Hydroxylation of Pregn-16-en-20-ones. Part I. Permanganate Oxidation of Pregn-16-en-20-ones.

By GEORGE COOLEY, BERNARD ELLIS, FRANK HARTLEY, and VLADIMIR PETROW.

[Reprint Order No. 6502.]

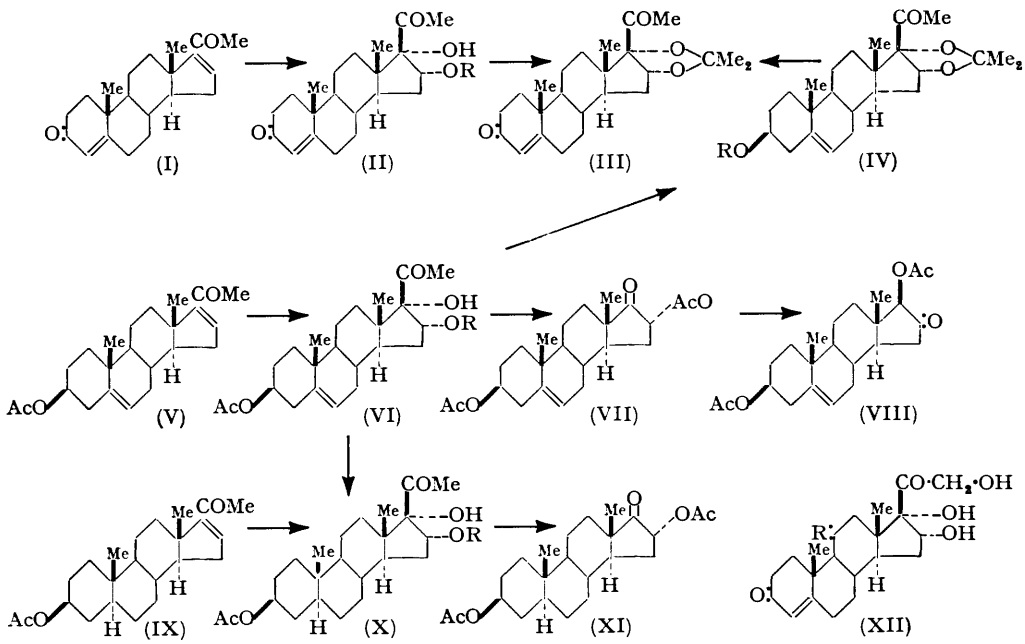
The conversion of certain pregn-16-en-20-ones into the corresponding 16α : 17α-dihydroxypregnan-20-ones by oxidation with potassium permanganate is described.

IN December, 1953, studies were initiated on the preparation of 16α-hydroxy-"Compound-S" (XII; R = H_2) and 16α-hydroxycortisone (XII; R = O), which were required for biological study. No information was available at the time on methods for building up the 16α : 17α-dihydroxyketol grouping. We consequently explored possible routes and first oxidised selected pregn-16-en-20-ones with potassium permanganate and with osmic acid. Unfortunately, both reagents gave rise not to one but to several series of hydroxy-compounds which, in addition, proved unexpectedly labile. At this stage a publication appeared by Inhoffen, Blomeyer, and Bruckner (*Chem. Ber.*, 1954, **87**, 593) (cf. B.P. 715,402, Løvens Kemiske Fabrik) covering some common ground. The views expressed therein on structural problems are largely at variance with conclusions since reached by ourselves and recorded in the present series. Their experimental results, in contrast, proved complementary to our own and provided valuable data for the elucidation

of structural issues. Our work is presented in three Parts, each dealing with a separate aspect of the problem.

Oxidation of 3β-acetoxypregna-5 : 16-dien-20-one (V) with potassium permanganate at 0° in acetone containing a limited quantity of acetic acid (cf. Part III) gave a product, $C_{23}H_{34}O_5$, which failed to show the ultraviolet absorption of an aβ-unsaturated ketone. The constitution 3β-acetoxy-16α : 17α-dihydroxypregn-5-en-20-one (VI; R = H) is assigned to this compound on the basis of the following transformations.

Reaction between this product and acetone containing a trace of hydrochloric acid gave the *iso*propylidene derivative (IV; R = Ac), the formation of which established the *cis*-configuration of the newly introduced α-glycol group (see Butenandt and Schäffer, *Z. Naturforsch.*, 1946, **1**, 82, and especially Huffman *et al.*, *J. Amer. Chem. Soc.*, 1944, **66**, 150; 1947, **69**, 1835). Acetylation of the triol monoacetate (VI; R = H) in pyridine furnished 3β : 16α-diacetoxy-17α-hydroxypregn-5-en-20-one (VI; R = Ac), which passed on catalytic hydrogenation into 3β : 16α-diacetoxy-17α-hydroxy*allo*pregnan-20-one (X; R = Ac), additionally obtained by oxidising 3β-acetoxy*allo*pregn-16-en-20-one (IX) with potassium permanganate, followed by acetylation of the resulting 3β-acetoxy-16α : 17α-dihydroxy*allo*pregnan-20-one (X; R = H).



Careful oxidation of the diol (X; R = Ac) with chromium trioxide in acetic acid led to 3β : 16α-diacetoxyandrostan-17-one (XI), a compound recently obtained by another method by Leeds, Fukushima, and Gallagher (*ibid.*, 1954, **76**, 2943), who have provided a rigorous proof of its structure. This observation establishes the 5-membered character of ring D in (X) (cf. Part II below), as well as the α-configuration of the 16-hydroxyl group. Further evidence on these points follows from the observation that the compound (VI; R = Ac) differs from the isomeric 3β : 16β-diacetoxy-17α-hydroxypregn-5-en-20-one recently obtained by Heusler and Wettstein (*Ber.*, 1954, **87**, 1301) by catalysed acetolysis of 3β-acetoxy-16α : 17α-epoxypregn-5-en-20-one. That the two compounds are, in fact, epimeric at $C_{(16)}$ has now been established by their conversion into the hitherto unknown 3β : 16α-diacetoxy- (VII) and 3β : 16β-diacetoxy-androst-5-en-17-one, respectively, by reduction of the 20-keto-groups with sodium borohydride, followed by oxidative cleavage of the

resulting (not isolated) 17 : 20-glycols with periodic acid. When either the diacetate (VII) or its $C_{(16)}$-epimer was treated with dilute methanolic potassium hydroxide at room temperature, a ketol rearrangement to the more stable 3β : 17β-dihydroxyandrost-5-en-16-one [isolated as the 3β : 17β-diacetate (VIII)] occurred in analogy to the formation of 3β : 17β-dihydroxyandrostan-16-one from the saturated diacetate (XI) under similar experimental conditions (cf. Leeds *et al.*, *loc. cit.*).

Oxidation of pregna-4 : 16-diene-3 : 20-dione (I) with potassium permanganate gave a hydroxylated derivative, $C_{21}H_{30}O_4$, with an absorption maximum at 240 mμ (ε 16,600) indicating the presence of the original 3-oxo-Δ^4-group and the absence of the 20-oxo-Δ^{16}-residue which had presumably been involved in the hydroxylation reaction. We consequently assign to this product the constitution 16α : 17α-dihydroxypregn-4-ene-3 : 20-dione (II; R = H) and in confirmation thereof find that it gives a monoacetate (II; R = Ac) on acetylation and an *iso*propylidene derivative identical with 16α : 17α-*iso*propylidenedioxypregn-4-ene-3 : 20-dione (III) formed by Oppenauer oxidation of the alcohol (IV; R = H) obtained from its acetate (IV; R = Ac) (see above) by alkaline hydrolysis.

Experimental

In this and the following two papers, optical rotations were measured in $CHCl_3$ in a 1-dm. tube, and ultraviolet absorption spectra (in propan-2-ol) were kindly determined by Mr. M. Davies, B.Sc.

3β-*Acetoxy*-16α : 17α-*dihydroxypregn*-5-*en*-20-*one* (VI; R = H).—A solution of potassium permanganate (18 g.) in aqueous acetone (1·05 l. of 85%) was added during 45 min. to a stirred ice-cooled solution of 3β-acetoxypregna-5 : 16-dien-20-one (40 g.) in a mixture of acetone (1·2 l.) and acetic acid (8 ml.). After treatment with sulphur dioxide, the pale yellow solution was decanted from inorganic salts, and most of the solvents were removed *in vacuo*. The product was extracted into ether (*ca.* 2 l.) and the extract washed with water, aqueous sodium hydrogen carbonate, and water and then dried. Concentration to 400 ml. gave crystals (14·7 g.), m. p. 140—170°, a further quantity of material (13·8 g.) of m. p. 170—185° being obtained by concentration of the mother-liquor to half its bulk. The combined products were crystallised several times from methanol to give 3β-*acetoxy*-16α : 17α-*dihydroxypregn*-5-*en*-20-*one* (8 g.), dense plates, m. p. 210—212°, $[\alpha]_D^{20}$ −65° (*c*, 1·37) (Found : C, 71·0; H, 9·0. $C_{23}H_{34}O_5$ requires C, 70·7; H, 8·8%).

Acetylation in pyridine for 30 min. at 100° gave 3β : 16α-*diacetoxy*-17α-*hydroxypregn*-5-*en*-20-*one*, silky needles (from methanol), m. p. 214—215°, $[\alpha]_D^{20}$ −74° (*c*, 1·02) (Found : C, 69·4; H, 8·45. $C_{25}H_{36}O_6$ requires C, 69·4; H, 8·4%).

3β-*Acetoxy*-16α : 17α-iso*propylidenedioxypregn*-5-*en*-20-*one* (IV; R = Ac).—3β-Acetoxy-16α : 17α-dihydroxypregn-5-en-20-one (350 mg.) in hot acetone (20 ml.) was treated with 2 drops of concentrated hydrochloric acid. The solution was gently boiled for 2 min., then set aside overnight, and the product obtained on dilution with water purified from aqueous methanol. The iso*propylidene* derivative (300 mg.) formed needles, m. p. 175°, $[\alpha]_D^{21}$ −18° (*c*, 0·75) (Found : C, 72·9; H, 8·8. $C_{26}H_{38}O_5$ requires C, 72·5; H, 8·9%).

3β : 16α-*Diacetoxy*-17α-*hydroxy*allo*pregnan*-20-*one* (X; R = Ac).—3β : 16α-Diacetoxy-17α-hydroxypregn-5-en-20-one (810 mg.) in ethyl acetate (35 ml.) containing prereduced Adams platinum catalyst (50 mg.) and a trace of perchloric acid was hydrogenated at room temperature and pressure until 1·1 equivs. of hydrogen were absorbed. The product was purified from aqueous methanol, giving needles (430 mg.) of 3β : 16α-*diacetoxy*-17α-*hydroxy*allo*pregnan*-20-*one*, m. p. 159°, $[\alpha]_D^{21}$ −16° (*c*, 0·54) (Found : C, 69·6; H, 9·0. $C_{25}H_{38}O_6$ requires C, 69·1; H, 8·8%).

3β-*Acetoxy*-16α : 17α-*dihydroxy*allo*pregnan*-20-*one* (X; R = H).—A stirred ice-cooled solution of 3β-acetoxy*allo*pregn-16-en-20-one (3·5 g.) in acetone (100 ml.) was treated with a solution of potassium permanganate (1·4 g.) in aqueous acetone (140 ml. of 90%) dropwise during 1 hr. After decolorisation with sulphur dioxide, the mixture was decanted from inorganic salts and concentrated *in vacuo* until solids began to separate. The product (1·75 g.), m. p. 183°, was purified from methanol to give lustrous plates of 3β-*acetoxy*-16α : 17α-*dihydroxy*allo*pregnan*-20-*one*, m. p. 222°, $[\alpha]_D^{21}$ +3° (*c*, 0·76) (Found : C, 70·3; H, 9·3. $C_{23}H_{36}O_5$ requires C, 70·4; H, 9·25%).

Acetylation in pyridine gave the 16α-acetoxy-derivative, m. p. 159°, not depressed on admixture with a specimen prepared by the method described above.

3β : 16α-*Diacetoxyandrostan*-20-*one* (XI).—3β : 16α-Diacetoxy-17α-hydroxy*allo*pregnan-20-one (200 mg.) in acetic acid (5 ml.) was treated for 21 hr. at room temperature with chromium trioxide (100 mg.) in acetic acid (5 ml. of 98%). The neutral fraction of the oxidation product was crystallised from aqueous methanol, giving 3β : 16α-diacetoxyandrostan-20-one (50 mg.), needles, m. p. 183—184°, $[\alpha]_D^{22}$ +55° (*c*, 0·5) (Found : C, 70·5; H, 8·8. Calc. for $C_{23}H_{34}O_5$: C, 70·7; H, 8·8%). Leeds *et al.* (*loc. cit.*) give m. p. 184—185°, $[\alpha]_D^{23}$ +57·1°.

3β : 16α-*Diacetoxyandrost*-5-*en*-17-*one* (VII).—To a suspension of 3β : 16α-diacetoxy-17α-hydroxypregn-5-en-20-one (1·65 g.) in methanol (100 ml.) was added a solution of sodium borohydride (500 mg.) in methanol. A few drops of acetic acid were added after 10 min.; then the mixture was poured into water and the product isolated with ether. The solids obtained were dissolved in methanol (60 ml.) and treated for 18 hr. with periodic acid (1·6 g.) in water (15 ml.). The product, isolated with ether, was purified from aqueous ethanol to give 3β : 16α-*diacetoxyandrost*-5-*en*-17-*one* (700 mg.), needles, m. p. 167—168°, $[\alpha]_D^{20}$ −18° (*c*, 0·97) (Found : C, 70·7; H, 8·4. $C_{23}H_{32}O_5$ requires C, 71·1; H, 8·3%).

3β : 16β-*Diacetoxyandrost*-5-*en*-17-*one*.—Sodium borohydride (300 mg.) was added to a suspension of 3β : 16β-diacetoxy-17α-hydroxypregn-5-en-20-one (900 mg.) in methanol (50 ml.). After 10 min., the clear solution was acidified with acetic acid and diluted with water, and the precipitate collected by filtration. The air-dried material in methanol (60 ml.) was treated for 18 hr. with periodic acid (1 g.) in water (10 ml.). Dilution with water gave a crystalline solid which was purified from aqueous methanol. 3β : 16β-*Diacetoxyandrost*-5-*en*-17-*one* separated in bright needles, m. p. 180—181° (sinters at 165°), $[\alpha]_D^{22}$ +6° (*c*, 0·82) (Found : C, 71·1; H, 8·1. $C_{23}H_{32}O_5$ requires C, 71·1; H, 8·3%).

3β : 17β-*Diacetoxyandrost*-5-*en*-16-*one* (VIII).—The foregoing compound (150 mg.) in methanolic 0·04N-potassium hydroxide (75 ml.) was kept at room temperature for 18 hr. After dilution with water, the product was isolated with methylene dichloride–ether, and acetylated in pyridine in the usual manner. 3β : 17β-Diacetoxyandrost-5-en-16-one separated from aqueous methanol in flat needles, m. p. and mixed m. p. 124—125°.

The same compound was obtained by similar treatment of 3β : 16α-diacetoxyandrost-5-en-17-one.

16α : 17α-*Dihydroxypregn*-4-*ene*-3 : 20-*dione* (II; R = H).—Potassium permanganate (8 g.) in aqueous acetone (230 ml. of 85%) was added during 1 hr. to a stirred ice-cooled solution of pregna-4 : 16-diene-3 : 20-dione (20 g.) in a mixture of acetone (600 ml.) and acetic acid (4 ml.). After treatment with sulphur dioxide, the solution was decanted from inorganic salts and concentrated *in vacuo* to a small volume. The product was extracted with ether, and the extract was washed with water, aqueous sodium hydrogen carbonate, and water, and dried. Concentration to 150 ml. gave dense crystals (7·3 g.), m. p. 209—220°, purified by extraction with a small volume of hot acetone followed by crystallisation from ethanol–methylene dichloride. 16α : 17α-*Dihydroxypregn*-4-*ene*-3 : 20-*dione* formed needles, m. p. 225°, $[\alpha]_D^{22}$ +95° (*c*, 0·81) (Found : C, 72·3; H, 8·75. $C_{21}H_{30}O_4$ requires C, 72·8; H, 8·7%).

On acetylation in pyridine, 16α-*acetoxy*-17α-*hydroxypregn*-4-*ene*-3 : 20-*dione* was obtained, needles (from aqueous methanol), m. p. 176—177°, $[\alpha]_D^{20}$ +49° (*c*, 1·25) (Found : C, 67·8, 68·7; H, 8·4, 8·3. $C_{23}H_{32}O_5,H_2O$ requires C, 68·1; H, 8·2%).

16α : 17α-iso*Propylidenedioxypregn*-4-*ene*-3 : 20-*dione* (III).—(*a*) 3β-Acetoxy-16α : 17α-*iso*propylidenedioxypregn-5-en-20-one (3 g.) in methanolic 2·5% potassium hydroxide1(40 ml.) was refluxed for 30 min. Careful addition of water gave needles (2·5 g.), m. p. 2 7—218° (Found : C, 74·2; H, 9·3. $C_{24}H_{36}O_4$ requires C, 74·2; H, 9·3%). A solution of thisproduct (2·3 g.) in toluene (80 ml.) and *cyclo*hexanone (20 ml.) was distilled until 25 ml. of distillate had been collected. Following the addition of aluminium *iso*propoxide in toluene (18 ml. of 25% solution), the mixture was refluxed for 30 min., cooled and extracted with concentrated aqueous, Rochelle salt solution. Removal of solvents by steam-distillation gave crystals (1·7 g.), m. p. 190—200°. Purified from aqueous ethanol, 16α : 17α-iso*propylidenedioxypregn*-4-*ene*-3 : 20-*dione* formed needles, m. p. 210°, $[\alpha]_D^{20}$ +137° (*c*, 0·7) (Found : C, 74·2; H, 9·0. $C_{24}H_{34}O_4$ requires C, 74·6; H, 8·9%).

(*b*) 16α : 17α-Dihydroxypregn-4-ene-3 : 20-dione (350 mg.) in hot acetone (20 ml.) was treated with 2 drops of concentrated hydrochloric acid. Next morning, the solution was poured into water and the precipitate crystallised from aqueous ethanol, to give the *iso*propylidene derivative (330 mg.), needles, identical with a sample prepared by method (*a*) in m. p., mixed m. p., and optical rotation.

The compound was recovered substantially unchanged after treatment with aqueous acetic acid (70%) for 2·5 hr. at 100°.

The authors thank the Directors of The British Drug Houses Ltd. for permission to publish the work in this and the two following papers.

CHEMICAL RESEARCH LABORATORIES,
THE BRITISH DRUG HOUSES, LTD., LONDON, N.1. [*Received, June 13th,* 1955.]