

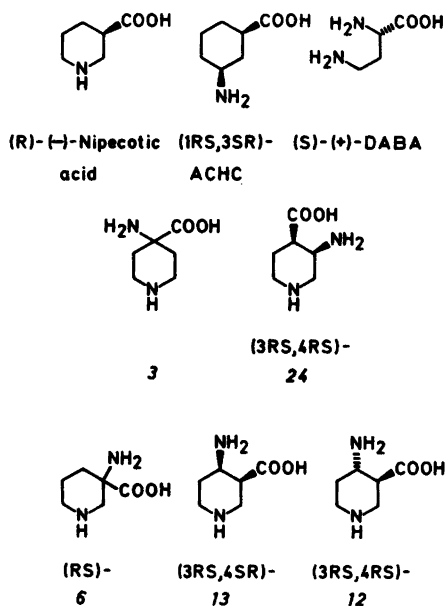
Potential GABA Uptake Inhibitors. Synthesis and Relative Stereochemistry of Some Aminopiperidinecarboxylic Acids

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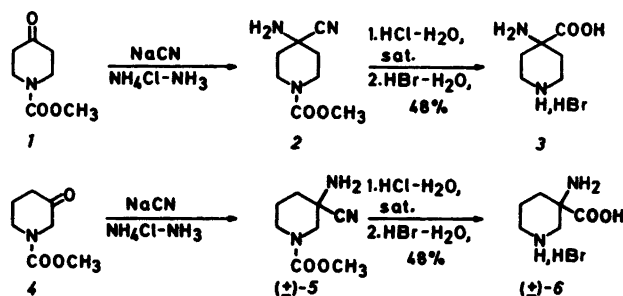
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The syntheses of 4-aminopiperidine-4-carboxylic acid (3), (*RS*)-3-aminopiperidine-3-carboxylic acid (6), (*3RS,4RS*)- and (*3RS,4SR*)-4-aminopiperidine-3-carboxylic acid (12) and (13) and (*3RS,4RS*)-3-aminopiperidine-4-carboxylic acid (24) are described. The compounds 3 and 6 were prepared from the corresponding *N*-protected 3- and 4-piperidones by Strecker reactions. The β -amino acids 12 and 13 were prepared *via* addition of benzylamine to the 1,2,5,6-tetrahydropyridine-3-carboxylic acid derivative 7. The *cis*-isomer 13 was also prepared *via* stepwise hydrogenation of 17, the 4-benzylamino derivative of 7. An analogous reaction sequence was used for the synthesis of the *cis*- β -amino acid 24, isomeric with 13. The relative stereochemistry of 12, 13, and 24 was established by 270 MHz ¹H NMR spectroscopy.

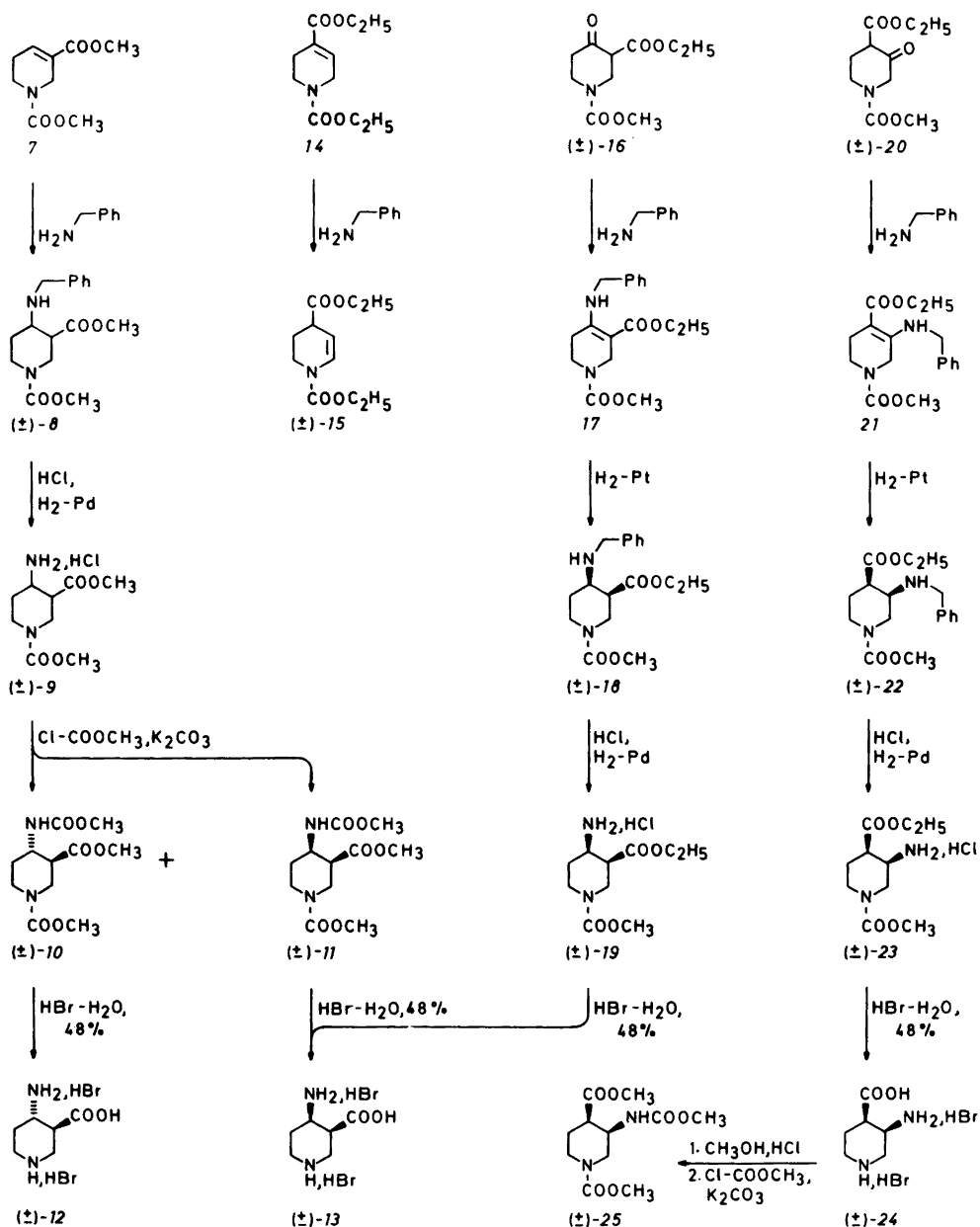
(*R*)-(-)-Nipecotic acid,^{1–3} (*S*)-(+)-2,4-diaminobutyric acid (DABA)^{4,5} and (1*RS,3SR*)-3-aminocyclohexanecarboxylic acid (ACHC)^{6,7} (Scheme 1) are substrate competitive inhibitors of the neuronal GABA uptake system. Of these inhibitors DABA and ACHC have selective effects on the neuronal



Scheme 1.



Scheme 2.



Scheme 3.

GABA uptake system, whereas nipecotic acid, which is the most potent inhibitor, also interacts with the transport process for GABA in glial cells.⁷ In an attempt to develop potent and selective neuronal GABA uptake inhibitors, we have now synthesized

a series of cyclic amino acids in which the structural characteristics of the above-mentioned inhibitors have been combined. Surprisingly, however, these compounds, *i.e.* 3, 6, 12, 13, and 24 (Scheme 1), have very little or no effect on neuronal GABA transport

in vitro,⁸ emphasizing the pronounced substrate specificity of this system.⁹

The α -amino acids 3 and 6 were prepared *via* Strecker reactions (Scheme 2). The β -amino acids 12 and 13 were prepared by addition of benzylamine to the 1,2,5,6-tetrahydropyridine-3-carboxylic acid derivative 7 and subsequent hydrogenolysis of the mixture of diastereomeric racemates 8 (Scheme 3). Treatment of 9 with methyl chloroformate gave a separable mixture of the diastereomeric compounds 10 and 11, which by acid treatment gave the amino acid dihydrobromides 12 and 13. Treatment of the 1,2,3,6-tetrahydropyridine-4-carboxylic acid derivative 14 with benzylamine in ethanol or toluene, however, caused migration of the double bond, and compound 15 was isolated as the only product (Scheme 3).

Condensation of the β -oxo-esters 16 and 20 with benzylamine gave the enamines 17 and 21, respectively. The high pressure hydrogenation of the enamine double bonds of 17 and 21 under neutral conditions proceeded stereospecifically and did not result in debenzylation. The benzyl groups of 18 and 22 were removed by low pressure hydrogenolysis in the presence of equivalent amounts of 0.1 N HCl, and subsequent acid treatment converted 19 and 23 into the dihydrobromides 13 and 24 of the desired amino acids. The protected compound 25 was prepared as part of the 270 MHz ¹H NMR spectroscopic analysis of 24.

The structures of the new products 2, 3, 5, 6, 8–13, 15, 17–19, and 21–25 were confirmed by elemental analysis, IR and ¹H NMR spectroscopy, in the cases of 15, 17, and 21 supported by UV spectroscopy. The relative configurations of 10–13, 24, and 25 were established by analysis of the 270 MHz ¹H NMR spectra.

In simple piperidine derivatives the equatorial proton on C(2) is found downfield from its axial counterpart.¹⁰ The coupling constants between the C(3) proton and the two C(2) protons in 24 are typical for axial-equatorial and axial-axial configurations of these protons. This is consistent with a predominantly equatorial orientation of the C(3) amino group (Table 1). Furthermore, the coupling constant for the C(3) and C(4) protons unequivocally indicates axial-equatorial orientation of the protons concerned and therefore a 3,4-*cis* configuration of 24. An analysis producing the coupling constants for the C(4) and C(5) protons supports this assignment. The coupling patterns of the C(2) and C(3) protons in the protected amino acid 25 indicates a

predominantly axial orientation of the methoxycarbonylamino group in position 3. Analysis of the mutual coupling constants for the C(4) and C(5) protons indicates an axial orientation of the proton at C(4), which is consistent with a 3,4-*cis* configuration of 25, but with the opposite orientation of the two substituents as compared with the unprotected compound 24.

In the spectrum of 12 a very complex pattern of signals was observed and it was not possible to establish the orientation of the substituents in this compound. However, the relative configuration of 12 was established from the coupling patterns in the ¹H NMR spectrum of the corresponding protected amino acid 10. The coupling constants between the C(3) proton and the two C(2) protons indicate an equatorial orientation of the C(3) methoxycarbonyl group. The coupling constant between the C(3) and C(4) protons in 10 is typical for axial-axial configuration of the protons. Therefore compound 10 has 3,4-*trans* configuration, which is further established by the axial-axial coupling constant between the C(4) and one of the C(5) protons.

The coupling patterns for the C(2), C(3), C(4), and C(5) protons in 13 are very similar to those observed in the ¹H NMR spectrum of 25 indicating axial and equatorial positions of the substituents at C(3) and C(4), respectively, and therefore a 3,4-*cis* configuration of 13. The spectrum of the protected amino acid 11 is complex and only the orientation of the C(3) substituent could be established. The coupling patterns for the C(2) and C(3) protons indicate an axial orientation of the C(3) methoxycarbonyl group, and therefore 11 and 13 must have the same orientation of the substituents.

The geminal coupling constants found in the present ¹H NMR spectroscopic analysis parallel data previously found in other piperidines.^{12,13} The interpretation of the vicinal coupling constants in terms of ring conformation follows the general treatment of 6-membered rings.¹¹ The observed values of the axial-axial coupling constants indicate the extent of conformational fixation of the substituents. The compounds 10 and 24 have almost identical axial-axial coupling constants, $J_{2a3a} = 11.0$ Hz and 10.8 Hz, respectively. The values are consistent with a dominating contribution from the equatorial-equatorial orientation in 10 and the equatorial-axial orientation in 24 of the C(3) and C(4) substituents. The J_{4a5a} value found in 13 (12.0 Hz) is higher than that in 10, indicating an increased fixation in an axial-equatorial arrangement of the

Table 1. Chemical shifts (δ) and coupling constants (Hz) from the 270 MHz ^1H NMR spectra of 10–13, 24 and 25.^a

	(±)-24	(±)-25	(±)-12	(±)-10	(±)-13	(±)-11
δ_{2a}	3.545	3.37		3.04	3.36	3.208
δ_{2e}	3.651	3.79	3.96	4.11	3.96	
δ_{3a}	3.97	—		2.43	—	—
δ_{3e}	—	4.11	—	—	—	2.853
δ_{4a}	—	2.789		3.86	3.8	
δ_{4e}	3.398	—	—	—	—	—
δ_{5a}	2.168	1.94	2.02	1.42	2.133	1.99
δ_{5e}	2.460	1.83	2.43	2.05	2.281	1.72
δ_{6a}	3.15	3.24		2.91	3.26	3.033
δ_{6e}	3.435	3.65	3.62	4.26	3.56	
J_{2a2e}	–12.6	–13.2		–13.6	–13.5	
J_{2a3a}	10.8	—		11.0	—	—
J_{2a3e}	—	3.0	—	—	3.5	3.5
J_{2e3a}	4.35	—	3.0	4.2	—	—
J_{2e3e}	—	6.0	—	—	4.0	3.0
J_{3a4a}	—	—		11.0	—	—
J_{3a4e}	4.35	—	—	—	—	—
J_{3e4a}	—	4.2	—	—	2.0	4.02
J_{3e4e}	—	—	—	—	—	—
J_{4a5a}	—	8.4		10.2	12.0	—
J_{4a5e}	—	4.2		—	3.0	—
J_{4e5a}	4.2	—	—	—	—	—
J_{4e5e}	4.35	—	—	—	—	—
J_{5a5e}	–15.6	–13.8		–12.5	–14.0	–13.0
J_{5a6a}	12.0	8.4		13.0	12.5	10.8
J_{5a6e}	4.3	5.4		4.5	4.5	
J_{5e6a}	3.3	4.2		2.5	3.5	3.0
J_{5e6e}	3.8	6.0		—	2.3	—
J_{6a6e}	–13.0	–13.2		–13.0	–13.2	–13.7

^a — indicates that the δ value or the coupling constant concerned does not exist. The missing values could not be deduced from the available data.

C(3) and C(4) substituents. The spectrum of 25 is quite different. The lower value of J_{4a5a} (8.4 Hz) points to a conformational equilibrium, where axial-equatorial orientations of the C(3) and C(4) substituents represent the preferred conformation. As mentioned above, the ^1H NMR spectra of 11 and 12 are very complex, and therefore the preferred conformations of these compounds cannot be deduced from the available data.

EXPERIMENTAL

Melting points, determined in capillary tubes, are corrected. Elemental analyses were made by Mr. P. Hansen, Chemical Laboratory II, University of Copenhagen. A Perkin-Elmer grating infrared spectrophotometer model 247, a Perkin-Elmer ultraviolet-visible spectrophotometer model 402 and a Jeol JMN-C-60HL (60 MHz) ^1H NMR instrument were used. ^1H NMR spectra were re-

corded using TMS as an internal standard. Compounds dissolved in D₂O were referenced to TSP. The 270 MHz ¹H NMR spectra were obtained on a Bruker HX 270 S instrument operating at 293 and 303 K. The Fourier transform method was used to obtain the spectra with a spectral width of 1500 Hz using 32 K data points, using quadrature detection.

Homodecoupling was used to verify the interpretations of the spectra and provide starting parameters for the analyses of the spectra. Nitrogen decoupling using a Bruker probehead equipped with an additional decoupling coil was utilized to remove nitrogen broadening of the signals. The frequency was derived from a Bruker synthesizer model BS 100 and fed to the amplifier of the decoupling channel through a band pass filter before entering the probe. The decoupling power was 5 W. The decoupling frequency used was 19.506 582 MHz. The proton spectra were analyzed using the programme SIMEQ¹⁴ and a Varian 620/i computer. Due to the large chemical shift differences at 270 MHz, it is permissible to divide the total spin system into several subsystems containing typically 3 to 6 nuclei. After the initial assignment the spectra were iterated to a best fit using the ITRCAL¹⁵ programme on a Nicolet 1180 computer with 80 K memory. Thin-layer chromatography (TLC) and column chromatography (CC) were accomplished by using silica gel F₂₅₄ plates (Merck) and silica gel (Woelm 0.063–0.1 mm), respectively. Columns were developed by stepwise gradient elution. The pK_A values were determined as described in a previous paper.¹⁶

1-Methoxycarbonyl-4-cyano-4-aminopiperidine (2). To a stirred mixture of sodium cyanide (2.39 g; 48.8 mmol) in water (4.5 ml), ammonium chloride (2.82 g; 52.7 mmol) in water (6 ml) and aqueous ammonia (6.45 g; ρ=0.88) was added 1¹⁷ (7.5 g; 47.8 mmol). Stirring was continued for 5 h at 60 °C. Upon cooling of the reaction mixture to 24 °C 2 (6.8 g; 77 %) crystallized. An analytical sample was recrystallized (water) to give 2, m.p. 112.0–113.0 °C. Anal. C₈H₁₃N₃O₂: C, H, N. IR (KBr): 3700–3200 (m), 3000–2800 (m), 2220 (w), 1680 (s), 1450 (s), 1400 (s), 1240 (s) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 4.2–2.9 (4 H, m), 3.67 (3 H, s), 2.2–1.3 (6 H, m).

4-Aminopiperidine-4-carboxylic acid hydrobromide (3). A suspension of 2 (2.6 g; 14.2 mmol) in water (34 ml) and aqueous hydrochloric acid (42 ml; 10 M) was saturated with hydrogen chloride at 0 °C. The solution was left for 2 days at 24 °C and then refluxed for 4 h. Evaporation *in vacuo* and recrystallization (water–ethanol) gave 1.85 g of 4-aminopiperidine-4-carboxylic acid as a mixture of mono- and dihydrochloride. To this compound aqueous hydrobromic acid (25 ml; 48 %) was added. Evaporation *in vacuo* and recrystallization (water–ethanol) gave 3 (1.20 g; 63 %), m.p. 255 °C (de-

comp.). Anal. C₆H₁₃N₂O₂Br: C, H, N, Br. IR (KBr): 3450 (w), 3200–2400 (m, several bands), 1600 (s), 1440 (m), 1300 (s) cm⁻¹. ¹H NMR (60 MHz, D₂O): δ 3.9–3.3 (4 H, m), 2.8–1.8 (4 H, m). pK_A values (H₂O, 24 °C): ca. 1.0, 7.47 ± 0.10, 10.96 ± 0.05.

(RS)-1-Methoxycarbonyl-3-cyano-3-aminopiperidine (5). To a stirred mixture of sodium cyanide (0.32 g; 6.5 mmol) in water (1 ml), ammonium chloride (0.38 g; 7.1 mmol) in water (2 ml) and aqueous ammonia (0.86 g; ρ=0.88) was added 4¹⁸ (1.0 g; 6.4 mmol). Stirring was continued for 5 min at 60 °C. The solvent was removed *in vacuo* and the residue was extracted with chloroform (3 × 6 ml). The combined and dried (K₂CO₃) chloroform phases were evaporated *in vacuo* to give crude 5 (1.1 g; 95 %). An analytical sample was recrystallized (benzene–light petroleum) to give 5, m.p. 97.0–98.0 °C. Anal. C₈H₁₃N₃O₂: C, H, N. IR (KBr): 3700–3200 (m, several bands), 2210 (w), 1690 (s), 1480 (s), 1410 (m) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 4.5–3.9 (2 H, m), 3.83 (3 H, s), 3.5–2.8 (2 H, m), 2.4–1.6 (6 H, m).

(RS)-3-Aminopiperidine-3-carboxylic acid hydrobromide (6). A suspension of 5 (1.2 g; 6.37 mmol) in water (16 ml) and aqueous hydrochloric acid (20 ml; 10 M) was saturated with hydrogen chloride at 0 °C. The solution was left for 2 days at 24 °C and then refluxed for 10 h. Evaporation *in vacuo* and recrystallization (water–acetic acid) gave 0.72 g of *(RS)-3-aminopiperidine-3-carboxylic acid* as a mixture of mono- and dihydrochloride. To this compound aqueous hydrobromic acid (25 ml; 48 %) was added. Evaporation *in vacuo* and recrystallization (water–acetic acid) gave 6 (0.6 g; 80 %), m.p. 260 °C (decomp.). Anal. C₆H₁₃N₂O₂Br: C, H, N, Br. IR (KBr): 3450 (w), 3200–2300 (m, several bands), 1640 (s), 1500 (s), 1360 (s) cm⁻¹. ¹H NMR (60 MHz, D₂O): δ 4.0–2.8 (4 H, m), 2.4–1.7 (4 H, m). pK_A values (H₂O, 24 °C): ca. 1.4, 6.59 ± 0.10, 9.86 ± 0.07.

(3RS,4RS)- and (3RS,4SR)-Methyl 1-methoxycarbonyl-4-benzylaminopiperidine-3-carboxylate (8). To a solution of 7¹⁹ (4.0 g; 20.1 mmol) in methanol (31 ml) was added benzylamine (2.4 g; 22.4 mmol). The mixture was refluxed for 4 days. CC [silica gel: 100 g; eluents: methylene chloride containing ethyl acetate (60–75 %)] of the evaporated reaction product gave starting material (1.9 g; 47.5 %) and 8 (2.1 g; 34.2 %), which by TLC [eluent: methylene chloride–ethyl acetate (2:3)] gave only one spot (R_F: 0.21). Anal. C₁₆H₂₂N₂O₄: C, H, N. IR (film): 3700–3300 (w), 3100–2800 (m, several bands), 1740 (s), 1700 (s), 1480 (m), 1460 (s) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 7.37 (5 H, s), 4.4–3.4 (5 H, m), 3.75 (3 H, s), 3.70 (3 H, s), 3.2–2.4 (5 H, m), 1.68 (1 H, s).

(3RS,4RS)-Methyl N,N-dimethoxycarbonyl-4-

aminopiperidine-3-carboxylate (10) and (3RS,4SR)-*methyl N,N-dimethoxycarbonyl-4-aminopiperidine-3-carboxylate* (11). A solution of 8 (2.1 g; 6.86 mmol) in aqueous hydrochloric acid (68.6 ml; 0.1 M) and aqueous ethanol (150 ml; 68 %) was hydrogenated (ca. 300 kPa) in a PARR hydrogenation apparatus by using a 10 % Pd-C catalyst (0.2 g) for 20 h. Evaporation *in vacuo* gave crude 9, which by TLC [eluent: 1-butanol-acetic acid-water (4:1:1)] gave only one spot (R_F : 0.29). To an ice-cooled solution of crude 9 in water (11 ml) was added with stirring an iced solution of potassium carbonate (2.8 g; 20.3 mmol) in water (3.8 ml) followed by addition of methyl chloroformate (3.0 g; 31.7 mmol). Stirring was continued at 0 °C for 1 h and at 24 °C for 1 h. The mixture was extracted with ether (4 × 30 ml). The combined and dried (Na₂SO₄) ether phases were evaporated *in vacuo* to give an oil, which by TLC [eluent: toluene-methanol (19:1)] was shown to consist of two compounds (R_F : 0.31 and 0.23). CC [silica gel: 40 g; eluents: toluene containing methanol (5-7 %)] gave 10 (0.82 g; 43.6 %) and 11 (0.61 g; 33.0 %). An analytical sample of 10 was recrystallized (benzene-cyclohexane), m.p. 103.0-104.0 °C. Anal. C₁₁H₁₈N₂O₆: C, H, N. IR (KBr): 3600-3300 (m), 3100-2800 (w, several bands), 1740 (s), 1700 (s), 1540 (m), 1460 (m), 1310 (s), 1240 (s) cm⁻¹.

An analytical sample of 11 was purified by ball-tube distillation at 100 Pa (oven temperature 250 °C). Anal. C₁₁H₁₈N₂O₆: Found: C 49.30; H 7.01; N 9.90. Calc. C 48.17; H 6.62; N 10.21. IR (film): 3700-3200 (m), 3100-2800 (m, several bands), 1740-1660 (s, several bands), 1530 (m), 1440 (m), 1240 (m) cm⁻¹.

(3RS,4RS)-4-Aminopiperidine-3-carboxylic acid dihydrobromide (12). A solution of 10 (0.3 g; 1.1 mmol) in aqueous hydrobromic acid (15 ml; 48 %) was refluxed for 2 h. Evaporation of the reaction mixture *in vacuo* and recrystallization (ethanol) gave 12 (0.14 g; 42 %) as a very hygroscopic substance, m.p. 220 °C (decomp.). Anal. C₆H₁₄N₂O₂Br₂. Found: C 25.45; H 5.56; N 8.21; Br 46.30. Calc. C 23.55; H 4.62; N 9.16; Br 52.22. IR (KBr): 3400 (w), 3200-2700 (m, several bands), 1720 (s), 1600 (s), 1460 (m), 1390 (s) cm⁻¹. pK_A values (H₂O, 24 °C): 2.82 ± 0.05, 8.24 ± 0.05, 10.77 ± 0.08.

(RS)-Ethyl 1-ethoxycarbonyl-1,2,3,4-tetrahydropyridine-4-carboxylate (15). To a solution of 14²⁰ (0.5 g; 2.2 mmol) in methanol (4 ml) was added benzylamine (0.26 g; 2.4 mmol). The mixture was refluxed for 4 days. CC [silica gel: 25 g; eluents: toluene containing ethyl acetate (10-20 %)] of the evaporated reaction product gave 15 (0.4 g; 80 %). Anal. C₁₁H₁₇NO₄: C, H, N. IR (film): 3450 (w), 3000 (s), 1720 (s), 1650 (m), 1410 (s) cm⁻¹. UV [methanol (log ϵ): 222 (4.08) nm. ¹H NMR (60

MHz, CDCl₃): δ 6.87 (1 H, d), 4.87 (1 H, m), 4.10 (4 H, q), 3.60 (2 H, t), 3.05 (1 H, m), 2.0 (2 H, m), 1.43 (6 H, t).

Ethyl 1-methoxycarbonyl-4-benzylamino-1,2,5,6-tetrahydropyridine-3-carboxylate (17). To a solution of 16²¹ (10.0 g; 43.7 mmol) in toluene (150 ml) was added benzylamine (5.0 g; 46.7 mmol) and Molecular Sieve, Union Carbide 3A (10 g). The mixture was refluxed for 20 h using a Dean-Stark water separator. Evaporation *in vacuo* followed by CC [silica gel: 200 g; eluents: methylene chloride containing ethyl acetate (60-80 %)] gave 17 (9.8 g; 71 %). IR (film): 3300 (w), 3100-2800 (m, several bands), 1700 (s), 1660 (s), 1600 (s), 1450 (s), 1250 (s) cm⁻¹. UV [methanol (log ϵ): 295 (4.21) nm. ¹H NMR (60 MHz, CDCl₃): δ 9.21 (1 H, broad signal), 7.26 (5 H, s), 4.36 (2 H, d), 4.1 (4 H, m), 3.71 (3 H, s), 3.51 (2 H, t), 2.36 (2 H, t), 1.32 (3 H, t).

(3RS,4SR)-Ethyl 1-methoxycarbonyl-4-benzylaminopiperidine-3-carboxylate (18). A solution of 17 (4.0 g; 12.6 mmol) in ethanol (300 ml) was hydrogenated (ca. 6 MPa) by using a 5 % Pt-C catalyst (2 g) for 72 h. Evaporation *in vacuo* followed by CC [silica gel: 125 g; eluents: methylene chloride containing ethyl acetate (60-90 %)] gave 18 (3.1 g; 78 %). An analytical sample was purified by ball-tube distillation at 50 Pa (oven temperature 250 °C). Anal. C₁₇H₂₄N₂O₄: C, H, N. IR (film): 3700-3300 (w), 3100-2800 (m, several bands), 1730 (s), 1700 (s), 1450 (s), 1240 (s), 1190 (s) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 7.25 (5 H, s), 4.11 (2 H, q), 3.79 (2 H, d), 3.67 (3 H, s), 3.6-3.2 (4 H, m), 3.2-3.0 (1 H, m), 2.8-2.6 (1 H, m), 2.38 (1 H, s), 2.0-1.6 (2 H, m), 1.22 (3 H, t).

(3RS,4SR)-4-Aminopiperidine-3-carboxylic acid dihydrobromide (13). a. A solution of 18 (0.7 g; 2.19 mmol) and aqueous hydrochloric acid (21.9 ml; 0.1 M) in aqueous ethanol (100 ml; 50 %) was hydrogenated (ca. 300 kPa) in a PARR hydrogenation apparatus by using a 5 % Pd-C catalyst (0.2 g) for 24 h. Evaporation *in vacuo* gave crude 19, characterized by TLC [R_F : 0.42; eluent: 1-butanol-acetic acid-water (4:1:1)]. A solution of crude 19 in aqueous hydrobromic acid (20 ml; 48 %) was refluxed for 2 h. Evaporation *in vacuo* and recrystallization (acetic acid-water) gave 13 (0.4 g; 60 % based on 18), m.p. 240 °C (decomp.). Anal. C₆H₁₄N₂O₂Br₂: C, H, N, Br. IR (KBr): 3450 (w), 3150-2600 (m, several bands), 1710 (s), 1570 (m), 1470 (s), 1380 (s), 1210 (s) cm⁻¹. pK_A values (H₂O, 24 °C): ca. 1.9, 7.82 ± 0.01, 10.34 ± 0.05.

b. A solution of 11 (0.3 g; 1.1 mmol) in aqueous hydrobromic acid (15 ml; 48 %) was refluxed for 2 h. Evaporation *in vacuo* and recrystallization (acetic acid-water) gave 13 (0.17 g; 52 %), the IR spectrum of which was identical with that of 13 prepared from 19.

Ethyl 1-methoxycarbonyl-3-benzylamino-1,2,5,6-

tetrahydropyridine-4-carboxylate (21). Compound 21 was synthesized as described above for 17 by using 20²² (10.0 g; 43.7 mmol). A yield of 12.1 g (87 %) of 21 was obtained. IR (film): 3300 (w), 3100–2800 (m, several bands), 1700 (s), 1660 (s), 1600 (s), 1450 (s), 1250 (s) cm⁻¹. UV [methanol (log ϵ): 295 (4.18) nm]. ¹H NMR (60 MHz, CDCl₃): δ 7.33 (5 H, s), 4.5–4.0 (6 H, m), 3.67 (3 H, s), 3.47 (2 H, t), 2.35 (2 H, m), 1.30 (3 H, t).

(3RS,4RS)-Ethyl 1-methoxycarbonyl-3-benzylaminopiperidine-4-carboxylate (22). Compound 22 was synthesized as described above for 18 by using 21 (5.0 g; 15.7 mmol). A yield of 3.2 g (63 %) of 22 was obtained. Anal. C₁₇H₂₄N₂O₄: C, H, N. IR (film): 3700–3300 (w), 3100–2800 (m, several bands), 1720 (s), 1700 (s), 1460 (s), 1200 (s) cm⁻¹. ¹H NMR (60 MHz, CDCl₃): δ 7.30 (5 H, s), 4.15 (2 H, q), 3.80 (2 H, d), 3.69 (3 H, s), 3.6–3.3 (2 H, m), 3.2–2.4 (4 H, m), 2.05 (1 H, s), 2.0–1.6 (2 H, m), 1.25 (3 H, t).

(3RS,4RS)-3-Aminopiperidine-4-carboxylic acid dihydrobromide (24). A solution of 22 (3.0 g; 9.4 mmol) in aqueous hydrochloric acid (94 ml; 0.1 M) and aqueous ethanol (300 ml; 50 %) was hydrogenated (ca. 300 kPa) in a PARR hydrogenation apparatus by using a 5 % Pd–C catalyst (0.8 g) for 24 h. Evaporation *in vacuo* gave crude 23, characterized by TLC [R_F : 0.37; eluent: 1-butanol–acetic acid–water (4:1:1)]. A solution of crude 23 in aqueous hydrobromic acid (60 ml; 48 %) was refluxed for 2 h. Evaporation *in vacuo* and recrystallization (acetic acid–water) gave 24 (1.64 g; 57 % based on 22), m.p. 235 °C (decomp.). Anal. C₆H₁₄N₂O₂Br₂: C, H, N, Br. IR (KBr): 3400 (w), 3200–2500 (m, several bands), 1720 (s), 1570 (m), 1460 (m), 1200 (s) cm⁻¹. pK_A values (H₂O, 24 °C): 2.54 ± 0.06, 6.96 ± 0.01, 10.12 ± 0.01.

(3RS,4RS)-Methyl N,N-dimethoxycarbonyl-3-aminopiperidine-4-carboxylate (25). A solution of 24 (0.7 g; 2.3 mmol) in methanolic hydrogen chloride (30 ml; 5 %) was refluxed for 24 h. The reaction mixture was evaporated *in vacuo*. To an ice-cooled solution of the residue in water (5 ml) was added with stirring an iced solution of potassium carbonate (1.4 g; 10.1 mmol) in water (1.8 ml) followed by addition of methyl chloroformate (3.0 g; 31.7 mmol). Stirring was continued at 0 °C for 1 h and at 24 °C for 1 h. The mixture was extracted with ether (4 × 20 ml). The combined and dried (Na₂SO₄) ether phases were evaporated *in vacuo*. CC [silica gel: 25 g; eluents: toluene containing methanol (5–7 %)] gave 25 (0.49 g; 79 %). Anal. C₁₁H₁₈N₂O₆: C, H, N. IR (film): 3700–3200 (m), 3100–2800 (m), 1740–1660 (s, several bands), 1540 (m), 1440 (m) cm⁻¹.

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REFERENCES

1. Krogsgaard-Larsen, P. and Johnston, G. A. R. *J. Neurochem.* 25 (1975) 797.
2. Johnston, G. A. R., Krogsgaard-Larsen, P., Stephanson, A. L. and Twitchin, B. *J. Neurochem.* 26 (1976) 1029.
3. Johnston, G. A. R., Stephanson, A. L. and Twitchin, B. *J. Neurochem.* 26 (1976) 83.
4. Martin, D. L. In Roberts, E., Chase, T. N. and Tower, D. B., Eds., *GABA in Nervous System Function*, Raven, New York 1976, p. 347.
5. Iversen, L. L. and Kelly, J. S. *Biochem. Pharmacol.* 24 (1975) 933.
6. Neal, M. J. and Bowery, N. G. *Brain Res.* 138 (1977) 169.
7. Schousboe, A., Krogsgaard-Larsen, P., Svenneby, G. and Hertz, L. *Brain Res.* 153 (1978) 623.
8. Schousboe, A., Thorbek, P., Hertz, L. and Krogsgaard-Larsen, P. *J. Neurochem.* 33 (1979) 181.
9. Krogsgaard-Larsen, P. In Fonnum, F., Ed., *Amino Acids as Chemical Transmitters*, Plenum, New York 1978, p. 305.
10. Lambert, J. B. and Keske, R. G. *J. Am. Chem. Soc.* 88 (1966) 620.
11. Jackman, L. M. and Sternhell, S. *Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry*, Pergamon, Oxford 1969, p. 288.
12. Lambert, J. B., Keske, R. G., Carhart, R. E. and Jovanovitch, A. P. *J. Am. Chem. Soc.* 89 (1967) 3761.
13. Krogsgaard-Larsen, P., Thyssen, K. and Schaumburg, K. *Acta Chem. Scand. B* 32 (1978) 327.
14. Kort, C. W. F., SIMEQ, Spectrometry Dept., Laboratory of Organic Chemistry, University of Amsterdam, Holland.
15. ITRCAL, Nicolet Instrument Corporation 5225 (Copyright), Verona Road, Madison, Wisconsin 53711, USA.
16. Hjeds, H. and Honoré, T. *Acta Chem. Scand. B* 32 (1978) 187.
17. Krogsgaard-Larsen, P. and Roldskov-Christiansen, T. *Eur. J. Med. Chem.* 14 (1979) 157.

18. Krogsgaard-Larsen, P. and Hjeds, H. *Acta Chem. Scand. B* 30 (1976) 884.
19. Christensen, S. B. and Krogsgaard-Larsen, P. *J. Labelled Compd.* 17 (1980) 191.
20. Krogsgaard-Larsen, P. and Johnston, G. A. R. *J. Neurochem.* 30 (1978) 1377.
21. Krogsgaard-Larsen, P. and Hjeds, H. *Acta Chem. Scand. B* 28 (1974) 533.
22. Krogsgaard-Larsen, P. *Acta Chem. Scand. B* 31 (1977) 584.

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