

Studies on Steroids. XXXVIII.¹⁾ A New Preparation of 1 α -Hydroxycholesterol²⁾

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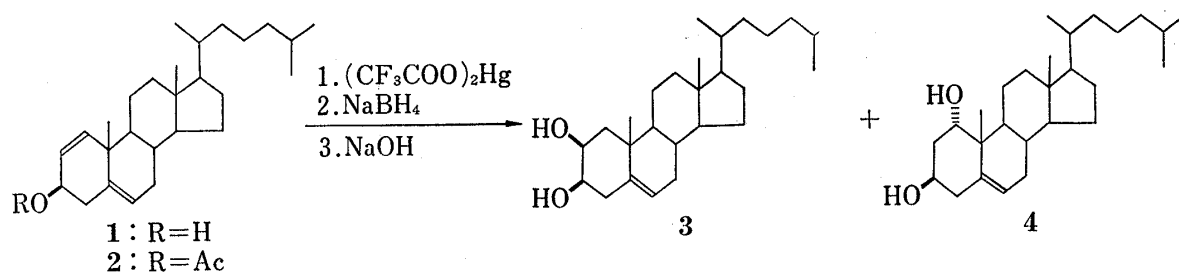
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Oxymercuration-demercuration of 3 β -acetoxycholesta-1,5-diene(2) followed by saponification gave 2 β -hydroxycholesterol (3) and 1 α -hydroxycholesterol (4) in 14% and 26% yield, respectively.

Since 1 α -hydroxyvitamin D₃ has been found⁴⁾ to elicit a comparable biological activity to 1 α ,25-dihydroxyvitamin D₃, a hormonal active form of vitamin D₃, several research groups⁵⁾ including ours have been actively exploring for synthetic route to 1 α -hydroxycholesterol (4),⁶⁾ the immediate precursor of 1 α -hydroxyvitamin D₃. Although the method of Kaneko, *et al.*,^{5d)} involves the short steps for preparation of 4 from cholesterol, the yield of 1 α -hydroxylation which was performed by hydroboration-oxidation reaction of cholesta-1,5-dien-3 β -ol (1), is low (10—15%). We have considered that oxymercuration-demercuration reaction⁷⁾ of the dienol 1 or its derivatives would be an alternative for the introduction of 1 α -hydroxyl function.

The dienol 1 prepared by the known 4 steps procedures from cholesterol,^{8,5d)} was converted to the acetate 2 in the usual manner. Reaction of the acetate 2 with mercuric acetate in tetrahydrofuran followed by reduction with alkaline sodium borohydride gave only the starting dienol 1. However, when mercuric trifluoroacetate was used⁷⁾ in place of mercuric acetate, there were obtained after saponification, the less polar diol 3 and the more polar diol 4 in yields of 14% and 26%, respectively, together with the recovered 1 (9%).²⁾ The trimethylsilyl derivative of the minor product 3 showed a mass ion peak at *m/e* 546, corresponding to the molecular ion of bis-trimethylsilyl ether of cholestenediol. In accordance with this, the nuclear magnetic resonance (NMR) spectrum of 3 indicated besides 3 α -H signal at 3.55 ppm, a one proton multiplet at 4.03 ppm, which may be assigned to a hydrogen attached to secondary hydroxy group. This hydroxyl should be located at C-2 position as deduced from a positive periodate oxidation test. All these facts and the marked difference of its physical data from 2 α -hydroxycholesterol,^{5d)} as well as the good agreement of melting point with the published 2 β -hydroxycholesterol,⁹⁾ we have concluded that this diol is 2 β -hydroxycholesterol (3). The major product of the oxymercuration-demercuration reaction was identified as 1 α -hydroxycholesterol by

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Chart

direct comparison (mp, thin-layer chromatography (TLC), gas-liquid chromatography (GLC), and NMR) with an authentic sample.^{5a)}

The yield of 1 α -hydroxylation of the present method appears to be better than that of hydroboration-oxidation reaction.^{5d)} The applicability of our procedures has recently been exemplified by synthesis of 1 α ,24-dihydroxyvitamin D₃.¹⁰⁾

Experimental

Melting points were determined on a hot stage microscope and are uncorrected. NMR spectra were run on a Varian T-60 spectrometer with deuteriochloroform as solvent and with tetramethylsilane as an internal standard. Mass spectra were determined with Shimadzu-LKB 9000S. Column chromatography was effected with Wakogel C-200. Abbreviations used for NMR data: s, singlet; d, doublet; m, multiplet.

3 β -Acetoxycholesta-1,5-diene (2)—Cholesta-1,5-dien-3 β -ol(1)^{5d)} (2.7 g) was heated in a mixture of acetic anhydride (7 ml), pyridine (7 ml) and benzene (15 ml) on a boiling water-bath for 2.5 hr. The reaction mixture was poured into ice-water and the aqueous layer was extracted with benzene. The combined benzene layer was washed with dil HCl and saturated NaHCO₃. Drying over K₂CO₃ and evaporation of the solvent gave a colorless residue, which was crystallized from methanol to give the acetate 2 (2.75 g) mp 68–70° (from ether-methanol), NMR δ 0.67 (3H, s, 13-Me), 1.09 (3H, s, 10-Me), 2.05 (3H, s, acetyl), 5.2 (1H, m, 3 α -H), 5.4 (1H, m, 6-H), 5.4 and 5.8 (2H, a pair of d, J =10 Hz, 1 and 2-H). *Anal.* Calcd. for C₂₉H₄₆O₂: C, 81.63; H, 10.87. Found: C, 81.54; H, 10.87.

Oxymercuration-demercuration of the Acetate (2)—To a stirred mixture of the acetate 2 (170 mg), tetrahydrofuran (2.0 ml) and water (0.54 ml), was added mercuric oxide (476 mg) and trifluoroacetic acid (0.34 ml) under cooling with an ice-bath. Stirring was continued at 3° for 8 hr and then at 15° for 16 hr. Two ml of 3 M NaOH and a solution of NaBH₄ (60 mg) in 3 M NaOH (2 ml) were added to the reaction mixture. After stirring 30 min, the mixture was extracted with ethyl acetate, washed with brine, 2 N HCl, sat. NaHCO₃ and brine. Evaporation of the solvent gave a colorless crystal, which was heated with a mixture of 3 N NaOH (1 ml) and methanol (10 ml) at 70° for 15 min. The mixture was partitioned between ethyl acetate and brine. The ethyl acetate layer was dried over MgSO₄ and evaporated to dryness. The resulting yellow amorphous materials were applied on a column of silica gel (3 g). Elution with benzene-ether (50:1) gave the recovered 1 (13 mg). Further elution with benzene-ether (10:1) afforded 2 β -hydroxycholesterol (3) (22 mg), mp 222–224° (from acetone) (ref.⁸⁾ mp 224–226°, NMR δ 0.68 (3H, s, 13-Me), 1.18 (3H, s, 10-Me), 3.55 (1H, m, 3 α -H), 4.03 (1H, m, 2 α -H) and 5.40 (1H, m, 6-H). Treatment of 3 with trimethylsilylimidazole gave the bis-trimethylsilyl ether, m/e 546 (M⁺), 531 (M-Me), 456 (M-TMSOH), 441 (M-TMSOH-Me), and 366 (M-2TMSOH). Continued elution on the above chromatography with benzene-ether (5:1) gave 1 α -hydroxycholesterol (4) (39 mg), which was identified with an authentic sample^{5a)} in mp, TLC, GLC, and NMR.

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