Unsaturated Steroids. Part 12.[1] Synthesis of 1α,3β-Dihydroxy-24-nor-9,10-secochola-5,7,10(19)trien-23-oic (Calcitroic) Acid and of the Cholic-and 25-Homocholic Acid Analogues

Brian R. de Costa, Nandor Makk, John M. Midgley, Najmuddin T. Modi, Robert A. Watt, and W. Basil Whalley*

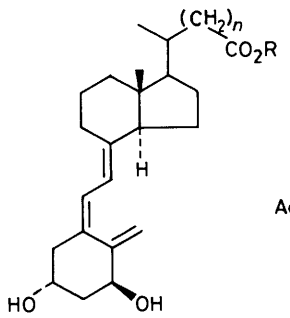
The School of Pharmacy, University of London, 29/39 Brunswick Square, London. WC1N 1AX

The three title compounds have been synthesized by the same general method. Thus, methyl 3β-hydroxy-24-nor-5α-chol-7-en-23-oate (**3**; R = CO_2Me), derived from 3β-acetoxypregn-7-ene-22-carbaldehyde (**2**), gave methyl 3-oxo-24-nor-5α-chol-7-en-23-oate (**4**; R = H,R′ = CH_3). Bromination of this to the 2ξ,4ξ-dibromo derivative (**4**; R = Br, R′ = Me) followed by dehydrobromination formed the corresponding 1,4,7-triene-3-one (**5**). The corresponding enol acetate (**6**) was reduced to the 1,5,7-triene (**7**) which was converted into the adduct (**8**; R = H) with 4-phenyl-1,2,4-triazoline-3,5-dione; the 3β-dimethyl-t-butylsilyl ether of (**8**; R = H) gave the 1α,2α-epoxide (**9**; R = Me_2Bu^tSi). Removal of the silyl ether group with acid, and then of the triazoline residue with pyridine–1,5-diazabicyclo[4.3.0]non-5-ene gave methyl 3β-hydroxy-1α,2α-epoxy-24-norchola-5,7-dien-23-oate (**10**; R = Me). Reduction of the corresponding acid (**10**; R = H) gave 1α,3β-dihydroxy-24-norchola-5,7-dien-23-oic acid (**11**; R = H). Photolysis/thermolysis of the corresponding methyl ester (**11**; R = Me) followed by saponification gave 1α,3β-dihydroxy-24-nor-9,10-secochola-5,7,10(19)trien-23-oic acid (calcitroic acid) (**1**; R = H, *n* = 1). The analogous cholic (**1**; R = H, *n* = 2) and 25-homocholic acid (**1**; R = H, *n* = 3) derivatives were similarly synthesized.

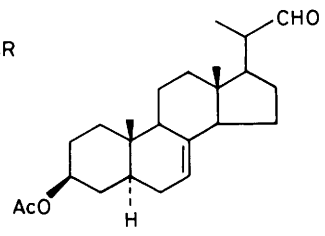
Intensive investigation[2] of the metabolism of 1α,25-dihydroxyvitamin D_3 has resulted in the identification[3] of 1α,3β-dihydroxy-9,10-seco-24-norchola-5,7,10,(19)-trien-23-oic acid (1; R = H, *n* = 1) (calcitroic acid) as a major degradation product. The synthesis of calcitroic acid[4] and of the 24-cholic acid[5] (1; R = H, *n* = 2) analogue have been described. However, the quantities of products available from both natural and synthetic sources have been minute; additionally the characterisation of the material (both natural and synthetic) has been minimal.

We now report rigorous syntheses of (**1**; R = H, *n* = 1), (**1**; R = H, *n* = 2), and (**1**; R = H, *n* = 3), with complete characterisation of the products, and their availability in mg quantities, starting from ergosterol. The three syntheses are similar, hence only that of calcitroic acid is described in detail; this synthesis is similar to, but has many significant differences from, our original approach[6] to this system.

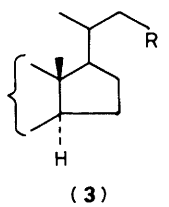
Thus, condensation of 3β-acetoxypregn-7-ene-22-carbaldehyde (**2**), (from 5,6-dihydroergosterol), with the Wittig reagent from methoxymethylphenylphosphonium chloride, gave 3β-hydroxy-24-nor-5α-chol-7-en-23-al (**3**; R = CHO). It has been established[7] that the natural configuration at C-20 is retained in this reaction. Oxidation of (**3**; R = CHO) with silver oxide gave



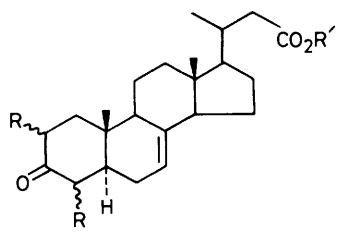
(1)



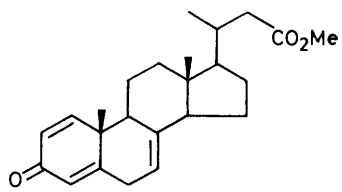
(2)



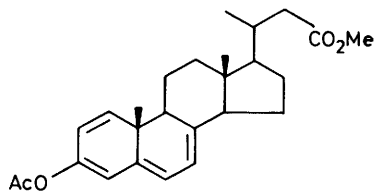
(3)



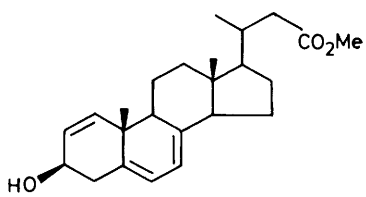
(4)



(5)



(6)

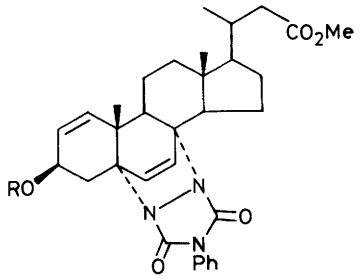


(7)

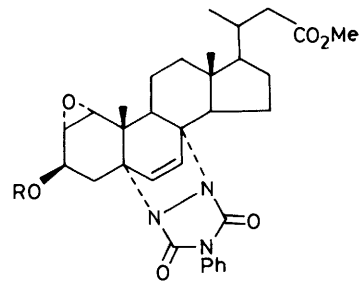
the acid (**3**; R = CO_2H). The methyl ester (**3**; R = CO_2Me) was oxidised with Jones' reagent to yield methyl 3-oxo-24-nor-5α-chol-7-en-23-oate (**4**; R = H, R′ = CH_3). This ketone and its analogues readily formed the 3,3-dimethyl acetal when recrystallised from methanol. This is in accord with the strain imposed[8] upon ring A by the C–7 double bond and the diminution of this strain by the conversion of C–3 into an sp^3 hybridised carbon atom. The sequence of bromination (using tri-*N*-methylanilinium perbromide) and dehydrobromination, with lithium carbonate–dimethylformamide–lithium bromide, gave methyl 3-oxo-chola-1,4,7-trien-23-oate (**5**), which was advantageously purified by 'flash' chromatography. With isopropenyl acetate–toluene-*p*-sulphonic acid, (**5**) gave methyl 3-acetoxy-24-norchola-1,3,5,7-tetraen-23-oate (**6**), which because of its incipient instability was reduced immediately with calcium borohydride to methyl 3β-hydroxy-24-norchola-1,5,7-trien-23-oate (**7**). To obtain high yields in this step particular attention must be paid to the purity and dryness of the calcium chloride used for the preparation of the calcium borohydride.

After conversion of (**7**) into the adduct (**8**; R = H) with 4-phenyl-1,2,4-triazoline-3,5-dione, the dimethyl-t-butylsilyl ether (**8**; R = Me_2Bu^tSi) was oxidised with *m*-chlorperbenzoic acid to give, exclusively, the 1α,2α-epoxide (**9**; R = Me_2Bu^tSi). As with analogous compounds, the adduct (**9**; R = H) retained solvent and did not furnish satisfactory elemental analyses. The n.m.r. and mass spectra (FAB), however, were satisfactory and the silyl ether (**8**; R = Me_2Bu^tSi) and its epoxide (**9**; R = Me_2Bu^tSi) furnished appropriate elemental analyses. Removal of the silyl ether group from (**9**; R = Me_2Bu^tSi) occurred satisfactorily with acetic acid–tetrahydrofuran at 60 °C, to yield the adduct (**9**; R = H), from which the triazoline residue was removed[9] by boiling pyridine–1,5-diazabicyclo[4.3.0]non-5-ene to yield (**10**; R = Me). Hydrolysis, at room temperature of (**10**; R = Me) gave the acid (**10**; R = H) which was reduced with lithium borohydride in boiling tetrahydrofuran to 1α,3β-dihydroxy-24-norchola-5,7-dien-23-oic acid (**11**; R = H). The ester (**11**; R = Me) was formed using diazomethane. Photolysis of this ester, or more satisfactorily of the 1α,3β-di-*O*-acetate, followed by thermal equilibration, saponification and final purification of the product by p.l.c. on silica–silver nitrate gave 1α,3β-dihydroxy-24-nor-9,10-seco-5,7,10(19)-cholatrien-23-oic acid (calcitroic acid) complete for the first time with melting point, elemental analysis, and n.m.r. spectrum. The cholic and 25-homocholic acid analogues were similarly prepared (and characterised) from the analogous acids; there was advantageous modification, in certain cases, of the experimental protocol. These compounds are presently being assayed for biological activity. During the development of this route to the 1α-functionalised system, we investigated the protection of the terminal carboxy residue as the trichloroethyl ester, since this group may be more easily removed reductively as opposed to hydrolytically.

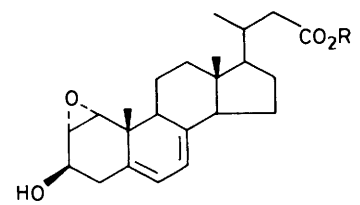
Thus methyl 3-oxo-5α-chol-7-en-24-oate (**4**; R = H, R′ = Me) was hydrolysed to the acid (**4**; R = R′ = H) which was esterified with 2,2,2-trichloroethanol to yield 2′,2′,2′-trichloroethyl-3-oxo-5α-chol-7-en-24-oate (**4**; R = H, R′ = CH_2CCl_3). This ester was converted into 2,2,2-trichloroethyl 3-acetoxy-chola-1,3,5,7-tetraen-24-oate, which unfortunately gave a complex mixture on reduction with calcium borohydride. Even this mild reducing reagent apparently removed the trichloroethyl residue (at least in part). Hence this approach was abandoned.



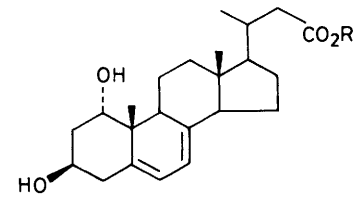
(**8**)



(**9**)



(**10**)



(**11**)

Experimental

Optical rotations were performed in chloroform, unless stated otherwise, and at 20 °C N.m.r. spectra were determined on a 60 MHz R12A Perkin-Elmer or an 80 MHz WP 80 SY Bruker in $CDCl_3$ unless stated otherwise. U.v. spectra were determined in 96% ethanol. Mass spectra and accurate mass measurement were recorded on a ZAB-1F (VG Analytical Ltd.,) mass spectrometer: FAB mass spectra were recorded from solutions in chloroform in a substrate of glycerol. Light petroleum refers to the fraction of b.p. 60—80 °C. Ether refers to diethyl ether.

Methyl 3β-*Hydroxy*-24-*nor*-5α-*chol*-7-*en*-23-*oate*.—A solution of 3β-acetoxy-23,24-dinor-5α-chol-7-en-22-al (10.4 g) in tetrahydrofuran (80 ml) was added with stirring during 15 min, at 20 °C, to a solution of methoxymethyltriphenylphosphorane [prepared from methoxymethyltriphenylphosphonium chloride (4.6 g) in tetrahydrofuran (400 ml), and n-butyl-lithium (1.65M; 72 ml) in the same solvent] in tetrahydrofuran (400 ml). After 1 h the reaction mixture was diluted with water (200 ml), and most of the organic solvent was removed under reduced pressure at 40 °C. The oily product was isolated with ether (400 ml), and the solvent removed. A solution of the residue in dioxane (300 ml) containing sulphuric acid (1M; 66 ml) was stirred at 70 °C during 30 min, when the cooled mixture was neutralised by the addition of aqueous sodium hydrogen carbonate and the dioxane removed under reduced pressure. A

solution of the residue in ethyl acetate (200 ml) was washed, dried, evaporated, and the impure residue chromatographed on silica from ether–light petroleum (1:5) to give 3β-*hydroxy*-24-*nor*-5α-*chol*-7-*en*-23-*al* (4.8 g) as prisms, m.p. 117 °C, from light petroleum–methylene chloride; $[\alpha]_D$ −24° (*c* 3.44); τ 0.27 (1 H, d, *J* 3 Hz, CHO) (Found: C, 79.6; H, 10.2%; M^+, 344.2715. $C_{23}H_{36}O_2$ requires C, 80.2; H, 10.5%; *M*, 344.2715). A suspension of silver oxide [prepared by the addition of aqueous sodium hydroxide (10% w/v; 80 ml) to silver nitrate (8.9 g) dissolved in water (150 ml)] was dissolved by the addition of aqueous ammonia (*d*, 0.88; 100 ml) and stirred and maintained at 70 °C, during the addition (15 min) of a solution of this aldehyde (3 g) in methanol (300 ml). After removal of the methanol under reduced pressure the reaction mixture was extracted with ether (2 × 200 ml) and the aqueous residue acidified with hydrochloric acid (10% w/v; 75 ml). The precipitated 3β-*hydroxy*-24-*nor*-5α-*chol*-7-*en*-23-*oic acid* (2.2 g), extracted with ethyl acetate, crystallised from acetone as needles, m.p. 212—213 °C; $[\alpha]_D$ +12° (*c* 1.8, in methanol) (Found: C, 76.9; H, 10.3%; M^+, 360.2666. $C_{23}H_{36}O_3$ requires C. 76.6; H, 10.1% *M*, 360.2664). Prepared quantitatively from this acid (5.9 g) with ethereal diazomethane *methyl* 3β-*hydroxy*-24-*nor*-5α-*chol*-7-*en*-23-*oate* formed prisms, m.p. 173—175 °C, from methanol; $[\alpha]_D$ −10.3° (*c* 4.63); τ 6.33 (3 H, s, CO_2CH_3) (Found: C, 77.0; H, 10.1%; M^+, 374.2820. $C_{24}H_{38}O_3$ requires C, 77.0; H, 10.2%; *M*, 374.2821).

Methyl 3β-*Hydroxy*-5α-*chol*-7-*en*-24-*oate*.—A solution of 3β-acetoxy-23,24-dinor-5α-chol-7-en-22-al (8.11 g) and carboxymethylenetriphenylphosphorane (8.37 g) in tetrahydrofuran (450 ml) was refluxed for 6 h. Next day the solvent was removed under reduced pressure and the product purified by chromatography from ethyl acetate–light petroleum (1:10) on silica to yield *methyl* 3β-*acetoxy*-5α-*chol*-7,22E-*dien*-24-*oate* (6.1 g) as needles, m.p. 131 °C, from ether–methanol; $[\alpha]_D$ + 44° (*c* 3.58): τ 3.11 (H, dd, $J_{22.23}$ 15 Hz, $J_{22.20}$ 9 Hz, 22-H), 4.24 (H, d, $J_{22.23}$ 15 Hz, 23-H), and 4.27 (3 H, s, CO_2CH_3) (Found: C, 76.0; H, 9.2%; M^+, 428.2926. $C_{27}H_{40}O_4$ requires C, 75.7; H, 9.4%; *M*, 428.2929).

Hydrogenation of a solution of this diene (5 g) in ethyl acetate (300 ml), containing platinic oxide (0.3 g) occurred during 2.5 h to yield *methyl* 3β-*acetoxy*-5α-*chol*-7-*en*-24-*oate* (5 g) as needles, m.p. 121 °C, from ether–methanol; $[\alpha]_D$ − 13.5° (*c* 3.1) (Found: C, 75.2; H, 9.8%; M^+, 430.3084. $C_{27}H_{42}O_4$ requires, C, 75.3; H, 9.8%; *M*, 430.3083). Hydrolysis of a solution of this acetate (2 g) in methanol (200 ml), by the addition of potassium hydroxide (2 g) dissolved in methanol (20 ml), during 5 min, at 40 °C gave *methyl* 3β-*hydroxy*-5α-*chol*-7-*en*-24-*oate* (1.7 g) as needles, m.p. 110—112 °C, from aqueous methanol; $[\alpha]_D$ −5.9° (*c* 3.73); τ 6.33 (3 H, s, CO_2CH_3) (Found: C, 76.2; H, 10.00%; M^+, 388.2978. $C_{25}H_{40}O_3 \cdot 0.5H_2O$ requires C, 76.0; H, 10.3%; $C_{25}H_{40}O_3$ requires *M*, 388.2977).

Methyl 3β-*Hydroxy*-25-*homo*-5α-*chol*-7-*en*-25-*oate*.—A stirred solution of 3β-acetoxy-23,24-dinor-5α-chol-7-en-22-al (9.27 g) and of (2-carboxyethyl)triphenylphosphonium bromide (20.76 g) in a mixture of dimethylformamide (100 ml) and toluene (100 ml) was treated (N_2) at 0 °C with sodium hydride (12 g). After 2 h at 0 °C, and a further 2.5 h at 20 °C, the excess of sodium hydride was destroyed (water added cautiously), and sulphuric acid (7% w/v; 340 ml) added. The product was isolated by extraction with isopropylalcohol–ether (1:9, 2 × 300 ml) and the extract esterified by the addition of an excess of ethereal diazomethane. The crude product was acetylated (pyridine–acetic anhydride) during 12 h to yield, after purification by chromatography on silica from ether–light petroleum (1:6) followed by crystallisation from ether–methanol, *methyl* 3β-*acetoxy*-25-*homo*-5α-*chol*-7,22-*dien*-25-*oate* (7 g) as plates, m.p. 115 °C; $[\alpha]_D$ −9.31° (*c* 5.69); τ 7.98 (3 H, s, $OCOCH_3$), 6.33 (3 H, s, CO_2CH_3), 4.58 (2 H, m, 22-, 23-H) (Found: C, 76.4; H, 9.8%; M^+, 442.3082. $C_{28}H_{42}O_4$ requires C, 76.0; 9.6%; *M*, 442.3083). Hydrogenation of this diene (5 g) in ethyl acetate (200 ml) containing platinic oxide (0.5 g) during 2.5 h gave *methyl* 3β-*acetoxy*-25-*homo*-5α-*chol*-7-*en*-25-*oate* (5 g) as prisms, m.p. 126 °C, from ether–methanol; $[\alpha]_D$ −9.9° (*c* 3.28); τ 8.00 (3 H, s, $OCOCH_3$) and 6.32 (3 H, s, CO_2CH_3) (Found: C, 75.5; H, 9.8%; M^+, 444.3239. $C_{28}H_{44}O_4$ requires C, 75.6; H, 10.0%; *M*, 444.3240).

Hydrolysis of this acetate (4.0 g) with potassium hydroxide (4.0 g) in methanol (400 ml) during 5 min at 40 °C, gave *methyl* 3β-*hydroxy*-25-*homo*-5α-*chol*-7-*en*-25-*oate* (3.55 g) as micro-needles, m.p. 122—124 °C, from ether–light petroleum; $[\alpha]_D$ +0.33° (*c* 6.15); τ 6.31 (3 H, s, CO_2CH_3) (Found: C, 77.3; H, 10.60; M^+, 402.3135. $C_{26}H_{42}O_3$ requires C, 77.6; H, 10.5%; *M*, 402.3134).

Methyl 3-*Oxo*-24-*norchola*-1,4,7-*trien*-23-*oate* (**5**).—Jones's reagent (2M) was added slowly with stirring to a solution of methyl 3β-hydroxy-24-nor-5α-chol-7-en-23-oate (6.13 g) until reaction (t.l.c.) was complete; excess of reagent was then destroyed by the addition of methanol (5 ml), followed by water (100 ml). Most of the solvent was removed under reduced pressure and the product extracted with ethyl acetate (2 × 200 ml) to yield *methyl* 3-*oxo*-24-*nor*-5α-*chol*-7-*en*-23-*oate* (5.6 g) as needles, m.p. 167—168 °C, from ether–light petroleum; $[\alpha]_D$ +20.7°; $\nu_{max.}$ 1 702 cm^{-1} (CO) (Found: C, 77.0; H, 9.8%; M^+, 372.2665. $C_{24}H_{36}O_3$ requires C, 77.4; H, 9.7%; *M*, 372.2664).

Methyl 3-*oxo*-5α-*chol*-7-*en*-24-*oate* formed plates, m.p. 121 °C, from ether–light petroleum; $[\alpha]_D$ +28.8°; $\nu_{max.}$ 1 705 cm^{-1} (CO) (Found: C, 77.8; H, 9.9%; M^+, 386.2817. $C_{25}H_{38}O_3$ requires C, 77.7; H, 9.9%; *M*, 386.2820). Crystallisation of this ester from methanol gave (quantitavely) *methyl* 3,3-*dimethoxy*-5α-*chol*-7-*en*-24-*oate* as needles, m.p. 128 °C; τ 6.80 (3 H, s, OCH_3), 6.88 (3 H, s, OCH_3), 6.34 (3 H, s, CO_2CH_3) (Found: C, 74.9; 10.3%; M^+, 432.3241. $C_{27}H_{44}O_4$ requires C, 75.0; H, 10.3%; *M*, 432.3239).

Methyl 3-*oxo*-25-*homo*-5α-*chol*-7-*en*-25-*oate* gave needles, m.p. 131 °C, from ether–light petroleum; $[\alpha]_D$ +23.1°; $\nu_{max.}$ 1 717 cm^{-1} (CO) (Found: C, 77.6; H, 10.1%; M^+, 400.2977. $C_{26}H_{40}O_3$ requires C, 78.0; H, 10.1%; *M*, 400.2977).

Phenyltrimethylammonium perbromide (4.13 g) was added during 0.5 h, to a stirred solution of methyl 3-oxo-24-nor-5α-chol-7-en-23-oate (4.1 g) in tetrahydrofuran (300 ml) at 0 °C, followed by more perbromide (4.1 g) during a further 40 min. After 4 h at room temperature the reaction mixture was diluted with water (500 ml) and the product extracted with ethyl acetate (2 × 200 ml). The resultant, unstable *methyl* 2ξ,4ξ-*dibromo*-3-*oxo*-24-*nor*-5α-*chol*-7-*en*-23-*oate* (5.9 g) formed micro-needles, m.p. 144—145 °C (decomp.), from ether–light petroleum; $[\alpha]_D$ −17.2° (*c* 2.18) (Found: C, 54.7; H, 6.4%; M^+, 530.0854. $C_{24}H_{34}{}^{79}Br^{81}BrO_3$ requires C, 54.4; H, 6.5%; *M*, 530.0856). The corresponding 2,4-*dibromocholate* formed needles, m.p. 148—149 °C, from ether–light petroleum; $[\alpha]_D{}^{19}$ −14.9° (*c* 2.17) (Found: C, 55.3; H, 6.5%; M^+, 544.1010. $C_{25}H_{36}{}^{79}Br^{81}BrO_3$ requires C, 55.2; H, 6.7%; *M*, 544.1010), whilst the 25-*homo*-*derivative* formed micro-needles, m.p. 144—145 °C (decomp.), from ether–light petroleum; $[\alpha]_D$ −14.1° (*c* 1.59) (Found: C, 55.7; H, 6.9%; M^+, 558.1165. $C_{26}H_{38}{}^{79}Br^{81}BrO_3$ requires C, 55.9; H, 6.9%; *M*, 558.1167).

A solution of unpurified methyl 2ξ,4ξ-dibromo-3-oxo-24-nor-5α-chol-7-en-23-oate (5.5 g) in dimethylformamide (190 ml) containing anhydrous lithium bromide (5.5 g) and lithium carbonate (11.0 g) was stirred at 150 °C, in a stream of nitrogen, during 45 min. Purification of the product by flash column chromatography on silica from ethyl acetate–light petroleum (1:4) gave *methyl* 3-*oxo*-24-*norchola*-1,4,7-*trien*-23-*oate* (2.7 g)

as prisms, m.p. 126—128 °C, from ether–light petroleum; $[\alpha]_D$ +9.1° (*c* 1.12); $\lambda_{max.}$ 242.5 nm (ε 13 700); τ 2.93 (1 H, d, $J_{1,2}$ 10 Hz, 1-H), 3.75 (1 H, dd, $J_{1,2}$ 10 Hz, $J_{2,4}$ 2 Hz, 2-H), 3.88 (1 H, m, 4-H), 4.74 (1 H, s br, 7-H) (Found: C, 77.8; H, 8.9%; M^+, 368.2351. $C_{24}H_{32}O_3$ requires C, 78.2; H, 8.8%; *M*, 368.2351).

Methyl 3-*oxochola*-1,4,7-*trien*-24-*oate* formed prisms, m.p. 134—136 °C; $[\alpha]_D$ +90.5° (*c* 4.75); $\lambda_{max.}$ 241 nm (ε 14 300) (Found: C, 78.6; H, 9.0%; M^+, 382.2508. $C_{25}H_{34}O_3$ requires C, 78.5; H, 9.0%; *M*, 382.2507).

The corresponding 25-*homo*-1,4,7-*triene* formed needles, m.p. 133—134 °C, from ether–light petroleum; $[\alpha]_D$ −1.90° (*c* 4.22); $\lambda_{max.}$ 242 nm (ε 14 100) (Found: C, 78.4; H, 9.2%; M^+, 396.2664. $C_{26}H_{36}O_3$ requires C, 78.7; H, 9.2%; *M*, 396.2664).

Methyl 3β-*Hydroxy*-24-*norchola*-1,5,7-*trien*-23-*oate* (**7**).—Prepared as in ref. 6 from methyl 3-oxo-24-norchola-1,4,7-trien-23-oate (2.5 g), *methyl* 3-*acetoxy*-24-*norchola*-1,3,5,7-*tetraen*-23-*oate* (2.7 g) formed pale yellow plates, m.p. 142—143 °C, from ether–methanol containing a trace of pyridine; $[\alpha]_D$ −470° (*c* 2.61); $\lambda_{max.}$ 251.5 nm (ε 11 200); τ 4.04 (3 H, m br), 4.30 (1 H, d), 6.35 (3 H, s, CO_2CH_3), and 7.83 (3 H, s, $OCOCH_3$) (Found: C, 76.3; H, 8.4%; M^+, 410.2455. $C_{26}H_{34}O_4$ requires C, 76.1; H, 8.4%; *M*, 410.2457).

The corresponding *methyl* 3-*acetoxychola*-1,3,5,7-*tetraen*-24-*oate* formed pale yellow needles, m.p. 122—123 °C, from the same solvent; $[\alpha]_D$ −490° (*c* 1.68) (Found: C, 76.3; H, 8.5; M^+, 424.2612. $C_{27}H_{36}O_4$ requires C, 76.4; H, 8.6%; *M*, 424.2614) whilst the 25-*homo-analogue* formed pale yellow plates, m.p. 93—95 °C; $[\alpha]_D$ −480 °C (*c* 1.06); $\lambda_{max.}$ 252 (ε 12 200) and 360 nm (ε 9 190) (Found: C, 76.8; H, 8.5%; M^+, 438.2771. $C_{28}H_{38}O_4$ requires C, 76.7; H, 8.7%; *M*, 438.2770). Reduction of unpurified methyl 3-acetoxy-24-norchola-1,3,5,7-tetraen-23-oate (2.75 g) as in ref. 6 gave *methyl* 3β-*hydroxy*-24-*norchola*-1,5,7-*trien*-23-*oate* (2.48 g) after purification from ether–light petroleum (1:5) on silica followed by crystallisation from ether–methanol, as plates, m.p. 170—171 °C; $[\alpha]_D$ −150° (*c* 1.9); $\lambda_{max.}$ 270 (ε 8 650), 280 (8 750), and 291 nm (4 970); τ 4.33 (3 H, m, 1-, 2-H and 6- or 7-H), 4.55 (1 H, m, 7- or 6-H), 5.72 (1 H, m, 3α-H), and 6.33 (3 H, s, CO_2CH_3) (Found: C, 77.9; H, 9.3%; M^+, 370.2506. $C_{24}H_{34}O_3$ requires C, 77.8; H, 9.3%; *M*, 370.2507).

The corresponding *methyl* 3β-*hydroxychola*-1,5,7-*trien*-24-*oate* formed plates, m.p. 101—102 °C, from ether–methanol; $[\alpha]_D$ − 190° (*c* 1.3) (Found: C, 78.1; H, 9.4%; M^+, 384.2662. $C_{25}H_{36}O_3$ requires C, 78.1; H, 9.4%; *M*, 384.2664), whilst the 25-*homo-analogue* formed microprisms, m.p. 93—95 °C, from ether–light petroleum; $[\alpha]_D$ −200° (*c* 0.41) (Found: C, 78.6; H, 9.7%; M^+, 398.2821. $C_{26}H_{38}O_3$ requires C, 78.4; H, 9.6%; *M*, 398.2820).

1α,3β-*Dihydroxy*-24-*norchola*-5,7-*dien*-23-*oic Acid* (**11**; R = H).—Prepared from methyl 3β-hydroxy-24-norchola-1,5,7-trien-23-oate (1.67 g) and 4-phenyl-1,2,4-triazoline-3,5-dione (0.6 g), as in ref. 6, the *adduct* (**8**; R = H) (1.23 g) formed needles, m.p. 169—170 °C (decomp.); $[\alpha]_D$ −30° (*c* 2.75) [Found: C, 70.8; H, 7.3; N, 7.4%; $(M + H)^+$(FAB), 546. $C_{32}H_{39}N_3O_5$ requires C, 70.4; H, 7.2; N, 7.7%; $(M + H)^+$, 546].

The *dimethyl-t-butylsilyl ether* (**8**; R = Me_2Bu^tSi) (1.5 g), prepared by the method of ref. 6 from this adduct (1.23 g), formed a non-crystalline wax, having *inter alia* τ 2.61 (5 H, m, Ph), 3.65 (2 H, q, $J_{6,7}$ 8.6 Hz, 6-, 7-H), 4.33 (2 H, bs, 1-, 2-H), 5.05 (1 H, m, 3α-H), 6.38 (3 H, s, CO_2CH_3), and 9.19 (9 H, s, t-Bu). Prepared as in ref. 6 from this ether (1.5 g), the *epoxide* formed a non-crystallisable pale yellow wax (1.4 g) having *inter alia* τ 2.61 (5 H, m, Ph), 3.70 (2 H, q, $J_{6,7}$ 8 Hz, 6-, 7-H), 5.08 (1 H, m, 3α-H), 6.35 (3 H, s, CO_2CH_3), and 9.09 (9 H, s, t-Bu) (Found: $(M + H)^+$ (FAB), 676. $C_{38}H_{53}N_3O_6Si + H^+$ requires 676).

A solution of crude epoxide (1.5 g) in tetrahydrofuran (75 ml), acetic acid (75 ml), and water (45 ml) was stirred at 60 °C. After 3 days the product was purified by chromatography from ether to yield the *adduct* of the 1α,2α-*epoxide* (**9**; R = H) (0.61 g) as needles, m.p. 153 °C, from methanol–methylene chloride; $[\alpha]_D$ −55° (*c* 1.56); τ 2.58 (5 H, m, Ph), 3.61 (2 H, q, $J_{6,7}$ 8 Hz, 6-, 7-H), 4.98 (1 H, m, 3α-H), and 6.33 (3 H, s, CO_2CH_3) [Found: C, 68.1; H, 7.2; N, 7.7%. M^+ (FAB), 562. $C_{32}H_{39}N_3O_6$ requires C, 68.4; H, 7.0; N, 7.5%; *M*, (for $C_{32}H_{39}N_3O_6{}^+H^+$) 562].

A solution of this epoxide adduct (0.44 g) in pyridine (40 ml) containing 1,5-diazabicyclo[4.3.0]non-5-ene (0.2 g) was refluxed (N_2) during 16 h, when the solvent was removed under reduced pressure and the residue dissolved in ethyl acetate. This extract was washed (i) with 5% (w/v) acetic acid and (ii) aqueous sodium hydrogen carbonate, dried, and evaporated when the residue was purified by flash chromatography on silica from ethyl acetate–light petroleum (1:3) to give *methyl* 1α,2α-*epoxy*-3β-*hydroxy*-24-*norchola*-5,7-*dien*-23-*oate* (**10**; R = Me) (0.18 g) which formed solvated needles, m.p. 181—182 °C, from light petroleum–methylene chloride; $[\alpha]_D$ −140° (*c* 0.94); $\lambda_{max.}$ 268 (ε 10 900), 278.5 (ε 11 300), and 290 nm (ε 6 290); τ 4.27 (1 H, d, $J_{6,7}$ 6 Hz, 6- or 7-H), 4.57 (1 H, d, $J_{6,7}$ 6 Hz, 7- or 6-H), 6.31 (3 H, s, CO_2CH_3), and 6.67 and 6.96 (2 H, ABq, $J_{1,2}$ 4 Hz, 1-, 2-H) [Found: C, 73.7; H, 9.0%; M^+, 386.2457. $C_{24}H_{34}O_4 0.5H_2O$ requires C, 73.5; H, 8.9%; *M*, (for $C_{24}H_{34}O_4$) 386.2457]. The *acetate* of this epoxide formed (quantitatively) needles, m.p. 170 °C., from methanol–methylene chloride; τ 4.29 (1 H, d, $J_{6,7}$ 6 Hz, 6- or 7-H), 4.52 (1 H, d, $J_{6,7}$ 6 Hz, 7- or 6-H), 6.37 (3 H, s, CO_2CH_3), 7.94 (3 H, s, $OCOCH_3$), and 6.63 and 6.90 (2 H, ABq, $J_{1,2}$ 4 Hz, 1-, 2-H) (Found: C, 73.0; H, 8.5%; M^+, 428.2564. $C_{26}H_{36}O_5$ requires C, 72.9; H, 8.5%; *M*, 428.2563). Methyl 3β-hydroxy-1α,2α-epoxy-24-norchola-5,7-dien-23-oate (0.18 g) dissolved in methanol (20 ml) containing potassium hydroxide (4.0 g) was maintained at 20 °C during 7.5 h. Purified from acetone, 1α,2α-*epoxy*-3β-*hydroxy*-24-*norchola*-5,7-*dien*-23-*oic acid* (**10**; R = H) (0.16 g) formed prisms, m.p. 212 °C (decomp.); $[\alpha]_D$ −110° (*c* 0.73, in tetrahydrofuran) (Found: C, 74.5; H, 8.5%; M^+, 372.2301. $C_{23}H_{32}O_4$ requires C, 74.2; H, 8.7%; *M*, 372.2301). A solution of this epoxide (0.15 g) in tetrahydrofuran (45 ml) containing lithium borohydride (1.0 g) was refluxed (N_2) for 2.5 h. Purified from aqueous tetrahydrofuran, 1α,3β-*dihydroxy*-24-*norchola*-5,7-*dien*-23-*oic acid* (**11**; R = H) (0.15 g) formed plates, m.p. 244 °C (decomp.); $[\alpha]_D$ −130° (*c* 0.61, in tetrahydrofuran); $\nu_{max.}$ 3 460, 3 040, 2 965 (OH), and 1 732 cm^{-1} (CO in CO_2H); $\lambda_{max.}$ 271 (ε 10 700), 281 (11 300), and 293 nm (6 690) (Found: C, 73.3; H, 9.1%; M^+, 374.2452. $C_{23}H_{34}O_4$ requires C, 73.8; H, 9.2%; *M*, 374.2457). Prepared (quantitatively) from this acid using diazomethane in ether–methanol, *methyl* 1α,3β-*dihydroxy*-24-*norchola*-5,7-*dien*-23-*oate* (**11**; R = Me_3) formed needles, m.p. 176 °C, from acetone–light petroleum; $[\alpha]_D$ −68° (*c* 0.69); $\lambda_{max.}$ 271 (ε 11 500), 282 (12 200), and 294 nm (7 300); τ 4.27 (1 H, d, $J_{6,7}$ 6 Hz, 6-or 7-H), 4.63 (1 H, d, $J_{6,7}$ 6 Hz, 7-or 6-H), 5.93 (1 H, m, 3α-H), and 6.35 (3 H, s, CO_2CH_3) (Found: C, 74.2; H, 9.2%; M^+, 388.2609. $C_{24}H_{36}O_4$ requires C, 74.2; H, 9.3%; *M*, 388.2614).

A solution of this ester (65 mg) in ether (900 ml) was purged for 15 min with nitrogen and then irradiated for 35 min, at 5 °C, with an Hanovia medium-pressure mercury-vapour lamp. Fluorenone (73 mg) was then added and irradiation continued for a further 30 min. After removal of the solvent under reduced pressure the yellow semi-crystalline residue was partially purified by chromatography from methanol–chloroform (1:10) on silica impregnated (3%, w/w) with silver nitrate. Unchanged 5,7-diene was separated and the residue (25 mg), dissolved in benzene (15 ml) and ethanol (15 ml), was refluxed (N_2) during 2.5 h. After removal of the solvent under reduced pressure at room temperature the residue was purified by p.l.c. using silica containing 3% (w/w) of silver nitrate and acetone–light petroleum (2:5) as the solvent. Elution of the more polar band with ether followed by three repetitions of this process with this

fraction gave *methyl* 1α,3β-*dihydroxy*-9,10-*seco*-24-*norchola*-5,7-10(19)-*trien*-23-*oate* (10 mg) as an oil; $\lambda_{max.}$ 261.5, $\lambda_{min.}$ 226.5 nm ($\varepsilon_{max.}/\varepsilon_{min.}$ 1.55); τ 4.44 (1 H, br s, 19*E*-H), 5.00 (1 H, br s, 19*Z*-H), 5.57 (1 H, m, 1β-H), 5.77 (1 H, m, 3α-H), and 6.31 (3 H, s, CO_2CH_3) (Found: M^+, 388.2610. $C_{24}H_{36}O_4$ requires *M*, 388.2614). A solution of this ester (10 mg) in methanol (4 ml) containing potassium hydroxide (0.4 g) was kept (N_2) at room temperature and the product isolated after 6 h. Purification from ether–light petroleum gave 1α,3β-*dihydroxy*-9,10-*seco*-24-*norchola*-5,7,10(19)-*trien*-23-*oic acid* (**1**; R = H, *n* = 1) (8.7 mg) as microprisms, m.p. 122—126 °C (decomp.). This product was homogenous on t.l.c. and had $\lambda_{max.}$ 262 (ε 18 000), $\lambda_{min.}$ 226 nm (ε 10 000); τ 3.57 and 3.94 (2 H, ABq, $J_{6,7}$ 11 Hz, 6-, 7-H), 4.44 (1 H, br s, 19*E*-H), 4.96 (1 H, br s, 19*Z*-H), 5.54 (1 H, m, 1β-H), 5.74 (1 H, m, 3α-H), 7.20 (1 H, m, 9β-H), 9.01 (3 H, d, $J_{21,20}$ 6 Hz, 21-Hz), and 9.41 (3 H, s, 18-Hz). (Found: C, 73.7; H, 9.1%; M^+, 374.2452. $C_{24}H_{34}O_4$ requires C, 73.8; H, 9.2%; *M*, 374.2457).

1α,3β-*Dihydroxy*-9,10-*secochola*-5,7,10(19)-*trien*-24-*oic Acid*.—The *adduct* of methyl 3β-hydroxychola-1,5,7-trien-24-oate with 4-phenyl-1,2,4-triazoline-3,5-dione formed microprisms, m.p. 187—191 °C, from ether; $[\alpha]_D$ −37° (*c* 1.83) [Found: C, 73.4; H, 7.7; N, 8.6%; $(M + H)^+$(FAB) 560. $C_{33}H_{41}N_3O_5$ requires C, 70.8; H, 7.4; N, 7.5%; $(M + H)^+$560]. The *t-butyldimethylsilyl ether* of this adduct formed needles, m.p. 178 °C, from methanol–methylene chloride (after chromatography); $[\alpha]_D$ +47.5° (*c* 3.00) (Found: C, 69.4; H, 8.2; N, 6.3. $C_{39}H_{55}N_3O_5Si$ requires C, 69.5; H, 8.2; N, 6.2%).

The 1α,2α-*epoxide* of this silyl ether formed needles, m.p. 174 °C, from methanol–methylene chloride; $[\alpha]_D$ −53° (*c* 2.14) [Found: C, 68.1; H, 8.1; N, 6.2%; $(M + H)^+$(FAB) 690. $C_{39}H_{55}N_3O_6Si$ requires C, 67.9; H, 8.0; N, 6.1%; $(M + H)^+$690].

This desilylated epoxide formed needles, m.p. 142—143 °C (decomp.), from acetone; $[\alpha]_D$ −55° (*c* 1.19) (Found: C, 68.4; H, 7.2; N, 7.5%; $(M + H)^+$(FAB) 576. $C_{33}H_{41}N_3O_6$ requires C, 68.8; H, 7.2; N, 7.3%; $(M + H)^+$576].

Removal of the triazoline residue from this epoxide gave *methyl* 3β-*hydroxy*-1α,2α-*epoxychola*-5,7-*dien*-24-*oate* as plates, m.p. 146—148 °C, from ether–methanol; $[\alpha]_D$ −110° (*c* 0.43) (Found: C, 75.0; H, 9.1; M^+, 400.2609. $C_{25}H_{36}O_4$ requires C, 75.0; H, 9.1%; *M*, 400.2613). Hydrolysis of this ester to 3β-*hydroxy*-1α,2α-*epoxychola*-5,7-*dien*-24-*oic acid* during 4.5 h, gave clusters of needles, m.p. 188—190 °C (decomp.), from ether; $[\alpha]_D$ −92° (*c* 0.61) (Found: C, 73.9; H, 8.7%; M^+, 386.2463. $C_{24}H_{34}O_4$ requires C, 74.6; H, 8.8%; *M*, 386.2457).

1α,3β-*Dihydroxychola*-5,7-*dien*-24-*oic acid* formed needles, m.p. 227—228 °C (decomp.), from aqueous tetrahydrofuran; $[\alpha]_D$ −120° (*c* 0.83 in tetrahydrofuran) (Found: C, 74.1; H, 9.2%; M^+, 388.2611. $C_{24}H_{36}O_4$ requires C, 74.2; H, 9.3%; *M*, 388.2613).

The *methyl ester* of this acid separated from acetone–light petroleum as needles, m.p. 120 °C; $[\alpha]_D$ −70° (*c* 1.25) (Found: C, 74.3; H, 9.4%; M^+, 402.2770. $C_{25}H_{38}O_4$ requires C, 74.6; H, 9.5%; *M*, 402.2770).

Prepared from this ester (0.1 g), pyridine (20 ml), 4-(dimethylamino)pyridine (0.2 g), and acetic anhydride (20 ml), *methyl* 1α,3β-*diacetoxychola*-5,7-*dien*-24-*oate* (0.09 g) formed needles, m.p. 142 °C, from methanol–methylene chloride; $[\alpha]_D$ −29° (*c* 1.19) (Found: C, 71.3; H, 8.6%; M^+, 486.2981. $C_{29}H_{42}O_6$ requires C, 71.6; H, 8.7%; *M*, 486.2981).

Photolysis of this ester (81.7 mg) as for the 24-nor analogue gave *methyl* 1α,3β-*diacetoxy*-9,10-*secochola*-5,7,10(19)-*trien*-24-*oate* (15 mg) as an oil, $\lambda_{max.}$ 264 nm (ε 18 000). τ 3.63, 4.08 (2 H, ABq, $J_{6,7}$ 11 Hz, 6-, 7-H), 4.52 (1 H, t, $J_{1.2\alpha}$ 5 Hz $J_{1.2\beta}$ 5 Hz, 1β-H), 4.68 (1 H, br s, 19*Z*-H), 4.81 (1 H, m, 3α-H), 4.95 (1 H, bs *H*-19E), 6.63 (3 H, s, $-CO_2CH_3$), 7.21 (1 H, m, 9β-H), 7.95 (3 H, s, $OCOCH_3$), 7.98 (3 H, s, $OCOCH_3$), 9.08 (3 H, d, $J_{21.20}$ 4.4 Hz, 21-H_3), and 9.48 (3 H, s, 18-H_3) (Found: M^+, 486.2986. $C_{29}H_{42}O_6$ requires *M*, 486.2981).

Hydrolysis of this ester (15 mg) gave 1α,3β-*dihydroxy*-9,10-*secochola*-5,7,10(19)-*trien*-24-*oic acid* (11.4 mg) as microprisms, m.p. 117—120 °C (decomp.), from ether–light petroleum; $\lambda_{max.}$ 263 (ε 18 200), $\lambda_{min.}$ 227 nm (ε 10 400); τ 3.58, 3.96 (2 H, ABq, $J_{6,7}$ 11.6 Hz, 6-H, 7-H), 4.65 (1 H, bs, 19*E*-H), 4.97 (1 H, bs, 19*Z*-H), 5.54 (1 H, m, 1β-H), 5.75 (1 H, m, 3α-H), 7.20 (1 H, m, 9β-H), 9.05 (H, d, $J_{21.20}$ 5.5 Hz, 21-H_3), and 9.45 (3 H, s, 18-H_3) (Found: C, 74.4; H, 9.3; M^+, 388.2621. $C_{24}H_{36}O_4$ requires C, 74.2; H, 9.3%; *M*, 388.2613).

1α,3β-*Dihydroxy*-9,10-*seco*-25-*homochola*-5,7,10(19)-*trien*-25-*oic acid*.—The *adduct* of methyl 3β-hydroxy-25-homochola-1,5,7-trien-25-oate and 4-phenyl-1,2,4-triazoline-3,5-dione formed small prisms, m.p. 153—155 °C (decomp.), from aqueous acetone; $[\alpha]_D$ −35° (*c* 3.01) [Found: C, 69.1; H, 7.4; N, 6.0; $(M + H)^+$(FAB) 574. $C_{34}H_{43}N_3O_5$ requires C, 71.2; H, 7.6; N, 7.3%; $(M + H)^+$574].

The dimethyl-t-butylsilyl ether of this adduct was an oil, having *inter alia* τ 2.65 (5 H, m, Ph), 3.68 (2 H, ABq, $J_{6,7}$ 8 Hz, 6-, 7-H), 4.34 (2 H, br s, 1-, 2-H), 5.05 (1 H, m, 3α-H), 6.33 (3 H, s, CO_2CH_3), and 9.14 (9 H, s, $SiBu^t$).

The 1α,2α-epoxide of this ether also failed to crystallise and exhibited *inter alia* τ 3.62 (5 H, s, Ph), 3.72 (2 H, ABq, $J_{6,7}$ 8 Hz, 6-, 7-H), 6.77 (3 H, s, CO_2Me), 6.84 (2 H, q, $J_{1.2}$ 4.4 Hz, $J_{2.1}$ 4.5 Hz, 2-H).

The *desilylated epoxide* formed needles, m.p. 194—195 °C (decomp.), from acetone–light petroleum; $[\alpha]_D$ −70° (*c* 1.13); τ 2.58 (5 H, m, Ph), 3.68 (2 H, ABq, $J_{6,7}$ 8 Hz, 6-, 7-H), 4.96 (1 H, m, 3α-H), 6.33 (3 H, s, CO_2Me), 6.76 (1 H, dd, $J_{4\alpha.4\beta}$ 14.8 Hz, $J_{3.4\alpha}$ 9 Hz, 4α-H), and 6.78 (2 H, br s, 1-, 2-H) [Found: C, 69.3; H, 7.4; N, 7.1; $(M + H)^+$(FAB) 590. $C_{34}H_{43}N_3O_6$ requires C, 69.2; H, 7.4; N, 7.1%; $(M + H)^+$ 590]. After removal of the triazoline residue from this adduct, *methyl* 1α,2α-*epoxy*-3β-*hydroxy*-25-*homochola*-5,7-*dien*-25-*oate* formed a non-crystallisable oil; $[\alpha]_D$ −110° (*c* 0.95); $\lambda_{max.}$ 260.5 (ε 8 020), 268 (11 000), 279 (11 300), and 290 nm (6 320), $\lambda_{min.}$ 273.5 nm (ε 9 450); τ 4.27 (H, d, $J_{6,7}$ 6 Hz, 6- or 7-H), 6.31 (3 H, s, CO_2Me), 4.58 (1 H, m, 7- or 6-H), and 6.67 and 6.96 (2 H, ABq, $H_{1.2}$ 3.8 Hz, 2-, 1-H) (Found: M^+, 414.2770. $C_{26}H_{38}O_4$ requires *M*, 414.2770).

1α,2α-*Epoxy*-3β-*Hydroxy*-25-*homochola*-5,7-*dien*-25-*oic acid* formed needles, m.p. 182—183 °C (decomp.) from acetone–light petroleum; $[\alpha]_D$ −74° (*c* 0.89) (Found: C, 74.8; H, 9.0%; M^+, 400.2613. $C_{25}H_{36}O_4$ requires C, 75.0; H, 9.1%; *M*, 400.2614).

1α,3β-*Dihydroxy*-25-*homochola*-5,7-*dien*-25-*oic acid* formed needles, m.p. 213 °C (decomp.), from aqueous methanol; $[\alpha]_D$ −150° (*c* 1.22 in tetrahydrofuran); $\nu_{max.}$ 3 405 cm^{-1} (OH) (Found: C, 74.0; H, 9.5%; M^+, 402.2767. $C_{25}H_{38}O_4$ requires C, 74.6; H, 9.5% *M*, 402.2770).

The *methyl ester* separated from acetone–light petroleum as needles, m.p. 112 °C; $[\alpha]_D$ −117° (*c* 0.93) (Found: C, 75.2; H, 9.60%; M^+, 416.2931. $C_{26}H_{40}O_4$ requires C, 75.0; H, 9.7%; *M*, 416.2927).

Methyl 1α,3β-*diacetoxy*-25-*homochola*-5,7-*dien*-25-*oate* separated from ether–methanol as needles, m.p. 137 °C; $[\alpha]_D$ −48° (*c* 1.16); τ 4.28 (1 H, d, $J_{6,7}$ 6 Hz, 6- or 7-H), 4.58 (1 H, m, 7- or 6-H), 4.97 (2 H, m, 1-, 2-H), 6.32 (3 H, s, CO_2Me), 7.90 (3 H, s, OCOMe), 7.96 (3 H, s, OCOMe). (Found: C, 71.9; H, 8.8%; M^+, 500.3142. $C_{30}H_{44}O_6$ requires C, 72.0; H, 8.9%; *M*, 500.3138).

Photolysis of this ester (95 mg) gave *methyl*-1α,3β-*diacetoxy*-9,10-*seco*-25-*homochola*-5,7,10(19)-*trien*-25-*oate* (14 mg) as an oil; $\lambda_{max.}$ 264 nm (ε 18 000); τ 3.64, 4.09 (2 H, ABq, $J_{6,7}$ 11 Hz, 6-, 7-H), 4.53 (1 H, t, $J_{1.2\alpha}$ 5 Hz, $J_{1.2\beta}$ 5 Hz, 1β-H), 4.69 (1 H, br s, 19*E*-H), 4.83 (1 H, m, 3α-H), 4.96 (1 H, br s, 19*Z*-H), 6.32 (3 H, s, CO_2Me), 7.21 (1 H, m, 9β-H), 7.95 (3 H, s, OCOMe), and 7.97 (3

H, s, OCOMe) (Found: M^+, 500.3147. $C_{30}H_{44}O_6$ requires M, 500.3138).

Hydrolysis of this ester (13.1 mg) gave 1α,3β-*dihydroxy*-9,10-*seco*-25-*homochola*-5,7,10(19)-*trien*-25-*oic acid* (10 mg) as micro-needles, m.p. 97—101 °C, from ether–light petroleum; $\lambda_{max.}$ 264 nm (ε 18 300), $\lambda_{min.}$ 228 nm (10 300); τ 3.58, 3.96 (2 H, ABq, $J_{6,7}$ 11.6 Hz, 6-, 7-H), 4.65 (1 H, br s, 19*E*-H), 4.97 (1 H, br s, 19*Z*-H), 5.55 (1 H, m, 1β-H), 5.75 (1 H, m, 3α-H), and 7.19 (1 H, m, 9β-H) (Found: C, 74.6; H, 9.6%; M^+, 402.2772. $C_{25}H_{38}O_4$ requires C, 74.6; H, 9.5%; M, 402.2770).

2,2,2-*Trichloroethyl* 3-*Acetoxychola*-1,3,5,7-*tetraen*-24-*oate*.—Hydrolysis of methyl 3-oxo-5α-chol-7-en-24-oate (1.84 g) in methanol (200 ml) containing potassium hydroxide (10 g) at 20 °C during 12 h gave 3-*oxo*-5α-*chol*-7-*en*-24-*oic acid* (1.63 g) which formed plates, m.p. 205—207 °C (decomp.) from acetone (Found: C, 77.5; H, 9.9. $C_{24}H_{36}O_3$ requires C, 77.4; H, 9.7%). A solution of this acid (1.63 g) in methylene chloride (80 ml) containing pyridine (0.7 g), 2,2,2-trichloroethanol (1.3 g) and dicyclohexylcarbodi-imide (0.9 g) was stirred overnight to yield 2,2,2-*trichloroethyl*-3-*oxo*-5α-*chol*-7-*en*-24-*oate* (1.3 g) as plates, m.p. 122 °C, from methanol containing a trace of pyridine; $[\alpha]_D$ +25° (*c* 1.62) (Found: C, 62.0; H, 7.3%; M^+, 502.1810. $C_{26}H_{37}{}^{35}Cl_3O_3$ requires C, 62.0 H, 7.4%; M, 502.1808).

Bromination of this ketone (0.6 g) with tri-*N*-methylanilinium perbromide (4.13 g) gave 2,2,2-*trichloroethyl* 2ξ,4ξ-*dibromo*-3-*oxo*-5α-*chol*-7-*en*-24-*oate* (0.46 g) as microprisms, m.p. 163—165 °C (decomp.), from light petroleum–ether; $[\alpha]_D$ −47.0° (*c* 1.26) (Found: C, 47.2; H, 5.3%; M^+, 661.9969. $C_{26}H_{35}{}^{79}Br^{81}Br^{35}Cl_2{}^{37}ClO_3$ requires, C, 47.2; H, 5.3%; M, 661.9968). Prepared from this dibromide 2,2,2-*trichloroethyl*-3-*oxochola*-1,4,7-*trien*-24-*oate* formed a yellow oil (Found: M^+, 499.1569. $C_{25}H_{34}{}^{35}Cl_3O_3$ requires M, 499.1574).

Prepared from this ketone (1.6 g), 2,2,2-*trichloroethyl*-3-*acetoxychola*-1,3,5,7-*tetraen*-24-*oate* (1.6 g) formed pale yellow needles, m.p. 150—152 °C, from ether–methanol containing 0.01% pyridine; $[\alpha]_D$ −430° (*c* 2.32) (Found: C, 61.9; H, 6.7%; M^+, 540.1605. $C_{28}H_{35}{}^{35}Cl_3O_4$ requires C, 62.1; H, 6.5%; M, 540.1601).

Acknowledgements

We are indebted to the S.E.R.C. for a studentship (to B. R. de C.) and to the British Council for awards to two of us (N. T. M. and N. M.). Glaxo Research Ltd. kindly prepared a large batch of 5,6-dihydroergosterol.

References

1 Part 11, J. M. Midgley, R. M. Upton, R. A. Watt, W. B. Whalley, and X-M. Zhang, *J. Chem. Res.* (*S*), 1983, 273; (*M*), 2513.
2 *Inter alia*. 'Vitamin D: Basic Research and its Clinical Applications: Proceedings of the Fourth Workshop on Vitamin D,' A. W. Norman, K. Schaefer, D. Herrath, H. G. Grigoleit, J. W. Coburn, H. F. DeLuca, E. B. Mawr, and T. Suda, Walter de Gruyter (Berlin), 1979, p. 445.
3 R. P. Esvelt, H. K. Schnoes, and H. F. DeLuca, *Biochemistry*, 1979, **18**, 3977.
4 R. P. Esvelt, M. A. Fivizzani, H. E. Paaren, H. K. Schnoes, and H. F. DeLuca, *J. Org. Chem.*, 1981, **46**, 456.
5 N. Koizumi, M. Morisaki, N. Ikekawa, Y. Tanake, and H. F. DeLuca, *J. Steroid Biochem.*, 1979, **10**, 261.
6 A. Emke, D. Hands, J. M. Midgley, and W. B. Whalley, *J. Chem. Soc., Perkin Trans. 1*, 1977, 820.
7 W. Sucrow and B. Girgensohn, *Chem. Ber.*, 1970, **103**, 745.
8 M. J. T. Robinson and W. B. Whalley, *Tetrahedron*, 1963, **19**, 2123 and references cited therein.
9 A. B. Garry, J. M. Midgley, W. B. Whalley, and B. J. Wilkins, *J. Chem. Soc., Perkin Trans. 1*, 1977, 809.

Received 27*th September* 1984; *Paper* 4/1664