SYNTHESIS OF PYRROLIZIDINE ALKALOID ANALOGUES AS POTENTIAL ANTI-TUMOUR AGENTS

A thesis presented in part fulfilment of the requirements for the Degree of

Doctor of Philosophy

by

GEORGE BAXTER

Department of Chemistry

University of Glasgow

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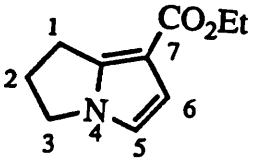
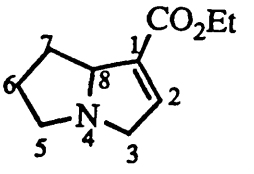
Finally, thanks go to my wife Beverley for proof-reading this Thesis and for many useful discussions on Chemistry.

NOTES ON NOMENCLATURE

Pyrrolizidine derivatives with one or two double bonds are named as derivatives of 1H- or 3H-pyrrolizine in accordance with Chemical Abstracts nomenclature.

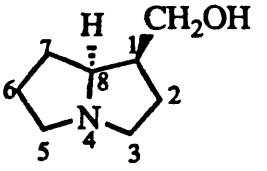


Ethyl 2,3-dihydro-1H-pyrrolizine-7-carboxylate



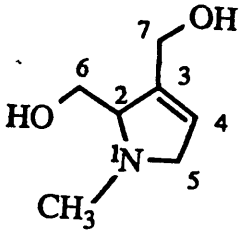
Ethyl 5,6,7,8-tetrahydro-3H-pyrrolizine-1-carboxylate

Fully saturated compounds are named as pyrrolizidine derivatives. Stereochemistry of substituents is indicated by α- and β-nomenclature to conform with the usual practice in this field.



1β-Hydroxymethyl-8α-pyrrolizidine (isoretronecanol).

Monocylic analogues of pyrrolizidine alkaloids are named as pyrroline or pyrrolidine derivatives.



2,3-Bishydroxymethyl-1-methyl-3-pyrroline (or -2,5-dihydropyrrole) (Synthanecine A).

Alternatively, they are named as derivatives of Synthanecine A itself.

ABBREVIATIONS

d	=	doublet
DBU	=	1,8-diazabicyclo[5.4.0]undec-7-ene
DIBAH	=	di-isobutylaluminium hydride
DMF	=	N,N-dimethyl formamide
DME	=	1,2-dimethoxyethane
HMPA	=	hexamethylphosphoramide
i.r.	=	infra-red
LDA	=	lithium diisopropylamide
m	=	multiplet
m.s.	=	mass spectrometry
m.p.	=	melting-point
n.m.r.	=	nuclear magnetic resonance
P.A.	=	pyrrolizidine alkaloid
s	=	singlet
t	=	triplet
TEA	=	triethylamine
THF	=	tetrahydrofuran
t.l.c.	=	thin layer chromatography
T.M.S.	=	Trimethylsilyl
U.V.	=	ultra-violet

TABLE OF CONTENTS

SUMMARY

This Thesis covers three areas of research relating to Pyrrolizidine Alkaloids: (a) Synthesis of PA analogues based on Synthanecine A [(±)-2,3-bis-hydroxymethyl-1-methyl-3-pyrroline]; (b) Synthesis of derivatives of supinidine, and (c) Use of 1-pyrroline aimed towards the synthesis of PA analogues.

(a) Synthesis of Pyrrolizidine Alkaloid Analogues based on Synthanecine A. - The synthesis of PA analogues based on Synthanecine A has been achieved. Treatment of Synthanecine A with various acid chlorides, chloroformates and isocynates, yielded ester, carbonate and carbamate derivatives respectively. Reaction of two of these derivatives with hydrogen peroxide gave the corresponding N-oxides, both of which proved to be unstable compounds. A new monocyclic analogue [(±)-3-hydroxymethyl-1-methyl-3-pyrroline] of supinidine was prepared by a route analagous to that used for Synthanecine A.

(b) Synthesis of supinidine derivatives. - The first synthesis of 3,3-dimethylsupinidine has been accomplished. N-Alkylation of proline using dimethyl 2-bromo-2-methylpropylidenemalonate, followed by iminium ion formation and intramolecular cyclisation gave the required pyrrolizidine skeleton. Deethoxycarbonylation, introduction of unsaturation and reduction of the ester led to 3,3-dimethylsupinidine. Extension of this approach aimed towards

8-methylsupinidine proved unsuccessful. New methods of iminium ion cyclisation were developed, using bases such as 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) and triethylamine (TEA).

(c) Use of 1-pyrroline aimed towards the synthesis of PA analogues. - 1-Pyrroline (an unstable oil) was prepared by hydrolysis of γ-aminobutanal diethylacetal and found to form a stable solid complex with zinc iodide. The 1-pyrroline could be regenerated when needed by treating the complex in organic solution with a base such as triethylamine. Attempts to use this complex as a dienophile source aimed towards the synthesis of pyrrolizidine and indolizidine alkaloids proved unsuccessful. However, the complex reacted as a source of 1-pyrroline with a range of β-ketoacids to produce acylpyrrolidines in high yield. Thus, a good synthon for the construction of five-membered rings containing nitrogen is now available in a stabilised form.

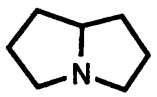
CHAPTER 1

INTRODUCTION

1.1 Pyrrolizidine Alkaloids

Pyrrolizidine Alkaloids (P.A.s) have been widely studied for many years, principally because of their toxicity and widespread occurrence.[1] The toxicity was first observed in the 19th Century by farmers whose cattle grazed on plants belonging to, commonly, the genera *Senecio* (family Compositae), *Crotalaria* (family Leguminosae), and *Heliotropium* (family Boraginaceae). The symptoms such as staggering and liver damage were known variously as "Winton disease" and "Pictou disease", amongst others.[2] Indeed, many diseases which were at one time thought to be unrelated have now been shown to be due to animals consuming plants which contained P.A.s.[3]

As their name implies, these alkaloids contain the pyrrolizidine skeleton (1), more formally known as 1-azabicyclo[3.3.0]octane, or related structures such as (2). The naturally occurring compounds are far more complex; many are unsaturated and contain a diol system such as in heliotridine (3). Mono-alcohols such as supinidine (4) are also known. These alcohols can be found as monoesters, diesters or macrocyclic diesters. The esterifying acids, known as necic acids (the amino-alcohols being known as necine bases) often contain 5 to 10 carbon atoms and are usually unsaturated and/or highly oxygenated.



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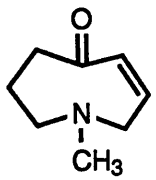

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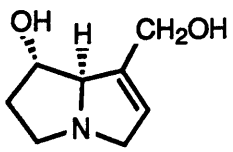

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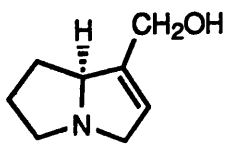

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1.2. Toxicity of Pyrrolizidine Alkaloids

P.A.s can cause heart and lung damage but their main toxic effect is on the liver. However, not all P.A.s are heptatotoxic: an investigation of the structural features of toxic P.A.s by Schoental[4] led her to suggest that there are two features common to toxic P.A.s:

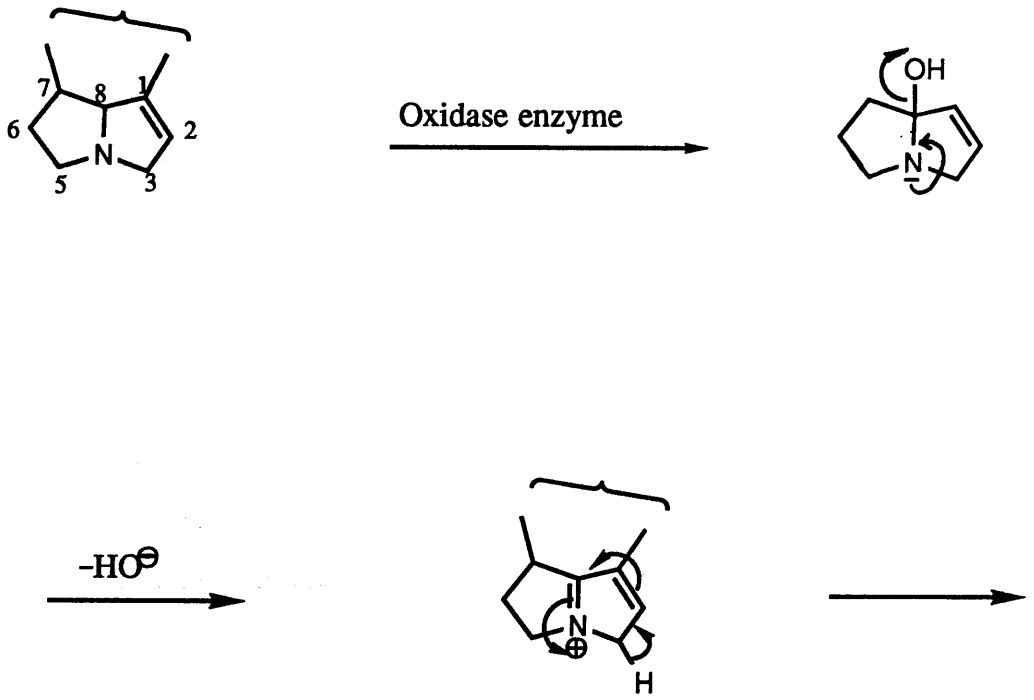
1. A 1,2 double bond;
2. An allylic ester group.

Whilst investigating the toxic action of P.A.s, Mattocks found dihydropyrrolizines in the urine of animals which had been exposed to P.A.s in the laboratory.[5] This led him to propose that it was not the P.A.s themselves that were responsible for the liver damage, but dihydropyrrolizines which were produced in the body from P.A.s. It is now thought that the dihydropyrrolizines are produced by the action of hepatic microsomal oxidase enzymes on P.A.s (Scheme 1). It was thought at one time that hydroxylation initially occurred at C-3.[6] However, more recent work points to hydroxylation on C-8 being favoured.[7]

This is only one possible metabolic pathway for P.A.s to follow in the body. It is possible for the alkaloids to be detoxified by either of the following routes:

1. N-oxidation by microsomal oxidases;
2. Hydrolysis by esterases.

SCHEME 1



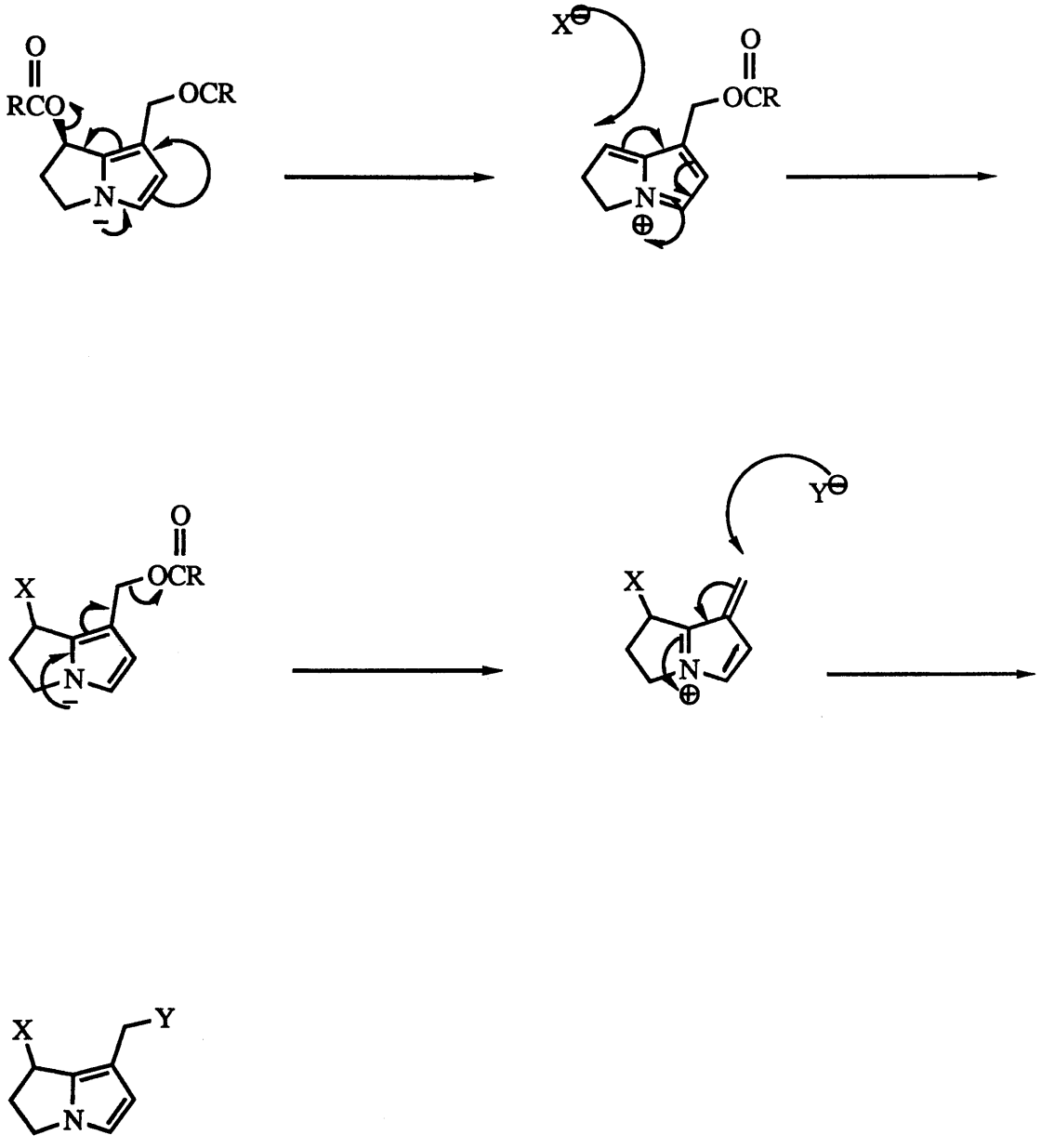
Both of these routes lead to detoxification as the products formed (N-oxides and amino-alcohols respectively) are much more water-soluble than esters or dihydropyrrolizines and so are excreted more quickly.

The toxic action of P.A.s is more complicated: Mattocks proposed that the pyrroles formed from P.A.s could act as bifunctional alkylating agents and that this was the mechanism of toxicity (Scheme 2). Possible reactants would be nucleic acids or other cell constituents bearing nucleophilic groups.

Structural factors play an important role in determining P.A. toxicity. One of the most important of these is the number of substituents close to the carbonyl group of the necic acid moiety or the amount of chain branching in the acid portion. As was stated earlier, one of the detoxification mechanisms is hydrolysis of the ester. Bulky substituents and chain branching will hinder esterase approach and therefore slow the rate of hydrolysis. For example, the hepatotoxicity of diacetyl retronecine (5) is only seen when the subject rat has been given an esterase inhibitor.[8] Confirming results have been obtained with a wide range of acids esterified to retronecine.[9]

Due to the numerous and varying symptoms associated with pyrrolizidine alkaloid poisoning and the delay usually encountered in observing these effects, it has often been difficult to locate the cause of these symptoms and remove the source. Indeed, in Jamaica[10] and other countries,[11] the inhabitants often use

SCHEME 2

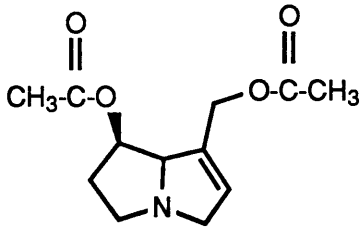


plants containing P.A.s in herbal remedies, and in the preparation of teas. Accidental contamination has also occurred[12, 13] due to the consumption of contaminated seeds and there are doubtless other cases where widespread poisoning has been attributed, perhaps mistakenly, to other factors.

1.3 Medicinal Uses of Pyrrolizidine Alkaloids

The alkylating ability of pyrrolizidine alkaloid metabolites has led several groups[14, 1] to investigate the tumour-inhibitory activity of P.A.s. Nine of the alkaloids tested showed activity against the tumours used and these all had structures capable of metabolism to pyrrolic derivatives. This work led others to investigate other alkaloids, notably indicine N-oxide (6) which has been shown to be the anti-tumour constituent of *Heliotropium indicum*.[15]. Due to its low hepatotoxicity relative to other P.A.s. with anti-tumour activity, indicine N-oxide has been used in clinical trials. These have shown that it is effective against advanced acute leukaemia[16] and gastro-intestinal cancer.[17] Zalkow *et al*.[18] prepared a semi-synthetic alkaloid (7) which was shown to be a more active anti-tumour agent than indicine N-oxide.

One other notable use of P.A.s has been the treatment of intestinal ulcers and hypertension in the U.S.S.R. using platyphylline (8). This compound lacks a 1,2-double bond and may therefore have reduced hepatotoxicity.



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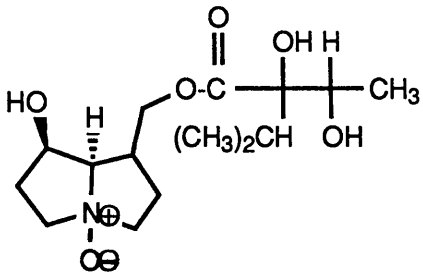

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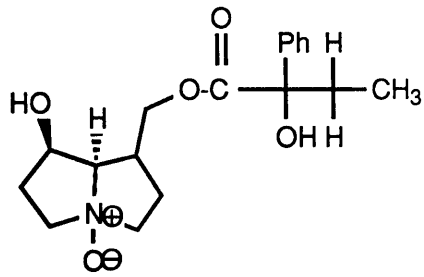

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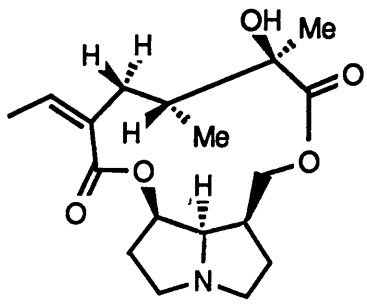


(8)

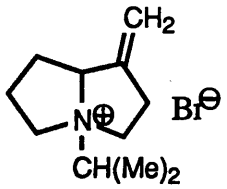
Success using N-oxides has led to the preparation of other derivatives containing quaternised nitrogen atoms; for example the quaternary ammonium derivatives (9) and (10) which showed ganglion and neuromuscular blocking activities respectively.[19]

1.4 Outline of Work Described in Thesis

The biological activity of P.A.s and the promise they have shown as anti-tumour agents has meant an increasing interest by P.A. chemists in the study of structure-activity relationships, and in the synthesis of analogues. The aims of this project were to select P.A. analogues and devise synthetic routes to them. These analogues might then be expected to show improved anti-tumour activity, combined with reduced hepatotoxicity. Existing routes to P.A.s and analogues are reviewed in Chapter 2.

Previous work in this area has shown that some monocyclic P.A. analogues produce similar toxic effects to P.A.s. The synthesis of new, monocyclic analogues is described in Chapter 3.

As has previously been discussed, one of the main causes of hepatotoxicity is thought to be pyrrole derivatives which can then form alkylating agents. P.A. analogues which cannot form pyrrole derivatives may show anti-tumour activity without hepatotoxicity. Selected examples of this type of compound have been synthesised and this work is discussed in Chapter 4.



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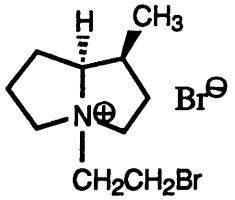

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1-pyrroline is a useful synthon for the construction of 5-membered systems. One of the major problems encountered in its use has been its instability. Methods for the stabilisation of 1-pyrroline and its use in alkaloid synthesis are described in Chapter 5.

CHAPTER 2

SYNTHESIS OF PYRROLIZIDINE ALKALOIDS AND ANALOGUES

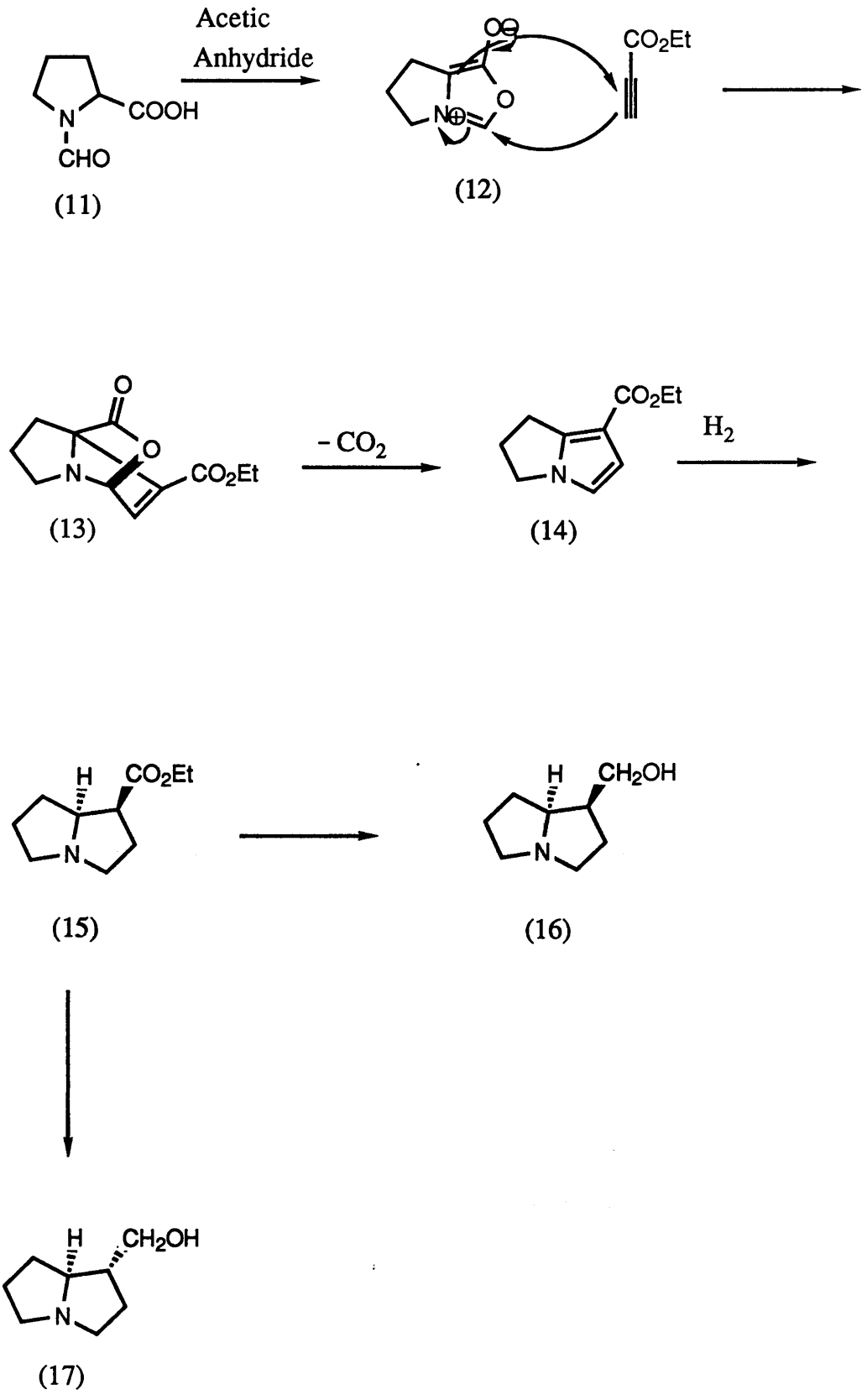
2.1 Introduction

It is not the intention of this section to provide a long, detailed list of previous syntheses of pyrrolizidine alkaloids, as these are already well documented.[1, 20] The following discussion will focus on some of the main routes used in pyrrolizidine alkaloid synthesis. It will also take into account work of direct relevance to this project, including the synthesis of P.A. analogues and the use of 1-pyrrolines as reagents in the synthesis of pyrrolizidine and indolizidine alkaloids.

2.2 The 1,3-Dipolar Cycloaddition Reaction

One particularly powerful method for construction of the pyrrolizidine system, which has attracted the attention of several groups is the 1,3-dipolar cycloaddition reaction, developed by Huisgen *et al*.[21] and adapted by Tufariello *et al*.[22] and Pizzorno and Albonico.[23] A convenient starting material for this reaction is *N*-formyl-proline (11) which when dehydrated by heating with acetic anhydride formed the oxazolium-5-oxide (12) (Scheme 3). Addition of ethyl propiolate gave the tricyclic structure (13) which spontaneously lost carbon dioxide to yield ethyl 2,3-dihydro-1H-pyrrolizine-7-carboxylate (14).

SCHEME 3

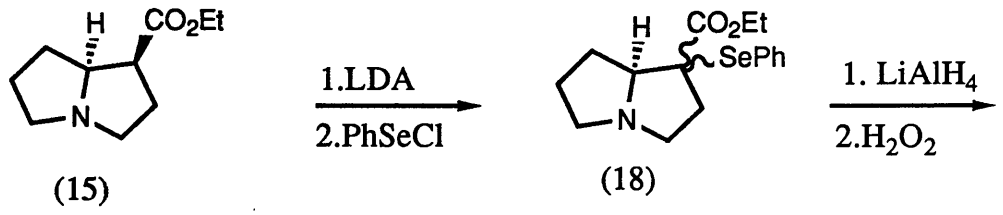

Acetic
Anhydride
COOH
N
CHO
(11)
CO2Et
O
N
(12)
O
N
O
CO2Et
(13)
- CO2
CO2Et
N
(14)
H2
H
CO2Et
N
(15)
H
CH2OH
N
(16)
H
CH2OH
N
(17)

Stereospecific cis-hydrogenation afforded ethyl endo-pyrrolizidine-1-carboxylate (15). This ester was then converted into (±)-isoretronecanol (16) by hydride reduction or into (±)-trachelanthamidine (17) by epimerisation, followed by reduction. A simple method for conversion of these bases into their 1,2-didehydro analogues was developed by Robins and Sakdarat.[24] Removal of the proton α to the ester of (15) gave an ester-stabilised enolate (Scheme 4). Reaction of this enolate with phenylselenenyl chloride afforded the intermediate (18). Reduction of the ester to the alcohol, followed by oxidation of the selenide to the corresponding selenoxide yielded (19). Thermal syn-elimination gave (±)-supinidine (20). This approach has been adopted in this project, aimed towards the synthesis of 3,3-dimethyl supinidine and is described in Chapter 4.

Tufariello and Tette[25] used similar methodology in their synthesis of supinidine (20) (Scheme 5). 1,3-Dipolar addition between 1-pyrroline 1-oxide and methyl 4-hydroxycrotonate afforded alcohol (21). The alcohol was converted into the mesylate and this was hydrogenated to give (22). Dehydration and reduction yielded (±)-supinidine (20). This procedure was extended by Tufariello and Lee[26] using the nitrone (24) to give (±)-retronecine (25).

Related to the 1,3-dipolar cycloaddition route is the synthesis of P.A.s by imidate methylide cycloaddition. This approach was developed by Vedejs and Martinez[27] who described the first viable route to nonstabilized iminium ylides, and 1,3-dipolar cycloadditions with these resulting in the synthesis of pyrrolines

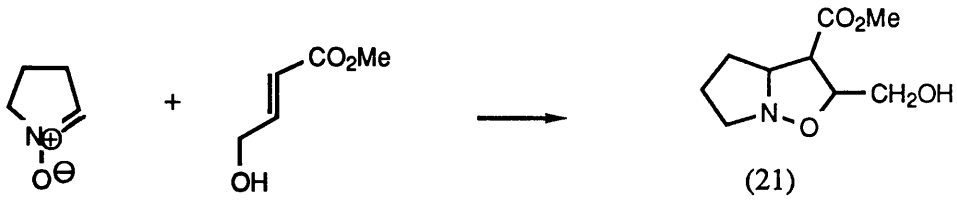
SCHEME 4



(15) (18)

(19) (20)

SCHEME 5



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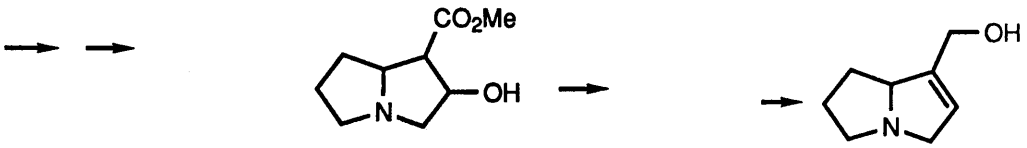


(22) (20)

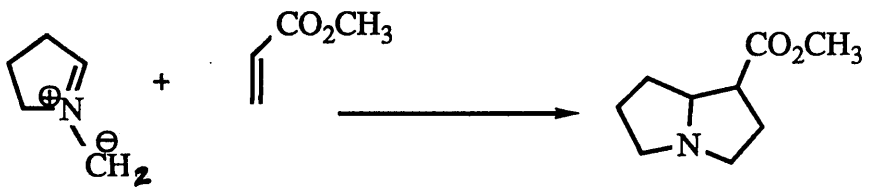
(24) (25)

and ultimately retronecine (25). In essence, the basis of this route is the cycloaddition as shown in Scheme 6 of an unsaturated ester with an imidate methylide. This latter species was previously unknown, but was prepared by the CsF desilylation of the corresponding (trimethylsilyl) methyl salt (30) (Scheme 7). This desilylation was carried out in the presence of methyl acrylate and the cycloadduct (33) was isolated in 51% yield. Conversion of (33) into retronecine was effected by catalytic hydrogenation to remove the unsaturation, followed by introduction of the 1,2-double bond following a method similar to that used by Robins and Sakdarat.[24] In this case, the selenenylating agent was diphenyl diselenide and meta-chloroperbenzoic acid was used as the oxidant. Retronecine was finally obtained by cleavage of the benzyl ether group using DIBAH.

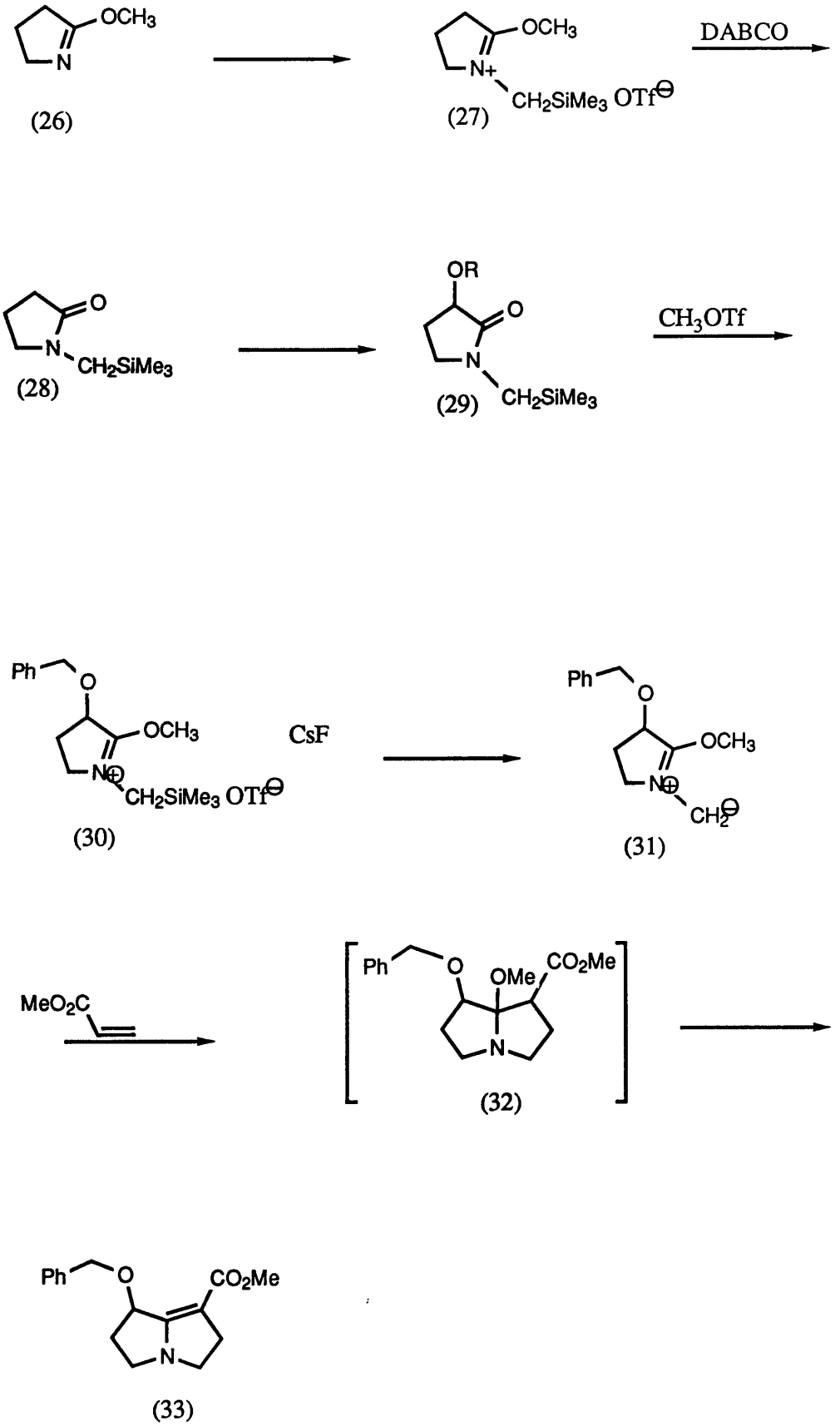
2.3 Intramolecular cyclisation of iminium ions

The utility of iminium salts in organic synthesis is well known.[28] Particular use has been made of the highly electrophilic carbon which presents a ready site for nucleophilic attack. However, the various methods employed to produce iminium salts have suffered from lack of regiospecificity and low yields.[29] Rapoport and co-workers developed a high-yielding, single-step, regiospecific method for preparing iminium salts. The starting materials were the readily accessible α-tertiary amino acids.[30] The experimental procedure for the production of these salts is relatively straightforward, involving the heating of the

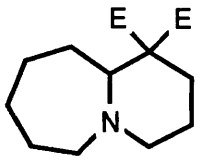
SCHEME 6



SCHEME 7

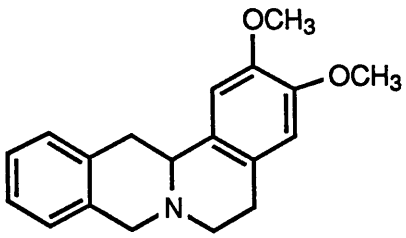

OCH3
N
(26)
OCH3
N+
CH2SiMe3 OTf⊖
(27)
DABCO
O
N
CH2SiMe3
(28)
OR
O
N
CH2SiMe3
(29)
CH3OTf
Ph
O
OCH3
N⊕
CH2SiMe3 OTf⊖
(30)
CsF
Ph
O
OCH3
N⊕
CH2⊖
(31)
MeO2C
Ph
O
OMe
CO2Me
N
(32)
Ph
O
CO2Me
N
(33)

α-tertiary amino acid with an excess of phosphorus oxychloride. The formation of the iminium ion was shown by examination of the infra-red spectrum, where the main absorption was between 1670 cm^{-1} and 1700 cm^{-1}. These iminium salts could be precipitated as crystalline perchlorates or reduced to the corresponding amines by catalytic (palladium on charcoal) hydrogenation. Rapoport used this generation of iminium ions to prepare a series of compounds, including diethyl 1-azabicyclo[5.4.0]undecane-8,8-dicarboxylate (34), 2,3-dimethoxy-5,8,13,13a-tetrahydro-6H-dibenzo[a,g]quinolizine hydrochloride (35), pyrrolizidines, quinolizidines, indolizidines and anatoxin a (36).[30] The synthesis of (34) is a good example of the general procedure (Scheme 8). Hexahydroazepine-2-carboxylic acid was converted into the corresponding benzyl ester (38) which was alkylated with diethyl 3-bromopropylmalonate,[31] to give (39). Hydrogenation removed the benzyl group, and the amino acid (40) formed was treated with phosphorus oxychloride to form the iminium salt. This was cyclised by adjusting the pH of the solution to 6.5 and allowing the mixture to stand overnight. This is the general approach also used in this project, aimed towards the synthesis of 8-methyl and 3,3-dimethyl substituted pyrrolizidines. This work is discussed in Chapter 4.



$E = CO_2Et$

(34)



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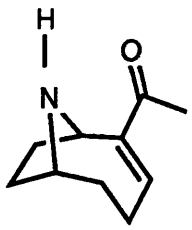


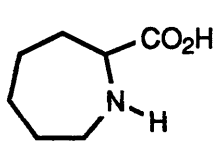
(36)

2.4 Synthesis of Pyrrolizidine Alkaloid Analogues

The major source of pyrrolizidine alkaloids is the extraction of plant material and purification of mixtures. This often leads to problems of scale due to the low concentrations of these alkaloids in plants and much effort has been directed towards the synthesis of analogues which would show similar properties in metabolic and toxicological studies. If we consider the structure of retronecine (25), then one monocylic analogue would be (41). The synthesis of this compound, Synthanecine A, was reported in 1974[32] and improved upon, also by Mattocks, in 1978[33] (Scheme 9). Methylamine was reacted with diethyl maleate (42) to give the amino-diester (43). N-Alkylation using ethyl bromoacetate gave the triester (44). Cyclisation to (45) was achieved by reaction with sodium hydride in a Dieckmann cyclisation. The introduction of unsaturation was accomplished by selectively reducing the ketone to the alcohol using sodium borohydride, followed by tosylation and elimination to afford (46). The ester groupings were then reduced to alcohols by reduction with DIBAH, yielding Synthanecine A (41). A range of similar compounds (47) - (50) was produced by varying the route (Synthanecines B - E).

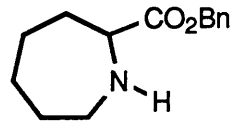
Synthanecine A (41) was chosen as the starting material for the preparation of a range of derivatives which included diesters, dicarbamates and the bisphenylcarbonate derivative, (51 - 56). Mattocks found that these derivatives were converted in vivo into pyrrole derivatives which were as hepatotoxic as naturally

SCHEME 8



(37)

Benzyl alcohol
H^+



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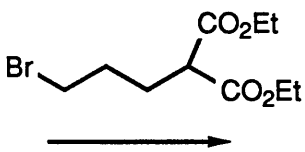
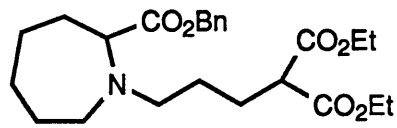
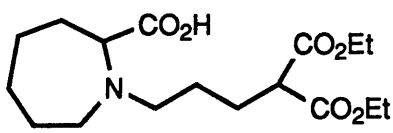




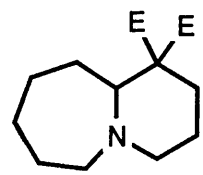
H_2

(39)



(40)

1. $POCl_3$
100^oC, 3min
2. H_2O
3. pH 6.5



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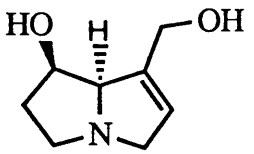

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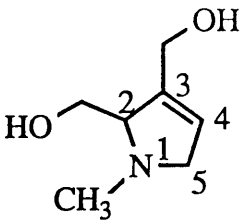
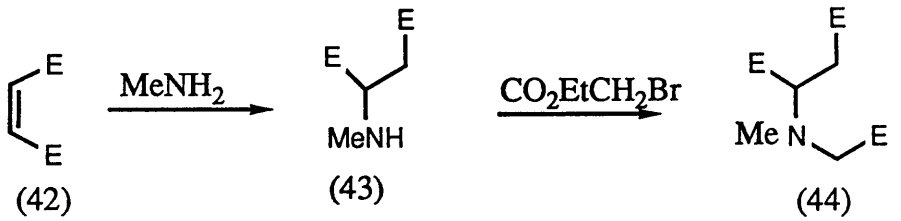
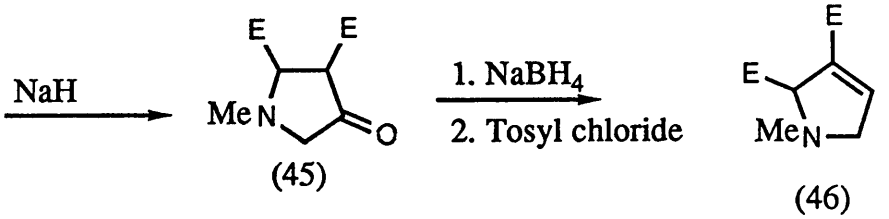


(41)

SCHEME 9

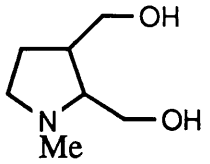


(42) (43) (44)



(45) (46)

(41)



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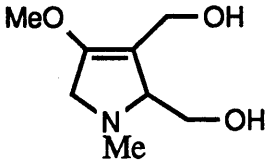

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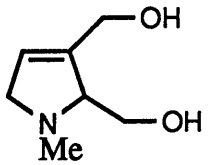

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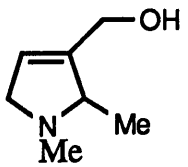
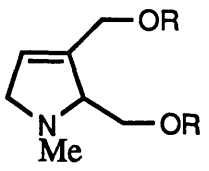


(50)



(51) R = COC(Me)(Et)OMe

(52) R = $PO(OEt)_2$

(53) R = CONHEt

(54) R = $CONMe_2$

(55) R = $CONEt_2$

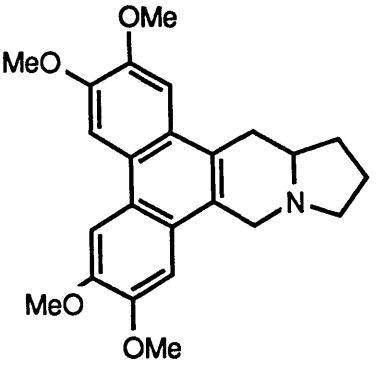
(56) R = CO_2Ph

occurring pyrrolizidine alkaloids, although simple diesters were less toxic than the others due to esterase hydrolysis.

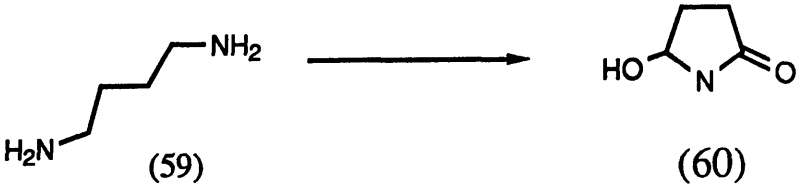
2.5.1 1-Pyrrolines in Organic Synthesis

1-Pyrroline (57) has proved of interest to chemists for many years, not only because of its role in the biosynthesis of alkaloids such as tylophorine (58)[34] and in the pathway from putrescine (59) to 5-hydroxy-2-pyrrolidone (60),[35] but also because of its potential use as a synthetic intermediate. Due to the electrophilic nature of the carbon atom of the imine, 1-pyrroline and its derivatives have been used in a wide range of syntheses. Much use of 1-pyrroline has been made by Herbert et al. in the synthesis of a series of precursors of the tylophorine (58) type of indolizidine alkaloids and in a synthesis of the alkaloid septicine (61).[34, 36] Central to these ideas was the synthesis of 2-phenacylpyrrolidines (62). The reasoning behind the use of this group of compounds as synthetic intermediates is the established biosynthetic pathway to tylophorine[37] from phenylalanine, tyrosine and ornithine (Scheme 10). The 2-phenacylpyrrolidines (62) were prepared by condensation of 1-pyrroline and an appropriately substituted benzoylacetic acid,[38] mimicking the biosynthetic pathway. Appropriate labelling with ^{14}C and ^{3}H confirmed that keto-amines such as (62) are important intermediates in the biosynthesis of phenanthroindolizidine alkaloids. The major advantage of using 1-pyrroline in this case is that it may be prepared either from

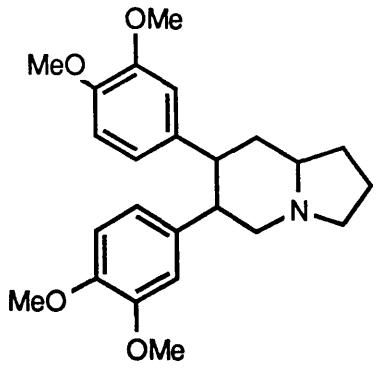
(57)



(58)



(59) (60)



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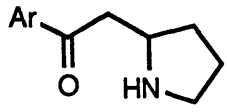
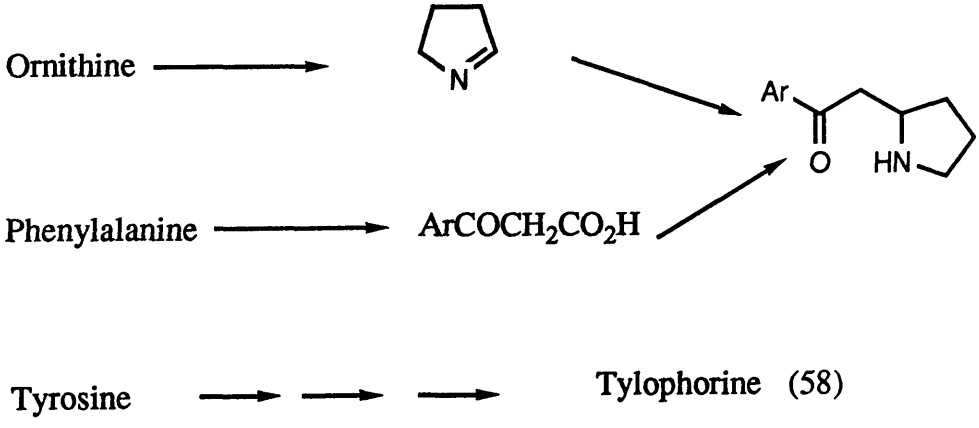


(62)

SCHEME 10

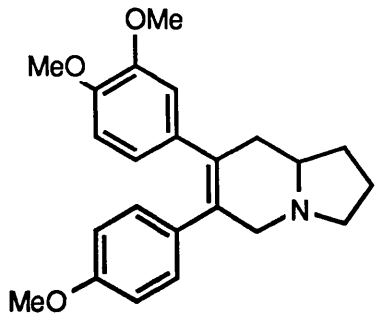


ornithine[39] or putrescine,[40] both of which are available with a variety of labels, and so the 5-membered ring in these alkaloids can be labelled with ^{14}C or ^{3}H. This methodology was further extended by Herbert in the synthesis of the alkaloids julandine (63) and cryptopleurine (64).[41] In these cases, instead of 1-pyrroline, 1-piperideine (65) was used. This was generated *in situ* from cadaverine by pea seedling diamine oxidase.[40]

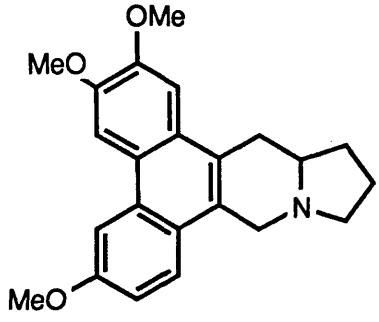
Substituted 1-pyrrolines have been used by Dannhardt and Obergrusberger in the synthesis of tetrahydroindolizidines.[42] The method employed makes use of the known reactivity of 1,4-dipolar molecules with alkynes to form 6-membered rings.[43] In this case (Scheme 11), acetylenedicarboxylic acid dimethyl ester (66) was reacted with a range of substituted 1-pyrrolines to produce the tetrahydroindolizidines (71) - (74). In these reactions 2 equivalents of the acetylenedicarboxylic acid dimethyl ester (66) were required and the reaction is thought to go through intermediates of the type (75) which then react with the alkyne to give (71) - (74).

2.5.2 Synthesis of 1-Pyrrolines

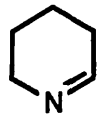
Although 1-pyrrolines are potentially very useful synthetic intermediates, their use has been limited by the relative instability of 1-pyrroline itself and by the lengthy and inconvenient routes used for its generation. Some of the earliest work in this area was carried out by Schöpf who developed several methods for the preparation of 1-pyrroline,[44] including the



(63)

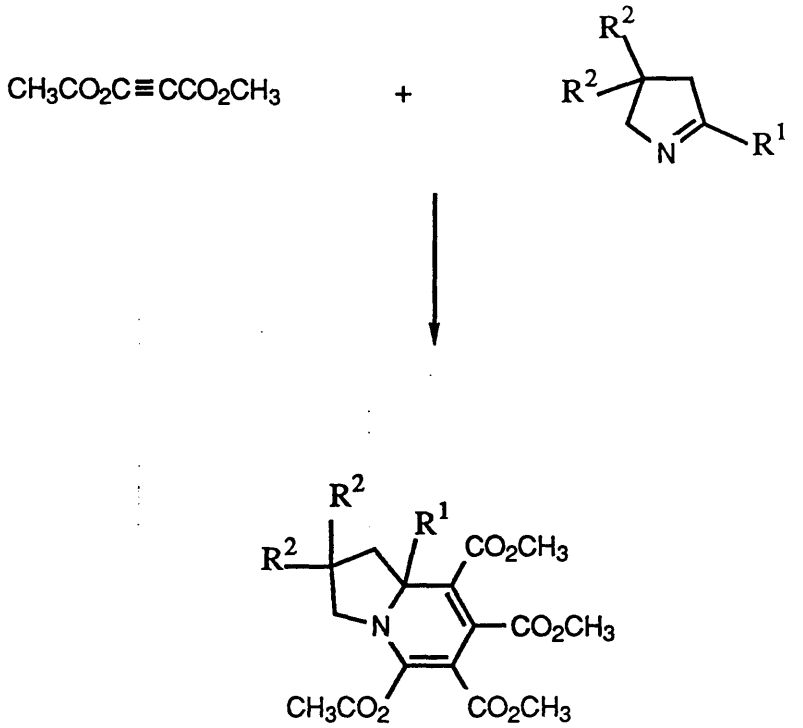


(64)



(65)

SCHEME 11

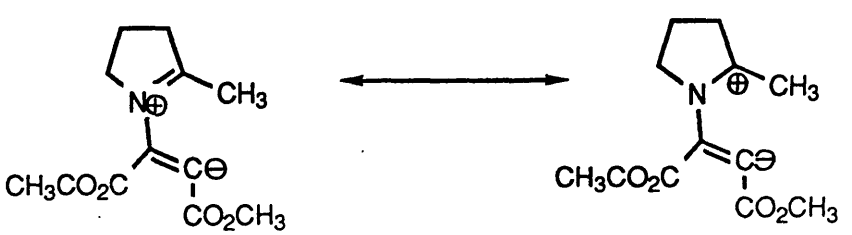


(67) , (71) $R^1 = CH_3$, $R^2 = H$

(68) , (72) $R^1 = CH_3$, $R^2 = CH_3$

(69) , (73) $R^1 = CH_2C_6H_5$, $R^2 = H$

(70) , (74) $R^1 = C_6H_5$, $R^2 = H$



(75)

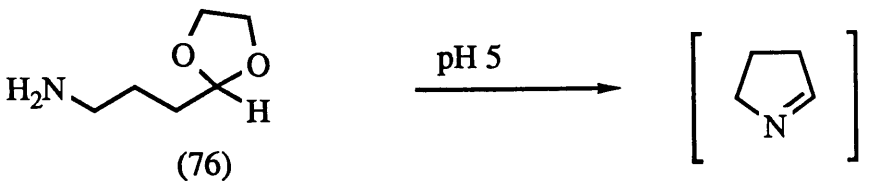
cyclisation of γ-aminobutanal acetal (76) which on hydrolysis at pH 2 gave 1-pyrroline in solution (Scheme 12). Schöpf also prepared 1-pyrroline from pyrrolidine (77) by N-chlorination, followed by dehydrohalogenation[44] (Scheme 13).

The 1-pyrroline was generally assayed by its reaction with ortho-aminobenzaldehyde. The use of 1-pyrroline in the study of biosynthetic pathways led to the extension of this work by Jakaby and Fredericks who synthesised 1-pyrroline from ornithine (79)[45] (Scheme 14) and Clarke and Mann who used putrescine (59) as starting material. In this case, pea seedling diamine oxidase was used to effect the oxidation.[46] One interesting aspect of this work lies in the relative instability of 1-pyrroline and its spontaneous trimerisation to the hexahydrotriazine derivative (80). Poisel[47] investigated this reaction and the ^1H n.m.r. spectra of the monomer and the trimer. The features of this will be discussed in Chapter 5. The monomer was found to be stable in methanolic solution; conversion into the trimer being achieved by heating with potassium hydroxide.

Several groups have reported the synthesis of 2-substituted pyrrolines.[48] Reaction of γ-chlorobutyronitrile (81) with various Grignard reagents led to the formation of 2-substituted pyrrolines of type (82).

Methods for the stabilisation of 1-pyrroline and its use in the synthesis of alkaloids and pyrrolidine derivatives are discussed in Chapter 5.

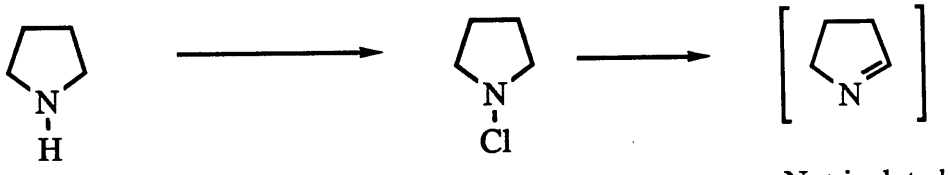
SCHEME 12



(76)

Not isolated

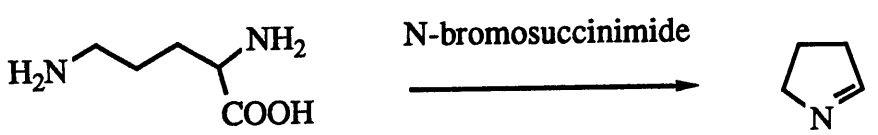
SCHEME 13



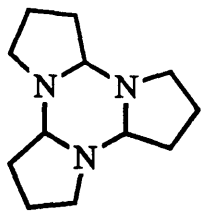
(77) (78)

Not isolated

SCHEME 14

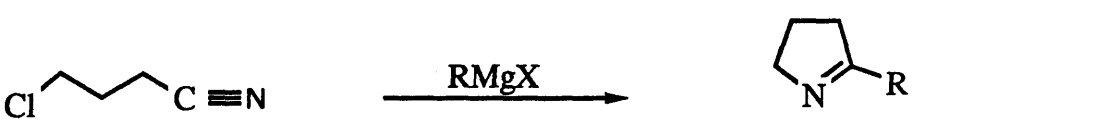


(79)



(80)

SCHEME 15



(81)

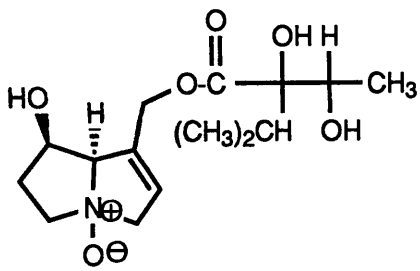
(82) R = Alkyl , aryl

CHAPTER 3

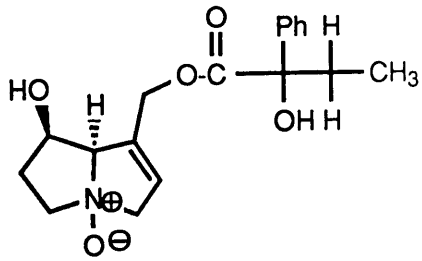
SYNTHESIS OF DIESTERS AND DIESTER N-OXIDES OF SYNTHANECINE A

3.1 Introduction

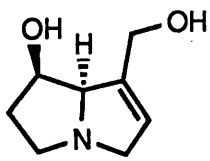
As has been previously discussed (Chapter 1), pyrrolizidine alkaloids display a wide range of biological activities.[20] This has led to the study of pyrrolizidine alkaloids and analogues as potential drugs where it is hoped that the therapeutic benefits of use against, for example, tumours, outweigh any toxic side-effects. Recently, indicine N-oxide (6) has been selected for clinical trials since it is less toxic than most pyrrolizidine alkaloids.[17] In the same area, the related unnatural ester (7) has been shown to be more active as an anti-cancer drug than (6).[18] The mechanism of this anti-tumour activity is not known, although the esterifying acid is important as can be shown by the differing anti-tumour activities of (6) and (7). Work in this field has been limited by the quantities of compounds available for testing as the necine base (±)-retronecine (25) is not readily available, either by synthesis, or from natural sources. Some seeds and plants (chiefly in India, South Africa and parts of the U.S.A.) do produce considerable quantities of P.A.s containing retronecine, but supplies of these seeds and plants can be difficult to obtain. For toxicological and metabolic studies, reasonable amounts of the chosen alkaloid or analogue are required



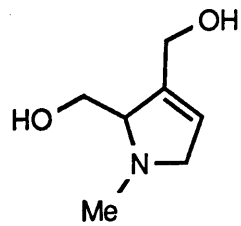
(6)



(7)



(25)



(41)

and thus a more accessible necine or necine analogue is needed. Synthanecine A (41), a monocyclic analogue of retronecine, was selected because it is readily available via a short and efficient route and its diester derivatives exhibit similar biological properties to retronecine diesters.[32, 33]

3.2 Synthesis of Synthanecine A

Synthanecine A was prepared by a modified version[49] of the route published by Mattocks[32, 33] (Scheme 9) as described in Chapter 2. At the final stage, selective reduction of the unsaturated diester (46) using DIBAH in toluene gave a mixture (88%) of Synthanecine A (41) and the corresponding pyrrole (83), 2,3-bishydroxymethyl-1-methylpyrrole in a ratio of approximately 85:15 (using ^1H n.m.r. spectroscopy). Pure Synthanecine A was obtained by column chromatography on silica gel, eluting with chloroform-methanol-triethylamine (85:14:1), and it was stored under argon at 0°C to prevent oxidation to the pyrrole (83). Alternatively, the crude mixture of Synthanecine A (41) and pyrrole (83) was used in further reactions and the products were then purified. Pure Synthanecine A (41) was characterised by ^1H n.m.r., i.r., and mass spectroscopy and as the picrolonate derivative, m.p. 174-175°C (literature value 176°C).[32]

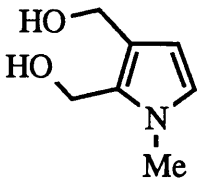
3.3 Synthesis of Synthanecine A derivatives

The range of derivatives chosen for synthesis was determined by two factors:

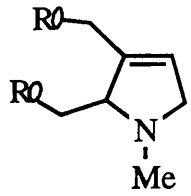
1. Similarity to compounds with known biological activity;
2. Ease of synthesis.

As has been stated previously, pyrrolizidine alkaloids exist naturally mostly as diesters, the esterifying acids having various levels of complexity. The examples which have shown most promise as anti-tumour agents have been N-oxides.[16, 17, 18, 50] This points to the preparation and evaluation of a range of bis-esters and similar compounds and the corresponding N-oxides. Mattocks[32] has synthesised the derivatives (51) - (56). Compounds (53) and (56) were used in this study, together with the novel compounds (84), (85), and (86).[33] Although Mattocks produced the bis(diethyl phosphate) (52), synthesis of the diphenyl phosphate using diphenyl phosphonyl chloride was unsuccessful here.

The simple esters such as the diacetate and dibenzoate were purified by column-chromatography on silica using petroleum ether (40°C - 60°C)-triethylamine (4:1) as solvent. An examination of the ^{1}H n.m.r. spectra of Synthanecine A and its dibenzoate derivative (85) showed the effect of esterification:



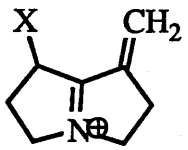
(83)



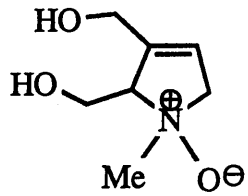
(84) R = COOEt

(85) R = COPh

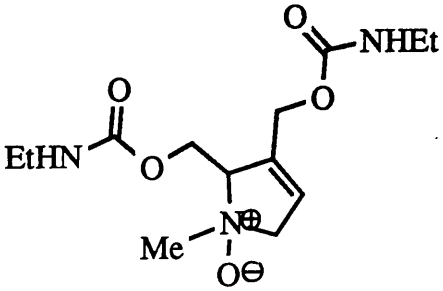
(86) R = COMe



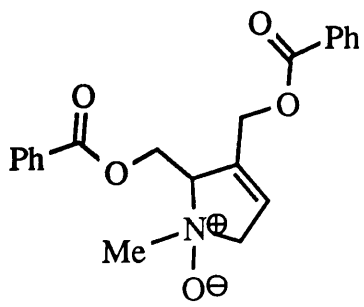
(87)



(88)



(89)



(90)

TABLE 1

^1H	Synthanecine A (Chemical shift p.p.m)	dibenzoate (Chemical shift (p.p.m)
N-$\underline{CH_3}$	2.50	2.59
2-H	3.53	3.18-3.58
4-H	5.73	5.92
5-H	3.25,3.88	3.80
6-H	3.67	4.45
7-H	4.16	4.96

All protons of the dibenzoate showed a downfield chemical shift which is most noticeable at 6-H and 7-H which lie closest to the ester functions. Work done by Robins and Barbour[49] using ^1H n.m.r. spectroscopic evidence has shown that esterification occurs in the first instance at the primary hydroxyl group of Synthanecine A rather than the less nucleophilic primary allylic hydroxyl. The diastereotopic protons at C-6 and C-7 of the diacetate (86) give rise to broad signals due to their near magnetic equivalence. However, when macrocyclic diesters are formed these signals change due to magnetic non-equivalence to an AB system (C-7 protons) and an AB part of an ABX system (C-6 protons).[49]

In Chapter 1, it was shown that one possible route to hepatotoxic molecules starting from pyrrolizidine alkaloids is *via* the alkylating agent (87). The nitrogen lone-pair plays a crucial role in the formation of this species and this, together with the known anti-tumour activity of indicine *N*-oxide (6) and the unnatural ester (7), indicates that *N*-oxides of these simple diesters would be suitable targets.

The two simplest methods of producing *N*-oxides are using a per-acid such as *meta*-chloroperbenzoic acid, or hydrogen peroxide as oxidising agent.[51] Due to the presence of a double bond in the starting materials which could be prone to some epoxidation, it was decided to use hydrogen peroxide as the oxidising agent. Destruction of excess reagent is also straightforward by the addition of manganese dioxide, as opposed to the use of gaseous ammonia after using a per-acid.

Synthanecine A *N*-oxide (88) itself has been prepared by Mattocks using hydrogen peroxide.[32, 33] However, it is a brown, viscous gum which rapidly decomposes to polymeric residues on standing. The *N*-oxides (89) and (90) of bases (53) and (85) were synthesised but proved difficult to characterise by conventional methods. Signals in the ^1H n.m.r. spectrum were very broad, possibly due to the presence of diastereoisomeric *N*-oxides. In fact, the only assignable peak in the spectrum of (89) was at δ= 3.20 p.p.m which was a singlet due to the methyl group on the positively charged nitrogen. This agrees with the normally

observed signal due to similar methyl groups of δ = 3.3 p.p.m[52] and the disappearance of the signal at δ = 2.5 p.p.m. due to the N-methyl protons of Synthanecine A. The product was shown to be homogeneous by t.l.c. using two different solvent systems and a range of stationary phases:

TABLE 2

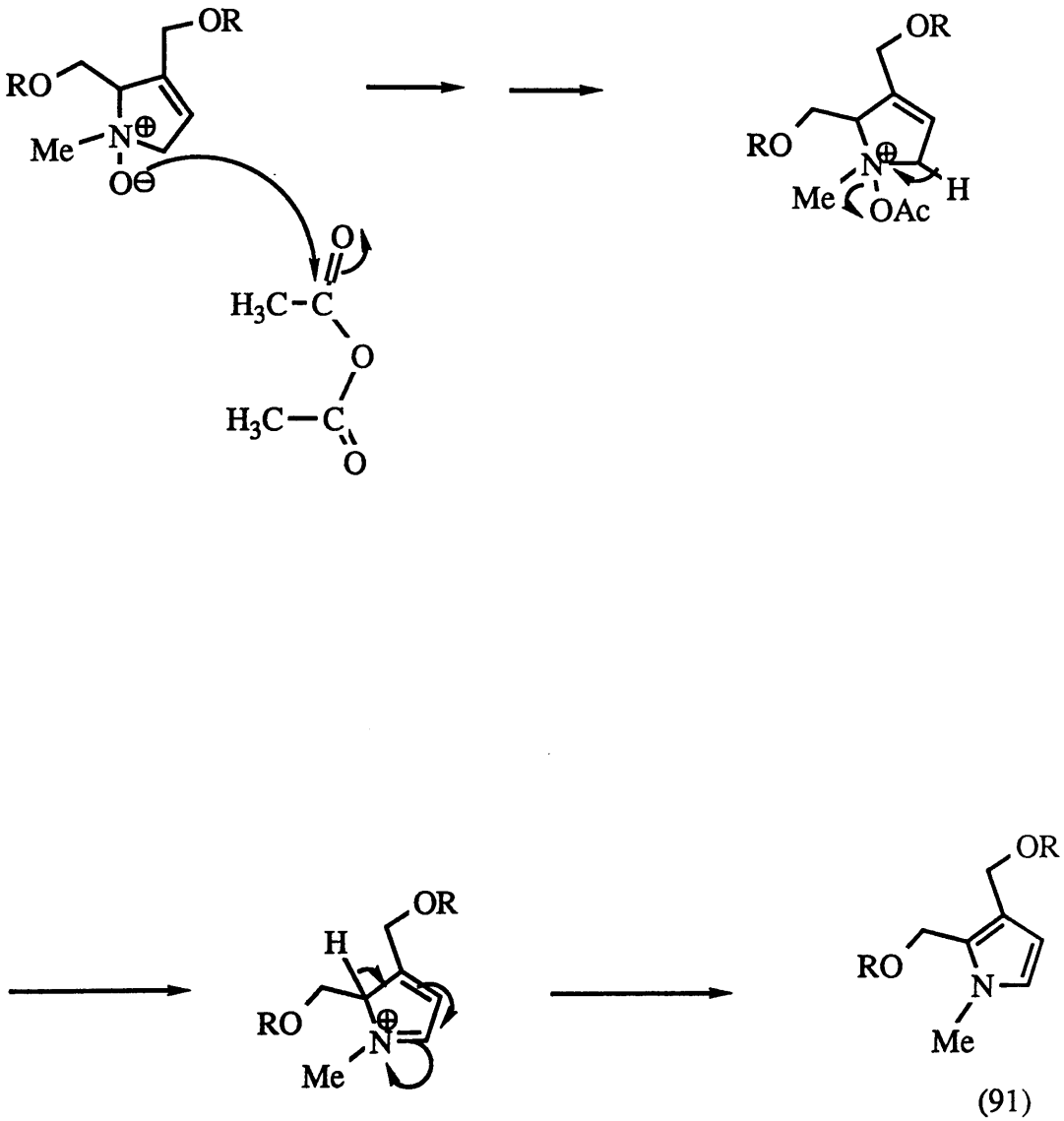
Stationary phase	solvent system	Rf
silica	1	0.61
silica	2	0.67
alumina	1	0.34
cellulose	1	0.82

1 = butanol-acetic acid-water (3:1:1).

2 = isopropanol-conc. ammonia (2:1).

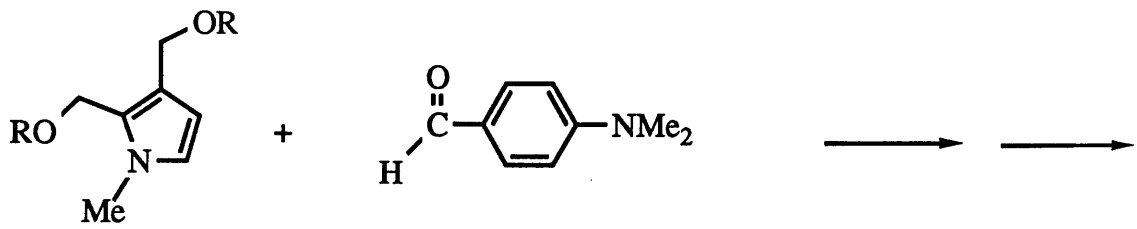
In all systems the product gave a single spot. The available evidence (^{1}H n.m.r. spectrum and t.l.c. data) indicates that only one isomer of (89) was produced in the oxidation. The fact that such polar solvent systems were needed also indicates N-oxide formation. Conclusive evidence was obtained by using a test developed by Mattocks.[53] The N-oxide was heated with acetic anhydride and a Polonovsky reaction took place (Scheme 16). This pyrrolic product (91) was then reacted with Ehrlich's reagent (4-dimethylaminobenzaldehyde) to give the highly conjugated

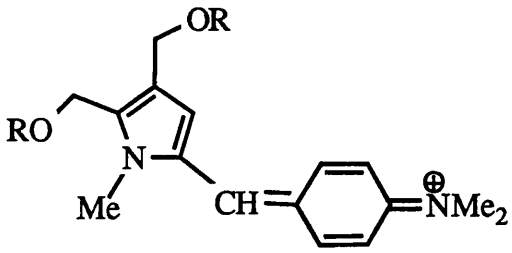
SCHEME 16



(91)

SCHEME 17





(92)

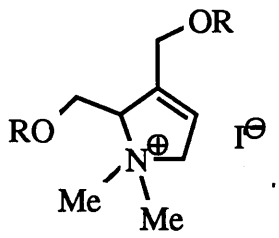
compound (92) (Scheme 17). The compounds produced by this procedure have characteristic U.V. absorptions with λ max. at around 565 nm. This method was adopted by D'Silva and Notari to estimate indicine N-oxide (6) in silica gel scraped from t.l.c. plates.[1] Both N-oxides (89) and (90) gave a positive reaction to this test.

Lack of stability of these N-oxides meant that they could not be used for anti-tumour testing. Other workers in this area sealed P.A. N-oxides under vacuum and sent them for testing.[50] One alternative possibility of producing a quaternary nitrogen compound is by forming the N-methyl ammonium iodide (93). Unfortunately, several attempts at this using methyl iodide proved unsuccessful.

3.4 Synthesis of a Supinidine Analogue (94)

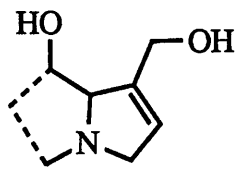
Synthanecine A (41) can be seen as a monocyclic analogue of retronecine (25). In the same way (94) can be seen as a monocyclic analogue of supinidine (4), another common necine base. Derivatives of (94) could be used in anti-tumour studies if this compound was available in sufficient quantity. Its similarity to Synthanecine A indicated a similar approach for the synthesis and this did indeed prove successful (Scheme 18).

Addition of methylamine (95) to acrylonitrile (96) gave β-cyanoethylmethylamine (97). Elemental analysis of the picrolonate of this compound gave the molecular formula $C_4H_8N_2$, confirmed by the appearance of the molecular ion at M/Z 84 in the mass spectrum. The infra-red spectrum showed NH at

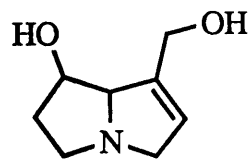


(93)

R = COMe



(41)



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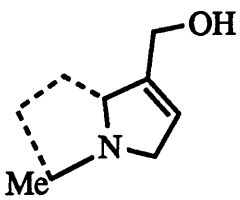
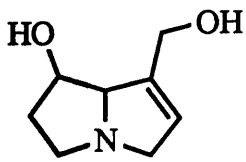
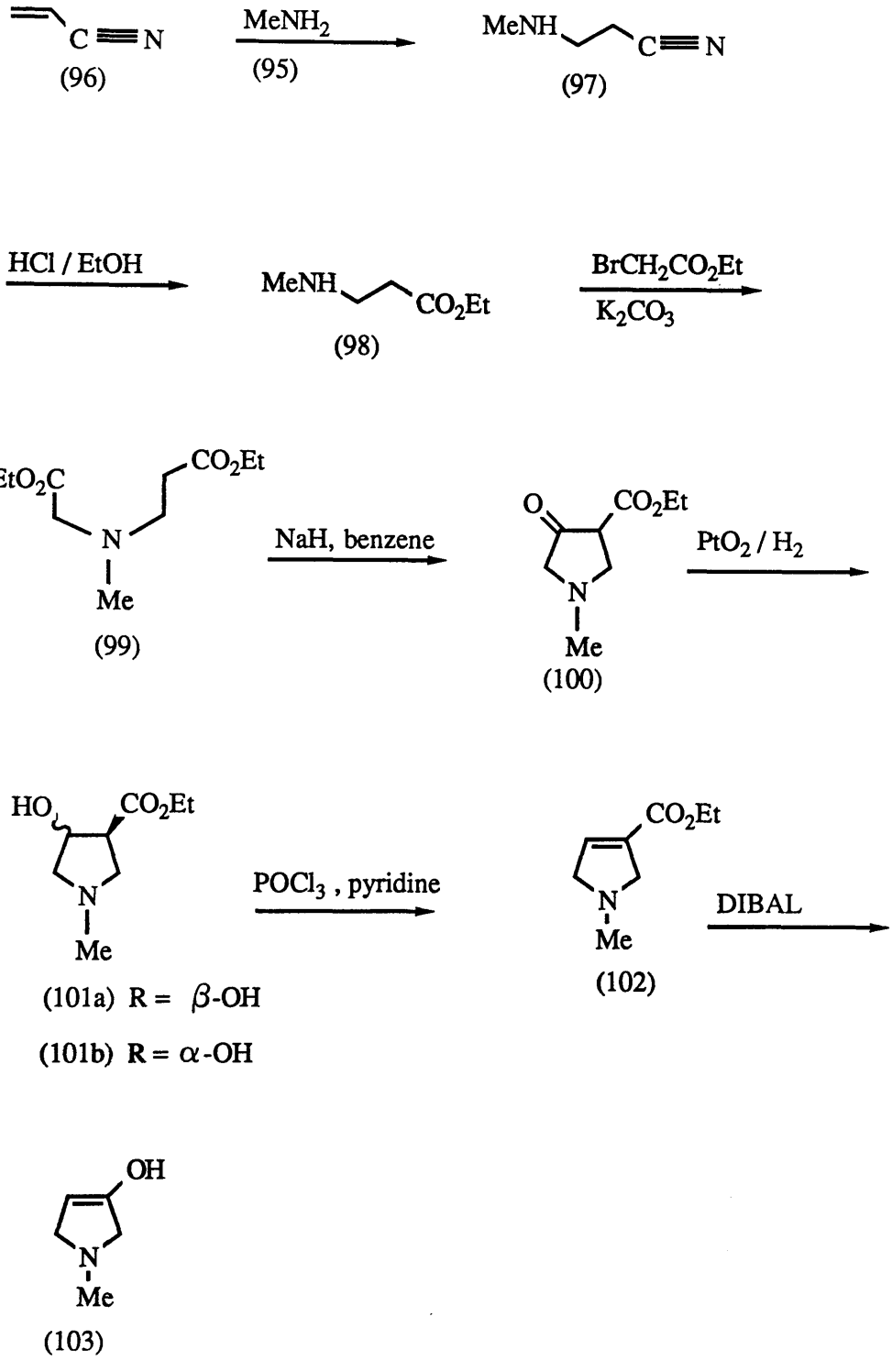


(94)



(4)

SCHEME 18


MeNH2
(95)
(96)
(97)
HCl / EtOH
(98)
BrCH2CO2Et
K2CO3
EtO2C
CO2Et
N
Me
(99)
NaH, benzene
O
(100)
PtO2 / H2
HO
POCl3 , pyridine
(101a) R = β-OH
(101b) R = α-OH
(102)
DIBAL
OH
(103)

3300cm^{-1} and the nitrile group at 2245 cm^{-1}. The ^{1}H n.m.r. spectrum revealed N-H at 1.30 p.p.m with the N-methyl signal at 2.46 p.p.m. The nitrile group of this compound was converted into the ethoxycarbonyl group by passing hydrogen chloride gas through a solution of (97) in ethanol. The yield of (98) was 85%. The picrolonate of this compound gave an analysis which showed (98) as $C_6H_{13}NO_2$ which was confirmed by a molecular ion at M/Z 131 in the mass spectrum. The i.r. spectrum showed the N-H grouping at 3320 cm^{-1}, but the nitrile at 2245 cm^{-1} has been replaced with the carbonyl absorption of the ester at 1730 cm^{-1}, with the C-O stretch at 1370 cm^{-1}. The ^{1}H n.m.r. spectrum showed N-H at 1.27 p.p.m. with the singlet due to N-methyl at 2.40 p.p.m. The methyl and methylene groups of the ethyl ester were present at 1.22 and 4.14 p.p.m. respectively. The diester (99) was produced by N-alkylation of (98) using ethyl bromoacetate in aqueous acetone containing potassium carbonate (yield 55%). Mass spectroscopy showed a molecular ion at 217 and analysis of the picrate of (99) suggested the formula $C_{10}H_{19}NO_4$. The absorbance due to N-H had disappeared from the i.r. spectrum which showed the carbonyl stretch of the two ester groups at 1730 cm^{-1} with the C-O stretch at 1170 cm^{-1}. The ^{1}H n.m.r. spectrum revealed two distinct groups of peaks attributable to two ester groups with two methyl signals at 1.22 and 1.24 p.p.m. and the corresponding methylene signals at 4.14 and 4.18 p.p.m. Dieckmann cyclisation of diester

(99) to the pyrrolidone (100) was accomplished using sodium hydride[33] in benzene. Accurate mass spectroscopy data on this product indicated a molecular formula of $C_8H_{13}NO_3$. The i.r. spectrum showed the presence of two distinct carbonyl absorbtions: the ester carbonyl at 1725 cm^{-1} and the ketone at 1675 cm^{-1}. The ^1H n.m.r. spectrum showed a single ethyl ester group with methylene and methyl signals at 4.22 and 1.20 p.p.m. respectively.

The next stage in the synthesis appeared relatively straightforward as it had been in the Synthanecine A route. Nevertheless, several attempts to reduce the pyrrolidone (100) to the hydroxypyrrolidine (101)[33] using sodium borohydride proved unsuccessful, producing only starting material. It proved necessary to change to catalytic hydrogenation using a platinum oxide catalyst in ethanol and aqueous hydrochloric acid. The acid was added to promote the formation of the enol which is thought to be the species hydrogenated. Due to the complexity of the ^1H n.m.r. spectrum with many overlapping signals, little could be ascertained about the relative stereochemistry at C-3 and C-4 of the hydroxyester (101). Presumably though, the catalytic hydrogenation occurs in a syn-fashion by addition of H_2 to the less hindered face of the pyrrolidone to give (101a). The hydroxyl proton was at 4.50 p.p.m. The accurate mass spectrum of (101) suggested the formula $C_8H_{15}NO_3$. The i.r. spectrum showed the presence of the hydroxyl group at 3400 cm^{-1} which had

replaced the ketone absorption from the pyrollidone. The i.r. spectrum also showed the ethyl ester remained, with carbonyl absorption at 1725 cm^{-1} and C-O stretch at 1170 cm^{-1}.

Dehydration of the hydroxyester (101) to (102) using p-toluenesulphonyl chloride was unsuccessful, yielding only starting material, despite long reaction times (1-2 days) and/or elevated temperatures (100°C). The unsaturated ester (102) was finally synthesised using phosphorus oxychloride at 130°C to effect the dehydration in 70% yield. It is surprising that such drastic conditions are required to dehydrate (101). This behaviour is more consistent with a trans-arrangement of the hydroxy and ester groups (101b). The formula $C_8H_{13}NO_2$ was suggested by the accurate mass spectrum. The i.r. spectrum indicated the formation of the double bond which had absorption at 1640 cm^{-1}. The two characteristic absorptions of the ethyl ester group were present at 1730 cm^{-1} and 1200 cm^{-1}. The ^1H n.m.r. spectrum showed the usual ethyl ester signals at 1.21 and 4.27 p.p.m. One interesting feature of this spectrum is the absorption due to the methylene group H-2 which contained a large geminal coupling constant of 15Hz. Similarly, H-4 and H-5 form an ABX system with J_{vic} 17Hz. H-4, the olefinic proton at 6.7 p.p.m., exhibited an absorption as a broad singlet, despite being coupled to H-5. This is characteristic of this type of system.

The final reduction step to the alcohol (103) was performed using DIBAH in toluene. Purification took place on an alumina

column eluting with methanol-ethyl acetate (2:3); yield 71%. Accurate mass data gave the molecular formula $C_6H_{11}NO$. The i.r. spectrum showed an OH band at 3410 cm^{-1} and a double bond at 1630 cm^{-1}. The ^1H n.m.r. spectrum contained the N-methyl singlet at 2.49 p.p.m., an olefinic proton at 5.81 p.p.m, a methylene group at 4.21 p.p.m. ($C\underline{H}_2OH$) and again, the 2-H absorptions at 3.42 and 3.94 p.p.m. with J_{gem} 18Hz. The 4-H, 5-Hz ABX system could again be seen with 5-H absorptions at 3.15 and 3.84 p.p.m., J_{gem} 16 Hz.

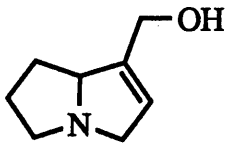
Lack of time did not permit a range of esters to be produced. However, the overall yield for this route is 4% from readily available and cheap starting materials (methylamine and acrylonitrile) and we have thus demonstrated a simple route to an interesting new pyrrolizidine alkaloid analogue.

CHAPTER 4

SYNTHESIS OF SUPINIDINE DERIVATIVES

4.1 Introduction

In Chapter 3 the work carried out in this project aimed towards the synthesis of potential anti-tumour agents was described, using the synthetic P.A. analogue synthanecine A. In order to prevent formation of an alkylating species as discussed in Chapter 1, the nitrogen lone-pair in the derivatives was involved in N-oxidation. An alternative approach can be seen if we examine the structure of supinidine (4). In order for the pyrrole derivative to form, the proton at C-8 must be removed, and also one of the protons at C-3. Obviously, if these protons were not available the pyrrole could not form. This suggests targets where C-3 and/or C-8 are substituted in some way. We chose the targets 3,3-dimethylsupinidine (104) and 8-methylsupinidine (105). The reason for basing the analogue work on supinidine (4) was that an efficient synthesis of (4) had already been accomplished by Robins and Sakdarat[55] and it was felt that this route could be adapted. Some of the possible intermediates en route to (104) had also been synthesised.[30]



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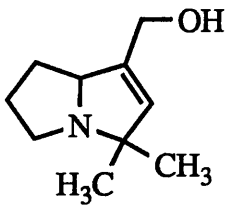
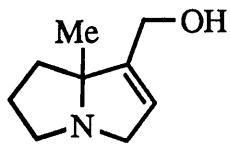


(104)

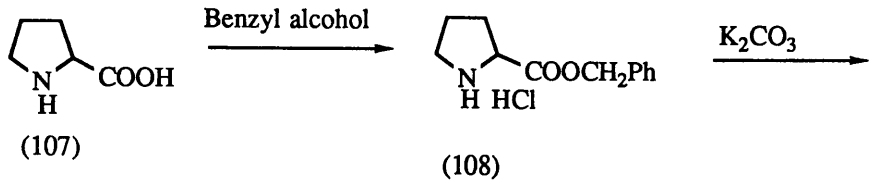


(105)

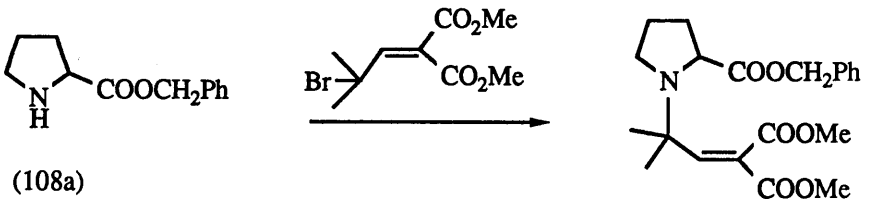
4.2 Synthesis of 3,3-dimethylsupinidine (104)

The methodology chosen for the synthesis of (104) was that of Padgett and his co-workers, as outlined in Chapters 1 and 2, involving the cyclisation of iminium ions. The key intermediate in our synthesis was dimethyl 3,3-dimethylpyrrolizidine-1,1-dicarboxylate (106) which had been prepared previously.[30] The route to this compound is outlined in Scheme 19 and takes a similar course to that described in Scheme 8. The carboxylic acid function of proline (107) was protected as its benzyl ester and isolated as the hydrochloride (108). The free base was easily obtained by extraction of a solution of (108) in 10% aqueous potassium carbonate with ether. N-Alkylation using 4-bromoisobutylidenemalonate afforded (109). The i.r. spectrum showed the ester carbonyls at 1725 cm^{-1} and the olefin at 1665 cm^{-1}. The key features of the ^1H n.m.r. spectrum were the two methyl singlets at 1.2 p.p.m., the methylene group of the benzyl ester at 5.0 p.p.m. and the olefinic proton at 6.7 p.p.m. The reason for the use of this unsaturated side-chain is that cyclisation attempts using the saturated equivalent failed due to intramolecular cyclopropane formation, as the carbanion formed α to the ester groups displaced the bromine. Generation of the required α-tertiary amino-acid (110) was accomplished by hydrogenation of (109) which also reduced the double bond. The infra-red spectrum of (110) showed clearly the OH of the carboxylic acid function with the carbonyl absorption at 1640 cm^{-1}. The ethyl ester carbonyl stretch was at 1720 cm^{-1}.

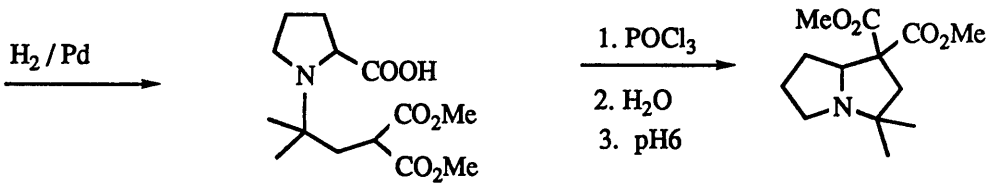
SCHEME 19



(107) (108)



(108a) (109)

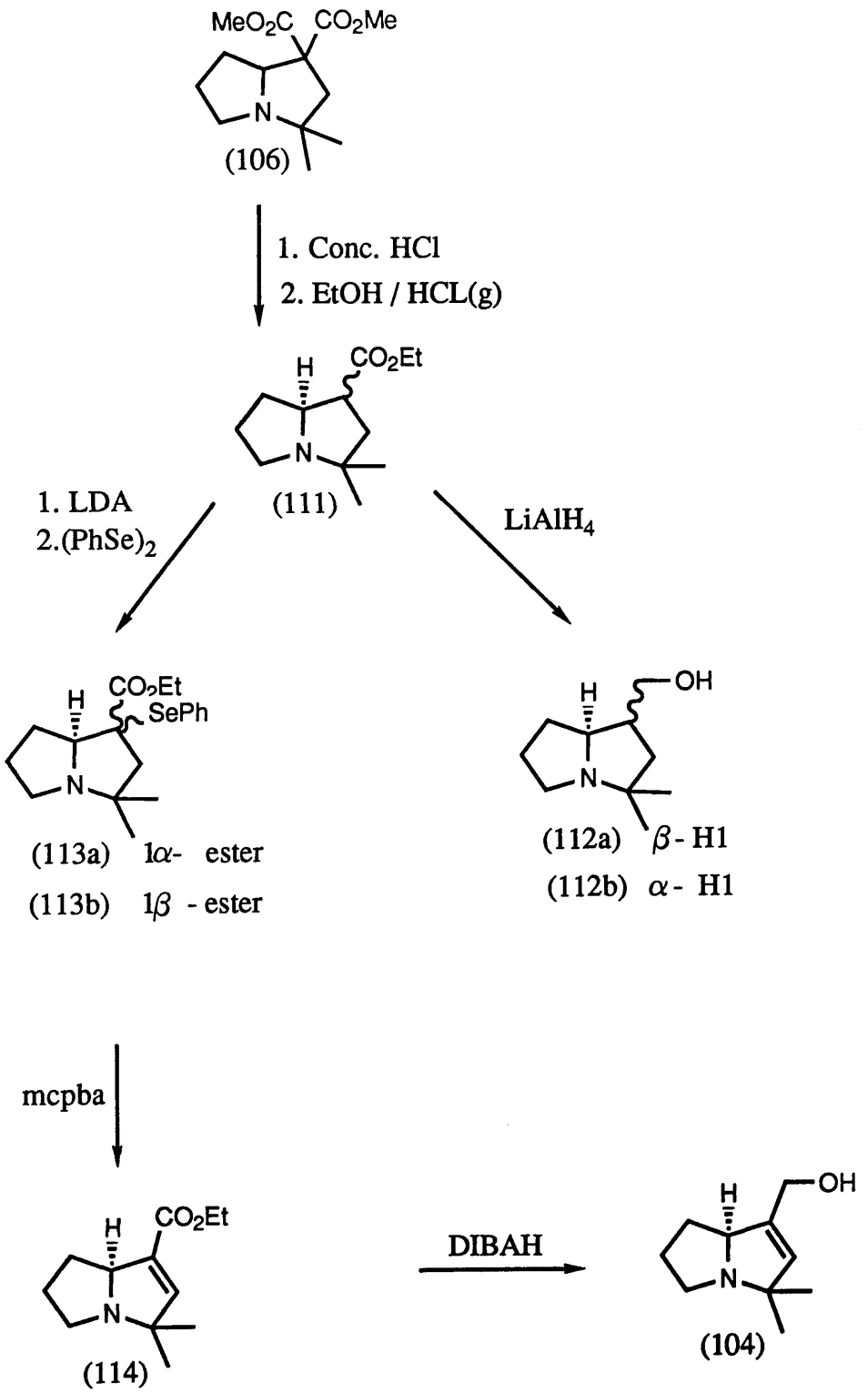


(110) (106)

The proton n.m.r. spectrum revealed the removal of the aromatic and the olefinic protons, with a new signal at 5.90 p.p.m. due to the proton alpha to the two ester groups in a "malonate" type position. Cyclisation to (106) via the iminium ion was performed in the standard manner using phosphorus oxychloride. We carried out this synthesis with some minor variations which are discussed later in this Chapter.

Having obtained (106) by a known route, the way to 3,3-dimethylsupinidine (104) appeared relatively straightforward. However, attempted demethoxycarbonylation using sodium and lithium chlorides in a variety of solvents proved unsuccessful, leading to a complex mixture of unidentified products. We eventually chose hydrolysis and decarboxylation using conc. hydrochloric acid, followed by reesterification with hydrochloric acid gas in ethanol (Scheme 20). This gave monoester (111) in 82% yield. Accurate mass data gave the molecular formula $C_{12}H_{21}NO_2$. The i.r. spectrum exhibited an ester carbonyl at 1730 cm^{-1}. In the ^1H n.m.r. spectrum two methyl singlets were visible at 1.18 and 1.20 p.p.m. These are the two ring geminal methyl groups which are magnetically non-equivalent. The methylene and methyl signals of the ethyl ester were clearly visible at 4.15 and 1.23 p.p.m. respectively. All other ring protons were found in the range 1.5 - 4 p.p.m. A ^{13}C n.m.r. spectrum of this monoester exhibited only one set of peaks with a distinct single carbonyl carbon signal at 176.6 p.p.m. This illustrates that only one of the two possible

SCHEME 20


MeO2C CO2Me
N
(106)
1. Conc. HCl
2. EtOH / HCL(g)
H
CO2Et
N
(111)
1. LDA
2.(PhSe)2
LiAlH4
H
CO2Et
SePh
N
(113a) 1α- ester
(113b) 1β - ester
H
OH
N
(112a) β - H1
(112b) α - H1
mcpba
H
CO2Et
N
(114)
DIBAH
H
OH
N
(104)

diastereoisomers of (111) has been formed. The stereochemistry of ester (111) was not clear from ^{1}H n.m.r. spectral data, but the ^{13}C n.m.r. spectrum showed no doubling of signals. It would be expected during the hydrolysis of racemic (106) that the more stable 1α-exo-isomer would be formed which would give the 1α-ester (111a) on reesterification. The epimerisation of endo-ester and carboxylic acid functions at C-1 under acidic or basic conditions is a well-known process[56] and has been used in the synthesis of some optically active pyrrolizidine bases by Robins and Sakdarat.[55] The ester (111) was reduced with lithium aluminium hydride to give the alcohol (112) (45%). Using the reasoning previously applied to the stereochemistry of ester (111), this alcohol (112a) would thus be (±)-3,3-dimethyltrachelanthamidine. T.l.c. data showed a lower R_F value in the standard solvent system (0.15 cf. 0.50 of the ester) which is characteristic of amino-alcohols. The i.r. spectrum confirmed the presence of a hydroxyl group with absorption at 3610 cm^{-1}. Accurate mass data gave the molecular formula $C_{10}H_{19}NO$. The ^{1}H n.m.r. spectrum showed the two methyl singlets due to the two methyl groups on C-3. The other main feature of this spectrum was the two proton doublet at 3.68 p.p.m. which is due to the methylene of the hydroxymethyl group. The ^{13}C n.m.r. spectrum revealed five methylene groups, two methine groups, two methyl groups and one quaternary carbon, all of which are consistent with the proposed structure.

Thermal elimination of a phenylseleno-group[27, 55, 57] was selected for the introduction of the olefinic double-bond into ester (111). Although phenylselenenyl chloride had been used with success in similar cases, it was decided to use diphenyl diselenide in this instance due to its lower toxicity and odour relative to phenylselenenyl chloride, and its relative ease of use.[27] Generation of the enolate of (111) using L.D.A. at -78°C in THF with 5% HMPA added, followed by addition of diphenyl diselenide and warming to 0°C gave the selenide (113). Accurate mass data gave the molecular formula $C_{18}H_{25}NO_2Se$. The i.r. spectrum showed the ester carbonyl stretch absorption at 1710 cm^{-1} with the two bands due to C-O absorptions at 1290 and 1070 cm^{-1}. The C-Se absorption was seen at 1600 cm^{-1}. The ^1H n.m.r. spectrum revealed the two ring methyl groups as singlets at 1.20 and 1.41 p.p.m. respectively. The ethyl ester was seen as a methyl triplet at 1.21 p.p.m. and a methylene quartet at 4.15 p.p.m. The presence of an aromatic substituent was obvious from the three proton peak at 7.40 p.p.m. and the two proton peak at 7.65 p.p.m. Removal of HMPA from solution was performed by thorough washing of the organic phase with brine and aqueous lithium chloride; the HMPA complexed readily with Li^+. The stereochemistry of this intermediate selenide (113) is uncertain. Addition of the diphenyl diselenide to the more-hindered endo-face of the intermediate lithium enolate would produce the 1α-ester (113a) whereas addition to the less-hindered exo-face would give the 1β-ester (113b).

The fragmentation of the selenoxide took place after oxidation of the selenide (113) using mcpba. Data from accurate mass measurement gave the molecular formula for (114) as $C_{12}H_{19}NO_2$. The i.r. spectrum exhibited ester carbonyl absorption at 1705 cm^{-1} and the double-bond at 1635 cm^{-1}. The ^1H n.m.r. spectrum revealed the effect of the introduction of a double bond into the ring: the 3,3-dimethyl signals were shifted downfield to 1.61 and 1.71 p.p.m. The difference in chemical shift (0.1 p.p.m.) was also reduced from that seen between these two signals in the selenide (0.21 p.p.m.) as they are more magnetically equivalent in (114). H-2 (the olefinic proton) appeared as a doublet with coupling constant 2Hz due to long-range coupling to H-8 through the π-system.

The fragmentation of the selenoxide produced from (113) is known to proceed in a syn-fashion and could thus produce both 1,2- and 1,8-didehydropyrrolizidines from (113b). The fact that no 1,8-didehydropyrrolizidine was detected on mcpba oxidation and fragmentation to the unsaturated ester (114) can be accounted for in two ways. Firstly, the 1β-ester (113b) could be formed and then no elimination takes place to give the 1,8-derivative, as elimination towards a bridgehead and an electron-withdrawing substituent is disfavoured. Secondly, the 1α-ester (113a) could be formed and the regioselectivity of the fragmentation is due to the favourable position of a hydrogen atom on C-2.

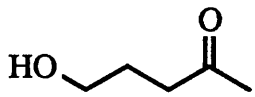
The final stage in this synthesis was the reduction of the ester group of (114) to a hydroxymethyl group to give 3,3-dimethylsupinidine (104). This was readily accomplished using DIBAH. Accurate mass data gave the molecular formula $C_{10}H_{17}NO$. In the i.r. spectrum a hydroxyl group was present at 3600 cm^{-1} and a double-bond at 1600 cm^{-1}. The ^{1}H n.m.r. spectrum again revealed the distinctive methyl signals at C-3 (1.19 and 1.21 p.p.m.). The methylene group of the hydroxymethyl appeared at 4.16 p.p.m. with the olefinic proton a broad singlet at 5.41 p.p.m. ^{13}C n.m.r. spectroscopy showed two methyl groups, four methylenes, two methines (1 olefinic) and two quaternary carbons (1 olefinic). This is consistent with the proposed structure for (104). There was insufficient time to prepare ester derivatives of 3,3-dimethylsupinidine for testing and evaluation of their biological activity.

4.3 Synthesis of 8-methylsupinidine (105)

As outlined in Section 4.1., the other possible "blocked" supinidine analogue is 8-methylsupinidine (105). As the procedures involving cyclisation of iminium ions proved successful in the route to 3,3-dimethylsupinidine, it was felt that an adaptation of this method aimed towards (105) could prove fruitful. The first obvious problem encountered here was introduction of the 8-methyl group at some stage. It was decided to do this as early as possible in the synthesis and this indicated using 2-methylproline (115).

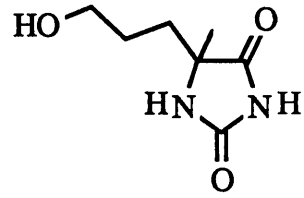
A literature preparation of this amino acid is well known[58,59] (Scheme 21). Reaction of 5-hydroxypentan-2-one (116) with sodium cyanide and ammonium carbonate gave the hydantoin (117). This was easily chlorinated to afford (118) using thionyl chloride. A high-pressure, basic fragmentation of (118) yielded 2-methylproline (115), m.p. 262-262.5°C (lit[58] 263-264.5°C). The i.r. spectrum showed hydroxyl at 3440 cm^{-1} and a carbonyl at 1600 cm^{-1}, a typical value for the ionised carboxylate anion of an amino acid. In an analagous fashion to the route to 3,3-dimethylsupinidine the acid function was protected as its benzyl ester. Treatment of 2-methylproline (115) with benzyl alcohol and P.T.S.A. afforded the p-toluene sulphonate of the benzyl ester (119) (47%). Elemental analysis of this compound confirmed the formula $C_{20}H_{25}NO_5S$. Dissolving this salt (119) in a basic solution followed by extraction with an organic solvent gave the free ester (120) in 95% yield. The i.r. spectrum showed the carbonyl absorption at 1720 cm^{-1}. The ^1H n.m.r. spectrum contained a three proton singlet (the 2-methyl group) at 1.43 p.p.m. together with the characteristic 2 proton singlet and five proton singlet at 5.23 p.p.m. and 7.27 p.p.m. due to the benzyl group of the benzyl ester. The ^{13}C n.m.r. spectrum revealed four methylene groups, one methyl group, one quaternary carbon, six aromatic carbon signals and a carbonyl carbon at 177.3 p.p.m. This is consistent with the proposed structure for (120).

SCHEME 21



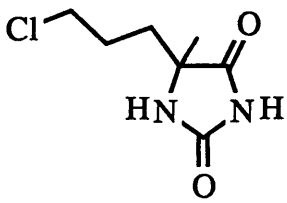
(116)

1. NaCN
2. $(NH_4)_2 CO_3$



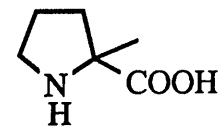
(117)

$SOCl_2$



(118)

$Ba(OH)_2$

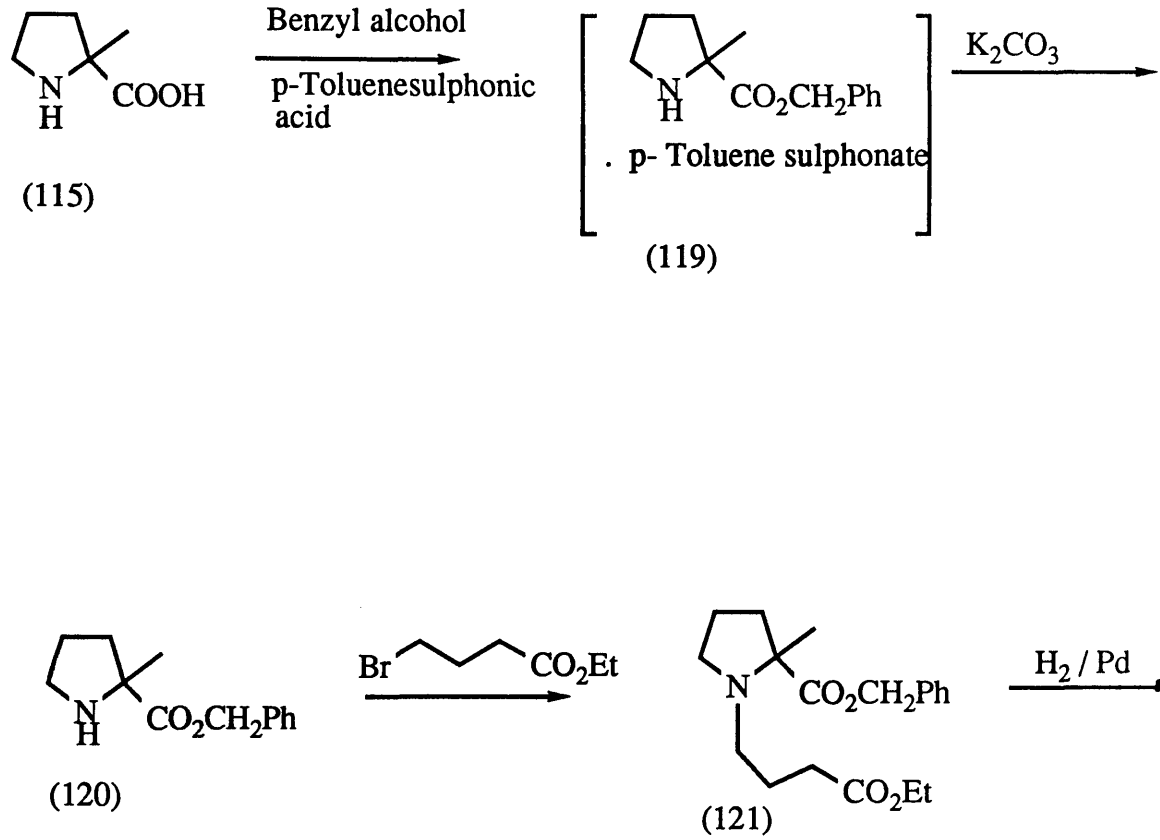


(115)

The relatively large amounts of 2-methylproline available enabled us to experiment with several options for the introduction of the N-alkyl chain. The first of these chosen was the simplest; N-alkylation using ethyl 4-bromobutanoate to give N-(3-ethoxycarbonylpropyl)-2-methylproline benzyl ester (121) (Scheme 22). Alkylation of (120) in benzene with potassium carbonate as base afforded (121) in 64% yield. In the i.r. spectrum two ester carbonyl absorptions were present at 1700 and 1725 cm^{-1}. The ^{1}H n.m.r. spectrum contained the ethyl ester signals at 1.22 and 4.13 p.p.m. The 2-methyl group was present as a singlet at 1.29 p.p.m. The benzyl ester had characteristic methylene absorption at 5.11 p.p.m. and the aromatic protons were at 7.32 p.p.m. ^{13}C n.m.r. spectroscopy revealed two carbonyl carbons (the ethyl and benzyl esters) at 173.7 and 175.0 p.p.m.

Deprotection of (121) to give the α-tertiary amino-acid (122) was carried out by catalytic hydrogenation to yield a gum which showed a complex ^{1}H n.m.r. spectrum with broad signals. However, all signals due to the benzyl ester group had disappeared and the ethyl ester remained with methylene absorption at 4.17 p.p.m. and methyl at 1.19 p.p.m. The 2-methyl signal was also recognisable as a singlet at 1.24 p.p.m. The i.r. spectrum showed two very different carbonyl absorptions as would be expected: the ester carbonyl at 1725 cm^{-1} and two signals for the carboxylate anion of an amino-acid at 1625 cm^{-1} and 3420 cm^{-1} (OH). Cyclisation of this compound was attempted as previously described using

SCHEME 22

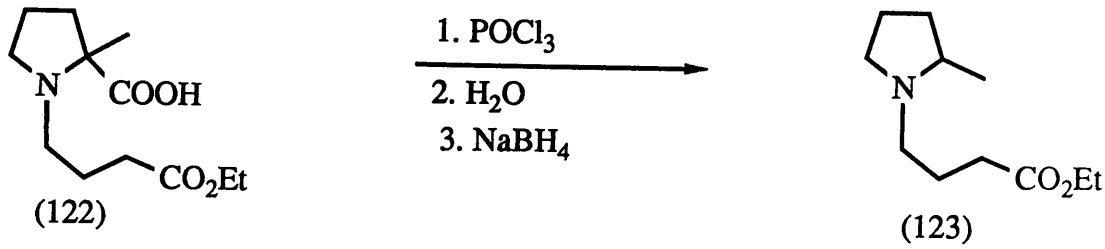


(115) (119) (120) (121) (122)

phosphorus oxychloride. However, a multi-component mixture was produced. One possibility was that the iminium ion had formed, but that no cyclisation had taken place. The other alternative was that no iminium ion formation had occurred, possibly being blocked by the 2-methyl group, and thus no cyclisation could take place. To investigate this second possibility, it was decided to repeat the cyclisation attempt, but to hydrogenate the reaction mixture after addition of phosphorus oxychloride and water. This was done and the ester (123) was isolated. Mass spectroscopy showed a molecular ion at 199 and elemental analysis of the picrolonate derivative indicated a molecular formula $C_{11}H_{21}NO_2$. I.r. spectroscopy showed a carbonyl absorption at 1725 cm^{-1} (ethyl ester). The ^1H n.m.r. spectrum revealed that the three proton singlet of the 2-methyl group of (122) had become a doublet and the ^{13}C n.m.r. had only one carbonyl absorption at 173.6 p.p.m. All of these pieces of information support the proposed structure for (123). An authentic sample of (123) was prepared by N-alkylation of 2-methylpyrrolidine with ethyl 4-bromobutanoate (Scheme 23). The mixed melting-point of the picrolonates of the compounds produced by both routes was undepressed and all spectroscopic and t.l.c. data were identical.

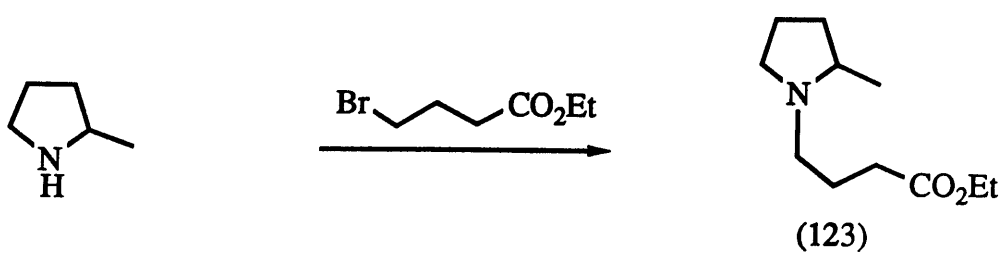
From this work, it was clear that iminimum ion formation had taken place, but no cyclisation had occurred. This could be attributed to two reasons:

SCHEME 23A



(122) (123)

SCHEME 23B



(123)

1. The 2-methyl group was blocking cyclisation;

   or

2. There was not a significant concentration of the "enol" form of the ester present to effect cyclisation.

A brief look at the acidity of α-protons shows the following:[60]

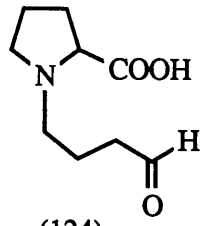
TABLE 3

Compound	pKa	% Enol (solvent)
$CH_3COC\underline{H}_2CO_2C_2H_5$	11	10-13% (C_2H_5OH)
$C\underline{H}_2(CO_2C_2H_5)_2$	13	-
$C\underline{H}_3COCH_3$	20	0.002% (H_2O)
$C\underline{H}_3CO_2C_2H_5$	25	less than 1 part in 10 million
$(CH_3)_2C\underline{H}CHO$	-	0.01 (H_2O)

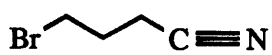
The percentage of enol available in each case should have a significant effect on the amount of cyclisation which takes place. Generally an enol tautomer is more acidic (4-5 pK units) than a keto form and thus a higher proportion of enol present should lead to easier cyclisation. Although no data could be found for the amount of enol form present in diethyl malonate (Table 3) it would be expected that this would be in the same region as the β-ketoester, ethyl acetoacetate. This would explain the relative ease of cyclisation of diester (110).

In order to investigate this effect more fully, it was decided to prepare a range of N-substituted proline derivatives. The first of these chosen was the aldehyde (124). As benzyl proline (108a) was available in greater quantity than the 2-methyl derivative, this was the compound chosen for N-alkylation (Scheme 24). Due to the reactivity of the aldehyde function, alkylation was performed using the protected compound 4-bromobutanal ethylene acetal (125). This was prepared by DIBAH reduction of 4-bromobutyronitrile to 4-bromobutanal,[61] followed by protection with ethylene glycol to give (125). The i.r. spectrum of this compound showed the C-O stretch at 1200 cm^{-1} and the C-Br absorption at 615 cm^{-1}. The ^1H n.m.r. spectrum consisted of 4 sets of peaks as would be expected with the proton to the 2 oxygen atoms at 4.87 p.p.m. The accurate mass spectrum confirmed the molecular formula as $C_6H_{11}O_2Br$ with a peak at 115 corresponding to M-Br. N-Alkylation was accomplished in 44% yield using two equivalents of benzyl proline, to give (126). The mass spectrum gave a peak at 319 (corresponding to M-1). The i.r. spectrum showed the carbonyl absorption due to the benzyl ester at 1740 cm^{-1} with the C-O stretch of the acetal at 1210 cm^{-1}. The ^1H n.m.r. spectrum was characterised by a number of overlapping absorptions. However, absorptions due to a benzyl ester (methylene singlet at 5.18 p.p.m. and five aromatic protons at 7.34 p.p.m.) were clearly visible, as was 4^1-H (the proton α to both oxygens) at 4.85 p.p.m. (triplet). Attempts at using a 1:1 ratio of

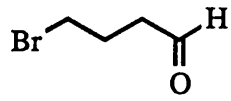
SCHEME 24

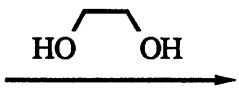


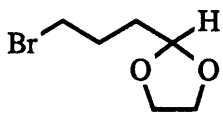
(124)



DIBAH

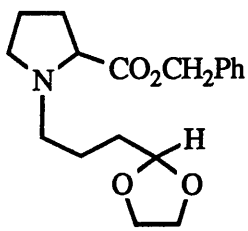




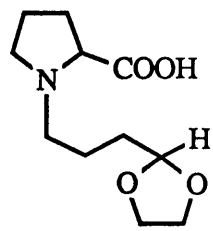


(125)

(108a)



(126)

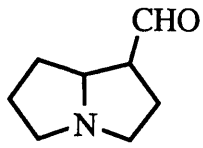


(127)

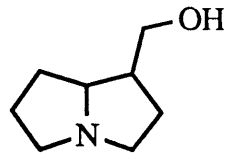
1. $POCl_3$
2. H_2O
3. pH6

No cyclised product

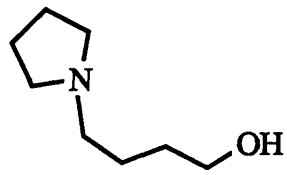
benzyl proline and acetal using bases such as DBU, TEA etc. led to yields of only ca. 20%. As benzyl proline hydrobromide is recoverable, a 44% yield using two equivalents of benzyl proline was acceptable. The α-tertiary amino-acid (127) was prepared in the usual manner using phosphorus oxychloride. ^1H n.m.r. spectroscopy showed that removal of the benzyl group was complete and the i.r. spectrum showed two peaks assignable to a carboxylate anion at 3385 cm^{-1} (OH) and 1645 cm^{-1} (carbonyl). Several attempted cyclisations of this compound proved unsuccessful. It had been hoped that the acidic conditions in which cyclisation normally occurred would de-protect the acetal to the free aldehyde which would then undergo cyclisation. One possibility was that cyclisation was occurring but that the aldehyde formed (128) was very reactive in basic conditions and underwent condensation processes. To investigate this, sodium borohydride was added to the reaction mixture to trap this aldehyde as the alcohol (129). This was unsuccessful. In fact, the sodium borohydride reduced both the iminium ion and the aldehyde to give the amino-alcohol (130) which was isolated from the reaction mixture. Accurate mass data gave the molecular formula $C_8H_{17}NO$. The i.r. spectrum showed a hydroxyl at 3590 cm^{-1}. An authentic sample of this compound was prepared for comparison as outlined in Scheme 25. All t.l.c. and spectral data were identical.



(128)

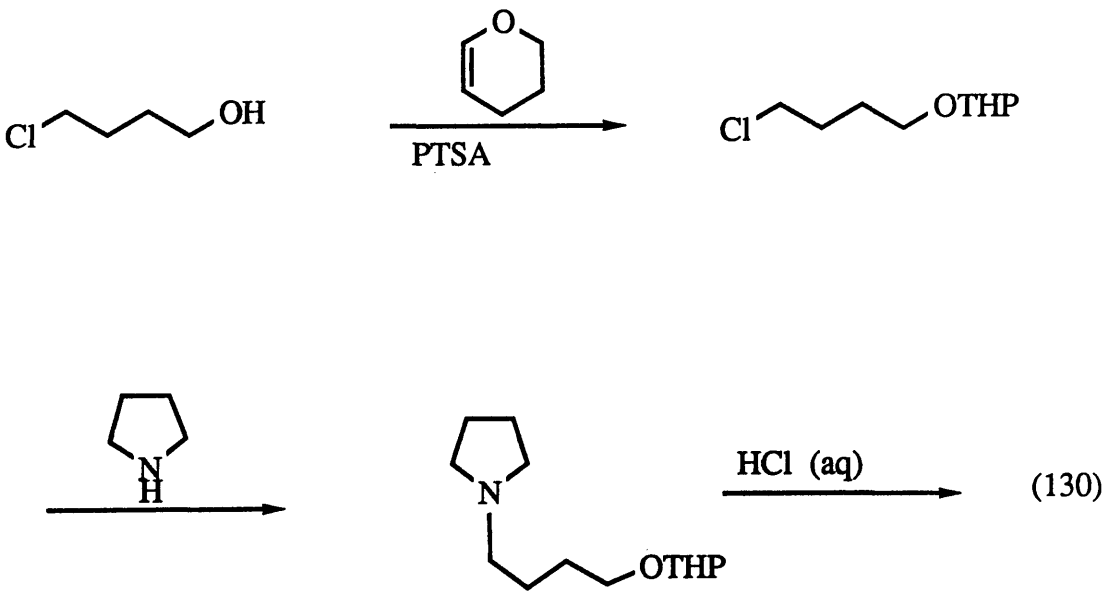


(129)



(130)

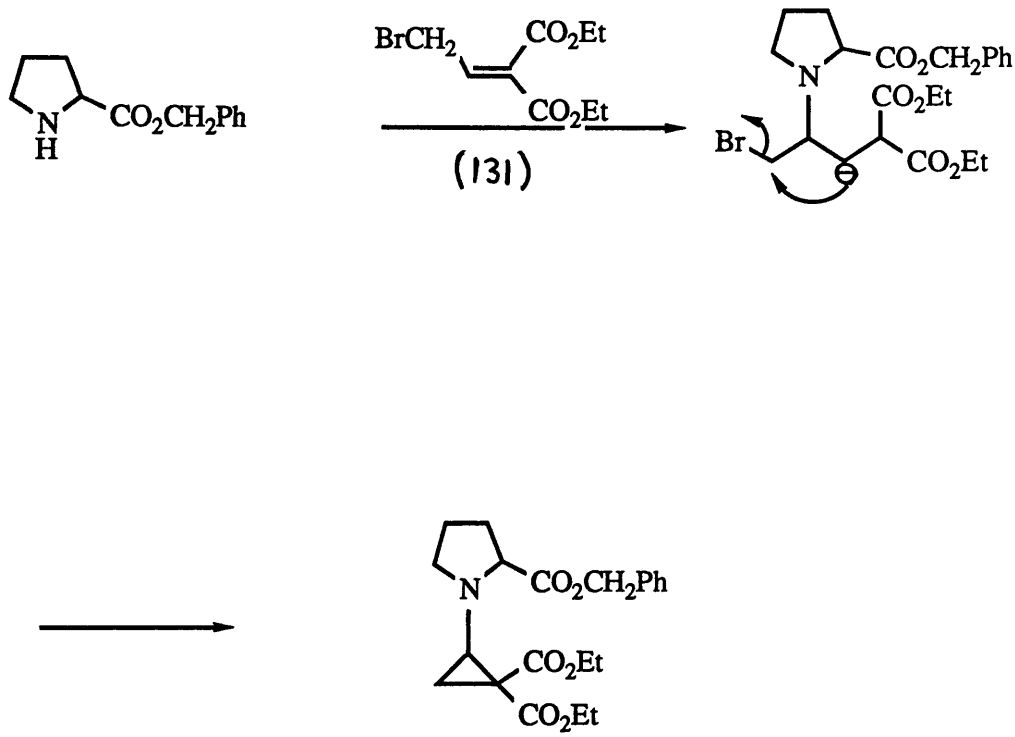
SCHEME 25



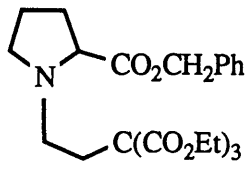
The failure of the aldehyde (127) to cyclise would be expected looking at Table 3. Moving up the table, the diesters, such as (110) had already been shown to cyclise readily. Unfortunately, the use of compounds such as (131) as alkylating agents had proved unfruitful, due to intramolecular cyclopropane formation[30] (Scheme 26). To get round this problem, a tri-ester such as (132) could be produced and then deethoxycarbonylated. The appropriate species for N-alkylation is triethyl 3-bromopropane-1,1,1-tricarboxylate (133) (Scheme 27). This was prepared according to a literature method.[62, 30] The alkylation of 2-methylproline benzyl ester was successful in benzene/DMF/potassium carbonate to give (134) (69%). The i.r. spectrum showed the existence of two distinct ester carbonyl absorptions at 1720 and 1730 cm^{-1}. The ^1H n.m.r. spectrum shows the three ethyl esters visible as methyl signals at 1.24 p.p.m. and methylenes at 4.21 p.p.m. The 2-methyl group appears as a singlet at 1.39 p.p.m. The benzyl ester group is clearly seen as a methylene absorption at 5.15 p.p.m. and a five proton aromatic signal at 7.30 p.p.m.

Deethoxycarbonylation of (134) using sodium benzylate proved unsuccessful. An alternative to this is to deprotect the benzyl ester by catalytic hydrogenation and then attempt the deethoxycarbonylation using sodium ethoxide. Hydrogenation did indeed afford the amino-acid (135). The i.r. spectrum showed a carboxylate set of signals at 3390 cm^{-1}(OH) and 1650 cm^{-1} (carbonyl)

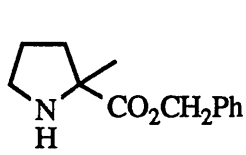
SCHEME 26


N
H
CO2CH2Ph
BrCH2
CO2Et
CO2Et
(131)
N
CO2CH2Ph
CO2Et
Br
CO2Et
N
CO2CH2Ph
CO2Et
CO2Et

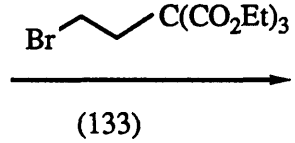
SCHEME 27



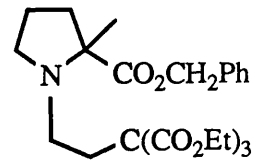
(132)



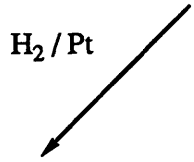
(120)

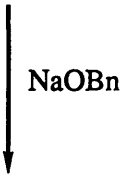


(133)

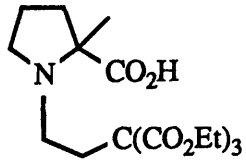


(134)

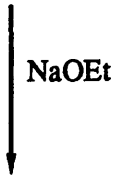




No reaction



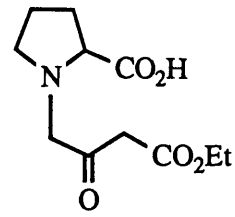
(135)



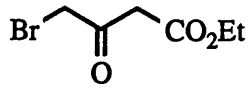
No reaction

together with the ethyl ester carbonyl absorption at 1725 cm^{-1}. The ^1H n.m.r. spectrum revealed the disappearance of all signals due to the benzyl ester group with all signals slightly broadened which is characteristic of this type of compound. All attempts, however, at deethoxycarbonylation proved unsuccessful.

Referring again to Table 3, a β-ketoester is a system which should encourage cyclisation due to the high proportion of the enol tautomeric form present. This suggested (136) as a target and (137) as the N-alkylating compound. Bromocompound (137) was prepared according to the method of Suendsen and Boll[63] (Scheme 28). N-Alkylation of proline benzyl ester with bromo-ketoester (137) gave (138). In the i.r. spectrum a broad signal was present at 1715 cm^{-1} (the benzyl ester carbonyl) and two further significant peaks at 1670 and 1570 cm^{-1} (β-ketoester in H-bonding enol form). The ^1H n.m.r. spectrum revealed a three proton triplet at 1.19 p.p.m. (methyl group of ethyl ester) and a two proton quartet at 4.09 p.p.m. (methylene group of ethyl ester). The benzyl ester group was also clearly seen as a two proton singlet at 5.09 p.p.m. and a five proton aromatic singlet at 7.30 p.p.m. Deprotection of (138) to give the amino-acid (139) was carried out by hydrogenation in the usual manner. All ^1H n.m.r. signals due to the benzyl group were seen to disappear. In the i.r. spectrum, the carboxylic acid group could be seen with absorptions at 3395 cm^{-1} (OH) and 1645 cm^{-1} (carbonyl). Analysis gave the molecular formula $C_{11}H_{17}NO_5$ which is consistent with the proposed structure. Numerous attempts

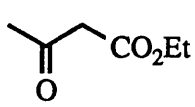


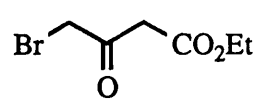
(136)



(137)

SCHEME 28





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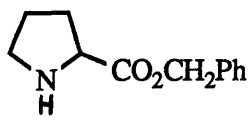

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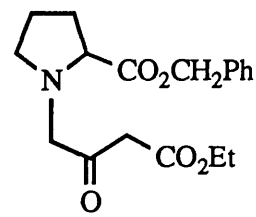
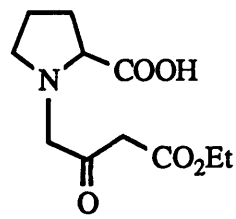


(138)

H_2 / Pd



(136)

1. $POCl_3$
2. H_2O
3. Base

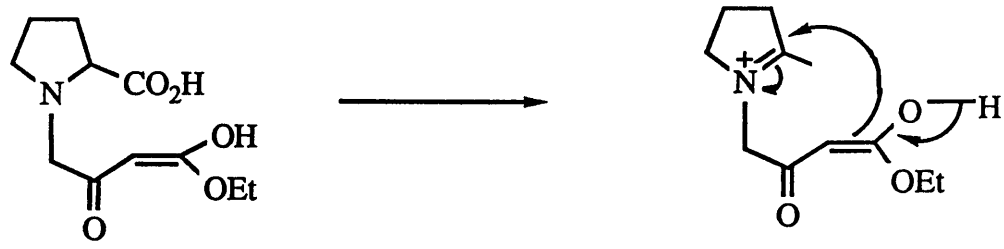
No cyclised product.

to cyclise the amino-acid (139) were unsuccessful. It is possible that the reduced flexibility of the N-alkyl chain due to the introduction of the ketone group contributed towards a degree of steric hindrance to cyclisation not seen in diesters. If the 2 compounds (110) and (136) are viewed in their "enol" forms in the context of Baldwin's Rules,[64] then it can be seen that cyclisation of (110) has an exo-component, whereas cyclisation of (136) would be classified as 5-endo-trig which is disfavoured. 5-Exo-trig cyclisations are favoured processes.

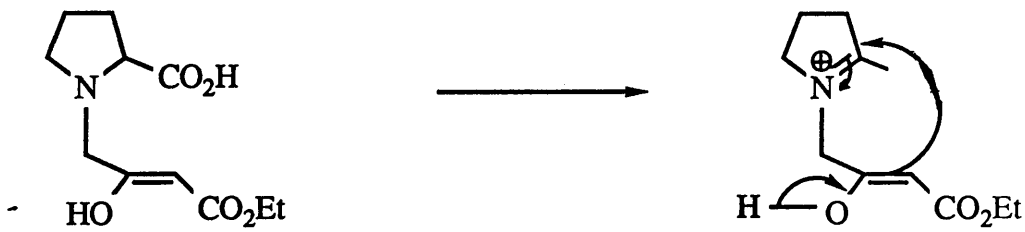
In conclusion, attempts to produce the target molecule, 8-methylsupinidine (105), using various N-alkyl groups and the iminium ion methodology proved unsuccessful. Various factors may contribute to this: steric hindrance due to the presence of a 2-methyl group in 2-methylproline; lack of sufficient enol form in the side-chains used to effect cyclisation and possible loss of flexibility in the side-chain when β-ketoesters are used i.e. difficulty in overlap of π-orbitals to form the new 5-membered ring.

4.4 Alternative procedures for iminium ion cyclisations

The experimental procedure developed by Rapoport[30] for the generation of iminium ions from α-tertiary amino-acids and which has been applied in this project, generally involves heating the amino-acid at 100°C with a large excess (10 equivalents) of phosphorus oxychloride followed by extraction of the product from an aqueous solution saturated with potassium carbonate. Typical

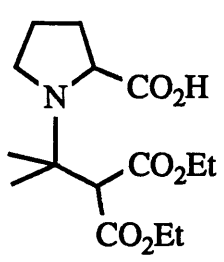


(110)

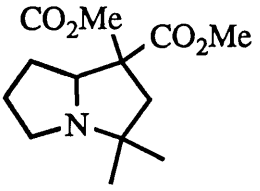


(136)

yields quoted for this process were ca.50%[30] (ca 35% in our hands). We decided to investigate possible variations in this procedure which might lead to a more general applicability over a wider range of compounds. The chosen model system was the cyclisation of (110) to (106). A range of reaction times and temperatures was investigated to establish relative yields. One other variation tried was the use of bases such as D.B.U. and T.E.A. to effect cyclisation, rather than saturating a solution of the iminium ion with potassium carbonate which could be too harsh for some compounds. The use of these bases meant that the solvent system used was changed from water to methanol-water (85:15) for solubility reasons. The effect of these changes is shown in Table 4.



(110)



(106)

TABLE 4

T(°C)	t(h)	base	solvent	yield %
100	0.2	K_2CO_3	water	36
100	24	K_2CO_3	water	34
100	0.2	DBU	methanol-water	35
100	24	DBU	methanol-water	30
25	24	K_2CO_3	water	37
25	24	DBU	methanol-water	32
100	0.2	TEA	methanol-water	29
25	24	TEA	methanol-water	31

There is remarkably little change in yield across a wide range of reaction times and temperatures with phosphorus oxychloride, even when the base and solvent used are changed. This then extends the use of the phosphorus oxychloride method of iminium ion formation to compounds unstable in saturated solutions of potassium carbonate, and at high temperature.

CHAPTER 5

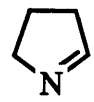
THE STABILISATION AND USE OF 1-PYRROLINE IN ALKALOID SYNTHESIS

5.1 Introduction

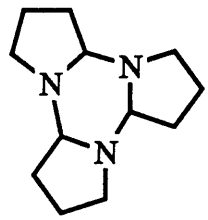
The use of 1-pyrroline (57) in organic synthesis was covered in Chapter 2.5.1 and its synthesis by conventional methods was discussed in Chapter 2.5.2. In this chapter, our work on 1-pyrroline is described under the following headings: the stabilisation of 1-pyrroline as a complex with zinc iodide; the regeneration of the free 1-pyrroline; the use of the complex in alkaloid synthesis; spectroscopic comparison of the complex and free 1-pyrroline, and the use of the 1-pyrroline zinc iodide complex in cycloaddition reactions aimed towards pyrrolizidine and indolizidine alkaloids.

5.2 Synthesis of 1-pyrroline zinc iodide complex (147)

The pioneering work in the study of 1-pyrroline and its derivatives was done by Schöpf[44] whilst looking at the reactions of γ-aminobutanal acetal (76). It was found that hydrolysis of this compound in acid did not yield the free γ-aminobutanal, but rather, 1-pyrroline (57), in aqueous solution. This compound was subsequently discovered to be stable in acid up to 100°C, but on treatment with base it trimerised to (80).[65, 47]



(57)

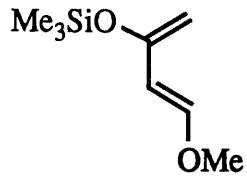


(80)

Our interest in 1-pyrroline (57) came as a result of work carried out by Danishefsky[66] who showed that imines undergo condensation with the activated diene, 1-methoxy-3-trimethylsilyoxydiene (141) in the presence of zinc chloride. If this reactivity could be extended to cyclic imines then this would open up some interesting areas of pyrrolizidine and indolizidine alkaloid synthesis.

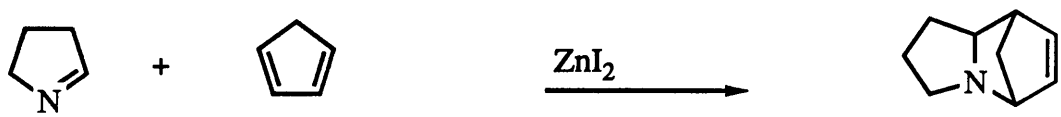
Our source of 1-pyrroline (57) was the dehydrohalogenation route used by Schöpf[44] (Scheme 13). Pyrrolidine was reacted with t-butyl hypochlorite to produce the N-chlorinated derivative and dehydrohalogenation was effected by treatment with sodium methoxide.[65] This left a solution of 1-pyrroline (57) in methanol. A mixture of (57) and the trimer (80) could be obtained by careful distillation. This mixture (approximately 3:1, (57)/(80) by ^1H n.m.r. spectroscopy was quickly dissolved in ether to minimise trimerisation.

The first diene chosen was cyclopentadiene (142) as it is readily available and should give access to a pyrrolizidine-type system (143) (Scheme 29). Cleavage of the double bond in (143) should then give a 1,3-disubstituted pyrrolizidine which could be further functionalised. The first reaction conditions chosen were to stir a mixture of 1-pyrroline (57), zinc iodide (144) and cyclopentadiene (142) under argon at room temperature. A yellow precipitate was observed which when washed with ether gave a grey



(141)

SCHEME 29



(57) (142) (143)

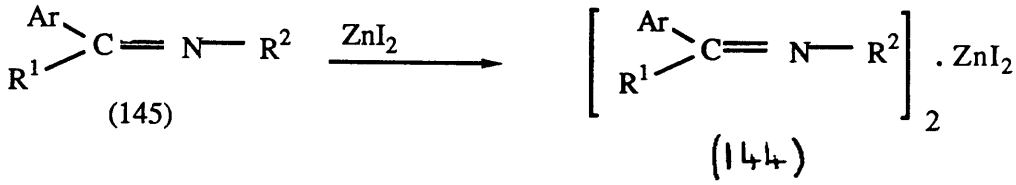
solid with a m.p. greater than 300°C. Previous work by Bartnik and Mloston[67] had established that aromatic imines such as (145) form complexes (144) with zinc iodide which can then react with a range of reagents (Scheme 30). In their work, these complexes had not been isolated or characterised, but had been reacted *in situ*. From this evidence, we postulated that this grey solid was in fact a complex (147) of zinc iodide with 1-pyrroline. Characterisation of this complex was carried out by ^{1}H n.m.r., i.r. and ^{13}C n.m.r. spectroscopy and elemental analysis. The ^{1}H n.m.r. spectrum revealed four sets of peaks. In Table 5 this spectrum is compared with that of 1-pyrolline (57) itself.[65]

TABLE 5

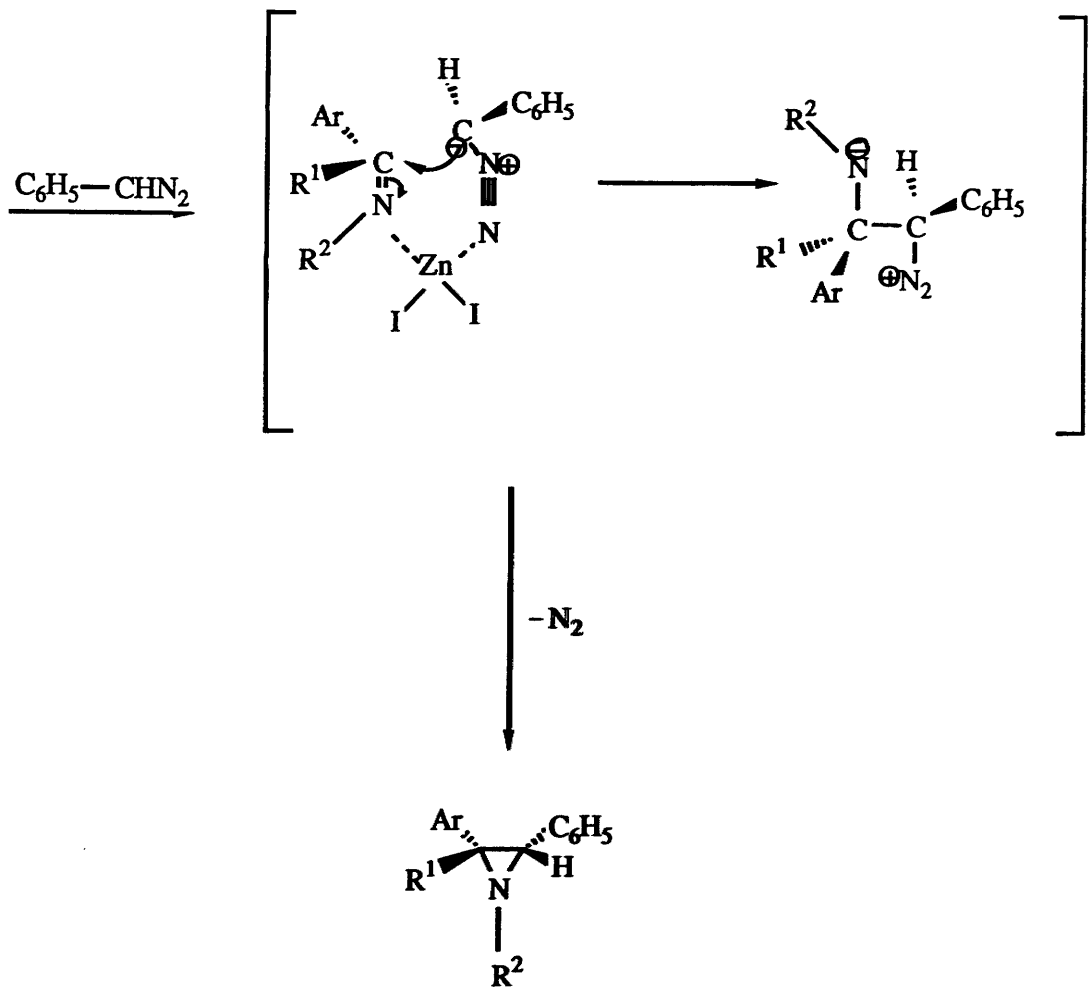
	1-pyrroline	complex	assignment
	1.62-1.92(m)	1.90-2.67(m)	C-4
chemical shift	2.50-2.75(m)	2.78-3.21(m)	C-3
(p.p.m.)	3.75-3.90(m)	3.92-4.32(m)	C-5
	7.70(t)	8.29(t)	C-2

As would be expected, all signals were shifted downfield due to the electron-withdrawing effect of the zinc. In the infra-red spectrum of the complex, the imine stretch was at 1630 cm^{-1} compared to 1620 cm^{-1} for free 1-pyrroline and the Zn-I stretch at 400 cm^{-1}

SCHEME 30



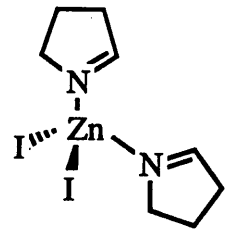
(145) (144)



$Ar = C_6H_5$

$R^1 = H, CH_3$

$R^2 = CH_3, C_2H_5$



(147)

was present. The ^{13}C n.m.r. spectrum showed four signals with C-2 at 178.5 p.p.m. compared to the more typical range of 145-160 p.p.m. for imines, again due to the effect of the zinc.

This evidence points to a zinc iodide-1-pyrroline complex of some sort. The nature of this complex was determined by elemental analysis and by comparison with zinc complexes most of which are known to be tetrahedral[69]. The elemental analysis shows an empirical formula of $C_8H_{14}N_2I_2Zn$, which when compared to known zinc complexes gives the likely molecular formula $(C_4H_7N)_2ZnI_2$ with a proposed tetrahedral arrangement of the 1-pyrroline and iodide ligands. This is consistent with the spectroscopic evidence.

5.3 Use of complex (147) in synthesis

The discovery that 1-pyrroline could be isolated as a complex with zinc iodide and easily stored as a stable solid meant that a wide range of reactions of 1-pyrroline could be attempted given two conditions:

1. 1-pyrroline could be freed easily from the complex (147) for reactions

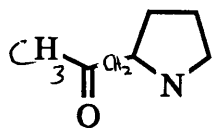
   or

2. The complex (147) could be used directly as a source of 1-pyrroline.

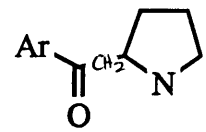
To investigate this first possibility, some n.m.r. spectroscopic experiments were performed. The complex (147) was dissolved in deuteriochloroform and 0.5 equivalents of diethylamine

were added. From comparison of the H-2 signal in the ^1H n.m.r. spectrum, the ratio of complex to free 1-pyrroline was 5:1. Further addition of diethylamine reduced this ratio, until, at 15 equivalents, the signal for the complex H-2 proton disappeared completely. Similar results were obtained using ethylene diamine, ammonia, ethylamine and triethylamine. The process here is the displacement of 1-pyrroline as a ligand by an amine. This was expected because of the existence of many complexes of Zn^{2+} with amine ligands.[70] We have thus demonstrated that 1-pyrroline can be freed from its complex with zinc iodide in organic solution by a wide range of amines.

More interesting would be the second option mentioned earlier; that the complex could be used directly in reactions as a source of 1-pyrroline, eliminating the need for regeneration of the free 1-pyrroline and possible changing of the solvent. The reactions chosen to investigate this possibility were those which were involved in the pathway towards phenanthroindolizidine alkaloids and related compounds as developed by Herbert and co-workers.[34, 36, 38, 41] As described in Chapter 2, Herbert *et al*. had generated 1-pyrroline enzymically on a small scale for use in the synthesis of norhygrine (148) and a range of 2-phenacylpyrrolidines (62). In our synthesis of norhygrine, the complex was added directly to a solution of acetoacetic acid and phosphate buffer (pH 7) in aqueous methanol. A straightforward



(148)



(62)

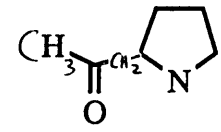
SCHEME 31

$(C_4H_7)_2ZnI_2$

(147)

CH_3CCH_2COOH

pH 7 , aqueous methanol



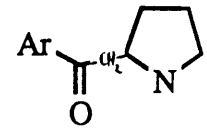
(148)

$(C_4H_7)_2ZnI_2$

(147)

$C_6H_5CCH_2COOH$

(149)

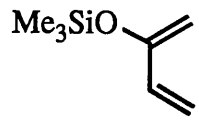


(150)

acid/base cycle for purification after three hours gave norhygrine in 85% yield. The i.r. spectrum showed the ketone carbonyl absorption at 1705 cm^{-1}. The ^1H n.m.r. spectrum displayed a three proton singlet at 2.15 p.p.m. for the methyl ketone. The semicarbazone hydrochloride had m.p. 203-205°C. All of these are in accordance with literature values.[71] A similar reaction using benzoyl acetic acid gave 2-phenacylpyrrolidine (150) in 88% yield (Scheme 31). We have therefore established that the complex (147) can be used directly as a source of 1-pyrroline in aqueous methanol. Addition of the complex to an excess of D_2O and examination of the ^1H n.m.r. spectrum showed the presence of free 1-pyrroline, so it is likely that in aqueous methanol, water displaces 1-pyrroline as a ligand.

5.4 Use of complex (147) in cycloaddition reactions

Having demonstrated the reactivity of complex (147) in reactions with β-keto acids, we set out to examine its possible uses as a dienophile. As previously discussed, the complex (147) was originally isolated and characterised after an unsuccessful attempt at the Lewis acid catalysed cycloaddition of cyclopentadiene and 1-pyrroline. The diene and reaction conditions now chosen reflected the failure of this reaction and the need to use more electron-rich dienes. The classic example of this type of diene is the trimethylsilylether of methyl vinyl ketone (151) which has been extensively used in Diels-Alder type reactions[66] and which is readily prepared from methyl vinyl ketone and



(151)

trimethylsilyl chloride. Table 6 shows a summary of some of the reaction conditions tried and results obtained.

TABLE 6

Solvent	T(°C)	time (h)	Vessel	Result
ether/acetone(50:50)	reflux	48	open	starting material
toluene	80°C	48	sealed tube	tar
neat	60°C	24	sealed tube	tar
acetonitrile	60°C	48	sealed tube	tar
toluene	25°C	24	open	starting material
toluene	50°C	24	sealed tube	tar

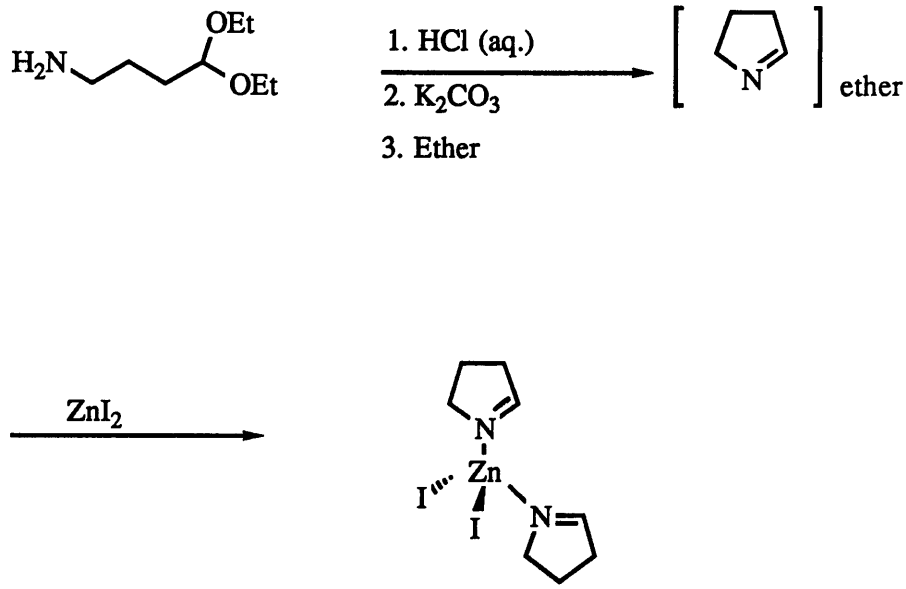
As can be seen from this table, no success was achieved despite using a range of solvents, times, pressures and temperatures. It is likely that a much more detailed investigation into this type of reaction with a range of dienes would be necessary to establish thoroughly whether the complex (147) is suitable for use as a source of 1-pyrroline as a dienophile.

5.5 Alternative routes to complex (147)

Conventional routes to 1-pyrroline (57) itself have already been discussed in Chapter 2. These often involve time-consuming and inefficient reactions or, in the case of enzymic production, problems of scaling up. Also, once the 1-pyrroline has been generated, it has to be stored in solution until needed and trimerisation is possible. In Schöpf's original work,[44]

1- pyrroline was generated from γ-aminobutanal acetal and assayed in solution with o-aminobenzaldehyde. We reasoned that significant quantities of 1-pyrroline could be obtained by the acidic hydrolysis of γ-aminobutanal diethyl acetal,which is readily available and then trapped as complex (147). This was successful as long as all organic extractions were carried out with ice-cold ether because 1-pyrroline is very volatile and significant quantities can escape (and be recognised by its strong smell) if warm ether is used. The preparation typically involved dissolving the γ-aminobutanal diethyl acetal in aqueous 2 M HCl at 0° C, adding ether and then stirring for 20 min.. The mixture was then basified with potassium carbonate and extracted with ether. After drying the ethereal solution, zinc iodide was added to the filtered solution and the precipitate was filtered off and washed with ether and hexane. Typical yields were 50% which fell to about 25% when warm ether was used for extractions (Scheme 31).Yields might be improved by further change of these conditions. In summary, we have demonstrated that 1-pyrroline (57) can be easily prepared in reasonable yield by acidic hydrolysis of γ-amino butanal diethyl acetal and isolated as a complex (147) with zinc iodide. This complex can act as a source of 1-pyrroline in a range of addition reactions and an organic solution of 1-pyrroline can be generated from the complex by

SCHEME 32



(147)

addition of a range of amines. The use of complex (147) as a source of an imine dienophile proved unsuccessful. Further work suggested in this area would include:

1. The preparation of other metal ion-1-pyrroline complexes (e.g. Cu^{+},Cd^{2+},Hg^{2+} etc.)
2. The preparation of complexes using substituted pyrrolines.
3. A thorough examination of the potential of complex (147) as a dienophile in Diels-Alder type processes.

CHAPTER 6

EXPERIMENTAL

6.1 General Notes

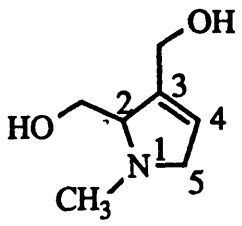
All melting points were measured with a Kofler hot-stage apparatus and are uncorrected. Infra-red spectra were obtained on a Perkin Elmer 580 spectrophotometer and ultra-violet spectra with a Pye-Unicam SP-100 spectrophotometer. Nuclear magnetic resonance spectra were recorded with a Perkin Elmer R32 spectrometer operating at 90 MHz (δ_H), a Varian XL-100 spectrometer operating at 25 MHz (δ_C) or with a Bruker WP-200 SY spectrometer operating at 200 MHz (δ_H) or 50 MHz (δ_C). Spectra were recorded for solutions in deuteriochloroform unless otherwise stated, with tetramethylsilane as internal standard. Mass spectra were determined with A.E.I. MS 12 or 902 spectrometers.

T.l.c. of the bases was carried out on Kieselgel G plates of 0.25 mm thickness and developed with chloroform-methanol-conc. ammonia (85:14:1) unless otherwise stated. The bases were located by oxidation with o-chloranil, followed by treatment with Ehrlich's reagent.[72]

Organic solutions were dried with anhydrous magnesium sulphate and solvents were evaporated off under reduced pressure below 50°C.

6.2 Experimental to Chapter 3

6.2.1 Preparation of Synthanecine A (41)

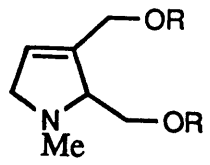


(41)

Synthanecine A (41) was prepared according to the method of Mattocks[32, 33] as modified by Barbour and Robins.[49] All analytical data agreed with literature values.

6.2.2 Preparation of derivatives of Synthanecine A (41)

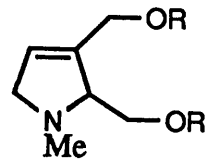
2,3-bis-ethylaminocarbonyloxymethyl-1-methyl-3-pyrroline (53).



(53) R = CONHEt

This compound was prepared according to the procedure used by Mattocks.[32, 33] All analytical data obtained agreed with literature values.

Synthanecine A bis (phenyl carbonate) (56)[32]

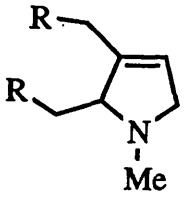


(56) $R = CO_2Ph$

Phenyl chloroformate (1.58g, 0.01 mol, 10 equivalents) was added, in portions, to a solution of Synthanecine A (41) (0.143g, 0.001 mol) in pyridine (10 ml) and then the solution was allowed to stir at room temperature for 24h. The mixture was then added to water (10 ml) with ice-cooling. The solution was acidified with 2M hydrochloric acid and washed with ether (3 x 30 ml). The aqueous solution was basified with conc. ammonia solution with ice-cooling and extracted with ether (3 x 50 ml). The combined ether extracts were dried ($MgSO_4$) filtered and concentrated under reduced pressure to give the title compound (56) (0.172g, 45%).

ν_{max} ($CHCl_3$) 3070, 2790, 1760, 1455 and 1260 cm^{-1}; δ_H(90MHz) 2.56 (3H, s, N-$\underline{CH_3}$), 3.16-3.54 (1H, complex, 2-H), 3.81 (2H, complex, 5-H), 4.52 (2H, d, $\underline{J}$ 7Hz, 6-H), 4.96 (2H, s, 7-H), 5.97 (1H, s, 4-H), 7.21-7.68 (6H, complex, aromatic) and 7.83-8.24 p.p.m. (4H, complex, aromatic); $\underline{M/Z}$ 383 ($\underline{M}^+$), 197, 169, 153, 127 and 112 [Found: $\underline{M}^+$, 383.1370. $C_{21}H_{21}NO_6$ requires $\underline{M}$, 383.1369.]

Synthanecine A bis (ethyl carbonate) (84)

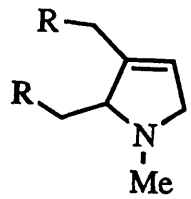


(84) R = COOEt

Ethyl chloroformate (1 ml, 1.135g, 0.01 mol, 10 equivalents) was added, in portions, to a solution of Synthanecine A (0.142g, 0.001 mol) in pyridine (10 ml) and the solution allowed to stir at room temperature for 24h. The mixture was then added to water (10 ml) with ice-cooling. The solution was acidified with 2M hydrochloric acid and washed with ether (3 x 30ml). The aqueous solution was basified with conc. ammonia solution with ice-cooling and extracted with ether (3 x 50ml). The combined ether extracts were dried ($MgSO_4$) filtered and concentrated under reduced pressure to give the title compound (84) (0.115g, 40%);

$\nu_{max}(CHCl_3)$ 2965, 2720, 1705, 1150 and 940 cm^{-1}; δ_H(90 MHz) 1.10-1.38 (6H, complex, 3 x $CO_2CH_2\underline{CH_3}$), 2.51 (3H, s, $NC\underline{H_3}$), 3.12-3.62 (1H, complex, 2-H), 3.80 (2H, complex, 5-H), 4.00-4.37 (4H, complex, 3 x $CO_2\underline{CH_2}$), 4.48 (2H, d, J 8Hz, 6-H), 4.90 (2H, s, 7-H) and 5.89 (1H, s, 4-H); M/Z 287 ($\underline{M}^+$), 260, 245, 230, 153 and 111 [Found : $\underline{M}^+$, 287.1367, $C_{13}H_{21}NO_6$ requires M, 287.1369.]

(±)-6,7,O,O-Dibenzoyl. Synthanecine A (Synthanecine A bis(benzoate))(85)



(85) R = COPh

Benzoyl chloride (0.37g, 2.64 mmol, 3 equivalents) was added dropwise, with ice-cooling, to a solution of Synthanecine A (0.12g, 0.88 mmol) in a mixture of THF (20ml) and pyridine (5ml). The mixture was stirred at room temperature for 1h, then poured into ice-water (25ml). The solution was acidified with 2M hydrochloric acid and washed with ether (3 x 50ml). The aqueous solution was basified with conc. ammonia solution with ice-cooling and extracted with chloroform (3 x 50ml). The chloroform extracts were dried, filtered and evaporated at 100°C under reduced pressure to give (±)-6,7,O,O-dibenzoyl Synthanecine A (85) as a brown oil (0.18g, 58%), R_F0.75, after column chromatography on basic alumina using methylene chloride with increasing proportions of chloroform;

ν_{max}($CHCl_3$), 2975, 2790, 1715, 1450 and 1270 cm^{-1};

δ_H (90MHz) 2.59 (3H, s, N-CH_3), 3.18-3.58 (1H, complex, 2-H),

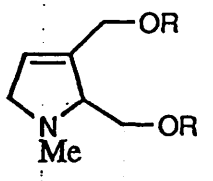
3.80 (2H, complex, 5-H), 4.45 (2H, d, J 6Hz, 6-H), 4.96 (2H, s, 7-H),

5.92 (1H, s, 4-H), 7.15-7.59 (6H, complex, aromatic) and 7.80-8.19

p.p.m. (4H, complex, aromatic); M/Z 351 (M^+), 274, 246, 110, 82

and 42 [Found: M^+, 351.1474. $C_{21}H_{21}NO_4$ requires M, 351.1471.]

(±)-6,7,0,0,-Diacetyl synthanecine A (86)



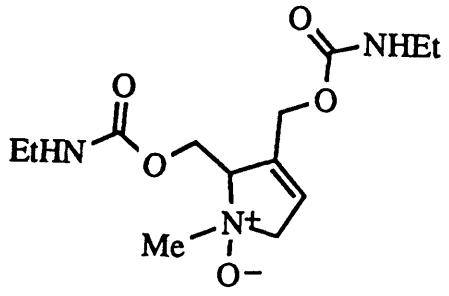
(86) R = COMe

This compound was prepared according to the procedure used by Barbour and Robins.[49] All analytical and spectral data were identical to those of an authentic sample.

Attempted synthesis of synthanecine A bis (diphenyl phosphate)

Diphenyl phosphoryl chloride (3 mmol) was added slowly with stirring to a solution of synthanecine A (1 mmol) in pyridine (3 ml) and the mixture allowed to stand at room temperature for 3h. After a conventional acid/base cycle, no basic material could be isolated.

Synthanecine A bis-(N-ethylcarbamate)N-Oxide (89)



(89)

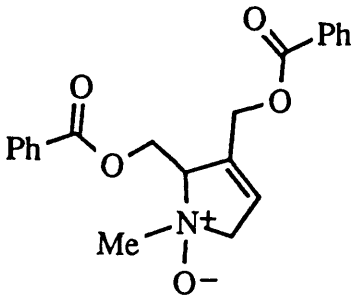
Synthanecine A bis-(N-ethylcarbamate) (0.094g, 0.33 mmol) was stirred at room temperature in methanol (5 ml) together with aqueous hydrogen peroxide (0.3 ml) which had been stablised with sodium pyrophosphate. After 16h, the mixture was heated at reflux for 2h. Excess of peroxide was decomposed by addition of manganese dioxide, and the solution filtered through Celite. The solution was then concentrated under reduced pressure to give the crude N-oxide (89) as a brown gum (0.08g, 81%).

t.l.c. - Product gave a single spot in the following solvent and plate systems;

Plate	Solvent	R_F
silica	1	0.61
silica	2	0.67
alumina	1	0.34
cellulose	1	0.82

The product gave a positive test for a N-oxide using the procedure of Mattocks;[53] δ_H(90MHz) 3.20(3H, s, N^+-CH_3).

(±)-6,7,0,0-Dibenzoyl synthanecine A N-oxide (90)



(90)

This compound was prepared in an identical way to (89), using (±)-6,7,0,0-dibenzoyl synthanecine A (84). The product was a brown gum (75%).

t.l.c. - Product gave a single spot in the following solvent and plate systems;

Plate	Solvent	R_F
silica	1	0.65
silica	2	0.73
alumina	1	0.48
cellulose	1	0.91

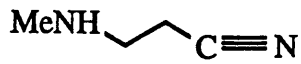
Solvent 1 was butanol/acetic acid/water, (3:1:1). Solvent 2 was isopropanol, 33% conc. ammonia. Compound (90) gave a positive test for an N-oxide using the procedure of Mattocks;[53] δ_H (90 MHz) 3.24 (3H, s, N^+-CH_3).

Attempted Synthesis of (±)-6,7,O,O-Diacetyl synthanecine A N-methyl ammonium iodide (93)

Methyl iodide (0.05g, 0.4 mmol, 1.5 equivalents) was added to a solution of (±)-6,7,O,O-diacetyl synthanecine A (86) in ether (5 ml) and the solution stirred at room temperature for 48h. T.l.c. at this time showed only starting material present. Evaporation of the solvent left only starting material. Various reaction temperatures and times were tried in several solvents (acetone and THF) but only starting material could be isolated.

6.3. Synthesis of 3-Hydroxymethyl-1-methyl-2,5-dihydropyrrole (103).

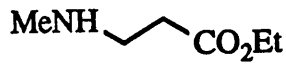
β-Cyanoethylmethylamine (97)



(97)

Acrylonitrile (60g, 1.13 mol) was added slowly, with stirring and cooling to 0°C, to a solution of methylamine (42g, 1.36 mol) in methanol (125 ml). The solution was allowed to stand at room temperature for 48h, then the solvent was removed in vacuo and the resultant oil was distilled (70°C/12mmHg, lit. 74°C/16 mmHg)[73] to leave the title compound (97) as a yellow oil, R_F0.52; ν_{max} (thin film) 3300, 2975, 2810 and 2245 cm^{-1}; δ_H(90 MHz) 1.30 (1H, s, N-H), 2.46 (3H, s, N-CH_3), 2.53 (2H, t, J 7Hz, NCCH_2) and 2.89 p.p.m. (2H, t, J 7Hz, N-CH_2); M/Z 84 (M^+), 69, 54, 53, 52 and 44. The picrolonate had m.p. 238-239°C (EtOH). [Found:C, 48.29; H, 4.35; N, 24.10; $C_{14}H_{16}N_6O_5$ requires C, 48.28; H, 4.63; N, 24.13%].

β-Ethoxycarbonylethylmethylamine (98)

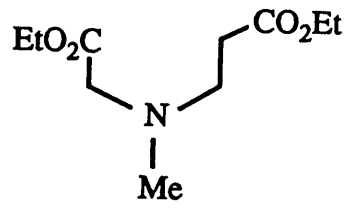


(98)

Hydrogen chloride gas was added to a solution of β-cyanoethylmethylamine (97) (70g, 0.84 mol) in ethanol (250 ml), until no more ammonium chloride precipitated. The mixture was left at room temperature for 2h then heated at reflux for 2h, cooled and filtered. The filtrate was concentrated to one quarter of its original volume under reduced pressure. An equal volume of water was then added and the solution was washed with chloroform (3 x 70 ml). The aqueous layer was basified with cooling with conc. ammonia solution, then extracted with chloroform (3 x 100 ml). The combined chloroform extracts were dried, filtered and evaporated to leave an oil which was distilled (b.p. 62°C/12 mmHg, lit[74] 65°C/15 mmHg) to leave the title compound (98) (93g, 85%) as a

green oil, R_F0.46; ν_{max} (thin film) 3320, 2980, 2790, 1730 and 1370 cm^{-1}; δ_H(90 MHz) 1.22 (3H, t, J 8Hz, $CO_2CH_2\underline{CH_3}$), 1.27 (1H, s, N-H), 2.40 (3H, s, NCH_3), 2.47 (2H, t, J 7Hz, $\underline{CH_2}CO_2Et$), 2.82 (2H, complex, N-$\underline{CH_2}$) and 4.14 p.p.m. (2H, q, J 8Hz, $CO_2\underline{CH_2}CH_3$); M/Z 131 (M^+), 85, 83, 55 and 44. The picrolonate had m.p. 175-177°C (EtOH). [Found: C, 48.67; H,5.24; N, 17.73. $C_{16}H_{20}N_5O_7$ requires C, 48.73; H, 5.11; N, 17.76%].

N- β-Ethoxycarbonylethyl-N-ethoxycarbonylmethyl methylamine (99)[32,74]

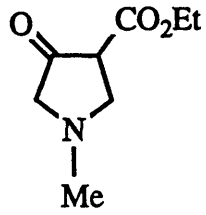


(99)

Potassium carbonate (26.7g) and ethyl bromoacetate (21.5 ml. 32.25g, 193 mol) were added to a solution of β-ethoxycarbonylethyl methylamine (98) (23g, 0.176 mol) in aqueous acetone (250 ml of 7% aqueous) and the mixture was heated at 60°C for 8h. The mixture was then cooled, filtered, acidified with 2M hydrochloric acid and washed with ether (3 x 200 ml). The aqueous solution was basified with cooling with conc. ammonia solution and extracted with ether (3 x 250 ml). The combined organic extracts were dried, filtered and concentrated under reduced pressure which on distillation gave sarcosine ethyl ester (b.p. 45°C/12mmHg, lit.[74] 43°C/10mmHg) and the title compound (99) (21g, 55%) (b.p. 128°C/12mmHg, lit.[74] 124-125°C/10mmHg,

R_F0.82; ν_{max} (thin film) 2980, 2810, 1730 and 1170 cm^{-1}; δ_H(90MHz) 1.22 (3H, t, J 8Hz, $CO_2CH_2\underline{CH_3}$), 1.24 (3H, t, J 8Hz, $CO_2CH_2\underline{CH_3}$), 2.39 (3H, s, N-$\underline{CH_3}$), 2.48 (2H, t, J 7Hz, $\underline{CH_2}CO_2Et$), 2.89 (2H, t, J 7Hz, $CH_2\underline{CH_2}N$), 3.28 (2H, s, N-$\underline{CH_2}$-CO_2Et), 4.14 (2H, q, J 8Hz, $\underline{CH_2}CH_3$) and 4.18 p.p.m. (2H, q, J 8Hz, $\underline{CH_2}CH_3$); M/Z 217 (M^+), 144, 130, 116, 102, 74 and 44. The picrate had m.p. 83-84°C (EtOH). [Found: C, 42.92; H, 4.64; N,12.46; $C_{16}H_{22}N_4O_{11}$ requires C, 43.05; H, 4.93; N, 12.56%].

Ethyl 1-methyl-3-pyrrolidone-4-carboxylate (100)

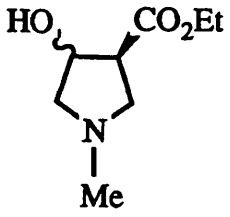


(100)

The diester (99) (10g, 46 mmol) was stirred in benzene (100 ml) with sodium hydride (2.44g of a 50% dispersion in oil, 1.22g, 0.05 mol, 1.1 equivalents) at 0°C under argon for 5h and then allowed to warm to room temperature. Water (100 ml) was added and the solution was stirred for 2h. The solution was then washed with ether (3 x 200 ml) and the aqueous layer was acidified with 2M hydrochloric acid. The acid solution was washed with ether (3 x 200 ml), then the pH adjusted to 6 with ice-cooling with conc. ammonia solution. The solution was extracted with ether (3 x 200 ml) and then the pH was adjusted to 8 and the solution was extracted with ether (3 x 200 ml). The combined organic extracts were dried, filtered and concentrated under reduced pressure to leave the title compound (100) (3.15g, 40%) as a brown oil;

R_F0.32; ν_{max} (thin film) 3050, 2800, 1725, 1675, 1580 and 1150 cm^{-1}; δ_H(90MHz) 1.20 (3H, t, J 8Hz, 9-H), 2.36 (1H, complex, 4-H), 2.51 (3H, s, 6-H), 2.71 (2H, complex, 2-H), 3.35 (2H, complex, 5-H) and 4.22 (2H, q, J 8Hz, 8-H); M/Z 171 (M^+). 142. 112, 100, 86, 73 and 70 [Found: M^+, 171.0890; $C_8H_{13}NO_3$ requires M, 171.0895]. Neither the pictrate nor the picrolonate could be formed.

Ethyl 4-hydroxy-1-methylpyrrolidine-3-carboxylate (101)



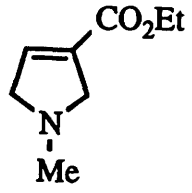
(101a) R = β-OH

(101b) R = α-OH

Platinum oxide catalyst (0.8g) and 2M hydrochloric acid (15 ml) were added to a solution of the pyrrolidone (100) (2.8g, 16.4 mmol) in ethanol (100 ml). This mixture was hydrogenated at 7 atm. for 72h, filtered through Celite and the solvent was removed under reduced pressure. The residue was dissolved in 2M hydrochloric acid (50 ml) washed with chloroform (3 x 50 ml) and then basified with cooling with conc. ammonia solution. The basic solution was extracted with chloroform (3 x 50 ml). The organic extracts were dried, filtered and evaporated under reduced pressure to leave a brown oil which was purified by column chromatography on alumina (methylene chloride/chloroform, 2:1) to leave the ester

(101) (1.1g, 39%); R_F 0.32; ν_{max} ($CHCl_3$) 3400, 2950, 2800, 1725 and 1170 cm^{-1}; δ_H(90 MHz) 1.27 (3H, t, J 8Hz, 9-H), 2.30-3.31 (6H, complex, 2-H, 3-H, 4-H and 5-H), 2.39 (3H, s, 6-H), 4.19 (2H, q, J 8Hz, 8-H) and 4.50 p.p.m. (1H, br, O-H); M/Z 173 (M^+), 144, 128, 126 and 98. [Found: M^+, 173.1059; $C_8H_{15}NO_3$ requires M, 173.1052]. Neither the picrate nor the picrolonate could be formed.

Ethyl 1-methyl-2,5-dihydropyrrole-3-carboxylate (102)

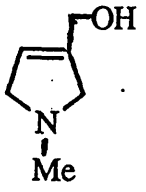


(102)

Phosphorus oxychloride (2.8 ml, 4.4g, 29 mmol, 5 equivalents) was added to the hydroxy pyrrolidine (101) (1g, 5.78 mmol) in pyridine (100 ml) and the mixture was heated at 110°C for 4h. The mixture was then cooled in an ice-bath and ice-water (40 ml) was added to destroy excess phosphorus oxychloride. The solution was evaporated to dryness under reduced pressure. The residue was then dissolved in dilute hydrochloric acid (40 ml), washed with chloroform (3 x 40 ml) and then basified with dilute ammonia solution. The basic solution was extracted with chloroform (3 x 100 ml). The organic extracts were then dried, filtered and evaporated under reduced pressure to leave the crude product, which was purified by column chromatography on alumina (methylene chloride/chloroform, 1:1) to leave

Ethyl 1-methyl-2,5-dihydropyrrole-3-carboxylate (102) (0.63g, 70%) as a brown oil; R_F 0.55; ν_{max} ($CHCl_3$) 2970, 2790, 1730, 1640 and 1200 cm^{-1}; δ_H (200MHz) 1.21 (3H, t, J 8Hz, 9-H), 2.51 (3H, s, 6-H), 3.40 (1H, d, J_{gem} 15Hz, 2-H), 3.90 (1H, d, J_{gem} 15 Hz, 2-H) 4.15 (1H, dd, J_{gem} 17Hz, J_{vic} 5Hz, 5-H), 4.27 (2H, q, J 8Hz, 8-H), 4.45 (1H, dd, J_{gem} 17Hz, J_{vic} 6Hz, 5-H) and 6.70 p.p.m. (1H, br s, 4-H); M/Z 155 (M^+), 140, 126, 110, 108 and 78 [Found: M^+, 155.0948; $C_8H_{13}NO_2$ requires M, 155.0946]. Neither the picrate nor the picrolonate could be formed.

3-hydroxymethyl-1-methyl-2,5-dihydropyrrole (103)



DIBAH in toluene (6.4 ml of a 1.5 M solution = 9.6 mmol, 3 equivalents) was added to a solution of the ester (102) (0.5g, 3.2 mmol) in toluene (10 ml) under argon and cooled to 0°C. The solution was stirred at 0°C for 2h. Glauber's salt (3g) was then added to the mixture, followed by alumina (1.2g) and the mixture was stirred at room temperature for 15 min. The whole mixture was then loaded onto an alumina column (50g) and eluted with methanol/ethyl acetate (2:3) to give 3-hydroxymethyl-1-methyl-2,5-dihydropyrrole (103)

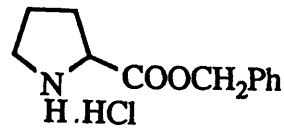
as an oil (0.26g, 71%); R_F 0.15; ν_{max} ($CHCl_3$) 3410, 2980, 2810 and 1630 cm^{-1}; δ_H (200 MHz) 2.49 (3H, s, 6-H), 3.15 (1H, dd, J_{gem} 16Hz, J_{vic} 4Hz, 5-H), 3.42 (1H, d, J_{gem} 18Hz, 2-H) 3.84 (1H, dd, J_{gem} 16Hz, J_{vic} 5Hz, 5-H), 3.94 (1H, d, J_{gem} 18 Hz, 2-H), 4.21 (br s, 7-H) and 5.81 p.p.m. (1H, br s, 4-H); M/Z 113 (M^+), 82, 52 and 37. [Found: M^+, 113.0844; $C_6H_{11}NO$ requires M, 113.0841].

Neither the picrate nor the picrolonate could be formed.

6.4 Experimental to Chapter 4

6.4.1 3,3-Dimethyl supinidine

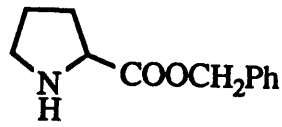
Proline benzyl ester hydrochloride (108)



(108)

This was prepared according to the method of Ramachandran and Li, and had m.p. 147-148°C (lit.[75] 148-149°C). [Found C,59.74; H, 6.81; N, 5.85%. $C_{12}H_{16}NO_2Cl$ requires C, 59.63; H, 6.67; N, 5.80%].

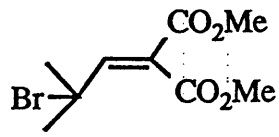
Proline benzyl ester (108a)



(108a)

Benzyl proline hydrochloride (10g, 0.044 mmol) was dissolved in a solution of potassium carbonate in water (30g in 300 ml). The resulting solution was extracted with ether (3 x 100 ml). The ether extracts were dried ($MgSO_4$), filtered, and the solvent was removed under reduced pressure to leave the free base (108a) (6.83g, 80%) as a brown oil, R_F 0.75; ν_{max} (thin film) 2960, 2870, 1730, 1660, 1500, 1450, 1200 and 1170 cm^{-1}; δ_H(90 MHz) 1.61-2.32 (4H, complex, 3-H and 4-H), 2.42 (1H, s, 1-H), 2.75-3.20 (2H, complex, H-5), 3.70-3.90 (1H, complex, 2-H), 5.16 (2H, s, 6-H), 7.31 (5H, s, aromatic); M/Z 206, 204, 160, 105, 92, 89, 77 and 70. The picrolonate had m.p. 157-158°C (EtOH) [Found: C,56.08; H,4.90; N, 15.04. $C_{22}H_{23}N_5O_7$ requires C, 56.29; H, 4.94; N, 14.92%].

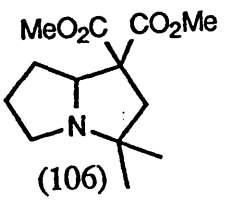
Dimethyl 2-bromo-2-methylpropylidenemalonate



This was prepared according to the method of Verhé et al.,[76] yield = 68%;

δ_C(25 MHz) 32.95 (2 x $\underline{C}H_3$-CBr), 52.54 ($CO_2\underline{C}H_3$), 52.84 ($CO_2\underline{C}H_3$), 57.92 ($\underline{C}$-Br), 124.35 (CH=$\underline{C}$), 148.35 ($\underline{C}$H=C), 163.91 ($\underline{C}O_2$Me) and 165.83 p.p.m. ($\underline{C}O_2$Me).

Dimethyl 3,3-dimethylpyrrolizidine-1,1-dicarboxylate (106)



(106)

This was prepared according to the method of Csendes et al.,[30] yield = 37%; R_F 0.60; ν_{max} (thin film) 2970, 1740, 1600, 1430 and 1350 cm^{-1}, δ_H (90 MHz) 1.07 (3H, s, CH_3), 1.24 (3H, s, CH_3), 1.56-2.41 (6H, m, 5-, 6-, 7-H), 2.59-3.20 (3H, m, 2-, 8-H), 3.71 (3H, s, CO_2CH_3) and 3.74 (3H, s, CO_2CH_3); M/Z 255 (M^+), 224, 193, 191, 181, 59 and 31 [Found: M^+, 255.1470; $C_{13}H_{21}NO_4$ requires M, 255.1471].

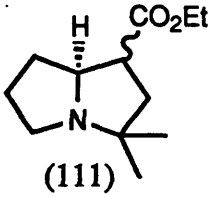
(106) - Alternative preparation

Phosphorus oxychloride (0.93 ml, 1.53g, 10 mmol, 10 equivalents) was added to N-[1,1-Dimethyl-3,3-bis(methoxycarbonyl)-n-propyl]-proline (110) (0.3g, 1 mmol) and the mixture was heated at 100°C for 3 min.. The mixture was rapidly cooled and ice-water (15 ml) was added, followed by methanol (85 ml). Diazabicyclo[5.4.0]undecene (D.B.U.) (9 ml, 9.15g, 30 mmol, 30 equivalents) was then added and the mixture was stirred at room temperature for 16h, and poured into water (100 ml). The solution was extracted with ether (3 x 150 ml). The ether extracts were washed with water (3 x 150 ml), dried, filtered and the solvent removed under reduced pressure to leave (106) as an oil (0.09g, 35%). All spectroscopic data were identical with those obtained from a sample prepared by the previous method.

Attempted demethoxycarbonylation of diester (106)

A mixture of the diester (106) (0.15g, 0.59 mmol), NaCl (0.04g, 0.68 mmol, 1.15 equivalents) and water (22μl, 0.022g, 1.22 mmol, 2.07 equivalents) was heated at reflux in dry dimethyl sulphoxide (1.5 ml) for 2h. The mixture was cooled and added to dilute aqueous ammonia (15 ml) and extracted with chloroform (3 x 30 ml). The chloroform extracts were washed with water (3 x 100 ml), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to leave a brown oil (0.066g) which showed a complex mixture of compounds by t.l.c. Reduction of the time of reflux and varying reaction temperatures gave similar results.

Ethyl 3,3-dimethylpyrrolizidine-1-carboxylate (111)

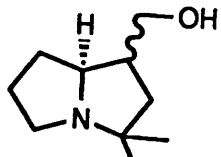


(111)

The diester (106) (0.15g, 0.59 mmol) was dissolved in 6M $HCl_{(aq)}$ (50 ml) and the solution was heated at reflux for 18h. After evaporation of the water, super-dry EtOH (40 ml) was added to the residue and dry HCl gas bubbled into the solution for 10 min. The mixture was then heated at reflux for 3h., cooled and brought to pH 9 or 10 (conc. ammonia) in the cold after addition of water (50 ml). The resulting mixture was extracted with ether (3 x 100 ml). The ether extracts were washed with water, dried, and the solvent removed under reduced pressure to leave the mono-ester (111) (0.106g, 82%) as a brown oil after chromatography on alumina

(CH_2Cl_2-$CHCl_3$, increasing polarity). R_F 0.50; ν_{max}(thin film) 2990, 1730, 1460 and 1270 cm^{-1}; δ_H(90MHz) 1.18 (3H, s), 1.20 (3H, s), 9-H and 10-H, 1.23 (3H, t, J 8Hz, 13-H), 1.50-2.30 (6H, complex, 2-, 6- and 7-H), 2.50-2.95 (3H, complex, 1- and 3-H), 3.69-4.00 (1H, complex, 8-H) and 4.15 p.p.m. (2H, q, J 8Hz, 12-H); δ_C(25 MHz) 176.6 (C-11); M/Z 211 (M^+), 182, 164, 162 and 134 [Found: M^+, 211.1567; $C_{12}H_{21}NO_2$ requires M, 211.1572]. Neither the picrate nor the picrolonate could be formed.

1-Hydroxymethyl-3,3-dimethylpyrrolizidine (112)



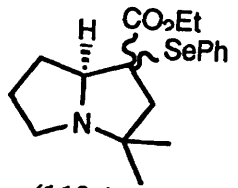
(112a) β-H1

(112b) α-H1

A solution of the ester (111) (0.25g, 1.19 mmol) in THF (2 ml) was added dropwise to a stirred suspension of lithium aluminium hydride (0.09g, 2.38 mmol) in THF (2 ml). The resultant solution was allowed to stir at room temperature for 4h., then sodium sulphate (0.3g) was added, followed by alumina (0.1g). This mixture was stirred for 15 min. and then loaded onto an alumina column (10g) and eluted with methanol/ethyl acetate (2:3) to leave the alcohol (112) as a brown oil (0.09g, 45%); R_F 0.15;

ν_{max} ($CHCl_3$) 3610, 2985, 1600 and 1460 cm^{-1}; δ_H (90MHz) 1.11 (3H, s), 1.16 (3H, s), 10-H and 11-H, 1.36-2.30 (6H, complex, 2-, 6- and 7-H), 2.42-2.91 (3H, complex, 1-H and 5-H), 3.61-3.94 (1H, complex, 8-H) and 3.68 p.p.m. (2H, d, J 7Hz, 9-H); δ_C (25 MHz) 24.7, 26.9 (C-10 and C-11), 29.9, 31.9 (C-6 and C-7), 44.2 (C-2), 46.2 (C-1), 47.2 (C-5), 61.3 (C-9), 65.4 (C-3) and 68.3 (C-8); M/Z 170 (M+1), 169 (M^+), 167, 155 and 154 [Found: M^+, 169.1474; $C_{10}H_{19}NO$ requires M, 169.1467]. Neither the picrate nor the picrolonate could be formed.

Ethyl 1-phenylseleno-3,3-dimethylpyrrolizidine-1-carboxylate (113)



(113a) 1α- ester

(113b) 1β - ester

A solution of n-butyl lithium in hexane (0.4 ml of a 1.44M solution = 0.062 mmol) was added to a solution of dry diisopropylamine (0.062 ml, 0.58 mmol) in dry THF (2 ml) over 10 min at 0°C under argon. Dry HMPA (0.5 ml) was then added and the solution was stirred for 10 min. at 0°C and then cooled to -78°C. A solution of the ester (111) (0.106g, 0.5 mmol) in dry THF (2 ml) was then added dropwise to the cooled solution. A solution of diphenyldiselenide (0.185g, 0.55 mmol) in dry THF (2 ml) was then added rapidly and the solution was stirred at -35°C for 2h, then it was allowed to warm to 0°C. Water (10 ml) was added and the mixture was extracted with ether (20 ml) and chloroform (3 x 20 ml). The combined organic layers were washed with brine (3 x 50 ml), aqueous lithium chloride (6 x 50 ml) and water (3 x 50 ml), dried,

and the solvent was removed under reduced pressure to leave a crude mixture. This was purified by column chromatography on silica (ethyl acetate/60°-80° Pet. ether, 2:1 followed by ethanol) to leave the phenylselenoester (113) (0.12g, 66%) as a red oil, R_F 0.5; ν_{max} (thin film) 2990, 1710, 1600, 1290 and 1010 cm^{-1}. δ_H (90 MHz) 1.20 (3H, s), 1.41 (3H, s), 9-H and 10-H, 1.21 (3H, t, J 9Hz, 13-H), 1.60 - 2.31 (4H, complex, 6-H and 7-H), 2.72-3.10 (4H, complex, 2-H and 5-H), 3.9 (1H, t, J 8Hz, 8-H), 4.15 (2H, q, J 9Hz, 12-H), 7.40 (3H, complex) and 7.65 p.p.m. (2H, complex), aromatic; M/Z 367, 365, 352, 350, 314, 194 and 164.

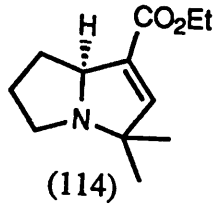
	M/Z	Reference	Measured Ratio	Measured Mass
1.	365	363.9807	1.003096	365.107584
2.	365	375.9807	1.029785	365.1060173

Theoretical mass for $C_{18}H_{25}NO_2{}^{78}Se$ = 365.1059072

	M/Z	Reference	Measured Ratio	Measured Mass
1.	367	363.9807	1.008584	367.1051103
2.	367	375.9807	1.024176	367.105556

Theoretical mass for $C_{18}H_{25}NO_2{}^{80}Se$ = 367.1049872

Ethyl 1,2-didehydro-2,3-dimethyl pyrrolizidine-1-carboxylate (114)

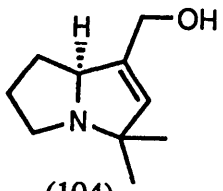


(114)

Meta-chloroperbenzoic acid (mcpba) (0.066g, 0.38 mmol, 2 equivalents) was added to the phenylseleno ester (113) (0.07g, 0.19 mmol) dissolved in methylene chloride (20 ml) at -78°C and the mixture was stirred at -78°C for 2h. Dimethyl sulphide (70 μl, 0.059g, 0.95 mmol, 5 equivalents) was then added and the cold solution was transferred by syringe into carbon tetrachloride (30 ml) heated at reflux. Concentration of the resulting solution gave the crude product which was purified by chromatography on silica [chloroform/methanol/triethylamine, 90:10:1] to leave (114) (0.034g, 85%) as a yellow oil [R_F same solvent system 0.65];

ν_{max} ($CHCl_3$) 2990, 1705, 1635 and 1380 cm^{-1}; δ_H (90MHz) 1.29 (3H, t, J 8Hz, 13-H), 1.61 (3H, s), 1.71 (3H, s), 10-H and 11-H, 1.51-1.96 (2H, complex, 6-H), 2.09-2.42 (2H, complex, 7-H), 2.55-3.09 (2H, complex, 5-H), 4.22 (2H, q, J 8Hz, 13-H), 4.36-4.70 (1H, complex, 8-H) and 6.55 p.p.m. (1H, d, J 2Hz, 2-H); M/Z 209 (M^+), 195, 194, 166, 164, 148 and 122 [Found: M^+, 209.1408; $C_{12}H_{19}NO_2$ requires M, 209/1416]. Neither the picrate nor the picrolonate could be formed.

1,2-Didehydro-1-hydroxymethyl-3,3-dimethyl pyrrolizidine (104)



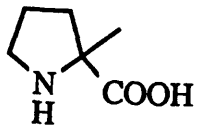
(104)

A solution of DIBAH in toluene (0.84 ml of a 1.5M solution = 1.26 mmol, 3.5 equivalents) was added to a solution of the ester (114) (0.075g, 0.36 mmol) in toluene (5 ml) cooled to 0°C and stirred under argon. This solution was stirred for 2h at 0°C and sodium sulphate (0.5g) was added followed by alumina (0.2g). This mixture was stirred for a further 15 min., then loaded on to an alumina column (10g) and eluted with methanol/ethyl acetate (2:3) to give the alcohol (104) (0.044g, 73%) as a brown oil, R_F 0.10; ν_{max} ($CHCl_3$) 3600, 2975, 1600 and 1460 cm^{-1}; δ_H (200 MHz) 1.19 (3H, s), 1.21 (3H, s), 9-H and 10-H, 1.39-2.09 (4H, complex, 6-H and 7-H), 2.76 (2H, complex, 5-H), 4.16 (2H, complex, 11-H), 4.08-4.28 (1H, complex, 8-H) and 5.41 (1H, br s, 2-H);

δ_C (50 MHz) 23.48, 30.52 (C-9 and C-10), 26.76, 29.05 (C-6 and C-7) 47.78 (C-5), 59.44 (C-11), 66.63 (C-3), 70.11 (C-8), 131.40 (C-2) and 141.95 (C-1); M/Z 167 (M^+) 154, 152, 149 and 134 [Found: M^+ 167.1306; $C_{10}H_{17}NO$ requires M, 167.1310]. Neither the picrate nor the picrolonate could be formed.

6.4.2 8-Methylsupinidine

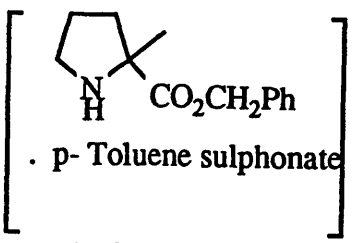
2-Methyl proline (115)



(115)

2-Methyl proline (115) was prepared according to the procedure of Ellington and Honigberg,[58], yield 85% (lit.[58] 90%); m.p. 262-262.5°C (lit.[58] 263-264.5°C) ν_{max} (KBr) 3440, 3195 and 1600 cm^{-1}; δ_H (90 MHz) 1.62 (3H, s, CCH_3), 1.94 (4H, m, CCH_2CH_2C), 3.30 (2H, m, NCH_2). All spectroscopic data were in accordance with literature values.

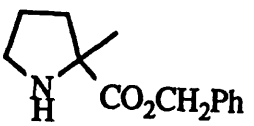
2-Methylproline benzyl ester p-toluenesulphonate (119)



(119)

2-Methylproline (0.11g, 0.853 mmol), p-toluenesulphonicacid monohydrate (0.4g, 2.27 mmol, 2.66 equivalents) and benzyl alcohol (0.6g, 5.56 mmol, 5.23 equivalents) were heated together at reflux for 16h in benzene (10 ml) using a Dean and Stark apparatus to remove water. Most of the benzene was removed by distillation, the reaction mixture was cooled to room temperature and ether (5 ml) was added with stirring. This mixture was stored at 0°C for 48h. The precipitate was washed with ether and hexane to give the p-toluenesulphonate (119) (0.155g, 47%) as a white, crystalline solid, m.p. 110-112°C (from benzene-ether). [Found: C,61.63; H, 6.43; N, 3.50; S, 8.0%. $C_{20}H_{25}NO_5S$ requires C, 61.36; H, 6.44; N, 3.58 and S, 8.19%).

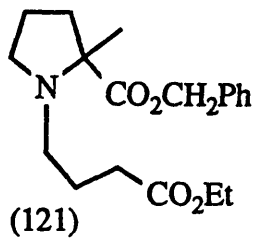
2-Methylproline benzyl ester (120)



(120)

2-Methylproline benzyl ester p-toluenesulphonate (119) (0.155g, 0.396 mmol) was dissolved in a 10% solution of aqueous potassium carbonate (20 ml). This solution was extracted with methylene chloride (4 x 30 ml). The organic extracts were dried, filtered, and the solution was evaporated under reduced pressure to leave the ester (120) as a brown oil (0.073g, 95%); R_F 0.81; ν_{max} (thin film) 2970, 2790, 1720, 1600, 1495, 1450 and 1380 cm^{-1}; δ_H (90 MHz), 1.43 (3H, s, 6-H), 1.71-2.24 (4H, complex, 3-H and 4-H), 2.30 (1H, s, 1-H), 2.67-3.18 (2H, complex, 5-H), 5.23 (2H, s, 8-H), 7.27 (5H, s, aromatic); δ_C (25 MHz) 25.2 (C-4), 26.0 (C-6), 36.8 (C-3), 46.4 (C-5), 65.8 (C-2), 66.8 (C-8), 127.9, 128.2, 128.6 and 128.9 (aromatic) and 177.3 (C-7); M/Z 201, 173, 105, 91, 85, 84.

N-(3 Ethoxycarbonylpropyl)-2-methylproline benzyl ester (121)

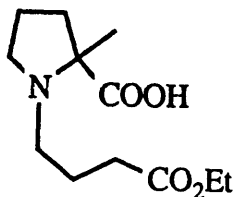


(121)

2-Methylproline benzyl ester (120) (0.197g, 1.125 mmol), ethyl 4-bromobutanoate (0.25g, 1.27 mmol, 1.13 equivalents) and potassium carbonate (0.5g) were heated together at reflux in benzene (20 ml) for 24h. A solution of potassium carbonate in water was then added (1g in 20 ml) and the mixture was stirred for 10 min., then extracted with ether (4 x 50 ml). The ether extracts were combined and extracted with 2M HCl_{aq}, (5 x 50 ml). The acidic solution was basified with potassium carbonate and extracted with ether (4 x 150 ml). The organic extracts were dried, filtered, and the solvent evaporated under reduced pressure to leave the amino diester (121) (0.23g, 64%) as a yellow oil;

R_F 0.63; ν_{max} (thin film) 2985, 2695, 1725, 1720, 1590, 1515, 1450 and 1380 cm^{-1}; δ_H (90 MHz) 1.22 (3H, t, J 7Hz, 6^1-H), 1.29 (3H, s, 6-H), 1.59-2.0 (6H, complex), 2.20-3.20 (6H, complex), 4.13 (2H, q, J 7Hz, 5^1-H), 5.11 (2H, s, 8-H) and 7.32 p.p.m. (5H, s, aromatic); δ_C (25 MHz) 14.3 (C-6), 21.6 (C-6^1), 24.3, 32.0, 37.6, 48.4, 51.1 and 60.1 (C-3, -4,-5, 1^1, 2^1 and 3^1), 66.0 (C-8), 67.8 (C-2), 128.0, 128.5 and 136.2 (aromatic), 173.7 and 175.0 p.p.m. (C-7 and C-4^1); M/Z 288, 232, 200, 199 and 198.

N-(3-Ethoxycarbonylpropyl)-2-methylproline (122)



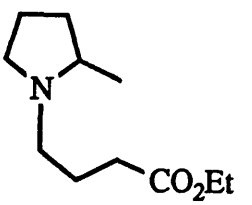
(122)

Palladium/charcoal catalyst (10%, 30 mg) was added under nitrogen to (121) (0.18g, 0.56 mmol) dissolved in absolute ethanol (20 ml). The mixture was hydrogenated at 7 atm. for 20h. The catalyst was removed by filtration through Celite under nitrogen and the solvent was evaporated under reduced pressure to give the acid ethyl ester (122) as a green gum. Trituration with ether/hexane (3:1) afforded a gum (122) (0.128g, 95%) which was used without further purification. R_F 0.0; ν_{max} ($CHCl_3$) 3420, 2995, 1725, 1625 and 1420 cm^{-1}; δ_H (90 MHz) 1.19 (3H, t, J 8Hz, 6^1-H), 1.24 (3H, s, 6-H), 1.64-2.10 (6H, complex), 2.31-3.21 (6H, complex) and 4.17 p.p.m. (2H, J 8Hz, 5^1-H).

Attempted cyclisation of N-(3-Ethoxycarbonylpropyl)-2-methylproline (122)

Phosphorus oxychloride (0.31 ml, 0.52g, 3.3 mmol, 10 equivalents) was added to the pyrrolidine ester (122) (80 mg, 0.33 mmol) and the mixture was heated at 100°C for 2 min. The solution was cooled and ice-water (10 ml) was cautiously added, then the pH was adjusted to 6.5 with potassium carbonate. The mixture was stirred at room temperature for 48h. The solution was then saturated with potassium carbonate and extracted with ether (4 x 15 ml). The organic extracts were combined, dried, filtered, and the solvent was removed under reduced pressure to leave a complex mixture which was multi-component by t.l.c..

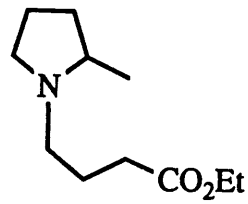
N-(3-Ethoxycarbonylpropyl)-2-methylpyrrolidine (123)



(123)

Phosphorus oxychloride (0.5 ml., 0.84g, 5.3 mmol, 10 equivalents) was added to N-(3-ethoxycarbonylpropyl)-2-methylproline (122) (0.128g, 0.53 mmol) and the mixture was heated at 100°C for 4 min.. The solution was cooled and ice-water (10 ml) was cautiously added. The pH of the solution was then adjusted to 4.0 with $NaHCO_3$. Palladium/charcoal catalyst (10%, 0.1g) was added and the mixture was hydrogenated at 3 atm. for 3h. The solution was filtered through Celite and the filtrate was made alkaline with potassium carbonate and extracted with ether (4 x 20 ml). The etheral extracts were washed with water (4 x 50 ml) and brine (2 x 50 ml), dried, and the solvent was removed under reduced pressure to leave the pyrrolidine (123) (0.02g, 19%) as a brown oil. All t.l.c. and spectral data were identical with the sample of (123) prepared by the alternative route.

N-(3-Ethoxycarbonylpropyl)-2-methylpyrrolidine (123)

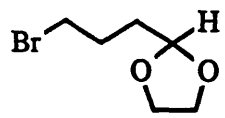


(123)

2-Methylpyrrolidine (2g, 0.024 mol), ethyl 4-bromobutanoate (4.8g, 0.025 mol, 1.04 equivalents) and potassium carbonate (10g) were heated together at reflux in benzene (200 ml) for 24h. A solution of potassium carbonate in water (10g in 200 ml) was then added and the solution was stirred for 60 min., then extracted with ether 4 x 250 ml). The organic extracts were extracted with 2M HCl (5 x 200 ml) and the aqueous extracts were made basic with potassium carbonate. The basic solution was extracted with ether (4 x 500 ml). The organic extracts were dried ($MgSO_4$), filtered and the solvent was removed under reduced pressure to leave the amino-ester (123) (3.34g, 70%) as a brown oil;

R_F 0.52; ν_{max} (thin film) 2970, 2800, 2400, 1725, 1380 and 1250 cm^{-1}; δ_H (90 MHz) 1.07 (3H, d, J 7Hz, 6-H), 1.29 (3H, t, J 8Hz, 6^1-H). 1.50-2.50 (10H, complex), 2.61-3.31 (3H, complex, 3^1-H and 2-H) and 4.15 p.p.m. (2H, q, J 7Hz, 5^1H); δ_C (25 MHz) 14.3 (C-6), 19.1 (C-6^1) 21.8, 24.2, 32.5, 32.9, 53.3 and 53.85 (C-3, -4, -5, -1^1, -2^1 and -3^1), 59.9 (C-5^1), 60.1 (C-2) and 173.6 p.p.m. (C-4^1); M/Z 199 (M^+) 184, 168, 167, 153 and 152. The picrolonate had m.p. 108°C (EtOH). [Found: C, 54.35; H, 6.26; N, 15.02%. $C_{21}H_{29}N_5O_7$ requires C, 54.42; H, 6.31; N, 15.11%]

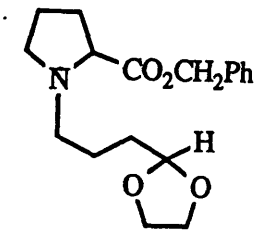
4-Bromobutanal ethylene acetal (125)



(125)

4-Bromobutanal (0.25g, 1.66 mmol), ethylene glycol (0.113g, 1.83 mmol, 1.1 equivalents) and p-toluenesulphonic acid (0.01g) in benzene (20 ml) were heated at reflux in a Dean and Stark apparatus for 2h. The solution was cooled, washed with 1M sodium hydroxide solution (1 x 20 ml) and with water (5 x 20 ml). The organic solution was dried, filtered, and concentrated to leave the acetal (125) (0.2g, 62%) as a clear oil after chromatography on silica [petroleum ether (40-60°C)/ether, 1:1]; R_F 0.50 (same solvent system); ν_{max} ($CHCl_3$) 2940, 1200 and 615 cm^{-1}; δ_H (90 MHz) 1.60-2.23 (4H, complex, 2-H and 3-H), 3.30-3.68 (4H, complex, 5-H and 6-H), 3.79-4.09 (2H, complex, 4-H) and 4.87 p.p.m. (1H, t, J 5Hz, 1-H; M/Z 196/194 (M^+), 115 (M^+-Br) [Found: M^+, 195.9924; $C_6H_{11}O_2Br$ requires M, 195.9923].

N(-3-Ethylene acetal carbonylpropyl)-proline benzyl ester (126)



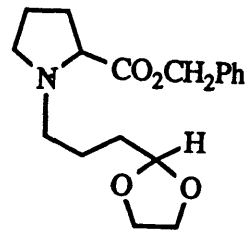
(126)

A solution of proline benzyl ester (0.41g, 2 mmol, 2 equivalents) in dry ether (5 ml) was added over 3h with stirring under argon to a solution of the acetal (125) (0.195g, 1 mmol) in dry ether (5 ml). The reaction mixture was heated at reflux for 18h, then cooled and poured into water (20 ml). The ethereal layer was separated and the aqueous layer was extracted with ether (3 x 50 ml). The organic extracts were combined, dried, filtered and concentrated to leave a brown oil, which was purified by chromatography on silica (chloroform/methanol/triethylamine, 90:5:1) to give the acetal (126) (0.28g, 44%) as a yellow oil,

R_F 0.85; ν_{max} ($CHCl_3$) 2950, 1740, 1670 and 1210 cm^{-1};

δ_H (90 MHz) 1.20-2.79 (8H, complex, H-3, -4, 3^1 and 2^1), 2.89-3.65 (5H, complex, H-2, -5 and 1^1), 3.75-4.11 (4H, complex, 5^1-H and 6^1H), 4.85 (1H, t, J 4Hz, 4^1-H), 5.18 (2H, s, 7-H) and 7.34 p.p.m. (5H, s, aromatic); M/Z 319 (M-1), 318, 184, 160 and 156.

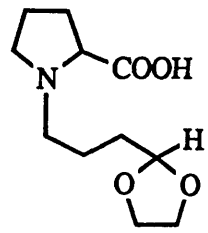
N-(3 Ethylene acetal carbonylpropyl)-proline benzyl ester (126)



(126)

The same procedure was used for the formation of acetal (126) except that 1 equivalent of D.B.U. replaced 1 equivalent of proline benzyl ester. The yield was 20%.

N-(3-Ethylene acetal carbonylpropyl)-proline (127)



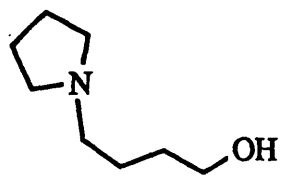
(127)

10% Palladium/charcoal catalyst (0.1g) was added to the amino acetal (126) (0.28g, 0.88 mmol) in ethanol (40 ml) and the mixture was hydrogenated at 5 atm. for 16h. The catalyst was removed by filtration through Celite and the solvent was removed under reduced pressure to leave the acid (127) as a green gum (0.19g 94%) which was used without further purification; R_F 0.0; ν_{max} ($CHCl_3$) 3385, 2970 and 1645 cm^{-1}; δ_H (90 MHz) 1.23-2.65 (8H, br, H-3, -4, -2^1 and 3^1), 2.93-3.76 (5H, br, H-2, -5 and 1^1), 3.77-4.20 (4H, br, 5^1-H and 6^1-H) and 4.90 p.p.m. (1H, broad t, J 5Hz, 4^1-H).

Attempted cyclisation of N-(3 ethylene acetal carbonylpropyl)-proline (127)

Phosphorus oxychloride (2 ml, 3.32g, 21.2 mmol, 12 equivalents) was added to (127) (0.3g, 1.79 mmol) and the mixture was heated at 100°C for 3 min. The solution was then rapidly cooled and ice-water (15 ml) was added followed by methanol (85 ml) and D.B.U. (10 ml). This mixture was stirred at room temperature for 48h, then poured into water (50 ml). The aqueous solution was extracted with ether (3 x 60 ml), dried, filtered, and the solvent was removed under reduced pressure to leave 10 mg of a brown oil which was a mixture of compounds by t.l.c.

N-(4-Hydroxybutyl)-pyrrolidine (130)

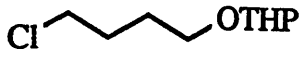


(130)

Phosphorus oxychloride (1 ml, 1.66g, 10.6 mmol, 12.77 equivalents) was added to N-(3-ethylene acetal carbonylpropyl)-proline (127) (0.19g, 0.83 mmol) and the resultant solution was heated at 100°C for 3 min.. The solution was then rapidly cooled, ice-water (10 ml) was added and the pH was adjusted to 6.5 using potassium carbonate. The mixture was stirred for 16h. The pH was then adjusted to 7 (potassium carbonate) and a solution of sodium borohydride (31 mg, 0.83 mmol, 1 equivalent in water (5 ml)) was added. The reaction mixture was stirred at room temperature for 24h, acidified with 2M hydrochloric acid and washed with methylene chloride (3 x 10 ml). The acidic solution was basified and saturated with potassium carbonate and extracted with methylene chloride (3 x 50 ml). The organic extracts were

dried, filtered and concentrated under reduced pressure to leave the amino-alcohol (130) (0.048g, 41%) as a yellow oil; R_F 0.34; ν_{max} ($CHCl_3$) 3590, 2970, 1590 and 1430 cm^{-1}; δ_H (90 MHz) 1.32-2.28 (8H, complex, 3-H, 4-H, 2^1-H and 3^1-H), 2.51-2.87 (6H, complex, 2-H, 5-H and 1^1-H) and 3.72 p.p.m. (2H, t, J 6Hz, 4^1-H); M/Z 143 (M^+) 126, 112 and 70 [Found: M^+, 143.1312, $C_8H_{17}NO$ requires M, 143.1310].

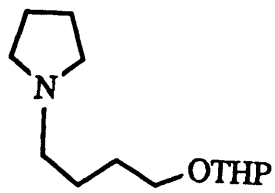
Tetrahydropyranyl ether of 4-chlorobutanol



p-Toluene sulphonic acid (53 mg, 0.28 mmol, 0.01 equivalents) was added to a solution of 4-chlorobutanol (3g, 0.028 mol) and 2,3-dihydro-4-pyran (4.94g, 0.059 mol, 2.13 equivalents) in dry methylene chloride (40 ml) with stirring and cooling to 0°C. The mixture was warmed to room temperature and stirred for 15h. The solution was then added to a mixture of ether (40 ml) and an aqueous layer comprising brine (40 ml), saturated sodium bicarbonate (40 ml) and water (80 ml). The organic layer was separated, washed with brine (2 x 80 ml), dried ($MgSO_4$-potassium carbonate), filtered and the solvent was removed under reduced pressure to leave a brown oil which was distilled (b.p. 72°C/0.5 mmHg) to give the THP protected alcohol (4.36g, 81%);

ν_{max} ($CHCl_3$) 2920, 2780, 1165 and 710 cm^{-1}; δ_H (90 MHz) 1.82 (4H, complex, H-8 and H-9), 1.97 (4H, complex, 2-H and 3-H), 3.41 (2H, t, J 7Hz, 1-H), 3.59 (2H, t, J 7Hz, 4-H), 3.85 (4H, complex, 7-H and 10-H), and 4.82 p.p.m. (1H, t, J 6Hz, 5-H); M/Z 194, 192, 156, 85, 38 and 36 [Found: M^+, 192.0916, $C_9H_{17}O_2{}^{35}Cl$ requires M 192.0917].

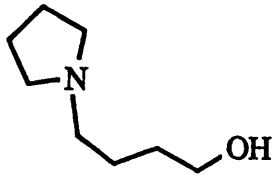
Tetrahydropyranyl ether of N-(4-hydroxybutyl) pyrrolidine



The tetrahydropyranyl protected 4-chlorobutanol (1g, 5.2 mmol), pyrrolidine (0.37g, 5.2 mmol, 1 equivalent), D.B.U. (0.871g, 5.72 mmol, 1.1 equivalents and sodium iodide (78 mg, 0.52 mmol, 0.1 equivalents) were heated together in dry DMF (30 ml) at 60°C for 5h. The mixture was cooled, poured into water (50 ml) and extracted with chloroform (3 x 50 ml). The organic extracts were washed with water (3 x 60 ml), dried, filtered and the solvent removed under reduced pressure to leave a brown oil. This was purified by chromatography on silica (chloroform/methanol/triethylamine, 90:9:1) to leave N-(4-Hydroxybutyl)-pyrrolidine tetrahydropyranyl derivative (0.75g, 64%) as a yellow oil;

R_F 0.35; ν_{max} 2950, 2810, 2785, 1165 and 950 cm^{-1}; δ_H (90MHz) 1.43-2.41 (12H, complex, 3-, 8^1-, 9^1-, 4-, 2^1- and 3^1-H), 2.58-3.40 (8H, complex, 2-, 5-, 1^1- and 4^1-H), 3.51-3.72 (4H, complex, 7^1- and 10^1-H) and 4.76 p.p.m. (1H, t, J 7Hz, 5^1-H). M/Z 227, 226, 142, 85 and 83 [Found M^+, 227.1886, $C_{13}H_{25}NO_2$ requires M, 227.1885].

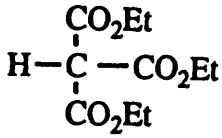
N-(4-Hydroxybutyl)pyrrolidine (130)



(130)

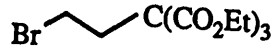
The tetrahydropyranyl protected N-(4-hydroxybutyl)pyrrolidine (0.35g, 1.54 mmol) was stirred at room temperature in 2M HCl for 16h. The solution was washed with methylene chloride and then basified and saturated with potassium carbonate. The basic solution was extracted with methylene chloride, dried, filtered, and the solvent was removed under reduced pressure to leave an oil (0.176g, 80%). All t.l.c. and spectroscopic data were identical with the sample previously prepared by a different route.

Triethyl methanetricarboxylate



This was prepared according to the method of Lund and Voigt,[77] yield 86% (lit.,[77] 88%); m.p. 27-28°C (lit.,[77] 28-29°C); b.p. 128°C/12 mmHg (lit.,[77] 130°C/10 mmHg); δ_H (90 MHz) 1.23 (9H, t), 4.15 (1H, s) and 4.22 (6H, q), identical to literature values.

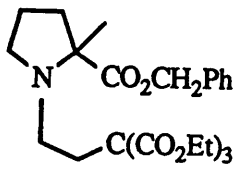
Triethyl 3-bromopropane-1,1,1-tricarboxylate (133)



(133)

This was prepared according to the method of Padgett et al.[30] yield 82% (lit.,[30] 91%); δ_H (90 MHz) 1.23 (9H, t), 2.53 (2H, multiplet), 3.42 (2H, multiplet) and 4.17 (6H, q), identical to literature values [Found M^+, 339.0443, 341.0426; $C_{12}H_{20}O_6Br$ requires M, 339.0449, 341.0427].

N-[3,3,3-Tris(ethoxycarbonyl)n-propyl]-2-methylproline benzyl ester (134)

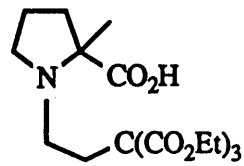


(134)

A mixture of 2-methylproline benzyl ester (120) (1g, 5.13 mmol) and potassium carbonate (2.1g, 15 mmol) in a solution of benzene/DMF (1:1, 10 ml) was stirred and heated at 80°C under argon. A solution of the bromotriester (133) (2.2g, 7 mmol, 1.36 equivalents in benzene/DMF (1:1, 2 ml) was added over 30 min. and the mixture was heated at 80°C for 20 h. T.l.c. showed no starting material remained after this time. The mixture was cooled, diluted to 40 ml with benzene and the inorganic residues were removed by filtering through Celite. The DMF was removed by washing the organic solution with water (6 x 20 ml). The organic phase was dried, filtered and the solvent removed under reduced pressure to leave the amino-ester (134) (1.7g, 69%) as a yellow

oil after chromatography on alumina (methylene chloride/chloroform); R_F 0.69; ν_{max} (thin film) 2990, 1730, 1720 and 1380 cm^{-1}; δ_H (90 MHz) 1.24 (9H, t, J 6Hz, 3 CH_3 of esters), 1.39 (3H, s, 6-H), 1.60-3.10 (8H, complex), 3.35-3.90 (2H, complex, 2^1-H), 4.21 (6H, q, J 6Hz, 3 CH_2 of esters), 5.15 (2H, s, 8-H) and 7.30 p.p.m. (5H, s, aromatic); M/Z 434, 390, 388, 345 and 344.

N-[3,3,3-Tris(ethoxycarbonyl)]-n-propyl-2-methylproline (135)



(135)

The tri-ester (134) (0.54g, 1.13 mmol) was dissolved in absolute ethanol (25 ml and 0.1g of 10% Pd/C catalyst was added to this mixture. The solution was then hydrogenated at 7 atm. for 18h. The catalyst was then filtered off and the solvent was removed under reduced pressure to leave the amino-acid derivative (135) after trituration with ether/hexane (4:1);
R_F 0.0; ν_{max} (thin film) 3390, 2980, 1725 and 1650 cm^{-1}; δ_H (90 MHz) 1.27 (9H, t, J 8Hz, 3 methyls of ethyl esters), 1.40 (3H, s, 6-H), 1.50-3.07 (8H, br, 3-, 4-, 5- and 2'-H), 3.30-4.01 (2H, br, 2'-H) and 4.25 p.p.m. (6H, q, J 8Hz, 3 methylenes of ethyl esters).

Attempted deethoxycarbonylation of

N-[3,3,3-tris(ethoxycarbonyl)-n-propyl]-2-methylproline (135)

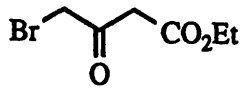
The amino-ester (135) (0.5g, 1.01 mmol) was added to a stirred solution of sodium benzylate (0.2g, 1.54 mmol, 1.5 equivalents) in anhydrous D.M.S.O. and the solution was stirred at room temperature for 15 min. The solution was then extracted with benzene (20 ml) and extracted with 1M phosphoric acid (3 x 30 ml). The acidic aqueous phase was made alkaline with potassium carbonate and extracted with benzene (3 x 50 ml). The combined organic extracts were washed with brine, dried and the solvent removed under reduced pressure to leave a brown oil which gave many spots by t.l.c.. Varying the temperature and length of time of the reaction gave similar results.

Attempted deethoxycarbonylation of

N-[3,3,3-Tris(ethoxycarbonyl)-n-propyl]-2-methylproline (135)

Sodium (0.17g, 7.4 mmol) was dissolved in super-dry ethanol (10 ml). The amino-ester (135) (1g, 2.6 mol) was added to this solution and the mixture was stirred at room temperature for 10 min.. The solution was cooled to 0°C and 2M HCl was added (200 mol % based on sodium used). The solution was concentrated to leave a thick oil which was triturated with hot t-butanol. The sodium chloride was filtered off and the solvent was evaporated to leave a brown oil (0.62g) which gave many spots by t.l.c.. Variations in the reaction time and temperature gave similar results.

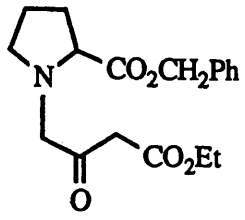
γ-Bromoethyl acetoacetate (137)



(137)

This compound was prepared according to the method of Svendsen and Boll,[63], yield 83%; (lit.[63] 85%); b.p. 107-112°C, 12 mmHg, (lit.[63] 110-120°C, 12 mmHg); ν_{max} (thin film) 2900, 1760, 1745 and 610 cm^{-1}; δ_H (90 MHz) 1.25 (3H, t, J 7Hz, methyl of ester), 3.71 (2H, s, γ-H), 4.07 (2H, s, α-H) and 4.23 p.p.m. (2H, q, J 7Hz, methylene of ester).

N-(2-Oxo-3-ethoxycarbonylpropyl)proline benzyl ester (138)

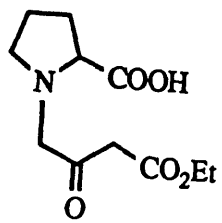


(138)

The bromoketoester (137) (8.15g, 3.9 mmol) was added, dropwise under nitrogen, to a solution of proline benzyl ester (108a) (5.1g, 2.49 mmol) and triethylamine (3.9g, 3.9 mmol) in sodium-dried ether (60 ml). The resultant solution was stirred at room temperature for 2h until t.l.c. showed no starting material present. The reaction mixture was poured into water (120 ml) and the aqueous layer was extracted with ether (3 x 50 ml). The organic layers were combined, washed with aqueous sodium metabisulphite (3 x 50 ml of 1M) and extracted with 2M HCl (4 x 100 ml). The acidic extracts were washed with ether, then basified to pH 10 with ice-cooling using conc. ammonia. The basic solution was extracted with ether (3 x 250 ml), dried, filtered and the solvent removed under reduced pressure to leave the

N-substituted proline benzyl ester (138) (5.85g, 71%) as a dark red oil; R_F 0.78; ν_{max} (thin film) 2980, 1715 (broad), 1670 and 1570 cm-1; δ_H (90 MHz) 1.19 (3H, t, J 7Hz, 6^1-H), 1.60-2.20 (4H, complex, 3-H and 4-H), 2.30-3.27 (3H, complex, 2-H and 5-H), 3.36-3.62 (4H, complex, 1^1-H and 3^1-H), 4.09 (2H, q, J 7Hz, 5^1-H), 5.09 (2H, s, 7-H) and 7.30 p.p.m. (5H, s, aromatic).

N-(2-Oxo-3-ethoxycarbonylpropyl) proline (139)



(139)

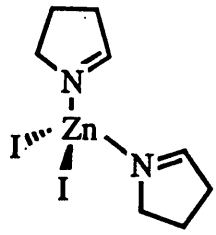
Palladium/charcoal catalyst (2g, 10%) was added to a solution of the ketoester (9g, 27 mmol) in ethanol (200 ml) and the mixture was hydrogenated at 3 atm. for 10h. The catalyst was filtered off through Celite and the solvent was evaporated to leave an orange solid which was triturated with ether/hexane (4:1) to give the proline ester (139) (5.67g, 86%), m.p. 128-130°C, R_F 0.0; ν_{max} ($CHCl_3$) 3395, 2975, 1735 and 1645 cm^{-1}; δ_H (90 MHz) 1.32 (3H, br t, J 7Hz, 6^1-H), 1.71-2.34 (4H, br, 3-H and 4-H), 3.40-3.91 (3H, br, 2-H and 5-H) and 3.90-4.51 p.p.m. (6H, br, 1^1-, 3^1- and 5^1-H); mass spectrum gave only low molecular weight fragments; [Found: C, 54.32; H, 7.08; N, 5.73%; $C_{11}H_{17}NO_5$ requires C, 54.31; H, 7.04; N, 5.75%].

Attempted cyclisation of N-(2-Oxo-3-ethoxycarbonylpropyl) proline (139)

Phosphorus oxychloride (3.9 ml, 6.47g, 41 mmol, 10 equivalents) was added to the proline derivative (139) (1g, 4.1 mmol) and the solution was heated at 100°C for 3 min. The reaction mixture was rapidly cooled, ice-water (60 ml) was added, and the pH was adjusted to 6 using potassium carbonate. The solution was stirred at room temperature for 16h, then saturated with potassium carbonate and extracted with ether (3 x 50 ml). The ether extracts were washed with water (3 x 100 ml), dried, filtered and concentrated to leave a brown oil (0.12g) which was a mixture of many compounds by t.l.c. Variations in the reaction temperature and pH showed similar results.

6.5 Experimental to Chapter 5

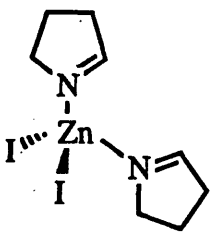
Formation of zinc iodide-1-pyrroline complex (147)



(147)

Cyclopentadiene (2.68g, 40 mmol, 4 equivalents) and zinc iodide (2.87g, 10 mmol, 1 equivalent) were added to a solution of 1-pyrroline (57) (0.69g, 10 mmol) in ether (30 ml). This mixture was stirred under argon at room temperature for 48h. The yellow precipitate was filtered off and washed with ether to leave the zinc iodide 1-pyrroline complex (147) as a grey solid (1.1g), m.p. > 300°C; ν_{max} ($CHCl_3$) 2980, 1630, 1330, 960, 930 and 420 cm^{-1}; δ_H (90 MHz) 1.90-2.67 (2H, complex, 4-H), 2.78-3.21 (2H, complex, 3-H), 3.92-4.32 (2H, complex, 5-H) and 8.29 p.p.m. (1H, t, J 6Hz, 2-H); δ_C (25 MHz) [$(CD_3)_2CO$] 20.81 (C-4), 37.5 (C-3), 59.9 (C-5) and 178.5 p.p.m. (C-2) [Found: C, 20.93; H, 2.78; N, 5.87; I, 55.56; Zn, 14.86%. $C_8H_{14}N_2I_2Zn$ requires C, 21.00; H, 3.09; N, 6.12; I, 55.49 and Zn, 14.29%]. (See p 150)

^{1}H N.m.r. experiments with the 1-pyrroline complex (147)



(147)

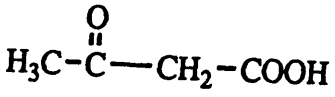
Diethylamine (3.7μl, 2.56 mg, 0.035 mmol, 0.5 equivalents) was added to a solution of the 1-pyrroline complex (147) (0.032g, 0.07 mmol) in deuteriochloroform (0.5 ml). ^{1}H N.m.r. spectroscopy showed the ratio of the complex:free pyrroline as 5:1. Addition of a further 3.7μl of diethylamine decreased this ratio to 4.5:1 and the spectrum showed some unreacted diethylamine. On addition of approximately 15 equivalents of diethylamine, the signals due to the complexed pyrroline disappeared. Similar results were obtained with ethylene diamine, ethylamine and triethylamine.

Attempted cycloaddition of the 1-pyrroline complex (147) and cyclopentadiene

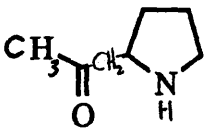
The pyrroline complex (147) (0.2g, 0.44 mmol) was dissolved in ether/acetone (5 ml of 4:1) and cyclopentadiene (0.14g, 2.12 mmol, 5 equivalents) was added to this solution. This mixture was stirred for 48h at room temperature under argon. After this time, t.l.c. and ^1H n.m.r. spectroscopy indicated only starting material was present. The experiment was repeated with a range of reaction temperatures and times but no product could be detected by t.l.c. or ^1H n.m.r. spectroscopy. A range of solvents, temperatures and time of reaction had the same result.

Acetoacetic Acid

This compound was prepared according to the method of Cragg et al. [78]



Norhygrine (148)



(148)

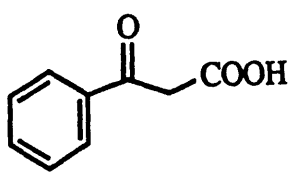
The complex (147) (0.97g, 2.13 mmol), methanol (50 ml) and 1 M phosphate buffer (pH 7, 4 ml) were added to a solution of acetoacetic acid (0.5g, 4.9 mmol, 2.3 equivalents) in water (10 ml). The pH of this solution was adjusted to 7 using 1M potassium hydroxide solution and the mixture was stirred at room temperature for 60h. The solution was acidified with 2M hydrochloric acid and heated at reflux for 3h. The cooled solution was washed with ether (3 x 50 ml) and basified with conc. ammonia solution and potassium carbonate. The basic solution was extracted with chloroform (3 x 80 ml). The combined organic extracts were dried, filtered, and concentrated under reduced pressure to leave

norhygrine (0.62g, 85%), R_F 0.5 (n-BuOH/acetic acid, 4:1, saturated with water); ν_{max} ($CHCl_3$) 2910, 1705 and 1530 cm^{-1}; δ_H (90 MHz) 1.5-4.5 (10H, complex), 2.15 p.p.m. (3H, s, $\underline{CH_3}$-).

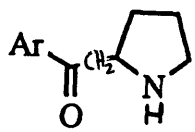
The semicarbazone hydrochloride had m.p. 203-205°C (lit.,[71] 204-206°C).

Benzoylacetic acid

This compound was prepared according to the method of Cragg et al.,[78] yield = 70%; ν_{max} (film) 3090, 2700, 1720 and 1700 cm^{-1}; δ_H (90 MHz) 5.10 (2H, s), 7.25-7.55 (3H, m) and 7.70-8.00 p.p.m. (2H, m).



2-Phenacylpyrrolidine (150)

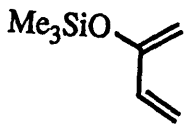


(150)

The 1-pyrroline complex (147) (0.7g, 1.53 mmol), methanol (50 ml) and 1M phosphate buffer (pH 7, 4 ml) were added to a solution of benzoylacetic acid (0.7g, 3.75 mmol, 2.46 equivalents) in water (10 ml). The pH of the solution was adjusted to 7 with 1M potassium hydroxide solution and the mixture was stirred at room temperature for 60h. The solution was then acidified with 2M hydrochloric acid to pH 2 and heated at reflux for 3h. The cooled solution was washed with ether (3 x 50 ml), basified with conc. ammonia solution, and saturated with potassium carbonate. The basic solution was extracted with chloroform (3 x 100 ml), and then continuously extracted with chloroform for 16h. The combined organic extracts were dried, filtered, and concentrated to leave an

oil which was purified by column chromatography on alumina (chloroform/methylene chloride, 50:50) to leave 2-phenacylpyrrolidine (150) (0.5g, 88%); ν_{max} (film) 3350, 1675, 1610 and 1590 cm^{-1}; δ_H (90 MHz) 1.18-2.10 (4H, complex), 2.78-3.10 (2H, complex), 2.95-3.20 (2H, m), 3.20 (1H, s, NH), 3.55 (1H, m), 7.10-7.55 (3H, m) and 7.60-8.20 p.p.m. (2H, m); M/Z 189 (M^+), 121, 84 and 75 [Found: M^+ 189.11489, $C_{12}H_{15}NO$ requires 189.11538]. All of these data are in accordance with literature values.[78]

2-Trimethylsilyloxy-1-buta-1,3-diene (151)[79]



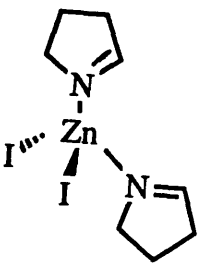
(151)

A solution of triethylamine (55.5 ml, 0.40 mol) in dry DMF (200 ml) was stirred under nitrogen at 80-90°C. To this was added, dropwise and simultaneously from 2 pressure equilibrated dropping funnels, a solution of freshly distilled methyl vinyl ketone (25.30g, 0.36 mol) in DMF (25 ml), and a solution of trimethylsilyl chloride (51.5 ml, 0.40 mol) in DMF (25 ml) and the resulting mixture was stirred overnight at this temperature. The reaction mixture was cooled, filtered twice through Celite and poured into n-pentane (300 ml). The organic solution was washed with 5% aqueous sodium hydrogen carbonate solution (3 x 100 ml) and the aqueous layer was extracted with n-pentane (2 x 100 ml). The combined organic extracts were dried, filtered, and the pentane was removed by fractional distillation at atmospheric pressure. The

brown residue was distilled (33-37°C, 12 mmHg) to afford 17.96g (35%) of pure diene (151); δ_H (90 MHz) 0.42 (9H, s), 4.45 (2H, br s), 5.10 (1H, m), 5.5 (m,1), 6.3 p.p.m. (m,1).

This was in accordance with literature values.[79]

1-Pyrroline zinc iodide complex (147)



(147)

γ-Aminobutanal diethylacetal (2.34g, 14.5 mmol) was dissolved in 2M hydrochloric acid (50 ml) and ether (50 ml) was added. The solution was stirred at 0°C for 20 min, then basified with potassium carbonate. The aqueous layer was extracted with ether (at 0°C) (3 x 50 ml). The combined organic extracts were cooled to 0°C, dried, filtered, and zinc iodide (4.17g, 14.5 mmol, 1 equivalent) was added. This mixture was stirred under argon for 30 min. The precipitate was filtered off and washed with ether and hexane to leave the complex (147) (3.31g, 50%) as a grey powder. All analytical and spectral data were identical with those of a sample prepared by a different route.

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