[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF THE UPJOHN COMPANY]

A Synthesis of Pregnane-3,20-dione from Stigmasterol and Ergosterol[1]

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A stereospecific hydrogenation of stigmastadienone (IV) and isoergosterone (III), obtained from stigmasterol and ergosterol, yielded 5β-stigmast-22-en-3-one (VIa) and 5β-ergost-22-en-3-one (VIb), respectively. A quantitative method is described for the determination of the ratio of *allo* and normal isomers in the reduction products. Ozonolysis of both ketones, of the normal series, yielded 3-ketobisnorcholan-22-al (VII) which was in turn converted to pregnane-3,20-dione (XI).

The microbiological oxygenation of pregnane-3,20-dione (XI) to the corresponding 11α-hydroxylated steroid has been described in a previous paper[2] from these laboratories. This 11-oxygenated steroid, possessing the normal A/B ring configuration, is a useful intermediate in the synthesis of certain cortical hormones. The present paper describes the preparation of pregnanedione from two naturally occurring sterols, stigmasterol (Ia) and ergosterol (Ib).

Stigmasterol was converted by the Oppenauer oxidation to 4,22-stigmastadien-3-one (IV) in 80% yield. Similar treatment of ergosterol yielded 4,7,22-ergostatrien-3-one (II) which was isomerized with acid to 4,6,22-ergostatrien-3-one (isoergosterone) (III).[3]

The next step in the synthesis was the selective hydrogenation of the nuclear double bonds of stigmastadienone and isoergosterone without reaction in the side chain. The catalytic hydrogenation of stigmastadienone yields a mixture of isomeric saturated-ring-A ketones. These were separated by chromatography and purified by several recrystallizations from acetone to yield the desired normal isomer VIa, m.p. 110.5–111.0°, $[\alpha]_D$ +11° (chf); and *allo* isomer Va, m.p. 170–171°, $[\alpha]_D$ +19° (chf). These compounds have been reported by Barton and Brooks[4] who obtained them, along with certain other reduction products, by catalytic reduction of stigmastadienone in ethyl acetate using platinum oxide as a catalyst. A stereospecific reduction yielding only the desired normal isomer was the first objective of this work.

The catalytic hydrogenation of a Δ^{14}-16-keto steroid to the corresponding saturated 16-keto steroid has been reported by Wilds, Johnson and Sutton.[5] They found that the inclusion of potassium hydroxide in the hydrogenation mixture favored, to a large extent, the formation of the isomer having the C/D ring fusion *cis*. Others[6a-d] have also hydrogenated certain Δ^4-3-ketones in alkali. With one exception,[7] the hydrogenations in the presence of alkali were described as highly stereospecific. Incomplete data were given on the exact isomer ratios found in the reduction products.

The reduction, employing alkali, of stigmastadienone was developed to a high order of stereospecificity. Such stereospecificity was obtained using as catalysts palladium supported on zinc oxide,[8] charcoal, calcium carbonate, cadmium carbonate, barium carbonate and barium sulfate. The isomer ratio depends to a certain extent on the conditions present during hydrogenation, hence, palladium-on-zinc oxide and palladium-on-charcoal were selected for detailed study of the effects of varying the amounts of catalyst, alkali, steroid and solvent. Conditions were found which allow reduction of stigmastadienone in 99% yield to a product assaying as high as 93% of the 5β-stigmast-22-en-3-one (VIa); the remaining 7% was 22-stigmasten-3-one (Va). This *allo* isomer was removed readily by recrystallization from acetone. For example, the normal isomer content of a sample was raised from 91 to 97% by one recrystallization from acetone in a recovery of 87%.

Optimal conditions varied somewhat from one catalyst to another. For example, in isopropyl alcohol at room temperature using 5% palladium-on-charcoal, a catalyst level of 4% of the steroid weight and a potassium hydroxide level of 21% was found to be effective; while for 5% palladium on zinc oxide a catalyst level of 8% and potassium hydroxide level of 5% was best.

Several solvents were evaluated. The reduction products were only slightly soluble in methanol. Their crystallization during the hydrogenation apparently coated the surface of the catalyst and sometimes stopped the reaction short of completion. Isopropyl alcohol showed good solubility characteristics and proved to be the solvent of choice. Ethanol caused about the same results but the rate was somewhat faster. Reduction in

(1) Presented before the Division of Organic Chemistry at the 124th Meeting of the American Chemical Society, Chicago, Ill., September 6–11, 1953, Abstracts p. 3-0; *Chem. Eng. News*, **31**, 3977 (1953). Two papers have since appeared on the preparation of pregnanedione from ergosterol by similar procedures: A. F. Daglish, J. Green and V. D. Poole, *Chemistry & Industry*, **45**, 1207 (1953); Francis Johnson, G. T. Newbold and F. S. Spring, *J. Chem. Soc.*, 1302 (1954).

(2) S. H. Epstein, D. H. Peterson, H. M. Leigh, H. C. Murray, A. Weintraub, L. M. Reineke and P. D. Meister, THIS JOURNAL, **75**, 421 (1953); H. C. Murray and D. H. Peterson, U. S. Patent 2,602,769.

(3) D. A. Shepherd, R. A. Donia, J. A. Campbell, B. A. Johnson, R. P. Holysz, G. Slomp, Jr., J. E. Stafford, R. L. Pederson and A. C. Ott, THIS JOURNAL, **77**, 1212 (1955).

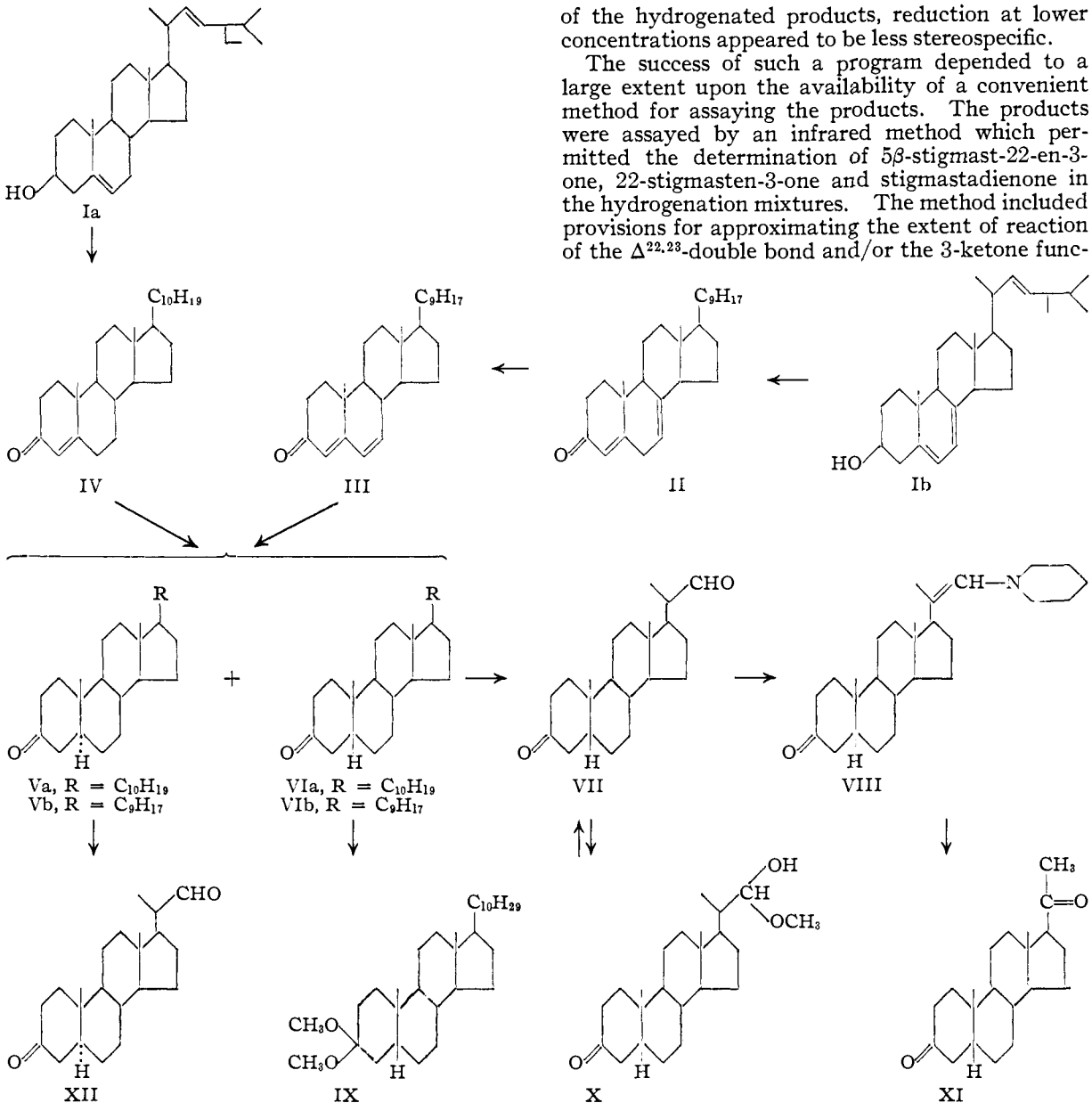
(4) D. H. R. Barton and C. J. W. Brooks, *ibid.*, **72**, 1633 (1950).

(5) A. L. Wilds, J. A. Johnson, Jr. and R. E. Sutton, *ibid.*, **72**, 5524 (1950).

(6) (a) P. L. Julian, "Recent Progress in Hormone Research," Vol. VI, Academic Press, Inc., New York, N. Y., 1951, pp. 207, 212; (b) O. Mancera, H. J. Ringold, C. Djerassi, G. Rosenkranz and F. Sondheimer, THIS JOURNAL, **75**, 1286 (1953); (c) M. Velasco, J. Rivera, G. Rosenkranz, F. Sondheimer and C. Djerassi, *J. Org. Chem.*, **18**, 92 (1953), and references cited therein; (d) R. Yashin, G. Rosenkranz and C. Djerassi, THIS JOURNAL, **73**, 4654 (1951), and references cited therein.

(7) C. Djerassi, R. Yashin and G. Rosenkranz, *ibid.*, **74**, 422 (1952).

(8) The preparation and utility of this catalyst will be described in a future communication from these Laboratories by Dr. G. S. Fonken.



methylcellosolve was less stereospecific and ethyl acetate, necessarily with no alkali present, showed poor selectivity, *i.e.*, there was considerable reduction of the side-chain double bond. At higher catalyst levels (50 to 100% of steroid weight) no decrease in selectivity was observed but the stereospecificity of the hydrogenation was less favorable. The amount of alkali present showed a definite inverse effect on reaction rate. The stereospecificity was not changed appreciably over a broad range varying somewhat with different catalyst supports, but at low levels poorer stereospecificity and selectivity were observed. Substitution of sodium hydroxide in place of potassium hydroxide resulted in lowered stereospecificity. Temperature showed the expected effect on reaction rate but the stereospecificity was only insignificantly improved at lower temperatures. Although the maximum steroid concentration was determined by the solubility of the hydrogenated products, reduction at lower concentrations appeared to be less stereospecific.

The success of such a program depended to a large extent upon the availability of a convenient method for assaying the products. The products were assayed by an infrared method which permitted the determination of 5β-stigmast-22-en-3-one, 22-stigmasten-3-one and stigmastadienone in the hydrogenation mixtures. The method included provisions for approximating the extent of reaction of the $\Delta^{22,23}$-double bond and/or the 3-ketone function, if it occurred. No attempt was made to determine quantitatively the compounds resulting from hydrogenation of the last-mentioned functions or from ketal formation.

It has been established that infrared spectra afford a useful means of differentiating between normal and *allo* isomers. In general, however, the spectra have not permitted an assignment of the *cis* or *trans* A-ring fusion to a specific compound. The 3-acetoxy steroids constitute an exception to this generalization.[9] Therefore, the configurations of the reference samples of the normal and *allo*-stigmastenone isomers, separated by chromatography over Florisil and purified by subsequent recrystallizations from methanol and acetone, were assigned initially on the basis of precedent.[4] The assignments were later proved to be correct when a

(9) R. N. Jones, P. Humphries, F. Herling and K. Dobriner, THIS JOURNAL, **73**, 3215 (1951).

sample tentatively designated as 5β-stigmast-22-en-3-one was converted to pregnane-3,20-dione.[10]

Portions of the spectra of 5β-stigmast-22-en-3-one, 22-stigmasten-3-one and stigmastadienone in chloroform are reproduced in Fig. 1. Stigmastadienone was characterized by its conjugated ketone absorption at 6.02 μ. The normal and *allo*-stigmastenones were characterized, respectively, by bands at 10.125 and 10.44 μ which probably resulted from wagging frequencies of the hydrogen atom at C-5. Figure 2 shows portions of the spectrum of a mixture of the three compounds. All of the compounds showed a strong absorption at 10.26 μ which originated with the 22,23-double bond. The intensity of this band in the spectra of hydrogenated mixtures measured the extent of side chain reduction. The band at 5.86 μ originated with the saturated 3-ketone function of the normal and *allo* stigmastenones. Thus, in the spectra of reduction mixtures, the intensity of this absorption measured any reaction of the saturated ketone.

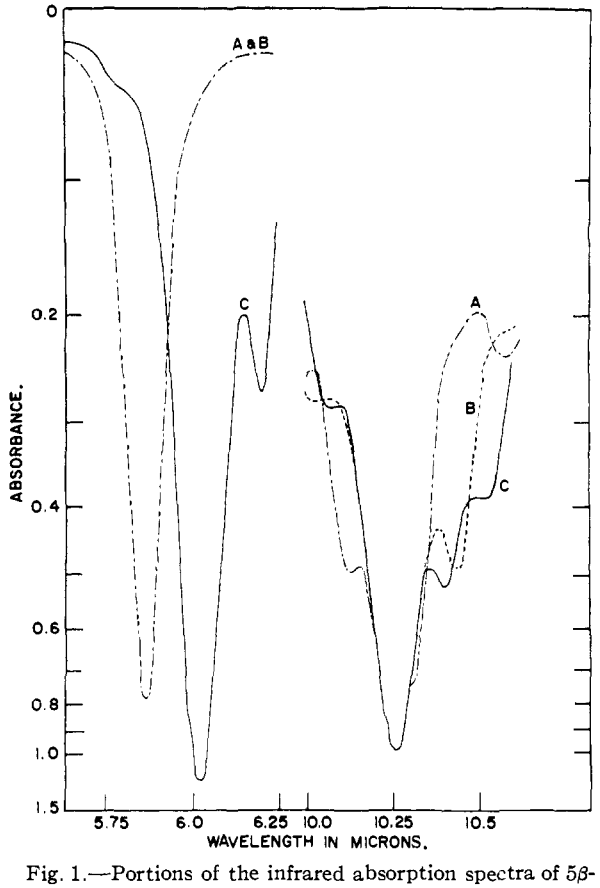


Fig. 1.—Portions of the infrared absorption spectra of 5β-stigmast-22-en-3-one (A), stigmast-22-en-3-one (B) and stigmastadienone (C); chloroform solutions.

The saturated ketone could have been destroyed in two ways: first, by reduction to an hydroxyl group and, secondly, by conversion to a ketal or hemiketal during recrystallization from methanol, if the solution was heated for an extended time.

(10) (a) A. Butenandt, *Ber.*, **63B**, 659 (1930); (b) A. Butenandt and J. Schmidt, *ibid.*, **67**, 1901 (1934); (c) A. Butenandt and G. Fleischer, *ibid.*, **68**, 2094 (1935); (d) C. W. Shoppee and T. Reichstein, *Helv. Chim. Acta*, **24**, 351 (1941); and others.

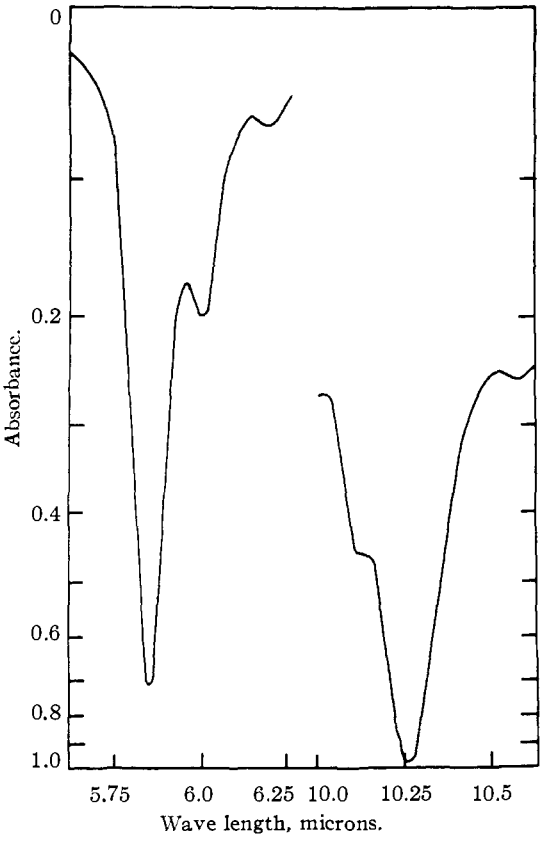


Fig. 2.—Portions of the infrared absorption spectrum of a reduction mixture (77.2% normal, 10.9% *allo*, 11.9% dienone).

In this study petrolatum mull spectra were measured for any sample showing weaker saturated ketone absorption than could be accounted for on the basis of its stigmastadienone content. In all such cases ketal formation had occurred, and the selectivity of the reduction was established as 4,5-double bond > 22,23-double bond > 3-ketone. The evidence for ketal formation was the absence of hydroxyl absorption at about 3 μ and the presence of strong ether C–O stretching absorption at 9.11 and 9.55 μ. A sample of the 3-dimethylketal (IX) was prepared by refluxing 5β-stigmast-22-en-3-one in methanol in the presence of a small amount of acid. The ketal was characterized and hydrolyzed to regenerate the 3-keto compound.

The results of several determinations made on synthetic mixtures indicated the accuracy of the *allo* and normal determinations to be ±3%. The reproducibility of results as determined by duplicate determinations on the same sample was ±2%. The accuracy of the stigmastadienone determinations was ±1% (see Table I).

The catalytic hydrogenation of isoergosterone also yielded a mixture of isomeric saturated-ring-A ketones Vb and VIb. These compounds have been reported by Barton, Cox and Holness.[11]

The same conditions which were used for the stereospecific reduction of stigmastadienone were applied to the reduction of isoergosterone. Reaction

(11) D. H. R. Barton, J. D. Cox and N. J. Holness, *J. Chem. Soc.*, 1771 (1949).

TABLE I

ASSAY RESULTS FOR PREPARED MIXTURES

Mixture	Composition Normal, %	*allo*, %	Stigmastadienone, %	Found Normal, %	*allo*, %	Stigmastadienone, %
1	75.0	25.0	0.0	72.0	27.0	0.0
				73.1	28.0	0.5
2	70.0	25.0	5.0	68.9	26.1	4.2
3	72.5	25.0	2.5	73.0	24.0	2.1
4	50.0	50.0	0.0	48.4	51.6	0.5
5	25.0	75.0	0.0	23.0	76.0	0.0
6	92.5	6.5	1.0	92.3	7.4	0.3
				92.7	6.8	0.5
				92.2	7.4	0.3
				91.1	8.7	0.3

with one mole of hydrogen yielded 4,22-ergostadien-3-one. Reaction with two moles of hydrogen yielded the isomeric ketones 22-ergosten-3-one (Vb) and 5β-ergost-22-en-3-one (VIb) in a ratio of 12 to 88, respectively. This ratio was determined by isolation, employing chromatography.

The purified normal isomers derived from both the stigmasterol and ergosterol sequences were converted to pregnane-3,20-dione (XI) by a modification of the method of Heyl and Herr.[12] The 3-ketobisnorcholan-22-al (VII) was obtained first in 85% yield by ozonolysis of 5β-ergost-22-en-3-one. Additional ozonolyses of 5β-stigmast-22-en-3-one were performed in order to establish the optimal conditions for the reaction, and yields of 90–95% resulted. The *allo* isomer from the ergosterol series Vb was ozonized to the corresponding *allo*-aldehyde XII in 73% yield.

The normal aldehyde VII had the unusual property of reacting with methanol to form a stable, isolable hemiacetal X. Thus, when methanol was used instead of isopropyl alcohol for crystallization, the hemiacetal was obtained instead of the aldehyde.

This aldehyde VII was converted to 22-N-piperidylbisnor-20(22)-cholen-3-one (VIII), and subsequent oxidation of the total product with sodium dichromate yielded the desired pregnanedione[10] (XI) in an over-all yield from the aldehyde of 71%.

Thus another starting material has been made readily available for the versatile bioxygenation process and for the preparation of C-11 oxygenated cortical hormones.

Acknowledgments.—The authors are indebted to Mrs. G. S. Fonken for assistance in the determination and interpretation of absorption spectra, to Mr. William A. Struck and his associates for analytical data, to Mr. L. M. Reineke and associates for papergram analyses, and to Mr. M. A. Rebenstrof for technical assistance.

Experimental[13]

Infrared Assay.—Measurements were made on a Perkin–Elmer model 21-double-beam spectrometer equipped with a sodium chloride prism. The following instrument settings were employed: resolution, 2; response, 1; gain, 4.5; speed, 0.5 μ per minute.

Chloroform solutions, 7% w./v., of the unknown samples and reference standards were used. A portion of each solution was loaded into a 0.489-mm. cell and placed in the sample beam. A 0.475-mm. cell containing chloroform was placed in the reference beam. The spectrum was scanned twice over the range 9.75 to 10.75 μ. A second portion of each solution was loaded into a 0.125-mm. sample cell. A 0.112-mm. cell containing chloroform was placed in the reference beam. The spectrum was recorded twice from 5.5 to 6.5 μ.

The compositions of samples were determined as follows: (1) Stigmastadienone was determined at 6.02 μ using the base-line technique.[14] The base line was drawn between points on the curve at 5.6 and 6.3 μ.

$$\% \text{ stigmastadienone} = Ab/K$$

where Ab = base-line absorbance of sample and K = specific absorbance of stigmastadienone.

(2) The total saturated 3-ketone content of the sample was measured at 5.86 μ. This was done by means of a simple one-component analysis since the intensity of the absorption was essentially independent of A/B ring configuration (Fig. 1).

$$\% \text{ 3-ketone} = A/K$$

where A = absorbance of sample and K = average specific absorbance of the *allo* and normal stigmastenones. If the total value for any sample was lower than could be explained by its content of unreduced stigmastadienone it was not carried further. Mull spectra were run on such samples; those containing ketals were hydrolyzed and the analyses repeated.

(3) Absorption intensities at 10.26 μ were inspected. A relatively low intensity indicated reduction of the side-chain double bond, and the analysis was not carried further.

(4) Absorbancies measured at 10.125 and 10.44 μ were corrected for any contribution from stigmastadienone.

$$A\lambda_{corr} = A\lambda - KsCs$$

where $A_{\lambda\,corr}$ = corrected absorbance at wave length λ, Ks = specific absorbance of stigmastadienone, and Cs = concentration of stigmastadienone expressed as a decimal fraction.

(5) Using the $A_{\lambda\,corr}$ values the percentages of *allo* and normal isomers were calculated according to the equations

$$\% n = 100 \times \frac{K_{2a}A_1 - K_{1a}A_2}{K_{1n}K_{2a} - K_{2n}K_{1a}}$$

$$\% a = 100 \times \frac{K_{2n}A_2 - K_{1n}A_1}{K_{2n}K_{1a} - K_{1n}K_{2a}}$$

where n is 5β-stigmast-22-en-3-one, a stigmast-22-en-3-one, 1 10.125 μ, 2 10.44 μ, A absorbance, and K specific absorbance.

5β-Stigmast-22-en-3-one (VIa).—A suspension of 0.247 g. of 6.8% palladium-on-zinc oxide catalyst (American Platinum Works) in 125 ml. of isopropyl alcohol, which had been redistilled from potassium hydroxide, containing 3.085 g. of stigmastadienone (IV) (m.m.p.(K) 128–129.5°, $[\alpha]_D$ +58° (*c* 1.06)) and 0.155 g. of potassium hydroxide (reagent grade, 85% minimum) was prepared in a Parr bottle. The bottle was quickly placed in a Parr hydrogenator, equipped with a mercury manometer for pressure readings, and the system was flushed with hydrogen three times in rapid succession. The pressure was raised to 500 mm. above atmospheric pressure, and the shaker was started, the whole procedure requiring about 3–4 minutes. Shaking was stopped when the pressure drop corresponded with the consumption of one molar equivalent of hydrogen (previously determined by calibration with sorbic acid). The pressure was released, and the mixture was filtered using Celite. The slightly green solution was neutralized with glacial acetic acid. The stoichiometric amount was just enough to discharge the color, and the solution was evaporated to dryness. The residue was triturated with 50 ml. of benzene, the resulting benzene solution was evaporated to

(12) (a) F. W. Heyl and M. E. Herr, THIS JOURNAL, **72**, 2617 (1950); (b) M. E. Herr and F. W. Heyl, *ibid.*, **74**, 3627 (1952).

(13) All melting points, m.p.'s, were determined with a Fisher–Johns block which had been checked against standard compounds; m.m.p.(K)'s are melting points determined between crossed polaroids on a Kofler micro hot-stage apparatus which had been checked against standard compounds. Infrared spectra were obtained with a Perkin–Elmer recording infrared spectrophotometer model 21. Rotations, $[\alpha]_D$, were determined in chloroform at 23–25°.

(14) J. J. Heigl, M. F. Bell and J. U. White, *Anal. Chem.*, **19**, 293 (1947).

dryness, and the residue dried in an oven at 60° and 30 mm. The yield of 5β-stigmast-22-en-3-one (VIa) was 3.077 g. (99.2%), $[\alpha]_D$ +11° (*c* 1.030), m.p. 107–111°; quantitative infrared assay: purity, 92.6 ± 3%; *allo*, 6.8 ± 3%; stigmastadienone, 0.6 ± 1%.

Several other catalysts yielded approximately equivalent results. In the same manner as described above the reduction of 3.00 g. of stigmastadienone in the presence of 0.63 g. of potassium hydroxide and 0.18 g. of 5% palladium-on-charcoal (American Platinum Works) resulted in a virtually quantitative yield of 5β-stigmast-22-en-3-one (VIa); quantitative infrared assay: purity, 91.0 ± 3.0%; *allo*, 8.7 ± 3.0%; stigmastadienone, 0.3 ± 1%.

The purity of this and similar samples was raised to 96.5–97% by recrystallization from acetone in which the first two small crops (about 13%) of *allo*-rich material were discarded.

In order to obtain reference standards for the infrared method of analysis, the product from a preliminary 60-g. hydrogenation was chromatographed over 6 kg. of Florisil. The normal isomer was eluted by 0.2% acetone–Skellysolve B and was followed by a very small quantity of an intermediate fraction. The *allo* isomer passed through in the remainder of the 0.2% acetone, in the 0.5% acetone, and in the acetone washings.

A middle portion of the normal fraction was recrystallized once from methanol and four times from acetone; m.p. 110.5–111°, m.m.p.(K) 111.5–113°, $[\alpha]_D$ +11° (*c* 1.095).

Anal. Calcd. for $C_{29}H_{48}O$: C, 84.45; H, 11.72. Found: C, 84.49; H, 11.33.

A middle portion of the *allo* fraction after five recrystallizations from acetone melted at 170–171°, $[\alpha]_D$ +19° (*c* 0.966).

Anal. Calcd. for $C_{29}H_{48}O$: C, 84.45; H, 11.72. Found: C, 84.18; H, 11.84.

5β-Stigmast-22-en-3-one 3-Dimethylketal (IX).—A solution of 25.0 g. of 5β-stigmast-22-en-3-one and 0.25 g. of *p*-toluenesulfonic acid monohydrate in 800 ml. of methanol was heated under reflux 4 hours. At the end of this time the reflux condenser was removed and the solution was concentrated to 300 ml. Crystallization occurred spontaneously during the concentration. The crystals were separated by filtration, after cooling the reaction mixture to room temperature. Recrystallization from 500 ml. of methanol afforded 22.0 g., m.p. 102–103°, $[\alpha]_D$ +8° (*c* 3.11). The infrared spectrum indicated the complete absence of hydroxyl and carbonyl absorption; however, strong side-chain double bond (10.26 μ) and C–O (9.12 μ) absorptions were present.

Anal. Calcd. for $C_{31}H_{54}O_2$: C, 81.16; H, 11.87; CH_3O, 13.53. Found: C, 81.17, 82.55; H, 11.71, 11.66; CH_3O, 12.41, 12.75.

5β-Ergost-22-en-3-one (VIb).—By the same procedure as described above 12.5 g. of isoergosterone (III) was hydrogenated in 3.75 l. of methanol (redistilled from potassium hydroxide) containing 1.4 g. of potassium hydroxide (reagent grade) in the presence of 2.0 g. of 5% palladium-on-charcoal (Amer. Plat. Works). After 63.0 millimoles of hydrogen had been absorbed, 12.4 g. (99% yield) of hydrogenated product was obtained. The product exhibited no absorption in the ultraviolet at either 242 or 286 mμ.

This material was chromatographed over Florisil, eluting with acetone in petroleum ether and resolved into 87% 5β-ergost-22-en-3-one (VIb), m.p. 106–111°, and 11% 22-ergosten-3-one (Vb), m.p. 160–167°, with an intermediate 2% m.p. 99–150°.

The normal fraction was recrystallized from acetone–methanol to yield 5β-ergost-22-en-3-one[11] (VIb), m.m.p.(K) 112.5–114°, $[\alpha]_D$ −5° (*c* 0.603).

The *allo* fraction was also recrystallized from acetone–methanol to yield 22-ergosten-3-one[11] (Vb), m.m.p.(K) 167–170°, $[\alpha]_D$ +7° (*c* 1.19).

3-Ketobisnorcholan-22-al (VII). From 5β-Ergost-22-en-3-one.—The saturated aldehyde was prepared by a modification of the procedure of Heyl and Herr.[12] A solution of 5.00 g. (12.5 millimoles) of 5β-ergost-22-en-3-one (m.m.p.(K) 109–112°, $[\alpha]_D$ −5°) in 225 ml. of methylene chloride and 2 ml. of pyridine in a 300-ml. pear-shaped flask equipped with a magnetic stirrer was sparged with ozone-rich oxygen at Dry-Ice temperature until 16.6 millimoles of ozone (1.33 moles of ozone per mole of steroid) was absorbed. The color of the reaction mixture changed from orange to yellow, and disappeared completely at the end-point.

The ozonized mixture was stirred for 1 hour at room temperature with 30 ml. of acetic acid and 10 g. of zinc dust. After filtration, the solution was washed with ice-water, 5% sodium bicarbonate, and again with water and then dried with sodium sulfate. The solution was concentrated at reduced pressure to about 20 ml., at which point 40 ml. of isopropyl alcohol was added. The solution was concentrated further at reduced pressure until a few crystals formed. The mixture was heated gently until the crystals redissolved. Slow cooling to room temperature followed by refrigeration at 0° for 12 hours resulted in crystallization. The yield of the first fraction was 2.60 g., m.m.p.(K) 140–142°. Concentration of the mother liquors afforded an additional 0.93 g., m.p. 135–141°, the total yield of 3-ketobisnorcholanal being 3.53 g. or 85%. Recrystallization from ether afforded an analytical sample (prisms), m.m.p.(K) 140–143°, $[\alpha]_D$ +19° (*c* 0.608).

Anal. Calcd. for $C_{22}H_{34}O_2$: C, 79.95; H, 10.37. Found: C, 79.73, 79.82; H, 10.28, 10.45.

The infrared spectrum indicated ketone and aldehyde absorption bands.

From 5β-Stigmast-22-en-3-one (VIa).—By the same procedure 18.22 g. of 5β-stigmast-22-en-3-one in 880 ml. of methylene chloride and 5.5 ml. of pyridine at −40 to −45° was allowed to react with 71 millimoles of ozone. The washed and dried solution was evaporated to dryness to remove α-ethylisovaleraldehyde, formed from the side-chain fragment, and crystallized from Skellysolve B–ether to yield 3-ketobisnorcholan-22-al (VII) in two crops: (1) 7.41 g., 50.7% yield, m.p. 136–138°; (2) 6.30 g., 43.2% yield, m.p. 129–132°; or 93.9% of material having $[\alpha]_D$ +19° (*c* 1.13). The infrared spectrum was like that of the aldehyde from ergosterol (above).

3-Ketobisnorallocholan-22-al (XII).—Ozonolysis of 3.97 g. (10 millimoles) of 22-ergosten-3-one (Vb) with 13.3 millimoles of ozone was accomplished in 120 ml. of methylene chloride solution containing 0.8 ml. of anhydrous pyridine at −78°. The ozonized reaction mixture was treated with 5 g. of zinc dust and 20 ml. of acetic acid. Processing in the usual manner afforded 2.40 g. (72.7% yield) of crude 3-ketobisnorallocholan-22-al (XII), m.p. 138–144° dec. Recrystallization from ether raised the melting point to 150–154°, $[\alpha]_D$ +25° (*c* 0.364). A second recrystallization gave material of analytical purity, m.p. 152–156° dec., $[\alpha]_D$ +28° (*c* 0.954).

Anal. Calcd. for $C_{22}H_{34}O_2$: C, 79.95; H, 10.37. Found: C, 79.95; H, 10.39.

The infrared spectrum differed from that of the isomeric normal aldehyde (VII).

3-Ketobisnorcholan-22-al Methyl Hemiacetal (X).—One gram of 3-ketobisnorcholanal was dissolved in 10 ml. of methanol; 2 ml. of water and 1 drop of acetic acid were added, and the solution was warmed on the steam-bath for 30 minutes. The precipitate which formed on cooling the solution to 0° was collected on a filter and washed with methanol and warm ether. The identity of this material was deduced from the following evidence. The infrared spectrum showed ketone and very strong hydroxyl absorption; chromatography over Florisil and elution with petroleum ether containing 2% acetone yielded the free aldehyde; the aldehyde was very soluble in methanol, ether or acetone, but the solubility of the hemiacetal was low; elemental analysis and Zeisel methoxyl determination confirmed the structure. The specific rotations of the aldehyde and hemiacetal were both +19° in chloroform.

Anal. Calcd. for $C_{23}H_{38}O_3$: C, 76.20; H, 10.56. Found: C, 76.41, 76.62; H, 10.79, 10.39; $[\alpha]_D$ +19° (*c* 0.752).

22-N-Piperidylbisnor-20(22)-cholen-3-one (VIII).—The "enamine" was prepared under the conditions described by Herr and Heyl[12b] modified to include the use of *p*-toluenesulfonic acid monohydrate as an acid catalyst. Reaction of 8.26 g. of 3-ketobisnorcholanal with 2.55 g. of piperidine in benzene solution in the presence of 25 mg. of *p*-toluenesulfonic acid catalyst afforded 9.73 g. (98% yield) of crude "enamine," m.p. 98–104°, $[\alpha]_D$ +20° (*c* 0.796).

Anal. Calcd. for $C_{27}H_{43}NO$: N, 3.52. Found: N, 3.42, 3.57.

A sample of this material was recrystallized twice from petroleum ether to yield 22-N-piperidylbisnor-20(22)-cholen-3-one, m.p. 105–108°, $[\alpha]_D$ +21° (*c* 0.879), $\lambda_{max}^{methanol}$ 227 mμ, ε 6,175.

Anal. Calcd. for $C_{27}H_{43}NO$: C, 81.55; H, 10.90; N, 3.52. Found: C, 81.74; H, 10.78; N, 3.43.

In a similar manner the "enamine" also was obtained from 3-ketobisnorcholanal methyl hemiacetal (X).

Pregnane-3,20-dione (XI).—A solution of 7.95 g. of 22-N-piperidylbisnor-20(22)-cholen-3-one (m.p. 98–104°, $[\alpha]_D$ +20°; N, 3.42, 3.57) in 60 ml. of benzene was added dropwise in approximately one hour to a stirring solution of 11.92 g. of sodium dichromate dihydrate in 60 ml. of benzene and 40 ml. of acetic acid. The temperature was maintained at 5–10° throughout addition of the "enamine" solution and for 2 hours thereafter. The very dark reaction mixture was diluted with 200 ml. of water, the layers were separated, the aqueous layer was extracted with 100 ml. of benzene, and the organic layers were combined. The benzene solution was washed successively with 35 ml. of water, two 35-ml. portions of 10% sodium hydroxide solution, 35 ml. of water, 35 ml. of 10% hydrochloric acid solution, and four 35-ml. portions of water. The resulting colorless solution was concentrated to dryness at 40–100° (15–20 mm.) leaving 5.02 g. (79.4% yield) of colorless oil which crystallized spontaneously on cooling. Recrystallization from ether gave pregnane-3,20-dione (XI) in two crops: (1) 2.60 g. (41% yield), m.p. 120–122°, $[\alpha]_D$ +111° (*c* 0.996); (2) 1.90 g. (30.0%), m.p. 120–122°, $[\alpha]_D$ +110° (*c* 1.048); total yield, 4.50 g. (71%). The residue (0.50 g.) was approximately 50% pregnanedione according to paper strip chromatographic assay. The above melting point agrees well with the literature values[10] which range from 120 to 123° and the infrared spectrum was identical to that for an authentic sample.

Pregnane-3,20-dione (XI), m.p. 118–121.5°, $[\alpha]_D$ +112° (*c* 2.215), also was obtained directly from aldehyde in a similar manner, without isolation of the "enamine," in a yield of 68.5%.

KALAMAZOO, MICHIGAN

[CONTRIBUTION FROM THE RESEARCH LABORATORIES, CHEMICAL DIVISION, MERCK & CO., INC.]

The Synthesis of 5α-Pregnane-3β,17α,21-triol-11,20-dione (Reichstein's Substance D) and 5α-Pregnane-3β,11β,17α,21-tetrol-20-one (Reichstein's Substance V) from 5α-Pregnan-3β-ol-11,20-dione

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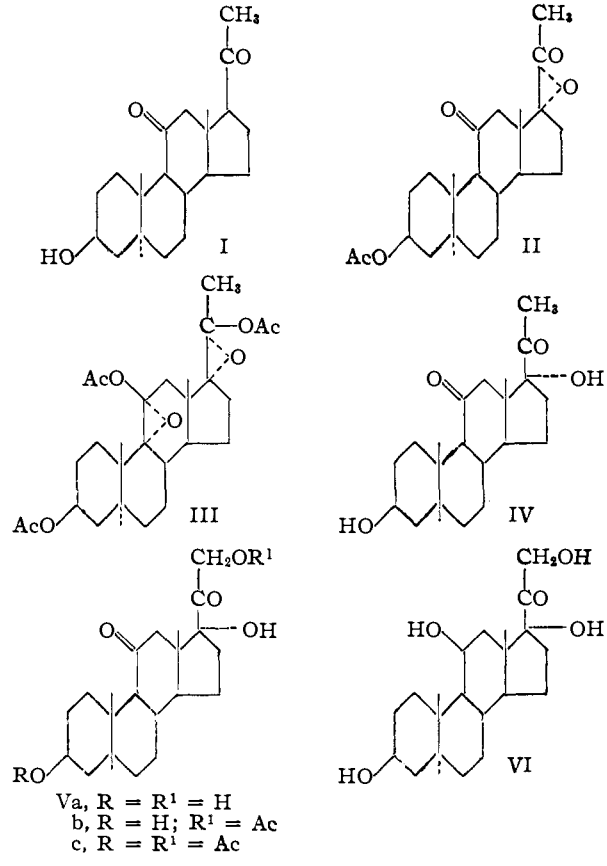
The synthesis of 5α-pregnane-3β,17α,21-triol-11,20-dione and 5α-pregnane-3β,11β,17α,21-tetrol-20-one from 5α-pregnan-3β-ol-11,20-dione is described. The latter compound is hydroxylated at C-17 by conversion to the 17,20-enol acetate followed by peracid oxidation. Hydrolysis affords 5α-pregnane-3β,17α-diol-11,20-dione which is brominated at C-21 and then acetoxylated by reaction with potassium acetate to 5α-pregnane-3β,17α,21-triol-11,20-dione 21-acetate. The triol is transformed to 5α-pregnane-3β,11β,17α,21-tetrol-20-one by reduction of the 11-keto group with sodium borohydride after protection of the 20-keto group by formation of the semicarbazone.

The ready availability of 5α-pregnan-3β-ol-11,20-dione[1] (I) along with well developed methods for elaborating the dihydroxyacetone side chain and reduction of 11-keto groups to 11-hydroxyl makes attractive the synthesis of some of the so-called "inactive companion substances" isolated from extracts of the adrenal cortex.[2]

The present paper is concerned with the synthesis of two of these compounds, 5α-pregnane-3β,17α,-21-triol-11,20-dione (Va) (Reichstein's Substance D)[3] and 5α-pregnane-3β,11β,17α,21-tetrol-20-one (VI) (Reichstein's Substance V).[4]

The introduction of the 17α-hydroxyl group into I was accomplished by a modification of the procedure of Kritchevsky and Gallagher.[5] In the original procedure of Gallagher the enolization of the 20-keto group was brought about by distilling off acetic anhydride in the presence of *p*-toluenesulfonic acid as a catalyst. In the present instance this treatment was too drastic in that the 11-keto group also was enolized and subsequent treatment of the enol acetate with peracid afforded largely the 9(11),17,20-diepoxy compound (III) and very lit-



Va, R = R¹ = H
b, R = H; R¹ = Ac
c, R = R¹ = Ac

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