ml. of water and 50 ml. of petroleum ether; 5.24 g. (85%) of 3-chlorosulfonyl-4'-hydroxybenzanilide; m.p. 170°.

1-Phenyl-3-*m*-nitrobenzamido-5-pyrazolone.—Ten grams (0.057 mole) of 1-phenyl-3-amino-5-pyrazolone, prepared by the procedure of Weissberger and Porter,[5] was stirred on the steam-bath with 10.6 g. (0.057 mole) of *m*-nitrobenzoyl chloride and 7 ml. of ethyl oxalate for 20 minutes. The product was broken up by refluxing and stirring with 50 ml. of ethyl alcohol, cooled, collected, and washed on the funnel with 50 ml. of 70% alcohol; 14.5 g. (78%); m.p. 215–220°.

1-Phenyl-3-*m*-aminobenzamido-5-pyrazolone (VI).—Sixteen grams (0.049 mole) of 1-phenyl-3-*m*-nitrobenzamido-5-pyrazolone was added to 145 ml. of glacial acetic acid, 15 ml. of water, and 80 ml. of ethyl alcohol which was refluxed in a 1-l. round-bottomed flask, and followed by 16.0 g. of iron powder, added at once. A vigorous reaction took place, no heating being necessary for about three minutes. The mixture was kept boiling for ten minutes, filtered hot, and the filtrate diluted with 100 ml. of water, cooled without stirring, collected, and washed successively on the funnel with 50 ml. of 50% acetic acid, 150 ml. of water and 50 ml. of alcohol; 11.0 g. (76%) lustrous brown crystals (VI); m.p. 220–222°.

(5) A. Weissberger and H. D. Porter, THIS JOURNAL, **64**, 2133 (1942).

1-Phenyl-3-[3'-(*m*-chlorosulfonyl)-benzamido]-benzamido-5-pyrazolone (VII).—To 2.39 g. (0.01 mole) of I in 25 ml. of glacial acetic acid was added 2.94 g. (0.01 mole) of 1-phenyl-3-*m*-aminobenzamido-5-pyrazolone and 1.64 g. (0.02 mole) of sodium acetate in 55 ml. of warm (50°) glacial acetic acid. The mixture was stirred at room temperature for three hours (a white solid began to separate after approximately one minute), collected, and washed on the funnel with 100 ml. of water, stirred into 300 ml. of water, refiltered, and washed successively with 10 ml. of ethanol and 25 ml. of ether; 5.2 g. (94%) white crystals; m.p. 188–190°. Analysis showed the presence of one mole of acetic acid.

Anal. Calcd. for $C_{25}H_{21}ClN_4O_7S$: C, 54.0; H, 3.8; Cl, 6.4. Found: C, 54.3; H, 4.0; Cl, 6.0.

1-(3'-Chlorosulfonylbenzamido)-anthraquinone (IVd).—To a hot mixture of 4.4 g. (0.02 mole) of 1-aminoanthraquinone in 130 ml. of dry toluene was added, all at once, 6.6 g. (0.028 mole) of I. As the mixture was refluxed for one and one-quarter hours, hydrogen chloride evolved and solution resulted. The solution was cooled to room temperature, and collected in long amber needles; 6.9 g. (81%); m.p. 216–219° (dec.); recrystallized from 180 ml. of toluene; 5.0 g. (59%); m.p. 219–222° (dec.).

Anal. Calcd. for $C_{21}H_{12}ClNO_5S$: Cl, 8.35. Found: Cl, 8.36.

ROCHESTER, N. Y. RECEIVED FEBRUARY 9, 1951

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF HARVARD UNIVERSITY AND THE RESEARCH LABORATORIES OF MERCK & CO., INC.]

The Conversion of Cholic Acid into 3α-Hydroxy-12-keto-Δ$^{9(11)}$-cholenic Acid

BY LOUIS F. FIESER AND SRINIVASA RAJAGOPALAN, EVELYN WILSON AND MAX TISHLER

Methyl 3α,7α-diacetoxy-12-ketocholanate, available in three steps from cholic acid, is convertible with selenium dioxide into the 9,11-unsaturated derivative, which on treatment with alkali affords 3α-hydroxy-12-keto-Δ$^{7,9(11)}$-choladienic acid (IV). Reduction of this dienone with zinc and acetic acid gives a non-conjugated mono-unsaturated ketone capable of being isomerized to 3α-hydroxy-12-keto-Δ$^{9(11)}$-cholenic acid (VI).

3-Hydroxy-12-keto-Δ$^{9(11)}$-cholenic acid (VI), a key intermediate in the Kendall procedure[1] for the production of an 11-oxygenated bile acid derivative from which cortisone can be prepared, hitherto has been available only *via* desoxycholic acid. We have now found a new route to VI that does not involve desoxycholic acid as an intermediate.

A previously reported procedure[2] for the preparation of methyl cholate 3,7-diacetate in 62–70% yield consists in acetylation of the bile acid ester with acetic anhydride and pyridine in benzene solution at room temperature. A still simpler procedure utilizes dioxane as solvent; on addition of a limited amount of water, after a suitable reaction period, the diacetate crystallizes in a state satisfactory for direct use in the next step of oxidation; the mother liquor can be processed for recovery of cholic acid (89% recovery). The ketone I is obtained in nearly quantitative yield by addition of aqueous potassium chromate to an acetic acid solution of the 12-hydroxy compound, and I is dehydrogenated readily to the Δ$^{9(11)}$-12-ketone II by selenium dioxide in acetic acid, as in the standard method for preparation of VI.[3,1] When refluxed with excess aqueous alcoholic alkali, the eneone diacetate II is converted in over 90% yield into the dienone acid IV. The course of the reaction was established by the observation that treatment of II with a limited amount of alkali for a brief period afforded a certain amount of the eneonediol III, which with excess alkali affords the dienone acid IV. The facile elimination of the elements of water from the eneonediol III is attributable to activation of the C_8-hydrogen atom by the 9,11-double bond; the saturated ketone I yields no dehydro product under the conditions used for conversion of II into IV.

Barring an unlikely rearrangement, the newly introduced double bond of the dienone must be at the 7,8-position, as in IV. This structure is consistent with the presence in the spectrum of an intense absorption band at 292.5 mμ, since the maximum calculated[4] for IV from available analogs is 303 mμ. Our dienone corresponds in properties to one obtained by Shimizu, Kazuno and Matsumoto[5] from 3,7-diacetoxy-11-bromo-12-ketocholanic acid; the Japanese investigators, evidently unaware of recent revisions of the literature, erroneously formulated the compound as the Δ$^{8(14),9(11)}$-dien-12-one.[6] On hydrogenation of the dienone, the Japanese chemists obtained a mono-unsaturated ketone that they formulate as having the Δ^8-structure V. We obtained the same compound by re-

(1) B. F. McKenzie, V. R. Mattox, L. L. Engel and E. C. Kendall, *J. Biol. Chem.*, **173**, 271 (1948).

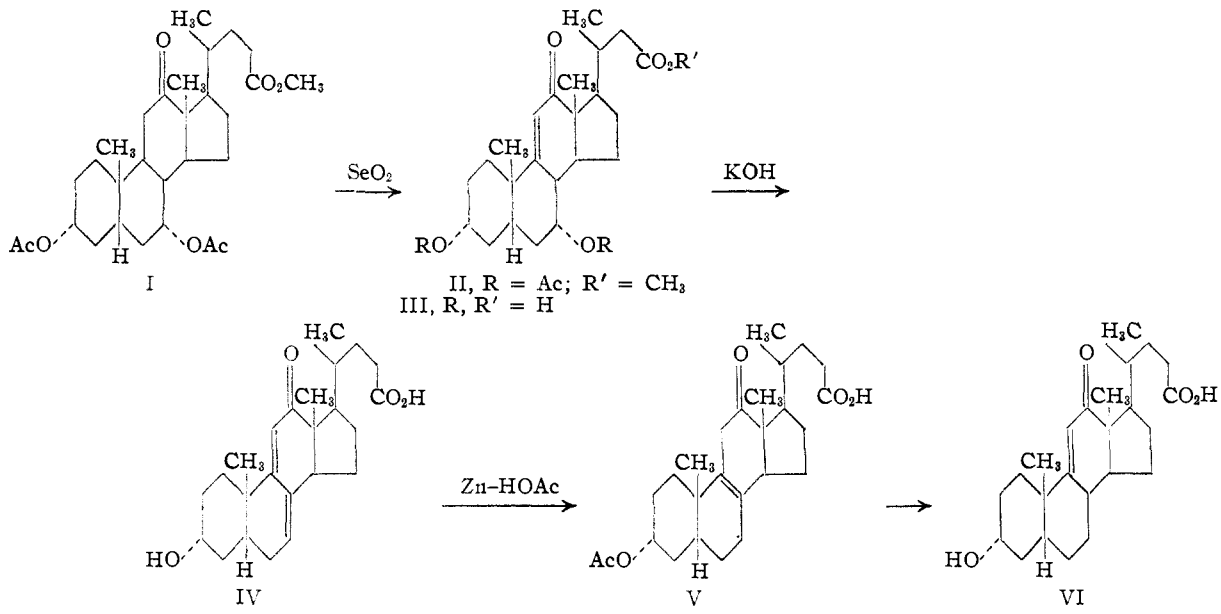
(2) L. F. Fieser and S. Rajagopalan, THIS JOURNAL, **72**, 5534 (1950).

(3) E. Schwenk and E. Stahl, *Arch. Biochem.*, **14**, 125 (1947).

(4) L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," 2nd ed., Reinhold Publishing Corp., New York, N. Y., 1949.

(5) T. Shimizu, T. Kazuno and K. Matsumoto, *J. Japan. Biochem. Soc.*, **20**, 164 (1948); *C. A.*, **44**, 164 (1950).

(6) *Chemical Abstracts* (see ref. 5) mistakenly quotes the authors as describing the compound as the Δ$^{7,9(11)}$-diene-12-one.



duction of the dienone IV with zinc and acetic acid. We found, further, that the compound is easily isomerized by acid or base to the known 3α-hydroxy-12-keto-$\Delta^{9(11)}$-cholenic acid VI. Structure V, α,β,γ-unsaturated ketone, seems established by the evidence that the substance has the optical properties of a non-conjugated ketone but can be isomerized to the α,β-unsaturated ketone VI.

Berner, Lardon and Reichstein[7] reported the preparation of methyl 3α-acetoxy-12-keto-Δ^7-cholenate and showed that the double bond in this substance readily migrates from the 7,8- to the 8,14-position. We prepared a sample of their substance and found that it depresses the melting point of the methyl ester acetate of V and that the two substances differ in infrared spectra. We found also that the Δ^7-compound, unlike the Δ^8-isomer V, is not isomerized by methanolic hydrogen chloride to the conjugated ketone VI.

Experimental

Methyl Cholate 3,7-Diacetate.—Further trials of the procedure of acetylation of methyl cholate in benzene solution[2] have given satisfactory material (m.p. 183–186°; $[\alpha]^{23}D$ +29.6, 1% in $CHCl_3$) in yield of about 70%. An alternate, simpler procedure is as follows (L.F.F.): 50 g. of methyl cholate was dissolved in 100 cc. of dioxane and 100 cc. of pyridine and the solution was cooled to room temperature (26–28°) and 150 cc. of acetic anhydride was added. After standing at 26–28° for 20 hours, 200 cc. of water was added and the solution warmed to effect solution and set aside for crystallization. The colorless crystallizate, collected after cooling for a few hours at 3°, was washed with methanol and dried to constant weight; yield 30.8–33.6 g. (51–56%), m.p. 181–183°. The yield was less when the reaction period was either shortened or extended.

The mother liquor and washings were combined and concentrated under reduced pressure to a sirupy residue, which was refluxed with 40 cc. of methanol and 200 cc. of aqueous 10% sodium hydroxide for 15 hours. The solution was filtered hot and most of the methanol removed by evaporation; the residual aqueous solution was cooled and acidified with dilute sulfuric acid. The crude acid that precipitated was dried to nearly constant weight at 80–85° (22.8 g., m.p. 155–160°, probably hydrated) and when crystallized from acetone afforded 20.3 g. of pure cholic acid, m.p. 194–196°. The yield of methyl cholate 3,7-diacetate based on the cholic acid consumed is 88.6%.

Methyl 3α,7α-Diacetoxy-12-ketocholanate (I).—Preparation of this ketone by oxidation of the diacetate alcohol with aqueous potassium chromate in acetic acid has been described (S.R.[2]). An oxidation (E.W.) of 87.5 g. of diacetate in 1 l. of acetic acid with a solution prepared from 17 g. of chromic anhydride, 22 cc. of water and 750 cc. of acetic acid was conducted on the steam-bath for one-half hour; the solution was concentrated *in vacuo* till solid began to separate and diluted with water to 3 liters. The solid was washed well with water; and the almost white crystalline product on one crystallization from methanol afforded 77.4 g. (90%) of material melting at 179–181°, $[\alpha]^{25.5}D$ +73° (dioxane), +84° (chloroform).

Methyl 3α,7α-Diacetoxy-12-keto-$\Delta^{9(11)}$-cholenate (II).—A mixture of 82 g. of methyl 3α,7α-diacetoxy-12-ketocholanate, 800 cc. of acetic acid and 48 g. of selenium dioxide was refluxed for 18 hours, and the orange solution was filtered and diluted with two volumes of water. The precipitated product was collected, washed and redissolved in 800 cc. of acetic acid. A solution of 25 g. of chromic anhydride in 100 cc. of water was added and the solution was maintained at 10–20° for four hours, filtered and diluted extensively with water. The precipitated product was nearly white and melted at 153–155°, yield 62 g. (75.5%). When the reflux time was extended to 60 hours the yield dropped to about 48%. The substance is very soluble in methanol, but crystallizes well when the solution is diluted extensively with water at the boiling point. The pure substance forms colorless needles, m.p. 159–161°, $[\alpha]^{25}D$ +75 ± 2° (1% in dioxane), $\lambda_{max.}^{C_2H_5OH}$ 237 mμ (log ε 4.06).

Anal. Calcd. for $C_{29}H_{42}O_7$ (502.63): C, 69.30; H, 8.42. Found: C, 69.25, 69.47; H, 8.35, 8.03.

Phase solubility analysis: 98.1 ± 0.5%.

Methyl 3α-Hydroxy-7α-acetoxy-12-keto-$\Delta^{9(11)}$-cholenate.—A solution of 40 g. of crude methyl 3α,7α-diacetoxy-12-keto-$\Delta^{9(11)}$-cholenate (II) in 300 cc. of methanol containing 15 g. of hydrogen chloride was let stand overnight at room temperature. The mixture was warmed, filtered from a little oil, and diluted to turbidity; on cooling and scratching colorless needles of the monoacetate separated; yield 30.8 g., m.p. 160–162°. Crystallization from aqueous methanol gave long, colorless needles, m.p. 162–163°, $[\alpha]^{22}D$ +65 ± 2° (dioxane), $\lambda_{mac.}^{alc.}$ 238 mμ (log ε 4.00).

Anal. Calcd. for $C_{27}H_{40}O_6$ (460.59): C, 70.38; H, 8.74. Found: C, 70.58; H, 8.88.

3α,7α-Dihydroxy-12-keto-$\Delta^{9(11)}$-cholenic Acid (III).—A solution of 6.5 g. (85% = 0.1 m.) of potassium hydroxide in 10 cc. of water was added to a solution of 15 g. of methyl 3α,7α-diacetoxy-12-keto-$\Delta^{9(11)}$-cholenic acid in 150 cc. of methanol and the solution was refluxed for 45 minutes and

(7) E. Berner, A. Lardon and T. Reichstein, *Helv. Chim. Acta*, **30**, 1642 (1947).

then concentrated in vacuum to about one-half the initial volume, cooled, and acidified with 2.5 *N* hydrochloric acid. A white gum separated and was extracted with chloroform; evaporation of the dried solution left an oil that afforded 3.3 g. of crystals, m.p. 204–210° dec. from benzene–methanol. A sample recrystallized from aqueous acetone melted at 202–206° dec., $\lambda_{max.}^{C_2H_5OH}$ 240 mμ (log ε 3.99).

Anal. Calcd. for $C_{24}H_{36}O_5$ (404.53): C, 71.26; H, 8.98. Found: C, 71.35; H, 8.70.

When III was refluxed with excess alcoholic alkali it yielded IV, m.p. 184–188°.

3α-Hydroxy-12-keto-$\Delta^{7,9(11)}$-choladienic Acid (IV).—A mixture of 25 g. of the diacetate ester II in 375 cc. of methanol and 25 g. of potassium hydroxide in 375 cc. of water was refluxed for six hours and the deep red-orange solution cooled and acidified by dropwise addition of hydrochloric acid. After chilling, the crystalline dienone acid was collected and washed. Crystallization from aqueous acetone gave 14.8 g. (92%) of hard, pale yellow needles and rods, m.p. 185–187° (2 crops). With a 5-g. batch of II the hydrolysis was apparently complete in one hour; with a 94-g. batch, the reaction was incomplete in three hours but complete in six hours. The product was obtained colorless by chromatographing through a column of acid-washed alumina. $[\alpha]^{20-22}D$ +250, +248 (dioxane), $\lambda_{max.}^{alc.}$ 292.5 mμ (log ε 4.11), 240 mμ (log ε 3.57).

Anal. Calcd. for $C_{24}H_{34}O_4$ (386.51): C, 74.58; H, 8.87. Found: C, 74.64; H, 8.52.

The acetate separated from aqueous methanol in almost colorless needles, m.p. 225–226° dec., $[\alpha]^{21}D$ +261 ± 5° (dioxane).

Anal. Calcd. for $C_{26}H_{36}O_5$ (428.55): C, 72.86; H, 8.47. Found: C, 73.01; H, 8.27.

The methyl ester of IV, prepared by esterification of the acid with 10 parts of methanol and 0.1 part of 36% hydrochloric acid at room temperature for ten hours, crystallized from methanol in pale yellow, prismatic needles, m.p. 148–149°, $[\alpha]^{20}D$ +251 ± 5° (dioxane), $\lambda_{max.}^{alc.}$ 290 mμ (log ε 4.13), 237.5 mμ (log ε 3.58).

Anal. Calcd. for $C_{25}H_{36}O_4$ (400.54): C, 74.96; H, 9.06. Found: C, 74.84, 75.06; H, 8.93, 8.95.

The methyl ester acetate of IV separated from dilute alcohol in almost colorless needles, m.p. 152–153°, $[\alpha]^{22}D$ +258 ± 5° (dioxane).

Anal. Calcd. for $C_{27}H_{38}O_5$ (442.57): C, 73.57; H, 8.65. Found: C, 73.07; H, 8.48.

3α-Acetoxy-12-keto-Δ^8-cholenic Acid (Acetate of V).—A solution of 15 g. of 3α-hydroxy-12-keto-$\Delta^{7,9(11)}$-choladienic acid (IV) in 500 cc. of acetic acid was refluxed with 50 g. of zinc dust for 60 hours. The solution was filtered hot, and the zinc was washed thoroughly with hot acetic acid; the solvent was removed from the combined filtrate and washings *in vacuo*. The gummy residue was rubbed with water and solidified within a few hours. The solid was washed with water and crystallized from methanol and the solvated product dried to constant weight; yield (two crops) 11.9 g. (73%), m.p. 148–152°, $[\alpha]^{22}D$ +110° (chloroform).

Anal. Calcd. for $C_{26}H_{38}O_5$ (430.56): C, 72.52; H, 8.90. Found: C, 72.45; H, 9.00.

The substance was found to be identical with a sample prepared by hydrogenation according to Shimizu, *et al.*[5]

The methyl ester acetate, prepared with diazomethane and crystallized from methanol, melted at 131–134° (unsolvated); $[\alpha]^{25}D$ +128° (chloroform).

Anal. Calcd. for $C_{27}H_{40}O_5$ (444.59): C, 72.93; H, 9.07; Ac, 9.64. Found: C, 72.89; H, 9.34; Ac, 9.70.

3α-Hydroxy-12-keto-$\Delta^{9(11)}$-cholenic Acid (VI).—3α-Acetoxy-12-keto-Δ^8-cholenic acid (2 g.) was dissolved under nitrogen in 30 cc. of 0.6 *N* methanolic hydrogen chloride. The solid dissolved in about one-half hour to a pale yellow solution, which was kept at room temperature for 40 hours, when the extinction coefficient appeared to have reached a maximum, and was then evaporated *in vacuo*. Water was added to the residue, and an off-white solid obtained. This was collected, washed with sodium carbonate solution and then with water. Two crystallizations from methanol gave material of m.p. 115–118°, $[\alpha]^{22}D$ +104° (chloroform), that did not depress the m.p. of authentic methyl 3α-hydroxy-12-keto-$\Delta^{9(11)}$-cholenate. The ester was then dissolved in 15 cc. of methanol and stirred while 0.38 g. of sodium hydroxide in 1 cc. of water was added dropwise, followed by 1 cc. of wash water. The yellow solution was heated at 40° for 45 minutes, diluted with 15 cc. of water, and concentrated *in vacuo* to a mush. The product was brought into solution with 60 cc. of acetone, 15 cc. of water and 1.1 cc. of acetic acid were added, and the mixture was diluted with 40 cc. of water and chilled for one hour. The resulting solid was crystallized twice from water and afforded 1.4 g. (80%) of hydroxy acid, m.p. 173–175°, $[\alpha]^{22}D$ +103° (chloroform), $\lambda_{max.}^{alc.}$ 240 mμ (log ε 4.03). The substance showed no depression when mixed with authentic 3α-hydroxy-12-keto-$\Delta^{9(11)}$-cholenic acid, m.p. 176–177.5°.

Alkaline isomerization of 3α-acetoxy-12-keto-Δ^8-cholenic acid was accomplished by adding 20 cc. of 30% sodium hydroxide with cooling to a suspension of 1 g. of the acetate acid in 20 cc. of methanol. After standing at room temperature overnight, the dark green solution was concentrated *in vacuo* and the residue was dissolved in hot water and acidified to litmus with acetic acid. Chilling afforded a solid product, which when crystallized twice from methanol gave 0.7 g. of 3α-hydroxy-12-keto-$\Delta^{9(11)}$-cholenic acid, m.p. 174–177° (no depression with authentic sample), $[\alpha]^{23}D$ +100° (chloroform), $\lambda_{max.}^{alc.}$ 240 mμ (log ε 4.03).

CAMBRIDGE, MASSACHUSETTS
RAHWAY, NEW JERSEY

RECEIVED MARCH 14, 1951