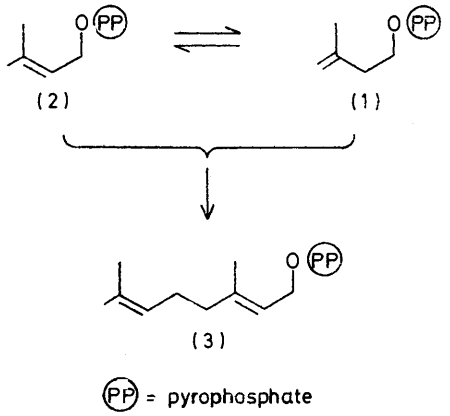
On Stereochemical Preference in the S_E2' Reaction

By Ian M. Cunningham and Karl H. Overton,* Department of Chemistry, University of Glasgow, Glasgow G12 8QQ

Attempts are described to detect stereochemical preference during an S_E2' reaction in a synthetic model system. 7β-Deuterio-4,5-secocholest-5-en-4-al ethylene acetal (20d) cyclised with Lewis acid to give 7-deuterio-4α-(2-hydroxyethoxy)-5β-cholest-6-ene (23c) with complete retention of deuterium. By contrast, 7α-deuterio-4,5-secocholest-5-en-4-yl *p*-bromobenzenesulphonate (29c) cyclised in 2,2,2-trifluoroethanol to 5β-cholest-6-ene (31) with total loss of deuterium. In both cases there is a *syn*-relationship between the newly formed C–C bond and the allylic hydrogen atom that is lost. The implications of these findings for the S_E2' reaction are discussed.

THE assembly of polyisoprenoid chains is a biosynthetic process of ancient origin [1] and apparently universal occurrence among living organisms. From it stem the host of naturally occurring terpenoids and steroids which sustain a multitude of essential functions in biological systems.

There are two fundamental steps in isoprenoid biosynthesis: (*a*) isomerisation of isopentenyl pyrophosphate (1) to dimethylallyl pyrophosphate (2), mediated by isopentenyl pyrophosphate isomerase; [2-4] and (*b*) chain extension, mediated by a prenyl transferase,[4-6] and exemplified by the coupling of isopentenyl and dimethylallyl pyrophosphates leading to geranyl pyrophosphate (3). Both may be formally regarded as

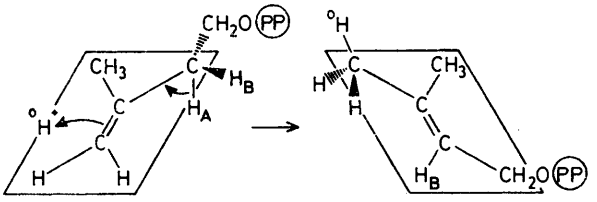


bimolecular electrophilic substitutions with allylic displacement (S_E2').

Cornforth, Popjak, and their colleagues have estab-

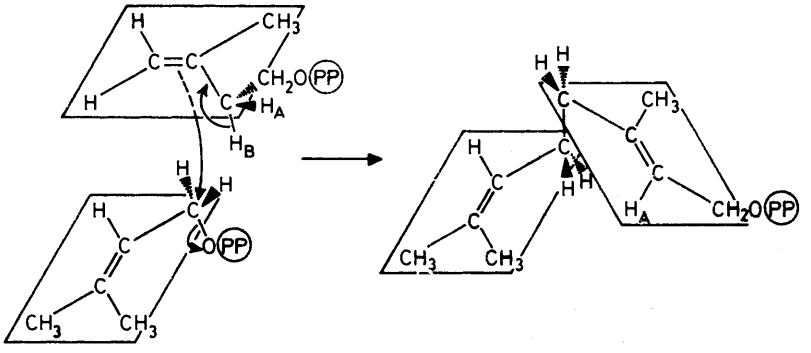
[1] J. R. Maxwell, C. T. Pillinger, and G. Eglinton, *Quart. Rev.*, 1971, **25**, 571.
[2] B. W. Agranoff, H. Eggerer, U. Henning, and F. Lynen, *J. Biol. Chem.*, 1960, **235**, 326.
[3] D. H. Shah, W. W. Cleland, and J. W. Porter, *J. Biol. Chem.*, 1965, **240**, 1946.
[4] P. Holloway and G. Popjak, *Biochem. J.*, 1967, **104**, 57; 1968, **106**, 835.
[5] F. Lynen, B. W. Agranoff, H. Eggerer, U. Henning, and E. M. Möslein, *Angew. Chem.*, 1959, **71**, 657.
[6] P. W. Holloway and G. Popjak, *Biochem. J.*, 1966, **100**, 61P.

lished the stereochemistry of these processes as *anti* for the isomerisation (Scheme 1) [7-9] and *syn* for the coupling (Scheme 2).[8,10] Intuitively,[11,12] the *anti*-mode might be



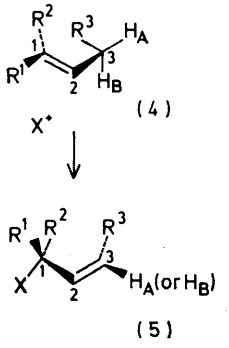
SCHEME 1

regarded as energetically preferable and this led Cornforth and Popjak to propose [11,12] a two-step sequence in order to rationalise the observed *syn* nature of the



SCHEME 2

coupling process. Predictions of preferred stereochemistry in the S_E2' reaction, made on the basis of orbital symmetry considerations, also indicate [13-15] *anti* preference for a concerted process. However, there is to



SCHEME 3

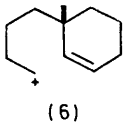
our knowledge no clear-cut demonstration, analogous to the model examined by Stork and White for the S_N2' reaction,[16] of the stereochemical outcome of a non-enzymic S_E2' reaction. This therefore merits attention in its own right, but additional interest derives from the question as to whether any stereochemical preference that may characterise a non-enzymic model will also be found in enzymic systems.[17-19] It was our aim to construct and examine a model that might shed light on this issue.

The essentials of the model are defined as follows (Scheme 3): (i) the electrophile X^+ must be made to react at C-1 in (4); (ii) the configuration at C-1 in the product (5) must be ascertainable; (iii) the electron deficiency that results at C-2 when X^+ attacks at C-1 must be neutralised by loss of H^+ from C-3; (iv) the exact proportion of H_A to H_B lost in the reaction must be measurable; and (v) accordingly it must be possible to replace stereoselectively H_A or H_B with deuterium or tritium and to know precisely the distribution of isotope between the H_A and H_B positions in (4).

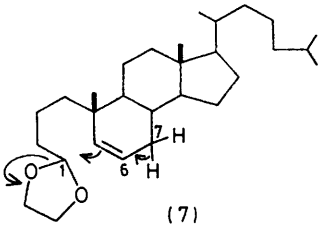
It was tempting for a variety of reasons to base the model on the part structure (6), and our choice fell on the 4,5-secocholest-5-ene derivative (7) which under acidic catalysis was expected to afford the cholest-6-ene (8). The acetal had the following attractions: (i) the possibility of intramolecular interaction between the potential cation, X^+, and the double bond, expected to lead to a six-membered ring *via* a geometrically favourable transition state; (ii) the probability of neutralising positive charge at C-6 by proton loss from C-7; (iii) prior work [20,21] on polyene cyclisations initiated by ethylene acetals; (iv) the ease of establishing the C-5 configuration in the product (8) by reference to C-10; and (v) the availability of cholesterol and the possibility

[7] J. W. Cornforth, R. H. Cornforth, C. Donninger, and G. Popjak, *Proc. Roy. Soc.* 1966, (*B*), **163**, 492.
[8] B. L. Archer, D. Barnard, E. G. Cockbain, J. W. Cornforth, R. H. Cornforth, and G .Popjak, *Proc. Roy. Soc.* 1966, (*B*), **163**, 519.
[9] J. W. Cornforth, K. Clifford, R. Mallaby, and G. T. Phillips, *Proc. Roy. Soc.* 1972, (*B*), **182**, 277.
[10] J. W. Cornforth, R. H. Cornforth, G. Popjak, and L. Yengoyan, *J. Biol. Chem.*, 1966, **241**, 3970.
[11] G. Popjak and J. W. Cornforth, *Biochem. J.*, 1966, **101**, 553.
[12] J. W. Cornforth, *Angew. Chem. Internat. Edn.*, 1968, **7**, 903.
[13] K. Fukui and H. Fujimoto, *Bull. Chem. Soc. Japan*, 1966, **39**, 2116.
[14] S. I. Miller, *Adv. Phys. Org. Chem.*, 1968, **6**, 185.
[15] N. T. Anh, *Chem. Comm.*, 1968, 1089.
[16] G. Stork and W. N. White, *J. Amer. Chem. Soc.*, 1956, **78**, 4609.
[17] I. A. Rose, *CRC Crit. Rev. Biochem.*, 1972, 33.
[18] I. A. Rose in 'The Enzymes,' 3rd edn., ed. P. D. Boyer, Academic Press, New York, vol. II, 1970, p. 281.
[19] K. R. Hanson, *Ann. Rev. Plant Physiol.*, 1972, **23**, 335.
[20] W. S. Johnson, *Accounts Chem. Res.*, 1968, **1**, 1.
[21] G. D. Abrams, W. R. Bartlett, V. A. Fung, and W. S. Johnson, *Bio-org. Chem.*, 1971, **1**, 243.

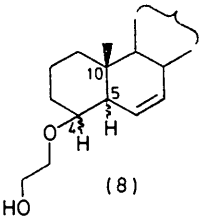
of stereoselective labelling at C-7 by well established biological methods.[22]



(6)



(7)

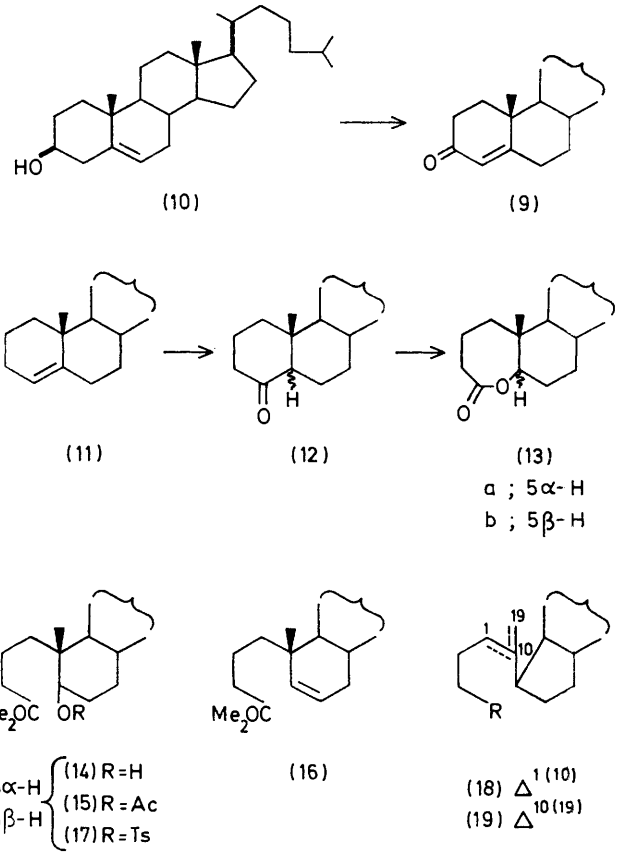


(8)

The acetal (7) was synthesised from cholesterol by the route depicted in Scheme 4. Cholestenone (9), obtained [23] from cholesterol (10), was converted into cholest-4-ene (11) with the mixed $AlCl_3$–$LiAlH_4$ reagent.[24] Hydroboration followed by oxidative work-up [25] afforded the 5α- and 5β-4-ones (12), separable by preparative layer chromatography (p.l.c.), initially in equal amounts, but after equilibration with base in the proportions 9 : 1 (α : β). Baeyer–Villiger oxidation of the equilibrium mixture with trifluoroperacetic acid in buffered methylene chloride [26] afforded the corresponding ε-lactones (13) (9 : 1), separable by p.l.c. in 85—90% yield. The lactone mixture was successively hydrolysed and esterified to afford the methyl esters (14a and b) which were separated by p.l.c.

Elimination of the C-5 alcohol function, to produce the 5,6-olefin proved troublesome. The major alcoholic ester (14a) has the β- (equatorial) configuration. Pyrolysis of the methyl acetate ester (15a) afforded variable yields of the required ester (16) and an unsaturated hydrocarbon which was not further investigated. Repeated passes through the furnace to effect conversion of unchanged acetate, resulted in increased amounts of the unsaturated hydrocarbon and diminished overall recovery. Elimination of the equatorial tosylate (17a) was attempted under a variety of conditions (see Experimental section). Heating in dry dimethyl sulphoxide afforded the required ester (16), characterised by its spectral properties and identical with the major ester from acetate pyrolysis. These conditions also produced several isomeric esters, among them (18) and (19) (R = CO_2Me) (see Experimental section) which result from migration of the 9,10-bond following tosylate solvolysis. Further experimentation showed the best reagent combination to be a ten-fold excess of lithium bromide with added lithium carbonate in dry dimethylformamide at 115 °C, and this afforded the derived ester (16) as the major component (>90%) of the olefinic fraction.

The olefinic ester (16) was finally converted into the ethylene acetal (20a) *via* the alcohol (28a) and aldehyde (21a) by standard procedures. In subsequent runs to procure the acetal in quantity for further elaboration, it was obtained in 10% overall yield from cholestenone without isolation of every intermediate (see Experimental section). In the course of one run, the aldehyde (19; R = CHO) was obtained as a by-product. An attempt to



(10) (9) (11) (12) (13) a ; 5α-H b ; 5β-H

a;5α-H, b;5β-H { (14) R=H, (15) R=Ac, (17) R=Ts } (16) (18) $\Delta^{1(10)}$ (19) $\Delta^{10(19)}$

SCHEME 4

characterise it by osmylation led to a mixture (2 : 5 by g.l.c.) of two internal acetals isomeric at C-10 whose i.r., n.m.r., and mass spectra supported their formulation as (22). The major fragment ions in the almost identical mass spectra of the two compounds are accommodated as in Scheme 5.

With a sufficient quantity of the olefinic acetal at our disposal, we now sought conditions to cyclise it to the tetracyclic olefin (8). Drawing on the extensive experience [20,21] of Johnson's school, a variety of Lewis acids and solvents were tried. An illustrative selection

[22] D. C. Wilton, K. A. Munday, S. J. M. Skinner, and M. Akhtar, *Biochem. J.*, 1968, **106**, 803; L. Canonica, A. Fiecchi, M. G. Kienle, A. Scala, G. Galli, E. G. Paoletti, and R. Paoletti, *Steroids*, 1968, **11**, 749; E. Caspi, J. B. Greig, P. J. Ramm, and K. R. Varma, *Tetrahedron Letters*, 1968, 3829; G. F. Gibbons, L. J. Goad, and T. W. Goodwin, *Chem. Comm.*, 1968, 1212.

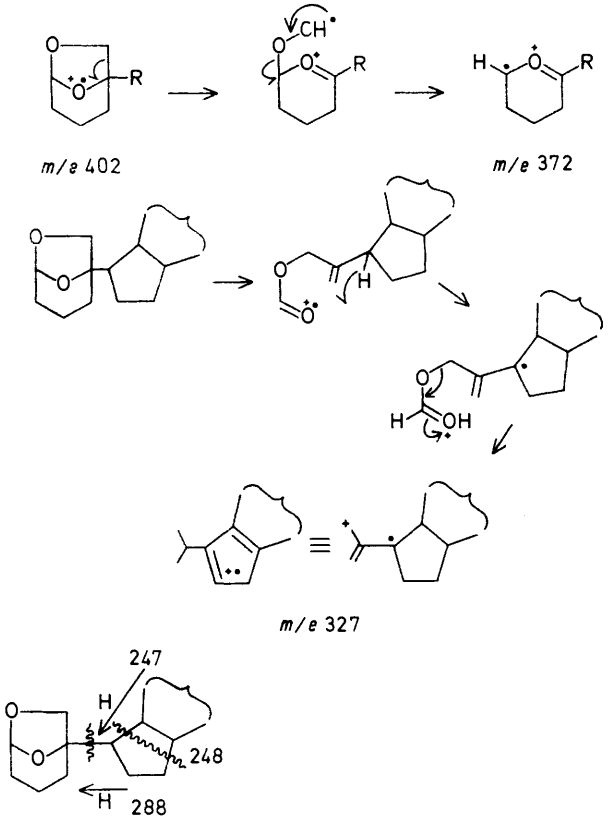
[23] L. F. Fieser, *Org. Synth.*, Coll. Vol. IV, 1963, p. 195.

[24] B. R. Brown and A. M. S. White, *J. Chem. Soc.*, 1957, 3755.

[25] J. R. Bull, E. R. H. Jones, and G. D. Meakins, *J. Chem. Soc.*, 1965, 2601.

[26] E. E. Smissman, J. R. Murer, and N. A. Dahle, *J. Org. Chem.*, 1964, **29**, 3517.

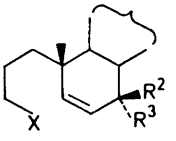
is recorded in the Experimental section. Initial results were discouraging. Typically, at that stage, reaction



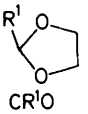
SCHEME 5

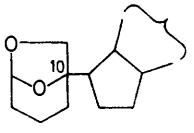
of the acetal with tin(IV) chloride in nitromethane at 20 °C afforded at least seven products (by g.l.c.); only one, produced in minor amount, showed in g.l.c.–mass spectrometric analysis a parent ion corresponding to the desired product (8). Systematic variation of temperature, solvent, and reactant concentrations eventually defined conditions (acetal : tin chloride, 1 : 20; methylene chloride; −78 °C; 30 s) that reproducibly afforded a much simplified product mixture with only four major components detected by g.l.c. Two of these, obtained pure by p.l.c. and each formed in 15% yield, were the expected olefinic ether [(8) ≡ (23a)] and the unexpected saturated alcohol (25). The structure of the ether (23a) was assigned on the basis of its i.r., n.m.r., and mass spectral characteristics and it was converted into 5β-cholestan-4α-ol (24) by successive hydrogenation with di-imide and removal of the ethanol chain by reductive fission of the tosylate with sodium iodide and zinc.[27] The expected position of the double bond in (23a) was supported in the n.m.r. spectrum by a quartet (2 H) centred at δ 5.62 whose upfield doublet was further split by a minor coupling, and by additional evidence adduced below. The saturated alcohol (25) was identical with an authentic specimen of 5β-cholestan-4β-ol and was oxidised to 5β-cholestan-4-one.[28] Of the other two major components from cyclisation detected by g.l.c.–mass spectrometry, one corresponds to a cholestadiene, possibly produced on the g.l.c. support since it does not appear in the g.l.c. trace of the trimethylsilyl ethers. The other, more inscrutably, has a parent ion corresponding to the loss of H_2 from the ether (23a). The mode of formation of (23a) and (25) is discussed below. The major cyclisation product (23a) thus had an AB-*cis*-fusion and hence H-5 must be β and the newly formed C–C bond α. Cyclisation products with rings A and B *trans*-fused were not formed. Models show that they are not to be expected.

We were next concerned to establish that proton loss occurred cleanly from C-7 in the cyclisation of (20a) to (23a), as would be required in an S_E2'-type process. To this end, C-7 was in the first instance doubly deuteriated by reducing the enone ester (26) (for its formation see Experimental section) with dichloroaluminium deuteride (generated from $LiAlD_4$–$AlCl_3$, 1 : 4). The derived

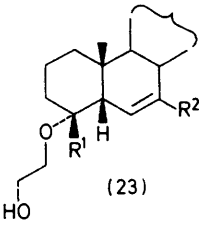


a ; $R^1=R^2=R^3=H$
b ; $R^1=R^2=R^3=D$
c ; $R^1=R^2=H$, $R^3=D$
d ; $R^1=R^3=H$, $R^2=D$

(20) X = 
(21) X = CR^1O
(28) X = CR^1_2OH
(29) X = CR^1_2OBs

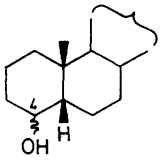


(22)



(23)

a ; $R^1=R^2=H$
b ; $R^1=R^2=D$
c ; $R^1=H$, $R^2=D$



(24) 4β-H
(25) 4α-H

trideuterioacetal (20b) cyclised with loss of one deuterium atom from C-7 and retention of the other in the product (23b) [$\nu_{max.}$ 2 235 cm^{-1}; δ 5.57 (1 H, d, –CH=CD–)].

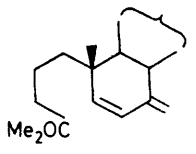
With the reaction course established in the desired sense, we now confronted our last experimental problem,

[27] W. S. Johnson and R. B. Kinnel, *J. Amer. Chem. Soc.*, 1966, **88**, 3861.

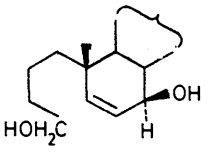
[28] C. Djerassi, R. Riniker, and B. Riniker, *J. Amer. Chem. Soc.*, 1956, **78**, 6362.

namely to replace in turn each C-7 hydrogen atom of the acetal (20a) by deuterium or tritium stereoselectively and to determine precisely how effectively this had been done. Two appropriate methods had been described; one,[22] biochemical, uses a rat-liver preparation and is capable of introducing tritium stereospecifically from either tritiated mevalonolactone or tritiated NADH at C-7 of cholesterol; the other,[29] chemical, is suitable for deuteriation and depends upon ketonisation of the enol of 3β-acetoxycholestan-6-one.

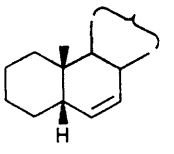
We chose a third method based on our synthesis of the 4,7,7-trideuterio-acetal (20b) (see above). Thus the allylic alcohol (27), available from the enone ester (26) by reduction with lithium aluminium hydride, was deuteriolised by dichloroaluminium deuteride to the olefinic alcohol (28c). The =CH– region of its n.m.r. spectrum



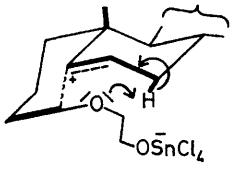
(26)



(27)



(31)



(30)

suggested that deuterium had been introduced predominantly with inversion into the 7α-position. However, as indicated previously, it was essential to know precisely the proportion of 7α- and 7β-deuterio-isomers formed in this reaction. Photo-oxygenation should, by analogy with the reaction of cholesterol,[30,31] selectively remove the 7α-hydrogen (or deuterium) atom in forming the 5α-hydroperoxy-6-ene, and so provide an ideal analytical method.

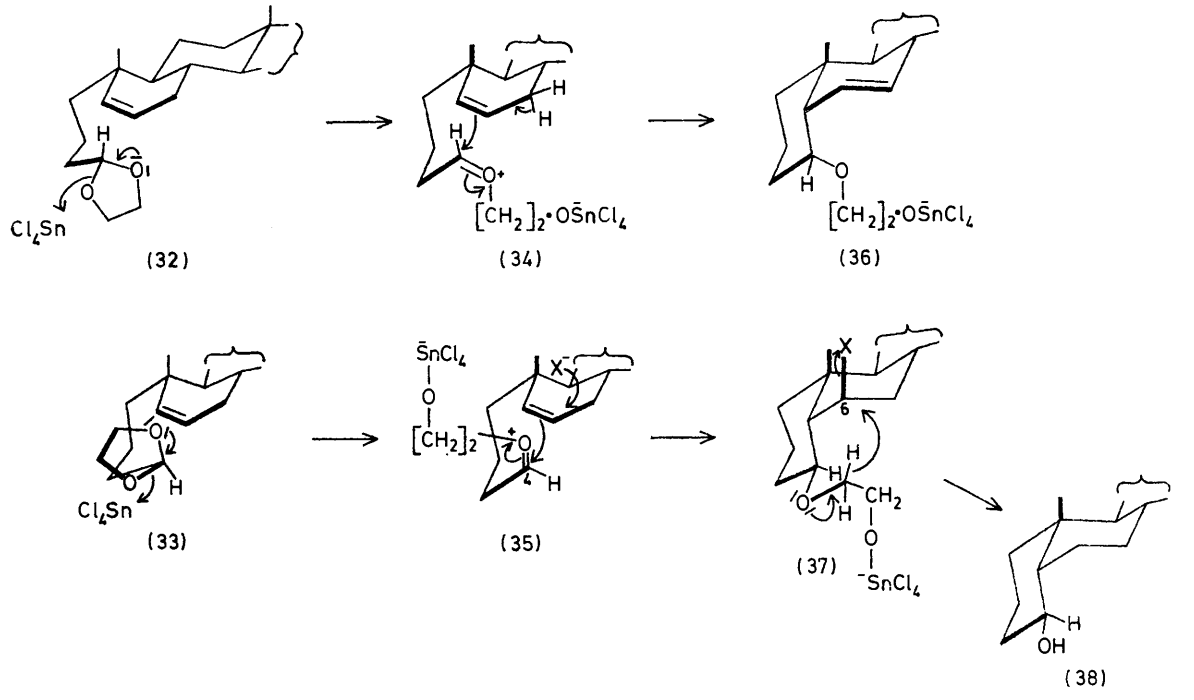
In our experience this photo-oxygenation failed to take place with the alcohol (28a). We were therefore forced to effect selective 7α- and 7β-deuteriation with cholesterol by using the two-step reduction described above, analyse each mixture of 7α- and 7β-deuterio-cholesterols by photo-oxygenation, and then convert it into the ring-A-cleaved aldehyde acetals (20c or d) by the route previously described. The details of the synthetic method and product analysis have been reported elsewhere.[32] In this way there became available the 7β-deuterio-aldehyde (21d) (97% 7β-D), which was converted into the acetal (20d) (97% 7β-D). Its n.m.r. spectrum clearly showed loss of the vicinal coupling between H-6 and H-7β seen in that of the diprotio-acetal (20a). Cyclisation of the acetal afforded the hydroxy-ether (23c), which retained deuterium (>96% D by mass spectrometry). The simple inference that might be drawn from this result is that in our model an S_E2' reaction has proceeded with *syn* stereochemistry, since formation of the α-C(4)–C(5) bond was accompanied by loss of the allylic α-hydrogen atom from C-7. This formally parallels the stereochemistry observed in the enzymic coupling of C_5 units in isoprenoid biosynthesis. However, we were concerned lest loss of the α-hydrogen atom from C-7 was assisted intramolecularly by the ether oxygen function of the cleaved acetal system as in (30). If this were the case, the observed result might not reflect the intrinsic stereochemical preference of the S_E2' reaction. In an attempt to allay these suspicions, cyclisation was induced under entirely different conditions. The sample of cholesterol deuteriated predominantly in the 7α-position (82.5% 7α-D, 17.5% 7β-D) was converted into the primary alcohol (28c) and then its *p*-bromobenzenesulphonate (29c). Trahanovsky and Doyle have shown [33] that in 2,2,2-trifluoroethanol, a non-nucleophilic but highly efficient ionising solvent, olefinic esters such as (29a) can be expected to cyclise readily. Solvolysis of the deuteriated brosylate (29c) in trifluoroethanol at 105 °C for 40 h afforded 5β-cholest-6-ene (31) as the predominant hydrocarbon, which by mass spectrometry had 16.5% D_1 and 83.5% D_0. Thus, under the entirely different solvolytic conditions of this second experiment (effectively two simultaneous experiments with the deuterium label in either the 7α- or the 7β-position) the observed stereochemistry was again stereospecifically *syn*.

Our experimental results thus clearly establish that for the two cyclisations examined, *i.e.* (20) ⟶ (23) and (29) ⟶ (31), there is a *syn* relationship between the newly formed C–C bond and the allylic hydrogen atom that is lost.

To assess how relevant these findings are to the stereochemical course of the S_E2' reaction, it is appropriate first to consider in some detail the steps that lead from (20) to (23) and (25). The first step must be complexation of tin(IV) chloride with one of the acetal oxygen atoms, followed by fission of its bond with C-4 (Scheme 6). Two acetal conformations, (32) or (33), could be involved, as shown, implying a degree of participation by the olefinic double bond, and each would lead to a conformation, (34) or (35), of the oxonium ion which in turn determines the configuration at C-4 of the product obtained on cyclisation. So far as formation of the olefin (23) ≡ (36) is concerned, C–C bond formation and allylic hydrogen atom loss could be concerted in the S_E2' manner [(34) ⟶ (36)]. The saturated alcohol (25) ≡ (38) on the other hand, is likely to result from *trans*-addition to the olefinic double bond of C-4 and a nucleophile X^- (Cl^-?), followed by hydride transfer

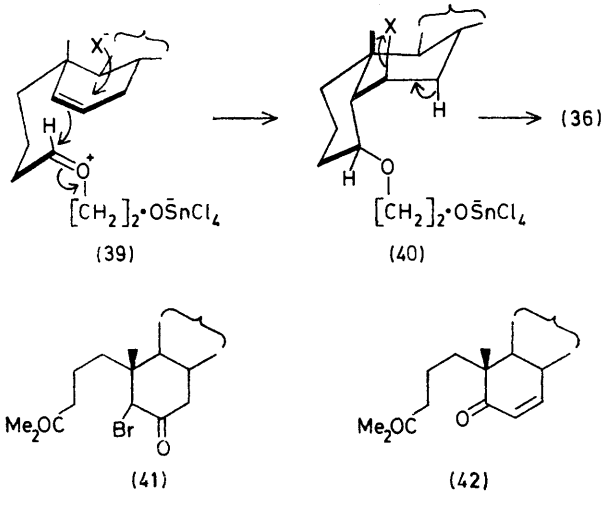
[29] E. J. Corey and G. A. Gregoriou, *J. Amer. Chem. Soc.*, 1959, **81**, 3127.
[30] A. Nickon and J. F. Bagli, *J. Amer. Chem. Soc.*, 1961, **83**, 1498.
[31] A. Nickon and W. L. Mendelson, *J. Amer. Chem. Soc.*, 1963, **85**, 1894.
[32] I. M. Cunningham and K. H. Overton, *J.C.S. Perkin I*, 1974, 2458.
[33] W. S. Trahanovsky and M. P. Doyle, *Tetrahedron Letters*, 1968, 2155.

from C-1′ of the ethylene glycol residue to C-6 with expulsion of X^-, as indicated [(35) ⟶ (37) ⟶ (38)]. Hydride transfer under similar circumstances has been observed.[34,35] An analogous two-step mechanism, which would correspond to Cornforth's suggestion for both cyclisations is quasi-axial. It is well known[36] that allylic σ–π interaction in cyclohexenes is stronger for the quasi-axial than for the quasi-equatorial allylic σ-bond. The loss in both cyclisations of the quasi-axial allylic hydrogen atom may simply reflect this preferential



SCHEME 6

the biological process, could account for formation of the olefin (23) also [(39) ⟶ (40) ⟶ (36)]. However,



cyclisation of the brosylate (29c) to the alkene (31) in the non-nucleophilic trifluoroethanol must surely be a concerted and not a two-step process.

If formation of (23) and (31) proceeds by an S_E2'-type mechanism, then our experimental findings raise one further doubt. The allylic hydrogen atom eliminated in interaction. Indeed, it is conceivable that S_E2' reactions might proceed with either *syn* or *anti* stereochemistry, provided that a quasi-axial allylic hydrogen atom can be lost. We are attempting to probe this hypothesis by devising suitable model systems, but we have also undertaken some preliminary studies by computational methods to detect stereochemical preference in the S_E2' reaction. Our findings and conclusions will be reported in a later paper.

EXPERIMENTAL

M.p.s were determined with a Kofler hot-stage apparatus. Routine i.r. spectra were recorded, for solutions in carbon tetrachloride, with a Perkin-Elmer 257 spectrophotometer, and high resolution spectra with a Unicam SP 100 or a Perkin-Elmer 225 double-beam spectrophotometer. N.m.r. spectra were recorded for solutions in $CDCl_3$ with Me_4Si as internal standard with a Varian T60 or HA100 spectrometer. Mass spectra were routinely determined with an A.E.I. MS12 spectrometer; mixtures were normally examined by means of an LKB 9000 g.l.c.-linked mass spectrometer. High resolution mass spectra were obtained with an A.E.I. MS902 spectrometer and all spectra were recorded at 70 eV unless otherwise stated. Analytical g.l.c. was effected on a Pye–Argon or Perkin-Elmer F11 chromatograph and peaks are identified by either their

[34] D. J. Goldsmith and B. C. Clark, *Tetrahedron Letters*, 1967, 1215.

[35] D. J. Goldsmith and C. F. Phillips, *J. Amer. Chem. Soc.*, 1969, **21**, 5862.

[36] H. L. Goering and R. R. Josephson, *J. Amer. Chem. Soc.*, 1962, **84**, 2779.

retention time in minutes or by their carbon number. Merck Kieselgel HF_{254} was used for all layer chromatography, analytical t.l.c. on 0.25 mm plates and preparative (p.l.c.) on 1.0 mm plates. 'Silver nitrate plates' were made by using a slurry of 15% by weight of silver nitrate in silica gel G. Light petroleum refers to the fraction of b.p. 60—80°. All organic extracts were washed to neutrality by appropriate acidic or basic treatment and were dried over anhydrous magnesium sulphate.

Cholest-4-en-3-one (9).—Oxidation [23] of cholesterol afforded cholestenone (9) in 65—70% yield, m.p. 79—80° (lit.,[23] 79.5—80.5°).

Cholest-4-ene (11).—Anhydrous aluminium chloride (48 g, 0.36 mol) was carefully added to dry ether (200 ml). The mixture was stirred under nitrogen at 0 °C, lithium aluminium hydride (4.6 g, 0.12 mol) suspended in dry ether (150 ml) was added, and the slurry was stirred for 10 min. Cholestenone (23 g, 0.06 mol) in dry ether (200 ml) cooled to 0 °C was then added over 20 min, the mixture was stirred a further 30 min, and the reaction was then quenched by dropwise addition of water until the inorganic salts coagulated. Work-up in the usual way afforded (from ether–methanol) cholest-4-ene (11) (19.1 g, 86%), m.p. 80—81°, $[\alpha]_D$ +68.5 (*c* 1.45) (lit.,[25] m.p. 79—80 and 82—83°, $[\alpha]_D$ +65° and +76°).

5α- and 5β-Cholestan-4-ones (12).—Hydroboration [25] of cholest-4-ene and oxidation of the intermediate alcohols with 8N-Jones reagent afforded a mixture (1 : 1) of 5α- and 5β-cholestan-4-ones (12), which were separated by p.l.c. with ethyl acetate–light petroleum (1 : 9). 5α-Cholestan-4-one had $\nu_{max.}$ 1 715, 1 210, 935, and 905 cm^{-1} δ_{Me} 0.89, 0.83, 0.74, and 0.64; g.l.c. (1% OV-1 at 225 °C) retention index * (r.i.) 2 917 and 2 975 (1 : 8). 5β-Cholestan-4-one had $\nu_{max.}$ 1 710, 1 180, 1 165, and 930 cm^{-1}; δ_{Me} 1.06, 0.85, 0.79, and 0.60, g.l.c. 2 917 and 2 975 (1 : 1). Equilibration of each sample had clearly occurred on g.l.c. Refluxing the crude product with methanolic 5% KOH afforded a 9 : 1 (g.l.c.) 5α : 5β mixture which was used without purification.

Baeyer–Villiger Oxidation of the Ketone Mixture (12): *the ε-Lactones* (13).—Trifluoroperacetic acid was prepared by carefully adding trifluoroacetic anhydride (10.2 ml, 0.07 mol) to ice-cold 90% H_2O_2 (1.6 ml, 0.06 mol) in dry CH_2Cl_2 (10 ml) under N_2. This was added over 30 min to a stirred suspension of anhydrous Na_2HPO_4 (16 g) in dry CH_2Cl_2 (80 ml) containing the ketones (12) (15.5 g, 0.04 mol). The mixture was kept at 0 °C during addition and for 1 h more and then at 20 °C for 2 h. Work-up gave a mixture (9 : 1) of lactones (13) (14.5 g). P.l.c. in ethyl acetate–light petroleum (1 : 3) afforded (sublimation at 180° and 0.7 Torr) the more polar *4,5-secocholestan-4,5β-carbolactone* (13a), m.p. 120—122°, $\nu_{max.}$ 1 740, 1 292, 1 278, 1 190, 1 100, and 1 035 cm^{-1}, δ 4.13 (1 H, q, lines at ±3 and ±10 Hz) (Found: C, 80.4; H, 11.4. $C_{27}H_{46}O_2$ requires C, 80.55; H, 11.5%). The less polar component was *4,5-secocholestan-4,5α-carbolactone* (13b) (sublimed at 170 °C and 0.1 Torr), a gum, $\nu_{max.}$ 1 735, 1 300, 1 290, 1 280, 1 265, 1 185, and 1 065 cm^{-1}, δ 4.64 (1 H, q, lines at ±2 and ±12 Hz) (Found: C, 80.65; H, 11.75%).

Hydrolysis of the Lactones (13).—The lactones were hydrolysed by stirring in methanolic 5% KOH at 20 °C for 16 h. The hydroxy-acids, isolated as usual, had $\nu_{max.}$ 3 600—2 500br, 1 705, 1 110, and 1 075 cm^{-1}; δ 5.06br (2 H, s, exchangeable with D_2O) and 3.60 (1 H, m).

The methyl esters, obtained with CH_2N_2, were separated by p.l.c. (ethyl acetate–light petroleum, 1 : 3), affording *methyl 5β-hydroxy-4,5-secocholestane-4-carboxylate* (14a), a gum, $\nu_{max.}$ 3 622, 3 550, 1 738, 1 235, 1 195, 1 169, 1 080, 1 040, and 980 cm^{-1}; δ 3.68 (3 H, s) and 3.60 (1 H, q); M^+ 434 ($C_{28}H_{50}O_3$), and the more mobile *5α-hydroxy-isomer* (14b), a gum, $\nu_{max.}$ 3 625, 3 540, 1 735, 1 235, 1 200, 1 168, 1 055, and 995 cm^{-1}; δ 3.67 (4 H, s); M^+ 434.

Acetylation of (14a) with pyridine–acetic anhydride at 20 °C for 17 h afforded after p.l.c. (ethyl acetate–light petroleum, 3 : 7) the *5β-acetate* (15a), a gum, $\nu_{max.}$ 1 740, 1 730, 1 210, 1 165, 1 020, 1 000, and 960 cm^{-1}; δ 4.73 (1 H, q, lines at ±4 and ±13 Hz), 3.66 (3 H, s), and 2.02 (3 H, s) (Found C, 75.55; H, 10.95. $C_{30}H_{52}O_4$ requires C, 75.6; H, 11.0%). Similarly acetylation of (14b) afforded the *5α-acetate* (15b), a gum, $\nu_{max.}$ 1 738, 1 240, 1 170, 1 015, and 965 cm^{-1}; δ 4.80 (1 H, s), 3.67 (3 H, s), and 2.06 (3 H, s) (Found: C, 75.7; H, 11.0%).

Treatment of the hydroxy-ester (14a) (1 g, 2.4 mmol) with freshly crystallised toluene-*p*-sulphonyl chloride (0.95 g, 5 mmol) in the minimum of anhydrous pyridine at 20 °C for 65 h afforded the *5β-tosylate* (17a) (1.25 g, 90%), purified by p.l.c. (ethyl acetate–light petroleum, 1 : 3); $\nu_{max.}$ 1 740, 1 180, 1 170, 1 095, 965, 930, 900, and 890 cm^{-1}; δ 7.56 (4 H, q, lines at ±16 and ±33 Hz), 4.50 (1 H, q, lines at ±3 and ±12 Hz), and 3.68 (3 H, s) and 2.44 (3 H, s).

The isomeric hydroxy-ester (14b) gave, by the same method, the *5α-tosylate* (17b) (65%), $\nu_{max.}$ 1 740, 1 180, 1 170, and 900 cm^{-1}; δ 7.50 (4 H, q, lines at ±16 and ±28 Hz), 4.65 (1 H, s), 3.65 (3 H, s), and 2.67 (3 H, s).

Methyl 5-Oxo-4,5-secocholestane-4-carboxylate.—Oxidation of the hydroxy-ester (14a) (100 mg, 0.24 mmol) with 8N-Jones reagent afforded the *5-ketone* (90 mg, 90%) which, purified by p.l.c. (ethyl acetate–light petroleum, 1 : 6), had $\nu_{max.}$ 1 740, 1 710, 1 190, 1 165, 1 090, and 950 cm^{-1}; δ 3.67 (3 H, s) (Found: M^+, 432.358 30. $C_{28}H_{48}O_3$ requires *M*, 432.360 32).

The Olefinic Ester (16).—(i) Dehydration of the hydroxy-acid (14a) with $POCl_3$ in anhydrous pyridine at 0 °C afforded only the lactone (13a) and unchanged hydroxy acid. The epimeric hydroxy-acid (14b) under similar conditions gave a complex mixture which was not further investigated.

(ii) The acetate methyl ester (15a) (40 mg) in benzene (1 ml) was introduced dropwise in a stream of dry N_2 into a 2 m narrow-bore silica tube, loosely packed with glass wool, which was clamped vertically and surrounded by a furnace maintained at 540 °C. The pyrolysate was collected in a cooled receiver at the lower end of the pyrolysis tube which projected from the furnace. It consisted of a minor component, apparently an olefinic hydrocarbon [$\nu_{max.}$ 3 010, 1 640, 910, and 880 cm^{-1}] and as major product the desired olefinic ester (16), contaminated with a minor impurity (g.l.c.) and more expeditiously prepared by elimination of the tosylate (17a) (see below), as well as *ca.* 50% unchanged acetate. Two further passes through the furnace transformed the remaining acetate but increased the proportion of hydrocarbon at the expense of the desired olefinic ester.

(iii) The tosylate (17a) (250 mg), stirred with neutral alumina (grade I; 10 g) in benzene–light petroleum (1 : 1) for 80 h, afforded unchanged tosylate (150 mg) and a product (30 mg), identical (t.l.c.) with the major product from the acetate pyrolysis in (ii).

* Obtained from the retention time of a compound by interpolation on a log (retention time) against retention index plot for n-alkanes taken under the same conditions. The r.i. of an n-alkane $C_n H_{2n+2}$ is defined as 100*n*.

(iv) The tosylate (17a) (900 mg, 1.5 mmol) in dry dimethyl sulphoxide (5 ml) was maintained at 115 °C for 17 h under N_2. The product (560 mg), obtained as usual, showed on t.l.c. one major band corresponding to the major product from acetate pyrolysis. It was isolated by p.l.c. (ethyl acetate–light petroleum, 1 : 9) but then showed three incompletely separated bands on t.l.c. ($AgNO_3$–SiO_2) and three peaks at t_R 50.5, 54 and 58.5 min (6 : 4 : 1) on g.l.c. over 1% OV-17 at 200 °C. P.l.c. on 0.5 mm $AgNO_3$–Kieselgel plates with multiple elution afforded the least mobile *methyl 4,5-secocholest-5-ene-4-carboxylate* (16), a gum, $\nu_{max.}$ 3 010, 1 740, 1 640, 1 190, 1 165, and 890 cm^{-1}; δ 5.45 (2 H, m, lines at ±10 and ±20 Hz, low-field doublet further split by ±3 Hz), 3.64 (3 H, s), and 2.25 (2 H, t, *J* 7 Hz) [on irradiation at δ 1.92, the multiplet at δ 5.45 was reduced to a quartet (lines at ±9 and ±20 Hz)]; g.l.c. on 1% OV-17 at 200 °C, t_R 54 min, identical, by co-injection, with the major product from acetate pyrolysis (Found: C, 80.8; H, 11.6. $C_{28}H_{48}O_2$ requires C, 80.7; H, 11.6%).

The band of intermediate polarity from t.l.c. on $AgNO_3$–SiO_2, consisting of a mixture of the two rearranged olefinic esters (18) and (19) (R = CO_2Me), in the ratio *ca.* 5 : 1, had $\nu_{max.}$ 1 740, 1 240, 1 190, and 1 165 cm^{-1}; δ 5.05 (0.75 H, t, HC=C), 4.63 (0.25 H, d, *J* 8 Hz, H_2C=C), 3.63 (3 H, s, $CO_2{\cdot}CH_3$), 2.33 (3 H, d, *J* 3 Hz, $CH_2{\cdot}CO_2Me$ and $CH_2{\cdot}C$=C), and 1.62 (2.5 H, s, C=C·CH_3). These assignments were supported by double irradiation. The most mobile band from t.l.c. was also a mixture (5 : 1) of the components having t_R on 1% OV-17 at 200 °C of 50.5 and 58.5 min, and $\nu_{max.}$ 1 740, 1 250, 1 185, and 1 165 cm^{-1}, δ 5.48 (0.8 H), 3.64 (3 H, s), and 1.60 (3 H, s).

The tosylate (17a) was treated under a variety of conditions intended to increase the proportion of (16) in the mixture of unsaturated esters. The conditions included $KOBu^t$–Me_2SO at 110 °C, pyridine at reflux, acetic acid–sodium acetate at reflux, and dimethylformamide at 100 °C.

Optimal conditions which afforded the desired ester (16) with least contamination by isomers were as follows. The tosylate (17a) (600 mg, 1.02 mmol), anhydrous LiBr (850 mg, 10 mmol), and anhydrous Li_2CO_3 (150 mg, 2 mmol) in dry dimethylformamide (12 ml) were kept at 115 °C under N_2 for 2 h. The product, extracted into light petroleum, when filtered through a short column of neutral alumina (grade III) in benzene, afforded the ester (16) (250 mg, 62%), g.l.c. on 1% OV-17 at 200° t_R 50.5 and 54 min (1 : 30), homogeneous on t.l.c. ($AgNO_3$–SiO_2).

The Olefinic Alcohol (28a).—The ester (16) (140 mg, 0.35 mmol) in dry ether (4 ml) was added dropwise to a stirred suspension of $LiAlH_4$ (80 mg, 2.1 mmol) in dry ether (5 ml) at 0 °C under N_2, and stirring was continued for 20 min. Work-up as usual, by dropwise addition of H_2O to coagulation, afforded *4,5-secocholest-5-en-4-ol* (28a) (122 mg, 94%), a gum. Purified by p.l.c. (ethyl acetate–light petroleum, 3 : 17) it had $\nu_{max.}$ 3 636, 3 480, 3 008, 1 652, 1 050, and 1 030 cm^{-1}; δ 5.46 (2 H, lines at ±8 and ±18 Hz, low-field doublet further split by 4 Hz) and 3.62 (1 H, t, *J* 6 Hz) (Found: C, 83.4; H, 12.65. $C_{27}H_{48}O$ requires C, 83.45; H, 12.45%).

The Olefinic Aldehyde (21a).—Oxidation of the above alcohol with CrO_3–pyridine–CH_2Cl_2 [37] afforded (70—90%) *4,5-secocholest-5-en-4-al* (21a), purified by p.l.c. [ethyl acetate–light petroleum (1 : 9)], a gum, $\nu_{max.}$ 3 010, 2 710, 1 730, and 1 655 cm^{-1}; δ 9.7 (1 H, t, *J* 2 Hz) and 5.46 (2 H, m, lines at ±6 and ±22 Hz, low-field doublet further split by 4 Hz) (Found: C, 83.95; H, 12.2. $C_{27}H_{46}O$ requires C, 83.85; H, 12.0%).

In one experiment, purification by p.l.c. at the aldehyde stage of the mixture of olefins obtained from tosylate elimination, afforded the minor rearranged aldehyde 9(10 ⟶ 5)abeo-*4,5-secocholest*-10(19)-*en*-4-*al* (19; R = CHO), $\nu_{max.}$ 3 080, 2 705, 1 730, 1 630, and 890 cm^{-1}; δ 9.78 (1 H, t, *J* 2 Hz), 4.68 (1 H, s), and 4.48 (1 H, s) (Found: C, 83.85; H, 12.2. $C_{27}H_{46}O$ requires C, 83.85; H, 12.0%).

The aldehyde (19; R = CHO) (30 mg, 0.075 mmol) and OsO_4 (40 mg, 0.16 mmol) were kept in dry benzene (3 ml) at 20 °C for 46 h. The product, *10,19-dihydroxy*-9(10 ⟶ 5)-abeo-*4,5-secocholestan*-4-*al internal acetal* (22), a homogeneous mixture of C-10 epimers, obtained by p.l.c. after decomposition of the osmate with H_2S, had $\nu_{max.}$ 1 145, 1 120, 1 105, 1 085, 1 040, 1 025, 1 010, 995, 870, and 840 cm^{-1}; δ 5.44 (1 H, $W_{\frac{1}{2}}$ 4 Hz), 4.29 (0.7 H, d, *J* 6 Hz), 4.09 (0.3 H, d, *J* 7 Hz), 3.26 (0.7 H, d, *J* 7 Hz), and 3.13 (0.3 H, d, *J* 7 Hz), g.l.c. (1% OV-1; 225 °C) r.i. 2 967 and 3 006 (2 : 5). G.l.c.–mass spectrometry gave for the minor component *m/e* 402 (M^+, 100%), 387 (10), 384 (15), 372 (70), 357 (15), 327 (55), 288 (40), 248 (58), and 247 (85); and for the major component *m/e* 402 (M^+, 83%), other peaks at 387 (11), 372 (81), 357 (32), 343 (16), 327 (100), 314 (29), 301 (34), 247 (47), and 215 (61).

The Acetal (20a).—The aldehyde (21) (1.13 g, 2.92 mmol), toluene-*p*-sulphonic acid (20 mg), and an excess of ethylene glycol in benzene (25 ml) were refluxed for 3 h with continuous water separation. Work-up afforded *4,5-secocholest-5-en-4-al ethylene acetal* (20a), purified by p.l.c. [ethyl acetate–light petroleum (1 : 9)]; $\nu_{max.}$ 1 652, 1 135, 1 128, 1 060, 955, and 940 cm^{-1}; δ 5.43 (2 H, m, lines at ±7 and ±17 Hz, low-field lines further split by 5 Hz) (Found: C, 80.8; H, 11.65. $C_{29}H_{50}O_2$ requires C, 80.9; H, 11.7%).

Conversion of Cholesterol (10) *into the Acetal* (20); *Preparative Procedure*.—Cholestenone (9) (23.0 g), prepared as described above and crystallised from MeOH, on reduction with dichloroaluminium hydride, afforded cholest-4-ene (11) (20.5 g). Hydroboration–oxidation of this crude product afforded 5α- and 5β-cholestan-4-ones (12) (9 : 1) (17 g); they were oxidised to the corresponding lactones (13) and these were converted into the hydroxy-acids, which, after separation from non-acidic material, were methylated to give the esters (14) (12 g). Tosylation of this ester mixture with a 2.5-fold excess of toluene-*p*-sulphonyl chloride afforded the tosylates (17) containing some unchanged hydroxy-esters (16.0 g).

Elimination in dimethylformamide, as already described, and filtration in benzene of the product (9.5 g) through grade III neutral alumina, afforded the olefinic ester (16) (5.1 g). Reduction, oxidation, and acetalisation furnished the olefinic acetal (20a) (2.6 g, 10% yield based on cholestenone).

Cyclisation of the Olefinic Acetal (20a).—(i) The olefin acetal (0.023M in benzene; 0.3 ml) and tin(IV) chloride (0.094M in benzene; 0.3 ml) were mixed at 20 °C and kept for 3 h. Quenching with dilute $NaHCO_3$ and extraction into ether afforded only starting material.

(ii) The olefin acetal (0.023M in CH_2Cl_2; 0.5 ml) and tin(IV) chloride (0.09M in CH_2Cl_2; 0.5 ml) (freshly distilled solvent) were pre-cooled at −78 °C, mixed, and kept at −78 °C for 1 min. G.l.c. of the product over 1% OV-1 (220 °C) showed peaks with r.i. 2 832, 2 870, 2 935, 2 966, and 3 078 (1 : 3 : 4 :

[37] R. Radcliffe and R. Rosehurst, *J. Org. Chem.*, 1970, **35**, 4000.

3 : 2). The component of r.i. 2 966 was separated by p.l.c. (ethyl acetate–light petroleum, 1 : 9) and had $\nu_{max.}$ 3 540, 3 010, 1 050, and 890 cm^{-1}; M^+ 430 ($C_{29}H_{50}O_2$). In an attempt to simplify the product, the mixture from cyclisation was converted into the tosylates (40 mg) (pyridine and toluene-*p*-sulphonyl chloride at 20 °C) and these were refluxed in dry dimethoxymethane (4 ml) with sodium iodide (100 mg) and freshly activated zinc powder (50 mg), added in portions over 2 h. Oxidation of the crude product with 8N-Jones reagent afforded a mixture showing on g.l.c. (OV-1; 200 °C) at least 17 peaks.

(iii) Tin(IV) chloride (4.5 mmol) in CH_2Cl_2 (10 ml) was added rapidly to the olefin acetal (100 mg, 0.23 mmol) in CH_2Cl_2 at −78 °C. The reaction was quenched after 30 s and the product isolated as usual. Two pure products were isolated by p.l.c. in ethyl acetate–light petroleum (1 : 9). The less polar component, 4-(2-*hydroxyethoxy*)-5β-*cholest*-6-*ene* (23a) (15 mg), had $\nu_{max.}$ 3 530, 3 010, 1 645, 1 150, 1 098, 1 050, 955, and 890 cm^{-1}; δ 5.62 (2 H, lines at ±4 and ±14 Hz, up-field doublet further split by 4 Hz), 3.62 (4 H, m), 3.46 (1 H, s), and 2.56 (1 H, t, *J* 6 Hz, exchangeable with D_2O) (irradiation at δ 1.76 removed the 4 Hz coupling from the up-field lines of the signal centred at δ 5.62); g.l.c. (1% OV-1; 200 °C) r.i. 2 966 (Found: M^+, 430.3778. $C_{29}H_{50}O_2$ requires *M*, 430.3811). The more polar component, 5β-cholestan-4β-ol (25) (16 mg), had $\nu_{max.}$ 3 630, 3 610, 1 050, 1 030, 1 020, 1 010, and 930 cm^{-1}; δ 3.94 (1 H, s, $W_{\frac{1}{2}}$ 20 Hz), and 0.96, 0.88, 0.82, and 0.64 (Me groups); g.l.c. (1% OV-1; 200 °C) r.i. 2 936, identical (co-injection) with an authentic sample (Found: M^+, 388.3700. Calc. for $C_{27}H_{48}O$: *M*, 388.3705). Jones oxidation afforded 5β-cholestan-4-one, identical (co-injection; 1% OV-1; 200°C) with an authentic sample.

Conversion of the Hydroxy-ether (23a) *into* 5β-*Cholestan*-4α-*ol*.—The hydroxy-ether (23a) (5 mg) and tosylhydrazine (40 mg) in bis-(2-methoxyethyl) ether (1 ml) were refluxed for 3 h under N_2. The product, obtained as usual, showed g.l.c. (1% OV-1; 225 °C) peaks at r.i. 2 965 and 3 045 (1 : 9). This was converted into the toluene-*p*-sulphonate, which was fragmented, as above, by treatment with NaI–Zn in dimethoxyethane at reflux. The product (2 mg), purified by p.l.c., gave 5β-cholestan-4α-ol, $\nu_{max.}$ 3 635, 3 619, 1 215, 1 170, 1 005, 968, 945, 940, and 900 cm^{-1}; g.l.c. (1% OV-1; 225 °C) r.i. 2 910, identical (co-injection) with an authentic sample. Authentic samples of 5β-cholestan-4α- and 4β-ols were prepared by reduction with lithium aluminium hydride of 5β-cholestan-4-one.

Allylic Oxidation of the Olefinic Ester (16).—(i) The ester (16) (22 mg, 0.053 mmol), *N*-bromosuccinimide (50 mg), and calcium carbonate (20 mg) in dioxan (2 ml) containing water (2 drops) were irradiated for 4 h at 20 °C with a 60 W lamp, and the product was isolated with ether. P.l.c. (ethyl acetate–light petroleum, 1 : 3) afforded *methyl* 5-*bromo*-6-*oxo*-4,5-*secocholestane*-4-*carboxylate* (41) (10 mg), $\nu_{max.}$ 1 740, 1 720, 1 275, 1 255, 1 240, 1 190, 1 160, and 925 cm^{-1}; δ 3.90 (1 H, s), 3.58 (3 H, s), 2.24 (2 H, d, *J* 6 Hz), M^+ 512 and 510 ($C_{28}H_{47}BrO_3$). The second product, isolated by p.l.c., was the corresponding alcohol, oxidised by Jones reagent to the ketone (41).

(ii) The ester (16) (400 mg, 0.96 mmol) and anhydrous sodium chromate (160 mg, 1 mmol) in acetic acid (4 ml) and acetic anhydride (2 ml) were kept at 40 °C for 24 h; more sodium chromate (160 mg) was then added. After 27 h more the product was isolated as usual. P.l.c. (ethyl acetate–light petroleum 1 : 4) gave, apart from starting material (10 mg), *methyl* 7-*oxo*-4,5-*secocholest*-5-*ene*-4-*carboxylate* (26) (58 mg), an oil, $\nu_{max.}$ 3 020, 1 740, 1 680, 1 260, 1 196, and 1 172 cm^{-1}; δ 6.51 (1 H, d, *J* 9 Hz), 5.80 (1 H, d, *J* 10 Hz), 3.62 (3 H, s), and 2.24 (2 H, g.l.c. (1% OV-1; 225 °C) r.i. 3 175 (Found: C, 77.9; H, 10.55. $C_{28}H_{46}O_3$ requires C, 78.1; H, 10.75%). The isomeric 6-*en*-5-*one* (42) (12 mg), just separated from this, had $\nu_{max.}$ 3 020, 1 740, 1 675, 1 270, 1 230, 1 190, 1 170, and 1 105 cm^{-1}; δ 6.79 (1 H, d, *J* 18 Hz further split by 1.5 Hz), 6.23 (1 H, d, *J* 18 Hz further split by 3 Hz), and 3.66 (3 H, s); g.l.c. (1% OV-1; 225 °C) r.i. 3 152 (Found: C, 77.9; H, 10.9. $C_{28}H_{46}O_3$ requires C, 78.1; H, 10.75%).

Preparation and Cyclisation of the Trideuterio-acetal (20b).—*Deuteriolysis of the enone* (26). The enone ester (26) (90 mg), reduced with $LiAlD_4$–$AlCl_3$, as described elsewhere,[32] afforded 4,4,7,7-*tetradeuterio*-4,5-*secocholest*-5-*en*-4-*ol* (28b). (70 mg), $\nu_{max.}$ 3 640, 3 550—3 150, 3 010, 2 180, 2 085, 1 640, 1 165, 1 110, 1 090, 950, and 885 cm^{-1}; δ 5.49 (2 H, q, lines at ±7 and ±21 Hz), M^+ 392 ($C_{27}H_{44}D_4O$) (<95% D_4 by comparison with unlabelled alcohol).

Oxidation, as described above for the unlabelled alcohol, gave the *trideuterio-aldehyde* (21b), M^+ 389 ($C_{27}H_{43}D_3O$) (<95% D_3 by comparison with unlabelled aldehyde).

Acetalisation, as previously, afforded the *trideuterio-acetal* (20b), $\nu_{max.}$ 3 010, 2 168, 2 085, 1 645, 1 265, 1 215, 1 200, 1 170, 1 060, and 890 cm^{-1}; δ 5.46 (2 H, q, lines at ±3 and ±21 Hz) and 3.91 (4 H, m); M^+ 433 ($C_{29}H_{47}D_3O_2$) (<95% D_3 by comparison with unlabelled acetal).

Cyclisation of the acetal (80 mg), as before, afforded the *dideuterio-ether* (23b) (13 mg), a gum, $\nu_{max.}$ 3 530, 3 012, 2 235, 2 110, 1 635, 1 290, 1 260, 1 160, 1 130, 1 120, 1 095, 1 055, and 895 cm^{-1}; δ 5.57 (1 H, d, *J* 3 Hz), 3.64 (4 H, s), and 2.56 (1 H, exchangeable with D_2O); M^+ 432 ($C_{29}H_{48}D_2O_2$) (<95% D_2 by comparison with unlabelled material). The more polar *trideuterio-alcohol* [4α,7,7-trideuterio- (25)] (11 mg) had $\nu_{max.}$ 3 625, 3 605, 2 180, 2 135, 2 100, 1 165, 1 090, 1 075, 1 050, 1 045, 1 020, 1 010, and 940 cm^{-1}; δ_{Me} 0.96, 0.89, 0.83, and 0.63; M^+ 391 ($C_{27}H_{45}D_3O$) (<95% D_3).

Synthesis and Deuteriolysis of 4,5-*Secocholest*-5-*ene*-4,7β-*diol*.—Reduction of the enone ester (26) with lithium aluminium hydride under standard conditions and p.l.c. of the product afforded the *diol* (27) (63%), m.p. 121—123°, $\nu_{max.}$ 3 640, 3 010, and 1 660 cm^{-1}; δ 5.49 (2 H, q, lines at ±1 and ±11 Hz), 3.86 (1 H, d, *J* 6 Hz), and 3.63 (2 H, t, *J* 6 Hz); M^+ 404 ($C_{27}H_{48}O_2$).

Deuteriolysis of this diol, as described elsewhere,[32] afforded 7α-*deuterio*-4,5-*secocholest*-5-*en*-4-*ol* (28c), $\nu_{max.}$ 3 638, 3 008, 2 155, 2 115, 1 650, and 1 120 cm^{-1}; δ 5.49 (2 H, q, lines at ±8 and ±18 Hz) and 3.84 (2 H, t, *J* 6 Hz), M^+ 389 ($C_{27}H_{47}DO$) (<98% D_1).

Attempted Photo-oxygenation of the Olefinic Acetal (20).—Exposure of the acetal (20) (50 mg) and hematoporphyrin (4 mg) in dry pyridine (5 ml) to a 60 W lamp or a high-pressure Hanovia u.v. lamp during passage of O_2 for 24 h led to no reaction.

7α- *and* 7β-*Deuteriocholesterol*.—These were prepared as detailed elsewhere.[34] By the analytical procedures there described, 7α-deuteriocholesterol (>97% D_1) had 82.5% 7α-D, and 17.5% 7β-D; 7β-deuteriocholesterol (>97% D_1) had 97% β-D and 3% α-D.

The 7β-*Deuterio-acetal* (20d).—Prepared from the above 7β-deuteriocholesterol by the route described for the unlabelled acetal, this had $\nu_{max.}$ 3 010, 2 155, 1 650, 1 140, 1 130, 1 050, 1 035, and 940 cm^{-1}, δ 5.46 (2 H, q, lines at

± 7 and ± 17 Hz; low-field doublet $W_{\frac{1}{2}}$ 2 Hz, up-field doublet $W_{\frac{1}{2}}$ 3 Hz), 4.82 (1 H, t, J 4 Hz), and 3.88 (4 H, m); M^+ 431 ($C_{29}H_{49}DO_2$).

Cyclisation of the 7β-*Deuterio-acetal* (20d).—Under the conditions described for the unlabelled acetal, cyclisation of the 7β-deuterio-acetal (20d) afforded the deuterio-hydroxy-olefin (23c), $\nu_{max.}$ 3 540, 3 010, 2 230, 1 630, 1 150, 1 095, 1 050, 955, and 900 cm^{-1}; δ 5.54 (1 H, d, J 4 Hz), 3.60 (4 H, m), 3.44 (1 H, s), and 2.55 (1 H, s, exchangeable with D_2O); M^+ 431 ($C_{29}H_{49}DO_2$) (< 96% D_1 by comparison with unlabelled material).

Brosylation of the Olefinic Alcohol (28a).—The alcohol (28a) (80 mg, 0.21 mmol) and 4-bromobenzenesulphonyl chloride (100 mg, 0.39 mmol) in dry pyridine (3 ml) were kept at 0 °C for 12 h. The *brosylate* (29a) (111 mg), purified by p.l.c. (ethyl acetate–light petroleum, 1 : 9), a gum, had $\nu_{max.}$ 3 010, 1 820, 1 650, 1 270, 1 185, 1 175, 1 095, 1 068, 1 010, 960, and 940 cm^{-1}; δ 7.72 (4 H, s) and 5.41 (2 H, m, lines at ± 11 and ± 21 Hz, low-field lines further split by 5 Hz).

Solvolysis of the Brosylate (29a).—(i) The brosylate (29a) (22 mg) and urea (4 mg) were refluxed in 2,2,2-trifluoroethanol for 72 h under N_2. P.l.c. afforded a sample (3 mg) of hydrocarbon, g.l.c. (1% OV-1/225°) t_R 4.6 and 5.5 min (8 : 1) (cholestane 5.5, 5α-cholest-6-ene 5.5, 5β-cholest-6-ene 4.65 min).

(ii) The brosylate (42 mg) and urea (8 mg) in trifluoroethanol (5 ml) were kept in a sealed glass tube at 105 ± 5 °C for 40 h. The hydrocarbon (4 mg) product gave g.l.c. peaks (above conditions) at t_R 2.5, 3.2, and 4.6 min (1 : 1.5 : 16). The longest retained component was identified as 5β-cholest-6-ene by co-injection and g.l.c.–mass spectrometric comparison with authentic material.

The 7α-*Deuterio-brosylate* (29c).—The sample of 7α-deuteriocholesterol was converted into 7α-*deuterio*-4,5-*seco-cholest-5-en*-4-*ol* (28c) as described above; δ 5.42 (2 H, m, lines at ± 10 and ± 20 Hz, low-field doublet further split by 5 Hz); M^+ 389 ($C_{27}H_{47}DO$) (< 97% D_1). The 7α-deuterio-brosylate was obtained as above.

Solvolysis of the 7α-*Deuterio-brosylate* (29c).—The 7α-deuterio-brosylate (29c) (30 mg) and urea (8 mg) were kept in 2,2,2-trifluoroethanol (3.5 ml) at 105 ± 5° for 40 h. The hydrocarbon product, obtained as before, was entirely 5β-cholest-6-ene (g.l.c. co-injection). G.l.c.–mass spectrometry gave peaks at *m/e* 370 (M^+, 100%) 355 (42), 301 (8), 274 (60), 257 (66), 247 (26), and 215 (66); isotopic ratio of *m/e* 370 to *m/e* 371, 79 : 39 (average of 5 scans); relative size of D_1 contribution = 39 − (29.7 × 79)/100 = 15.5; hence D_0/D_1 = 79/15.5, and D_1 = 16.5%. Authentic 5β-cholest-6-ene had *m/e* 370 (M^+, 100%), 355 (44), 301 (6), 274 (62), 257 (68), 247 (30), and 215 (62).

[5/801 *Received*, 29*th April*, 1975]