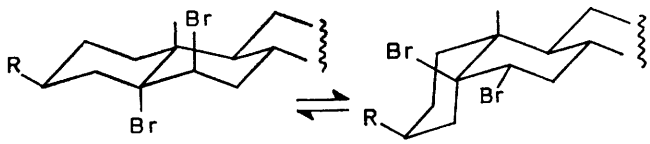
Equilibration of 24,25-Dibromides of Lanosterol and Derivatives

By **D. H. R. Barton, H. MacGrillen,** and **P. D. Magnus,** Department of Chemistry, Imperial College, London SW7 2AY
C. H. Carlisle * and **P. A. Timmins,** Department of Crystallography, Birkbeck College, London WC1E 7HX

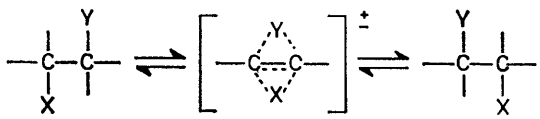
Bromination of lanosteryl acetate under conditions of kinetic control gives two 24,25-dibromides, A and B, in equal amounts. Thermodynamically controlled equilibration of these dibromides results in a 5:1 mixture of the two. The driving force is considered to be interaction between the 20-methyl group and the 24-bromine atom in the 24*R*-configuration. Configurations have been assigned to the dibromides from the results of an *X*-ray crystal structure determination on dibromide A.

THE diaxial–diequatorial rearrangement of cholestene 5α,6β-dibromides to give 5β,6α-dibromides (Scheme 1) has been studied extensively.[1] The driving force is the



SCHEME 1

relief of 1,3-diaxial interactions in going to the di-equatorial dibromide. The position of equilibrium varies to a first approximation with the size of the group



SCHEME 2

R at C-3. When R = H the equilibrium is >97% (at 40°) in favour of the diequatorial dibromide. The related rearrangements of 2β-bromo-3α-chloro- and of 3α-bromo-2β-chloro-cholestane proceed in the same fashion, the driving force for the rearrangement being clearly of conformational origin. The generalised diaxial–diequatorial rearrangement[2] is summarised in Scheme 2.

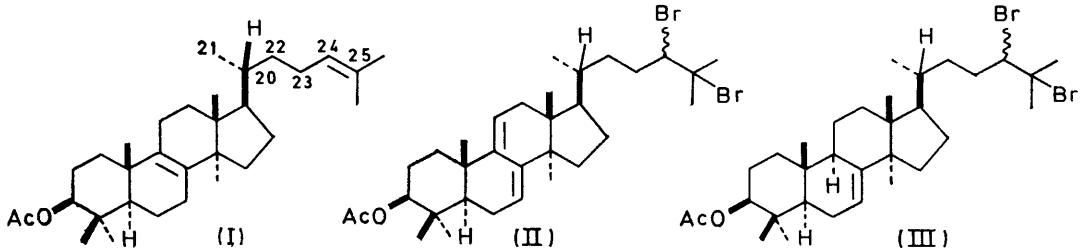
Bromination of lanosteryl acetate (I) is reported to give two 24,25-dibromides designated:[3] A (less soluble in acetic acid–ether), m.p. 168—169°, $[\alpha]_D^{25}$ +44° (*c* 1·1 in chloroform), and B (more soluble in acetic acid–ether), m.p. 176—177°, $[\alpha]_D^{25}$ +7·6° (*c* 0·9 in chloroform). It would be expected that an isolated double bond flanked by achiral centres, in the absence of any directing functions, would not demonstrate any conformational preference in bromination. Thus the bromination of lanosteryl acetate would be expected to give, under kinetic control, approximately equal amounts of dibromides A and B. Furthermore, in equilibration of dibromides A and B (thermodynamic control) it would not be expected that one dibromide would predominate over the other because of any conformational preference.

TABLE 1

Bromination of lanosteryl acetate in chloroform

Temp. (°C)	$[\alpha]_D^{25}$ (°)	A (%)	B (%)
0	+29·0	55·0	45·0
−5	+29·2	55·3	44·7
−10	+28·8	54·3	45·7
	+27·0	49·8	50·2
−20	+27·8	51·8	48·2
	+29·0	54·8	45·2
−55	+27·2	50·0	49·9
	+29·0	54·8	45·2

Mean % A 53·4. Mean % B 46·6.



Lanosteryl acetate was treated with bromine in chloroform at a variety of temperatures (0 to −55°) and the rotation of the completely brominated product was measured (Table 1). The reaction consistently gave a mixture (*ca.* 53 : 47) of lanosteryl acetate 24,25-dibromides A and B, as expected from random attack from

[1] (*a*) D. H. R. Barton and A. J. Head, *J. Chem. Soc.*, 1956, 932; (*b*) C. Grob and S. Winstein, *Helv. Chim. Acta*, 1952, **35**, 782; (*c*) G. H. Alt and D. H. R. Barton, *J. Chem. Soc.*, 1954, 4284; (*d*) D. H. R. Barton and E. Miller, *J. Amer. Chem. Soc.*, 1950, **72**, 1066.

[2] D. H. R. Barton and J. F. King, *J. Chem. Soc.*, 1958, 4398.

[3] D. A. Lewis and J. F. McGhie, *Chem. and Ind.*, 1956, 550.

either side of the 24,25-double bond. Epoxidation of the 24,25-double bond gives equal amounts of the two possible epoxides.*

Pure lanosteryl acetate 24,25-dibromide B was heated in refluxing chloroform and the rate of conversion into dibromide A was measured (see Experimental section). The reaction was strictly first-order at the temperatures and concentrations studied. No acid catalysis was detected. The results are given in Table 2.

TABLE 2

Rate of conversion of dibromide B into dibromide A

Temp. (°C)	Trichloroacetic acid (mol. equiv.)	$10^5k/s^{-1}$ [a]
42		0·501
56		2·69
62		6·31
62	0·308	6·34
62	0·616	6·29
62	0·924	6·27
62	1·232	6·32

$\Delta G^{\ddagger} = 25{\cdot}5$ kcal mol^{-1}

[a] Average of three runs. Values are accurate to ±2%.

The equilibrium constants, determined in the usual manner, are given in Table 3. For lanosteryl acetate dibromides form A is thermodynamically by far the more stable (equilibrium ratio 5·2 : 1). Agnosteryl acetate dibromides A and B (II) and lanosta-7,24-dien-3-yl acetate dibromides A and B (III) also equilibriate to give the thermodynamically more stable A form (equilibrium ratio *ca.* 5 : 1).

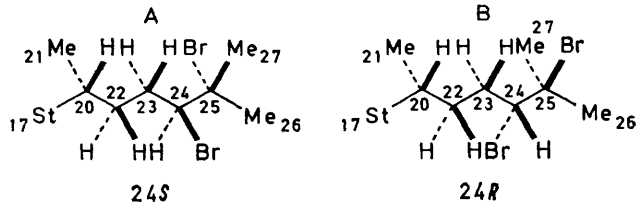
The pronounced thermodynamic stability of dibromide A over B indicates a strong long-range conformational effect that provides the driving force for the formation of the former.

TABLE 3

Equilibrium constants for the rearrangement of lanosteryl acetate dibromides, agnosteryl acetate dibromides, and lanosta-7,24-dien-3-yl acetate dibromides

Starting material		$K = [A]_{equil}/[B]_{equil}$
Lanosteryl acetate dibromide	A	5·22
	B	5·12
Lanosteryl acetate dibromide	A, B 4 : 1	5·12
Lanosteryl acetate dibromide	A, B 9 : 1	5·08
Agnosteryl acetate dibromide	A	4·97
	B	4·85
Lanosta-7,24-dien-3-yl acetate dibromide	A	4·94
	B	4·86

X-Ray crystallographic analysis (see later) of lanosteryl acetate dibromide A demonstrated that the C(17)–C(20)–C(22—26) chain is fully extended and nearly planar (Scheme 3). The molecule has the *S*-configuration at C-24. Consequently, the configuration at C-24 of dibromide B, if Scheme 2 is accepted, must be *R*. A model of structure B demonstrates the possibility of interaction between the 20-methyl group and the 24-bromine atom that does not exist in structure A. This appears to be the only interaction that can explain the driving force for the formation of A. Presumably the situation in the agnosterol and lanosta-7,24-dienol cases is the same. Such a conformational preference, in a site well removed from the usual steric interactions,



SCHEME 3

would have been difficult to predict. But our results clearly demonstrate its existence and ascribe it to the 20-methyl group.

With regard to the details of the *X*-ray determination, the crystal structure of lanosteryl acetate 24,25-dibromide A is substantially the same as that found in lanosterol iodoacetate.[4] At the present stage of refinement there appears to be some disorder in the structure, as indicated by the large anisotropic thermal vibrations of the bromine atoms. All bond lengths and angles lie within expected limits and the average carbon–carbon single bond length is 1·545 Å. The packing of the molecules in the unit cell is interesting in that it gives a very open structure with virtually no overlap of molecules in the *b*-axis projection, as shown in the Figure. The unit cell dimensions and molecular shape are in general agreement with those found in similar structures.[5] A list of observed structure factors is available on request.

The *X*-ray structure of 24,25-dibromokulactone has recently been reported and the configuration at C-24 assigned as *S*.[6]

EXPERIMENTAL

Chloroform was distilled from phosphorus pentoxide prior to use. All $[\alpha]_D$ values were determined for solutions in chloroform (*c ca.* 1·0).

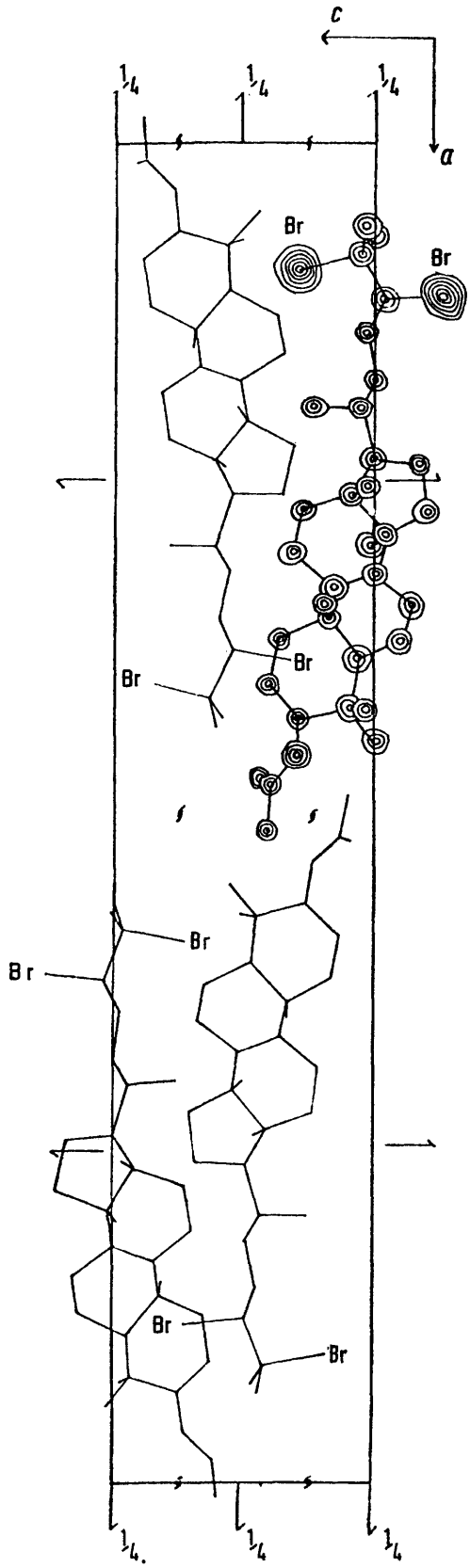
Lanosteryl acetate dibromide B was crystallised from chloroform–methanol; m.p. 176—177°, $[\alpha]_D^{25}$ +7·6° (*c* 0·9). Lanosteryl acetate dibromide A was prepared by heating dibromide B in chloroform at reflux for 12 h and purified by crystallisation; m.p. 168—169° (from chloroform–methanol), $[\alpha]_D^{25}$ +44° (*c* 1·1). Agnosteryl acetate dibromides [3] A and B (II) were supplied by Dr. U. Kempe and purified similarly: A, m.p. 179—180°, $[\alpha]_D^{25}$ +65°; B, m.p. 193—195°, $[\alpha]_D$ +38°.

* We thank Professor J. F. McGhie and Dr. R. B. Boar, Chelsea College of Science and Technology, for this information.

[4] J. Fridrichsons and A. McL. Mathieson, *J. Chem. Soc.*, 1953, 2159.

[5] J. D. Bernal, D. Crowfoot, I. Fankuchen, *Phil. Trans.*, 1940, *A* **239**, 135.

[6] K. W. Ma, F. C. Chang, and J. C. Clardy, *Chem. Comm.*, 1971, 424.



Packing of molecules in the unit cell; *b*-axis projection

Lanosta-7,24-dien-3-yl acetate 24,25-dibromide B was obtained by passing dry hydrogen chloride gas into lanosteryl dibromide at 0° for 3 h. Work-up in the usual way gave the *dibromide* B (III), m.p. 171—171·5° (from chloroform–methanol), $[\alpha]_D^{25}$ −7·0° (Found: C, 61·0; H, 8·2; Br, 25·7. $C_{32}H_{52}Br_2O_2$ requires C, 61·2; H, 8·3; Br, 25·5%).

Lanosta-7,24-dien-3-yl acetate 24,25-dibromide A was prepared either by heating dibromide B in chloroform or by acid-catalysed rearrangement of the corresponding Δ^8-isomer. It had m.p. 161—162° (from chloroform–methanol), $[\alpha]_D^{25}$ +31·6° (Found: C, 61·4; H, 8·3; Br, 25·7. $C_{32}H_{52}Br_2O_2$ requires C, 61·2; H, 8·3; Br, 25·5%).

Treatment of the Δ^7-dibromide A or B (III) with zinc in glacial acetic acid followed by catalytic hydrogenation (Adams catalyst) gave the known [7] 3β-acetoxylanost-7-ene, identified by m.p., $[\alpha]_D$ value, and n.m.r. spectrum.

Kinetics.—To obtain a constant reaction temperature a two-necked flask equipped with condenser was suspended in another flask partially filled with an appropriate solvent. By this means, the reaction flask could be bathed in the vapour of the refluxing liquid. To the reaction flask were added lanosteryl acetate dibromide B (0·03—0·07 mol) and chloroform (25 ml). The second neck of the flask was then closed with a neoprene serum cap through which samples were withdrawn by syringe at 30 min intervals. The samples were rapidly quenched to room temperature and their specific rotations determined (Table 2).

Equilibrium Constants.—Equilibrium constants were measured with the apparatus described in the previous section for 0·03—0·07M-solutions of the substrate in chloroform. The reactions were allowed to proceed for *ca.* 12—15 half-lives (*ca.* 99·8% approach to equilibrium). The solvent was then removed *in vacuo* at 0°. The residue was weighed and made up to a known concentration in chloroform, and the rotation was determined (Table 3).

Crystal and Molecular Structure of Lanosteryl Acetate 24,25-Dibromide A.—Crystal data. $C_{32}H_{52}Br_2O_2$, M = 627·8, Orthorhombic, a = 40·893 ± 0·006, b = 9·990 ± 0·002, c = 7·839 ± 0·002 Å, U = 3203 Å^3, D_m = 1·306, Z = 4, D_c = 1·299, $F(000)$ = 1320. Space group $P2_12_12_1$, μ(Cu-K_α) = 36·0.

Crystallographic measurements. Most of the crystals were large, fragmented, and opaque, but there were, in a batch recrystallised from 50% methanol–benzene, a few crystals which were suitable for *X*-ray structure analysis.

A diamond-shaped plate of dimensions *ca.* 0·8 × 0·6 × 0·2 mm was examined under a polarising microscope and found to be a single crystal. By permission of Prof. D. Rogers intensity data from this crystal were collected with the Siemens four-circle diffractometer at Imperial College, London. It was noted from Weissenberg photographs that the recorded data diminished to zero intensity at θ = 55°; thus no data were collected beyond this θ value. After the collection of about 1800 reflections the intensity of a reference reflection had diminished by about 50%. The last 600 reflection were therefore discarded and only the first 1200 considered, after which the reference reflection intensity had fallen by about 20%. A second, similar crystal was mounted and another 1168 reflections were measured. The apparent decrease in intensity of the reference reflection was assumed to be due to radiation damage; other crystals, which had not been irradiated, did

[7] J. F. Cavella, J. F. McGhie, and M. K. Pradhan, *J. Chem. Soc.*, 1951, 3142.

not appear to deteriorate over a period of several months. To allow for this effect the intensities in each block of 25 reflections were scaled to the average value of the reference reflection at the beginning and end of each block. The data for the two crystals were also scaled relative to this reference reflection and were finally corrected for Lorentz and polarisation factors by use of the *X*-Ray '63 system of programs.

Structure determination. A sharpened Patterson synthesis was calculated and from this the bromine atom co-ordinates were unambiguously determined. Both bromine atoms were found to lie on the plane $y = 0$ with one of the z co-ordinates almost equal to 0·25. This arrangement of heavy atoms in space group $P2_12_12_1$ gives rise to a false centre of symmetry in any Fourier synthesis calculated by use of only the phases derived from the heavy atom model, therefore producing the true structure and its enantiomorph. A partial Fourier synthesis phased on the bromine positions and weighted by use of the scheme due to Sim [8] was calculated, and with the help of a model based on the known configuration of lanosterol, ring A of the correct enantiomorph was selected (Figure).

All atoms except C-24, C-25, and C-29 were clearly visible in the first few Fouriers but C-24 and C-25 lay within an elongated strip of electron density and could not be located with any great accuracy. Carbon atom 29 was located from a difference Fourier after several cycles of least-squares refinement.

Structure refinement.—The least-squares refinement was commenced with $R = 0{\cdot}45$; *ca.* ten cycles of full matrix calculations have reduced this to a present value of 0·153. No absorption correction has yet been applied. The parameters refined to date are: all positional parameters, anisotropic thermal parameters for the bromine atoms, and isotropic thermal parameters for the other non-hydrogen atoms.

We thank Mr. D. Williams, Department of Chemical Crystallography, Imperial College, London, for his help in collecting the data and in its subsequent processing. We also thank Professor D. W. J. Cruickshank and Dr. J. G. F. Smith for permission to use their SFLS program for structure refinement, Dr. D. S. Moss for his modification and help in the use of this program, and Dr. P. F. Lindley for help with computing throughout the course of the structure determination. We thank Mrs. Diana Knight for the determination of kinetically-controlled dibromide ratios and Drs. P. G. Sammes and J. P. Poyser for preparing crystal specimens. One of us (H. MacG.) acknowledges the award of a Research Studentship by the Ministry of Education, Northern Ireland. One of us (P. A. T.) is in receipt of a grant from Birkbeck College, London.

[1/2467 *Received*, 23*rd December*, 1971]

[8] G. A. Sim, *Acta Cryst.*, 1960, **13**, 511.