[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, COLLEGE OF MEDICINE, UNIVERSITY OF UTAH]

Δ^4-CHOLESTENE-6-ONE AND RELATED COMPOUNDS[1]

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In connection with studies carried out in this laboratory, Δ^4-cholestene-6-one (VI) was required for comparison. In 1939 Ladenburg *et al.* (1) obtained an unsaturated ketone by refluxing 3α-bromocholestane-6-one with quinoline and assumed that this compound was Δ^4-cholestene-6-one. Several years later Blunschy *et al.* (2) demonstrated that it was actually Δ^2-cholestene-6-one.

For our preparation of Δ^4-cholestene-6-one a series of reactions somewhat similar to the synthesis of Δ^4-androstene-6,17-dione (3) was employed. The starting material, cholestane-3β,5α-diol-6-one (I) was obtained in excellent yield by oxidation of cholesterol with hydrogen peroxide in formic acid, followed by saponification and further oxidation with N-bromosuccinimide (4). From the mother liquor of I, after acetylation, a small amount of 7-ketocholesteryl acetate was isolated by chromatography. Recently Clemo and co-workers (5) reported the formation of this unsaturated ketone as a by-product of the action of hydrogen peroxide on cholesteryl acetate and discussed a mechanism for this unusual reaction.

Cholestane-3β,5α-diol-6-one (I) gave, as expected, the monotosylate II on treatment with tosyl chloride in pyridine. By chromatographic separation, a sulfur-free by-product was obtained the analysis of which was in accordance with the empirical formula $C_{27}H_{46}O_3$. This compound was found to be a ketone, different from I, and might be either cholestane-5α,6β-diol-3-one or cholestane-3α,5α-diol-6-one. Both of these compounds have not been described in the literature.

The infrared spectrum of the tosylate II showed bands at 2.70, 5.78, 8.40 and 8.50μ. The latter two bands seem to be characteristic for the tosyloxy group, since they also appeared in the spectra of cholesteryl tosylate (6), methyl 3α-tosyloxy-12α-hydroxycholanate (7) and methyl 3α-tosyloxy-12-ketocholanate (see experimental part).

Upon refluxing with pyridine, the tosylate II gave an unsaturated ketol which is presumed to be Δ^2-cholestene-5α-ol-6-one (III), analogous to the formation of Δ^2-cholestene-6-one from 3-chloro- and 3-bromo-cholestane-6-one upon refluxing with quinoline (2). The unsaturated ketol III formed an oxime and a semicarbazone. On treatment with 2,4-dinitrophenylhydrazine in the presence of hydrochloric acid, it gave a red dinitrophenylhydrazone with maximum absorption at 408 mμ. Since this maximum is characteristic for dinitrophenylhydrazones of $\alpha,\beta;\gamma,\delta$-doubly unsaturated ketones, it is evident that the hydroxyl group in the 5-position was split off during the reaction with dinitrophenylhydrazine

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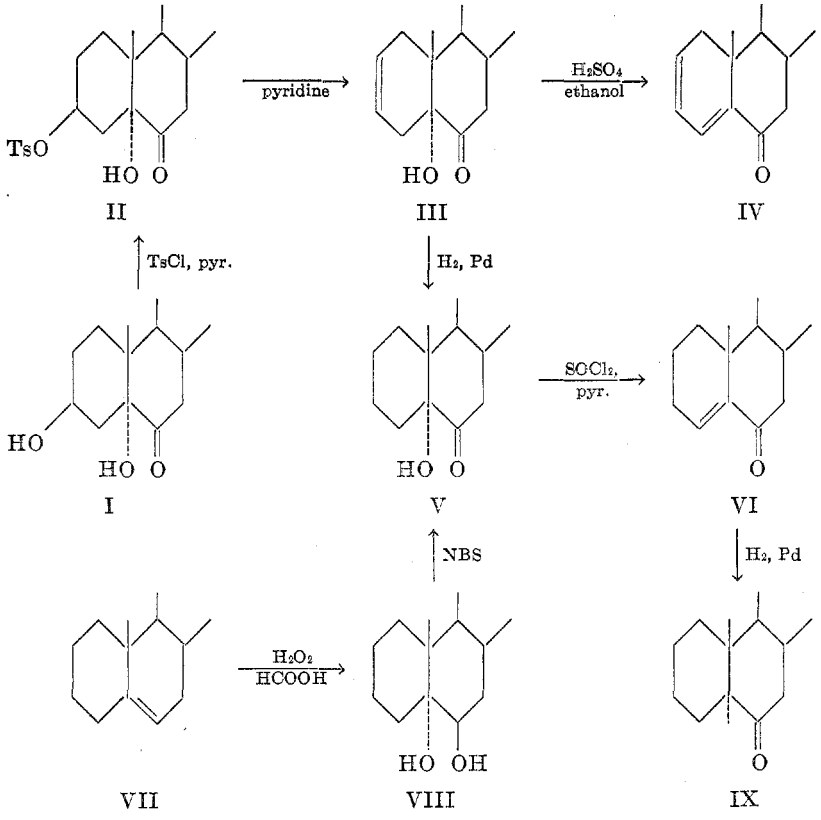
in acidic solution, and that the dinitrophenylhydrazone of IV was formed. Similar reactivity of the 5-hydroxyl group was observed by Ehrenstein *et al.* (8), who found that 3-keto-5-hydroxy compounds gave rise to Δ^4-3-ketosteroids when treated with Girard's reagent T. The loss of water during formation of a dinitrophenylhydrazone also resembles the loss of hydrogen bromide from 3-keto-4-bromo compounds on treatment with dinitrophenylhydrazine (9).



The doubly unsaturated ketone IV was prepared both by introduction of hydrogen chloride into a cold solution of III in chloroform, and by refluxing a solution of III in a mixture of ethanol and sulfuric acid. The ketone IV showed maximum absorption at 314 mμ (log ϵ 3.882), indicating that the two double bonds are located in the same ring. As mentioned in an article by Ross (10) the ketone IV has been preparted by E. R. H. Jones (unpublished work) who found maximum absorption at 316 mμ (log ϵ 3.875).

The dinitrophenylhydrazone of IV was identical with that prepared from III. The semicarbazone of IV which was obtained only in poor yield proved to be a mixture of a saturated and an unsaturated semicarbazone, since it showed two maxima in the ultraviolet, at 225 mμ and at 305–310 mμ. It is assumed that not only the keto group, but to some extent also the double bond in the 4,5-

position has entered into reaction with semicarbazide to give a semicarbazino-semicarbazone, which should absorb like a saturated semicarbazone between 225 and 230 mμ.

On hydrogenation with palladium-on-calcium carbonate the unsaturated ketol III gave cholestane-5α-ol-6-one (V). This saturated ketol was also synthesized using Δ^5-cholestene (VII) as the starting material. When VII was treated with hydrogen peroxide in formic acid, a diol was obtained which is designated as cholestane-5α,6β-diol (VIII) in analogy to the formation of cholestane-3β,5α,6β-triol from cholesterol. The diol VIII is probably identical with that prepared by Ruzicka and Thomann (11) by hydrolysis of both isomeric cholestene oxides. It gave a monacetate, but failed to react with 3,5-dinitrobenzoyl chloride. Upon oxidation with N-bromosuccinimide or chromium trioxide cholestane-5α-ol-6-one (V) was obtained, which was characterized by its oxime and its semicarbazone. On refluxing with acetic anhydride, V gave an acetate and a small amount of an α,β-unsaturated ketone (probably VI), which was recognized by its ultraviolet absorption. The 2,4-dinitrophenylhydrazone prepared from cholestane-5α-ol-6-one (V) in the presence of hydrochloric acid was bright orange and showed maximum absorption at 375 mμ. The analysis of this derivative indicated that it was the dinitrophenylhydrazone of the unsaturated ketone VI, and that the hydroxyl group in the 5-position was split off during the reaction. Further proof for the correctness of this assumption was provided by preparation of the same dinitrophenylhydrazone from VI. The wavelength of the absorption maximum, 375 mμ, is surprisingly low in comparison with the maxima of dinitrophenylhydrazones of Δ^4-3-keto- and Δ^1-3-keto-steroids, which were found to be at 391 mμ and 383 mμ respectively (see table I).

The ketone VI was obtained from cholestane-5α-ol-6-one (V) by treatment with thionyl chloride in pyridine (12). It melted at 109° and showed maximum absorption at 243 mμ in abs. ethanol. The dinitrophenylhydrazone was identical with that prepared from V. The oxime failed to crystallize, while the semicarbazone was obtained only in poor yield and could not be purified completely. It showed maximum absorption at 253 mμ, a wavelength considerably shorter than the maxima of other semicarbazones of α,β-unsaturated ketones (approximately 270 mμ). For comparison the semicarbazone of Δ^4-cholestene-3β-ol-6-one acetate (13) was prepared, which showed maximum absorption at 258 mμ, similar to the semicarbazone of VI. The unsaturated ketone VI, on hydrogenation in the presence of palladium-on-calcium carbonate, gave the known cholestane-6-one (IX), which was characterized by its oxime (19).

The absorption maxima of Δ^4-cholestene-6-one (VI), Δ^4-cholestene-3-one, Δ^1-cholestene-3-one, Δ^1-coprostene-3-one and of their derivatives are listed in Table I.

EXPERIMENTAL[2]

Semicarbazones were prepared according to two different methods. Method I: One hundred mg. of semicarbazide hydrochloride and 150 mg. of sodium acetate trihydrate were

[2] All melting points were taken on a Kofler micro hot stage and are corrected. The micro analyses were carried out by Drs. Weiler and Strauss, Oxford, England. Infrared spectra

ground until the mixture liquified; it was then taken up in abs. ethanol. The sodium chloride was removed, washed with ethanol, and the filtrate diluted to 10 cc. Ten mg. of the ketone was refluxed for 1.5 hours with 1–2 cc. of the semicarbazide acetate solution. After addition of water the ethanol was taken off *in vacuo*, and the semicarbazone was either filtered or extracted with ether.

Method II: To a solution of 500 mg. of semicarbazide hydrochloride in 1.5 cc. of water a solution of 500 mg. of potassium acetate in 5 cc. of abs. ethanol was added, and the potassium chloride was filtered off. Twenty mg. of the ketone was dissolved in 0.25 cc. of abs. ethanol and 0.25 cc. of pyridine, and 0.5 cc. of the semicarbazide acetate solution was added. The mixture was allowed to stand at room temperature for three to four days. After addition of water the semicarbazone was either filtered or taken up in ether. In the latter case the ether solution was washed several times with 2 *N* hydrochloric acid and water to remove the pyridine completely.

TABLE I

ULTRAVIOLET MAXIMA OF α,β-UNSATURATED KETOSTEROIDS AND THEIR DERIVATIVES

	KETONE		DINITROPHENYLHYDRAZONE		SEMICARBAZONE	
	mμ	log ε	mμ	log ε	mμ	log ε
Δ^4-Cholestene-6-one	243	3.804	377	4.396	253	3.852
	ethanol		chloroform		ethanol	
Δ^4-Cholestene-3-one	240.5	4.255	391	4.505	273	4.420
	ethanol (14)		chloroform (14)		ethanol (15)	
Δ^1-Cholestene-3-one	230	4.033	383	4.48	unknown	
	ethanol (16)		chloroform (17)			
Δ^1-Coprostene-3-one	232	4.009	unknown		270	3.985
	ethanol (18)				chloroform (18)	

Cholestane-3β,5α-diol-6-one (I). This ketone was prepared by hydroxylation of cholesterol, followed by oxidation with N-bromosuccinimide according to the method of Fieser (4). The last mother liquor was acetylated and chromatographed. By elution with hexane-benzene 2:3 and recrystallization from methanol and 85% acetic acid a small amount of 7-ketocholesteryl acetate, m.p. 152–155° was obtained; u.v. maximum at 235 mμ, infrared bands at 5.71, 5.92, 8.05, 8.46 and 9.58 μ. Cholestane-3β,5α-diol-6-one 3-acetate was eluted with benzene and mixtures of benzene and ether.

Cholestane-3β,5α-diol-6-one 3-tosylate (II). To a solution of 1.34 g. of I in 4 cc. of abs. pyridine 670 mg. of tosyl chloride was added. The mixture was allowed to stand at room temperature overnight. After addition of water the precipitate was filtered, washed with water and dried. It weighed 1.715 g. and melted at 145–153° (dec.). For purification it was chromatographed on 51 g. of aluminum oxide. The fractions eluted with benzene and mixtures of benzene and ether up to 20% ether yielded 691 mg. of the tosylate II. It was recrystallized from acetone-hexane and gave prisms with m.p. 161–163° (dec.); infrared bands at 2.70, 5.78, 8.40 and 8.50 μ.

Anal. Calc'd for $C_{34}H_{52}O_5S$: C, 71.29; H, 9.15; S, 5.60.
Found: C, 71.39; H, 9.05; S, 5.38.

were taken in carbon disulfide on a Beckman spectrophotometer model IR 2. For all chromatographies aluminum oxide Merck "Suitable for Chromatographic Adsorption" was used.

The fractions eluted with benzene-ether 3:2 and 2:3 were recrystallized twice from acetone-hexane and gave crystals, m.p. 195–197° which proved to be sulfur-free; infrared bands at 2.83 and 5.80 μ.

Anal. Calc'd for $C_{27}H_{46}O_3$: C, 77.46; H, 11.08.
Found: C, 77.38; H, 11.08.

The analysis indicates that this material is a cholestanediolone. From the last fractions, eluted with chloroform and ethanol, 400 mg. of unchanged starting material was isolated.

Δ^2-*Cholestene-5α-ol-6-one* (III). A solution of 670 mg. of tosylate II in 35 cc. of abs. pyridine was refluxed for 36 hours. The pyridine was evaporated *in vacuo* and the residue extracted several times with ether; some material remained undissolved. The combined ether solutions were washed with hydrochloric acid, water, sodium carbonate solution and water, dried and evaporated. The residue, 388 mg., was chromatographed on 12 g. of aluminum oxide. The unsaturated ketol III was eluted with mixtures of hexane and benzene up to 70% benzene. After sublimation in a high vacuum at 160° and recrystallization from ether-pentane, needles were obtained, m.p. 140–141.5°, $[\alpha]_D^{23} - 25.1° \pm 3°$ (abs. ethanol); infrared bands at 2.70 and 5.79 μ.

Anal. Calc'd for $C_{27}H_{44}O_2$: C, 80.94; H, 11.07.
Found: C, 80.79; H, 11.11.

The *oxime* was obtained as needles from dil. ethanol; it melted over a wide range between 160 and 180°.

Anal. Calc'd for $C_{27}H_{45}O_2N$: C, 78.02; H, 10.91; N, 3.37.
Found: C, 78.07; H, 10.96; N, 3.32.

The *semicarbazone* prepared according to method II was chromatographed on aluminum oxide and eluted from the column with ether and chloroform. It was recrystallized from dil. ethanol; m.p. 200–202°, u.v. maximum at 226 mμ (log ϵ 4.082, ethanol).

Anal. Calc'd for $C_{28}H_{47}O_2N_3$: C, 73.47; H, 10.35; N, 9.18.
Found: C, 73.10; H, 10.27; N, 8.91.

When method I was employed for the preparation of the semicarbazone, 56% of unchanged starting material was recovered by chromatography.

$\Delta^{2,4}$-*Cholestadiene-6-one* (IV). A solution of 64.3 mg. of Δ^2-cholestene-5α-ol-6-one (III) in 3 cc. of abs. ethanol and 0.1 cc. of conc. sulfuric acid was refluxed for two hours. An aqueous solution of 260 mg. of potassium carbonate was added, and the mixture was extracted with ether. The ether solutions were washed with water, dried and evaporated, and the residue (65.7 mg.) was chromatographed on 2.5 gm. of aluminum oxide. All fractions eluted with hexane were combined, sublimed in a high vacuum at 135° and recrystallized from acetone and from abs. ethanol. Needles were obtained, m.p. 129–130°; u.v. maximum at 314 mμ (log ϵ 3.882, ethanol), infrared band at 5.93 μ.

Anal. Calc'd for $C_{27}H_{42}O$: C, 84.75; H, 11.07.
Found: C, 84.54; H, 10.72.

The same substance was obtained, when a stream of dry hydrogen chloride was introduced into a cooled solution of the unsaturated ketol III in chloroform for three hours. In this case the yield of IV was rather low, and a considerable amount of starting material was recovered by chromatography.

The *semicarbazone* was prepared from 13.9 mg. of the diene IV according to method I. Chromatography of the crude reaction product gave 6.9 mg. of starting material and two fractions of semicarbazones; the first, eluted with benzene-ether 2:3, weighed 3.9 mg.; the second, eluted with ether and chloroform, weighed 2.3 mg. The first fraction was recrystallized from dil. ethanol; m.p. 125–130°, u.v. maxima at 225 and 305–310 mμ (log ϵ 3.851 and 3.749 respectively, ethanol). The second fraction showed only one maximum at 225 mμ, and failed to crystallize.

$\Delta^{2,4}$-*Cholestadiene-6-one 2,4-dinitrophenylhydrazone.* (*a*) *From* Δ^2-*cholestene-5α-ol-6-one* (III). A solution of 21 mg. of the unsaturated ketol III in 2 cc. of abs. ethanol was mixed with a solution of 17 mg. of dinitrophenylhydrazine in 2 cc. of abs. ethanol and four drops of conc. hydrochloric acid, and refluxed for five minutes. Red crystals precipitated which

were washed with ethanol and dried. They were purified by chromatography (eluted with hexane-benzene 9:1) and recrystallized from ethanol. On slow cooling fine red needles with m.p. 189–193° were obtained; u.v. maxima at 257, 320 and 408 mμ (log ε 4.067; 3.894 and 4.400 respectively, chloroform).

Anal. Calc'd for $C_{33}H_{46}O_4N_4$: C, 70.43; H, 8.24; N, 9.96.

Found: C, 70.44; H, 8.03; N, 9.99.

(*b*) *From* $\Delta^{2,4}$*-cholestadiene-6-one* (IV). The dinitrophenylhydrazone obtained from 3 mg. of the diene IV was purified by chromatography and recrystallized from ethanol; m.p. 187–191°. The u.v. spectrum was identical with that of the dinitrophenylhydrazone prepared according to (*a*).

Cholestane-5α,6β-diol (VIII). To a suspension of 1.915 g. of Δ^5-cholestene (VII) in 20 cc. of 88% formic acid two cc. of 30% hydrogen peroxide was added. The mixture was shaken at room temperature overnight; some wax-like material remained undissolved. After addition of 200 cc. of water the mixture was extracted with ether and the ether solutions were washed with water, 2 *N* sodium hydroxide solution and water, dried and evaporated. The residue was refluxed with 22 cc. of 2.5% methanolic potassium hydroxide solution, and the solution acidified, concentrated *in vacuo*, diluted with water and extracted with ether. After washing with sodium carbonate solution and water the ether solution was dried and evaporated. The residue (2.00 g.) was chromatographed on 60 g. of aluminum oxide. The first fraction, eluted with hexane, weighed 430 mg. and gave after recrystallization from ether-ethanol 381 mg. of starting material, m.p. 91–93°. The diol was eluted with benzene, 1.1 g. From dil. ethanol, there were obtained crystals which melted at 50° and after solidification at 123.5–126.5° [lit. (11), m.p. 125.5°]. After drying in a high vacuum only the higher m.p. was observed; infrared band at 2.70 μ. The diol VIII can be sublimed in a high vacuum at 160°. When recrystallized from dil. acetone the needles thus obtained melted at 116–118° and after solidification at 125–127°. This diol failed to give a 3,5-dinitrobenzoate, when refluxed for 10 min. with dinitrobenzoyl chloride in benzene-pyridine solution.

The *6-monoacetate* was prepared as follows: 100 mg. of diol VIII was acetylated with 2 cc. of pyridine and 1 cc. of acetic anhydride overnight at room temperature. The crude acetate was chromatographed on 3.5 g. of aluminum oxide and eluted from the column with mixtures of hexane and benzene containing from 40–70% benzene. After sublimation in a high vacuum at 150° and recrystallization from methanol it had m.p. 112–114°; infrared bands at 2.69, 5.72 and 8.02 μ.

Anal. Calc'd for $C_{29}H_{50}O_3$: C, 77.97; H, 11.28.

Found: C, 78.00; H, 11.22.

Cholestane-5α-ol-6-one (V). (*a*) *From cholestane-5α,6β-diol* (VIII). A suspension of 220 mg. of diol VIII in 4.5 cc. of ether, 0.75 cc. of water, and 0.75 cc. of methanol was shaken in a separatory funnel after addition of 120 mg. of N-bromosuccinimide. After five minutes 4 cc. of water and more ether were added to the orange-yellow solution, and the ether layer was washed with bisulfite solution, 2 *N* sodium hydroxide solution and water, dried and evaporated. On standing with pentane the residue crystallized in big prisms which were sublimed in a high vacuum at 150° and recrystallized from dil. acetone; m.p. 153–154°; $[\alpha]_D^{23}$ − 44.5° ± 3° (abs. ethanol), infrared bands at 2.70 and 5.80 μ.

Anal. Calc'd for $C_{27}H_{46}O_2$: C, 80.54; H, 11.52.

Found: C, 80.45: H, 11.58.

The same substance was obtained when the diol VIII was oxidized with chromium trioxide.

(*b*) *From* Δ^2*-cholestene-5α-ol-6-one* (III). A solution of 106 mg. of the unsaturated ketol III in 6 cc. of abs. ethanol was hydrogenated with 50 mg. of 5% palladium-on-calcium carbonate catalyst. The hydrogenation was complete after 2.5 hours. The filtrate was evaporated *in vacuo* and the residue was recrystallized from ether-pentane, prisms, m.p. 152.5–154°. No depression was observed in mixture with a specimen prepared according to (*a*).

The *oxime* was recrystallized from ether-pentane and from methanol and melted at 195–197°. When chromatographed on aluminum oxide, it was eluted from the column with mixtures of benzene and ether.

Anal. Calc'd for $C_{27}H_{47}O_2N$: C, 77.64; H, 11.34; N, 3.35.
Found: C, 77.69; H, 11.40; N, 3.39.

The *semicarbazone* prepared according to method II was chromatographed on aluminum oxide and eluted with ether and chloroform. After recrystallization from methanol it melted in a wide range between 226 and 232°; u.v. maximum at 228 mμ (log ϵ 4.114, ethanol).

Anal. Calc'd for $C_{28}H_{49}O_2N_3$: C, 73.15; H, 10.74; N, 9.14.
Found: C, 73.63; H, 10.57; N, 8.92.

When method I was used for the preparation of the semicarbazone, only 25% of the ketone was found to have reacted.

The *acetate* was prepared as follows: A solution of 25 mg. of V in 2 cc. of acetic anhydride was refluxed for 24 hours. The crude reaction product weighed 34 mg. and was chromatographed on 1.2 g. of aluminum oxide. One fraction eluted with hexane-benzene 4:1 was a mixture of an acetate and an α,β-unsaturated ketone (u.v. maximum at 240 mμ, presumably VI). The acetate was eluted with hexane-benzene 7:3, 3:2 and 1:1. It was sublimed in a high vacuum at 160° and recrystallized from methanol; fine needles, m.p. 124–125.5°; infrared bands at 5.70, 5.78 and 8.05 μ.

Anal. Calc'd for $C_{29}H_{48}O_3$: C, 78.32; H, 10.88.
Found: C, 77.62; H, 10.71.

The sample for analysis was probably not completely dry.

Some unchanged starting material was eluted from the column with hexane-benzene 2:3 and 1:4.

Δ_4-*Cholestene-6-one* (VI). To a solution of 462 mg. of V in 8 cc. of abs. pyridine, 0.2 cc. of thionyl chloride was added with shaking. The mixture was refluxed for ten minutes, the pyridine evaporated *in vacuo*, and the residue taken up in ether. The ether was washed with hydrochloric acid and water, dried and evaporated. Chromatography on 15 g. of aluminum oxide and elution with hexane and mixtures of hexane and benzene up to 20% benzene yielded the unsaturated ketone VI, which was sublimed in a high vacuum at 150° and recrystallized from acetone. Needles were obtained, m.p. 108–109°; u.v. maximum at 243 mμ (log ϵ 3.804, ethanol); infrared band at 5.89 μ; $[\alpha]_D^{25}$ + 36.2° ± 3° (ethanol).

Anal. Calc'd for $C_{27}H_{44}O$: C, 84.31; H, 11.53.
Found: C, 84.17; H, 11.61.

Some unchanged starting material was eluted from the column with hexane-benzene 1:1, 2:3 and benzene.

The *oxime* of the unsaturated ketone VI failed to crystallize. After chromatography on aluminum oxide very few crystals with m.p. between 150° and 165° were obtained which could not be recrystallized in a satisfactory manner.

The *semicarbazone* prepared according to method II was chromatographed on aluminum oxide, but could not be purified completely. After recrystallization from methanol it melted in a wide range between 100° and 165°; u.v. maximum at 253 mμ (log ϵ 3.852, ethanol).

Anal. Calc'd for $C_{28}H_{47}N_3O$: N, 9.52. Found: N, 8.73.

Δ^4-*Cholestene-6-one 2,4-dinitrophenylhydrazone.* (*a*) *From* Δ^4-*cholestene-6-one* (VI). A solution of ten mg. of the unsaturated ketone VI and eight mg. of dinitrophenylhydrazine in two cc. of abs. ethanol and two drops of conc. hydrochloric acid was refluxed for five minutes. The orange precipitate was filtered, washed with ethanol, dried and chromatographed on 500 mg. of aluminum oxide. It was eluted from the column with hexane-benzene 1:1 and recrystallized from ethanol. The orange prisms had m.p. 162–164°; u.v. maxima at 257 and 377 mμ (log ϵ 4.090 and 4.396 respectively, chloroform).

Anal. Calc'd for $C_{33}H_{48}N_4O_4$: C, 70.18; H, 8.57; N, 9.92.
Found: C, 70.14; H, 8.52; N, 9.88.

(*b*) *From cholestane-5α-ol-6-one* (V). The dinitrophenylhydrazone was prepared from 26 mg. of V and 21 mg. of dinitrophenylhydrazine. It was chromatographed on aluminum oxide and eluted with large amounts of hexane-benzene 9:1. After recrystallization from ethanol it showed m.p. 163–165° and u.v. maxima at 255 and 375 mμ (log ϵ 4.137 and 4.384 respectively, chloroform). No depression was observed in mixture with a specimen prepared according to (*a*).

Anal. Calc'd for $C_{33}H_{43}O_4N_4$: N, 9.92. Found: N, 9.46.

Cholestane-6-one (IX). A solution of 50 mg. of Δ^4-cholestene-6-one (VI) in 3.5 cc. of abs. ethanol was hydrogenated with 25 mg. of 5% palladium-on-calcium carbonate catalyst. The hydrogenation was complete after two hours. The filtrate was evaporated to dryness, the residue chromatographed on aluminum oxide, and the column washed exhaustively with hexane. The first five fractions were combined and recrystallized from ethanol, giving crystals of m.p. 99–101°; infrared band at 5.79 μ.

The *oxime* was chromatographed, eluted with hexane-benzene 1:4, and benzene, and recrystallized from ethanol; needles, m.p. 194–196° [lit. (19), m.p. 195°].

Δ^4-Cholestene-3β-ol-6-one acetate semicarbazone. This semicarbazone was prepared according to method II and chromatographed on aluminum oxide. It was eluted with benzene-ether 2:3 and with ether. The crystals obtained from methanol melted in a wide range between 141° and 196°; u.v. maximum at 258 mμ (log ϵ 3.999, ethanol); infrared bands at 5.71, 5.87 and 8.08 μ.

Anal. Calc'd for $C_{30}H_{49}N_3O_3$: C, 72.10; H, 9.88; N, 8.41.
Found: C, 72.06; H, 9.88; N, 8.33.

Methyl 3α-tosyloxy-12-ketocholanate.[3] To a solution of 56 mg. of methyl 3α-tosyloxy-12α-hydroxycholanate in 0.5 cc. of glacial acetic acid 0.5 cc. of 2% solution of chromium trioxide in glacial acetic acid was added in five portions over a period of 2.5 hours. The oxidation product was recrystallized from acetone-hexane and gave elongated prisms, m.p. 167.5–168.5°; infrared bands at 5.71, 5.82, 8.40 and 8.49 μ.

Anal. Calc'd for $C_{32}H_{46}O_6S$: C, 68.78; H, 8.30; S, 5.74.
Found: C, 68.39; H, 8.34; S, 5.44.

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SUMMARY

$\Delta^{2,4}$-Cholestadiene-6-one and Δ^4-cholestene-6-one have been prepared by a series of reactions from cholesterol. Δ^4-cholestene-6-one was also obtained from Δ^5-cholestene.

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REFERENCES

(1) LADENBURG, CHAKRAVORTY, AND WALLIS, *J. Am. Chem. Soc.*, **61**, 3483 (1939).
(2) BLUNSCHY, HARDEGGER, AND SIMON, *Helv. Chim. Acta*, **29**, 199 (1946).
(3) RUZICKA, GROB, AND RASCHKA, *Helv. Chim. Acta*, **23**, 1518 (1940).
(4) FIESER AND RAJAGOPALAN, *J. Am. Chem. Soc.*, **71**, 3938 (1949).
(5) CLEMO, KELLER, AND WEISS, *J. Chem. Soc.*, 3470 (1950).
(6) FREUDENBERG AND HESS, *Ann.*, **448**, 121 (1926).
(7) BARNETT AND REICHSTEIN, *Helv. Chim. Acta*, **21**, 926 (1938).
(8) EHRENSTEIN AND NEUMANN, *J. Org. Chem.*, **16**, 335 (1951);
EHRENSTEIN, BARBER, AND GORDON, *J. Org. Chem.*, **16**, 349 (1951).
(9) MATTOX AND KENDALL, *J. Am. Chem. Soc.*, **70**, 882 (1948).
(10) ROSS, *J. Chem. Soc.*, 737 (1946).
(11) RUZICKA AND THOMANN, *Helv. Chim. Acta*, **16**, 216 (1933).
(12) PRELOG AND TAGMANN, *Helv. Chim. Acta*, **27**, 1867 (1944).
(13) HEILBRON, JONES, AND SPRING, *J. Chem. Soc.*, 801 (1937).
(14) JONES, WILKINSON, AND KERLOGUE, *J. Chem. Soc.*, 391 (1942).
(15) MENSCHICK, PAGE, AND BOSSERT, *Ann.*, **495**, 225 (1932).
(16) BUTENANDT, MAMOLI, DANNENBERG, MASCH, AND PALAND, *Ber.*, **72**, 1617 (1939).
(17) DJERASSI AND RYAN, *J. Am. Chem. Soc.*, **71**, 1000 (1949).
(18) INHOFFEN AND HUANG-MINLON, *Ber.*, **71**, 1720 (1938).
(19) WINDAUS AND DALMER, *Ber.*, **52**, 162 (1919).

[3] This experiment was carried out by Dr. G. Fujimoto.