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80. Shiro Terashima, Kazuo Achiwa, and Shun-ichi Yamada : Studies on Optically Active Amino Acids. Ⅸ.*1 Studies on α-Alkyl-α-amino Acids. Ⅳ.*2 Chemical Correlation of Absolute Configuration of α-Methylaspartic Acid to Isovaline.*3

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In our previous paper the absolute configuration of (+)−isovaline ((+)−I) was established to be *S* (+)−isovaline, L-α-methylbutyrine or D-α-ethylalanine by the chemical correlation with D-(−)-quinic acid.[1,2] Furthermore, an attempt has been made to examine the correlation of the absolute configuration of α-methylaspartic acid to that of (+)-isovaline already established, and this correlation studied will be reported in this paper. α-Methylaspartic acid seems to be an important key amino acid for the determination of the absolute configuration of several amino acids of this series.

The chemical scheme which we employed is shown in Chart 1. Before the optically active amino acids were attempted, favorable reaction conditions were examined using the racemic compounds.

2-Acetamido-2-methyl-4-benzyloxybutyric acid (Ⅱ)[3,4] was chosen as a starting material as shown in Chart 1. Reduction of DL-Ⅱ with 30% palladium-carbon at room temperature and atmospheric pressure underwent debenzylation to give DL-Ⅲ, which, without isolation, was refluxed with acetic anhydride and acetic acid to afford the lactone, DL-2-acetamido-2-methyl-γ-butyrolactone (DL-Ⅳ), m.p. 126~127.5° in a 68% yield. A mixed melting point of DL-Ⅳ thus obtained with the authentic sample prepared from DL-α-methylhomoserine (DL-Ⅺ)[5] according to the procedure of Murata, *et al.*[6] was undepressed and also their IR spectra were superimposed. Optically active (−)-Ⅱ was easily obtained by the resolution method employing l-menthyl ester reported from our laboratory.[3] Reduction of (−)-Ⅱ under the similar procedures gave (+)-Ⅳ, m.p. 117~118°, $[\alpha]_D^{25}$ +18.6° (methanol) in a 55% yield. Reflux of DL-Ⅳ in dimethylformamide with sodium thiophenolate for 4 hours[7] afforded quantitatively DL-2-acetamido-2-methyl-4-phenylthiobutyric acid (DL-Ⅴ) m.p. 188.5~189.5, and similarly (−)-Ⅴ, m.p. 198.5~200°, $[\alpha]_D^{20}$ −19.1° (methanol), was obtained from (+)-Ⅳ. Desulfurization of DL-Ⅴ in ethanol with Raney nickel according to the Mozingo procedure[8] yielded N-acetyl-DL-isovaline (DL-Ⅵ) m. p. 186.5~187° in an 80% yield. A mixed melting point with the authentic sample prepared from DL-isovaline (DL-I)[3] was undepressed and the IR spectra of the two samples were superimposable and then the elemental analysis was consistent

*1 Part Ⅷ : This Bulletin, **14**, 537 (1966).
*2 Part Ⅲ : *Ibid.*, **14**, 537 (1966).
*3 A part of this work was communicated in This Bulletin, S. Yamada, S. Terashima, K. Achiwa : *Ibid.*, **13**, 227 (1965).
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1) S. Yamada, K. Achiwa : This Bulletin, **12**, 1525 (1964).
2) K. Achiwa, S. Yamada : *Ibid.*, **14**, 537 (1966).
3) S. Terashima, K. Achiwa, S. Yamada : This Bulletin, **13**, 1399 (1965).
4) *a*) J. Murata, H. Arai, M. Tanaka : Kogyo Kagaku Zasshi, **60**, 1206 (1957). *b*) J. Murata, H. Arai : *Ibid.*, **56**, 628 (1953). *c*) U. Shimodoi, M. Sashio, J. Murata : *Ibid.*, **63**, 2140 (1960).
5) H. Brockman, H-B, König, R. Oster : Ber., **87**, 856 (1954).
6) U. Shimodoi, K. Masuda, J. Murata : Kogyo Kagaku Zasshi, **65**, 1664 (1962).
7) cf. S. Kukolja : Tetr., **18**, 1161 (1962).
8) R. Mozingo, D. E. Wolf, S. A. Harris, K. Folkers : J. Am. Chem. Soc., **65**, 1013 (1943).

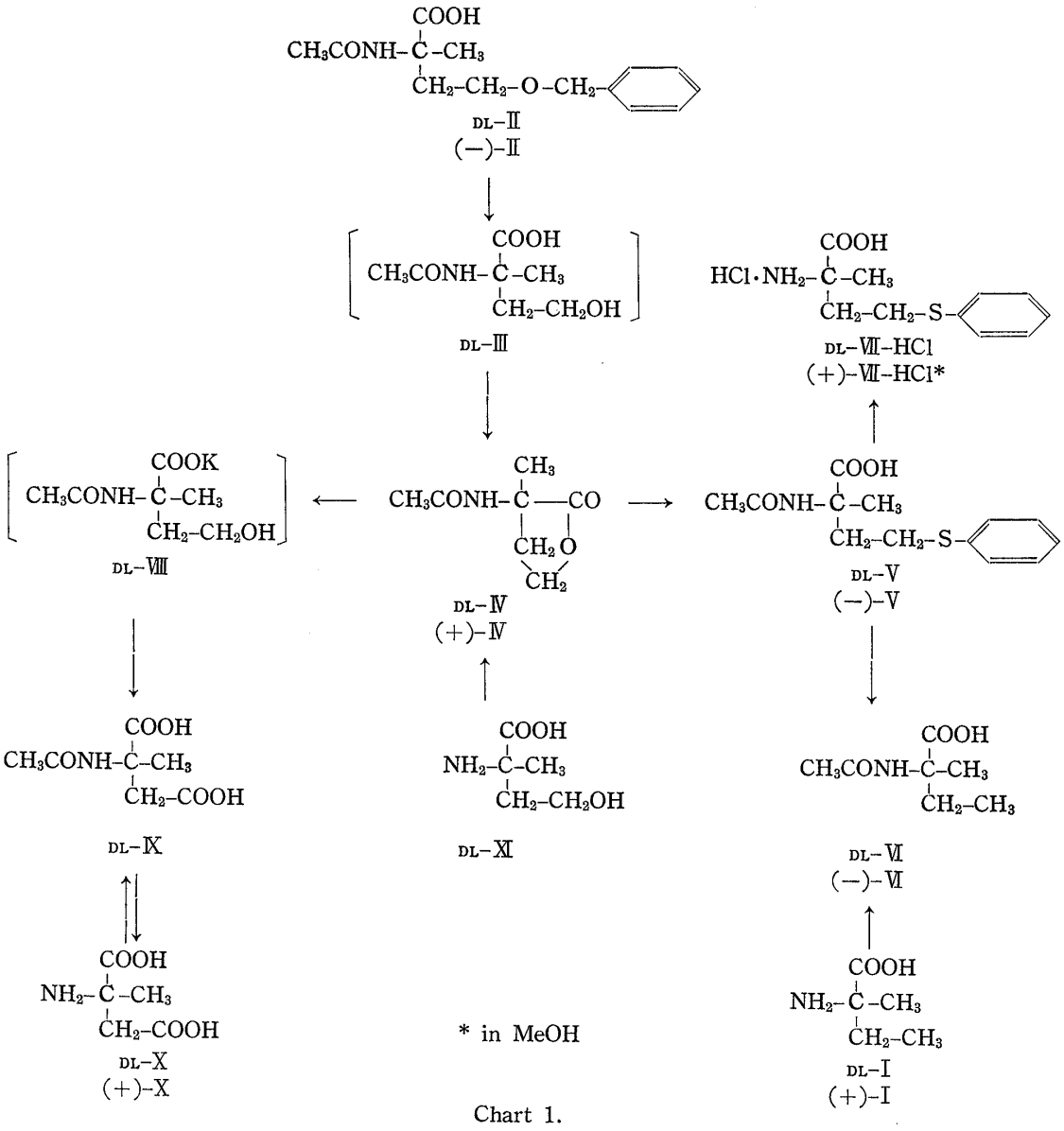

Chart 1.

with that expected for structure VI. Similar procedures on (−)-V afforded (−)-VI in a 48% yield m.p. 193~193.5°, $[\alpha]_D^{18}$ −1.3° (methanol), whose IR spectrum in solid state was superimposable with the authentic (−)-VI m.p. 190~191.5°, $[\alpha]_D^{34}$ −1.3° (methanol) prepared from (+)-isovaline, and a mixed melting point showed no depression.

(+)-Isovaline ((+)-I) was obtained from the resolution of racemic modification according to the procedure of Akabori, *et al.*[9] The $[\alpha]_D$ value of (−)-VI was so small that optical rotatory dispersion (ORD) curves[10] of (−)-VI prepared from both (−)-V and (+)-I were measured in the range from 700 to 250 mμ. The well consistency of both curves was observed in the range measured as shown in Fig. 1.

Accordingly, it has been proved that the absolute configuration of (+)-IV, (−)-V, (−)-VI and (+)-I belonged to the same series concerning the α-asymmetric carbon atom.

Reflux of (−)-V with 12% hydrochloric acid for 3 hours afforded (+)-2-amino-2-methyl-4-phenylthiobutyric acid (VII) hydrochloride m.p. 251~253° (decomp.), $[\alpha]_D^{20}$ +21.2°

9) S. Akabori, T. Ikenaka, K. Matsuki : Nippon Kagaku Zasshi, **73**, 112 (1952).
10) The measurements of ORD curves were performed using a Spectrophotometer model ORD/UV-5, Japan Spectroscopic Co., Ltd.

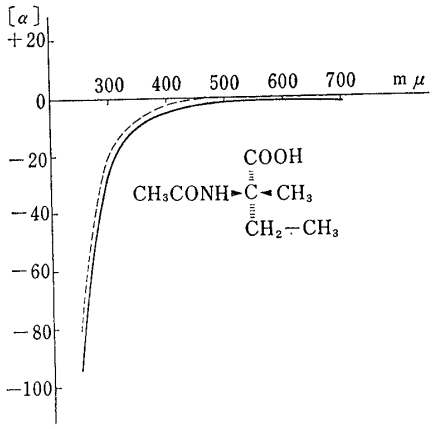

Fig. 1. Optical Rotatory Dispersion Curves of (−)-Ⅵ from (−)-Ⅴ and (+)-I
——— (−)-Ⅴ from (−)-Ⅴ
- - - - (−)-Ⅴ from (+)-I

(methanol) whose absolute configuration was to belong to the same series as (+)-I.

Subsequently, the chemical correlation of (+)-Ⅳ to optically active α-methylaspartic acid (X) was undertaken. An aqueous solution of DL-Ⅳ containing 2 molar equivalent of potassium hydroxide afforded N-acetyl-DL-α-methylhomoserine potassium salt (DL-Ⅷ) which, without separation, was oxidized with potassium permanganate to give N-acetyl-DL-α-methylaspartic acid (DL-Ⅸ) in a 61% yield m.p. 157.5~158.5° (decomp.). A mixed melting point with the authentic sample[11] prepared from the reaction of DL-α-methylaspartic acid (DL-X) with acetic anhydride and water, was undepressed and IR spectra of the both compounds were superimposed. However recrystallization of DL-Ⅸ was found to be very difficult, so the crude DL-Ⅸ obtained after oxidation was, without isolation, treated with ca. 12% hydrochloric acid to hydrolyse N-acetyl group. The amino acid hydrochloride (DL-X·HCl) obtained, was purified by a column chromatography on cellulose powder, and neutralization of the obtained DL-X·HCl with pyridine in alcohol afforded amino acid DL-X, as monohydrate m.p. 234~235° (decomp.) in a 50% yield based on DL-Ⅳ. The amino acid exhibited only a single spot on paper chromatogram developed by three different solvent systems, and each Rf value was consistent with that of an authentic sample DL-X[12] as shown in Table I. Its IR spectrum was superimposable with that of an authentic sample. And hence, (+)-Ⅳ was treated as the same manner as DL-Ⅳ to give (+)-X as colorless small needles, m.p. 256~257° (decomp.), $[\alpha]_D^{18}$ +49.0° (water)[13] in a 30% yield.

Table I. Rf Values of α-Methylaspartic Acid on Paper Chromatography[a]

Samples	Rf Values for various solvents[b]		
	A[c,d]	B[c,e]	C[c,e]
Authentic DL-α-methylaspartic acid (DL-X)	0.30	0.22	0.03
DL-α-Methylaspartic acid from DL-Ⅳ	0.29	0.23	0.03
S(+)-α-Methylaspartic acid from (+)-Ⅳ	0.31	0.24	0.03

a) Toyo filter paper No. 51, ascending method.
b) Reagent for coloring, 0.5% ninhydrin in acetone. After sprayed, the chromatogram was heated for a short time.
c) Solvent systems.
A: n-BuOH-AcOH-H_2O=4:1:2. B: n-BuOH-Pyridine-H_2O=1:1:1. C: n-BuOH-EtOH-2N-NH_4OH=5:1:2.
d) Reddish violet.
e) Violet.

11) P. Pfeiffer, E. Heinrich: J. prakt. Chem., **146**, 105 (1936). In this reference, it was reported that DL-Ⅸ was obtained from the reaction of DL-X with acetyl chloride and recrystallized from ethanol, and the further recrystallization from acetone-ligroin gave pure DL-Ⅸ. We repeated their procedures and obtained the compound, m.p. 156.5~158°, which was found to be different from DL-Ⅸ, even though its m.p. was very similar to that of the authentic DL-Ⅸ, and this compound was supposed to be monoester of DL-Ⅸ from its IR spectrum and elementary analysis. Therefore the authentic sample of DL-Ⅸ was prepared from DL-X using acetic anhydride and water by our hand.
12) See Ref. 11 and the experimental part.
13) In ref. 11, it was reported that the resolution of DL-α-methylaspartic acid with strychinine gave two antipodal optically active amino acids, one is m.p. 240°, $[\alpha]_D^{23}$ +3.46° (c=19.37, water), the other $[\alpha]_D^{24}$ −3.66° (c=15.84, water) and m.p. is not described. There was a remarkable difference of $[\alpha]_D$ value in-between. We followed the procedure reported by Pfeiffer, *et al.* to obtain optically active X, but the resolution was unsuccessful.

This (+)-X showed only a single spot on paper chromatogram by three different solvent systems respectively, and their Rf-values were in accordance with those of DL-X, as shown in Table I. IR spectrum of (+)-X in a solid state was very different from that of DL-X as shown in Fig. 2.

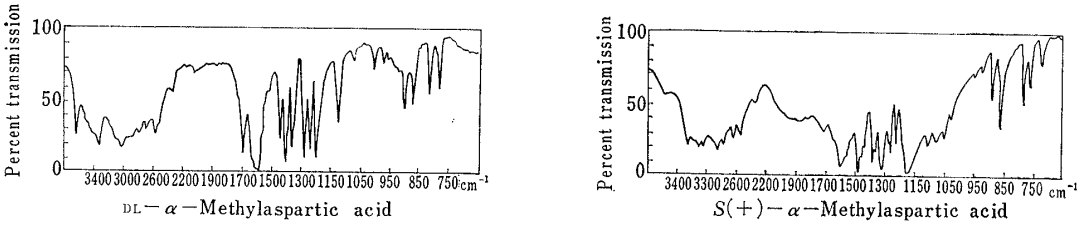

Fig. 2. Infrared Spectra of DL-α-Methylaspartic Acid and S(+)-α-Methylaspartic Acid in Solid State (KBr)

Based on the above results, the absolute configuration of α-asymmetric carbon atom of (+)-α-methylaspartic acid ((+)-X) is the same as that of (+)-I, whose absolute configuration has been correlated unequivocally to glyceraldehyde.[1,2] Consequently as shown in Chart 2, (+)-α-methylaspartic acid ((+)-X) can be named as S (+)-α-methylaspartic acid or L (+)-α-methylaspartic acid according to the nomenclature of α-alkyl-α-amino acid proposed in our previous report.[2]

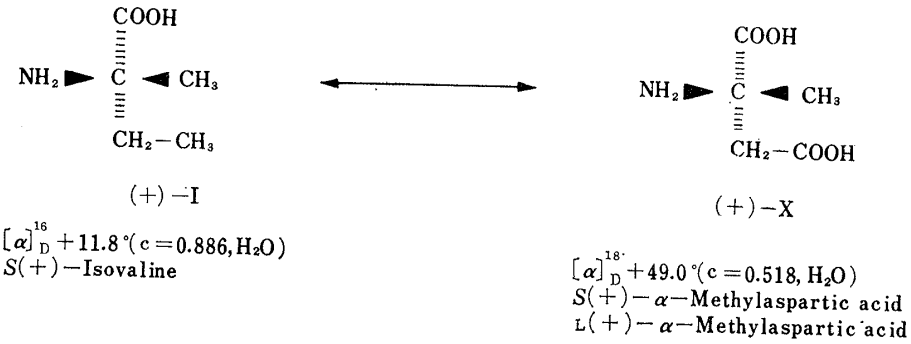

Chart 2.

Experimental[14]

DL-2-Acetamido-2-methyl-γ-butyrolactone (DL-IV)——a) DL-IV from DL-2-acetamido-2-methyl-4-benzyloxybutyric acid (DL-II): A mixture of DL-II[3,4] (1.0 g., 0.0038 mole) and 30% Pd-C (0.5 g.) in EtOH (75 ml.) was hydrogenated at room temperature and atmospheric pressure. After the theoretical amount of H_2 was absorbed (3 hr.), the catalyst was filtered and washed with EtOH. The combined ethanolic filtrate and the washings were evaporated to dryness *in vacuo* to afford a clear oil (0.47 g.). A mixture of AcOH (10 ml.) and Ac_2O (15 ml.) was added to the residual oil. The resulted solution was refluxed for 2.5 hr. in an oil bath, and then evaporated to dryness *in vacuo* to give a brown oil. This oil was dissolved in a small amount of $CHCl_3$, and ether was added dropwise to the $CHCl_3$ solution. The brown oil separated solidified on cooling. After filtration, the brownish white solid (0.40 g., 68%) showed m.p. 124.5~127°. Several recrystallizations from $CHCl_3$-*n*-hexane gave small needles, m.p. 126~127.5°. IR ν_{max}^{KBr} cm^{-1} :3380, 3310, 1778, 1660, 1545. This IR spectrum was identical with that of an authentic sample (see b). The mixed melting point with an authentic sample (see b, m.p. 126.5~128°) showed no depression (m.p. 126.5~127.5°).

b) DL-IV from DL-α-methylhomoserine: A mixture of DL-α-methylhomoserine[5] (5.5 g., 0.041 mole) and Ac_2O (55 ml.) was refluxed for 6 hr. in an oil bath according to the method reported by Murata, *et al.*[6] Evaporation and recrystallization from $CHCl_3$-*n*-hexane gave white powdery crystals (5.0 g., 77%), m.p.

14) All melting points are uncorrected. IR spectra measurements were carried on with a Spectrophotometer, Model DS-301. Japan Spectroscopic Co., Ltd. Optical activities were measured with a Yanagimoto Photo Magnetic Direct Polarimeter, Model OR-20.

125.5～127°. Pure sample was obtained by the further recrystallization from $CHCl_3$–*n*-hexane, m.p. 126.5～128°. (lit.,[6] m.p. 126.5～128°). IR ν_{max}^{KBr} cm^{-1}: 3390, 3320, 1777, 1657, 1543.

(+)-2-Acetamido-2-methyl-γ-butyrolactone ((+)-IV)——A mixture of (−)-2-acetamido-2-methyl-4-benzyloxybutyric acid[3] ((−)-II) (m.p. 151～153°, $[\alpha]_D^{25}$ −6.7° (c=2.46, MeOH)) (12.0 g., 0.0452 mole) and 30% Pd-C (5.8 g.) in EtOH (200 ml.) was treated as same as the case of DL-II. A mixture of AcOH (75 ml.) and Ac_2O (25 ml.) was added to the debenzylated product and refluxed for 3 hr. Evaporation *in vacuo* gave a pale yellow oil (8.0 g.), which was dissolved in $CHCl_3$ (20 ml.). $CHCl_3$ solution was cooled, and stimulated after dropwise addition of ether (30 ml.) to afford white solid (4.1 g.), m.p. 68～90° (clear at ca. 100°). This solid (4.1 g.) was purified on column chromatography using silica gel (410 g.). The eluting solvent was changed successively from CH_2Cl_2, $CHCl_3$ and $CHCl_3$ containing 10% EtOH. Fractions eluted by $CHCl_3$ containing 10% EtOH gave prisms (3.94 g., 55%) after an evaporation of solvent *in vacuo*, m.p. 116～117°, $[\alpha]_D^{25}$ +19.0° (c=1.28, MeOH). Further recrystallization from dioxane-iso-Pr_2O afforded pure (+)-IV as prisms, m.p. 117～118°, $[\alpha]_D^{25}$ +18.6° (c=1.28, MeOH). *Anal.* Calcd. for $C_7H_{11}O_3N$: C, 53.49; H, 7.05; N, 8.91. Found: C, 53.60; H, 6.85; N, 8.79. IR ν_{max}^{KBr} cm^{-1}: 3320, 1760, 1671, 1540. This IR spectrum was different from that of DL-IV.

DL-2-Acetamido-2-methyl-4-phenylthiobutyric Acid (DL-V)——A mixture of Na (0.15 g., 0.0065 mole) and thiophenol (0.85 g., 0.0077 mole) in absolute MeOH (20 ml.) was evaporated to dryness *in vacuo* to give sodium thiophenolate as white solid. DL-IV (1.1 g., 0.0070 mole) and dimethylformamide (20 ml.) were added to the sodium thiophenolate. The resulted mixture was refluxed for 4 hr. in an oil bath and evaporated to dryness after kept standing overnight to afford reddish brown oil, which was dissolved in H_2O (20 ml.) and acidified with dil. HCl. The yellowish white solid crystallized out was filtered, washed with aq. EtOH and dried. Crude DL-V (1.9 g., quantitative) showed m.p. 177～180°. Once recrystallization from 40% aq. EtOH and later twice from EtOH afforded white needles, m.p. 188.5～189.5°. *Anal.* Calcd. for $C_{13}H_{17}O_3NS$: C, 58.35; H, 6.36; N, 5.24. Found: C, 58.52; H, 6.38; N, 5.02. IR ν_{max}^{KBr} cm^{-1}: 3360, 1714, 1627, 1587, 1555, 744, 690.

(−)-2-Acetamido-2-methyl-4-phenylthiobutyric Acid ((−)-V)——(+)-IV (m.p. 115.5～117°, $[\alpha]_D^{24}$ +18.1° (c=1.82, MeOH)) (2.0 g., 0.0127 mole) was treated as same as DL-IV to give crude (−)-V as yellowish white solid (3.4 g., quantitative), m.p. 189.5～192°. Recrystallizations similar to those of DL-IV gave white needles, m.p. 198.5～200°. $[\alpha]_D^{20}$ −19.1° (c=1.40, MeOH). *Anal.* Calcd. for $C_{13}H_{17}O_3NS$: C, 58.52; H, 6.38; N, 5.24. Found: C, 58.48; H, 6.31; N, 5.18. IR ν_{max}^{KBr} cm^{-1}: 3370, 1717, 1621, 1583, 1560, 732. This IR spectrum was different from that of DL-V.

N-Acetyl-DL-isovaline (DL-VI)——a) DL-VI from DL-2-acetamido-2-methyl-4-phenylthiobutyric acid (DL-V): A mixture of DL-V (0.50 g., 0.0019 mole) and Raney Ni for desulfurization[8] (ca. 5 g.) in absolute EtOH (50 ml.) was refluxed on a water bath for 8 hr. Raney Ni was filtered and washed with ca. 1% NaOH solution preheated at 50°. A combined filtrate and the washings were concentrated to ca. 10 ml. *in vacuo*, and acidified with conc. HCl. The acidic aqueous solution was extracted with AcOEt (20 ml.×3). The combined AcOEt layer was washed with satd. NaCl (20 ml.), dried over anhyd. Na_2SO_4, and evaporated to dryness *in vacuo* to give white powder (0.24 g., 80%), m.p. 186～187°. Recrystallization from H_2O gave colorless crystals, m.p. 186.5～187°. The mixed melting point with an authentic DL-VI (see b, m.p. 187～187.5°) showed no depression m.p. 187～187.5°. *Anal.* Calcd. for $C_7H_{13}O_3N$: C, 52.81; H, 8.23; N, 8.80. Found: C, 52.88; H, 7.94; N, 8.77. IR ν_{max}^{KBr} cm^{-1}: 3390, 1700, 1611, 1538. This IR spectrum was superimposable with that of an authentic DL-VI (see b).

b) DL-VI from DL-isovaline (DL-I): Anhyd. DL-I[3] was treated with pyridine–Ac_2O as usual to give DL-VI (88%). Recrystallization from H_2O afforded colorless crystals, m.p. 187.5～189°. *Anal.* Calcd. for $C_7H_{13}O_3N$: C, 52.81; H, 8.23; N, 8.80. Found: C, 52.80; H, 8.31; N, 8.76. IR ν_{max}^{KBr} cm^{-}: 3400, 1700, 1610, 1540.

(−)-N-Acetylisovaline ((−)-VI)——a) (−)-VI from (−)-2-acetamido-2-methyl-4-phenylthiobutyric acid ((−)-V): A mixture of (−)-V (m.p. 197～199°, $[\alpha]_D^{21}$ −16.6° (c=1.25, MeOH) (1.50 g., 0.0056 mole) and Raney Ni for desulfurization[8] (ca. 15 g.) in absolute EtOH (100 ml.) was refluxed for 8 hr. and then treated similary to the case of DL-V to give white residue (0.60 g., 68%), which was recrystallized from H_2O (ca. 8 ml.) to give colorless pillars (0.42 g., 48%), m.p. 192～193°. Further recrystallization from H_2O afforded pure (−)-VI as pillars, m.p. 193～193.5°, $[\alpha]_D^{18}$ −1.3° (c=1.32, MeOH). This sample showed m.p. 192～193.5° on mixed melting point with the authentic (−)-VI (see b, m.p. 190～191.5°, $[\alpha]_D^{34}$ −1.3° (c=1.98, MeOH)). *Anal.* Calcd. for $C_7H_{13}O_3N$: C, 52.81; H, 8.23; N, 8.80. Found: C, 53.03; H, 8.42; N, 8.66. IR ν_{max}^{KBr} cm^{-1}: 3370, 1711, 1620, 1558. This IR spectrum was identical with that of the authentic (−)-VI (see b) and not identical with that of DL-VI. ORD[10]: $[\alpha]^{27}$ (c=1.32, MeOH) (mμ): −94.6° (250), −26.1° (300), −10.0° (350), −5.5° (400), −1.8° (500), −1.2° (589), −1.5° (700). ORD curve showed the same feature as that of the authentic (−)-VI (see b).

b) (−)-VI from *S*(+)-isovaline[1～3] (*S*(+)-I): (−)-VI was obtained from *S*(+)-I after the same treatment as in the case of DL-VI from DL-I. Recrystallizations from H_2O afforded pure (−)-VI as pillars, m.p. 190～191.5°, $[\alpha]_D^{34}$ −1.3° (c=1.98, MLeOH). *Anal.* Calcd. for $C_7H_{13}O_3N$: C, 52.81; H, 8.23; N, 8.80. Found: C, 52.62; H, 8.03; N, 8.86. IR ν_{max}^{KBr} cm^{-1}: 3365, 1712, 1622, 1557. This IR spectrum was not identical

with that of DL-Ⅵ. ORD[10]: $[\alpha]^{41}$ (c=1.98, MeOH) (mμ): −80.8°(252), −19.4°(300), −7.9°(350), −2.4° (400), −1.0°(500), ±0°(589), ±0°(700).

S(+)-Isovaline[1~3] (**S(+)-I**)——N-Formyl-DL-isovaline was resolved by brucine and hydrolysed to S(+)-isovaline according to the method of Akabori, *et al.*[9] Recrystallization from H_2O-EtOH-ether gave S(+)-I as fine needles, m.p. >250°, $[\alpha]_D^{16}$ +11.8°(c=0.886, H_2O) (lit.,[15a] $[\alpha]_D^{19}$ +11.0° (H_2O) as d-isovaline; lit.,[15b] $[\alpha]_D^{25}$ +11.13°(H_2O) as L-isovaline). IR ν_{max}^{KBr} cm^{-1}: 3510, 3070, 1641, 1589, 1559.

DL-2-Amino-2-methyl-4-phenylthiobutyric Acid Hydrochloride (**DL-VII·HCl**)——Reflux of DL-2-acetamido-2-methyl-4-phenylthiobutyric acid (DL-V) in ca. 12% HCl gave DL-Ⅵ·HCl. Recrystallization from dil. aq. HCl afforded pure DL-Ⅵ·HCl as white plates, m.p. 246°(decomp.). *Anal.* Calcd. for $C_{11}H_{16}O_2SNCl$: C, 50.46; H, 6.16; N, 5.35. Found: C, 51.04; H, 6.34; N, 5.35. IR ν_{max}^{KBr} cm^{-1}: 3000~2900, 1745, 1597, 1589, 1504, 739, 693. Paper chromatograms developed with two different solvent systems[16] showed a single spot respectively. The Rf value: 0.81 (solvent A), 0.82 (solvent B).

(+)-2-Amino-2-methyl-4-phenylthiobutyric Acid Hydrochloride (**(+)-VII·HCl**)——(−)-2-Acetamido-2-methyl-4-phenylthiobutyric acid ((−)-V) (m.p. 197~199°, $[\alpha]_D^{21}$ −16.6°(c=1.25, MeOH)) (0.30 g., 0.0011 mole) in ca. 12% HCl (12 ml.) was refluxed for 3 hr. After cooled, the HCl solution which crystallized out the white powder was kept standing in an ice bath for 3 hr., to give crude (+)-Ⅵ·HCl (0.27 g., 92%), m.p. 250~254°(decomp.), $[\alpha]_D^{12}$ +19.8° (c=0.366, MeOH). Several recrystallizations from dil. aqueous HCl gave white crystals, m.p. 251~253°(decomp.), $[\alpha]_D^{20}$ +21.2°(c=0.774, MeOH). *Anal.* Calcd. for $C_{11}H_{16}O_2SNCl$: C, 50.46; H, 6.16; N, 5.35. Found: C, 50.61; H, 6.15; N, 5.18. IR ν_{max}^{KBr} cm^{-1}: 3135, 3015, 1743, 1598, 1589, 1505, 739, 693. This IR spectrum was different from that of DL-Ⅵ·HCl. Paper chromatograms developed with two different solvent systems[16] showed a single spot respectively. The Rf value: 0.80 (solvent A), 0.81 (solvent B).

N-Acetyl-DL-α-methylaspartic Acid (**DL-IX**)——a) DL-Ⅸ from DL-α-methylaspartic acid (DL-X): DL-X monohydrate (0.50 g., 0.0030 mole) in Ac_2O (20 ml.) was refluxed for 2.5 hr. After cooling, the reaction mixture was evaporated to dryness *in vacuo* to afford a brown oil. H_2O (20 ml.) was added to this oil and evaporated to dryness *in vacuo*. This procedure was repeated again. The residue was dissolved in EtOH (10 ml.) and EtOH solution was evaporated to dryness to give a yellowish white solid (0.75 g.). After this solid was dissolved in EtOH (20 ml.), the undissolved materials were filtered off. The filtrate was concentrated to ca. 3 ml. and kept standing at room temperature. When the plates began to crystallize out, EtOAc was dropped into the EtOH solution as slowly as possible (ca. 10 drops). After kept standing overnight at room temperature, the upper layer was decanted off. The twice rinsings of the residual crystals with EtOH (1 ml.) gave crude DL-Ⅸ (0.42 g., 74%), m.p. 161~162°(decomp.). Recrystallization from EtOH-AcOEt gave pure DL-Ⅸ, m.p. 160.5~161°(decomp.) (lit.,[13] m.p. 156~157°). *Anal.* Calcd. for $C_7H_{11}O_5N$: C, 44.44; H, 5.86; N, 7.41. Found: C, 44.48; H, 6.25, N, 7.36. IR ν_{max}^{KBr} cm^{-1}: 3410, 1728, 1718, 1617, 1531.

b) DL-Ⅸ from DL-2-acetamido-2-methyl-γ-butyrolactone (DL-Ⅳ): A mixture of DL-Ⅳ (0.50 g., 0.0032 mole) and KOH (0.50 g., 0.0089 mole) in H_2O (15 ml.) was stirred and cooled to 0~5° in an ice bath. $KMnO_4$ (0.70 g., 0.0044 mole) was added portionwise to the cooled reaction mixture under stirring for 1 hr. Ice bath was taken off and the stirring was continued for additional 3 hr. at room temperature. The color of $KMnO_4$ was faded out after 2 hr.' stirring, an additional $KMnO_4$ (0.10 g.) was added to the reaction mixture. An excess of $KMnO_4$ was converted to MnO_2 using EtOH (2 ml.). MnO_2 was filtered and washed with hot H_2O (50 ml.). The combined filtrate and washings were poured through column (Amberlite IR-120, H^+-form, 128 ml.). The column was eluted with H_2O. The eluates showing pH value from 1 to 7 was combined and evaporated to dryness to give white solid (0.64 g.). After this solid was dissolved in EtOH (20 ml.), the undissolved materials were filtered off. The filtrate was concentrated to ca. 2 ml., cooled, and stimulated. The filtration and drying of the white crystals gave crude DL-Ⅸ (0.37 g., 61%), m.p. 149~153° (decomp.). Recrystallization from EtOH gave DL-Ⅸ, m.p. 157.5~158.5°(decomp.). This DL-Ⅸ showed no depression (m.p. 158.5~159° (decomp.)) on the mixed melting point with the authentic DL-Ⅸ (see a, m.p. 159.5~160° (decomp.)). IR ν_{max}^{KBr} cm^{-1}: 3400, 1730, 1720, 1613, 1532. This IR spectrum was superimposable with that of authentic DL-Ⅸ (see a).

DL-α-Methylaspartic Acid (**DL-X**)——a) DL-X from ethyl acetacetate: According to the method reported by Bucherer, *et al.*[17] ethyl acetacetate was converted to 5-(2-ethoxycarbonylethyl)-5-methylhydantoin, pillars, m.p. 137.5~138°, 49% (lit.,[11] m.p. 138°; lit.,[17] m.p. 135~137°, 60%). This hydantoin was hydrolyzed by reflux with 20% aq. NaOH to gave DL-X as monohydrate and white needles. m.p. 235°(decomp.). Yield, 37% (lit.,[11] m.p. 233° (decomp.); lit.,[18a] m.p. 232~234° (decomp.); lit.,[18b] m.p. 229~230° (decomp.)). IR ν_{max}^{KBr} cm^{-1}: 3580, 3275, 1696, 1630 (sh), 1611, 1593, 1566 (sh). The paper chromatograms developed with three different solvent systems showed just one spot respectively (see Table I).

15) *a*) E. Fischer, R. von Grävenitz: Ann., **406**, 5 (1914). *b*) C. G. Baker, S-C. J. Fu, S. M. Birnbaum, H. A. Sober, J. P. Greenstein: J. Am. Chem. Soc., **74**, 4701 (1952).
16) Solvent A. *n*-BuOH-AcOH-H_2O (4:1:2). Solvent B. *n*-BuOH-Pyridine-H_2O (1:1:1).
17) H. T. Bucherer, V. A. Lieb: J. prakt. Chem., **141**, 5 (1934).
18) *a*) A. Piutti: Ber., **31**, 2044 (1898). *b*) W. Cocker, A. Lapworth: J. Chem. Soc., **1931**, 1391.

b) DL-X from DL-2-acetamido-2-methyl-γ-butyrolactone (DL-Ⅳ): A mixture of DL-Ⅳ (1.0 g., 0.0064 mole) and KOH (0.75 g., 0.0134 mole) in H_2O (30 ml.) was stirred and cooled in an ice bath. $KMnO_4$ (1.5 g., 0.0095 mole) was added to this mixture portionwise for ca. 1 hr. between 0° and 10°. After the addition of $KMnO_4$, further $KMnO_3$ (0.2 g) and H_2O (20 ml.) were added to the reaction mixture, which was stirred for 2 hr. and then kept standing overnight at room temperature. An excess of $KMnO_4$ was converted to MnO_2 using EtOH (4 ml.). MnO_2 was filtered and washed with hot H_2O (50 ml.). The combined filtrate and washings were poured through column (Amberlite IR-120, H^+-form, 154 ml.). The column was eluted until the eluate became neutral. All the eluates (ca. 800 ml.) were combined and evaporated to dryness to give colorless caramel (1.1 g.). Ca. 12% HCl (15 ml.) was added to this caramel and refluxed for 3 hr. Evaporation to dryness gave yellowish white solid (1.1 g.), which showed two spots (Rf value : 0.02, 0.09) on paper chromatography.[19] The spot showing Rf value 0.02 was DL-X by the comparison with the Rf value of the authentic DL-X (see a). The yellowish white solid (1.1 g.) was dissolved in EtOH (ca. 12 ml.). The EtOH solution was submitted to a cellulose powder column chromatography (50 g.) employing *n*-BuOH-EtOH-2*N*-NH_4OH (5:1:2) as eluting solvent system. 25 ml. each of the eluate was collected and tested with ninhydrin and paper chromatography[19] to find out whether the fractions would contain DL-X only. The fractions required were combined and evaporated to dryness *in vacuo* to afford colorless caramel (0.65 g.). H_2O (30 ml.) containing NaOH (0.50 g.) was added to this caramel and concentrated to ca. 10 ml. on a water bath. The residual alkaline solution was acidified to pH <1 with conc. HCl and evaporated to dryness. The residue was extracted with EtOH (10 ml. × 3), and the combined EtOH solution was evaporated to dryness *in vacuo*. A small amount of ca. 12% HCl was again added to the residue and evaporated to dryness to give yellow solid (0.71 g.), which showed single spot on the paper chromatography,[19] whose Rf value was as same as that of the authentic DL-X (see a). This solid was dissolved in a mixture of EtOH (14 ml.) and H_2O (1 ml.), and then pyridine (20 drops) was added to give crude DL-X (0.52 g., 50%), m.p. 231~233° (decomp.). Several recrystallizations from H_2O-acetone gave pure DL-X as monohydrate and white powderish needles, m.p. 234~235° (decomp.). *Anal.* Calcd. for $C_5H_9O_4N \cdot H_2O$: C, 36.36; H, 6.71; N, 8.48. Found : C, 36.59; H, 6.64; N, 8.38. IR ν_{max}^{KBr} cm^{-1} : 3575, 3270, 1697, 1633 (sh), 1613, 1593, 1567 (sh). This IR spectrum was identical with that of authentic sample (see a). The paper chromatographical data were shown in Table I.

(+)-α-Methylaspartic Acid ((+)-X)——(+)-2-Acetamido-2-methyl-γ-butyrolactone ((+)-Ⅳ) (m.p. 115.5~117°, $[\alpha]_D^{24}$ +18.1° (c=1.82, MeOH)) (1.0 g., 0.0064 mole) was treated as same as DL-Ⅳ to give crude (+)-X as white powder (0.28 g., 30%), m.p. 245~246° (decomp.), $[\alpha]_D^{18}$ +47.5° (c=0.864, H_2O). Several recrystallizations from H_2O-acetone afforded pure (+)-X as white small needles, m.p. 256~257° (decomp.), $[\alpha]_D^{18}$ +49.0° (c=0.518, H_2O).[15] *Anal.* Calcd. for $C_5H_9O_4N$: C, 40.81; H, 6.17; N, 9.52. Found : C, 40.98; H, 6.23; N, 9.54. IR ν_{max}^{KBr} cm^{-1} : 3200, 1731, 1640 (sh), 1619, 1603 (sh), 1565. This IR spectrum was quite different from that of DL-X. The paper chromatographical data were shown in Table I.

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Summary

The absolute configuration of (+)-α-methylaspartic acid has been established to be *S*-configuration by the chemical correlation with (+)-isovaline, which was already correlated to D-(−)-quinic acid. The chemical scheme employed was shown in Chart 1. Preliminary examinations using racemic compounds were also reported.

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19) Solvent : *n*-BuOH-EtOH-2*N* NH_4OH (5:1:2). Other conditions were similar to those described in Table I.