[Chem. Pharm. Bull. 18(3) 608—616 (1970)]

UDC 547.92.04

Thiosteroids. XXV.[1] Syntheses of the Epimers of Cholest-4- and -5-en-2,3-episulfide and Related Compounds

Taichiro Komeno and Hikaru Itani

Shionogi Research Laboratory, Shionogi & Co., Ltd.[2]

(Received August 25, 1969)

Cholest-4-en-2β,3β-episulfide and the epimers of cholest-5-en-2,3-episulfide were synthesized upon treatment of the corresponding thiocyanatohydrin acetate with alkali. However, synthesis of cholest-4-en-2α,3α-episulfide in a similar way failed. This could be achieved successfully but in low yield by treatment of 3β-acetoxycholest-4-en-2α-ethyl xanthate with alkali. CD and ORD curves of these episulfides were studied.

Although a number of stilbene episulfide derivatives are known,[3] only a few aliphatic episulfides containing a conjugated double bond were synthesized.[4] In connection with our continuing CD and ORD studies on the steroidal episulfides,[5] this paper describes the syntheses of epimers of cholest-4- and -5-en-2,3-episulfide.

Oxidation of cholest-2-en-5α-ol (I)[6] with *m*-chloroperbenzoic acid afforded 5α-hydroxy-2α,3α-epoxide (II), configuration of which was confirmed by the fact that an absorption band due to intramolecularly hydrogen bonded hydroxyl group was observed at 3503 cm^{-1} in the infrared (IR) spectrum ($\Delta\nu$=117 cm^{-1}). Diaxial ring-opening of the oxide (II) with thiocyanic acid gave 3α,5α-dihydroxy-2β-thiocyanate (IIIa) in 64% yield, which was converted into the monoacetate (IIIb) and the monomesylate (IIIc) with pyridine-acetic anhydride and pyridine-mesyl chloride, respectively. The mesylate (IIIc) was treated with alkali to give 5α-hydroxy-2β,3β-episulfide (IV), whose IR spectrum exhibits no absorption band due to hydrogen bonded hydroxyl group (ν_{OH}=3610 cm^{-1}). An attempt to dehydrate the hydroxyepisulfide (IV) with pyridine-thionyl chloride failed and afforded an ambiguous result. On the other hand, dehydration of the thiocyanatohydrin acetate (IIIb) with the same reagent gave a mixture of the crystalline 5-ene (V) and the oily 4-ene derivative (VI) in a ratio of 10: 5.5. The structure of the former compound (V) was determined by conversion with alkali to cholest-5-en-2β,3β-episulfide (VII), which exhibits an absorption maximum characteristic of the episulfide group at 261.5 mμ in the ultraviolet (UV) spectrum. Alkaline treatment of the 4-ene derivative (VI) afforded an unstable substance which decomposed upon attempted recrystallization. However, the substance isolated by direct filtration from the reaction mixture was found to be the expected cholest-4-en-2β,3β-episulfide (VIII) from the following observation. In its nuclear magnetic resonance (NMR) spectrum two protons attached to the episulfide moiety occur centered at 6.53 τ as a multiplet and in the UV spectrum the absorption maximum appears at 288 mμ which is shifted to the longer wave length ($\Delta\lambda$=26 mμ) compared to that

1) Part XXIV: T. Komeno and K. Takigawa, *Chem. Pharm. Bull.* (Tokyo), **18**, 43 (1970).
2) Location: *Fukushima-ku, Osaka.*
3) Recently the review of episulfides was published: M. Sander, *Chem. Rev.*, **66**, 297 (1966).
4) *e.g.*, 1,2-Epithio-3-butene: C.C.J. Culvenor, W. Davies and N.S. Heath, *J. Chem. Soc.*, **1949**, 278. Unsuccessful synthesis of α,β-epithioester was also reported. C.C. Tung and A.J. Speziale, *J. Org. Chem.*, **29**, 1577 (1964) and cited ref.
5) *a*) C. Djerassi, H. Wolf, D.A. Lightner, E. Bunnenberg, K. Takeda, T. Komeno and K. Kuriyama, *Tetrahedron*, **19**, 1547 (1963); *b*) K. Kuriyama, T. Komeno and K. Takeda, *Tetrahedron*, **22**, 1039 (1966); *c*) K. Kuriyama, T. Komeno and K. Takeda, *Ann. Rept. Shionogi Res. Lab.*, **17**, 66 (1967).
6) R.B. Clayton, H.B. Henbest and M. Smith, *J. Chem. Soc.*, **1957**, 1982.

of VII. This bathochromic shift indicates the conjugation of the episulfide group with the double bond as is found in butyl vinyl sulfide.[7]

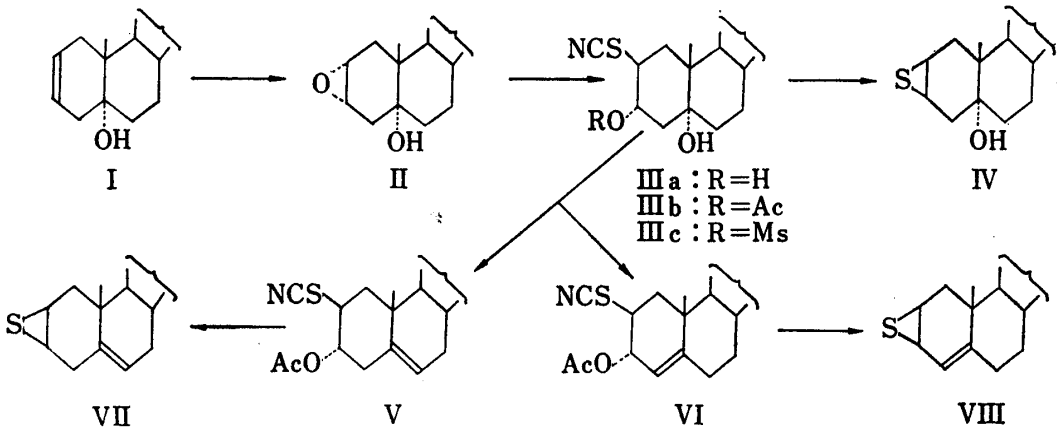

Chart 1

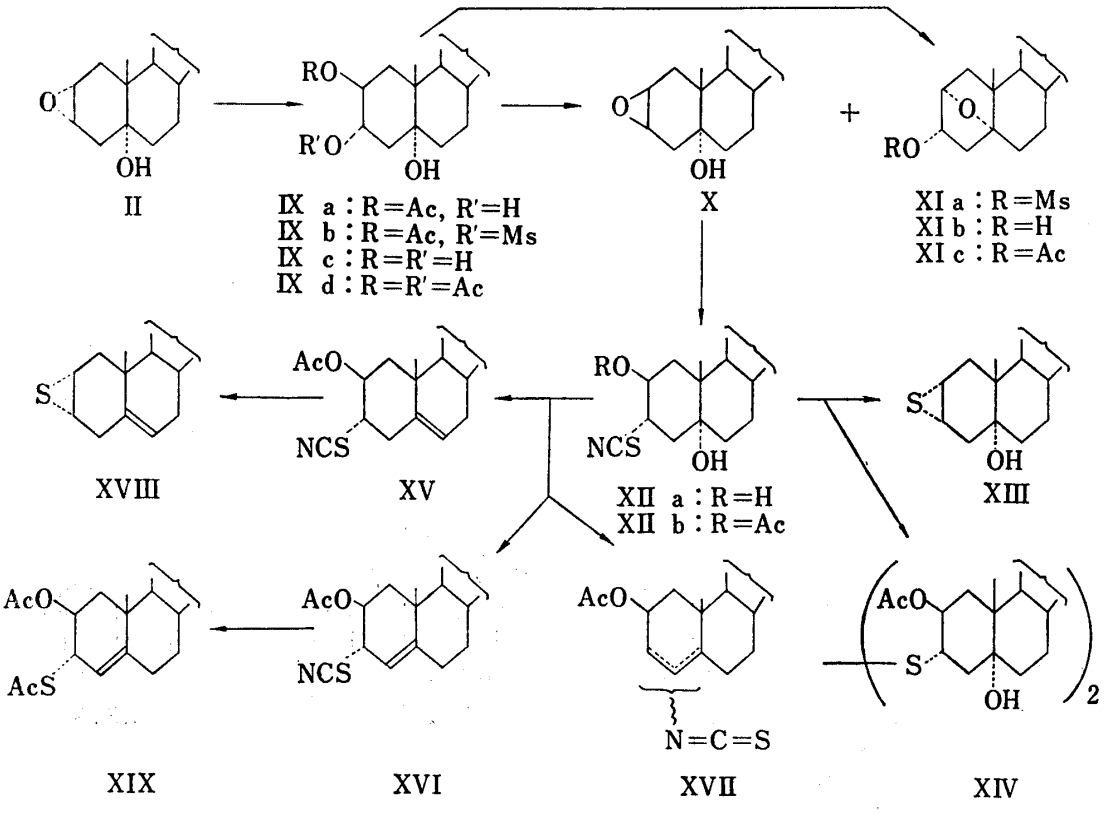

Chart 2

Since the reaction of cholest-2-en-5α-ol with hypobromous acid afforded a complex mixture, 5α-hydroxycholestan-2β,3β-epoxide (X) was synthesized from the 5α-hydroxy-2α,3α-epoxide (II) in 3 steps involving the cleavage of the oxide with acetic acid, followed by mesylation and alkaline treatment in a similar manner as described by Fürst and Plattner.[8] In the course of the synthesis of X when the reactions were run without isolation of the intermediates in each step a small amount of the unexpected compound (XIa), $C_{28}H_{48}O_4S$, which shows characteristic absorption bands due to a mesyloxy group in the IR spectrum, was accompanied.

7) In the UV spectrum of butylvinylsulfide the absorption maximum was observed at 229 mμ (*cf.* dibutylsulfide λ_{max} 209 mμ) M. Procházka and M. Palecek, *Collection Czech. Chem. Commun.*, 32, 3149 (1967).

This finding led us to investigate the reactions stepwise. After the reaction of II with acetic acid separation of the product using thin-layer chromatography (TLC) on a preparative scale gave 73.9% of the triol 2-monoacetate (IXa), 10.7% of an unidentified triol monoacetate, 1.7% of the triol (IXc), and 4.9% of the starting material. On the other hand, treatment of the α-epoxide (II) with perchloric acid in aqueous dioxane afforded 19.3% of the compound (XIb) and 65% of the triol (IXc). This triol (IXc) on treatment with mesyl chloride-pyridine furnished quantitatively the foregoing mesylate (XIa), which was reduced with lithium aluminum hydride to the compound (XIb). In the NMR spectra of the compounds (XIa) and (XIb) one proton attached to the carbon bearing the ether-oxygen occurs at 5.47 and 5.82 τ as a doublet ($J \approx 6.0$, 6.5 cps) respectively and vicinal coupling between its proton and the well-resolved proton attached to the carbon carrying a mesyloxy and a hydroxyl group, respectively, was not observed. These NMR spectra can be interpreted as *exo* substituted 7-oxabicyclo[2,2,1]heptane derivatives.[9] Therefore, we assumed tentatively the structures for XIa and XIb as 3α-mesyloxy- and 3α-hydroxy-2α,5α-epoxide. The elucidation by chemical means will be reported in the following paper.

Treatment of the β-epoxide (X) with thiocyanic acid afforded dihydroxythiocyanate (XIIa) in 57% yield, which was further transformed into the acetate (XIIb). Whereas the alkaline treatment of IIIb gave only the episulfide, the same treatment of XIIb yielded 45% of the hydroxyepisulfide (XIII) along with 35% of the disulfide (XIV). A similar and more facile dimerization of the 3α-thiol as compared to the 2β-thiol was stated in the previous report already.[10] The structure of the hydroxyepisulfide (XIII) was supported by the IR spectrum which shows an absorption band due to the hydrogen bonded hydroxyl group at 3448 cm^{-1} ($\Delta\nu = 162$ cm^{-1}). Dehydration of the dihydroxythiocyanate monoacetate (XIIa) with pyridine-thionyl chloride afforded a mixture of three compounds, from which the 5-ene derivative (XV) was readily isolated by crystallization and converted to cholest-5-en-2α,3α-episulfide

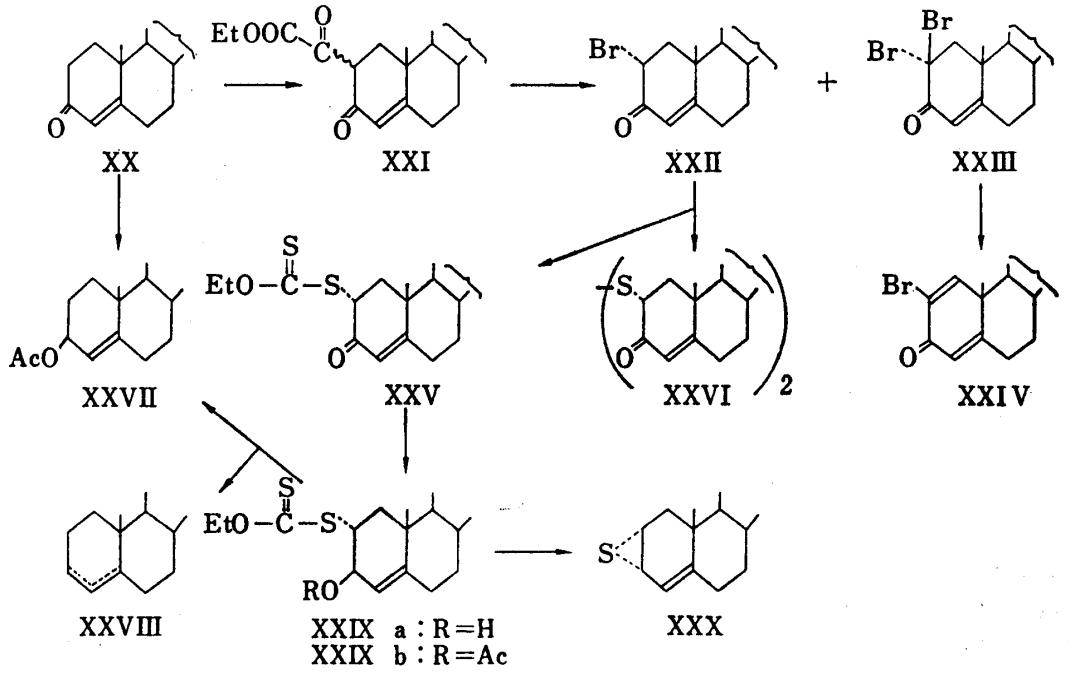

Chart 3

8) A. Fürst and P.A. Plattner, *Helv. Chim. Acta*, 32, 275 (1949).
9) In general, the NMR spectrum of 2-*exo*-substituted 7-oxabicyclo[2,2,1]heptane shows its bridge head proton at C_1 as a doublet ($J \approx ca.$ 5.0 cps). D. Gagnaire and E.P. Subiza, *Bull. Soc. Chim. France*, **1963**, 2627; F.A.L. Anet, *Can. J. Chem.*, **39**, 789 (1961); T.J. Flautt and W.F. Erman, *J. Am. Chem. Soc.*, **85**, 3212 (1963).

(XVIII) with alkali. The remaining two compounds, exhibiting a strong absorption band characteristic of an isothiocyanate group at 2810 cm^{-1} in the IR spectrum, were separated by preparative TLC. But each component isolated here showed again original two spots on TLC. This observation indicates that the rearrangement of the allyl thiocyanate (XVI) to the allyl isothiocyanate (XVII) is taken place. Reduction of the mixture with lithium aluminum hydride, followed by acetylation gave the products, from which the pure dihydroxythiol O,S-diacetate (XIX), was isolated as a crystalline form. The structure of XIX was characterized on the basis of the NMR spectrum. Unfortunately, treatment of XIX with alkali afforded a complex mixture and the expected episulfide was not isolated. This failure led us to synthesize 3β-acetoxycholest-4-en-2α-ethyl xanthate (XXIXb), in which the trans relationship between substituents leading to the episulfide group is maintained and the acetoxyl function is in an allylic position.

Slightly different from the observation by Allen and Weiss,[11] bromination of 2-ethyloxalylcholestenone (XXI) gave the 2α-bromide (XXII) as well as 2,2-*gem*-dibromide (XXIII). Physical constants of the latter compound are quite different from those described by Shoppee.[12] However, the structure of our compound is supported on the basis of the NMR spectrum, which shows no proton attached to the carbon bearing bromine atom but AB pattern at 6.90 τ as a pair of doublets due to the 1-methylene moiety in addition to one vinyl proton at 4.18 τ. Dehydrobromination of the compound gave 2-bromocholesta-1,4-dien-3-one, whose physical constants are in accord with those reported by Shoppee.[12] Treatment of the monobromide (XXII) with potassium ethyl xanthate yielded 41% of the ethyl xanthate (XXV) accompanied by 11% of the disulfide (XXVI). In both compounds 2α-configuration of the substituent was confirmed by the NMR spectra.[13] Reduction of the ethyl xanthate (XXV) with sodium borohydride, followed by acetylation afforded 3β-acetoxycholest-4-en-2α-ethyl xanthate (XXIXb). The proof for the structure was gained from the fact that desulfurization with Raney nickel yielded cholest-4-en-3β-ol acetate (XXVII) and a mixture of cholest-4- and -3-ene (XXVIII). Although the reaction of the ethyl xanthate (XXIXb) with alkali seemed to proceed smoothly judging from TLC, actual isolation by means of filtration after

Table I. CD and ORD Data of Epimers of Cholesten-2,3-episulfides (in Isooctane)

Compound	Configuration of episulfide	Position of unsaturation	CD		ORD				
					Peak		Trough		
			mμ	$[\theta]\times10^{-2}$	mμ	$[\phi]\times10^{-2}$	mμ	$[\phi]\times10^{-2}$	a
	β[a]		264	+ 6.0[b]	272	+ 12.7	254	+ 7.0[b]	+ 6
VII	β	5.6	261	− 34.8[c]	249	+ 18.2	274	−27.7[c]	− 46
					225	+ 23.1	236	+ 8.7	
VIII	β	4.5	289	−164.8	274.5	+143.3	309	−65.9	−209
			243	− 66.9	233.5	+157.1	251	+98.4	
			226.5	+ 86.0			216.5	−60.1	
	α[d]		268	− 38.4[b]	257	+ 53.4	280	−11.0[b]	− 64
XVIII	α	5.6	267	− 39.3	250[sh]	+ 33.0	279	−26.3	− 59
XXIX	α	4.5	282	+143.0	297	+122.8	268	−73.0	+196
			210	+253.6	221	+102.1	210	+86.5	

a) 5α-cholestan-2β,3β-episulfide b) ref. 4a) c) values determined in MeOH solution d) 5α-cholestan-2α,3α-episulfide

10) K. Takeda, T. Komeno, J. Kawanami, S. Ishihara, H. Kadokawa, H. Tokura and H. Itani, *Tetrahedron*, **21**, 329 (1965).

11) G.R. Allen, Jr. and M.J. Weiss, *J. Am. Chem. Soc.*, **82**, 2840 (1960). They reported that bromination of 2-ethyloxalyl-16α,17α-isopropylidene-dioxyprogesterone gave 2α-bromide and 2α,4-dibromide.

12) C.W. Shoppee, R.E. Lack and J. Scott, *J. Chem. Soc.*, **1962**, 2233. The compound assigned by them to this structure was obtained by bromination of 2α-bromocholestanone in hot AcOH–AcOK, followed by alumina chromatography.

13) R.J. Abraham and J.S.E. Holker, *J. Chem. Soc.*, **1963**, 806.

dilution of the reaction mixture with water resulted in low yield (18%) of cholest-4-en-2α,3α-episulfide (XXX), whose UV spectrum exhibits a red-shifted absorption maximum at 279 mμ. Attempted extraction of the mother liquor in the usual way was unsuccessful and underwent the decomposition of XXX. Heating of the solution of XXX in petroleum ether also caused its decomposition to give the less polar substance (probably cholestadiene) and a material insoluble in the solvent.

The CD and ORD data of cholest-4- and -5-en-2,3-episulfides are summarized in Table. These observed Cotton effects are in good agreement with those predicted by the sector rule for episulfides.[5b] Moreover, the Cotton effects of the conjugated episulfides reflect those UV maxima and are shifted to the longer wave length.

Experimental[14]

5α-Hydroxycholestan-2α,3α-epoxide (II)——To a stirred solution of 1.0 g of cholest-2-en-5α-ol (I)[6] in 4 ml of $CHCl_3$, a solution of 600 mg of *m*-chloroperbenzoic acid in 10 ml of $CHCl_3$ was added dropwise for 12 min. The resulting mixture was agitated at room temperature for 35 min, poured into 10% Na_2CO_3, and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with water, dried over Na_2SO_4, and evaporated to dryness under reduced pressure. The residual solid (1.052 g) was recrystallized from acetone to give 994 mg (95.5%) of II, mp 143—145°, $[\alpha]_D^{22}$ +22.5±2° (c=0.952). IR ν_{max} cm^{-1}: 3480, 3450, 877, 810, 803. *Anal.* Calcd. for $C_{27}H_{46}O_2$: C, 80.54; H, 11.52. Found: C, 80.81; H, 11.32.

3α,5α-Dihydroxycholestane-2β-thiocyanate (IIIa)——To a mixture of 2.5 g of KSCN dissolved in a small volume of ice water and 10 ml of ether, 3.5 g of H_3PO_4 was added in small portions and the mixture was shaken to extract the HSCN formed into the ether layer. The pink-colored HSCN–ether solution was dried over Na_2SO_4 and added to a solution of 502 mg of the epoxide (II) in 5 ml of ether. After stirring for 2 hr, the ethereal layer was washed with 10% Na_2CO_3 and water, dried, and evaporated to dryness under reduced pressure. The residue (579 mg) was recrystallized from MeOH giving 385 mg (66.7%) of IIIa, mp 139—141°, $[\alpha]_D^{23}$ +19.9±0.4° (c=1.101). IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3443, 2153, 1055, 870. CD (in MeOH) $[\theta]_{251m\mu}^{max}$ +1550. NMR τ: (18-H) 9.35, (19-H) 8.96, (2α-H) 6.08, (3β-H) 5.87. *Anal.* Calcd. for $C_{28}H_{47}O_2NS$: C, 72.83; H, 10.26; N, 3.06; S, 6.94. Found: C, 72.60; H, 10.41; H, 2.94; S, 6.91. Acetylation of IIIa with pyridine–Ac_2O in the usual way afforded the monoacetate (IIIb), which was recrystallized from acetone–hexane to yield the pure sample, mp 163—165°, $[\alpha]_D^{24}$ +81.1±1° (c=1.060). IR ν_{max} cm^{-1}: 3440, 2172, 1717, 1284, 1044. CD (in MeOH) $[\theta]_{248m\mu}^{max}$ +3300. NMR τ: (18-H) 9.33; (19-H) 8.96; (AcO) 7.89; (2α-H) 5.91 d-t (J≈9.0, 8.3 cps), (3β-H) 4.81 d-q (J≈9.0, 8.0, 5.0 cps). *Anal.* Calcd. for $C_{30}H_{49}O_3NS$: C, 71.53; H, 9.80; N, 2.78; S, 6.37. Found: C, 71.78; H, 10.02; N, 3.00; S, 6.57.

5α-Hydroxycholestan-2β,3β-episulfide (IV)——To a cooled solution of 230 mg of IIIa in 2.3 ml of pyridine, 0.23 ml of MsCl was added. The resulting mixture was allowed to stand at room temperature overnight, then poured into ice water and extracted with ether. The ethereal layer was washed successively with 10% HCl, 10% Na_2CO_3 and water, and dried over Na_2SO_4. After removal of the solvent 267 mg of the solid (IIIc) was obtained. Without purification the mesylate (IIIc) was treated with 270 mg of K_2CO_3 in a mixture of 3.6 ml of dioxane, 3.6 ml of MeOH, and 1.0 ml of water. The reaction mixture was stirred for 2 hr, poured into ice water, and extracted with ether. The ethereal layer was washed with water, dried, and evaporated to dryness. The residue (222 mg) was recrystallized from pentane to afford 120 mg of IV, mp 105—107°, $[\alpha]_D^{24}$ +11.5±0.3° (c=1.102). IR ν_{max} cm^{-1}: 3428. NMR τ: (18-H) 9.36, (19-H) 8.95, (2α- and 3α-H) *ca.* 6.70m (2H). *Anal.* Calcd. for $C_{27}H_{46}OS$: C, 77.45; H, 11.07; S, 7.66. Found: C, 77.02; H, 11.03; S, 7.69.

Dehydration of 3α,5α-Dihydroxycholestane-2β-thiocyanate 3-Acetate (IIIb)——To a stirred solution of 705 mg of IIIb in 17 ml of pyridine, 3.5 ml of $SOCl_2$ was added dropwise at 0° for 4 min. The reaction mixture was agitated with cooling for an additional 10 min, then poured into ice water, and extracted with ether. The ethereal layer was washed successively with 10% HCl, 10% Na_2CO_3, and water, dried over

14) All melting points were measured on a Kofler hot-stage apparatus and are uncorrected. Optical rotations were determined in 1% EtOH–$CHCl_3$ with a Perkin Elmer Polarimeter, type 141. Unless otherwise stated UV spectra were recorded in isooctane with a Hitachi EPS-2 spectrophotometer and IR spectra in Nujol mulls by use of a Koken DS-201B spectrophotometer. Intramolecular hydrogen bonding was measured in about 1.5×10^{-3}M CCl_4-solution using 20 mm cell. CD and ORD curves were determined with a Jasco Model ORD/UV-5 equipped with CD. All NMR spectra were run in $CDCl_3$ solutions with a Varian A-60 spectrometer, TMS serving as internal standard. Apparent coupling constants were obtained from the 1st order analysis. For preparative TLC silicagel G (E. Merck Co.) was used as an adsorbent.

Na_2SO_4, and evaporated under reduced pressure. The residue, which shows 2 spots on TLC, was recrystallized from ether-petr. ether yielding 389 mg (56.8%) of V, mp 132—133°, $[\alpha]_D^{24}$ +44.6±0.7° (c=0.944). IR ν_{max} cm^{-1}: 2166, 1746, 1238, 1030, 789. CD (in MeOH) $[\theta]_{249m\mu}^{max}$ +3580. NMR τ: (18-H) 9.31, (19-H) 8.94, (OAc) 7.94, (2α-H) 6.56, (3α- and 6-H) 4.93 (1H), 4.60 (1H). *Anal.* Calcd. for $C_{30}H_{47}O_2NS$: C, 74.18; H, 9.75; N, 2.88; S, 6.60. Found: C, 74.42; H, 9.59; N, 2.95; S, 6.67. The mother liquor afforded 305 mg (44.5%) of an oily substance, which exhibits two spots on TLC and was purified by preparative TLC using cyclohexane–AcOEt (4:1) as developing solvent. Elution of the adsorbent corresponding to the spot (*Rf* =0.50, *cf.* *Rf* for V=0.43) with 800 ml of ether gave 212 mg (30.9%) of pure VI, which could not be crystallized and was used for the next step. IR $\nu_{max}^{CCl_4}$ cm^{-1}: 2190, 1744, 1669, 1213, 1024. NMR τ: (18-H) 9.32 (19-H) 8.90, (OAc) 7.89, (2α-H) 6.60, (3β- and 4-H) 4.83 (1H) 4.65 (1H).

Cholest-5-en-2β,3β-episulfide (VII)——To a solution of 163 mg of KOH in 6 ml MeOH, a solution of 150 mg of V in 2 ml of tetrahydrofuran was added. The reaction mixture was stirred at room temperature for 1 hr, then poured into ice water, and extracted with ether. The usual work-up afforded 119 mg of crystals, which were recrystallized from ether–acetone to give 110 mg (92.2%) of VII, mp 103—104°, $[\alpha]_D^{23}$ −50.8±0.8° (c=0.921). IR ν_{max} cm^{-1}: 999, 955, 848, 801. UV λ_{max} mμ (ε): 261.5 (58). NMR τ: (18-H) 9.33, (19-H) 8.99, (2β- and 3β-H) *ca.* 6.83 m (2H), (6-H) 4.59. *Anal.* Calcd. for $C_{27}H_{44}S$: C, 80.93; H, 11.07; S, 8.00. Found: C, 80.94; H, 10.96; S, 8.21.

Cholest-4-en-2β,3β-episulfide (VIII)——To a solution of 250 mg of KOH in 5 ml of MeOH, a solution of 194 mg of VI in 1.1 ml of tetrahydrofuran was added. The reaction mixture was stirred for 2 hr. The appeared crystals were collected by filtration, washed with water, and dried at room temperature under reduced pressure. mp 74—76°, $[\alpha]_D^{23}$ −37.0±0.6° (c=0.960). IR ν_{max} cm^{-1}: 3031, 1594, 865, 785, 724, 692. UV λ_{max} mμ (ε): 288 (57) 221.5 (8400). NMR τ: (18-H) 9.33, (19-H) 8.89, (2β- and 3β-H) *ca.* 6.53 (2H), (4-H) 4.28. *Anal.* Calcd. for $C_{27}H_{44}S \cdot \frac{1}{3}H_2O$: C, 79.74; H, 11.07; S, 7.88. Found: C, 79.79; H, 10.83; S, 7.85.

5α-Hydroxycholestan-2β,3β-epoxide (X)——a) A solution of 1.005 g of II in 10 ml of AcOH was heated on a steam bath for 3.5 hr. After dilution with ice water the mixture was extracted with ether. The ethereal layer was washed with 10% Na_2CO_3 and water, dried, and evaporated to dryness. To a cooled solution of the residual solid (1.123 g) in 20 ml of pyridine 2 ml of MsCl was added. The mixture was allowed to stand under cooling for 3.5 hr, then poured into ice water, and extracted with ether. The ethereal layer was washed successively with 10% HCl, 10% Na_2CO_3 and water, dried, and concentrated under reduced pressure. The residue (1.613 g) was heated under reflux in a solution of 2 g of KOH in 90 ml of absolute MeOH for 30 min. The reaction mixture was poured into ice water and extracted with ether. The usual work-up gave 1.045 g of the product, which was chromatographed over 20 g of standardized Al_2O_3. The fractions eluted with petr. ether–benzene (1:1) yielded 336 mg of a mixture. The fractions eluted with petr. ether–benzene (1:1) and benzene afforded 461 mg of X, which was recrystallized from aqueous acetone to yield 446 mg of the pure sample, mp 137—139°. $[\alpha]_D^{20}$ +33.2±0.6° (c=1.122). IR ν_{max} cm^{-1}: 3492, 1060, 813. NMR τ: (18-H) 9.36, (19-H) 9.02, (OH), 8.72, (2α- and 3α-H) 6.79 m (2H). *Anal.* Calcd. for $C_{27}H_{46}O_2$: C, 80.54; H, 11.52. Found: C, 80.39; H, 11.34. The fractions eluted with benzene–ether (9:1–1:1) gave 40 mg of a mixture. The combined mixture (376 mg) was again chromatographed over 10 g of Al_2O_3. The fractions eluted with petr. ether–benzene (1:1) afforded 20 mg (1.7%) of XIa, mp 116—118°. The fractions eluted with the same solvent gave 42 mg of a mixture of X and XIa. The fractions eluted with petr. ether–benzene (1:1) yielded 228 mg of the solid, which upon recrystallization from aqueous acetone afforded 176 mg of X. The combined yield of X was 61.8%.

b) The purified triol monoacetate (IXa) (159 mg) was treated with MsCl–pyridine, followed by cyclization with KOH–MeOH as described in a) and 114 mg (82.5%) of X was obtained without formation of XIa.

Cleavage of 5α-Hydroxycholestan-2α,3α-oxide (II) with Acetic Acid——A solution of 345 mg of II in 10 ml of AcOH was heated on a steam–bath for 2.5 hr. The product was extracted with CH_2Cl_2 and submitted to preparative TLC using cyclohexane–AcOEt (1:1) as developing solvent. This provided 17 mg (4.9%) of unchanged starting material (II), 293 mg (73.9%) of IXa, mp 179—180° (from MeOH). $[\alpha]_D^{22}$ +21.1±0.5° (c=1.060). IR $\nu_{OH}^{CCl_4}$ cm^{-1}: 3616, 3519 (1:4.7). ν_{max} cm^{-1}: 3343, 3223, 1735, 1251, 1058. NMR τ: (18-H) 9.33, (19-H) 8.93, (OAc) 7.98, (3β-H) 6.16 (Wh/2≈8.0), (2α-H) 4.98 (Wh/2≈7.0 cps). *Anal.* Calcd. for $C_{29}H_{50}O_4$: C, 75.28; H, 10.89. Found: C, 75.23; H, 10.87. 43 mg (10.7%) of triol monoacetate, which is different from IXa, mp 190—192°. IR ν_{max} cm^{-1}: 3424, 3390, 1713, 1293, 1285, 1047. *Anal.* Calcd. for $C_{29}H_{50}O_4$: C, 75.28; H, 10.89. Found: C, 75.51; H, 11.05, and 7 mg (1.7%) of triol (IXc), mp 257—260° (decomp.).

Cleavage of 5α-Hydroxycholestan-2α,3α-oxide (II) with Perchloric Acid in Aqueous Dioxane——To a stirred solution of 1.00 g of II in 33 ml of 90% dioxane, 1.5 ml of 30% $HClO_4$ was added. The reaction mixture was agitated for 2.5 hr, during that period precipitates appeared. After dilution with water, the precipitates were collected by filtration, washed with water, and dried. Recrystallization from $CHCl_3$–MeOH afforded 658 mg (63.0%) of IXc, mp 257—260° (decomp.), IR ν_{max} cm^{-1}: 3220, 1056, 1016. *Anal.* Calcd. for $C_{27}H_{48}O_3$: C, 77.07; H, 11.50. Found: C, 77.08; H, 11.46. Evaporation of the mother liquor gave 395 mg of the residue, which was crystallized from ether to yield 154 mg of IXc. Recrystallization from $CHCl_3$–MeOH gave 34 mg of IXc. Combined yield of IXc was 65.0%. The ether-soluble fraction

was chromatographed over 7 g of Florisil. The fractions eluted with benzene and benzene–ether (4:1) afforded 212 mg (19.3%) of XIb, which was recrystallized from aqueous acetone, mp 151—152°. $[\alpha]_D^{29}$ +24.2±0.5° (c=1.003). IR $\nu_{OH}^{CCl_4}$ cm^{-1}: 3589. ν_{max} cm^{-1}: 3403, 963, 811. NMR τ: (18-H) 9.33, (19-H) 9.09, (4β-H) 7.55 d-d (J≈7.0, 14.0), (3β-H) 6.19d (J≈7.0), (2β-H) 5.82d (J≈6.0 cps). *Anal.* Calcd. for $C_{27}H_{46}O_2$: C, 80.54; H, 11.52. Found: C, 80.48; H, 11.55. Acetylation of this compound with pyridine–Ac_2O gave the acetate (XIc) which was recrystallized from M.OH. mp 83.5—84.5°. $[\alpha]_D^{21}$ +13.3±0.4° (c=1.036). IR ν_{max} cm^{-1}: 1738, 1242, 1035, 975. NMR τ: (18-H) 9.33, (19-H) 9.09, (AcO) 7.97, (4β-H) 7.54q (J≈−14.0, 7.0), (2β-H) 5.66d (J≈6.2), (3β-H) 5.28 d-d (J≈7.0, 3.0 cps). *Anal.* Calcd. for $C_{29}H_{48}O_3$: C, 78.32; H, 10.88. Found: C, 78.35; H, 10.72.

Acetylation of Cholestane-2β,3α,5α-triol (IXc)——To a solution of 130 mg of triol (IXc) in 3.6 ml of pyridine, 0.4 ml of Ac_2O was added. The reaction mixture was allowed to stand at room temperature for 2 days. After the usual work-up, the product (155 mg) was submitted to preparative TLC using cyclohexane–AcOEt (2:1) as developing solvent. The more mobile fraction was recrystallized from MeOH yielding 100 mg (64.1%) of the diacetate (IXd), mp 122—123°. $[\alpha]_D^{21}$ +32.1±0.6° (c=1.088). IR ν_{max} cm^{-1}: 3496, 1727, 1245. NMR τ: (18-H) 9.35, (19-H) 8.93, (AcO) 7.96, 7.93, (OH) 7.37, (2α- and 3β-H) 4.93 (Wh/2≈8.0 cps). *Anal.* Calcd. for $C_{31}H_{52}O_5$: C, 73.76; H, 10.38. Found: C, 73.95; H, 10.32. The less mobile fraction was recrystallized from MeOH to give 33 mg (23.1%) of IXa, mp 179—180°, identical with the product obtained by treatment of the oxide (II) with AcOH.

3α-Mesyloxycholestan-2α,5α-epoxide (XIa)——a) To a solution of 490 mg of triol (IXc) in 30 ml of pyridine, 0.5 ml of MsCl was added. The reaction mixture was kept standing at room temperature overnight. After the usual work-up, the product was recrystallized from hexane affording 532 mg (95.0%) of XIa identical with the by-product formed in the conversion of II to X, mp 116—118°. $[\alpha]_D^{19}$ +13.2±0.4° (c=1.065). IR ν_{max} cm^{-1}: 1343, 1336, 1179, 999, 990, 935. NMR τ: (18-H) 9.34, (19-H) 9.09 (Ms) 6.99, (4β-H) 7.47q (J≈−14.0, 7.0), (2β-H) 5.47d (J≈6.5), (3β-H) 5.23 d-d (J≈7.0, 3.0 cps). *Anal.* Calcd. for $C_{28}H_{48}O_4S$: C, 69.95; H, 10.06; S, 6.70. Found: C, 70.05; H, 10.02; S, 6.78. Reduction of XIa (230 mg) was carried out with 50 mg of $LiAlH_4$ in 10 ml of dry ether under reflux. After the usual work-up, recrystallization from aqueous acetone afforded 135 mg (69.3%) of XIb, identical with the by-product in the reaction of the α-oxide (II) with perchloric acid in aqueous dioxane.

b) Treatment of XIb with pyridine–MsCl gave the mesylate (XIa) in quantitative yield.

2β,5α-Dihydroxycholestane-3α-thiocyanate (XIIa)——To a solution of 4.95 g of the epoxide (X) in a mixture of 7 ml of CH_2Cl_2 and 5 ml of ether, HSCN-solution prepared from 9 g of KSCN, 7 g of H_3PO_4, and 15 ml of ether was added. After agitation for 3 hr, the crystals deposited were filtered off, and washed with ether. The filtrate was washed with 10% Na_2CO_3 and water, dried, and evaporated to dryness. The residue was treated with ether giving additional crystals. The combined crystals were recrystallized from acetone to afford 3.217 g (56.7%) of XIIa, mp 214—215.5°. $[\alpha]_D^{20}$ +3.3±0.3° (c=1.040). IR ν_{max} cm^{-1}: 3404, 2176, 1297, 1289, 1049, 1010. NMR τ: (18-H) 9.34, (19-H) 8.84. *Anal.* Calcd. for $C_{28}H_{47}O_2NS$: C, 72.83; H, 10.26; N, 3.06; S, 6.94. Found: C, 73.07; H, 10.00; N, 2.98; S, 7.01. The mother liquor of ether exhibits many spots on TLC and was not further studied. The thiocyanate (XIIa) (3.45 g) was acetylated with 20 ml of pyridine and 8 ml of Ac_2O in the usual way. After the work-up recrystallization from hexane gave 3.204 g (85.5%) of XIIb, mp 157—158°. $[\alpha]_D^{20}$ −4.6±0.3° (c=0.963). IR ν_{max} cm^{-1}: 3438, 2176, 1745, 1234, 1016. NMR τ: (18-H) 9.35, (19-H) 8.93, (AcO) 7.94, (2α-H) 4.68, (3β-H) 6.34.

5α-Hydroxycholestan-2α,3α-episulfide (XIII)——To a solution of 300 mg of XIIb in 4.5 ml of dioxane, 8 ml of 0.94M KOH–MeOH was added. The mixture was stirred at room temperature for 50 min. After dilution with ice water, the precipitate was collected by filtration, washed with water, and dried. Crystallization from hexane gave the solid which was recrystallized from CH_2Cl_2–MeOH affording 55 mg (19.3%) of XIV, mp 242—244°. $[\alpha]_D^{22}$ −88.0±1.2° (c=1.016). IR ν_{max} cm^{-1}: 3464, 3195, 1737, 1252. CD (in dioxane) $[\theta]_{250m\mu}^{inf}$ −4590, $[\theta]_{235m\mu}^{inf}$ −6160, $[\theta]_{220m\mu}^{max}$ −29870, Mol. wt.; Calcd. 955.55. Found: 956. *Anal.* Calcd. for $C_{58}H_{98}O_6S_2$: C, 72.90; H, 10.34; S, 6.71. Found: C, 73.07; H, 10.39; S, 7.05. The mother liquor of hexane was evaporated and the residue was recrystallized from aqueous acetone yielding 110 mg (45.2%) of XIII, mp 116.5—118°. $[\alpha]_D^{22}$ +19.7±0.5° (c=0.930). IR ν_{max} cm^{-1}: 3408, 3384, 1054, 1035, 980, 870. CD(in isooctane) $[\theta]_{261m\mu}^{max}$ −2480. NMR τ: (18-H) 9.37, (19-H) 9.04, (2β- and 3β-H) *ca.* 6.71 m (2H).

Dehydration of 2β,5α-Dihydroxycholestane-3α-thiocyanate 2-Acetate (XIIb)——To a stirred solution of 1.010 g of (XIIb) in 10 ml of pyridine, 0.35 ml of $SOCl_2$ was added dropwise with cooling to 0° for 5 min. After on standing for 5 min, the reaction mixture was worked up as described in the preparation of V and VI. The product showing 3 spots on TLC was crystallized from petr. ether to afford 276 mg of XV, which was recrystallized from ether–petr. ether, mp 145.5—146.5°. $[\alpha]_D^{22}$ +5.9±0.5° (c=1.002). IR ν_{max} cm^{-1}: 2156, 1746, 1237, 1022. CD (in dioxane) $[\theta]_{247m\mu}^{max}$ +12210. NMR τ: (18-H) 9.33, (19-H) 8.89, (OAc) 7.92, (3β-H) 6.22, (2α-H) 4.85, (6-H) 4.50. *Anal.* Calcd. for $C_{30}H_{47}O_2NS$: C, 74.18; H, 9.75; N, 2.88; S, 6.60. Found: C, 74.44; H, 9.97; N, 2.69; S, 6.67. The mother liquor was submitted to preparative TLC using cyclohexane–AcOEt (9:1) as developing solvent. The more mobile fraction (25 mg) exhibits 2 spots corresponding to XVI and XVII on TLC. The less mobile fraction (347 mg) was obtained as an oil and again shows the same 2 spots as above on TLC. The fraction exhibits absorption bands at $\nu_{max}^{CS_2}$ 2810, 1743, 1654,

1231, 1021 cm^{-1} in the IR spectrum. The least mobile fraction (58 mg) was identical with XV. The combined yield was 334 mg (34.4%).

Cholest-5-en-2α,3α-episulfide (XVIII)——A mixture of 242 mg of XV, 2 ml of dioxane, and 4 ml of 1.56M KOH–MeOH was kept at room temperature with stirring for 2 hr and then poured into ice water. The precipitate was collected by filtration, washed with water, and dried. Recrystallization from petr. ether afforded 108 mg (54.1%) of XVIII, mp 113—115°. $[\alpha]_D^{22}$ -43.3 ± 0.8 ($c=1.117$). IR ν_{max} cm^{-1}: 1669, 1004, 961, 834. UV λ_{max} mμ (ε): 263 (53). NMR τ: (18-H) 9.33, (19-H) 8.96, (4-H) 4.64, (2β- and 3β-H) *ca.* 6.70 m (2H). *Anal.* Calcd. for $C_{27}H_{44}S$: C, 80.93; H, 11.07; S, 8.00. Found: C, 81.07; H, 11.20; S, 8.23.

2β-Acetoxycholest-4-ene-3α-thiol Acetate (XIX)——A cooled solution of 430 mg of a mixture of XVI and XVII was reduced with 103 mg of $LiAlH_4$ in 4 ml of dry ether for 1.5 hr. The usual work-up gave the product (386 mg), which was acetylated with 1 ml of pyridine and 0.5 ml of Ac_2O. After the work-up, the product (460 mg) was submitted to preparative TLC using cyclohexane–AcOEt (23:2) as developing solvent. The most mobile fraction was recrystallized from ether–MeOH affording 223 mg (50.3%) of XIX, mp 90—91.5°. $[\alpha]_D^{22}$ $+158.3\pm1.9°$ ($c=1.040$). IR ν_{max} cm^{-1}: 1742, 1694, 1663, 1247. NMR τ: (18-H) 9.32, (19-H) 8.91, (OAc) 7.97, (S-Ac) 7.70, (3β-H) 5.88 (Wh/2$\approx$10.0), (2α-H) 4.97 (Wh/2$\approx$12.0), (4-H) 4.77d ($J\approx4.5$ cps). *Anal.* Calcd. for $C_{31}H_{50}O_3S$: C, 74.05; H, 10.02; S, 6.38. Found: C, 73.96; H, 9.92; S, 6.34.

Treatment of 2β-Acetoxycholest-4-ene-3α-thiolacetate (XIX) with Alkali——To a solution of 55 mg of XIX in 0.5 ml of ether, 1 ml of 1.17M KOH–MeOH was added. The reaction mixture was stirred for 1 hr and poured into ice water. The deposited oily substance was extracted with ether–petr. ether (1:1). The organic layer was washed with water, dried, and concentrated at 25° under reduced pressure. The residue (44.6 mg) exhibiting many spots on TLC, was separated by preparative TLC using cyclohexane–AcOEt (9:1) as developing solvent. The main product (12 mg) was isolated as an oily substance. IR $\nu_{max}^{CS_2}$ cm^{-1}: 1739, 1659, 1231, 1003, which was not further studied.

Bromination of Cholestenone (XX)——A mixture of 1.6 g of NaH (52.9%), 31 ml of MeOH and 140 ml of benzene was distilled until 90 ml of the distillate was collected. After cooling to room temperature, 6 ml of diethyl oxalate was added to the mixture and stirred for 10 min. To the orange-colored solution, a solution of 10.0 g of XX in 60 ml of benzene was added dropwise with stirring for 15 min. The dark colored mixture was stirred at room temperature for 39 hr. The precipitate formed was collected by filtration, washed with benzene, and dried. A solution of the pricipitate in water was acidified with HCl and extracted with ether. The ethereal layer was washed with water, dried and evaporated to dryness under reduced pressure. The residue (12.6 g) was dissolved in 250 ml of MeOH. To the cooled solution, 9 g of anhydrous AcOK was added, and then a solution of 4.6 g of Br_2 in 57 ml of CCl_4 was added dropwise with stirring over 15 min. After agitation for 5 min, the yellow colored solution was concentrated at 20—40° under reduced pressure. After dilution with ice water, the mixture was extracted with ether. The ethereal layer was washed with 10% Na_2CO_3 and water, dried, and evaporated to dryness. The product (12.7 g) was chromatographed over 300 g of Florisil. The eluates with petr. ether–benzene (1:1–1:2) gave 420 mg of an oily substance. The fractions eluted with petr. ether–benzene (1:4) afforded 811 mg (5.8%) of the dibromide (XXIII), which was recrystallized from ether–MeOH, mp 120—121.5°. $[\alpha]_D^{24}$ $-12.7\pm0.6°$ ($c=0.883$). IR ν_{max} cm^{-1}: 1683, 1628, 1238, 870, 858, 845, 740. UV λ_{max}^{EtOH} mμ (ε) 259 (11700). NMR τ: (18-H) 9.28, (19-H) 8.53, (1-CH_2) 6.99d, 6.81d ($J_{AB}\approx-16.0$), (4-H) 4.18. *Anal.* Calcd. for $C_{27}H_{42}OBr_2$: C, 59.78; H, 7.80; Br, 29.46. Found: C, 59.90; H, 7.84; Br, 29.34. (reported[12]) mp 189—191°. $[\alpha]_D$ $+86°$. UV λ_{max}^{EtOH} 266 mμ (log ε 4.0)). The eluates with the same solvent yielded 715 mg of a complex mixture. The fractions eluted with benzene and benzene–ether (20:1) afforded 5.195 g of the solid, which was recrystallized from ether–MeOH giving 3.83 g (31.8%) of XXII, mp 125—126°. $[\alpha]_D^{24}$ $+99.3\pm1.3°$ ($c=1.090$). IR ν_{max} cm^{-1}: 1683, 1620, 1238, 1168, 864. UV λ_{max}^{EtOH} mμ (ε): 245 (15000). NMR τ: (18-H) 9.29, (19-H) 8.73, (1α-H) 7.86t ($J\approx$13.1, $-$13.0), (1β-H) 7.38d-d ($J\approx-13.0$, 6.5), (2β-H) 5.19d-d ($J\approx$13.1, 6.5). (4-H) 4.19. *Anal.* Calcd. for $C_{27}H_{43}OBr$: C, 69.96; H, 9.35; Br, 17.24. Found: C, 70.04; H, 9.34; Br, 17.52. (reported[15]) mp 117—119°. $[\alpha]_D$ $+81°$. UV λ_{max}^{EtOH} 243 mμ (log ε 4.15)).

2-Bromocholesta-1,4-dien-3-one (XXIV)——A mixture of 260 mg of the dibromide (XXIII), 156 mg of Li_2CO_3 and 2 ml of DMF was refluxed for 50 min. After dilution with ether, precipitate was removed by filtration. The filtrate was washed with 10% Na_2CO_3 and water, dried and concentrated under reduced pressure. The product (260 mg) was submitted to preparative TLC using cyclohexane as developing solvent. The main fraction was recrystallized from acetone–MeOH yielding 205 mg (92.8%) of XXIV mp 132—132.5°. $[\alpha]_D^{28}$ $-22.7\pm0.6°$ ($c=1.060$). IR ν_{max} cm^{-1}: 1670, 1603, 973, 947, 870. UV λ_{max}^{EtOH} 256 mμ (log ε 4.18). NMR τ: (18-H) 9.26, (19-H) 8.73, (4-H) 3.84s (Wh/2$\approx$3.0). (1-H) .250s (Wh/2$\approx$1.5). *Anal.* Calcd. for $C_{27}H_{41}OBr$: C, 70.27; H, 8.95; Br, 17.32. Found: C, 70.04; H, 8.84; Br, 17.59 (reported mp 130°. $[\alpha]_D$ $-13°$. UV λ_{max}^{EtOH} 255 mμ (log ε 4.2)).

3-Oxocholest-4-en-2α-ethyl Xanthate (XXV)——A mixture of 4.20 g of the bromide (XXII), 2.03 g of KSC(=S)OEt, and 50 ml of acetone was stirred at room temperature for 40 hr. The mixture was poured into ice water and extracted with ether. The product insoluble in ether was filtered off and recrystallized from CH_2Cl_2–acetone yielding 403 mg (10.7%) of XXVI, mp 209.5—210.5°. $[\alpha]_D^{24}$ $-119.0\pm1.8°$ ($c=0.878$). IR ν_{max} cm^{-1}: 1681, 1636, 1239, 1219, 1172, 903, 870. UV $\lambda_{max}^{dioxane}$ 241 mμ (31100) NMR τ: (18-H) 9.28, (19-H)

8.70, (1β-H) 7.43d-d ($J\approx-13.2$, 5.0), (2β-H) 5.99d-d ($J\approx14.0$, 5.0 cps), (4-H) 4.26. Mol. wt., Calcd. 831.4. Found: 822. *Anal.* Calcd. for $C_{54}H_{86}O_2S_2$: C, 78.01; H, 10.43; S, 7.71. Found: C, 78.26; H, 10.37; S, 7.89. The filtrate was washed with 10% Na_2CO_3 and water, dried, and evaporated. The residue was chromatographed over 90 g of Florisil. The eluates with petr. ether–benzene (7:3) gave a small amount of an oil. The fractions eluted with petr. ether–benzene (6:4, 1:1, 1:2 and 1:5) were recrystallized from acetone to afford 1.87 g (40.9%) of XXV, mp 115.5—116.5°. $[\alpha]_D^{24}$ $-198.1\pm2.5°$ ($c=0.952$). IR ν_{max} cm^{-1}: 1681, 1620, 1228, 1111, 1051, 899. UV $\lambda_{max}^{dioxane}$ mμ (ε): 282 (11400), 235.5 (18800). NMR τ: (18-H) 9.29, (19-H) 8.65, (CH_3CH_2O) 8.95t ($J\approx7.0$), (CH_3CH_2O) 5.36q ($J\approx7.0$), (1β-H) 7.50d-d ($J\approx-13.0$, 5.0), (2β-H) 5.25d-d ($J\approx14.5$, 5.0 cps). *Anal.* Calcd. for $C_{30}H_{48}O_2S_2$: C, 71.38; H, 9.58; S, 12.70. Found: C, 71.22; H, 9.56; S, 12.89.

3β-Acetoxycholest-4-en-2α-ethyl Xanthate (XXIXb)——The keto xanthate (XXV) (500 mg) was reduced with 37 mg of $NaBH_4$ in a mixture of 5 ml of tetrahydrofuran and 5 ml of MeOH for 1 hr. The usual work-up afforded 510 mg of the product as an oil, which was submitted to preparative TLC using cyclohexane as developing solvent. The fraction of the main product gave 379 mg of the solid, which was acetylated with 1.5 ml of pyridine and 0.5 ml of Ac_2O in the usual way. The isolated product was recrystallized from aqueous acetone to yield 345 mg (63.4%) of (XXIXb), mp 90.5—91°. $[\alpha]_D^{24}$ -157.1 ± 2.0 ($c=1.012$). IR ν_{max} cm^{-1}: 1745, 1665, 1248, 1239, 1206, 1050. UV $\lambda_{max}^{dioxane}$ mμ (ε) 282 (12400), 224 (7900). NMR τ: (18-H) 9.32, (19-H) 8.78, (OAc) 7.96, (CH_3CH_2O) 8.58t ($J\approx7.1$), (CH_3CH_2O) 5.35q ($J\approx7.1$), (2β-H) 6.00 oct ($J\approx13.1$, 10.5, 3.1), (4-H) 4.80, (3α-H) 4.70bd, ($J\approx10.5$), (in C_6D_6): (4-H) 4.69 (Wh/2$\approx$2.0), (3α-H) 4.37bd ($J\approx10.5$ cps). *Anal.* Calcd. for $C_{32}H_{52}O_3S_2$: C, 70.02; H, 9.55; S, 11.68. Found: C, 70.12; H, 9.59; S, 12.00.

Desulfurization of 3β-Acetoxycholest-4-en-2α-ethyl Xanthate (XXIXb)——A mixture of 73 mg of the xanthate (XXIXb), 660 mg of freshly prepared W-2 Raney Ni, and 3.6 ml of dioxane was warmed to 55° with stirring. After removal of Raney Ni by filtration, the filtrate was evaporated to dryness under reduced pressure. The residue (54 mg) was submitted to preparative TLC using benzene–cyclohexane (4:1) as developing solvent. The more mobile fraction gave 20 mg (42.5%) of a mixture of cholest-4-ene and -3-ene, IR spectrum of which indicated predominant 4-ene over 3-ene. The less mobile fraction was recrystallized from ether-MeOH affording 30 mg (52.8%) of XXVII,[16] which was identified with the authentic sample derived from XX by a mixed mp and comparison of the IR spectrum, mp 87—88°. $[\alpha]_D^{23}$ $+93\pm0.9°$ ($c=0.503$). IR ν_{max} cm^{-1}: 1746, 1658, 1241, 1019. *Anal.* Calcd. for $C_{29}H_{48}O_2$: C, 81.25; H, 11.29. Found: C, 81.45; H, 11.28.

Cholest-4-en-2α,3α-episulfide (XXX)——To a solution of 300 mg of XXIXb in 2.5 ml of tetrahydrofuran, a clear solution of 100 mg of KOH in 1.5 ml of absolute EtOH was added. The reaction mixture was stirred at room temperature for 20 min and poured into water. The deposited crystals were collected by filtration washed with water, and dried under reduced pressure and weighed 39 mg (17.8%), mp 72—74.5°. NMR τ: (18-H) 9.32, (19-H) 8.94, (2β- and 3β-H) *ca.* 6.65 (2H), (4-H) 4.30. UV λ_{max} mμ (ε) 279 (670), 220 (10180). *Anal.* Calcd. for $C_{27}H_{44}S\cdot\frac{1}{3}H_2O$: C, 79.74; H, 11.07; S, 7.88. Found: C, 79.85; H, 10.99; S, 8.05.

Acknowledgement The authors thank Dr. K. Takeda, Director of this laboratory, for his encouragement throughout this work. Thanks are also due to Dr. K. Kuriyama for his measurement of CD and ORD curves and his helpful discussion.

15) C. Djerassi, *J. Am. Chem. Soc.*, **71**, 1003 (1949).
16) H. McKennis Jr. and G.W. Gattney, *J. Biol. Chem.*, **175**, 217 (1948); Pl. A. Plattner, H. Heusser and A.B. Kulkarni, *Helv. Chim. Acta*, **32**, 265 (1949).