Syntheses and $^1$H NMR Spectroscopic Investigations of Some Pyrrolidine Carboxylic Acids Designed as Potential Glial GABA Uptake Inhibitors

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The syntheses of 3-pyrroline-3-carboxylic acid (5), cis-4-hydroxyazepropylidene-3-carboxylic acid (6), 3-hydroxyazepropylidene-3-carboxylic acid (11), pyrrolidine-3-acetic acid (homo-β-proline) (15) and cis-4-aminoazepropylidene-3-carboxylic acid (20) are described. Catalytic hydrogenation of appropriate cyclic β-oxoesters are key steps in the preparation of 5 and 6. Compound 11 was synthesized from the ketone 8 via the corresponding protected cyanohydrin 9, and homo-β-proline (15) was prepared via a Knoevenagel reaction. The β-amino acid 20 was prepared by stepwise hydrogenation of the enamine 16 followed by acid treatment of the protected product 19. 270 MHz $^1$H NMR spectroscopic analyses of 4a, 7 and 19 were carried out in order to establish the relative stereochemistry of 6 and 20.

The glial uptake system probably contributes to the termination of GABA mediated synaptic transmission in the central nervous system. 1–3 β-Alanine 1,4 and in particular β-proline 3,5,6 are inhibitors of the glial GABA uptake system. In an attempt to develop more potent and more selective glial GABA uptake inhibitors we have synthesized a number of cyclic amino acids structurally related to β-proline, the structure of which combines the structural elements of GABA as well as of β-alanine.

Most of these compounds, i.e. 5, 6, 11 and 20 have little effect on glial as well as on neuronal GABA transport in vitro. 5, 6, 10, 11, 12–8 Pyrrolidine-3-acetic acid (homo-β-proline) (15), on the other hand, turned out to be a very potent competitive inhibitor of both GABA transport systems. The compound, however, is not a specific GABA uptake inhibitor but has also a high affinity for the GABA receptors. 7

The compounds were prepared as described below. Dieckmann condensation of 2a with methyl acrylate gave the β-oxoester 3a, which according to the $^1$H NMR spectrum exists as the enol-form (Scheme 1). High pressure hydrogenation of 3a, b proceeded stereospecifically to give racemic 4a, b. Treatment of 4a, b with hydrochloric acid gave the hydroxy amino acid 6, whereas prolonged treatment of 4a, b with hydrobromic acid gave the α,β-unsaturated amino acid 5. The ethyl ester 7 was

\[
\begin{align*}
\text{COOH} & \quad \text{NH}_2 \\
\text{GABA} & \quad \beta-\text{Proline} & \quad \beta-\text{Alanine}
\end{align*}
\]

\[
\begin{align*}
\text{COOH} & \quad \text{HO} \\
\text{5} & \quad \text{COOH} \\
(3RS,4RS) & \quad (RS)
\end{align*}
\]

\[
\begin{align*}
\text{COOH} & \quad \text{H}_2\text{N} \\
\text{11} & \quad \text{COOH}
\end{align*}
\]

\[
\begin{align*}
\text{COOH} & \quad \text{H}_2\text{N} \\
\text{15} & \quad \text{COOH}
\end{align*}
\]

\[
\begin{align*}
\text{COOH} & \quad \text{H}_2\text{N} \\
\text{20} & \quad \text{COOH}
\end{align*}
\]
synthesized to facilitate the 270 MHz $^1$H NMR spectroscopic analysis.

Reaction of the ketone 8 with potassium cyanide in glacial acetic acid under acylating conditions gave 9.

In order to prepare compound 13, cyanoacetic acid was used as an active methylene compound in a Knoevenagel condensation with 8. As the reaction was performed in pyridine using piperidine as a catalyst the obtained product was the decarboxylated $\beta,\gamma$-unsaturated nitrile derivative 12, which by low pressure hydrogenation was converted into the saturated compound 13.

The nitriles 9 and 13 were transformed into the amino acids 11 and 15, respectively, via the corresponding methyl esters 10 and 14.

The $\beta$-amino acid 20 was prepared from 3b via the enamine 16 (Scheme 2). High pressure hydrogenation of 16 using Pt – C as a catalyst resulted in reduction of the enamine double bond and yielded 17. Hydrogenolysis of 17 as a hydrochloride gave a compound considered to be 18. Crude 18 was transformed into the protected compound 19, which was used in the 270 MHz $^1$H NMR spectroscopic analysis. Treatment of 19 with hydrobromic acid gave compound 20.

The structure elucidation of the new compounds 2a – 4a, 4b, 5 – 7, 9 – 17, 19 and 20 was based on elemental analyses, IR and $^1$H NMR spectroscopy, in the cases of 12 and 16 supported by UV spectroscopy.

The relative configurations of 4a, 7 and 19 were
deduced by analysis of the 270 MHz $^1$H NMR spectra.

In the literature comparatively little is reported concerning chemical shifts and coupling constants of pyrrolidines$^{10-14}$ The scarcity of data reflects the lack of success NMR data have had in characterizing conformations of this ring system. The pyrrolidine ring is an almost planar pentagon with low barrier to pseudorotation. The NMR data represent accordingly the average over a number of probable conformations. The vicinal coupling constants are claimed to be represented by the Karplus equation$^{15}$ but the extensive averaging results in typical values in the range of 4 to 8 Hz for both cis and trans isomers. The key problem in connection with the 3,4-disubstituted compounds is the determination of the relative configuration of the substituents. The observed values of $J_{3,4}$ of the three compounds 4a, 7 and 19 are 4.59, 5.0 and 6.0 Hz, respectively, permitting no final conclusion as to the cis-trans isomerism. The same consideration is reached based on the values of $J_{2x,3}$ and $J_{2y,3}$. The sum $J_{2x,3}+J_{2y,3}$ is expected to be less influenced by the pseudorotation and is quoted to be 13.3 Hz in pyrrolidine$^{16}$. In all of the three compounds the sum $J_{2x,3}+J_{2y,3}+J_{4,5x}+J_{4,5y}$ is close to twice this value, but in 4a and 7 $J_{2x,3}+J_{2y,3}$ has increased to 18 Hz, while the contribution from $J_{4,5x}+J_{4,5y}$ has fallen to 6 Hz. This indicates that 4a as well as 7 are present in solution with strongly biased conformations.

Since no reliable conclusion regarding the cis–trans isomerism can be drawn from the NMR data above, we have attempted to gain this from proton–proton nuclear Overhauser effect (NOE)$^{17}$ On the qualitative level the experiment may be used to distinguish protons on basis of their spatial separation. The NOE depends on $r^{-6}$, $r$ being the interproton distance, and this ensures that only protons with a distance of $r \leq 6$ Å will normally be capable of showing measurable effects.$^{17}$ For the NOE experiment $H_4$ is chosen for saturation, since it is well separated from the remaining signals of interest. The NOE difference spectrum and the reference spectrum of 19 are reproduced in Fig. 1. It is seen that a NOE of 12% is found for $H_3$ in 19. In a similar experiment concerning compound 4a a NOE of 12% is found for $H_3$. These effects are sufficiently large$^{17}$ to ensure that the protons $H_3$ and $H_4$ of 4a and 19 are located in a cis configuration.

The chemical processes, which transform these compounds to the final products 6 and 20, will retain the configuration at $C_3$ and $C_4$, leading to the conclusion, that 6 and 20 are also characterized by cis arrangement of the substituents at $C_3$ and $C_4$.

**EXPERIMENTAL**

Thin-layer chromatography (TLC) and column chromatography (CC) were accomplished by using silica gel GF254 plates (Merck) and silica gel

Fig. 1. In trace a the $^1$H NMR spectrum of 19 is reproduced, obtained in FT mode at 270 MHz. Trace b shows the proton—proton difference nuclear Overhauser effect obtained by irradiating $H_4$. The appearance of a positive NOE of 12% is determined for $H_3$. 

(Woelm 0.063 – 0.100 mm), respectively. Columns were developed by stepwise gradient elution. Melting points, determined in capillary tubes, are corrected. Elemental analyses were made by Mr. P. Hansen, Chemical Laboratory II, University of Copenhagen. The $pK_a$-values were determined as described in a previous paper. A Perkin-Elmer grating infrared spectrophotometer model 247, a Perkin-Elmer ultraviolet-visible spectrophotometer model 402 and a JEOL JMN-C-60HL (60 MHz) $^1$H NMR instrument were used. The 270 MHz $^1$H NMR spectra were obtained on a Bruker HX 270 S instrument operating at 293 and 353 K.

The samples were contained in 5 mm o.d. sample tubes. The concentrations of the substances were ca. 4 w/v %. The analysis of the spectra was supplemented with selective decoupling experiments wherever appropriate. $^1$H NMR spectra were recorded using TMS as an internal standard, except for the compounds dissolved in $D_2$O, where DSS was used.

The spectra of 4a, 7 and 19 have been simulated using the programme MIMER and the simulations were found to coincide with the experimental spectra.

The proton—proton nuclear Overhauser experiments were performed by difference technique. Two spectra are alternatingly accumulated and swapped from disc. In the active spectrum $C_4$–$H$ was saturated using a 4 s pulse, after which the FID was sampled with the decoupler off. The passive spectrum is a reference spectrum where the irradiation is displaced outside the spectral region. The NOE spectrum corresponds to the difference between the two spectra accumulated. The magnitude of the NOE was determined by computer subtraction of a suitable fraction of the reference spectrum whereby nulling was obtained for the signal for which the NOE was to be determined. In the NOE difference spectrum a broad signal due to traces of water was observed at 2.91 ppm. The delay of 4 s between the experiments did not permit full relaxation of water. As a consequence the outbalancing was not complete.

**Methyl N-Methoxycarbonylglycinate (2a).** To a stirred ice-cooled solution of 1a (50.2 g; 0.4 mol) in water (150 ml) was added an iced solution of potassium carbonate (138.2 g; 1.0 mol) in water (150 ml) followed by methyl chloroformate (45.4 g; 0.48 mol). The mixture was stirred at 0 °C for 1 h and at 24 °C for 1 h followed by extraction with ether (4 x 300 ml). The combined and dried (K$_2$CO$_3$) ether phases were evaporated in vacuo. Distillation of the residue gave 2a (27.3 g; 46 %), collected at 82 – 84 °C/50 Pa. Anal. C$_3$H$_9$NO$_4$: C, H, N. IR (film): 3370 (m), 2930 (w), 1750 (s), 1720 (s), 1530 (m), 1280 (m), 1210 (s) cm$^{-1}$. $^1$H NMR (60 MHz, CDCl$_3$): $\delta$ 5.66 (1 H, broad signal), 3.93 (2 H, m), 3.77 (3 H, s), 3.70 (3 H, s).

**Methyl 1-methoxycarbonyl-4-oxopyrroolidine-3-carboxylate (3a).** To a suspension of sodium (3.3 g; 0.145 g-atom) in CaH$_2$-dried toluene – xylene [250
cis-Methyl 1-methoxycarbonyl-4-hydroxypyrrrolidine-3-carboxylate (4a). A solution of 3a (7.2 g; 36 mmol) in methanol (400 ml) was hydrogenated (ca. 5 MPa) for 24 h using ca. 3 g PtO₂-W₂ catalyst. The filtered and evaporated reaction mixture gave 4a (7.2 g; 90%) as a crude product. An analytical sample was purified by CC [silica gel; eluents: toluene containing ethyl acetate (78-84%)] followed by ball-tube distillation at 50 Pa (oven temperature 200 °C). Found: C 46.75; H 6.93; N 6.77. Calc. for C₇H₁₂NO₃: C 47.29; H 6.45; N 6.89.

1H NMR (270 MHz, DMSO-d₆, 353 K): δ₁ = 3.50, δ₂ = 3.58, δ₃ = 3.16, δ₄ = 4.41, δ₅ = 3.31, δ₆ = 3.42, δₓ = 5.2, δᵧ = 3.63, δₒ = 3.59, J₁₂₋₃₋₄ = 10.92 Hz, J₂₋₃ = 8.43 Hz, J₃₋₄ = 9.63 Hz, J₄₋₅ = 4.59 Hz, J₅₋₆ = 1.74 Hz, J₆₋₇ = 4.43 Hz, J₅₋₆ = 11.55 Hz.

cis-Ethyl 1-methoxycarbonyl-4-hydroxypyrrrolidine-3-carboxylate (4b). A solution of 3b (2.0 g; 9.3 mmol) in ethanol (200 ml) was hydrogenated (ca. 3.5 MPa) for 24 h using ca. 1 g PtO₂-W₂ catalyst. Ball-tube distillation of the evaporated reaction mixture at 50 Pa (oven temperature 210 °C) gave 4b (1.8 g; 89%). Found: C 48.35; H 6.69; N 6.49. Calc. for C₉H₁₅NO₃: C 49.76; H 6.96; N 6.45. IR (film): 3430 (m), 2980-2870 (several bands, w), 1730 (s), 1690 (s), 1460 (s), 1395 (m), 1200 (m) cm⁻¹. 1H NMR (60 MHz, CDCl₃): δ = 5.00 (1H, broad signal), 4.49 (1H, m), 4.13 (2H, q), 3.8-3.4 (m) and 3.65 (a total of 7 H), 3.23 (1H, m), 1.27 (3 H, t).

3-Carboxy-3-pyrrolidinyl bromide (5). A mixture of 4a (2.0 g; 10 mmol) or 4b (2.2 g; 10 mmol) and 48% aqueous hydrobromic acid (10 ml) was refluxed for 24 h. Filtration of the hot reaction mixture, followed by cooling gave pure 5 (605 mg; 31%) m.p. 255-257 °C (decomp.). Calc. for C₅H₄BrNO; C, 37.56; H, 2.87; N, 4.84. Anal. for C₅H₄BrNO; C, 37.56; H, 2.87; N, 4.84. 1H NMR (60 MHz, DMSO-d₆, D₂O [9:1]): δ = 6.75 (1H, m), 4.4-4.1 (4H, m). pKa values (H₂O, 25 °C): 2.93 ± 0.01; 9.77 ± 0.03.

cis-3-Carboxy-4-hydroxy-3-pyrrolidinyl chloride (6). A solution of 4a (900 mg; 4.5 mmol) or 4b (1.0 g; 4.6 mmol) in 5 M hydrochloric acid (10 ml) was refluxed for 1.5 h. Evaporation of the reaction mixture to dryness in vacuo and recrystallization (acetic acid) of the residue gave 6 (200 mg; 26%). m.p. ca. 159 °C (decomp.). Anal. C₅H₄BrNO; C, 37.56; H, 2.87; N, 4.84. 1H NMR [60 MHz, DMSO-d₆, D₂O (9:1)]: δ = 5.34 (1H, q), 4.12 (4H, s), 3.3 (1H, m). pKa values (H₂O, 25 °C): 3.39 ± 0.01; 10.06 ± 0.02.

cis-3-Ethoxycarbonyl-4-hydroxy-3-pyrrolidinyl chloride (7). A solution of 6 (100 mg; 0.60 mmol) in 9 w/v% ethanol hydrochloric acid (2 ml) was refluxed for 2 h. Evaporation in vacuo and recrystallization (ethanol—ether) gave 7 (40 mg; 34%), m.p. 190-191 °C. Anal. C₅H₄BrNO; C, 37.56; H, 2.87; N, 4.84. 1H NMR (270 MHz, DMSO-d₆, 353 K): δ₁ = 3.25, δ₂ = 3.39, δ₃ = 3.16, δ₄ = 4.50, δ₅ = 3.04, δₓ = 3.19, δₒ = 4.11 and 4.13, δᵧ = 1.22. J₁₂₋₃₋₄ = 11.2 Hz, J₂₋₃ = 8.3 Hz, J₃₋₄ = 9.9 Hz, J₄₋₅ = 5.0 Hz, J₅₋₆ = 1.8 Hz, J₆₋₇ = 4.0 Hz, J₇₋₈ = 12.0 Hz, J₈₋₉ = 7.1 Hz, J₉₋₁₀ = 10.78 Hz.

1-Methoxycarbonyl-3-acetoxy-3-cyanopyrrolidine (9). A solution of 8 (20 mg; 21 mmol) and potassium cyanide (2.1 g; 32 mmol) in glacial acetic acid (12 ml) was stirred at room temperature for 1 h. After addition of acetic anhydride (2.7 g; 26 mmol) the mixture was heated at 50-60 °C for 70 h. The evaporated reaction mixture was dissolved in water (50 ml) and extracted with ether (4 × 25 ml). The combined, dried and evaporated ether phases were subjected to CC [silica gel; eluent: toluene—ethyl acetate—methanol (80:20:2)] to give 9 (3.4 g; 76%), m.p. 74-75 °C. Anal. C₆H₁₀N₂O₄; C, 44.25; H, 6.45; N, 16.67; O, 32.98. 1H NMR (60 MHz, CDCl₃): δ = 3.9-3.4 (m) and 3.67 (a total of 7 H), 2.50 (2 H, t), 2.12 (3 H, s).

Methyl 1-methoxycarbonyl-3-hydroxy-3-pyrrolidinyl-3-carboxylate (10). A solution of 9 (2.0 g; 9.4 mmol) in 10 w/v% methanolic hydrochloric acid (40 ml) was stirred at 24 °C overnight. The reaction mixture was concentrated in vacuo to ca. 15 ml. Upon addition of water (30 ml) and stirring at room temperature for 1/4 h the solvents were removed in vacuo and the residue was extracted with ethyl acetate—toluene (4:1) (3 × 15 ml). The combined and dried (MgSO₄) organic extract was evaporated to give 10 (1.5 g; 79%). An analytical sample was purified by CC [silica gel; eluents: toluene containing ethyl acetate (80-84%)] followed by ball-tube distillation at 25 Pa (oven temperature 170 °C). Anal. C₁₂H₁₄NO₃. Found: C 45.70, H 7.00, N 6.47. Calc.:
C 47.29, H 6.45, N 6.89. IR (film): 3400 (m), 2960 (m), 2890 (w), 1730 (s), 1690 (s), 1460 (s), 1400 (m), 1230 (s) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 5.06 (1 H, broad signal), 4.0–3.5 (m), 3.88 (s), and 3.77 (s) (a total of 10 H), 2.5–2.0 (2 H, m).

3-Carboxy-3-hydroxypropyridinium chloride (11). A solution of 10 (300 mg; 1.5 mmol) in 5 M hydrochloric acid (6 ml) was refluxed for 1 ½ h. The reaction mixture was evaporated to dryness in vacuo. The crystalline residue was recrystallized (water–acetic acid–ether) to give I1 (100 mg; 40 %), m.p. 224–225 °C (decomp.). Anal. C₈H₁₀ClNO₃: C, H, Cl, N, IR (KBr): 3460–2730 (several bands, s–m), 1730 (s), 1575 (w), 1400 (m), 1210 (m) cm⁻¹. ¹H NMR [60 MHz, DMSO-d₆–D₂O (9:1)]: δ 3.5–3.0 (4 H, m), 2.4–1.9 (2 H, m). pKₐ values (H₂O, 25 °C): 2.82 ± 0.03; 10.11 ± 0.03.

1-Methoxy carbonyl-3-cyanomethyl-2-pyrrole (12). To a solution of 8²⁰ (10.0 g; 70 mmol) and cyanacetic acid (12.0 g; 140 mmol) in pyridine (100 ml) was added piperidine (2 ml). The mixture was refluxed for 12 h. The solvent was removed in vacuo and CC [silica gel: 500 g; eluents: toluene containing ethyl acetate (20–30 %) and formic acid (1 %)] of the residue gave 12 (7.2 g; 62 %). An analytical sample was purified by ball-tube distillation at 65 Pa (oven temperature 150 °C). Found: C 57.00; H 6.21; N 16.62. Calc. for C₈H₁₆N₂O₂: C 57.82; H 6.07; N 16.86. IR (film): 3120 (w), 2970–2850 (several bands, m–w), 2250 (w), 1710 (s), 1660 (m), 1460 (s), 1410 (s), 1210 (m) cm⁻¹. UV [methanol (log ε)]: 234 (4.07) nm. ¹H NMR (60 MHz, CDCl₃): δ 6.63 (1 H, m), 3.87 (2 H, m), 3.77 (3 H, s), 3.17 (2 H, m), 2.65 (2 H, m).

1-Methoxy carbonyl-3-cyanomethyl-pyrroline (13). A solution of 12 (3.0 g; 18 mmol) in ethanol (125 ml) was hydrogenated (ca. 300 kPa) for 21 h in a PARR low pressure hydrogenation apparatus using a 10 % Pd – C catalyst (0.6 g). Ball-tube distillation of the evaporated solution at 130 Pa (oven temperature 170 °C) gave 13 (2.7 g; 89 %). Anal. C₈H₁₆N₂O₂: C, H, N, IR (film): 2970 (m), 2880 (m), 2250 (w), 1700 (s), 1640 (m), 1390 (s), 1200 (m) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 3.8–2.9 (m) and 3.72 (s) (a total of 7 H), 2.6–2.3 (3 H, m), 2.3–1.6 (2 H, m).

Methyl 1-methoxy carbonyl pyridinium chloride-3-acetate (14). A solution of 13 (2.0 g; 12 mmol) in 20 w/v % methanolic hydrochloric acid (50 ml) was stirred at room temperature overnight. The reaction mixture was concentrated in vacuo at 30 °C to ca. 10 ml. Upon addition of water (35 ml) the mixture was extracted continuously for 2 h in a Kutscher-Steuvel apparatus with ether – methylene chloride (4:1) (200 ml). The dried (Na₂SO₄) and evaporated organic phase was submitted to CC [silica gel: 100 g; eluents: toluene containing ethyl acetate (50–54 %)] to give 14 (1.2 g; 50 %). An analytical sample was purified by ball-tube distillation at 50 Pa (oven temperature 175 °C). Found: C 53.10; H 7.68; N 6.78. Calc. for C₈H₁₇N₂O₄: C 53.72; H 7.51; N 6.96. IR (film): 2950 (m), 2870 (w), 1730 (s), 1695 (s), 1450 (s), 1390 (s), 1190 (m), 1170 (m) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 3.8–2.7 (m) and 3.66 (s) (a total of 10 H), 2.6–2.3 (3 H, m), 2.3–1.5 (2 H, m).

Pyrridine-3-acetic acid (homo-β-proline) hydrochloride (15). A solution of 14 (500 mg; 2.5 mmol) in 5 M hydrochloric acid (10 ml) was refluxed for 1 ½ h. Evaporation to dryness in vacuo and recrystallization (water–acetic acid–ether) gave 15 (280 mg; 68 %), m.p. 90–91.5 °C. Anal. C₈H₁₃ClNO₂: C, H, N, IR (KBr): 3460–2700 (several bands, s–m), 1720 (s), 1590 (w), 1410 (m), 1210 (m) cm⁻¹. ¹H NMR (60 MHz, D₂O): δ 3.8–3.0 (4 H, m), 3.0–2.4 (m) and 2.64 (d) (a total of 3 H), 2.4–1.5 (2 H, m). pKₐ values (H₂O, 25 °C): 3.96 ± 0.01; 11.03 ± 0.05.

Ethyl 1-methoxycarbonyl-4-benzylamino-3-pyrrole-3-carboxylate (16). To a stirred solution of 3b²⁰ (5.0 g; 23.2 mmol) in toluene (75 ml) was added benzylamine (2.7 g; 24.8 mmol) and 5 g of Molecular Sieve (Union Carbide 3 A). The mixture was refluxed for 20 h using a Dean–Stark water separator. Upon evaporation in vacuo the reaction product was purified by CC [silica gel: 100 g; eluents: methylene chloride containing ethyl acetate (50–65 %)] to give 16 (6.0 g; 85 %). IR (film): 3340 (m), 3100–2850 (several bands, m–w), 1700 (s), 1670 (s), 1620 (s), 1450 (s), 1390 (s), 1290 (s) cm⁻¹. UV [methanol (log ε)]: 288 (4.11) nm. ¹H NMR (60 MHz, CDCl₃): δ 7.25 (5 H, s), 4.4–3.8 (8 H, m), 3.63 (3 H, s), 1.22 (3 H, t).

cis-Ethyl 1-methoxycarbonyl-4-benzylaminopyrrole-3-carboxylate (17). A solution of 16 (4.8 g; 15.8 mmol) in ethanol (350 ml) was hydrogenated (ca. 9 MPa) using a 5 % Pt – C catalyst (2.5 g) for 72 h. Filtration and evaporation in vacuo followed by CC [silica gel: 150 g; eluents: methylene chloride containing ethyl acetate (45–70 %)] yielded starting material (1.8 g; 37 %) and 17 (1.1 g; 23 %). Anal. C₁₅H₁₆ClNO₄: Found: C 61.75, H 7.23, N 9.00. Calc.: C 62.72, H 7.24, N 9.14. IR (film): 3350 (w), 3100–2850 (several bands, m–w), 1730 (s), 1700 (s), 1450 (s), 1390 (s), 1190 (m) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 7.17 (5 H, s), 4.05 (2 H, q), 3.8–3.4 (m), and 3.58 (s) (a total of 9 H), 3.4–2.7 (3 H, m), 1.18 (3 H, t).

cis-Ethyl 1-methoxycarbonyl-4-methoxycarbonylaminopyrrole-3-carboxylate (19). A solution of 17 (800 mg; 2.6 mmol) and 0.1 M aqueous hydrochloric acid (26 ml) in 50 % aqueous ethanol (120 ml) was hydrogenated (ca. 300 kPa) for 20 h in a PARR low pressure hydrogenation apparatus using a 5 % Pd – C catalyst (250 mg). The reaction mixture was concentrated in vacuo to ca. 30 ml and washed with methylene chloride (2 × 20 ml). The aqueous
phase was evaporated in vacuo to give an oil (0.7 g), characterized by TLC \([R_f; 0.33];\) eluent: butanol—acetic acid—water (4:1:1). The compound was considered to be 18. To a stirred ice-cooled solution of the crude product (0.7 g) and potassium carbonate (900 mg; 6.5 mmol) in water (9 ml) was added methyl chloroformate (300 mg; 3.1 mmol). After stirring for further 1 h at 0 °C and 1 h at room temperature the reaction mixture was extracted with ether (6 \(\times\) 10 ml). Evaporation of the combined and dried \((\text{Na}_2\text{SO}_4)\) ether phases gave 19 (50 mg; 7% based on 17), which was shown to be TLC-pure in a variety of eluent-systems. An analytical sample was purified by CC [silica gel; eluent: toluene containing ethyl acetate (78–82%)] followed by ball-tube distillation at 65 Pa (oven temperature 250 °C). Anal. C\(_{11}\)H\(_{18}\)N\(_2\)O\(_6\); C, H, N. IR (film): 3320 (m), 2990–2850 (several bands, m–w), 1730 (s), 1710 (s), 1690 (s), 1550 (m), 1460 (s), 1400 (s), 1200 (s) cm\(^{-1}\). \(^1\)H NMR (270 MHz, DMSO-\(d_6\), 353 K): \(\delta_{25} = 3.53, \delta_2 = 3.68, \delta_3 = 3.25, \delta_4 = 4.43, \delta_5 = 3.35, \delta_6 = 3.58, \delta_{\text{NH}} = 6.83, \delta_{\text{CH}_2} = 4.09\) and 4.13, \(\delta_{\text{NH} = \text{COOCH}_3} = 3.63, \delta_{\text{COOCH}_3} = 3.59, \delta_{\text{CH}_2} = 1.22, J_{J_2, J_7} = 10.8\) Hz, \(J_{J_3, J_8} = 7.8\) Hz, \(J_{J_4, J_5} = 7.05\) Hz, \(J_{J_5, J_6} = 6.0\) Hz, \(J_{\text{NH} = \text{CH}_2} = 8.1\) Hz, \(J_{J_4, J_5} = 5.15\) Hz, \(J_{J_5, J_6} = 6.45\) Hz, \(J_{J_5, J_7} = 11.03\) Hz, \(J_{J_4, J_5} = 7.1\) Hz, \(J_{J_3, J_8} = 11.22\) Hz.

cis-3-Carboxy-4-aminopyrrolidinum dibromide (20). A mixture of 19 (500 g; 1.8 mmol) and aqueous hydrobromic acid (6 ml) was refluxed for 2 h. Evaporation to dryness in vacuo gave crude 20 (500 mg) as an oil, which slowly crystallized. Recrystallization from water–acetic acid–ether [45 ml; (2:5:2)] gave pure 20 (175 mg; 33%), m.p. 178–181 °C. Anal. C\(_{13}\)H\(_{12}\)Br\(_2\)N\(_2\)O\(_2\); C, H, Br, N. IR (KBr): 3440 (m), 3200–2600 (several bands, s–m), 1735 (s), 1590 (m), 1490 (m), 1390 (m) cm\(^{-1}\).

\(^1\)H NMR (60 MHz, DMSO-\(d_6\)–\(D_2\)O (9:1)): \(\delta\) 4.15 (1 H, m), 3.8–3.3 (5 H, m).

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