Anal. Calcd. for $C_{24}H_{33}N_3O_5$: N, 9.50. Found: N, 9.57.

11-Dehydrocorticosterone-3,20-bis-semicarbazone (XII).—Prepared in the same manner as described for the preparation of (VIII). Obtained as a micro-crystalline solid, m.p. over 300° (dec.).

Anal. Calcd. for $C_{23}H_{34}N_6O_4$: N, 18.03. Found: N, 18.02.

Reichstein's Substances E and U Diacetate (XIII) and (XIV).—A solution of 4.2 g. of (V) in 600 cc. of dry tetrahydrofuran was added with stirring at 25° to a solution of 2.4 g. of lithium borohydride in 120 cc. of tetrahydrofuran. Some separation of material was observed at this point. The mixture was stirred for one hour after complete addition and then the excess lithium borohydride was decomposed under cooling with 10% aqueous acetic acid. Concentration of the resulting clear solution and trituration of the residue with water afforded, on filtration and drying at 90°, 3.3 g. of crude, reduced product. The latter was acetylated with 19 cc. of pyridine and 18 cc. of acetic anhydride at 90° for ten minutes. The solvents were removed *in vacuo* and the residue washed with water and dried at room temperature. This crude reacetylated material was treated with 35 cc. of acetic acid, 11 cc. of water, 5.95 g. of sodium acetate and 5.6 cc. of 90% pyruvic acid and heated for four hours at 75°. At the end of this period the solvents were removed *in vacuo* and the residue dissolved in chloroform. The chloroform solution was washed with 5% aqueous sodium bicarbonate solution and dried over anhydrous sodium sulfate. The solvent was evaporated *in vacuo* and the residue chromatographed on acid-washed alumina. Fractional elution with benzene–chloroform afforded Reichstein's Substance E diacetate, obtained as needles from ethyl acetate–ether, m.p. 229–231°.

Anal. Calcd. for $C_{25}H_{36}O_7$: C, 66.94; H, 8.09. Found: C, 67.08; H, 7.98.

A mixed melting point of this material with an authentic specimen[4] was not depressed.

Further fractions from the chromatography yielded Reichstein's Substance U diacetate obtained as thick prisms from ethyl acetate, m.p. 252–253°.

Anal. Calcd. for $C_{25}H_{34}O_7$: C, 67.24; H, 7.67. Found: C, 67.46; H, 7.24.

A mixed melting point of this material with an authentic sample prepared by chromic acid oxidation[5] of Reichstein's Substance E diacetate showed no depression.

17(α)-Hydroxycorticosterone (XV).—A solution of 0.8 g. of the disemicarbazone (VIII) in 5 cc. of dimethylformamide and 10 cc. of tetrahydrofuran was added dropwise to a stirred solution of 0.5 g. of lithium borohydride in 25 cc. of tetrahydrofuran. The temperature was maintained at 25° and stirring was continued for two hours after addition was complete. At the end of this period the excess lithium borohydride was decomposed with 50 cc. of 10% aqueous acetic acid and the resulting clear solution was concentrated *in vacuo* nearly to dryness. Trituration of the residue with water gave a colorless solid which was filtered, washed with water and dried first at room temperature and finally at 70° for two hours. This crude reduction product (m.p. over 340°) was reacetylated as described above, with 10 cc. of pyridine and 10 cc. of acetic anhydride at 90° for ten minutes. The solvents were evaporated *in vacuo*, replaced by methanol and the methanol solution treated with Norite. The colorless filtrate from this treatment gave an evaporation of the solvent 0.59 g. of material. To this reacetylated reduction product were added 7.5 cc. of glacial acetic acid, 2.5 cc. of water, 1.28 g. of anhydrous sodium acetate and 1.2 cc. of 90% aqueous pyruvic acid. The mixture was heated in a nitrogen atmosphere at 75° for four hours. At the end of this period of heating the mixture was concentrated *in vacuo* at 50° nearly to dryness. Water was added and the organic material was extracted with ethyl acetate. The ethyl acetate solution was washed with water, 5% aqueous sodium bicarbonate, treated with anhydrous sodium sulfate and Norite and filtered. The colorless filtrate was concentrated to a small volume and seeded. In this way there was obtained 75 mg. of colorless sandy crystals, m.p. 211–216°. Recrystallization first from ethyl acetate and finally from acetone gave pure material, m.p. 219–220°, which was not depressed on admixture with authentic 17-(α)-hydroxycorticosterone acetate[1] $\lambda_{max.}^{C_2H_5OH}$ 2425 Å., $E_{1\,cm.}^{1\%}$ 380.

Anal. Calcd. for $C_{23}H_{32}O_6$: C, 68.29; H, 7.98. Found: C, 68.56; H, 8.17.

Corticosterone Acetate (XVI).—11-Dehydrocorticosterone 3,20-bis-semicarbazone (XII) was transformed to corticosterone acetate (XVI) in the same manner described above for the conversion of (VIII) to (XV). After chromatography of the final reaction product a very small amount of (XVI) was obtained as colorless crystals from acetone–ether, m.p. 147–152°.[8] A mixed melting point of this material with a sample prepared by acetylation of authentic corticosterone[9] showed no depression.

(8) Reichstein (*Helv. Chim. Acta*, **20**, 953 (1937)), observed that this compound gave a double melting point at 145–146.5° and 152.5–153°.

(9) We are indebted to the Worcester Foundation for Experimental Biology, Shrewsbury, Mass., for an authentic sample of corticosterone.

RAHWAY, NEW JERSEY RECEIVED MARCH 19, 1951

[JOINT CONTRIBUTION FROM THE INSTITUTO DE QUÍMICA DE LA UNIVERSIDAD NACIONAL AUTÓNOMA DE MÉXICO AND THE RESEARCH LABORATORIES OF SYNTEX, S. A.]

Steroidal Sapogenins. IX.[1] Oxidation of $\Delta^{5,16,20(22)}$-Furostatriene-3β,26-diol[2]

BY A. SANDOVAL, J. ROMO, G. ROSENKRANZ, ST. KAUFMANN AND CARL DJERASSI

$\Delta^{5,16,20(22)}$-Furostatriene-3β,26-diol diacetate (ψ-kryptogenin diacetate) (IIb) is oxidized with chromium trioxide to $\Delta^{5,17(20)}$-cholestadiene-3β,26-diol-16,22-dione diacetate (IV), which on saponification leads to 22,26-oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,22-diol-16-one (VIIa). Both IV and VIIa yield the pyridazine derivative V of Δ^5-cholestene-3β,26-diol-16,22-dione (kryptogenin) when refluxed with hydrazine. Lithium aluminum hydride reduction of VII gives $\Delta^{5,17(20)}$-22-isospirostadien-3β-ol (Xa).

As pointed out in an earlier article,[3] Δ^5-cholestene-3β,26-diol-16,22-dione (kryptogenin) (I) occurs in a number of species of Mexican *Dioscoreae* and is obtained in appreciable amounts as a by-product in the commercial extraction of such plants. The conversion of I, by reductive methods,[3] to $\Delta^{5,20(22)}$-furostadiene-3β,26-diol (ψ-diosgenin) (III) affords one path for utilizing Δ^5-cholestene-3β,26-diol-16,22-dione (I) for the production of steroid hormones. Since this sapogenin is readily transformed[4] into $\Delta^{5,16,20(22)}$-furostatriene-3β,26-diol (ψ-kryptogenin) (II), it was of interest to study the behavior of this substance toward oxidizing agents and compare it with that of its 16,17-dihydro derivative III (ψ-diosgenin). As has been discovered by Marker,[5] the latter substance (III)

(1) Paper VIII, C. Djerassi, J. Romo and G. Rosenkranz, *J. Org. Chem.*, **16**, 754 (1951).

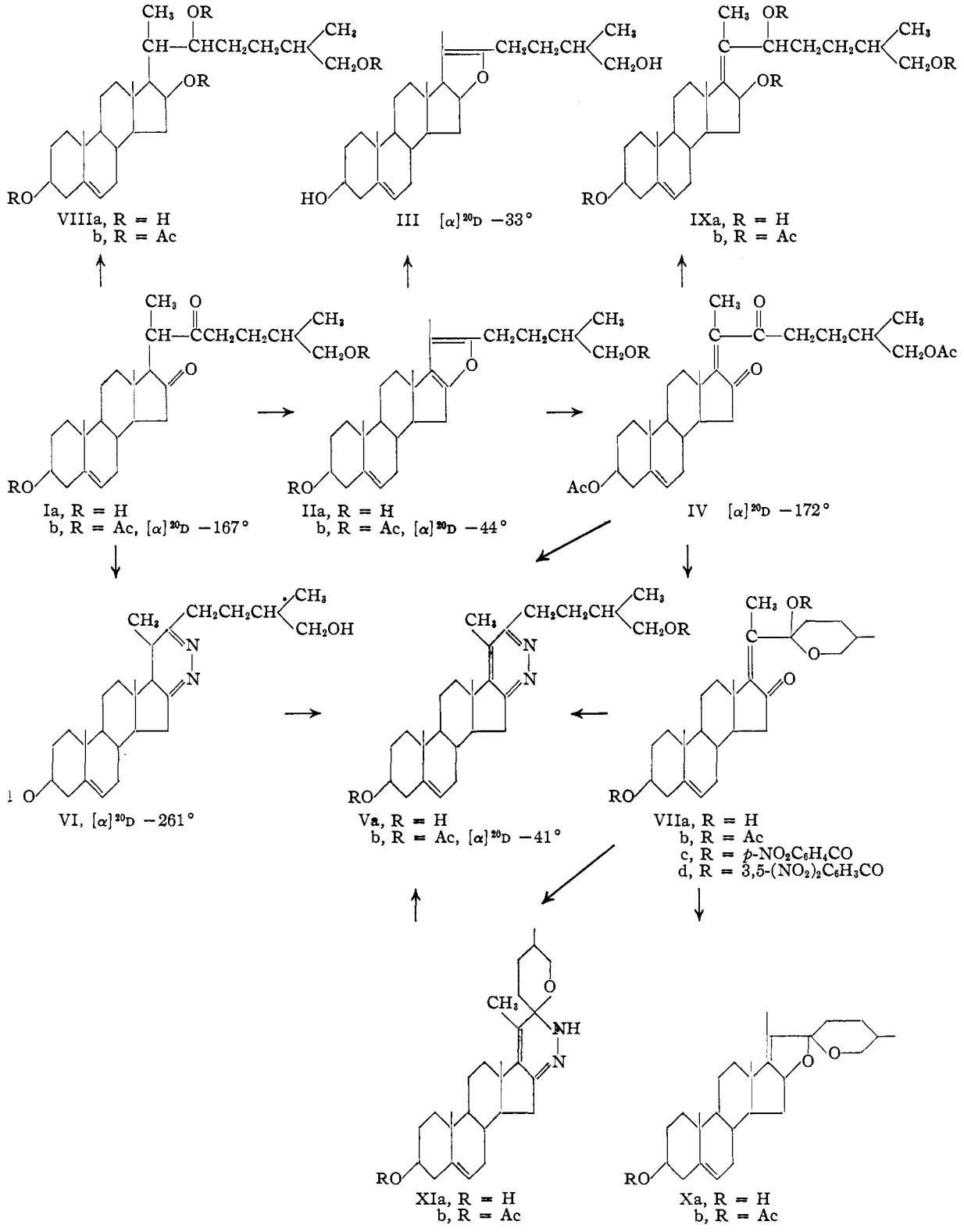
(2) For nomenclature of steroidal sapogenins see G. Rosenkranz and C. Djerassi, *Nature*, **166**, 104 (1950).

(3) St. Kaufmann and G. Rosenkranz, THIS JOURNAL, **70**, 3502 (1948).

(4) (a) R. E. Marker, R. B. Wagner, P. R. Ulshafer, E. L. Wittbecker, D. P. J. Goldsmith and C. M. Ruof, *ibid.*, **69**, 2200 (1947); (b) St. Kaufmann and G. Rosenkranz, U. S. Patent 2,535,073.

(5) R. E. Marker, THIS JOURNAL, **62**, 3350 (1940).

FLOWSHEET

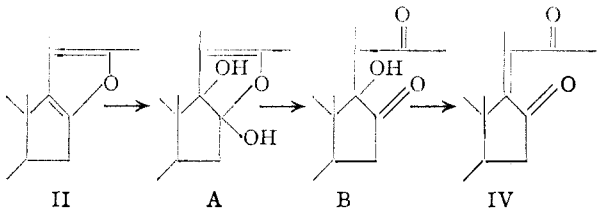


on chromium trioxide oxidation suffers oxidative fission of the 20–22 bond and after hydrolysis leads to $\Delta^{5,16}$-pregnadien-3β-ol-20-one, a convenient starting material for the preparation of the corpus luteum hormone progesterone.

Oxidation (one equivalent of oxygen) of $\Delta^{5,16,20(22)}$-furostatriene-3β,26-diol diacetate (IIb) ([α]^{20}D −44°, ultraviolet maximum at 226 mμ) with chromium trioxide in acetic acid at room temperature produced in nearly 70% yield a substance with m.p. 110–111°, [α]^{20}D −172°, ultraviolet absorption maximum at 246 mμ (log ϵ 4.13),

whose analysis agreed with the empirical formula $C_{31}H_{44}O_6$. The strongly negative rotation of the oxidation product was similar to that of Δ^5-cholestene-3β,26-diol-16,22-dione diacetate (Ib) ($[\alpha]^{20}D$ $-167°$), while the ultraviolet absorption spectrum indicated the presence of an α,β-unsaturated carbonyl system. When the oxidation product was refluxed with hydrazine hydrate in ethanol solution, there was obtained in excellent yield the pyridazine derivative Va of Δ^5-cholestene-3β,26-diol-16,22-dione, which had previously been synthesized[6] by condensation of Δ^5-cholestene-3,26-diol-16,22-dione (I) with hydrazine hydrate and dehydrogenation of the resulting dihydropyridazine VI. The fact that the pyridazine V was formed directly from the oxidation product without the intermediate formation of the dihydro compound VI clearly establishes its constitution as that of $\Delta^{5,17(20)}$-cholestadiene-3β,26-diol-16,22-dione diacetate (17,20-dehydrokryptogenin diacetate) (IV). The structure of IV was confirmed by zinc dust reduction to Δ^5-cholestene-3β,26-diol-16,22-dione diacetate (Ib). The striking changes in the optical rotation of compounds I–VI[7] are given in the accompanying flowsheet and illustrate the utility of rotatory data in this series. It is of interest to note that the ultraviolet absorption spectra (see experimental section) of the pyridazine (V) and dihydropyridazine (VI) derivatives differ markedly.

The formation of the unsaturated diketone IV from the oxidation of the furan derivative IIb can be rationalized as follows. It has already been demonstrated by the behavior toward catalytic hydrogenation[3] that the 16–17 double bond of the furostatriene II is the most reactive one and it is not unexpected that oxidative attack with a limited amount of reagent should occur in that position. A dihydroxy intermediate (A), possibly arising initially from an oxide, constitutes a ketal system which should be quite labile toward hydrolysis and the resulting hydroxydiketone (B), undoubtedly, is dehydrated quite readily to afford the unsaturated diketone IV.



II A B IV

Saponification of the diacetate IV produced an alcohol with the expected composition $C_{27}H_{40}O_4$ but a markedly lower ultraviolet absorption spectrum at 236 mμ (log ϵ 4.17). Reacetylation produced a diacetate, different from IV, which also exhibited an ultraviolet maximum at 236 mμ. These observations are in agreement with the formulation VII (22,26-oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,22-diol-16-one) for the saponification product. The change in the ultraviolet absorption spectrum is strictly analogous to that observed with Δ^4-cholesten-3-one (max. at 242 mμ) and Δ^4-cholestene-3,6-dione (max.[8] at 252 mμ). Treatment of the hemiketal VIIa with hydrazine in ethylene glycol solution produced the pyridazine derivative V, identical with that previously obtained from both I and IV. When the reaction with hydrazine was carried out in ethanol solution, there was isolated a substance which appeared to be an isomeric dihydropyridazine (XI), but when this material was refluxed with ethylene glycol it was converted smoothly into the pyridazine V.

The above structure assignments are supported further by reduction experiments with lithium aluminum hydride. Δ^5-Cholestene-3β,26-diol-16,22-dione diacetate (Ib) and the 17,20-dehydro derivative IV yield the corresponding tetrols VIIIa and IXa, which were characterized by formation of tetraacetates. On the other hand, similar treatment of 22,26-oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,22-diol-16-one (VII) produced an alcohol to which is assigned tentatively the structure of $\Delta^{5,17(20)}$-22-isospirostadien-3β-ol (Xa), since it formed only a monoacetate, which exhibited no hydroxyl band in the infrared.[9] 22,26-Oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,16,22-triol, the expected initial product in the lithium aluminum hydride reduction of VIIa, apparently is dehydrated very readily, as is to be anticipated by analogy to the behavior[3] of its 17,20-dihydro derivative ("16-dihydrokryptogenin"). The substance (Xa) showed no carbonyl bands in the infrared[9] nor did it exhibit selective absorption in the ultraviolet.

Experimental[10]

$\Delta^{5,16,20(22)}$-Furostatriene-3β,26-diol (ψ-Kryptogenin) (IIa).—This substance, m.p. 189–192°, was obtained by Marker, *et al.*,[4a] in 24% yield by heating Δ^5-cholestene-3β,26-diol-16,22-dione diacetate (Ib) with acetic anhydride at 200°. The following procedure[4b] is preferable: A solution of 256 g. of the diacetate Ib and 2.6 g. of *p*-toluenesulfonic acid in 770 cc. of acetic anhydride was refluxed for one hour, poured into water, extracted with benzene and washed free of acid with carbonate solution. The residue was dissolved in 2 l. of methanol and refluxed with a solution of 100 g. of potassium hydroxide in 100 cc. of water for one-half hour. After cooling, the crystals were collected and recrystallized once from benzene–methanol; yield 151 g. (62%), m.p. 189–191°, $[\alpha]^{20}D$ $-39°$, ultraviolet absorption maximum at 226 mμ (log ϵ 4.12). The **diacetate IIb** was obtained in nearly quantitative yield on boiling IIa with acetic anhydride for one hour. Two recrystallizations from methanol afforded the analytical sample with m.p. 94–95° (Kofler), $[\alpha]^{20}D$ $-44°$, ultraviolet absorption maximum at 226 mμ (log ϵ 4.16). The melting point is in agreement with that recorded for the diacetate Ib prepared by the conventional acetic anhydride method[3] but not with the one reported by Marker.[4a]

$\Delta^{5,17(20)}$-Cholestadiene-3β,26-diol-16,22-dione 3,26-Diacetate (IV).—To a vigorously stirred solution of 100 g. of $\Delta^{5,16,20(22)}$-furostatriene-3β,26-diol diacetate (IIb) in 800 cc. of glacial acetic acid was added over a period of one-half hour 13.5 g. of chromium trioxide dissolved in 15 cc. of water and 400 cc. of acetic acid. The temperature was

(6) G. Rosenkranz, St. Kaufmann, A. Landa, J. J. Corona and A. Olalde, THIS JOURNAL, **70**, 3518 (1948).

(7) The rotation ($[\alpha]^{20}D$ $-103°$) of $\Delta^{5,20(22)}$-furostadiene-3β,26-diol (III) given earlier (ref. 2) is in error.

(8) H. Dannenberg, *Abhandl. Preuss. Akad. Wissensch.*, No. 21 (1940), p. 30.

(9) We are indebted to Dr. K. Dobriner and Mrs. P. Humphries of the Sloan-Kettering Institute for Cancer Research for this information.

(10) Melting points marked "Kofler" are corrected and were determined in the Kofler block. Unless indicated otherwise, rotations were determined in chloroform and ultraviolet absorption spectra in 95% ethanol solution. We are indebted to Srta. Paquita Revaque for these measurements and to the Srtas. Amparo Barba and Raquel Cervera for the microanalyses. Acknowledgment is due to Sr. L. Miramontes for preparing certain comparison samples.

maintained at 12–15° during the addition and the mixture was then left at room temperature for an additional two hours. The product was isolated by dilution with water, extraction with ether, washing until neutral with water and sodium bicarbonate solution and evaporating to dryness. Crystallization from methanol yielded 60–70 g. (58–68%) of colorless solid with m.p. 108–110°. The analytical sample crystallized from hexane–benzene; m.p. 110–111° (Kofler), $[\alpha]^{20}D$ −171.7° (chloroform), $[\alpha]^{20}D$ −166.6° (dioxane), ultraviolet absorption maximum at 246 mμ (log ϵ 4.13).

Anal. Calcd. for $C_{31}H_{44}O_6$: C, 72.62; H, 8.65. Found: C, 72.70, 72.47; H, 8.66, 8.52.

When 10 g. of the diacetate IV was refluxed with 250 u. of acetic acid and 40 g. of zinc dust for seven hours, there was isolated in nearly 40% yield Δ^5-cholestene-3β,26-diol-16,22-dione diacetate (Ib).

22,26-Oxido-$\Delta^{5,17(20)}$-Cholestadiene-3β,22-diol-16-one (VIIa).—The saponification of the above diacetate IV (30 g.) was carried out by refluxing for one hour with 15 g. of potassium hydroxide and 1.5 l. of methanol and then diluting with water. Filtration yielded 22.7 g. of crystals with m.p. 220–223°; recrystallization from methanol–ethyl acetate afforded the analytical sample with m.p. 225–226° (Kofler), $[\alpha]^{20}D$ −173°, ultraviolet absorption maximum at 236 mμ (log ϵ 4.17). The same product was obtained when the saponification was carried out with bicarbonate solution.

Anal. Calcd. for $C_{27}H_{40}O_4$: C, 75.66; H, 9.41. Found: C, 75.48; H, 9.48.

The diacetate VIIb was produced by acetylating VIIa at room temperature (one day) with pyridine and acetic anhydride. When crystallized from hexane–ether, the product had m.p. 122–123° (Kofler), while a m.p. of 85–87° (Kofler) was observed when the material was recrystallized from methanol. Both forms exhibited $[\alpha]^{20}D$ −160° and an ultraviolet absorption maximum at 236 mμ (log ϵ 4.19).

Anal. Calcd. for $C_{31}H_{44}O_6$: C, 72.62; H, 8.65. Found: C, 72.44; H, 8.53.

The bis-*p*-nitrobenzoate VIIc was obtained in an analogous manner with *p*-nitrobenzoyl chloride in pyridine solution and was recrystallized from ethanol–benzene; m.p. 185–187° (Kofler), $[\alpha]^{20}D$ −104°.

Anal. Calcd. for $C_{41}H_{46}O_{10}N_2$: C, 67.75; H, 6.38. Found: C, 67.87; H, 6.37.

The bis-3,5-dinitrobenzoate VIId, crystallized from ethyl acetate–methanol, exhibited m.p. 164–166°, $[\alpha]^{20}D$ −94°.

Anal. Calcd. for $C_{40}H_{44}O_{14}N_4$: C, 59.69; H, 5.51. Found: C, 59.63; H, 5.24.

Pyridazine Derivative of Δ^5-Cholestene-3β,26-diol-16,22-dione (V) (a) From $\Delta^{5,17(20)}$-Cholestadiene-3β,26-diol-16,-22-dione Diacetate (IV).—A solution of 10 g. of the diacetate IV in 100 cc. of ethanol and 25 cc. of 85% hydrazine hydrate was refluxed for four hours and diluted with water. The precipitate, obtained in nearly quantitative yield, was recrystallized twice from ethyl acetate, yielding 6.7 g. of colorless crystals with m.p. 221–223° (Kofler), $[\alpha]^{20}D$ −50°, ultraviolet absorption maxima[11] at 258 mμ (log ϵ 3.33) and 296 mμ (log ϵ 2.65). A comparison sample of the pyridazine Va prepared by refluxing "kryptogenin dihydropyridazine (VI)" in nitrobenzene solution[6,12] possessed the same physical constants and a mixture of the two specimens showed no depression of the melting point.

Anal. Calcd. for $C_{27}H_{40}O_2N_2$: N, 6.59. Found: N, 6.74.

One gram of the pyridazine Va was heated at 100° for four hours with 2 cc. of pyridine and 20 cc. of acetic anhydride. Dilution with water afforded 1.1 g. of the **diacetate Vb** (m.p. 186–188°), which on recrystallization from ethyl acetate showed m.p. 192–193° (Kofler), $[\alpha]^{20}D$ −41°, ultraviolet absorption maxima at 258 mμ (log ϵ 3.30) and 294 mμ (log ϵ 2.61).

Anal. Calcd. for $C_{31}H_{44}O_4N_2$: N, 5.51. Found: N, 5.76.

The product proved to be identical in all respects with a comparison sample prepared from Δ^5-cholestene-3β,26-diol-16,22-dione (I). Saponification of the diacetate Vb with methanolic potassium bicarbonate solution regenerated the free diol Va.

(b) From 22,26-Oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,22-diol-16-one (VIIa).—When a solution of 1.0 g. of the hemiketal VIIa was refluxed with 20 cc. of ethylene glycol and 0.6 cc. of 85% hydrazine hydrate for 45 minutes, there was obtained 0.85 g. of the pyridazine derivative Va identical in all respects (mixed m.p., $[\alpha]^{20}D$, spectrum) with specimens prepared from I or IV.

On carrying out the reaction in ethanol solution (4 g. of VIIa, 10 cc. of hydrazine of hydrate, 40 cc. of ethanol) for eight hours, there was obtained 2.3 g. of a product, m.p. 213–215° (Kofler), $[\alpha]^{20}D$ −228° (dioxane), maxima at 238 mμ (log ϵ 4.30) and 256 mμ (log ϵ 4.25), which appeared to be a dihydropyridazine derivative (XIa). The strongly negative rotation is also suggestive of such a formulation, but completely satisfactory analytical results could not be obtained.

Anal. Calcd. for $C_{27}H_{40}O_2N_2$: C, 76.37; H, 9.50. Found: C, 75.46; H, 9.39.

Acetylation with refluxing acetic anhydride–pyridine led to a monoacetate (XIb) with m.p. 198–200°, maxima at 238 mμ (log ϵ 4.34) and 256 mμ (log ϵ 4.29).

Anal. Calcd. for $C_{29}H_{42}O_3N_2$: C, 74.64; H, 9.07; N, 6.00. Found: C, 74.57; H, 9.07; N, 5.84.

The dihydropyridazine XIa was converted into the pyridazine Va after refluxing in ethylene glycol solution for 45 minutes.

Δ^5-Cholestene-3β,16,22,26-tetrol (VIIIa).—A mixture of 5 g. of Δ^5-cholestene-3β,26-diol-16,22-dione diacetate (Ib) (m.p. 151–154°, $[\alpha]^{20}D$ −167°), 2 g. of lithium aluminum hydride and 120 cc. of tetrahydrofuran was refluxed for one hour. After working up as usual, the residue was dissolved in chloroform and passed through a short column of alumina. Evaporation of the eluate and recrystallization from a mixture of ethyl acetate (slightly soluble) and methanol (very soluble) afforded 3.2 g. of the analytical sample with m.p. 205–207° (Kofler), $[\alpha]^{20}D$ −29.5° (dioxane). The substance exhibited no selective absorption in the ultraviolet.

Anal. Calcd. for $C_{27}H_{46}O_4$: C, 74.61; H, 10.67. Found: C, 74.63; H, 10.72.

The tetraacetate VIIIb exhibited m.p. 111–113° (Kofler), $[\alpha]^{20}D$ +5° after recrystallization from methanol.

Anal. Calcd. for $C_{35}H_{54}O_8$: C, 69.74; H, 9.03. Found: C, 69.63; H, 9.16.

$\Delta^{5,17(20)}$-Cholestadiene-3β,16,22,26-tetrol (IXa).—The reduction was carried out by refluxing 7.0 g. of $\Delta^{5,17(20)}$-cholestadiene-3β,26-diol-16,22-dione diacetate (IV) with 5 g. of lithium aluminum hydride and 150 cc. of tetrahydrofuran for one hour, decomposing the excess reagent with acetone, adding water and dilute acid, and filtering the insoluble product. A small sample of the tetrol IXa was purified by recrystallization from benzene whereupon it showed m.p. 222–225°, $[\alpha]^{20}D$ +5.7° (pyridine), no selective absorption in the ultraviolet.

Anal. Calcd. for $C_{27}H_{44}O_4$: C, 74.95; H, 10.25. Found: C, 75.19; H, 10.46.

The rest of the material was acetylated by refluxing for one hour with 4 cc. of pyridine and 35 cc. of acetic anhydride and then purified by chromatographing over 150 g. of ethyl acetate–washed alumina. The benzene–hexane (1:1) eluates on evaporation and recrystallization from methanol–chloroform yielded 4.3 g. of the **tetraacetate IXb** with m.p. 174–175°. The analytical sample possessed the following constants: m.p. 180–181° (Kofler), $[\alpha]^{20}D$ +20°, no selective absorption in the ultraviolet. Saponification regenerated IXa.

Anal. Calcd. for $C_{35}H_{52}O_8$: C, 69.97; H, 8.73; acetyl, 28.66. Found:[13] C, 69.89; H, 8.81; acetyl, 28.49.

$\Delta^{5,17(20)}$-22-Isospirostadien-3β-ol (Xa).[14]—The lithium aluminum hydride reduction of 22,26-oxido-$\Delta^{5,17(20)}$-cholestadiene-3β,22-diol-16-one (VIIa) was carried out exactly

(11) The spectrum of 3-methylpyridazine in ethanol exhibits two bands at 251 mμ (log ϵ 3.11) and 310 mμ (log ϵ 2.60) [W. G. Overend, L. M. Turton and L. F. Wiggins, *J. Chem. Soc.*, 3500 (1950)].

(12) The dihydropyridazine derivative (VI) of Δ^5-cholestene-3β,26-diol-16,22-dione was previously (ref. 6) not characterized. By refluxing I with 85% hydrazine hydrate in ethanol solution for two hours and recrystallizing from methanol, the dihydropyridazine VI could be obtained in 75% yield with m. p. 155–159° (Kofler), $[\alpha]^{20}D$ −260.6°, ultraviolet absorption maxima at 232 mμ (log ϵ 3.86) and 286 mμ (log ϵ 1.83). *Anal.* Calcd. for $C_{32}H_{42}O_2N_2$: C, 75.99; H, 9.92; N, 6.57. Found: C, 75.82; H, 9.56; N, 6.85.

(13) This analysis was carried out by Mr. Joseph F. Alicino, Metuchen, N. J.

(14) The configuration of C-22 has not been proved, but is only assumed by analogy to the conversion of "16-dihydrokryptogenin" to Δ^5-22-isospirosten-3β-ol (ref. 3).

as above and led in 82% yield to the alcohol Xa. Recrystallization from methanol–chloroform gave the analytical sample with m.p. 236–237° (Kofler), $[\alpha]^{20}D$ −81.3°. The substance showed no selective absorption in the ultraviolet and exhibited no carbonyl bands in the infrared.[9] It was recovered unchanged on treatment with hydrazine hydrate.

Anal. Calcd. for $C_{27}H_{40}O_3$: C, 78.59; H, 9.77. Found: C, 78.92; H, 9.66.

The acetate Xb, m.p. 260–262° (Kofler), $[\alpha]^{20}D$ −70.7°, after recrystallization from chloroform—methanol, showed no hydroxyl band in the infrared.[9]

Anal. Calcd. for $C_{29}H_{42}O_4$: C, 76.61; H, 9.31. Found: C, 76.33; H, 9.01.

LAGUNA MAYRAN 413
MEXICO CITY 17, D. F.

RECEIVED FEBRUARY 19, 1951

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF S. B. PENICK & COMPANY]

The Glycosides of the Seeds of *Strophanthus intermedius* Pax.

BY M. R. SALMON, ERIC SMITH AND W. G. BYWATER

The seeds of *Strophanthus intermedius* were found to contain two crystalline cardioactive glycosides that appear to be identical with those previously isolated by v. Euw and Reichstein[1] from *Strophanthus gerrardii* and provisionally called Substance 761 and Substance 762. We find that Substance 761 is a glycoside of sarverogenin.[2]

The Upjohn–Penick Expedition[3] for African Botanical Exploration in 1949–1950 collected a large series of the seeds of *Strophanthus* species for study of their glycoside content in an effort to make a complete survey of the availability of glycosides containing 11-oxygenated steroid nuclei suitable for conversion to cortisone or related steroids. The elegant work of Reichstein and his group has outlined the general method for isolation of glycosides from *Strophanthus* species, details of which have been described.[1,2,4] One species of *Strophanthus* which has not previously been studied in detail has now been subjected to the general isolation technique developed in Professor Reichstein's laboratories. This species was identified as *Strophanthus intermedius* (B&W No. 20932) by Mr. Joseph Monachino of the New York Botanical Garden. The seeds were collected in northwestern Angola.

The seeds were worked up according to the general procedure of Katz.[4a] The resulting aqueous solution of the total glycosides was extracted successively with ether and with chloroform. The glycosides remaining in the aqueous phase after extraction have not yet been examined.

The ether extract on concentration gave 350 mg. of crystalline material which agreed in properties with Reichstein's Substance 761 except for a slightly higher rotation. In addition there was 389 mg. of oily glycosides that did not yield crystalline material after chromatographic adsorption.

The chloroform extract yielded 6.25 g. of total glycosides which, when crystallized from methanol, gave 3.0 g. of white crystals. The properties of the crystalline material did not change significantly on recrystallization but an attempt to prepare the aglycone from a small portion showed conclusively that it was a mixture of an easily hydrolyzable and a difficultly hydrolyzable glycoside. Accordingly, the crystalline mixture was chromatogramed and there was isolated 636 mg. of Reichstein's Substance 761 and 392 mg. of Reichstein's Substance 762, and other crystalline fractions which may have been mixtures of these two.

The oily mother liquors from the 3 g. of crystalline glycosides were separately chromatogramed and we found some chromatogram fractions with a small negative rotation, suggesting the presence of sarmentocymarin, but we were unable to isolate this glycoside.

The total yield of ether- and chloroform-soluble components from *Strophanthus intermedius* seeds was 4.5%, of which 2.2% was crystalline glycosides.

The preparation identified as Reichstein's Substance 761 on hydrolysis with dilute acid gave sarverogenin, characterized as the crystalline benzoate. Since Reichstein's Substance 761 appears to be isomeric with sarveroside, the sugar portion of the molecule is probably isomeric with sarmentose. We have not examined the sugar formed on hydrolysis.

The occurrence of Reichstein's Substance 761 in the seeds of *Strophanthus intermedius* has been announced in a footnote (reference 1, page 525). The simultaneous occurrence of Reichstein's Substance 762 and the characterization of Substance 761 as a glycoside of sarverogenin have not previously been reported. In addition to *Strophanthus gerrardii* and *S. intermedius*, Substance 761 and Substance 762 have been found in *S. courmonti*[4b] and Substance 762 has been found in *S. sarmentosus*.[4g]

We are indebted to Professor T. Reichstein who has examined our two crystalline glycosides and reported to us that the colors formed with 84% sulfuric acid are identical with the corresponding glycosides prepared in his laboratory and there is no depression in melting point when corresponding

(1) J. v. Euw and T. Reichstein, *Helv. Chim. Acta*, **33**, 522 (1950).

(2) A. Buzas, J. v. Euw and T. Reichstein, *ibid.*, **33**, 465 (1950).

(3) An expedition sponsored jointly by The Upjohn Company of Kalamazoo, Michigan, and S. B. Penick & Company of New York, of which Mr. L. J. Brass of the Archbold Expeditions unit of The American Museum of Natural History was field director and Mr. E. F. Woodward of S. B. Penick & Company was pharmacognosist and business manager. Herbarium specimens of all the materials collected by the expedition have been submitted to the New York Botanical Garden for taxonomic study. Such specimens have been assigned field numbers under the name of Brass and Woodward; for example, one of the *Strophanthus intermedius* samples collected by the expedition is identified as B&W No. 20932. A set of the collection will also be on deposit with the Royal Botanic Gardens, Kew, Surrey, England. When the taxonomic studies are completed it will be possible to identify and locate the species of *Strophanthus*, upon which chemical isolation work is being done in these laboratories, by reference to the appropriate numbers in the collection.

(4) For example (a) A. Katz, *Helv. Chim. Acta*, **31**, 993 (1948); (b) J. v. Euw and T. Reichstein, *ibid.*, **31**, 883 (1948); (c) J. v. Euw and T. Reichstein, *ibid.*, **33**, 544, 666 (1950); (d) A. Lardon, *ibid.*, **33**, 639 (1950); (e) John W. Rothrock, E. E. Howe, Klaus Florey and Max Tishler, THIS JOURNAL, **72**, 3827 (1950); (f) J. v. Euw and T. Reichstein, *Helv. chim. acta*, **33**, 1006 (1950); (g) **33**, 2153 (1950).