[CONTRIBUTION FROM THE DIVISION OF STEROID METABOLISM AND BIOCHEMISTRY, SLOAN-KETTERING INSTITUTE FOR CANCER RESEARCH]

Synthesis of 17α,20α-Dihydroxysteroids[1]

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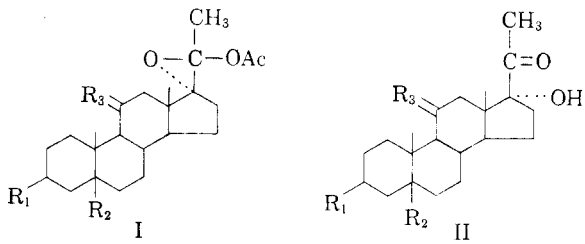
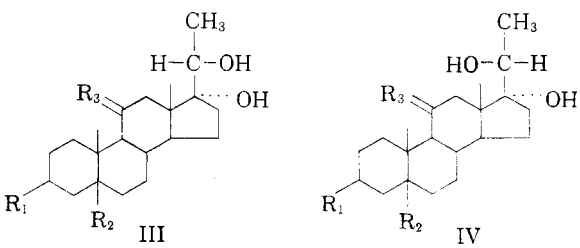
The preparation of 17α,20α-dihydroxysteroids by the lithium aluminum hydride reduction of 17α,20β-epoxy-20α-acetoxysteroids and the corresponding 17α-hydroxy-20-ketosteroids has been studied. Reduction of the epoxyacetates afforded the 17α,20α-glycols in higher yields.

The present study on the synthesis of 17α,20α-dihydroxysteroids was initiated because of the interest in the role of such glycols in the metabolism of adrenal steroid hormones.[2] There are a number of methods for the preparation of 17α,20-dihydroxysteroids but many of these yield the glycol with the β-orientated C-20 hydroxyl group as the principal product. Reduction of 17α-hydroxy-20-ketosteroids with lithium aluminum hydride affords the 17α,20α-dihydroxy epimer as the main product[3] but the yields are not so high as desired. Soloway and coworkers[4] reported that reduction of 17α,20β-epoxyallopregnane-3β,20α-diol diacetate (I B) with lithium aluminum hydride gave rise to a single product, allopregnane-3β,-17α,20α-triol (IV B) in 76% yield. Although the reduction of epoxyacetates with this reagent is highly stereoselective, it does not proceed with the formation of only a single α-glycol.[5] A study on the yields of the epimeric steroids obtained by the reduction of 17α,20β-epoxy-20-acetoxysteroids with lithium aluminum hydride has therefore been made. The corresponding 17α-hydroxy-20-ketosteroids were also reduced in order to evaluate the merits of the two methods for the synthesis of 17α,20α-dihydroxysteroids. It was found that the reduction of epoxyacetates with lithium aluminum hydride gave higher yields of 17α,-20α-glycols than the reduction of the corresponding ketols. Furthermore, since the 17α,20β-epoxyacetates are intermediates in the synthesis of 17α-hydroxy-20-ketosteroids, the direct reduction of the former is the method of choice for the preparation of 17α,20α-dihydroxysteroids.



I II

A. R_1 = α-OAc R_2 = α-H R_3 = H_2
B. R_1 = β-OAc R_2 = α-H R_3 = H_2
C. R_1 = α-OAc R_2 = β-H R_3 = H_2
D. R_1 = α-OAc R_2 = β-H R_3 = O



III IV

A. R_1 = α-OH R_2 = α-H R_3 = H_2
B. R_1 = β-OH R_2 = α-H R_3 = H_2
C. R_1 = α-OH R_2 = β-H R_3 = H_2
D. R_1 = α-OH R_2 = β-H R_3 = H, β-OH
E. R_1 = α-OH R_2 = β-H R_3 = O

Chart 1

The following epoxyacetates, 17α,20β-epoxyallopregnane-3α,20α-diol diacetate (I A), 17α,20β-epoxypregnane-3α,20α-diol diacetate (I C), and 11-keto-17α,20β-epoxypregnane-3α,20α-diol diacetate (I D), and their corresponding 17α-hydroxy-20-ketosteroid analogs II A–D, were reduced with lithium aluminum hydride. The resulting epimeric 17α,20-glycols (III and IV) were separated as their 3,20-diacetates by a partition type chromatography on silica gel containing *tert*-butyl alcohol and elution with increasing amounts of *tert*-butyl alcohol in methylene chloride. In some cases the reduction products were directly separated on silica gel containing ethanol and eluted with increasing amounts of ethanol in chloroform. The 3,20α-diacetates were eluted first in the former system but the or-

(1) This investigation was supported in part by a grant from the American Cancer Society and a research grant (CY-3207) from the National Cancer Institute of the National Institutes of Health, United States Public Health Service.

(2) (a) A. M. Bongiovanni, W. R. Eberlein, and J. Cara, *J. Clin. Endocrinol. and Metab.*, **14**, 409 (1954). (b) E. C. Reifenstein, 16, 1262 (1956). (c) J. P. Rosselet, J. W. Jailer, and S. Lieberman, *J. Biol. Chem.*, **225**, 977 (1957). (d) D. K. Fukushima and T. F. Gallagher, **226**, 725 (1957).

(3) (a) H. Hirschmann and F. B. Hirschmann, **187**, 137 (1951). (b) R. B. Turner, *J. Am. Chem. Soc.*, **75**, 3489 (1953).

(4) A. H. Soloway, W. J. Considine, D. K. Fukushima, and T. F. Gallagher, **76**, 2941 (1954).

(5) (a) M. Mousseron, R. Jacquier, M. Mousseron-Canet, and R. Zagdoun, *Compt. rend.*, **235**, 177 (1952). (*b*) N. S. Leeds, D. K. Fukushima, and T. F. Gallagher, *J. Am. Chem. Soc.*, **76**, 2943 (1954). (c) W. Biggerstaff and T. F. Gallagher, *J. Org. Chem.*, **22**, 1220 (1957).

der was reversed for the alcohols in the latter system so that the 20β-hydroxy epimers were eluted before the 20α-hydroxy derivatives.

It was found that reduction of 17α,20β-epoxy-20α-acetoxysteroids resulted in about 70% yield of the 17α,20α-dihydroxysteroids whereas less than 10% of the epimeric 17α,20β-glycols was obtained (Table I). The yields of 17α,20α-glycols by the lithium aluminum hydride reduction of the 17α-hydroxy-20-ketosteroids in the present study were about 30–55% whereas the yields of the 17α,20β-epimers were from 30–45% (Table I). These yields are comparable with the results in the literature[3,6] although other investigators have reported only the 17α,20β-dihydroxy epimer from this reduction.[7]

TABLE I

REDUCTION OF STEROIDS WITH LITHIUM ALUMINUM HYDRIDE TO 17α,20-DIHYDROXYSTEROIDS

Epoxyacetates	17α,20α-Glycol, %	17α,20β-Glycol, %
17α,20β-Epoxyallopregnane-3α,20α-diol diacetate	68	3
17α,20β-Epoxyallopregnane-3β,20α-diol diacetate[a]	76	
17α,20β-Epoxypregnane-3α,20α-diol diacetate	70	8
3α,20α-Diacetoxy-17α,20β-epoxypregnane-11-one[b]	62[c]	8[c]
Ketols		
3α-Acetoxy-17α-hydroxyallopregnane-20-one	55	4[d]
3β-Acetoxy-17α-hydroxyallopregnane-20-one	45	29
3α-Acetoxy-17α-hydroxypregnane-20-one	41	31
3α,17α-Dihydroxypregnane-11,20-dione[b]	29[c]	45[c]

[a] A. H. Soloway, W. J. Considine, D. K. Fukushima, and T. F. Gallagher, *J. Am. Chem. Soc.*, **76**, 2941 (1954). [b] Isolated as pregnane-3α,11β,17β,20-tetrol. [c] Includes the 11α-hydroxy epimer. [d] A mixture of the two epimers was obtained in 22% yield which was not resolved.

In contrast to lithium aluminum hydride, the metal borohydrides reduce 20-ketones predominantly to the 20β-hydroxy epimer. Thus the reduction of 3α,17α-dihydroxypregnane-11,20-dione (II D) with lithium borohydride or sodium borohydride in aqueous methanol overnight at room temperature gave 3α,17α,20β-trihydroxypregnane-11-one (IV E) in 71% yield, the 20α-hydroxy epimer (III E) in 5% yield and pregnane-3α,11β,17α,-20β-tetrol (IV D) in 8% yield. However, the use of a large excess of sodium borohydride under similar conditions has afforded only pregnane-3α,11β,17α,-20β-tetrol.[8] The large excess of the metal hydride is necessary for the reduction of the 11-ketone because the rate of reduction of this carbonyl group is slow[8c] and there is loss of the reagent by reaction with the solvent.

The assignment of the orientation of the C-20 hydroxy group in the heretofore undescribed 17α,-20-dihydroxysteroids, III A and D and IV A, was made by the application of molecular rotation differences (Table II). Further evidence was furnished by comparison of the order of elution in the partition chromatogram of the epimeric 17α,20-glycols, either as the aclohol or acetate, with that of known epimeric compounds. The molecular rotation differences resultant from acetylation of the 20-hydroxy epimers have been described by Sarett.[9] Since it would be tedious to prepare the necessary 3-acetoxy-17α,20-dihydroxysteroids[10] as reference compounds, the molecular rotation difference due to the conversion of 3-acetoxy-17α-hydroxy-20-ketosteroid to the epimeric 3,20-diacetoxy-17α-hydroxysteroid has been calculated (Table II). The differences for the compounds studied show good agreement. The ΔM is about −70 for the 17α,-20α-dihydroxysteroids (Δ^{α}) and about +125 for the 17α,20β-epimer (Δ^{β}) when the optical rotations were taken in chloroform. However, when the molecular rotation differences are calculated with optical rotation values obtained in acetone solution, the Δ^{α} is about −200 whereas the Δ^{β} is approximately 0. The discrepancy is primarily due to a solvent effect on the optical rotations of the 17α-hydroxy-20-ketosteroids since there is no solvent effect on the 17α-hydroxy-20-acetoxy derivatives. The effect of solvent on the optical rotations of α-ketols has previously been pointed out by Norymberski.[11]

After the reduction of 3α,17α-dihydroxypregnane-11,20-dione to the epimeric pregnane-3α,-11β,17α,20-tetrols with lithium aluminum hydride, two other isomeric pregnanetetrols were isolated in small amounts. These were pregnane-3α,11α,17α,-20α-tetrol and its 20β-hydroxy epimer; both were isolated as the 3,11,20-triacetates. The former was also obtained from the lithium aluminum hydride reduction of 3α,20α-diacetoxy-17α,20β-epoxypregnan-11-one and 11-ketopregnane-3α,17α,20α-triol-3,20-diacetate. That these two 11α-acetoxy compounds are epimeric at C-20 is borne out by the

(6) G. I. Poos, *J. Am. Chem. Soc.*, **77**, 4932 (1955).

(7) J. Romo, M. Romero, C. Djerassi, and G. Rosenkranz, **73**, 1528 (1951).

(8) (a) H. Herzog, M. Jevnik, P. Perlman, A. Nobile, and E. B. Hershberg, *J. Am. Chem. Soc.*, **75**, 266 (1953). (b) E. P. Oliveto and E. B. Hershberg, **75**, 488 (1953). (c) J. I. Appleby, G. Gibson, J. K. Norymberski, and R. D. Stubbs, *Biochem. J.*, **60**, 453 (1955).

(9) L. H. Sarett, *J. Am. Chem. Soc.*, **71**, 1175 (1949).

(10) In the acetylation of the reduction products of the allopregnane derivatives to the diacetate a small amount of 20-monoacetates was obtained. This fact indicates that it would be very difficult to isolate 3-monoacetylated products in large amount. The selective monoacetylation of the 20-hydroxyl group has also been observed by Sarett.[9]

(11) J. K. Norymberski, *J. Chem. Soc.*, 762 (1954).

TABLE II

MOLECULAR ROTATION DIFFERENCES IN THE REDUCTION OF 17α-HYDROXY-20-KETOSTEROIDS TO 17α-HYDROXY-20-ACETOXYSTEROIDS

	M_D		Δ^α		Δ^β		$\Delta^{\alpha-\beta}$	
	Chl	An	Chl	An	Chl	An	Chl	An
3α-Acetoxy-17α-hydroxypregnane-20-one	+102[a]							
Pregnane-3α,17α,20α-triol 3,20-diacetate	+ 12[b]		−90				+220	
Pregnane-3α,17α,20β-triol 3,20-diacetate	+232[b]				+130			
3β-Acetoxy-17α-hydroxyallopregnane-20-one	−60[a]	+60[c]						
Allopregnane-3β,17α,20α-triol 3,20-diacetate		−126[d]		−186				+239
Allopregnane-3β,17α,20β-triol 3,20-diacetate		+113[d]				+53		
17α-Hydroxypregnane-3,11,20-trione		+245[a]						
17α-Hydroxy-20α-acetoxypregnane-3,11-dione		+47[e]		−198				+185
17α-Hydroxy-20β-acetoxypregnane-3,11-dione		+232[f]				−13		
11β,17α-Dihydroxy-Δ⁴-pregnene-3,20-dione	+363[a]							
11β,17α-Dihydroxy-20α-acetoxy-Δ⁴-pregnene-3-one	+307[g]		−56				+356	
11β,17α-Dihydroxy-20β-acetoxy-Δ⁴-pregnene-3-one	+663[g]				+300			
3α-Acetoxy-17α-hydroxyallopregnane-20-one	+6[h]							
Allopregnane-3α,17α,20α-triol 3,20-diacetate	−66[b]		−72				+222	
Allopregnane-3α,17α,20β-triol 3,20-diacetate	+156[b]				+150			
3α-Acetoxy-17α-hydroxypregnane-11,20-dione	+196[i]	+328[j]						
3α,20α-Diacetoxy-17α-hydroxypregnane-11-one	+144[b]	+104[j]	−52	−224			+168	+207
3α,20β-Diacetoxy-17α-hydroxypregnane-11-one	+312[k]	+311[k]			+116	−17		
3α-Acetoxy-11β,17α-dihydroxypregnane-20-one	+156[a]	+324[l]						
Pregnane-3α,11β,17α,20α-tetrol 3,20-diacetate	+71[b]	+73[b]	−85	−251			+188	+222
Pregnane-3α,11β,17α,20β-tetrol 3,20-diacetate	+259[b]	+295[m]			+103	−29		

$\Delta^\alpha = \Delta M_D$ (20-C=O → 20α-OAc) $\Delta^\beta = \Delta M_D$ (20-C=O → 20β-OAc) $\Delta^{\alpha-\beta} = \Delta M_D$ (20α-OAc → 20β-OAc)
Chl = chloroform An = acetone

[a] This laboratory. [b] This investigation. [c] T. H. Kritchevsky and T. F. Gallagher, *J. Am. Chem. Soc.*, **73**, 184 (1951). [d] D. A. Prins and T. Reichstein, *Helv. Chim. Acta*, **23**, 1490 (1940). [e] L. H. Sarett, *J. Am. Chem. Soc.*, **71**, 1169 (1949). [f] E. P. Oliveto, C. Gerold and E. B. Hershberg, *J. Am. Chem. Soc.*, **76**, 6113 (1954). [g] G. I. Poos, *J. Am. Chem. Soc.*, **77**, 4932 (1955). [h] D. K. Fukushima, A. D. Kemp, R. Schneider, M. B. Stokem, and T. F. Gallagher, *J. Biol. Chem.*, **210**, 129 (1954). [i] E. P. Oliveto and E. B. Hershberg, *J. Am. Chem. Soc.*, **76**, 5167 (1954). [j] L. H. Sarrett, *J. Am. Chem. Soc.*, **70**, 1690 (1948). [k] M. Finkelstein, J. v. Euw and T. Reichstein, *Helv. Chim. Acta*, **36**, 1266 (1953). [l] E. P. Oliveto, T. Clayton, and E. B. Hershberg, *J. Am. Chem. Soc.*, **75**, 486 (1953). [m] E. P. Oliveto and E. B. Hershberg, *J. Am. Chem. Soc.*, **75**, 488 (1953).

molecular rotation difference of the triacetates. This value is +172, in good agreement with +200 for $\Delta^{\alpha-\beta}$ found in the present study (Table II). The production of an 11α-hydroxy isomer is quite general for adrenal steroids and is reported by Poos[6] in the reduction of 3-ethylenedioxy-17α-hydroxy-Δ⁵-pregnene-11,20-dione.

It has been recently postulated that the lithium aluminum hydride reduction of an epoxyacetate proceeded *via* the intermediate formation of a ketol (Fig. 1).[12] However, from the present study it is quite apparent that this mechanism does not apply. This follows from the fact that the yield of 17α,20α-glycols differs between the reduction of 17α,20β-epoxy-20-acetoxy- and 17α-hydroxy-20-ketosteroids. From this it is presumed that the reduction of the epoxyacetate proceeds in part by a mechanism similar to the one proposed by Gaylord for the reduction of the –N–C–O grouping.[12]

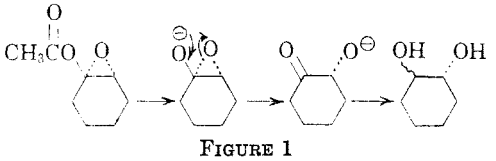


FIGURE 1

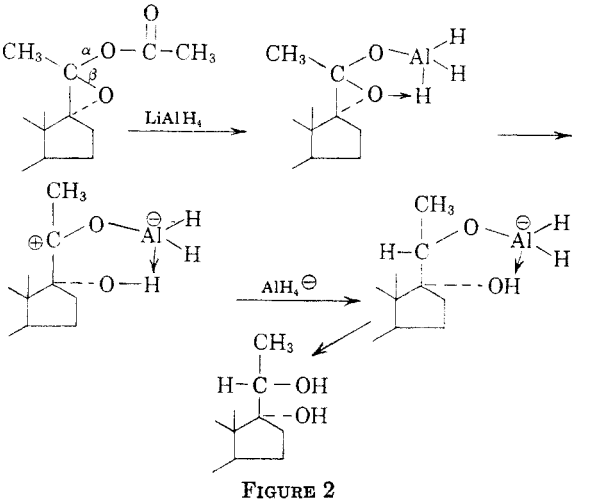


FIGURE 2

EXPERIMENTAL[13]

Allopregnane-3α,17α,20-triols. A. From 17α,20β-epoxyallopregnane-3α,20α-diol diacetate (I A). A solution of 2.2 g. of 17α,20β-epoxyallopregnane-3α,20α-diol diacetate[14] in 200 ml. of ether was added with stirring to a suspension of

(12) N. G. Gaylord, *Experientia*, **10**, 351 (1954).

(13) All melting points are corrected. The optical rotations were taken in chloroform solution unless stated otherwise.

(14) D. K. Fukushima, A. D. Kemp, R. Schneider, M. B. Stokem, and T. F. Gallagher, *J. Biol. Chem.*, **210**, 129 (1954).

1.3 g. of lithium aluminum hydride in 250 ml. of ether. The mixture was then refluxed for 2 hr. and the excess reagent destroyed with ethyl acetate. After acidification with dilute sulfuric acid, the crude triol was extracted with ethyl acetate. The extract was washed with base and brine, dried, and the solvent evaporated to give 1.8 g. of crude reduction product. The epimeric triols were separated on 800 g. of silica gel containing 320 ml. of ethanol by elution with ethanol in chloroform at a rate of 15 ml./hr. With 5% ethanol in chloroform, 70 mg. of a substance judged to be allopregnane-3α,17α,20β-triol by infrared spectrometry was obtained. Recrystallization from ethyl acetate and from methanol gave 26 mg. of allopregnane-3α,17α,20β-triol (IV A), m.p. 226–229°; $[\alpha]_D^{25}$ −5.8°.

Anal. Calcd. for $C_{21}H_{36}O_3$: C, 74.95; H, 10.78. Found: C, 74.63; H, 10.89.

Elution with 6% ethanol in chloroform afforded 1.20 g. of the triol epimeric at C-20. Recrystallization from methanol gave 1.03 g. of allopregnane-3α,17α,20α-triol (III A), m.p. 224–228°; the analytical sample melted at 228–230°; $[\alpha]_D^{26}$ −13.2°. The mixture with the 20β-hydroxy epimer melted at 204–219°.

Anal. Calcd. for $C_{21}H_{36}O_3$: C, 74.95; H, 10.78. Found: C, 74.45; H, 10.43.

Acetylation with acetic anhydride and pyridine at room temperature afforded allopregnane-3α,17α,20α-triol 3,20-diacetate which had a double m.p. 123–124° and 130–131°; $[\alpha]_D^{27}$ −15.7°.

Anal. Calcd. for $C_{25}H_{40}O_5$: C, 71.39; H, 9.59. Found: C, 71.44; H, 9.77.

B. From 3α-acetoxy-17α-hydroxyallopregnane-20-one (II A) *with lithium aluminum hydride.* A solution of 200 mg. of 3α-acetoxy-17α-hydroxyallopregnane-20-one[14] in 15 ml. of ether and 10 ml. of benzene was added with stirring to a suspension of 100 mg. of lithium aluminum hydride in 20 ml. of ether. The mixture was refluxed for 3 hr. and worked up in the manner described above. The crude reduction product (180 mg.) was acetylated with pyridine and acetic anhydride at room temperature for 2 hr., yielding 221 mg. of epimeric triol diacetates. The epimers were separated on 100 g. of silica gel containing 40 ml. of *tert*-butyl alcohol. Elution was started with 1% *tert*-butyl alcohol in petroleum ether-methylene chloride (1:1) at the rate of 10 ml./hr. With 1% *tert*-butyl alcohol in methylene chloride, 91 mg. of allopregnane-3α,17α,20α-triol 3,20-diacetate, as judged by infrared spectrometry, was obtained. Recrystallization from petroleum ether afforded 72 mg. of the 3α,20α-diacetate, m.p. 122–129°. The analytical sample had a double m.p., 123–124° and 130–131°; it was found that most of the samples had a melting point range between these two.[15]

Further elution with the same solvent gave 50 mg. of a mixture of the epimeric 3α,20α- and 3α,20β-diacetates of allopregnanetriol from which neither of the pure epimers could be obtained by recrystallization. A small amount of allopregnane-3α,17α,20β-triol 3,20-diacetate was then eluted from the chromatogram. Recrystallization gave 3,20-diacetate, m.p. 203–206°.

Elution with 5% *tert*-butyl alcohol in methylene chloride afforded 30 mg. of allopregnane-3α,17α,20α-triol 20-monoacetate. Recrystallization from acetone-petroleum ether gave the triol monoacetate, m.p. 213–216.5°; $[\alpha]_D^{28}$ −20.7°.

Anal. Calcd. for $C_{23}H_{38}O_4$: C, 72.98; H, 10.12. Found: C, 72.52; H, 10.27.

Acetylation with acetic anhydride and pyridine yielded allopregnane-3α,17α,20α-triol 3,20-diacetate. The triol monoacetate was recovered unchanged upon treatment with periodic acid. Oxidation with chromic acid yielded a substance which had an absorption band at 1716 cm.$^{-1}$ indicative of a ketone in a 6-membered ring (3-ketone).

C. From 3α,17α-dihydroxyallopregnane-20-one with sodium borohydride. A solution of 320 mg. of 3α,17α-dihydroxyallopregnane-20-one[14] in 13 ml. of methanol was reduced with 50 mg. of sodium borohydride in 6 ml. of methanol at room temperature overnight. The reduction mixture was diluted with equal volume of brine and extracted with ethyl acetate. The organic layer was washed with brine and dried and the solvent was evaporated to give 313 mg. of crude reduction product. The epimeric triols were acetylated (393 mg.) and chromatographed on silica gel containing *tert*-butyl alcohol in the manner described in *B*. Elution with 1% *tert*-butyl alcohol in methylene chloride afforded 264 mg. of crystalline material judged to be allopregnane-3α,17α,20β-triol 3,20-diacetate by infrared spectrometry. Recrystallization from acetone yielded 189 mg. of the 3α,20β-diacetate, m.p. 198–205°. The analytical sample melted at 203.5–206°; $[\alpha]_D^{28}$ +37.1°.

Anal. Calcd. for $C_{25}H_{40}O_5$: C, 71.39; H, 9.59. Found: C, 71.12; H, 9.52.

Elution with 5% *tert*-butyl alcohol in methylene chloride afforded 50 mg. of allopregnane-3α,17α,20β-triol monoacetate. Chromatography on silica gel and recrystallization from methanol gave prisms, m.p. 179.5–184°, clear at 192°. Acetylation yielded allopregnane-3α,17α,20β-triol 3,20-diacetate. The compound was proved to be the 20-monoacetate since it was recovered unchanged after treatment with periodic acid and afforded 3-ketoallopregnane-17α,20β-diol 20-monoacetate on oxidation with chromic acid.

Pregnane-3α,17α,20-triols. A. From 17α,20β-epoxypregnane-3α,20α-diol diacetate (I C). 17α,20β-Epoxypregnane-3α,20α-diol diacetate[16] (108 mg.) was reduced with 300 mg. of lithium aluminum hydride in the manner described above to give 87 mg. of crude pregnane-3α,17α,20-triols, m.p. 247–250.5°. Recrystallization from ethyl acetate gave 56 mg. of triol, m.p. 250–253°. Further recrystallization from methanol gave 31 mg. of pregnane-3α,17α-triol (III C), m.p. 253–254.5°.

The combined mother liquors (52 mg.) were acetylated and chromatographed on silica gel containing *tert*-butyl alcohol. Elutions with 1% *tert*-butyl alcohol in methylene chloride afforded 37 mg. of pregnane-3α,17α,20α-triol 3,20-diacetate. Recrystallization from methanol gave 29 mg. of the diacetate, m.p. 157.5–160.5°; $[\alpha]_D^{26}$ +3.4°.

Further elution yielded 9 mg. of the epimeric triol diacetate. Recrystallization from methanol gave 3 mg. of pregnane-3α,17α,20β-triol 3,20-diacetate, m.p. 186–188.5°; $[\alpha]_D^{27}$ +57.5°.

B. From 3α-acetoxy-17α-hydroxypregnane-20-one (II C). 3α-Acetoxy-17α-hydroxypregnane-20-one (209 mg.)[16] was reduced with 400 mg. of lithium aluminum hydride as described in method *B* above to give 184 mg. of pregnane-3α,17α,20-triols. Acetylation with acetic anhydride and pyridine for 3 hr. yielded 224 mg. of triol diacetate. Chromatography on silica gel containing *tert*-butyl alcohol and elution with 1% *tert*-butyl alcohol in methylene chloride yielded 72 mg. of pregnane-3α,17α,20α-triol 3,20-diacetate. Recrystallization from benzene gave 54 mg. of the diacetate, partial melt at 144° with transformation of prisms to needles and final melt at 157.5–158.5°. Further elution gave 24 mg. of compound judged to be pregnane-3α,17α,20α-triol diacetate by infrared spectrometry. Recrystallization gave 19 mg. of the triol diacetate, m.p. 140–147°, no depression of the m.p. when mixed with the above sample.

Continued elution with 1% *tert*-butyl alcohol in methylene chloride gave 72 mg. of pregnane-3α,17α,20β-triol 3,20-diacetate as judged by infrared spectrometry. Recrystallization from acetone-petroleum ether gave 60 mg. of the triol diacetate, m.p. 186–188°.

Allopregnane-3β,17α,20-triols. From 3β-acetoxy-17α-hydroxyallopregnane-20-one (II B). 3β-Acetoxy-17α-hydroxyallopregnane-20-one (570 mg.)[3b] was reduced with 600 mg. of lithium aluminum hydride as described in method *B*

(15) Many of the steroids in this investigation exhibited polymorphism and therefore the m.p. of a compound was not a good criterion of its purity.

(16) T. H. Kritchevsky and T. F. Gallagher, *J. Am. Chem. Soc.*, **73**, 184 (1951).

above to give 484 mg. of triol. Acetylation afforded 572 mg. of diacetate. Chromatography on silica gel containing *tert*-butyl alcohol afforded 284 mg. of allopregnane-3β,17α,20α-triol 3,20-diacetate as judged by infrared spectrometry. Recrystallization from ethyl acetate–methanol afforded 225 mg. of triol diacetate, m.p. 245–247°.

Further elution gave 54 mg. of a mixture of the diacetates of the epimeric 3β,17α,20-triols with the 20β epimer predominating as judged by infrared spectrometry. Following the mixture, 96 mg. of allopregnane-3β,17α,20β-triol 3,20-diacetate was obtained. Recrystallization from methanol gave 69 mg. of prisms, m.p. 151–159°. Further recrystallization from methanol gave 33 mg. of triol diacetate, m.p. 158–160°.

Pregnane-3α,11β,17α,20-tetrols. A. From 3α,20α-diacetoxy-17α,20β-epoxypregnane-11-one (I D). 3α,20α-Diacetoxy-17α,20β-epoxypregnane-11-one (170 mg.)[17] was reduced with 400 mg. of lithium aluminum hydride as described in method *A* above to give 153 mg. of pregnane-3α,11β,17α,-20-tetrols. Acetylation with acetic anhydride and pyridine at room temperature afforded 181 mg. of the 3,20-diacetate which was chromatographed on silica gel containing *tert*-butyl alcohol.

Elution with 2% *tert*-butyl alcohol in methylene chloride gave 11 mg. of substance judged to be pregnane-3α,11α,17α,-20α-tetrol 3,11,20-triacetate by infrared spectrometry. Further elution with the same solvent yielded 96 mg. of pregnane-3α,11β,17α,20α-tetrol 3,20-diacetate (III D diacetate). Recrystallization from methanol gave 70 mg. of the diacetate, m.p. 211–219°. The analytical sample melted at 214.5–219.5°; $[\alpha]_D^{20}$ +16.2°, +16.7° (acetone).

Anal. Calcd. for $C_{25}H_{40}O_6$: C, 68.77; H, 9.24. Found: C, 69.09; H, 9.44.

The diacetate III D had two crystalline forms, prisms and needles, melting at the same temperature. Some samples of pure pregnane-3α,11β,17α,20α-tetrol 3,20-diacetate which melted at 209–217° melted at 216.5–222° when pulverized. Chromic acid oxidation of the tetrol diacetate afforded 11-ketopregnane-3α,17α,20α-triol 3,20-diacetate, m.p. 225–227° (III E diacetate).

Saponification of III D diacetate and recrystallization from acetone–benzene gave pregnane-3α,11β,17α,20α-tetrol which had a double m.p. 133–136° and 200–201°; $[\alpha]_D^{29}$ +10.2°.

Anal. Calcd. for $C_{21}H_{36}O_4$: C, 71.55; H, 10.30. Found: C, 71.45; H, 10.28.

Elution with 3% *tert*-butyl alcohol in methylene chloride afforded 13 mg. of crystalline substance judged to be pregnane-3α,11β,17α,20β-tetrol 3,20-diacetate by infrared spectrometry.

B. From 3α,17α-dihydroxypregnane-11,20-dione. 3α,17α-Dihydroxypregnane-11,20-dione (8.0 g.)[17] was placed in a Soxhlet thimble and continuously extracted into a flask containing 4 g. of lithium aluminum hydride in 500 ml. of benzene and 300 ml. of ether. The excess reagent was destroyed with ethyl acetate. Acidification of the mixture with dilute sulfuric acid gave insoluble crystalline material. Filtration of the solid and several washings with brine, base and water afforded 2.18 g. of pregnane-3α,11β,17α,20β-tetrol, m.p. 274–281°. Ethyl acetate was added to the aqueous filtrate and the organic layer was separated. It was washed successively with brine, base and water. After drying and concentrating the ethyl acetate solution, an additional 1.33 g. of pregnane-3α,11β,17α,20β-tetrol, m.p. 274–278°, was collected by filtration. The two crystalline fractions were combined and recrystallized from methanol to give 2.95 g. of pregnane-3α,11β,17α,20β-tetrol, m.p. 278–283°, reported m.p. 275–282°[8a] and 282–284°.[8c]

The solvent from the ethyl acetate filtrate was removed *in vacuo* to give 4.50 g. of yellow oil. Acetylation with acetic anhydride and pyridine at room temperature for 2 hr. afforded 5.22 g. of pregnanetetrol diacetate. Chromatography on 1.5 kg. of silica gel containing 600 ml. of *tert*-butyl alcohol in methylene chloride afforded 500 mg. of pregnane-3α,11α,17α,20α-tetrol 3,11,20-triacetate, m.p. 202–215.5°. Recrystallization from methanol gave 390 mg. of triacetate, m.p. 228–229°. The analytical sample from acetone melted at 229.5–230.5°; $[\alpha]_D^{30}$ −29°, M_D −140.

Anal. Calcd. for $C_{27}H_{42}O_7$: C, 67.75; H, 8.85. Found: C, 67.80; H, 8.94.

The α-orientation of the C-20 hydroxyl group has been assigned since this compound was also a side product of lithium aluminum hydride reduction of the known 11-ketopregnane-3α,17α,20α-triol 3,20-diacetate to pregnane-3α,-11β,17α,20α-tetrol.

Further elution with the same solvent gave 2.41 g. of pregnane-3α,11β,17α,20α-tetrol 3,20-diacetate, m.p. 218–223°. Recrystallization from methanol gave 2.00 g. of tetrol diacetate, m.p. 212–219.5°.[15]

A mixture (618 mg.) of pregnane-3α,11β,17α,20α- and 20β-tetrol 3,20-diacetate and pregnane-3α,11α,17α,20β-tetrol 3,11,20-triacetate was then eluted.

Elution with 4% *tert*-butyl alcohol in methylene chloride gave 243 mg. of material judged to be pregnane-3α,11β,17α,-20β-tetrol 3,20-diacetate by infrared spectrometry. Recrystallization from methanol gave 72 mg. of the tetrol diacetate, partially melted from 120° and all clear at 189°; $[\alpha]_D^{27}$ +59.3°; reported m.p. 111–112.5° and 186–187°.[2b] An additional 90 mg. of tetrol diacetate, m.p. 128°, clear at 165°, was obtained from the mother liquor.

The mixture (618 mg.) obtained above was oxidized with chromic acid in acetic acid. Upon chromatography 11-ketopregnane-3α,17α,20-triol 3,20-diacetate was separated from a new substance, pregnane-3α,11α,17α,20β-tetrol 3,11,20-triacetate (86 mg.), m.p. 264–266°, $[\alpha]_D^{27}$ +7.0, M_D +32.

Anal. Calcd. for $C_{27}H_{42}O_7$: C, 67.75; H, 8.85. Found: C, 67.83; H, 8.58.

11-Ketopregnane-3α,17α,20-triols. A solution of 200 mg. of lithium borohydride in 15 ml. of methanol was added to a solution of 500 mg. of 3α,17α-dihydroxypregnane-11,20, dione (II D) in 35 ml. of methanol. The reaction mixture was allowed to stand at room temperature overnight and then diluted with equal volume of brine. The solution was acidified to destroy the excess reagent and then neutralized with base. The reduction product was extracted with ethyl acetate and washed with brine. The extract was dried and the solvent was evaporated to give 500 mg. of triolone. Recrystallization from benzene gave 446 mg. of 11-ketopregnane-3α,17α,20β-triol (IV E), m.p. 222–226°. The crystals and the mother liquor were combined and acetylated with pyridine and acetic anhydride at room temperature for 2 hr. to give 658 mg. of triolone diacetate. Chromatography on silica gel containing *tert*-butyl alcohol and elution with 3% *tert*-butyl alcohol in methylene chloride yielded 29 mg. of 11-ketopregnane-3α,17α,20α-triol 3,20-diacetate. Recrystallization from methanol gave 17 mg. of diacetate-m.p. 220–224°. Further recrystallization gave 11-ketopregnane-3α,17α,20α-triol 3,20-diacetate, m.p. 225–227°; $[\alpha]_D^{27}$ +32.2°, reported m.p. 227–228°.[18] Saponification and recrystallization from benzene gave 11-ketopregnane-3α,-17α,20α-triol (III E), m.p. 184–188°. The triolone had a tendency to gel from this solvent as Sarett[18] noted. Recrystallization from acetone gave needles, m.p. 193.5–194.5°; 7 months later the same samples melted at 203.5–206°. Concentration of the mother liquor yielded prisms, m.p. 205–209°. Sarett[18] reported m.p. 189–191° (from ether) and 210–212° (from benzene) for 11-ketopregnane-3α,17α,20α-triol. The infrared spectra of the two crystalline forms were identical in chloroform solution but differed when taken in potassium bromide disc. This work will be reported elsewhere.

Further elution with the same solvent gave 441 mg.

(17) H. V. Anderson, E. R. Garrett, F. H. Lincoln, Jr., A. H. Nathan, and J. H. Hogg, *J. Am. Chem. Soc.*, **76**, 743 (1954).

(18) L. H. Sarett, *J. Am. Chem. Soc.*, **70**, 1690 (1948).

of 11-ketopregnane-3α,17α,20β-triol 3,20-diacetate, m.p. 240–246°. Recrystallization from methanol gave the diacetate, m.p. 243–245.5°; $[\alpha]_D^{26}$ +72.1°; reported m.p. 244–246°; $[\alpha]_D^{24}$ +71.9°[19]; m.p. 249–250°.[20] Saponification and recrystallization from acetone gave 11-ketopregnane-3α,17α,20β-triol, m.p. 218–220.5°; reported m.p. 179° and 220°.[20] A small amount (50 mg.) of pregnane-3α,11β,17α,20β-tetrol 3,20-diacetate was eluted from the chromatogram. The reduction of 3α,17α-dihydroxypregnane-11,20-dione with sodium borohydride under the same conditions gave essentially the same result.

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(19) M. Finkelstein, J. v. Euw and T. Reichstein, *Helv. Chim. Acta*, **36**, 1266 (1953).

(20) L. H. Sarett, *J. Am. Chem. Soc.*, **71**, 1169 (1940).

[CONTRIBUTION FROM THE INSTITUTE OF PAPER CHEMISTRY]

Liriodendrin, a New Lignan Diglucoside from the Inner Bark of Yellow Poplar (*Liriodendron tulipifera* L.)

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A new di-β-D-glucoside was isolated from an alcohol extract of the inner bark of yellow poplar, *Liriodendron tulipifera* L., in yields of 0.05–0.08% of the fresh bark. The glucoside was colorless, odorless, tasteless, crystalline, m.p. 269–270°, and was hydrolyzed by dilute acids to D-glucose and a new lignan. The name "liriodendrin" is suggested for the glucoside and "lirioresinol" for the lignan. Liriodendrin octaacetate and octamethyl ether were prepared as crystalline substances. Lirioresinol was obtained in two forms, lirioresinol-*A* and -*B* from which the corresponding crystalline dimethyl and dibromodimethyl ethers were prepared. The dibromodimethyl ethers were degraded to 4-bromo-5,6-dinitropyrogallol trimethyl ether and bis-(hydroxymethyl)succinic acid dilactone to establish lirioresinol as a tetrahydro-1,4-bis(4-hydroxy-3,5-dimethoxyphenyl)-furo[3,4-*c*]furan, stereoisomeric with syringaresinol, and liriodendrin the corresponding di-β-D-glucoside. A diastereoisomeric form, lirioresinol-*C*, was obtained upon hydrolysis of liriodendrin with crude almond emulsin.

Introduction. The yellow poplar or tulip tree, *Liriodendron tulipifera* L., is ranked among the most beautiful and valuable of the hardwoods which are native to the North American continent.[1] The Indians made canoes from its strong, light wood. The colonists used the tree extensively for lumber, and developed the use of its bark for medicinal purposes. Morel and Totain[2] stated that without extracts of yellow poplar bark as a substitute for quinine, the War of Independence might have been lost!

During the 19th century, European scientists studied the extractives of the yellow poplar's wood and bark, but the isolation of specific substances was rarely reported.[3] In 1831, Emmet[4] isolated 2–3% of a bitter principle, from the fresh, winter-gathered root bark. He named the substance "liriodendrine," but it has not been reported by later investigators. Bouchardat[5] isolated a crystalline material which was alkaloidal in character but which was not further described. The Lloyds[6] named a material "tulipiferin" which, though not crystalline, was apparently an alkaloid. Since then the extractives of this tree have remained essentially uninvestigated, but the increasing utilization of yellow poplar along with other hardwoods for pulp and paper has renewed interest in its chemistry.

Studies in progress at The Institute of Paper Chemistry indicate that alcoholic extracts of fresh yellow poplar bark consist largely of sugars, and of lesser amounts of unknown phenolic substances, coloring matter, and an essential oil with a distinctive pleasant odor. In addition to these materials, a new colorless substance was crystallized from the extracts in amounts of 0.05–0.08% based on the fresh bark. This substance has been characterized as a di-β-D-glucoside of a new lignan built on a nucleus of tetrahydrofurofuran. *Liriodendrin* is proposed for the name of the glucoside, and *lirioresinol* for the lignan.

Lignans derived from tetrahydrofurofuran. A group of naturally occurring phenylpropane dimers which are linked through the beta-carbon atoms of the side chains are known as lignans, a compre-

(1) C. D. Mell, *Textile Colorist*, **63**, 349 (1941).

(2) P. Morel and P. Totain, *Assoc. franc. avance. sci. Congrés Nîmes, 41 Session*, 810 (1912).

(3) C. Wehmer, *Die Pflanzenstoffe*, p. 336, Jena, G. Fischer, 1929; J. von Wiesner, *Die Rohstoffe des Pflanzenreichs*, Vierte auflage, p. 146. Leipzig, W. Engelmann, 1927.

(4) J. P. Emmet, *J. pharm. chim.*, **17**, 334, 400 (1831).

(5) A. Bouchardat, *Bull. de therapeut.*, **19**, 243 (1842).

(6) J. U. Lloyd and C. G. Lloyd, *Pharm. Rundsch.*, **4**, no. 8, 169–72 (1886); *Jahresber. Pharm.*, **46**, 61 (1886); *Am. Druggist*, **15**, no. 6, 101 (June, 1886).