Organic Syntheses

A Publication
of Reliable Methods
for the Preparation
of Organic Compounds

Working with Hazardous Chemicals

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These paragraphs were added in September 2014. The statements above do not supersede any specific hazard caution notes and safety instructions included in the procedure.

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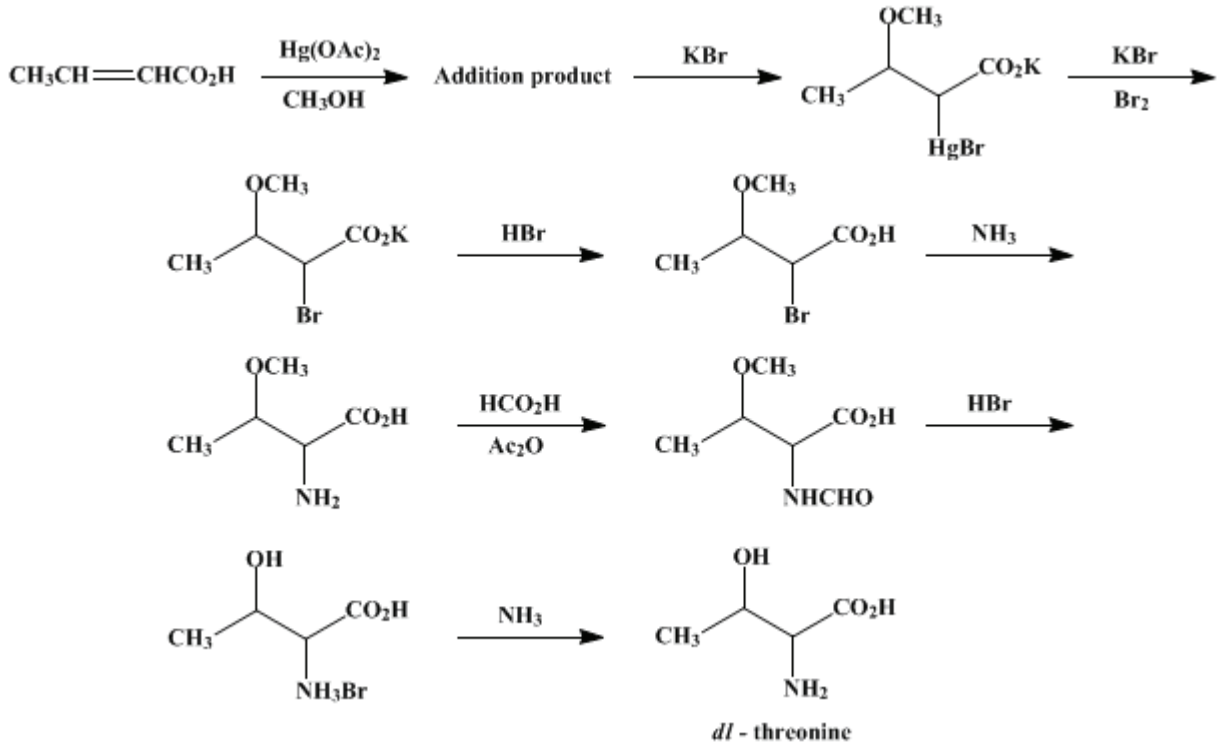
dl-THREONINE



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1. Procedure

A. *α-Bromo-β-methoxy-n-butyric acid.* In a 5-l. flask are placed 3 l. of methanol, 640 g. (2 moles) of mercuric acetate (Note 1), and 172 g. (2 moles) of crotonic acid (Note 2). The flask is warmed on a steam cone and shaken vigorously until the mercuric acetate dissolves (about 10 minutes). The solution is allowed to stand at room temperature for 48 hours (Note 3), then the precipitate is filtered, washed twice with 300-ml. portions of methanol, and air-dried. The yield is 625–650 g. (Note 4). This material is powdered and dissolved in a solution of 360 g. (3 moles) of potassium bromide in 2 l. of water, and the solution is placed in a 4-l. beaker, which is cooled in an ice bath and exposed to direct sunlight (Note 5). A solution of 320 g. (2 moles) of bromine and 360 g. (3 moles) of potassium bromide in 600 ml. of water is added with stirring during 20–40 minutes, a large excess of bromine being avoided. After 10–15 minutes' standing, any excess bromine is destroyed with sodium bisulfite. The bromo acids are isolated as follows: The solution is extracted once with 300 ml. of ether to remove a small amount of lachrymatory material and, after acidification with 400 ml. of 40% hydrobromic acid, is extracted again with six 800-ml. portions of ether. The ether extracts are combined, washed once with a small volume of cold water, and dried over anhydrous sodium sulfate. The ether is removed by distillation, leaving the crude bromo acid mixture.

The yield of crude bromo acids is 350–370 g. (88–93% based on the crotonic acid used in the first step). This material is used without purification in the preparation of aminomethoxybutyric acid.

The crude bromo acids may be freed from impurities by fractionation under reduced pressure. The fraction distilling below 125°/10 mm. (105°/3 mm.) is discarded; the remainder distils at 125–128°/10 mm. (105–107°/3 mm.) and consists of a mixture of stereoisomeric acids. The yield is 75–85% based on the crotonic acid used in the first step.

B. *dl-Threonine.* One hundred and seventy-five grams of crude bromomethoxybutyric acid is heated with 2 l. of concentrated ammonium hydroxide for 6 hours at 90–100° in a glass-lined autoclave (p. 777) (Note 6), The solution is concentrated to a thick gum under reduced pressure (Note 7), water is added, and the solution is reconcentrated under reduced pressure. The residue is allowed to stand under acetone with frequent shaking (Note 8) until the material has crystallized completely (1–2 days). The acetone is decanted, and the residue dissolved in 1 l. of 85–90% formic acid (Note 9). The solution is warmed to 45°, and 350 g. (330 ml.) of acetic anhydride is added with stirring during 10 minutes. The heat of reaction causes the temperature of the solution to rise to 70–80°, and the temperature of the mixture is maintained within this range for about 15 minutes. The solution is next evaporated to dryness under reduced pressure. The residue is dissolved, while being warmed on the steam bath, in the minimum amount of water (Note 10), and the solution is cooled overnight in the icebox. The crystals are filtered and air-dried. This material is a mixture of formyl derivatives (Note 11). One recrystallization from 150 ml. of hot water yields about 25 g. of practically pure formyl-*dl*-O-methylthreonine melting at 174–176°. An additional 3–5 g. is obtained by working up the filtrates. The yield is 25% (Note 12).

Twenty-five grams (0.16 mole) of formyl-*dl*-O-methylthreonine is refluxed for 2 hours with 360 ml. of constant-boiling hydrobromic acid. The solution is concentrated under reduced pressure (Note 13). Sufficient water is added to dissolve all the residue, and the solution is reconcentrated under reduced pressure. The gummy residue is next dissolved in 450 ml. of absolute ethanol, and concentrated ammonium hydroxide is added until the odor of ammonia persists after vigorous shaking. The solution is cooled in the icebox overnight, and the crystals are filtered and dissolved in 3 volumes of hot water (about 5 ml. of water per gram of crude material). Seven volumes of absolute ethanol are added, and the solution is cooled to room temperature with scratching of the flask to induce crystallization. It is then cooled in an icebox overnight. The crystals are filtered and washed twice with 90-ml. portions of absolute ethanol and once with ether. The yield is 18–20 g. (85–90% based on the formyl-*dl*-O-methylthreonine) of pure *dl*-threonine, melting with decomposition at 234–235° (Note 14).

2. Notes

1. The mercuric acetate may be replaced by equivalent amounts of mercuric oxide and glacial acetic acid.
2. The crotonic acid was obtained from the Niacet Chemical Company and used without purification.
3. It is advisable to scratch the flask with a glass rod after 3–4 hours. This usually initiates precipitation of the addition product in a finely divided form. If this is not done, the addition product may crystallize slowly on the sides of the flask in a cake which is removed only with the greatest difficulty. It is also advantageous to stir the mixture mechanically for several hours after crystallization begins in order to prevent caking.
4. The exact yield cannot be calculated, since the structure of the addition product is unknown. The yield is almost quantitative, however, since only a small amount of mercury remains in the filtrate.
5. The bromination can be carried out equally successfully under the illumination of two No. 2 Photoflood lamps in suitable reflectors placed directly above the surface of the liquid. Under these conditions, however, the addition of the bromine requires 10–15 minutes longer.
6. According to the submitters the amination can be carried out in ordinary 500-ml. glass bottles if the temperature does not exceed 85°. The time of heating should then be extended to 8–10 hours.
7. All the concentrations under reduced pressure required in this preparation may be carried out at the pressure provided by an efficient water pump.
8. The material, if allowed to stand without shaking, solidifies to a hard cake. The shaking furthers extraction of certain gummy impurities which interfere with the separation to be carried out later.
9. If a mixture of *dl*-threonine and *dl*-allothreonine is desired instead of *dl*-threonine alone, the residue may be dissolved directly in 1.2 l. of 48% hydrobromic acid and refluxed for 2 hours. After removal of the hydrobromic acid under reduced pressure, the gummy residue is dissolved in warm water, and concentrated ammonium hydroxide is added slowly until a faint odor of ammonia persists after vigorous shaking. The solution is concentrated until crystals appear, and 3–4 volumes of ethanol is added. The acids are recrystallized by dissolving in the minimum amount of hot water (4–5 ml. per gram) and adding 4–5 volumes of ethanol. The solution is allowed to cool and permitted to stand overnight at room temperature.
10. If, after solution is complete on the steam bath, 10% more water is added, the quality of the product

is better, but the yield is slightly less.
11. Formyl-*dl*-O-methylthreonine melts at 173–174°. Formyl-*dl*-O-methylallothreonine melts at 152–153°.
12. Crude *dl*-allothreonine may be obtained from the mother liquors by concentrating them to dryness, refluxing the residue with 10 volumes of 48% hydrobromic acid, and working up the solution as described for *dl*-threonine. The product contains a small amount of *dl*-threonine which can be largely removed by three or four recrystallizations from 50% ethanol. *dl*-Allothreonine of this purity melts at 242–243°.
13. Constant-boiling hydrobromic acid can be recovered by fractionating the distillate at atmospheric pressure.
14. The melting points obtained by the checkers were consistently 3–4° above those given. The melting points of these compounds vary with the method of determination.

3. Discussion

α-Amino-β-hydroxybutyric acid has been prepared by a procedure similar to the one described, using ethyl crotonate as the starting material.[1] A mixture of the α-amino-β-hydroxy- and α-hydroxy-β-aminobutyric acids has been secured by treating crotonic acid with hypochlorous acid and heating the resulting product with dry ammonia under pressure.[2] A mixture containing threonine has been obtained by treatment of acetoacetic ester with sodium nitrite and acetic acid; the resultant ethyl oximinoacetoacetate was then converted by means of diethyl sulfate into ethyl O-ethyloximinoacetoacetate. This product was reduced by hydrogen and Raney nickel to an impure ethyl α-amino-β-hydroxybutyrate, which was then hydrolyzed to a mixture of *dl*-threonine and *dl*-allothreonine.[3] The method described has been published.[4]

Improvements in this method have been reported,[5] and an excellent synthesis from ethyl acetamidoacetoacetate has been described.[6]

Interconversion methods for obtaining *dl*-threonine from *dl*-allothreonine via the azlactone[7] or the oxazoline[6] have been reported.

References and Notes

1. Abderhalden and Heyns, *Ber.*, **67**, 530 (1934).
2. Burch, *J. Chem. Soc.*, **137**, 310 (1930).
3. Adkins and Reeve, *J. Am. Chem. Soc.*, **60**, 1330 (1938).
4. West and Carter, *J. Biol. Chem.*, **119**, 109 (1937).
5. Pfister, Howe, Robinson, Shabica, Pietrusza, and Tishler, *J. Am. Chem. Soc.*, **71**, 1096 (1949).
6. Pfister, Robinson, Shabica, and Tishler, *J. Am. Chem. Soc.*, **71**, 1101 (1949).
7. Carter, Handler, and Melville, *J. Biol. Chem.*, **129**, 362 (1939).

Appendix
Chemical Abstracts Nomenclature (Collective Index Number); (Registry Number)

acetoacetic ester

α-amino-β-hydroxy- and α-hydroxy-β-aminobutyric acids

ethanol (64-17-5)

acetic acid (64-19-7)

ammonia (7664-41-7)

methanol (67-56-1)

ether (60-29-7)

acetic anhydride (108-24-7)

hydrogen (1333-74-0)

HYDROBROMIC ACID (10035-10-6)

bromine (7726-95-6)

sodium sulfate (7757-82-6)

formic acid (64-18-6)

sodium nitrite (7632-00-0)

sodium bisulfite (7631-90-5)

mercuric acetate (1600-27-7)

mercury (7439-97-6)

mercuric oxide (21908-53-2)

Raney nickel (7440-02-0)

acetone (67-64-1)

potassium bromide (7758-02-3)

hypochlorous acid (7790-92-3)

ammonium hydroxide (1336-21-6)

diethyl sulfate (64-67-5)

crotonic acid (3724-65-0)

ethyl crotonate (623-70-1)

aminomethoxybutyric acid

bromomethoxybutyric acid

α-Amino-β-hydroxybutyric acid

threonine,

DL-Threonine (72-19-5)

ethyl oximinoacetoacetate

ethyl O-ethyloximinoacetoacetate

ethyl α-amino-β-hydroxybutyrate

ethyl acetamidoacetoacetate

oxazoline

formyl-dl-O-methylthreonine

dl-allothreonine (28954-12-3)

Formyl-dl-O-methylallothreonine

α-Bromo-β-methoxy-n-butyric acid (26839-91-8)