18-Substituted Steroids. Part 10.[1] Synthesis of 16α,18,21-Trihydroxypregn-4-ene-3,20-dione (16α,18-Dihydroxy-11-deoxycorticosterone)

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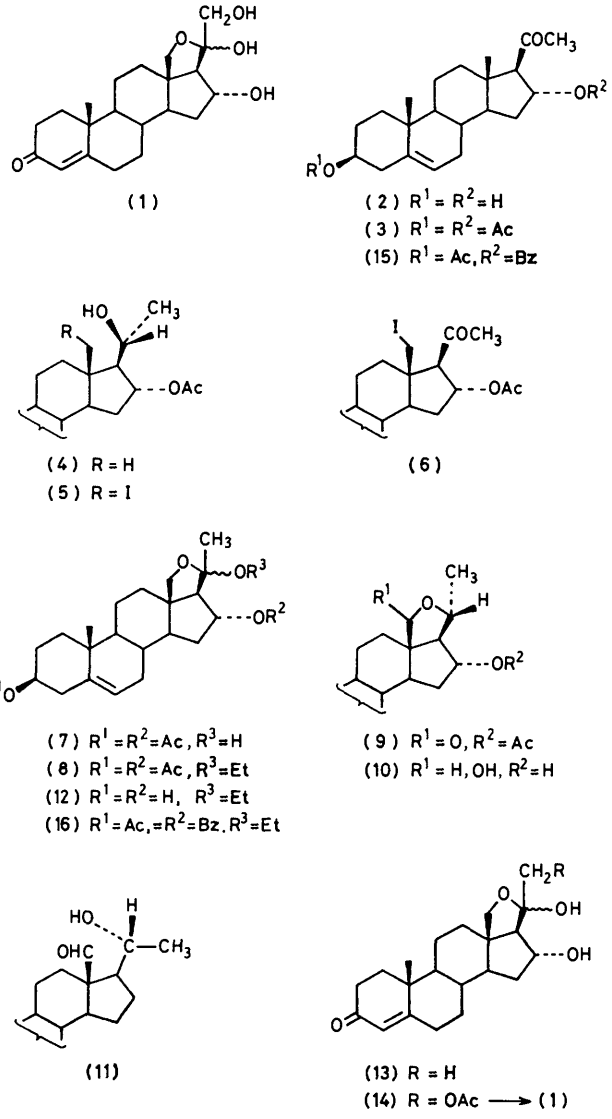
The title compound has been prepared from 3β-acetoxy-16α-hydroxypregn-5-en-20-one by two routes, each involving application of the ' hypoiodite ' reaction sequence [$Pb(OAc)_4$–I_2–*hν*; oxidation; solvolysis] to a 16α-acyloxypregnan-20β-ol derivative, to obtain the corresponding 18-hydroxypregnan-20-one (as 18,20-hemiacetal), with subsequent acetoxylation at C-21, and selective oxidation at C-3.

An abnormality in steroid biosynthesis, which may be associated with ' low renin ' hypertension,[2] leads to increased production by the adrenal cortex of a compound identified[3] as 16α,18,21-trihydroxypregn-4-ene-3,20-dione (16α,18-dihydroxy-11-deoxycorticosterone; 16α,18-dihydroxy-DOC), which exists in the 18,20-hemiacetal form (1). The biological role of 16α,18-dihydroxy-DOC is as yet poorly understood: it appears to have no direct effect on sodium levels in adrenolectomised rats, but is reported to enhance the sodium-retaining effect of sub-threshold doses of aldosterone.[3,4]

Although 16α,18-dihydroxy-DOC (1) has been produced on a milligram scale by microbial hydroxylation of 18-hydroxy-DOC,[4] no chemical synthesis has yet been reported. We now describe a synthesis of 16α,18-dihydroxy-DOC (Scheme 1) which made use of our recently improved method for the preparation of 3β,16α-dihydroxypregn-5-en-20-one (2).[5] The derived 3,16-diacetate (3) was carefully reduced at C-20 with borohydride to afford the pregnan-20β-ol (4) without loss of acetate groups. Application of the ' hypoiodite ' sequence of reactions[6,7] [photolysis in the presence of $Pb(OAc)_4$–I_2, followed by oxidation and Ag^+-assisted solvolysis] gave either the 18,20-hemiacetal (7) or the corresponding 20-ethoxy-derivative (8), depending upon the choice of either an aqueous solvent or ethanol, respectively, for the solvolytic step. In contrast to our experience of ' hypoiodite ' reactions in those pregnan-20β-ols which have no substitution at C-16, where a steroid–I_2–$Pb(OAc)_4$ molar ratio of 1 : 0.56 : 2.44 was found most satisfactory,[8-10] it was necessary in the presence of a 16α-acetoxy-group to halve the proportion of lead tetra-acetate in order to optimise the yield of the required hemiacetal. Larger amounts of lead tetra-acetate caused over-oxidation in the photolysis step, giving the 18,20-lactone (9) in dominant proportion.[11]

As recommended by Choay and his co-workers,[12] yields of the desired hemiacetal could be improved by isolating the intermediate 18-iodo-20β-alcohol (5) by crystallisation, in preference to carrying the crude photolysis product through subsequent stages before purification of the hemiacetal or its 20-ethoxy-derivative (see Experimental section). The formation of by-products[13,14] during the Ag^+-assisted solvolysis of the iodo-ketone (6) was minimised by bringing the reaction mixture *rapidly* to the required temperature.[14]

When the lactone (9) was found to be a significant part of the product from the hypoiodite reaction, an attempt was made to convert it into the required hemiacetal (7). The lactone was easily reduced by di-isobutylaluminium hydride (DIBAL)[15-17] to the corresponding lactol (10),



Scheme 1

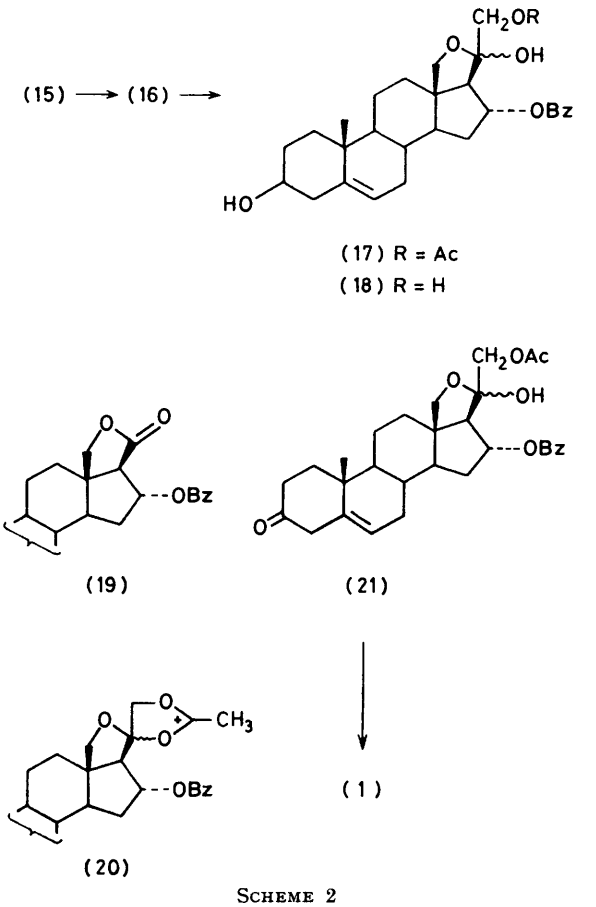
which is the hemiacetal form of the 20β-hydroxy-18-aldehyde (11). Intramolecular hydride transfer has been shown to occur in comparable structures under Oppenauer/Meerwein–Pondorff conditions,[18-20] and we accordingly treated the lactol with aluminium t-butoxide in refluxing toluene, conditions which we had found to effect at least a partial isomerisation of a 16-deoxy-analogue of the lactol (10) into its corresponding 18,20-hemiacetal.[20] The 16α-hydroxy-lactol (10), however, did not isomerise so that approach was abandoned.

The main reaction sequence (Scheme 1) required the protection of the 18,20-hemiacetal (7) as a derivative which would allow (*a*) selective oxidation at C-3, to generate the 4-en-3-one, and (*b*) acetoxylation at C-21. The 20-ethoxy-derivative (8) of the hemiacetal was a convenient protected form for use during the alkaline hydrolysis of the 3β- and 16α-acetoxy-groups, but unfortunately failed to provide sufficient steric hindrance in the vicinity of ring *D* to permit a selective hydrolysis at C-3 while retaining the 16-acetate. Since the common oxidants based upon Cr^{VI} are unsuitable for a selective oxidation of the 3β,16α-diol (12) only at C-3, the more sterically-demanding Oppenauer method was used, with *N*-methyl-4-piperidone[21] as hydride acceptor. Selective oxidation at C-3 of another pregnane-3,16α-diol has been reported,[22] although the yield was only 35%. The oxidised product in the present case was a complex mixture (t.l.c.), containing several products of relatively low polarity as well as the required 16α,18-dihydroxy-pregn-4-ene-3,20-dione (16α,18-dihydroxyprogesterone) in the 18,20-hemiacetal form (13), which was obtained in 20% yield by direct crystallisation. It seems possible that products of lower polarity could have resulted from the 16α-hydroxy-group having suffered elimination, possibly associated with instability of the masked 20-oxo-function to Oppenauer conditions. The formation of 16-oxo-derivatives and of dimeric products[14,23] are also possibilities. No attempt was made to separate or identify the numerous by-products. The structure of the required compound (13) followed from its spectral characteristics (see Experimental section).

Disappointing results were obtained from attempts to carry out the oxidation at C-3 on solid alumina,[24,25] with either trichloroacetaldehyde or cyclohexanone[26] as hydride acceptor, although with the latter reagent t.l.c. evidence was obtained for the formation of a little of the required 4-en-3-one (13) among a large number of products.

Acetoxylation of 16α,18-dihydroxyprogesterone with lead tetra-acetate in acetic acid[8] gave the 21-acetoxy-derivative, which comprised a 5 : 4 mixture of the 18,20-hemiacetals (14) isomeric at C-20, from n.m.r. evidence. Careful hydrolysis with methanolic potassium hydrogen carbonate then afforded 16α,18-dihydroxy-DOC (1).

The least satisfactory stage in the foregoing synthesis was the selective Oppenauer oxidation of the 3β,16α-diol (12) to obtain 16α,18-dihydroxyprogesterone (13). In an attempt to overcome this problem, a modified reaction sequence (Scheme 2) was explored, in which the 16α-hydroxy-group was protected at an early stage as its benzoate (15). Conversion into the corresponding acetal (16) followed a route parallel to that described above for the 3,16-diacetate. The presence of the less reactive 16-benzoate permitted selective alkaline hydrolysis of the 3-acetate. The 21-acetoxylation step [$Pb(OAc)_4$–HOAc] was carried out at this point to give 21-acetoxy-16α-benzoyloxy-3β,18-dihydroxypregn-5-en-20-one, 18,20-hemiacetal (17), although in unusually low



SCHEME 2

yield (43%). The product (17) was a 1 : 1 mixture of isomers at C-20 (n.m.r.). The corresponding 3,21-diol (18) and the 21-nor-16α-benzoyloxy-lactone (19) were obtained as by-products. Loss of C-21 has been reported previously[27] as a consequence of prolonged contact with the 21-acetoxylation reagents. Both the 3,21-diol (18) and the 21-nor-lactone (19) are thought to arise *via* sequences of reactions which include intramolecular attack of the 21-acetoxy-group on a C-20 cationic centre to form a 20,21-acetoxonium ion (20).

Careful oxidation[28] of the 3β-hydroxy derivative (17) with Jones' reagent afforded the corresponding 5-en-3-one (21). Alkaline hydrolysis, with concomitant $\Delta^5 \longrightarrow \Delta^4$ isomerisation, then gave 16α,18-dihydroxy-DOC (1), although again only in low yield. It was found necessary to use potassium hydroxide to remove the 16α-*O*-benzoyl group, and the complexity (t.l.c.) of the

hydrolysis product indicated that the 16α-hydroxy-hemiacetal system is somewhat intolerant of such strongly alkaline conditions.

EXPERIMENTAL

M.p.s were determined with a Reichert microscope. Unless otherwise stated i.r. spectra refer to KBr discs. N.m.r. spectra (δ values) were determined at 100 MHz for solutions in $CDCl_3$ unless otherwise indicated, with Me_4Si as internal standard. For t.l.c. analyses Merck Kieselgel 60 $PF_{254+366}$/Kieselgel G type 60 (1 : 1) was used. 'Light petroleum' refers to the fraction of b.p. 60—80 °C.

Pregn-5-ene-3β,16α,20β-triol 3,16-*Diacetate* (4).—A solution of 3β,16α-diacetoxypregn-5-en-20-one (3) (26.5 g) in tetrahydrofuran (400 ml) and methanol (400 ml) was cooled to 0—5 °C, then sodium borohydride (15.5 g) was added and the mixture was stirred at the same temperature for 2 h. T.l.c. then showed complete disappearance of starting material. The excess of sodium borohydride was destroyed with acetone, and the solution was concentrated under reduced pressure, then poured into water. The washed and dried precipitate (25.6 g) was dissolved in benzene and chromatographed on silica gel. Elution with benzene containing 5% ether gave the triol diacetate (4) (15.4 g, 60%); m.p. 186—187° (from benzene) (lit.,[29] 184—186°); $\nu_{max.}$ 3 440, 1 735, 1 705, 1 240, and 1 035 cm^{-1}; $\nu_{max.}$ (2% w/v in CCl_4; 0.5 mm path) 3 620, 3 040, 1 735, 1 240, and 1 035 cm^{-1}; δ 0.82 (s, 18-H_3), 1.03 (s, 19-H_3), 1.16 (d, *J* 6.5 Hz, 21-H_3), 2.02 (6 H, s, 3-OAc and 16-OAc), 2.33 (m, 4-H_2), 3.90 (m, 20α-H), 4.40—5.00 (complex, 3α-H and 16β-H), and 5.36 (m, 6-H) (Found: C, 71.6; H, 9.2. Calc. for $C_{25}H_{38}O_5$: C, 71.7; H, 9.2%).

Elution with ether gave crude pregn-5-ene-3β,16α,20β-triol 16-acetate (8%); $\nu_{max.}$ 3 420, 1 735, 1 250, 1 050, and 1 020 cm^{-1}; δ 0.82 (s, 18-H_3), 1.02 (s, 19-H_3), 1.16 (d, *J* 6.5 Hz, 21-H_3), 2.02 (s, 16-OAc), 3.54 (m, 3α-H), 3.91 (m, 20α-H), 4.85 (m, 16β-H), and 5.35 (m, 6-H).

18-*Iodopregn-5-ene-3β,16α,20β-triol* 3,16-*Diacetate* (5).—A solution of the triol diacetate (4) (4 g) in boiling cyclohexane (1 000 ml) was treated with lead tetra-acetate (5.164 g); iodine (1.334 g) was added and the mixture was stirred under reflux while being irradiated with two tungsten lamps (each 500 W) for 2.5 h. After cooling to room temperature, the mixture was filtered through a bed of Celite 535, and the filter cake was washed with cyclohexane. The filtrate was washed with aqueous 5% sodium thiosulphate and water, dried over anhydrous potassium carbonate, and, after the addition of a few drops of pyridine, was evaporated under reduced pressure at 40 °C. The residue was dissolved in acetone, which was then removed under reduced pressure. This operation was repeated to remove traces of cyclohexane. Crystallisation from acetone gave the *iodo-derivative* (5) (1.665 g, 32%), m.p. 156—158°; $\nu_{max.}$ 3 510, 1 730—1 720 (split carbonyl absorption, as a result of hydrogen bonding in the crystal lattice), 1 250 and 1 040 cm^{-1}; δ 1.03 (s, 19-H_3), 1.17 (d, *J* 6.5 Hz, 21-H_3), 2.02 (6 H, s, 3-OAc and 16-OAc), 3.06 and 3.40 (d,d, *J* 11 Hz, 18-H_2), 4.22 (m, 20α-H), 4.64 (m, 3α-H), 4.8 (m, 16β-H), and 5.37 (m, 6-H) (Found: C, 55.1; H, 6.9; I, 23.3. $C_{25}H_{37}IO_5$ requires C, 55.2; H, 6.9; I, 23.3%).

3β,16α-*Diacetoxy*-18-*iodopregn-5-en*-20-*one* (6).—A solution of the 18-iodo-alcohol (5) (1.3 g) in acetone (40 ml) was cooled to 0—5 °C, in an ice-bath, then Jones' reagent (*ca.* 0.8 ml) was added slowly with vigorous stirring until the solution acquired a permanent brown colour. The solution was further stirred for 30 min, then aqueous 5% sodium acetate was added and the mixture was extracted twice with benzene. The organic layer was washed with water, dried over anhydrous potassium carbonate, and, after the addition of a few drops of pyridine, evaporated under reduced pressure at 40 °C. The residue crystallised from acetone–light petroleum, to yield the 18-*iodo*-20-*ketone* (6) (1.174 g, 91%); m.p. 120—121°; $\nu_{max.}$ 1 740, 1 730, 1 720, 1 235, and 1 025 cm^{-1}; δ 1.02 (s, 19-H_3), 1.99 (s, 16-OAc), 2.03 (s, 3-OAc), 2.31 (s, 21-H_3), 2.67 (d, *J* 6.5 Hz, 17α-H), 3.19 and 3.25 (d,d, *J* 11 Hz, 18-H_2), 4.60 (m, 3α-H), 5.38 (m, 6-H), and 5.55 (m, 16β-H) (Found: C, 55.0; H, 6.3; I, 23.6. $C_{25}H_{35}IO_5$ requires C, 55.3; H, 6.5; I, 23.4%).

18,20-*Epoxy*-20-*ethoxypregn-5-ene*-3β,16α-*diol Diacetate* (8).—The 18-iodo-ketone (6) (845 mg) was dissolved in dry ethanol (50 ml), and silver acetate (728 mg) was added. The mixture was heated rapidly to 85—90 °C with stirring, kept at this temperature for 4 h, then cooled to 25 °C, and filtered through a bed of Celite. The solvent was then removed under reduced pressure. To remove traces of inorganic residues, the residue was taken up in ether, and the solution was washed with water, dried over anhydrous potassium carbonate, and evaporated under reduced pressure, to furnish the 18,20-*acetal* (8) (472 mg, 66%), m.p. 151—154° (from ethanol containing 0.1% triethylamine); $\nu_{max.}$ 1 740, 1 250, 1 175, 1 110, 1 030, 950, and 870 cm^{-1}; δ 0.95 (s, 19-H_3), 1.16 (t, *J* 7 Hz, 20-OCH_2CH_3), 1.52 (s, 21-H_3), 2.00 and 2.02 (s,s, 3-OAc, 16-OAc), 3.47 (q, *J* 7 Hz, 20-OCH_2CH_3), 3.48 and 3.74 (d,d, *J* 9 Hz, 18-H_2), *ca.* 4.60 (m, 3α-H), 5.12 (m, 16β-H), and 5.39 (m, 6-H) (Found: C, 70.7; H, 8.8. $C_{27}H_{40}O_6$ requires C, 70.4; H, 8.8%).

3β,16α-*Diacetoxy*-18-*hydroxypregn-5-en*-20-*one* 18,20-*Hemiacetal* (7).—The 18-iodo-ketone (6) (200 mg) was dissolved in aqueous 90% dioxan (12 ml), and silver acetate (172.3 mg) was added. The mixture was heated rapidly with stirring to 60—65 °C, and kept at this temperature for 4 h. Work-up as above gave the 18,20-*hemiacetal* (7) (83 mg, 52%), which did not crystallise, but was obtained as an amorphous solid on trituration with light petroleum; $\nu_{max.}$ 3 460, 1 740, 1 720, 1 250, 1 120, and 1 030 cm^{-1}; $\nu_{max.}$ (1% w/v in CCl_4, 1.00 mm path) 3 608, 3 450, 1 740, 1 230, 1 100, and 1 025 cm^{-1}; δ 0.95 (s, 19-H_3), 1.64 (s, 21-H_3), 2.01 (6 H, s, 3-OAc and 16-OAc), 3.75 (s, 18-H_2),[30] *ca.* 4.60 (m, 3α-H), 5.08 (m, 16β-H), and 5.38 (m, 6-H). The product was a mixture of C-20 isomers (*ca.* 4 : 1), as was apparent from the weaker singlets at 1.54 (21-H_3) and 3.65 (18-H_2) attributed to the minor component.

Optimization of the Reaction Conditions in the Hypoiodite Photolysis.—The three steps (photolysis, Jones oxidation, and silver-ion-promoted hydrolysis) were performed as described above, but without isolation of the iodo-intermediates (5) and (6). The proportion of iodine (0.56 mol per mol of steroid) was kept constant while lead tetra-acetate was used at different molar ratios. Product mixtures were analysed by n.m.r. on the basis of 19-H_3 peak heights for the individual steroids, with the following results: 2.44 mol $Pb(OAc)_4$: hemiacetal (7) 2.6%, lactone (9) 58%, ketone (2) 16%; 1.22 mol $Pb(OAc)_4$ (two runs): hemiacetal (7) 37 or 42%, lactone (9) 29 or 30%, ketone (2) 34 or 28%.

The products were separated by t.l.c. and identified by i.r. and ^1H n.m.r. The *lactone* (9) was obtained as a gum, $\nu_{max.}$ 1 760, 1 740, 1 240, 1 100, and 1 030 cm^{-1}; δ 1.12 (s,

19-H_3), 1.41 (d, *J* 6.5 Hz, 21-H_3), 2.02 and 2.04 (s,s, 3-OAc, 16-OAc), *ca.* 4.63 (m, 3α-H), 4.70 (q, *J* 6.5 Hz, 20-H), 4.93 (m, 3α-H), and 5.38 (m, 6-H).

Attempt to prepare the 18,20-*Hemiacetal* (7) *from the Lactone* (9).—*Reduction with DIBAL.* A solution of the lactone (9) (75 mg) in toluene (5.3 ml) was stirred under nitrogen at −70 °C, while a 20% solution of di-isobutylaluminium hydride in hexane (1 ml) was added dropwise. After 1 h the lactone had reacted completely (t.l.c.). A 2M-solution of propan-2-ol in toluene (0.3 ml) was then added, followed by saturated aqueous magnesium sulphate (*ca.* 4 ml), and the mixture was stirred for 30 min. The organic layer was then washed with water and dried, and the solvent was removed under reduced pressure to give crude 3β,16α,20β-*trihydroxypregn*-5-*en*-18-*al* 20,18-*hemiacetal* (10) (56.1 mg), which did not crystallise; δ[($^2H_5$)pyridine] 1.06 (s, 19-H_3), 1.42 (d, *J* 6.5 Hz, 21-H_3), *ca.* 3.85 (m, 3α-H), 4.2—4.8 (complex, 16β-H and 20-H), 5.35 (m, 6-H), and 5.72 (s, 18-H). The product was a mixture of C-18 isomers (*ca.* 7 : 3), as was apparent from the weaker singlet at 5.66 (18-H).

Complete acetylation of a sample of the crude triol (9) in pyridine–acetic anhydride at room temperature, yielded the crude *triacetate*, $\nu_{max.}$ 1 740, 1 240, 1 110, and 1 035 cm^{-1}; δ 0.98 (s, 19-H_3), 1.33 (d, *J* 6.5 Hz, 21-H_3), 2.01 (9 H, s, 3-OAc, 16-OAc, and 18-OAc), 4.00—4.75 (complex, 3α-H and 20-H), *ca.* 5.00 (16β-H), 5.38 (m, 6-H), and 6.27 (s, 18-H). A weaker singlet observed at δ 6.17 was attributed to 18-H in the less abundant component of a mixture of C-18 isomers.

Attempted Isomerization of 3β,16α,20β-*Trihydroxypregn*-5-*en*-18-*al* 20,18-*Hemiacetal* (10).—Compound (10) (60 mg) was treated with aluminium t-butoxide (54 mg) in refluxing toluene (30 ml). After 6 h the mixture was poured into a saturated solution of magnesium sulphate, and extracted into ether; the extract was washed, dried (K_2CO_3), and evaporated. T.l.c. and the ^1H n.m.r. spectrum of the crude product showed that no isomerization of compound (10) had occurred.

18,20-*Epoxy*-20-*ethoxypregn*-5-*ene*-3β,16α-*diol* (12).—The 20-ethoxy-3β,16α-diacetate (8) (400 mg) was heated under reflux in methanolic potassium hydroxide (0.12M; 20 ml) under nitrogen for 30 min. The product was precipitated by evaporation of most of the solvent and addition of water. After being dried over P_2O_5 the 3β,16α-*diol* (252.2 mg) crystallized from acetone containing 0.1% triethylamine; m.p. 189—191°; $\nu_{max.}$ 3 465, 3 300, 1 165, 1 120, 1 100, 1 050, 1 020, 950, and 870 cm^{-1}; δ 0.93 (s, 19-H_3), 1.16 (t, *J* 7 Hz, 20-OCH_2CH_3), 1.46 (s, 21-H_3), 3.46 (q, *J* 7 Hz, 20-OCH_2CH_3), 3.47 and 3.73 (d,d, *J* 9 Hz, 18-H_2), 3.2—3.8 (broad complex signal under the dd for 18-H_2 and the q for 20-OCH_2CH_3; three more protons were assigned as the 3α-H and two OH), 4.25 (m, 16β-H), and 5.36 (m, 6-H) (Found: C, 73.1; H, 9.7. $C_{23}H_{36}O_4$ requires C, 73.4; H, 9.6%).

16α,18-*Dihydroxypregn*-4-*ene*-3,20-*dione* 18,20-*Hemiacetal* (13).—The 3β,16α-diol (12) (100 mg) in dry toluene (9 ml) was treated with *N*-methyl-4-piperidone (0.5 ml), then heated with stirring under nitrogen, and 2.5 ml of toluene were distilled off. Aluminium isopropoxide (178.5 mg) in dry toluene (0.7 ml) was added and the mixture was heated under reflux and stirred under nitrogen for 6 h. The mixture was then cooled, diluted with ether, and a Rochelle salt solution (saturated aqueous) was added. The mixture was stirred for 15 min and the organic phase was separated. The extraction with ether was repeated, and the combined organic layers were washed with water, and stirred for 30 min with aqueous 5% H_2SO_4 (75 ml). The organic solution was then washed with brine and dried (K_2CO_3). A further extraction of the aqueous phase with dichloromethane gave a total of 88 mg of crude material, which after crystallization from acetone containing 0.1% triethylamine, yielded the *hemiacetal* (13), m.p. 185—186° (34 mg, 37%); $\nu_{max.}$ 3 370br, 1 660, 1 610, 1 120, 1 100, and 1 030 cm^{-1}; δ[($^2H_5$)pyridine], 0.96 (s, 19-H_3), 1.88 (s, 21-H_3), 4.03 (s, 18-H_2), 4.66 (m, 16β-H), 4.80 (m, OH), and 5.85 (s, 4-H); δ[(^2H)chloroform containing a few drops of ($^2H_5$)pyridine] 1.08 (s, 19-H_3), 1.62 (s, 21-H_3), 3.80 (s, 18-H_2), 3.46—4.14 (complex, two OH), 4.30 (m, 16β-H), and 5.73 (s, 4-H). When the amount of ($^2H_5$)pyridine in the solvent was increased the OH signal moved downfield.

21-*Acetoxy*-16α,18-*dihydroxypregn*-4-*ene*-3,20-*dione* 18,20-*Hemiacetal* (16α,18-*Dihydroxy-DOC* 21-*Acetate*) (14).—16α,18-Dihydroxyprogesterone 18,20-hemiacetal (13) (10 mg) was treated with lead tetra-acetate (13.4 mg) followed immediately by anhydrous acetic acid (1.4 ml). After swirling for 15 min at room temperature to dissolve the reactants, the mixture was poured into water. The product was extracted with dichloromethane and the extract was washed with water, saturated aqueous sodium hydrogen carbonate, and water until neutral, then dried and evaporated to give 16α,18-dihydroxy-DOC 21-acetate (14), m.p. 168—170° (from benzene) (8.2 mg, 71%); $\nu_{max.}$ (1.6% w/v in $CHCl_3$; 1.00 mm path) 3 570, 3 420, 1 735, 1 665, and 1 620 cm^{-1}; δ 1.14 (s, 19-H_3), 2.09 (s, 21-OAc), 3.82 (s, 18-H_2), 4.00—4.60 (m,m, 21-H_2 and 16β-H), 4.8 (m, OH), and 5.75 (s, 4-H). The product was a mixture of isomers at C-20 (*ca.* 5 : 4), as was apparent from the relatively less intense singlets at 2.07 (21-OAc) and 3.72 (18-H_2) from the minor isomer.

16α,18,21-*Trihydroxypregn*-4-*ene*-3,20-*dione* 18,20-*Hemiacetal* (16α,18-*Dihydroxy-DOC*) (1).—The crude 21-monoacetate (14) (8 mg) in methanol (0.5 ml) was treated with an aqueous 10% solution (20 μl) of potassium hydrogen carbonate. The mixture was kept at room temperature for 24 h, then the solvent was removed under reduced pressure and the solid residue was washed with water by decantation. Benzene was then added and evaporated off under reduced pressure to remove traces of water, and the crude product (5 mg) was dried over P_2O_5. T.l.c. of the crude material showed mainly one u.v.-absorbing polar spot, which developed a yellow colour with sulphuric acid. After isolation by preparative t.l.c., 16α,18-dihydroxy-DOC was obtained as a white solid which would not crystallise well and showed variable m.p.s over the range 176—184 °C, but appeared to resolidify in part and failed to melt completely even at 350 °C. This behaviour suggests spontaneous dimerisation during the melting process. The solid sample exhibited $\nu_{max.}$ 3 450—3 280, 1 660, 1 610, and 1 040 cm^{-1}; δ 1.15 (s, 19-H_3), 3.4—4.1 (complex, 18-H_2 and 21-H_2), 4.29 (m, 16β-H), and 5.75 (s, 4-H); mass spectrum [as 3,20-bis(methyloxime) 16,18,21-tris(trimethylsilyl) ether] *m/z* 636 (M^+), 605 (M^+ − OMe), 574 [M^+ − $(OMe)_2$], 515 (M^+ − OMe − Me_3SiOH), 425 [M^+ − OMe − $Me_3SiOH)_2$], 386, 346, 296, 148, and 103. The mass spectrum and g.l.c. retention time were identical with those of a sample kindly provided by Dr. S. L. Dale, Boston University.

Alternative Route via 16α-*Benzoate Derivatives* (*Scheme* 2).—3β-*Acetoxy*-16α-*benzoyloxypregn*-5-*en*-20-*one* (15). 3β-Acetoxy-16α,17α-epoxypregn-5-en-20-one [31] (20 g) in a mixture

of tetrahydrofuran (332 ml) and 95% ethanol (332 ml) was stirred at room temperature with freshly prepared aluminium amalgam (106 g). The reduction was monitored until t.l.c. showed complete disappearance of epoxide, then the mixture was diluted with chloroform and filtered through Celite. Evaporation left 3β-acetoxy-16α-hydroxypregn-5-en-20-one (16 g), m.p. 167—170° (from benzene) (lit.,[32] 169—170°); $\nu_{max.}$ 3 385, 1 735, 1 700, 1 245, and 1 040 cm^{-1}; δ 0.64 (s, 18-H_3), 1.01 (s, 19-H_3), 2.01 (s, 3-OAc), 2.26 (s, 21-H_3), 2.53 (d, *J* 6.5 Hz, 17α-H), *ca.* 4.60 (m, 3α-H), 4.83 (m, 16β-H), and 5.37 (m, 6-H).

The 3-acetate (15 g) in pyridine (74 ml) and benzene (74 ml) was treated with benzoyl chloride (15.2 ml) at room temperature for 3 h, then the mixture was poured with stirring into aqueous 10% sodium carbonate. Extraction by use of ether, followed by washing with 3M-HCl and water, and evaporation, afforded the 3-*acetate* 16-*benzoate* (15) (13.8 g), m.p. 134—135.5° (from methanol); $\nu_{max.}$ 3 030, 1 730, 1 715, 1 700, 1 600, and 1 240 cm^{-1}; δ 0.74 (s, 18-H_3), 1.04 (s, 19-H_3), 2.03 (s, 3-OAc), 2.19 (s, 21-H_3), 2.84 (d, *J* 6.5 Hz, 17α-H), *ca.* 4.60 (m, 3α-H), 5.38 (m, 6-H), 5.74 (m, 16β-H), and 7.3—8.1 (complex, 5 aromatic protons) (Found: C, 75.2; H, 8.2. $C_{30}H_{38}O_5$ requires C, 75.3; H, 8.0%).

18,20-*Epoxy*-20-*ethoxypregn*-5-*ene*-3β,16α-*diol* 3-*acetate* 16-*benzoate* (16). 3β-Acetoxy-16α-benzoyloxypregn-5-en-20-one (15) (8.31 g) in tetrahydrofuran (108 ml) and dry methanol (108 ml) was cooled to 0—5 °C, then sodium borohydride (4.27 g) was added and the mixture was stirred at 0—5 °C for 1 h. The product (7.1 g), isolated as for compound (4), was 3β-*acetoxy*-16α-*benzoyloxypregn*-5-*en*-20β-*ol*, m.p. 165—166° (EtOH); $\nu_{max.}$ 3 490, 3 030, 1 725, 1 710, 1 600, 1 585, 1 490, 1 275, 1 110, 1 040, and 715 cm^{-1}; δ 0.88 (s, 18-H_3), 1.03 (s, 19-H_3), 1.19 (d, *J* 6.5 Hz, 21-H_3), 2.01 (s, 21-OAc), 2.50 (m, OH), 3.98 (m, 20α-H), 4.61 (m, 3α-H), 5.06 (m, 16β-H), 5.35 (m, 6-H), and 7.3—8.1 (complex, 5 aromatic protons) (Found: C, 74.7; H, 8.5. $C_{30}H_{40}O_5$ requires C, 75.0; H, 8.4%).

The above 20-alcohol (6.45 g) was submitted to the ' hypoiodite ' reaction as described above to give a product from which the 18-*iodo*-20-*alcohol* (2.2 g) crystallized in 27% yield, m.p. 166—168° (MeOH); $\nu_{max.}$ 3 570, 3 030, 1 735, 1 705, 1 600, 1 285, and 1 245 cm^{-1}; δ 1.03 (s, 19-H_3), 1.21 (d, *J* 6.5 Hz, 21-H_3), 2.02 (s, 3-OAc), 2.50 (m, OH), 3.13 and 3.46 (d,d, *J* 11 Hz, 18-H_2), 4.34 (m, 20α-H), *ca.* 4.60 (m, 3α-H), 5.14 (m, 16β-H), 5.35 (m, 6-H), and 7.3—8.2 (complex, 5 aromatic protons) (Found: C, 59.0; H, 6.4; I, 20.9. $C_{30}H_{39}IO_5$ requires C, 59.4; H, 6.5; I, 20.9%).

The 18-iodo-alcohol (2.08 g) was treated with Jones' reagent, as described for compound (5), to give the 18-*iodo*-20-*ketone* (1.7 g), m.p. 128—131° (acetone–light petroleum); $\nu_{max.}$ 3 060, 1 735, 1 720, 1 700, 1 600, 1 280, and 1 240 cm^{-1}; δ 1.03 (s, 19-H_3), 2.02 (s, 3-OAc), 2.35 (s, 21-H_3), 2.82 (d, *J* 6.5 Hz, 17α-H), 3.28 and 3.30 (d,d, *J* 11 Hz, 18-H_2), 4.63 (m, 3α-H), 5.38 (m, 6-H), 5.79 (16β-H), and 7.3—8.2 (complex, 5 aromatic protons) (Found: C, 59.4; H, 6.1; I, 21.2. $C_{30}H_{37}IO_5$ requires C, 59.6; H, 6.2; I, 21.0%).

The 18-iodo-ketone (1.69 g) in dry ethanol (120 ml) was treated with silver acetate (1.45 g), under the conditions described for compound (6), to give the *acetal* (16) (1.46 g). The crude product, although apparently homogeneous (t.l.c.), did not crystallize; $\nu_{max.}$ 1 740, 1 710, 1 600, 1 580, 1 275, and 1 240 cm^{-1}; δ 0.95 (s, 19-H_3), 1.16 (t, *J* 7 Hz, 20-OCH_2CH_3), 1.59 (s, 21-H_3), 2.01 (s, 3-OAc), 3.49 (q, *J* 7 Hz, 20-OCH_2CH_3), 3.53 and 3.80 (d,d, *J* 9 Hz, 18-H_2), *ca.* 4.60 (m, 3α-H), 5.37 (complex, 16β-H and 6-H), and 7.3—8.2 (complex, 5 aromatic protons) (Found: C, 73.3; H, 8.4. $C_{32}H_{42}O_6$ requires C, 73.5; H, 8.1%).

21-*Acetoxy*-16α-*benzoyloxy*-3β,18-*dihydroxypregn*-5-*en*-20-*one* 18,20-*hemiacetal* (17). The acetal (16) (1.1 g) in dioxan (34.3 ml) and water (2.8 ml) was treated with potassium hydrogen carbonate (aqueous 2% solution; 11.6 ml) and heated under nitrogen at reflux for 16 h to hydrolyse the 3-acetate selectively. The solution was then concentrated under reduced pressure and extracted with ether; the extract was washed with water, dried ($MgSO_4$), and evaporated. Preparative t.l.c. [benzene–ether (1 : 3) containing 0.1% triethylamine] gave the 3β,16α-diol 16-benzoate (566 mg), $\nu_{max.}$ 3 440, 1 720, 1 600, 1 580, 1 490, and 1 270 cm^{-1}; δ 0.95 (s, 19-H_3), 1.16 (t, *J* 7 Hz, 20-OCH_2CH_3), 1.59 (s, 21-H_3), 3.49 (q, *J* 7 Hz, 20-OCH_2CH_3), 3.53 and 3.80 (d,d, *J* 9 Hz, 18-H_2), 3.2—3.9 (br, m under the dd for the 18-H_2 and the q for 20-OCH_2CH_3, 3α-H), 5.37 (complex, 16β-H and 6-H), and 7.3—8.2 (complex, 5 aromatic protons).

The crude 16-benzoate (386 mg) was treated with lead tetra-acetate (422 mg) followed immediately by anhydrous acetic acid (23 ml). The mixture was swirled from time to time for 30 min, then a little water was added and the solution was stirred at room temperature for 1 h (to convert any 18,20-acetal into 18,20-hemiacetal). The solution was then poured into water and the product was extracted with dichloromethane; the extract was washed with water, saturated aqueous sodium hydrogen carbonate, and water until neutral, dried, and evaporated, to give 356 mg of crude product. Preparative t.l.c. gave the 21-*acetoxy-derivative* (17) (191 mg, 43%), $\nu_{max.}$ 3 420, 1 745, 1 720, 1 600, 1 580, 1 275, 1 115, 1 055, 1 025, and 720 cm^{-1}; the product was a mixture of C-20 isomers (*ca.* 1 : 1) as was apparent from the presence of pairs of signals in the n.m.r. spectrum: δ 0.95 and 0.97 (s,s, 19-H_3), 2.05 and 2.08 (s,s, 21-OAc), *ca.* 3.60 (m, 3α-H), 3.76 and 3.86 (s,s, 18-H_2), 4.21 and 4.53 (s,s, 21-H_2), 5.35 (complex, 16β-H and 6-H), and 7.3—8.2 (complex, 5 aromatic protons); a small multiplet at 5.76 was assigned to 16β-H in one of the isomers; δ [($^2H_5$)-pyridine] 0.97 and 1.01 (s,s, 19-H_3), 1.92 and 2.00 (s,s, 21-OAc), *ca.* 3.80 (m, 3α-H), 3.94 and 4.10 (s,s, 18-H_2), 4.53 and 4.65 (d,d, *J* 11 Hz, 21-H_2), 4.87 and 5.01 (d,d, *J* 11 Hz, 21-H_2), 5.4 (complex, 6-H and 16β-H), 5.83 (m, 16β-H), and 7.4—8.5 (complex, 5 aromatic protons).

A second compound, less polar than (17), was isolated from the preparative t.l.c. in 11% yield and identified as 16α-*benzoyloxy*-3β-*hydroxyandrost*-5-*ene*-17β,18-*carbolactone* (19) (26 mg); $\nu_{max.}$ 3 400, 1 775, 1 725, 1 600, 1 580, and 1 275 cm^{-1}; δ 0.97 (s, 19-H_3), 2.80 (s, 17α-H), *ca.* 3.50 (m, 3α-H), 4.02 and 4.21 (d,d, *J* 11 Hz, 18-H_2), 5.32 (m, 6-H), 5.52 (m, 16β-H), and 7.3—8.2 (complex, 5 aromatic protons).

A third compound, more polar than (17), was isolated from the preparative t.l.c. in 6% yield and identified as 16α-*benzoyloxy*-3β,18,21-*trihydroxypregn*-5-*en*-20-*one* 18,20-*hemiacetal* (18) (15 mg) as a mixture of C-20 isomers; δ 0.97 (br,s, 19-H_3), 3.3—4.25 (complex, 18-H_2, 21-H_2, and 3α-H), 5.20 (m, 16β-H), 5.35 (m, 6-H), and 7.3—8.2 (complex, 5 aromatic protons). Acetylation of a sample (acetic anhydride–pyridine; room temperature overnight) gave the 3β,21-diacetate as a mixture of C-20 isomers; δ 0.97 (br,s, 19-H_3), 2.02 (s, 3-OAc), 2.07 and 2.09 (s,s, 21-OAc), 3.75 and 3.85 (s,s, 18-H_2), 4.20 and 4.53 (s,s, 21-H_2), *ca.*

4.60 (m, 3α-H), 5.35 (complex, 6-H and 16β-H), 5.76 (m, 16β-H), and 7.3—8.15 (complex, 5 aromatic protons).

21-*Acetoxy*-16α-*benzoyloxy*-18-*hydroxypregn*-5-*ene*-3,20-*dione* 18,20-*hemiacetal* (21). The 3β-hydroxy-compound (17) (150 mg) in acetone (7.5 ml) was stirred at 0 °C for 5 min with Jones' reagent (138 μl). Nitrogen was bubbled through the solvents and reaction solution before and during the oxidation. The excess of oxidant was destroyed with aqueous 10% sodium hydrogen sulphite, and the product was extracted with ether. The extract was washed with water, dried, and evaporated under reduced pressure to give the 3-*ketone* (21) (108 mg); $\nu_{max.}$ 3 420, 1 745, 1 725, 1 700sh, 1 600, 1 580, and 1 275 cm^{-1}; δ 1.13 and 1.15 (s,s, 19-H_3), 2.05 and 2.08 (s,s, 21-OAc), 3.78 and 3.88 (s,s, 18-H_2), 4.21 and 4.53 (s,s, 21-H_2), 5.34 (complex, 6-H and 16β-H), 5.76 (m, 16β-H), and 7.3—8.1 (complex, 5 aromatic protons). A mixture of C-20 isomers (*ca.* 1 : 1) was apparent from the doubling of most of the signals.

16α,18-*Dihydroxy-DOC* (1). Compound (21) (46 mg) in aqueous 70% dioxan (5 ml) containing aqueous 6% potassium hydroxide (0.2 ml) was kept at 50—70 °C under nitrogen for 4 h, then the product was extracted with ether. The extract was washed with water until neutral, dried, and evaporated to give a crude mixture (17 mg) which, after preparative t.l.c., gave 16α,18-dihydroxy-DOC (1) (3.5 mg), identical with the material described above.

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REFERENCES

[1] Part 9, D. N. Kirk and B. W. Miller, *J. Steroid Biochem.*, in the press.
[2] J. C. Melby and S. L. Dale, *Mayo Clin. Proc.*, 1977, **52**, 317.
[3] S. L. Dale and J. C. Melby, *Steroids*, 1973, **21**, 617.
[4] R. L. Braham, S. L. Dale, and J. C. Melby, *Steroids*, 1975, **26**, 697.
[5] D. N. Kirk and M. L. Sá e Melo, *Steroids*, 1979, **34**, 683.
[6] Ch. Meystre, K. Heusler, J. Kalvoda, P. Wieland, G. Anner, and A. Wettstein, *Helv. Chim. Acta*, 1962, **45**, 1317.
[7] K. Heusler and J. Kalvoda, *Angew. Chem., Int. Ed. Engl.*, 1964, **3**, 525.
[8] D. N. Kirk and M. S. Rajagopalan, *J. Chem. Soc., Perkin Trans. 1*, 1975, 1860.
[9] D. N. Kirk and M. S. Rajagopalan, *J. Chem. Soc., Chem. Commun.*, 1976, 77.
[10] D. N. Kirk and M. S. Rajagopalan, *Steroids*, 1976, **27**, 269.
[11] H. O. House, 'Modern Synthetic Reactions,' Benjamin, New York, 1972, p. 370.
[12] P. Choay, C. Monneret, and Khuong-Hua, *Bull. Soc. Chim. Fr.*, 1973, **4**, 1456.
[13] K. Heusler, J. Kalvoda, P. Wieland, G. Anner, and A. Wettstein, *Helv. Chim. Acta*, 1962, **45**, 2575.
[14] D. N. Kirk and M. S. Rajagopalan, *J. Chem. Soc., Perkin Trans. 1*, 1976, 1064.
[15] E. R. H. Walker, *Chem. Soc. Rev.*, 1976, **5**, 23.
[16] E. J. Corey, N. M. Weinshenker, T. K. Schaaf, and W. Huber, *J. Am. Chem. Soc.*, 1969, **91**, 5675.
[17] J. Schmidlin and A. Wettstein, *Helv. Chim. Acta*, 1963, **46**, 2799.
[18] V. Černý, A. Kasal, and F. Šorm, *Collect. Czech. Chem. Commun.*, 1966, **31**, 1752.
[19] L. Eignerová and A. Kasal, *Collect. Czech. Chem. Commun.*, 1976, **41**, 1056; A. Kasal and A. Trka, *ibid.*, 1977, **42**, 1389.
[20] D. N. Kirk and N. Whittaker, unpublished observation.
[21] R. Reich and J. F. W. Keana, *Synth. Commun.*, 1972, **2**, 323.
[22] S. Bernstein, M. Heller, and S. M. Stolar, *J. Am. Chem. Soc.*, 1954, **76**, 5674.
[23] J. T. Edward, M. Kaufman, R. W. Wojtowsky, D. M. S. Wheeler, and T. M. Barrett, *Can. J. Chem.*, 1973, **51**, 1610.
[24] G. H. Posner, R. B. Perfetti, and A. W. Runquist, *Tetrahedron Lett.*, 1976, 3499.
[25] G. H. Posner and M. J. Chapdelaine, *Synth. Commun.*, 1977, **8**, 555.
[26] A. Bodor, R. Schwartz, and D. Breazu, *Rev. Chim. (Bucharest)*, 1976, **27**, 929.
[27] W. D. Slaunwhite and A. J. Solo, *J. Pharm. Sci.*, 1975, **64**, 168.
[28] C. Djerassi, R. R. Engle, and A. Bowers, *J. Org. Chem.*, 1956, **21**, 1547.
[29] T. Miki, K. Morita, S. Noguchi, T. Kishi, K. Hiraga, and H. Nawa, *Chem. Pharm. Bull.*, 1963, **11**, 90.
[30] P. Génard, 'Contribution à la Détermination de la Configuration et de la Conformation Moléculaires des Corticosteroides,' Masson, Paris, 1974, p. 129.
[31] D. Löken, S. Kaufmann, G. Rosenkranz, and F. Sondheimer, *J. Am. Chem. Soc.*, 1956, **78**, 1738.
[32] K. I. H. Williams, M. Smulowitz, and D. K. Fukushima, *J. Org. Chem.*, 1963, **28**, 2101.