The Stereoselective Synthesis of *threo*-3-Hydroxy-4-amino Acids

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Two *threo*-3-hydroxy-4-amino acids, *dl*-4-amino-3-hydroxy-6-methylheptanoic acid (**1**) and *dl*-4-amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic acid (**2**), the optically active forms of which had been isolated by the acid hydrolysis of some antibiotics, were synthesized stereoselectively from L-leucine and DL-3-(3-pyridyl)-alanine respectively through 2-pyrrolidinone intermediates.

Two unusual *threo*-type hydroxy amino acids, (3*S*,4*S*)-4-amino-3-hydroxy-6-methylheptanoic acid (**1**) and (2*R*,3*S*,4*S*)-4-amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic acid (**2**), have been isolated by the acid hydrolysis of various pepstatins[1a-e] and pyridomycin[2] respectively. The first synthesis of **1** was carried out by Morishima *et al.* in a *non*-stereoselective way.[3] Kinoshita *et al.* recently synthesized **1** and **2** in a highly stereospecific manner starting from the appropriate optically active deoxy-sugar derivatives.[4,5]

The present authors have now attempted the synthesis of this type of hydroxy amino acid according to a general plan starting from common amino acids and involving the stereoselective reduction of 4-hydroxy-3-pyrrolin-2-one intermediates, such as **6** (Scheme 1) or **13** (Scheme 2). The *dl*-**1** could be synthesized smoothly along those lines. In the case of **2**, the reduction of the 4-hydroxy-3-pyrrolin-2-one (**13**) proceeded in the desired manner, but the subsequent steps led to a mixture of two *dl*-hydroxy amino acids, epimeric at the C-2 carbon atom. The major (90%) *non*-natural-type isomer (**23**) could easily be obtained in a pure form, but the isolation of the minor (10%) natural-type isomer (**2**) was so tedious that no crystalline sample could be obtained.

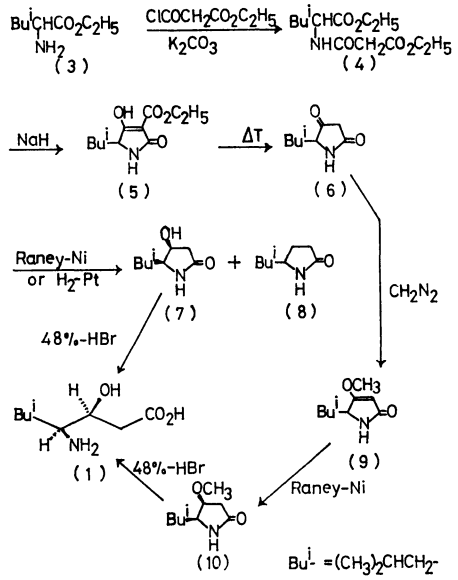
Results and Discussion

dl-*4-Amino-3-hydroxy-6-methylheptanoic Acid (1) (Scheme 1).* The *N*-(ethoxycarbonylacetyl)leucine ethyl ester (**4**) derived from **3** was subjected to Dieckmann condensation in a usual way[6] to give 3-ethoxycarbonyl-4-hydroxy-5-isobutyl-3-pyrrolin-2-one (**5**), which was then decarboxylated to 5-isobutyl-2,4-pyrrolidinedione (**6**) in an 87.7% overall yield from **4**. The pyrrolidinedione (**6**) thus obtained was a racemate, and it was used without resolution.

When **6** was hydrogenated with Raney nickel, it gave a mixture of 4-hydroxy-5-isobutyl-2-pyrrolidinone (**7**) and 5-isobutyl-2-pyrrolidinone (**8**). Compound (**7**) could be separated from **8** by crystallization and was proved, from the results of the subsequent hydrolysis, to be the *cis*-compound with regard to the two substituents at C-4 and C-5.

The hydrogenation of **6** in the presence of platinum oxide in ethyl acetate gave mostly **7**, but in a mixture of ethanol and acetic acid, it led to the hydrogenolyzed product (**8**) quantitatively. Alternatively, **6** was converted into 4-methoxy-5-isobutyl-3-pyrrolin-2-one (**9**) by diazomethane and was hydrogenated with Raney nickel to give *cis*-4-methoxy-5-isobutyl-2-pyrrolidinone (**10**) in an 87% yield.

The hydrolysis of **7** or **10** with 48% hydrobromic acid, followed by chromatographic purification on a Dowex 50 column, gave the *dl*-**1**[7] in a fair yield (74% from **10**). The NMR spectrum of synthetic *dl*-**1** agreed with that of the natural specimen.[3]



Scheme 1.

dl-*4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic Acid (2) (Scheme 2).* DL-3-(3-Pyridyl)alanine[8] was converted to its methyl ester (**11**), which was then condensed with 2-ethoxycarbonylpropionyl chloride. The *N*-(2-ethoxycarbonylpropionyl)-3-(3-pyridyl)alanine methyl ester (**12**) thus obtained was subjected to Dieckmann condensation with sodium methoxide to afford 4-hydroxy-3-methyl-5-(3-pyridylmethyl)-3-pyrrolin-2-one (**13**) in a 32% overall yield from pyridylalanine.

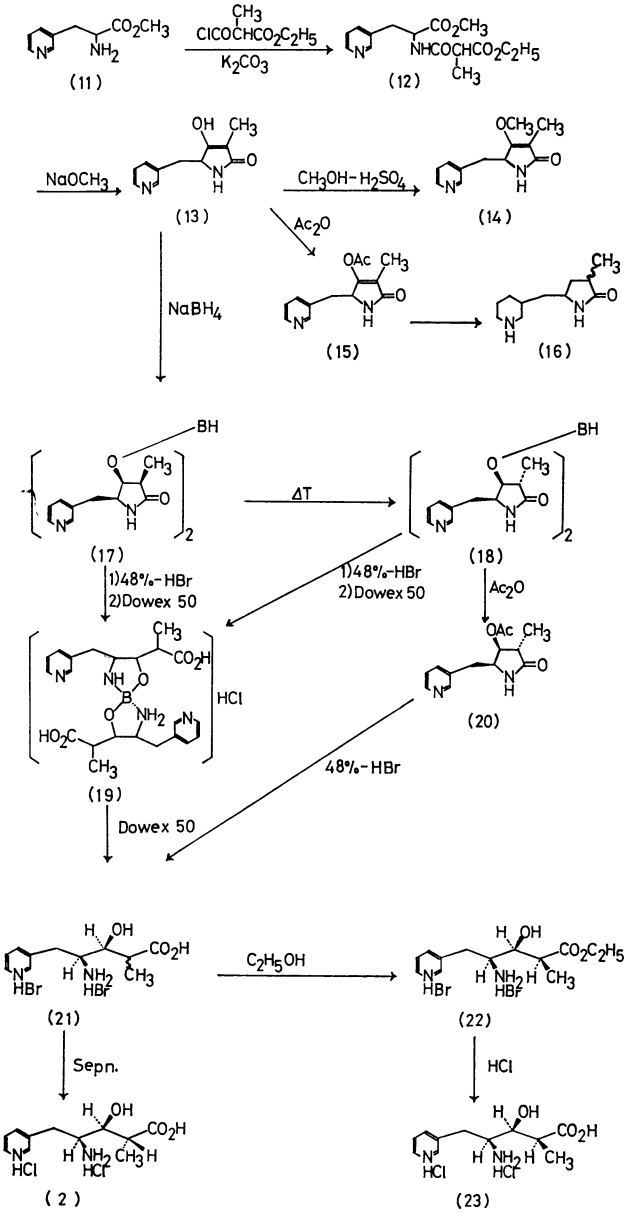
The catalytic hydrogenation of **13**, its methyl ether (**14**), or its acetate (**15**) was, however, unsuccessful because of the hydrogenolysis of the *O*-function at C-4 and/or of the reduction of the pyridine nucleus, leading to the formation of **16**, for example. Compound (**13**) was then reduced with sodium borohydride in 2-propanol to give, stereoselectively, a single compound, bis[4-methyl-5-oxo-2-(3-pyridylmethyl)-3-pyrrolidinyl]-boronate, which was considered to have the desired *cis*-configuration, as represented by **17**. Unfortunately, the compound was unstable to heat, and, on recrystallization from a mixture of chloroform and ethyl acetate, it isomerized into the more stable isomer (**18**).

The compound (**17** or **18**) was hydrolyzed with

48% hydrobromic acid at 50 °C. The product obtained was a mixture of the boron complexes of the two isomeric hydroxy amino acids with the composition represented by **19**.

The elimination of boric acid from **19** was effected by treatment with 48% hydrobromic acid, followed by ion-exchange chromatography, thus giving a mixture of two racemic hydroxy amino acids (**21**, a mixture of **2** and **23**, 30% from **13**). The isomer with the configuration corresponding to the natural amino acid (**2**) in the mixture was the minor component (10% from NMR). Alternatively, boron was removed by the acetylation of **18** with acetic anhydride and pyridine, and the acetate (**20**) was hydrolyzed by hydrobromic acid to give **21** with the same isomeric ratio as above in a 40% overall yield from **13**.

The *non*-natural-type major isomer (**23**) could be easily separated from the minor counterpart by the recrystallization of the mixture from ethanol, whereupon



Scheme 2.

the isomer crystallized out as its ethyl ester (**22**);[9] the hydrolysis of the ester gave pure **23**. The isolation of the natural-type isomer (**2**) was, however, very tedious. The above hydroxy amino acid mixture (**21**) was repeatedly chromatographed on the Dowex-50 column, and finally a small amount of an almost pure sample was obtained in the form of a *non*-crystalline concentrate, which gave the same NMR spectrum as that of authentic natural amino acid (**2**).

Experimental

The melting points are uncorrected. The IR spectra were taken on a Hitachi R-215 spectrometer. The NMR spectra were recorded on Hitachi R-20B and Hitachi R-22 spectrometers.

*N-(Ethoxycarbonylacetyl)leucine Ethyl Ester (**4**), 3-Ethoxycarbonyl-4-hydroxy-5-isobutyl-3-pyrrolin-2-one (**5**), and 5-Isobutyl-2,4-pyrrolidinedione (**6**).* Compounds (**4** and **5**) were prepared from the L-leucine ethyl ester in a way similar to that described by Achiwa and Yamada for the preparation of 5-ethyl-5-methyl-2,4-pyrrolidinedione.[6] **4** (60% from **3**); bp 145 °C/2 mmHg. **5** (87% from **4**), NMR (CD_3OD): δ 0.96(d, 6H), 1.31(t, 3H), 1.40—2.05 (m, 3H), 4.08(t, 1H), 4.3(q, 2H). The recrystallization of **5** from a mixture of ethyl acetate and ligroin gave **6** as colorless needles. Mp 124 °C. IR: ν_{max} 3180, 2960, 1765, 1680, 1384, 1290, 1240, 853, 800, 682 cm^{-1}. NMR ($CDCl_3$):[10] δ 0.96(d, 6H), 1.45—2.10(m, 3H), 3.03(s, 2H), 4.06(m, 1H), 7.85 (broad s, 1H). Found: C, 61.57; H, 8.42; N, 8.86%. Calcd for $C_8H_{13}NO_2$: C, 61.91; H, 8.44; N, 9.03%.

*4,5-cis-4-Hydroxy-5-isobutyl-2-pyrrolidinone (**7**).* a) A mixture of **6** (97 mg), excess Raney nickel, and ethanol (10 ml) was refluxed for 3 h. The Raney nickel was then removed by filtration, and the solvent was removed *in vacuo* to give a colorless solid. Subsequent recrystallization from a mixture of dichloromethane and hexane gave **7** as colorless prisms (48 mg, 48%). Mp 131—132 °C. NMR ($CDCl_3$): δ 0.96(d, 6H), 1.58(m, 3H), 2.51(m, 2H), 3.70 (m, 1H), 4.40(m, 1H), 6.28(broad s, 1H). Found: C, 60.95; H, 9.62; N, 8.76%. Calcd for $C_8H_{15}NO_2$: C, 61.12; H, 9.62; N, 8.91%.

b) Compound (**6**, 32 mg) was dissolved in ethyl acetate (20 ml) and hydrogenated over the platinum oxide catalyst. After the removal of the catalyst, the solution was concentrated and the residue was crystallized from a mixture of dichloromethane and hexane to give **7** as colorless prisms (31 mg, 94%).

*5-Isobutyl-2-pyrrolidinone (**8**).* Compound (**6**, 29 mg) was dissolved in a mixture of ethanol(9 ml) and acetic acid (1 ml) and then hydrogenated over the platinum oxide catalyst. After the removal of the catalyst and the solvent, the residue was sublimed at 80 °C/20 mmHg to afford **8** as colorless needles (26 mg, quantitative). Mp 74.5—75°C. NMR ($CDCl_3$): δ 0.94 (d, 6H), 1.29—1.85 (m, 3H), 2.18—2.52(m, 4H), 3.70(m, 1H), 6.60(1H). Found: C, 67.88; H, 10.65; N, 9.83%. Calcd for $C_8H_{15}NO$: C, 68.04; H, 10.71; N, 9.92%.

*4-Methoxy-5-isobutyl-3-pyrrolin-2-one (**9**) and 4,5-cis-4-Methoxy-5-isobutyl-2-pyrrolidinone (**10**).* The ethereal diazomethane was added to a solution of **6** in methanol at 0 °C. After the removal of the excess diazomethane and the solvent, **9** was obtained as colorless needles quantitatively. NMR ($CDCl_3$): δ 0.96(d, 6H), 1.32—1.95 (m, 3H), 3.78(s, 3H), 4.05 (m, 1H), 5.00 (d, 1H), 6.95 (broad s, 1H). The ether (**9**) was used without purification in the following reaction. A mixture of **9** (139 mg) and excess Raney nickel

in ethanol was refluxed for 5 h. The mixture was then filtered and concentrated under reduced pressure. The residue was recrystallized from a mixture of dichloromethane and hexane to give **10** as colorless needles (120 mg, 86%). Mp 87—88 °C. NMR ($CDCl_3$): δ 0.95(6H), 1.35—1.70 (m, 3H), 2.44 (d, 2H), 3.32 (s, 3H), 3.69—4.10 (m, 2H), 7.35(broad s, 1H). Found: C, 62.97; H, 10.03; N, 8.04%. Calcd for $C_9H_{17}NO_2$: C, 63.13; H, 10.00; N, 8.18%.

dl-*4-Amino-3-hydroxy-6-methylheptanoic Acid (1).*

a) A mixture of **10** (60 mg) and 48% hydrobromic acid (5 ml) was stirred for 10 h at 78 °C. The mixture was then concentrated to dryness under reduced pressure, redissolved in water (5 ml), and applied to a column of Dowex 50(0.8 cm × 25 cm). The column was washed with water until the eluent became neutral, and then the amino acid was eluted with 2 M-ammonia. The eluate was concentrated under reduced pressure, and the residue was recrystallized from a mixture of methanol and ethanol to afford *dl*-**1** as colorless crystals (45 mg, 74%). Mp 211.5—213.5 °C (dec). NMR (D_2O): δ 0.98 (d, 6H), 1.38—1.72 (m, 3H), 2.44—2.62(m, 2H), 3.13—3.47(m, 1H), 3.88—4.22(m, 1H). The NMR spectrum ($J_{3,4}$=5.5 Hz) of *dl*-**1** agreed with that ($J_{3,4}$=5.5 Hz)[3] of natural amino acid. Found: C, 54.66; H, 9.68; N, 7.99%. Calcd for $C_8H_{17}NO_3$: C, 54.83; H, 9.68; N, 7.99%.

b) By the same procedure as was used in the hydrolysis of **10**, **7** was hydrolyzed to give *dl*-**1** (80%).

N-*(2-Ethoxycarbonylpropionyl)-3-(3-pyridyl)alanine Methyl Ester (12).* By the same procedure as was described for the preparation of **4**, **11** (2.8 g) was *N*-acylated with 2-ethoxycarbonylpropionyl chloride. The crude product was purified by chromatography (silica gel, ethyl acetate) to give **12** (3.99 g, 88%). It was then recrystallized from diisopropyl ether. Colorless needles; mp 70.5—71.5 °C. IR: ν_{max} 3300, 3100, 3000, 2940, 1740, 1640, 1550, 1200, 1100, 1045, 1000, 960, 830, 810, 770, 715 cm^{-1}. NMR ($CDCl_3$): δ 1.22(t, 3H), 1.35(d, 3H), 3.05—3.43(m, 3H), 3.72(s, 3H), 4.14(q, 2H), 4.87(m, 1H), 7.15—7.70 and 8.30—8.60(m, 4H), 9.42(brosd s, 1H). Found: C, 58.43; H, 6.67; N, 8.80%. Calcd for $C_{15}H_{20}N_2O_5$: C, 58.43; H, 6.54; N, 9.09%.

4-Hydroxy-3-methyl-5-(3-pyridylmethyl)-3-pyrrolin-2-one (13). Sodium (3 g) was dissolved in absolute methanol (100 ml). To this solution, **12** (3 g) was added, after which the whole mixture was refluxed for 10 h. After the removal of the solvent under reduced pressure, the residue was dissolved in water (10 ml) and applied to a column of Dowex 50 (3 cm × 40 cm). After the column had then been washed with water, the **13** was eluted with 2 M-ammonia. Fractions containing **13** were concentrated, and the residue was recrystallized from methanol to give **13** as colorless prisms (1.1 g, 41%). Mp 172—174 °C. IR: ν_{max} 1665, 1600, 1580, 1490, 1225, 760, 700 cm^{-1}. NMR (CD_3SOCD_3): δ 1.39(s, 3H), 2.87—3.05(m, 2H), 4.13(m, 1H), 7.24—7.75 and 8.32—8.55 (m, 4H). Found: C, 64.52; H, 5.90; N, 14.10%. Calcd for $C_{11}H_{12}N_2O_2$: C, 64.69; H, 5.92; N, 13.72%.

4-Methoxy-3-methyl-5-(3-pyridylmethyl)-3-pyrrolin-2-one (14). A mixture of **13** (67 mg) and concentrated sulfuric acid (0.1 ml) in anhydrous methanol was refluxed for 30 h. This solution was then neutralized with dry Dowex 1 (OH-type). After filtration, the whole solution was concentrated to an oil (47 mg, 76%). NMR ($CDCl_3$): δ 1.78(s, 3H), 2.75—3.01(m, 2H), 3.90—4.20(m, 1H), 3.97(s, 3H), 6.80—7.56 and 8.20—8.50(m, 4H).

4-Acetoxy-3-methyl-5-(3-pyridylmethyl)-3-pyrrlolin-2-one (15). A mixture of **13** (45 mg) and acetic anhydride (0.4 ml) was stirred for 3 h. The solution was then concentrated, and the residue was recrystallized from a mixture of chloroform and diisopropyl ether to give colorless needles (50 mg, 89%). Mp 134—134.5 °C. IR: ν_{max} 3180, 3080, 1780, 1700, 1575, 1480, 1420, 1200, 760, 705 cm^{-1}. NMR ($CDCl_3$): δ 1.57 (d, 3H), 2.16(s, 3H), 2.73—2.96(m, 2H), 4.40—4.68(m, 1H), 5.65(broad s, 1H), 7.05—7.65 and 8.22—8.48(m, 4H). Found: C, 63.16; H, 5.83; N, 11.45%. Calcd for $C_{13}H_{14}N_2O_3$: C, 63.40; H, 5.73; N, 11.38%.

*Bis[4-methyl-5-oxo-2-(3-pyridylmethyl)-3-pyrrolidinyl]boronate (17, 3,4-*cis*-4,5-*cis*) and (18, 3,4-*trans*-4,5-*cis*).* Sodium borohydride (200 mg) was added to a solution of **13** (200 mg) in 2-propanol (20 ml), after which the mixture was stirred at room temperature for 48 h. The mixture was then diluted with water and extracted with chloroform. The extract was washed with water, dried over anhydrous sodium sulfate, and concentrated under a vacuum to give **17** as colorless oil (165 mg, 82%). NMR (CD_3OD): δ 1.16(d, 3H, J= 6.5 Hz), 2.30(m, 1H), 2.79—3.15 (m, 2H), 3.70—4.18 (m, 2H), 7.20—7.90 and 8.26—8.60(m, 4H). The recrystallization of **17** from a mixture of methanol and diisopropyl ether afforded a stable isomer, **18**, as colorless prisms. Mp 210—218 °C. IR: ν_{max} 1710, 1585, 1485, 1115, 780, 720, 710 cm^{-1}. NMR (CD_3OD): δ1.15(d, 3H, J=7.5 Hz), 2.29 (m, 1H), 2.80—3.10(m, 2H), 3.53—4.20(m, 2H), 7.24—7.90 and 8.25—8.57(m, 4H). Found: C, 62.45; H, 6.67; N, 13.21%. Calcd for $C_{22}H_{27}N_4O_4B$: C, 62.57; H, 6.44; N, 13.27%.

Boron Complex of 4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)-pentanoic Acid (19). Compound (**18**, 100 mg) was dissolved in 48% hydrobromic acid, and the solution was stirred at 70 °C for 72 h. The solution was then concentrated and applied to an ion-exchange column of Dowex 50 (0.8 cm × 25 cm). The amino acid was eluted by 3 M-hydrochloric acid, and the residue of the eluate was recrystallized from a mixture of ethanol and 2-propanol to give **19** as colorless needles (70 mg, 60%). Mp 211.5—212 °C. IR: ν_{max} 1700, 1575, 1475, 1105, 1040, 773, 720, 700 cm^{-1}. Found: C, 53.78; H, 6.20; N, 11.49%. Calcd for $C_{22}H_{30}N_4O_6$·BCl: C, 53.62; H, 6.14; N, 11.37%.

*3,4-*trans*-4,5-*cis*-4-Acetoxy-3-methyl-5-(3-pyridylmethyl)-2-pyrrolidinone (20).* A mixture of **18** (125 mg), pyridine (2 ml), and acetic anhydride (2 ml) was stirred at room temperature for 24 h, diluted with water, and extracted with chloroform. The extract was dried over anhydrous sodium sulfate and concentrated to give a residue which was then recrystallized from a mixture of methanol and diisopropyl ether. Colorless needles (80 mg, 54.5%); mp 143.5—144 °C. IR: ν_{max} 1730, 1710, 1580, 1485, 1218, 785, 710 cm^{-1}. NMR (CD_3OD): δ 1.17 (d, 3H), 2.10 (s, 3H), 2.46 (m, 1H), 2.82 (d, 2H), 4.20 (q, 1H), 5.01 (t, 1H), 7.25—7.88 and 8.30—8.60 (m, 4H). Found: C, 62.91; H, 6.52; N, 11.33%. Calcd for $C_{13}H_{16}N_2O_3$: C, 62.89; H, 6.50; N, 11.28%.

4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic Acid Dihydrochloride (21, a Mixture of 2 and 23). a) The complex (**19**, 120 mg) was dissolved in 48% hydrobromic acid in the presence of ion-exchange resin (Dowex 50, 5 g). The mixture was stirred for 10 h, and the solvent was removed under reduced pressure. The resin was washed with water, and the amino acid was eluted with 3 M-hydrochloric acid. The concentration of the eluate gave **21**, an isomeric mixture of the dihydrochlorides of **2** and **23**, in the form of a viscous syrup (90 mg, 62%). The NMR spectrum of the sample gave an approximate ratio of **23** : **2**=9 : 1.

b) The acetate (**20**, 200 mg) was dissolved in 48% hydrobromic acid. The mixture was stirred at 70 °C for 40 h and then concentrated to a viscous syrup in the form of

dihydrobromide, with the same isomeric ratio as above (280 mg, 90%).

*Ethyl 4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoate Dihydrobromide (**22**).* The above dihydrobromide (**21**, 250 mg) was crystallized from ethanol to give **22** as colorless needles (150 mg, 56%). Mp 191 °C (dec). IR: ν_{max} 1725, 1610, 1590, 1510, 1204, 1095, 790, 675 cm^{-1}. NMR (CD_3OD): δ 1.24 (t, 3H), 1.25 (d, 3H), 2.85 (m, 1H), 3.25—3.50 (m, 2H), 3.65—4.20 (m, 2H), 4.14 (q, 2H), 8.00—8.30 and 8.61—9.10 (m, 4H). Found: C, 37.41; H, 5.30; N, 6.75%. Calcd for $C_{13}H_{20}N_2O_3 \cdot 2HBr$: C, 37.70; H, 5.35; N, 6.76%.

*4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic Acid Dihydrochloride (**23**).* The ethyl ester (**22**, 100 mg) was stirred with 4 M-hydrochloric acid (25 ml) at room temperature for 10 h; then the mixture was concentrated, and the residue was treated with dioxane. As the precipitate was very hygroscopic, it was filtered with the exclusion of moisture, washed with dioxane, and dried to yield **23** (68.5 mg, 90%). IR: ν_{max} 1710, 1605, 1548, 1466, 1185, 780, 673 cm^{-1}. NMR (D_2O): δ 1.14 (d, 3H, J=7.0 Hz), 2.80 (m, 1H), 3.18—3.42 (m, 2H), 3.58—3.94 (m, 2H), 7.91—8.21 and 8.44—8.84 (m, 4H). Found: C, 42.21; H, 6.21; N, 8.87%. Calcd for $C_{11}H_{16}N_2O_3 \cdot 2HCl \cdot H_2O$: C, 41.92; H, 6.40; N, 8.89%.

*4-Amino-3-hydroxy-2-methyl-5-(3-pyridyl)pentanoic Acid Dihydrochloride (**2**).* A hydroxy amino acid mixture (**21**, 185 mg) which contained *dl*-**2** (10%) and its diastereomer (**23**, 90%) was applied to a column of Dowex 50 (H-form, 100—200 mesh, 0.8 cm × 50 cm). The column was washed with water, and then the amino acid was eluted with 2M-hydrochloric acid. The diastereomer (**23**) was eluted a little faster than **2**. The fractions containing *dl*-**2** were combined and concentrated. The concentrate was, again, applied to the column; this process was repeated five times. The glassy concentrate thus obtained (4 mg) gave an NMR spectrum identical with that of natural (−)-**2**. NMR (D_2O): δ 1.13 (d, 3H, J=7.0 Hz), 2.80 (quintet, 1H), 3.18—3.44 (m, 2H), 3.67—3.91 (m, 2H), 7.95—8.25 (m, 1H), 8.50—8.85 (m, 3H).

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References

1) (a) H. Umezawa, T. Aoyagi, H. Morishima, M. Hamada, and T. Takita, *J. Antibiot.* (*Tokyo*), **23**, 259 (1970); (b) H. Morishima, T. Takita, T. Aoyagi, T. Takeuchi, and H. Umezawa, *ibid.*, **23**, 263 (1970); (c) S. Murao and S. Satoi, *Agric. Biol. Chem.*, **34**, 1265 (1970); (d) T. Miyano, M. Tomiyasu, H. Iizuka, S. Tomisaka, T. Takita, T. Aoyagi, and H. Umezawa, *J. Antibiot.* (*Tokyo*), **25**, 489 (1972); (e) T. Aoyagi, Y. Yagisawa, M. Kumagai, M. Hamada, H. Morishima, T. Takeuchi, and H. Umezawa, *ibid.*, **26**, 539 (1973).

2) H. Ogawara, K. Maeda, G. Koyama, H. Nagawara, and H. Umezawa, *Chem. Pharm. Bull.*, **16**, 679 (1968).

3) H. Morishima, T. Takita, and H. Umezawa, *J. Antibiot.* (*Tokyo*), **26**, 115 (1973).

4) M. Kinoshita, A. Hagiwara, and S. Aburaki, *Bull. Chem. Soc. Jpn.*, **48**, 570 (1975).

5) M. Kinoshita and S. Mariyama, *Bull. Chem. Soc. Jpn.*, **48**, 2081 (1975).

6) K. Achiwa and S. Yamada, *Chem. Pharm. Bull.*, **14**, 537 (1966).

7) *dl*-**1** was converted into its diastereomer as follows. *dl*-**1** was *N*-benzoylated, and the product was treated with diazomethane and then with thionyl chloride. The imino ester thus obtained was a mixture of two diastereomers (50 : 50), which was then hydrolyzed to give a mixture of *dl*-**1** and its diastereomer.

8) R. K. Griffith and H. J. Harwood, *J. Org. Chem.*, **29**, 2658 (1964).

9) The ethyl ester (**22**) was converted with silver oxide to 4-hydroxy-3-methyl-5-(3-pyridyl)methyl-2-pyrrolidinone, the hydrolysis of which gave an amino-acid mixture (**21**).

10) The 2,4-pyrrolidinedione (**6**) existed in the keto-form in chloroform, but in the enol-form in methanol (NMR).