The faster moving component was isolated by thick layer chromatography (silica gel H or alumina plates, 10% ethyl acetate–benzene) and had mp 140–142°. Attempts to isolate the slower moving component were unsuccessful, the band containing significant amounts of the faster moving substance.

When the isoflavanol (1.0 g), mp 130–139°, was oxidized by the Jones procedure, 7-benzyloxy-4′-methoxyisoflavanone (0.57 g), mp 130.5–132.5°, was obtained as needles after recrystallization from methanol. The infrared spectrum contained major bands at 1689 (s), 1620 (s), 1587 (s), and 1521 (s), cm^{-1}. The ultraviolet spectrum contained λ_{max}^{MeOH} 315 mμ (log ε 3.95) and 274 mμ (log ε 4.27).

Anal. Calcd for $C_{23}H_{20}O_4$ (360.4): C, 76.65; H, 5.59. Found: C, 76.61; H, 5.55.

C. 7-Benzyloxy-4′-methoxy-Δ³-isoflavene.—A solution of the isoflavanol (4.0 g), mp 131–139°, in chloroform (200 ml) was stirred at 0–5° and a stream of dry hydrogen chloride passed through with the aid of a gas dispersion tube. The reaction was allowed to proceed for 4 hr, after which tlc (10% ethyl acetate–benzene) indicated complete dehydration. The deep orange organic solution was washed repeatedly with water until the aqueous layer afforded a pH reading of 5 and the organic layer was nearly colorless. After drying over a mixture of anhydrous sodium sulfate and sodium carbonate, the solvent was removed under reduced pressure to give a white, crystalline residue that, when dissolved in a minimum amount of boiling benzene and precipitated with methanol, provided a chromatographically uniform, white, microcrystalline solid (3.8 g), mp 142–150°. When recrystallized from benzene, the product was isolated as colorless plates (2.0 g), mp 154–155°. The infrared spectrum contained major bands at 1605 (s), 1570 (m), and 1500 (s) cm^{-1}. The ultraviolet spectrum contained λ_{max}^{MeOH} 335 mμ (log ε 4.43) and 251 mμ (log ε 4.22).

Anal. Calcd for $C_{23}H_{20}O_3$ (344.4): C, 80.21; H, 5.85. Found: C, 80.48; H, 6.02.

Hydrogenation of 7-Benzyloxy 4′-methoxy-Δ³-isoflavene.—A suspension of the Δ³-isoflavene (0.3 g, 0.874 mmole) in glacial acetic acid (20 ml) was hydrogenated at atmospheric pressure over 10% palladium–charcoal (50 mg). Complete dissolution of starting material was observed after approximately 43 ml of hydrogen had been consumed (theoretical value for 1 mole = 20.8 ml). The solution was filtered free of the catalyst by gravity and the filtrate was diluted with water to precipitate a crystalline solid. This solid consisted of two components (tlc, benzene–PMA), which were facilely separated by fractional crystallization. When dissolved in methanol (10 ml), 7-benzyloxy-4′-methoxyisoflavan (53 mg), mp 126–127.5°, crystallized as chromatographically pure platelets. The infrared spectrum contained bands at 1620 (s), 1585 (m), and 1510 (s) cm^{-1}. The ultraviolet spectrum contained λ_{max}^{MeOH} 284 mμ (log ε 3.76) and inflections at 289 (3.63) and 280 (3.73). Another crop (10 mg) was obtained by careful concentration of the mother liquor.

Anal. Calcd for $C_{23}H_{22}O_3$ (346.4): C, 79.74; H, 6.40. Found: C, 79.51; H, 6.74.

The mother liquor from the above recrystallization was allowed to evaporate to dryness at room temperature, and the white, crystalline residue was recrystallized from benzene–petroleum (bp 30–60°) ether to give 7-hydroxy-4′-methoxyisoflavan (66 mg), mp 157–158.5°, with slight softening at 153°. This crop was chromatographically uniform in two solvent systems, benzene and 10% ethyl acetate–benzene. The infrared spectrum contained major bands at 3424 (s), 1605 (s), 1590 (s), and 1500 (s) cm^{-1}. The ultraviolet spectrum contained λ_{max}^{MeOH} 284 mμ (log ε 3.73) and inflections at 289 (3.67) and 279 (3.69); $\lambda_{max}^{0.1\ N\ NaOH}$ 296 mμ (log ε 3.72) and inflections at 286 (3.70) and 278 (3.60). A second crop (17 mg) was isolated as a result of further dilution with petroleum ether. Tlc showed this fraction to be contaminated to a very slight extent by the benzyloxy derivative.

Anal. Calcd for $C_{16}H_{16}O_3$ (256.3): C, 74.98; H, 6.29. Found: C, 75.14; H, 6.31.

Registry No.—**2a**, 10499-07-7; **2b**, 10499-08-8; **2c**, 10499-09-9; **4a**, 10499-10-2; **4b**, 10499-11-3; **6**, 10499-12-4; **7a**, 10499-13-5; **7b**, 10499-14-6; 7-hydroxy-4′-methoxy-Δ³-isoflavene, 10535-63-4; 7-benzyloxy-4′-methoxyisoflavanol, 10535-64-5; 7-benzyloxy-4′-methoxyisoflavone, 10499-15-7; 7-benzyloxy-4′-methoxy-Δ³-isoflavene, 10499-16-8; 7-benzyloxy-4′-methoxyisoflavan, 10535-65-6; 7-hydroxy-4′-methoxyisoflavan, 10499-17-9.

The Synthesis of 17-Bromo-16α-methylprogesterones

HANS REIMANN AND OLGA ZAGNEETKO SARRE

Natural Products Research Department, Schering Corporation, Bloomfield, New Jersey 07003

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In situ bromination of 16α-methylpregnane-20-magnesium enolates gives a mixture of the 17α-bromo- and 17β-bromo-20-keto derivatives. Utilizing this reaction, 17α-bromo-16α-methylprogesterone and 17β-bromo-16α-methyl-17-isoprogesterone were prepared from 16-dehydropregnenolone acetate.

In recent years there has been considerable interest in the synthesis of 17α-bromoprogesterone[1] and some of its 6-substituted derivatives[2–5] in view of the enhanced progestational activity exhibited by this class of compounds. An earlier communication from these laboratories[6] described the preparation of a number of 16-alkylated progesterones. We now report the synthesis of 17α-bromo-16α-methylprogesterone (VI) and 17β-bromo-16α-methyl-17-isoprogesterone (VII).

The reaction of 16-dehydropregnenolone acetate (I) with methylmagnesium bromide in the presence of cuprous chloride, followed by *in situ* bromination of the resulting Grignard complex, afforded, after treatment with sodium iodide to debrominate any 5,6-dibromide, a mixture of 17α-bromo-16α-methylpregnenolone acetate (II)[7] and 17β-bromo-16α-methyl-17-isopregnenolone acetate (III). The 17α-bromo compound II, the major component of the mixture (90–95%), was obtained pure after column chromatography on silica gel. Fermentation of II with a culture of *Flavobacterium dehydrogenans*[6,8,9] gave 17α-bromo-16α-methylprogesterone (VI).

Direct dehydrohalogenation of the bromination mixture (II and III) with lithium bromide and lithium

(1) Ch. R. Engel and H. Jahnke, *Can. J. Biochem. Physiol.*, **35**, 1047 (1957).

(2) Ch. R. Engel and R. Deghenghi, *Can. J. Chem.*, **38**, 452 (1960).

(3) D. J. Marshall and R. Gaudry, *ibid.*, **38**, 1495 (1960).

(4) J. S. Mills, O. Candiani, and C. Djerassi, *J. Org. Chem.*, **25**, 1056 (1960).

(5) S. Rakhit, R. Deghenghi, and Ch. R. Engel, *Can. J. Chem.*, **41**, 703 (1963).

(6) E. Shapiro, T. Legatt, L. Weber, M. Steinberg, A. Watnick, M. Eisler, M. G. Hennessey, C. T. Coniglio, W. Charney, and E. P. Oliveto, *J. Med. Pharm. Chem.*, **5**, 975 (1962).

(7) P. DeRuggieri, *Farmaco, Ed. Sci.*, **16**, 583 (1961).

(8) C. Arnaudi, *Zentr. Bakteriol. Parasitenk., Abt. II*, **105**, 352 (1942).

(9) A. L. Nussbaum, F. E. Carlon, D. Gould, E. P. Oliveto, E. B. Hershberg, M. L. Gilmore, and W. Charney, *J. Am. Chem. Soc.*, **79**, 4814 (1957).

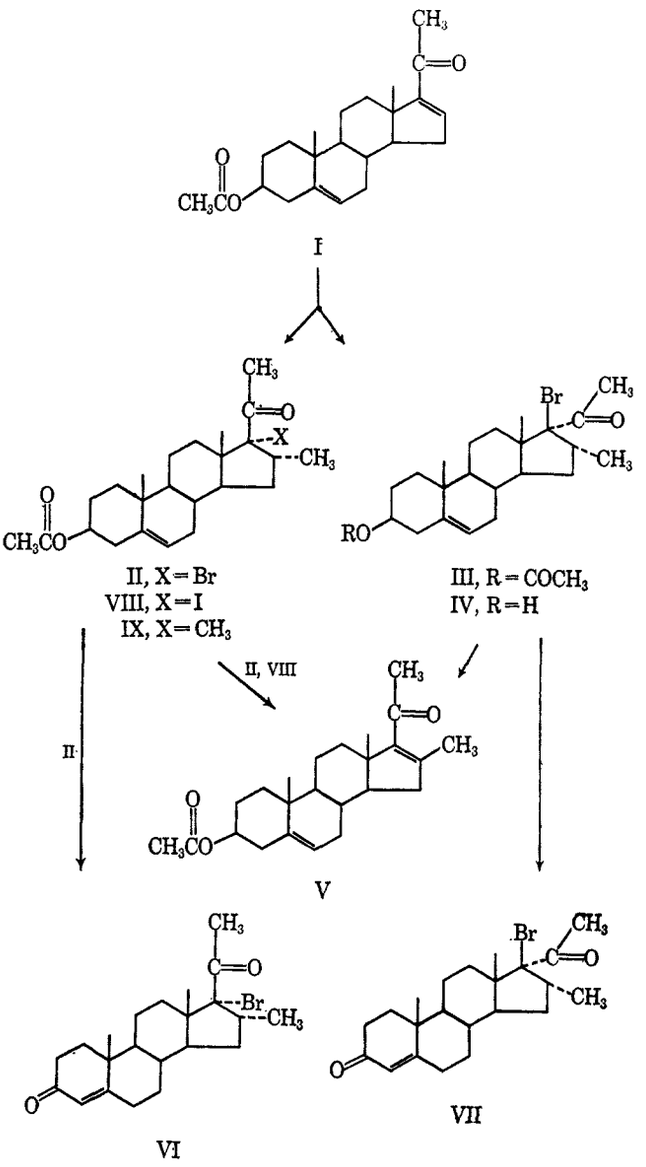
carbonate in dimethylformamide[7,10] resulted in selective reaction of II to give 16-methyl-16-dehydropregnenolone acetate (V).[11] The 17β-bromo isomer III was isolated from the dehydrobromination mixture by selective crystallization of V, followed by chromatography of the liquors on silica gel. The purity of compounds II and III was established by their nmr spectra, in which the 17β-bromo compound III exhibits a downfield shift of 19 cps for the C-18 methyl protons compared with the 17α-bromo isomer II (1.15 and 0.83 ppm, respectively).[12]

The structure of III follows from the method of synthesis and from the dehydrobromination under more forcing conditions in refluxing dimethylacetamide, to give the 16-methyl-16-dehydro compound V. Rotational differences between II and III (see the Experimental Section) and the infrared spectra are in accord with this assignment. It is of interest that 17β-bromo derivatives have not previously been reported from the bromination of 20-keto steroids or their enol derivatives. In fact, a study by Wendler and coworkers[13] showed that in the 16-desmethyl series such brominations give the pure 17α-bromo analogs, while Heusler and co-workers[14] found no evidence of 17,20β-oxide formation in the epoxidation of a 16α-methyl-17(20)-enol acetate. On the other hand, 17β attack has been observed to a minor extent in the reaction of 17-keto steroids with potassium acetylide,[15] sodium phenylacetylide,[16] and methyl Grignard reagent.[17] The reaction of 5α-9(11),16-pregnadien-3β-ol-20-one acetate with methyl Grignard reagent followed by *in situ* bromination as described above similarly gave a mixture of the 17α-bromo-16α-methyl derivative and 5 to 10% of the 17β-bromo isomer.[18]

Hydrolysis of compound III with potassium hydroxide gave the 3β-hydroxy analog IV, which on Oppenauer oxidation was converted to 17β-bromo-16α-methyl-17-isoprogesterone (VII). Alternatively, III was fermented with a culture of *Flavobacterium dehydrogenans* to afford VII directly, albeit in rather low yield. (See Scheme I.)

The synthesis of 17-iodo-20-keto steroids has not been reported. In connection with the present work it was desirable to prepare a 17-iodoprogesterone derivative. The addition of iodine to the Grignard complex from I, *in situ*, afforded 17α-iodo-16α-methylpregnenolone acetate (VIII), the stereochemistry at C-17 being assigned on the basis of rotation and predominant α attack. Dehydroiodination of VIII with lithium bromide and lithium carbonate in dimethylformamide yielded the 16-dehydro compound V. In addition to VIII, there was isolated from the iodination a minor, halogen-free product which was assigned structure IX, 16α,17α-dimethyl-5-pregnen-3β-ol-20-one 3-acetate, on the basis of spectroscopic and analytical data. In particular, the nmr spectrum[19] of IX exhibited an additional singlet methyl resonance at 1.01 ppm (or 1.04 ppm, one of the two close three-proton singlets being due to the C-19 protons) which is ascribed to the new 17α-methyl group. The formation of IX is rationalized

SCHEME I



as proceeding by methylation of the intermediate Grignard enolate complex[20] by methyl iodide, the latter being formed on reaction of excess methyl Grignard reagent with iodine. Compound VIII was rather unstable and attempts to convert it to a pure progesterone derivative, either chemically or microbiologically, were unsuccessful.

(10) R. Joly, J. Warnant, G. Nomine, and D. Bertin, *Bull. Soc. Chim. France*, 366 (1958).

(11) A. Sandoval, G. Rosenkranz, and C. Djerassi, *J. Am. Chem. Soc.*, **73**, 2383 (1951).

(12) These nmr spectra were determined on a Varian A-60 instrument in deuteriochloroform. We express our thanks to Professor L. Mandell for obtaining the nmr spectra.

(13) N. L. Wendler, R. P. Graber, and G. G. Hazen, *Tetrahedron*, **3**, 144 (1958).

(14) K. Heusler, J. Kebrle, C. Meystre, H. Ueberwasser, P. Wieland, G. Anner, and A. Wettstein, *Helv. Chim. Acta*, **42**, 2043 (1959).

(15) T. Reichstein and Ch. Meystre, *ibid.*, **22**, 728 (1939).

(16) L. Mamlok, A. M. Giroud, and J. Jacques, *Bull. Soc. Chim. France*, 1806 (1961).

(17) K. Miescher and W. Klarer, *Helv. Chim. Acta*, **22**, 962 (1939).

(18) H. Reimann and O. Zagneetko Sarre, unpublished observations.

(19) This spectrum was determined on a Varian A-60A instrument in deuteriochloroform. We thank Mr. M. Yudis of the Physical Organic Chemistry Department, Schering Corp., for the measurement and for helpful discussions.

(20) The alkylation of steroid 17(20)-enolates with alkyl halides has been reported by R. Deghenghi and R. Gaudry [*Tetrahedron Letters*, 489 (1962)], and by M. J. Weiss, R. E. Schaub, J. R. Poletto, G. R. Allen, Jr., and C. J. Coscia [*Chem. Ind.* (London), 118 (1963)].

Experimental Section[21]

17α-Bromo-16α-methyl-5-pregnen-3β-ol-20-one Acetate (II).—To a solution of methylmagnesium bromide, prepared from 0.240 g of magnesium turnings and methyl bromide, in 20 ml of tetrahydrofuran, was added under argon, with stirring, 50 mg of cuprous chloride, followed by a solution of 1.75 g of 16-dehydropregnenolone acetate in 10 ml of tetrahydrofuran. The mixture was stirred at room temperature for 30 min, then cooled in an ice bath, and a solution of 2.37 g of bromine in 5 ml of carbon tetrachloride was added with stirring at a rapid dropwise rate. The mixture was stirred for 5 min, then 50 ml of ether was added, and the excess bromine was reduced by the addition of 10% aqueous sodium bisulfite solution. The organic phase was separated, washed with 5% sodium bicarbonate solution followed by saturated sodium chloride solution, dried, and concentrated to a residue under reduced pressure. The residue was dissolved in 50 ml of acetone, 3.0 g of sodium iodide was added, and the mixture kept at room temperature for 16 hr. It was then decolorized with 10% aqueous sodium bisulfite solution and poured into cold water. The crystalline precipitate was filtered, washed with water, and dried to give 2.19 g of crude II, containing 5–10% of the 17β-bromo isomer III. A sample of 1.00 g of the crude product was chromatographed on silica gel. Fractions eluted with 2% ether–hexane were combined and crystallized from methylene chloride–methanol to give II (0.867 g), mp 185–187°. The analytical sample was recrystallized from the same solvent system: 185°; $[\alpha]_D$ −126°; $\lambda\lambda_{max}^{Nujol}$ 5.78, 5.87, 8.10 μ (lit.[7] mp 174–176°).

Anal. Calcd for $C_{24}H_{35}BrO_3$: C, 63.85; H, 7.82; Br, 17.70. Found: C, 63.78; H, 8.01; Br, 17.48.

17β-Bromo-16α-methyl-17-iso-5-pregnen-3β-ol-20-one Acetate (III).—To a stirred suspension of 69.9 g of anhydrous lithium bromide and 60.0 g of anhydrous lithium carbonate in 1100 ml of dry dimethylformamide was added 65.0 g of the crude mixture of II and III (prepared as above) and the mixture was heated under reflux with stirring for 1 hr. It was then cooled and poured into cold, dilute hydrochloric acid and the resulting precipitate was filtered, washed well with water, and dried to give 53.0 g of crude material. The latter was taken up in ether–methylene chloride, treated with decolorizing carbon, and crystallized with methanol, affording 29.7 g of 16-methyl-5,16-pregnadien-3β-ol-20-one acetate (V), mp 171–172°. A sample was recrystallized from methylene chloride–methanol: mp 173–174°; λ_{max}^{MeOH} 252 mμ (ε 9000); infrared spectrum identical with that of an authentic sample (lit.[11] mp 176–178°).

The filtrate from the original crystallization was concentrated to a residue which was chromatographed on silica gel. Fractions eluted with 2% ether–hexane were combined and crystallized from ether–methanol to give a total of 3.02 g of the 17β-bromo compound III, mp 142–143°. The analytical sample was recrystallized from ether–methanol: mp 143°; $[\alpha]_D$ +10°; $\lambda\lambda_{max}^{Nujol}$ 5.73, 5.87, 8.00 μ.

Anal. Calcd for $C_{24}H_{35}BrO_3$: C, 63.85; H, 7.82; Br, 17.70. Found: C, 63.55; H, 7.63; Br, 17.53.

Dehydrobromination of 17β-Bromo-16α-methyl-17-iso-5-pregnen-3β-ol-20-one Acetate (III).—To a solution of 100 mg of III in 5 ml of dry N,N-dimethylacetamide was added 235 mg of anhydrous lithium bromide and 190 mg of anhydrous lithium carbonate. The mixture was heated under reflux for 3.5 hr, then poured into 10% hydrochloric acid–ice, and the resulting precipitate was filtered, washed well with water, and dried, giving 75 mg of crude product. Crystallization from ether–methanol, after treatment with decolorizing charcoal, afforded 36 mg of 16-methyl-5,16-pregnadien-3β-ol-20-one acetate (V): mp 172–173°; infrared spectrum identical with that of an authentic sample.

17α-Bromo-16α-methyl-4-pregnene-3,20-dione (17α-Bromo-16α-methylprogesterone) (VI).—17α-Bromo-16α-methyl-5-pregnen-3β-ol-20-one acetate (500 mg) was incubated with a culture of *Flavobacterium dehydrogenans* for 67 hr. The fermentation mixture was extracted with chloroform and the extracts were concentrated to a residue which was crystallized from acetone, after treatment with decolorizing charcoal, to give 231 mg, mp 192–195° dec. Crystallization from methylene chloride–methanol afforded VI (199 mg), mp 199–203° dec. The analytical sample was further purified by thick layer chromatography on silica gel. The ultraviolet absorbing zone was eluted with methylene chloride and the product was crystallized with methanol: mp 197–201° dec; $[\alpha]_D$ −36°; λ_{max}^{MeOH} 240 mμ (ε 16,900); $\lambda\lambda_{max}^{Nujol}$ 5.86, 6.01, 6.16 μ.

Anal. Calcd for $C_{22}H_{31}BrO_2$: C, 64.86; H, 7.67; Br, 19.62. Found: C, 64.76; H, 7.64; Br, 20.00.

17β-Bromo-16α-methyl-17-iso-5-pregnen-3β-ol-20-one (IV).—A solution of 500 mg of compound III in a mixture of 30 ml of a 5% solution of potassium hydroxide in 95% aqueous methanol and 5 ml of methylene chloride was stirred under nitrogen at room temperature for 1 hr. The mixture was diluted with ice water, acidified with acetic acid, and extracted with methylene chloride. The extracts were concentrated and the product was crystallized from acetone–hexane to afford IV (410 mg), mp 197°. Recrystallization from acetone–hexane gave the analytical sample: mp 197–198°; $[\alpha]_D$ +10°; $\lambda\lambda_{max}^{Nujol}$ 2.82, 5.90 μ.

Anal. Calcd for $C_{22}H_{33}BrO_2$: C, 64.54; H, 8.12; Br, 19.52. Found: C, 64.80; H, 8.30; Br, 19.58.

17β-Bromo-16α-methyl-17-iso-4-pregnene-3,20-dione (17β-Bromo-16α-methyl-17-isoprogesterone) (VII). A. From IV by Oppenauer oxidation.—To a solution of 600 mg of the 3β-hydroxy-Δ⁵-compound IV in 12 ml of anhydrous toluene containing 2.5 ml of cyclohexanone was added a solution of 500 mg of aluminum isopropoxide in 10 ml of toluene and the stirred mixture was heated under reflux for 1.5 hr. The mixture was slightly cooled and 2 ml of water was added. The excess water was azeotroped off, the cooled solution was filtered, and the filtrate was steam distilled. The solid obtained on cooling of the pot residue was filtered and chromatographed on silica gel. Fractions eluted with 8% ether–hexane were combined and crystallized from methylene chloride–methanol to give a total of 390 mg of VII, mp 172–173°. Recrystallization afforded the analytical sample: mp 173°, $[\alpha]_D$ +133°; λ_{max}^{MeOH} 239 mμ (ε 17,600); $\lambda\lambda_{max}^{Nujol}$ 5.90, 5.99, 6.21 μ.

Anal. Calcd for $C_{22}H_{31}BrO_2$: C, 64.86; H, 7.67; Br, 19.62. Found: C, 64.65; H, 7.79; Br, 19.36.

B. From III by Fermentation with *Flavobacterium dehydrogenans*.—A sample of 500 mg of the 3-acetate III was incubated with a culture of *Flavobacterium dehydrogenans* for 12 hr. The fermentation mixture was extracted with chloroform and the extracts were concentrated to a residue which was taken up in acetone, treated with decolorizing charcoal, and crystallized from acetone–methanol to afford a total of 273 mg of crude VII. The material was chromatographed on silica gel, giving 128 mg of pure VII: mp 170–172°; $[\alpha]_D$ +133°; λ_{max}^{MeOH} 241 mμ (ε 16,900); infrared spectrum very similar to that of VII prepared as described in part A.

17α-Iodo-16α-methyl-5-pregnen-3β-ol-20-one Acetate (VIII) and 16α,17α-Dimethyl-5-pregnen-3β-ol-20-one Acetate (IX).—To a solution of methylmagnesium bromide, prepared from 0.240 g of magnesium in 20 ml of tetrahydrofuran, containing 50 mg of cuprous chloride, was added with stirring a solution of 1.75 g of 16-dehydropregnenolone acetate in 10 ml of tetrahydrofuran. The mixture was stirred at 10° for 25 min; then a solution of 2.5 g of iodine in 10 ml of tetrahydrofuran was added dropwise until a permanent coloration was obtained (about 5 ml of the solution was added). The mixture was stirred for 5 min, then poured into cold water. The resulting precipitate was filtered, suspended in ether, and shaken with a saturated, aqueous solution of ammonium chloride. The latter was separated and washed with ether and the ethereal solutions were combined, washed with ammonium chloride solution and water, dried, and concentrated to an oil which solidified on cooling. Trituration with cold methanol gave 1.90 g of solids. Chromatography of 600 mg of the crude product on silica gel and elution with 2% ether–hexane, followed by crystallization from methanol gave 400 mg of VIII: mp 129° dec; $[\alpha]_D$ −148°; $\lambda\lambda_{max}^{Nujol}$ 5.76, 5.90, 8.05 μ.

Anal. Calcd for $C_{24}H_{35}IO_3$: C, 57.83; H, 7.08; I, 25.46. Found: C, 57.87; H, 7.20; I, 25.02.

Further elution with the same solvent system gave, after a short break, additional solid fractions which were combined and crystallized from methylene chloride–methanol to afford 45 mg of IX: mp 218–219°; $[\alpha]_D$ −64°; $\lambda\lambda_{max}^{Nujol}$ 5.76, 5.90, 8.12 μ.

Anal. Calcd for $C_{25}H_{38}O_3$: C, 77.67; H, 9.91. Found: C, 77.36; H, 9.79.

Dehydroiodination of 17α-Iodo-16α-methyl-5-pregnen-3β-ol-20-one Acetate (VIII).—To a solution of 200 mg of 17α-iodo compound VIII in 5 ml of anhydrous dimethylformamide was added 235

(21) Melting points were determined on a Fisher-Johns apparatus. Rotations were measured in dioxane. The physical and analytical data were obtained by the Physical Chemistry Department, Schering Corp. Purity of compounds was checked by thin layer chromatography on silica gel.

mg of dry lithium bromide and 190 mg of dry lithium carbonate. The mixture was heated under reflux for 45 min, then cooled, and poured into dilute hydrochloric acid. The precipitate was filtered, washed, and dried, giving 124 mg of crude V. Chromatography on silica gel, elution with 2% ether–hexane, and crystallization from methylene chloride–methanol gave 38 mg of 16-dehydro-16-methylpregnenolone acetate (V): mp 170–172°; λ_{max}^{MeOH} 251 mμ (ε 8600); infrared spectrum matches that of an authentic sample.

Registry No.—II, 983-23-3; III, 13116-47-7; IV, 13116-48-8; V, 982-06-9; VI, 13116-50-2; VII, 13116-51-3; VIII, 13143-64-1; IX, 13116-52-4.

Acknowledgments.—We thank Dr. E. B. Hershberg for helpful suggestions in the course of this work. We are indebted to Miss C. Federbush for carrying out the microbiological transformations.

Chemistry of Erythronolide B. Acid-Catalyzed Transformations of the Aglycone of Erythromycin B

THOMAS J. PERUN

Organic Chemistry Department, Research Division, Abbott Laboratories, North Chicago, Illinois 60064

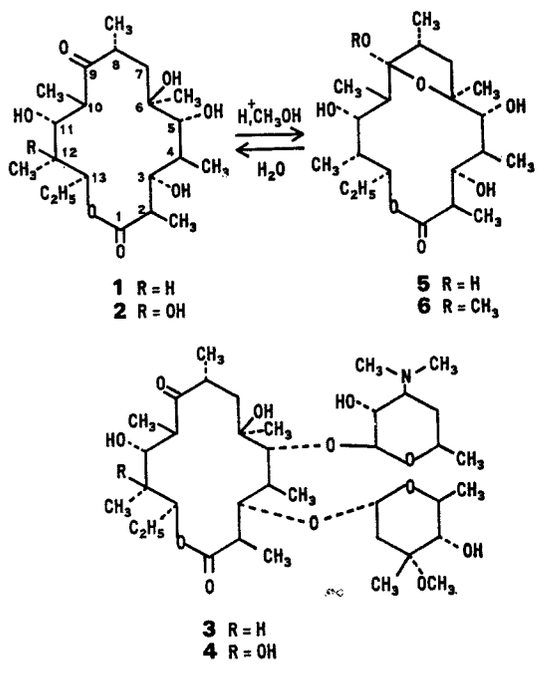
Received January 17, 1967

The treatment of erythronolide B and its derivatives with aqueous acid establishes an equilibrium between the hydroxy ketone and its corresponding 6,9-hemiketal or derived enol ether. An enol ether has been isolated from the treatment of triacetylerythronolide B. Further reaction of this enol ether with anhydrous acid caused elimination of the allylic acetoxy group giving a conjugated enol ether. Hydrolysis of this compound led to the formation of two anhydroerythronolide B derivatives, one of which was a member of the previously unknown 8-*epi*-erythronolide B series.

Erythronolide B (**1**)[1] is the 14-membered lactone portion of erythromycin B[4] (**3**), one of the macrolide family of antibiotics.[5] It has been shown to be an effective biological precursor of the erythromycins.[6] Even though erythromycin B has been shown to be more stable to acid than erythromycin A,[7] it is not possible to obtain erythronolide B by removal of the sugars, cladinose and desosamine, because the lactone ring is degraded during the severe hydrolysis procedure needed to remove the amino sugar. Similarly, erythronolide A (**2**) cannot be obtained from erythromycin A (**4**) because of extensive acid-catalyzed degradation.[8] Since we had available to us a large supply of erythronolide B from a fermentation procedure,[9] we decided to examine the nature of the acid-catalyzed degradation of this ring system (Scheme I).

It has been postulated that the acid-catalyzed degradation of erythromycin A involves the formation of a hemiketal bond between the carbonyl at C-9 and one of the tertiary hydroxyls at C-6 or C-12, followed by or concomitant with participation of the second tertiary hydroxyl giving a spiroketal.[8] Erythromycin B (and erythronolide B) does not have the tertiary hydroxyl at C-12, precluding the possibility of spiro ketal formation.[4] It should be possible, however, for a hemiketal bond to form between the carbonyl and the tertiary hydroxyl at C-6, and formation of the hemiketal **5** may be the first step in the acid-catalyzed degradation of the erythronolide B ring system. Since the ultraviolet absorption spectrum of erythronolide B contains an absorption maximum at 288 mμ (ε 39) due to the C-9 ketone, it was felt that the formation of a hemiketal could be followed by observing the disappearance of this peak.[10] Solutions were made of erythronolide B (1%) in methanol containing different concentrations of hydrochloric acid, and the absorbance of each solution at 288 mμ was recorded as a function of time. The rate of decrease in absorbance was very fast, even at low concentrations of acid. The half-life of the ketone function at room temperature in 10^{-4} *M* methanolic hydrochloric acid was slightly less than 4 min.

SCHEME I



(1) Stereochemistry is based on that determined for erythromycin A.[2,3] The structural formula used does not imply a particular conformation of the molecule but is merely a convenient planar representation of the 14-membered ring. Similarly, the double-bond geometry of the olefinic derivatives is not specified by the use of this planar structure.

(2) D. R. Harris, S. G. McGeachin, and H. H. Mills, *Tetrahedron Letters*, 679 (1965).

(3) W. D. Celmer, *J. Am. Chem. Soc.*, **87**, 1801 (1965).

(4) P. F. Wiley, M. V. Sigal, Jr., O. Weaver, R. Monahan, and K. Gerzon, *ibid.*, **79**, 6070 (1957).

(5) M. Berry, *Quart. Rev.* (London), **17**, 343 (1963).

(6) P. P. Hung, C. L. Marks, and P. L. Tardrew, *J. Biol. Chem.*, **240**, 1322 (1965).

(7) R. K. Clark, Jr., and M. Taterka, *Antibiot. Chemotherapy*, **5**, 206 (1955).

(8) P. F. Wiley, K. Gerzon, E. H. Flynn, M. V. Sigal, Jr., O. Weaver, U. C. Quarck, R. R. Chauvette, and R. Monahan, *J. Am. Chem. Soc.*, **79**, 6062 (1957).

(9) P. L. Tardrew and M. A. Nyman, U. S. Patent 3,127,315 (1964).

(10) This experiment was suggested by Dr. P. H. Jones. Preliminary observations of this effect were carried out by Dr. M. A. Nyman.