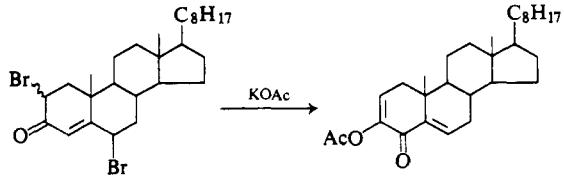
Mechanism of the Reaction of 2ξ, 6β-Dibromocholest-4-en-3-one with Potassium Acetate

Percy L. Julian, Ludwig Bauer, Charles L. Bell, and Richard E. Hewitson[1]

Contribution from The Julian Research Institute of Franklin Park, Franklin Park, Illinois 60131, and the Department of Chemistry, College of Pharmacy, University of Illinois at the Medical Center, Chicago, Illinois 60680. *Received August 29, 1968*

Abstract: The reaction of 2ξ,6β-dibromocholest-4-en-3-one (**1**) with ^{18}O-labeled acetate ion, yielded 3-acetoxycholesta-2,5-dien-4-one (**2**) labeled only in the C=O of the ester at C-3 and the ketone at C-4. A mechanism is proposed in which initial attack by the carboxylate ion on C-4 transforms **1** into 2-bromo-4-acetoxycholest-5-en-3-one (**3**), which subsequently undergoes a 1,3-elimination rearrangement. To substantiate the possible intermediacy of **3** in the title reaction, **3** was synthesized independently. It was found that **3** could be converted readily to **2** by carboxylate ions, collidine, and N,N-dimethylaniline. Attempted preparation of 4,4-dibromocholest-5-en-3-one from the reaction of either of the two isomeric 4-bromo-4,5-epoxycholestan-3-ones with hydrogen bromide unexpectedly produced 2α,4-dibromocholest-4-en-3-one. A mechanism to account for the last transformation is proposed.

The conversion of 2ξ,6β-dibromocholest-4-en-3-one (**1**) by potassium acetate into 3-acetoxycholesta-2,5-dien-4-one (**2**) (acetate is represented by OAc throughout this paper) has been the subject of numerous investigations,[2-6] since its discovery almost simultaneously by Inhoffen[3a] and Butenandt.[3b] This type of elimination–rearrangement reaction has been extended to a variety of 2,6-dibromo-Δ^4-3-oxo steroids to yield the corresponding esters of 2,5-dien-3-ol-4-ones and other derived products which exhibited interesting physiological properties.[6] Thus, this transformation can be considered as a rather general reaction for 2,6-dibromo-Δ^4-3-oxo steroids of the normal steroid series.



1 **2**

Mass Spectral Investigation. In order to establish the mechanism for this reaction, **1** was treated with ^{18}O-labeled potassium acetate[7] in hot tetrahydrofuran. The attacking acetate ion was unlabeled and mono- and dilabeled with ^{18}O in the ratio of 1:2:1.

The relevant fragmentation paths in the mass spectrum of **2** are shown in Scheme I. The mass peaks in Scheme I under N refer to the ions from the natural compound and those under L, to the ions obtained from the ^{18}O-labeled compound. Probable structures are shown for most of the charged fragments and the decompositions for which metastable ions are found are marked by an asterisk. The parent molecular ion radical for the natural material, **2**, is at *m/e* 440, while in the labeled compound mass peaks occur at *m/e* 440, 442, and 444 in the same ratio of 1:2:1 as in the labeled potassium acetate. This ratio represents unlabeled, monolabeled, and dilabeled species. No trilabeling is observed although the compound does possess three oxygen atoms thus eliminating the possibility of random exchange or mixing between all three oxygen atoms. The loss of a methyl radical from the parent ion to give an ion, a, at *m/e* 425, as expected, does not alter the oxygen isotope distribution since three fragment ions appear at *m/e* 425, 427, and 429. The base peak, b, in the spectrum arises from the loss of ketene from the acetoxy group to give a fragment ion at *m/e* 398. Significantly, the labeled material resulted in only two species for this fragment representing unlabeled and monolabeled ions. This indicates that one of the ^{18}O atoms is lost with ketene. The site of the remaining label in b could not be located from identifiable subsequent decompositions as shown in Scheme I. The ions produced from b either possess both oxygen atoms as in c, d, and e, or no oxygen atoms as in f. This leaves the question undecided as to whether the remaining ^{18}O label, in each of these species, is at C_3 or C_4. Since no fragment from **2** contained only one oxygen atom, attention was directed to the possible catalytic hydrogenolysis of **2** to 5α-cholestan-4-one (*cf.* ref 8) in order to pinpoint the site of the ^{18}O label.

The reduction of **2** actually afforded a mixture of 3β-acetoxy-5α-cholestan-4-one (g, see Scheme II), 5α-

(1) Abstract from the Ph.D. Thesis of Richard E. Hewitson, University of Illinois at the Medical Center, June 1968.

(2) For a review of this reaction see L. Fieser and M. Fieser, "Steroids," Reinhold Publishing Corp., New York, N. Y., 1959, pp 291–294, 302–303.

(3) (a) H. H. Inhoffen, *Ber.*, **69**, 1702 (1939); **70**, 1695 (1937); (b) A. Butenandt and G. Schramm, *ibid.*, **69**, 2289 (1936).

(4) A. Butenandt and A. Wolff, *ibid.*, **68**, 2091 (1935).

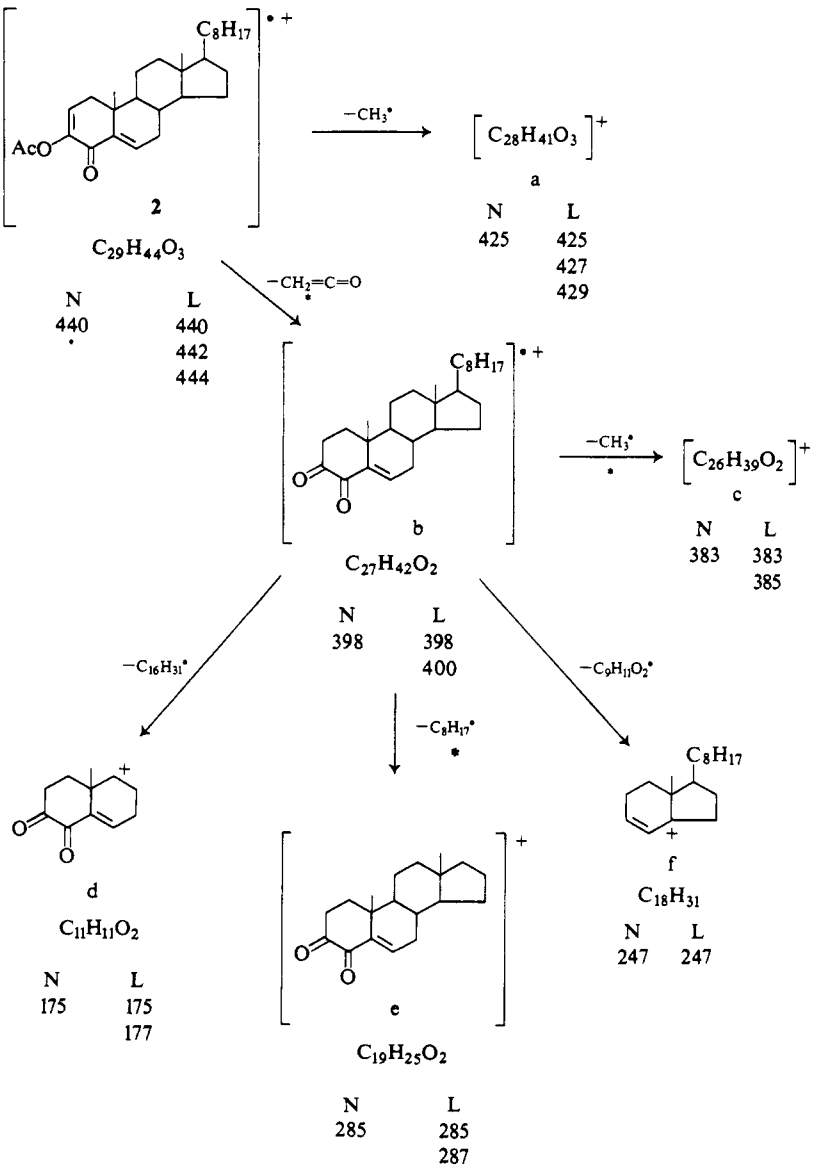
(5) (a) L. F. Fieser, M. Fieser, and S. Rajagopalan, *J. Org. Chem.*, **13**, 800 (1948); (b) D. H. R. Barton and E. Miller, *J. Amer. Chem. Soc.*, **72**, 1066 (1950); (c) E. J. Corey, *ibid.*, **76**, 175 (1954); (d) C. Djerassi, G. Rosenkranz, J. Romo, St. Kaufmann, and J. Pataki, *ibid.*, **72**, 4534 (1950); (e) L. F. Fieser, M.A. Romero, and M. Fieser, *ibid.*, **77**, 3305 (1955); (f) B. Ellis and V. Petrow, *J. Chem. Soc.*, 1179 (1956).

(6) P. J. Julian and H. C. Printy, U. S. Patent 2,891,974 (June 23, 1959); *Chem. Abstr.*, **54**, 1622*d* (1960); P. L. Julian and H. C. Printy, U. S. Patent 2,891,975 (June 23, 1959); *Chem. Abstr.*, **54**, 1624*b* (1960); P. L. Julian, E. Huang, and A. Magnani, U. S. Patent 3,153,061 (Oct 13, 1964); *Chem. Abstr.*, **61**, 16130*h* (1964).

(7) The reaction of CH_3COCl with $H_2{}^{18}O$ (97.1% in ^{18}O, purchased from International Chemical and Nuclear Corp., City of Industry, Calif.) by the procedure of Gash and Yuen [*J. Org. Chem.*, **31**, 4234 (1966)] produced acetic acid, molecular ions at *m/e* 60, 62, and 64 (ratio 1:2:1) indicating unlabeled, mono-, and dilabeled ^{18}O material.

(8) P. L. Julian and H. C. Printy, U. S. Patent 2,900,399 (Aug 18, 1959); *Chem. Abstr.*, **54**, 1622*i* (1960).

Scheme I. Partial Fragmentation of 3-Acetoxycholest-2,5-dien-4-one (2)



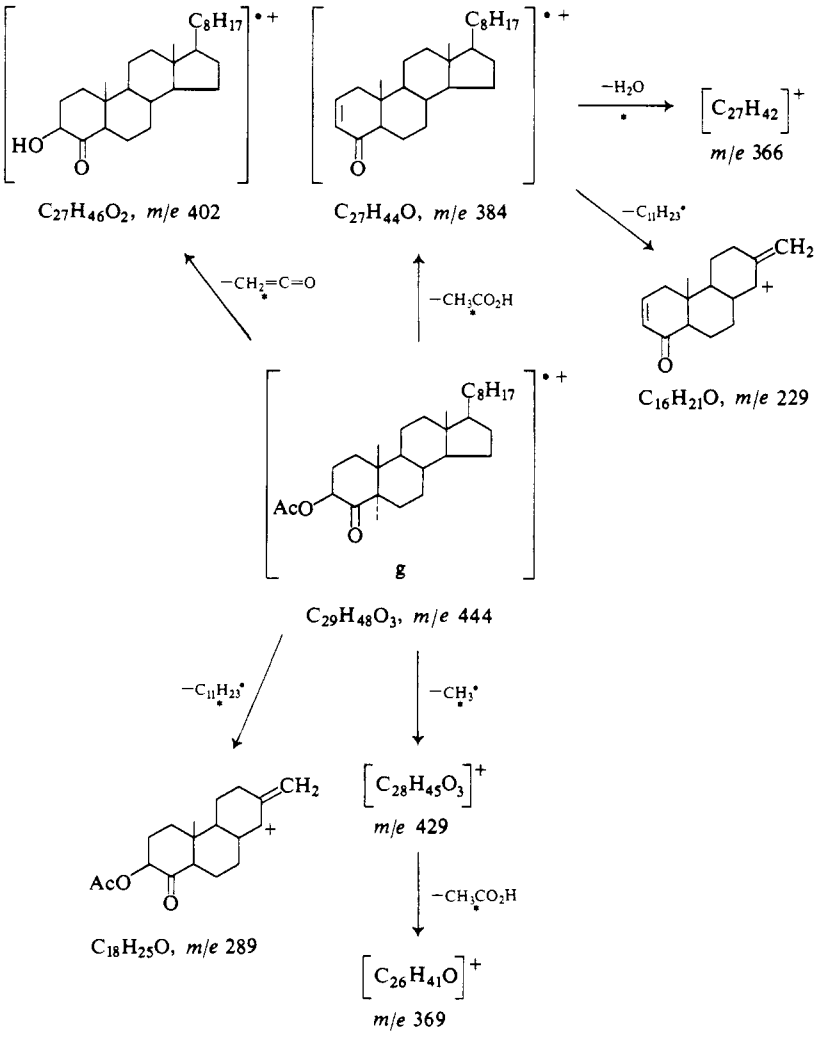
cholestan-4-one (h, see Scheme III), and cholestane, which were separated by column chromatography over alumina and identified. Six of the seven transformations of 3β-acetoxy-5α-cholestan-4-one (g, Scheme II) are supported by appropriate metastable ions. It should be noted that in addition to the loss of ketene, the molecular ion also loses acetic acid. A similar presentation for fragmentation of 5α-cholestan-4-one (h) is shown in Scheme III. Again, six of the seven fragmentations shown yield metastable ions.

A check was made to determine if chromatographic separation over alumina could alter the isotope distribution in the reduction products. ^{18}O-Labeled 5α-cholestan-4-one (50%) was prepared from a sample of pure 5α-cholestan-4-one, dissolved in dry petroleum ether (bp 30–60°) and adsorbed onto an alumina column. After elution with a dry solution of 10% benzene in petroleum ether the ketone was completely free of ^{18}O-label, establishing that a facile oxygen exchange[9] occurs on alumina. Additional evidence for this exchange is found in the mass spectrum of the labeled reduction mixture. The component of highest mass corresponds to 3β-acetoxy-5α-cholestan-3-one, which gives molecular ions (see g) at *m/e* 444, 446, and 448, representing unlabeled, monolabeled, and dilabeled compound. After chromatographic separation over alumina only unlabeled and monolabeled material is found, again indicating the ready exchange of one of the oxygen atoms, presumably the ketone oxygen of g.

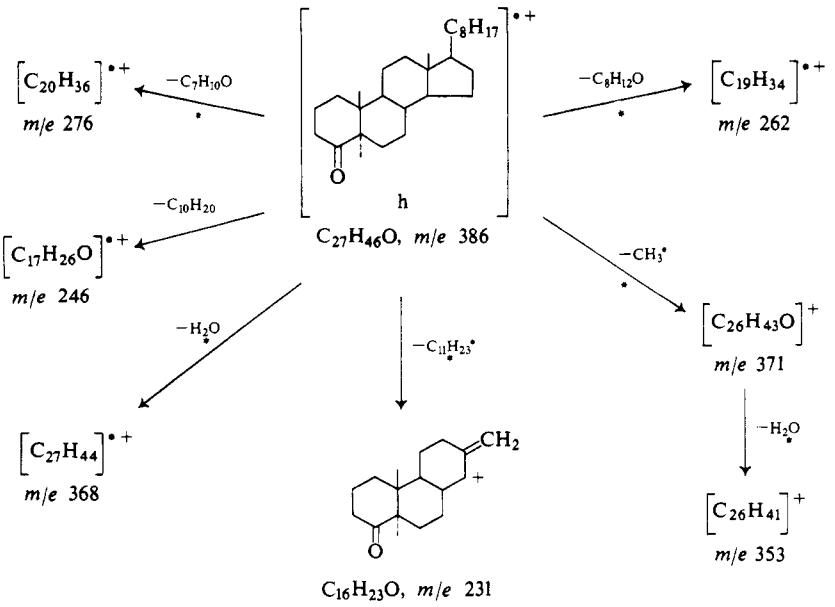
In the mass spectrum of labeled 5α-cholestan-4-one (h) peaks would be expected at *m/e* 388 and 233, since these peaks represent, respectively, the labeled parent ion and the loss of side chain plus 42 mass units from this ion. Actually the mass spectrum of the reduction mixture showed ions of near equal abundance at *m/e* 386 and 388 and at *m/e* 231 and 233 proving that the C-4 carbonyl group is labeled. Thus it is quite clear that during the

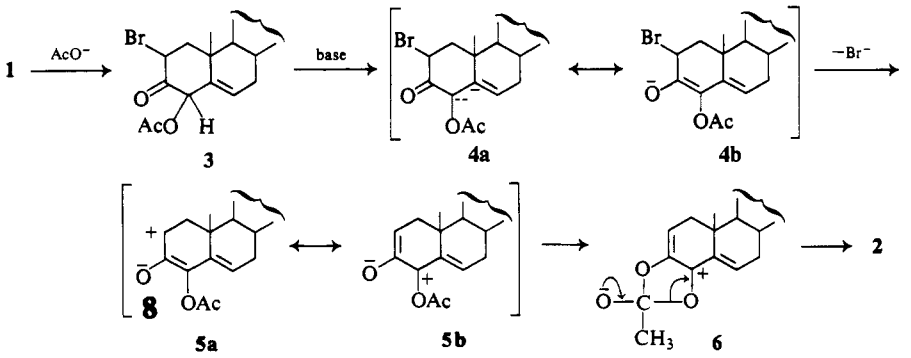
(9) (a) G. Aksnes, D. Aksnes, and T. Albrikten, *Acta Chem. Scand.*, **20**, 325 (1966); (b) M. Byrn and M. Calvin, *J. Amer. Chem. Soc.*, **88**, 1916 (1966).

Scheme II. Partial Fragmentation of 3β-Acetoxy-5α-cholestan-4-one



Scheme III. Partial Fragmentation Pattern of 5α-Cholestan-4-one

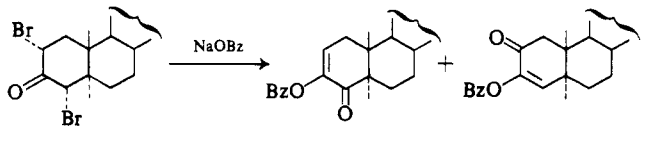




conversion of **1** → **2** with ^{18}O-labeled acetate ion, a label is introduced in the ester C=O at C-3 and the ketone C=O at C-4. This is substantiated by the partial fragmentation schemes for **2** and for the two oxygen-containing reduction products. On this basis, the original oxygen atom at C-3 in **2** does not bear a label.

Mechanism of the Reaction. The mass spectral study clearly indicated that the ^{18}O-labeled acetate ion never attacks C-3 of **1**, but is attached to C-4 sometime during the transformation of **1** to **2**. It is suggested, therefore, that the first step in the reaction consists of an SN2′ displacement of the allylic bromo group at C-6 of **1** to form the bromo acetate **3** (this step was also postulated previously by Wendler[10]). When **3** was synthesized independently and shown to react with a number of bases to form **2**, the possibility that **3** might very well be an intermediate in this reaction was enhanced.

Attempts to detect the presence of **3** by thin layer chromatography (tlc) during the reaction of **1** with acetate ion proved unsuccessful, which might be attributed to a very fast reaction of **3** with bases. To complete the conversion of **3** to **2**, it is postulated that the first step is the abstraction of the acidic proton at C-4 to create **4**, from which bromide ion is lost to give rise to the dipolar structure **5**.[11-13] Transacetylation of **5** *via* **6** would complete the reaction to furnish **2**. Such a mechanism is consistent with the one advanced by Bordwell and Wellman to explain, for example, the 1,3-elimination–rearrangement reaction of a 2-acetoxy-4,4-dimethyl-6-bromocyclohexanone by acetate ion to form 2-acetoxy-5,5-dimethylcyclohex-2-enone.[12] Furthermore, it should be noted that the alkene at C-5 in **1** is not an essential structural requirement for this reaction. It was also reported[12] that 2α,4α-dibromo-cholestan-3-one (**7**) reacted with sodium benzoate to give a mixture of 3-benzoyloxycholest-2-en-4-one (**8**) and 3-benzoyloxycholest-3-en-2-one (**9**) in 3 and 12% yield, respectively (Bz is C_6H_5CO), a reaction which is considerably slower



than that of **1** with carboxylate ions. The fact that a related 1,3-elimination–rearrangement reaction takes place in 2,6-dibromocyclohexanones is important since this supports the mechanisms advanced above. It is felt that the 5,6-alkene in **3** increases the acidity of H-4 and provides additional resonance stabilization for both **4** and **5**, thereby enhancing the reaction, **1** → **2**.

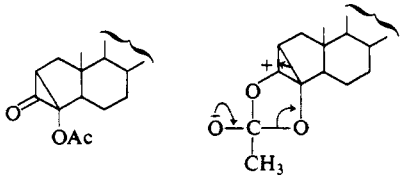
The first mechanism suggested for the conversion of **1** to **2** was advanced by Fieser[2a] and is considered briefly.

(10) Private communication by N. L. Wendler to L. F. Fieser quoted in footnote 5 of ref 2, p 303.

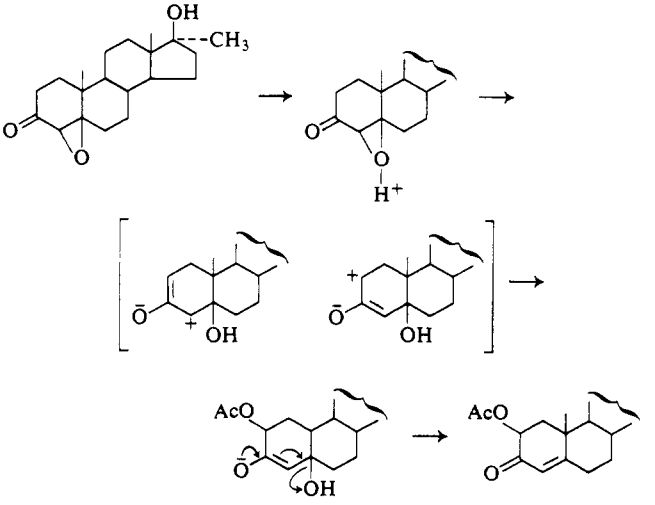
(11) Dipolar ion intermediates have been suggested a number of times for Favorskii-like reactions: (a) E. W. Warnhoff, C. M. Wong, and W. T. Tai, *J. Amer. Chem. Soc.*, **90**, 514 (1968); (b) N. J. Turro and W. B. Hammond, *ibid.*, **87**, 3258 (1965); (c) H. R. Nace and B. A. Olson, *J. Org. Chem.*, **32**, 3438 (1967).

(12) F. G. Bordwell and K. M. Wellman, *ibid.*, **31**, 351 (1966).

(13) Although the dipolar ion is preferred to a cyclopropanone intermediate, there is no way of distinguishing between **5** and a cyclopropanone intermediate, shown below. The latter could ring open and transacetylate equally well to give **1**. A number of related reactions can be rationalized to proceed either *via* dipolar or cyclopropanone

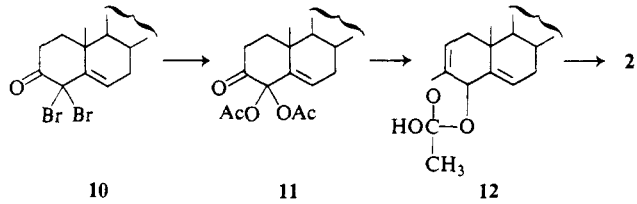


intermediates. For example, on heating 4β-bromo-5β-cholestan-3-one with potassium acetate in boiling acetic acid, 2β-acetoxy-5β-cholestan-3-one was formed [Y. Satoh, M. Mukoh, Y. Ogaki, T. Takahasi, T. Kimura, H. Aoki, and A. Hagitani, *Chem. Pharm. Bull.* (Tokyo), **39**, 855 (1966)]. Under the same conditions, 2β-bromo-5α-cholestan-3-one always yielded a complex consisting of a 1:1 mixture of 2α- and 4α-acetoxy-5α-cholestan-3-ones [(L. F. Fieser and M. A. Romero, *J. Amer. Chem. Soc.*, **75**, 4716 (1953); K. L. Williamson and W. S. Johnson, *J. Org. Chem.*, **26**, 4563 (1961)]. It was also shown that under these reaction conditions, 6β-bromocholest-4-en-3-one yielded 2α-acetoxycholest-4-en-3-one (Fieser and Romero, see above). A cognate elimination rearrangement was discovered when 4β,5-epoxy-17α-methyl-5β-androstan-17β-ol-3-one was converted to 2α-acetoxy-17α-methyltestosterone. The dipolar intermediate (or corresponding cyclopropanone) could be formed if the epoxide is first protonated and opens as shown. Subsequent attack by acetate ion at the electrophilic C-2 furnishes an intermediate which eliminates the alcoholic function at C-5



to give the product [P. L. Julian, V. Georgian, and H. C. Printy, U. S. Patent 2,910,487 (Oct 27, 1959); *Chem. Abstr.*, **54**, 2444c (1960)].

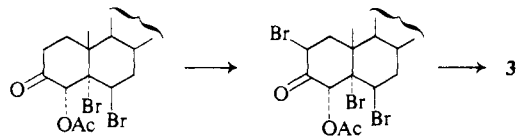
Fieser suggested that the two bromo groups in **1** undergo "allylic rearrangements" to produce the *gem*-dibromide **10** and subsequently suffer displacement by acetate ion to form the ketal acetate **11**. The final step would be a transacetylation *via* **12** to yield **2**. Whereas, an SN2′



10 **11** **12**

displacement on **1** by bromide or acetate ion is quite conceivable, and indeed was postulated for **1** → **3**, there is no precedent in the literature for an "allylic" migration of a bromo group in a ketone from one α position to the other under the basic conditions employed in the reaction **1** → **2**. Moreover, the facile conversion of the "Wendler intermediate," **3**, into **2**, employing bases other than potassium acetate, makes the participation of a second molecule of acetate, as such, unnecessary. Unfortunately, the labeling experiments cannot distinguish between the proposed and Fieser's mechanism, and partly for this reason synthesis of Fieser's *gem*-dibromide was attempted (*vide infra*).

Synthesis and Elimination–Rearrangement of 3 ("Wendler's Intermediate"). Bromination of 4α-acetoxycholest-5-en-3-one (prepared from lead tetraacetate oxidation of cholest-5-en-3-one[14]) afforded first the known relatively stable dibromide **13**,[14] into which a third bromo group was introduced by bromine in methylene chloride to furnish **14**. Addition of sodium

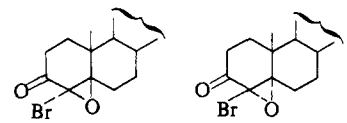


13 **14**

iodide in acetone to **14**, followed by sodium thiosulfate, produced **3** (as the 2β isomer). Treatment of **3** with a number of bases induced elimination (of HBr) and transfer of the ester group to C-3. Anhydrous potassium acetate in boiling acetone, or *sym*-collidine at 100° or N,N-dimethylaniline, each caused the smooth conversion of **3** to **2**. The fact that carboxylate ion acted as a base only received confirmation when it was shown that the reaction of **3** with 2 equiv of potassium benzoate in hot acetone yielded *only* **2** and *none* of the known benzoate analog of **2**. The last reaction was monitored by withdrawing samples from the reaction mixture at short intervals (5–10 min). Critical examination of the mass spectra of these crude products failed to reveal the presence of the benzoate analog of **2**. It was concluded, therefore, that benzoate ion served as a base to neutralize the proton at C-4, in **3** to form the anion **4**, which is subsequently transformed to **2**. Although not absolute, the facile conversion of the now synthesized "Wendler intermediate," **3** to **2**, by a number of basic reagents strongly suggests its intermediacy in the title reaction.

(14) L. F. Fieser and R. Stevenson, *J. Amer. Chem. Soc.*, **76**, 1728 (1954).

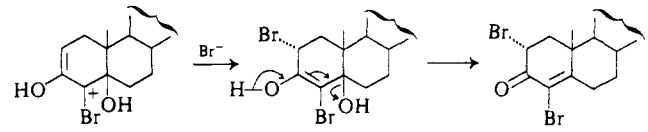
Attempted Preparation of 10 ("Fieser's Intermediate"). A Novel Synthesis of 2α,4-Dibromocholest-4-en-3-one (19). In approaching a synthesis of "Fieser's intermediate," **10**, oxidation of 4-bromocholest-4-en-3-one by alkaline hydrogen peroxide to produce the isomeric epoxides **15** and **16** was the first step. Exposure of



15 **16**

either **15** or **16** to HBr was expected to give the 4,4-dibromo-5-ol which, on losing water, should have yielded **10**. The product which was isolated failed to give a positive tetranitromethane test and its pmr spectrum ($CDCl_3$) did not show an alkene proton resonance of δ *ca.* 5.9 which would have been expected from model compounds. Instead, the pmr spectrum exhibited a clear doublet of doublets, δ 4.93 (J = 6 and 14 Hz). The chemical shift and multiplicities are characteristic of a CHBr proton signal of 2α-bromo steroids, expected at δ 4.5–5.0 and quite distinct from a methine resonance of a 6β-bromo steroid.

Although a number of different mechanisms can be written to account for this conversion, they must involve attack by bromide ion at an electrophilic C-2. To arrive at such a situation, it is suggested that HBr first protonates the epoxides, which open, and the more planar carbonium ion is best accommodated sterically and electronically by the enolic form of the ketone, see **17**. The reaction is completed by attack of Br^- at the least hindered site, C-2, to provide **18** which loses water to form **19**.



17 **18** **19**

Experimental Section

Melting points were taken on a Thomas–Hoover melting point apparatus and are uncorrected. Optical rotations, unless otherwise stated, were obtained on 5% solutions in chloroform in a 1-dm cell. Pmr data were obtained on a Varian A-60 spectrometer in $CDCl_3$ with chemical shifts measured downfield in parts per million (ppm) (δ) from tetramethylsilane as internal reference. Mass spectra were obtained on an Hitachi-Perkin-Elmer Model RMU-6D mass spectrometer with direct inlet being used in all cases. The ionizing voltage was 70 eV and the inlet system temperature (dial temperature, true sample temperature was considerably lower) was maintained at 70–120° except for 3β-acetoxy-5α-cholestan-4-one and 2β-bromo-4α-acetoxycholest-5-en-3-one for which the temperature was 330–350°. Elemental analyses were performed by Micro-Tech Laboratories, Skokie, Ill.

2α,6β-Dibromocholest-4-en-3-one (1). A solution of 5α,6β-dibromocholestan-3-one[3b] (27.2 g, 0.05 mol) in acetone (250 ml) containing fused potassium acetate (98 g, 1.0 mol) was heated under reflux for 3.5 hr. The solvent was evaporated *in vacuo*, the residue was taken up in 250 ml of methylene chloride, the solution was washed twice with 250 ml of water, and the combined water fractions were back-extracted with methylene chloride (250 ml). The combined methylene chloride fractions were evaporated to dryness *in vacuo* to give 23.1 g of 6β-bromocholest-4-en-3-one, which upon recrystallization from aqueous acetone yielded 15.2 g (66%); mp 130–132°; $[\alpha]_D$ 6.1° (lit.[5b] mp 130–132°, $[\alpha]_D$ 6°), nmr δ 4.97 (d, 1, J = 3 Hz, H_6) and 5.88 (s, 1, H_4).

To a solution of 6β-bromocholest-4-en-3-one (46.3 g, 0.1 mol) in ether (1 l.), at 0°, was added, with stirring, 20% hydrogen bromide in glacial acetic acid (2 ml), followed by a dropwise addition of a solution of bromine (16 g, 0.1 mol) in glacial acetic acid (50 ml). When the addition was completed, the solution was stirred for 0.5 hr, and the solid was filtered, giving 15.5 g. The filtrate was washed with water (two 50-ml portions), dried over sodium sulfate, and concentrated *in vacuo* to approximately one-fifth of its original volume. The filtered solid weighed 21.9 g. The two crops were combined and upon recrystallization from acetone–methanol afforded 26.8 g (50%); mp 160–162°; $[\alpha]_D$ 48.6° (lit.[5d] mp 162–163°, $[\alpha]_D$ 52°); nmr δ 4.88 (d of d, 1, J = 5 and 14 Hz, H_2), 4.93 (m, 1, H_6), and 5.97 (s, 1, H_4).

The 2β,6β isomer was prepared by literature methods.[5c,f] Most experiments were performed with the 2α,6β isomer.

3-Acetoxycholesta-2,5-dien-4-one (2). To a solution of 2α,6β-dibromocholest-4-en-3-one (**1**) (5.4 g, 0.01 mol) in acetone (50 ml) was added fused potassium acetate (19.6 g, 0.2 mol) and the mixture was heated under reflux for 5 hr. The mixture was evaporated to dryness *in vacuo*. Methylene chloride (50 ml) and water (50 ml) were added and the methylene chloride fraction was washed with water (two 50-ml portions). The aqueous fractions were back-extracted with methylene chloride (50 ml), and the combined methylene chloride fractions were evaporated to dryness *in vacuo* to give 4.6 g of material melting 148–156°, which upon recrystallization from hexane yielded 3.85 g (88%) of 3-acetoxycholesta-2,5-dien-4-one (**2**), mp 158–160°, $[\alpha]_D$ 11.7°. Thin layer chromatography showed the reaction to be essentially complete within 2 hr.

The reaction proceeded equally as well in benzene–absolute ethanol[3b,5a] and in tetrahydrofuran. The latter was employed because of its convenience in carrying out the reaction of **1** with ^{18}O-labeled potassium acetate.

Preparation of Potassium Acetate-^{18}O. Water containing 97.1 atom % oxygen-18 (0.5 g, 0.025 mol) was frozen in an ice–methanol bath at approximately −20°. To this was added acetyl chloride (2.3 g, 0.03 mol) and the mixture was agitated by bubbling nitrogen through it. The mixture was allowed to warm slowly to above the melting point of the water-^{18}O. Soon a vigorous exothermic reaction ensued. This was controlled by immediately, at the first sign of reaction, returning the flask to the ice–methanol bath. After the reaction was complete, the nitrogen stream continued to be fed through the solution for 0.5 hr to remove the hydrogen chloride produced and most of the excess acetyl chloride. The solution was used directly in the next step without purification or isolation.

To a suspension of potassium hydride (2.2 g of 40% suspension of potassium hydride in oil, 0.022 mol) which had been washed free of oil by decantation with dry tetrahydrofuran,[15] in dry tetrahydrofuran (25 ml), cooled to 0°, was added, in rapid drops, the acetic acid-^{18}O prepared above. This mixture was allowed to warm slowly (left in the ice bath which was allowed to melt and warm) to room temperature and was stirred overnight. This suspension was used in the next step without isolation.

Reaction of 2α,6β-Dibromocholest-4-en-3-one (1) with Potassium Acetate-^{18}O. To the suspension of potassium acetate-^{18}O prepared above was added a solution of **1** (2.7 g, 0.005 mol) in dry tetrahydrofuran (25 ml), and the reaction mixture was heated under reflux for 5 hr. The salts were removed by filtration, and the solvent was evaporated to dryness *in vacuo* to give 2 g of 3-acetoxycholesta-2,5-dien-4-one-^{18}O (**2**) which upon recrystallization from hexane yielded 0.75 g (34%), mp 156–158°, $[\alpha]_D$ 12.7°. Thin layer chromatography showed but a single spot.

Reduction of 3-Acetoxycholesta-2,5-dien-4-one (2). To 10% palladium on charcoal catalyst (0.02 g) was added methanol (5 ml), then a solution of 3-acetoxycholesta-2,5-dien-4-one (**2**) (0.1 g, 0.0023 mol) in methanol (20 ml). The mixture was then placed on a Parr shaker under hydrogen (40 psi). Within 15 min the hydrogen uptake was 0.5 lb and held at this pressure. The catalyst was filtered off and the solvent removed under reduced pressure. The residue was dissolved in petroleum ether (bp 30–60°) and chromatographed over activated alumina (10 g). Elution with petroleum ether afforded cholestane; 10% benzene in petroleum ether gave cholestan-4-one and benzene–petroleum ether (1:1) gave 3β-acetoxy-5α-cholestan-4-one. Identification was made by comparison of thin-layer chromatograms and mass spectra with authentic samples of 3β-acetoxy-5α-cholestan-4-one and 5α-cholestan-4-one (see below). Cholestane was identified by comparison with its published mass spectrum.[16]

Reduction of 3-Acetoxycholesta-2,5-dien-4-one-^{18}O (2). This was run as was the unlabeled compound to give upon chromatography by elution with petroleum ether, cholestane; with 10% benzene in petroleum ether, 5α-cholestan-4-one (unlabeled); and with 50% benzene–50% petroleum ether (1:1), 3β-acetoxy-5α-cholestan-4-one (labeled only in the acetate carbonyl). A portion of the reduced mixture before chromatography showed mass peaks expected for 5α-cholestan-4-one and an expected fragment thereof (*vide supra*).

5α-Cholestan-4-one-^{18}O. Water-^{18}O (one drop) was added to a solution of 5α-cholestan-4-one (10 mg, 0.026 mol) in tetrahydrofuran (five drops) in a screw-cap vial. Dry hydrogen chloride gas was blown into the vial, the vial was capped, and the solution was allowed to stand at room temperature for 1 hr. The solvent was removed *in vacuo*. Mass spectral analysis of the residue indicated approximately 50% oxygen-18-labeled 5α-cholestan-4-one as evidenced by a 1:1 ratio of the parent peaks *m/e* 386 and 388.

This material without further treatment was dissolved in petroleum ether and chromatographed over 10 g of alumina. Elution with petroleum ether (200 ml) yielded nothing; however, elution with 10% benzene in petroleum ether (300 ml) afforded approximately 8 mg of solid. Mass spectral analysis proved it to be identical with unlabeled 5α-cholestan-4-one.

3β-Acetoxy-5α-cholestan-4-one. A mixture of 3-acetoxycholesta-2,5-dien-4-one (**2**) (10 g, 0.023 mol) and 10% palladium on carbon (1 g) in methanol (300 ml) was placed on a Parr shaker under hydrogen pressure (50 psi). In 1.5 hr a total of 115 lb was taken up. The catalyst was removed by filtration through Celite and the filtrate was evaporated to dryness. The resulting oil was dissolved in methylene chloride (100 ml) and was well washed with water. The methylene chloride was removed under reduced pressure. Repeated recrystallizations of the residue from methanol yielded 3 g (30%) of product, mp 114–116°, $[\alpha]_D$ 2.2°.

Anal. Calcd for $C_{29}H_{48}O_3$: C, 78.31; H, 10.88. Found: C, 78.67; H, 10.98.

Preparation of 5α-Cholestan-4-one. a. 3β-Hydroxy-5α-cholestan-4-one. A solution of 3β-acetoxy-5α-cholestan-4-one (9 g, 0.02 mol) and potassium bicarbonate (6 g, 0.06 mol) in methanol (100 ml) was heated under reflux for 1 hr. The solution was diluted with methylene chloride (100 ml), well washed with water, and evaporated to dryness. The resulting solid (5.2 g) melted at 108–115°. One recrystallization from acetone raised the melting point to 115–121°, but subsequent recrystallizations did not improve the melting point. It was, therefore, employed as such for the next step.

b. 3β-*p*-Toluenesulfonyloxy-5α-cholestan-4-one. *p*-Toluenesulfonyl chloride (7 g, 0.944 mol) was added to a solution of 3β-hydroxy-5α-cholestan-4-one (8 g, 0.018 mol) in pyridine (30 ml) and the solution was allowed to stand overnight. It was then diluted with ether (150 ml), washed with water, then 1% hydrochloric acid (two 100-ml portions), and finally with water. The ether layer was dried and concentrated to dryness. The residue was crystallized from ether–petroleum ether yielding 7 g (70%), mp 182–184°.

c. 5α-Cholestan-4-one. A solution of 3β-*p*-toluenesulfonyloxy-5α-cholestan-4-one (1 g, 0.0018 mol) and sodium iodide (2 g) in methyl ethyl ketone (50 ml) was heated under reflux for 18 hr. The methyl ethyl ketone was removed under reduced pressure. The residue was treated with water (100 ml) and ether (100 ml). The organic layer was well washed with water and concentrated to 50 ml. The dark solution was intermittently shaken with a saturated sodium bisulfite solution (50 ml) until no further darkening of the ether layer was observed upon standing.[17] The ethereal layer was then washed with water, dilute aqueous potassium hydroxide solution, and then water, dried, and evaporated to dryness. The residue was recrystallized several times from methanol to give 0.43 g (62%), mp 99–101°, $[\alpha]_D$ 29° (lit.[18] mp 96°, $[\alpha]_D$ 27°).

Anal. Calcd for $C_{27}H_{46}O$: C, 83.86; H, 11.99. Found: C, 83.66; H, 12.00.

The "Wendler Intermediate." a. 4α-Acetoxy-2β,5α,6β-tribromocholestan-3-one (14). To a solution of 4α-acetoxy-5α,6β-dibromocholestan-3-one (**13**)[14] (10 g, 0.017 mol) in methylene chloride (100 ml) was added, dropwise, 10% bromine in glacial acetic acid

(15) Tetrahydrofuran was dried first over potassium hydroxide, then distilled from sodium hydride.

(16) K. Bieman, "Mass Spectrometry, Organic Chemical Applications," McGraw-Hill Book Co., Inc., New York, N. Y., 1962, p 339.

(17) P. L. Julian and W. J. Karpel, *J. Amer. Chem. Soc.*, **72**, 362 (1950).

(18) D. N. Jones, J. R. Lewis, C. W. Shoppee, and G. H. R. Summers, *J. Chem. Soc.*, 2876 (1955).

(28 ml). The reaction was initiated with a few drops of 30% hydrogen bromide in acetic acid. The reaction mixture was diluted with methylene chloride (100 ml) and the organic layer washed with water until the layer was almost colorless. The methylene chloride solution was evaporated to dryness *in vacuo*, and the residue was recrystallized from acetone to yield 8.4 g (74%), mp 173–174°, $[\alpha]_D$ 17°.

Anal. Calcd for $C_{29}H_{45}Br_3O_3$: C, 51.07; H, 6.66; Br, 35.23. Found: C, 51.33; H, 6.59; Br, 35.05.

b. 2β-Bromo-4α-acetoxycholest-5-en-3-one (3). To a solution of 4α-acetoxy-2β,5α,6β-tribromocholestan-3-one (**14**) (8.2 g, 0.012 mol) in acetone (250 ml) was added with rapid stirring, sodium iodide (8.2 g, 0.055 mol) and the mixture was stirred for 5 min. To this mixture was then added 5% aqueous sodium thiosulfate solution (200 ml). The resulting oil quickly crystallized from ether–methanol, and then from ethanol; 3.4 g (54%); mp 116.5–118°; $[\alpha]_D$ 114°; nmr δ 2.23 (s, 3, Ac), 2.32 (d, 1, J = 6.2 Hz, H_2), 5.78 (m, 1, H_6), and 6.32 (m, 1, H_4).

Anal. Calcd for $C_{29}H_{45}BrO_3$: C, 66.75; H, 8.70; Br, 15.34. Found: C, 66.68; H, 8.58; Br, 15.41.

The configuration of the bromine atom at the 2 position was determined by molecular rotation data (in degrees) as follows {$[M]_D$ ($\Delta[M]_D$)}: cholest-5-en-3-one,[19] −9.6; 2α-bromocholest-5-en-3-one,[5f] +2.3 (+11.9); 2β-bromocholest-5-en-3-one,[5f] +532.0 (+541.6); 4α-acetoxycholest-5-en-3-one,[14] −139; 2-bromo-4α-acetoxycholest-5-en-3-one (**3**), +568 (+697). The $[M]_D$ of that product *vs.* its parent much more closely resembles that of 2β-bromocholest-5-en-3-one *vs.* its parent as compared to the 2α isomer.

Reaction of 2β-Bromo-4α-acetoxycholest-5-en-3-one (3). a. With Acetate Ion. To a solution of **3** (2.08 g, 0.004 mol) in acetone (20 ml) was added fused potassium acetate (7.8 g, 0.08 mol) and the mixture was heated under reflux for 5 hr. The acetone was evaporated *in vacuo* and the residue was dissolved in ether (50 ml) and water (50 ml). The ether layer was washed with water (two 50 ml portions) and the combined ether extracts were dried over sodium sulfate and evaporated to dryness to give 1.4 g, mp 157–159°, $[\alpha]_D$ 11.8°. A mixture melting point with an authentic specimen of 3-acetoxycholesta-2,5-dien-4-one (**2**) showed no depression.

b. With Benzoate Ion. To a solution of 2β-bromo-4α-acetoxycholest-5-en-3-one (**3**) (2.08 g, 0.004 mol) in acetone (20 ml) was added potassium benzoate (1.28 g, 0.009 mol). The resulting mixture was heated under reflux for 2 hr. During the reaction 0.5-ml samples were removed each minute for the first 10 min, then each 5 min to 1 hr. Each sample was immediately diluted with ether, washed with water, dried over sodium sulfate, and concentrated to dryness under reduced pressure. The remainder of the reaction mixture was treated similarly at the end of the 2 hr. Mass spectra of these fractions showed only starting material and/or 3-acetoxycholesta-2,5-dien-4-one (**2**). No 3-benzoyloxy compound (*m/e* 502) was formed, indicating that the acetate moves from C-4 to C-3 *via* an intramolecular pathway which could very likely be an ortho ester, and the potassium benzoate acted solely as a base.

c. With Collidine. A solution of 2β-bromo-4α-acetoxycholest-5-en-3-one (**3**) (0.6 g, 0.001 mol) in collidine (2.6 ml) was heated on a steam bath for 20 hr. The mixture was poured into 10% hydrochloric acid (75 ml) and extracted with ether (100 ml). The ether solution was washed with 10% hydrochloric acid (25 ml), water (three 100-ml portions), dried, and evaporated to dryness. The residue was recrystallized from acetone to yield 0.4 g (91%) of 3-acetoxycholesta-2,5-dien-4-one (**2**), mp 158.5–159°. Mixture melting point with an authentic sample showed no depression.

d. With N,N-Dimethylaniline. A solution of **3** (0.9 g, 0.0015 mol) and N,N-dimethylaniline (3.6 g, 0.03 mol) in acetone (10 ml) was heated under reflux for 5 hr. It was concentrated to near dryness and diluted with water (50 ml) and ether (50 ml). The organic layer was washed with water (two 50-ml portions), 10% concentrated hydrochloric acid (two 50-ml portions), then water (two 50-ml portions), dried, and concentrated to dryness. The residue was recrystallized from acetone to yield 0.51 g (77%) of 3-acetoxycholest-2,5-dien-4-one (**2**), mp 157–159°. Mixture melting point with authentic material showed no depression.

Attempted Preparation of 10 ("Fieser's Intermediate"). 4-Bromocholest-4-en-3-one. The literature preparation of the intermediate epoxide by Shaw and Stevenson[20] did not give satisfactory results in our hands and the following modification was found to give a better product.

4β,5-Epoxy-5β-cholestan-3-one. To a solution of cholest-4-en-3-one (76.8 g, 0.2 mol) in methanol (7.5 l.) was added at 20°, simultaneously and with stirring, 30% hydrogen peroxide solution (310 ml), and 5 *N* potassium hydroxide solution (310 ml). Addition time was *ca.* 20 min. The reaction was followed by monitoring the ultraviolet absorption at 241 mμ.

The solid (39.5 g) was filtered and was recrystallized from acetone–methanol to yield the pure product, 35.2 g (44%), mp 116.5–118.5°, $[\alpha]_D$ 118.5° (lit.[20] mp 118–119°, $[\alpha]_D$ 128°), but we have not been able to confirm this rotation. Thin layer chromatography showed but one spot.

4-Bromocholest-4-en-3-one. The substitution of HBr in acetic acid for aqueous HBr gave more reproducible results for this conversion.

A suspension of 4β,5-epoxy-5β-cholestan-3-one (60 g, 0.15 mol) in glacial acetic acid (180 ml) was chilled to 15°. To this was added, slowly, cold 20% hydrogen bromide in acetic acid (240 ml). The temperature rose spontaneously to 25°, and the mixture became viscous upon initial addition. The reaction mixture was stirred for 0.5 hr at 15–16°, then poured into ice water (3 l.) and collected by filtration. The solid weighed 69.5 g and yielded upon recrystallization from acetone–methanol, 57 g (82%), mp 114–116°, $[\alpha]_D$ 107° (lit.[20] mp 114–115°, $[\alpha]_D$ 107°).

4-Bromo-4,5-epoxycholestan-3-ones (15 and 16). To a suspension of 4-bromocholest-4-en-3-one (13.9 g, 0.03 mol) in methanol (1.4 l.) at 20°, was added, with stirring, and simultaneously, in rapid drops, 30% hydrogen peroxide solution (42 ml) and 5 *N* potassium hydroxide solution (42 ml). There was an initial temperature rise to 23° and the addition took place at 23–25°. The reaction was monitored by ultraviolet absorption at 260 mμ.

The product (7.5 g) which had separated from the solution was filtered. The filtrate was diluted with water (1 l.) and made neutral with acetic acid (40 ml). Although turbidity developed, no filterable material was obtained. The product was purified as follows. It was dissolved in ethyl acetate (22 ml) in the cold, filtered, and treated with methanol (22 ml). The first crop gave 2.5 g which yielded upon a second recrystallization from cold ethyl acetate (60 ml) and methanol (30 ml), 1.4 g (10%), mp 120–121°, $[\alpha]_D$ −64.2°.

Anal. Calcd for $C_{27}H_{43}BrO_2$: C, 67.59; H, 9.04; Br, 16.68. Found: C, 67.74; H, 8.88; Br, 16.28.

The second crop (obtained without concentration) gave 1.0 g which yielded upon recrystallization, in the cold, from ethyl acetate (10 ml) and methanol (10 ml), 0.4 g (6%), mp 106° sharp, $[\alpha]_D$ 149.2°.

Anal. Calcd for $C_{27}H_{43}Br_2O$: C, 67.59; H, 9.04; Br, 16.68. Found: C, 67.74; H, 9.01; Br, 16.59.

Comparing the following data—4β,5-epoxy-5β-cholestan-3-one[20] $[\alpha]_D$ +128°, 4α,5-epoxy-5α-cholestan-3-one[21] $[\alpha]_D$ −42°—it seems apparent that the following configuration can be assigned: 4α-bromo-4β,5-epoxy-5β-cholestan-3-one (**15**), mp 106°, $[\alpha]_D$ 149.2°; 4β-bromo-4α,5-epoxy-5α-cholestan-3-one (**16**), mp 120–121°, $[\alpha]_D$ −64.2°.

2α,4-Dibromocholest-4-en-3-one (19). To a suspension of 4β-bromo-4α,5-epoxy-5α-cholestan-3-one (**16**) (1.1 g, 0.019 mol) in glacial acetic acid (60 ml), at 15°, was added slowly 20% hydrogen bromide in acetic acid (75 ml), and the reaction mixture was stirred for 0.5 hr. The mixture was poured into cold water (2.5 l.). The solid was collected by filtration and weighed 9.5 g. Three recrystallizations from acetone yielded analytical material, 1.5 g (14.7%), mp 177–178°, $[\alpha]_D$ 82.5°, nmr δ 4.83 (d/d, 1, J = 5 and 14 Hz, H_2).

For comparison, 2α-bromocholest-5-en-3-one shows nmr δ 4.77 (d/d, 1, J = 5 and 13 Hz, H_2) and 5.95 (s, 1, H_4).

Anal. Calcd for $C_{27}H_{42}Br_2O$: C, 59.74; H, 7.81; Br, 29.50. Found: C, 60.06; H, 8.08; Br, 29.51.

Treatment of the product (1.1 g, 0.002 mol) with sodium iodide (1.5 g, 0.01 mol) in acetone (25 ml) under reflux for 20 hr, followed by treatment with 10% sodium bisulfite solution as per Julian and Karpel[17] afforded 4-bromocholest-4-en-3-one, 0.58 g (64.5%), mp and mmp 112–114°.

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