was taken to dryness *in vacuo*, dissolved in chloroform and precipitated with petroleum ether; after several recrystallizations the residue melted at 157–160° dec., λ_{max}^{alc} 256 mμ (ε 3730).

Anal. Calcd. for $C_{32}H_{51}O_5NSK$: N, 2.3; S, 5.3; K, 6.5. Found: N, 2.2; S, 4.7; K, 6.1.

The analogous derivative of 7β-hydroxycholesterol, prepared as above, m.p. 192–197° dec., λ_{max}^{alc} 256 mμ (ε 3850).

Anal. Found: K, 5.9.

Pyridinium 3,7α-Cholestanyl Disulfate.—Prepared as for the sterols; m.p. 132–135° dec., λ_{max}^{alc} 256 mμ (ε 8050).

Anal. Calcd. for $C_{37}H_{58}O_8N_2S_2$: C, 61.4; H, 8.04; N, 3.89; S, 8.81. Found: C, 61.0; H, 7.93; N, 3.92; S, 8.86.

The 3,7β-cholestanol derivative was prepared as above, m.p. 157–162° dec., $[\alpha]^{25}D$ +48° (chloroform), λ_{max}^{alc} 256 mμ (ε 7000).

Anal. Found: C, 60.9; H, 7.75; N, 3.99; S, 8.90.

Potassium 3,7α-Cholestanyl Disulfate.—Prepared in the same manner as the sterol derivative, m.p. 132–135° dec., λ_{max}^{alc} 256 mμ (ε 8050).

Anal. Calcd. for $C_{27}H_{46}O_8S_2K_2$: K, 12.2. Found: K, 13.3.

The 3,7β-cholestanol derivative was prepared similarly, m.p. 178–181° dec.

Anal. Found: K, 12.8.

BROOKLYN, NEW YORK

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF G. D. SEARLE AND COMPANY]

Derivatives of Steroids Containing a Small Ring Fused to the D Ring

BY GEORGE P. MUELLER AND BYRON RIEGEL

RECEIVED FEBRUARY 25, 1954

Ethyl diazoacetate adds to the conjugated double bond of 3β-acetoxy-5,16-pregnadien-20-one to form the Δ²-pyrazoline carboxylic ester. Pyrolysis of the corresponding free acid yields the 16-methyl-16-dehydro steroid; pyrolysis of the ester gives rise to two cyclopropanecarboxylic esters isomeric at carbon 16a. Alkaline hydrolysis of the latter proceeds with epimerization of one isomer so that the same acid, assigned the 16aβ-configuration, is derived from both esters.

Addition of ethyl diazoacetate to 3β-acetoxy-5,16-pregnadiene-20-one, an extension of the well known addition of diazomethane to 16-dehydro-20-keto steroids,[1,2,3] affords the corresponding pyrazoline carboxylic ester I. The products of diazomethane addition have been assigned Δ¹-pyrazoline structures. Examination of a typical example, XI, revealed the absence of any absorption in the infrared region corresponding to the =N—H stretching but showed a weak band at 6.45 μ probably associated with the —N=N— linkage. These observations further confirm the accepted structure of this adduct.

The diazoacetic ester adduct shows entirely different absorption characteristics. It was assigned the conjugated structure I which is in accord with the classical work in this field[4] and is supported by the infrared spectrum. Thus compound I shows a weak =N—H stretching band at 2.92 μ and a medium band at 6.39 μ due to the conjugated C=N stretching, also present in the spectra of II and III. The same bands occur at 2.93 and 6.39 μ in the model compound ethyl Δ²-pyrazoline-3-carboxylate. The conjugated C=N bond has been assigned at 6.50 μ in a similar model.[5] Additional evidence for the Δ²-pyrazoline structures was obtained from ultraviolet spectra where XI (λ_{max}^{MeOH} 227 mμ (ε 1,275); 335 mμ (ε 239)) shows entirely different absorption from the model, ethyl Δ²-pyrazoline-3-carboxylate (λ_{max}^{MeOH} 293 mμ (ε 9,260)). Compound I (λ_{max}^{MeOH} 290 mμ (ε 7577); 317 mμ (6,777)), is similar to II and III. All of the latter have 290 mμ absorption characteristic of the aforementioned model but not found in XI.

On pyrolytic decomposition these steroid adducts behave according to the predictions of von Auwers based on monocyclic systems,[6] in that 3β-acetoxy-16,17-[3,1-(1-pyrazolino)]-5-pregnen-20-one, yields mainly the 16-dehydro-16-methyl steroid with very little of the 16,17-cyclopropano derivative.[1,3] In pursuing this analogy the acetyl group at C-17 may be considered equivalent to the ester groups discussed by von Auwers. Thus, with two such groups attached, the pyrazoline I led to a mixture nearly half of which consisted of cyclopropane derivatives IV and V. If the pyrolysis of the free acid II involved initial decarboxylation, the resulting monosubstituted pyrazoline would yield little of the cyclopropane derivative. Our recovery of 3β-hydroxy-16-methyl-5,16-pregnadiene-20-one as the only product from a small-scale reaction lends support to this thesis.

Cyclopropane structures were assigned in this series on the basis of the isomerism at C-16a, discussed below, and analogy between their infrared spectra and those of 3β-acetoxy-16,17-cyclopropano-5-pregnen-20-one,[3] *i*-cholestenone and related *i*-steroid ketones.[7] The same 5.90 μ carbonyl band was present in the spectra of V, VI, IX and X and corresponds well with the presence of an adjacent cyclopropane ring. The shift in absorption of the C-20 carbonyl from 5.85 μ in II to its characteristic position in the nitrogen free derivatives was clearly evident and characteristic of the structural changes involved. The carbonyl band for IV could not be resolved but was apparent from the dissymmetry of the larger acetate band.

Configurations about the new centers of asymmetry are still subject to confirmation but may be deduced from the known course of reactions at the positions involved and from study of molecular models. Since epoxidation and catalytic addition of hydrogen, alcohols, mercaptans and other sub-

(1) A. Wettstein, *Helv. Chim. Acta*, **27**, 1803 (1944).

(2) C. Djerassi and C. R. Scholz, *J. Org. Chem.*, **14**, 660 (1949).

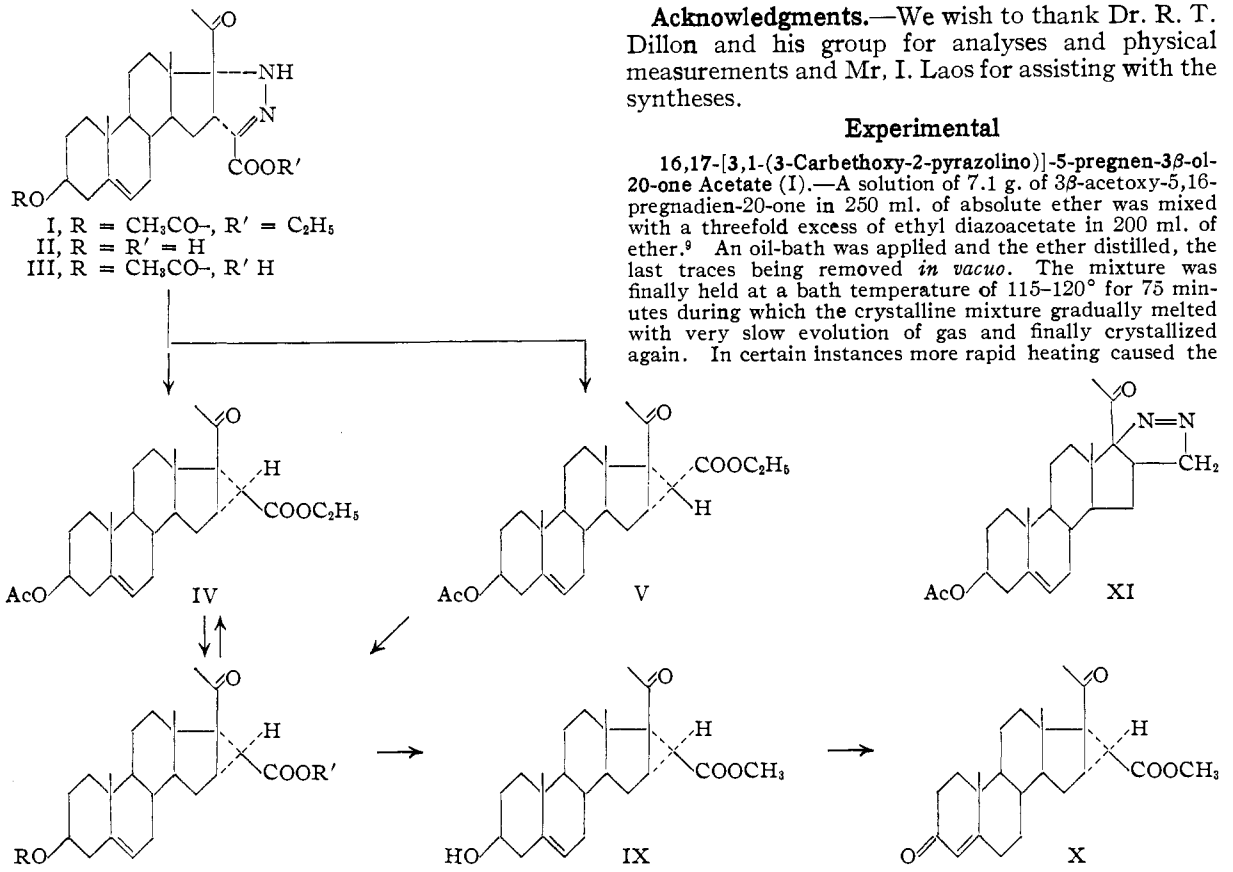
(3) A. Sandoval, G. Rosenkranz and C. Djerassi, THIS JOURNAL, **73**, 2383 (1951).

(4) K. von Auwers and O. Ungemach, *Ber.*, **66B**, 1198 (1933).

(5) H. M. Randall, R. G. Fowler, N. Fuson and J. R. Dangl, "Infrared Determination of Organic Structures," D. Van Nostrand Co., Inc., New York, N. Y., 1949, p. 34.

(6) K. von Auwers and F. König, *Ann.*, **496**, 252 (1932).

(7) M. Josien, N. Fuson and A. S. Cary, *ibid.*, **73**, 4445 (1951).



stances occur at the backside of the C-16 double bond, we assume that the pyrazoline and cyclopropane rings are fused in the 16α- and 17α-positions. Compounds IV and V have similar infrared spectra and the suspected isomerism at C-16a was confirmed by alkaline hydrolysis of each to the same hydroxy acid VI. Re-esterification and acetylation of the latter gave IV which showed that inversion had occurred during the hydrolysis of V. Molecular models demonstrated less hindrance and a higher degree of symmetry in the 16aβ-isomer, so this configuration has been assigned to the more stable, higher melting isomer, IV.

The slight solubility of hydroxy acid VI in ether gave some difficulty in preparing the methyl ester with diazomethane in the usual manner and it was more convenient to prepare IX in methanol with acetyl chloride. Similarly the convenient use of steam distillation in cleaning up the product of Oppenauer oxidation twice led to complications, presumed due to partial hydrolysis of the product X, so that chromatographic separation became necessary instead.

Cleavage of the cyclopropane ring of IV was attempted several times with hydrogen chloride in boiling acetic acid without success. Similar stability of cyclopropane-1,2-dicarboxylic acids and esters was noted by Kohler,[8] verifying earlier observations by Buchner.

(8) E. P. Kohler and L. L. Steele, *ibid.*, 41, 1093 (1919).

Acknowledgments.—We wish to thank Dr. R. T. Dillon and his group for analyses and physical measurements and Mr. I. Laos for assisting with the syntheses.

Experimental

16,17-[3,1-(3-Carbethoxy-2-pyrazolino)]-5-pregnen-3β-ol-20-one Acetate (I).—A solution of 7.1 g. of 3β-acetoxy-5,16-pregnadien-20-one in 250 ml. of absolute ether was mixed with a threefold excess of ethyl diazoacetate in 200 ml. of ether.[9] An oil-bath was applied and the ether distilled, the last traces being removed *in vacuo*. The mixture was finally held at a bath temperature of 115–120° for 75 minutes during which the crystalline mixture gradually melted with very slow evolution of gas and finally crystallized again. In certain instances more rapid heating caused the reaction to proceed with explosive violence. The cake was dissolved in benzene and recrystallized overnight from a 60-ml. volume. The product was collected and washed with cold ether; the yield was 7.3 g. Two recrystallizations at room temperature from 300 ml. of methanol yielded 5.9 g. of beautiful hexagonal rods, m.p. 223.5–225.5° (with gas evolution, sweating at 217°)[10]; $[\alpha]_D$ +188° (dioxane); λ_{max}^{MeOH} 290 mμ (ϵ 7577), 317 mμ (ϵ 6777). *Anal.* Calcd. for $C_{27}H_{38}O_5N_2$: C, 68.91; H, 8.14; N, 5.95. Found: C, 68.99; H, 8.07; N, 5.95.

16,17-[3,1-(3-Carboxy-2-pyrazolino)]-5-pregnen-3β-ol-20-one (II).—A solution of 5 g. of I was refluxed one hour with 25 g. of potassium hydroxide in 50 ml. of water. An atmosphere of nitrogen was used, and the cooled solution was diluted with 400 ml. of water and filtered to remove a small quantity of flocculent precipitate. After acidifying to congo red with 6 *N* hydrochloric acid the clear solution was concentrated *in vacuo* on the steam-bath. Crystallization proceeded as methanol was removed and the aqueous suspension was filtered to give 4.18 g. (98%) of analytically pure product in the form of short hexagons, m.p. 241–243° (vigorous gas evolution, sweating at 238°); $[\alpha]_D$ +177° (methanol); λ_{max}^{MeOH} 285 mμ (ϵ 6809), 316 mμ (ϵ 5207). *Anal.* Calcd. for $C_{23}H_{32}O_4N_2$: C, 68.97; H, 8.05; N, 7.00. Found: C, 69.16; H, 8.12; N, 7.22.

16,17-[3,1-(3-Carboxy-2-pyrazolino)]-5-pregnen-3β-ol-20-one Acetate (III).—Acetylation of II overnight in pyridine-acetic anhydride gave an ether-soluble product which was recrystallized from ethyl acetate. The product crystallized as micro hexagonal plates and appeared to be solvated; m.p. 211–213° dec., $[\alpha]_D$ +119° (chloroform); λ_{max}^{MeOH} 286 mμ (ϵ 7,300), 316 mμ (ϵ 5,710). *Anal.* Calcd. for $C_{25}H_{34}O_5N_2 \cdot CH_3COOC_2H_5$: C, 65.63; H, 7.98; N, 5.28. Found: C, 65.81; H, 7.51; N, 5.36.

Pyrolysis of 16,17-[3,1-(3-Carboxy-2-pyrazolino)]-5-pregnen-3β-ol-20-one (II).—Two hundred milligrams of II in a small test-tube was heated in a metal-bath at 250°. The

(9) Alternatively the crystalline steroid may be heated directly with the liquid diazo ester without the use of a solvent.

(10) All melting points were observed microscopically on the Kofler apparatus and are corrected.

vigorous evolution of gas had nearly ceased after 15 minutes. Three treatments of an acetone solution with Norite removed most of the dark color and two recrystallizations from the same solvent produced small rosettes of plates, melting diffusely at 185–190°. Further recrystallization from ethyl acetate gave 50 mg. of beautiful plates with the same melting range, $[\alpha]_D$ −79° (alcohol), λ_{max}^{MeOH} 252 mμ (ε 8206). *Anal.* Calcd. for $C_{22}H_{32}O_2$: C, 80.44; H, 9.82. Found: C, 80.22; H, 9.67. Wettstein[1] has reported 3β-hydroxy-16-methyl-5,16-pregnadien-20-one, m.p. 197–198°, $[\alpha]_D$ −78° (alcohol); λ_{max} 252 mμ (log ε 4.25). While not entirely pure, the pyrolytic product is obviously this compound.

Pyrolysis of 16,17-[3,1-(3-Carbethoxy-2-pyrazolino)]-5-pregnen-3β-ol-20-one Acetate (I).—Small-scale experiments with the pure compound I had demonstrated the quantitative evolution of one mole of nitrogen on pyrolysis. Therefore 16.1 g. of a crude preparation of the pyrazoline was mixed with a large quantity of boiling stones and heated in a metal-bath at 250° for 15 minutes. The cooled melt was dissolved in 2 l. of ether and crystallized from a volume of 450 ml. to give 3.32 g. of impure 3β-acetoxy-16,17-cyclopropano-16aβ-carbethoxy-5-pregnen-20-one (IV), which on further crystallization from methanol yielded the pure product as fine needles, m.p. 223.0–223.5°, $[\alpha]_D$ −34° (dioxane). *Anal.* Calcd. for $C_{27}H_{38}O_5$: C, 73.27; H, 8.65. Found: C, 73.23; H, 8.43.

Concentration of the ethereal mother liquors with slow crystallization led to a mixture of fine needles and heavy rhombs, easily separable by hand picking or by swirling the mixture and decanting the needles with the solvent. The rhombs were most suitably recrystallized several times from ethyl acetate, appearing finally as stubby prisms of pure 3β-acetoxy-16,17-cyclopropano-16aα-carbethoxy-5-pregnen-20-one (V), m.p. (subliming to needles at 180°) 201–203°, $[\alpha]_D$ −17° (dioxane). *Anal.* Found: C, 73.12; H, 8.67.

The complete separation involved a long series of fractional crystallizations and yielded a total of 4.8 g. of IV, 3.4 g. of V and an uncrystallizable glass.

3β-Hydroxy-16,17-cyclopropano-16aβ-carboxy-5-pregnen-20-one (VI).—Hydrolysis of IV was carried out by refluxing 6.2 g. under nitrogen in 1 l. of methanol containing 25 g. of potassium hydroxide in 50 ml. of water for one hour. Although the product was isolable by conventional means its insolubility rendered crystallization impractical. The following method gave an entirely suitable product without further purification. The hot solution was acidified with 40 ml. of concentrated hydrochloric acid in 300 ml. of hot water and the clear solution further diluted with 250 ml. of hot water to incipient cloudiness. Methanol was removed *in vacuo* to a total volume of 1 l. and the suspension cooled and filtered to yield 5.1 g. (98%) of crystalline product, m.p. (sweating at 280°) 285–297° dec. Recrystallization from acetone produced small prismatic needles, m.p. 297.5–300.5° dec., $[\alpha]_D$ −36.4° (dioxane). *Anal.* Calcd. for $C_{23}H_{32}O_4$: C, 74.16; H, 8.66. Found: C, 74.18; H, 8.57.

Similarly, 2 g. of V was hydrolyzed in 400 ml. of methanol with 10 g. of potassium hydroxide in 20 ml. of water. Isolation as described above yielded the theoretical quantity of a product, m.p. 261–281° dec. A small sample purified from acetone appeared as tiny, flat needles, m.p. (sweating at 287°) 297–301° dec.; $[\alpha]_D$ −34.8° (dioxane). *Anal.* Found: C, 74.04; H, 8.63.

3β-Acetoxy-16,17-cyclopropano-16aβ-carboxy-5-pregnen-20-one (VII).—Two hundred milligrams of the hydroxy acid VI was dissolved in 5 ml. of warm pyridine, cooled to room temperature, treated with 5 ml. of acetic anhydride and allowed to stand overnight. After hydrolysis in ice and water and extraction with ether, the residue from the washed ethereal solution was dissolved in benzene and crystallized by adding petroleum ether to this concentrated solution; the yield of fine needles was 152 mg., m.p. 243–245°, $[\alpha]_D$ −37.5° (dioxane). *Anal.* Calcd. for $C_{25}H_{34}O_6$: C, 72.43; H, 8.27. Found: C, 72.60; H, 8.25.

Acetylation and Esterification of 3β-Acetoxy-16,17-cyclopropano-16aβ-carbethoxy-5-pregnen-20-one (VI).—A 200-mg. sample of the hydroxy acid VI in 50 ml. of absolute alcohol was treated with 0.5 ml. of acetyl chloride. After standing 24 hours the mixture was evaporated *in vacuo* and the residue dissolved in 10 ml. of pyridine and 10 ml. of acetic anhydride. This mixture stood for 24 hours, was decomposed in ice and water, and the residue from the washed ethereal extract crystallized from methanol yielding 139 mg. of needles. After further recrystallization the melting point and mixed melting point with IV was 224–226°, identical with the latter; $[\alpha]_D$ −30° (dioxane).

β-Diethylaminoethyl Ester of 3β-Hydroxy-16,17-cyclopropano-16aβ-carboxy-5-pregnen-20-one (VIII).—A mixture of 600 mg. of VI in 30 ml. of isopropyl alcohol and 270 mg. of β-diethylaminoethyl chloride was refluxed four hours during which the acid slowly dissolved. An excess of water and sodium bicarbonate were added to the cooled solution. Extraction with ether, drying and evaporation gave 670 mg. of the crude ester. Recrystallization from ethyl acetate yielded the pure product as transparent plates, m.p. 151.3–151.9°, $[\alpha]_D$ −30° (dioxane). *Anal.* Calcd. for $C_{29}H_{45}O_4N$: C, 73.84; H, 9.62; N, 2.97. Found: C, 73.96; H, 9.68; N, 2.98.

3β-Hydroxy-16,17-cyclopropano-16aβ-carbomethoxy-5-pregnen-20-one (IX).—A solution of 300 mg. of the hydroxy acid VI in 1 l. of ether was treated with excess diazomethane. Treatment with acetic acid, washing and concentration gave elongated plates which were recrystallized from ether, m.p. 234.4–236.1°, $[\alpha]_D$ −40° (dioxane). *Anal.* Calcd. for $C_{24}H_{34}O_4$: C, 74.58; H, 8.87. Found: C, 74.70; H, 8.78.

The same product was obtained by treating a methanolic solution of hydroxy acid with a few drops of acetyl chloride, as described for the ethyl ester above.

16,17-Cyclopropano-16aβ-carbomethoxy-4-pregnene-3,20-dione (X).—Compound IX, 2.35 g., in 200 ml. of dry toluene under nitrogen was treated with 10 ml. of cyclohexanone and 3 g. of aluminum isopropoxide. After refluxing 90 minutes the cooled mixture was decomposed with 20 ml. of saturated Rochelle salt solution and extracted with ether. The dried ethereal solution was distilled to 100° at 0.5 mm. and the oily residue in benzene adsorbed on a column of 50 g. of silica gel. Much oily material was eluted with benzene, after which crystalline product was eluted with 700 ml. of 10% ether in benzene. Recrystallization from aqueous acetone gave 1.92 g. (83%) of product, m.p. 198–200°. Further crystallization from 50% aqueous acetone produced small, irregular plates, m.p. 202.5–204°, $[\alpha]_D$ +104° (dioxane), λ_{max}^{MeOH} 241 mμ (ε 15,572). *Anal.* Calcd. for $C_{24}H_{32}O_4$: C, 74.96; H, 8.39. Found: C, 74.96; H, 8.29.

SKOKIE, ILLINOIS