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The Synthesis, Resolution and Configuration of β-Hydroxyvaline[1]

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ABSTRACT

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DL-β-Hydroxyvaline (III) was synthesized from sodium β,β'-dimethylglycidate by aminolysis with benzylamine followed by hydrogenolysis. N-Benzoyl-DL-β-hydroxyvaline (IV) was resolved by use of (-)-α-methylbenzylamine to yield (-) N-benzoyl β-hydroxyvaline (VI). The configuration of VI was determined by converting it to methyl N-benzoyl-β-chlorovalinate (IX) and subsequent hydrogenolysis of IX yielded (-) methyl N-benzoylvalinate (X). From X, optically pure D-(-)-valine was obtained upon hydrolysis.

The finding of β-hydroxyvaline in nature has been reported.[2-4] However, later investigations showed that the natural occurrence of the amino acid is questionable.[5-8] Syntheses of the racemic amino acid have been reported by use of the addition reaction of mercuric acetate and bromine to β,β-dimethylacrylic acid.[5,9-11] Ammonolysis of ethyl β,β-dimethylglycidate resulted in β-amino α-hydroxy derivative.[12-14] Recently Kaneko and Inui[15] synthesized and

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[3] E. J. Czarnetsky and C. L. A. Schmidt, *J. Biol. Chem.*, *92*, 453 (1931).

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[8] B. H. Nicolet and L. A. Shinn, *ibid.*, *142*, 139 (1942).

[9] W. Schrauth and H. Geller, *Ber.*, *55*, 2783 (1922).

[10] S. Tatsuoka, T. Ueno and K. Hirata, *Yakugaku Zasshi*, *70*, 229 (1950).

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[12] S. N. Danilov and V. F. Martynov, *Zhur. Obschei Khim.*, *22*, 1572 (1952).

[13] O. von Schickh, German Patent 583,243 (1933) [*Chem. Abst.* *28*, 260 (1934)].

[14] V. F. Martynov, *Doklady Akad. Nauk*, U.S.S.R. *89*, 869 (1953).

[15] T. Kaneko and T. Inui, *J. Chem. Soc. Japan*, pure chem. sec. (Nippon Kagaku Zasshi), *82*, 743 (1961).

characterized racemic β-hydroxyvaline and an α-hydroxy-β-aminoisovaleric acid which were separated from the reaction mixture of ammonolysis of sodium β,β-dimethylglycidate (I).

Optically active β-hydroxyvaline has not been prepared. In this investigation, DL-β-hydroxyvaline was prepared by treatment of I with benzylamine in aqueous condition in a way similar to the method employed by Liwschitz _et_ _al_.[16,17] in which they obtained only α-amino β-hydroxy acids. The resulting N-benzyl-β-hydroxyvaline (II) was debenzylated by hydrogenolysis with palladium on charcoal. The prepared DL-β-hydroxyvaline (III)[18] was benzoylated and was resolved by (-) methylbenzylamine[19,20] to yield (-) N-benzoyl-β-hydroxyvaline (-) amine salt (V). From the salt (V), (-) N-benzoyl-β-hydroxyvaline (VI) was isolated. Free (-)-β-hydroxyvaline (VII) was obtained by hydrolysis of VI. To determine the configuration of the resolved (-)-β-hydroxyvaline (VII), (-) N-benzoyl-β-hydroxyvaline (VI) was converted to the methyl ester (VIII) by the use of diazomethane. The ester (VIII) was treated with thionyl chloride to convert VIII to

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[16] Y. Liwschitz, Y. Rabinsohn and D. Perera, _J_. _Chem_. _Soc_. 1116 (1962).

[17] Y. Liwschitz, Y. Rabinsohn and A. Haber, _ibid_., 3589 (1962).

[18] III showed a single spot in paper chromatography and also showed a single peak in the Phoenix model K-5000 automatic amino acid analyzer.

[19] W. Theilacker, _Chem_. _Ber_., _87_, 690 (1954).

[20] W. Leithe, _ibid_., _64_, 2831 (1931).

β-chloro ester (IX). The β-chloro ester was catalytically hydrogenolyzed by the use of palladium on charcoal to yield methyl N-benzoyl-valinate (X) as was done in the study of β-phenylserine.[21] The ester (X) was hydrolyzed and optically pure [illegible](-)-valine (XI) was obtained. Accordingly, the α-carbon atom of (-)-β-hydroxyvaline is assigned the [illegible]-configuration. The schematic route of this work is shown in Fig. 1.

Attempts to obtain active glycidic acid by resolution were unsuccessful, because of instability of the free glycidic acid.

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[21] K. Vögler, *Helv. Chim. Acta*, **33**, 2111 (1950).

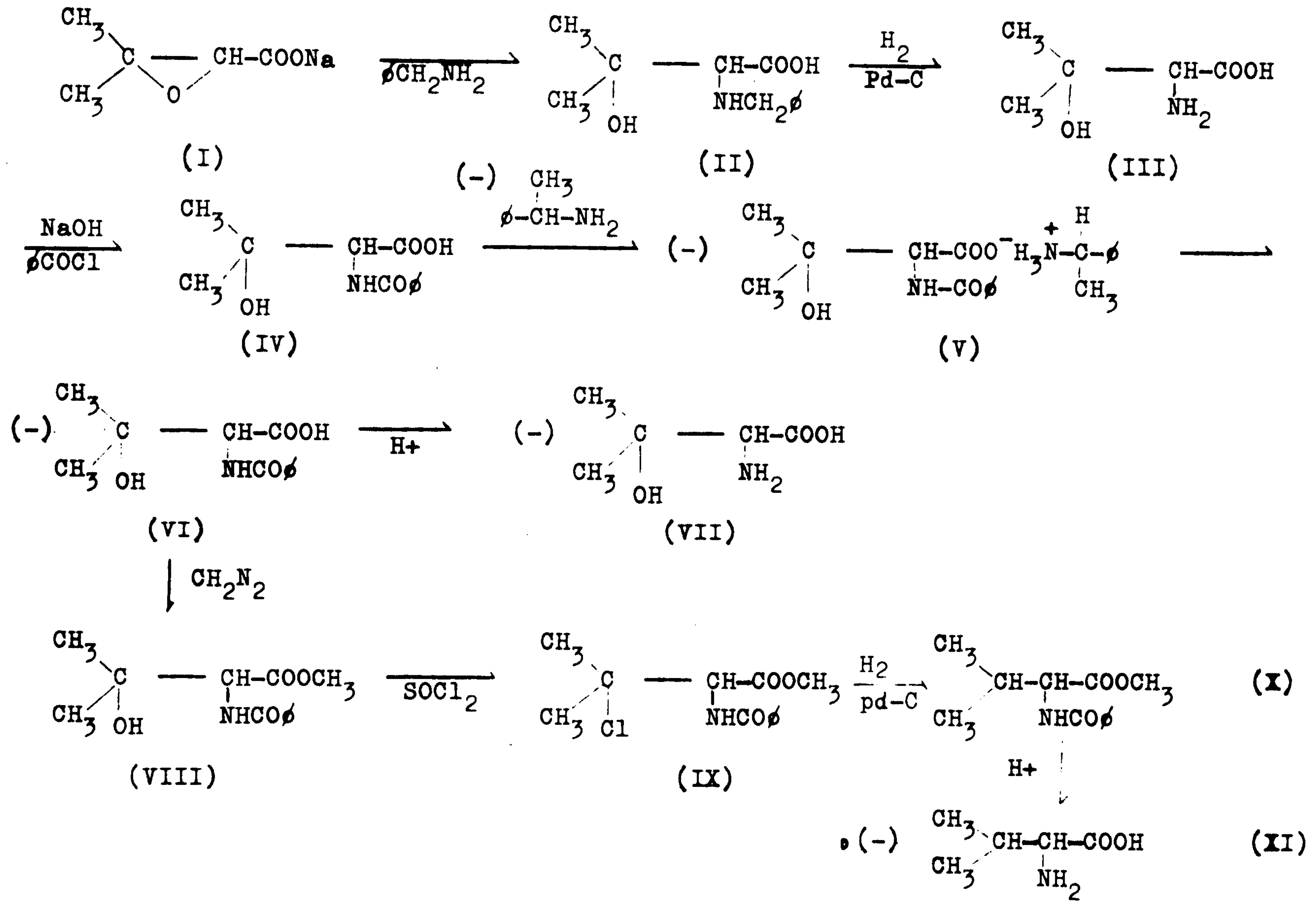

Fig. 1

Schematic route of synthesis, resolution and configuration determination of β-hydroxyvaline

EXPERIMENTAL[22]

DL-N-Benzyl-β-hydroxyvaline (II) —— A mixture of sodium 2,3-epoxyisovalerate[15] (I), 6.9 g. (0.05 mol.), water, 7 ml., and benzylamine, 8.0 g., was heated under reflux for 6 hr. After cooling, the excess of benzylamine was removed by distillation *in vacuo*. The pH of the aqueous solution was adjusted to 5.5 with concentrated hydrochloric acid. The precipitated crystals of DL-N-benzyl-β-hydroxyvaline (II) were separated by filtration and washed with acetone, then ether. The crude product, 7.6 g. (68%), was recrystallized from N,N'-dimethylformamide. Yield, 6.9 g. (62%), m.p. 204°C.

Found: C, 64.38; H, 7.80; N, 6.35. Calcd. for $C_{12}H_{17}NO_3$: C, 64.55; H, 7.67; N, 6.27%.

DL-β-Hydroxyvaline (III) —— DL-N-Benzyl-β-hydroxyvaline (II), 2.2 g. (0.01 mol.), was dissolved in 50 ml. of 50% methanol. To this solution, 1.0 g. of 5% palladium on charcoal was added and the hydrogenolysis was carried out at room temperature. When the hydrogen uptake ceased, the catalyst was removed by filtration. The filtrate was concentrated *in vacuo*. The crystalline residue was recrystallized from water and methanol. Yield, 1.21 g. (93%). This was further purified

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[22] All temperature measurements are uncorrected. All optical rotation measurements were carried out by the use of the Rudolph model 80 polarimeter with PEC-101 photometer. Elemental analyses were carried out by Micro-Tech Laboratories, Inc., Skokie, Illinois, U.S.A. Infrared absorption spectra were recorded by the use of the Perkin Elmer Model 137 B Infracord Spectrophotometer.

from water and ethanol. 1.13 g. (85%), m.p. 218°C (decomp.), [reported m.p. 226-227°C (decomp.)[15/]]. IR spectra showed[15/] absorption bands at 1670, 1620, 1600, 1560, 1420 cm^{-1} (amino acid), 1145 cm^{-1} ($\geqslant$C-OH). R_f value 0.44, BuOH:AcOH:H_2O = 4:1:2. The compound showed a single peak in the Phoenix model K-5000 automatic amino acid analyzer.

Found: C, 45.12; H, 8.57; N, 10.42. Calcd. for $C_5H_{11}NO_3$: C, 45.10; H, 8.33; N, 10.52%.

DL-N-Benzoyl-β-hydroxyvaline (IV) —— DL-β-Hydroxyvaline (III), 6.65 g. (0.05 mol.), was dissolved in 25 ml. of 2 N sodium hydroxide solution. The solution was chilled in an ice bath and benzoylated in the usual Schotten-Baumann method with a total of 7.0 g. of benzoyl chloride and 25 ml. of 2 N sodium hydroxide solution. Upon completion of the addition of reagents, the reaction mixture was shaken for 15 min. at room temperature and then acidified to pH 3.0 under cooling with concentrated hydrochloric acid. DL-N-Benzoyl-β-hydroxyvaline (IV) was extracted with ethyl acetate 2 times (100 ml. x 2) and this ethyl acetate layer was washed with water. The ethyl acetate solution was dried with anhydrous sodium sulfate. After evaporation of ethyl acetate *in vacuo*, 8.9 g. (75%) of crude IV was obtained. Contaminating benzoic acid was removed from the product by boiling with 30 ml. chloroform and subsequently filtering this mixture. These crystals were purified by recrystallization from ethanol and ether. Yield, 8.3 g. (70%), m.p. 156°C (decomp.), [reported

155-157°C (decomp.)[15]]. IR spectra showed bands at 1720 cm^{-1} (-COOH), 1635; 1520 cm^{-1} (-CONH-); 1152 cm^{-1} ($\geqslant$C-OH). The benzoyl derivative seems to correspond to the stable form of IV reported by Kaneko and Inui.[15] (The unstable form was reported m.p. 135-136°C.[15])

Found: C, 60.73; H, 6.53; N, 5.80. Calcd. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90%.

(-)-α-Methyl benzylamine salt of N-benzoyl-β-hydroxyvaline (V) —— DL-N-Benzoyl-β-hydroxyvaline (IV), 94.8 g. (0.4 mol.), and (-)-α-methylbenzylamine, 48.4 g. (0.4 mol.), ($[\alpha]_D^{25}$ = 42.3°, benzene), were dissolved in 50 ml. methanol. The solvent was evaporated *in vacuo*. The residual syrup was dissolved in hot acetone (150 ml.). After cooling, crystallization of (-)-α-methylbenzylamine salt (V) began by seeding with an authentic specimen which was obtained in another crystallization experiment. After standing overnight in a refrigerator, the crystals were filtered and washed with cold acetone. The crystals, 51.8 g. (72%), were purified from a hot mixture of methanol and acetone (5:95). Yield, 47.3 g. (66%), m.p. 155°, $[\alpha]_D^{25}$ = -22.9° (c 2.32, water).

Found: C, 67.12; H, 7.43; N, 7.64. Calcd. for $C_{20}H_{26}N_2O_4$: C, 67.02; H, 7.31; N, 7.82%.

(-) N-Benzoyl-β-hydroxyvaline (VI) —— (-)-α-Methylbenzylamine salt (V), 36.0 g. (0.1 mol.), was dissolved in 100 ml. of water. To this solution, 110 ml. of 2 N sodium hydroxide was added. The alkaline solution was extracted with ether twice (100 ml. x 2) to remove (-) α-methylbenzylamine. Concentrated hydrochloric acid was added to the aqueous solution to bring the pH to 3.0. (-)-N-Benzoyl-β-hydroxyvaline (VI) was extracted with ethyl acetate (100 ml. x 2). The ethyl acetate solution was washed with water and dried with anhydrous sodium sulfate. After evaporation of ethyl acetate <u>in vacuo</u>, 20.7 g. (87%) of crude VI was obtained. This was recrystallized from methanol and ether. Yield, 19.7 g. (83%), m.p. 112°C, $[\alpha]_D^{25} = -30.5°$ (c 2.01, ethanol). IR spectra showed bands at 1745 cm^{-1} (-COOH); 1645, 1550 cm^{-1} (-CONH-); 1150 cm^{-1} ($>$C-OH). The optically active N-benzoyl-β-hydroxyvaline also showed polymorphism which was detected by IR spectrographically as was reported for DL-N-benzyl β-hydroxyvaline.[15]

Found: C, 60.85; H, 6.32; N, 5.70. Calcd. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90%.

<u>(-)-β-Hydroxyvaline (VII)</u> —— A mixture of (-)-N-benzoyl-β-hydroxyvaline (VI), 2.37 g. (0.01 mol.), and 5 N hydrochloric acid, 50 ml., was refluxed for 5 hr. After cooling, the precipitated benzoic acid was separated by filtration. The filtrate was extracted with chloroform (50 ml. x 2) to remove the residual benzoic acid. After evaporation of the hydrochloric acid solution under reduced pressure, the residual syrup was dissolved in 50 ml. of water. The solution was treated with silver carbonate to remove hydrogen chloride. This mixture was filtered and washed with water. Hydrogen

sulfide gas was bubbled through the solution. Silver sulfide was removed by filtration. The filtrate was concentrated *in vacuo*. A yield of 1.18 g. (89%) of crude VII was obtained. This was recrystallized from water and ethanol, 0.997 g. (75%), m.p. 208° (decomp.), $[\alpha]_D^{25} = -13.5°$ (c 4.82, 5 N HCl). IR spectra showed bands at 1660, 1600, 1555, 1418 cm^{-1} (amino acid), 1145 cm^{-1} ($-\!\!\gt$C-OH). R_f value 0.44, BuOH:AcOH:H_2O = 4:1:2. The phenomenon of polymorphism of the compound was also observed by IR spectra. IR absorption curve varied depending on the ratio of water and ethanol in the recrystallization.

Found: C, 45.08; H, 8.31; N, 10.69. Calcd. for $C_5H_{11}NO_3$: C, 45.10; H, 8.33; N, 10.52%.

N-Benzoyl-β-hydroxyvaline methyl ester (VIII) ——
(-)-N-Benzoyl-β-hydroxyvaline (VI), 2.37 g. (0.01 mol.), was dissolved in 20 ml. of methanol. To this solution, diazomethane in ether solution was added. After the reaction was completed, excess diazomethane was decomposed by adding 5 per cent acetic acid in methanol solution. After evaporation of the solvent under reduced pressure, the residual syrup was dissolved in ethyl acetate. The solution was washed with sodium bicarbonate, then water. The ethyl acetate solution was dried with anhydrous sodium sulfate. After evaporation of the solution *in vacuo*, an oily residue was obtained. This did not crystallize.

(±)-N-Benzoyl-β-hydroxyvaline methyl ester, (±)-(VIII), was obtained from IV by diazomethane. Yield, 92%, m.p. 105°C.

Found: C, 62.36; H, 6.75; N, 5.55. Calcd. for $C_{13}H_{7}NO_{4}$: C, 62.14; H, 6.82; N, 5.57%.

N-Benzoyl-β-chloro-valine methyl ester (IX) —— N-Benzoyl-β-hydroxyvaline methyl ester (VIII) (oil obtained above) was chilled in an ice bath and 1.2 ml. of thionyl chloride was added. This mixture was warmed in a water bath (about 40°C) to dissolve the oil. After 4 hr., 10 ml. of water was added to the reaction mixture to decompose the excess thionyl chloride. The mixture was extracted with ethyl acetate. The ethyl acetate layer was washed with water, aqueous sodium bicarbonate, and water. The ethyl acetate solution was dried with anhydrous sodium sulfate. After evaporation of ethyl acetate in vacuo, a yellow oil was obtained. This did not crystallize.

(±)-(IX) was obtained from (±)-(VIII) by treatment with thionyl chloride under the same conditions as above.

N-Benzoylvaline methyl ester (X) —— The yellow oil (IX) was dissolved in 50 ml. of 80% methanol. To this solution, 2.0 g. of sodium acetate, 2.0 ml. of acetic acid, and 2.0 g. of 5% palladium on charcoal were added and the hydrogenolysis was carried out at room temperature. When hydrogen uptake ceased, the catalyst was removed by filtration. The filtrate was concentrated in vacuo. This residue was dissolved in water and extracted with ethyl acetate. The ethyl acetate layer was washed with aqueous sodium bicarbonate and then with water.

The ethyl acetate layer was dried with anhydrous sodium sulfate. After evaporating the ethyl acetate in vacuo, 1.5 g. (63.8% based on VI) of N-benzoyl-valine methyl ester (X) was obtained. This was recrystallized from ethanol and ether. Yield, 1.3 g. (55% based on VI), m.p. 128°C. The specific rotation is too small to measure in alcohol or in chloroform. The direction of rotation seems to be slightly (+).

Found: C, 66.29; H, 7.33; N, 6.01. Calcd. for $C_{13}H_{17}NO_3$: C, 66.36; H, 7.28; N, 5.95%.

N-Benzoyl-DL-valine methyl ester, (±)-(X), was obtained from (±)-(IX). Yield, 64.2%, m.p. 86°C [based on (±)-(VIII)].

Found: C, 66.10; H, 7.23; N, 5.97%.

D(-) Valine (XI) —— The mixture of N-benzoylvaline methyl ester (X), 4.7 g. (0.02 mol.), and 5 N hydrochloric acid, 50 ml., was refluxed for 6 hr. After cooling, the precipitated benzoic acid was separated by filtration. The filtrate was extracted with chloroform (50 ml. x 2) to remove the residual benzoic acid. After evaporating the hydrochloric acid solution in vacuo, residual crystals were dissolved in a minimum amount of absolute alcohol at room temperature and pyridine was added to the solution. Crude D-(-) valine (XI), 1.78 g., was obtained. This was recrystallized from water and ethanol. Yield, 1.50 g. (65%), m.p. 310°C (sublimation, sealed capillary), $[\alpha]_D^{25}$ = -28.7° (c 2.92, 5 N HCl).[23]

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[23] L-Valine, $[\alpha]_D^{25}$ = +28.2° (5 N HCl). J. P. Greenstein and M. Winitz, "Chemistry of the Amino Acids", Vol. 3, p. 2368, John Wiley and Sons, Inc., New York, 1961.

Found: C, 51.05; H, 9.68; N, 12.01. Calcd. for $C_5H_{11}NO_2$: C, 51.26; H, 9.46; N, 11.96%.

In a similar way, DL-valine was obtained from (±)-(X). Yield, 75%. Found: C, 51.02; H, 9.47; N, 11.80%.

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