(77% yield) of hard crystals, m.p. 158–160° dec. alone or in mixture with an authentic sample.[4a] The infrared spectra for the two samples were identical. The material had $[\alpha]^{24}D$ +71° (*c* 1.06, chloroform) and λ_{max}^{MeOH} 229 mμ (ε 30,800). Reported [4a] values are m.p. 158–159°, $[\alpha]^{25}D$ +69° (chloroform) and λ_{max}^{MeOH} 231 mμ (ε 31,400).

Anal.[4a] Calcd. for $C_{30}H_{36}O_7S$: C, 66.64; H, 6.71; S, 5.93. Found: C, 66.75; H, 6.89; S, 6.18.

Conversion of this compound to pregnatriene III is described above.

PEARL RIVER, N.Y.

[CONTRIBUTION FROM THE ORGANIC CHEMICAL RESEARCH SECTION, LEDERLE LABORATORIES DIVISION, AMERICAN CYANAMID CO.]

Studies in the Synthesis of Triamcinolone. The Condensation of 16α,17α-Isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione with Ethyl Oxalate

BY GEORGE R. ALLEN, JR., AND MARTIN J. WEISS[1]

RECEIVED JANUARY 24, 1959

Ethoxalylation of 16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI), which was prepared from 16α,17α-epoxy-11α-hydroxyprogesterone (II), in the presence of a molar equivalent of sodium methoxide gave exclusively the 2-ethoxalyl derivative XVIII, which was converted by a six-step procedure into 9α-fluoro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII). The 9α-chloro analog of XXVII also was prepared. Bis-ethoxalylation of XI led to the preparation of the 2α,21-dibromide XXII. Attempts to convert XXII into 16α,21-diacetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione were unsuccessful.

The importance of 9α-fluoro-11β,16α,17α,21-tetrahydroxy-1,4-pregnadiene-3,20-dione (I)[2a] (Aristocort triamcinolone[2b]) as a therapeutic agent[3] made it desirable to explore other routes for the synthesis of this material.[4] An attractive starting point for a synthesis of I is 16α,17α-epoxy-11α-hydroxyprogesterone (II) which has been obtained from 16α,17α-epoxyprogesterone by fermentation with *Rhizopus nigricans*.[5] In the present paper we wish to describe the conversion of II into 16α,17α-dihydroxy-4,9(11)-pregnadiene-3,20-dione (X), and efforts to transform the latter material and certain of its derivatives into intermediates that have been utilized for the preparation of I.[9]

Our preparation of X was accomplished in the following manner. Reaction of II with methanesulfonyl (mesyl) chloride gave the 11α-mesyloxy derivative III from which 16α,17α-epoxy-4,9(11)-pregnadiene-3,20-dione (IV) was prepared by elimination of the elements of methanesulfonic acid. This procedure, which is reported in the patent literature by Bergstrom,[11] in our hands has afforded a consistent over-all yield of 80%. The conversion of the 16α,17α-epoxy group of IV into a 16α,17α-diol system was accomplished by using the procedure of Romo and Romo de Vivar.[12] Reaction of the oxide IV with hydrogen bromide in glacial acetic acid gave the bromohydrin V in 62% yield. Acetylation of this material with acetic anhydride in the presence of *p*-toluenesulfonic acid afforded the enol diacetate VII in 95% yield. Treatment of VII with sodium acetate in refluxing acetic acid produced 16α-acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX) in 57% yield. This step, wherein the 16β-bromo group is displaced, involves participation of the neighboring 17-acetoxy group, presumably *via* an intermediate *ortho* ester type cation.[12] The diol acetate IX also was prepared by two variations of the above method. Reaction of the oxide IV with sodium iodide in glacial acetic acid gave in 69% yield the iodohydrin VI[13] which was converted into IX *via* the enol acetate VIII. Finally, IX also was obtained from 16α,17α-epoxy-11α-mesyloxyprogesterone (III). Reaction of this substance with hydrogen bromide in glacial acetic acid gave XII in almost quantitative yield. The bromohydrin XII was converted into the enol diacetate XIII which, without isolation, was treated with sodium acetate in refluxing acetic acid to give IX in 39% yield. Of the above three methods, the last gives the most satisfactory yield, whereas the method based on the iodohydrin VI gives the poorest. Hydrolysis of the diol acetate IX with 0.5 *N* hydrochloric acid gave 16α,17α,dihydroxy-4,9(11)-

(1) To whom inquiries concerning this paper should be addressed.

(2) (a) S. Bernstein, R. H. Lenhard, W. S. Allen, M. Heller, R. Littell, S. M. Stolar, L. I. Feldman and R. H. Blank, THIS JOURNAL, **78**, 5693 (1956); **81**, in press (1959); (b) the trademark of American Cyanamid Co. for triamcinolone is Aristocort.

(3) L. Hellman, B. Zumoff, M. K. Schwartz, T. F. Gallagher, C. A. Berntsen and R. H. Freyberg, Abstracts of papers presented at the 3rd Interim Meeting of the American Rheumatism Association, Bethesda, Md., November 30, 1956.

(4) The original method for the preparation of I utilized 21-acetoxy-4,9(11),16-pregnatriene-3,20-dione[1,5] as the key intermediate. This latter substance has been prepared from 21-acetoxy-17α-hydroxyprogesterone (Reichstein's Substance S)[6] and the bis-ethylene ketal of cortisone acetate.[8] More recently, Fried and his co-workers have described the preparation of I from 9α-fluorohydrocortisone; this elegant procedure utilized microbiological fermentation to introduce the required 16α-hydroxy function.[7]

(5) W. S. Allen and S. Bernstein, THIS JOURNAL, **77**, 1028 (1955).

(6) W. S. Allen, S. Bernstein and R. Littell, *ibid.*, **76**, 6116 (1954).

(7) R. W. Thoma, J. Fried, S. Bonanno and P. Grabowich, *ibid.*, **79**, 4818 (1957).

(8) D. H. Peterson, P. D. Meister, A. Weintraub, L. M. Reineke, S. H. Eppstein, H. C. Murray and H. M. L. Osborn, *ibid.*, **77**, 4428 (1955).

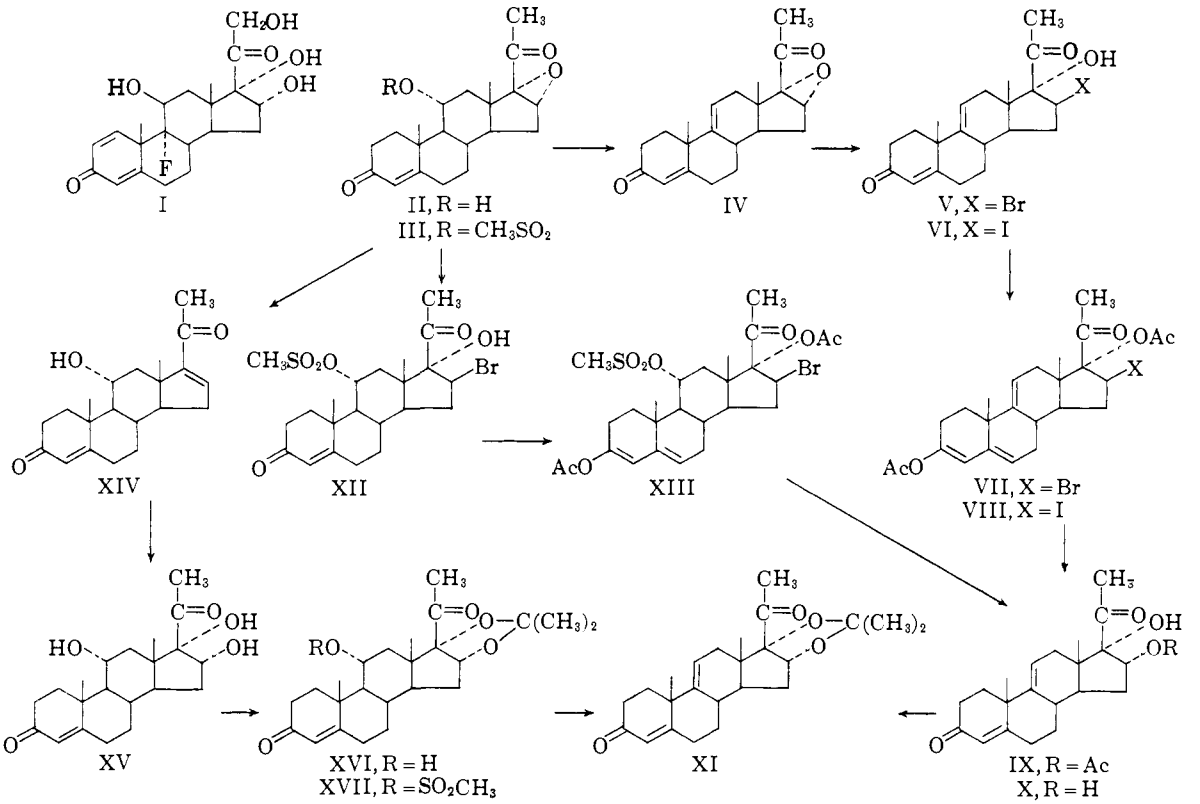
(9) Other efforts to utilize 16α,17α-epoxy-11α-hydroxyprogesterone for the preparation of triamcinolone are the subject of an accompanying paper.[10] The general utility of 16α,17α-epoxy steroids for the synthesis of triamcinolone will be discussed in a forthcoming publication (W. S. Allen, S. Bernstein, L. I. Feldman and M. J. Weiss).

(10) R. E. Schaub, G. R. Allen, Jr., and M. J. Weiss, THIS JOURNAL, **81**, 4962 (1959).

(11) C. G. Bergstrom, U. S. Patent 2,703,799.

(12) J. Romo and A. Romo de Vivar, *J. Org. Chem.*, **21**, 902 (1956).

(13) The opening of 16α,17α-oxides to yield iodohydrins has been reported by A. Ercoli and P. de Ruggieri [*Gazz. chim. ital.*, **84**, 479 (1954)].



pregnadiene-3,20-dione (X) which has been converted into its acetonide derivative (XI).[14,15]

In an alternate preparation of XI, 16α,17α-epoxy-11α-hydroxyprogesterone (II) was treated with chromous chloride[16] to give, in 50% yield, the known[17] 11α-hydroxy-4,16-pregnadiene-3,20-dione (XIV). Hydroxylation of the 16,17-double bond in XIV with osmium tetroxide gave the triol XV in only 12% yield, but with potassium permanganate[18] this transformation was accomplished in 46% yield. The 16α,17α-diol system of XV was blocked with an isopropylidene group in 87% yield, and the 11α-hydroxy function of the resulting XVI was converted into the 11α-mesylate XVII in 69% yield. Elimination of the elements of methanesulfonic acid with sodium acetate in refluxing acetic acid gave a quantitative yield of the desired XI.

16α-Acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX) and the corresponding free alcohol X require only the introduction of a 21-acetoxy function to give intermediates that have already been converted into triamcinolone (I).[1] A useful method for such a conversion is the "ethoxalylation" procedure which was first applied to the introduction of a 21-acetoxy group by Ruschig.[19a] Application of this method to a 21-deoxysteroid which contains a 16-hydroxy or 16-acetoxy substituent has not been reported. When 16α,17α-dihydroxy-4,9(11)-pregnadiene-3,20-dione (X) was treated with 1.7 molar equivalents of ethyl oxalate and 1.1 molar equivalents of sodium methoxide,[19b] no ethoxalyl derivative could be isolated. Only a low yield of starting material (isolated after chromatography as the acetonide XI) was obtained. Since X has been shown to undergo D-homoannulation in the presence of base,[14] it is possible that the remainder of this material suffered from a base-catalyzed rearrangement. Similar treatment of the 16α-acetoxy derivative IX merely resulted in de-acetylation with the formation of the free diol X in 84% yield.

The failure of IX and X to undergo a condensation reaction with ethyl oxalate prompted us to investigate this reaction with the blocked steroid, 16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI), which is not susceptible to D-homoannulation.[20] When XI was allowed to react with 1.7 molar equivalents of ethyl oxalate and 1.1 molar equivalents of sodium methoxide,[19b] the sodium salt of a monoethoxalyl derivative was obtained as an amorphous solid. The free mono-

(14) S. Bernstein, J. J. Brown, L. I. Feldman and N. E. Rigler, THIS JOURNAL, **81**, 4956 (1959).

(15) It should be noted that 16α,17α-dihydroxy-4,9(11)-pregnadiene-3,20-dione (X) may be prepared from the oxide IV in an over-all yield that is superior to that of the above sequence. Treatment of IV with chromous chloride[16] gives 4,9(11),16-pregnatriene-3,20-dione in 80% yield.[14] This material has been converted into X in 90% yield by hydroxylation of the 16,17-double bond with osmium tetroxide.[14] However, in the present investigation we were interested in a sequence that would circumvent the expensive and somewhat hazardous osmium tetroxide.

(16) W. Cole and P. L. Julian, *J. Org. Chem.*, **19**, 131 (1954).

(17) B. J. Magerlein, D. A. Lyttle and R. H. Levin, *ibid.*, **20**, 1709 (1955).

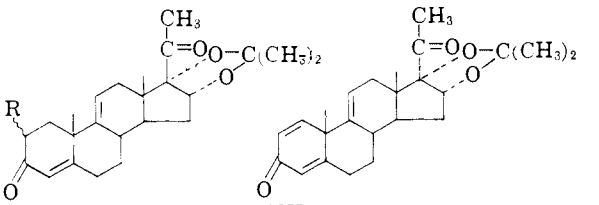
(18) Method of V. Petrow and co-workers [*J. Chem. Soc.*, 4373 (1955)] as modified by L. L. Smith and M. Marx of the Chemical Process Improvement Department of these laboratories.

(19) (a) H. Ruschig, *Ber.*, **88**, 878 (1955); (b) J. A. Hogg, P. F. Beal, A. H. Nathan, F. H. Lincoln, W. P. Schneider, B. J. Magerlein, A. R. Hanze and R. W. Jackson, THIS JOURNAL, **77**, 4436 (1955). Leading references to the patent literature on this subject may also be found in this communication.

(20) We wish to thank Dr. M. D. Heller of these laboratories for helpful discussions on this aspect of the problem.

ethoxalyl derivative of XI was isolated (75% over-all yield from XI) by acidification of an aqueous solution of the sodium salt. Although satisfactory analytical values could not be obtained for the ethoxalyl derivative, treatment with a molar equivalent of bromine followed by brief heating with acetate gave a crystalline monobromo derivative of XI in 95% yield.[21,22] Acetolysis by the iodide–acetate procedure[23] gave only halogen-containing material. The failure of this standard acetolysis procedure to give a 21-acetoxy-steroid indicated that the monoethoxalyl derivative of XI and the corresponding monobromide were not the desired 21-substituted steroids. The precise constitution of these substances was determined in the following manner.

Treatment of the monobromide with boiling collidine gave, in 54% yield, a bromine-free material to which the structure 16α,17α-isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XX) was assigned on the basis of infrared spectral evidence,[24] the ultraviolet absorption maxima of its semicarbazone,[25,26] and polarographic assays. Since triene XX was prepared from diene XI by the sequence ethoxalylation, bromination, deacylation and dehydrobromination, it follows that the monoethoxalyl and monobromo derivatives of XI must be the C-2 substituted compounds (XVIII and XIX, respectively). Assignment of the α-orientation to the bromo group in XIX is predicated on infrared spectral evidence[27a,b] and mechanistic considerations.[27c] It is probable that the preparation of the 2-bromo derivative proceeds *via* the intermediate carbanion ii. This carbanion, which offers ample opportunity for assumption of the more stable conformation (presumably the equatorial 2α),[27c] would result from the base-catalyzed deacylation of the brominated ethoxalyl derivative i.[28]

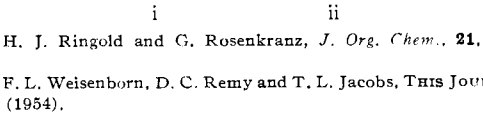


XVIII, R = $COCOOC_2H_5$ XX
XIX, R = α-Br

The availability of position C-2 in 3-ketosteroids for acylation reactions is well known. Other workers have reported the condensation of ethyl oxalate or ethyl formate at position C-2 with testosterone,[29,30] androstane derivatives[29] and cholestenone.[30,31] However, to our knowledge, the formation of the 2-ethoxalyl derivative XVIII constitutes the first known example wherein ethyl oxalate reacts with a progesterone derivative exclusively at position C-2. This result was surprising since previous workers have reported that progesterone,[32] 11-ketoprogesterone,[19b] 11β-hydroxyprogesterone[19b] and 11α-hydroxyprogesterone[19b] react preferentially at position C-21.

Since our efforts to convert the acetonide XI into a Δ^4-3-keto intermediate which had been utilized for the preparation of triamcinolone were thwarted by preferential condensation of ethyl oxalate at position C-2,[33] an attractive alternative was to proceed *via* a 2,21-bis-ethoxalyl derivative to 16α,21-diacetoxy-17α-hydroxy-1,4,9(11)-pregnatriene-3,20-dione which has been converted in four steps to triamcinolone.[1] The formation of 2,21-bis-ethoxalylsteroids[35] and their transformation to 21-acetoxy-$\Delta^{1,4}$-3-ketosteroids[10] had already been achieved at the time of this investigation.

Treatment of the acetonide XI with 3.4 molar equivalents of ethyl oxalate and 2.2 molar equivalents of sodium methoxide[35] gave a crude disodium salt of 2,21-bis-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXI). Preliminary bromination experiments with the anior-



i ii

(21) Bromine is preferable to iodine for the addition of halogen to this ethoxalyl derivative. Treatment of this derivative with a molar equivalent of iodine resulted in an iodine uptake amounting to 84–98% of theory as indicated by titration of an aliquot of the reaction mixture (several experiments). However, despite this high iodine consumption, we were unable to isolate an iodo derivative of good quality.

(22) Subsequently, it was found that heating with acetate could be eliminated without affecting the yield of this monobromo derivative.

(23) G. Rosenkranz, J. Pataki, St. Kaufmann, J. Berlin and C. Djerassi, THIS JOURNAL, **72**, 4081 (1950).

(24) *Cf.* J. Fried, R. W. Thoma and A. Klingsberg, *ibid.*, **75**, 5764 (1953); H. L. Herzog, C. C. Payne, M. A. Jevnik, D. Gould, E. L. Shapiro, E. P. Oliveto and E. B. Hershberg, *ibid.*, **77**, 4781 (1955).

(25) L. Dorfman, *Chem. Revs.*, **53**, 85 (1953).

(26) It is interesting to note that both XX and XI formed only the 3-semicarbazones even though they were treated with excess semicarbazide. This apparently is another example of the steric effect exerted by the 16α,17α-isopropylidenedioxy group on the 17β-acetyl sidechain (see ref. 38). It is possible that the enhancement of biological activity exhibited by the ketal and acetal derivatives of 16α-hydroxylated glucocorticoids [J. Fried, *et al.*, THIS JOURNAL, **80**, 2339 (1958)] is related to the fact that the alkylidenedioxy group sterically hinders the 20-carbonyl function. Since it is known that one of the metabolic pathways for the inactivation of cortisone or hydrocortisone involves reduction of the 20-carbonyl group to an alcohol [E. Caspi, H. Levy and O. M. Hechter, *Arch. Biochem. Biophys.*, **45**, 169 (1953); D. K. Fukushima, Abstr. of 131st A.C.S. Meeting, Miami, Fla., 1957, p. 56C; E. M. Glenn, *et al.*, *Endocrinol.*, **61**, 128 (1957)], it is conceivable that this steric effect may interfere with catabolic inactivation of the molecule. A similar hypothesis has been advanced in connection with the enhancement of biological activity resulting from 16α-methyl substitution [G. E. Arth, *et al.*, THIS JOURNAL, **80**, 3160 (1958)].

(27) (a) M. Fieser, M. A. Romero and L. F. Fieser, *ibid.*, **77**, 3305 (1955); (b) E. G. Cummins and J. E. Page, *J. Chem. Soc.*, 3847 (1957); (c) see E. J. Corey [THIS JOURNAL, **76**, 175 (1954)] for a discussion of the factors determining the relative stability of epimeric α-bromoketones.

(28) In their preparation of certain 2-alkyl substituted steroids from the corresponding ethoxalyl derivatives, Hogg and co-workers [*ibid.*, **77**, 6401 (1955)] concluded that the method of preparation made it likely that the alkyl substituent was α-oriented.

(29) H. J. Ringold and G. Rosenkranz, *J. Org. Chem.*, **21**, 1333 (1956).

(30) F. L. Weisenborn, D. C. Remy and T. L. Jacobs, THIS JOURNAL, **76**, 552 (1954).

(31) J. G. Burr, Jr., W. F. Holton and C. N. Webb, *ibid.*, **72**, 4903 (1950); L. Ruzicka and Pl. A. Plattner, *Helv. Chim. Acta*, **21**, 1717 (1938); Pl. A. Plattner and L. M. Jampolsky, *ibid.*, **24**, 1459 (1941).

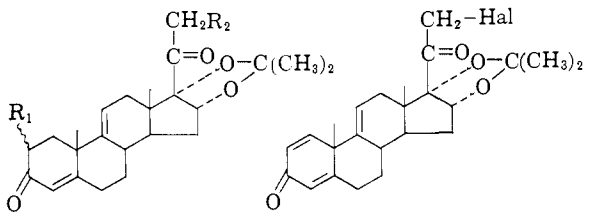
(32) A. H. Nathan and J. A. Hogg, U. S. Patent 2,727,905 (1955); *C. A.*, **50**, 10806g (1956).

(33) Several attempts to introduce the 21-acetoxy function into acetonide XI *via* the recently described[34] modified haloform reaction failed.

(34) H. J. Ringold and G. Stork, THIS JOURNAL, **80**, 250 (1958).

(35) J. A. Hogg, F. H. Lincoln, A. H. Nathan, A. R. Hanze, W. P. Schneider, P. F. Beal and J. Korman, *ibid.*, **77**, 4438 (1955).

phous free bis-ethoxalyl derivative resulted in the isolation of small quantities of the monobromosteroid XIX which was isolated satisfactorily by chromatography. Presumably, the formation of XIX results from the presence of the 2-monoethoxalyl derivative XVIII in the crude bis-ethoxalyl preparation. Since the conversion of XVIII to XIX had been demonstrated already to be essentially quantitative (see above), it was possible on the basis of these preliminary experiments to estimate the amount of monoethoxalyl derivative XVIII present in a given preparation of crude XXI. Hence, reaction of the mixed ethoxalyl derivatives with a calculated theoretical amount (estimated from preliminary experiments) of bromine and treatment of the intermediates with sodium methoxide gave the 2α,21-dibromide XXII (56% over-all yield from XI) and the 2α-bromo derivative XIX (7% over-all yield from XI). Only when the bromination was carried out with a controlled amount of bromine could the crystalline dibromo derivative XXII be isolated.[36] The structure of this dibromide follows from infrared spectral evidence[27] and the known ability of C-2 and C-21 to undergo acylation by ethyl oxalate.

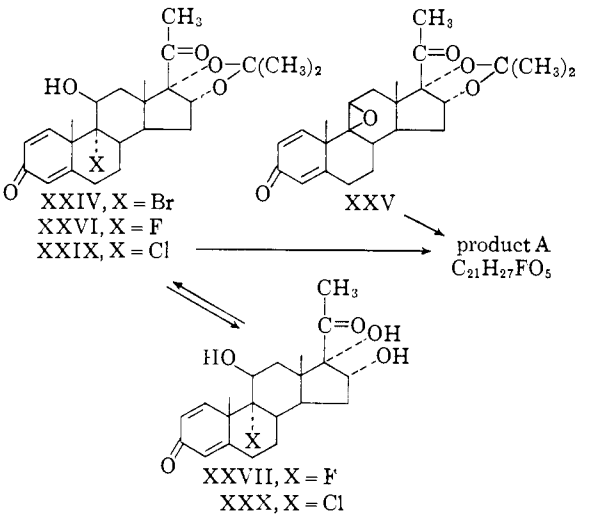


XXI, $R_1 = R_2 =$ COCOOEt XXIII
XXII, $R_1 = \alpha$-Br, $R_2 =$ Br

In an attempt to introduce the 21-acetoxy group, the dibromo derivative XXII was stirred with potassium acetate in acetone at room temperature for three days. This treatment was ineffective, and XXII was recovered to the extent of 92%.[37] Moreover, treatment of XXII with potassium acetate in refluxing acetone gave a 65% recovery; none of the desired 21-acetoxy steroid could be isolated.[38] Since it was believed that more vigorous acetolysis conditions with XXII would cause displacement of the 2α-bromo substituent,[40] the initial introduction of the $\Delta^{1,4}$-3-ketone system by dehydrobromination was investigated.

When the dibromo compound XXII was treated with lithium chloride in dimethylformamide[43] under varying conditions of temperature and time, inseparable mixtures always were obtained. Although the desired 21-halo-$\Delta^{1,4}$-3-ketone XXIII probably was present in these mixtures, the appearance of absorption bands at 6.31 and 6.43 μ in the infrared spectra and at 385 mμ in the ultraviolet spectra of these mixtures indicated that a steroid with a more highly unsaturated chromophore also was present. Treatment of XXII with refluxing collidine gave an extremely low yield of material that appeared to be a mixture on the basis of its elemental analyses and infrared spectrum. (Details of these attempts to introduce the $\Delta^{1,4}$-3-ketone system are given in the Experimental section.) These discouraging results caused the abandonment of efforts to convert dibromide XXII into 16α,21-diacetoxy-17α-hydroxy-1,4,9(11)-pregnatriene-3,20-dione.

The unexpected preparation of 16α,17α-isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XX) (see above) made possible a second synthesis of 9α-fluoro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII),[14] the 21-deoxy analog of triamcinolone. This synthesis proceeded as follows. Reaction of the triene XX with N-bromoacetamide and perchloric acid gave in 65% yield the bromohydrin XXIV, which on treatment with potassium acetate in refluxing ethanol afforded a 95% yield of the 9β,11β-oxide XXV.[44] Reaction of this oxide with anhydrous hydrogen fluoride (17–22 volume per cent.) in tetrahydrofuran pro-



(36) Treatment of 2,21-bis-ethoxalyl steroids with more than two (three) molar equivalents of bromine has been shown to give tribromosteroids.[35]

(37) When 2,21-dibromo-4,9(11),16-pregnatriene-3,20-dione is treated with potassium acetate under these conditions, preferential acetolysis of the 21-bromo group occurs in good yield.[10]

(38) H. Ringold, G. Rosenkranz and F. Sondheimer [THIS JOURNAL, **78**, 820 (1956)] have reported that the bromo substituent in 17α-acetoxy-21-bromopregnenolone does not undergo displacement with potassium acetate at 100°. Similarly, Salamon and Reichstein [*Helv. Chim. Acta*, **30**, 1616 (1947)] were unable to displace the bromo group of 3β,17β-diacetoxy-21-bromoallopregnane-20-one on treatment with acetate. The non-reactivity of XXII and of the above examples toward acetate may be the result of hindrance to the approach of the acetoxy anion at the rear face of C-21. Inspection of molecular models[39] revealed that the number of possible approaches available to the acetoxy anion is restricted markedly by the 16α,17α-isopropylidenedioxy (or 17-acetoxy) substituent. Moreover, the presence of these substituents in a 21-bromosteroid clearly hampers the freedom of rotation about the C_{20}–C_{21} bond. This further minimizes the number of possible approaches to the rear face of C-21. However, it could be demonstrated that under sufficiently vigorous conditions the 21-bromo group was susceptible to displacement, for when XXII was treated with sodium hydroxide in refluxing 80% 2-methoxyethanol for 24 hours, 92% of the bromine was isolated as silver bromide. (For another apparent example of the steric hindrance exerted by the 16α, 17α-isopropylidenedioxy group, see ref. 26.)

(39) Molecular models manufactured by Catalin Ltd., Waltham Abbey, Essex, Eng.

(40) Treatment of 2-bromo-4-androstene-3,17-dione[41] and 2-bromocholestanone[42] with potassium acetate in refluxing acetic acid has been reported to give 2α-acetoxy-4-androstene-3,17-dione and a mixture of 2α-acetoxy- and 4α-acetoxycholestanone, respectively.

(41) G. Rosenkranz, O. Mancera and F. Sondheimer, THIS JOURNAL, **77**, 145 (1955).

(42) L. F. Fieser and M. A. Romero, *ibid.*, **75**, 4716 (1953).

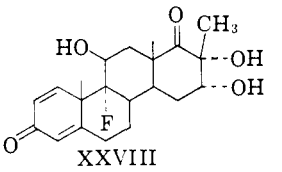
(43) R. P. Holysz, *ibid.*, **75**, 4432 (1953).

(44) J. Fried and E. F. Szabo, *ibid.*, **79**, 1130 (1957).

duced the fluorohydrin acetonide XXVI in 73% yield.[45] Treatment of the oxide XXV with a higher concentration (30 volume per cent.) of anhydrous hydrogen fluoride resulted in the formation of a deacetonated fluorohydrin A. The structure of product A, which was not the desired triol XXVII, will be discussed below. Hydrolysis of the fluorohydrin acetonide XXVI with 0.5 *N* hydrochloric acid in refluxing aqueous methanol for three hours gave a mixture of materials which was resolved successfully by partition chromatography on Celite.[14,46] In this way there was isolated a 44% recovery of the fluorohydrin acetonide XXVI, an 11.5% yield of product B, and a 5% yield of material which was identical with product A. Product B was identical, according to the usual criteria, with a sample of the desired 9α-fluoro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII), prepared in another manner.[14] Treatment of B with acetone and hydrochloric acid afforded the fluorohydrin acetonide XXVI in 93% yield. This observation eliminated any possibility that B (XXVII) was a compound resulting from an acid-catalyzed rearrangement of the desired 21-deoxy analog of triamcinolone.

Product A, which was obtained from either the 9β,11β-oxide XXV or the fluorohydrin acetonide XXVI, differed from, but was isomeric with, 21-deoxytriamcinolone (XXVII). Furthermore, A afforded in 65% yield an acetonide which was not 21-deoxytriamcinolone acetonide (XXVI). The limited quantity of A prevented the further work that would be required for the definitive assignment of its structure. However, the available data indicated that this material was probably 9α-fluoro-11β,16α,17α-trihydroxy-17β-methyl-1,4-D-homoandrostadiene-3,17a-dione (XXVIII). Recent work has shown that D-homoannulation on the presence of Lewis acids affords 17a-keto-D-homosteroids as the predominant product[47]; rearrangement of a 16α,17α-dihydroxy-20-ketosteroid gives a 16α,17α-dihydroxy-17a-ketosteroid.[48] In view of these reports, structure XXVIII is assigned to product A, which was formed in acid media. It may be noted that the *cis* arrangement of the vicinal hydroxyl groups is supported by the ease with which A formed an isopropylidene derivative.[49]

It was also desirable to prepare the 9α-chloro analog of 21-deoxytriamcinolone [9α-chloro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione



XXVIII

(XXX)]; this was accomplished in the following manner. Reaction of the oxide XXV with hydrogen chloride gave the chlorohydrin acetonide in 86% yield. Treatment of this material with 0.5 *N* hydrochloric acid in refluxing aqueous methanol gave a mixture which was successfully resolved by partition chromatography on Celite.[46] In this manner there was isolated a 63% recovery of the acetonide XXIX and a 15% yield of the desired 9α-chlorotriol XXX. This latter material reacted with acetone and hydrochloric acid to reform the chlorohydrin acetonide XXIX in 90% yield.

Bioassays.[50]—Preliminary assay by the rat liver glycogen procedure of 9α-chloro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXX) indicated an activity less than that of hydrocortisone. The assay results with 9α-fluoro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII) are presented elsewhere.[14]

Acknowledgment.—We are indebted to Dr. S. Bernstein for suggesting the possible utility of 16α,17α-epoxy-11α-hydroxyprogesterone as a starting material for the preparation of triamcinolone and to Drs. N. Bohonos, L. Feldman, P. Shu, Mr. C. Pidacks and their associates of the Biochemical Research Section of these laboratories for supplying us with large quantities of the fermentation product. We would like to thank Mr. Charles Pidacks and his staff for advice and help in carrying out the partition chromatograms and Mr. Anthony Pellicano for the large-scale preparations of certain intermediates. Polarographic assays were kindly determined and interpreted by Dr. M. Halwer of the Chemical Process Improvement Department of these laboratories. The technical advice of Mr. W. S. Allen is gratefully acknowledged. The microanalyses were carried out by Mr. L. Brancone and staff, and the spectroscopic and polarimetric data were supplied by Mr. W. Fulmor and staff.

Experimental[51]

16α,17α-Epoxy-11α-mesyloxyprogesterone (III).—A solution of 5.00 g. (0.015 mole) of 16α,17α-epoxy-11α-hydroxyprogesterone (II)[8] and 5.0 ml. of methanesulfonyl chloride in 25 ml. of pyridine was allowed to stand at 5° during 18 hours. The solution was protected from the atmosphere by a calcium sulfate drying tube. Some solid separated from the solution, and the mixture was diluted with 100 ml. of water and 100 ml. of methylene chloride. The organic

(45) R. F. Hirschmann, R. Miller, J. Wood and R. E. Jones, THIS JOURNAL **78**, 4956 (1956).

(46) H. M. Kissman, C. Pidacks and B. R. Baker, *ibid.*, **77**, 18 (1955).

(47) (a) D. K. Fukushima, S. Dobriner, M. S. Heffler, T. H. Kritchevsky, F. Herling and G. Roberts, *ibid.*, **77**, 6585 (1955); (b) N. L. Wendler, D. Taub, S. Dobriner and D. K. Fukushima, *ibid.*, **78**, 5027 (1956); (c) R. B. Turner, M. Perelman and K. T. Park, Jr., *ibid.*, **79**, 1108 (1957).

(48) N. L. Wendler and D. Taub, *Chemistry & Industry*, 1237 (1957).

(49) See S. J. Angyal and C. G. MacDonald, *J. Chem. Soc.*, 686 (1952). We are familiar with only two examples in cyclic systems wherein a *trans*-glycol reacts with acetone to form a ketal. However, each involves the formation of a tri-*O*-isopropylideneinositol from a di-*O*-isopropylideneinositol. Angyal and MacDonald have explained the formation of a tri-*O*-isopropylidene derivative as a special situation in which the normal steric hindrance to ketal formation between *trans*-hydroxyl groups is diminished by withdrawal of axial groups from their usual position by prior ketal formation between *cis*-hydroxyl groups.

(50) The assays were done by L. Bortle, E. Heyder, J. Perrine, E. Ross and I. Ringler (Experimental Therapeutics Research Section of these laboratories).

(51) All melting points were determined in a capillary tube and are uncorrected. The ultraviolet spectra were determined in methanol solution on a Cary recording spectrophotometer unless otherwise specified. The infrared spectra (pressed potassium bromide disk) were determined with a Perkin-Elmer spectrophotometer (model 21). Optical rotations were determined in a 1-dm. semi-micro tube (unless otherwise noted) at wave length 5893 Å. (D). All evaporations were carried out under reduced pressure unless otherwise specified. Except where otherwise noted, the petroleum ether used was that fraction boiling at 60–70°.

layer was washed with 5% hydrochloric acid solution (100 ml.) and water (100 ml.). The solution was taken to dryness, and the residue was dissolved in 75 ml. of hot methanol and chilled to give 4.109 g. of solid, m.p. 159–161° dec. The mother liquor gave an additional 0.700 g. of solid, m.p. 159–161° dec. (79% yield). This material was of suitable purity for the subsequent step. The material was recrystallized four times from acetone–petroleum ether to give shiny white plates, m.p. 170–172° dec.; λ_{max} 238.5 mμ (ε 16,500); λ_{max} 5.85, 5.96, 6.18, 7.38, 7.50, 8.57, 10.83, 11.04 μ; $[\alpha]^{25}D$ +121° (*c* 1.0, chloroform). Reported values[11] are m.p. 160–161° dec.; λ_{max} 238.5 mμ (ε 15,300); λ_{max} 5.90, 5.98, 6.23, 7.56, 8.52, 10.82, 11.03 μ.

Anal. Calcd. for $C_{22}H_{30}O_6S$: C, 62.53; H, 7.16; S, 7.59. Found: C, 62.77; H, 7.46; S, 7.67.

Two experiments utilizing 0.57 mole and 1.1 moles of 16α,17α-epoxy-11α-hydroxyprogesterone afforded the mesylate III in 81 and 80% yield, respectively.

16α,17α-Epoxy-4,9(11)-pregnadiene-3,20-dione (IV).—A mixture of 0.900 g. (2.13 mmoles) of 16α,17α-epoxy-11α-mesyloxyprogesterone (III) and 0.900 g. of anhydrous sodium acetate in 10 ml. of glacial acetic acid was allowed to reflux for two hours. All solid dissolved at the boiling point. The dark solution was concentrated to half volume, and water was added slowly until the volume was 50 ml. The solid that had precipitated was collected by filtration to give 0.650 g. of crystals, m.p. 172–177°. The material was chromatographed on 16 g. of silica gel[52] (column size: 16 × 145 mm.). The column was washed with 200 ml. of benzene, and these washings were discarded. The column then was washed with 100 ml. of 25% ether in benzene. This wash was taken to dryness, and the residue was recrystallized from acetone–petroleum ether to give 0.572 g. (80% yield) of white flat needles, m.p. 181–182° (reported[11] 174–179°), $[\alpha]^{25}D$ +187° (*c* 1.0, chloroform), λ_{max} 239 mμ (ε 17,150); λ_{max} 5.89, 6.04, 6.11 (inflection), 6.22 μ.

Anal. Calcd. for $C_{21}H_{26}O_3 \cdot {}^1/_2C_3H_6O$: C, 76.00; H, 8.17. Found: C, 76.27; H, 8.17.

A sample dried under reduced pressure (oil-pump) over phosphorus pentoxide at 140° for 18 hours gave essentially the same analytical values. Furthermore, the analysis of five other samples indicated the above degree of solvation.

Subsequently, two experiments utilizing 0.42 mole and 0.92 mole of the 11α-mesylate III gave 16α,17α-epoxy-4,9(11)-pregnadien-3,20-dione (IV) in quantitative yield.

16β-Bromo-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (V).—A mixture of 2.00 g. (6.14 mmoles) of 16α,17α-epoxy-4,9(11)-pregnadiene-3,20-dione (IV) in 16 ml. of glacial acetic acid was treated with 2.0 ml. of a solution of 30% hydrogen bromide in glacial acetic acid; all suspended solid dissolved immediately. The solution was allowed to stand at room temperature during 30 minutes. Sufficient water was added to make the volume 65 ml.; this precipitated a gum. The mixture was extracted with 70 ml. of methylene chloride; the methylene chloride solution was washed with 70 ml. of water, 70 ml. of saturated sodium carbonate solution and again with 70 ml. of water. After drying the methylene chloride solution over magnesium sulfate, the solvent was removed to give 2.60 g. of semi-crystalline residue. This material was crystallized from acetone–petroleum ether to give 1.55 g. (62% yield) of fine needles, m.p. 177–180° dec. Two additional recrystallizations from acetone–petroleum ether gave fine white needles, m.p. 182–184° dec., $[\alpha]^{25}D$ +114° (*c* 0.98, methanol), λ_{max} 238 mμ (ε 17,900); λ_{max} 2.95, 5.82, 6.02, 6.17 μ.

Anal. Calcd. for $C_{21}H_{27}BrO_3$: C, 61.92, H, 6.68; Br, 19.62. Found: C, 61.92; H, 6.95; Br, 19.59.

3,17α-Diacetoxy-16β-bromo-3,5,9(11)-pregnatriene-20-one (VII).—A solution of 0.412 g. (1.0 mmole) of 16β-bromo-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (V) and 0.140 g. of *p*-toluenesulfonic acid hydrate in 25 ml. of acetic anhydride was allowed to stand at room temperature during 17 hours. The solution was poured into 200 ml. of water, chilled in an ice-bath, and magnetically stirred until the acetic anhydride hydrolyzed. The solid was collected by filtration, and this material was recrystallized three times from dilute acetone to give 0.383 g. (80% yield) of white needles, m.p. 94–98° dec., $[\alpha]^{25}D$ −91.6° (*c* 2.0, chloroform); λ_{max} 236 mμ (ε 19,700); λ_{max} 5.70, 5.78 (shoulder), 5.95, 6.07, 8.25 (broad), 9.75 μ.

(52) A product of Davison Chemical Co., Baltimore, Md.

Anal. Calcd. for $C_{25}H_{31}BrO_5$: C, 61.10; H, 6.36; Br, 16.24. Found: C, 60.88; H, 6.64; Br, 15.97.

This material was unstable, and, after five days at room temperature, the white needles became buff colored. The sample became black after standing at room temperature for two weeks. For this reason the crude material, obtained consistently in 95% yield, was used without purification for the subsequent step.

17α-Hydroxy-16β-iodo-4,9(11)-pregnadiene-3,20-dione (VI).—A mixture of 7.385 g. (0.024 mole) of 16α,17α-epoxy-4,9(11)-pregnadiene-3,20-dione (IV) and 31 g. of sodium iodide in 80 ml. of glacial acetic acid was heated on the steam-bath for two hours. All solid dissolved initially with the aid of occasional agitation, and solid then separated from the dark colored solution. The mixture was diluted with 500 ml. of methylene chloride and 500 ml. of water. The organic solution was washed with saturated sodium thiosulfate solution (500 ml.) and water (500 ml.) and dried over magnesium sulfate. The solution was taken to dryness, and the residual glass was dissolved in 50 ml. of benzene. Crystals soon began separating, and the material was collected by filtration to give 4.330 g. of needles, m.p. 166–168° dec. The mother liquor was chromatographed on 200 g. of silica gel.[52] The column was washed with two liters of 3% ether in benzene and two liters of 5% ether in benzene; these washings were discarded. The column was then eluted with 4 liters of 7% ether in benzene; the eluate was taken to dryness. The residue was recrystallized from acetone–petroleum ether to give 1.328 g. (18% recovery) of **16α,17α-epoxy-4,9(11)-pregnadiene-3,20-dione (IV)**, m.p. 178–180° alone or when mixed with an authentic sample.

The column was then eluted with 2.5 liters of 20% ether in benzene. The eluate was taken to dryness, and the residue was combined with the 4.330 g. of crystalline material. The combined solids were recrystallized from benzene–petroleum ether to give 5.846 g. (69% yield based on unrecovered oxide) of white needles, m.p. 164–166° dec. after darkening from 155°, $[\alpha]^{25}D$ +118° (*c* 1.0, methanol), λ_{max} 238 mμ (ε 17,300); λ_{max} 2.96, 5.74, 6.03, 6.20 μ.

Anal. Calcd. for $C_{21}H_{27}IO_3$: C, 55.51; H, 5.95; I, 27.93. Found: C, 55.12; H, 6.15; I, 27.64.

3,17α-Diacetoxy-16β-iodo-3,5,9(11)-pregnatriene-20-one (VIII).—In the manner described previously, 3.474 g. (7.65 mmoles) of 17α-hydroxy-16β-iodo-4,9(11)-pregnadiene-3,20-dione (VI) was allowed to react with 1.00 g. of *p*-toluenesulfonic acid and 125 ml. of acetic anhydride. The product (3.842 g., 93% yield) was obtained as white needles, m.p. 113–115° dec., by recrystallization from dilute acetone, $[\alpha]^{25}D$ −62° (*c* 1.0, chloroform), λ_{max} 235 mμ (ε 20,000); λ_{max} 5.75, 5.81 (shoulder), 5.99, 6.09, 8.26 (broad), 9.76 μ.

Anal. Calcd. for $C_{25}H_{31}IO_5$: C, 55.77; H, 5.81; I, 23.57. Found: C, 55.92; H, 6.02; I, 23.33.

16β-Bromo-17α-hydroxy-11α-mesyloxyprogesterone (XII).—A solution of 5.00 g. (0.012 mole) of 16α,17α-epoxy-11α-mesyloxyprogesterone (III) in 100 ml. of carbon tetrachloride–acetic acid (1:1 v./v.) was cooled to 18°. This solution was treated with 10 ml. of a solution of 32% hydrogen bromide in glacial acetic acid; an oily layer separated. The mixture was stirred magnetically for 30 minutes and then poured into 1000 ml. of cracked ice. The solid which separated was collected by filtration and dried to give 6.00 g. (99% yield) of solid, m.p. 165–168° dec. Recrystallization from acetone gave white needles, m.p. 169.5–170.5° dec., $[\alpha]^{25}D$ +142° (*c* 0.99, pyridine), λ_{max} 238 mμ (ε 17,600); λ_{max} 2.92, 5.82, 6.05, 6.22, 7.39, 8.54 μ.

Anal. Calcd. for $C_{22}H_{31}BrO_6S$: C, 52.49; H, 6.21; Br, 15.87; S, 6.25. Found: C, 52.69; H, 6.19; Br, 15.97; S, 6.32.

16α-Acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX). Method A.—A mixture of 3.300 g. (6.72 mmoles) of 3,17α-diacetoxy-16β-bromo-3,5,9(11)-pregnatriene-20-one (VII), 6.60 g. of anhydrous sodium acetate and 100 ml. of glacial acetic acid was allowed to reflux during six hours. All solid dissolved when the reflux temperature was reached. The cooled solution was concentrated to near dryness; the residue was triturated with 200 ml. of water. The solid was collected by filtration to give 2.489 g. of solid, m.p. 135–160°, which was dissolved in 15 ml. of benzene and chromatographed on a silica gel[52] column (20 × 100 mm.). The column was washed with 200 ml. of methylene chloride–benzene (30:70), 500 ml. of methylene chloride, 200 ml. of chloroform–methylene chloride (20:80), 400 ml. of chloroform–

methylene chloride (50:50); these washings were discarded. The column was then washed with 1400 ml. of chloroform; 100-ml. fractions were collected, and the material eluted in fractions 8–12 was combined. Recrystallization from methylene chloride–petroleum ether gave 1.446 g. (57% yield) of white feathery needles, m.p. 170–180° (bubbling), $[\alpha]^{25}D$ +36.8° (*c* 0.57, chloroform), λ_{max} 240 mμ (ε 16,300); λ_{max} 2.91, 5.75, 5.86, 6.02, 6.19 μ. Reported[14] values are m.p. 183–187°, $[\alpha]^{25}D$ +37.6° (methanol), and λ_{max} 240 mμ (ε 16,500).

Anal. Calcd. for $C_{23}H_{30}O_5 \cdot {}^1/_4CH_2Cl_2$: C, 67.75; H, 7.53; Cl, 4.34. Found: C, 67.73; H, 7.54; Cl, 4.36.

When this material was recrystallized from acetone–petroleum ether, it was obtained as white crystals, m.p. 179–188° (bubbling).

Anal. Calcd. for $C_{23}H_{30}O_5 \cdot C_3H_6O$: C, 70.24; H, 8.16. Found: C, 70.21; H, 8.10.

Method B.—In the manner described in the previous experiment, 3.228 g. (6.0 mmoles) of 3,17α-diacetoxy-16β-iodo-3,5,9(11)-pregnatriene-2-one (VIII), 6.40 g. of sodium acetate and 100 ml. of glacial acetic acid were heated six hours to give 1.307 g. (37% yield) of needles, m.p. 170–180° (bubbling) after recrystallization from methylene chloride–petroleum ether. The infrared spectrum of this material was identical with that of the material obtained by method A.

Method C.—A suspension of 2.000 g. (4.0 mmoles) of 16β-bromo-17α-hydroxy-11α-mesyloxyprogesterone (XII) in 150 ml. of methylene chloride was treated with 0.65 g. of *p*-toluenesulfonic acid hydrate and 100 ml. of acetic anhydride. The mixture was stirred magnetically during 18 hours; all solid dissolved. The methylene chloride was removed at 40°, and the residual solution was magnetically stirred with 250 ml. of water while being chilled in an ice-bath. After the acetic anhydride had hydrolyzed, the solution was diluted with 500 ml. of water and extracted with methylene chloride (3 × 150 ml.). The solvent was removed to give a gummy residue which was heated with 8.0 g. of sodium acetate in 100 ml. of boiling glacial acetic acid as described in method A. This procedure gave 0.607 g. (39% yield) of IX as white needles, m.p. 173–188° (bubbling), after chromatography and recrystallization from benzene–petroleum ether. The infrared spectra of this material and that obtained by method A were identical.

16α,17α-Dihydroxy-4,9(11)-pregnadiene-3,20-dione (X).—A solution of 0.384 g. (1.0 mmole) of 16α-acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX), 15 ml. of methanol, 5 ml. of water and 0.17 ml. of 37% hydrochloric acid solution was allowed to boil during three hours. The solution was concentrated until solid began crystallizing, chilled in an ice-bath and filtered to give 0.231 g. (67% yield) of crystals, m.p. 197–203°. The solid was recrystallized two times from acetone–petroleum ether to give white prisms, m.p. 220–223°, $[\alpha]^{25}D$ +68.5° (*c* 0.99, methanol), λ_{max} 239 mμ (ε 17,200); λ_{max} 2.97, 5.85, 6.05, 6.20 μ. Reported[14] values are m.p. 206–216°, $[\alpha]^{25}D$ +73.3° (methanol) and λ_{max} 239 mμ (ε 17,800).

Anal. Calcd. for $C_{21}H_{28}O_4$: C, 73.22; H, 8.19. Found: C, 73.44; H, 8.27.

11α-Hydroxy-4,16-pregnadiene-3,20-dione (XIV).—A solution of 5.345 g. (0.0155 mole) of 16α,17α-epoxy-11α-hydroxyprogesterone (II) in 250 ml. of glacial acetic acid was treated with 88 ml. of 0.43 *N* chromous chloride solution under an atmosphere of carbon dioxide.[16] The solution was allowed to stand at room temperature for 30 minutes. Water (250 ml.) and methylene chloride (500 ml.) were added; the methylene chloride extract was washed with water (250 ml.) saturated sodium carbonate solution (2 × 250 ml.) and again with water (2 × 250 ml.). The organic solution was dried over magnesium sulfate and taken to dryness. The residue was allowed to reflux during two hours with 200 ml. of acetone and 2 ml. of 37% hydrochloric acid solution. The solution was concentrated to 25 ml. and diluted with 250 ml. of methylene chloride. The solution was washed with water (2 × 100 ml.), dried over magnesium sulfate and taken to dryness. The residue was recrystallized twice from ethyl acetate to give 2.530 g. (50% yield) of long white needles, m.p. 179–180°, $[\alpha]^{25}D$ +143° (*c* 0.43, chloroform), λ_{max} 240 mμ (ε 24,800); λ_{max} 2.98, 6.00, 6.04, 6.20, 6.30 μ. Reported[17] values are m.p. 179–180°, $[\alpha]D$ +142° (chloroform), λ_{max}^{MeOH} 241 mμ (ε 24,100).

Anal. Calcd. for $C_{21}H_{28}O_3$: C, 76.79; H, 8.59. Found: C, 76.94; H, 8.71.

11α,16α,17α-Trihydroxyprogesterone (XV). Method A.—A solution of 10.0 g. (0.039 mole) of osmium tetroxide in 250 ml. of benzene was added dropwise to a solution of 12.00 g. (0.037 mole) of 11α-hydroxy-4,16-pregnadiene-3,20-dione (XIV) in 240 ml. of benzene and 6 ml. of pyridine with continuous stirring. The solution became black, and a black solid separated before the addition was completed. The mixture was stirred for two hours, after which it was treated with 380 ml. of methanol; this dissolved the black solid. A solution of 72 g. of potassium bicarbonate and 72 g. of sodium sulfite in 1200 ml. of water was added; within five minutes a black solid again separated from the solution. Additional benzene (500 ml.) and chloroform (500 ml.) were added; however, the dark material failed to dissolve. The mixture was stirred for 19 hours; at no time did the separated material assume a red color. The mixture was filtered, and the organic layer was separated, washed with saturated saline, and taken to dryness. The residue was recrystallized from acetone–petroleum ether to give 1.576 g. (12% yield) of white crystals, m.p. 213–215°, $[\alpha]^{25}D$ + 82.6° (*c* 0.5, chloroform), λ_{max} 241 mμ (ε 14,600); λ_{max} 2.96, 5.86, 6.01, 6.20, 8.24, 9.43 μ.

Anal. Calcd. for $C_{21}H_{30}O_5$: C, 69.58; H, 8.34. Found: C, 69.77; H, 8.56.

Method B.[18]—A solution of 0.652 g. (2.0 mmoles) of 11α-hydroxy-4,16-pregnadiene-3,20-dione (XIV) in 21 ml. of acetone and 0.24 ml. of glacial acetic acid was chilled in an ice-bath to 3°. There was added in one portion a solution of 0.316 g. (2.1 mmoles) of potassium permanganate in 15 ml. of 85% aqueous acetone. After three minutes, the mixture was treated with 3 ml. of a saturated aqueous solution of sodium sulfite. The mixture was filtered through a bed of Celite,[53] and the filtrate was extracted with methylene chloride (2 × 50 ml.). The combined organic extracts were washed with water (2 × 150 ml.), dried over magnesium sulfate and taken to dryness. The residue, which crystallized upon trituration with acetone, was recrystallized from acetone–petroleum ether to give 0.336 g. (46% yield) of white crystals, m.p. 213–215°. A mixture of this material and that prepared by method A melted at 213–215°. Furthermore, the material prepared by the two methods had identical infrared spectra.

11α-Hydroxy-16α,17α-isopropylidenedioxyprogesterone (XVI).—A solution of 0.750 g. (2.1 mmoles) of 11α,16α,17α-trihydroxyprogesterone (XV) in 50 ml. of acetone and containing two drops of 37% hydrochloric acid solution was allowed to stand at room temperature during 16 hours. The solution was filtered, and the filtrate was concentrated to a volume of about 20-ml. Water (5 ml.) was added slowly, and long needles began separating. The mixture was chilled and filtered. The material was recrystallized from acetone–petroleum ether to give 0.700 g. (87% yield) of long white needles, m.p. 252–254°, $[\alpha]^{25}D$ +117° (*c* 1.1, chloroform), λ_{max} 241 mμ (ε 16,200); λ_{max} 2.90, 5.85, 6.03, 6.23, 7.29, 9.60 μ.

Anal. Calcd. for $C_{24}H_{34}O_5$: C, 71.61; H, 8.51. Found: C, 71.35; H, 8.77.

16α,17α-Isopropylidenedioxy-11α-mesyloxyprogesterone (XVII).—A solution of 0.400 g. (1.0 mmole) of 11α-hydroxy-16α,17α-isopropylidenedioxyprogesterone (XVI) and 0.75 ml. of methanesulfonyl chloride in 2.5 ml. of pyridine was allowed to stand at 5° during 17 hours. The dark solution was diluted with 20 ml. of methylene chloride, washed with 20 ml. of 5% hydrochloric acid solution and with 20 ml. of water. The organic solution was dried over magnesium sulfate and taken to dryness. The residual gum was crystallized from acetone–petroleum ether to give 0.332 g. (69% yield) of hard crystals, m.p. 165–167° dec. The material was recrystallized from acetone–petroleum ether to give white needles, m.p. 171–172° dec., $[\alpha]^{25}D$ +94° (*c* 1.1, chloroform), λ_{max} 238 mμ (ε 17,100); λ_{max} 5.83, 5.97, 6.17, 7.24, 7.28, 7.50, 8.55, 9.55, 11.00 μ.

Anal. Calcd. for $C_{25}H_{36}O_7S$: C, 62.47; H, 7.55; S, 6.68. Found: C, 62.28; H, 7.77; S, 6.32.

16α,17α-Isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI).—A solution of 0.132 g. (0.28 mmole) of 16α,17α-isopropylidenedioxy-11α-mesyloxyprogesterone (XVII) and

(53) Celite is Johns-Manville's registered trademark for diatomaceous silica products.

0.132 g. of anhydrous sodium acetate in 2 ml. of glacial acetic acid was allowed to reflux during two hours. The pale yellow solution was diluted slowly with 7 ml. of water to give 0.109 g. (100% yield) of long needles, m.p. 193–194°. The material was recrystallized from petroleum ether to give long white needles, m.p. 198–199.5°, $[\alpha]^{25}D$ +111° (*c* 0.98, chloroform), λ_{max} 238 mμ (ε 18,300); λ_{max} 5.85, 5.95, 6.10, 6.19, 7.32, 7.38, 9.57 μ. Reported[14] values are m.p. 200–201°, $[\alpha]^{25}D$ +107° (chloroform), and λ_{max} 239 mμ 17,700).

Anal. Calcd. for $C_{24}H_{32}O_4$: C, 74.97; H, 8.39. Found: C, 74.76; H, 8.59.

16α,17α-Isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione 3-Semicarbazone.—A solution of 0.384 g. (1.0 mmole) of 16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI) in 25 ml. of methanol was treated with a solution of 0.790 g. (7.0 mmoles) of semicarbazide hydrochloride and 0.60 g. (7.7 mmoles, 0.4 ml.) of pyridine in 4.0 ml. of water. The solution was allowed to reflux during 22 hours, concentrated to turbidity and chilled to give crystals, m.p. 234–240° dec. Two recrystallizations from methanol gave white crystals, m.p. 244–247° dec., $[\alpha]^{25}D$ +145° (*c* 1.0, chloroform), λ_{max} 269 mμ (ε 33,700); λ_{max} 2.90, 5.90, 6.35, 6.90 μ.

Anal. Calcd. for $C_{25}H_{35}N_3O_4$: N, 9.52. Found: N, 9.60.

Attempted Reaction of Ethyl Oxalate with 16α,17α-Dihydroxy-4,9(11)-pregnadiene-3,20-dione (X).—A solution of 1.1 ml. of 1 *N* methanolic sodium methoxide in 4 ml. of benzene was distilled until 3.5 ml. of distillate was collected. The residual mixture was diluted with 4 ml. of benzene and 0.249 g. (1.7 mmoles, 0.23 ml.) of ethyl oxalate was added with magnetic stirring; all solid dissolved. The solution was treated with 0.344 g. (1.0 mmole) of 16α,17α-dihydroxy-4,9-(11)-pregnadiene-3,20-dione (X). Almost instantaneously, a gelatinous solid separated from the solution. The mixture was stirred for three hours, diluted with 6 ml. of ether, and stirred for an additional hour. The solid was collected by filtration and dried to give 0.349 g. of solid.

The material was dissolved in 30 ml. of acetone containing three drops of 37% hydrochloric acid solution and allowed to stand at room temperature for 21 hours. The solid which was obtained by dilution of the solution with water was chromatographed on silica gel[52] (column size: 16 × 135 mm.). The column was washed with 100 ml. of benzene, 150 ml. of 5% ether in benzene and 75 ml. of 7% ether in benzene; these washings were discarded. Elution with 225 ml. of 10% ether in benzene and evaporation of the eluate gave 0.121 g. of crystals. The material was recrystallized from petroleum ether to give 0.077 g. (20% yield) of **16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI)** as white needles, m.p. 196–198°, $[\alpha]^{25}D$ +111° (*c* 1.0, chloroform), λ_{max} 238 mμ (ε 16,800). Its complete infrared spectrum was identical with that of an authentic sample.

Further elution of the column with 100 ml. of 20% ether in benzene, 175 ml. of ether, 75 ml. of 10% chloroform in ether, 100 ml. of chloroform, 125 ml. of acetone and 150 ml. of methanol failed to give any additional material.

Attempted Reaction of Ethyl Oxalate with 16α-Acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX).—A solution of 1.1 ml. of 1 *N* sodium methoxide in methanol and 2 ml. of benzene was taken to dryness; the residue was suspended in 2 ml. of benzene and treated with 0.23 ml. (1.7 mmoles) of ethyl oxalate. A solution of 0.386 g. (1.0 mmole) of 16α-acetoxy-17α-hydroxy-4,9(11)-pregnadiene-3,20-dione (IX) in 4 ml. of warm benzene was added. The resulting yellow solution was stirred during 20 hours at room temperature. Ether (6 ml.) was added and the resulting turbid solution was stirred for one hour. An additional 18 ml. of ether was added and stirring was continued for three hours. The mixture was filtered to give 0.173 g. of solid. This solid was combined with the partially evaporated filtrate, treated with Norite activated carbon while hot, and filtered. The filtrate was diluted with hot petroleum ether and chilled to give 0.266 g. (84% yield) of white prisms, m.p. 202–210°.

An additional recrystallization raised the melting range to 209–211°, λ_{max} 239 mμ (ε 16,000). The complete infrared spectrum was identical with that of an authentic sample of **16α,17α-dihydroxy-4,9(11)-pregnadiene-3,20-dione (X)**.

2-Ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XVIII).—A solution of 25 ml. of 1 *N* methanolic sodium methoxide in 100 ml. of anhydrous benzene was distilled until 76 ml. of distillate was collected. The residual mixture was allowed to cool to room temperature, diluted with 40 ml. of benzene and treated with 6.00 g. (0.041 mole, 5.54 ml.) of ethyl oxalate with magnetic stirring. All solid immediately dissolved, and 9.30 g. (0.024 mole) of 16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI) was added. The solution became progressively darker, and after one hour, solid began precipitating from the solution. The mixture was stirred at room temperature for 22 hours. Ether (100 ml.) was added, and the mixture was stirred for one hour. An additional 200 ml. of ether was added, and stirring was continued for three hours. The mixture was filtered to give 6.74 g. (55% yield) of crude **sodium salt of 2-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione**. The filtrate was evaporated to a sirup which was triturated with 100 ml. of ether. Filtration gave an additional 2.735 g. (23% yield) of the crude sodium salt. The mother liquor was taken to dryness, and the residue was recrystallized from petroleum ether to give 0.982 g. (10% recovery) of **16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI)**.

The combined crude sodium salt was dissolved in 400 ml. of water, and the turbid solution was filtered. The filtrate was acidified with 5% hydrochloric acid solution, and the precipitated solid was collected by filtration and dried over phosphorus pentoxide to give 7.919 g. (75% yield based on unrecovered XI) of pale yellow amorphous solid, λ_{max} 242 mμ (ε 14,200), 352 mμ (ε 7760); λ_{max}^{HCl} 244 mμ (ε 14,500), 320 mμ (ε 2230); λ_{max}^{NaOH} 244 mμ (ε 14,500), 352 mμ (ε 11,000)[54]; λ_{max} 5.75, 5.82, 6.11, 6.92, 7.22, 7.26, 7.83, 9.55 μ.

Anal. Calcd. for $C_{28}H_{36}O_7$: C, 69.40; H, 7.48. Found: C, 67.85; H, 7.21.

2α-Bromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XIX).—A solution of 3.20 g. (0.0326 mole) of potassium acetate in 100 ml. of methanol was chilled in an ice-bath with magnetic stirring. 2-Ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XVIII) (7.917 g., 0.0163 mole) was added using 20 ml. of methanol to aid in the transfer. A 19.8-ml. aliquot of a solution prepared by diluting 6.60 g. of bromine to 50-ml. volume with carbon tetrachloride was added dropwise over a period of 50 minutes; the aliquot is equivalent to 0.0163 mole. The resulting turbid, yellow solution was treated with 100 mg. of phenol and 16.3 ml. of 1 *N* sodium methoxide in methanol. The solution was allowed to reflux for 10 minutes on the steam-bath and chilled in an ice-bath. The crystals were collected by filtration to give 4.732 g. of pale yellow solid, m.p. 175–179° dec. The mother liquor was concentrated to a volume of about 25 ml. to give an additional 2.420 g. of crystals, m.p. 179–183° dec. (95% yield). This material was suitable for subsequent work without purification.

A 0.500-g. sample was dissolved in 7 ml. of benzene and chromatographed on 15 g. of silica gel[52] (column size: 16 × 140 mm.). The column was washed with 100 ml. of benzene, and these washings were discarded. The column was then washed with 250 ml. of ether–benzene (3:97 v./v.); 50-ml. fractions were collected. Fractions 2–4 were combined and taken to dryness; the residue was recrystallized from methanol to give 0.303 g. of glistening white plates, m.p. 177–179° dec. When this material was dried under reduced pressure (oil-pump) at 56° for two hours, it became colored; therefore, the analytical sample was dried at room temperature, $[\alpha]^{25}D$ +122° (*c* 0.98, chloroform), λ_{max} 241 mμ (ε 16,300); λ_{max} 5.85, 5.88 (less intense than the 5.85 band), 6.09, 6.17, 7.25, 7.30, 8.54, 9.58 μ.

Anal. Calcd. for $C_{24}H_{31}BrO_4$: C, 62.20; H, 6.74; Br, 17.25. Found: C, 62.40; H, 6.82; Br, 17.16.

In a subsequent experiment the above procedure was followed except that the phenol and methanolic sodium methoxide addition was omitted. In this manner a 90% yield of XIX was obtained.

16α,17α-Isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XX).—A solution of 7.243 g. (0.0156 mole) of 2α-bromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XIX) in 15 ml. of 2,4,6-collidine was allowed to reflux during 45 minutes. Solid began precipitating from the solution almost immediately, and the mixture became dark. The cooled mixture was diluted with sufficient ether to make the total volume 80 ml. Filtration gave crude 2,4,6-collidine hydrobromide which was washed with 100 ml.

(54) For the acid and base spectra, a methanolic solution was diluted 1:1 with 0.1 *N* hydrochloric acid and 0.1 *N* sodium hydroxide, respectively.

of ether. The combined filtrate and washing was washed with 10% sulfuric acid solution (3 × 50 ml.) and water (2 × 50 ml.). The combined acid and water washes were extracted with 100 ml. of ether. The combined ether solutions were dried over a mixture of Norite and magnesium sulfate. The mixture was filtered and the ether solution was taken to a volume of about 10 ml. and filtered to give 3.231 g. (54% yield) of needles, m.p. 196.0–198.5°. A sample was recrystallized three times from acetone–petroleum ether to give fine, white needles, m.p. 204.5–205.5°, $[\alpha]^{25}D$ +22.2° (*c* 1.0, chloroform), λ_{max} 238 mμ (ε 15,600); λ_{max} 5.84, 5.98, 6.13, 6.21, 7.24, 7.30, 8.56, 9.55 μ.

Anal. Calcd. for $C_{24}H_{30}O_4$: C, 75.36; H, 7.91. Found: C, 75.36; H, 7.62.

In polarographic assays a solution (*c* 1 mg./ml.) of triene XX in 50% methanol and 0.1 *M* tetra-*n*-butylammonium hydroxide buffered to *p*H 3 with phosphoric acid showed a half-wave potential of −1.04 volts, whereas its Δ^4-3-keto counterpart XI under the same conditions showed a half-wave potential of −1.19 volts.[55]

In another experiment the triene XX was isolated in 52% yield from 0.073 mole of XIX.

A solution of 73 mg. (0.19 mmole) of XX, 150 mg. (1.33 mmoles) of semicarbazide hydrochloride and 118 mg. (1.5 mmoles, 0.08 ml.) of pyridine in 5 ml. of methanol and 0.8 ml. of water was allowed to reflux during 22 hours. The solution was concentrated to turbidity and chilled to give 45 mg. of the semicarbazone of XX as an amorphous solid, m.p. 204–210° dec., λ_{max} 241 mμ (ε 10,300), 298 mμ (ε 25,800); λ_{max} 2.90, 5.90, 6.35, 6.92 μ.

Anal. Calcd. for $C_{25}H_{33}N_3O_4$: N, 9.56. Found: N, 10.82.

2,21-Bis-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXI).—A solution of 154 ml. of 1 *N* methanolic sodium methoxide in 560 ml. of benzene was distilled until 430 ml. of distillate was collected. The residual mixture was cooled, and was treated with 32.2 ml. (0.238 mole) of ethyl oxalate with magnetic stirring; all solid dissolved. This solution was treated with 27.90 g. (0.070 mole) of 16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XI) to give a yellow solution which, after 15 minutes of stirring, became deep red. The solution was stirred for three hours and then diluted with 600 ml. of ether. This precipitated an orange solid. Stirring was continued for 45 minutes and the mixture was allowed to stand at room temperature during 16 hours. The solid was collected by filtration and dried under reduced pressure to give 47.0 g. (109% yield) of the crude sodio derivative as a yellow solid. The solid was dissolved in 500 ml. of water and the mixture was filtered through a bed of Celite.[53] The resulting deep red filtrate was acidified with the necessary amount of 5% hydrochloric acid solution, and the precipitated amorphous solid was collected by filtration and dried under reduced pressure over phosphorus pentoxide to give 26.7 g. (65% yield) of crude product.

In a preliminary experiment, the product was obtained in 54% yield from 0.384 g. (1.0 mmole) of the isopropylidene derivative. The solid gave a wine color with an alcoholic solution of ferric chloride, and it had λ_{max} 244 mμ (ε 13,000), 330 mμ (ε 4390); λ_{max}^{HCl} 244 mμ (ε 13,000), 320 mμ (ε 2400); λ_{max}^{NaOH} 244 mμ (ε 14,500), 350 mμ (ε 11,100)[54]; λ_{max} 5.75, 5.82, 6.11, 6.94, 7.25, 7.29, 7.90 (broad), 9.56 μ.

Anal. Calcd. for $C_{32}H_{40}O_{10}$: C, 65.74; H, 6.90. Found: C, 66.60, 66.50; H, 7.26, 7.26.

Determination of the Quantity of 2-Ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20 (XVIII) in the Crude 2,21-Bis-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXI).—To a magnetically-stirred, ice-chilled solution of 0.705 g. (7.2 mmoles) of potassium acetate in 10 ml. of methanol there was added 1.045 g. (1.79 mmoles) of crude 2,21-bis-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXI). The resulting orange solution was treated with 4.17 ml. of 0.86 *M* bromine in carbon tetrachloride during 10 minutes. The yellow solution was stirred for 10 minutes; phenol (25 mg.) and 3.58 ml. of 1 *N* sodium methoxide in methanol were added. The resulting solution was concentrated on the steam-bath at atmospheric pressure to remove the carbon tetrachloride. The concentrate was diluted with 100 ml. of water; this precipitated an amorphous solid which was collected by filtration and dried under reduced pressure over phosphorus pentoxide. This material was dissolved in 25 ml. of benzene and chromatographed on silical gel[52] (column size: 20 × 200 mm.). The column was eluted with 5% ether in benzene solution; 50-ml. fractions were collected. The material eluted in fractions 4–9 was combined and recrystallized from methanol to give 0.335 g. of **2α-bromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XIX)** as white crystals, m.p. 177–179° dec. alone or when mixed with an authentic sample. Furthermore, the infrared spectrum of this material and that of the authentic specimen were identical.

From the data it was calculated that the 1.045 g. of crude 2,21-bis-ethoxalyl derivative contained 0.352 g. of **2-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XVIII)**. These data were used for the calculation of the amount of bromine required for the following preparation.

2α,21-Dibromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXII).—A solution of 6.46 g. (0.066 mole) of potassium acetate in 200 ml. of methanol was chilled in an ice-bath with magnetic stirring. To the solution was added 9.107 g. of crude 2,21-bis-ethoxalyl-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXI); the material dissolved to give a red-orange solution. Bromine (4.64 g., 0.029 mole, 34 ml. of a solution prepared by dilution of 6.835 g. of bromine to 50-ml. volume with carbon tetrachloride) in carbon tetrachloride was added dropwise over a period of 70 minutes, and the solution was stirred for an additional 10 minutes. The solution was treated with 29 ml. of 1 *N* methanolic sodium methoxide, stirred for 15 minutes, and concentrated to remove the carbon tetrachloride. The solid which separated was collected by filtration to give 4.809 g. of crystals, m.p. 199–201° dec. The filtrate was concentrated to about 75 ml. and cooled to give an additional 2.924 g. of crystals, m.p. 201–204° dec. The combined material was recrystallized from ethyl acetate to give 7.527 g. of white crystals, m.p. 215–217° dec.

The mother liquor from the separation of the second crop of material was diluted with water, and the amorphous solid was collected by filtration and dried over phosphorus pentoxide. The material was chromatographed on 100 g. of silica gel[52] (column size: 40 × 130 mm.). The column was washed with 1750 ml. of 5% ether in benzene. The first 875 ml. of eluate was discarded; the material in the second 875 ml. of eluate was recrystallized from methanol to give 0.776 g. of **2α-bromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XIX)**, m.p. 175–179° dec. Admixture with an authentic sample of this material did not alter the melting point behavior. Furthermore, when this material was treated with collidine as described previously, **16α,17α-isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XX)** was formed in 59% yield.

In a preliminary experiment, the dibromo compound XXII was obtained after recrystallization from ethyl acetate as white crystals, m.p. 215–217°, $[\alpha]D$ +113° (*c* 1.1, chloroform), λ_{max} 240 mμ (ε 15,500); λ_{max} 5.77, 5.91, 6.10, 6.17, 7.25, 8.52, 9.55 μ.

Anal. Calcd. for $C_{24}H_{30}Br_2O_4$: C, 53.15; H, 5.58; Br, 29.48. Found: C, 53.55; H. 5.62; Br, 29.47.

When the dibromo material was stirred with potassium acetate in acetone at room temperature for three days as described in an accompanying paper,[10] XXII was recovered to the extent of 92%. Moreover, when attempted acetolysis of XXII was carried out in refluxing acetone for 16 hours, there resulted a 65% recovery of XXII.

A solution of 54.1 mg. (0.1 mmole) of XXII in 4 ml. of 2-methoxyethanol was treated with 1 ml. of 1 *N* sodium hydroxide solution. The resulting black solution was allowed to reflux during 24 hours. The solution was treated with Norite activated carbon and filtered to give a yellow solution from which 33.9 mg. (92% of two molar equivalents) of silver bromide was isolated in the usual way.

Attempted Preparation of 21-Bromo-16α,17α-isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XXIII) from 2α,21-Dibromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXII). Method A.—In a typical experiment 0.271 g. (0.5 mmole) of 2α,21-dibromo-16α,17α-

(55) At *p*H 2.8, prednisone and prednisolone have half-wave potentials of −1.05 volts and −1.15 volts, respectively; and cortisone and hydrocortisone have half-wave potentials of −1.20 volts and −1.26 volts, respectively.[56]

(56) P. Kabasakalian and J. McGlotten, THIS JOURNAL, **78**, 5032 (1956).

isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXII) and 0.165 g. (1.5 mmoles) of anhydrous lithium chloride were dissolved in 10 ml. of dimethylformamide.[43] The solution was swept thoroughly with nitrogen, and the container was sealed and heated at 98–103° for 3.5 hours. The solution was diluted to turbidity with water and cooled to give 168 mg. of a solid which was recrystallized from acetone–petroleum ether to give yellow needles, m.p. 223–233° dec., λ_{max} 241, 285 (shoulder), 385 mμ ($E^{1\%}_{1cm}$ 355, 65, 80); 5.75, 5.98, 6.12, 6.20, 6.31, 6.43 μ.

A material with the same ultraviolet and infrared spectra was obtained when the time of heating was extended to 30 hours.

A third experiment run at 90–92° for two hours gave yellow needles, m.p. 230–236° dec.; λ_{max} 241, 285, 385 mμ ($E^{1\%}_{1cm}$ 300, 100, 65); λ_{max} 5.77, 5.91, 5.99, 6.13, 6.20 (inflection), 6.31, 6.43 μ.

When the experiment was conducted at 71° for two hours, the isolated material had m.p. 204–207°, λ_{max} 241 mμ ($E^{1\%}_{1cm}$ 315); λ_{max} 5.77, 5.91, 5.99, 6.08, 6.15 μ. The relative intensities of the 5.91 and 5.99 μ absorption bands indicated that the bulk of this mixture was unreacted XXII.

All attempts (silica gel absorption chromatography, partition chromatography and fractional crystallization) to resolve the mixtures from the first and second experiments were unsuccessful.

Method B.—A solution of 0.524 g. (1.0 mmole) of 2α,21-dibromo-16α,17α-isopropylidenedioxy-4,9(11)-pregnadiene-3,20-dione (XXII) in 5 ml. of redistilled collidine was allowed to reflux during 40 minutes. The resulting mixture was diluted with 10 ml. of dry ether; this precipitated an amorphous solid. The mixture was filtered, and the residue was washed thoroughly with 40 ml. of ether. The combined filtrate and washings were washed with a 10% sulfuric acid solution (2 × 25 ml.) and finally with water (2 × 25 ml.). The ethereal solution was dried over magnesium sulfate and taken to dryness. The residue was crystallized from acetone–petroleum ether to give 50 mg. of rosettes of needles, m.p. 212–216° dec. One recrystallization raised the melting range to 218–220° dec., λ_{max} 240 mμ ($E^{1\%}_{1cm}$ 300); λ_{max} 5.77, 5.83, 5.98, 6.13, 6.20 μ. The appearance of the carbonyl absorption band at 5.83 μ seemed to indicate the presence of a 17β-acetyl steroid in the mixture.

Anal. Calcd. for $C_{24}H_{29}BrO_4$: C, 62.46; H, 6.34; Br, 17.36. Found: C, 65.50; H, 6.86; Br, 10.63.

The ether-insoluble residue was washed with two 15-ml. portions of acetone. This gave 0.165 g. (82% of one molar equivalent) of collidine hydrobromide, λ_{max} 265 mμ (ε 7800). From the extinction coefficient this material was estimated to be 95% pure.

The acetone solution was taken to dryness, and the residue was dissolved in 75 ml. of methylene chloride. The organic solution was washed with 50 ml. of 10% sulfuric acid solution and 50 ml. of water. After drying the solution over magnesium sulfate, the solvent was removed to give 0.226 g. of an amorphous residue; λ_{max} 5.80, 6.02, 6.16 μ.

9α-Bromo-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIV).—A solution of 2.179 g. (5.7 mmoles) of 16α,17α-isopropylidenedioxy-1,4,9(11)-pregnatriene-3,20-dione (XX) in 62 ml. of dioxane (peroxide-free) and 12 ml. of water was chilled to 15°. The solution was treated with 10.7 ml. of 20% perchloric acid solution and 0.980 g. (7.1 mmoles) of N-bromoacetamide.[44] The solution was allowed to stand at room temperature for 30 minutes, treated with 25 ml. of saturated sodium sulfite solution, and diluted with sufficient water to make the total volume 250 ml. The resulting mixture was extracted with three 150-ml. portions of methylene chloride. The combined extracts were dried over magnesium sulfate and concentrated to a volume of about 15 ml. The solid which had separated was collected by filtration and washed with 10 ml. of methanol to give 1.585 g. of crystals, m.p. 218–221° dec. after darkening from 195°. The mother liquor gave an additional 0.257 g. of material (65% yield). The solid was recrystallized from acetone to give white crystals, m.p. 219.0–220.5° dec. after darkening from 195°, $[\alpha]^{25}D$ +115° (*c* 0.2, chloroform), +126° (c 1.0, pyridine), λ_{max} 242 mμ (ε 14,700); λ_{max} 3.00, 5.84, 6.02, 6.17, 6.24(inflection), 7.26, 8.54, 9.51 μ.

Anal. Calcd. for $C_{24}H_{31}BrO_5$: C, 60.13; H, 6.52; Br, 16.67. Found: C, 60.18; H, 6.71; Br, 16.68.

9β,11β-Epoxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXV).—A solution of 1.20 g. (2.5 mmoles) of 9α-bromo-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIV) and 1.20 g. of anhydrous potassium acetate in 180 ml. of ethanol was allowed to reflux during 16 hours.[44] The solution was concentrated to near dryness, and the moist residue was triturated with 50 ml. of water. The mixture was filtered to give 0.947 g. (95% yield) of near white solid, m.p. 238–242°.

In a preliminary experiment the oxide XXV was obtained in 78% yield from 0.110 g. of XXIV. The material was recrystallized from acetone–petroleum ether to give white needles, m.p. 243–245°, $[\alpha]^{25}D$ +50.5° (*c* 0.51, methanol), λ_{max} 247 mμ (ε 15,700); λ_{max} 5.85, 6.00, 6.12, 6.22, 7.23, 7.28, 8.58, 9.10 μ.

Anal. Calcd. for $C_{24}H_{30}O_5$: C, 72.33; H, 7.59. Found: C, 72.21; H, 7.78.

9α-Bromo-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIV). From 9β,11β-Epoxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXV).—A solution of 0.398 g. (1.0 mmole) of XXV in 10 ml. of carbon tetrachloride, 10 ml. of acetic acid and 0.8 ml. of 32% hydrogen bromide in glacial acetic acid was allowed to stand at room temperature during 10 minutes. The solution was diluted with 50 ml. of water and extracted with 50 ml. of methylene chloride. The organic solution was washed with water (2 × 50 ml.), saturated sodium bicarbonate solution (50 ml.) and water (2 × 50 ml.). The solution was dried over magnesium sulfate and taken to dryness. The white solid residue was recrystallized from acetone–petroleum ether to give 0.423 g. (89% yield) of white crystals, m.p. 219–220.5° dec. alone or when mixed with an authentic sample. The material had $[\alpha]^{25}D$ +127° (*c* 0.99, pyridine) and its infrared spectrum was identical with that of an authentic specimen.

9α-Fluoro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXVI).—A solution of 0.636 g. (1.60 mmoles) of 9β,11β-epoxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXV) in 6 ml. of methylene chloride and 10 ml. of tetrahydrofuran was chilled to −30° while in a stainless steel bomb.[45] To this solution was added 5 ml. of hydrogen fluoride in 5 ml. of methylene chloride (previously chilled to −30°). The bomb was sealed and shaken at 5° during 18 hours. The contents of the bomb were poured into 100 ml. of saturated sodium bicarbonate solution contained in a stainless steel beaker. The bomb was rinsed twice with 60 ml. of chloroform and the rinsings were combined with the sodium bicarbonate solution. The organic layer was separated and washed with a saturated saline solution (2 × 50 ml.). The solution was taken to dryness, and the moist residue was triturated with 7 ml. of acetone and collected by filtration to give 0.488 g. (73% yield) of solid, m.p. 294–296° dec. The material was recrystallized from acetone–petroleum ether to give shiny white platelets, m.p. 308–310° dec. (inserted in oil–bath at 275°) (m.p. 294–296° when sample taken up from room temperature), $[\alpha]^{25}D$ +102° (*c* 1.0, chloroform), λ_{max} 238 mμ (ε 15,500); λ_{max} 2.99, 5.85, 6.02, 6.17, 6.24, 7.25, 7.28, 8.54, 9.46 μ.

Anal. Calcd. for $C_{24}H_{31}FO_5$: C, 68.86; H, 7.47; F, 4.54. Found: C, 68.67; H, 7.45; F, 4.83.

Product A (9α-Fluoro-11β,16α,17α-trihydroxy-17β-methyl-1,4-D-homoandrostadiene-3,17α-dione) (XXVIII).—A solution of 0.600 g. (1.25 mmoles) of 9β,11β-epoxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXV) in 15 ml. of chloroform and 10 ml. of tetrahydrofuran was treated with 10 ml. of anhydrous hydrogen fluoride at 5° during 20 hours. The reaction was worked up as described in the previous experiment to give 0.142 g. (35% yield) of fine white needles, m.p. 290–292° after recrystallization from acetone–petroleum ether, $[\alpha]^{25}D$ +37.8° (*c* 0.66, methanol), λ_{max} 238 mμ (ε 14,800); λ_{max} 2.92, 2.99, 5.90, 6.02, 6.14, 6.22, 9.48, 10.02 μ.

Anal. Calcd. for $C_{21}H_{27}FO_5$: C, 66.65; H, 7.19; F, 5.02. Found: C, 66.34; H, 7.61; F, 5.23.

When this material (27 mg., 0.07 mmole) was treated with 6 ml. of acetone and one drop of 37% hydrochloric acid solution, 25.5 mg. of its isopropylidene derivative was obtained. The material was recrystallized from acetone–petroleum ether to give 19 mg. (65% yield) of white needles, m.p. 308–310° dec. A mixture with 9α-fluoro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXVI) melted at 268–274° dec. The material had $[\alpha]^{25}D$

+23.4° (*c* 0.4, methanol),[57] λ_{max} 237 mμ (ε 16,100); λ_{max} 2.90, 5.87, 6.00, 6.11, 6.21, 7.25, 7.30, 9.30, 9.44, 10.03 μ.

Anal. Calcd. For $C_{24}H_{31}FO_5$: F, 4.54. Found: F, 4.81.

9α-Fluoro-11β-16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII).—A solution of 0.418 g. (1.0 mmole) of 9α-fluoro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXVI) in 40 ml. of methanol, 15 ml. of water and 2.5 ml. of 37% hydrochloric acid solution was allowed to reflux during three hours. The hot solution was concentrated until solid began crystallizing, and then was extracted immediately with 100 ml. of methylene chloride. The organic solution was extracted with water (2 × 50 ml.), dried over magnesium sulfate and taken to dryness to give 0.292 g. of white solid.

The solid was dissolved in 15 ml. of the lower and 15 ml. of the upper phase of the system cyclohexane–dioxane–water (3:5:1) and the liquid was mixed thoroughly with 30 g. of Celite diatomaceous earth. This mixture was packed on top of a column which had been prepared from 250 g. of Celite[58] diatomaceous earth and 125 ml. of the lower phase of the solvent system just described. The column (80 × 3.8 cm.) was eluted with the upper phase, and the effluent was allowed to pass through a recording spectrophotometer which had been set at 240 mμ. A small amount of material having absorption at that wave length was eluted from the column in the first 26 ml., whereas the following 17 ml. did not contain such material. These fractions were discarded. The next 163 ml. contained much ultraviolet-absorbing material (fraction A). The following 525 ml. contained no ultraviolet-absorbing material. The following 310 ml. contained fraction B as shown by its ultraviolet absorption. After 63 ml. of effluent that contained no ultraviolet-absorbing material, the next 302 ml. contained fraction C.

Fraction A was taken to dryness and the residue was recrystallized from acetone–petroleum ether to give 0.131 g. of shiny white platelets, m.p. 308–310° dec. (inserted in oil-bath at 275°) alone or when mixed with known **9α-fluoro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXVI)**. Concentration of the mother liquor gave an additional 0.053 g. of this material (44% recovery).

Fraction B was taken to dryness and the residue was recrystallized from acetone–petroleum ether to give 44 mg. (11.5% yield) of **9α-fluoro-11β-16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII)** as small white prisms, m.p. 290–291° dec. after darkening from 275°; when inserted in the oil-bath at 270°, a sample of this material melted at 298–300° with dec., $[\alpha]^{25}D$ +53.3° (*c* 0.41, methanol),[57] +36.8° (*c* 1.09, pyridine), λ_{max} 238 mμ (ε 15,700); λ_{max} 2.85, 2.93, 5.86, 6.01, 6.19, 6.25, 9.34 μ. Reported[14] values are m.p. 286–287° dec., $[\alpha]^{25}D$ +62° (*c* 0.50, methanol), +41.5° (*c* 1.06, pyridine), λ_{max} 238 mμ (ε 15,100); λ_{max} (ν) 2.85 (3509), 2.93 (3401), 5.86 (1709), 6.01 (1667), 6.18 (1618) and 6.24 μ (1603 cm.$^{-1}$).

Anal. Calcd. for $C_{21}H_{27}FO_5$: C, 66.65; H, 7.19; F, 5.02. Found: C, 66.38; H, 7.41; F, 4.91.

Fraction C was taken to dryness and the residue was recrystallized from acetone–petroleum ether to give 18 mg. (5% yield) of fine white needles, m.p. 290–292° dec. alone or when mixed with **9α-fluoro-11β-16α,17α-trihydroxy-17β-methyl-1,4-D-homoandrostadiene-3,17α-dione (XXVIII)**. The material had λ_{max} 238 mμ (ε 15,100), $[\alpha]^{25}D$ +52.3° (*c* 0.8, pyridine), +35.3° (*c* 0.33, methanol) and its infrared spectrum was identical with that of the D-homo material obtained by reaction of hydrogen fluoride with the oxide XXV.

Preparation of 9α-Fluoro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXVI) from XXVII.—A solution of 13 mg. (0.034 mmole) of 9α-fluoro-11β,16α,-17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXVII) in 10 ml. of acetone and containing two drops of 37% hydrochloric acid solution was allowed to stand at room temperature during 16 hours. The solution was concentrated to about 2-ml. volume and diluted with 10 ml. of water. The solid was collected by filtration and recrystallized from acetone–petroleum ether to give 13 mg. (93% yield) of white crystals, m.p. 308–310° dec. (inserted in the oil-bath at 275°) alone or when mixed with an authentic specimen. The material had $[\alpha]^{25}D$ +101° (*c* 0.65, chloroform) and its complete infrared spectrum was identical with that of the authentic sample.

9α-Chloro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIX).—To a solution of 0.400 g. (1.0 mmole) of 9β,11β-epoxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXV) in 20 ml. of chloroform was added 10 ml. of chloroform that had been saturated with hydrogen chloride at 5°. The resulting solution was allowed to stand at 0° during four hours, and then taken to dryness at 25–30°. The solid residue was triturated with 10 ml. of acetone and filtered to give 0.386 g. of white solid. The material was recrystallized from chloroform–petroleum ether to give 0.375 g. (86% yield) of white crystals, m.p. 288–290° dec. after darkening from 255°. This material retained chloroform tenaciously as shown by low carbon and high chlorine combustion values. For acceptable analysis the material was recrystallized from 2-methoxyethanol to give white crystals, m.p. 288–290° dec. after darkening from 255°, $[\alpha]^{25}D$ +125° (*c* 1.0, chloroform), λ_{max} 238 mμ (ε 15,200); λ_{max} 2.98, 5.84, 6.02, 6.16, 6.23, 7.24, 7.28, 8.48 and 9.50 μ.

Anal. Calcd. for $C_{24}H_{31}ClO_5$: C, 66.27; H, 7.18; Cl, 8.15. Found: C, 66.26; H, 7.49; Cl, 8.12.

9α-Chloro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXX).—A mixture of 0.266 g. (0.6 mmole) of 9α-chloro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIX), 32 ml. of methanol, 12 ml. of water and 4 ml. of 37% hydrochloric acid solution was allowed to reflux during one hour. Methanol (25 ml.) was added to aid in solution of the solid, and the mixture was allowed to reflux for an additional two hours. The hot mixture was filtered to give 82 mg. of white crystals, m.p. 285–286° dec. after darkening from 245°. The melting point behavior was not altered by admixture with authentic starting material.

The filtrate was concentrated to a volume of about 25 ml. and the concentrate was extracted with two 100-ml. portions of methylene chloride. The combined extracts were dried over magnesium sulfate and taken to dryness.

The residue was suspended in 25 ml. of the lower and 25 ml. of the upper phase of the system cyclohexane–dioxane–water (3:5:1); the undissolved material was collected by filtration to give 38 mg. of crystals, m.p. 275–281° dec. after darkening 220°. The filtrate was mixed thoroughly with 50 g. of Celite[58] diatomaceous earth and this mixture was packed on a column which had been prepared from 200 g. of Celite diatomaceous earth and 100 ml. of the lower phase of the solvent system just described. The column (3.8 × 68 cm.) was eluted with the upper phase of the solvent system, and the effluent was allowed to pass through a recording spectrophotometer which had been set at 240 mμ. A small amount of material having absorption at that wave length was eluted in the first 32 ml.; this material was discarded. The next 128 ml. contained much ultraviolet-absorbing material (fraction A). After 290 ml. of eluate that contained no ultraviolet-absorbing material, the next 299 ml. contained more ultraviolet-absorbing material (fraction B). The following 179 ml. contained no such material.

Fraction A was taken to dryness to give 58 mg. of white solid which was recrystallized from chloroform–petroleum ether to give 47 mg. of white crystals, m.p. 284–286° dec. after darkening from 270°. This material was identical with the starting material according to mixture melting point and infrared spectra comparisons. The total recovery of starting material was 63%.

Fraction B was taken to dryness to give 48 mg. of white solid which was recrystallized from acetone–petroleum ether to give 36 mg. (15% yield) of white crystals, m.p. 242–243° dec., $[\alpha]^{25}D$ +73° (*c* 0.89, pyridine), λ_{max} 238 mμ (ε 15,300); λ_{max} 2.85, 2.94, 5.86, 6.01, 6.20, 6.26, 9.35 μ.

Anal. Calcd. for $C_{21}H_{27}ClO_5$: C, 63.87; H, 6.89. Found: C, 64.13; H, 7.12.

9α-Chloro-11β-hydroxy-16α,17α-isopropylidenedioxy-1,4-pregnadiene-3,20-dione (XXIX). From XXX.—A solution of 10 mg. (0.025 mmole) of 9α-chloro-11β,16α,17α-trihydroxy-1,4-pregnadiene-3,20-dione (XXX) in 10 ml. of acetone and two drops of 37% hydrochloric acid solution was allowed to stand at room temperature during 2.5 days. The solution was concentrated on the steam-bath until solid be-

(57) Determined in a 2-dm. semi-micro tube.

(58) The material used in these partition columns was Celite[58] 545 diatomaceous earth which had been washed with 6 *N* hydrochloric acid and then distilled water until neutral and finally with methanol. The substance was dried to give a fluffy powder.

gan separating, and the mixture was chilled to ice-bath temperature to give 9.8 mg. (90% yield) of white crystals, m.p. 286–288° dec. (darkened from 270°), after recrystallization from 2-methoxyethanol. A mixture of this material with an authentic sample melted at 287–289° dec. after prior darkening. This material had $[\alpha]^{25}D$ +121° (*c* 0.49, chloroform) and its entire infrared spectrum was identical with that of an authentic specimen.

PEARL RIVER, N. Y.

[CONTRIBUTION FROM THE MERCK SHARP & DOHME RESEARCH LABORATORIES, DIVISION OF MERCK & CO., INC.[a] AND THE DEPARTMENTS OF MICROBIOLOGY, BIOCHEMISTRY AND PEDIATRICS, SCHOOL OF MEDICINE, UNIVERSITY OF PENNSYLVANIA[b]]

The Structure of a New Product from Yeast: 2-(6-Hydroxy-2-methoxy-3,4-methylenedioxyphenyl)-benzofuran

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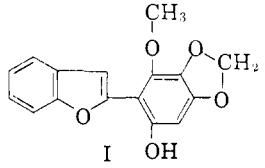
A new crystalline compound from yeast has been shown to be 2-(6-hydroxy-2-methoxy-3,4-methylenedioxyphenyl)-benzofuran (I).

The isolation from yeast of a new crystalline compound has been described by Forbes, Zilliken, Roberts and Gyorgy.[1] It was reported to be an antioxidant, and to be highly active in tests involving the inhibition of hemolysis of red cells. It was characterized as a colorless, solvent-soluble, optically inactive aromatic compound of the composition $C_{16}H_{12}O_5$. We have repeated the isolation and wish to present new studies on the degradation of this substance to products of proven structure[2] which led to the elucidation of the substance as 2-(6-hydroxy-2-methoxy-3,4-methylenedioxyphenyl)-benzofuran (I).

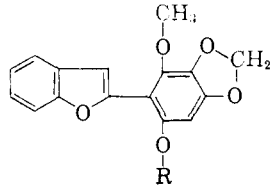


I

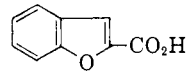
The initial studies[1] on this new substance showed the presence of one methoxyl group by a Zeisel determination, and a second methoxyl group was introduced by reaction of the compound with diazomethane; thus, the presence of a free phenolic hydroxyl group was indicated.

In this Laboratory, reaction of this substance with dimethyl sulfate in alkali gave in better yield the same methyl derivative II. Confirmation of the presence of a free hydroxyl group was obtained by acetylation to the monoacetyl derivative III. Evidence for the presence of a methylenedioxy group was obtained by hydrolysis with 90% sulfuric acid. The formaldehyde produced was detected by its color reaction with chromatropic acid.[3]

The fifth oxygen atom in the molecule was presumed to be a member of a nucleus, and this interpretation was established readily. Oxidation of the molecule with sodium chromate in glacial acetic acid resulted in the formation of coumarilic acid (IV). Alkaline fusion of the substance resulted in extensive decomposition, but some salicylic acid was isolated from the reaction products. The salicylic acid is *corollarial* in structural proof to the coumarilic acid. The initial studies on alkali fusion[1] led to actual recovery of the molecule in 50% yield, but a low yield of an impure and unidentified acid was obtained.



II, R = CH_3
III, R = $COCH_3$
XI, R = C_2H_5
XVIII, R = Ts



IV

These degradation reactions taken in conjunction with the other evidence made it apparent that the substance contains a benzofuran ring substituted in the 2-position and that this 2-substituent is a benzene ring bearing a phenolic hydroxyl, a methoxyl and a methylenedioxy group.

A nuclear magnetic resonance spectrum[4] was determined. Chemical shifts of 35, 50, 100 and approximately zero cycles were observed relative to benzene with relative intensities, respectively, of 1:2:3:5–6. These data suggested the presence of a phenolic hydroxyl group (35 cycles), a methylenedioxy group (50 cycles), a methoxyl group (100 cycles) and a benzene ring or rings (zero cycles) containing about five protons. These interpretations are in accord with structure I.

The methyl derivative II was used in a number of fruitless attempts to degrade the compound to a substituted benzoic acid in order to establish the positions of the oxygen substituents in the unelucidated portion of the molecule. Thus, oxidations with either permanganate or chromate did not destroy the benzofuran portion of this methyl derivative. Instead, the highly oxygenated benzene ring was attacked, and coumarilic and salicylic acids were isolated.

(1) M. Forbes, F. Zilliken, G. Roberts and P. Gyorgy, THIS JOURNAL, **80**, 385 (1958).

(2) A. F. Wagner, E. Walton, A. N. Wilson, J. O. Rodin, F. W. Holly, N. G. Brink and K. Folkers, *ibid.*, **81**, 4983 (1959).

(3) E. Hegriwe, *Z. anal. Chem.*, **110**, 22 (1937).

(4) We are indebted to Dr. Joseph Shoolery, Varian Associates, Palo Alto, Calif., for this measurement and interpretation.