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Synthesis of (2*S*, 3*R*, 4*S*), (2*S*, 3*S*, 4*R*)-Epoxyprolines and Aminohydroxyprolines

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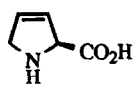
Abstract: 2*S*, 3*R*, 4*S*- and 2*S*, 3*S*, 4*R*-epoxy-*L*-prolines were synthesised from *trans*-4-hydroxy-*L*-proline. Assignment of the stereochemical configurations of the epoxy prolines was achieved by n.O.e. studies and chemical correlation. The synthetic utility of the protected epoxides was investigated briefly by ring opening with NaN_3, followed by deprotection to give aminohydroxy prolines.

trans-4-Hydroxy-*L*-proline **3** is a non-proteinogenic amino acid which has been extensively used in synthesis.[1] Presently, it is produced commercially by purification of collagen hydroxylates. The discovery of proline 3- and 4-hydroxylase enzymes has opened up the possibility of utilizing these enzymes for the production of hydroxy-prolines.[2, 3, 4] Known proline hydroxylases are all non-haem iron dependent enzymes with a requirement for 2-oxoglutarate as a cofactor. Since a number of enzymes from this family of oxygenases/oxidases have been shown to have a relatively lax substrate selectivity[5] we envisaged that proline 4-hydroxylase might be used for the synthesis of synthetically useful amino acids. It has been demonstrated that proline 4-hydroxylase from *Streptomyces griseoviridus* P8648 catalyses *in vitro* the epoxidation of its unnatural substrate 3,4-dehydro-*L*-proline **1**.[6] The product was shown to be (2*S*, 3*R*, 4*S*)-epoxy-*L*-proline **2a** by comparison with synthetic samples of (2*S*, 3*R*, 4*S*)-epoxy-*L*-proline **2a** and (2*S*, 3*S*, 4*R*-)-epoxy-*L*-proline **2b**. Herein, we describe the experimental details for the preparation of epoxides **2a** and **2b**. An alternative synthesis of epoxide **2a** has been reported recently by Herdeis *et al.*[7]



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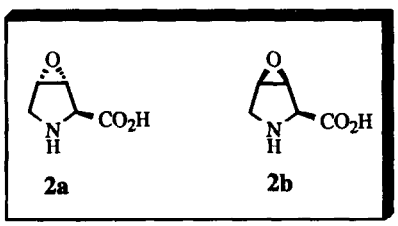

2a **2b**

Discussion

Epoxidation of *N*-benzenesulphonyl-3, 4-dehydro-*L*-proline methyl ester using trifluoroperacetic acid has been described by Hudson *et al.*[8] The resultant epoxide mixture were reported to be chromatographically inseparable and unstable towards deprotection to form the free amino acids [i.e. **2a** or **2b**], but was apparently

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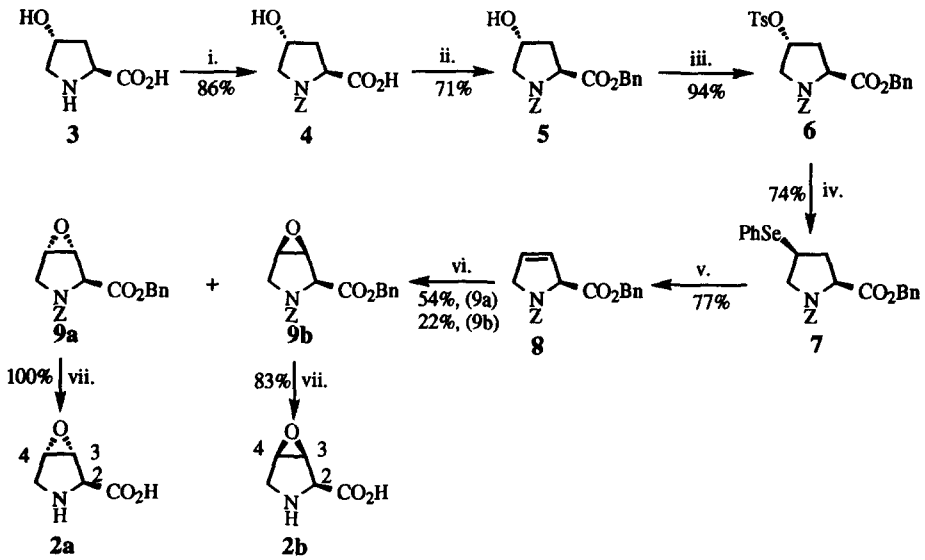
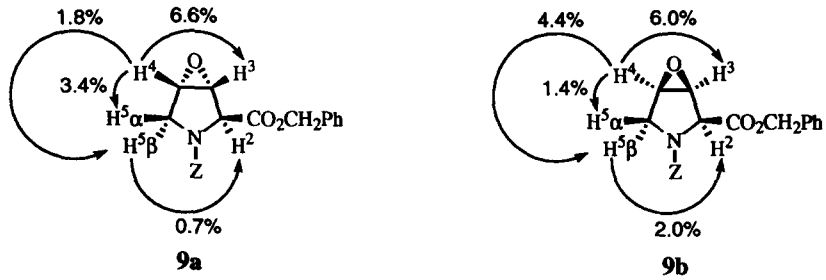
resistant to catalytic hydrogenolysis. Hence, we investigated the synthesis and deprotection of the *N*-benzyloxycarbonyl-3, 4-epoxy-*L*-proline benzyl esters **9a** and **9b**. The requisite 3, 4-dehydro-*L*-proline derivative **8** (**Scheme 1**) was synthesised from *trans*-hydroxy-*L*-proline **3** using a modified version of the literature method.[9] After diprotection[10], attempted tosylation of alcohol **5** with tosyl chloride/pyridine failed to generate the desired tosylate **6**[9] in acceptable yield, even with the addition of *N, N*-dimethylaminopyridine and/or with heating. Tosylation of **5** was achieved by using 1-(toluenesulphonyl)-3-methylimidazolium triflate.[11, 12] Selenation ($PhSeSePh/NaBH_4$) of **6** to give **7** was carried out in ^tBuOH to avoid ester exchange which occurred when using ethanol as a solvent. Selenide **7** was converted to the required dehydro-*L*-proline derivative **8** by oxidative elimination (H_2O_2/pyridine). Treatment of **8** with m-CPBA in refluxing 1,2-dichloroethane with the presence of a radical inhibitor[13, 14] gave *trans*-epoxide **9a** (54%) and *cis*-epoxide **9b** (22%) which were separated by flash chromatography. Deprotection by catalytic hydrogenlysis (Pd/ C/ H_2) gave the desired epoxides **2a** and **2b** without (by 500 MHz ^1H NMR analysis) any observable cleavage of the epoxide ring.[15]



Scheme 1: (i) $PhCH_2OCOCl$ / NaOH / THF / H_2O then HCl; (ii) $PhCH_2Br$ / NaI / K_2CO_3 / DMF; (iii) 1-(toluenesulphonyl)-3-methylimidazolium triflate / *N*-methylimidazole / THF; (iv) $NaBH_4$ / PhSeSePh / ^tBuOH / reflux; (v) H_2O_2 / pyridine / DCM; (vi) 3-chloroperoxybenzoic acid / 3-*tert*-butyl-4-hydroxy-5-methylphenylsulphide / 1,2-dichloroethane / reflux; (vii) H_2 / Pd / C/ THF / H_2O.

Initial structural assignments of the *trans*-**9a** and *cis*-**9b** epoxides were based on the observed coupling constants between H-2 and H-3. In **9a**, there was no observed coupling between H-2 and H-3 whereas a coupling constant of 2.5 Hz was observed in the case of **9b**. Nuclear Overhauser effect studies on epoxide **9a** revealed a small (0.7%) signal enhancement of the H-2 resonance when that of H-5β (assigned *via* a COSY spectrum) was irradiated. Signal enhancements of 3.4% and 1.8% were observed for the H-5α and H-5β resonances, respectively when H-4 was irradiated. These results implied that **9a** was the *trans*-epoxide. Analogous n.O.e. experiments led to the tentative assignment of **9b** as the *cis*-epoxide. Thus, a 2.0% signal

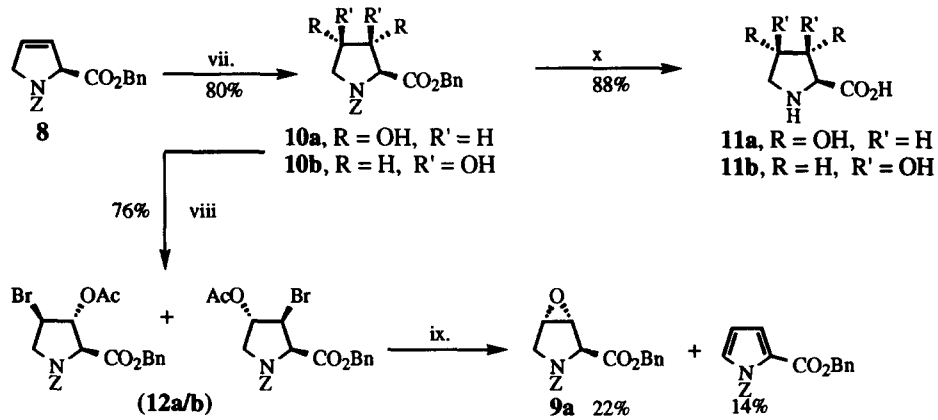
enhancement of the H-2 resonance was observed when H-5β (assigned *via* COSY) was irradiated. Irradiation of the H-4 resonance produced 1.4% and 4.4% signal enhancements of the H-5α and H-5β resonances, respectively (**Figure 1**).



Selected n.O.e. data for **9a** and **9b**

Figure 1

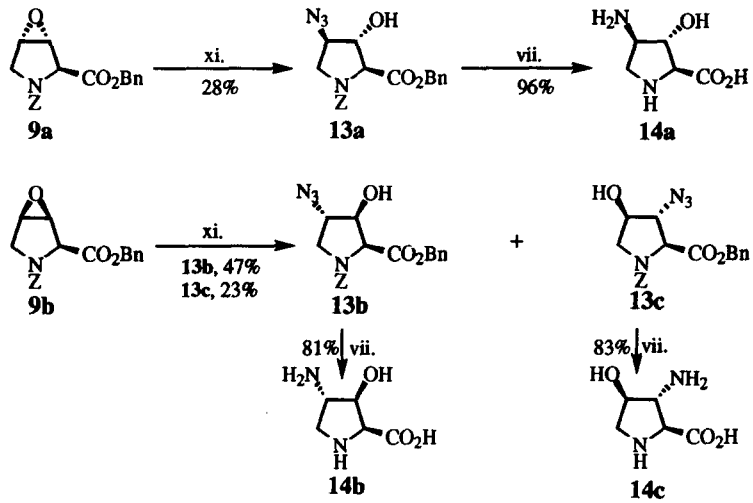
However, due to the absence of cross ring interactions in the n.O.e. studies on **9a** or **9b** their assignments were regarded as provisional. Similarly, n.O.e. studies on **2a** and **2b** failed to unequivocally confirm their relative sterochemistries. Thus synthetic structural correlation studies were carried out (**Scheme 2**).



Scheme 2: (vii) cat. OsO_4 / *N*-methylmorpholine-*N*-oxide / ^tBuOH / H_2O; (viii) 2-acetoxyisobutyryl bromide / CH_3CN; (ix) K_2CO_3 / $PhCH_2OH$; (x) H_2 / Pd / THF / H_2O.

Reaction of **8** with *N*-methylmorpholine-*N*-oxide and catalytic OsO_4[16] in ^tBuOH gave a mixture of diols **10a and 10b**, with the former predominating [**10a**:**10b**, >10:1 by 500 MHz ^1H NMR analysis]. When acetone was used as the reaction solvent, instead of ^tBuOH, a conspicuous amount of black precipitate formation was observed and the mixture of diols (**10a/b**) was isolated in a lower yield and less pure form. Deprotection by hydrogenolysis (H_2/ Pd/ C) of mixture (**10a/b**) gave mainly amino acid **11a** together with a small amount (*ca.* 4 %, by 500 MHz ^1H NMR analysis) of its (3*S*, 4*R*)-diastereomer **11b**. **11a** is a constituent of an adhesive protein from *Mytilus edulis*[17]. Spectroscopic data for amino acid **11a** [contaminated with small amount of **11b**] was consistent with that previously reported for its enantiomer as synthesised by Fleet

et al.[18, 19] Furthermore, diols mixture (**10a/b**) were converted in two steps, to a single isolated epoxide **9a**. Subsequent treatment of (**10a/b**) with 2-acetoxyisobutyryl bromide[20, 21] in acetonitrile gave a mixture of regioisomers (**12a/b**), which when stirred with K_2CO_3 in benzyl alcohol gave epoxide **9a**, identical to that previously prepared from m-CPBA epoxidation of dehydroproline **8**. Benzyl-*N*-benzyloxycarbonylpyrrole-2-carboxylate (14%) and unreacted starting material (17%) were also isolated. These chemical correlation studies confirmed the assignment of **9a** and by implication **9b**, **2a** and **2b**.



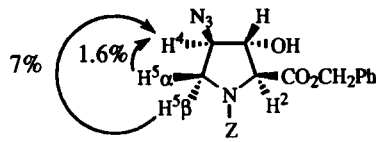
Scheme 3: (xi) NaN_3 / NH_4Cl / acetone / H_2O; (vii) H_2/ Pd / C / THF / H_2O

The synthetic potential of the epoxyproline derivatives **9a** and **9b** was also exemplified by the synthesis of three hydroxyaminoprolines (**Scheme 3**). Ring opening of the epoxides **9a** or **9b** was achieved using NaN_3 in the presence of NH_4Cl.[22] A single product **13a** was isolated from reaction of the *trans*-epoxide **9a** after chromatography. In contrast *cis*-epoxide **9b** gave two products which were separated by flash chromatography. The azidohydroxyproline derivatives **13a**, **13b** and **13c** were deprotected by catalytic hydrogenation to give hydroxyaminoproline **14a**, which has also been prepared by Herdeis *et al.*[7] **14b** and **14c**, respectively.

The regiochemistry of azides **13a**, **13b** and **13c** were established by ^1H COSY 2-D NMR. Observed coupling constants for the H-2 and H-3 hydrogens supported the depicted stereochemcial assignments: for **13a**, $J_{2,3}$ = 3 Hz suggesting a trans stereo-relationship; for **13b**, $J_{2,3}$ = 7 Hz suggesting a *cis*-stereo-relationship; and for **13c**, $J_{2,3}$ = 1.5 Hz suggesting a *trans* stereo-relationship. In each case two conformational isomers were clearly present. The depicted stereochemcial assignments were supported by both conventional n.O.e difference and plused field gradient n.O.e. experiments. The n.O.e. studies of **13a** were conducted in both C_6D_6/acetone-d^6 (1:1) and $CDCl_3$ since no single NMR solvent allowed for complete resolution of all its hydrogen resonances in the pyrrolidine ring.

For **13a**, in C_6D_6/acetone-d^6 (1:1), irradiation of the lower frequency H-5 hydrogen caused a 1.6% enhancement of the H-4 resonance, whereas a 7.0% enhancement in the H-4 resonance was observed when

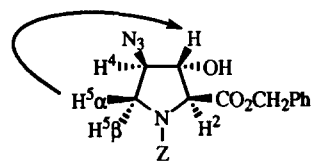
the high frequency H-5 resonance was irradiated. This result implies the lower and higher frequency resonating H-5 hydrogens are *syn* and *anti* relative to the azide respectively **(Figure 2)**.



13a
(in C_6D_6/acetone-d_6 1:1)
Selected n.O.e data for **13a**

Figure 2

Double pulsed field gradient spin echo n.O.e. studies[23, 24, 25] on **13a** (in $CDCl_3$) revealed that H-3 signal enhancement was observed when H-5α resonance was selectively inverted, confirming the weak enhancement as depicted **(Figure 3)**.



13a
(in $CDCl_3$)
DPFGSE- n.O.e. for **13a**

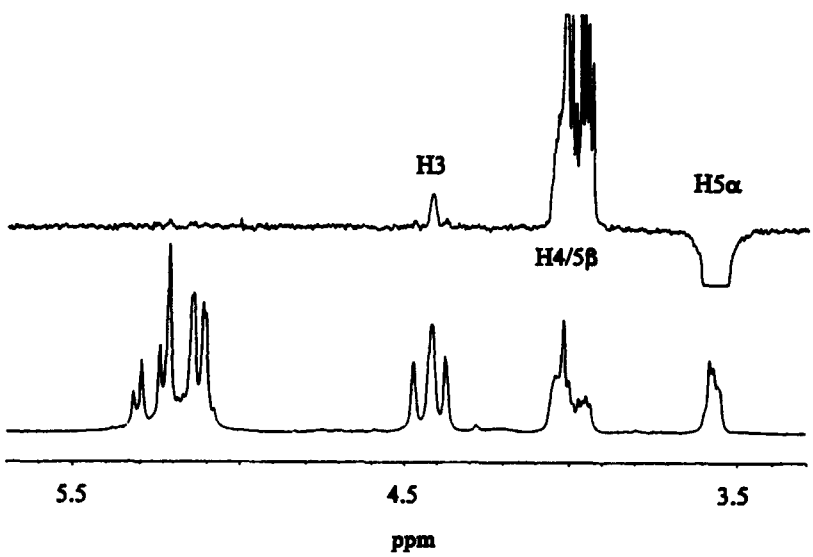
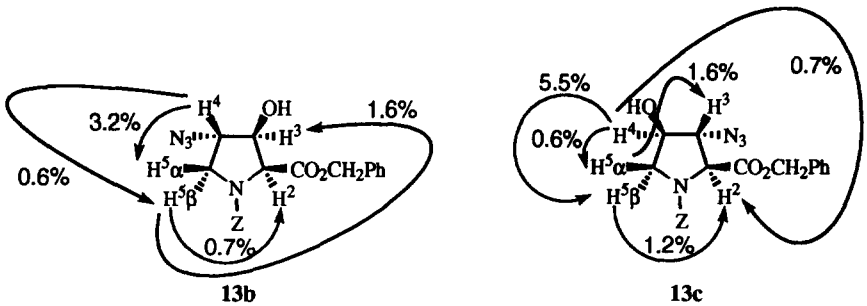


Figure 3

For **13b** irradiation of the H-5β resonance caused 0.7% and 1.6% signal enhancements for the H-2 and H-3 resonances, respectively. Signals enhancements of 3.2% and 0.6% for H-5α and H-5β, respectively, were observed when H-4 resonance was irradiated (**Figure 4**).



13b **13c**

Selected n.O.e. data for **13b** and **13c**

Figure 4

For **13c**, irradiation of the H-5β resonance produced a 1.2% signal enhancement of the H-2 resonance. Signal enhancement of 0.7%, 0.6% and 5.5% were observed for the H-2, H-5α and H-5β resonances, respectively when H-4 was irradiated. Irradiation of the H-5α resonance caused a 1.6% signal enhancement in the H-3 resonance (**Figure 4**).

In summary, we have described routes to both *trans* -**2a** and *cis*-epoxides **2b**, the former of which has been prepared in small amounts using proline 4-hydroxylase. The work of Herdeis[7] and the results presented herein demonstrate that protected versions of these epoxides may be useful synthetic building blocks. They should help to stimulate work directed towards more efficient preparations of these epoxides and related functionalised prolines, possibly *via* catalysis using modified proline hydroxylases.

General Experimental

Infrared spectra were recorded on a Perkin-Elmer 1750 Fourier Transform spectrometer with only selected absorbencies being recorded. Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on either a Varian Gemini 200 operating at 200 MHz or a Brüker AM500 operating at 500 MHz. The spectra were referenced to residual protonated solvent residues, for example, $CHCl_3$ δ_H = 7.27 p.p.m., as an internal standard. Carbon-13 nuclear magnetic resonance (^{13}C NMR) spectra were recorded on either a Varian Gemini 200 operating at 50.3 MHz or a Brüker AM500 operating at 125.8 MHz. The spectra were referenced to solvent carbon residues or 1,4-dioxane, δ_C = 67.3 p.p.m, for samples in deuterium oxide. DEPT editing was used for spectra obtained at 50.3 MHz. All chemical shifts (δ_H, δ_C) are quoted in parts per million (p.p.m.), in the solvent indicated in parentheses. N.O.e. experiments were performed on a Bruker AMX500 equipped with an inverse broadband gradient probehead. Difference experiments utilised decoupler frequency cycling within each multiplet for improved selectivity[26] with total saturation periods of 8 s (80 cycles of 100 ms saturation). Difference FIDs were processed with 3 Hz line broadening and the resulting

spectra were baseline corrected prior to integration. Double pulsed field gradient spin echo n.O.e. experiments used a 40 ms 180° Gaussian pulse for selective inversion of the target multiplet and plused field gradients of 8.5:8.5:5.5:5.5 G cm^{-1} within the double spin-echo. A single non-selective 180° inversion pulse bracketed by gradients of 4:-4 Gcm^{-1} were placed at the midpoint of the 1 s mixing time. 128 transients were collected within a recycle delay of 2s. Mass spectra were recorded on either a V. G. 20-250 or a BIO-Q instruments, with the ionisation modes used were Desorption Chemical Ionisation (DCI), Probe Chemical Ionisation (CI) or Direct Electron Impact (DEI). Only major peaks are reported as their mass-to-charge (*m/z*) ratios. Accurate masses were recorded by the EPSRC mass spectrometry service centre (University of Wales, Swansea). Optical rotations were determined using a Perkin-Elmer 241 polarimeter with concentrations given in g/100ml. Melting points (m.p.) were determined using a Büchi 510 capillary melting point apparatus and are uncorrected.

Thin layer chromatography was performed on Merck DC-Alufólien 60F254 0.2mm precoated plates. Spots were detected by quenching of ultraviolet fluorescence (λ_{max} 254nm) or 5%w/v dodecamolybdophosphoric acid in ethanol followed by heat. Amino acids were located on t.l.c. by 3%w/v ninhydrin in ethanol. Flash chromatography was carried out on Baker silica gel (30-60 mm, pore diameter 6nm). Reagents were used as obtained from commercial sources except for benzyl bromide which was passed through neutral alumina before use. Toluenesulphonyl chloride was purified by dissolving it in the minimum volume of chloroform and adding petroleum ether to precipitate impurities which were filtered off. Light petroleum (PE) refers to that fraction of petroleum ether which boils between 40-60°C, which was distilled before use, as were ethyl acetate, diethyl ether, benzene, dichloromethane, 1,2-dichloroethane and water. Tetrahydrofuran was distilled from sodium benzophone ketyl under an atmosphere of nitrogen, *N*-methylimidazole and *tert*-butanol were dried by distillation from calcium hydride and stored over activated (flame dried) 4 Å molecular sieves under argon. Ion exchange chromatography was carried out using Dowex® 50W-X8(H) resin which was pre-equilibrated using 3 M HCl (aq). The amino acid products were eluted from the resin with 3 M NH_3 (aq).

(2*S*, 4*R*)-*N*-Benzyloxycarbonyl-4-hydroxyproline 4[4]

To a cooled solution (ice/water bath) of (2*S*, 4*R*)-hydroxyproline (11.61g, 88.5mmol) in 1M NaOH(aq) (88.5ml) and THF (88.5ml) was added dropwise a solution of benzyl chloroformate (16.8ml, 111.8mmol) in THF (88.5ml) and a solution of 1M NaOH(aq) (115ml) simultaneously. The mixture was allowed to reach room temperature and stirred overnight. The reaction mixture was diluted with water (100ml) and washed with diethyl ether (3 x 75ml). The aqueous layer was acidified to pH 3 with 6M HCl(aq) (25ml) and extracted with EtOAc (3 x 75ml). The combined organic extracts were washed with water (50ml), brine (sat., 50ml) and dried (Na_2SO_4). The solvent was removed *in vacuo* to give a colourless oil which crystallised from ether/petrol to give **4** as a white solid (20.1g, 86%), m.p. 104.5-105°C (from ether/PE), (lit.[4] m.p. 106-107°C (from EtOAc/petrol)); $[\alpha]_D^{24}$ -94.1 (c 0.76 in $CHCl_3$); Found C 59.00, H 5.57, N 5.16 %, $C_{13}H_{15}NO_5$ requires C 58.86, H 5.70, N 5.28 %; ν_{max}/cm^{-1} (nujol) 3399(br, O-H), 2950(m), 1754(s, acid C=O), 1685(s, urethane C=O), 1427(m), 1173(m); δ_H(200 MHz, $CDCl_3$) 2.05-2.60 (2H, m, H-3), 3.50-3.75 (2H, m, H-5), 4.40-4.60 (2H, m, H-2 + H-4), 4.95-5.40 (2H, m, $PhCH_2O$-), 7.10-7.55 (5H, m, Ar-H); δ_C (50.3 MHz,

acetone-d6) (2 conformational isomers) 38.2 and 39.2 (C-3), 54.6 and 55.0 (C-5), 57.6 and 58.0 (C-2), 66.4 ($PhCH_2O$-), 68.7 and 69.4 (C-4), 127.5, 127.9, 128.0, 128.5 and 128.6 (ArC-H), 137.4 (ArC_{ipso}), 154.6 and 155.1 (C=O urethane), 173.58 and 173.96 (C=O acid); [*m/z* CI (NH_3)] 283 (MNH_4^+,25), 266 (MH^+,100), 222 (MH^+-CO_2,55), 132 (90), 108 (38), 91 (53).

(2*S*, 4*R*)-*N*-Benzyloxycarbonyl-4-hydroxyproline benzyl ester 5

To a solution of (2*S*, 4*R*)-*N*-benzyloxycarbonyl-4-hydroxyproline **4** (4.45g, 16.8mmol) in DMF (30ml) under an atmosphere of nitrogen was added anhydrous potassium carbonate (4.64g, 33.6mmol), sodium iodide (0.27g, 1.80mmol) and benzyl bromide (6.0ml, 50.4mmol). The mixture was stirred overnight. The reaction mixture was diluted with water (80ml) and extracted with EtOAc (3 x 60ml). The combined organic extracts were washed with water (2 x 30ml), brine (sat., 30ml) and dried (Na_2SO_4). The solvent was removed *in vacuo* and the crude product was purified by flash chromatography (PE/EtOAc 2:3) to give the desired product as a colourless oil (4.21g, 71%); R_f 0.29 (PE/EtOAc 2:3); $[\alpha]_D^{24}$ -70.0 (c 0.75 in $CHCl_3$); Found C 67.37, H 5.79, N 3.98 %, $C_{20}H_{21}NO_5$ requires C 67.59, H 5.96, N 3.94 %; ν_{max}/cm^{-1} (film) 3450(br, O-H), 2952(m), 1747(s, C=O ester), 1705(s, C=O urethane), 1420(m), 1358(m), 1190(m); δ_H(200 MHz, $CDCl_3$) 1.90-2.35 (2H, m, H-3+ OH), 3.40-3.70 (2H, m, H-5), 4.30-4.70 (2H, m, H-2 + H-4), 4.83-5.23 (4H, m, $PhCH_2O$-), 7.04-7.45 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 38.3 and 39.1 (C-3), 54.6 and 55.2 (C-5), 57.8 and 58.0 (C-2), 66.9, 67.3, 67.0 ($PhCH_2O$), 69.3 and 70.1 (C-4), 128.0, 128.2, 128.3, 128.6, 128.8 (ArC-H), 135.5 and 135.8 (ArC_{ipso}), 136.4 (ArC_{ipso}), 154.9 and 155.4 (urethane C=O), 172.7 and 172.9 (ester C=O); [*m/z* CI (NH_3)] 373 (MNH_4^+, 8), 356 (MH^+, 67), 312 (MH^+-CO_2, 15), 220 (26), 176 (30), 108 (56), 91 (100).

(2*S*, 4*R*)-*N*-Benzyloxycarbonyl-4-*para*-toluenesulphonylimidazoleproline benzylester 6

To a solution of *para*-toluenesulphonylimidazole[5] (3.52g, 15.8mmol) in THF (20ml) under an atmosphere of nitrogen at 0°C was added methyl triflate (1.8ml, 15.9mmol). The mixture was stirred for 30 minutes. A solution of (2*S*, 4*R*)-*N*-benzyloxycarbonyl-4-hydroxyproline benzyl ester **5** (3.75g, 10.6mmol) and *N*-methylimidazole (1.3ml, 16.0mmol) in THF (10ml) was added to the reaction mixture which was allowed to reach room temperature and stirred overnight at 30°C. Water (50ml) was added and the mixture extracted with EtOAc (3 x 45ml). The combined extracts were washed successively with $KHSO_4$(aq) (5%, 30ml), $NaHCO_3$(aq) (sat., 30ml), brine (sat., 30ml), dried (Na_2SO_4) and the solvent removed *in vacuo*. The crude product was purified by flash chromatography (PE/EtOAc 1:1) to give **6** as a colourless oil (5.06g, 94%) which gradually changed into a semi-solid; R_f 0.54 (PE/EtOAc 1:1), $[\alpha]_D^{24}$ -33.7 (c 0.77 in $CHCl_3$); ν_{max}/cm^{-1} (film) 3034(m), 2954(m), 1748(s, C=O ester), 1713(s, C=O urethane), 1598(m), 1498(m), 1417(m), 1359(m), 1190(m); δ_H(200 MHz, $CDCl_3$) 2.01-2.30 (1H, m, H-3), 2.40-2.70 (4H, m, H-3 + Ar-CH_3), 3.55-3.85 (2H, m, H-5), 4.51 (1H, *ca.* quartet, *J* 8.0 Hz, H-2), 4.90-5.30 (4H, m, $PhCH_2O$-), 7.20-7.50 (12H, m, Ar-CH), 7.65-7.90 (2H, m, Ar-CH); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 21.6 (CH_3), 36.0 and 37.1 (C-3), 52.1 and 52.5 (C-5), 57.3 and 57.6 (C-2), 67.1, 67.2 and 67.5 ($PhCH_2O$-), 78.2 and 78.9

(C-4), 127.9, 128.1, 128.3, 128.7, 128.8 (ArC-H), 130.3, 133.4 (ArC-CH_3), 133.5, 135.3, 135.4, 135.6, 136.3, (ArC_{ipso}), 145.6 (ArC-SO_2-), 154.2 and 154.7 (urethane C=O), 171.8 and 172.0 (ester C=O); [*m/z* DCI (NH_3)] 527 (MNH_4^+,12), 510 (MH^+,23), 466 (MH^+-CO_2,10), 356 (36), 108 (60), 91 (100); Accurate mass : MH^+ requires 510.1585, found 510.1590.

(2*S*, 4*S*)-*N*-Benzyloxycarbonyl-4-phenylselenoproline benzyl ester 7

To a solution of diphenyldiselenide (1.30g, 4.16mmol) in ^{t}BuOH (10ml) under an atmosphere of nitrogen was added $NaBH_4$ (0.305g, 8.06mmol). The mixture was heated under reflux until the yellow colour disappeared. A solution of tosylate **6** (3.33g, 6.53mmol) in ^{t}BuOH (10ml) was added and the reflux was continued for 2.5 hours. The mixture was diluted with EtOAc (50ml), washed with water (3 x 40ml), then dried (Na_2SO_4) and the solvent removed *in vacuo*. Purification of the crude product by flash chromatography (PE/EtOAc 4:1) gave **7** as a colourless oil (2.39g, 74%); R_f 0.24 (PE/EtOAc 4:1); $[\alpha]_D^{24}$ -58.8 (c 0.41 in $CHCl_3$); Found C 63.48, H 5.09, N 2.67 %, $C_{26}H_{25}NO_4Se$ requires C 63.17, H 5.10, N 2.83 %; ν_{max}/cm^{-1} (film) 3033(m), 2952(m), 1749(s, C=O ester), 1708(s, C=O urethane), 1579(m), 1415(s), 1355(s), 1113(m), 1080(m); δ_H(200 MHz, $CDCl_3$) 2.00-2.22 (1H, m, H-3), 2.60-2.82 (1H, m, H-3), 3.42-3.75 (2H, m, H-5), 3.92-4.15 (1H, m, H-4), 4.30-4.55 (1H, m, H-2), 4.90-5.35 (4H, m, 2xPhC$\underline{H}_2$O), 7.05-7.60 (15H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 36.2 and 36.8 (C-4), 36.8 and 37.7 (C-3), 52.8 and 53.3 (C-5), 58.8 and 59.0 (C-1), 67.0, 67.1 and 67.2 (2xPh$\underline{C}H_2$O), 128.1, 128.2, 128.3, 128.6 and 128.7 (ArC-H), 128.8, 129.5, 135.3 and 135.5 (ArC_{ipso}), 136.6 and 136.7 (ArC_{ipso}), 154.2 and 154.8 (C=O urethane), 172.3 and 172.5 (ester); [*m/z* DCI(NH_3)] 496 (MH^+,34), 450 (8), 360 (37), 316 (75).

(2*S*)-*N*-Benzyloxycarbonyl-3,4-dehydroproline benzyl ester 8

To a stirred solution of selenide **7** (2.25g, 4.61mmol) and pyridine (0.50ml, 6.18mmol) in dichloromethane (25ml) at 0°C was added H_2O_2 (30%, 1.17ml, 11.5mmol) dropwise. The mixture was allowed to reach room temperature and stirred for a further 1.5 hours. Dichloromethane (25ml) was added and the mixture washed successively with $KHSO_4$(aq) (5%, 2 x 30ml), $NaHCO_3$(aq) (sat., 30ml) and water (2 x 30ml). The organic layer was dried (Na_2SO_4), the solvent removed *in vacuo* and the residue was purified by flash chromatography (PE/EtOAc 3:2) to give **8** as a colourless oil (1.20g, 77%). The product crystallised from ether/petrol to give **8** as a white solid, m.p. 44.5-45.5°C (from ether/petrol); R_f 0.40 (PE/EtOAc 3:2); $[\alpha]_D^{23}$-95.6 (c 0.74 in $CHCl_3$); Found C 71.09, H 5.48, N 4.14%, $C_{20}H_{19}NO_4$ requires C 71.20, H 5.68, N 4.15%; ν_{max}/cm^{-1} (film) 3033(m), 2869(m), 1750(s, C=O ester), 1708(s, C=O urethane), 1622(m), 1418(s), 1355(s), 1175(s) 1082(s), 754(s), 698(s); δ_H(200 MHz, $CDCl_3$) 4.25-4.45(2H, m, H-5), 4.95-5.35 (4H, m, $PhCH_2O$-), 5.70-5.85 (1H, m, H-3 or H-4), 5.90-6.10 (1H, m, H-3 or H-4), 7.10-7.50 (10H, m, Ar-H); δ_C(50.3MHz, $CDCl_3$) (2 conformational isomers) 53.4 and 53.9 (C-5), 66.4 and 66.8 (C-2), 67.0,and 67.2 (Ph$\underline{C}H_2$O), 124.8 (C-3 and C-4), 127.1, 128.2, 128.3, 128.5, 128.7, 128.8 and 129.5 (ArC-H), 135.6 and 135.8 (ArC_{ipso}), 136.6 and 136.8 (ArC_{ipso}), 154.7 and 154.2 (urethane C=O), 170.1 and 170.4 (ester C=O); [*m/z* CI(NH_3)] 355 (MNH_4^+,13), 338 (MH^+,95), 294 (MH^+-CO_2,15), 202 (28), 158 (38), 108 (36), 91 (100).

(2*S*, 3*R*, 4*R*)-*N*-Benzyloxycarbonyl-3, 4-epoxyproline benzyl ester 9a and (2*S*, 3*R*, 4*S*)-*N*-Benzyloxycarbonyl-3, 4-epoxyproline benzyl ester 9b

To a solution of olefin **8** (1.00g, 2.96mmol) in 1,2-dichloroethane was added m-CPBA (50%, 1.23g, 3.6mmol) and 4,4'-thio-(6-*tert*-butyl-3-methylphenol) (65mg, 0.18mmol). The mixture was refluxed under an atmosphere of nitrogen for 4 hours and then a further batch of m-CPBA (0.434g, 1.26mmol) and 4,4'-thio-(6-*tert*-butyl-3-methylphenol) (35mg, 98μmol) was added. Refluxing was continued overnight. The reaction mixture was diluted with dichloromethane (30ml), washed with sodium metabisulphate (aq) (5%, 2 x 25ml), saturated $NaHCO_3$ (aq) (3 x 25ml), brine (sat., 25ml), dried (Na_2SO_4) and the solvent removed *in vacuo*. The diastereomeric epoxides were separated by flash chromatography (PE/EtOAc 4:3) to give **9a** (0.57g, 54%) and **9b** (0.23g, 22%) as colourless oils. **9b** crystallised from ether/petrol, forming a white solid.

9a

R_f 0.53 (PE/EtOAc 4:3); $[\alpha]_D^{23}$ -93.6 (c 0.41 in $CHCl_3$); ν_{max}(film)/cm^{-1} 2957(m), 1753(s, C=O ester), 1713(s, C=O urethane), 1585(m), 1425(s), 980(s), 850(s), 747(s), 630(s); δ_H(500 MHz, $CDCl_3$) (2 conformational isomers) 3.59 and 3.61 (1H, dd, J_1 12.5Hz, J_2 1.0 Hz, H-5α), 3.70 and 3.73 (1H, dd, J_1 3.0 Hz, J_2 1.0 Hz, H-4), 3.80 (1H, d, J 3.0 Hz, H-3), 3.95 and 4.00 (1H, d, J 12.5 Hz, H-5β), 4.72 and 4.81 (1H, s, H-2), 5.05-5.35 (4H, m, Ph-C$\underline{H_2}$-O), 7.20-7.45 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 47.1 and 47.4 (C-5), 54.2 and 54.8 (C-3 or C-4), 56.5 and 57.2 (C-3 or C-4), 60.6 and 60.8(C-2), 67.4, 67.5 (2xPh$\underline{C}H_2$O), 128.0, 128.2, 128.4, 128.7 and 128.9 (ArC-H), 133.1, 135.2 and 135.3 (ArC_{ipso}), 136.3 (ArC_{ipso}), 155.0 and 155.5 (C=O urethane), 169.1 (C=O ester); [*m/z* CI(NH_3)] 371 (MNH_4^+,30), 354 (MH^+,94), 310 (MH^+-CO_2,50), 174 (24), 108 (28), 91 (100); Accurate mass : MH^+ requires 354.1340, found 354.1341.

9b

m.p. 128.5-129°C (from ether/petrol); R_f 0.25 (PE/EtOAc 4:3); $[\alpha]_D^{24}$ -21.3 (c 0.75 in $CHCl_3$); Found C 68.06, H 5.29, N 3.82%, $C_{20}H_{19}NO_5$ requires C 67.98, H 5.42, N 3.96%; ν_{max}/cm^{-1} (film) 2951(m), 1762(s, C=O ester), 1713(s, C=O urethane), 1499(m), 1425(s), 1174(s), 1124(s), 988(m), 861(m), 752(s), 669(s); δ_H(500 MHz, $CDCl_3$) 3.59 and 3.63 (1H, dd, J_1 12.5 Hz, J_2 2.0Hz, H-5β), 3.78 and 3.80 (1H, t, J 2.5 Hz, H-4), 3.90 and 3.92 (1H, d, J12.5 Hz, H-5α), 3.97 (1H, br s, H-3), 4.45 and 4.51 (1H, d, J2.5 Hz, H-2), 4.92-5.35 (4H, m, 2xPh-C$\underline{H_2}$-O), 7.20-7.40 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 47.8 and 48.3 (C-5), 55.7 and 56.1 (C-3 or C-4), 57.3 and 58.1 (C-3 or C-4), 60.1 and 60.3 (C-2), 67.2 and 67.4 (Ph$\underline{C}H_2$O), 128.1, 128.2, 128.4, 128.5 and 128.8 (ArC-H), 135.6 and 135.7 (ArC_{ipso}), 136.3 and 136.4 (ArC_{ipso}), 154.7 and 155.2 (C=O urethane), 167.4 and 167.9 (C=O ester); [*m/z* CI(NH_3)] 371 (MNH_4^+,14), 354 (MH^+,100), 310 (MH^+-CO_2,34), 174 (25), 108 (27), 91 (95).

(2*S*, 3*R*, 4*R*)-3,4-Epoxyproline (2a) and (2S, 3S, 4S)-3,4-Epoxyproline 2b

To a solution of epoxide **9a** (0.15g, 0.42mmol) or **9b** (0.10g, 0.28mmol) in THF (15ml) and water (5ml) was added palladium on charcoal catalyst (10%, 50mg). The mixture was left to stir under hydrogen at

atmospheric pressure for 3 hours. After filtration through cellulose to remove the catalyst the solvent was evaporated to give **2a** (54mg, 100%), and **2b** (30mg, 83%) respectively as white solids.

2a

$[\alpha]_D^{23}$ -109.2 (c 0.25 in H_2O); ν_{max}/cm^{-1} (KBr disc) 2980(br), 1632(s, C=O), 1411(s), 1372(s), 1268(m), 1079(m), 854(m); δ_H(200 MHz, D_2O) 3.32 (1H, d, *J* 13.0 Hz, H-5), 3.47 (1H, d, *J* 13.0 Hz, H-5), 3.81 (1H, d, *J* 3.0 Hz, H-4), 3.91 (1H, d, *J* 3.0 Hz, H-3), 4.21 (1H, *ca.*s, H-2); δ_C(50.3 MHz, D_2O) 45.3 (C-5), 53.8 (C-3 or C-4), 56.4 (C-3 or C-4), 61.4 (C-2), 169.4 (CO_2H); [*m/z* DCI(NH_3)] 147 (MNH_4^+,3), 130 (MH^+,52), 129 (30), 111 (72), 94 (35), 69 (100); Accurate mass : MH^+ requires 130.0504, found 130.0504.

2b

$[\alpha]_D^{24}$ -14.3 (c 0.99 in H_2O); ν_{max}/cm^{-1} (KBr disc) 3431(br), 1625(s, C=O), 1397(s); δ_H(200 MHz, D_2O) 3.27 (1H, d, *J* 13.0 Hz, H-5), 3.52 (1H, d, *J* 13.0 Hz, H-5), 3.83 (1H, d, *J* 3.0 Hz, H-4), 3.96 (1H, d, *J* 3.0 Hz, H-3), 4.10(1H, *ca.*s, H-2); δ_C(50.3 MHz, D_2O) 45.7 (C-5), 54.9 (C-3 or C-4), 56.5 (C-3 or C-4), 61.0(C-2), 169.9 (CO_2H); [*m/z* DCI(NH_3)] 151 (MNa^+-H, 17), 140 (10), 111 (66), 94 (27), 68 (100), 67 (91); Accurate mass : MH^+ requires 130.0504, found 130.0504.

(2*S*, 3*R*, 4*R*)-*N*-Benzyloxycarbonyl-3,4-dihydroxyproline benzyl ester (10a/b)

To a solution of (2*S*)-*N*-benzyloxycarbonyl-3,4-dehydroproline benzyl ester **8** (0.25g, 0.74mmol) and *N*-methylmorpholine-*N*-oxide (83mg, 0.61mmol) in ^{t}BuOH/water (7ml:3ml) was added a catalytic amount of osmium tetroxide. The solution was left to stir overnight. The reaction was quenched by the addition of 5% sodium metabisulpate (aq) (30ml). The mixture was stirred for 30 minutes and then extracted with EtOAc (3 x 20ml). The combined organic extracts were washed with 5% $KHSO_4$(aq) (2 x 15ml), brine (sat., 15ml), dried (Na_2SO_4) and the solvent removed. Flash chromatography (PE/EtOAc 3:7) gave the product as a colourless oil (0.22g, 80%); R_f 0.23 (PE/EtOAc 3:7); $[\alpha]_D^{20}$ +7.8 (c 1.02 in $CHCl_3$); Accurate mass : MH^+ requires 372.1445, found 372.1447; ν_{max}/cm^{-1} (film) 3419(br, O-H), 2979(m), 1740(s, ester C=O), 1708(s, urethane C=O), 1500(m), 1424(s), 1360(s), 1161(s); δ_H(500 MHz, $CDCl_3$) 2.62 and 2.65 (1H, d, *J* 5.0Hz, OH), 2.96 and 2.99 (1H, d, *J* 5.0Hz, OH), 3.53 and 3.61 (1H, dd, J_1 4.5Hz, J_2 11.5Hz, H-5), 3.72-3.80 (1H, m, H-5), 4.20-4.40 (3H, m, H-2 + H-3 + H-4), 4.98-5.30 (4H, m, $PhCH_2O$-), 7.16-7.44 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (conformational isomers) 50.6, 50.8, 50.9 and 51.1 (C-5), 64.5, 64.8, 65.5 and 65.8 (C-2), 67.2, 67.4 and 67.5($PhCH_2O$), 69.8 and 70.5 (C-3 or C-4), 74.8 and 75.8(C-3 or C-4), 128.0, 128.3, 128.6 and 128.8 (ArC-H), 135.4 and 135.5 (ArC_{ipso}), 136.2, 136.3 and 136.5(ArC_{ipso}), 155.0, 155.2 and 155.5 (C=O urethane), 170.6, 170.7, 171.4 and 171.6 (C=O ester); [*m/z* CI(NH_3)] 372 (MH^+,4), 355 (10), 338 (42), 299 (57), 282 (100), 238 (40), 192 (32), 108 (31), 91 (72).

(2*S*, 3*R*, 4*R*)-Dihydroxyproline (11a/b)

To a solution of diol **(10a/b)** (0.15g, 0.40mmol) in THF (10ml) and water (5ml) was added palladium on charcoal catalyst (10%, 90mg). The mixture was left to stir overnight under hydrogen at atmospheric pressure. After filtration to remove the catalyst and evaporation of solvent *in vacuo* the crude product was purified by ion exchange to give **(11a/b)** (52mg, 88%) as a white solid; $[\alpha]_D^{20}$ +4.8 (c 0.50 in H_2O); Accurate mass : MH^+ requires 148.0609, found 148.0610; vmax/cm-1 (KBr disc) 3398, 3122(br), 1602(s, C=O), 1409(m), 1372(m), 1333(m), 1139(m), 1102(m); δ_H(500 MHz, D_2O) 3.13 (1H, dd, J_1 4.5 Hz, J_2 12.5 Hz, H-5), 3.37 (1H, dd, J_1 5.0 Hz, J_2 12.5 Hz, H-5) 3.81 (1H, d, J 5.0 Hz, H-2), 4.10-4.25 (2H, m, H-3 + H-4); δ_C(125 MHz, D_2O) 51.2 (C-5), 67.3 (C-2), 72.7 (C-3 or C-4), 76.9 (C-3 or C-4), 174.7 (CO_2H); [*m/z* CI(NH_3)] 148 (MH^+,89), 102 (37), 84 (6).

(2*S*, 3*R*, 4*S*)-*N*-Benzyloxycarbonyl-3-acetyl-4-bromoproline benzyl ester and (2*S*, 3*S*, 4*R*)-*N*-Benzyloxycarbonyl-4-acetyl-3-bromoproline benzyl ester (12a/b)

To a stirred solution of (2*S*, 3*R*, 4*R*)-*N*-benzyloxycarbonyl-3, 4-dihydroxyproline benzyl ester **(10a/b)** (0.30g, 0.81mmol) in acetonitrile (10ml) was added acetoxyisobutyrl bromide (0.26ml, 1.77mmol) and the mixture was stirred at room temperature for 1 hour. Saturated $NaHCO_3$ (aq) (20ml) was added and the mixture was extracted with EtOAc (3 x 20ml). The combined organic extracts were washed with water (15ml), brine (sat., 15ml), dried (Na_2SO_4) and the solvent removed *in vacuo*. The crude product was purified by flash chromatography (PE/EtOAc 3:2) to give **(12a/b)** as a colourless oil, (0.29g, 76%); R_f 0.5 (PE/EtOAc 1:1); $[\alpha]_D^{24}$ -23.9 (c 4.9 in $CHCl_3$); Found C 55.71, H 4.48, N 2.88 %, $C_{22}H_{22}NO_6Br$ requires C 55.48, H 4.66, N 2.94 %; ν_{max}/cm^{-1} (film) 3033(m), 1752(s, C=O ester), 1713(s, C=O urethane), 1499(m), 1456(m), 1416(s), 1353(m), 1214(s), 1120(m), 1038(m), 753(s), 698(s); δ_H(200 MHz, $CDCl_3$) 2.09 (3H, s, CH_3-), 3.83-4.05 (1H, m, C<u>H</u>-Br), 4.05-4.35 (2H, m, H-5), 4.40-4.60 (1H, m, H-2), 5.00-5.33 (4H, m, 2xPhC<u>H</u>$_2$O-), 5.55-5.65 (1H, m, C<u>H</u>-OAc), 7.15-7.52 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 20.6 (CH_3), 53.6 and 53.8 (C-5), 63.4 and 63.6 (<u>C</u>H-Br), 67.5 and 67.7 (2xPh<u>C</u>H_2O), 80.3 and 81.3 (<u>C</u>H-OAc), 128.1, 128.4, 128.5 and 128.7 (ArC-H), 135.3 and 136.2 (ArC_{ipso}), 154.4 and 155.0 (C=O urethane), 169.6 and 170.4 (C=O ester). [*m/z* CI(NH_3)] 493 (MNH_4^+,3), 478/476 (MH^+,25), 434/432 (MH^+-CO_2,13), 336 (24), 292 (16), 91 (100).

(2*S*, 3*R*, 4*R*)-*N*-Benzyloxycarbonyl-3, 4-epoxyproline benzyl ester 9a from (12a/b)

To a solution of (2*S*, 3*R*, 4*R*)-*N*-benzyloxycarbonyl-3-acetyl-4-bromoproline benzyl ester and (2*S*, 3*S*, 4*R*)-*N*-benzyloxycarbonyl-4-acetyl-3-bromoproline benzyl ester **(12a/b)** (0.17g, 0.35mmol) in benzyl alcohol was added anhydrous K_2CO_3 (9mg, 0.35mmol) and the mixture was stirred at 50°C for 24 hours. Water (20ml) was added and the mixture extracted with EtOAc (3 x 15ml). The combined organic extracts were washed with water (15ml), brine (sat., 15ml) and dried (Na_2SO_4). The solvent was removed *in vacuo* to give

a mixture of starting material and epoxide **9a** which were separated by flash chromatography (PE/EtOAc 4:1), **9a** (27mg, 22%) and benzyloxycarbonylpyrrole-2-carboxylate benzyl ester (16mg, 14%).
Benzyloxycarbonylpyrrole-2-carboxylate benzyl ester
δ_H (200MHz, $CDCl_3$) 5.24-5.33 (4H, m. $PhCH_2O$), 6.18-6.22 (1H, m), 6.92-6.94 (1H, m), 7.27-7.39 (11H, m); [*m/z* CI(NH_3)] 353 (MNH_4^+, 5), 336 (MH^+, 100), 292 (25), 260 (25), 108 (40), 91 (50).

(2*S*, 3*R*, 4*R*)-*N*-Benzyloxycarbonyl-3-hydroxy-4-azidoproline benzyl ester 13a

To a stirred solution of (2*S*, 3*R*, 4*S*)-*N*-benzyloxycarbonyl-3, 4-epoxyproline benzyl ester **9a** (99mg, 0.28mmol) and NH_4Cl (35mg, 0.65mmol) in acetone/H_2O 8:1 (9ml) was added sodium azide (91mg, 1.40mmol) and the mixture heated at 70°C for 36 hours. The mixture was diluted with water (20ml) and extracted with EtOAc (3 x 20ml). The combined organic extracts were washed with water (20ml), brine (sat., 20ml) and dried (Na_2SO_4) and the solvent removed *in vacuo*. Flash chromatography (PE/EtOAc 3:2) gave **13a** as a colourless oil (31mg, 28%), (unreacted epoxide (28mg, 28%) was also recovered); R_f 0.35 (PE/EtOAc 3:2); $[\alpha]_D^{25}$ -43.6 (c 1.0 in $CHCl_3$); ν_{max}/cm^{-1} (film) 3418(br, O-H), 2955(m), 2110(s, azide), 1749(s, C=O ester), 1694(s, C=O urethane), 1424(s), 1313(s), 1212(m), 1120(m), 990(m); δ_H(500 MHz, C_6D_6/acetone-d^6) (2 conformational isomers) 3.59 and 3.65 (1H, dd, J_1 11.5Hz, J_2 4.5Hz, H-5α), 4.02 and 4.10 (1H, dd, J_1 11.5Hz, J_2 6.5Hz, H-5β), 4.15-4.22 (1H, m, H-4), 4.55-4.62 (m, H-3 + one of H-2 rotamer signal), 4.66 (*J*3.0Hz, part of H-2 rotamer signal), 5.10-5.40 (4H, m, 2x$PhC\underline{H}_2O$), 7.30-7.60 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 48.9 (C-5), 63.7 and 64.5 (C-4), 65.7 and 66.0 (C-2), 67.4, 67.5 and 67.7 ($Ph\underline{C}H_2O$), 77.5 and 78.6 (C-3), 128.0, 128.1, 128.3, 128.5, 128.7 and 128.8 (ArC-H), 135.3, 135.5 (ArC_{ipso}) and 136.2 (ArC_{ipso}), 154.9 and 155.4 (C=O urethane), 169.5 and 169.8 (C=O ester). [*m/z* CI(NH_3)] 414 (MNH_4^+,7), 397 (MH^+,13), 371 (57), 351 (100), 307 (13), 108 (40), 91 (60); Accurate mass : MH+ requires 397.1511, found 397.1512.

(2*S*, 3*S*, 4*S*)-*N*-Benzyloxycarbonyl-3-hydroxy-4-azidoproline benzyl ester 13b
and (2*S*, 3*R*, 4*R*)-*N*-Benzyloxycarbonyl-3-azido-4-hydoxyproline benzyl ester 13c

To a stirred solution of (2*S*, 3*S*, 4*R*)-*N*-benzyloxycarbonyl-3, 4-epoxyproline benzyl ester **9b** (97mg, 0.27mmol) and NH_4Cl (34mg, 0.64mmol) in acetone/H_2O 8:1 (9ml) was added sodium azide (93mg, 1.43mmol) and the mixture heated at 70°C for 24 hours. The mixture was diluted with water (20ml) and extracted with EtOAc (3 x 20ml). The combined organic extracts were washed with water (20ml), brine (sat., 20ml) and dried (Na_2SO_4) and the solvent removed *in vacuo* to give a mixture of **13b** and **13c** which were separated by flash chromatography (PE/EtOAc 3:2) on silica gel to give the products as colourless oils.

13b
(50mg, 47%); R_f 0.25 (PE/EtOAc 3:2); $[\alpha]_D^{25}$ +4.98 (c 3.0 in $CHCl_3$); ν_{max}/cm^{-1} (film) 3419(br, O-H), 2958(m), 2113(s, azide), 1738(s, C=O ester), 1708(s, C=O urethane), 1422(s), 1358(m), 1250(m), 1215(m),

755(s); δ_H(500 MHz, $CDCl_3$) 3.14 (1H, dd, J_1 4.5Hz, J_2 11.0Hz), 3.35-3.46 (1H, m), 3.88-3.99 (1H, m), 4.04-4.14 (1H, m), 4.34-4.42 (1H, m, H-3), 4.56 (1H, dd, J_1 7.0Hz, J_2 12.0Hz) 5.05-5.31 (4H, m, 2xPhCH_2O-), 7.26-7.36 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 48.2 and 48.3 (C-5), 62.4 and 62.5 (C-4), 63.4 and 64.0 (C-2), 67.1, 67.2 and 67.5 (PhCH_2O), 74.7 and 75.5 (C-3), 128.0, 128.1, 128.3 and 128.7 (ArC-H), 135.5, 136.2 and 136.3 (ArC_{ipso}), 154.4 (C=O urethane), 169.7 (C=O ester). [*m/z* DCI(NH_3)] 414 (MNH_4^+,18), 397 (MH^+,42), 351 (26), 108 (34), 91 (100); Accurate mass : MH^+ requires 397.1511 found, 397.1512.

13c

(25mg, 23%); R_f 0.20 (PE/EtOAc 3:2); $[\alpha]_D^{25}$ +11.5 (c 0.3 in $CHCl_3$); ν_{max}/cm^{-1} (film) 3428(br, O-H), 2954(m), 2110(s, azide), 1751(s, C=O ester), 1708(s, C=O urethane), 1422(s), 1357(m), 1260(m), 1191(s), 1119(m), 972(m), 752(m), 698(m); δ_H(500 MHz, $CDCl_3$) 2.84 and 2.88 (1H, d, *J* 8.0 Hz, O-H), 3.65 and 3.88 (1H, dd, J_1 12.0 Hz, J_2 2.0 Hz, H-5α), 3.80 and 3.82 (1H, dd, J_1 12.0Hz, J_2 5.0Hz, H-5β), 4.09 and 4.11 (1H, br. s, H-3), 4.17-4.24 (1H, m, H-4), 4.36 and 4.46 (1H, d, *J* 1.5 Hz, H-2), 5.03-5.35 (4H, m, 2xPhCH_2-O), 7.20-7.45 (10H, m, Ar-H); δ_C(50.3 MHz, $CDCl_3$) (2 conformational isomers) 52.7 and 52.9 (C-5), 63.2 and 63.5 (C-3), 67.8, 67.9 and 68.0 (2xPhCH_2O), 68.3 and 69.2 (C-2), 73.4 and 74.3 (C-4), 128.1, 128.4, 128.5 and 128.8 (ArC-H), 136.1 (ArC_{ipso}), 136.3 (ArC_{ipso}), 154.5 and 155.0 (C=O urethane), 170.9 (C=O ester). [*m/z* CI(NH_3)] 414 (MNH_4^+,3), 397 (MH^+,10), 351 (12), 108 (32), 91 (100); Accurate mass : MH^+ requires 397.1511, found 397.1512.

(2*S*, 3*R*, 4*R*)-3-hydroxy-4-aminoproline 14a, (2*S*, 3*S*, 4*S*)-3-hydroxy-4-aminoproline 14b, and (2*S*, 3*R*, 4*R*)-3-amino-4-hydoxyproline 14c

To a solution of **13a** (61mg, 0.15mmol), **13b** (63mg, 0.16mmol) or **13c** (56mg, 0.14mmol) in THF (10ml) and water (5ml) was added palladium on charcoal catalyst (10%, 15mg). The mixture was left to stir overnight under hydrogen at atmospheric pressure. After filtration to remove the catalyst the solvent was evaporated to give **14a** (21mg, 96%), **14b** (19mg, 81%) or **14c** (17mg, 83%) respectively as solids.

14a

$[\alpha]_D^{25}$ -4.7 (c 1.0 in H_2O); ν_{max}/cm^{-1} (KBr disc) 3428(br, s), 1635(s), 1397(m); δ_H(200 MHz, D_2O) 2.90-3.08 (1H, m, H-5), 3.08-3.45 (2H, m, H-4 + H-5), 3.64 (1H, d, *J* 3.5Hz, H-2), 4.03 (1H, t, *J* 3.0Hz, H-3); δ_C(50.3 MHz, D_2O) 48.8 (C-5), 56.6 (C-4), 66.8 (C-2), 78.6 (C-3), 173.4 (CO_2H); [*m/z* DCI(NH_3)] 147 (MH^+,25), 140 (29), 101 (68), 99 (28), 98 (30), 96 (38), 86 (47), 85 (28), 84 (75), 83 (100), 68 (40); Accurate mass : MH^+ requires 147.0769, found 147.0770.

14b

$[\alpha]_D^{25}$ -32.3 (c 0.55 in H_2O);ν_{max}/cm^{-1} (KBr disc) 3428(br, s), 1637(s), 1400(m), 1340(m), 1310(m),1000(m); δ_H(200 MHz, D_2O) 2.95 (1H, dd, J_1 1.5Hz, J_2 12.0Hz, H-5), 3.30-3.65 (2H, m, H-4 + H-5), 4.12 (1H, d, *J* 4.5Hz, H-2), 4.15-4.25 (1H, m, H-3); δ_C(50.3 MHz, D_2O) 49.6 (C-5), 57.1 (C-4), 65.1 (C-

2), 75.8 (C-3), 170.9 (CO_2H); [*m/z* DCI(NH_3)] 147 (MH^+,45), 101 (35), 86 (31), 84 (74), 83 (100), 68 (40); Accurate mass : MH^+ requires 147.0769, found 147.0770.

14c

$[\alpha]_D^{25}$ -20.9 (c 0.45 in H_2O); ν_{max}/cm^{-1} (KBr disc) 3402(br, s), 1632(s), 1397(m); δ_H(200 MHz, D_2O) 3.19 (1H, dd, J_1 2.5Hz, J_2 12.5Hz), 3.30-3.52 (2H, m), 3.63 (1H, d, J 3.5Hz, H-2), 3.95-4.10 (1H, m, H-4); δ_C(50.3 MHz, D_2O) 50.3 (C-5), 57.3 (C-3), 66.4 (C-2), 74.6 (C-4), 172.7 (CO_2H, tentative); [*m/z* DCI(NH_3)] 147 (MH^+, 89), 129 (29), 101 (32), 84 (34), 88 (42), 83 (100), 68 (30); Accurate mass : MH^+ requires 147.0769, found 147.0780.

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