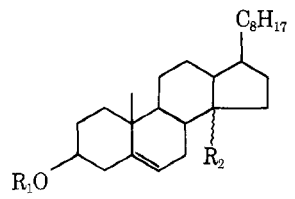
Synthesis of 14β-Cholest-5-en-3β-ol

Mario Anastasia,* Antonio Scala, and Giovanni Galli

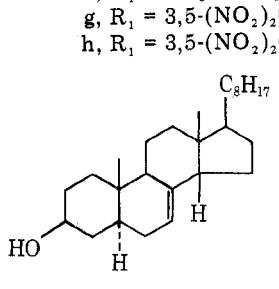
Institute of Chemistry, School of Medicine, University of Milan, I-20133 Milano, Italy, and Institute of Pharmacology and Pharmacognosy, Laboratory of Applied Biochemistry, School of Pharmacy, University of Milan, I-20129 Milano, Italy

Received July 25, 1975

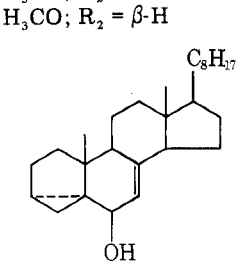
A previous work from this laboratory on the intermediary role of some sterols in the biosynthesis of cholesterol (**1a**) indicates that a cis B/C ring junction, opposite to the trans ring junction of natural compounds, does not prevent the conversion of a sterol molecule to **1a** by liver enzymes.[1] On the basis of this observation it could be postulated that also sterols with a cis C/D ring junction might be enzymatically transformed. In addition, sterols with a modified stereochemistry with respect to that of the natural compounds could exhibit an inhibitory effect on the biosynthesis of **1a** as it has been shown for the triterpenoid euphol, which differs from lanosterol only in stereochemistry.[2] The synthesis of 5α,14β-cholest-7-en-3β-ol (**2**), a sterol suitable to check the influence of a cis fusion of C/D rings on sterol biosynthesis, has been described elsewhere.[3] A simple synthesis of 14β-cholest-5-en-3β-ol (**1b**) from 3α,5α-cyclocholest-7-en-6β-ol (**3**) is now described, since **1b** may be a metabolite in the enzymatic transformation of **2.** The oxidation of **3** to the corresponding ketone **4** either with manganese dioxide in chloroform[4] or with chromium trioxide in pyridine[5] gave unsatisfactory results in our hands; the compound was recovered unchanged after treatment with manganese dioxide at temperatures below 40 °C, whereas dehydration took place with the formation of consistent amounts of the diene **5a** both by oxidation with manganese dioxide at temperatures above 40 °C or by oxidation with chromium trioxide in pyridine. Approximately 80% yield of pure 3α,5α-cyclocholest-7-en-6-one (**4**) was obtained, with a solution of chromium trioxide in pyridine–dichloromethane (1:3.6).



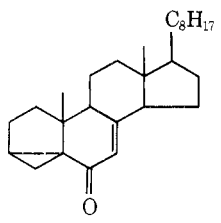
1a, R_1 = H; R_2 = α-H
b, R_1 = H; R_2 = β-H
c, R_1 = C_2H_5CO; R_2 = α-H
d, R_1 = C_2H_5CO; R_2 = β-H
e, R_1 = CH_3CO; R_2 = α-H
f, R_1 = CH_3CO; R_2 = β-H
g, R_1 = 3,5-$(NO_2)_2C_6H_3CO$; R_2 = α-H
h, R_1 = 3,5-$(NO_2)_2C_6H_3CO$; R_2 = β-H



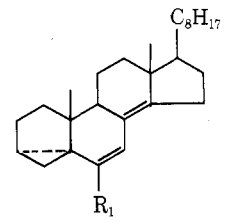
2



3



4



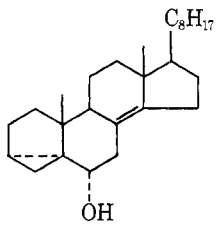
5a, R_1 = H
b, R_1 = *O*-alkyl
c, R_1 = *O*-acyl
d, R_1 = $OSi(CH_3)_3$

A simple method to obtain 14β-steroids was proposed recently by Mincione et al., consisting in hydroboration of 8(14)-ene steroids at controlled temperature.[6] In our case it was necessary to isomerize the Δ^7 double bond of the unsaturated ketone **4** to the 8(14) position. Treatment of **4** with various bases results in an equilibrium mixture of 7- and 8(14)-double bond isomers. Unfortunately, the 8(14)-double bond isomer is unstable and reverts to the original during work-up.[4b] On the other hand, a heteroannular 6,8(14) diene structure **5b** or **5c** can be anticipated for dienol derivatives of ketone **4,** the 6,8(9) homoannular isomer being less stable as shown by uv absorption of simple unsubstituted dienes.[7] Reduction of the dienol derivative would give a homoallylic alcohol **6** suitable for the following hydroboration reaction. Dienol ethers **5b** and their thio analogues were not considered as suitable derivatives of **4** because of their stability under basic conditions, which precludes generation of the $\Delta^{8(14)}$-6-one system under the usual reducing conditions (i.e., $NaBH_4$). Dienol esters **5c** were also excluded since the acidic conditions required for their preparation[8] cleave the cyclopropane ring.[4b,9]

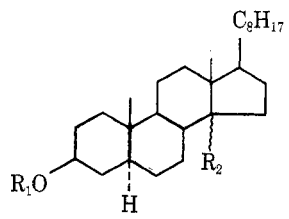
Trimethylsilyl enol ethers can be obtained by addition of trimethylsilyl chloride to sodium enolates generated by heating a solution of the ketone in glyme with sodium hydride.[10] Under these conditions the cyclopropane ring is unaffected.[5a] Application of this procedure with minor modifications to the unsaturated ketone **4** resulted in the formation of 6-trimethylsilyloxy-3α,5α-cyclocholesta-6,8(14)-diene (**5d**) in nearly quantitative yield. This compound was crystallized from 2-propanol. Its spectral properties were in agreement with the proposed structure; the uv absorption spectrum (λ_{max} 263 nm, ε 24 200) was consistent with a 6,8(14) heteroannular diene;[7] double bond stretching frequencies were at 1620 and 1650 cm^{-1}.[11] The ^1H NMR spectrum showed the 10β- and 13β-methyl signals at δ 0.79 and 0.88, respectively, in accordance with the signals at δ 0.76 and 0.89 of the corresponding methyl groups of diene **5a;**[12] the vinylic proton at position 7 gave a sharp singlet and multiplet at very high fields confirming the presence of the cyclopropane ring. The mass spectrum of the compound showed the molecular ion at the expected value. No ion corresponding to the formal loss of trimethylsilanol was present. This behavior under the electron impact which has been already observed for 3-trimethylsilyloxycholesta-3,5-diene[13] confirmed the presence of a trimethylsilyl dienol ether system in the molecule. No hydrolysis of the trimethylsilyloxy group was observed during the

isolation and the purification both by chromatographic methods and by crystallization.

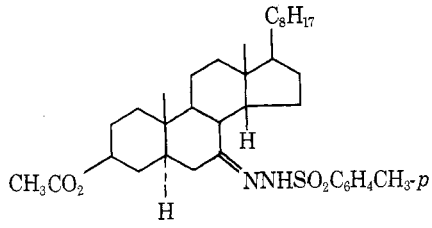
The trimethylsilyl derivative **5d** was reduced with sodium borohydride in 2-propanol to 3α,5α-cyclocholest-8(14)-en-6α-ol (**6**). The structure was assigned on the basis of ^{1}H NMR information. A downfield shift of the 13β-methyl signal with respect to the 13β-methyl signal of 3α,5α-cyclocholestan-6α-ol[14] was observed which was attributable to the presence of the 8(14) double bond.[15a] As expected, cyclopropane protons were present at very high fields. A quadruplet centered at δ 2.73 ($J_{6\beta,7\beta}$ = 5, $J_{7\alpha,7\beta}$ = −13 Hz) was assigned to the equatorial 7β proton whereas absorption of axial 7α proton was included in the methylene envelope. The axial (i.e., β) configuration in a chair conformation was attributed to the 6 hydrogen since this proton signal was present in the ^{1}H NMR spectrum as the X part of an AMX system;[15b] the quadruplet centered at δ 3.83 ($J_{6\beta,7\alpha}$ = 11.5, $J_{6\beta,7\beta}$ = 5 Hz) collapsed to a doublet ($J_{6\beta,7\alpha}$ = 11.5 Hz) by irradiation at δ 2.73. The mass spectrum of **6** was almost identical with that of diene **5a** owing



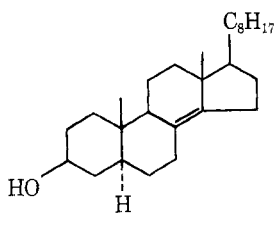
6



7a, R_1 = H; R_2 = β-H
b, R_1 = CH_3CO; R_2 = β-H
c, R_1 = CH_3CO; R_2 = α-H
d, R_1 = 3,5-$(NO_2)_2C_6H_3CO$; R_2 = β-H
e, R_1 = C_2H_5CO; R_2 = α-H
f, R_1 = C_2H_5CO; R_2 = β-H



8



9

to the thermal or electronic loss of water; a prominent ion at *m/e* 199 attributable to the presence of the 3α,5α-cyclo-6,8(14)-diene system was present in the spectrum.[16]

It is known that hydroboration of 8(14)-ene steroids occurs with the addition of boron predominantly at the 15 position[6] whereas the cyclopropane ring is opened only at elevated temperature.[17] Therefore the alcohol **6** was hydroborated at 50–60 °C in diglyme and protonolysis of boron was obtained by refluxing in diglyme–propionic acid for 48 h.[18] Under these conditions the 3β-propionyloxy-5-ene system was generated from the 3α,5α-cyclo-6α-hydroxy system in agreement with the results obtained by other authors by treatment of a 3α,5α-cyclo-6β-methoxy steroid with acetic acid.[19] A 1:1.5 mixture of propionates **1c** and **1d** was obtained from the reaction.

It has been reported that addition of boron occurs to the carbon atom β to the enol ether substituent.[20] This fact prompted us to check the hydroboration reaction of the trimethylsilyloxy derivative **5d.** Treatment of the compound with diborane under a variety of conditions, followed by protonolysis, resulted in a mixture of compounds containing only traces of the two isomers **1c** and **1d.**

Separation of pure 14β diastereoisomer **1d** from **1c** was obtained by one crystallization of the mixture of the two isomers from methanol; **1c** crystallized whereas **1d** was recovered 95–99% pure by evaporation of the mother liquors. Attempts to crystallize **1d** and **1f** from a number of solvents were unsuccessful: crystallization of the free alcohol **1b** was achieved by keeping a solution of the compound in methanol for 1 month at −20 °C. An easily crystallizable derivative was found to be the 3,5-dinitrobenzoate **1h.** Considering solubilities in common organic solvents and GLC retention time values it can be deduced that 14β isomers are more soluble and less retained than the corresponding 14α isomers.

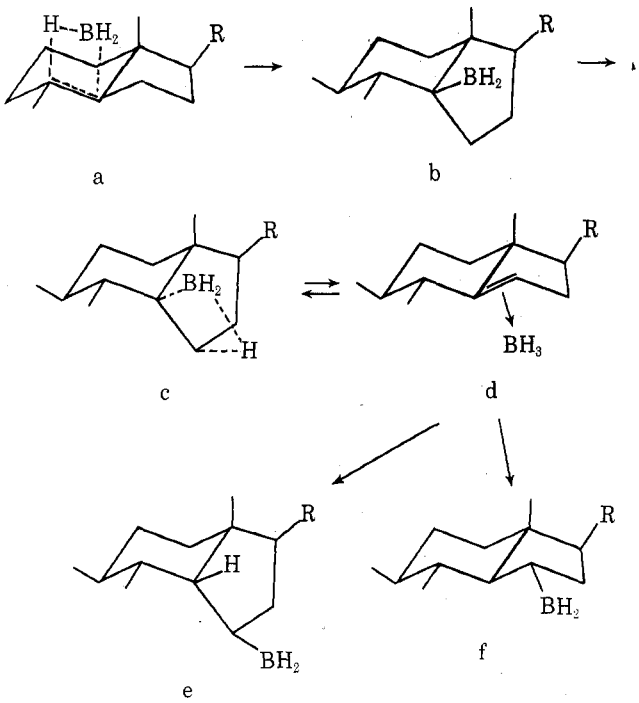
The structure of **1f** was assigned on the basis of its spectroscopic properties; the δ values of signals of 10β- and 13β-methyl groups in the ^{1}H NMR spectrum were in agreement (±0.04 ppm) with those calculated on the basis of Zürcher's rule;[15c,d] 3α- and 6-proton signals in the ^{1}H NMR spectra of both **1g** and **1h** were observed at the same fields and showed identical half-band widths. The mass spectra of **1e**[21] and **1f** showed similar fragmentation patterns differing only for the relative intensities of peaks at *m/e* 120, 213, 247, 255, and 260. Moreover, the evaporation temperature of **1f** was about 40 °C lower with respect to that of **1e** when analyzed by direct introduction in the ion source.

The structure of **1f** was confirmed by hydrogenation and transformation to crystalline **7d,** which was also obtained either by sodium borohydride reduction of 5α,14β-cholestan-3β-ol-7-one acetate tosylhydrazone (**8**)[3] and subsequent saponification and esterification or by hydroboration of 5α-cholest-8(14)-en-3β-ol (**9**) followed by protonolysis, fractional crystallization, saponification, and esterification. The ^{1}H NMR information for **7b** was consistent with the assigned structure; 10β- and 13β-methyl signals were at the calculated values (±0.04 pm);[15a,d] the 3α proton absorbed at the same field and with identical half-band width as the 3α proton of **7c.** The acetates **7b** and **7c**[22] showed mass spectra differing only for the relative intensities of a number of peaks. The diagnostic value for the 14β isomer can be attributed to triplets centered at *m/e* 216 and 276, respectively.

Hydroboration of 8(14)-ene sterols yields 15-boron compounds as the end products.[6] The excess diborane produced at the hydroboration stage may catalyze the low-temperature isomerization.[23] However, a reaction mechanism can be postulated implying relatively nonpolar transition states, by which the influence of steric factors on the direction of the addition might be explained.[24] According to this mechanism, diborane adds first to the β side of the molecule since addition to the α side would give rise to an highly unstable B/C ring junction. On the basis of model examination, the conformation of ring C in transition state a should revert to chair in compound b. Compounds e and f with a boron at 15β and 15α position, respectively, would be formed from compound b through transition state c and π complex d (Scheme I).

It has been reported that protonolysis of 15α-boron compounds requires 8 h.[25] However, the above reported 1.5:1 ratio of 14β to 14α isomers was obtained after only 47 h; on the other hand only the 14α isomer was detectable after 8 h. The different rate is attributable to the lower rate of formation of the cyclic transition state owing to hindrance of

Scheme I



the boron atom in the 15β configuration.[26] The observed 1.5:1 ratio between 14β and 14α isomers significantly differs from the reported 5:1 ratio.[6] This discrepancy might be caused by the more drastic reaction conditions utilized by us (time and temperature) which favor side reactions as demonstrated by the decreased yields of **1c** and **1d** after 72 reaction h.

The availability of **1b** opens the way to various biological investigations. The inhibition of **1a** biosynthesis is a first possibility. The transformation of **1b** in tissues should also be examined with special regard to the possible role of the compound in modulating the synthesis of steroid hormones.

Experimental Section

All melting points are uncorrected. Infrared spectra were taken as Nujol mulls and absorptions are reported as reciprocal centimeters, NMR spectra were taken on a Perkin-Elmer R-24 as chloroform-d_1 solutions and are reported as δ units relative to Me_4Si, and optical rotations were taken as chloroform solutions. Gas–liquid chromatography (GLC) was done on a 3% SE-30 column (2 m × 2.5 mm). The mass spectra were determined on an LKB 9000 spectrometer either by GLC (on 3% SE-30 column, 2 m × 2.5 mm) or by direct inlet (d.i.).

3α,5α-Cyclocholest-7-en-6-one (4). Finely ground crude 7-dehydrocholesteryl tosylate[27] (12g) (contaminated with cyclohydrocarbon **5a**, 3% as determined by uv and TLC) was added to a solution of potassium hydrogen carbonate (6 g), water (0.5 l.), and acetone (2 l.) at 56 ± 0.5 °C. After 5 min the solution was concentrated to 800 ml under vacuum at 30 ± 0.5 °C. The resulting mixture was cooled to −15 °C. The obtained cyclopropyl alcohol **3** (9.5 g), mp 96 °C,[4a] containing 3% of hydrocarbon **5a** (TLC), was dissolved in pyridine (10 ml) and added to a solution of chromium trioxide (9.5 g) in pyridine (90 ml) and methylene chloride (360 ml). The mixture was stirred for 1.5 h at room temperature under nitrogen. The reaction mixture was poured into ice-cold water and extracted with methylene chloride. Evaporation of the solvent yielded crude ketone **4** which was crystallized from diisopropyl ether (7.8 g): mp 131 ° C (lit.[4a] mp 129–130 °C); $[\alpha]^{21}D$ + 87°; uv λ_{max} (EtOH) 249 nm (ε 12 400); ir 1650 cm^{-1}; NMR δ 5.82 (m, C-7 H), 1.08 (s, C-10 Me), 0.66 (s, C-13 Me); mass spectrum (d.i.) *m/e* 382 (M^+), 380, 367, 365, 269, 267, 243, 229.

Anal. Calcd for $C_{27}H_{42}O$: C, 84.75; H, 11.07. Found: C, 84.90; H, 11.33.

6-Trimethylsilyloxy-3α,5α-cyclocholesta-6,8(14)-diene (5d). To a 80% dispersion (395 mg) of sodium hydride in mineral oil previously washed with anhydrous hexane and dried under nitrogen, 980 mg of ketone **4** in 15 ml of diglyme was added and the mixture was heated to 80 °C under nitrogen. After 2 hr the evolution of hydrogen ceased and the solution was cooled to room temperature. Triethylamine (10 ml) and trimethylsilyl chloride (6.5 ml) were added, and after the mixture was stirred at 25 °C for 1 h it was diluted with hexane and filtered through a pad of Celite. The filtrate was washed with a saturated NaCl solution, dried on Na_2SO_4, filtered, and evaporated in vacuo to give 950 mg of dienol silyl ether **5d**. *Crystallization from 2-propanol gave pure* **5d** *(796 mg):* mp 82 °C; $[\alpha]^{21}D$ +112°; uv λ_{max} (isooctane) 263 nm (ε 24 200); ir 1650, 1620 cm^{-1}; NMR δ 5.5 (s, C-7 H), 0.88 (s, C-13 Me), 0.79 (s, C-10 Me), 0.3–0.6 (m, cyclopropyl H); mass spectrum (GLC) *m/e* 454 (M^+), 439, 341, 336, 314, 299.

Anal. Calcd for $C_{30}H_{50}OSi$: C, 79.22; H, 11.18. Found: C, 79.23; H, 11.21.

3α,5α-Cyclocholest-8(14)-en-6α-ol (6). Trimethylsilyl dienol ether **5d** (650 mg) and $NaBH_4$ (70 mg) were dissolved in 2-propanol (60 ml). The mixture was stirred at 60 °C for 2 h. The solvent was removed under reduced pressure, ice-cold water was added, and the mixture was extracted with ether. The organic layer was washed with water and saturated NaCl solution. After drying (Na_2SO_4), the solvent was evaporated and alcohol **6** was crystallized (450 mg) from acetone: mp 107 °C; $[\alpha]^{21}D$ +72°; ir 3400, 1620 cm^{-1}; NMR δ 3.83 (q, C-6β H), 2.73 (q, C-7β H), 0.92 (s, C-10 Me), 0.86 (s, C-13 Me), 0.3–0.6 (m, cyclopropyl H); mass spectrum (d.i.) *m/e* 366 ($M^+ - H_2O$), 351, 253, 199.

Anal. Calcd for $C_{27}H_{44}O$: C, 84.31; H, 11.53. Found: C, 84.01; H, 11.28.

3β-Propionyloxy-14β-cholest-5-ene (1d). Diborane generated by addition of $NaBH_4$ (7 g) in diglyme (50 ml) to a stirred solution of boron trifluoride etherate (50 g, freshly distilled) in dry diglyme (50 ml) was bubbled under a hydrogen flow over a period of 2 h into a stirred solution of alcohol **6** (2 g) in dry diglyme (50 ml) previously heated at 55 °C. After completion of $NaBH_4$ addition, the flask containing the diborane generating mixture was heated for 1 h at 70–80 °C, the hydrogen flow being maintained to ensure the complete transfer of diborane to the hydroboration flask. The solution was then cooled to −80 °C (dry ice–acetone), propionic acid (5 ml, freshly distilled) was added carefully, and the solution was stirred for 1 h. Upon return to room temperature, more propionic acid (45 ml) was added and the solution was heated at 140 °C for 48 h at which time it was evaporated in vacuo at 70 °C. The residue was treated with $NaHCO_3$ saturated solution and extracted with petroleum ether (bp 30–50 °C); the organic layer was washed with a saturated NaCl solution, dried (Na_2SO_4), and evaporated in vacuo to give a pale yellow oil (2.58 g) which was chromatographed on silica gel G–Celite (50:50 v/v) (50 g).

Fractions eluted with hexane–benzene (90:10 v/v) (996 mg) contained a mixture of propionates **1c** and **1d** (GLC and mass spectrometry). Keeping a methanolic solution of the two isomers at −15 °C for 12 h, crystalline **1c** (410 mg) was obtained containing 3% **1d** (GLC). Recrystallization from the same solvent gave pure **1c**, mp 123–124 °C,[28] whose physical constants (GLC, ir, and mass spectrum) was identical with those of an authentic specimen.

Solvent removal from first mother liquors gave propionate **1d** (580 mg), which was purified from trace impurities by column chromatography on silica gel G–Celite (50:50 v/v). The residue from evaporation of hexane–benzene (90:10 v/v) fractions was pure **1d**, a colorless oil. The presence of **1c** was excluded by GLC (240 °C), since a single peak was present in the gas chromatogram with relative retention time (rrt) 0.86 (**1c**: rrt 1): ir 1730 cm^{-1}; NMR δ 5.62 (m, C-6 H), 4.8 (m, C-3α H), 1.0 (s, C-10 Me), 0.98 (s, C-13 Me); mass spectrum (GLC) *m/e* 368 ($M^+ - C_3H_6O_2$), 353, 260, 255, 213.

Anal. Calcd for $C_{30}H_{50}O_2$: C, 81.39; H, 11.07. Found: C, 81.62; H, 10.98.

14β-Cholest-5-en-3β-ol (1b). The ester **1d** (290 mg) was saponified in methanolic KOH and worked up as usual. The oily residue was crystallized from methanol at −20 °C. After 1 month the crystalline **1b** (240 mg) was filtered: mp 38 °C; $[\alpha]^{21}D$ +29°; ir 3310 cm^{-1}; mass spectrum *m/e* 386 (M^+), 371, 368, 301, 273, 255; GLC (240 °C) rrt 0.87 (rrt of **1a**, 1).

The acetate **1f** was a colorless, viscous oil: ir 1730 cm^{-1}; NMR δ 5.62 (m, C-6 H), 4.79 (m, C-3α H), 2.01 (s, CH_3CO), 1.02 (s, C-10 Me), 0.97 (s, C-13 Me) (calcd,[15a,d] 1.000 and 0.984, respectively); GLC (24 °C) rrt 0.86 (rrt of **1e**, 1).

The crystalline 3,5-dinitrobenzoate **1h** had mp 157 °C from acetone; $[\alpha]^{21}D$ +32°; ir 3095, 1740 cm^{-1}; NMR δ 9.13 (m, aromatic

H), 5.44 (m, C-6 H), 4.93 (m, C-3α H), 1.08 (s, C-10 Me), 1.02 (s, C-13 Me) (calcd,[15a,29] 1.086 and 1.010, respectively).

Anal. Calcd for $C_{34}H_{48}N_2O_6$: C, 70.31; H, 8.33; N, 4.82. Found: C, 70.63; H, 8.16; N, 4.77.

5α,14β-Cholestan-3β-ol (7a). A. The acetate **1f** (180 mg) was dissolved in acetic acid (20 ml) and hydrogenated over PtO_2 (20 mg) at room temperature and atmospheric pressure. After the stoichiometric hydrogen was taken up, the catalyst was removed by filtration and the filtrate was concentrated to dryness. The solution of the residue in ether was washed acid free with 5% $NaHCO_3$ solution, dried over Na_2SO_4, and evaporated. The residue was chromatographed on silica gel G–Celite (50:50 v/v). The hexane–benzene eluates (95:5 v/v) were concentrated to dryness to yield acetate **7b:** oil; ir 1730 cm^{-1}; NMR δ 4.9 (m, C-3α H), 2.03 (s, CH_3CO), 0.95 (s, C-13 Me), 0.84 (s, C-10 Me) (calcd,[15a,d] 0.950 and 0.800, respectively); GLC (240 °C) rrt 0.87 (rrt of **7c**, 1).

After saponification of **7b** with methanolic KOH, the oily alcohol **7a** was obtained, mass spectrum (GLC) *m/e* 388 (M^+).

Anal. Calcd for $C_{27}H_{48}O$: C, 83.43; H, 12.45. Found: C, 83.32; H, 12.30.

The alcohol **7a** was transformed as usual into 3,5-dinitrobenzoate **7d:** mp 155–156 °C from acetone; $[\alpha]^{21}D$ +38°; ir 3120, 1740 cm^{-1}.

Anal. Calcd for $C_{34}H_{50}N_2O_6$: C, 70.07; H, 8.65; N, 4.81. Found: C, 70.10; H, 8.79; N, 4.51.

B. Compound **8**[3] (300 mg) and $NaBH_4$ (600 mg) were dissolved in methanol (20 ml) and refluxed for 4 h. The solution was worked up as usual to yield oily **7a,** which after esterification was transformed into 3,5-dinitrobenzoate **7d** (260 mg), mp 155–156 °C.

C. The alcohol **9** (950 mg) in dyglime (50 ml) was hydroborated under the same conditions described for the preparation of propionate **1d.** After addition of propionic acid the mixture was heated at 140 °C for 48 h, at which time the solution was worked up as above. The residue was chromatographed on silica gel G–Celite (50:50 v/v).

Fractions eluted with hexane–benzene (95:5 v/v) (420 mg) contained a mixture of propionates **7e** and **7f** (GLC and GLC–mass spectrometry). Upon standing of their solution at −15 °C for 12 h and filtration of crystalline material the mother liquor was evaporated to yield propionate **7f** (250 mg) as an oil: ir 1730 cm^{-1}; mass spectrum (GLC) *m/e* 444 (M^+), 370, 355, 290, 257, 230, 217, 216, 215.

Compound **7f** was saponified to alcohol **7a,** which was transformed into 3,5-dinitrobenzoate **7d,** mp 155–156 °C.

Acknowledgments. This research was supported by the Italian Research Council. We thank Professor A. Fiecchi for helpful discussions.

Registry No.—**1b,** 57759-45-2; **1c,** 633-31-8; **1d,** 57759-46-3; **1f,** 57759-47-4; **1h,** 57759-48-5; **3,** 57679-64-8; **4,** 57674-64-3; **5d,** 57674-65-4; **6,** 57674-66-5; **7a,** 57759-49-6; **7b,** 57759-50-9; **7d,** 57759-51-0; **7e,** 57674-67-6; **7f,** 57759-52-1; **8,** 57674-68-7; **9** 566-99-4; 7-dehydrocholesteryl tosylate, 57674-69-8; trimethylsilyl chloride, 75-77-4.

References and Notes

(1) A. Fiecchi, M. Galli Kienle, A. Scala, G. Galli, R. Paoletti, and E. Grossi Paoletti, *J. Biol. Chem.*, **247,** 5898 (1972).
(2) D. J. Baisted, S. R. Klemp, and M. L. Smith, *Biochem. Biophys. Res. Commun.*, **29,** 258 (1967).
(3) M. Anastasia, A. Fiecchi, and A. Scala, *J. Chem. Soc., Perkin Trans. 1,* submitted for publication.
(4) (a) W. E. Harvey and K. Bloch, *Chem. Ind. (London),* 595 (1961); (b) D. H. R. Barton, P. G. Feakins, J. P. Poyser, and P. G. Sammes, *J. Chem. Soc. C,* 1584 (1970).
(5) (a) G. H. R. Summers, *J. Chem. Soc.* 4489 (1958); (b) M. J. Thompson, C. F. Cohen, and S. M. Lancaster, *Steroids,* **5,** 745 (1965).
(6) E. Mincione and F. Feliziani, *J. Chem. Soc., Chem. Commun.,* 942 (1973).
(7) L. Dorfman, *Chem. Rev.,* **52,** 53 (1953).
(8) H. J. E. Lowenthal, "Protective Groups in Organic Chemistry", J. F. W. McOmie, Ed., Plenum Press, New York, N.Y., 1973, p. 337.
(9) J. Hora, L. Labler, A. Kasal, V. Cerny, F. Sorm, and K. Slama, *Steroids,* **8,** 887 (1966).
(10) G. Stork and P. F. Huldrick, *J. Am. Chem. Soc.,* **90,** 4462 (1968).
(11) H. O. House, L. J. Czuba, M. Gall, and H. O. Olmstead, *J. Org. Chem.,* **34,** 2324 (1969).
(12) I. F. Eckhard, H. Heaney, and B. A. Marples, *J. Chem. Soc. C,* 2098 (1969).
(13) H. Hoellinger, N.-H. Nam, and L. Pichat, *C. R. Acad. Sci., Ser. C,* **276,** 715 (1973).
(14) V. Cerny, A. Kasal, and F. Sorm, *Collect. Czech. Chem. Commun.,* **34,** 1235 (1970).
(15) (a) N. S. Baccha and D. M. Williams, "Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry", Holden-Day, San Francisco, Calif., 1966, p 13; (b) *ibid., pp 47 and 70;* (c) *H. Mory, K. Shibata, K. Tsuneda, and M. Saway, Chem. Pharm. Bull.,* **16,** 1593 (1968); (d) R. F. Zürcher, *Helv. Chim. Acta,* **46,** 2054 (1963).
(16) D. H. R. Barton, J. P. Poser, and P. G. Sammes, *J. Chem. Soc., Perkin Trans. 1,* 53 (1972).
(17) B. Rickborn and S. E. Wood, *Chim. Ind. (Milan),* 162 (1966).
(18) H. C. Brown and K. Murray, *J. Am. Chem. Soc.,* **81,** 4108 (1959).
(19) C. W. Shoppee and G. H. R. Summers, *J. Chem. Soc.,* 3374 (1952).
(20) (a) H. C. Brown and R. L. Sharp, *J. Am. Chem. Soc.,* **90,** 2915 (1968); (b) D. J. Pasto and C. C. Cumbo, *Ibid.,* **86,** 4343 (1964).
(21) G. Galli and S. Maroni, *Steroids,* **10,** 189 (1967).
(22) H. Budzikiewicz and C. Djerassi, *J. Am. Chem. Soc.,* **84,** 1430 (1962).
(23) H. C. Brown and G. Zweifel, *J. Am. Chem. Soc.,* **88,** 1433 (1966); **89,** 561 (1967).
(24) (a) A. Stretwieser, Jr., L. Verbit, and R. Bittman, *J. Org. Chem.,* **32,** 1530 (1967); (b) J. Klein, E. Dunkelblum, and M. A. Wolff, *J. Organomet. Chem.,* **7,** 377 (1967).
(25) G. Jones and C. Djerassi, *Steroids,* **10,** 653 (1967).
(26) L. H. Toporcer, R. E. Dessy, and S. I. E. Green, *J. Am. Chem. Soc.,* **87,** 1236 (1965).
(27) P. Karrer and H. Asmis, *Helv. Chim. Acta,* **35,** 1926 (1952).
(28) A. Windaus and C. Ulbrig, *Ber. Dtsch. Chem. Ges.,* **47,** 2384 (1914).
(29) W. Arnold, W. Meister, and G. Englert, *Helv. Chim. Acta,* **57,** 1561 (1974).

Synthesis of 3-*epi*-Cholecalciferol and 5,6-*trans*-3-*epi*-Cholecalciferol

D. John Aberhart,* John Yeou-Ruoh Chu, and Adam Chi-Tung Hsu

Department of Chemistry, The Catholic University of America, Washington, D.C. 20064

Received August 27, 1975

It is now known that vitamin D_3 undergoes a two-stage metabolic process involving hydroxylation, first at C-25 (occurring in liver), then at the C-1α position (in kidney) to produce what is apparently the final biologically active form, 1α,25-dihydroxycholecalciferol (I).[1] Recent studies on synthetic ring A analogs of vitamin D_3 and its hydroxylated metabolites have provided considerable information on structure–activity relationships in such compounds. In particular, the continued presence of intestinal calcium transport ability, even in nephrectomized rats, of 25-hydroxydihydrotachysterol (**2**), 5,6-*trans*-cholecalciferol (**3a**), and 5,6-*trans*-25-hydroxycholecalciferol (**3b**), suggests that the principal requirement for such activity may be the presence of a hydroxyl group in ring A having approximately the same position, relative to the transoid diene system, as that of the 1α-hydroxyl in the normal metabolite, **1**.[1b] Further, such results suggest that the 3β-hydroxyl of **1** may be of little importance in determining the biological activity of vitamin D_3 derivatives. This is supported by the recent finding that 3-deoxy-1α-hydroxycholecalciferol exhibits high biological activity and produced a greater maximum in intestinal calcium transport than did the natural metabolite.[2a] Furthermore, 3-methoxy-1α-hydroxycholecalciferol has pronounced intestinal calcium transport activity in vitamin D deficient rats.[1b]

In order to provide further information on the relationships between structure and biological function in ring A analogues of cholecalciferol, we undertook syntheses of the analogues in which the configuration of the 3-hydroxyl group is inverted, namely 3-*epi*-cholecalciferol (**4**) and 5,6-*trans*-3-*epi*-cholecalciferol (**5**). The latter is of particular interest because it possesses a hydroxyl in the same relative position as the 1β-hydroxyl of the (as yet) unknown 3-deoxy-1β-hydroxycholecalciferol. The hydroxyl will also be similar in location to that of the biologically active 3-