Synthesis of Carolic Acid

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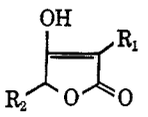
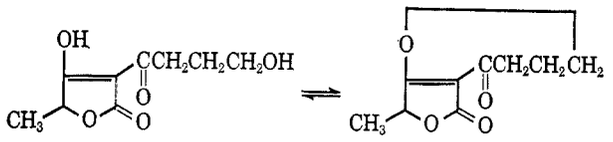
The total synthesis of carolic acid, α-(γ'-hydroxybutyryl)-γ-methyltetronic acid anhydride (**1**), has been described. Ethyl ε-benzyloxy-β-ketocaproate, which is the component of the carolic acid side chain, was prepared by condensation of diethyl ethoxymagnesium malonate and mixed γ-benzyloxybutyric–ethyl carbonic anhydride and by following decarboxylation. Treatment of the product with acetyllactyl chloride gave α-(γ-benzyloxybutyryl)-γ-methyltetronic acid, which was converted to **1** by selective reduction.

Carolic acid (**1**, anhydride of **1a**), carolinic acid (**1b**), carlic acid (**1c**), and carlosic acid (**1d**) were isolated in 1934 from cultures of *Penicillium charlesii G. Smith*,[1] and the structures of these acids have been suggested as **1a**, **1b**, **1c**, and **1d**, respectively.[2] They have a unique structural relationship to ascorbic acid (**1e**).



1a, $R_1 = COCH_2CH_2CH_2OH$; $R_2 = CH_3$
b, $R_1 = COCH_2CH_2COOH$; $R_2 = CH_3$
c, $R_1 = COCH_2CH_2CH_2OH$; $R_2 = CH_2COOH$
d, $R_1 = COCH_2CH_2CH_3$; $R_2 = CH_2COOH$
e, $R_1 = OH$; $R_2 = CHOHCH_2OH$

It was postulated that **1a** crystallized with loss of water and the crystalline compound should possess structure **1**.[3]

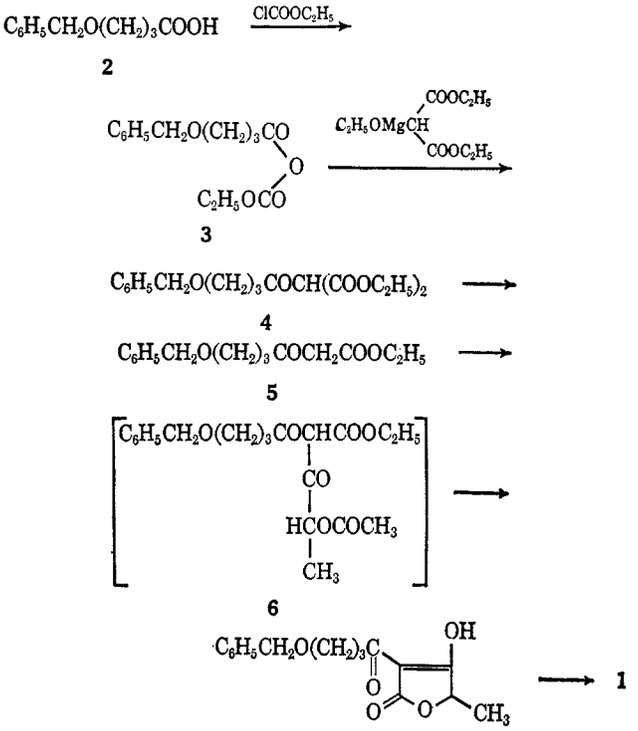


1a **1**

Of these products, only **1b** has been synthesized by Haynes and co-workers.[4] One difficulty in the synthesis of **1a** or **1c** arose from the presence of a hydroxy group in the 4 position of the side chain, because condensation of γ-bromobutyryl chloride with ethoxymagnesium malonic ester gave diethyl tetrahydro-2-furylidene malonate,[4] instead of the expected acyl malonate. To avoid this side reaction, we attempted to protect the hydroxy group of carolic acid side chain with a benzyl group, and succeeded in the total synthesis of carolic acid according to Scheme I.

The preparation of γ-benzyloxybutyric acid (**2**) had been reported,[5,6] but the method was tedious and the yields were low in our hands; so we prepared it in 40% yield by fusion of γ-butyrolactone with sodium benzylate,[7] followed by neutralization with acid. Compound **2** was converted by treatment with ethyl chlorocarbonate according to the method of Tarbell and Price[8] into the mixed γ-benzyloxybutyric ethylcarbonic

SCHEME I



anhydride (**3**), which was treated with diethyl ethoxymagnesium malonate to give diethyl γ-benzyloxybutyrylmalonate (**4**). In an attempt to obtain **4** by condensation of ethoxymagnesium or sodium malonate with the acid chloride, γ-benzyloxybutyric acid (**2**) was treated with thionyl chloride. However, the treatment gave γ-butyrolactone and benzyl chloride. These compounds seem to be formed by decomposition of the cyclic oxonium intermediate (**9**) derived from the initially produced acid chloride (**8**). This mechanism is supported by the fact that γ-methoxybutyryl chloride (**10**) on heating was transformed into methyl γ-chlorobutyrate (**12**) through the oxonium intermediate (**11**)[9] (Scheme II).

When **4** was boiled in dilute acetic acid, hydrolysis was followed or accompanied by decarboxylation, and ethyl ε-benzyloxy-β-ketocaproate (**5**) was obtained.[10] Compound **4** was the key compound in this synthesis and could be lactonized with α-acetoxyacetyl chloride derivatives to tetronic acid derivatives according to the method of Haynes and co-workers.[4] The ethoxymagnesium salt of **5** was treated with α-acetoxypro-

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(2) P. W. Clutterbuck, H. Raistrick, and F. Reuter, *ibid.*, **29**, 300 (1935); **29**, 871 (1935).
(3) J. R. Plimmer, *J. Org. Chem.*, **29**, 2511 (1964).
(4) L. J. Haynes, J. R. Plimmer, and A. H. Stanners, *J. Chem. Soc.*, 4661 (1956).
(5) G. M. Bennett and A. L. Hock, *ibid.*, **472** (1927).
(6) T. Haga, *J. Chem. Soc. Japan*, **81**, 1113 (1960).
(7) W. Reppe, *Ann.*, **596**, 163 (1955).
(8) D. S. Tarbell and J. A. Price, *J. Org. Chem.*, **22**, 245 (1957).
(9) F. F. Blicke, W. B. Wright, and M. F. Zienty, *J. Am. Chem. Soc.*, **63**, 2488 (1941).
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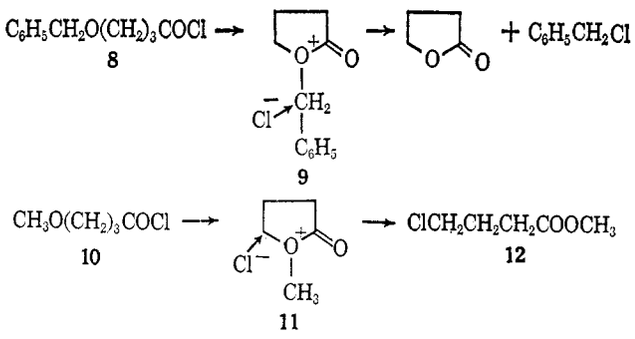
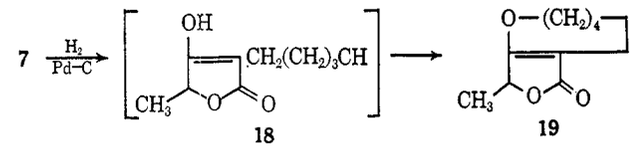
SCHEME II

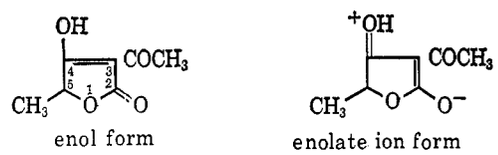


pionyl chloride to give ethyl α-(α-acetoxypropionyl)-β-keto-γ-benzyloxycaproate (**6**), which was saponified by dissolving in aqueous sodium hydroxide followed by acidifying with dilute sulfuric acid under cooling to afford α-(γ'-benzyloxybutyryl)-γ-methyltetronic acid (**7**). The product **7** gave an orange precipitate with aqueous ferric chloride and its infrared spectrum showed sharp bands characteristic for α-acyltetronic acid at 1605, 1660, 1690, and 1765 cm^{-1} [11] (liquid film). Similarly α-acetyl-γ-phenyltetronic acid (**13**) and α-acetyl-γ-methyltetronic acid (**14**) were obtained.

Hydrogenation of α-acetyltetronic acid (**15**) with palladium–charcoal catalyst is reported to yield α-ethyltetronic acid (**16**).[12] In our study, α-acetyl-γ-methyltetronic acid (**14**) absorbed 2 mole equiv of hydrogen with palladium–charcoal catalyst to give α-ethyl-γ-methyltetronic acid (**17**). The hydrogenation of **7** under similar conditions was therefore expected to cause not only debenzylation but also transformation of the carbonyl group of the side chain to a methylene group. In fact when **7** was reduced in methanolic solution, 3 mole equiv of hydrogen was absorbed. The oily product (**18**) showed the characteristic infrared spectrum of an α-alkyltetronic acid at 1650 and 1720 cm^{-1} (liquid film) and was converted on distillation to a solid, mp 54° ($C_9H_{12}O_3$). This product may be identical with the reduced product of carolic acid anhydride, for which a melting point of 45° was recorded by Clutterbuck and co-workers.[2]



The ultraviolet absorption spectrum of **14** in methanolic solution exhibited two absorption bands [λ_{max}^{EtOH} 232 mμ (ε 7130) and λ_{max}^{EtOH} 267 mμ (ε 13,400)] the former being attributed to the enol form and the latter to the enolate ion form.[13,14] However in methanolic solution containing a little hydrochloric acid, only the enolate absorption band appeared at 263 mμ (ε 11,200).



enol form enolate ion form

The structural difference depending on pH might be expected to cause some effect in this reduction course, because the conjugation of the C=O double bond of the acyl group with the 2,3-C=C double bond would differ from that with the 3,4-C=C double bond, owing to the presence of an additional oxygen in β position of this α,β-unsaturated carbonyl conjugation system. When **14** was reduced with palladium–charcoal catalyst in methanolic solution containing a little hydrochloric acid, no hydrogen was absorbed and **14** was recovered. When **7** was reduced under similar conditions, 1 mole of hydrogen was absorbed and α-(γ-hydroxybutyryl)-γ-methyltetronic acid anhydride (carolic acid, **1**), mp 113°, was formed. It gave an orange color with aqueous ferric chloride and had infrared sharp bands at 1590, 1695, and 1750 cm^{-1} (liquid film); nmr signals (in $CDCl_3$ with TMS as internal reference) appeared at δ = 1.45 ppm, a doublet; δ = 2.25 ppm, a multiplet; δ = 3.40 ppm, a triplet; δ = 4.62 ppm, a quartet; and δ = 4.73 ppm, a triplet of λ_{max}^{EtOH} 273 mμ (ε 18,900). The properties of the synthetic material were essentially identical with those of the natural product.

Experimental Section

γ-Benzyloxybutyric Acid (**2**).—To a suspension of 46 g (2 g-atoms) of sodium in 30 ml of xylene was added 216 g (2 moles) of benzyl alcohol diluted with 500 ml of dry toluene. After completion of the addition, the mixture was refluxed to complete the reaction, 172 g (2 moles) of γ-butyrolactone was then added, and the mixture was heated gradually up to 220°. During this time the solvent evaporated and the residue was fused for 30 min at 220°. After cooling, the crude sodium salt was dissolved in 500 ml of water. The solution was washed with 200 ml of ether and then acidified with 120 g of acetic acid. The oily layer was separated and extracted with four portions of 200 ml of ether. The combined ether solution was dried over anhydrous sodium sulfate and filtered, and the solvent was removed. The residue was distilled under reduced pressure to give **2**: 140 g (40%), bp 150° (2.5 mm). *Anal.* Calcd for $C_{11}H_{14}O_3$: C, 68.02; H, 7.27. Found: C, 68.28; H, 7.47.

Diethyl γ-Benzyloxybutyrylmalonate (**4**).—To the mixture of 97 g (0.5 mole) of γ-benzyloxybutyric acid and 50.5 g (0.5 mole) of triethylamine, dissolved in 500 ml of dry toluene, was added 54.3 g (0.5 mole) of ethyl chlorocarbonate under cooling at such a rate that the temperature did not rise above 0°. Triethylamine hydrochloride precipitated both during the addition and while the mixture was stirred for 15–25 min thereafter. To this solution was added an ether solution of diethyl ethoxymagnesium malonate prepared according to the method of Lund,[15] with stirring as the temperature was held at −5 to 0°. After the mixture had been allowed to stand over night and come to room temperature, it was acidified cautiously with 400 ml of 5% sulfuric acid. The aqueous solution was separated and extracted once with ether. The two organic layers were combined, washed once with dilute sulfuric acid and with a saturated sodium bicarbonate solution, and then with water, and dried over anhydrous sodium sulfate. The solvent was removed and the residue was distilled under reduced pressure to give **4**: 109 g (65%), bp 175–180° (bath temperature, 0.05 mm). *Anal.* Calcd for $C_{18}H_{24}O_6$: C, 64.27; H, 71.9. Found: C, 64.40; H, 7.27.

Ethyl ε-Benzyloxy-β-ketocaproate (**5**).—Ethyl γ-benzyloxy-β-ketobutyrylmalonate (84 g, 0.25 mole) was boiled with 1000 ml of 2 *N* acetic acid for 6 hr. Carbon dioxide evolved during this time. After cooling, the organic layer was extracted with four portions (200-ml each) of ether. The ether solution was washed once with a saturated sodium bicarbonate solution and then with water and dried over anhydrous sodium sulfate. The ether was removed and the residue was distilled under reduced pressure

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(14) J. R. Plimmer and L. J. Haynes, unpublished observation. We are grateful to a referee for pointing this out.
(15) H. Lund, *Ber.*, **67**, 935 (1934).

to give **5**: 49.5 g (75%), bp 143.5° (1 mm). *Anal.* Calcd for $C_{15}H_{20}O_4$: C, 68.16; H, 7.63. Found: C, 68.41; H, 7.76.

Carolic Acid (1).—A solution of 50 g of ethyl ε-benzyloxy-β-ketocaproate in 20 ml of absolute alcohol, 1 ml of carbon tetrachloride, and 100 ml of dry ether was added to 4.7 g of magnesium. After all of the magnesium had dissolved the solvents were removed by distillation under reduced pressure. The residue was dissolved in 60 ml of dry ether and to this solution was added 30 g of acetyllactyl chloride[16] diluted with 30 ml of dry ether. The reaction mixture was set aside overnight and then treated cautiously with 100 ml of 5% sulfuric acid under cooling with an ice–salt mixture. The ether layer was separated and the water layer was washed with 50 ml of ether. The combined ether solution was washed once with dilute sulfuric acid and then with water, dried, and concentrated. The residue was dissolved in 200 ml of 4 *N* sodium hydroxide and set aside for 24 hr. Addition of an excess of 5% sulfuric acid under cooling separated 26 g of an oily substance (**7**) which was extracted with ether. The product could not be purified by either distillation or crystallization and was used directly for the following reaction.

The crude product (**7**, 2.9 g, 0.01 mole) was reduced at atmospheric pressure with 1 g of 10% palladium–charcoal catalyst in 100 ml of methanol containing 0.5 ml of hydrochloric acid at room temperature (about 240 ml of hydrogen was absorbed). After absorption had ceased the solvent was removed and the residue was recrystallized from ethanol to give 1.6 g of **1** as needles, mp 113°. *Anal.* Calcd for $C_9H_{10}O_4$: C, 59.33; H, 5.53. Found: C, 59.32; H, 5.58.

Reduced Product of Carolic Acid Anhydride (19).—Crude **7** (2.9 g, 0.01 mole) was reduced with 1 g of 10% of palladium–charcoal catalyst at atmospheric pressure in 100 ml of methanol at room temperature (about 720 ml of hydrogen was absorbed). After absorption had ceased the solvent was removed and the residue was heated under reduced pressure. The solid product was recrystallized from ether to give 0.8 g of white crystals, mp 54°. *Anal.* Calcd for $C_9H_{12}O_3$: C, 64.27; H, 7.19. Found: C, 64.01 H, 7.45.

α-Acetyl-γ-phenyltetronic acid (13) was prepared in a similar manner from ethyl acetoacetate and acetylmandelyl chloride in 65% yield and was recrystallized from petroleum ether (bp 104.5°). It melted at 104.5°. The infrared spectrum (KBr) of this compound exhibited sharp bands at 1600, 1660, and 1745 cm^{-1}. *Anal.* Calcd for $C_{12}H_{12}O_4$: C, 66.05; H, 4.62. Found: C, 66.09; H, 4.62.

α-Acetyl-γ-methyltetronic acid (14) was prepared similarly from ethyl acetoacetate and α-acetoxypropionyl chloride in 60% yield and was recrystallized from petroleum ether. This compound melted at 55° and exhibited the sharp infrared bands (KBr) at 1625, 1665, and 1755 cm^{-1}. *Anal.* Calcd for $C_7H_8O_4$: C, 53.84; H, 5.16. Found: C, 53.94; H, 5.37.

α-Ethyl-γ-methyltetronic Acid (17).—A solution of 1.56 g (0.01 mole) of **14** in 100 ml of methanol was reduced with 1 g of 10% palladium–charcoal catalyst at atmospheric pressure and room temperature (about 480 ml of hydrogen was absorbed). After absorption had ceased the solvent was removed to give 1.3 g of **17** which was recrystallized from petroleum ether to give white needles, mp 81.5°. The infrared spectrum (KBr) of this compound exhibited sharp bands at 1655 and 1725 cm^{-1}. *Anal.* Calcd for $C_7H_{10}O_3$: C, 59.14; H, 7.09. Found: C, 58.92; H, 7.13.

Reaction of γ-Benzyloxybutyric Acid with Thionyl Chloride.—To a mixture of **2** (19.4 g, 0.1 mole) and 30 ml of petroleum ether was added 23.8 g (0.2 mole) of thionyl chloride at room temperature. After an evolution of gas had ceased, solvent and excess thionyl chloride were distilled under reduced pressure and then the residue was distilled under reduced pressure to yield 8.0 g of benzyl chloride, bp 41° (3.0 mm), and 6.0 g of γ-butyrolactone, bp 56° (3.0 mm). The former was characterized by its isothiourea hydrochloride, mp 148°, and the latter was characterized by its infrared sharp bands at 1770 cm^{-1} (liquid film). *Anal.* Calcd for C_7H_7Cl: C, 66.42; H, 5.57. Found: C, 66.30; H, 5.50. Calcd for $C_4H_6O_2$: C, 55.80; H, 7.03. Found: C, 55.91; H, 6.95.

Registry No.—**1**, 485-40-5; **2**, 10385-30-5; **4**, 10385-31-6; **5**, 10378-08-2; γ-butyrolactone, 96-48-0; benzyl chloride, 100-44-7; isothiourea hydrochloride, 538-28-3; **13**, 10385-32-7; **14**, 10385-33-8; **17**, 10385-34-9; **19**, 10385-35-0.

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Synthesis of Epimeric 3-Ureido-Δ⁴-Androsten-17-ones[1]

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3β-Ureido-Δ⁴-androsten-17-one was prepared from 3-oximino-Δ⁴-androsten-17-one ethylene ketal by zinc reduction to 3β-amino-Δ⁴-androsten-17-one followed by carbamylation of the amine with silicon tetraisocyanate or nitrourea. 3α-Ureido-Δ⁴-androsten-17-one was formed stereoselectively from 3-hydroxy-Δ⁴-androsten-17-one ethylene ketal and urea. The corresponding 17β-hydroxy derivatives were prepared. The orientation of the ureido group was proved by chemical and physical methods.

Recently a nitrogen-containing steroid was isolated following administration of 11β-hydroxy-Δ⁴-androstene-3,17-dione to man.[3] The compound, ureasterone (I), was characterized as 3α-ureido-11β-hydroxy-Δ⁴-androsten-17-one. It was subsequently demonstrated that ureasterone could be readily synthesized[4] stereoselectively from both 3α,11β- and 3β,11β-dihydroxy-Δ⁴-androsten-17-one and urea in aqueous acetic acid solution at 50°. Since the α orientation of the ureido group in ureasterone (I) was assigned principally from nmr studies, a stereospecific synthesis of 3β-ureido-Δ⁴-androsten-17-one (IIa) was undertaken to compare it with the product formed from the 3-hydroxy Δ⁴-steroids and urea. This paper reports the synthesis and reactions of the epimeric 3-ureido Δ⁴-steroids.

In 1956, Joska and Šorm[5] described the reduction of testosterone oxime with zinc dust in ethanolic acetic acid to 3-amino-Δ⁴-androsten-17β-ol (IIIa), but they did not assign the orientation of the 3-amino group. In the present study this reduction was repeated and the product was shown to be the 3β-amino epimer from its nmr spectrum. This stereoselective method was therefore employed for the synthesis of 3β-amino-Δ⁴-androsten-17-one (VIa) which was carbamylated to yield the desired 3β-ureido-Δ⁴-androsten-17-one (IIa). In order to retain the 17-ketone in the synthesis, this group was protected by ketalization in the starting material, Δ⁴-androstene-3,17-dione 17-ethylene ketal

(1) This work was supported by a research grant from the American Cancer Society and by a grant (CA 07304) from the National Cancer Institute, National Institutes of Health.

(2) Visiting Scientist, 1965–1966.

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