The precipitate was collected by filtration, washed with cold water, ethanol, (50:50) ethanol–ether, and finally with ether, then dried *in vacuo*.

The yield for various pteridines formed, based on dry weights, after an additional recrystallization from water, was 50–60%.

2-Amino-4-hydroxy-6-hydroxymethylpteridine (IIa),—was characterized by its infrared spectra. For comparison, an authentic sample was obtained by the $NaBH_4$ reduction[6] of an authentic sample of 2-amino-4-hydroxy-6-pteridinecarboxaldehyde.[21] A pure sample of IIa was oxidized to 2-amino-4-hydroxy-6-pteridinecarboxylic acid (III) with $KMnO_4$ in alkali.[22] Quantitative conversion to the acid III was obtained as judged by the change in spectra.[23] No trace of the 7-acid was seen either spectrally[24] or on paper chromatography in the solvent system of Weygand, *et al.*[25] The absence of the 7-carboxylic acid was judged by the lack of absorption at 400 mμ in 0.1 *N* NaOH where this isomer has a maxima as well as by the correspondence of the spectrum of the oxidation product to that of the 6-carboxylic acid III in all details.[23]

Spectral properties of IIa: in 0.1 *N* HCl, λ_{max} 247 mμ (ε 10,340), λ_{max} 322 mμ (ε 7690), λ_{min} 272 mμ (ε 1760); in 0.1 *N* NaOH, λ_{max} 253 mμ (ε 22,600), λ_{max} 362 mμ (ε 7000), λ_{min} 230 mμ (ε 8400), λ_{min} 300 mμ (ε 1000).

Anal. Calcd. for $C_7H_7N_5O_2$: C, 43.52; H, 3.65; N, 36.26; O, 16.57. Found: C, 43.26; H, 3.94; N, 34.97; O, 17.67.

2,4-Diamino-6-hydroxymethylpteridine (IIb) was characterized by deamination[17] to IIa and the $KMnO_4$ oxidation of IIa to III. The deamination to IIa was quantitative, no trace of the 7-isomer was detected. The selective removal of the 5-amino group in IIb was also accomplished by heating in a boiling water bath for 30 min. in 0.1 *N* NaOH.

Spectral properties of IIb: in 0.1 *N* HCl, λ_{max} 243 mμ (ε 15,730), 283 (4620), 336.5 (9840), λ_{min} 228 mμ (10,670), 265 (3520), 296 (4290); in 0.1 *N* NaOH, λ_{max} 227 mμ (ε 11,300), 257 (21,000), 368 (7200), λ_{min} 235 mμ (ε 10,600), 305 (1200).

Anal. Calcd. for $C_7H_8N_6O$: C, 43.74; H, 4.19; N, 43.74. Found: C, 43.63; H, 4.78; N, 43.78.

2-Hydroxy-4-amino-6-hydroxymethylpteridine (IIc) was characterized by its deamination to 2,4-dihydroxy-6-hydroxymethylpteridine (IV). This was accomplished by heating IIc in 0.1 *N* HCl in a boiling water bath for 30 min. This deamination also proceeds readily in 0.1 *N* NaOH at 100°. The lumazine IV was oxidized to the corresponding carboxylic acid V in alkaline permanganate. As a reference standard lumazine-6-carboxylic acid (V) was prepared by the method of Angier, *et al.*[18] No trace of the lumazine-7-carboxylic acid could be detected spectrally.[19]

Spectral properties of IIc: in 0.1 *N* HCl, λ_{max} 275 mμ (ε 5290), λ_{max} 342 mμ (ε 5170), λ_{min} 266 mμ (ε 4860), λ_{min} 300 mμ (ε 2860); in 0.1 *N* NaOH, λ_{max} 257.5 mμ (ε 13,840), λ_{max} 376 mμ (ε 4200), λ_{min} 236 mμ (ε 9600), λ_{min} 315 mμ (ε 1000).

Anal. Calcd. for $C_7H_9N_5O_3 \cdot H_2O$: C, 39.80; H, 4.29; N, 33.16. Found: C, 39.45; H, 4.29; N, 33.97.

Chromatographic data in four solvent systems for the synthetic pteridines and their oxidation products are given in Table I.

TABLE I

R_f VALUES IN FOUR SOLVENT SYSTEMS[a]

Pteridine	*i*-PrOH–NH_4OH–H_2O (7:1:2)	*n*-BuOH–EtOH–H_2O (100:35:72)	*n*-BuOH–HOAc–H_2O (4:1:5)	*sec*-BuOH–HOAc–H_2O (8:2:5)
IIa	0.30	0.37	0.38	0.52
IIb	0.36	0.39	0.35	0.59
IIc	0.24	0.26	0.40	0.58
III	0.10	0.17	0.22	0.44
V	0.14	0.19	0.20	0.47

[a] Paper chromatography was carried out on 10 × 16 in. sheets of Whatman No. 3 MM paper in closed jars in the dark. The ascending method was employed. Spots were detected by observing the completely dried papers in the dark with a Mineralite ultraviolet source, output maxima at 254 mμ.

Acknowledgment.—We express our appreciation to Dr. Carlos Krumdieck for his interest and encouragement and to Miss Lynn Capel for her technical assistance. We are also indebted to the American Cancer Society (T-67) and to the U. S. Public Health Service (CA-05294 of the National Cancer Institute) for support of this work.

(21) A gift from Dr. Robert B. Angier, Lederle Laboratories, Pearl River, N. Y.

(22) F. Weygand and G. Shaefer, *Chem. Ber.*, **85**, 307 (1952).

(23) E. L. R. Stokstad, B. L. Hutchings, J. H. Mowat, J. H. Boothe, C. W. Waller, R. B. Angier, J. Semb, and Y. SubbaRow, *J. Am. Chem. Soc.*, **70**, 5 (1948).

(24) J. H. Mowat, J. H. Boothe, B. L. Hutchings, E. L. R. Stokstad, C. W. Waller, R. B. Angier, J. Semb, D. B. Cosulich, and Y. SubbaRow, *ibid.*, **70**, 14 (1948).

(25) F. Weygand, A. Wacker, and V. Schmied-Koworzik, *Experientia*, **6**, 184 (1950).

5α-Androstano[3,2-*b*]pyrroles[1]

THEODORE C. MILLER AND ROBERT G. CHRISTIANSEN

Sterling-Winthrop Research Institute, Rensselaer, New York

Received July 28, 1964

5α-Androstano[3,2-*b*]pyrrol-17β-ol (**7a**) was synthesized by the following sequence of steps: 17β-hydroxy-5α-androstan-3-one (**1a**) → 17β-hydroxy-2-(hydroxymethylene)-5α-androstan-3-one (**2a**) → 2α-allyl-17β-hydroxy-5α-androstan-3-one acetate (**4b**) → 2α-(formylmethyl)-17β-hydroxy-5α-androstan-3-one acetate (**5a**) → 5α-androstano[3,2-*b*]-1′-benzylpyrrol-17β-ol acetate (**6a**) → **7a**. The 2α-allylation of **1a** was also accomplished by direct allylation of **1a** and by the allylation of the pyrrolidine enamine (**3**) of **1a**. A similar series of steps afforded 17-methyl-5α-androstano[3,2-*b*]pyrrol-17β-ol (**7b**) from 17β-hydroxy-17-methyl-5α-androstan-3-one (**1b**). A small amount of alkylation at the 4-position was observed in the direct allylation of **1a** and **1b**.

As part of a program to prepare analogs of the excellent anabolic agent, 17α-methyl-5α-androstano[3,2-*c*]-pyrazol-17β-ol (stanozolol, Winstrol®), and related steroid-fused pyrazoles[2a,b] and isoxazoles,[3a,b] the synthesis of the 5α-androstano[3,2-*b*]pyrroles (**7a** and **7b**) was undertaken. This paper reports the successful synthesis of **7a** and **7b**[4] by the sequence of steps shown in the flow chart: first, introduction of the allyl group into the 2α-position of the 5α-androstan-3-one (**1**) either directly, *via* the 2-(hydroxymethylene)-3-one

(1) Steroidal Heterocycles. XI. Paper X: J. H. Ackerman, G. O. Potts, A. L. Beyler, and R. O. Clinton, *J. Med. Chem.*, **7**, 238 (1964).

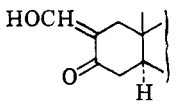
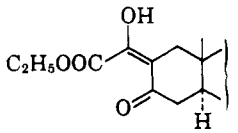
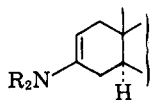
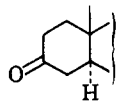
(2) (a) R. O. Clinton, A. J. Manson, F. W. Stonner, A. L. Beyler, G. O. Potts, and A. Arnold, *J. Am. Chem. Soc.*, **81**, 1513 (1959); (b) R. O. Clinton, A. J. Manson, F. W. Stonner, H. C. Neumann, R. G. Christiansen, R. L. Clarke, J. H. Ackerman, D. F. Page, J. W. Dean, W. B. Dickinson, and C. Carabateas, *ibid.*, **83**, 1478 (1961).

(3) (a) R. O. Clinton, A. J. Manson, F. W. Stonner, R. G. Christiansen; A. L. Beyler, G. O. Potts, and A. Arnold, *J. Org. Chem.*, **26**, 279 (1961); (b) A. J. Manson, F. W. Stonner, H. C. Neumann, R. G. Christiansen, R. L. Clarke, J. H. Ackerman, D. F. Page, J. W. Dean, D. K. Phillips, G. O. Potts, A. Arnold, A. L. Beyler, and R. O. Clinton, *J. Med. Chem.*, **6**, 1 (1963).

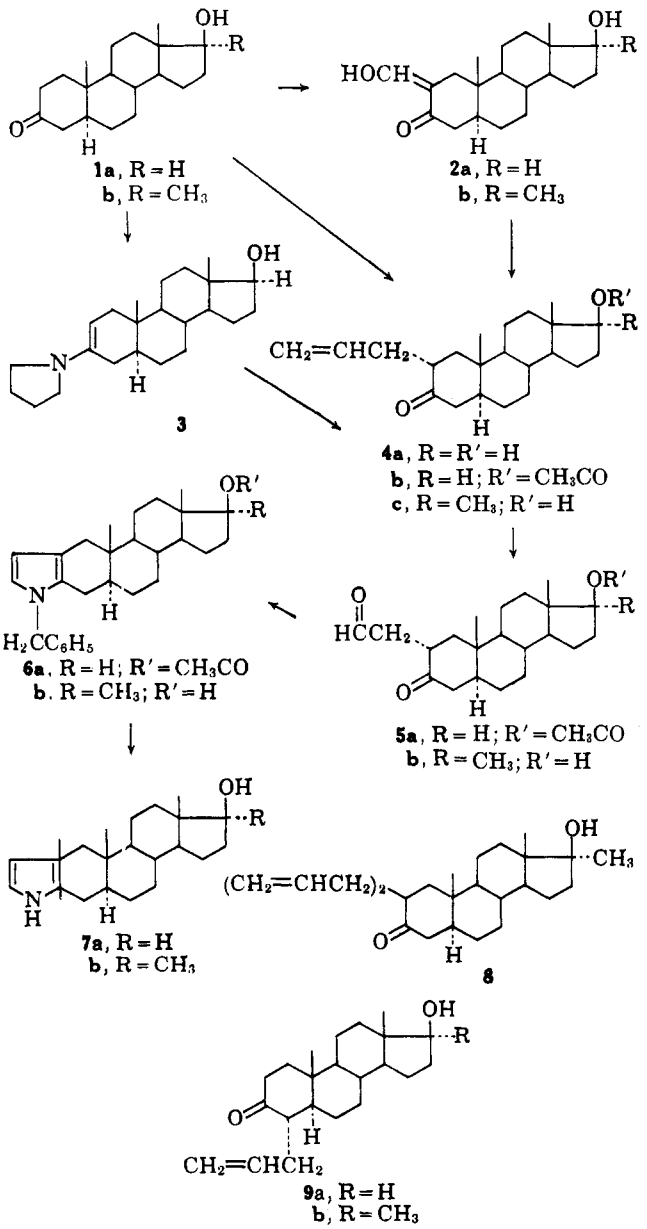
(4) A recent patent [J. C. Orr and A. Bowers, U. S. Patent 3,032,551 (May 1, 1962)] reported the synthesis of 5α-androstano[3,2-*b*]pyrroles by the reaction of 5α-androstan-3-ones with α-aminoaldehydes or α-amino ketones, but no physical data for the pyrroles were given.

TABLE I

2α-METHYLATION OF 5α-STEROID 3-ONES

Starting materials	Yield of 2α-methyl-3-one,[a] % 5α-Cholestan-3-one	5α-Androstan-3-one
 + CH_3I + CH_3ONa	30[b] ?[c]	
 + CH_3I + base	10[d]	9–17[e]
 + CH_3I	?[c]	<11[f]
 + CH_3I + base	50[b] >20[d]	?[g]

[a] Yields are based on 3-one. [b] Ref. 6. [c] B. Fuchs and H. J. E. Loewenthal, *Tetrahedron*, **11**, 199 (1960). [d] Ref. 5. [e] H. J. Ringold, E. Batres, O. Halpern, and E. Necoechea, *J. Am. Chem. Soc.*, **81**, 427 (1959). [f] Upjohn Co., British Patent 889,330 (Nov. 25, 1958). [g] O. Engelfried and M. Schenck, German Patent 1,117,112 (May 17, 1962).



(2), or *via* the 3-one pyrrolidine enamine (**3**); secondly, oxidative cleavage of the double bond of the 2α-allyl-3-one (**4**) to form the ketoaldehyde (**5**); then cyclization of **5** to the N-benzylpyrrole (**6**); and, finally, reductive debenzylation of **6** to the 5α-androstano[3,2-*b*]pyrrole (**7**).

Although none of the published methods for the selective alkylation of 5α-steroid 3-ones at the 2α-position promised good yields of the 2α-alkyl-3-ones, several of them appeared applicable to the preparation of 2α-allyl-17β-hydroxy-5α-androstan-3-one (**4a**) and its 17α-methyl homolog (**4c**). Table I summarizes the literature on methylation in the 2α-position of 5α-cholestan-3-ones and 5α-androstan-3-ones. Direct methylations of the 3-ones produced significant amounts of 2,2-dimethyl-3-ones in addition to 2α-methyl-3-ones, the relative amount of the former increasing with the quantity of methyl iodide used.[5] In one case cited in the table, 2α-methyl-5α-cholestan-3-one (50% yield) was accompanied by a 30% yield of 2,2-dimethyl-5α-cholestan-3-one.[6]

While this paper was being written, the synthesis of 2α-allyl-17β-hydroxy-5α-androstan-3-one acetate (**4b**) by two methods was reported by Gardi and Castelli.[7] The first involved the thermal rearrangement (70% yield) of 3-(allyloxy)-5α-androst-2-en-17β-ol acetate, which was prepared in 55% yield from 3-ethoxy-5α-androst-2-en-17β-ol acetate, which[8] in turn was prepared by partial hydrogenation of 3-ethoxyandrosta-3,5-dien-17β-ol acetate in unstated yield. Thus, the length of this route (four steps) and the uncertain yield from testosterone would have made it of questionable synthetic utility in the present work. The second method (3.7% yield), which was used to confirm the structure of the thermal rearrangement product, involved allylation of 2-(ethoxalyl)-17β-hydroxy-5α-androstan-3-one followed by acetylation of the crude 2α-

(5) Y. Mazur and F. Sondheimer, *J. Am. Chem. Soc.*, **80**, 5220 (1958).
(6) M. Mousseron, F. Winternitz, and A. Crastes de Paulet, *Compt rend.*, **245**, 1859 (1957).
(7) R. Gardi and P. P. Castelli, *Gazz. chim. ital.*, **93**, 1681 (1963).
(8) R. Gardi, P. P. Castelli, and A. Ercoli, *Tetrahedron Letters*, 497 (1962).

allyl-17β-hydroxy-5α-androstan-3-one (**4a**), which was not characterized.

In the present work, three of the alkylation procedures in Table I were employed to prepare 2α-allyl-17β-hydroxy-5α-androstan-3-one (**4a**). While none of the three gave good yields of **4a**, one (the reaction of 17β-hydroxy-2-(hydroxymethylene)-5α-androstan-3-one (**2a**)[2b] as its sodium salt with allyl bromide in dimethylformamide) was preferred because the product could be completely freed from by-products by column chromatography. Based on 17β-hydroxy-5α-androstan-3-one (**1a**), the yield was 23%. In another run in which the crude allylation product was acetylated before chromatography, the yield of the acetate ester **4b** of **4a** was 29%. The reaction of crude 17β-hydroxy-5α-androstan-3-one pyrrolidine enamine (**3**) with allyl bromide in dimethylformamide afforded **4a** in 14% yield based on **1a**. Accompanying **4a** in the chromatographic fractions containing it was a large amount of oily by-product, which impeded the crystallization of **4a** and precluded quantitative isolation of it. Direct allylation of **1a** by means of allyl bromide and potassium *t*-butoxide in refluxing benzene afforded **4a** in 15% yield. In this case too, the chromatographic fractions containing **4a** contained appreciable amounts of oily by-product, which made purification of **4a** difficult. The physical constants of the 2α-allyl-17β-hydroxy-5α-androstan-3-one acetate (**4b**) prepared in this work agreed with those of Gardi and Castelli.[7]

2α-Allyl-17β-hydroxy-17-methyl-5α-androstan-3-one (**4c**) was obtained both by allylation of the 2-(hydroxymethylene)-3-one (**2b**) and by direct allylation of the 3-one (**1b**). A different base–solvent combination was tried for the allylation of **2b**. Whereas allylation of **2a** produced **4a** in 23% yield with sodium hydride–dimethylformamide, **4c** was obtained in only 12% yield with potassium *t*-butoxide–dimethyl sulfoxide. Although **4c** was not crystalline, except as an unstable solvate with acetonitrile, it showed a single spot on a thin layer chromatographic plate and its analysis and infrared spectrum confirmed its structure. The direct allylation of **1b** by means of potassium *t*-butoxide and allyl bromide in refluxing benzene produced a mixture which could be partially resolved by column chromatography. The first fractions contained an intimate mixture of **4c** and one other component of higher R_f value than **4c** as shown by thin layer chromatography.[9] Vapor phase chromatographic analysis of these fractions revealed the presence of 65% of **4c**. Since the other component did not interfere with the completion of the synthesis of **7b**, the direct allylation of **1b** effectively produced a 32% yield of **4c**.

Formation of the ketoaldehydes (**5**) could be accomplished either by treatment of the 2α-allyl-3-ones (**4**) with sodium metaperiodate and a catalytic amount of osmium tetroxide in aqueous dioxane[10] or by ozonization followed by reductive work-up. 2α-(Formylmethyl)-17β-hydroxy-5α-androstan-3-one acetate (**5a**) was obtained by the first method in 38% yield. Three different reductive work-ups of the ozonide of **4b** afforded **5a** in yields of 16–22%. Neither method of oxidation permitted isolation of **5a** by crystallization of the crude product, but both gave pure **5a** after column chromatography. 2α-(Formylmethyl)-17β-hydroxy-17-methyl-5α-androstan-3-one (**5b**), obtained by the sodium metaperiodate–osmium tetroxide oxidation of **4c**, was not crystalline even after column chromatography. Fortunately, crude **5b**, which resulted from the oxidation of **4c** containing the impurity of higher R_f value, could be carried successfully through the cyclization step.

Cyclization of 1,4-diketones, 1,4-ketoaldehydes, and 1,4-dialdehydes into pyrroles by heating them with ammonia or primary amines at temperatures above 100° is the long-known Paal–Knorr synthesis.[11a,b] What is believed to be the first instance of the application of this synthesis to the preparation of cycloalkane-fused pyrroles, namely a series of N-substituted cyclopenta[*b*]-2-methylpyrroles and cyclohexa[*b*]-2-methylpyrroles, was only recently reported.[12] In the present work, and prior to that report, it was found that the cyclization of the 1,4-ketoaldehydes **5a** and **5b** with benzylamine took place in refluxing benzene solution. *p*-Toluenesulfonic acid was used as a catalyst, although its use was not proved to be necessary, and the water formed was collected by azeotropic distillation with benzene. 5α-Androstano[3,2-*b*]-1′-benzylpyrrol-17β-ol acetate (**6a**) was thus obtained from **5a** in 71% yield. 17-Methyl-5α-androstano[3,2-*b*]-1′-benzylpyrrol-17β-ol (**6b**) was readily purified by crystallization of the crude product from methanol, even though the intermediates (**4c** and **5b**) had been quite impure. Since the crystals of **6b** were weakly solvated by methanol in unreproducible amounts, the compound was not characterized.

Insofar as the authors are aware, the alkali metal–liquid ammonia debenzylation of N-benzylpyrroles has not been reported, although the debenzylation of N-benzylindole by sodium–liquid ammonia has been described.[13] The use of lithium in liquid ammonia–tetrahydrofuran provided a ready means of debenzylating **6a** and **6b**. Debenzylation of **6a** was accompanied by deacetylation of the 17β-ol acetate, affording 5α-androstano[3,2-*b*]pyrrol-17β-ol (**7a**) in 71% yield. The methanol of solvation of **6b** did not interfere with its debenzylation. 17-Methyl-5α-androstano[3,2-*b*]-pyrrol-17β-ol (**7b**) was characterized as crystals containing 0.25 mole of ethyl acetate, yet melting at 215° with no evidence of desolvation. In a later experiment, crystals of **7b**, which were dried exactly as previously, contained no ethyl acetate of solvation and melted only slightly higher (216°). Whereas the proton magnetic resonance spectrum of **7a** was not clear enough to discern the aromatic proton coupling, the coupling of the three pyrrole protons in the spectrum of **7b** was clearly that special type of coupling observed in 2,3-dimethylpyrrole, in which the C-4 and C-5 protons interact equally with each other and with the N-proton.[14]

(9) It is believed that this component of higher R_f value than **4c** was 2,2-diallyl-17β-hydroxy-17-methyl-5α-androstan-3-one (**8**), first, because direct methylation of 5α-steroid 3-ones has been shown to produce 2,2-dimethyl-3-one as well as 2α-methyl-3-one,[5,6] and, secondly, because the infrared spectrum of the intimate mixture of **4c** and **8** differed from that of **4c** only in the relative intensities of the vinyl group absorption bands at 10.03 and 10.93 μ, which were more intense in the spectrum of the mixture.

(10) R. Pappo, D. S. Allen, Jr., R. U. Lemieux, and W. S. Johnson, *J. Org. Chem.*, **21**, 478 (1956).

(11) (a) C. Hollins, "The Synthesis of Nitrogen Ring Compounds Containing a Single Heteroatom," E. Benn, Ltd., London, 1924, pp. 28–33; (b) A. H. Corwin in "Heterocyclic Compounds," Vol. I, R. C. Elderfield, Ed., John Wiley and Sons, Inc., New York, N. Y., 1950, pp. 289–290.

(12) M. A. Volodina, V. G. Mishina, A. P. Terent'ev, and G. V. Kiryushkina, *Zh. Obshch. Khim.*, **32**, 1922 (1962); *Chem. Abstr.*, **58**, 5612*d* (1963).

Direct alkylation of 5α-steroid 3-ones in the 4-position is heretofore unrecorded. Direct allylation of **1a** and **1b** by allyl bromide–potassium *t*-butoxide in refluxing benzene produced as by-products minor amounts of monoallyl-3-ones, which have been shown by indirect proof to be 4α-allyl-17β-hydroxy-5α-androstan-3-one (**9a**) and 4α-allyl-17β-hydroxy-17-methyl-5α-androstan-3-one (**9b**). The analyses and infrared spectra of **9a** and **9b** indicated their monoallyl-3-one character. When a solution of **9a** and a catalytic amount of *p*-toluenesulfonic acid monohydrate was heated at reflux, **9a** was recovered unchanged. Since 2β-methyl-5α-cholestan-3-one was isomerized to the 2α-methyl-3-one and 4β-methyl-5α-cholestan-3-one was isomerized to the 4α-methyl-3-one by refluxing ethanolic sulfuric acid,[5] it appeared unlikely that **9a** was either the 2β-allyl-3-one or the 4β-allyl-3-one. Furthermore, since the 2α-, 2β-, 4α-, and 4β-positions were the only reasonable sites for the allyl group, **9a** had to be the 4α-allyl-3-one by elimination. The acid-catalyzed isomerization of **9b** was not attempted. However, two additional observations served to relate **9a** and **9b**, thus also establishing **9b** as the 4α-allyl-3-one. First, they appeared at the same relative positions in column and thin layer chromatograms. Secondly, the difference in molecular rotations between **9b** and **4a** (64°) was approximately equal to the difference in molecular rotations between **9b** and **4c** (62°).

Experimental

General.—Reagent grade solvents and inorganic chemicals were used in reactions. Melting points were taken in capillaries and are uncorrected. Chromatographic silica gel was supplied by the Davison Company (Grade 923, 100–200 mesh). Thin layer chromatographic plates were coated with silica gel supplied by Merck A. G. (GF_{254}). The spots were brought out on the plates by spraying with 20% ethanolic sulfuric acid, then 0.5% ethanolic vanillin,[15] and heating on a hot plate. Infrared spectra were determined on 1% potassium bromide pellets on a Perkin-Elmer Model 21 spectrophotometer, unless otherwise noted. Proton magnetic resonance spectra were determined on deuteriochloroform solutions, unless otherwise noted, on a Varian Model A-60 spectrometer, using tetramethylsilane as an external reference. Rotations were determined on 1% chloroform solutions.

2α-Allyl-17β-hydroxy-5α-androstan-3-one (4a). A.—A mixture of 17β-hydroxy-2-(hydroxymethylene)-5α-androstan-3-one[2b] (m.p. 168–172°, 3.18 g., 0.0100 mole), dimethylformamide (dried over potassium hydroxide and distilled, 20 ml.), and sodium hydride (0.30 g., 0.013 mole) was stirred for 0.5 hr. at room temperature under nitrogen. Allyl bromide (redistilled, 1.51 g., 0.0125 mole) was added and the mixture was stirred for 1 hr. on the steam bath. Aqueous potassium hydroxide (2 g. in 5 ml. of water) was added, and stirring was continued on the steam bath for 1 hr. Methylene dichloride (50 ml.) was added to the reaction mixture, followed by careful addition of water (300 ml.). The organic phase was separated and the aqueous phase was extracted again with methylene dichloride (50 ml.). The combined methylene dichloride extracts were washed with water, dried over sodium sulfate, filtered, and chromatographed on silica gel (200 g.). The 80:20 pentane–ether eluates contained the product, which was recrystallized from ether–hexane as colorless prisms, 0.85 g., 26% yield (based on **2a**, 23% based on **1a**), m.p. 118–119°, $[\alpha]^{25}D$ +13.6°. Its infrared spectrum showed absorption bands at 2.83 (—O—H), 5.82 (—C(=O)—), 6.09 (—C=C—), and 10.03 and 11.04 μ (CH_2=CH—). Its proton magnetic resonance spectrum showed a broad signal at 6.17 p.p.m. (—C=CH—) and multiplets at 5.37 and 5.58 p.p.m. (CH_2=C—).

Anal. Calcd. for $C_{22}H_{34}O_2$: C, 79.95; H, 10.37. Found: C, 80.08; H, 10.38.

B.—A solution of 17β-hydroxy-5α-androstan-3-one (m.p. 177–181°, 43.5 g., 0.150 mole) and pyrrolidine (redistilled, 32.0 g., 0.450 mole) in benzene (500 ml.) was refluxed under a water separator until no further water was removed (1.5 hr.). The solution was evaporated to dryness on the steam bath under reduced pressure. The residue was dissolved in dimethylformamide (redistilled, 600 ml.) and a few milliliters of liquid was distilled in order to remove last traces of benzene and pyrrolidine. Allyl bromide (redistilled, 85.0 g., 0.700 mole) was added and the solution was heated for 2 hr. on the steam bath. Water (100 ml.) was added and heating was continued for 2 hr. The reaction mixture was poured into water (3 l.) and the aqueous suspension was extracted with four 200-ml. portions of ether. The ether extracts were washed with saturated sodium chloride solution, dried over sodium sulfate, filtered, and concentrated to a red-brown oil, which was chromatographed on silica gel (1.50 kg.). The partly crystalline fractions (18.9 g.) containing **4a** were eluted by 95:5 methylene dichloride–ether and recrystallized from acetone–hexane, yielding 6.89 g. (14%), m.p. 117–119°, undepressed by material prepared by procedure A.

C.—Freshly cut potassium (0.98 g., 0.025 g.-atom) was dissolved in refluxing *t*-butyl alcohol (distilled from calcium hydride, 100 ml.). The excess *t*-butyl alcohol was distilled and the potassium *t*-butoxide was baked dry on the steam bath at reduced pressure. Benzene (100 ml.), 17β-hydroxy-5α-androstan-3-one (5.81 g., 0.0200 mole), and allyl bromide (redistilled, 2.42 g., 0.0200 mole) were added, and the mixture was refluxed for 5 hr. under nitrogen. It was then allowed to stand overnight at room temperature. Water (20 ml.) and 2 *N* hydrochloric acid (20 ml.) were added. The benzene layer was separated, washed with saturated sodium chloride solution, dried over sodium sulfate, filtered, and concentrated on the steam bath to a clear, colorless oil. Trituration of the oil with ether–hexane gave some unchanged starting material (0.66 g., 11%, m.p. 173–180°, undepressed by starting material). Chromatography of the mother liquor on silica gel (300 g.) gave the product as an oil (2.35 g.) in the 80:20 pentane–ether eluates. The oil crystallized from acetone–hexane, yielding 0.96 g. (15%), m.p. 117–119°, undepressed by material prepared by procedure B.

4α-Allyl-17β-hydroxy-5α-androstan-3-one (9a) was eluted as a crystalline solid (0.32 g.) from the column with 70:30 pentane–ether and was recrystallized from ether as colorless prisms, 0.22 g., 3.3% yield, m.p. 150–151°, $[\alpha]^{25}D$ +34.7°. The infrared spectrum showed absorption bands at 2.81 (—O—H), 5.84 (—C(=O)—), and 6.08 μ (—C=C—).

Anal. Calcd. for $C_{22}H_{34}O_2$: C, 79.95; H, 10.37. Found: C, 80.07; H, 10.22.

When a solution of **9a** (22 mg.) and *p*-toluenesulfonic acid monohydrate (1 mg.) in 95% ethanol (2 ml.) was heated under reflux for 2 hr., unchanged **9a** (identified by mixture melting point and infrared spectrum) was recovered quantitatively.

2α-Allyl-17β-hydroxy-5α-androstan-3-one acetate (4b) was prepared from **4a** by treatment with acetic anhydride–pyridine on the steam bath: colorless prisms from hexane, m.p. 130–131°, $[\alpha]^{25}D$ +4.8°.

Anal. Calcd. for $C_{24}H_{36}O_3$: C, 77.37; H, 9.74. Found: C, 77.42; H, 9.59.

Compound **4b** was also obtained in 32% yield (based on **2a**, 29% based on **1a**) by acetylating the crude **4a** obtained by procedure A, followed by chromatography on silica gel.

2α-Allyl-17β-hydroxy-17-methyl-5α-androstan-3-one (4c). A. A solution of 17β-hydroxy-2-(hydroxymethylene)-17-methyl-5α-androstan-3-one[2b] (3.32 g., 0.0100 mole), potassium *t*-butoxide (M. S. A. Research Co., 1.23 g., 0.0110 mole), allyl bromide (redistilled, 1.45 g., 0.0120 mole), and dimethyl sulfoxide (Matheson Coleman and Bell, 50 ml.) was stirred for 2 hr. at room temperature under nitrogen. Aqueous sodium hydroxide (2 *M*, 5 ml.) was added dropwise and the solution was warmed for 1 hr. on the steam bath. The red-brown solution was cooled and diluted with benzene (30 ml.) and water (0.5 l.). The benzene layer was separated, washed with water, dried over sodium sulfate, filtered, concentrated to 10–20 ml., and chromatographed on silica gel (100 g.). The product was eluted by 80:20 pentane–

(13) M. Julia, P. Manoury, and J. Igolen, *Compt. rend.*, **251**, 394 (1960).

(14) J. A. Pople, W. G. Schneider, and H. J. Bernstein, "High Resolution Nuclear Magnetic Resonance," McGraw-Hill Book Co., Inc., New York, N. Y., 1959, pp. 270–271.

(15) J. S. Matthews, *Biochim. Biophys. Acta*, **69**, 163 (1963).

ether as a yellow oil showing a single spot on a t.l.c. plate, 0.44 g., 13% yield, $[\alpha]^{25}D$ +3.0°. It crystallized from acetonitrile at ice temperature, but the crystals decomposed at room temperature. Thus, the oil itself was characterized. Its infrared spectrum (carbon tetrachloride solution) showed absorption bands at 2.79 and 2.88 (—O—H), 5.85 (—C(=O)—), 6.16 (—C=C—), and 10.03 and 10.93 μ (CH_2=CH—).

Anal. Calcd. for $C_{23}H_{36}O_2$: C, 80.18; H, 10.53. Found: C, 79.98; H, 10.31.

B.—Two runs of this experiment are described. In the first, a mixture of 17β-hydroxy-17-methyl-5α-androstan-3-one (m.p. 190–195°, 24.36 g., 0.0800 mole), potassium *t*-butoxide (M. S. A. Research Co., 11.22 g., 0.100 mole), allyl bromide (redistilled, 9.68 g., 0.0800 mole), and benzene (400 ml.) was stirred at reflux under nitrogen for 4 hr. The reaction mixture was cooled and shaken with excess dilute hydrochloric acid. The benzene layer was washed with water, dried over anhydrous sodium sulfate, filtered, and concentrated on the steam bath under reduced pressure to a yellow oil. Trituration of the oil with 60:40 pentane–ether gave unchanged starting material (5.10 g., 21%) which was recrystallized from ethyl acetate, yielding 3.85 g. (16%), m.p. 193–194.5°, undepressed on admixture with starting material. Chromatography of the mother liquors on alumina (Merck reagent grade, 650 g.) afforded a colorless oil in the 40:60 pentane–ether eluates, 14.86 g. ("impure **4c**"). This "impure **4c**" was initially thought to be pure and was carried through the next two steps of the synthesis as described later.

In the second run, 60.09 g. (0.200 mole) of **1b** was used and 33.63 g. of "impure **4c**" was obtained. On a t.l.c. plate "pure **4c**" (that obtained from **2b**, R_f 0.60) could readily be distinguished from **1b** (R_f 0.45) and from **9b** (R_f 0.55) when the plate was developed with ether. However, when the plate was developed three times with 50:50 pentane–ether, allowing the plate to dry between passes, "impure **4c**" was clearly resolved into **4c** (R_f 0.19) and a component having an R_f value of 0.24. The colors of the spots differed which also aided in distinguishing them. Quantitative analysis of this "impure **4c**" by vapor phase chromatography[16] indicated the presence of 65.4% of **4c** (retention time 20.8 min.) and 33.6% of the component (retention time 33.0 min.) having R_f 0.24.

Continuation of the column chromatography in the first run afforded crude **4α-allyl-17β-hydroxy-17-methyl-5α-androstan-3-one** (**9b**, 2.83 g.) mixed with **4c** in the 70:30 pentane–ether eluates. Three recrystallizations from ethyl acetate gave the analytical sample, which was shown by t.l.c. to contain no detectable amounts of impurity: 0.58 g., 2.1% yield, m.p. 152–154°, $[\alpha]^{25}D$ +23.4°. Its infrared spectrum showed bands at 2.99 (O—H), 5.85 (—C(=O)—), 6.11 (—C=C—), and 10.11 and 10.95 μ (CH_2=C—).

Anal. Calcd. for $C_{23}H_{36}O_2$: C, 80.18; H, 10.53. Found: C, 80.41; H, 10.45.

2α-(Formylmethyl)-17β-hydroxy-5α-androstan-3-one Acetate (5a). A.—To a solution of **4b** (5.00 g., 0.0134 mole) in dioxane (80 ml.) and water (10 ml.) was added dropwise with stirring a freshly prepared solution of osmium tetroxide (0.157 *M*, 4.3 ml., 0.00067 mole). After being stirred for 15 min. at room temperature, the deep brown solution was treated with finely ground sodium metaperiodate (5.75 g., 0.0268 mole), added in portions during 30 min. Stirring was continued for 1 hr. at room temperature, after which time the colorless solution contained a bulky, white precipitate, and the mixture was poured with stirring into water (1 l.). The resulting white solid was collected, washed with water, dried (5.21 g.), and chromatographed on silica gel (200 g.). Elution with 75:25 pentane–ether and recrystallization of the product from ether–hexane gave reddish needles: 1.93 g., 38% yield, m.p. (evacuated capillary) 174–176°. The instability of **5a** was evidenced by the fact that solutions of it turned red and the fact that, in an open melting point capillary, melting was accompanied by appreciable decomposition. The analytical sample had m.p. 175–176° and $[\alpha]^{25}D$ +6.5°. The infrared spectrum (Beckman IR-7 spectrophotometer, chloroform solution) showed a weak absorption band at 3.65 μ (H—CO—) and carbonyl group bands (carbon tetrachloride solution) at 5.77, 5.79, and 5.85 μ. The proton magnetic resonance spectrum showed a singlet signal at 10.27 p.p.m. (H—CO—).

Anal. Calcd. for $C_{23}H_{34}O_4$: C, 73.76; H, 9.15. Found: C, 73.48; H, 8.98.

B.—A solution of **4b** (1.00 g., 0.00269 mole) and pyridine (0.25 ml., 0.0031 mole) in methylene dichloride (100 ml.) was treated with a stream of ozone (0.0032 mole) in oxygen with stirring at −70°.[17] The blue solution was shaken with 10% palladium on carbon (0.10 g.) and hydrogen (30–45 p.s.i.g.) for 1.5 hr. in a Parr apparatus. Separation of the catalyst by filtration and concentration of the filtrate afforded a deep red (colloidal palladium?) oil, which was chromatographed on silica gel (40 g.). The yield of product having m.p. (evacuated capillary) 172–174.5° was 0.22 g. (22%). Alternative work-up involving treatment of the ozonization solution with zinc (2 g.) and glacial acetic acid (10 ml.) afforded product having m.p. 171–175° in 20% yield. When the ozonization was carried out at 2–5° in ethyl acetate and the ozonization solution was worked up by hydrogenation over 30% palladium hydroxide on strontium carbonate, the yield of product having m.p. (evacuated capillary) 171–173° was 16%.

5α-Androstano[3,2-*b*]-1′-benzylpyrrol-17β-ol Acetate (6a).—A solution of **5a** (2.00 g., 0.00534 mole), benzylamine (1.18 ml., 0.0107 mole), and *p*-toluenesulfonic acid monohydrate (0.10 g. in benzene (50 ml.) was refluxed for 2 hr. under a water separator. The benzene solution was washed with 5% hydrochloric acid, water, then saturated sodium bicarbonate solution, dried over sodium sulfate, filtered, and concentrated on the steam bath under reduced pressure. The residue was recrystallized twice from methanol as colorless needles, 1.68 g., 71% yield, m.p. 188–190°, $[\alpha]^{25}D$ +54.1°. The ultraviolet spectrum (recorded on a Cary Model 11 spectrophotometer in 95% ethanol solution) showed λ_{max} 209 mμ (ε 15,000). The absorption reported[18] for N-benzylpyrrole is λ_{max} 208 mμ (ε 13,700).

Anal. Calcd. for $C_{30}H_{39}NO_2$: C, 80.85; H, 8.82; N, 3.14. Found: C, 81.18; H, 8.55; N, 3.09.

17-Methyl-5α-androstano[3,2-*b*]-1′-benzylpyrrol-17β-ol (6b).—Efforts to prepare the pure intermediate **5b** by the procedure below followed by column chromatography on silica gel gave a noncrystalline product, slightly contaminated by a less polar material as shown by t.l.c. Thus, **6b** was prepared as follows without isolation of **5b**.

Aqueous osmium tetroxide (1.0 g., 0.0043 mole in 20 ml. of water) was added dropwise to a stirred solution of "impure **4c**" (33.63 g., obtained by method B, second run) in dioxane (520 ml.) and water (110 ml.). After 15 min., powdered sodium metaperiodate (66.0 g., 0.308 mole) was added in portions with continued stirring during 30 min. The reaction mixture was kept at 25–27° by means of a water bath. After 1.5 hr. more, the mixture, now nearly colorless, was quenched in ice–water (2.5 l.). The product was extracted with methylene dichloride. The methylene dichloride extracts were washed with water, dried over sodium sulfate, filtered, and concentrated on the steam bath under reduced pressure to a dark brown oil. A solution of the oil, benzylamine (21.4 g., 0.200 mole), *p*-toluenesulfonic acid monohydrate (1.90 g., 0.0100 mole), and benzene (350 ml.) was heated for 2 hr. at reflux under a water separator. The solution was concentrated on the steam bath under reduced pressure to a dark brown oil. Recrystallization from methanol gave glistening tan crystals, 18.1 g., dried in the air at room temperature. This material was a methanol solvate whose methanol content varied with the extent of drying. It lost its solvent at about 80° to become an amorphous glass. The infrared spectrum of **6b** showed absorption bands at 2.9 (—O—H), and 6.68, 6.71, and 6.88 μ (aromatic ring).

5α-Androstano[3,2-*b*]pyrrol-17β-ol (7a).—Lithium (0.56 g., 0.081 g.-atom) was added in small pieces to a refluxing slurry of **6a** (1.84 g., 0.00404 mole) in tetrahydrofuran (10 ml.) and liquid ammonia (about 40 ml.). The mixture was stirred under a Dry Ice condenser for 1 hr., then treated with ammonium chloride (about 5 g.), added in small portions. The resulting thick slurry was worked with a spatula to discharge all of the blue color. The ammonia was allowed to evaporate and the residue was diluted with water (200 ml.). The solid was collected, washed with water, dried (1.27 g.), and recrystallized twice from methanol as pale peach blades, 0.75 g., 59% yield, m.p. (evacuated capillary) 244–245°, $[\alpha]^{25}D$ +62.4°. The infrared spectrum

(16) The vapor phase chromatographic analysis was performed on an F and M Model 400 instrument equipped with a 3.8% silicone gum (SE-30) column (4 ft. × 4 mm. i.d.) at 225° with a helium flow rate of 40 ml./min. A flame ionization detector was used.

(17) G. Slomp, Jr., and J. L. Thompson, *J. Am. Chem. Soc.*, **80**, 915 (1958).

(18) J. Davoll, *J. Chem. Soc.*, 3802 (1953).

showed absorption bands at 2.85 (—N—H), 3.00 (—O—H), 6.15, 6.30, 6.43, and 6.57 μ (aromatic ring). Since **7a** was insoluble in chloroform, a proton magnetic resonance spectrum was run on a dioxane solution in order to observe the aromatic proton signals. A noisy base line obscured the fine structure. Signals were seen at about 9.0 (—N—H, very broad), 6.58 (=C—H at 2'), and 5.93 p.p.m. (=C—H at 3').

Anal. Calcd. for $C_{21}H_{31}NO$: C, 80.46; H, 9.97; N, 4.47. Found: C, 80.73; H, 10.10; N, 4.50, 4.56.

17-Methyl-5α-androstano[3,2-*b*]pyrrol-17β-ol (7b).—Lithium (2.4 g., 0.34 g.-atom) was added in small pieces during 10 min. to a stirred slurry of **6b** (7.02 g.) in tetrahydrofuran (dried over alumina, 35 ml.) and liquid ammonia (140 ml.). The mixture was stirred for 50 min. under a Dry Ice condenser. Ammonium chloride (21.4 g., 0.400 mole) was added with continued stirring during 10 min. After most of the ammonia had evaporated, the mixture was diluted with water and the resulting mixture was extracted with methylene dichloride (130 ml.). The methylene dichloride extracts were washed with water, dried over sodium sulfate, filtered, and concentrated with ethyl acetate. The product crystallized from ethyl acetate as irregular prisms containing 0.25 mole of ethyl acetate (confirmed by the infrared spectrum) and having adsorbed red-orange color: 3.78 g., 13.2% yield based on **1b**, m.p. (evacuated capillary) 213–214.5°. Two further recrystallizations gave the final product: 2.59 g., dried at 80° at 0.01 mm., m.p. (evacuated capillary) 214–215°. The infrared spectrum showed absorption bands at 2.78 (shoulder), 2.87, 3.02, and 3.06 μ (sh) (—N—H and —O—H), 5.75 ($CH_3COOC_2H_5$), 6.10, 6.26, and 6.56 (aromatic ring), and 8.0 μ ($CH_3COOC_2H_5$). The proton magnetic resonance spectrum of **7b** showed signals at 8.09 (broad multiplet, —N—H), 6.95 (triplet, =C—H at 2'), 6.30 (triplet, =C—H at 3'), 4.6 (quartet, $—CH_2—$ of $CH_3COOC_2H_5$), 2.52 (singlet, CH_2 of $CH_3COOC_2H_5$), 1.72 (singlet, CH_3 at C-17), 1.37 and 1.28 p.p.m. (C-18 H_3 and C-19 H_3).

Anal. Calcd. for $C_{22}H_{33}N \cdot 0.25CH_3COOC_2H_5$: C, 79.03; H, 10.09; N, 4.01. Found: C, 78.95; H, 10.16; N, 3.90.

In another run, two recrystallizations from ethyl acetate of the crude product of the debenzylation of 8.99 g. of **6b** gave 4.85 g. of **7b**, which was dried at 84° at 0.01 mm. and had m.p. (evacuated capillary) 215.5–216° and no ethyl acetate of crystallization.

Anal. Calcd. for $C_{22}H_{33}NO$: C, 80.68; H, 10.15; N, 4.28 Found: C, 80.74; H, 10.29; N, 4.42.

Acknowledgment.—The authors wish to thank the following members of the staff of the Sterling-Winthrop Research Institute for their contributions to this work: Mr. K. D. Fleischer and co-workers for microanalyses, Dr. F. C. Nachod and Miss C. M. Martini and co-workers for spectral determinations, Dr. R. K. Kullnig for assistance in the interpretation of proton magnetic resonance spectra, Mr. A. V. R. Crain and co-workers for the preparation of thin layer chromatographic plates, Mr. H. P. Warrington for the vapor phase chromatographic analysis, and Drs. R. L. Clarke and J. W. Dean for their criticisms of the manuscript.

The Oxidation of Gossypol. I. Early Stages in the Reaction of Gossypol and Oxygen

EKKEHARD W. SCHEIFFELE AND DAVID A. SHIRLEY[1]

Department of Chemistry, University of Tennessee, Knoxville, Tennessee

Received June 8, 1964

The oxidation of gossypol (I) in alkaline solution by oxygen proceeds *via* 1,1',6,6'-tetrahydroxy-5,5'-diisopropyl-3,3'-dimethyl(2,2'-binaphthalene)-7,7',8,8'-tetrone (II) which is formed by a Dakin-type reaction. This labile *o*-binaphthoquinone was isolated and characterized along with several of its derivatives.

It has been long known[2] that gossypol (I), the common pigment of the cottonseed, is rapidly attacked in alkaline solution by atmospheric oxygen yielding deep purple, labile compounds of intense color. The presence and fixation of such oxidation products in cottonseed oil has been suspected as a causative agent in the formation of off-color oils, but few data are available which bear directly on this problem. We have undertaken an investigation of the nature of the route(s) by which gossypol is degraded by oxygen and hydrogen peroxide in alkaline solution.

Prior work on oxidation of gossypol is confined to studies of ozonization, made as a part of the original work on structure determination,[3,4] the reported isolation of a crystalline product from the methylation of the reaction mixture from a 500-hr. treatment with oxygen of gossypol in alkaline solution,[5] and an investigation in this laboratory (to be published later) on the oxidation of gossypol with alkaline hydrogen peroxide.[6]

Principal attention in this work has been given to the isolation and characterization of labile, highly colored intermediates formed in the early stages of the reaction between oxygen and gossypol in alkaline solution. The techniques of paper and thin layer chromatography were used to monitor experiments designed to develop reaction conditions which would yield maximal amounts of the colored compounds. It was observed that uptake of 1.0–1.7 moles of oxygen/mole of gossypol corresponded to maximum production of a red-brown (neutral) or deep purple (alkaline) compound, and at this point there were also present unreacted gossypol and light-colored compounds probably representing later stages of oxidation. Workable quantities of the colored compound (II) were first obtained from column chromatographic separation of the reaction products on polyamide powder,[7] and larger amounts were later obtained directly by crystallization of a fraction of the reaction product.

The colored intermediate (II) was found to be an *o*-binaphthoquinone of the structure shown in the accompanying chart. The n.m.r. spectrum of the quinone II is summarized in Table I, and the assignments of bands given in the table are straightforward with the

(1) To whom communications concerning this paper should be addressed.
(2) L. Marchlewski, *J. prakt. Chem.*, **60**, 84 (1899).
(3) P. Karrer and E. Tobler, *Helv. Chim. Acta*, **15**, 1204 (1932).
(4) R. Adams, T. A. Geissman, W. R. Dial, and J. T. Fitzpatrick, *J. Am. Chem. Soc.*, **63**, 2439 (1941).
(5) A. Ismailov and A. S. Sadykov, *Uzbeksk. Khim. Zh.*, 34 (1959).
(6) (a) L. Farmer, Doctoral Dissertation, University of Tennessee, 1963. (b) P. W. Alley, unpublished study.
(7) H. Endres and E. Hoermann, *Angew. Chem.*, **75**, 288 (1963).