The Preparation of Optically Active α-*C*-Substituted Glutamic Acid

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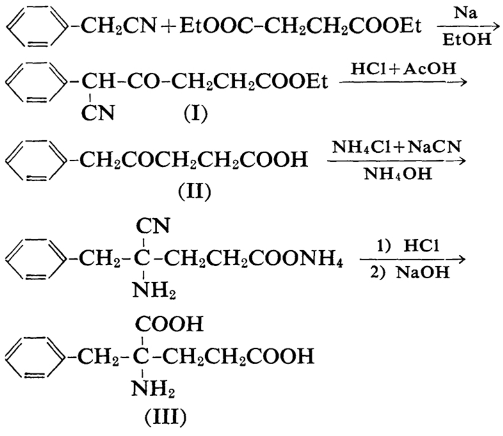
α-*C*-Dialkyl-substituted glycine is an interesting substance from the biochemical point of view, with regard to α-amino acid and also with ragard to the relation between its optical rotation and its absolute configuration. However, although several α-*C*-dialkyl-substituted glycines have been synthesized, only two have been resolved into the optically active isomers.[1,2]

In the present work, two optically active α-*C*-dialkyl-substituted glycines, 2-methyl and 2-benzylglutamic acid, were prepared. DL-2-Methylglutamic acid was synthesized from levulinic acid by Takenishi's procedure,[3] and then its optical resolution was performed. *N*-Benzoylated DL-2-methylglutamic acid was resolved with brucine by the method of the differential solubility of the isomers in water. The optical rotation values of the isomers were as follows; (+) isomer, $[\alpha]_D^{20}$ +12.1° (*c* 4, 6 N HCl); (−) isomer, $[\alpha]_D^{20}$ −12.1° (*c* 4, 6 N HCl).

DL-2-Benzylglutamic acid was prepared from benzyl cyanide and diethyl succinate by the following process:



The optical resolution of this amino acid was performed by means of a methanolic solution with strychinine. The optical rotation values of two optically active isomers of this amino acid were as follows:

(+) isomer, $[\alpha]_D^{20}$ +1.65° (*c* 10, 4 N HCl);
(−) isomer, $[\alpha]_D^{20}$ −1.60° (*c* 10, 4 N HCl).

Experimental

The Preparation of DL-2-Methylglutamic Acid.—DL-2-Methylglutamic acid was prepared from levulinic acid by Takenishi's procedure.[3]

The Preparation of *N*-Benzoyl-2-methylglutamic Acid.—One hundred and fifty milliliters of 2 N sodium hydroxide and 42.5 g. of benzoyl chloride in 100 ml. of dioxane were simultaneously stirred, drop by drop, into a solution of 48.3 g. of DL-2-methylglutamic acid in 600 ml. of N sodium hydroxide at 5—10°C; the addition took about 2 hr. Stirring was then continued for one more hour at the same temperature. The reaction mixture was washed with two 100 ml. portions of ethyl ether and concentrated to about 500 ml. in vacuo. This solution was then acidified with 100 ml. of concentrated hydrochloric acid, and stored for two days in an ice box. The crude material obtained was washed several times with boiling ligroin. The resulting crystals were dissolved in 230 ml. of methyl alcohol and filtered. The filtrate was stirred into 500 ml. of water, and the mixture was allowed to stand overnight in an ice box. White crystalline *N*-benzoyl-2-methylglutamic acid was collected, washed with cold water, and dried in air. m. p. 198°C. Yield, 50 g.

Found: C, 58.48; H, 5.66; N, 5.15. Calcd. for $C_{13}H_{15}NO_5$: C, 58.86; H, 5.70; N, 5.28%.

The Optical Resolution of *N*-Benzoyl-2-methylglutamic Acid.—After 69 g. of *N*-benzoyl-2-methylglutamic acid and 255 g. of brucine had been dissolved in 3.4 l. of boiling water, the solution was left to stand overnight in a refrigerator. The crystals were then collected, washed with cold water, and dried in air at room temperature. Two recrystallizations of this crude product from 1.4 parts of water gave 125 g. of pure *N*-benzoyl-2-methylglutamic acid brucine salt. One hundred and ten grams of the brucine salt were decomposed with 150 ml. of 2 N sodium hydroxide solution, and the brucine was removed by filtration. The alkaline solution was neutralized with hydrochloric acid and then evaporated to about 100 ml. in vacuo. This solution was boiled with 100 ml. of concentrated hydrochloric acid for 4 hr. and then left to stand overnight in a refrigerator. Benzoic acid was removed by filtration and extraction with 100 ml. of

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1) T. Kaneko, *J. Chem. Soc. Japan* (*Nippon Kwagaku Kwaishi*), **60**, 531 (1939).

2) C. G. Baker, S. C. J. Fu, S. M. Birnbaum, H. A. Sober and J. P. Greenstein, *J. Am. Chem. Soc.*, **74**, 4701 (1952).

3) T. Takenishi and O. Shimamura, This Bulletin, **27**, 207 (1954).

ethyl ether. The aqueous layer was evaporated to dryness in vacuo. To the residue a small amount of water was added, and the solution was evaporated to dryness in order to remove the free hydrochloric acid. Then the residue was taken up in 30 ml. of water, decolorized with charcoal, and adjusted to pH 3.2 with 20% sodium hydroxide. The mixture was stored overnight in a refrigerator, and the resulting crude crystals were collected, washed with 5 ml. of 50% methanol, and dried in air, yielding 7.1 g. of (+)-2-methylglutamic acid. On recrystallization from 10 ml. of boiling water, pure (+)-2-methylglutamic acid was obtained as fine needles. Yield, 6.3 g. $[\alpha]_D^{20}$ +12.1° (*c* 4, 6 N HCl).

Found: C, 44.99; H, 6.80; N, 8.60. Calcd. for $C_6H_{11}NO_4$: C, 44.71; H, 6.88; N, 8.69%.

The mother liquor of the *N*-benzoyl-(+)-2-methylglutamic acid brucine salt was concentrated to a paste. After it had stood for a week in a refrigerator, the brucine salt was collected. (−)-2-Methylglutamic acid was similarly obtained from the brucine salt by the procedure described above. Yield, 1.8 g. $[\alpha]_D^{20}$ −12.1° (*c* 4, 6 N HCl).

Found: C, 43.39; H, 6.77; N, 8.44%.

The Preparation of DL-2-Benzylglutamic Acid (III).—*Ethyl 5-Phenyl-5-cyanolevulinate (I).*—A mixture of 37.2 g. of freshly-distilled benzyl cyanide and 26.1 g. of diethyl succinate was added, with vigorous shaking, to 120 ml. of a sodium ethoxide solution containing 8.4 g. of sodium. The mixture was then refluxed for 2 hr., stored overnight at room temperature, poured into 350 ml. of cold water, and neutralized slowly with glacial acetic acid. An oily brown ester was separated, and the aqueous layer was extracted with ethyl ether. The ether layer was combined with the oily ester, washed twice with water, and dried over magnesium sulfate. Then the ether was evaporated off, and unreacted benzyl cyanide and diethyl succinate were removed by distillation in vacuo. The residual oily substance was allowed to stand for a few hours at −20°C. Brown crystals were thus obtained. Recrystallization from benzene - petroleum ether (1 : 1) gave white crystals. Yield, 54.8 g. M. p. 154°C. Betts and Davey[4] reported a m. p. of 152°C for this material.

Found: C, 68.05; H, 6.15; N, 5.69. Calcd. for $C_{14}H_{15}NO_3$: C, 68.55; H, 6.16; N, 5.71%.

5-Phenyllevulinic Acid (II).—Crude I (44 g.) was added to a mixture of glacial acetic acid (300 ml.), concentrated hydrochloric acid (100 ml.) and water (90 ml.), and the mixture was refluxed for 14.5 hr. Then it was evaporated to dryness in vacuo, and the residue was taken up in ether. The ether layer was washed twice with water and repeatedly extracted with a 5% potassium carbonate solution. The alkaline solution was then washed with ether and adjusted to pH 3—4 with 3 N hydrochloric acid. II was obtained as a brown oil. For the purification of crude II, this oil was dissolved in a small amount of water containing equimolar potassium hydroxide, and then a large quantity of acetone was added to the solution. After the mixture had stood in an ice box for 4 hr., the colorless crystalline potassium salt of II was collected. Recrystallization from water-acetone (1 : 4) gave 32 g. of the pure salt. This was dissolved in water, adjusted to pH 3—2 with concentrated hydrochloric acid, and extracted with ether. The ether layer was washed with water and dried over magnesium sulfate. When the ether was evaporated off, II was obtained as white crystals. Yield, 25 g. M. p. 57°C.

Found: C, 68.89; H, 6.29. Calcd. for $C_{11}H_{12}O_3$: C, 68.73; H, 6.29%.

DL-2-Benzylglutamic Acid (**III**).—After a solution containing 23 g. of II and 34 ml. of 12% aqueous ammonia had been cooled to 0—5°C in an ice water bath, 7.4 g. of sodium cyanide in 27 ml. of water and 8 g. of ammonium chloride in 27 ml. of water were added. The flask was tightly stoppered, and the mixture was shaken vigorously in a water bath at 50—70°C for 6 hr. Then the reaction mixture was cooled in an ice water bath and poured into 1.5 l. of concentrated hydrochloric acid; the mixture was allowed to stand overnight at room temperature before it was refluxed for 2 hr. The hydrolyzate was concentrated in vacuo, and during this procedure inorganic salts were removed by filtration. The residue was dissolved in a small amount of water and redried in vacuo in order to remove the free hydrochloric acid. It was taken up in a small amount of water, and the solution was decolorized, adjusted to pH 3.2 with dilute sodium hydroxide, and allowed to stand in a refrigerator. White crystals of III were collected, washed with cold water, and dried in air. Yield, 21 g. M. p. 214—215°C (decomp.).

Found: C, 60.71; H, 6.35; N, 5.88. Calcd. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90%.

The Preparation of *N*-Benzoyl-DL-2-benzylglutamic Acid.—*N*-Benzoyl-DL-2-benzylglutamic acid was prepared from 47.4 g. of III and 91 g. of benzoyl chloride using a procedure similar to that described for the preparation of *N*-benzoyl-DL-2-methylglutamic acid. Yield, 50 g. M. p. 193°C.

Found: C, 66.80; H, 5.61; N, 4.08. Calcd. for $C_{19}H_{19}NO_5$: C, 66.85; H, 5.61; N, 4.10%.

The Optical Resolution of *N*-Benzoyl-DL-2-benzylglutamic Acid.—Seventy-three grams of strychinine and 37 g. of *N*-benzoyl-DL-2-benzylglutamic acid were dissolved in 600 ml. of boiling methanol. The solution was then cooled to room temperature, 20 ml. of water was added with vigorous shaking, and the mixture was allowed to stand overnight in a refrigerator. The crystalline precipitate was collected and recrystallized twice from methanol. An optically pure *N*-benzoyl-2-benzylglutamic acid distrychinine salt was obtained. Yield, 44 g. M. p. 173—175°C, $[\alpha]_D^{20}$ +11.9° (*c* 1.2, methanol).

Forty-four grams of the distrychinine salt were decomposed with 270 ml. of 0.5 N sodium hydroxide at 5°C, and crystallized strychinine was removed. The alkaline solution was washed twice with 50 ml. of chloroform, and the aqueous layer was concentrated to half its volume and then acidified with 15 ml. of concentrated hydrochloric acid. The acidic solution was then left to stand overnight in an ice box, and 15 g. of *N*-benzoyl-(+)-2-benzylglutamic acid was obtained. $[\alpha]_D^{20}$ +52.6° (*c* 2.5, methanol). *N*-Benzoyl-(+)-2-benzylglutamic acid

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[4] B. E. Betts and W. Davey, *J. Chem. Soc.*, **1961**, 3336.

(14.7 g.) were hydrolyzed with 90 ml. of 20% hydrochloric acid for 4 hr. After standing overnight in a refrigerator, the crystallized benzoic acid was removed from the hydrolyzate and the filtrate was washed twice with ether. The solution was evaporated to drynesss in vacuo. The residue was dissolved in a small amount of water, and the solution was evaporated to dryness again in order to remove the free hydrochloric acid. To the residual syrup, 20 ml. of water was added. The resulting solution was decolorized with charcoal, neutralized with alkali, and allowed to stand in an ice box. The crude product was collected, and for purification it was converted to the hydrochloride and neutralized with sodium hydroxide. Crystals of optically pure (+)-2-benzylglutamic acid were collected, washed with methanol-water, and dried. Yield, 6.0 g. M. p. 209°C, $[\alpha]^{20}_{\mathrm{D}}$ +1.65° (*c* 10, 4 N HCl).

Found: C, 60.67; H, 6.28; N, 5.86. Calcd. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90%.

When the mother liquor of the *N*-benzoyl-(+)-2-benzylglutamic acid distrychinine salt was treated in the manner described above, crystals of (−)-2-benzylglutamic acid were obtained. Yield, 3.2 g. M. p. 208°C, $[\alpha]^{20}_{\mathrm{D}}$ +1.60° (*c* 10, 4 N HCl).

Found: C, 60.70; H, 6.33; N, 5.83. Calcd. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90%.

Summary

1) The optical resolution of DL-2-methylglutamic acid has been performed using brucine as a resolving reagent; the two optically active isomers have been obtained.

2) DL-2-Benzylglutamic acid has been synthesized using benzyl cyanide and diethyl succinate as the starting materials. Moreover, the optical resolution of this amino acid has been performed using strychinine as a resolving reagent. The two optically active isomers were obtained.

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