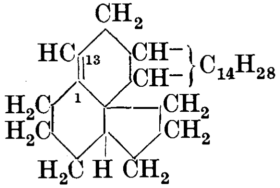
CCLXXXIX.—*Studies in the Sterol Group. Part V. The Constitution of Cholesterilene.*

By ATUL CHANDRA BOSE and WILLIAM DORAN.

IT was pointed out by Heilbron, Morton, and Sexton (J., 1928, 47) that the absorption spectrum of cholesterilene closely resembles that of ergosterol, and it was inferred that, of the three double bonds in the latter compound, two occupy the same positions as in cholesterilene. This inference has since been supported by Rosenheim (*Biochem. J.*, 1929, **23**, 50). It was also shown (Heilbron and Sexton, J., 1928, 347) that one of the ethenoid linkages in cholesterilene must occupy the same position as in ψ-cholestene (I) (see also Rosenheim, *loc. cit.*).

(I.)



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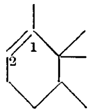


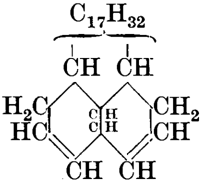
A review of the literature of cholesterilene reveals, as shown in Table I, a considerable divergence between the data recorded by various investigators. (The product obtained in the experiments of Tschugaev and Fomin consisted of 66% of α- and 33% of β-cholesterylene.)

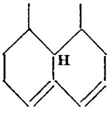
TABLE I.

		α-Cholesterilene.	
Author.	Reaction.	M. p.	$[\alpha]_D$.
Mauthner & Suida (*Monatsh.*, 1896, **17**, 29).	Cholesterol + anhydrous $CuSO_4$ at 200°.	78°	− 81·63°
Mauthner & Suida (*Monatsh.*, 1903, **24**, 660).	Cholesteryl chloride heated with lime.	79	− 61·55
,, ,, ,,	Cholesteryl chloride heated with quinoline.	77	− 86·09
Bloch (*Bull. Soc. chim.*, 1904, **31**, 73).	Cholesteryl phenylurethane heated.	75	—
Tschugaev & Gasteff (*Ber.*, 1909, **42**, 4631).	Methyl cholesterylxanthate heated.	77	−107
Tschugaev & Fomin (*Annalen*, 1910, **375**, 288).	Methyl cholesterylxanthate heated in a vacuum at 200°.	{77 {59	−109·3 (α) − 76·68 (β)
Steinkopf & Blümner (*J. pr. Chem.*, 1911, **84**, 466).	Cholesteryl chloride + potassium cholesterolate heated with Zn dust.	80	−116·2
Steinkopf (*J. pr. Chem.*, 1920, **100**, 70).	Cholesterol + kieselguhr heated in a vacuum at 300°.	79	− 46·6
,, ,, ,,	Cholesteryl chloride distilled at 200°.	79	− 47·04
Fantl (*Monatsh.*, 1926, **47**, 251).	Cholesterol + Zn dust distilled at 10 mm. pressure.	75	− 4·49

Some of these discrepancies may, as suggested by Mauthner and Suida (*loc. cit.*), be explained by stereochemical considerations such as racemisation of one or more of the asymmetric carbon atoms in the molecule, and yet a further complication arises through the well-known tendency of compounds in the sterol group to form " mixed crystals " containing in various proportions isomerides of different rotatory powers. The chemistry of cholesterilene also becomes involved for other reasons : for instance, it would appear that the so-called α- and β-cholesterylenes of Tschugaev and Fomin (*loc. cit.*), to which the formulæ (II) and (III) were ascribed, are structurally different from the cholesterilene of Mauthner and Suida, since they are stated to yield only cholestane upon hydrogenation, whereas Windaus and Seng (*Z. physiol. Chem.*, 1921, **117**, 158) have shown that cholesterilene prepared according to Mauthner and Suida's copper sulphate method yields both cholestane and ψ-cholestane.

(II.)





(III.)

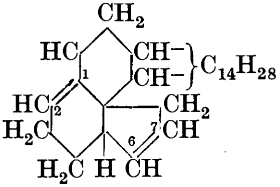
Apart, therefore, from any question of stereoisomerism, the evidence to be adduced from the foregoing suggests the existence of three structurally different cholesterilenes.

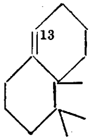
Before accepting these conclusions, the authors deemed it advisable to re-examine the whole question, particularly from the standpoint of hydrogenation. The experiments of Tschugaev and Fomin have been repeated and extended and it has been found that the so-called " β-cholesterylene " is not a definite compound and when care is taken to eliminate every trace of sulphur from the reaction product, only the hydrocarbon of m. p. 79—80° can be isolated.

The experiments described in this paper also place beyond all doubt the identity of the hydrocarbons prepared by Mauthner's and by Tschugaev's process, for when hydrogenated under precisely similar conditions each yielded a mixture of cholestane and ψ-cholestane. The name " cholesterylene " should therefore be deleted from the literature, and since it has been shown that only one hydrocarbon $C_{27}H_{44}$ (m. p. 79—80°, lævorotatory) containing two ethenoid linkages has up to the present been obtained from cholesterol by any of the simple reactions outlined above (Table I), this hydrocarbon should be designated *cholesterilene* without qualification.

Assuming, as seems probable, that during the reactions involved no change takes place in the position of the double bond originally present at 6 : 7, there are two possible structures for cholesterilene (IV and V), either of which is in agreement with the production, on hydrogenation, of the stereoisomeric saturated hydrocarbons cholestane and ψ-cholestane.

(IV.)





(V.)

EXPERIMENTAL.

Action of Dehydrating Agents upon Cholesterol.—Attempts were made to ascertain whether reagents other than anhydrous copper sulphate could be successfully employed for the preparation of cholesterilene. Anhydrous sodium sulphate did not appear to dehydrate cholesterol after 15 minutes' heating at 210—215°, and anhydrous potassium sulphate under the same conditions afforded dicholesteryl ether in high yield.

Cholesterilene was satisfactorily prepared by the method of Mauthner and Suida (*Sitzungsber. Akad. Wiss. Wien*, 1896, **104**, 822), but a cleaner product was obtained by heating the reactants at 200° under reduced pressure (5 mm.).

Methyl Cholesterylxanthate.—Following the directions of Tschugaev and Gasteff (*loc. cit.*), low yields of ester (5—10%) were repeatedly obtained and cholesterol was always recovered. The initial reaction between potassium amyloxide and cholesterol was defective whether heating was carried out for long or short periods. Methyl amyl ether was invariably detected in the course of working up the final product, pointing to methylation of potassium amyloxide in the second stage of the process. The following technique avoids the use of amyl alcohol, considerably shortens the duration of the reaction, and ensures high yields (90%) of pure colourless ester.

A solution of cholesterol (20 g.) in hot benzene (30 g.) was added in one portion with shaking to "emulsified" potassium (1·9 g.) in sodium-dried benzene (30 g.). The mixture was heated on the water-bath for 15 minutes to complete the formation of potassium cholesteryloxide, and cooled quickly to 40°, dry carbon disulphide (15 g.) was slowly added with shaking, and the pink reaction mass refluxed for 3 hours with methyl iodide (20 g.). Ether (200 c.c.) was added to the cooled product, the deposited potassium iodide washed away with water, and the dried ethereal layer distilled under reduced pressure to half its bulk; on addition of absolute alcohol, colourless needles (16 g.) of the ester, m. p. 127°, $[\alpha]_D^{25^\circ}$ $-51{\cdot}1^\circ$ in toluene ($c = 6{\cdot}4$), were deposited (Found: S, 13·2. Calc. for $C_{29}H_{48}OS_2$: S, 13·4%).

Preparation of "β-Cholesterylene."—When methyl cholesterylxanthate was heated at about 200° in a vacuum (compare Tschugaev and Fomin, *loc. cit.*), the product obtained contained both α- and β-cholesterylene. The proportion of the supposed β-isomeride varied with the temperature of decomposition: at 155°, equal amounts of the two isomerides were obtained; at 185°, the ratio of α to β was 2 : 1, and at 205° it became 3 : 1.

When the β-compound, m. p. 58°, was kept at 200° under reduced pressure for ½ hour, or heated with animal charcoal in benzene, it was apparently partly converted into the higher-melting α-cholesterylene.

Removal of Sulphur from "β-Cholesterylene."—Tschugaev and Fomin (*loc. cit.*) used alkali-containing alcohol to precipitate the hydrocarbons from an ethereal solution of the reaction mixture, presumably in order to remove sulphur-containing impurities. We have found that this treatment is of primary importance and that only after repeated precipitation with concentrated alcoholic potash is all sulphur removed from the more soluble hydrocarbon fractions (so-called β-cholesterylene).

(i) Supposed pure β-cholesterylene, m. p. 59° [Found: S (micro), 1·2%] was dissolved in the minimum quantity of ether and treated with concentrated alcoholic potash solution. Two crops of needles of α-cholesterylene, m. p. 78°, were separated and the mother-liquor gave a small amount of low-melting material.

(ii) β-Cholesterylene, m. p. 59°, was dissolved in heptane and shaken with sodium–potassium liquid amalgam. Rapid tarnishing of the metal was observed. When, after 15 minutes, the heptane layer was filtered and allowed to evaporate, sulphur-free α-cholesterylene (needles, m. p. 78°) was obtained; no β-cholesterylene remained in the mother-liquor.

Catalytic Hydrogenation of Cholesterilene and α-Cholesterylene.—Cholesterilene (5 g.; m. p. 79°) in ether (80 c.c.) in the presence of palladium-black (0·6 g.) was hydrogenated for 27 hours under 1½ atmospheres pressure. On crystallisation, cholestane appeared first as plates, m. p. 80° (Found: C, 87·0; H, 13·1. Calc. for $C_{27}H_{48}$: C, 87·1; H, 12·9%), and then, after successive cropping, long needles of ψ-cholestane, m. p. 70—71°, separated (Found: C, 86·9; H, 12·9%).

Under precisely similar conditions, α-cholesterylene, m. p. 79·5°, prepared by the modification of Tschugaev's process here given, yielded a mixture of cholestane, m. p. 79—80° (Found : C, 87·2; H, 13·2%), and ψ-cholestane, m. p. 70—71° (Found : C, 87·2; H, 12·8%).

The identity of the saturated hydrocarbons in each case was confirmed by the fact that they showed no alteration of melting point in admixture respectively with cholestane and ψ-cholestane obtained by hydrogenation of cholestene and ψ-cholestene, both of authentic origin.

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THE UNIVERSITY OF LIVERPOOL. [*Received, July 9th*, 1929.]