cending technique. The phospholipids were applied to the paper (5″ × 16″) at a concentration of 50 μg. each per 20 μl. of isoamyl alcohol–benzene 1:1. Since mixtures were run the total amount of lipid applied to the paper varied from 50 to 150 μg. Good separations were also obtained using 300–400 μg. of total lipids.

Whatman No. 1 filter paper was used which had previously been washed successively with 1 *N* acetic acid, water and methanol. The papers were first dried in air and then in an oven at 100° for 15 minutes. The solvent systems used were as follows: solvent A, lutidine–methanol 3:2; solvent B, methanol–lutidine–acetic acid 4:16:1; solvent C, 2-octanol–lutidine–acetic acid 90:5:5.

In the chloroform–lutidine–acetic acid system reported previously,[7] which gave useful separations of unmodified phospholipids, the N-acetyl derivatives of I and II moved with an R_f value of 0.93 and were not separated from each other but were completely resolved from phosphatidylcholines. On the other hand, solvent systems A, B and C were able to resolve the N-acetyl derivatives of I and II.

The solvent mixtures were prepared on a volume to volume basis. The lutidine used was obtained from the Eastman Kodak Co. (T4908), and was a mixture of lutidines. Pure 2-octanol, absolute methanol and J. T. Baker analyzed glacial acetic acid were used. In preparing solvents B and C the acetic acid and lutidine were first mixed, cooled and then the third component added. The chambers were lined internally with filter paper and equilibrated with 150 ml. of the developing solvent for 8 hours before use. All chromatograms were run at about 27°. The solvent front was allowed to run about 14–15″.

Detection of the Phospholipids on Chromatograms.—The chromatograms were air-dried, washed with distilled water for 10 minutes, dried and then immersed in an aqueous solution of 0.001% Rhodamine B or G (National Aniline Division, Allied Chemical and Dye Corp.) for 10 minutes. The excess dye was washed out with distilled water and the papers air-dried. The lipid spots were observed under ultraviolet light as fluorescent areas. In addition, the phosphatidylcholines can be detected by the method of Levine and Chargaff[15] and the lipids containing a free amino group by spraying with a 0.3% solution of ninhydrin in acetone–lutidine 95:5. The blue color resulting from the latter reaction was allowed to develop at room temperature in the dark.

(15) C. Levine and E. Chargaff, *J. Biol. Chem.*, **192**, 465 (1951).

DEPARTMENT OF BIOCHEMISTRY
UNIVERSITY OF ROCHESTER
ROCHESTER 20, N. Y.

24-Hydroxycholane-3,7,12-trione

BY ROBERT H. MAZUR AND EDWARD A. BROWN

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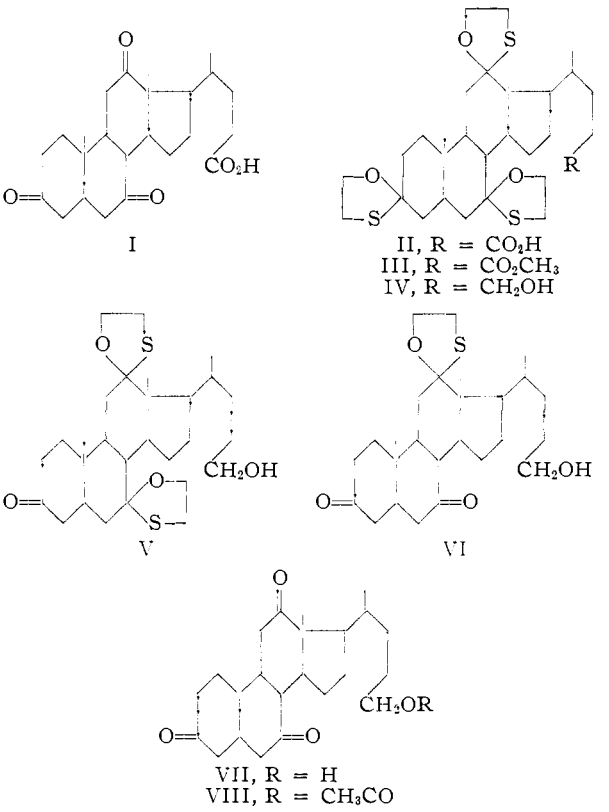
In view of the physiological activity of dehydrocholic acid (I)[1] it seemed desirable to prepare 24-hydroxycholane-3,7,12-trione (VII) for pharmacological evaluation. A suitable synthesis was found by protection of the three keto groups of dehydrocholic acid, reduction of the carboxylic acid (in the form of its ester) and regeneration of the keto groups. The commoner reagents for blocking ketones are ethylene glycol[2] and ethanedithiol.[3] In the present case, ethylene glycol failed even on long refluxing to form a triethylene ketal. Although ethanedithiol reacted readily, the trimercaptol was attacked by lithium aluminum hydride to yield unidentifiable sulfur-containing products. The use of β-mercaptoethanol[4,5] proved successful and gave the desired dehydrocholic acid trihemithioethylene ketal II. The latter was



esterified with ethereal diazomethane and the resulting ester III reduced with lithium aluminum hydride to the alcohol IV. It was found possible to hydrolyze IV selectively so that the mono-, di- and triketo alcohols were prepared. Structures were assigned on the basis of the known relative reactivities of the keto groups of dehydrocholic acid.[6] The 3-ketone V was obtained by acid-catalyzed methanolysis while hydrolysis with sulfuric acid in aqueous dioxane gave a separable mixture of the 3,7-diketone VI and 24-hydroxycholane-3,7,12-trione (VII). A more convenient procedure was to hydrolyze IV with concentrated hydrochloric acid in acetic acid with simultaneous acetylation to yield VIII. The latter could readily be saponified to VII. Compounds II–VIII had the expected infrared spectra (taken at 0.5% concentration in a potassium bromide disc).

Experimental[7]

Dehydrocholic Acid Trihemithioethylene Ketal (II).—A mixture of 82 g. of dehydrocholic acid, 88 g. of β-mercaptoethanol and 1 g. of *p*-toluenesulfonic acid monohydrate in 1.8 liters of toluene was heated under reflux (continuous water separator) for 18 hours. The toluene was distilled and the residue dissolved in 900 ml. of 90% aqueous methanol containing 80 g. of potassium hydroxide. The solution was heated under reflux for two hours, poured into an excess of cold, dilute hydrochloric acid and the product taken up in benzene. The benzene was distilled to a small volume and the residue diluted with one liter of

(1) V. A. Drill, "Pharmacology in Medicine," McGraw-Hill Book Co., Inc., New York, N. Y., 1954, sec. 43, p. 6.

(2) M. B. Fernholz, U. S. Patent 2,378,918; *C. A.*, **39**, 5051 (1945).

(3) H. Hauptmann, THIS JOURNAL, **69**, 562 (1947).

(4) J. Romo, G. Rosenkranz and C. Djerassi, *ibid.*, **73**, 4961 (1951).

(5) C. Djerassi and M. Gorman, *ibid.*, **75**, 3704 (1953).

(6) L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," 3rd Ed., Reinhold Publ. Corp., New York, N. Y., 1949, p. 125.

(7) We are indebted to Robert T. Dillon and his associates for analyses and determinations of physical properties.

methanol. On standing overnight in the ice-box, there was obtained 54.5 g. of crystalline material. The mother liquors gave an additional 1.5 g. on dilution with methanol. Recrystallization from benzene–methanol gave 35 g. (30%) of long needles, m.p. 140–155°. An analytical sample had m.p. 155° (bubbly melt) with sintering at 150°, $[\alpha]_D$ +92° (dioxane). The analytical sample was dried under vacuum at 118° for 5 hours.

Anal. Calcd. for $C_{30}H_{46}O_5S_3$: C, 61.82; H, 7.96. Found: C, 61.64; H, 7.95.

Methyl Dehydrocholate Trihemithioethylene Ketal (III).—The above acid (33.5 g.) in 400 ml. of ether was treated with an excess of ethereal diazomethane. The crude product after drying and distillation of the ether weighed 34 g. (99%), m.p. 173–178°. Crystallization from methanol gave small feathery needles, m.p. 183° with sintering at 177°, $[\alpha]_D$ +90° (dioxane). The analytical sample was dried under vacuum at 100° for one hour.

Anal. Calcd. for $C_{31}H_{48}O_5S_3$: C, 62.38; H, 8.11. Found: C, 62.30; H, 7.86.

24-Hydroxycholane-3,7,12-trione Trihemithioethylene Ketal (IV).—The above ester (31 g.) in one liter of dry ether was added dropwise with stirring to a suspension of 3.8 g. of lithium aluminum hydride in 400 ml. of dry ether. The mixture was heated at the reflux temperature for two hours and allowed to stand overnight. The reaction mixture was worked up with dilute sulfuric acid in the usual manner. The crude product (29 g., 98%) had m.p. 222–228° and two recrystallizations from benzene–methanol gave needles with m.p. 233–235°, $[\alpha]_D$ +95° (dioxane). The analytical sample was dried under vacuum at 118° for 5 hours.

Anal. Calcd. for $C_{30}H_{48}O_4S_3$: C, 63.34; H, 8.51. Found: C, 62.95; H, 8.57.

Hydrolysis of 24-Hydroxycholane-3,7,12-trione Trihemithioethylene Ketal. A.—The above alcohol (2.0 g.) was dissolved in 150 ml. of dry methanol containing 3 ml. of acetyl chloride and the solution heated under reflux for five hours. The methanol was distilled, the residue taken up in ether, washed with sodium bicarbonate, the ether distilled and the residue chromatographed on silica. Elution with 15% ethyl acetate–benzene gave 0.74 g. (41%) of 24-hydroxycholane-3,7,12-trione 7,12-dihemithioethylene ketal (V). The product, thick needles after two crystallizations from benzene–cyclohexane, had m.p. 212.6–214.2°, $[\alpha]_D$ +43° (dioxane). The analytical sample was dried under vacuum at 118° for 5 hours.

Anal. Calcd. for $C_{28}H_{44}O_4S_2$: C, 66.10; H, 8.72. Found: C, 66.15; H, 8.77.

B.—Compound IV (7.0 g.) was dissolved in 315 ml. of dioxane containing 35 ml. of water and 8 ml. of concentrated sulfuric acid. The solution was heated under reflux for 22 hours, neutralized with solid sodium bicarbonate, and filtered. The filtrate was concentrated under reduced pressure and the residue taken up in methanol. On standing, 0.5 g. of material crystallized, m.p. 192–206°. The mother liquor was distilled to dryness, taken up in 15 ml. of benzene and diluted with 75 ml. of ether to give 1.7 g., m.p. 202–207°. The mother liquor was diluted with ether and gave 0.4 g., m.p. *ca.* 178° with previous sintering. Evaporation of the mother liquor to dryness and trituration with ether gave 1.5 g., m.p. *ca.* 170°.

Crops 3 and 4 (1.9 g.) were combined and crystallized twice from methanol, yielding 0.55 g. of 24-hydroxycholane-3,7,12-trione 12-hemithioethylene ketal (VI), m.p. 182–183°, $[\alpha]_D$ +57.5° (dioxane).

Anal. Calcd. for $C_{26}H_{40}O_4S$: C, 69.31; H, 8.99; S, 7.15. Found: C, 69.33; H, 8.93; S, 7.13.

Crops 1 and 2 (2.2 g.) were crystallized from methanol and gave 1.3 g. of 24-hydroxycholane-3,7,12-trione (VII), m.p. 212–213°, $[\alpha]_D$ +28° (dioxane).

Anal. Calcd. for $C_{24}H_{36}O_4 \cdot {}^1/_2CH_3OH$: C, 72.74; H, 9.47. Found: C, 72.47; H, 9.12; S, 0.0.

The triketone alcohol (0.5 g.) was acetylated with pyridine and acetic anhydride. The product was recrystallized from benzene–petroleum ether (b.p. 60–70°) and gave 24-acetoxycholane-3,7,12-trione (VIII), m.p. 203–204°, $[\alpha]_D$ +26° (dioxane). The analytical sample was dried under vacuum at 56°.

Anal. Calcd. for $C_{26}H_{38}O_5$: C, 72.52; H, 8.90. Found: C, 72.37; H, 8.95.

C.—Trihemithioethylene ketal alcohol (2.0 g.) was dissolved in 50 ml. of glacial acetic acid. One ml. of concentrated hydrochloric acid was added and the solution heated under reflux for 22 hours. The acid was distilled under reduced pressure and the residue triturated with ether which gave 0.9 g. (59%) of 24-acetoxycholane-3,7,12-trione, m.p. 191–200°. Crystallization from benzene–petroleum ether (b.p. 60–70°) yielded 0.7 g. of needles, m.p. 202°, which did not depress the melting point of authentic cholanetrione acetate (m.p. 202–203°).

The acetate (0.207 g.) was dissolved in 8 ml. of 4% potassium hydroxide in methanol, the solution concentrated to 5 ml. and allowed to stand, giving in two crops 0.139 g. (74%) of long colorless needles. The analytical sample was crystallized from 80% methanol, had m.p. 212.2–212.8°, and was dried under vacuum at 118° for 5 hours. The infrared spectrum was identical with that of the triketone alcohol (m.p. 212–213°) obtained previously.

DIVISION OF CHEMICAL RESEARCH
G. D. SEARLE AND CO.
CHICAGO 80, ILLINOIS

Heterocyclic Derivatives of Arsenic: Some Corrected Statements

BY F. G. MANN

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It has been stated by Weston[1] that Chatt and Mann[2] have resolved 5,10-di-*p*-tolyl-5,10-dihydroarsanthrene into optically active forms. A statement to the same effect is made by Costain and Sutherland.[3] Actually Chatt and Mann separated this compound into two geometric isomers. Each isomer possesses a plane of symmetry and therefore no question of optical resolution arises.

Garascia and Mattei[4] have stated that Cookson and Mann[5] have "prepared several 9-substituted arsafluorenes." Our compounds were, however, 10-substituted 9,10-dihydroarsanthridines, the central ring system being six-membered and not five-membered. Garascia and Mattei's further statement that we record 9-arsafluorinic acid, m.p. 299°, is therefore also incorrect, for no arsafluorene compounds are described in our paper.

(1) R. E. Weston, THIS JOURNAL, **76**, 2645 (1954).
(2) J. Chatt and F. G. Mann, *J. Chem. Soc.*, 1184 (1940).
(3) C. C. Costain and G. B. B. M. Sutherland, *J. Phys. Chem.*, **56**, 321 (1952).
(4) R. J. Garascia and I. V. Mattei, THIS JOURNAL, **75**, 4589 (1953).
(5) G. H. Cookson and F. G. Mann, *J. Chem. Soc.*, 2888 (1949).

UNIVERSITY CHEMICAL LABORATORY
CAMBRIDGE, ENGLAND

Condensation of Chloral Hydrate with 8-Quinolinol

BY KONOMU MATSUMURA AND MOTOKO ITO

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Chloral hydrate reacts with 8-quinolinol to form predominantly 5-(α-hydroxy-β-trichloroethyl)-8-quinolinol (I). On alkaline hydrolysis with methanolic potassium hydroxide I yielded 5-carboxy- and 5-formyl-8-quinolinol (IV) and a compound which is soluble in sodium bisulfite. Hydrolysis with sodium methoxide in methanol gave IV and a compound V which is insoluble in sodium bisulfite and which gives some indication of possessing the carboxy and methoxy groups. Thus, I on alkaline hydrolysis appears to undergo only partial hydrolysis to V; the remainder of V undergoes