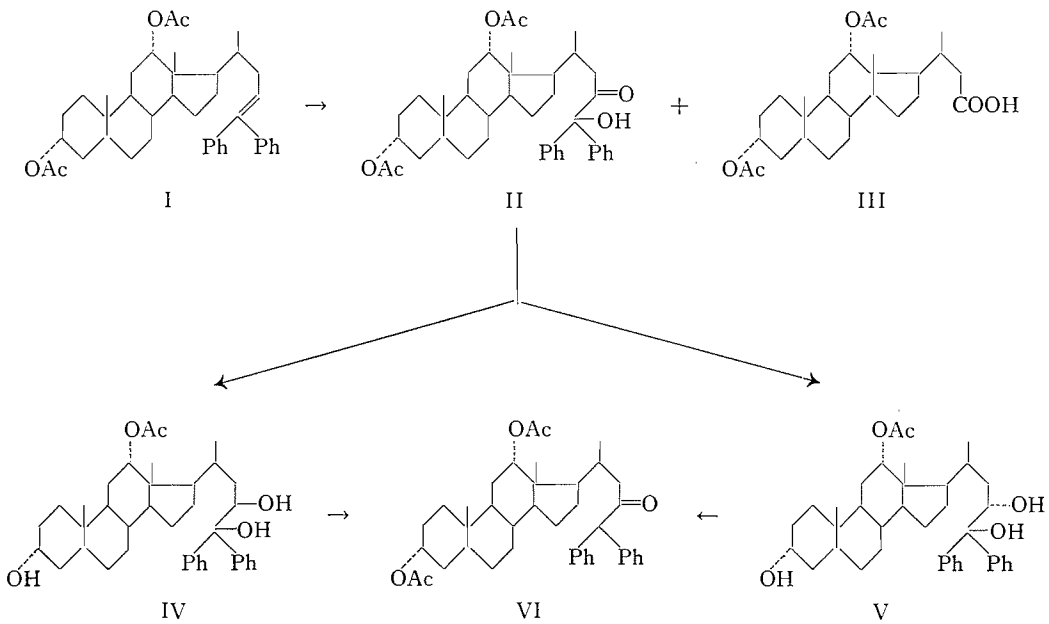
BILE ACID DERIVATIVES[1]

A. A. Amos and P. Ziegler

ABSTRACT

The potassium permanganate oxidation of 3α,12α-diacetoxy-24,24-diphenyl-Δ^{23}-cholene yielded a new compound which was identified as 3α,12α-diacetoxy-23-keto-24-hydroxy-24,24-diphenylcholane. The latter steroid, on reduction with sodium borohydride, produced two isomeric glycols which were subsequently dehydrated to provide the corresponding 23-keto-24,24-diphenyl derivative.

Desoxycholic acid is used extensively as a starting material for the preparation of steroidal hormones. One important phase of this conversion consists of the partial degradation of the bile acid side chain which is attached to C-17 of the steroid nucleus. This degradation can be accomplished by the classical Barbier–Wieland procedure, or preferably by the Miescher–Wettstein (1) scheme. Both methods produce the acetylated diphenylethylene derivative I as an intermediary product. According to the directions of Miescher and Wettstein, I is brominated in the allylic position, then dehydrobrominated to the corresponding diene, which in turn is oxidized to the 20-ketone. It has been found in our laboratories that the dienes thus produced are often contaminated with the ethylene derivative I; this impurity can be detected by its absorption at 252 mμ. When the crude diene was then treated with potassium permanganate, a useful (2) oxidizing agent for such compounds, the isolation of 3α,12α-diacetoxypregnan-20-one proved to be difficult; in fact, it was found that the yields of 20-ketone increased markedly with increasing purity of diene. This observation indicated that I or its oxidation products interfered with the normal course of the permanganate oxidation of the diene, or with the subsequent isolation of the 20-ketone. It was therefore decided to investigate the products derived from I by this oxidation.



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The oxidation was carried out in refluxing aqueous acetone, containing potassium dihydrogen phosphate as a buffer, which provided the desired neutral pH. The reaction products were then separated to give 30% of acidic material which was identified as nordesoxycholic acid diacetate (III). The neutral fraction, after crystallization, afforded 45% of a compound (II) which in turn was converted to III by oxidation with chromic acid. Compound II failed to react with the Malaprade reagent or with acetic anhydride and pyridine at 20°, and on the basis of this evidence a 23,24-glycol structure was eliminated. The presence of a ketone group in II was indicated clearly by its ultraviolet and infrared absorption, though the compound did not form a 2,4-dinitrophenylhydrazone or an oxime. This result is attributed to steric effects which are known (3) to be operative in similar cases. The ultraviolet spectrum of the ketol II showed bands at 260 mμ (ϵ = 440) and at 297 mμ (ϵ = 360). The absorption at the lower wavelength is undoubtedly due to the two unconjugated phenyl groups (4, 5) in the molecule; the band at the higher wavelength results from the interaction of the ketone function with the phenyl groups. In studies of α,α-diphenylketones, it has previously been shown (4, 6, 7) that the carbonyl absorption peak in the ultraviolet region is more intense and is shifted towards longer wavelength than that of most ketones. The above data as well as the results of elemental analyses for II and its hydrolysis product provided the evidence for the ketol structure in these compounds.

When the ketol II was reduced with sodium borohydride and the reaction products were hydrolyzed, two isomeric glycols (IV and V) were isolated. Both compounds gave positive Malaprade tests, and the predominant isomer (IV) formed a well-defined acetonide; since V was obtained in only small yield, its reactivity towards acetone could not be determined. The two isomers showed the ultraviolet absorption bands at 260 mμ due to the phenyl groups, but the ketone peak was absent; in the infrared region there appeared the characteristic absorption of associated hydroxyl groups. On treatment with acidic reagents, both glycols yielded 3α,12α-diacetoxy-23-keto-24,24-diphenylcholane (VI). The latter compound again showed the ketone band at 293 mμ, and in the infrared the hydroxyl peak was absent. These findings, in conjunction with the results of elemental analyses, established the structures for IV, V, and VI.

The oxidation of 3α,12α-diacetoxy-24,24-diphenyl-Δ^{23}-cholene (I) by chromic acid has been reported (8, 9) to yield 60–70% of the noracid III; ozonization (10) of the diphenylethylene derived from cholic acid afforded the corresponding noracid in lower yield. Invariably, these oxidations produced relatively large amounts of neutral, unidentified fractions which may well contain the related ketols. The good yield of II obtained from the permanganate oxidation is not surprising in view of some recent reports. In the steroid field, this oxidizing agent has been employed to produce ketols from the corresponding $\Delta^{5,6}$-unsaturated compounds (11) as well as from $\Delta^{17,20}$-analogues (12, 13). The mild permanganate treatment (14) of oleic acid similarly provided high yields (70–75%) of ketohydroxystearic acid.

EXPERIMENTAL[2,3]

The Permanganate Oxidation of 3α,12α-Diacetoxy-24,24-diphenyl-Δ^{23}-cholene (I)

Compound I (20 g) and potassium dihydrogen phosphate (2 g) were suspended in acetone (195 ml) and water (5 ml). The mixture was stirred and there was then added, in

[2]*The microanalyses were kindly performed by Mr. E. Thommen, Basel, Switzerland.*

[3]*The infrared absorption data were obtained through the courtesy of Dr. G. D. Laubach of Chas. Pfizer & Co., Brooklyn, N.Y.*

four equal portions, at intervals of 30 minutes, potassium permanganate (32 g). On addition of each batch of oxidizing agent, vigorous refluxing began and gradually subsided after several minutes. After all the permanganate had been added, the reaction was continued under reflux conditions for an additional $2\frac{1}{2}$ hours. The excess oxidizing agent was destroyed by charcoal (5 g) and, after 1 hour's stirring, the solids were filtered off. The filtrate was evaporated to dryness, the residue (16 g) was dissolved in ethyl acetate and extracted with 2.5% sodium carbonate solution. After being washed with water, the solvent extract was evaporated, the residual gum was taken up in ether to provide, on cooling, 9.45 g (45%) of II, m.p. 169–171° C. Recrystallization from acetone gave material, m.p. 175–175.5° C, and $[\alpha]_D^{25}$ +81.3° (*c*, 0.994, dioxane). This compound failed to react with a solution of bromine in carbon tetrachloride, with 2,4-dinitrophenylhydrazine, trichloroacetic acid, the Zimmermann reagent, and was negative in the Malaprade test. Its ultraviolet spectrum showed weak bands at 260 mμ (ϵ = 440) and at 297 mμ (ϵ = 360). Analysis: Calculated for $C_{40}H_{52}O_6$: C, 76.39; H, 8.34; O, 15.27. Found: C, 76.37, 76.48; H, 8.56, 8.47; O, 15.37, 15.41.

The carbonate extract as well as the water washings from the cake of solids (manganese salts, etc.) were acidified and extracted with ether. The solvent phase was washed, dried, and evaporated; crystallization of the residue from ether–hexane afforded 4.5 g (29%) of crude 3α,12α-diacetoxy-norcholanic acid (III), m.p. 180–190° C. This material was recrystallized from hexane–ether to give a melting point of 203–205° C.

3α,12α-Diacetoxy-norcholanic Acid (III) *from* II

Compound II (1 g) was suspended in acetic acid (15 ml) and there was then added dropwise, over a period of $\frac{1}{2}$ hour, a solution of chromic acid (1 g) in acetic acid (24 ml) and water (1 ml). The mixture was kept at 20° C for 2 hours and an equal period of time at 35° C with occasional shaking. The excess oxidizing agent was decomposed by sodium bisulphite; water was added, and the products were extracted with ether. The solvent phase, after having been washed, dried, and evaporated, yielded a residue (1.12 g) which crystallized readily from acetone. The noracid III (472 mg), m.p. 203–205° C, thus obtained, was recrystallized from ether–hexane to give pure III, m.p. 211–213° C.

3α,24-Dihydroxy-12α-acetoxy-23-keto-24,24-diphenylcholane

The diacetoxy-ketol II (0.5 g) was dissolved by warming in methanol (24 ml) and water (1 ml), containing potassium hydroxide (0.2 g). The solution was kept at room temperature for 66 hours, after which time most of the methanol was removed *in vacuo*. The residue was taken up in ether and dilute mineral acid, the ether phase was washed to neutrality, dried, and evaporated to give a residue (0.46 g) which crystallized from ether–hexane to yield 367 mg of material, m.p. 188–190° C. Two further recrystallizations from ether–hexane provided needles, m.p. 190–191.5° C and $[\alpha]_D^{24}$ +68.9° (*c*, 0.779, dioxane). Analysis: Calculated for $C_{38}H_{50}O_5$: C, 77.78; H, 8.59; O, 13.63. Found: C, 77.83, 77.91; H, 8.75, 8.75; O, 13.63.

Sodium Borohydride Reduction of the Ketol II

Compound II (1 g) was partially dissolved in ether (50 ml) and sodium borohydride (0.5 g) was added. The mixture was kept at 20° C for 15 hours, and was subsequently refluxed for $3\frac{1}{2}$ hours. The excess reducing agent was destroyed by addition of acetic acid, the mixture was diluted with water and extracted exhaustively with ether. The combined solvent extracts were washed, dried, and evaporated to provide a residue (1 g), which

was saponified at room temperature for 15 hours with 2.5% methanolic potassium hydroxide. Dilution with water, extraction with ether, and working up in the usual fashion gave the residue (0.98 g) which crystallized from ether–hexane to provide, in two crops, 451 mg of material; these crystals melted at 127–135° C, the melt resolidified at 145–150° C and finally melted at 215–218° C. Two recrystallizations from ethyl acetate yielded pure IV, m.p. 216.5–219° C and $[\alpha]_D^{25}$ +121.2° (*c*, 0.366, dioxane). This substance gave a positive Malaprade test; in the ultraviolet region, it exhibited a weak band at 260 mμ (ϵ = 380). Analysis: Calculated for $C_{38}H_{52}O_5$: C, 77.51; H, 8.90; O, 13.59. Found: C, 77.66, 77.55; H, 9.07, 8.99; O, 13.65, 13.74.

The filtrate from IV was reduced in volume and, on refrigeration, three more crops of crystals were isolated. These weighed 295 mg and had a melting point of 177–185° C. Several recrystallizations from ether–hexane provided a small, pure sample of V, m.p. 193–194° C and $[\alpha]_D^{23}$ +16.9° (*c*, 0.655, dioxane). This compound, on admixture with IV, had a melting point of 178–183° C; V, like IV, gave a positive Malaprade test. Analysis: Calculated for $C_{38}H_{52}O_5$: C, 77.51; H, 8.90; O, 13.59. Found: C, 77.78; H, 9.15; O, 13.24.

The Acetonide of IV

Compound IV (250 mg) was dissolved in dry acetone (20 ml), *p*-toluenesulphonic acid (50 mg) was added, and the solution was kept at 25° C for 16 hours. The solution was neutralized with solid sodium bicarbonate, diluted with water, and extracted with ether. The solvent extract was washed, dried, and evaporated to a white solid (270 mg) which crystallized from ether–hexane to give 220 mg of material, m.p. 227–228° C. Three recrystallizations from the same solvent system yielded the acetonide of IV, m.p. 228–230° C and $[\alpha]_D^{25}$ +158.3° (*c*, 0.647, dioxane). Analysis: Calculated for $C_{41}H_{56}O_5$: C, 78.30; H, 8.98; O, 12.72. Found: C, 78.42, 78.34; H, 9.17, 9.22; O, 12.79, 12.68.

3α,12α-Diacetoxy-23-keto-24,24-diphenylcholane (VI)

(a) From IV

Compound IV (120 mg) was refluxed gently for 2 hours with acetic acid (8 ml) and concentrated hydrochloric acid (0.8 ml); an additional 0.8 ml of hydrochloric acid was added then, and refluxing was continued for 2 more hours. The reaction mixture was poured into water, extracted with ether, and the solvent phase worked up in the customary manner. Crystallization of the residue from hexane–ether yielded 70 mg of substance which melted at 127–129° C, partially recrystallized, and finally melted at 155–158° C. Three recrystallizations from hexane–ether provided a pure sample of VI, having a double melting point 133–136° and 167–170° C. When VI is crushed to a very fine powder, it melts at 167–168° C; $[\alpha]_D^{23}$ +67.5° (*c*, 0.591, dioxane). This compound absorbs in the ultraviolet at 260 mμ (ϵ = 515) and at 293 mμ (ϵ = 270). Analysis: Calculated for $C_{40}H_{52}O_5$: C, 78.39; H, 8.55; O, 13.05. Found: C, 78.54, 78.77; H, 8.88, 8.53; O, 13.14, 13.05.

(b) From V

Compound V (80 mg) and *p*-toluenesulphonic acid (300 mg) were refluxed for 3 hours in acetic acid (8 ml). The solution was diluted with water and then extracted with ether; washing, drying, and evaporating the solvent phase provided a residue (86 mg) which crystallized from hexane–ether to give 27 mg of crude VI, m.p. 160–164° C. Recrystallization from ethanol gave pure VI, showing the double melting point, 132–135° C and 165.5–167° C. This material gave no melting point depression on admixture with a sample of VI obtained by dehydration of IV.

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