

HETEROCYCLES, Vol. 61, 2003, pp. 259 - 269

Received, 26th June, 2003, Accepted, 1st August, 2003, Published online, 4th August, 2003

HEXAFLUOROACETONE AS PROTECTING AND ACTIVATING REAGENT: AN EFFICIENT STRATEGY FOR ACTIVATION OF PYROGLUTAMIC ACID AND HOMOLOGUES

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Abstract - Hexafluoroacetone-protected glutamic acid on treatment with thionyl chloride is transformed into a carboxy-activated pyroglutamic acid derivative. Likewise, the reaction sequence can be applied to aminoadipic acid and homologues.

INTRODUCTION

Pyroglutamic acid and its derivatives are important amino acids, since they are constituents of many bioactive compounds like azaprostaglandin analogues,¹ monocyclic thienyl- γ -lactam² and thioliberin (TRH).³ Pyroglutamic acid and substituted pyroglutamic acid derivatives are interesting targets as they confer unique structural constraints in peptide chains⁴ and hence may play a major role in protein folding. Furthermore, glutamic acid which can be derived from pyroglutamic acid acts as one of the major neurotransmitters at excitatory synapses in the mammalian central nervous system (CNS).⁵ Substituted 2-pyrrolidinones are an important class of nitrogen-containing heterocycles.⁶ They exhibit a variety of biological activities,⁷ *i.a.* they have been applied for treatment of brain and cognitive problems,⁸ as anticonvulsant agents,⁹ and as inhibitors for HIV-1 replication.¹⁰ Furthermore, they have been used as intermediates in the synthesis of alkaloids,¹¹ α -amino¹² and γ -amino acids.¹³ Therefore, the development of new methodology for incorporation of pyroglutamic

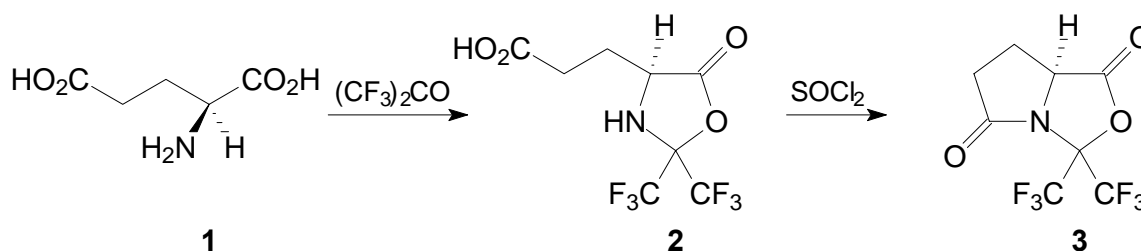
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** Dedicated to Prof. J. Sieler on the occasion of his 65th birthday.

acid into target molecules is of current interest, especially when the reaction sequence is suitable for generation of combinatorial libraries.

RESULTS AND DISCUSSION

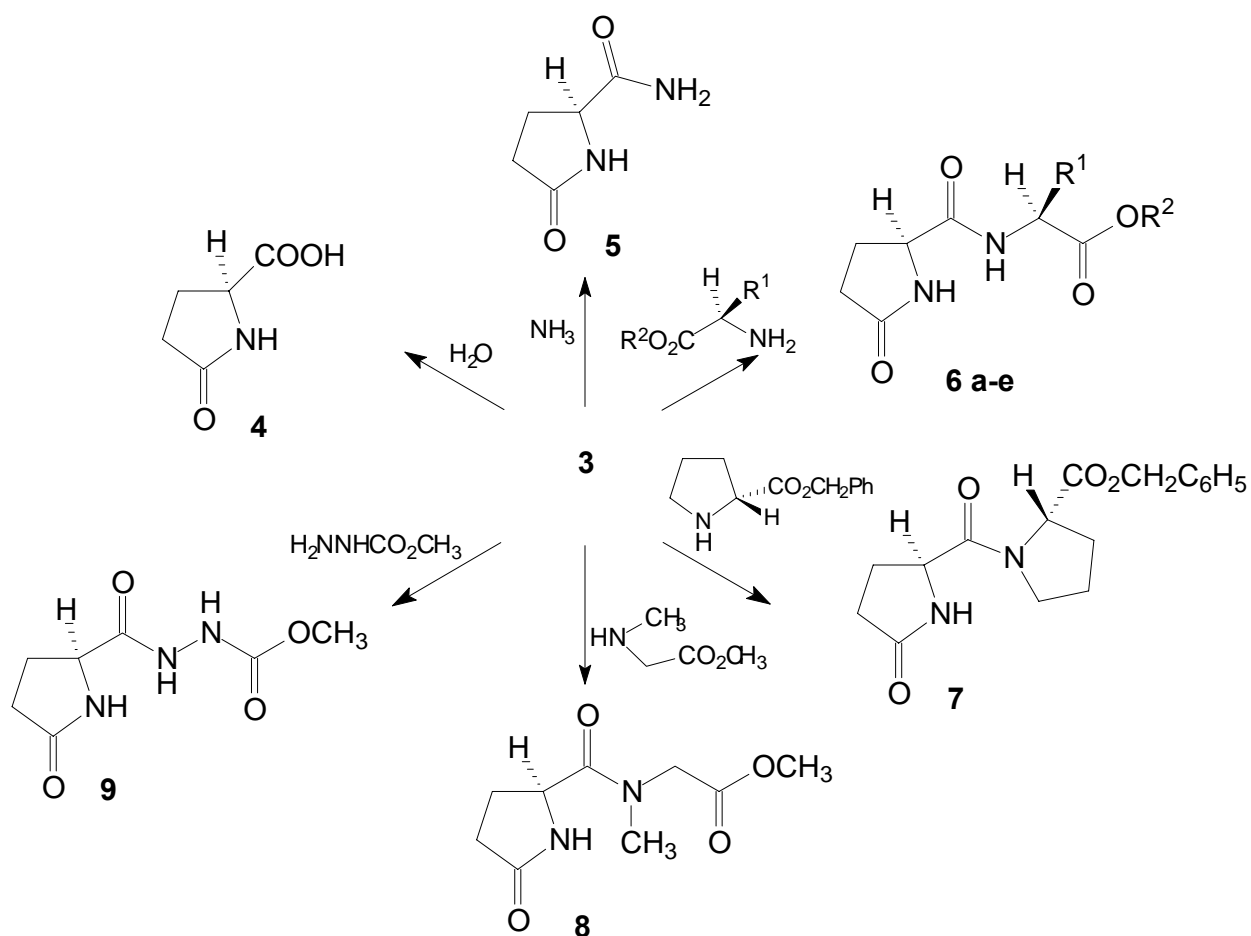
Hexafluoroacetone (HFA) (2 equivalents) reacts readily with (*S*)- and (*R*)-glutamic acids in DMSO at room temperature to give 2,2-bis(trifluoromethyl)-1,3-oxazolidin-5-one (**2**).^{14,15} The second equivalent of hexafluoroacetone is necessary to trap the water which is eliminated during lactone formation. ¹⁹F NMR spectral control of the reaction indicates that the five-membered lactone is formed quantitatively, but there is some loss during work-up, because of the water solubility of **2**. Hexafluoroacetone hydrate is formed as the only by-product. With the formation of the five-membered heterocyclic ring system, amino group protection and selective activation of the α -carboxy group are achieved in one step. The ω -carboxy group remains unaffected and can be derivatized regioselectively after separate activation.¹⁶



Scheme 1

There exist two fundamentally different strategies for the incorporation of the pGlu moiety into peptides: Incorporation of Glu into the peptide chain and construction of the 2-pyrrolidinone ring in a consecutive step,¹⁷ or incorporation of the complete pGlu moiety. When we tried to activate the ω -carboxy group of HFA-protected Glu selectively on treatment with thionyl chloride we obtained the HFA-protected pGlu (**3**) in almost quantitative yield. The presence of a lactone as well as a lactam function can be identified by IR absorptions at $\nu = 1855$ and 1755 cm^{-1} , respectively. The two diastereotopic trifluoromethyl groups bound to the C-(2) atom of the oxazolidin-5-one ring resonate at $\delta = -1.99$ (q, $J = 8.3$ Hz, 3F) and 5.76 (q, $J = 8.3$ Hz, 3F) ppm in the ¹⁹F NMR spectrum. **3** is a crystalline compound, easy to handle. It can be stored without decomposition in a fridge for months on exclusion of water.

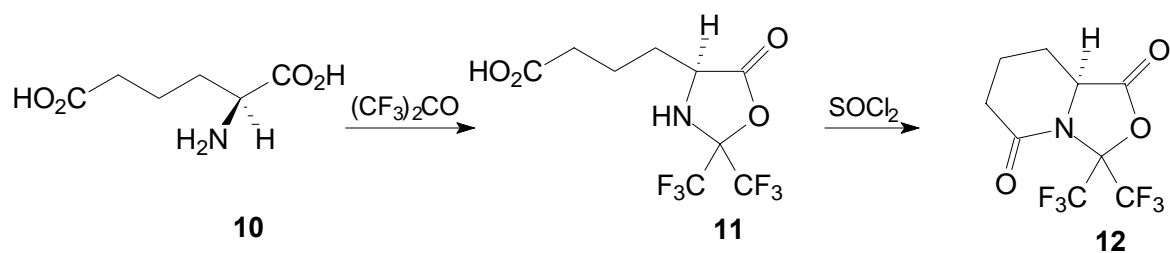
Compound (**3**) represents an activated ester and reacts readily with nucleophiles like water, alcohols and amines to give the corresponding acid, esters and amides in very good yields (56-90%), respectively.



	R¹	R²
6a	CH ₃	C ₂ H ₅
6b	CH(CH ₃) ₂	CH ₃
6c	CH ₂ C ₆ H ₅	CH ₃
6d	<i>p</i> -HOC ₆ H ₄ CH ₂	CH ₃
6e	CH ₂ CO ₂ C ₂ H ₅	C ₂ H ₅

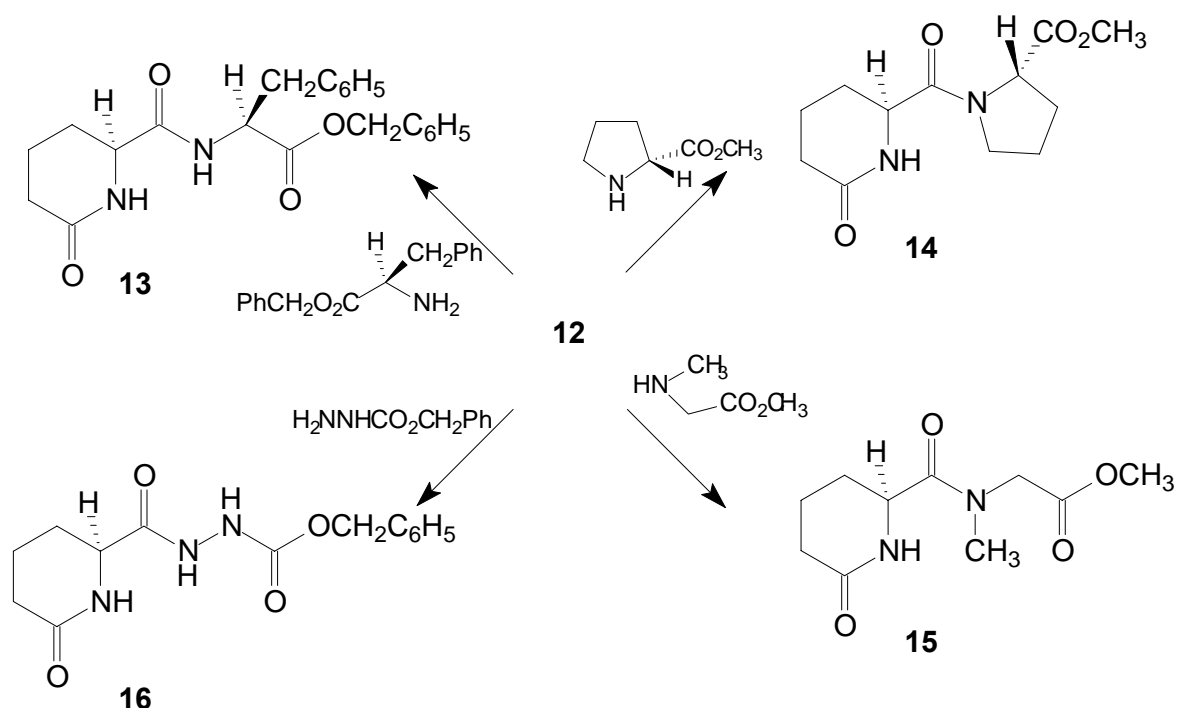
Scheme 2

Dipeptides and dipeptide surrogates are obtainable in high yields on reaction with amino acid esters, *N*-alkylamino acid esters and *N*-protected amino acid hydrazides, respectively. Dipeptide (**8**) formed on reaction of **3** with sarcosine methyl ester exists in solution (CDCl₃) at room temperature as a mixture of rotamers (ratio 5:1).



Scheme 3

α -Aminoadipic acid (**10**) and hexafluoroacetone react analogously to give a 2,2-bis(trifluoromethyl)-1,3-oxazolidin-5-one (**11**). On treatment with thionyl chloride **11** is converted into an acid chloride which spontaneously undergoes ring closure to give a bicyclic six-membered lactam (**12**). The chemistry of **11** and **12** parallels that of **2** and **3**. Best yields are obtained, when appropriate solvents are used where the products crystallize spontaneously. In general the products are analytically pure after careful trituration with dry diethyl ether.



Scheme 4

EXPERIMENTAL

Melting points were determined on a Boetius heating table. IR spectra were obtained with a FTIR spectrometer (Genesis ATI Mattson/Unicam). ^1H NMR spectra were recorded with VARIAN Gemini 2000 spectrometers at 200 and 300 MHz. Chemical shifts were reported in ppm relative to tetramethylsilane (TMS, $\delta = 0$ ppm); J values are given in Hertz (Hz). ^{13}C NMR spectroscopy was performed at 50 and 75 MHz. ^{19}F NMR spectra were recorded at 188 and 282 Hz with trifluoroacetic acid (TFA, $\delta = 0$ ppm) as external standard. Optical rotations ($[\alpha]_D$) were measured using a Polarotronic polarimeter (Schmidt & Haensch) in a 5 cm cell. For C, H, N analyses a CHNO-Rapid-Elemental-Analyzer (Heraeus) was used. For flash chromatography, silica gel (32-63 μm) was used with solvent systems given in the text. Organic solvents were dried and distilled prior to use.

(S)-[2,2-Bis(trifluoromethyl)-5-oxo-1,3-oxazolidin-4-yl]propionic acid (2). (S)-Glutamic acid (**1**) (14.71 g, 100 mmol) was reacted for 6-8 h in DMSO (50 mL) with an excess of hexafluoroacetone at rt in a glass apparatus sealed with a dry-ice condenser. When the reaction was complete, water (500 mL) was added and the reaction mixture was extracted with CH₂Cl₂ (4 x 50 mL). The organic phase was extracted with water (3 x 50 mL), dried with MgSO₄ and evaporated to dryness. Yield 19.78 g (67%), mp 117 - 118 °C (CH₂Cl₂), [α]_D = +6.4° (c = 1.1, DMSO). IR (KBr) ν 3355, 1810, 1700 cm⁻¹. ¹H NMR (acetone-d₆) δ 1.91 (1H, m), 2.17 (1H, m), 2.46 - 2.61 (2H, m), 4.30 (1H, m), 5.31 (1H, d, *J* = 6.5 Hz). ¹³C NMR (acetone-d₆) δ 29.0, 29.6, 54.3, 89.3 (qq, *J* = 34.0 Hz, *J* = 34.0 Hz), 121.5 (q, *J* = 285.0 Hz), 122.4 (q, *J* = 288.0 Hz), 172.3, 174.0. ¹⁹F NMR (acetone-d₆) δ -2.02 (3F, q, *J* = 9.0 Hz), -2.96 (3F, q, *J* = 9.0 Hz). MS *m/z* = 295 [M]⁺, 277 [M - H₂O]⁺, 249 [M - H₂O, - CO]⁺, 235 [M - H₂O, - CO, - CH₂]⁺, 180 [M - H₂O, - CO, - CF₃]⁺, 166 [M - H₂O, - CO, - CH₂, - CF₃]⁺, 152 [M - H₂O, - CO, - CH₂, - CF₃, - CH₂]⁺, 136 [M - H₂O, - CO, - CF₃, - CO₂]⁺, 85 [CH₂CH₂CHCO₂]⁺, 69 [CF₃]⁺. Anal. Calcd for C₈H₇NO₄F₆: C, 32.56; H, 2.39; N, 4.75. Found: C, 32.64; H, 2.54; N, 4.78.

(5S)-2,2-Bis(trifluoromethyl)-1-aza-3-oxabicyclo[3.3.0]octan-4,8-dione (3). To a solution of (4S)-3-[2,2-bis(trifluoromethyl)-5-oxo-1,3-oxazolidin-4-yl]propionate (**2**) (10.44 g, 35.4 mmol) in CH₂Cl₂ (100 mL) four drops of dimethyl formamide were added. After cooling to 0 °C to the stirred reaction mixture thionyl chloride (5.95 g, 50 mmol) was added dropwise. Stirring was continued at rt for 16 - 24 h. The progress of the reaction was monitored by ¹⁹F NMR spectroscopy. Then the excess of the thionyl chloride and of the solvent was evaporated *in vacuo*, the remaining solid was recrystallized from hexane. Yield 9.66 g (99%), mp 85 - 86 °C (hexane), [α]_D = +24.0° (c = 1.0, CHCl₃). IR (KBr) ν 1855, 1755 cm⁻¹. ¹H NMR (CDCl₃) δ 2.28 (1H, m), 2.60 - 2.72 (2H, m), 2.87 (1H, ddd, *J* = 17.2 Hz, *J* = 12.9 Hz, *J* = 8.3 Hz), 4.68 (1H, dd, *J* = 10.1 Hz, *J* = 7.3 Hz). ¹³C NMR (CDCl₃) δ 24.9, 34.2, 57.7, 88.0 (qq, *J* = 36.0 Hz, *J* = 36.0 Hz), 118.8 (q, *J* = 289.0 Hz), 120.0 (q, *J* = 288.0 Hz), 166.5, 172.1. ¹⁹F NMR (CDCl₃) δ -1.99 (3F, q, *J* = 8.3 Hz), 5.76 (3F, q, *J* = 8.3 Hz). MS *m/z* = 277 [M]⁺, 233 [M - CO₂]⁺, 208 [M - CF₃]⁺, 180 [M - CO, - CF₃]⁺, 152 [M - CO, - CF₃, - CO]⁺, 83 [M - CO, - (CF₃)₂CO]⁺, 69 [CF₃]⁺, 55 [M - CO, - (CF₃)₂CO, - CO]⁺. Anal. Calcd for C₈H₅NO₃F₆: C, 34.67; H, 1.81; N, 5.05. Found: C, 34.74; H, 1.94; N, 5.10.

(S)-2-Pyrrolidinone-5-carboxylate (4). A solution of **3** (2.77 g, 10 mmol) in 2-propanol/ water (40 mL, 1:1) was stirred at rt until the starting material was consumed (¹⁹F NMR analysis). The solvent was evaporated *in vacuo*, then the crystalline residue was carefully triturated with dry ether and filtered off. Yield 1.12 g (87%), mp 158 - 159 °C (ether) (lit.,¹⁸ mp 155-157 °C), [α]_D = -15.0° (c = 1.0, H₂O), [α]_D = -12.5° (c = 2.0, H₂O) (lit.,¹⁹ [α]_D = -10.1° (c = 5.0, H₂O)). IR (KBr) ν 3410, 1725, 1650, 1235 cm⁻¹. ¹H

NMR (DMSO- d_6) δ 1.97 (1H, m), 2.06 - 2.21 (2H, m), 2.33 (1H, m), 4.67 (1H, dd, $J = 8.9$ Hz, $J = 4.3$ Hz), 7.93 (1H, s, NH). ^{13}C NMR (DMSO- d_6) δ 24.7, 29.2, 54.9, 174.6, 177.3. MS $m/z = 129$ $[\text{M}]^+$, 84 $[\text{M} - \text{COOH}]^+$, 56 $[\text{M} - \text{COOH}, - \text{CO}]^+$, 41 $[\text{M} - \text{COOH}, - \text{CO}, - \text{NH}]$. Anal. Calcd for $\text{C}_5\text{H}_7\text{NO}_3$: C, 46.51; H, 5.46; N, 10.85. Found: C, 46.10; H, 5.76; N, 10.78.

(S)-2-Pyrrolidinone-5-carboxamide (5). A solution of **3** (2.77 g, 10 mmol) in 2-propanol (20 mL) was stirred for 6 d with conc. ammonia (20 mL) at rt until the starting material was consumed (^{19}F NMR analysis). After evaporation of the solvent *in vacuo*, the residue was triturated with ethyl acetate and then recrystallized from methanol. Yield 0.76 g (59%), mp 148 - 150 °C (methanol) (lit.,²⁰ mp 168 °C), $[\alpha]_{\text{D}} = -42.5^\circ$ ($c = 1.0$, H_2O). IR (KBr) ν 3460, 3400, 3210, 1700 - 1665, 1650 cm^{-1} . ^1H NMR (DMSO- d_6) δ 1.85 (1H, m), 2.01 - 2.29 (3H, m), 3.93 (1H, dd, $J = 8.2$ Hz, $J = 4.5$ Hz), 7.07 (1H, s, NH), 7.41 (1H, s, NH), 7.77 (1H, s, NH). ^{13}C NMR (DMSO- d_6) δ 25.4, 29.4, 55.7, 174.7, 177.7. MS $m/z = 128$ $[\text{M}]^+$, 84 $[\text{M} - \text{CONH}_2]^+$, 56 $[\text{M} - \text{CONH}_2, - \text{CO}]^+$. Anal. Calcd for $\text{C}_5\text{H}_8\text{N}_2\text{O}_2$: C, 46.87; H, 6.29; N, 21.86. Found: C, 46.56; H, 5.89; N, 21.72.

Pyroglutamyl dipeptides (6), general procedure. To a stirred solution of **3** in ether (or ethyl acetate) 1 - 1.5 equivalents of the corresponding C-terminal protected amino acid was added. The progress of the reaction is controlled by ^{19}F NMR spectroscopy. Crystalline products were filtered off and carefully triturated with dry ether and afterwards dried *in vacuo*. If the dipeptide does not crystallize from the crude reaction mixture, the solvent is evaporated *in vacuo*. The crude products are purified by reversed phase chromatography.

(S)-Pyroglutamyl-(S)-alanine ethyl ester (6a). **3** (1.00 g, 3.6 mmol) was reacted for 24 h with (S)-alanine ethyl ester (0.70 g, 6.0 mmol) in ether (20 mL). Yield 0.72 g (88%), mp 113 °C (ether), $[\alpha]_{\text{D}} = -76.0^\circ$ ($c = 2.0$, H_2O). IR (KBr) ν 3310, 1740, 1700, 1670, 1545 cm^{-1} . ^1H NMR (CDCl_3) δ 1.28 (3H, t, $J = 7.2$ Hz), 1.42 (3H, d, $J = 7.3$ Hz), 2.13 - 2.62 (4H, m), 4.19 (2H, q, $J = 7.2$ Hz), 4.21 (1H, m), 4.60 (1H, dq, $J = 7.5$ Hz, $J = 7.3$ Hz), 7.45 (1H, br s, NH), 7.56 (1H, d, $J = 7.7$ Hz, NH). ^{13}C NMR (CDCl_3) δ 14.1, 17.9, 25.7, 29.4, 48.0, 57.4, 61.7, 172.3, 173.4, 179.7. MS $m/z = 228$ $[\text{M}]^+$, 183 $[\text{M} - \text{OC}_2\text{H}_5]^+$, 155 $[\text{M} - \text{OCOC}_2\text{H}_5]^+$, 84 $[\text{C}_4\text{H}_6\text{NO}]^+$; Anal. Calcd for $\text{C}_{10}\text{H}_{16}\text{N}_2\text{O}_4$: C, 52.62; H, 7.07; N, 12.27 Found: C, 52.38; H, 7.07; N, 12.25.

(S)-Pyroglutamyl-(S)-valine methyl ester (6b). To a solution of (S)-valine methyl ester (1.27 g, 9.7 mmol) in ethyl acetate (30 mL) a solution of **3** (2.69 g, 9.7 mmol) in ethyl acetate (10 mL) was added.

The reaction mixture was stirred for 3 d. **6b** was purified by reversed phase chromatography (eluent: water/methanol, 3:1). Yield 1.56 g (66%), mp 99 - 100 °C (water/methanol), $[\alpha]_D = -45.5^\circ$ ($c = 2.0$, H₂O). IR (KBr) ν 3500 - 2900, 1740, 1700 - 1650 cm⁻¹. ¹H NMR (CDCl₃) δ 0.94 (3H, d, $J = 7.0$ Hz), 0.95 (3H, d, $J = 7.0$ Hz), 2.14 - 2.55 (5H, m), 3.73 (3H, s), 4.29 (1H, dd, $J = 8.6$ Hz, $J = 5.3$ Hz), 4.53 (1H, dd, $J = 8.8$ Hz, $J = 5.9$ Hz), 7.55 (1H, d, $J = 8.8$ Hz, NH), 7.66 (1H, s, NH). ¹³C NMR (CDCl₃) δ 17.9, 18.8, 25.5, 29.3, 30.8, 52.0, 57.1, 57.2, 172.6, 172.7, 179.5. MS $m/z = 242$ [M]⁺, 211 [M - OCH₃]⁺, 183 [M - COOCH₃]⁺, 85 [C₄H₇NO]⁺, 84 [C₄H₆NO]⁺. Anal. Calcd for C₁₁H₁₈N₂O₄: C, 54.53; H, 7.49; N, 11.56. Found: C, 54.72; H, 7.40; N, 11.48.

(S)-Pyroglutamyl-(S)-phenylalanine methyl ester (6c). **3** (1.0 g, 3.6 mmol) and (S)-phenylalanine methyl ester (1.12 g, 6.2 mmol) were reacted for 3 d in ether (30 mL). Yield 0.80 g (77%), mp 74 °C (ether), $[\alpha]_D = -31.0^\circ$ ($c = 2.0$, H₂O). IR (CHCl₃) ν 3340, 1732, 1711, 1658 cm⁻¹. ¹H NMR (CDCl₃) δ 1.76 - 1.79 (1H, m), 2.16 - 2.34 (3H, m), 3.01 (1H, dd, $J = 13.8$ Hz, $J = 9.1$ Hz), 3.28 (1H, dd, $J = 13.8$ Hz, $J = 5.2$ Hz), 3.73 (3H, s), 4.08 (1H, m), 4.93 (1H, ddd, $J = 9.1$ Hz, $J = 8.6$ Hz, $J = 5.2$ Hz), 7.18 - 7.30 (6H, m), 7.51 (1H, $J = 8.6$ Hz, NH). ¹³C NMR (CDCl₃) δ 25.6, 29.0, 37.6, 52.6, 53.0, 57.2, 127.1, 128.5, 129.2, 136.2, 172.6, 172.7, 179.9. MS $m/z = 290$ [M]⁺, 246 [M - CO₂]⁺, 162 [M - CO₂, - C₄H₆NO]⁺, 84 [C₄H₆NO]⁺. Anal. Calcd for C₁₅H₁₈N₂O₄: C, 62.06; H, 6.25; N, 9.65. Found: C, 61.79; H, 6.33; N, 9.42.

(S)-Pyroglutamyl-(S)-tyrosine methyl ester (6d). **3** (0.83 g, 3.0 mmol) and (S)-tyrosine methyl ester (0.59 g, 3.0 mmol) were reacted for 3 d in ether (30 mL). Yield 0.59 g (64%), mp 236 °C (ether), $[\alpha]_D = -27.0^\circ$ ($c = 1$, H₂O). IR (KBr) ν 3600 - 2900, 1725, 1710, 1640, 1520 cm⁻¹. ¹H NMR (DMSO-d₆) δ 1.77 (1H, m), 2.01 - 2.08 (2H, m), 2.21 (1H, m), 2.81 (1H, dd, $J = 13.8$ Hz, $J = 9.1$ Hz), 2.92 (1H, dd, $J = 13.8$ Hz, $J = 5.5$ Hz), 3.59 (3H, s), 3.99 (1H, dd, $J = 8.8$ Hz, $J = 3.8$ Hz), 4.40 (1H, m), 6.65 (2H, m), 6.99 (2H, m), 7.75 (1H, s, NH), 8.31 (1H, d, $J = 7.8$ Hz), 9.23 (1H, br s, NH). ¹³C NMR (DMSO-d₆) δ 25.3, 29.0, 35.8, 51.9, 53.9, 55.3, 115.1, 127.2, 130.1, 156.1, 172.0, 172.8, 177.6. MS $m/z = 306$ [M]⁺, 247 [M - COOCH₃]⁺, 178 [HOC₆H₄CHCHCOOCH₃]⁺, 84 [C₆H₄NO]⁺. Anal. Calcd for C₁₅H₁₈N₂O₅ x H₂O: C, 55.55; H, 6.22; N, 8.64. Found: C, 55.71; H, 5.99; N, 8.30.

(S)-Pyroglutamyl-(S)-aspartic acid diethyl ester (6e). **3** (1.0 g, 3.6 mmol) and aspartic acid diethyl ester (1.05 g, 5.5 mmol) were reacted for 48 h in ether (20 mL). Yield 0.77 g (71%), mp 74 - 75 °C (ether), $[\alpha]_D = -35.0^\circ$ ($c = 1.0$, H₂O). IR (KBr) ν 3320, 2990, 1750 - 1650 cm⁻¹. ¹H NMR (CDCl₃) δ 1.25 (3H, t, $J = 7.1$ Hz), 1.27 (3H, t, $J = 7.2$ Hz), 2.15 - 2.55 (4H, m), 2.88 (1H, dd, $J = 16.8$ Hz, $J = 4.8$ Hz), 2.99 (1H, dd, $J = 16.8$ Hz, $J = 5.7$ Hz), 4.14 (2H, q, $J = 7.1$ Hz), 4.17 - 4.23 (3H, m), 4.85 - 4.89 (1H, m), 7.14 (1H,

br s), 7.45 (1H, d, $J = 8.2$ Hz). ^{13}C NMR (CDCl_3) δ 13.9, 14.0, 25.7, 29.2, 36.1, 48.7, 56.9, 61.1, 61.9, 170.6, 170.7, 172.3, 179.3. MS $m/z = 300$ $[\text{M}]^+$, 255 $[\text{M} - \text{OC}_2\text{H}_5]^+$, 227 $[\text{M} - \text{OC}_2\text{H}_5, - \text{CO}]^+$, 84 $[\text{CH}_2\text{CONHCHCH}_2]^+$. Anal. Calcd for $\text{C}_{13}\text{H}_{20}\text{N}_2\text{O}_6$: C, 51.99; H, 6.71; N, 9.33. Found: C, 51.96; H, 6.74; N, 9.38.

(S)-Pyroglutamyl-(S)-proline benzyl ester (7). 3 (1.39 g, 5 mmol) and (S)-proline benzyl ester (1.44 g, 7.5 mmol) were stirred for 30 min in dry ether (25 mL). Yield 0.89 g (56%), mp 76 °C (ether), $[\alpha]_{\text{D}} = -131.5^\circ$ ($c = 2$, H_2O). IR (KBr) ν 3470, 1735, 1675, 1645, 1440 cm^{-1} . ^1H NMR (CDCl_3) δ 1.97 - 2.38 (8H, m), 3.54 - 3.59 (2H, m), 4.39 (1H, dd, $J = 8.6$ Hz, $J = 5.0$ Hz), 4.58 (1H, dd, $J = 8.4$ Hz, $J = 3.7$ Hz), 5.07 (1H, d, $J = 12.3$ Hz), 5.20 (1H, d, $J = 12.3$ Hz), 7.01 (1H, br s, NH), 7.24 - 7.38 (5H, m). ^{13}C NMR (CDCl_3) δ 24.4, 24.9, 28.6, 29.2, 46.2, 54.8, 59.2, 67.0, 128.2, 128.5, 128.6, 135.5, 170.7, 171.7, 179.0. MS $m/z = 316$ $[\text{M}]^+$, 209 $[\text{M} - \text{OCH}_2\text{C}_6\text{H}_5]^+$, 208 $[\text{M} - \text{HOCH}_2\text{C}_6\text{H}_5]^+$, 180 $[\text{M} - \text{COOCH}_2\text{C}_6\text{H}_5]^+$, 108 $[\text{C}_6\text{H}_5\text{CH}_2\text{OH}]^+$, 107 $[\text{C}_6\text{H}_5\text{CH}_2\text{O}]^+$, 91 $[\text{C}_7\text{H}_7]^+$, 84 $[\text{CH}_2\text{CONHCHCH}_2]^+$, 77 $[\text{C}_6\text{H}_5]^+$. Anal. Calcd for $\text{C}_{17}\text{H}_{20}\text{N}_2\text{O}_4 \times \text{H}_2\text{O}$: C, 61.06; H, 6.63; N, 8.38. Found: C, 60.76; H, 6.75; N, 8.17.

(S)-Pyroglutamylsarcosine methyl ester (8). A solution of **3** (1.52 g, 5.5 mmol) in acetonitrile (40 mL) was treated for 24 h with sarcosine methyl ester (0.68 g, 6.6 mmol). Compound **(8)** was purified by flash chromatography (eluent: methanol/ether, 1:2). Yield 0.99 g (84%), colorless oil; mixture of rotamers, ratio 5:1, $[\alpha]_{\text{D}} = -21.8^\circ$ ($c = 1.1$, CHCl_3). IR (KBr) ν 3475 - 3075, 2950, 1745, 1650 cm^{-1} . ^1H NMR (CDCl_3) δ 1.96 - 2.02 (1H, m), 2.14 - 2.21 (2H, m), 2.29 - 2.34 (1H, m), 2.94 / 2.80 (3H, s), 3.55 / 3.61 (3H, s), 3.76 (1H, d, $J = 17.2$ Hz), 4.15 (1H, d, $J = 17.2$ Hz), 4.46 (1H, dd, $J = 8.0$ Hz, $J = 4.8$ Hz), 6.99 / 6.96 (1H, s, NH). ^{13}C NMR (CDCl_3) δ 24.3 / 24.6, 29.0 / 29.2, 35.6 / 35.0, 49.4 / 50.3, 51.9 / 52.4, 53.5 / 53.2, 169.3, 172.3, 178.3. MS $m/z = 214$ $[\text{M}]^+$, 182 $[\text{M} - \text{CH}_3\text{OH}]^+$, 142 $[\text{C}_6\text{H}_{10}\text{N}_2\text{O}_2]^+$, 84 $[\text{C}_4\text{H}_6\text{NO}]^+$, 69 $[\text{C}_4\text{H}_5\text{O}]^+$, 59 $[\text{CO}_2\text{CH}_3]^+$, 44 $[\text{CONH}_2]^+$. Anal. Calcd for $\text{C}_9\text{H}_{14}\text{N}_2\text{O}_4$: C, 50.46; H, 6.59; N, 13.08. Found: C, 50.43; H, 6.65; N, 13.02.

(S)-Pyroglutamylazaglycine methyl ester (9). **3** (2.77 g, 10 mmol) were treated for 3 h with methoxycarbonylhydrazine (0.90 g, 10 mmol) in ethyl acetate (50 mL). Yield 1.47 g (73%), mp 148 - 149 °C (AcOEt), $[\alpha]_{\text{D}} = -12.0^\circ$ ($c = 1.0$, H_2O). IR (KBr) ν 3500 - 3100, 1760, 1730 - 1650, 1525, 1255 cm^{-1} . ^1H NMR ($\text{DMSO}-d_6$) δ 1.88 (1H, m), 2.04 - 2.33 (3H, m), 3.57 (3H, s), 4.01 (1H, dd, $J = 8.5$ Hz, $J = 3.6$ Hz), 7.86 (1H, s, NH), 9.11 (1H, s, NH), 9.83 (1H, s, NH). ^{13}C NMR ($\text{DMSO}-d_6$) δ 25.3, 29.1, 52.0, 54.2, 156.6, 172.4, 177.5. MS $m/z = 201$ $[\text{M}]^+$, 173 $[\text{M} - \text{CO}]^+$, 84 $[\text{C}_4\text{H}_6\text{NO}]^+$. Anal. Calcd for $\text{C}_7\text{H}_{11}\text{N}_3\text{O}_4$: C, 41.79; H, 5.51; N, 20.89. Found: C, 41.79; H, 5.61; N, 20.90.

4-[2,2-Bis(trifluoromethyl)-5-oxo-1,3-oxazolidin-4-yl]butanoate (11). A vigorously stirred solution of (4*S*)- α -aminoadipic acid (**10**) (10.0 g, 61.7 mmol) in dry DMSO (30 mL) was treated with an excess of hexafluoroacetone. Procedure see under compound (**2**). Yield 16.0 g (90%), mp 60-62 °C (CH₂Cl₂), [α]_D = -6.0° (c = 2.1, CHCl₃). IR (KBr) ν 3385 br, 1815, 1705 cm⁻¹. ¹H NMR (acetone-d₆) δ 1.71 - 2.02 (4H, m), 2.43 (2H, t, *J* = 7.0 Hz), 4.26 (1H, m), 5.29 (1H, d, *J* = 6.5 Hz, NH). ¹³C NMR (acetone-d₆) δ 22.1, 33.9, 34.1, 55.5, 90.0 (sept., *J* = 34.0 Hz), 122.2 (q, *J* = 284.0 Hz), 123.2 (q, *J* = 288.0 Hz), 173.1, 175.3. ¹⁹F NMR (acetone-d₆) δ -2.91 (3F, q, *J* = 9.0 Hz), -2.00 (3F, q, *J* = 9.0 Hz). MS *m/z* = 309 [M]⁺, 291 [M - H₂O]⁺, 263 [M - H₂O, -CO]⁺, 235 [M - H₂O, -CH₃CHCO]⁺, 166 [(CF₃)₂CO]⁺. Anal. Calcd for C₉H₉NO₄F₆: C, 34.96; H, 2.93; N, 4.53. Found: C, 34.90; H, 2.99; N, 4.56.

(6*S*)-1-Aza-9,9-bis(trifluoromethyl)-8-oxabicyclo[4.3.0]nonane-2,7-dione (12). **11** (3.09 g, 10 mmol) was stirred with an excess of thionyl chloride (5.95 g, 50 mmol) due to protocol applied for compound (**3**). Yield 2.05 g (70%), mp 51-52 °C (CHCl₃/hexane), [α]_D = +4.0° (c = 1.4, CH₂Cl₂). IR (KBr) ν 1855, 1710 cm⁻¹. ¹H NMR (CDCl₃) δ 1.68 - 1.80 (1H, m), 1.88 - 2.04 (1H, m), 2.06 - 2.18 (1H, m), 2.41 - 2.50 (1H, m), 2.53 - 2.62 (2H, m), 4.25 (1H, dd, *J* = 11.5 Hz, *J* = 4.0 Hz). ¹³C NMR (CDCl₃) δ 19.5, 25.2, 31.7, 55.4, 90.3 (sept., *J* = 33.0 Hz), 119.5 (q, *J* = 289.0 Hz), 120.6 (q, *J* = 290.0 Hz), 166.7, 167.0. ¹⁹F NMR (CDCl₃) δ 0.05 (3F, m), 5.99 (3F, m). MS *m/z* = 291 [M]⁺, 222 [M - CF₃]⁺, 194 [M - CF₃, -CO]⁺, 55 [C₄H₇]⁺. Anal. Calcd for C₉H₇NO₃F₆: C, 37.13, H, 2.40; N, 4.81. Found: C, 37.19; H, 2.74; N, 4.71.

(*S*)-6-Oxopipicolyl(*S*)-phenylalanine benzyl ester (13). A solution of **12** (1.46 g, 5.0 mmol) in dry acetonitrile (40 mL) was treated with (*S*)-phenylalanine benzyl ester (1.53 g, 6.0 mmol). Compound (**13**) was purified by column chromatography (eluent: methanol/ether, 1:2). Yield: 1.32 g (69%), colorless oil; [α]_D = -4.5° (c = 1.1, DMSO). IR (Film) ν 3450 - 3060, 1740, 1680, 1655 cm⁻¹. ¹H NMR (CDCl₃) δ 1.45 - 1.49 (2H, m), 1.73 - 1.87 (2H, m), 2.18 - 2.24 (2H, m), 3.03 (1H, dd, *J* = 14.0 Hz, *J* = 9.5 Hz), 3.21 (1H, dd, *J* = 14.0 Hz, *J* = 5.5 Hz), 3.91 (1H, m), 4.90 - 4.94 (1H, m), 5.11 (2H, s), 7.00 - 7.32 (11H, m), 7.88 (1H, d, *J* = 8.5 Hz, NH). ¹³C NMR (CDCl₃) δ 18.9, 26.3, 31.8, 37.5, 54.5, 55.4, 66.9, 127.0, 127.1, 128.5, 128.6, 129.2, 129.3, 136.0, 136.5, 171.2, 171.8, 172.6. MS (FAB) *m/z* = 381 [M + H]⁺, 289 [M - C₇H₇]⁺. Anal. Calcd for C₂₂H₂₄N₂O₄: C, 69.46; H, 6.36; N, 7.36. Found: C, 69.41; H, 6.45; N, 7.39.

(*S*)-6-Oxopipicolyl(*S*)-proline methyl ester (14). **12** (1.46 g, 5.0 mmol) and (*S*)-proline methyl ester (0.77 g, 6.0 mmol) were reacted for 2 d in acetonitrile (40 mL). Compound (**14**) was purified by column chromatography (eluent: methanol/ether, 1:2). Yield: 0.83 g (65%), colorless oil; [α]_D = -120.9° (c = 1.1, CHCl₃). IR (Film) ν 3490-3305, 1745, 1680, 1630, 1440 cm⁻¹. ¹H NMR (CDCl₃) δ 1.71-1.80 (2H, m),

1.81-1.95 (3H, m), 1.98-2.02 (3H, m), 2.28-2.32 (2H, m), 3.53-3.60 / 3.45-3.52 (2H, m), 3.66 / 3.74 (3H, s), 4.25 (1H, m), 4.50 / 4.42 (1H, dd, $J = 5.0$ Hz, $J = 8.5$ Hz), 6.76 / 6.62 (1H, s). ^{13}C NMR (CDCl_3) δ 18.8 / 19.1, 24.8 / 25.0, 25.4 / 25.1, 28.9 / 29.0, 31.1 / 31.1, 46.9 / 47.0, 52.5 / 52.5, 54.0 / 54.3, 59.4 / 59.6, 170.3 / 170.2, 172.7 / 172.4, 172.8 / 172.8. MS $m/z = 254$ $[\text{M}]^+$, 222 $[\text{M} - \text{CH}_3\text{OH}]^+$, 156 $[\text{M} - \text{C}_5\text{H}_8\text{NO}]^+$, 128 $[\text{M} - \text{C}_5\text{H}_8\text{NO}, - \text{CO}]^+$, 113 $[\text{M} - \text{C}_5\text{H}_8\text{NO}, - \text{CO}, - \text{CH}_3]^+$, 98 $[\text{C}_5\text{H}_8\text{NO}]^+$, 70 $[\text{C}_4\text{H}_8\text{N}]^+$, 59 $[\text{CO}_2\text{CH}_3]^+$, 43 $[\text{CONH}]^+$. Anal. Calcd for $\text{C}_{12}\text{H}_{18}\text{N}_2\text{O}_4$: C, 56.68; H, 7.13; N, 11.02. Found: C, 56.73; H, 6.98; N, 11.16.

(S)-6-Oxopipercolylsarcosine methyl ester (15). A solution of **12** (1.46 g, 5.0 mmol) in dry acetonitrile (40 mL) was treated for 24 h with sarcosine methyl ester (0.84 g, 6.0 mmol). Compound (**15**) was purified by flash chromatography (eluent: methanol/ether, 1:2). Yield: 0.86 g (75%), colorless oil; mixture of rotamers, ratio 4:1; $[\alpha]_{\text{D}} = 3.0^\circ$ ($c = 1.0$, CHCl_3). IR (Film) ν 3370-3250, 2955, 1750, 1670 cm^{-1} . ^1H NMR (CDCl_3) δ 1.68-1.98 (4H, m), 2.25 (2H, m), 3.00 / 2.86 (3H, s), 3.62 / 3.68 (3H, s), 3.81 (1H, d, $J = 17.5$ Hz), 4.21 (1H, d, $J = 17.0$ Hz), 4.39 (1H, m), 6.51 (1H, br s). ^{13}C NMR (CDCl_3) δ 18.7, 24.8, 30.8, 36.2, 49.9, 52.0, 52.4, 169.3, 171.7, 172.1. MS $m/z = 228$ $[\text{M}]^+$, 197 $[\text{M} - \text{OCH}_3]^+$, 169 $[\text{M} - \text{OCH}_3, - \text{CO}]^+$, 130 $[\text{M} - \text{C}_5\text{H}_8\text{NO}]^+$, 98 $[\text{C}_5\text{H}_8\text{NO}]^+$. Anal. Calcd for $\text{C}_{10}\text{H}_{16}\text{N}_2\text{O}_4$: C, 52.62; H, 7.07; N, 12.27. Found: C, 52.91; H, 7.13; N, 11.98.

N'-Benzyloxycarbonyl-(S)-6-oxopipercolylhydrazide (16). **12** (1.46 g, 5.0 mmol) and *N*-benzyloxycarbonylhydrazine (1.25 g, 7.5 mmol) were stirred for 6 h in dry ether (30 mL). Yield: 0.99 g (68 %), mp 182 °C (ether), white crystals; $[\alpha]_{\text{D}} = 9.3^\circ$ ($c = 1.0$, MeOH). IR (KBr) ν 3530-3350, 3330-3120, 1745, 1680, 1635 cm^{-1} . ^1H NMR ($\text{DMSO}-d_6$) δ 1.60 - 1.79 (4H, m), 2.07 - 2.09 (2H, m), 3.88 (1H, m), 5.05 (2H, s), 7.34 (5H, m), 7.50 (1H, s, NH), 9.23 (1H, s, NH), 9.80 (1H, s, NH). ^{13}C NMR ($\text{DMSO}-d_6$) δ 17.9, 25.8, 31.1, 53.2, 65.9, 127.8, 128.0, 128.4, 136.6, 156.0, 170.5, 171.8. MS $m/z = 291$ $[\text{M}]^+$, 263 $[\text{M} - \text{CO}]^+$, 247 $[\text{M} - \text{CHNO}]^+$, 184 $[\text{M} - \text{OCH}_2\text{Ph}]^+$, 107 $[\text{OCH}_2\text{Ph}]^+$, 98 $[\text{C}_5\text{H}_8\text{NO}]^+$, 91 $[\text{CH}_2\text{Ph}]^+$, 77 $[\text{C}_6\text{H}_5]^+$. Anal. Calcd for $\text{C}_{14}\text{H}_{17}\text{N}_3\text{O}_4$: C, 57.72; H, 5.88; N, 14.42. Found: C, 58.00; H, 5.75; N, 14.10.

ACKNOWLEDGEMENTS

We are grateful to Stiftung Volkswagenwerk, Hannover, for financial support.

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