Synthesis and X-ray structure of functionalised proline mimics

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Dedicated to Rod Rickards on the occasion of his 70th birthday

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Abstract

Methyl (2*R*)-(1-benzoyl-2-benzyl-1,2,3,6-tetrahydropyridin-2-yl)carboxylate **1** reacts with osmium tetroxide to give two isomeric *syn* diols **2a** and **2b** and also with Br_2 to give two isomeric *anti* dibromides **4a** and **4b**. The major dibromide **4a** gave rise to the lactone **5** on standing, the crystal structure of which is reported.

Keywords: Proline mimetic, crystal structure, lactone, diol, dibromide

Introduction

For some time, we[1-4] and others,[5-7] have had an interest in the design and preparation of analogues of proline of varying ring size and substitution. This interest stems from the fact that proline, and its hydroxylated derivatives, are known to induce secondary structure in peptides and proteins.[8] This induced secondary structure plays an important role in structures such as collagen,[9-11] an insoluble fibrous protein found in bone, teeth, blood vessels, connective tissue, tendons, cartilage, and hide. In addition, many bioactive peptides and natural products contain one or more functionalised proline, or proline-like, residues that are critical to their biological activity.[12,13] As a result, there has been much interest in the synthesis and functionalisation of proline analogues. However, of the many mimetics known few possess substitution to the α-carbon. With this in mind we prepared the ring-expanded and benzyl-substituted proline mimetic, methyl (2*R*)-(1-benzoyl-2-benzyl-1,2,3,6-tetrahydropyridin-2-yl)carboxylate (**1**), and also solved its X-ray structure.[1] We now present results of studies on the functionalisation of its constituent olefin.

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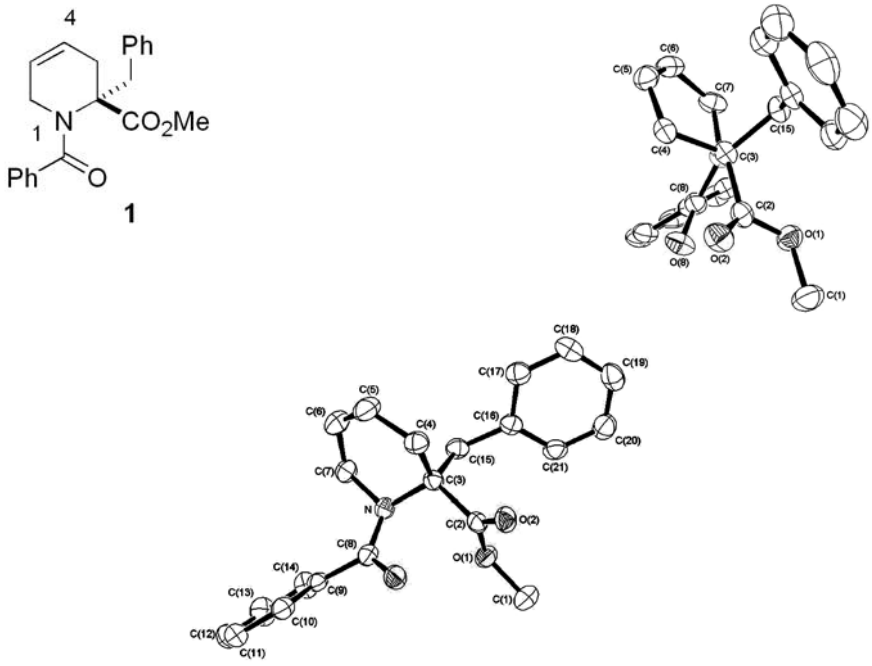
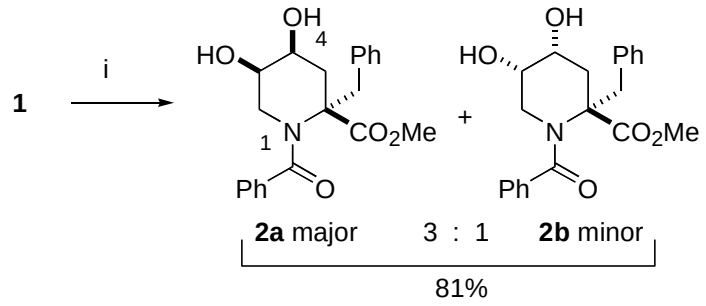


Figure 1. Compound **1** and perspective view of its X-ray crystal structure.

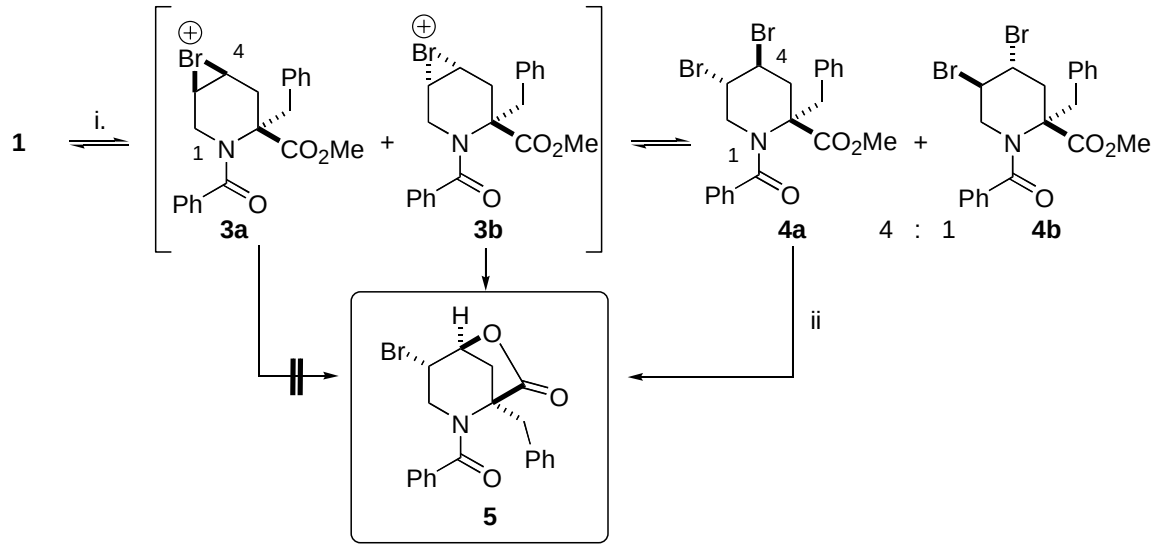
Results and Discussion

In the first instance, the peptidomimetic **1** was treated with potassium osmate and N-methylmorpholine-*N*-oxide (NMO) to give a mixture of *syn* diols in a ratio of 3:1 by ^{1}H NMR (Scheme 1). The crude mixture was purified by column chromatography to give the major isomer in 55% yield and an enriched sample of the minor isomer (2:1, minor to major). The major isomer was assigned the absolute configuration **2a** based on the known configuration of **1** and the fact that osmylation would be expected to predominate from the less hindered face of **1**, i.e. the side opposite the pseudo axial benzyl group as revealed in the X-ray structure (see Fig 1). The assignment of absolute configuration to **2a**, and hence **2b**, was supported by ^{1}H NMR spectral data. In particular, the resonance at 3.96 ppm (H5) gave a positive nOe to the benzyl methylene group at 3.06 ppm. This is consistent with a *syn* relationship between these two groups as in **2a** but not **2b**. The assignment of the resonances at 4.20 and 3.96 to H4 and H5, respectively, was consistent with observed COSY cross peaks between (i) the resonance at 4.20 ppm and the C3 methylene at 2.04 and 2.27 ppm and (2) the resonance at 3.96 ppm and the C6 methylene at 2.37 and 3.51 ppm. A positive nOe was also observed between the H4 and one of the C3 methylene resonances.



Scheme 1. i. K_2OsO_4, NMO, *t*-BuOH, H_2O/acetone (8:3), rt, 24 h.

With this result in hand we next turned our attention to the bromination of **1** (Scheme 2). Treatment of **1** with a slight excess of bromine gave two *anti* dibromides in a ratio of 4:1 by ^{1}H NMR. Purification of this mixture by silica chromatography resulted in isolation of the major isomer in 67% yield. Bromination of **1** would be expected to initially give rise to the two isomeric bromonium ions **3a** and **3b**, and again addition *anti* to the pseudo axial C2 benzyl group would be expected to predominate to give **3a** as the major isomer. The bromonium ion **3a** would then give rise to **4a** via *anti* bromide attack at the less hindered C5 position. A similar bromide attack on **3b** would give rise to **4b**. Thus the major dibromide isomer is assigned the absolute configuration **4a**. ^{1}H NMR and nOe data is again consistent with this assignment: as for **2a** the resonance for H4 (4.61 ppm) of **4a** is downfield relative to that of H5 (4.03 ppm) and a diagnostic positive nOe was observed between H4 and the C3 methylene. Unlike **2a**, an nOe was not observed between C2 benzyl methylene and H5 since these groups are *syn* in **2a** but *anti* in **4a**.



Scheme 2. i. Br_2, CH_2Cl_2, rt; ii. Crystallisation from ethyl acetate/petroleum ether.

The major isomer **4a** was left to stand for 6 months at which time a single crystal was obtained and its structure solved by X-ray crystallography (Figure 2). This revealed, somewhat to our surprise, that lactonisation had occurred to give **5** with the absolute configuration being assigned based on the known absolute configuration of C2 in **1**. Lactonisation of **4a** to give **5** requires hydrolysis of the methyl ester followed by displacement of the C4 bromide, which would be expected to occur with inversion of configuration at that centre. However, this is not possible since the methoxycarbonyl group and the C4 bromide of **4a** are *syn*. In addition, while attack of the axial 'carboxylate' of **4b** at C4 is possible, it would give rise to a lactone with the incorrect configuration at C5. Therefore, lactonisation to give **5** must occur by reversion to the bromonium ions **3a** and **3b**, of which only **3b** has suitable stereochemistry for cyclisation. This is supported by the observation that **5** and **3b** have the same absolute configuration at the stereogenic centre bearing the bromine.

Lactone **5** crystallized in the chiral, orthorhombic space group $P2_12_12_1$ with four molecules in the unit cell (see Figure 2 for a perspective view and atom numbering). It is apparent that the piperidine ring atoms represented by N, C3-C7 adopt a distinct boat conformation, with a five-membered lactone ring bridge existing between C5 and the carboxyl group extending from C3. A distorsion of normal tetrahedral geometry (bond angle is 109.5º) is observed about C3 due to the formation of this lactone. Bond angles around C3, internal to the ring system, for C2-C3-C4, C2-C3-N and C4-C3-N, are 100.3 (2)°, 106.8 (2)° and 106.7 (2)° respectively. Bond angles for C3, external to the ring system, for N-C3-C15, C2-C3-C15 and C4-C3-C15 are 115.0 (3)°, 110.7 (2)° and 116.0 (2)° respectively. The torsion angle about the peptide backbone of C8-N-C3-C2 is –78.9 (3)º. A slight twisting of the amide bond is also observed, with the magnitude of the torsion angle about O8-C8-N-C3 being 15.1 (4)°. The torsion angle about the amide bond represented by C9-C8-N-C3 is –159.9 (2)° indicating a *trans* amide bond relationship exists along the peptide backbone. No significant pyrimidalisation of the amide nitrogen is observed with the angles at N summing to 359.2°.

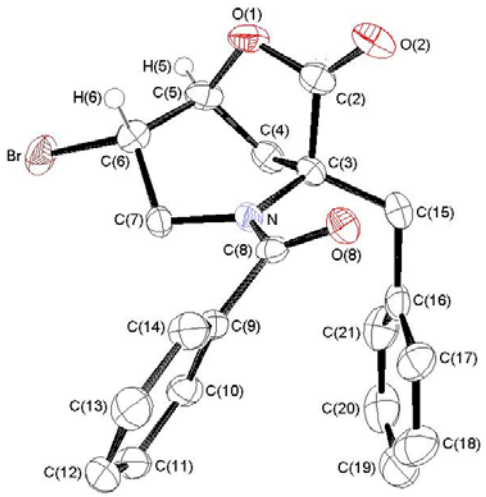


Figure 2. Perspective drawing of **5** showing crystallographic numbering.

Experimental Section

General Procedures. Proton NMR and nOe spectra were recorded on a Varian 500 MHz NMR spectrometer. Carbon NMR spectra were recorded on a Varian 300 MHz NMR spectrometer operating at 75 MHz. Chromatography was carried out on a Harrison and Harrison chromatotron using Merk type P.F. 254 silica gel.

***cis*-(2*S*,4*S*,5*R*) and *cis*-(2*S*,4*R*,5*S*)-1-Benzoyl-2-benzyl-4,5-dihydroxy-piperidine-2-carbo xylic acid methyl esters (2a) and (2b).** A solution of **1** (45 mg, 0.13 mmol, 1 equiv) in acetone (0.03 mL) was added, under nitrogen, to a mixture of K_2OsO_4 (2.5 mg, 0.05 equiv), ^{t}BuOH (0.02 mL), NMO (17 mg, 0.15 mmol, 1.06 equiv), water (0.08 mL) and acetone (0.03 mL). The mixture was stirred at rt for 24 h, following which a slurry of magnesium silicate (17 mg), and sodium dithionate (2.5 mg) in water (0.1 mL) was added. The magnesium silicate was removed by filtration and the filtrate adjusted to pH 7 with 1N H_2SO_4. Acetone was removed under reduced pressure and the resulting aqueous phase acidified to pH 2 with 1M aqueous HCl. The aqueous phase was then saturated with NaCl and extracted with ethyl acetate (2 x 2 mL). The organic extracts were combined, dried ($MgSO_4$) and the solvent removed under reduced pressure to give a crude mixture of **2a** and **2b** (40 mg, 3:1 by ^{1}H NMR). The residue was purified by radial chromatography (ethyl acetate/dichloromethane 1:1) to give a fraction containing 27 mg (55%) of the major *cis* isomer **3a**, and a second fraction containing 13 mg of a 3:1 mixture of **2a** and **2b** which could not be separated further.
Data for **2a**: ^{1}H NMR ($CDCl_3$) δ 2.04 (dd *J*=14.5 and 4.8Hz, 1H, CC**H$_a$**CHOH), 2.27 (dd *J*=14.2 and 11.1Hz, 1H, CC**H$_b$**CHOH), 2.37 (dd *J*=9.5 and 13.8Hz, 1H, NC**H$_a$**CHOH), 3.06 (d *J*=13.9Hz, 1H, C**H$_a$**Ph) 3.49 (dd *J*=13.7 and 5.8Hz, 1H, H, C**H$_a$**Ph, 3.51 (dd, 5.7 and 13.8Hz, 1H, NC**H$_b$**CHOH), 3.79 (s, 3H, OMe), 3.96 (m, 1H, NCH$_2$C**H**OH), 4.01 (d, 13.9Hz, 1H, C**H$_b$**Ph), 4.20 (m, 1H, CCH$_2$C**H**OH), 7.22-7.45 (m, 10H, Ph**H**). ^{13}C NMR ($CDCl_3$) δ 35.0, 38.9, 49.0, 52.6, 63.6, 64.0, 65.0, 126.9, 127.1, 128.3, 128.6, 130.1, 131.0, 135.8, 136.0, 171.9, 173.0. HRMS Calcd for $C_{21}H_{24}NO_5$ (M+H) 370.1654; Found 370.1655.
Selected data for **2b** (from mixture): ^{1}H NMR ($CDCl_3$) δ 18, 2.58 (dd *J*=14.2 and 1.5Hz, 1H), 3.01 (d *J*=15.4Hz, 1H, CH$_a$Ph), 3.75 (d *J*=15.4Hz, 1H), 3.86 (s, 3H, OMe), 3.97 (d *J*=14.2Hz, 1H, CH$_b$Ph).

Preparation of (2*S*,4*S*,5*S*) and (2*S*,4*R*,5*R*)-1-benzoyl-2-benzyl-4,5-dibromo-piperidine-2-carboxylic acid methyl esters (4a) and (4b), and (2*S*,4*S*,5*S*)-2-benzoyl-1-benzyl-4-bromo-6-oxa-2-aza-bicyclo[3,2,1]octan-7-one (5). Bromine was added dropwise to a solution of **1** (15 mg, 0.045 mmol) in CH_2Cl_2 (5 mL) under nitrogen, until a permanent brown colour remained and the resulting mixture was stirred under nitrogen for 3 h. The solvent was removed under reduced pressure to give a brown residue containing a mixture of **4a** and **4b** (4:1 by ^{1}H NMR). Purification by radial chromatography (ethyl acetate/petroleum ether, 1:3) gave the major isomer **4a** (15 mg, 67%).

Data for **4a**: ^{1}H NMR ($CDCl_3$) δ 2.43 (dd *J*=4.0 and 14.7Hz, 1H, CC**H$_a$**CHBr), 2.61 (t *J*=14.2Hz, 1H, CC**H$_b$**CHBr), 2.85 (dd *J*=5.1 and 16.1Hz, 1H, NC**H$_a$**CHBr), 3.07 (d *J*=14.3Hz, 1H, C**H$_a$**Ph), 3.63 (dd *J*=2.2 and 16.1Hz, 1H, NC**H$_b$**CHBr), 3.91 (s, 1H, OC**H$_3$**), 4.03 (m, 1H, NCH_2C**H**Br), 4.11 (d *J*=14.3Hz, 1H, C**H$_b$**Ph), 4.61 (m, 1H, CCH_2C**H**Br), 7.23-7.53 (m, 10H, Ph**H**). ^{13}C NMR ($CDCl_3$) δ 38.4, 39.5, 48.5, 51.1, 52.2, 52.8, 64.5, 127.0, 127.3, 128.3, 128.5, 130.0, 130.9, 134.7, 135.1, 171.8, 171.9. HRMS Calcd for $C_{21}H_{22}Br_2NO_3$ (M+H) 495.9946; Found 495.9942.
Selected data for **4b** (from crude mixture): ^{1}H NMR ($CDCl_3$) δ 2.22 (dd *J*=13.4 and 5.5Hz, 1H), 3.14 (d *J*=13.1Hz, 1H), 3.22 (d *J*=14.3Hz, 1H, C**H$_a$**Ph), 3.80 (s, 3H, O**Me**), 4.27 (d *J*=14.3Hz, 1H, C**H$_b$**Ph), 4.86 (m, 1H).
Crystallisation by the diffusion of petroleum ether into a solution of **4a** in ethyl acetate, over a period of 6 months, gave **5**, the structure of which was determined by X-ray crystal structure analysis.

X-ray crystallography. Data were collected with a Siemens SMART CCD area detector, using graphite monochromatized MoKα radiation (λ = 0.71073 Å). The intensities were corrected for Lorentz and polarisation effects and for absorption.[14] The structure was solved by direct methods using SHELXS[15] and refined on F^2, using all data, by full-matrix least-squares procedures using SHELXTL.[16] Complete crystallographic data, as a CIF file, have been deposited with the Cambridge Crystallographic Data Centre (CCDC No 234281). Copies can be obtained free of charge from: The Director, CCDC, 12 Union Road, Cambridge CB2 1EZ, U.K. (email: *deposit@ccdc.cam.ac.uk*).

Crystal data for 5. $C_{20}H_{18}BrNO_3$, MW 400.26, orthorhombic, $P2_12_12_1$, *a* = 6.271(11), *b* = 15.83(3), *c* = 17.57(3) Å, V = 1745(5) Å^3, Z = 4, T = -105°C(2), F(000) = 816, $2\theta_{max}$ 52.568°, $wR(F^2)$ = 0.0745 (all 6974 data), R = 0.0315 (3395 data with I > 2σI).

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