Stereodivergent Synthesis of (*2S,3S,4R,5R*)- and (*2S,3S,4R,5S*)-[3,4,5-D_3]Proline Depending on the Substituent of the γ-Lactam Ring

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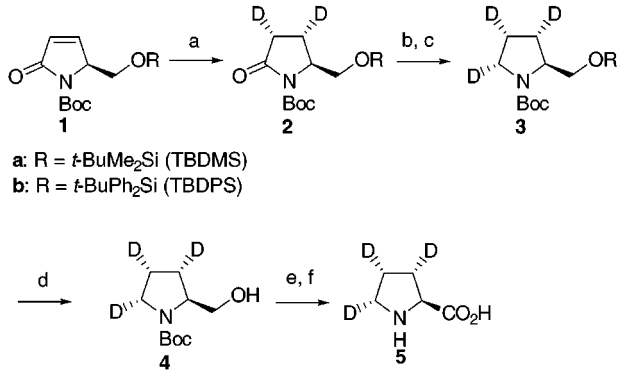
Introduction

Nowadays, a combination of a stable isotope labeling technique and multidimensional NMR spectroscopy is the principal method for obtaining a detailed solution structure of proteins.[1,2] For more precise structure determination, stereospecific assignments for the diastereotopic methylene protons or the methyl group are indispensable. In view of the above background, we have recently been engaged in preparing [3,4,5-D_3]proline[2] as part of our continuing studies on the synthesis of stereoselectively deuterated amino acids.[3] Since the residue plays a prominent role in the structure of proteins owing to its unusual conformational preference,[4] monitoring the proline ring dynamics should provide information regarding the protein interior around the residue. Our latest paper dealt with the preparation of such prolines starting from 4-hydroxyproline;[3] however, incorporation of a ^{13}C label into the proline framework is very difficult by this method. This difficulty becomes a crucial drawback when the sample is incorporated into larger biomolecules as an analytical probe, because the selective ^{13}C labeling is necessary to extract NMR signals for specific residues independently from the rest of the structure by a 2D NMR technique such as $^1H-^{13}C$ HSQC. Therefore, we herein disclose a novel protocol to access [3,4,5-D_3]proline starting from L-glutamic acid, the ^{13}C isotopomers of which are commercially available.

Results and Discussion

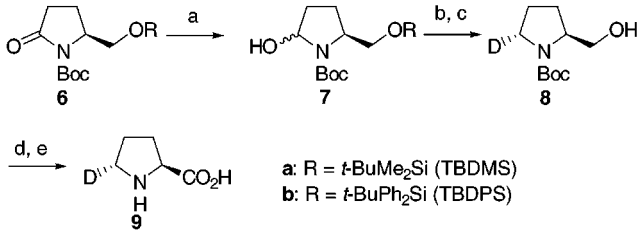
The synthetic course of [3,4,5-D_3]proline is shown in Scheme 1. The unsaturated γ-lactam derivatives **1a** and **1b** were readily prepared from L-glutamic acid according to the reported procedure.[5] When a catalytic deuteration of the olefin **1a** was carried out using 10% palladium on carbon in EtOD under medium pressure (5 kgf/cm^2) of deuterium gas, the corresponding 3,4-dideuterated lactam **2a** was obtained in 92% yield as a single diastereomer, indicating a stereospecific deuterium addition occurred. In this case, the H−D scrambling, which had been encountered in the catalytic deuteration of the dehydroproline derivatives,[3] was not observed.

Scheme 1[a]



[a] Reagents and conditions: (a) D_2 (5 kgf/cm^2), 10% Pd/C, MeOD; (b) $LiEt_3BH$, $BF_3{\cdot}OEt_2$, 51% (two steps); (d) TBAF, 84%; (e) CrO_3, H_2SO_4; (f) 1 M HCl, 110 °C, then Dowex 50W-X8, 66% (two steps).

Scheme 2[a]



[a] Reagents and conditions: (a) $LiEt_3BH$; (b) deuteride, $BF_3{\cdot}OEt_2$; (c) TBAF (R = TBDPS); (d) CrO_3, H_2SO_4; (e) 1 M HCl, 110 °C, then Dowex 50W-X8.

To obtain the [3,4,5-D_3]proline, a stereoselective reduction of the lactam carbonyl moiety with an appropriate deuteride seems to be the most straightforward route.[6] As shown in Scheme 2, a preliminary examination was carried out using the corresponding unlabeled lactam **6a** or **6b** as the starting material, and the results are compiled in Table 1. For example, Lewis acid-promoted reduction of the aminal **7a** derived from the lactam **6a** was performed using a $Bu_3SnD-BF_3{\cdot}OEt_2$ system to afford the deuterated prolinol derivative **8**. During the course of the reaction, the TBDMS group was removed.[7] After oxidation of the hydroxymethyl moiety with Jones reagent, the deprotection procedure gave [5-D]proline in

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Table 1. Stereoselective Reduction of the Amido Carbonyl Moiety of γ-Lactam 6

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run	lactam	deuteride	yield, %[a]	5R:5S[b]
1	**6a**	Bu_3SnD	57	81:19
2	**6a**	Et_3SiD	38	87:13
3	**6a**	Ph_3SiD	41	92:8
4	**6a**	$(TMS)_3SiD$	42	97:3
5	**6b**	$(TMS)_3SiD$	43	99:1

[a] Isolated yield based on the γ-lactam derivative. [b] Determined by ^1H NMR analysis.

57% yield based on the starting lactam **6a**, and the stereoselectivity of the deuterium substitution at the C-4 was determined by ^1H NMR integration to be 5R:5S = 81:19 (run 1). This stereochemical outcome can be attributed to a preferential delivery of the deuterium atom anti to the resident siloxymethylene group in the intermediate iminium ion produced from the aminal **7a**.

When deuteriosilanes were employed as a deuterium source instead of Bu_3SnD, a remarkable improvement of the stereoselectivity in the deuterium addition was observed (runs 2−4), especially in the case of tris(trimethylsilyl)deuteriosilane (TMS_3SiD, 5R:5S = 97:3). The advantage of changing the reducing agent from stannane to silane cannot be explained at the present stage; however, steric interaction between the silane and the siloxymethylene group is obviously responsible for the observed anti-selective deuteration. In fact, on replacing the TBDMS protecting group by the more bulky *tert*-butyldiphenylsilyl (TBDPS) group, the ratio of 5R:5S rose to 99:1 (run 5). Consequently, the reaction conditions shown in run 5 were adopted for the reduction of the amido carbonyl moiety of the deuterated lactam **2b**, giving the [3,4,5-D_3]prolinol derivative **3** in 51% yield. After removal of the TBDPS group by tetrabutylammonium fluoride (TBAF), oxidation and deprotection procedures afforded the [3,4,5-D_3]proline **5** in 66% yield. The optical purity was checked by HPLC analysis using a chiral stationary phase column to be 98% ee (L-form). The 400 MHz ^1H NMR spectrum of the proline **5** is depicted in Figure 1 (top). Coupled with the fact that the signal assigned to the 3S-proton completely disappeared, the relative intensity ratio for the 5R- and 5S-proton signals is 99:1, which indicates a stereospecific formation of the (2S,3S,4R,5R)-isomer.

To prepare the corresponding (5S)-isomer, changing the deuterium source in the reduction step seems to be the most convenient. We actually performed the reduction of the lactam **2b** using $LiEt_3BD$; however, the efficiency of the deuterium incorporation was unsatisfactory. The source of the hydrogen could not be specified.

On the other hand, our previous paper dealt with the preparation of (2S,3S,4R,5S)-[3,4,5-D_3]proline in which the 5S configuration was established on the basis of the syn-selective deuteration of the aminal derived from pyroglutamate derivatives, even though the selectivity was moderate (5R:5S = 10:90).[3] Therefore, we next carried out the interconversion of a siloxymethylene group into the *tert*-butyl ester prior to the reduction of the amido carbonyl moiety with optimization of the reaction conditions for the selective deuteration.

As shown in Scheme 3, deprotection of the silyl ether **2b** and subsequent oxidation of the alcohol **10** with RuO_4 gave the [3,4-D_2]pyroglutamic acid derivative **11** in quantitative yield. The *tert*-butyl ester **12** was obtained

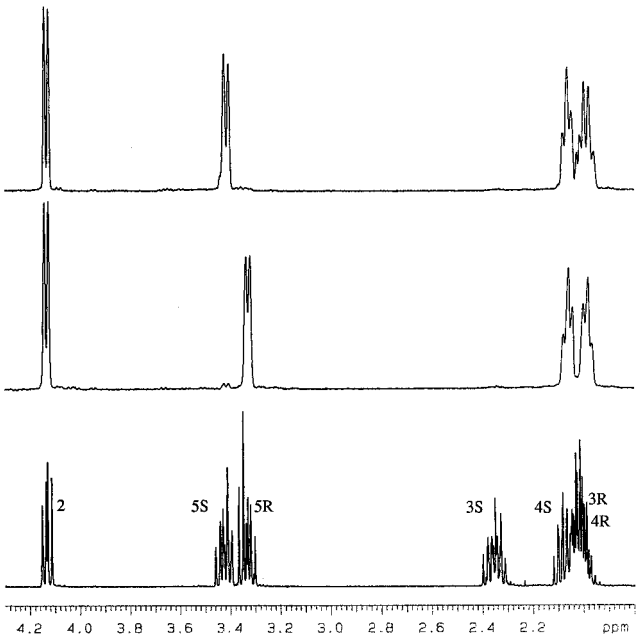
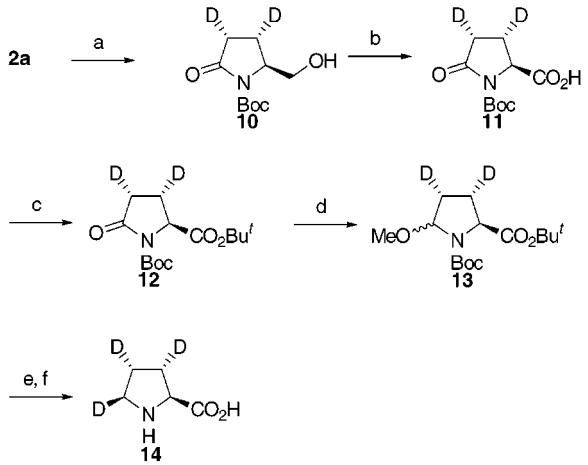


Figure 1. 400 MHz ^1H NMR spectra of (2S,3S,4R,5R)-[3,4,5-D_3]proline (**5**; top), (2S,3S,4R,5S)-[3,4,5-D_3]proline (**14**; middle), and unlabeled proline (bottom) in D_2O.

Scheme 3[a]



[a] Reagents and conditions: (a) TsOH, MeOH, 99%; (b) RuO_2, $NaIO_4$ quant; (c) $Me_2NCH(OCH_2CMe_3)_2$, *t*-BuOH, 50%; (d) $LiEt_3BH$, then TsOH, MeOH, 80% (two steps); (e) Bu_3SnD, $BF_3{\cdot}OEt_2$, in toluene; (f) 1 M HCl, 110 °C, then Dowex 50W-X8, 96% (two steps).

in 50% yield by reacting the acid **11** with dimethylformamide di-*tert*-butyl acetal prepared in situ from the corresponding dineopentyl acetal with *tert*-butyl alcohol.[8] The amido function was reduced at this stage. Thus, the [3,4-D_2]pyroglutamate **12** was treated with $LiEt_3BH$, followed by methanol in the presence of TsOH to afford 5-methoxy[3,4-D_2]prolinate **13** in 80% yield. After optimization of the reaction conditions for selective reduction of the aminal **13**, highly syn-selective deuteration (5R:5S = 3:97) was achieved by employment of a $Bu_3SnD-BF_3{\cdot}OEt_2$ system in toluene. This 1,3-stereocontrol can be attributed to the known Cieplak-type stereoelectronic effect[9] observed in a few Lewis acid-catalyzed addition reactions, although most of the reported syn additions

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were based on 1,2-stereocontrol directed by an adjacent OTBDMS group.[11,12] It was also pointed out in the foregoing works that reaction with a tin reagent in a toluene solution was the most efficient;[11] however, an explanation of the effects of the reagents and solvents awaits theoretical verification.

The obtained [3,4,5-D_3]prolinate, without purification, was submitted to the deprotection procedures to give [3,4,5-D_3]proline **14** in 96% yield with 92% ee (C-2). The 400 MHz ^{1}H NMR spectrum of the proline **14** is shown in Figure 1 (middle). The complete reversal of the relative intensities of the 5R- and 5S-proton signals also indicates the selective formation of the (2S,3S,4R,5S)-isomer.

In conclusion, we have achieved a stereodivergent synthesis of (2S,3S,4R,5R)- and (2S,3S,4R,5S)-[3,4,5-D_3]-proline starting from L-glutamic acid. The synthesis was based on the catalytic deuteration of the olefin **1** and the stereoselective reduction of the amido function under steric or stereoelectronic control depending on the substituent of the γ-lactam ring. Incorporation of a ^{13}C label into the deuterated proline framework has now become feasible using the corresponding ^{13}C-labeled L-glutamic acid as the starting material. Furthermore, the NMR spectra of selectively labeled proteins produced by incorporating such a proline will provide useful structural information. Studies that address these issues will be published in due course.

Experimental Section

General Procedures. ^{1}H and ^{13}C NMR spectra were measured at 400 and 100 MHz, respectively. All chemical shifts are reported as δ values (ppm) relative to residual chloroform (δ_H 7.26), sodium 3-(trimethylsilyl)[2,2,3,3-D_4]propionate (δ_H 0.00), or the central peak of deuteriochloroform (δ_C 77.0). High-resolution mass spectra (EI) were obtained at an ionization potential of 70 eV. Optical purity was determined on a HPLC system equipped with a chiral column and 2 mM $CuSO_4$ solution as an eluent. All other reagents were of commercial grade and used as supplied.

(5S)-*N*-*tert*-Butoxycarbonyl-5-*tert*-butyldimethylsiloxymethyl-2-[3,4-D_2]pyrrolidone (2a). A mixture of olefin **1a** (2.46 g, 7.51 mmol) and 10% Pd/C (491 mg) in MeOD (10 mL) was stirred at room temperature for 1 h under medium pressure (5 kgf/cm^2) of deuterium gas. After removal of the catalyst using a Celite pad, evaporation of the solvent gave 2-[3,4-D_2]pyrrolidone **2a** (2.27 g, 92%) as an oil. The structure was confirmed by comparing its spectral data with those of the known unlabeled compound.[6] ^{1}H NMR ($CDCl_3$) δ 0.02 (s, 3 H), 0.03 (s, 3 H), 0.86 (s, 9 H), 1 52 (s, 9 H), 1.98 (d, J = 9 Hz, 1 H), 2.66 (d, J = 10 Hz, 1 H), 3.67 (dd, J = 2 and 10 Hz, 1 H), 3.90 (dd, J = 4 and 10 Hz, 1 H), 4.15 (br, 1 H); HRMS m/z 216.1156 [(M − OTBDMS)$^+$, calcd for $C_{10}H_{14}D_2O_4N$ 216.1205].

(5S)-*N*-*tert*-Butoxycarbonyl-5-*tert*-butyldiphenylsiloxymethyl-2-[3,4-D_2]pyrrolidone (2b). Using the procedure described for the synthesis of compound **2a**, 2-[3,4-D_2]prrolidone **2b** (2.06 g) was obtained as an oil from the olefin **1b** (1.97 g, 4.37 mmol) in quantitative yield. The structure was confirmed by comparing its spectral data with those of the known unlabeled compound.[6] ^{1}H NMR ($CDCl_3$ δ 1.04 (s, 9 H), 1.43 (s, 9 H), 2.08 (d, J = 10 Hz, 1 H), 2.76 (d, J = 10 Hz, 1 H), 3.69 (dd, J = 2 and 11 Hz, 1 H), 3.89 (dd, 4 and 11 Hz, 1 H), 4.2 (br, 1 H), 7.41 (m, 6 H), 7.62 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 18.87, 20.36 (t, J = 20 Hz), 26.56, 27.73, 31.54 (J = 20 Hz), 58.39, 64.77, 82.20, 127.54 and 127.56, 129.57 (4 C), 140.98 and 141.33, 135.20 and 135.24, 149.57, 174.28; MS (EI) m/z 382 [(M − OBu^t)$^+$]; HRMS m/z 382.1820 [(M − OBu^t)$^+$, calcd for $C_{22}H_{24}D_2NO_3Si$ 382.1808].

(2S)-*N*-*tert*-Butoxycarbonyl-2-*tert*-butyldiphenylsiloxymethyl[3,4,5-D_3]pyrrolidine (3b). To a solution of 2-[3,4-D_2]-pyrrolidone **2b** (2.06 g, 4.37 mmol) in THF (45 mL) was added a solution of 1 M $LiEt_3BH$ in THF (5.2 mL, 5.2 mmol) at −78 °C under an Ar atmosphere, and the reaction mixture was stirred for 0.5 h. Then, the reaction was quenched with saturated aqueous $NaHCO_3$ (9 mL) at −78 °C, and the mixture was warmed to 0 °C. After addition of 30% H_2O_2 (2 mL), the mixture was stirred for an additional 20 min and concentrated to dryness. The residue was extracted with ether. The ethereal solution was washed with H_2O and dried over $MgSO_4$. Evaporation of the solvent gave a crude 5-hydroxy[3,4-D_2]prolinol derivative (2.08 g) in quantitative yield which was used in the next step without purification.

To a solution of the 5-hydroxy[3,4-D_2]prolinol (694 mg, 3.50 mmol) in CH_2Cl_2 (15 mL) was added tris(trimethylsilyl)deuteriosilane (870 mg, 3.50 mmol) and trifluoroborane etherate (454 mg, 3.20 mmol) in two portions at intervals of 0.5 h at −78 °C under an argon atmosphere, and the resulting solution was stirred for 2 h. Then the reaction mixture was quenched with saturated aqueous $NaHCO_3$ (7.5 mL), and the organic layer was dried over $MgSO_4$ and evaporated. After removal of the solvent, the residue was chromatographed on silica gel. Elution with a mixture of hexane and ethyl acetate (96:4) afforded oily [3,4,5-D_3]prolinol **3b** (350 mg, 51%): ^{1}H NMR ($CDCl_3$) δ 1.04 (s, 9 H), 1.33 and 1.45 (2 s, 9 H), 1.77 and 1.88 (2 m, 1 H), 2.01 and 2.10 (2 m, 1 H), 3.32 (m, 1 H), 3.51 and 3.77 (2 m, 1 H), 3.71 (m, 1 H), 3.84 and 3.95 (2 m, 1 H), 7.38 (m, 6 H), 7.65 (m, 4 H); ^{13}C NMR ($CDCl_3$) δ 19.28, 22.65 (br), 26.88, 27.84 (br), 28.51, 46.55 (br), 58.28, 64.67 (br), 79.03 (br), 127.66 (4 C), 129.60 (4 C), 133.70, 133.82, 135.58, 135.60, 154.53; MS (EI) m/z 442 (M$^+$); HRMS m/z 442.2703 (M$^+$, calcd for $C_{26}H_{34}D_3NO_3Si$ 442.2731).

(2S)-*N*-*tert*-Butoxycarbonyl-2-hydroxymethyl[3,4,5-D_3]-pyrrolidine (4). To a solution of compound **3b** (325 mg, 0.735 mmol) in THF (5 mL) was added 1 M Bu_4NF in THF (1.6 mL, 1.60 mmol), and the resulting solution was stirred at room temperature for 1.5 h. After removal of the solvent, the residue was chromatographed on silica gel. Elution with a mixture of hexane and ethyl acetate (70:30) afforded oily [3,4,5-D_3]prolinol **4** (126 mg, 84%). The structure was confirmed by comparing its spectral data with those of the commercially available unlabeled compound. ^{1}H NMR ($CDCl_3$) δ 1.47 (s, 9 H), 1.52 and 1.80 (2 m, 2 H), 3.29 (d, J = 7 Hz, 1 H), 3.58 (dd, J = 11 and 8 Hz, 1 H), 3.63 (dd, J = 11 and 3 Hz, 1 H), 3.95 (br, 1 H); ^{13}C NMR ($CDCl_3$) δ 23.29 (t, J = 19 Hz), 28.16 (t, J = 20 Hz), 28.38, 46.97 (t, J = 20 Hz), 59.93, 67.22, 79.94, 156.85; MS (EI) m/z 204 (M$^+$); HRMS m/z 173.1387 [(M − CH_2OH)$^+$, calcd for $C_9H_{13}D_3O_2N$ 173.1369].

(2S,3S,4R,5R)-[3,4,5-D_3]Proline (5). To a solution of [3,4,5-D_3]prolinol **4** (126 mg, 0.608 mmol) in acetone (10 mL) was added Jones reagent, prepared from CrO_3 (243 mg, 2.43 mmol), sulfuric acid (0.2 mL), and H_2O (0.5 mL), and the resultant suspension was stirred at room temperature for 0.5 h. Then the reaction mixture was quenched by 2-propanol (0.2 mL), and the insoluble materials were filtered off. The organic layer was separated, diluted with ethyl acetate (100 mL), washed with H_2O (20 mL × 5) and brine (20 mL), dried over $MgSO_4$, and evaporated. Deprotection of the resultant crude *N*-*tert*-butoxycarbonyl[3,4,5-D_3]proline was carried out in 1 M HCl (15 mL) at 110 °C for 3 h followed by treatment with Dowex 50W-X8 to give [3,4,5-D_3]-proline **5** (47 mg, 66%) as a colorless solid, mp 207−213 °C dec. The spectral data were identical with those reported by us.[3] ^{1}H NMR (D_2O) δ 1.97 (dd, J = 8 and 7 Hz, 1 H), 2.05 (dd, J = 7 and 7 Hz, 1 H), 3.32 (d, J = 7 Hz, 0.01 H), 3.41 (d, J = 7 Hz, 0.99 H), 4.13 (d, J = 7 Hz, 1 H).

***tert*-Butyl (2S,3S,4R)-*N*-*tert*-Butoxycarbonyl[3,4-D_2]pyroglutamate (12).** To a solution of compound **2a** (1.97 g, 6.02 mmol) in MeOH (60 mL) was added *p*-toluenesulfonic acid (103 mg, 0.6 mmol), and the resulting solution was stirred at room temperature overnight. After removal of the solvent, the residue was extracted with ethyl acetate. The organic layer was washed with saturated aqueous $NaHCO_3$ and dried over $MgSO_4$. Evaporation of the solvent afforded oily 5-hydroxymethyl[3,4-D_2]-pyrrolidone **10** (1.30 g, 99%), which was used in the next step without purification. ^{1}H NMR ($CDCl_3$) δ 1.55 (s, 9 H), 1.94 (dd, J = 2 and 10 Hz, 1 H), 2.31 (br t, J = 6 Hz, 1 H), 2.66 (d, J = 10

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(12) Thaning, M.; Winstrand, L.-G. *J. Org. Chem.* **1990**, *55*, 1406−1408; Jeroncic, L. O.; Cabal, M. P.; Danishefsky, S. J.; Shulte, G. M. *J. Org. Chem.* **1991**, *56*, 387−395.

Hz, 0.99 H), 3.74 (ddd, J = 4, 6 and 11 Hz, 1 H), 3.86 (ddd, J = 4, 6 and 11 Hz, 1 H), 4.23 (dt, J = 2 and 4 Hz, 1 H).

To a suspension of sodium metaperiodate (3.34 g, 15.6 mmol) and $RuCl_3 \cdot nH_2O$ (110 mg) in H_2O (9 mL) was added a solution of compound **10** (340 mg, 1.56 mmol) in acetone (15 mL). The resulting two-phase mixture was vigorously stirred at room temperature for 1 h. The layers were separated, to the organic phase was added 2-propanol (4.6 mL), and the mixture was stirred for 1 h. After removal of the precipitated RuO_2 using a Celite pad, the filtrate was extracted with chloroform, concentrated, and dried over $MgSO_4$. Evaporation of the solvent gave [3,4-D_2]pyroglutamic acid **11** (362 mg) in quantitative yield as a colorless solid: mp 117−120 °C; ^{1}H NMR ($CDCl_3$) δ 1.50 (s, 9 H), 2.11 (dd, J = 9 and 3 Hz, 1 H), 4.64 (d, J = 3 Hz, 1 H), 6.10 (br, 1 H).

To a refluxing solution of the crude [3,4-D_2]pyroglutamic acid **11** (195 mg, 0.84 mmol) in benzene (5 mL) was added a mixture of *N*,*N*-dimethylformamide dineopentyl acetal (352 mg, 1.51 mmol) and *tert*-butyl alcohol (186 mg, 2.52 mmol), and the reaction mixture was stirred for 0.5 h. Then the cooled reaction mixture was diluted with ethyl acetate (30 mL), washed with saturated aqueous $NaHCO_3$ and brine, and dried over $MgSO_4$. After removal of the solvent, the residue was chromatographed on silica gel. Elution with a mixture of hexane and ethyl acetate (90:10) afforded oily *tert*-butyl [3,4-D_2]pyroglutamate **12** (120 mg, 50%). The spectral data were identical with those reported by us.[3] ^{1}H NMR ($CDCl_3$) δ 1.47 (s, 9 H), 1.49 (s, 9 H), 1.96 (dd, J = 10 and 2 Hz, 1 H), 2.57 (d, J = 10 Hz, 1 H), 4.46 (d, J = 2 Hz, 1 H); ^{13}C NMR ($CDCl_3$) δ 21.19 (t, J = 21 Hz), 27.85 (6 C), 30.64 (t, J = 21 Hz), 59.47, 82.10, 83.09, 149.37, 170.32, 173.11.

***tert*-Butyl (2S,3S,4R,5RS)-*N*-*tert*-Butoxycarbonyl-5-methoxy[3,4-D_2]prolinate (13).** To a solution of *tert*-butyl [3,4-D_2]-pyroglutamate **12** (303 mg, 1.06 mmol) in THF (10 mL) was added a solution of 1 M $LiEt_3BH$ in THF (1.27 mL, 1.27 mmol) at −78 °C under an Ar atmosphere, and the reaction mixture was stirred for 0.5 h. Then, the reaction was quenched with saturated aqueous $NaHCO_3$ (5 mL) at −78 °C, and the mixture was warmed to 0 °C. After addition of 30% H_2O_2 (1 mL), the mixture was stirred for an additional 30 min and concentrated. The residue was extracted with ether, washed with H_2O, and dried over $MgSO_4$. After removal of the solvent, the crude *tert*-butyl 5-hydroxyprolinate was directly treated with MeOH (10 mL) in the presence of *p*-toluenesulfonic acid (17.1 mg, 0.0996 mmol) for 1 h. The concentrated reaction mixture was diluted with ethyl acetate (50 mL). The organic layer was washed three times with saturated aqueous $NaHCO_3$ (10 mL) and dried over $MgSO_4$. After removal of the solvent, the residue was chromatographed on silica gel. Elution with a mixture of hexane and ethyl acetate (96:4) afforded oily 5-methoxy[3,4-D_2]prolinate **13** (256 mg, 80%) as unseparable diastereoisomers. The spectral data were identical with those reported by us.[3] ^{1}H NMR ($CDCl_3$) δ 1.437, 1.440, 1.45, 1.46, 1.48, and 1.49 (6 s, 18 H), 1.84−2.11 (m, 2 H), 3.34, 3.39, 3.40, and 3.43 (4 s, 3 H), 4.14−4.20 (m, 1 H), 5.12−5.29 (m, 1 H).

(2S,3S,4R,5S)-[3,4,5-D_3]Proline (14). To a solution of 5-methoxy[3,4-D_2]prolinate **13** (78.0 mg, 0.257 mmol) in toluene (10 mL) were added Bu_3SnD (178 mg, 0.616 mmol) and trifluoroborane etherate (84 mg, 0.566 mmol) in two portions at intervals of 0.5 h at −78 °C under an argon atmosphere, and the resulting solution was stirred for 2 h. Then the reaction mixture was quenched with saturated aqueous $NaHCO_3$ (5 mL), and the organic layer was dried over $MgSO_4$ and evaporated. The deprotection of the resultant crude [3,4,5-D_3]prolinate was carried out in 1 M HCl (15 mL) at 110 °C for 3 h followed by a treatment with Dowex 50W-X8 to give [3,4,5-D_3]proline **14** (29.0 mg, 96%) as a colorless solid, mp 223−225 °C dec. The spectral data were identical with those reported by us.[3] ^{1}H NMR (D_2O) δ 1.97 (dd, J = 7 and 7 Hz, 1 H), 2.05 (dd, J = 7 and 7 Hz, 1 H), 3.32 (d, J = 7 Hz, 0.97 H), 3.41 (d, J = 7 Hz, 0.03 H), 4.13 (d, J = 7 Hz, 1 H).

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