Towards the Total Synthesis of Asperparaline C

Peter John Crick

Thesis submitted to the University of Birmingham
for the degree of Doctor of Philosophy

School of Chemistry
University of Birmingham

September 2011

Abstract

The asperparalines belong to a large family of prenylated indole alkaloids possessing a characteristic bicyclo[2.2.2]diazaoctane core structure. More than 70 examples are known to have been isolated from various diverse sources. Chapter 1 discusses the isolation and structure of these compounds along with an introduction to their biosynthesis and biological activities.

A combination of unique structural features and intriguing biological profiles has inspired a large body of work regarding the chemical synthesis of this alkaloid family. Chapter 2 gives an overview of the most important strategies for the construction of the central bicyclo[2.2.2]diazaoctane and presents these in the context of several total syntheses.

Chapter 3 details an approach to the bicyclo[2.2.2]diazaoctane core using a free radical cascade comprised of a 1,6-hydrogen atom translocation followed by *6-exo-trig* and *5-exo-trig* cyclisations. An initial model system is presented consisting of a series of DKPs synthesised from a propargylated proline derived from a modification of Seebach's procedure for the self-reproduction of chirality. Addition of a sulfur radical to the acetylene triggers the desired reaction in good to excellent yield and favours the asperparaline stereochemistry.

In Chapter 4 the application of this strategy to the synthesis of an advanced asperparaline core structure is presented. While construction of the key cyclisation intermediate proved difficult, the radical cascade proceeds in modest yield to furnish a bicyclo[2.2.2]diazaoctane possessing the key structural features of the asperparalines.

Acknowledgments

First and foremost I would like to thank Professor Nigel Simpkins for giving me the opportunity to carry out the work reported here. I am also very grateful to Dr Adrian Highton (my CASE supervisor) for all of his helpful input and for organising my time in the AstraZeneca laboratories in Charnwood.

None of this work would be possible without the financial support generously provided by the University of Birmingham and AstraZeneca. The analytical facilities in the School of Chemistry were also vital and I would particularly like to mention Dr Neil Spencer for help with running NMR experiments and numerous helpful discussions.

During my time in the Simpkins group I've had the pleasure of working with some great chemists and friends all of whom are appreciated. In particular I've learnt a lot from Fred, Ilias and Bick. Of course I've had a lot of fun with everyone in the group – Mike, Jimm, Valerie, Mark, Cynthia and Damian.

Thanks also to my other friends and family for all fun I've had, in particular Ann, John, Martin, Duncan, Giorgio, Nats, Tom, Bene, Freeman and Rachel. Also thanks to everyone who I've lived with over the past 4 years – Haf, Lorraine, Polly, Vicky and Susanna.

Finally, thanks to Jen – I know she's had to put up with a lot, especially over the final few months – and I appreciate that a lot.

Table of Contents

Chapter 1, Introduction

Chapter 2, Previous Synthetic Work

Chapter 3, Model System Studies

Chapter 4, Towards Asperparaline C

Chapter 5, Experimental

Glossary of Abbreviations

1,2-DCE	1,2-dichloroethane
1D	one dimensional
2D	two dimensional
4-DMAP	4-dimethylaminopyridine
9-BBN	9-borabicyclo[3.3.1]nonane
AIBN	azobis*iso*butyronitrile
Boc	*tert*-butoxycarbonyl
BOP	benzotriazole-1-yl-oxy-tris(dimethylamino)-phosphonium hexafluorophosphate
Bz	benzoyl
CaM	calmodulin
Cbz	carboxybenzyl
COSY	correlation spectroscopy
DABCO	1,4-diazabicyclo[2.2.2]octane
DBU	1,8-diazabicycloundec-7-ene
DCB	*ortho*-dichlorobenzene
DCC	*N*,*N'*-dicyclohexylcarbodiimide
DDQ	2,3-dichloro-5,6-dicyano-1,4-benzoquinone
DIBAL	di*iso*butylaluminium hydride
DIPEA	di*iso*propylethylamine (Hünig's base)
DKP	diketopiperazine
DMF	dimethylformamide
DMP	Dess-Martin periodinane

DMSO	dimethylsulfoxide
ESI	electrospray ionization mass spectrometry
ESR	electron spin resonance spectroscopy
FAB	fast atom bombardment
Fm	fluorenylmethyl
Fmoc	fluorenylmethyloxycarbonyl
FT	Fourier transform
GI_{50}	concentration of compound required for 50% growth inhibition
GOESY	gradient enhanced nuclear Overhauser effect spectroscopy
HATU	*O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N'*,*N'*-tetramethyluronium hexafluorophosphate
HMBC	heteronuclear multiple-bond correlation spectroscopy
HMDS	hexamethyldisilazane/bis(trimethylsilyl)amine
HOAt	1-hydroxy-7-azabenzotriazole
HOMO	highest occupied molecular orbital
HPLC	high pressure (or performance) liquid chromatography
HSQC	heteronuclear single-quantum correlation spectroscopy
IC_{50}	concentration of compound required for 50% inhibition of activity
IMDA	intramolecular Diels-Alder reaction
IR	infrared spectroscopy
J	coupling constant (in nuclear magnetic resonance spectroscopy)
kcal	kilocalorie
LC-MS	liquid chromatography-mass spectrometry
LD_{50}	dose of compound required to kill 50% of subjects
LDA	lithium di*iso*propylamide

LUMO	lowest unoccupied molecular orbital
mCPBA	*meta*-chloroperoxybenzoic acid
MIC	minimum inhibitory concentration
MKP	monoketopiperazine
MOM	methoxymethyl
Ms	methylsulfonyl
MS	mass spectrometry
MTM	methylthiomethyl
nOe	nuclear Overhauser effect
NOESY	nuclear Overhauser effect spectroscopy
NMM	*N*-methylmorpholine
NMR	nuclear magnetic resonance spectroscopy
OTf	trifluoromethanesulfonate (triflate)
Piv	pivaloyl
PMB	*para*-methoxybenzyl
ppm	parts per million
PTLC	preparative thin layer chromatography
Rose Bengal	4,5,6,7-tetrachloro-2',4',5',7'-tetraiodofluorescein disodium salt
RNA	ribonucleic acid
RSE	radical stabilisation enthalpy
SAR	structure-activity relationship
SEM	2-(trimethylsilyl)ethoxymethyl
SOMO	singly occupied molecular orbital
TBAF	tetra-*n*-butylammonium fluoride

TBAI	tetra-*n*-butylammonium iodide
TBAT	tetra-*n*-butylammonium difluorotriphenylsilicate
TBDPS	*tert*-butyldiphenylsilyl
TBS	*tert*-butyldimethylsilyl
TFA	trifluoroacetic acid
THF	tetrahydrofuran
TKP	triketopiperazine
TLC	thin layer chromatography
TMM	trimethylenemethane
TMP	2,2,6,6-tetramethylpiperidine
TMS	trimethylsilyl
UV	ultraviolet spectroscopy

Chapter 1

Introduction

1.1 The Prenylated Indole Alkaloid Family

A large and diverse family of natural products classified as prenylated indole alkaloids are characterised by a central 2,5-diketopiperazine (DKP) core structure or a partially reduced derivative (monoketopiperazine, MKP). This heterocycle is often present as part of a bridged bicyclo[2.2.2]diazaoctane motif and may possess various substituents as outlined in Figure 1.1.

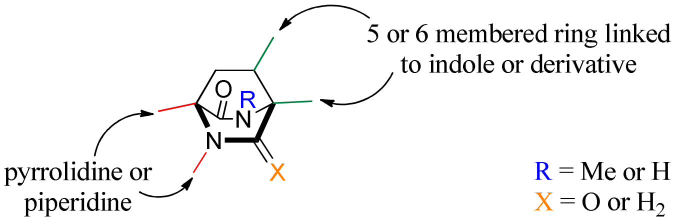
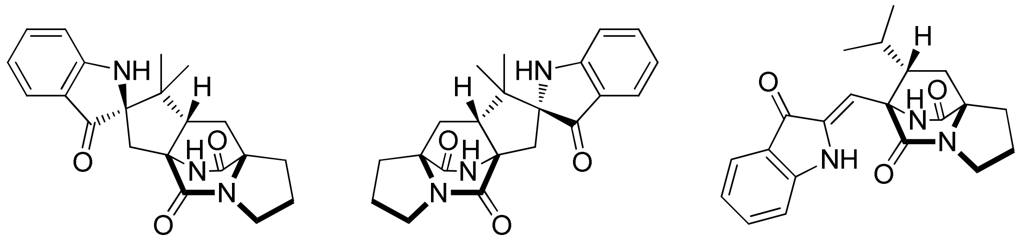


Figure 1.1

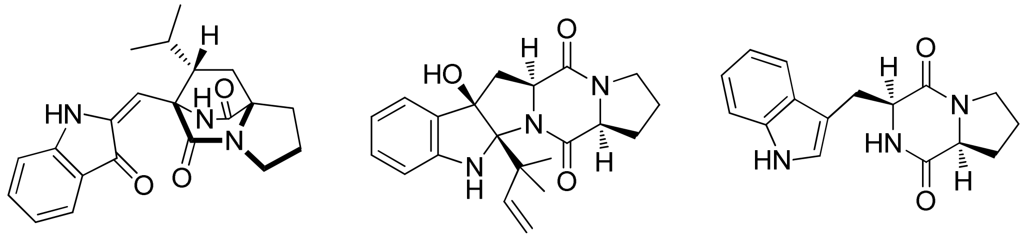
In total, the structures of more than 70 members of this family have been reported leading to a number of total syntheses and some detailed investigations of their biological activities.

1.2 Isolation and Structure Elucidation

The first report detailing members of this family dates from an initial communication in 1969 when Birch and Wright reported the isolation of brevianamides A-E as a mixture of pigments from *Penicillium brevi-compactum* (Figure 1.2).[1] Elemental analysis of brevianamide A (**1**) revealed an empirical formula of $C_{21}H_{23}N_3O_3$. An indoxyl chromophore along with two adjacent but non-equivalent methyl groups suggested a *spiro* fused ring junction. No double bonds were observed apart from those in the indoxyl portion of the molecule. Two peaks in the infrared (IR) spectrum at 1670 cm^{-1} and 1690 cm^{-1} along with further studies of **1** suggested the presence of a DKP. However, no evidence was found for a proton alpha to a carbonyl and amide nitrogen suggesting a bridged bicyclo[2.2.2]diazaoctane structure. Further spectroscopic evidence along with proposed biosynthetic routes led the authors to propose the presence of a pyrrolidine ring derived from proline. This was confirmed by feeding experiments incorporating ^{14}C labelled proline.



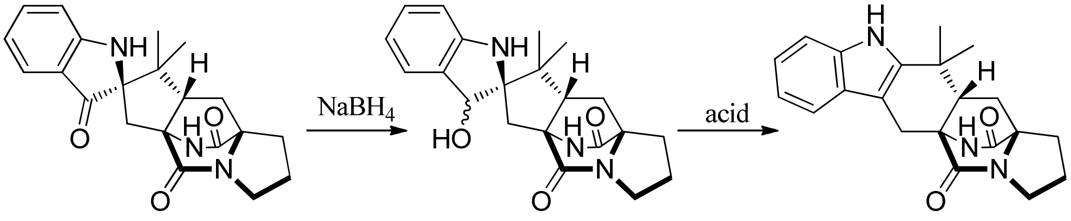
1 brevianamide A **2** brevianamide B **3** brevianamide C



4 brevianamide D **5** brevianamide E **6** brevianamide F

Figure 1.2

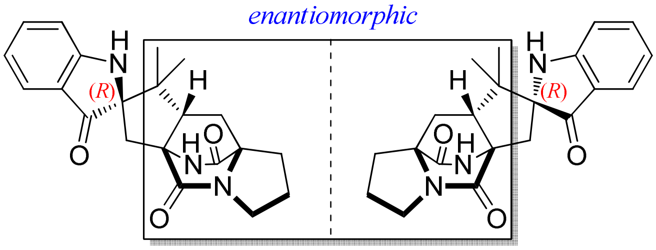
A second paper in 1970 provided a more complete analysis of both brevianamides A (**1**) and E (**5**) along with further speculation of their biosynthesis (*vide infra*).[2] Further proof of the *spiro*-indoxyl moiety was obtained by reduction of **1** with sodium borohydride followed by treatment with acid to give the 2,3-disubstituted indole deoxybrevianamide A (**8**) in an analogous manner to a previously reported transformation (Scheme 1.1).[3] This presumably proceeds through the hydroxyindoline **7**, although this compound was never isolated.



1 brevianamide A **7** **8** deoxybrevianamide A

Scheme 1.1

The structures of brevianamides B (**2**), C (**3**), D (**4**) and F (**6**) were reported in 1972 using broadly similar analytical techniques.[4] Brevianamide B possesses the same *spiro*-indoxyl motif as brevianamide A and can be converted into the latter by a sequence involving reduction, dehydration, re-oxidation and rearrangement. This suggested brevianamides A and B are stereoisomers differing only by the configuration of the *spiro*-centre. However, more detailed analysis showed that the absolute configuration of the *spiro* centres are the same (*R*) and brevianamides A and B are, in fact, enantiomorphic with respect to the DKP core (Figure 1.3).



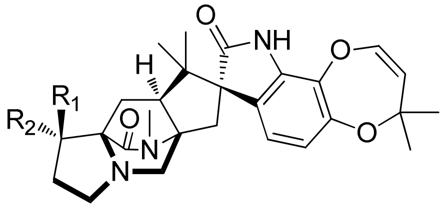
1 (+)-brevianamide A **2** (+)-brevianamide B

Figure 1.3

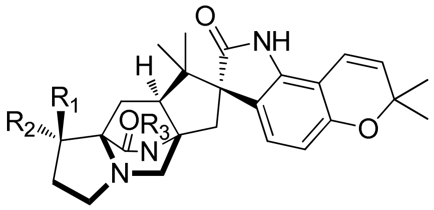
In a similar way, brevianamides C and D differ only by their configuration around the double bond connecting the DKP to the indole. Interestingly, brevianamides C and D were not isolated when *Penicillium brevi-compactum* was grown in the dark, suggesting that they are actually photochemical artefacts of isolation.[4] More recently a number of other brevianamides have been described, although none possessing the bridged DKP core.[5]

In 1981, a similar but more structurally complex compound was isolated from *Penicillium paraherquei* by the group of Yamazaki and named paraherquamide (now known as paraherquamide A, **9**, Table 1.1).[6] The IR spectrum for this compound indicated two carbonyl groups, but only one of them was characteristic of a DKP amide implying a different core structure. The ^{1}H NMR spectrum showed doublets at δ 4.87 and 6.30 corresponding to an enol ether functional group. These signals were replaced by two upfield doubled doublets at δ 2.10 and 4.28 after catalytic hydrogenation using palladium on carbon, presumably corresponding to a saturated ether motif.

To determine the structure, paraherquamide A was recrystallised from ethyl acetate. Solving the crystal structure revealed a *spiro*-oxindole ring system in contrast to the *spiro*-indoxyl found in the brevianamides along with a partially reduced central DKP. The proton NMR signals that had shifted after hydrogenation turned out to be a double bond in a dioxygenated seven-membered ring. Another marked contrast to the brevianamides is the presence of substitution on the pyrrolidine ring – in this case a methyl and hydroxyl group. It is also important to note the *syn* relationship between the proline residue and the carbon possessing the *gem* dimethyl at the ring junction of the reduced DKP. This is in contrast to the *anti* configuration of the brevianamides.



paraherquamide	R$_1$	R$_2$
9, A (VM29919)	OH	Me
10, B	H	H
11, E (VM54159)	H	Me

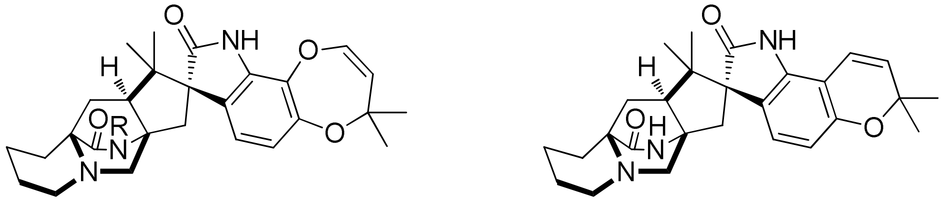


paraherquamide	R$_1$	R$_2$	R$_3$
12, F (VM55594)	H	Me	Me
13, G (VM54158)	OH	Me	Me
14, VM55595	H	Me	H

Table 1.1

More recently, a number of other members of this family have been reported.[7] Paraherquamide B (**10**) possesses the same basic structure as A but without decoration on the proline residue while paraherquamide E (**11**) lacks the hydroxyl group of **9**. Other examples incorporate a pyran ring in place of the unusual seven-membered heterocycle (e.g. paraherquamides F and G, **12** and **13** along with VM55595, **14**). As well as those shown above, a number of other members of the paraherquamide family have been isolated from various sources, including *Aspergillus japonicus* and *Penicillium fellutanum*.[8]

At around the same time as the initial reports of the paraherquamides, three compounds named the marcfortines were isolated (Figure 1.4).[9] They were found in extracts from *Penicillium roqueforti* – a fungus used in the production of a number of blue cheeses. The structure of marcfortine A was determined both by NMR analysis and crystallography. This showed a structure very similar to the paraherquamides. In fact, the only structural difference between paraherquamide B (**10**) and marcfortine A (**15**) is a single CH_2 moiety – the marcfortines all possess a piperidine ring in place of the proline-like pyrrolidine found in the paraherquamides. Further studies showed that marcfortine B (**16**) differs only by incorporating a secondary rather than tertiary amide in the monoketopiperazine ring while marcfortine C (**17**) has a slightly simpler structure with a pyran in place of the seven-membered ring.[10]



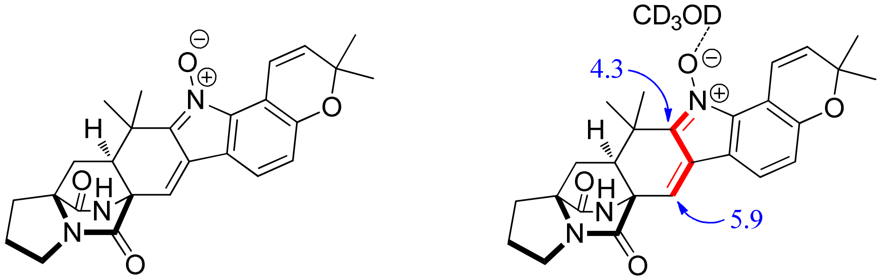
15 marcfortine A (R = Me)
16 marcfortine B (R = H)

17 marcfortine C

Figure 1.4

Interest in this family of compounds grew rapidly after a patent was filed in 2000 describing the isolation a novel compound named avrainvillamide or CJ-17,655 (**18**, Figure 1.5) from an *Aspergillus* species designated CNC358.[11] The following year the group of Sugie at the Pfizer laboratories reported the same compound, this time from fermentation of *Aspergillus ochraceus* CL41582.[12]

Various 2D NMR techniques were employed to determine the empirical formula of avrainvillamide, but the proposed molecular structure differed from that expected from mass spectral analysis by one oxygen atom. Reasoning that the missing oxygen could be attached to one of the three nitrogens the authors carried out more detailed studies. Grigor'ev and colleagues have previously reported a method for the determination of the structure of cyclic nitrones by analysing the ^{13}C NMR spectra in different solvents.[13] Solvents that are capable of forming hydrogen bonds with the *N*-oxide group cause the signal for the nitrone carbon to move downfield. This can be exploited by comparing the chemical shifts in chloroform-*d* and methanol-*d₄*, expressed as δ_C (CD_3OD) - δ_C ($CDCl_3$). A difference of 3.5-7.5 ppm is considered enough to determine the position of the oxygen. Figure 1.5 shows that in the case of avrainvillamide the difference for the nitrone carbon is 4.3 ppm, leading Sugie *et al.* to assign the missing oxygen as being on the indole nitrogen.



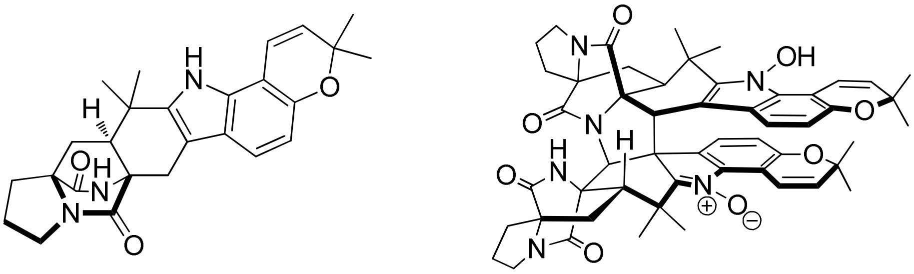
18 avrainvillamide
(CM-17,665)

values indicate chemical
shift difference in ppm
δ_C (CD_3OD) - δ_C ($CDCl_3$)

Figure 1.5

Soon after the isolation of avrainvillamide, the related compounds stephacidins A (**19**) and B (**20**) were discovered from fermentation of *Aspergillus ochraceus* WC76466 by researchers at Bristol-Myers Squibb (Figure 1.6).[14]

The UV spectrum of stephacidin A (**19**) suggested the presence of a highly conjugated aromatic moiety, supported by FAB-MS (fast atom bombardment mass spectrometry) analysis (showing 14 degrees of unsaturation) as well as ten signals in the ^{13}C NMR spectrum between δ 148.1 and 104.0 (showing five double bonds). A number of signals in the IR spectrum were characteristic of an amide or lactam, again supported by two carbonyl signals in the ^{13}C NMR with shifts at δ 174.0 and 169.3. The connectivity of all of these functional groups and the overall structure of the molecule was determined by detailed analysis using a variety of 2D-NMR studies. The molecular mass of stephacidin B (**20**) indicated it was approximately a dimer of stephacidin A (**19**). After some difficulties with solvent selection, the authors were able to obtain well resolved NMR spectra by using a 1:1 mixture of DMSO-*d*$_6$ and acetonitrile-*d*$_3$ but were still unable to determine the nature of the connectivity between the two monomers. In the end the structure was solved unambiguously by X-ray analysis of crystals grown from acetonitrile.

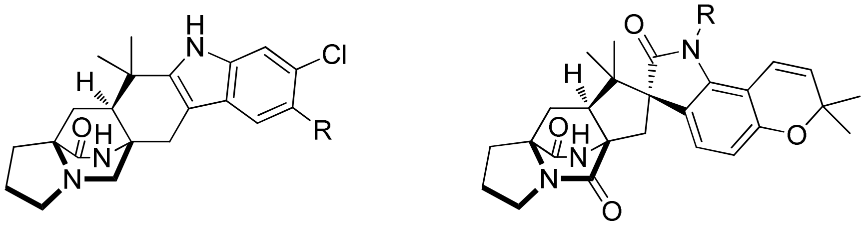


19 stephacidin A **20** stephacidin B

Figure 1.6

Avrainvillamide and both stephacidins can all be isolated from the same fungal species and are closely related to one another. Stephacidin A can be oxidised to form avrainvillamide which, in turn, dimerises to form stephacidin B. Stephacidin B is of particular interest as one of the most structurally complex alkaloids known – possessing 15 rings and 9 asymmetric centres.[15]

In total, more than 70 members of this family are known, with reports of related compounds appearing frequently. Table 1.2 shows another two classes, the malbrancheamides and notoamides. Malbrancheamide (**21**) was first reported in 2006 with malbrancheamide B (**22**) isolated two years later.[16] They are unique in incorporating either one or two chlorine atoms on the indole part of the molecule, along with a partially reduced DKP core. Notoamides A and B (**23** and **24**) are a good example of the close relationship between various family members. They possess similar structural features to some of the paraherquamides but with a DKP core.[17]



malbrancheamide	**R**
21, –	Cl
22, B	H

notoamide	**R**
23, A	OH
24, B	H

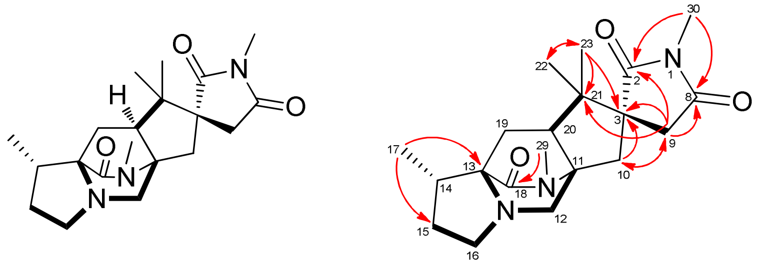
Table 1.2

The asperparalines are a sub-family of natural products related to those described above. While they have the same bridged core structure (present as a MKP) they differ significantly by incorporating a *spiro*-succinimide in place of the more common indole derivatives. Despite the apparent differences, the asperparalines share a common biological precursor with the paraherquamides (*vide infra*).

The first compound isolated was asperparaline A (**25**) in two reports published in 1997. The group of Hayashi identified the fungal strain *Aspergillus japonicus* JV-23 as a potential source of paralytic compounds using a bioassay with silkworm (*Bombyx mori*).[18] At the same time, scientists based at the Pfizer laboratories in Sandwich, England, noted the anthelmintic activity of an aspergillus strain named IMI 337664.[19] After culture and purification, both groups were able to identify fractions of extract that exhibited the desired biological activities.

Analysis of the relevant fractions by mass spectrometry revealed a compound with mass 359, indicating a molecular formula of $C_{20}H_{29}N_3O_3$. The ^{13}C NMR spectra had resonances for three carbonyl groups along with four additional quaternary centres, five methyl groups, six CH_2

and two CH carbons. The similarity of the data with those from another alkaloid – paraherquamide F (VM55594)[20] and further analysis by ^{1}H NMR, HMBC and COSY experiments suggested the structure shown in Figure 1.7.



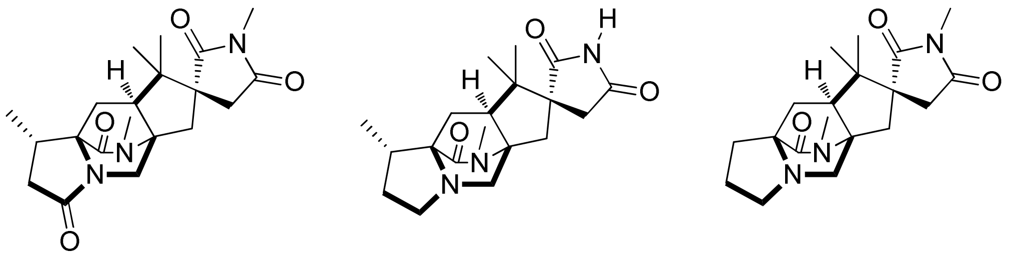
25 asperparaline A

arrows indicate relevant HMBC correlations

Figure 1.7

As shown, HMBC correlations from H-30 to C-2 and C-8 demonstrated that C-30 is an *N*-methyl in a succinimide motif. The presence of this structural feature was confirmed by the HMBC data around C-9 and C-21, and the *spiro* fused nature of the ring shown by correlations around C-3. Additionally, Hayashi reasoned that IR signals at 1773 and 1698 cm^{-1} corresponded with two imide carbonyls in a five membered ring. The amide portion of the bicyclo[2.2.2]diazaoctane motif was shown by connections from H-29 to C-18, while interactions around C-11, C-12, C-13 and C-19 confirmed the remainder of the core structure. The make-up of the pyrrolidine moiety was elucidated by a combination of both HMBC and ^{1}H-^{1}H COSY interactions. The relative stereochemistry was assigned by nOe experiments by irradiation of the protons at the *gem* dimethyl group (C-22/C-23) and comparison with the known paraherquamide F. The proposed structure was finally secured by X-Ray crystallography. It should be noted that this compound has also been known as aspergillimide and VM55598 but will hereon be referred to as asperparaline A.

In addition to asperparaline A, one of the 1997 reports detailed a related compound known as 16-keto aspergillimide or SB202327 (**26**, Figure 1.8). The structure was determined using similar methods to those discussed above. In 2000, another paper appeared in the literature by Hayashi and co-workers reporting two more alkaloids named asperparalines B (**27**) and C (**28**).[21]



26 16-keto aspergillimide (SB202327) **27** asperparaline B **28** asperparaline C

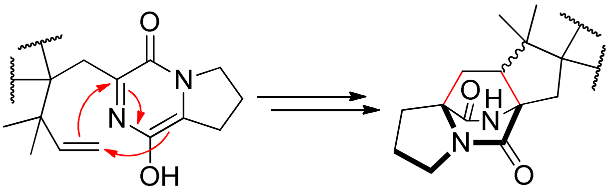
Figure 1.8

Analysis by mass spectrometry showed that both of these compounds had the molecular formula $C_{19}H_{27}N_3O_3$ suggesting structures similar to **25** but each lacking one methyl group. Further analysis by ^1H and ^{13}C NMR experiments showed that asperparaline B (**27**) possesses an N-H succinimide, while asperparaline C (**28**) lacks the methyl decoration on the pyrrolidine ring. The structure of the asperparalines are unique as no other natural products have been reported to incorporate a *spiro*-succinimide motif, leading to some interesting biosynthetic suggestions (*vide infra*).[22]

1.3 Biosynthesis

1.3.1 Origin of the bicyclo[2.2.2]diazaoctane motif

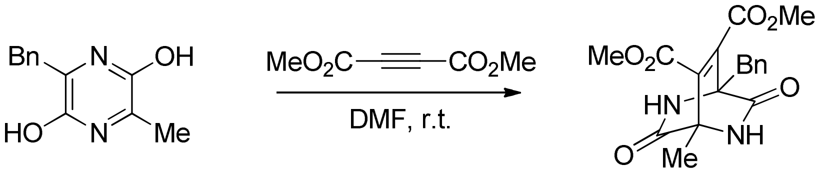
Soon after the isolation of the brevianamides, a short communication by Porter and Sammes reported a novel Diels-Alder reaction "of possible biosynthetic importance."[23] They postulated that the bridged bicyclic core could arise from a [4+2] cycloaddition of an isoprene unit to a DKP (Scheme 1.2).



29 **30**

Scheme 1.2

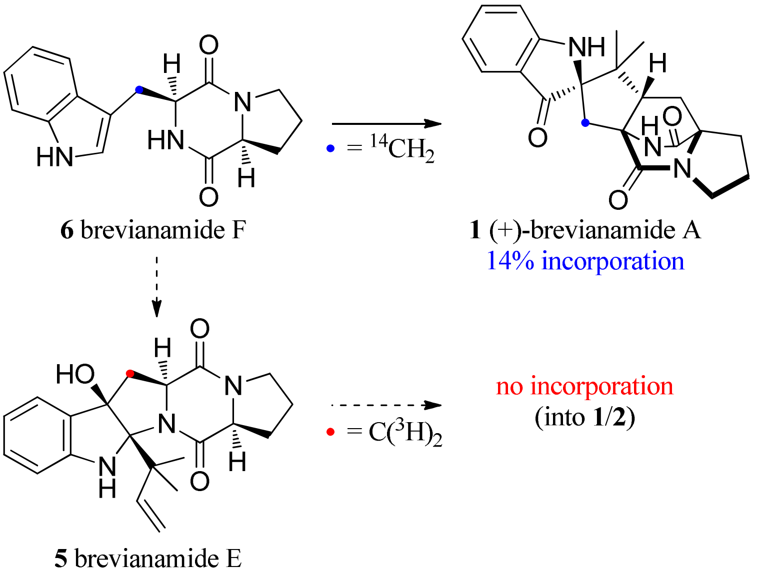
To test this hypothesis, they treated substituted pyrazine **31** with dimethyl acetylenedicarboxylate in DMF at room temperature and successfully isolated bicyclic DKP **32** (Scheme 1.3).[24]



31 **32**

Scheme 1.3

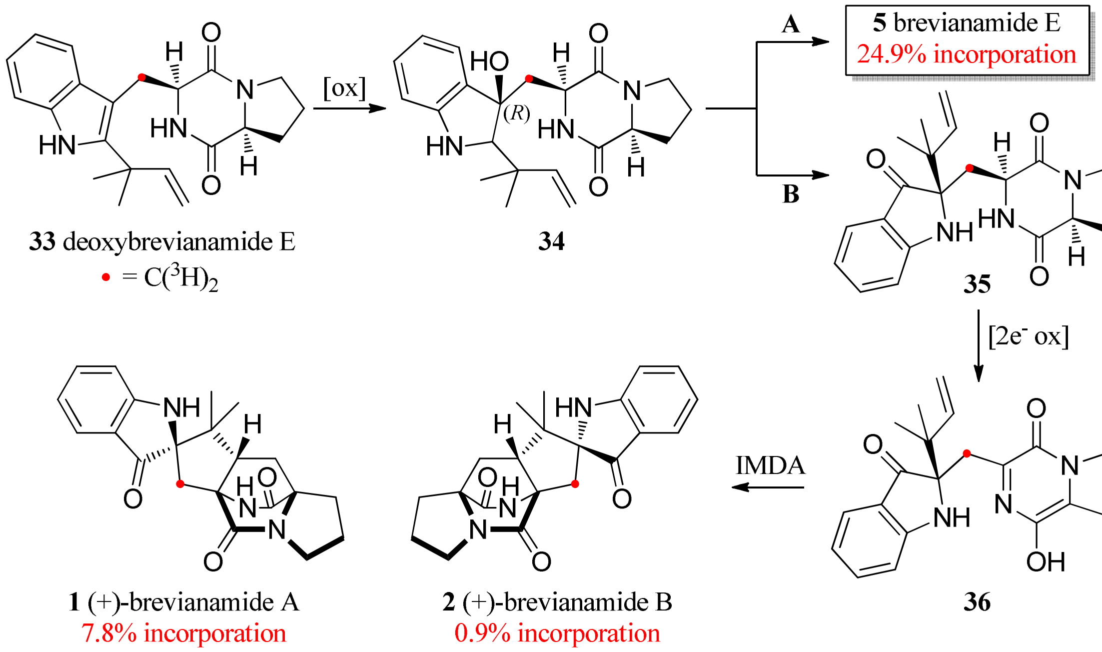
In 1974, Birch and co-workers showed that labelled brevianamide F (*cyclo*-L-tryptophyl-L-proline, **6**) was incorporated into brevianamide A when fed to *Penicillium brevicompactum*,[25] an organism it had previously been isolated from.[4] This led to the suggestion that brevianamides A and B could arise from isoprene addition to **6** followed by oxidation to form brevianamide E (**5**) as a precursor for the Diels-Alder cyclisation (Scheme 1.4).



Scheme 1.4

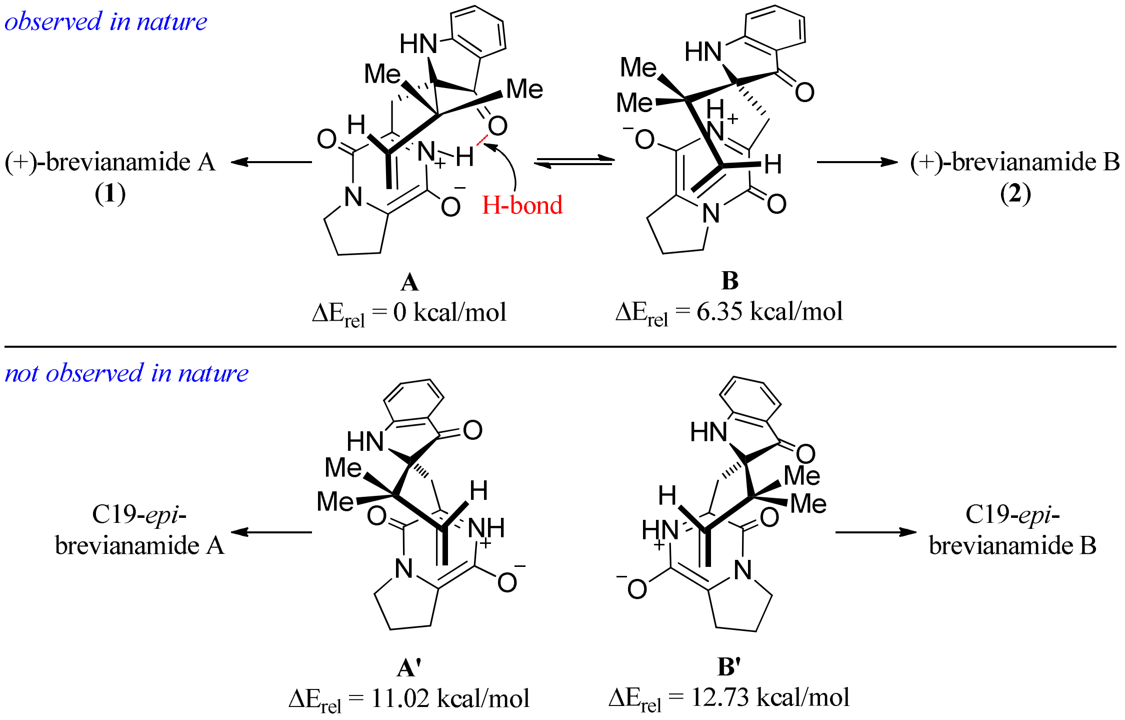
However, **5** has never been isolated from *Penicillium brevicompactum* and more recently it was demonstrated that synthetic brevianamide E was not incorporated into brevianamides A and B.[26] Instead, Williams and co-workers suggested that brevianamide E could arise from a 'shunt' pathway involving nucleophilic addition (Scheme 1.5, pathway **A**) whereas *spiro* rearrangement via pathway **B** would lead to **35**. Further oxidation and enol formation to give **36** would precede intramolecular Diels-Alder (IMDA) cycloaddition to give brevianamides A and B.

Using a ^3H labelled precursor, Williams observed incorporation of deoxybrevianamide E **33** into **1** and **2**, lending strong support to this proposed pathway. Interestingly only (+)-**1** and (+)-**2** have been isolated, presumably showing that the key IMDA proceeds with total facial selectivity. It has been proposed that this is the result of the involvement of an (*R*)-selective oxidase in the conversion of **33** to **34** giving rise to a single enantiomer of the IMDA precursor **36**.[27]



Scheme 1.5

Brevianamide A is produced in much larger quantities than brevianamide B. Calculations of the relative energies of the four possible transition states suggest that cyclisation from the two lowest energy transition states (**A** and **B**, Scheme 1.6) would lead to the naturally occurring alkaloids while cycloaddition proceeding through **A'** or **B'** would result in the unobserved C-19 epimers.[28] The lowest energy transition state could plausibly be stabilised by hydrogen bonding between the DKP and indole portions of the molecule helping to explain why brevianamide A is the more abundant natural product. However, as similar reactions in the laboratory do not show a significant bias for the *syn* or *anti* relationship it is not clear if the energy differences in these transition states have a strong effect.[28b]



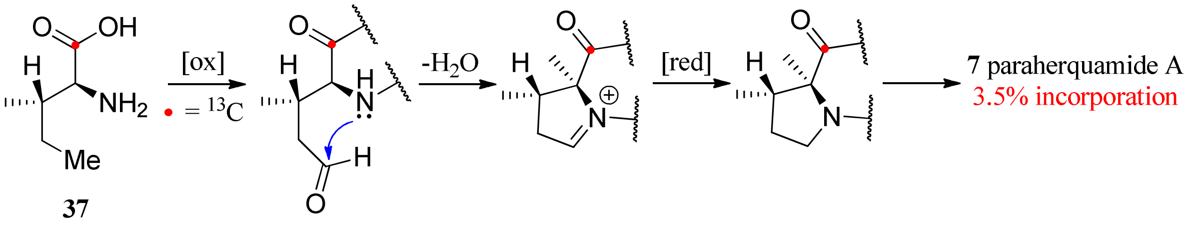
Scheme 1.6

The apparent stereoselectivity of this reaction raises the intriguing prospect of the involvement of an enzyme or other biomolecule.[29] Some evidence of this type of transformation exists, although not for the specific case of bridged DKPs. For example, Hilvert and co-workers have reported the catalysis of a Diels-Alder reaction by an antibody.[30] Ribonucleic acid (RNA) has also been shown to catalyse [4+2] cycloadditions[31] and some proteins have been isolated and identified as potential Diels-Alderases.[32]

1.3.2 Biosynthesis of the asperparalines and paraherquamides

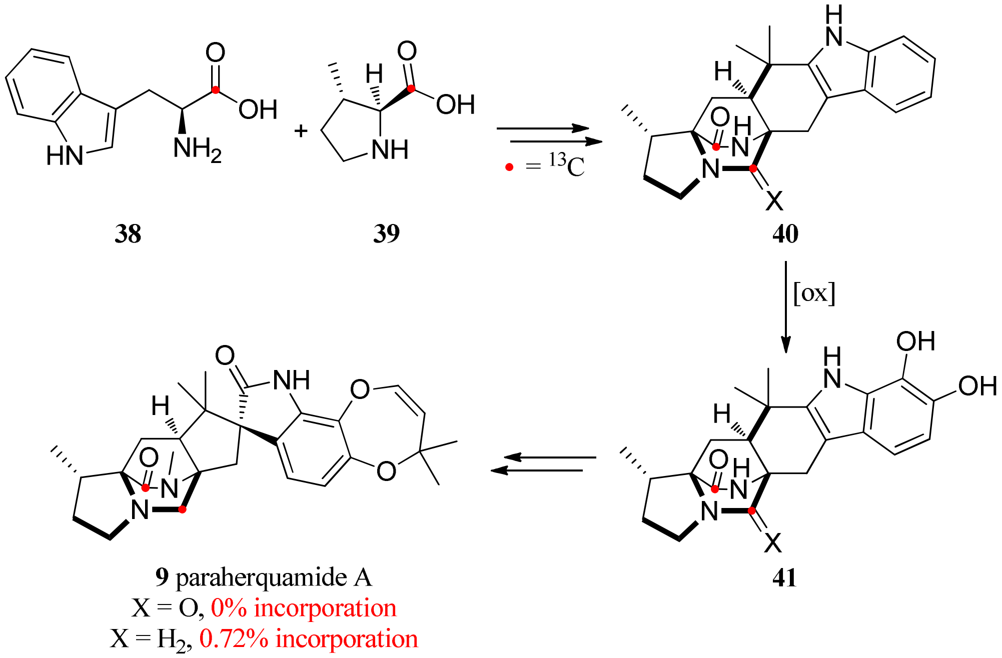
The biosynthetic origins of the asperparalines are particularly interesting due to the presence of the unique *spiro*-succinimide motif and are thought to be linked to that of the paraherquamides.[8c]

Early work on this combined pathway focused on the precursor for the β-methyl proline residue common to some members of the two families of natural products. The stereochemistry of the β-methyl group led to the theory that an oxidative cyclisation of isoleucine could be involved (Scheme 1.7). Feeding experiments with ^{13}C labelled isoleucine (**37**) suggest that this is indeed the case.[33]



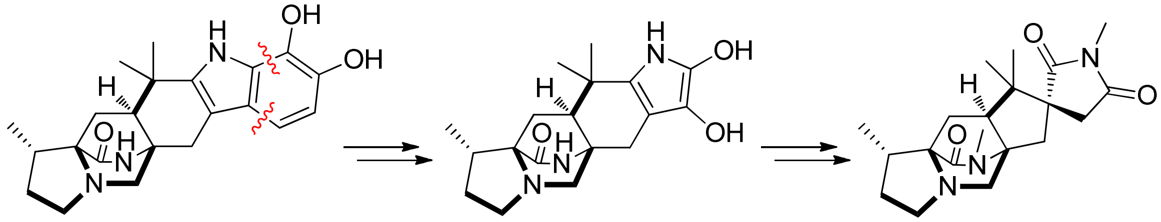
Scheme 1.7

Coupling of β-methyl proline with L-tryptophan followed by prenylation and Diels-Alder cyclisation (as outlined above) would lead to a common intermediate for both series of natural products (**40**, Scheme 1.8).



Scheme 1.8

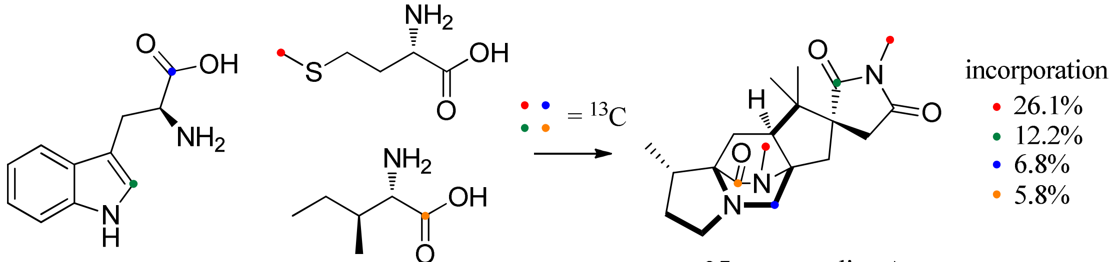
Williams has shown in feeding experiments that the diketopiperazine version of **40** is not a viable intermediate for the synthesis of paraherquamide, so reduction to the monoketopiperazine must occur at a relatively early stage. In paraherquamide synthesis, oxidation to dihydroxyindole **41** would be followed by prenylation and cyclisation to afford the dioxepin ring as shown in Scheme 1.8. Alternatively, cleavage of four carbons from the indole moiety would be followed by rearrangement to form the *spiro*-succinimide found in the asperparalines (Scheme 1.9).



41 **42** **25**, asperparaline A

Scheme 1.9

A number of further feeding experiments have been carried out to determine the amino acids incorporated into the asperparalines (Scheme 1.10). It appears that the β-methyl proline residue is derived from isoleucine (as above); the succinimide motif arises from tryptophan; and the amide and imide methyl groups come from methionine.[22]



25 asperparaline A

Scheme 1.10

It is also worth noting that the stereochemistry of the *spiro*-oxindole ring in the paraherquamides is the same as the *spiro*-succinimide in the asperparalines, lending weight to the idea of a common biosynthetic pathway.

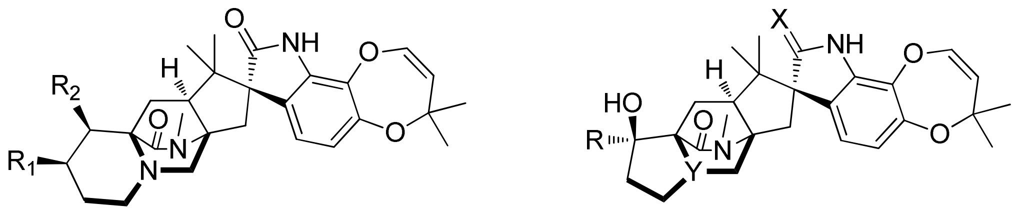
1.4 Biological Activity

Much of the interest around this family of alkaloids has been driven by the potent biological activities of some of the compounds.[34]

1.4.1 The paraherquamides and marcfortines

The paraherquamides and marcfortines have been investigated as antiparasitic agents. Paraherquamide A (**7**) was shown to be effective in sheep against a number of infections including *H. contortus*, *T. colubriformis* and *C. curticei*. A single oral dose was enough to remove the adult parasites. The research also demonstrated similar efficacy against nine nematodes in cattle with no side effects reported in either animal.[35] However, the same compound was found to be toxic in much lower doses in canine models.[36] Interestingly, there is no decrease in activity against strains resistant to other agents and no cross-resistance has been reported; this has led to speculation of a novel mode of action.[37]

The reported activity has led to a number of groups modifying the paraherquamides and marcfortines to probe their structure-activity relationship (SAR). Some examples of this are shown in Table 1.3.



compound	R_1	R_2
15	H	H
43	OH	H
44	OH	Me

compound	R	X	Y
9	Me	O	N
45	Et	O	N
46	Me	O	$^{+}$N–O^{-}
47	Me	H_2	N

Table 1.3

The initial results of these experiments were disappointing. For example, modification of marcfortine A (**15**) by adding a hydroxyl group to the piperidine ring to give **43** (Table 1.3) led to no improvement in efficacy.[38] Methylation adjacent to this position also had no effect (**44**).[39] More success was had through modifications to paraherquamide A (**9**). For example, replacing the pyrrolidine methyl group with an ethyl function (**45**) led to a 3-fold *in vitro* increase in activity against *C. elegans*.[40] Oxidising the tertiary amine in the monoketopiperazine gave **46** which was found to be equipotent to **9**,[7d] but reduction of the oxindole motif provided compound **47** with a 2-4 fold increase in efficacy.[41] Perhaps most interesting was the toxicity data for this compound. Whereas paraherquamide A has an LD_{50} in a mouse model of < 15 mg/kg, **47** showed no significant side effects at doses of 200 mg/kg. The same was seen in dogs where **9** was lethal at 0.5 mg/kg while a 25 mg/kg dose of **47** caused only mild side effects.

1.4.2 Avrainvillamide and the stephacidins

When avrainvillamide was first isolated, it was reported to have anti-proliferative activity against the breast cancer cell lines βT-549 and T-47D as well as the lung cancer line MALME-3M.[11-12] Interesting antibacterical effects were also reported with minimum inhibitory concentrations (MICs) against resistant strains considerably lower than some established antibiotics (Table 1.4).

microorganism	**MIC (µg/mL)** **avrainvillamide (18)**	**erythromycin**	**azithromycin**	**vancomycin**
S. aureus 01A1105	12.5	>100	>100	1.56
S. pyogenes 02C1068	12.5	>100	>100	0.39
E. faecalis 03A1069	25	>100	>100	12.5
E. coli 51A0266	>100	100	1.56	>100

Table 1.4

Two years later, Qian-Cutrone reported that the stephacidins have potent cytotoxic effects against a number of cancer cell lines including a Taxol resistant line.[14] Stephacidin B (**20**) seems to be the more potent of the two compounds (Table 1.5) and the authors noted that the effects are not mediated by p53, mdr, bcl2, tubulin or topoisomerase II suggesting an unusual mode of action.

cell line	**histotype**	**characteristic**	**IC_{50} (µM)** **19**	**20**
PC3	prostate	testosterone-independent	2.10	0.37
LNCaP	prostate	testosterone-sensitive	1.00	0.06
A2780	ovarian	parental	4.00	0.33
A2780/DDP	ovarian	mutp53/bcl2+	6.80	0.43
A2780/Tax	ovarian	Taxol-resistant	3.60	0.26
HCT116	colon	parental	2.10	0.46
HCT116/mdr+	colon	overexpress mdr+	6.70	0.46
HCT116/topo	colon	etoposide-resistant	13.10	0.42
MCF-7	beast	estradiol-sensitive	4.20	0.27
SKBR3	breast	estradiol-independent	2.15	0.32
LX-1	lung	sensitive	4.22	0.38

Table 1.5

As stephacidin B is the dimer of avrainvillamide, Myers and co-workers carried out a side-by-side analysis of the cytotoxicity of these two compounds as well as their unnatural enantiomers.[42] Interestingly, the unnatural enantiomers of the two compounds (i.e. *ent*-**18** and *ent*-**20**) also have reasonably low GI_{50} values (concentration at which cancer cell growth is inhibited by 50%). This is especially apparent in the case of the βT-549 cell line where the unnatural compounds are only about 2-fold less potent than their natural counterparts (Table 1.6).

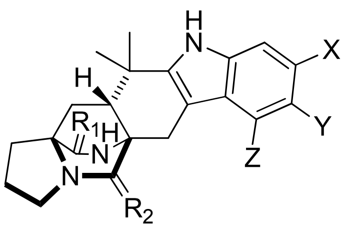
	GI$_{50}$ (nM)			
cell line	**18**	**20**	***ent*-18**	***ent*-20**
LNCaP	135	241	952	1514
βT-549	346	621	550	786
T-47D	91	205	942	1485
MALME-3M	289	406	987	1854

Table 1.6

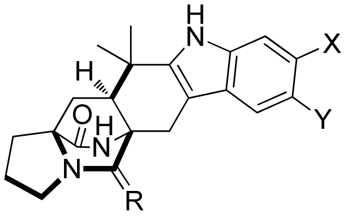
It is also worth noting that, correcting for stoichiometry, it seems the two compounds have almost the same activity (i.e. for equimolar solutions, stephacidin B is twice as active as avrainvillamide). This raises the question of which compound is actually responsible for the observed cytotoxic effects. To answer this, Myers followed the conversion of **20** (the dimer) into **18** (the monomer) by HPLC analysis of solutions of the compounds in cell culture medium. The half-life of stephacidin B was found to be about 50 minutes at 23 °C and only 10 minutes at 37 °C. As Myers reports it takes around 3 hours to prepare the solutions for cell culture assays, these data suggest that only negligible amounts of stephacidin B would be present. This indicates that avrainvillamide is probably responsible for the majority of the biological effects observed.

1.4.3 The malbrancheamides

Malbrancheamide has been reported to inhibit calcium modulated protein (calmodulin, CaM).[16a] Williams has synthesised a number of analogues of the malbrancheamides to further investigate this effect (Table 1.7).[43]



entry	R_1	R_2	X	Y	Z	IC_{50} (μM)
1	O	O	Cl	Cl	H	33.92 ± 4.64
2	O	O	H	Cl	Cl	45.41 ± 2.39
3	O	H_2	Cl	Cl	H	134.47 ±
4	H_2	O	Cl	Cl	H	11.95 ± 1.05
5	O	H_2	H	Cl	Cl	81.24 ± 4.46
6	H_2	O	H	Cl	Cl	27.51 ± 3.13
7	O	O	Cl	H	H	62.34 ± 4.58
8	O	O	H	Cl	H	42.77 ± 1.66
9	O	O	H	H	H	23.61 ± 4.21



entry	R	X	Y	IC_{50} (μM)
10	O	Cl	Cl	18.57 ± 2.87
11	O	H	Cl	31.49 ± 2.52
12	O	Cl	H	61.79 ± 5.48
13	O	H	H	150.66 ±
14 (±)-**21**	H_2	Cl	Cl	15.99 ± 0.87
15 (+)-**21**	H_2	Cl	Cl	19.33 ± 1.40
16	H_2	H	Cl	60.33 ± 6.84
17 (±)-**22**	H_2	Cl	H	42.85 ± 4.22
18 (+)-**22**	H_2	Cl	H	183.28 ±
19	H_2	H	H	35.73 ± 3.01

Table 1.7

Whilst it is difficult to draw firm conclusions from these data, it appears possible that the *unnatural* enantiomers of malbrancheamide (**21**) and malbrancheamide B (**22**) may be more active than their natural counterparts as the racemic mixtures have lower IC_{50} values than the corresponding genuine samples (entries 14/17 vs. 15/18). The chlorine present on the indole ring seems to improve potency in some cases, for example a dichlorinated compound (entry 10) is more active than the equivalent monochlorinated derivatives (entries 11 and 12) which in turn have lower IC_{50} values than the non-halogenated indole (entry 13). However, in other series the opposite appears to be true, for example entry 9 shows a higher activity than the chlorinated compounds in entries 7 and 8.

1.4.4 The asperparalines

Whilst both of the initial isolation papers contained preliminary biological testing data, little else is known about the activity of these compounds. Hayashi reported that asperparalines A, B and C all showed similar paralytic effects in silkworms.[18] A dose of 10 μg/g of diet was sufficient to cause paralysis for 7 to 10 hours within 1 hour of oral administration. A lower dose of 3 μg/g of body weight led to paralysis for 4 to 5 hours within 20 minutes of administration via syringe. The group at Pfizer carried out experiments using gerbils infected with the parasite *Trichostrongylus colubriformis*, measuring a reduction in faecal egg count after oral administration. A dose of 20 mg/kg of asperparaline A led to a 98% reduction while 16-keto aspergillimide had no effect.[19]

1.5 Summary

A wide variety of alkaloids possessing a characteristic bicyclo[2.2.2]diazaoctane core have been isolated. Numerous studies have investigated the biosynthesis of these molecules and probed their biological effects. Of particular interest are the various anti-cancer properties that have been reported.

Chapter 2

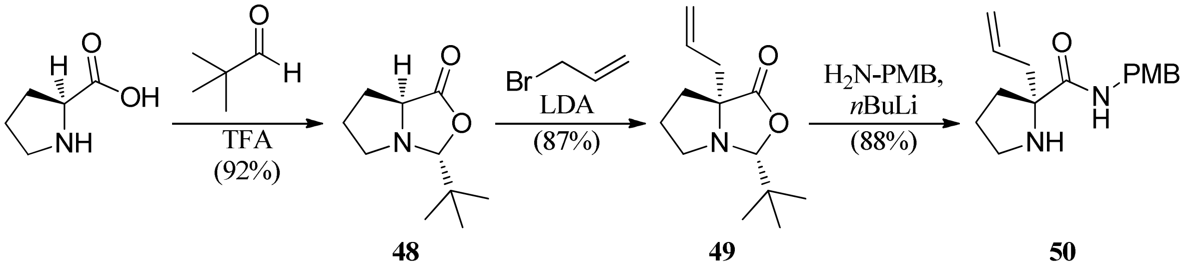
Previous Synthetic Work

2.1 Introduction

Since their isolation in the late 1960's, the family of alkaloids possessing a bicyclo[2.2.2]diazaoctane core has inspired numerous total syntheses. Driven by an interest in their biological profiles and intriguing biosynthesis, several groups have reported work in this area. Most notable is the group of Williams who have published in excess of twenty syntheses since completing brevianamide B in 1988.[44] This Chapter presents some of the most significant work in this field and is broadly arranged by the strategy employed for construction of the bridged core structure.

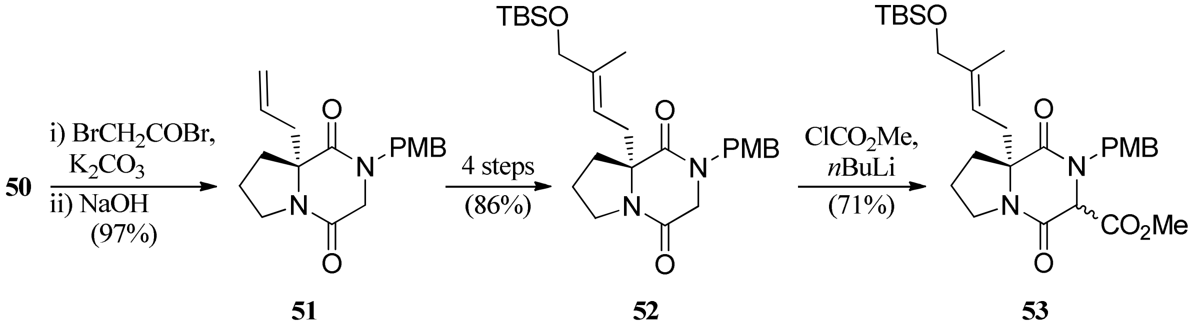
2.2 S_N2' Cyclisations

The Williams group's first synthesis of brevianamide B relied on an S_N2' reaction for the formation of the bridged core.[45] They began by using Seebach's method for the self-reproduction of chirality to synthesise pyrrolidine **50** (Scheme 2.1). This powerful sequence of reactions allows L-proline to be substituted with retention of the original stereochemistry. First, condensation of L-proline with pivalaldehyde affords oxazolidinone **48** as a single diastereomer in 92% yield. This compound is extremely sensitive to hydrolysis but can be purified by distillation and substituted with a variety of electrophiles. In this case deprotonation with LDA was followed by external quench with allyl bromide to afford oxazolidinone **49** in 87% yield. Next, treatment with lithiated *para*-methoxybenzylamine (PMB) led to clean conversion to amide **50**.



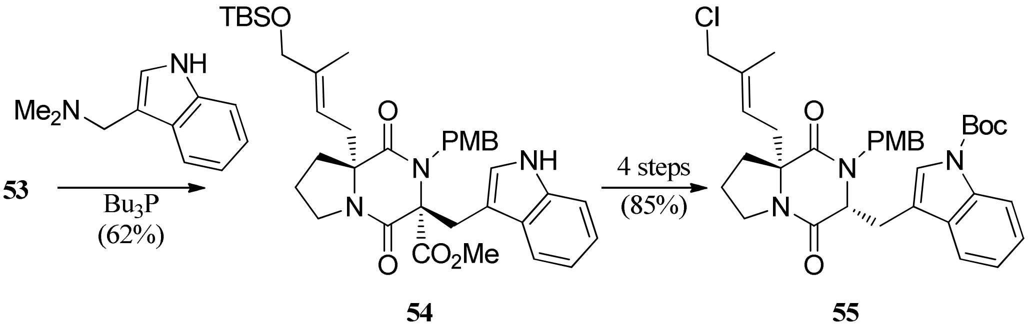
Scheme 2.1

Conversion of **50** into DKP **51** was achieved by acylation of the proline nitrogen with bromoacetyl bromide followed by treatment with sodium hydroxide in DCM (Scheme 2.2). From this intermediate a four-step procedure consisting of oxidative cleavage of the double bond, Wittig olefination, reduction, and protection with *tert*-butyldimethylsilyl chloride (TBS-Cl) gave **52** in 86% yield. The lithium enolate of **52** was then quenched with methyl chloroformate to afford ester **53** as a mixture of diastereomers.



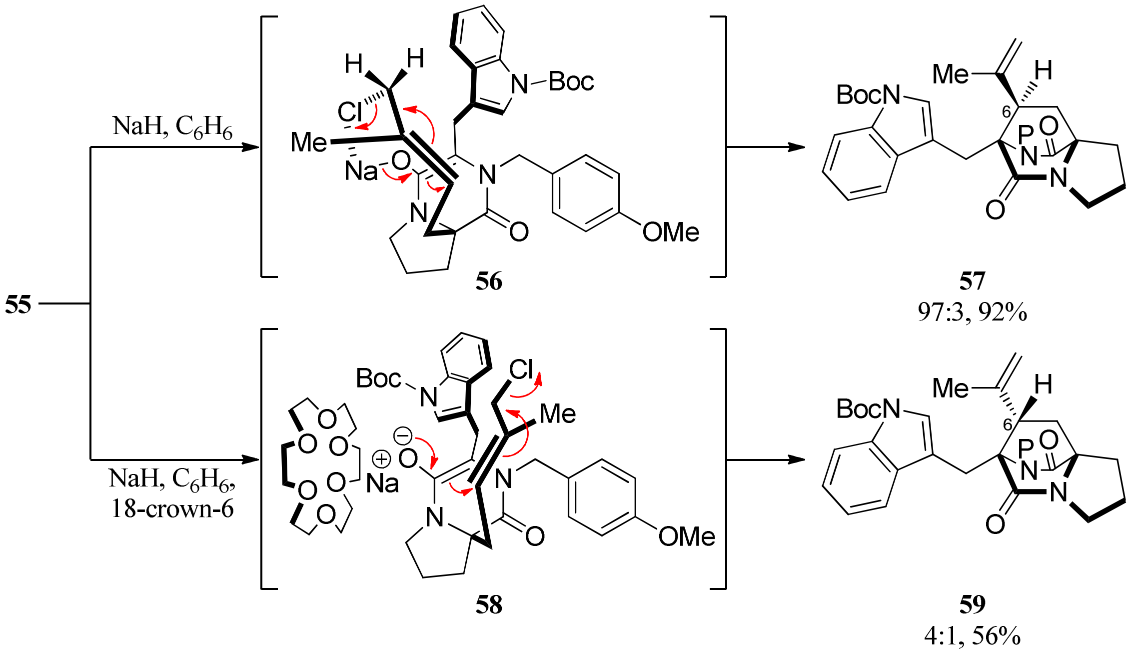
Scheme 2.2

To install the indole moiety, Williams used a method described by Kametani for the tributylphosphine catalysed coupling of gramine with 1,3-dicarbonyl **53** (Scheme 2.3).[46] This was followed by decarboxylation, indole protection, desilylation, and conversion to chloride **55** in 85% yield over the four steps.



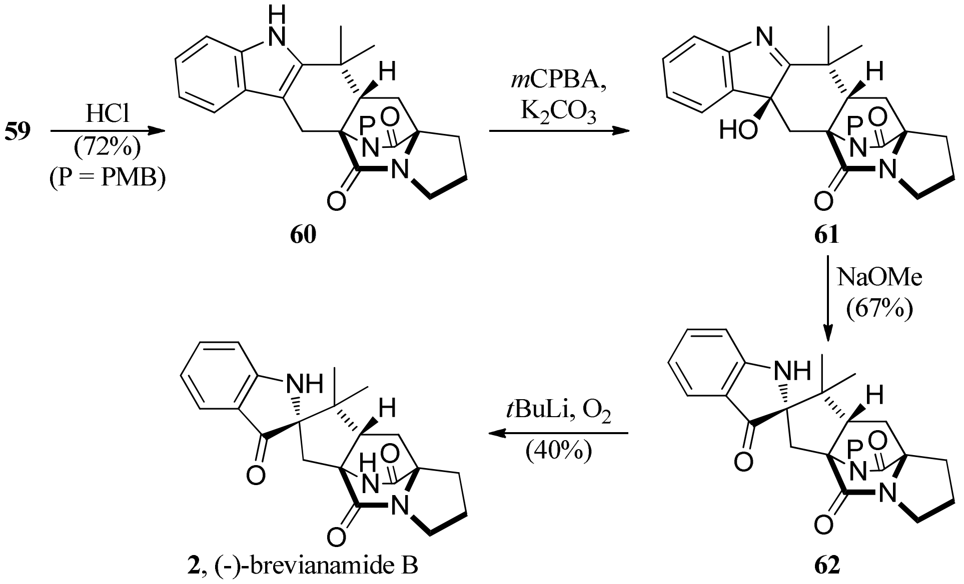
Scheme 2.3

Next, exposure to sodium hydride in benzene promoted the key S_N2' reaction to afford bridged DKP **57** in 92% yield (Scheme 2.4). However, analysis of the stereochemistry at the C-6 position showed that the undesired epimer had been formed. The authors postulated that this would arise from a “closed” transition state such as **56** where the chloride ion is in close proximity to the sodium enolate. Repeating the reaction with the addition of 18-crown-6 resulted in formation of the desired epimer (**59**). The authors suggest this is a result of solvation of the enolate to form the sterically crowded intermediate **58** and favour the desired stereochemistry.



Scheme 2.4

To complete the synthesis **59** was exposed to concentrated HCl causing Boc cleavage and cyclisation to form hexacycle **60**. Oxidation with *m*CPBA gave **61** as a single isomer which rearranged on treatment with sodium methoxide to give indoxyl **62**. Removal of the PMB protecting group proved difficult but was eventually achieved by deprotonation of the benzylic position with *tert*-butyllithium followed by quenching with oxygen to give (−)-brevianamide B (**2**) in 20 steps and 4% overall yield.



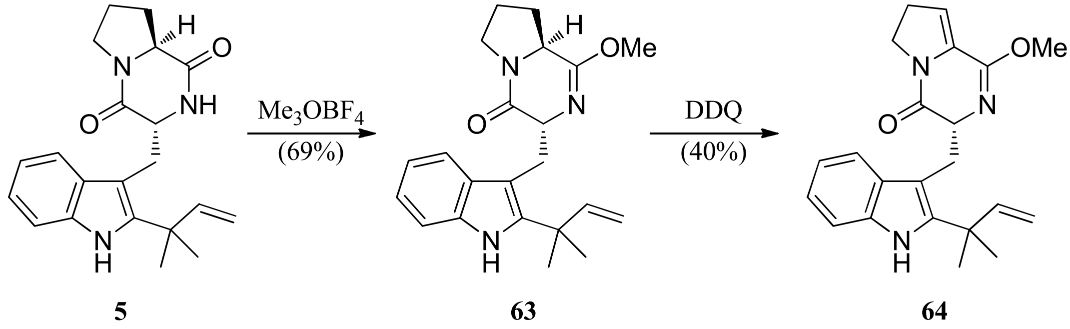
Scheme 2.5

The oxidation of **60** is interesting in that the epimer produced leads to brevianamide B rather than brevianamide A – the more abundant natural product. This suggests that if the biosynthesis follows a similar sequence, enzyme control is necessary for the oxidation to give the precursor to brevianamide A.

Following this work, the Williams group applied the same methodology to the synthesis of paraherquamides A[47] and B[48] as well as stephacidins A and B.[49]

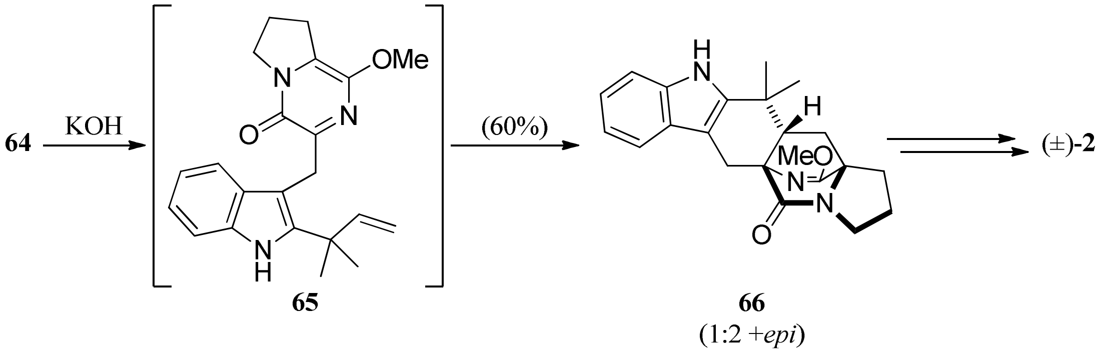
2.3 Intramolecular Diels-Alder Cyclisations

It has been proposed that in Nature the bicyclo[2.2.2]diazaoctane core arises through an intramolecular Diels-Alder (IMDA) cyclisation (*vide supra*). Testing this using a biomimetic approach to the synthesis of these compounds has always been an attractive proposition. The first demonstration of this strategy in the laboratory was Williams' synthesis of brevianamide B.[50] They began with brevianamide E (**5**) which had previously been synthesised by Kametani.[51] Protection of the DKP as the lactim ether was accomplished by treatment with trimethyloxonium tetrafluoroborate to furnish **63** in 69% yield. Next, oxidation with DDQ gave the unsaturated Diels-Alder precursor **64** (Scheme 2.6).



Scheme 2.6

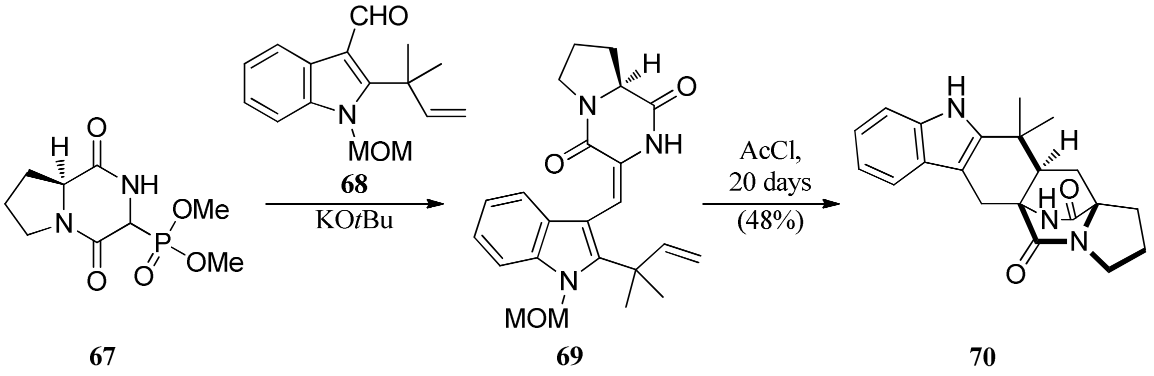
Treatment of **64** with potassium hydroxide afforded azadiene **65** which spontaneously cyclised to give bridged DKP **66** as a 2:1 mixture of epimers (Scheme 2.7). In this case, the minor epimer was converted to brevianamide B (**2**) in a sequence similar to that shown above in Section 2.2.



Scheme 2.7

Unfortunately, as the proposed intermediate **65** is prochiral the products are necessarily racemic. Nevertheless, the extremely rapid access to the bicyclo[2.2.2]diazaoctane core results in a very brief access to these natural products.

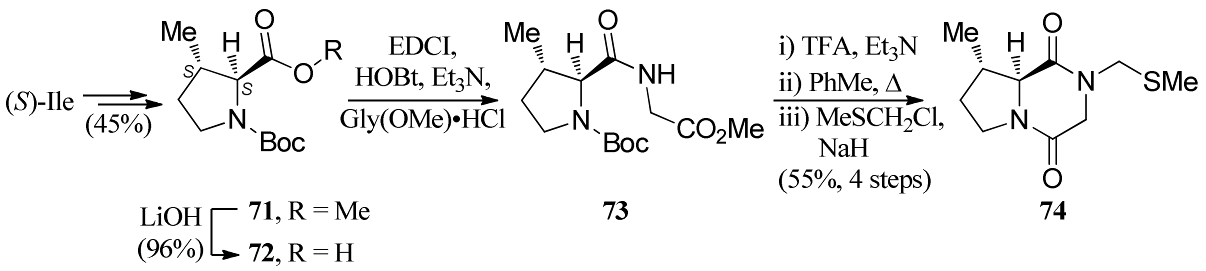
Independently of Williams, Liebscher and co-workers reported a similar Diels-Alder reaction for the construction of a model system (Scheme 2.8).[52]



Scheme 2.8

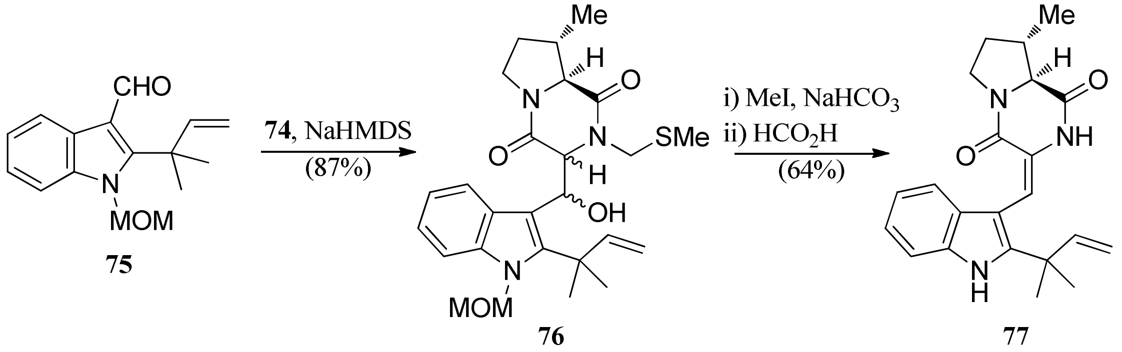
The cyclisation precursor **69** was synthesised from DKP **67** and substituted indole **68**. Exposure of this compound to neat acetyl chloride afforded the bridged core **70** as a single diastereomer in 48% yield after twenty days.

While Liebscher never applied this methodology to a total synthesis, Williams realised that the use of this method would allow the stereochemistry of a substituent on the pyrrolidine ring to be retained throughout the sequence. The alkaloid VM55599 provided a suitable target for this approach.[53] Starting from (*S*)-isoleucine, methylproline **71** was synthesised in an overall yield of 45% (Scheme 2.9). Hydrolysis of the methyl ester under standard conditions was followed by amide bond formation to furnish dipeptide **73**. Treatment with trifluoroacetic acid resulted in Boc cleavage before heating in toluene to effect cyclisation. Protection of the amide as the methylthiomethyl (MTM) derivative was accomplished using sodium hydride as the base to give DKP **74** in 55% yield over four steps.



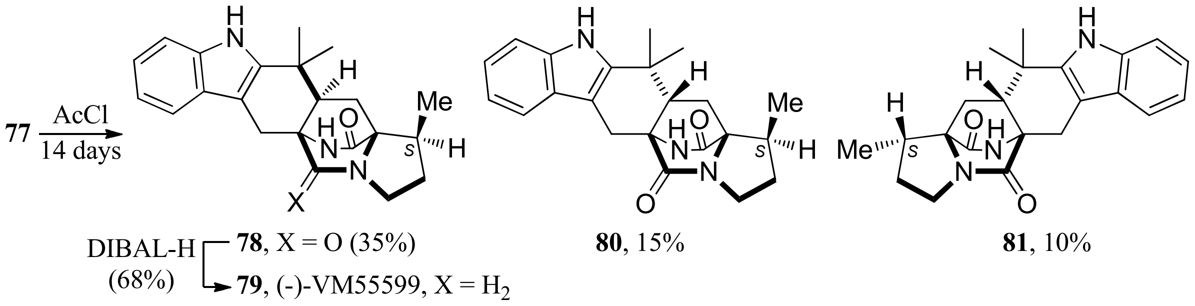
Scheme 2.9

Next, the enolate of **74** was generated with sodium bis(trimethylsilyl)amide and this was condensed with aldehyde **75** to afford DKP **76** as a mixture of diastereomers (Scheme 2.10). Treatment with methyl iodide in the presence of sodium hydrogen carbonate resulted in MTM cleavage and exposure of the resulting intermediate to formic acid led to dehydration and hydrolysis of the methoxymethyl (MOM) group to yield **77**.



Scheme 2.10

Using the conditions previously described by Liebscher, the Diels-Alder reaction proceeded through three of the four possible transition states to give **78**, **80** and **81** – all with retention of the stereochemistry on the methylproline (Scheme 2.11). A DIBAL mediated reduction of the tertiary amide of the major component (**78**) furnished (−)-VM55599 (**79**) as a single enantiomer.



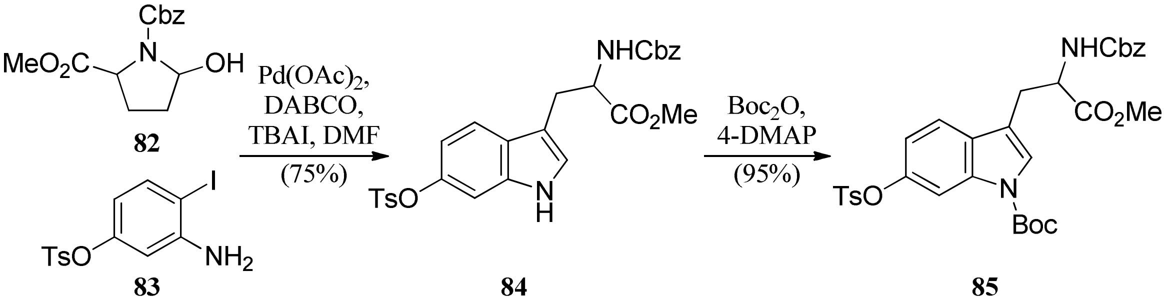
Scheme 2.11

Although the selectivity of the penultimate step of this route is poor, this synthesis allowed the determination of the absolute configuration of VM55599.

As well as the examples shown here, Williams has applied this IMDA strategy to the synthesis of stephacidin A,[54] marcfortine C[55] and the malbrancheamides.[56]

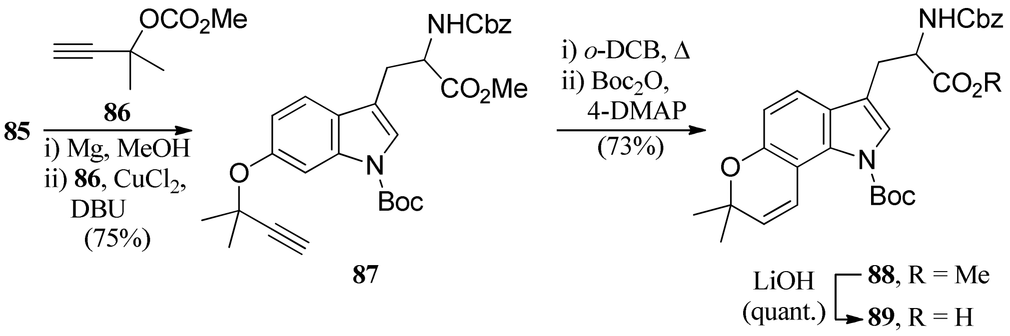
2.4 Oxidative Enolate Coupling

Baran and co-workers devised an approach to avrainvillamide and the stephacidins using an oxidative enolate coupling to synthesise the core bridged DKP.[57] These natural products are somewhat complex as they incorporate an acid sensitive pyran ring. To synthesise this component, Baran began with hydroxypyrrolidine **82** and aniline **83** (Scheme 2.12). After extensive optimisation studies, they found that treatment with palladium acetate, 1,4-diazabicyclo[2.2.2]octane (DABCO) and tetrabutylammonium iodide (TBAI) afforded the desired tryptophan **84** in good yield. The authors propose that this proceeds through condensation of the aniline with the aldehyde tautomer of **82** followed by a palladium catalysed cyclisation. Subsequent selective protection of the amine nitrogen was accomplished under standard conditions (Boc_2O, 4-DMAP) to yield **85**.



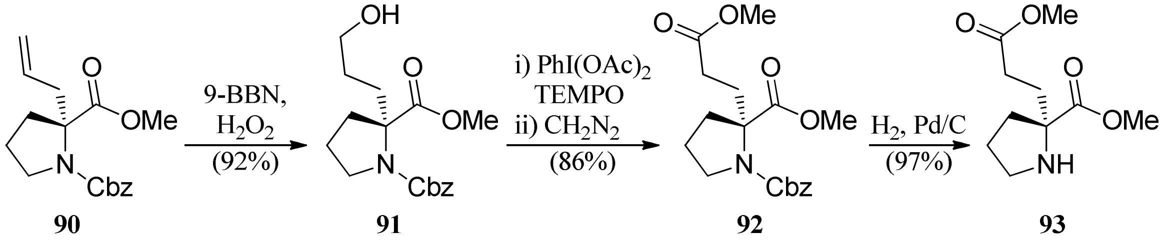
Scheme 2.12

To install the pyran ring, a magnesium promoted removal of the tosyl group was followed by a copper catalysed addition of acetylene **86** to give **87** in 75% yield over the two steps (Scheme 2.13). Heating this compound in *ortho*-dichlorobenzene caused cyclisation via an allenyl Claisen rearrangement, but this unfortunately also led to deprotection of the indole nitrogen. Reprotection with Boc anhydride was then followed by ester saponification using lithium hydroxide to afford the completed indole section as carboxylic acid **89**.



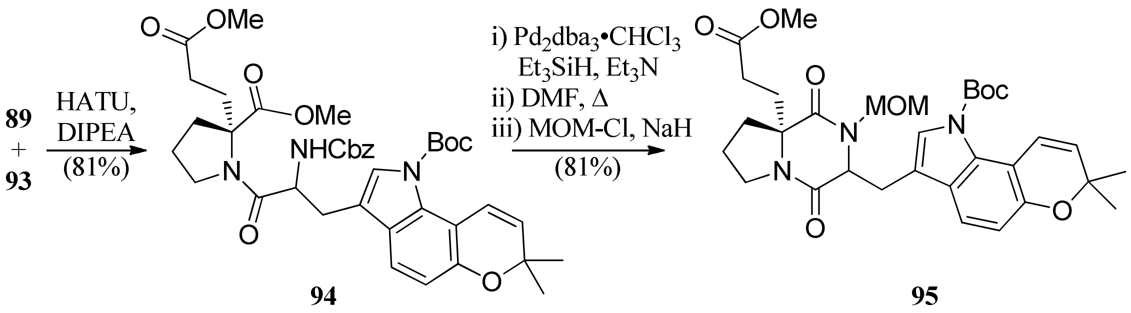
Scheme 2.13

In a similar way to previous syntheses by Williams (*vide supra*) pyrrolidine **90** was obtained using Seebach's method for the self-reproduction of chirality. Hydroxylation of the olefin gave **91** which was oxidised to the corresponding acid and methylated with diazomethane to give ester **92** in good yield (Scheme 2.14). Removal of the Cbz protecting group afforded the free amine **93** ready for coupling with acid **89**.



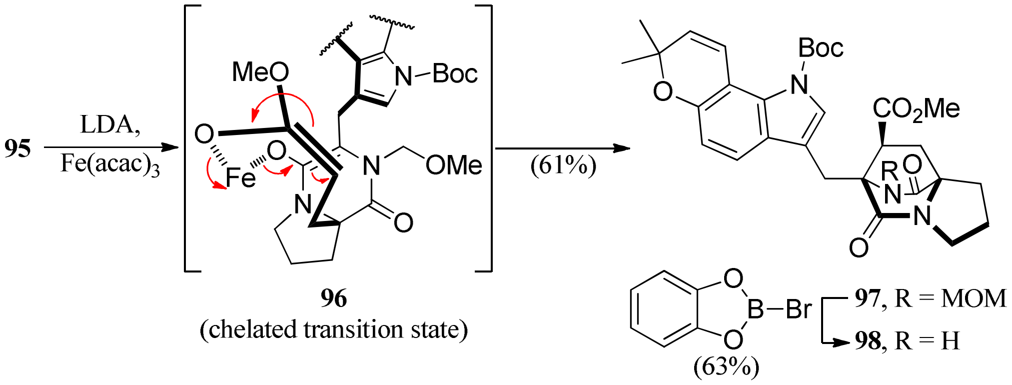
Scheme 2.14

Amide bond formation proceeded well under standard conditions (HATU, Hünig's base) to yield dipeptide **94** (Scheme 2.15). To obtain the desired DKP, a palladium catalysed Cbz removal was followed by thermal cyclisation on heating in DMF. At this point the amide nitrogen was protected with a MOM group to afford **95** primed for the key oxidative cyclisation step.



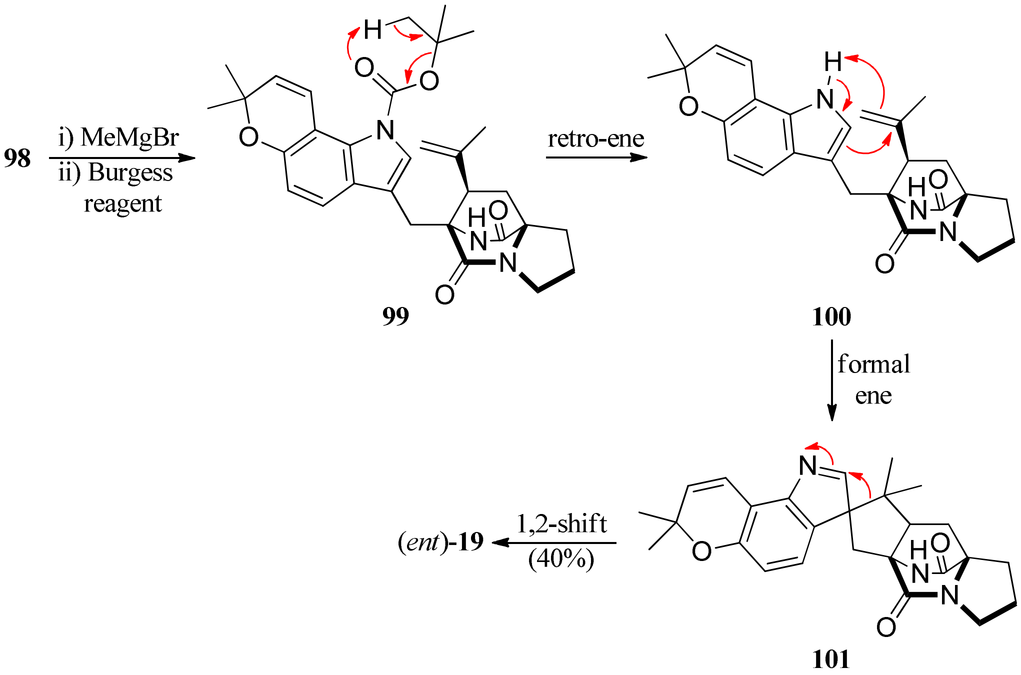
Scheme 2.15

Using conditions investigated on a simpler model system, deprotonation of **95** with LDA was followed by treatment of the resulting enolate with the oxidant Fe(acac)$_3$ leading to bridged DKP **97** (Scheme 2.16). The authors propose that the reaction occurs through the chelated transition state **96** in an analogous manner to that described by the Williams group in their S$_N$2' work. As well as the concerted process shown in Scheme 2.16 the reaction could proceed through a radical mechanism either by initial ester oxidation, or initial amide oxidation, or diradical formation. In any case, **97** was obtained as single diastereomer which was deprotected with *B*-bromocatecholborane to afford bridged DKP **98**.



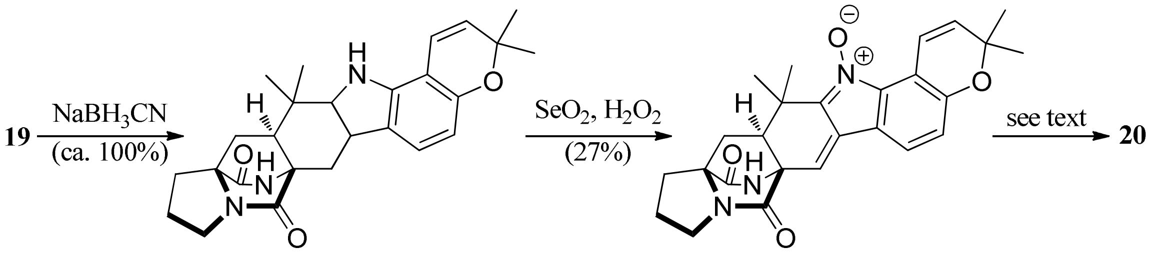
Scheme 2.16

To complete the synthesis the ester was converted into the corresponding olefin by treatment with methyl Grignard followed by dehydration using the Burgess reagent. The acid-sensitive nature of the pyran ring made Boc removal non-trivial, but Baran found that heating to 200 °C effected deprotection, formal ene cyclisation, and 1,2-rearrangement to afford stephacidin A (**19**) as shown in Scheme 2.17.



Scheme 2.17

At the time of this work the absolute configuration of stephacidin A was not known. Comparison of a synthetic sample with a genuine isolation product showed that the authors had, in starting from L-proline, produced *ent*-**19**. To confirm this, Baran repeated the synthesis with D-proline and found that the resulting compound matched exactly with the natural sample. In addition, Baran carried this material forward to avrainvillamide (**18**) and stephacidin B (**20**) as outlined in Scheme 2.18. Reduction of the indole to the corresponding indoline proceeded in near-quantitative yield on treatment with sodium cyanoborohydride to afford intermediate **102**. This was then oxidised by exposure to selenium oxide and hydrogen peroxide to yield avrainvillamide (**18**) in 27% yield.



102 **18**, avrainvillamide

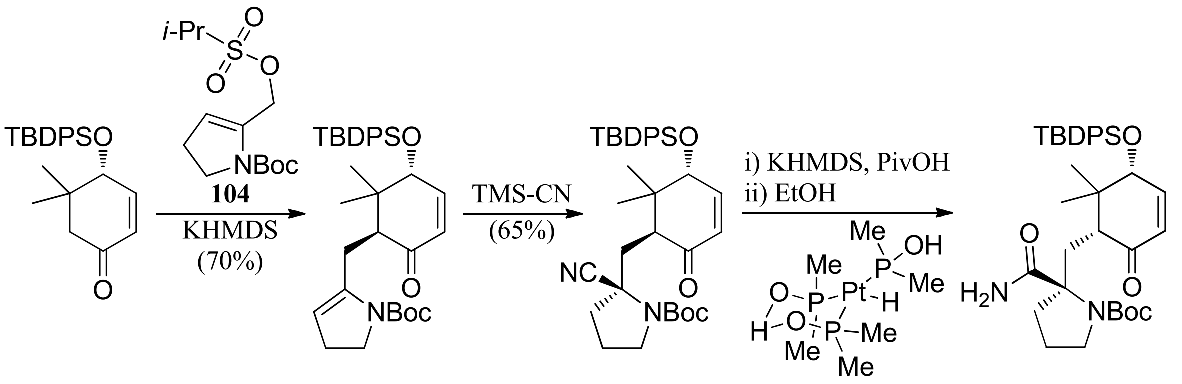
Scheme 2.18

The authors found that **18** dimerises to stephacidin B (**20**) under a variety of conditions such as purification by preparative TLC (PTLC; SiO_2, EtOAc) or dissolution in DMSO followed by solvent removal *in vacuo*.

2.5 Radical Cyclisations

2.5.1 Work by Myers

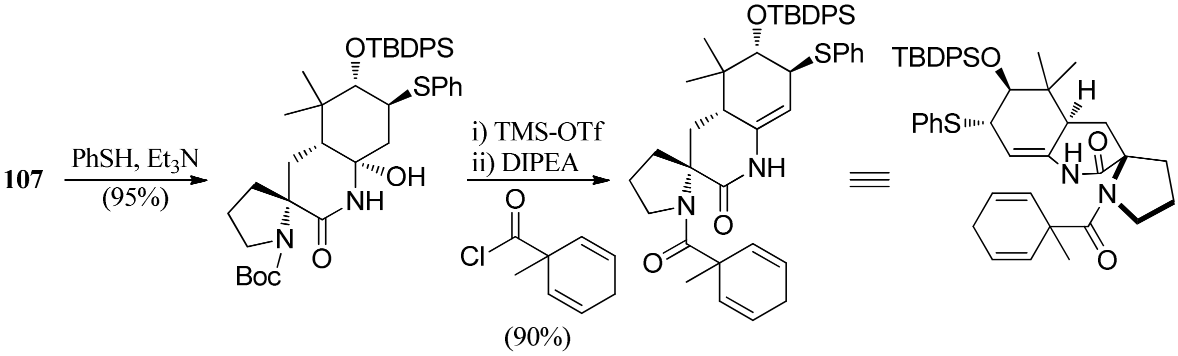
In 2005, Myers and co-workers published a synthesis of avrainvillamide and stephacidin B using a radical cyclisation to form the bicyclo[2.2.2]diazaoctane core.[58] Their synthesis began with the known cyclohexanone **103** (Scheme 2.19).[59] Alkylation of the corresponding enolate with dihydropyrrole sulfonate **104** proceeded with complete stereoselectivity to give the *trans* product **105** in 70% yield. Next, addition of hydrogen cyanide under Strecker conditions gave 65% of **106** along with 16% of the corresponding diastereomer (not shown). Epimerisation *alpha* to the carbonyl was accomplished by quenching of the potassium enolate with pivalic acid, before hydration of the nitrile using a platinum catalyst developed by Ghaffar and Parkins gave primary amide **107** in good yield.



103 **105** **106** **107**

Scheme 2.19

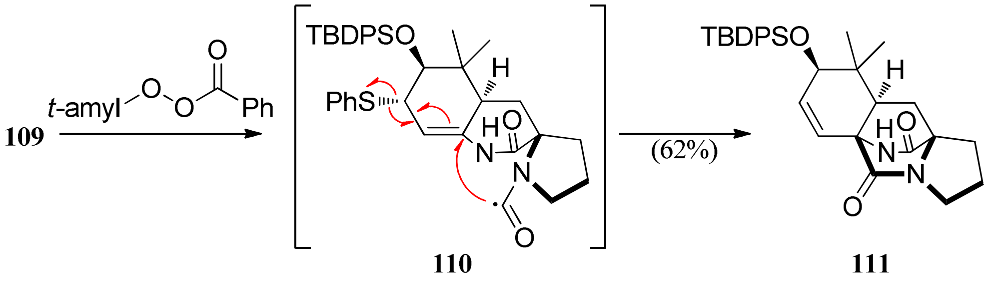
Treatment of **107** with thiophenol in the presence of triethylamine led to Michael addition of sulfur along with hemiaminal formation to afford **108**. Next, trimethylsilyl triflate (TMS-OTf) was used to cleave the Boc protecting group with simultaneous dehydration and the pyrrolidine nitrogen was acylated in the presence of Hünig's base to give radical precursor **109**.



108 **109**

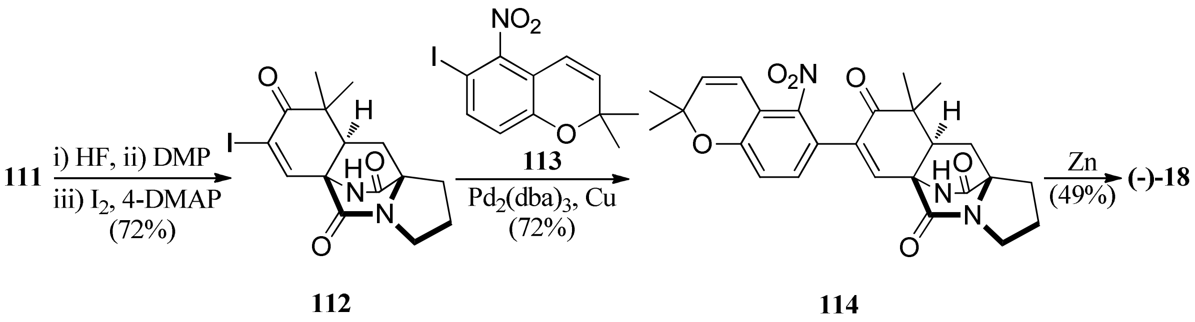
Scheme 2.20

To initiate the key cyclisation, **109** was heated with *tert*-amyl peroxybenzoate to form the bridged DKP **111** in 62% yield (Scheme 2.21). The authors postulate that this proceeds through formation of aminoacyl radical **110** followed by attack of the enamide at the more substituted carbon leading to migration of the double bond and expulsion of a sulfur radical.



Scheme 2.21

With the central bicyclo[2.2.2]diazaoctane constructed, Myers used an Ullmann coupling to introduce the indole portion of the molecule.[60] Thus, desilylation using hydrogen fluoride followed by oxidation with Dess-Martin periodinane yielded an enone which was converted to the α-iodo derivative (**112**) with iodine and 4-DMAP (Scheme 2.22). Next, coupling with known aryl iodide **113**[61] proceeded in good yield to afford **114** with all of the carbons required for the total synthesis in place. The synthesis was completed by reduction of the nitro group with zinc to afford (−)-avrainvillamide (**18**) in 49% yield.

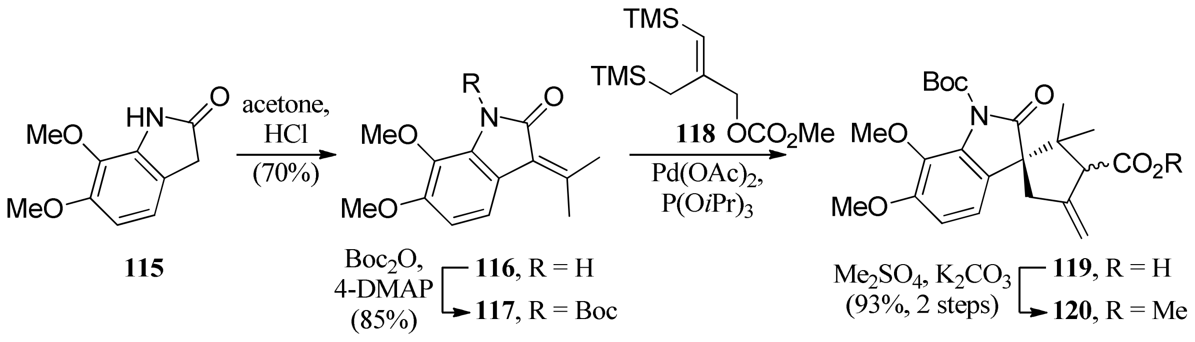


Scheme 2.22

In a similar way to Baran (*vide supra*), Myers found that dimerisation of **18** to stephacidin B (**20**) occurred on exposure to triethylamine, again helping to confirm the biosynthetic relationship between these two components.

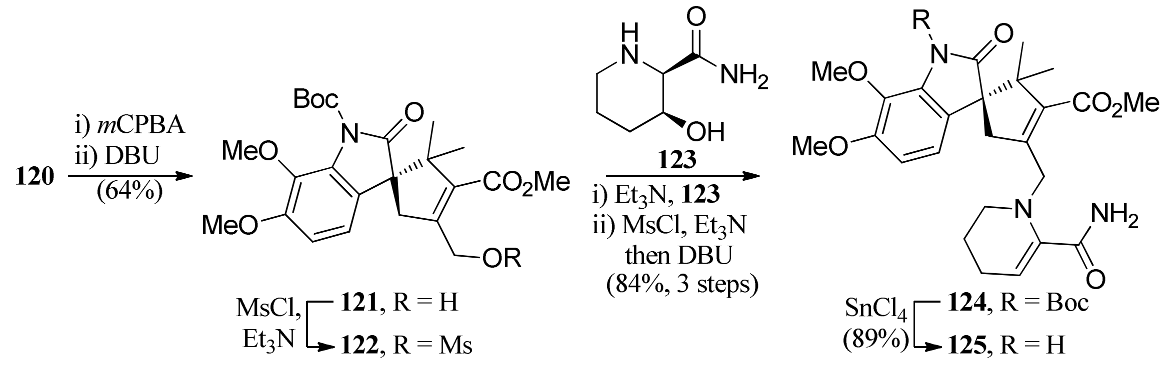
2.5.2 Work by Trost

Trost and co-workers applied a radical cyclisation for the construction of the MKP ring found in marcfortine B.[62] Their synthesis relied on a trimethylenemethane (TMM) [3+2]-cycloaddition between **117** (accessible from known oxindole **115**)[63] and previously prepared **118**[64] (Scheme 2.23). The cycloaddition proceeded well to afford tricycle **120** in 93% yield after methylation with dimethyl sulfate.



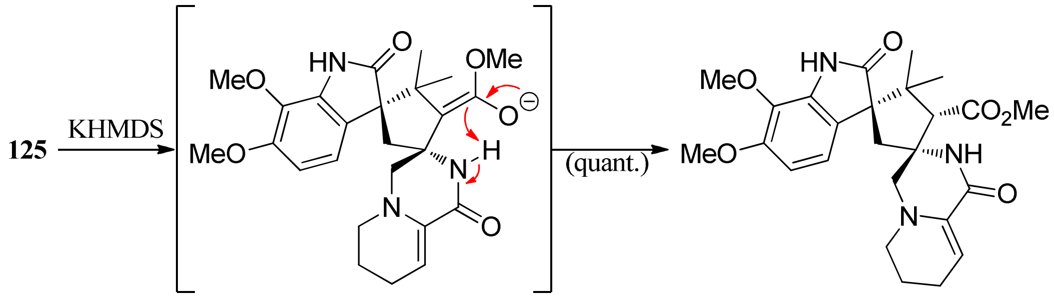
Scheme 2.23

Epoxidation of the exocyclic double bond was achieved with *m*CPBA before ring opening with DBU gave primary alcohol **121** in 64% yield (Scheme 2.24). Formation of the mesylate (**122**) was followed by substitution with piperidine **123** which, after elimination of the secondary alcohol, gave amide **124**. At this stage, the authors found that subsequent steps led to degradation of the material, but this could be avoided by Boc removal using tin tetrachloride to afford **125**.



Scheme 2.24

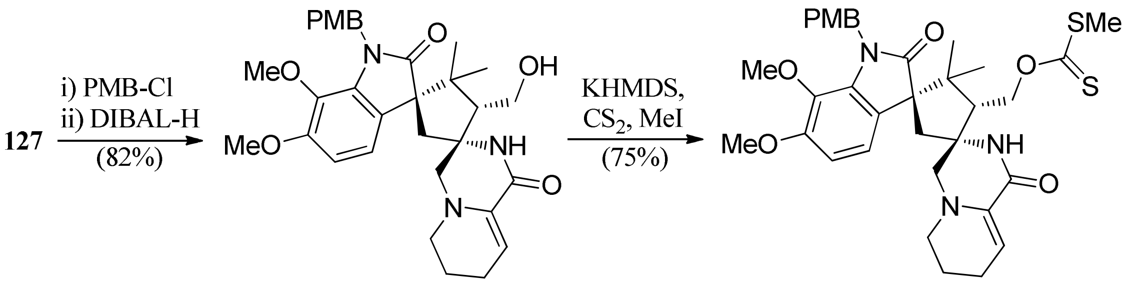
Treatment of this compound with KHMDS promoted a Michael addition of the primary amide to generate the *spiro* ring junction of **127** as a single diastereomer (Scheme 2.25). Trost speculates that the stereocontrol in this step is a result of intramolecular protonation of the intermediate enolate by the amide N-H of **126**. Replacing this with the corresponding *N*-methyl amide results in a complete reversal to give exclusively the undesired isomer.



126 **127**

Scheme 2.25

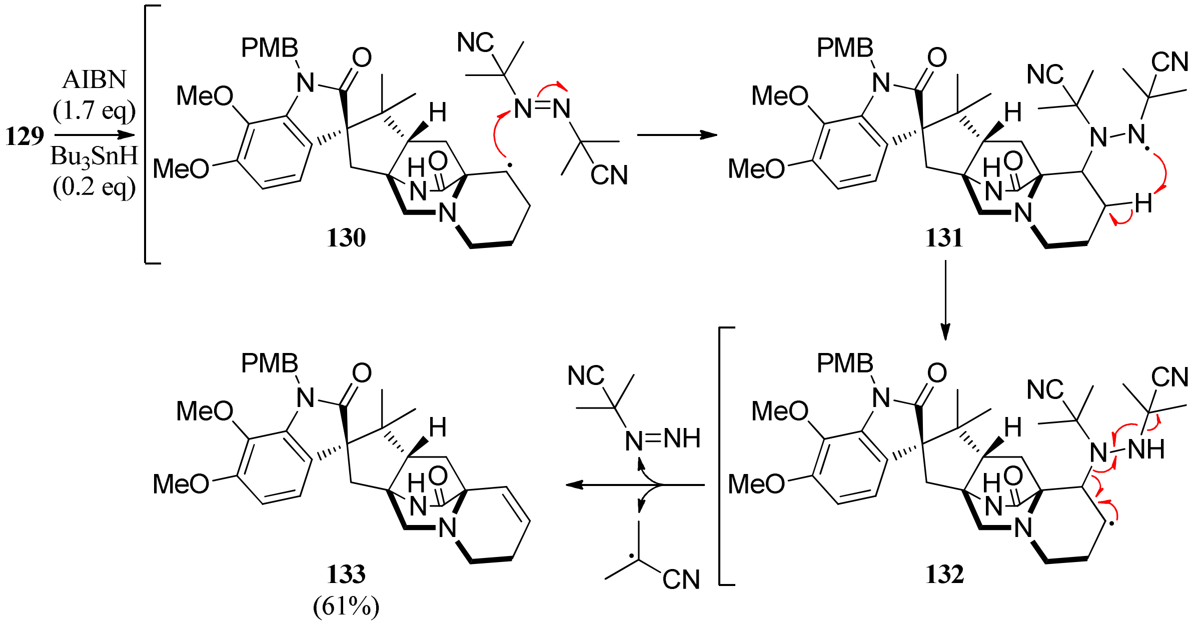
To improve solubility, the oxindole of **127** was protected with PMB chloride before DIBAL mediated reduction of the methyl ester to the corresponding alcohol (**128**, Scheme 2.26). Substitution with carbon disulfide was followed by *S*-methylation to form xanthate **129** as the precursor for the radical cyclisation.



128 **129**

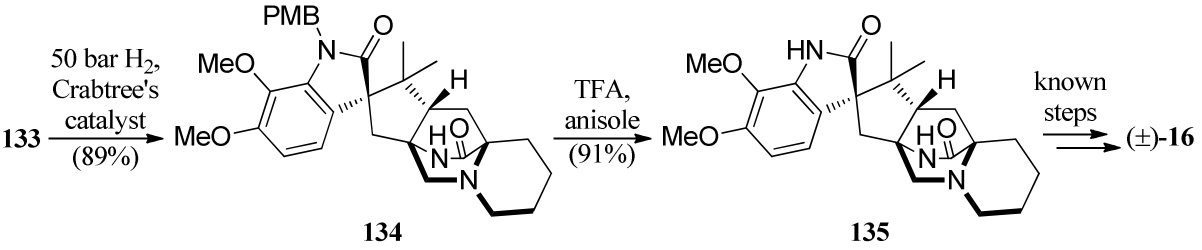
Scheme 2.26

Despite investigating numerous conditions, the authors were unable to isolate the desired saturated product **134** (Scheme 2.28). Instead, unsaturated compound **133** was isolated which they speculate could arise from reaction of intermediate **130** with AIBN to give **131** (Scheme 2.27). Subsequent 1,4-H abstraction to give **132** would then be followed by elimination to afford the observed product, **133**. After optimisation studies, the best yield (61%) was achieved using 1.7 equivalents of AIBN along with sub-stoichiometric quantities of tin hydride.



Scheme 2.27

To obtain the desired saturated compound, reduction of the unwanted double bond in **133** was accomplished using Crabtree’s catalyst under a hydrogen atmosphere. Cleavage of the PMB protecting group failed under several standard conditions but proceeded cleanly on exposure to TFA in refluxing anisole to afford **135**.

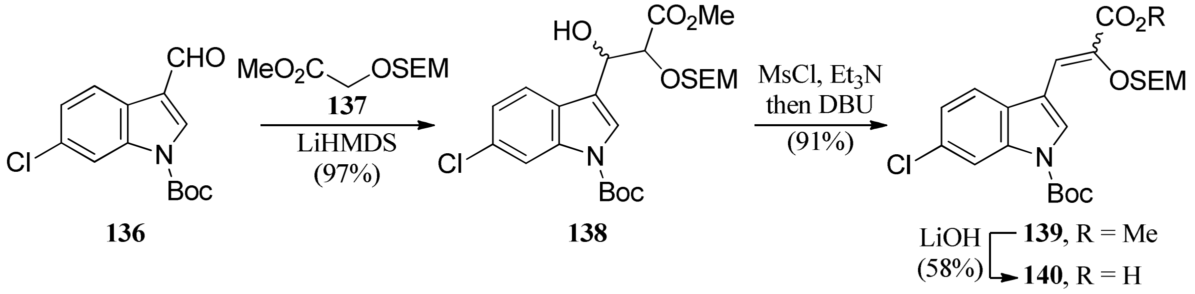


Scheme 2.28

To complete the synthesis, the dioxepine ring was constructed in four steps following Williams' procedure to furnish (±)-marcfortine B (**16**).[65]

2.6 Cationic Cascade

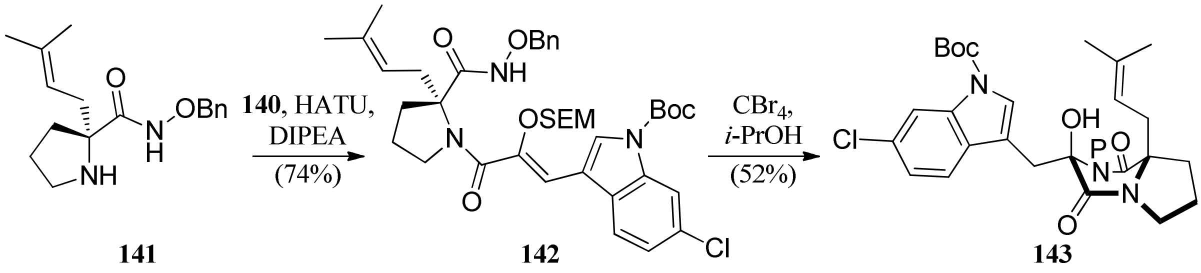
In 2009, our group published a concise synthesis of malbrancheamide B using a cationic cascade to construct the central bicyclo[2.2.2]diazaoctane.[66] The synthesis began with chloroindole **136**, prepared using a method reported by Appella (Scheme 2.29).[67] Condensation with ester **137** proceeded in near-quantitative yield in the presence of LiHMDS to afford aldol product **138** as a 4:1 mixture of diastereomers. Elimination of the secondary alcohol was accomplished by mesylate formation followed by treatment with DBU to give **139**. Lithium hydroxide mediated ester cleavage gave the free acid **140**, albeit in relatively modest yield due to partial cleavage of the Boc group before completion of the desired reaction.



Scheme 2.29

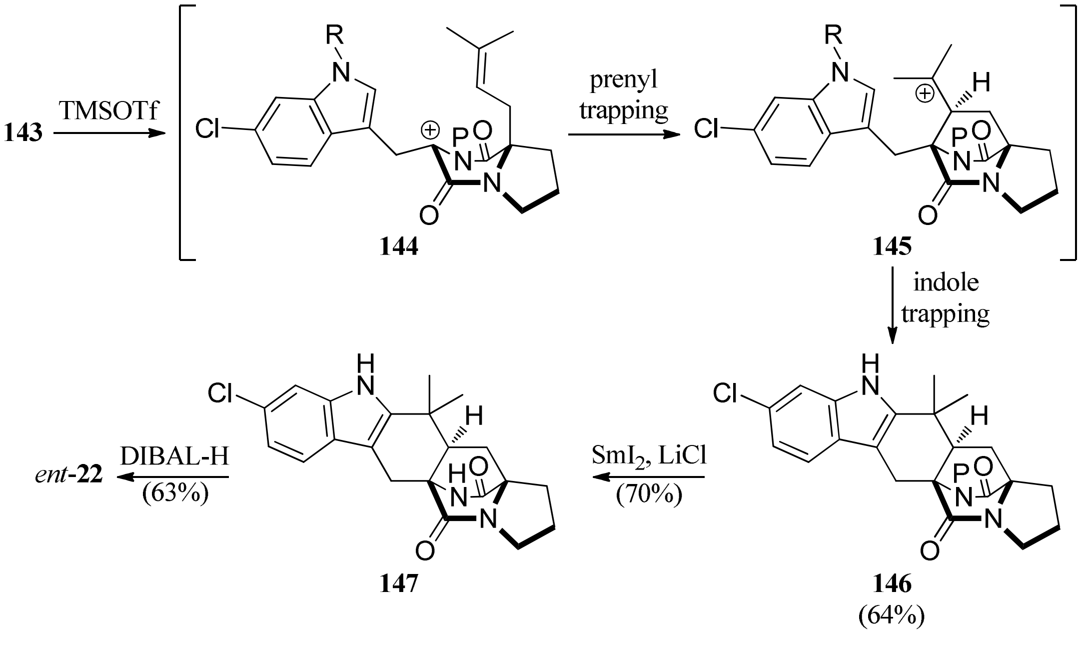
Prenylated proline **141** was synthesised using Seebach's method for the self-reproduction of chirality (*vide supra*). The *O*-benzyl protecting group was selected to avoid the problems previously observed with PMB cleavage from bridged DKPs. Coupling of **141** with acid **140** was accomplished using HATU and Hünig's base to afford dipeptide **142** in 74% yield

(Scheme 2.30). Surprisingly, SEM ether **142** was stable to a variety of standard deprotection conditions such as TBAF, TBAT and HF. In the end, treatment with carbon tetrabromide in warm (50 °C) *iso*propanol led to SEM cleavage along with DKP formation to give the cyclisation precursor **143**.



Scheme 2.30

To initiate the key cationic cascade, alcohol **143** was exposed to TMS triflate in dichloromethane (Scheme 2.31). This presumably leads to formation of **144** which undergoes prenyl trapping to give tertiary cation **145**. Indole trapping of this intermediate then affords the malbrancheamide core **146** in 64% yield as a 4:1 mixture of diastereomers. At some point in this sequence Boc deprotection occurs to reveal the N-H indole.



Scheme 2.31

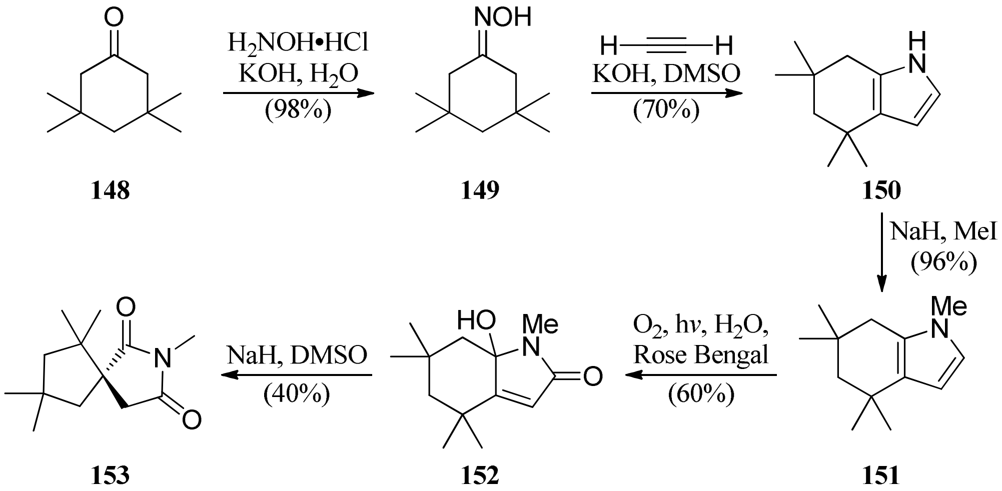
To complete the synthesis, samarium iodide mediated *O*-benzyl removal was followed by reduction using Williams' protocol to afford *ent*-malbrancheamide B (*ent*-**22**). While the overall yield of this sequence is lower than Williams' corresponding IMDA approach, a single enantiomer of malbrancheamide B is delivered in only 10 steps from commercially available 6-chloroindole.

2.7 Work Towards the Asperparalines

As outlined above, there have been numerous total syntheses of alkaloids possessing a bicyclo[2.2.2]diazaoctane core. However, no synthesis of the asperparalines has been forthcoming. The synthetic challenges of these molecules include the unusual *spiro*-succinimide motif; decoration on the proline ring; and the partially reduced nature of the central DKP. There have, however, been a number of reports detailing the synthesis of various components of the asperparalines.

2.7.1 Work by Williams

As with many other prenylated indole alkaloids, the first published work towards the asperparalines was by the Williams group in 1999.[68] This involves an approach to a *spiro* fused 5,5 ring system incorporating a succinimide and methylated cyclopentane (Scheme 2.32). Commercially available cyclohexanone **148** was converted into oxime **149** by treatment with hydroxylamine hydrochloride. Pyrrole **151** was synthesised in good yield using the Trofimov reaction followed by methylation of the resulting amine. Oxidation was accomplished under photooxygenation conditions before the key *spiro*-rearrangment was carried out by heating **152** in DMSO in the presence of sodium hydride.

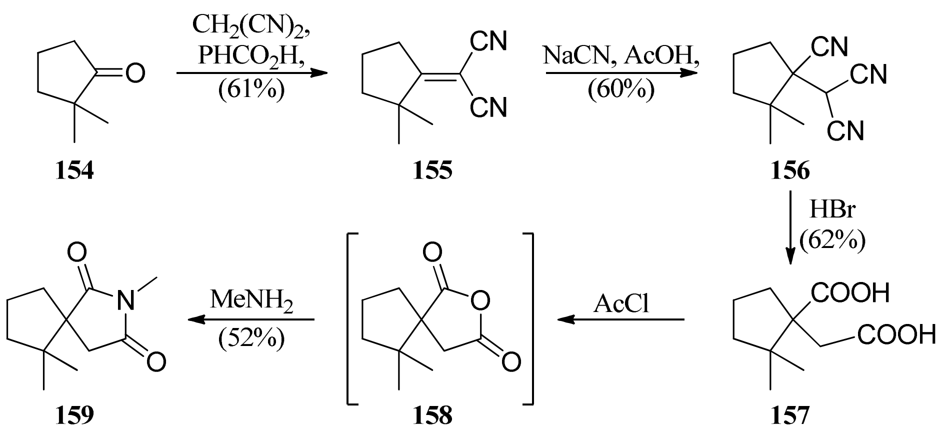


Scheme 2.32

Since this paper, Williams has published no further work on the synthesis of the asperparalines.

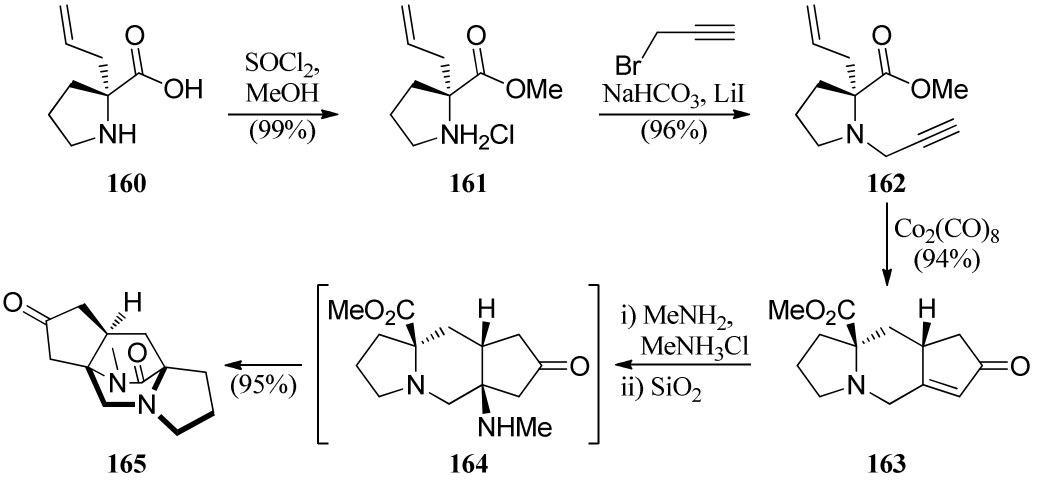
2.7.2 Work by Tanimori

The Tanimori group has published several papers discussing the synthesis of the asperparalines. In 2000, they published an approach to the *spiro*-succinimide motif starting from cyclopentanone **154** (Scheme 2.33).[69] Treatment with malononitrile afforded dinitrile **155** in reasonable yield and a third nitrile group was incorporated by Michael addition of sodium cyanide to give **156**. Hydrolysis and decarboxylation then led to diacid **157** which, in turn, was transformed to anhydride **158**. The synthesis was completed by conversion of the anhydride to succinimide **159** by treatment with methylamine.



Scheme 2.33

Soon after this approach to the succinimide portion of the molecule, the same group published a synthesis of the core bicyclo[2.2.2]diazaoctane motif using a Pauson-Khand reaction (Scheme 2.34).[70] Allyl proline **160** was prepared using the Seebach protocol for the self-reproduction of chirality. Formation of the methyl ester **161** and *N*-propargylation afforded enyne **162**. Treatment of **162** with $Co_2(CO)_8$ in a mixture of THF and DMSO triggered the desired Pauson-Khand [2+2+1] cycloaddition to form tricycle **163** in up to 94% yield and as a single diastereomer. Finally, treatment with methylamine and exposure to silica gel furnished bridged tetracycle **165** in excellent yield.



Scheme 2.34

In principle, combining these two approaches would lead to the asperparaline core structure but no further reports have been forthcoming.

2.8 Summary

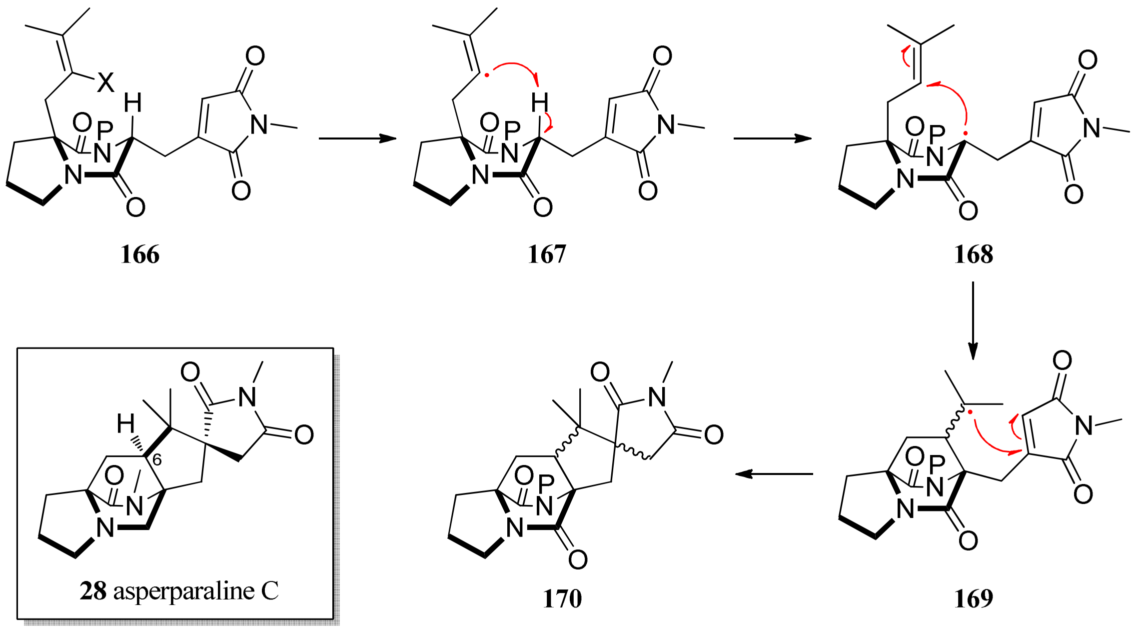
Since their discovery, the family of prenylated indole alkaloids possessing a bridged bicyclo[2.2.2]diazaoctane core have provided inspiration for numerous total syntheses. A number of different methods have been used for the construction of key parts of these molecules driving the development of new chemistry and helping to further understand the biosynthetic origins of these molecules. The asperparalines are yet to succumb to total synthesis but their unique structure has inspired novel approaches to individual features.

Chapter 3

Model System Studies

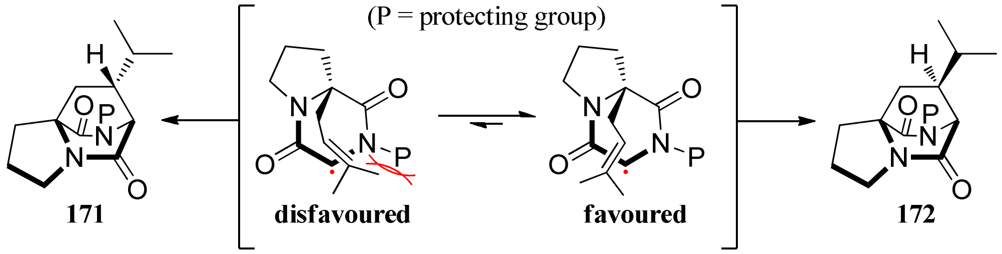
3.1 Introduction

As discussed in Chapter 2, previous work in our group has led to the total synthesis of both (+)-brevianamide B and (−)-malbrancheamide B using a cationic cascade.[66] We were interested in applying an analogous radical cyclisation cascade to the synthesis of the asperparaline C core structure (Scheme 3.1). Starting with a DKP such as **166** possessing a substituted proline along with a maleimide moiety, generation of a vinylic radical by scission of a suitable group (e.g. X = Br, I) would be followed by 1,6-hydrogen atom transfer to give radical **168**. A *6-exo-trig* cyclisation would then form a new carbon-carbon bond to afford intermediate **169**. We hoped that this would then undergo a *5-exo-trig* cyclisation onto the maleimide to afford the asperparaline core structure **170**.



Scheme 3.1

A number of potential problems are evident in this process. Firstly, the vital stereochemistry at C-6 needs to be controlled. Previous work in our group has shown that the analogous cationic cascade tends to favour the *syn* relationship between the pyrrolidine and DKP ring junction – the same configuration as the majority of the natural products. However, the extent of this bias depends on the protecting group on the amide nitrogen. As shown in Scheme 3.2, this is presumably due to steric constraints – the favoured transition state positions the bulky dimethyl group away from protecting group P leading to products such as **172**. We hoped that similar interactions in our system would give the same stereochemical outcome.



Scheme 3.2

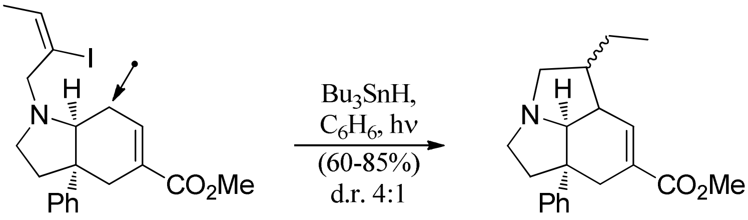
Secondly, as well as the 1,6-H abstraction there is a competing 1,5-H transfer alpha to the nitrogen in the pyrrolidine motif. We hope that a combination of conformational and stereoelectronic effects will promote the pathway we desire (*vide infra*).

Finally, the cyclisation onto the maleimide might proceed in *6-endo-trig* mode rather than the *5-exo-trig* attack that we desire. The stereocontrol at the *spiro* centre is also important. These issues are discussed further in Chapter 4.

3.2 Radical Translocations

While there are numerous methods for the radical cleavage of various C-X bonds (e.g. with AIBN/tributyltin hydride) direct intermolecular radical scission of an unactivated C-H bond is typically significantly endothermic. Intramolecular abstraction of a hydrogen atom from a remote position has often been used as an elegant solution to this problem.[71]

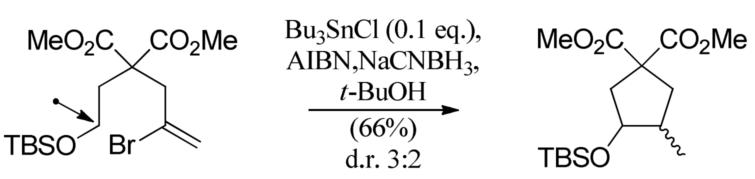
Parsons and co-workers published an early example where they showed that a radical generated from a vinyl iodide would abstract an allylic hydrogen in 1,5 mode (Scheme 3.3).[72] This abstraction was followed by a *5-exo-trig* cyclisation to afford the pyrrolizidine ring system of **174**. As all of the mechanisms in this section are broadly similar to that shown above in Scheme 3.1, individual steps are not shown but an arrow (•⟶) is used to indicate the position from which the hydrogen atom is transferred.



173 **174**

Scheme 3.3

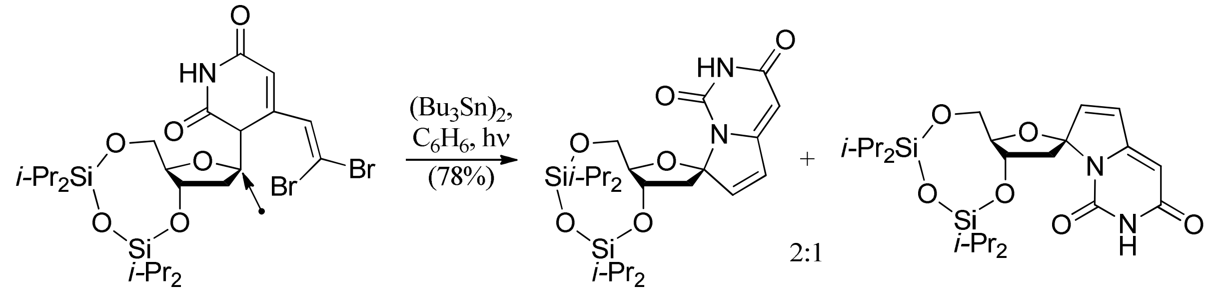
Seminal work by Curran demonstrated a more general method for hydrogen atom transfer. Radical scission of a vinyl bromide was again followed by cyclisation in *5-exo-trig* mode (Scheme 3.4).[73] In this case, the site of abstraction on **175** is alpha to a functional group that weakens the C-H bond and stabilises the newly formed radical. This example uses a protected alcohol but acetals, esters and phenyl rings were also shown to be effective.



175 **176**

Scheme 3.4

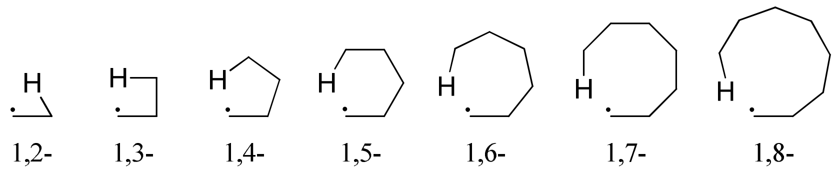
Other examples broadly similar to those shown above include Robertson's synthesis of pyrrolizidines[74] and our group's work on *spiro* acetals.[75] Chatgilialoglu reported a particularly elegant application of this methodology for the synthesis of *spiro* nucleosides (Scheme 3.5).[76] In this case 1,5-abstraction is followed by *5-endo-trig* cyclisation and loss of a bromine radical to give **178** and **179** still possessing a double bond.



177 **178** **179**

Scheme 3.5

In all of the examples shown above, the hydrogen atom is abstracted in 1,5 mode, but other transformations are also possible. The groups of Lendvay[77] and Dannenberg[78] have published theoretical studies on the activation energy required for 1,*x*-H abstractions over various different chain lengths (Table 3.1). These calculations are based on simple hydrocarbon chains and the situation is very different when bulky substituents are present. Nevertheless, it is interesting to note that in terms of activation energies 1,5-H abstraction is the most favourable, closely followed by 1,6-H.

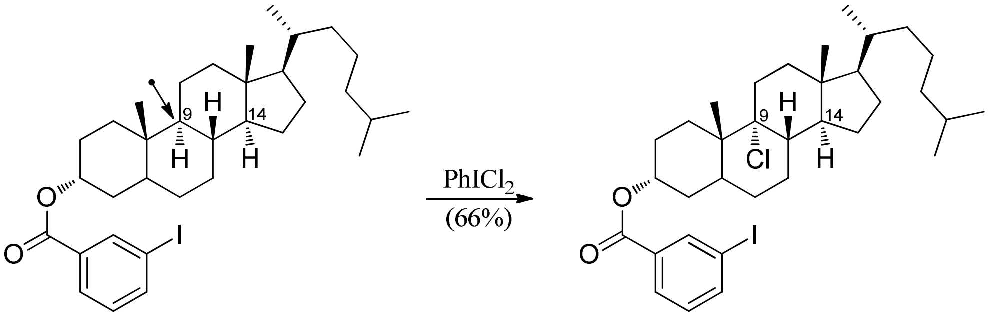


	activation energy		**bond angle**
transformation	**kcal/mol**[77]	**kcal/mol**[78]	**(C-H--C)**[78]
1,2-	41.1	-	-
1,3-	41.6	42.5	98.5
1,4-	24.6	27.5	125.2
1,5-	17.2	20.9	144.5
1,6-	-	21.0	158.0
1,7-	-	23.0	163.6
1,8-	-	24.9	178.1

Table 3.1

Table 3.1 also shows the effect of chain length on the C-H--C bond angle of the transition state. While the stereoelectronic preference is for an angle of as close to 180° as possible, Dannenberg calculated that a distortion of 35° has less than a 1 kcal/mol effect on the activation energy. As shown, the 6-membered transition state necessary for a 1,5-abstraction has a C-H--C bond angle of 145° so suffers no severe enthalpic penalty. Abstractions over shorter distances (1,2-H, 1,3-H, 1,4-H) result in an increase in ring strain and therefore higher activation energies.

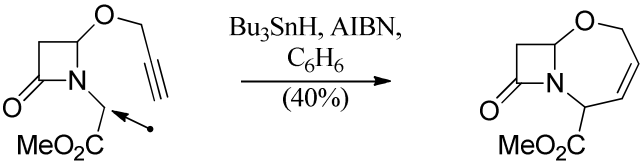
However, as well as enthalpic considerations, the conformation of the molecule can be very important in determining which hydrogen is transferred. An example of this is provided by Breslow's classic work on the remote functionalisation of steroids. In the example shown in Scheme 3.6 a radical generated by complexation of a chlorine species with the aryl iodide abstracts a hydrogen in 1,11-mode from C-9 to give the haloalkane **181** in good yield.[79] This selectivity is entirely due to the shape of the molecule – extending the linker to the aryl iodide by one carbon completely changes the selectivity to C-14.



180 **181**

Scheme 3.6

Another interesting example is Bosch and Bachi's synthesis of bicyclic β-lactams where both 1,5- and 1,6-abstraction are possible but the strained nature of the four membered ring helps favour the 1,6-H transfer. Cyclisation in *7-endo-trig* mode is then followed by expulsion of a stannane radical, leaving a double bond in the product (**183**) and making the reaction catalytic in tributyltin hydride. (Scheme 3.7).[80]



182 **183**

Scheme 3.7

In this example it is also worth noting that the intermediate radical SOMO (singly occupied molecular orbital) is stabilised by a one-electron interaction with the carbonyl LUMO (lowest unoccupied molecular orbital) as well as a three-electron interaction with the nitrogen HOMO (highest occupied molecular orbital). This demonstrates an unusual property of radicals – they are stabilised by both electron-releasing and electron-withdrawing groups as shown in Figure 3.1.[81] Radicals positioned adjacent to one of each of these are known as capto-dative radicals and are highly stabilised.

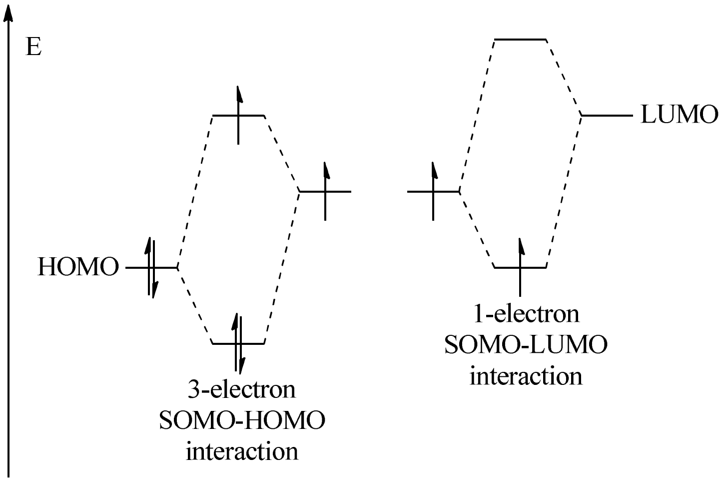
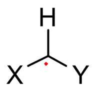


Figure 3.1

The extent of this stabilisation has been the subject of a number of investigations. For example, Rüchardt and co-workers used electron spin resonance (ESR) spectroscopy to analyse the energy differences for radicals next to various substituents and quantified these as radical stabilisation enthalpies (RSEs, Table 3.2).[82] Their work demonstrates that radicals flanked by both a carbonyl group and a nitrogen are considerably more stabilised than those by a carbonyl or nitrogen only (entry 10 vs. entries 5 and 3 respectively). Again, these are simple systems and the effect is likely to be less pronounced in the amide variations of these functionalities.

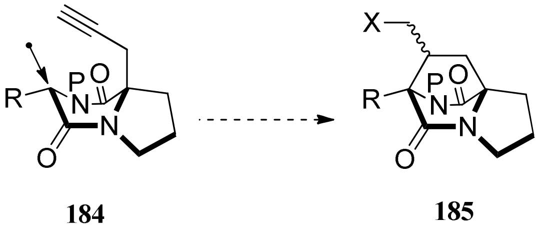


entry	X	Y	RSE (kcal mol^{-1})
1	C	C	≡ 0
2	C	COOR	−2.8
3	**C**	**NR_2**	**−3.9**
4	C	OR	−5.9
5	**C**	**CO**	**−6.0**
6	CO	CO	−8.1
7	C	Ph	−8.4
8	Ph	Ph	−12.9
9	COOR	NR_2	−14.4
10	**CO**	**NR_2**	**−20.7**

Table 3.2

A number of other groups have published theoretical investigations into this effect with broadly the same conclusions.[83]

To investigate the conformational and stereoelectronic effects in our proposed synthesis, we designed a model system comprised of a proline-fused DKP possessing a propargyl unit (**184**, Scheme 3.8).



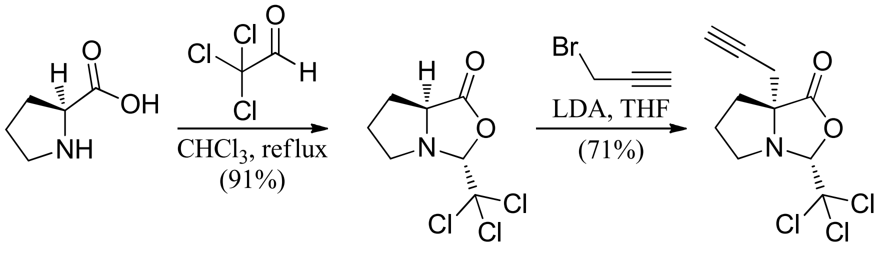
Scheme 3.8

We hoped that radical addition to the triple bond would be followed by 1,6-H transfer and *6-exo-trig* cyclisation to afford the bridged bicyclic DKP **185**. Importantly, the intermediate radical would be capto-dative; and molecular models suggest that the conformation should favour this process rather than the potential 1,5-H abstraction alpha to the pyrrolidine nitrogen.

At this stage, it is important to note that we have used the cheaper L-proline for all of our work so the compounds in Scheme 3.8 are of the opposite enantiomeric series to the natural products shown in Chapter 1. Of course, all of the chemistry we have developed should be equally applicable to the D-proline series.

3.3 Mixed Proline-Glycine Model Systems

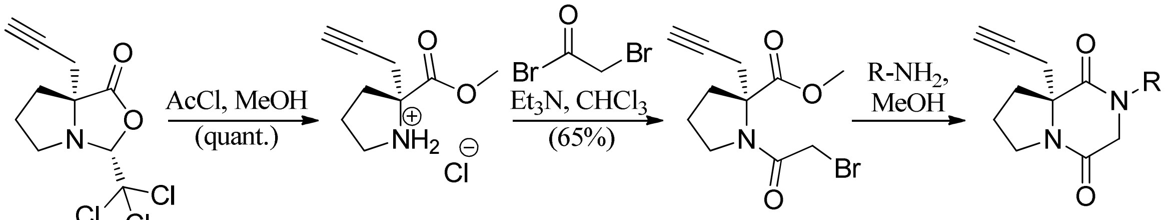
To synthesise our model system as a single enantiomer, we opted to use Seebach's method for the self-reproduction of chirality (Scheme 3.9).[84] The original sequence of reactions was discussed in Chapter 2 in the context of Williams' synthetic studies. In 1999, Wang and Germanas reported a modification of this procedure employing chloral (trichloroacetaldehyde) in place of pivalaldehyde.[85] More recently, Amedjkouh and Ahlberg published an improvement to this method increasing the yield of **186** to 91% (Scheme 3.9).[86] This method is advantageous in that the resulting oxazolidinone is a stable crystalline solid that can be stored at room temperature without degradation; and chloral is considerably cheaper than pivalaldehyde. Cordero and co-workers used this methodology for the synthesis of propargylated oxazolidinone **187**, albeit in poor yield (24% based on 63% conversion).[87]



186 **187**

Scheme 3.9

Carrying out brief optimisation studies, we were able to improve this yield to 71% by using a large excess (five equivalents) of propargyl bromide and keeping the reaction at −78 °C until total comsumption of oxazolidinone **186**. Opening of the oxazolidinone ring was achieved using acetyl chloride in refluxing methanol to afford substituted proline **188** in quantitative yield (Scheme 3.10).[88]

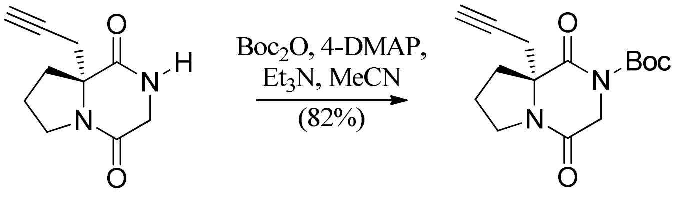


187 **188** **189**

190a R = H (quant.)
190b R = Me (93%)
190c R = PMB (89%)

Scheme 3.10

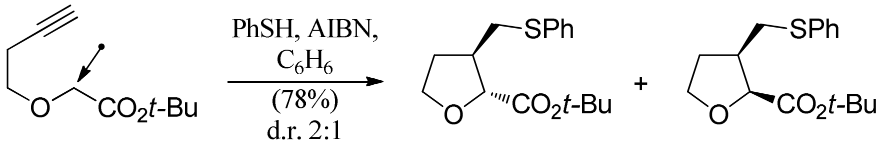
This hydrochloride salt was then treated with bromoacetyl bromide and triethylamine to afford the acylation product **189**. Formation of the DKPs **190a-c** was accomplished smoothly by treatment with ammonia, methylamine or *para*-methoxybenzylamine respectively. Exposure of **189** to the less nucleophilic *tert*-butyl carbamate failed to furnish the DKP, so the Boc group was instead introduced by treating **190a** with Boc$_2$O in the presence of 4-DMAP and triethylamine to yield **190d** (Scheme 3.11).



190a **190d**

Scheme 3.11

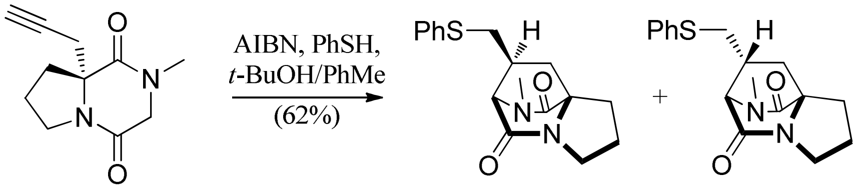
With DKPs **190a-d** in hand, we were keen to test our key radical cyclisation. While a large body of work describes the use of tributyltin hydride in radical reactions, we were attracted to reports by Burke and co-workers describing the addition of sulfur to a triple bond (Scheme 3.12).[89] The use of thiophenol avoids toxic tin derivatives and introduces the possibility of further functionalisation by oxidation of the sulfide.[90] In this example the first step is the addition of a sulfur centred radical to the triple bond of **191** followed by 1,5-H transfer and *5-exo-trig* cyclisation to give a 2:1 mixture of **192a** and **192b**.



191 **192a** **192b**

Scheme 3.12

Following a more general approach described by the group of Renaud,[91] a refluxing solution of the DKP **190b** in *tert*-butanol was treated with solutions of thiophenol and AIBN, each in toluene. This addition took place over twenty hours via syringe pump and we were delighted to find that we obtained the desired compounds **193a** and **193b** in 62% yield as a 5:4 mixture of inseparable diastereoisomers (Scheme 3.13).



190b **193a** **193b**

Scheme 3.13

Detailed NMR analysis allowed us to identify the signals corresponding to each diastereomer, but determining the stereochemistry was somewhat problematic. Figure 3.2 shows a section of a ^1H NMR spectrum for the mixture of diastereomers along with two nOe experiments. As shown, the crucial H-6 is poorly resolved presenting a broad multiplet corresponding to both diastereomers. Selectively exciting the signal for the *N*-methyl group of the minor component shows no nOe correlation to H-6 suggesting a *syn* relationship between this proton and the pyrrolidine ring (blue spectrum). Carrying out the same experiment with the major diastereomer shows a good correlation between H-6 and the *N*-methyl (red spectrum).

However, in this case we were not able to selectively excite only the peak corresponding to H-10 leaving this assignment somewhat ambiguous.

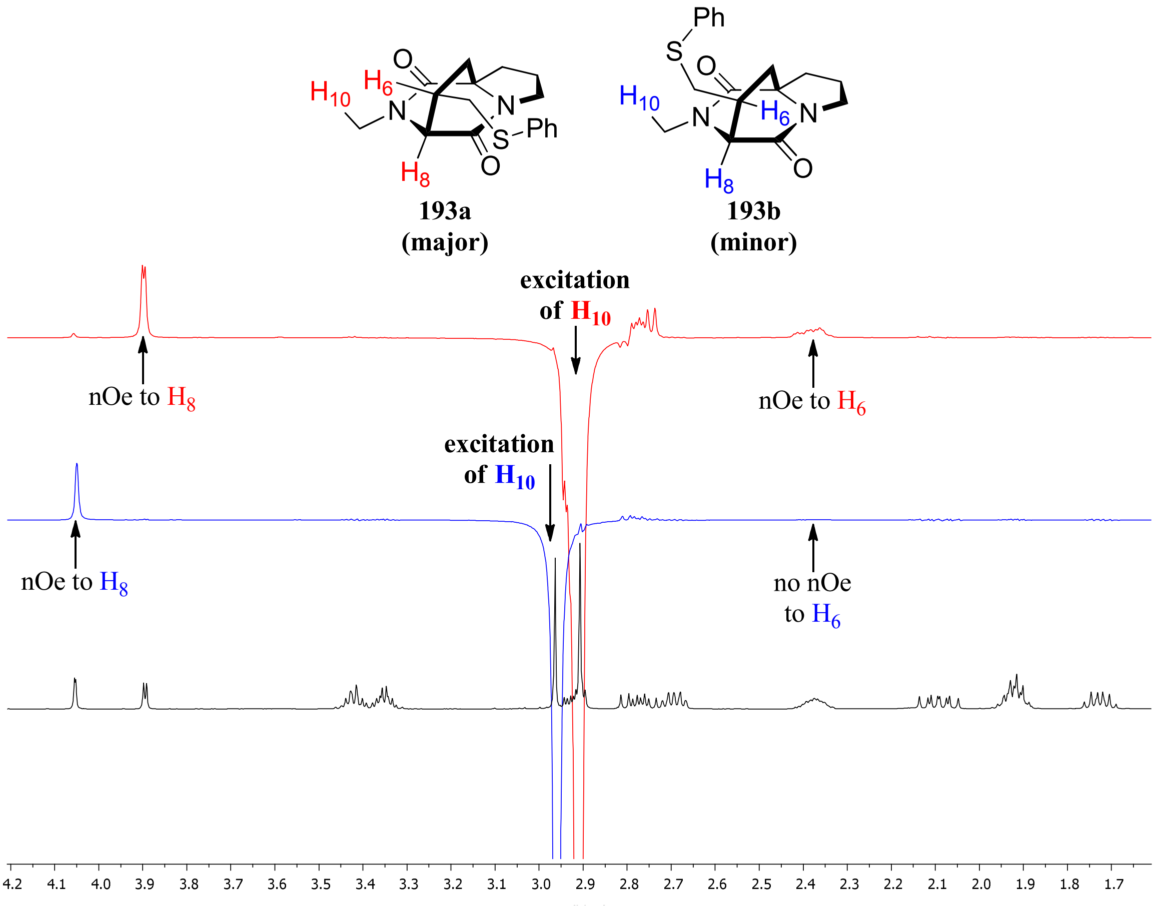
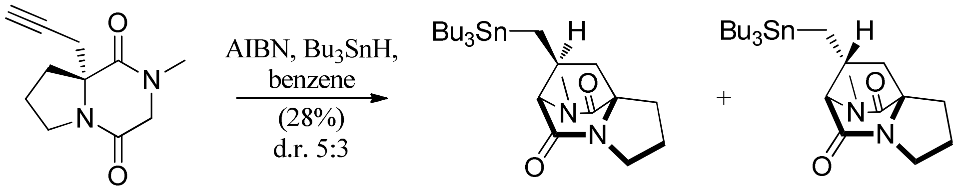


Figure 3.2

Although we were pleased with our success using thiophenol, we were also interested in other methods for the initiation of the radical process. While all of our attempts using Renaud's dialkyl phosphite reagents failed,[92] we were able to effect cyclisation of **190b** using the classic conditions of tributyltin hydride and AIBN.[80] This afforded the bridged bicycles **194a** and **194b** in 28% yield as a 5:3 mixture of diastereomers (Scheme 3.14).



Scheme 3.14

In this case determination of the stereochemistry was more straightforward. While the signal corresponding to H-6 was still poorly resolved, we were easily able to selectively excite both *N*-methyl groups as shown in Figure 3.3. Excitation of H-10 of the major component showed a nOe correlation with H-6 (red spectrum) while excitation of H-10 of the minor component did not (blue spectrum). These experiments demonstrate that the major isomer (**194a**) has an *anti* relationship between H-6 and the pyrrolidine ring – the same as that found in the majority of natural products.

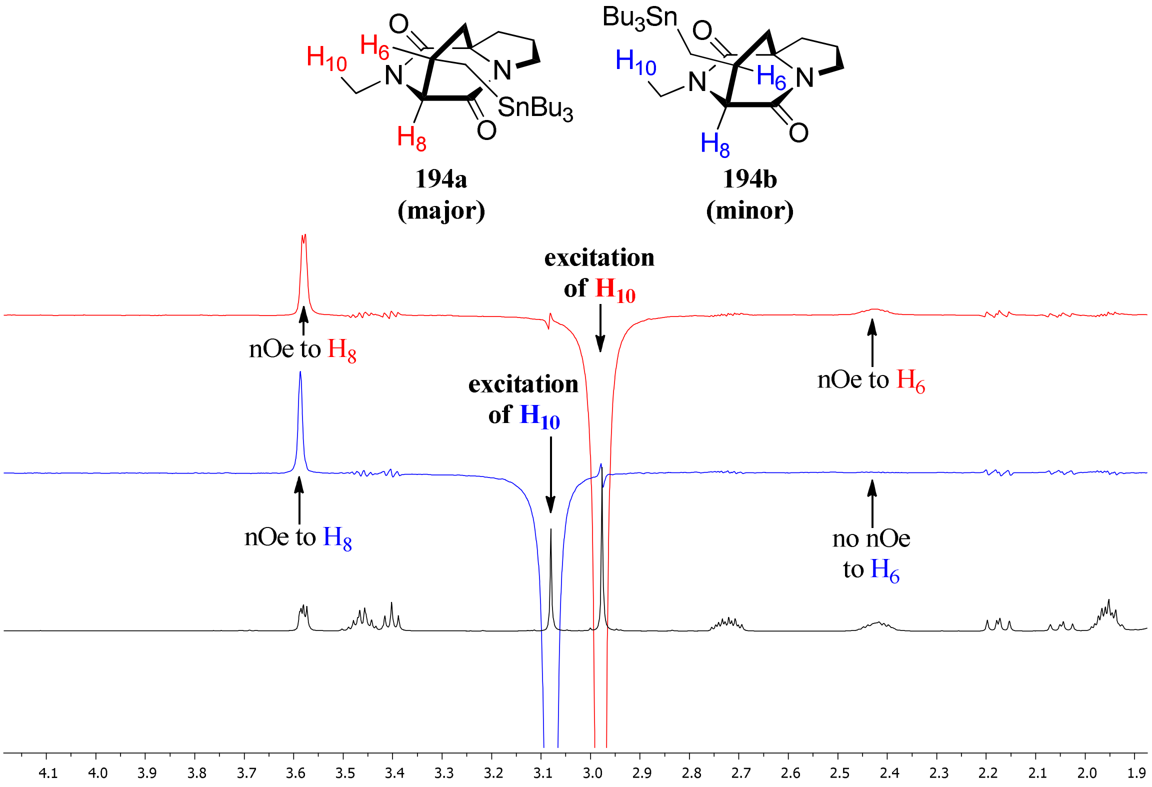
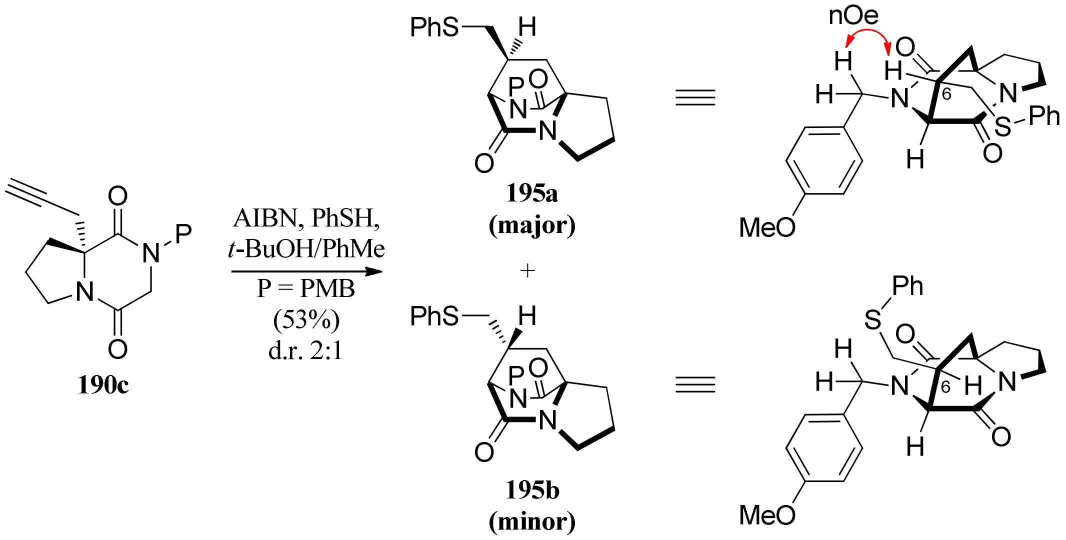


Figure 3.3

As well as securing the configuration of these compounds, these experiments lend weight to our initial assignment of *N*-methyl compounds **193a** and **193b** by demonstrating that there is a strong nOe from the *N*-methyl to H-6 in only one of the diastereomers.

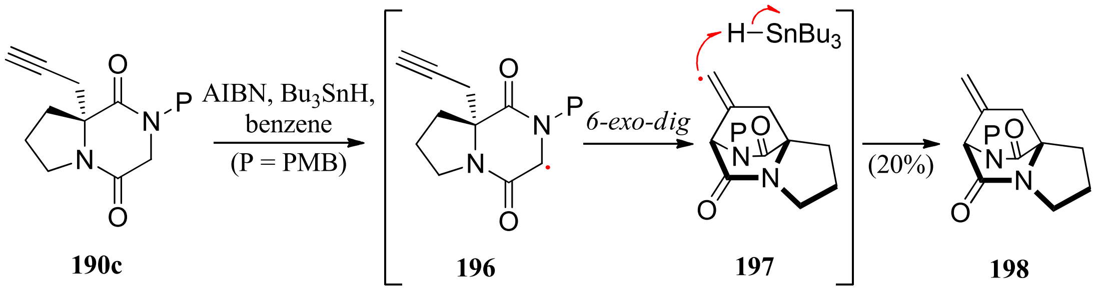
Having successfully cyclised *N*-Me DKP **190b**, we were interested in the effect the *N*-protecting group would have on the stereochemical outcome of the reaction. Thus *N*-PMB derivative **190c** was treated with AIBN and thiophenol using the same conditions as described above to give the desired compounds **195a** and **195b** in 53% yield. We were pleased to find that in this case the bridged bicycle was isolated as a 2:1 mixture of diastereomers. Again, nOe experiments showed that the major diastereomer has an *anti* relationship between H-6 (brevianamide numbering) and the pyrrolidine ring (Scheme 3.15).



Scheme 3.15

While the *N*-PMB derivative gave an improvement in the diastereomeric ratio, previous work both in our group[66b] and by Williams[45, 93] has identified difficulty in removing this group at the end of the synthesis. Therefore, we did not plan to use this protecting group in more complex systems, but we again attempted this cyclisation using a tin-centred radical.

However, in this case we isolated compound **198** possessing an *exo*-methylene motif in 20% yield (Scheme 3.16). Presumably this comes about from *inter*molecular hydrogen atom abstraction from the captodative position of DKP **190c** followed by *6-exo-dig* cyclisation and reaction with tributyltin hydride to give **198**. We are unsure why this only happens when the PMB protecting group is present, but perhaps a steric or electronic interaction with the triple bond slows down the addition of a stannane radical enough to allow this alternative process.

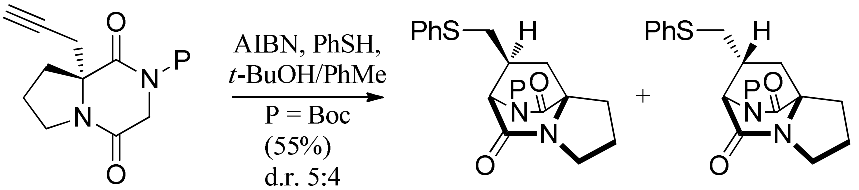


190c **196** **197** **198**

Scheme 3.16

This example along with the corresponding tin mediated reaction with *N*-methyl DKP **190b** demonstrate that tributyltin hydride can be used for our cyclisation – albeit with somewhat unpredictable results. As the yields are disappointing and there is little change in the stereoselectivity, we have used thiophenol/AIBN in all subsequent work with acetylene units.

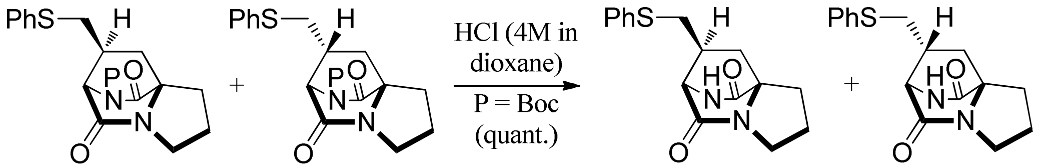
The last of our simple model systems, **190d**, has an *N*-Boc protecting group. We hoped that this would be much easier to remove than a PMB group but display some similar characteristics in terms of controlling the diastereoselectivity. As in the previous examples, treatment with AIBN and thiophenol furnished the desired bridged DKPs **199a** and **199b** in reasonable yield (Scheme 3.17). The diastereomeric ratio was again 5:4, but determining the configuration of each epimer proved more difficult. The only protons on the Boc group are positioned some distance from the DKP so no nOe was observed in either diastereomer.



190d **199a** **199b**

Scheme 3.17

After studying molecular models, we reasoned that the conversion to the corresponding N-H DKP might aid our assignment. Thus, **199a** and **199b** were treated with HCl in dioxane resulting in clean Boc removal to afford **200a** and **200b** (Scheme 3.18). While this served to demonstrate the ease of removal of the Boc group, unfortunately it was of no help in determining the stereochemistry at the vital C-6 position.

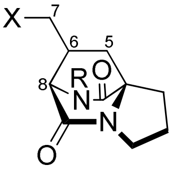


199a **199b** **200a** **200b**

Scheme 3.18

Having had no success with nOe experiments, we turned our attention to the ^{1}H NMR spectra of our previous examples. In all cases, the signals for H-8 in the major and minor epimers are well resolved doublets separated by between 0.01 and 0.16 ppm. As summarised in entries 1 to 6 of Table 3.3, we found that the signal corresponding to a *syn* relationship between H-6 and the pyrrolidine ring is slightly further downfield and has a smaller coupling constant (J = 1.4 – 1.5 Hz) than the *anti* diastereomer (J = 3.1 – 3.5 Hz). Comparing this with the data for *N*-Boc compounds **199a** and **199b** (entries 7 and 8) we observed the same relationship so have tentatively assigned the 5:4 diastereomeric ratio as favouring the *anti* epimer. In an attempt to confirm this, we analysed the other protons close to the stereocentre.

Unfortunately, the signals for the H-6 and H-7 are generally poorly resolved so we found it difficult to reliably assign exact chemical shifts for each epimer. However, one of the H-5 protons is a well resolved doubled doublet at around 2 ppm. We found that in all cases the *anti* proton was slightly downfield of the corresponding *syn* signal – and this relationship was maintained in the *N*-Boc series.



					H-8 (d)		H-5 (dd)	
entry	**compound**	**R**	**X**	***syn*/*anti***	**ppm**	***J* (Hz)**	**ppm**	***J* (Hz)**
1	**194a**	Me	$SnBu_3$	*anti*	3.58	3.5	2.18	12.8, 9.6
2	**194b**	Me	$SnBu_3$	*syn*	3.59	1.4	2.05	12.9, 9.5
3	**193a**	Me	SPh	*anti*	3.90	3.3	2.12	12.6, 9.1
4	**193b**	Me	SPh	*syn*	4.06	1.4	2.08	12.6, 8.9
5	**195a**	PMB	SPh	*anti*	4.04	3.1	2.15	12.7, 9.6
6	**195b**	PMB	SPh	*syn*	4.10	1.4	2.10	12.2, 8.4
7	**199a**	Boc	SPh	*anti*	5.03	3.3	2.21	13.8, 9.8
8	**199b**	Boc	SPh	*syn*	5.05	1.5	2.12	13.9, 10.3

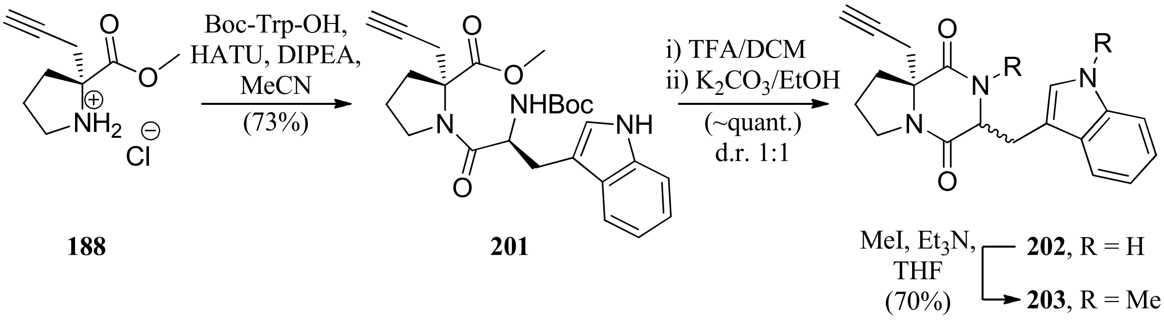
Table 3.3

Taken together, these results show that our key cyclisation works for a simple model system in reasonable yield. While the stereocontrol is relatively poor, we hoped that the extra steric bulk present in more complex systems would lead to an improvement in this.

3.4 Other Mixed DKPs

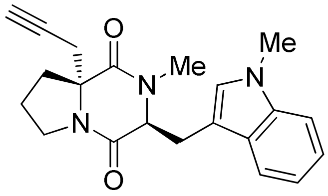
Having demonstrated our key cyclisation on simple proline-glycine DKPs, we turned our attention to mixed DKPs made up of proline along with various other amino acids. We hoped that by incorporating an indole motif we would include a radical trap suitable for a second cyclisation to occur. Thus, substituted proline **188** was coupled with Boc-tryptophan using HATU in the presence of Hünig's base to afford dipeptide **201** in 73% yield (Scheme

3.19).[94] Cleavage of the Boc group was accomplished using TFA and the resulting free amine underwent ring closure by heating with potassium carbonate to give DKP **202**.[95]



Scheme 3.19

Unfortunately the use of such harsh basic conditions resulted in epimerisation at the tryptophan moiety leading to a ca. 1:1 mixture of diastereomers. As we required a single diastereomer for our subsequent radical cyclisation to be successful, we needed to separate this mixture. Exposure of **202** to triethylamine and methyl iodide led to smooth methylation of both the DKP and the indole to afford **203**. We were then able to separate the two diastereomers by preparative HPLC to afford the desired cyclisation precursor and confirm the structure by X-ray crystallography (Figure 3.4).



203

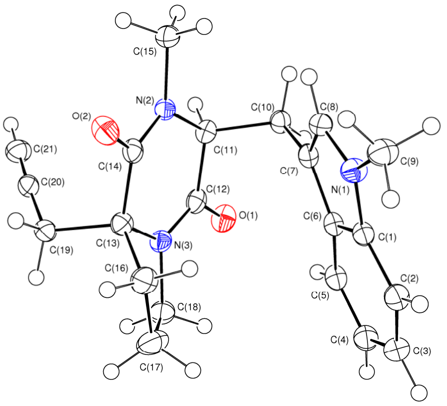
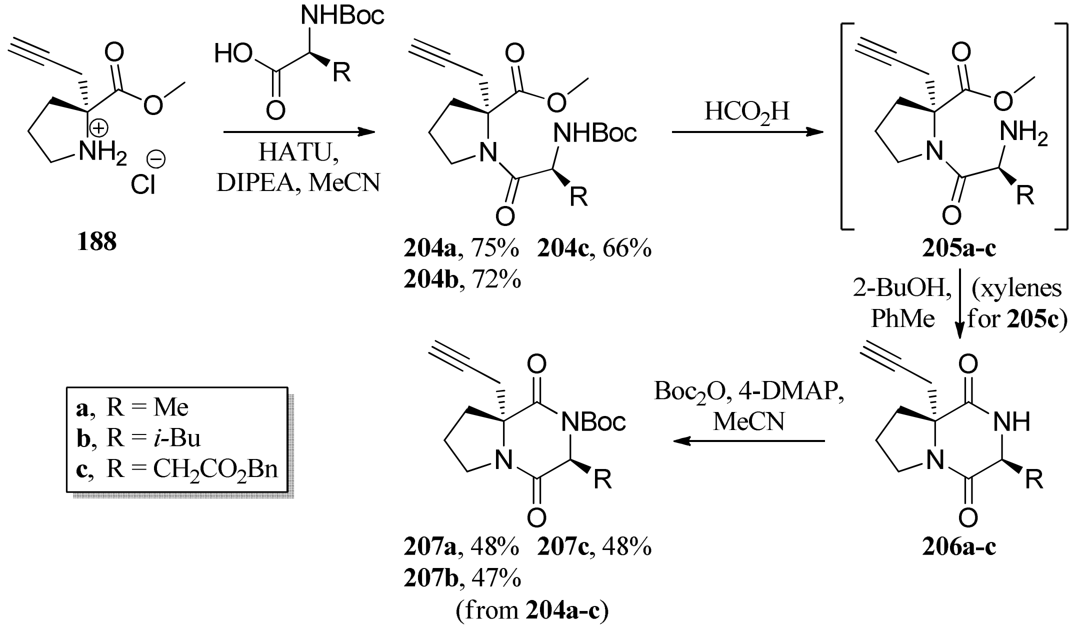


Figure 3.4

With DKP **203** in hand, we turned to our established optimal conditions for the radical cyclisation (thiophenol/AIBN, syringe pump addition). This example is, of course, considerably more complicated that those discussed above and a number of products are possible. We presume that the first cyclisation (analogous to our earlier results) should lead to the formation of two diastereomers. These could each then generate two regioisomers by attack on the indole either in *5-exo-trig* or *6-endo-trig* mode. The regioisomeric products could occur as a mixture of diastereoisomers, potentially leading to a very complex mixture – which is what we in fact observed. While analysis by mass spectrometry showed the correct molecular weight, we were disappointed to find that despite our best efforts we were unable to isolate any pure compounds.

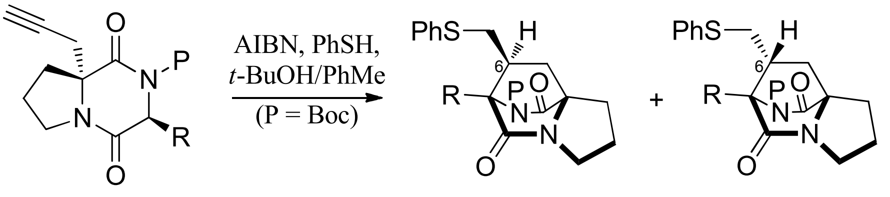
Taking a step back, we turned our attention to slightly simpler mixed DKPs incorporating proline along with a substituted but non-participating amino acid. Again, HATU and Hünig's base mediated coupling afforded dipeptides **204a-c** in good yield incorporating an alanine, leucine or a protected aspartic acid residue (Scheme 3.20). To avoid the epimerisation we had witnessed with the Pro-Trp series, we adopted a different approach for the DKP formation.[96] Cleavage of the Boc group was accomplished with neat formic acid which was then removed *in vacuo*. For the alanine and leucine derived systems DKP formation was achieved by heating the primary amine in a mixture of 2-butanol and toluene. The dipeptide derived from aspartic acid (**205c**) would not react under these conditions, presumably as it is more sterically hindered. However, we found that using xylenes and employing a higher temperature led to formation of the desired DKP **206c**. To avoid the use of strong base we decided to use a Boc protecting group that could be introduced under mild conditions. Thus treatment of DKPs

206a-c with Boc anhydride in the presence of 4-DMAP gave **207a-c** ready for our cyclisation reactions.



Scheme 3.20

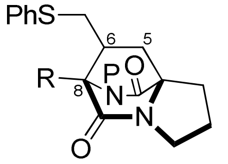
To initiate our radical cyclisation we again employed Renaud's conditions using thiophenol and AIBN and were delighted to find that alanine derived DKP **207a** formed the bridged bicycles **208a** and **208b** in near-quantitative yield as a 2:1 mixture of diastereomers (Table 3.4). The proline-leucine DKP **207b** gave **209a** and **209b** as a 4:1 mixture of epimers again in excellent yield. Finally, the aspartic acid derived system **207c** cyclised to furnish **210a** and **210b** in 73% yield and with a d.r. of 2:1.



entry	compounds	R	% yield	d.r.
1	**208a/208b**	Me	98	2:1
2	**209a/209b**	*i*-Bu	96	4:1
3	**210a/210b**	CH_2CO_2Bn	73	2:1

Table 3.4

While we were delighted by the improved yields, we found it difficult to conclusively determine the stereochemistry of each epimer. As with the proline-glycine system we observed no nOe from the *N*-Boc group to H-6 in either diastereomer. Moreover, we were unable to use the chemical shift and coupling constant of the bridgehead proton as it has been replaced by a carbon in these examples. The only other proton for which we had previously observed a useful difference was H-5, where the signal corresponding to the *anti* epimer was always shifted slightly further downfield than the *syn* signal. In all of the examples in Table 3.4 the H-5 signal of the major diastereomer is downfield of the minor component (Table 3.5). While this is consistent with the major epimer having the desired *anti* relationship between H-6 and the pyrrolidine, it should not be considered as conclusive proof.



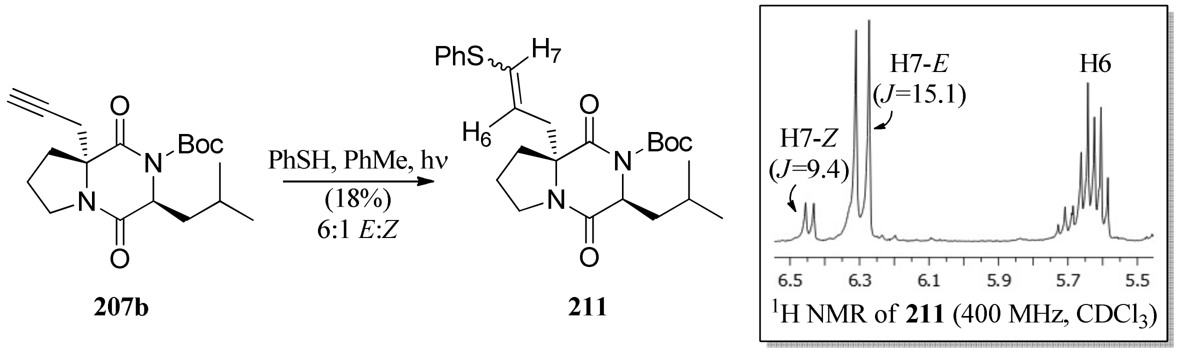
(P = Boc)

compounds	R		H-5 (dd) ppm	H-5 (dd) *J* (Hz)
208a/208b	Me	major	2.24	13.8, 9.6
		minor	2.15	13.2, 9.8
209a/209b	*i*-Bu	major	2.24	13.8, 9.4
		minor	2.10	13.5, 8.8
210a/210b	CH_2CO_2Bn	major	2.28	13.8, 9.7
		minor	2.15	13.5, 9.9

Table 3.5

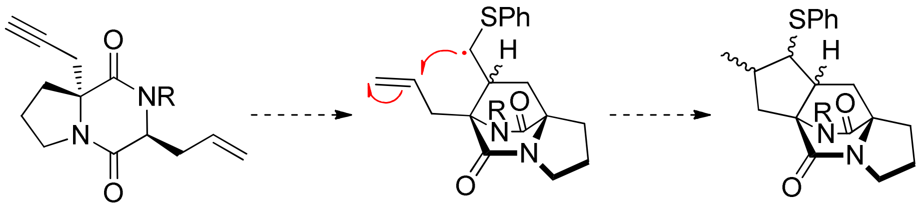
Nevertheless, as all of the examples of this bond formation using *N*-acyliminium chemistry,[66, 96] IMDA cycloaddition,[50, 53, 55-56, 97] S_N2' reactions[45, 47-49] and our earlier radical cyclisations (*vide supra*) have resulted in predominantly the *anti* isomer, we were reasonably confident that these systems would show the same bias.

We wondered if we could improve the ratio of epimers by carrying out the reaction at a lower temperature. In our first attempt, we used a 300 W sunlamp to irradiate a solution of DKP **207b** in toluene with the temperature maintained below 35 °C.[98] As before, thiophenol and AIBN were added via syringe pump over twenty hours. While we were able to isolate bridged bicycles **209a** and **209b** in 28% yield, the stereoselectivity was not improved from our previous attempts and some starting material remained. Using slightly different conditions we exposed a solution of DKP **207b** and thiophenol to a 400 W mercury lamp.[99] Again, the temperature was maintained below 35 °C and again the reaction did not go to completion. This time though, the bridged DKP was not formed and instead we isolated 18% of vinyl sulfide **211** as a 6:1 *E*:*Z* mixture (Scheme 3.21). Presumably this compound arises from addition of a sulfur radical to the triple bond followed by either a failure of the 1,6-H abstraction step or the subsequent *6-exo-trig* cyclisation.



Scheme 3.21

While we were somewhat disappointed at being unable to improve the diastereoselectivity of this reaction, further investigation on such a simple model system was considered a low priority. With this in mind we considered a more complex DKP (**212**) substituted with an allyl group to allow a second cyclisation to occur (Scheme 3.22).

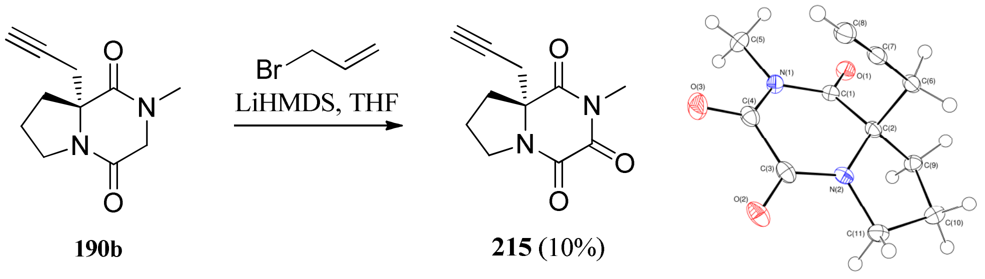


212 **213** **214**

Scheme 3.22

We hoped that treatment of **212** with our established cyclisation conditions would, as before, afford radical intermediate **213**. Instead of terminating the chain process by picking up a hydrogen, this radical would be able to cyclise in *5-exo-trig* mode to give tetracycle **214** as a simplified model of our real system.

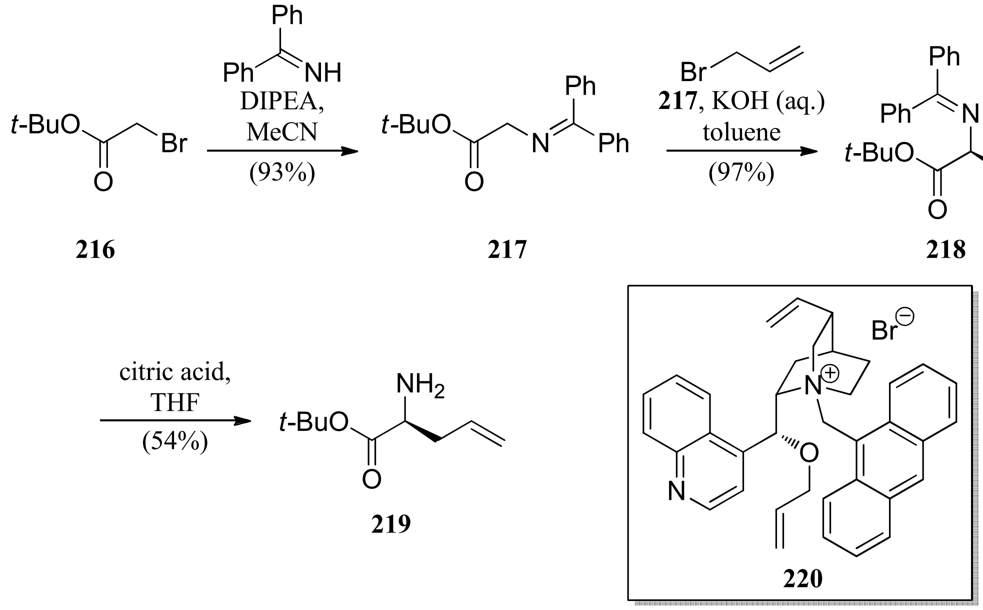
To obtain allyl DKP **212** we first considered deprotonation of a simple Pro-Gly mixed DKP followed by electrophilic quench with allyl bromide (Scheme 3.23). Thus, DKP **190b** was treated with LiHMDS followed by allyl bromide. Unfortunately we did not observe any of the desired product and instead isolated triketopiperazine (TKP) **215** in low yield along with starting material. We were able to confirm the structure unambiguously by X-ray crystallography. This presumably arises through either direct radical oxidation at the capto-dative position of the DKP or oxidation of the intermediate enolate. The synthesis of TKPs is relatively unexplored[100] and oxidation of DKPs could provide a route to single enantiomers of these interesting compounds although we have not investigated this any further at present.



190b **215** (10%)

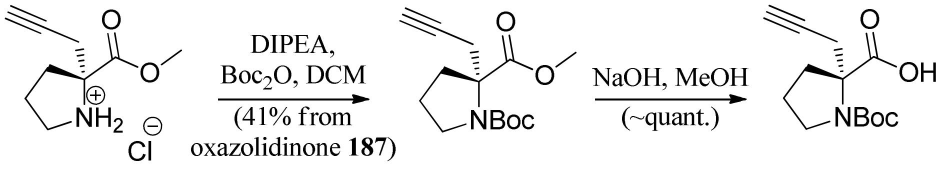
Scheme 3.23

To circumvent this problem we set about the synthesis of an allyl glycine derivative suitable for coupling with a substituted proline residue. To do this we used *Cinchona* alkaloid **220** as a phase transfer catalyst in chemistry that has been developed by Lygo[101] and Corey.[102] Known glycine imine **217** was prepared by a simple procedure using benzophenone imine and *tert*-butyl bromoacetate (Scheme 3.24).[103] Treatment with allyl bromide in the presence of the *Cinchona* alkaloid **220**, aqueous sodium hydroxide and toluene then afforded protected allylglycine **218** in 97% yield. While we did not determine the ee by chiral HPLC, we reasoned that once we had coupled this piece with another amino acid we would be able to indirectly determine the ee by analysis of the diastereomeric ratio. Continuing the synthesis, we were unable to selectively cleave the *tert*-butyl ester under a variety of conditions. Instead, imine hydrolysis was accomplished by treatment with citric acid in THF to afford amine **219** which we were able to prepare on a multi-gram scale.



Scheme 3.24

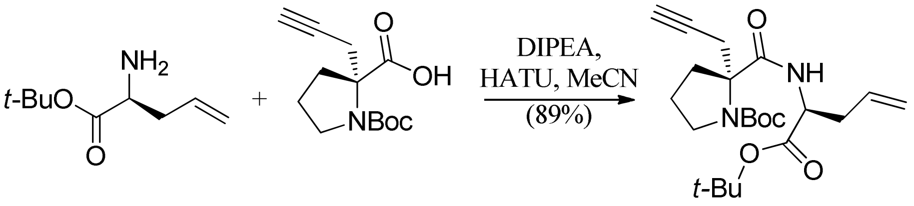
With amine **219** in hand we turned our attention to the synthesis of a suitably protected proline as a coupling partner. Thus, previously prepared substituted proline **188** was Boc protected in the presence of Hünig's base before ester hydrolysis under standard conditions afforded carboxylic acid **222** in good yield.



188 **221** **222**

Scheme 3.25

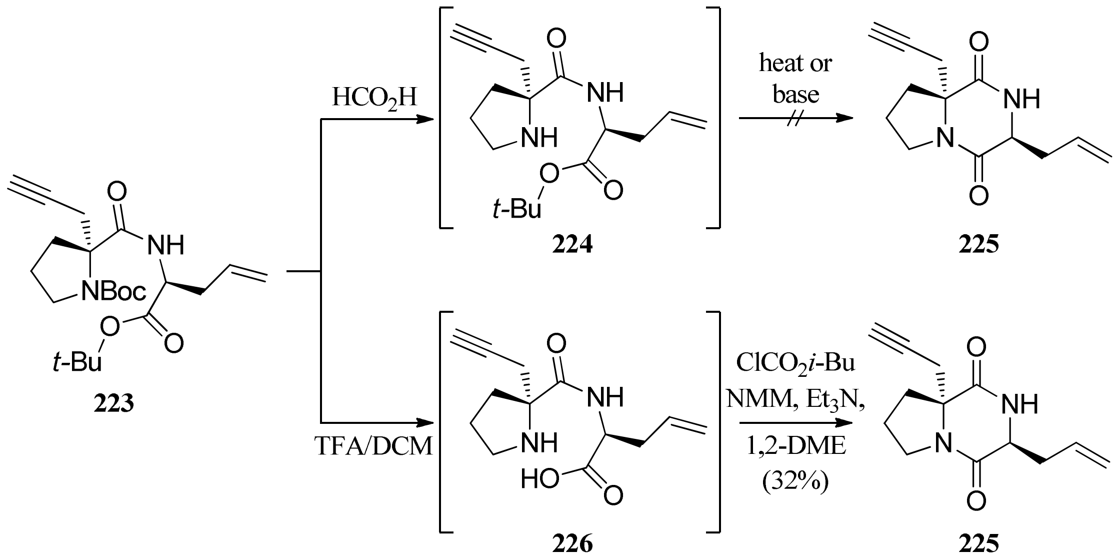
Exposing a mixture of proline **222** and allylglycine **219** to the same peptide coupling conditions we had used previously, we were pleased to obtain dipeptide **223** in 89% yield (Scheme 3.26). This is somewhat more efficient than the couplings detailed above, possibly because in this example we have used a primary amine rather than a sterically hindered pyrrolidine.[104]



219 **222** **223**

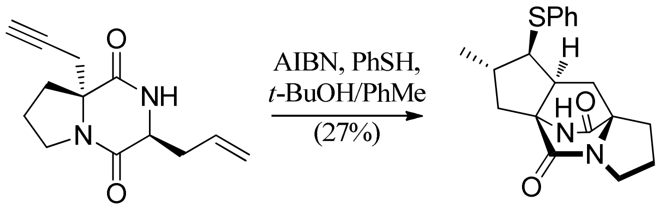
Scheme 3.26

At this stage, we hoped to effect DKP formation by Boc cleavage followed by thermal cyclisation as we had carried out before. Treatment of **223** with formic acid resulted in smooth Boc deprotection to give the corresponding secondary amine **224** with the *tert*-butyl ester remaining intact (Scheme 3.27). However, heating this compound either neat or in a range of solvents failed to provide any of the desired allyl DKP **225**. Despite having concerns of causing epimerisation we also screened a number of both organic and inorganic bases, again without any useful reaction occurring.



Scheme 3.27

Trying another approach, we treated dipeptide **223** with TFA in DCM to cause both Boc deprotection and hydrolysis of the ester. Hoping to effect DKP formation via a conventional peptide coupling reaction, we treated **226** with a number of common reagents such as DCC, HATU and 4-DMAP, but without any success. In the end we managed to obtain DKP **225**, albeit in poor yield, by formation of a mixed anhydride followed by amide bond construction. Presumably the low yield is due to a fast attack on *iso*-butyl chloroformate by the pyrrolidine amine preventing DKP formation from occurring. We were surprised to find that **225** resisted all attempts of amide Boc protection with recovery of unreacted starting material in all cases. Although this was frustrating, we reasoned that the desired radical cyclisation should not be adversely affected by a free N-H. Thus, following our standard procedure, AIBN and thiophenol were slowly added to a solution of DKP **225** in *tert*-butanol. As this reaction constructs two carbon-carbon bonds along with three new stereocentres we anticipated a complex mixture of products. While this was indeed the case, we were able to isolate the desired tetracycle **227** in 27% yield (Scheme 3.28).



225 **227**

Scheme 3.28

We were able to determine the stereochemistry of **227** using nOe experiments. A correlation between the N-H and one H-10 allowed us to differentiate between H-10a and H-10b. As shown in Figure 3.5 we were able to use this to establish the stereochemistry at C-6, C-11 and C-12. An nOe between H-10b and H-6 shows an *anti* relationship between the pyrrolidine nitrogen and the vital H-6. Another correlation demonstrates that H-6, H-10b and H-12 are all on the same side of the cyclopentane ring. Finally, an nOe between H-10a and H-11 shows these protons are on the same side as each other – i.e. the opposite side to H-6.

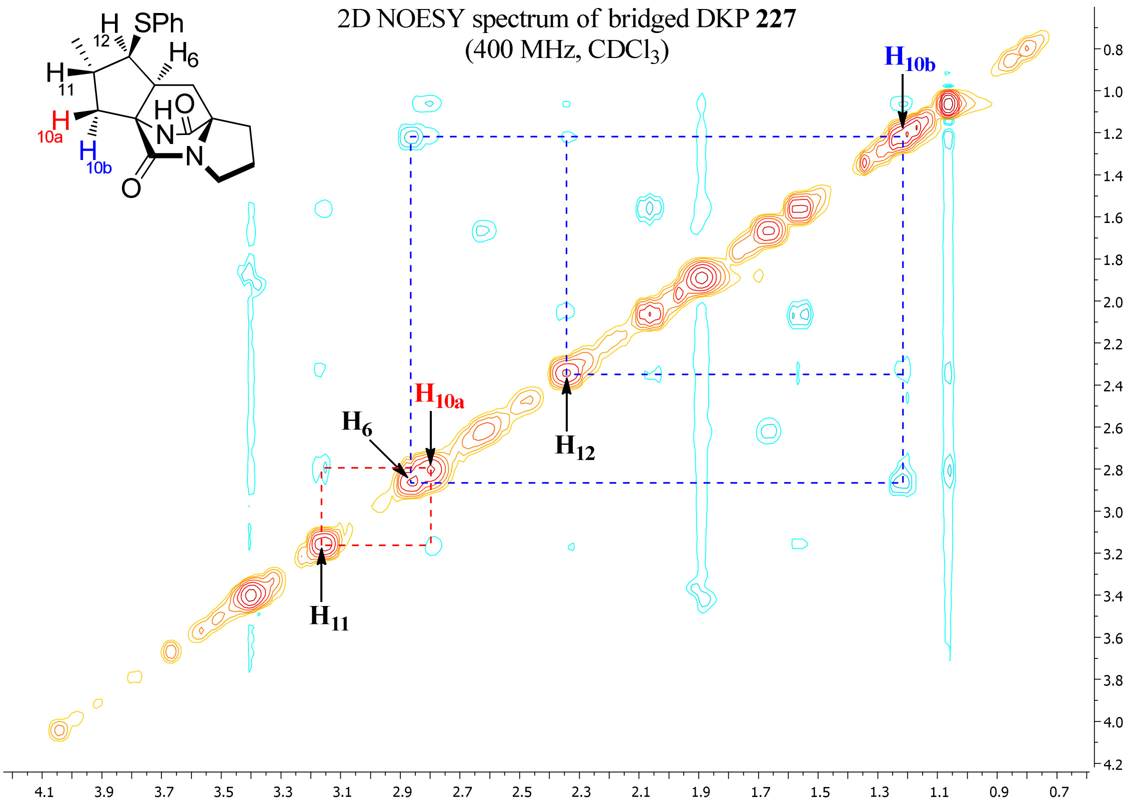


Figure 3.5

While the yield of this reaction was somewhat disappointing, we were pleased that the major component had the desired stereochemistry at C-6. This was also the first demonstration of a complete radical cascade analogous to our proposed synthesis of asperparaline C.

3.5 Summary

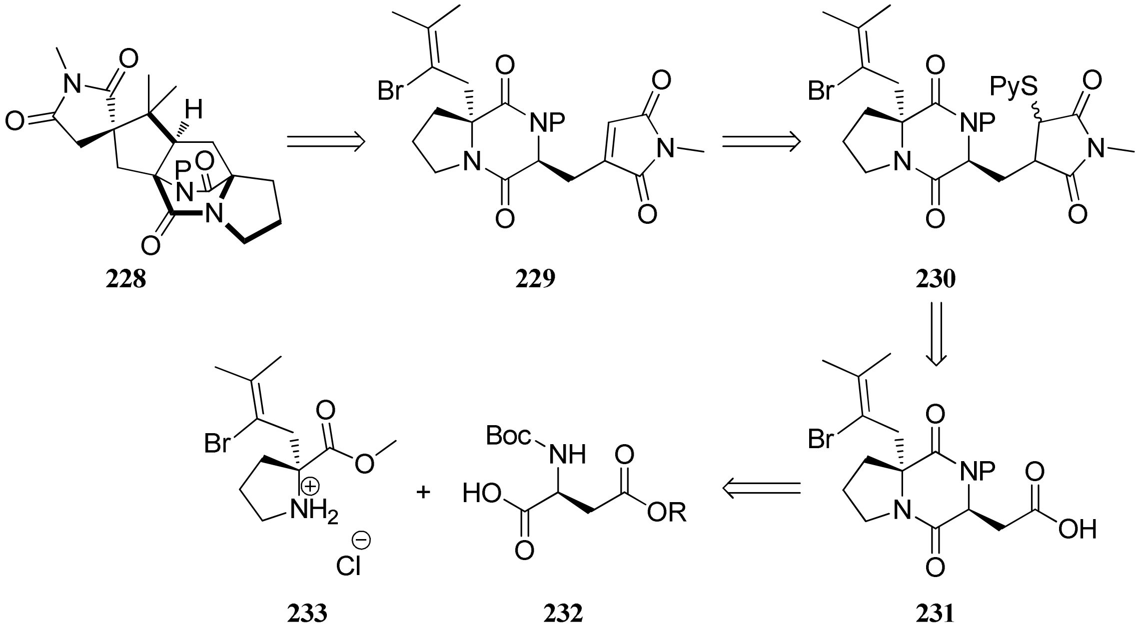
In this Chapter we have shown that our key cyclisation proceeds in good to excellent yields using a range of simple model systems. Additionally, a more complex system incorporating an allyl group was successfully cyclised with the major component possessing the desired stereochemistry at the vital C-6 position.

Chapter 4

Towards Asperparaline C

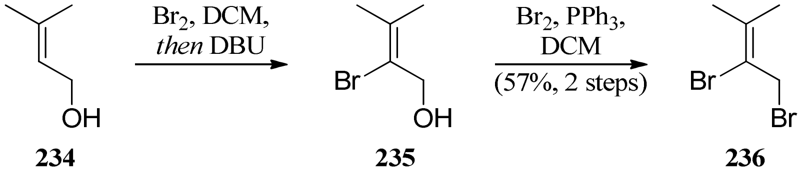
4.1 Retrosynthetic Analysis

While we were pleased with the success of our model system studies, we were keen to construct more complex systems that would allow us to make progress toward the synthesis of asperparaline C. As described in Chapter 3, we hoped that a DKP such as **229** would undergo a radical cascade to form the core bicyclo[2.2.2]diazaoctane along with the *spiro* fused succinimide (Scheme 4.1). We hoped that **229** would arise from oxidation and *syn* elimination of sulfide **230**, itself a product of a Barton decarboxylation and intermolecular radical addition to *N*-methylmaleimide starting from a suitable carboxylic acid such as **231**. DKP **231** should be relatively straightforward to synthesise from protected aspartic acid **232** and modified proline **233** using a similar series of reactions to those described previously.



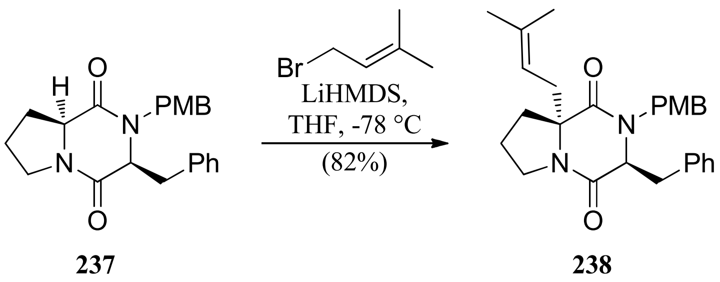
Scheme 4.1

Several protected aspartic acids are commercially available and we expected to be able to synthesise substituted proline **233** using the same Seebach chemistry we had previously employed. However, in this case the required electrophile (a prenyl group substituted with two bromines) is not commercially available. This component has to be synthesised from prenyl alcohol **234** by bromination to give first **235**[105] then the desired compound **236** (Scheme 4.2).[106]



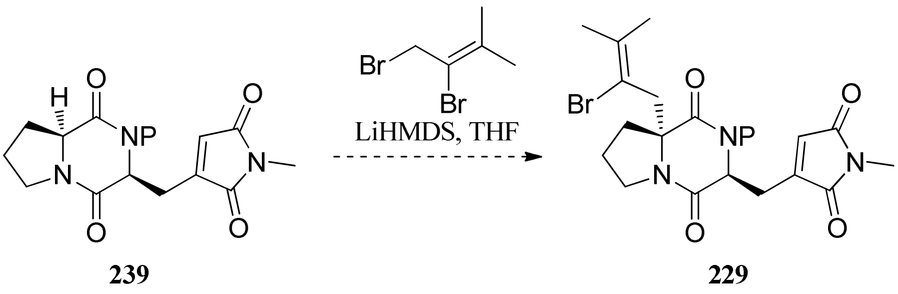
Scheme 4.2

As this piece is somewhat valuable, we also considered introducing it later in the synthesis. Previous work in our group has shown that substitutions on mixed DKPs of this type proceed with total regioselectivity for the proline residue and give solely the *anti* diastereomer. For example, deprotonation of a protected proline-phenylalanine DKP (**237**) with lithium bis(trimethylsilyl)amide followed by an external quench with prenyl bromide afforded **238** in 82% yield (Scheme 4.3).[96]



Scheme 4.3

In our case this would require substitution of a DKP such as **239** incorporating a maleimide along with a simple proline (Scheme 4.4). However, there is an additional complication as DKP **239** possesses an acidic methylene unit adjacent to the maleimide.

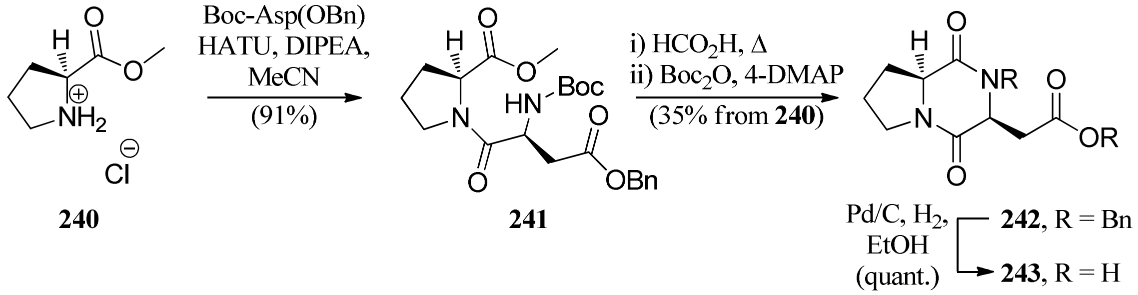


Scheme 4.4

Whether using the Seebach method or a late-stage prenylation, we envisaged synthesising cyclisation precursor **229** in 5 or 6 steps from commercially available starting materials.

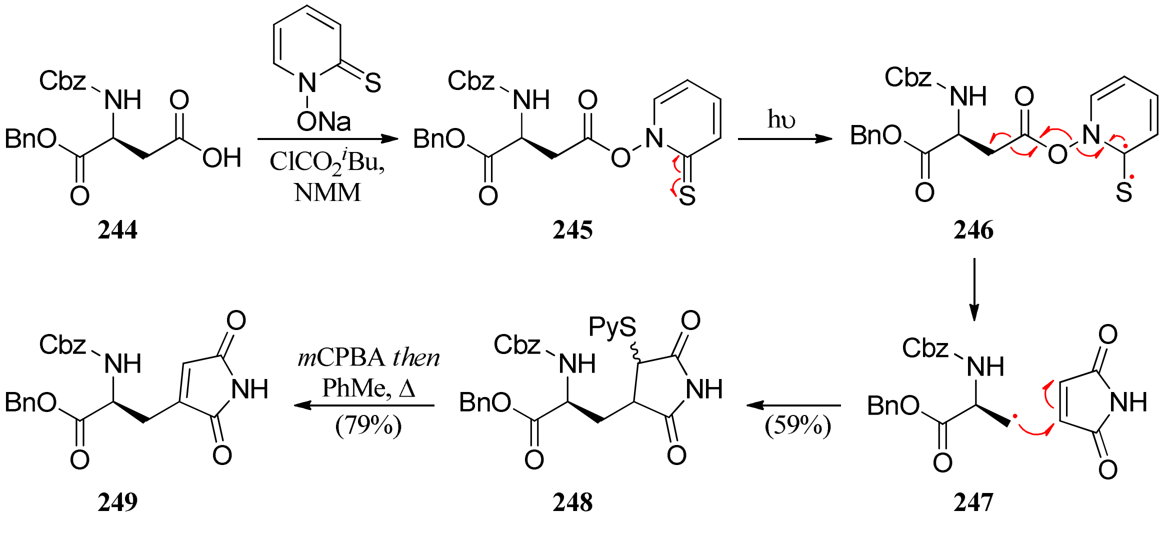
4.2 Initial Studies

In our first approach, commercially available L-proline *O*-methyl ester **240** was coupled with an aspartic acid piece. Our early studies used the aspartic acid *O*-methyl ester, but after difficulties with deprotection we changed to the *O*-benzyl series. Thus, amide bond formation in the presence of HATU and Hünig's base afforded dipeptide **241** in 91% yield (Scheme 4.5). As previously described, DKP formation was accomplished by acid-mediated Boc removal followed by heating in a mixture of toluene and 2-butanol to furnish **242** after Boc protection. Finally, hydrogenation of the benzyl ester gave free carboxylic acid **243** in a total of four steps.



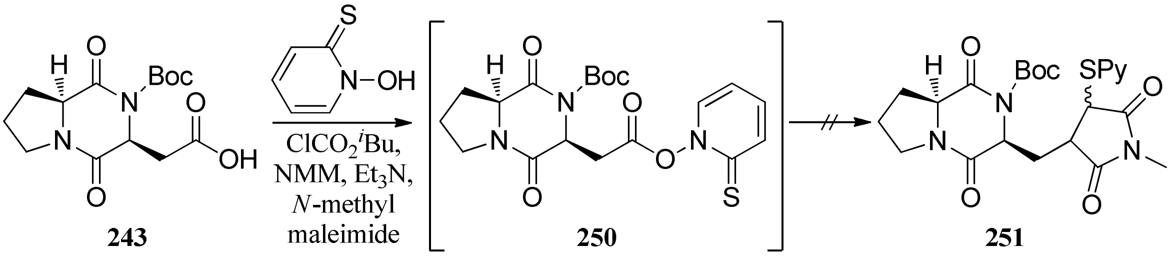
Scheme 4.5

With DKP **243** in hand we were ready to introduce the maleimide moiety. Barton and co-workers have previously demonstrated this reaction on a simple aspartic acid (Scheme 4.6).[107] In their work, coupling of **244** with the sodium salt of 2-mercaptopyridine *N*-oxide was achieved via formation of a mixed anhydride with isobutyl chloroformate. The resulting ester **245** was not isolated but irradiated in the presence of maleimide to afford succinimide **246** in 59% yield. This proceeds through radical scission of the ester to give a carbon centred radical along with loss of carbon dioxide and a 2-mercaptopyridine radical. Addition to the maleimide is followed by trapping through a sulfur radical to give the desired compound. To complete the process, oxidation of the sulfide to its corresponding sulfoxide was followed by *syn* elimination giving protected amino acid **249** in 79% yield.



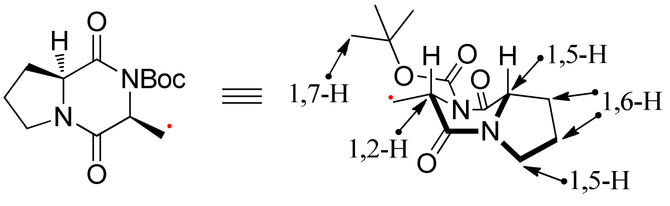
Scheme 4.6

Applying this to our system, DKP **243** was treated with *N*-methyl morpholine (NMM) and isobutyl chloroformate (Scheme 4.7). After addition of a solution of 2-mercaptopyridine *N*-oxide and triethylamine the mixture was stirred in the dark to prevent any premature photolysis of the intermediate ester. At this stage we were able to identify a peak possessing the correct mass for **250** by LC-MS. Unfortunately, however, after irradiation of the mixture in the presence of *N*-methyl maleimide, we were unable to isolate any of the desired compound, **251**.



Scheme 4.7

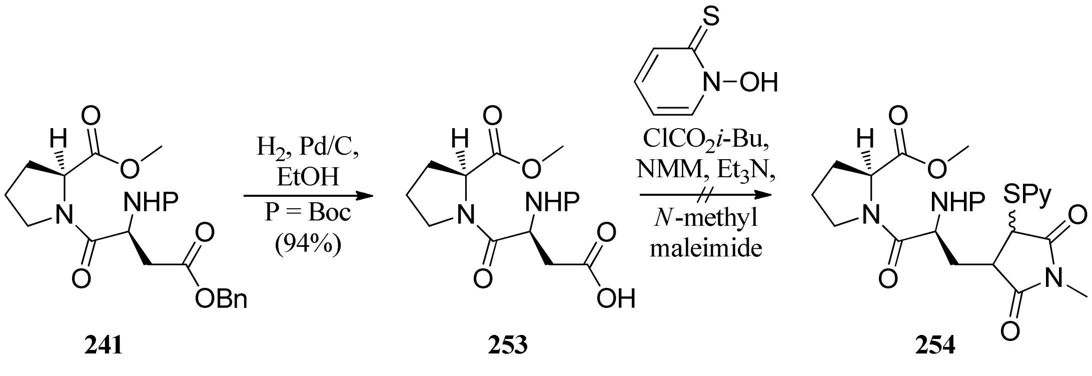
As we could isolate no clean compounds, it is difficult to identify what alternative process is interfering with the desired reaction. Assuming that the intermediate radical is generated, the desired next step is an intermolecular addition to an electron poor olefin. However, it is possible that one or more rapid intramolecular hydrogen atom abstractions can occur before this happens. Transfer of the radical centre in 1,2-mode to the captodative position on the DKP seems plausible, while abstraction from the pyrrolidine ring looks geometrically impossible (Figure 4.1). A 1,5-transfer to the alternative captodative position looks unlikely as the hydrogen on this carbon is positioned on the opposite side of the ring from the methyl radical we expect to generate. Lastly, a longer distance transfer in 1,7-mode from the Boc group is an outside possibility. If one or more of these processes did occur, we would expect a complex mixture of products as the chain process could terminate by addition of maleimide to various positions of the molecule. As the rate of intermolecular addition to an olefin is concentration dependent, it is difficult to compare the rate constant to that of an intramolecular hydrogen atom transfer. However, qualitatively we would expect the intramolecular reaction to be considerably faster.



252

Figure 4.1

As the shape of the molecule is often an important part of hydrogen atom transfers we also synthesised carboxylic acid **253** by hydrogenation of the benzyl ester of dipeptide **241** (Scheme 4.8). Unfortunately, however, we were again unable to isolate the desired compound incorporating a maleimide.

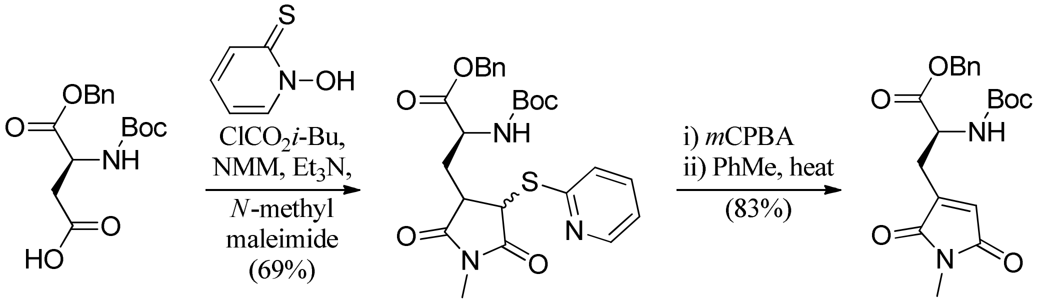


Scheme 4.8

With no obvious solution to this problem we abandoned this approach in favour of a more convergent synthesis of our key DKP.

4.3 Progress Towards Asperparaline C

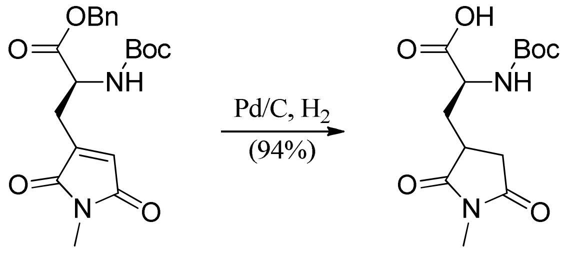
We hoped to synthesise an amino acid incorporating a maleimide that we could then couple with a proline piece to form our desired cyclisation precursor. Thus, using a modification of Barton's procedure reported by Johnson,[108] *N*-Boc aspartic acid *O*-benzyl ester **255** was decarboxylated in the presence of *N*-methylmaleimide to afford **256** in good yield (Scheme 4.9). Oxidation of the sulfide to the corresponding sulfoxide was accomplished with *m*CPBA before *syn* elimination on heating in toluene to give maleimide **257** in 83% yield.



255 **256** **257**

Scheme 4.9

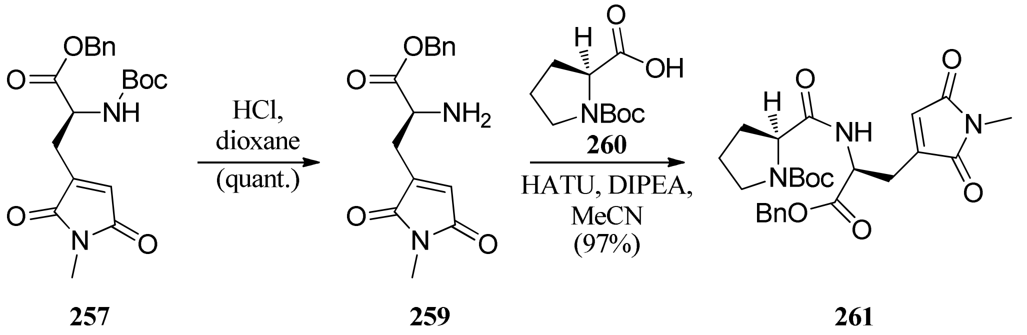
To convert amino acid **257** into a suitable partner for coupling with a proline moiety, we hoped to cleave the benzyl ester to give a free carboxylic acid. Unfortunately, cleavage under standard conditions (hydrogen gas in the presence of palladium on carbon) resulted in concurrent reduction of the maleimide double bond to give succinimide **258** in near-quantitative yield (Scheme 4.10). Other standard conditions for the saponification of esters (e.g. LiOH) also failed, leading to rapid degradation of the starting material.



257 **258**

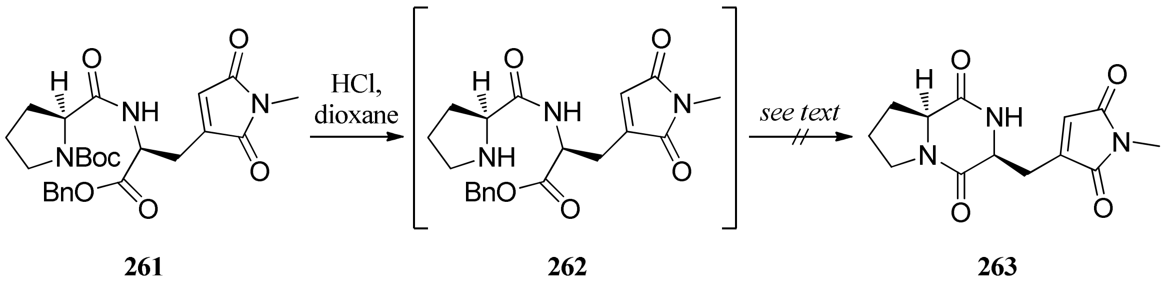
Scheme 4.10

Instead, we opted to cleave the *N*-Boc group to afford a suitable amine for coupling with *N*-Boc proline **260**. Thus, treatment of **257** with a solution of HCl in 1,4-dioxane led to rapid formation of **259** (Scheme 4.11). The subsequent peptide coupling proceeded well under the conditions we had previously used to give dipeptide **261** in near-quantitative yield.



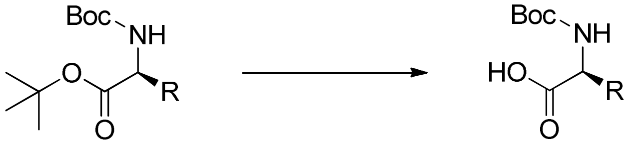
Scheme 4.11

Next, we hoped that removal of the pyrrolidine Boc group would be followed by DKP formation on heating in a suitable solvent. Treatment with acid led to smooth deprotection to give secondary amine **262** (Scheme 4.12). However, under our usual conditions for DKP formation (refluxing in 2-butanol/toluene) we recovered only starting material. Switching to the higher boiling *m*-xylene resulted in decomposition and mixtures of these solvents were similarly ineffective. Attempts to effect cyclisation in a sealed tube or microwave reactor had the same outcome – no reaction at low temperatures and decomposition under harsher conditions.



Scheme 4.12

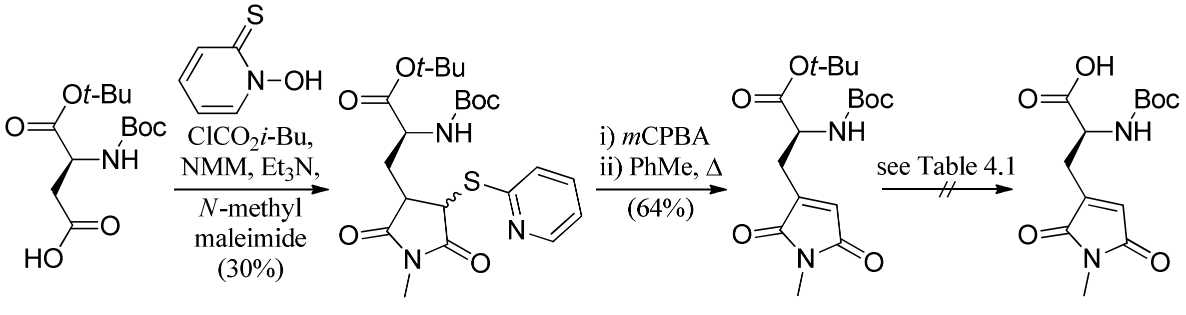
When taken together with the corresponding allylglycine derived dipeptide **223** (Chapter 3) these results demonstrate the difficulty in forming DKPs in this manner. Realising we could avoid this problem if we could selectively hydrolyse the ester of **257** we turned our attention to other protecting groups. We were keen to use a commercially available amino acid as the two carboxyl groups of aspartic acid complicate selective protection of one over the other. With this in mind, the *tert*-butyl ester was identified as a possibility. While we were initially reluctant to use a compound with two similarly reactive protecting groups (*N*-Boc vs. *tert*-butyl ester) a search of the literature revealed several methods for selective ester hydrolysis in the presence of a Boc group. For example, Marcantoni and co-workers reported a cerium mediated ester cleavage on a range of simple *N*-Boc amino acids.[109] They were able to isolate clean carboxylic acids in good to excellent yield (Table 4.1). Similarly, the group of Wu published a method for *tert*-butyl ester cleavage using zinc bromide,[110] while Yadav has employed montmorillonite KSF clay.[111]



entry	conditions	R	yield	reference
1	$CeCl_3 \cdot 7H_2O$-NaI	Bn	78	[109]
2	$ZnBr_2$	H	86	[110]
3	KSF clay	Bn	85	[111]

Table 4.1

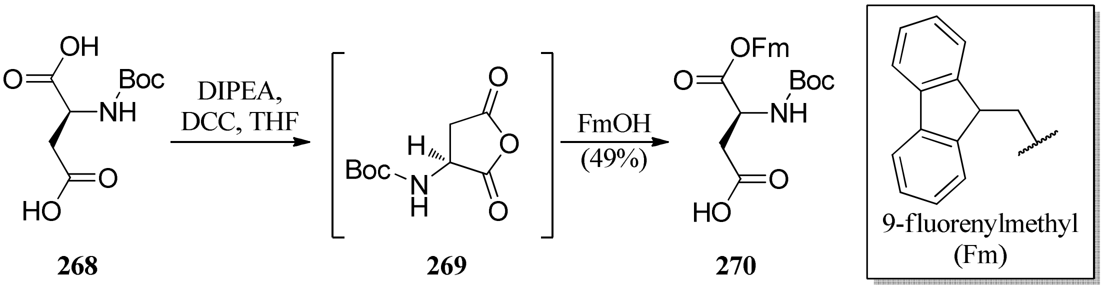
Our synthesis began with decarboxylation of aspartic acid *tert*-butyl ester **264** to afford **265** in 30% isolated yield (Scheme 4.13). The low yield in this case can be accounted for by the difficulty in separating the product from the remaining excess of *N*-methylmaleimide. Still, we were able to synthesise gram quantities of **265** to carry through to further steps. Thus, oxidation with *m*CPBA followed by *syn* elimination of the sulfur gave known maleimide **266** with moderate efficiency.[108] With this compound in hand, we were disappointed to find that all attempts at ester cleavage using the reported conditions were unsuccessful. As would be expected, treatment with protic acids gave only global deprotection forcing us to abandon this approach and consider other routes to **267**.



264 **265** **266** **267**

Scheme 4.13

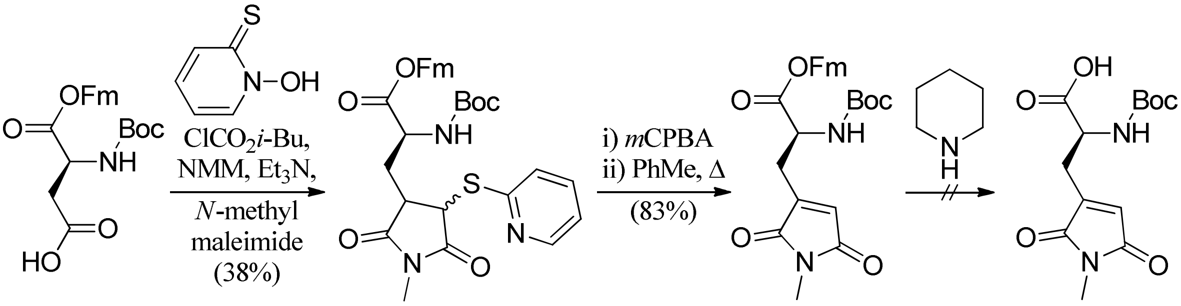
To better differentiate between the two protecting groups, we wanted to introduce a base-labile ester that could be readily removed without affecting the acid-sensitive Boc group. The 9-fluorenylmethyl (Fm) ester seemed suitable especially as a one-step access has been reported via the cyclic anhydride corresponding to *N*-Boc aspartic acid.[112] Treatment of diacid **268** with DCC in the presence of Hünig's base afforded **269** which was immediately treated with 9-fluorenylmethanol to give exclusively the alpha Fm ester **270**.



268 **269** **270**

Scheme 4.14

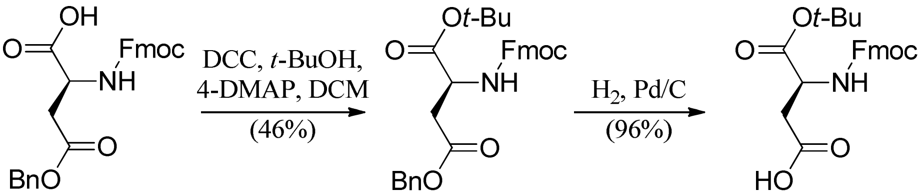
Next, Barton decarboxylation under the established conditions was followed by oxidation and elimination to give maleimide **272** in good yield. However, we were surprised to discover that treatment with piperidine led to immediate degradation of the material – even after only a few minutes at 0 °C. Other common conditions for the removal of fluorenylmethyl esters such as diethylamine and TBAF had similar effects.



270 **271** **272** **267**

Scheme 4.15

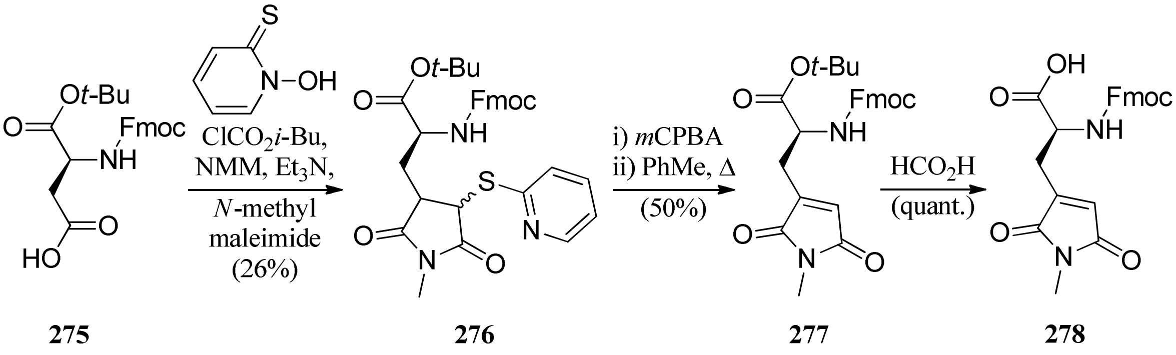
Contemporaneously with these studies, we had synthesised aspartic acid **275** possessing an acid-labile *tert*-butyl ester along with an *N*-Fmoc protecting group. Two steps were necessary for this – esterification of benzyl ester **273** followed by hydrogenation to afford acid **275** (Scheme 4.16).



273 **274** **275**

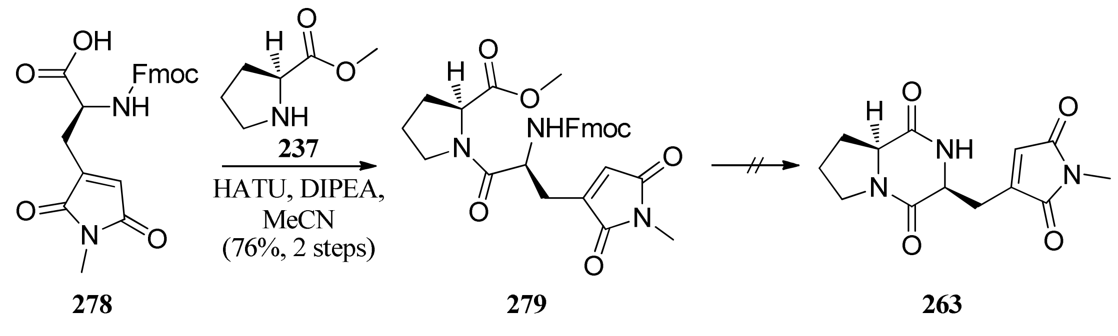
Scheme 4.16

Again, installation of the maleimide proceeded with modest yield through radical decarboxylation followed by sulfoxide formation and *syn* elimination (Scheme 4.17). This time, however, exposure to neat formic acid led to clean ester hydrolysis to yield **278** without affecting the Fmoc protected amine.



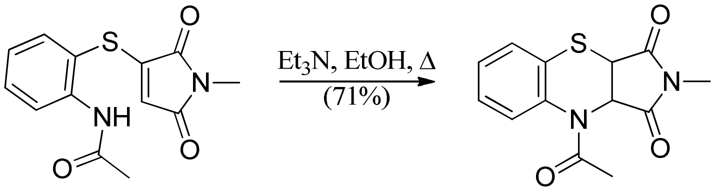
Scheme 4.17

We were pleased to find that peptide coupling under our established conditions (HATU, Hünig's base) afforded dipeptide **279** in 76% yield (Scheme 4.18). Frustratingly, all attempts at Fmoc cleavage and DKP formation were unsuccessful despite screening numerous bases (piperidine, diethylamine, TBAF, TMP, sodium methoxide, DBU).



Scheme 4.18

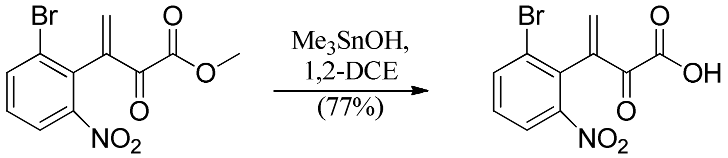
During this work, we frequently observed rapid degradation of compounds possessing a maleimide upon treatment with base. While we were unable to isolate any clean by-products, we hypothesised that base-promoted Michael addition of nitrogen onto the maleimide could be responsible. Maleimides are well known Michael acceptors. For example, Igarashi and Watanabe found that treatment of sulfur substituted maleimides such as **280** with 5 equivalents of triethylamine led to intramolecular 1,4-addition to afford tricycle **281** in 71% yield (Scheme 4.19).[113] Interestingly, the reaction proceeded with similar efficiency when sub-stoichiometric (10 mol%) quantities of base were used. This implies that this system is primed for cyclisation even under mild conditions.



280 **281**

Scheme 4.19

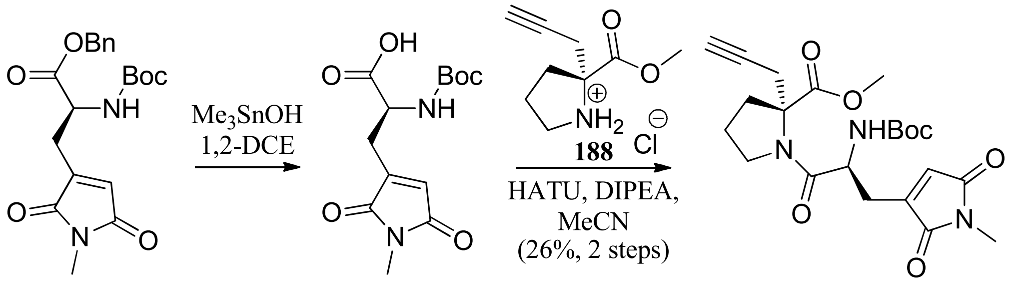
With this hypothesis in mind, we focused our literature studies on systems incorporating both an ester and Michael acceptor. Alongside numerous examples using common ester hydrolysis conditions (e.g. LiOH or NaOH) we found an isolated example reported by the group of Nicolaou employing trimethyltin hydroxide.[114] In particular, the example shown in Scheme 4.20 was of interest. Methyl ester **282** was converted to the corresponding carboxylic acid **283** by treatment with trimethyltin hydroxide in dichloroethane at 80 °C. While the authors noted a minor by-product caused by intramolecular conjugate addition of the carboxylate group to the neighbouring Michael acceptor, the desired compound was obtained in 77% yield. The same conditions were reported to be successful on a range of esters including benzyl, ethyl and allyl derivatives.



282 **283**

Scheme 4.20

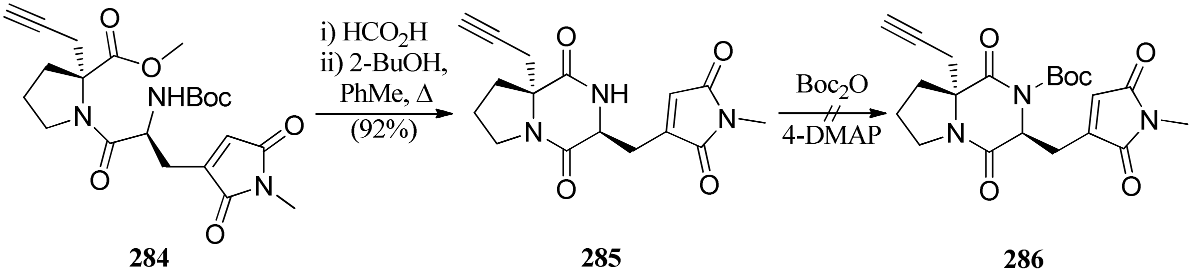
Applying these conditions to benzyl ester **257**, we were delighted to obtain carboxylic acid **267** after 20 hours at 80 °C (Scheme 4.21). Unfortunately this compound proved to be unstable on silica gel but we were able to isolate reasonably clean material by repeated washes with dilute acid. With a free carboxylic acid finally in hand, we were eager to continue our synthesis. Thus, coupling of **267** with substituted proline **188** was carried out under standard conditions. We were disappointed that, after purification, the yield over these two steps was only 26%, possibly due to impurities remaining with acid **267**. Still, the brevity of this route (4 steps is the longest linear sequence to **284**) allowed us to carry through sufficient material to continue.



257 **267** **284**

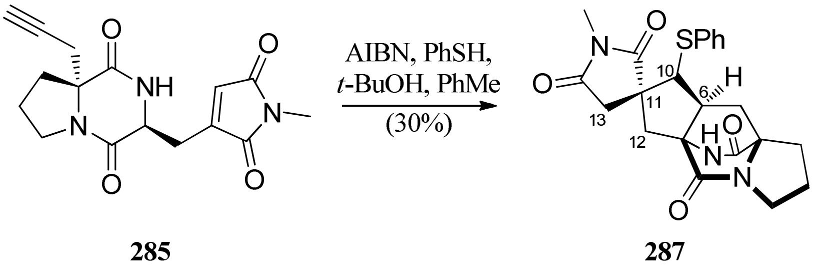
Scheme 4.21

Having obtained suitably protected dipeptide **284**, formation of the corresponding DKP was trivial. Cleavage of the Boc group with formic acid was followed by heating in 2-butanol and toluene to afford **285** in 92% yield (Scheme 4.22). Attempted Boc protection of **285** again demonstrated the sensitivity of these compounds to base – this time exposure to 0.1 equivalents of 4-DMAP led to degradation of the starting material.



Scheme 4.22

As we were unable to protect the lactam *N*-H of **285**, we carried out our key cyclisation with the *N*-H DKP. This reaction is considerably more complex than many of those we had previously carried out as we hoped to generate two new carbon-carbon bonds along with three new stereocentres. Although we obtained a complex mixture of compounds, we were delighted to find that the major component possessed a bridged bicyclo[2.2.2]diazaoctane core along with a *spiro*-fused succinimide (Scheme 4.23).



Scheme 4.23

While deducing the basic structure of **287** was relatively straightforward, assigning the stereochemistry proved more difficult. We found that ^{1}H NMR spectra recorded in deuterated chloroform had peaks corresponding to H-6, H-10 and one H-13 within a region spanning just 0.2 ppm. While heteronuclear correlation experiments (HSQC/HMBC) allowed us to assign the individual peaks, we were unable to selectively excite each proton for nOe studies. However, we were pleased to find that recording spectra in benzene-d_6 resulted in some of these peaks spreading further apart. We were then able to carry out nOe experiments to determine the stereochemistry of **287**. As we knew the stereochemistry of the proline residue, we hoped to use the amide N-H of the DKP to establish the configuration at C-6, C-10 and C-11. While H-6 was still in a cluttered area of the spectrum, exciting one H-12 showed a strong nOe correlation to both H-6 and the N-H, along with a coupling to the other geminal H-12 (blue spectrum, Figure 4.2). This established the stereochemistry at C-6 as well as differentiating between the H-12a and H-12b. Next we needed to determine the configuration at the *spiro* ring junction. After studying models of **287**, we reasoned that if C-13 was 'up' H-13b would be positioned close to H-12a, while if C-13 was 'down' the correlation would be between H-13b and H-12b. We were delighted to find that excitation of one H-13 showed an nOe to H-12a, along with coupling to the other geminal H-13 (red spectrum, Figure 4.2).

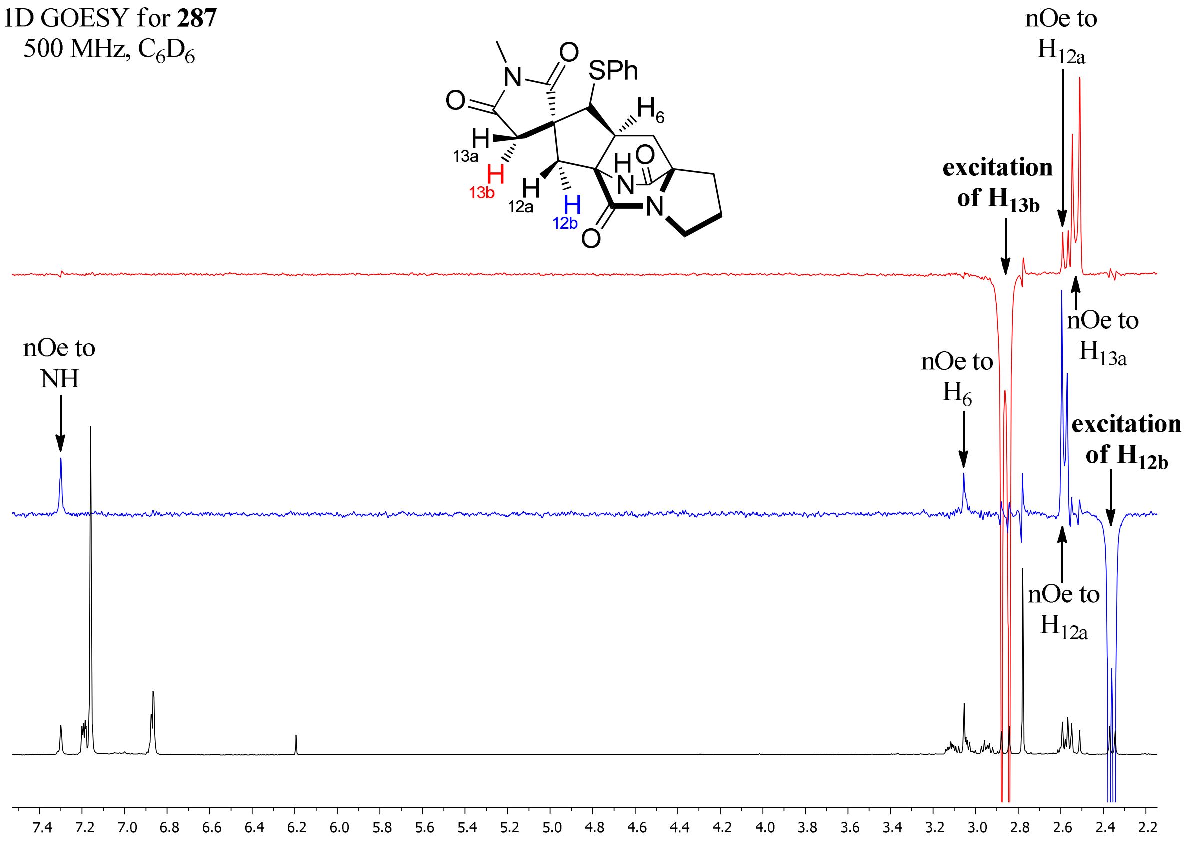


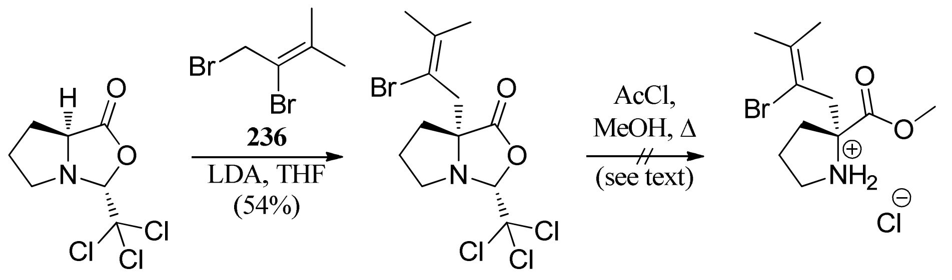
Figure 4.2

Unfortunately, we were unable to establish the configuration adjacent to the sulfur (C-10) but as the natural product bears a *gem*-dimethyl group at this position this stereocentre is of little importance. Taken together, our nOe experiments allowed us to confirm that the vital stereocentres of **287** match with those of the asperparalines.

The sequence to **287** is exceptionally short – just 6 steps in the longest linear sequence from commercially available *N*-Boc aspartic acid benzyl ester. While several of the yields are modest, this represents the first synthesis of a compound incorporating a DKP along with a *spiro*-succinimide so is a significant step towards a total synthesis of the asperparalines.

4.4 Further Work and Alternative Routes

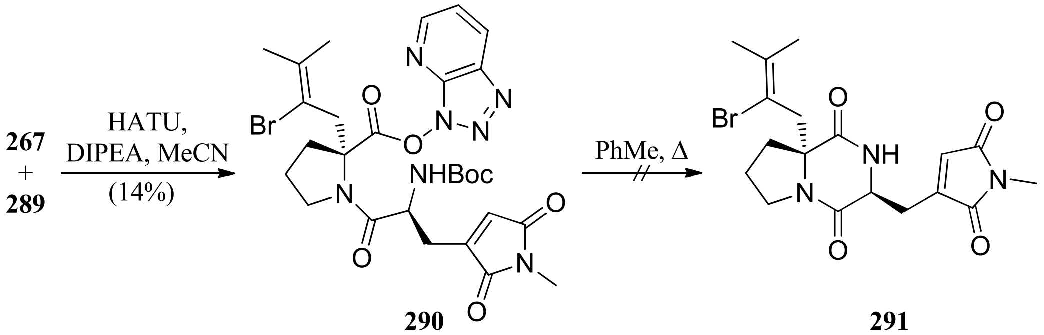
Having accomplished the construction of the asperparaline core, we hoped that installing vinyl bromide **236** would allow incorporation of the missing *gem*-dimethyl group. Although the original plan was to introduce **236** by deprotonation of a DKP, the sensitivity of our substrates to basic conditions forced us to reconsider this and again use oxazolidinone **186**. Thus, deprotonation of **186** with LDA was followed by an electrophilic quench with vinyl bromide **233** to afford **288** in 54% yield (Scheme 4.24). However, attempted ring-opening under our usual conditions (acetyl chloride in refluxing methanol) did not give clean **289**. The product of this reaction had ambiguous spectral data, although analysis by mass spectrometry suggested the correct molecular formula.[115]



186 **288** **289**

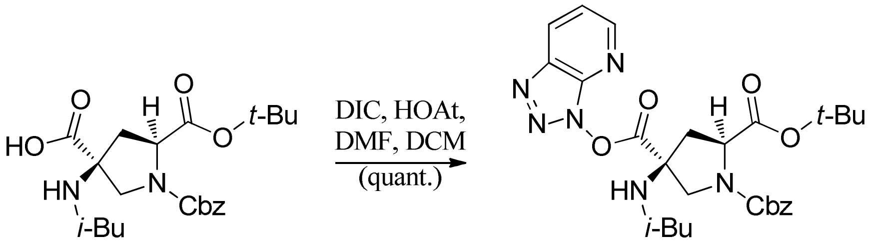
Scheme 4.24

We hoped that by coupling this compound to maleimide **267** we would be able to purify the product and resolve this ambiguity. However, we were unable to isolate the desired dipeptide from this reaction but instead obtained a by-product tentatively identified as triazole **290** in 14% yield (Scheme 4.25). All attempts to convert this to DKP **291** by heating at high temperatures were unsuccessful leading to recovery of the starting material.



Scheme 4.25

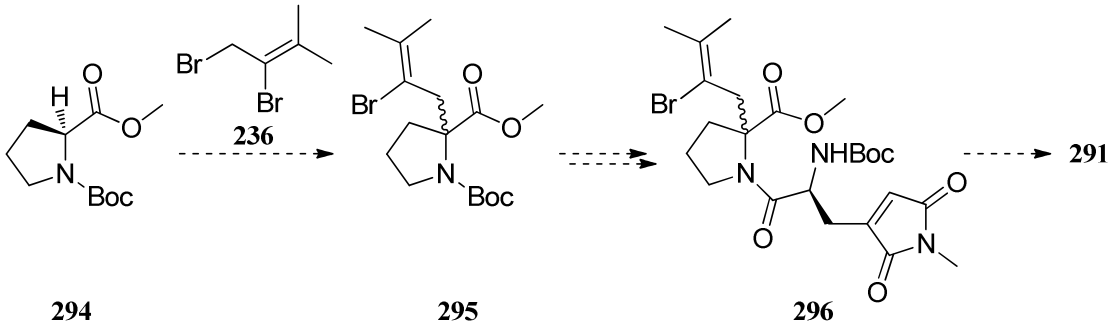
Triazoles analogous to **290** have previously been synthesised by Brown and Schafmeister by treating carboxylic acids such as **292** with 1-hydroxy-7-azabenzotriazole (HOAt) in the presence of di*iso*propylcarbodiimide (DIC) to obtain **293** (Scheme 4.26).[116] They reported that these compounds were surprisingly stable to self-condensation despite the presence of a relatively nucleophilic secondary amide and could only be made to react under harsh thermal conditions.



292 **293**

Scheme 4.26

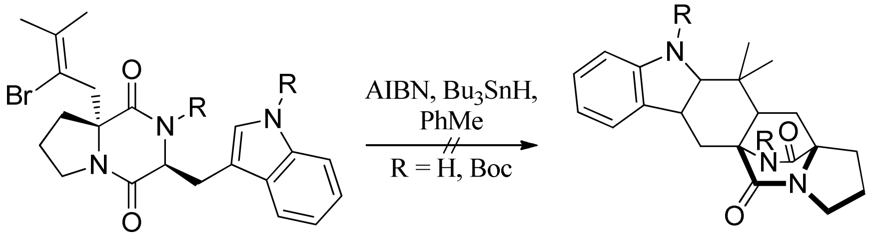
If this problem cannot be solved, an alternative approach could be to prenylate a simpler protected proline such as **294** to obtain **295** as a racemate (Scheme 4.27). This would avoid the problematic cleavage of the Seebach oxazolidinone and Baran has previously shown that deprotection using *para*-toluenesulfonic acid can be followed by coupling promoted by BOP chloride to give dipeptide products.[57c] Separation of the resulting diastereomers and DKP formation would then afford our key cyclisation precursor **291**.



294 **295** **296**

Scheme 4.27

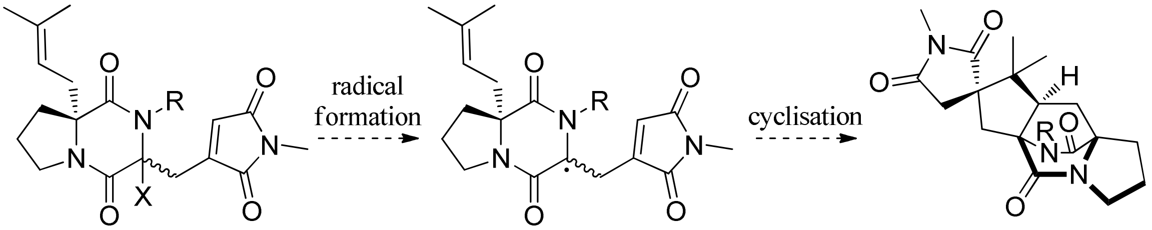
In addition to the problems in forming DKP **291**, recent studies in our laboratory have identified difficulties in initiating a radical from a fully substituted vinyl halide. Treatment of **297** with AIBN and tributyltin hydride resulted in recovery of the starting material without any trace of **298** or reduction of the vinyl bromide (Scheme 4.28).



297 **298**

Scheme 4.28

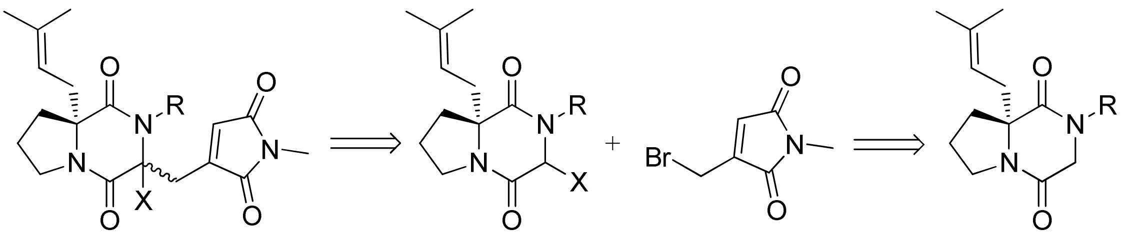
In light of this, we have also considered alternative methods for generating a radical at the desired position of our DKP. For example, instead of relying on a 1,6-H abstraction, scission of a suitable substituent on the DKP (ie. X = SPh, CO_2R) could directly give intermediate **165** which would then cyclise to give bridged DKP **228** (Scheme 4.29).



299 **168** **228**

Scheme 4.29

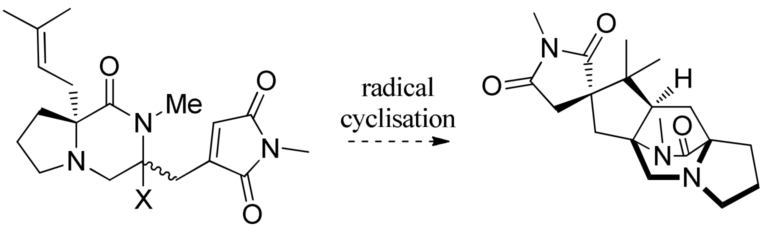
Maleimide **299** could arise from addition of a suitable electrophile to the enolate of a prenylated DKP such as **300**. This, in turn, should be accessible by substitution of the simple proline-glycine DKP **302** which is similar to compounds we have previously synthesised (Scheme 4.30).



299 **300** **301** **302**

Scheme 4.30

If one of these approaches does give access to the bridged core structure of the asperparalines, reduction of the extra carbonyl could still be a significant problem. While we expect any approach incorporating a hydrogen atom abstraction to require the captodative position of the DKP, a cyclisation based on direct radical formation might be possible without this in place (Scheme 4.31).



303 *ent*-**28**

Scheme 4.31

While the MKP precursor **303** for this reaction could be challenging to synthesise, if a successful strategy was developed it would lead directly to asperparaline C without requiring any later-stage functional group modifications.

4.5 Summary

We have demonstrated a radical cascade for the synthesis of the asperparaline core structure incorporating a bridged DKP along with a *spiro*-fused succinimide in just 6 steps. At present the sequence to this compound is compromised by several moderate yields, but we hope that future modification of our route will lead to the total synthesis of this fascinating compound.

Chapter 5

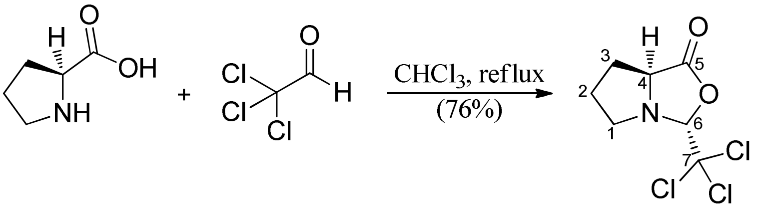
Experimental

5.1 General Methods

All reactions were performed under an atmosphere of nitrogen or argon in dry glassware unless otherwise stated. THF was distilled immediately prior to use from sodium and benzophenone; CH_2Cl_2 was distilled from calcium hydride. All other solvents and reagents were used as received from commercial suppliers unless otherwise stated. Solvents and solutions for radical reactions were degassed before use by bubbling a steady stream of dry N_2 gas through them for 30 minutes. Infra-red spectra were recorded neat on a Perkin-Elmer Spectrum 100 FTIR spectrometer. Wavelengths (ν) are reported in cm^{-1}. Optical rotations were recorded as dilute solutions in the indicated solvent in a 25 mm glass cell using a JASCO DIP370 digital polarimeter at 294 nm. Mass spectra were obtained using a VG Micromass 70E or VG Micron Autospec spectrometer, using electrospray ionization (ESI) with meta-nitrobenzyl alcohol as matrix. All ^1H NMR and ^{13}C NMR experiments were recorded using Bruker AVIII300, AVIII400, and DRX500 spectrometers at 300 K. ^{13}C NMR were recorded using the PENDANT pulse sequence from the Bruker standard pulse program library. Chemical shifts (δ) are quoted in ppm and coupling constants (J) are quoted in Hz to one decimal place. For spectra recorded in chloroform-*d* the 7.27 ppm resonance of residual $CHCl_3$ for proton spectra and 77.16 ppm resonance of $CDCl_3$ for carbon spectra were used as internal references. For spectra recorded in methanol-d_4 the 3.34 ppm resonance of residual CH_3OH for proton spectra and 49.86 ppm resonance of CD_3OD for carbon spectra were used as internal references. Spectral data for ^1H NMR is reported as follows: chemical shift (multiplicity, coupling constant, number of protons, assignment); and for ^{13}C NMR: chemical shift (assignment). The following abbreviations were used for multiplicity in ^{1}H NMR: s (singlet), d (doublet), t (triplet), q (quadruplet), dd (doublet of doublets), td (triplet of doublets), qd (quadruplet of doublets), br (broad), app. (signal with well defined multiplicity that may not be expected). In the case of ambiguous assignments, 2-dimensional homonuclear (^{1}H-^{1}H) and heteronuclear (^{1}H-^{13}C) NMR experiments were used. For simplicity of assignments all atoms are labelled, although these numbers do not necessarily follow IUPAC naming conventions. Melting points were recorded on a Gallenkamp melting point apparatus and are uncorrected. Reaction progress was monitored by thin layer chromatography (TLC) performed on plastic plates coated with keiselgel F254 with 0.2 mm thickness. Visualisation was achieved by a combination of ultraviolet light (254 nm) and acidic potassium permanganate or anisaldehyde. Flash column chromatography was performed using silica gel 60 (230-400 mesh, Merck and Co.).

5.2 Experimental for Chapter 3

Oxazolidinone 186



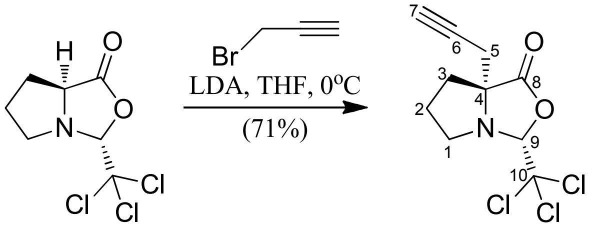
186

A solution of L-proline (7.6 g, 66.0 mmol, 1.0 eq.) in chloroform (100 mL) was treated with anhydrous trichloroacetaldehyde (9.66 mL, 99.0 mmol, 1.5 eq.). The flask was fitted with a Dean-Stark receiver and the reaction heated at reflux for 4 hours. The resulting brown solution was cooled to room temperature, washed with water (2 x 40 mL) and the aqueous phases extracted with dichloromethane (2 x 40 mL). The combined organic layers were dried over $MgSO_4$ and concentrated under reduced pressure. The resulting brown solid was purified by recrystallisation from ethanol to yield the title compound **186** as white crystalline needles (12.34 g, 50.5 mmol, 76%).

(**186**, white needles) m.p. 106 °C – 110 °C [lit. 108 °C – 109 °C];[117] $[\alpha]_D^{20}$ +44.0 (*c* 0.50, $CHCl_3$) [lit. +34.0 (*c* 2.00, C_6H_6);[117] FTIR (film) ν_{max} 2961, 1781, 1450, 1322, 1245, 1175, 1001, 958; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.15 (s, 1H, H-6), 4.08 (dd, *J*=8.8, 4.8, 1H, H-4), 3.39 (ddd, *J*=10.9, 7.9, 6.3, 1H, H-1), 3.12 (dd, *J*=11.2, 5.7, 1H, H-1), 2.00 (m, 4H, H-2,3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 175.3 (C-5), 103.4 (C-6), 101.0 (C-7), 62.3 (C-4), 57.7 (C-1), 29.8 (C-3), 25.2 (C-2); HRMS (ESI) calculated for $C_7H_8Cl_3NO_2$ $[M+Na]^+$ 265.9518 found 265.9520.

Data in agreement with those previously reported.[85, 87, 117-118]

Propargylated Oxazolidinone 187

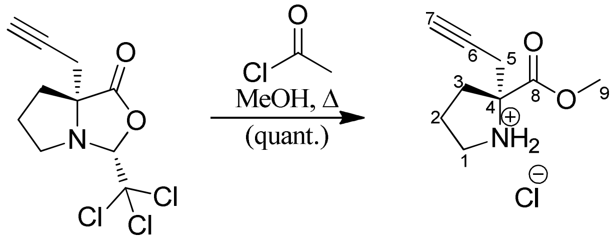


186 **187**

A solution of diisopropylamine (9.63 mL, 68.71 mmol) in THF (120 mL) was cooled to –78 °C and treated with *n*-butyllithium (1.6M solution in hexanes, 42.9 mL, 68.71 mmol). The resulting LDA solution was allowed to stir for 30 minutes. Oxazolidinone **186** (12.0 g, 49.08 mmol) in THF (90 mL) was added via cannula and the reaction mixture stirred for 1 hour before addition of propargyl bromide (80% wt. solution in toluene, 27.4 mL, 245.4 mmol). After 4 hours at -78 °C the reaction was quenched with NH_4Cl (saturated aqueous solution, 15 mL) and allowed to warm to room temperature. The volatile organics were removed under reduced pressure and the resulting oily residue partitioned between dichloromethane and water. The layers were separated and the aqueous phase extracted with dichloromethane (3 x 100 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure. Purification of the brown oily residue by column chromatography (eluent petroleum ether/ethyl acetate, 9/1 gradually to 8/2) afforded the title compound **187** as a white solid (9.9 g, 35.04 mmol, 71%).

(**187**, white solid) m.p. 87 °C – 90 °C; $[\alpha]_D^{20}$ +32.6 (*c* 1.13, $CHCl_3$); FTIR (film) ν_{max} 3298, 2970, 1784, 1328, 1194, 1104, 834, 631; ^1H NMR (300 MHz, $CDCl_3$) δ 1.61-2.45 (m, 4H, H-2, H-3), 2.06 (t, *J*=2.9, 1H, H-7), 2.81 (d, *J*=2.9, 2H, H-5), 3.24-3.48 (m, 2H, H-1), 4.99 (s, 1H, H-9); ^{13}C NMR (75 MHz, $CDCl_3$) δ 174.8 (C-8), 101.9 (C-9), 78.8 (C-10), 71.1 (C-7), 70.5 (C-6), 60.2 (C-4), 58.5 (C-1), 35.6 (C-3), 28.0 (C-5), 25.2 (C-2); HRMS (ESI) calculated for $C_{10}H_{10}Cl_3NO_2$ $[M+Na]^+$ 303.9675 found 303.9665.

Propargylated Proline *O*-Methyl Ester 188

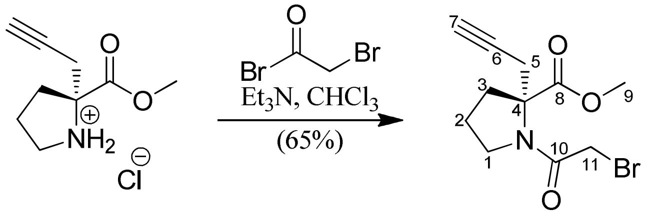


187 **188**

A stirred solution of **187** (9.9 g, 35.04 mmol) in methanol (200 mL) was treated with a solution of acetyl chloride (5.23 mL, 73.58 mmol) in methanol (100 mL). The resulting mixture was heated at reflux for 20 hours before concentration under reduced pressure. The residue was dried by azetrope with toluene to afford proline *O*-methyl ester **188** as an off-white solid (7.27 g, ~quantitative) that was used without further purification in subsequent steps.

(**188**, off-white solid) m.p. 180 °C (dec.); $[\alpha]_D^{20}$ −63.7 (*c* 0.91, MeOH); FTIR (film) ν_{max} 3171, 2918, 2647, 1757, 1416, 1219, 1098; ^{1}H NMR (300 MHz, CD_3OD) δ 3.88 (s, 3H, H-9), 3.40-3.45 (m, 2H, H-1), 3.13 (dd, *J*=17.7, 2.6, 1H, H-5), 2.84 (dd, *J*=17.7, 2.6, 1H, H-5), 2.73 (t, *J*=2.6, 1H, H-7), 1.97-2.19 (m, 4H, H-2, H-3); ^{13}C NMR (75 MHz, CD_3OD) δ 108.7 (C-8), 77.4 (C-6), 75.3 (C-7), 73.0 (C-4), 54.8 (C-9), 47.7 (C-1), 35.6 (C-3), 26.3 (C-5), 24.3 (C-2); HRMS (ESI) calculated for $C_9H_{14}NO_2$ (free amine) $[M+H]^+$ 168.1025 found 168.1027.

Acylated Proline *O*-Methyl Ester 189

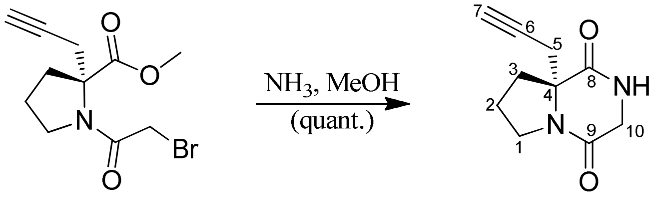


188 **189**

A suspension of crude **188** (3.8 g, ~18.66 mmol) in chloroform (50 mL) was cooled to –10 °C and treated sequentially with triethylamine (6.22 mL, 44.78 mmol) and bromoacetyl bromide (3.90 mL, 44.78 mmol) in chloroform (50 mL). After stirring for 3 hours the reaction was quenched with water (50 mL) and the layers separated. The organic phase was washed with sodium hydrogen carbonate (sat. aq. soln., 50 mL), brine (50 mL) and dried over $MgSO_4$. Removal of the solvent under reduced pressure afforded a brown oil which was purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 then 7/3) to yield the title compound **189** as a colourless oil (3.5 g, 1.18 mmol, 65%).

(**189**, colourless oil) $[\alpha]_D^{20}$ +52.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3236, 2958, 1733, 1635, 1417, 1263, 1216; ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.85 (d, *J*=2.9, 2H, H-11), 3.72 (s, 3H, H-9), 3.66-3.89 (m, 2H, H-1), 3.45 (dd, *J*=17.3, 2.8, 1H, H-5), 2.86 (dd, *J*=17.3, 2.8, 1H, H-5), 2.04-2.51 (m, 4H, H-2, H-3), 1.96 (t, *J*=2.8, 1H, H-7); ^{13}C NMR (75 MHz, $CDCl_3$) δ 172.5 (C-8/10), 164.9 (C-8/10), 79.9 (C-6), 70.6 (C-7), 67.8 (C-4), 52.6 (C-9), 49.6 (C-1), 35.7 (C-3), 27.5 (C-5), 26.4 (C-11), 23.9 (C-2); HRMS (ESI) calculated for $C_{12}H_{14}BrNO_3$ $[M+Na]^+$ 310.0055 found 310.0057.

Proline-Glycine *N*-H DKP 190a

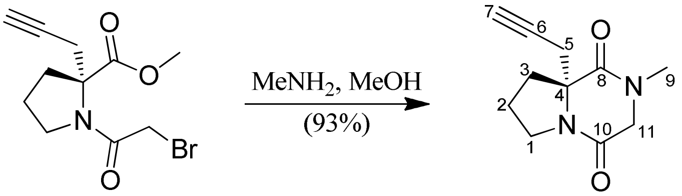


189 **190a**

Bromide **189** (1.06 g, 3.68 mmol) was dissolved in an ammonia solution (2M in methanol; 18.4 mL, 36.8 mmol) and allowed to stir at room temperature for 4.5 hours. The volatiles were removed under reduced pressure and DCM was added to the residue. The resulting precipitate was removed by filtration and the filtrate was concentrated under reduced pressure to afford the title compound **190a** (716 mg ~quantitative) as a sticky yellow oil that was dried under high vacuum then used without further purification.

(**190a**, yellow oil) $[\alpha]_D^{20}$ −138.8 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3262, 2982, 2941, 1776, 1724, 1645, 1440, 1148; ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.70 (s, br, 1H, N-H), 4.38 (d, *J*=17.0, 2H, H-10), 3.93-3.79 (m, 2H, H-1), 2.77 (dd, *J*=16.9, 2.7, 1H, H-5), 2.61 (dd, *J*=16.9, 2.7, 1H, H-5), 2.23-2.14 (m, 2H, H-3), 2.08 (t, *J*=2.7, 1H, H-7), 2.05-1.94 (m, 2H, H-2); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.5 (C-8/9), 161.8 (C-8/9), 76.6 (C-6), 70.5 (C-7), 64.7 (C-4), 45.1 (C-10), 43.3 (C-1), 33.1 (C-3), 26.4 (C-5), 18.4 (C-2); HRMS (ESI) calculated for $C_{10}H_{12}N_2O_2$ $[M+Na]^+$ 215.0796 found 215.0784.

Proline-Glycine *N*-Methyl DKP 190b

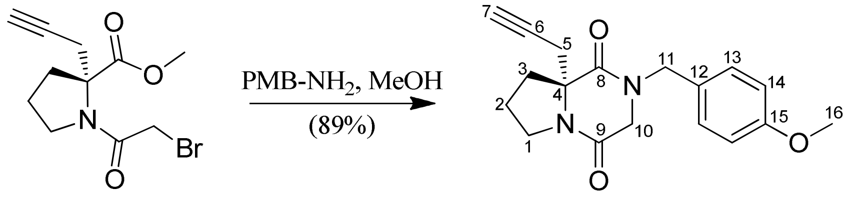


189 **190b**

Bromide **189** (1.90 g, 6.58 mmol) in methanol (10 mL) was treated with a methylamine solution (2M in methanol; 33.0 mL, 65.8 mmol) at 0 °C. After 1.5 hours the reaction mixture was concentrated under reduced pressure and the resulting yellow residue treated with dichloromethane (20 mL). The resulting white precipitate was removed by filtration and the filtrate concentrated under reduced pressure. The residue was purified by column chromatography (eluent 100% ethyl acetate then ethyl acetate/methanol, 9/1) to afford the title compound **190b** as a white solid (1.17 g, 6.09 mmol, 93%).

(**190b**, white solid) m.p. 128 °C – 130 °C; $[\alpha]_D^{20}$ −106.8 (*c* 1.00, CHCl$_3$); FTIR (film) ν_{max} 3284, 2974, 2888, 1651, 1454, 1408, 1339, 1311, 1224, 1013; ^{1}H NMR (300 MHz, CDCl$_3$) δ 4.46 (d, *J*=16.9, 1H, H-11), 3.89 (m, 1H, H-1), 3.80 (d, *J*=16.9, 1H, H-11), 3.57 (m, 1H, H-1), 3.02 (s, 3H, H-9), 2.79 (dd, *J*=17.3, 2.7, 1H, H-5) 2.59 (dd, *J*=17.3, 2.7, 1H, H-5), 2.20-1.95 (m, 4H, H-2, H-3), 2.05 (t, *J*=2.7, 1H, H-7); ^{13}C NMR (75 MHz, CDCl$_3$) δ 168.2 (C-8/10), 162.9 (C-8/10), 78.6 (C-6), 72.0 (C-7), 66.7 (C-4), 53.8 (C-11), 44.8 (C-1), 35.3 (C-3), 33.5 (C-9), 28.4 (C-5), 20.1 (C-2); HRMS (ESI) calculated for $C_{11}H_{14}N_2O_2$ $[M+Na]^+$ 229.0953 found 229.0947.

Proline-Glycine *N*-PMB DKP 190c

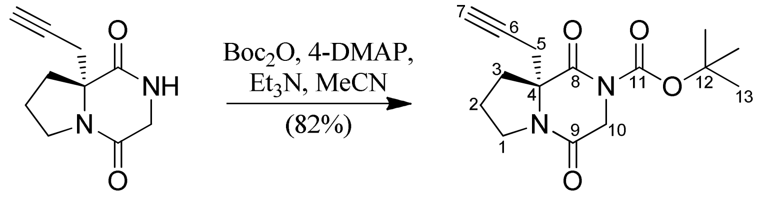


189 **190c**

Bromide **189** (211 mg, 0.73 mmol, 1.0 eq.) was dissolved in methanol (3.7 ml) and treated with 4-methoxybenzyl amine (956 μL, 7.32 mmol, 10.0 eq.). The resulting solution was allowed to stir at room temperature for 2.5 hours, then the volatiles removed under reduced pressure. DCM was added to the residue and the resulting white solid precipitate removed by filtration. The filtrate was concentrated under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 gradually to 100% ethyl acetate) to afford the title compound **190c** as a white solid (202 mg, 0.65 mmol, 89%).

(**190c**, white solid) m.p. 112 °C – 114 °C; $[\alpha]_D^{20}$ –72.8 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 3234, 2962, 1662, 1641, 1512, 1436, 1330, 1244, 1112; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.24-7.12 (m, 2H, H-13/14), 6.84-6.79 (m, 2H, H-13/14), 4.80 (d, *J*=14.3, 1H, H-10), 4.25 (d, *J*=14.3, 1H, H-10), 4.17 (d, *J*=17.1, 1H, H-11), 3.82 (m, 1H, H-1), 3.75 (s, 3H, H-16), 3.66 (d, *J*=17.1, 1H, H-11), 3.49 (m, 1H, H-1), 2.78 (dd, *J*=16.9, 2.7, 1H, H-5), 2.56 (dd, *J*=16.9, 2.7, 1H, H-5), 2.24-2.07 (m, 2H, H-3), 2.01-1.90 (m, 2H, H-2), 1.86 (t, *J*=2.7, 1H, H-7); ^{13}C NMR (75 MHz, $CDCl_3$) δ 168.0 (C-8/9), 163.1 (C-8/9), 159.3 (C-15), 129.9 (C-13), 127.2 (C-12), 114.0 (C-14), 78.3 (C-6), 72.1 (C-7), 66.6 (C-4), 55.1 (C-16), 50.9 (C-10), 48.9 (C-11), 44.7 (C-1), 35.3 (C-3), 28.2 (C-5), 20.0 (C-2); HRMS (ESI) calculated for $C_{18}H_{20}N_2O_3$ $[M+Na]^+$ 335.1372 found 335.1381.

Proline-Glycine *N*-Boc DKP 190d



190a **190d**

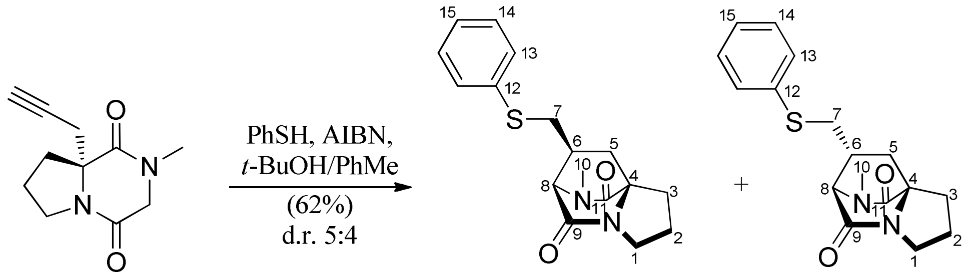
DKP **190a** (134 mg, 0.70 mmol) and Boc_2O (229 mg, 1.05 mmol) were dissolved in THF (10 mL) and treated with triethylamine (146 µL, 1.05 mmol) and 4-DMAP (17 mg, 0.14 mmol). The reaction mixture was allowed to stir at room temperature overnight then concentrated under reduced pressure. The residue was partitioned between water (20 mL) and dichloromethane (20 mL) and the layers separated. The aqueous phase was extracted with dichloromethane (2 x 20 mL) and the combined organic layers dried over $MgSO_4$. The volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 gradually to 100% ethyl acetate) to afford the desired compound **190d** as a colourless oil (168 mg, 0.57 mmol, 82%).

(**190d**, colourless oil) $[\alpha]_D^{20}$ −50.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2984, 1777, 1726, 1661, 1369, 1288, 1147; ^1H NMR (400 MHz, $CDCl_3$) δ 4.42 (d, *J*=2.5, 2H, H-10), 3.77 (m, 1H, H-1), 3.52 (m, 1H, H-1), 2.73 (dd, *J*=17.0, 2.7, 1H, H-5), 2.58 (dd, *J*=17.0, 2.7, 1H, H-5), 2.32-2.16 (m, 2H, H-3), 2.07 (t, *J*=2.7, 1H, H-7), 2.04-1.87 (m, 2H, H-2), 1.48 (s, 9H, H-13); ^{13}C NMR (75 MHz, $CDCl_3$) δ 168.3 (C-8/9), 162.7 (C-8/9), 150.0 (C-11), 84.5 (C-12), 77.7 (C-6), 72.7 (C-7), 67.8 (C-4), 49.7 (C-10), 45.2 (C-1), 35.4 (C-3), 28.0 (C-5), 27.7 (C-13), 20.4 (C-2); HRMS (ESI) calculated for $C_{15}H_{20}N_2O_4$ $[M+Na]^+$ 315.1321 found 315.1325.

General Procedure A: Thiophenol Mediated Radical Cyclisation of DKPs

To a refluxing solution of DKP (1.0 eq.) in *tert*-butanol (50 mL) was added AIBN (2.0 eq.) in toluene (10 mL) and thiophenol (2.0 eq.) in toluene (10 mL) over 20 hours via a syringe pump. Once the addition was complete, the reaction was kept at reflux for a further 2 hours, cooled to room temperature, and concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 gradually to 100% ethyl acetate) to afford the bridged bicyclo[2.2.2]diazaoctanes.

N-Methyl Bridged DKPs 193a and 193b

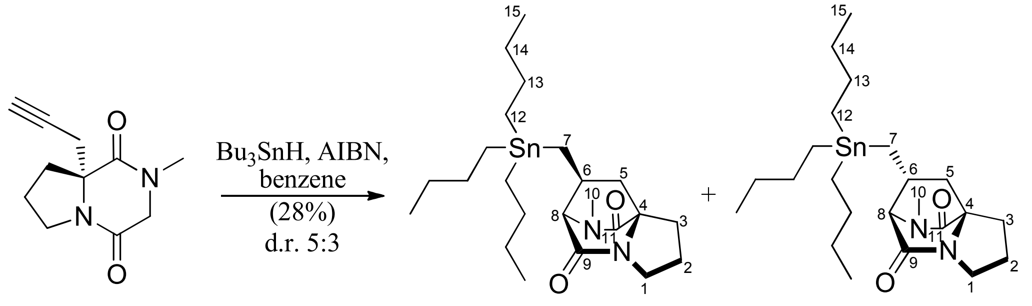


190b **193a** **193b**

General procedure A was followed with DKP **190b** (119 mg, 0.58 mmol), AIBN (189 mg, 1.15 mmol) and thiophenol (127 μL, 1.15 mmol) to afford the title compounds **193a** and **193b** as a colourless oil (114 mg, 0.36 mmol, 62%) and a 5:4 mixture of inseparable diastereomers. (**193a/193b**, colourless oil) $[\alpha]_D^{22}$ +17.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2962, 2882, 1674, 1582, 1482, 1426, 1396, 1209; ^{1}H NMR (500 MHz, $CDCl_3$) (**193a**) δ 7.32 (m, 1H, H-13) 7.26 (t, *J*=7.7, 2H, H-14), 7.20 (m, 2H, H-15), 3.90 (d, *J*=3.3, 1H, H-8), 3.48-3.39 (m, 2H, H-1), 2.97 (s, 3H, H-10), 2.92 (m, 1H, H-7), 2.80 (m, 1H, H-7), 2.70 (dt, *J*=13.3, 6.2, 2H, H-3), 2.39 (m, 1H, H-6), 2.12 (dd, *J*=12.6, 9,1, 1H, H-5), 1.94-1.87 (m, 2H, H-2), 1.71 (m, 1H, H-3), 1.51 (m, 1H, H-5); (**193b**) δ 7.32 (m, 1H, H-13) 7.26 (t, *J*=7.7, 2H, H-14), 7.22-7.17 (m, 2H, H-15), 4.06 (d, *J*=1.4, 1H, H-8), 3.39-3.31 (m, 2H, H-1), 2.91 (s, 3H, H-10), 2.93 (m, 1H, H-

7), 2.76 (m, 1H, H-7), 2.70 (dt, *J*=13.3, 6.2, 2H, H-3), 2.39 (m, 1H, H-6), 2.08 (dd, *J*=12.6, 8.9, 1H, H-5), 1.98-1.90 (m, 2H, H-2), 1.75 (m, 1H, H-3), 1.55 (m, 1H, H-5); ^{13}C NMR (125 MHz, $CDCl_3$) (**193a**) δ 170.7 (C-11), 167.5 (C-9), 134.3 (C-12), 130.5 (C-13), 129.1/129.0 (C-14), 127.0/126.8 (C-15), 67.0 (C-4), 65.0 (C-8), 43.4 (C-1), 37.2 (C-7), 37.0 (C-6), 36.8 (C-5) 33.5 (C-10), 29.0 (C-3), 24.4 (C-2); (**193b**) δ 170.8 (C-11), 166.0 (C-9), 134.5 (C-12), 130.3 (C-13), 129.1/129.0 (C-14), 127.0/126.8 (C-15), 66.8 (C-4), 65.7 (C-8), 43.6 (C-1), 37.6 (C-7), 37.3 (C-6), 36.8 (C-5) 31.9 (C-10), 29.1 (C-3), 24.3 (C-2); HRMS (ESI) calculated for $C_{17}H_{20}N_2O_2S$ $[M+Na]^+$ 339.1143 found 339.1139.

N-Methyl Bridged DKPs 194a and 194b



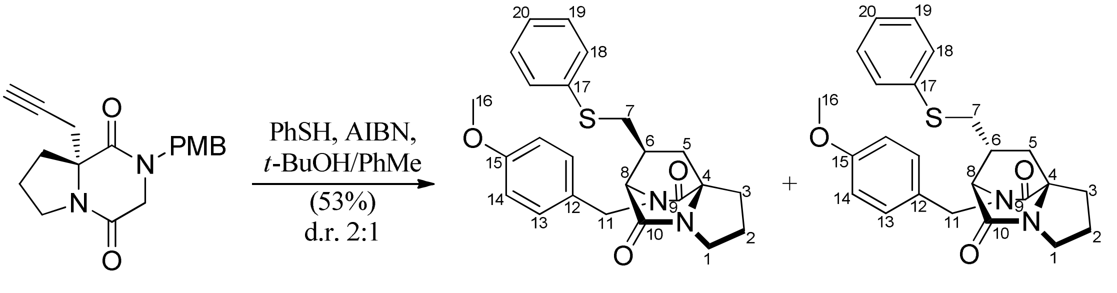
190b **194a** **194b**

Diketopiperazine **190b** (119 mg, 0.58 mmol) was dissolved in benzene (50 mL) and heated at reflux. Solutions of tributyltin hydride (310 μL, 1.15 mmol) in benzene (10 mL) and AIBN (189 mg, 1.15 mmol) in benzene were added over 20 hours via syringe pump then the reaction mixture kept at reflux for a further 2 hours. The volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 then 100% ethyl acetate) to afford the desired compounds **194a** and **194b** as a colourless oil (80 mg, 0.16 mmol, 28%) and 5:3 mixture of diastereoisomers.

(**194a**/**194b**, colourless oil) $[\alpha]_D^{20}$ +13.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2923, 1685, 1422, 1395, 1220, 1069; ^{1}H NMR (500 MHz, $CDCl_3$) (**194a**) δ 3.58 (d, *J*=3.5, 1H, H-8), 3.50-3.43

(m, 2H, H-1), 2.98 (s, 3H, H-10), 2.79-2.64 (m, 2H, H-3), 2.42 (m, 1H, H-6), 2.18 (dd, *J*=12.8, 9.6, 1H, H-5), 1.96 (td, *J*=7.0, 3.5, 2H, H-2), 1.78-1.67 (m, 2H, H-3), 1.52-1.37 (m, 6H, H-13), 1.28 (dd, *J*=14.7, 7.3, 6H, H-14), 1.20 (d, *J*=4.6, 1H, H-5), 0.95 (dd, *J*=13.0, 4.3, 1H, H-7), 0.88 (t, *J*=7.4, 9H, H-15), 0.85-0.80 (m, 6H, H-12), 0.74 (m, 1H, H-7); (**194b,** only signals that differ from **194a**) δ 3.59 (d, *J*=1.4, 1H, H-8), 3.40 (t, *J*=6.8, 2H, H-1), 3.08 (s, 3H, H-10), 2.05 (dd, *J*=12.9, 9.5, 1H, H-5), 1.22 (d, *J*=4.6, 1H, H-5); ^{13}C NMR (125 MHz, $CDCl_3$) (**194a**) δ 171.1 (C-11), 167.1 (C-9), 70.3 (C-8), 67.0 (C-4), 43.7 (C-1), 40.5 (C-5), 36.8 (C-6), 32.2 (C10), 29.6 (C-3), 29.3 (C-13), 27.5 (C-14), 24.6 (C-2), 14.8 (C-7), 13.8 (C-15), 9.4 (C-12); (**194b**) δ 170.9 (C-11), 168.7 (C-9), 71.5 (C-8), 67.5 (C-4), 43.6 (C-1), 40.2 (C-5), 36.6 (C-6), 34.6 (C-10), 29.4 (C-3), 29.2 (C-13), 27.5 (C-14), 24.7 (C-2), 14.4 (C-7), 13.8 (C-15), 9.4 (C-12); HRMS (ESI) calculated for $C_{23}H_{42}N_2O_2$ $[M+Na]^+$ 521.2166 found 521.2176.

N-PMB Bridged DKPs 195a and 195b

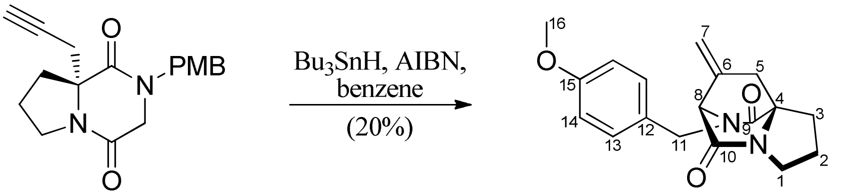


190c **195a** **195b**

General procedure A was followed with DKP **190c** (157 mg, 0.50 mmol), thiophenol (103 μL, 1.00 mmol) and AIBN (165 mg, 1.00 mmol) to afford the title compounds **195a** and **195b** as a colourless oil (114 mg, 0.27 mmol, 53%) and 2:1 mixture of inseparable diastereoisomers. (**195a/195b**, colourless oil) $[\alpha]_D^{20}$ −24.7 (*c* 1.20, $CHCl_3$); FTIR (film) ν_{max} 2930, 1677, 1511, 1414, 1244, 1175, 1026; ^{1}H NMR (400 MHz, $CDCl_3$) (**195a**) δ 7.27-7.09 (m, 7H, H-Ar), 6.77

(m, 1H, H-Ar), 4.81 (d, *J*=14.4, 1H, H-11), 4.10 (d, *J*=14.4, 1H, H-11), 4.04 (d, *J*=3.1, 1H, H-8), 3.78 (s, 3H, H-16), 3.53-3.43 (m, 2H, H-1), 2.85-2.72 (m, 2H, H-3, H-7), 2.66 (d, *J*=7.9, 1H, H-7), 2.27 (m, 1H, H-6), 2.15 (dd, *J*=12.7, 9.6, 1H, H-5), 2.98 (m, 1H, H-2), 1.79 (m, 1H, H-3), 1.52 (m, 1H, H-5); (**195b**, only signals that differ from **195a**) δ 6.84 (m, 1H, H-Ar), 4.59 (d, *J*=14.4, 1H, H-11), 4.36 (d, *J*=14.4, 1H, H-11), 4.10 (d, *J*=1.4, 1H, H-8), 3.76 (s, 3H, H-16), 2.10 (dd, *J*=12.2, 8.4, 1H, H-5); ^{13}C NMR (125 MHz, CDCl$_3$) (**195a**) δ 170.6 (C-10), 166.3 (C-9), 159.3 (C-15), 134.5 (C-17), 130.2 (C-19), 129.5 (C-13), 129.0 (C-18), 127.9 (C-12), 126.7 (C-20), 114.2 (C-14), 67.3 (C-4), 63.3 (C-8), 55.1 (C-16), 48.0 (C-11), 43.7 (C-1), 37.8 (C-6), 36.8 (C-5), 36.6 (C-7), 29.3 (C-3), 24.5 (C-2); (**195b**) δ 170.8 (C-10), 167.9 (C-9), 159.5 (C-15), 134.8 (C-17), 130.1 (C-19), 129.9 (C-13), 129.1 (C-18), 128.3 (C-12), 126.7 (C-20), 114.3 (C-14), 67.1 (C-4), 63.4 (C-8), 55.2 (C-16), 49.2 (C-11), 43.6 (C-1), 37.8 (C-6), 36.8 (C-5), 36.8 (C-7), 29.2 (C-3), 24.5 (C-2); HRMS (ESI) calculated for C$_{24}$H$_{26}$N$_2$O$_3$S [M+Na]$^+$ 445.1562 found 445.1554.

N-PMB Bridged DKP 198

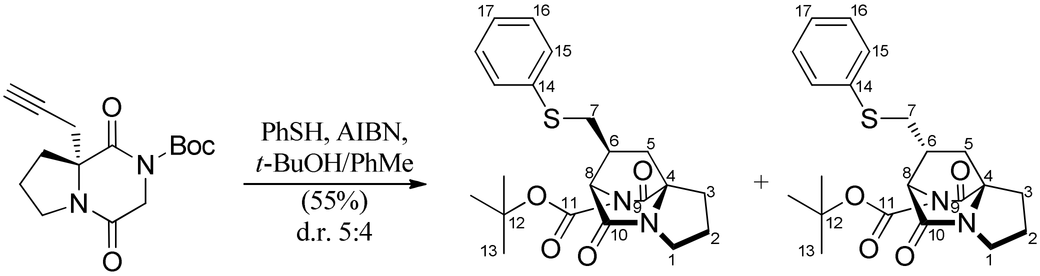


190c **198**

A solution of diketopiperazine **190c** (184 mg, 0.59 mmol) in benzene (50 mL) was heated to reflux and treated with solutions of AIBN (194 mg, 1.18 mmol) in benzene (10 mL) and tributyltin hydride (317 μL, 1.18 mmol) in benzene (10 mL) over 20 hours via syringe pump. Once the addition was complete the reaction mixture was allowed to stir at reflux for a further two hours then cooled to room temperature and concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 gradually to 100% ethyl acetate) to afford the title compound **198** as a colourless oil (37 mg, 0.12 mmol, 20%).

(**198**, colourless oil) $[\alpha]_D^{20}$ −33.6 (*c* 0.25, $CHCl_3$); FTIR (film) ν_{max} 2921, 2852, 1689, 1514, 1413, 1249, 1023; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.12 (d, *J*=8.6, 2H, H-13), 6.83 (d, *J*=8.7, 2H, H-14), 4.97 (t, *J*=2.2, 1H, H-7), 4.91 (t, *J*=2.2, 1H, H-7), 4.63 (d, *J*=14.7, 1H, H-11), 4.41 (d, *J*=14.7, 1H, H-11), 4.15 (s, 1H, H-8), 3.78 (s, 3H, H-16), 3.54-3.36 (m, 2H, H-1), 2.82 (dt, *J*=13.3, 6.8, 1H, H-3), 2.66-2.45 (m, 2H, H-5), 2.09-1.94 (m, 2H, H-2), 1.85 (m, 1H, H-3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.6 (C-10), 166.4 (C-9), 159.5 (C-15), 138.6 (C-6), 129.7 (C-13), 127.9 (C-12), 114.2 (C-14), 112.8 (C-7), 67.6 (C-8), 67.4 (C-4), 55.4 (C-16), 48.1 (C-11), 43.9 (C-1), 37.1 (C-5), 29.0 (C-3), 24.8 (C-2); HRMS (ESI) calculated for $C_{18}H_{20}N_2O_3$ $[M+Na]^+$ 335.1372 found 335.1367.

N-Boc Bridged DKPs 199a and 199b



190d **199a** **199b**

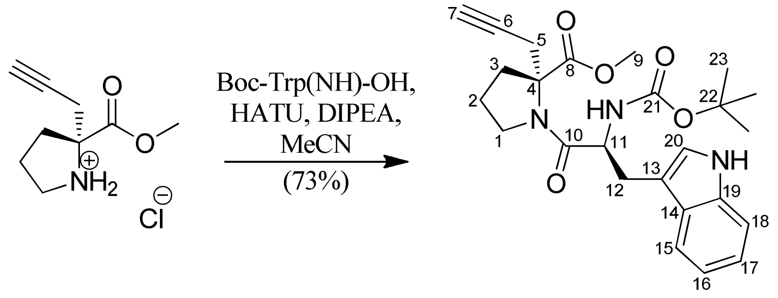
General procedure A was followed with DKP **190d** (168 mg, 0.57 mmol), thiophenol (117 µL, 1.14 mmol) and AIBN (187 mg, 1.14 mmol) to afford the title compounds **199a** and **199b** as a colourless oil (125 mg, 0.31 mmol, 55%) and 5:4 mixture of inseparable diastereoisomers.

(**199a/199b**, colourless oil) $[\alpha]_D^{20}$ +8.80 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2978, 1780, 1692, 1649, 1439, 1366, 1251, 1144; ^1H NMR (400 MHz, $CDCl_3$) δ (**199a**) 7.45-7.18 (m, 5H, H-Ar), 5.03 (d, *J*=3.3, 1H, H-8), 3.59-3.35 (m, 2H, H-1), 3.03 (m, 1H, H-7), 2.84-2.69 (m, 2H, H-3, H-7), 2.44 (m, 1H, H-6), 2.21 (dd, *J*=13.8, 9.8, 1H, H-5), 2.03-1.92 (m, 2H, H-2), 1.79 (m, 1H, H-3), 1.70 (m, 1H, H-5), 1.55 (s, 9H, H-13); (**199b**, only signals that differ from **199a**) δ 5.05 (d, *J*=1.5, 1H, H-8), 2.12 (dd, *J*=13.9, 10.3, 1H, H-5), 1.52 (s, 9H, H-13); ^{13}C NMR (75 MHz, $CDCl_3$) (**199a**) δ 168.7 (C-10), 166.3 (C-9), 149.5 (C-11), 134.3 (C-14), 130.5 (C-15), 129.1 (C-16), 126.9 (C-17), 84.5 (C-12), 68.1 (C-4), 61.0 (C-8), 43.7 (C-1), 36.7 (C-5), 36.1 (C-6), 35.7 (C-7), 29.2 (C-3), 27.8 (C-13), 24.2 (C-2); (**199b**) δ 168.5 (C-10), 165.0 (C-9), 148.0 (C-11), 134.2 (C-14), 130.3 (C-15), 129.0 (C-16), 126.9 (C-17), 84.4 (C-12), 68.0 (C-4), 60.9 (C-8), 43.8 (C-1), 37.7 (C-5), 37.1 (C-6), 35.8 (C-7), 29.3 (C-3), 27.8 (C-13), 24.1 (C-2); HRMS (ESI) calculated for $C_{21}H_{26}N_2O_4S$ $[M+Na]^+$ 425.1511 found 425.1509.

General Procedure B: Peptide Coupling with *N*-Boc Amino Acids

To a stirred suspension of a carboxylic acid (1 eq.), an *N*-Boc amino acid (1 eq.) and HATU (1.1 eq.) in acetonitrile was added diisopropylethylamine (2.1 eq.) causing a dark yellow solution to form. This solution was allowed to stir at room temperature overnight then concentrated under reduced pressure. The resulting brown oily residue was purifed by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 gradually to 1/1) to afford the dipeptide products.

Proline-Tryptophan Dipeptide 201

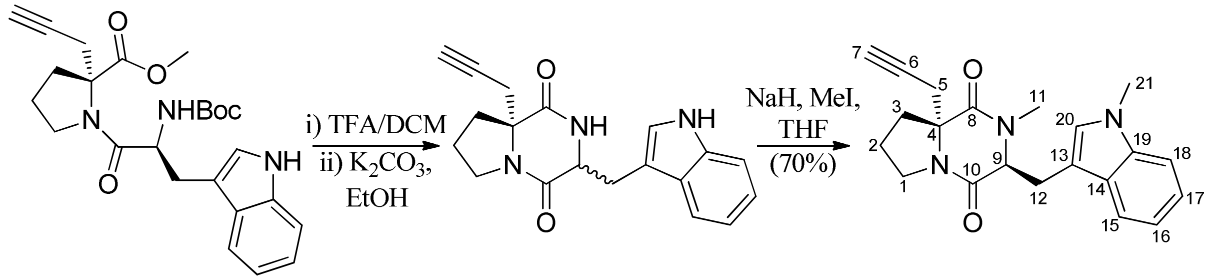


188 **201**

General procedure B was followed with modified proline **188** (1.0 g, 4.91 mmol), Boc-Trp(NH)-OH (1.5 g, 4.91 mmol), HATU (2.0 g, 5.40 mmol) and diisopropylethylamine (1.8 mL, 10.3 mmol) to afford the title compound as a colourless oil (1.63 g, 3.60 mmol, 73%).

(**201**, colourless oil) $[\alpha]_D^{20}$ +17.6 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 3305, 2978, 1738, 1694, 1636, 1433, 1163, 729; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.83 (s, 1H, N-H), 7.67 (d, *J*=7.5, 1H, H-Ar), 7.32 (d, *J*= 7.5, 1H, H-Ar), 7.20-7.01 (m, 3H, H-Ar), 5.40 (d, *J*=9.1, 1H, N-H), 4.82 (dd, *J*=15.3, 7.4, 1H, H-11), 3.71-3.47 (m, 2H, H-1), 3.60 (s, 3H, H-9), 3.36 (dd, *J*=17.2, 2.6, 1H, H-5), 3.20 (dd, *J*=14.6, 6.8, 1H, H-12), 3.06 (dd, *J*=14.6, 6.8, 1H, H-12), 2.83 (dd, *J*=17.2, 2.6, 1H, H-5), 2.37-1.63 (m, 4H, H-2, H-3), 1.92 (t, *J*=2.6, 1H, H-7); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.0 (C-8/10), 170.9 (C-8/10), 155.4 (C-21), 136.2 (C-14/19), 128.0 (C-14/19), 123.4 (CH-Ar), 122.1 (CH-Ar), 119.7 (CH-Ar), 118.9 (CH-Ar), 111.2 (CH-Ar), 110.7 (C-13), 79.6 (C-6), 70.7 (C-7), 67.7 (C-22), 60.5 (C-4), 52.61 (C-9/11), 52.58 (C-9/11), 48.9 (C-1), 36.0 (C-3), 28.7 (C-12), 28.5 (C-23), 24.4 (C-5), 24.0 (C-2); HRMS (ESI) calculated for $C_{25}H_{31}N_3O_5$ $[M+Na]^+$ 476.2161 found 476.2163.

Methylated Proline-Tryptophan DKP 203



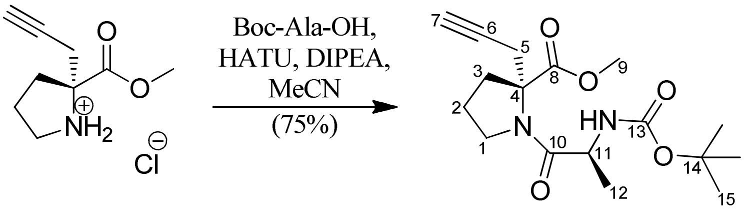
201 **202** **203**

(1:1 +C-9 *epi*)

Dipeptide **201** (1.57 g, 3.47 mmol) was dissolved in dichloromethane (7.6 mL) and treated with trifluoroacetic acid (7.6 mL) then water (80 μL). The resulting mixture was stirred at room temperature for 2 hours then concentrated under reduced pressure. The residue was dissolved in ethanol and exaporated to dryness; then dissolved in toluene and exaporated to dryness. The final residue was dissolved in ethanol and the pH adjusted to 9 with potassium carbonate. The reaction mixture was heated at reflux for 16 hours then cooled to room temperature and filtered through Celite®. The filtrate was concentrated under reduced pressure to afford the DKP **202** as a yellow oil (1.14 g). A solution of this compound in tetrahydrofuran (5 mL) was added to a solution of sodium hydride (60% dispersion in mineral oil; 416 mg, 10.4 mmol) in tetrahydrofuran (10 mL) at 0 °C. Methyl iodide (2.2 mL, 34.7 mmol) was added dropwise and the reaction mixture allowed to warm to room temperature and stir overnight. Water (1 mL) was added and the volatiles removed under reduced pressure. The residue was partitioned between water (20 mL) and dichloromethane (20 mL) and the layers separated. The aqueous phase was extracted with dichloromethane (2 x 20 mL) and the combined organic layers dried over $MgSO_4$ then concentrated under reduced pressure to afford the desired *N*-methyl DKP as an off-white foam (824 mg, 2.36 mmol, 70%) and 1:1 mixture of diastereoisomers. A portion of the material was purified by preparative HPLC to afford the desired diastereomer **203** as a white solid.

(**203**, white solid) m.p. 211 °C (dec.); $[\alpha]_D^{20}$ +3.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3192, 2928, 1642, 1547, 1455, 1428, 1404, 1326; ^1H NMR (300 MHz, $CDCl_3$) δ 7.67-7.62 (m, 1H, H-Ar), 7.35-7.03 (m, 3H, H-Ar), 6.76 (s, 1H, H-20), 4.33 (dd, *J*=4.3, 2.5, 1H, H-9), 3.73 (s, 3H, H-21), 3.72 (m, 2H, H-12), 3.46 (m, 1H, H-1), 3.28 (dd, *J*=14.8, 4.5, 1H, H-12), 3.16 (s, 3H, H-11), 3.06 (m, 1H, H-1), 2.61 (dd, *J*=16.7, 2.6, 1H, H-5), 2.29 (dd, *J*=16.7, 2.6, 1H, H-5), 2.00 (t, *J*=2.6, 1H, H-7), 1.66-1.25 (m, 4H, H-2, H-3); ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.8 (C-8/10), 164.5 (C-8/10), 136.6 (C-14/19), 128.3 (C-14/19), 127.9 (C-20), 122.0 (CH-Ar), 120.0 (CH-Ar), 119.2 (CH-Ar), 109.0 (CH-Ar), 107.0 (C-13), 78.5 (C-6), 72.1 (C-7), 66.7 (C-4), 63.4 (C-9), 43.6 (C-1), 33.6 (C-3), 32.8 (C-11/21), 32.6 (C-11/21), 28.9 (C-12), 27.7 (C-5), 18.9 (C-2); HRMS (ESI) calculated for $C_{21}H_{23}N_3O_2$ $[M+Na]^+$ 372.1688 found 372.1696.

Proline-Alanine Dipeptide 204a

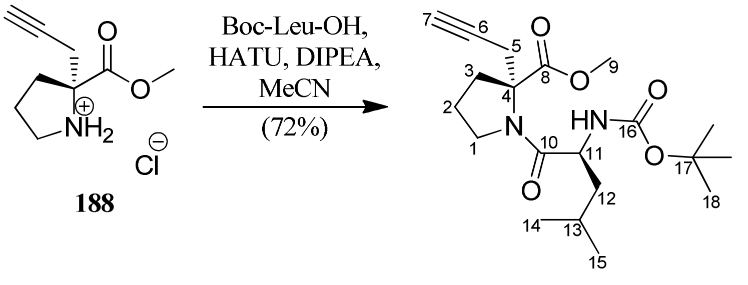


188 **204a**

General procedure B was followed with modified proline **188** (1.03 g, 5.06 mmol), Boc-Ala-OH (957 mg, 5.06 mmol), HATU (2.11 g, 5.57 mmol) and diisopropylethylamine (1.85 mL, 10.6 mmol) to afford the title compound **204a** as a colourless oil (1.29 g, 3.80 mmol, 75%).

(**204a**, colourless oil) $[\alpha]_D^{20}$ +14.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3307, 2985, 2887, 1741, 1702, 1645, 1432, 1243, 1161, 1025; ^1H NMR (400 MHz, $CDCl_3$) δ 5.38 (d, *J*=8.2, 1H, N-H), 4.45 (m, 1H, H-11), 3.74-3.63 (m, 2H, H-1), 3.65 (s, 3H, H-9), 3.35 (dd, *J*=17.2, 2.7, 1H, H-5), 2.83 (dd, *J*=17.2, 2.7, 1H, H-5), 2.33 (m, 1H, H-2), 2.15 (m, 1H, H-3), 2.08 (m, 1H, H-2), 1.99 (m, 1H, H-3), 1.92 (t, *J*=2.7, 1H, H-7), 1.38 (s, 9H, H-15), 1.26 (d, *J*=6.8, 3H, H-12); ^{13}C NMR (75 MHz, $CDCl_3$) δ 172.7 (C-8/10), 171.3 (C-8/10), 155.0 (C-13), 80.0 (C-6), 79.4 (C-14), 70.7 (C-7), 67.7 (C-4), 52.5 (C-9), 48.7 (C-1), 47.9 (C-11), 35.7 (C-3), 28.3 (C-15), 24.5 (C-5), 23.9 (C-2), 17.9 (C-12); HRMS (ESI) calculated for $C_{17}H_{26}N_2O_5$ $[M+Na]^+$ 361.1739 found 361.1747.

Proline-Leucine Dipeptide 204b



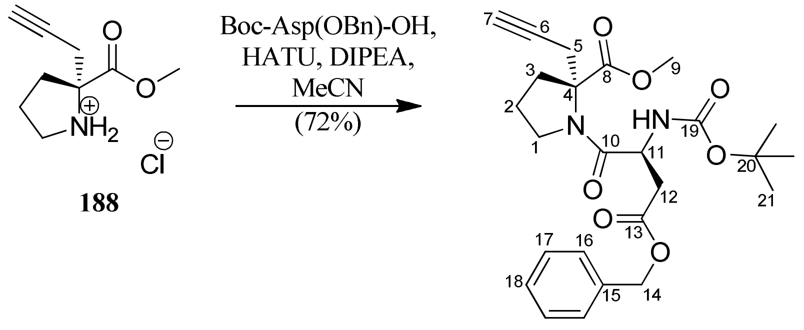
204b

General procedure B was followed with modified proline **188** (640 mg, 3.14 mmol), Boc-Leu-OH (727 mg, 3.14 mmol), HATU (1.31 g, 3.45 mmol) and diisopropylethylamine (1.15 mL, 6.59 mmol) to afford the title compound **204b** as a slightly yellow oil (864 mg, 2.27 mmol, 72%).

(**204b**, yellow oil) $[\alpha]_D^{20}$ +11.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3329, 3292, 3226, 2956, 1738, 1690, 1644, 1526, 1424, 1246, 1160, 1048; ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.23 (s, br, N-H), 4.43 (q, *J*=8.5, 1H, H-11), 3.76 (m, 1H, H-1), 3.63 (m, 1H, H-1), 3.60 (s, 3H, H-9), 3.29 (dd, *J*=17.2, 2.4, 1H, H-5), 2.78 (dd, *J*=17.2, 2.4, 1H, H-5), 2.20 (m, 1H, H-3), 2.15 (m, 1H, H-2), 2.05 (m, 1H, H-3), 1.97 (m, 1H, H-2), 1.87 (t, *J*=2.4, 1H, H-7), 1.63 (m, 1H, H-13), 1.36 (s, 2H, H-12), 1.34 (s, 9H, H-18), 0.91 (d, *J*=6.5, 3H, H-14/15), 0.87 (d, *J*=6.5, 3H, H-14/15); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.0 (C-8/10), 171.7 (C-8/10), 155.7 (C-16), 80.2 (C-6), 79.6 (C-17), 70.7 (C-7), 67.9 (C-4), 52.6 (C-9), 50.6 (C-11), 48.9 (C-1), 41.8 (C-3), 36.0

(C-12), 28.5 (C-18), 24.6 (C-5), 24.1 (C-2), 23.5 (C-14/15), 23.0 (C-13), 22.1 (C-14/15); HRMS (ESI) calculated for $C_{20}H_{32}N_2O_5$ $[M+Na]^+$ 403.2209 found 403.2211.

Proline-Aspartic Acid *O*-Benzyl Ester Dipeptide 204c



188

204c

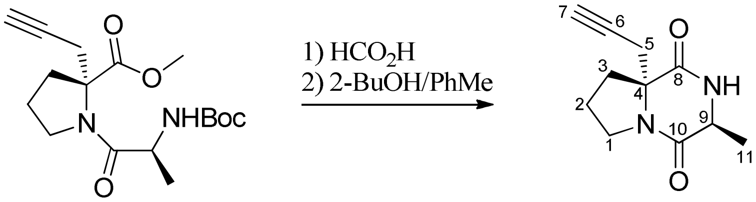
General procedure B was followed with modified proline **188** (1.0 g, 4.91 mmol), Boc-Asp(OBn)-OH (1.59 g, 4.91 mmol), HATU (2.05 g, 5.40 mmol) and diisopropylethylamine (1.80 mL, 10.31 mmol) to afford the title compound **204c** as a colourless oil (1.43 g, 3.26 mmol, 72%).

(**204c**, colourless oil) $[\alpha]_D^{20}$ +5.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3300, 2976, 1735, 1646, 1434, 1244, 1161, 1022; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32-7.16 (m, 5H, H-Ar), 5.38 (d, *J*=9.7, 1H, N-H), 5.05 (d, *J*=4.4, 2H, H-14), 4.84 (dt, *J*=9.6, 6.6, 1H, H-11), 3.83 (m, 1H, H-1), 3.68 (m, 1H, H-1), 3.55 (s, 3H, H-9), 3.32 (dd, *J*=17.2, 2.6, 1H, H-5), 2.81-2.69 (m, 2H, H-5, H-12), 2.58 (dd, *J*=16.1, 5.9, 1H, H-12), 2.26-1.87 (m, 4H, H-2, H-3), 1.85 (t, *J*=2.6, 1H, H-7), 1.34 (s, 9H, H-21); ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.8 (C=O), 170.5 (C=O), 169.4 (C=O), 155.1 (C=O), 135.8 (C-15), 128.6 (CH-Ar),128.4 (CH-Ar), 128.3 (CH-Ar), 80.2 (C-6), 80.1 (C-4), 70.6 (C-7), 67.8 (C-20), 66.7 (C-14), 52.6 (C-9), 49.3 (C-1, C-11), 37.2 (C-12), 35.9 (C-3), 28.4 (C-21), 24.3 (C-5), 24.1 (C-2); HRMS (ESI) calculated for $C_{25}H_{32}N_2O_7$ $[M+Na]^+$ 495.2107 found 495.2105.

General Procedure C: DKP Formation from Dipeptides

The dipeptide was dissolved in formic acid (10 mL/g) and allowed to stir at room temperature until TLC indicated complete consumption of the starting material (ca. 3-4 hours). The volatiles were removed under reduced pressure and the residue dissolved in toluene (7 mL/g) and 2-butanol (20 mL/g). The resulting solution was heated at reflux for 2 hours then concentrated under reduced pressure to afford the DKP that could be used without further purification.

Proline-Alanine *N*-H DKP 206a

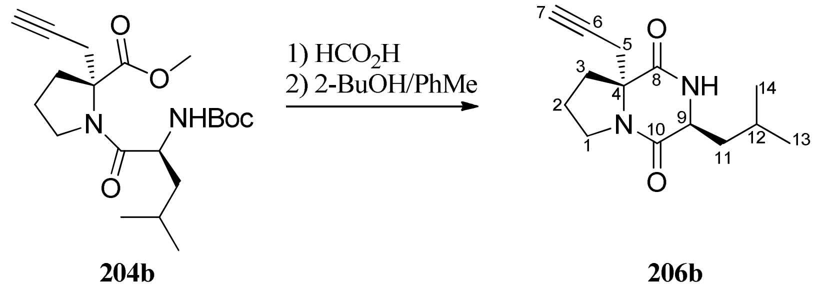


204a **206a**

General procedure C was followed with dipeptide **204a** (820 mg, 2.68 mmol) to afford the title compound **206a** as a white solid (550 mg, ~quantitative).

(**206a**, white solid) m.p. 109 °C – 111 °C; $[\alpha]_D^{20}$ −49.6 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 3242, 2982, 2893, 2245, 1645, 1420, 1160, 1128, 727; ^1H NMR (300 MHz, $CDCl_3$) δ 7.65 (s, br, 1H, N-H), 4.39 (q, *J*=6.9, 1H, H-9), 3.78 (m, 1H, H-1), 3.48 (m, 1H, H-1), 2.70 (dd, *J*=17.0, 2.7, 1H, H-5), 2.55 (dd, *J*=17.0, 2.7, 1H, H-5), 2.16-2.07 (m, 2H, H-3), 2.02 (t, *J*=2.7, 1H, H-7), 2.00-1.86 (m, 2H, H-2), 1.38 (d, *J*=6.9, 3H, H-11); ^{13}C NMR (75 MHz, $CDCl_3$) δ 171.1 (C-8/10), 166.6 (C-8/10), 78.4 (C-6), 72.0 (C-7), 67.0 (C-4), 51.1 (C-9), 45.1 (C-1), 34.6 (C-3), 27.3 (C-5), 20.1 (C-2), 17.0 (C-11); HRMS (ESI) calculated for $C_{11}H_{14}N_2O_2$ $[M+Na]^+$ 229.0953 found 229.0955.

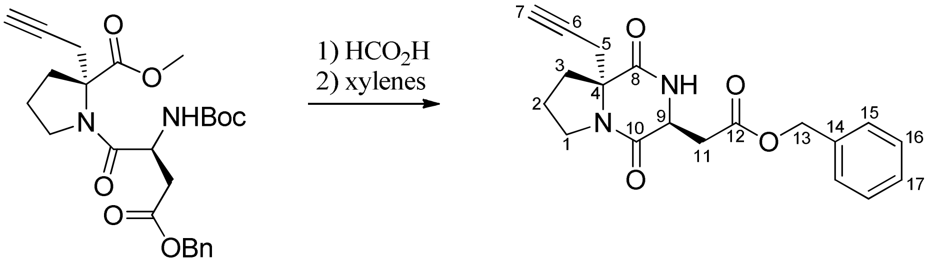
Proline-Leucine *N*-H DKP 206b



General procedure C was followed with dipeptide **204b** (680 mg, 1.79 mmol) to afford the title compound **206b** as an off-white solid (500 mg, ~quantitative).

(**206b**, off-white solid) m.p. 103 °C – 105 °C; $[\alpha]_D^{20}$ −79.2 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 3310, 3242, 2958, 2878, 1651, 1422, 910; ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.63 (s, br, 1H, N-H), 4.38 (dd, *J*=9.4, 3.8, 1H, H-9), 3.85 (m, 1H, H-1), 3.56 (m, 1H, H-1), 2.77 (dd, *J*=16.9, 2.7, 1H, H-5), 2.63 (dd, *J*=16.9, 2.7, 1H, H-5), 2.27-2.14 (m, 2H, H-3), 2.06 (t, *J*=2.7, 1H, H-7), 2.03 (m, 1H, H-11), 2.00-1.92 (m, 2H, H-2), 1.55 (m, 1H, H-11), 1.49-1.41 (m, 2H, H-12), 0.97 (d, *J*=6.6, 3H, H-13/14), 0.93 (d, *J*=6.6, 3H, H-13/14); ^{13}C NMR (75 MHz, $CDCl_3$) δ 171.2 (C-8/10), 166.2 (C-8/10), 78.4 (C-6), 72.1 (C-7), 66.0 (C-4), 53.6 (C-9), 45.2 (C-1), 39.8 (C-11), 34.8 (C-3), 27.6 (C-5), 24.4 (C-12), 23.2 (C-13/14), 21.1 (C-13/14), 20.2 (C-2); HRMS (ESI) calculated for $C_{14}H_{20}N_2O_2$ $[M+Na]^+$ 271.1422 found 271.1425.

Proline-Aspartic Acid *N*-H DKP 206c



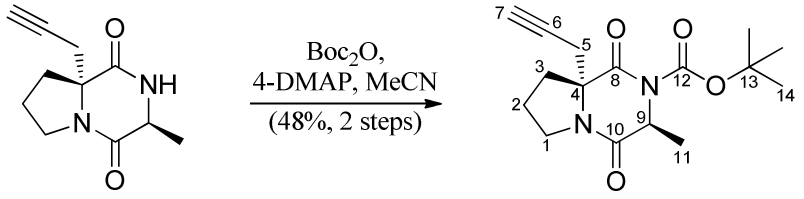
Dipeptide **204c** (1.48 g, 3.13 mmol) was dissolved in formic acid (10 mL) and allowed to stir at room temperature for 4 hours then concentrated under reduced pressure. The residue was dissolved in xylene (30 mL, mixture of isomers) and 2-butanol (5 mL) then heated at reflux for 3 hours. The volatiles were removed under reduced pressue to afford the desired compound **206c** as an orange oil that could be used without further purifcation (1.33 g, ~quantitative)

(**206c**, orange oil) $[\alpha]_D^{20}$ −85.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2963, 1716, 1483, 1396, 1258, 1008; ^1H NMR (400 MHz, $CDCl_3$) δ 7.33-7.21 (m, 5H, H-Ar), 5.07 (d, *J*=2.2, 2H, H-13), 4.66 (dd, *J*=7.8, 4.1, 1H, H-10), 3.74 (dt, *J*=12.3, 8.4, 1H, H-1), 3.54 (d, *J*=4.8, 1H, N-H), 3.45 (m, 1H, H-1), 3.11 (dd, *J*=17.4, 4.1, 1H, H-11), 2.73 (dd, *J*=17.4, 7.8, 1H, H-11), 2.68 (dd, *J*=17.0, 2.6, 1H, H-5), 2.51 (dd, *J*=17.0, 2.6, 1H, H-5), 2.05-1.99 (m, 2H, H-3), 1.98 (t, *J*=2.6, 1H, H-7), 1.92-1.74 (m, 2H, H-2); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.7 (C-8/9), 170.5 (C-8/9), 164.7 (C-12), 135.4 (C-14), 128.7 (C-15/16/17), 128.5 (C-15/16/17), 128.4 (C-15/16/17), 78.5 (C-6), 72.4 (C-7), 67.1 (C-4), 67.0 (C-13), 52.4 (C-10), 45.2 (C-1), 36.4 (C-11), 34.8 (C-3), 28.0 (C-5), 20.1 (C-2); HRMS (ESI) calculated for $C_{19}H_{20}N_2O_4$ $[M+Na]^+$ 363.1321 found 363.1315.

General Procedure D: *N*-Boc Protection of DKPs

A solution of DKP (1.0 eq.), 4-DMAP (0.1 eq.) and Boc_2O (1.7 eq.) in acetonitrile (20 mL/g) was allowed to stir at room temperature overnight. The mixture was diluted with ethyl acetate and washed with HCl (0.5 N aq. soln.), $NaHCO_3$ (sat. aq. soln.) and NaCl (sat. aq. soln.) then dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 gradually to 3/7) to afford the desired *N*-Boc DKPs.

Proline-Alanine *N*-Boc DKP 207a

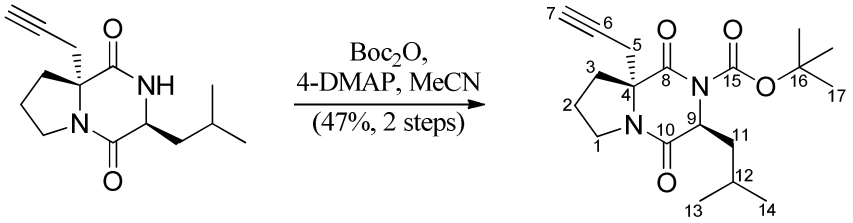


206a **207a**

General procedure D was followed with DKP **206a** (203 mg, 0.99 mmol), 4-DMAP (12 mg, 0.10 mmol) and Boc$_2$O (367 mg, 1.68 mmol) to afford the title compound **207a** as a colourless oil (145 mg, 0.47 mmol, 48% from **204a**).

(**207a**, colourless oil) $[\alpha]_D^{22}$ +31.5 (*c* 1.00, CHCl$_3$); FTIR (film) ν_{max} 2986, 1741, 1685, 1655, 1391, 1372, 1248, 1146; ^{1}H NMR (300 MHz, CDCl$_3$) δ 4.74 (q, *J*=6.7, 1H, H-9), 3.86 (m, 1H, H-1), 3.50 (m, 1H, H-1), 2.70 (dd, *J*=17.0, 2.7, 1H, H-5), 2.58 (dd, *J*=17.0, 2.7, 1H, H-5), 2.24-2.07 (m, 2H, H-3), 2.05 (t, *J*=2.7, 1H, H-7), 2.01-1.84 (m, 2H, H-2), 1.49 (s, 9H, H-14), 1.48 (d, *J*=6.8, 3H, H-11); ^{13}C NMR (75 MHz, CDCl$_3$) δ 168.6 (C-8/10), 165.1 (C-8/10), 150.9 (C-12), 84.6 (C-6), 77.7 (C-13), 72.6 (C-7), 67.4 (C-4), 55.6 (C-9), 44.9 (C-1), 35.0 (C-3), 28.8 (C-5), 27.6 (C-14), 19.7 (C-2), 18.2 (C-11); HRMS (ESI) calculated for $C_{16}H_{22}N_2O_4$ $[M+Na]^+$ 329.1477 found 329.1471.

Proline-Leucine *N*-Boc DKP 207b

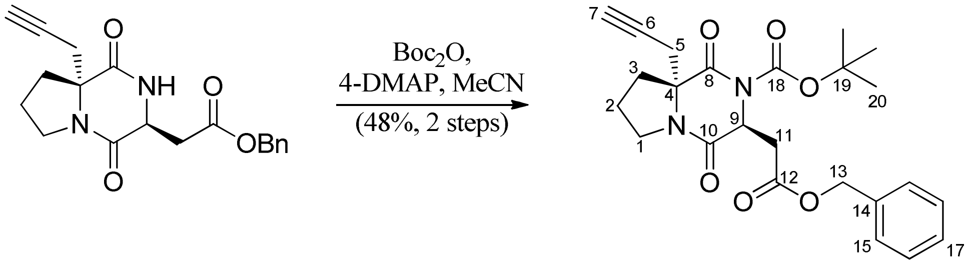


206b **207b**

General procedure D was followed with DKP **206b** (276 mg, 1.11 mmol), 4-DMAP (14 mg, 0.11 mmol) and Boc_2O (412 mg, 1.89 mmol) to afford the title compound **207b** as a white solid (180 mg, 0.52 mmol, 47% from **204b**).

(**207b**, white solid) m.p. 82 °C – 85 °C; $[\alpha]_D^{20}$ +68.8 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3274, 2960, 2872, 1777, 1726, 1662, 1368, 1245, 1155; ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.79 (dd, *J*=5.2, 4.3, 1H, H-9), 3.88 (m, 1H, H-1), 3.52 (m, 1H, H-1), 2.70 (dd, *J*=16.9, 2.6, 1H, H-5), 2.60 (dd, *J*=16.9, 2.6, 1H, H-5), 2.19-2.09 (m, 2H, H-3), 2.06 (t, *J*=2.6, 1H, H-7), 2.02-1.92 (m, 2H, H-11), 1.92-1.88 (m, 2H, H-2), 1.72 (m, 1H, H-12), 1.52 (s, 9H, H-17), 0.88 (d, *J*=6.6, 3H, H-13/14), 0.81 (d, *J*=6.6, 3H, H-13/14); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.8 (C-8/10), 165.0 (C-8/10), 150.5 (C-15), 84.2 (C-6), 77.6 (C-16), 72.7 (C-7), 67.7 (C-4), 59.1 (C-9), 44.7 (C-1), 41.2 (C-3), 34.7 (C-11), 29.4 (C-5), 27.8 (C-17), 24.5 (C-12), 23.6 (C-13/14), 22.0 (C-13/14), 19.2 (C-2); HRMS (ESI) calculated for $C_{19}H_{28}N_2O_4$ $[M+Na]^+$ 371.1947 found 371.1942.

Proline-Aspartic Acid *N*-Boc DKP 207c

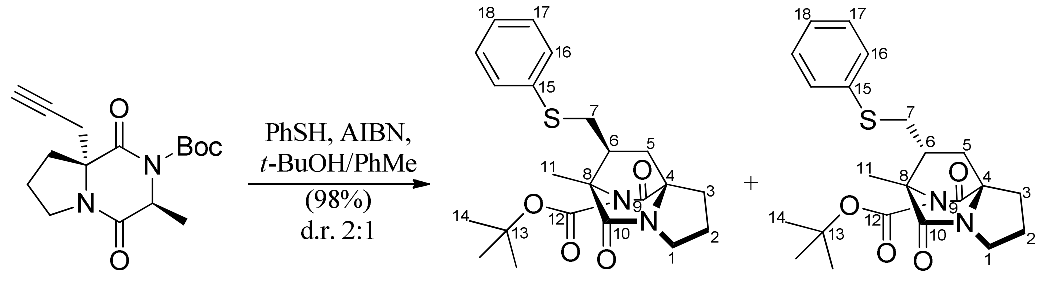


206c **207c**

General procedure D was followed with DKP **206c** (1.33 g, 3.91 mmol), 4-DMAP (48 mg, 0.39 mmol) and Boc_2O (1.45 g, 6.65 mmol) to afford the title compound **207c** as a yellow oil (821 mg, 1.87 mmol, 48% from **204c**).

(**207c**, yellow oil) $[\alpha]_D^{20}$ +153.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3259, 2979, 1727, 1656, 1455, 1369, 1242, 1148; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.36-7.24 (m, 5H, H-Ar), 5.02 (d, *J*=7.9, 2H, H-13), 4.84 (dd, *J*=3.8, 3.2, 1H, H-9), 3.87 (dt, *J*=12.3, 8.8, 1H, H-1), 3.48 (m, 1H, H-1), 3.37 (dd, *J*=18.1, 3.1, 1H, H-5), 3.09 (dd, *J*=18.1, 3.1, 1H, H-5), 2.63 (dd, *J*=2.6, 1.1, 2H, H-11), 2.32 (m, 1H, H-3), 2.06 (t, 1H, *J*=3.1, H-7), 2.03 (m, 1H, H-3), 1.99-1.81 (m, 2H, H-2), 1.49 (s, 9H, H-20); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.9 (C-8/10/12), 169.1 (C-8/10/12), 164.5 (C-8/10/12), 150.4 (C-18), 135.2 (C-14), 128.6 (C-15/16/17), 128.4 (C-15/16/17), 128.3 (C-15/16/17), 84.1 (C-6), 77.9 (C-19), 72.3 (C-7), 68.8 (C-4), 66.8 (C-13), 56.6 (C-9), 45.6 (C-1), 36.8 (C-3), 33.8 (C-11), 29.9 (C-5), 27.9 (C-20), 19.8 (C-2); HRMS (ESI) calculated for $C_{24}H_{28}N_2O_6$ $[M+Na]^+$ 463.1845 found 463.1854.

Alanine-Derived Bridged DKPs 208a and 208b

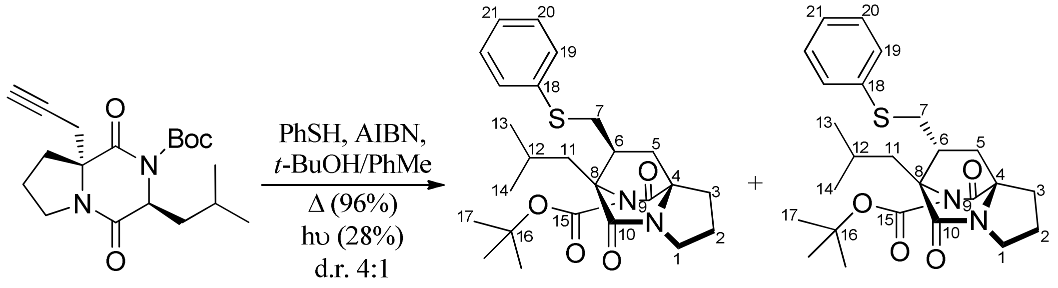


207a **208a** **208b**

General procedure A was followed with DKP **207a** (145 mg, 0.47 mmol), thiophenol (97 µL, 0.95 mmol) and AIBN (156 mg, 0.95 mmol) to afford the title compounds **208a** and **208b** as a colourless oil (194 mg, 0.47 mmol, 98%) and 2:1 mixture of inseparable diastereoisomers.

(**208a**/**208b**, colourless oil) $[\alpha]_D^{20}$ +12.5 (*c* 0.80, $CHCl_3$); FTIR (film) ν_{max} 2985, 2938, 1772, 1721, 1689, 1369, 1254, 1143; ^{1}H NMR (400 MHz, $CDCl_3$) (**208a only**) δ 7.39-7.03 (m, 5H, H-Ar), 3.60-3.37 (m, 2H, H-1), 3.21 (dd, *J*=12.5, 3.4, 1H, H-7), 2.73 (m, 1H, H-3), 2.55 (m, 1H, H-6), 2.35 (t, *J*=12.3, 1H, H-7), 2.24 (dd, *J*=13.8, 9.6, 1H, H-5), 1.99-1.88 (m, 2H, H-2), 1.81 (m, 1H, H-3), 1.62 (s, 3H, H-11), 1.46 (s, 9H, H-14); ^{13}C NMR (75 MHz, $CDCl_3$) (**208a**) δ 170.0 (C-9/10), 165.8 (C-9/10), 150.2 (C-12), 134.6 (C-15), 129.7 (C-Ar), 129.0 (C-Ar), 126.6 (C-Ar), 84.6 (C-13), 66.5 (C-4), 65.5 (C-8), 44.4 (C-1), 42.5 (C-6), 37.0 (C-5), 36.8 (C-7), 29.5 (C-3), 27.7 (C-14), 23.9 (C-2), 15.7 (C-11); (**208b**) δ 169.6 (C-9/10), 167.4 (C-9/10), 150.4 (C-12), 134.6 (C-15), 129.7 (C-Ar), 129.0 (C-Ar), 126.6 (C-Ar), 84.6 (C-13), 66.3 (C-4), 66.1 (C-8), 44.4 (C-1), 40.8 (C-6), 37.0 (C-5), 35.6 (C-7), 29.5 (C-3), 27.7 (C-14), 23.9 (C-2), 14.9 (C-11); HRMS (ESI) calculated for $C_{22}H_{28}N_2O_4S$ $[M+Na]^+$ 439.1667 found 439.1660.

Leucine-Derived Bridged DKPs 209a and 209b



207b **209a** **209b**

Thermal Initiation

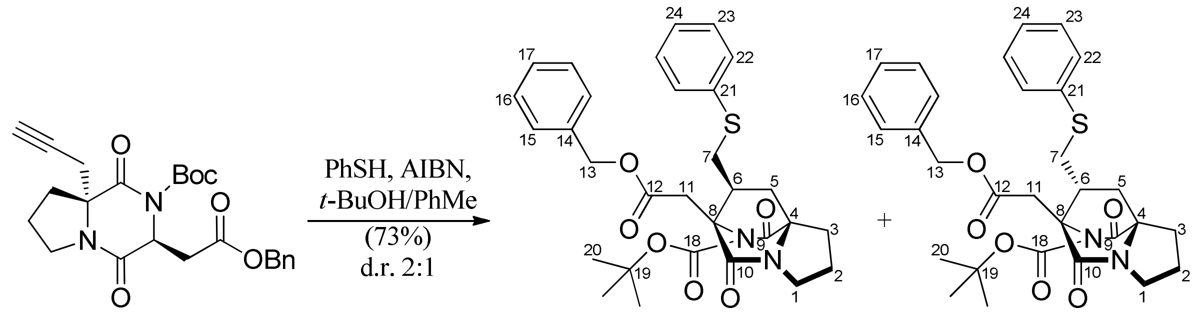
General procedure A was followed with DKP **207b** (180 mg, 0.52 mmol), thiophenol (106 μL, 1.03 mmol) and AIBN (169 mg, 1.03 mmol) to afford the title compounds **209a** and **209b** as a colourless oil (227 mg, 0.50 mmol, 96 %) and 4:1 mixture of inseparable diastereoisomers.

Photolytic Initiation

A solution of DKP **207b** (146 mg, 0.42 mmol) in toluene (20 mL) was treated with AIBN (138 mg, 0.84 mmol) in toluene (5 mL) and thiophenol (86 μL, 0.84 mmol) in toluene (5 mL) over 20 hours via syringe pump. Throughout this period the mixture was irradiated with a 300W sunlamp while the temperature was maintained below 35 °C. Once the addition was complete the mixture was irradiated for a further two hours then concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 to 1/1) to afford the title compounds **209a** and **209b** as a colourless oil (53 mg, 0.12 mmol, 28%) and 4:1 mixture of inseparable diastereoisomers.

(**209a**/**209b**, colourless oil) $[\alpha]_D^{20}$ +38.8 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2978, 1723, 1687, 1428, 1368, 1253, 1146; ^{1}H NMR (300 MHz, $CDCl_3$) (**209a only**) δ 7.43-7.15 (m, 5H, H-Ar), 3.68-3.40 (m, 2H, H-1), 3.24 (dd, *J*=12.2, 3.2, 1H, H-7), 2.82-2.67 (m, 2H, H-3, H-6), 2.43-2.31 (m, 2H, H-7, H-11), 2.24 (dd, *J*=13.8, 9.4, 1H, H-5), 2.05-1.92 (m, 3H, 2 x H-2, H-11), 1.90-1.76 (m, 2H, H-3, H-5), 1.62 (m, 1H, H-12), 1.51 (s, 9H, H-17), 0.97 (s, 3H, H-13/14), 0.95 (s, 3H, H-13/14); ^{13}C NMR (100 MHz, $CDCl_3$) (**209a only**) δ 171.4 (C-9/10), 165.7 (C-9/10), 151.4 (C-15), 134.9 (C-18), 129.9 (CH-Ar), 129.0 (CH-Ar), 126.7 (CH-Ar), 84.7 (C-16), 70.2 (C-4), 66.2 (C-8), 44.3 (C-1), 41.8 (C-6), 37.3 (C-5), 37.1 (C-7), 35.5 (C-11), 29.6 (C-3), 27.7 (C-17), 24.8 (C-13/14), 24.7 (C-12), 23.8 (C-2), 22.9 (C-13/14); HRMS (ESI) calculated for $C_{25}H_{34}N_2O_4S$ $[M+Na]^+$ 481.2131 found 481.2137.

Aspartic Acid-Derived Bridged DKPs 210a and 210b

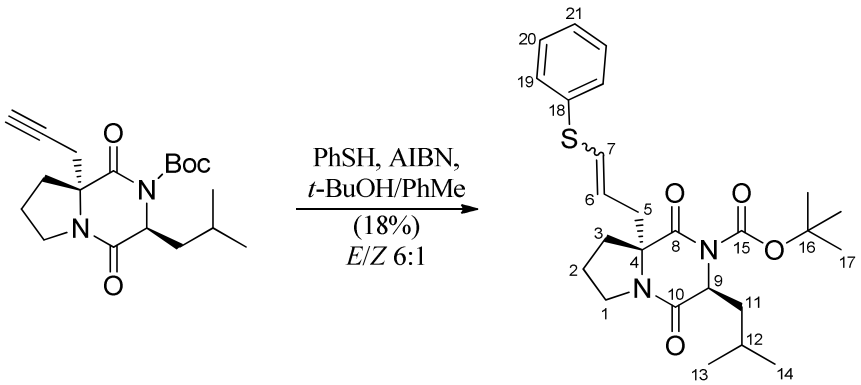


207c **210a** **210b**

General procedure A was followed with DKP **207c** (169 mg, 0.38 mmol), thiophenol (79 μL, 0.77 mmol) and AIBN (126 mg, 0.77 mmol) to afford the title compounds **210a** and **210b** as a colourless oil (155 mg, 0.28 mmol, 73 %) and 2:1 mixture of inseparable diastereoisomers.

(**210a**/**210b**, colourless oil) $[\alpha]_D^{20}$ +37.6 (*c* 0.80, $CHCl_3$); FTIR (film) ν_{max} 2979, 1724, 1692, 1390, 1363, 1275, 1257, 1148; ^{1}H NMR (300 MHz, $CDCl_3$) (**210a only**) δ 7.36-7.05 (m, 10H, H-Ar), 5.03 (s, 2H, H-13), 3.67 (d, *J*=17.1, 1H, H-11), 3.57-3.33 (m, 2H, H-1), 3.33 (dd, *J*=13.0, 3.9, 1H, H-7), 3.12 (dd, *J*=12.5, 3.7, 1H, H-5), 2.90 (d, *J*=17.1, 2H, H-11), 2.71 (td, *J*=13.2, 6.6, 1H, H-3), 2.65-2.53 (m, 2H, H-6, H-7), 2.28 (dd, 1H, *J*=13.8, 9.7, H-5), 1.88 (dt, *J*=9.8, 5.3, 2H, H-2), 1.76 (m, 1H, H-3), 1.37 (s, 9H, H-20, major); ^{13}C NMR (100 MHz, $CDCl_3$) (**210a**) δ 169.5 (C-9/C-10), 169.2 (C-9/C-10), 164.4 (C-12), 151.1 (C-18), 135.8 (C-14/C-21), 134.6 (C-14/C-21), 130.2 (CH-Ar), 130.1 (CH-Ar), 129.3 (CH-Ar), 128.6 (CH-Ar), 128.2 (CH-Ar), 127.1 (CH-Ar), 84.5 (C-19), 67.4 (C-4), 66.6 (C-13), 60.5 (C-8), 44.7 (C-1), 41.7 (C-6), 37.1 (C-7), 37.0 (C-5), 33.6 (C-11), 29.9 (C-3), 27.8 (C-20), 24.0 (C-2); (**210b**) δ 170.1 (C-9/C-10), 169.1 (C-9/C-10), 166.0 (C-12), 151.6 (C-18), 135.9 (C-14/C-21), 134.5 (C-14/C-21), 130.2 (CH-Ar), 130.1 (CH-Ar), 129.3 (CH-Ar), 128.6 (CH-Ar), 128.2 (CH-Ar), 127.1 (CH-Ar), 84.6 (C-19), 67.8 (C-4), 66.6 (C-13), 66.0 (C-8), 44.6 (C-1), 40.7 (C-6), 37.3 (C-7), 36.1 (C-5), 33.8 (C-11), 29.8 (C-3), 27.8 (C-20), 24.0 (C-2); HRMS (ESI) calculated for $C_{30}H_{34}N_2O_6S$ $[M+Na]^+$ 573.2035 found 573.2040.

Leucine-Derived Vinyl Sulfide 211

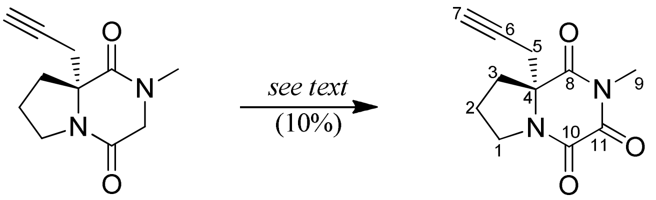


207b **211**

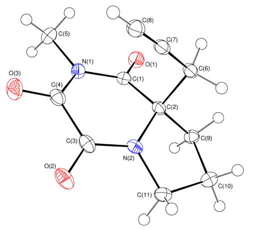
A solution of DKP **207b** (117 mg, 0.34 mmol) and thiophenol (52 µL, 0.50 mmol) in toluene (20 mL) was irradiated with a 400W mercury lamp with the temperature maintained below 35 °C. After 5 hours the volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 to 1/1) to afford the title compound **211** as a colourless oil (28 mg, 0.06 mmol, 18%) and 6:1 mixture of *E*:*Z* isomers.

(**211**, colourless oil) $[\alpha]_D^{20}$ +40.0 (*c* 0.25, $CHCl_3$); FTIR (film) ν_{max} 2962, 1662, 1260, 1089, 1018, 797; ^{1}H NMR (400 MHz, $CDCl_3$) (***E***) δ 7.37-7.20 (m, 5H, H-Ar), 6.28 (d, *J*=15.1, 1H, H-7), 5.61 (dt, *J*=15.5, 7.9, 1H, H-6), 4.57 (dd, *J*=6.5, 3.1, 1H, H-9), 3.90 (m, 1H, H-1), 3.47 (m, 1H, H-1), 2.69-2.45 (m, 2H, H-5), 2.22-2.13 (m, 2H, H-3), 2.06-1.84 (m, 4H, H-2, H-11), 1.76 (m, 1H, H-12), 1.53 (s, 9H, H-17), 0.91 (d, *J*=1.8, 3H, H-13/14), 0.85 (d, *J*=3.9, 3H, H-13/14); (***Z***) δ 7.37-7.20 (m, 5H, H-Ar), 6.43 (d, *J*=9.4, 1H, H-7), 5.70 (m, 1H, H-6), 4.83 (dd, *J*=5.7, 3.6, 1H, H-9), 3.89 (m, 1H, H-1), 3.69 (m, 1H, H-1), 2.82-2.57 (m, 2H, H-5), 2.22-2.13 (m, 2H, H-3), 2.06-1.84 (m, 4H, H-2, H-11), 1.76 (m, 1H, H-12), 1.55 (s, 9H, H-17), 0.90 (d, *J*=1.7, 3H, H-13/14), 0.83 (d, *J*=3.9, 3H, H-13/14); ^{13}C NMR (100 MHz, $CDCl_3$) (***E***) δ 169.7 (C-8/10), 164.9 (C-8/10), 150.7 (C-15), 134.4 (C-18), 130.3 (CH-Ar), 130.0 (C-7), 129.3 (CH-Ar), 127.3 (CH-Ar), 123.7 (C-6), 84.6 (C-16), 69.0 (C-4), 58.7 (C-9), 44.8 (C-1), 42.3 (C-5), 41.5 (C-11), 34.7 (C-3), 28.0 (C-17), 24.8 (C-12), 23.8 (C-13/14), 22.1 (C-13/14), 19.8 (C-2); (***Z***) δ 169.1 (C-8/10), 165.2 (C-8/10), 151.0 (C-15), 133.4 (C-18), 130.2 (CH-Ar), 129.7 (C-7), 129.3 (CH-Ar), 127.0 (CH-Ar), 124.0 (C-6), 84.5 (C-16), 68.0 (C-4), 59.3 (C-9), 44.9 (C1), 41.3 (C-11), 34.9 (C-3), 29.6 (C-5), 28.5 (C-17), 24.8 (C-12), 24.0 (C-13/14), 22.1 (C-13/14), 19.8 (C-2); HRMS (ESI) calculated for $C_{25}H_{34}N_2O_4S$ $[M+Na]^+$ 481.2137 found 481.2122.

Triketopiperazine 215



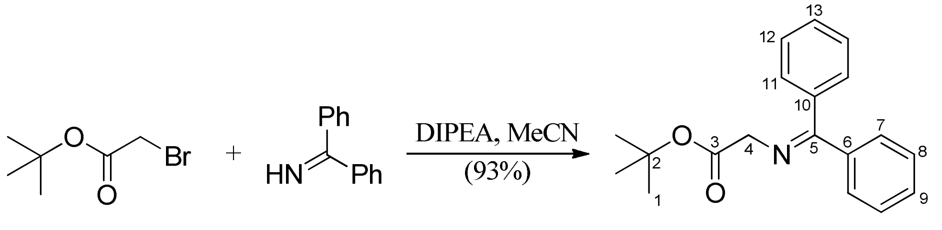
190b **215**



Diketopiperazine **109b** (241 mg, 1.17 mmol) in THF (40 mL) at −78 °C was treated with LiHMDS (1.0 M solution in THF, 1.23 ml, 1.23 mmol) and stirred for 45 minutes. Allyl bromide (202 μL, 2.34 mmol) was added and the reaction mixture allowed to warm to −40 °C and stirred for a further two hours. Ammonium chloride (7 mL) was added and the volatiles removed under reduced pressure. The residue was partitioned between water and dichloromethane and the layers separated. The aqueous phase was extracted with dichloromethane (2 x 30 mL) and the organics combined, dried ($MgSO_4$) and concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 gradually to 100% ethyl acetate) to afford triketopiperazine **215** as a white solid (26 mg, 0.12 mmol, 10%). See Chapter 3 for a discussion of the mechanism.

(**215**, white solid) m.p. 174 °C (dec.); $[\alpha]_D^{20}$ +38.6 (*c* 1.10, $CHCl_3$); FTIR (film) ν_{max} 2921, 1674, 1647, 1451, 1417, 1363, 1318, 1086; ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.94 (dt, *J*=13.0, 8.6, 1H, H-1), 3.71 (ddd, *J*=13.2, 9.6, 3.9, 1H, H-1), 3.26 (s, 3H, H-9), 2.72 (t, *J*=2.5, 2H, H-5), 2.37-2.24 (m, 2H, H-3), 2.20 (t, *J*=2.5, 1H, H-7), 2.18-1.94 (m, 2H, H-2); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.7 (C-8/10/11), 158.7 (C-8/10/11), 152.3 (C-8/10/11), 77.0 (C-6), 76.0 (C-7), 68.3 (C-4), 45.2 (C-1), 34.7 (C-3), 30.5 (C-5), 27.2 (C-9), 19.7 (C-2); HRMS (ESI) calculated for $C_{11}H_{12}N_2O_3$ $[M+Na]^+$ 243.0746 found 243.0740.

Glycine Imine 217

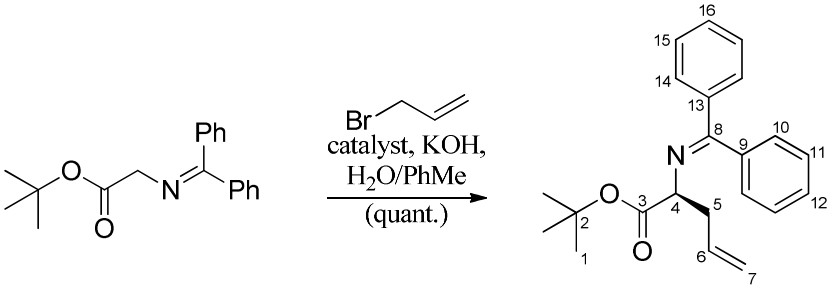


217

Benzophenone imine (9.29 g, 51.3 mmol) and *tert*-butyl bromoacetate (10.0 g, 51.3 mmol) were dissolved in acetonitrile (50 mL). The mixture was treated with di*iso*propylethylamine (8.9 mL, 51.3 mmol) then heated at reflux for 24 hours before cooling to room temperature. The mixture was diluted with dichloromethane (500 mL) and the organic layer washed with water then brine. After drying over $MgSO_4$ the volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 9/1) to afford the title compound **217** as colourless oil (14.1 g, 47.8 mmol, 93%).

(**217**, colourless oil) FTIR (film) ν_{max} 2981, 2891, 1734, 1622, 1367, 1216, 1142, 1053, 909, 848, 679, 708, 692; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.70-7.61 (m, 2H, H-Ar), 7.51-7.40 (m, 3H, H-Ar), 7.39-7.29 (m, 3H, H-Ar), 7.24-7.13 (m, 2H, H-Ar), 4.13 (s, 2H, H-3), 1.47 (s, 9H, H-1); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.7 (C-3/5), 169.9 (C-3/5), 139.5 (C-6/10), 136.3 (C-6/10), 130.5 (CH-Ar), 128.9 (CH-Ar), 128.7 (CH-Ar), 128.2 (CH-Ar), 127.9 (CH-Ar), 81.2 (C-2), 56.4 (C-4), 28.2 (C-1); HRMS (ESI) calculated for $C_{19}H_{21}NO_2$ $[M+Na]^+$ 318.1470 found 318.1465.

Allylated Glycine Imine 218

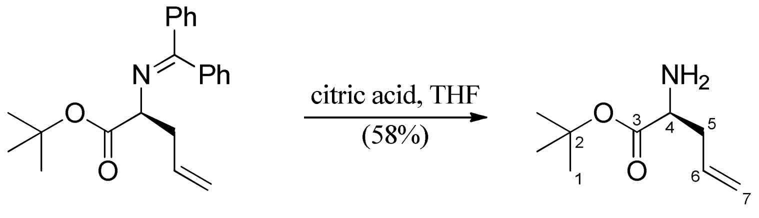


217 **218**

Protected glycine **217** (14.0 g, 47.5 mmol) and *N*-(9-anthracenylmethyl)cinchonindinium chloride (2.47 mg, 4.75 mmol) were dissolved in toluene and allyl bromide (4.52 mL, 52.3 mmol) was added. The resulting mixture was treated with an aqueous solution of potassium hydroxide (50% w/v, 107 mL, 0.95 mol) and allowed to stir for 24 hours. The layers were separated and the aqueous phase extracted with ethyl acetate. The organic layers were combined, dried ($MgSO_4$) and concentrated under reduced pressure to afford the title compound **218** as a yellow oil (16.3 g, ~47.5 mmol, quant.) that could be used without further purification.

(**218**, yellow oil) $[\alpha]_D^{22}$ −10.4 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2979, 1731, 1657, 1598, 1447, 1276, 1150; ^1H NMR (300 MHz, $CDCl_3$) δ 7.86-7.78 (m, 1H, H-Ar), 7.69-7.62 (m, 1H, H-Ar), 7.55-7.29 (m, 6H, H-Ar), 7.23-7.15 (m, 2H, H-Ar), 5.74 (m, 1H, H-6), 5.18-4.94 (m, 2H, H-7), 4.03 (dd, *J*=7.4, 5.5, 1H, H-4), 2.71-2.57 (m, 2H, H-5), 1.46 (s, 9H, H-1); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.0 (C-3/8), 170.2 (C-3/8), 139.9 (C-9/13), 136.8 (C-9/13), 134.9 (C-6), 130.2 (CH-Ar), 128.9 (CH-Ar), 128.6 (CH-Ar), 128.5 (CH-Ar), 128.4 (CH-Ar), 128.1 (CH-Ar), 117.4 (C-7), 81.1 (C-2), 66.0 (C-4), 38.3 (C-5), 28.2 (C-1); HRMS (ESI) calculated for $C_{22}H_{25}NO_2$ $[M+Na]^+$ 358.1783 found 358.1792.

Allylated Glycine 219

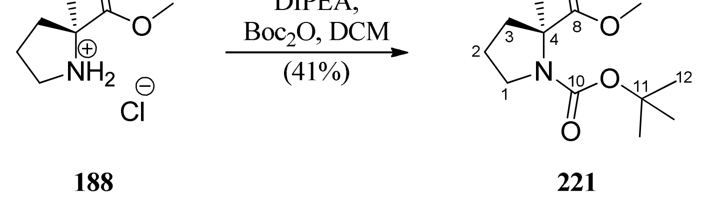


218 **219**

Protected allylglycine **218** (16.3 g ~47.5 mmol) was dissolved in tetrahydrofuran (250 mL) and treated with an aqueous solution of citric acid (15% w/v, 125 mL). The resulting mixture was allowed to stir at room temperature for 24 hours then concentrated under reduced pressure. The residue was partitioned between water (150 mL) and petroleum ether (150 mL) and the layers separated. The aqueous phase was adjusted to pH 13 with potassium carbonate then extracted with dichloromethane (3 x 150 mL). The combined dichloromethane layers were dried over $MgSO_4$ and concentrated under reduced pressure to afford the title amine **219** as a yellow oil (6.07 g, 27.5 mmol, 58%).

(**219**, yellow oil) $[\alpha]_D^{22}$ +3.6 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2979, 1727, 1648, 1535, 1369, 1154; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.76 (m, 1H, H-6), 5.25-4.96 (m, 2H, H-7), 3.42 (dd, *J*=6.9, 5.2, 1H, H-4), 2.60-2.17 (m, 2H, H-5), 1.47 (s, 9H, H-1); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.7 (C-3), 133.8 (C-6), 118.5 (C-7), 81.2 (C-2), 54.5 (C-4), 39.5 (C-5), 28.2 (C-1); HRMS (ESI) calculated for $C_9H_{17}NO_2$ $[M+Na]^+$ 194.1157 found 194.1155.

N-Boc Propargylated Proline *O*-Methyl Ester 221

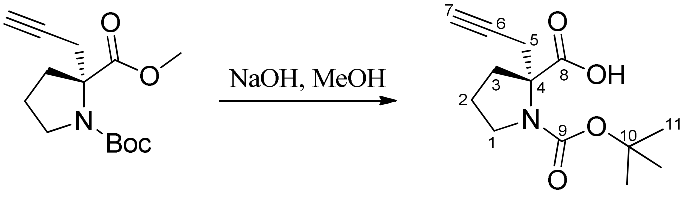


188 **221**

Substituted proline **188** (3.21 g, ~15.4 mmol) was dissolved in dichloromethane (45 mL) and treated with di*iso*propylethylamine (5.36 mL, 30.8 mmol). A solution of Boc$_2$O (8.40 g, 38.5 mmol) in dichloromethane (30 mL) was added and the resulting mixture allowed to stir at room temperature for three days. The volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 9/1 to 8/2) to afford *N*-Boc proline **221** as a colourless oil (1.69 g, 6.33 mmol, 41% from oxazolidinone **187**).

(**221**, colourless oil) $[\alpha]_D^{22}$ +65.2 (*c* 1.00, CHCl$_3$); FTIR (film) ν_{max} 2976, 1740, 1692, 1387, 1367, 1245, 1161, 1133; ^{1}H NMR (300 MHz, CDCl$_3$) (**major rotamer**) δ 3.71 (s, 3H, H-9), 3.80-3.39 (m, 2H, H-1), 3.07 (dd, *J*=17.1, 2.7, 1H, H-5), 2.83 (dd, *J*=17.1, 2.7, 1H, H-5), 2.47-1.81 (m, 4H, H-2, H-3), 1.96 (t, *J*=2.7, 1H, H-7), 1.41 (s, 9H, H-12); (**minor rotamer**) δ 3.70 (s, 3H, H-9), 3.80-3.39 (m, 2H, H-1), 3.31 (dd, *J*=17.1, 2.7, 1H, H-5), 2.89-2.76 (m, 1H, H-5), 2.47-1.81 (m, 4H, H-2, H-3), 1.94 (t, *J*=2.7, 1H, H-7), 1.44 (s, 9H, H-12); ^{13}C NMR (100 MHz, CDCl$_3$) (**major rotamer**) δ 174.2 (C-8), 153.3 (C-10), 80.5 (C-6/11), 80.3 (C-6/11), 70.7 (C-7), 66.5 (C-4), 52.5 (C-9), 48.7 (C-1), 37.9 (C-3), 28.4 (C-12), 26.4 (C-2), 23.0 (C-5); (**minor rotamer**) δ 174.1 (C-8), 154.1 (C-10), 80.9 (C-6/11), 79.9 (C-6/11), 70.2 (C-7), 67.0 (C-4), 52.5 (C-9), 48.8 (C-1), 36.5 (C-3), 28.5 (C-12), 25.0 (C-5), 23.5 (C-2); HRMS (ESI) calculated for $C_{14}H_{21}NO_4$ $[M+Na]^+$ 290.1368 found 290.1373.

N-Boc Propargylated Proline 222

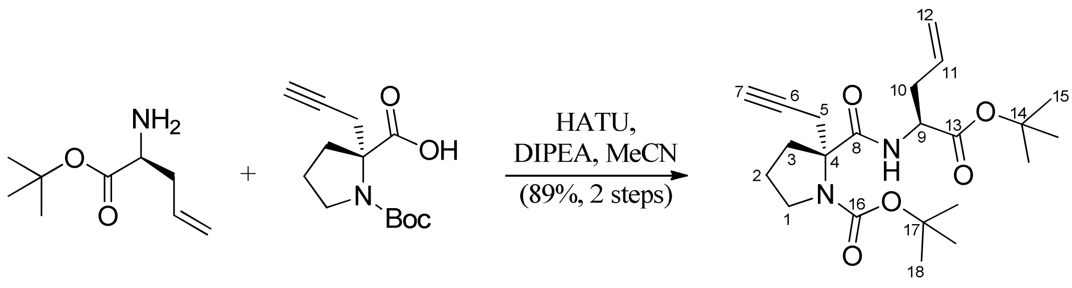


221 **222**

Protected proline **221** (1.3 g, 4.87 mmol) was dissolved in methanol (30 mL) and treated with an aqueous solution of sodium hydroxide (2 N, 30 mL). The resulting mixture was stirred at 50 °C for 3 hours then cooled to room temperature and concentrated under reduced pressure. The residue was partitioned between water (30 mL) and diethyl ether (30 mL) and the layers separated. The aqueous phase was cautiously acidified to pH 4 with an aqueous solution of citric acid (20% w/v) and extracted with diethyl ether (3 x 50 mL). The combined organic phases were dried over $MgSO_4$ and concentrated under reduced pressure to afford the title compound **222** as an off-white solid (1.26 g, ~4.87 mmol) that could be used without further purification.

(**222**, off-white solid) m.p. 93 °C – 96 °C; $[\alpha]_D^{22}$ +58.0 (*c* 1.00, MeOH); FTIR (film) ν_{max} 3323, 2977, 1741, 1631, 1414, 1370, 1226, 1149; ^{1}H NMR (300 MHz, $CDCl_3$) δ 9.80 (s, br, 1H, OH), 3.81-3.39 (m, 2H, H-1), 3.34 (dd, *J*=17.0, 2.6, 1H, H-5, major rotamer), 3.11 (dd, *J*=17.2, 2.6, 1H, H-5, minor rotamer), 2.81 (dd, *J*=17.2, 2.6, 1H, H-5, minor rotamer), 2.78 (dd, *J*=17.0, 2.6, 1H, H-5, major rotamer), 2.54-1.82 (m, 4H, H-2, H-3), 1.99 (m, 1H, H-7), 1.48 (s, 9H, H-11, major rotamer), 1.43 (s, 9H, H-11, minor rotamer); ^{13}C NMR (100 MHz, $CDCl_3$) (**major rotamer**) δ 179.4 (C-8), 156.0 (C-9), 81.6 (C-6/11), 79.6 (C-6/11), 71.0 (C-7), 68.1 (C-4), 49.5 (C-1), 35.9 (C-3), 28.6 (C-11), 25.0 (C-2), 23.1 (C-5); (**minor rotamer**); δ 176.1 (C-8), 153.4 (C-9), 81.1 (C-6/11), 80.0 (C-6/11), 71.0 (C-7), 66.4 (C-4), 48.8 (C-1), 38.0 (C-3), 28.4 (C-11), 26.1 (C-5), 23.0 (C-2); HRMS (ESI) calculated for $C_{13}H_{18}NO_4$ $[M-H]^-$ 252.1236 found 252.1245.

Proline-Allylglycine Dipeptide 223



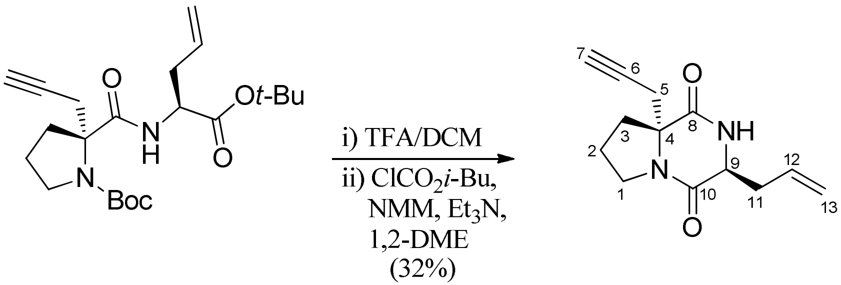
219 **222** **223**

General procedure B was followed with allylglycine **219** (1.05 g, 4.74 mmol), *N*-Boc proline **222** (1.2 g, 4.74 mmol), HATU (1.98 g, 5.21 mmol) and di*iso*propylethylamine (1.73 mL, 9.95 mmol) to afford the title compound **223** as a near-colourless oil (1.71 g, 4.21 mmol, 89%).

(**223**, colourless oil) $[\alpha]_D^{22}$ +9.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2978, 1673, 1513, 1366, 1251, 1154, 1002; ^1H NMR (300 MHz, $CDCl_3$) δ 7.46 (d, *J*=6.5, 1H, N-H, minor rotamer), 6.74 (d, *J*=7.1, 1H, N-H, major rotamer), 5.67 (m, 1H, H-11), 5.17-4.98 (m, 2H, H-12), 4.46 (m, 1H, H-9), 3.79-3.33 (m, 2H, H-1), 3.43 (dd, *J*=17.1, 2.7, 1H, H-5, minor rotamer), 3.13 (dd, *J*=17.3, 2.5, 1H, H-5, major rotamer), 2.88 (m, 1H, H-5, major rotamer), 2.81 (m, 1H, H-5, minor rotamer), 2.67-2.15 (m, 4H, H-3, H-10), 1.96 (m, 1H, H-7), 2.00-1.63 (m, 2H, H-2), 1.47 (s, 9H, H-15/18, minor rotamer), 1.46 (s, 9H, H-15/18, major rotamer), 1.45 (s, 9H, H-15/18, minor rotamer), 1.44 (s, 9H, H-15/18, major rotamer); ^{13}C NMR (100 MHz, $CDCl_3$) (**major rotamer**) δ 173.3 (C-8), 170.4 (C-13), 153.7 (C-17), 132.5 (C-11), 118.9 (C-12), 82.4 (C-6), 81.1 (C-14/17), 80.4 (C-14/17), 71.1 (C-7), 67.8 (C-4), 51.9 (C-9), 49.4 (C-1), 38.8 (C-3), 36.7 (C-10), 28.3 (C-15/18), 28.1 (C-15/18), 25.9 (C-2), 22.2 (C-5); (**minor rotamer**) δ 172.9 (C-8), 170.6 (C-13), 154.5 (C-17), 132.7 (C-11), 118.7 (C-12), 82.1 (C-6), 80.8 (C-14/17), 80.4 (C-14/17), 70.5 (C-7), 68.6 (C-4), 52.6 (C-9), 49.7 (C-1), 37.1 (C-

3), 36.1 (C-10), 28.5 (C-15/18), 28.1 (C-15/18), 24.8 (C-5), 22.7 (C-2); HRMS (ESI) calculated for $C_{22}H_{34}N_2O_5$ $[M+Na]^+$ 429.2365 found 429.2372.

Proline-Allylglycine DKP 225



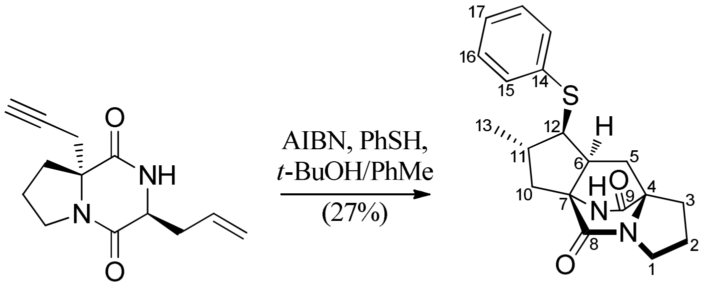
223 **225**

Dipeptide **223** (893 mg, 2.20 mmol) was dissolved in dichloromethane (14 mL) and treated with trifluoroacetic acid (21 mL) followed by water (250 μL). After stirring for 4 hours the volatile organics were removed under reduced pressure to afford a sticky oil (1.16 g). A portion of this (129 mg, ~0.35 mmol) was dissolved in 1,2-dimethoxyethane (2 mL) at 0 °C and treated sequentially with *N*-methylmorpholine (77 μL, 0.70 mmol) then *iso*butyl chloroformate (92 μL, 0.70 mmol). After stirring for 3 hours at 0 °C the precipitate was removed by filtration and washed with 1,2-dimethoxyethane (2 x 2 mL). The filtrate was combined, cooled to 0 °C and treated with triethylamine. After stirring at room temperature for 20 hours the volatile organics were removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 to 100% ethyl acetate) to afford the desired DKP **225** as a colourless oil (26 mg, 0.11 mmol, 32%).

(**225**, colourless oil) $[\alpha]_D^{22}$ −15.0 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 3275, 3236, 1669, 1638, 1435, 1306, 1142, 990; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.91 (s, 1H, N-H), 5.76 (m, 1H, H-12), 5.32-5.19 (m, 2H, H-13), 4.37 (dd, *J*=9.7, 3.3, 1H, H-9), 3.87 (dt, *J*=12.4, 8.3, 1H, H-1), 3.56 (m, 1H, H-1), 3.01 (m, 1H, H-11), 2.79 (dd, *J*=17.0, 2.7, 1H, H-5), 2.63 (dd, *J*=17.0, 2.7, 1H, H-5), 2.43-1.89 (m, 5H, H-2, H-3, H-11), 2.07 (t, *J*=2.7, 1H, H-7); ^{13}C NMR (100 MHz,

$CDCl_3$) δ 170.3 (C-8/10), 165.3 (C-8/10), 133.3 (C-12), 120.6 (C-13), 78.6 (C-6), 72.4 (C-7), 67.3 (C-4), 54.1 (C-9), 45.3 (C-1), 36.4 (C-11), 35.0 (C-3), 27.9 (C-5), 20.4 (C-2); HRMS (ESI) calculated for $C_{13}H_{16}N_2O_2$ $[M+Na]^+$ 255.1109 found 255.1105.

Tetracyclic Bridged DKP 227



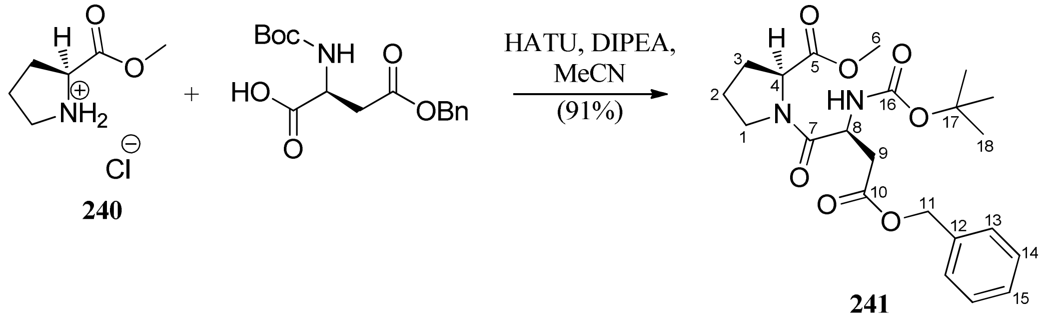
225 **227**

General procedure A was followed with DKP **225** (26 mg, 0.11 mmol), AIBN (0.2 M solution in toluene, 1.1 mL, 0.22 mmol), thiophenol (23 μL, 0.22 mmol) and *tert*-butanol (25 mL) to afford the desired bridged compound **227** as a colourless oil (10 mg, 0.03 mmol, 27%).

(**227**, colourless oil) $[\alpha]_D^{20}$ −4.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2963, 2928, 1685, 1439, 1402, 1262, 1072; ^1H NMR (400 MHz, $CDCl_3$) δ 7.38-7.14 (m, 5H, H-Ar), 6.85 (s, 1H, N-H), 3.57-3.43 (m, 2H, H-1), 3.26 (dd, *J*=13.1, 8.7, 1H, H-11), 2.98 (m, 1H, H-6), 2.90 (m, 1H, H-10), 2.72 (m, 1H, H-3), 2.45 (ddd, *J*=13.1, 9.7, 6.1, 1H, H-12), 2.17 (dd, *J*=13.5, 9.7, 1H, H-5), 2.06-1.93 (m, 2H, H-2), 1.77 (dt, *J*=13.5, 7.6, 1H, H-3), 1.67 (dd, *J*=13.4, 6.1, 1H, H-5), 1.30 (m, 1H, H-10), 1.17 (d, *J*=7.1, 3H, H-13); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.6 (C-9), 168.0 (C-8), 136.5 (C-14), 129.2 (CH-Ar), 128.7 (CH-Ar), 126.1 (CH-Ar), 68.8 (C-4), 66.1 (C-7), 52.8 (C-11), 51.6 (C-12), 44.0 (C-1), 35.4 (C-6), 35.3 (C-10), 34.7 (C-5), 29.5 (C-3), 24.8 (C-2), 19.5 (C-13); HRMS (ESI) calculated for $C_{19}H_{22}N_2O_2S$ $[M+Na]^+$ 365.1300 found 365.1308.

5.3 Experimental for Chapter 4

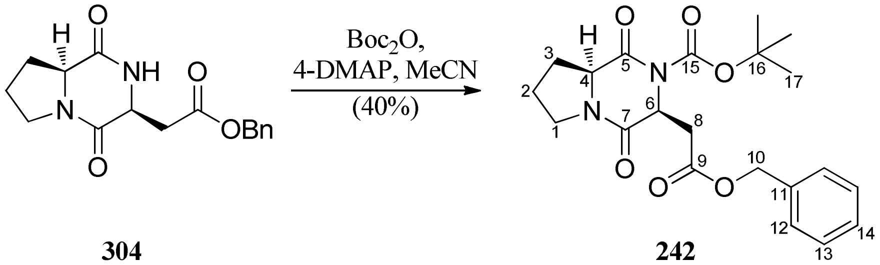
Proline-Aspartic Acid *O*-Benzyl Ester Dipeptide 241



General procedure B was followed using L-proline *O*-methyl ester (2.0 g, 12.1 mmol), *N*-Boc aspartic acid β-*O*-benzyl ester (3.9 g, 12.1 mmol), HATU (5.1 g, 13.3 mmol) and di*iso*propylethylamine (4.4 mL, 25.4 mmol) to afford dipeptide **241** (5.20 g, 11.0 mmol, 91%) as a colourless oil.

(**241**, colourless oil) $[\alpha]_D^{21}$ −50.8 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2963, 1738, 1646, 1436, 1257, 1161, 1005; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.42-7.28 (m, 5H, H-Ar), 5.41 (d, *J*=9.3, 1H, N-H), 5.15 (d, *J*=3.7, 2H, H-11), 4.89 (dt, *J*=9.3, 6.4, 1H, H-8), 4.50 (dd, *J*=8.2, 3.3, 1H, H-4), 3.69 (s, 3H, H-6), 3.82-3.57 (m, 2H, H-1), 2.82 (dd, *J*=15.9, 5.7, 1H, H-9), 2.68 (dd, *J*=15.9, 6.9, 1H, H-9), 2.30-1.77 (m, 4H, H-2, H-3), 1.42 (s, 9H, H-18); ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.3 (C=O), 170.4 (C=O), 169.7 (C=O), 155.2 (C-16), 135.8 (C-12), 128.6 (CH-Ar), 128.4 (CH-Ar), 128.4 (CH-Ar), 80.2 (C-17), 66.9 (C-11), 59.1 (C-4), 52.3 (C-6), 49.1 (C-8), 47.1 (C-1), 37.6 (C-9), 29.1 (C-3), 28.4 (C-18), 25.0 (C-2); HRMS (ESI) calculated for $C_{22}H_{30}N_2O_7$ $[M+Na]^+$ 457.1951 found 457.1961.

N-Boc Proline-Aspartic Acid *O*-Benzyl Ester DKP 242

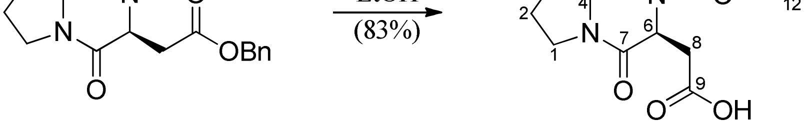


304 **242**

General procedure D was followed with DKP **304** (2.50 g, 8.27 mmol), Boc_2O (3.07 g, 14.1 mmol), 4-DMAP (101 mg, 0.83 mmol) and acetonitrile (75 mL) to afford *N*-Boc DKP **242** as a colourless oil (1.84 g, 4.57 mmol, 55%).

(**242**, colourless oil) $[\alpha]_D^{21}$ +15.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2979, 1733, 1713, 1668, 1368, 1248, 1148; ^1H NMR (300 MHz, $CDCl_3$) δ 7.44-7.30 (m, 5H, H-Ar), 5.78 (dd, *J*=9.1, 5.1, 1H, H-6), 5.15 (app. s, 2H, H-10), 3.88 (m, 1H, H-4), 3.82-3.68 (m, 2H, H-1), 3.28 (dd, *J*=16.1, 9.1, 1H, H-8), 2.68 (dd, *J*= 16.1, 5.1, 1H, H-8), 2.62-2.47 (m, 2H, H-3), 2.18-1.89 (m, 2H, H-2), 1.55 (s, 9H, H-17); ^{13}C NMR (100 MHz, $CDCl_3$) δ 194.4 (C=O), 193.5 (C=O), 184.8 (C=O), 179.9 (C=O), 133.6 (C-11), 128.7 (CH-Ar), 128.3 (CH-Ar), 124.9 (CH-Ar), 85.3 (C-16), 66.8 (C-10), 53.4 (C-4), 46.1 (C-1), 39.6 (C-6), 34.3 (C-8), 33.4 (C-3), 28.1 (C-17), 17.8 (C-2); LRMS (LC-MS/ESI) calculated for $C_{21}H_{26}N_2O_6$ $[M+H]^+$ 402.2 found 402.2.

N-Boc Proline-Aspartic Acid DKP 243

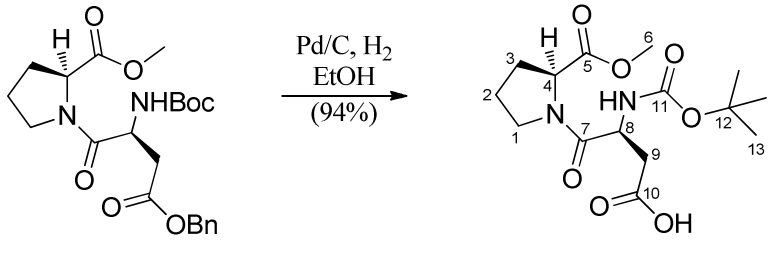


242 **243**

A 50 mL round bottomed flask was charged with benzyl ester **242** (1.56 g, 4.0 mmol), palladium on carbon (10 wt. %, 43 mg, 0.4 mmol) and ethanol (20 mL). The flask was flushed with nitrogen then hydrogen and stirred under this atmosphere for 20 hours. The catalyst was removed by filtration through Celite® and the filtrate concentrated under reduced pressure to afford the title compound **243** as a colourless oil (1.04 g, 3.33 mmol, 83%) that was used without further purification.

(**243**, colourless oil) $[\alpha]_D^{21}$ −2.8 (*c* 1.00, MeOH); FTIR (film) ν_{max} 3329, 2977, 1708, 1648, 1399, 1253, 1160, 1050; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.06 (br. s, 1H, O-H), 5.70 (dd, *J*=9.2, 4.7, 1H, H-6), 4.26 (dd, *J*=10.2, 6.2, 1H, H-4), 4.06-3.51 (m, 2H, H-1), 3.52-3.00 (m, 2H, H-8), 2.45-1.76 (m, 4H, H-2, H-3), 1.53 (s, 9H, H-12); ^{13}C NMR (100 MHz, $CDCl_3$) δ 180.7 (C=O), 174.9 (C=O), 174.7 (C=O), 155.8 (C-10), 69.6 (C-11), 49.9 (C-4), 46.1 (C-1), 36.8 (C-8), 31.1 (C-6), 29.1 (C-3), 28.2 (C-12), 24.9 (C-2); HRMS (ESI) calculated for $C_{14}H_{19}N_2O_6$ $[M-H]^-$ 311.1243 found 311.1238.

Proline-Aspartic Acid Dipeptide 253

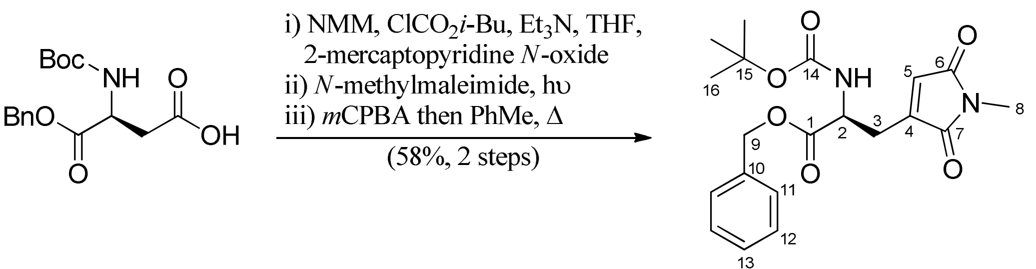


241 **253**

A 100 mL round bottomed flask was charged with dipeptide **241** (5.0 g, 10.6 mmol), palladium on carbon (10 wt. %, 113 mg, 0.11 mmol) and ethanol (50 mL). The flask was flushed with nitrogen followed by hydrogen and stirred under this atmosphere for 20 hours. The catalyst was removed by filtration through Celite® and the filtrate concentrated under reduced pressure to afford the desired compound **253** as a colourless oil (3.42 g, 9.93 mmol, 94%).

(**253**, colourless oil) $[\alpha]_D^{22}$ −60.8 (*c* 1.20, $CHCl_3$); FTIR (film) ν_{max} 2979, 1707, 1515, 1367, 1158, 1049; ^1H NMR (300 MHz, $CDCl_3$) δ 7.88 (s, br, 1H, O-H), 5.73 (d, *J*=9.1, 1H, N-H), 4.84 (m, 1H, H-8), 4.49 (m, 1H, H-4), 3.83-3.64 (m, 2H, H-1), 3.69 (s, 3H, H-6), 3.00-2.50 (m, 2H, H-9), 2.33-1.83 (m, 4H, H-2, H-3), 1.42 (s, 9H, H-13); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.0 (C=O), 172.4 (C=O), 170.4 (C=O), 155.5 (C-11), 80.4 (C-12), 59.3 (C-4), 52.5 (C-6), 49.0 (C-8), 47.3 (C-1), 37.2 (C-9), 29.1 (C-3), 28.4 (C-13), 24.9 (C-2); HRMS (ESI) calculated for $C_{15}H_{24}N_2O_7$ $[M+Na]^+$ 367.1481 found 367.1475.

N-Methylmaleimide Substituted Aspartic Acid *O*-Benzyl Ester 257

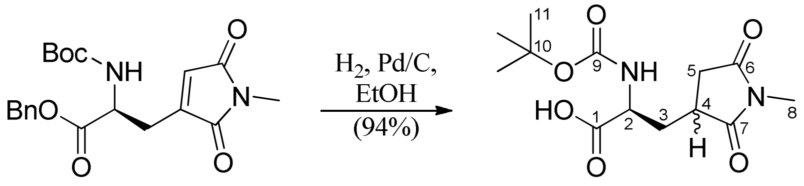


255 **257**

N-Boc Aspartic acid α-*O*-benzyl ester **255** (5.55 g, 17.2 mmol) was dissolved in THF (150 mL), cooled to −20 °C, and treated sequentially with *N*-methyl morpholine (1.89 mL, 17.2 mmol) then *iso*butyl chloroformate (2.26 mL, 17.2 mmol). After 5 minutes a solution of 2-mercaptopyridine *N*-oxide (2.62 g, 20.6 mmol) and triethylamine (3.1 mL, 22.3 mmol) in THF (50 mL) was added. The resulting mixture was stirred with the exclusion of light for 4 hours. The precipitate was removed by filtration and the filtrate irradiated in the presence of *N*-methylmaleimide (9.55 g, 86.0 mmol) for 1 hour with the temperature maintained below 10 °C. After removal of the volatile organics under reduced pressure, the residue was partitioned between dichloromethane (200 mL) and $NaHCO_3$ (150 mL, saturated aqueous solution). The layers were separated and the organic phase washed with HCl (0.5 N, 150 mL) then brine (150 mL) and dried over $MgSO_4$. The mixture was concentrated under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 to 1/1) to afford a white foam (6.1 g, ~12.2 mmol, 71%). Signals in the ^{1}H NMR spectrum (300 MHz, $CDCl_3$) at 8.24-8.14, 7.56-7.44, 7.23-7.12 and 7.04-6.89 ppm and mass spectrometry (HRMS (ESI) calculated for $C_{25}H_{29}N_3O_6S$ $[M+Na]^+$ 522.1675 found 522.1680) indicated that the 2-thiopyridyl moiety was present. The white foam was dissolved in chloroform (100 mL), cooled to 0 °C and treated with a solution of *m*CPBA (70-75% in H_2O, 2.8 g, ~12.2 mmol) in chloroform (100 mL). Further small portions of *m*CPBA were added until TLC indicated that all of the starting material had been consumed. The mixture was diluted with dichloromethane (200 mL) and washed with $NaHCO_3$ (saturated aqueous solution, 300 mL), HCl (0.5 N, 300 mL) and brine (300 mL). After drying over $MgSO_4$, the mixture was concentrated under reduced pressure and the resulting white foam dissolved in toluene (200 mL) then heated at reflux for 1.5 hours. The solvent was removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 to 1/1) to afford the desired compound **257** as a colourless oil (3.84 g, 9.88 mmol, 81%).

(**257**, colourless oil) $[\alpha]_D^{20}$ −4.8 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3379, 2989, 1752, 1700, 1682, 1522, 1213, 1156; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.47-7.17 (m, 5H, H-Ar), 6.30 (s, 1H, H-5), 5.36 (d, *J*=7.7, 1H, N-H), 5.26-5.05 (m, 2H, H-9), 4.62 (dd, *J*=12.9, 7.3, 1H, H-2), 2.93 (s, 3H, H-8), 3.09-2.75 (m, 2H, H-3), 1.41 (s, 9H, H-16); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.3 (C-1/6/7), 170.8 (C-1/6/7), 170.4 (C-1/6/7), 155.2 (C-14), 144.5 (C-4), 135.0 (C-10), 129.2 (C-11/12/13), 128.8 (C-11/12/13), 128.7 (C-11/12/13), 125.7 (C-5), 80.5 (C-15), 67.8 (C-9), 52.1 (C-2), 28.8 (C-3), 28.3 (C-16), 23.9 (C-8); HRMS (ESI) calculated for $C_{20}H_{24}N_2O_6$ $[M+Na]^+$ 411.1532 found 411.1533.

N-Methylsuccinimide Substituted *N*-Boc Aspartic Acid 258

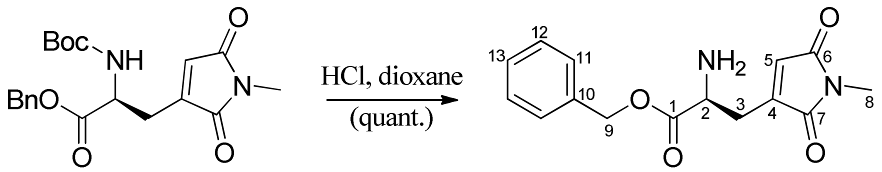


257 **258**

A 100 mL round bottomed flask was charged with benzyl ester **257** (1.18 g, 3.04 mmol), palladium on carbon (10 wt. %, 32 mg, 0.30 mmol) and ethanol (25 mL). The flask was flushed with nitrogen followed by hydrogen and stirred under this atmosphere for 20 hours. The catalyst was removed by filtration through Celite® and the filtrate concentrated under reduced pressure to afford the desired compound **258** as a colourless oil (859 mg, 2.86 mmol, 94%) which was used without further purification.

(**258**, colourless oil) $[\alpha]_D^{20}$ +5.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3322, 2977, 1730, 1681, 1276, 1159, 1117; ^1H NMR (300 MHz, CD_3OD) δ 7.52 (s, br, 1H, O-H), 5.36 (d, *J*=7.8, 1H, N-H), 4.46 (m, 1H, H-2), 3.14-2.86 (m, 2H, H-3), 3.00 (s, 3H, H-8), 2.64-2.23 (m, 2H, H-5), 2.02 (m, 1H, H-4), 1.45 (s, 9H, H-11); ^{13}C NMR (100 MHz, CD_3OD) δ 181.7 (C=O), 178.7 (C=O), 175.3 (C=O), 158.1 (C-9), 80.7 (C-10), 52.8 (C-2), 38.6 (C-4), 35.2 (C-3), 33.9 (C-5), 28.7 (C-11), 25.0 (C-8); HRMS (ESI) calculated for $C_{13}H_{20}N_2O_6$ $[M+Na]^+$ 323.1219 found 323.1205.

N-Methylmaleimide Substituted Aspartic Acid 259

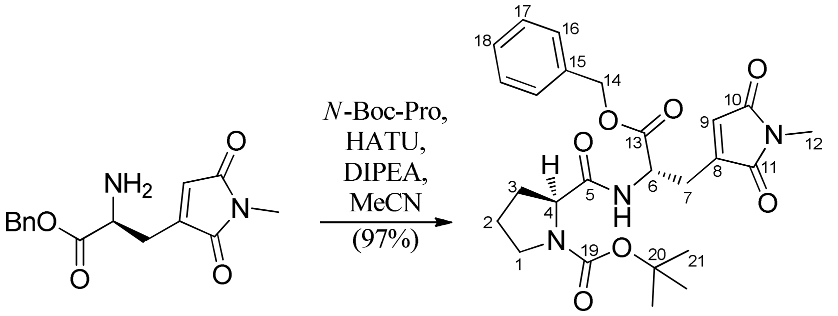


257 **259**

O-Benzyl ester **257** (693 mg, 1.78 mmol) was dissolved in hydrogen chloride in dioxane (4M, 7.0 mL) and allowed to stir at room temperature for 20 hours. The volatile organics were removed under reduced pressure to afford the title compound **259** as a yellow oil (514 mg, 1.78 mmol, quant.) which was used without further purification.

(**259**, yellow oil) $[\alpha]_D^{20}$ +5.0 (*c* 0.50, MeOH); FTIR (film) ν_{max} 2925, 1699, 1447, 1391, 1219, 1119, 1011; ^{1}H NMR (300 MHz, CD_3OD) δ 7.54-7.09 (m, 5H, H-Ar), 6.53 (s, 1H, H-5), 5.26 (app. q, *J*=11.9, 2H, H-9), 4.52 (t, *J*=6.6, 1H, H-2), 3.21-2.90 (m, 2H, H-3), 2.86 (s, 3H, H-8); ^{13}C NMR (100 MHz, CD_3OD) δ 172.4 (C=O), 171.3 (C=O), 169.1 (C=O), 143.4 (C-4), 136.0 (C-10), 131.8 (C-5), 130.0 (CH-Ar), 129.8 (CH-Ar), 128.0 (CH-Ar), 69.6 (C-9), 52.1 (C-2), 27.4 (C-3), 23.9 (C-8); HRMS (ESI) calculated for $C_{15}H_{17}N_2O_4$ $[M+H]^+$ 289.1188 found 289.1181.

N-Boc-Proline-*N*-Methylmaleimide Substituted Aspartic Acid Dipeptide 261

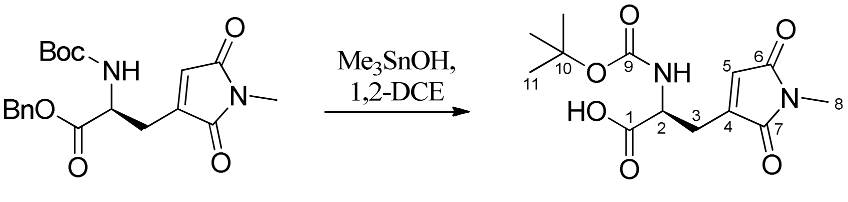


259 **261**

General procedure B was followed with maleimide **259** (514 mg, 1.78 mmol), *N*-Boc proline (384 mg, 1.78 mmol), HATU (746 mg, 1.96 mmol) and di*iso*propylethylamine (0.65 mL, 3.74 mmol) to afford the title compound **261** as a yellow oil (841 mg, 1.73 mmol, 97%).

(**261**, yellow oil) $[\alpha]_D^{21}$ −25.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2969, 1738, 1714, 1396, 1261, 1168, 1090, 1020; ^{1}H NMR (300 MHz, $CDCl_3$) δ (rotamers) 7.38-7.32 (m, 5H, H-Ar), 6.31 (br. s, 1H, H-N), 5.18 (d, *J*=3.0, 2H, H-14), 4.85 (dd, *J*=12.7, 6.9, 1H, H-9), 4.37 (m, 1H, H-6), 4.26 (m, 1H, H-4), 3.65-3.29 (m, 2H, H-1), 2.94 (s, 3H, H-12), 3.21-2.76 (m, 2H, H-7), 2.19-1.73 (m, 4H, H-2, H-3), 1.45 (br. s, 9H, H-21); ^{13}C NMR (100 MHz, CDCl$_3$) δ 177.9 (C=O), 174.9 (C=O), 171.4 (C=O), 170.3 (C=O), 154.0 (C-19), 144.3 (C-8), 135.0 (C-15), 129.3 (C-9), 128.9 (CH-Ar), 128.8 (CH-Ar), 128.7 (CH-Ar), 81.4 (C-20), 67.8 (C-14), 59.2 (C-4), 50.8 (C-6), 47.1 (C-7), 46.4 (C-1), 31.0 (C-3), 28.4 (C-21), 24.4 (C-2), 23.9 (C-12); HRMS (ESI) calculated for $C_{25}H_{31}N_3O_7$ [M+Na]$^+$ 508.2060 found 508.2069.

N-Methylmaleimide Substituted Aspartic Acid 267

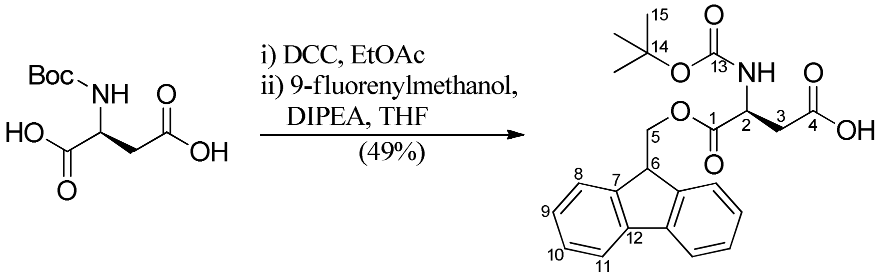


257 **267**

A pressure tube was charged with protected aspartic acid **257** (1.50 g, 3.86 mmol), trimethyltin hydroxide (1.0 g, 5.79 mmol) and 1,2-dichloroethane (15 mL). The tube was sealed and heated at 80 °C for 20 hours. After cooling to room temperature the volatile organics were removed under reduced pressure and the residue partitioned between ethyl acetate and HCl (5% v/v aqueous solution). The layers were separated and the organic phase washed with further portions of HCl until ^{1}H NMR indicated no trimethyltin species were present. After drying over MgSO$_4$ the mixture was concentrated under reduced pressure to afford **267** as a colourless oil (1.34 g, ~3.86 mmol) that could be used without further purification. An analytically pure sample was obtained by flash column chromatography (eluent petroleum ether/ethyl acetate, 7/3 to 1/1).

(**267**, colourless oil) $[\alpha]_D^{21}$ −19.2 (*c* 0.50, CHCl$_3$); FTIR (film) ν_{max} 3362, 2980, 1696, 1515, 1445, 1390, 1158; ^{1}H NMR (300 MHz, CDCl$_3$) δ 6.49 (s, 1H, H-5), 5.69 (s, br, 1H, O-H), 5.38 (d, *J*=7.4, 1H, N-H), 4.62 (dd, *J*=12.4, 7.4, 1H, H-2), 3.02 (s, 3H, H-8), 3.22-2.77 (m, 2H, H-3), 1.44 (s, 9H, H-11); ^{13}C NMR (100 MHz, CDCl$_3$) δ 173.9 (C-1/6/7), 173.8 (C-1/6/7), 170.4 (C-1/6/7), 155.5 (C-9), 144.4 (C-4), 129.4 (C-5), 80.9 (C-10), 51.9 (C-2), 28.4 (C-3), 28.2 (C-11), 23.9 (C-8); HRMS (ESI) calculated for $C_{13}H_{18}N_2O_6$ $[M+Na]^+$ 321.1063 found 321.1058.

N-Boc Aspartic Acid *O*-Fluorenylmethyl Ester 270

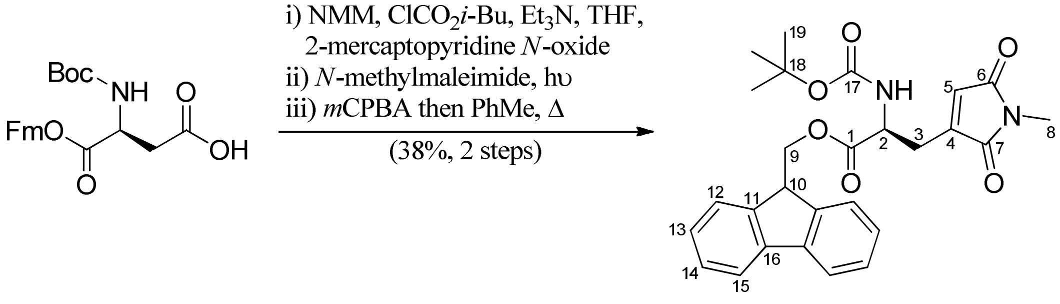


268 **270**

Dicyclohexylcarbodiimide (2.13 g, 10.3 mmol) was added in portions to a solution of *N*-Boc aspartic acid **268** (2.29 g, 9.82 mmol) in ethyl acetate (20 mL) at 0 °C. After stirring for one hour the mixture was warmed to room temperature and stirred for a further one hour when TLC showed consumption of the starting material. The precipitate was removed by filtration and the filtrate concentrated under reduced pressure. The residue was dissolved in THF (20 mL) and treated with 9-fluorenylmethanol (2.12 g, 10.8 mmol) followed by di*iso*propylethylamine (1.41 mL, 11.8 mmol). After stirring at room temperature for 20 hours, the volatile organics were removed under reduced pressure. The residue was dissolved in ethyl acetate (50 mL) and washed with $KHSO_4$ (0.5 M, 2 x 50 mL) followed by brine (2 x 50 mL). After drying over $MgSO_4$ the mixture was concentrated under reduced pressure to give an off-white solid which was purified further by recrystallisation from ethyl acetate to afford the title compound **270** as a white solid (2.0 g, 4.86 mmol, 49%).

(**270**, white solid) m.p. 159 °C – 162 °C; $[\alpha]_D^{20}$ −12.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2977, 1739, 1697, 1500, 1446, 1238, 1207, 1157; ^1H NMR (300 MHz, $CDCl_3$) δ 7.82-7.71 (m, 2H, H-Ar), 7.62-7.52 (m, 2H, H-Ar), 7.45-7.36 (m, 2H, H-Ar), 7.35-7.28 (m, 2H, H-Ar), 6.44 (s, br, 1H, O-H), 5.51 (d, *J*=8.6, 1H, N-H), 4.65 (dt, *J*=8.6, 4.2, 1H, H-2), 4.56-4.40 (m, 2H, H-5), 4.22 (t, *J*=6.6, 1H, H-6), 2.99 (dd, *J*=17.6, 4.2, 1H, H-3), 2.83 (dd, *J*=17.6, 4.2, 1H, H-3), 1.47 (s, 9H, H-15); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.3 (C-1/4), 170.9 (C-1/4), 160.8 (C-13), 143.6 (C-7/12), 141.4 (C-7/12), 128.0 (CH-Ar), 127.3 (CH-Ar), 125.1 (CH-Ar), 120.2 (CH-Ar), 77.7 (C-14), 67.8 (C-5), 49.9 (C-2), 46.8 (C-6), 36.4 (C-3), 28.5 (C-15); HRMS (ESI) calculated for $C_{23}H_{25}NO_6$ $[M+Na]^+$ 434.1580 found 434.1573.

N-Methylmaleimide Substituted *N*-Boc Aspartic Acid Fluorenylmethyl Ester 272



270 **272**

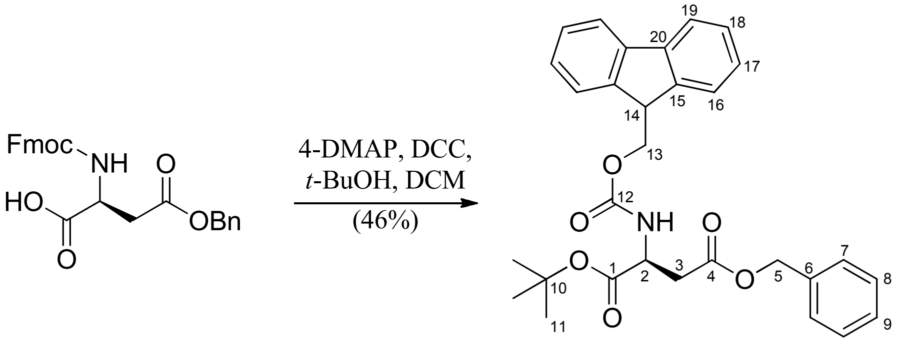
Carboxylic acid **270** (1.0 g, 2.43 mmol) was dissolved in THF (20 mL) and treated with *N*-methyl morpholine (270 μL, 2.43 mmol) and isobutyl chloroformate (320 μL, 2.43 mmol) at −20 °C. After 5 minutes a solution of 2-mercaptopyridine *N*-oxide (370 mg, 2.92 mmol) and triethylamine (440 μL, 3.16 mmol) in THF (10 mL) was added. The resulting mixture was stirred in the dark for 4 hours and the resulting white solid removed by filtration. The filtrate was irradiated in the presence of *N*-methylmaleimide (1.36 g, 12.2 mmol) for 1 hour with the temperature maintained below 10 °C. The volatile organics were removed under reduced pressure and the residue partitioned between dichloromethane (75 mL) and $NaHCO_3$ (sat. aq.

soln., 75 mL). The layers were separated and the organic phase washed with water (75 mL), HCl (0.5 N, 75 mL), water (75 mL) and brine (75 mL) before drying over $MgSO_4$. After removal of the solvent under reduced pressure diethyl ether (75 mL) was added and the resulting white preciptate removed by filtration. The filtrate was concentrated under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 3/2 gradually to 1/1) to afford a white foam (544 mg, 0.93 mmol, 38%). Signals in the ^{1}H NMR spectrum (300 MHz, $CDCl_3$) at 8.23-8.16, 7.60-7.49, 7.20-7.12 and 7.02-6.92 ppm and mass spectrometry (HRMS (ESI) calculated for $C_{32}H_{33}N_3O_6S$ $[M+Na]^+$ 610.1988 found 610.1981) indicated that the 2-thiopyridyl moiety was present. A portion of the white foam (349 mg, 0.59 mmol) was dissolved in chloroform (6 mL), cooled to 0 °C and treated with a solution of *m*CPBA (70-75% in H_2O, 140 mg, ~0.59 mmol) in chloroform (6 mL). Further small portions of *m*CPBA were added until TLC indicated that all of the starting material had been consumed. The mixture was diluted with dichloromethane (20 mL) and washed with $NaHCO_3$ (saturated aqueous solution, 20 mL), HCl (0.5 N, 20 mL) and brine (20 mL). After drying over $MgSO_4$, the mixture was concentrated under reduced pressure and the resulting white foam dissolved in toluene (15 mL) then heated at reflux for 1.5 hours. The solvent was removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 8/2 to 1/1) to afford the desired compound **272** as a colourless oil (233 mg, 0.49 mmol, 83%).

(**272**, colourless oil) $[\alpha]_D^{22}$ −9.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2979, 1700, 1505, 1448, 1389, 1158, 1006; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.76 (dd, *J*=7.4, 2.9, 2H, H-Ar), 7.62-7.55 (m, 2H, H-Ar), 7.46-7.37 (m, 2H, H-Ar), 7.36-7.28 (m, 2H, H-Ar), 6.30 (s, 1H, H-5), 5.28 (d, *J*=8.5, 1H, N-H), 4.71-4.41 (m, 3H, H-2, H-9), 4.22 (t, *J*=6.4, 1H, H-10), 2.97 (s, 3H, H-8), 2.90 (dd, *J*=15.3, 4.8, 1H, H-3), 2.66 (dd, *J*=15.3, 7.5, 1H, H-3), 1.42 (s, 9H, H-19); ^{13}C NMR

(100 MHz, $CDCl_3$) δ 171.3 (C=O), 170.9 (C=O), 170.4 (C=O), 155.2 (C-17), 144.5 (C-4), 143.4 (C-11/16), 141.4 (C-11/16), 129.0 (C-5), 128.1 (CH-Ar), 127.3 (CH-Ar), 125.0 (CH-Ar), 120.2 (CH-Ar), 80.5 (C-18), 67.4 (C-9), 52.0 (C-2), 46.9 (C-10), 28.8 (C-3), 28.3 (C-19), 23.9 (C-8); HRMS (ESI) calculated for $C_{27}H_{28}N_2O_6$ $[M+Na]^+$ 499.1845 found 499.1868.

N-Fmoc Aspartic Acid Diester 274

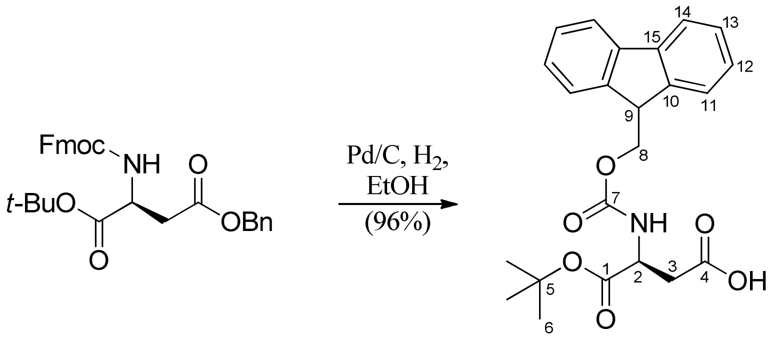


273 **274**

Fmoc-Aspartic acid β-*O*-benzyl ester **273** (5.38 g, 12.1 mmol) was dissolved in dichloromethane (10 mL) and treated with *tert*-butanol (1.32 mL, 13.9 mmol) followed by dicyclohexylcarbodiimide (1 M in DCM, 14 mL, 14.0 mmol) then 4-dimethylaminopyridine (148 mg, 1.21 mmol). After stirring at room temperature for 20 hours, the white precipitate was removed by filtration and the filtrate concentrated under reduced pressure. The residue was purified by column chromatography (eluent 100% dichloromethane) to afford the desired compound **274** as a colourless oil (2.82 g, 5.62 mmol, 46%).

(**274**, colourless oil) $[\alpha]_D^{21}$ +5.6 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2979, 1723, 1503, 1450, 1218, 1150, 1044; ^1H NMR (300 MHz, $CDCl_3$) δ 7.78 (d, *J*=7.5, 2H, H-Ar), 7.61 (d, *J*=7.5, 2H, H-Ar), 7.52-7.27 (m, 9H, H-Ar), 5.81 (d, *J*=8.2, 1H, N-H), 5.17 (q, *J*=12.2, 2H, H-5), 4.58 (m, 1H, H-2), 4.50-4.14 (m, 3H, H-13, H-14), 3.06 (dd, *J*=17.0, 4.6, 1H, H-3), 2.91 (dd, *J*=17.0, 4.6, 1H, H-3), 1.45 (s, 9H, H-11); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.8 (C-1/4), 170.0 (C-1/4), 156.1 (C-12), 144.0 (C-15/20), 143.9 (C-15/20), 141.4 (C-6), 128.7 (C-Ar), 128.6 (C-Ar), 128.5 (C-Ar), 127.8 (C-Ar), 127.2 (C-Ar), 125.3 (C-Ar), 120.1 (C-Ar), 82.8 (C-10), 67.3 (C-5/13), 66.9 (C-5/13), 51.1 (C-2), 47.3 (C-14), 37.1 (C-3), 28.0 (C-11); HRMS (ESI) calculated for $C_{30}H_{31}NO_6$ $[M+Na]^+$ 524.2049 found 524.2054.

N-Fmoc Aspartic Acid *O-tert*-Butyl Ester 275

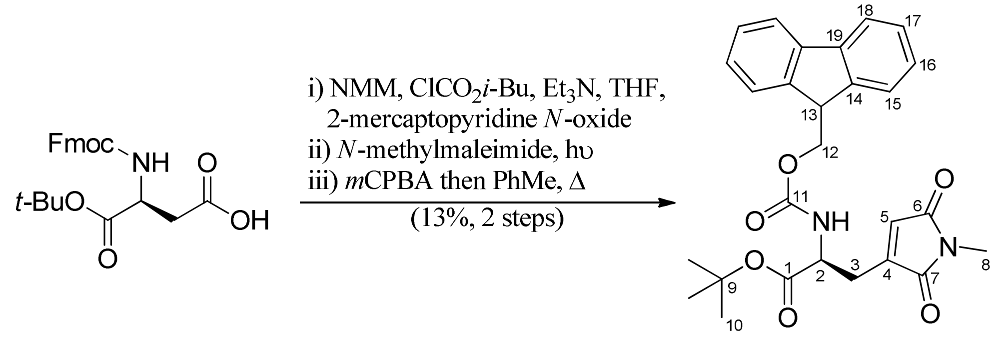


274 **275**

A round bottomed flask was charged with aspartic acid **274** (2.82 g, 5.62 mmol), palladium on carbon (10 wt. %, 119 mg, 0.11 mmol) and ethanol (50 mL). The flask was carefully flushed with dry nitrogen followed by hydrogen and then stirred under an atmosphere of hydrogen for 20 hours. The catalyst was removed by filtration through Celite® and the filtrate concentrated under reduced pressure to afford the desired carboxylic acid **275** as a colourless oil (2.22 g, 5.40 mmol, 96%) which was used without further purification.

(**275**, colourless oil) $[\alpha]_D^{21}$ −3.6 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3341, 2980, 1713, 1452, 1369, 1224, 1151, 1046; ^1H NMR (300 MHz, $CDCl_3$) δ 7.77 (d, *J*=7.4, 2H, H-Ar), 7.61 (d, *J*=7.4, 2H, H-Ar), 7.46-7.25 (m, 4H, H-Ar), 6.29 (s, br, 1H, O-H), 5.85 (d, *J*=8.1, 1H, N-H), 4.56 (dt, *J*=8.5, 4.4, 1H, H-2), 4.48-4.15 (m, 3H, H-8, H-9), 3.06 (dd, *J*=17.2, 4.5, 1H, H-3), 2.90 (dd, *J*=17.2, 4.5, 1H, H-3), 1.48 (s, 9H, H-6); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.6 (C-1/4), 169.5 (C-1/4), 156.1 (C-7), 143.7 (C-10/15), 141.3 (C-10/15), 127.7 (C-Ar), 127.1 (C-Ar), 125.1 (C-Ar), 120.0 (C-Ar), 82.9 (C-5), 67.3 (C-8), 50.8 (C-2), 47.1 (C-14), 36.7 (C-3), 27.8 (C-6); HRMS (ESI) calculated for $C_{23}H_{25}NO_6$ $[M+Na]^+$ 434.1580 found 434.1597.

N-Methylmaleimide Substituted *N*-Fmoc Aspartic Acid *O-tert*-Butyl Ester 277

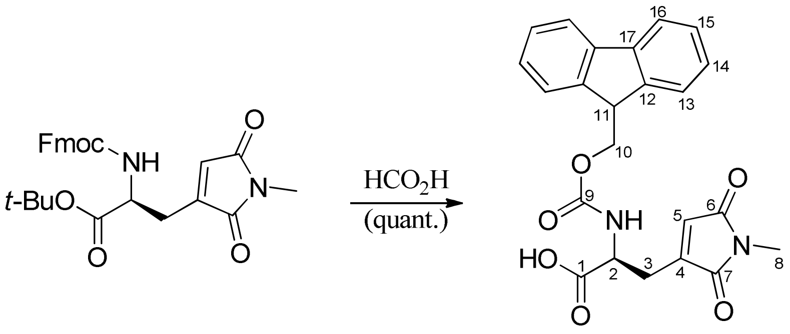


275 **277**

Aspartic acid **275** (2.10 g, 5.11 mmol) was dissolved in THF (60 mL), cooled to −20 °C, and treated sequentially with *N*-methyl morpholine (562 μL, 5.11 mmol) then *iso*butyl chloroformate (671 μL, 5.11 mmol). After 5 minutes a solution of 2-mercaptopyridine *N*-oxide (780 mg, 6.13 mmol) and triethylamine (921 μL, 6.64 mmol) in THF (20 mL) was added. The resulting mixture was stirred with the exclusion of light for 4 hours. The precipitate was removed by filtration and the filtrate irradiated in the presence of *N*-methylmaleimide (2.84 g, 25.6 mmol) for 1 hour with the temperature maintained below 10 °C. After removal of the volatile organics under reduced pressure, the residue was partitioned between dichloromethane (50 mL) and NaHCO$_3$ (40 mL, saturated aqueous solution). The layers were separated and the organic phase washed with HCl (0.5 N, 40 mL) then brine (40 mL) and dried over MgSO$_4$. The mixture was concentrated under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 3/1 to 1/1) to afford a white foam (792 mg, ~1.35 mmol, 26%). Signals in the ^{1}H NMR spectrum (300 MHz, CDCl$_3$) at 8.27-8.13, 7.68-7.54, 7.24-7.07 and 7.02-6.90 and mass spectrometry (HRMS (ESI) calculated for $C_{32}H_{33}N_3O_6S$ $[M+Na]^+$ 610.1988 found 620.1980) indicated that the 2-thiopyridyl moiety was present. A portion of the white foam (750 mg, 1.28 mmol) was dissolved in chloroform (12 mL), cooled to 0 °C and treated with a solution of *m*CPBA (70-75% in H$_2$O, 294 mg, ~1.28 mmol) in chloroform (12 mL). Further small portions of *m*CPBA were added until TLC indicated that all of the starting material had been consumed. The mixture was diluted with dichloromethane (40 mL) and washed with $NaHCO_3$ (saturated aqueous solution, 30 mL), HCl (0.5 N, 30 mL) and brine (30 mL). After drying over $MgSO_4$, the mixture was concentrated under reduced pressure and the resulting white foam dissolved in toluene (30 mL) then heated at reflux for 1 hour. The solvent was removed under reduced pressure and the residue purified by column chromatography (eluent petroleum ether/ethyl acetate, 7/3 to 2/3) to afford the desired compound **277** as a yellow foam (306 mg, 0.64 mmol, 50%).

(**277**, yellow foam) $[\alpha]_D^{21}$ +5.2 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2975, 1704, 1450, 1260, 1149, 1050, 1007; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.77 (d, *J*=7.5, 2H, H-Ar), 7.59 (d, *J*=7.5, 2H, H-Ar), 7.49-7.27 (m, 4H, H-Ar), 6.41 (s, 1H, H-5), 5.58 (d, *J*=7.4, 1H, N-H), 4.55 (dd, *J*=12.9, 7.4, 1H, H-2), 4.41 (m, 2H, H-12), 4.22 (t, *J*=6.8, 1H, H-13), 3.02 (s, 3H, H-8), 3.15-2.77 (m, 2H, H-3), 1.47 (s, 9H, H-10); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.9 (C=O), 175.8 (C=O), 171.5 (C=O), 171.3 (C=O), 143.9 (C-5/14/19), 143.8 (C-5/14/19), 141.5 (C-5/14/19), 129.0 (C-Ar), 127.9 (C-Ar), 127.2 (C-Ar), 125.1 (C-Ar), 120.2 (C-4), 83.5 (C-9), 67.3 (C-12), 52.92 (C-2/13), 52.91 (C-2/13), 28.7 (C-3), 28.1 (C-10), 24.0 (C-8); HRMS (ESI) calculated for $C_{27}H_{28}N_2O_6$ $[M+Na]^+$ 499.1845 found 499.1855.

N-Methylmaleimide Substituted *N*-Fmoc Aspartic Acid 278

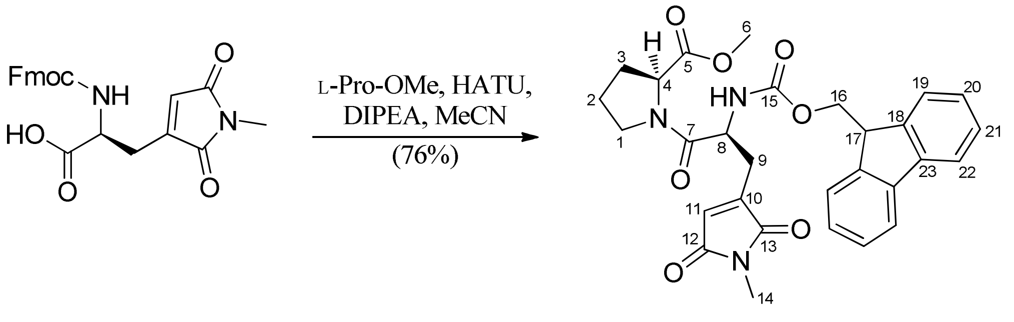


277 **278**

Aspartic acid **277** (198 mg, 0.42 mmol) was dissolved in neat formic acid (2 mL) and allowed to stir for 20 hours at room temperature. The volatile organics were removed under reduced pressure to afford the desired compound **278** as a yellow oil (186 mg, ~0.42 mmol) that was used without further purification.

(**278**, yellow oil) $[\alpha]_D^{20}$ −18.0 (*c* 1.00, MeOH); FTIR (film) ν_{max} 2963, 2925, 2855, 1709, 1642, 1450, 1263, 1122; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.45 (s, br, 1H, O-H), 7.75 (d, *J*=7.5, 2H, H-Ar), 7.57 (d, *J*=5.0, 2H, H-Ar), 7.47-7.22 (m, 4H, H-Ar), 6.44 (s, 1H, H-5), 5.79 (d, *J*=7.8, 1H, N-H), 4.67 (dd, *J*=12.7, 7.4, 1H, H-2), 4.41 (dd, *J*=6.7, 3.0, 2H, H-10), 4.20 (t, *J*=6.5, 1H, H-11), 3.15-2.87 (m, 2H, H-3), 2.99 (s, 3H, H-8); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.7 (C-1/6/7), 171.5 (C-1/6/7), 170.3 (C-1/6/7), 156.0 (C-9), 143.6 (C-4/12/17), 143.5 (C-4/12/17), 141.3 (C-4/12/17), 129.4 (C-Ar), 127.8 (C-Ar), 127.1 (C-Ar), 125.0 (C-Ar), 120.0 (C-5), 67.3 (C-10), 52.2 (C-2), 47.1 (C-11), 28.1 (C-3), 23.9 (C-8); HRMS (ESI) calculated for $C_{23}H_{19}N_2O_6$ $[M+Na]^+$ 419.1243 found 419.1245.

Proline-Maleimide Substituted *N*-Fmoc Aspartic Acid Dipeptide 279

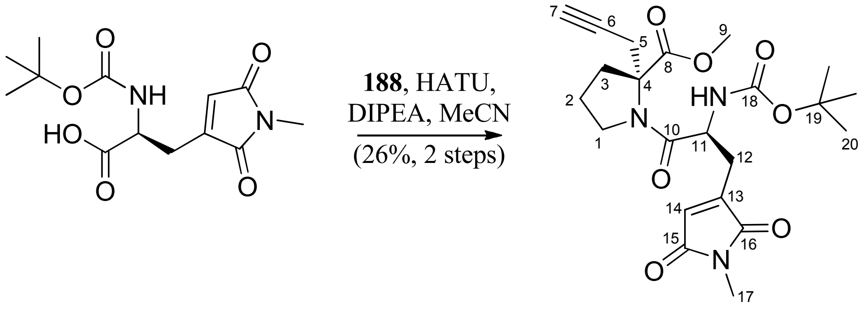


278 **279**

Aspartic acid **278** (186 mg, 0.42 mmol), L-proline *O*-methyl ester (73 mg, 0.44 mmol) and HATU (184 mg, 0.48 mmol) were dissolved in acetonitrile (10 mL) and treated with di*iso*propylethylamine (161 µL, 0.92 mmol). The resulting mixture was allowed to stir at room temperature for 20 hours then concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 to 1/3) to afford the desired dipeptide **279** as an orange oil (168 mg, 0.32 mmol, 76% from **277**).

(**279**, orange oil) $[\alpha]_D^{20}$ −7.0 (*c* 0.50, $CHCl_3$); FTIR (film) ν_{max} 2966, 2855, 1707, 1645, 1450, 1264, 1102, 1024; ^1H NMR (300 MHz, $CDCl_3$) δ 7.82-7.73 (m, 2H, H-Ar), 7.61-7.53 (m, 2H, H-Ar), 7.45-7.23 (m, 4H, H-Ar), 6.45 (s, 1H, H-11), 5.82 (d, *J*=8.4, 1H, N-H), 4.88-4.78 (m, 1H, H-8), 4.55 (m, 1H, H-4), 4.46-4.26 (m, 2H, H-16), 4.18 (m, 1H, H-17), 3.87-3.65 (m, 2H, H-1), 3.74 (s, 3H, H-6), 3.03 (s, 3H, H-14), 3.13-2.61 (m, 2H, H-9), 2.37-1.91 (m, 4H, H-2, H-3); ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.0 (C=O), 171.9 (C=O), 171.6 (C=O), 171.4 (C=O), 156.0 (C-15), 143.9 (C-10), 143.7 (C-18/23), 141.3 (C-18/23), 129.5 (C-11), 127.7 (C-Ar), 127.1 (C-Ar), 125.0 (C-Ar), 120.0 (C-Ar), 67.3 (C-16), 59.0 (C-4), 52.5 (C-6), 50.9 (C-8), 47.4 (C-1), 47.0 (C-17), 29.1 (C-3/9), 29.0 (C-3/9), 24.9 (C-2), 23.9 (C-14); HRMS (ESI) calculated for $C_{29}H_{29}N_3O_7$ $[M+Na]^+$ 554.1903 found 554.1909.

Proline-*N*-Methylmaleimide Substituted Aspartic Acid Dipeptide 284



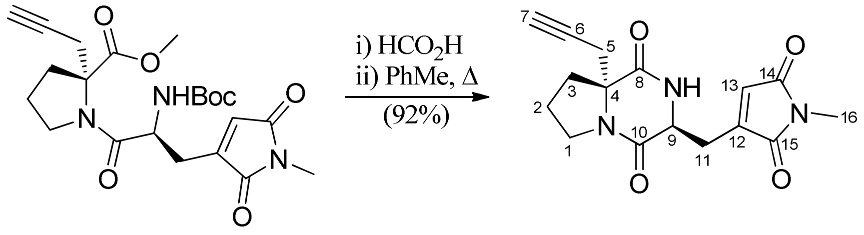
267 **284**

General procedure B was followed with aspartic acid **267** (1.0 g, 3.35 mmol), proline **188** (327 mg, 1.61 mmol), HATU (673 mg, 1.77 mmol) and di*iso*propylethylamine (0.59 mL, 3.38 mmol) to afford the title compound **284** as a colourless oil (184 mg, 0.41 mmol, 26%).

(**284**, colourless oil) $[\alpha]_D^{22}$ −20.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2927, 1702, 1643, 1434, 1390, 1247, 1161; ^1H NMR (300 MHz, $CDCl_3$) δ 6.39 (s, 1H, H-14), 5.37 (d, *J*=9.0, 1H, N-

H), 4.74 (td, *J*=9.4, 3.8, 1H, H-11), 4.36 (m, 1H, H-1), 3.76 (m, 1H, H-1), 3.67 (s, 3H, H-9), 3.40 (dd, *J*=17.2, 2.7, 1H, H-5), 3.00 (s, 3H, H-17), 2.88 (m, 1H, H-12), 2.74 (m, 1H, H-5), 2.53 (dd, *J*=14.5, 9.6, 1H, H-12), 2.44-1.85 (m, 4H, H-2, H-3), 1.94 (t, *J*=2.7, 1H, H-7), 1.34 (s, 9H, H-20); ^{13}C NMR (100 MHz, CDCl$_3$) δ 172.6 (C=O), 171.6 (C=O), 170.7 (C=O), 168.9 (C=O), 155.3 (C-18), 145.1 (C-13), 128.7 (C-14), 80.1 (C-6/14), 80.0 (C-6/14), 70.9 (C-7), 68.0 (C-4), 52.8 (C-9), 50.7 (C-11), 49.3 (C-1), 36.1 (C-3), 29.7 (C-12), 28.3 (C-20), 24.5 (C-5), 24.1 (C-2), 23.9 (C-17); HRMS (ESI) calculated for $C_{22}H_{29}N_3O_7$ [M+Na]$^+$ 470.1903 found 470.1894.

N-Methylmaleimide DKP 285

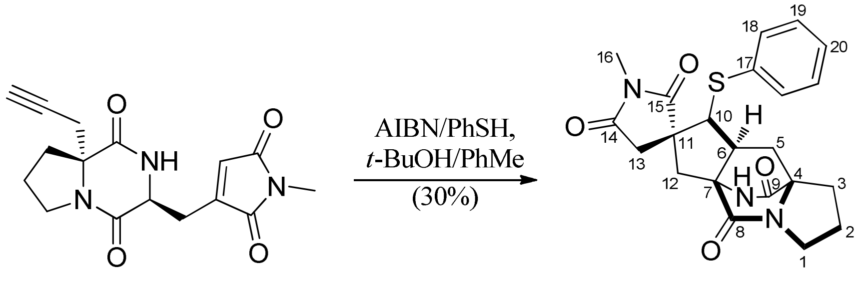


284 **285**

Dipeptide **284** (280 mg, 0.63 mmol) was dissolved in formic acid (3 mL) and allowed to stir at room temperature for 4 hours. The volatile organics were removed under reduced pressure and the residue dissolved in toluene. The resulting solution was heated at reflux for 2 hours then concentrated under reduced pressure. The residue was purified by column chromatography (eluent petroleum ether/ethyl acetate, 1/1 to 100% ethyl acetate) to afford the desired DKP **285** (183 mg, 0.58 mmol, 92%) as a colourless oil.

(**285**, white solid) m.p. 155 °C (dec.); $[\alpha]_D^{20}$ −61.5 (*c* 1.00, CHCl$_3$); FTIR (film) ν_{max} 2962, 2896, 1704, 1651, 1422, 1388, 1259, 1004; ^{1}H NMR (300 MHz, CDCl$_3$) δ 7.05 (s, 1H, N-H), 6.48 (t, *J*=1.4, 1H, H-13), 4.78 (dd, *J*=6.6, 4.7, 1H, H-9), 3.83 (m, 1H, H-1), 3.57 (m, 1H, H-1), 3.21 (ddd, *J*=15.3, 4.7, 1.4, 1H, H-11), 3.00 (s, 3H, H-16), 2.96 (m, 1H, H-11), 2.75 (dd, *J*=17.0, 2.7, 1H, H-5), 2.61 (dd, *J*=17.0, 2.7, 1H, H-5), 2.10 (t, *J*=2.7, 1H, H-7), 2.21-1.91 (m, 4H, H-2, H-3); ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.0 (C=O), 171.0 (C=O), 170.5 (C=O), 164.4 (C=O), 144.4 (C-12), 130.4 (C-13), 78.5 (C-6), 72.6 (C-7), 67.4 (C-4), 54.3 (C-9), 45.5 (C-1), 35.1 (C-3), 27.8 (C-11), 27.3 (C-5), 24.1 (C-16), 20.4 (C-2); HRMS (ESI) calculated for $C_{16}H_{17}N_3O_4$ $[M+Na]^+$ 338.1117 found 338.1103.

Asperparaline Core Structure 287

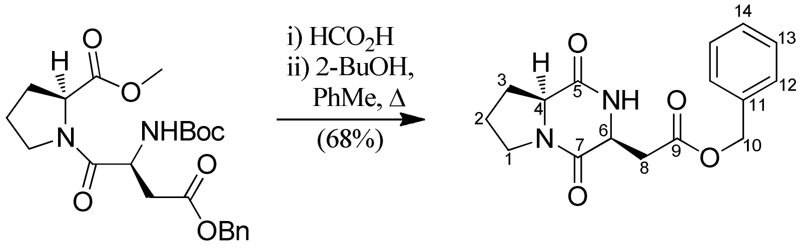


285 **287**

General procedure A was followed with DKP **285** (48 mg, 0.15 mmol), AIBN (0.2 M solution in toluene, 1.50 mL, 0.30 mmol), thiophenol (31 μL, 0.30 mmol) and *tert*-butanol (25 mL) to afford the desired bridged compound **287** as a slightly yellow solid (20 mg, 0.05 mmol, 30%). (**287**, yellow solid) m.p. 163 °C – 165 °C; $[\alpha]_D^{20}$ −32.5 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 2920, 1699, 1683, 1437, 1424, 1280, 1002; ^{1}H NMR (500 MHz, C_6D_6) δ 7.30 (s, 1H, N-H), 7.19 (dd, *J*=6.4, 3.0, 2H, H-Ar), 6.87 (m, 3H, H-Ar), 6.30 (s, 1H, N-H), 3.11 (m, 1H, H-1), 3.05 (br. s, 1H, H-6), 3.04 (m, 1H, H-10), 2.95 (dt, *J*=11.3, 7.4, 1H, H-1), 2.86 (d, *J*=18.6, 1H, H-13), 2.78 (s, 3H, H-16), 2.58 (m, 1H, H-3), 2.58 (d, *J*=12.9, 1H, H-12), 2.53 (d, *J*=18.6, 1H, H-13), 2.36 (d, *J*=12.9, 1H, H-12), 1.75 (dd, *J*=13.3, 8.6, 1H, H-5), 1.39-1.30 (m, 2H, H-2, H-5), 1.26-1.11 (m, 2H, H-2, H-3); ^{13}C NMR (100 MHz, C_6D_6) δ 180.2 (C=O), 174.4 (C=O), 173.0 (C=O), 167.8 (C=O), 133.8 (C-17), 133.0 (CH-Ar), 129.7 (CH-Ar), 128.5 (CH-Ar), 68.7 (C-4), 66.1 (C-7), 62.5 (C-10), 55.2 (C-11), 52.7 (C-6), 44.2 (C-1), 43.8 (C-18), 37.2 (C-

12), 34.3 (C-5), 30.2 (C-3), 24.6 (C-2, C-23); HRMS (ESI) calculated for $C_{22}H_{23}N_3O_4S$ $[M+Na]^+$ 448.1307 found 448.1306.

Proline-Aspartic Acid *O*-Benzyl Ester DKP 304

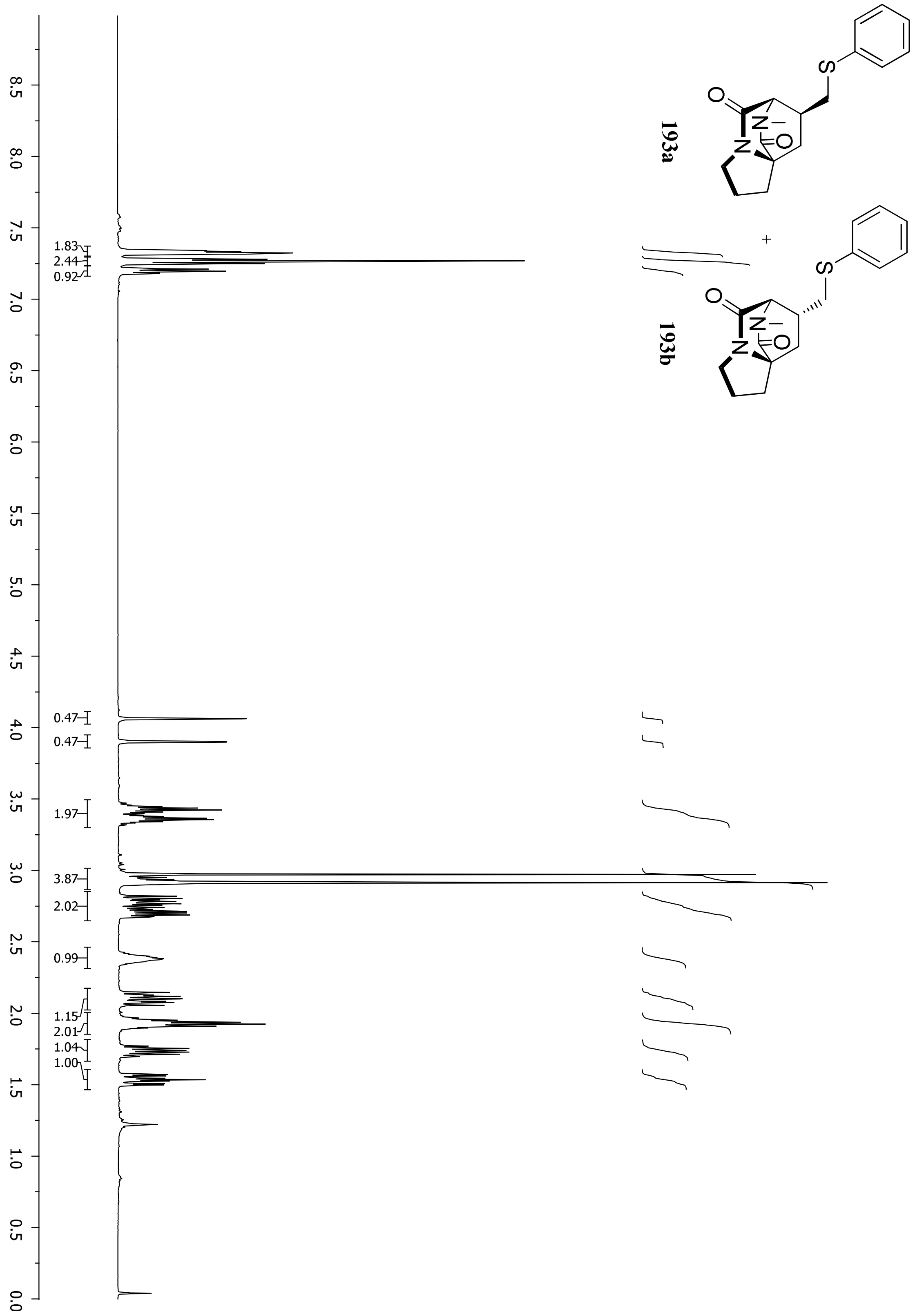


241 **304**

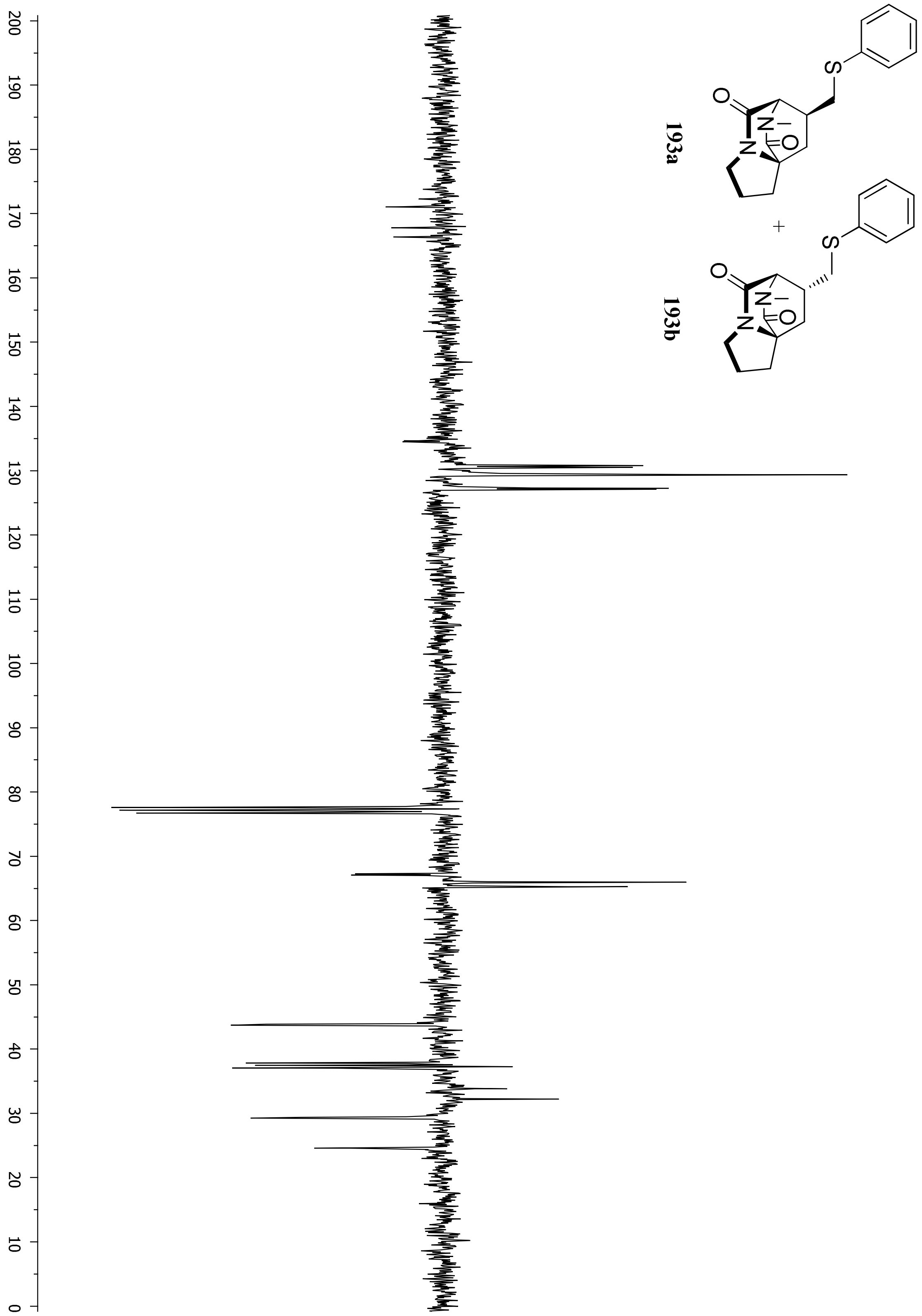
General procedure C was followed with dipeptide **241** (4.48 g, 10.3 mmol), formic acid (40 mL), 2-butanol (75 mL) and toluene (25 mL) to afford the title compound **304** as a colourless oil (2.11 g, 6.98 mmol, 68%).

(**304**, colourless oil) $[\alpha]_D^{21}$ −43.0 (*c* 1.00, $CHCl_3$); FTIR (film) ν_{max} 3221, 1731, 1675, 1636, 1453, 1319, 1262, 1171, 1007; ^1H NMR (300 MHz, $CDCl_3$) δ 7.44-7.30 (m, 5H, H-Ar), 6.77 (s, 1H, N-H), 5.15 (s, 2H, H-10), 4.51 (d, *J*=8.8, 1H, H-6), 4.19-3.88 (m, 3H, H-1, H-4), 3.58 (m, 1H, H-1), 3.17 (dd, *J*=17.6, 3.1, 1H, H-8), 2.88 (dd, *J*=17.6, 9.0, 1H, H-8), 2.81–2.68 (m, 2H, H-3), 2.13-1.76 (m, 2H, H-2); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.7 (C=O), 161.1 (C=O), 157.0 (C=O), 135.2 (C-11), 128.8 (CH-Ar), 128.8 (CH-Ar), 128.6 (CH-Ar), 67.5 (C-10), 67.3 (C-1), 54.3 (C-6), 54.0 (C-4), 45.8 (C-8), 39.0 (C-3), 28.0 (C-2); HRMS (ESI) calculated for $C_{16}H_{18}N_2O_4$ $[M+Na]^+$ 325.1164 found 325.1171.

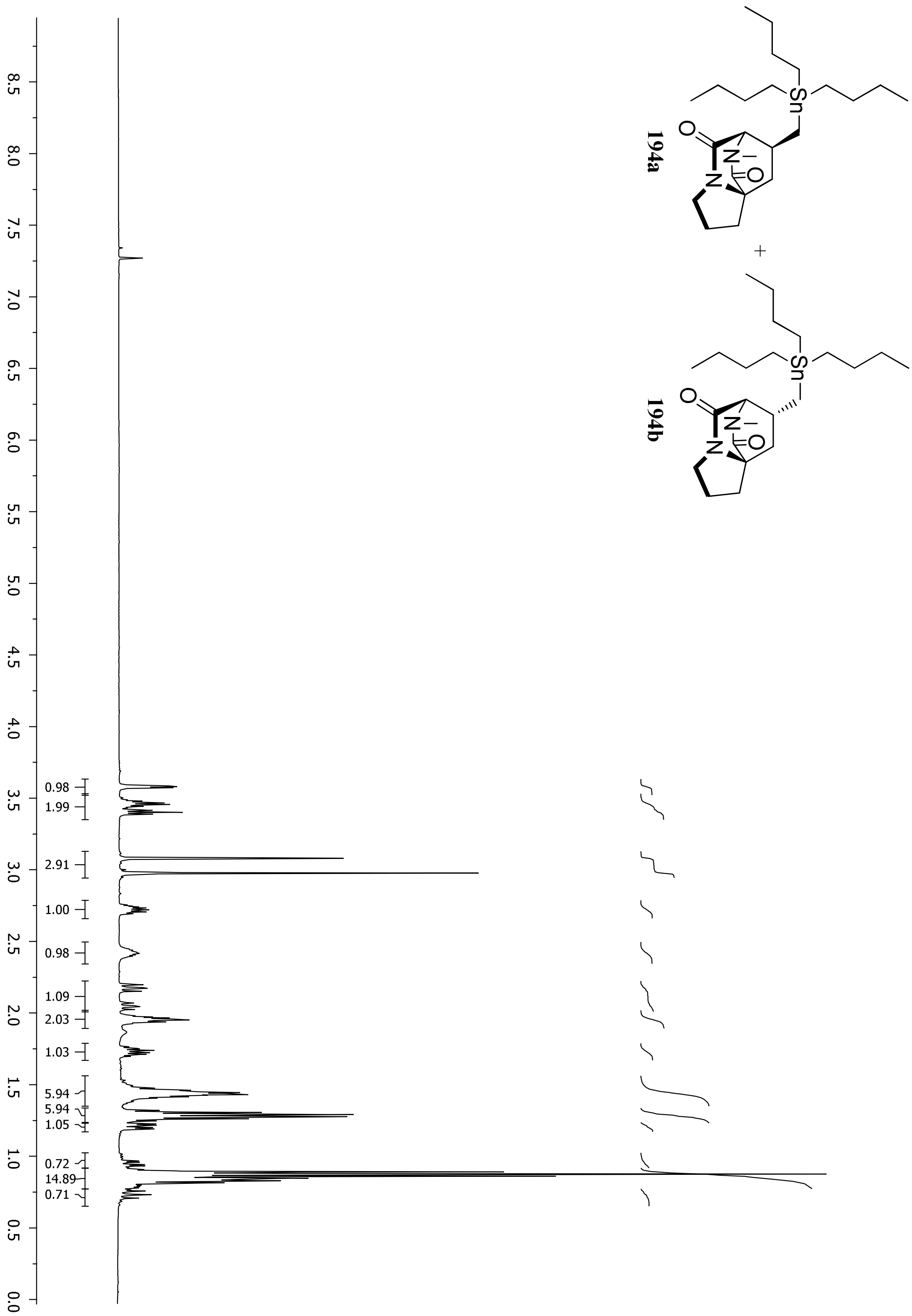
^{1}H NMR spectrum of bridged DKPs **193a** and **193b** (500 MHz, $CDCl_3$)



^{13}C NMR spectrum of bridged DKPs **193a** and **193b** (75 MHz, CDCl$_3$)

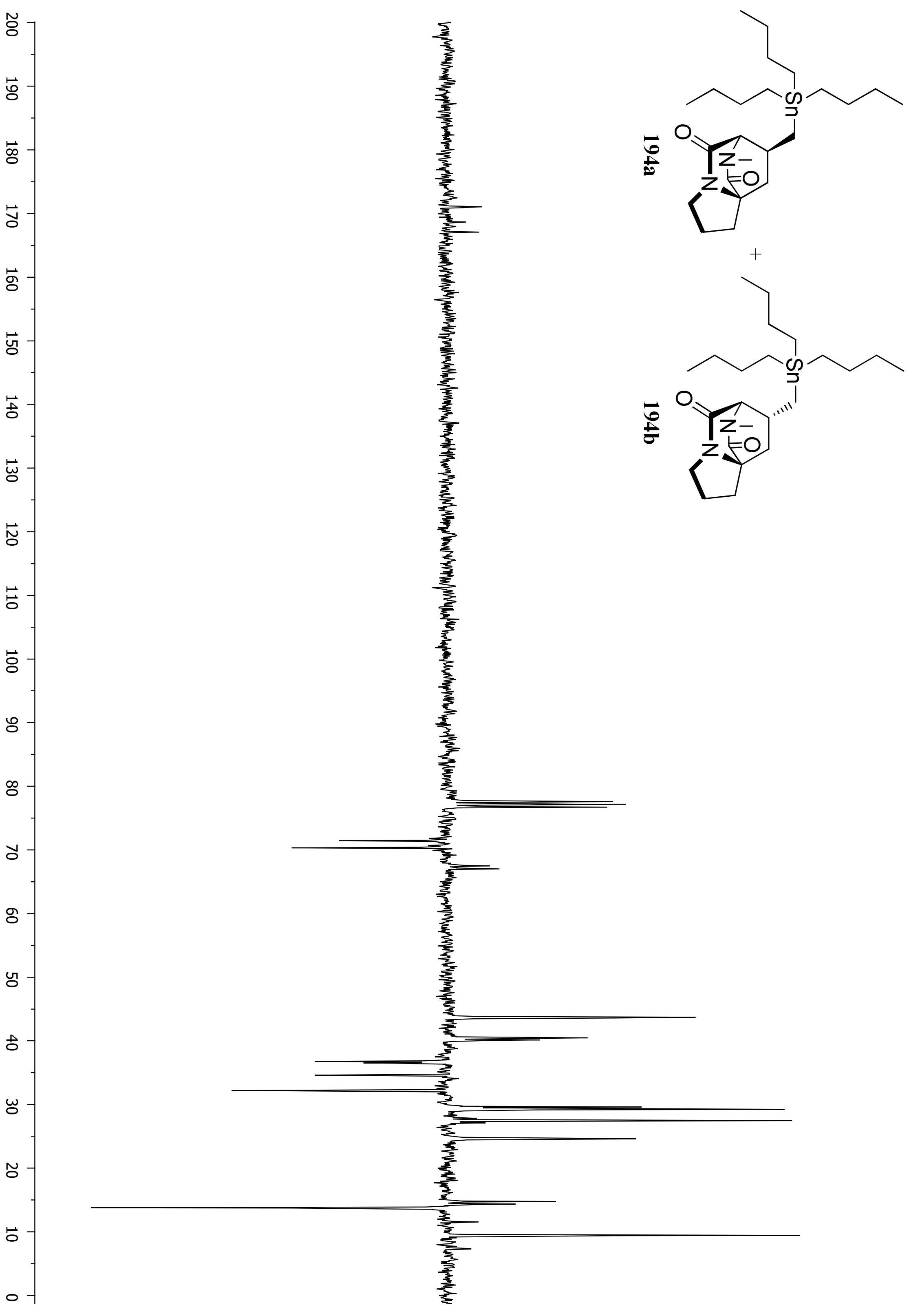


^{1}H NMR spectrum of bridged DKPs **194a** and **194b** (500 MHz, $CDCl_3$)

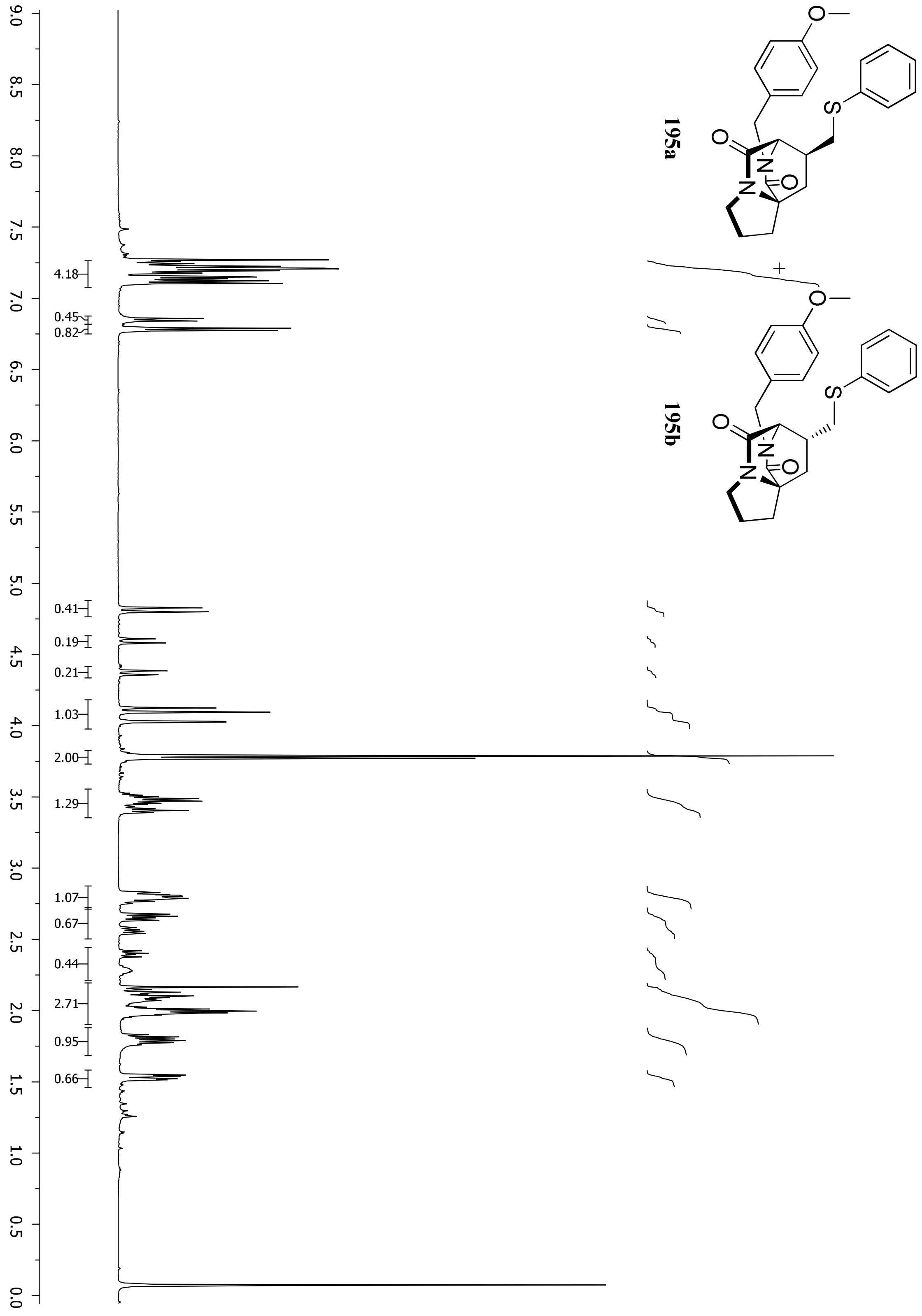

194a

194b

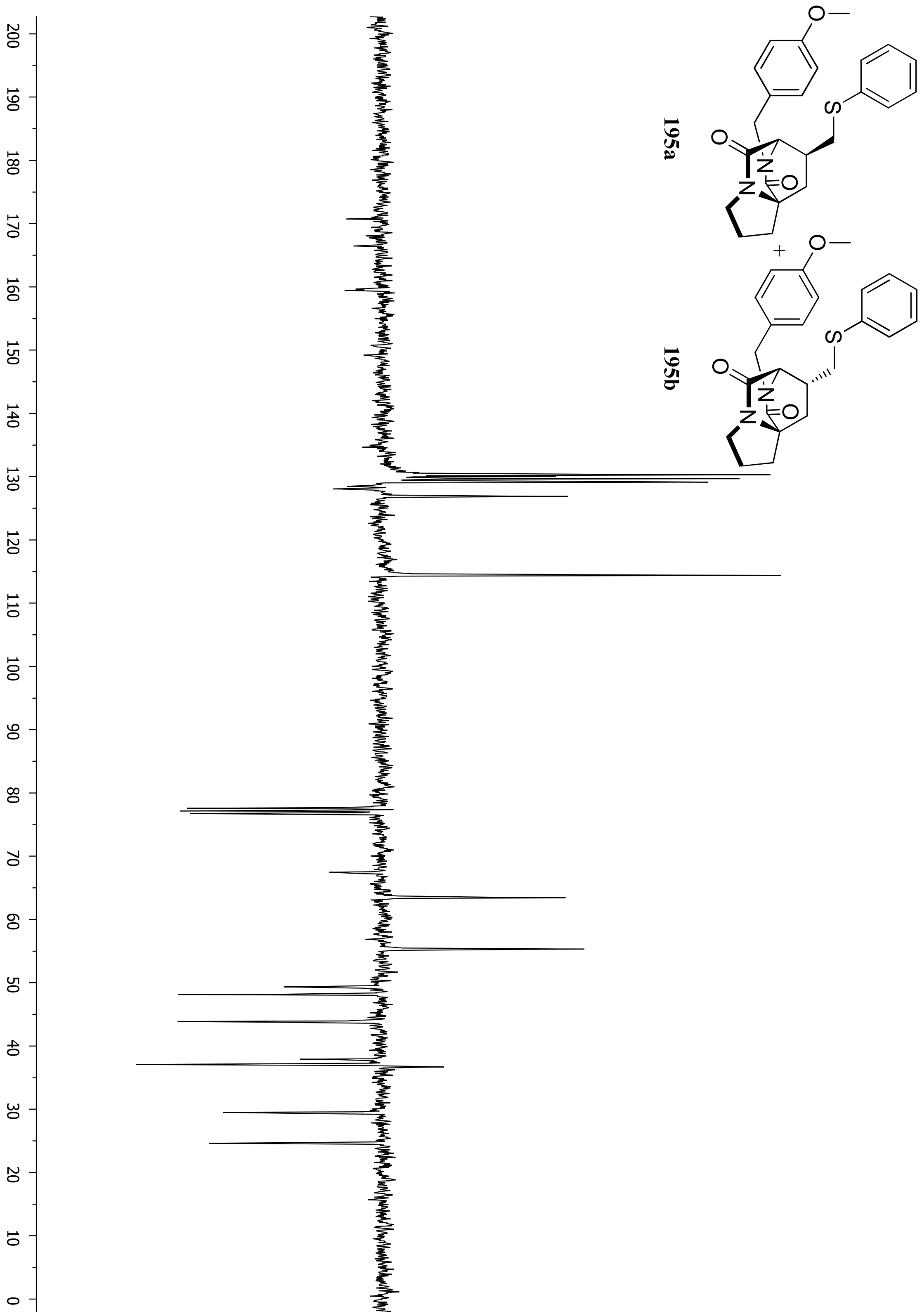
^{13}C NMR spectrum of bridged DKPs **194a** and **194b** (75 MHz, $CDCl_3$)



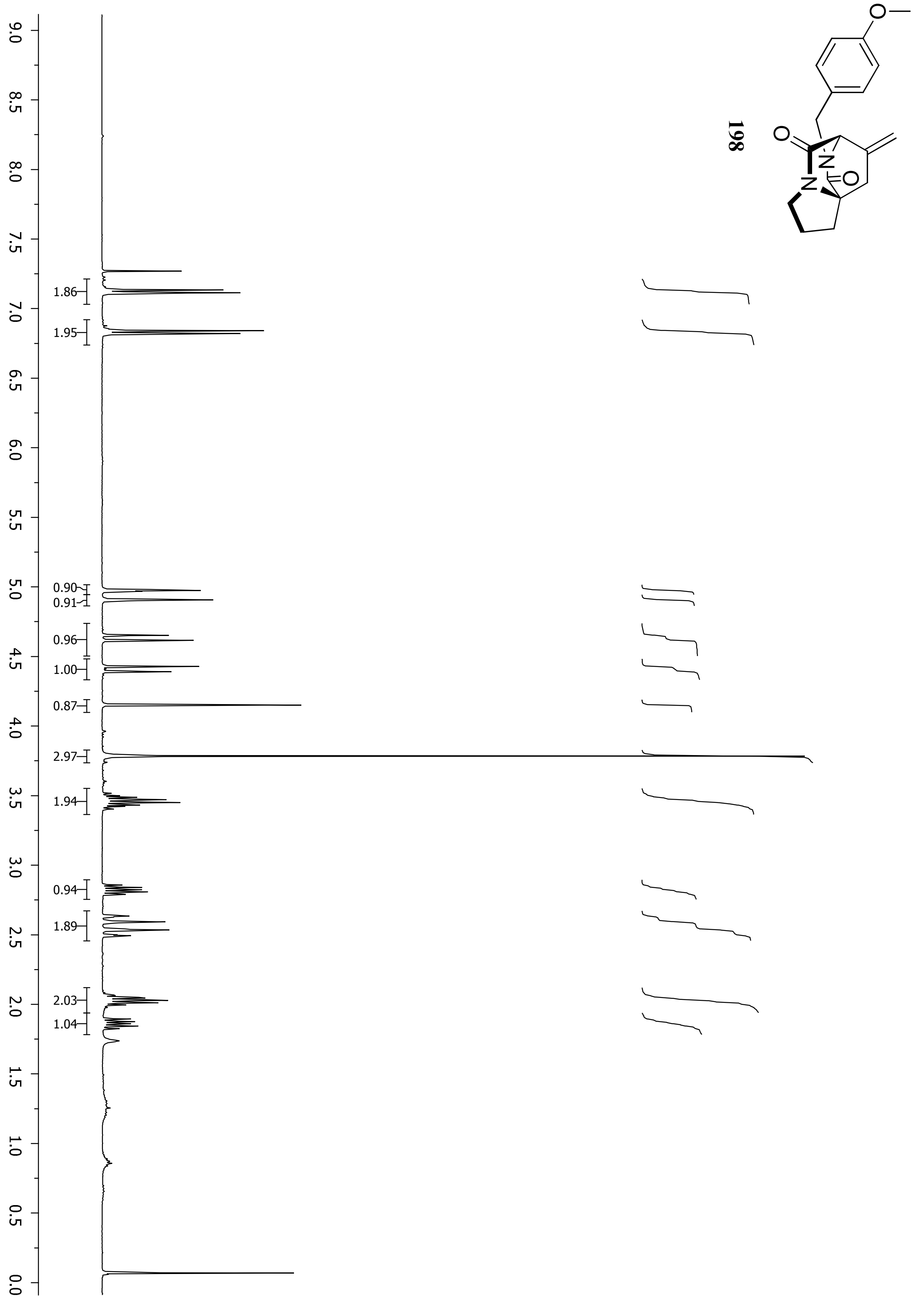
^{1}H NMR spectrum of bridged DKPs **195a** and **195b** (500 MHz, CDCl$_3$)



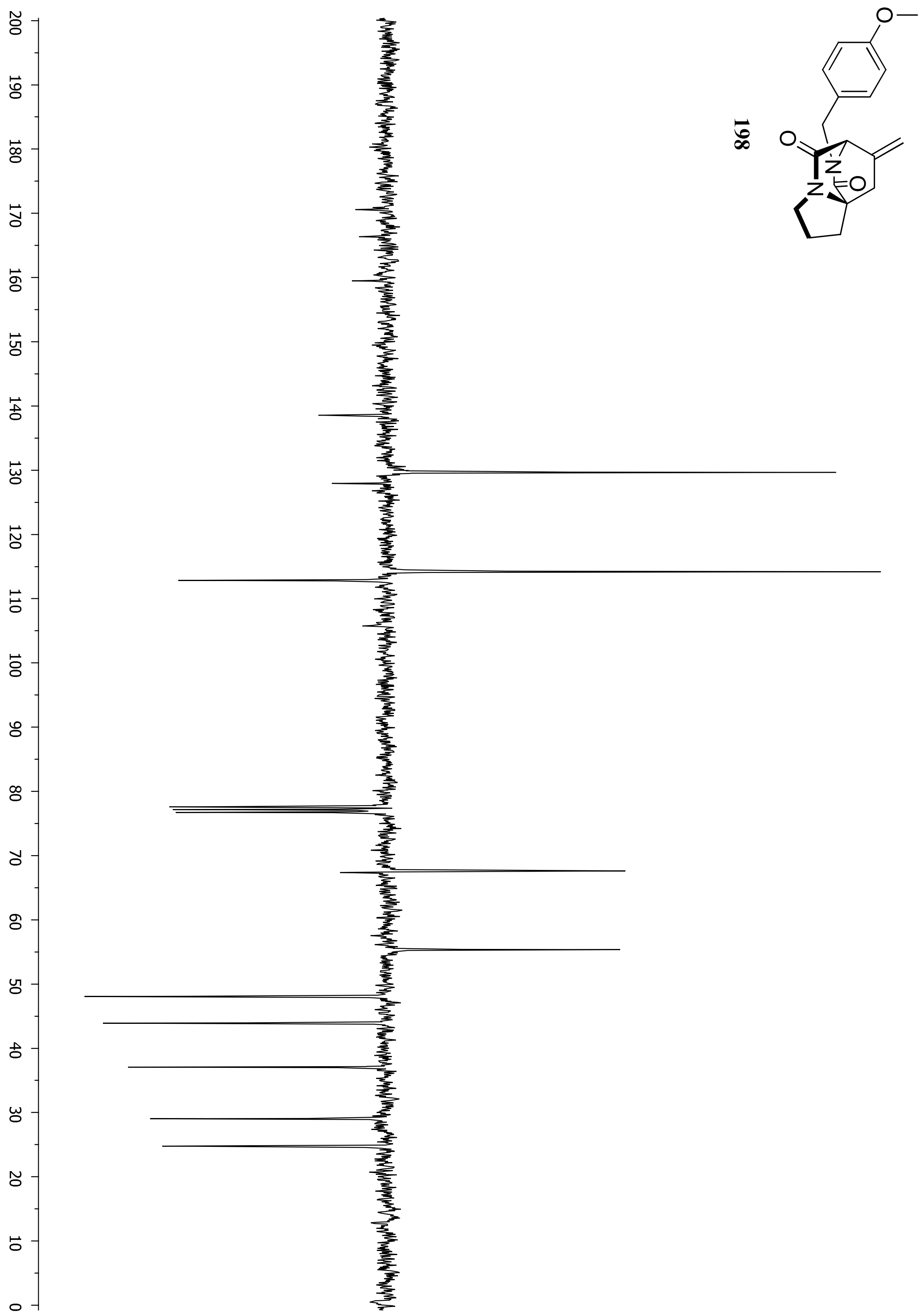
^{13}C NMR spectrum of bridged DKPs **195a** and **195b** (75 MHz, $CDCl_3$)



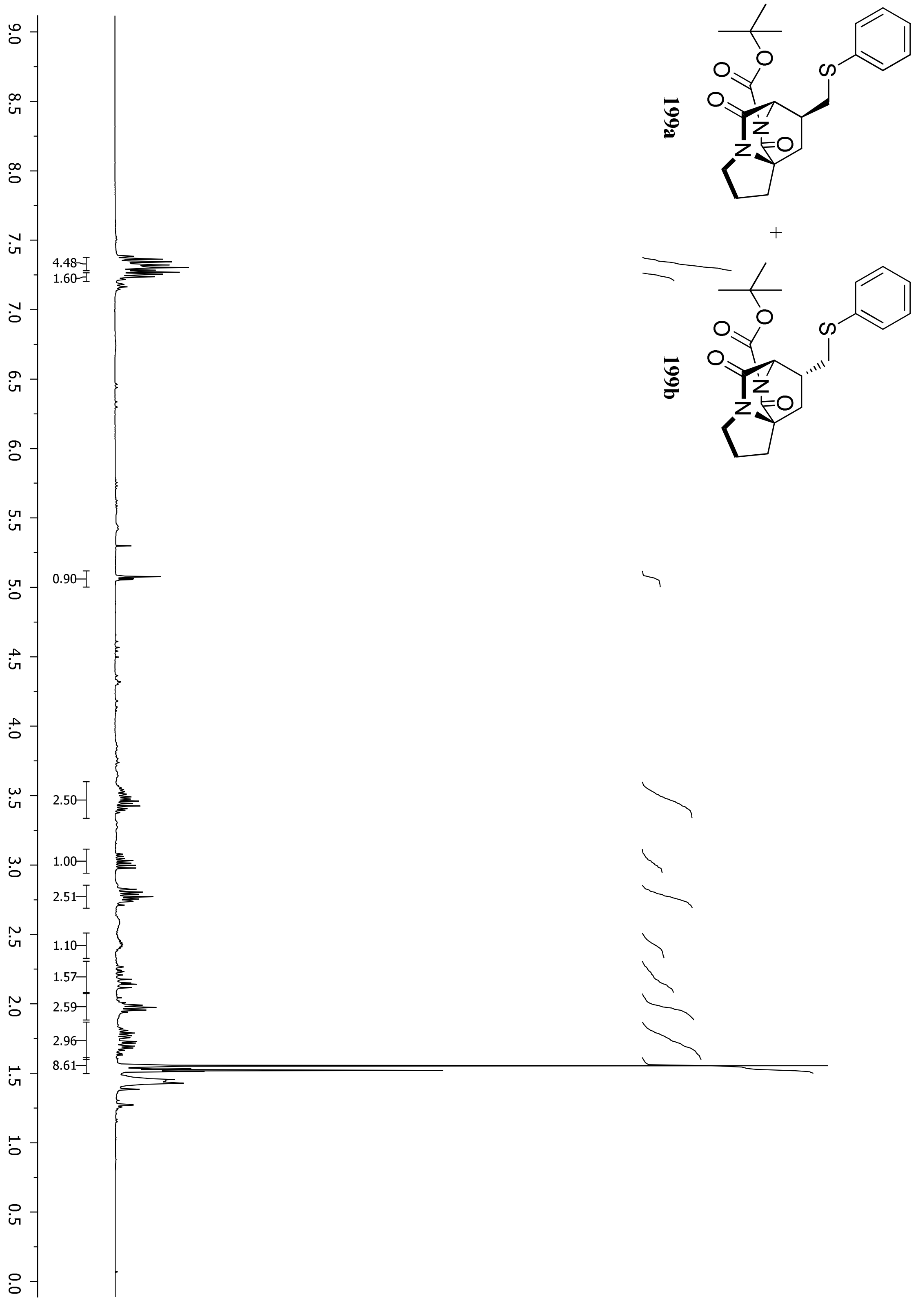
^{1}H NMR spectrum of bridged DKP **198** (400 MHz, $CDCl_3$)



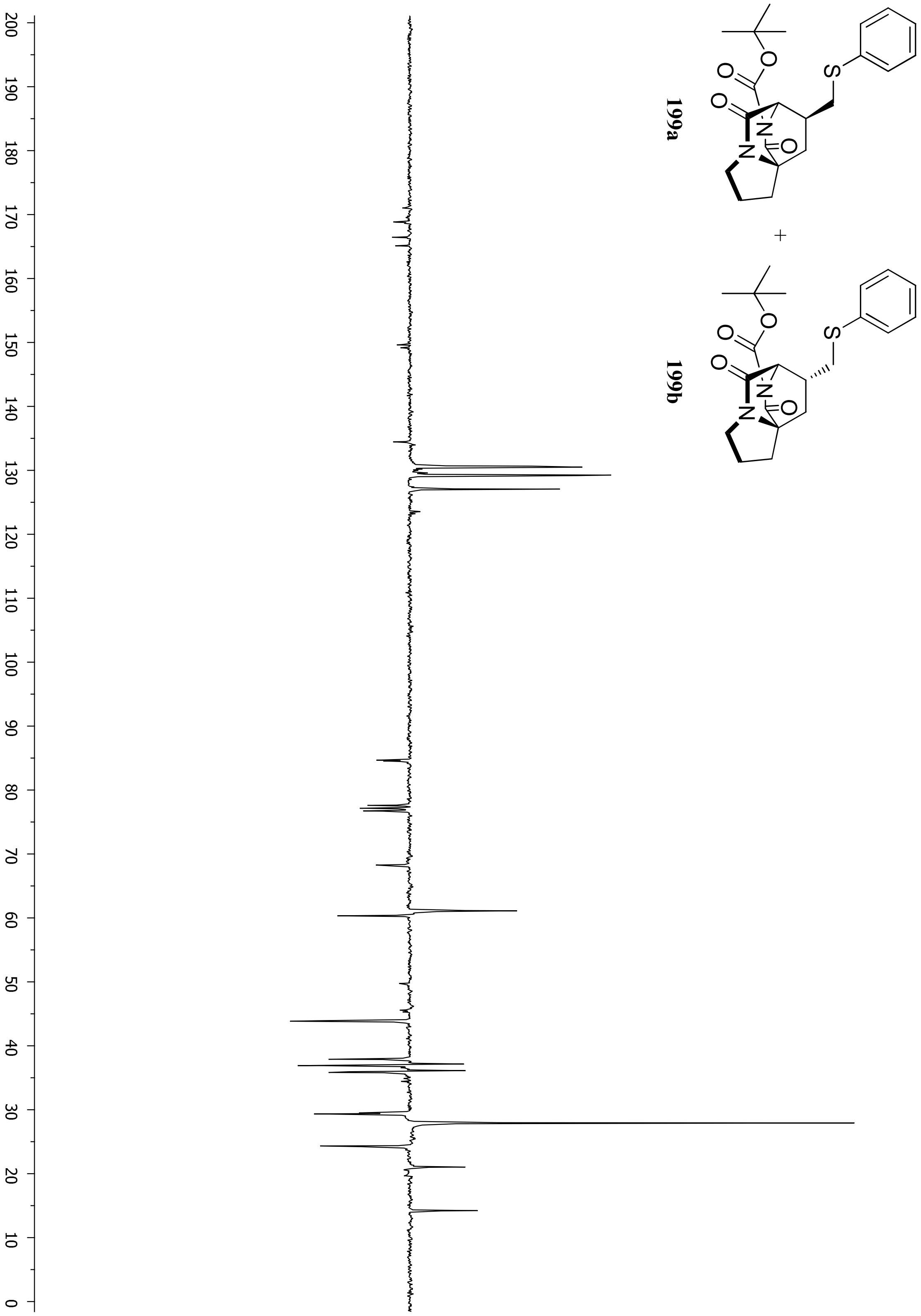
^{13}C NMR spectrum of bridged DKP **198** (75 MHz, CDCl$_3$)



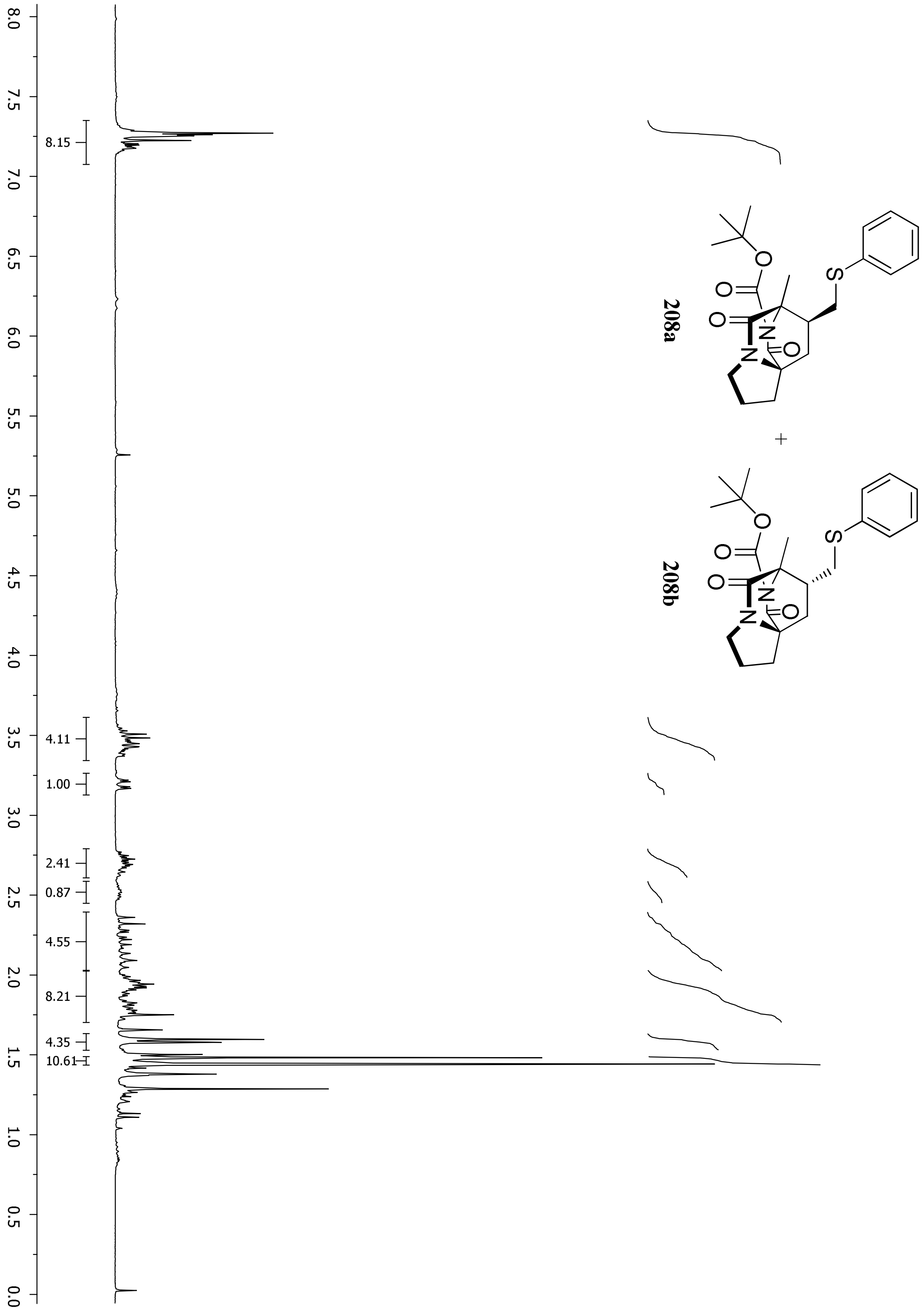
^{1}H NMR spectrum of bridged DKPs **199a** and **199b** (400 MHz, $CDCl_3$)



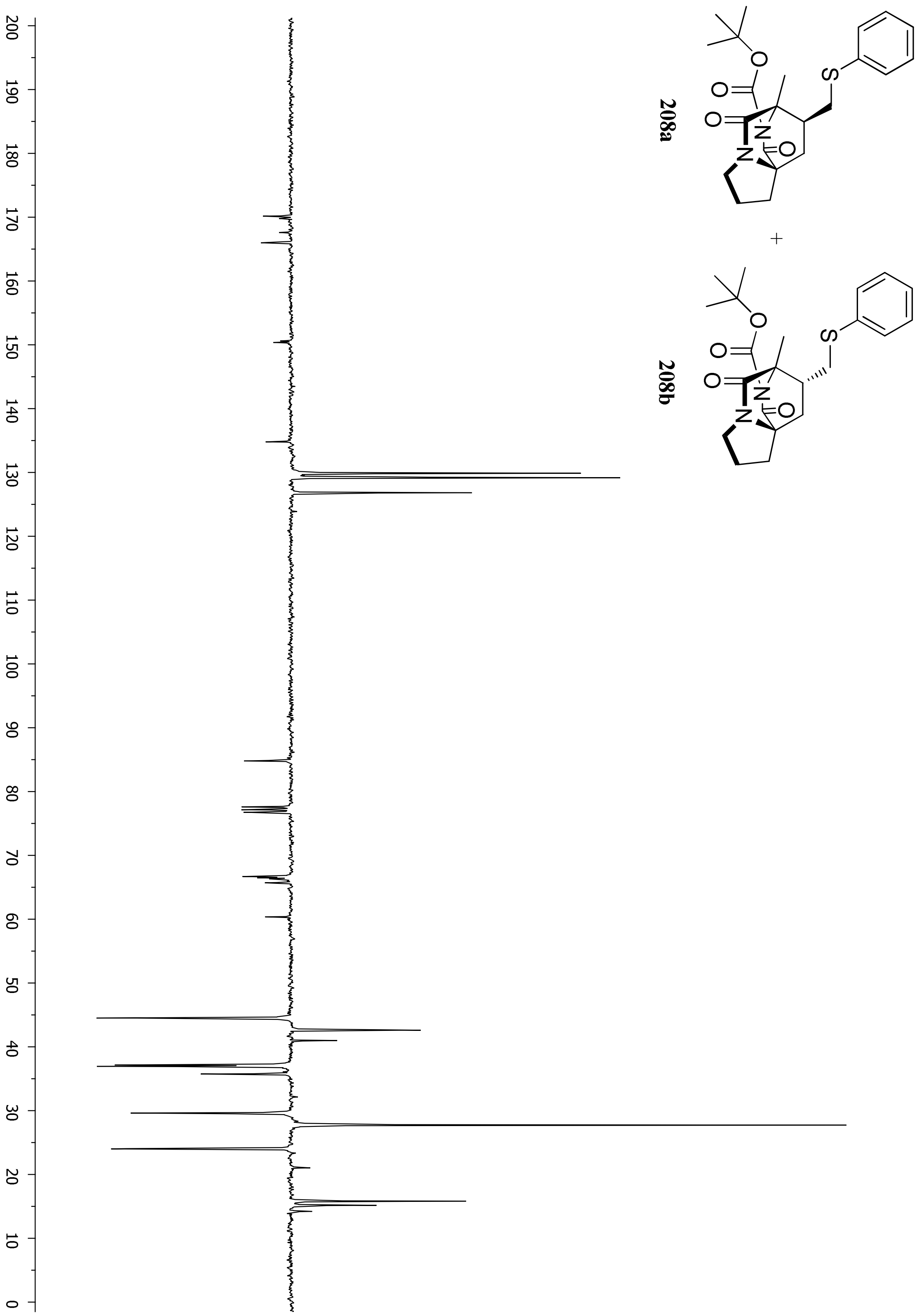
^{13}C NMR spectrum of bridged DKPs **199a** and **199b** (75 MHz, CDCl$_3$)



^{1}H NMR spectrum of bridged DKPs **208a** and **208b** (300 MHz, CDCl$_3$)



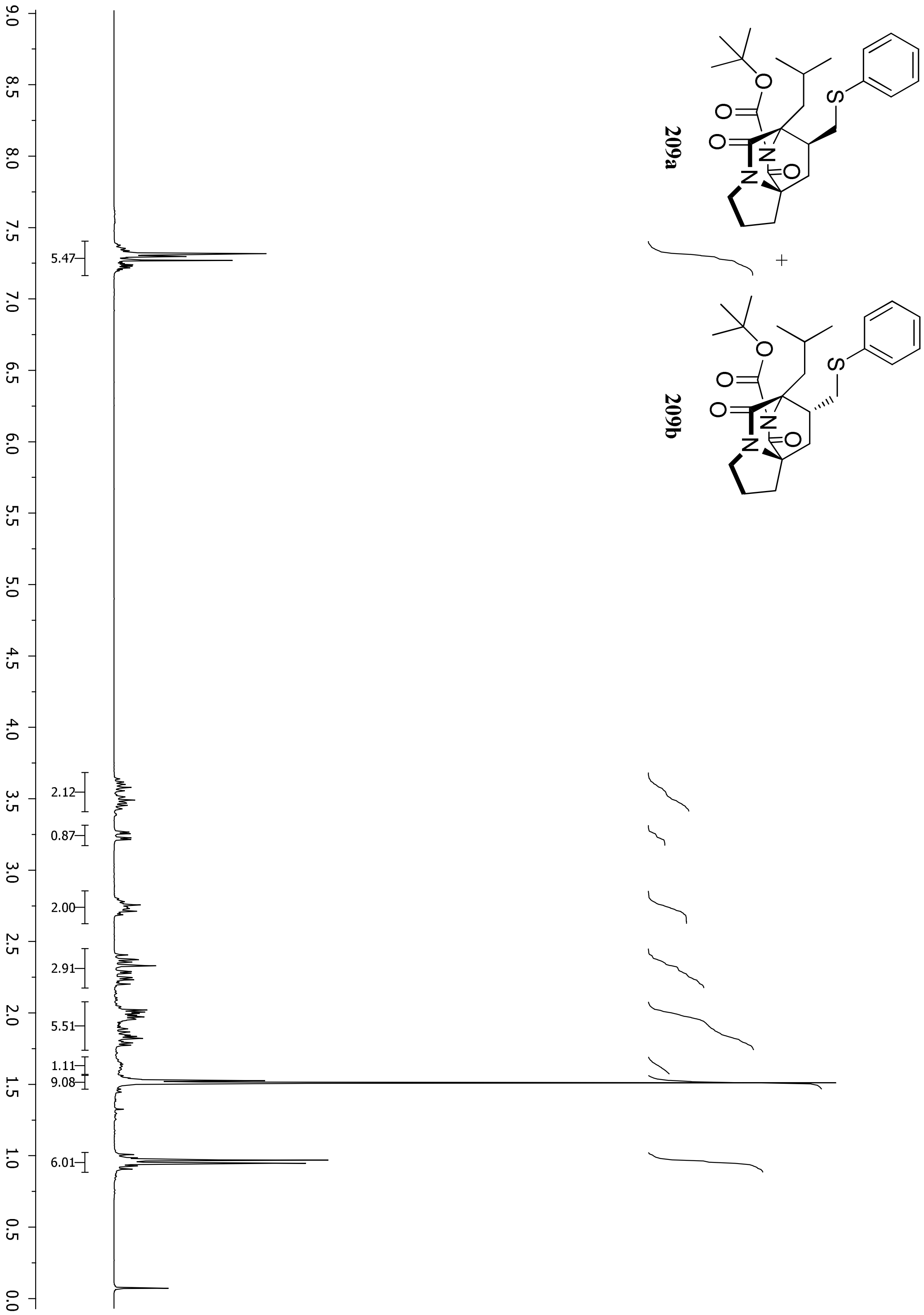
^{13}C NMR spectrum of bridged DKPs **208a** and **208b** (75 MHz, CDCl$_3$)



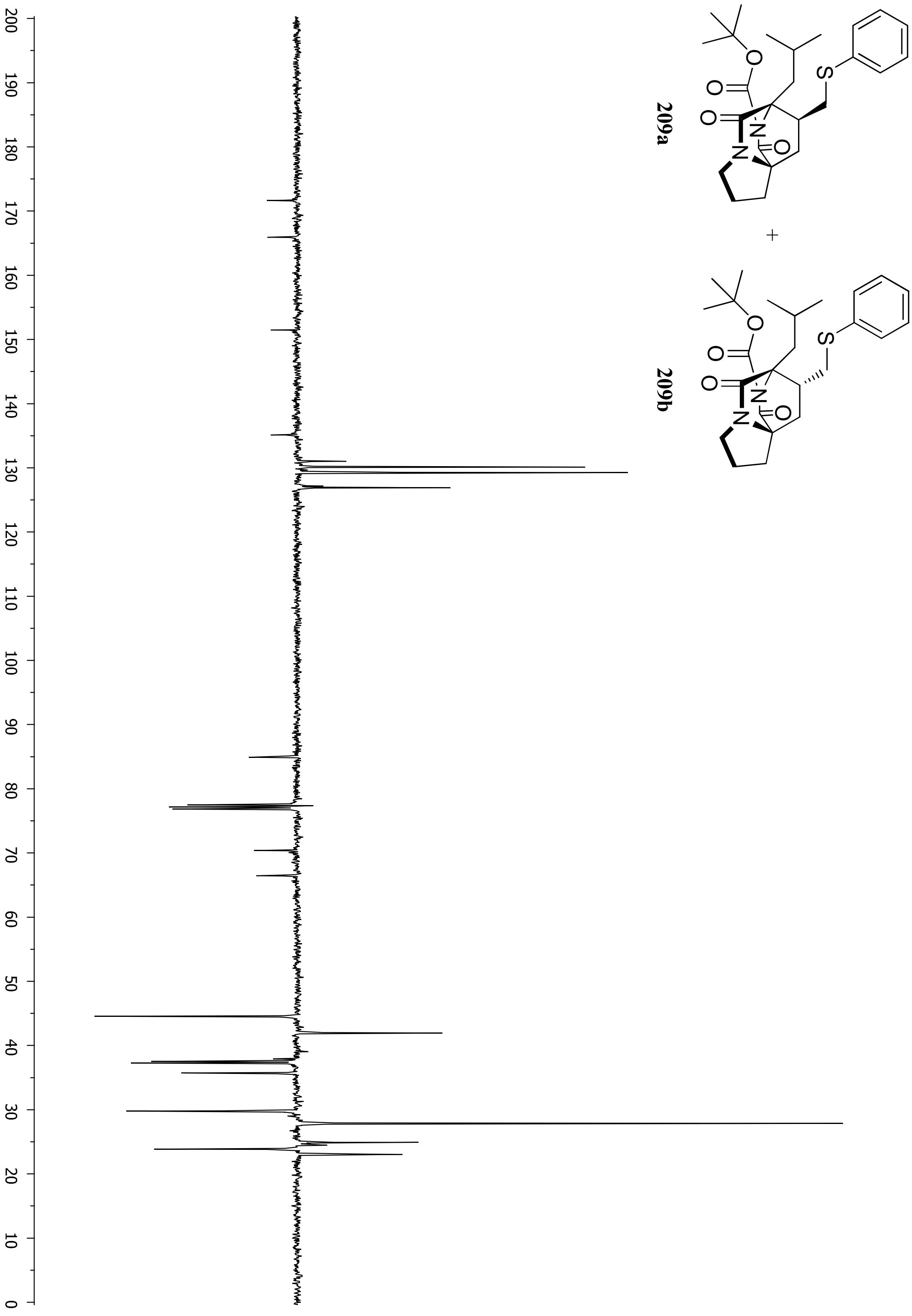
208a

208b

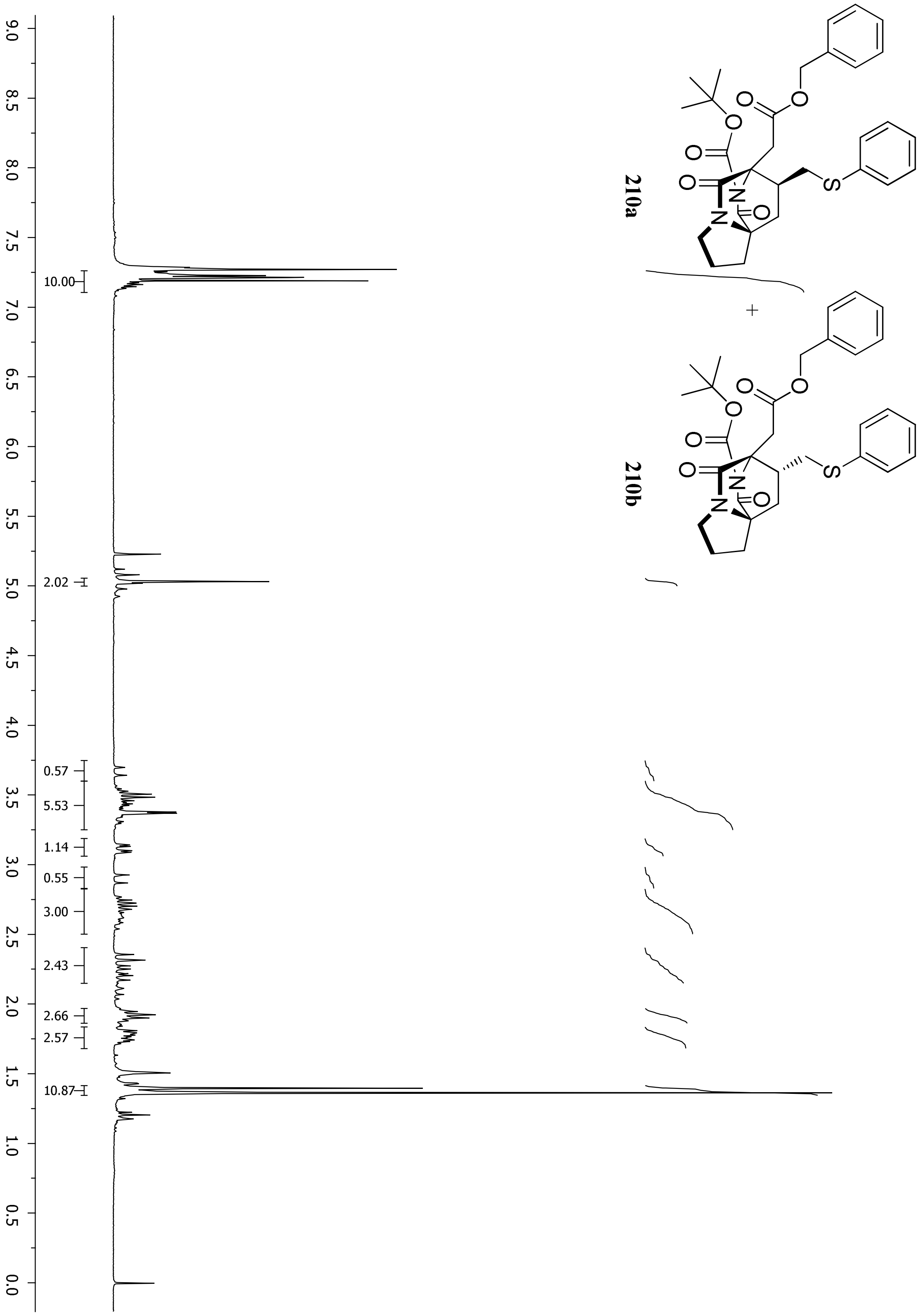
^{1}H NMR spectrum of bridged DKPs **209a** and **209b** (300 MHz, $CDCl_3$)



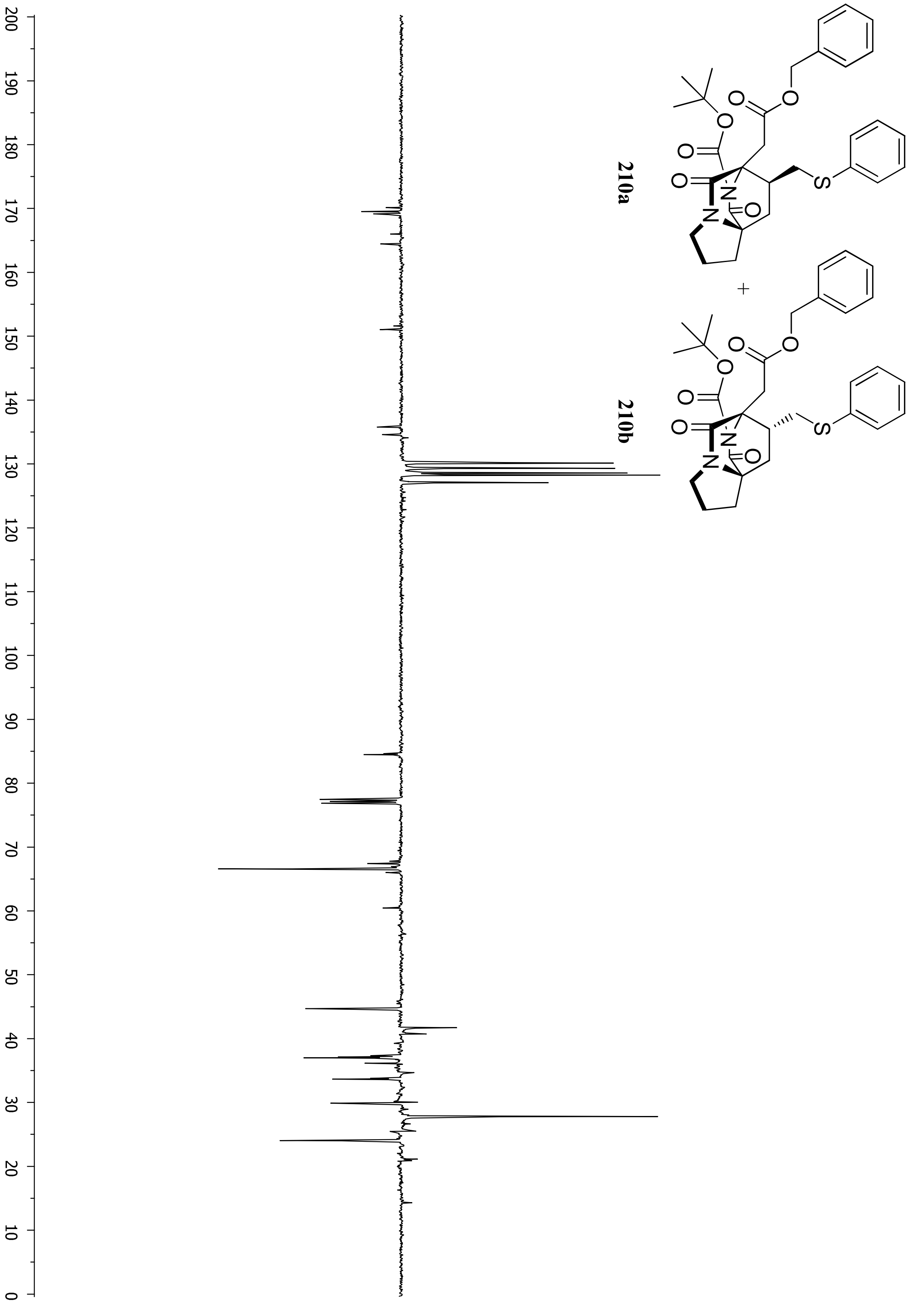
^{13}C NMR spectrum of bridged DKPs **209a** and **209b** (100 MHz, $CDCl_3$)



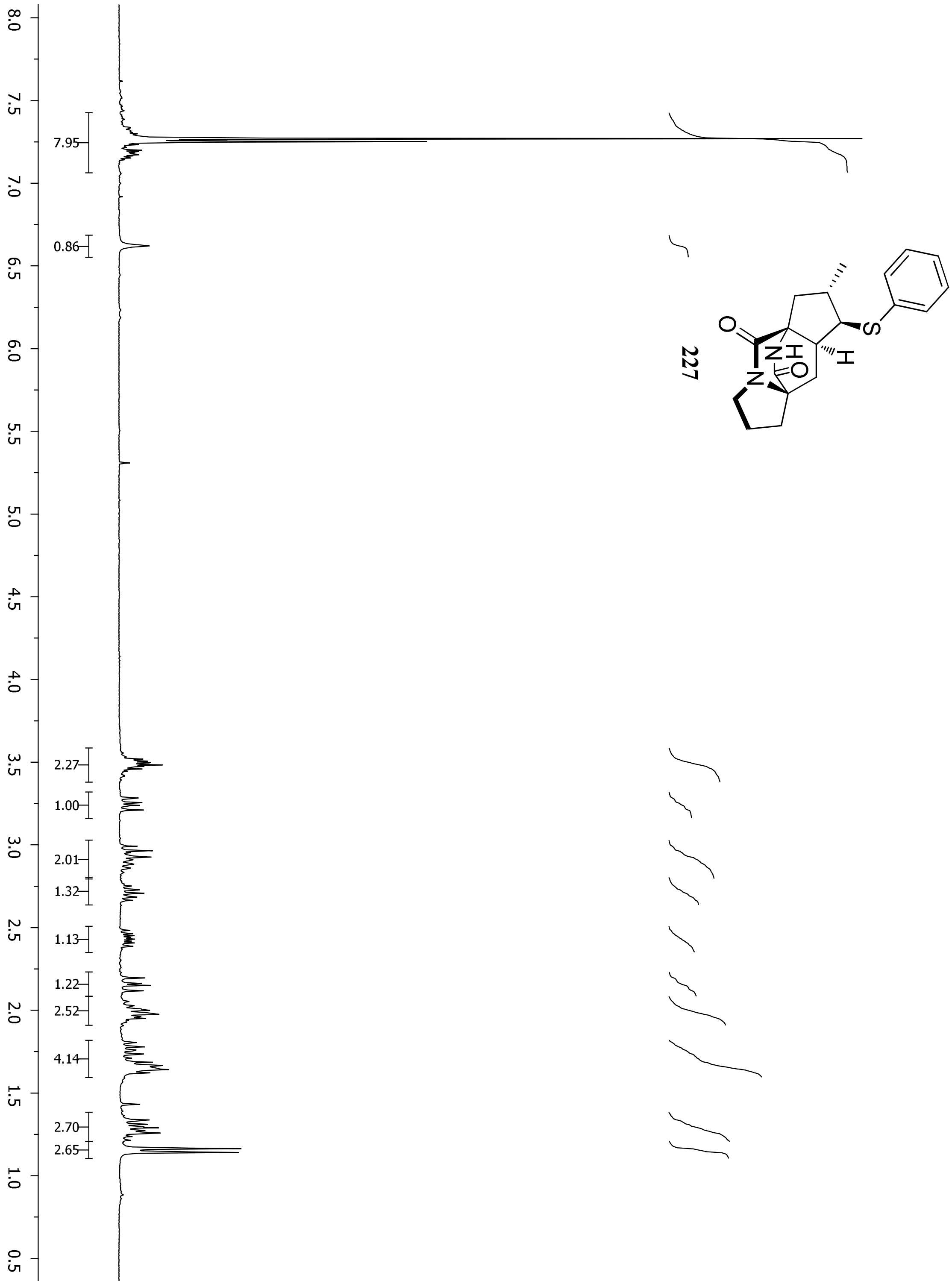
^{1}H NMR spectrum of bridged DKPs **210a** and **210b** (300 MHz, $CDCl_3$)



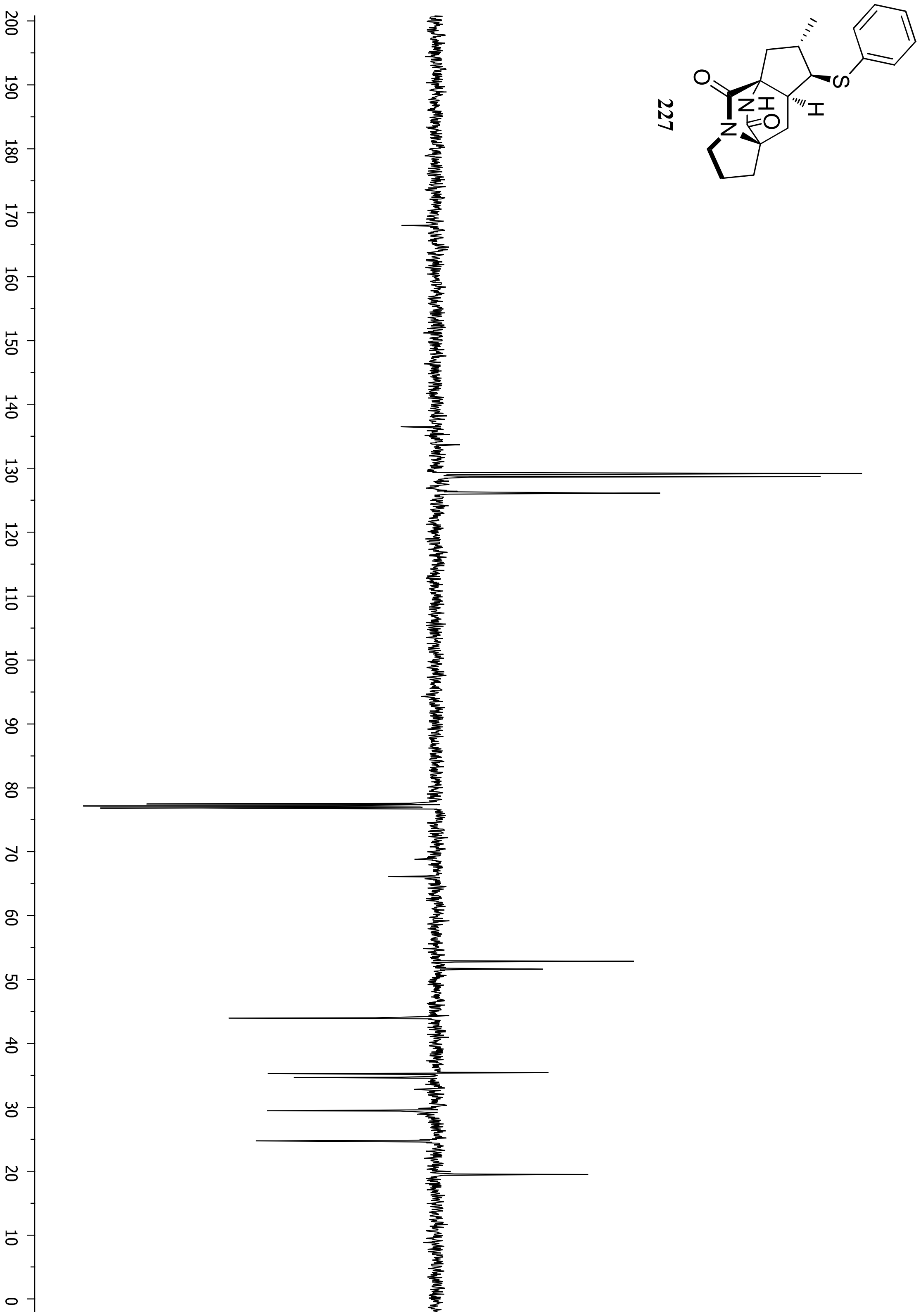
^{13}C NMR spectrum of bridged DKPs **210a** and **210b** (100 MHz, $CDCl_3$)



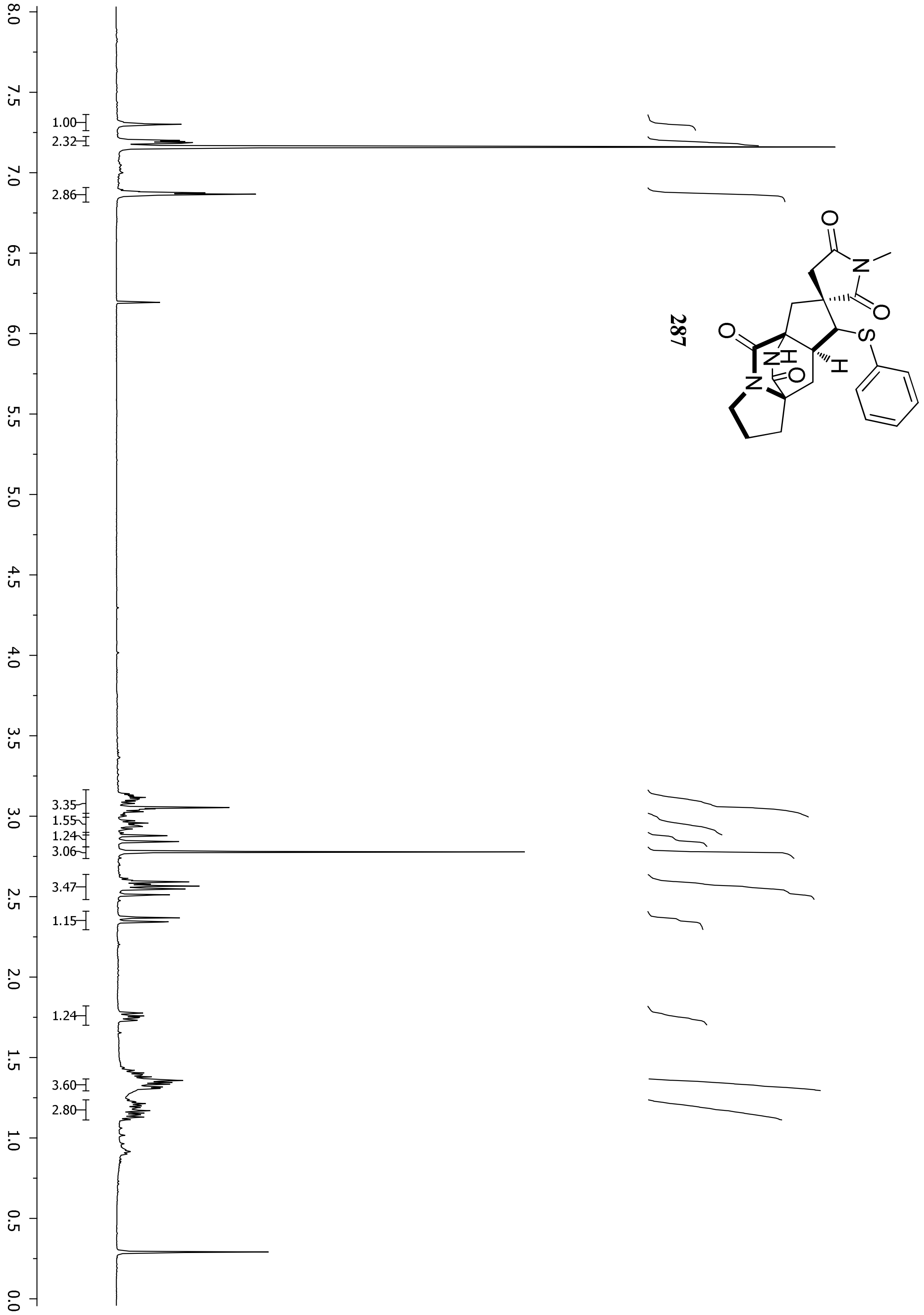
^{1}H NMR spectrum of bridged DKP **227** (300 MHz, $CDCl_3$)



^{13}C NMR spectrum of bridged DKP **227** (100 MHz, CDCl$_3$)

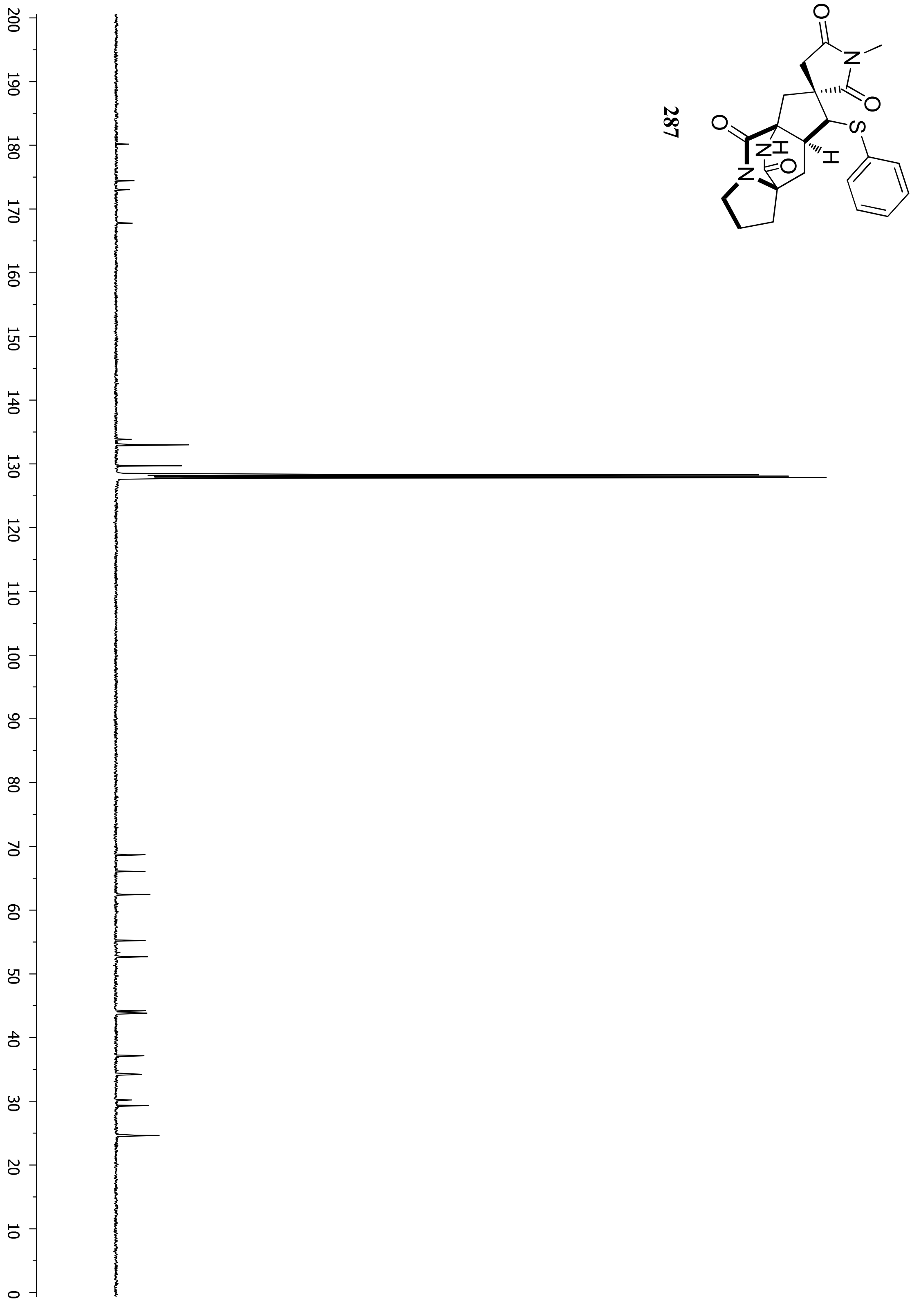


^{1}H NMR spectrum of bridged DKP **287** (500 MHz, CDCl$_3$)

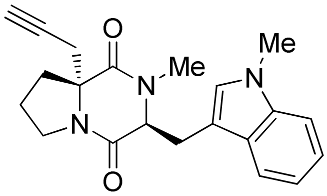


287

^{13}C NMR spectrum of bridged DKP **287** (100 MHz, CDCl$_3$)



Crystal Structure Data for DKP **203**



203

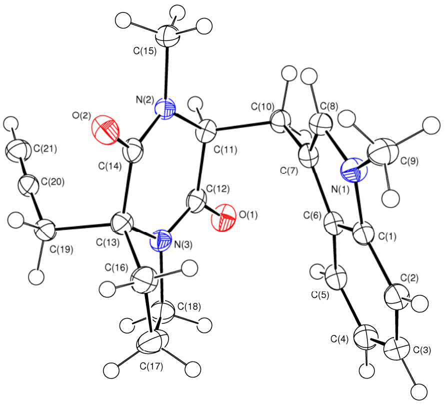


Table 1. Crystal data and structure refinement for 2009NS1.

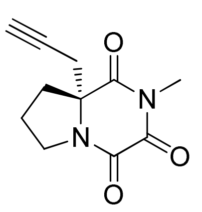
Identification code	2009NS1	
Empirical formula	C21 H23 N3 O2	
Formula weight	349.42	
Temperature	120(2) K	
Wavelength	1.54178 Å	
Crystal system	Orthorhombic	
Space group	P 2(1) 2(1) 2(1)	
Unit cell dimensions	a = 8.0642(4) Å	□= 90°.
	b = 9.4021(5) Å	□= 90°.
	c = 23.4940(12) Å	□ = 90°.
Volume	1781.32(16) Å^3	
Z	4	
Density (calculated)	1.303 Mg/m^3	
Absorption coefficient	0.680 mm^{-1}	
F(000)	744	
Crystal size	0.30 x 0.30 x 0.20 mm^3	
Theta range for data collection	6.66 to 70.02°.	
Index ranges	-8<=h<=9, -11<=k<=11, -28<=l<=28	
Reflections collected	15134	
Independent reflections	1940 [R(int) = 0.0347]	
Completeness to theta = 70.02°	99.0 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.8759 and 0.8219	
Refinement method	Full-matrix least-squares on F^2	
Data / restraints / parameters	1940 / 0 / 237	
Goodness-of-fit on F^2	1.066	
Final R indices [I>2sigma(I)]	R1 = 0.0256, wR2 = 0.0642	
R indices (all data)	R1 = 0.0262, wR2 = 0.0647	
Absolute structure parameter	?	
Largest diff. peak and hole	0.163 and -0.175 e.Å^{-3}	

Notes:
The hydrogen atoms were fixed as riding models.
The absolute structure could not be determined from the diffraction data as there are no heavy atoms present. However, it can be confirmed that the chirality of atom C(11) is opposite to that of atom C(13).

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3) for 2009NS1. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
C(1)	4096(2)	5259(2)	-119(1)	21(1)
C(2)	5169(2)	6270(2)	-345(1)	24(1)
C(3)	6749(2)	6337(2)	-116(1)	26(1)
C(4)	7250(2)	5424(2)	324(1)	25(1)
C(5)	6175(2)	4414(2)	540(1)	23(1)
C(6)	4569(2)	4307(2)	317(1)	20(1)
C(7)	3178(2)	3391(2)	425(1)	21(1)
C(8)	1953(2)	3841(2)	65(1)	22(1)
C(9)	1529(2)	5625(2)	-711(1)	27(1)
C(10)	3063(2)	2125(2)	811(1)	21(1)
C(11)	2604(2)	2462(2)	1439(1)	20(1)
C(12)	4072(2)	3197(2)	1712(1)	21(1)
C(13)	2356(2)	5368(2)	1855(1)	21(1)
C(14)	910(2)	4624(2)	1571(1)	20(1)
C(15)	-429(2)	2473(2)	1266(1)	24(1)
C(16)	2924(2)	6725(2)	1554(1)	29(1)
C(17)	4694(2)	6949(2)	1778(1)	36(1)
C(18)	5294(2)	5493(2)	1989(1)	29(1)
C(19)	1809(2)	5705(2)	2476(1)	21(1)
C(20)	1479(2)	4407(2)	2797(1)	23(1)
C(21)	1248(2)	3313(2)	3039(1)	28(1)
N(1)	2470(2)	4967(1)	-259(1)	23(1)
N(2)	1039(2)	3230(1)	1466(1)	19(1)
N(3)	3888(2)	4549(1)	1869(1)	22(1)
O(1)	5396(1)	2549(1)	1745(1)	29(1)
O(2)	-381(1)	5295(1)	1490(1)	28(1)

Crystal Structure Data for TKP **215**



215

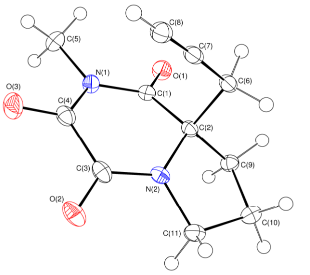


Table 1. Crystal data and structure refinement for 2008NS3.

Identification code	2008NS3	
Empirical formula	$C_{11} H_{12} N_2 O_3$	
Formula weight	220.23	
Temperature	120(2) K	
Wavelength	1.54178 Å	
Crystal system	Monoclinic	
Space group	P 2(1)	
Unit cell dimensions	a = 7.7088(1) Å	□= 90°.
	b = 8.0240(1) Å	□= 105.641(1)°.
	c = 8.5966(1) Å	□ = 90°.
Volume	512.055(11) $Å^3$	
Z	2	
Density (calculated)	1.428 Mg/m^3	
Absorption coefficient	0.881 mm^{-1}	
F(000)	232	
Crystal size	0.12 x 0.04 x 0.04 mm^3	
Theta range for data collection	6.85 to 70.04°.	
Index ranges	-9<=h<=9, -9<=k<=8, -10<=l<=10	
Reflections collected	4297	
Independent reflections	1685 [R(int) = 0.0168]	
Completeness to theta = 70.04°	98.2 %	
Absorption correction	Semi-empirical from equivalents	
Max. and min. transmission	0.9656 and 0.9017	
Refinement method	Full-matrix least-squares on F^2	
Data / restraints / parameters	1685 / 1 / 146	
Goodness-of-fit on F^2	1.059	
Final R indices [I>2sigma(I)]	R1 = 0.0261, wR2 = 0.0697	
R indices (all data)	R1 = 0.0263, wR2 = 0.0700	
Absolute structure parameter	0.06(16)	
Largest diff. peak and hole	0.157 and -0.193 e.$Å^{-3}$	

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters (Å^{2}x 10^3) for 2008NS3. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
C(1)	8282(2)	5695(2)	2143(2)	16(1)
C(2)	7269(2)	6237(2)	3349(2)	15(1)
C(3)	4835(2)	7322(2)	1077(2)	19(1)
C(4)	6206(2)	7347(2)	69(2)	19(1)
C(5)	8952(2)	6211(2)	-452(2)	25(1)
C(6)	8369(2)	7710(2)	4333(1)	17(1)
C(7)	8230(2)	9251(2)	3400(1)	18(1)
C(8)	8037(2)	10493(2)	2608(2)	21(1)
C(9)	7035(2)	4857(2)	4495(2)	17(1)
C(10)	5387(2)	5449(2)	5030(2)	21(1)
C(11)	4142(2)	6193(2)	3492(2)	19(1)
N(1)	7775(1)	6438(2)	625(1)	18(1)
N(2)	5410(1)	6725(1)	2573(1)	16(1)
O(1)	9576(1)	4784(1)	2542(1)	23(1)
O(2)	3280(1)	7759(1)	447(1)	25(1)
O(3)	5822(1)	8034(2)	-1233(1)	30(1)

Bibliography

[1] A. J. Birch, J. J. Wright, *J. Chem. Soc. D* **1969**, 644-645.

[2] A. J. Birch, J. J. Wright, *Tetrahedron* **1970**, *26*, 2329-2344.

[3] B. Witkop, *J. Am. Chem. Soc.* **1950**, *72*, 614-620.

[4] A. J. Birch, R. A. Russell, *Tetrahedron* **1972**, *28*, 2999-3008.

[5] G.-Y. Li, T. Yang, Y.-G. Luo, X.-Z. Chen, D.-M. Fang, G.-L. Zhang, *Org. Lett.* **2009**, *11*, 3714-3717.

[6] M. Yamazaki, E. Okuyama, M. Kobayashi, H. Inoue, *Tetrahedron Lett.* **1981**, *22*, 135-136.

[7] (a) J. G. Ondeyka, R. T. Goegelman, J. M. Schaeffer, L. Kelemen, L. Zitano, *J. Antibiot.* **1990**, *43*, 1375-1379. (b) J. M. Liesch, C. F. Wichmann, *J. Antibiot.* **1990**, *43*, 1380-1386. (c) S. E. Blanchflower, R. M. Banks, J. R. Everett, B. R. Manger, C. Reading, *J. Antibiot.* **1991**, *44*, 492-497. (d) S. E. Blanchflower, R. M. Banks, J. R. Everett, C. Reading, *J. Antibiot.* **1993**, *46*, 1355-1363.

[8] (a) B. H. Lee, M. F. Clothier, F. E. Dutton, S. J. Nelson, S. S. Johnson, D. P. Thompson, T. G. Geary, H. D. Whaley, C. L. Haber, V. P. Marshall, G. I. Kornis, P. L. McNally, J. I. Ciadella, D. G. Martin, J. W. Bowman, C. A. Baker, E. M. Coscarelli, S. J. Alexander-Bowman, J. P. Davis, E. W. Zinser, V. Wiley, M. F. Lipton, M. A. Mauragis, *Curr. Top. Med. Chem.* **2002**, *2*, 779-793. (b) M. P. Lopez-Gresa, M. C. Gonzalez, L. Ciavatta, I. Ayala, P. Moya, J. Primo, *J. Agric. Food Chem.* **2006**, *54*, 2921-2925. (c) Y. Ding, S. Gruschow, T. J. Greshock, J. M. Finefield, D. H. Sherman, R. M. Williams, *J. Nat. Prod.* **2008**, *71*, 1574-1578.

[9] J. Polonsky, M. A. Merrien, T. Prange, C. Pascard, S. Moreau, *J. Chem. Soc., Chem. Commun.* **1980**, 601-602.

[10] T. Prange, M. A. Billion, M. Vuilhorgne, C. Pascard, J. Polonsky, S. Moreau, *Tetrahedron Lett.* **1981**, *22*, 1977-1980.

[11] W. Fenical, P. R. Jensen, X. C. Cheng, U.S. Patent 6,066,635, **2000**.

[12] Y. Sugie, H. Hirai, T. Inagaki, M. Ishiguro, Y. J. Kim, Y. Kojima, T. Sakakibara, S. Sakemi, A. Sugiura, Y. Suzuki, L. Brennan, J. Duignan, L. H. Huang, J. Sutcliffe, N. Kojima, *J. Antibiot.* **2001**, *54*, 911-916.

[13] I. A. Grigor'ev, V. I. Mamatyuk, G. I. Shchukin, V. V. Martin, L. B. Volodarskii, *Khim. Geterotsikl. Soedin.* **1986**, 1065-1072.

[14] (a) J. F. Qian-Cutrone, K. D. Krampitz, Y.-Z. Shu, L.-P. Chang, S. E. Lowe, U.S. Patent 6,291,461, **2001**. (b) J. F. Qian-Cutrone, S. Huang, Y. Z. Shu, D. Vyas, C. Fairchild, A. Menendez, K. Krampitz, R. Dalterio, S. E. Klohr, Q. Gao, *J. Am. Chem. Soc.* **2002**, *124*, 14556-14557.

[15] (a) F. von Nussbaum, *Angew. Chem., Int. Ed.* **2003**, *42*, 3068-3071. (b) C. Escolano, *Angew. Chem., Int. Ed.* **2005**, *44*, 7670-7673.

[16] (a) S. Martinez-Luis, R. Rodriguez, L. Acevedo, M. C. Gonzalez, A. Lira-Rocha, R. Mata, *Tetrahedron* **2006**, *62*, 1817-1822. (b) M. Figueroa, C. G. M. Del, R. Mata, *Nat. Prod. Res.* **2008**, *22*, 709-714.

[17] H. Kato, T. Yoshida, T. Tokue, Y. Nojiri, H. Hirota, T. Ohta, R. M. Williams, S. Tsukamoto, *Angew. Chem., Int. Ed.* **2007**, *46*, 2254-2256.

[18] H. Hayashi, Y. Nishimoto, H. Nozaki, *Tetrahedron Lett.* **1997**, *38*, 5655-5658.

[19] R. M. Banks, S. E. Blanchflower, J. R. Everett, B. R. Manger, C. Reading, *J. Antibiot.* **1997**, *50*, 840-846.

[20] S. E. Blanchflower, R. M. Banks, J. R. Everett, B. R. Manger, C. Reading, *J. Antibiot.* **1991**, *44*, 492-497.

[21] H. Hayashi, Y. Nishimoto, K. Akiyama, H. Nozaki, *Biosci., Biotechnol., Biochem.* **2000**, *64*, 111-115.

[22] C. R. Gray, J. F. Sanz-Cervera, L. A. Silks, R. M. Williams, *J. Am. Chem. Soc.* **2003**, *125*, 14692-14693.

[23] A. E. A. Porter, P. G. Sammes, *J. Chem. Soc. D* **1970**, 1103.

[24] P. J. Machin, A. E. A. Porter, P. G. Sammes, *J. Chem. Soc., Perkin Trans. 1* **1973**, 404-409.

[25] J. Baldas, A. J. Birch, R. A. Russell, *J. Chem. Soc., Perkin Trans. 1* **1974**, 50-52.

[26] J. F. Sanz-Cervera, T. Glinka, R. M. Williams, *Tetrahedron* **1993**, *49*, 8471-8482.

[27] R. M. Williams, *J. Org. Chem.* **2011**, *76*, 4221-4259.

[28] (a) L. R. Domingo, J. F. Sanz-Cervera, R. M. Williams, M. T. Picher, J. A. Marco, *J. Org. Chem.* **1997**, *62*, 1662-1667. (b) R. M. Williams, J. F. Sanz-Cervera, F. Sancenon, J. A. Marco, K. M. Halligan, *Bioorg. Med. Chem.* **1998**, *6*, 1233-1241.

[29] E. M. Stocking, R. M. Williams, *Angew. Chem., Int. Ed.* **2003**, *42*, 3078-3115.

[30] (a) D. Hilvert, K. W. Hill, K. D. Nared, M. T. M. Auditor, *J. Am. Chem. Soc.* **1989**, *111*, 9261-9262. (b) A. C. Braisted, P. G. Schultz, *J. Am. Chem. Soc.* **1990**, *112*, 7430-7431.

[31] (a) T. M. Tarasow, B. E. Eaton, *Cell. Mol. Life Sci.* **1999**, *55*, 1463-1472. (b) B. Seelig, A. Jaschke, *Chem. Biol.* **1999**, *6*, 167-176. (c) B. Seelig, S. Keiper, F. Stuhlmann, A. Jaschke, *Angew. Chem., Int. Ed.* **2000**, *39*, 4576-4579.

[32] (a) K. Auclair, A. Sutherland, J. Kennedy, D. J. Witter, d. H. J. P. Van, C. R. Hutchinson, J. C. Vederas, *J. Am. Chem. Soc.* **2000**, *122*, 11519-11520. (b) K. Watanabe, T. Mie, A. Ichihara, H. Oikawa, M. Honma, *J. Biol. Chem.* **2000**, *275*, 38393-38401.

[33] E. M. Stocking, J. F. Sanz-Cervera, R. M. Williams, C. J. Unkefer, *J. Am. Chem. Soc.* **1996**, *118*, 7008-7009.

[34] S.-M. Li, *Nat. Prod. Rep.* **2010**, *27*, 57-78.

[35] W. L. Shoop, J. R. Egerton, C. H. Eary, D. Suhayda, *J. Parasitol.* **1990**, *76*, 349-351.

[36] W. L. Shoop, H. W. Haines, C. H. Eary, B. F. Michael, *Am. J. Vet. Res.* **1992**, *53*, 2032-2034.

[37] J. H. Gill, E. Lacey, *Int. J. Parasitol.* **1993**, *23*, 375-381.

[38] B. H. Lee, M. F. Clothier, *Tetrahedron Lett.* **1997**, *38*, 4009-4012.

[39] G. A. Conder, S. S. Johnson, P. M. Guimond, D. L. Cox, B. L. Lee, *J. Parasitol.* **1991**, *77*, 621-623.

[40] T. A. Blizzard, H. Mrozik, M. H. Fisher, J. M. Schaeffer, *J. Org. Chem.* **1990**, *55*, 2256-2259.

[41] B. H. Lee, M. F. Clothier, S. S. Johnson, *Bioorg. Med. Chem. Lett.* **2001**, *11*, 553-554.

[42] J. E. Wulff, S. B. Herzon, R. Siegrist, A. G. Myers, *J. Am. Chem. Soc.* **2007**, *129*, 4898-4899.

[43] K. A. Miller, M. Figueroa, M. W. N. Valente, T. J. Greshock, R. Mata, R. M. Williams, *Bioorg. Med. Chem. Lett.* **2008**, *18*, 6479-6481.

[44] K. A. Miller, R. M. Williams, *Chem. Soc. Rev.* **2009**, *38*, 3160-3174.

[45] R. M. Williams, T. Glinka, E. Kwast, *J. Am. Chem. Soc.* **1988**, *110*, 5927-5929.

[46] T. Kametani, N. Kanaya, M. Ihara, *J. Am. Chem. Soc.* **1980**, *102*, 3974-3975.

[47] R. M. Williams, J. Cao, H. Tsujishima, R. J. Cox, *J. Am. Chem. Soc.* **2003**, *125*, 12172-12178.

[48] T. D. Cushing, J. F. Sanz-Cervera, R. M. Williams, *J. Am. Chem. Soc.* **1993**, *115*, 9323-9324.

[49] G. D. Artman, III, A. W. Grubbs, R. M. Williams, *J. Am. Chem. Soc.* **2007**, *129*, 6336-6342.

[50] R. M. Williams, J. F. Sanz-Cervera, F. Sancenon, J. A. Marco, K. Halligan, *J. Am. Chem. Soc.* **1998**, *120*, 1090-1091.

[51] T. Kametani, N. Kanaya, M. Ihara, *J. Chem. Soc., Perkin Trans. 1* **1981**, 959-963.

[52] S. Jin, P. Wessig, J. Liebscher, *J. Org. Chem.* **2001**, *66*, 3984-3997.

[53] J. F. Sanz-Cervera, R. M. Williams, *J. Am. Chem. Soc.* **2002**, *124*, 2556-2559.

[54] T. J. Greshock, A. W. Grubbs, S. Tsukamoto, R. M. Williams, *Angew. Chem., Int. Ed.* **2007**, *46*, 2262-2265.

[55] T. J. Greshock, A. W. Grubbs, R. M. Williams, *Tetrahedron* **2007**, *63*, 6124-6130.

[56] K. A. Miller, T. R. Welch, T. J. Greshock, Y. Ding, D. H. Sherman, R. M. Williams, *J. Org. Chem.* **2008**, *73*, 3116-3119.

[57] (a) P. S. Baran, C. A. Guerrero, B. D. Hafensteiner, N. B. Ambhaikar, *Angew. Chem., Int. Ed.* **2005**, *44*, 3892-3895. (b) P. S. Baran, C. A. Guerrero, N. B. Ambhaikar, B. D. Hafensteiner, *Angew. Chem., Int. Ed.* **2005**, *44*, 606-609. (c) P. S. Baran, B. D. Hafensteiner, N. B. Ambhaikar, C. A. Guerrero, J. D. Gallagher, *J. Am. Chem. Soc.* **2006**, *128*, 8678-8693.

[58] S. B. Herzon, A. G. Myers, *J. Am. Chem. Soc.* **2005**, *127*, 5342-5344.

[59] L. A. K. Nelson, A. C. Shaw, S. R. Abrams, *Tetrahedron* **1991**, *47*, 3259-3270.

[60] M. G. Banwell, B. D. Kelly, O. J. Kokas, D. W. Lupton, *Org. Lett.* **2003**, *5*, 2497-2500.

[61] (a) A. Elomri, S. Michel, F. Tillequin, M. Koch, *Heterocycles* **1992**, *34*, 799-806. (b) R. J. Cox, R. M. Williams, *Tetrahedron Lett.* **2002**, *43*, 2149-2152.

[62] B. M. Trost, N. Cramer, H. Bernsmann, *J. Am. Chem. Soc.* **2007**, *129*, 3086-3087.

[63] B. M. Savall, W. W. McWhorter, E. A. Walker, *J. Org. Chem.* **1996**, *61*, 8696-8697.

[64] B. M. Trost, S. M. Mignani, T. N. Nanninga, *J. Am. Chem. Soc.* **1986**, *108*, 6051-6053.

[65] R. M. Williams, T. D. Cushing, *Tetrahedron Lett.* **1990**, *31*, 6325-6328.

[66] (a) F. Frebault, N. S. Simpkins, A. Fenwick, *J. Am. Chem. Soc.* **2009**, *131*, 4214-4215. (b) F. C. Frebault, N. S. Simpkins, *Tetrahedron* **2010**, *66*, 6585-6596.

[67] T. Hara, S. R. Durell, M. C. Myers, D. H. Appella, *J. Am. Chem. Soc.* **2006**, *128*, 1995-2004.

[68] F. Gonzalez, J. F. Sanz-Cervera, R. M. Williams, *Tetrahedron Lett.* **1999**, *40*, 4519-4522.

[69] S. Tanimori, K. Fukubayashi, M. Kirihata, *Biosci., Biotechnol., Biochem.* **2000**, *64*, 1758-1760.

[70] (a) S. Tanimori, K. Fukubayashi, M. Kirihata, *Tetrahedron Lett.* **2001**, *42*, 4013-4016. (b) S. Tanimori, T. Sunami, K. Fukubayashi, M. Kirihata, *Tetrahedron* **2005**, *61*, 2481-2492.

[71] J. Robertson, J. Pillai, R. K. Lush, *Chem. Soc. Rev.* **2001**, *30*, 94-103.

[72] D. Lathbury, P. J. Parsons, I. Pinto, *J. Chem. Soc., Chem. Commun.* **1988**, 81-82.

[73] D. P. Curran, D. Kim, H. T. Liu, W. Shen, *J. Am. Chem. Soc.* **1988**, *110*, 5900-5902.

[74] J. Robertson, M. A. Peplow, H. Pillai, *Tetrahedron Lett.* **1996**, *37*, 5825-5828.

[75] C. D. S. Brown, N. S. Simpkins, K. Clinch, *Tetrahedron Lett.* **1993**, *34*, 131-132.

[76] T. Gimisis, C. Chatgilialoglu, *J. Org. Chem.* **1996**, *61*, 1908-1909.

[77] B. Viskolcz, G. Lendvay, T. Kortvelyesi, L. Seres, *J. Am. Chem. Soc.* **1996**, *118*, 3006-3009.

[78] X. L. Huang, J. J. Dannenberg, *J. Org. Chem.* **1991**, *56*, 5421-5424.

[79] R. Breslow, R. J. Corcoran, B. B. Snider, *J. Am. Chem. Soc.* **1974**, *96*, 6791-6792.

[80] E. Bosch, M. D. Bachi, *J. Org. Chem.* **1993**, *58*, 5581-5582.

[81] S. Z. Zard, *Radical Reactions in Organic Synthesis*, Oxford University Press, Oxford, **2003**.

[82] F. M. Welle, H.-D. Beckhaus, C. Ruechardt, *J. Org. Chem.* **1997**, *62*, 552-558.

[83] (a) D. J. Pasto, *J. Am. Chem. Soc.* **1988**, *110*, 8164-8175. (b) G. Leroy, M. Sana, C. Wilante, *J. Mol. Struct.* **1991**, *80*, 303-328.

[84] (a) D. Seebach, M. Boes, R. Naef, W. B. Schweizer, *J. Am. Chem. Soc.* **1983**, *105*, 5390-5398. (b) D. Seebach, A. R. Sting, M. Hoffmann, *Angew. Chem., Int. Ed.* **1997**, *35*, 2708-2748.

[85] H. Wang, J. P. Germanas, *Synlett* **1999**, 33-36.

[86] M. Amedjkouh, P. Ahlberg, *Tetrahedron: Asymmetry* **2002**, *13*, 2229-2234.

[87] F. Pisaneschi, F. M. Cordero, M. Lumini, A. Brandi, *Synlett* **2007**, 2882-2884.

[88] P. W. R. Harris, M. A. Brimble, V. J. Muir, M. Y. H. Lai, N. S. Trotter, D. J. Callis, *Tetrahedron* **2005**, *61*, 10018-10035.

[89] S. D. Burke, K. W. Jung, *Tetrahedron Lett.* **1994**, *35*, 5837-5840.

[90] S. D. Burke, K. W. Jung, R. E. Perri, *Tetrahedron Lett.* **1994**, *35*, 5841-5844.

[91] (a) F. Beaufils, F. Denes, P. Renaud, *Org. Lett.* **2004**, *6*, 2563-2566. (b) F. Denes, F. Beaufils, P. Renaud, *Org. Lett.* **2007**, *9*, 4375-4378.

[92] F. Beaufils, F. Denes, P. Renaud, *Angew. Chem., Int. Ed.* **2005**, *44*, 5273-5275.

[93] R. M. Williams, E. Kwast, *Tetrahedron Lett.* **1989**, *30*, 451-454.

[94] L. A. Carpino, *J. Am. Chem. Soc.* **1993**, *115*, 4397-4398.

[95] D. J. C. Pappin, S. Pillai, J. M. Coull, U.S. Patent 7,307,169, **2005**.

[96] M. Pichowicz, N. S. Simpkins, A. J. Blake, C. Wilson, *Tetrahedron* **2008**, *64*, 3713-3735.

[97] (a) E. M. Stocking, J. F. Sanz-Cervera, R. M. Williams, *J. Am. Chem. Soc.* **2000**, *122*, 1675-1683. (b) L. A. Adams, M. W. N. Valente, R. M. Williams, *Tetrahedron* **2006**, *62*, 5195-5200. (c) T. J. Greshock, R. M. Williams, *Org. Lett.* **2007**, *9*, 4255-4258.

[98] K. C. Majumdar, P. Debnath, *Tetrahedron* **2008**, *64*, 9799-9820.

[99] G. E. Keck, T. T. Wager, J. F. D. Rodriquez, *J. Am. Chem. Soc.* **1999**, *121*, 5176-5190.

[100] (a) P. D. Bailey, A. N. Boa, S. R. Baker, J. Clayson, E. J. Murray, G. M. Rosair, *Tetrahedron Lett.* **1999**, *40*, 7557-7560. (b) P. D. Bailey, N. Bannister, M. Bernad, S. Blanchard, A. N. Boa, *J. Chem. Soc., Perkin Trans. 1* **2001**, 3245-3251.

[101] B. Lygo, B. I. Andrews, *Acc. Chem. Res.* **2004**, *37*, 518-525.

[102] (a) E. J. Corey, F. Xu, M. C. Noe, *J. Am. Chem. Soc.* **1997**, *119*, 12414-12415. (b) E. J. Corey, M. C. Noe, F. Xu, *Tetrahedron Lett.* **1998**, *39*, 5347-5350.

[103] B. R. Lichtenstein, J. F. Cerda, R. L. Koder, P. L. Dutton, *Chem. Commun.* **2009**, 168-170.

[104] C. A. G. N. Montalbetti, V. Falque, *Tetrahedron* **2005**, *61*, 10827-10852.

[105] (a) M. Ito, T. Koyama, K. Ogura, *Chem. Lett.* **1986**, 101-104. (b) P. A. Jacobi, Y. Li, *Org. Lett.* **2003**, *5*, 701-704.

[106] J. B. P. A. Wijnberg, P. G. Wiering, H. Steinberg, *Synthesis* **1981**, 901-903.

[107] D. H. R. Barton, Y. Herve, P. Potier, J. Thierry, *Tetrahedron* **1987**, *43*, 4297-4308.

[108] N. Subasinghe, M. Schulte, R. J. Roon, J. F. Koerner, R. L. Johnson, *J. Med. Chem.* **1992**, *35*, 4602-4607.

[109] E. Marcantoni, M. Massaccesi, E. Torregiani, G. Bartoli, M. Bosco, L. Sambri, *J. Org. Chem.* **2001**, *66*, 4430-4432.

[110] Y.-q. Wu, D. C. Limburg, D. E. Wilkinson, M. J. Vaal, G. S. Hamilton, *Tetrahedron Lett.* **2000**, *41*, 2847-2849.

[111] J. S. Yadav, B. V. S. Reddy, K. S. Rao, K. Harikishan, *Synlett* **2002**, 826-828.

[112] C. G. H. White, A. B. Tabor, *Tetrahedron* **2007**, *63*, 6932-6937.

[113] Y. Igarashi, S. Watanabe, *Nippon Kagaku Kaishi* **1992**, 1392-1396.

[114] K. C. Nicolaou, A. A. Estrada, M. Zak, S. H. Lee, B. S. Safina, *Angew. Chem., Int. Ed.* **2005**, *44*, 1378-1382.

[115] HRMS (ESI) calculated for $C_{11}H_{19}NO_2Br$ 276.0599 found 276.0605

[116] Z. Z. Brown, C. E. Schafmeister, *Org. Lett.* **2010**, *12*, 1436-1439.

[117] G. D. Artman, III, R. J. Rafferty, R. M. Williams, *Org. Synth.* **2009**, *86*, 262-273.

[118] T. Shinada, H. Yoshida, Y. Ohfune, *Synthesis* **2009**, 3751-3756.