Synthesis of 1α-Hydroxycorticosterone

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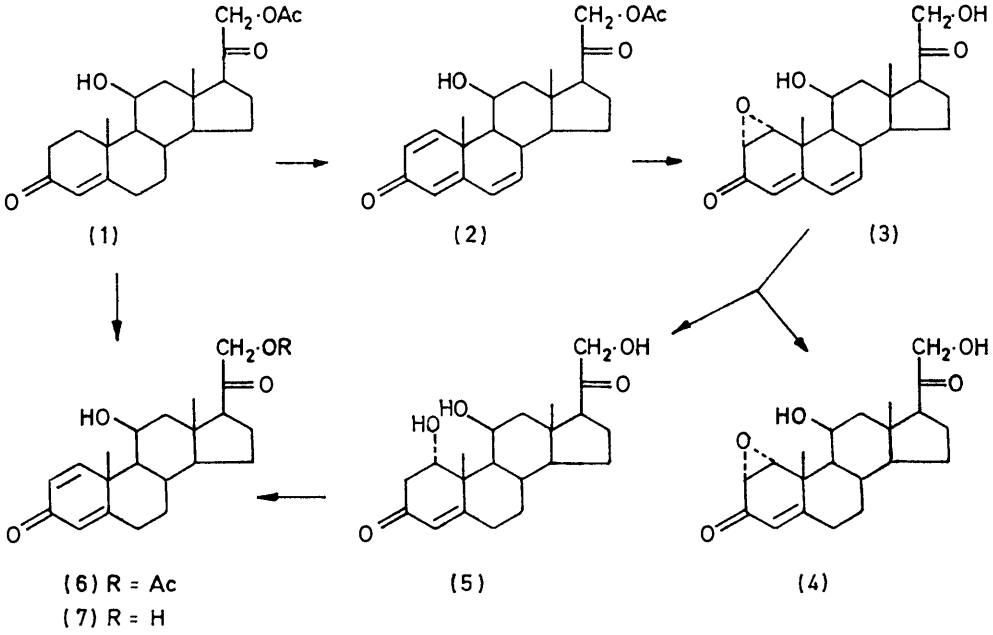
1α-Hydroxycorticosterone (5), previously isolated from the skate (genus *Raja*), has been prepared from corticosterone 21-acetate *via* its 1,2,6,7-tetradehydro- and 1α,2α-epoxy-6,7-didehydro-derivatives. The configuration at C-1 has been established by polarimetry.

THE major corticosteroid hormone of the skate (genus *Raja*) has been identified as 1α-hydroxycorticosterone [1α,11β,21-trihydroxypregn-4-ene-3,20-dione (5)] on the basis [1] of (i) its ready conversion (*via* the diacetate) into the known 1,2-didehydrocorticosterone [11β,21-dihydroxypregna-1,4-diene-3,20-dione (7)] and (ii) a comparison of its n.m.r. spectrum and chromatographic mobility with those of a number of related compounds. Our objectives were to confirm this assignment by synthesis, to characterise the compound more rigorously, and to make it more readily available for biological experimentation. Our synthesis essentially follows that of 1α-hydroxycholest-4-en-3-one [2] (the only reported example of a chemical synthesis of a steroidal 1α-hydroxy-4-en-3-one) although the product was obtained in an impure state and its structure not rigorously established. Microbiological 1α-hydroxylation of steroidal 4-en-3-ones of

[1] D. R. Idler and B. Truscott, *Steroids*, 1967, **9**, 457.
[2] B. Pelc and E. Kodicek, *J. Chem. Soc.* (*C*), 1971, 1568.

the oestrane and androstane series has been reported [3] but its application to compounds of the pregnane series was unsuccessful.[4]

Corticosterone 21-acetate (1) was dehydrogenated with chloranil [5] to 1,2,6,7-tetradehydrocorticosterone acetate (21-acetoxy-11β-hydroxypregna-1,4,6-triene-3,20-dione) (2). Treatment of the triene (2) with alkaline hydrogen peroxide [2] gave 1α,2α-epoxy-11β,21-dihydroxypregna-4,6-diene-3,20-dione (3), which showed peaks characteristic of a 1,2-epoxy-4-en-3-one system in the τ 6 region of its n.m.r. spectrum.[6,7] Hydrogenation of the epoxide (3) in pyridine over palladium–calcium carbonate [2] gave 1α-hydroxycorticosterone (5) and 1α,2α-epoxy-11β,21-dihydroxypregn-4-ene-3,20-dione (4). The epoxy-group



of the latter resisted reduction under the conditions employed.

The 1α,2α-configuration of the epoxides (3) and (4) was confirmed by c.d. measurements. The prediction of the Octant Rule [8] that the Cotton effect due to the $n\rightarrow\pi^*$ transition of an αβ-unsaturated oxo-group should be strongly positive for the 1α,2α-epoxide and strongly negative for the 1β,2β-epoxide has been confirmed for model compounds.[7] The strongly positive Cotton curves observed for the epoxides (3) ($\Delta\varepsilon_{334}$ + 2.63) and (4) ($\Delta\varepsilon_{327}$ + 2.57) indicate a 1α,2α-orientation for both compounds. Consequently the reductive opening of the epoxide ring of (3) would be expected to give the α-oriented 1-alcohol (5). This was confirmed by molecular rotation measurements. The low negative ΔM_D value (−14°) due to the 1-hydroxy-group indicates its α-configuration (reported ΔM_D for 1α-OH −14 to −26°; for 1β-OH −116 to −218°).[9]

The i.r., u.v., and n.m.r. spectra of the synthetic 1α-hydroxycorticosterone agreed with the data published [1] for the natural product. The small quantity of the natural product available was sufficient to determine its mass spectrum and its mobilities in three chromatographic systems. These were identical with those of the synthetic material. Both the synthetic and the natural product were readily dehydrated with dilute acid to give the 1,4-diene (7), which was also prepared by dehydrogenation [10] of corticosterone acetate (1) with selenium dioxide. The properties of this diene agreed with those reported for the compound prepared by microbiological dehydrogenation of corticosterone.[11-13]

EXPERIMENTAL

M.p.s were determined with a Kofler hot-stage apparatus. Preparative t.l.c. was performed on glass plates 20 cm square

[3] R. M. Dodson, A. H. Goldkamp, and R. D. Muir, *J. Amer. Chem. Soc.*, 1957, **79**, 3921; 1960, **82**, 4026; G. Ambrus, E. Szarka, I. Barta, Gy Horvath, L. Radics, and M. Kajtar, *Steroids*, 1975, **25**, 99.

[4] R. C. Tweit, R. M. Dodson, and R. D. Muir, *J. Org. Chem.*, 1962, **27**, 3654.

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[8] G. Snatzke, *Tetrahedron*, 1965, **21**, 413, 421.

[9] L. L. Smith, *Steroids*, 1963, **1**, 570; G. Greenspan, R. W. A. Rees, G. D. Link, C. P. Boyd, R. C. Jones, and H. E. Alburn, *Experientia*, 1974, **30**, 328; L. L. Smith, G. Greenspan, R. Rees, and T. Foell, *J. Amer. Chem. Soc.*, 1966, **88**, 3120; R. M. Dodson, S. Kraychy, R. T. Nicholson, and S. Mizuba, *J. Org. Chem.*, 1962, **27**, 3159.

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[13] H. L. Herzog, C. C. Payne, M. Tully Hughes, M. Jevnik-Gentles, E. B. Hershberg, A. Nobile, W. Charney, C. Federbush, D. Sutter, and P. L. Perlman, *Tetrahedron*, 1962, **18**, 581.

with a layer of silica gel GF_{254} (Merck) 1 mm thick. I.r. spectra were measured for dispersions in KBr with a Unicam SP 200 spectrophotometer, u.v. spectra for solutions in ethanol with a Unicam SP 8000 spectrophotometer, and n.m.r. spectra for solutions in [^{2}H]chloroform with a Varian HA 100 spectrometer. Optical rotations were measured for solutions in chloroform at 20—25 °C and c.d. data were obtained with a Thorn–Bendix Polarmatic 62 spectropolarimeter for solutions in ethanol. Mass spectra were obtained with an A.E.I. MS12 spectrometer.

21-Acetoxy-11β-hydroxypregna-1,4,6-triene-3,20-dione (2).—Corticosterone 21-acetate (1) (5 g) and chloranil (19 g) in pentan-2-ol (250 ml) were refluxed for 3 h. The cooled solution was filtered to remove the excess of reagent and the filtrate taken to dryness. The residue was extracted with chloroform (400 ml); the extract was filtered, washed with water (3 × 40 ml), M-sodium hydroxide solution (3 × 40 ml), and water (4 × 40 ml), then dried (Na_2SO_4) and evaporated. The crude product (4.3 g) was purified by column chromatography on aluminium oxide (150 g; 100—150 mesh, Neutral Activity I). Elution, in 25 ml fractions, with hexane–chloroform (1 : 1; 500 ml) and hexane–chloroform (2 : 3; 500 ml) gave, in fractions 24—36, a product (1.9 g) which on crystallisation from acetone–hexane afforded crystals of the *acetate* (2) (1.10 g), m.p. 169°, $[\alpha]_D$ +194°, $\nu_{max.}$ 3 500, 1 750, 1 720, 1 650, and 1 600 cm^{-1}, $\lambda_{max.}$ 223, 256, and 299 nm (ε 15 800, 13 000, and 16 200), τ 2.69 (d, *J* 10 Hz, 1 H), 3.66—4.04 (m, 2-,4-,6-,and 7-H), 5.39 (q, 21-H), 5.51 (q, 11α-H), 7.86 (s, OAc), 8.56 (10-Me), and 8.99 (13-Me). (Found: C, 72.0; H, 7.5. $C_{23}H_{28}O_5$ requires C, 71.85; H, 7.3%). More pure acetate (2) (424 mg) was obtained from the mother liquors and impure column fractions 20—23 and 37—57 by t.l.c. in chloroform–methanol (98 : 2).

1α,2α-Epoxy-11β,21-dihydroxypregna-4,6-diene-3,20-dione (3).—The acetate (2) (500 mg) in methanol (15 ml) was treated with 10M-sodium hydroxide (0.3 ml) and 30% hydrogen peroxide (1.5 ml) for 3 h at room temperature. Sodium sulphite (1 g) was added, followed by water (100 ml), and the product was extracted twice with ethyl acetate (100 ml). The organic phase was washed twice with water, dried, and evaporated to give the crude epoxide (163 mg). Acidification of the aqueous phases and extraction with chloroform gave a mixture of unidentified by-products (250 mg). Further purification of the crude epoxide (3) by t.l.c. in chloroform–methanol (98 : 2) gave a product (119 mg) which on crystallisation from acetone–hexane afforded fine *needles* (98 mg) m.p. 237—239°, $[\alpha]_D$ +390°, $\nu_{max.}$ 3 510, 1 700, 1 650, and 1 615 cm^{-1}, $\lambda_{max.}$ 290 nm (ε 20 460), τ 3.87 (s, 6- and 7-H), 4.41 (d, $J_{2,4}$ 2 Hz, 4-H), 5.33 (q, 11α-H), 5.79 (s, 21-H), 6.12 (d, $J_{1,2}$ 4 Hz, 1β-H), 6.57 (q, $J_{2,4}$ 2, $J_{2,1}$ 4 Hz, 2β-H), 8.54 (10-Me), 8.98 (13-Me), c.d. $\Delta\varepsilon_{334}$ + 2.63pk, $\Delta\varepsilon_{278}$ 0tr, $\Delta\varepsilon_{250}$ + 0.24br,pk (Found: C, 70.0; H, 7.7. $C_{21}H_{26}O_5$ requires C, 70.4; H, 7.3%).

1α-Hydroxycorticosterone (5).—The epoxide (3) (68 mg) and 5% palladium–calcium carbonate (70 mg) in pyridine (4 ml) were hydrogenated overnight. The catalyst was removed by filtration through a small Celite column and the pyridine removed in a stream of air. T.l.c. in chloroform–methanol (9 : 1) gave two main products. The more polar compound (13 mg) was crystallised from acetone–hexane to give *1α-hydroxycorticosterone* (5) (8 mg), m.p. 201—202° (lit.,[1] 201—202°), $[\alpha]_D$ +159.5°, M_D + 577° (corticosterone M_D + 591°), $\nu_{max.}$ 3 500, 1 700, and 1 650 cm^{-1}, $\lambda_{max.}$ 242 nm (ε 11 255), *m/e* 362 (M^+, trace), 344 (24%), 331 (100), 313 (14), 285 (46), 267 (96), 221 (58), 173 (28), 161 (30), 147 (41), and 135 (29), τ (C_5D_5N) 4.13 (4-H), 5.13 (11α-H), 5.43 (1β-H), 5.67 (21-H), 8.40 (10-Me), and 8.86 (13-Me), τ ($CDCl_3$) 4.27 (4-H), 5.47 (11α-H), 5.69 (1β-H), 5.83 (21-H), 8.56 (10-Me), and 9.08 (13-Me) (Found: C, 69.9; H, 8.6. $C_{21}H_{30}O_5$ requires C, 69.6; H, 8.4%).

The synthetic compound (5) had chromatographic properties identical with those of the natural product [1] in the t.l.c. systems chloroform–methanol (9 : 1), and ethyl acetate–ethanol (95 : 5) and in the Bush B_5 paper system benzene–methanol–water (2 : 1 : 1). The mass spectra of the two compounds were identical.

The less polar compound (37 mg) isolated from t.l.c. was crystallised from acetone–hexane to give *1α,2α-epoxy-11β,21-dihydroxypregn-4-ene-3,20-dione* (4) (30 mg), m.p. 208—211°, $[\alpha]_D$ +241°, $\nu_{max.}$ 3 500, 1 705, and 1 665 cm^{-1}, $\lambda_{max.}$ 247 nm (ε 9 250), τ 4.37 (4-H), 5.29 (11α-H), 5.91 (21-H), 6.19 (d, $J_{1,2}$ 4 Hz, 1β-H), 6.59 (q, $J_{1,2}$ 4, $J_{2,4}$ 2 Hz, 2β-H), 8.43 (10-Me), and 9.00 (13-Me), c.d. $\Delta\varepsilon_{327}$ + 2.57pk, $\Delta\varepsilon_{305}$ + 2.28tr, $\Delta\varepsilon_{286}$ + 2.62pk (Found: C ,70.0; H, 7.8. $C_{21}H_{28}O_5$ requires C, 70.0; H, 7.8%). This epoxide could not be further hydrogenated to 1α-hydroxycorticosterone.

C.d. data for corticosterone are: $\Delta\varepsilon_{324}$ −0.79tr, $\Delta\varepsilon_{312}$ 0, $\Delta\varepsilon_{282}$ + 2.68pk.

21-Acetoxy-11β-hydroxypregna-1,4-diene-3,20-dione (6).—Corticosterone acetate (1) (1.8 g) and selenium dioxide (0.9 g) in 2-methylpentan-2-ol (100 ml) were refluxed for 2 days under nitrogen. The cooled mixture was filtered through Celite and the filtrate evaporated. The residue was taken up in chloroform (60 ml), washed with saturated sodium chloride solution, dried (Na_2SO_4), and then evaporated. The residue was chromatographed on a silica gel column (100 g; 60—120 mesh). Elution with chloroform (200 ml) gave an unidentified non-polar fraction (221 mg). Elution with chloroform–acetone (99 : 1; 300 ml) gave impure acetate (6) (1.43 g), and further elution with chloroform–acetone (98 : 2; 500 ml) yielded an almost pure product (393 mg) which after two crystallisations from acetone–hexane afforded the acetate (6) (232 mg), m.p. 158—160° (lit.,[11] 159—161°), $[\alpha]_D$ +135° (lit.,[11] +151° in dioxan), $\nu_{max.}$ 1 710, 1 660, 1 610, and 1 600 cm^{-1}, $\lambda_{max.}$ 242 nm (ε 15 600) (Found: C, 71.1; H, 7.6. Calc. for $C_{23}H_{30}O_5$: C, 71.5; H, 7.8%).

11β,21-Dihydroxypregna-1,4-diene-3,20-dione (7).—The acetate (6) (20 mg) in aqueous 1.5% potassium hydrogen carbonate-ethanol (3 : 7; 20 ml) was refluxed for 1 h; the solution was then cooled and concentrated under reduced pressure, and the product extracted with ethyl acetate. The crude product (18 mg) after crystallisation from acetone–hexane gave the alcohol (7) (15 mg), m.p. 217—219° (lit.,[11] 216—220°; lit.,[12] 227.5—230.5°), $[\alpha]_D$ +157° (in ethanol) (lit.,[11] +158° in ethanol), $\nu_{max.}$ 3 500, 1 710, 1 660, 1 620, and 1 600 cm^{-1}, $\lambda_{max.}$ 242 nm (ε 13 800), *m/e* 344 (M^+, 52%), 326 (10), 313 (56), 267(78), 223(100), 173(74), 159(69), and 147 (95) (Found: C, 72.9; H, 8.2. Calc. for $C_{21}H_{28}O_4$: C, 73.2; H, 8.2%).

Dehydration of 1α-Hydroxycorticosterone.—1α-Hydroxycorticosterone (5) (1 mg) in ethanol (0.5 ml) was treated with 2M-hydrochloric acid (0.2 ml) for 3 h at 58 °C. The acid was neutralised with saturated sodium hydrogen carbonate solution and the product extracted with chloroform (3 ml). T.l.c. of the product in chloroform–methanol (98 : 2) and in 2,2,4-trimethylpentane–2-methylpropan-2-ol (1 : 1) and paper chromatography in the Bush systems benzene–methanol–water (2 : 1 : 1) and toluene–methanol–water (10 : 7 : 3) showed only one spot, R_F identical with that of 1,2-didehydrocorticosterone (7). The mass spectrum of the

dehydration product was identical with that of (7). A sample of Idler's 1α-hydroxycorticosterone [1] gave identical results.

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