

SYNTHESIS AND RESOLUTION OF SUBSTITUTED PIPECOLIC ACIDS

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Abstract—A new general method for the preparation of 2-alkyl-pipecolic acids has been developed. The syntheses of 2-methyl-, 2-benzyl- and *cis*-6-methylpipecolic acids are described. (–)S- and (+)R-2-methylpipecolic acids were resolved by fractional crystallization of the quinine salt of their *N*-carbobenzoxy derivative. Isolation of (–)S-*cis*-6-methylpipecolic acid required the use of 4-phenylbenzyloxycarbonyl as protecting group to achieve selective crystallization of the quinine salt. The absolute configurations of these compounds were determined by circular dichroism.

INTRODUCTION

For some time we have been involved in the study of substituent effects on the polymerization of cyclic imino acids and of the conformational behaviour of the corresponding polypeptides [2–6]. This paper deals with the syntheses of 2-methyl-, 2-benzyl- and *cis*-6-methylpipecolic acids (2MPA, 2BPA and *c*6MPA, respectively) and with the resolution of 2-methyl- and *cis*-6-methylpipecolic acids. The following papers in this series will describe the dimerization of *N*-carboxyanhydrides of 2MPA and *c*6MPA [7] and report on the conformations of some diketopiperazines derived from 2MPA *c*6MPA [7] and report on the conformations of some diketopiperazines derived from 2MPA and *t*6MPA in solution and in the solid state [8].

EXPERIMENTAL

All melting points were determined in open capillary tubes on a Thomas Hoover Capillary Melting Point Apparatus and were uncorrected. Elemental analyses were performed by Spang Laboratories, Inc., Knoxville, Tennessee. Specific rotations at the sodium D line were measured by a Perkin–Elmer 241 MC and Rudolf polarimeter. Circular dichroism spectra were recorded on a JASCO J-40A spectropolarimeter using 0.1–1.0 mm cells. I.R. spectra were recorded on a Perkin–Elmer Model 257 spectrophotometer. NMR spectra were recorded on a Varian T-60 spectrophotometer.

Ethyl-1-methyl-2-oxocyclopentanecarboxylate (2b)

Compound **2b** was prepared according to the method of Claisen as modified by Barco, Benetti and Pollini [14]. Vigorous stirring of a mixture of K_2CO_3 (351.6 g, 2.54 mol), acetone (500 ml), ethyl 2-oxocyclopentanecarboxylate (99.4 g, 0.636 mol) and iodomethane (180.5 g, 1.27 mol), produced ethyl 1-methyl-2-oxocyclopentanecarboxylate (106.0 g, 0.62 mol) in 97.5% yield; b.p. 117–118°/20 mmHg (lit. [14] 107°/15 mmHg).

NMR ($CDCl_3$) δ : 1.23 (3H, t, $J = 7$ Hz), 4.1 (2H_X, q, $J = 7$ Hz); 1.27 (3H, S); 1.80–2.40 (6H, m).

I.R. (neat): 2880–2980, 1750, 1725, 1460, 1450, 1410, 1380, 1320, 1275, 1240, 1190, 1150, 1070, 1030, 950, 865 and 815 cm^{-1} .

Methyl 1-methyl-2-oxocyclopentanecarboxylate (2a)

The same procedure used for the preparation of **2b** was employed here. The scale was three times that of the above example; yield 87–97%; b.p.; 96°/11 mmHg, (lit. [7] 102°C/13 mmHg).

NMR ($CDCl_3$) δ : Methyl ester, 3.6 (3H, s); methylene group, 1.8–2.5 (6H, m); α -methyl, 1.25 (3H, s).

I.R. (neat): 2890–2990, 1760, 1730, 1465, 1455, 1440, 1410, 1380, 1320, 1275, 1240, 1200, 1160, 1070, 985 and 945 cm^{-1} .

Ethyl 2-methyl-6-oxopiperolate (3b)

The procedure of Plotniewski [13] was modified in the following manner. In a 2 l, 3-necked flask equipped with mechanical stirrer, pressure equalizing funnel, thermometer, and gas outlet were placed a dry solution of hydrazoic acid in chloroform (700 ml/0.45 N, 0.315 mol) and **2b** (34 g, 0.2 mol). The solution was stirred and cooled to –10°. Conc. sulphuric acid (70 ml) was added dropwise (2 hr), keeping the temperature below –5°. About 5.5 l of N_2 were evolved. Stirring was continued at room temperature for one additional hour and the excess of hydrazoic acid evolved was trapped in aqueous NaOH. The chloroform layer was decanted from the H_2SO_4 layer which contained most of the product. The H_2SO_4 layer was poured over ice (500 g) with stirring and external cooling, neutralized (Na_2CO_3), and extracted with methylene chloride. The chloroform layer was combined with the methylene chloride extracts. After drying, filtration and evaporation of the solvents in a rotary evaporator, a yellow oily residue was obtained which solidified upon standing. Crystallization was carried out in 1,2-dimethoxyethane to give 25 g (67% yield) of colorless crystals, m.p. 64–65°.

Analysis. Calculated for $C_9H_{15}NO_3$: C, 58.35; H, 8.11; N, 7.57. Found: C, 58.38; H, 8.05; N, 7.65.

NMR ($CDCl_3$) δ : Amide, 7.2 (1H, s); ethyl ester (A_3X_2 pattern), 4.15 (2H_X, q, $J_{AX} = 7$), 1.3 (3H_A, t, $J_{AX} = 7$); methylene group, 1.6–2.5 (6H, m); α -methyl, 1.45 (3H, s).

I.R. (KBr): 3200, 3080, 2980–2900, 1740, 1670, 1460, 1410, 1340, 1300, 1245, 1180, 1125, 1030 and 830 cm^{-1} .

Methyl 2-methyl-6-oxopiperolate (3a)

The above procedure was used with the following modifications. Hydrazoic acid in chloroform (2000 ml/0.65 N, 1.3 mol), **2a** (156 g, 1.0 mol), and H_2SO_4 (200 ml) were treated as in the above example (4 hr). During the work-up procedure, the solid sodium sulphate, resulting

from neutralizing the H_2SO_4 , was filtered before extraction. The yellow solid product was triturated with diethyl ether to remove impurities and colour. Crystallization from 1,2-dimethoxyethane raised the melting point from 98–99 to 98.5–99.5° and gave pure colourless crystals (128.0 g, yield 75%).

Analysis. Calculated for $\text{C}_8\text{H}_{13}\text{NO}_3$: C, 56.13; H, 7.65; N, 8.18. Found: C, 56.20; H, 7.56; N, 8.27.

NMR (CDCl_3) δ : Amide, 7.60 (1H, s); methyl ester, 3.75 (3H, s); methylene group, 1.6–2.5 (6H, m); α -methyl, 1.45 (3H, s).

I.R. (KBr): 3340, 3220, 2890–2990, 1740, 1680, 1450, 1380, 1350, 1250, 1190, 1125, 1075, 1040 and 770 cm^{-1} .

Ethyl 2-methylpiperolate (6b)

The procedure of Borch [15] for the reduction of secondary amides was used. Triethyloxonium tetrafluoroborate [16] (6.64 g, 35 mmol), **3b** (6.0 g, 32.4 mmol), methylene chloride (25 ml), absolute ethanol (30 ml) and sodium borohydride (3.0 g, 79 mmol) were used to give ethyl 2-methylpiperolate (3.75 g, 21.9 mmol) in 67.6% yield; b.p. 78°/11 mmHg (n_D^{25} , 1.4497).

Analysis. Calculated for $\text{C}_9\text{H}_{17}\text{NO}_2$: C, 63.12; H, 10.00; N, 8.17. Found: C, 62.98; H, 9.92; N, 7.92.

NMR (neat) δ : Ethyl ester (A_3X_2 pattern), 4.10 (2H_X, q, $J_{\text{AX}} = 7$ Hz), 1.20 (3H_A, t, $J_{\text{AX}} = 7$ Hz); methylene group, (CH₂)₂, 2.65 (2H, m); (CH₂)₂, 2.20 (1H, m), (CH₂)₂, 1.30–1.55 (5H, m); amine 1.40 (1H, s), α -CH₃, 1.15 (3H, s).

I.R. (neat): 3330, 2950, 2860, 1730, 1475, 1450, 1375, 1295, 1240, 1130–1190, 1105, 1055, 1030 and 700 cm^{-1} .

Methyl 2-methylpiperolate (6a)

The procedure of Borch [15] was modified slightly for large-scale synthesis: (1) The reduction of the imino ether salt was carried out at 0° for 6 hr, and at 25° for 12 hr; (2) the work-up procedure was to filter the solid material, evaporate the ethanol and then to treat with water and extract the product with methylene chloride. Triethyloxonium tetrafluoroborate [16] (102.5 g, 0.54 mol), **3a** (85.5 g, 0.5 mol), methylene chloride (400 ml), absolute ethanol (600 ml) and sodium borohydride (46.3, 1.2 mol) were used to obtain methyl 2-methylpiperolate (57.0 g) in 73% yield after vacuum distillation; b.p. 67°/8 mmHg.

Analysis. Calculated for $\text{C}_8\text{H}_{15}\text{NO}_2$: C, 61.10; H, 9.62; N, 8.91. Found: C, 61.15; H, 9.61; N, 8.85.

NMR (neat) δ : Methyl ester, 3.65 (3H, s); methylene group, (CH₂)₂, 2.70 (2H, m), (CH₂)₂, 2.10 (1H, m), (CH₂)₂, 1.3–1.55 (5H, m), α -CH₃, 1.15 (3H, s).

I.R. (neat): 3340, 2870, 2950, 1735, 1450, 1370, 1300, 1240, 1100–1200, 1050, 1000, 990, 930 and 750 cm^{-1} .

Methyl 2-methyl-6-ethoxy-2,3,4,5-tetrahydropicolinate (5a)

The free imino ether was obtained by washing its fluoroborate salt in methylene chloride with ice-cold sodium carbonate solution. An analytical sample was prepared and distilled under vacuum; b.p. 125°/20 mmHg.

Analysis. Calculated for $\text{C}_{10}\text{H}_{17}\text{NO}_3$: C, 60.28; H, 8.60; N, 7.03. Found: C, 60.23; H, 8.60; N, 6.99.

NMR (CDCl_3): Ethyl ether (A_3X_2 pattern), 4.1 (2H_X, q, $J_{\text{AX}} = 7$ Hz), 1.25 (3H_A, t, $J_{\text{AX}} = 7$ Hz) methyl ester: 3.65 (3H, s), methylene group, 2.3–1.6 (6H, m); α -methyl 1.40 (3H, s).

I.R. (neat): 2990–2960, 1740, 1675, 1450, 1375, 1350, 1245–1170, 1120, 1070, 1040 and 770 cm^{-1} .

2-Methylpiperolic acid monohydrate (7)

Compound **6a** (200 g, 1.27 mol) and water (2000 ml) were refluxed for 1 hr (the ethyl ester needed a longer reaction time, 10 hr). The reaction mixture was evaporated to dryness and the white residue was crystallized from an ethanol–water mixture (70:30 by vol.). White crystalline needles of **7** (193.0 g) were obtained in 94% yield; m.p. 340° with sublimation.

Analysis. Calculated for $\text{C}_7\text{H}_{13}\text{NO}_2 \cdot \text{H}_2\text{O}$: C, 52.16; H, 9.38; N, 8.69. Found: C, 52.21; H, 9.36; N, 8.72.

Mass spectrum (m/e , abundance): (144, 0.04), (128, 1.0), (99, 5.4), (98, 100), (82, 5.9), (70, 16.0), (69, 3.9), (55, 9.0), (42, 18.0), (41, 10.8), (39, 5.7), (30, 7.2), (28, 14.0).

NMR (D_2O) δ : (CH₂)₂, 3.3–3.6 (2H, m), (CH₂)₂, 1.8–2.7 (6H, m), α -CH₃, 1.7 (3H, s).

I.R. (KBr): 3400–2400, 1590–1620, 1515, 1450, 1395, 1365, 1345, 1290, 1180, 1110, 1060, 1040, 980, 870, 850 and 780 cm^{-1} .

N-carbobenzoxy-2-methylpiperolic acid (17)

Compound **7** (32.2 g, 0.20 mol) was dissolved in aqueous NaOH (50 ml 4 N, 0.20 mol). The reaction flask was immersed in an ice-bath and the solution was stirred vigorously with a mechanical stirrer. Benzyl chloroformate (51.1 g, 0.30 mol) and aqueous NaOH (75 ml 4 N, 0.30 mol) were added alternately in portions over a period of 30 min, maintaining the temperature below 5°. The pasty reaction mixture was stirred and cooled for one additional hour, and the excess benzyl chloroformate was extracted with ether. About 1 litre of water was added to the reaction mixture and brought to boiling. The heterogeneous mixture turned homogeneous after 2.5 hr was subsequently cooled and acidified to pH 4. The resultant milky solution was left at 0° overnight. Crystalline solid was formed, filtered, dried and recrystallized from chloroform/petroleum ether. Colourless crystals were obtained (35.7 g, 68% yield); m.p. 119.5–120.5°.

Analysis. Calculated for $\text{C}_{15}\text{H}_{19}\text{NO}_4$: C, 64.97; H, 6.91; N, 5.05. Found: C, 64.94; H, 6.85; N, 4.88.

NMR (d_6 -acetone) δ : Carboxylic, 9.20 (1H, s), aromatic 7.35 (5H, s), benzylic, 5.10 (2H, s), (CH₂)₂, 4.05–0.65 (1H, m), 3.5–2.9 (H, m), (CH₂)₂, 2.0–1.6 (6H, m), α -CH₃, 1.50 (3H, s).

I.R. (KBr): 3300–3100, 2945, 2875, 1755, 1670, 1505, 1480, 1425, 1360, 1280, 1240, 1210, 1185, 1145, 1090, 1035, 990, 915, 885, 780, 750 and 710 cm^{-1} .

(+)-N-carbobenzoxy-2-methyl-S-piperolic acid (18)

Compound **17** (28 g, 0.10 mol) and quinine (32.4 g, 0.10 mol) were dissolved in hot acetone (2.5 l). The hot solution was filtered and allowed to stand at 5° overnight. White fluffy crystals were collected and the filtrate saved for the preparation of **21**. After four recrystallizations from acetone the [(+A)·(–B)] salt was obtained: 17.0 g, 56% yield, m.p. 164–165° [$\alpha_D^{25} = -93.3^\circ$ (MeOH, $c = 1.5$ g/dl)]. The same values were obtained after two additional recrystallizations. This salt was dissolved in 100 ml chloroform and 100 ml aqueous NH_4OH (1 N) were added dropwise with stirring. The chloroform was evaporated under vacuum, quinine precipitated and removed from the aqueous layer by filtration. The filtrate was brought to pH 4 with 2 N HCl, the precipitated solid was extracted with chloroform. After drying and removing the solvent, white crystals were obtained and recrystallized from chloroform–hexane to give white needles: 7.6 g, 54% overall yield; m.p. 137.5–139°, [$\alpha_D^{25} = +25.4^\circ$ (MeOH, $c = 5$ g/dl)].

NMR and *i.r.* spectra are the same as for the racemic compound.

Analysis. Calculated for $\text{C}_{15}\text{H}_{19}\text{NO}_4$: C, 64.97; H, 6.91; N, 5.05. Found: C, 64.70; H, 6.73; N, 4.99.

(–)-N-carbobenzoxy-2-methyl-4-piperolic acid (21)

The [(–A)·(–B)] enriched filtrate obtained from the crystallization of (+)-N-carbobenzoxy-2-methyl-S-piperolic acid quinine salt [(+A)·(–B)] was evaporated to dryness (34.0 g). This salt was recrystallized six times from acetone at –5° to give the quinine salt of **21** (12.5 g, 41% yield). This salt was treated as described in the previous example to give white crystalline needles: 5.6 g, 40% overall yield; m.p. 137.5–138.5°, [$\alpha_D^{25} = -25.5^\circ$ (MeOH, $c = 5$ g/dl)].

Analysis. Calculated for $C_{15}H_{19}NO_4$: C, 64.97; H, 6.91; N, 5.05. Found: C, 64.95; H, 6.85; N, 4.88.

(-)-2-Methyl-S-pipecolic acid (20)

(a) *Hydrogen bromide method.* A solution of HBr in acetic acid (10 ml) was added dropwise to **18** (2.0 g, 7.2 mmol) with stirring. The mixture was allowed to react for 30 min at room temperature. The reaction mixture was evaporated to dryness and the residue triturated with ethyl ether. The product precipitated as fine white crystals and was recrystallized from a methanol-ethyl ether mixture to give *(-)-2-methyl-S-pipecolic acid hydrobromide salt (19)* (1.5 g, 93% yield); m.p. 262–265°, $[\alpha]_D^{25} = -5.25$ (MeOH, $c = 2$ g/dl).

This hydrobromide salt was dissolved in water and passed through a column of Amberlite i.r.-45. The residue obtained after rotary evaporation of the water was crystallized from methanol-ethyl ether to give free amino acid **20** (0.80 g, 85% yield); m.p. 350° (sublimation).

(b) *Catalytic hydrogenation method.* A solution of **18** (1.0 g, 3.6 mmol) in methanol (50 ml) and catalytic amounts of platinum oxide was hydrogenated at 45 psi. When hydrogen uptake ceased (8 hr) the solution was filtered, the filtrate was evaporated to dryness and the residue crystallized from methanol-ethyl ether to give colourless crystals (0.50 g, 98% yield); m.p. 350° (sublimation), $[\alpha]_D^{25} = -3.7$ (H_2O , $c = 2$ g/dl).

Analysis. Calculated for $C_7H_{13}NO_2$: C, 58.72; H, 9.15; N, 9.78. Found: C, 58.76; H, 9.19; N, 9.82.

NMR (D_2O) δ : (CH_3), 3.3–3.6 (2H, distorted t), (CH_2) $_{\beta,\gamma,\delta}$: 1.8–2.7 (6H, m), α - CH_3 , 1.7 (3H, s).

I.R. (KBr): 3100–2300, 1590–1620, 1450, 1395, 1360, 1295, 1290, 1210, 1180, 1115, 1060, 1045, 1000, 985, 875, 850 and 780 cm^{-1} .

(+)-2-Methyl-R-pipecolic acid (23)

Compound **23** was prepared in the same fashion as described for the *l*-rotatory isomer. *(+)-2-Methyl-R-pipecolic acid hydrobromide (22)* possessed the following properties: m.p. 261–263°, $[\alpha]_D^{25} = +5.1$ (MeOH, $c = 2$ g/dl). The free amino acid **23** had the following properties: m.p. 350° (sublimation), $[\alpha]_D^{25} + 5 \pm 2^\circ$ (H_2O , $c = 2$ g/dl).

Analysis. Calculated for $C_7H_{13}NO_2$: C, 58.72; H, 9.15; N, 9.78. Found: C, 58.75; H, 9.09; N, 9.80.

The NMR and i.r. spectra are identical to those of the *(-)-S*-isomer **20**.

Methyl 1-benzyl-2-oxocyclopentanecarboxylate (2c)

The procedure used for the preparation of methyl 1-methyl-2-oxocyclopentane was employed here. The benzylation of methyl 2-oxocyclopentane-carboxylate (250 g, 1.76 mol) gave, after distillation, pure product (330 g, 81% yield); b.p. 187°/23 mmHg.

Analysis. Calculated for $C_{14}H_{16}O_3$: C, 72.39; H, 6.94. Found: C, 72.34; H, 6.90.

NMR (neat) δ : 7.25 (5H, m); 3.65 (3H, s); benzylic $-CH_2-O-S$ AB pattern at 3.30 and 3.0 (2H, $J_{AB} = 14$ Hz); 2.6–1.5 (6H, m).

Methyl 2-benzyl-6-oxopipecolate (3c)

The procedure used for the preparation of **3a** was used except that methyl 2-oxocyclopentane-carboxylate was added dropwise to a solution of HN_3 and H_2SO_4 in chloroform at -10 over a period of 3 hr. Crystallization of the product from ethyl acetate-hexane gave colourless crystals in 79% yield; m.p. 111–113°.

Analysis. Calculated for $C_{14}H_{17}NO_3$: C, 68.00; H, 6.93; N, 5.66. Found: 67.89; H, 6.99; N, 5.69.

NMR ($CDCl_3$) δ : Aromatic 7.2 (5H, m), amide 6.5 (1H, s), methyl ester 3.65 (3H, s), benzylic AM pattern, $M = 3.25$, $A = 2.87$ (2H, $J_{AM} = 14$ Hz), (CH_2) $_{\beta,\gamma,\delta}$: 2.45–1.40 (6H, m).

I.R. (KBr): 3190, 3075, 2960, 1735, 1660, 1490, 1450, 1435, 1400, 1330–1300, 1265, 1240, 1190, 1095, 1010, 985 and 830–690 cm^{-1} .

Methyl 2-benzylpipecolate (6c)

The method of Borch [15] was used except that the reduction with sodium borohydride was carried out at 0° for 6 hr and continued at 25° for an additional 12 hr. Methyl 2-benzyl-6-oxopipecolate (130 g, 0.49 mol) was reduced to give **6c** (86.0, 70%), as a viscous oil (b.p. 114°/0.25 mmHg) which crystallized upon standing; m.p. 39–40°.

Analysis. Calculated for $C_{14}H_{19}NO_2$: C, 72.07; H, 8.21; N, 6.00. Found: C, 71.92; H, 8.00; N, 6.12.

NMR ($CDCl_3$) δ : Aromatic 7.20 (5H, m), 3.60 (3H, s), benzylic AB pattern 3.0–2.75 (2H, $J_{AB} = 12$ Hz), NH, 2.0 (1H, s), 2.40–1.20 (8H, m).

I.R. (neat): 3340, 3040, 2950, 2870, 1735, 1610, 1500, 1450, 1225–1175, 1135, 1100, 1020, 765 and 710 cm^{-1} .

2-Benzylpipecolic acid (8)

Compound **6c** (80 g, 0.34 mol) was suspended in 800 ml 1 N NaOH and refluxed for 2 hr. The clear solution was cooled to 70° and acidified (1 N HCl) with stirring using Congo red as an indicator; the free amino acid precipitated upon cooling to room temperature; it was collected by filtration and washed with excess water and dried. The free amino acid was insoluble in water or organic solvents (DMSO, MeOH) but was soluble in acidic or basic solution, yield 94%; m.p. 360° (dec).

Analysis. Calculated for $C_{13}H_{17}NO_2$: C, 71.20; H, 7.81; N, 6.39. Found: C, 71.92; H, 8.00; N, 6.12.

NMR (D_2O -DCl) δ : Aromatic 7.40 (5H, m), benzylic 3.23 (2H, m), (CH_2) $_{\gamma}$: 3.25 (2H, m), (CH_2) $_{\beta,\gamma,\delta}$: 2.65–1.50 (6H, m).

I.R.: 3450, 3110, 3030, 2970, 2930, 2860, 2500–2400, 1615, 1595, 1450, 1425, 1390, 1345, 1330, 1310, 1280, 1235, 1185, 1100, 870, 850, 750 and 710 cm^{-1} .

6-Methylpicolinic acid (9)

The method of Black, Depp and Corson [18] was used for the partial oxidation of 2,6-lutidine.

Ethyl 6-methylpicolinate (10)

A solution of **9** (200 g, 1.46 mol) in absolute ethanol (1000 ml) was saturated with HCl gas, and allowed to reflux for 2 hr. The solvent was evaporated, the residue was washed with cold aqueous potassium carbonate, and extracted with ether. The extracts were dried ($MgSO_4$), filtered, evaporated, and the ester was distilled under vacuum, yielding 217 g (90%) of pure colourless liquid; b.p. 125°/20 mmHg (lit. [17] 79–81°/0.25 mmHg).

NMR (neat) δ : Aromatic 7.95 (2H, m), 7.40 (1H, m), ethyl ester, 4.45 (2H, 9), 1.40 (3H, t), 6-methyl 2.60 (3H, s).

Methyl 6-methylpicolinate (12)

The esterification procedure described above was not successful, instead the methyl ester was obtained in 46% yield when thionyl chloride [17] was used; b.p. 62°/10 mm.

Analysis. Calculated for $C_8H_9NO_2$: C, 63.56; H, 6.00; N, 6.27. Found: C, 63.54; H, 6.15; N, 9.24.

NMR (neat) δ : Aromatic 7.85 (2H, m), 7.40 (1H, m), methyl ester 4.03 (3H, s), 6-methyl 2.62 (3H, s).

Ethyl cis-6-methylpipecolate (11)

The catalytic hydrogenation of **10** was carried out according to the literature [17]; b.p. 58–60°/10 mmHg (lit. [17] 99–100°/13 mm). The product was analyzed by gas chromatography with three different columns (Chrom W deg, Propak P, and $C_{18}A_w$ -DMCS); it was found that the ethyl ester is pure *cis* isomer.

NMR ($CDCl_3$) δ : Ethyl ester 4.10 (2H, 9), 1.15 (3H, t), (CH_2) $_{\gamma}$: 3.30 (1H, m), (CH) 2.60 (1H, m), NH 2.05 (1H, s), (CH_3) $_{\gamma}$: 1.05 (3H, d, $J = 6$ Hz), (CH_2) $_{\beta,\gamma,\delta}$: 1.0–2.0 (6H, m).

Methyl 6-methylpipercolates (13, 14)

Compound **12** (28 g, 0.185 mol), 400 ml methanol (containing 2.5% HCl gas), and platinum oxide (2.5 g) were mixed and hydrogenated at 40 psi at room temperature for 12 hr. The purification was carried out as described in the previous example. A mixture of methyl *cis*- and *trans*-6-methyl pipercolate was obtained (16 g, 55% yield); b.p. 47°/10 mmHg. The composition of this mixture was found by gas chromatography (using Chrom W OF-1) to be 87% *cis* and 13% *trans* with retention times of 1'30" and 2'20", respectively.

Analysis. Calculated for C₈H₁₅NO₂: C, 61.51; H, 9.03; N, 8.97. Found: C, 61.42; H, 9.06; N, 8.94.

NMR (neat) δ : Methyl ester 3.60 (3H, s), (CH)₂: 3.20 (1H, m), (CH)₁: 2.50 (1H, m), (CH₂) _{β,γ,δ} : 1.90–0.90 (6H, m), (CH₃)₁: 1.00 (*cis*, d, J = 6 Hz), 0.98 (*trans*, d, J = 6 Hz).

Cis-6-methylpipercolic acid hydrochloride (16)

A solution of **11** (20 g, 0.117 mol) in 100 ml 2 N HCl solution was refluxed for 2 hr. The solvent was evaporated to dryness and the residue was crystallized from methanol-ether (14 g, yield 56%), m.p. 262°.

Analysis. Calculated for C₇H₁₄ClNO₂: C, 46.80; H, 7.85; N, 7.80. Found: C, 46.74; H, 7.77; N, 7.82.

NMR (D₂O) δ : (CH)₂: 4.0 (1H, m), (CH)₁: 3.30 (1H, m), (CH₂) _{β,γ,δ} : 2.6–1.4 (5H, m), (CH₃)₁: 1.36 (3H, d, J = 6 Hz).

Cis-6-methylpipercolic acid (15)

Compound **11** (150 g, 0.88 mol) was suspended in water (1000 ml) and refluxed for 6 hr. Water was evaporated and the dry residue was crystallized from ethanol-ether, yielding 110 g (88%), m.p. 252–253° (dec).

Analysis. Calculated for C₇H₁₃NO₂: C, 58.72; H, 9.14; N, 9.78. Found: C, 58.77; H, 9.12; N, 9.78.

NMR (D₂O) δ : (CH)₂ and (CH)₁: 4.0–3.0 (2H, m), (CH₂) _{β,δ,γ} : 2.4–1.3 (6H, m), (CH₃)₁: 1.32 (3H, d, J = 6 Hz).

I.R. (KBr): 3450–2500, 1610, 1480, 1400, 1380, 1160, 1050, 1000, 770 and 690 cm⁻¹.

N-carbobenzoxy-cis-6-methylpipercolic acid (24)

A solution of **15** (10.0 g, 0.07 mol) in 17.5 ml 4 N NaOH was chilled to 0–5°. NaOH (21 ml, 4 N) and benzylchloroformate (13.0 g, 0.076 mol) were alternately added in portions over a period of 30 min with stirring. The reaction mixture was stirred for one additional hour, extracted with ether to remove excess acid chloride and the aqueous fraction was acidified carefully using Congo red as the indicator, with 5 N HCl and cooling in an ice bath. The separated oily substance was extracted with ethyl acetate and dried (MgSO₄). Ethyl acetate was removed and the oily residue was crystallized from ethyl acetate-hexane to give 13.0 g (67% yield) of white crystals; m.p. 80°.

Analysis. Calculated for C₁₅H₁₉NO₄: C, 64.96; H, 6.91; N, 5.05. Found: C, 64.92; H, 6.83; N, 4.98.

NMR (CDCl₃) δ : Aromatic protons, 7.40 (5H, s); benzylic protons, 5.20 (2H, s); CH₂: 4.90 (1H, m); CH₁: 4.40 (1H, m); (CH₂) _{β,γ,δ} : 1.50–2.20 (6H, m); CH₃: 1.2 (3H, d, J = 6 Hz); COOH 11.15 (1H, s).

N-(4-phenylbenzyloxycarbonyl)-cis-6-methylpipercolic acid (26)

Compound **25** was prepared from 4-biphenylmethanol and phosgene by a procedure similar to that for the preparation of benzyl chloroformate [20] (92% yield). A solution of **15** (26.0 g, 0.18 mol) in 45 ml 4 N NaOH was mixed with 200 ml of dioxane-water (50:50 by vol.). The mixture was cooled to 5°; 45 ml 4 N NaOH and a solution of 47.0 g (0.19 mol) of crude **25** in 60 ml dioxane were added alternately to the cooled reaction mixture with vigorous stirring. When the addition was completed (0.5 hr) the reaction mixture was stirred for an additional 3 hr, keeping the temperature between 0 and 5°. The suspension was

extracted 3 times with ethyl ether and once with ethyl acetate to remove the unreacted chloroformate derivative, dioxane and di-(4-phenylbenzyl)-carbonate formed as a side product. The aqueous layer was acidified with 5 N HCl to pH 4.0 and extracted with chloroform. The organic extracts were dried (MgSO₄); filtration and evaporation of the solvent afforded 47.5 g (74%) of a viscous oil. Several attempts to crystallize the oily product were not successful. The compound was used directly in the following step, since its NMR indicated high purity.

NMR (CDCl₃) δ : COOH 11.0 (1H, s); aromatic 7.7–7.2 (9H, m); ArCH₂: 5.2 (2H, s); CH₂: 4.9 (1H, m); CH₁: 4.4 (1H, m); (CH₂) _{β,γ,δ} : 2.3–1.4 (6H, m); CH₃: 1.20 (3H, d, J = 6 Hz).

(-)-N-(4-Phenylbenzyloxycarbonyl)-cis-6-methyl-S-pipercolic acid (27)

Compound **26** (48.0 g, 0.133 mol) and quinine (43.2 g, 0.133 mol) were dissolved in 500 ml methanol and refluxed for 30 min. Solvent was evaporated under aspirator pressure and the residue was dissolved in hot toluene (450 ml). The clear solution was slowly cooled to -5° and allowed to crystallize for 24 hr. A white crystalline salt (40.0 g) was collected by filtration and recrystallized seven times from a mixture of acetone-water (90:10, by volume). The quinine salt of the *l*-rotatory acid was thus obtained (12.0 g, 27% yield); m.p. 163–164°, [α]_D²⁵ = -110.7° (MeOH, c = 2 g/dl).

Analysis. Calculated for C₄₁H₄₇N₃O₆: C, 72.65; H, 6.99; N, 6.20. Found: C, 72.63; H, 6.96; N, 6.28.

An aqueous solution of NH₃ (100 ml 2 N) was added dropwise to a chloroform solution of the above quinine salt (11.0 g, 16 mmol in 100 ml CHCl₃) with cooling and stirring. The chloroform was evaporated; the aqueous solution was extracted with ether to remove the free quinine, acidified to a pH of 4.0 with 2 N HCl, and extracted with chloroform. The chloroform layer was dried (MgSO₄) and the solvent was evaporated. The foamy residue was crystallized by slow evaporation of chloroform-hexane solution to give 5.2 g (94%) of white crystals, m.p. 99–100°, [α]_D²⁵ = -31.9 (MeOH, c = 2 g/dl).

Analysis. Calculated for C₂₁H₂₃NO₄: C, 71.37; H, 6.56; N, 3.96. Found: C, 71.32; H, 6.67; N, 3.88.

The NMR spectrum was identical to that of the racemic compound.

I.R. (KBr): 3140, 3095, 3060, 3030, 2980, 2950, 1735, 1665, 1490, 1450, 1430, 1410, 1385, 1360, 1345, 1335, 1315, 1290, 1210, 1160, 1135, 1110, 1080, 1045, 1010, 970, 950, 870, 830, 800, 780, 760 and 700 cm⁻¹.

(-)-cis-6-methyl-S-pipercolic acid (28)

Compound **27** (1.1 g, 3.1 mmol) in 20 ml methanol was hydrogenated by a procedure similar to that for the preparation of 2-methyl-S-pipercolic acid. The free amino acid was crystallized from ethanol-ether, yielding 0.4 g (90%); m.p. 272–274° (dec), [α]_D²⁵ = -14.9° (MeOH, c = 1.5 g/dl).

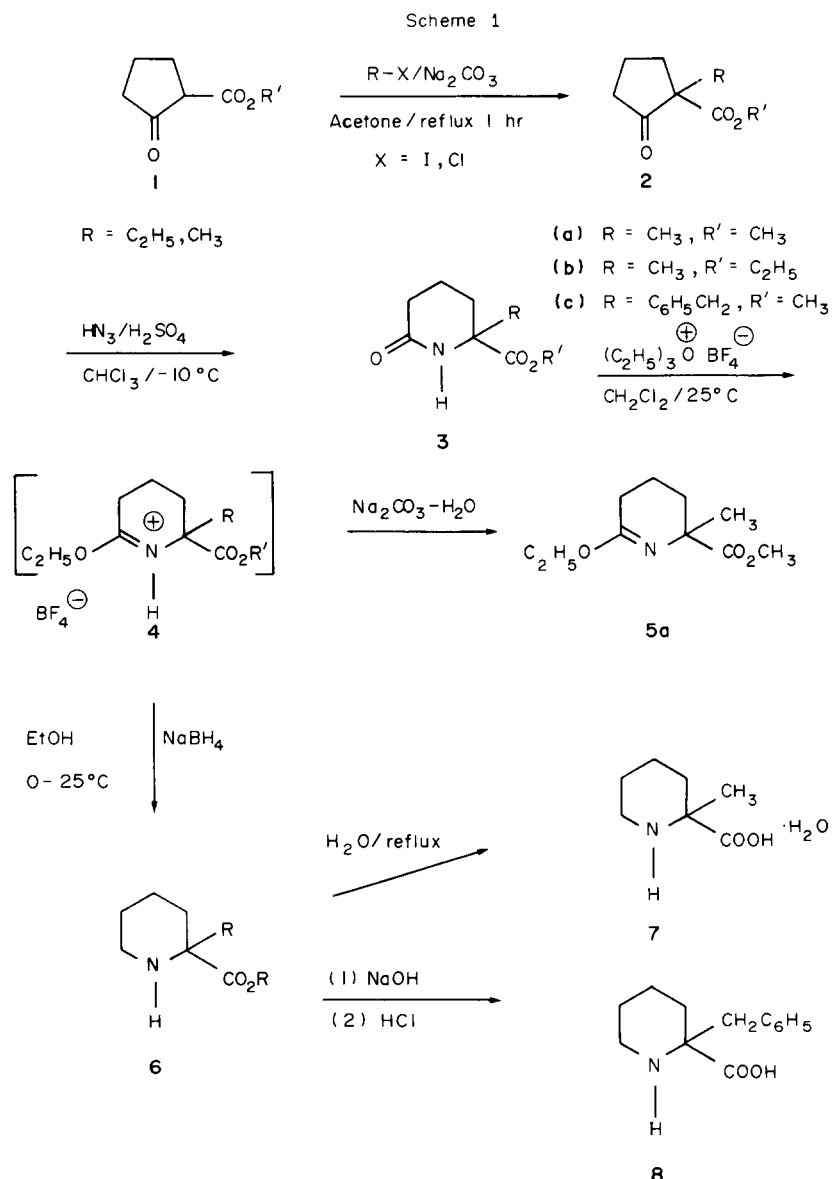
Analysis. Calculated for C₇H₁₃NO₂: C, 58.72; H, 9.15; N, 9.78. Found: C, 58.75; H, 9.09; N, 9.69.

RESULTS AND DISCUSSION

Syntheses of 2-benzyl- and 2-methylpipercolic acids

(-)-S-pipercolic acid is a naturally occurring amino acid present in the fruit of beans and other legumes [9]. Pipercolic acid was shown to be formed from lysine by plants and subsequently was implicated in lysine metabolism by mammals [10]. Various methods for its synthesis are detailed by Greenstein and Winitz [11]. Katchalski *et al.* [12] have reported on the properties of poly-S-pipercolic acid.

The only 2-alkylpipercolic acid reported previously is pipercolic acid [13]. A convenient and general



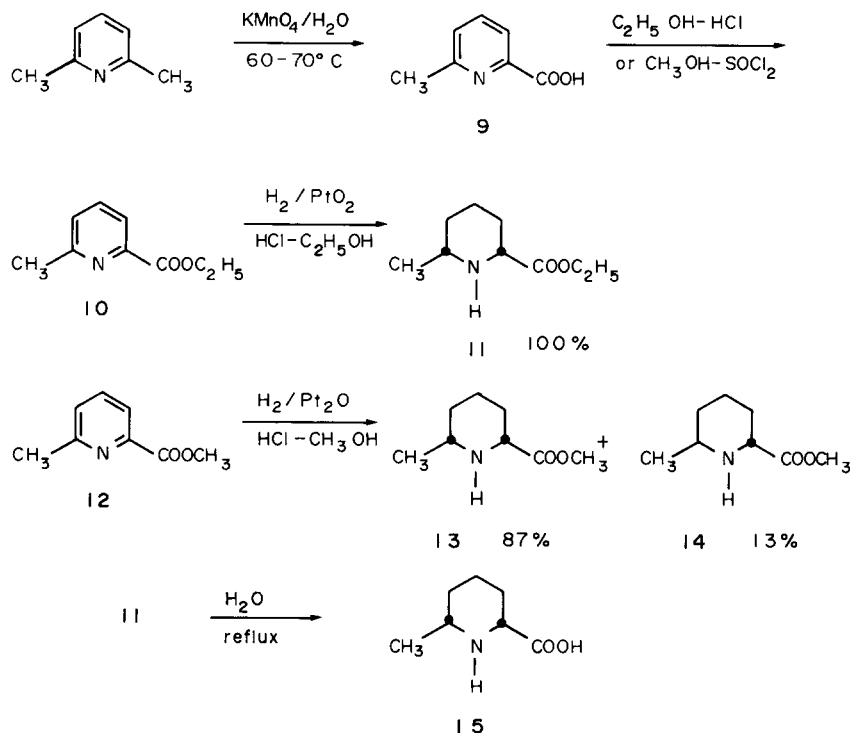
approach for the preparation of 2-alkylpiperelic acids is outlined in Scheme 1. Alkyl 2-oxocyclopentanecarboxylates (**1**) were alkylated *in situ* according to the method of Claisen as simplified by Barco *et al.* [14]. C-alkylation was quantitative and the yields of the distilled products were 81% for benzylation and 97% for methylation. A Schmidt reaction was utilized for ring enlargement, using conc. sulphuric acid as catalyst and 1.3 equivalents of hydrazoic acid in dry chloroform at -10° . These conditions gave oxopiperelate **3** in 67–79% yield. The selective reduction of the amide group in **3** is best accomplished by the method of Borch [15], using triethyloxonium tetrafluoroborate [16] directly to convert the secondary amide (**3**) to the iminoether fluoroborate salt (**4**). This was reduced directly with sodium borohydride to give alkyl 2-alkylpiperelate (**6**) in an overall yield of 67–73% from **3**. The free imino ether (**5a**) was obtained by neutralizing its salt (**4a**) with cold aqueous sodium carbonate. Hydrolysis

of **6a** or **6b** was accomplished in water at reflux; whereas, **6c** required base catalyzed hydrolysis. 2-Methylpiperelic acid (**7**) was obtained (94% yield) in its monohydrate form upon crystallization from 70% aqueous ethanol. 2-Benzylpiperelic acid (**8**) was obtained upon acidifying its sodium salt solution. The free amino acid **8** is insoluble in water and precipitated from the solution as a white solid. It is soluble only in water at high or low pHs. 2-Methylpiperelic acid is soluble in water, hot 70% aqueous ethanol, and hot DMSO, slightly soluble in hot ethanol and insoluble in chloroform, acetone or cold ethanol.

Synthesis of *cis*-6-methylpiperelic acid

6-Methylpiperelic acid has previously been synthesized but no indication was given about its stereochemistry [17]. 2,6-Lutidine was partially oxidized (see Scheme 2) to give 6-methylpicolinic acid (**9**) according to the literature [18]. Ethyl 6-methylpico-

Scheme 2



linatate (10) was prepared in 90% yield by heating at reflux an ethanolic solution of compound 9 saturated with gaseous hydrogen chloride. The methyl ester 12 was obtained by reaction with thionyl chloride [17]. Catalytic hydrogenation of compounds 10 and 12 with platinum oxide in ethanol, saturated with hydrogen chloride [17], proceeded quantitatively. Reduction of the ethyl ester 10 gave pure ethyl 6-*cis*-methylpipercolate (11), whereas the methyl ester 12 upon hydrogenation afforded a mixture of 87% *cis* isomer 13 and 13% *trans* isomer 14, as determined by NMR and gas chromatography. Hydrolysis of 11 yielded pure *cis*-6-methylpipercolic acid (15) upon heating with water at reflux (80% yield). Acidic hydrolysis of 11 gave the hydrochloride salt of 15 in 56% yield.

Resolution of 2-methylpipercolic acid (2MPA)

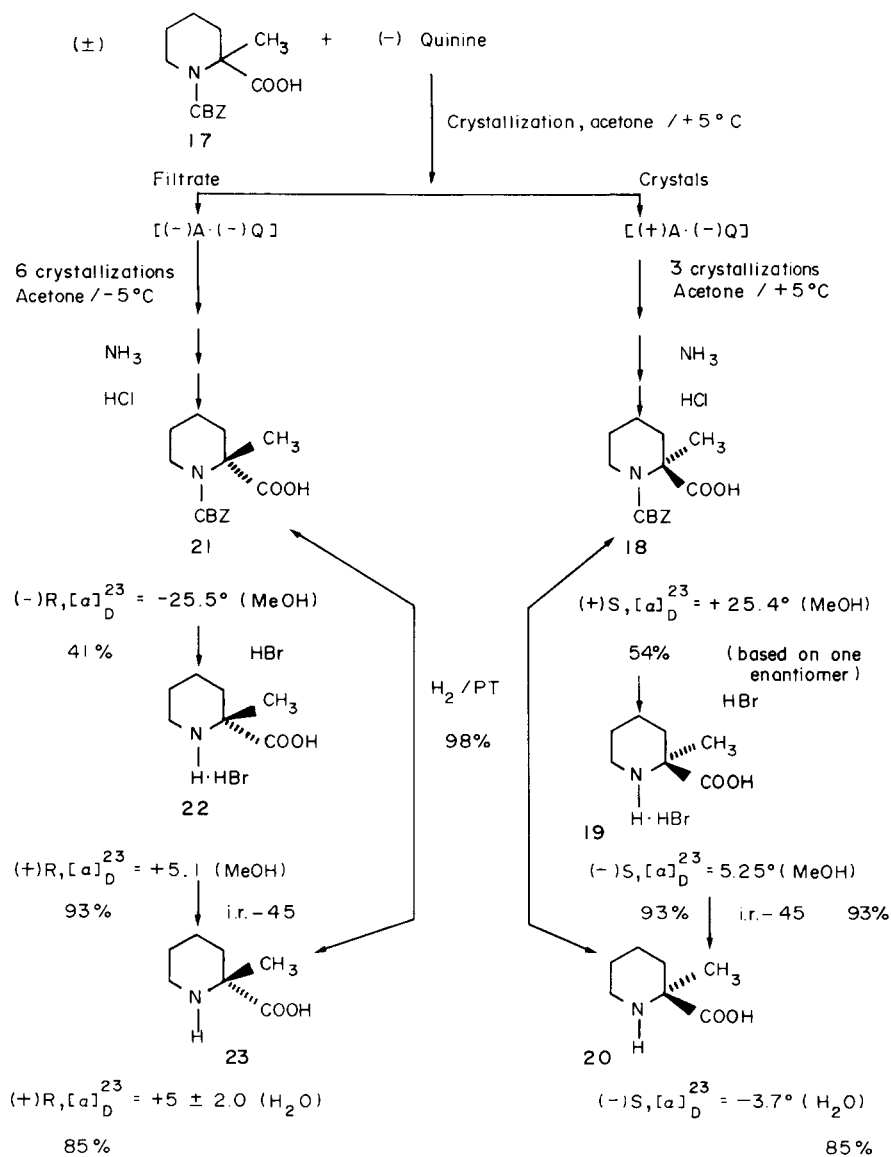
The resolution of pipercolic acid was achieved by Mende [19] in 1896 through fractional crystallization of its salt with *l* and *d*-tartaric acid. This method was not successful for 2-methylpipercolic acid. However, fractional recrystallization of the quinine salt of the *N*-carbobenzyloxy derivative [3] proved successful. Reaction of 2MPA with an excess of benzyl chloroformate yielded *N*-carbobenzyloxy-2-methylpipercolic acid (17) in addition to the corresponding acid anhydride (approx. 1:1). Consequently, the reaction mixture was allowed to reflux with water in order to hydrolyze the anhydride, resulting in a 68% yield of pure 17. Resolution then proceeded according to Scheme 3. The quinine salt of 17 was prepared and

crystallized from acetone at $+5^\circ$. The less soluble salt [(+)-A·(-)Q] was crystallized and purified further by three recrystallizations from acetone at $+5^\circ$. The free acid was liberated by treatment with ammonia with subsequent acidification. (+)-*S*-*N*-carbobenzyloxy-2-methylpipercolic acid (18) was obtained in 54% overall yield, based on one enantiomer, $[\alpha]_D^{23} +25.4^\circ$ (MeOH). The carbobenzyloxy group was removed either by catalytic hydrogenation or by acidic cleavage with hydrogen bromide. (-)-*S*-2-methylpipercolic acid hydrobromide (19) and the free amino acid were obtained with $[\alpha]_D^{23} -5.25^\circ$ (MeOH) and -3.7° (H_2O), respectively. The filtrate of the initial crystallization of the diastereomeric salts was determined to be enriched with the [(-)-A·(-)Q] salt. It was evaporated to dryness and recrystallized six times at -5° from acetone. Dissociation of this salt with ammonia followed by acidification produced (-)-*R*-*N*-carbobenzyloxy-2-methylpipercolic acid (21) in 41% of the theoretical yield for one enantiomer; $[\alpha]_D^{23} -25.5^\circ$ (MeOH). (+)-*R*-2-methylpipercolic acid hydrobromide (22) and the free amino acid (23) had $[\alpha]_D^{23} +5.1^\circ$ (MeOH) and $5 \pm 2^\circ$ (H_2O), respectively. The specific rotations of all enantiomeric compounds in Scheme 3 are of comparable values with opposite signs of rotation, indicating high efficiency of the resolution.

Resolution of *cis*-6-methylpipercolic acid

We have made several attempts to resolve 6-methylpipercolic acid or its ethyl ester by means of fractional crystallization of the corresponding di-

Scheme 3



astereomeric salts but none of these attempts were successful. Several diastereoisomeric salts were prepared by treating *N*-carbobenzyloxy-*cis*-6-methylpipercolic acid with an optically active resolving agent such as tyrosine hydrazone, ephedrine, quinine or brucine. Several solvents or solvent combinations with variable polarities were used in an attempt to crystallize these salts, but unfortunately not one of these was crystallizable. In most cases the salt separated from its solution as an oil upon cooling.

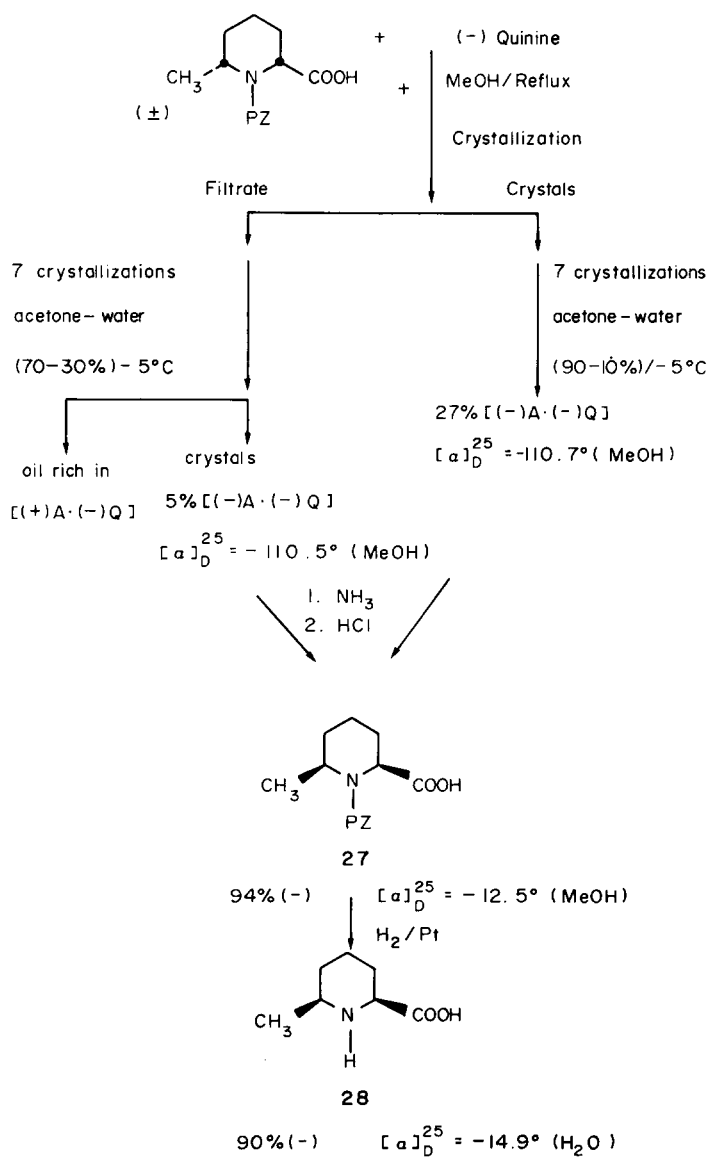
Eventually, we decided on 4-phenylbenzyloxycarbonyl as protecting group, due to its enhanced proclivity towards crystallization. 4-Phenylbenzyl chloroformate (**25**) was prepared in 92% yield from the reaction of phosgene with 4-biphenylmethanol by a procedure similar to that for the preparation of benzyl chloroformate [20]. Compound **25** was reacted with *c*6MPA in a dioxane-water (1:1) mixture. Dioxane was used as co-solvent in order to dissolve the

solid chloroformate derivative **23**. *N*-(4-phenylbenzyloxycarbonyl)-*cis*-6-methylpipercolic acid (**26**) was obtained as a viscous oil in 74% yield.

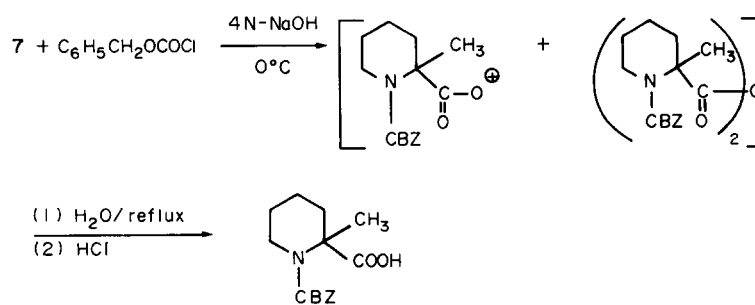
The quinine salt of compound **26** was formed in methanol (Scheme 4). It could be fractionally crystallized from toluene at -5°. The crystals were recrystallized seven times from a mixture of acetone-water (9:1, v/v) at -5°, to give the quinine salt of the *l*-rotatory acid [(−)A·(−)Q] in 27% yield based on one enantiomer, [α]_D²⁵ = -110.7° (MeOH). Treating this salt with ammonia followed by acidification gave the *l*-rotatory acid (**27**) in 94% yield, [α]_D²³ = -12.5° (MeOH). The removal of the protecting group by catalytic hydrogenation afforded (−)-*cis*-6-methyl-*S*-pipercolic acid (**28**) in 90% yield, [α]_D²⁵ = -14.9° (H₂O).

The filtrate retained from the first crystallization in toluene was evaporated to dryness. The residue contained the quinine salts of the *d*-rotatory acid and

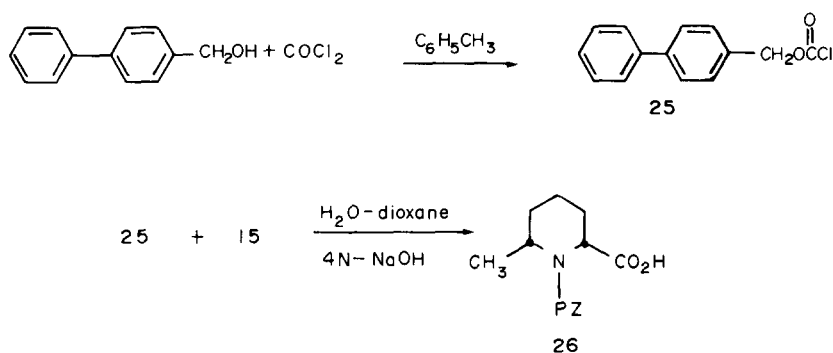
Scheme 4



Scheme 5



Scheme 6



some of the *l*-rotatory acid. An attempt to crystallize this residue from a mixture of acetone–water (7:3, v/v) gave only the minor isomer as a crystalline salt in 5% yield. The quinine salt of the *d*-rotatory acid separated as an oil from the hot acetone–water solution upon cooling to room temperature. No further attempt was made to purify the *d*-rotatory acid.

Absolute configurations

Optical rotatory dispersion (ORD) and circular dichroism (CD) can be used for a rapid assignment

of the absolute configuration of α -amino acids [2, 21–23]. All naturally-occurring and most synthetic α -amino acids show positive Cotton effects around 200 nm in water and 208–210 in acid.

Figure 1 shows the CD spectra of (–)-*S*-pipecolic acid (S-PA) (–)-*c*6MPA, (–)-2MPA, (–)-*S*-proline (S-P) and (–)-2-methyl-*S*-proline (2M-S-P) in water and in acid. The CD data are given in Table 1 as measured from Fig. 1. S-PA and (–)-*c*6MPA each exhibits a single positive Cotton effect in water and in acid around 207–208 nm. The similarity of their CD curves suggests that both compounds possess the

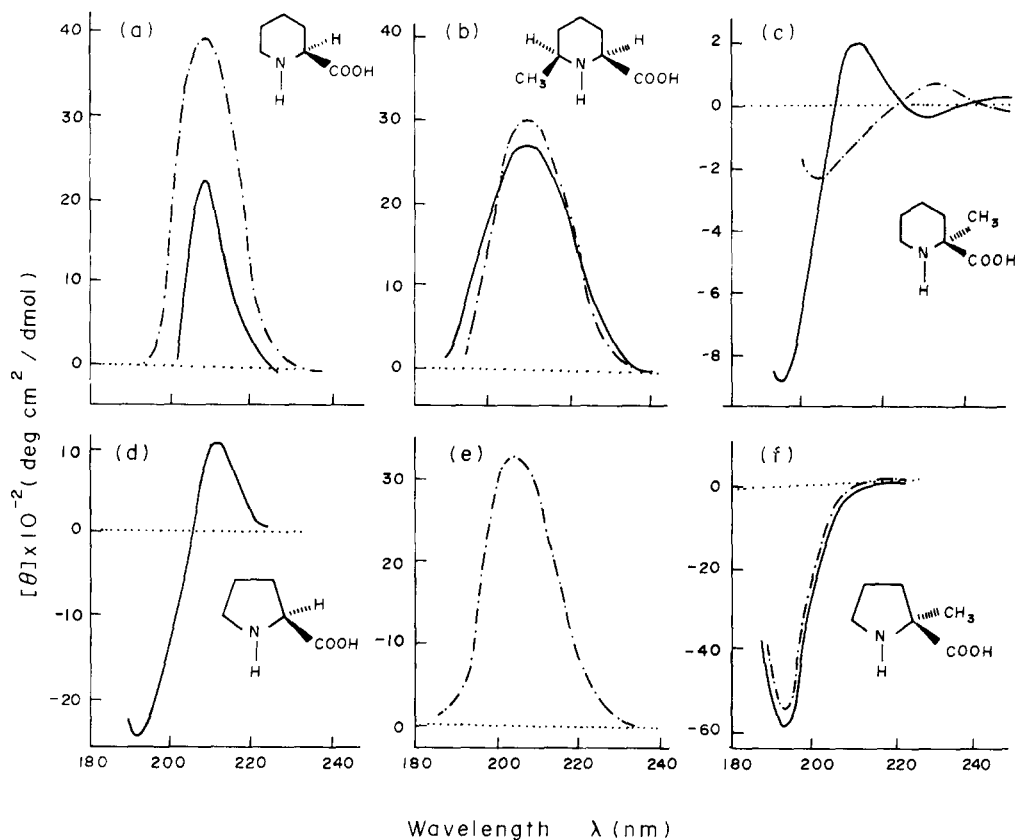


Fig. 1. CD Spectra of (a) (–)-*S*-pipecolic acid; (b) (–)-*cis*-6-methyl-*S*-pipecolic acid; (c) (–)-2-methyl-*S*-pipecolic acid; (d) and (e) (–)-*S*-proline; (f) (–)-2-methyl-*S*-proline in water (—) and in 1N HCl solution (---).

Table 1. CD data of pipecolic acid, proline and related imino acids

Compound	H ₂ O		HCl	
	$[\theta] \times 10^{-2}$ (deg cm ² /dmol)	(nm)	$[\theta] \times 10^{-2}$ (deg cm ² /dmol)	(nm)
(-)-S-pipecolic acid	+21.20	207	+39.30	208
(-)-Cis-6-methyl-S-pipecolic acid	+27.90	207	+30.10	207
(-)-2-Methyl-S-pipecolic acid	-0.60	230	+0.50	231
	+1.85	212		
	-8.85	195	-2.30	202
(-)-S-proline	+9.80	211	+31.00	207
	-24.60	193	—	—
(-)-2-Methyl-S-proline	-58.50	195	-55.00	195

S configuration. The CD spectrum of (-)2MPA in water shows three Cotton effects, a negative one at 195 nm, a positive one at 212 nm and another negative one at 230 nm, whereas the CD spectrum in acid shows only negative and positive Cotton effects at 202 and 231 nm.

Two Cotton effects have been observed in water for S-azetidine-2-carboxylic acid, S-proline and 4-methyl-S-proline [23] and three Cotton effects were reported for the CD spectrum of S-proline methyl ester [24]. More than one Cotton effect may arise from different conformations of the molecules [25], from different states of solvation [26], from the existence of "allowed" and "forbidden" transitions of closely similar energy levels [27], or from a combination of these factors [23]. Craig and Pereira [24] suggest that the three Cotton effects observed for proline methyl ester may be due to an optically active $n \rightarrow \sigma^*$ transition of the nitrogen for the positive CD band at 202 nm, $n \rightarrow \pi^*$ transition of the ester carbonyl group for the positive CD band at 209 nm, and an interaction through space between the non-bonding orbital of the nitrogen atom and the chromophoric transition of the carbonyl group for the negative weak CD band at 232 nm. However, only one Cotton effect was observed in the CD spectra of the above compounds in acidic solutions around 208–212 nm [23, 24].

The S-configuration can be assigned to (-)2MPA based on the similarity of its CD spectra in water with that of proline and other related amino acids [23]. However, we have as yet no satisfactory explanation for the abnormal CD spectrum in acid of (-)2MPA.

The sector rule of Jorgensen [28], which relates the sign and amplitude of the Cotton effect to the conformation and absolute configuration of α -amino acids, can be applied here. His sector diagram is being referred to in the following discussion. The slightly higher positive rotation of pipecolic acid over proline can be attributed to the different geometry of the six-membered ring *versus* the five-membered ring. The C _{β} and C _{γ} methylene groups in pipecolic acid are closer to the positive and negative sectors, respectively, than the corresponding C _{β} and C _{γ} methylene groups in proline. Therefore, the net effect is a larger positive contribution. The Cotton effect of (-)6MPA is more positive than that of S-PA in water; the methyl group is residing in the null sector as determined from an examination of the molecular model, thus creating a more positive value by decreasing the negative contribution to the net rotation.

Similar results have been found for *cis*-5-methylproline [2].

The sector rule of Jorgensen does not provide a satisfactory explanation for the observed CD spectra of (-)2MPA or (-)S-2MP. The CD spectrum of the latter shows a single negative Cotton effect at 195 nm. The absolute configuration of (-)2MP was assigned the S-configuration [3] based on the method of Lutz-Jirgensons [29]. However, the sector rule predicts opposite configuration for (-)2MP, namely the R-configuration. In the case of (-)2MPA the sector rule predicts a single positive Cotton effect with higher amplitude than that of S-PA because of the presence of an α -methyl group in or nearby the positive sector. The experimental result does not agree with the prediction of the sector rule. Perhaps this deviation is due to a change in the conformation of the molecules by substitution at the 2-position and/or a change in the curvatures of the sectors. We are currently attempting to determine the absolute configurations of (-)2MPA and (-)2MP by chemical means in order to resolve this problem.

At present we are inclined to assign the S-configuration for the *l*-rotatory amino acids based on the above results and on the CD curves of their cyclic dimers [8] or polymer [3].

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Dedication—The senior author (C. G. Overberger) dedicates this paper to Professor Oto Wichterle for his enormous contribution to macromolecular science, and in honour of his 70th birthday.

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