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PERSPECTIVE

The synthesis of dehydrotryptophan and dehydrotryptophan-containing peptides

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Dehydrotryptophan and its derivatives are non-proteinogenic amino acids commonly found in peptide-based natural products produced by microorganisms, marine organisms and plants. These non-proteinogenic amino acids are found in secondary metabolites and are formed by post translational modification processes. Although comprehensive reviews on the synthesis of dehydroamino acids are available, this perspective focuses solely on methods to synthesise the dehydrotryptophan-containing segment of naturally occurring peptides, amino acids and their derivatives.

1. Introduction

α,β-Dehydroamino acids are non-proteinogenic residues often found in natural products produced by microorganisms, marine organisms and plants.[1] Dehydroamino acids usually exist as the *Z*-isomer as it is thermodynamically more stable than the *E*-isomer.[1–4] The *E*- and *Z*-isomers of certain dehydroamino acids, such as dehydrophenylalanine and dehydrotryptophan, are able to interconvert photochemically, either when incorporated into peptides or as part of smaller derivatives.[4,5] The *E*/*Z* configuration of the double bond affects the conformation of dehydroamino acid containing peptides, and hence the biological activity of these compounds.[5]

Dehydroamino acid residues reduce the conformational flexibility of peptides, a property that is useful for structure–activity relationship studies and the design of secondary structures in peptides.[6–8] For example, dehydroalanine rigidifies the peptide backbone into extended conformations. In contrast dehydrophenylalanine induces β-turns in short peptides and promotes longer peptides to adopt helical conformations.[2,9] This property is due to both the formation of a cross-conjugated system and the influence on the φ and ψ torsion angles (Fig. 1). Other dehydroamino acid residues also induce peptides and proteins to adopt highly ordered secondary structures such as helices, β sheets and γ turns.[2]

Importantly the presence of dehydroamino acids in peptides can also confer resistance to enzymatic degradation and alter bioactivity.[10,11] Thus, the development of pharmaceutical compounds wherein dehydroamino acids are incorporated into analogues of biologically active peptides is a valuable strategy to enhance the potency and improve the stability of a bioactive peptide.[11]

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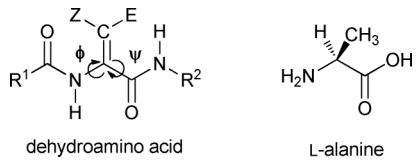


Fig. 1 Dehydroamino acids can exist in either *Z* or *E* conformations with modified φ and ψ angles compared to saturated amino acids.[2]

In the last two decades comprehensive reviews of the synthesis of dehydroamino acids have been published by Blaskovich (2010), Hughes (2009), Bonauer (2006), Humphrey (1997) and Schmidt (1988).[12–16] Due to scant attention in these reviews to the synthesis of the dehydroamino acid α,β-dehydrotryptophan, wherein a double bond exists between C^{α} and C^{β} of the natural tryptophan residue, this review focuses exclusively on the synthesis of the dehydrotryptophan-containing segment of naturally occurring peptides, amino acids and their derivatives.

Dehydrotryptophan and derivatives thereof occur naturally in several peptides, all of which are cyclic (Fig. 2). The dehydrotryptophan residue is synthesised biosynthetically by the enzymatic oxidation of tryptophan residues by tryptophan side chain α,β oxidase from *Pseudomonas* or L-tryptophan 2′,3′-oxidase from *Chromobacterium violaceum*.[17–21] L-Tryptophan 2′,3′-oxidase dehydrogenates tryptophan directly in a *syn* specific manner to give *Z*-dehydrotryptophan whereas tryptophan side chain α,β oxidase forms an indolyloxazoline intermediate, which undergoes isomerisation resulting in a mixture of *E* and *Z* dehydrotryptophan.[17–22]

2. Synthesis of dehydrotryptophan and derivatives

2.1 Erlenmeyer synthesis

The Erlenmeyer reaction is a well-known procedure for the synthesis of amino acids and dehydroamino acids from *N*-protected glycine derivatives that proceeds *via* an azlactone intermediate

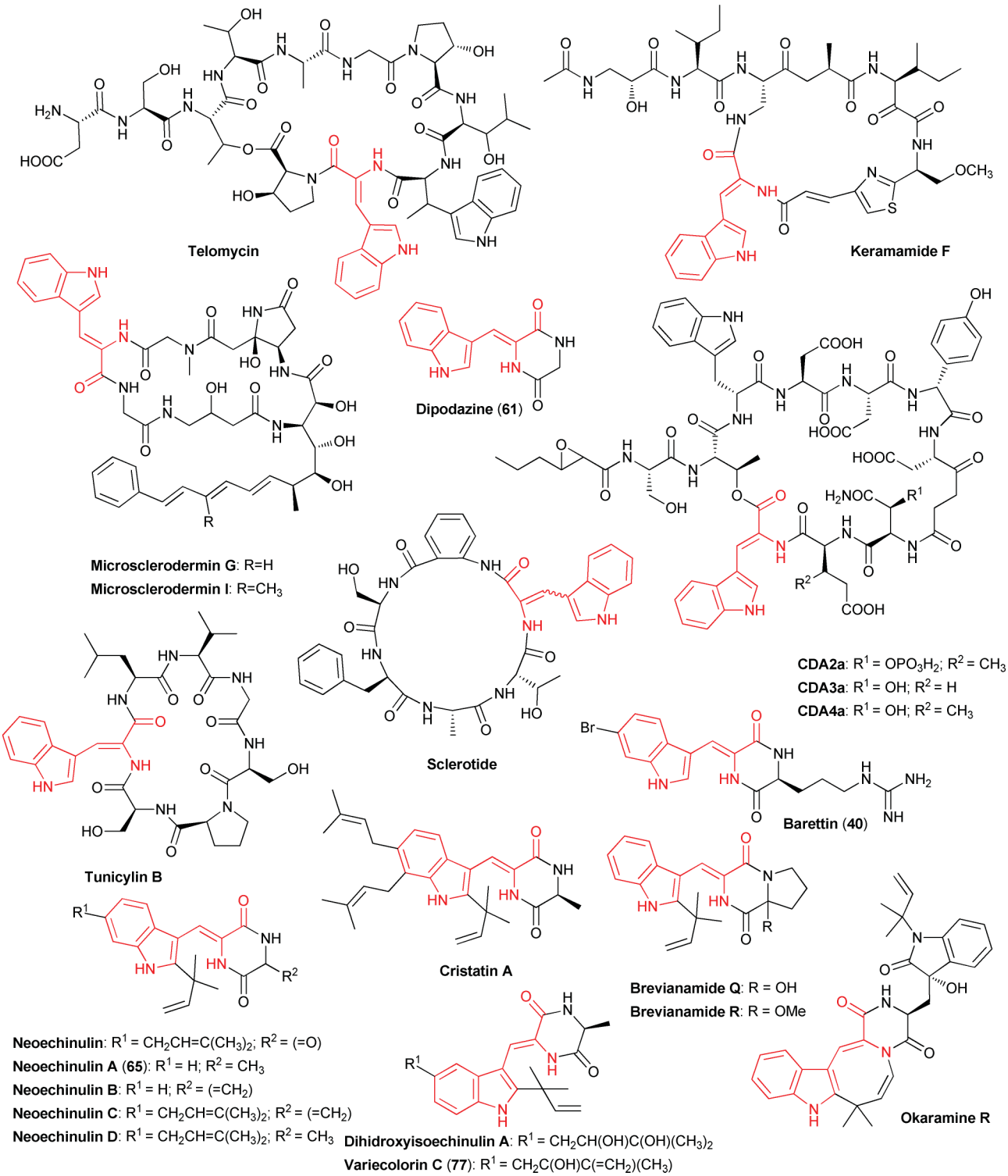
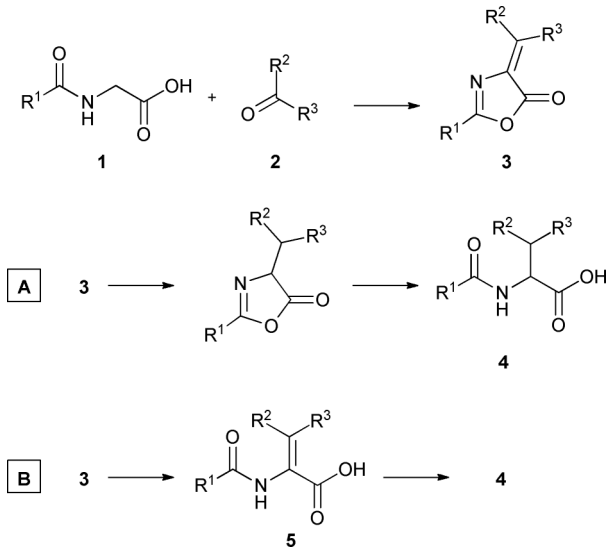


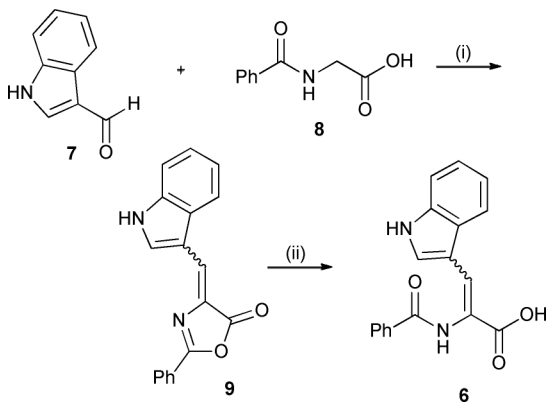
Fig. 2 Dehydrotryptophan and its derivatives in naturally occurring peptides.

(Scheme 1).[14,23] Condensation of glycine derivative **1** with aldehyde or ketone **2** affords azlactone **3** which upon reduction of the double bond (Scheme 1A) followed by ring opening yields amino acids of structure **4**.[23] In contrast, hydrolysis of azlactone **3** initially affords the dehydroamino acid **5** (Scheme 1B) from which the natural amino acid can be obtained by subsequent hydrogenation.[23]

The amino acid tryptophan was first synthesised by Erlenmeyer's method over a century ago *via* the dehydrotryptophan intermediate **6** (Scheme 2).[23] Ellinger *et al.* reacted indole-3-carbaldehyde **7** with the glycine derivative hippuric acid **8**, after which hydrolysis of the azlactone intermediate **9** afforded the dehydrotryptophan intermediate **6**.[23]

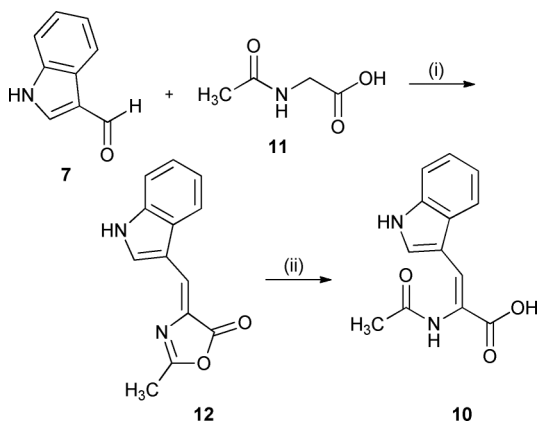


Scheme 1 Erlenmeyer synthesis of amino acids.



Scheme 2 Synthesis of dehydrotryptophan intermediate **6** *en route* to the synthesis of tryptophan. i) Ac_2O, NaOAc. ii) NaOH, H_2O.

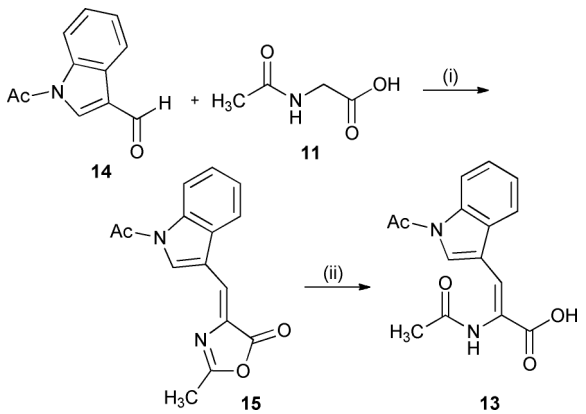
Although Erlenmeyer's azlactone method has been used to synthesise several dehydrotryptophan derivatives, variable yields are obtained.[24–28] *N*-Acetyl dehydrotryptophan **10** (Scheme 3) was prepared in good yield by Oba *et al.* by reacting indole-3-carbaldehyde **7** with *N*-acetyl glycine **11** and heating to 120 °C.[24]



Scheme 3 Synthesis of *N*-acetyl dehydrotryptophan **10**. i) NaOAc (1 eq), Ac_2O (4 eq), 120 °C, 2–3 h. ii) 0.2 M Na_2CO_3, 4 h.

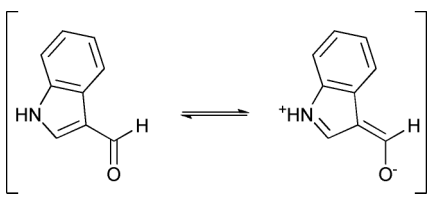
Hydrolysis of the resultant crude azlactone **12** using aqueous Na_2CO_3 afforded the desired product **10** in 77% yield.[24]

In contrast, low yields were obtained by both Kirby *et al.*[25] and Skrabal *et al.*[26] for the synthesis of indole-protected *N*-acetyl dehydrotryptophan **13** (Scheme 4). Condensation of *N*-acetyl-3-formylindole **14** with *N*-acetylglycine **11** followed by hydrolysis of the intermediate azlactone **15** afforded the dehydrotryptophan derivative **13** in 20%[25] and 26%[26] yield, respectively.



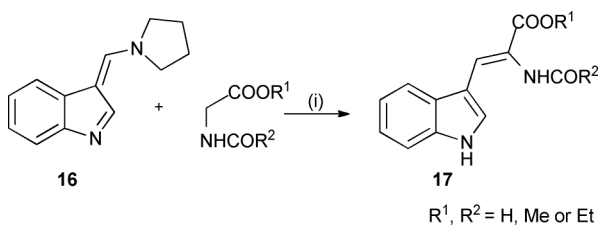
Scheme 4 Synthesis of *N*-acetyl-dehydrotryptophan derivative **13**. i) Ac_2O, K_2CO_3, 100 °C, 1 h. ii) acetone : H_2O (2 : 1), reflux, 24 h; K_2CO_3, H_2O, 20 h.

It has been postulated that the low yields obtained for the synthesis of dehydrotryptophan derivatives using Erlenmeyer's synthesis is due to the low reactivity of the contributing resonance structures of the indole-aldehyde (Scheme 5).[29]



Scheme 5 Resonance structures of indole-3-carbaldehyde.

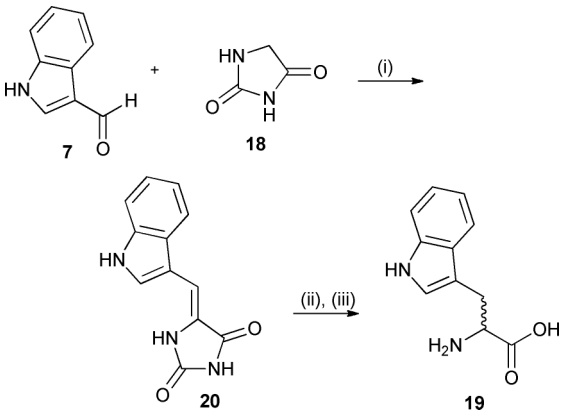
As an alternative strategy 3-[(1-pyrrolidinyl)methylene]-3*H*-indole **16** was used by Moriya *et al.* as a 1,4-dipolar synthon of indole-3-carbaldehyde, from which *N*-acyl-α,β-dehydrotryptophan esters **17** were synthesised (Scheme 6).[29] This modification improved the yield of dehydrotryptophan derivatives to 50–70%.[29]



Scheme 6 Synthesis of *N*-acyl-α,β-dehydrotryptophan esters **17**. i) DMF : MeOH (10 : 1), 110 °C, 6 h.

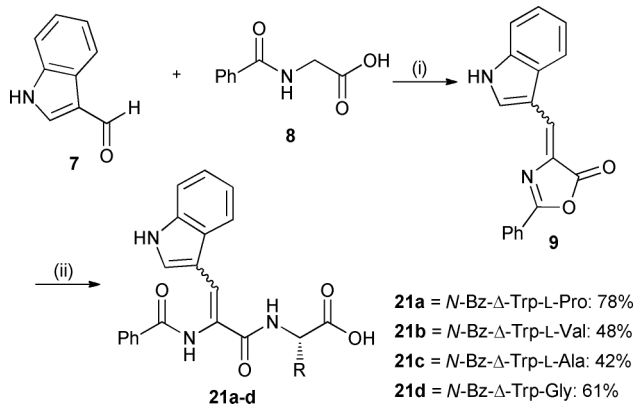
Another alternative to Erlenmeyer's original procedure involved the use of hydantoin **18** as a glycine equivalent for the synthesis of tryptophan **19**. The cyclic dehydrotryptophan hydantoin

intermediate **20** was obtained in 95% yield by heating indole-3-carboxaldehyde **7** with hydantoin **18** under reflux (Scheme 7).[30]



Scheme 7 Synthesis of tryptophan **19**. i) piperidine, 150 °C, 20 min, 95%. ii) RANEY® nickel, H_2, NaOH, 24 h, 71%. iii) $Ba(OH)_2$, H_2O, reflux, 40 h, 88%.

N-Benzoyl protected dehydrotryptophan dipeptides **21** have been directly accessed using Erlenmeyer's procedure, whereby condensation of indole-3-carbaldehyde **7** with hippuric acid **8** afforded the azlactone intermediate **9**, after which azlactone hydrolysis and concomitant condensation with a second amino acid enabled peptide bond formation (Scheme 8).[31,32]

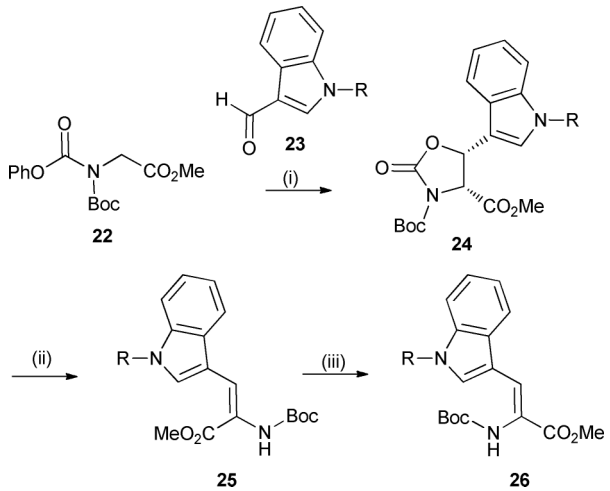


Scheme 8 Synthesis of *N*-Bz-Δ-Trp-amino acids **21**. i) Ac_2O, $KHCO_3$, rt, 24 h. ii) amino acid, acetone, NaOH (1 N), 20 °C, several h.

In a similar manner to the above dehydrotryptophan dipeptide synthesis, this reaction has been carried out in the absence of base. Azlactone **9** was opened with a small molecule primary amine, resulting in the formation of an amide bond at the *C*-terminus of a dehydrotryptophan derivative.[33]

2.2 *Via* an oxazolidinone derivative

Dehydrotryptophan amino acid derivatives have been prepared *via* oxazolidinone intermediates (Scheme 9).[34] Condensation of glycine derivative **22** with indole-3-carbaldehyde **23** formed oxazolidione intermediate **24**, after which treatment with base afforded dehydrotryptophan derivative **25** as an isomeric mixture composed predominantly of the *E* isomer (Scheme 9).[34] It was then found that the *E* isomer **25** could be isomerised to the *Z* isomer **26** using catalytic quantities of iodine.[34]

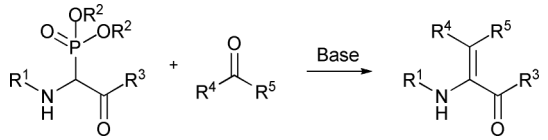


Scheme 9 Synthesis of *E*-dehydrotryptophan derivative **25** and isomerisation to *Z*-dehydrotryptophan derivative **26**. i) LDA (R = Cbz) or LiHMDS (R = Boc), THF, −78 °C, 1 h then **23**, $Ti(O^iPr)_4$, −78 °C, 4 h, R = Cbz (66%), Boc (41%). ii) LiHMDS, THF, R = Cbz (−10 °C, 10 min, 100%, 93 : 7 *E* : *Z*), R = Boc (−78 °C, 5 min, 90%, 41 : 4 *E* : *Z*). iii) catalytic I_2, THF, rt, R = Cbz (24 h, 97%, 1 : 24 *E* : *Z*), Boc (43 h, 93%, 9 : 91 *E* : *Z*).

2.3 Olefination reactions

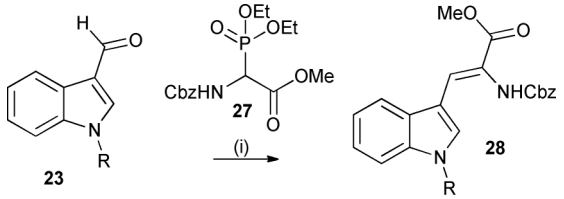
The Horner–Wadsworth–Emmons and Wittig type olefination reactions are popular methods for the preparation of dehydrotryptophan containing amino acids and peptide derivatives.

2.3.1 Horner–Wadsworth–Emmons olefination. The Horner–Wadsworth–Emmons (HWE) olefination reaction is a frequently used synthetic method for the preparation of dehydrotryptophan derivatives and can be generalized by the following transformation (Scheme 10) in which a phosphonate, is treated with a base and reacted with a ketone or aldehyde resulting in the formation of a new carbon–carbon double bond.[35–48]



Scheme 10 The HWE reaction used to prepare dehydroamino acid derivatives.

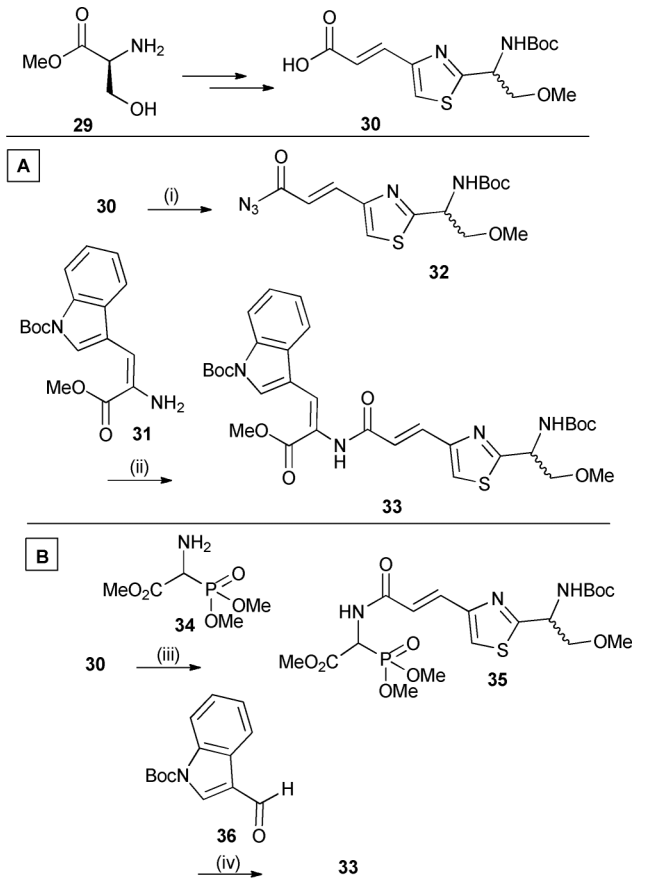
This method was used by Shin *et al.* to synthesise dehydrotryptophan derivatives **28** by reacting indole protected aldehyde **23** with *N*-Cbz-(diethoxyphosphinyl)glycine methyl ester **27** (Scheme 11).[49]



Scheme 11 Synthesis of dehydrotryptophan derivatives **28** using the HWE olefination reaction. i) KO^tBu, CH_2Cl_2, −70 °C to rt, 3 h.

2.3.1.1 Use of the HWE olefination towards the total synthesis of keramamide F. Keramamide F (Fig. 2), is a cytotoxic natural product isolated in 1992 from a *Theonella* sponge by Itagaki *et al.*[50] The presence of the dehydrotryptophan residue was confirmed by the characteristic ultraviolet absorption at λ_{max} 339 nm, due to the conjugated system of dehydrotryptophan, and the detection of a tryptophan residue after keramamide F was subjected to hydrogenation, acid hydrolysis and amino acid analysis.[50,51]

A total synthesis of keramamide F was initiated by Sowinski and Toogood a few years later.[52] The molecule was disconnected into three fragments, of which the dehydrotryptophan containing fragment was synthesised from serine methyl ester **29** (Scheme 12).[52]



Scheme 12 Synthesis of the dehydrotryptophan fragment **33** of keramamide F. i) DPPA, Et_3N, CH_2Cl_2, 81%. ii) LiHMDS, THF, 35%. iii) BOP, Et_3N, CH_2Cl_2, 56%. iv) KDA, THF, 26%.

Three different methods were evaluated to introduce the dehydrotryptophan moiety. Initial attempts to directly form an amide bond between enoic acid **30** and dehydrotryptophan methyl ester **31** using a range of reagents were unsuccessful.[52] The next method attempted involved converting enoic acid **30** to azide **32** (Scheme 12A) which was then reacted with dehydrotryptophan methyl ester **31** to form amide **33** in 28% yield over two steps.[52] The third route to prepare dehydrotryptophan derivative **33** involved initial condensation of enoic acid **30** with trimethylphosphonate **34** using BOP (Scheme 12B). The resultant phosphonate **35** was then reacted with *N*-Boc-indole-3-carboxaldehyde **36** *via* a HWE reaction to afford the desired dehydrotryptophan fragment **33** albeit in 15% yield over the two steps.[52] Although Sowinski *et al.*[52] has reported synthetic procedures to access all three required fragments of keramamide F, the total synthesis of keramamide F is yet to be published.

2.3.1.2 Use of the HWE olefination for the total synthesis of barettin. Barettin was isolated in 1986 from *Geodia baretti*, a cold water sponge found in the deep waters off the Swedish coast (Fig. 3).[53]

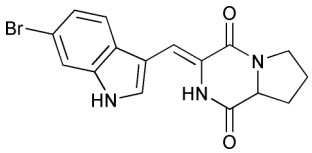
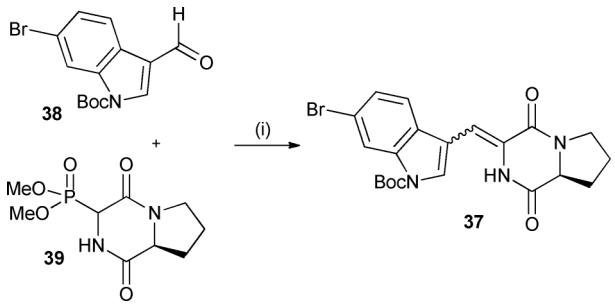


Fig. 3 Original proposed structure of barettin.

The following year a mixture of *Z*- and *E*-isomers of **37** of the proposed barettin structure were synthesised by HWE coupling of aldehyde **38** with phosphonate **39** (Scheme 13).[54] The isomers were able to be separated using column chromatography to afford diketopiperazine **37** with an *E* : *Z* isomeric ratio of 3 : 7. Disappointingly the spectral data obtained for both the *Z*- and *E*-isomers did not match that of the isolated material and the proposed structure was disputed.[54]

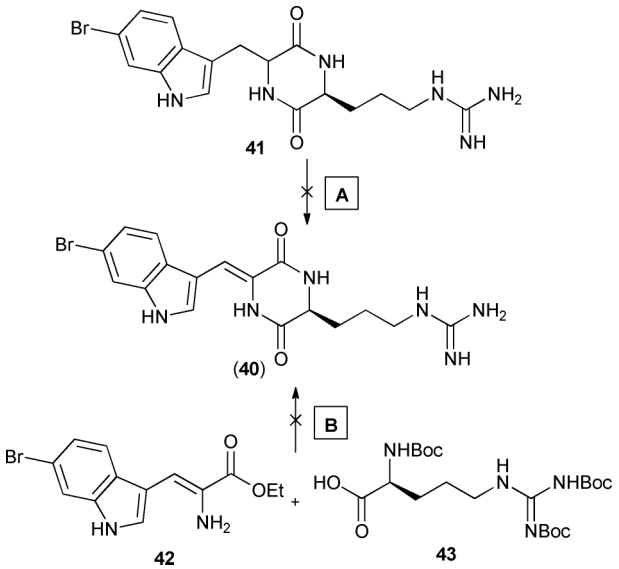


Scheme 13 Synthesis of original barettin structure. i) KO^{t}Bu (1 eq), CH_2Cl_2, −70 °C to rt.

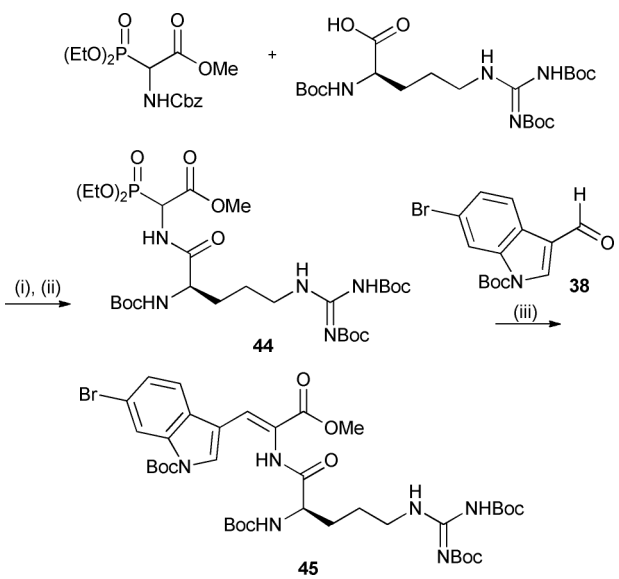
Due to the above findings and the isolation of an indole-arginine diketopiperazine derivative from *G. baretti* in Norway, which exhibited spectral data that closely matched that of barettin, the original structure of barettin was revised (**40**) in 2002 (Scheme 14).[55]

Two years later three synthetic routes were assessed to introduce the dehydrotryptophan residue during the total synthesis of the revised barettin structure (Scheme 14). Initially direct dehydrogenation of tryptophan **41** using oxidising reagents DDQ and TCCA failed to generate 6-bromo-dehydrotryptophan (**40**) (Scheme 14A). Similarly all attempts to couple dehydrotryptophan **42** with arginine **43** also failed (Scheme 14B).[56]

Finally, a HWE reaction used to couple phosphoglycinate **44** with aldehyde **38** using DBU (Scheme 15) afforded the desired product **45** in a moderate 55% yield.[56] When KO^{t}Bu was used as the base, lower yields were obtained. The spectral data obtained from the total synthesis of the revised structure was in agreement with that of the isolated natural product and thus the structure of barettin was confirmed (Fig. 2).[56]

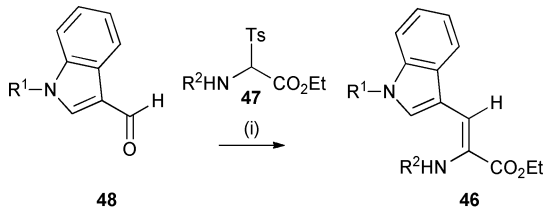


Scheme 14 Initial unsuccessful attempts to introduce the dehydrotryptophan moiety during the total synthesis of the revised structure of barettin (**40**).



Scheme 15 Successful introduction of dehydrotryptophan *en route* to barettin. i) H_2, Pd/C, EtOH, 4.5 h. ii) EDCI, HOBt, DIPEA, CH_2Cl_2, 64% over two steps. iii) DBU, CH_2Cl_2, −78 °C to rt, 20 h, 55%.

2.3.2 Wittig-type olefination reaction. An alternative olefination reaction was used by Kimura *et al.* to synthesise dehydrotryptophan derivatives **46**.[57] It was proposed that dehydrotryptopan derivatives **46** were synthesised by a Wittig-type reaction which first involved generating a Schiff base from the treatment of tosylglycinate **47** with base (Scheme 16).[57] The resultant Schiff base then reacts with tributylphosphine to form a phosphonium ylide, which upon treatment with indole-3-carbaldehyde **48** and elimination of tributylphosphine oxide affords *Z* dehydrotryptophan derivatives **46**.[57]

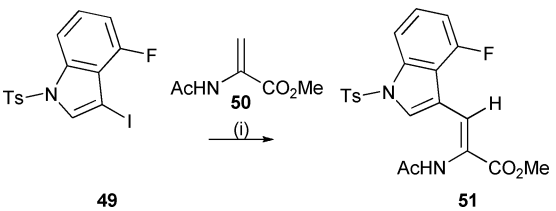


R^1 = Cbz, R^2 = Boc, 73%; R^1 = Boc, R^2 = Boc, 77%;
R^1 = Cbz, R^2 = Cbz, 60%; R^1 = Boc, R^2 = Cbz, 77%

Scheme 16 Synthesis of dehydrotryptophan derivatives **46** by a Wittig–type olefination. i) Na_2CO_3, Bu_3P, catalytic $Bu_4N^+Br^-$, $PhCH_3$, 67 h.

2.4 Heck reaction

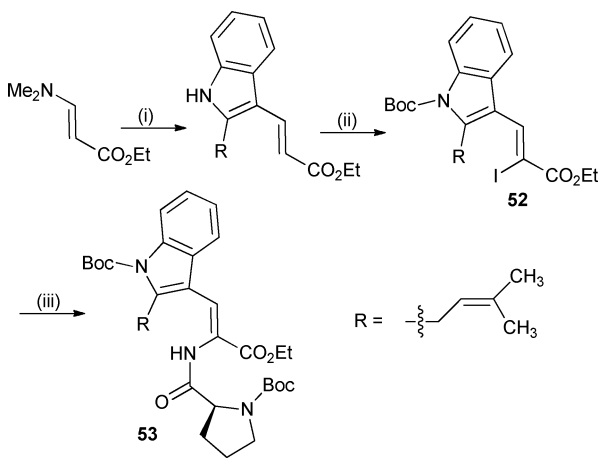
The palladium-catalysed Heck reaction has been used to synthesise dehydrotryptophan derivatives (Scheme 17).[58,59] Coupling of halide **49** with dehydroalanine derivative **50** afforded dehydrotryptophan derivative **51** in 40% yield (Scheme 17).[58] It was found that the addition of lithium chloride (2 eq) increased the yield to 77%.[58]



Scheme 17 Synthesis of dehydrotryptophan derivative **51** using the Heck reaction. i) K_2CO_3, Pd/C, DMF, 100 °C, 40%.

2.5 Copper catalysed amidation of vinyl halides

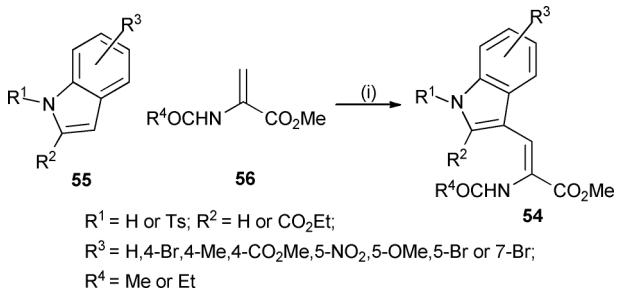
The copper(I) catalysed amidation of vinyl iodides or bromides[60] has been used to form proline-dehydrotryptophan dipeptides.[61,62] Vinyl iodide **52** was reacted with *N*-Boc-protected prolinamide to afford dehydrotryptophan dipeptide **53** in 57% yield (Scheme 18).[62]



Scheme 18 Synthesis of dehydrotryptophan dipeptide **53** by copper catalysed vinyl amidation. i) indole derivative, AcOH, 80 °C, 66%. ii) I_2, KOH, DMF, rt; $(Boc)_2O$, dioxane, DMAP, 0 °C to rt, 44% (2 steps). iii) *N*-Boc-protected prolinamide, CuI, *N*,*N*′-dimethylethylenediamine, $PhCH_3$, 100 °C, Cs_2CO_3, 57%.

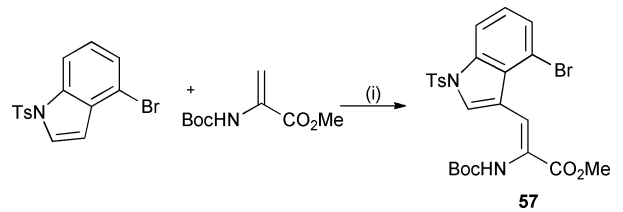
2.6 C–H vinylation of indole derivatives with dehydroalanine derivatives

The vinylation of indole derivatives with protected dehydroalanine derivatives using palladium has been reported in the literature.[63–66] *N*- and *C*-terminal protected dehydrotryptophan derivatives **54** were synthesised by Yokoyama *et al.* by the vinylation of indole derivatives **55** with protected dehydroalanine **56** using stoichiometric amounts of palladium(II) chloride, excess sodium acetate and heating at 120 to 130 °C (Scheme 19).[63]



Scheme 19 Synthesis of dehydrotryptophan derivatives **54** by C–H vinylation of indole. i) $PdCl_2$, AcONa, AcOH, 120–130 °C, 2 h, 14–59%.

Modification of the reaction conditions by replacing palladium(II) chloride with a stoichiometric amount of palladium(II) acetate and sodium acetate with $NaHCO_3$ afforded dehydrotryptophan derivatives **57** in 31% yield (Scheme 20).[64] It was found that the addition of chloranil (0.25–1.0 eq) improved the yield of **57** significantly (74–87%).[64] This led to the initial assumption that chloranil oxidised Pd(0) to Pd(II), however its role remains to be confirmed as a repeat of the experiment with a catalytic amount of Pd(II) in the presence of stoichiometric amounts of chloranil, afforded the dehydrotryptophan derivative **57** with a reduced 38% yield.[64]

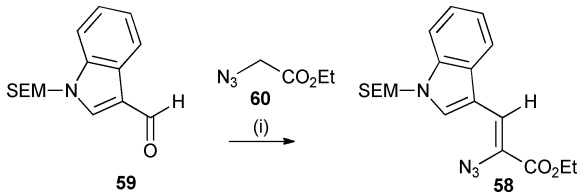


Scheme 20 Synthesis of dehydrotryptophan derivative **57**. i) $Pd(OAc)_2$, $NaHCO_3$, O_2, 70 °C, 7 h, 31%.

2.7 Condensation reactions

Condensation of indole aldehydes or imine derivatives with various substrates is a popular synthetic strategy to forming dehydrotryptophan amino acid and peptide derivatives. Examples of the use of various condensation reactions to form dehydrotryptophan amino acid derivatives are summarised below.

2.7.1 Aldol condensation to form α-azido dehydrotryptophan derivatives. Fresneda *et al.* synthesised the enantiopure azido dehydrotryptophan derivative **58** by the low temperature condensation of aldehyde **59** with azidoacetate **60** using sodium ethoxide as base (Scheme 21).[67]

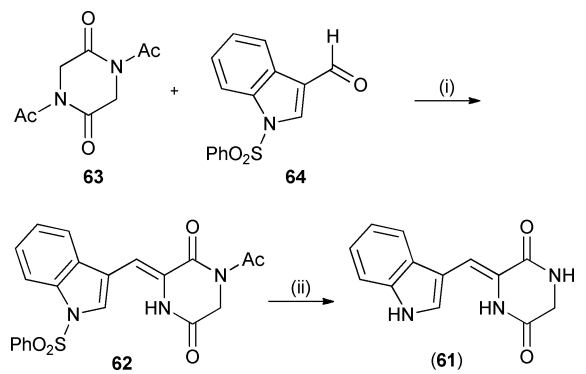


Scheme 21 Synthesis of azido dehydrotryptophan derivative **58**. i) NaOEt, EtOH, –15 °C, 80%.

The condensation of indole aldehyde derivatives with azidoacetates to form azido dehydrotryptophan derivatives having differing substituents has been reported using similar reaction conditions.[68–71]

2.7.2 Use of the aldol condensation for the total synthesis of dipodazine. Dipodazine (**61**) (Fig. 2), a cyclic dipeptide comprised of dehydrotryptophan and glycine, is a major component of the blue-fluorescent metabolites of the mould *Penicillium dipodomyis*, obtained from cheek pouches of kangaroo rats, and *Penicillium nalgiovense* from mould-fermented sausages.[72,73]

The synthesis of dipodazine was attempted using an aldol condensation to introduce the dehydrotryptophan moiety (Scheme 22) rather than effecting an intramolecular cyclisation of the linear dehydrotryptophan-glycine precursor, as dehydrotryptophan was 'not readily available'.[73] The semi-protected dipeptide **62** was obtained by aldol condensation between 1,4-diacetyl-2,5-piperazinedione **63** and (1-benzenesulfonyl)indole-3-carbaldehyde **64** using cesium carbonate as the base and proceeded with concomitant loss of an acetyl group.[73,74] The *Z*-configuration of dipeptide **62** was confirmed by NMR analysis and the remaining protecting groups were removed using ethanolic aqueous NaOH to afford dipodazine (**61**).[73]

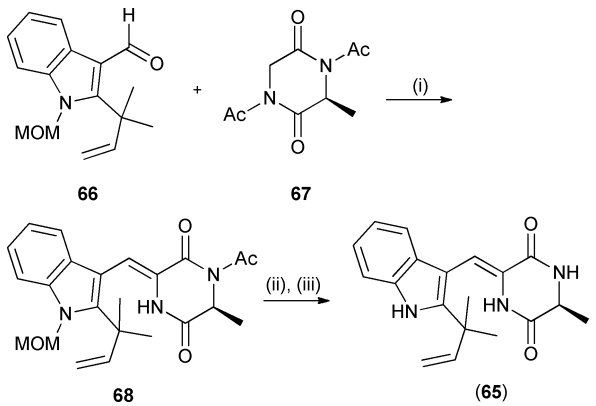


Scheme 22 Synthesis of dipodazine (**61**) by aldol condensation. i) Cs_2CO_3, 4 Å MS, DMF, rt, 18 h, 77%. ii) NaOH, H_2O/EtOH, reflux, 3 h, 53%.

2.7.3 Use of the aldol condensation for the total synthesis of Neoechinulin A. Neoechinulin A (**65**) (Fig. 2), a dehydrotryptophan-alanine diketopiperazine derivative, was isolated by several groups from different species of the fungus *Aspergillus*.[75,76] It exhibits strong antioxidant activity with significant radical scavenging ability.[76]

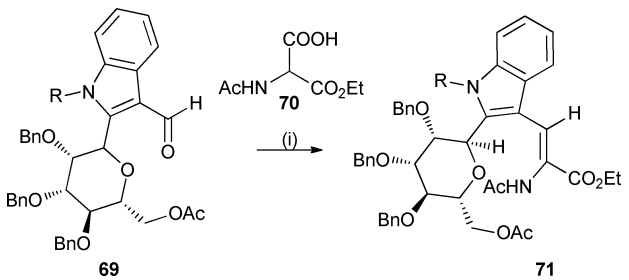
An aldol condensation procedure was used for the synthesis of neoechinulin A to introduce the dehydrotryptophan moiety by condensation of aldehyde **66** with diketopiperazine **67**

(Scheme 23).[76] The *Z*-configuration of **68** was assigned by NMR analysis after which deprotection of the acetyl group was achieved using hydrazine monohydrate followed by removal of the methoxymethyl group using formic acid to afford neoechinulin A (**65**).[76]



Scheme 23 Synthesis of neoechinulin A (**65**) utilising an aldol condensation. i) KO^{t}Bu, DMF, 43%. ii) $H_2NNH_2{\cdot}H_2O$, DMF. iii) HCO_2H, H_2O.

2.7.4 Decarboxylative aldol condensation. Decarboxylative aldol condensation of indole aldehyde derivatives with appropriately substituted carboxylic acids is another way of accessing dehydrotryptophan containing compounds.[77] This method has been used to access *C*-mannosyltryptophan derivatives by treatment of indole aldehyde **69** and acid **70** with acetic anhydride and pyridine to afford dehydrotryptophan derivative **71** in 49%[78] and 66%[79] yield (Scheme 24).

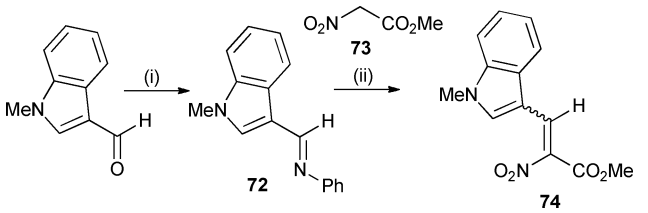


Scheme 24 Synthesis of dehydrotryptophan derivative **71** by decarboxylative aldol condensation. i) Ac_2O, pyridine, 49%, 6 h, rt.

2.7.5 Knoevenagel condensation. The Knoevenagel condensation has been used to form dehydrotryptophan derivatives with varying *E* : *Z* ratios of isomers by reaction of imines derived from indole-3-carbaldehyde derivatives.[80–83] For example, the condensation of *N*-phenyl imine **72** with methyl nitroacetate **73** in the presence of acetic anhydride afforded the nitro dehydrotryptophan derivative **74** with an isomeric ratio of 40 : 1 (*Z* : *E*) (Scheme 25).[80]

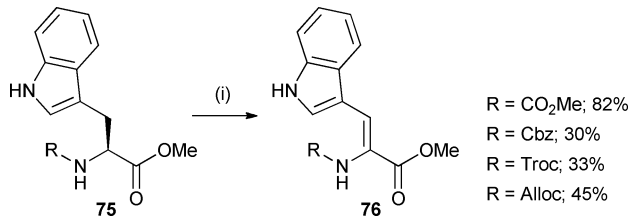
2.8 Elimination reactions

2.8.1 Direct dehydrogenation of a tryptophan derivative. The direct dehydrogenation of partially protected tryptophan derivatives **75** to afford dehydrotryptophan derivatives **76** was achieved by Baran *et al.* during the synthesis of the complex natural products avrainvillamide and the stephacidins.[84] The use of



Scheme 25 Synthesis of nitro dehydrotryptophan derivative **74** by Knoevenagel condensation. i) $PhNH_2$, $MgSO_4$, 70 °C, 4–6 h, 99%. ii) Ac_2O, 50 °C, 17 h, 74%.

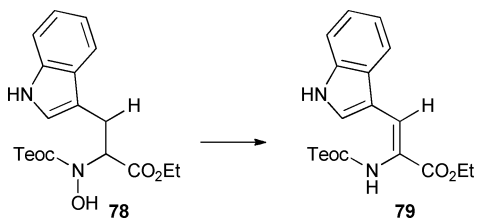
nitrosobenzene in the presence of the Lewis acid zirconium tetrachloride effected the desired transformation after initial screening of a variety of oxidants including CAN, DDQ, IBX and $Pd/C/O_2$ proved to be unsuccessful. The reaction was proposed to proceed *via* the formation of a tryptophan-nitrosobenzene intermediate followed by elimination and tautomerisation to form dehydrotryptophan derivatives of structure **76** (Scheme 26).[84]



Scheme 26 Synthesis of dehydrotryptophan derivatives by direct dehydrogenation. i) PhNO, $ZrCl_4$, CH_2Cl_2, 0 °C to 20 °C.

This direct dehydrogenation method has recently been used to introduce the dehydrotryptophan moiety during the total synthesis of variecolorin C (**77**) (Fig. 2), a dehydrotryptophan-alanine dipeptide derivative.[85]

2.8.2 Direct dehydration of a tryptophan derivative. Undesired dehydration occurred during attempted *O*-alkylation of protected *N*-hydroxy tryptophan **78**. Dehydrotryptophan derivative **79** (Scheme 27) was obtained in near quantitative yield when **78** was treated with 4-bromo-1,1-dimethoxybutane in the presence of either DMSO/KO^{t}Bu or DME/NaH.[86]

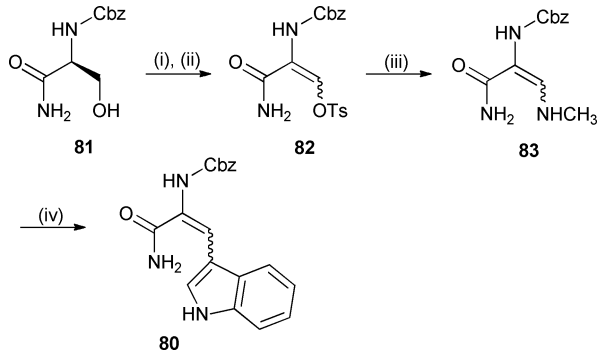


Scheme 27 Dehydration of *N*-hydroxy tryptophan **78**. i) DMSO/KO^{t}Bu or DME/NaH.

2.9 Substitution reactions of dehydroamino acid derivatives

Displacement of a vinyl leaving group from dehydroamino acid derivatives using an indole nucleophile is a popular method for the formation of dehydrotryptophan containing compounds.

An *N*- and *C*-terminal protected dehydrotryptophan derivative **80** was synthesised by Nakazawa *et al.* starting from the protected serine derivative **81** (Scheme 28).[87]

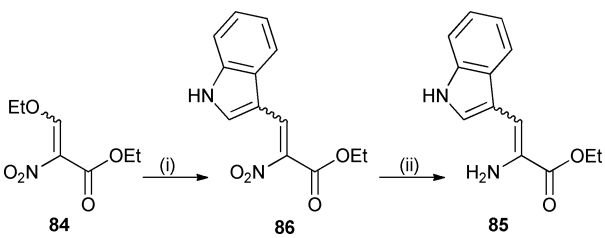


Scheme 28 Synthesis of dehydrotryptophan derivatives by displacement. i) TsCl (3 eq), DMSO/DMF (1 : 2), –5 °C, 10 min. ii) Et_3N, 1 h, 48%. iii) CH_3NH_2/MeOH. iv) indole (1 eq), AcOH, 40 °C, 3 d, 49%.

This procedure involved initial oxidation of the primary alcohol of serine **81** to the corresponding aldehyde using DMSO activated by *p*-toluenesulfonyl chloride and afforded enol tosylate **82**.[88] Displacement of the tosyl group with indole to form the final compound occurred in low yield, hence the tosyl group was replaced with other leaving groups. Displacement of the methylamino group of **83** by indole formed the final compound **80** in 49% yield (*Z* : *E* 11 : 1) and it was proposed that the yield could be improved if a better leaving group was used.[87]

The formation of dehydrotryptophan was confirmed by its characteristic ultraviolet absorption in which the *Z* isomer exhibited maxima at 275 and 332 nm. It was proposed that this method can be used to synthesise other dehydroamino acids from serine amino acid derivatives or from serine residues incorporated into peptides.[87]

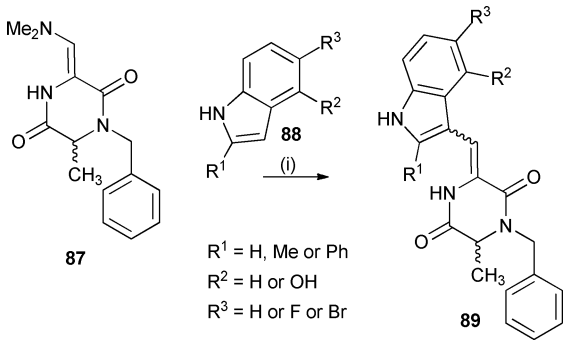
In a similar manner, an alternative dehydroserine derivative **84** was used by Hammadi *et al.* to synthesise dehydrotryptophan ethyl ester **85**, also as a mixture of *E*- and *Z*-isomers that were not separated (Scheme 29).[22] Displacement of the ethoxy group of nitroacrylate **84** with indole afforded the nitro-indole adduct **86** in a comparatively high 80% yield. Selective hydrogenation of the nitro-functional group of **86** afforded dehydrotryptophan ethyl ester **85** in 96% yield as an isomeric mixture (*Z* : *E* 1 : 1), where the proportion of the *E*-isomer was maximised by controlling the temperature during solvent evaporation.[22]



Scheme 29 Synthesis of (*E*/*Z*)-dehydrotryptophan ethyl ester **85**. i) 1*H*-indole (0.8 eq), rt, 18 h, 80%. ii) H_2, Pt/C, EtOAc, 2 h, 96%.

This reaction has also been reported in the literature using bromo-substituted indoles.[89]

Displacement of a vinyl dimethylamino group by indole derivatives is another method used to synthesise dehydrotryptophan derivatives.[61,90–92] Displacement of the dimethylamino group of **87** with indole **88** in the presence of acetic acid was used to form dehydrotryptophan dipeptides **89** (Scheme 30).[61,92]



Scheme 30 Synthesis of dehydrotryptophan dipeptide **89** by dimethylamino displacement. i) AcOH, reflux, 1–6 h, 22–80%.

2.10 Using biosynthetic engineering

The calcium-dependent antibiotics (CDAs) were isolated from *Streptomyces coelicolor* A(3)2 fermentations in 1983 by Hopwood *et al.* and were found to inhibit the growth of Gram-positive bacteria in the presence of Ca^{2+} ions.[93] The acidic amino acid residues are essential for antibiotic activity as they are responsible for coordinating to calcium ions, which is then followed by aggregation of CDAs and penetration and disruption of the membrane of Gram-positive bacteria, which ultimately results in cell death.[94]

Several CDA variants have been discovered, of which the a-series (Fig. 4) was shown to contain a *Z*-dehydrotryptophan residue adjacent to the ester linkage of the cyclodepsipeptide, as evident by its characteristic ultraviolet absorption at λ_{max} 349 nm for CDA2a and CDA4a.[93–95]

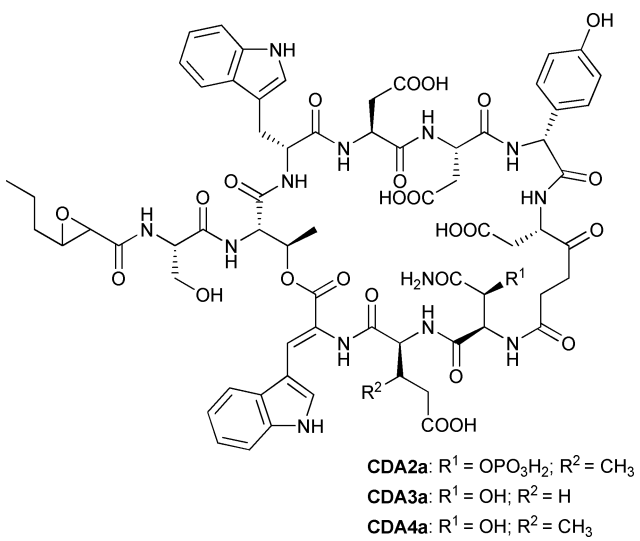


Fig. 4 Structure of the a-series of calcium-dependent antibiotics.

To date the chemical synthesis of these compounds has not been reported, however several other a-series CDA variants containing the *Z*-dehydrotryptophan residue have recently been synthesised by Micklefield using biosynthetic engineering, whereby genes are reprogrammed to generate new 'non-natural' compounds.[94]

3. Conclusions

Dehydrotryptophan is an interesting amino acid of which the synthesis remains to be reported in the free amino acid form for ready modification and use in solid phase peptide synthesis. Several dehydrotryptophan amino acid and dipeptide derivatives have been synthesised by the various synthetic methods described herein, however the total chemical synthesis of large peptides containing dehydrotryptophan are yet to be reported.

Abbreviations

BOP	Benzotriazol-1-oxy-tris(demethylamino)phosphonium hexafluorophosphate
CAN	Cerium(IV) ammonium nitrate
DBU	1,8-Diazobicyclo[5.4.0]undec-7-ene
DDQ	2,3-Dichloro-5,6-dicayno-1,4-benzoquinone
DIPEA	*N*,*N*-diisopropylethylamine
DMAP	4-*N*,*N*-dimethylaminopyridine
DME	Dimethyl ether
DMF	Dimethylformamide
DMSO	Dimethyl sulfoxide
DPPA	Diphenylphosphoryl azide
EDCI	1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide
HOBt	1-Hydroxybenzotriazole
IBX	2-Iodoxybenzoic acid
KDA	Potassium diisopropylamide
LDA	Lithium diisopropylamide
LiHMDS	Lithium hexamethyldisilazane
MS	Molecular sieves
NMR	Nuclear magnetic resonance
TCCA	Trichloroisocyanuric acid
THF	Tetrahydrofuran

References

1 F. S. Nandel and T. R. Sahrawat, *Biopolymers*, 2009, **92**, 44–51.
2 M. Thormann and H. J. Hofmann, *THEOCHEM*, 1998, **431**, 79–96.
3 T. J. Nitz, E. M. Holt, B. Rubin and C. H. Stammer, *J. Org. Chem.*, 1981, **46**, 2667–2671.
4 Z. Kubica, T. Kozlecki and B. Rzeszotarska, *Chem. Pharm. Bull.*, 2000, **48**, 296–297.
5 C. Dugave and L. Demange, *Chem. Rev.*, 2003, **103**, 2475–2532.
6 J. S. Davies, *J. Pept. Sci.*, 2003, **9**, 471–501.
7 A. S. Abreu, P. M. T. Ferreira, L. S. Monteiro, M. J. R. P. Queiroz, I. C. F. R. Ferreira, R. C. Calhelha and L. M. Estevinho, *Tetrahedron*, 2004, **60**, 11821–11828.
8 C. Cativiela, M. D. Diaz-de-Villegas, J. A. Galvez and G. Su, *Arkivoc*, 2004, 59–66.
9 M. Lisowski, R. Latajka, B. Picur, T. Lis, I. Bryndal, M. Rospenk, M. Makowski and P. Kafarski, *Biopolymers*, 2008, **89**, 220–234.
10 P. Mathur, S. Ramakumar and V. S. Chauhan, *Biopolymers*, 2004, **76**, 150–161.
11 J. C. Jimenez, N. Bayo, B. Chavarria, A. Lopez-Macia, M. Royo, E. Nicolas, E. Giralt and F. Albericio, *Lett. Pept. Sci.*, 2002, **9**, 135–141.
12 M. A. Blaskovich, *Handbook on Syntheses of Amino Acids*, Oxford University Press, New York, 2010.
13 A. B. Hughes, *Amino Acids, Peptides and Proteins in Organic Chemistry*, 2009.
14 C. Bonauer, T. Walenzyk and B. Konig, *Synthesis*, 2006, 1–20.
15 J. M. Humphrey and A. R. Chamberlin, *Chem. Rev.*, 1997, **97**, 2243–2266.
16 U. Schmidt, A. Lieberknecht and J. Wild, *Synthesis*, 1988, 159–172.
17 B. Amir-Heidari, J. Thirlway and J. Micklefield, *Org. Lett.*, 2007, **9**, 1513–1516.
18 A. Hammadi, H. Lam, M. Gondry, A. Menez and R. Genet, *Tetrahedron*, 2000, **56**, 4473–4477.
19 A. Hammadi, A. Menez and R. Genet, *Tetrahedron*, 1997, **53**, 16115–16122.
20 Y. Noda, K. Takai, T. Tokuyama, S. Narumiya, H. Ushiro and O. Hayaishi, *J. Biol. Chem.*, 1978, **253**, 4819–4822.
21 M. E. Gustafson, D. Miller, P. J. Davis, J. P. Rosazza, C. J. Chang and H. G. Floss, *J. Chem. Soc., Chem. Commun.*, 1977, 842–843.
22 A. Hammadi, A. Menez and R. Genet, *Tetrahedron Lett.*, 1996, **37**, 3309–3312.
23 R. J. Block, *Chem. Rev.*, 1946, **38**, 501–571.
24 M. Oba, R. Ueno, M. Fukuoka, M. Kainosho and K. Nishiyama, *J. Chem. Soc., Perkin Trans. 1*, 1995, 1603–1609.
25 G. W. Kirby and M. J. Varley, *J. Chem. Soc., Chem. Commun.*, 1974, 833–834.
26 P. Skrabal, V. Rizzo, A. Baici, F. Bangerter and P. Luigiluisi, *Biopolymers*, 1979, **18**, 995–1008.
27 T. Bach and B. Pruger, *Synthesis*, 2007, 1103–1106.
28 M. Fujita and I. Ojima, *Tetrahedron Lett.*, 1983, **24**, 4573–4576.
29 T. Moriya, N. Yoneda, M. Miyoshi and K. Matsumoto, *J. Org. Chem.*, 1982, **47**, 94–98.
30 H. W. Bond, *J. Biol. Chem.*, 1948, **175**, 531–534.
31 M. Bakhra, G. S. Katrukha and A. B. Silaev, *Khim. Prir. Soedin.*, 1973, 280–281.
32 J. C. Sheehan, D. Mania, S. Nakamura, J. A. Stock and K. Maeda, *J. Am. Chem. Soc.*, 1968, **90**, 462–470.
33 T. G. Barros, S. Pinheiro, J. S. Williamson, A. Tanuri, H. S. Pereira, R. M. Brindeiro, J. B. A. Neto, O. A. C. Antunes and E. M. F. Muri, *Synthesis*, 2009, 620–626.
34 H. Kinoshita, M. Kometani, K. Ihara and R. Kimura, *Bull. Chem. Soc. Jpn.*, 2009, **82**, 364–380.
35 C. J. Moody and J. Sperry, *Tetrahedron*, 2010, **66**, 6483–6495.
36 T. V. RajanBabu, H. J. Lim and J. C. Gallucci, *Org. Lett.*, 2010, **12**, 2162–2165.
37 F. von Nussbaum, S. Anlauf, C. Freiberg, J. Benet-Buchholz, J. Schamberger, T. Henkel, G. Schiffer and D. Habich, *ChemMedChem*, 2008, **3**, 619–626.
38 Y. X. Li, E. Hayman, M. Plesescu and S. R. Prakash, *Tetrahedron Lett.*, 2008, **49**, 1480–1483.
39 J. R. Harrison and C. J. Moody, *Tetrahedron Lett.*, 2003, **44**, 5189–5191.
40 W. Wang, M. Y. Cai, C. Y. Xiong, J. Y. Zhang, D. Trivedi and V. J. Hruby, *Tetrahedron*, 2002, **58**, 7365–7374.
41 K. S. Feldman, K. J. Eastman and G. Lessene, *Org. Lett.*, 2002, **4**, 3525–3528.
42 W. Wang, C. Y. Xiong, J. Q. Yang and V. J. Hruby, *Tetrahedron Lett.*, 2001, **42**, 7717–7719.
43 M. C. Bagley, S. L. Hind and C. J. Moody, *Tetrahedron Lett.*, 2000, **41**, 6897–6900.
44 F. Micheli, R. Di Fabio, D. Baraldi, N. Conti, A. Cugola, P. Gastaldi, S. Giacobbe, C. Marchioro, M. Mugnaini, L. Rossi, A. Pecunioso and G. Pentassuglia, *Arch. Pharm.*, 1999, **332**, 271–278.
45 D. J. Bentley, A. M. Z. Slawin and C. J. Moody, *Org. Lett.*, 2006, **8**, 1975–1978.
46 D. J. Bentley and C. J. Moody, *Org. Biomol. Chem.*, 2004, **2**, 3545–3547.
47 E. A. Couladouros and A. D. Magos, *Mol. Diversity*, 2005, **9**, 99–109.
48 R. T. Buck, P. A. Clarke, D. M. Coe, M. J. Drysdale, L. Ferris, D. Haigh, C. J. Moody, N. D. Pearson and E. Swann, *Chem.–Eur. J.*, 2000, **6**, 2160–2167.
49 C. G. Shin, N. Takahashi and Y. Yonezawa, *Chem. Pharm. Bull.*, 1990, **38**, 2020–2023.
50 F. Itagaki, H. Shigemori, M. Ishibashi, T. Nakamura, T. Sasaki and J. Kobayashi, *J. Org. Chem.*, 1992, **57**, 5540–5542.
51 K. K. Babievskii, Y. A. Davidovich, V. I. Bakhmutov and S. V. Rogozhin, *Bull. Acad. Sci. USSR, Div. Chem. Sci. (Engl. Transl.)*, 1988, **37**, 549–552.
52 J. A. Sowinski and P. L. Toogood, *Tetrahedron Lett.*, 1995, **36**, 67–70.
53 G. Lidgren and L. Bohlin, *Tetrahedron Lett.*, 1986, **27**, 3283–3284.
54 A. Lieberknecht and H. Griesser, *Tetrahedron Lett.*, 1987, **28**, 4275–4278.
55 S. Solter, R. Dieckmann, M. Blumenberg and W. Francke, *Tetrahedron Lett.*, 2002, **43**, 3385–3386.
56 A. L. Johnson, J. Bergman, M. Sjogren and L. Bohlin, *Tetrahedron*, 2004, **60**, 961–965.
57 R. Kimura, T. Nagano and H. Kinoshita, *Bull. Chem. Soc. Jpn.*, 2002, **75**, 2517–2525.

58 C. A. Merlic and M. F. Semmelhack, *J. Organomet. Chem.*, 1990, **391**, C23–C27.
59 P. J. Harrington, L. S. Hegedus and K. F. Mcdaniel, *J. Am. Chem. Soc.*, 1987, **109**, 4335–4338.
60 L. Jiang, G. E. Job, A. Klapars and S. L. Buchwald, *Org. Lett.*, 2003, **5**, 3667–3669.
61 B. Stanovnik, J. Wagger, U. Groselj, A. Meden and J. Svete, *Tetrahedron*, 2008, **64**, 2801–2815.
62 B. Stanovnik, J. Wagger and J. Svete, *Synthesis*, 2008, 1436–1442.
63 Y. Yokoyama, M. Takahashi, M. Takashima, Y. Kohno, H. Kobayashi, K. Kataoka, K. Shidori and Y. Murakami, *Chem. Pharm. Bull.*, 1994, **42**, 832–838.
64 Y. Yokoyama, T. Matsumoto and Y. Murakami, *J. Org. Chem.*, 1995, **60**, 1486–1487.
65 P. Lloyd-Williams, M. Prieto, S. Mayor and E. Giralt, *J. Org. Chem.*, 2009, **74**, 9202–9205.
66 K. L. Lee, J. B. Goh and S. F. Martin, *Tetrahedron Lett.*, 2001, **42**, 1635–1638.
67 P. M. Fresneda, P. Molina and M. A. Sanz, *Synlett*, 2000, 1190–1192.
68 P. Molina, P. Almendros and P. M. Fresneda, *Tetrahedron*, 1991, **50**, 2241–2254.
69 P. Molina, P. M. Fresneda and P. Almendros, *Tetrahedron Lett.*, 1992, **33**, 4491–4494.
70 C. J. Moody and J. G. Ward, *J. Chem. Soc., Perkin Trans. 1*, 1984, 2903–2909.
71 C. J. Moody and J. G. Ward, *J. Chem. Soc., Perkin Trans. 1*, 1984, 2895–2901.
72 D. Sorensen, T. O. Larsen, C. Christophersen, P. H. Nielsen and U. Anthoni, *Phytochemistry*, 1999, **51**, 1181–1183.
73 A. L. Johnson, T. Janosik and J. Bergman, *Arkivoc*, 2002, 57–61.
74 C. Gallina and A. Liberato, *Tetrahedron*, 1974, **30**, 667–673.
75 S. Kobayashi, K. Kuramochi, T. Aoki, A. Nakazaki, S. Kamisuki, M. Takeno, K. Ohnishi, K. Kimoto, N. Watanabe, T. Kamakura, T. Arai and F. Sugawara, *Synthesis*, 2008, 3810–3818.
76 T. Aoki, S. Kamisuki, M. Kimoto, K. Ohnishi, Y. Takakusagi, K. Kuramochi, Y. Takeda, A. Nakazaki, K. Kuroiwa, T. Ohuchi, F. Sugawara, T. Arai and S. Kobayashi, *Synlett*, 2006, 677–680.
77 U. Hengartner, D. Valentine, K. K. Johnson, M. E. Larscheid, F. Pigott, F. Scheidl, J. W. Scott, R. C. Sun, J. M. Townsend and T. H. Williams, *J. Org. Chem.*, 1979, **44**, 3741–3747.
78 T. Nishikawa, Y. Koide, S. Kajii, K. Wada, M. Ishikawa and M. Isobe, *Org. Biomol. Chem.*, 2005, **3**, 687–700.
79 T. Nishikawa, M. Ishikawa and M. Isobe, *Synlett*, 1999, 123–125.
80 F. P. J. T. Rutjes, D. Blanco-Ania, P. H. H. Hermkens, L. A. J. M. Sliedregt and H. W. Scheeren, *Tetrahedron*, 2009, **65**, 5393–5401.
81 Z. M. Xiong, D. H. A. Gao, D. A. Cogan, D. R. Goldberg, M. H. Hao, N. Moss, E. Pack, C. Pargellis, D. Skow, T. Trieselmann, B. Werneburg and A. White, *Bioorg. Med. Chem. Lett.*, 2008, **18**, 1994–1999.
82 R. S. Fornicola, E. Oblinger and J. Montgomery, *J. Org. Chem.*, 1998, **63**, 3528–3529.
83 R. Rodriguez, I. VInets, A. Diez, M. Rubiralta and E. Giralt, *Synth. Commun.*, 1996, **26**, 3029–3059.
84 P. S. Baran, B. D. Hafensteiner, N. B. Ambhaikar, C. A. Guerrero and J. D. Gallagher, *J. Am. Chem. Soc.*, 2006, **128**, 8678–8693.
85 X. G. She, Q. P. Dai, X. G. Xie, S. Y. Xu, D. H. Ma and S. B. Tang, *Org. Lett.*, 2011, **13**, 2302–2305.
86 P. H. H. Hermkens, J. H. Vonmaarseveen, H. W. Berens, J. M. M. Smits, C. G. Kruse and H. W. Scheeren, *J. Org. Chem.*, 1990, **55**, 2200–2206.
87 T. Nakazawa, M. Ishii, H. J. Musiol and L. Moroder, *Tetrahedron Lett.*, 1998, **39**, 1381–1384.
88 T. Nakazawa, T. Suzuki and M. Ishii, *Tetrahedron Lett.*, 1997, **38**, 8951–8954.
89 D. C. Horwell, P. D. Nichols, G. S. Ratcliffe and E. Roberts, *J. Org. Chem.*, 1994, **59**, 4418–4423.
90 L. Jukic, J. Svete and B. Stanovnik, *J. Heterocycl. Chem.*, 2001, **38**, 869–876.
91 L. Selic, R. Jakse, K. Lampic, L. Golic, S. Golic-Grdadolnik and B. Stanovnik, *Helv. Chim. Acta*, 2000, **83**, 2802–2811.
92 J. Wagger, D. Bevk, A. Meden, J. Svete and B. Stanovnik, *Helv. Chim. Acta*, 2006, **89**, 240–248.
93 M. A. Marahiel and M. Strieker, *ChemBioChem*, 2009, **10**, 607–616.
94 J. Micklefield, *Pure Appl. Chem.*, 2009, **81**, 1065–1074.
95 Z. Hojati, C. Milne, B. Harvey, L. Gordon, M. Borg, F. Flett, B. Wilkinson, P. J. Sidebottom, B. A. M. Rudd, M. A. Hayes, C. P. Smith and J. Micklefield, *Chem. Biol.*, 2002, **9**, 1175–1187.