Anal. Calcd. for $C_{23}H_{33}NO_6$: C, 65.85; H, 7.93; N, 3.34; O, 22.88. Found: C, 66.05; H, 7.62; N, 3.45; O, 22.78.

Pregna-3,5-diene-17α,21-diol-11,20-dione 21-Acetate (V).—Hydrolysis of the 17α,20,20,21-bismethylenedioxy group and elimination of the 3-acetate from 4-pregnene-3,17α,21-triol-11,20-dione 3-acetate (IIIa) (0.5 g.) was effected by heating with 50% aqueous acetic acid (100 cc.) on the steam bath under a nitrogen atmosphere for 24 hr. After removal of the acid under vacuum and extraction with ethyl acetate, the solution was washed to neutrality and evaporated. The residue was acetylated at room temperature with acetic anhydride and pyridine for 20 hr. Isolation of the crude acetate by extraction with ethyl acetate and chromatography on silica gel (30 g.) furnished two compounds: a) **Pregna-3,5-diene-17α,21-diol-11,20-dione-BMD, IVb** (50 mg., m.p. 150–160°) eluted with benzene–hexane (9:1) and crystallized from ether–hexane to yield needles with m.p. 170–172°, $[\alpha]_D$ −164°, λ_{max} 228, 234 mμ, log ε 4.25, 4.27, and ν_{max} 1710 cm.$^{-1}$.

Anal. Calcd. for $C_{23}H_{30}O_5$: C, 71.48; H, 7.82. Found: C, 71.44; H, 7.76.

b) **Pregna-3,5-diene-17α,21-diol-11,20-dione 21-Acetate (V)** was isolated by elution with benzene–ether (30 mg.). It had m.p. 154–156°, raised to 173–175° by one additional crystallization, and $[\alpha]_D$ +26°, λ_{max} 228, 234, 290–292 mμ, log ε 4.27, 4.28, 2.13, ν_{max} 3500, 1720, 1260 cm.$^{-1}$. This compound gave a positive α-ketol reaction with triphenyltetrazolium chloride.

Anal. Calcd. for $C_{23}H_{30}O_5$: C, 71.48; H, 7.82; O, 20.70. Found: C, 71.15; H, 7.92; O, 21.06.

The Cleavage Reaction of 16-Oximino-17-keto Steroids[1]

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Received July 19, 1961

16-Oximinoandrostane-3β-ol-17-one (Ib) and its 5,6-dehydro analog Ia form oxime acetates, the structures, reactions, and spectral characteristics of which have been determined. Cleavage of the oxime acetates was found to be extremely facile, aqueous solvents giving nitrile acids, anhydrous alcohols yielding nitrile esters. The structure of the products was proved by independent synthesis and chemical conversions. Interpretation of the results to reconcile previously reported data is given. Spectral properties of the oxime acetates are discussed.

Kendall and co-workers[2] and later Regan and Hayes[3] found that the acylation of 16-oximino-5-androstene-3β-ol-17-one (Ia) with acetic anhydride gave a product which was assigned the structure 3β - acetoxy - 16 - oximino - 5 - androstene - 17-one (IIa) on the basis of elemental analysis and conversion with thionyl chloride, conditions for a Beckmann transformation, to a ring D imide XVIa. Recently Heard, Ryan, and Bolker,[4] evidently unaware of the previous work,[2,3] reported that acylation of 16-oximinoandrostane-3β-ol-17-one (Ib), the 5,6-dihydro derivative of Ia, gave a diacetate which was assigned the structure of the Beckmann rearrangement intermediate XVb.[5] This assignment was based[4] on the hypsochromic shift in the ultraviolet absorption maximum on the conversion of the oxime Ib to the acetate XVb, and on the reaction of XVb with base to form a compound, soluble in sodium carbonate, and believed to be the imide XVIb.

In the course of related studies the acylation of 16-oximino-5-androstene-3β-ol-17-one (Ia) was attempted in this laboratory and gave two products A and B which corresponded in analyses to a monoacetate (B) and a diacetate (A). Compound B was identical[6] with the material isolated by Kendall[2] and Regan[3]; compound A was very unstable and was converted readily into compound B by aqueous acid or base, on recrystallization from aqueous solvents or even on standing in air. This explains the failure of these authors[2,3] to isolate A. Compound A was shown to be analogous to the diacetate obtained from Ib by Heard[4] (*vide infra*).

The incorrectness of the assignment of structure IIa to compound B was immediately obvious from the infrared spectrum which showed absorption bands at 2250 cm.$^{-1}$, characteristic of a nitrile group, and at 2600–2700 cm.$^{-1}$ and 1700 cm.$^{-1}$ indicative of a carboxy group. These results and the chemical properties of the material, namely formation of a methyl ester and solubility in aqueous sodium carbonate, suggested the structures VIa or Xa for compound B.

The direct cleavage[7] of Ia or IIIa would be expected to lead to VIa with a primary nitrile function; however, the resistance of the nitrile function of B to hydrolysis required the consideration of structure Xa with a tertiary nitrile.[8] Thus,

(1) Presented in part before the Division of Organic Chemistry at the 138th Meeting of the American Chemical Society, New York, N. Y., September, 1960.

(2) F. H. Stodola, E. C. Kendall, and B. F. McKenzie, *J. Org. Chem.*, **6**, 843 (1941).

(3) B. M. Regan and F. N. Hayes, *J. Am. Chem. Soc.*, **78**, 639 (1956).

(4) R. D. Heard, M. T. Ryan, and H. I. Bolker, *J. Org. Chem.*, **24**, 172 (1959).

(5) These results have subsequently been quoted as evidence for the isolation of a Beckmann rearrangement intermediate, *cf.*, A. R. Surrey, "Name Reactions in Organic Chemistry," 2nd ed., Academic Press, New York, 1961, p. 15.

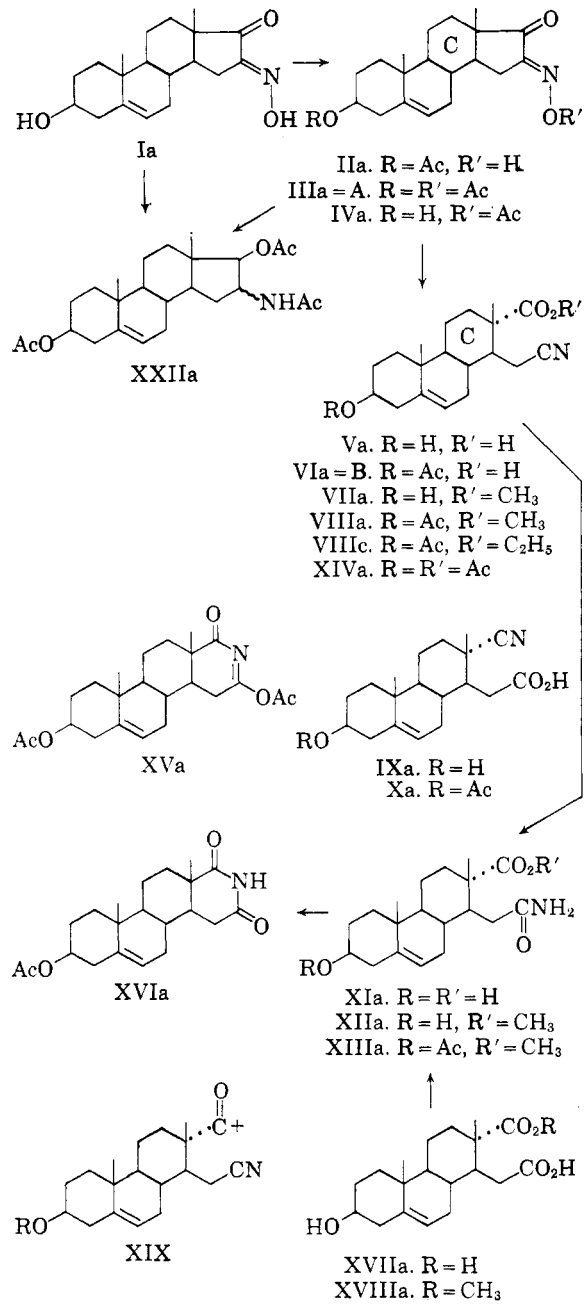
(6) We are indebted to Dr. Regan for samples of Ia and its acetate used for comparison of melting points and infrared spectra with our materials.

(7) Such reactions have been referred to as "second-order Beckmann rearrangement" (*cf.*, ref. 12, 13), but we feel a more appropriate term is "cleavage of oximes."

(8) Mechanistically it is possible to conceive of a path leading from IIIa to Xa *via* intermediates of type XVa.

upon heating for twenty-four hours in methanolic potassium hydroxide or upon warming with alkaline hydrogen peroxide,[9] the acetate B was converted into an alcohol (Va or IXa) with the nitrile group still intact. Hydrolysis under acid conditions led to dehydration in the A-ring of the steroid. The hydrolysis of the nitrile group in B was accomplished by heating with potassium hydroxide in glycerol to yield the known[10] 3-β-hydroxy-16,17-seco-5-androstene-16,17-dioic acid (XVIIa). The reaction of 3β-hydroxy-16,17-seco-5-androstane-16-nitrile-17-oic acid (Va) with potassium hydroxide in 95% ethanol caused partial conversion to the amide acid XIa. The structure of the latter was proved by conversion to methyl 3β-acetoxy-16,17-seco-5-androstene-16-amide-17-oate (XIIIa) which was prepared by an alternate route from authentic 3β-hydroxy-16,17-seco-5-androstene-16,17-dioic acid 17-methyl ester (XVIIIa). This confirmed the structure of B as VIa. The tertiary nature of the carbomethoxy function in VIIIa, the methyl ester of VIa, was also indicated by selective hydrolysis to methyl 3β-hydroxy-16,17-seco-5-androstene-16-nitrile-17-oate (VIIa). The resistance of the nitrile function in VIa to mild hydrolysis in basic solution is probably due to a proximity effect by the carboxylate ion.

We established the structure of the intermediate diacetate A as IIIa rather than as the Beckmann rearrangement intermediate XV proposed by Heard,[4] by proving that the steroid nucleus was still intact in A. Reduction of A with lithium aluminum hydride gave an amine identical with that obtained by the reduction of the oxime Ia.[11] The diacetate A was partially hydrolyzed on neutral alumina to the monoacetate IIa which in turn could be saponified to 16-oximino-5-androstene-3β-ol-17-one (Ia). This showed that acylation had not changed the ring system of Ia, and that A must have structure IIIa in which the steroid ring system was retained. 16-Acetoximino-5-androstene-3β-ol-17-one (IVa) the other possible monoacetate of Ia was prepared from the latter by careful acetylation in dioxane in the absence of pyridine. The oxime acetate IVa resembled the diacetate IIIa but not the monoacetate IIa in its chemical behavior. It was easily converted to the nitrile acid Va upon treatment with base or aqueous acetone. Acids VIa and Va were interconvertible by hydrolysis and acetylation, respectively. The fact that the oxime acetylates more readily than the 3-hydroxyl group in 16-oximino-5-androstene-3β-ol-17-one (Ia) and that the oxime acetate hydrolyzes more readily than the 3-acetate in 3β-acetoxy-16-acetoximino-5-androstene-17-one (IIIa) is consistent with expectations.



Series b is the 5,6-dihydro derivative of series a.

With the establishment of the structure of A and B an interpretation of the reactions observed by Kendall,[2] Regan,[3] and Heard[4] was possible. The labile 3β-acetoxy-16-acetoximinoandrostane-17-one (IIIb), obtained on acetylation of oxime Ib, was cleaved to nitrile acids VIb or Vb on treatment with aqueous sodium bicarbonate or potassium hydroxide, respectively. Nitrile acid Vb was unaffected by alkaline hydrogen peroxide but was converted by more vigorous hydrolysis into a

(9) K. B. Wiberg, *J. Am. Chem. Soc.*, **75**, 3961 (1953); most nitriles can be hydrolyzed to amides under these conditions.

(10) (a) S. Kuwada, *J. Pharm. Soc. Japan*, **56**, 75 (1936); (b) A. Butenandt, S. Schmidt-Thome, T. Weiss, D. von Dressler, and U. Meinerts, *Ber.*, **72**, 417 (1939); (c) A. Wettstein, H. Fritzsche, F. Hunzicker, and K. Miescher, *Helv. Chim. Acta*, **24**, 332E (1941).

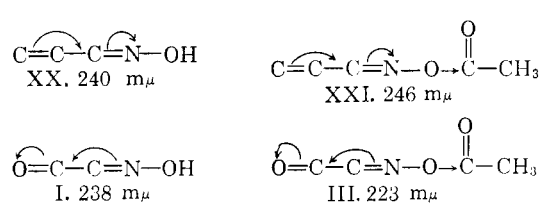
(11) When this investigation was practically completed we became aware of T. A. Lies' dissertation [*Dissertation Abstr.*, **19**, 1565 (1959)] where the acetylation of Ia is described. The diacetate product was assigned structure IIIa on the basis of hydride reduction in low yield followed by acetylation to an aminodiol triacetate identical with XXIIa obtained *via* Ia. The conversion of Ia into IVa and of IIIa into VIa was also reported there without structure proof for the latter. We are grateful to Dr. Lies for making his Ph.D. thesis available to us.

mixture of amide acid XIb and diacid XVIIb. These results coupled with the information gained in the 5,6-dehydro series led to the following conclusions. The conversion of Heard's[4] imidoyl ester XVb by alcohol to an oil with loss of absorption in the ultraviolet followed by base hydrolysis to an imide XVIb, soluble in aqueous sodium carbonate, is actually the cleavage of IIIb to the nitrile ester VIIIb followed by hydrolysis to the carbonate soluble acid Vb. Further hydrolysis of the latter leads to XIb and XVIIb.

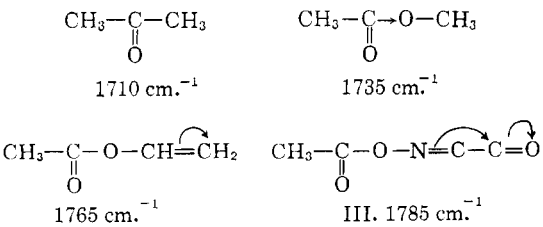
3β-Acetoxy-16,17-seco-5-androstene-16,17-dioic acid imide (XVIa) had previously[3] been prepared by what was believed to be a Beckmann rearrangement of oxime IIa with thionyl chloride. Since these workers actually had the nitrile acid VIa rather than the isomeric oxime IIa in hand (*vide supra*), ring closure to the imide must take place *via* an acid chloride of VIa followed by ring closure and hydrolysis. In this laboratory the reaction of VIa with thionyl chloride in the cold or at room temperature followed by work-up with methanol[3] led to the imide XVIa as well as to methyl 3β-acetoxy-16,17-seco-5-androstene-16-nitrile-17-oate (VIIIa). The imide XVIa was also obtained from the amide ester XIIIa, upon attempts to dehydrate the latter to the nitrile ester VIIIa by means of potassium hydrogen sulfate or by means of sodium borohydride.

The easy conversion of α-ketoacetoximes III and IV to nitrile acids VI and V can be best explained as a solvolysis reaction passing through an intermediate state[12] of type XIX on the basis of the following facts. 16-Oximino-5-androstene-3β-ol-17-one (Ia) did not undergo Beckmann rearrangement nor cleavage when heated with polyphosphoric acid or with concentrated hydrochloric acid in dioxane; yet cleavage of its monoacetate IVa or diacetate IIIa occurred with aqueous organic solvents and proceeded faster in the presence of acid or of increased ionic strength of the medium. Attack by water as a nucleophile at the hindered C-17 position in IIIa is unlikely. Solvolysis of 3β-acetoxy-16-acetoximino-5-androstene-17-one (IIIa) in absolute methanol or ethanol[13] rather than in aqueous solvents led to the methyl ester VIIIa or ethyl ester VIIIc, respectively. Methyl 3β-acetoxy-16,17-seco-5-androstene-16-nitrile-17-oate (VIIIa) thus prepared was identical with the diazomethane reaction product of VIa.

The *anti* configuration of the oximino hydroxyl group with respect to the 17-carbonyl in Ia and Ib is indicated by a shift in the ultraviolet[14] from 240 mμ to 288 mμ upon addition of base to these compounds. Similarly the hypsochromic shift in the ultraviolet observed in going from the α-oximino ketone I (λ_{max} 238 mμ) to its acetate III (λ_{max} 233 mμ) seems to be general for a system[14] where the oximino hydroxyl is *anti* to the keto group. An explanation for the latter phenomenon lies in the recognition that the chromophore in α-oximinoketones (*cf.* I) is polarized in a different direction as is the chromophore in α,β-unsaturated oximes (*cf.* XX). In the latter, (*e.g.*, oximes of Δ^4-3-ketones), acetylation of the oxime causes a bathochromic shift of a few millimicrons (see XX–XXI), and it was this fact that led Heard and co-workers[4] to propose an alternate structure XVb rather than IIIb for the diacetate obtained from 16-oximinoandrostane-3β-ol-17-one (Ib). This bathochromic shift can be rationalized on the basis that the electron-with-



drawing properties of acetyl are in the same direction as the polarization of the conjugated system (see arrows). In III, however, the polarization of acetyl is in the direction opposite to the polarization of the original chromophore in the α-keto oxime I and a hypsochromic shift can be expected. An interpretation of the infrared spectra of the acetoximes IIIa and IVa leads to the same conclusion. These compounds have a strong band at 1785 cm.$^{-1}$, due to the carbonyl absorption of the oximino acetate. A shift to higher wave numbers can be observed when a group with electron withdrawing properties is attached to a carbonyl. This requires a higher force constant or carbonyl stretching frequency and thus a polarization as in III is indicated.



The nature of the reactions of the esters of benzil monoximes and of α-oximinocamphor with base has been shown to be indicative of the stereochemistry of the oxime.[15] Thus the esters of the

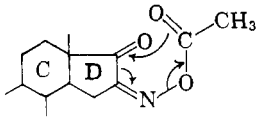
(12) There has been some discussion recently [J. P. Freeman, *J. Org. Chem.*, **26**, 3507 (1961)] whether cleavage of oximes (*cf.*, ref. 7) proceeds by a concerted process or *via* a carbonium ion of type XIX.

(13) This reaction is analogous to the conversion of α-oximino ketones to nitriles and esters by means of acetic anhydride–sodium ethoxide as reported independently by A. F. Ferris, *J. Org. Chem.*, **25**, 12, 1813 (1960).

(14) We have made similar observations on *anti*-α-oximinocamphor and *anti*- but not *syn*-benzil monoxime. D. H. R. Barton and J. M. Beaton, *J. Am. Chem. Soc.*, **83**, 4083 (1961) found similar results with the two isomers of 4-oximino-3-keto steroids. We are indebted to Prof. Barton for communication of these results prior to publication.

(15) (a) R. P. Barnes and A. H. Blatt, *J. Am. Chem. Soc.*, **57**, 1330 (1935); (b) J. Meisenheimer and W. Theilacker, *Ann.*, **493**, 33 (1932).

anti oximes underwent cleavage while the *syn* isomers were hydrolyzed. The similarity of the reactions of IIIa with those of the above-mentioned *anti* oximes offers support to the stereochemical assignment based on spectral data (*vide supra*). Furthermore the possibility of an intramolecular reaction of a *cis* isomer of IIIa to give XIVa as an intermediate was eliminated. Thus the mixed anhydride XIVa was shown to give the nitrile acid VIa, rather than the ester VIIIa, on reaction with methanol, while IIIa underwent cleavage in methanol to give VIIIa (*vide supra*).



cis IIIa

Experimental[16]

16-Oximino-5-androstene-3β-ol-17-one (Ia).—To a solution of 0.223 mole of potassium in *t*-butyl alcohol (380 ml.) under nitrogen was added 8.0 g. (0.0277 mole) of 5-androstene-3β-ol-17-one[17] followed by two 8-ml. portions of isoamyl nitrite, the latter being added 2 hr. apart. The resulting orange salt was stirred with ice water and the aqueous solution extracted with two 20-ml. portions of ether. The alkaline aqueous solution was acidified with concd. hydrochloric acid in the presence of chloroform. The aqueous layer was further extracted with chloroform until colorless. The combined chloroform layers were in turn extracted with 0.5 *N* potassium hydroxide until the organic phase remained colorless.[18] Acidification of the aqueous extracts yielded 7.4 g. of crude oximino ketone Ia, m.p. 215–220° dec.

The acidic, aqueous portion remaining after chloroform extraction yielded upon standing an additional 375 mg. of Ia (total 90%), m.p. 234–236° dec., and 2% of Va (*vide infra*). Recrystallization from methanol gave white needles (66% yield) melting at 242.5–244° dec. The analytical sample melted at 250–251° dec. (lit.,[2,3] m.p. 248–249° dec.).

Anal. Calcd. for $C_{19}H_{27}O_3N$: C, 71.89; H, 8.57; N, 4.41. Found: C, 72.04; H, 8.63; N, 4.48.

ν_{max} 3400 (3 OH), 3200 (Oxime-OH), 1740 (17 C=O), 1640 cm.$^{-1}$ (C=N). λ_{max} 238 mμ (ε 10,400); upon addition of aqueous potassium hydroxide the maximum shifted to 288 mμ. $[\alpha]^{25}D$ −77.8° ± 2 (*c* 2, in chloroform).

Various fractions obtained by recrystallization of Ia melted at 239° dec., 242–244° dec., or 245–247° dec., but all had an infrared spectrum identical to that of the analytical sample.

16-Acetoximino-5-androstene-3β-ol-17-one (IVa).—A solution of 200 mg. of oxime Ia and 2.8 ml. of acetic anhydride in 20 ml. of dry dioxane was kept at room temperature in the dark for 2 days. Upon decomposition with water 214 mg. of oxime acetate IVa, m.p. 157–163°, was isolated. Two recrystallizations from dry ethyl acetate gave an analytical sample, m.p. 168.5–170.5°.

Anal. Calcd. for $C_{21}H_{29}O_9N$: C, 70.17; H, 8.13; N, 3.90; O, 17.80. Found: C, 70.09; H, 8.13; N, 3.89; O, 18.04.

ν_{max} 3430 (OH), 1775 (C=O of oximino acetate), 1740 (17 C=O), 1640 cm.$^{-1}$ (C=N). λ_{max} 223 mμ (ε ~ 9000).

The same product was also obtained by stirring in acetic anhydride for 4 days under nitrogen.

3β-Acetoxy-16-acetoximino-5-androstene-17-one (IIIa).

A. From Ia.—A solution of 300 mg. of oxime Ia in 4 ml. of dry pyridine and 4.5 ml. of acetic anhydride was allowed to stand for 24 hr., then poured into ice water or into dilute hydrochloric acid. The product (330 mg.) melted at 163–165° and was recrystallized from ether (distilled from lithium aluminum hydride) yielding 110 mg. of IIIa., m.p. 173–175°.

Anal. Calcd. for $C_{23}H_{31}O_5N$: C, 68.80; H, 7.78; N, 3.49. Found: C, 68.69; H, 7.84; N, 3.54.

ν_{max} 1785 and 1755 (C=O of oximino acetate), 1730 (acetate C=O), 1640 cm.$^{-1}$ (C=N). λ_{max} 223 mμ (ε ~ 10,000); ε decreased steadily in alcohol solution, faster in the presence of hydrochloric acid, sulfuric acid, pyridine, or sodium methoxide. $[\alpha]^{25}D$ −89.4° ± 2° (*c* 2.1, in chloroform).

The product is readily cleaved by aqueous acid or base, or by heating with aqueous organic solvents but may be stored in the dark over phosphorus pentoxide under reduced pressure for at least 6 months.

B. From IIa.—Monoacetate IIa (60 mg.) was treated with acetic anhydride in dioxane. After the usual work-up 45 mg. of crude diacetate IIIa, m.p. 160–162°, was collected by filtration.

C. From IVa.—The diacetate IIIa was prepared from monoacetate IVa by heating with acetic anhydride on the steam bath until the solid had dissolved (1 hr.) or by allowing IVa to stand for 36 hr. in acetic anhydride–pyridine at room temperature. After the usual work-up, crude diacetate IIIa was obtained in 85% yield.

3β-Acetoxy-16-oximino-5-androstene-17-one (IIa).—Diacetate IIIa (282 mg.) in chloroform was chromatographed over neutral Woelm aluminum oxide. From the fractions eluted with chloroform and with chloroform–ethyl acetate there were obtained 60 mg. of pure monoacetate IIa, m.p. 221.5–222.5° dec.

From later chromatographic fractions there was also obtained impure cleavage product containing the nitrile group (by infrared).

Hydrolysis of IIa to Ia.—A portion of 20 mg. of IIa was warmed in a 0.5 *N* potassium hydroxide solution for 2 hr. Acidification gave a solid melting at 238–240° dec., identical by infrared and melting point with authentic 16-oximino-5-androstene-3β-ol-17-one (Ia).

3β-Acetoxy-16,17-seco-5-androstene-16-nitrile-17-oic Acid (VIa). A. From Ia *via* IIIa.—Recrystallization from aqueous methanol of the crude product from the preparation of IIIa gave VIa in 79% yield as white needles, m.p. 174–176°. Recrystallization from aqueous acetone and then from ethyl acetate raised the melting point[6] to 184–185°.

Anal. Calcd. for $C_{21}H_{29}O_4N$: C, 70.17; H, 8.13; N, 3.90. Found: C, 69.39, 69.23; H, 8.11, 8.05; N, 4.10. Neut. equiv.: Calcd. 359; Found: 349.

ν_{max} 2700–2600 (bonded OH of CO_2H), 2250 (C≡N), 1735 (acetate C=O), 1700 cm.$^{-1}$ (CO_2H).

B. From Va.—Acetylation of the 3-hydroxynitrile acid Va with acetic anhydride–pyridine for 14 hr. at room temperature yielded the 3-acetoxynitrile acid VIa, m.p. 179–180°. The product showed no melting point depression when mixed with authentic VIa.

3β-Hydroxy-16,17-seco-5-androstene-16-nitrile-17-oic Acid Va. A. From IIIa.—Hydrolysis of 50 mg. of acetoxime IIIa with 0.25 *N* potassium hydroxide solution at room temperature (90 min.) yielded 35 mg. of silvery crystals of Va. The product sintered at 118° and melted at 178–180°. Larger quantities of Va were synthesized in 92% over-all

(16) Melting points were taken on a Fisher melting block and are uncorrected. Analyses were performed by Pascher Laboratories, Bonn, Germany. Ultraviolet spectra were run in methanol. Infrared spectra were run in potassium bromide pellets unless otherwise indicated.

(17) We are indebted to Dr. Mancera (Syntex Co.) for generous samples of the material.

(18) M. Huffman and N. Lott, *J. Am. Chem. Soc.*, **76**, 4038 (1954); *J. Biol. Chem.*, **207**, 431 (1954).

yield by acetylation of oximino ketone Ia with acetic anhydride in pyridine as described above, and stirring the crude diacetate IIIa with aqueous alkali.

B. From VIa.—The 3β-acetoxynitrile acid VIa (100 mg.) was stirred into 0.25 *N* aqueous potassium hydroxide until it had dissolved, or was heated in 5% methanolic potassium hydroxide under reflux for 1 hr.. Usual work-up yielded 79 mg. of Va, m.p. 176–178°, with partial sintering and resolidification in the range of 115–125°. Two recrystallizations from ethyl acetate gave an analytical sample of Va melting at 178–179° with partial sintering and resolidification at 125°. Mixed with starting material (VIa) it melted at 165–170°.

Anal. Calcd. for $C_{19}H_{27}O_3N$: C, 71.89; H, 8.57; N, 4.41. Found: C, 71.95, 72.09; H, 8.78; 8.78; N, 4.36.

ν_{max} 3400 (OH); 3280 and 2650–2550 (carboxy OH); 2250 (C≡N); 1710 cm.$^{-1}$ (CO_2H).

C. From IVa.—Hydrolysis of monoacetate IVa (150 mg.) with 0.5 *N* aqueous potassium hydroxide for 1 hr. yielded some unchanged starting material (12 mg.), m.p. 171–172° dec., and mainly nitrile acid Va (109 mg.), m.p. 180–180.5°.

D. From VIIa.—When a solution of 95 mg. of 3-hydroxymethyl ester nitrile VIIa was heated in 12.5% methanolic potassium hydroxide for 3 hr., the product isolated (64 mg., m.p. 172–174°) was Va, identified by melting point and infrared spectrum.

Methyl 3β-Acetoxy-16,17-seco-5-androstene-16-nitrile-17 oate (VIIIa). A. From VIa *via* Its Acid Chloride.—The nitrile acid Va (200 mg.) was treated at ice bath temperature with thionyl chloride for 2 hr. The residue from evaporation under reduced pressure was recrystallized from methylene chloride, then from methanol and finally from ethyl acetate and 50 mg. of product VIIIa melting at 177–178° was obtained. Its melting point was depressed by 10° on admixture with VIa.

Anal. Calcd. for $C_{23}H_{31}O_4N$: C, 70.75; H, 8.37; N, 3.75. Found: C, 70.95; H, 8.44; N, 3.64.

ν_{max} 2250 cm.$^{-1}$ (C≡N), 1725 cm.$^{-1}$ ($OCOCH_3$ and CO_2CH_3).

Reaction of VIa with thionyl chloride at room temperature for 16 hr. led to the isolation of some imide XVIa in addition to nitrile methyl ester VIIIa.

B. From VIa with Diazomethane.—To a solution of 200 mg. of nitrile acid VIa in ether there was added a solution of excess diazomethane in the same solvent. After usual work-up and recrystallization from ethyl acetate the ester VIIIa (m.p. 177–177.5°) was obtained in 60% yield.

C. From IIIa.—A solution of 100 mg. of oxime acetate IIIa in 10 ml. of absolute methanol was heated under reflux for 8 hr. The volume was reduced to one-fourth and 78 mg. of white needles of VIIIa, m.p. 176–177°, was collected by filtration.

D. From VIIa.—3-Hydroxynitrile methyl ester VIIa (80 mg.) was treated with acetic anhydride in pyridine solution at room temperature for 48 hours. Work-up with water afforded 76 mg. of crude product, m.p. 168–173°. Recrystallization from methanol gave 50 mg. of VIIIa, m.p. 177–178°.

Methyl 3β-Hydroxy-16,17-seco-5-androstene-16-nitrile-17-nitrile-17-oate (VIIa). A. From VIIIa by Partial Hydrolysis.—When 110 mg. of acetate methyl ester VIIIa was heated under reflux in a 1.5% solution of potassium hydroxide in methanol for 3.5 hr. and worked up as usual, the product (60 mg.) proved to be the alcohol VIIa with the methyl ester grouping still intact. Recrystallization from ethanol gave white crystals, m.p. 150–152°, ν_{max} 3505 cm.$^{-1}$ (OH), 2250 cm.$^{-1}$ (C≡N), 1725 cm.$^{-1}$ (CO_2CH_3). The ester VIIa was converted by hydrolysis in 12.5% methanolic potassium hydroxide to the acid Va as described above.

B. From IVa.—The cleavage of oxime acetate IVa to nitrile ester VIIa, m.p. 148–151°, was accomplished in 60% yield by heating with methanol in a manner analogous to the cleavage of IIIa to VIIIa.

Ethyl 3β-Acetoxy-16,17-seco-5-androstene-16-nitrile-17-oate (VIIIe).—Absolute ethanol (3 ml.) was added to a solution of 117 mg. of the diacetate IIIa in pyridine (3 ml.). After 2 days the solution was worked up as usual and 38 mg. of VIIIc, m.p. 115–116.5° was obtained. The analytical sample (from ethanol) melted at 117–118°. ν_{max} 2240 cm.$^{-1}$ (C≡N), 1735 cm.$^{-1}$ (C=O of acetate), 1720 cm.$^{-1}$ (C=O of $CO_2C_2H_5$).

Anal. Calcd. for $C_{23}H_{33}O_4N$: C, 71.29; H, 8.58; N, 3.61. Found: C, 71.09; H, 8.57; N, 3.73.

Hydrolysis of Va to 3-β-Hydroxy-16,17-seco-5-androstene-16,17-dioic Acid (XVIIa).—A solution of 150 mg. of the acetoxynitrile acid VIa in 2% of methanolic potassium hydroxide was heated under reflux for 24 hr. The product isolated proved to be the hydroxynitrile acid Va. It was redissolved in a 20% solution of potassium hydroxide in glycerol and heated at 160° for 24 hr., during which time evolution of ammonia could be observed. Work-up with dilute hydrochloric acid gave diacid XVIIa, m.p. 243–252°, which after recrystallization from methanol–water melted at 227–228°, $[\alpha]^{27}D$ −67 ±3° (*c* 2, ethanol); neut. equiv.: 168 (calcd: 168.2). Reported[10] m.p. for XVIIa (from aqueous alcohol) 250–251°, $[\alpha]^{27}D$ −74° ±3°. Elemental analysis indicated one-half mole of water of crystallization.

Anal. Calcd. for $C_{19}H_{28}O_5 \cdot \frac{1}{2}\ H_2O$: C, 66.09; H, 8.43. Found: C, 66.17, 66.32; H, 8.28, 8.43.

ν_{max} 3245 (OH of CO_2H), 3400 shoulder (OH), 2650 (bonded OH of CO_2H), 1700 (CO_2H), 1275 cm.$^{-1}$ (C—O).

The diacid XVIIa, m.p. 249–252°, was obtained on recrystallization of the hemihydrate from glacial acetic acid. The diacid (m.p. 227°) was converted by means of acetic anhydride to the known **3β-acetoxy-16,17-seco-5-androstene-16-17-dioic anhydride** in 80% yield, m.p. 186–187.5°; ν_{max} 1800 and 1750 (anhydride), 1720 cm.$^{-1}$ ($OCOCH_3$); lit.,[10] m.p. 186°.

Anal. Calcd. for $C_{21}H_{28}O_5$: C, 69.97; H, 7.83. Found: C, 69.81; H, 7.53.

Heating of nitrile acid Va in 10% aqueous potassium hydroxide solution for 1 hr. (evolution of ammonia was detected) resulted in a product, the infrared spectrum of which indicated the presence of unchanged nitrile acid Va, diacid XVIIa and amide acid XIa.

Reaction of 3-acetoxynitrile methyl ester VIIIa or of the alcohol VIIa with concd. sulfuric acid at 7° or at room temperature gave after laborious work-up a material melting at 226–235°. Spectra indicated the absence of a nitrile, the presence of an amide, and an ester function but also the presence of a homoannular diene[19] (λ_{max} 266, 277, and 288 mμ (ε 8000, 10,000, and 9000).

Methyl 3β-Acetoxy-16,17-seco-5-androstene-16-amide-17-oate (XIIIa). A.—A solution of 1.58 g. of nitrile acid Va and 8.0 g. of potassium hydroxide in 160 ml. of 95% ethanol was heated under reflux for 20 hr. The volume of the solution was reduced *in vacuo*, water was added, and the resulting solution was washed with ether and then acidified. The suspension of solids formed was shaken with chloroform and the separated aqueous phase was filtered to collect the chloroform-insoluble solids. Thus 600 mg. of amide acid XIa containing some unchanged nitrile acid (by infrared) was obtained. Recrystallization from methanol afforded different crops of material of which 210 mg. was reasonably pure amide acid XIa (as judged by infrared), m.p. 233–236° dec.; ν_{max} 3460, 3430, 3350, 3215, 2650–2550 (OH of CO_2H), 1705 (CO_2H), 1645 and 1570 cm.$^{-1}$ ($CONH_2$). Upon work-up of the chloroform layer there was obtained 815 mg. of nitrile acid Va, m.p. 173–175°. An ether suspension of 190 mg. of 3-hydroxyamide acid XIa, described above, was esterified with diazomethane to yield 150 mg. of white needles melting at 233–236°. The melting point was lowered 12° on admixture with starting XIa. The infrared spectrum was consistent with that expected for

(19) L. Dorfman *Chem. Revs.*, **53**, 54 (1953).

XIIa; 3415, 3200, 1720 (CO_2CH_3), 1670 and 1620 cm.$^{-1}$ ($CONH_2$).

This product (120 mg.) was acetylated in acetic anhydride–pyridine at room temperature. Work-up afforded 105 mg. of crude XIIIa melting at 99–101°. Recrystallization from aqueous methanol in the cold gave two crops of needles. The first crop, 57 mg., melted at 149–151°. The second crop (30 mg.) melted at 120–124°. Further recrystallization of first crop material caused a lowering of the melting point. The infrared spectra of the crude as well as of the two recrystallized fractions were nearly identical. The analytical sample melted at 146.5–148°, $[\alpha]^{28}D$ −85° ± 4° (*c* 0.72, in chloroform); lit.,[20] m.p. 135°, $[\alpha]^{21}D$ −103° ± 4° (*c* 0.637, in chloroform).

Anal. Calcd. for $C_{22}H_{33}O_5N$: C, 67.49; H, 8.50; N, 3.58. Found: C, 67.22; H, 8.62; N, 3.63.

B. From XVIIIa *via* its Acid Chloride.—Acetylation of 3β-hydroxy-16,17-seco-5-androstene-16,17-dioic acid 17-methyl ester (XVIIIa),[21] m.p. 208–210°, (lit.,[22] m.p. 214–216°, lit.,[23] m.p. 200–202°), ν_{max} 3350 (OH), 2650 (bonded OH of CO_2H), 1730 (CO_2CH_3), 1700 (CO_2H), gave 3β-acetoxy-16,17-seco-5-androstene-16,17-dioic acid-17-methyl ester (85%), m.p. 156.5–158.5° (lit.,[21,23] m.p. 167°). ν_{max} 3230 and 2650 (OH of CO_2H), 1725–1740 (esters), 1700 (CO_2H), 1265 cm.$^{-1}$ (C=O of $OCOCH_3$). The acid chloride (490 mg.) of the above acid (90% yield) melted at 126–130° (lit.,[21,23] m.p. 135°) and without purification was treated with anhydrous ammonia in benzene at 15°. Work-up afforded 460 mg. of white powder, m.p. 47–150°. Recrystallization from aqueous methanol gave amide XIIIa as needles melting at 147–149°.

Anal. Calcd. for $C_{22}H_{33}O_5N$: N, 3.58. Found: N, 3.38.

This product was identical, by mixed melting point and infrared, with XIIIa obtained from hydrolysis of nitrile acid Va as described above.

3β-Acetoxy-16,17-seco-5-androstene-16-nitrile-17-oic Ethanoic Anhydride (XIVa).—A solution of 155 mg. of 3β-hydroxy-16,17-seco-5-androstene-16-nitrile-17-oic acid (V) or its acetate VIa in acetic anhydride–pyridine (1:1) was allowed to stand for 48 hr. at room temperature and then was poured into cold 3 *N* hydrochloric acid. The initially formed oil slowly solidified and afforded after washing with water and drying 155 mg. of white powder, m.p. 98–109°. Recrystallization from *n*-heptane–acetone gave feathery needles of XIVa melting at 105.5–107°. ν_{max} 2250 (C≡N), 1817 and 1750 (anhydride), 1735 and 1240 cm.$^{-1}$ ($OCOCH_3$).

Anal. Calcd. for $C_{23}H_{31}O_5N$: C, 68.80; H, 7.78; O, 19.93; N, 3.49. Found: C, 68.85; H, 7.87; O, 20.04; N, 3.51.

When crude XIVa was recrystallized from ethanol or ethanol–water, nitrile acid VIa was formed.

3β-17β-Diacetoxy-17-acetamino-5-androstene (XXIIa). A. By Reduction of Oxime Acetate IIIa.—A solution of 500 mg. of diacetate IIIa in ether was added to lithium aluminum hydride (425 mg.) in dry ether over a period of 2 hr. After heating for 5 additional hours, the mixture was worked up with water and 106 mg. of acid-soluble product, m.p. 195–205°, was isolated from the ether layer.

Acetylation of a portion of this crude product gave a triacetate which upon repeated recrystallization from benzene–petroleum ether (b.p. 60–80°) furnished 10 mg. of needles, m.p. 254–258° dec., identical by infrared spectrum with 3β,17β-diacetoxy-16-acetamino-5-androstene (XXIIa) obtained by hydride reduction of Ia. Further material isolated from the reduction reaction was difficult to purify and was not characterized.

B. By Reduction of Oxime Ia.—16-Oximino-5-androstene-3β-ol-17-one (Ia) was heated with lithium aluminum hydride in tetrahydrofuran for 26 hr., and the product was worked up as usual. The basic material obtained in 50% yield was acetylated with acetic anhydride in pyridine.

The product XXIIa was recrystallized from ethyl acetate and melted at 257–258° (lit.,[11] m.p. 249–254°). ν_{max} 3300 (NH), 1730 (C=O of acetate), 1640 and 1550 cm.$^{-1}$ (CONH).

Anal. Calcd. for $C_{25}H_{37}O_5N$: C, 69.57; H, 8.64; N, 3.25. Found: C, 69.65; H, 8.45; N, 3.27.

3β-Acetoxy-16,17-seco-5-androstene-16,17-imide (XVIa).—Attempts to dehydrate amide XIIIa to nitrile VIIIa by means of potassium hydrogen sulfate or of sodium borohydride[24] led to formation of imide XVIa. Thus, when 87 mg. of XIIIa was heated with an equal weight of potassium hydrogen sulfate at 130° for 12 min., then at 140–150° for 30 min., 72 mg. of imide XVIa, m.p. 242–252°, was obtained. Upon heating of a solution of 98 mg. of XIIIa with 9.5 mg. of sodium borohydride in 6 ml. of diethylene glycol dimethyl ether at 115° for 20 min., there was obtained 22 mg. of imide XVIa, m.p. 258–260°, and 44 mg. of starting material.

The imide obtained in the above dehydration procedures was identical by infrared with imide XVIa (25%) obtained upon action of thionyl chloride on nitrile acid VIa for 16 hr. at 25°, m.p. 262–264°, upon recrystallization from ethyl acetate, $[\alpha]^{28}D$ −128° ± 2° (*c* 1.79 in chloroform); lit.,[3] m.p. 257–259°, $[\alpha]^{22}D$ −134° ± 4°. ν_{max} 3290 (NH), 1730, 1270 and 1240 (acetate), 1685 cm.$^{-1}$ (imide). $\nu_{max}^{CHCl_3}$ 3300, 1720, 1690 cm.$^{-1}$.

The imide XVIa was insoluble in dilute sodium carbonate or hydrochloric acid solution but dissolved in 0.5*N* aqueous potassium hydroxide on warming.

16-Oximinoandrostane-3β-ol-17-one (Ib).—The procedure described for the preparation of Ia was used. From 2.36 g. of 3β-acetoxyandrostane-17-one there was obtained 1.80 g. of crude Ib, m.p. 227–228° dec. Recrystallization from aqueous methanol afforded three crops of needles melting at 236–238° dec. (980 mg.), 236–238° dec. (650 mg.), and 213–216° dec. (30 mg.), respectively. Lit.,[4,18] m.p. 218–219.5° dec., 220° dec.

An infrared spectrum of material melting 236–238° dec. showed the expected absorption at 3400 (OH), 3150 oxime OH), 1740 (C=O), 1640 cm.$^{-1}$ (C=N). λ_{max} 238 mμ (ε 10,000); after addition of potassium hydroxide λ_{max} 288 mμ (ε 14,000).

Anal. Calcd. for $C_{19}H_{29}O_3N$: C, 71.44; H, 9.15; N, 4.39. Found: C, 71.34; C, 71.34; H, 9.11; N, 4.45.

3β-Acetoxy-16-acetoximinoandrostane-17-one (IIIb).—Acetylation of 135 mg. of oxime Ib yielded 193 mg. of crude diacetate IIIb, m.p. 162–164°. Recrystallization from anhydrous ether gave 70 mg. of needles, m.p. 166–167° (lit.,[4] m.p. 163–165°). The infrared spectrum showed characteristic absorption peaks at 1785 (C=O of oximino acetate), 1755 (shoulder, C=O), 1735 ($OCOCH_3$), 1640 cm.$^{-1}$ (C=N). λ_{max} 222 mμ (ε ~ 9000).

3β-Acetoxy-16,17-seco-androstane-16-nitrile-17-oic Acid (VIb).—When crude diacetate IIIb was dissolved in ether, the solution was washed with 3% aqueous sodium bicarbonate, and the basic extract was acidified, a 50% yield of crystals, m.p. 165.5–166.5°, was obtained. Recrystallization of the product from ethyl acetate gave an analytical sample of VIb, m.p. 166–167.5°. On admixture with diacetate IIIb the melting point was depressed to 134–143°. ν_{max} 3320 and 2600 (wk, OH of CO_2H), 2250 (C≡N), 1740 ($OCOCH_3$), 1710 cm.$^{-1}$ (shoulder, CO_2H).

Anal. Calcd. for $C_{21}H_{31}O_4N$: C, 69.77; H, 8.65; N, 3.88. Found: C, 69.60; H, 8.64; N, 3.97. Neut. equiv. Calcd.: 361. Found: 356.

3β-Hydroxy-16,17-seco-androstane-16-nitrile-17-oic Acid (Vb).—Crude diacetate IIIb (620 mg., m.p. 143–147°) was stirred into 50 ml. of 0.25 *N* aqueous potassium hy-

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droxide. After 1 hr. 8 ml. of methanol was added to the suspension and stirring was continued for 1.5 hr. The resulting solution was acidified, diluted with ice water, and extracted with ether. The ether solution was extracted with 10% aqueous sodium carbonate. The ethereal layer upon evaporation gave a semisolid (36 mg.) melting at 200–217° which was not further characterized. The aqueous portion on acidification and filtration yielded 325 mg. of crude Vb, m.p. 160–162.5°. Recrystallization from 50% aqueous methanol raised the melting point to 177–179° (reported[4] for a presumed imide XVIb, m.p. 180–182°). ν_{max} 3340 (OH), 2600 (OH of CO_2H), 2250 (C≡N), 1690 cm.$^{-1}$ (CO_2H).

Hydrolysis of 3β-hydroxy-16,17-seco-androstane-16-nitrile-17-oic Acid (Vb) in Aqueous Alkali.—A solution of 100 mg. of nitrile acid Vb in 10 ml. of 10% aqueous potassium hydroxide was heated under reflux for 10 hr. At the end of this heating period evolution of ammonia was noted. Acidification afforded 10 mg. of a solid, m.p. 208–210° (reported[4] for crude amide XIb, m.p. 210–214°). An infrared spectrum of the material indicated the presence of amide XIb (3320, 1640, 1600 cm.$^{-1}$) and diacid XVIIb (2600, 1710 cm.$^{-1}$). Recrystallization from methanol lowered the melting point to 190–192°.

Chromatography of the above product on silicic acid (Mallinckrodt A. R., 100 mesh, processed by stirring with water and drying at 110°) gave a small amount of impure diacid XVIIb, eluted with benzene, m.p. 227–237° (lit.,[4] m.p. 234–237°), and crude amide acid XIb, eluted with acetone–benzene (1:1), m.p. 209–214° dec. (lit.,[4] m.p. 210–214°). There was not enough material for further purification but the infrared spectra were consistent with the assignments as stated above.

Acknowledgment.—This work was supported in part by a grant from the Research Corp., N. Y. and by Grant CY-4474 from the National Institutes of Health.

Partial Synthesis of Evomonoside[1]

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Received December 6, 1961

Digitoxigenin was treated with 2,3,4-tri-*O*-benzoyl-α-L-rhamnosyl bromide to give an *O*-benzoylated glycoside which, after removal of the protecting groups, yielded digitoxigenin α-L-rhamnopyranoside, identical with the naturally occurring cardiac-active principle, evomonoside. Thus, the proof of structure of evomonoside has been completed. The α-D-rhamnopyranoside of digitoxigenin was made by similar methods and was found to have an infrared absorption spectrum nearly identical to that of evomonoside. Finally, digitoxigenin coupled readily with methyl 2,3,4-tri-*O*-acetyl-1-bromo-1-deoxy-α-D-glucuronate to give an acetylated glucosiduronate which could be isolated. Repeated attempts to saponify the latter resulted in failure.

In 1953, Tamm and Rosselet[5] elucidated the structure of evomonoside [3β-(α-L-rhamnopyranosyl)-14β-hydroxy-5β-card-20(22)-enolide (VI)], but they were unable to complete the proof of its structure by a partial synthesis. When they treated digitoxigenin [3β,14β-dihydroxy-5β-card-20(22)-enolide (V)] with 2,3,4-tri-*O*-acetyl-α-L-rhamnosyl bromide in the presence of silver carbonate, elimination of the C-14 hydroxyl group of the genin (V) took place simultaneously with glycoside formation. Saponification of the *O*-acetylated intermediate gave, instead of the desired evomonoside (VI), 3β(α-L-rhamnopyranosyl)-5β-carda-14,20(22)-dienolide.

In an effort to resolve this problem we turned to a consideration of *O*-benzoylated halides as carbohydrate coupling intermediates inasmuch as the latter are considerably less reactive than the corresponding *O*-acetylglycosyl halides. The known 2,3,4-tri-*O*-benzoyl-α-L-rhamnosyl bromide (I)[6] was prepared easily from L-rhamnose but when I was treated with digitoxigenin (V) in the presence of silver carbonate under conditions essentially the same as described by Meystre and Miescher,[7] only 6% of evomonoside (VI) could be obtained after saponification of the reaction products.

Studies by Helferich and co-workers[8] have shown that mercuric cyanide may advantageously replace silver carbonate as an acid acceptor in the preparation of glycosides. Following Helferich's procedure except for substitution of 1,2-dichloroethane for the solvent nitromethane, we were able to couple the bromide I with the genin V; saponification of the reaction products *in toto* gave 44% of evomonoside (VI). Identity of the synthetic material with authentic naturally occurring evomonoside (VI)[9] was established on the basis of mixture melting point, paper chromatographic comparisons, and infrared spectra.

Natural cardenolides containing rhamnose as a

(1) This work was supported initially by funds generously awarded by Eli Lilly and Co., Indianapolis, Ind. Concluding portions were supported by U. S. Public Health Service Grant H-5839.

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(3) The preparation of 2,3,4-tri-*O*-benzoyl-α-D-rhamnosyl bromide which is described in this paper is taken from a dissertation submitted by G. D. Valiaveedan to the Graduate School of Georgetown University, June, 1960, in partial fulfillment of the M.S. degree.

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(9) The authors are grateful to Prof. T. Reichstein for supplying a sample of natural evomonoside.