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Crystal Structure and Synthesis of 5α,14β-Cholest-7-ene-3β,15β-diol Di-*p*-bromobenzoate

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5α,14β-Cholest-7-ene-3β,15β-diol di-*p*-bromobenzoate has been synthesized from 5α,14β-cholest-7-ene-3β,15β-diol and the crystal structure determined. $C_{41}H_{52}O_4Br_2$, monoclinic, space group $P2_1$; $a = 14{\cdot}12$ (2), $b = 7{\cdot}46$ (1), $c = 18{\cdot}53$ (4) Å, $\beta = 97{\cdot}08$ (6)°; $Z = 2$, $M_r = 768{\cdot}7$, $D_c = 1{\cdot}33$, $D_m = 1{\cdot}32$ g cm^{-3}. The structure was solved by the heavy-atom method and refined by full-matrix anisotropic least squares to a final R of 0·064. This structure analysis was undertaken to establish the configuration of the O substituent of C(15) and to determine the absolute stereochemistry of the *C–D* ring junction of the steroid nucleus. The configuration of the 15-hydroxyl function is β. In addition, the steroid nucleus has two distinctive features: a *cis C–D* ring junction and the *C* ring in a twist conformation.

Introduction

Barton & Laws (1954) reported that boron trifluoride–ether treatment of the reaction mixture obtained by the action of perphthalic acid upon 3β-acetoxyergosta-7,14,22-triene yielded 3β-acetoxyergosta-7,22-dien-15-one [configuration at C(14) not specified]. Our recent success in the isolation and characterization of the product of the action of *m*-chloroperbenzoic acid on 3β-benzoyloxy-5α-cholesta-7,14-diene as 3β-benzoyloxy-14α,15α-epoxy-5α-cholest-7-ene (Parish, Spike & Schroepfer, 1977; Conner, Parish, Schroepfer & Quiocho, 1977) has permitted the demonstration that treatment of the 14α,15α-epoxy-Δ^7-steryl ester with boron trifluoride–ether yields 3β-benzoyloxy-5α,14β-cholest-7-en-15-one (Parish, Newcomer, Gilliland, Quiocho & Schroepfer, 1976; Parish & Schroepfer, 1977). Reduction of the latter compound with lithium aluminum hydride yielded 5α,14β-cholest-7-ene-3β,15β-diol and 5α,14β-cholest-7-ene-3β,15α-diol, both of which have been found to be potent inhibitors of sterol biosynthesis (Schroepfer, Parish, Chen & Kandutsch, 1976).

Presented herein are the chemical synthesis and a crystal structure determination of the di-*p*-bromobenzoate derivative of 5α,14β-cholest-7-ene-3β,15β-diol. This report constitutes the first X-ray crystallographic analysis of a Δ^7-sterol derivative with the 'unnatural' *cis C–D* ring junction. A preliminary account of a portion of this work has been published (Parish *et al.*, 1976).

Experimental

Synthesis

p-Bromobenzoyl chloride (2·0 g; 9·11 mmol) was added to a solution of 5α,14β-cholest-7-ene-3β,15β-diol (0·5 g; 1·24 mmol), prepared as described previously (Parish *et al.*, 1976), in dry pyridine (150 ml). After gentle warming to dissolve the acid chloride, the reaction mixture was allowed to stand at room temperature for 36 h under an atmosphere of nitrogen. The mixture was poured into water and thoroughly extracted with ether containing methylene chloride (5%). The combined extracts were washed successively with water, cold aqueous 5% hydrochloric acid, and water and dried over anhydrous magnesium sulfate. The residue obtained upon evaporation of the solvent was chromatographed on a silica gel (60–200 mesh) column (2·0 × 100 cm) with benzene as the eluting solvent (1·5 ml min^{-1}; 24 ml per fraction). The contents

Table 1. *Spectroscopic data on 5α,14β-cholest-7-ene-3β,15β-diol di-p-bromobenzoate*

Infrared (ν_{max}^{KBr}; cm^{-1}):* 1793, 1725, 1593, 1281

Nuclear magnetic resonance (δ; p.p.m.):* 1·20 (*m*, methylene envelope), 4·95 [*m*, 2H, H(C3) and H(C15)], 5·40 [*m*, 1H, H(C7)], 7·85 (*m* 8H, aromatic)

Mass (*m/e*):* 570 and 568 (7% and 6%; *M*-bromobenzoic acid), 455 and 453 (8% and 6%; *M*-bromobenzoic acid–side chain), 441 and 439 (3% and 2%; *M*-bromobenzoic acid–side chain–CH_3), 366 (3%; *M*-bromobenzoic acid–bromobenzoic acid), 351 (19%; *M*-bromobenzoic acid–bromobenzoic acid–CH_3), 253 (24%; *M*-bromobenzoic acid–bromobenzoic acid–side chain), 239 (11%), 238 (8%), 227 (4%), 225 (4%), 215 (3%), 213 (6%), 211 (9%), 202 (14%), 200 (17%), 198 (9%), 196 (2%), 185 (100%), 183 (88%)

* Recorded as described previously (Knapp, Wilson & Schroepfer, 1976).

of fractions 15 through 25 were pooled and, after evaporation of the solvent under reduced pressure, crystallized from acetone–water to give the di-*p*-bromobenzoate of 5α,14β-cholest-7-ene-3β,15β-diol (0·86 g; 88% yield) melting at 175–176°C (spectral data presented in Table 1). The compound showed a single component on thin-layer chromatographic analyses on silica gel *G* plates (solvent systems, benzene and 10% ether in hexane). The component was recrystallized from a 1 : 1 mixture of methylene chloride and ethanol. The resulting prismatic crystals (elongated along the *b* axis) were dried in a vacuum desiccator for 24 h. The density was determined by flotation in sodium bromide solution.

Structure analysis

Precession photographs using Ni-filtered Cu $K\alpha$ radiation indicated space group $P2_1$ (systematic extinctions, $0k0$ for $k = 2n + 1$). The unit-cell parameters are given in Table 2. In order to minimize radiation damage, the crystals were mounted in vacuum-sealed glass capillaries. A Syntex $P2_1$ diffractometer with Ni-filtered Cu $K\alpha$ radiation was used in the 2θ–θ scan mode (range 2 to 25° min^{-1}) to collect a total of 5799 reflections ($\sin\theta/\lambda = 0{\cdot}6$ or $d_{min} =$ 1 Å). Five monitor reflections were checked every 100 reflections. Because of a slight decay of the monitor reflections, three crystals were required in the data collection; at no time did the decrease in intensity of the monitor exceed 6% for any of the crystals. Of the total number of reflections, 431 were rejected because of unequal background and 49 were given zero weight because of poor agreement between intensities of symmetry-related reflections. An additional 275 reflections were collected as overlaps to correlate the three crystal data sets by a linearized least-squares method (Rae, 1965). The total number of unique reflections was 2215. The final correlation R value ($R = \Sigma_h\ |s_i I_{hi} - s_j I_{hj}|/\Sigma_h\ |s_i I_{hi} + s_j I_{hj}|$; s = scale factor, i,j = data sets) was 0·04.

Table 2. *Crystal data*

$C_{41}H_{52}O_4Br_2$, $M_r = 768{\cdot}7$
$a = 14{\cdot}12\ (2)$ Å
$b = 7{\cdot}46\ (1)$
$c = 18{\cdot}53\ (4)$
$\beta = 97{\cdot}08\ (6)°$
$V = 1927\ (6)$ Å^3

Radiation: Cu $K\alpha$
Space group: $P2_1$
$D_c = 1{\cdot}329$ g cm^{-3}
$D_m = 1{\cdot}319$
$\mu = 47{\cdot}57$ cm^{-1}

The non-hydrogen atoms were located by Patterson and Fourier methods. The structure was refined by a least-squares method using diagonal, block, and full matrices. The function minimized was $\Sigma\, w(k^2|F_o|^2 - |F_c|^2)^2$. A weighting scheme suggested by Hughes (1941) was used: $w = 1/F_o$ for $F_o > 4(F_o)$min and, for $F_o < 4(F_o)$min, $w^{1/2} = 1/[(F_o)4(F_o)\text{min}]^{1/2}$. Prior to the final cycles of refinement, the Br and O atoms were refined with anisotropic temperature factors. Before application of anisotropic temperature factors of the C atoms, the H atom positions were calculated or found from a difference synthesis (see Table 4). The H atoms were given isotropic temperature factors corresponding to the value estimated from the Wilson plot. The C atoms were then refined with anisotropic temperature factors, the H positional and thermal parameters being fixed. Iteration of a refinement and readjustment of the H atom positions obtained convergence in four cycles. The final R was 0·064 and R_w was 0·016. All crystallographic calculations were made with the

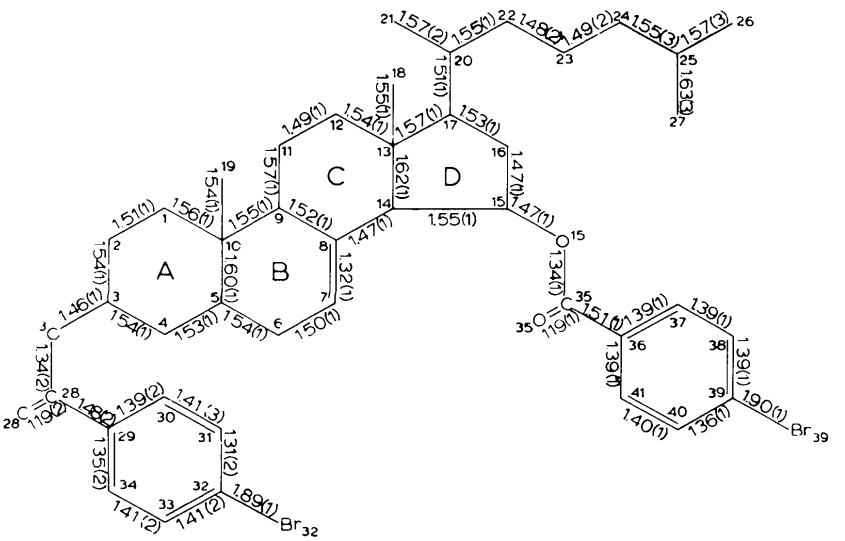

Fig. 1. The numbering scheme and bond distances (Å) for the nonhydrogen atoms of 5α,14β-cholest-7-ene-3β,15β-diol di-*p*-bromobenzoate.

Table 3. *Fractional atomic coordinates* ($\times 10^3$) *and e.s.d.'s for the non-hydrogen atoms*

	x	*y*	*z*
C(1)	224 (1)	739 (2)	380 (1)
C(2)	179 (1)	720 (2)	302 (1)
C(3)	188 (1)	525 (2)	277 (1)
C(4)	142 (1)	393 (2)	326 (1)
C(5)	185 (1)	418 (2)	405 (1)
C(6)	138 (1)	286 (2)	454 (1)
C(7)	167 (1)	326 (2)	533 (1)
C(8)	210 (1)	474 (2)	559 (1)
C(9)	235 (1)	626 (2)	510 (1)
C(10)	176 (1)	619 (2)	434 (1)
C(11)	228 (1)	810 (2)	550 (1)
C(12)	159 (1)	807 (2)	605 (1)
C(13)	189 (1)	680 (2)	669 (1)
C(14)	234 (1)	500 (2)	638 (1)
C(15)	341 (1)	520 (2)	668 (1)
C(16)	341 (1)	604 (2)	740 (1)
C(17)	271 (1)	760 (2)	725 (1)
C(18)	100 (1)	620 (2)	704 (1)
C(19)	71 (1)	670 (2)	437 (1)
C(20)	240 (1)	847 (2)	792 (1)
C(21)	183 (1)	1026 (3)	774 (1)
C(22)	327 (1)	877 (2)	850 (1)
C(23)	304 (1)	926 (3)	923 (1)
C(24)	387 (1)	911 (4)	981 (1)
C(25)	368 (2)	953 (5)	1060 (1)
C(26)	463 (2)	897 (4)	1108 (1)
C(27)	343 (2)	1165 (4)	1067 (1)
C(28)	183 (1)	519 (3)	148 (1)
C(29)	120 (1)	511 (3)	78 (1)
C(30)	163 (1)	527 (5)	15 (1)
C(31)	107 (1)	513 (4)	−53 (1)
C(32)	14 (1)	499 (3)	−57 (1)
C(33)	−33 (1)	494 (3)	6 (1)
C(34)	24 (1)	500 (3)	74 (1)
C(35)	429 (1)	283 (2)	621 (1)
C(36)	478 (1)	108 (2)	642 (1)
C(37)	500 (1)	−5 (2)	587 (1)
C(38)	544 (1)	−169 (2)	603 (1)
C(39)	569 (1)	−212 (2)	676 (1)
C(40)	552 (1)	−104 (2)	732 (1)
C(41)	506 (1)	59 (2)	714 (1)
O(3)	134 (1)	505 (1)	205 (1)
O(15)	386 (1)	343 (1)	677 (1)
O(28)	266 (1)	549 (3)	153 (1)
O(35)	431 (1)	355 (1)	564 (1)
Br(32)	−65 (1)	500 (0)	−147 (1)
Br(39)	634 (1)	−431 (1)	699 (1)

Table 4. *Fractional atomic coordinates* ($\times 10^3$) *for the hydrogen atoms*

Hydrogen atoms located by the difference synthesis are indicated by asterisks.

	x	*y*	*z*
H(C1)	219	877	396
H′(C1)	298	703	383
H(C2)	214	807	268
H′(C2)	104	755	299
H(C3)	262	498	279
H(C4)	155	258	309
H′(C4)	67	417	321
H(C5)	260	391	408
H(C6)	160	151	442
H′(C6)	62	296	442
H(C7)	151	227	572
H(C9)	307	610	501
H(C11)	205	910	510
H′(C11)	297	844	576
H(C12)	90	765	579
H′(C12)	154	940	627
H(C14)	205	377	657
H(C15)	379	597	632
H(C16)	411	652	760
H′(C16)	317	511	778
H(C17)	303	875	702
H(C18)*	66	738	726
H′(C18)*	122	525	749
H″(C18)*	49	554	663
H(C19)*	26	609	390
H′(C19)*	63	817	434
H″(C19)*	48	620	488
H(C20)	192	756	813
H(C21)*	122	1027	806
H′(C21)*	157	1027	716
H″(C21)*	228	1144	789
H(C22)	370	982	831
H′(C22)	368	754	855
H(C23)	248	839	937
H′(C23)	279	1063	922
H(C24)	441	1002	968
H′(C24)	413	775	981
H(C25)	309	881	1076
H(C26)*	509	819	1076
H′(C26)*	446	818	1155
H″(C26)*	500	1021	1128
H(C27)*	375	1242	1026
H′(C27)*	372	1213	1122
H″(C27)*	265	1184	1059
H(C30)	239	549	18
H(C31)	141	513	−102
H(C33)	−110	485	3
H(C34)	−10	495	123
H(C37)	483	34	530
H(C38)	559	−262	561
H(C40)	574	−142	789
H(C41)	491	150	756

CRYM system (unpublished program system developed at Caltech and Harvard Universities).

The atomic numbering scheme and bond lengths for the non-hydrogen atoms are shown in Fig. 1.* Table 3 gives the atomic coordinates. Table 4 presents the atomic coordinates for the H atoms. Table 5 lists torsional angles for the steroid nucleus.

The most conspicuous feature of the overall conformation is the *cis C–D* ring junction (Fig. 2). Because of this *cis* junction, the pseudotorsion angle, C(19)–C(10)···C(13)–C(18), is large: 35·9°. This gives an overall apparent twist to the steroid nucleus. The *A–B* ring junction is *trans*, and the *B–C* ring junction is quasi-*trans*. The 3β-*p*-bromobenzoate group

* Lists of structure factors and anisotropic thermal parameters have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 32639 (36 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 13 White Friars, Chester CH1 1NZ, England.

is almost perpendicular to the *A* ring of the steroid nucleus. This conformation is the one commonly observed for 3β-*p*-bromobenzoate substituents of other steroids (Duax, Weeks & Rohrer, 1976).

Ring *A* has a highly symmetrical chair conformation. This is evident from both the mirror plane and twofold asymmetry parameters (Duax & Norton, 1975), ΔC_s and ΔC_2, respectively $\{\Delta C_s = [\Sigma_{i=1}^{m} (\varphi_i + \varphi_i')^2/m]^{1/2}$; $\Delta C_2 = [\Sigma_{i=1}^{m} (\varphi_i - \varphi_i')^2/m]^{1/2}$; φ_i and φ_i' are symmetry-related torsion angles, and m is the number of pairs of symmetry-related torsion angles$\}$. These asymmetry parameters are: $\Delta C_s^1 = 1{\cdot}3$, $\Delta C_s^2 = 1{\cdot}7$, $\Delta C_s^3 = 2{\cdot}5$, $\Delta C_2^{1,2} = 0{\cdot}3$, $\Delta C_2^{1,10} = 2{\cdot}6$, and $\Delta C_2^{2,3} = 2{\cdot}9$. The average of the endocyclic torsion angles for ring *A* is 57·3°.

Ring *B* has a 5α,10β-half-chair conformation induced by the double bond between C(7) and C(8). The only asymmetry parameter is a twofold rotational axis, $\Delta C_2^{5,10} = 3{\cdot}6$. The average of ring *B*'s endocyclic torsion angles is 31·8°.

Ring *C* has a twist conformation, which is unusual for this ring of the steroid nucleus. The twist conformation may be a result of the combination of the *cis C–D* ring junction and the C(7)–C(8) double bond. Of considerable importance in this structural determination was the establishment of the

Table 5. *Torson angles* (°)

Ring *A*	
C(10)–C(1)–C(2)–C(3)	−61
C(1)–C(2)–C(3)–C(4)	57
C(2)–C(3)–C(4)–C(5)	−56
C(3)–C(4)–C(5)–C(10)	56
C(4)–C(5)–C(10)–C(1)	−56
C(2)–C(1)–C(10)–C(5)	58
Ring *B*	
C(10)–C(5)–C(6)–C(7)	−46
C(5)–C(6)–C(7)–C(8)	13
C(6)–C(7)–C(8)–C(9)	1
C(7)–C(8)–C(9)–C(10)	20
C(8)–C(9)–C(10)–C(5)	−50
C(6)–C(5)–C(10)–C(9)	65
Ring *C*	
C(14)–C(8)–C(9)–C(11)	−31
C(8)–C(9)–C(11)–C(12)	−27
C(9)–C(11)–C(12)–C(13)	65
C(11)–C(12)–C(13)–C(14)	−39
C(12)–C(13)–C(14)–C(8)	−18
C(9)–C(8)–C(14)–C(13)	55
Ring *D*	
C(17)–C(13)–C(14)–C(15)	−11
C(13)–C(14)–C(15)–C(16)	36
C(14)–C(15)–C(16)–C(17	−48
C(15)–C(16)–C(17)–C(13)	39
C(14)–C(13)–C(17)–C(16)	−16

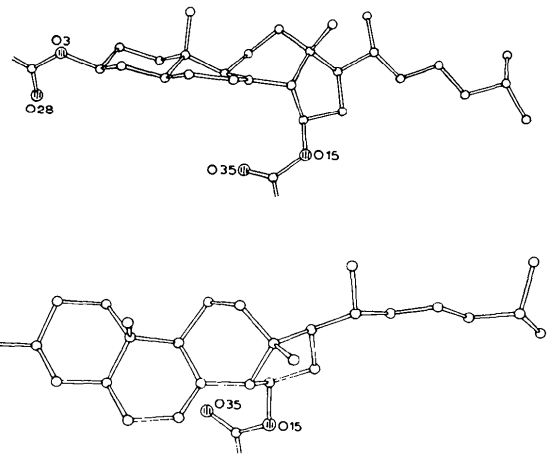

Fig. 2. Two views of the molecule without the *p*-bromophenyl substituents (*ORTEP*).

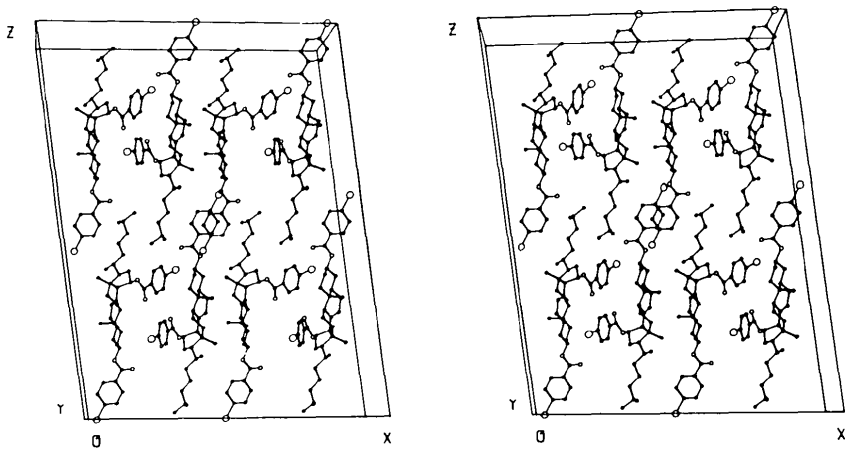

Fig. 3. Crystal packing diagram (*ORTEP*). The dimensions of the box of enclosure are $X = 2a$, $Y = b$, and $Z = 2c$.

configuration of the O substituent at C(15). The results clearly indicate that the O is β. The asymmetry parameters are large, indicating some distortion in the twist structure: $\Delta C_2^9 = 9{\cdot}4$ and $\Delta C_2^{8,14} = 8{\cdot}9$. The average of the endocyclic torsion angles is 38·7°.

Ring *D* has a 15α,16β-half-chair conformation. The asymmetry parameter is small, indicating good correlation: $\Delta C_2^{13} = 2{\cdot}57$. The pseudorotation parameters (Altona, Geise & Romers, 1968) are unusual, $\Delta = -153{\cdot}1°$ and $\varphi = -46{\cdot}6°$. These values are probably a function of the strain induced by the *cis* *C–D* ring structure. The average of the endocyclic torsional angles is 30·7°.

A stereoview of the crystal packing of the molecules is shown in Fig. 3. The unit cell is two molecules thick, one molecule wide and one long. [The length of the steroid is the dimension parallel to the C(10) → C(13) direction and the width is the dimension parallel to the C(14) → C(12) direction. The thickness of the steroid is the dimension orthogonal to the length and the width.] This packing arrangement is frequently observed for steroid structures with two molecules in a unit cell (Duax & Norton, 1975). The width of the steroid is at 27° to the screw axis. The length of the molecule is at 21° to the *c* axis and the thickness at 25° to the *a* axis. The modified Hodgkin notation is $Ma_{25°}b_{27°}c_{21°}$ 211 (Duax & Norton, 1975).

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