4 : 4-*Dimethyl Steroids. Part I.* 4 : 4-*Dimethylcalciferol.*

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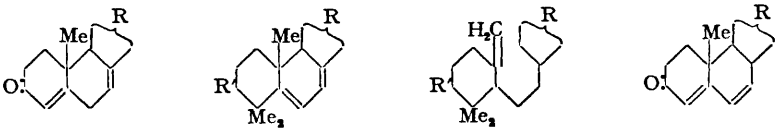
Ergosterol has been converted into 4 : 4-dimethylergosterol (II; R′ = β-OH) and thence by irradiation into 4 : 4-dimethylcalciferol (III; R′ = β-OH).

Biosynthesis of cholesterol in the animal organism appears to proceed from acetate, which is then converted into branched-chain acids such as β-methylcrotonic acid and thence into linked *iso*prenoid precursors which may be of the squalene type (see recent reviews: Liebermann and Teich, *Pharm. Reviews*, 1953, **5**, 285; Ruzicka, *Experientia*, 1953, **9**, 357. Also Popják, *Arch. Biochem. Biophys.*, 1954, **48**, 102; Rabinovitch and Gurin, *J. Amer. Chem. Soc.*, 1954, **76**, 5168; Eidinoff, Rosenfeld, Knoll, Marano, and Hellman, *J. Clin. Invest.*, 1954, **33**, 333; Bloch, Clark, and Isaac, *J. Biol. Chem.*, 1954, **211**, 687). 4 : 4-Dimethyl-steroids may consequently be implicated in steroid biogenesis (Woodward and Bloch, *J. Amer. Chem. Soc.*, 1953, **75**, 2023; Ruzicka, *loc. cit.*; Clayton and Bloch, *Fed. Proc.*, 1955, **14**, 194) and are therefore worthy of study.

4 : 4-Dimethylcholesterol, required by Woodward, Packett, Barton, Ives, and Kelley (*J. Amer. Chem. Soc.*, 1954, **76**, 2852) for conversion into lanosterol, was prepared by them by dimethylation of cholestenone followed by reduction of the resulting 4 : 4-dimethyl-

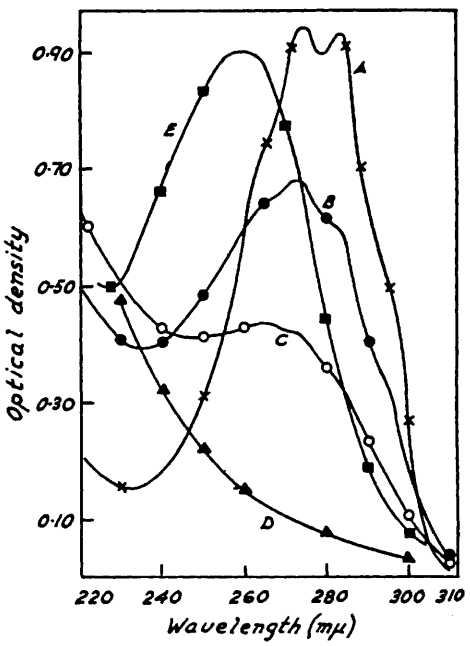


(I) (II) (III) (IV)

(R = $C_{17}H_{30}$)

cholest-5-en-3-one. Extension of the methylation procedure to the ergosterol series is now reported.

Ergosterol was converted into *iso*ergosterone (IV) and ergosterone (I) as described by Johnson, Newbold, and Spring (*J.*, 1954, 1302). Attempts to methylate the ketone (IV)



Ultra-violet absorption spectra of 4 : 4-*dimethylergosterol* (4·0 × 10^{-3} *g./*100 *ml. of ethanol*): *A*, ×, *original*; *B*, ●, *after 2-min. irradiation*; *C*, ○, *after 5-min. irradiation*; *D*, ▲, *after 15-min. irradiation*; (*E*), ■, 4 : 4-*dimethylcalciferol* (1·84 × 10^{-3} *g./*100 *ml. of ethanol*).

in dry *tert.*-butanol with potassium *tert.*-butoxide (3 mols.) and methyl iodide (6 mols.) (Woodward *et al.*, *loc. cit.*; cf. Conia, *Bull. Soc. chim. France*, 1954, 943) yielded mostly unchanged material. Ergosterone (I), in contrast, passed smoothly under these conditions into crystalline 4 : 4-dimethylergosta-5 : 7 : 22-trien-3-one (II; R′ = :O). This structure is supported, not only by the mode of formation, but also by the ultraviolet absorption which closely resembles that of the parent ergosterol. Reduction of the product (II; R′ = :O) with lithium aluminium hydride gave in high yield a homogeneous alcohol, considered to be 4 : 4-dimethylergosterol (II; R′ = β-OH) as its ultraviolet absorption is almost identical with that of the ketone (II; R′ = :O). Its alternative formulation as 4 : 4-dimethyl*epi*ergosterol (II; R′ = α-OH) is rendered unlikely as reduction of similar 4 : 4-dimethyl-3-ketones such as 4 : 4-dimethylcholest-5-en-3-one (Woodward *et al.*, *loc. cit.*), lanostadienone, and euphadienone (Dr. J. F. McGhie, personal communication) with lithium aluminium hydride leads exclusively to 3β-hydroxy-compounds (see also Klyne, " Progress in Stereochemistry," Butterworths Scientific Publ., 1954, Vol. I, p. 74).

Parallel study of the spectral changes undergone by ergosterol and by the alcohol (II; R′ = β-OH) in ethanolic solution on irradiation with ultraviolet light revealed closely similar behaviour (cf. Figure, curves *A* to *D* obtained in small-scale experiments in which

the irradiation was carried out in 5-mm. bore silica tubes with data, for example, in Morton, Heilbron, and Kamm, *J.*, 1927, 2002). Extension of these experiments to the preparative scale led to the isolation as the 3 : 5-dinitrobenzoates of calciferol from ergosterol and of a photochemical transformation product from the alcohol (II; R′ = β-OH) to which we assign the constitution of 4 : 4-dimethylcalciferol (III; R′ = β-OH) on the basis of its ultraviolet absorption.

Oxidation of dimethylcalciferol (III; R′ = β-OH) by the method of Heilbron, Jones, Samant, and Spring (*J.*, 1936, 905) gave a viscous oil with aldehydic properties (Schiff's reagent). Treatment with semicarbazide gave a small quantity of semicarbazone which was probably derived from the $C_{19}H_{33}O$ ketone of Windaus and Thiele (*Annalen*, 1936, **521**, 160), but paucity of material prevented full identification. We also examined the irradiation of the ketone (II; R′ = :O), but obtained the photochemical product only as its semicarbazone, to which we tentatively assign the formulation of 4 : 4-dimethylcalciferone semicarbazone on the basis of its ultraviolet absorption. Cleavage of the semicarbazone by Hershberg's method (*J. Org. Chem.*, 1948, **13**, 542) gave a viscous oil; its ultraviolet absorption resembled that of 4 : 4-dimethylcalciferol (III; R′ = β-OH).

Experimental

Optical rotations were measured in $CHCl_3$ in a 1-dm. tube, unless otherwise stated. Absorption spectra, measured in EtOH, were kindly determined by Mr. M. T. Davies, B.Sc.

4 : 4-*Dimethylergosta*-5 : 7 : 22-*trien*-3-*one* (II; R′ = :O).—Ergosterone (28·5 g.) was added to a solution of potassium (10·9 g.) in *tert.*-butanol (350 ml.; previously distilled over potassium *tert.*-butoxide) under nitrogen, and the mixture stirred and warmed gently until dissolution was complete. After addition of methyl iodide (35 ml.) to the ice-cooled mixture, stirring under nitrogen was continued for 2½ hr. at room temperature, then most of the solvent was removed under reduced pressure. The residue was treated with water, and the product isolated with ether. 4 : 4-*Dimethylergosta*-5 : 7 : 22-*trien*-3-*one* (74%) formed plates (from acetone), m. p. 167—168°, $[\alpha]_D^{24}$ −37° (*c*, 0·75) (Found: C, 85·3; H, 11·0. $C_{30}H_{46}O$ requires C, 85·2; H, 11·0%). Light absorption: $\lambda_{max.}$ 274 and 283 mμ (log ε 4·00, 4·01); infl. 265 and 292 mμ. The *semicarbazone* formed plates, m. p. 259°, $[\alpha]_D^{23}$ −59° (*c*, 1·0) (Found: C, 76·5; H, 10·1; N, 8·8. $C_{31}H_{49}ON_3$ requires C, 77·6; H, 10·3; N, 8·8%), after crystallisation from chloroform–methanol. Light absorption: $\lambda_{max.}$ 228, 273, and 283 mμ (log ε 4·13, 4·09, 4·09); infl. 266 and 292 mμ.

Attempted Methylation of iso*Ergosterone* (IV).—The ketone (1·53 g.) in a solution of potassium (0·6 g.) in *tert.*-butanol was treated, under nitrogen, with methyl iodide (2 ml.). After 2½ hr. the product was isolated and chromatographed in benzene on a column of acid-washed alumina (40 g.). Early fractions provided unchanged material. Later fractions afforded a *substance* (20 mg.), rhombs, m. p. 124° (Found: C, 84·7; H, 11·0%), after crystallisation from acetone–methanol. Light absorption: single peak with $\lambda_{max.}$ 283 mμ.

4 : 4-*Dimethylergosterol* (II; R′ = β-OH).—The ketone (II; R′ = :O) (9·2 g.) in ether (1·4 l.) was added during 90 min. to an ice-cooled suspension of lithium aluminium hydride (6 g.) in ether (500 ml.). The mixture was stirred for 15 min. at room temperature and then heated under reflux for 2 hr. The product, isolated in the usual way and purified from ethanol, gave 4 : 4-*dimethylergosterol* (89%), plates, m. p. 181°, $[\alpha]_D^{25}$ −171·5° (*c*, 0·76) (Found: C, 85·1; H, 11·5. $C_{30}H_{48}O$ requires C, 84·8; H, 11·4%). Light absorption: $\lambda_{max.}$ 274 and 283 mμ (log ε 4·00, 4·00); infl. 265 and 292 mμ. The *acetate* formed plates, m. p. 173—174°, $[\alpha]_D^{25}$ −108° (*c*, 0·64) (Found: C, 81·7; H, 10·8. $C_{32}H_{50}O_2$ requires C, 82·3; H, 10·8%), after crystallisation from ethanol. The 3 : 5-*dinitrobenzoate* separated from acetone in plates, m. p. 196—199°, $[\alpha]_D^{23}$ −47° (*c*, 1·11) (Found: C, 71·7; H, 7·9; N, 4·5. $C_{37}H_{50}O_6N_2$ requires C, 71·8; H, 8·1; N, 4·5%). Light absorption: $\lambda_{max.}$ 262, 273, and 282 mμ (log ε 4·18, 4·18, 4·13); infl. 292 mμ (cf. spectrum of ergosteryl 3 : 5-dinitrobenzoate, Huber, Ewing, and Kriger, *J. Amer. Chem. Soc.*, 1945, **67**, 609).

4 : 4-*Dimethylcalciferyl* 3 : 5-*Dinitrobenzoate* [III; R′ = β-$C_6H_3(NO_2)_2$·CO_2].—A suspension of the alcohol (II; R′ = β-OH) (33·5 g.) in ethanol (6·5 l. of 96%) under carbon dioxide was irradiated for 1 hr. with a mercury-vapour lamp. The solution was concentrated under reduced pressure to *ca.* 300 ml.; unchanged material separated and was removed by filtration. Complete removal of solvent gave a yellow resin (17·9 g.), which was treated in pyridine (36 g.) at 0° with 3 : 5-dinitrobenzoyl chloride (15·2 g.), stirred vigorously at room temperature for 20 min., and digested with water. Fractionation of this material from acetone led, after removal of further

quantities of unchanged material, to 4 : 4-*dimethylcalciferyl* 3 : 5-*dinitrobenzoate* (5·7 g.), pale yellow prisms, m. p. 132—135°, $[\alpha]_D^{23}$ +99° (*c*, 0·84) (Found : C, 71·7; H, 8·0; N, 4·5. $C_{37}H_{50}O_6N_2$ requires C, 71·8; H, 8·1; N, 4·5%). The spectrum showed an absorption increasing steadily in intensity from 280 to 220 mμ with just discernible shoulders at 235 and 260 mμ (cf. calciferyl 3 : 5-dinitrobenzoate, Huber *et al.*, *loc. cit.*).

4 : 4-*Dimethylcalciferol* (III; R′ = β-OH).—The foregoing compound (5 g.) in benzene (10 ml.) was treated with methanolic sodium hydroxide (8·3 ml. of a solution prepared from 700 mg. of sodium and 23 ml. of methanol, diluted with 1·5 ml. of water). The mixture was stirred for 5 min., then filtered, and the sodium 3 : 5-dinitrobenzoate washed once with benzene (2 ml.). The filtrate was washed with water and dried, the solvent removed, and the residue crystallised from methanol. 4 : 4-*Dimethylcalciferol* (2 g.) formed flat needles, m. p. 122—124°, $[\alpha]_D^{23}$ +85° (*c*, 1·01 in EtOH) (Found : C, 84·4; H, 11·4. $C_{30}H_{48}O$ requires C, 84·8; H, 11·4%). Light absorption : $\lambda_{max.}$ 260 mμ (log ε 4·32).

The foregoing product (550 mg.) in glacial acetic acid (30 ml.) was treated dropwise with stirring during 1 hr. with chromium trioxide (250 mg.) in 75% acetic acid (1 ml.). After 3 hr. at room temperature the mixture was diluted with water, and the product isolated with ether. The neutral fraction, a yellow viscous oil (530 mg.), was treated with semicarbazide acetate in ethanol, yielding a very small quantity of crystals, m. p. 199°, $\lambda_{max.}$ 231 mμ, intense absorption (cf. Windaus and Thiele, *loc. cit.*).

Irradiation of 4 : 4-*Dimethylergosta*-5 : 7 : 22-*trien*-3-*one* (II; R′ = :O).—The ketone (II; R′ = :O) (30 g.), suspended in ethanol (6 l. of 96%) and under carbon dioxide, was irradiated for 1 hr. with a mercury-vapour lamp. The solution was concentrated in stages under reduced pressure to *ca.* 45 ml., unchanged material which separated being removed by filtration. Removal of solvent left a yellow brown oil (6·9 g.) which was treated with semicarbazide acetate in ethanol under reflux for 1½ hr. Fractionation of the product from ethanol gave 4 : 4-*dimethylcalciferone semicarbazone* (420 mg.), pale yellow plates, m. p. 205°, $[\alpha]_D^{20}$ +29° (*c*, 0·72) (Found : C, 76·8; H, 10·1; N, 8·9. $C_{31}H_{49}ON_3$ requires C, 77·6; H, 10·3; N, 8·8%). Light absorption : $\lambda_{max.}$ 235 and 270 mμ (log ε 4·24 and 4·23).

The foregoing compound (400 mg.) in warm glacial acetic acid (4 ml.) was treated with pyruvic acid (110 mg.) and anhydrous sodium acetate (144 mg.) in acetic acid (1 ml.); after 10 min. under reflux the mixture was diluted with water to turbidity, and refluxing continued for a further 50 min. The product, isolated by precipitation with water (12 ml.) and decantation, failed to crystallise : it had $\lambda_{max.}$ 269—270 mμ and formed a yellow dinitrophenylhydrazone.

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