When refluxed with acetic anhydride it gave a diacetate which was crystallized from methanol and melted at 150–152°. When mixed with chlorogenin acetate (from natural chlorogenin), m. p. 151–153°, it melted at 151–153°.

Anal. Calcd. for $C_{31}H_{48}O_6$: C, 72.0; H, 9.4. Found: C, 72.2; H, 9.7.

Oxidation of Digitogenin Residues.—The residues from the crystallization of 100 g. of digitogenin as the acetate were hydrolyzed and extracted with ether. These weighed 38 g. This was dissolved in 1 liter of acetic acid. Chromic acid (50 g.) in 500 cc. of 90% acetic acid was added slowly, while keeping the temperature at 20°. It was allowed to stand at this temperature for twenty minutes, water was added and the product extracted with ether. The acids were removed by shaking with water and sodium carbonate solution, and the ether was evaporated. The residue was refluxed for one hour with 1% potassium hydroxide in methanol. The solution was then diluted with water, extracted with ether and the product sublimed in a high vacuum at 130–160°. The sublimate was crystallized from methanol to give a product melting at 233–236°. When mixed with chlorogenone, m. p. 235–237°, it melted at 234–237°.

Anal. Calcd. for $C_{27}H_{40}O_4$: C, 75.7; H, 9.4. Found: C, 75.5; H, 9.4.

Summary

Evidence has been presented indicating that the hydroxyl groups of chlorogenin are 3-beta and 6-alpha.

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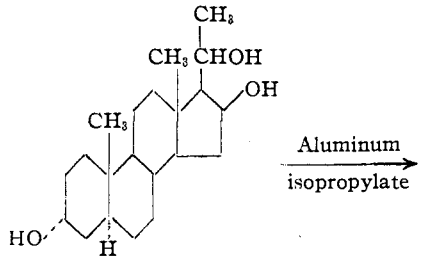
[CONTRIBUTION FROM THE SCHOOL OF CHEMISTRY AND PHYSICS OF THE PENNSYLVANIA STATE COLLEGE]

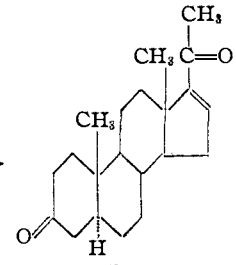
Sterols. CIII. The Oxidation of Pregnanetriols

BY RUSSELL E. MARKER AND D. L. TURNER

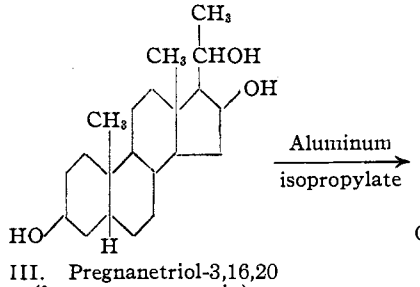
An *allo*-pregnanetriol isolated from the urine of pregnant mares[1] was believed by Odell and Marrian[2] to be pregnanetriol-3(α),6,20. Marker and Wittle[3] obtained much evidence indicating that the compound was *allo*-pregnanetriol-3,16,20.



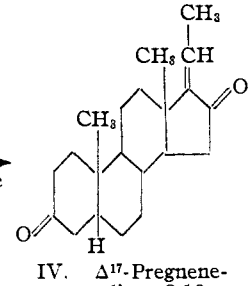
I. *allo*-Pregnanetriol-3,16,20 (from mares' urine)



II. Δ^{16}-*allo*-Pregnenedione-3,20



III. Pregnanetriol-3,16,20 (from sarsasapogenin)



IV. Δ^{17}-Pregnenedione-3,16

Recently pseudo-tigogenin was converted to Δ^{16}-*allo*-pregnenedione-3,20 by oxidation with chromic acid under mild conditions. This was reduced with hydrogen using a barium sulfate–palladium catalyst to *allo*-pregnanedione-3,20.[4] Oxidation using aluminum isopropylate and a large excess of cyclohexanone[5] has now been applied to the *allo*-pregnanetriol-3,16,20 from mares' pregnancy urine. The product obtained was Δ^{16}-*allo*-pregnenedione-3,20, identical with that obtained from tigogenin. This compound has also been obtained by Butenandt, Mamoli and Heusner[6] from androsterone. The oxidation evidently occurs first at the 3 and 20 positions and the product loses water under the conditions used to give the unsaturated diketone. Reduction with palladium–barium sulfate catalyst and hydrogen gives *allo*-pregnanedione-3,20, a compound previously prepared from *allo*-pregnanetriol-3,16,20 in poor yield.[7]

The preparation of Δ^{16}-*allo*-pregnenedione-3,20 from the triol of Marrian proves conclusively that the structure proposed by Marker and Wittle[3] is correct.

Sarsasapogenin has been oxidized to pregnanetriol-3,16,20 of the coprostane series.[8] By using the dry persulfate reagent of von Baeyer[9] the yield of triol in this oxidation

(1) Haslewood, Marrian and Smith, *Biochem. J.*, **28**, 1316 (1934).
(2) Odell and Marrian, *J. Biol. Chem.*, **125**, 333 (1938).
(3) Marker and Wittle, THIS JOURNAL, **61**, 855 (1939).
(4) Marker and Rohrmann, *ibid.*, **62**, 898 (1940).
(5) *Cf.* Oppenauer, *Rec. trav. chim.*, **56**, 137 (1937).
(6) Butenandt, Mamoli and Heusner, *Ber.*, **72**, 1614 (1939).
(7) Marker, Kamm, Wittle, Oakwood and Lawson, THIS JOURNAL, **60**, 1067 (1938).
(8) Marker, Rohrmann, Crooks, Wittle, Jones and Turner, *ibid.*, **62**, 525 (1940).
(9) Von Baeyer and Villiger, *Ber.*, **32**, 3625 (1899).

has been increased considerably when the oxidation is carried out at room temperature. Under these conditions there are no acid products formed.

When the pregnanetriol-3,16,20 from sarsasapogenin was oxidized with cyclohexanone–aluminum isopropylate, a pregnenedione was obtained. Since this is not identical with Δ^{16}-pregnenedione-3,20 prepared by the mild chromic acid oxidation of pseudosarsasapogenin,[10] it is probably Δ^{17-20}-pregnenedione-3,16, a compound analogous to one obtained from the *allo*-pregnanetriol-3,16,20 by Marker and Wittle.[3] The latter *allo*-pregnenedione was made by the partial hydrolysis of the triacetate of Marrian's triol followed by oxidation of the diol monoacetate and hydrolysis of the product.

We wish to thank Parke, Davis and Company for their help.

Experimental Part

Oxidation of *allo*-Pregnanetriol-3,16,20 with Aluminum Isopropylate.—A mixture of 3 g. of *allo*-pregnanetriol-3,16,20 (from mares' urine), 15 g. of aluminum isopropylate, 40 cc. of cyclohexanone and 250 cc. of toluene was refluxed for eighteen hours. Water and ether were added; the ethereal layer was shaken with hydrochloric acid, then with dilute sodium hydroxide solution and finally washed well with water. The solvents were removed *in vacuo* and the residue was steam distilled for two hours. The product was then sublimed in a high vacuum at 130–140°. The sublimate was crystallized from ether–pentane, and finally directly from ethyl ether. It melted at 209–211°. When mixed with Δ^{16}-*allo*-pregnenedione-3,20, prepared from the oxidation of pseudotigogenin,[4] m. p. 210–212°, there was no depression; yield, 1.4 g.

Anal. Calcd. for $C_{21}H_{30}O_2$: C, 80.2; H, 9.6. Found: C, 80.2; H, 9.68.

Reduction of Δ^{16}-*allo*-Pregnenedione-3,20 with Palladium.—A solution of 200 mg. of Δ^{16}-*allo*-pregnenedione-3,20, obtained from the above triol, in 50 cc. of ether was shaken with 200 mg. of palladium–barium sulfate catalyst under a pressure of 1.7 atm. of hydrogen for five hours. The catalyst was filtered and the product was crystallized from methanol and acetone, m. p. 200°. Mixed with *allo*-pregnanedione-3,20, m. p. 200–201°, there was no depression in melting point.

Anal. Calcd. for $C_{21}H_{32}O_2$: C, 79.7; H, 10.2. Found: C, 79.5; H, 10.2.

Preparation of Pregnanetriol-3(β),16,20.—A solution of sarsasapogenin acetate (10 g.) in 500 cc. of glacial acetic acid was mixed with 100 g. of the dry reagent of von Baeyer and Villiger (prepared from 20 g. of potassium persulfate, 22 g. of concentrated sulfuric acid and 60 g. of potassium sulfate). The mixture was stirred vigorously at room temperature for three days. Sufficient dilute sodium hydroxide solution was added to neutralize the sulfuric acid and the mixture was concentrated to 50 cc. *in vacuo*. The residue was poured into water and filtered. This was hydrolyzed by refluxing for fifteen minutes in ethanolic potassium hydroxide. Water was then added and the precipitated material was filtered, washed with water and dried. This product had m. p. 224–228°. The yield was 4.0 g. After recrystallization from methanol it melted at 227–228°. Mixed with pregnanetriol-3,16,20 previously prepared,[8] m. p. 223–226°, it melted at 223–228°.

Oxidation of Pregnanetriol-3,16,20 with Aluminum Isopropylate.—The oxidation of the pregnanetriol from sarsasapogenin (5 g.) was carried out as described above with *allo*-pregnanetriol-3,16,20 except that the product was separated from by-products containing hydroxyl groups by boiling in pyridine for one hour with succinic anhydride. The pyridine solution was poured into water and extracted with ether. The pyridine was removed by washing with water and freed of the half succinates by washing with saturated sodium carbonate solution. The ether solution on evaporation gave a white solid which was recrystallized from methanol, m. p. 179–182°. When mixed with Δ^{16}-pregnenedione-3,20, it melted at 150–168°.

Anal. Calcd. for $C_{21}H_{30}O_2$: C, 80.2; H, 9.6. Found: C, 80.0; H, 9.7.

Summary

The oxidation of pregnanetriol-3,16,20 and *allo*-pregnanetriol-3,16,20 by the method of Oppenauer has been studied.

(10) Marker and Rohrmann, THIS JOURNAL, **62**, 518 (1940).

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