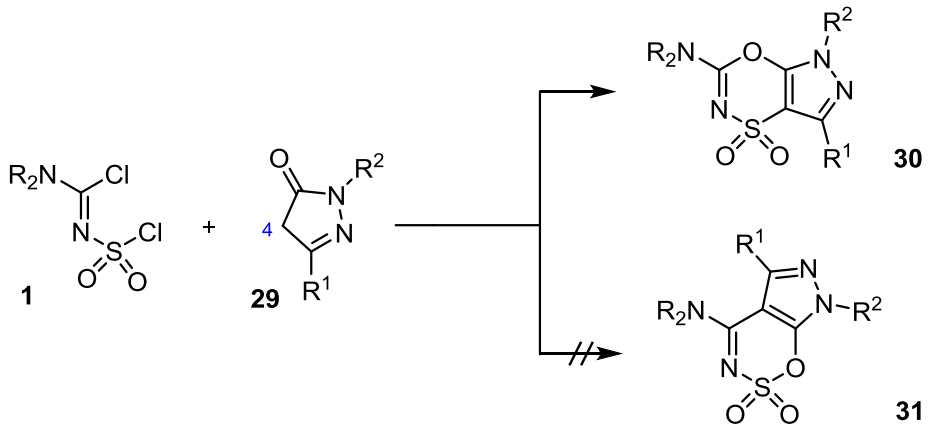
The structures of some representative compounds of each of these ring systems (**7, 9-12, 14-16, 18,** and **21-23**) were confirmed by X-ray crystallography to circumvent ambiguity. The remaining compounds were then assigned based on similarities in the NMR spectral data, and physical properties such as relative solubilities and mobility during TLC analyses. The investigations which spanned over seven papers[19,22–27] established the stronger electrophilic nature of the amidinyl carbon in comparison to the sulfonyl group with the nucleophiles and conditions employed. While the amidinyl moiety appears to be more reactive in general, the regioselectivity of condensation reactions is also likely to be influenced by compounding factors. These include steric and electronic demands of the nucleophile; and by alterations to the reaction conditions such as solvent, base or temperature.

Prior to this PhD study, the reactions between dichlorides **1** and pyrazol-3-ones **24**, which can act as either 1,2 or 1,3-dinucleophiles, were studied.[28] Four different isomeric products **25-28** were possible from this reaction, and at least one example of each was obtained from various reaction conditions and different substitution patterns on the pyrazol-3-one starting material. Under all conditions, the fused pyrazolo[5,1-*b*][1,4,3,5]oxathiadiazine **26** were isolated as the major product; however it was also observed that regioselectivity could be adjusted to a limited extent by varying the reaction conditions (**Scheme 13**).[28] Examples of all four possible ring systems were eventually isolated. Compounds **25** and **26** each represented a previously unreported ring system.



Scheme 13: Four isomeric products obtained by reaction of dielectrophiles **1** with various pyrazol-3-ones **24.**

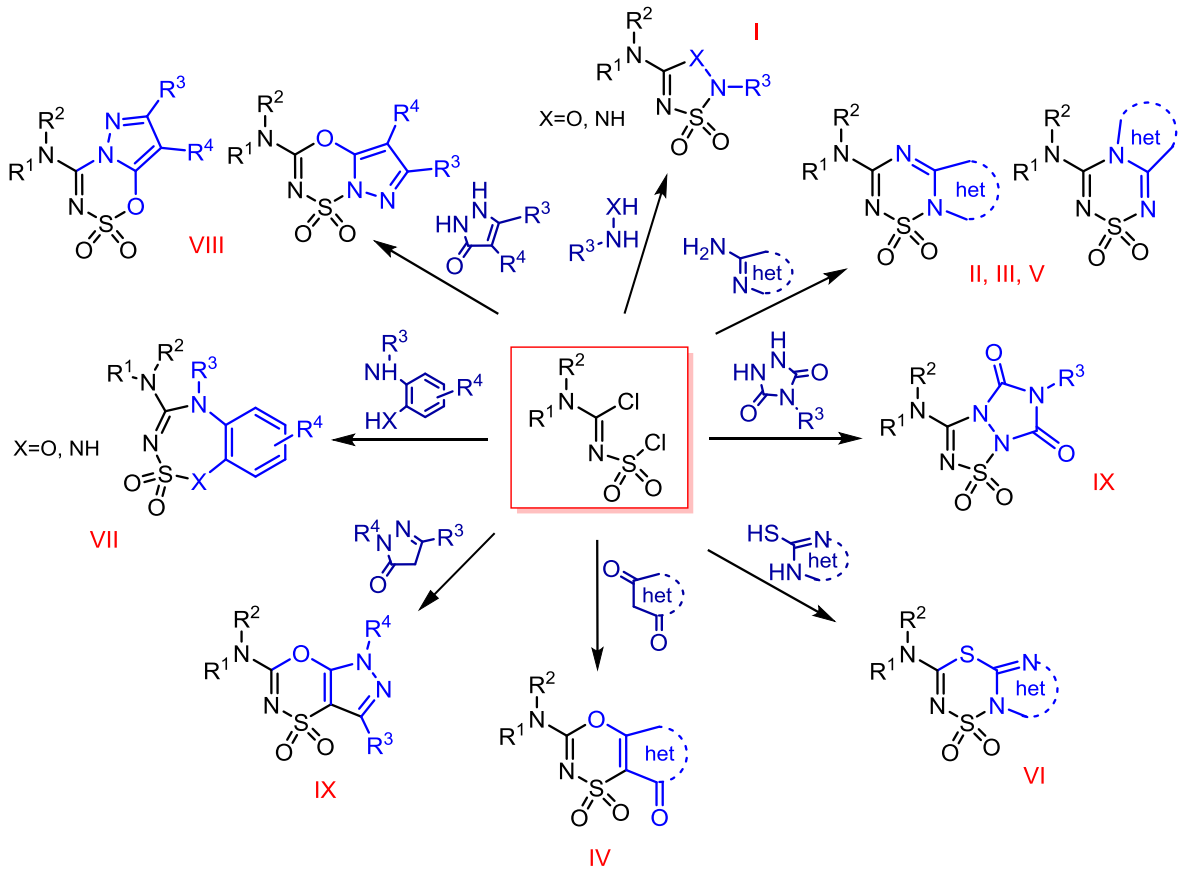
Continuing from this work, 1-substituted pyrazol-5-ones **29** were utilised as a means of removing one nucleophilic nitrogen, thus restricting the tautomeric forms available to the pyrazolone starting material.[29] The only isolated products in this case resulted from reaction of the oxygen atom at the amidinyl chloride and C4 reacting at the sulfonyl chloride moiety to give the fused pyrazolo[4,3-*e*][1,4,3]oxathiazines **30.** The pyrazolo[4,3-*e*][1,2,3]oxathiazines **31** were not observed (**Scheme 14**).



Scheme 14: Reactions of N-substituted-pyrazol-5-ones **29** with dichloro compounds **1**.

As described above, dichloro compounds **1** gave regioselective access to a variety of new or uncommon, stable heterocyclic ring systems of possible biological or pharmacological significance. The results from the research group at CSIRO were

published over a series of nine papers[19,22–29] and are summarised in **Scheme 15**. This is an ongoing project at CSIRO which has also involved collaborations with the Flinders University of South Australia and Monash University in Victoria.



Scheme 15: The range of heterocyclic ring products afforded from dichlorides **1**, with the number of the corresponding paper represented by Roman Numerals.

These new and unusual ring systems have the potential for use as a scaffold modification (known as "scaffold-hopping") from structurally similar known active/drug templates. Lower molecular weight examples could be employed in fragment based drug discovery investigations. These heteroatom-rich systems are likely to occupy less explored regions of chemical space in terms of properties such as polar surface area, number of hydrogen-bond acceptors and solubility profile when compared to the majority of compounds found in conventional screening libraries.

While the inherent chemical novelty of these products had much to commend them as candidates for biological testing, in most cases broader evaluation would be constrained by a lack of reactive sites where a diversity of substituents could be readily introduced and hence, opportunities for structural variations were limited. Accordingly, the potential for extending this synthetic methodology to generate related heterocycles bearing an NH group was evaluated, which would then allow subsequent substitution and diversification within the pool of available test compounds. Employing 3-aminopyrazoles in place of *N*-unsubstituted pyrazol-3-ones in reactions with *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1** appeared to offer the desired product versatility.

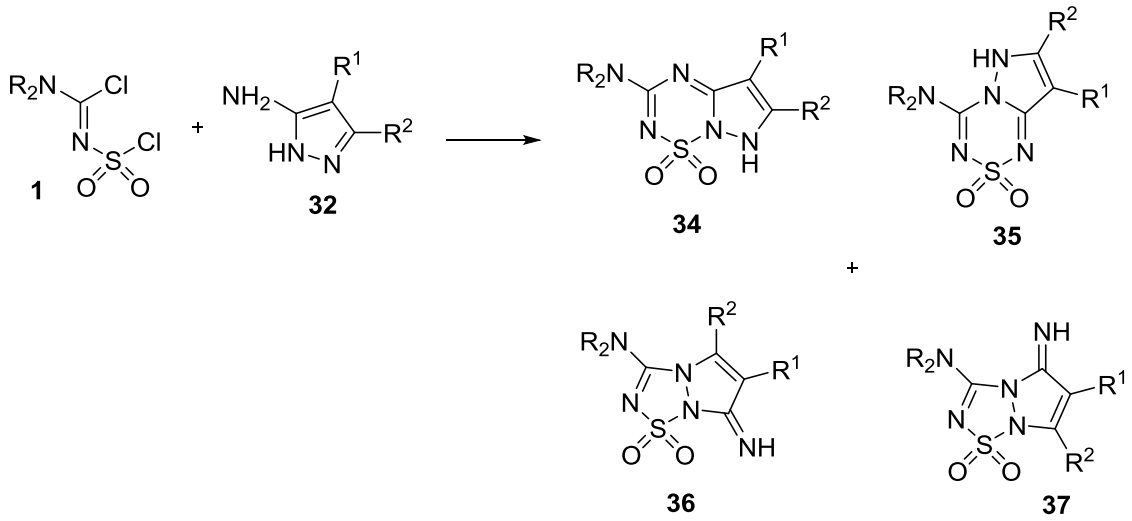
1.5 Project aims

As mentioned previously, the program at CSIRO have recently been pursuing the generation of new or uncommon heterocyclic systems (from dichlorides **1**) bearing one or more centres for potential substitution reactions, thus enabling the ready production of a small, focused screening library based on each ring system. The focus of the research described in this thesis is directed at evaluating the potential of dinucleophilic substituted pyrazoles to produce a further suite of novel heterocyclic systems. A systematic "diversity-oriented synthesis" approach[2] was taken in order to efficiently generate multiple ring systems.

The aim of this research is to explore a small set of examples of novel ring systems afforded from the treatment of pyrazoles **32** and **33** (**Figure 7**) with dichlorides **1**.

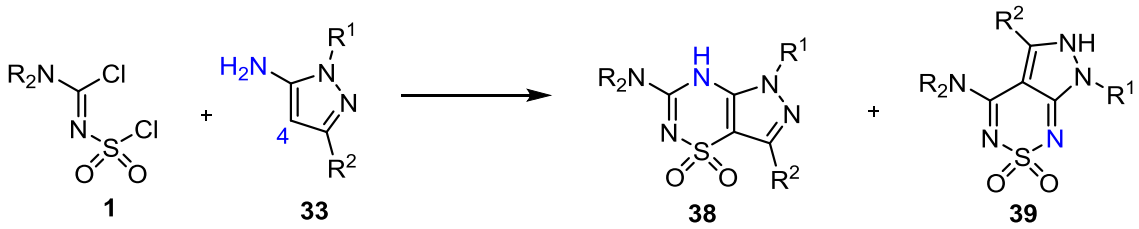
Fig 7. The 3-aminopyrazole **32** system and 1-substituted 5-amino pyrazoles **33**.

Each of these aminopyrazole derivatives contains multiple reactive sites (which are outlined in more detail in Chapter 2 and Chapter 3) and thus has the potential to afford isomeric products (**Scheme 16**). The 3-aminopyrazole ring system **32** contains three nucleophilic atoms and therefore dinucleophilic substitution could lead to four possible bicyclic ring structures: the six-membered fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **34** and pyrazolo[5,1-*c*][1,2,4,6]thiatriazine **35**; and also the five membered fused pyrazolo[1,2-*b*][1,2,3,5]thiatriazole **36** and pyrazolo[1,2-*b*][1,2,3,5]thiatriazole **37**.



Scheme 16: Possible isomeric products from the reaction of dichloro compound **1** with 3-aminopyrazoles **32**.

The reaction of 5-aminopyrazoles **33** with dichlorides **1** could give rise to two possible bicylic ring products: the fused pyrazolo[3,4-*e*][1,2,5]thiadiazines **38** and the fused pyrazolo[3,4-*c*][1,2,6]thiadiazines **39** (**Scheme 17**).



Scheme 17: The potential reaction of 1-substituted 5-amino pyrazoles **33** with dichlorides **1**.

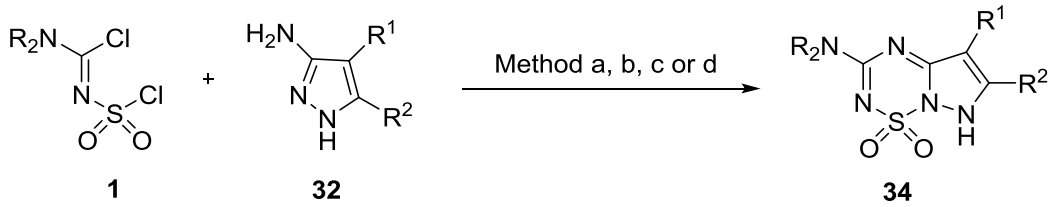
Complete structural identification of these novel ring systems is thus essential to develop effective methods for synthesis of these compounds and derivatives thereof. This entails characterisation by High Resolution Mass Spectrometry, Nuclear Magnetic Resonance Spectroscopy, melting point analysis and elemental analysis where required; and lastly X-ray crystallography of important representatives. The most efficient methods of synthesis of these compounds were sought along with determination of the regioselectivity of the reactions, and the effects of substrate substitution patterns or changes to the reaction conditions employed. The utility of product ring systems as reactive scaffolds was to be determined by treatment with common electrophilic reagents such as alkyl halides, acyl halides, acid anhydrides, and sulfonate esters. This also serves to identify any dominant nucleophilic sites providing insight into the chemical behaviour of these novel systems. Any bicyclic ring products are to be added to the CSIRO Compound Library.

1.6 Previous studies of 3-aminopyrazoles with *N*,*N*–dialkyl *N*'-chlorosulfonyl chloroformamidines

Preliminary studies on the 3-aminopyrazole system were conducted during the author's Honours research as part of the synthesis of a small library of novel compounds. Aminopyrazoles **32** were expected to react as :N-C-N: 1,3-dinucleophiles to form fused pyrazolothiatriazines **34** and/or **35** with perhaps some possibility of forming fused pyrazolothiatriazines **36** and **37** by an :N-N: 1,2-nucleophilic reaction (**Scheme 16**). It

was hypothesised that these reactions might have been influenced by steric interactions, or by the nature of substituents on C4 and C5 positions. Different reaction conditions offered the possibility of altered yields and/or ratio of isomeric products obtained.

The reaction of selected pyrazoles **32** with dichlorides **1** under various solvent, base and heating conditions all afforded derivatives of the novel pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system **34** as the only isolated products in each case (**Scheme 18**).



Scheme 18: Synthesis of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** from 3-aminopyrazoles **32** and dichlorides **1**. Method a = DMPU, 80^{o}C; b = DMPU, $^{i}Pr_2NEt$; c = K_2CO_3/H_2O, $Bu^{n}_4NHSO_4/CH_2Cl_2$; d = CH_2Cl_2, Et_3N.

The 3-aminopyrazoles **32a-d** outlined in **Figure 7** were utilised as examples of different functionally substituted starting materials. The relative yields of the condensation with dielectrophiles **1** under a small set of conditions was reported and is outlined in **Figure 8**, which would be a consequence of both the speed of reaction and also the ease of product isolation.

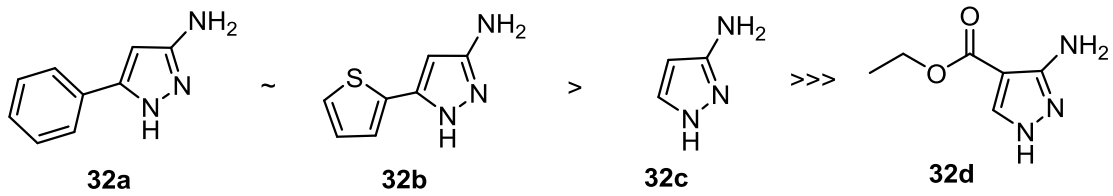


Fig 8. The 3-amino pyrazole **32** employed in reactions with dichlorides **1**, and the relative success based on comparative reaction times and yields during preliminary studies.

It became apparent that further investigation into the synthesis of this novel ring system was required, which included the exploration of other pyrazole substrates and modifications to the reaction. These studies were continued into the PhD work and the final results from this investigation are outlined in Chapter 2.

1.7 Thiatriazine and Thiadiazine Rings in Organic Synthesis

The potential ring products from condensations of aminopyrazoles **32** or **33** with dichlorides **1** are fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34**, pyrazolo[3,4-e][1,2,5]thiadiazines **38** and pyrazolo[3,4-c][1,2,6]thiadiazines **39** which contain the unusual thiadiazine or thiatriazine rings (**Figure 9**).

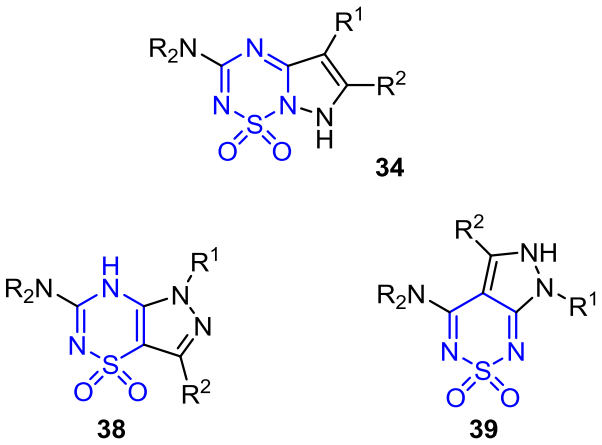


Fig 9. Thiatriazine and thiadiazine motif highlighted in structures of fused pyrazolo-thiadiazines **38** and **39** and –thiatriazines **34**.

There are a limited number of ring systems described in the literature which contain more than three N, O or S atoms, and the majority of those that have been authenticated have only been obtained in the form of reduced derivatives (or as S,S-dioxides in the case of those containing a sulfur atom).[30] In fact, thiadiazines and thiatriazines are not naturally occurring compounds and can only be obtained by synthetic methods.

Some functionalized thiatriazine dioxides are efficient anticholesteremic agents,[31] gastric secretion inhibitors,[32,33] herbicides,[34–38] and H2-antagonists.[39] Thiadiazine dioxides have been reported for antagonist activity against cannabinoid receptors,[40] vasorelaxant properties,[41] lowering serum uric acid,[42] and as chemosensory receptor therapies for metabolic disorders.[43]

The few reported examples of [1,2,6]thiadiazines and [1,2,4,6]thiatriazines are most often synthesised from sulfamide as a precursor;[44–46] and a comprehensive review by Spillane and Malaubier was published in 2013.[47] Reported methods of synthesis of thiadiazines and thiatriazines containing the sulfamide moiety (**Figure 10**) include the condensation of sulfonamides with formaldehyde and primary amines,[45] and the treatment of aminotetrazoles[48] or thiadiazoles[49] with chlorosulfonyl isocyanate.

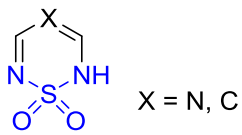


Fig 10. Sulfamide moiety within thiadiazine or thiatriazine rings.

These methods have disadvantages such as: multistep reaction procedures, long reaction times, low yields of target compounds, and the necessity to use highly reactive reagents such as chlorosulfonylamides, chlorosulfonyl isocyanate, and sodium amide.[46,47] Other methods have been outlined[47,50–52] including ring expansion of thiatriazoles or condensation of other fused systems to form rings, and these are often narrow in scope.

Given that the synthesis of cyclic sulfamides is a sparingly reported subject, the applications of such products have not yet been adequately explored. Furthermore, these unusual ring systems are generally free of patent competition and this means that the area of chemical space surrounding these compounds is likely to be open for further exploration.

1.8 Nomenclature of Uncommon Sulfur and Nitrogen Containing Heterocycles

Since these thiadiazine and thiatriazine rings are intrinsically rare compounds, they are not known by common names. It is worth briefly outlining the nomenclature of such systems by IUPAC standards. The latest revision of the extended Hantzsch-Widman system of nomenclature for heteromonocycles was reported in 1983.[53] The 'IUPAC recommendations for nomenclature of fused and bridged-fused ring systems' published in 1998[54] constitute a comprehensive documentation for naming of bicyclic ring systems that this thesis entails.

Ortho-fused rings are two rings that have only two atoms and one bond in common. Since the fused ring systems proposed during this study will fall into this category, only the naming rules for otho-fused systems will be outlined.

Noteworthy rules within this naming system are as follows:

a) Valence II Sulfur atoms are prefixed with thia- and nitrogen atoms are prefixed as aza-.
b) If a position of fusion is occupied by a heteroatom, both the components (ring systems) are considered to possess that heteroatom.
c) Unsaturated 6- membered rings have the suffix –ine (5-membered is –ole) i.e. thiatriazine or thiatriazole. (**Figure 11**).

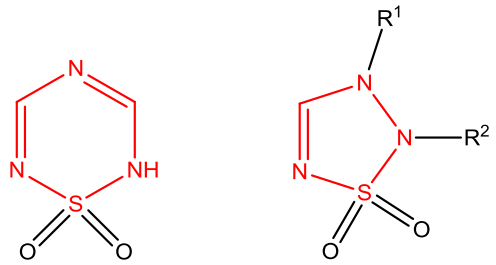
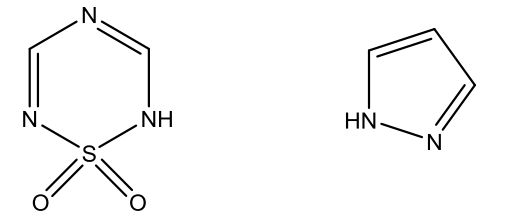


Fig 11. Thiatriazine and thiatriazole rings.

d) Fused rings must be named by assigning one ring as the *parent* component and the other(s) as *attached* component(s) (see **Figure 12**).

a. The *parent* component is the one with highest seniority according to the criteria given in the Hantzsch-Widman system of nomenclature FR-2.3, and is represented in the fusion name by citing that ring/ring system last in the name.

b. The *attached* component is the component of a fused ring system which is not the parent compound and is expressed by fusion prefixes (see part e)). The first order prefixes are for those directly attached to the parent ring but there may be second order or third order prefixes and so on for additional rings.



thiatriazine (parent) pyrazole (attached)

Fig 12. *Thiatriazine takes priority over a pyrazole ring*

e) Fused rings in which one ring has no accepted trivial or semi-systematic name are named by selecting the component with the trivial/semi-systematic name as the *principal* component. The other components become a *prefix*. If one or more rings with semi-systematic names are present then the order of priority is outlined in the Hantzsch-Widman rules.

f) The prefixes designated for an attached component are formed by changing the terminal 'e' of a trivial or Hantzsch-Widman name of a component to into 'o' i.e. Pyrazolo from pyrazole.

g) Numbering occurs from the highest priority substituent and proceeds in a clockwise fashion around the bridged system:

a. A component containing the greatest number of heteroatoms is most preferred. When two rings contain the same number of heteroatoms, the ring with the greater variety of heteroatoms is selected.

b. If there are alternative numbering systems of a ring system which are equally preferred then give low numbers of heteroatoms when considered in the order: O, S, Se, Te, N, P, As, Sb, Bi, Si, Ge, Sn, Pb, B (Table 1 of the revised Hantzsch-Widman system).

c. If two rings have the same heteroatom(s) then the component with the larger ring is given higher priority.

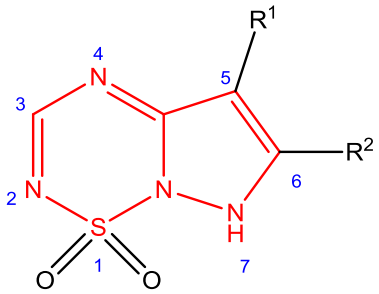
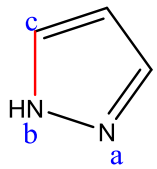


Fig 13. An example of a simple fused *pyrazolothiatriazine* ring system.

In ortho fused compounds, parent rings are denoted by numbering and additional rings are given roman letters (“a”, “b”, “c”, etc.) for each point of fusion. These letters are given to the number of the position immediately preceding. This should begin from the first heteroatom, and should follow in such a direction that heteroatoms are given the earliest letters possible. If this is not applicable then the letters will follow a clockwise fashion.



In the above example, the point of fusion is atom “b”

The fusion indicated by appropriate letters and numbers are enclosed in a square bracket and placed immediately after the prefix of the attached component. The numbers (positions of attachment) of the second component are placed in the sequence in which they are attached to the base component.

The name is formed of: name of attached ring [number, number-*letter*] name of parent ring.

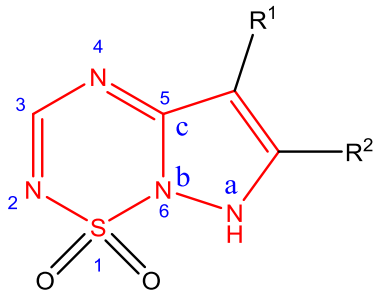


Fig 14. Numbering of a fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system.

These example compounds are thus named as fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxides according to the most recently updated, standard IUPAC nomenclature.

1.9 X-Ray Crystallography

It should be mentioned that some of the claims of heterocyclic systems with maximum unsaturation reported in the older literature have proved erroneous. In most cases this was not unexpected due to limitations of the techniques available at the time. Due to a lack of proton coupling and carbon environments in the heterocyclic core, the ring systems afforded from *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines (**1**) have typically suffered from NMR and mass-spectral data ambiguity. Publications arising from the collaborative research with CSIRO stemming from dichlorides **1** have included X-ray crystal structures as definitive proof of the correct structural assignment. X-ray crystallography was therefore an integral aspect of this project, and the theory of this fundamental technique will be briefly outlined.

1.9.1 X-Ray diffraction

The reason X-rays are such a valuable form of electromagnetic radiation in the structural elucidation of compounds is because the wavelengths of the X-rays are on the same order of magnitude (typically 0.5Å to 1.6Å) as the spacing for atoms in solids.[55]

Individual atoms in a molecule can diffract X-rays but they have only a weak scattering effect so they may pass through a single molecule without any diffraction.[56] A crystal diffraction pattern however can be measured because a crystal is composed of a number of patterns (ie. the unit cells) in a regular and ordered arrangement.

1.9.2 The crystal lattice

To best comprehend how X-ray crystallography works, one must first understand the basic chemistry of a crystal lattice. Solid materials are often classified according to the regularity with which atoms are arranged with respect to one another, and a crystalline material is defined as one in which the atoms are situated in a repeating or periodic array so that a long-range order exists.[57] An ideal crystal, one that is necessary for X-ray diffraction experiments, is a repetition of identical structural units in three dimensional space.[55] The term lattice is regularly used in the context of crystal structures meaning a three dimensional array of points coinciding with atom positions. The crystal structure can be divided into seven repeat entities specified as the unit cells (**Figure 15**).[58]

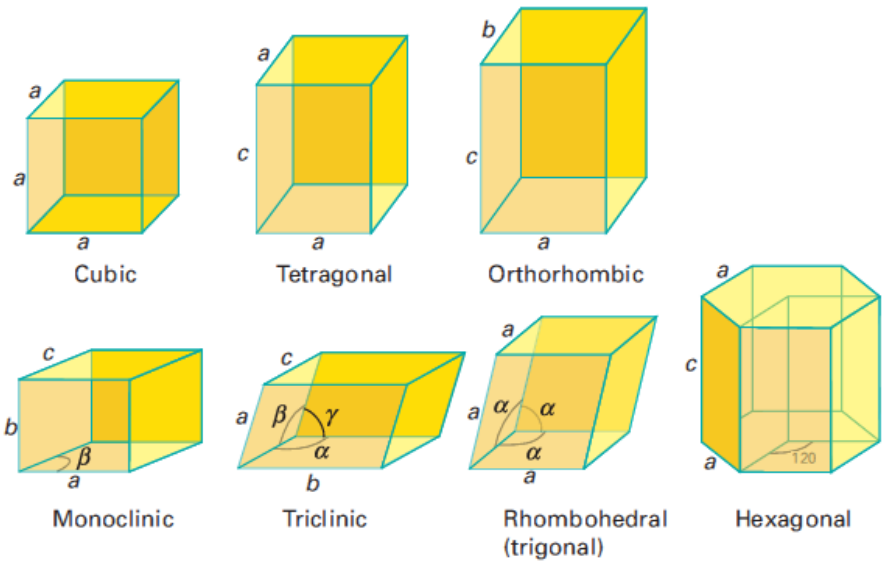


Fig 15.[58] The seven crystal systems. These lattices are also classified according to some basic symmetry groups which gives rise to 14 Bravais lattices in total.[57]

Furthermore, the symmetry group of a 3-dimensional configuration in space is used to provide 219 distinct types of space groups. A definitive source regarding 3-dimensional space groups is the *International Tables for Crystallography*.[59] All unit cell and space group types will not be outlined here but can be found in the Acta Crystallographica paper by Janssen et al which encompasses the symbols for all arithmetic crystal classes.[60]

There is a large degree of mathematical theory behind the crystal lattice which is beyond the scope of this study and will not be discussed here any further. An appreciation of the crystal lattice as a repeating unit in a single crystal is, however, imperative in understanding the underpinning theory behind X-ray crystallography.

1.9.3 An Overview of Single Crystal X-ray Diffraction

When a beam of X-rays encroaches on a crystal, a portion of the beam will be scattered by the electrons within the molecules. Since the crystal should be a repeating set of the same unit cell, collection of X-ray scattering patterns from different angles will allow the determination of the location of electron density within each repeating unit.[56] The relationship between X-ray wavelength, interatomic spacing and the angle of diffraction are described by Bragg's law which is a necessary condition for diffraction by real crystals.[61] Scattering can be described as reflection from two parallel planes of distance d then the angle at which consecutive interference occurs between waves of wavelength (λ) is given by the Bragg equation ($2d\sin\theta = n\lambda$) where d is path distance between the X-rays and θ is the angle of reflection (**Figure 16**).[55]

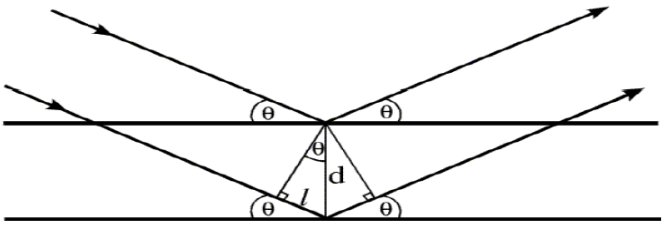


Fig 16.[62] The geometry of diffraction and derivation of the Bragg equation.

Bragg's law describes the relationship between the wavelength of electromagnetic radiation and the diffraction angle with the lattice spacing in a crystalline sample. W.H and W. I Bragg demonstrated that this angle could be calculated in terms of the path difference between a ray reflected by one plane and that reflected by the next plane after it in the lattice. Only those angles θ are allowed, where the path difference 2d sin θ is an integral multiple of the wavelength.[62] This law is the basis behind the mathematical transformation of X-ray scattering data or 'reflections' into an 'electron density map' from which chemists can interpret crystal data.

1.9.4 The X-ray Diffractometer

The X-ray diffractometer is an apparatus used to determine the angles at which diffraction occurs from a crystalline solid. The image below (**Figure 17**) is a photograph taken of an X-Calibur 3 Diffractometer (Oxford Diffraction) for use in data collection of single crystal X-ray diffraction patterns.

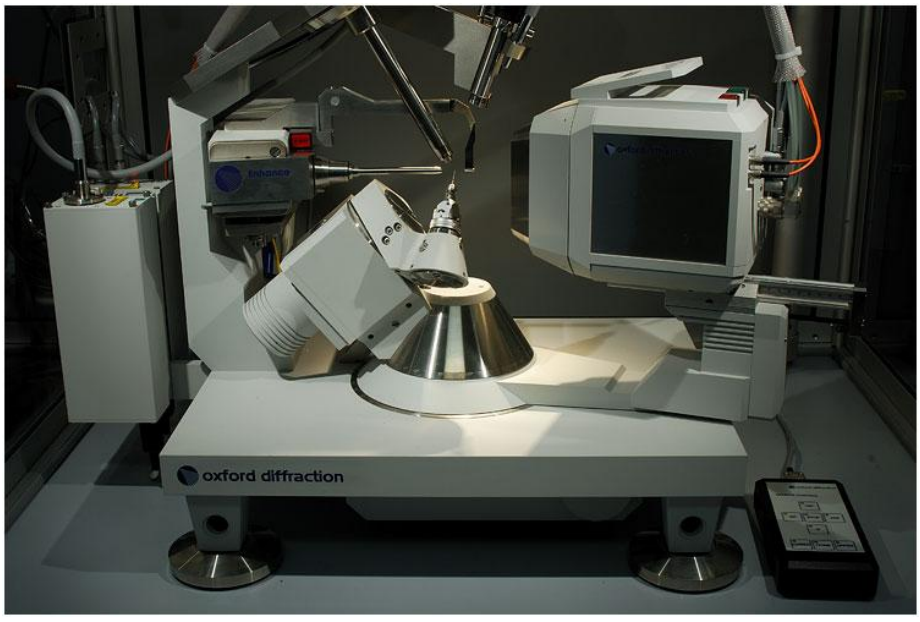


Fig 17. A Mo-target Oxford Diffraction X-Calibur X-ray diffractometer for small molecule structure determination. This image was taken in the Bragg crystallography facility at Adelaide University (SA).

These X-rays are generated by a cathode ray tube and filtered to produce monochromatic radiation.[56] A sample which is placed in the path of the X-rays is then exposed to the X-ray beam and the interaction of the incident rays with the sample produces constructive interference (they are scattered by the electrons in the atoms), when conditions satisfy Bragg's Law.[56] These diffracted X-rays can be detected (by a photon detector for example) when they are reflected on a target, then they are processed and counted. **Figure 18** shows a pattern of reflections obtained from protein X-ray diffraction experiments of an enzyme.

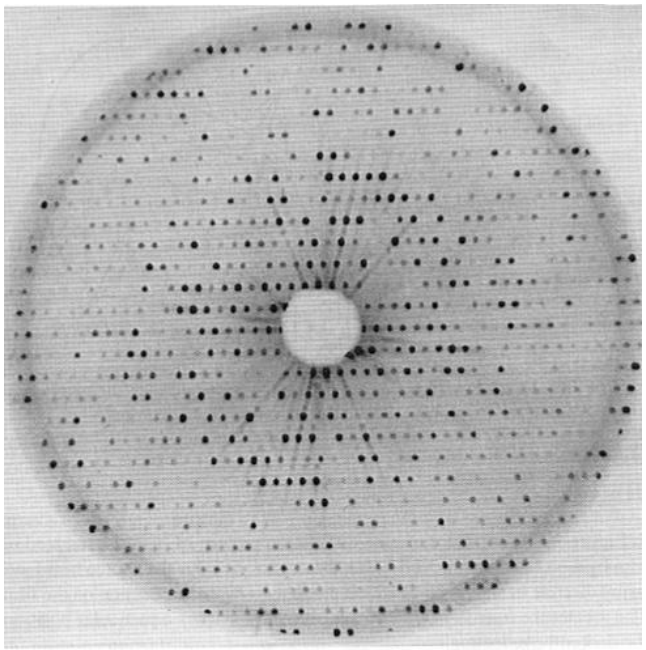


Fig 18.[56] An image of the raw crystal data of an enzyme which forms a diffraction pattern around the crystal which lies in the centre of the image. Repeating patterns can be seen surrounding the crystal sample in the centre of the image, and some reflections are notably stronger (darker) than others. A well diffracting crystal will give rise to strong reflections further away from the sample.

The geometry of the incident rays, the orientation of the centred crystal (which is mounted on a rotating arm), and the location of the detector can be changed in order for all possible diffraction directions of the lattice to be obtained. Periodic arrays of scattering centres are separated by distances similar to the wavelength of the radiation (nearby 1Å) such as that which exists in a crystal.[57] The intensity of the diffraction

depends on the details of the crystal structure and the identities of the atoms. How well an atom scatters X-rays depends on how many electrons are in the crystal lattice, their size and where they are located in the cell. A more detailed description of crystal structure determination including theoretical considerations and a description of X-ray diffraction techniques can be found in the text 'Crystal Structure Determination' by W. Massa.[62]

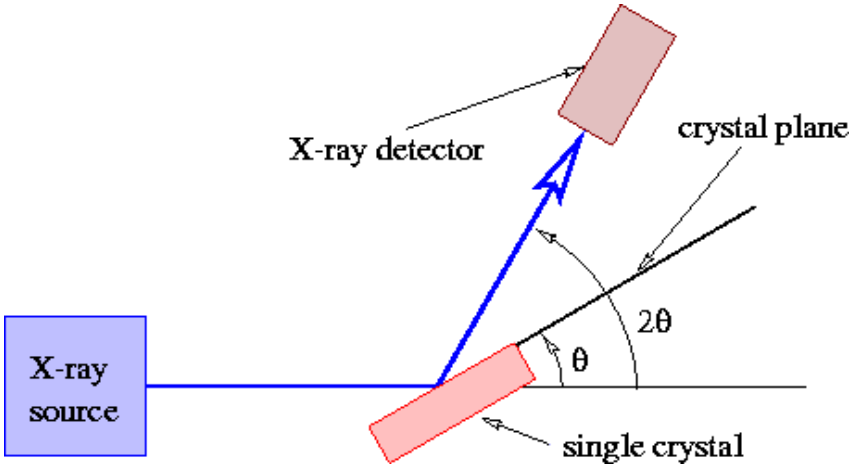


Fig 19. A trivial schematic of an X-ray diffractometer.

Figure 19 shows a simplified schematic of an X-ray diffractometer. X-rays are produced in a device called an X-ray tube which consists of an evacuated chamber with a tungsten filament at one end (the cathode), and a metal target at the other end (usually Cu or Mo) as the anode.[62] When an electrical current is passed through the filament, excited electrons from the tungsten are emitted.[62] The electron will strike atoms at the target, dislodging inner shell electrons of the target atom. As a result, electrons on the outer shell jump down to fill the void in the inner shell. Then because the inner shells have lower energy than the outer shells, they emit radiation in the form of high energy X-rays. The X-rays are then filtered through a monochromator (a single crystal flake of graphite, quartz or germanium) before striking the crystal of study. The diffracted X-rays are collected by an area detector.[56] Diffraction angles and intensities can subsequently be measured which then delivers information to the crystallographer about the bond angles, bond lengths, sizes of the atoms, areas of high and low electron densities and ultimately the overall framework of the repeating molecule(s).

1.9.5 X-Ray refinement

X-ray crystal structure determination is heavily assisted by modern computational methods; however a refinement process must still be carried out by the crystallographer in order to obtain accurate and publishable data. Only a brief summary of refinement will be required to provide the reader with a rudimentary understanding of the X-ray refinement process. The following information is a summary from an historic *Acta Crystallographica* entry from Zbigniew Dauter which outlines common data-collection strategies.[63]

For data collection purposes, the X-ray data is measured in completeness which includes: the indices of the lattice, their intensities and standard uncertainties; and also some geometric and informative content.

The geometric factor is a qualitative factor which arises from aspects such as the symmetry of the crystal lattice itself and the detector setup. Examples include the angular rotating method, the rotation range, crystal-to-detector distance, beam divergence and the mosaicity of the crystal (a measure of long-range disorder).

The informative factor includes the quality of the data, the range of the detector itself, and the R-factors. The R-factors are universally applied as a measure of the quality of data for publication purposes where acceptable R^1 values usually lie between 0.03 and 0.08. The smaller the R-factor, the greater agreement there is between the modelled structure and the electron density from the data itself. R^1 values will never be zero due mostly to the movements of atoms when contacted by the X-ray beam (even at cold temperatures) and the electron smearing effect of protons.

The direct results from the experimental measurements for a crystal structure are the unit cell parameters, the most probable space group(s), and the intensity data.[62] The diffraction data is converted into the individual structure factor files by a process of *Fourier transformation*.[62] A *Fourier synthesis* gives the electron density for every point X,Y,Z in the unit cell (**Figure 20**).

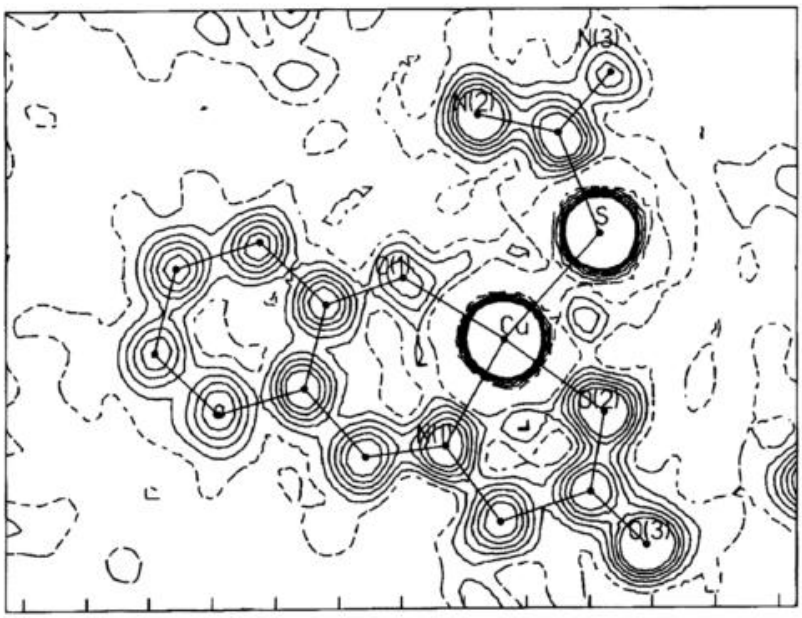


Fig 20.[62] A Fourier synthesis section through the molecular plane of the thiourea derivative of N-Salicylideneglycinatocopper (II). The contours are drawn at an interval of 0.1eÅ^{-3} which illustrates heavy atoms such as Cu and S with high density; however the bonding electrons in the rings are also visible.

This provides an 'electron contour map' of the various electron densities within the unit cell which can be *refined*.[62] The word *refinement* refers to the statistical adjustment of atomic coordinates from modelled data to fit the electron density data collected from a single crystal.[56] In layman terms, this means that the molecular structure is built with a modelling program and then the calculated parameters for that structure (electron density, bond lengths, bond angles etc.) are compared with the experimental results. A common misconception about X-ray crystallography is that the diffraction patterns are used to determine the positions of the hydrogen atoms as well as the atoms in the 'skeleton' structure. This is not true, since in many cases the hydrogen atoms cannot be located at all; in fact the hydrogen atoms are generally too small for detection by X-ray techniques and present an electron smearing effect due to slight vibrations of the atoms.[64] The ability to detect the location of hydrogen atoms is highly dependent on the quality of the data set, the presence or absence of heavy atoms, and the extent of thermal motion of all the atoms.[62] Hydrogen atoms are frequently placed in geometrically calculated positions and then refined using a 'riding model' which means applying

constraints to the X—H bond lengths and H−−X−−H or H−−X−−Y angles, setting them to certain values.[64] The refinement programs used for this PhD research project (X-Seed, ShelX 1997) are universally applied in modern day crystallography.[56,65–67] No further details regarding the use of the refinement program will be included in this thesis. The official ShelX website[67] contains instructions on the operation of the ShelX program, and experimental details of the use of the ShelX program can be found in the literature.[66]

The use of X-ray crystallography will allow the unequivocal reporting of the structures of these novel compounds and will remove any ambiguity arising from the lack of structural information from NMR spectroscopy and mass spectrometry due to the potential for isomeric forms.

1.10 Doctoral research contributions to this study

The work presented in this thesis builds upon the examples presented by the research group in papers I-IX[19,22–29] of the series "*N,N-Dialkyl-N'-Chlorosulfonyl Chloroformamidines in Heterocyclic Synthesis*" which have been outlined in Section 1.4. This PhD study focused on the synthesis and characterisation of two novel ring systems derived from dichlorides **1**, and subsequent chemistry of these novel compounds. Readily commercially available amino pyrazoles were chosen as representative bidentate nucleophiles due to the various reactive sites on this class of compounds, and the availability of literature precedent.

Chapter 2 details the synthesis of pyrazolo-[1,5-*b*][1,2,4,6]thiatriazines and other side products from the condensation of 3-aminopyrazoles **32** with dichlorides **1**. Chapter 3 discusses the use of N1-substituted 5-aminopyrazoles **33** to afford the pyrazolo[3,4-*e*][1,2,6]thiadiazine ring system, and also details some unusual side products from these condensation reactions. Chapter 4 describes alkylated derivatives of systems **34** and **38**, including an unusual ring expansion reaction. Chapter 5 summarises reactions involving the extrusion of the sulfur dioxide moiety in ring system **34** and the application of this

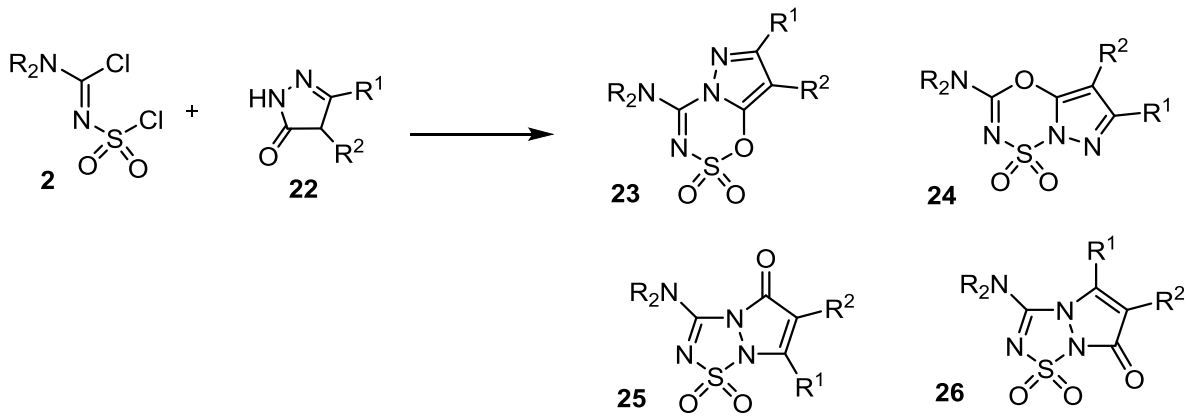
chemistry to synthesis of pyrazolo-triazine compounds. Chapter 6 discusses pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system modifications including acylation, and reactions of the nucleophilic carbon atom C5. Chapter 7 describes ring cleavage reactions of benzylated derivatives of compounds **34** which include an unusual thermal rearrangement, a ring expansion, and alcoholysis and aminolysis at the SO_2 moiety.

CHAPTER 2: SYNTHESIS OF PYRAZOLO[1,5-*B*][1,2,4,6]THIATRIAZINE 1,1 DIOXIDES

2.1 Introductory remarks

2.1.1 Novel, low molecular weight ring systems containing synthetic handles

The CSIRO and Flinders group recently documented[28] that the reaction between *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1** and *N*-unsubstituted pyrazol-3-ones **22** furnished two previously unknown pyrazolo-fused oxathiadiazine ring systems **23** and **24**, along with one or two isomers of the rare pyrazolo[1,2-*b*][1,2,3,5]thiatriazole ring system **25** and **26** in minor proportions (**Scheme 19**). The four possible regioisomeric products from this reaction seemingly stemmed from the numerous tautomeric forms adopted by *N*-unsubstituted pyrazol-3-ones. [28]



Scheme 19: Four isomeric products synthesised from the condensation of N-unsubstituted pyrazolones **22** with dichlorides **1**.

The distinctive chemical originality of these products makes them attractive candidates for biological testing; however, as mentioned earlier, further elaboration was inhibited by a lack of reactive sites. This meant that a diversity of substituents could not be

readily introduced and hence, opportunities for structural variations were limited. This synthetic methodology was re-evaluated to provide the potential for ring products which contain synthetic handles.

As aforementioned in Chapter 1, 3-aminopyrazoles **32** are bidentate nucleophiles and presented the possibility for the synthesis of novel, low molecular weight heterocyclic products. The main focus of this PhD study was to generate related heterocycles bearing an NH group, which would then allow subsequent substitution and diversification within the pool of available test compounds. Employing 3-aminopyrazoles **32** in place of *N*-unsubstituted pyrazol-3-ones **22** in reactions with *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines **1** appeared to offer the desired product versatility. Aminopyrazoles, which are often favoured as precursors to fused pyrazole ring systems, are readily available and are known to provide useful precursors in the dye, pharmaceutical, and agrochemical industries.[68]

2.1.2 Reactions of aminopyrazoles as 1,2/1,3 dinucleophilic systems

Interestingly, as there are very few examples of naturally occurring pyrazoles, probably due to the difficulty in the construction of N-N bonds by living organisms, the availability of pyrazole-containing compounds is predicated upon synthetic methods.[69,70] 3-aminopyrazoles are low-cost, commercially available compounds which can also be synthesised readily following a number of well-established literature methods.[71] A review by Anwar and Elnagdi[72] outlines recent developments in aminopyrazole chemistry, including synthesis from the condensation of hydrazines with α,β-unsaturated nitriles or 3-oxoalkanenitriles; or from substituted hydrazones. More recently, Aggarwal *et al*[70] reviewed the production of 3- and 5-aminopyrazoles from conventional methods such as the reaction of β-ketonitriles, malononitrile and its derivatives with hydrazines in addition to modern methods such as resin supported solid-phase synthesis, multi-component synthesis and ring transformations. The 3-aminopyrazole (**32**) system exists as two dominant tautomeric forms (**Figure 21**) and constitutes as an ambident dinucleophile.[71,73]

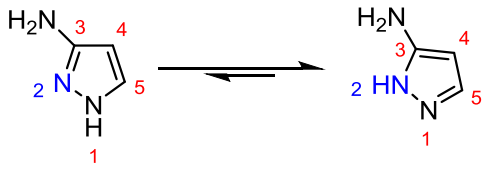


Fig 21. Tautomeric structures of 3-amino pyrazole **32**.

Aminopyrazoles have been explored as templates and scaffolds towards higher structures for both biological and industrial purposes. Pharmacophoric requirements of 3-aminopyrazoles as inhibitors of Cyclin Dependant Kinase (CDK2) enzymes were investigated by QSAR studies.[74] 3-Aminopyrazole derivatives have also been studied for use as hypnotic drugs,[75] inhibitors of protein kinases,[76] dyes,[77] and templates to influence β-sheet formation in peptides.[78] Further information on the reactivity of the 3-aminopyrazole ring system towards various bidentate electrophiles can be found within a 2009 review by Anwar and Elnagdi[72], and a number of textbook chapters within 'Comprehensive Heterocyclic Chemistry' also cover this reactivity up until 2008.[69,79,80] Since they are low molecular weight heterocycles, 3-aminopyrazoles have amenability for rapid class expansion; hence investigation of new derivatives for future applications is an attractive proposal. Additionally, pyrazole rings are known to be relatively stable to metabolism compared to other five-membered ring heteroaromatic compounds[7] and unlike isoxazoles and isothiazoles, this ring has not been shown to undergo reductive metabolism,[6] so it is unsurprising that pyrazoles are a common motif in drug molecules.

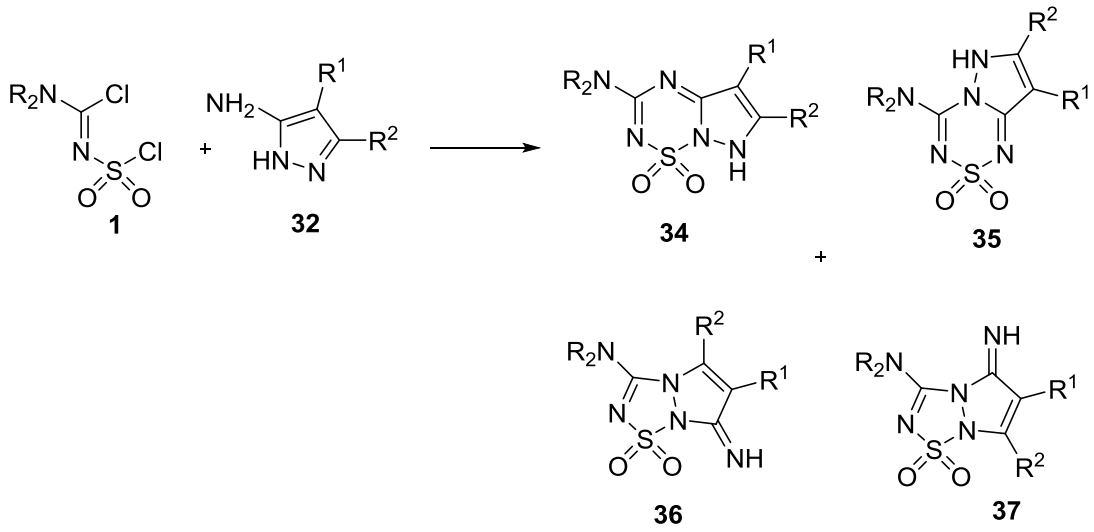
The reactivity of atoms in the 3-aminopyrazole **32** system has also been well documented. It now established that the ring nitrogen (N2 in **Figure 21**) is the most basic nitrogen, and in many examples the strongest nucleophile, but that it is also the most hindered site in the molecule.[81–83] Nucleophilic attack is therefore governed by steric consideration; hence, difficulty arises in predicting the dominant site of attack with certain electrophiles. Reactions of carbon-disulfides with **32** in attempts to selectively substitute on specific nitrogens have failed to provide a single isomer.[84] Selective alkylation at the exocyclic NH_2 group has been reported.[85] Acylation with ^{t}butoxy-carbonyl and carbo-benzyloxy groups proved selective; conversely, at the ring nitrogen N2.[86] Blake *et al*[87] strategised an increase in steric bulk at the 5-position dramatically increases regioselectivity towards the ring nitrogen N2. Initial substitution

on N2 has been used to selectively react agents such as acid chlorides at ring nitrogen N1 and the amino group.[88,89]

Preparations of ring-fused products from this dinucleophile and its derivatives have been achieved using a variety of dielectrophiles including β-diketones,[90] β-keto esters,[91] β-ketoaldehydes[80,92,93] and oxaldiimidoyl dichlorides.[73] In these cases however, the condensations are not completely regioselective and as a result, the separation and structural assignment of isomers is often tedious. Competing reactivities and differing steric effects of the electrophilic sites contribute to the formation of more than one isomer.[81,82] Enaminocarbonyl compounds have been used in place of the corresponding 1,3-dicarbonyl compounds to provide good yields of a single isomeric product. By altering the substituents on either carbon of a vinyl group, such as in 3-dimethyl-aminopropiophenone and cinamaldehyde, one isomer could be obtained from their reactions with 3-aminopyrazole derivatives.[82,94] Thomas et al[95] were able to suitably modify electrophilic centres on α-oxoketene dithioacetals to afford a sole product upon reaction with 3-aminopyrazoles. Other dielectrophiles utilised in condensations with 3-aminopyrazoles to produce one isomer include α,β-unsaturated nitriles,[75,96] and cyanothioacetamides.[97] In reactions that are highly regioselective such as these, attempts to alter selectivity under alternative reaction conditions were often unsuccessful. This demonstrates that factors such as steric bulk and electron withdrawing groups may greatly bias competition between electrophilic sites, with greater potential to produce a single isomer due to the reactive dominance of one electrophilic site.

2.1.3 The selectivity of condensation between *N*,*N*-dialkyl *N*'-chlorosulfonyl chloroformamidines and 3-aminopyrazoles

Reactions of dichlorides **1** with some 3-aminopyrazole derivatives have the potential to deliver four isomeric fused ring systems with marked similarities to the compounds **23-26** (**Scheme 20**) with the exception of a nitrogen in place of the oxygen from the pyrazolone moiety. Nonetheless, these may exhibit alternate physical and chemical behaviour since amino pyrazoles and pyrazolones are themselves different in reactivity.



Scheme 20: Possible isomeric products from the reaction of dichloro compound **1** with 3-aminopyrazoles **32**.

The condensation of pyraz-3-olones **22** with dichlorides **1** demonstrated that literature precedent does not always allow the prediction of the major isomer from a bidentate reaction.[28] The major differences between 3-pyrazolones **22** and 3-aminopyrazoles **32** are that pyrazolones **22** comprise of four tautomeric forms (**Figure 22**) where pyrazoles **32** only comprise of two dominant forms (**Figure 21**).[79,98] Additionally, the NH_2 group of a pyrazole ring is generally more nucleophilic than the OH group of the pyrazolol tautomers which often react preferentially at the pyrazole ring; and so in the case of 3-aminopyrazoles **32** the reactivity at the NH_2 group tends to dominate.[69,79,80,98] As outlined in Section 2.1.2, this reactivity is highly directed by substrate substitution patterns and the class of electrophile.

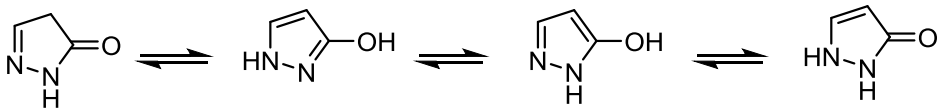


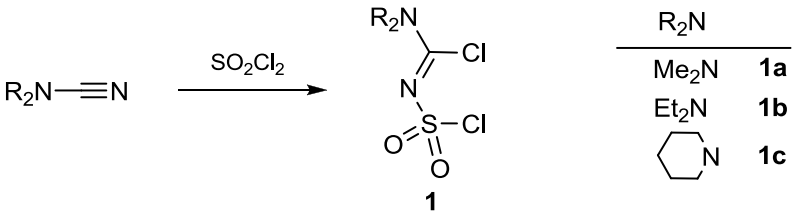
Fig 22. Tautomers of N-unsubstituted pyrazolones **22**.

The effects of steric interactions around the dichloride also need to be taken into consideration as these could affect reactivity with the two pyrazole derivatives differently. The effects of steric and electron withdrawing interactions arising from

substituents on the pyrazole ring is also important. The previous studies during the initial investigation provided a starting point for the exploration into new ring systems from the condensation of dichlorides **1** with pyrazoles **32**; however the substrate scope was limited, and the conditions had not yet been optimised. A more extensive series of reactions between various 3-amino pyrazole derivatives **32** and a few examples of dichloride compounds **1** needed to be carried out in order to establish the regioselectivity of the reaction to a greater level of detail. Alternative reaction conditions may also lead to apparent changes in the ratio of isomeric products if the overall selectivity of the condensation reaction is found to be low, and so the product isolation process needed to account for all of the potential products.

2.2 Synthesis of the *N,N*-dialkyl *N*'-chlorosulfonyl chloroformamidines

Dichloride compounds such as 1-pyrrolidino-*N*'-chlorosulfonyl chloroformamidine[19,22,23] and *N*-methyl, *N*-butyl-*N*'-chlorosulfonyl chloroformamidine[23,25–27] which had been utilised in previous work were not explored in this PhD study because it was primarily an individual project that needed to be completed within time boundaries. The dichlorides **1a-c** (**Scheme 21**) were readily prepared from sulfuryl chloride and the corresponding dialkyl cyanamides following the previous literature procedures.[99] The three substrates outlined in **Scheme 21** offer examples of dialkyl and cyclic amino groups flanking the formamidine moiety and were deemed sufficient for this study.



Scheme 21: General synthesis of dichlorides **1**.

Two molar equivalents of sulfuryl chloride typically afforded the highest yields which had been established by previous research which outlined that decomposition products formed when lesser equivalents were used.[18,22] The three dielectrophiles **1a-c** were synthesised following methods from Fallon *et al*[19] by the application of different alkyl groups on the cyanamide precursor; dimethyl **1a**, diethyl **1b** and piperidino **1c**. Removal of the excess sulfuryl chloride was achieved under vacuum (60-70°C) using a sodium hydroxide trap (to neutralise the sulfuric and hydrochloric acid byproducts which form by reaction with water). The dichloro compounds **1a**, **1b** and **1c** were obtained as pale yellow, low melting point crystalline solids which formed in near quantitative (>95%) yields and were not further purified.

The dichlorides **1** underwent reaction with DMSO and water; and so these were avoided as solvents for NMR analysis or reaction solvents. DMF was also avoided because of the hydroscopic nature of the compound. It is also important to note that these compounds slowly react with moisture which meant that special care was taken upon purification to promptly remove the excess sulfuryl chloride under the vacuum preparation, and then the remaining sulfuryl chloride was evaporated under a stream of nitrogen gas. The use of high vacuum systems such as portable oil pumps was avoided because the sulfuryl chloride would cause damage to expensive high-vacuum equipment. Acid chlorides **1** were unstable in air,[10] hence the dichlorides were stored under 5°C in an atmosphere of nitrogen. Evidence of decomposition was apparent after a number of months from hardening and slight discoloration (which was confirmed by NMR analysis) meaning that additional batches needed to be synthesised.

2.3 Synthesis of the novel fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1 dioxides

2.3.1 General methods and materials for synthesis

A set of commercially available 3-aminopyrazoles **32** (**Figure 23**) with various substitution patterns were utilised to investigate the overall regioselectivity observed in reactions with **1**.

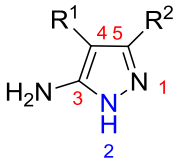


Fig 23. Pyrazoles **32a-g**

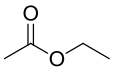
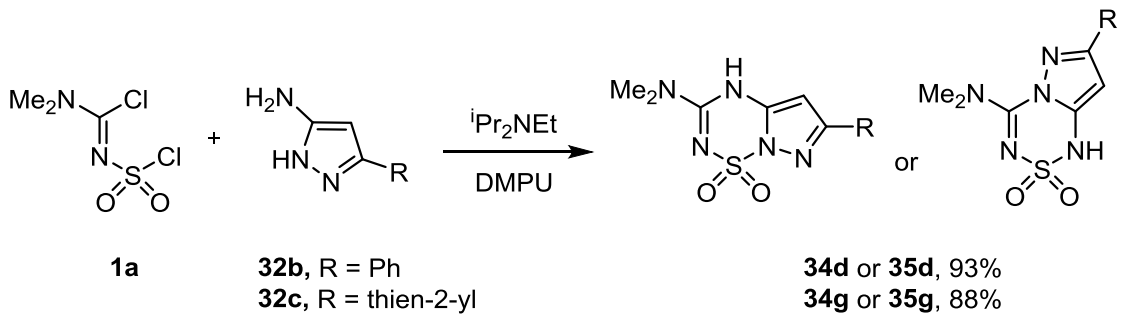
R^1	R^2	
H	H	**32a**
H	Ph	**32b**
H		**32c**
	H	**32d**
H	Me	**32e**
H		**32f**
Ph	H	**32g**
I	H	**32h**

Table 1: The range of 3-amino pyrazole **32** substrates selected for this study.

As with the pyrazol-3-one reactions described in Section 2.1.1, the condensation of dichlorides **1** with 3-aminopyrazoles **32** was examined under a variety of conditions:- (A) heating at 80°C in a polar, aprotic solvent, 1,3-dimethyl-3,4,5,6-tetrahydro-2(*1H*)-pyrimidinone (dimethylpropylene urea, DMPU); (B) in DMPU at ambient temperature with an organic base, *N*,*N*-diisopropylethylamine (Hünig's base); (C) a two-phase system (aqueous/non-polar organic solvent) containing an inorganic base and (D) a homogeneous system using a non-polar, organic solvent with an organic base (see Table 1). These conditions exemplify a range of solvent polarities and classes of base; and also an example of higher temperature conditions. The previous research efforts into synthesis of ring systems from chloride compounds **1** have explored a series of conditions, and reported methods A-D to be the most desirable in terms of reaction timeframe, yields, ease of isolation and cost effectiveness.[19,22,23,25]

2.3.2 Reactions of dichlorides with 3-amino pyrazole bidentate nucleophiles

A sample of dichloride **1a** was treated with two pyrazoles substituted at the 5-position (**32b** and **32c**) using the Hünig's base in DMPU method (B) which provided a single product in each case (**Scheme 22**). These compounds were confirmed by high resolution mass spectrometry to contain the formula of a fused ring product. At this point the structures of each compound were not certain; but they were hypothesised to be one of the 6-membered isomers **34** or **35** due to literature knowledge on the prevalent reactivity of 3-aminopyrazoles at ring nitrogen N1 or the exocyclic amine as discussed in section 2.1.



1a

32b, R = Ph
32c, R = thien-2-yl

34d or **35d**, 93%
34g or **35g**, 88%

Scheme 22: The formation of unknown ring products from treatment of dichloride **1a** with two pyrazole substrates **32b** and **32c**.

An X-ray crystal structure was used to determine the core structure of one sample (**Figure 24**), and was identified as a representative of the fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system **34**. The results from X-ray diffraction experiments and additional detail are further discussed in Chapter 8.

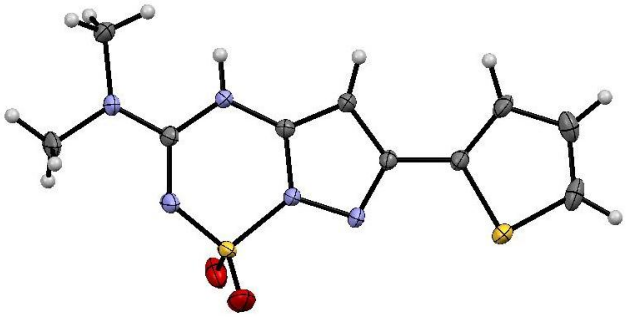
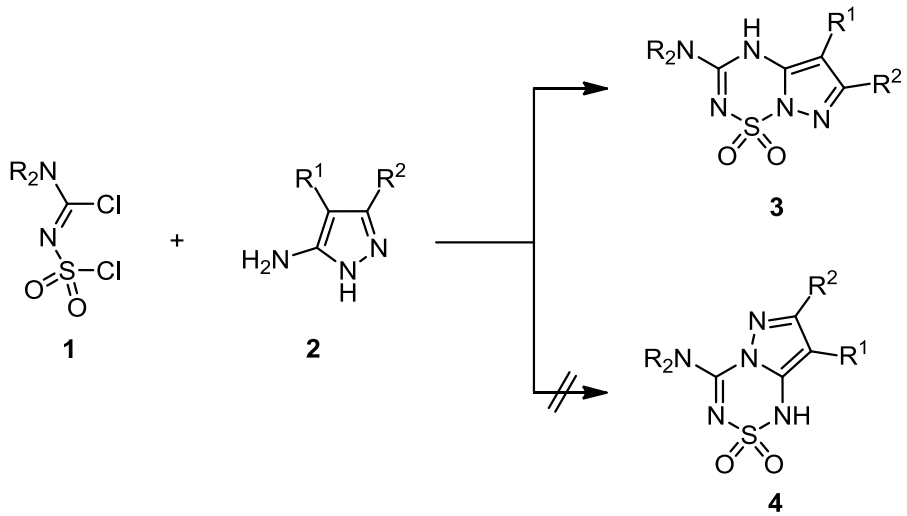


Fig 24. An ORTEP diagram of compound **34g**. A molecule of DMSO has been omitted for clarity.

The set of pyrazole substrates **32a**-**32h** were reacted with the dichloride compounds **1a**-**c** under the four sets of conditions. This was to furnish a series of novel derivatives, as well as establish the extent to which the condensation between species **1** and **32** was selective. Under all conditions employed, these reactions provided pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** (**Scheme 23**, **Table 2**) as generally stable, colourless, crystalline solids.[99] Compounds **34** are also representatives of a formerly unreported ring system. No other ring-fused products, such as the isomeric pyrazolo[5,1-*c*][1,2,4,6]thiatriazines **35**, were isolated from the product mixtures.



Scheme 23: Selective condensation of dichlorides **1** with 3-aminopyrazoles **32**.

Table 2: Synthesis of pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxides **34**.

R_2N	R^1	R^2	Method	Product(s)	Yield [%]
Me_2N (**1a**)	H	H (**32a**)	C	**34a**[#]	39
			D	**34a**[#]	76
Et_2N (**1b**)	H	H (**32a**)	D	**34b**[#]	64
 (**1c**)	H	H (**32a**)	D	**34c**	28
Me_2N (**1a**)	H	Ph (**32b**)	B	**34d**	93
Et_2N (**1b**)	H	Ph (**32b**)	B	**34e**	82
			A	**34e**	53
 (**1c**)	H	Ph (**32b**)	B	**34f**[#]	68
			A	**34f**[#]	36
Me_2N (**1a**)	H	Thien-2-yl (**32c**)	B	**34g**[#]	88
Et_2N (**1b**)	H	Thien-2-yl (**32c**)	A	**34h**[#]	42
			B	**34h**[#]	79
			C	**34h**[#]	24
			D	**34h**[#]	40
Me_2N (**1a**)	CO_2Et	H (**32d**)	B	**34i**[#]	28
			D	**34i**[#]	26
Me_2N (**1a**)	H	Me (**32e**)	D	**34j**	79
Me_2N (**1a**)	H	 (**32f**)	D	**34k**	90

 (**1c**)	H	 (**32f**)	D	**34l**	30

[#]X-ray crystal structure obtained.

Method A = DMPU/80°C; Method B = $^{i}Pr_2NEt$/DMPU; Method C = $KHCO_3$(aq.)/C_6H_6/$Bu^{n}_4NHSO_4$; Method D = Et_3N/CH_2Cl_2

In the case of compound **34c**, the reaction did not proceed to completion and the starting aminopyrazole **32a** was recovered in 43% yield. In the case of reactions between 3-amino pyrazole **32a** and each dichloride compound, the starting materials were not recovered to account for all of the material, and ^{1}H NMR experiments of the product mixture indicated some decomposition rather than incomplete conversion. In the case of compound **34i**, the reactions also did not proceed to completion and the starting aminopyrazole **32d** was recovered in 36% yield (Method B) and 49% (Method D).

Generally, the highest yields were afforded from pyrazoles containing thien-2-yl-, phenyl-, and cyclopropyl- substituents at the 5-position on the pyrazole ring. The poor aqueous solubility was exploited in precipitation of products by dilution of DMPU reaction mixtures with water, although the formation of product **34k** was also excellent (90%) from the application of method D. Comparison of each dichloride precursor indicated that the dimethylamino electrophile (**1a**) afforded the highest yields of isolated material, and this is typically attributed to lower solubility of derivatives containing methyl groups in comparison to those containing a diethylamino group (from **1b**) or a piperidine ring (from **1c**).

Compound **34i** was isolated in comparatively low yields (28 and 26%) from methods B and D which is indicative of a lower reactivity of the carbethoxy pyrazole **32d.** This observation could be explained by the electron withdrawing nature of the ester group at C4 reducing the electron density, and therefore nucleophilicity, of the exocyclic amino nitrogen (**Figure 25**). These results also confirm that the nucleophilicity of the NH_2 group is a great limiting factor on the cyclisation reaction.

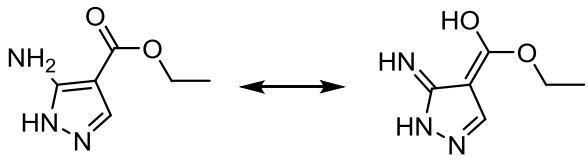


Fig 25. Resonance contributions of the 4-carbethoxy-3-amino pyrazole **32d**

There is an intramolecular N–H…O hydrogen bond between the exoamino group and the carbonyl of the ester in the pyrazole **32d**.[79,100] This hydrogen bond further contributes to the poor reactivity of the amino group (**Figure 26**).

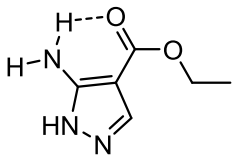
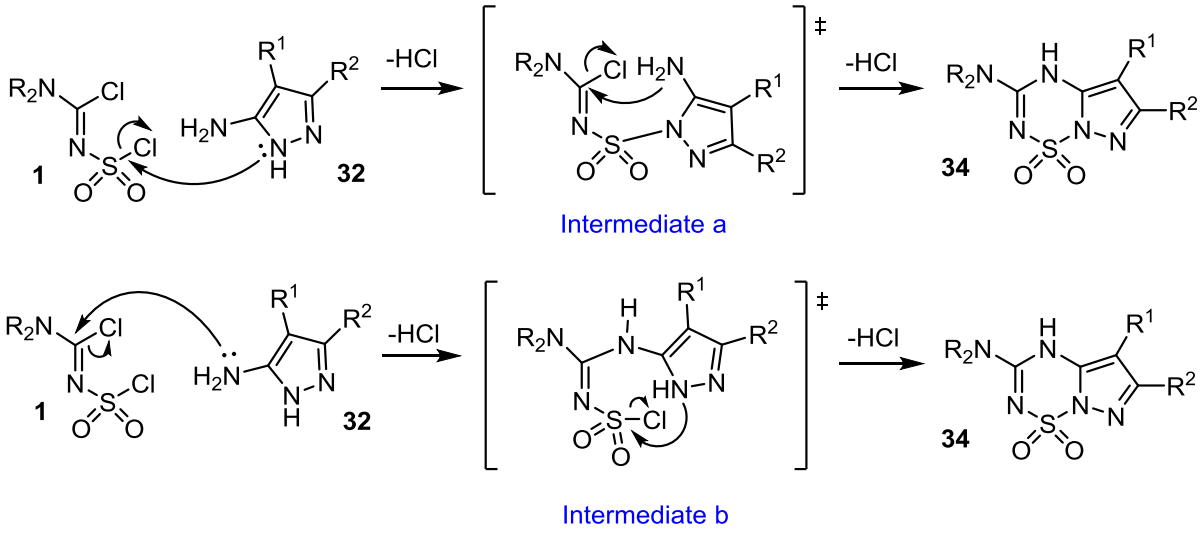


Fig 26. The hydrogen bonding between the amino group and the carbonyl group of the carbethoxy substituent of aminopyrazole **32d.**

2.3.4 Mechanism of formation

The selective formation of fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34** infers that the exocyclic amino group and the pyrazole nitrogen β to this position (N2) are the two most reactive sites towards dielectrophiles **1a-c**. Since there are two electrophilic sites on the formamidine species **1**, there exist two mechanisms that could account for the same product (**Scheme 24**).



Scheme 24: Proposed mechanisms for bidentate condensation to form fused thiatriazines **34**.

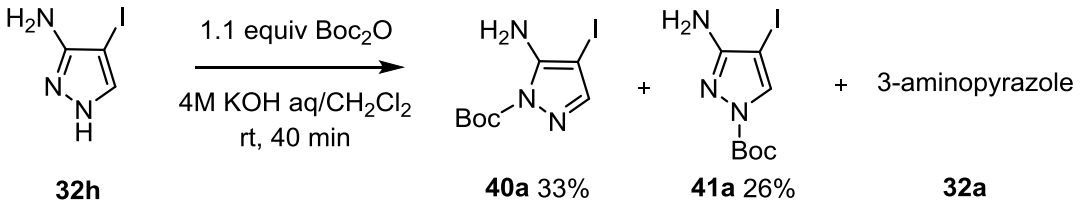
It is assumed that intermediates *a* or *b*, formed initially by a nucleophilic substitution, were not isolatable because the subsequent cyclisation to form a ring fused product was fast. It is important to note here that these intermediate chloride compounds (*a* and *b*) may not have been stable, and so in the cases of reactions where low yields of product were formed and little starting material returned, this could be the cause of decomposition material observed. Given the established greater electrophilicity of the amidinyl chloride in the previous body of work which has been outlined in Chapter 1, the results in this study suggest the exocyclic amine reacts at the amidinyl carbon initially to form intermediate *b*, then the ring nitrogen subsequently cyclises onto the sulfamoyl group to form **34** (**Scheme 24**).

The preference for reactivity at the amidinyl carbon over the sulfamoyl moiety ensured that the product formed was always the fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **34** and not fused pyrazolo[5,1-*c*][1,2,4,6] **35**. The production of a single isomer is attractive given that separation of isomers is not required, meaning purification becomes less time consuming since there is no longer a need for a difficult chromatographic separation or special recrystallisation techniques.

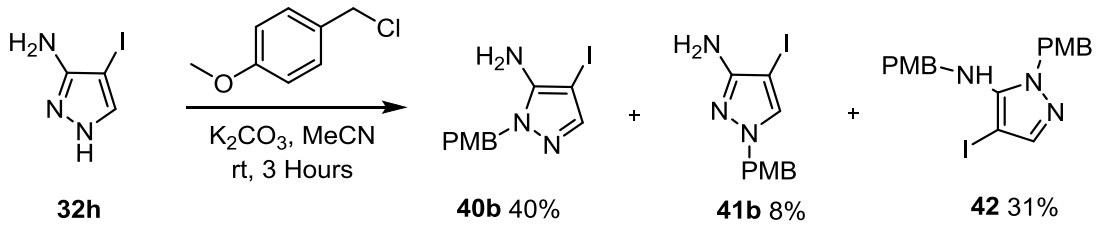
2.3.5 Bulky substituents on the C4 position of the pyrazole ring

Attempted reactions of 3-amino 4-phenyl pyrazole **32g** and also 3-amino 4-iodo pyrazole **32h** with the dichloro compounds **1** did not result in any detectable formation of pyrazolothiatriazines **34**. Several experiments using methods B and D resulted in varying degrees of degradation, most probably due to potential reaction intermediates that had formed which did not cyclise and were not stable. In most cases the starting materials, **32g** and **32h,** were recovered unchanged which suggested that the bulky phenyl- and iodo- substituents caused significant hindrance of the NH_2 substituent, thus preventing reaction at the amidinyl substituent (or the sulfonyl chloride moiety).

The 4-iodo pyrazole **32h** was prone to decomposition by removal of the iodine substituent to return 3-amino pyrazole (**32a**) material when more forceful conditions were applied. Attempts were also made to synthesise derivatives from compound **32h** by initially protecting with di-tert-butyl-dicarbonate (Boc_2O)[86] or by means of a 4-methoxybenzyl chloride protection[101,102] as illustrated in **Scheme 25** and **Scheme 26**.



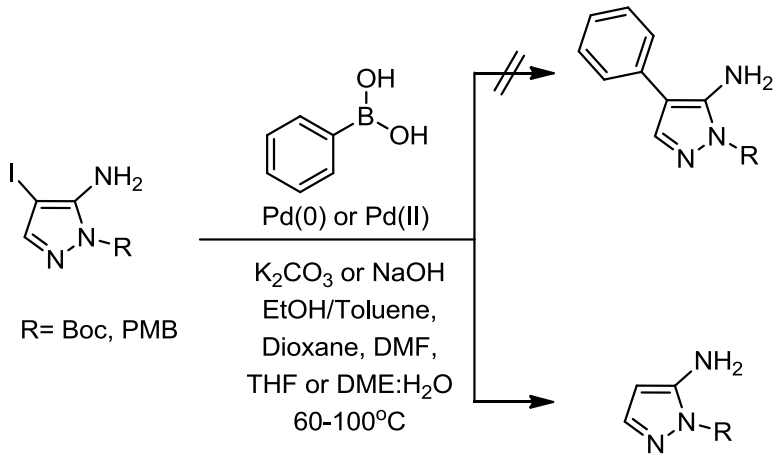
Scheme 25: The protection of compound **32h** with di-tert-butyl-dicarbonate.[1]



Scheme 26: The protection of compound **32h** with 4-methoxybenzyl chloride.

[1] Experimental data for these previously reported products is contained in the reference (Seelen 2003).

These protected materials were to be utilised as synthons to couple to aryl boronic acids following Suzuki conditions,[103,104] and then deprotected in situ to couple to dichlorides **1**. Under the harsh heating and basic conditions required to couple a boronic acid to an aryl halide, the iodine moiety was often lost and the resultant product was 3-amino pyrazole (**32a**) (**Scheme 27**).



Scheme 27: Attempts to form derivatives from the 4-iodopyrazole were unsuccessful due to the propensity for the loss of the iodine moiety.

2.3.6 The influence of reaction conditions

In exploring the reactions of the pyrazoles **32** with dichloride compounds **1**, the most effective methods in the literature were used in this study and included Methods A-D listed in **Table 2**. These methods suggest using 1.3 equivalents of the dielectrophile, as some deterioration of the dichloro compound **1** occurs.

Method A which applied 80°C temperature to a stirred reaction in DMPU was employed successfully in three reactions to form **34e** and **34f** and **34h**. Compared to Method B, which employed an amine base ($^{i}Pr_2NEt$) and no heating, afforded the highest yields of bicyclic ring products **34** with faster reaction completion. Since HCl is produced as a byproduct from the reaction, a weak base such as a tertiary amine is effective in removing this which ensured the reaction solution did not become acidic.

Ring products from unsubstituted pyrazole **32a** could not be collected using DMPU as a solvent as they did not precipitate from solution. The products in these cases were water soluble, hence a mixture of DMPU and water prevented extraction of product with dichloromethane, diethyl ether, ethyl acetate, chloroform, or mixtures thereof.

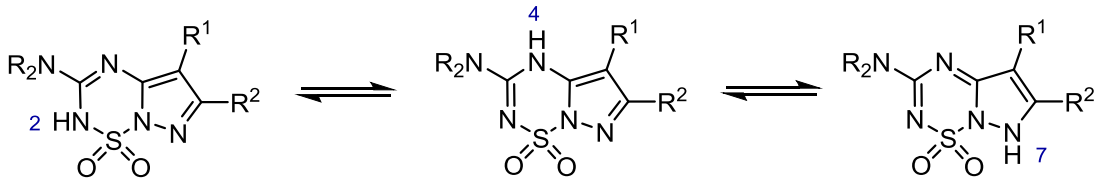
Method C was a biphasic system which utilised potassium hydrogen carbonate base in water and tetra-*n*butylammonium hydrogensulfate ($Bu^n_4NHSO_4$) phase transfer catalyst in benzene. In previous studies, this had proven to afford a different ratio of isomers, [27,28]however the treatment of 3-aminopyrazoles **32** with dielectrophiles **1** was proven to be highly selective and only a single product was generated. Method C was only used in two examples to obtain ring structures **34a** and **34h** because the starting pyrazoles were poorly soluble in both benzene and water, and so the reaction times were long. Method D utilised a different base and solvent system (triethylamine and dichloromethane). An advantage of dichloromethane over DMPU is that it can be readily evaporated under reduced pressure. Method D was most successful for reactions of the unsubstituted pyrazole **32a** to obtain structures **34a, 34b** and **34c.** In cases of other pyrazoles **32**, the starting materials were less soluble in dichloromethane than they were in DMPU. This method proceeded slowly in comparison to methods A and B which is best illustrated again by comparing synthesis of thienyl product **34h** with its formation from other methods (**Table 2**). Methods A and B were deemed the more suitable reaction conditions for those pyrazoles.

2.4 Characteristic NMR behaviour of pyrazolo[1,5-b][1,2,4,6]thiatriazine dioxides

^{1}H and ^{13}C NMR analyses were performed on all novel ring products. DMSO-d_6 was applied as an NMR solvent in the analysis of the fused pyrazolothiatriazines **34** as they were not adequately soluble in other deuterated solvents. ^{1}H NMR spectroscopy was limited by the lack of proton coupling within the heterocyclic core structure. Two isolated spin systems were present in all cases which did not couple to each other; and therefore did not provide evidence of ring size or orientation. The lack of protonated

carbon atoms in the ring structures meant long and short range heteronuclear correlation experiments were not useful. Weak signals due to the amidinyl moiety were present at δ 153-148 ppm from the ^{13}C NMR spectrum of each ring structure; these signals, coupled with those from the dialkylamino group, were the main indicators of product formation.

It is significant to mention here that the fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxide ring system **34** is capable of tautomerism in solution which gives rise to three plausible forms (**Scheme 28**).



Scheme 28: The NH moiety of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **34** can exist at N2, N4, or N7.

A common characteristic of isomers **34** was the presence of an NH proton signal in the ^{1}H NMR spectrum at δ 10.5-11.5 ppm. This seemed to indicate a single tautomer was dominant in DMSO-d_6. The chemical shift of this could correspond to the pyrazole N7 proton (pyrazoles typically contribute signals at ~ δ 12-13 ppm,[79,105]) as opposed to a sulfamide proton that would be expected to provide a signal at ~ δ7-9 ppm.[46,106] The thiatriazine NH tautomer at the N4 position could also give rise to this signal as there is limited literature precedent for [1,2,4,6]thiatriazines which are capable of forming an NH tautomer at this position.

The ^{1}H NMR spectra of compounds **34** offered a common peculiarity for the signals attributable to the protons connected to the exocyclic amino groups. These sets of protons gave rise to broad signals at 26°C (**Figure 27**), and similarly in the ^{13}C NMR the corresponding carbon signals were very broad and difficult to distinguish (**Figure 28**).

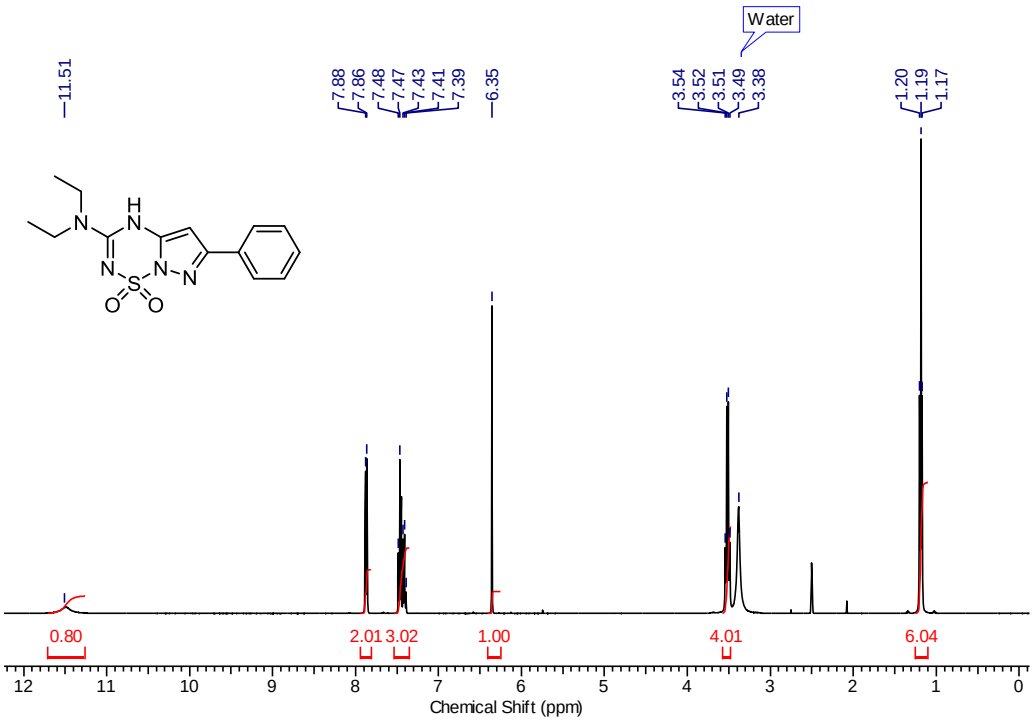


Fig 27. ^{1}H NMR spectrum of **34e** (DMSO-d^{6}) at 26°C

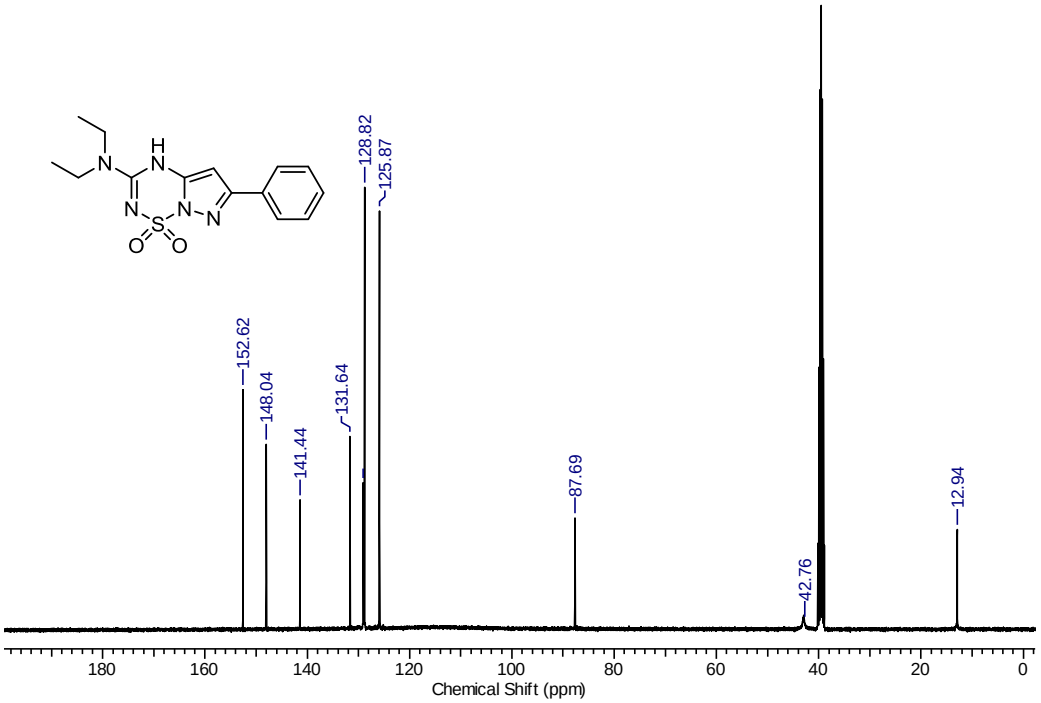


Fig 28. ^{13}C NMR spectrum of **34e** (DMSO-d^{6}) at 26°C

The characteristic NMR behaviour of dichlorides **1** has been discussed in Chapter 1, and similarly fused pyrazolothiatriazines **34** could feasibly hold the same resonance contributors. The compounds **34** may exist as two rotamers that interconvert slowly on the NMR timescale at the amino substituent, which is best illustrated by the zwitterionic contributions for the formamidinyl moiety (**Figure 29**). Variable temperature NMR studies of related systems[22] resulted in peak broadening or collapsing into two separate sets of peaks at low temperatures, and sharpening to single peaks at higher temperatures.

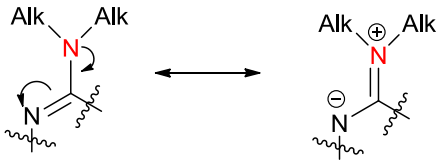


Fig 29. Restricted rotation around the tertiary amine heterocyclic products obtained from sulfamoyl-formamidines **1**

These alkyl substituents on products **34** all provided single signals in the ^{1}H NMR, hence the zwitterionic resonance contribution would not have been as great in comparison with some other ring-fused products derived from dichlorides **1**. The signals observed were presumably an average of the two non-identical chemical environments.

2.5 General physical properties of the pyrazolothiatriazine ring system

2.5.1 Stability

As mentioned in Section 2.2.1, compounds **34a-l** were obtained as stable, crystalline solids. The materials **34a** and **34b** and **34c** in **Figure 30** were the only examples of ring products which were not stable in air at room temperature. These were isolated as

crystalline materials initially and gradually formed amorphous materials with coloured impurities over time. No signs of degradation or transformation by means of acid were ever observed during acidic workup, or from the amine bases which were utilised in their formation. The ring products **34a**, **34b** and **34c** were stored at temperatures below 5°C under an atmosphere of nitrogen to slow this process.

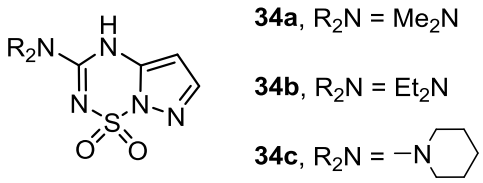
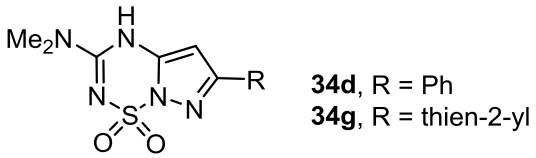


Fig 30. The pyrazolo[1,5-*b*][1,2,4,6]thiatriazines synthesised from unsubstituted 3-amino pyrazole **32a**.

All ring structures **34** were high melting point solids. Melting point analyses in many cases resulted in decomposition of the material at temperatures between 220-300°C. This indicates a reasonable level of thermal stability of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system **34** which indicated that they could be heated in organic solvents for subsequent reactions with little need for thermal degradation concerns.

2.5.2 Solubility and polarity

Fused pyrazolothiatriazines **34d** and **34g** in **Figure 31** were highly insoluble in water, non-polar solvents such as hexanes, and solvents of medium polarity such as diethyl ether. They could be dissolved in mixtures of methanol and dichloromethane, or methanol and chloroform. The high yields of these materials could best be explained by this inherent lack of solubility which meant that they readily precipitated upon acidification of DMPU and water mixtures. This is best compared with the lower yields of products **34a, 34b, 34c** and **34j** which were water soluble; and hence a different reaction solvent was necessary. Ring structures **34** overall were very polar compounds which were most soluble in polar solvents such as methanol, DMF or DMSO.



34d, R = Ph
34g, R = thien-2-yl

Fig 31. The pyrazolo[1,5-*b*][1,2,4,6]thiatriazines synthesised from 5-substituted 3-aminopyrazoles **32b** and **32c**.

In the cases of phenyl and thienyl substituted ring products (**34d**, **34e**, **34f**, **34g** and **34h**), material precipitated after the addition of aqueous hydrochloric acid to DMPU mixtures (methods A and B) which was difficult to dissolve in many common chromatography solvents. Recrystallisation techniques were preferred; however in cases where chromatography was essential, a dry-loading method of silica gel chromatography was often used. Most compounds were purified by silica gel chromatography with 10-15% methanol in dichloromethane or chloroform as the mobile phase. Other mobile phases were trialled, although the only other mixtures giving adequate separation were diethyl ether and ethyl acetate in dichloromethane for ring structures from pyrazole **32d**.

Materials which contained a diethylamino substituent (**34b**, **34e**, **34h**) were comparatively more soluble in methanol, acetonitrile, chloroform and dichloromethane than the analogous materials which contained the dimethylamino moiety (**34a**, **34d**, **34g**). This direct comparison demonstrated that although the diethylamino derived products were generated in lower yields, the added benefits of higher solubility and ease of handling could mean that they would be more amenable as synthons due to a larger pool of solvents to choose from. Generally, the products arising from chloride **1c** had similar solubility issues to **1a** derived products.

2.5.3 Miscellaneous properties

All products **34** and starting materials **32** were UV active and could be visualised under UV light; however Ninhydrin dip offered the advantage in that coloured complexes

resulted. Compounds spread on the silica to give a 'streaky' appearance or the impression of multiple spots. A drop of water or 5% aqueous ammonia in the mobile phase to reduce adhesion to the polar silica gel often helped tighten spots when TLC monitoring.

2.6 Bis-adducts and uncyclised dichlorides

2.6.1 Two bis-adducts – characterisation by X-ray crystallography

Pyrazolothiatriazine **34i** was obtained from pyrazole **32d** by the reaction with dichloride **1a** from methods B and D in modest yields (28% and 26% respectively). The same starting materials were combined using method C which produced a material that appeared by NMR to have introduced two pyrazole subunits into its structure (**Scheme 29**). Mass spectrometry data indicated that two pyrazole units had added to the dichloride compound at each of the electrophilic sites; however the precise connectivity of each pyrazole ring to the sulfamoyl and amidinyl groups was unclear by NMR. The structure of bis-fused adduct **43a** (**Figure 32**) was determined by means of X-ray crystallography.

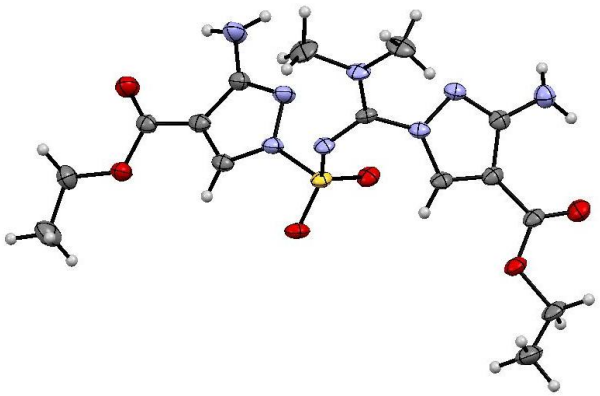
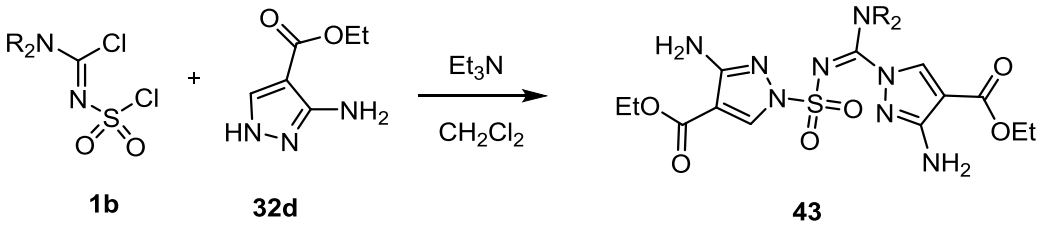


Fig 32. ORTEP diagram of **43a**.



Scheme 29: The reaction of pyrazole **23d** with dichlorides **1** provided bis adducts **43**.

The X-ray crystal structure of **43a** obtained showed that ring nitrogen N1 had reacted at each electrophilic site of dichloride **1a**. HMBC NMR experiments on **43b** were consistent with the assigned structure. The preferential nucleophilic attack of the pyrazole nitrogen as opposed to the exocyclic amino group could be explained by the electron withdrawing effect of the ester substituent. Apparently, the intermediate compound from the reaction of N1 with the dielectrophile was incapable of cyclising because the other ring nitrogen N2 was not sufficiently reactive. Instead, another aminopyrazole molecule reacted at the remaining electrophilic site to obtain the bis-adduct **43a** in 24% yield. The formation of a 5-membered ring from cyclisation at N2 was also not likely to be a favourable process compared to cyclisation to form 6-membered rings, and this was generally observed in the pyrazol-3one work.[28]

Table 3: The formation of bis-adducts **5** arising from reaction of 4-carbethoxy 3-aminopyrazole **32d** at N1.

R_2N	Method*	**Product**	Yield
Me_2N (**1b**)	C	**43a**$^{\#}$	24
Et_2N (**1b**)	B	**43b**	54

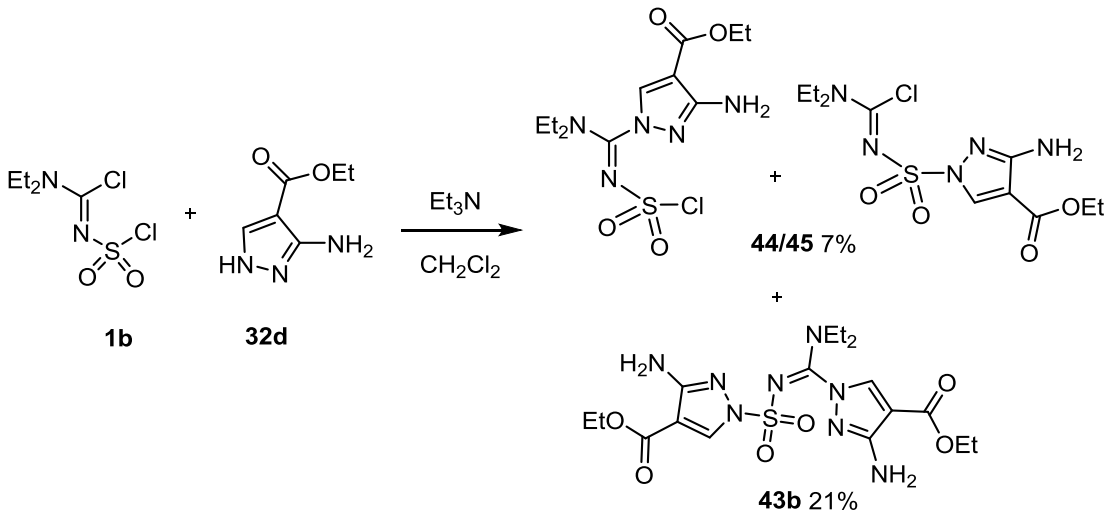
*Method B = $^{i}Pr_2NEt$/DMPU; Method C = $KHCO_3$(aq.)/C_6H_6/$Bu^{n}_4NHSO_4$

Interestingly, compound **43b**, but none of the ring-fused compound **34**, was obtained from the diethylamino-dichloro compound **1b** using methods B and D. Method C may have acted to enhance the nucleophilicity of ring nitrogen N1, or reduce the relative

nucleophilicity of the exocyclic amino substituent to prevent the reaction which forms product **34i**.

2.6.2 Dilution effects (and formation of mono adducts)

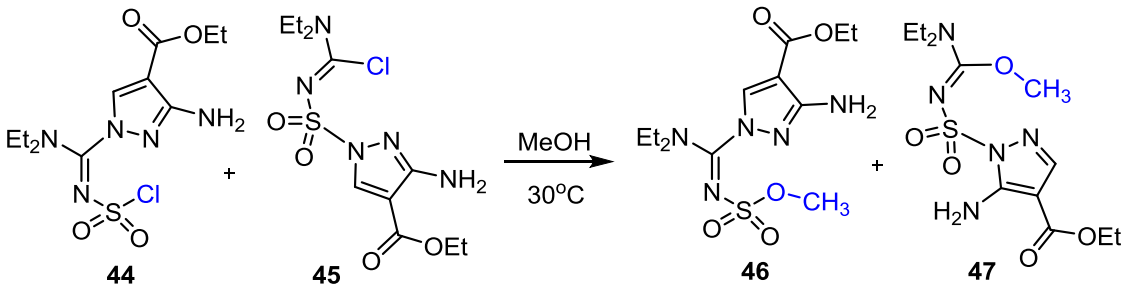
Attempts to obtain a cyclised product from dichloride **1b** and pyrazole **32d** were unsuccessful. A method D experiment between compounds **1b** and **32d** which used ten-fold dilution was conducted with the aim of reducing bis-pyrazole product formation and facilitating ring-fused product formation. Bis-adduct **43b** was obtained in 21% yield along with two "intermediate chloride" products **44** and **45** (in a ratio of 5:1) isolated in minor quantities (7% total) (**Scheme 30**). The information obtained from the ^{1}H NMR spectrum such as chemical shift and proton integration was not suggestive of an uncyclised adduct, however the mass spectrum obtained indicated that these materials contained chlorine.



Scheme 30: The formation of acid chlorides **44** and **45** along with bis-adduct **43b**.

These results suggest that, in addition to reduced nucleophilicity due to the electron withdrawing group at C4, steric hindrance also retards reaction of the exocyclic amino group with the dielectrophile, especially in the case of diethylamino-substituted **1b**. Compounds **44** and **45** were not stable in air for long periods of time and proved very

difficult to separate by chromatography. The chloride materials appeared to be decomposing in air (or from the silica gel) and so instead, a stable derivative was envisioned. A mixture of **44** and **45** in methanol was gently heated with the aim of producing stable, crystalline products which could be purified by chromatography and would be potentially more amenable to structure determination by X-ray crystallography. A mixture of the corresponding methoxy derivatives **46** and **47** was obtained (in a ratio of 6:1), which also could not be adequately separated, either chromatographically or by crystallization (**Scheme 31**).



Scheme 31: Isomers **44** and **45** from incomplete cyclisation of **32d** and **1b** reacted with methanol to form **46** and **47**.

High resolution mass spectrometry confirmed the molecular formula of **46** and **47** more adequately as they were stable materials. No evidence of formation of the cyclised ring product **34i** was found.

2.7 Conclusions

The versatility of the 1,3-dielectrophilic reagents **1** was demonstrated by a series of reactions with readily available 3-aminopyrazoles **32**. These selectively furnished representatives of the previously unreported pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system, compounds **34,** with no indication of other isomeric ring products having formed. The regioselectivity of this reaction was proposed to have resulted from the initial nucleophilic attack of the exocyclic amino substituent of the pyrazole onto the

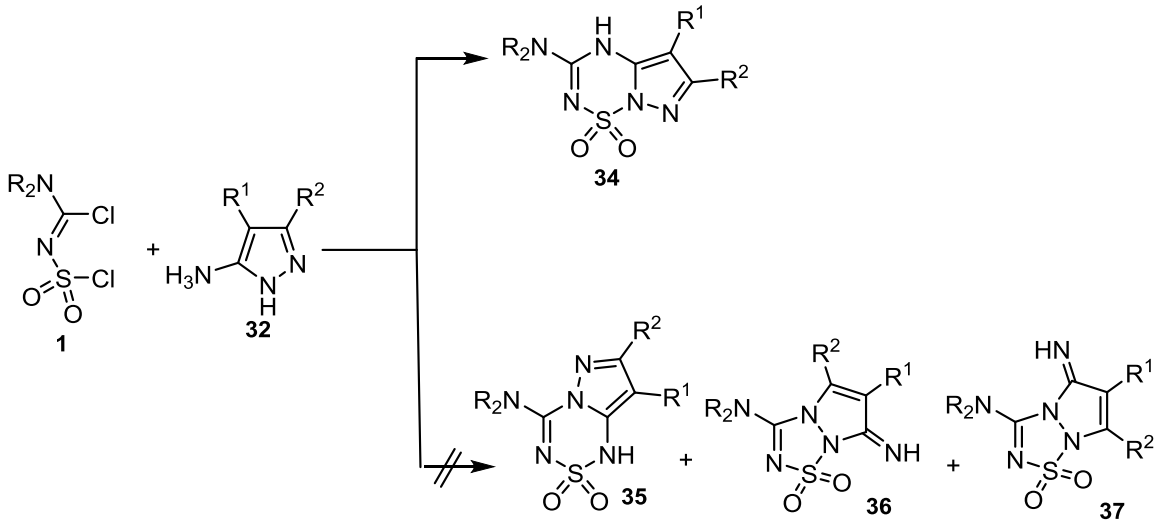
amidinyl moiety of the dichloride, followed by rapid cyclisation onto the sulfamoyl chloride. Substrate substitution patterns appeared to affect the yields of this condensation process, particularly when the pyrazole ring was substituted at the 4-position. In some circumstances, the 4-ester substituted pyrazole **32d** gave rise to bis-adducts **43** or intermediate chlorides **44/45** by reaction at the ring nitrogen N1 instead.

CHAPTER 3: REACTIONS OF *N,N*-DIALKYL *N'*-CHLOROSULFONYL CHLOROFORMAMIDINES WITH 1-SUBSTITUTED 5-AMINO PYRAZOLES

3.1 Introductory remarks

3.1.1 Modified aminopyrazoles in the pursuit of an isomeric core structure to the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides

The reaction between *N,N*-dialkyl *N'*-chlorosulfonyl chloroformamidines **1** and *N*-unsubstituted 3-aminopyrazoles **32** described in Chapter 2 delivered the unprecedented pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring system **34** as the sole isolated product under a variety of conditions. Four isomeric products were possible from the three nucleophilic nitrogen atoms; however, compounds **35**, **36** and **37** were never observed (**Scheme 32**).



Scheme 32: The regioselective condensation reaction between 3-aminopyrazoles **32** and dichlorides **1**.

The reaction of dichlorides **1** with 1-substituted 5-aminopyrazoles **33**, in place of simple 3-aminopyrazoles **32**, was investigated with the aim of varying the regiochemical outcome of the multi-site reaction to a dinucleophilic substitution involving C4 and the exocyclic amino group.

3.1.2 The N-substituted 5-amino pyrazole system as a reactive 1,3-CCN dinucleophile

The 1-substituted 5-aminopyrazoles **33** differ from simple 3-aminopyrazoles **32** in that the pyrazole ring nitrogen N1 is prevented from acting as a nucleophile due to an *N*-alkyl or *N*-aryl substituent (**Figure 33**).

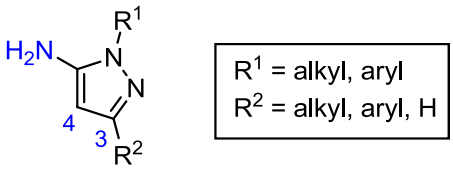
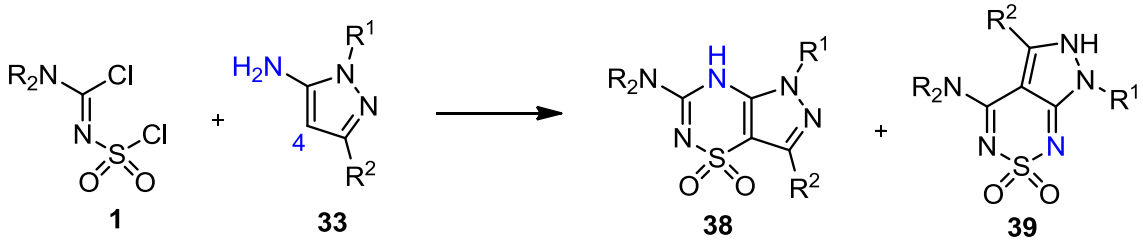


Fig 33. The 1-substituted 5-aminopyrazole structure **33**.

1-Substituted 5-aminopyrazoles **33** are conventionally synthesised following the methods outlined in Section 1.3; most of which involve the condensation of mono-substituted hydrazines with either α,β-unsaturated nitriles or 3-oxoalkanenitriles.[70–72,107] As aforementioned, electrophilic substitution at either ring nitrogen N1 or N2 is unfavourable, particularly in comparison to the exocyclic amine substituent, and produces a quaternary salt.[79,98] These aminopyrazolium salts upon heating undergo migration from the nuclear to the extra-nuclear nitrogen.[98] Furthermore, it has been well documented that the 5-amino group is the most nucleophilic site on the molecule, followed by C4 under selected conditions.[69,71,79] Heterocyclization of 1-substituted 5-aminopyrazoles **33** with β-ketoesters occurs by initial reaction at the exoxylic amino group in most reported cases.[108,109] Tabak, Grandberg and Kost[110,111] reported the condensation of acetoacetic esters with aminopyrazoles to provide pyrazolo[3,4-*b*]pyridinones from reaction of either electrophilic site at the NH_2 group initially, followed by cyclisation into the C4 substituent. 1-Substituted 5-

aminopyrazoles have been reacted with pyrazolidines at the C4 position in boiling benzene; whereas in the presence of an excess of alumina in benzene at 60°C, the sole product isolated was substituted at the amino group.[112]

Two cyclised products are conceivable via the action of compounds **33** as 1,3-CCN dinucleophiles towards dichlorides **1**: the fused pyrazolo[3,4-*e*][1,2,5]thiadiazines **38** and the fused pyrazolo[3,4-*c*][1,2,6]thiadiazines **39** (**Scheme 33**).



Scheme 33: The potential reaction of 1-substituted 5-amino pyrazoles **33** with dichlorides **1**.

These two products **38** and **39** are derivatives of a very rare ring system (only one derivative has been reported from a six-step synthesis[113]) and represent the first examples, in the CSIRO/Flinders body of work, of a fused ring system produced from reactions of dichlorides **1** with 1,3-CCN dinucleophiles. Compounds **38** and **39** are also isomeric in core structure to the fused pyrazolothiatriazines **34** and **35**. A range of substrates **33** were selected to investigate electronic and steric effects of substituents on C3 and N1 on the nucleophilicity of the NH_2 group and C4.

3.2 Synthesis of the novel fused pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1 dioxides

3.2.1 General methods and substrate synthesis

A set of commercially available 1-substituted 5-aminopyrazoles **33** (**Figure 34**) with various substitution patterns were selected to investigate the overall regioselectivity observed in condensations with dichlorides **1**.

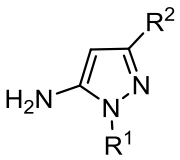
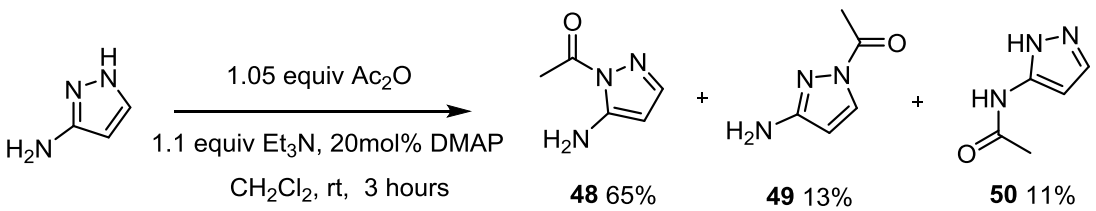


Fig 34. Pyrazoles **33a-f**

R^1	R^2	
Me	H	**33a**
Me	Me	**33b**
Ph	Me	**33c**
Me	Ph	**33d**
Me		**33e**
Me	*t*-butyl	**33f**

Table 4: The 1-substituted 3-amino pyrazole **33** substrates selected for this study.

A sample of 1-acetyl-5-amino pyrazole (**48**) was synthesised from 3-aminopyrazole in order to establish the effect of an electron withdrawing substituent at the pyrazole nitrogen N1 upon the condensation reaction (**Scheme 34**). Isomers **49** and **50** were also isolated in minor quantities. Structural assignments were made based on literature reports.[114]

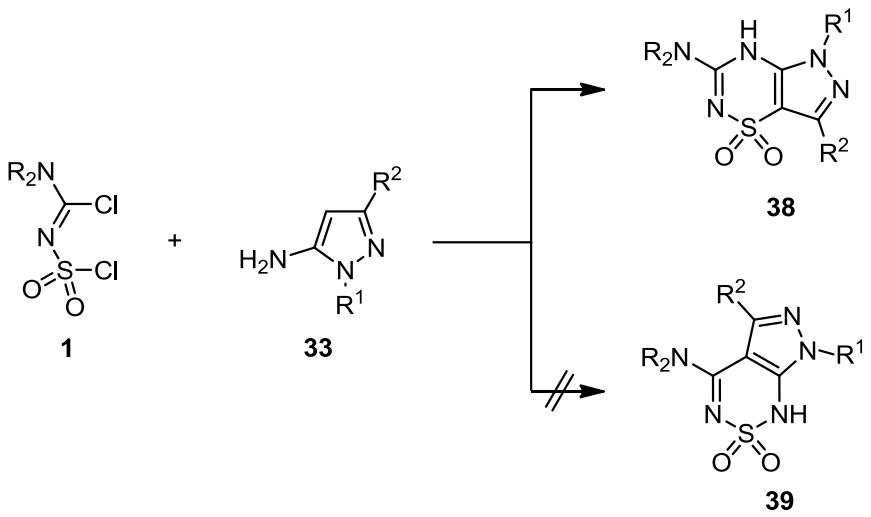


Scheme 34: The synthesis of *N*-acetyl pyrazoles **48-50**.

The reaction of dichlorides **1** with 1-alkyl 5-aminopyrazoles **33** was examined under a variety of conditions:- (A) heating at 80°C in a polar, aprotic solvent, 1,3-dimethyl-3,4,5,6-tetrahydro-2(*1H*)-pyrimidinone (dimethylpropylene urea, DMPU); (B) in DMPU at ambient temperature with an organic base, *N*,*N*-diisopropylethylamine (Hünig's base); (C) and (D) homogeneous systems using triethylamine in either dichloromethane or acetonitrile (see **Table 5**).

3.2.2 Summary of results

Syntheses following methods A-D all afforded only the pyrazolo[3,4-*e*][1,2,4]thiadiazines **38** (**Scheme 35**, **Table 5**) as stable, colourless, crystalline solids. No other ring-fused products, such as the isomeric pyrazolo[3,4-c][1,2,6]thiadiazines **39**, were isolated. The structures of compounds **38** were confirmed by X-ray crystallography of two representatives (see Chapter 8) and represent new derivatives of a very rare ring system. They are the first examples in the CSIRO/Flinders work of a fused-ring system prepared from reactions of dichlorides **1** with 1,3-CCN dinucleophiles, although similar compounds were reported by other research groups.[12,113]



Scheme 35: Selective formation of pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides **38**.

Table 5: Synthesis of pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides **38**.

R_2N	R^1	R^2	Method	Product(s)	Yield [%]
Me_2N (**1a**)	Me	H (**33a**)	C	**38a**	78
Me_2N (**1a**)	Me	Me (**33b**)	A	**38b**[#]	60
			B	**38b**[#]	71
Et_2N (**1b**)	Me	Me (**33b**)	D	**38c**	13
 (**1c**)	Me	Me (**33b**)	B	**38d**	15
			C	**38d**	36
Me_2N (**1a**)	Ph	Me (**33c**)	B	**38e**	14
			C	**38e**	42
Me_2N (**1a**)	Me	Ph (**33d**)	B	**38f**	44
 (**1c**)	Me	Ph (**33d**)	D	**38g**	49
Me_2N (**1a**)	Me	Thien-2-yl (**33e**)	B	**38h**	44
			D	**38h**	70
Et_2N (**1b**)	Me	Thien-2-yl (**33e**)	D	**38i**	11
Me_2N (**1a**)	Me	t-butyl (**33f**)	B	**38j**[#]	12

	D	**38j**[#]	33

[#]X-ray crystal structure obtained. Method A = DMPU/80°C; Method B = $Pr^{i}_{2}NEt$/DMPU; Method C = Et_3N/CH_2Cl_2/rt; Method D = Et_3N/MeCN/rt or 50°C.

Treatment of pyrazoles **33b** and **33d** with dichloride **1b** did not afford a precipitate with methods A or B, and efforts to isolate product from the mother liquor were not successful. The attempted condensation between dichloride **1c** and pyrazole **33b** using method A gave rise to mixtures of possible chloride intermediates by NMR studies; however these could not be adequately purified. In the case of compounds **38c, 38d, 38e, 38g** and **38i** when using methods D and E, complete conversion did not ensue and the starting aminopyrazoles were recovered in yields of 37%, 30%, 25%, 19%, and 42%, respectively.

Generally, the highest yields were afforded from pyrazoles **33a** and **33b** containing methyl substituents at the 1-position and either a hydrogen or a methyl substituent on the 3-position of the pyrazole ring; in other words, substrates bearing substituents with little steric bulk. Treatment of **33a** with dichloride **1a** using method C supplied product **38a** in 78% yield whereas **33d** under the same conditions only afforded 42% of ring product **38f**. The synthesis of **38f** and **38h** from method B (44% yields) can also be compared to that of **38b** from the same method (71% yield).

The N-phenyl substituted compound **33c** was used as representative substrate containing steric bulk at the ring nitrogen. Reaction of this material with the methyl dichloride **1a** with method B afforded a ring product in 22% yield (**38c**). This contrasts with the formation of **38b** (71% yield) under the same conditions from the pyrazole **33b** which contains a methyl group at the pyrazole nitrogen.

The treatment of 1-acetyl 5-amino pyrazole (**48**) with dichloride **1a** under conditions from methods B, C or D did not result in formation of a ring product. The pyrazole starting material was collected from methods C and D unchanged in 62% and 48% yields respectively. The electron withdrawing effect of the acyl group likely reduced the

nucleophilicity of the exocyclic amine to such an extent that no reaction occurred under the conditions employed. There is also a hydrogen bonding effect between the carbonyl group and the adjacent amine which further inhibits reactivity, similar to the effect of the carbethoxy substituent on C4 of 3-aminopyrazole outlined in Chapter 2 (**Figure 34**).[100]

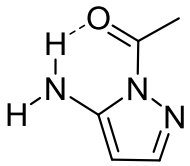


Fig 34. The intramolecular hydrogen bond between the exocyclic amino substituent and the carbonyl group of 1-acetyl 5-aminopyrazole.

A comparison of product yields from each dichloride precursor indicated that the dimethylamino bis-electrophile (**1a**) afforded the highest yields of isolated products in comparison to those containing a diethylamino group (from **1b**) or a piperidine ring (from **1c**). A similar observation was made during the work described in Chapter 2. This phenomenon cannot be attributed solely to the comparatively poorer solubility of products obtained from the dimethylamino dichloride **1a** as methods where the products could be readily separated from the reaction mixture (methods C and D) were also employed in these cases. Another probable contributing factor could be that the dichlorides **1b** and **1c** were not as reactive as **1a** towards the dinucleophile, particularly in the less polar solvents (acetonitrile and dichloromethane) compared to the polar, aprotic solvent DMPU. However, the ease of isolation due to solubility still appears to be the most significant causative factor of the general yield differences.

Structural elucidation of compounds **38** was achieved via X-ray crystallography of the two representative compounds **38b** and **38c** shown in **Figure 35.** Additional details of the crystallographic studies are outlined in Chapter 8.

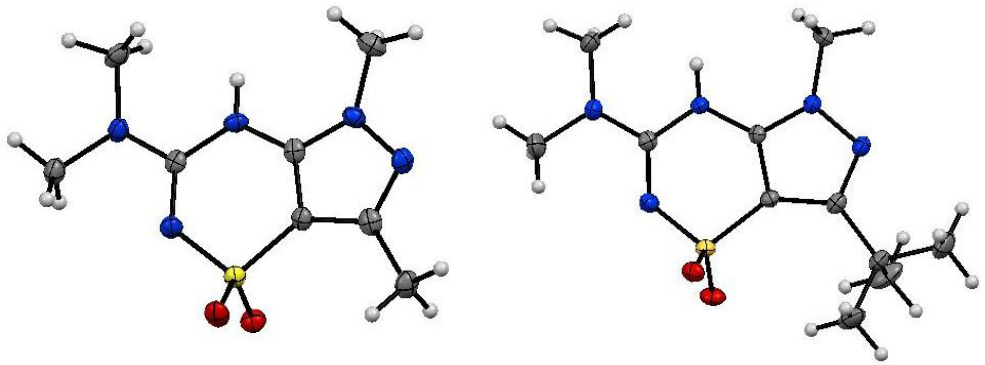
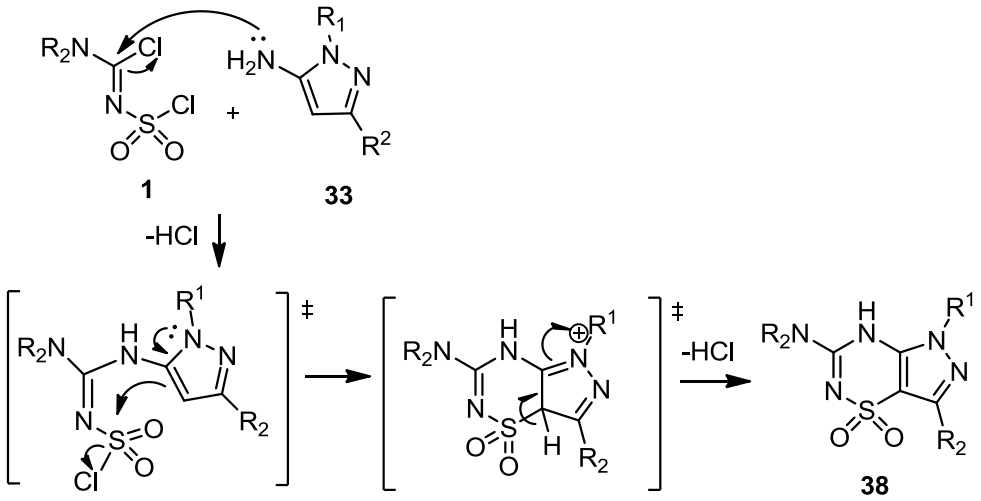


Fig 35. ORTEP diagrams of **38b** and **38j**.

3.2.3 Mechanism of formation

Based on earlier work with the dichlorides **1**,[23,27–29] carbon nucleophiles are most likely to react at the sulfur atom and the more reactive (and less hindered) exocyclic amine would be expected to react at the more electrophilic carbon atom. The selective formation of compounds **38** is consistent with this general trend, confirming that the major reaction pathway involves the nucleophilic attack of the exocyclic amino group at the amidinyl carbon followed by cyclisation of the pyrazole carbon onto the sulfonyl moiety (**Scheme 36**).



Scheme 36: The proposed mechanism of formation of fused pyrazolo-thiadiazines **38**.

Others have reported that when a ring nitrogen of an amino pyrazole is substituted, the exocyclic NH_2 group typically is the first to react because it is a stronger nucleophile and is less hindered than the nucleophilic carbon atom C4.[79,98,115] Regioselectivity is nevertheless dependant on factors including the nature of the alkyl group on the pyrazole ring, the specific electrophile and, to an extent, the conditions employed.[108,116] Only one of the two possible reaction pathways was observed during this study, which confirms the greater nucleophilicity of the amino group of **33** compared to C4 towards dichlorides **1**.

3.2.4 The influence of reaction conditions

The outcomes from methods A and B were comparable for the synthesis of pyrazolo-thiadiazine **38b** (60 and 71% respectively); however, attempts to generate other ring products from DMPU-based methods were often unsuccessful and we were unable to isolate any products from the mother liquor. Reaction of the pyrazole **33c** with dichloride **1a** afforded ring product **38c** in 22% yield from method B. The same starting materials were reacted in dichloromethane (method C) which afforded a 36% yield of the same product. This increase in yield could be due to the poorer solubility of the product in dichloromethane, from which **38c** precipitates as it forms, leaving the starting materials in solution. Moreover, it might be simply that the product can be isolated more readily from the more volatile solvent.

Use of Method D, with acetonitrile as solvent, proved to be more effective for substrates containing bulky substituents on the 3-position of the pyrazole ring. Nucleophilic substitution reactions are generally improved in polar aprotic solvents (a class which includes acetonitrile); however the ease of product isolation is important to consider as well as the rate and quality of the reaction. DMPU appeared to be a powerful solvent and products often did not readily precipitate from aqueous DMPU mixtures. Dichloromethane is a less powerful and more volatile solvent, thus product isolation was much easier. Dichloromethane as a reaction solvent generally provided lower yields than the polar aprotic acetonitrile (method C compared to method D) since the less polar

solvent would not dissolve the starting materials as effectively. Acetonitrile delivered improved yields when used as a solvent compared to DMPU despite the higher solubilising power of the DMPU. This indicated that the lower yields from methods A and B were owing to the lack of precipitation from the aqueous product mixture. Acetonitrile presented a good balance between a rate-enhancing polar aprotic solvent and an easily removable solvent, allowing isolation of products in acceptable yields.

3.2.5 Chemical and physical properties of the new ring system

The pyrazolo-thiadiazine ring products **38** were obtained as stable, colourless crystalline solids with no signs of decomposition at room temperature in air. In contrast to the isomeric pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34**, compounds **38** did not become contaminated with coloured impurities. Dioxides **38** were all high melting point solids which decomposed at temperatures >250°C with the exception of **38g** which had a decomposition point at ~191°C. This indicated a practical level of thermal stability of the pyrazolo[3,4-*e*][1,2,4]thiadiazine ring system **38** suggesting that they could be heated in organic solvents for subsequent reactions. No signs of acid, light, or thermal sensitivity under the conditions employed were observed.

Solubility became the biggest issue when handling compounds **38** which were less soluble in organic solvents such as DMSO, chloroform, methanol and acetone than the pyrazolothiatriazines **34**. The bicyclic ring products **38** were very polar, with mobile phases of up to 20% methanol in dichloromethane required for elution during silica gel chromatographic purification. Compounds **38** could not be dissolved in any other common chromatography solvents such as ethyl acetate, diethyl ether, or light petroleum. Fortunately, many of the poorly soluble compounds containing dimethylamino groups were isolated by precipitation and recrystallisation. When chromatographic separation was necessary, a dry-loading method for silica gel chromatography was most often used. Compounds **38** tended to spread over the silica gel to give a ‘streaky’ appearance and so 1% aqueous ammonia was often added to the mobile phase during TLC analysis.

Compounds which contained a diethylamino substituent (**38c** and **38i**) were comparatively more soluble in methanol, acetonitrile, chloroform and dichloromethane than the analogous compounds bearing a dimethylamino moiety (**38b** and **38h**). Due to isolation difficulties, only two examples of compounds **38** containing the diethylamino substituent were synthesised, along with only two examples containing the piperidine ring at this position (**38d** and **38g**).

All products **38** and starting materials **33** were UV active and could be visualised under UV light; however in contrast to the isomeric dioxides **34**, the ninhydrin reagent did not react with the ring products **38** to form coloured complexes. This worked as an advantage since the pyrazole starting materials were highly reactive with the ninhydrin reagent to give coloured yellow and orange complexes.

3.2.6 Characteristic NMR behaviour

^{1}H and ^{13}C NMR spectroscopic analyses were performed on all novel ring products **38**. The usefulness of ^{1}H NMR spectroscopy in product structure elucidation was limited by the low number of hydrogen atoms within the heterocyclic core structure. In most cases, there was only a single proton directly attached to a ring atom, an NH. Heteronuclear correlation experiments were not informative due to a lack of protonated carbon atoms in the ring structures. Low intensity signals in the ^{13}C NMR spectrum of each ring structure were present at δ 150-147 ppm. These were assigned to the amidinyl carbon which has a slow relaxation time on the NMR timescale. The amidinyl signals and those from the dialkylamino group were the main indicators of product formation.

The fused pyrazolo [3,4-*e*][1,2,4]thiadiazine dioxide ring system **38** is capable of tautomerism in solution which gives rise to three plausible forms (**Figure 36**).

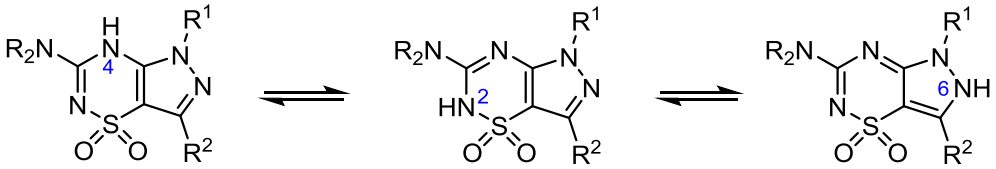


Fig 36. The NH moiety of the pyrazolo**[**3,4-*e*][1,2,4]thiatriazine **38** can exist at N4, N2, or N6.

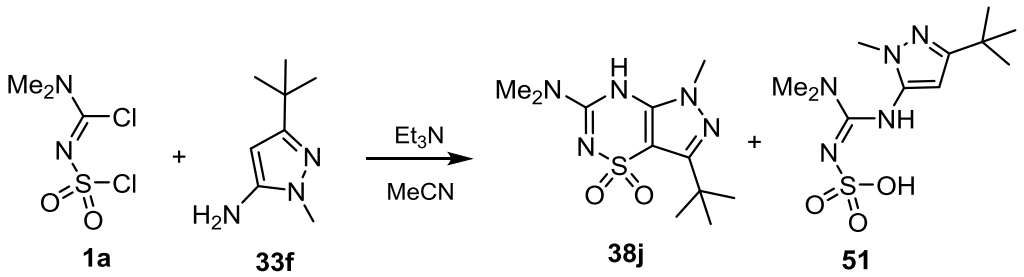
A common characteristic of ring products **38** was the presence of an NH signal in the ^{1}H NMR spectrum at δ 10.8-10.4 ppm. This seemed to indicate a single tautomer was dominant in DMSO-d_6. The chemical shift of this could correspond to the pyrazole N6 proton (typically ~ δ 12-13 ppm,[79,105]), the N2-sulfonamide proton (typically ~ δ 7-9 ppm[46,106]), or the thiatriazine NH tautomer at the N4 position which has no literature precedent. Identification of the major tautomer based on NMR experiments alone was speculative, although the observation of a single signal meant that one tautomer dominated.

The ^{1}H NMR spectra of compounds **38** presented broad signals for the hydrogens flanking the dialkylamino substituent. This occurrence has been attributed to the partial double bond character and restricted rotation of this functionality (discussed in chapters 1 and 2). The corresponding carbon signals were very broad and difficult to discern in the ^{13}C NMR spectra of compounds **38**. The restricted rotation effects were generally more pronounced than with the pyrazolothiatriazines **34** and the ^{1}H NMR and ^{13}C NMR signals owing to the dialkyl substituents were broad, often disappearing into the baseline. These effects combined with limited solubility made ^{13}C NMR spectra difficult to collect. DMSO-d_6 was applied as an NMR solvent in the analysis of the fused pyrazolothiatriazines **38** as they were not adequately soluble in other deuterated solvents, however DMSO is known to produce broader signals due to the viscosity of the solvent itself. The solubility of compounds **38** was markedly lower than the pyrazolothiatriazines **34** which meant that NMR samples usually contained no more than 5mg of analyte, despite the abundance of available sample. Higher temperature NMR experiments (36-56^{o}C) were needed to produce adequate ^{13}C NMR spectra, which also enabled a higher concentration of solute and faster rate averaging signal.

3.3 Sulfamic acid side products

Many of the reactions of dichlorides **1** with pyrazoles **33** furnished lower yields of ring products than analogous experiments with unsubstituted pyrazoles **32**. Section 2.2.1 mentions the possible formation of "uncyclised chloride intermediates" which could account for the complex mixture of compounds often observed during NMR analyses of crude products from methods C and D. Any uncyclised chloride compounds that may have formed could conceivably undergo side reactions, decompose, or hydrolyse upon workup, and hence they were difficult to isolate. In the previous chapter characterisation of an isomeric mixture of uncyclised chloride intermediates (**44:** an amidinyl chloride, and **45:** a sulfamoyl chloride) was described.[99]

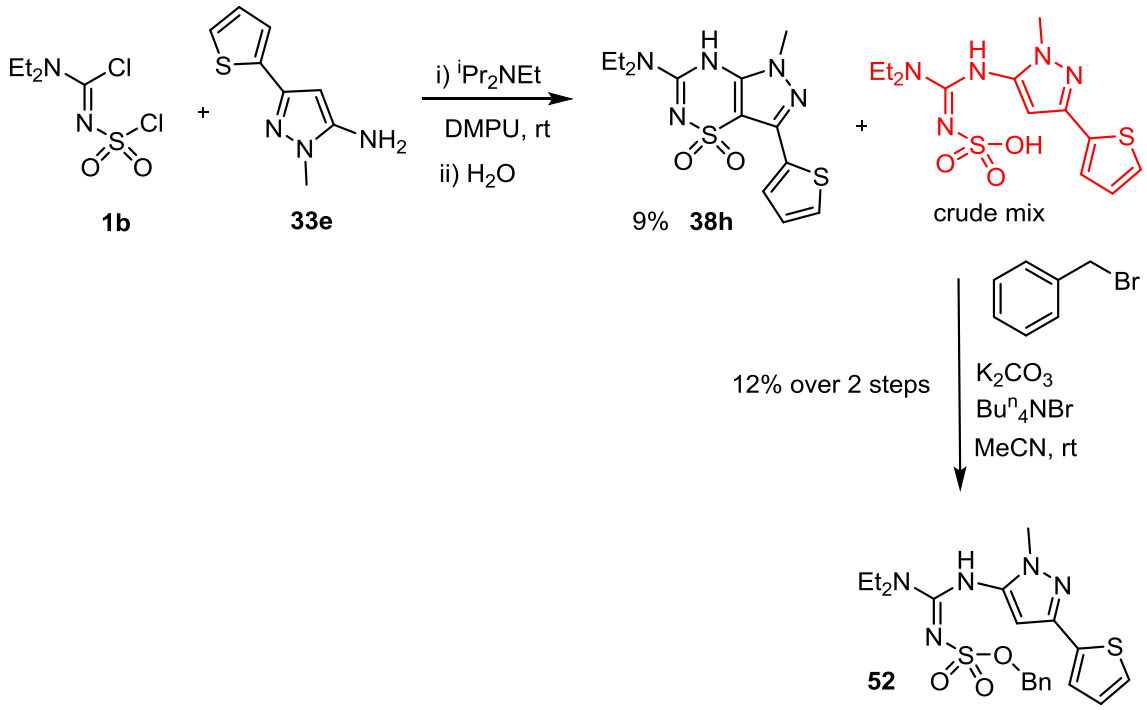
During the synthesis of t-butyl substituted compound **38j**, a side product which could conceivably be the sulfamic acid **51** (**Scheme 37**), from hydrolysis of the corresponding sulfamoyl chloride under the alkaline, aqueous workup conditions, was isolated. The structure **51** was hypothesised because of the presence of a singlet at δ 6.27ppm in the ^{1}H NMR spectrum, attributable to the hydrogen atom at C4 of the pyrazole ring. An uncyclised product **51** would be unsurprising due to the steric bulk of the *t*-butyl group at C3 of the pyrazole that would hinder ring closure at C4. Compound **51** could not be isolated and adequately characterised due to low stability. Nonetheless, poor yields of cyclised material can be rationalised by the formation of uncyclised chloride "intermediate" compounds which undergo hydrolysis or decomposition reactions.



Scheme 37: The formation of sulfamic acid side-product **51** from treatment of dichloride **1a** with pyrazole **33f**.

3.3.1 Trapping the sulfamic acid intermediate

To confirm the structure of these sulfonic acid side-products **51** or an analogue thereof, a stable derivative of one of these intermediates was sought, which could be adequately characterised. A set of substrates and conditions was selected which would give rise to a low yield of pyrazolo-thiadiazine **38**; hence method B was employed so the ring product could be collected conveniently by filtration. The organic phase of the filtrate (after collecting **38i**) was evaporated and the residue was treated with benzyl bromide and a base (**Scheme 38**) to afford a product tentatively assigned the benzyl sulfamate structure **52**. The molecular formula was established by high resolution mass spectrometry and the likely structure by NMR spectroscopy.

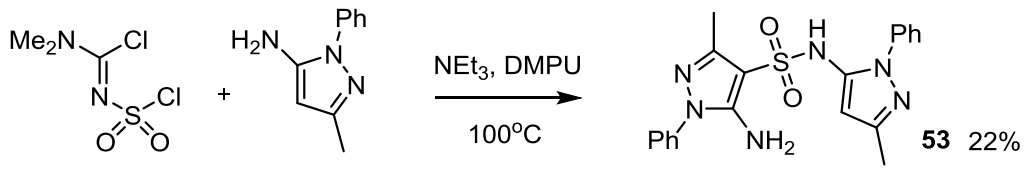


Scheme 38: The structural assignment of sulfamic acid side products by formation of a benzyl ester derivative **52.**

The benzyl sulfonate structure **52** was proposed over the alternative isomeric benzylamine structure due to the presence of the broad singlet at δ 5.34 ppm which was consistent with an N-H moiety rather than the alternative (and more acidic) sulfamic acid O-H, which would be expected at a much higher shift. The reason for isolation and characterisation difficulties with uncyclised intermediate compounds may be the low stability of a sulfamic acid or sulfamoyl chloride on silica gel or in solution.

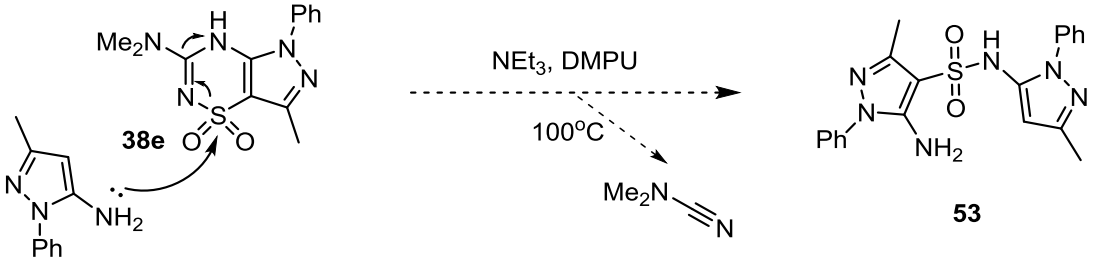
3.3.2 An unusual sulfur dioxide bridged dimer

A heated mixture of **1a** and **33c** in DMPU in the presence of triethylamine produced an unanticipated result. The product isolated was the sulfonamide **53** in place of an expected bicyclic ring structure **38e** (**Scheme 39**).



Scheme 39: The formation of sulfonamide bridged dimer **53** from heating in DMPU.

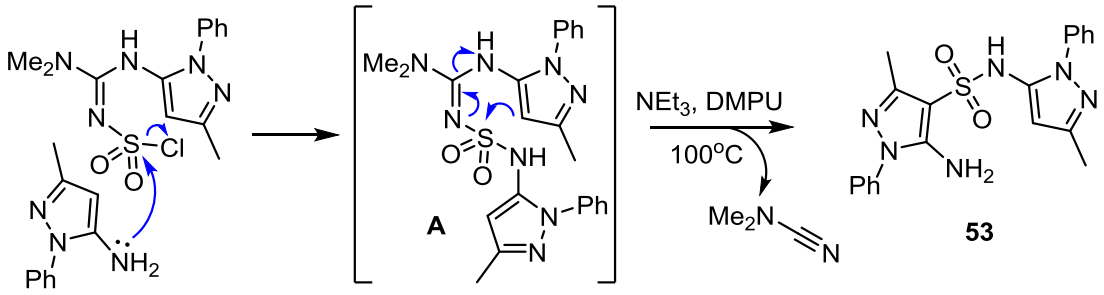
A plausible explanation for this result was that compound **53** could have arisen from the action of another pyrazole molecule on the fused thiatriazine compound **38e** (**Scheme 40**).



Scheme 40: Proposed mechanism for the formation of sulfonamide **53** from pyrazolo-thiadiazine **38e**.

This assumes that compounds **38** are susceptible to ring opening by moderate nucleophiles at higher temperatures, which has not been demonstrated. Attempts to replicate this result by treating **38e** with one equivalent of pyrazole under the same conditions were not successful; instead, the starting compound was returned along with small quantities of apparent decomposition products after 27 hours. No reaction between the pyrazole and the thiadiazine ring was indicated and no other compounds could be extracted from the mother liquor. Analogues of compound **53** were not observed throughout this body of work, which suggests that a mechanism by which an intermediate bis-adduct decomposes at higher temperatures is more likely. A second mechanism is hence suggested which involves the formation of a bis-adduct **A**, analogous to the bis-pyrazole materials **43** from Chapter 2, which subsequently rearranges under the high temperature conditions via an intramolecular cyclisation of

the pyrazole carbon C4 onto the sulfamide group with expulsion of dimethylcyanamide to give product **53** (**Scheme 41**).



Scheme 41: Proposed mechanism for the formation of sulfonamide **53** from a bis-adduct intermediate **A.**

Sulfamides differ from structurally similar ureas because the sulfur atom remains in the tetrahedral configuration such as in structure **a**.[45] The sulfamide moiety cannot adopt the resonance contributor **b** which ureas are capable of achieving (**Figure 36**).[45] This means that sulfamides can be susceptible towards nucleophilic attack although it is important to note that it would also be dependent on which other substituents are attached.

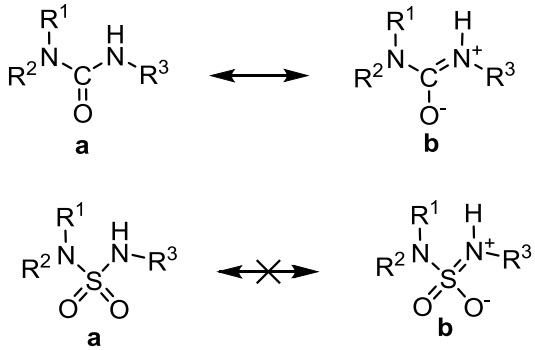


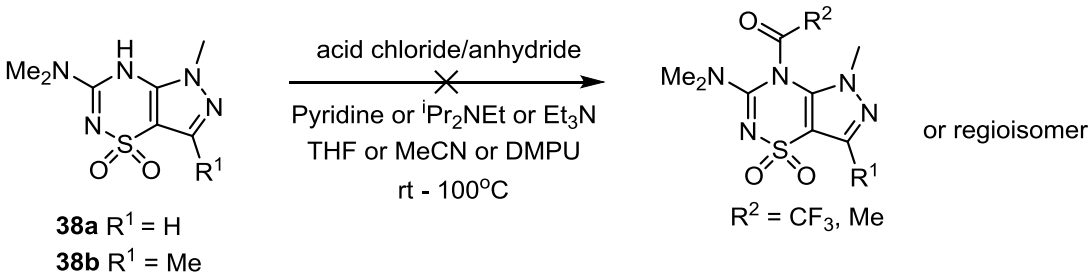
Fig 36. Canonical forms of a urea (top) and sulfamide (bottom).

Some examples of sulfamides which have been readily cleaved and displaced by nitrogen nucleophiles have been reviewed by Spillane and Malaubier.[47] A susceptibility of the sulfamide moiety in intermediate **A** toward nucleophilic attack by the nearby carbon nucleophile is thus plausible. Unfortunately, no bis-adduct material **A** was ever

isolated and so this mechanism remains unproven. No evidence of the formation of this type of product **53** was observed during experiments at room temperature or 50°C.

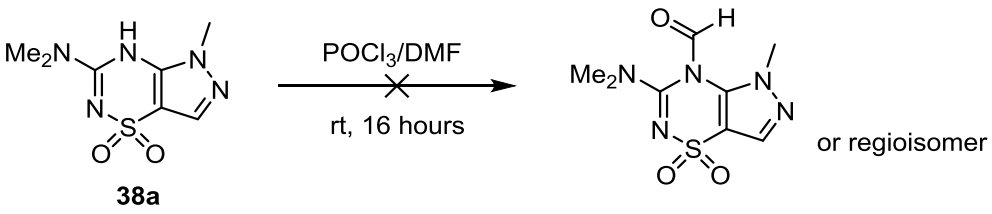
3.4 Reactivity of fused pyrazolo[3,4-*e*][1,2,4]thiadiazines with acylating agents

Trials were conducted to test the nucleophilicity of the NH moiety of the pyrazolo[3,4-*e*][1,2,4]thiadiazine heterocyclic system **38** towards acylation. Representative compound **38a** was treated with acetyl chloride in pyridine at room temperature and also acetic anhydride and Hünig's base in DMPU at 100°C. Substrate **38b** was stirred at room temperature with triethylamine and acetyl chloride in THF; room temperature with acetic anhydride in pyridine; and trifluoroacetic anhydride with triethylamine in acetonitrile at 50°C. No reactivity was observed under these conditions, and greater than 90% yields of the starting materials were recollected (**Scheme 42**).



Scheme 42: Attempted acylation of pyrazolothiatriazines **38a** or **38b** returned the starting materials.

A sample of **38a** was exposed to Vilsmeier conditions (phosphoryl chloride ($POCl_3$)/DMF) at room temperature for 16 hours (**Scheme 43**). No product could be isolated from the mother liquor, and evaporation to dryness revealed a mixture of likely decomposition products and starting material.



Scheme 43: Attempted acylation of pyrazolothiatriazine **38a** by Vilsmeier-Haack formylation returned the starting materials and caused unknown side-reactions.

In light of the failed acylation results, the research on derivatisation of this ring system was subsequently directed at exploration of reactions with some alkylating agents. The results from these studies form the basis of Chapter 5.

3.5 Conclusions

The versatile 1,3-dielectrophilic dichlorides **1** were condensed with readily available 1-substituted 5-aminopyrazoles **33** to generate the novel pyrazolo[3,4-*e*][1,2,4]thiadiazine dioxides **38**. No examples of the isomeric pyrazolo[3,4-*c*][1,2,6]thiadiazines **39** ring system were isolated, which implied selective nucleophilic action of the exocyclic amino group on the amidinyl carbon of dichlorides **1**, followed by cyclisation of the nucleophilic pyrazole C4 onto the sulfamoyl moiety. Steric factors were shown to have strong influence on the product yields which meant that substrates with small substituents needed to be selected in order to achieve adequate product yields. Complex mixtures of products were formed in some of these cases which may have included sulfamic acid 'intermediate' compounds such as **51**. One such example was characterised by means of benzyl ester **52**.

An unusual side product containing a sulfonamide bridge between two pyrazole units (**53**) was also isolated from a method which employed heating to 100°C. This indicated the potential electrophilic nature of the sulfamide group, although an exact mechanism of formation remains unclear.

CHAPTER 4: PYRAZOLO[1,5-*B*][1,2,4,6]THIATRIAZINE RING SYSTEM MODIFICATIONS ON A NUCLEOPHILIC CARBON

4.1 Introductory remarks

This research project was focused on the generation of new and uncommon heterocyclic ring systems from readily available precursor dinucleophiles and bis-electrophilic dichlorides **1**. As described in Chapters 2 and 3, this provided derivatives of one previously unreported and one very rare ring system (**34** and **38** respectively). Due to the novel nature of these unusual compounds, exploration into the reactivity of each ring system was thought to be of value to the synthetic chemistry community. Furthermore, some unique derivatives could be added to the CSIRO compound library.

Tautomerism of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** shows that the NH moiety can exist at N2, N4, or N7 and thus indicates the potential for the introduction of various substituents at these positions (**Figure 37**).

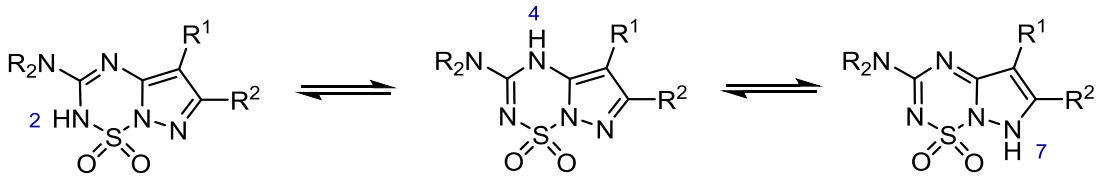
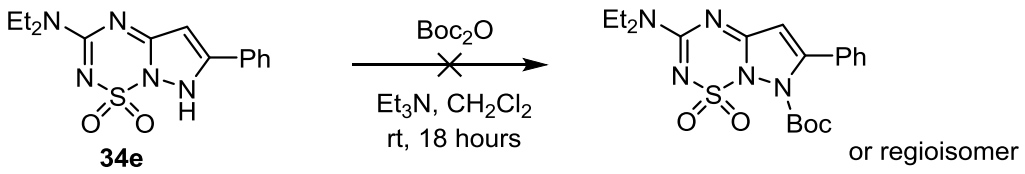


Fig 37. Tautomeric forms of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34**.

This chapter describes results from the exploration of the nucleophilicity of the fused pyrazoles **34** towards various carbonyl, sulfonyl, or halide based electrophiles. The results of alkylation experiments can be found in the subsequent chapter.

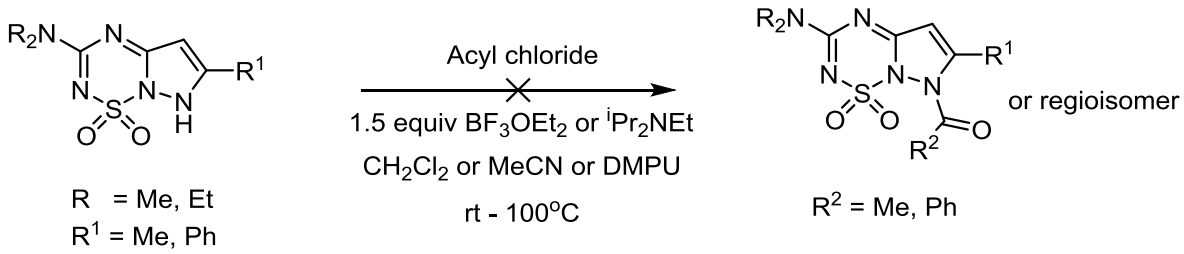
4.2 The susceptibility of pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides towards acylation

Experiments were carried out to test the nucleophilicity of the NH moiety of the novel heterocyclic system **34** towards acylation in order to form various amide and related derivatives. The substrate **34e** was stirred with di-tertiary-butyl dicarbonate (Boc_2O) and triethylamine in dichloromethane which resulted in the recovery of the starting material in 95% yield (**Scheme 44**).



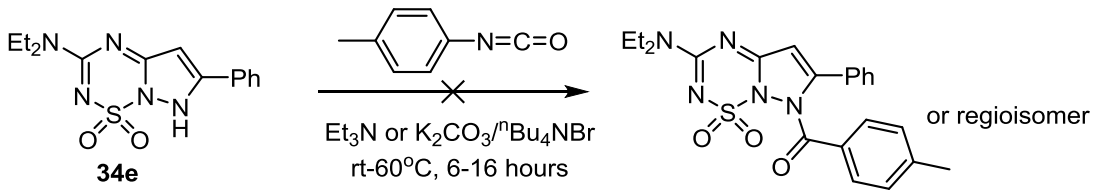
Scheme 44: The attempted synthesis of an *N*-Boc functionalised substrate.

The mild conditions were ineffective for this ring system which identified the fact that ring products **34** were not as nucleophilic as originally anticipated. A mixture of **34e**, acetyl chloride, and Hünig's base in acetonitrile was stirred at room temperature overnight after which no conversion was detected. An experiment in which a mixture of acetyl chloride and **34e** was heated to 55°C in acetonitrile for 16 hours returned the starting material in 94% yield. Treatment of **34e** with benzoyl chloride in DMPU and heating to 100^{o}C for 14 hours resulted in recovery of the starting material in 96% yield. Other attempts to afford an *N*-acyl derivative from either substrates **34j** or **34e** employed (1) the use of acetyl or benzoyl chloride with an amine base in dichloromethane, acetonitrile or DMPU at elevated temperatures (2) Friedel crafts conditions employing $BF_3.OEt_2$ as a Lewis acid catalyst in acetonitrile or dichloromethane with heating to reflux temperature, and (3) dicyclohexylcarbodiimide, simple carboxylic acids, *N*-hydroxy succinimide, and 20-30 mol% 4-dimethylaminopyridine in acetonitrile or DMF. No reactions were observed under a variety of conditions, with return of the starting material in >80% yield (**Scheme 45**).



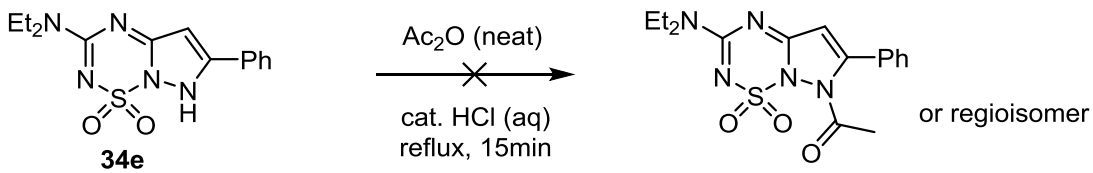
Scheme 45: Attempted acylation of pyrazolothiatriazines **34e** or **34j**.

Treatment of **34e** with 1.5 equivalents of *p*-tolyl isocyanate and a catalytic amount of triethylamine overnight resulted in recovery of only starting material. A similar experiment involving treatment of **34e** with *p*-tolyl isocyanate, an equivalent of potassium carbonate, and 0.1 equivalents of tetrabutylammonium bromide in acetonitrile at 60°C for 6 hours also returned starting material in near quantitative yield (**Scheme 46**).



Scheme 46: Attempted acylation of pyrazolothiatriazines **34e** with p-tolyl isocyanate.

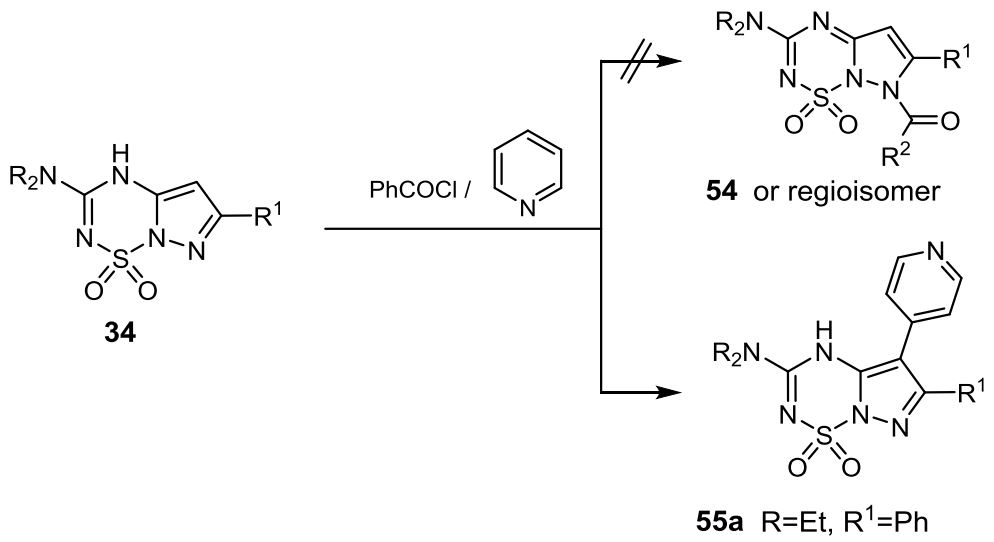
Heating a sample of **34e** in acetic anhydride to reflux temperature in the presence of catalytic aqueous hydrochloric acid gave rise to a complex mixture of products which could not be adequately purified. The harsh acidic conditions appeared to have acted to cause the decomposition of the parent ring structure and acylation of the breakdown products (**Scheme 47**).



Scheme 47: Attempted acylation of pyrazolothiatriazines **34e** with acetic anhydride.

4.3 Reactions of pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides with N–acylpyridinium species

Efforts towards acylation of fused pyrazolothiatriazines **34** were continued, and conditions which had been employed successfully with related ring systems were thus explored.[27] A representative compound **34e** was treated with benzoyl chloride in pyridine with the aim of introducing a benzoyl moiety at a nucleophilic nitrogen atom. Neither the expected N-benzoyl derivative **54** (**Scheme 48**), nor either of the N2- or N4-benzoylated regioisomers were observed. In addition to recovered starting material (17%), the only isolated product, in 51% yield, was a bright yellow crystalline solid; the ^{1}H NMR spectral data for which were not consistent with structure **54**. The only new aromatic signals were two broad doublets at δ 8.40 ppm and δ 7.87 ppm, each integrating for 2H, and there was no signal attributable to H5 on the pyrazole ring. Mass spectrometric data and X-ray crystallography confirmed the structure of the product to be the 5-(pyridin-4-yl) derivative **55a** (**Figure 38**).



Scheme 48: The formation of 5-(pyridine-4-yl) derivative **55a**.

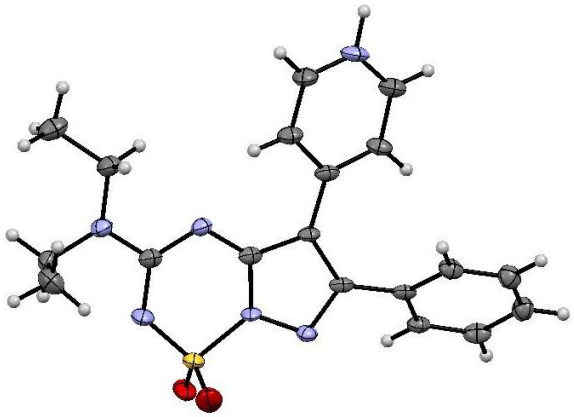
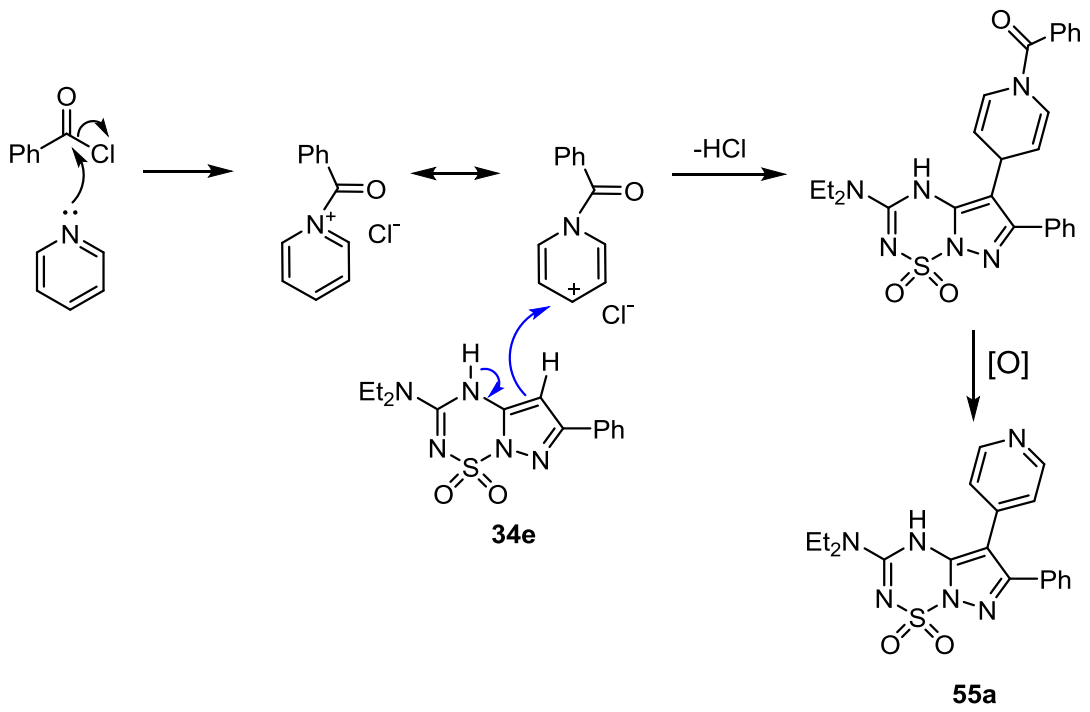


Fig 38. X-ray crystal structure of 5-(pyridin-4-yl)-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **55a** in a zwitterionic salt form.

4.3.1 Proposed Mechanism of the Reaction with Pyridine

A plausible mechanism for the formation of the pyridine adduct **55a** is shown in **Scheme 49**. Interestingly, C5 of the pyrazolothiatriazine ring system **34** appears to be more nucleophilic towards the *N*-acylpyridinium species than any of the ring nitrogen atoms. The nucleophilic aromatic substitution described in **Scheme 49** indicates a mechanism where the *N*-acylpyridinium species acts as an electrophile activated towards nucleophilic attack at C4 of the pyridine ring. Oxidative removal of the benzoyl or acetyl substituents generated the re-aromatised product observed.

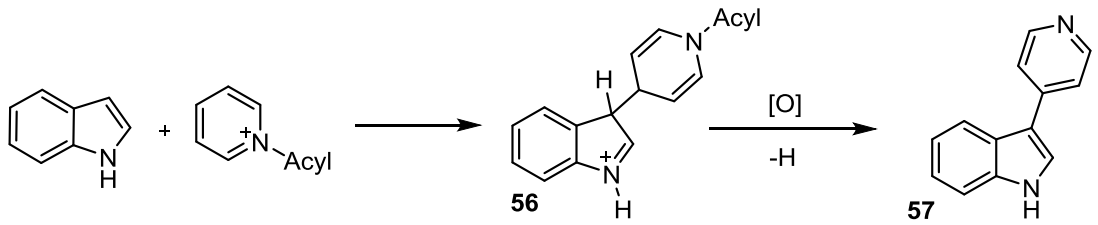


Scheme 49: The proposed mechanism of formation of pyridine adduct **55a.**

4.3.2 Literature precedent for nucleophilic aromatic substitution of pyridine

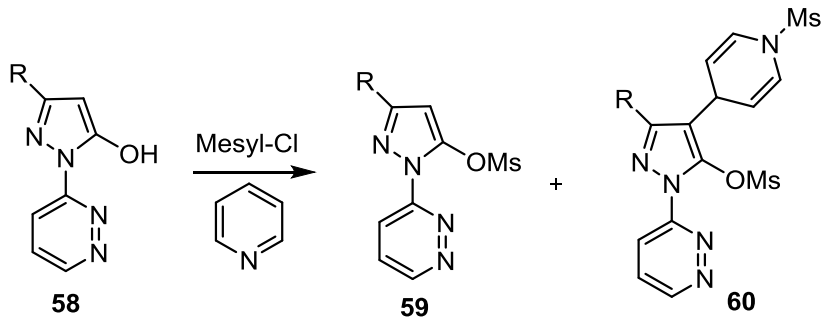
Reports of similar transformations have been documented in the literature. The reactivity of *N*-acylpyridinium and *N*-acylquinoline salts towards indoles and other nucleophiles was observed by Von Dobeneck *et al* in 1962.[117] Lyle and coworkers[118] were able to react Grignard reagents with the alkylformate salts of 3,4-lutidine, followed by oxidation with elemental sulfur to re-generate the aromatic pyridine ring. Akiba et al[119] reacted 1-ethoxycarbonylpyridinium chloride with trimethylsilyl ethers to provide 1,4-dihydropyridines with high regioselectivity, although 1,2 dihydropyridines were isolated in minor quantities. Intermediate dihydropyridines were oxidised with oxygen gas or silver(I)nitrate to furnish 4-(2-oxoalkyl)pyridines. The reactivity with Grignard reagents was expanded to study 1,4- vs 1,2- pyridine addition.[120,121] Bergman and collegues[122] demonstrated that, depending on solvent and the ratio of reactants, N-acylpyridinium salts condense with indole to give 3-(N-acyl-1,4-dihydro-4-

pyridyl)indole **56** or 4-(N-acyl-3-indolyl)pyridinium chloride. Hydrolysis of **56** under alkaline conditions gave the 3-(4-pyridyl)indoles **57** (**Scheme 50**).



Scheme 50: The selective reaction of indole at the 4-position of pyridine.

Matyus et al[123] observed the reactivity of the 1-acylpyridinium salt when acylating 5-hydroxy-1-(pyridazin-3-yl)pyrazoles **58** with mesyl chloride in pyridine. These reactions afforded the expected *O*-mesyl derivatives **59**, along with the 4-(*N*-mesyl-dihydropyridine) derivative **60** as a side product (**Scheme 51**).

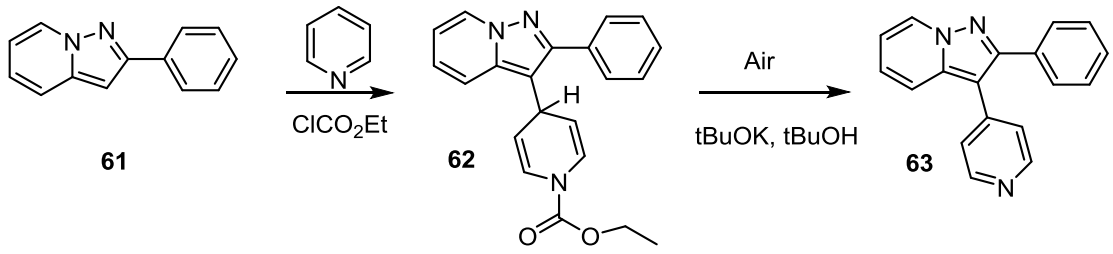


Scheme 51: The side product **60** in the mesylation reactions of 1-(pyridazin-3-yl)-pyrazoles **58**.

It has also been reported that the addition of carbon nucleophiles to *N*-alkylpyridinium salts can result in nucleophilic substitution at both the C4 and C2 atoms of the pyridine ring. The instability of dihydropyridinium intermediates from these reactions was also established. For this reason, most of the previous works were accomplished using *N*-acylpyridinium species, although Volochnyuk and collegues[124] reported the reactions of *N*-benzyl 3-cyanopyridinium chloride with various carbon nucleophiles including 5-aminopyrazoles, 5-aminoisoxazoles, 2-aminothiazoles and 6-aminouracils. The

reactions took place solely at the 4-position of pyridine and the dihydropyridines were not oxidised.

A small group of 3-heteroaryl-2-phenylpyrazolo[1,5-*a*]pyridines **63** were prepared as adenosine A1 receptor antagonists.[125] The 2-phenyl pyrazolopyridine **61** was treated with ethyl chloroformate and pyridine to afford dihydropyridine **62**, which was then aromatised to the pyridinyl-substituted compound **63** using potassium *tert*butoxide in *tert*-butyl alcohol (**Scheme 52**). Analogously, 3-(pyridazin-4-yl)pyrazolo[1,5-*a*]pyridine was obtained from a similar reaction with ethyl chloroformate and pyridazine.



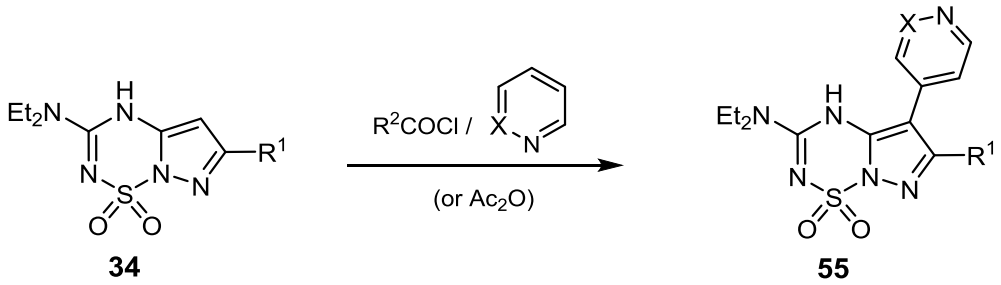
Scheme 52: The selective formation of pyrazolopyridine **63** utilising pyridine ethylchloroformate salts.

Follot and coworkers[126] synthesised three 3-(pyridin-4-yl)imidazo[1,2-*a*]pyridine derivatives to evaluate as anti-apoptosis agents. 3-(Pyridin-4-yl)-2-(4-fluorophenyl)-6-iodoimidazo[1,2-*a*]pyridine was synthesised from the imidazo-pyridine precursor using ethyl chloroformate in pyridine followed by oxidation of the diydropyridinyl intermediate with *O*-chloranil.

Significantly, the reaction between compounds **34** and pyridine occurs selectively in good yield without the need for heat or catalysis or a subsequent oxidation step, whereas in most of the previously reported reactions, a subsequent oxidation step had to be introduced to re-aromatize the dihydropyridine substituent. The frequency with which the pyridine ring is found in biologically active molecules of significance suggests potential significance of this type of chemistry for drug discovery. The materials required for this reaction to take place are relatively inexpensive and the preparation is simple in terms of both the reaction and isolation processes.

4.3.3 Summary of results

This unusual synthesis was explored in more detail to determine preferred conditions for pyridine and pyridazine as two representative azaheterocycles (**Scheme 53**, **Table 6**).



Scheme 53: Pyrid(az)ine derivatives were generated from electrophilic attack of the pyrazole.

Table 6: The synthesis of pyrid(az)ine derivatives from pyrazolothiatriazines **34**.

R^1	X	Conditions	Product	Yield [%]
Ph	C	PhCOCl, pyridine, rt	**55a**	51*
Ph	C	Ac_2O, pyridine, rt	**55a**	35*
Thien-2-yl	C	PhCOCl, pyridine, rt	**55b**	70
Thien-2-yl	C	MeOCOCl, pyridine, MeCN, rt	**55b**	68
Thien-2-yl	N	MeOCOCl, pyridazine, THF, rt	**55c**	55
Thien-2-yl	N	MeCOCl, pyridazine, THF, rt	**55c**	46
Thien-2-yl	N	Ac_2O, pyridazine, THF, rt	**55c**	8*

* Recovery of the starting material

Attempted acetylation of compound **34e** with acetic anhydride/pyridine[27] resulted in isolation of **55a** in 35% yield along with a 41% yield of recovered starting material. The reaction of 6-(thien-2-yl) substrate **34h** with benzoyl chloride in pyridine afforded the 5-

(pyridin-4-yl) derivative **55b** in 70% yield. Compound **55b** was also prepared from **34h** in 68% yield by treatment with two equivalents of both methyl chloroformate and pyridine in acetonitrile. A similar reaction of **34h** with methyl chloroformate and pyridazine in tetrahydrofuran afforded the 5-(pyridazin-4-yl) derivative **55c** in 55% yield. Substituting acetyl chloride for methyl chloroformate in the above reaction also provided **55c** in 46% yield. The analogous reaction with acetic anhydride was much less efficient, resulting in only an 8% yield of **55c**, and recovery of most of the starting material. From these experiments, it appeared that use of methyl chloroformate offered no obvious advantage over acetyl or benzoyl chloride. Further optimization of these reactions would require more systematic and detailed work (such as process chemistry) which was beyond the capacity of this PhD research.

4.3.4 Extension to other pyridine derivatives

The selective reactions of the pyrazolothiatriazines **34** with pyridine and pyridazine, indicating the significant nucleophilicity of C5 in this ring system, encouraged the consideration of similar reactions with analogues of these two simple heterocycles. The scope of this uncommon reaction was explored in terms of other pyridine analogues, which conceivably could give rise to similar derivatives to **55**. Shilcrat *et al*[127] reported that the ethyl chloroformate salts of a variety of benzo-fused six-membered π-deficient aza heteroaromatics reacted with 6-(4-fluorophenyl)-2,3-dihydroimidazo[2,1-*b*]thiazole at the 5-position. These results provided us with confidence that other aza heteroaromatics could be applied to the pyrazolothiatriazine system **34**. It is also significant to mention that, aside from the work by Volochnyuk and co-workers,[124] all of the literature reports of analogous reactions have involved unsubstituted pyridine. This meant that the area of nucleophilic addition to *N*-acyl derivatives of pyridine or similar azaheterocycles remains undeveloped.

Treatment of pyrazolothiatriazine **34h** with 4-methylpyridine and either of acetyl chloride, acetic anhydride, or benzoyl chloride in acetonitrile or tetrahydrofuran at either ambient temperature (20°C) or 60°C resulted in negligible reaction and >90% recovery of starting material. Similar experiments with 2,6-dimethylpyridine and

quinoline also resulted in >90% recovery of starting material. A summary of the attempted substitutions on pyridine analogues is outlined in **Table 7**.

Table 7: Attempted synthesis of substituted pyridine adducts of pyrazolo[1,5-b][1,2,4,6]thiatriazine dioxides **34**.

R^1	Conditions*
Ph	4-methylpyridine, Ac_2O, MeCN, rt
Ph	4-methylpyridine, MeCOCl, THF, rt
Thien-2-yl	4-methylpyridine, PhCOCl, MeCN, rt
Ph	4-methylpyridine, PhCOCl, rt
Ph	4-methylpyridine, MeCOCl, THF, Et_3N (5 drops), rt
Ph	2,6-lutidine, Ac_2O, MeCN, 60°C
Ph	2,6-lutidine, PhCOCl, MeCN, Et_3N (3 drops), rt
Ph	Quinoline, MeCOCl, THF, rt

* Recovery of the starting material >90% after 1-2 days.

Volochnyuk *et al*[124] found that 5-aminopyrazoles reacted with *N*-benzyl-3-cyanopyridinium chloride to afford 1-benzyl-3-cyano-4-(5-aminopyrazol-4-yl)-1,4-dihydropyridines with high regioselectivity. 5-Aminoisoxazole, 2-aminothiazole, and 6-aminouracil reacted analogously. However, treatment of pyrazolothiatriazine **34h** under a series of conditions: either *N*-benzyl-3-cyanopyridinium chloride or *N*-benzyl-3-cyanopyridinium bromide in either methanol, aqueous methanol, or DMSO, at temperatures from 20°C to 80°C, resulted in greater than 80% recovery of starting material and minor amounts of decomposition products. The addition of triethylamine to the reaction mixture also did not result in product formation.

Unfortunately, throughout the course of this study, no analogues other than pyridine or pyridazine could be substituted onto the pyridine ring, and so research efforts were directed elsewhere.

4.3.5 NMR analysis of pyrid(az)ine adducts 55

Compound **55c** displayed interesting behaviour during NMR spectroscopy experiments. In the ^{1}H NMR spectrum, initially recorded in DMSO-d_6 solution at 26°C, all of the resonances were broader than expected. Broad resonances from the hydrogens on the 3-dialkylamino substituent have often been observed in ring systems produced from dichlorides **1**, and this is due to restricted rotation of the dialkylamino group.[22] However, the resonances due to the pyridazine and thiophene substituents were also broad, suggesting restricted rotation of these heteroaryl substituents. A similar effect was observed in the ^{13}C NMR spectrum (DMSO-d_6 solution at 26°C). Long and short range HETCOR (HMBC, HMQC) data confirmed the small, broad carbon peaks to be those from the pyridazine substituent (see Appendix A). Initially this was attributed to the viscosity of the solvent which gave a poor signal to noise ratio, and therefore; NMR experiments were conducted in methanol-d_4. ^{1}H NMR and ^{13}C NMR signal strength was inconveniently limited by the poor solubility of **55c** in methanol. A variable temperature ^{1}H NMR spectroscopic study on this compound (30 to 70°C in DMSO-d_6) showed a gradual sharpening of the diethylamino group resonances into the expected quartet and triplet, but the pyridazine substituent resonances were not sharpened to a similar extent (**Figure 39**).

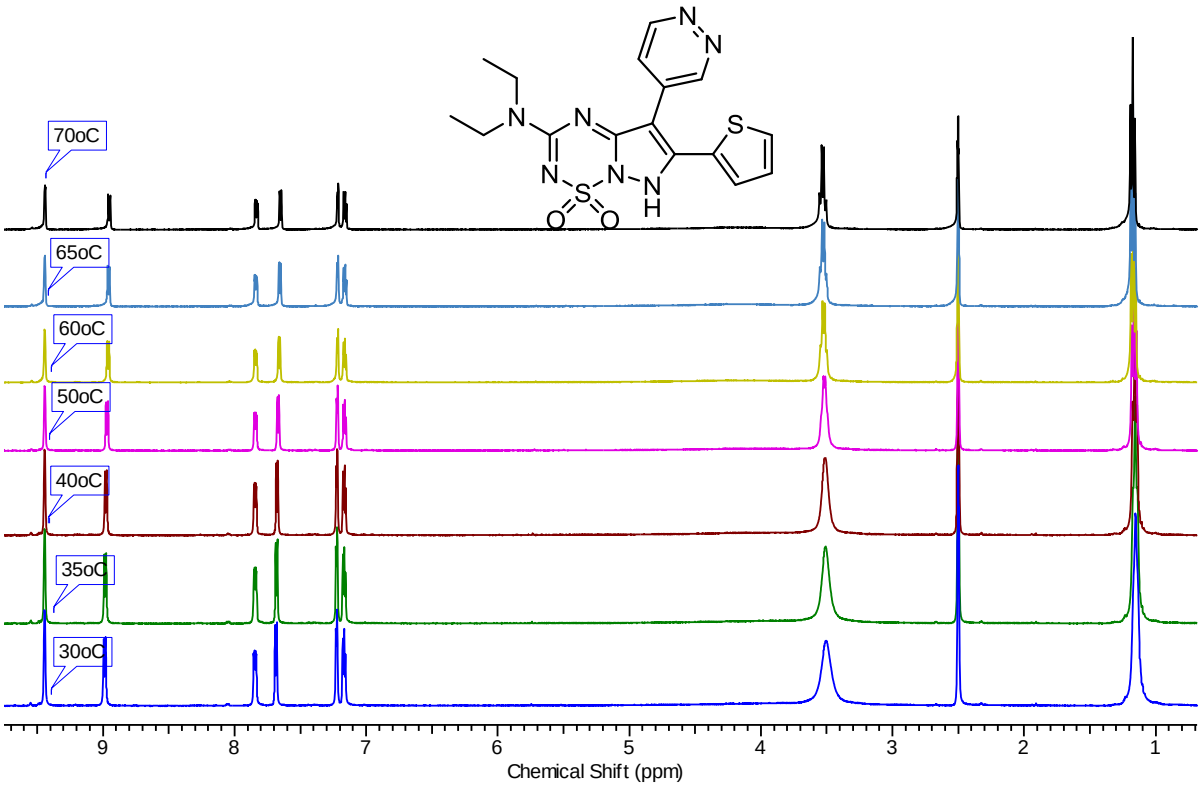


Fig 39. Variable temperature ^{1}H NMR spectra of pyridazine adduct **55c** (DMSO-d_6)

Plausibly, in the original ^{13}C NMR spectrum, the pulse-repetition time was insufficient to allow adequate relaxation between scans. The relevant spectrometer parameter (D1) was adjusted from the usual setting of 2 seconds to 5 seconds to allow for additional relaxation by these carbons. At 26°C with D1=5 seconds, the signals attributed to the diethylamino protons were broad and the signals attributable to the pyridazine substituent were poorly resolved. A ^{13}C NMR spectrum (DMSO-d_6) recorded at 65°C overnight with D1=5 seconds clearly displayed all carbon resonances as sharp signals compared to the experiment at 26^{o}C with D1=2 seconds (**Figure 40**).

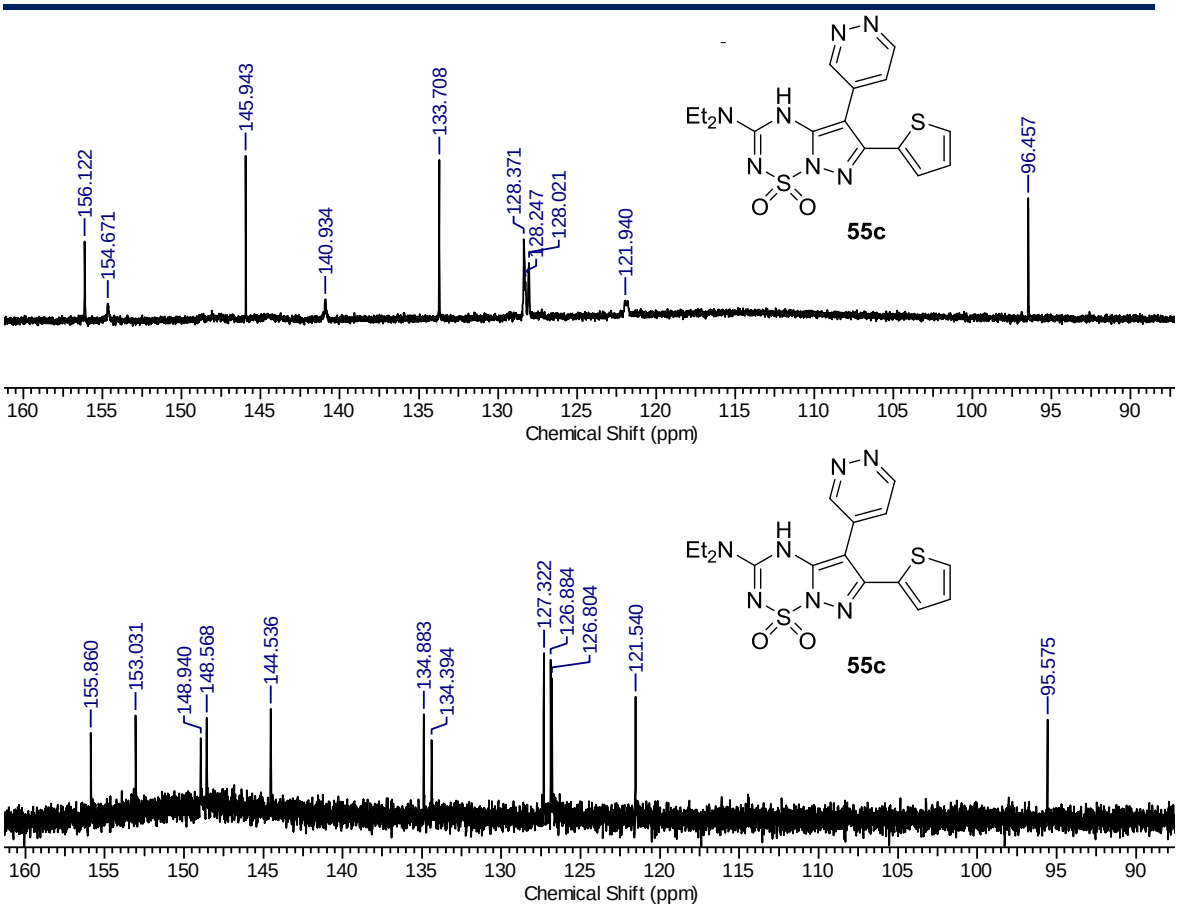
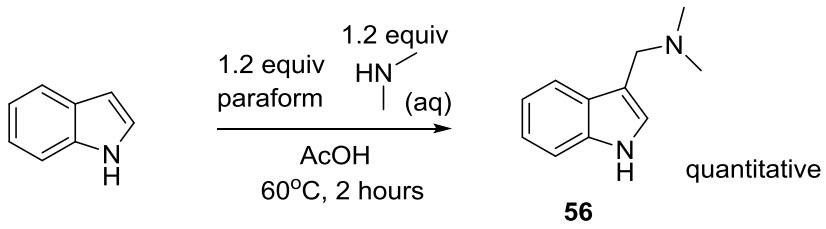


Fig 40. Region of the ^{13}C NMR spectra of compound **55c** in DMSO-d_6 showing signals from the parent ring system and the thienyl and pyridazine substituents. The spectrum recorded at 26^{o}C with D1=2 (above) gave broad resonances which were difficult to distinguish. Comparatively, the spectrum taken at 65^{o}C with D1=5 (below) gave sharper signals.

The improvement in signal strength can also be accredited to the greater solubility of the analyte **55c** in the higher temperature solution; whereas at 26°C only a very minor quantity (~5mg) could be dissolved in the NMR solvent. The products **55** were highly planar, rigid compounds as illustrated by the X-ray crystal structure of **55a** (**Figure 38**) which seemingly did not rotate freely on the NMR timescale in DMSO-d[6]. The effect of altering the time delay parameter before each scan (D1) demonstrated that the relaxation times were indeed longer than those for carbons within the precursor materials **34**.

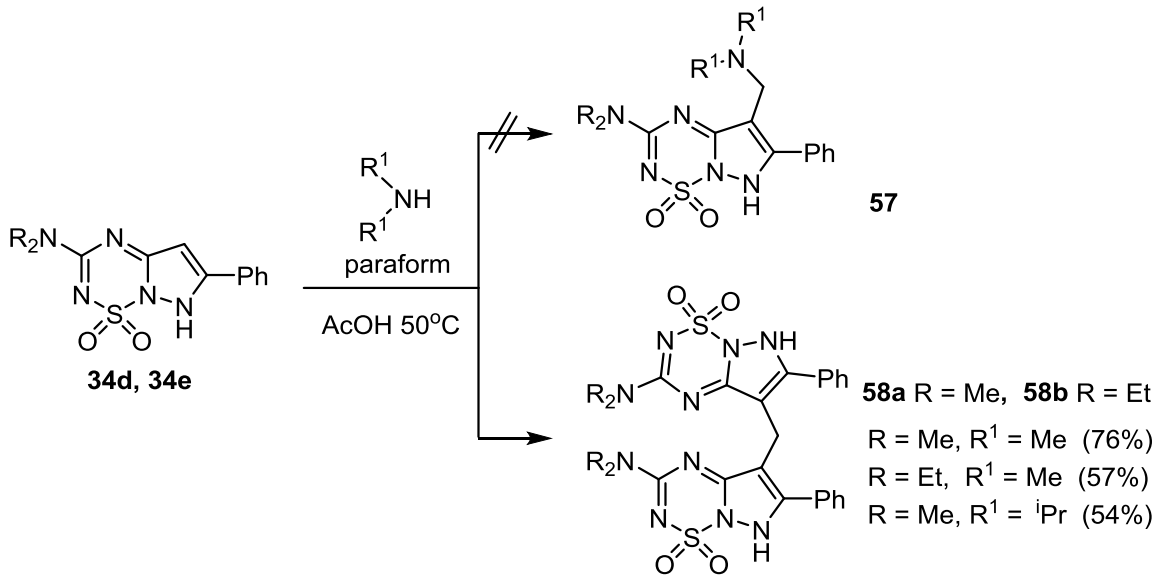
4.4 Mannich Reaction at C5

Given that the ring system **34** demonstrated reactivity at the nucleophilic pyrazole carbon C5, some literature searches of similar structures (such as indoles, imidazoles and other fused pyrazoles) were conducted. A Mannich-type reaction was envisaged to introduce a variety of dialkylaminomethyl substituents from simple secondary amines and formaldehyde utilising methodology successfully applied to imidazo-pyridines.[128,129] Indole was employed as a model substrate, and under literature conditions [128] this reaction furnished the alkylamine **56** in quantitative yield without the requirement for further purification (**Scheme 54**).



Scheme 54: The synthesis of 3-(N,N-dimethylamino methylene)indole from a Mannich base.

Treatment of substrate **34d** with aqueous formaldehyde and aqueous dimethylamine[128] rapidly produced a complex mixture of unknown materials. Treatment of pyrazolothiatriazines **34d** and **34e** with secondary amines and paraformaldehyde in acetic acid[129] did not provide the expected products **57**; instead, the methylene 5,5'-bis-adducts **58** were obtained (**Scheme 55**). This reaction occurred despite changes to the secondary amine used. The conditions were modified to form the imine species first, followed by the addition of the pyrazolothiatriazine but the result was a complex mixture.



Scheme 55: Methylene dimers **58** were formed from the treatment of substrates **34d** and **34e** with secondary amines and paraformaldehyde in acetic acid.

The bis-methylene compounds **58** were stable white solids which were insoluble in most organic solvents and only moderately soluble in DMF and DMSO, making characterisation by NMR spectroscopy problematic. Mass spectrometry and single crystal X-ray diffraction of **58a** allowed structural confirmation (**Figure 41**).

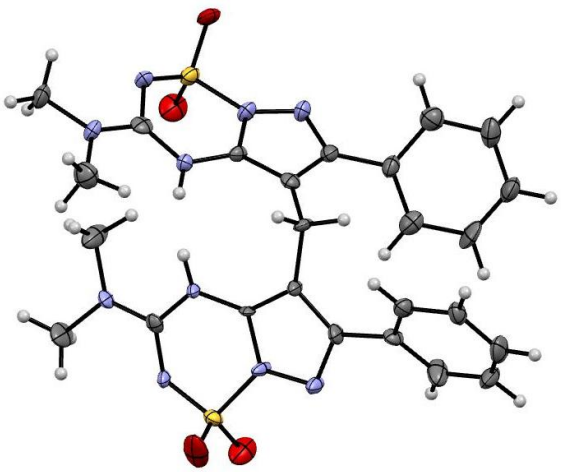
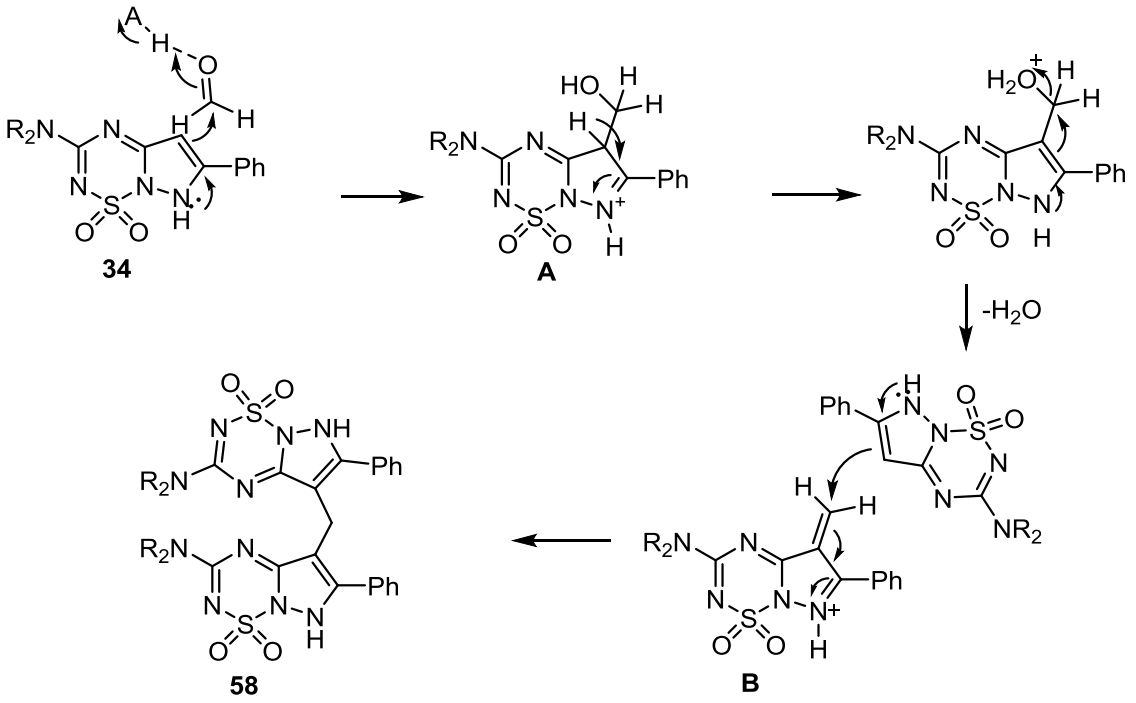


Fig 41. ORTEP diagram of methylene-bridged dimer **58a**. A foreign entity (potentially a HCl salt) was omitted from the crystal structure for clarity.

4.4.1 Mechanism of Formation of Methylene-bridged Dimers

Analogous "dimerisations" of indole rings have been documented in the past as side products of the Mannich condensation conditions.[130] Indoles possess an electron rich carbon nucleophile at the C3-position which reacts with electrophilic formaldehyde to form a 3-carbinol indole or 3-hydroxymethyl indole tautomer and, under acidic conditions, these have been shown to condense with another indole moiety.[131,132] The intrinsic instability of indole-3-carbinol in acidic media arises from the vinyl hemiaminal moiety of the indole ring.[133] This unique structural feature underlies the high susceptibility of indole-3-carbinol to acid-mediated dimerization and oligomerisation.

Methylene-bridged dimers **58** were thus assumed to arise from the reaction between formaldehyde and the pyrazole carbon nucleophile on the substrate **34** to create an alcohol intermediate **A** (**Scheme 56**). The alcohol dehydrated under acidic conditions to generate the iminium species **B** which subsequently reacted with another molecule of **34** to furnish the dimer **58**.

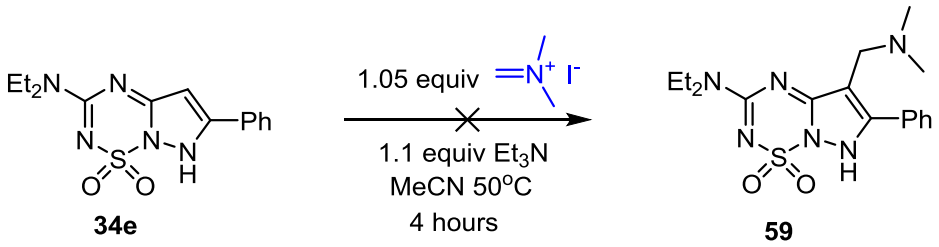


Scheme 56: Proposed mechanism of formation of methylene dimers **58**.

No 5-dialkylaminomethyl derivatives were isolated from these experiments, which indicates that under the conditions employed, either compounds **34** react instead with formaldehyde, or there is a possibility that reaction of the iminium (Mannich) intermediate could produce the dimer **58**. Unfortutaley, the remaining materials constituting the product mixture could not be separated and identified individually to confirm which of these two reagents react with the pyrazole carbon. Possible side products might be those resulting from the reaction of formaldehyde on an NH group or some other decomposition from the acidic conditions; however this was not verified. A different method was required in order to selectively form dialkylaminomethyl derivatives without the competing reaction with formaldehyde.

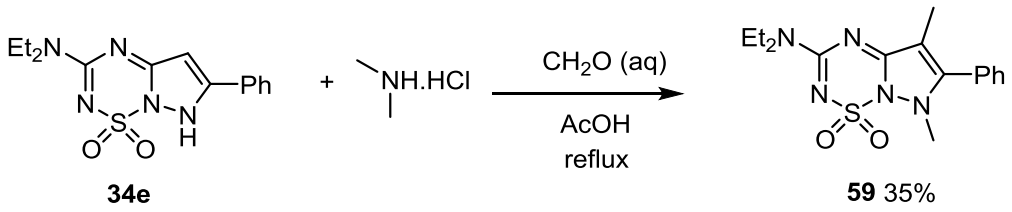
4.4.2 Alternative Approaches to Synthesis of Dialkylaminomethyl Derivatives

Brown and McGeary documented the formation of Mannich bases from pyrazolo-pyridine with imine salts in high yields.[134] Treatment of compound **34e** with Eschenmoser's salt (*N*,*N*-dimethylmethyleneammonium iodide) and triethylamine in acetonitrile resulted in only degradation products forming over a 14 hour period (**Scheme 57**).



Scheme 57: The attempted reaction of substrate **34e** with Eschenmoser's salt.

Treatment of substrate **34e** with dimethylamine hydrochloride and aqueous formaldehyde in acetic acid at reflux temperature resulted in the formation of a dimethylated derivative **59** (**Scheme 58**).



Scheme 58: A dimethylated derivative resulting from the treatment of substrate **34e** with aqueous formaldehyde.

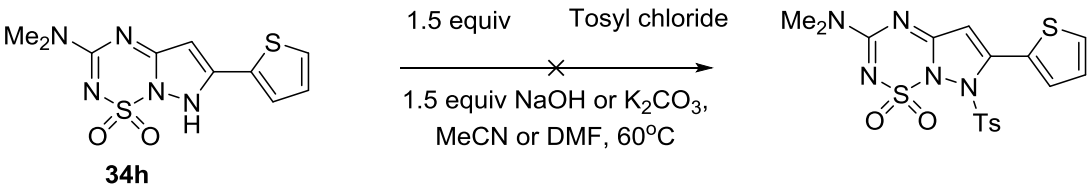
Formaldehyde solutions are known to decompose to formic acid as well as polymerisation to the paraform trimer.[135] The potential presence of formic acid in the old bottle of formalin used (and emptied) in this experiment suggested an Eschweiler-Clark methylation could be the cause of this result. Subsequent efforts to produce larger quantities of this material for further characterization included: (1) formaldehyde in water at reflux; (2) formaldehyde in neat formic acid at room temperature or at reflux; (3) Paraform in neat formic acid at reflux; (4) and one equivalent of Paraform and formic acid in acetic acid at 60^{o}C. In all cases, complex mixtures were generated. The formic acid present in the old bottle of formalin could account for the methylation result; however the exact concentration of this reagent in the solution was not known and any attempts to duplicate the methylation without the original bottle were not likely to succeed. None of the efforts to obtain a dialkylaminomethyl derivative from this chemistry were successful and it was decided that it would be more productive to direct our efforts elsewhere.

4.5 Sulfonylation of pyrazolothiatriazines

The susceptibility of pyrazolothiatriazines **34** towards sulfonylation was explored through attempts to introduce a 4-toluenesulfonyl (tosyl) moiety at either of N2, N4, N7, or C5. A number of tosylation reactions have been reported including Ag_2O-KI-TsCl,[136] TsCl-Et_3N-Bu_2SnO[137], commonly in the presence of base such as potassium hydroxide or potassium carbonate. Most of these papers report tosylation of primary alcohols and require catalysts in order to achieve adequate yields. Conditions for tosyl

protection of indoles were considered as a guide for the tosylation of the N or C nucleophilic sites on the pyrazolothiatriazine ring system.

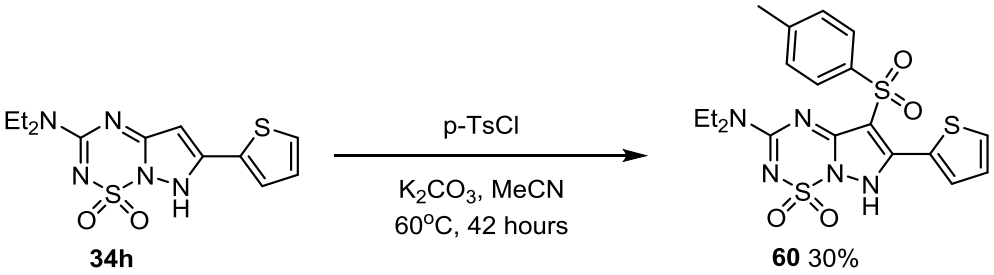
Initial attempts to synthesise a sulfonylated derivative of **34g** were unsuccessful and this may have been due to the low solubility of the starting material (**Scheme 59**).



Scheme 59: Attempted tosyl protections of substrate **34g** under standard sulfonylation conditions.

Further experiments were aimed at forming an N-tosyl derivative from compound **34h** and tosyl chloride involving (1) potassium iodide (KI) and silver(I)oxide (Ag_2O) in catalytic or stoichiometric quantities,[136] (2) 20mol% indium(III)chloride ($InCl_3$),[138] (3) Friedel crafts conditions using boron trifluoride (BF_3OEt_2) or (4) the addition of pyridine as a sulfonyl transfer agent. These additives did not cause a reaction between tosyl chloride and substrate **34h** under a range of base/solvent combinations or with heating. The starting materials were returned in 50-90% yields.

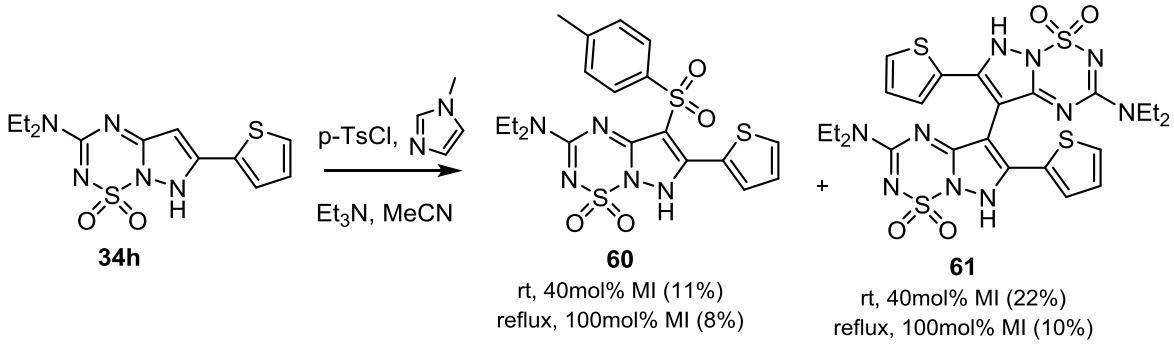
Treatment of 6-(thien-2-yl)pyrazolothiatriazine **34h** with tosyl chloride and potassium carbonate in acetonitrile at 60°C resulted in a 30% yield of 5-tosyl derivative **60** along with 38% of the recovered starting material (**Scheme 60**) after 42 hours.



Scheme 60: C5-tosyl derivative **60** from the reaction of **34e** with 4-toluene sulfonyl chloride and potassium carbonate.

Remarkably, the reaction was selective towards the C5 position of the pyrazolothiatriazine fused ring, rather than N2, N4, or N7. This mirrors the regioselectivity of the acylation experiments and provides further insight into the reactivity of this new ring system. We also noted that the sulfonyl-substituted compound **60** fragmented to return the starting material under the acidic conditions of silica gel chromatography, and gradually decomposed upon storage and handling.

The use of 1-methyl imidazole (MI) for catalysis of tosylation of sterically hindered alcohols has been reported.[139] Treatment of **34d** with tosyl chloride and both catalytic and stoichiometric amounts of MI provided low yields of compound **60** along with the 5,5'-dimer **61** which was identified by HRMS, with NMR data supportive of this structure (**Scheme 61**). Prolonged reaction times, heating or employing dichloromethane as a solvent did not improve the reaction and after >20 hours, mostly starting material was recovered.



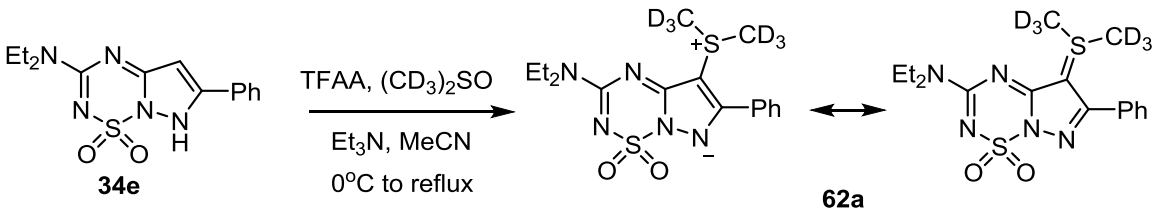
Scheme 61: N-Methylimidazole catalyzed sulfonylation of compound **34d**.

The formation of dimer **61** likely resulted from nucleophilic attack of C5 of **34h** at the 5-tosyl group of **60**. The displacement of sulfonyl groups by carbon-centred nucleophiles is known in the cases of imidazole,[140] pyrazole,[141,142] and pyrazole-fused[143] ring systems, usually in compounds where the sulfonyl-containing ring also bears a strongly electron-withdrawing substituent. In the case of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxide derivative **60**, presumably the electron-withdrawing nature of the sulfamide moiety facilitates such a reaction.

This result suggests potential for tosyl derivative **60** to be exploited as a synthetic intermediate, although the degradation under acidic conditions and the propensity towards dimer formation remains a concern. Displacement of the tosyl group of **60** by other nucleophiles, for example amines, thiol(ate)s, or alkoxides, might be possible.

4.6 Dimethylthiolation of C5 by dimethylsulfoxide and trifluoroacetic anhydride

Through the course of investigating the action of acylating agents on the fused pyrazolothiatriazines **34**, a sample of **34d** was treated with an excess of both trifluoroacetic anhydride (TFAA) and triethylamine. A dimethylsulfyl derivative **62a** was generated in one instance (**Scheme 62**), the structure of which was revealed by X-ray crystallography (**Figure 42**). Attempts to replicate this result (with TFAA/Et_3N) were unsuccessful and only trace amounts of a 5-$COCF_3$ compound were collected after 46 hours, along with the starting material (77%). This led to the conclusion that the unexpected product from the initial experiment was due to a contaminant in the reaction mixture, and it was soon deduced that residual DMSO-d_6 from recovered NMR samples.



Scheme 62: Thiolation of substrate **34e** with DMSO-d^6 under Swern-type conditions.

The activation of DMSO-d_6 by the TFAA would generate an electrophilic salt (F_3CCO_2-S(+)Me_2) which had apparently reacted with the nucleophilic pyrazole carbon. The product structure was not evident from the ^{1}H NMR spectrum because the deuterated methyl groups did not provide signals. The structure was confirmed by X-ray crystallography, which does not differentiate between deuterium and hydrogen (**Figure 42**), and also by accurate mass spectrometry. **Scheme 62** outlines the contributing ylide and resonance forms which are discussed in more detail in Chapter 8.

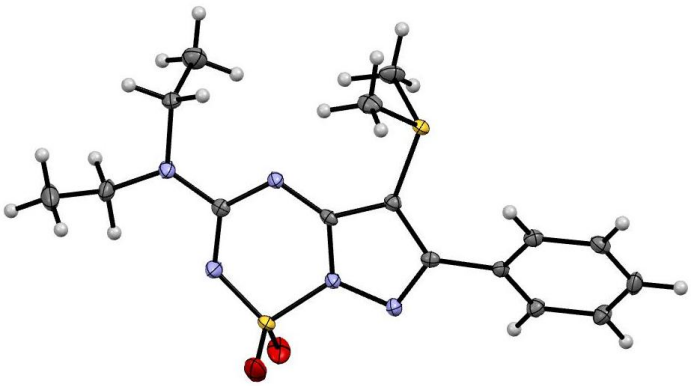
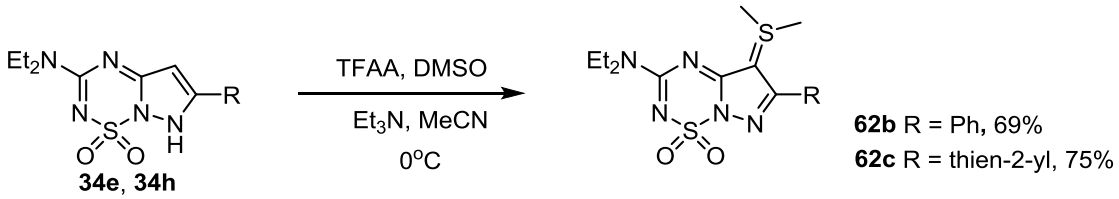


Fig 42. ORTEP diagram of **62a**. A molecule of DMSO has been removed for clarity.

This discovery prompted the synthesis of analogues of **62a** such as **62b** and **62c** (**Scheme 63**). At room temperature, the F_3CCO_2-S(+)Me_2 reagent was not stable, however at 0°C the product **62b** was obtained in in 69% yield and **34h** was utilised in a similar reaction to prepare **62c** in 75% yield.



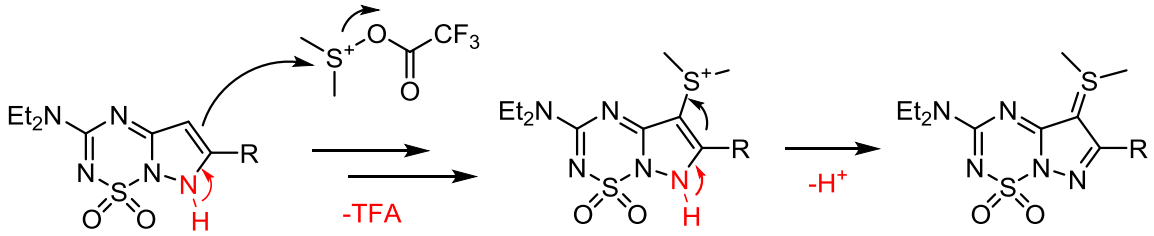
Scheme 63: Thiolation of substrate **34e, 34h** with DMSO under Swern-type conditions.

Curiously, when the reaction was attempted at -50°C, a complex yellow mixture was the result. Presumably, an oxidative decomposition of the precursor may have resulted, since it is known that at these cold temperatures, the DMSO-TFA reagent acts as an oxidising agent.[144,145] Since the products from this mixture were not adequately identified, this remains speculation. The result was not explored in further detail as the mechanisms of oxidative decomposition were not seen as beneficial to the research unless they could selectively generate a sufficient amount of isolable product(s).

4.6.1 Mechanism of dimethylthiolation

Nucleophiles such as alcohols and indoles[146] have been shown to react with the activated DMSO-trifluoroacetate molecule. Omura and Swern[145,147] published a detailed study of the interaction of DMSO with TFAA and proved that the resulting activated DMSO species is stable at low temperatures. Methylthio derivatives have also been reported as side products from reactions under Swern conditions whereby a nucleophile reacts with the electrophilic sulfur atom on the activated DMSO species.[144,148,149] A methylthiolation reported by Yang and co-workers[150] involved the attack of indole on activated DMSO to introduce a $–S(+)Me_2$ group which subsequently transformed to the –SMe by demethylation.

A mechanism by which nucleophilic attack of the pyrazole carbon of compounds **34** on the F_3CCO_2-$S(+)Me_2$ reagent followed by oxidation to regenerate the aromatic pyrazole system is proposed for the formation of these dimethylthio compounds **62** (**Scheme 64**).

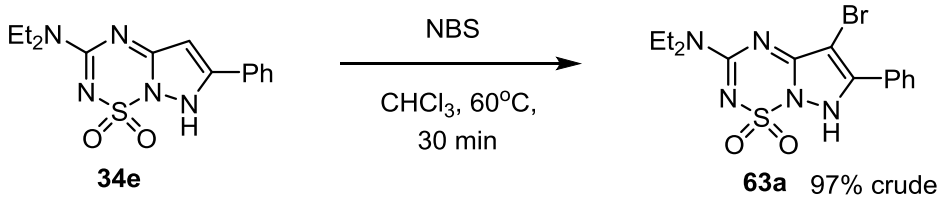


Scheme 64: The proposed mechanism of formation of dimethylthio-pyrazole derivatives from the activation of DMSO with TFAA.

This chemistry may have medicinal chemistry applications in instances where derivatives at a carbon nucleophile are to be investigated. Most readily prepared sulfur derivatives of carbon nucleophiles are sulfones or sulfoxides and so this simple and inexpensive method of dimethylthiolation has potential synthetic significance. Certainly, reactions such as these are not abundant in the literature, hence further exploration of the reactions of pyrazoles and fused pyrazole systems (privileged scaffolds in drug discovery) with the F_3CCO_2-S(+)Me_2 reagent would be valuable to the synthetic chemistry community.

4.7 Halogenation of the pyrazole ring

At this point, it had been established that the pyrazole carbon of compounds **34** exhibits considerable nucleophilic activity. Therefore it is plausible that a halogen might be installed at that site to furnish a potential reactive intermediate. A sample of **34e** was treated with N-bromo succinimide (NBS) in chloroform to generate the 5-bromo derivative in 97% yield (**Scheme 65**).



34e **63a** 97% crude

Scheme 65: Bromination of C5 with N-bromosuccinimide.

These derivatives were unstable to heat, light, particular solvents, and acidity; therefore they were not suitable synthetic intermediates.

A fluorine derivative was considered likely to be more stable compound than the analogous bromo or iodo derivatives (or one which would be less prone to side-reactions) and could be used to establish reactivity towards fluorination. The SelectFluor® reagent (**Figure 43**) is known to be an effective and versatile source of electrophilic fluorine.[151,152]

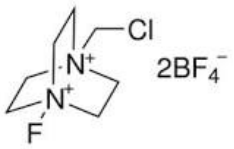
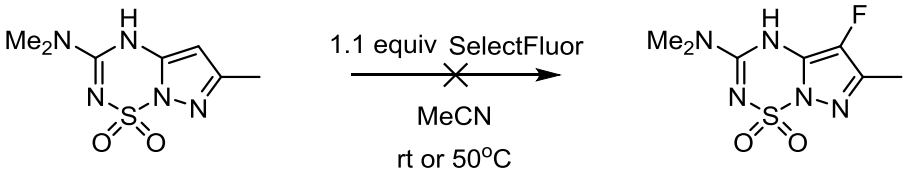


Fig 43. 1-Chloromethyl-4-fluoro-1,4-diazoniabicyclo[2.2.2]octane bis(tetrafluoroborate) (SelectFluor®)

A parent derivative **34j** was treated with the SelectFluor® reagent, unfortunately a fluoro derivative was not obtained after stirring with SelectFluor® in acetonitrile at room temperature or 50°C. No conversion of the starting material was detected (**Scheme 66**).



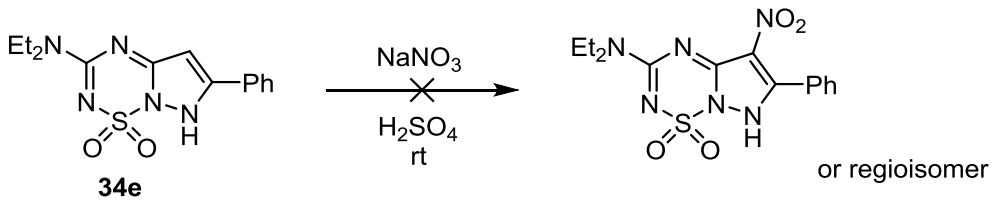
Scheme 66: Attempted fluorination of pyrazolothiatriazine derivative **34j** with SelectFluor®.

The SelectFluor® reagent presumably was not sufficiently activated to react with the substrate under the conditions employed. The aim of these experiments was to ascertain whether substrates **34** would react readily with a fluorinating agent. Since this reaction

would not proceed under these conditions, its use as a synthon might prove to be problematic and so the attempts were not made to further force the conditions.

4.8 Attempted Nitration of C5

Nitration of the nucleophilic carbon of substrate **34e** was attempted through treatment with stoichiometric quantities of aqueous sodium nitrate and sulfuric acid at room temperature. The result was a complex mixture as well as 42% recovery of the starting material after stirring at room temperature overnight (**Scheme 67**).



Scheme 67: Attempted nitration of compound **34e**.

These conditions were deemed too harsh for pyrazolothiatriazines **34** and this reaction was not pursued under any additional conditions. It was speculated at this point that the compounds **34** were stable to many of the basic conditions applied, but that the addition of acid has some problematic outcomes.

4.9 Conclusions

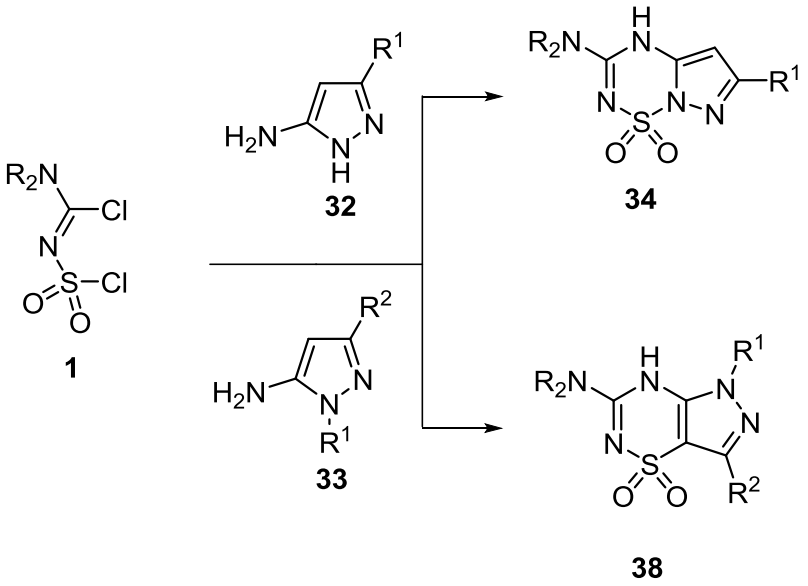
The carbon atom (C5) of the pyrazolo [1,5-*b*][1,2,4,6]thiatriazine ring system **34** has been demonstrated as a versatile carbon nucleophile towards a series of commercial electrophiles. The pyrazole carbon (or nucleophilic N atoms) did not appear to be readily susceptible to acylation with common electrophilic carbonyl compounds such as acid halides or anhydrides. In fact, under many of the conditions which were aimed at

synthesising an N-acyl derivatives using pyridine as an acyl transfer agent, only 5-(pyridin-4-yl) derivatives were obtained. Such reactions with pyridine and pyridazine occurred readily without a separate oxidation step, which contrasts with reports on several similar systems. Sulfonylation, thiolation and bromination reactions also occurred readily and selectively at C5. This led to the conclusion that despite the nucleophilicity of the ring N atoms dominating in alkylation experiments; other electrophiles such as tosyl chloride, formaldehyde, *N*-bromosuccinimide and DMSO were shown to react preferentially at C5. The ring system of compounds **34** also displayed a tendency to generate dimers if the carbon substituent was a good leaving group, such as tosyl, and this is likely to be a cause of the complex mixtures which formed from halogenated derivatives **63**.

CHAPTER 5: ALKYLATION OF PYRAZOLO[3,4-*E*][1,2,4]THIADIAZINES AND PYRAZOLO[1,5-*B*][1,2,4,6]THIATRIAZINES.

5.1 Introductory remarks

As aforementioned in Chapters 2 and 3, dichlorides **1** underwent regioselective reactions with 3-aminopyrazoles **32** and 1-substituted 5-amino pyrazoles **33** to give rise to the two fused ring systems: pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** and pyrazolo[3,4-*e*][1,2,4]thiadiazines **38**, respectively (**Scheme 68**).



Scheme 68: The selective formation of ring compounds **34** and **38** from reaction of dichlorides **1** with amino-pyrazoles **32** or **33** respectively.

The core ring structures of **34** and **38** represent a new and a very rare heterocyclic ring system, respectively. Tautomerism of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** stipulates that the NH moiety can exist at N2, N4, or N7 and thus these positions have potential for the introduction of various substituents (**Figure 44**).

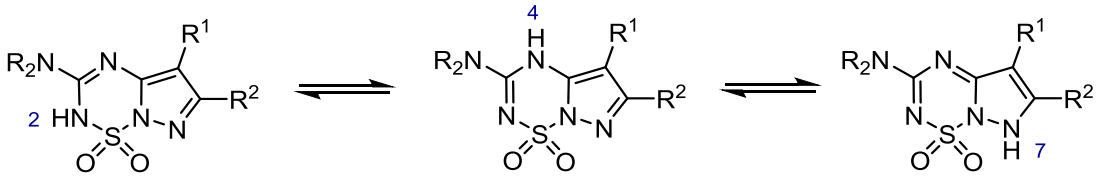


Fig 44. Tautomeric forms of the pyrazolo**[**1,5-*b*][1,2,4,6]thiatriazines **34**.

Similarly, pyrazolo[3,4-*e*][1,2,4]thiadiazines **38** may form three possible tautomeric structures which indicates three prospective nucleophilic NH sites at either N2, N4, or N6 for reaction with electrophilic reagents (**Scheme 70**).

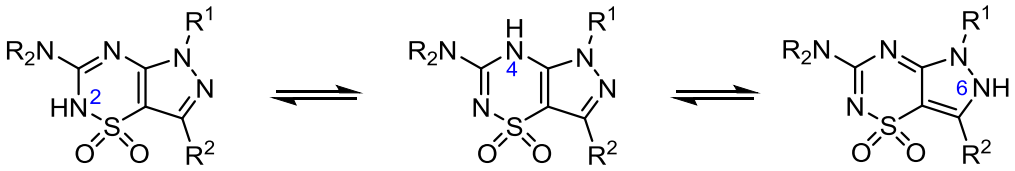
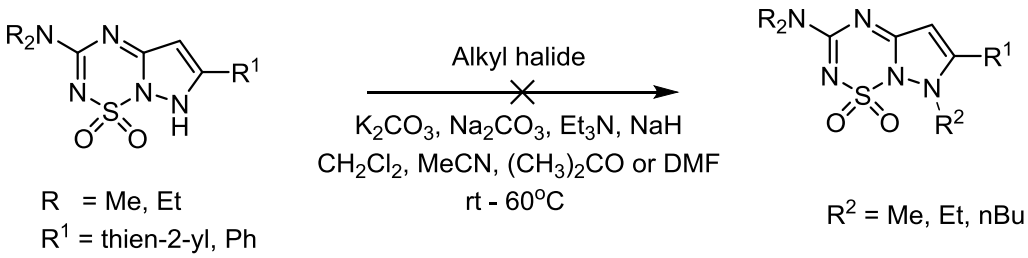


Fig 45. Tautomeric forms of the pyrazolo[3,4-*e*][1,2,4]thiadiazines **38**.

The potential for substitution reactions with compounds **34** and **38** has not been explored. Research to establish the suitability of each ring system as a template/scaffold for production of a library of novel derivatives was of interest in order to demonstrate their use as potential drug candidates. Such studies would aim to reveal the nucleophilicity of each ring system towards different classes of alkylating agents. This would be a beneficial endeavour because heterocyclic systems containing thiatriazine and thiadiazine rings are rare in the literature. The formation of *N*-substituted derivatives would represent an efficient synthetic approach to a novel compound library, allowing these new heterocyclic templates to find use in bioactive molecule discovery programs. Therefore, in addition to further exploring the nucleophilicity of C5, it was important to determine if the nitrogen atoms in the pyrazolothiatriazine core structure of **34** and **38** possessed a significant degree of nucleophilicity. This could be evaluated by investigating the treatment of some representative compounds with a variety of electrophilic reagents.

5.2 Alkylation Reactions of fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1 dioxides

Despite the surprising intrinsic lack of reactivity of the NH moiety towards common acylating agents, the susceptibility to alkylation of ring systems **34** and **38** was subsequently investigated. Attempts to alkylate the fused pyrazolothiatriazine compounds **34e**, **34d** and **34h** with iodomethane, iodoethane, or 1-bromobutane in the presence of sodium carbonate in acetone or acetonitrile between room temperature and 40°C resulted in return of the starting materials. Switching to the organic amine base triethylamine under similar conditions or with DMF at 60°C also led to return of the precursor compound. Samples of **34e** were treated with (1) 1.5 equivalents of ethyl iodide or methyl iodide and 2 equivalents of sodium hydride in dichloromethane and stirred at room temperature, (2) 1.1 equivalents of ethyl iodide and potassium carbonate with 10mol% Bu^n_4NBr in acetonitrile at room temperature, (3) 2.0 equivalents of ethyl iodide and potassium carbonate with 20mol% Bu^n_4NBr in DMF at 50°C (**Scheme 69**).

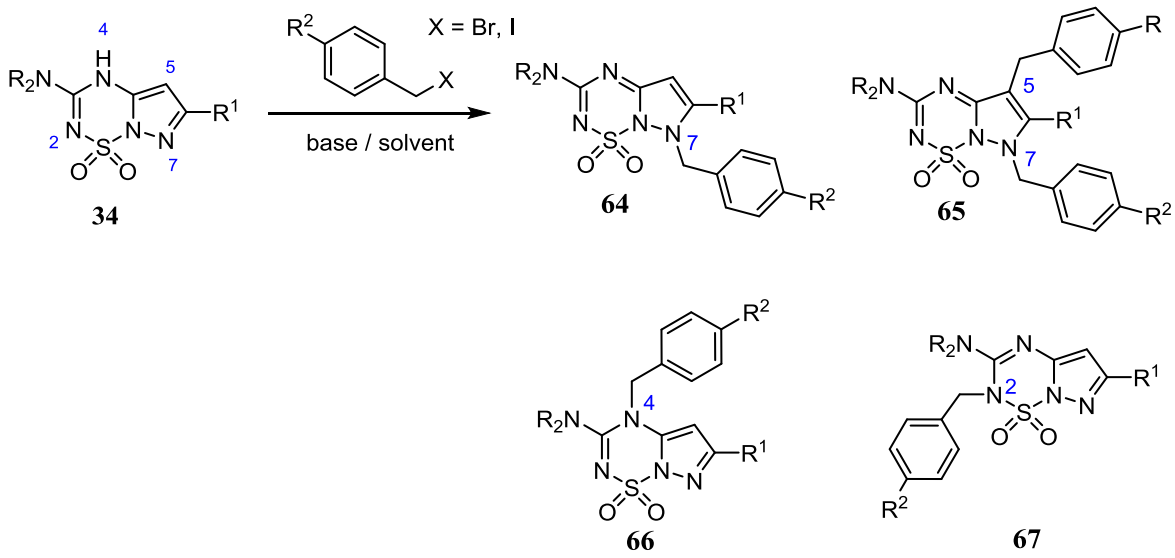


Scheme 69: Attempted alkylation of pyrazolothiatriazines **34d, 34e** or **34h**.

In all cases, the starting material was recovered. Although trace amounts of a suspected alkylated compound were observed in some instances, the low quantity meant that full characterisation and isolation were not practicable. It became apparent that these alkylating agents were not sufficiently electrophilic to react with pyrazolothiatriazines **34.**

5.2.1 Benzylation

A series of pyrazolothiatriazines **34** were treated with benzylic halides and a base under conditions which have been successfully employed with related ring systems.[24,27] In every case, these reactions afforded the N7 alkylated regioisomer **64** as the major product (**Scheme 70**). The regioselectivity was not greatly influenced by solvent or base, or by substitution on the pyrazole ring. Pyrazole carbon C5 also reacted as a nucleophile to afford bis-benzylated compounds **65**. Minor proportions of 4-benzylated products **66** and 2-benzyl compounds **67** were isolated in several cases. Results are summarised in **Table 8**.



Scheme 70: The alkylation of compounds **34** with benzylic halides.

Table 8: Synthesis of pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **64-67**.

R_2N	R^1	R^2	Method	Product(s)	Yield(s) [%]
Me_2N	Ph (**34d**)	Cl	A	**64a***	49
			B	**64a*** + **65a***	60 + 15

Et_2N	Ph (**34e**)	Cl	C	**64b*** + **65b**	86 + 1
			D	**64b*** + **65b** + **66b**	73 + 10 + 1
Me_2N	Thien-2-yl (**34g**)	Cl	B	**64c*** + **65c**$^{*\#}$ + **66c**$^{*\#}$ + **67c**	46 + 13 + 4 + 1
			C	**64c*** + **65c**$^{*\#}$ + **66c**$^{*\#}$	77 + 13 + 3
Et_2N	Thien-2-yl (**34h**)	Cl	C	**64d*** + **65d*** + **66d**	72 + 20 + 1
		H	C	**64e*** + **65e** + **66e***	69 + 14 + 2
			D	**64e*** + **65e** + **66e***	72 + 6 + 12
Me_2N	Me (**34j**)	H	C	**64f** + **65f** + **66f** + **67f**	60 + 11 + 6 + 2
Et_2N	Thien-2-yl (**34e**)	OMe, X=Br	C	**64g*** + **65g*** + **66g*** + **67g**	44 + 8 + 16 + 4
		OMe, X=I	C	**64g*** + **65g*** + **66g*** + **67g**	26 + 10+ 12+ 5

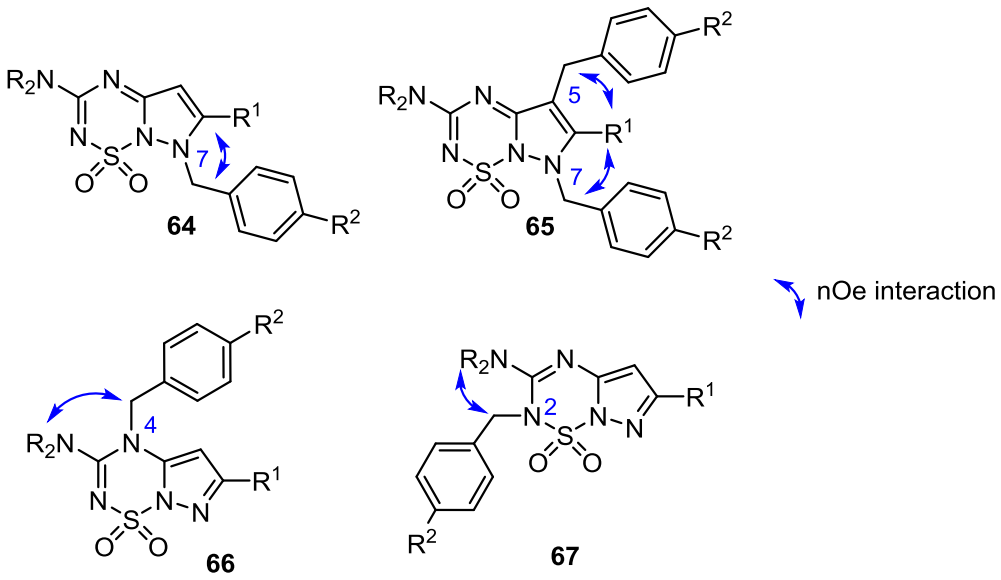
$^{\#}$X-ray crystal structure obtained, *NOESY experiments conducted. Methods: A: K_2CO_3, cat. Bu^n_4NBr, CH_2Cl_2; B: Et_3N, THF; C: K_2CO_3, cat. Bu^n_4NBr, MeCN; D: K_2CO_3, cat. Bu^n_4NBr, MeCN, MWI 50^{o}C.

The most effective conditions employed were potassium carbonate with catalytic tetrabutylammonium bromide (Bu^n_4NBr) in acetonitrile. When the solvent was changed to tetrahydrofuran or dichloromethane, low substrate solubility slowed reaction progress. Microwave irradiation (MWI) to 50°C allowed conversion within 2-3 hours compared to reaction times of 1-2 days when stirring at room temperature; although selectivity was similar.

The reaction of 4-methoxybenzyl chloride in place of benzyl bromide or 4-methoxy benzyl bromide was very slow with only ~10% conversion after 4 days at room temperature.[153] A sample of 4-methoxybenzyl iodide was synthesised via a Finkelstein reaction of 4-methoxybenzyl chloride with sodium iodide[154] and reacted with substrate **34h** under method C conditions. The combined yield from this reaction including all isomers was 52% with recovery of the starting material; and this was attributed to the fact that the 4-methyoxybenzyl iodide, while very reactive,[155] is thermally and photochemically unstable.[156] The synthesis of 4-methoxybenzyl derivatives **64g**-**67g** was successful; however these compounds were prone to decomposition and proved difficult to handle and purify. The resultant decomposition products are specified in Chapter 6.

Substrates containing exocyclic diethyl amino groups provided higher yields compared to the analogous dimethylamino counterparts, which was likely due to the greater solubility of the substrates in the reaction mixture.

The structural assignments for products **64**-**67** were evidenced by 2D NOESY NMR spectroscopic experiments. Key nOe interactions were observed between the benzylic methylene protons and protons from substituents on the pyrazole ring for compounds **64** and **65**, or with those flanking the dialkylamino groups for compounds **66** and **67** (**Scheme 71**).



Scheme 71: The characteristic nOe interactions from 2D NOESY experiments of compounds **64-67**.

Notably, isomers **66** and **67** could conceivably give rise to the same nOe interaction since the N4 benzyl and N2 benzyl substituents are close in space to the dialkylamino groups. NMR experiments alone would not allow the unambiguous structural elucidation of these two isomeric products. The NMR evidence was thus combined with X-ray crystallographic analyses of representative compounds **65c** and **66c** (**Figure 46**). A more detailed discussion of these X-ray results can be found in Chapter 8. Consistent trends in TLC mobility, NMR chemical shifts, and solubility supported the structural assignments of the regioisomeric products in each case.

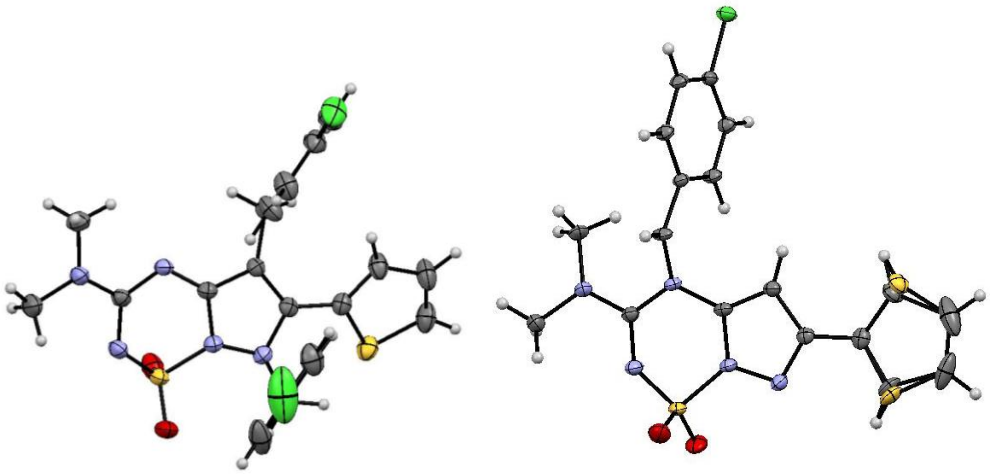


Fig 46. ORTEP diagrams of **65c** and **66c**. A dichloromethane molecule in the crystal lattice of **65c** has been omitted for clarity. The crystal structure of **66c** is disordered around the sigma bond of the thiophene moiety.

A consistent trend between the site of benzylation and the NMR signals corresponding to the protons on the exocyclic dialkylamino substituent was observed in compounds **64-67** (**Figure 47**). Products **64** and **65** which contain benzyl groups on the pyrazole ring showed different NMR environments for each alkyl group of the dialkylamino moiety. Products containing benzyl substituents on N4 and N2 positions (compounds **66** and **67**) presented single, broad signals for the two alkyl groups, representing an average of two environments. These trends were used to assist with structural assignment. This phenomenon of two NMR environments for dialkylamino substituents as a consequence of contributions from zwitterionic resonance forms which restrict rotation has been observed previously in related ring systems.[22]

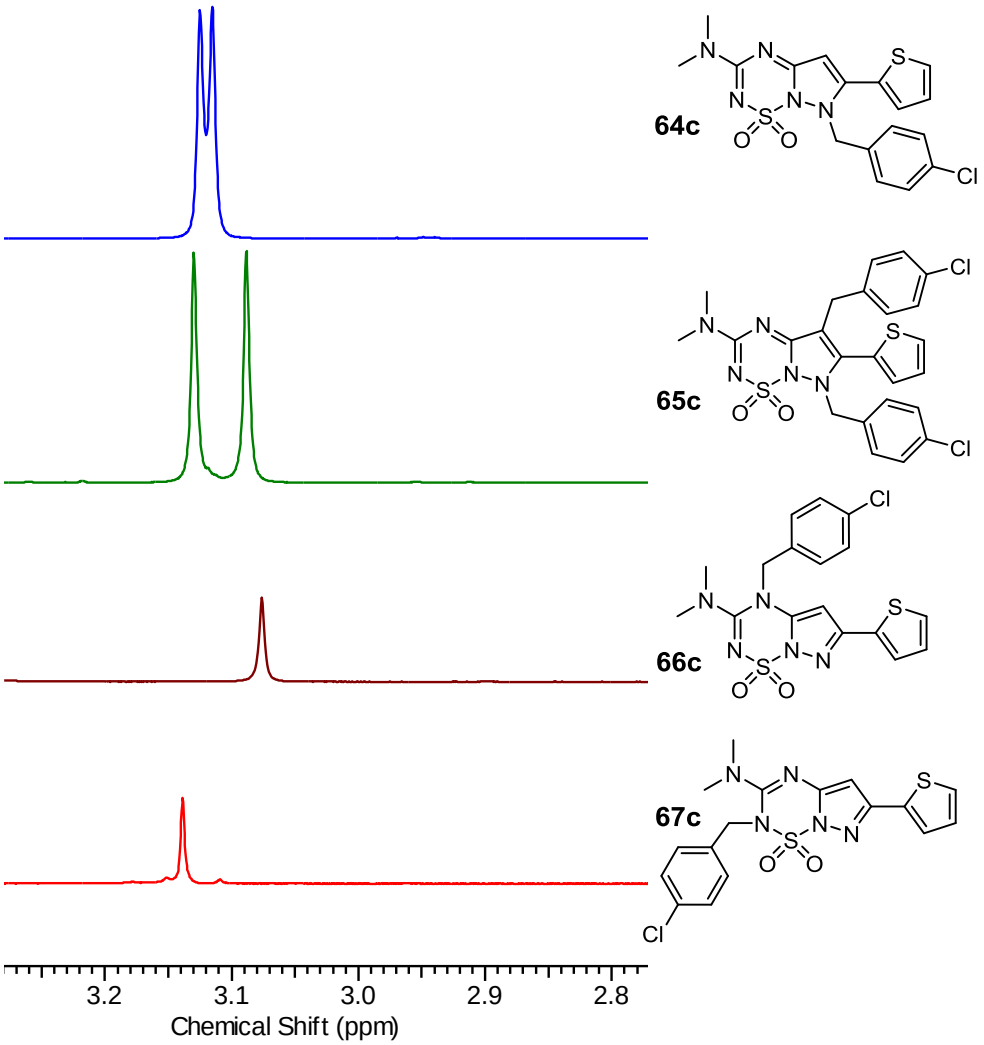


Fig 47. Dimethylamino resonances in the 400MHz, ^{1}H NMR spectra of **64-67c**.

The disparity between the different sites of alkylation and the NMR signals may also be explained from a perspective of planarity. The crystal structure of **65c** however does not indicate any significant difference between the shape of the thiatriazine ring compared to that of **66c**; however, this is in the solid state and not in solution and the characteristics of these products in $CDCl_3$ solution may differ. It could be the case that the dialkylamino substituent in **66c** rotates relatively freely in solution and the dimethylamino signals are an average of the two environments. The zwitterionic contributions may be more substantial in compounds **64** and **65** to give a partial double bond character of the bond between the exocyclic amine and the thiatriazine ring. A

larger resonance contribution from this zwitterion would lead to different chemical environments for the two alkyl substituents (**Figure 48**).

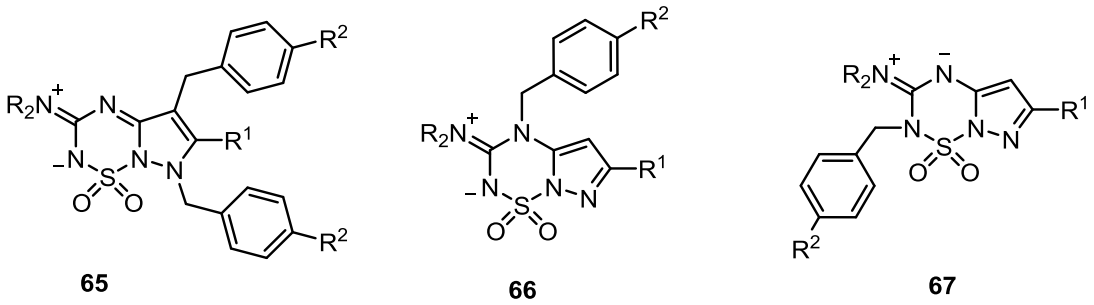
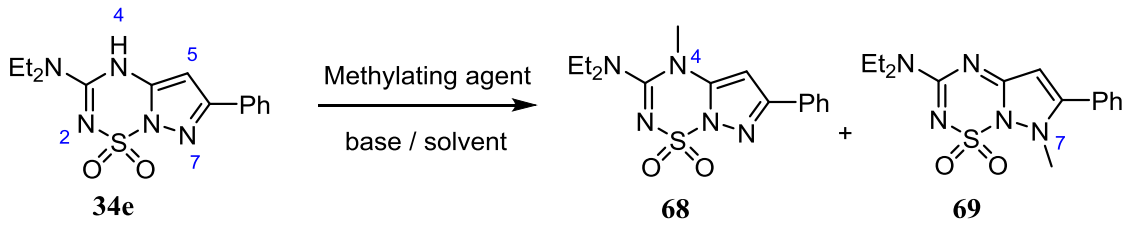


Fig 48. Zwitterionic structures of compounds **65**, **66** and **67**.

5.2.2 Methylation

The successful benzylation reactions described above encouraged investigation of the reactivity of the new ring system **34** towards other common alkylating agents. The 4-methyl derivative **68** and the 7-methyl derivative **69** were produced from reaction of pyrazolothiatriazine **34b** with methyl tosylate (MeOTs) or dimethyl sulfate (Me_2SO_4) (**Scheme 72**, **Table 9**).



Scheme 72: Methylation of fused pyrazolothiatriazine **34e**.

Table 9: Synthesis of methyl-substituted pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **68** and **69**.

Methylating agent	Conditions	Product 68	Yields [%] 69
MeOTs	Na_2CO_3, MeCN, 55°C, 3 days	28	1*
	Na_2CO_3, $CHCl_3$, 50°C, 2 days	2	18#
Me_2SO_4	Na_2CO_3, MeCN, 55°C, 8 hours	60	39
	Na_2CO_3, $CHCl_3$, 50°C, 6 hours	30	65

*__34e__ recovered in 64% yield; #__34e__ recovered in 76% yield

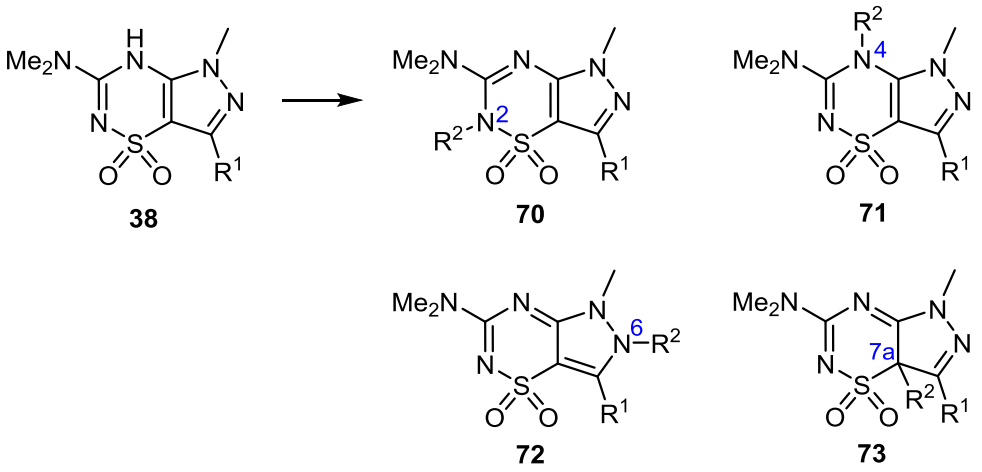
Structural assignments for compounds **68** and **69** were based on 2D NOESY NMR experiments. In the case of 4-methyl compound **68**, a definitive nOe cross peak was observed between the N-methyl resonance at δ 3.52ppm and the H-5 resonance at δ 6.16ppm. With 7-methyl compound **69**, a definitive nOe interaction was observed between the N-methyl resonance at δ 3.86ppm and resonances from the 6-phenyl group at approximately δ 7.5ppm.

Compounds **68** and **69** formed slowly over two to three days from alkylation by methyl tosylate; and low yields were also accompanied by recovery of the majority of the starting material. A change of solvent to DMF or DMSO, which solvate the fused thiatriazines **34** more effectively, was not desirable because these solvents could become reactive with methylating agents at temperatures greater than 100°C. Alternative methods utilising Me_2SO_4 proved to be more effective with near-quantitative conversion, although elevated temperatures were required for product formation to occur within a practical time-frame. Interestingly, the choice of solvent had a major influence on the regioselectivity of the methylation reactions. The 4-methyl

derivative **68** was the major product in the polar aprotic solvent acetonitrile, whereas the use of chloroform as solvent favoured formation of the 7-methyl product **69** (**Table 9**). This solvent dependence stands in contrast to the benzylation experiments summarized in **Table 8**, in which the product ratio was apparently independent of solvent.

5.3 Alkylations of pyrazolo[3,4-e][1,2,4]thiadiazine 1,1 dioxides

Representatives of fused pyrazolo-thiadiazines **38** were treated with benzylic halides and dimethyl sulfate following conditions employed in with pyrazolo[1,5-*b*][1,2,4,6]thiatriazines (**Scheme 73**, **Table 10**).



Scheme 73: Methylation of the three nucleophilic nitrogen atoms of compounds **38.**

Table 10: Synthesis of alkylated pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides **70-73**.

R^1	R^2	Method	Product(s)	Yield [%]
Me (**38b**)	Me	Me_2SO_4, Na_2CO_3, DMF, 50°C	**70a**[#~]	47
		Me_2SO_4, K_2CO_3, DMSO,	**70a**[#~]	31

		80°C		
		Me_2SO_4, K_2CO_3, MeCN, reflux	**70a**[#~] + **71a** + **72a**[#]	14 + 15 + 65
Thien-2-yl (**38h**)	Me	Me_2SO_4, K_2CO_3, MeCN, reflux	**70b** + **71b**[#] + **72b**[~]	30 + 30 + 21
Me (**38b**)	4-ClBn	4-ClBnBr, K_2CO_3, cat. Bu^n_4NBr, MeCN, 60°C	**70c*** + **71c**[~] + **73c**	47 + 5 + 31
Thien-2-yl (**38h**)	4-ClBn	4-ClBnBr, K_2CO_3, cat. Bu^n_4NBr, MeCN, 40°C	**70d*** + **71d** + **73d**	67 + 16 + 7

[#]X-ray crystal structure obtained, *2D NOESY interactions observed, ~2D HETCOR experiments conducted

During methylation reactions with dimethyl sulfate, it was noted that some of the isomers formed (namely **71** and **72**) had significant water solubility; hence to minimize isolation problems, acetonitrile was used as solvent in place of dimethylformamide or dimethyl sulfoxide. Minimal addition of water was used to quench the dimethyl sulfate and a methanol/dichloromethane or methanol/ethyl acetate extraction was performed on workup.

Methylation of **38b** and **38h** was not very selective and a mixture of products was obtained in each case. Methylation of substrate **38b**, with a (relatively small) methyl subsituent at C7, resulted in a higher yield of N6 methylated product **72a** (65%) than with substrate **38h**, with a 7-(thien-2-yl) substituent (**72b**; 21%).

Alkylation of **38b** and **38h** with 4-chlorobenzyl bromide occurred at N2 and N4; preferentially at N2, providing products **70**, and to a lesser extent at N4, giving **71**. Interestingly, benzylation also occurred at the ring junction carbon C7a, adjacent to the SO_2 moiety, affording compounds **73**. Initially, the N6-alkylated structure **72** was assigned to compounds **73c** and **73d**, but the NMR data were not compatible with structure **72**. In each case, an AB quartet at ~3.5 ppm in the ^{1}H NMR spectrum and a resonance at ~70 ppm in the ^{13}C NMR spectrum were observed. These data are

consistent with structure **73**, which bears a benzyl substituent on an asymmetric, quaternary carbon C7a.

As might be expected, the compound with less steric hindrance at C7 produced a greater proportion of alkylation at C7a, furnishing isomers **73** (**73c**; 31%, compared with **73d**; 16%).

The observation that all three possible *N*-alkylated products and a C-alkylated product could be isolated was interesting; however, the purpose of the present study was simply to establish that the core ring system could undergo substitution reactions.

The methyl groups in products **70-72** were not in close proximity to the protons flanking other alkyl substituents and hence, nOe data could not be used effectively to elucidate regioisomers of these alkylated products. It was therefore necessary to collect an X-ray crystal structure of an example of each **70, 71**, and **72** (**Figure 49**).

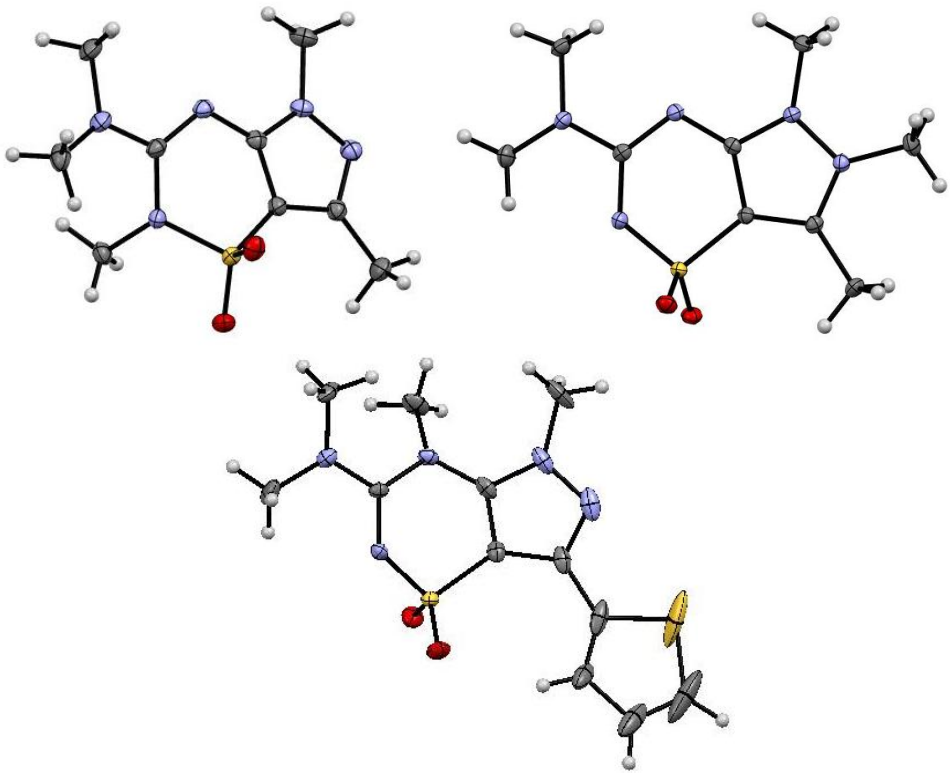
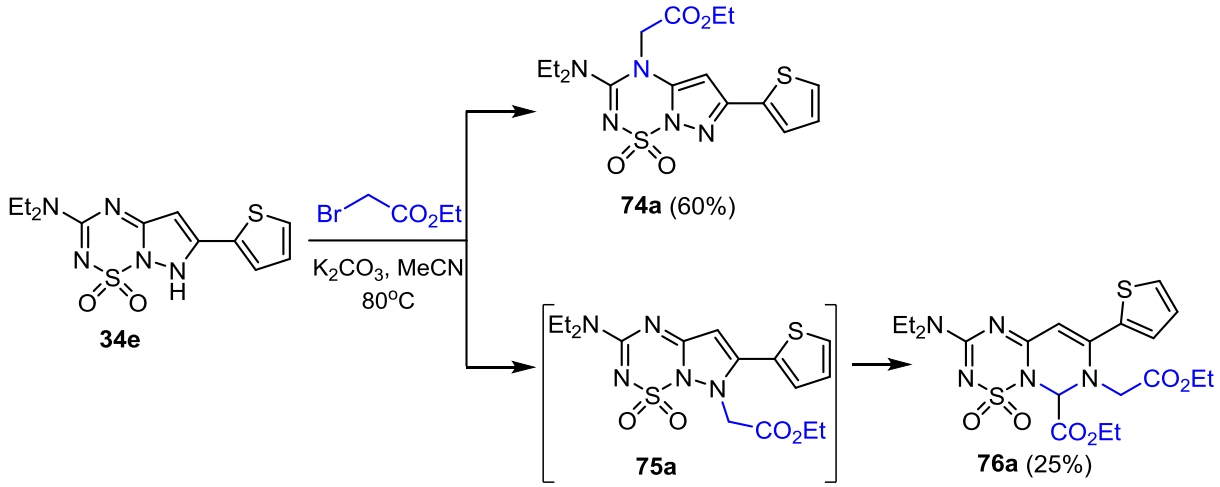


Fig 49. ORTEP diagrams of methylated derivatives **70a** (top left), **72a** (top right) and **71b** (bottom) for structural elucidation.

However, the NOESY spectra of N-benzylated compounds **70c**-**71d** displayed nOe crosspeaks which enabled structural assignments. N2-benzylated compounds **70c** and **70d** presented nOe crosspeaks between the methylene singlets at δ 4.58 ppm and δ 4.67 ppm, and the dimethylamino resonances at δ 3.10 ppm and δ 3.46 ppm, respectively. The NOESY spectra of N4-substituted products **71c** and **71d** contained weak nOe correlations between the methylene singlets at δ 4.93 ppm and δ 4.91 ppm, and the pyrazole ring N-methyl signals at δ 3.74 ppm and δ 3.07 ppm, respectively. The same methylene signals weakly correlated to those of the dialkylamino substituent at δ 3.07 ppm and δ 3.06 ppm, respectively. Compounds **73c** and **73d** did not present any convincing nOe crosspeaks.

5.4 Ring expansions of fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazines

An unexpected result followed from an experiment aimed at N-alkylation of pyrazolothiatriazine **34d** with ethyl bromoacetate and potassium carbonate in acetonitrile (**Scheme 74**). In addition to a 60% yield of the expected N4-alkylated product **74a**, a second compound containing two acetate substituents was isolated from this reaction in 25% yield. X-Ray crystallographic analysis revealed the pyrimido-thiatriazine structure **76a** (**Figure 50**), presumably formed from ring expansion of the pyrazole moiety of N7-alkylated compound **75a**. The structure of N4-alkylated product **74a** was also established by X-ray crystallography (**Figure 50**).



Scheme 74: Treatment of compound **34d** with ethyl bromoacetate.

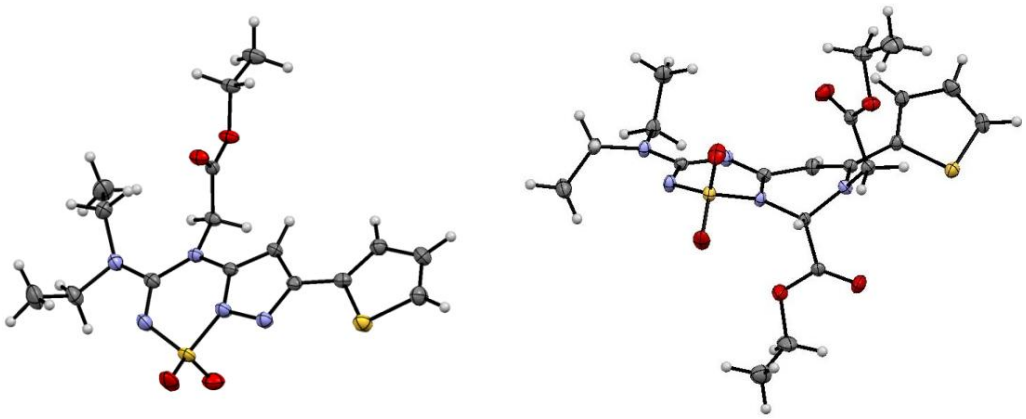
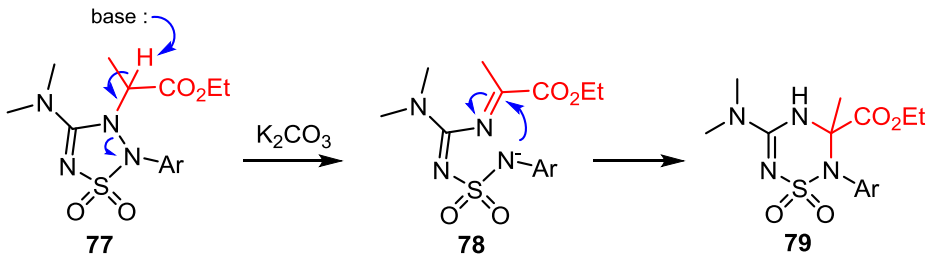


Fig 50. ORTEP diagrams of **74a** (left) and **76a** (right).

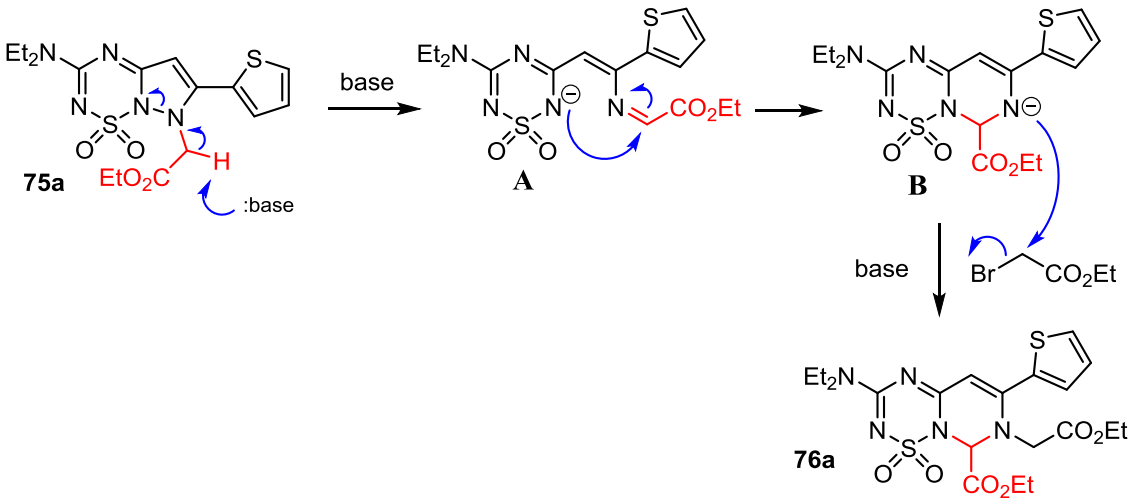
Compound **76a** is the first reported example of the pyrimido[1,6-*b*][1,2,4,6]thiatriazine ring system. Ring expansion of a pyrazole moiety by means of an alkylating agent had not been reported previously in the literature; however, Duggan and co-workers[157] described related ring expansion reactions of [1,2,3,5]-thiatriazoles with α-halo esters. The ring systems are similar and it appears that the mechanism of pyrazole ring opening has some similarity to that of the thiatriazole ring expansion in **77** to form thiatriazine rings **79** (**Scheme 75**).



Scheme 75: Ring expansion of 4-amino 1,1-dioxo [1,2,3,5]thiatriazole **77** upon alkylation with methyl 2-bromo propanoate.

5.4.1 Mechanism of formation

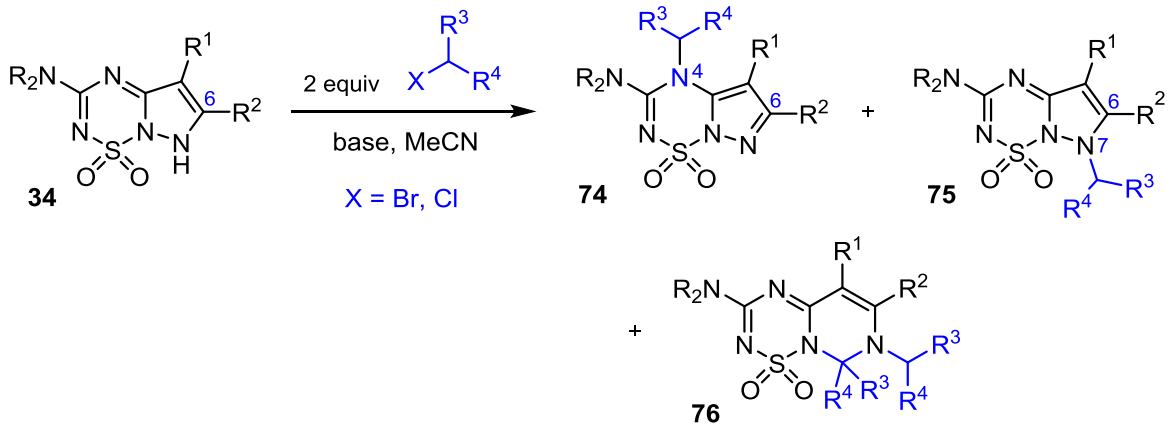
By analogy with the mechanism proposed for the thiatriazole ring expansion reactions reported by Duggan and coworkers,[157] an account for formation of **76a** is outlined in **Scheme 76**. Both the electron withdrawing nature of the alkyl substituent on N7 and the sulfamide moiety of the core ring system of **75a** contribute to the acidic nature of the α-proton which could be removed under basic conditions to form an imine, with simultaneous cleavage of the pyrazole ring to form sulfamide anion **A**. The pyrimidine ring subsequently forms via addition of the sulfamide nitrogen to the imine carbon to give intermediate **B** which is alkylated again to provide the fused pyrimidine **76a**.



Scheme 76: The proposed mechanism of formation of pyrimido[1,6-*b*][1,2,4,6]thiatriazine product **75a**.

5.4.2 Substrate scope

This unusual result prompted investigations into the scope of this rare ring-expansion reaction; the results are summarised in **Scheme 77** and **Table 11**.



Scheme 77 (above), **Table 11** (below): Synthesis of N-alkylated pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides (**74** and **75**) and dihydropyrimido[1,6-*b*][1,2,4,6]thiatriazine dioxides (**76**).

R_2N	R^1	R^2	R^3	R^4	Conditions†	Product(s)*	Yield [%]
Et_2N	H	Thien-2-yl (**34h**)	CO_2Et	H	K_2CO_3, cat. Bu^n_4NBr, 80°C	**74a** + **76a**#	60 + 25
Me_2N	H	H (**34a**)	CO_2Et	H	K_2CO_3, cat. Bu^n_4NBr	**74b** + **76b** + **74b**#	25 + 4 + 31
					K_2CO_3, cat. Bu^n_4NBr, 80°C	**74b**	25
Me_2N	H	Me (**34j**)	CO_2Et	H	K_2CO_3, cat. Bu^n_4NBr	**74c** + **76c**#	24 + 40
					$^iPr_2NEt$	**74c** + **76c**#	40 + 33

Me_2N	CO_2Et	H (**34i**)	CO_2Et	H	K_2CO_3, cat. Bu^n_4NBr	**74d** + **75d** + **76d**[#]	12 + 1 + 34
Me_2N	H	Cyclopropyl (**34k**)	CO_2Et	H	K_2CO_3, cat. Bu^n_4NBr	**74e** + **76e**	40 + 31
Me_2N	H	H (**34a**)	4-bromo phenacyl	H	K_2CO_3	**74f**	18
Me_2N	H	Me (**34j**)	CO_2Me	CO_2Me	K_2CO_3, cat. Bu^n_4NBr	**76g**[#]	25

†Reactions performed in MeCN at 50°C, unless otherwise specified; *All product structures confirmed by NOESY experiments with the exception of **76g**; [#]Structural assignment based on HMBC and HMQC experiments.

Conditions employed generally involved mild heating in acetonitrile since no appreciable reaction conversion was observed at room temperature or in dichloromethane. In the case of substrates **34a** and **34j**, which lacked a bulky substituent at C6, reaction with ethyl bromoacetate and dimethyl chloromalonate resulted in the ring-expanded compound **76b-c** as the major product as well as low yields of (ring-expansion precursor) N7-alkylated products **75b-c**. Substrates with bulky substituents at C6 led to a greater proportion of alkylation at thiatriazine nitrogen N4, presumably due to steric hindrance at N7. This illustrates the effects of substrate substitution patterns on the propensity towards alkylation of the pyrazole ring in preference to the thiatriazine ring. A pyrazolothiatriazine **34i** containing the electron withdrawing carbethoxy substituent at the C5 position reacted with ethyl bromoacetate to give rise to a higher proportion of the fused pyrimidine species **76d** in 34% yield compared to compound **74d** which only formed in 12% yield. The bulk of the crude material consisted of unknown yellow mixtures from which no pure products could be isolated. The ester substituent on the pyrazole ring could be affecting the intermediate **A** (**Scheme 76**) by accelerating its formation or increasing the δ^+/electrophilicity of the imine carbon leading to a greater ratio of ring expansion to N4 alkylation. Unfortunately, no appreciable quantities of **75** (the precursor to **76**) were isolated and so experiments to

prove the ring expansion mechanism by treatment of a representative of **75** with a single equivalent of ethyl bromoacetate and base could not be performed.

The electrophiles 2-chloroacetamide, 2-bromoacetamide, ethyl 2-bromobutyrate, ethyl 2-chloroacetoacetate, and ethyl 2,2-dichloroacetate were unreactive towards compounds **34** over long periods of time with potassium carbonate in acetonitrile or DMF at 60-80°C.

Products **74-76** were characterised by 2D NOESY experiments combined with consistent NMR chemical shifts and relative TLC mobilities. During the course of these preparations, it became apparent that excessive heating and lengthy reaction times all served to accelerate the decomposition of the ring expanded compounds. Each reaction was hence monitored by TLC until the consumption of starting material, at which time the reaction was quenched.

To investigate the scope of this rare ring-expansion reaction, compounds **34** were treated with another class of electrophile, the phenacyl bromides. These were expected to react similarly to ethyl bromoacetate and potentially afford a ring expanded product if substitution on N7 was favoured. The reaction of **34a** with 4-bromophenacyl bromide and potassium carbonate at 50°C provided only the N4-alkylated product **74f** in low yield (18%). The remainder of the crude material was a complex yellow mixture from which no starting material or other pure products could be isolated. A sample of **34a** was treated with phenacyl bromide and potassium carbonate at 90°C, resulting in a similar assortment of products which could not be adequately purified by column chromatography. No compounds could be isolated from the product mixture when **34h** or **34a** were treated with 4-bromo phenacylbromide and either sodium bicarbonate or potassium carbonate in DMF at room temperature. A sample of **34a** was also treated with phenacyl bromide at room temperature with potassium carbonate in DMF; however, no compound(s) could be purified from the resultant mixture.

5.4.3 General characteristics of pyrimido[1,6-b][1,2,4,6]thiatriazine dioxides

N4-Substituted compounds **74** were stable crystalline solids, however pyrimido-thiatriazines **76** (with the exception of **76e**) were obtained as off-white semi-solids or oils which gradually decomposed when stored in solution or in air. Analysis of an NMR sample solution of fused pyrimido-thiatriazine **76a** in deuterated chloroform revealed a gradual transformation to an unknown derivative upon standing in air (**Figure 51**, **Appendix B**). The ratio of unknown derivative to the original compound was approximately 0.6:1. Attempts to allow this transformation to reach full conversion by the addition of 1M aqueous hydrochloric acid led to a mixture of degradation products. Further analysis of the NMR data suggested that the H8 proton of the pyrimidine ring had disappeared from the region of the ^{1}H NMR spectrum at ~δ 5.8-6.0 ppm. All other signals which are due to the same functional groups of the precursor compound were accounted for, and had simply shifted slightly (within δ ±0.5 ppm). **Figure 51** illustrates the disappearance of the singlet due to the H8 proton and also the emergence of an additional AB quartet at δ ~3.4ppm. This was observed in conjunction with the AB quartet resulting from the methylene protons of the ester substituent adjacent to the thiophene ring, which implied that the cleavage of the pyrimidine ring could have occurred.

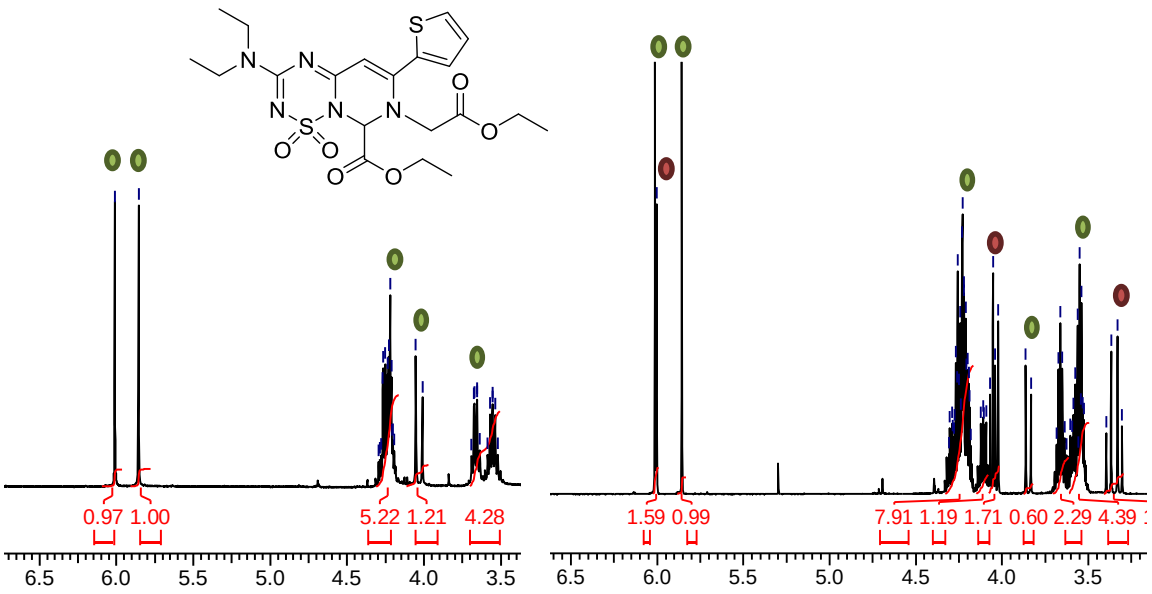
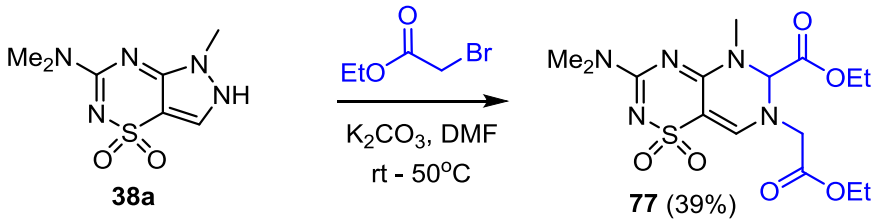


Fig 51. ^{1}H NMR spectrum of **76a** (left) and an expanded region from δ 8.0-3.0 ppm of the ^{1}H NMR spectrum of **76a** upon standing in $CDCl_3$ (right) (600MHz, $CDCl_3$)

Efforts to isolate this compound from column chromatography led to further degradation along with co-elution of the precursor compound with the new, unknown decomposition product. Further analysis by mass spectrometry was not informative and we were unable to identify the degradation product. However, the fused pyrimidine compounds **76** were stored cold and under a nitrogen atmosphere, with minimal exposure to acidity. Column chromatography of crude product mixtures from the 'ring expansion' reactions was carried out using buffered silica gel[2] (pH = 7) to minimize unwanted transformations. Deuterated chloroform for NMR analysis was stored over potassium carbonate and silver foil under a nitrogen atmosphere in the dark.

5.5 Ring expansions of fused pyrazolo[3,4-e][1,2,4]thiadiazines

The pyrazolo[3,4-*e*][1,2,4]thiadiazine ring system **38** was treated with ester containing alkylating agents to compare the regioselectivity with that of the isomeric compounds **34**. Compounds **38** were considerably less soluble than compounds **34** and a more powerful solvent (DMF) was utilised in place of acetonitrile to allow reaction to occur within a satisfactory time frame. Upon treatment of compounds **38** with ethyl bromoacetate, a ring expansion of the pyrazole ring to form a pyrimidine ring was encountered (**Scheme 77**).

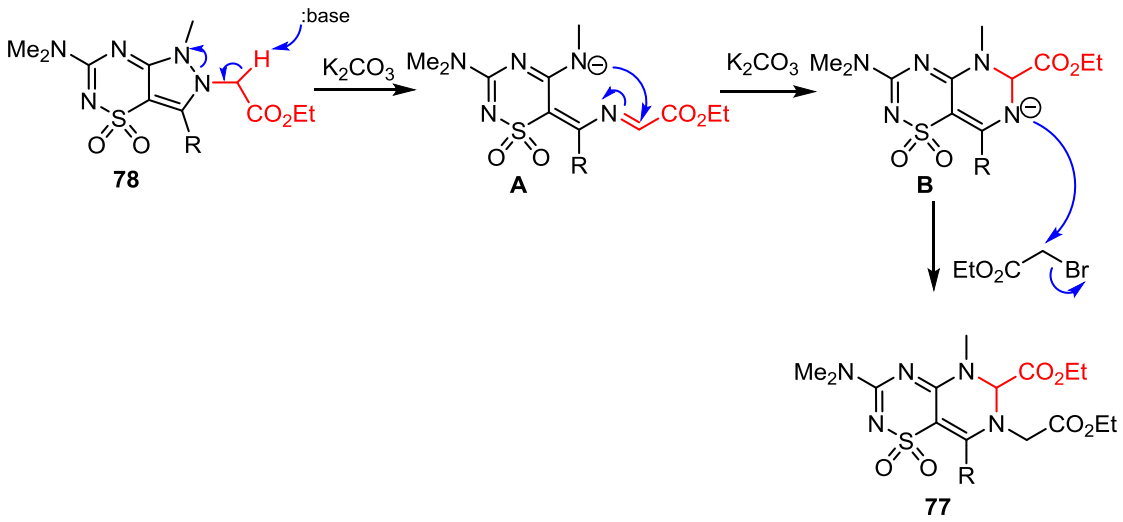


Scheme 77: Treatment of **38a** with ethylbromoacetate.

[2] Silica gel was added to deionised water (2.5:1w/v) with 1 phosphate buffer tablet (pH = 7.0) per 100mL of water. The mixture was stirred overnight while being allowed to dry in air.

5.5.1 Mechanism of formation

A proposed mechanism for the formation of these compounds is outlined in **Scheme 78**. This follows a similar pathway to that of the pyrazolothiatriazine **34** ring expansions illustrated in **Scheme 76**.

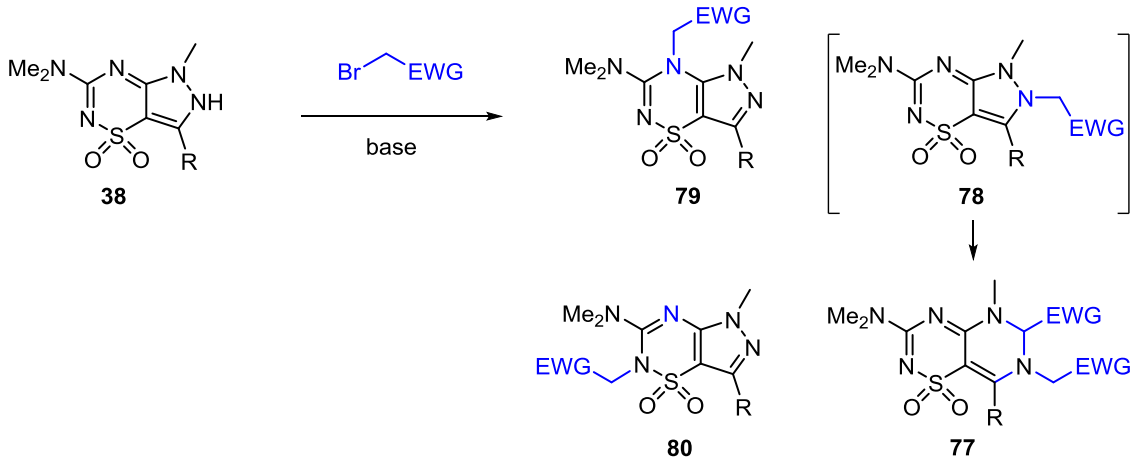


Scheme 78: The proposed mechanism of formation of pyrimido[4,5-*e*][1,2,4]thiadiazine products **77**.

The anion in intermediate **A** would be stabilised by conjugation through the thiadiazine ring and readily condenses at the imine carbon to form the pyrimidine ring in intermediate **B** which alkylates a second time. Intermediates (**A** or **B**) were never isolated, and hence it can be assumed that these two steps are fast.

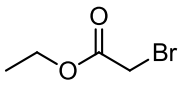
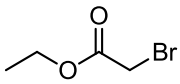
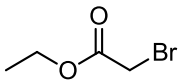
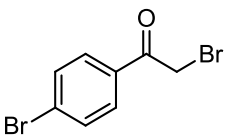
5.5.2 Substrate scope

A summary of the treatment of compounds **38** with electrophiles containing electron withdrawing groups is presented in **Table 12**, **Scheme 79**.



Scheme 79: Ring expansion reactions of pyrazolo-thiadiazines **38**.

Table 12: Reactions of pyrazolo-thiadiazines **38** with ethyl bromoacetate and 4-bromophenacyl bromide.

Electrophile	R	Method	Product(s)	Yield [%]
	H (**38a**)	A	**77a**	39
		B	**79a**$^{\#}$ + **77a**	11 + 39
	Me (**38b**)	A	**77b**$^{\#}$*****	61
		C	**79b** + **77b**$^{\#}$*****	5 + 53
	Ph (**38f**)	A	**79c**$^{\#}$***** + **81c***	62 + 3
		B	**79c**$^{\#}$*****	57
	Me (**38b**)	D	**79d***	24

[#]X-ray crystal structure obtained, *2D NOESY experiments conducted. Method A = K_2CO_3, Bu^n_4NBr, DMF, 50°C; Method B = K_2CO_3, Bu^n_4NBr, DMF, 60°C; Method C = K_2CO_3, Bu^n_4NBr, MeCN, 55°C; Method D = i. $NaHCO_3$ ii. K_2CO_3, DMF, rt.

Substrates **38** with minimal steric interference at the pyrazole ring gave rise to appreciable quantities of the ring expanded compound **77**. One example with a bulky phenyl substituent at C7 (**38f**), from which mostly N4 –alkylated product **79c** and little N2-alkylated product **81c** was isolated, demonstrated the steric influences of R groups towards the regioselectivity of N-alkylation. The reaction of compound **38a** with 4-bromo phenacyl bromide in DMF furnished only the N4 alkylated product **79d** in 24% yield. Unknown compounds were also produced from this reaction and no starting material was isolated. Heating **38a** with dimethylchloromalonate, methyl 2-bromopropionate and ethyl dichloroacetate in DMF largely returned the starting materials.

Structural assignments of compounds **79** and **77** were established by X-ray crystallographic studies on representative compounds **79c** and **77b** (**Figure 52**). Only one example of a product alkylated on a ring nitrogen atom other than N4, and which had not undergone a ring expansion, was isolated (compound **81c**). This led to the conclusion that this compound was alkylated on the N2 position since no N6-alkylated compounds were isolated in this study, and were thought to undergo ring expansion under the reaction conditions applied. The chemical shifts of the methylene protons were consistent with those obtained for the 4-chlorobenzyl alkylated derivatives of this system at N2 (**70c**, **70d**). Attempts to grow suitable crystals for X-ray analysis were unsuccessful.

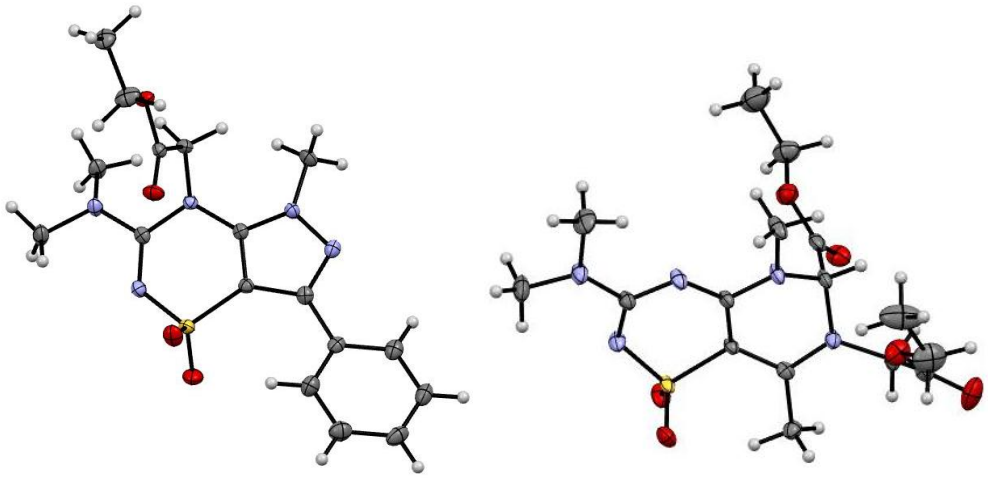


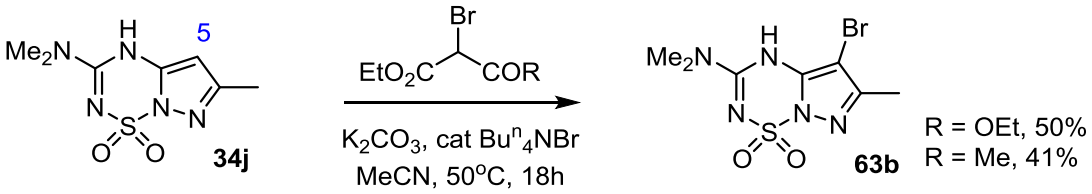
Fig 52. ORTEP diagrams of **79c** and **77b**.

The derivatives of the ring expanded pyrimido[3,4-*e*][1,2,4]thiadiazine ring system **77** described above exhibited similar poor stability to compounds **76** when exposed to acidic media such as residual acid in chloroform or silica gel; and hence these products were purified by chromatography over buffered silica and expeditiously characterised.

5.5.3 Susceptibility of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines to bromination

Section 5.2.1 demonstrates the slow reactivity of pyrazolothiatriazines **34** towards alkyl chlorides such as *p*-methoxy benzylchloride, diethyl chloromalonate and ethyl chloroacetoacetate. The application of the corresponding benzylic bromide (or iodide in the case of *p*-methoxy benzyl substitution) enabled conversion to the desired alkylated products shown in **Table 9**. Therefore, synthesis of the bromide or iodide derivatives of the β-keto compounds was proposed to work in a similar fashion and increase reactivity with the pyrazolothiatriazines **34**. A sample of methyl 2-iodoacetoacetate was prepared by Finkelstein reaction of the corresponding chlorides;[154] however the iodoester was very unstable and prone to elimination or polymerisation similar to the benzylic iodide synthesised in Section 5.2.1. Instead, samples of diethyl bromomalonate[158] and ethyl 2-bromo acetoacetate[159] were synthesised from the corresponding β-ketoester compounds. Treatment of **34j** with either diethyl bromomalonate or ethyl 2-bromoacetoacetate under

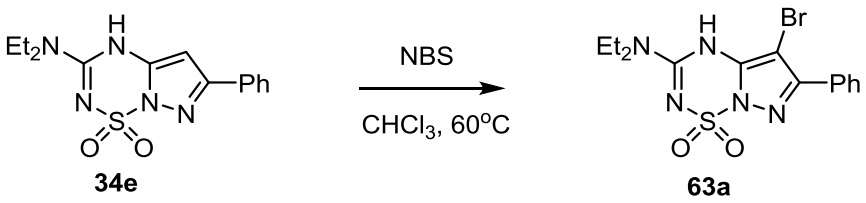
the conditions described in **Table 11** did not result in alkylation or ring expansion. Curiously, a bromine atom was introduced to the pyrazole carbon C5 to afford the unstable bromide **63b** (**Scheme 80**).



Scheme 80: Bromination at C5 of **34e** by α-bromoesters.

Other researchers[158–160] have reported bromination by diethyl bromomalonate; however, these results in **Scheme 80** are the first reported examples of diethyl bromomalonate and ethyl 2-bromoacetoacetate acting as brominating agents for a pyrazole ring**.** These results further highlight the considerable nucleophilicity of the C5 atom of pyrazolothiatriazines **34**.

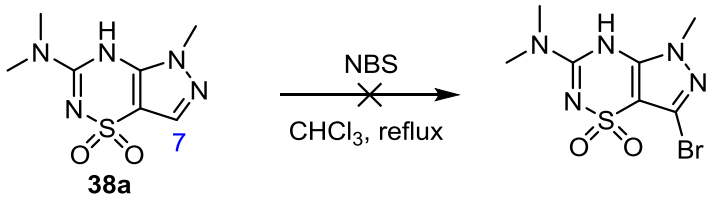
This is consistent with the nucleophilic nature of the pyrazole carbon in substrate **34e** whereby a 5-bromo substituent was introduced into in high yield by treatment with *N*-bromosuccinimide (NBS) in hot chloroform (**Scheme 81**) which was reported in Chapter 4.



Scheme 81: Bromination at C5 of **34e** with *N*-bromosuccinimide.

We considered the bromo-compounds **63** as potential substrates for further chemistry, such as Suzuki-couplings and related transformations; however, these compounds proved to be unstable in solution, to chromatography over silica gel, and were also sensitive to heat and light, making them unsuitable as synthetic intermediates.

A sample of pyrazolo-thiadiazine **38a** was treated with NBS in chloroform in order to evaluate the reactivity of C7 (**Scheme 82**).

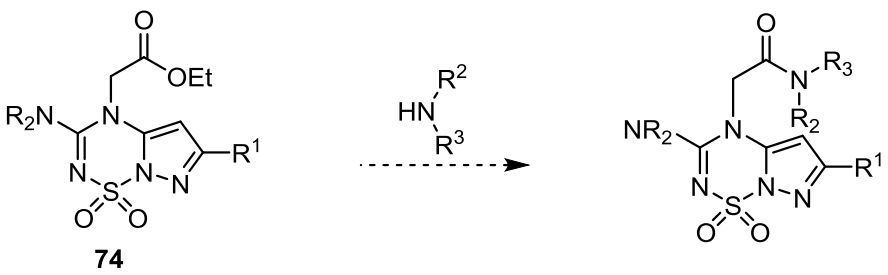


Scheme 82: The attempted bromination of **38a** by *N*-bromosuccinimide

After heating to reflux temperature overnight, mostly starting material was observed in the crude ^{1}H NMR spectrum. The starting material was subsequently collected in 59% yield after recrystallisation.

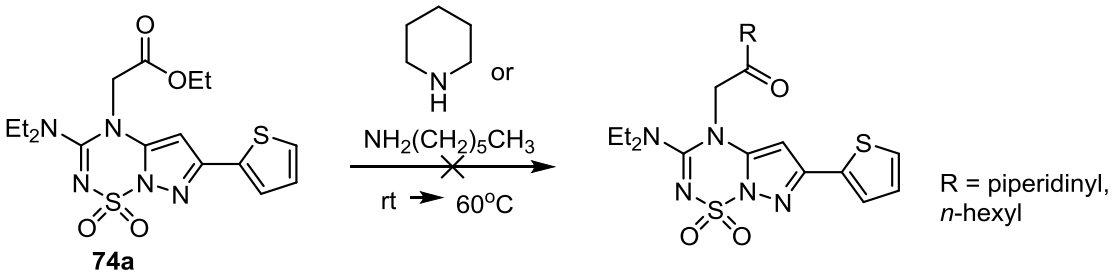
5.6 The application of ester functionality as a precursor to additional derivatives.

Compounds **74**, **76**, **77** and **79** containing ester functionalities were of interest as potential synthetic intermediates towards amide derivatives, which would add structural diversity to the library of derivatives of ring systems **34** and **38**. A proposed formation of amide derivatives of compounds **74** was envisioned from reactions of a series of small, primary or secondary amines with the electrophilic ester group (**Scheme 83**).



Scheme 83: The proposed formation of amides from esters **77**.

A series of experiments were carried out involving treatment of **74a** with two commercially available amines under a small set of conditions (**Scheme 84**, **Table 13**); however the starting material was returned in each case.

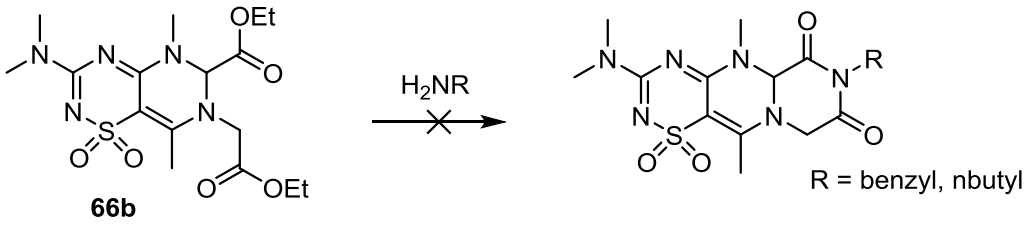


Scheme 84: The attempted aminolysis of the N4-ester substituent

Table 13: Reaction conditions attempted in order to form amide derivatives from ester containing precursor materials.

Amine	Conditions	Outcome
Piperidine	1.5 equivalents, MeCN, 50°C, 5 hours	No conversion
Piperidine	Neat, room temperature, 2 hours	No conversion
Piperidine	Neat, 60°C, 2 hours	No conversion
n-hexylamine	Neat, room temperature, 2 hours	No conversion
n-hexylamine	Neat, 60°C, 6 hours	Mostly starting material; signs of degradation

Compounds **65** and **66** containing bis-ester functionality were of interest as these could conceivably give rise to additional ring systems from condensation with primary amines. A sample of **66b** was treated with benzylamine, or n-butylamine under a series of previously reported conditions[161,162] (**Scheme 85**, **Table 14**).



Scheme 85: The attempted condensation of bis-ester groups with primary amines.

Table 14: Reaction conditions attempted in order to form additional fused pyrimidine rings from a bis-ester precursor.

Amine	Conditions	Outcome
Benzylamine	2 equivalents, xylene, reflux, 23 hours	Complex mixture
Benzylamine	2 equivalents, triethylamine, 2-methoxyethanol, 80°C, 24 hours	Complex mixture
n-Butylamine	Neat, reflux temperature, 3 hours	Complex mixture

In all cases, attempts to form a cyclised product from the condensation of an amine with the bis-ester dielectrophile **66b** resulted in a complex mixture of products. The colour of the reaction solution altered to a vibrant yellow or orange colour, and the crude product could not be adequately purified via silica gel chromatography or recrystallisation. ^{1}H NMR analysis of the crude product indicated that the core heterocyclic structure was altered when exposed to the amine nucleophile under the heating conditions. Previously, these fused pyrimidines **65** and **66** were observed to be unstable when exposed to acid, so these compounds were deemed unsuitable as intermediates for subsequent transformations via the ester substituents because it was apparent that the conditions required to initiate reactions were causing the degradation of the compounds.

A sample of **66b** was treated with hydrazine hydrate in attempts to generate a 7-membered ring following relevant literature reports.[163–166] Similar problems to those encountered with primary amines arose whereby room temperature trials in acetonitrile

did not result in any changes, and heating or concentrating the solution caused decomposition of the starting material.

5.7 General notes on the reactivity of pyrazolo[1,5-b][1,2,4,6]thiatriazines and pyrazolo[3,4-e][1,2,4]thiadiazines as nucleophiles

The pyrazolothiatriazines **34** and pyrazolo–thiadiazines **38** possessed low solubility in many common organic solvents such as dichloromethane, acetone, diethyl ether, ethyl acetate and tetrahydrofuran. This meant that comparative experiments between these solvents and the solvents with greater polarity (DMF, DMSO and acetonitrile) were not practicable. The use of a phase transfer catalyst (Bu^n_4NBr) was often required to allow action of the base (often potassium carbonate) upon the substrate. In a few instances, comparison between the less polar tetrahydrofuran, dichloromethane, chloroform and the application of the more polar acetonitrile as a solvent for reaction was explored (**Table 8**, **Table 9**). Generally, the specific alkylating agent used had a great influence on selectivity as shown in methylation and benzylation experiments. In several examples, specifically from methylation and ethyl bromoacetate alkylation, substrate substitution influences such as steric hindrance of NH groups were obvious.

It is interesting to note that with benzylic alkylation, where the same conditions were employed, the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34** exhibited dominant reactivity at the NH group on the pyrazole ring (N7) rather than the thiatriazine ring, whereas the pyrazolo[3,4-*e*][1,2,4]thiadiazine dioxides **38** were benzylated mostly at N2 on the thiadiazine ring. This demonstrated that the two classes of compounds have different regiochemical reactivity patterns and could be used to produce diverse molecular architectures in synthesis of small compound libraries for biological screening.

5.8 Conclusion

The recently discovered pyrazolo[1,5-*b*][1,2,4,6]thiatriazine template **34** has been shown to possess four nucleophilic sites (N2, N4, C5, N7) which underwent a range of substitution reactions. Methylation occurred at both N4 and N7; the ratio was dependent on choice of solvent. Benzylation occurred preferentially at N7, regardless of solvent, but also occurred at C5, and to a lesser extent at N4 and N2. Alkylation with α-halo esters occurred at both N4 and N7, but the latter derivatives, under the reaction conditions, underwent a ring expansion reaction to afford the first reported pyrimido[1,6-*b*][1,2,4,6]thiatriazine derivatives **76**. This represents the first published example of a pyrazole ring expansion mediated by an alkylating agent. Bromination of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34** afforded unstable 5-bromo derivatives **63** which were not synthetically useful.

Compounds **38** were shown to possess three nucleophilic sites (N2, N4, and N6) which underwent substitution reactions. Methylation occurred at all three sites. Alkylation with benzylic halides occurred preferentially at N2, but some also occurred at N6, and to a lesser extent at N4. Similar alkylation with ethyl bromoacetate occurred at both N4 and N6, but the latter derivatives underwent a pyrazole ring expansion to afford pyrimido[4,5-*e*][1,2,4]thiadiazine derivatives. There has been no evidence of a carbon nucleophile on the pyrazole ring of compounds **38**.

CHAPTER 6: REACTIONS TO FORM PYRAZOLO[1,5-*A*][1,3,5]TRIAZINE COMPOUNDS FROM PYRAZOLO-[1,5-*B*][1,2,4,6]THIATRIAZINES

6.1 Introductory remarks

The selective synthesis of novel fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34** as described in Chapter 2 provided a range of substrates of **34** for NH substitution reactions. Unsuccessful attempts to react the NH moiety with common acid chlorides or anhydrides led to the synthesis of alkylated derivatives as described in Chapter 5. N7-Benzyl derivatives of compounds **34** (compounds **65** and **66**) were found to be sensitive to acidic conditions or heat. This contrasted with the relatively robust parent compounds **34**; which under the same conditions were unaffected. Ring expanded materials containing fused pyrimidine rings (**76** and **77**) were also prone to decomposition and required careful handling. The causes for decomposition are explored within this chapter, to establish which structural factors influence the degradation of these fused thiatriazine compounds, and under which conditions these materials are prone to such changes. It was of interest to determine the mechanism of decomposition and identify the degradation material(s) which would aid in the understanding of these unusual compounds. Such understanding might also be of some synthetic value.

6.2 The extrusion of SO_2 from N7-benzylated pyrazolothiatriazines

In the course of characterising the various pyrazolo[1,5-*b*][1,2,4,6]thiatriazine products described above, compounds bearing benzyl substituents at the pyrazole nitrogen N7 specifically (i.e. **64** and **65**) decomposed via an unknown mechanism. Samples of **65** were especially prone to degradation in chlorinated solvents and silica gel, making

characterisation difficult. This was thought to be a result of residual acidity to which these compounds may have been sensitive. Analysis of NMR sample solutions of compounds **64** and **65** in deuterated chloroform showed additional peaks forming between δ 8-10 ppm after standing for long periods of time, with material precipitating from solution. A sample of **65e** was left to stand for several days in deuterated chloroform until a precipitate formed; then the solvent was evaporated and a ^{1}H NMR sample analysed in DMSO-d_6 which revealed that a complete transformation had occurred (**Figure 53**).

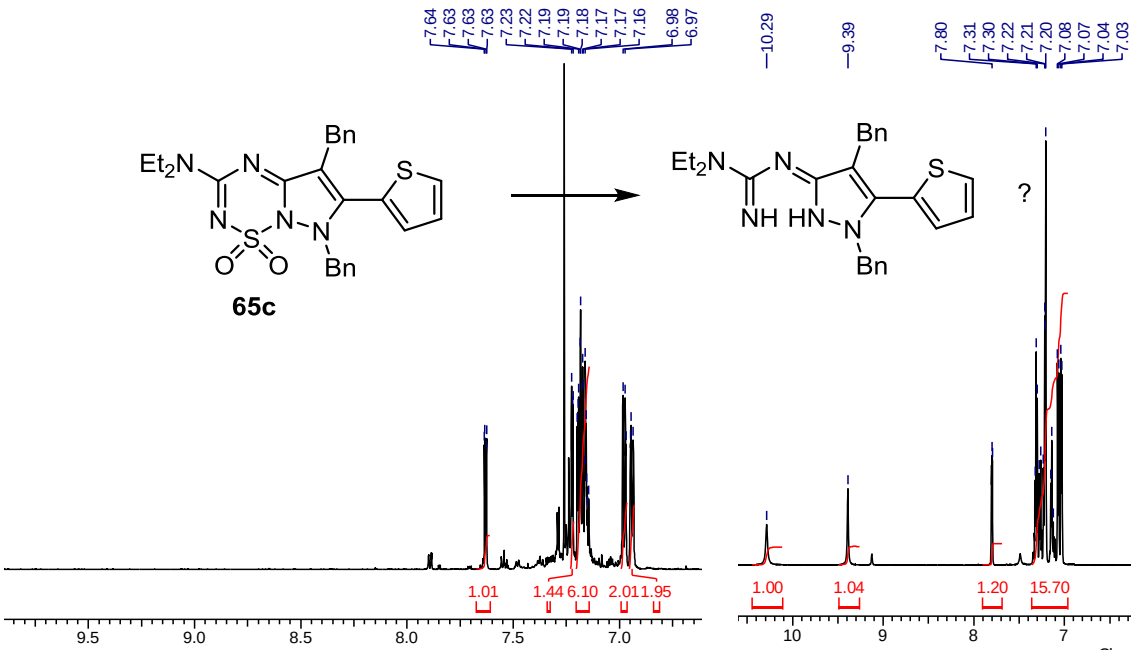
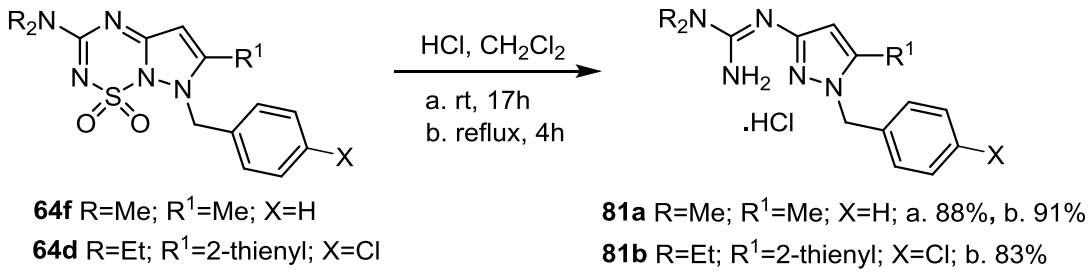


Fig 53. ^{1}H NMR (600MHz) spectra of **65e** (left, $CDCl_3$) and the precipitate formed after standing **65e** in $CDCl_3$ (right, DMSO-d_6).

The spectral data from this product revealed a potential guanidine structure (which was not fully characterised due to limitations of scale) which could conceivably result from the extrusion of sulfur dioxide from the thiatriazine ring. This appeared to be caused by the residual HCl present in the chloroform or perhaps from the silica gel. A dichloromethane solution of representative compound **64f** was treated with aqueous hydrochloric acid which afforded the guanidine hydrochloride **81a** in high yield (**Scheme 86**). This result supported the proposed acid-mediated expulsion of SO_2 from the thiatriazine ring.



64f R=Me; R^1=Me; X=H
64d R=Et; R^1=2-thienyl; X=Cl

81a R=Me; R^1=Me; X=H; a. 88%, b. 91%
81b R=Et; R^1=2-thienyl; X=Cl; b. 83%

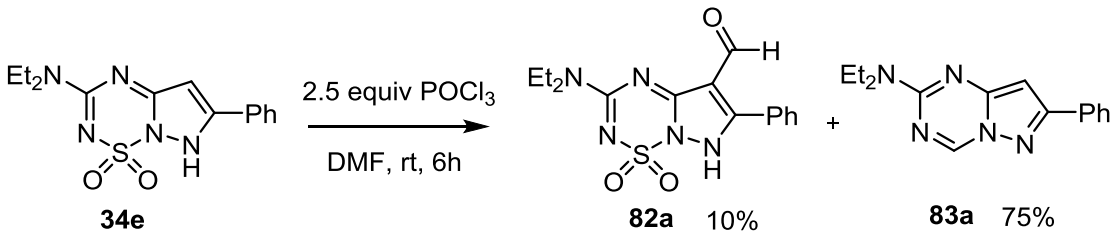
Scheme 86: The formation of pyrazolo guanidine hydrochlorides **81** from treatment of N7-benzyl pyrazoles **64f** & **64d** with hydrochloric acid.

Similar treatment of **64d** afforded **81b** in 83% yield (**Scheme 86**). A comparison between the precursor materials **34** and the N7-benzyl derivatives **64** to check relative stability under acidic conditions was carried out by exposure of the parent compound to the conditions described in **Scheme 86**. A sample of **34d** in dichloromethane was treated with 3 drops of 10% aqueous hydrochloric acid and heated at reflux for 24 hours. The starting material was recovered in 94% yield, indicating much greater stability under the acidic conditions.

The propensity of these pyrazolothiatriazines to extrude sulfur dioxide may explain the observed instability of N7 alkylated compounds. Compounds **64, 65** and **74** (Chapter 5) were particularly sensitive to acidic conditions when compared to the precursor compounds **34**. Similar behaviour was observed with the pyrimido-thiatriazines **76**, although attempts to isolate and characterise these decomposition products were not successful. Filtration of chlorinated solvents such as chloroform and dichloromethane through alumina, followed by storage over potassium carbonate prior to addition to samples of **64, 65, 75** and **76** slowed the degradation of these compounds significantly. The parent compounds **34** and the N4-benzylated compounds **71** were stable to the conditions which caused the extrusion of the sulfur dioxide moiety from the N7-benzyl derivatives **64** and **65**. However, if the ejection of sulfur dioxide could be achieved with the parent compounds **34,** then this may enable the establishment of a synthon for efficient production of other novel compounds.

6.3 The action of phosphoryl chloride and DMF to form fused pyrazolo[1,5-*a*][1,3,5]triazines

When a sample of fused pyrazolothiatriazine dioxide **34e** was treated with standard Vilsmeier-Haack conditions,[167] an unexpected side product formed. Substrate **34e** was stirred in DMF in the presence of phosphoryl(V) chloride ($POCl_3$) to furnish two compounds, a *C*-formylated derivative **82a** and the fused pyrazolo-triazine **83a** (**Scheme 87**).



Scheme 87: The formation of triazine **83a** from Vilsmeier-Haack conditions.

The expected products from this synthesis would contain either an *N*-formyl group or a *C*-formyl group by reaction of a nucleophilic site with the electrophilic Vilsmeier reagent. The reaction of the Vilsmeier reagent at the pyrazole carbon formed the aldehyde **82a** in the conventional manner.

The structure of compound **83a** was confirmed by X-ray crystallography (**Figure 54**) which revealed the highly conjugated and planar bicyclic system.

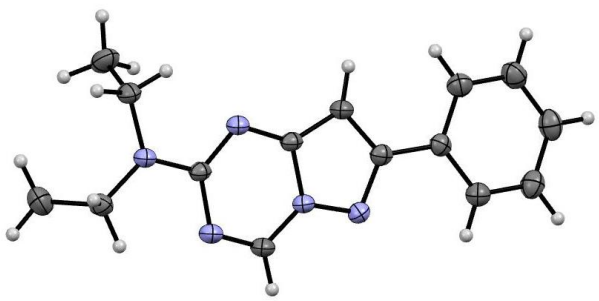
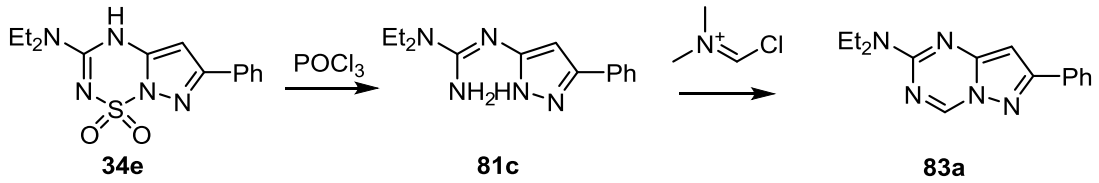


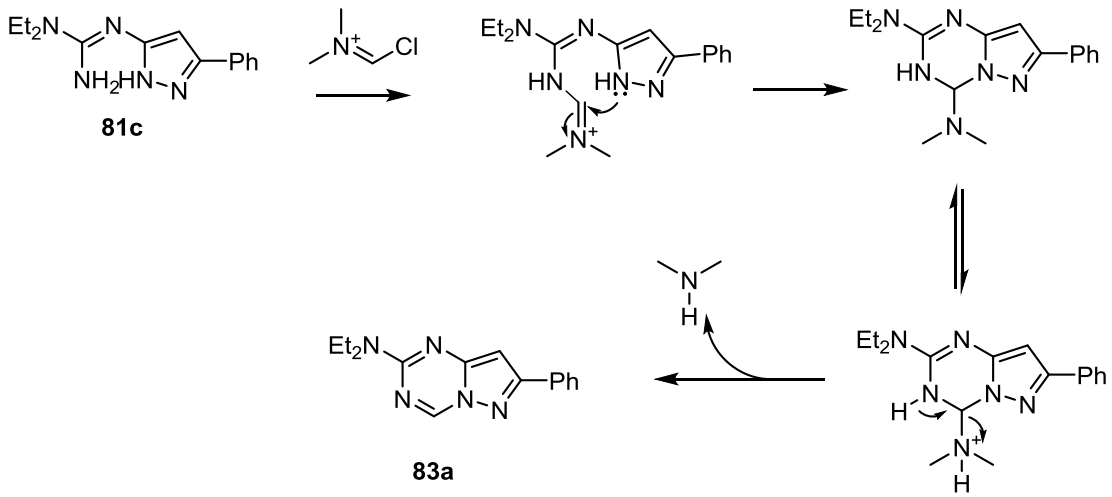
Fig 54. ORTEP diagram of **83a**.

The formation of the triazine ring from the thiatriazine parent compound must arise from the initial cleavage of an N-S sulfamide bond. This is proposed to follow a similar mechanism to the formation of guanidines **81a** and **81b**, where the reaction conditions produced by the combination of $POCl_3$ and DMF led to extrusion of the sulfur dioxide motif (**Scheme 88**) to form a guanidine **81c**. Then in the presence of the Vilsmeier reagent, compound **81c** condenses to form **83a** in situ before it can be isolated.



Scheme 88: The in situ generation of triazine **82a** from initial extrusion of the SO_2 moiety.

The condensation to form the triazine ring from the guanidine intermediate **81c** is likely favoured by the aromatisation to form the triazine ring, readily emitting dimethyl amine in the process (**Scheme 89**).



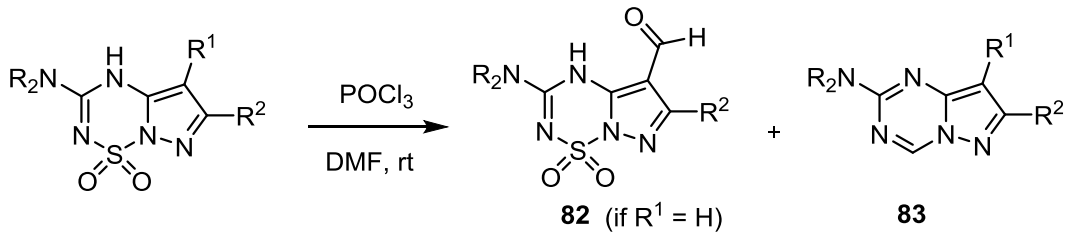
Scheme 89: The proposed mechanism for the formation of triazine **83a** from the pyrazolo-guanidine intermediate.

The initial elimination of the SO_2 moiety under the reactions was unexpected considering that these 'parent' pyrazolothiatriazines **34** were not prone to degradation under the mild acidic conditions described in Section 6.2, **Scheme 86**. The acidity of phosphoryl chloride is less than that of aqueous HCl, which meant that the mechanism of this sulfamide bond cleavage was not simply driven by acidity alone. At this stage, there is insufficient evidence to propose a mechanism of formation; however, this will be further discussed in Section 6.5.

An experiment was performed to investigate the extrusion of the sulfur dioxide group under these Vilsmeier-Haack conditions. A solution of the substrate **34e** in acetonitrile was treated with 3.3 equivalents of $POCl_3$ and stirred at room temperature for 15 hours; after which time the starting material was recovered. This led to the conclusion that the presence of $POCl_3$ alone was not sufficient to cause degradation of the starting material. The $POCl_3$ reagent is known to release by-products upon reaction with DMF (Cl^- and $POCl_2^-$)[168] which could have initiated cleavage of the thiatriazine ring at the electrophilic sulfur atom, leading to the eventual loss of the SO_2 moiety.

6.3.1 Substrate scope of the Vilsmeier-Haack reagent mediated triazine formation

Three representative compounds **34** were each exposed to Vilsmeier conditions ($DMF/POCl_3$) to determine the optimal quantities of the phosphoryl reagent, and the influence of steric and electronic properties of the substrate (**Scheme 90**, **Table 15**).



Scheme 90: The formation of pyrazolo-triazines **82** and 5-formyl derivatives **81**.

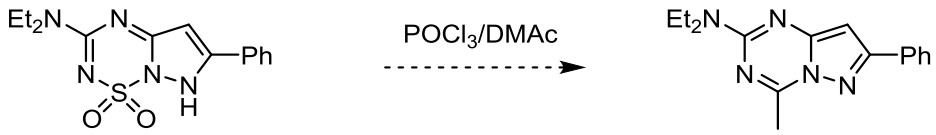
Table 15: Formation of compounds **82** and **83**.

R_2N	R^1	R^2	Equiv. $POCl_3$	Product (s)	Yield(s) [%]
Et	H	Ph (**34e**)	2.1	**82a + 83a**	Trace + 50
			3.1	**82a + 83a**	10 + 75
			3.5	**82a + 83a**	9 + 88
Me	CO_2Et	H (**34i**)	2.0	**83b**	18*
			3.2	**83b**	20*
Me	H	Me (**34j**)	3.3	**83c**	57

* starting material recovered

The carbethoxy functionalised substrate **34i** lacked the electron rich pyrazole ring and the *C*-nucleophile moiety that the other fused pyrazolothiatriazines possessed. Under the best conditions, the analogous pyrazolo-triazine **83b** was afforded in 20% yield after 40 hours as well as recovery of the starting material. It was noted that the reactions did not proceed smoothly when the mixture was heated, and this was likely due to the known instability of the Vilsmeier reagent at higher temperatures.[168]

The scope of this methodology was also investigated with respect to the group which could be inserted in place of the SO_2 substituent. Modified Vilsmeier conditions have historically applied the use of other amide derivatives than DMF.[169,170] For example, a methyl substituent might be placed at the C6 position of the triazine ring using N,N-dimethyl acetamide (DMAc) (**Scheme 91**).



Scheme 91: The proposed application of DMAc as a substitute for DMF in a Vilsmeier-Haack reaction.

An attempt to react substrate **34e** with a mixture of DMAc and $POCl_3$ returned the starting material after stirring at room temperature overnight. On this basis, it appeared that the scope of Vilsmeier-type chloroiminium reagents for the formation of triazines **82** from thiatriazine rings was not adequately diverse to merit further attempts at synthesis of pyrazolo-triazine derivatives from this method.

6.4 Pyrazolo[1,5-*a*][1,3,5]triazines as biologically relevant compounds

These new compounds **83** represent examples of the pyrazolo[1,5-*a*][1,3,5]triazine ring system and were obtained as stable crystalline solids. Triazines **83** are structurally similar to purine bases and have often been documented as isosteres of purine.[171,172]

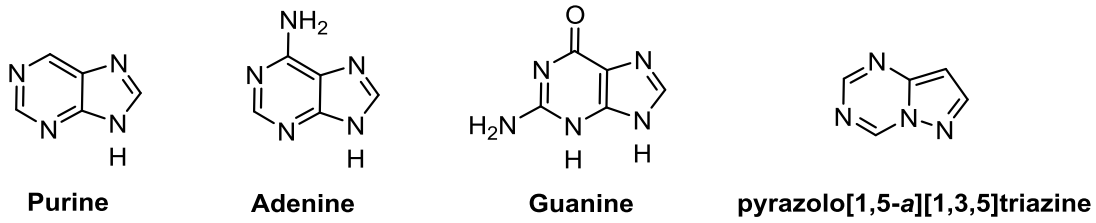


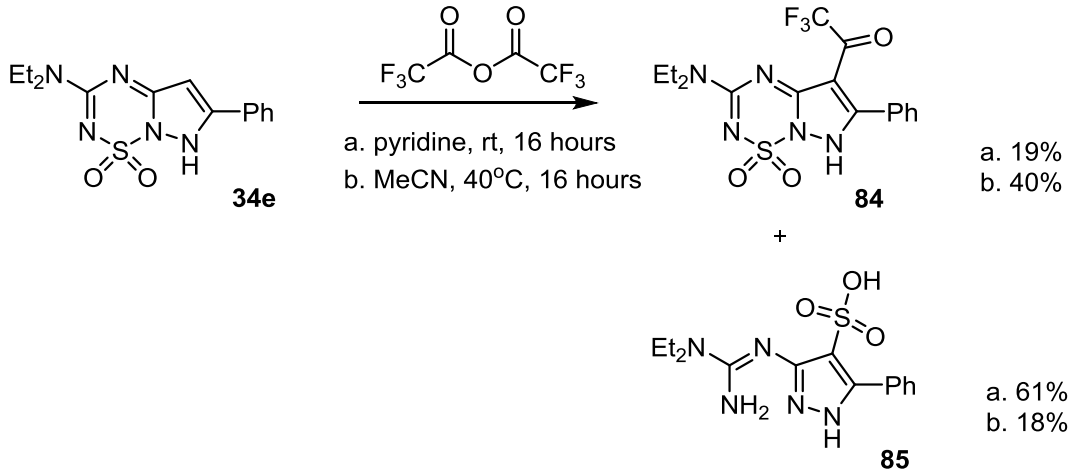
Fig 55. The structure of the purine heterocylic core including representative purine DNA bases adenine and guanine, and a pyrazolo-triazine **83** core.

Purine bases are common structural motifs for DNA and RNA as well as a number of biological coenzymes such as nicotinic adenosine diphosphate (NAD^+), adenosine triphosphate (ATP), guanosine triphosphate (GTP) and flavin adenosine diphosphate

(FAD).[173] The structural likeness of purine to the pyrazolo[1,5-*a*][1,3,5]triazine ring system has warranted the development of compounds **83** as biologically active agents to target the purinergic signalling receptors and enzymes involved in the metabolism of purine derivatives. Pyrazolo[1,5-*a*][1,3,5]triazines **83** have therefore remained of biological interest for common purine enzyme and receptor targets such as antagonists of protein kinase CK2,[174] phosphodiesterase type 4,[172] corticotropin releasing factor 1 (CRF1),[175] A1 adenosine receptors[176] and for antiproliferative activity by means of tubulin inhibition.[177]

6.5 The action of trifluoroacetic anhydride on the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine system.

The lack of reactivity of the NH group of compounds **34** towards activated carbonyl reagents[99] led to the use of trifluoroacetic anhydride (TFAA) as a stronger electrophile. The trifluoroacetylated product **84** was furnished from the reaction of TFAA with the precursor compound **34e** in acetonitrile or in neat pyridine (**Scheme 92**) and no NH acylated product(s) were isolated, again indicating the nucleophilicity of C5 in these systems. That no N acylated products were isolated may be explained by their potential instability, as it is known that *N*-trifluoroacetylated pyrazoles are labile and are readily cleaved under mildly basic conditions.[178] It is thus possible that any trifluoroacetamide which formed was hydrolysed on workup or cleaved in the reaction mixture. Unexpectidly the major product afforded from the reaction in pyridine was the guanidine sulfonic acid **85** which was structurally elucidated by X-ray crystallography (**Figure 56**).



Scheme 92: The action of TFAA on fused pyrazolothiatriazine **34e**.

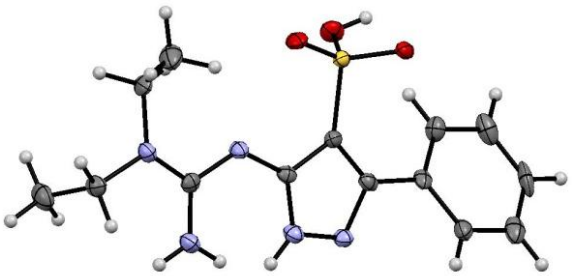


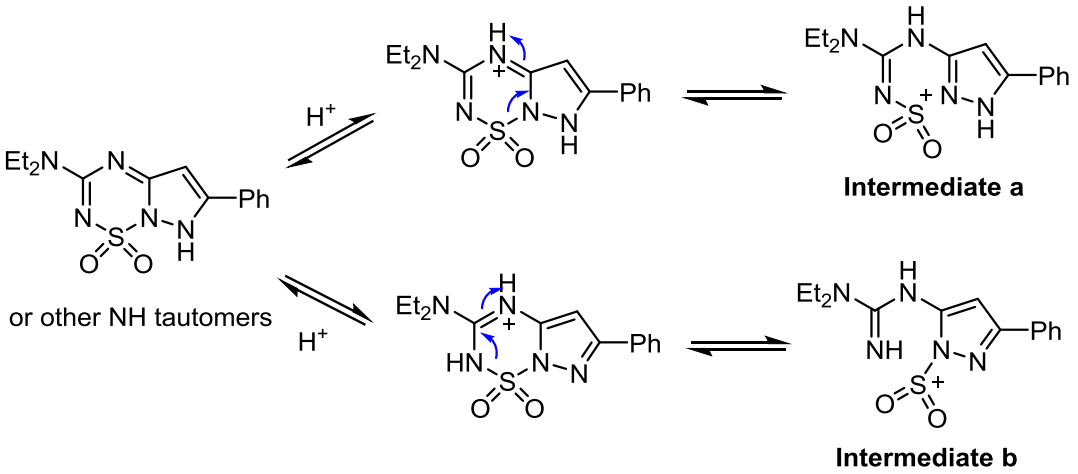
Fig 56. ORTEP diagram from X-ray crystal structure of **85**. A molecule of H_2O has been omitted for clarity.

It appears that there is a competing reaction between trifluoroacetylation and this sulfonation reaction, and it is suggested that conditions a), which utilise pyridine, provide more of the sulfonic acid **85** simply because the formation of compound **84** is slower under these conditions.

The migration of the sulfonic acid to the pyrazole carbon and the previous loss of sulfur from the ring (Section 6.3) suggested that simple acidity is not the sole factor in causing the extrusion of the SO_2 group from the thiatriazine ring, and that perhaps other factors are involved.

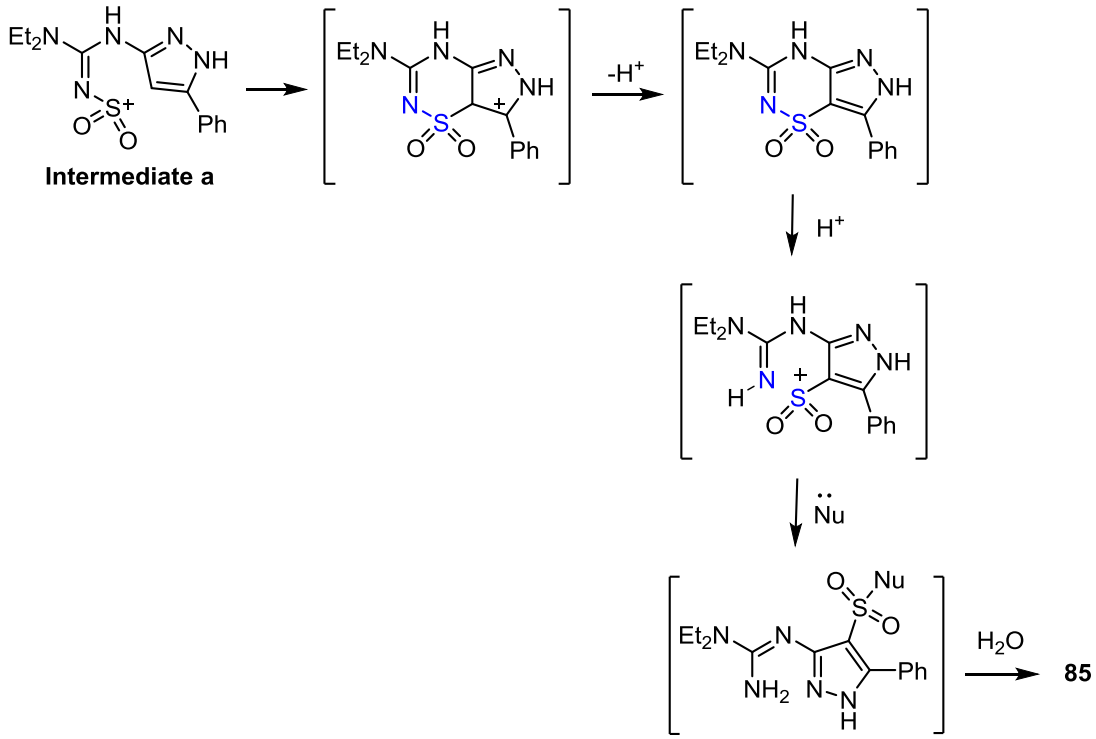
6.5.1 Mechanism of the formation of pyrazolo-guanidines substituted with sulfonic acid

While not the only cause, it is apparent that acid, such as that generated from residual TFA in the mixture, may mediate the cleavage of a sulfamide S-N bond. Considering the mechanism, it may be possible to cleave either of the two sulfur to nitrogen bonds giving intermediates *a* and *b* as shown in **Scheme 93**. In an alternative mechanism, it is possible that another species may initiate this ring cleavage presumably by nucleophilic attack at the sulfur atom. Under these conditions however, the nucleophiles present such as trifluoroacetate or chloride anions have very weak reactivity as nucleophiles and are unlikely to react in this way. This alternative mechanism will be futher discussed in Section 7.4.2 for a series of similar reactions.



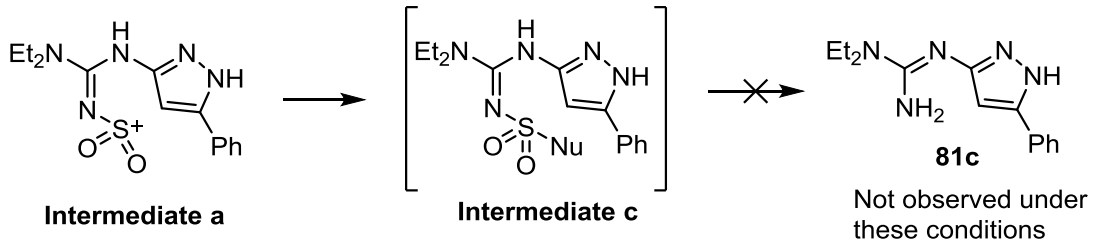
Scheme 93: A proposed mechanism for the cleavage of the sulfamide S-N bond.

A possible mechanism for the formation of compound **85** is indicated in **Scheme 94** whereby the sulfur atom in intermediate *a* reacts with the nucleophilic pyrazole carbon. This would form a thiadiazine intermediate which may not be stable, and could be prone to nucleophilic attack from a weak nucleophile such as TFA, or water in an aqueous separation. The sulfonic acid **85** could result from aqueous hydrolysis of the S-N bond.



Scheme 94: The proposed pathway to compound **85** via intermediate *a* with a nucleophile such as TFA or water.

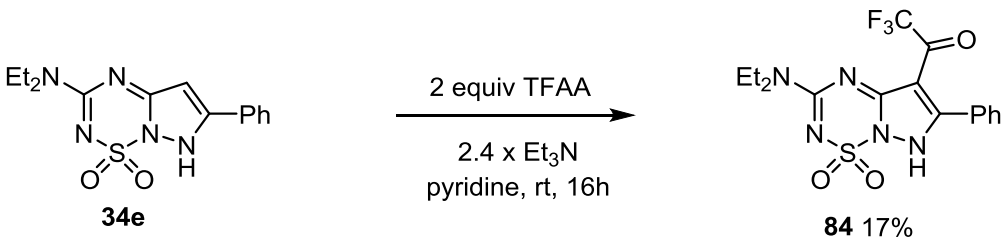
Reaction of intermediate *a* with an external nucleophile would give intermediate *c*, which would be expected to hydrolyse on product isolation to give compound **81c** as shown in **Scheme 95**. As this product is not observed under these reaction conditions this pathway does not occur. It is thus apparent that intermediate a must react quickly with the pyrazole C5 to give compound **85**.



Scheme 95: Reaction of intermediate *a* with a nucleophile.

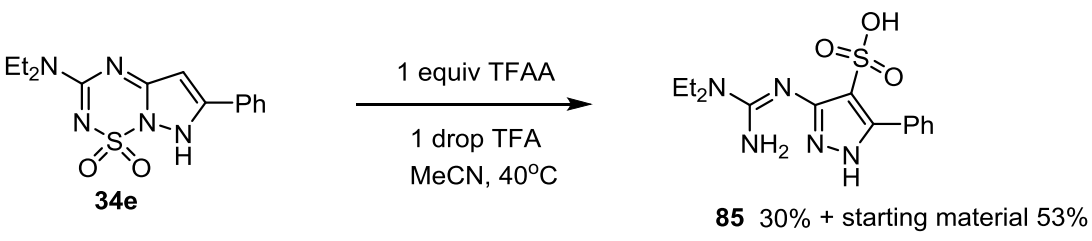
The absence of any products which have the CF_3 group attached at C5 and loss of the SO_2 group seems to indicate that attachment of this goup stabilises the ring system possibly by reducing the tendacy to form intermediate *a* or intermediate *b*.

To establish the role of the residual TFA in the reaction to form the sulfonic compound **85**, two sets of conditions were employed which involved the presence of excess of base for comparison. The conditions from **Scheme 92** (b) with the addition of an excess of potassium carbonate gave no appreciable quantity of product(s) after 30 hours and 86% recovery of the starting material. The conditions from **Scheme 92** (a) with the addition of excess of triethylamine generated **84** in 17% yield along with a 76% recovery of the starting material (**Scheme 96**).



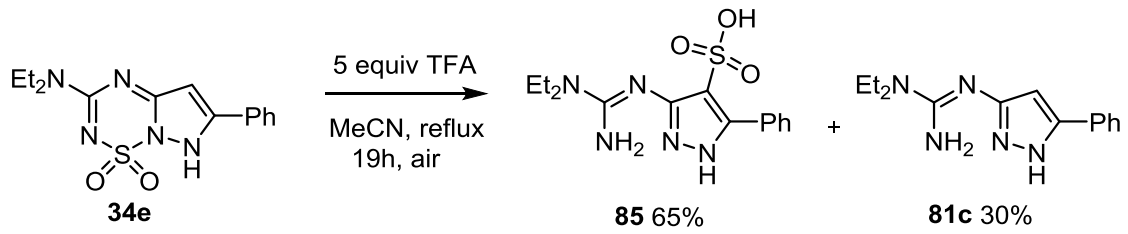
Scheme 96: Trifluoroacetylation of **34e** from the action of TFAA and excess of base.

It was hypothesised that a combination of TFA and the TFAA should furnish the sulfonic acid **85** in the absence of base. Compound **34e** was treated with TFAA in the presence of a single drop of TFA to furnish compound **85** in 30% yield along with 53% recovery of the starting material (**Scheme 97**).



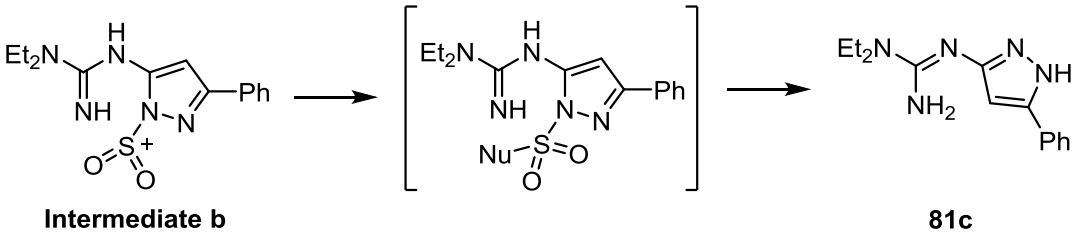
Scheme 97: Treatment of **34e** with TFAA/TFA to establish the role of the acid in the extrusion of sulfur dioxide.

The formation of sulfonic acid **85** was explored in more detail by adding an excess of acid in an attempt to force the formation of the one product. A mixture of TFA and acetonitrile was heated to reflux temperature in air and after 19 hours, a mixture of the sulfonic acid **85** and also a guanidine **81c** (notably without the sulfonic substituent on the pyrazole) was obtained under these conditions (**Scheme 98**).



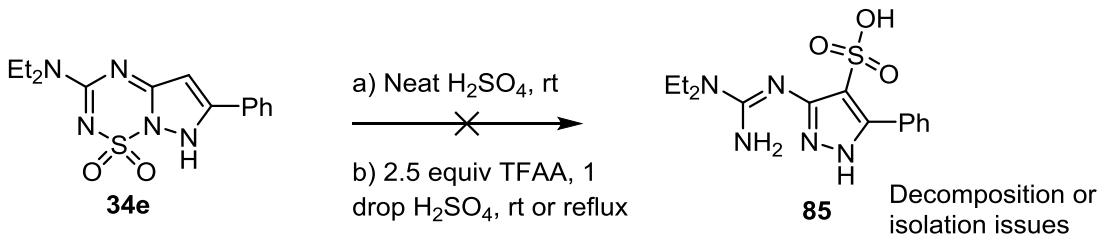
Scheme 98: The formation of guanidines **85** and **81c** from the action of TFA on compound **34e**.

One possible explanation for this formation of product **81c** is that these conditions led to preferential formation of intermediate *b*. Previously, it appeared that intermediate *a* was the precursor to product **85** (**Scheme 94**) and was unikely to give product **81c** (**Scheme 95**). Intermediate *b* is thus proposed as the likely precursor to compound **81c** by nucleophilic attack and hydrolysis (**Scheme 99**).



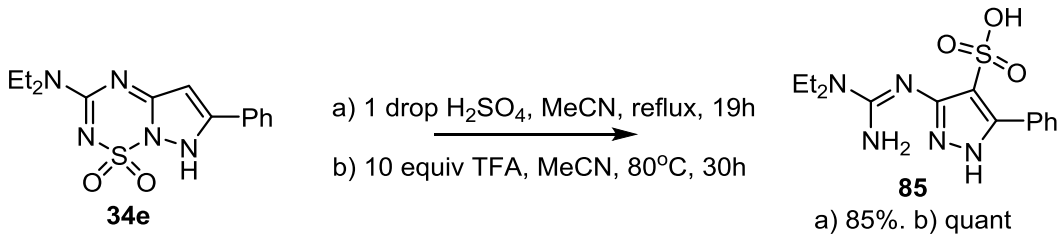
Scheme 99: The proposed formation of compound **81c** from intermediate *b*.

An experiment was performed at room temperature with concentrated sulfuric acid in neat conditions; however, after 12 hours only decomposition was detected (**Scheme 100**). Treatment of the same substrate **34e** with 2.5 equivalents of TFAA and a single drop of concentrated sulfuric acid in acetonitrile at room temperature returned the starting material after stirring overnight. When the conditions were repeated at reflux temperature, very little material could be extracted from the aqueous layer.



Scheme 100: Treatment of compound **34e** with sulfuric acid (neat) or H_2SO_4/TFAA.

Compound **34e** was then treated with a single drop of concentrated sulfuric acid in acetonitrile and heated to reflux temperature in air for 19 hours, after which an 85% yield of the sulfonic acid **85** was collected (**Scheme 101**). The reaction of **34e** with dry TFA in acetonitrile at 80°C gave the sulfonic acid material **85** in quantitative yield.

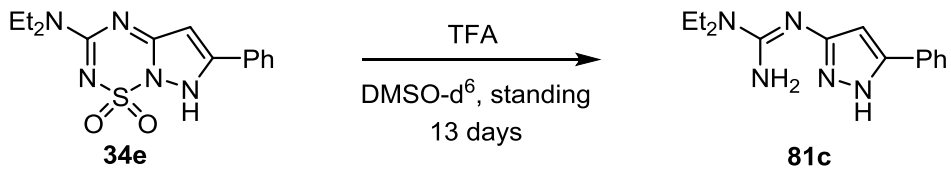


Scheme 101: Sulfonation of pyrazolothiatriazine **34e**.

It should be noted here that sulfuric acid is a known sulfonating agent for pyrazoles and, in some cases, a dessicant such as an anhydride for example is added to favour the formation of the sulfonic acid product.[116] The role of the sulfuric acid here however, is most probably to the protonation of the precursor compound **34e**. The sulfonation did not work effectively at higher concentrations of acid because the compound in question was most seemingly prone to degradation by other pathways.

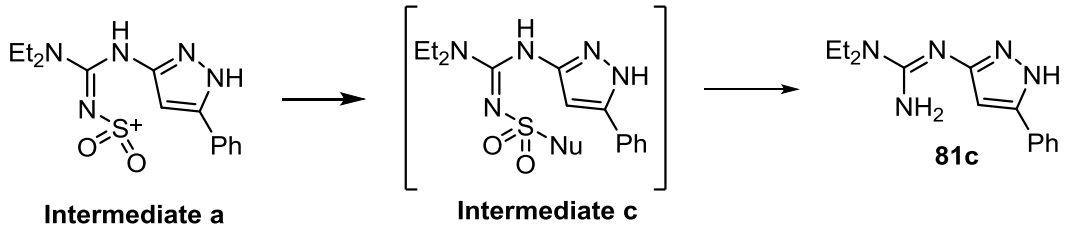
The action of TFA on compound **34e** was investigated in air with an excess of TFA within an NMR tube in DMSO-d_6 (which is a notoriously hydroscopic solvent). This generated the ring opened product **81c** upon standing at room temperature over a period of 13 days (**Scheme 102**). At this point it was speculated that a large excess of acid under dry conditions and in the absence of a moderate nucleophile seemed to favour the

formation of the sulfonic acid **85**, but in the presence of air or moisture and with less acid, the product **81c** seemed to form.



Scheme 102: Transformation of **34e** to guanidine **81c** with TFA at room temperature.

This result could mean that, in addition to the source of acid, the presence of nucleophiles in the reaction mixure (such as water for example) could intercept with the formation of the sulfonic acid **85** from intermediate *a*. In such cases, the reaction of an external nucleophile competes with the reaction of the pyrazole carbon (**Scheme 103**). This means that, although it has been suggested that in the absence of external nucleophiles the formation of compound **85** does not proceed from intermediate *b* and comes from intermediate *a*, compound **81** could form via either intermediate *a* or *b*.



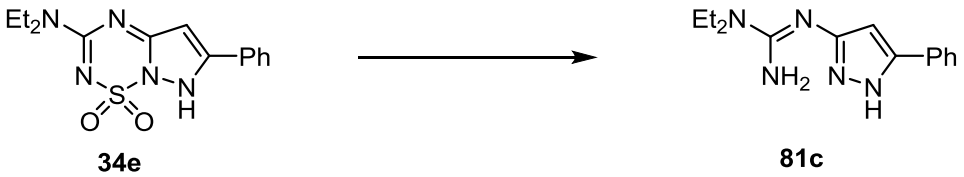
Scheme 103: The proposed formation of compound **81c** from intermediate *a* with the action of a nucleophile such as water.

The addition of excess of base to a mixture of **34e** and TFA (such as triethylamine or potassium carbonate) prevented formation of either product **81** or **85**. An experiment in acetonitrile at 50°C in the presence of potassium carbonate returned mostly the starting material. The starting materials were also isolated from mixtures of acetonitrile or DMF and potassium carbonate in the presence of TFA and/or TFAA, further demonstrating that the ejection of the sulfur dioxide moiety could be avoided by the addition of base. This also supported the hypothesis that expulsion of sulfur dioxide is acid mediated. Notably, the conditions required to form a guanidine compound **81** from compounds **34**

are harsher than those required to extrude the SO_2 moiety from the benzylated pyrazolothiatriazines **64** and **65**. The parent ring structures **34** seemed more resistant to the acidic decomposition of the thiatriazine ring as discussed in Chapter 5.

6.6 The formation of pyrazolo guanidines as precursors to triazine rings

The preparation of compound **81c** was explored in more detail to obtain an adequate quantity of the material as a possible synthetic precursor (**Table 16**). The table represents the most successful conditions from this research.



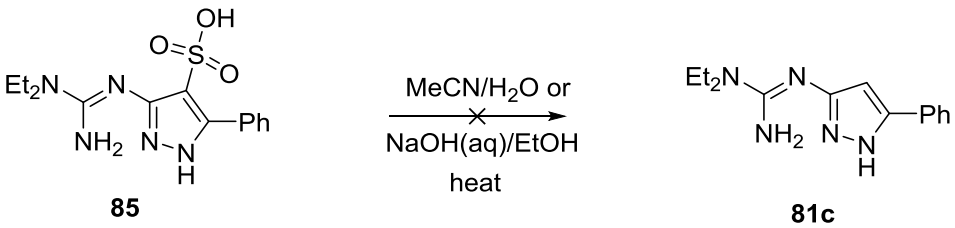
Scheme 104 (above) and **Table 16** (below): The acid facilitated extrusion of sulfur dioxide from pyrazolothiatriazine **34e** to form pyrazolo-guanidine **81c**.

Conditions	Yield [%]
5 equivalents TFA, MeCN, 75°C, 43h	78
Excess of concentrated HCl (aq), 3:1 MeCN:H_2O, reflux, 17h	79
Concentrated HCl (aq), reflux, 2.5h	100% crude
1.8 equivalents *p*-TsOH, MeCN, 60°C, 20h	75
10 equivalents TFA, 1:1 MeCN:H_2O, 55°C, 72h	86

Notably, little to no reactivity was observed when *p*-toluene sulfonic acid (*p*TsOH) or TFA were employed at room temperature. To avoid anticipated isolation issues with other substrates from aqueous mixtures, the reaction was attempted in chloroform which had been treated with hydrogen chloride gas; however no conversion was detected.

Concentrated aqueous hydrochloric acid functioned effectively in a mixture of acetonitrile and water over a shorter time frame to give 79% of the guanidine compound **81c**. A basic workup was performed to remove the hydrochloride salt and extract the polar compound from the aqueous layer. The reaction to form compound **81c** from neat concentrated aqueous hydrochloric acid presented problems in separating the product from the aqueous acid mixture. Treatment of substrate **34e** in neat TFA at room temperature also led to the formation of decomposition products which implied that the guanidine product possibly degrades under harsh conditions. The reaction with excess of TFA in a water/acetonitrile mixture was deemed the most effective and convenient method because it afforded the desired product in high (86%) yield with minimal isolation issues.

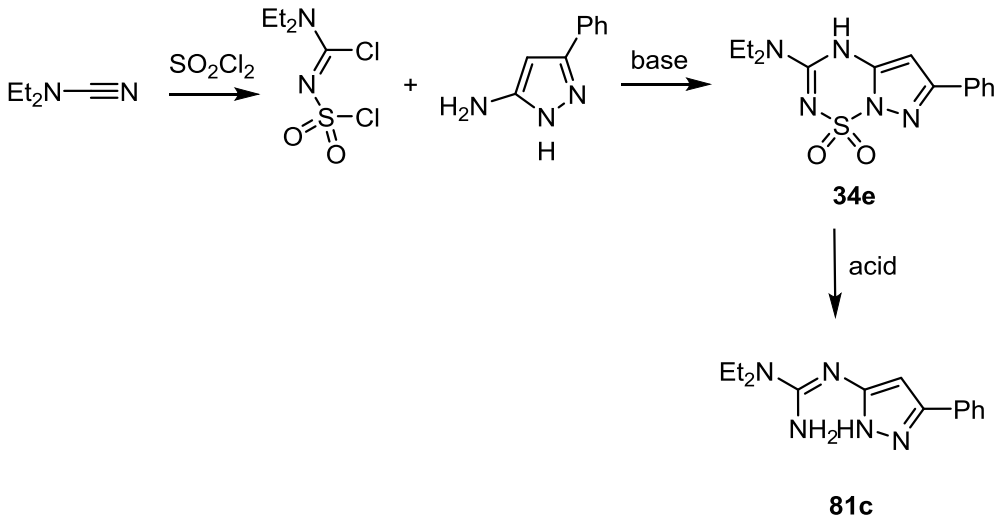
To test whether the sulfonic acid group could be removed, a small quantity of **85** was heated in acetonitrile at reflux temperature (**Scheme 105**). After 72 hours, NMR analysis indicated the mixture contained >90% starting material and a complex mixture; nevertheless, no signals for **81c** were distinguished. Sulfonic acid **85** was heated in 10%v/v water in acetonitrile for 24 hours and similarly, there was no conversion. The sulfonic acid adduct **85** was heated in acetonitrile to reflux temperature and also with 5M aqueous sodium hydroxide in ethanol at 60°C but mostly starting material was observedfrom the crude ^{1}H NMR spectrum. No evidence for the formation of compound **81c** was observed.



Scheme 105: The attempted removal of the sulfonic acid group from compound **85**.

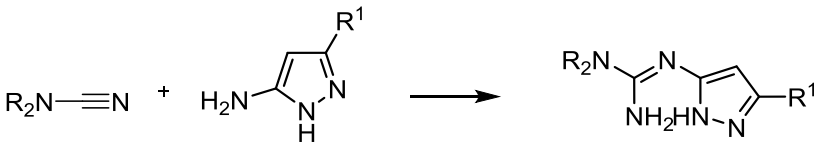
6.6.1 The formation of pyrazolo-guanidines by other means

The formation of the pyrazolo-guanidine **81c** has thus been accomplished in good yields, albeit over several steps (**Scheme 106**).



Scheme 106: The formation of pyrazolo-guanidines **81c** from readily accessible starting materials.

Zahariev et al[179] have documented the treatment of amines with dialkylcyanamides to provide guanidine derivatives. If this could be applied to 3-aminopyrazoles, then the formation of this pyrazolo-guanidine moiety would be possible over a single step from commercially available materials (**Scheme 107**).



Scheme 107: The proposed formation of pyrazolo-guanidines **81** from direct reaction of an aminopyrazole with a dialkylcyanamide.

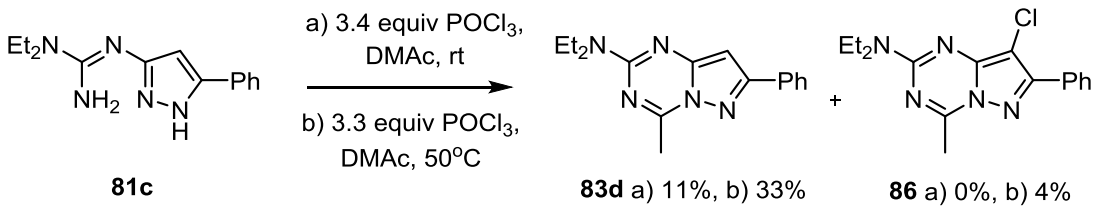
A mixture of 3-aminopyrazole and 1-piperidine carbonitrile was heated in ethanol at reflux temperature, however no reaction was observed. The reaction was repeated in the

absence of solvent with a slight excess of the cyanamide at 150°C which afforded a complex mixture of products by NMR observation. An experiment was also conducted with the two starting materials under neat conditions at 150°C and a catalytic amount of concentrated aqueous hydrochloric acid. A complex product mixture was again the outcome. These results indicated that the pyrazole and cyanamide would not react satisfactorily when diluted in solvent; but at elevated temperatures in neat conditions, the condensation was not sufficiently selective and many side reaction pathways became favourable. It was therefore reasoned that the original method of synthesising these pyrazolo-guanidines by SO_2-extrusion from the pyrazolothiatriazines was suitable as it was selective and achieved from cheap, commercially available precursors and reagents.

6.7 Formation of fused pyrazolo-triazines from dielectrophilic reagents

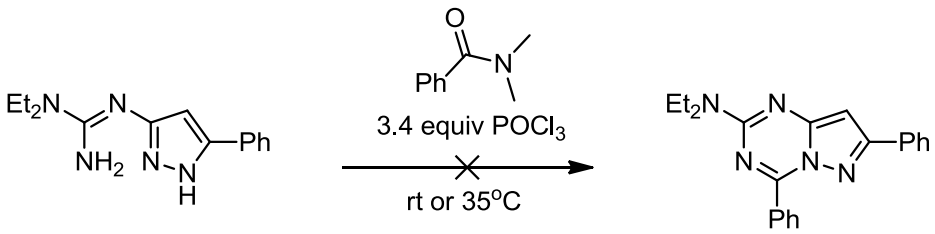
6.7.1 Triazine rings from amides and phosphoryl chloride

The possibility of forming a range of differently 4-substituted pyrazolo[1,5-*a*][1,3,5]triazines by the use of alternative dimethylamides under the same Vilsmeier-type conditions used previously to prepare compounds **83** was investigated. DMAc was first employed as an alternative to DMF; however, the conversion rate was markedly slower and after 24 hours, the product **83d** was furnished in 11% yield (**Scheme 108**). Heating the mixture gently improved the yield to 33%, along with a minor quantity (4%) of the 8-chloro compound **86**. The reagents formed in the Vilsmeier reaction are documented as prone to decomposition at higher temperatures and this would account for the cessation in conversion after the first 8 hours.[168]



Scheme 108: The formation of pyrazolo-triazines **82d** and **85** from $POCl_3$/DMAc.

A sample of *N'N'*-dimethyl benzamide was synthesised following literature procedures[180] and then subjected to $POCl_3$ in the presence of compound **81c**. Upon treatment to the same Vilsmeier conditions and some additional mild heating, the starting material was returned in near quantitative yield (**Scheme 109**).

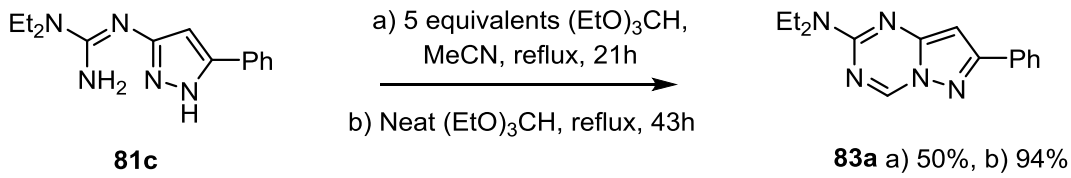


Scheme 109: Attempted synthesis of a triazine adduct from the reaction of a guanidine precursor with *N',N'*-dimethyl benzamide and $POCl_3$.

Substituted amides are less reactive than formamide towards phosphoryl chloride and furthermore the corresponding imine carbon intermediate is more sterically hindered by methyl or phenyl groups, for example, than by a simple hydrogen atom. It is for these reasons that the reaction or modified Vilsmeier-Haack reagents does not proceed readily. The application of activated amides to form triazine rings was rejected in favour of an alternative route from other commercial electrophiles.

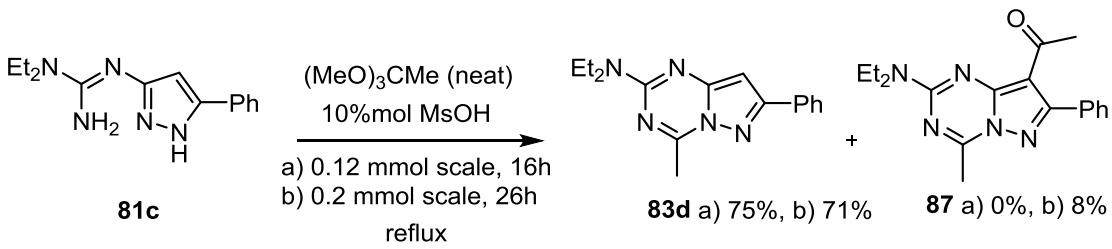
6.7.2 Formation of fused pyrazolo-triazines by condensation with orthoesters

Reports on the synthesis of triazine rings from guanidine type precursors have included the use of orthoesters.[181,182] Triethyl orthoformate proved an effective electrophile for the formation of the triazine **83a** albeit over longer reaction times in comparison to the Vilsmeier conditions (**Scheme 110**).



Scheme 110: The formation of pyrazolo-triazine **82a** from triethyl orthoformate.

The reaction between **81c** and trimethyl orthoacetate required more forcing conditions with the addition of a catalytic amount of methanesulfonic acid to provide the triazine **83d** in 75% yield under neat conditions. The reaction was repeated on a larger scale which also generated acylated compound **87** in 8% yield (**Scheme 111**).



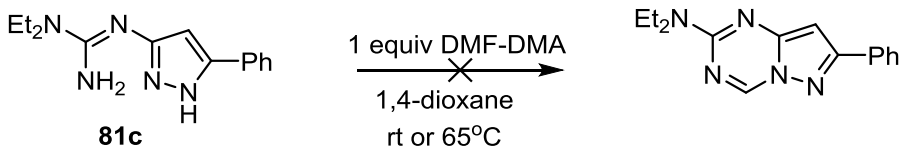
Scheme 111: The formation of 4-methyl-pyrazolo-triazine **83d** from trimethyl orthoacetate.

Orthoesters other than orthoacetate or orthoformate are not currently abundant commercially. The few which were available (and affordable) were only available from Europe or the United States via sea freight. Orthoesters can be readily synthesised from primary alcohols and ketene dimethyl acetals,[183] or primary alcohols and nitriles via the Pinner reaction.[184] However, these syntheses would involve labour-intensive processes

and purifications. It was therefore important to explore other means of electrophilic condensation to form a triazine ring before attempting the orthoester route of synthesis.

6.7.3 Condensation of amines by DMF-DMA

Abu-Shanab et al have reported *N*,*N*-dimethyl formamide dimethyl acetal (DMF-DMA) as a condensation agent for various cyclic amines.[183] Compound **81c** was treated with a single equivalent of DMF-DMA in dioxane, and only starting material was returned (**Scheme 112**).



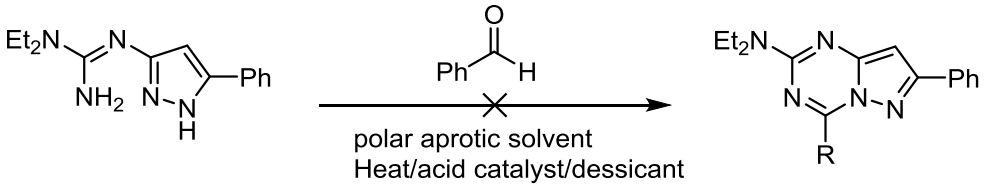
Scheme 112: The treatment of compound **81c** with DMF-DMA returned the starting material.

This reaction may require neat conditions or more excessive heating but did not warrant further investigation as a good yield was already obtained with cheaper reagents.

6.7.4 Condensation of amines by aldehydes

Following a preparation for condensations to form imidazo-quinazolines,[184] substrate **81c** was exposed to a stoichiometric quantity of benzaldehyde in acetonitrile with some sodium sulfate desiccant. No conversion was observed after stirring at room temperature overnight. Similarly with one equivalent of benzaldehyde in a 1:1 mixture of acetonitrile in acetic acid at reflux temperature, the starting material was returned in 69% yield after 43 hours. Further attempts were made to prepare triazines **83** from **81c**: (1) 1.5 equivalents of benzaldehyde and catalytic mesylic acid in acetonitrile at reflux

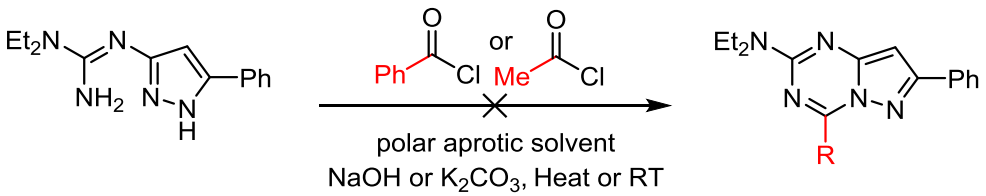
temperature, (2) 1.5 equivalents of benzaldehyde in methanol at reflux temperature and (3) in neat benzaldehyde at 100°C. After 2 days under these conditions, no product(s) were isolated and the starting materials were recovered in 84%, 98%, and 79% yields respectively (**Scheme 113**).



Scheme 113: Aldehydes were not sufficiently activated to react with guanidine **81c** under the conditions investigated.

6.7.5 Condensation of amines by acid chlorides

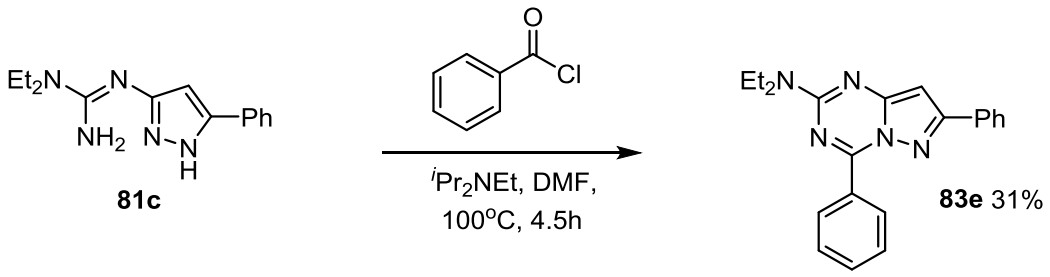
Acid chlorides were then explored in the place of corresponding aldehydes as more reactive electrophiles by employing benzoyl chloride or acetyl chloride. Treatment of guanidine **81c** under a series of common *N*-acylation conditions led to complex mixtures, presumably due to the lack of selectivity of the acylation reaction (**Scheme 114**).



Scheme 114: The reaction of benzoyl or acetyl chloride with guanidine **81c** led to complex mixtures.

Treatment with a single equivalent of benzoyl chloride returned the staring material under conditions including: (1) acetonitrile at reflux temperature (80% recovery), (2) similar conditions in the presence of triethylamine (71% recovery), and (3) with

triethylamine and 4-dimethylamino pyridine (50% recovery). In the case of the third reaction in the presence of 4-dimethylaminopyridine, other products had formed; however selective formation of a predominant product was not apparent and hence further purification was not attempted. When the conditions were altered to include DMF as a solvent and more liberal use of amine base, a triazine product **83e** was isolated from the product mixture in 31% yield (**Scheme 115**).



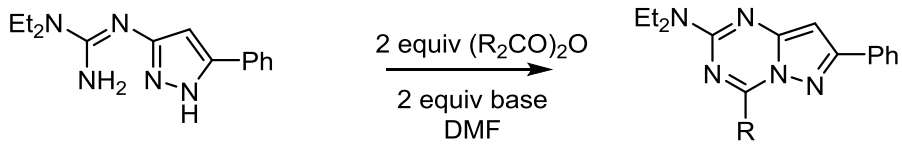
Scheme 115: The formation of 4-phenyl substituted pyrazolo-triazine **83e** from benzoyl chloride.

These results were nonetheless encouraging, and the condensation reaction was consequently investigated through use of anhydrides as an alternative means of acylation.

6.7.6 Condensation of amines by anhydrides

A sample of pyrazolo-guanidine **81c** was heated in neat acetic anhydride at reflux temperature for 15 minutes following a preparation from Giovanonni et al.[181] NMR analysis of the crude product suggested that the result was a complex mixture of acylated materials and possible dimerisation products. The reaction was repeated with the addition of catalytic aqueous hydrochloric acid; again no triazine product was observed. When a sample of compound **81c** was treated with 2 equivalents of benzoic anhydride under acidic conditions with methane sulfonic acid in acetonitrile, no single product could be isolated from the mixture.

Insuasty et al reported condensations of 1-acyl-3-(1H-pyrazol-5-yl) thiourea derivatives to triazine rings by means including triethylamine and DMF.[185,186] Similar conditions were successfully applied to reactions of pyrazolo-guanidine **81c** with anhydrides to afford the respective triazines, and results are summarised in **Table 17**.



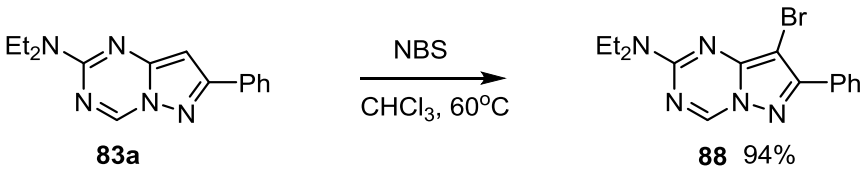
Scheme 116 (above), **Table 17** (below): The formation of pyrazolo-triazines **83** by condensation of anhydrides with pyrazolo-guanidine **81c**.

R	Conditions	Products	Yield [%]
Me	$^{i}Pr_2NEt$, 70°C	**83d**	42
Et	$^{i}Pr_2NEt$, 100°C	**83f**	66
Ph	Et_3N, 100°C	**83e**	60
Ph	$^{i}Pr_2NEt$, 100°C	**83e**	71

NMR analysis of crude products suggested that mixtures of other products were present (especially in the case of acetic anhydride reactions) which would likely be owing to the lower selectivity with decreasing steric bulk of the anhydride. Nonetheless, the formation of a small set of 4-substituted pyrazolo-triazines **83** was achieved from inexpensive and readily available electrophiles with overall satisfactory yields.

6.8 The reaction of pyrazolo[1,5-*a*][1,3,5]triazines with electrophilic bromine

The pyrazolo-triazine **83a** reacted with *N*-bromosuccinimide to form the brominated derivative **88** in 94% yield (**Scheme 117**) as a stable white solid. This result contrasts with bromination of the pyrazolothiatriazine ring system **34** where product stability issues prevented the application as a synthetically viable precursor (Chapter 4).



Scheme 117: Bromination of pyrazolo[1,5-*b*][1,2,4]triazine **83a** with NBS.

This result confirmed that the pyrazole carbon of compounds **83** is nucleophilic. The bromination reaction was slow compared to the formation of the analogous bromide from fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **34e** under similar conditions; however the stability of the final product was much greater. Pyrazolo[1,5-*a*][1,3,5]triazines are suitable synthons for subsequent chemistry at the carbon position by means of electrophiles or via aryl halide intermediates to facilitate C-C cross coupling reactions.[171,172] The reactivity of derivatives **88** is beyond the scope of this study and was not explored further during the course of this PhD project. This section of research was focused on synthesising triazine rings from cost effective and simple methods, which has been competently explored.

6.9 General conclusions

The tendency of some representative pyrazolo[1,5-*b*][1,2,4,6]thiatriazines to undergo ring cleavage at the sulfamide moiety was observed under a variety of conditions. Derivatives alkylated at the pyrazole ring such as compounds **64**, **65** and **75** were most

susceptible to this change. It was established that the parent compounds could undergo a similar conversion; however under comparatively stronger acidic conditions. The propensity to extrude the sulfur dioxide moiety was exploited for supplementary chemical transformations to form pyrazolo[1,5-*a*][1,3,5]triazines which included the application of Vilsmeier-Haack conditions to form triazines directly from parent thiatriazines **34**; or by treatment with acids (typically TFA or HCl) first to form a pyrazolo-guanidine intermediate **81**. Guanidines **81** were reasonably versatile reagents and could react with a range of cheap and commercially available electrophiles such as acid anhydrides or orthoesters to afford the pyrazolo-triazine ring product **83**. The fused triazine **83** was verified to contain an active C nucleophile at the pyrazole ring. This methodology has potential application in medicinal chemistry because pyrazolo[1,5-*a*][1,3,5]triazines, which are isosteres of the purine bases abundant in biological systems, are found to be active against a range of biological targets which interact with purines.

CHAPTER 7: REARRANGEMENTS AND OTHER TRANSFORMATIONS OF *N*-ALKYLATED PYRAZOLO[1,5-*B*][1,2,4,6]THIATRIAZINE DIOXIDES.

7.1 Introductory remarks

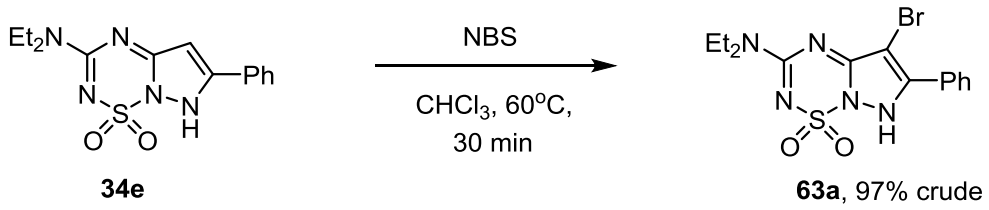
The results from Chapter 2 demonstrated that aminopyrazoles **32** which contained a bulky or electron withdrawing substituent at the 4-position were not reactive with dichloride compounds **1**. In light of this, it would be synthetically useful to establish methodology to install such a substituent after the formation of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine bicyclic core. It would also be important to not only demonstrate that protection with benzylic substituents could be achieved, but also to determine what conditions were required to remove these protecting groups and highlight when this was not possible. This chapter outlines the unusual transformations which resulted from attempted aryl cross-couplings and de-protections of pyrazolothiatriazines **34**.

As outlined in Chapters 5 and 6, the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34** and particularly N7-benzylated derivatives thereof are prone to cleavage of the thiatriazine ring and to the expulsion of sulfur dioxide under a variety of acidic conditions. This indicates that the fused pyrazolothiatriazine ring may be susceptible to cleavage by reaction with nucleophilic species. This chapter reports some such substitutions at the sulfamide group of compounds **34**, reflecting the inherent electron deficiency of the sulfur atom.

Given that the pyrazole carbon (C5) of compounds **34** has demonstrated a degree of nucleophilicity towards common electrophiles (Chapters 4 and 5), it was plausible that C5 could be activated in some fashion (by a halogen or stannyl group) or directly coupled in C-C cross-coupling reactions.

7.2 Forming activated intermediates towards aryl coupling

In Chapter 4 it was established that the pyrazole carbon of compounds **34** exhibits nucleophilicity and that a bromine atom could be installed at that site. For example, the 5-bromo derivative **63a** was obtained in 97% yield (**Scheme 118**).

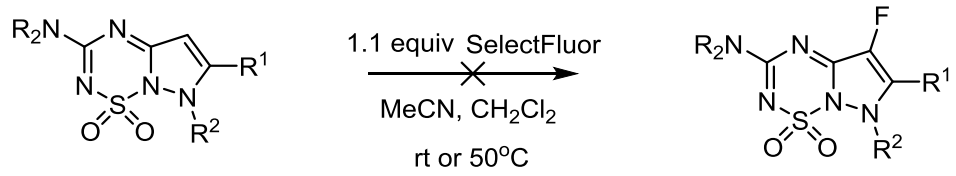


Scheme 118: Halogenation of C5 with N-bromosuccinimide (NBS).

Many of the C-C bond formation reactions catalysed by palladium complexes such as the Suzuki, Stille, Sonogashira, and Negishi couplings require activation of substrates, often by a halogen. Due to the unstable nature of the aryl bromide derivative **63** (see Chapter 4), the halogenation of an *N*-substituted material was explored with the aim of obtaining a more stable product with greater synthetic potential. The use of a protecting group or *N*-alkylated or *N*-acylated derivative of similar systems such as indoles,[104,189–191] fused or substituted pyrazoles,[103,192] and pyrroles[193] was conveyed in the literature to provide a stabilised product upon bromination. Acylation of pyrazolothiatriazines **34** had not been successful; therefore the *N*-benzyl derivatives **64** were explored. The reactivity of the methylene group of the benzyl substituent meant that there could be competing reactions at this site and at C5 with radical halogenating agents. However, there have been several reported examples of selective bromination of the nucleophilic C2 and/or C3 carbons of *N*-benzyl substituted pyrroles or at C4 of N-benzylated pyrazoles, using NBS, without apparent reactivity at the benzyl group.[193–196] Avoidance of an excess of halogenating reagent might enable similar selective reactions at C5 of with *N*-benzyl substituents pyrazolothiatriazines **64**.

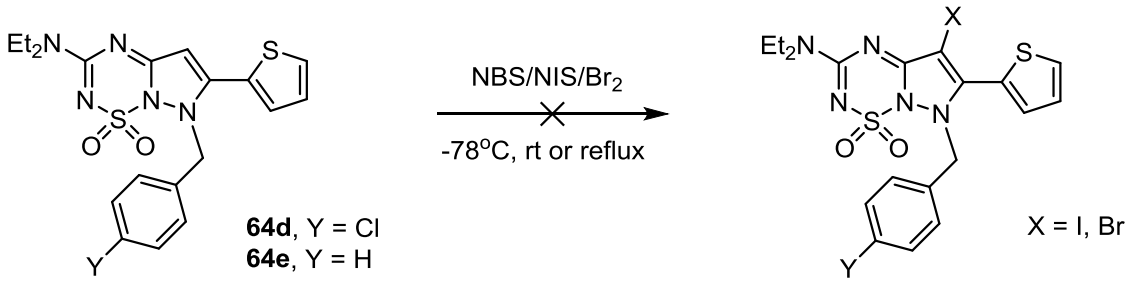
In the event, all attempts to form stable C5 bromo, iodo, or fluoro derivatives were unsuccessful. These experiments included the use of NBS, bromine, *N*-iodosuccinimide,

and SelectFluor®. In all cases, complex mixtures were formed which could not be adequately purified (**Schemes 119** and **120**).



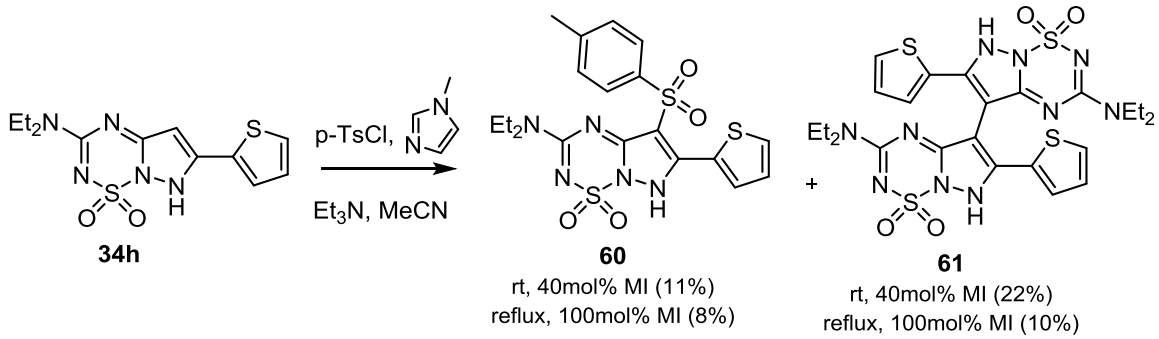
69 R_2N = Et, R^1=Ph, R^2=Me
68 N4 regioisomer of **69**
64d R_2N= Et, R^1=Thien-2-yl, R^2=4-Chlorobenzyl
34j R_2N =Me, R^1=Me, R^2=H

Scheme 119: Attempted fluorination of pyrazolothiatriazine derivatives with SelectFluor.



Scheme 120: Attempted bromination of benzyl protected substrates.

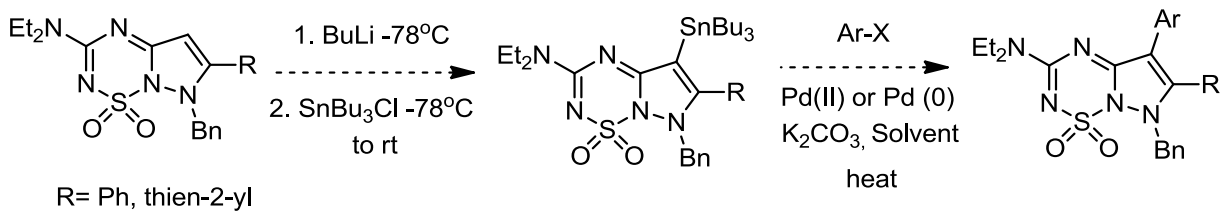
It is possible that the halogenation was not selective towards the pyrazole ring and that the mixtures of products arose from halogenation at the thiophene ring, benzyl substituent, and pyrazole ring. Another explanation is that the halogenation does proceed at one site but that the product may undergo various reactions in solution. One example of a dimer **61** was isolated from the reaction of a *C*-tosyl substrate **60** with the nucleophilic C5 of the precursor compound **34h** in situ (**Scheme 121**) which was outlined in Chapter 4.



Scheme 121: *N*-Methylimidazole catalysed sulfonylation of **34d**.

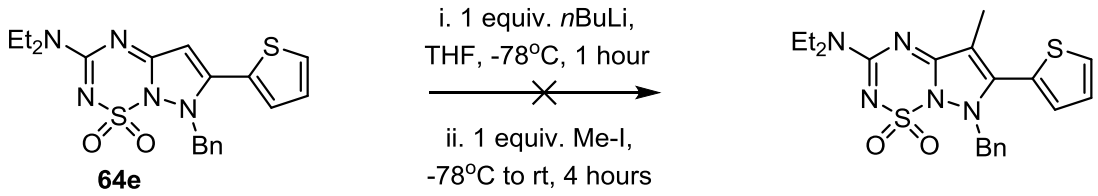
The C-tosyl product **60** had presented instability issues such as the decomposition to form **34h**, unknown side reactions in solution, and the formation of dimer **61** when in the presence of methyl imidazole as a sulfonyl transfer agent. Since a halide is also a good leaving group, similar reactions are plausible with a compound containing a halogen at the C5 position. It has not been possible to confirm this result because appreciable quantities of purified side products could not be isolated; however, these results do indicate that activation of the pyrazole carbon with a leaving group may lead to stability problems.

Because of the difficulties with handling 5-bromo derivatives, a different approach was taken. A Stille coupling reaction was attempted via the formation of a lithium intermediate at a much colder temperature (-78°C), and stannylating with tributyl tin chloride (**Scheme 122**).



Scheme 122: The proposed Stille coupling of benzyl protected pyrazolothiatriazines with aryl halides.

Preliminary treatment of substrate **64e** with *n*-butyl lithium, followed by quenching with methyl iodide was undertaken to determine if the substrate deprotonated selectively, or formed a mixture of methylated products (**Scheme 123**).



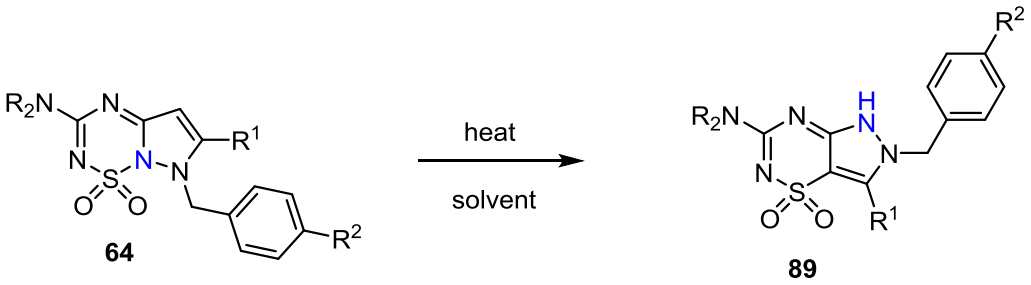
Scheme 123: The treatment of substrate **34e** with *n*-butyl lithium and methyl iodide.

Upon consumption of the starting material (as determined by TLC analysis), a ^{1}H NMR spectrum of the crude product suggested that a number of compounds were present. Two singlets at δ 2.4-2.5 ppm indicated methyl groups on a carbon; however, the complex nature of the ^{1}H NMR spectrum indicated that this reaction was not selective in the manner that was necessary and this approach was therefore abandoned.

7.3 The rearrangement of N7 benzyl pyrazolothiatriazines to the isomeric N6 benzyl pyrazolo-thiadiazines

Recent reports of direct aryl couplings of heterocyclic nucleophiles such as indoles,[197,198] imidazoles[199,200] and other carbon nucleophiles to aryl halides have been published.[201,202] In an attempt to apply this chemistry, bromobenzene was employed as a representative aryl halide and heated in polar aprotic solvents with either of substrates **64e** and **64d**, base and catalytic palladium sources. Under these conditions, the desired aryl coupled products were not observed, and instead a compound that did not appear to have incorporated the benzene substituent was isolated (see **Appendix C** for comparison). The products were eventually structurally assigned as the isomeric fused pyrazolo-thiadiazines **89** by mass spectrometry and X-ray crystallography of one representative **89b** (**Figure 57**). Originally, the cleavage of the N-S bond was

tentatively attributed to the palladium; however the rearrangement was also observed in the absence of a palladium source. This unusual reaction (**Scheme 124**) was investigated in further detail and results are summarised in **Table 18.**



Scheme 124: Rearrangement of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **64** to form fused pyrazolo[3,4-*e*][1,2,4]thiadiazines **89**.

Table 18: The formation of fused pyrazolo-thiadiazines **89**.

R_2N	R^1	R^2	Conditions	Product	Yield [%]
Et		Cl (**64d**)	$Pd(OAc)_2$, PPh_3, K_2CO_3, dioxane, 100°C, 17h	**89a**	Quant.
			PEPPSI-^{i}pr, PPh_3, K_2CO_3, dioxane, reflux, 18h	**89a**	92
			$Pd(dba)_2$, K_2CO_3, dioxane, reflux, 3.5h	**89a**	75
			$Pd(dba)_2$, dioxane, reflux, 6.5h	**89a**	74
			Dioxane, reflux, 4h	**89a**	99
			$Pd(OAc)_2$, DMAc, KOAc, 140°C, 15.5h	**89a**	78
			$PdCl_2(PPh_3)_2$, K_2CO_3, Dioxane, 80°C, 16h	**89a**	38
			DMF, K_2CO_3, 80°C, 18h	**89a**	74
			MeCN, reflux, 14.5h	**89a**	82

			Toluene, reflux, 46h	**89a**	79
			EtOAc, reflux, 44h	**89a**	74
Et		H (**64e**)	$Pd(dba)_2$, K_2CO_3, dioxane, reflux	**89b**	70
			$Pd(OAc)_2$, PPh_3, K_2CO_3, DMAc, 100^oC	**89b**	78
Me	Me	H (**64f**)	$Pd(OAc)_2$, PPh_3, K_2CO_3, dioxane, reflux	**-**	-
			PEPPSI-ipr, PPh_3, K_2CO_3, dioxane, reflux	**-**	-
Et	Ph	Cl (**64b**)	$Pd(OAc)_2$, PPh_3, K_2CO_3, dioxane, reflux	**89c**	91
			Dioxane, reflux, 8 hours		88
			EtOAc, reflux, 2 days		83

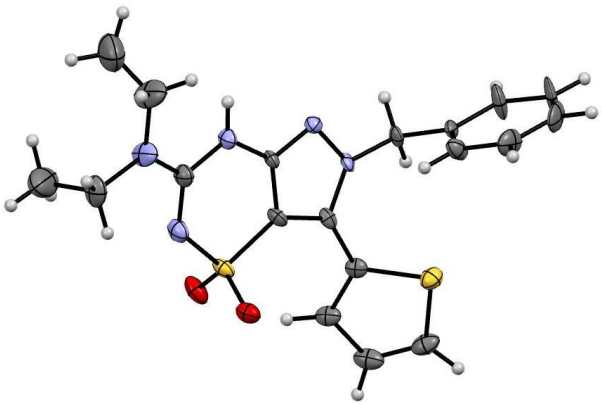


Fig 57. ORTEP diagram of compound **89b**. The structure shown is one of five crystallographically independent molecules in the crystal lattice.

The same product **89b** also formed when iodobenzene was added in place of bromobenzene. Experiments were conducted in the absence of base and also in the absence of palladium with base in order to explore their influence on the reaction. It was noted that in some cases, yields were lower and decomposition products had formed but

that the product **89** was still the only compound isolated. This might be attributable to the amount of insoluble solids within the reaction media which slowed the formation of the rearranged product. It also seemed that basic conditions favoured the reaction; likely due to the removal of residual acid which was known to cleave N7-benzylated derivatives **64** (Chapter 5). Such degradation would be exacerbated with heating.

The results in **Table 18** also illustrate the effect of solvent on this transformation to form fused pyrazolo-thiadiazines **89**. Noticeable differences in reaction times between solvents with higher boiling points were evident. There did not appear to be a clear correlation between product yields and solvent polarity; however, the solubility of the starting material did enable shorter reaction time. The lower yields from DMF and DMAc are attributable to isolation difficulties.

The desired aryl-aryl coupling reaction was not successful with substrates **64f**, **68**, or **69** (**Figure 58**), nor was ring rearrangement observed. In the cases of compounds **64f** (6-methyl and 7-benzyl substituted) and **69** (6-phenyl and 7-methyl substituted), complex mixtures of products were obtained. Compound **68** (4-methyl substituted) placed under the same conditions was recovered unchanged.

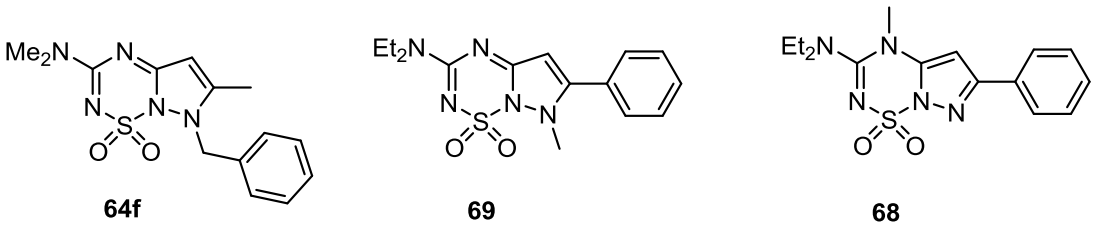
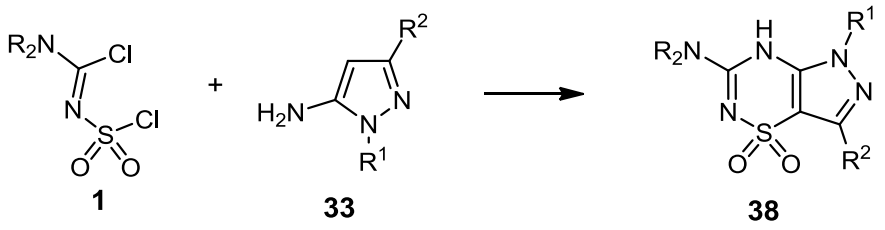


Fig 58.

These results indicated that only compounds bearing both 6-aryl and 7-benzyl substituents were prone to ring rearrangement, suggesting that a high level of aromaticity of the substrates and substituents which can stabilize charge (e.g. benzyl) may serve to lower the activation barrier for the reaction.

This simple transformation gave access to a series of novel *N*-benzyl pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides **89**. Related derivatives of the pyrazolo[3,4-

e][1,2,4]thiadiazine ring system (compounds **38**) were synthesised in Chapter 3 by means of *N*-substituted pyrazoles (**Scheme 125**); however yields were often low when dichloride **1b** was used, or when substituents on the pyrazole ring were sterically demanding.

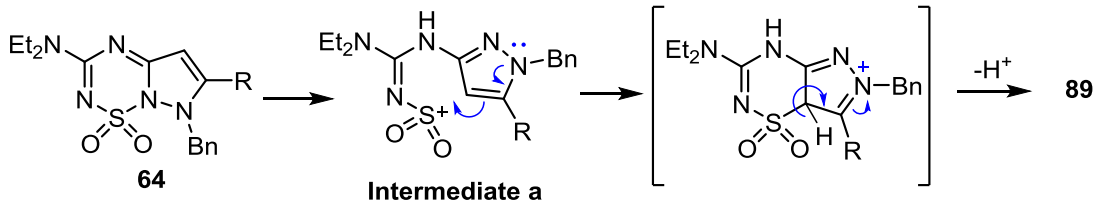


Scheme 125: The formation of dioxides **38** from treatment of dichlorides **1** with N-substituted 5-amino pyrazoles **33**.

The examples **89a**, **89b** and **89c** all contain two large substituents on the pyrazole ring, and were afforded in near quantitative yields from simply heating in a suitable solvent. This ring rearrangement reaction represents an alternative synthetic method for preparation of pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides.

7.3.1 Mechanism of formation

The cleavage of the sulfamide bond in compounds **64** appeared to be facilitated by the increased temperature, and the pyrazolo-thiadiazine **89** would be the thermodynamically favourable product. The proposed mechanism in **Scheme 126** is consistent with the mechanism that was proposed for the formation of the sulfonic acid **85** outlined in Chapter 6.



Scheme 126: The proposed mechanism for the formation of compounds **89**.

The pathway to intermediate *a* remains unclear for this reaction. It was established earlier (Section 6.2) that when acid is present with compounds **64**, they undergo the expulsion of SO_2 from the ring. However, despite the absence of acid in the present set of experiments, the N-S bond in the thiatriazine ring must break initially in order to form the observed product.

The unusual reactivity of these particular substrates **64** could be attributed to the presence of a suitable *N*-substituent on the pyrazole ring, and to the higher temperatures. Also the presence of the *N*-benzyl group might mean that the product **89** is not as susceptible to further ring opening, and so a sulfonic acid such as that in compound **85** (Chapter 6) was not isolated. Compounds **34**, which do not contain an N7-benzyl substituent, did not show clearly noticeable signs of similar ring rearrangement on heating during the wide range of reactions carried out on these compounds as discussed in earlier chapters. Compounds **38**, which contain the same core ring structure as compounds **89**, also did not exhibit signs of degradation under mildly acidic conditions or applied heat. At this point, it is unclear why compounds **64** specifically are so prone to rearrangement and acid-mediated loss of SO_2, but it is thought to be a factor of stability.

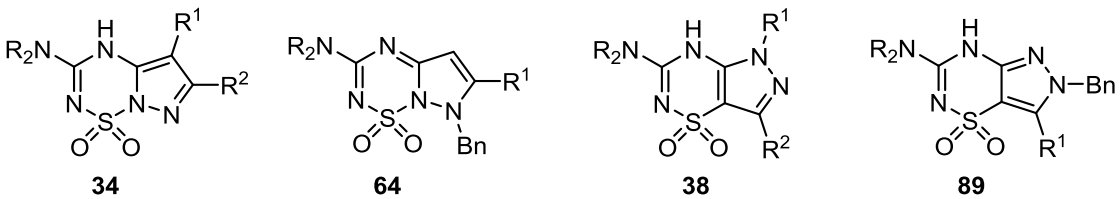
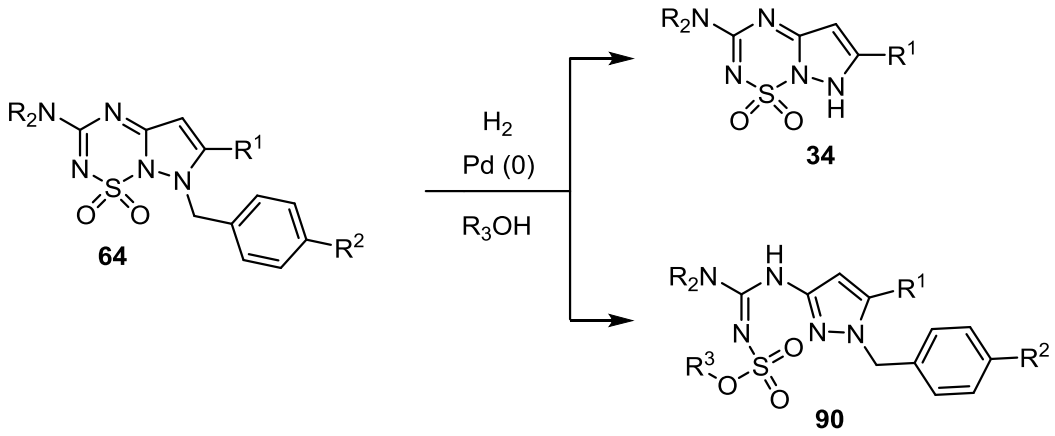


Fig 59. Fused pyrazolothiatriazines **34** and **64**, and pyrazolo-thiadiazines **38** and **89**.

7.4 Cleavage of the thiatriazine ring by nucleophilic attack of the sulfamide moiety

7.4.1 Alcoholysis of benzylated pyrazolo[1,5-*b*][1,2,4,6]thiatriazines

The results in chapter 5 outlined the successful synthesis of a small series of N-benzyl protected pyrazolothiatriazines **64** to provide novel derivatives as well as investigate the reactivity and selectivity of compounds **34** towards alkylation with benzyl groups. The next study was to determine whether or not the benzyl groups of 7-benzyl-pyrazolothiatriazines **64** could be readily cleaved by standard deprotection procedures. The first approach evaluated was catalytic hydrogenolysis. Treatment of benzylic substrates **64** with hydrogen and catalytic amounts of activated palladium in alcohols often did not afford the expected deprotected compounds **34**. Instead, alcoholysis of the thiatriazine ring afforded sulfamate derivatives **90** (**Scheme 127**, **Table 19**) with significant substrate dependence.



Scheme 127: The competing reaction of hydrogenolysis of the benzyl substituent and alcoholysis of the thiatriazine ring.

Table 19: Synthesis of compounds **34** and **90**.

R_2N	R^1	R^2	R^3	Conditions	Product	Yield [%]
Et		OMe (**64g**)	Et	20mol% $Pd(OH)_2/C$, $H_2(g)$, rt	**90a**	27
				rt	**90a**	65
		Cl (**64d**)	Et	20mol% $Pd(OH)_2/C$, $H_2(g)$, rt	**90b**	50
				20mol% Pd/C, $H_2(g)$, rt	**90b**	71
		Cl (**64d**)	Me	10mol% Pd/C, NH_4CO_2H, reflux	**90c**	43
				10mol% Pd/C, rt	**90c**	71
Et	Ph	Cl (**64b**)	Et	10mol% Pd/C, $H_2(g)$, rt	**34e**	70
			N/A	50mol% $Pd(OH)_2/C$, $H_2(g)$, rt, EtOAc	**34e**	93
Me	Me	H (**64f**)	Me	rt	**90d**	100 crude
				reflux	**90d**	100 crude
			Et	10mol% Pd/C, $H_2(g)$, rt	**34j, 90e**	48, 52
			^{t}Bu	20mol% $Pd(OH)_2/C$, $H_2(g)$, 35^{o}C	**34j**	79
			N/A	20mol% $Pd(OH)_2/C$, $H_2(g)$, rt, EtOAc	**34j**	83
			N/A	50mol% $Pd(OH)_2/C$, $H_2(g)$, rt, EtOAc	**34j**	96

Notably, substrates containing the phenyl ring at C6 or a methyl group at this position (**64b** and **64f** respectively) were shown to undergo hydrogenolysis to provide the parent compounds **34**; however, in the case of the substrate **64f** this reaction competed with the alcoholysis and compounds **34j** and **90e** were formed in similar yields. The alcoholysis of the substrate with a phenyl substituent at C6 (**64b**) was remarkably slower than that for the substrate containing a methyl group at this position (**64f**). The exact reason remains unclear as the increased steric interference of the phenyl substituent adjacent to the benzyl group would be expected to decrease the rate of the hydrogenolysis reaction. It did appear that the hydrogenolysis of **64b** took longer than hydrogenolysis of **64f**; which meant that the substrate containing the phenyl ring was not as prone to nucleophilic attack at the sulfamide group. Interestingly, when the hydrogenolysis of **64f** was carried out in *t*-butanol, a good yield of the debenzylated product **34j** was obtained, indicating that this alcohol solvent was too sterically bulky to cleave the thiatriazine ring.

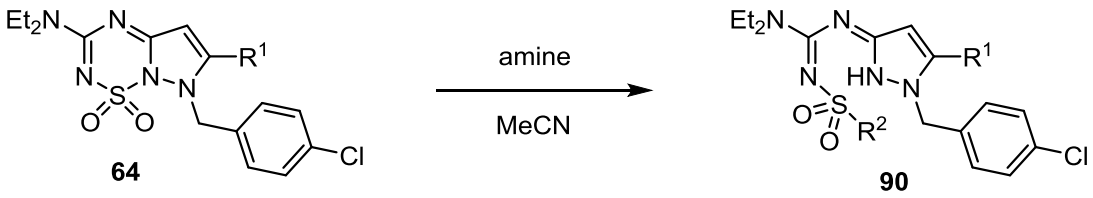
Use of ammonium formate as an alternative hydrogen source gave rise to other products. Singlets at around δ 9 ppm were observed in the crude ^{1}H NMR spectrum which indicated that some formylation or triazine formation (Chapter 6.3) had occurred. The reaction of substrate **64g** which contained a PMB group on the N7 atom did not proceed smoothly and only decomposition products were observed. The product from this reaction (**90a**) suffered from poor stability, similar to the parent compound **64g**, which was documented earlier (Chapter 5).

The 6-(thien-2-yl)-substituted substrates **64d** and **64g** only afforded alcoholysis products, unlike the 6-phenyl analogue **64b**, which provided the debenzylated product **34e**, perhaps indicating poisoning of the catalyst by the thiophene sulphur atom. The mild catalyst poisoning effects of the sulfur atom in thiophenes on palladium/carbon surfaces has been recognised.[203,204] Ethyl acetate was utilised as an alternative solvent for the deprotection which afforded the debenzylated compounds **34** in good yields with minimal side reactions. Reports in the literature[205] have described the lower efficiency of hydrogen uptake by ethyl acetate compared to alcohols which affects the rate of hydrogenation or hydrogenolysis reactions. After two days, complete

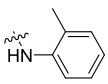
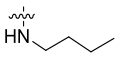
conversion was not reached using 10% Pd/C catalyst and instead a higher loading of palladium (20-50%) became necessary.

7.4.2 Aminolysis of benzylated pyrazolo[1,5-*b*][1,2,4,6]thiatriazines

The propensity towards nucleophilic attack at the sulfamide group was further investigated employing amines as nucleophiles. Cleaved sulfamides **90f-h** were afforded from treatment of N7 benzyl substrates **34b** or **34d** with a primary or secondary amine in acetonitrile (**Scheme 128**, **Table 20**). The structural assignments for these compounds were based on X-ray crystallography of one representative **90f** (**Figure 60**).



Scheme 128 (above), **Table 20** (below): The synthesis of sulfamides **90** by aminolysis of the thiatriazine ring.

R^1	R^2	Conditions	Product	Yield [%]
Ph (**64b**)		1.5 equivalents morpholine, 82°C, 1h	**90f**	42
Ph (**64b**)		2 equivalents o-toluidine, reflux, 2h	**90g**	66
Thien-2-yl (**64d**)		1.5 equivalents ^{n}Butylamine, 50°C, 45min	**90h**	71

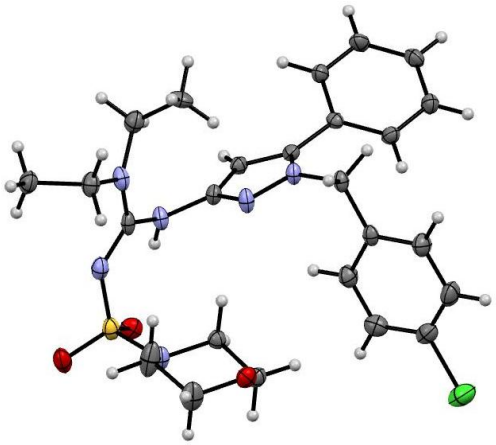
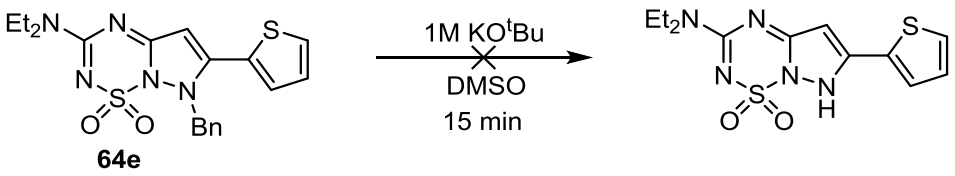


Fig 60. ORTEP diagram of the morpholine sulfamide **90f**.

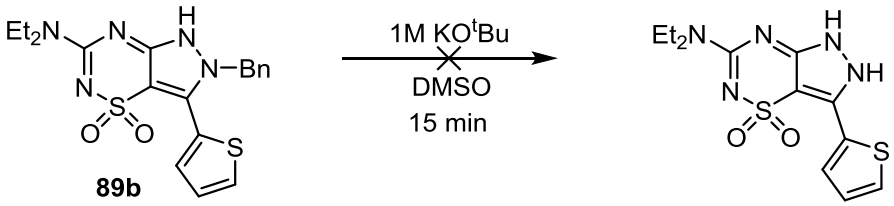
The cleavage of the sulfamide bond to form compounds **90** by amines or alcohols indicates that the pyrazole-N-SO_2 bond is preferentially cleaved over the guanidine-N-SO_2 bond. These results may also provide some indirect evidence of "intermediate *a*" in the proposed mechanisms of formation of guanidines **81** and **85**, which were discussed in Chapter 6.

7.5 Attempts to remove *N*-benzyl and *N*-(*p*-chloro)benzyl substituents by other means

Alternative deprotection methods for *N*-benzyl compounds, specifically those for azaheterocycles, were investigated in order to avoid the use of palladium or platinum. Haddach, Kelleman and Deaton-Rewolinski reported the *N*-debenzylation of aromatic heterocycles, including fused imidazoles and pyrroles, with a combination of potassium tert-butoxide (KO^tBu) and DMSO in air or with oxygen.[206] Samples of benzyl protected substrates **64d** and **89b** were treated with a suspension of KO^tBu in anhydrous DMSO in air (**Scheme 129, 130**).



Scheme 129

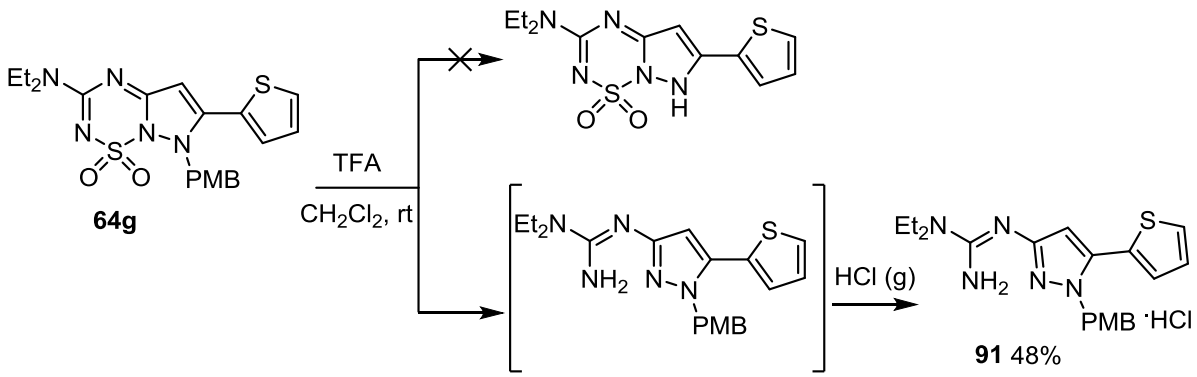


Scheme 130

Within a 15 minute time period, the harsh conditions had led to the decomposition of the substrate as indicated by NMR spectroscopy and TLC analysis. No further attempts were made to debenzylate these materials, and the conditions described above were deemed too harsh for substrates **64d** and **89c**.

7.6 Attempted removal of PMB amine protecting group

Deprotection of the fused-pyrazolothiatriazine containing a thiophene moiety was not achieved by catalytic hydrogenolysis. A 4-methoxybenzyl (PMB) protected substrate **64g** was selected in order to demonstrate whether or not the PMB group could be cleaved. Literature methods for removal of PMB from amines include the use of DDQ, TFA or dilute aqueous hydrochloric acid.[207] under these conditions, the acid simply caused extrusion of the sulfur dioxide moiety and no de-protection product was observed. One reaction which led to an appreciable quantity of the guanidine is outlined below (**Scheme 131**).

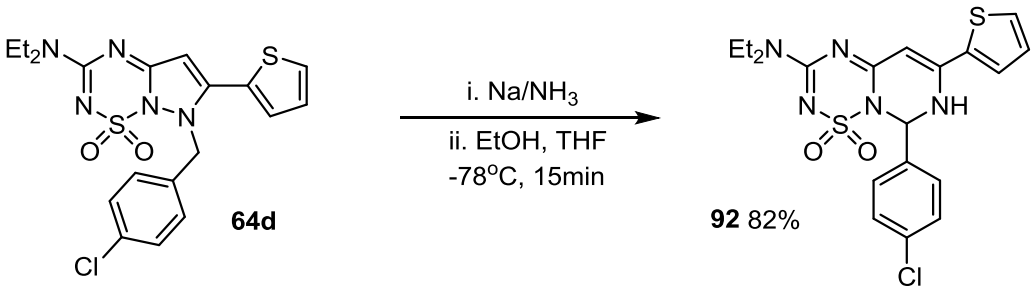


Scheme 131: The formation of guanidine hydrochloride salt **91** by treatment of **64g** with TFA and HCl.

The guanidine intermediate was evidently unstable as the free base which was apparent by physical observations and NMR experiments. The hydrochloride salt **91** was obtained as a stable material by bubbling HCl gas through a dichloromethane solution of the crude product.

7.7 The ring expansion of pyrazolothiatriazines by "dissolving metal reduction"

Birch reduction conditions have been reported in the literature as successful means to remove benzylic protecting groups from aromatic azaheterocycles or hindered amines.[208,209] Compound **64d** was allowed to react with sodium in liquid ammonia which furnished the ring expanded compound **92** in place of the expected debenzylated compound (**Scheme 132**).



Scheme 132: Ring expansion of **64d** by means of sodium in liquid ammonia.

It is worth mentioning that this rearrangement represents the first ring expansion of the pyrazole ring without the requirement of a strongly electron withdrawing ester substituent. This could be seen as an alternative to the ring expansion reactions using electron withdrawing substituents, which has the added benefit of leaving a free NH group as a synthetic handle.

Attempts to replicate this rare example of a ring expansion from other similar conditions thus far have remained unsuccessful, in part due to the sensitive nature of the substrates. These include the use of ethylene diamine in place of liquid ammonia, and lithium in place of sodium. In these cases, complex mixtures were the result. Unfortunately, due to time constraints of the PhD research, this chemistry was not explored any further. Nevertheless, understanding of this reaction may be developed through additional studies with other related substrates.

7.8 General conclusions

The potential for C-C cross coupling reactions was envisioned due to the inherent nucleophilicity of the pyrazole carbon C5; however throughout the course of these studies, compounds with C5-activating groups such as a halide or a tosyl moiety were not sufficiently stable to be synthetically useful. Instead, these products underwent side-reactions, including dimerisations. This did lead to the exploration of *N*-benzyl derivatives as substrates for direct aryl couplings to aryl halides, and thus the thermal rearrangement to form pyrazolo-thiadiazines **89** was discovered. It appeared that the

products **89** were more thermodynamically favoured than the precursor compounds **64**, and interestingly the regioisomers **66** did not undergo this transformation.

The difficulties in deprotecting the N-benzyl compounds **64** have provided further insight into the general stability of these unusual compounds. In addition to being prone to the extrusion of the SO_2 moiety under acidic conditions, and rearrangement from heating, the compounds **64** were not stable to oxidising agents such as DDQ or the combination of $KO^tBu/DMSO/O_2$. The susceptibility of compounds **64** towards nucleophilic attack by alcohols and amines has also been demonstrated.

The unusual rearrangement of pyrazolothiatriazine dioxides **64** to form isomeric pyrazolo-thiadiazine dioxides **89** provides insight into the relative thermal stabilities of the ring systems of compounds **34** and **38**. Compounds **34** are more prone to nucleophilic attack, extrusion of sulfur dioxide and also ring expansions, perhaps due to the relatively lower stability of the heterocyclic core. The rare ring expansion to form the pyrimido-thiatriazine **92** from Birch reduction conditions outlines the potential for further “ring expansion” transformations from radical sources.

CHAPTER 8: X-RAY CRYSTALLOGRAPHY

8.1 Introductory remarks

As is usually the case with bicyclic ring-condensed products generated from dichlorides **1**, the lack of contiguous NMR-responsive nuclei in the core heterocyclic systems of fused ring systems **34** and **35** meant that X-ray crystallographic studies of several representative compounds were critical in confirming the exact connectivity of the atoms constituting the heterocyclic core. X-ray crystallography has enabled the elucidation of the heteroatom-rich compounds which have been synthesised throughout this body of research and the results have been discussed in brief throughout Chapters 2-7. This chapter details the technical and experimental aspects of collecting crystal data; beginning at the crystal growing phase and continuing on to the data collection and refinement details for some representative compounds.

8.2 Crystallisation methods

With the easy availability of modern high powered computers and automated software, obtaining a diffraction-quality crystal is arguably the largest barrier to solving the molecular crystal structure of a sample. There are a series of techniques recommended for assistance in growing single crystals; often for the purposes of protein crystallography but which also apply to small molecules.[210,211] Complex crystallisation methods such as sublimation and hydrothermal syntheses are utilised to grow single crystals of a variety of organic and inorganic materials; anything from silicon metal or a protein to various transition metal complexes.[212] Crystals of the small heterocyclic organic molecules afforded from the present research on dichloride compounds **1** were obtained from simple solvent recrystallisation methods. These straightforward methods have provided single, X-ray quality crystals for Bragg diffraction studies. The

techniques which were utilised in this body of work all fall into the category of "solvent recrystallisation" methods which includes five distinct techniques.

1) The **cooling method** was the simplest procedure where a saturated solution of the solute to be crystallised was produced by heating a sample of the analyte to the boiling temperature of the solvent, and then allowing the solution to cool to room temperature. If necessary, the solution was then cooled further in an ice bath. This method provided rapid crystal growth; however this was not optimal since, the solvent molecules become more likely to crystallise with the analyte which in some instances complicated the refinement process.

2) The **slow evaporation** technique was favoured with these ring systems which involved simply evaporating solvent from the solution of the compound until saturation was reached and crystals began to "grow" or form. Generally this was achieved with two solvents of different volatility and in which the analyte had a greater solubility in the more volatile solvent. When the first solvent began to evaporate, the crystals would form. Solvents that were selected mainly included dichloromethane or chloroform as the volatile solvent and acetonitrile, methanol or ethyl acetate as the co-solvent.

3) The **vapour diffusion** method was the slowest but generally most successful method for growing crystals. Two vials were selected where one would fit inside the other. The inner vial contained the analyte in a small quantity of slow evaporating solvent such as acetonitrile, chloroform or methanol. The larger vial contained a solvent which is more volatile than the first solvent, and this worked best with diethyl ether which did not dissolve pyrazolothiatriazines very effectively. The vial was then capped and as the second solvent diffused into the first, the product would crystallise slowly.

4) The **Liquid/liquid diffusion** method involved carefully layering a low density solvent on top of a higher one in a thin tube. One solvent must dissolve the analyte more than the other. This technique was never successful in the present work.

8.3 Loading of single crystals into the diffractometer

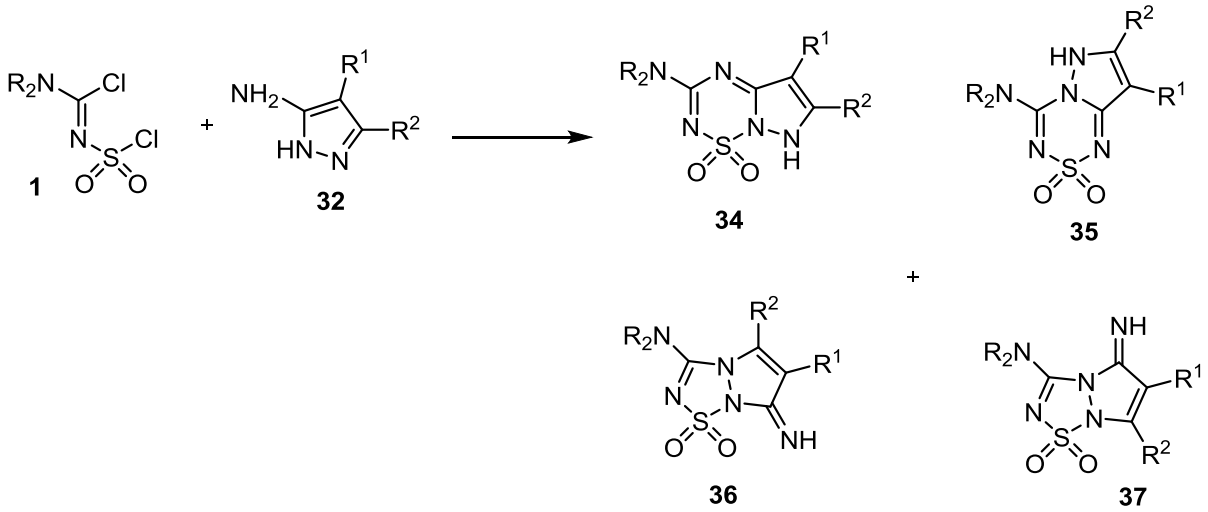
Single crystals were grown and observed under a microscope to assure the crystal quality. A suitable crystal was one which reflected light from the microscope lamp, was free of observable cracks/chips or other impurities, had a high clarity (lack of cloudy appearance), and had well-formed faces. The size of the desired crystal needed to be between 0.1-0.5mm in every dimension. Crystals of which a dimension was less than 0.1mm would not diffract X-rays efficiently, and one which had a dimension greater than 0.5mm would be outside of the path range for the X-ray beam. This meant that often the crystals required cutting with a small scalpel under the microscope into smaller fragments while ensuring that these fragments did not have damaged or chipped edges as a result. Once an acceptable sample was obtained, the individual crystal was mounted on a small loop in inert oil (**Figure 61**) then transferred to the cold gas stream of the diffractometer (typically performed at 150 K to ensure a minimal atomic vibrational motion). Further experimental features such as specific solvents of crystallisation for each crystal sample, diffractometer settings, experimental refinement and crystal parameters are detailed in Chapter 9.

Fig 61. A sample of **34a** loaded onto a loop in Paratone-N® oil. The image was captured on the X-Calibur video microscope at Adelaide University.

8.4 Characterisation of pyrazolo[1,5-*b*][1,2,4,6]thiatriazines by X-ray crystallography

8.4.1 Structural elucidation and ellipsoid plots

Crystallography was essential for samples **34** to be discriminated from the other three possible isomers **35-37** which could have arisen from the condensation between dichlorides 1 and 3-aminopyrazoles **32** (**Scheme 134**).



Scheme 134: The four possible isomeric products from the condensation between 3-aminopyrazoles **32** and dichlorides **1**.

Six crystal structures of the twelve different pyrazolo[1,5-*b*][1,2,4,6]thiatriazine ring products were collected and published.[99] Generally, the most effective method of crystal growth was **slow evaporation** from methanol and dichloromethane mixtures and the **cooling method** which provided adequate single crystals for X-ray analysis. The **vapour diffusion** method provided crystals which were opaque upon viewing under a microscope. **Figure 62** (below) illustrates the ORTEP (The Oak Ridge Thermal Ellipsoid Plot) diagrams obtained from single crystal X-ray diffraction experiments of some pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxides.

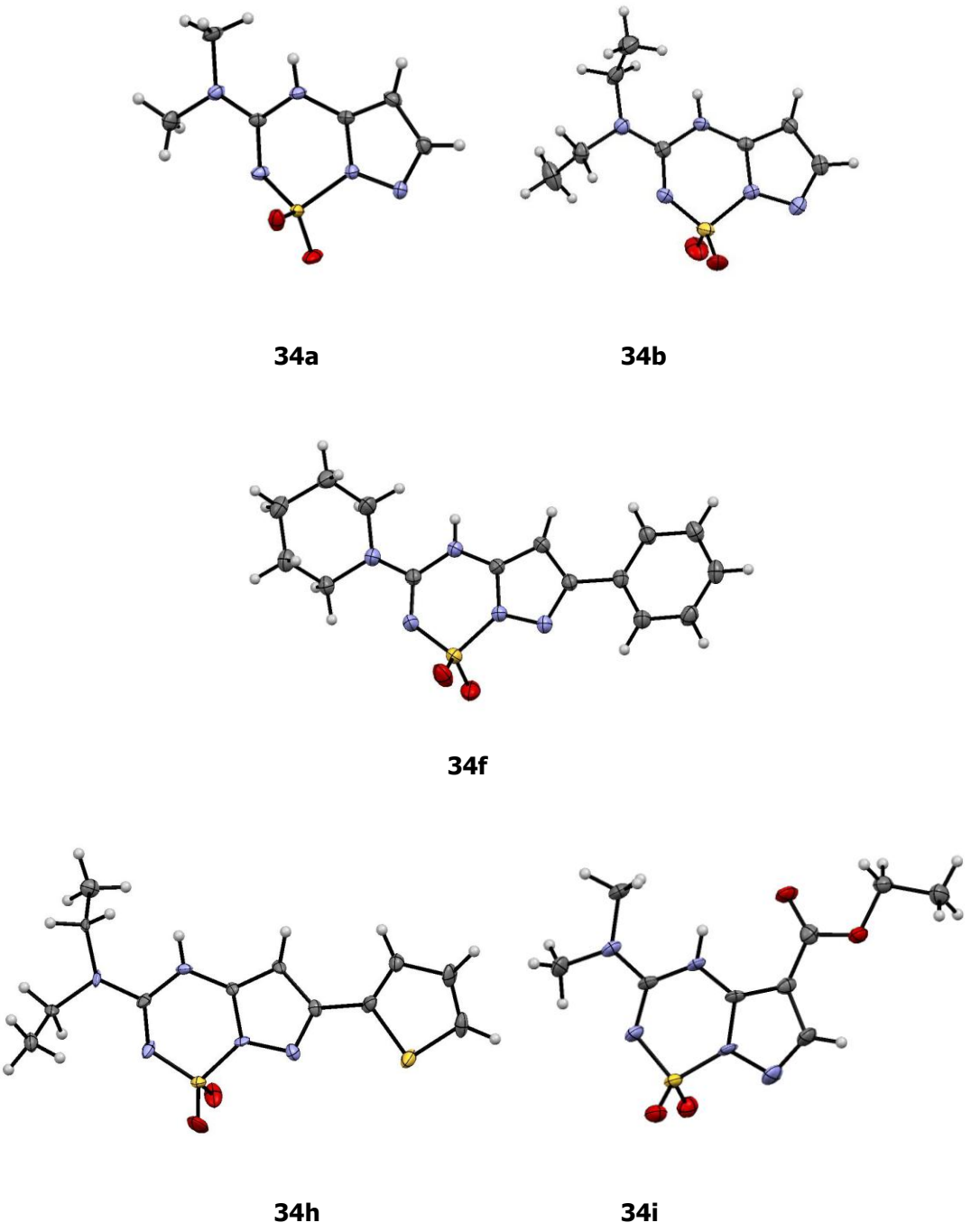


Fig 62. ORTEP diagrams of **34a**, **34b, 34f**, **34h**, and **34i**. In **34f** and **34h** the lattice molecules, DMSO and H_2O, respectively, have been omitted for clarity.

The crystal structure of **34a** contained two crystallographically independent, but structurally identical, molecules. In **34f** and **34h** the lattice molecules, DMSO and H_2O respectively, were present in the repeating crystal lattice which is a common occurrence when crystals are grown from a solvent with similar polarity to the analyte.

Sample **34g** was the first problematic structure to solve because it appeared to contain two heavy atoms on the thiophene moiety. Furthermore, the compound had co-crystallised with a molecule of DMSO solvent from the crystal growth process, which further complicated the refinement process due to the presence of several large areas of electron density which could not be easily clarified. **Figure 63** below contains the ORTEP diagram of the solved structure, whereby the thiophene sulfur atoms were placed in partially occupied sites.

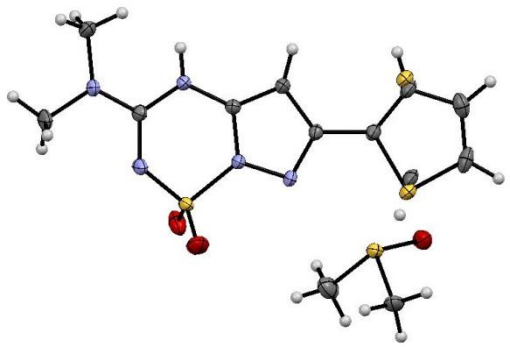
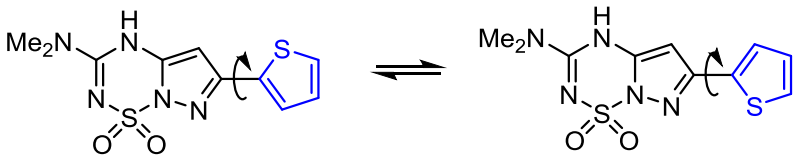


Fig 63. An ORTEP diagram of the repeating lattice of **34g**.DMSO crystals.

This is a case of *disorder* which means that the crystal structure of the compound exists in two or more energetically similar conformations.[65] The example with rotation about a single bond in the thiophene moiety is such a case. In the structure to be refined, a superposition of both cases became apparent (the thiophene moiety rotated "up" and "down" as shown in **Scheme 135**). Once recognized, such a disorder can be refined relatively easily and is called *positional disorder* where one atom occupies more than a single site (split atomic sites).[62] In a single unit cell this might be *dynamic disorder* which is a real motion in the solid state; or it could be distributed among different unit cells as *static disorder* which means that the repeating unit determined by the computer was not in fact identical.[62] Both of these disorder types are treated in the same way during refinement, and so there is usually no need to determine the cause of the disordered crystal structure.[62,65]



Scheme 135: Two conformations of compound **34g** with rotation about the σ-bond to the thiophene substituent.

The thiophene ring in the crystal structure of product **34g** was disordered by rotation of 180° about the linking C—C bond, thus was refined with 50% occupancy of the sulfur atom in each location.

8.4.2 General crystal properties

All six samples were run as "well diffracting" which meant that the reflection patterns were strong compared to a poorly diffracting sample. Determination of a well-diffracting sample was a qualitative observation based on the diffraction patterns observed during the pre-experiment, and thus can provide high-resolution diffraction data.[56,63] This is most attributable to i) the uniform, close packing of the polar product molecules in the crystal lattice, and ii) the heavy sulfur atom which contains a larger electron cloud than the N, O and C atoms. Elements containing more electron shells have much greater atomic radii due to the scale in which the electron cloud increases; and therefore provide easily distinguishable, strong reflections from the diffraction of the X-ray beam.[55] Most samples were packed in a *monoclinic* pattern which has the second highest symmetry of the seven Bravais lattice types (**Figure 64**).[59] This meant that they are generally simple to identify from reflection data in comparison to lattice types with lower symmetry; however this is not taking into account the other geometric factors discussed in Chapter 1.[56,62]

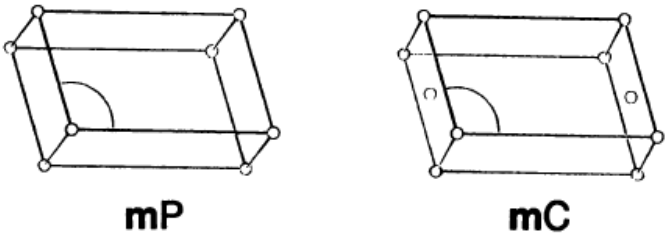


Fig 64. The two monoclinic space group classes, 1) mP (primitive) and 2) mC (C-centered).[62]

Interestingly, in all crystal structures of compounds **34** that were obtained (**34a**, **34b**, **34f**.DMSO, **34g**.DMSO, **34h**.2H_2O and **34l),** a single tautomeric form was apparent; where the hydrogen atom was situated on the thiatriazine ring nitrogen atom N4, rather than N2 or N7. A larger region of electron density was indicated during the structural refinement process with SHELX which may imply the presence of a proton at this position. The reason that the hydrogen atom was affixed at the N4 position during H-FIX refinement[67] was because the assignment of an NH group was required for publication purposes. An X-ray crystal structure is not considered "complete" until the hydrogen atoms have been placed in geometrically sensible positions. The refinement of hydrogen atoms has been outlined in Chapter 1.9.

8.5 Characterisation of pyrazolo[3,4-*e*][1,2,4]thiadiazines by X-ray crystallography

8.5.1 Structural elucidation and ellipsoid plots

As was the case with fused pyrazolo[1,5-*b*][1,2,4,6]thiatriazines **34**, the information obtained from mass spectrometry and NMR experiments was not sufficient to differentiate between the potential isomers from the condensation of 1-substittued 5-aminopyrazoles **33** and dichlorides **1**. The exact connectivity of atoms in the core of the

pyrazolo[3,4-*e*][1,2,4]thiatriazine ring system of dioxides **38** required clarification by X-ray crystallographic studies.

The most effective method of crystal growth for this system was **slow evaporation** from methanol and dichloromethane mixtures to afford single crystals for X-ray analysis. The **vapour diffusion** method provided crystals which were opaque or were too small (thin) in a single dimension, and the **cooling method** led to crystal clusters. X-ray crystal structures of representative compounds **38b** and **38j** are shown in **Figure 65**.

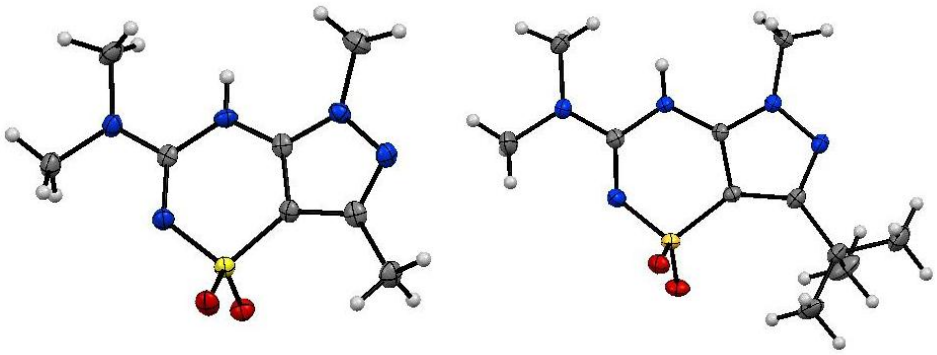


Fig 65. ORTEP diagrams of **38b** and **38j**.

Only two representatives of the unsubstituted pyrazolo[3,4-*e*][1,2,4]thiadiazine ring system were analysed by X-ray studies. Other structural assignments of compounds **38** were based on consistent ^{1}H and ^{13}C NMR chemical shifts and TLC mobility. The results of these condensations were accordant with other research on the dichloride chemistry whereby the amidinyl carbon was indicated to be the most electrophilic site[99] (to react with the NH_2 group); and carbon nucleophiles typically condensed with the sulfonyl group.[28] This evidence was sufficient to claim the formation of the single ring system; although some crystalline derivatives of the compounds **38** were also analysed by X-ray crystallography, thus proving the correct assignment of the precursor material.

8.5.2 General crystal properties

The two samples of **38b** and **38j** were defined by the detector as "well diffracting" with strong reflection patterns. The heavy sulfur atom of the SO_2 moiety allowed for rapid determination of the repeating lattice unit. The **38b** sample was packed in a *triclinic* pattern and the **38j** crystal in an *orthorhombic* cell (**Figure 66**). These required longer experimental collection times than crystals of the pyrazolo[1,5-*b*][1,2,4,6]thiatriazine dioxides **34**, although the data was collected with low R1 parameters (<0.5) to indicate satisfactory data.

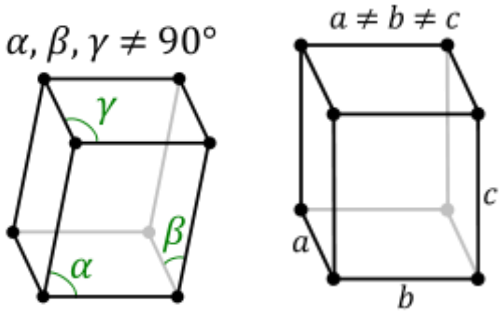


Fig 66. Unit cell representations of a triclinic cell (**38b**) on the left and an orthorhombic cell (**38j**) on the right.

Both crystal structures of products **38b** and **38j** were refined with the hydrogen atom attached to the thiadiazine ring at the N4 position rather than N2 or N6 atoms. A larger region of electron density was indicated during the structural refinement process, similar to the ring system **34** outlined in Chapter 2.5.2, with SHEL-X which indicated the presence of a proton at this position. Once more, the reason that the hydrogen atom was affixed at the N4 position during H-FIX refinement[67] was because the assignment of an NH group was necessary for publication.

8.6 Characterisation of alkyl substituted pyrazolo[3,4-*e*][1,2,4]thiadiazines by X-ray crystallography

Derivatives of products **38** which had been methyl substituted (**70-72**) as described in Chapter 5 were analysed by X-ray crystallography. An X-ray crystal structure of an example of each **70, 71**, and **72** became necessary given that 2D NOESY data was not informative (**Figure 67**). Benzylated compounds **70c-72d** were identified via consistent NMR trends, TLC mobility and solubility which were comparable to the methylated derivatives **70a-72b**.

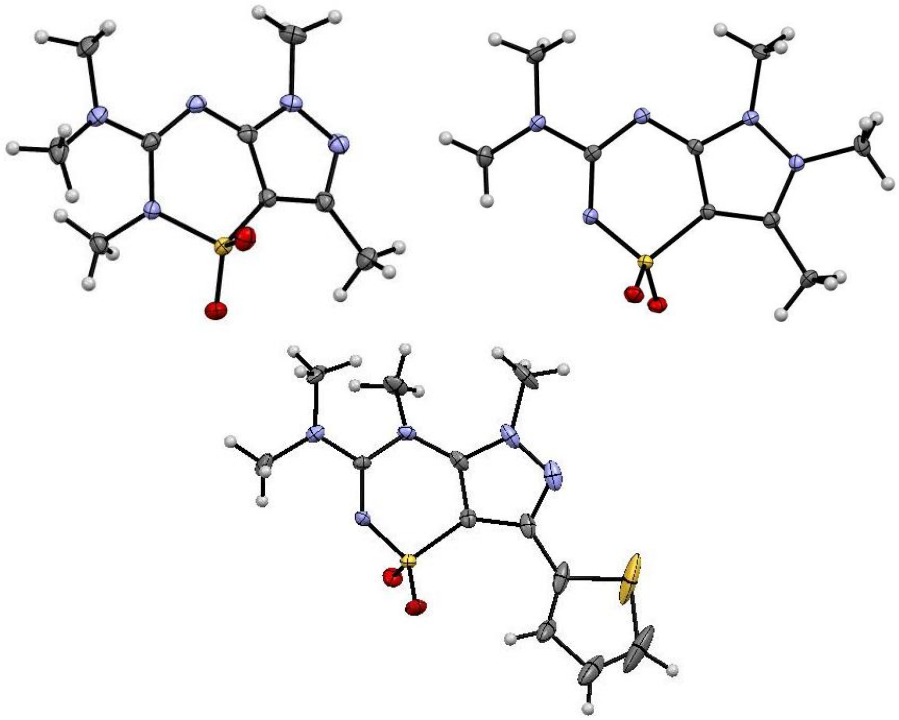


Fig 67. ORTEP diagrams of methylated derivatives **70a** (top left), **72a** (top right) and **71b** (bottom).

The X-ray crystal structure of **71b** presented minor disorder about the σ-bond between the thienyl group and the pyrazole ring. The R1 parameter (R1 = 0.0419) for this crystal structure lies well within acceptable parameters for publication, and further refinement was not necessary. It is important to note that the crystal diffraction patterns for these three compounds were collected at room temperature, and not 150°K, due to issues with the cryogenic device. This could have led to the higher instances of atomic vibrations or

rotations during sample collection (continuous disorder). It could also be a result of damage by X-ray radiation, although no damage to the crystal was indicated. Cryo-crystallography is usually carried out (and is especially important for protein crystallography) because of the potential for atomic vibrations and bond movement as well as radiation damage to the crystal from the X-rays.[62] These types of experiments had previously been carried out at sub-ambient temperature to minimise these possibilities, but it appeared that in fact the crystals from this body of work were not particularly sensitive to either of these problems.

A sample of compound **79c** which was alkylated on the thiadiazine ring nitrogen N4 was also analysed by X-ray diffraction (**Figure 68**). This was essential in determining the mode of connectivity of the ethyl acetate moiety from a choice of the N2 or N4 atoms, because the 2D NOESY data was not sufficient to confidently assign the structure.

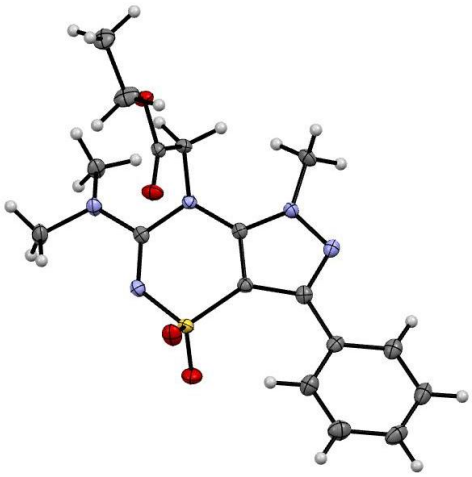


Fig 68. An ORTEP diagram of ethyl acetate derivative 7**9c**.

8.7 Characterisation of alkyl substituted pyrazolo[1,5-*b*][1,2,4,6]thiatriazines by X-ray crystallography

The X-ray experimental results of a series of alkyl substituted pyrazolothiatriazines **34** are provided below. **Table 21** in Section 8.10 summarises the experimental detail and provides access to CCDC deposition numbers and hence the full detail will not be described in the thesis. However, some general conclusions and refinement issues are outlined.

A derivative of product **34e** which had been substituted by ethyl bromoacetate (**74a**) as described in Chapter 5 was structurally elucidated by X-ray diffraction (**Figure 68**). A crystal structure of one example of an N4 alkylated product meant that (along with solid 2D NOESY data evidence) the structures of compounds **74** were less likely to be disputed.

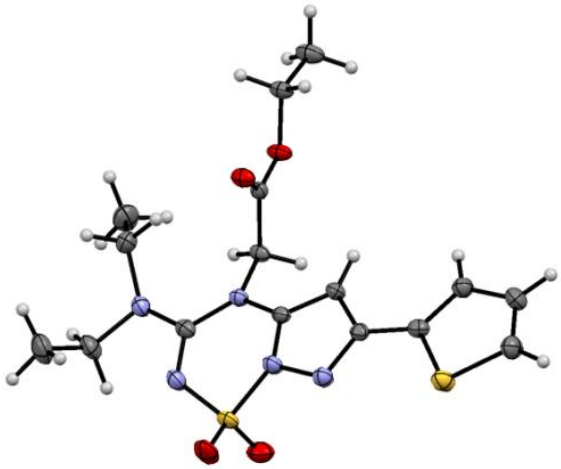


Fig 68. An ORTEP diagram of ethyl acetate derivative **74a**

The structure of the dimethylsulfanylidenederivative reported in Chapter 4 (compound **62a**, **Figure 69**) was the cause of much confusion, since the atoms flanking the sulfur atom were in fact deuterium (and not hydrogen). X-ray diffraction does not differentiate between deuterium and hydrogen because the electron density is the same for both.[64] Furthermore, hydrogen atoms were affixed in calculated positions after the refinement

process owing to the fact that they cannot be accurately located by current X-ray diffraction technology.[64]

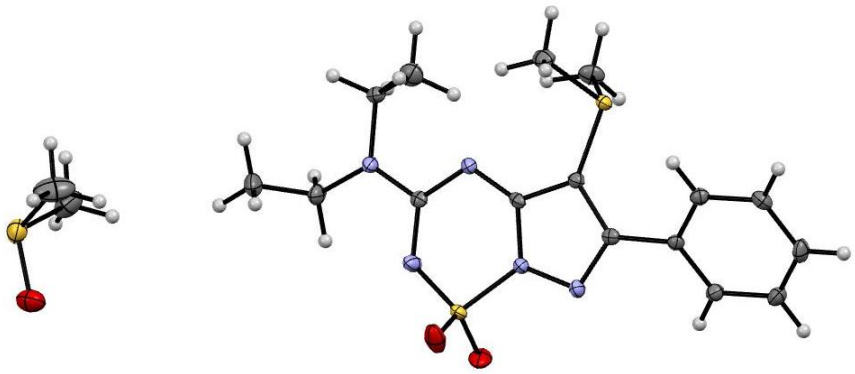


Fig 69. ORTEP diagram of **62a** which co-crystallised with a molecule of DMSO.

Additionally it should be noted that although the structure of such compounds has typically been represented in the form of the dimethyl sulfanylidene derivative with a neutral sulfur atom, an ylide form (such as in **Figure 71**) is most likely a more accurate representation due to the geometry depicted in the X-ray structure.

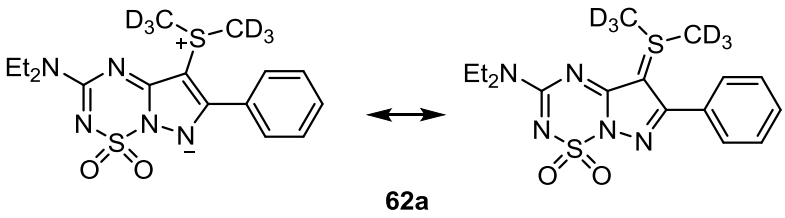


Fig 71. Some resonance contributors of compound **62a**.

It should be noted that resonant forms of the ylide with the negative charge on the other N atoms N2 or N4 would also exist are not shown. The presence of an ylide form is similar to the true composition of DMSO, which is best represented by the ylide structure but is often depicted with the sulfoxide group (S=O) for simplicity.

The crystal structure of pyridine adduct **55a** (**Figure 72**) was solved as the zwitterionic salt form as opposed to the neutral species shown in **Figure 73**.

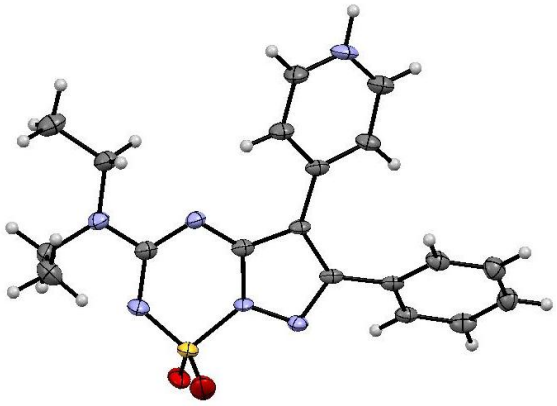


Fig 72. X-ray crystal structure of 5-(pyridin-4-yl)-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine **55a** in a zwitterionic salt form.

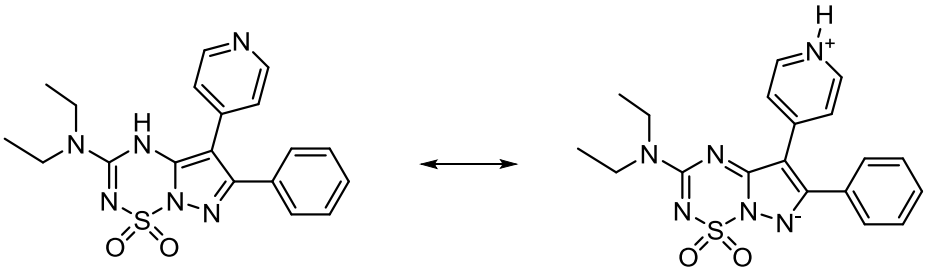


Fig 73. Resonance of the neutral pyridine species **55a** (left) to form the zwitterionic salt (right).

The crystal packing of compound **55a** was stabilised by an intermolecular N-H --- N hydrogen bond between the pyridine substituent and N7 of the pyrazole unit in an adjacent lattice (**Figure 74**). SHELX refinement suggested the H atom was present on the pyridine substituent and a negative charge present on the pyrazole nitrogen N7.

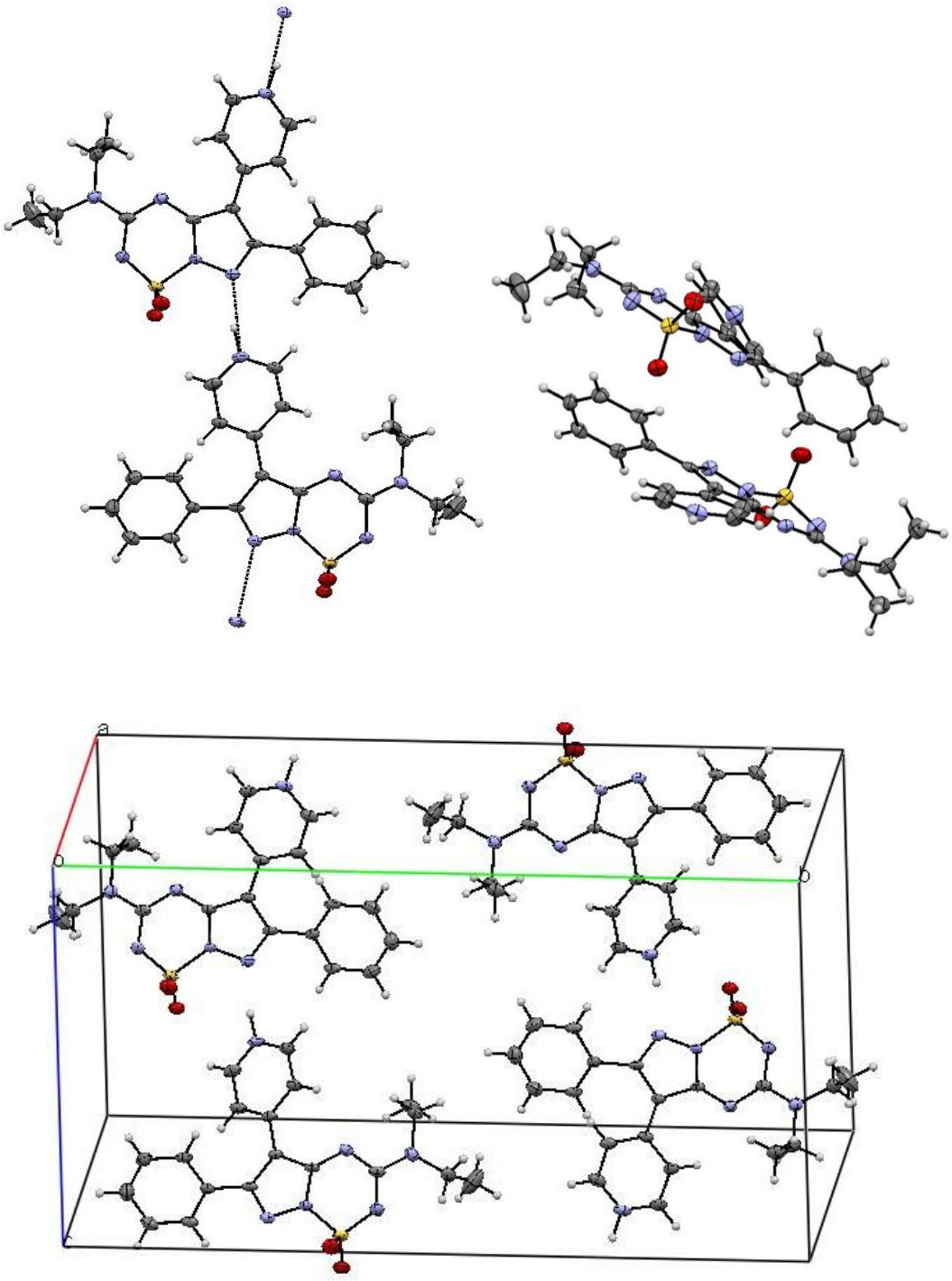


Fig 74. ORTEP diagrams of pyridine adduct **55a** generated in *Mercury -version 3.0* showing H-bonding between pyridine ring NH and N7 (pyrazole ring) of an adjacent molecule (upper left); packing within the crystal lattice (upper right, below).

The crystal structures of compounds **65c** and **66c** are shown in **Figure 75** below.

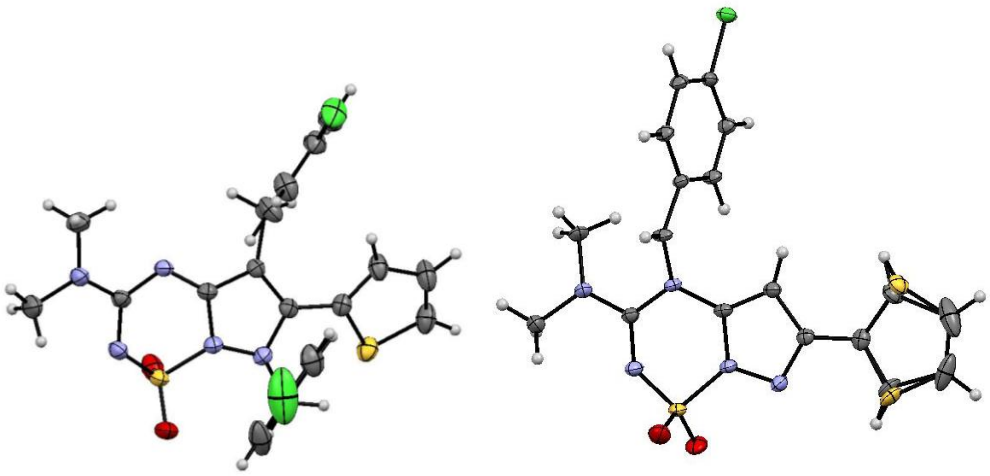


Fig 75. ORTEP diagrams of **65c** and **66c**. A dichloromethane molecule in the crystal lattice of **65c** has been omitted for clarity. The crystal structure of **66c** is disordered around the sigma bond of the thiophene moiety.

The crystal data of compounds **65c** and **66c** were obtained through longer acquisition times, which can be a consequence of a range of factors such as: crystal size and quality; the complexity of the crystal lattice; exposure time of the X-ray beam; and the inherent ability of the atoms to diffract the beam intensely. Compound **65c** contained multiple fragments of a dichloromethane molecule within each crystal lattice (**Figure 76**).

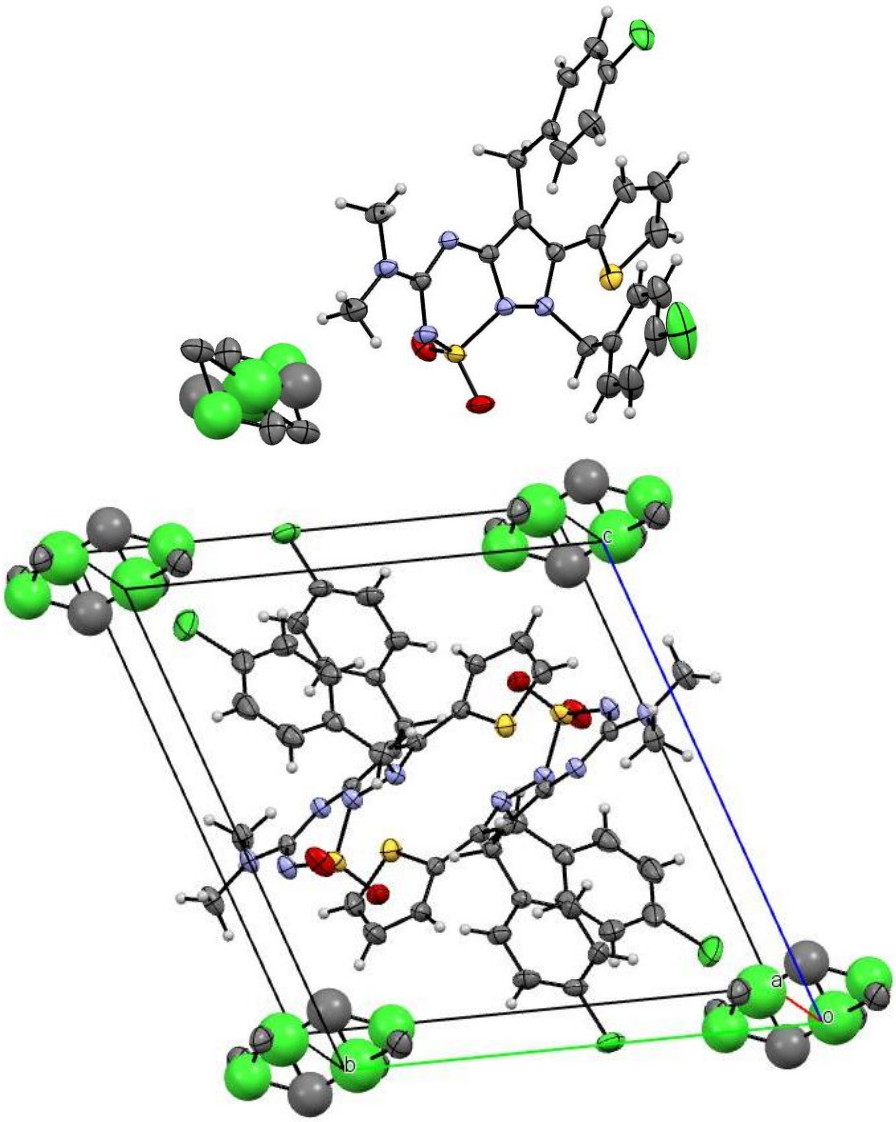


Fig 76. ORTEP diagrams of the repeating crystal lattice of **65a** (above) and packing of **65a** with partial molecules of suspected dichloromethane in the crystal lattice (below).

The approximate ratio of carbon to chlorine within the lattice was approximately 3.4:1 with a sum of C 2.35 Cl 0.7 in solvent molecules present within the lattice. It appeared that fragments of mostly carbon were present, and that the exact structure of a single solvent molecule could not be determined. Hydrogens were also not assigned to this molecule during the refinement process because, in this case, their locations were not apparent due to the small size and the disorder of the solvent molecules. The crystal data was not modified in any other way (such as the squeeze process) to remove the solvent molecules, and was reported with a redundancy restraint of R1=0.051.

The crystal structure of **66c** is disordered around the σ-bond between the thienyl group and the pyrazole ring in a 1:1 ratio, similar to structure **71b** as shown in **Figure 77**.

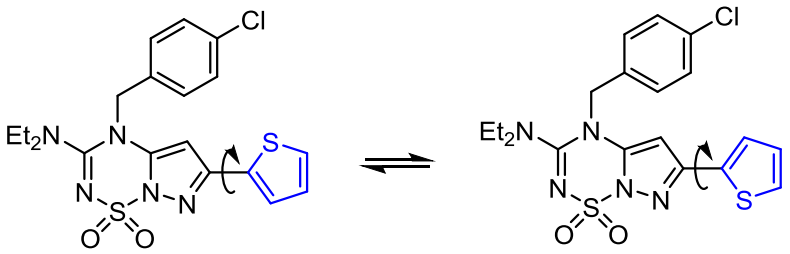


Fig 77. Two conformations of compound **66a** with rotation about the σ-bond to the thiophene substituent.

This meant that refinement could easily be carried out by assigning the sulfur atom at both orientations (with a partial occupancy or 0.5 occupancy in this case), to show that it inhabits half of each location. This gave a redundancy value of R1=0.038, well within acceptable limits.

8.8 Characterisation of bis-adduct 43a

The crystal structure of compound **43a** was obtained to determine the mode of connectivity of the pyrazole ring as either N1 or N2 (**Figure 78**).

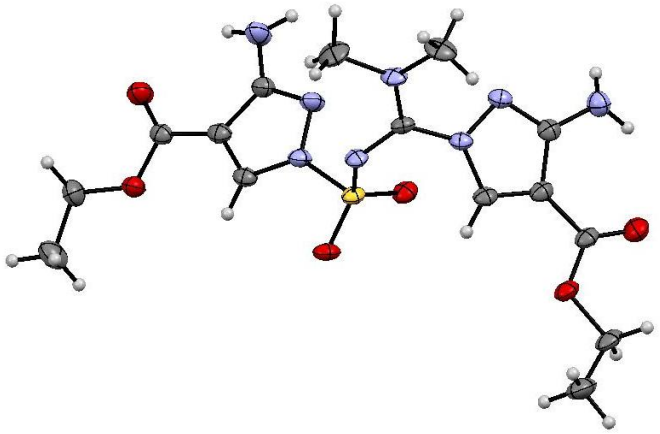


Fig 78. ORTEP diagram of **43a**.

A dichloromethane molecule was disordered over a spacial position at around 50% occupancy. This could not be adequately modelled and the 'SQUEEZE' routine of Platon was used to remove the electron density corresponding to this solvate.

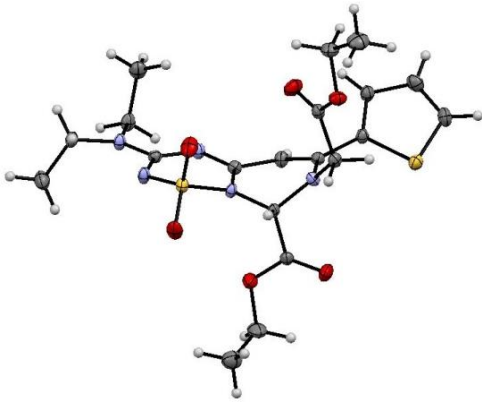
A significant number of compounds crystallize together with molecules of the solvent of crystallization. Solvent molecules often reside on a site with higher symmetry than their molecular symmetry, resulting in disorder.[65] The stacking can be disproportionate, resulting in points of inconsistent electron density in three dimensions.

Several approaches are available to take the contribution of disordered solvents into account.[56,62,65,213] The SQUEEZE technique is based on the concept that the total density shown in a correctly phased Fourier map can be divided into a portion that can be described and refined with discrete model parameters (i.e. positional and displacement parameters) and a portion corresponding to the disordered solvent.[213,214] The contribution to the calculated structure factors of the disordered solvent is taken into account by back-Fourier transformation of all density found in the disordered solvent area.[213] The procedure is highly automated and compatible with the widely used SHELXL refinement package.[213] A crystallographer must be able to justify the use of the SQUEEZE process, since essentially the technique removes the electron density from the raw data set and therefore corresponds to a reduced redundancy error associated with the crystal structure. In this case, the structure of the relevant moiety

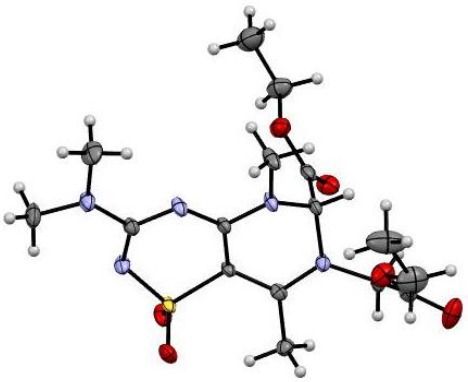
(the bis-adduct **43a**) was not in question, only the presence of an unidentifiable solvent, and so the use of the SQUEEZE function was defensible.

8.9 Characterisation of miscellaneous transformation products of the pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxides and pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides.

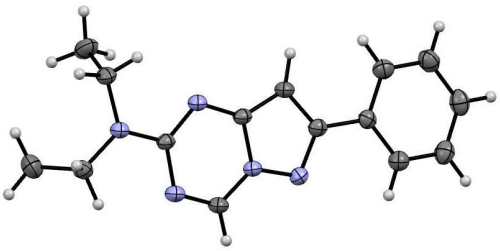
In many cases, the information gained from 1D and 2D NMR experiments and mass-spectral data was not enough to deduce the structures of these heteroatom-rich compounds, especially in cases where the product(s) formed were not the expected outcome. **Figure 79** below contains the ORTEP diagrams of: the 'ring expanded' fused pyrimidines **76a** and **77b**; a pyrazolo[1,5-*a*][1,2,4]triazine **83a**, the guanidine sulfonic acid **85**, the 'rearranged' pyrazolo[3,4-*e*][1,2,4]thiadiazine thermal product **89b** and the morpholine sulfamide **90f**. All of these structures were confirmed by unambiguous X-ray structures without complications such as disorder or erroneous solvent molecules, in a direct manner.



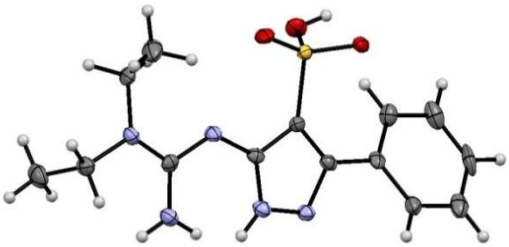
76a



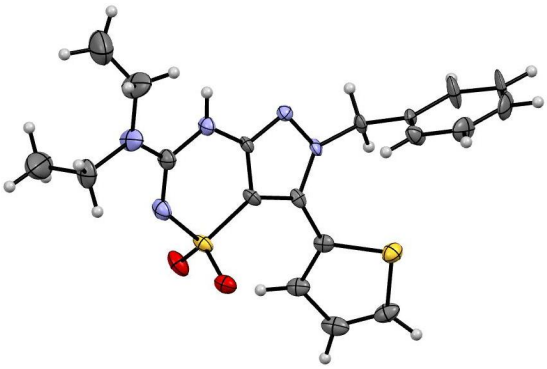
77b



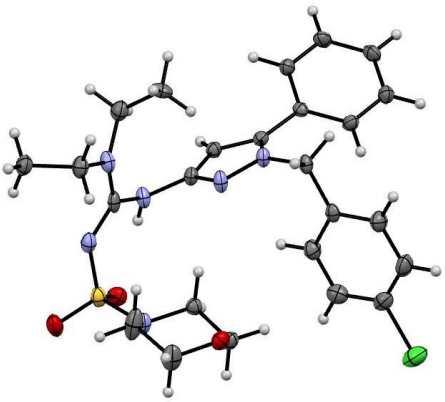
83a



85



89b



90f

Fig 79. ORTEP diagrams of crystal structures **76a**, **77b**, **83a**, **85**, **89b** and **90f**. A water molecule contained within the crystal lattice of **85** was removed for clarity. The structure shown of **89b** is one of five crystallographically independent molecules in the crystal lattice.

The crystal structure of Mannich adduct (**58a**) contained a foreign entity within the crystal lattice which was structurally independent of the compound shown in **Figure 80**. Speculation that a hydrochloride salt might be present could not be confirmed by refinement with a molecule of HCl in the lattice, as this did solve to within publishable parameters.

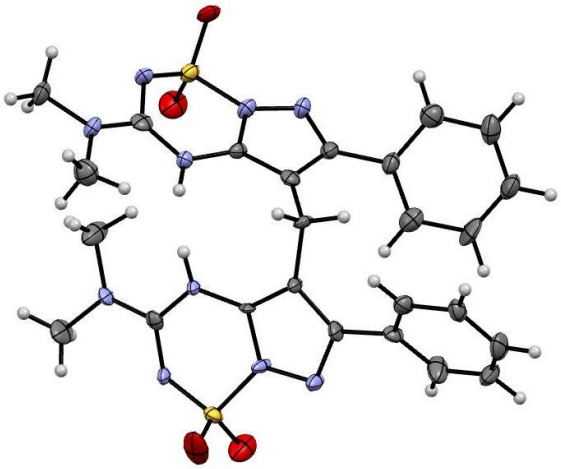


Fig 80. ORTEP diagram of methylene dimer **58a**. A foreign entity (potentially an HCl salt) present in the crystal structure of **58a** was removed for simplicity.

Many attempts were made to solve the structure of the crystal repeat unit with the presence of solvent, HCl, sodium atom, or fragments of another molecule from an adjacent lattice. The compound did not crystallise readily, and other efforts to grow adequate X-ray quality crystals were unsuccessful. Rather than use the SQUEEZE function on this structure to remove the undesired entity, this compound was left with the large pocket of electron density unaccounted for. The structure of the methylene dimer moiety was apparent from the X-ray result and also from mass spectrometry results, and so the structure has been included into the thesis as well as the CCDC website.

8.10 A summary of X-ray crystal diffraction experiments

A summary of results is shown in **Table 21** below including: crystal systems and space group settings; R1 values to give an indication of the data quality; and the CCDC numbers for reference.

Table 21: Crystallographic data and structure refinement for samples analysed by single crystal X-ray diffraction.

Sample name	Crystal System + Space Group Setting	R1	Reflections used	Temperature (°K)	CCDC number
34a	monoclinic 'P 1 21 1'	0.0402	7570	150	939838
34b	monoclinic 'P 1 21 1'	0.0412	3884	150	939839
34f	triclinic 'P - 1'	0.0433	4699	150	939840
34g	triclinic 'P - 1'	0.0766	5980	150	939841
34h	monoclinic 'p 1 21/n 1'	0.0372	6700	150	939842
34i	monoclinic 'C c'	0.0521	3198	150	939843
38b	triclinic 'P - 1'	0.0602	2092	150	1035053
38j	orthorhomic 'p b c a'	0.041	8046	148	1035054
70a	monoclinic 'p 21/n 1'	0.0382	4906	298	1035055
72a	monoclinic 'p 21/c 1'	0.035	4925	298	1035057

71b	orthorhomic 'P c 21 n'	0.0419	4127	298	1035056
79c	monoclinic 'P 1 21 1'	0.0797	3957	150	1035059
74a	triclinic 'P - 1'	0.0476	12967	150	1022794
65c	triclinic 'P - 1'	0.0512	13157	150	1022792
66c	triclinic 'P - 1'	0.0382	14237	150	1022793
76a	triclinic 'P - 1'	0.0332	11797	150	1022791
77c	triclinic 'P - 1'	0.077	7012	150	1035058
43a	triclinic 'P - 1'	0.0717	5339	150	939844
55a	monoclinic 'P 1 21/c 1'	0.0887	8650	150	939845
83a	triclinic 'P - 1'	0.0473	2985	150	*
85	orthorhomic 'P 21 21 21'	0.0455	1501	150	*
62a	orthorhomic 'P 21 21 21'	0.0422	4970	298	*
90f	triclinic 'P - 1'	0.0341	9464	123	*
89b	monoclinic 'P 1 21/c 1'	0.0873	11502	150	*
58a	monoclinic 'p 21/n 1'		7920	150	1042481

* The cif files of these structures have not yet been uploaded to the CCDC database. These will be uploaded for publication purposes. The crystal data is included in **Appendix D**.

Overall, X-ray crystallography has enabled the unequivocal structural elucidation of these unusual heterocyclic compounds, which typically did not provide informative NMR signals. With the current availability of X-ray diffraction technology it is now straightforward to characterise many of these heteroatom-rich compounds with little room for argument. Furthermore, X-ray crystallography provided information on some of the chemical interactions between the molecules in the crystalline state (such as key hydrogen bonding sites and packing arrangements) which could be related to the properties of these materials such as the high melting points and low solubility.

CHAPTER 9: EXPERIMENTAL

9.1 Methods and Materials

Reactions were carried out under an atmosphere of nitrogen. Solvents (dichloromethane, chloroform, acetonitrile, acetone, benzene, pyridine, methanol) and triethylamine were distilled over calcium hydride. THF was distilled from sodium benzophenone ketyl. DMF and DMSO were distilled under vacuum onto 3Å molecular sieves. Phosphoryl chloride was distilled over potassium carbonate. Samples of $CDCl_3$ were passed through activated aluminium oxide 70–290 mesh (Scharlau, activity degree 1, grain size 0.05–0.2 mm) to remove residual HCl and stored over silver foil/molecular sieves. Other reagents were used without further purification.

Analytical TLC was performed on Merck Kiesegel 60 F_{254} silica aluminium backed sheets, and was visualised under UV light and/or developed using ninhydrin dip. Column chromatography was performed on Merck Kiesegel (particle size: 0.04-0.063mm) silica gel.

^{1}H (δ_H) and ^{13}C (δ_C) NMR spectra were recorded on a Bruker Ultrashield 400 (400MHz and 100MHz respectively), a Bruker DRX500 (500MHz and 125MHz respectively), or a Bruker Ultrashield 600 (600MHz and 150MHz respectively). 400MHz nuclear Overhauser effect correlation spectroscopy (NOESY) two dimensional ^{1}H NMR experiments were performed on a Bruker Ultrashield 400. $CDCl_3$ and DMSO-d_6 were used as solvents and as an internal lock. Chemical shifts (δ) are measured in ppm. ^{1}H NMR chemical shifts were referenced to δ7.26 for $CDCl_3$ and δ2.50 for DMSO-d_6. ^{13}CNMR chemical shifts were referenced to δ77 for $CDCl_3$ and δ39.52 for DMSO-d_6. Spectral data were reported using the following format: (1) chemical shift (ppm), (2) integration, (3) multiplicity (s=singlet, d=doublet, t=triplet, q=quartet, dd=doublet of doublets, dq = doublet of quartets, m=multiplet, br=broad), (4) J-coupling constant (Hz), (5) assignment.

Melting points were determined using a Gallenkamp hotstage microscope and are uncorrected. X-Ray crystallography was performed at the Bragg Crystallography

Facility at The University of Adelaide on a Mo-target Oxford Diffraction X-Calibur X-ray diffractometer. Electron Impact (EI) mass spectrometry (70eV) was performed on a ThermoQuest MAT95XP mass spectrometer. Only the major fragments are given with their relative abundances shown in parentheses. Accurate mass measurements were taken with a resolution of 5000-10000 using PerFluoroKerosene (PFK) as an internal reference. High resolution mass spectrometry using electrospray ionization, positive ion with lockspray, was performed on a Waters Synapt HDMS instrument. Calibration: infusion of 0.5 mM sodium formate solution. Data acquisition: infusion of sample, *m/z* 527.1588 from the sodium attached ion of raffinose employed as a lock mass signal in positive ion mode and the proton abstracted ion of raffinose (*m/z* 503.1612) in the negative ion mode.

Experiments involving microwave irradiation were performed in a CEM Discover S-Class microwave reactor in reaction vessels (10 mL) loaded with combined starting materials in solvent (no more than 2 mL). The microwave reactor was operated in variable power (dynamic) mode with the following parameters: power 150 W, pressure maximum 20 bar, temperature 50 °C, stirring high, air/nitrogen cooling off. Ramp time to 50°C was 2 min. The sample was held at this temperature until the reaction reached completion, automatically modulating the power between 20–110W.

9.2 General synthesis procedure for 3-dialkylamino-4*H*-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxides 34a-l

Method A

A stirred mixture of 3-aminopyrazole **2** (2.5 mmol) and dichloro compound **1** (3.25 mmol) in DMPU (5 mL) was heated to 80^{o}C for 4-9 hrs. The reaction mixture was allowed to cool, diluted slowly with water (20 mL), stirred for several hours and the resulting precipitate was collected by filtration, washed with water and dried under vacuum. The mother liquor was allowed to stand overnight and any additional precipitate was treated as described above. If no precipitate formed, an extractive

workup was carried out with dichloromethane. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method B

A stirred solution of 3-aminopyrazole **2** (2.5 mmol) and dichloro compound **1** (3.3 mmol) in DMPU (5 mL) was warmed gently to dissolve the starting materials. The resulting solution was cooled with an ice/water bath and *N,N'*-diisopropylethylamine (6.5 mmol) was added dropwise. Stirring was continued overnight. The mixture was diluted slowly with water (20 mL), stirred for several hours and the resulting precipitate was collected, washed with hexane (5 mL) and water (5 mL) and dried under vacuum. The mother liquor was adjusted to pH 3 by dropwise addition of HCl (conc.), allowed to stand overnight and any additional precipitate was treated as described above. If no precipitate formed, an extractive workup was carried out with dichloromethane. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method C

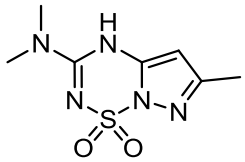
The dichloro compound **1** (3.3 mmol) in benzene (2 mL) was added to a stirred mixture of 3-aminopyrazole **2** (2.5 mmol), $KHCO_3$ (2 g), water (5 mL), $^nBu_4NHSO_4$ (0.1 g), and benzene (5 mL). The reaction mixture was warmed gently to dissolve the dichloro compound and stirred vigorously overnight at room temperature. The aqueous layer was removed. The precipitate and benzene layer were washed with 5% aq NaOH (2 x 10 mL) and water (2 x 10 mL) and the precipitate was dried under vacuum. The pH of the aqueous layer was adjusted to pH 3 by dropwise addition of HCl (2M) and allowed to stand overnight. Any additional precipitate was treated as above. If no precipitate formed, an extractive workup was carried out with dichloromethane. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method D

A stirred mixture of 3-aminopyrazole **2** (2.5 mmol), dichloro compound **1** (3.3 mmol), and dichloromethane (5 mL) was warmed gently to dissolve the starting materials and then cooled in an ice/water bath. Triethylamine (5 mmol) was added dropwise and the resulting mixture was stirred at room temperature overnight. The reaction mixture was diluted slowly with water (20 mL) and extracted with dichloromethane. The solvent was then evaporated then the crude product mixture was chromatographed over silica gel and the product was recrystallised.

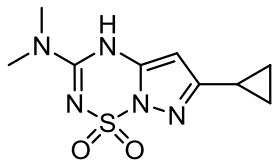
Compounds **34a-i** and **43a**, **43b**, **44**, **45**, **46** and **47** were synthesised originally during the author's Honours research and experimental data for these can be obtained from Paper X in the *Aust. J. Chem.* series.[99] The following compounds were also prepared by the aforementioned procedures.

3-Dimethylamino-6-methyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***34j***



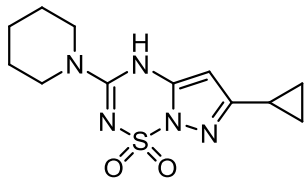
Method D. The crude brown solid was purified by column chromatography (10% MeOH in CH_2Cl_2) to provide the *title compound* as an off-white solid in 79% yield. Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave off-white needles, m.p. 218-220°C decomp. (Found: C 36.89, H 4.74, N 30.48; $[M+Na]^+$ 252.0504; $C_7H_{11}N_5O_2{}^{32}SNa$ requires C 36.67, H 4.84, N 30.55; $[M+Na]^+$ 252.0531). δH (400MHz, DMSO-d_6) 11.33 (1H, s, NH), 5.76 (1H, s, H5), 3.08 (6H, s, NCH_3 x 2), 2.19 (3H, br s, CH_3). δC (100MHz, DMSO-d_6) 151.6, 149.2, 140.9, 90.3, 37.7, 13.7**.**

6-Cyclopropyl-3-dimethylamino-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***34k***



Method D. After stirring at room temperature for 48 hrs, the reaction mixture was diluted with CH_2Cl_2 (5 mL), washed with dilute (1M) HCl (2 x 10 mL) and the aqueous layer was extracted with CH_2Cl_2 (2 x 10 mL). The solvent was evaporated and the brown crystalline residue was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* (0.60 g, 90%) as white wedges. Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless wedges, m.p. 244-245°C decomp. (Found: C 42.48, H 5.13, N 27.55; $[M+Na]^+$ 278.0680; $C_9H_{13}N_5O_2{}^{32}SNa$ requires C 42.34, H 5.13, N 27.43; $[M+Na]^+$ 278.0688). δH (600MHz, DMSO-d_6) 11.37 (1H, s, NH), 5.64 (1H, s, H5), 3.07 (6H, s, NCH_3 x 2), 1.90 (1H, m, C*H*(CH_2-CH_2), 0.92 (2H, ddd, *J* 8.4, 6.6, 4.1, CH(C*H*$_2$-C*H*$_2$)), 0.72 (2H, m, CH(C*H*$_2$-C*H*$_2$)). δC (150 MHz, DMSO-d_6) 157.9, 149.3, 140.9, 87.4, 37.8, 9.4, 8.2.

6-Cyclopropyl-3-(piperidin-1-yl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***34l***

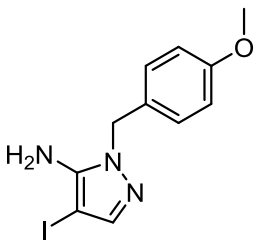


Method D (as for **34k** above). The crude black gum was recrystallised from CH_2Cl_2/MeOH to give yellow crystals which were washed with cold CH_2Cl_2 (1mL), then purified by column chromatography. Elution with 10% MeOH in CH_2Cl_2 gave the *title compound* (0.22 g, 30%) as white needles. Recrystallisation from CH_2Cl_2 gave white wedges, m.p. 198°C decomp. (Found: C 48.91, H 5.71, N 23.77; $[M+Na]^+$ 318.1001; $C_{12}H_{17}N_5O_2{}^{32}SNa$ requires C 48.80, H 5.80, N 23.71; $[M+Na]^+$ 318.1000). δH (600MHz, DMSO-d_6) 11.45 (1H, s, NH), 5.64 (1H, s, H5),

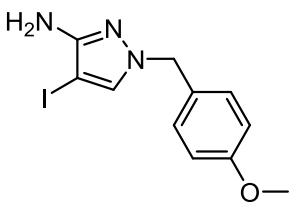
3.57 (4H, m, NCH_2 x 2), 1.91 (1H, m, C*H*(CH_2-CH_2)), 1.61 (2H, m-br, $NCH_2CH_2CH_2$), 1.57 (4H, m br, 2 x NCH_2CH_2), 0.92 (2H, m, CH(CH_2-CH_2)), 0.72 (2H, m, CH(CH_2-CH_2)). δC (150 MHz, DMSO-d_6) 158.0, 147.9, 140.8, 87.4, 46.1, 25.1, 23.4, 9.4, 8.2.

2-(4-Methoxybenzyl)-3-amino-4-iodo-pyrazole ***40b****, 1-(4-methoxybenzyl)-3-amino-4-iodo pyrazole* ***41b*** *and 2-(4-methoxybenzyl)-3[(4-methoxybenzyl)amino]-4-iodo-pyrazole* ***42***

A mixture of 4-iodo-3-aminopyrazole **32h** (0.52 g, 2.5 mmol), 4-methoxybenzyl chloride (0.42 g, 2.75 mmol), K_2CO_3 (0.38 g, 2.75 mmol), and acetonitrile (3 mL) was stirred at room temperature for 3 hrs. The reaction mixture was diluted slowly with water (5 mL) and extracted with dichloromethane (2 x 5 mL). The organic phase was evaporated and the crude product mixture was separated by column chromatography to give 3 compounds, in order of elution i) **40b** as a beige powder (0.33 g, 40%), **41b** as a tan solid (64 mg, 8%) and iii) **42** as a beige solid (0.17 g, 31%). Structural assignment of pyrazoles **40**-**42** were based on literature reports.[102]

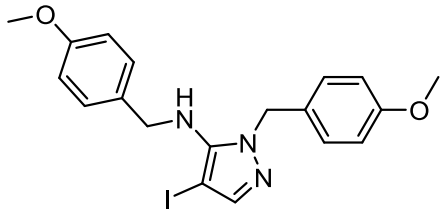


40b was obtained as a beige powder. δH (400MHz, $CDCl_3$) 7.20 (2H, d, *J* 8.6, ArH), 7.17 (1H, s, H5), 6.82 (2H, d, *J* 8.6, ArH), 4.25 (2H, s, CH_2Ar), 3.78 (3H, s, OCH_3). δC (100 MHz, $CDCl_3$) 158.7, 130.3, 129.6, 113.7, 55.3, 54.0, 50.3.



41b was obtained as a tan solid. δH (400MHz, $CDCl_3$) 7.31 (1H, s, H5), 7.12 (2H, d, *J* 8.6, ArH), 6.86 (2H, d, *J* 8.6, ArH), 5.17 (2H, s, CH_2Ar), 3.78 (3H, s, OCH_3). δC (100

MHz, $CDCl_3$) 159.5, 145.2, 141.9, 128.5, 127.7, 114.5, 55.3, 52.6, 43.0.



42 was obtained as a beige solid. δH (400MHz, $CDCl_3$) 7.26 (2H, d, J 8.5, ArH), 7.22 (1H, s, H5), 7.13 (2H, d, *J* 8.5, ArH), 6.84 (2H, d, *J* 8.6, ArH), 6.79 (2H, d, *J* 8.6, ArH), 5.57 (1H, s, NH), 4.35 (2H, s, CH_2Ar), 4.33 (2H, s, CH_2Ar), 3.76 (3H, s, OCH_3), 3.75 (3H, s, OCH_3). δC (100 MHz, $CDCl_3$) 158.7, 130.3, 129.6, 113.7, 55.3, 54.0, 50.26.

9.3 General synthesis procedure for 3-dialkylamino-4*H*-pyrazolo[3,4-*e*][1,2,4]thiadiazine 1,1-dioxides 38a-i

Method A

A stirred mixture of 1-substituted 5-aminopyrazole **2** (2.5 mmol) and dichloro compound **1** (3.25 mmol) in DMPU (5 mL) was heated to 80^{o}C for 9-18 hrs. The reaction mixture was allowed to cool, diluted slowly with water (20 mL), stirred for several hours and the resulting precipitate was collected by filtration under vacuum. The collected solid was washed with water and dried under vacuum. The mother liquor was allowed to stand overnight and any additional precipitate was treated as described above. If no precipitate formed, an extractive workup was carried out with dichloromethane. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method B

A stirred mixture of 1-substituted 5-aminopyrazole **2** (2.5 mmol), dichloro compound **1** (3.3 mmol), and DMPU (5 mL) was warmed gently to dissolve the starting materials.

The stirred solution was cooled with an ice/water bath and *N,N'*-diisopropylethylamine (6.5 mmol) was added dropwise. Stirring was continued overnight or until completion by TLC. The mixture was diluted slowly with 1M HCl (20 mL), stirred for several hours and the resulting precipitate was collected by filtration, washed with hexane (5 mL) and then water (5 mL) and dried under vacuum. The mother liquor was adjusted to pH 3 by dropwise addition of HCl (conc.), allowed to stand overnight and any additional precipitate was treated as described above. If no precipitate formed, an extractive workup was carried out with dichloromethane. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method C

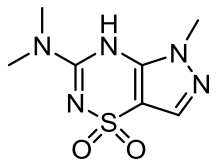
A stirred mixture of 1-substituted 5-aminopyrazole **2** (2.5 mmol), dichloro compound **1** (3.3 mmol), and CH_2Cl_2 (5 mL) was warmed gently to dissolve the starting materials and then cooled in an ice/water bath. Triethylamine (5 mmol) was added dropwise and the resulting mixture was stirred at room temperature until conversion was no longer observed by TLC. The reaction mixture was diluted slowly with water (20 mL) and extracted with CH_2Cl_2. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

Method D

A stirred mixture of 1-substituted 5-aminopyrazole **2** (2.5 mmol), dichloro compound **1** (3.3 mmol), and acetonitrile (5 mL) was warmed gently to dissolve the starting materials and then cooled in an ice/water bath. Triethylamine (5 mmol) was added dropwise and the resulting mixture was stirred at room temperature or 50^{o}C until conversion was no longer observed by TLC. The reaction mixture was diluted slowly with water (20 mL) and extracted with CH_2Cl_2. The crude product mixture was chromatographed over silica gel and the product was recrystallised.

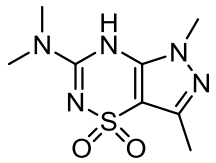
The following compounds were prepared by the above procedures:

3-Dimethylamino-5-methyl-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38a***



Method C. Elution with 20% MeOH in CH_2Cl_2 provided the *title compound* as a white solid in 78% yield. R_F 0.35 (20%MeOH:CH_2Cl_2). Recrystallisation of **38a** from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 235 °C decomp. (Found $[M+H]^+$ 230.0714; $C_7H_{12}N_5O_2{}^{32}S$ requires $[M+H]^+$ 230.0712). δH (400MHz, DMSO-d_6) 10.56 (1H, s, NH), 7.78 (1H, s, H7), 3.84 (3H, s, N5-CH_3), 3.09 (6H, s, NCH_3 x 2). δC (100MHz, DMSO-d_6, 36°C) 149.0, 139.3, 131.4, 103.6, 38.0, 35.9, 35.9.

5,7-Dimethyl-3-(dimethylamino)-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38b***

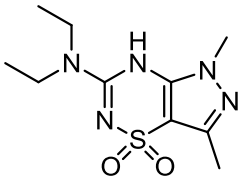


Method A was performed with extractive workup using CH_2Cl_2 and washing with 5% aq. NaOH solution to afford the *title compound* as a white powder in 61% yield.

Method B followed by elution with 10% MeOH in CH_2Cl_2 provided the *title compound* as white flakes in 71% yield. R_F 0.31 (10% MeOH:CH_2Cl_2).

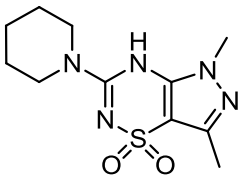
Recrystallisation of **38b** from 1:1 MeOH:CH_2Cl_2 gave white needles, m.p. 224-226°C decomp. (Found $M^{+\bullet}$ 243.0784; $C_8H_{13}N_5O_2{}^{32}S$ requires $M^{+\bullet}$ 243.0796). δH (600MHz, DMSO-d_6) 10.46 (1H, br s, NH), 3.74 (3H, s, N5-CH_3), 3.08 (6H, s, NCH_3 x 2), 2.20 (3H, s, C7-CH_3). δC (150MHz, DMSO-d_6) 149.7, 141.4, 140.4, 102.0, 38.4, 35.9, 12.8. EIMS m/z (%) 271.1 (19.63) 244.1 (13.08) 243.0 (100) 199.0 (8.10).

3-(Diethylamino)-5,7-dimethyl-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38c***



Method D was employed at 50°C. Elution with 10%EtOH in CH_2Cl_2 afforded the *title compound* as colourless blocks in 13% yield, along with 37% recovery of the starting material. R_F 0.48 (10%EtOH:CH_2Cl_2). Recrystallisation of **38c** from CH_2Cl_2 gave colourless prisms, m.p. 168-170 °C decomp. (Found $[M+Na]^+$ 294.1001. $C_{10}H_{17}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 294.1001). δH (400MHz, DMSO-d_6, 36°C) 10.18 (1H, s, NH), 3.75 (3H, s, NCH_3), 3.53 (4H, q, *J* 7.1Hz, NCH_2CH_3 x 2), 2.23 (3H, s, CH_3), 1.16 (3H, t, *J* 7.1Hz, NCH_2CH_3 x 2). δC (100MHz, DMSO-d_6, 56°C:) 147.7, 140.5, 139.6, 102.0, 42.6, 35.6, 13.0, 12.5.

5,7-Dimethyl-3-(piperidin-1-yl)-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38d***



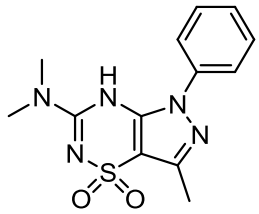
Method B was employed with the addition of 10% aq. HCl during workup to afford the *title compound* as a colourless gum in 15% yield.

Method C followed by elution with 5% MeOH in CH_2Cl_2 provided the *title compound* as a beige powder in 36% yield. R_F 0.30 (5% MeOH: CH_2Cl_2).

Recrystallisation of **38d** from CH_2Cl_2 gave colourless blocks, m.p. 272-274 °C decomp. (Found $[M+Na]^+$ 306.0996; $C_{11}H_{17}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 306.1001). δH (600MHz, DMSO-d_6) 10.73 (1H, s, NH), 3.73 (3H, s, N5-CH_3), 3.55 (4H, m, NCH_2 x

2), 2.21 (3H, s, C7-CH_3), 1.61 (2H, m, CH_2), 1.56 (4H, m, CH_2 x 2). δC (100MHz, DMSO-d_6) 148.1, 140.6, 139.7, 102.0, 46.8, 35.6, 25.2, 23.7, 12.5.

3-Dimethylamino-7-methyl-5-phenyl-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38e***

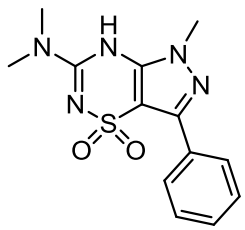


Method B gave the *title compound* as a white powder in 14% yield.

Method C gave the *title compound* as a beige powder in 42% yield.

Recrystallisation of **38e** from 1:1 MeOH:CH_2Cl_2 gave white needles m.p. 298°C decomp. (Found $[M+Na]^+$ 328.0849; $C_{13}H_{15}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 328.0844). δH (400MHz, DMSO-d_6) 10.78 (1H, s, NH), 7.69 (2H, d, *J* 7.1, ArH), 7.57 (2H, t, *J* 7.6, ArH), 7.44 (1H, t, *J* 7.2, ArH), 3.06 (6H, s, NCH_3 x 2), 2.34 (3H, s, C7-CH_3). δC (100MHz, DMSO-d_6, 70°C:) 149.7, 142.5, 137.6, 129.4, 129.2, 127.4, 122.7, 103.3, 37.6, 12.2.

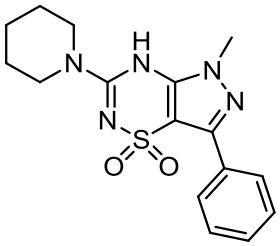
3-Dimethylamino-5-methyl-7-phenyl-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38f***



Method B provided the *title compound* as a white solid in 44% yield.

Recrystallisation of **38f** from 1:1 MeOH:CH_2Cl_2 gave white needles m.p. >320°C. (Found $[M+Na]^+$ 328.0849; $C_{13}H_{15}N_5O_2{}^{32}SNa$ requires 328.0844). δH (400MHz, DMSO-d_6) 10.55 (1H, s, NH), 8.05 (2H, d, *J* 7.2, ArH x 2), 7.46 (2H, t, *J* 7.2, ArH x 2), 7.39 (1H, d, *J* 7.2, ArH), 3.90 (3H, s, CH_3), 3.12 (6H, s, NCH_3 x 2). δC (100MHz, DMSO-d_6, 56°C) 148.5, 143.2, 140.6, 131.1, 128.6, 128.5, 126.8, 100.9, 38.0, 36.2.

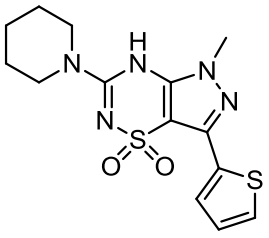
5-Methyl-7-phenyl-3-(Piperidin-1-yl)-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38g***



Method D was employed at 50°C. Elution with 40% EtOAc in CH_2Cl_2 afforded the *title compound* as a white solid in 49% yield. R_F 0.30 (40%EtOAc:CH_2Cl_2).

Recrystallisation of **38g** from 1:1 MeOH:CH_2Cl_2 gave colourless prisms, m.p. 191 °C decomp. (Found $[M+Na]^+$ 368.1154; $C_{16}H_{19}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 368.1157). δH (400MHz, DMSO-d_6) 10.83 (1H, s, NH), 8.04 (2H, d, *J* 7.2Hz, ArH x 2), 7.46 (2H, t, *J* 7.2Hz, ArH x 2), 7.39 (1H, t, *J* 7.2Hz, ArH), 3.89 (3H, s, NCH_3), 3.61 (4H, m, NCH_2 x 2), 1.62 (6H, m, CH_2 x 3). δC (150MHz, DMSO-d_6, 56°C) 147.7, 143.2, 140.7, 131.2, 128.7, 128.6, 126.9, 101.1, 46.8, 36.2, 25.3, 23.7.

3-Dimethylamino-5-methyl-7-(thien-2-yl)-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38h***

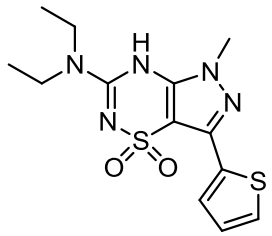


Method B provided *the title compound* as a white powder in 44% yield.

Method D followed by elution with 20% MeOH in CH_2Cl_2 provided the *title compound* as a beige powder in 70% yield. R_F 0.34 (20%MeOH:CH_2Cl_2).

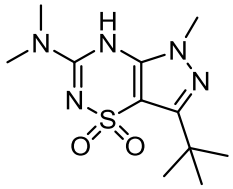
Recrystallisation of **38h** from 1:1 MeOH:CH_2Cl_2 gave white wedges, m.p. 295°C decomp. (Found $[M+Na]^+$ 334.0418; $C_{11}H_{13}N_5O_2{}^{32}S_2Na$ requires $[M+Na]^+$ 334.0408). δH (400MHz, DMSO-d_6) 10.60 (1H, s, NH), 7.79 (1H, d, *J* 3.6, 1.1, thienyl H5), 7.56 (1H, d, *J* 5.0, 1.1, thienyl H3), 7.15 (1H, dd, *J* 5.0, 3.7, thienyl H4), 3.86 (3H, s, N5-CH_3), 3.12 (6H, s, NCH_3 x 2). δC (100MHz, DMSO-d_6) 149.7, 142.0, 139.0, 133.9, 128.3, 128.1, 126.9, 99.9, 38.4, 36.4.

3-(Diethylamino)-5-methyl-7-(thien-2-yl)-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***38i***



Method D was employed at 50°C. Elution with 10% MeOH in CH_2Cl_2 afforded the *title compound* as white prisms in 11% yield. R_F 0.46 (10%MeOH:CH_2Cl_2). Recrystallisation of **38i** from CH_2Cl_2 gave colourless blocks, m.p. 248 °C dec. (Found $[M+Na]^+$ 362.0724; $C_{13}H_{17}N_5O_2{}^{32}S_2Na$ requires $[M+Na]^+$ 362.0721). δH (400MHz, DMSO-d_6, 36°C) 10.39 (1H, s, NH), 7.79 (1H, dd, *J* 3.5, 0.9, thienyl-H5), 7.57 (1H, d, *J* 4.9, 0.9, thienyl-H3), 7.16(1H, dd, *J* 4.9, 3.5, thienyl-H4), 3.86 (3H, s, NCH_3), 3.55 (4H, q, *J* 7.2, NCH_2CH_3 x 2), 1.19 (6H, t, *J* 7.2, NCH_2CH_3 x 2). δC (150MHz, DMSO-d_6, 56 °C) 147.4, 140.7, 138.4, 133.2, 127.5, 127.4, 126.2, 100.0, 42.4, 35.8, 12.7.

*3-Dimethylamino)-5-methyl-7-*tert*-butyl-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***38j***

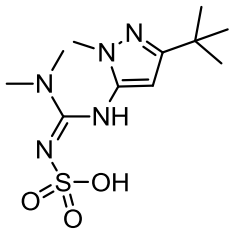


Method B followed by extraction with EtOAc, washing with saturated aqueous NaCl provided the *title compound* as white wedges in 10% yield.

Recrystallisation of **38j** from 1:1 MeOH:CH_2Cl_2 gave off-white prisms m.p. 276^{o}C decomp. (Found $[M+Na]^+$ 308.1157; $C_{11}H_{19}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 308.1157). δH (400MHz, DMSO-d_6) 10.4 (1H, s, NH), 3.75 (3H, s, N5-CH_3), 3.08 (6H, s, NCH_3 x 2), 1.34 (9H, s, $C(CH_3)_3$). δC (100MHz, DMSO-d_6) 153.1, 148.9, 140.1, 100.9, 38.0, 35.6, 33.0, 29.1.

*3-Dimethylamino)-5-methyl-7-*tert*-butyl-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***38j*** *and [(3-*tert*-butyl-1-methyl-1H-pyrazol-5-ylamino)-dimethylamino-methylene]-sulfamic acid* ***51***

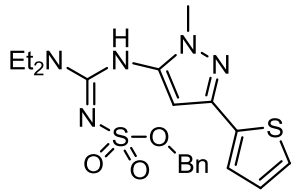
Triethylamine (0.9 mL, 6.5 mmol) was added to a stirred, ice-cooled mixture of 5-amino-3-*tert*-butyl-1-methyl pyrazole (0.38 g, 2.5 mmol), dichloride **1a** (0.67 g, 3.3 mmol), and acetonitrile (3 mL) and the resulting mixture was stirred at room temperature for 18 hrs. The mixture was then diluted with CH_2Cl_2 (10 mL), washed with water (2 x 5 mL), then dried and evaporated to a brown oil. The products were separated via column chromatography to afford compound **34j** (0.233g, 33%) R_F 0.33 (20%MeOH:CH_2Cl_2) and **51** (0.105g, 13%) R_F 0.25 (60%EtOAc: CH_2Cl_2).



Recrystallisation of **51** from CH_2Cl_2 gave white needles, m.p. 147-148 °C. (Found $[M+Na]^+$ 312.1924**;** $C_{14}H_{23}N_7Na$ requires $[M+Na]^+$ 312.1913).* δH (400MHz, DMSO-

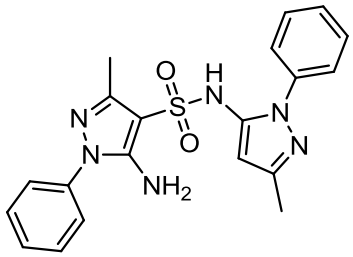
d_6) 8.31 (1H, s, NH); 6.30 (1H, s, pyrazole CH); 3.63 (3H, s, py-NCH_3); 3.17 (3H, s, NCH_3); 3.13 (3H, s, NCH_3); 1.22 (9H, s, $C(CH_3)_3$). δC (100MHz, DMSO-d_6) 161.0, 158.1, 157.7, 148.1, 91.9, 36.3, 35.6, 33.6, 31.8, 30.5. *Mass spectral data does not match the structure; hence obtaining the benzyl derivative **52**.

*Diethylamino-[2-methyl-5-(thien-2-yl)-2H-pyrazol-3-ylamino)-methylene]sulfamic acid benzyl ester **52***



N,N'-diisopropylethylamine (1.13 mL, 6.5 mmol) was added slowly to a stirred mixture of 1-methyl-3-(thien-2-yl)-5-aminopyrazole (0.45 g, 2.5 mmol) and dichloride **1b** (0.75 g, 3.3 mmol) in DMPU (3 mL) at 5°C. The resulting solution was then stirred at room temperature for 15 hrs, then treated with concentrated HCl (5 mL), followed by 1M aqueous HCl (10 mL) and stirred for a further 20 minutes to afford a precipitate. The solid was collected via vacuum filtration and washed with water (5 mL) to afford **38i** (71 mg, 9%). The filtrate was was extracted with CH_2Cl_2 (2 x 10 mL) and the organic phase was dried and concentrated to a brown oil. The crude material was used for the next step. A mixture of crude compound, K_2CO_3 (0.315 g, 2.30 mmol), benzyl bromide (0.388 g, 2.28 mmol) and $^nBu_4NBr$ (74 mg, 0.23 mmol) in acetonitrile (3 mL) was stirred at room temperature for 18 hrs until TLC indicated no further reaction. The mixture was then treated with water (5 mL) and extracted with CH_2Cl_2 (5 mL). The organic phase was dried and concentrated to a brown oil which was purified by column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* (0.129 g, 12%) as a colourless oil. (Found $[M+Na]^+$ 470.1295; $C_{20}H_{25}N_5O_3{}^{32}S_2Na$ requires $[M+Na]^+$ 470.1297). δH (600MHz, $CDCl_3$) 8.18 (1H, s, NH), 7.41 (1H, dd, *J* 5.0, 1.0, thienyl H5), 7.32-7.27 (3H, m, ArH), 7.08 (2H, d, *J* 6.9, ArH), 7.07 (1H, dd, *J* 5.0, 3.6, thienyl H3), 7.04 (1H, dd, *J* 3.6, 1.0, thienyl H4), 6.13 (1H, s, pyrazole CH), 5.34 (2H, s, CH_2), 3.84 (3H, s, NCH_3), 3.36 (4H, q, *J* 7.1, NCH_2CH_3 x 2), 1.10 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (150MHz, $CDCl_3$) 156.8, 147.2, 138.5, 136.9, 130.0, 128.9, 128.0, 127.9, 127.6, 127.2, 126.9, 100.1, 56.3, 53.5, 43.5, 12.7.

5-Amino-3-methyl-N-(3-methyl-1-phenyl-1H-pyrazol-5-yl)-1-phenyl-1H-pyrazole-4-sulfonamide ***53***

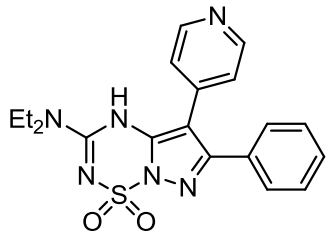


A stirred mixture of 1-phenyl 3-methyl 5-aminopyrazole (0.433 g, 2.5 mmol), dichloro compound **1a** (0.69 g, 3.5 mmol), triethylamine (0.1 mL, 6.7 mmol), and DMPU (3 mL) was heated to 100^{o}C overnight. The mixture was cooled to room temperature and water (10 mL) and 2M aqueous HCl (2 x 5 mL) were added. The aqueous mixture was extracted with ethyl acetate (3 x 15 mL). The extracts were combined and washed with saturated NaCl (aq) (2 x 15 mL) then dried and evaporated to give a yellow oil. The remaining product was obtained upon distillation of the DMPU under vacuum to give a brown oil. The crude oils were combined and purified via column chromatography (30% EtOAc in CH_2Cl_2) to give the *title compound* (0.113 g, 22%).

Recrystallisation of **53** from 1:1 MeOH:CH_2Cl_2 gave white needles m.p. 208-209^{o}C decomp. (Found $[M+Na]^+$ 431.1266; $C_{20}H_{20}N_6O_2{}^{32}SNa$ requires $[M+Na]^+$ 431.1266). δH (400MHz, DMSO-d_6): 9.98 (1H, s, NH), 7.55-7.32 (10H, m, Ph H x 2), 5.98 (1H, s, pyrazole CH), 5.78 (2H, s, NH_2), 3.33 (6H, s, NCH_3 x 2), 2.18 (3H, s, CH_3), 1.98 (3H, s, CH_3). δC (100MHz, DMSO-d_6): 147.8, 147.5, 146.6, 138.4, 137.5, 134.8, 129.4, 128.7, 127.6, 127.1, 123.9, 123.8, 103.8, 98.1, 13.8, 12.7.

9.4 Synthesis of 5-(pyridin-4-yl)- and 5-(pyridazin-4-yl)-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine adducts

3-(Diethylamino)-6-phenyl-5-(pyridin-4-yl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***55a***



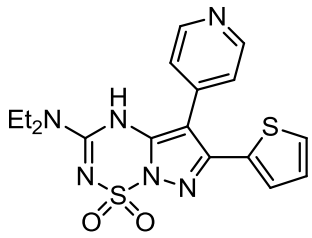
Using benzoyl chloride: Benzoyl chloride (1.2 mL, 1.0 mmol) was added dropwise to a stirred solution of **34e** (0.160 g, 0.5 mmol) in pyridine (5 mL). The solution was stirred overnight at room temperature and then water (20 mL) was added and the mixture extracted with CH_2Cl_2 (2 x 20 mL). Extracts were combined, dried, and evaporated to give a red oil which was purified via column chromatography (10% MeOH in CH_2Cl_2) to yield the *title compound* (0.10 g, 51%) as a yellow solid and recovered starting material **34e** (17%). R_F 0.15 (20%MeOH: CH_2Cl_2).

Using acetic anhydride: A mixture of pyrazolothiatriazine **34e** (0.160 g, 0.5 mmol), acetic anhydride (1.5 mL) and pyridine (4 drops) was heated at 90^{o}C for 3 hrs. More pyridine (0.2 mL) was added and the heating was continued overnight at 90^{o}C. The mixture was cooled to room temperature then water (5 mL), and ether (5 mL) were added and the resulting precipitate collected by vacuum filtration. The yellow solid was purified by column chromatography as above to yield the *title product* (70 mg, 35%) and recovered starting material **34e** (41%).

Recrystallisation of **55a** from 1:1 MeOH:CH_2Cl_2 gave bright yellow needles, m.p. 228.5^{o}C decomp. (Found: C 57.30, H 5.11, N 21.01; $M^{+\bullet}$ 396.1363; $C_{19}H_{20}N_6O_2S$ requires C 57.56, 5.08, 21.20; $M^{+\bullet}$ 396.1358). δH (600MHz, DMSO-d^6) 8.40 (2H, br d, pyridine-H2,H6), 7.87 (2H, d, *J* 5.6, pyridine-H3,H5), 7.50-7.46 (5H, m, ArH), 3.60 (2H, br-m, NCH_2), 3.47 (2H, br-m, NCH_2), 1.17 (6H, br m, 2 x CH_3). δC (100MHz,

DMSO-d^6) 156.26, 154.12, 152.11, 139.94, 133.96, 128.95, 128.87, 128.76, 120.16, 97.78, 41.81, 13.46. *m/z* (EI) 396 (100%, $M^{+\bullet}$), 262 (26), 205 (17), 179 (37), 99 (26).

3-(Diethylamino)-5-(pyridin-4-yl)-6-(thien-2-yl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***55b***



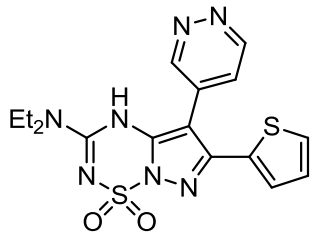
Using benzoyl chloride: A stirred mixture of **34h** (0.16 g, 0.5 mmol) in acetonitrile (2 mL) was cooled to -30°C. Pyridine (0.1 mL, 1.0 mmol) was added, followed by benzoyl chloride (0.1 mL, 1.0 mmol) and the resulting mixture was stirred at -30°C for 20 minutes. The reaction was left to warm to room temperature and stirred for 24 hrs. 2M HCl (5mL) was added and the resulting mixture was extracted with CH_2Cl_2 (5 mL). The organic layer was washed with 2M HCl (2 x 5 mL) then dried over Na_2SO_4 and evaporated at reduced pressure and dried under vacuum. The residual yellow solid was purified by column chromatography (20% MeOH in CH_2Cl_2) to give the *title compound* (0.14 g, 70%) as a yellow powder. R_F 0.25 (20%MeOH: CH_2Cl_2).

Using methyl chloroformate: A stirred mixture of **34h** (0.16 g, 0.5 mmol) in acetonitrile (2 mL) was cooled to -30°C. Pyridine (0.1 mL, 1 mmol) was added, followed by methyl chloroformate (0.1 mL, 1 mmol) and the mixture was stirred at -30°C for 20 minutes. The reaction was left to warm to ambient temperature and stirred for 24 hrs. 2M HCl (5 mL) was added and the resulting mixture was extracted with CH_2Cl_2 (5 mL). The organic layer was washed with 2M HCl (2 x 5 mL) then dried over Na_2SO_4 and evaporated under reduced pressure. The residue was purified by column chromatography as above to give the *title compound* (0.13 g, 68%) as a yellow solid.

Recrystallisation of **55b** from 1:1 MeOH:CH_2Cl_2 gave bright yellow needles, m.p. 250°C dec. (Found: C 50.6, H 4.7, N 20.6; $M^{+\bullet}$ 402.0927; $C_{17}H_{18}N_6O_2S_2$ requires C

50.7, 4.5, 20.9; $M^{+\bullet}$ 402.0932). δH (400MHz, DMSO-d_6) 8.49 (2H, d, *J* 7.2, pyridine-H2,H6), 8.11 (2H, d, *J* 7.2, pyridine-H3,H5), 7.73 (1H, dd, *J* 5.1, 1.1, thienyl-H5), 7.27 (1H, dd, *J* 3.6, 1.1, thienyl-H3), 7.19 (1H, dd, *J* 5.1, 3.6, thienyl-H4), 3.59 (2H, br m, NCH_2); 3.47 (2H, br m, NCH_2), 1.16 (6H, br-m, 2 x CH_3). δC (100MHz, DMSO-d_6) 156.2, 154.1, 150.3, 145.5, 139.6, 134.4, 128.0, 127.8, 127.8, 120.5, 98.0, 41.8, 13.4. *m/z* (EI) 402 (100%, $M^{+\bullet}$), 185 (33), 105.0 (30).

3-(Diethylamino)-5-(pyridazin-4-yl)-6-(thien-2-yl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***55c***



Using methyl chloroformate: Pyridazine (0.1 ml, 1.4 mmol) was added to a stirred mixture of **34h** (0.16 g, 0.5 mmol) in THF (3 mL). The mixture was cooled to -50^{o}C, methyl chloroformate (0.08 mL, 1.5 mmol) was added, and the resulting mixture was left to stir for 30 minutes. The reaction was left to warm to room temperature and stirred overnight. Water (10 mL) was added and the product was extracted with CH_2Cl_2 (2 x 5 mL). The combined extracts were washed sequentially with water (5 mL) and saturated aqueous NaCl (5 mL) and dried over Na_2SO_4 then evaporated *in vacuo*. The red residue was purified by column chromatography (20% MeOH in CH_2Cl_2) to give the *title product* (0.11 g, 55%) as orange-red crystals. R_F 0.3 (20%MeOH: CH_2Cl_2).

Using acetyl chloride: A stirred mixture of **34h** (0.16 g 0.5 mmol) in THF (2 mL) was cooled to -30°C. Pyridazine (0.1 mL, 1.0 mmol) was added, followed by acetyl chloride (0.08 mL, 1.0 mmol) and the resulting mixture was stirred at -30°C for 20 minutes. The reaction was left to warm to ambient temperature and stirred for 24 hrs. 2M HCl (5 mL) was added and the resulting mixture was extracted with CH_2Cl_2 (5 mL). The organic layer was washed with 2M HCl (2 x 5 mL) then dried over Na_2SO_4 and evaporated under reduced pressure. The residue was purified by column chromatography as above to give the *title compound* (93 mg, 46%) as an orange/red solid.

Recrystallisation of **55c** from 1:1 MeOH:CH_2Cl_2 gave small bright red crystals, m.p. 268-271°C dec. (Found: C 47.65, H 4.28, N 24.41; $[M+Na]^+$ 426.0779; $C_{16}H_{17}N_7O_2S_2$ requires C 47.63, 4.25, 24.30; $[M+Na]^+$ 426.0783). δH (400MHz, DMSO-d_6) 9.59 (1H, br s, pyridazine-H3); 9.12 (1H, br d, *J* 6.6, pyridazine-H6); 8.23 (1H, dd, *J* 6.4, 2.2, pyridazine-H5); 7.78 (1H, dd, *J* 5.0, 0.9, thienyl-H5); 7.37 (1H, dd, *J* 3.5, 0.8, thienyl-H3); 7.22 (1H, dd, *J* 5.0, 3.6, thienyl-H4); 3.60 (2H, br s, NCH_2); 3.48 (2H, br s, NCH_2); 1.19 (6H, br s, 2 x CH_3). δC (100MHz, DMSO-d_6, 65°C) 155.9, 153.0, 148.9, 149.0, 144.5, 134.9, 134.4, 127.3, 126.9, 126.8, 121.5, 96.6, 41.3, 13.2.

9.5 Synthesis of "methylene dimers" and methylation from attempted Mannich reactions

General Procedures:

Method A

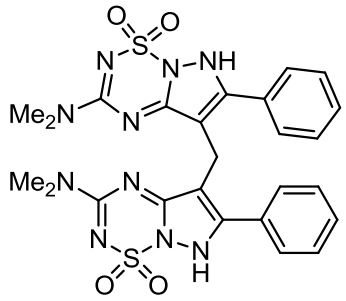
A stirred mixture of compound **34** (0.25 mmol) and paraformaldehyde (0.3 mmol) in acetic acid (1.5 mL) was treated with 40% aqueous dimethylamine (0.28 mmol) in acetic acid (1 mL) slowly at room temperature. The mixture was stirred at 50°C until completion, cooled to room temperature, then diluted with water (5 mL). The pH was adjusted to 8 with 20%w/v aqueous NaOH to form a precipitate which was collected by gravity filtration. The crude solid was purified by column chromatography over silica gel.

Method B

N,*N*'-diisopropylamine (0.3 mmol) was added slowly to a stirred mixture of compound **34** (73 0.25 mmol) and paraformaldehyde (0.3 mmol) in acetic acid (3 mL). The mixture was stirred at 60°C until completion, cooled to room temperature, then diluted with water (5 mL). The pH was adjusted to 8 with 20%w/v aqueous NaOH and the

resulting precipitate was collected by gravity filtration. The crude solid was purified by column chromatography over silica gel.

5,5'-Methylene-bis[3-dimethylamino-6-phenylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide] **58a**

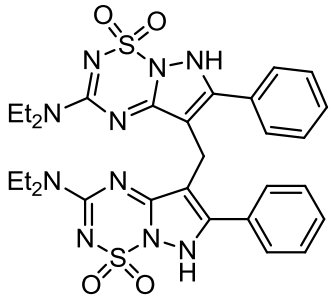


Method A was performed with compound **34d** (15h) to form a white solid. Elution with 15% EtOAc in CH_2Cl_2 provided the *title compound* (58 mg, 76%) as white crystals. R_F 0.1 (15%EtOAc:CH_2Cl_2).

Method B was performed with compound **34d** (15h) to form a white solid. Elution with 10% EtOAc in CH_2Cl_2 afforded the *title compound* (40 mg, 54%) as white crystals.

Recrystallisation of **58a** from DMSO gave white wedges, m.p. 196^{o}C dec. (Found: $[M+H]^+$ 593.1514; $C_{25}H_{25}N_{10}O_4{}^{32}S_2$ requires $[M+H]^+$ 593.1502). δH (600MHz, DMSO-d_6) 10.45 (2H, s, NH), 7.41 (4H, m, ArH), 7.31-7.26 (6H, m, ArH), 3.71 (2H, s, CH_2), 3.02 (12H, s, NCH_3 x 2). δC (150 MHz, DMSO-d_6) 153.4, 151.4, 144.6, 132.6, 128.4, 128.2, 127.8, 99.8, 45.8, 37.3.

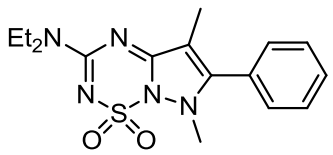
5,5'-Methylene-bis[3-diethylamino-6-phenylpyrazolo [1,5-b][1,2,4,6] thiatriazine 1,1-dioxide] **58b**



Method A was performed with compound **34e** (20h) but no precipitate formed after basification. The solvent was thus evaporated to give a white powder which was purified via column chromatography (50% EtOAc in CH_2Cl_2) to provide the *title compound* (46 mg, 57%) as a white solid. R_F 0.2 (50%EtOAc: CH_2Cl_2).

Recrystallisation of **58b** from DMSO gave white wedges, m.p. 192°C dec. (Found: $[M-H]^-$ 649.2127; $C_{29}H_{34}N_{10}O_4{}^{32}S_2$ requires $[M-H]^-$ 649.2128). δH (600MHz, DMSO-d_6) 10.29 (2H, s, NH), 7.43 (4H, m, ArH), 7.31-7.26 (6H, m, ArH), 3.73 (2H, s, CH_2), 3.42 (8H, q, *J* 6.6, NCH_2CH_3 x 2), 1.10 (12H, m, NCH_2CH_3 x 2). δC (150 MHz, DMSO-d_6) 152.2, 151.5, 144.7, 132.7, 128.4, 128.2, 127.8, 99.4, 45.8, 42.1, 13.1.

5,7-Dimethyl-3-(dimethylamino)-6-phenylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***59***



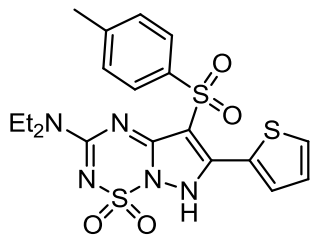
A mixture of **34e** (0.160 g, 0.5 mmol) and dimethylamine.HCl (0.05 g, 0.57 mmol) in acetic acid (3.5 mL) was stirred at room temperature for 5 minutes. Aqueous formaldehyde (40%, 0.05 mL, 0.75 mmol) was then added slowly and the mixture heated to 105°C for 20 hrs. The red solution was then cooled and concentrated *in vacuo*. The syrupy residue was diluted with water (10 mL) and pH adjusted to 13 by 5M NaOH

(aq). The product was extracted with CH_2Cl_2 (2 x 10 mL) and purified via column chromatography (40% EtOAc in CH_2Cl_2) to give the *title compound* (0.035 g, 35%). R_F 0.50 (40% EtOAc:CH_2Cl_2)

Recrystallisation of **59** from CH_2Cl_2 as soft white needles, m.p. 210-212 °C. (Found: $[M+Na]^+$ 370.1318. $C_{16}H_{21}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 370.1314). δH (400MHz, $CDCl_3$) 7.67 (2H, d, J 2.0Hz, o-H x2), 7.51 (3H, m, p + o H x 3), 3.86 (3H, s, NC*H*$_3$), 3.54 (3H, s, C5-CH_3), 3.35 (4H, q, J 7.08, NC*H*$_2$$CH_3$ x 2), 1.24 (6H, t, J 7.12, NCH_2C*H*$_3$ x 2). δC (100MHz, $CDCl_3$) 155.0, 149.3, 140.7, 130.3, 129.8, 129.2, 127.00, 106.8, 44.8, 38.0, 37.1, 13.1.

9.6 Synthesis of products from *p*-toluenesulfonylation experiments

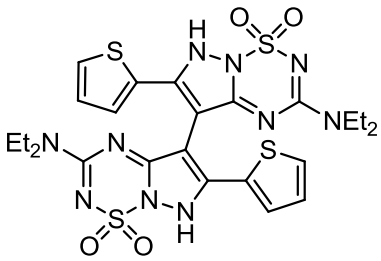
3-Diethylamino-6-(thien-2-yl)-5-tosyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***60***



A mixture of compound **34h** (0.163 g, 0.5 mmol), K_2CO_3 (0.11 g, 0.75 mmol), *p*-toluene sulfonyl chloride (0.146 g, 0.75 mmol), and acetonitrile (3 mL) was stirred at 60°C for 42 hrs. The mixture was cooled to room temperature, diluted with CH_2Cl_2 (10 mL), and washed with deionised water until pH was neutral (2 x 10 mL), followed by washing with saturated aqueous NaCl (5 mL). The aqueous layers were combined and extracted with CH_2Cl_2 (2 x 10 mL), dried, and evaporated to afford a dark green oil which was purified by column chromatography (30% EtOAc in CH_2Cl_2) to yield the *title compound* (72 mg, 30%) as a white powder. R_F 0.40 (30% EtOAc:CH_2Cl_2).

Recrystallisation of **59** from CH_2Cl_2 gave white needles, m.p. 120°C dec. (Found: $[M-H]^-$ 478.0673; $C_{19}H_{20}N_5O_4{}^{32}S_3$ requires $[M-H]^-$ 478.0683). δH (400MHz, $CDCl_3$) 7.81 (1H, dd, *J* 3.7, 1.1, thienyl H5), 7.63 (1H, s, NH), 7.34 (1H, dd, *J* 5.0, 1.1, thienyl H3), 7.08-7.01 (5H, m, ArH + thienyl H4), 3.49 (4H, br-s, NCH_2 x 2), 2.28 (3H, s, CH_3), 1.24 (6H, t, *J* 7.0, NCH_2CH_3 x 2). δC (150MHz, CD_3CN, 35°C) 151.2, 149.1, 144.9, 137.2, 134.0, 133.7, 131.1, 128.7, 128.6, 128.5, 127.3, 89.8, 44.6, 20.9, 13.1.

3-Diethylamino-6-(thien-2-yl)-5-tosyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***60*** *and 3,3'-bis(diethylamino)-6,6'-di(thien-2-yl)-7H,7'H-[5,5'-bipyrazolo[1,5-b][1,2,4,6]thiatriazine] 1,1,1',1'-tetraoxide* ***61***



A solution of compound **34d** (0.163 g, 0.5 mmol), *p*-toluene sulfonyl chloride (0.116 g, 0.6 mmol) and 1-methylimidazole (18 mg, 0.21 mmol) in acetonitrile (1.5 mL) was treated slowly with triethylamine (0.1 mL, 0.7 mmol) at room temperature. The mixture was stirred at room temperature for 21 hrs, quenched with saturated aqueous $NaHCO_3$ (5 mL) and extracted with CH_2Cl_2 (2 x 5 mL). The organic layers were combined and dried, then evaporated *in vacuo* to give a brown oil. Purification by column chromatography (10% EtOAc in CH_2Cl_2) gave two products, in order of elution: **60** (26 mg, 11%) as a fawn solid and **61** (36 mg, 22%) as colourless blocks. **61**: R_F 0.2 (10% EtOAc:CH_2Cl_2) The starting material was also isolated from the column (94 mg, 58%).

A solution of compound **34d** (0.161 g, 0.5 mmol), *p*-toluene sulfonyl chloride (0.160 g, 0.6 mmol) and 1-methyl imidazole (40 mg, 0.5 mmol) in acetonitrile (1.5 mL) was treated slowly with triethylamine (0.1 mL, 0.7 mmol) at room temperature. The mixture was heated at reflux for 21 hours, quenched with saturated aqueous $NaHCO_3$ (5 mL) and extracted with CH_2Cl_2 (2 x 5 mL). The combined organic layers were dried and evaporated *in vacuo* to give a green oil, which was purified by column chromatography.

Elution with 40% EtOAc in CH_2Cl_2 gave two products, in order of elution: **60** (19 mg, 8%) as a beige solid and **61** (16 mg, 11%) as colourless blocks. The starting material was also isolated from the column (46 mg, 28%).

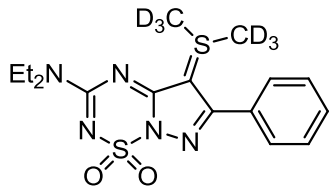
Recrystallisation of **61** from 1:1 CH_2Cl_2:MeOH gave colourless blocks, m.p. 220°C dec. (Found: $[M+Na]^+$ 671.1080; $C_{24}H_{28}N_{10}O_4{}^{32}S_4Na$ requires $[M+Na]^+$ 671.1076). δH (400MHz, DMSO-d_6) 10.98 (1H, s, NH), 7.51 (1H, br-d, *J* 4.6, thienyl H5), 7.06 (1H, dd, *J* 4.7, 1.0, thienyl H3), 6.99 (1H, dd, *J* 4.7, 3.9, thienyl H4), 3.50 (4H, m, NCH_2CH_3 x 2), 1.03 (6H, t, *J* 6.7, NCH_2CH_3 x 2). δH (150MHz, DMSO-d_6, 40°C) 155.5, 155.3, 146.0, 132.5, 127.7, 127.7, 127.6, 74.0, 41.7, 13.4.

9.7 Synthesis of 5-dimethylthio derivatives

General Procedure

A mixture of **34** (0.5 mmol or 0.25 mmol), DMSO (5.5 equiv) in acetonitrile (2 mL) was treated slowly with triethylamine (5 equiv) and then cooled via ice bath to 5°C for the dropwise addition of TFAA (5 equiv). The mixture was stirred at room temperature until completion and treated with water (5 mL) to form a precipitate which was filtered under vacuum and washed with 1M aqueous HCl. If no precipitate formed an extractive workup with water and EtOAc (2 x 5mL) was performed, combined extracts dried *in vacuo*, then purified via column chromatography over silica gel.

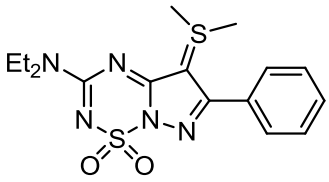
5-(Bis(methyl-d3)sulfanylidene)-3-diethylamino-6-phenyl-5H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***62a***



The general procedure was followed with **34e** (0.162 g, 0.5 mmol) and DMSO-d_6 (41h) with an extractive workup using EtOAc (2 x 5 mL). Elution with 40% EtOAc in CH_2Cl_2 provided the *title compound* as a white powder (0.12 g, 67%) R_F 0.35 (40%EtOAc:CH_2Cl_2) and the starting material (12 mg, 7%).

Recrystallisation of **62a** from 1:1 MeOH:CH_2Cl_2 gave colourless prisms, m.p. 209°C dec. (Found: $[M+Na]^+$ 408.1416; $C_{16}H_{15}D_6N_5O_2{}^{32}S_2Na$ requires $[M+Na]^+$ 408.1416). δH (600MHz, $CDCl_3$) 7.73-7.71 (2H, m, ArH); 7.53-7.50 (3H, m, ArH); 3.55 (2H, m, NCH_2); 3.44 (2H, s, NCH_2); 1.16-1.13 (6H, m, CH_3 x 2). δC (150MHz, $CDCl_3$) 155.5, 155.4, 151.8, 130.8, 129.3, 128.6, 128.4, 74.4, 48.6, 13.6. *When at 56°C d1=4 the signal as 155.4 merges with that at 155.5 and one signal at 155.4 is seen.

3-Diethylamino-5-(dimethylsulfanylidene)-6-phenyl-5H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***62b***

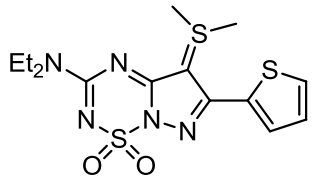


The general procedure was followed with **34e** (82 mg, 0.25 mmol) and DMSO (15min) with an extractive workup using EtOAc (2 x 5 mL). Elution with 30% EtOAc in CH_2Cl_2 provided the *title compound* as white needles (65 mg, 69%). R_F 0.2 (30%EtOAc:CH_2Cl_2).

Recrystallisation of **62b** from MeOH gave white needles, m.p. 196°C dec. (Found: $[M+Na]^+$ 402.1037; $C_{16}H_{21}N_5O_2{}^{32}S_2Na$ requires $[M+Na]^+$ 402.1034). δH (600MHz, DMSO-d^6, 25°C) 7.73-7.71 (2H, m, ArH); 7.52-7.50 (3H, m, ArH); 3.55 (2H, m, NCH_2); 3.46 (2H, s, NCH_2); 3.32 (6H, s, $S(CH_3)_2$), 1.16 (6H, m, NCH_2CH_3 x 2). δH (600MHz, DMSO-d^6, 65°C) 7.73-7.71 (2H, m, ArH); 7.53-7.49 (3H, m, ArH); 3.52 (4H, br m, NCH_2 x 2); 3.34 (6H, s, $S(CH_3)_2$), 1.17 (6H, t, *J* 7.0, NCH_2CH_3 x 2). δC (150MHz, DMSO-d^6) 155.5, 155.4, 151.8, 130.7, 129.3, 128.6, 128.4, 74.9, 41.8, 41.7,

26.3, 13.5. #There is a short range hector correlation (HMQC) between the proton singlet at δ 3.32 ppm and the carbon signal at δ 26.3 ppm.

3-(Diethylamino)-5-(dimethylsulfanylidene)-6-(thien-2-yl)-5H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***62c***

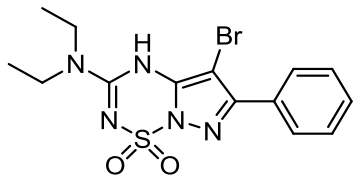


The general procedure was followed with **34h** (82 mg, 0.25 mmol) and DMSO (10min) to give the *title compound* as white needles (72 mg, 75%).

Recrystallisation of **62c** from MeOH gave colourless blocks, m.p. 201°C dec. (Found: $[M+Na]^+$ 408.0610; $C_{14}H_{19}N_5O_2{}^{32}S_3Na$ requires $[M+Na]^+$ 408.0599). δH (600MHz, DMSO-d^6, 25°C) 7.70 (1H, dd, *J* 0.6, 5.0, thienyl H5), 7.66 (1H, dd, *J* 0.6, 3.7, thienyl H3), 7.21 (1H, dd, J 5.0, 3.7, thienyl H4), 3.56 (2H, m, NCH_2), 3.44 (2H, m, NCH_2), 3.34 (6H, s, $S(CH_3)_2$), 1.18-1.13 (6H, m, NCH_2CH_3 x 2). δC (150MHz, DMSO-d^6) 155.5, 155.4, 146.2, 132.6, 127.9, 127.8, 127.7, 74.3, 41.8, 41.7, 26.3, 13.6, 13.5.

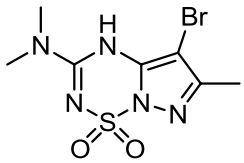
9.8 Synthesis of 5-bromo-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine derivatives

5-Bromo-3-diethylamino-6-phenyl-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***63a***



A mixture of **34e** (0.161 g, 0.5 mmol) and N-bromosuccinimide (100 mg, 0.5 mmol) in $CHCl_3$ (3 mL) was heated to 60°C for 30 minutes. The reaction mixture was then cooled and diluted with $CHCl_3$ (5 mL) then washed with saturated aqueous $NaHCO_3$ (2 x 5 mL). The organic layer was dried and evaporated to afford the *title compound* (0.193 g, 97% crude) as a red semisolid. (Found [low resolution] $M^{+\bullet}$ 398.0; $C_{14}H_{16}N_5O_2{}^{32}S^{79}Br$ requires $M^{+\bullet}$ 398.2). δH (600MHz, $CDCl_3$) 8.29 (2H, d, *J* 7.5, ArH), 7.58 (1H, t, *J* 5.6, ArH), 7.52 (2H, t, *J* 7.3, ArH), 3.78 (2H, q, *J* 7.1, NCH_2), 3.64 (2H, q, *J* 7.1, NCH_2), 1.31 (6H, t, J 7.1, CH_3 x 2). δC (150MHz, $CDCl_3$) 163.9, 155.3, 155.8, 132.8, 129.4, 129.0, 128.4, 127.3, 127.3, 125.5, 44.1, 44.0, 14.2, 12.6.

5-Bromo-3-dimethylamino-6-methyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***63b***



From ethyl bromoacetoacetate: A mixture of compound **34e** (0.113 g, 0.5 mmol), K_2CO_3 (0.150 g, 1.1 mmol), $^nBu_4NBr$ (29 mg, 0.1 mmol) in acetonitrile (3 mL) was treated with ethyl bromoacetoacetate (0.23 g, 1.1 mmol) and stirred at 50°C for 18 hrs. The mixture was then cooled to room temperature, diluted with CH_2Cl_2 (10 mL) and washed with water (2 x 5 mL). The combined aqueous phases were extracted with CH_2Cl_2 (10 mL). The combined organic layers were dried and evaporated to give a brown oil which was purified by column chromatography. Elution with 40% EtOAc in CH_2Cl_2 afforded the *title compound* (68 mg, 41%) as a white powder. R_F 0.3 (40%EtOAc:CH_2Cl_2).

From diethyl bromomalonate: A similar reaction, employing diethyl bromomalonate (1.1 mmol) in place of ethyl bromoacetoacetate, and purification under the conditions described above afforded the *title compound* (82 mg, 50%) as a white powder.

Recrystallisation of **63a** from 1:1 MeOH:CH_2Cl_2 as white needles, m.p 175-177°C dec. (Found: $[M+Na]^+$ 329.9636. $C_7H_{10}N_5O_2{}^{32}S^{79}BrNa$ requires $[M+Na]^+$ 329.9636). δH

(400MHz, DMSO-d^6) 3.11 (6H, s, NCH_3 x 2), 2.18 (3H, s, CCH_3). δC (100MHz, DMSO-d^6) 151.6, 150.4, 140.3, 90.9, 38.7, 13.0.

9.9 General synthesis procedures for benzylated 3-dialkylamino-pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-dioxides 64a-67g

Method A

A mixture of pyrazolothiatriazine **34** (0.5 mmol), benzyl halide (0.55 mmol), K_2CO_3 (0.55 mmol), $^nBu_4NBr$ (0.055 mmol), and CH_2Cl_2 (5 mL) was stirred at room temperature overnight or until TLC indicated completion. The mixture was washed sequentially with water (5 mL) and saturated aqueous NaCl (5 mL). The organic phase was dried and evaporated and the residue was purified by column chromatography over silica gel.

Method B

A mixture of compound **34** (0.5 mmol), benzyl halide (0.57 mmol), and THF (5 mL) was cooled to 5°C in an ice-water bath and triethylamine (1.1 mmol) was added dropwise. The mixture was stirred at room temperature overnight or until TLC indicated completion, then diluted with CH_2Cl_2 (10 mL) and washed sequentially with water (5 mL) and saturated aqueous NaCl (5 mL). The organic layer was then dried and evaporated. The residue was purified by column chromatography over silica gel.

Method C

A mixture of compound **34** (0.5 mmol), benzyl halide (0.55 mmol), K_2CO_3 (0.55 mmol), $^nBu_4NBr$ (0.055 mmol), and acetonitrile (5 mL) was stirred at room temperature overnight or until TLC indicated completion. The solvent was evaporated and the residual gum was washed with water (5 mL), then extracted with CH_2Cl_2 (2 x

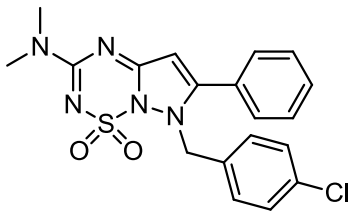
10 mL). The organic phases were combined and washed with saturated aqueous NaCl (5 mL), dried, and evaporated. The residue was purified by column chromatography over silica gel.

Method D

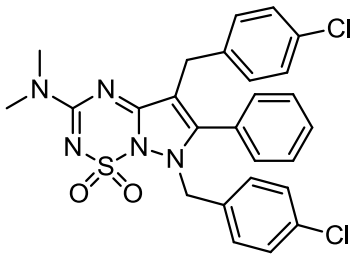
A stirred mixture of **34** (0.5 mmol), benzyl halide (0.55 mmol), K_2CO_3 (0.55 mmol), $^{n}Bu_4NBr$ (0.055 mmol), and acetonitrile (3 mL) was irradiated under microwave conditions at 50°C for 130 minutes. The mixture was diluted with water (5 mL), extracted with CH_2Cl_2 (2 x 5 mL), dried and evaporated. The residue was purified by column chromatography over silica gel.

The following compounds were prepared using the above procedures. All products are listed in order of elution from chromatography:

7-(4-Chlorobenzyl)-3-dimethylamino-6-phenyl-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64a*** *and 5,7-Bis(4-chlorobenzyl)-3-dimethylamino-6-phenyl-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65a***



Method A. Elution with CH_2Cl_2 then gradient to 2.5% EtOAc in CH_2Cl_2 gave the *title compound* (103 mg, 49%) as a white solid. Recrystallisation from DMSO gave white needles, m.p. 179-180°C. (Found: C 53.35, H 4.67, N 16.35; $M^{+\bullet}$ 415.0865; $C_{19}H_{18}{}^{35}ClN_5O_2{}^{32}S.{}^{1/2}H_2O$ requires C 53.71, H 4.51, N 16.48; $M^{+\bullet}$ 415.0864). δH (400MHz, $CDCl_3$) 7.57-7.50 (5H, m, ArH), 7.12 (2H, d, *J* 8.4, ArH), 6.86 (2H, d, *J* 8.4, ArH), 5.95 (1H, s, H5), 5.33 (2H, s, NCH_2Ar), 3.17 (3H, s, NCH_3), 3.16 (3H, s, NCH_3). δC (100MHz, $CDCl_3$) 162.0, 159.2, 159.0, 134.5, 131.9, 131.7, 129.9, 129.6, 129.1, 128.7, 127.5, 101.3, 54.1, 37.6, 37.1. *m/z* (EI) 415/417 (23/9%, $M^{+\bullet}$), 125/127 (100/35).

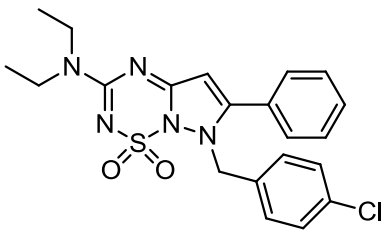


Method B. Elution with CH_2Cl_2 then gradient to 2.5% EtOAc in CH_2Cl_2 afforded two products: **65a** (22 mg, 15%) and **64a** (0.125 g, 60%), both as white solids. Recrystallisation of **65a** from MeOH:CH_2Cl_2 gave white prisms, m.p. 177.5-179°C. (Found $M^{+\bullet}$ 539.0944; $C_{26}H_{23}{}^{35}Cl_2N_5O_2{}^{32}S$ requires $M^{+\bullet}$ 539.0945). δH (400MHz, $CDCl_3$) 7.60-7.50 (3H, m, ArH), 7.31 (2H, d, *J* 8.0, ArH), 7.15 (4H, m, ArH), 6.86 (4H, m, ArH), 5.20 (2H, s, NC*H*$_2$Ar), 3.65 (2H, s, CC*H*$_2$Ar), 3.16 (3H, s, NCH_3), 3.12 (3H, s, NCH_3). δC (100MHz, $CDCl_3$) 161.4, 158.9, 155.9, 137.6, 134.5, 132.1, 131.9, 131.4, 130.0, 129.6, 129.4, 129.2, 128.8, 128.6, 128.5, 126.9, 112.3, 54.1, 37.4, 37.1, 27.5. *m/z* (EI) 539/541/543 (22/16/3%, $M^{+\bullet}$), 125/127 (100/35).

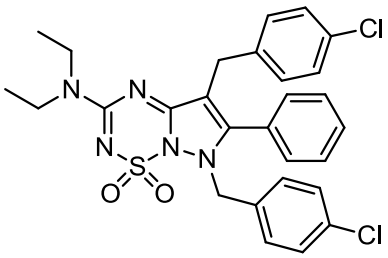
7-(4-Chlorobenzyl)-3-diethylamino-6-phenylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64b*** *and 5,7-Bis(4-chlorobenzyl)-3-diethylamino-6-phenyl-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65b*** *and 4-(4-Chlorobenzyl)-3-diethylamino-6-phenyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66b***

Method C. Elution with CH_2Cl_2 gave two products: **65b** (2 mg, 1%) and **64b** (0.19 g, 86%)**,** both isolated as white solids.

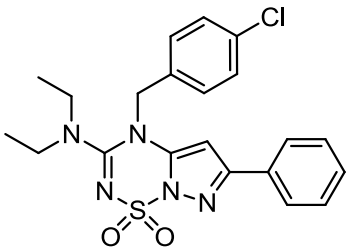
Method D. Elution with CH_2Cl_2 then gradient to 2.5% EtOAc in CH_2Cl_2 afforded three products: **65b** (15 mg, 10%) as an off-white powder, **64b** (0.161 g, 73%) as a white powder, and **66b** (1 mg, 1%) as a beige oil.



64b: Recrystallisation from CH_2Cl_2 gave white blocks, m.p. 188-190°C. (Found: C 57.01, H 5.19, N 15.39; $M^{+\bullet}$ 443.1177; $C_{21}H_{22}{}^{35}ClN_5O_2{}^{32}S$ requires C 56.81, H 4.99, N 15.78; $M^{+\bullet}$ 443.1177). δH (400MHz, $CDCl_3$) 7.63-7.49 (5H, m, ArH), 7.13 (2H, d, *J* 8.4, ArH), 6.88 (2H, d, *J* 5.2, ArH), 5.93 (1H, s, H5), 5.33 (2H, s, NC*H*$_2$PhCl), 3.62-3.53 (4H, m, NC*H*$_2$ x 2), 1.23(3H, t, *J* 7.1, NCH_2CH_3), 1.19 (3H, t, *J* 7.1, NCH_2CH_3). δC (100MHz, $CDCl_3$) 162.0, 158.6, 158.2, 134.5, 132.0, 131.6, 129.9, 129.5, 129.0, 128.6, 127.6, 101.4, 54.0, 42.57, 42.2, 13.5, 13.3. *m/z* (EI) 443/445 (24/10%, $M^{+\bullet}$), 364/366 (27/9), 125/127 (100/37), 72 (69).



65b: Recrystallisation from CH_2Cl_2 gave white needles m.p. 149-150°C. (Found $M^{+\bullet}$ 567.1249; $C_{28}H_{27}{}^{35}Cl_2N_5O_2{}^{32}S$ requires $M^{+\bullet}$ 567.1257). δH (400MHz, $CDCl_3$) 7.60-7.49 (3H, m, ArH), 7.31 (2H, d, *J* 7.2, ArH), 7.16-7.13 (4H, m, ArH), 6.90-6.86 (4H, m, ArH), 5.21 (2H, s, NC*H*$_2$Ar), 3.63 (2H, s, CC*H*$_2$Ar), 3.58-3.49 (4H, m, NC*H*$_2$ x 2), 1.23 (3H, t, *J* 7.1, NCH_2CH_3), 1.09 (3H, t, *J* 7.1, NCH_2CH_3). δC (100MHz, $CDCl_3$) 161.2, 157.9, 155.3, 137.7, 134.4, 132.1, 132.0, 131.3, 130.0, 129.6, 129.4, 129.3, 128.6, 128.5, 126.9, 112.1, 53.9, 42.7, 42.5, 27.6, 13.3. *m/z* (EI) 567/569/571 (11/8/2%, $M^{+\bullet}$), 434 (21), 432 (17), 125/127 (100/34), 72 (47).

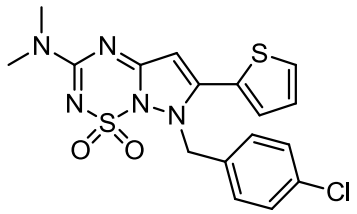


66b: Isolated as a beige oil. δH (400MHz, $CDCl_3$), 7.75 (2H, m, ArH), 7.37-7.35 (5H, m, ArH), 7.22 (2H, d, *J* 8.4, ArH), 5.95 (1H, s, H5), 5.00 (2H, s, C*H*$_2$Ar), 3.43 (4H, q, *J* 7.2, NC*H*$_2$CH$_3$ x 2), 1.23 (6H, t, *J* 7.2, NCH_2CH_3 x 2).

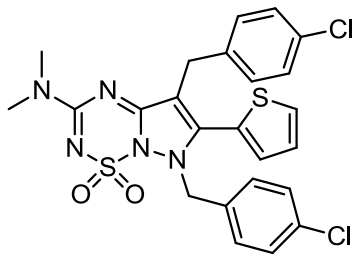
7-(4-Chlorobenzyl)-3-dimethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64c****, 5,7-Bis(4-chlorobenzyl)-3-dimethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65c****, 4-(4-Chlorobenzyl)-3-dimethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66c****, and 2-(4-Chlorobenzyl)-3-dimethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***67c***

Method B. Elution with CH_2Cl_2 then gradient to 2.5% EtOAc in CH_2Cl_2 gave four products: **65c** (20 mg, 13%) as a white powder, **67c** (1 mg, 1%) as a yellow oil, **64c** (96 mg, 46%) as a white powder, and **66c** (9 mg, 4%) as a white powder.

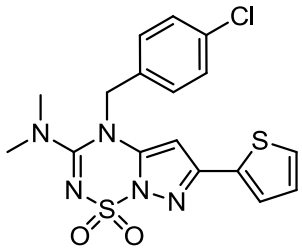
Method C. Elution with CH_2Cl_2 then gradient to 2.5% EtOAc in CH_2Cl_2 afforded three products: **65c** (22 mg, 13%) as a white powder, **64c** (162 mg, 77%) as white rods, and **66c** (6 mg, 3%) as a white amorphous solid.



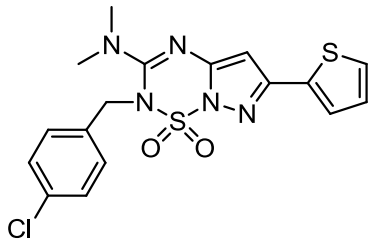
64c: Recrystallisation from CH_2Cl_2 gave white blocks, m.p. 188-190°C**.** (Found: C 57.01, H 5.19, N 15.39; $M^{+\bullet}$ 421.0422; $C_{17}H_{16}{}^{35}ClN_5O_2{}^{32}S_2$ requires C 56.81, H 4.99, N 15.78; $M^{+\bullet}$ 421.0428). δH (400MHz, $CDCl_3$) 7.66 (1H, dd, *J* 5.0, 1.0, thienyl-H5), 7.43 (1H, dd, *J* 3.7, 1.0, thienyl-H3), 7.24 (1H, dd, *J* 5.0, 3.8, thienyl-H4), 7.17 (2H, d, *J* 8.4, ArH), 6.98 (2H, d, *J* 8.4, ArH), 5.98 (1H, s, H5), 5.45 (2H, s, NC*H*$_2$Ar), 3.17 (3H, s, NCH_3), 3.16 (3H, s, NCH_3). δC (100MHz, $CDCl_3$) 161.9, 158.0, 151.9, 134.6, 131.9, 131.1, 130.9, 129.8, 128.9, 128.8, 128.0, 101.3, 54.7, 37.6, 37.2. *m/z* (EI) 421/423 (68/31%, $M^{+\bullet}$), 134 (38), 125/127 (100/34).



65c: Recrystallisation from CH_2Cl_2 gave white needles, m.p. 170-171.5°C. (Found: $M^{+\bullet}$ 545.0508; $C_{24}H_{21}{}^{35}Cl_2N_5O_2{}^{32}S_2$ requires $M^{+\bullet}$ 545.0514). δH (400MHz, $CDCl_3$) 7.67 (1H, dd, *J* 4.5, 1.6, thienyl-H5), 7.24-7.23 (2H, m, thienyl-H3,H4), 7.16 (4H, m, ArH), 6.91 (2H, d, *J* 8.4, ArH), 6.83 (2H, d, *J* 8.8, ArH), 5.31 (2H, s, NC*H*$_2$Ar), 3.75 (2H, s, CC*H*$_2$Ar), 3.15 (3H, s, NCH_3), 3.11 (3H, s, NCH_3). δC (100MHz, $CDCl_3$) 161.8, 159.0, 150.1, 137.2, 134.6, 132.2, 131.6, 131.6, 131.2, 130.2, 129.2, 128.6, 128.6, 128.5, 127.0, 113.4, 55.2, 37.5, 37.1, 27.7. *m/z* (EI) 545/547/549 (24/19/4%, $M^{+\bullet}$), 125/127 (100/33).



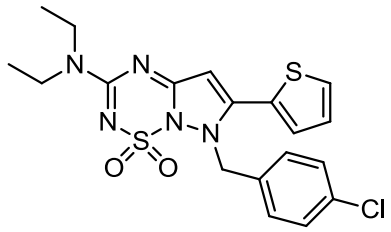
66c: Recrystallisation from CH_2Cl_2 gave white blocks, m.p. 221-222°C. (Found: $M^{+\bullet}$ 421.0428; $C_{17}H_{16}{}^{35}ClN_5O_2{}^{32}S_2$ requires $M^{+\bullet}$ 421.0428). δH (400MHz, DMSO-d_6) 7.60 (1H, dd, *J* 5.1, 1.0, thienyl-H5), 7.48 (1H, dd, *J* 3.5, 1.1, thienyl-H3), 7.39 (2H, d, *J* 8.0, ArH), 7.32 (2H, d, *J* 7.6, ArH), 7.12 (1H, dd, *J* 5.0, 3.6, thienyl-H4), 6.58 (1H, s, H5), 5.19 (2H, s, NC*H*$_2$Ar), 3.09 (6H, s, NCH_3 x 2). δC (100MHz, DMSO-d_6) 155.5, 148.8, 143.8, 134.1, 133.9, 132.7, 129.4, 128.5, 127.9, 127.3, 126.7, 91.9, 54.0, 40.7. *m/z* (EI) 421/423 (21/9%, $M^{+\bullet}$), 125/127 (100/33).



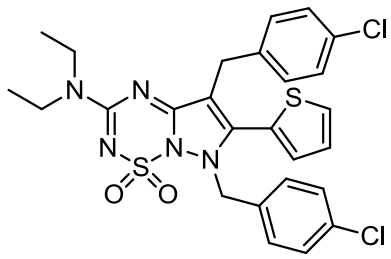
67c: Obtained as a yellow oil. δH (400MHz, $CDCl_3$) 7.35 (1H, dd, *J* 3.5, 0.8, thienyl-H3), 7.32 (1H, dd, *J* 5.1, 0.8, thienyl-H5), 7.14 (2H, d, *J* 8.8, ArH), 7.03 (2H, d, *J* 8.8, ArH), 7.05 (1H, dd, *J* 5.2, 3.6, thienyl-H4), 5.86 (1H, s, H5), 4.66 (2H, s, NC*H*$_2$ArH), 3.13 (6H, s, NC*H*$_3$ x 2).

7-(4-Chlorobenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64d****, 5,7-Bis(4-chlorobenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65d****, and 4-(4-Chlorobenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66d***

Method C. Elution with CH_2Cl_2 gave three products: **65d** (31 mg, 20%) as a yellow powder, **64d** (162 mg, 72%) as a beige solid, and **66d** (1 mg, 0.5%) as a yellow oil.

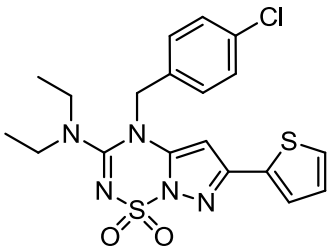


64d: Recrystallisation from CH_2Cl_2 gave light yellow crystals, m.p. 173-174°C. (Found: C 50.60, H 4.59, N 15.44; $M^{+\bullet}$ 449.0737; $C_{19}H_{20}{}^{35}ClN_5O_2{}^{32}S_2$ requires C 50.71, H 4.48, N 15.56; $M^{+\bullet}$ 449.0741). δH (400MHz, $CDCl_3$) 7.65 (1H, dd, *J* 5.1, 1.1, thienyl-H5), 7.42 (1H, dd, *J* 3.7, 1.1, thienyl-H3), 7.24 (1H, dd, *J* 5.1, 3.8, thienyl-H4), 7.18 (2H, d, *J* 8.5, ArH), 6.98 (2H, d, *J* 8.5, ArH), 5.97 (1H, s, H5), 5.46 (2H, s, NCH_2Ar), 3.58 (2H, q, *J* 7.1, NCH_2CH$_3$), 3.54 (2H, q, *J* 7.1, NCH_2CH$_3$), 1.22 (3H, t, *J* 7.2, NCH$_2$CH_3), 1.17 (3H, t, *J* 7.2, NCH$_2$CH_3). δC (100MHz, $CDCl_3$) 161.8, 158.1, 151.4, 134.5, 132.0, 130.9, 130.7, 129.8, 128.8, 128.7, 128.0, 101.3, 54.5, 42.6, 42.2, 13.4, 13.2. *m/z* (EI) (%) 449/451 (34/15%, $M^{+\bullet}$), 370/372 (31/12), 134 (28), 125/127 (100/32), 72 (61).



65d: Recrystallisation from CH_2Cl_2 gave light yellow crystals, m.p. 154-156°C. (Found: $M^{+\bullet}$ 573.0821; $C_{26}H_{25}{}^{35}Cl_2N_5O_2{}^{32}S_2$ requires $M^{+\bullet}$ 573.0828). δH (400MHz, $CDCl_3$) 7.66 (1H, dd, *J* 4.6, 2.7, thienyl-H5), 7.24-7.22 (2H, m, thienyl-H3,H4), 7.14 (4H, m, ArH), 6.92 (2H, d, *J* 8.4, ArH), 6.86 (2H, d, *J* 8.3, ArH), 5.31 (2H, s, NCH_2Ar), 3.73 (2H, s,

CCH_2Ar), 3.57-3.49 (4H, m, NCH_2CH$_3$ x 2), 1.23 (3H, *J* 7.1, t, NCH$_2$CH_3), 1.09 (3H, *J* 7.1, t, NCH$_2$CH_3). δC (100MHz, $CDCl_3$) 161.5, 157.9, 149.4, 137.3, 134.5, 132.1, 131.8, 131.5, 130.1, 129.5, 128.7, 128.7, 128.6, 128.5, 126.9, 113.3, 55.0, 42.7, 42.5, 27.8, 13.3, 13.2. *m/z* (EI) (%) 573/575/577 (11/8/2%, $M^{+\bullet}$), 440 (17), 438 (15), 125/127 (100/32), 72 (42).

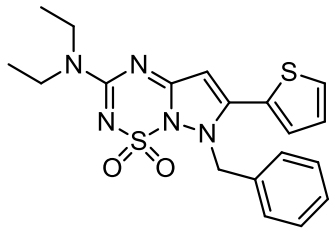


66d: Obtained as a yellow oil. δH (400MHz, $CDCl_3$) 7.36 (2H, d, *J* 8.4, ArH), 7.33 (1H, dd, 3.6, 0.9, thienyl-H3), 7.30 (1H, dd, *J* 5.1, 0.9, thienyl-H5), 7.20 (2H, d, *J* 8.4, ArH), 7.01 (1H, dd, *J* 5.0, 3.6, thienyl-H4), 5.84 (1H, s, H5), 4.98 (2H, s, NCH_2Ar), 3.42 (4H, q, *J* 7.1, NCH_2CH$_3$ x 2), 1.17 (6H, t, *J* 7.1, NCH$_2$CH_3 x 2).

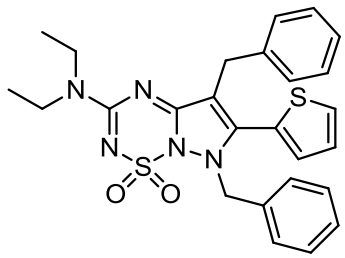
7-Benzyl-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64e****, 5,7-*Bis*-benzyl-3-(diethylamino)-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65e*** *and 4-Benzyl-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66e***

Method C (45h) followed by elution with CH_2Cl_2 gave three compounds: **65e** (44 mg, 14%) as a beige solid, **64e** (288 mg, 69%) as a white solid, and **66e** (7 mg, 2%) as a colourless oil.

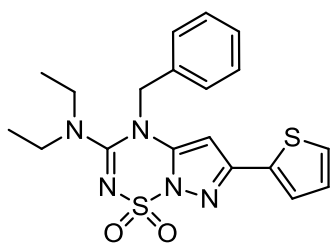
Method D (3h) followed by elution with CH_2Cl_2 gave three compounds: **65e** (9 mg, 6%) as a white solid, **64e** (150 mg, 72%) as a white powder, and **66e** (25 mg, 12%) as a white semi-solid.



64e: Recrystallisation from EtOAc:CH_2Cl_3 gave white blocks, m.p. 149-150°C. (Found: $[M+H]^+$ 416.1203; $C_{19}H_{22}N_5O_2{}^{32}S_2$ requires $[M+H]^+$ 416.1215). δH (600MHz, $CDCl_3$) 7.62 (1H, dd, *J* 4.7, 1.2, thienyl-H5), 7.42 (1H, dd, *J* 3.6, 1.2, thienyl-H3), 7.23-7.19 (4H, m, thienyl H4 & ArH), 7.04 (2H, d, *J* 7.2, ArH), 5.97 (1H, s, H5), 5.52 (2H, s, CH_2Ar), 3.60-3.52 (4H, m, NCH_2CH_3 x 2), 1.22 (3H, t, *J* 7.2, NCH_2CH_3), 1.17 (3H, t, *J* 7.2, NCH_2CH_3). δC (600MHz, $CDCl_3$) 161.7, 158.2, 151.4, 133.9, 131.0, 130.8, 128.8, 128.7, 128.6, 128.3, 128.3, 101.0, 55.2, 42.7, 42.3, 13.6, 13.4.



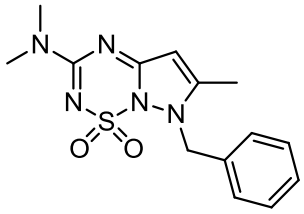
65e: Recrystallisation from CH_2Cl_2 gave yellow needles, m.p. 64-66°C. (Found: $[M+H]^+$ 506.1696; $C_{26}H_{28}N_5O_2{}^{32}S_2$ requires $[M+H]^+$ 506.1684). δH (600MHz, $CDCl_3$) 7.63 (1H, dd, *J* 4.8, 1.2, thienyl-H5), 7.24-7.15 (8H, m, thienyl H4 & H3, ArH), 6.98 (2H, d, *J* 7.2, ArH), 6.94 (2H, d, *J* 7.2, ArH), 5.37 (2H, s, CH_2Ar), 3.77 (2H, CH_2Ar), 3.55-3.49 (4H, m, NCH_2CH_3 x 2), 1.22 (3H, t, *J* 7.2, NCH_2CH_3), 1.07 (3H, t, *J* 7.2, NCH_2CH_3). δC (600MHz, $CDCl_3$) 161.7, 158.1, 149.7, 139.1, 133.7, 131.6, 130.9, 129.6, 128.7, 128.6, 128.5, 128.5, 128.1, 127.4, 126.3, 113.7, 55.8, 42.8, 42.6, 28.4, 13.4.



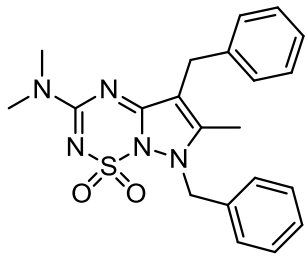
66e: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave white needles, m.p. 158-160°C. (Found: $[M+Na]^+$ 438.1033; $C_{19}H_{21}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 438.1032). δH (600MHz, $CDCl_3$) 7.38-7.35 (2H, m, ArH), 7.33-7.24 (5H, m, ArH, thienyl-H3,H5), 6.99 (1H, dd, *J* 3.7, 1.3, thienyl H4), 5.85 (1H, s, H5), 5.01 (2H, s, CH_2Ar), 3.41 (4H, q, *J* 7.1, NCH_2CH_3 x 2), 1.20 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (600MHz, $CDCl_3$) 155.6, 149.7, 144.1, 134.6, 133.6, 129.4, 128.7, 127.5, 126.6, 126.4, 126.1, 91.6, 56.4, 45.2, 12.8.

7-Benzyl-3-dimethylamino-6-methylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64f****, 5,7-bis-benzyl-3-dimethylamino-6-methylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65f,*** *4-Benzyl-3-dimethylamino-6-methylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66f*** *and 2-Benzyl-3-dimethylamino-6-methylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***67f***

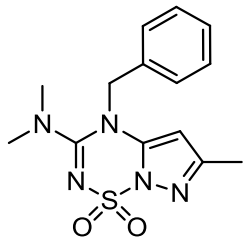
Method C (41h) followed by elution with CH_2Cl_2 gave four compounds: **65f** (98 mg, 11%) as a colourless oil; **67f** (3 mg, 2%) as a colourless oil, **64f** (0.407 g, 60%) as an off white solid, and **66f** (85 mg, 6%) as a white powder.



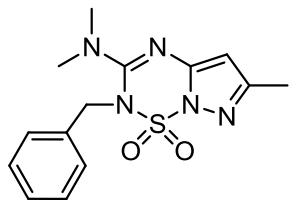
64f: Recrystallisation of **6f** from CH_2Cl_2 gave white blocks, m.p. 158-159°C. (Found: $[M+H]^+$ 320.1179; $C_{14}H_{18}N_5O_2{}^{32}S$ requires $[M+H]^+$ 320.1181). δH (600MHz, $CDCl_3$) 7.32-7.29 (3H, m, ArH), 7.21 (2H, d, *J* 6.8, ArH), 5.60 (1H, s, H5), 5.46 (2H, s, NCH_2Ar), 3.14 (3H, s, NCH_3), 3.12 (3H, s, NCH_3), 2.26 (3H, s, CH_3). δC (150MHz, $CDCl_3$) 159.9, 158.6, 153.0, 134.1, 129.1, 128.6, 127.2, 98.8, 52.5, 37.6, 37.1, 12.5.



65f: Obtained as a colourless gum. (Found: $[M+H]^+$ 410.1641; $C_{21}H_{24}N_5O_2{}^{32}S$ requires $[M+H]^+$ 410.1651). δH (600MHz, $CDCl_3$) 7.30-7.27 (5H, m, ArH), 7.25-7.14 (5H, m, ArH), 5.44 (2H, s, NC*H*$_2$Ar), 3.74 (2H, s, CC*H*$_2$Ar), 3.16 (3H, s, NCH_3), 3.15 (3H, s, NCH_3), 2.15 (3H, s, CH_3). δC (150MHz, $CDCl_3$) 159.6, 158.7, 151.5, 139.2, 134.4, 129.0, 128.6, 128.5, 128.2, 127.3, 126.4, 110.3, 52.7, 37.5, 37.1, 28.0, 10.9.



66f: Recrystallisation from CH_2Cl_2 gave white blocks, m.p. 150-151^{o}C. (Found: $[M+Na]^+$ 342.1000; $C_{14}H_{20}N_5{}^{32}SNa$ requires $[M+Na]^+$ 342.1001). δH (600MHz, $CDCl_3$) 7.39-7.31 (3H, m, ArH), 7.23 (2H, d, *J* 6.8, ArH), 5.44 (1H, s, H5), 4.99 (2H, s, NC*H*$_2$Ar), 3.03 (6H, s, NCH_3 x 2), 2.22 (3H, s, CH_3). δC (150MHz, $CDCl_3$) 156.0, 153.3, 143.9, 133.8, 129.3, 128.6, 126.3, 94.2, 60.5, 56.2, 41.2, 14.3.

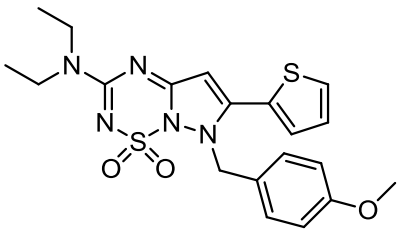


67f: Obtained as a colourless oil. δH (600MHz, $CDCl_3$) 7.43 (2H, d, *J* 7.5, ArH), 7.37-7.31 (3H, m, ArH), 5.41 (1H, s, H5), 4.84 (2H, s, NC*H*$_2$Ar), 2.17 (3H, s, CH_3). δC (150MHz, $CDCl_3$) 153.6, 148.3, 146.6, 134.8, 128.9, 128.3, 128.0, 92.3, 49.7, 14.4.

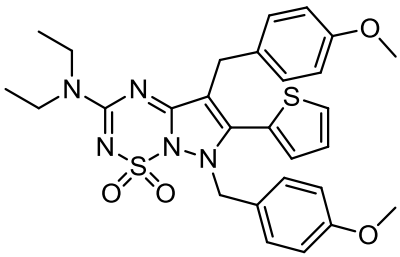
7-(4-Methoxybenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***64g****, 5,7-Bis(4-methoxybenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***65g,*** *4-(4-Methoxybenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***66g****, and 2-(4-Methoxybenzyl)-3-diethylamino-6-(thien-2-yl)pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***67g***

Method C (23h). A second wash with 10% aqueous sodium thiosulfate (5 mL) was included. Silica gel chromatography (CH_2Cl_2) gave four compounds: **65g** (14 mg, 10%) as a yellow semi-solid, **67g** (10 mg, 5%) as a colourless oil, **64g** (57 mg, 26%) as a colourless semi-solid, and **66g** (26 mg, 12%) as a beige powder followed by starting material (56 mg, 35%).

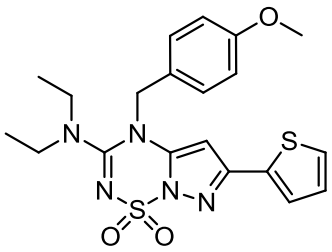
Method C (42h). A second wash with 10% aqueous sodium thiosulfate (5 mL) was included. Silica gel chromatography (CH_2Cl_2) gave four compounds: **65g** (10 mg, 8%) as a colourless gum, **67g** (8 mg, 4%) as a colourless oil, **64g** (98 mg, 44%) as a beige semi-solid, and **66g** (37 mg, 16%) as a beige powder. Further elution returned the starting material (42 mg, 26%).



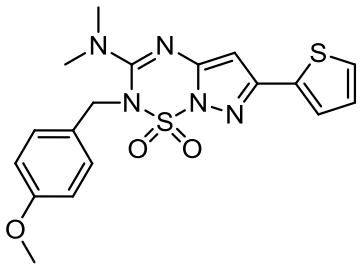
64g: Obtained as a colourless amorphous semi-solid. (Found: $[M+Na]^+$ 468.1125; $C_{20}H_{23}N_5O_3{}^{32}S_2Na$ requires $[M+Na]^+$ 468.1140). δH (400MHz, $CDCl_3$) 7.65 (1H, dd, *J* 5.1, 0.9, thienyl H5), 7.44 (1H, dd, *J* 2.7, 0.9, thienyl H3), 7.24 (1H, dd, *J* 5.0, 3.8, thienyl H4), 6.97 (2H, d, *J* 8.7, ArH), 6.72 (2H, d, *J* 8.7, ArH), 6.00 (1H, s, H5), 5.46 (2H, s, C*H*$_2$Ar), 3.74 (3H, s, OCH_3), 3.62-3.52 (4H, m, NC*H*$_2$$CH_3$ x 2), 1.23 (3H, t, *J* 7.0, NCH_2CH_3), 1.18 (3H, t, *J* 7.0, NCH_2CH_3). δC (100MHz, $CDCl_3$) 159.6, 151.4, 130.8, 130.8, 129.8, 128.7, 128.5, 128.2, 125.8, 113.8, 101.0, 55.2, 54.8, 42.6, 42.2, 13.4, 13.2.



65g: Obtained as a colourless amorphous gum. (Found: $[M+H]^+$ 566.1890; $C_{28}H_{32}O_4N_5{}^{32}S_2$ requires $[M+H]^+$ 566.1890). δH (600MHz, $CDCl_3$) 7.64 (1H, d, *J* 5.0, thienyl H5), 7.25 (1H, dd, *J* 3.6, thienyl H3), 7.21 (1H, dd, *J* 4.8, 3.8, thienyl H4), 6.90 (2H, d, *J* 8.6, ArH), 6.84 (2H, d, *J* 8.6, ArH), 6.71-6.69 (4H, m, ArH x 2), 5.29 (2H, s, CH_2Ar), 3.76 (3H, s, OCH_3), 3.75 (3H, s, OCH_3), 3.70 (2H, s, CH_2Ar), 3.56-3.53 (4H, m, NC$H_2$$CH_3$ x 2), 1.22 (3H, t, *J* 7.1, NCH_2CH_3), 1.10 (3H, t, *J* 7.1, NCH_2CH_3). δC (150MHz, $CDCl_3$) 161.8, 159.5, 158.1, 157.9, 149.7, 131.5, 131.1, 130.7, 130.2, 128.9, 128.3, 127.5, 125.6, 114.1, 113.7, 113.5, 55.4, 55.2, 55.1, 42.6, 42.4, 27.4, 13.31, 13.26.



66g: Recrystallisation from CH_2Cl_2 gave white needles, m.p. 76-77^{o}C. (Found: $[M+H]^+$ 446.1316; $C_{20}H_{24}O_3N_5{}^{32}S_2$ requires $[M+H]^+$ 446.1316). δH (600MHz, $CDCl_3$) 7.33 (1H, dd, *J* 3.6, 1.0, thienyl H3), 7.29 (1H, dd, *J* 5.04, 1.0, thienyl H5), 7.16 (2H, d, *J* 8.6, ArH), 7.00 (1H, dd, *J* 4.9, 3.8, thienyl H4), 6.88 (2H, d, *J* 8.6, ArH), 5.87 (1H, s, H5), 4.95 (2H, s, CH_2Ar), 3.78 (3H, s, OCH_3), 3.43 (4H, q, *J* 7.1, NC$H_2$$CH_3$ x 2), 1.22 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (150MHz, $CDCl_3$) 159.7, 155.6, 149.5, 143.8, 134.6, 127.8, 127.4, 126.5, 125.9, 125.2, 114.6, 91.6, 56.0, 55.3, 45.1, 12.8.



67g: Obtained as a colourless oil. (Found: $[M+Na]^+$ 468.1135; $C_{20}H_{24}O_3N_5{}^{32}S_2Na$ requires $[M+Na]^+$ 468.1135). δH (600MHz, $CDCl_3$, 40°C) 7.48 (1H, dd, *J* 3.6, 1.0, thienyl H3), 7.40 (1H, dd, *J* 5.1, 1.0, thienyl H5), 7.33 (2H, d, *J* 8.5, ArH), 7.12 (1H, dd, *J* 5.0, 3.7, thienyl H4), 6.98 (2H, d, *J* 8.5, ArH), 6.46 (1H, s, H5), 4.03 (2H, s, C*H*$_2$Ar), 3.83 (3H, s, OCH_3), 3.07 (4H, br-s, N*CH*$_2$$CH_3$ x 2), 1.04 (6H, br-s, NCH_2*CH*$_3$ x 2). δC (150MHz, $CDCl_3$, 40°C) 159.2, 147.7, 146.8, 136.6, 134.3, 130.5, 128.9, 127.4, 126.7, 126.6, 115.2, 99.8, 55.4, 43.0, 29.9, 13.0.

3-Diethylamino-4-methyl-6-phenyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***68*** *and 3-Diethylamino-6-phenyl-7-methyl-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***69***

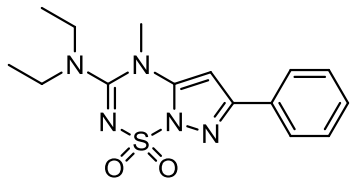
From methyl *p*-toluene sulfonate in MeCN: A stirred mixture of compound **34e** (82 mg, 0.25 mmol), methyl *p*-toluene sulfonate (71 mg, 0.375 mmol), Na_2CO_3 (40 mg, 0.375 mmol), and acetonitrile (1.5 mL) was heated at 55°C for 71 hours. The mixture was cooled to room temperature and then quenched with water (5 mL). The aqueous phase was extracted with CH_2Cl_2 (2 x 5 mL). The combined organic phases were dried and evaporated to afford a yellow semi-solid. Purification by column chromatography (CH_2Cl_2) gave, in order of elution, **69** (1 mg, 1%) as a colourless oil, and **68** (23 mg, 28%) as a white amorphous solid. Further elution with 10% MeOH in CH_2Cl_2 recovered starting material (52 mg, 64%).

From methyl *p*-toluene sulfonate in $CHCl_3$: A stirred mixture of compound **34e** (0.162 g, 0.5 mmol), methyl *p*-toluene sulfonate (0.140 g, 0.75 mmol) and Na_2CO_3 (81 mg, 0.76 mmol) in $CHCl_3$ (3 mL) was heated at 50°C for 48 hours. The mixture was cooled to room temperature and then quenched with water (5 mL). The aqueous phase was extracted with CH_2Cl_2 (5 mL). The combined organic phase was washed with saturated

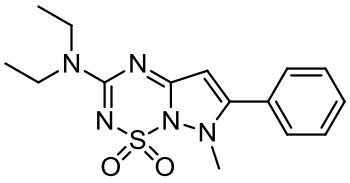
aqueous sodium chloride solution (5 mL), dried, and evaporated to afford a brown semi-solid. Purification by column chromatography (5% EtOAc in CH_2Cl_2) gave **69** (30 mg, 18%) as colourless prisms and **68** (4 mg, 2%) as a white amorphous solid. Further elution with 10% MeOH in CH_2Cl_2 recovered starting material (107 mg, 76%).

From dimethyl sulfate in $CHCl_3$: A stirred mixture of compound **34e** (81 mg, 0.25 mmol), Na_2CO_3 (41 mg, 0.38 mmol) and $CHCl_3$ (3 mL) was treated slowly with dimethyl sulfate (0.04 mL, 0.38 mmol). The mixture was heated at 50°C and stirred for 6 hours. The reaction was cooled to room temperature and treated with water (5 mL), stirred for 10 minutes, then extracted with CH_2Cl_2 (5 mL x 2). Extracts were combined and dried and the solvent was evaporated to leave an opaque oil. The residue was purified by column chromatography (CH_2Cl_2) to afford **69** (54 mg, 65%) as colourless prisms and **68** (25 mg, 30%) as white needles.

From dimethyl sulfate in MeCN: A mixture of compound **34e** (80 mg, 0.25 mmol), Na_2CO_3 (41 mg, 0.38 mmol) and acetonitrile (2 mL) was treated slowly with dimethyl sulfate (0.04 mL, 0.38 mmol). The mixture was heated at 55°C for 8 hours, then cooled to room temperature and treated with water (5 mL), stirred for 5 minutes then extracted with CH_2Cl_2 (5 mL x 2). Extracts were combined and dried, then solvent evaporated to give a yellow oil which was purified by column chromatography (CH_2Cl_2) to afford **69** (33 mg, 39%) as colourless prisms and **68** (51 mg, 60%) as white needles.



68: Recrystallisation from CH_2Cl_2 gave colourless wedges, m.p. 196-197°C. (Found: $[M+Na]^+$ 356.1154; $C_{15}H_{19}N_5O_2{}^{32}SNa$ requires $[M+Na]^+$ 356.1157). δH (400MHz, $CDCl_3$) 7.86 (2H, d, *J* 1.0, ArH), 7.40 (3H, m, ArH), 6.16 (1H, s, H5), 3.52 (3H, s, CH_3), 3.42 (4H, q, *J* 7.1, NC*H*$_2$ x 2), 1.28 (6H, t, *J* 7.1, NCH$_2$C*H*$_3$ x 2). δC (100MHz, $CDCl_3$) 155.2, 154.5, 144.8, 131.5, 129.1, 128.6, 126.3, 89.8, 44.7, 39.9, 12.7.



69: Recrystallisation from CH_2Cl_2 gave colourless blocks, m.p. 127-129°C. (Found: $[M+H]^+$ 334.1339; $C_{15}H_{20}N_5O_2{}^{32}S$ requires $[M+H]^+$ 334.1338). δH (400MHz, $CDCl_3$) 7.57-7.47 (5H, m, ArH), 5.93 (1H, s, H5), 3.86 (3H, s, NCH_3), 3.62-3.53 (4H, m, NCH_2CH_3 x 2), 1.24-1.18 (6H, m, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 157.9, 157.0, 154.5, 131.4, 129.5, 129.2, 127.0, 97.7, 42.8, 42.2, 37.7, 13.6, 13.5.

*General procedures for methylation of pyrazolo-thiadiazines **38:***

Method A

A mixture of **38** (0.5 mmol) and Na_2CO_3 (0.75 mmol) in acetonitrile (3 mL) was treated with dimethyl sulfate (0.075 mmol) and stirred at 65°C. The mixture was then cooled to room temperature, quenched with water (5 mL), extracted with CH_2Cl_2 (2 x 5 mL), dried and evaporated. The product(s) were separated by column chromatography over silica gel.

Method B

A mixture of **38** (0.5 mmol) and Na_2CO_3 (0.75mmol) in DMF (2.5 mL) was treated with dimethyl sulfate (0.075 mmol) and stirred at 50°C. The mixture was then cooled to room temperature and quenched with water (5 mL). Product was extracted with CH_2Cl_2 (2 x 5 mL) and the combined organic layers washed with 5% aqueous LiCl (5 mL) then dried and evaporated. The product(s) were separated by column chromatography over silica gel.

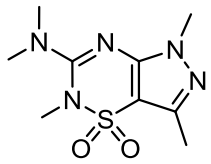
Method C

A mixture of **38** (0.5 mmol) and K_2CO_3 (1.5 mmol) in DMSO (2 mL) was treated slowly with dimethyl sulfate (1.5 mmol) then stirred at 80°C. The mixture was then cooled to room temperature and treated with water (5 mL), neutralised with 2M aqueous HCl and extracted with EtOAc (3 x 5 mL). The combined extracts were dried and evaporated. The product(s) were separated via column chromatography over silica gel.

Method D

A mixture of **38** (0.5 mmol) and K_2CO_3 (1.5 mmol) in acetonitrile (3 mL) was treated slowly with dimethyl sulfate (1.5 mmol) then stirred at reflux temperature. The mixture was then cooled to room temperature and treated with water (3 mL), stirred vigorously for 10 minutes and then extracted with CH_2Cl_2 (3 x 5 mL) or EtOAc (2 x 5mL). The combined extracts were dried and evaporated. The products were separated via column chromatography over silica gel.

3-Dimethylamino-2,5,7-trimethyl-2H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***70a***



Method A was employed at 65°C (42h) followed by elution with 40% EtOAc in CH_2Cl_2 to give the starting material (96 g, 78%) and the *title compound* (16 mg, 12%). R_F 0.3 (20% EtOAc in CH_2Cl_2).

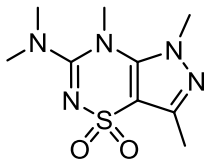
Method B was employed with **38b** at 50°C (19h) followed by elution with 20% EtOAc in CH_2Cl_2 to give the starting material (60 mg, 49%) and *the title compound* (61 mg, 47%) R_F 0.3 (20%EtOAc in CH_2Cl_2).

Method C was employed with **38b** at 80°C (26h) followed by elution with 40% EtOAc in CH_2Cl_2 to give *the title compound* (40 mg, 31%). R_F 0.6 (40%EtOAc in CH_2Cl_2).

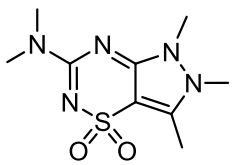
Recrystallisation of **70a** from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 152-153°C. (Found $[M+H]^+$ 258.1019; $C_9H_{16}N_5O_2{}^{32}S$ requires 258.1025). δH (400MHz, $CDCl_3$) 3.71 (3H, s, N5-CH_3), 3.17 (6H, s, NCH_3 x 2), 3.05 (3H, s, N2-CH_3), 2.40 (3H, s, C7-CH_3). δC (100MHz, $CDCl_3$) 157.3, 149.4, 141.5, 101.4, 38.6, 36.6, 33.6, 13.1.

3-Dimethylamino-2,5,7-trimethyl-2H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***70a****, 3-dimethylamino-4,5,7-trimethyl-4H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***71a*** *and 3-dimethylamino-5,6,7-trimethyl-5H-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***72a***

Method D was employed with **38b** at reflux temperature (38.5h) and extraction with EtOAc. Further extraction with 5:1 EtOAc:MeOH (5 mL) was required. Elution with 10% MeOH in CH_2Cl_2 provided compounds: i) **70a** (18 mg, 14%); ii) **71a** (19 mg, 15%); and iii) **72a** (84 mg, 65%).



71a: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless needles, m.p. 254-256°C dec. (Found $[M+H]^+$ 258.1023; $C_9H_{16}N_5O_2{}^{32}S$ requires 258.1025). δH (600MHz, $CDCl_3$): 3.71 (3H, s, N5-CH_3), 3.70 (3H, s, N4-NCH_3), 3.16 (6H, s, NCH_3 x 2), 2.59 (3H, s, CCH_3). δH (400MHz, DMSO-d_6) 3.86 (3H, s, N4-CH_3), 3.52 (3H, s, N5-NCH_3), 3.01 (6H, s, NCH_3 x 2), 2.23 (3H, s, CCH_3). δC (150MHz, DMSO-d_6): 156.2, 143.16, 139.98, 106.8, 40.3, 37.3, 36.5, 12.3.

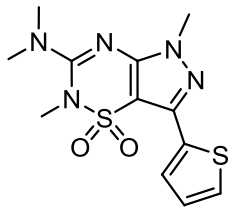


72a: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 293-295°C dec. (Found $[M+H]^+$ 258.1024; $C_9H_{16}N_5O_2{}^{32}S$ requires 258.1025). δH (400MHz,

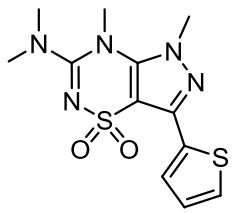
DMSO-d_6) 3.76 (3H, s, N6-CH_3), 3.70 (3H, s, N5-NCH_3), 3.01 (6H, s, NCH_3 x 2), 2.46 (3H, s, CCH_3). δC (100MHz, DMSO-d_6) 158.4, 154.1, 140.4, 101.4, 36.3, 32.0, 30.1, 10.8.

2,5-Dimethyl-3-dimethylamino- 7-(thien-2-yl)-2,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***70b****, 4,5-dimethyl-3-dimethylamino- 7-(thien-2-yl)-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***71b*** *and 5,6-dimethyl-3-dimethylamino- 7-(thien-2-yl)-5,6-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***72b***

Method D was employed with **38h** (95 mg, 0.31 mmol) at reflux temperature (20h) and extraction with CH_2Cl_2 (3 x 5 mL). Elution with 10%MeOH in CH_2Cl_2 provided: i) **70b** (30 mg, 30%); ii) **71b** (29 mg, 30%); and iii) **72b** (21 mg, 21%).

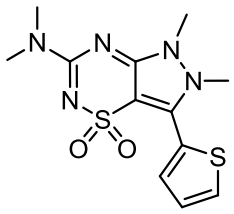


70b: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 174-175°C. (Found $[M+H]^+$ 326.0747; $C_{12}H_{16}N_5O_2{}^{32}S$ requires 326.0745). δH (400MHz, $CDCl_3$) 7.82 (1H, dd, *J* 3.7, 1.0, thienyl-H3), 7.30 (1H, dd, *J* 5.0, 0.9, thienyl-H5), 7.09 (1H, dd, *J* 5.0, 3.7, thienyl H4), 3.80 (3H, s, N5-CH_3), 3.20 (6H, s, NCH_3 x 2), 3.10 (3H, s, N2-CH_3). δC (100MHz, $CDCl_3$) 157.1, 150.0, 139.4, 133.7, 127.8, 127.8, 125.8, 99.4, 38.5, 37.0, 34.1.



71b: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 266-269°C dec. (Found $[M+H]^+$ 326.0745; $C_{12}H_{16}N_5O_2{}^{32}S$ requires 326.0745). δH (400MHz,

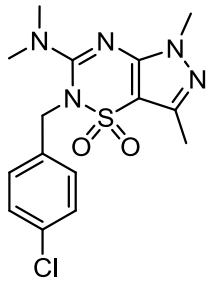
DMSO-d_6) 8.05 (1H, dd, *J* 3.6, 1.1, thienyl H3), 7.89 (1H, dd, *J* 5.1, 1.1, thienyl H5), 7.35 (1H, dd, *J* 5.1, 3.6, thienyl H4), 3.93 (3H, s, N5-CH_3), 3.80 (3H, s, NCH_3), 3.05 (6H, s, 2 x NCH_3). δC (100MHz, DMSO-d_6) 156.0, 144.3, 138.0, 133.0, 127.9, 127.7, 126.8, 105.0, 47.4, 40.7, 37.9.



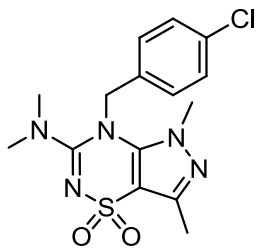
72b: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. 266-268^oC dec. (Found $[M+H]^+$ 326.0751; $C_{12}H_{16}N_5O_2{}^{32}S$ requires 326.0745). δH (400MHz, DMSO-d_6) 7.74 (1H, dd, *J* 3.6, 1.2, thienyl H3), 7.55 (1H, dd, *J* 5.1, 1.2 thienyl H5), 7.15 (1H, dd, *J* 5.1, 3.6, thienyl H4), 3.78 (3H, s, N5-CH_3), 3.41 (9H, br-s, N6-NCH_3 + NCH_3 x 2). δC (100MHz, DMSO-d_6) 149.7, 137.6, 134.2, 133.6, 127.8, 127.2, 126.0, 98.5, 53.0, 47.3, 33.7.

2-(4-Chlorobenzyl)-5,7-dimethyl-3-dimethylamino-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***70c,*** *4-(4-chlorobenzyl)-5,7-dimethyl-3-dimethylamino-pyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***71c*** *7a-(4-Chlorobenzyl)-3-(dimethylamino)-5,7-dimethyl-5,7a-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***73c***

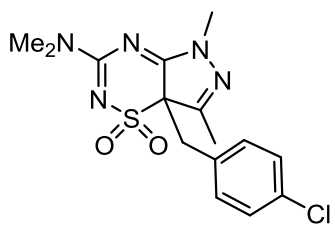
A mixture of compound **38b** (0.122 g, 0.5 mmol), 4-chlorobenzyl bromide (0.115 g, 0.55 mmol), K_2CO_3 (76 mg, 0.6 mmol) and $^nBu_4NBr$ (15 mg, 0.055 mol) in acetonitrile (4 mL) was stirred at 40^oC for 41 hrs. The mixture was then cooled to room temperature, diluted with CH_2Cl_2 (10 mL) then washed with water (2 x 5 mL), dried and evaporated down to a yellow semi-solid. The mixture was separated via column chromatography (5% EtOAc in CH_2Cl_2 to 60% EtOAc in CH_2Cl_2) to afford **70c** as white needles (87 mg, 47%) R_F 0.25 (5% EtOAc:CH_2Cl_2); **71c** as white prisms (9 mg, 5%) R_F 0.34 (5%EtOAc in CH_2Cl_2) and **73c** as white prisms (57 mg, 31%) R_F 0.20 (60%EtOAc:CH_2Cl_2).



70c: Recrystallisation from CH_2Cl_2 gave colourless needles, m.p. 154-156°C. (Found $[M+Na]^+$ 390.0778; $C_{15}H_{18}N_5O_6{}^{32}SClNa$ requires $[M+Na]^+$ 390.0767). δH (400MHz, $CDCl_3$) 7.11 (2H, d, *J* 8.4, ArH), 6.90 (2H, d, *J* 8.4, ArH), 4.58 (2H, s, NC*H*$_2$Ar), 3.42 (3H, s, N5-CH_3), 3.10 (6H, s, NC*H*$_3$ x 2), 2.36 (3H, s, C7-CH_3). δC (100MHz, $CDCl_3$) 155.1, 149.6, 140.9, 134.6, 131.6, 130.1, 128.2, 103.8, 53.4, 38.6, 33.4, 13.1. # An nOe correlation was observed between the methylene signal at δ 4.58 ppm and the dimethylamino protons at δ 3.10 ppm.



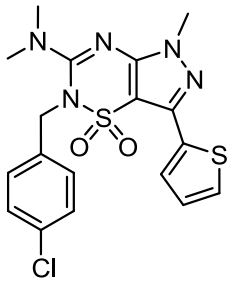
71c: Recrystallisation from CH_2Cl_2 gave colourless needles, m.p. 204-206 °C dec. (Found: $[M+Na]^+$ 390.0758; $C_{15}H_{18}N_5O_6{}^{32}SClNa$ requires $[M+Na]^+$ 390.0767). δH (400MHz, $CDCl_3$) 7.26 (2H, d, *J* 8.4, ArH), 7.13 (2H, d, *J* 8.4, ArH), 4.93 (2H, s, NC*H*$_2$Ar), 3.74 (3H, s, N5-CH_3), 3.07 (6H, s, 2 x NCH_3), 2.36 (3H, s, C7-CH_3). δC (100MHz, $CDCl_3$) 156.8, 134.2, 143.2, 142.1, 135.3, 131.7, 129.5, 129.1, 110.1, 56.7, 40.4, 37.2, 12.9. #Weak nOe correlations were observed between the methylene signal at δ 4.93 ppm and the N-methyl signals at δ 3.74 ppm and δ 3.07 ppm.



73c: Recrystallisation from CH_2Cl_2 gave white blocks, m.p. 178°C decomp. (Found $[M+H]^+$ 368.0950; $C_{15}H_{19}N_5O_2{}^{32}SCl$ requires $[M+H]^+$ 368.0948). δH (400MHz, $CDCl_3$) 7.23 (2H, d, *J* 8.4, ArH); 6.92 (2H, d, *J* 8.4, ArH); 3.49 and 3.35 (each 1H, ABq, J_{AB} 16.0, NC*H*$_2$-BnCl); 3.30 (3H, s, N5-CH_3); 3.21 (3H, s, NC*H*$_3$); 3.16 (3H, s, NC*H*$_3$); 2.37 (3H, s, C7-CH_3). δC (100MHz, $CDCl_3$) 172.1, 161.8, 157.9, 134.3, 130.8, 130.7, 128.7, 69.9, 38.5, 38.0, 37.1, 33.3, 14.9.

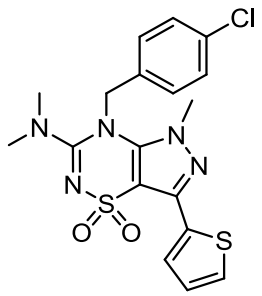
2-(4-Chlorobenzyl)-5-methyl-3-dimethylamino- pyrazolo[3,4-e][1,2,4]thiadiazine 7-(thien-2-yl) 1,1-dioxide ***70d****, 3-dimethylamino-4-(4-chlorobenzyl)-pyrazolo[3,4-e][1,2,4]thiadiazine 5-methyl, 7-(thien-2-yl) 1,1-dioxide* ***71d*** *and 7a-(4-Chlorobenzyl)-3-(dimethylamino)-5-methyl-7-(thein-2-yl)-5,7a-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide* ***73d***

A mixture of compound **38h** (0.133 g, 0.5 mmol), 4-chlorobenzyl bromide (0.115 g, 0.55 mmol), K_2CO_3 (78 mg, 0.6 mmol) and $^nBu_4NBr$ (15 mg, 0.55 mmol) in acetonitrile (4 mL) was stirred at room temperature for 16 hrs. The reaction was heated to 60°C for a further 24 hrs. The mixture was then cooled to room temperature, diluted with CH_2Cl_2 (10 mL) then washed with water (2 x 5 mL), dried and evaporated to a yellow oil. The mixture was separated via column chromatography to afford **70d** as white needles (0.146 g, 67%) R_F 0.74 (CH_2Cl_2); **71d** as white prisms (34 mg, 16%) R_F 0.20 (60%EtOAc in CH_2Cl_2); and **73d** as yellow prisms (16 mg, 7%) R_F 0.50 (CH_2Cl_2).

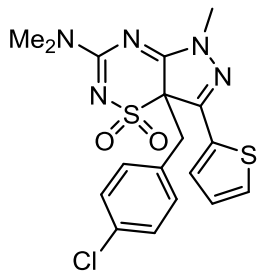


70d: Recrystallisation from CH_2Cl_2 as colourless needles, m.p. 134-135 °C. (Found $[M+Na]^+$ 458.0490; $C_{18}H_{18}N_5O_2{}^{32}S_2ClNa$ requires $[M+Na]^+$ 458.0488). δH (600MHz, $CDCl_3$) 7.84 (1H, dd, *J* 3.7, 1.1, thienyl-H5), 7.32 (1H, dd, *J* 4.0, 1.1, thienyl-H3), 7.13-7.11 (3H, m, ArH & thienyl-H4), 6.94 (2H, d, *J* 8.4, ArH), 4.67 (2H, s, NC*H*$_2$Ar), 3.53

(3H, s, N5-CH_3), 3.16 (6H, s, 2 x NCH_3). δC (150MHz, $CDCl_3$) 155.0, 150.2, 138.9, 134.8, 133.7, 131.4, 130.2, 128.4, 128.0, 126.1, 101.9, 53.9, 38.7, 33.9. #An nOe correlation was observed between the methylene signal at δ 4.67ppm and the dimethylamino protons at δ 3.16 ppm.



71d: Recrystallisation from CH_2Cl_2 as colourless needles, m.p. 208-210 °C (dec.). (Found: $[M+Na]^+$ 458.0486. $C_{18}H_{18}N_5O_2{}^{32}S_2ClNa$ requires $[M+Na]^+$ 458.0488). δH (600MHz, $CDCl_3$) 7.91 (1H, dd, *J* 3.7, 1.0, thienyl-H5), 7.27 (1H, dd, *J* 4.5, 1.0, thienyl-H3), 7.24 (2H, d, *J* 8.5, ArH), 7.11 (2H, d, *J* 8.5, ArH), 7.05 (1H, dd, *J* 5.0, 1.3, thienyl-H4), 4.91 (2H, s, NC*H*$_2$Ar), 3.77 (3H, s, N5-CH_3), 3.06 (6H, s, NC*H*$_3$ x 2). δC (150MHz, $CDCl_3$) 156.2, 143.0, 140.5, 135.2, 132.7, 131.5, 129.5, 129.2, 129.1, 128.1, 126.3, 107.9, 56.9, 40.3, 37.7. #An nOe correlation was observed between the CH_2 peaks at δ 4.91 ppm and the methyl protons on the pyrazole at δ 3.77 ppm as well as the dimethylamino substituent at δ 3.06 ppm.



73d: Recrystallisation from CH_2Cl_2 as yellow blocks, m.p. 198 °C (dec.). (Found: $[M+Na]^+$ 458.0488. $C_{18}H_{18}N_5O_2{}^{32}S_2ClNa$ requires $[M+Na]^+$ 458.0488). δH (600MHz, $CDCl_3$) 8.28 (1H, dd, *J* 3.8, 0.9, thienyl-H5), 7.52 (1H, dd, *J* 5.0, 0.9, thienyl-H3), 7.21 (1H, dd, *J* 5.0, 1.1, thienyl-H4), 7.15 (2H, d, *J* 8.4, ArH), 6.70 (2H, d, *J* 8.4, ArH), 3.56 (2H, ABq, J_{AB} 13.9, NCH_AH_BAr), 3.34 (3H, s, N5-CH_3), 3.29 (3H, s, NCH_3), 3.26 (3H,

s, NCH_3). δC (150MHz, $CDCl_3$) 170.8, 161.2, 152.4, 134.3, 133.3, 132.3, 130.7, 130.6, 129.0, 128.6, 69.9, 41.0, 38.2, 37.3, 33.6.

9.10 Synthesis of N4-alkylated pyrazolo[1,5-*b*][1,2,4,6] thiatriazines 74, N7-alkylated pyrazolo[1,5-*b*][1,2,4,6] thiatriazines 75, and 7,8-dihydropyrimido[1,6-*b*][1,2,4,6] thiatriazines 76

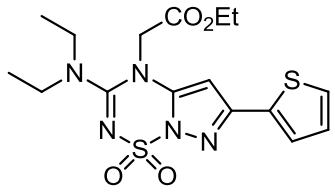
General procedure:

A mixture of compound **34** (0.5 mmol), $^{n}Bu_4NBr$ (0.1 mmol), electrophile (1 mmol), K_2CO_3 (1 mmol), and acetonitrile (3 mL) was stirred at 50°C for 16-48 hours. The reaction mixture was cooled to room temperature and evaporated. The residue was diluted with CH_2Cl_2 (10 mL) and washed with water (5 mL). The aqueous phase was extracted with CH_2Cl_2 (5 mL). The combined organic extracts were dried and evaporated and the residue was chromatographed over silica gel. Products were recrystallised as described below.

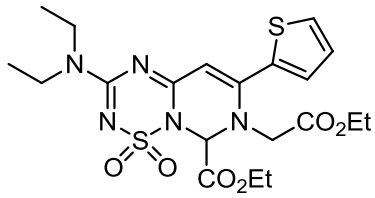
The following compounds were prepared using the above procedure. All products are listed in order of elution from chromatography.

Ethyl 2-(3-diethylamino-1,1-dioxo-6-(thien-2-yl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazin-4-yl)acetate ***74a*** *and Ethyl 3-diethylamino-7-(2-ethoxy-2-oxoethyl)-6-(thien-2-yl)-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine-8-carboxylate 1,1-dioxide* ***76a***

Reaction at 80°C for 6 h; purification (SiO_2; 10% EtOAc in CH_2Cl_2) gave two products: **74a** (62 mg, 60%) as a yellow gum and **76a** (32 mg, 25%) as white flakes.



74a: Recrystallisation from 1:1 CH_2Cl_2:EtOAc gave white needles, m.p. 151-152°C. (Found: $M^{+\bullet}$ 411.1023; $C_{16}H_{21}N_5O_4{}^{32}S_2$ requires $M^{+\bullet}$ 411.1029). δH (600MHz, $CDCl_3$) 7.43 (1H, dd, *J* 3.7, 1.0, thienyl-H5), 7.34 (1H, dd, *J* 5.0, 3.7, thienyl-H3), 7.05 (1H, dd, *J* 3.7, 1.0, thienyl-H4), 6.00 (1H, s, H5), 4.47(2H, s, C*H*$_2$CO$_2$Et), 4.34 (2H, q, *J* 7.2, CO$_2$C*H*$_2$CH$_3$) 3.41 (4H, q, *J* 7.2, NC*H*$_2$CH$_3$ x 2), 1.31 (3H, t, *J* 7.2, CO$_2$CH$_2$C*H*$_3$), 1.27 (6H, t, *J* 7.2, NCH$_2$C*H*$_3$ x 2). δC (150MHz, $CDCl_3$) 166.6, 155.2, 149.5, 143.7, 134.1, 127.2, 126.4, 125.8, 90.7, 62.6, 54.1, 44.6, 13.8, 12.4. *m/z* (EI) 411/412/543 (26/6%, $M^{+\bullet}$), 221 (13), 148 (5), 108/109/110 (100/9/5).



76a: Obtained as a yellow semi-solid. Recrystallization from 1:1 CH_2Cl_2:light petroleum gave yellow prisms, m.p. 56-58.5°C. (Found: $[M+Na]^+$ 498.1473; $C_{20}H_{27}N_5O_6{}^{32}S_2$ requires $[M+Na]^+$ 498.1481). δH (600MHz, $CDCl_3$) 7.56 (1H, d, *J* 5.0, thienyl-H5), 7.46 (1H, d, *J* 3.7, thienyl-H3), 7.15 (1H, dd, *J* 3.9, 0.9, thienyl-H4), 6.01 (1H, s, H5), 4.26-4.20 (4H, m, C*H*$_2$CO$_2$C*H*$_2$CH$_3$), 3.69-3.53 (4H, m, NC*H*$_2$CH$_3$ x 2), 1.26 (3H, t, *J* 4.3, CO$_2$CH$_2$C*H*$_3$), 1.22 (6H, m, NCH$_2$C*H*$_3$ x 2). δC (150MHz, $CDCl_3$) 168.0, 165.9, 157.7, 155.6, 151.4, 135.3, 131.0, 130.6, 128.6, 103.6, 68.4, 63.0, 62.0, 53.6, 42.5, 42.4, 14.1, 14.0, 13.7, 12.8.

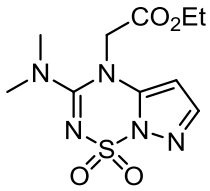
Ethyl 2-(3-dimethylamino-1,1-dioxo-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazin-4-yl)acetate ***74b*** *and Ethyl 2-(3-dimethylamino-1,1-dioxo-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazin-7-yl)acetate* ***75b*** *and Ethyl 3-dimethylamino-7-(2-ethoxy-2-oxoethyl)-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine-8-carboxylate 1,1-dioxide* ***76b***

Method A

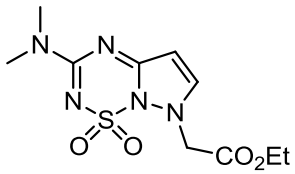
Reaction at 50°C for 24 h; purification (SiO_2; 15% EtOAc in CH_2Cl_2) furnished three products: **76b** (55 mg, 31%) as a colourless oil, **75b** (6 mg, 4%) as a colourless gum, and **74b** (36 mg, 24%) as colourless blocks. Further elution gave starting material (21 mg, 19%).

Method B

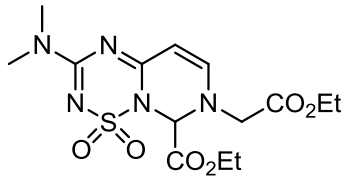
Reaction at 80°C for 16 h; purification (SiO_2; 10% EtOAc in CH_2Cl_2) gave **74b** (38 mg, 25%) as colourless blocks.



74b: Recrystallisation from 1:1 EtOAc:CH_2Cl_2 gave colourless prisms, m.p. 164-165°C. (Found: $[M+Na]^+$ 324.0746; $C_{10}H_{15}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 324.0742). δH (400MHz, $CDCl_3$) 7.67 (1H, d, *J* 1.8, H6), 5.80 (1H, d, *J* 1.8, H5), 4.47 (2H, s, CH_2), 4.29 (2H, q, *J* 7.1, O*CH*$_2$CH$_3$), 3.08 (6H, s, NCH_3 x 2), 1.28 (3H, t, *J* 7.1, OCH_2*CH*$_3$). δC (100MHz, $CDCl_3$) 166.8, 155.8, 142.9, 142.8, 93.1, 62.8, 54.1, 40.7, 14.0.



75b: Isolated as a colourless oil. (Found $[M+Na]^+$ 324.0749; $C_{10}H_{15}N_5O_4{}^{32}SNa$ requires 324.0742). δH (600MHz, $CDCl_3$) 7.62 (1H, d, *J* 2.0, H6), 5.72 (1H, d, *J* 2.0, H5), 4.44 (2H, s, CH_2), 4.25 (2H, q, *J* 7.1, O*CH*$_2$CH$_3$), 3.59 (3H, br-s, NCH_3), 3.27 (3H, br-s, NCH_3), 1.28 (3H, t, *J* 7.1, OCH_2*CH*$_3$). δC (150MHz, $CDCl_3$) 166.8, 148.2, 146.0, 143.5, 91.5, 62.1, 47.1, 42.0, 41.1, 14.1.

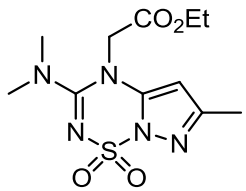


76b: Recrystallisation from CH_2Cl_2 gave colourless prisms, m.p. 93-94.5°C. (Found: $[M+Na]^+$ 410.1102; $C_{14}H_{21}N_5O_6{}^{32}SNa$ requires 410.1110). δH (400MHz, $CDCl_3$) 6.86 (1H, dd, *J* 7.3, 1.20, H6), 5.94 (1H, d, *J* 1.1, H8), 5.24 (1H, d, *J* 7.3, H5), 4.33 (1H, d, *J* 17.7, C*H*$_A$H$_B$CO$_2$Et), 4.23-4.11 (4H, m, OC*H*$_2$CH$_3$ x 2), 4.08 (1H, d, *J* 17.7, CH$_A$*H*$_B$CO$_2$Et), 3.20 (3H, s, NCH$_3$), 3.11 (3H, s, NCH$_3$), 1.29 (3H, t, *J* 7.2, OCH$_2$C*H*$_3$), 1.27 (3H, t, *J* 7.2, OCH$_2$C*H*$_3$). δC (100MHz, $CDCl_3$) 167.5, 165.8, 158.7, 155.3, 147.2, 95.6, 66.5, 63.0, 62.3, 55.1, 37.3, 37.2, 14.0, 13.9.

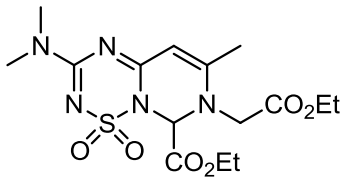
Ethyl 2-(3-dimethylamino-6-methyl-1,1-dioxo-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazin-4-yl)acetate ***74c*** *and Ethyl 3-dimethylamino-7-(2-ethoxy-2-oxoethyl)-6-methyl-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine-8-carboxylate 1,1-dioxide* ***76c***

Chromatography (SiO_2; 40% EtOAc in CH_2Cl_2) gave two compounds: **76c** (81 mg, 40%) as a white powder and **74c** (38 mg, 24%) as white needles.

A similar reaction, employing *N*,*N*'-diisopropylethylamine (1.1 mmol) in place of K_2CO_3, afforded, after purification as above: **76c** (66 mg, 33%) as a white powder and **74c** (62 mg, 40%) as white needles.



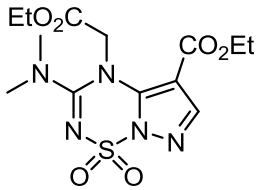
74c: Recrystallisation from CH_2Cl_2 gave white needles, m.p. 157-158°C. (Found: $[M+Na]^+$ 338.0895; $C_{11}H_{17}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 338.0899). δH (400MHz, $CDCl_3$) 5.63 (1H, s, H5), 4.46 (2H, s, NC*H*$_2$CO), 4.35 (2H, q, *J* 7.2, OC*H*$_2$CH$_3$), 3.10 (6H, s, NC*H*$_3$ x 2), 2.34 (3H, s, C*H*$_3$), 1.34 (3H, t, *J* 7.2, OCH$_2$C*H*$_3$). δC (100MHz, $CDCl_3$) 166.9, 155.7, 153.3, 143.7, 93.2, 62.8, 53.9, 40.6, 14.2, 14.0.



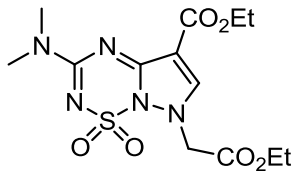
76c: Recrystallisation from CH_2Cl_2 gave white flakes, m.p. 88-90°C. (Found: $[M+H]^+$ 402.1448; $C_{15}H_{24}N_5O_6{}^{32}S$ requires $[M+H]^+$ 402.1447). δH (400MHz, $CDCl_3$) 5.92 (1H, s, H8), 5.33 (1H, s, H5), 4.33-4.20 (6H, m, C*H*$_2$CO$_2$Et + C*H*$_2$CH$_3$ x 2), 3.22 (3H, s, NCH$_3$), 3.12 (3H, s, NCH$_3$), 2.10 (3H, s, CH$_3$), 1.31-1.23 (6H, m, CH$_2$C*H*$_3$ x 2). δC (100MHz, $CDCl_3$) 167.3, 166.3, 159.0, 156.7, 155.6, 154.9, 96.7, 67.9, 62.9, 62.4, 52.1, 37.1, 19.3, 14.0, 14.0.

Ethyl 3-dimethylamino-4-(2-ethoxy-2-oxoethyl)-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazine-5-carboxylate 1,1-dioxide ***74d****, Ethyl 3-dimethylamino-7-(2-ethoxy-2-oxoethyl)-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazine-5-carboxylate 1,1-dioxide* ***75d*** *and Diethyl 3-dimethylamino-7-(2-ethoxy-2-oxoethyl)-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine-5,8-dicarboxylate 1,1-dioxide* ***76d***

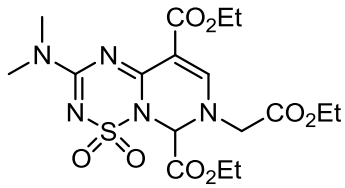
Chromatography (SiO_2; 15% EtOAc in CH_2Cl_2) gave 3 compounds: **75d** (2 mg, 1%) as a yellow oil, **74d** (21 mg, 12%) as a yellow gum, and **76d** (82 mg, 34%) as a white powder.



74d: Recrystallisation from 1:1 EtOAc:CH_2Cl_2 as colourless prisms, m.p 136-137.5°C. (Found: $[M+Na]^+$ 396.0945; $C_{13}H_{19}N_5O_6{}^{32}SNa$ requires 396.0954). δH (400MHz, $CDCl_3$) 7.99 (1H, s, H6), 4.95 (2H, s, C*H*$_2$CO$_2$Et), 4.29 (2H, q, *J* 7.2, C5CO$_2$C*H*$_2$CH$_3$), 4.14 (2H, q, *J* 7.1, CH$_2$CO$_2$C*H*$_2$CH$_3$), 3.13 (6H, s, NCH$_3$ x 2), 1.36 (3H, t, *J* 7.2, 5-CO$_2$CH$_2$C*H*$_3$), 1.22 (3H, t, *J* 7.1, CH$_2$CO$_2$CH$_2$C*H*$_3$). δC (100MHz, $CDCl_3$) 168.1, 161.4, 156.8, 143.3, 142.9, 102.3, 62.4, 61.1, 54.4, 40.6, 14.2, 13.9.



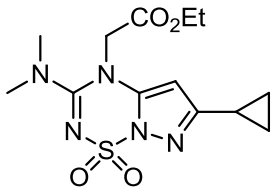
75d: Obtained as a yellow oil. δH (400MHz, $CDCl_3$) 8.01 (1H, s, H6), 5.07 (2H, s, CH_2), 4.33 (2H, q, *J* 7.1, C*H*$_2$CH$_3$), 4.13 (2H, q, *J* 7.2, C*H*$_2$CH$_3$), 3.37 (6H, s, NC*H*$_3$), 1.36 (3H, t, *J* 7.1, CH$_2$*CH*$_3$), 1.23 (3H, t, *J* 7.2, CH$_2$*CH*$_3$). δC (100MHz, $CDCl_3$) 168.7, 160.9, 149.2, 146.8, 143.8, 103.9, 61.7, 61.1, 50.3, 14.2, 14.0.



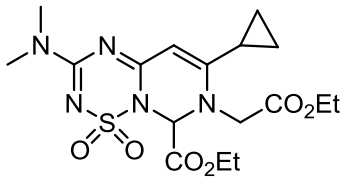
76d: Recrystallisation from CH_2Cl_2 gave yellow prisms, m.p 158-160^{o}C. (Found: $[M+Na]^+$ 482.1314; $C_{17}H_{25}N_5O_8{}^{32}SNa$ requires 482.1322). δH (400MHz, $CDCl_3$) 8.01 (1H, d, *J* 1.1, H6); 6.07 (1H, d, *J* 1.1, H8), 4.50 (1H, d, *J* 17.7, C*H*$_A$H$_B$CO$_2$Et), 4.33-4.21 (7H, m, O*CH*$_2$CH$_3$ x 3 + CH$_A$*H*$_B$CO$_2$Et), 3.33 (3H, s, NCH$_3$), 3.14 (3H, s, NCH$_3$), 1.31 (6H, m, OCH$_2$*CH*$_3$ x 2), 1.27 (3H, t, *J* 7.2, OCH$_2$*CH*$_3$). δC (100MHz, $CDCl_3$) 166.4, 165.1, 163.1, 158.7, 154.8, 152.4, 98.3, 66.2, 63.5, 62.9, 60.6, 55.9, 37.4, 37.1, 14.3, 14.0, 13.9.

Ethyl 2-(6-cyclopropyl-3-dimethylamino-1,1-dioxo-4H-pyrazolo[1,5-b][1,2,4,6]thiatriazin-4-yl)acetate ***74e*** *and Ethyl 6-cyclopropyl-3-dimethylamino-7-(2-ethoxy-2-oxoethyl)-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine-8-carboxylate 1,1-dioxide* ***76e***

Chromatography (SiO_2; 50% EtOAc in CH_2Cl_2) afforded two compounds: **76e** (65 mg, 31%) as off-white flakes and **74e** (69 mg, 40%) as a white powder.

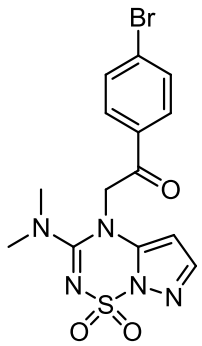


74e: Recrystallisation from CH_2Cl_2 gave white needles, m.p. 109-110°C. (Found: $[M+Na]^+$ 364.1062. $C_{13}H_{19}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 364.1055). δH (400MHz, $CDCl_3$) 5.46 (1H, s, H5), 4.42 (2H, s, NCH_2CO), 4.28 (2H, q, *J* 7.2, OCH_2CH$_3$), 3.03 (6H, s, NCH$_3$ x 2), 1.94-1.87 (1H, m, CH(CH_2CH$_2$)), 1.28 (3H, t, *J* 7.2, OCH$_2$CH_3), 0.94-0.91 (2H, m, C*H*(CH$_2$CH$_2$)), 0.83-0.81 (2H, m, CH(CH$_2$CH_2)). δC (100MHz, $CDCl_3$) 166.9, 159.5, 155.6, 143.7, 90.5, 90.4, 62.7, 53.8, 40.6, 40.6, 14.0, 9.6, 8.4.



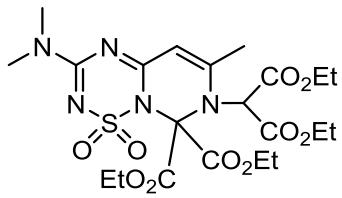
76e: Recrystallisation from CH_2Cl_2 gave white flakes, m.p. 106-108.5°C. (Found: $[M+Na]^+$ 450.1416. $C_{17}H_{25}N_5O_6{}^{32}SNa$ requires $[M+Na]^+$ 450.1423). δH (400MHz, $CDCl_3$) 5.92 (1H, s, H8), 5.07 (1H, s, H5), 4.58,4.32 (2H, ABq, J_{AB} 17.8, C$H_A$$H_BCO_2$Et), 4.28-4.15 (4H, m, OCH_2CH$_3$ x 2), 3.18 (3H, s, NCH$_3$), 3.10 (3H, s, NCH$_3$), 1.55-1.48 (1H, m, C*H*(CH$_2$CH$_2$)), 1.29-1.23 (6H, m, OCH$_2$CH_3 x 2), 1.06-0.91 (2H, m, CH(CH_2CH$_2$)), 0.63-0.58 (2H, m, CH(CH$_2$CH_2)). δC (100MHz, $CDCl_3$) 167.7, 166.2, 161.4, 159.0, 156.2, 93.3, 68.0, 62.8, 62.1, 52.2, 37.3, 37.1, 14.0, 14.0, 12.6, 9.2, 5.3.

1-(4-Bromophenyl)-2-(2-dimethylamino-4,4-dioxo-1H-pyrazolo[1,5-b][1,2,4]thiadiazin-1-yl)ethanone ***74f***



Chromatography (SiO_2; 30% EtOAc in CH_2Cl_2) afforded the *title compound* (28 mg, 18%) as an orange semi-solid. Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave orange wedges, m.p. 183-185°C dec. (Found $[M+Na]^+$ 433.9901; $C_{14}H_{14}N_5O_3{}^{32}S^{79}BrNa$ requires $[M+Na]^+$ 433.9898). δH (400MHz, DMSO-d_6) 7.99 (2H, d, *J* 6.8, ArH), 7.83 (2H, d, *J* 6.8, ArH), 7.73 (1H, d, *J* 1.9, H6), 6.04 (1H, d, *J* 1.9, H5), 5.69 (2H, s, C*H*$_2$COAr), 2.98 (6H, s, NCH_3 x 2). δC (100MHz, DMSO-d_6) 192.7, 155.6, 144.2, 143.4, 133.4, 132.4, 130.8, 129.1, 94.1, 59.0, 40.8.

Dimethyl 7-(1,3-dimethoxy-1,3-dioxopropan-2-yl)-3-dimethylamino-6-methylpyrimido[1,6-b][1,2,4,6]thiatriazine-8,8(7H)-dicarboxylate 1,1-dioxide ***76g***



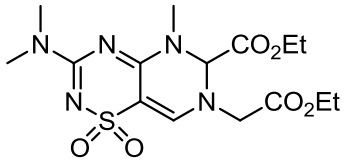
Chromatography (SiO_2; 40% EtOAc in CH_2Cl_2) gave the *title compound* (62 mg, 25%) as a white powder and the starting material (55 mg, 47%). Recrystallisation from CH_2Cl_2 gave white flakes, m.p. 201-203°C dec. (Found: $[M+Na]^+$ 512.1065; $C_{17}H_{23}N_5O_{10}{}^{32}SNa$ requires $[M+Na]^+$ 512.1063). δH (400MHz, $CDCl_3$) 9.65 (1H, s, H5), 4.90 (1H, s, NC*H*(CO_2CH_3)$_2$), 3.81 (6H, s, CO_2CH_3 x 2), 3.64 (6H, s, CO_2CH_3 x 2), 3.20 (6H, s, NCH_3 x 2), 2.04 (3H, s, CH_3). δC (100MHz, $CDCl_3$) 168.1, 167.9, 150.1, 148.9, 138.0, 95.4, 57.7, 57.2, 54.1, 53.4, 50.0, 13.3.

9.11 Synthesis of N4-alkylated pyrazolo[3,4-*e*][1,2,4] thiadiazines 79, N6-alkylated pyrazolo[3,4-*e*][1,2,4] thiadiazines 78, 6,7-dihydropyrimido[4,5-*e*][1,2,4] thiadiazines 77 and N2-alkylated pyrazolo[3,4-*e*][1,2,4] thiadiazines 80

General procedure:

A mixture of compound **38** (0.5 mmol or 0.25 mmol), $^{n}Bu_4NBr$ (2 equiv), electrophile (2 equiv), K_2CO_3 (2 equiv), and DMF or MeCN (3 mL) was stirred at 50-55°C for 27-69 hrs. The reaction mixture was cooled to room temperature and an extractive workup was performed as described below. The product(s) were then separated by chromatography over buffered silica gel.

Ethyl 3-(dimethylamino)-7-(2-ethoxy-2-oxoethyl)-5-methyl-6,7-dihydro-5H-pyrimido[4,5-e][1,2,4]thiadiazine-6-carboxylate 1,1-dioxide ***77a***

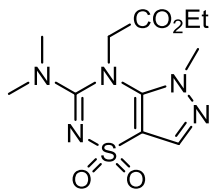


Compound **38a** (0.5 mmol) and ethyl 2-bromoacetate (1.05 mmol) (50°C;27h; DMF). The mixture was cooled to room temperature and diluted with CH_2Cl_2 (5 mL) then washed with water (5 mL) and extracted with CH_2Cl_2 (2 x 5 mL). The combined extracts were dried and solvent evaporated to an orange/red oil. Elution with 40% EtOAc in CH_2Cl_2 gave the *title compound* as a yellow amorphous solid (62 mg, 39%) R_F 0.24 (40% EtOAc:CH_2Cl_2).

(Found: $[M+Na]^+$ 424.1257; $C_{15}H_{23}N_5O_6{}^{32}SNa$ requires $[M+Na]^+$ 424.1267). δH (600MHz, $CDCl_3$): 7.66 (1H, s, H8), 5.24 (1H, s, H6), 4.43 (1H, ABq, J_{AB} 18.0, C*H*$_2$CO$_2$Et), 4.29-4.20 (5H, m, O-C*H*$_2$ x 2, C*H*$_2$CO$_2$Et), 3.30 (3H, s, N6C*H*$_3$), 3.19 (3H, s, NCH$_3$), 3.13 (3H, s, NCH$_3$), 1.30 (6H, m, CH$_2$C*H*$_3$ x 2). δC (600MHz, $CDCl_3$): 167.6, 167.3, 159.0, 156.1, 146.4, 101.2, 73.1, 63.5, 62.6, 55.1, 37.6, 37.2, 35.8, 14.2.

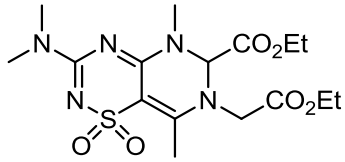
Ethyl 2-(3-(dimethylamino)-5-methyl-pyrazolo[3,4-e][1,2,4]thiadiazin-4(5H)-yl)acetate 1,1-dioxide ***79a*** *and Ethyl 3-(dimethylamino)-7-(2-ethoxy-2-oxoethyl)-5-methyl-6,7-dihydro-5H-pyrimido[4,5-e][1,2,4]thiadiazine-6-carboxylate 1,1-dioxide* ***77a***

Compound **38a** (0.25 mmol) and ethyl 2-bromoacetate (0.5 mmol) (55^{o}C;50h; acetonitrile). The mixture was then cooled to room temperature, and the solvent evaporated. The residue was diluted with CH_2Cl_2 (15 mL) and filtered to remove K_2CO_3. The filtrate was dried and evaporated in vacuo to a yellow semi-solid. Elution with 60% EtOAc in CH_2Cl_2 afforded two compounds: i) **77a** (31 mg, 39%) R_F 0.25 (60% EtOAc in CH_2Cl_2); and ii) **79a** (8 mg, 11%) R_F 0.15 (60% EtOAc in CH_2Cl_2).



Recrystallisation of **79a** from CH_2Cl_2 gave off-white flakes, m.p. 161^{o}C decomp. (Found $[M+Na]^+$ 338.0891; $C_{11}H_{17}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 338.0899). δH (400MHz, $CDCl_3$) 7.78 (1H, s, H7), 4.50 (2H, s, CH_2), 4.18 (2H, q, *J* 7.1, C*H*$_2$CH$_3$), 3.88 (3H, s, N5-CH_3), 3.06 (6H, s, 2 x NCH_3), 1.19 (3H, t, *J* 7.1, CH_2CH_3). δC (100MHz, $CDCl_3$) 166.1, 156.1, 141.9, 133.2, 110.9, 63.1, 53.6, 40.5, 37.7, 14.1. #A weak nOe interaction was observed between the methylene signal at δ 4.50 ppm and the dimethylamino proton signal at δ 3.06 ppm.

7-Carbethoxy-6-ethylacetoxy-5,8-dimethyl-3-dimethylamino, diazino [1,5-b][1,2,4,6] thiatriazine 1,1-dioxide ***77b***



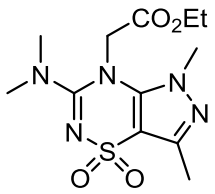
Compound **38b** (0.5 mmol) and ethyl 2-bromoacetate (0.57 mmol) (50^{o}C;40h; DMF). The mixture was then cooled to room temperature, diluted with water (10 mL) then

product extracted with EtOAc (2 x 10 mL) and washed with 10% aqueous LiCl (2 x 15 mL), then dried and evaporated to give a brown residue. Elution with 40% EtOAc in CH_2Cl_2 afforded the *title compound* (0.126 g, 61%) R_F 0.30 (40%EtOAc:CH_2Cl_2).

Recrystallisation from CH_2Cl_2 gave off-white flakes, m.p. 157-159 °C (dec.). (Found $[M+Na]^+$ 438.1425; $C_{16}H_{25}N_5O_6{}^{32}SNa$ requires $[M+Na]^+$ 438.1423). δH (400MHz, $CDCl_3$) 5.06 (1H, s, H6), 4.50, 4.17 (2H, ABq, J_{AB} 18.04, C*H*$_A$*H*$_B$CO$_2$Et), 4.22 (4H, m, C*H*$_2$CH$_3$ x 2), 3.25 (3H, s, N5-CH$_3$), 3.16 (6H, s, 2 x NCH$_3$), 2.62 (3H, s, C7-CH$_3$), 1.33-1.26 (6H, m, CH$_2$C*H*$_3$ x 2). δC (100MHz, $CDCl_3$): 167.8, 167.4, 158.0, 157.9, 157.3, 101.4, 74.3, 63.3, 62.6, 51.4, 37.2, 36.0, 17.0, 14.3, 14.2. #nOe interactions are present between the pyrazole N-CH$_3$ protons and the proton at δ 5.06 ppm. There is also an nOe between the AB quartet and the pyrazole C7 methyl protons.

Ethyl 2-(3-(dimethylamino)-5,7-dimethyl-pyrazolo[3,4-e][1,2,4]thiadiazin-4(5H)-yl)acetate 1,1-dioxide ***79b*** *and 7-carbethoxy-5,8-dimethyl, 3-dimethylamino- 6-ethylacetoxy, diazino [1,5-b][1,2,4,6] thiatriazine 1,1-dioxide* ***77b***

Compound **38b** (0.6 mmol) and ethyl 2-bromoacetate (1.2 mmol) (55^{o}C;48h; DMF). The mixture was diluted with water (5 mL) and extracted with CH_2Cl_2 (2 x 5 mL). The combined organic phase was washed with 1M aqueous HCl (2 x 3 mL), dried and concentrated *in vacuo* to give an orange residue. Elution with 40% EtOAc in CH_2Cl_2 gave two compounds: i) **77b** (0.132 g, 53%) R_F 0.3 (40% EtOAc:CH_2Cl_2) and ii) **79b** (9 mg, 5%) R_F 0.50 (40% EtOAc:CH_2Cl_2).



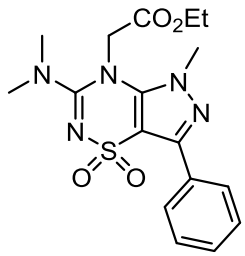
Obtained as a yellow oil, (Found $[M+Na]^+$ 352.1056; $C_{12}H_{19}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 352.1055). δH (600MHz, $CDCl_3$) 4.31 (2H, s, CH_2), 4.12 (2H, q, *J* 7.1, CO$_2$C*H*$_2$CH$_3$), 3.76 (3H, s, N5-CH$_3$), 3.11 (6H, s, 2 x NCH$_3$), 2.42 (3H, s, CH$_3$), 1.16 (3H, t, *J* 7.1, CO$_2$CH$_2$C*H*$_3$). δC (150MHz, $CDCl_3$) 167.4, 155.6, 149.2, 140.8, 103.6,

62.1, 49.7, 39.3, 33.8, 14.0, 13.0. #An nOe correlation was observed between the CH_2 protons at δ 4.12 ppm and the dimethylamino protons at δ 3.11 ppm.

Ethyl 2-(3-(dimethylamino)-5-methyl-7-phenylpyrazolo[3,4-e][1,2,4]thiadiazin-2(5H)-yl)acetate 1,1-dioxide ***78c*** *and ethyl 2-(3-(dimethylamino)-5-methyl-7-phenylpyrazolo[3,4-e][1,2,4]thiadiazin-4(5H)-yl)acetate 1,1-dioxide* ***79c***

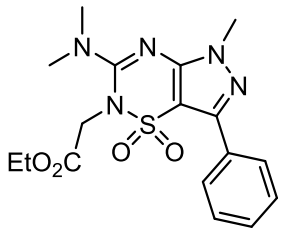
Compound **38f** (0.5 mmol) and ethyl 2-bromoacetate (1.1 mmol) (55°C;21h; DMF). The mixture was cooled to room temperature, then diluted with EtOAc (10 mL) and washed with 10% aqueous LiCl (2 x 5 mL). Product was extracted with EtOAc (10 mL) and the organic layer was dried and evaporated to an orange residue. Elution with 20% EtOAc in CH_2Cl_2 provided **79c** (0.112 g, 57%) R_F 0.33 (40%EtOAc:CH_2Cl_2).

Compound **38f** (0.5 mmol) and ethyl 2-bromoacetate (1.0 mmol) (50°C;69h; DMF). The mixture was cooled to room temperature and water (5 mL) was added. Product was then extracted with CH_2Cl_2 (10 mL) and washed with 1M aqueous HCl (2 x 5 mL). The extracts were dried and evaporated to an orange oil. Elution with 40% EtOAc in CH_2Cl_2 gave two products: i) **79c** (0.122 g, 62 %) R_F 0.30, and ii) **81c** (5 mg, 3%) R_F 0.65 (40%EtOAc:CH_2Cl_2).



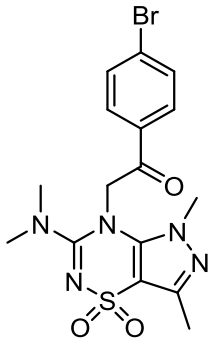
79c: Recrystallisation from CH_2Cl_2 gave white needles, m.p. 188-190°C dec. (Found $[M+Na]^+$ 414.1212; $C_{17}H_{21}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 414.1212). δH (400MHz, $CDCl_3$) 8.07 (2H, d, *J* 7.4, ArH); 7.44 (2H, t, *J* 7.1, ArH); 7.38 (1H, t, *J* 7.3, ArH); 4.50 (2H, s, CH_2); 4.21 (2H, q, *J* 7.1, CH_2CH_3); 3.89 (3H, s, N5-CH_3); 3.08 (6H, s, 2 x NCH_3); 1.20 (3H, t, *J* 7.1, CH_2CH_3). δC (100MHz, $CDCl_3$): 165.9, 155.4, 145.8, 142.8, 130.5, 129.2, 128.7, 128.6, 127.7, 127.6, 108.1, 62.9, 53.7, 40.3, 37.6, 14.0, 13.9. # An

nOe correlation was observed between the methylene signals at δ 4.50 ppm and the dimethylamino signal at δ 3.08 ppm.



81c: Obtained as a yellow oil (Found $[M+Na]^+$ 414.1216; $C_{17}H_{21}N_5O_4{}^{32}SNa$ requires $[M+Na]^+$ 414.1212). δH (400MHz, $CDCl_3$) 7.98 (2H, d, *J* 7.2, ArH); 7.44-7.33 (3H, m, ArH); 4.35 (2H, s, CH_2); 4.11 (2H, q, *J* 7.0, $CO_2C\mathit{H}_2CH_3$); 3.86 (3H, s, N5-CH_3); 3.13 (6H, s, 2 x NCH_3); 1.13 (3H, t, *J* 7.0, $CO_2CH_2C\mathit{H}_3$). δC (100MHz, $CDCl_3$); 167.4, 155.2, 149.7, 143.9, 131.2, 130.5, 129.0, 128.7, 127.6, 102.4, 62.0, 50.0, 39.2, 34.3, 13.9. #An nOe correlation was observed between the CH_2 protons and the dimethylamino protons.

4-(4-Bromobenzoylmethyl)-5,7-dimethyl-3-dimethylamino-4,5-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***79d***



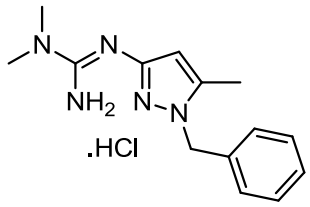
A mixture of **38b** (81 mg, 0.33 mmol), 4-bromophenacyl bromide (0.139 g, 0.66 mmol), $NaHCO_3$ (60 mg, 0.66 mmol), and DMF (2 mL) was stirred at room temperature for 22 hrs. K_2CO_3 (0.125 g, 0.7 mmol) was added and the mixture was stirred for a further 16 hrs. 5% aqueous LiCl (5 mL) was added and stirring was continued for 10 minutes. The red precipitate which formed was collected by filtration under vacuum and purified by chromatography over buffered silica gel (10% EtOAc in CH_2Cl_2 gradient to 40%

EtOAc in CH_2Cl_2) to afford the *title compound* (32 mg, 24%) R_F 0.42 (40%EtOAc:CH_2Cl_2).

Recrystallisation from CH_2Cl_2 gave off-white blocks, m.p. 196-198.5 °C (dec.). (Found $[M+Na]^+$ 462.0217; $C_{16}H_{18}N_5O_3{}^{32}SBrNa$ requires $[M+Na]^+$ 462.0211). δH (400MHz, $CDCl_3$) 7.66 (2H, d, *J* 8.7, ArH), 7.55 (2H, d, *J* 8.7, ArH), 4.98 (2H, s, CH_2), 3.71 (3H, s, N5-CH_3); 3.03 (6H, s, 2 x NCH_3); 2.36 (3H, s, C7-CH_3). δC (100MHz, $CDCl_3$): 191.5, 155.5, 149.0, 140.6, 132.94, 132.3, 129.4, 129.4, 103.4, 54.1, 39.2, 33.8, 33.8, 13.2. #An nOe interaction was observed between the methylene signal at δ 4.98 ppm and the diemthylamino signal at δ 3.03 ppm.

9.12 Synthesis of guanidine derivatives via the extrusion of SO_2

N-(Amino(diethylamino)methylene)-1-benzyl-5-methyl-1H-pyrazol-3-aminium chloride ***81a***



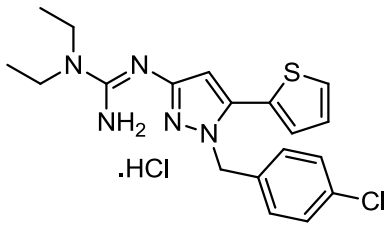
A solution of compound **64f** (44 mg, 0.14 mmol) in CH_2Cl_2 (1.5 mL) was treated with two drops of 10%w/v aqueous HCl and stirred vigorously for 17 hrs at room temperature. The crude mixture was evaporated to a colourless oily residue, which was purified by column chromatography. Elution with 20% MeOH in CH_2Cl_2 gave the *title compound* (31 mg, 87%) as colourless blocks.

A similar reaction heated at reflux for 4 hrs afforded **81a** in 91% yield.

Recrystallisation from MeOH gave white needles, m.p. 95-96°C. (Found: $[M+H]^+$ 258.1721; $C_{14}H_{20}N_5$ requires $[M+H]^+$ 258.1719). δH (400MHz, DMSO-d_6)

10.05 (1H, s, NH), 8.41 (2H, s, NH_2), 7.37-7.27 (3H, m, ArH), 7.14 (2H, d, *J* 7.4, ArH), 6.08 (1H, s, CH), 5.28 (2H, s, C*H*$_2$Ar), 3.10 (6H, s, NCH_3 x 2), 2.25 (3H, s, CH_3). δC (100MHz, DMSO-d_6) 154.2, 146.1, 140.8, 137.5, 129.1, 128.0, 127.5, 97.9, 52.2, 39.2, 11.2.

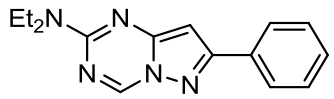
(E)-N-(Amino(diethylamino)methylene)-1-(4-chlorobenzyl)-5-(thien-2-yl)-1H-pyrazol-3-aminium chloride ***81b***



Two drops of 10%w/v aqueous HCl were added to a stirred solution of compound **64d** (53 mg, 0.12 mmol) in CH_2Cl_2 (1.5 mL). The mixture was heated at reflux for 4.5 hrs. The solvent was evaporated and the residue was recrystallised from MeOH:CH_2Cl_2 (1:1) to give the *title compound* (42 mg, 83%) as colourless blocks, m.p. 149-151°C. (Found: $[M+H]^+$ 388.1351; $C_{19}H_{23}N_5{}^{32}S^{35}Cl$ requires [M+H]+ 388.1363). δH (400MHz, DMSO-d_6) 9.78 (1H, s, NH), 8.24 (2H, s, NH_2), 7.75 (1H, dd, *J* 5.1, 1.1, thienyl H5), 7.41 (2H, d, *J* 8.6, ArH), 7.31 (1H, d, *J* 3.6, 1.1, thienyl H3), 7.19 (1H, d, *J* 5.1, 3.6, thienyl H4), 7.12 (2H, d, *J* 8.6, ArH), 6.49 (1H, s, CH), 5.49 (2H, s, C*H*$_2$Ar), 3.50 (4H, q, *J* 7.0, NC*H*$_2$$CH_3$ x 2), 1.17 (6H, t, *J* 7.0, NCH_2C*H*$_3$ x 2), 2.25 (3H, s, CH_3). δC (100MHz, DMSO-d_6) 152.2, 146.0, 137.8, 136.0, 132.3, 128.8, 128.7, 128.62, 128.58, 128.4, 128.2, 99.4, 52.3, 43.2, 12.8.

9.13 Synthesis of pyrazolo[1,5-*a*][1,3,5]triazine derivatives

2-(Diethylamino)-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***83a***



A suspension of **81c** (60 mg, 0.23 mmol) in acetonitrile (1.5 mL) was treated with triethyl orthoformate (0.5 mL, 4.6 mmol) and the mixture heated to reflux temperature for 21 hrs, then cooled to room temperature and solvent evaporated. The crude yellow oil was purified via column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* as beige needles (31 mg, 50%) R_F 0.8 (5%EtOAc:CH_2Cl_2).

A mixture of **81c** (49 mg, 0.19 mmol) in neat triethyl orthoformate (1.5 mL) was heated to reflux temperature for 43 hrs. The crude solution was then cooled to room temperature and solvent evaporated to give a yellow oil. The crude mixture was purified via column chromatography (5% EtOAc in CH_2Cl_2) to afford the *title compound* as off-white needles (48 mg, 94%).

Recrystallisation of **83a** from MeOH gave yellow needles, m.p. 118-120^{o}C. (Found: $[M+H]^+$ 268.1561; $C_{15}H_{18}N_5$ requires $[M+H]^+$ 268.1562). δH (600MHz, DMSO-d_6) 9.33 (1H, s, H4), 7.96 (2H, d, *J* 6.8, ArH), 7.49-7.43 (3H, m, ArH), 6.52 (1H, s, H8), 3.65 (2H, s, NCH_2), 3.60 (2H, s, NCH_2), 1.17 (6H, t, *J* 7.0, CH_3 x 2). δC (100MHz, DMSO-d_6) 157.8, 154.9, 151.0, 147.8, 132.3, 129.3, 128.8, 126.2, 87.6, 41.9, 41.7, 13.6, 12.5. #Short and long range heteronuclear correlations (HMBC/HMQC) were consistent with the assigned structure.

The following general procedure applies for the $POCl_3$-mediated syntheses of pyrazolo-triazines:

$POCl_3$ (2.1-3.3 equiv) was added dropwise to a stirred, cooled solution of **34** (0.5 mmol) in DMF (3 mL) at 5°C. The mixture was stirred at room temperature until TLC indicated consumption of the starting material, then water (5 mL) was added and the

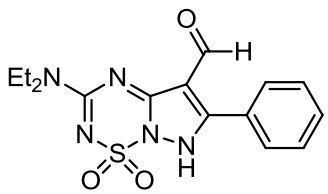
mixture stirred for 5 minutes. The product was extracted with CH_2Cl_2 (10 mL x 2) and the organic phase was washed with 2M aqueous HCl (2 x 10 mL) then 5% aqueous LiCl (10 mL). The organic phase was evaporated and the residue was separated by column chromatography over silica gel.

*2-(Diethylamino)-7-phenyl-pyrazolo[1,5-a][1,3,5]-triazine **83a** and 3-(diethylamino)-5-formyl-6-phenylpyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide **82a***

Compound **34e** (0.162 g, 0.5 mmol) was treated with $POCl_3$ (2.1 equiv) under the general conditions at room temperature (14h). Elution with (10% EtOAc in CH_2Cl_2) provided **83a** (67 mg, 50%) R_F 0.75 (10%EtOAc:CH_2Cl_2) and the starting material (50 mg, 31%).

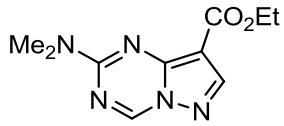
Compound **34e** (0.14 g, 0.43 mmol) was treated with $POCl_3$ (2.5 equiv) under the general conditions at room temperature (3h). Elution with 10% EtOAc in CH_2Cl_2 gave two compounds: i) **83a** (87mg, 75%) R_F 0.75 (10%EtOAc:CH_2Cl_2); and ii) **82a** (14mg, 10%) R_F 0.3 (10%EtOAc:CH_2Cl_2).

Compound **34e** (99 mg, 0.31 mmol) was treated with $POCl_3$ (3.3 equiv) under the general conditions at room temperature (6h). Elution with 15% EtOAc in CH_2Cl_2 gave two compounds; i) **83a** (72mg, 88%) isolated as yellow crystals R_F 0.5 (CH_2Cl_2) and ii) **82a** (8mg, 9%) as off-white crystals R_F 0.35 (15%EtOAc:CH_2Cl_2).



Recrystallisation of **82a** from MeOH gave off-white needles, m.p. 166-168°C dec. (Found: $[M-H]^-$ 346.0975; $C_{15}H_{16}N_5O_3{}^{32}S$ requires $[M-H]^-$ 346.0975). δH (400MHz, DMSO-d_6) 9.85 (1H, s, CO*H*), 7.93 (2H, m, ArH), 7.45 (3H, m, ArH), 3.52 (4H, s, NCH_2 x 2), 1.17 (6H, s, NCH_2CH_3 x 2). δC (100MHz, DMSO-d_6) 181.5, 156.7, 151.7, 133.4, 129.4, 129.0, 128.9, 128.2, 126.9, 41.8, 14.0.

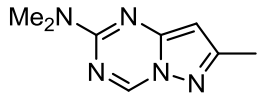
Ethyl 2-Dimethylaminopyrazolo[1,5-a][1,3,5]triazine-8-carboxylate ***83b***



Compound **34i** (0.144 g, 0.5 mmol) was treated with $POCl_3$ (3.2 equiv) under the general conditions at room temperature (40h). Elution with 10% EtOAc in CH_2Cl_2 provided the *title compound* (23 mg, 20%) as colourless prisms R_F 0.3 (10%EtOAc:CH_2Cl_2). Further elution gave the starting material (86 mg, 60%).

Recrystallisation of **83b** from CH_2Cl_2 gave colourless blocks, m.p. 99-101°C. (Found: $[M+Na]^+$ 258.0969; $C_{10}H_{13}N_5O_2Na$ requires $[M+Na]^+$ 258.0969). δH (600MHz, $CDCl_3$) 8.85 (1H, s, H4), 8.29 (1H, s, H7), 4.35 (2H, q, *J* 7.1, OCH_2CH_3), 3.32 (6H, s, 2 x CH_3), 1.39 (3H, t, *J* 7.1, OCH_2CH_3). δC (150MHz, $CDCl_3$) 162.9, 157.9, 151.1, 149.9, 147.3, 93.4, 60.1, 37.7, 37.4, 14.6.

2-Dimethylamino-7-methylpyrazolo[1,5-a][1,3,5]triazine ***83c***



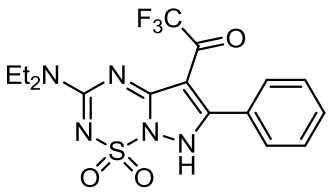
Compound **34j** (0.115 g, 0.5 mmol) was treated with $POCl_3$ (3.3 equiv) under the general conditions at room temperature (25h). The extractive workup was altered in that the mixture was basified slowly with 5M aqueous NaOH then extracted with EtOAc (3 x 5 mL). Elution with 20%EtOAc in CH_2Cl_2 gave the *title compound* (50 mg, 57%) as colourless blocks, R_F 0.4 (20%EtOAc:CH_2Cl_2).

Recrystallisation of **83c** from CH_2Cl_2 gave colourless blocks, m.p. 124-125°C. (Found: $[M+H]^+$ 178.1098; $C_8H_{12}N_5$ requires $[M+H]^+$ 178.1093). δH (400MHz, DMSO-d_6) 9.15 (1H, s, H4), 5.78 (1H, s, H8), 3.30 (6H, s, CH_3 x 2), 2.27 (3H, s, CH_3). δC (100MHz, DMSO-d_6) 157.5, 156.0, 150.3, 147.0, 90.1, 36.8, 14.3. #There is a long range correlation between H4 and two carbons at δ 156.0 ppm and δ 150.3 ppm and a short range between H4 and the carbon at δ 147.0 ppm.

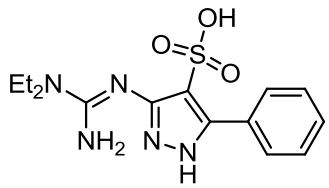
3-Diethylamino- 5-trifluoroacetyl, 6-phenyl [1,5-b][1,2,4,6] thiatriazine 1,1-dioxide ***84*** *and 1H- (E)-3-((amino(diethylamino)methylene)amino)-5-phenyl-1H-pyrazole-4-sulfonic acid* ***85***

Using acetonitrile: A mixture of **34e** (0.161 g, 0.5 mmol) and acetonitrile (2.5 mL) was cooled to 5°C and treated slowly with TFAA (0.14 mL, 1 mmol), then heated to 40°C and stirred for 16 hrs. The mixture was cooled to room temperature and treated with water (5 mL) then extracted with CH_2Cl_2 (5 mL x 2). Extracts were combined and dried, then solvent was evaporated to a red oil which was purified via column chromatography (20% EtOAc in CH_2Cl_2) to afford i) **84** (82 mg, 40%) as yellow needles R_F 0.80 (20% EtOAc:CH_2Cl_2) and ii) **85** (31 mg, 18%) as white needles R_F 0.20 (10%MeOH:CH_2Cl_2). Further elution with 10%MeOH in CH_2Cl_2 returned the starting material (65 mg, 40%).

Using pyridine: A mixture of **34e** (81 mg, 0.25 mmol) and pyridine (2 mL) was cooled to 5°C and treated slowly with TFAA (0.14 mL, 1 mmol). The mixture was stirred for 16 hrs at room temperature then treated with water (5 mL) and stirred for a further 20 minutes. The mixture was extracted with CH_2Cl_2 (10 mL) the extract was washed with saturated aqueous $CuSO_4$ (5 mL x 3), dried and evaporated to a brown oil which was purified via column chromatography (20% EtOAc in CH_2Cl_2) to afford i) **84** (20 mg, 19%) as yellow needles R_F 0.80 (20% EtOAc:CH_2Cl_2) and ii) **85** (52 mg, 61%) as white needles R_F 0.20 (20% EtOAc:CH_2Cl_2).



84: Recrystallisation from CH_2Cl_2 gave yellow needles, m.p. 92-94°C. (Found: $[M-H]^-$ 414.0848; $C_{16}H_{15}{}^{19}F_3N_5O_3{}^{32}S$ requires $[M-H]^-$ 414.0848). δH (500MHz, $CDCl_3$) 11.43 (1H, s, NH), 7.56-7.55 (2H, m, ArH), 7.46-7.44 (3H, m, ArH), 3.76 (4H, *J* 7.1, NCH_2 x 2), 1.28 (6H, t, *J* 7.1, CH_3 x 2). δC (125MHz, $CDCl_3$) 159.7, 158.9, 151.4, 151.0, 150.7, 150.3, 146.2, 133.1, 129.2, 128.7, 128.0, 121.6, 118.8, 100.8, 42.9, 42.7, 13.2, 12.8.



85: Recrystallisation from 1:1 MeOH:CH_2Cl_2 gave colourless blocks, m.p. >300°C. (Found: $[M+Na]^+$ 360.1105; $C_{14}H_{19}N_5O_3{}^{32}SNa$ requires $[M+Na]^+$ 360.1106). δH (600MHz, $CDCl_3$) 13.37 (1H, s, SO_3H), 10.62 (1H, s, NH), 8.62 (2H, s, NH_2), 8.02 (2H, d, *J* 7.1, ArH), 7.49-7.44 (3H, m, ArH), 3.47 (4H, q, *J* 7.1, 2 x NCH_2CH_3), 1.20 (6H, t, *J* 7.1, 2 x NCH_2CH_3). δC (150MHz, $CDCl_3$) 150.9, 144.5, 140.7, 129.4, 128.6, 128.5, 128.1, 112.2, 43.5, 12.7. #HMQC (short range) shows those protons (δ 13 ppm, δ 10.6 ppm and δ 8.6 ppm) do not correlate to carbons. HMBC (long range) confirms assignment of protons and structure.

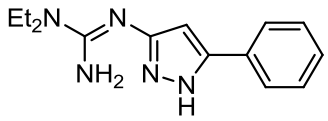
(E)-3-((amino(diethylamino)methylene)amino)-5-phenyl-1H-pyrazole-4-sulfonic acid ***85***

Using TFAA and TFA: A solution of **34e** (80 mg, 0.24 mmol) in acetonitrile (1.5 mL) was treated slowly with TFAA (0.033 mL, 0.24 mmol) followed by TFA (5 µL, 0.05 mmol) and the mixture was warmed to 40°C for 23 hrs. The mixture was cooled to room temperature and evaporated *in vacuo*. The residual yellow oilwas chromatographed over silica gel (20%EtOAc in CH_2Cl_2) to give the starting material (42 mg, 53%) and *the title compound* as white needles (25 mg, 30%) R_F 0.20 (20%EtOAc:CH_2Cl_2).

Using H_2SO_4: A solution of **34e** (81 mg, 0.25 mmol) in acetonitrile (2 mL) was treated with a single drop of concentrated H_2SO_4 and heated to reflux temperature for 19 hrs. The solvent was evaporated and the crude mixture was purified by column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* as white needles (72 mg, 85%) R_F 0.5 (10%MeOH:CH_2Cl_2).

Using TFA: A mixture of **34e** (0.162 g, 0.5 mmol) in acetonitrile (2.5 mL) was treated with TFA (0.4 mL, 5 mmol) and heated to 80°C for 30 hrs. The mixture was evaporated to give the *title compound* as a beige powder (0.168 g, 100%).

(E)-3-((amino(diethylamino)methylene)amino)-5-phenyl-1H-pyrazole-4-sulfonic acid ***85*** *and (E)-1,1-Diethyl-2-(5-phenyl-1H-pyrazol-3-yl)guanidine* ***81c***



A solution of **34e** (0.162 g, 0.5 mmol) in acetonitrile (3 mL) was treated with TFA (0.2 mL, 2.6 mmol) then heated to 80°C in air for 19 hrs. The mixture was cooled to room temperature and solvent evaporated *in vacuo*. The residue was dissolved in hot CH_2Cl_2 and gradually cooled via ice/water bath to form a white precipitate. The solid was collected by filtration under vacuum to give **85** (0.109 g, 65%). The filtrate was evaporated and the residue was purified via column chromatography (20% MeOH in CH_2Cl_2) to afford **81c** (38 mg, 30%) R_F 0.32 (10%MeOH:CH_2Cl_2).

81c: Obtained as a beige gum. (Found: $[M+H]^+$ 258.1722; $C_{14}H_{20}N_5$ requires $[M+H]^+$ 258.1719). δH (600MHz, DMSO-d_6) 9.79 (1H, s, pyrazole H1), 8.35 (2H, s, NH_2), 7.76 (2H, d, J 7.5Hz, ArH), 7.50 (2H, t, J 7.5Hz, ArH), 7.41 (1H, t, J 7.4Hz, ArH), 6.63 (1H, s, pyrazole H4), 3.52 (4H, q, *J* 7.0Hz, NCH_2 x 2), 1.18 (6H, t, *J* 7.0Hz, NCH_2CH_3 x 2). δC (150MHz, DMSO-d_6) 152.4, 143.3, 129.2, 128.9, 128.6, 125.7, 125.4, 95.2, 43.2, 12.8. #HMBC (long range) correlation between singlet at δ 6.5 ppm and two carbons at δ 143 ppm and δ 147 ppm. Short range between singlet at δ 6.5 ppm and carbon at δ 95 ppm. No correlations between peaks at δ 9.74 ppm and δ 8.31 ppm with any carbons long or short range.

(E)-1,1-Diethyl-2-(5-phenyl-1H-pyrazol-3-yl)guanidine ***81c***

Using TFA in acetonitrile: A solution of **34e** (82 mg, 0.25 mmol) in acetonitrile (2 mL) was treated with TFA (0.1 mL, 1.3 mmol) then the mixture was heated to 75°C for 43 hrs. The mixture was concentrated to a brown oil and purified via silica gel chromatography (20% MeOH in CH_2Cl_2) to give the *title compound* as a colourless oil (50 mg, 78%) R_F 0.45 (20%MeOH:CH_2Cl_2).

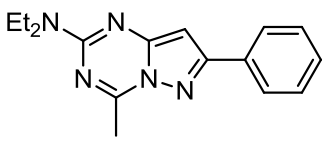
Using HCl in acetonitrile:water: A mixture of **34e** (0.161 g, 0.5 mmol) in 1:3 water:acetonitrile (2 mL) was treated with concentrated HCl (5 drops) then heated to reflux temperature for 17 hrs. The mixture was neutralised with 1M aqueous NaOH and the solvent was evaporated. The residue was dissolved in MeOH and filtered to remove the NaCl. The filtrate was evaporated to give a brown oil, which was purified by silica gel chromatography (20% MeOH in CH_2Cl_2) to give the *title compound* as a beige gum (102 mg, 79%).

Using HCl: A sample of **34e** (0.162 g, 0.5 mmol) was heated to reflux temperature in concentrated aqueous HCl (4 mL) for 2.5 hrs. The mixture was cooled to room temperature and concentrated to give the crude compound **81c** (0.163 g, 100%) as a yellow-brown oil.

Using tosylic acid in acetonitrile: A mixture of **34e** (0.159 g, 0.5 mmol) and *p*-toluenesulfonic acid hydrate (0.16 g, 0.84 mmol) in acetonitrile (2.5 mL) was heated to 60°C for 20 hrs then cooled to room temperature. The mixture was evaporated to give a brown oil which was purified via column chromatography (20% MeOH in CH_2Cl_2) to afford the *title compound* as a beige oil (96 mg, 75%).

Using TFA in acetonitrile:water: A mixture of **34e** (0.250 g, 0.78 mmol) in 1:1 acetonitrile:water (4 mL) was treated with TFA (0.4 mL, 5 mmol) and heated to 55°C for 72 hrs. The mixture was evaporated and the residual brown oil was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* as a beige viscous gum (0.172 g, 86%).

2-Diethylamino-4-methyl-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***83d***



Using DMAc and $POCl_3$: $POCl_3$ (60 µL, 0.65 mmol, 3.4 equiv) was added slowly to a stirred solution of **81c** (50 mg, 0.19 mmol) in DMAc (0.5 mL) at 5°C. The resulting mixture was stirred at room temperature for 24 hrs. The mixture was diluted with water

(10 mL) and extracted with EtOAc (5 mL x 2). The combined organic extracts were washed with 5% aqueous LiCl (5 mL), dried, and evaporated. The residue was purified by column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* (6 mg, 11%) as an off-white amorphous solid. R_F 0.8 (5%EtOAc:CH_2Cl_2).

Using trimethyl orthoacetate: A mixture of **81c** (34 mg, 0.16 mmol), methane sulfonic acid (10 µL, 0.016 mmol) and trimethyl orthoaceate (1.2 mL) was heated to reflux temperature for 16 hrs, then cooled to room temperature and evaporated to a yellow semi-solid. The crude material was purified via column chromatography (CH_2Cl_2) to give the *title compound* (28 mg, 75%) as colourless prisms. R_F 0.45 (CH_2Cl_2).

Using acetic anhydride: A mixture of **81c** (61 mg, 0.24 mmol) , acetic anhydride (44 µL, 0.47 mmol), and DMF (1 mL) was treated with *N*,*N*'-diisopropylethylamine (80 µL, 0.46 mmol) and heated to 70°C for 25 hrs. The mixture was quenched with 1M aqueous NaOH (5 mL) to form a white precipitate which was collected by filtration and washed with 1M aqueous NaOH. The crude product was purified via column chromatography (10%X4 in CH_2Cl_2) to give the *title compound* as colourless needles (28 mg, 42%), R_F 0.6 (10%X4 in CH_2Cl_2).

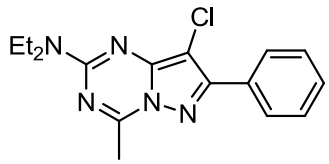
Recrystallisation of **83d** from 1:1 MeOH:CH_2Cl_2 gave colourless needles, m.p. 112-114°C. (Found: $[M+H]^+$ 282.1718; $C_{16}H_{20}N_5$ requires $[M+H]^+$ 282.1719). δH (400MHz, $CDCl_3$) 7.95 (2H, d, *J* 8.4, ArH), 7.46-7.36 (3H, m, ArH), 6.29 (1H, s, H8), 3.67 (4H, q, *J* 7.0, NCH_2 x 2), 2.82 (3H, s, CH_3), 1.23 (6H, t, *J* 7.0, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 157.7, 156.6, 155.3 152.0, 133.1, 129.0, 128.6, 126.6, 88.1, 42.0, 19.5, 13.2.

2-Diethylamino-4-methyl-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***83d*** *and 2-diethylamino-4-methyl-7-phenyl-8-chloropyrazolo[1,5-a][1,3,5]triazine* ***86***

A solution of **81c** (57 mg, 0.22 mmol) in DMAc (400 µL) was treated with $POCl_3$ (70 µL, 0.75 mmol) at room temperature and the mixture was gently heated to 50°C for 23 hrs. The mixture was cooled to room temperature and water (5 mL) was added. The mixture was extracted with EtOAc (5 mL) and the organic phase was washed with 5% aq. LiCl (5 mL), dried, and evaporated, The residual orange oil was purified via column

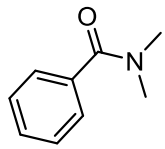
chromatography (10% X4 in CH_2Cl_2) to give i) **86** as a yellow oil (3 mg, 4%), R_F 0.6 (10%X4:CH_2Cl_2) and ii) **83d** (21 mg, 33%) as colourless needles, R_F 0.35 (10%X4:CH_2Cl_2). Further elution (10% MeOH in CH_2Cl_2) afforded the starting material (18 mg, 32%).

One drop of concentrated aqueous HCl was added to a solution of **81c** (30 mg, 0.12 mmol) in acetic anhydride (1 mL) and the resulting mixture was heated to reflux temperature for 15 minutes. The mixture was cooled, diluted with water (5 mL) and the precipitate was collected by filtration under vacuum. The crude solid was purified via column chromatography to give **86** (5 mg, 13%) R_F 0.85 (10%EtOAc:CH_2Cl_2).



86: Obtained as an amorphous off-white solid. (Found: $[M+Na]^+$ 316.1335; $C_{16}H_{19}N_5{}^{35}ClNa$ requires $[M+Na]^+$ 316.1329). δH NMR (600 MHz, $CDCl_3$) 8.04 (2H, d, J 8.6, ArH), 7.49-7.42 (3H, m, ArH), 3.71 (4H, q, J 7.1, NCH_2 x 2), 2.80 (3H, s, CH_3), 1.25 (6H, m, NCH_2CH_3 x 2).

N'N-Dimethyl benzamide



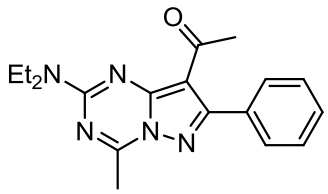
Benzoyl chloride (1.20 mL, 0.01 mol) was added dropwise over 10 minutes to a mixture of 40% aqueous dimethylamine (1.24 g, 0.011 mol), triethylamine (1.74 mL, 0.0125 mol), and CH_2Cl_2 (20 mL) . After 30 minutes at room temperature the solution was diluted with CH_2Cl_2 (20 mL) and washed sequentially with 1M aqueous HCl (20 mL)

and saturated aqueous NaCl (20 mL). The organic layer was then dried and evaporated to a beige semi-solid (1.36 g, 91%).[3]

δH (600MHz, $CDCl_3$) 7.35-7.31 (5H, m, ArH), 3.04 (3H, s, NCH_3), 2.90 (3H, s, NCH_3). δC (150MHz, $CDCl_3$) 171.8, 136.4, 129.6, 128.4, 127.2, 39.4, 35.5.

2-Diethylamino-4-methyl-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***83d*** *and 1-(2-Diethylamino)-4-methyl-7-phenylpyrazolo[1,5-a][1,3,5]triazin-8-yl)ethan-1-one* ***87***

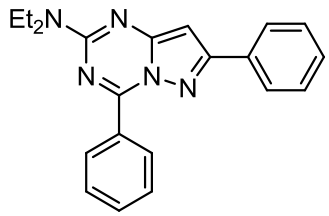
A solution of **81c** (53 mg, 0.2 mmol), trimethyl orthoacetate (1.5 mL) and methanesulfonic acid (15 µL, 0.022 mmol) was heated to reflux temperature for 26 hrs. The mixture was cooled to room temperature and evaporated. The residual brown oil was purified via column chromatography (CH_2Cl_2) to give i) **83d** as yellow-white needles (40 mg, 71%) R_F 0.5 (CH_2Cl_2) and ii) **87** (5 mg, 8%) as a white powder, R_F 0.3 (CH_2Cl_2).



Recrystallisation of **87** from CH_2Cl_2 gave colourless needles, m.p. 90-92°C. (Found: $[M+H]^+$ 296.1871; $C_{17}H_{22}N_5$ requires $[M+H]^+$ 296.1875). δH (400MHz, $CDCl_3$) 7.62 (2H, d, *J* 7.8, ArH), 7.48-7.41 (3H, m, ArH), 3.93 (3H, s, CH_3), 3.74 (4H, q, *J* 7.0Hz, NCH_2 x 2), 2.44 (3H, s, $COCH_3$), 1.23 (6H, t, *J* 7.0, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 163.3, 159.8, 156.5, 145.6, 134.0, 129.4, 128.7, 128.3, 126.6, 105.1, 42.0, 32.8, 23.6, 13.2.

[3] The data collected matches literature NMR data.

2-Diethylamino-4,7-diphenylpyrazolo[1,5-a][1,3,5]triazine ***83e***



General procedure:

A solution of **81c**, amine base (2 equiv), and electrophile (2 equiv) in DMF (1 mL) was stirred at 100°C. The mixture was cooled to room temperature and quenched with 1M aqueous NaOH (5 mL) to form a precipitate which was collected by filtration, and washed with 1M aqueous NaOH. If a precipitate did not form, an extractive workup with EtOAc (5mL) was performed and then the organic phase was dried and evaporated. The crude product was purified via column chromatography over silica gel with 20% X4 in CH_2Cl_2 as the mobile phase.

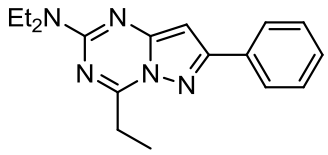
Using Benzoyl chloride and $^{i}Pr_2NEt$: The general procedure was applied with **81c** (0.21 mmol), *N,N'*-diisopropylethylamine (0.4 mmol) and benzoyl chloride (0.22 mmol) for 4.5hrs. An extractive workup was performed with EtOAc to give a yellow oil. The *title compound* (22 mg, 31%) was obtained as a bright yellow powder. R_F 0.7 (20% X4 in CH_2Cl_2)

Using Benzoic anhydride and Et_3N: The general procedure was applied with **81c** (0.21 mmol), triethylamine (0.4 mmol) and benzoic anhydride (0.4 mmol) for 39 hrs. The *title compound* (43 mg, 60%) was obtained as a bright yellow powder.

Using Benzoic anhydride and $^{i}Pr_2NEt$: The general procedure was applied with **81c** (0.19 mmol), *N,N'*-diisopropylethylamine (0.4 mmol) and benzoic anhydride (0.38 mmol) for 48 hrs. An extractive workup was performed with EtOAc to give a yellow oil. The *title compound* (46 mg, 71%) was obtained as a bright yellow powder (46 mg, 71%)

Recrystallisation of **82e** from EtOAc gave bright yellow needles, m.p. 89-90°C. (Found: $[M+H]^+$ 344.1871; $C_{21}H_{22}N_5$ requires $[M+H]^+$ 344.1875). δH (400MHz, $CDCl_3$) 8.81 (2H, d, *J* 6.8, ArH), 8.0 (2H, d, *J* 6.8, ArH), 7.65-7.56 (3H, m, ArH), 7.48-7.38 (3H, m, ArH), 6.40 (1H, s, H5), 3.76 (4H, broad m, NCH_2 x 2), 1.30 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 157.9, 155.4, 154.0, 153.4, 133.1, 132.3, 131.3, 131.2, 129.1, 128.6, 128.1, 126.7, 87.8, 42.2, 13.3.

2-Diethylamino-4-ethyl-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***83f***



General procedure:

A solution of **81c**, *N,N'*-diisopropylethylamine (2 equiv), and propionic anhydride (2 equiv) in DMF (1 mL) was stirred at 100°C or 70°C until TLC indicated completion. The mixture was cooled to room temperature and quenched with 1M aqueous NaOH (5 mL) then extracted with EtOAc (5mL). The organic phase was dried and evaporated and the crude product was purified via column chromatography over silica gel.

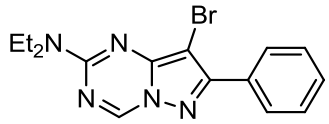
At 70°C: The general procedure was applied with **81c** (51 mg, 0.19 mmol) for 18 hrs. Elution with 10% X4 in CH_2Cl_2 provided the *title compound* as a colourless amorphous solid (15 mg, 27%). R_F 0.65 (10%X4 in CH_2Cl_2).

At 100°C: The general procedure was applied with **81c** (53 mg, 0.2 mmol) for 17 hrs. The mixture was not treated with 1M aqueous NaOH and, instead the solvent was evaporated *in vacuo*. Elution with 15% X4 in CH_2Cl_2 provided the *title compound* as a colourless semi-solid (39 mg, 66%) R_F 0.45 (15%X4 in CH_2Cl_2).

Recrystallisation of **83f** from MeOH gave colourless blocks, m.p. 106-108°C. (Found: $[M+H]^+$ 296.1864; $C_{17}H_{21}N_5$ requires $[M+H]^+$ 296.1875). δH (400MHz, $CDCl_3$) 7.95 (2H, d, J 7.3, ArH), 7.45-7.36 (3H, m, ArH), 6.29 (1H, s, H5), 3.68 (4H, broad q, J 6.6,

NCH_2 x 2), 3.24 (2H, q, J 7.4, CH_2CH_3), 1.43 (3H, t, J 7.4, CH_2CH_3), 1.23 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 160.0, 157.5, 155.5, 152.1, 133.2, 128.9, 128.6, 126.6, 88.1, 42.1, 24.9, 13.2, 9.2.

8-Bromo-2-diethylamino-7-phenylpyrazolo[1,5-a][1,3,5]triazine ***88***



From **34e**: A solution of **34e** (85 mg, 0.27 mmol) and *N*-bromosuccinimide (49 mg, 0.28 mmol) in chloroform (2 mL) was heated to reflux temperature for 45 minutes. The reaction mixture was cooled to room temperature, diluted with CH_2Cl_2 (10 mL), then washed with aqueous $NaHCO_3$ (2 x 10 mL). The solvent was evaporated and the red sticky solid was dried *in vacuo*. DMF (2 mL) was added under nitrogen and $POCl_3$ (0.1 mL, 0.58 mmol) was added dropwise. The reaction was stirred at room temperature for 2 hrs, then quenched with water (3 mL) and stirred at room temperature for 10 minutes. Saturated aqueous K_2CO_3 solution (5 mL) was added and the mixture was extracted with EtOAc (10 mL). The organic phase was washed with 5% aqueous LiCl solution (2 x 5 mL), dried and evaporated to obtain a brown sticky solid which was purified via column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* as a white solid (32 mg, 30%) R_F 0.85 (5% EtOAc:CH_2Cl_2).

From **83a** as a precursor: A mixture of **83a** (44 mg, 0.13 mmol) and *N*-bromosuccinimide (25 mg, 0.14 mmol) in chloroform (2 mL) was heated to reflux temperature for 16 hrs. The mixture was cooled to room temperature, diluted with $CHCl_3$ (5 mL), washed with aqueous $NaHCO_3$ (2 x 10 mL), dried and evaporated *in vacuo*. The residual white powder was purified using column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* as white crystals (52 mg, 94%).

Recrystallisation of **88** from CH_2Cl_2 to give fine white needles, m.p. 105-106°C. (Found: $M^{+\bullet}$ 345.0584; $C_{15}H_{16}N_5{}^{79}Br$ requires $M^{+\bullet}$ 345.0589). δH (400MHz, $CDCl_3$) 8.8 (1H, s, H4), 8.02 (2H, d, *J* 5.6, ArH), 7.45 (3H, m, ArH), 3.72 (4H, br-s, NCH_2 x 2), 1.26 (3H, br-s, NCH_2CH_3), 1.23 (3H, br-s, NCH_2CH_3). δC (100.1MHz, $CDCl_3$) 155.9,

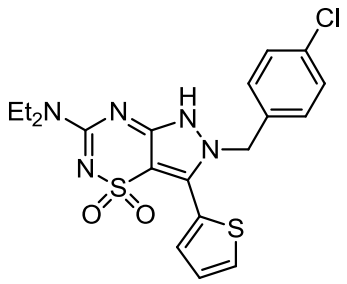
155.7, 148.8, 146.4, 131.7, 131.3, 129.4, 129.4, 128.5, 128.4, 128.4, 128.1, 42.6, 42.4, 13.6, 12.4. *m/z* (EI) 347/345 (100/99.5%, $M^{+\bullet}$), 332/330 (69/70).

9.14 Synthesis of pyrazolo-thiadiazines from rearrangement of pyrazolothiatriazines

General procedure for attempted aryl coupling:

A mixture of **64**, bromobenzene (2 equiv), K_2CO_3 (2 equiv), palladium catalyst (10-20 mol %), PPh_3 (20-40 mol %), and dioxane (1.5-2 mL) was heated to reflux temperature. The mixture was cooled to room temperature and filtered through a short pad of Celite. Water (5 mL) was added to the filtrate and the mixture was extracted with CH_2Cl_2 (2x 5 mL). The combined organic extracts were dried and evaporated. The product was purified by chromatography over silica gel.

6-(4-Chlorobenzyl)-3-diethylamino-7-(thien-2-yl)-5,6-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***89a***



The general procedure was carried out with **64d** (51 mg, 0.11 mmol), bromobenzene (36 mg, 0.22 mmol), K_2CO_3 (31 mg, 0.22 mmol), PEPPSI-^{i}pr (4 mg, 0.0055 mmol) and PPh_3 (16 mg, 0.055mmol) (20 hrs). Elution with 10% EtOAc in CH_2Cl_2 gave the *title compound* (46 mg, 90%) as colourless blocks, R_F 0.48 (20%EtOAc:CH_2Cl_2).

The general procedure was carried out with **64d** (50 mg, 0.11 mmol), bromobenzene (22 mg, 0.14 mmol), K_2CO_3 (20 mg, 0.13 mmol), $Pd(OAc)_2$ (3 mg, 0.01 mmol) and PPh_3 (6 mg, 0.02 mmol) (25 h) Elution with 20% EtOAc in CH_2Cl_2 afforded the *title compound* (50 mg, 100%) as off-white needles, R_F 0.48 (20%EtOAc:CH_2Cl_2).

The general procedure was carried out with **64d** (53 mg, 0.12 mmol), K_2CO_3 (25 mg, 0.18 mmol), iodobenzene (38 mg, 0.18 mmol) and $Pd(PPh_3)Cl_2$ (~2.5 mg, 0.036 mmol) at 50°C (22h). The mixture was cooled to room temperature, diluted with water (5mL), and extracted with EtOAc (5mL). The organic layer was washed with 5% aqueous LiCl (5mL), dried and evaporated. Elution with 10% EtOAc in CH_2Cl_2 afforded the *title compound* as a white powder (20 mg, 38%).

The general procedure was carried out with **64d** (27 mg, 0.06 mmol) and $Pd(dba)_2$ (6 mg, 0.01 mmol) (6.5 h). Elution with 10% EtOAc in CH_2Cl_2 provided the *title compound* as a white powder (20 mg, 74%).

Without palladium: A mixture of **64d** (23 mg, 0.05 mmol), K_2CO_3 (18 mg, 0.13 mmol) and PPh_3 (15 mg, 0.06 mmol) in dioxane (1 mL) was heated to reflux temperature for 19 hrs. The mixture was cooled to room temperature and evaporated *in vacuo*. The residue was purified by column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* as a white powder (2 mg, 8%).

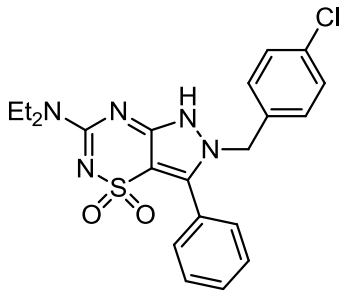
Using DMF: A mixture of **64d** (27 mg, 0.06 mmol), K_2CO_3 (19 mg, 0.12 mmol), iodobenzene (20 mg, 0.10 mmol), $Pd(dba)_2$ (10 mg, 0.011 mmol), and DMF (1 mL) was stirred at 80°C for 28 hrs. The mixture was cooled to room temperature, diluted with water (5 mL) and extracted with CH_2Cl_2 (5 mL). After general workup, the organic layer was washed with 5% aqueous LiCl (5 mL), dried and evaporated to give a yellow oil which was purified via column chromatography (10%EtOAc in CH_2Cl_2) to yield the *title compound* as white flakes (20 mg, 74%).

Using DMAc: A mixture of **64d** (27 mg, 0.06 mmol), KOAc (9 mg, 0.09 mmol), bromobenzene (150 mg, 0.095 mmol) and $Pd(OAc)_2$ (3 mg, 0.012 mmol) in DMAc (1.2 mL) was heated to 140°C for 15.5 hrs. The mixture was cooled to room temperature,

diluted with water (5 mL), and extracted with EtOAc (5 mL). The organic phase was washed with 5% aqueous LiCl (5 mL), dried, and evaporated to a yellow powder. The crude material was purified via column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* as white flakes (21 mg, 78%).

Recrystallisation of **89a** from CH_2Cl_2 gave white needles, m.p. 210°C dec. (Found: $[M-H]^-$ 448.0676; $C_{19}H_{19}N_5O_2Cl^{32}S_2$ requires $[M-H]^-$ 448.0669). δH (400MHz, $CDCl_3$) 7.74 (1H, dd, *J* 3.7, 1.1, thienyl H5), 7.56 (1H, s, NH), 7.53 (1H, dd, *J* 5.1, 1.1, thienyl H3), 7.31 (2H, d, *J* 8.4, ArH), 7.19 (1H, dd, *J* 5.1, 3.7Hz, thienyl H4), 7.06 (2H, d, *J* 8.4, ArH), 5.40 (2H, s, CH_2Ar), 3.50 (4H, q, *J* 7.2, NCH_2CH_3 x 2), 1.27 (3H, t, *J* 7.2, NCH_2CH_3 x 2). δC (400MHz, $CDCl_3$) 147.0, 146.3, 134.9, 134.4, 134.2, 132.3, 129.5, 129.4, 129.3, 128.5, 125.9, 104.9, 53.4, 43.1, 13.4.

6-(4-Chlorobenzyl)-3-diethylamino-7-phenyl-5,6-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***89c***



The general procedure was carried out with **64b** (53 mg, 0.12 mmol), K_2CO_3 (0.022 g, 0.145 mmol), $PdOAc_2$ (5 mg, 0.021 mmol) and PPh_3 (19 mg, 0.039 mmol) (19 h). Elution with 20% EtOAc in CH_2Cl_2provided the *title compound* (48 mg, 91%) as white flakes.

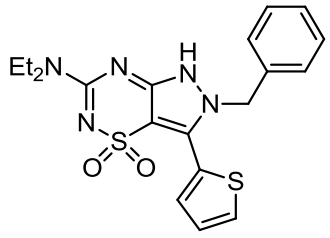
The general procedure was carried out with **64b** (53 mg, 0.12 mmol), K_2CO_3 (20 mg, 0.14 mmol), PEPPSI-ipr (4 mg, 0.006 mmol) and PPh_3 (16 mg, 0.014 mmol) (17h). Elution with 20% EtOAc in CH_2Cl_2provided the *title compound* (30 mg, 57%) as white needles.

Using EtOAc: A solution of pyrazole **64b** (85 mg, 0.19 mmol) in ethyl acetate (4.5 ml) was boiled until TLC indicated completion (2 days). The mixture was diluted with CH_2Cl_2 and filtered to remove insoluble impurities. The filtrate was evaporated and the amorphous residue was recrystallised from aqueous MeOH to give the *title compound* as white needles (71 mg, 83%).

Without palladium, ligand or base: A solution of pyrazole **64b** (60 mg, 0.135 mmol) in dioxane (4.5 ml) was heated at reflux temperature for 8 hrs. The solvent was evaporated and the amorphous residue was recrystallised from aqueous MeOH to give the *title compound* as long flat needles (53 mg, 88%).

Recrystallisation of **89c** from 1:1 MeOH:CH_2Cl_2 gave white flakes, m.p. 128-129°C. (Found: $[M+Na]^+$ 466.1081; $C_{21}H_{22}N_5O_2Cl^{32}SNa$ requires $[M+Na]^+$ 466.1080). δH (400MHz, $CDCl_3$) 8.39 (1H, s, NH), 7.60-7.57 (2H, m, ArH), 7.46-7.44 (3H, m, ArH), 7.23 (2H, d, *J* 8.4, ArH), 6.97 (2H, d, *J* 8.4, ArH), 5.12 (2H, s, C*H*$_2$Ar), 3.43 (4H, q, *J* 7.2, NCH_2 x 2), 1.18 (6H, t, *J* 7.2, CH_3 x 2). δC (100MHz, $CDCl_3$) 147.2, 146.5, 140.8, 134.3, 133.9, 130.3, 129.4, 129.1, 129.0, 128.4, 126.7, 104.6, 52.5, 42.8, 13.1.

6-Benzyl-3-diethylamino-7-(thien-2-yl)-5,6-dihydropyrazolo[3,4-e][1,2,4]thiadiazine 1,1-dioxide ***89b***



The general procedure was carried out with **64e** (50 mg, 0.12 mmol), K_2CO_3 (20 mg, 0.14 mmol) and $Pd(dba)_2$ (10 mg, 0.012 mmol) (24h). Elution with 20%EtOAc in CH_2Cl_2 gave the *title compound* (35 mg, 70%) as white flakes.

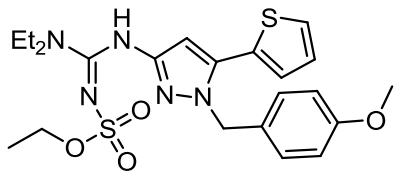
The general procedure was carried out with **64e** (50 mg, 0.12 mmol), K_2CO_3 (21 mg, 0.14 mmol) and $Pd(PPh_3)_2Cl_2$ (10 mg, 0.014 mmol) (16h). Elution with CH_2Cl_2 gave the *title compound* (15 mg, 30%) and the starting material (5 mg, 10%).

Using DMAc: A mixture of **64e** (45 mg, 0.1 mmol), K_2CO_3 (32 mg, 0.22 mmol), $Pd(OAc)_2$ (5.6 mg, 0.24 mmol), PPh_3 (18 mg, 0.067 mmol), bromobenzene (37 mg, 0.22 mmol) in DMAc (1.5 mL) was stirred at 100°C for 24 hrs. The reaction mixture was cooled to room temperature and filtered through a pad of Celite. The filtrate was treated with water (10mL) and extracted with CH_2Cl_2 (10mL, then 2 x 5mL). The combined organic phase was concentrated *in vacuo* and the residue was purified by column chromatography (20% EtOAc in CH_2Cl_2) to give the *title compound* (35 mg, 78%) as white flakes, R_F 0.50 (20%EtOAc:CH_2Cl_2).

Recrystallisation of **89b** from 1:1 MeOH:CH_2Cl_2 gave white needles, m.p. 218°C dec. (Found $[M+Na]^+$ 438.1040; $C_{19}H_{20}N_5O_2{}^{32}S_2Na$ requires $[M+Na]^+$ 438.1034). δH (600MHz, $CDCl_3$) 7.73 (1H, dd, J 3.7, 1.0Hz, thienyl H5), 7.65 (1H, s, NH), 7.52 (1H, dd, J 5.0, 1.0Hz, thienyl H3), 7.36-7.29 (3H, m, ArH), 7.17 (1H, dd, *J* 5.0, 3.7Hz, thienyl H4), 7.12 (2H, d, *J* 7.2, ArH); 5.43 (2H, s, CH_2), 3.48 (4H, q, *J* 7.0, NCH_2CH_3 x 2), 1.27 (3H, t, *J* 7.0, NCH_2CH_3 x 2). δH (400MHz, DMSO-d_6) 11.16 (1H, s, NH),7.86 (1H, dd, J 3.9, 1.1Hz, thienyl H5), 7.62 (1H, d, J 3.7, 1.1Hz, thienyl H3), 7.37-7.25 (4H, m, ArH + thienyl H4), 7.10 (2H, d, 7.0Hz, ArH), 5.49 (2H, s, CH_2Ar), 3.48 (4H, q, J 7.0, 2 x NCH_2CH_3), 1.12 (3H, t, J 7.0Hz, 2 x NCH_2CH_3). δC (150MHz, DMSO-d_6) 147.7, 146.9, 136.4, 132.5, 131.1, 130.2, 128.8, 128.1, 127.9, 126.9, 126.2, 104.5, 53.4, 42.3, 13.2. #HMBC long range correlation seen between thienyl protons and a carbon in the baseline at ~ δ 95 ppm. HMQC and NOESY also consistent with assigned structure

9.15 Synthesis of compounds arising from nucleophilic attack on the sulfamide moiety

Ethyl (E)-((diethylamino)((1-(4-methoxybenzyl)-5-(thien-2-yl)-1H-pyrazol-3-yl)amino)methylene)sulfamate ***90a***

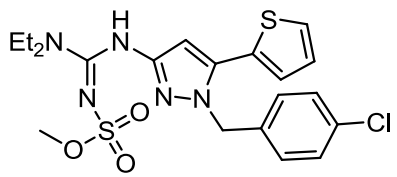


A solution of **64g** (15 mg, 0.034 mmol) in ethanol (0.5 mL) was stirred at room temperature until TLC indicated completion (16 hrs). The mixture was evaporated and the residue was purified by column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* (11 mg, 65%) as a colourless oil, R_F 0.45 (10%EtOAc:CH_2Cl_2).

A mixture of **64g** (30 mg, 0.067 mmol) and $Pd(OH)_2/C$ (7 mg, 0.013 mmol) in EtOH (1 mL) was placed under a H_2 atmosphere and stirred vigorously for 10 hrs. The mixture was poured onto a short pad of Celite and washed through with CH_2Cl_2 (5 mL). The combined filtrate was evaporated to a brown oil which was purified by column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* (9 mg, 27%) as a colourless oil.

90a obtained as a colourless oil (Found: $[M+H]^+$ 492.1735; $C_{20}H_{25}N_5O_4{}^{32}S_2$ requires $[M+H]^+$ 492.1734). δH (400MHz, $CDCl_3$) 8.11 (1H, s, NH), 7.41 (2H, dd, *J* 5.04, 1.2, thienyl H3), 7.08-7.04 (4H, m, ArH + theinyl H4, H5), 6.83 (2H, d, *J* 8.8, ArH), 6.08 (1H, s, CH), 5.26 (2H, s, C*H*$_2$Ar), 4.23 (2H, q, *J* 7.1, OC*H*$_2$CH$_3$), 3.78 (3H, s, OCH_3), 3.35 (4H, q, *J* 7.1, NCH_2 x 2), 1.32 (3H, t, *J* 7.1, OCH_2CH_3), 1.09 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 159.2, 156.9, 147.0, 138.1, 130.0, 129.2, 128.8, 128.3, 127.9, 127.8, 127.4, 114.1, 100.0, 66.2, 55.3, 52.9, 43.4, 14.9, 12.5.

Methyl ((Z)-(((Z)-1-(4-chlorobenzyl)-5-(thien-2-yl)-1,2-dihydro-3H-pyrazol-3-ylidene)amino)(diethylamino)methylene)sulfamate ***90b***

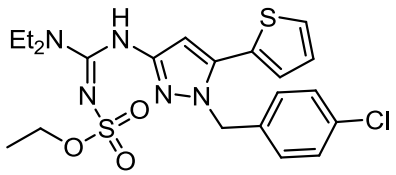


A solution of **64d** (52 mg, 0.12 mmol) and ammonium formate (36 mg, 0.6 mmol) in methanol (2.5 mL) was treated with 10%Pd/C (0.013 g, 10 mol%) and stirred at reflux temperature for 16 hrs. The mixture was filtered through a short pad of Celite, which was rinsed with CH_2Cl_2 (5 mL). The combined filtrate was evaporated and residue was purified via column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* as a beige gum (25 mg, 43%) R_F 0.45 (5%EtOAc:CH_2Cl_2).

A solution of **64d** (54 mg, 0.12 mmol) in methanol (1.5 mL) was treated with 10%Pd/C (13 mg, 0.012 mmol) and stirred at room temperature for 21 hrs. The mixture was then filtered through a short pad of Celite, which was rinsed with CH_2Cl_2 (5mL). The combined filtrate was evaporated and the residue was purified via column chromatography (5% EtOAc in CH_2Cl_2) to give the *title compound* as a colourless gum (41 mg, 71%).

90b obtained as a colourless gum. (Found: $[M+H]^+$ 482.1084; $C_{20}H_{25}N_5O_3{}^{32}S_2Cl$ requires $[M+H]^+$ 482.1082). δH (600MHz, $CDCl_3$) 8.12 (1H, s, NH), 7.41 (2H, dd, *J* 5.1, 1.0, thienyl H5), 7.27 (2H, d, *J* 6.8, ArH), 7.07 (1H, dd, *J* 5.1, 3.7, theinyl H5), 7.04-7.01 (3H, m, ArH + thienyl H3), 6.10 (1H, s, CH), 5.29 (2H, s, CH_2Ar), 3.85 (3H, s, OCH_3), 3.36 (4H, q, *J* 7.1, NCH_2 x 2), 1.10 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 156.6, 147.3, 138.4, 135.2, 133.8, 129.6, 128.9, 128.3, 127.9, 127.8, 127.6, 100.1, 56.1, 52.7, 43.4, 12.5.

Ethyl ((Z)-(((Z)-1-(4-chlorobenzyl)-5-(thien-2-yl)-1,2-dihydro-3H-pyrazol-3-ylidene)amino)(diethylamino)methylene)sulfamate ***90c***



General procedure: A solution of **64d** (53 mg, 0.12 mmol) in ethanol was treated with palladium agent (20 mol%) and stirred at room temperature under an atmosphere of H_2. The mixture was filtered through a short pad of Celite, which was rinsed with CH_2Cl_2 (5

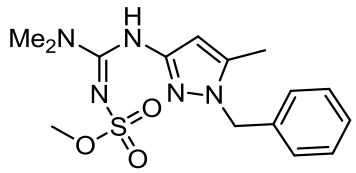
mL). The combined filtrate was evaporated and the residue was purified via column chromatography.

The general procedure was carried out with 1.5 mL of ethanol and 10%Pd/C (25 mg, 0.024 mmol) (25h). Elution with 10% EtOAc in CH_2Cl_2 gave the *title compound* as a colourless gum (42 mg, 71%), R_F 0.6 (10%EtOAc:CH_2Cl_2).

The general procedure was carried out with 2.5 mL of ethanol and 20%Pd/$(OH)_2$ (0.025 g, 0.024 mmol) (18h). Elution with CH_2Cl_2 gave the *title compound* as a colourless gum (32 mg, 50%), R_F 0.6 (10%EtOAc:CH_2Cl_2).

90c obtained as a colourless gum. (Found: $[M+H]^+$ 496.1240; $C_{21}H_{27}N_5O_3{}^{32}S_2Cl$ requires $[M+H]^+$ 496.1238). δH (400MHz, $CDCl_3$) 8.13 (1H, s, NH), 7.42 (2H, dd, *J* 2.4, 1.0, thienyl H3), 7.28 (2H, d, *J* 6.8, ArH), 7.07 (1H, dd, *J* 5.1, 1.0Hz, theinyl H5), 7.04-7.01 (3H, m, ArH + thienyl H4), 6.10 (1H, s, CH), 5.29 (2H, s, C*H*$_2$Ar), 4.26 (2H, q, *J* 7.1, OC*H*$_2$CH$_3$), 3.36 (4H, q, 7.1, NCH_2 x 2), 1.34 (3H, t, *J* 7.1, OCH_2CH_3), 1.10 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 156.6, 147.3, 138.3, 135.2, 133.7, 129.7, 128.9, 128.3, 127.8, 127.8, 127.5, 100.0, 66.2, 52.7, 43.3, 14.8, 12.5.

Methyl (E)-(((1-benzyl-5-methyl-1H-pyrazol-3-yl)amino)(dimethylamino)methylene)sulfamate ***90d***

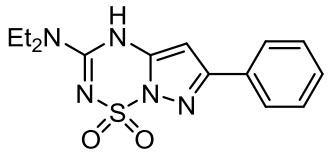


A solution of **64f** (41 mg, 0.128 mmol) in methanol (1.5 mL) was stirred at room temperature for 21 hrs. The solvent was then evaporated to give the *title compound* as a colourless oil (45 mg, 100% crude).

A solution of **64f** (40 mg, 0.125 mmol) in methanol (1.5 mL) was stirred at reflux temperature for 4 hrs. The mixture was cooled and solvent evaporated to give the *title compound* as a colourless oil (44 mg, 100% crude).

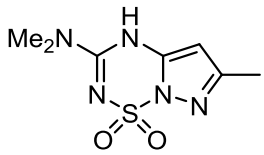
90d obtained as a colourless oil. (Found: $[M+Na]^+$ 374.1264; $C_{15}H_{21}N_5O_3{}^{32}SNa$ requires $[M+H]^+$ 374.1263). δH (600MHz, $CDCl_3$) 8.21 (1H, s, NH), 7.32-7.29 (3H, m, ArH), 7.06 (2H, d, *J* 7.3, ArH), 5.71 (1H, s, CH), 5.15 (2H, s, C*H*$_2$Ar), 3.80 (3H, s, OCH_3), 2.88 (6H, s, NCH_3 x 2), 2.17 (3H, s, CH_3). δC (100MHz, $CDCl_3$) 157.0, 146.1, 140.6, 136.6, 128.9, 127.9, 126.8, 89.8, 56.2, 52.9, 39.5, 11.5.

3-Diethylamino-6-phenyl-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***34e***



A solution of **64b** (51 mg, 0.11 mmol) in EtOAc (1 mL) was treated with $Pd(OH)_2/C$ (15 mg, 0.056 mmol) and the mixture stirred vigorously under a H_2 atmosphere at room temperature for 46 hrs. The suspension was diluted with CH_2Cl_2 (5 mL) and filtered through a short plug of Celite which was washed with CH_2Cl_2 (1 mL). The combined filtrate was evaporated and the residue was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* (34 mg, 93%) as colourless blocks.

3-Dimethylamino-6-methyl-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide ***34j***



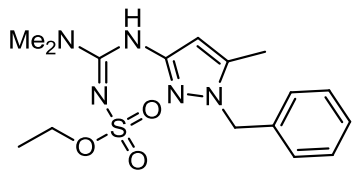
A solution of **64f** (33 mg, 0.1 mmol) in dry *tert*-butanol (1.5 mL) was treated with 20wt% $Pd(OH)_2/C$ (11 mg, 0.02 mmol) and stirred vigorously under a H_2 atmosphere at 35°C for 22 hrs. The mixture was diluted with CH_2Cl_2 (5 mL) and filtered through a short pad of Celite. The filtrate was evaporated and the residue was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* as colourless needles (18 mg, 79%) R_F 0.5 (10%MeOH:CH_2Cl_2).

A solution of **64f** (32 mg, 0.1 mmol) in ethyl acetate (1.2 mL) was treated with 20%wt $Pd(OH_2)/C$ (12 mg, 0.02 mmol) and stirred vigorously under a H_2 atmosphere at room temperature for 46 hrs. The suspension was diluted with CH_2Cl_2 (5 mL) and filtered through a short pad of Celite. The filtrate was evaporated and the residue was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* as colourless needles (19 mg, 83%).

A solution of **64f** (33 mg, 0.1 mmol) in ethyl acetate (1.0 mL) was treated with 20%wt $Pd(OH_2)/C$ (26 mg, 0.05 mmol) and stirred vigorously under a H_2 atmosphere at room temperature for 44 hrs. The suspension was diluted with CH_2Cl_2 (5 mL) and filtered through a short pad of Celite. The filtrate was then evaporated and the residue was purified via column chromatography (10% MeOH in CH_2Cl_2) to give the *title compound* as colourless prisms (22 mg, 96%).

Ethyl (E)-(((1-benzyl-5-methyl-1H-pyrazol-3-yl)amino)(dimethylamino)methylene)sulfamate ***90e*** *and 3-dimethylamino-6-methyl-7H-pyrazolo[1,5-b][1,2,4,6]thiatriazine 1,1-dioxide* ***34j***

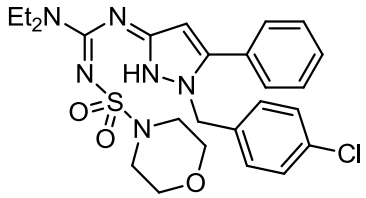
A suspension of **64f** (32 mg, 0.1 mmol) in ethanol (1.5 mL) was treated with 10%wt Pd/C (12 mg, 0.011 mmol) and stirred vigorously under an atmosphere of H_2 for 42 hrs. The reaction mixture was diluted with CH_2Cl_2 (5 mL) and filtered through a short pad of Celite. The filtrate was evaporated and the reside was purified by column chromatography to give i) **90e** as a colourless oil (19 mg, 52%) R_F 0.5 (10%EtOAc in CH_2Cl_2) and ii) **34j** (11 mg, 48%) R_F 0.4 (30%EtOAc:CH_2Cl_2).



90e obtained as a colourless oil. (Found: $[M+Na]^+$ 388.1433; $C_{16}H_{23}N_5O_3{}^{32}SNa$ requires $[M+Na]^+$ 388.1419). δH (400MHz, $CDCl_3$) 8.22 (1H, s, NH), 7.34-7.25 (3H, m, ArH), 7.07 (2H, d, J 6.7, ArH), 5.72 (1H, s, pyrazole H4), 5.16 (2H, s, CH_2Ar), 4.21 (2H, q, J 7.1, OCH_2CH_3); 2.89 (6H, s, NCH_3 x 2), 2.18 (3H, s, CH_3), 1.31 (3H, t, J 7.1,

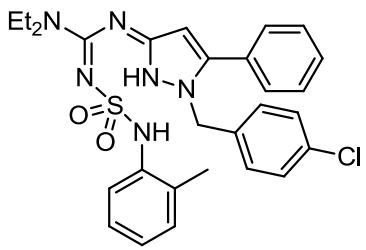
OCH_2CH_3). δC (100MHz, $CDCl_3$) 156.9, 146.1, 140.3, 136.5, 128.8, 127.8, 126.7, 98.6, 66.1, 52.8, 39.3, 14.8, 11.3.

Morpholino ((Z)-(((Z)-1-(4-chlorobenzyl)-5-phenyl-1,2-dihydro-3H-pyrazol-3-ylidene)amino)(diethylamino)methylene)sulfonamide ***90f***



Morpholine (100 mg, 1.1 mmol) was added to a stirred suspension of **34e** (300 mg, 0.68 mmol) in acetonitrile (6 ml) and the mixture was simmered for 1 hr. The mixture was evaporated *in vacuo* and the residue was dissolved in hot isopropanol (4 ml). On cooling the solution a colourless tar was deposited which gradually solidified upon standing for several days. The solid was collected and recrystallised from ethanol to give the *title compound* (238 mg, 66%) as colourless prisms, m.p. 151-153°C. (Found: $[M+Na]^+$ 553.1751; $C_{25}H_{31}N_6O_3{}^{32}S_2ClNa$ requires $[M+Na]^+$ 553.1765). δH (400MHz, $CDCl_3$) 8.27 (1H, s, NH), 7.43-7.41 (3H, m, ArH), 7.29-7.24 (4H, m, ArH (Ph & p-ClBn)), 6.98 (1H, d, *J* 8.4, ArH), 6.01 (1H, d, pyrazole CH), 5.16 (2H, s, NCH_2Ar), 3.74 (4H, m, morpholino NCH_2 x 2), 3.34 (2H, q, *J* 7.1, NCH_2CH_3 x 2), 3.15 (4H, m, morpholino OCH_2 x 2), 1.09 (6H, t, *J* 7.1, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 156.5, 147.6, 145.7, 135.5, 133.6, 129.6, 129.2, 128.9, 128.8, 128.4, 99.2, 66.2, 52.4, 46.8.

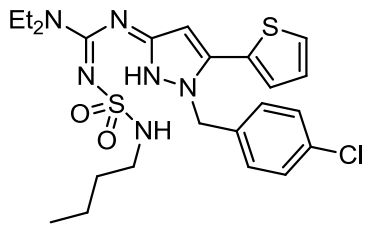
N-((Z)-(((Z)-1-(4-Chlorobenzyl)-5-phenyl-1,2-dihydro-3H-pyrazol-3-ylidene)amino)(diethylamino)methylene)o-tolyl-amino-4-sulfonamide ***90g***



A solution of **64b** (55 mg, 0.124 mmol) and o-toluidine (20 µl, 0.25 mmol) in acetonitrile (1.2 mL) was heated to reflux temperature until TLC indicated completion (2 hrs). The mixture was evaporated to give a green oil which was purified via column chromatography (10% EtOAc in CH_2Cl_2) to give the *title compound* as a colourless gum (45 mg, 68%) R_F 0.4 (10%EtOAc:CH_2Cl_2).

90g obtained as a colourless gum. (Found: $[M+Na]^+$ 573.1815; $C_{28}H_{31}N_6O_2{}^{32}SClNa$ requires $[M+Na]^+$ 573.1815). δH (400MHz, $CDCl_3$) 8.29 (1H, s, NH), 7.50 (1H, d, *J* 8.0, ArH), 7.39-7.37 (3H, m, ArH), 7.24-7.18 (4H, m, ArH), 7.06-7.03 (2H, m, ArH), 6.94-6.88 (3H, m, ArH), 6.28 (1H, s, N*H*Ar), 5.72 (1H, s, pyrazole CH), 5.09 (2H, s, NCH_2Ar), 3.20 (4H, q, *J* 7.0, NCH_2CH_3 x 2), 2.17 (3H, s, CH_3), 0.91 (6H, t, *J* 7.0, NCH_2CH_3 x 2). δC (100MHz, $CDCl_3$) 156.3, 147.4, 145.5, 136.7, 135.4, 133.6, 130.4, 129.6, 129.2, 128.9, 128.7, 128.5, 128.4, 126.8, 124.0, 121.2, 98.9, 52.4, 43.0, 17.7, 12.4.

Butyl ((Z)-(((Z)-1-(4-chlorobenzyl)-5-(thien-2-yl)-1,2-dihydro-3H-pyrazol-3-ylidene)amino)(diethylamino)methylene)sulfonamide ***90h***

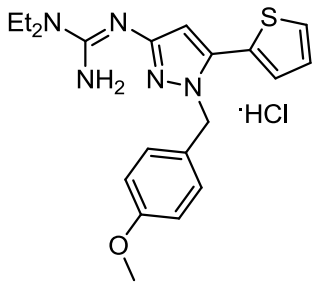


A solution of **64d** (54 mg, 0.12 mmol) and *n*-butyl amine (13 mg, 0.18 mmol) in acetonitrile (1 mL) was heated to 50°C for 45 minutes. The solvent was evaporated to give a colourless oil which was purified via column chromatography (20% EtOAc in CH_2Cl_2) to give the *title compound* (45 mg, 72%) as a colourless oil.

90h obtained as colourless oil. (Found: $[M+Na]^+$ 545.1539; $C_{23}H_{31}N_6O_2{}^{32}S_2ClNa$ requires $[M+Na]^+$ 545.1536). δH (400MHz, $CDCl_3$) 8.32 (1H, s, NH), 7.41 (1H, dd, *J* 5.1, 0.8, thienyl-H5), 7.26 (2H, m, ArH), 7.06 (1H, dd, *J* 3.6, 1.3, thienyl-H3), 7.06-7.00 (3H, m, thienyl-H4, ArH), 6.09 (1H, d, pyrazole CH), 5.28 (2H, s, NCH_2Ar), 4.10 (1H, br m, N*H*Butyl), 3.34 (2H, q, *J* 7.1, NCH_2CH_3 x 2), 3.08 (2H, br m, $NCH_2CH_2CH_2CH_3$),

1.50 (4H, q, J 7.4, $NCH_2CH_2CH_2CH_3$), 1.31 (2H, q, J 7.6, $NCH_2CH_2CH_2CH_3$), 1.08 (6H, t, J 7.1, NCH_2CH_3 x 2), 0.88 (3H, t, J 7.4, $NCH_2CH_2CH_2CH_3$). δC (100MHz, $CDCl_3$) 156.5, 147.9, 138.3, 135.3, 133.7, 129.8, 128.9, 128.2, 127.8, 127.4, 99.9, 52.7, 43.5, 43.0, 31.6, 29.7, 19.9, 13.6, 12.6.

(E)-1,1-Diethyl-2-(1-(4-methoxybenzyl)-5-(thien-2-yl)-1H-pyrazol-3-yl)guanidinium chloride ***91***

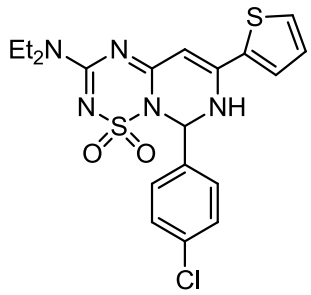


A solution of **64g** (40 mg, 0.09 mmol) in CH_2Cl_2 (0.5 mL) was treated with TFA (0.1 mL, 1.3 mmol) and the solution was stirred at room temperature until TLC indicated consumption of starting material (5 hrs). The mixture was evaporated to leave a yellow residue which was dissolved in CH_2Cl_2 (5 mL) and HCl was bubbled through the solution to generate the hydrochloride salt. The crude salt was purified via column chromatography (20% MeOH in CH_2Cl_2) to afford the *title compound* (18 mg, 48%) as a colourless semi-solid.

Recrystallisation of **91** from MeOH gave colourless blocks, m.p. 116-119°C (Found: $[M+H]^+$ 384.1852; $C_{20}H_{26}N_5O_1{}^{32}S$ requires $[M+H]^+$ 384.1853). δH (400MHz, DMSO-d_6) 8.45 (1H, s, NH), 7.69 (1H, dd, *J* 5.1, 1.2, thienyl H5), 7.27 (1H, dd, *J* 3.5, 1.2, thienyl H3), 7.16 (1H, dd, *J* 5.1, 3.5, thienyl H4), 7.01 (2H, d, *J* 8.8, ArH), 6.88 (2H, d, *J* 8.8, ArH), 6.49 (2H, s, NH_2), 6.22 (1H, s, CH), 5.31 (2H, s, CH_2Ar), 3.71 (3H, s, OCH3), 3.25 (4H, q, *J* 7.0, NCH_2 x 2), 1.02 (6H, t, *J* 7.0, CH_3 x 2). δC (100MHz, DMSO-d_6) 159.1, 155.2, 148.0, 137.7, 130.1, 129.6, 128.6, 128.5, 128.4, 128.2, 114.4, 99.2, 55.5, 52.7, 42.9, 13.0. #Short and long range HETCOR also consistent with assigned structure.

9.16 Synthesis of a dihydropyrimido[1,6-*b*][1,2,4,6]thiatriazine

8-(4-Chlorophenyl)-3-diethylamino-6-(thien-2-yl)-7,8-dihydropyrimido[1,6-b][1,2,4,6]thiatriazine 1,1-dioxide ***92***



Sodium metal (20 mg, 0.87 mmol) was added to NH_3 (3 mL)* at -78°C. The deep blue mixture was treated with a solution of **64d** (53 mg, 0.12 mmol) in THF (5 mL) added dropwise over 1 minute. The vibrant yellow mixture was treated with ethanol (15 µL, 0.25 mmol) and stirred for a further 15 minutes then warmed to room temperature. The reaction mixture was quenched with saturated aqueous NH_4Cl (1 mL), diluted with water (10 mL), and extracted with EtOAc (2 x 10 mL). The combined organic extracts were dried and evaporated to a yellow oil which was purified via column chromatography (CH_2Cl_2) to afford the *title compound* (44 mg, 82%) as a bright yellow semi-solid. R_F 0.35 (CH_2Cl_2).

*A round-bottomed flask with solid NaOH was fitted with a fractionating column packed with CaO, and a still-head leading into a 3-necked round bottom flask under at atmosphere of nitrogen at -65°C. Saturated aqueous NH_4Cl was added slowly to the NaOH, and the ammonia evolved was collected at -65°C until approximately 3 mL of liquid formed.

Recrystallisation of **92** from CH_2Cl_2 gave yellow blocks, m.p. 230-232°C. (Found $[M+H]^+$ 450.0821; $C_{19}H_{21}O_2N_5{}^{32}S_2Cl$ requires $[M+H]^+$ 450.0820). δH (400MHz, $CDCl_3$) 7.59 (1H, dd, *J* 3.8, 2.9, thienyl H3), 7.55 (1H, dd, *J* 5.0, 0.9, theinyl H5), 7.43 (2H, d, *J* 8.6, ArH), 7.26 (2H, d, *J* 8.6, ArH), 7.15 (1H, dd, *J* 5.0, 3.8, thienyl H4), 6.93 (1H, d, *J* 6.3, CH), 6.84 (1H, br s, NH), 5.62 (1H, s, H5), 3.64-3.53 (4H, m, 2 x NCH_2CH_3), 1.26 (3H, t, *J* 8.3, NCH_2CH_3), 1.19 (3H, t, *J* 7.1, NCH_2CH_3). δC (150MHz,

$CDCl_3$) 158.1, 156.2, 148.9, 136.0, 135.8, 134.6, 130.7, 129.1, 128.6, 128.6, 127.5, 127.4, 94.1, 63.1, 42.27, 42.25, 13.7, 12.9. #Short and long range HETCOR collected. NOESY experiments also collected, nOe between benzyl and thienyl substituents.

9.17 X-ray Crystallography

Single crystals suitable for X-ray diffraction experiments were covered in Paratone-N oil and mounted on a glass fibre. Data ($2\Theta_{max}$ 55°) were collected at 298 K, 150 K or 110 K using an Oxford Diffraction X-Calibur X-ray diffractometer and $Mo_{k\alpha}$ (λ 0.71073 Å) radiation. After integration and scaling, the datasets were merged into N unique reflections (R_{int}). The structures were solved using conventional methods and refined by using full-matrix least-squares using the SHELX-97 software[214] in conjunction with the X-Seed interface.[215] Non-hydrogen atoms were refined with anisotropic thermal parameters and hydrogen atoms were placed in calculated positions.

CCDC 939838-939845, 1035053-1035059, 1022791-1022794, 1042481 contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre via www.CCDC.cam.ac.uk/data_request/cif.

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APPENDICES

The appendices herein contain supplementary data which has been referenced within the body of text in the results/discussion sections of this thesis. For additional supplementary information, please refer to the publications associated with this body of work:

Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2013**, *66*, 1323. 'The First Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine Derivatives and their Unusual Reactions with Acylating Agents'

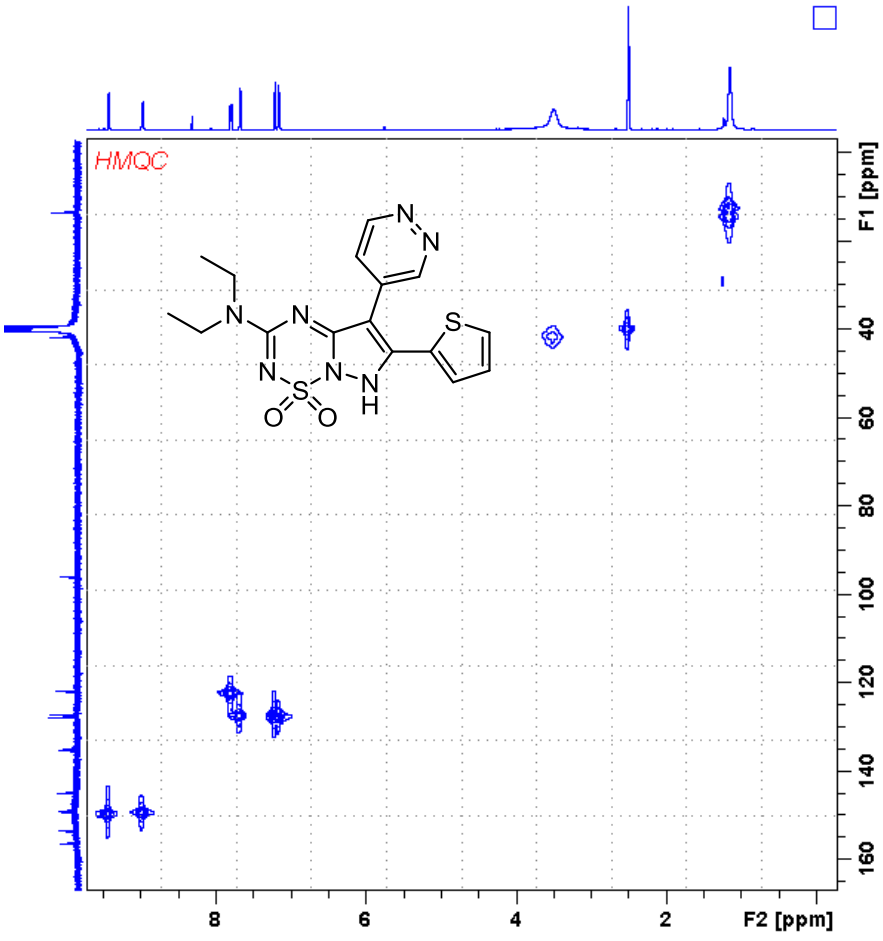
Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2015**, 68, *In press.* 'Substitution Reactions of Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-Dioxides.'

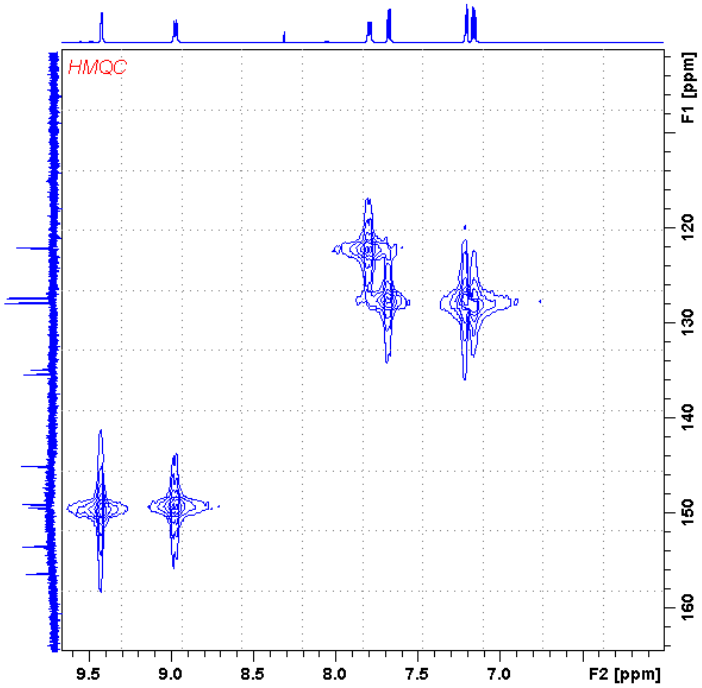
Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2015**, *In press.* 'Synthesis and Reactivity of Novel Pyrazolo[3,4-*e*][1,2,4]Thiadiazine Derivatives.'

Rebecca E. Norman; Michael V. Perkins; Andris J. Liepa; Craig L. Francis, *Australian Journal of Chemistry* **2014**, *In CSIRO internal review.* 'Cleavage and Rearrangement of Pyrazolo[1,5-*b*][1,2,4,6]thiatriazine 1,1-Dioxides.'

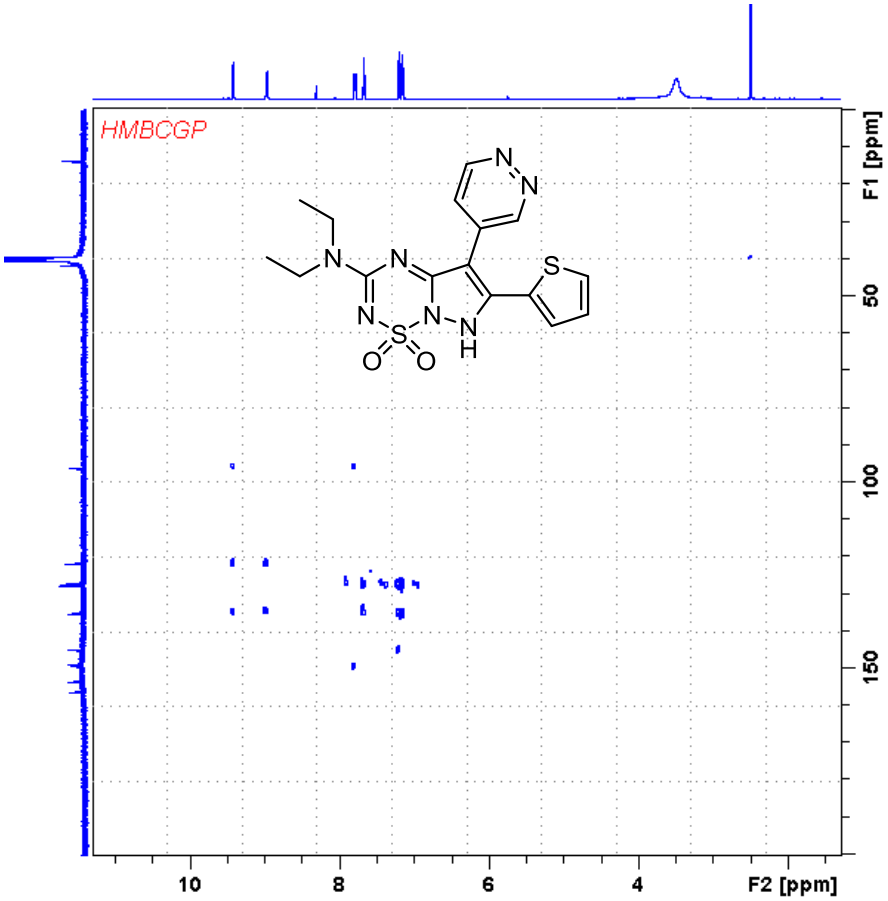
Appendix A: Spectral data for chapter 4

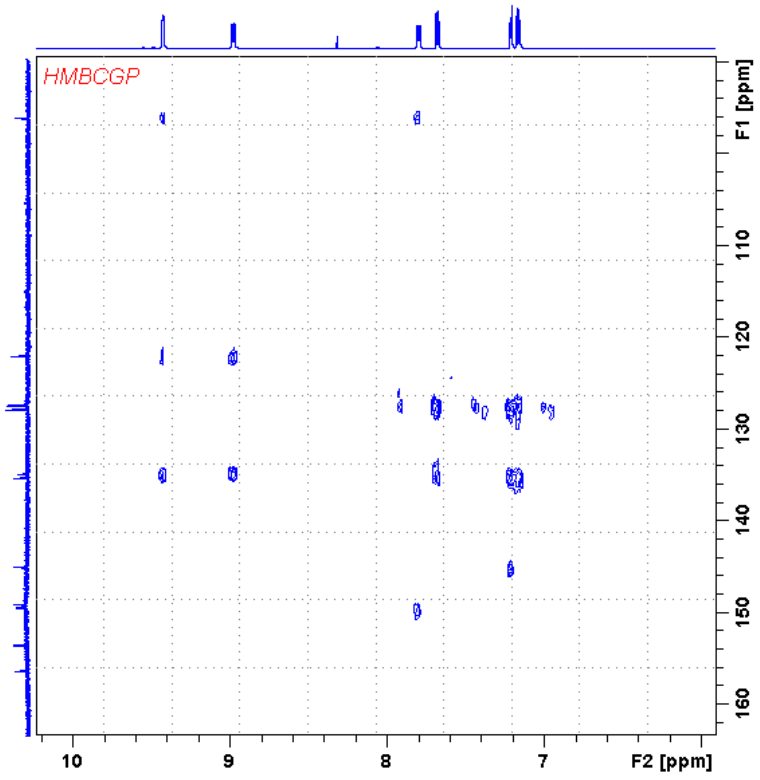
Short range correlation HMQC spectrum of pyridazine adduct **55c** (DMSO-d^6)





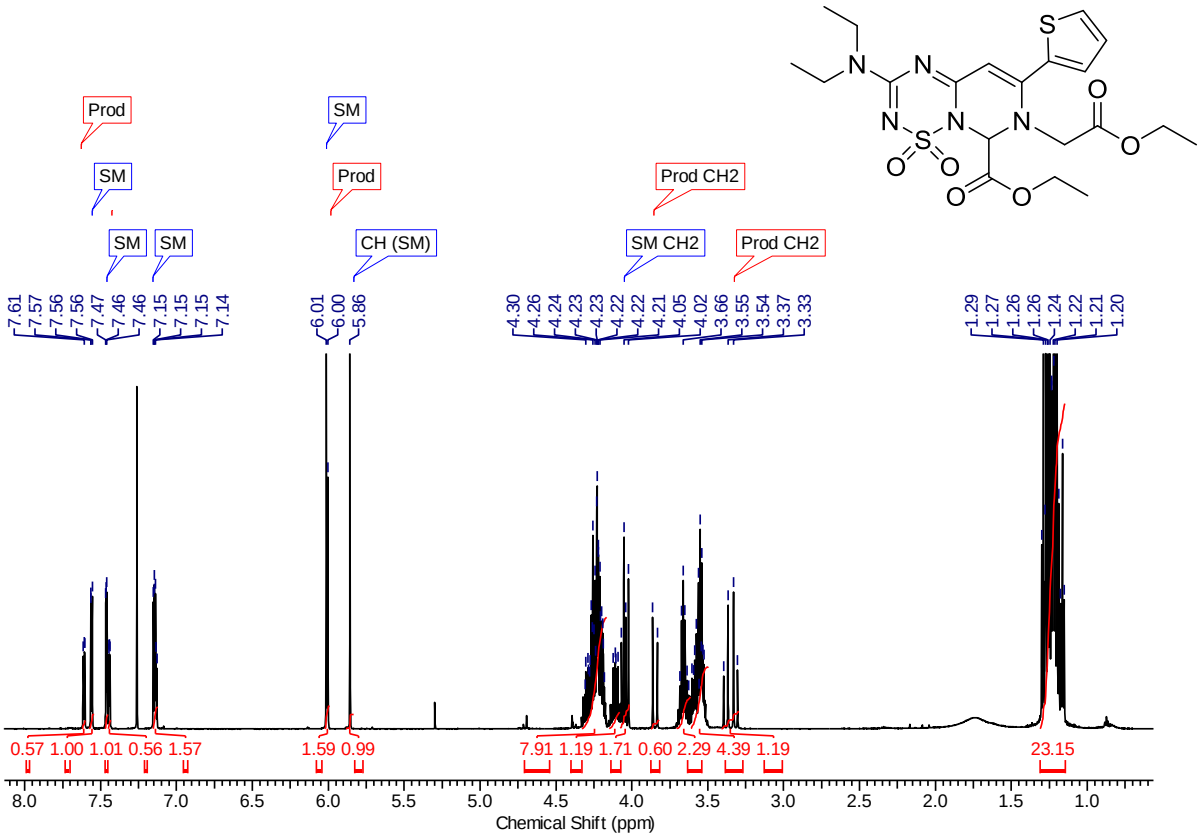
Long range correlation HMBC spectrum of pyridazine adduct **55c** (DMSO-d^6)

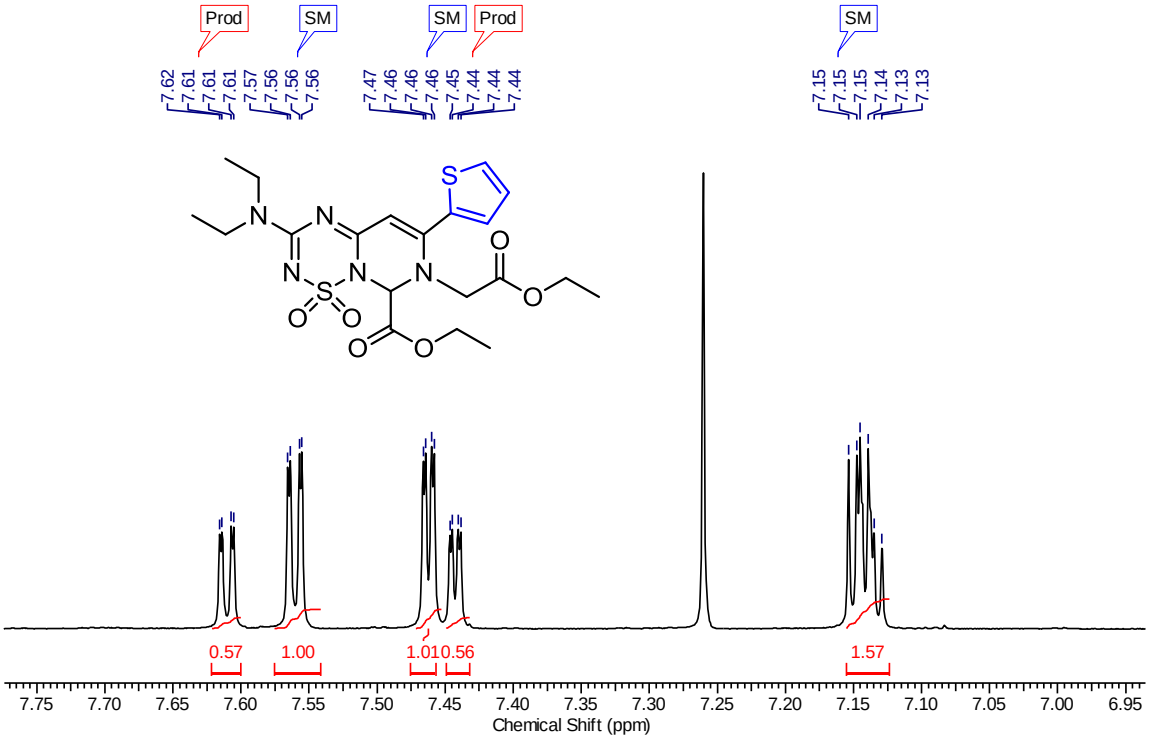


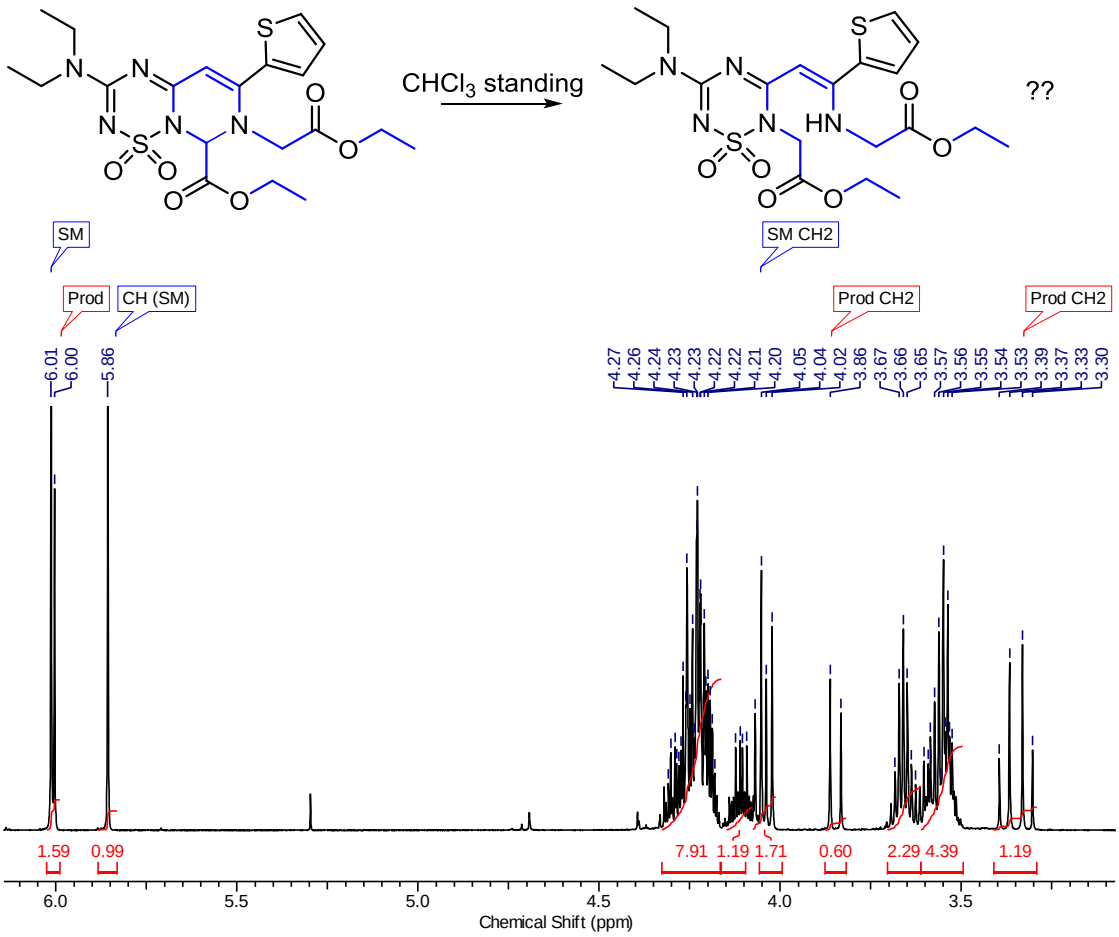


Appendix B: Spectral data for chapter 5

^{1}H NMR spectrum of **74a** upon standing in $CDCl_3$ 600MHz ($CDCl_3$)

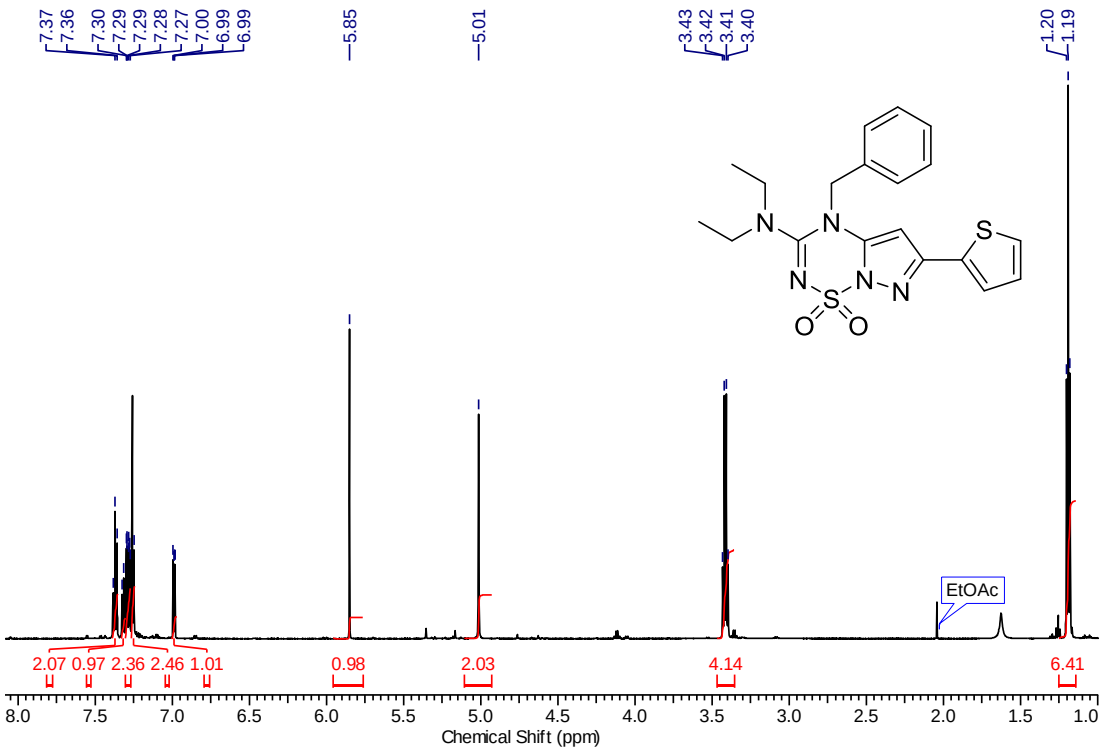




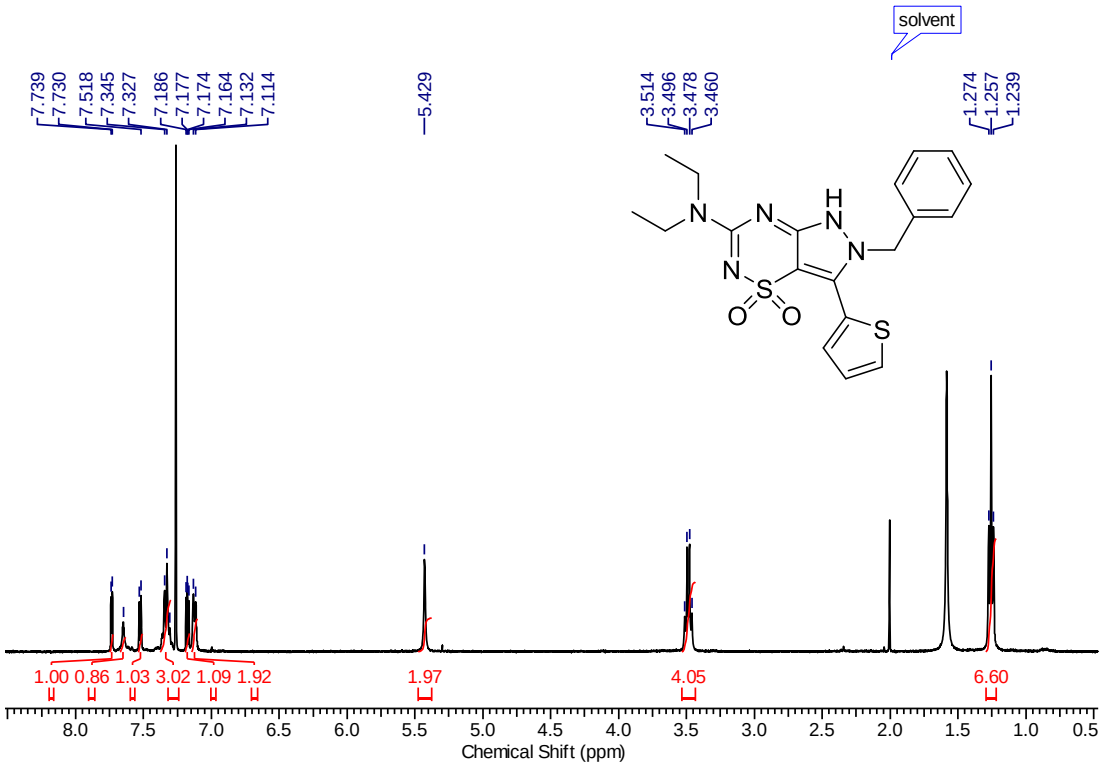

CHCl3 standing
??
SM
Prod
CH (SM)
SM CH2
Prod CH2
Prod CH2
6.01
6.00
5.86
4.27
4.26
4.24
4.23
4.23
4.22
4.22
4.21
4.20
4.05
4.04
4.02
3.86
3.67
3.66
3.65
3.57
3.56
3.55
3.54
3.53
3.39
3.37
3.33
3.30
1.59
0.99
7.91
1.19
1.71
0.60
2.29
4.39
1.19
6.0
5.5
5.0
4.5
4.0
3.5
Chemical Shift (ppm)

Appendix C: Spectral data for chapter 7

^{1}H NMR spectrum of **64e** 600MHz ($CDCl_3$)



^{1}H NMR spectrum of **89b** 600MHz ($CDCl_3$)



Appendix D: X-ray crystallography data

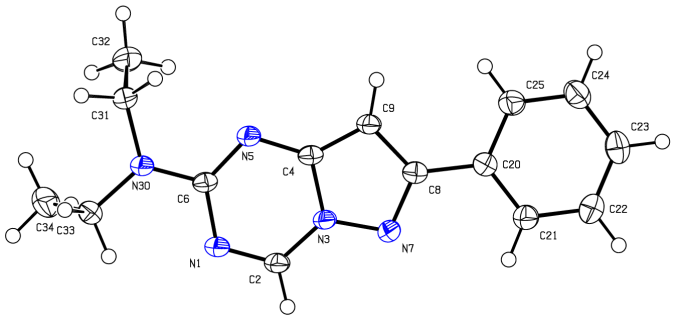


Table 22: Crystal data and Structure Refinement for **83a**

Parameter	**83a**
Empirical formula	C15 H17 N5
Formula weight	267.34
Temperature (K)	150
Wavelength (Å)	0.071073
Crystal system	triclinic
Space group	'P - 1'
Unit cell dimensions	a=6.0239(3) b=8.1736(4) c=14.5925(7)
Theta max	29.27
Reflections used	2985
Final R indices [I > 2σ(I)]	
R1	0.0473
*w*R2	0.1511
Data completeness	0.894

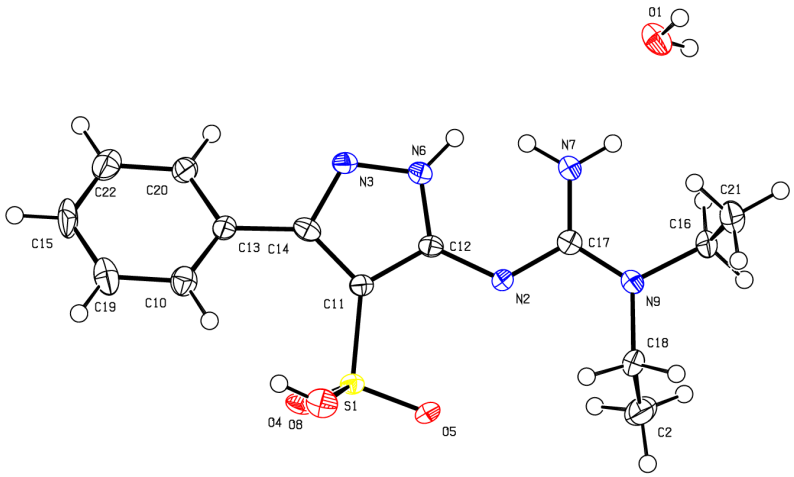


Table 22: Crystal data and Structure Refinement for **85**

Parameter	**85**
Empirical formula	C14 H19 N5 O3 S, H2 O
Formula weight	355.42
Temperature (K)	150
Wavelength (Å)	0.071073
Crystal system	orthorhomic
Space group	'P 21 21 21'
Unit cell dimensions	a=7.1232(3) b=12.4682(6) c=18.9247(6)
Theta max	28.85
Reflections used	1501
Final R indices [I > 2σ(I)]	
R1	0.0455
*w*R2	0.1179
Data completeness	1.11/0.64

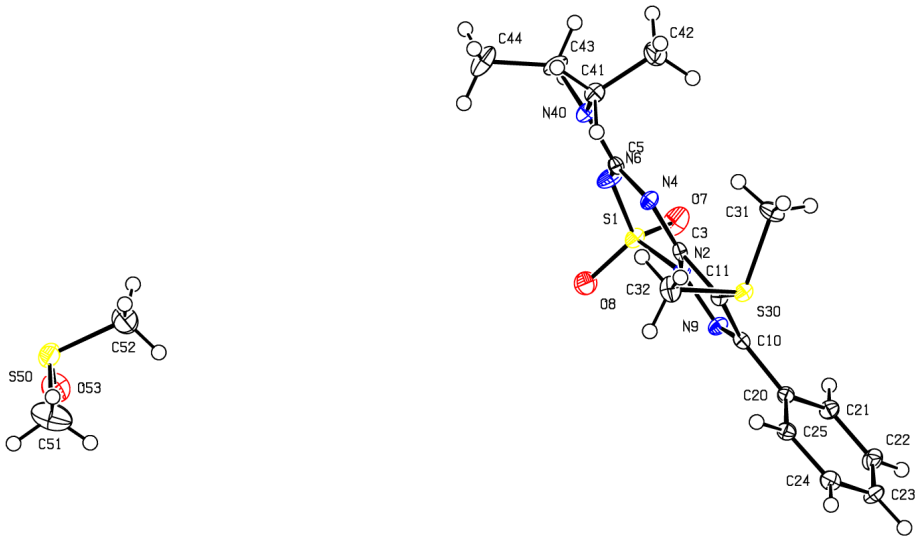


Table 23: Crystal data and Structure Refinement for **62a**

Parameter	**62a**
Empirical formula	C16 H21 N5 O2 S2, C2 H6 O S
Formula weight	457.63
Temperature (K)	298
Wavelength (Å)	0.071073
Crystal system	orthorhomic
Space group	'P 21 21 21'
Unit cell dimensions	a=8.1953(4) b=14.2276(7) c=18.5117(9)
Theta max	29.36
Reflections used	1501
Final R indices [I > 2σ(I)]	
R1	0.0422
*w*R2	0.1067
Data completeness	1.53/0.86

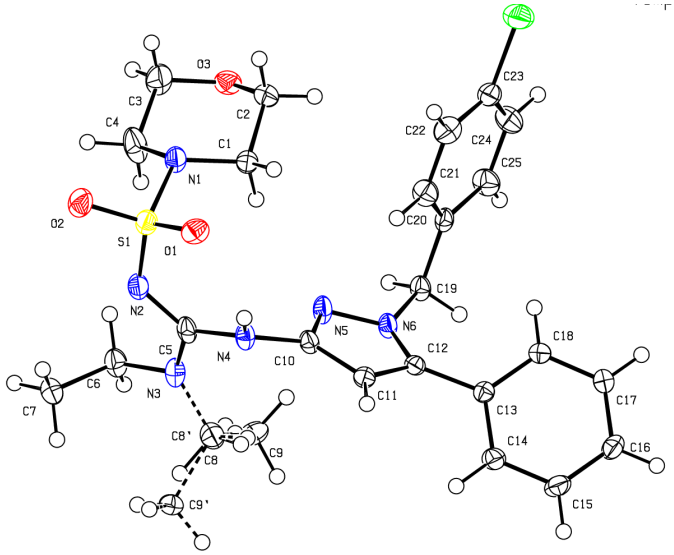


Table 24: Crystal data and Structure Refinement for **90f**

Parameter	**90f**
Empirical formula	C25 H31 Cl N6 O3 S
Formula weight	531.08
Temperature (K)	123
Wavelength (Å)	0.071073
Crystal system	triclinic
Space group	'P - 1'
Unit cell dimensions	a=9.4776(5) b=9.8831(7) c=14.1671(7)
Theta max	27.50
Reflections used	9464
Final R indices [I > 2σ(I)]	
R1	0.0341
*w*R2	0.0895
Data completeness	1.00