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Two Non-Racemic Preparations of a Piperidine-Based NMDA Antagonist with Analgesic Activity

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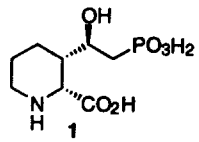
Abstract—(2*R*,3*S*,1′*R*)-3-(1-Hydroxy-2-phosphonoethyl)-2-piperidinecarboxylic acid **1** has been synthesized by two different methods. The NMDA receptor binding affinities (K_i values) are 74 nM for compound **1**, and 64 nM for the corresponding ketone **2**. The analgesic effects were evaluated using the mouse hot-plate test and the mouse formalin model. The ED_{50} values for the racemates of compounds **1** and **2**, using the mouse hotplate and intrathecal injection, were 0.53 and 0.51 nmol, respectively. © 1997 Elsevier Science Ltd.

Introduction

Excitatory amino acids, such as aspartate and glutamate, play an important role in central synaptic transmission. Consequently the modulation of glutamate receptors may be of interest in the treatment of central nervous system disorders. The best characterized of the glutamate receptors are those activated selectively by *N*-methyl-D-aspartate (NMDA). These receptors are involved in a phenomenon known as excitotoxicity, which may accompany stroke or cerebral trauma. Overstimulation of NMDA receptors may also be a factor in several other neurological disorders such as convulsive disorders, neuropathic pain and anxiety.[1] Several selective competitive antagonists of the NMDA receptor have been described[1] and a few years ago we reported the racemates of compounds **1** and **2**[2] as potent competitive NMDA antagonists. The synthesis of compound **2** (also named MDL 100,925) has been reported by us and others.[2,3] We showed that these piperidine compounds adopt a chair conformation, with internal hydrogen bonding that stabilizes the conformation with the 2 and 3 substituents in the *equatorial* and *axial* position, respectively (Fig. 1). This conformation appears to be crucial for activity since the analogous deoxypiperidine has 10-fold lower activity.[4] In this paper we report the synthesis of **1**, the receptor binding affinities of **1** and its antipode **3**, as well as some of the pharmacological properties of the racemates of compounds **1** and **2** as analgesic agents.



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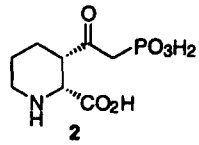

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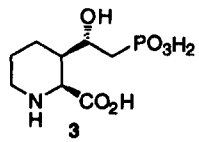

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Chemistry

For toxicological and pharmacological studies, larger quantities of the pure enantiomer **1** were needed, and we developed a synthetic method suitable for pilot plant facilities. The earlier reported pyridinecarboxylic acid chloride **4**[4] (Scheme 1) served as starting material for **1**. Acid chloride **4** was subjected to a magnesium chloride-catalyzed acylation reaction with diethyl malonate in the presence of triethyl amine;[5] subsequent decarboxylation in wet DMSO produced methyl ketone **5**. Ketone **5** was then treated with Br_2 in HBr/AcOH to give bromoketone **6**. Several unsuccessful attempts at enantioselective reduction of bromoketone **6** with (+)-chlorodiisopino-campheylborane[6] were made (yields <5%). However, catalytic reduction of ketone **6** with

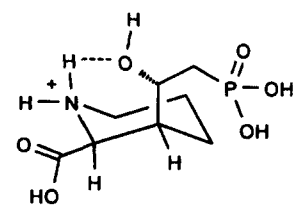

Figure 1.

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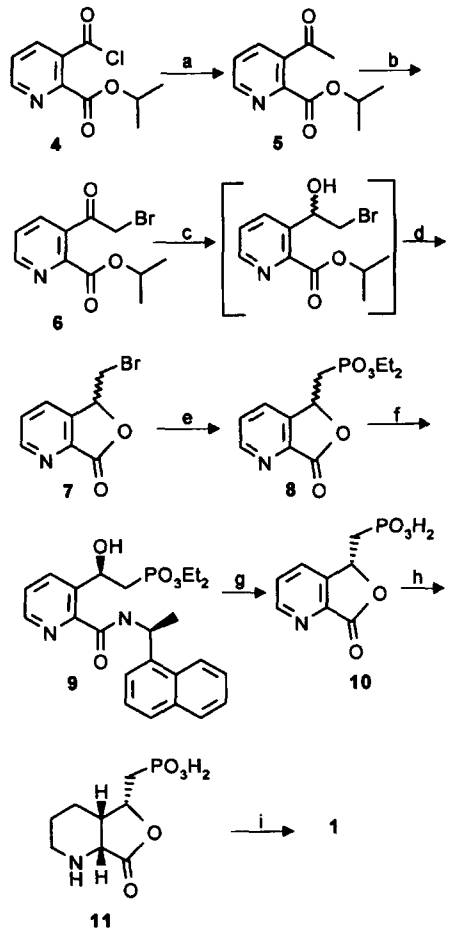
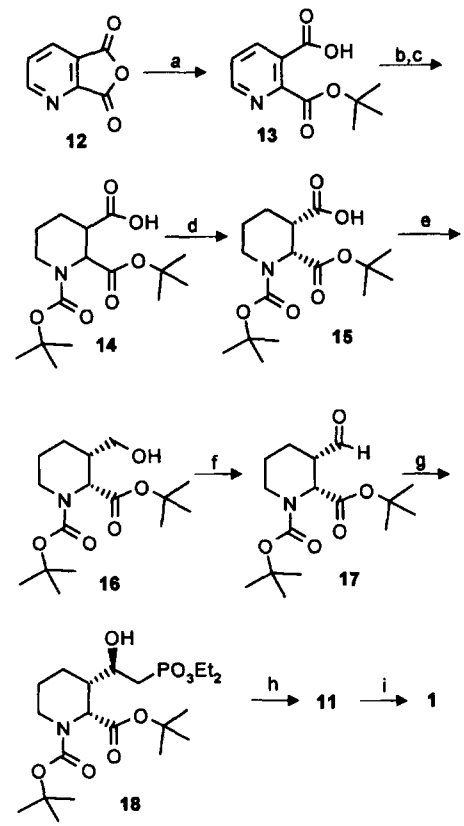
1,3,2-oxazaborolidine catalyst derived from (*S*)-α,α-diphenyl-2-pyrrolidinemethanol,[7] gave good yields (90%, 5% ee). Higher enantiomeric excess (ee) was obtained with (4*R*,5*S*)-*B*-methyl-4,5-diphenyl-1,3,2-oxazaborolidine[8] as catalyst and borane dimethylsulfide as reductant (yield 90%, 50% ee[9]). Although an ee of about 50% might be acceptable, extensive purification would still be required to produce compounds with good enantiomeric purity. We thus chose to pursue a different approach via simple sodium borohydride reduction of ketone **6** to the racemic alcohol and resolution of the diastereomeric mixture later in the synthetic scheme.

Sodium borohydride in acetic acid was found to be a mild and selective reducing agent for the reduction of ketone **6** to racemic alcohol, which was subsequently converted to lactone **7** by heating in toluene/AcOH. An Arbuzov reaction of **7** with triethyl phosphite gave phosphonate **8** in moderate yields; a major side reaction was the elimination of HBr. Treatment of lactone **8** with (*S*)-1-(1-naphthyl)ethylamine afforded amide **9** as a diastereomeric mixture. After two recrystallizations the desired diastereomer **9** was obtained in over 95% de (^{1}H NMR of the well-separated benzylic proton multiplets at 5.50 and 5.62 ppm). Treatment of **9** with 6 M HCl at reflux yielded the lactone **10**.

Attempts to reduce pyridine lactone **10** using catalytic hydrogenation over Pd/C or PtO_2 failed to give piperidine lactone **11** in acceptable yields. These hydrogenations tended to stop at the 2,3-dehydropiperidine stage. However, catalytic hydrogenation over Rh/C was successful and gave the desired piperidine lactone **11**. Hydrogenation of pyridine lactone **10** was expected to occur from the less-hindered side and produce the 2*R*,3*S*,1′*R* lactone **11**. We were able to prove the relative stereochemistry by ^{1}H NMR analysis of the racemic diethylester lactone (Fig. 2).[2] The measured coupling constants in $CDCl_3$ closely match the calculated values for a piperidine ring chair conformer, with the 2 and 3 substituents in *axial* (2*R*) and *equatorial* (3*S*) positions respectively, and with the phosphonomethyl sidechain in the *endo* (1′*R*) position.[10] The absolute configuration of compound **2** had previously been determined to be 2*R*,3*S*,[2,3] and because we used the common intermediate **14** (Scheme 2) for the synthesis of **1** and **2**, the absolute configuration of



Scheme 1. (a) 1. $CH_2(CO_2Et)_2$, $MgCl_2$, Et_3N, 2. DMSO, H_2O, 50%. (b) Br_2, HBr, CH_3COOH, 50%. (c) $NaBH_4$, CH_3COOH, THF. (d) CH_3COOH, toluene, 69%. (e) $P(OEt)_3$, toluene, 25%. (f) (*S*)-1-(1-naphtyl)ethylamine, 43%. (g) 6 M HCl, 72%. (h) H_2, Rh/C, H_2O. (i) NaOH, Dowex 50WX8-100, 1 M NH_3, 1 M NaOH, 63%.



Scheme 2. (a) *t*-BuOH, pyridine, 42%. (b) H_2, Pd/C, CH_3COOH, 96%. (c) di-*tert*-butyl dicarbonate, Et_3N, CH_2Cl_2, MeOH, 66%. (d) (+)-ephedrine, EtOAc, 25%. (e) $(COCl)_2$, toluene, $NaBH_4$, THF, 94%. (f) $(COCl)_2$, CH_2Cl_2, DMSO, 40%. (g) BuLi, $CH_3PO_3Et_2$, THF, 51%. (h) TMSBr, $CHCl_3$, H_2O, 90%. (i) LiOH, Dowex 50WX8-100, 1 M NH_3, 82%.

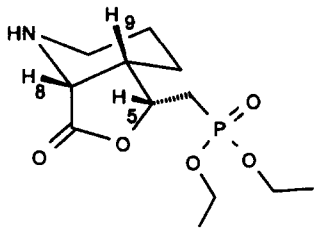


Figure 2.

compound **1** was assigned as 2*R*,3*S*,1′*R*. After hydrolysis of piperidine lactone **11** with sodium hydroxide followed by ion exchange on an ammonium satd ion exchange resin, the product was precipitated as a disodium salt of **1** (LAS 250). Using an analogous synthetic scheme we were also able to produce the 2*S*,3*R*,1′*S* enantiomer **3**.

For autoradiographic and pharmacokinetic studies, radioactively labeled **1** was needed, and we developed a synthetic route that would be suitable for [^{14}C] incorporation (i.e., late introduction of [^{14}C] in the synthetic scheme). This synthesis (Scheme 2) started with commercially available quinolinic acid anhydride **12**, which was converted to *t*-butyl ester **13** with *t*-butyl alcohol in pyridine solution. Hydrogenation of pyridine **13** over Pd/C, followed by Boc-protection of the amino group gave compound **14**. This acid was resolved by resolution via the formation of diastereomeric salts using 0.5 equiv of (+)-ephedrine. After two recrystallizations from ethyl acetate the pure enantiomer **15** was obtained in over 95% ee.[2] Carboxylic acid **15** was treated with oxalyl chloride affording the corresponding acid chloride, which was subsequently reduced with sodium borohydride to alcohol **16**. Swern oxidation of alcohol **16** yielded aldehyde **17** which was susceptible to epimerization during silica gel chromatography, but could be obtained in an almost enantiomerically pure form by rapid flash chromatography. We anticipated that it would be possible to add lithiated diethyl methylphosphonate to the enantiomerically pure aldehyde **17** and that this would produce mainly the desired diastereomer **18**. According to Cram's rule the addition would occur preferentially from the less-hindered *re*-face, thus giving mainly the product with 1′*R* configuration. The addition of lithiated diethyl methylphosphonate to aldehyde **17** in THF at −70 °C afforded a diastereomeric mixture which contained 85% of the diastereomer **18**. Compound **18** was treated with an excess of TMSBr in $CHCl_3$, followed by addition of water to produce piperidine lactone **11**, which was hydrolyzed with lithium hydroxide and passed through an ammonium satd ion exchange resin to yield the ammonium salt of **1**. This synthetic sequence was also used to prepare [^{14}C]-labeled compound **1**. Carbon-14 was introduced as diethyl[^{14}C]-methylphosphonate which was synthesized from commercially available [^{14}C]-methyl iodide and triethyl phosphite.

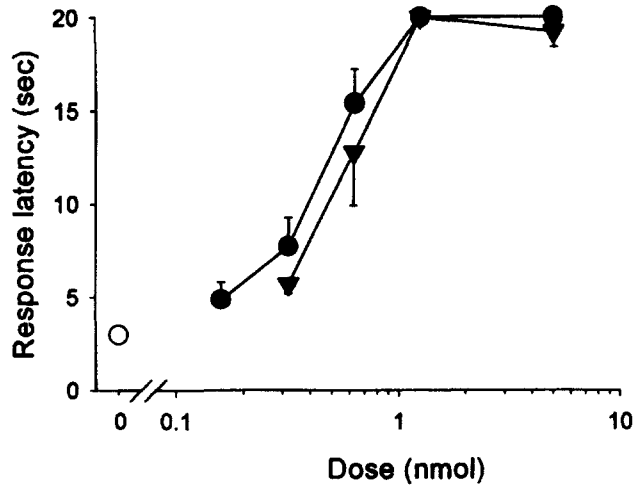


Figure 3. Hot-plate (triangles = compound (±)-**1**, circles = compound (±)-**2**).

Biological results

The binding affinities (K_i) to the glutamate site of the NMDA receptor were determined by measuring the competitive inhibition of binding of [^{3}H]-CGS 19755 to rat cerebral cortex membranes.[11] Compound **1** (LAS 250) and its antipode, the 2*S*,3*R*,1′S compound **3** yielded values of 75 and 11,700 nM, respectively. In the same assay CGS 19755 exhibited a K_i value of 40 nM and the ketone **2** of 64 nM.

The analgesic effects were evaluated using the mouse hot-plate test (58 °C) 15 min after direct intrathecal injection of the test substance into the lumbar subarachnoidal space. Kicking, shaking, and licking of a hind paw were used as criterion responses, and a cut-off of 20 s was observed in the absence of any response. Data are presented as mean ± SEM, n = 6–12 per dose (Fig. 3). ED_{50} (95% confidence limits): compound (±)-**1** 0.53 (0.42–0.65) nmol and compound (±)-**2** 0.51 (0.41–0.64) nmol.

The analgesic effects of the racemates of compounds **1** and **2** were also evaluated using the mouse formalin test.[12] Whereas the hot-plate test uses an acute thermal stimulus, the formalin test employs a tonic painful stimulus with both a first phase of direct chemical activation and a second phase involving central sensitization and peripheral inflammation.[12] The test substances were given by direct intrathecal injection into the lumbar subarachnoidal space 15 min before formalin. A 20 μL quantity of 1% formaldehyde were injected into a hindpaw, and licking of the paw was scored 0–5 min (first phase) and 15–30 min (second phase) after formalin (Fig. 4). ED_{50} (95% confidence limits) for compound (±)-**2** were 0.27 (0.22–0.36) nmol for the first phase and 0.16 (0.02–0.22) nmol for the second phase.

The racemate of compound **1** showed similar potency; 0.32 nmol reducing the response during the first phase by 72%, and the response during the second phase by

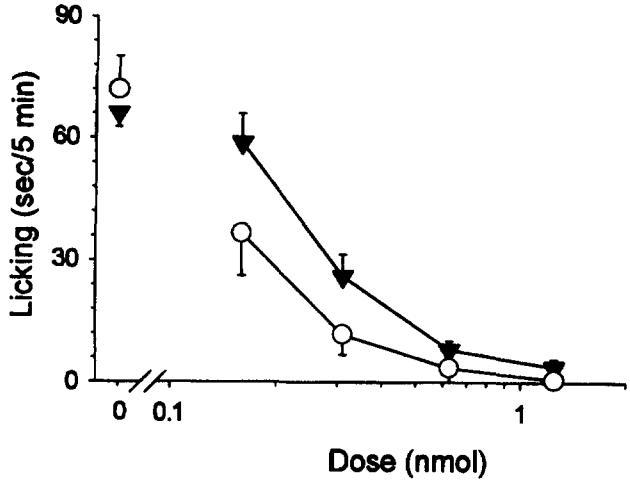


Figure 4. Formalin test compound (±)-**2** (triangles = first phase, circles = second phase).

81%. It can be concluded that compounds **1** and **2** have a very good analgesic effect when given intrathecally, being two to 10 times more potent than morphine under the same testing conditions (data not shown).

Experimental

Melting points were determined on a Büchi capillary melting point apparatus and were uncorrected. All chemicals and reagents were used as received from suppliers. ^{1}H and ^{13}C NMR spectra were recorded on a Varian Unity 400 (400 MHz) spectrometer in $CDCl_3$ with TMS as an internal standard, unless otherwise stated. Mass spectra were recorded on a Finnigan MAT SSQ 7000 spectrometer. The GC-analyses were performed on a Carlo-Erba HRGC 4160 chromatograph equipped with a SE-54 column. Optical rotations were measured on a Perkin–Elmer 241 polarimeter at the sodium D line at 25 °C. Flash column chromatography was carried out on silica gel 60 (230–400 mesh). The elemental microanalyses were performed at Mikro Kemi AB in Uppsala, Sweden.

Isopropyl 3-acetylpyridine-2-carboxylate (5). Dimethylmalonate (53 mL, 455 mmol), triethylamine (126.8 mL, 910 mmol), and anhydrous magnesium chloride (25.0 g, 273.0 mmol) were mixed in toluene (350 mL) and stirred at ambient temperature for 1.5 h. The mixture was cooled to 0 °C and **4**[4] (87 g, 382.4 mmol) in toluene (50 mL) was added dropwise over 30 min. The cooling bath was removed and 37% HCl (100 mL) was added dropwise. The mixture was washed with water (2 × 100 mL). The organic phase was concentrated at reduced pressure. The residue was dissolved in DMSO (380 mL) and water (14 mL) and heated to 125 °C for 2 h (gas evolution was observed). The tan-colored solution was allowed to cool and then diluted with water (760 mL). The mixture was extracted with ethyl acetate (760 mL + 2 × 300 mL). The combined organic phases were washed with satd $NaHCO_3$ solution (360 mL), water (200 mL), and concentrated at reduced pressure to afford crude **5** (65.4 g) as a brownish oil, which was distilled on a Büchi kügel rohr apparatus at 135–140 °C, 0.2 mbar yielding 28.2 g (50%) of **5**. ^{1}H NMR: δ 1.41 (s, 3H, CH_3), 1.42 (s, 3H, CH_3), 2.59 (s, 3H, CH_3), 5.32 (m, 1H, CH–), 7.53 (q, 1H, ArH), 7.91 (q, 1H, ArH), 8.76 (q, 1H, ArH); ^{13}C NMR: δ 21.2 (CH_3), 29.0 (CH_3), 69.9 (CH–), 125.0 (CH=), 134.9 (CH=), 136.0 (C=), 147.6 (C=), 150.5 (CH=), 165.1 (COO–), 199.3 (C=O). MS (TSP) *m/z* 208 (M+1).

Isopropyl 3-(bromoacetyl)pyridine-2-carboxylate (6). A mixture of **5** (179.5 g, 805.8 mmol), AcOH (180 mL), and HBr, 30% in AcOH (15 mL) was heated to 100 °C before dropwise addition of Br_2 (44 mL, 858.5 mmol). After the addition was complete, the reaction mixture was allowed to cool to ambient temperature. The mixture was concentrated in vacuo giving a crystalline residue, which was suspended in water (230 mL). The pH was adjusted to 3–4 with NaOH (45%), and the crystals that were isolated by filtration were washed with water (120 mL) and recrystallized from ethanol (300 mL) yielding 122.7 g (50%) of **6**. ^{1}H NMR: δ 1.41 (s, 3H, CH_3), 1.42 (s, 3H, CH_3), 4.39 (s, 2H, CH_2–Br), 5.34 (m, 1H, CH–), 7.59 (q, 1H, ArH), 7.81 (q, 1H, ArH), 8.86 (q, 1H, ArH); ^{13}C NMR: δ 21.6 (CH_3), 34.2 (CH_2–Br), 63.4 (CH–), 125.9 (CH=), 136.4 (C=), 136.5 (CH=), 145.7 (C=), 150.9 (CH=), 164.7 (COO–), 195.0 (C=O). MS (TSP) *m/z* 286,288 (M+1)

5-Bromomethyl-5,7-dihydrofuro[3,4-*b*]pyridin-7-one (7). To $NaBH_4$ (21.0 g, 555.1 mmol) suspended in dry THF (515 mL), AcOH (106 mL, 1.85 mol) was added at such a rate that the reaction temperature did not exceed 30 °C and the gas evolution could be kept under control. The mixture was stirred until the gas evolution ceased, whereupon **6** (64.4 g, 211.3 mmol) was added in one portion. After stirring for 40 h, water (40 mL) was carefully added (gas evolution) and the reaction mixture was filtered and concentrated in vacuo. The residue was dissolved in toluene (320 mL) and washed with water (2 × 320 mL). Half the amount of toluene added was evaporated before AcOH (32 mL) was added. The resulting solution was heated to 70 °C for 8 h, and then slowly cooled to 20 °C, with repeated seeding of the solution with pure **7**. The resulting slurry was allowed to stand at 20 °C for 2.5 h and filtered. The crystals were washed with toluene (60 mL) and then dried in vacuo yielding 34.8 g (69%) of **7**. ^{1}H NMR: δ 3.76 (m, 1H, CH_2–Br), 3.90 (m, 1H, CH_2–Br), 5.80 (m, 1H, CH–O), 7.63 (q, 1H, ArH), 8.13 (d, 1H, ArH), 8.94 (d, 1H, ArH); ^{13}C NMR: δ 31.4 (CH_2–Br), 76.6 (CH–O), 127.3 (CH=), 131.5 (CH=), 141.45 (C=), 144.9 (C=), 152.3 (CH=), 166.9 (COO–).

5-Diethylphosphonomethyl-5,7-dihydrofuro[3,4-*b*]pyridin-7-one (8). A mixture of **7** (10.0 g, 43.0 mmol) and triethyl phosphite (100 mL, 583 mmol) was heated at 100 °C under a gentle flow of nitrogen gas. After 12 h, *n*-heptane (100 mL) was slowly added to the vigorously stirred mixture. The resulting slurry was cooled to 0 °C and the solid was filtered off and washed with heptane (40 mL). The solid was suspended in water (50 mL) and

stirred vigorously for 1 h. The slurry was neutralized with satd sodium bicarbonate solution and the insoluble material was filtered off and discarded. The mother liquid was saturated with NaCl and extracted with ethyl acetate (150 mL). The organic phase was concentrated in vacuo affording 3.48 g (25%) of **8**. The crude product was used without any further purification. An analytical sample was recrystallized from toluene–diethyl ether. ^{1}H NMR: δ 1.26 (t, 3H), 1.34 (t, 3H), 2.35 (m, 1H, –CH_2–P), 2.45 (m, 1H, –CH_2–P), 4.07 (q, 2H, OCH_2–), 4.17 (q, 2H, OCH_2–), 5.79 (m, 1H, CH–O), 7.55 (q, 1H, ArH), 8.17 (d, 1H, ArH), 8.84 (d, 1H, ArH); ^{13}C NMR: δ 16.3 (d, CH_3), 31.4 (d, J_{CP} = 142 Hz, CH_2–P), 62.3 (d, OCH_2), 74.2 (CH–O), 127.1 (CH=), 131.9 (CH=), 142.9 (d, C=), 144.2 (C=), 152.8 (CH=), 167.1 (COO–). Anal. calcd for $C_{12}H_{16}NO_5P$: C 50.5; H 5.6; N 4.9. Found C 50.4; H 5.6; N 4.9.

(1′*R*)-3-(1-Hydroxy-2-diethylphosphonoethyl)-pyridine-2-[N-(*S*)-α-1-naphthylethyl]-carboxamide (9). A suspension of **8** (57.0 g, 200 mmol), (*S*)-(−)-α-(1-naphtylethylamine) (51.6 mL, 320 mmol), and 2-hydroxypyridine (21.0 g, 220 mmol) in toluene (560 mL) was heated to 45 °C. The resulting clear solution was stirred at room temperature for 16 h, washed with satd ammonium chloride solution (3 × 200 mL) and concentrated in vacuo. The resulting viscous oil was dissolved in diethylether (75 mL), and the racemate of **9** immediately precipitated. Filtration followed by washing with diethylether (45 mL) and drying afforded 73.8 g of the racemic product which was recrystallized twice from diethylether:ethanol 25:1 (2 × 735 mL) yielding 19.7 g (43%) of **9** in more than 95% de, as determined by proton NMR. ^{1}H NMR: δ 1.20 (t, 3H), 1.24 (t, 3H), 1.79 (d, 3H, –CH_3), 2.40 (m, 1H, –CH_2–P), 2.49 (m, 1H, –CH_2–P), 4.07 (q, 2H, OCH_2–), 4.17 (q, 2H, OCH_2–), 5.50 (m, 1H, CH–Ar), 6.12 (m, 2H, CH–O, OH), 7.38–8.64 (m, 11H, ArH, NH); ^{13}C NMR: δ 16.3 (d, CH_3), 21.3 (CH_3), 34.0 (d, J_{CP} = 137 Hz, CH_2–P), 44.9 (CH), 62.3 (d, OCH_2), 68.2 (CH–O), 122.0, 122.5, 125.0, 125.8, 126.0, 126.2, 128.0, 128.2, 131.0, 134.0, 138.0, 138.4, 140.8, 145.8, 147.0 (Ar), 164.9 (COO–). MS (TSP) *m/z* 457 (M+1).

5-Phosphonomethyl-(5*R*)-5,7-dihydrofuro[3,4-*b*]pyridin-7-one (10). A mixture of **9** (19.96 g, 43.7 mmol) in 6 M HCl (400 mL) was heated at reflux for 12 h. The solution was cooled to room temperature, filtered, and concentrated at reduced pressure. The residue was dissolved in ethanol (50 mL) and water (5 mL), and stirred at ambient temperature during the crystallization phase. The resulting slurry was cooled to 0 °C and crude **10** was filtered off. The product was recrystallized from ethanol:water 2:1 (145 mL) affording 7.34 g (72%) of **10**. ^{1}H NMR (D_2O, 400 MHz): δ 2.25 (m, 1H, –CH_2–P), 2.45 (m, 1H, –CH_2–P), 5.84 (m, 1H, CH–O), 7.65 (q, 1H, ArH), 8.17 (d, 1H, ArH), 8.84 (d, 1H, ArH); ^{13}C NMR: δ 31.0 (d, J_{CP} = 138Hz, CH_2–P), 74.6 (CH–O), 128.1 (CH=), 133.9 (CH=), 142.0 (d, C=), 144.2 (C=), 152.1 (CH=), 169.8 (COO–). Anal. calcd for $C_8H_8NO_5P$: C 41.9; H 3.5; N 6.1; P 13.5. Found C 41.7; H 3.4; N 6.0; P 13.7. $[\alpha]_D$ −77.3 (*c* 1.0, 1M HCl)

5-Phosphonomethyl-(4a*S*,5*R*,7a*R*)-perhydrofuro[3,4-*b*]pyridin-7-one (11). Rh/C 5% (23.56 g) was added to **10** (30.81 g, 134.47 mmol) dissolved in water (1760 mL). The mixture was hydrogenated for 16 h in a Büchi pressure reactor at 3.5 bar. The catalyst was filtered off, and 1700 mL of the solvent was evaporated. The residual solution was used in the next step without any further purification. An analytical sample was recrystallized from water. **11** was 90% diastereomerically pure as determined by proton NMR. mp 278–280 °C. ^{1}H NMR (D_2O, 400 MHz): δ 1.24 (qd, 1H, C(4)Hax), 1.54 (qt, 1H, C(5)Hax), 1.85–2.12 (m, 4H, C(5)Heq, C(4)Heq, CH_2–P), 2.76 (td, 1H, C(6)Hax), 2.89 (m, 1H, C(3)Hax), 3.30 (br d, 1H, C(6)Heq), 4.84 (m, 1H, CH–O); ^{13}C NMR: δ 18.2 (CH_2–), 19.2 (CH_2–), 26.5 (d, J_{CP} = 138 Hz, CH_2–P), 36.8 (d, CH–), 41.7 (CH_2–), 54.5 (CH–), 77.6 (–CH–O), 171.1 (COO–). MS (TSP) *m/z* 236 (M+1), 156 (M–PO_3H_2). Anal. calcd for $C_8H_{14}NO_5P$: C 40.8; H 6.0; N 5.9; P 13.2. Found C 40.8; H 5.9; N 5.9; P 13.5. $[\alpha]_D$ −21.6 (*c* 1.0, 1M HCl).

Disodium salt of (2*R*,3*S*,1′*R*)-3-(1-hydroxy-2-phosphonoethyl)-2-piperidinecarboxylic acid (1). Solid NaOH (15.51 g, 387.8 mmol) was added in one portion to a solution of **11** (38.0 g, 161.6 mmol) in water (1500 mL). The solution was allowed to stand at ambient temperature for 1.5 h before being passed through a column filled with Dowex 50WX8-100 (1275 g) saturated with NH_3. The product was eluted with 1 M ammonium hydroxide solution yielding 48.3 g of **1**. This product was dissolved in water (45 mL), and solid NaOH (12.93 g, 323.3 mmol) was added in small portions. The solution was stirred for 1 h and then concentrated under reduced pressure. The residue was dissolved in water (45 mL) and methanol (225 mL) was added dropwise along with concomitant seeding of small amounts of **1** (disodium salt). A thick slurry formed and the product was filtered and dried to afford 30.6 g (63%, 90% diastereomeric purity) of **1** (disodium salt). ^{1}H NMR (D_2O, 400 MHz): δ 1.45–2.00 (m, 6H, C(4)H_2, C(5)H_2, CH_2-P), 2.12 (br s, C(3)Heq), 2.85 (td 1H, C(6)Hax), 3.24 (br d, 1H, C(6)Heq), 3.71 (d, *J* = 3.8 Hz, 1H, C(2)Hax), 4.09 (m, 1H, CH–O); ^{13}C NMR: δ 19.6 (CH_2–), 21.6 (CH_2–), 33.9 (d, J_{CP} = 128 Hz, CH_2–P), 37.6 (d, CH–), 43.9 (CH_2–), 62.1 (CH–), 69.1 (–CH–O), 173.2 (COO–). MS (FAB) *m/z* 254 (M+1). Anal. calcd for $C_8H_{14}NO_6P$*2Na*H_2O: C 30.4; H 5.1; N 4.4; P 9.8. Found C 30.3; H 5.0; N 4.4; P 9.9. $[\alpha]_D$ +26.0 (*c* 1.0, H_2O).

2-*tert*-Butyloxycarbonylnicotinic acid (13). A solution of pyridine-2,3-dicarboxylic acid anhydride (120 g, 0.56 mol), in *tert*-butanol (150 mL) and pyridine (100 mL), was stirred for 16 h at 40 °C. The solution was concentrated at reduced pressure and filtered through silica gel (ethyl acetate:acetone 4:1). The product was crystallized from diethylether affording 53 g (42%) of **13**. ^{1}H NMR: δ 1.63 (s, 9H, CH_3), 7.53 (dd, 1H), 8.34 (dd, 1H), 8.87 (d, 1H), 10.95 (br s, 1H); ^{13}C NMR: δ

27.9 (CH_3–), 84.0 (–O–C–), 124.9 (CH=), 125.0 (C=), 139.2 (CH=), 152.7 (CH=), 153.4 (C=), 165.8 (COO–), 170.1 (COO–).

***cis*-1,2-Di-*tert*-butyloxycarbonyl-3-piperidinecarboxylic acid (14).** Pd/C 10% (5.5 g) was added to a solution of **13** (24.5 g, 109 mmol) in acetic acid (275 mL). The mixture was hydrogenated in a Parr apparatus at 300 kPa for 16 h. The catalyst was filtered off and the solvent was evaporated to give 24 g (96%) of *tert*-butyl *cis*-3-carboxy-2-piperidinecarboxylate as a white solid, which was dissolved in methanol (300 mL). Triethylamine (16.2 mL, 116 mmol), dichloromethane (175 mL), and di-*tert*-butyl dicarbonate (25.6 g, 114 mmol) were added and the mixture was stirred for 16 h at room temperature. The reaction solution was concentrated and partitioned between ethyl acetate and 1 M HCl. The organic phase was washed with water, dried over sodium sulfate, and concentrated. The residue was purified by flash chromatography (ethyl acetate) to give 22.6 g (66%) of **14** as a colorless oil. ^{1}H NMR: δ 1.44 (s, 9H, CH_3), 1.48 (s, 9H, CH_3), 1.48–1.77 (m, 3H, C(5)Hax, C(5)Heq, C(4)Hax), 2.05 (m, 1H, C(4)Heq), 2.57–2.80 (m, 2H, C(6)Hax, C(3)Hax), 3.93 (br d, 0.5H, C(6)Heq), 4.05 (br d, 0.5H, C(6)Heq), 5.18 (d, 0.5H, C(2)Heq), 5.45 (d, 0.5H, C(2)Heq); ^{13}C NMR: δ 22.0, (d, CH_2–), 24.2 (d, CH_2–), 27.8 (CH_3–), 28.3 (CH_3–), 40.8 (d, CH_2–), 43.1 (d, CH–), 55.6 (d, CH–), 80.2 (d, C–), 82.5 (C–), 155.2 (d, N–COO–), 168.3 (d, COO–), 177.6 (d, COO–).

(2*R*,3*S*)-1,2-Di-*tert*-butyloxycarbonyl-3-piperidinecarboxylic acid (15). (+)-Ephedrine ((+)-2-methylamino-1-phenyl-propane-1-ol) (8.23 g, 49.8 mmol) was added to a solution of **14** (32.8 g, 99.6 mmol) in ethyl acetate (110 mL). The resulting mixture was left overnight at room temperature, and the precipitate was filtered off. Two recrystallizations from ethyl acetate yielded 12.5 g of **15** as a (+)-ephedrine salt, mp 166–169 °C. This salt was partitioned between ethyl acetate (150 mL) and 1 M HCl (35 mL). The organic phase was washed with water, dried over sodium sulfate, and concentrated in vacuo to yield 8.3 g (25% overall yield) of **15** as a colorless oil. GLC analysis of the Mosher acid derivative showed a 95% ee.[2] ^{1}H NMR: δ 1.43 (s, 9H, CH_3), 1.47 (s, 9H, CH_3), 1.46–1.76 (m, 3H, C(5)Hax, C(5)Heq, C(4)Hax), 2.03 (m, 1H, C(4)Heq), 2.57–2.80 (m, 2H, C(6)Hax, C(3)Hax), 3.90 (br d, 0.5H, C(6)Heq), 4.04 (br d, 0.5H, C(6)Heq), 5.17 (d, 0.5H, C(2)Heq), 5.44 (d, 0.5H, C(2)Heq).

(2*R*,3*S*)-1,2-Di-*tert*-butyloxycarbonyl-3-hydroxymethylpiperidine (16). Oxalyl chloride (2.3 mL, 25.2 mmol), followed by DMF (50 μL), were added to a stirred solution of **15** (8.3 g, 25.2 mmol) in toluene (150 mL) at 0 °C. The solution was stirred for 1 h at 0 °C and concentrated at reduced pressure. The residue was dissolved in THF (150 mL) and $NaBH_4$ (0.95 g, 25.2 mmol) was added in small portions at 0 °C. After stirring at 0 °C for 30 min, water (40 mL) was added carefully followed by ethyl acetate. The organic phase was washed with brine, dried over sodium sulfate, and concentrated in vacuo to yield 7.5 g (94%) of **16** as a colorless oil. ^{1}H NMR: δ 1.10 (qd, 1H, C(4)Hax), 1.43 (s, 9H, CH_3), 1.47 (s, 9H, CH_3), 1.57 (m, 1H, C(5)Hax), 1.76 (m, 1H, C(5)Heq), 2.01 (m, 1H, C(4)Heq), 3.05–3.20 (m, 2H, C(6)Hax, C(3)Hax), 3.60 (m, 2H,CH_2–O), 3.90 (br d, 0.5H, C(6)Heq), 4.05 (br d, 0.5H, C(6)Heq), 4.69 (d, 0.5H, C(2)Heq), 4.85 (d, 0.5H, C(2)Heq); ^{13}C NMR: δ 23.1 (d, CH_2–), 24.5 (d, CH_2–), 28.1 (CH_3–), 28.3 (CH_3–), 41.0 (d, CH–), 41.6 (d, CH_2–), 56.5 (d, CH–), 64.7 (d, CH_2–OH), 80.0 (d, C–), 82.3 (C–), 155.8 (d, N–COO–), 171.1 (COO–). $[\alpha]_D$ +27.8 (*c* 6.0, 95% ethanol).

(2*R*,3*S*)-1,2-Di-*tert*-butyloxycarbonyl-3-piperidinecarbaldehyde (17). DMSO (3.9 mL, 55 mmol) in CH_2Cl_2 (15 mL) was added to a stirred solution of oxalyl chloride (2.4 mL, 27.5 mmol) in CH_2Cl_2 (110 mL) at −75 °C. After 5 min, **16** (7.9 g, 25 mmol) in CH_2Cl_2 (25 mL) was added dropwise, and the resulting solution was stirred at −75 °C for 25 min. Triethylamine (17.5 mL, 126 mmol) was added dropwise, after which the temperature was raised to room temperature over 40 min. The solvent was evaporated. The residue was dissolved in toluene and washed with satd citric acid solution, water, satd sodium bicarbonate solution, and water again. The organic phase was dried over sodium sulfate and concentrated in vacuo. The residue was purified by rapid flash chromatography (hexane:ethyl acetate 9:1) affording 3.2 g (40%) of **17** as a colorless oil. ^{1}H NMR: δ 1.20–1.60 (m, 2H, C(5)Hax, C(5)Heq), 1.40 (s, 9H, CH_3), 1.45 (s, 9H, CH_3), 1.75 (m, 1H, C(4)Hax), 2.16 (m, 1H, C(4)Heq), 2.43 (m, 1H, C(3)Hax), 2.74 (t, 0.5H, C(6)Hax), 2.87 (t, 0.5H, C(6)Hax), 3.95 (br d, 0.5H, C(6)Heq), 4.08 (br d, 0.5H, C(6)Heq), 5.18 (d, 0.5H, C(2)Heq), 5.42 (d, 0.5H, C(2)Heq), 9.71 (s, 1H, CHO); ^{13}C NMR: δ 20.2 (d, CH_2–), 23.8 (d, CH_2–), 27.8 (CH_3–), 28.2 (CH_3–), 41.4 (d, CH_2–), 50.4 (d, CH–), 55.4 (d, CH–), 80.2 (C–), 83.0 (d, C–), 155.2 (d, –NCOO–), 168.2 (d, COO–), 198.2 (d, CHO). $[\alpha]_D$ +39 (*c* 2.45, 95% ethanol).

***tert*-Butyl-(2*R*,3*S*,1′*R*)-1-*tert*-butyloxycarbonyl-3-(1-hydroxy-2-diethylphosphonoethyl)-2-piperidinecarboxylate (18).** *n*-Butyl lithium (2.2 M in hexane, 0.86 mL, 1.9 mmol) was added dropwise to a stirred solution of diethyl methylphosphonate (0.29 mL, 1.9 mmol) in THF (10 mL) at −78 °C. After 25 min at −78 °C the solution was added to **17** (0.5 g, 1.6 mmol) in THF (10 mL) while maintaining the temperature at −78 °C. The reaction mixture was stirred at −78 °C for an additional 45 min, followed by addition of ammonim chloride (0.6g) and ethanol (2 mL). The temperature was raised to room temperature. The mixture was filtered and concentrated. The residue was purified by flash chromatography (hexane:ethyl acetate) to yield 0.38 g (51%, 85% diastereomeric purity) of **18** as a slightly yellow oil. ^{1}H NMR: δ 1.20–1.50 (m, 2H, C(5),C(4)Hax), 1.34 (d t, CH_3–), 1.45 (s, 9H, CH_3–), 1.48 (s, 9H, CH_3–), 1.75 (m, 1H, C(5)Heq), 2.05 (m, 1H, C(4)Heq), 2.30 (m, 2H, CH_2–P), 3.09 (m, 2H, C(3)Hax, C(6)Hax), 3.85 (m, 1H,C(6)Heq) 4.10 (m, 5H, OCH_2–, CH–O), 4.59 (d, 0.5H, C(2)Heq), 4.80 (d, 0.5H,

C(2)Heq); ^{13}C NMR: δ 16.2 (CH_3–), 20.9 (d, CH_2–), 24.6 (d, CH_2–), 27.9 (CH_3–), 28.2 (CH_3–), 31.1 (dd, J_{CP} = 138 Hz, CH_2–P), 41.1 (d, CH_2–), 45.9 (m, CH–), 56.7 (d, CH–), 61.7 (d, OCH_2–), 67.3 (–CHOH), 79.8 (C–), 82.0 (C–), 155.3 (d, –NCOO–), 170.4 (d, COO–). $[\alpha]_D$ +5.2 (*c* 5.8, 95%ethanol).

5-Phosphonomethyl-(4a*S*,5*R*,7a*R*)-perhydrofuro[3,4-*b*-]pyridin-7-one (11). TMSBr (0.3 mL, 2.3 mmol) was added to a solution of **18** (0.13 g, 0.28 mmol) in $CHCl_3$ (4 mL). The mixture was stirred for 4 h at 70 °C in a sealed vessel. After evaporation of the solvent, water (2 mL) was added and the resulting solution stirred for 15 min. The solution was concentrated and the residue crystallized from the water–ethanol mixture to afford 55 mg (90%, 90% diastereomeric purity) of **11** as a white solid, mp 278–280 °C. ^{1}H NMR (D_2O, 400 MHz): δ 1.24 (qd, 1H, C(4)Hax), 1.54 (qt, 1H, C(5)Hax), 1.85–2.12 (m, 4H, C(5)Heq, C(4)Heq, CH_2-P), 2.76 (td, 1H, C(6)Hax), 2.89 (m, 1H, C(3)Hax), 3.30 (br d, 1H, C(6)Heq), 4.84 (m, 1H, CH–O); ^{13}C NMR: δ 18.2 (CH_2–), 19.2 (CH_2–), 26.5 (d, J_{CP} = 132Hz, CH_2–P), 36.8 (d, CH–), 41.7 (CH_2–), 54.5 (CH–), 77.6 (–CH–O), 171.1 (COO–). MS (TSP) *m/z* 236 (M+1), 156 (M–PO_3H_2). Anal. calcd for $C_8H_{14}NO_5P$: C 40.8; H 6.0; N 5.9; P 13.2. Found C 40.8; H 5.9; N 5.9; P 13.5. $[\alpha]_D$ −7.7 (*c* 3.28, H_2O).

Ammonium salt of (2*R*,3*S*,1′*R*)-3-(1-hydroxy-2-phosphonoethyl)-2-piperidinecarboxylic acid (1). LiOH (0.04 g) was added to a solution of **11** (0.037 g, 0.15 mmol) in water (0.5 mL). The mixture was stirred for 3 h at room temperature and eluted through an ammonium satd Dowex 50WX8-100 column, with 1 M NH_3 as eluent, to give 0.033 g (82%, 90% diastereomeric purity) of **1** as an ammonium salt. ^{1}H NMR (D_2O, 400 MHz): δ 1.45–2.00 (m, 6H, C(4)H_2, C(5)H_2, CH_2–P), 2.12 (br s, C(3)Heq), 2.85 (td 1H, C(6)Hax), 3.24 (br d, 1H, C(6)Heq), 3.71 (d, *J* = 3.8Hz, 1H, C(2)Hax), 4.09 (m, 1H, CH–O); ^{13}C NMR: δ 19.6 (CH_2–), 21.6 (CH_2–), 33.9 (d, J_{CP} = 128 Hz, CH_2–P), 37.6 (d, CH–), 43.9 (CH_2–), 62.1 (CH–), 69.1 (–CH–O), 173.2 (COO–). MS (FAB) *m/z* 254 (M+1). Anal. calcd for $C_8H_{15}NO_6P^*NH_4$: C 35.6; H 7.1. Found C 35.6; H 7.0. $[\alpha]_D$ +23.3 (*c* 1.65, H_2O).

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10. Measured ^{1}H-coupling constants (400 MHz in $CDCl_3$) *J* = 6.3 Hz for 8H–9H and *J* = 4.1 Hz for 9H–5H. Calculated coupling constants MMX minimized model for *endo J* = 6.0 Hz and *J* = 4.1 Hz, respectively, and for *exo J* = 6.5 Hz and *J* = 1.0 Hz, respectively.

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