Synthesis of Conformationally Constrained Arginine and Ornithine Analogues Based on the 3-Substituted Pyrrolidine Framework

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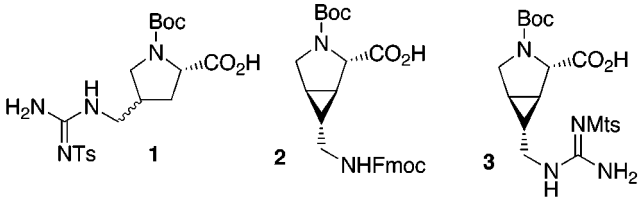
Introduction

As part of our program focused on the synthesis of mimics of the poly-L-proline type II (PPII) secondary structure, we are pursuing the synthesis of unnatural amino acids we call proline-templated amino acids (PTAAs).[1] OligoPTAAs are specifically designed to preferentially populate the PPII conformation in solution due to a combination of cyclic and acyclic conformational control elements (Figure 1).[1] OligoPTAAs are thus unusual in that they represent conformationally constrained linear peptides. Since a number of receptor-bound PPII helices include basic residues, we are particularly interested in the synthesis of arginine and ornithine PTAAs.[2] Some of these analogues are known. For example, the synthesis of the cis and trans 4-substituted arginine analogues **1** was reported by Webb and Eigenbrot, while our group has reported the synthesis of ornithine (**2**) and arginine (**3**) analogues based on the 3-aza-bicyclo[3.1.0]-hexane system.[3,4,5] This work describes our contributions to the synthesis of the 3-substituted ornithine and arginine analogues suitably protected for Fmoc/Boc solid-phase peptide synthesis.



Results and Discussion

Both syntheses use the α,β-unsaturated lactam **4** as a common intermediate. Conjugate addition of $LiCH_2CN$ to the α,β-lactam **4** was smoothly accomplished in THF at −78 °C in 79% yield (Scheme 1).[6] This conjugate addition reaction serves to introduce the desired stereochemistry at C2 (which will become Cβ of the PTAA) and also the requisite ϵ-nitrogen.[7] Interestingly, in one instance when we scaled-up the conjugate addition reaction, another product was obtained in 29% yield. The mass spectrum of this compound was consistent with dimerization of the bicyclic lactam and incorporation of CH_2CN. It is well precedented that with basic nucleophiles this class of bicyclic lactams is prone to enolization (**11**) and dimerization affording structures such as **12** (Scheme 2).[8] At first, it was surmised that addition of $LiCH_2CN$ to **12** would afford the observed dimer (after workup). However, COSY spectroscopy revealed that the correct structure was the tetracycle **16** that presumably

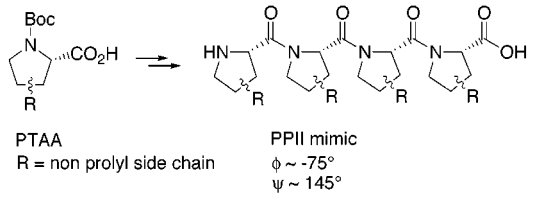
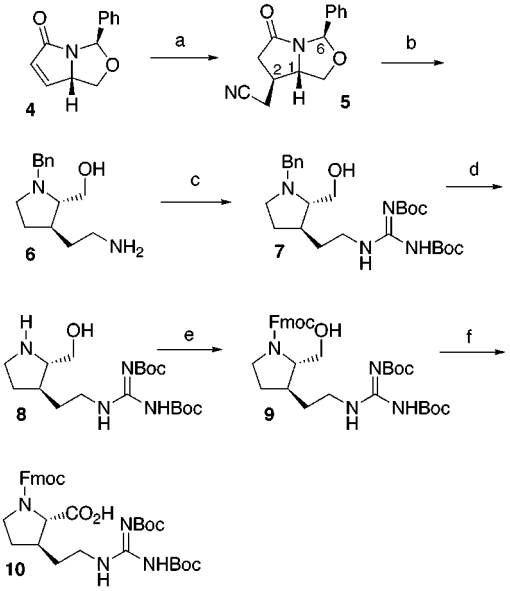


Figure 1. PPII mimic design strategy.

Scheme 1[a]



[a] Reagents and conditions: (a) $LiCH_2CN$, THF, −78 °C; (b) 3.6 equiv of BH_3·THF, THF, reflux; (c) $(BocNH)_2C{=}NTf$, Et_3N, CH_2Cl_2; (d) Pd−C, HCO_2NH_4, MeOH, reflux; (e) FmocCl, $NaHCO_3$, dioxane/H_2O; (f) TEMPO, bleach, $NaClO_2$.

(1) (a) Zhang, R.; Madalengoitia, J. S. *Tetrahedron Lett.* **1996**, *37*, 6235. (b) Zhang, R.; Brownewell, F.; Madalengoitia, J. S. *J. Am. Chem. Soc.* **1998**, *120*, 3894. (c) Zhang, R.; Madalengoitia, J. S. *J. Org. Chem.* **1999**, *64*, 330. (d) Mamai, A.; Zhang, R.; Natarajan, A.; Madalengoitia, J. S. *J. Org. Chem.* **2001**, *66*, 455.

(2) Siligardi, G.; Drake, A. F. *Pept. Sci.* **1995**, *37*, 281.

(3) Webb, T. R.; Eigenbrot, C. *J. Org. Chem.* **1991**, *56*, 3009.

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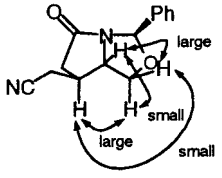
(5) Mamai, A.; Madalengoitia, J. S. *Org. Lett.* **2001**, *3*, 561.

(6) The stereochemical assignment is based on relative NOE intensities. The NOEs were determined in DMSO-d_6 as it resolves the resonances. The CH_2O resonances were initially assigned based on the relative NOE intensities with the methine hydrogen. From these assignments, the C2-stereochemistry could then be assigned based on the NOE intensities of endo and exo C8-hydrogens with the C2-hydrogen.



(7) The intermediate **5** was reported by the Moloney group in the synthesis of a pyroglutamate-derived ornithine analogue. Goswami, R.; Moloney, M. G. *Chem. Commun.* **1999**, 2333.

(8) Nagasaka, T.; Imai, T. *Chem. Pharm. Bull.* **1997**, *45*, 36.

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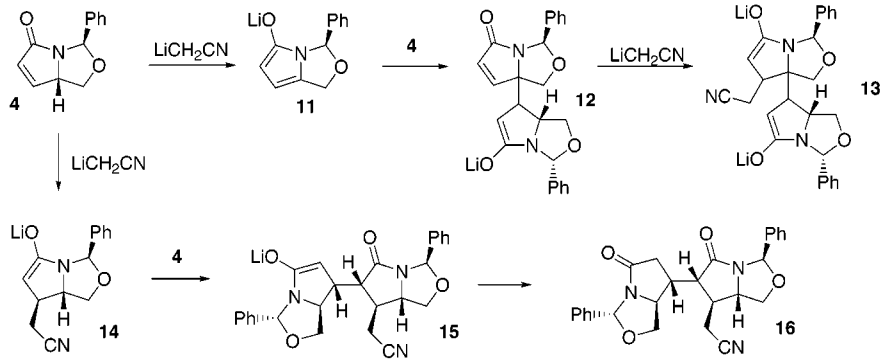
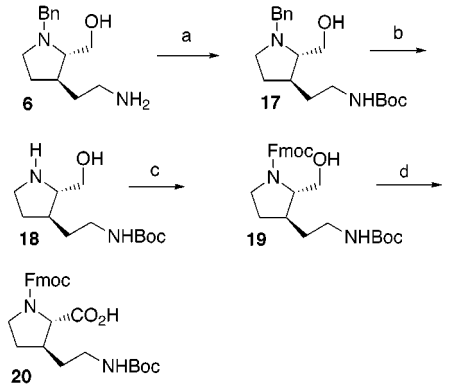
Scheme 2



arises from two tandem conjugate addition reactions (Scheme 2). Interestingly, the dimer is formed as a single isomer with five contiguous stereocenters. The stereochemistry of the dimer has not been rigorously established; however, the stereochemistry shown is the most likely from the precedented behavior of this bicyclic lactam.[9] The formation of the dimer is most likely a result of the low solubility of $LiCH_2CN$ in THF. Since there is little $LiCH_2CN$ in solution, the enolate **14** formed from the initial conjugate addition reaction can then begin to compete with $LiCH_2CN$ for the α,β-unsaturated lactam (even when lactam is added to $LiCH_2CN$). The procedure described in the Experimental Section utilizes a dilution factor that minimizes formation of the dimer **16** to 1−5%. With the introduction of the desired functionality at what will become Cβ of the PTAA, a method for the reduction of the amide, oxazolidine, and nitrile was next investigated. When the lactam **5** was subjected to LAH in refluxing THF, the amine **6** was obtained with good mass recovery (~100%).[10] However, inspection of the crude ^{1}H NMR spectrum revealed significant baseline impurities. Consequently, when the crude amine **6** was guanidinylated with the Goodman reagent $(BocNH)_2NTf$, the prolinol **7** was obtained, but in 35% yield for both steps.[11] In contrast, reduction of the nitrile **5** with BH_3 in refluxing THF afforded the amine **6** with significantly improved purity such that guanidinylation of the primary amine functionality with $(BocNH)_2NTf$ afforded the prolinol **7** in 77% yield for both steps. N-Debenzylation of prolinol **7** was accomplished with HCO_2NH_4, Pd−C.[12] Protection of the resultant secondary amine **8** with FmocCl in aqueous $NaHCO_3$/dioxane gave the Fmoc-prolinol **9** in 50% yield for both steps. Finally, TEMPO oxidation of the primary alcohol afforded the PTAA **10** appropriately protected for Fmoc/Boc solid-phase peptide synthesis.[13] To demonstrate the practical nature of this route, 22 g of PTAA **10** were synthesized in 11 steps from pyroglutamic acid.

The ornithine PTAA was obtained in a manner similar to the arginine PTAA (Scheme 3). Starting from the pyrrolidine **6**, the primary amine was protected with Boc_2O in 61% yield. N-Debenzylation of the tertiary amine (HCO_2NH_4, Pd−C) followed by reprotection of the resulting secondary amine with FmocCl gave the N-Fmoc protected prolinol **19** in 46% yield for both steps. Oxidation of the primary alcohol with TEMPO gave the PTAA **20** in 96% yield.

Scheme 3[a]



[a] Reagents and conditions: (a) Boc_2O, CH_2Cl_2; (b) Pd−C, HCO_2NH_4, MeOH, reflux; (c) FmocCl, $NaHCO_3$, dioxane/H_2O; (d) TEMPO, bleach, $NaClO_2$.

In conclusion, we have developed practical routes to two conformationally constrained PTAAs appropriately protected for Fmoc/Boc solid-phase peptide synthesis. The routes are concise and amenable to the synthesis of multigram quantities of PTAAs. These amino acids should find utility in the synthesis of conformationally constrained peptides. The results from these studies will be reported in due course.

Experimental Section

General. Unless otherwise specified, all reagents were purchased from commercial sources and were used without further purification. Reagent $(BocHN)_2C{=}NTf$ was prepared according to Goodman's procedure.[11] THF was distilled from sodium benzophenone ketyl, and CH_2Cl_2 was distilled from CaH_2. When required, reactions were carried out under a N_2 atmosphere. Flash chromatography was carried out on Selecto silica gel (230−400 mesh). NMR spectra were collected in $CDCl_3$ unless otherwise noted using standard pulse sequences provided by Brucker.

(1*S*,2*S*,6*R*)-2-(5-Aza-7-oxa-4-oxo-6-phenylbicyclo[3.3.0]-oct-2-yl)ethanenitrile (5). Freshly distilled CH_3CN (6.00 mL, 116 mmol) in THF (80 mL) was cooled to −78 °C. LDA (60.0 mL, 120 mmol) was added dropwise over 30 min as a 2 M solution in hexanes. After 20 min, a solution of lactam **4** (11.67 g, 58.00 mmol) in THF (60 mL) was added dropwise to the mixture. The solution was maintained at −78 °C for 20 min then treated with saturated aqueous $NaHCO_3$ (20 mL). After warming to room temperature, the layers were separated and the aqueous layer was extracted with EtOAc (3 × 60 mL). The combined organic fractions were dried over Na_2SO_4 and concentrated.

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Purification by flash chromatography with EtOAc:/hexanes (50:50 then 75:25) afforded the nitrile **5** as a colorless solid (11.14 g, 79%): mp 97−98 °C; $[\alpha]^{20}_D = +177.4°$ (c = 0.83, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$) δ 7.47−7.24 (m, 5H), 6.40 (s, 1H), 4.32 (dd, J = 6.5, 8.6, 1H), 3.96 (apparent q, ddd, J = 6.6, 6.6, 6.6 Hz, 1H), 3.72 (dd, J = 6.8, 8.6 Hz, 1H), 2.78 (m, 1H), 2.70−2.53 (m, 4H); ^{13}C NMR (125 MHz, $CDCl_3$) 175.2, 137.9, 128.8, 128.6, 126.0, 117.1, 87.5, 63.5, 60.4, 39.6, 36.3, 21.6 ppm; IR (film) 2248, 1707 cm^{-1}; MS (CI) m/z 243 (MH). Anal. Calcd for $C_{14}H_{14}N_2O_2$: C, 69.41; H, 5.82; N, 11.56. Found: C, 69.22; H, 5.76; N, 11.45. **Dimer 16.** colorless solid: mp 178−180 °C; [a]$^{25}_D = +154.2°$ (c 0.9, acetone); ^1H NMR (500 MHz, DMSO d_6) δ 7.27−7.22 (m, 10H), 5.98 (s, 1H), 5.97 (s, 1H), 4.14−4.08 (m, 2H), 3.97 (dd, J = 5.6, 13.6 Hz, 1H), 3.85 (dd, J = 7.3, 11.5 Hz, 1H), 3.57 (dd apparent t, J = 8.0 Hz, 1H), 3.49 (dd apparent t, J = 8.0 Hz, 1H), 3.41 (dd apparent t, J = 6.0 Hz, 1H), 2.63−2.51 (m, 5H) 2.39−2.33 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) 175.4, 175.0, 139.2, 129.1, 128.8, 128.5, 126.5, 126.2, 119.8, 87.1, 86.7, 70.8, 68.9, 62.9, 62.6, 41.6, 40.4, 40.1, 38.8, 37.6, 35.4, 30.0 ppm; FTIR (KBr) 3060, 3035, 2361, 1705, 1701, 1399, 1356 cm^{-1}; MS (CI) m/z 444 (MH). Anal. Calcd for $C_{26}H_{25}N_3O_4$: C, 70.41; H, 5.68; N, 9.47. Found: C, 70.21; H, 5.89; N, 9.64.

***tert*-Butyl 3-({2-[(2*S*,3*S*)-2-(Hydroxymethyl)-1-benzylpyrrolidin-3-yl]ethyl}amino)-2-aza-3-[(*tert*-butoxycarbonylamino]prop-2-enoate (7). Method A**. A solution of nitrile **5** (10.74 g, 44.33 mmol) in THF (60 mL) was slowly added to a suspension of LAH (5.93 g, 156 mmol) in 100 mL of THF. The mixture was heated at reflux for 10 h and then cooled to 0 °C. H_2O (6 mL), NaOH (1 M in H_2O, 6 mL), and then H_2O (18 mL) were successively added to the solution. The mixture was stirred for 30 min at room temperature and filtered. The filtrate was concentrated to afford the amine **6** (10.68 g) as a yellow oil which was used without further purification: ^1H NMR (500 MHz, $CDCl_3$) δ 7.27−7.13 (m, 5H), 3.91 (d, J = 13.0 Hz, 1H), 3.54 (dd, J = 4.2, 10.7 Hz, 1H), 3.24 (d, J = 13.0 Hz, 1H), 2.85−2.80 (m, 1H), 2.66−2.60 (m, 2H), 2.37−2.20 (m, 6H), 2.10−2.01 (m, 1H), 1.84−1.74 (m, 1H), 1.51−1.47 (m, 1H), 1.40−1.34 (m, 1H) 1.31−1.25 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) 139.3, 128.7, 128.2, 127.1, 71.2, 61.5, 58.9, 52.7, 40.3, 38.7, 38.1, 29.7 ppm; FTIR (film) 3350, 3290, 3200 cm^{-1}; MS (CI) m/z 235 (MH). The crude amine **6** (10.68 g) in CH_2Cl_2 (100 mL) was slowly added to a solution of $(BocNH)_2C{=}NTf$ (16.0 g, 41.0 mmol) and triethylamine (6.4 mL, 46 mmol). After 24 h at room temperature, the solution was washed with a saturated aqueous $NaHCO_3$ (50 mL) and the aqueous layer was back extracted with CH_2Cl_2 (3 × 200 mL). The combined organic layers were dried over Na_2SO_4 and then concentrated. The crude residue was purified by flash chromatography (100% CH_2Cl_2 then 100% EtOAc) to afford the guanidine **7** (7.58 g, 35% from **5**) as a colorless foam: $[\alpha]^{25}_D = -6.1°$ (c 0.3, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$) δ 10.55−10.45 (br s, 1H), 8.25 (s, 1H), 7.21−7.16 (m, 5H), 3.9 (d, J = 12.8 Hz, 1H), 3.58 (d, J = 8.6 Hz, 1H), 3.45−3.38 (m, 5H), 3.26 (d, J = 12.8 Hz, 1H), 2.87−2.84 (m, 1H), 2.33−2.27 (m, 2H), 2.12−2.06 (m, 2H), 1.87−1.79 (m, 1H), 1.69−1.63 (m, 1H), 1.43 (s, 9H), 1.41 (s, 9H); ^{13}C NMR 163.5, 156.1, 153.2, 138.9, 128.7, 128.3, 127.1, 83.0, 79.1, 71.1, 60.8, 58.6, 52.7, 39.5, 38.2, 34.7, 29.5, 28.3, 28.0 ppm; FTIR (film) 3330, 3250, 1721, 1642 cm^{-1}; MS (CI) m/z 477 (MH). Anal. Calcd for $C_{25}H_{40}N_4O_5$: C, 63.00; H, 8.46; N, 11.76. Found: C, 62.88; H, 8.36; N, 11.72. **Method B**: BH_3·THF (380 mL, 380 mmol) was added dropwise as 1.0 M solution in THF to a solution of the nitrile **5** (25.5 g, 105 mmol) in THF (200 mL), and the resultant mixture was heated at reflux for 1 h. After the solution cooled to room temperature, the solvent was removed under reduced pressure, and the residue was dissolved in methanolic HCl (500 mL). The solution was heated at reflux for 2 h, and the methanol was removed under reduced pressure. The resulting oil was dissolved in CH_2Cl_2 (200 mL) and washed with a 20% aqueous K_2CO_3 (200 mL). The aqueous layer was extracted with CH_2Cl_2 (4 × 200 mL), and the combined organic fractions were dried over Na_2SO_4 and concentrated to afford the amino-alcohol **6** (25.5 g) as a yellow oil. The amino-alcohol **6** (25.5) was allowed to react with $(BocNH)_2C{=}NTf$ (38.3 g, 97.9 mmol) according to the procedure described above. This modified procedure afforded 38.8 g (77% from the nitrile **5**) of the guanidine **7** as a colorless foam.

***tert*-Butyl 3-[(2-{(2*S*,3*S*)-1-[(Fluoren-9-ylmethyl)oxycarbonyl]-2-(hydroxymethyl)pyrrolidin-3-yl}ethyl)amino]-2-aza-3-[(*tert*-butoxy)carbonylamino]prop-2-enoate (9).** A solution of the guanidine **7** (38.0 g, 79.8 mmol), 10% Pd−C (12 g), and ammonium formate (30.7 g, 479 mmol) in MeOH (400 mL) was heated at reflux for 30 min. After cooling, the mixture was filtered through Celite, and the Celite plug was washed with MeOH (3 × 150 mL) and CH_2Cl_2 (3 × 200 mL). The filtrate was concentrated to yield the corresponding amino-alcohol **8** (30.3 g) as a colorless foam which was used without further purification: ^1H NMR (500 MHz, $CDCl_3$) δ 11.51 (br s, 1H) 8.48 (br s, 1H), 6.35−6.25 (m, 2H), 3.90 (dd, J = 3.7, 10.2 Hz, 1H) 3.74−3.70 (m, 1H), 3.46−3.34 (m, 2H), 3.26−3.21 (m, 1H), 3.11−3.05 (m, 1H), 2.42−2.32 (m, 1H), 2.05−2.01 (m, 1H), 1.79−1.75 (m, 1H), 1.62−1.55 (m, 3H), 1.48 (s, 18H); ^{13}C NMR (125 MHz, $CDCl_3$), 163.4, 156.0, 153.2, 83.0, 79.1, 65.8, 61.3, 45.2, 39.8, 38.4, 34.7, 33.4, 28.2, 28.0; FTIR (film) 3328, 3292, 1723, 1717, 1684; MS (CI) m/z 387 (MH). The amino-alcohol **8** was dissolved in a 1:1 mixture of dioxane and a saturated aqueous $NaHCO_3$ solution (600 mL) and cooled to 0 °C. FmocCl (20.3 g, 78.2 mmol) was slowly added, and the solution was maintained at 0 °C for 4 h. The mixture was extracted with EtOAc (4 × 200 mL). The combined organic layers were dried over Na_2SO_4 and concentrated. The residue was purified on silica gel eluting with EtOAc/hexanes (30:70) and then CH_2Cl_2/acetone (10:90) to afford The Fmoc-prolinol **9** (24.3 g, 50%) as a colorless foam: $[\alpha]^{25}_D = -9.9°$ (c 2.0, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$) δ 11.48 (s, 1H), 8.30 (br s, 1H), 7.72 (d, J = 7.4 Hz, 2H), 7.55 (d, J = 7.4 Hz, 2H), 7.35 (t, J = 7.4 Hz, 2H), 7.27 (t, J = 7.4 Hz, 2H), 4.43−4.39 (m, 2H), 4.23−4.18 (m, 2H), 3.69−3.65 (m, 1H), 3.60−3.52 (m, 2H), 3.49−3.41 (m, 2H), 3.27−3.21 (m, 2H), 2.18−2.12 (m, 1H), 2.07−2.01 (m, 2H), 1.94−1.88 (m, 1H), 1.82−1.76 (m, 1H), 1.48 (s, 9H), 1.47 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$) 163.5, 156.5, 156.2, 153.3, 143.9, 141.4, 127.7, 127.1, 125.0, 119.9, 83.2, 79.2, 67.4, 66.1, 65.5, 60.3, 47.4, 46.1, 39.3, 39.0, 32.8, 28.3, 28.1 ppm; FTIR (film) 3330, 3295, 1718, 1700, 1685, 1636, cm^{-1}; MS (CI) m/z 609 (MH).

(2*S*,3*S*)-3-[2-({-2-Aza-2-[(*tert*-butyl)oxycarbonyl]-1-[(*tert*-butoxy)carbonylamino]vinyl}amino)ethyl]-1-[fluoren-9-ylmethyl)oxycarbonyl]pyrrolidine-2-carboxylic Acid (10). The primary alcohol **9** (23.9 g, 39.2 mmol), TEMPO (0.42 g, 2.5 mmol), and $NaClO_2$ (7.4 g, 79 mmol) were added to a mixture of acetonitrile (85 mL) and NaH_2PO_4 (0.67 M) (75 mL). This mixture was warmed to 35 °C, and bleach (1.1 mL) was slowly added over 15 min. After 5 h at 35 °C, the reaction mixture was cooled to room temperature and poured over a solution of Na_2SO_3 (10.5 g), water (22 mL), and ice (60 g). The layers were separated, and the aqueous layer was extracted with EtOAc (3 × 150 mL). The combined organic fractions were dried over Na_2SO_4 and concentrated. Purification of the residue on silica gel eluting with 96.5:3.3:0.2 CH_2Cl_2−MeOH−AcOH afforded the acid **10** (22.2 g, 90%) as a colorless solid: mp 116−118 °C; $[\alpha]^{25}_D = -23.8°$ (c 0.3, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$) δ 11.42−11.19 (br s, 1H), 8.41 (br s, 1H), 7.74 (d, J = 7.4 Hz, 1H), 7.69 (d, J = 7.4 Hz, 1H), 7.59−7.52 (m, 2H), 7.39−7.20 (m, 4H), 4.45−4.39 (m, 1H), 4.34 (dd apparent t, J = 8.7 Hz, 1H), 4.23 (dd apparent t, J = 6.9 Hz, 1H), 4.15−4.09 (m, 1H), 3.64−3.48 (m, 4H), 2.41−2.36 (m, 1H), 2.17−2.08 (m, 1H), 1.92−1.84 (m, 1H), 1.72−1.60 (m, 2H), 1.53−1.45 (m, 1H), 1.49 (s, 18H); ^{13}C NMR (125 MHz, $CDCl_3$) 175.9, 174.8, 163.3, 156.3, 155.3, 154.5, 153.3, 144.1, 143.8, 141.3, 127.7, 127.1, 125.1, 119.9, 83.4, 79.5, 67.7, 64.5, 63.9, 47.3, 46.1, 45.7, 42.4, 41.0, 39.2, 33.2, 33.0, 31.1, 30.2, 29.5, 28.3, 28.1 ppm; FTIR (KBr) 3326, 3284, 1725, 1718, 1701, 1685, 1653, cm^{-1}; MS (CI) m/z 623 (MH). Anal. Calcd for $C_{33}H_{42}N_4O_8$: C, 63.65; H, 6.80; N, 9.00. Found: C, 63.40; H, 6.85; N, 8.99.

***N*-{2-[(2*S*,3*S*)-2-(Hydoxymethyl)-1-benzylpyrrolidin-3-yl]-ethyl}(*tert*-butoxy)carboxamide (17).** A solution of the amine **6** (1.00 g, 4.27 mmol), Boc_2O (1.0 g, 4.7 mmol), and DIEA (0.82 mL, 4.7 mmol) in CH_2Cl_2 (40 mL) was stirred for 6 h under N_2 and then concentrated. The residue was purified by flash chromatography over silica eluting with EtOAc to afford 1.03 g (61%) of compound **17** as a colorless foam: $[\alpha]^{25}_D = -6.5°$ (c 2.1, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$) δ 7.31−7.22 (m, 5H), 4.66 (br s, 1H), 3.96 (d, J = 13.1 Hz, 1H), 3.67 (dd, J = 3.6, 11.0 Hz, 1H), 3.47 (dd, J = 1.4, 11.0 Hz, 1H), 3.32 (d, J = 13.0 Hz, 1H), 3.23−3.17 (m, 1H), 3.12−3.05 (m, 1H), 2.96−2.91 (m, 1H), 2.37

(dd, J = 9.3, 17.1 Hz, 1H), 2.33−2.30 (m, 1H), 2.18−2.09 (m, 1H), 1.93−1.85 (m, 1H), 1.65−1.61 (m, 1H), 1.52−1.43 (m, 3H), 1.47 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$) 156.1, 139.0, 128.7, 128.3, 127.1, 79.0, 71.1, 60.8, 58.7, 52.8, 39.1, 37.9, 35.2, 29.4, 28.5 ppm; FTIR (film) 3353, 1691, cm^{-1}; MS (CI) m/z 335 (MH). Anal. Calcd for $C_{19}H_{30}N_2O_3$: C, 68.23; H, 9.04; N, 8.38. Found: C, 68.21; H, 9.02; N, 8.24.

Fluoren-9-ylmethyl(2*S*,3*S*)-3-{2-{(*tert*-butoxy)carbonylamino]ethyl}-2-(hydroxymethyl)pyrrolidinecarboxylate (19). Compound **17** (0.54 g, 1.6 mmol) was debenzylated following the same procedure as **7** to afford the corresponding secondary aminol **18** (0.37 g) as a white foam: ^{1}H NMR (500 MHz, $CDCl_3$) δ 4.96 (br s, 1H), 4.66 (br s, 2H), 3.60 (dd, J = 3.6, 11.5 Hz, 1H), 3.42 (dd, J = 6.6, 11.5 Hz, 1H), 3.10−3.01 (m, 2H), 2.99−2.94 (m, 1H), 2.92−2.86 (m, 2H), 2.02−1.96 (m, 1H), 1.79−1.69 (m, 1H), 1.64−1.55 (m, 1H), 1.42−1.34 (m, 2H), 1.38 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$) 156.3, 78.9, 65.8, 62.4, 53.5, 45.0, 38.0, 34.0, 32.1, 28.4 ppm; FTIR (film) 3327, 1700, 1690, 1455 cm^{-1}; MS (CI) m/z 245 (MH). The amino-alcohol **18** was *N*-Fmoc protected using the same conditions as for the protection of **8** above to afford **19** (0.35 g, 46%) as a colorless foam: $[\alpha]^{25}_D$ = −20.3° (*c* 1.2, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.73 (d, J = 7.4 Hz, 2H), 7.56 (d, J = 7.4 Hz, 2H), 7.37 (dd apparent t, J = 7.4 Hz, 2H), 7.29 (dd apparent t, J = 7.0 Hz, 2H), 4.86 (dd apparent t, J = 5.6 Hz, 1H), 4.44−4.36 (m, 2H), 4.20 (dd apparent t, J = 6.5 Hz, 1H). 3.65−3.50 (m, 3H), 3.26 (dd, J = 7.5, 17.3 Hz, 1H), 3.16−3.04 (m, 3H), 2.06−2.01 (m, 1H), 1.94−1.89 (m, 1H), 1.70−163 (m, 1H), 1.47−1.36 (m, 3H), 1.43 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$) 156.5, 156.1, 143.9, 141.4, 127.8, 127.1, 125.0, 120.0, 79.2, 67.4, 65.9, 65.4, 60.4, 47.4, 46.2, 38.6, 33.7, 29.9, 28.5 ppm; FTIR (film) 3360, 3065, 1700, 1684, 1653 cm^{-1}; MS (CI) m/z 467 (MH). Anal. Calcd for $C_{27}H_{34}N_2O_5$: C, 69.50; H, 7.35; N, 6.00. Found: C, 69.35; H, 7.78; N, 6.33.

(2*S*,3*S*)-3-{2-[(*tert*-Butoxy)carbonylamino]ethyl}-1-[(fluoren-9-ylmethyl)oxycarbonyl]pyrrolidine-2-carboxylic Acid (20). The primary alcohol **19** (0.35 g, 0.75 mmol) was oxidized following the same procedure described for the oxidation of **9** to afford the acid **20** (0.34 g, 96%) as a colorless solid: mp 88−90 °C; $[\alpha]^{25}_D$ = −25.5° (*c* 0.2, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ 11.16−11.01 (br s, 1H), 7.76 (d, J = 7.4 Hz, 1H), 7.73 (d, J = 7.4 Hz, 1H), 7.62 (d, J = 7.4 Hz, 1H), 7.59 (d, J = 7.4 Hz, 1H), 7.43−7.35 (m, 2H), 7.33−7.26 (m, 2H), 5.03−4.98 (m, 1H), 4.49−4.42 (m, 1H), 4.37 (dd apparent t, J = 8.9 Hz, 1H), 4.35−4.31 (m, 1H), 4.26 (dd apparent t, J = 6.7 Hz, 0.5H), 4.19 (dd apparent t, J = 6.2 Hz, 0.5H), 4.15−4.10 (m, 0.5H), 4.08−4.04 (m, 0.5H), 3.65−3.59 (m, 1H), 3.24−3.17 (m, 2H), 2.40−2.36(m, 1H), 217−2.12 (m, 1H), 1.80−1.75 (m,1H), 1.60−1.52 (m, 2H), 1.57 (s, 9H); ^{13}C NMR (125 MHz, $CDCl_3$) 175.6, 175.1, 156.5, 155.4, 154.7, 144.2, 144.1, 143.8, 141.3, 127.7, 127.1, 125.2, 120.0, 81.3, 79.8, 79.6, 67.9, 67.7, 64.3, 63.9, 47.3, 47.2, 46.1, 45.8, 42.0, 40.7, 39.7, 38.8, 33.8, 30.7, 29.7, 29.5, 28.5 ppm; FTIR (KBr) 3337, 3090, 1725, 1701, 1705, 1695, cm^{-1}; MS (CI) m/z 461 (MH).

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Supporting Information Available: ^{1}H NMR and ^{13}C NMR spectra for compounds **5**−**10** and **16**−**20**. This material is available free of charge via the Internet at http://pubs.acs.org.

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