Conformational Aspects of Some Ring A α-Acetoxy-ketones in the Cholestane Series

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In contrast to 2α-acetoxy-3-oxo- and 3α-acetoxy-2-oxo-5α-cholestane, which gave n.m.r. and c.d. spectra consistent with a chair conformation of ring A, the corresponding 5α-hydroxy-derivatives have spectral data indicative of twisted conformations of ring A. Conversely, 2β-acetoxy-5-hydroxy-3-oxo-5α-cholestane has ring A in the same boat-like conformation as the corresponding 5-deoxy-compound. 3β-Acetoxy-2-oxo- and 3β-acetoxy-5-hydroxy-2-oxo-5α-cholestane have ring A in the chair conformation.

MILD oxidation of 2β-acetoxy-3β,5-dihydroxy-5α-cholestane (2) [1] afforded the corresponding acetoxy-ketone (15), which is also obtainable by oxidation of the *trans*-triol monoacetate (7).[2] The structure assigned to (15) was confirmed by reduction with sodium borohydride in methanol, at room temperature, to give the starting *cis*-triol monoacetate (2) in *ca.* 70% yield. In contact with silica gel, the acetoxy-ketone (15) was smoothly epimerised at C-2 to give 2α-acetoxy-5-hydroxy-5α-cholestan-3-one (17).[1] Reduction of this compound with borohydride under the same conditions as above took place with almost complete hydrolysis of the acetate group to give a mixture of the triols (9) and (12). Acetylation of the crude mixture afforded the triol monoacetate (10), accompanied by the triol diacetates (11) and (13).

For purposes of comparison the corresponding 5-deoxy-acetoxy-ketones (14) [1,3a] and (16) [3a] were prepared. 2β-Acetoxy-5α-cholestan-3-one (14) was reported [3a] to undergo ready epimerisation at C-2 on treatment with hydrobromic acid or on chromatography over Florisil. In our hands, compound (14) was isomerised in the presence of hydrobromic acid to a 1 : 2 mixture of the acetoxy-ketones (16) and (18), which were separated by chromatography. In contrast to the acetoxy-ketone (15), compound (14) remained unchanged even after prolonged contact with silica gel.

Oxidation of 3β-acetoxy-2β,5-dihydroxy-5α-cholestane (4) [1] with chromium trioxide (Jones reagent) afforded the acetoxy-ketone (19), the structure of which was confirmed by reduction with sodium borohydride to give quantitatively the original triol monoacetate (4). 3α-Acetoxy-5-hydroxy-5α-cholestan-2-one (21) was prepared by acetolysis of 2β,3β-epoxy-5-hydroxy-5α-cholestane to the *trans*-triol monoacetate (8a) [2] and subsequent oxidation (Sarett reagent); in contact with silica gel, it was partially isomerised to the C-3 epimeric compound (19). This isomerisation takes place even in the crystalline form, at room temperature; after a few weeks, compound (21) is partially transformed into compound (19) (t.l.c. evidence). Reduction with sodium borohydride of the acetoxy-ketone (21) was incomplete; however it was accompanied by extensive hydrolysis of the acetate group to yield a complex mixture of products, which was re-acetylated and fractionated by chromatography. Two compounds could be isolated pure, the *trans*-triol diacetate (8b) [4] (*ca.* 10%) and the acetoxy-ketone (19) (*ca.* 30%). According to the n.m.r. spectrum of the crude product, before re-acetylation, the isomerisation of the acetate group took place, at least in part, during treatment of acetoxy-ketone (21) with sodium borohydride. The different behaviour of the acetate groups in compounds (19) and (21) in the slightly alkaline methanolic solution of sodium borohydride, is consistent with previous observations concerning the rates of hydrolysis of 3β- and 3α-acetoxy-5α-hydroxy-steroids.[5]

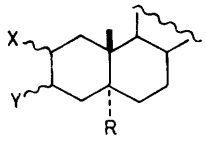
The corresponding 5-deoxy-acetoxy-ketone (20) was

[1] E. Glotter and A. Schwartz, *J.C.S. Perkin I*, 1976, 1660.
[2] T. Komeno, H. Itani, H. Iwagura, and K. Nabeyana, *Chem. and Pharm. Bull.* (*Japan*), 1970, **18**, 1145.
[3] K. L. Williamson and W. S. Johnson, (*a*) *J. Org. Chem.*, 1961, **26**, 4563; (*b*) *J. Amer. Chem. Soc.*, 1961, **83**, 4623.
[4] T. Komeno and H. Itani, *Chem. and Pharm. Bull.* (*Japan*), 1970, **18**, 608.
[5] H. B. Henbest and B. J. Lovell, *J. Chem. Soc.*, 1957, 1965.

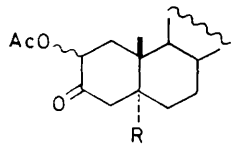
prepared [3a] by oxidation of 3α-acetoxy-2β-hydroxy-5α-cholestane (6). On treatment with hydrobromic acid,[3a] compound (20) was isomerised to a 4 : 1 mixture of 3β-acetoxy-5α-cholestan-2-one (18) and 2α-acetoxy-5α-cholestan-3-one (16). Compound (18) was also obtained by oxidation of the diol monoacetate (3).

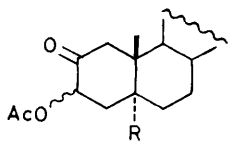
On the basis of the double doublet pattern (Table) of the 2α-H signal in the n.m.r. spectrum of 2β-acetoxy-5α-cholestan-3-one (14), Williamson and Johnson concluded [3b] that the repulsion between the axial 2β-acetoxy-group and the angular 10-methyl group forces ring A into a boat-like conformation in which the acetoxy-group assumes a quasi-equatorial orientation, and does not contribute, therefore, to the deshielding of the 10-methyl group. A similar conformation can be assigned to the corresponding 5α-hydroxy-derivative (15). In both cases, the boat conformation with C-3 and C-10 ' upward,' is supported by the significant upfield shift of the angular methyl signal, entering the shielding region of the carbonyl group [by 0.17 p.p.m. in (14) and 0.21 p.p.m. in (15); comparisons were made with 5α-cholestan-3-one and 5-hydroxy-5α-cholestan-3-one, respectively]. The conformational assignment for compound (15) is supported by the benzene-induced shift [$\Delta(CDCl_3 - C_6D_6)$] (0 p.p.m., as compared to +0.43 p.p.m. in 5-hydroxy-5α-cholestan-3-one).

Although the axial hydroxy-group in 5-hydroxy-5α-cholestan-3-one is in a positive octant, the circular dichroism band of this compound is almost similar to



	R	X	Y		R	X	Y
(1)	H	β-OAc	β-OH	(8a)	OH	β-OAc	α-OAc
(2)	OH	β-OAc	β-OH	(8b)	OH	β-OAc	α-OAc
(3)	H	β-OH	β-OAc	(9)	OH	α-OH	α-OH
(4)	OH	β-OH	β-OAc	(10)	OH	α-OAc	α-OH
(5)	H	β-OAc	α-OH	(11)	OH	α-OAc	α-OAc
(6)	H	β-OH	α-OAc	(12)	OH	α-OH	β-OH
(7)	OH	β-OAc	α-OH	(13)	OH	α-OAc	β-OAc





(14)	R = H	β-OAc	(18)	R = H	β-OAc
(15)	OH	β-OAc	(19)	OH	β-OAc
(16)	H	α-OAc	(20)	H	α-OAc
(17)	OH	α-OAc	(21)	OH	α-OAc

that of 5α-cholestan-3-one.[6] The contribution of 5α-oriented substituents to the chiroptical properties of 3-oxo-steroids has been amply discussed,[6,7] and related to possible conformational changes in ring A of these compounds. According to the pattern of the 2β-H signal and the weak positive c.d. band of the ketone chromophore, 2α-acetoxy-5α-cholestan-3-one (16) should have ring A in a normal chair conformation with the acetoxy-group equatorially oriented. A quite different picture is given by 2α-acetoxy-5-hydroxy-5α-cholestan-3-one (17). The c.d. band is positive; however, much more intense than that of compound (16). (Several c.d. measurements were repeated for solutions in acetonitrile, in order to avoid misinterpretations due to eventual hemiacetal formation in the presence of methanol.) Since the 2α-acetoxy-group in compound (16), as well as the 5α-hydroxy-group in 5-hydroxy-5α-cholestan-3-one, does not appreciably contribute to the intensity of the c.d. band of the 3-ketone, the only reasonable assumption is that ring A in compound (17) underwent a significant conformational change, due to the interaction between the 2α-acetoxy- and the 5α-hydroxy-groups. In our opinion, the c.d. spectrum of compound (17) could be interpreted by assuming ring A in a twisted conformation with C-2 and C-5 ' downwards,' such that the 5α-hydroxy- and the 2α-acetoxy-groups approach each other to hydrogen-bond distance. Such a conformation would also account for the ready hydrolysis of the acetate group during sodium borohydride reduction of (17), in contrast to the reduction of (15) which proceeds without hydrolysis.

According to the octant rule, the contribution of the quasi-axial 2α-acetoxy-group (twisted ring A), should lead to a strong negative and not to a positive c.d. curve. Although it is not a general feature, there are several well documented [8] examples of steroidal α-acetoxy-ketones in which an axial acetoxy-group reverses the expected sign of the Cotton effect (anti-octant contribution). It is well known that in α-hydroxy-ketones, the position of the c.d. band is almost unaffected by an equatorial hydroxy-group, but it is significantly shifted towards longer wavelengths by an axial hydroxy-group. Such effects are not always encountered [9] in α-acetoxy-ketones; we therefore attribute more importance to the intensity and sign of the Cotton effect than to the position of the band ($\lambda_{max.}$ 284 nm) in compound (17).

The twisted conformation assigned above is supported by i.r. measurements in dilute carbon tetrachloride solution ($\nu_{max.}$ 3 535 cm^{-1}), clearly pointing towards an intramolecular hydrogen bonding of the type O–H · · · O. Since previous work [10] has revealed that the stretching of the hydroxy-group in 5-hydroxy-5α-cholestan-3-one is at 3 599 cm^{-1} (the lowering is attributed to a weak O–H · · · π interaction), there remains only the possibility that hydrogen bonding in compound (17) is due to a rather strong interaction between the acetoxy- and hydroxy-groups. This hydrogen bonding may be due to interaction with the lone pair of the singly bonded, or of the doubly bonded oxygen of the acetate group. Of these two possibilities we incline to the former, involving

[6] J. C. Jacquesy and J. Levisalles, *Bull. Soc. chim. France*, 1965, 1538.
[7] H. J. C. Jacobs and E. Havinga, *Tetrahedron*, 1972, **28**, 135.
[8] J. R. Bull and P. R. Enslin, *Tetrahedron*, 1970, **26**, 1525.
[9] See, for instance, J. R. Bull and A. Tuinman *J.C.S. Perkin I*, 1976, 212.
[10] M. Oki, H. Iwamura, J. Aihara, and H. Jida, *Bull. Chem. Soc. Japan*, 1968, **41**, 176.

a quasi-seven-membered-ring structure, with the carbonyl moiety of the acetate group pointing outwards, such as to allow the lone pair of the singly bonded oxygen to be directed towards the hydroxy-group. Intramolecular hydrogen bonding may well involve a quasi-cyclic structure comprising more than five or six atoms. A good example is (25*R*)-11α-hydroxy-2-oxo-5β-spirostan, having the O–H stretching at 3 534 cm^{-1}.[10]

The n.m.r. data of this compound (17) are instructive as well. Although the 2β-H signal appears as a triplet in chloroform solution and as a double doublet in benzene solution, the trend of the benzene induced shift of the 10-methyl signal clearly points to similar conclusions as obtained from c.d. and i.r. measurements. Of all simple ring-A ketones in 5α-steroids, the most important upfield shift [$\Delta(CDCl_3 - C_6D_6)$] of the 10-methyl signal is given by the 3-ketone (+0.37 p.p.m.).[11] The considerable enhancement of this solvent shift in compound (17) (+0.48 p.p.m.) can be accounted for by the suggested conformation of ring A, in which the 3-ketone and the 10-methyl groups are in almost perpendicular dihedral planes.

The n.m.r. data of the acetoxy-ketones (18) and (20) are easily accounted for by assuming a chair conformation of ring A; in the former, the acetate is equatorial (broad multiplet in the n.m.r.), whereas in the latter it is axial (narrow multiplet). Furthermore, the c.d. band of compound (18) is in agreement with the requirements of the octant rule, whereas that of the isomeric compound (20) ($\Delta\varepsilon_{300}$ +1.27), in which the acetoxy-group is axial, can be explained only as the result of an anti-octant contribution.[8] With the exception of the n.m.r. pattern of the 3α-H signal, other spectral data of acetoxy-ketones (18) and (19) are almost similar and do not point to any major conformational differences. Although the 3α-H signal in compound (19) looks like a triplet in chloroform solution, it becomes a double doublet in benzene. As expected, the O–H stretching ($\nu_{max.}$ 3 630 cm^{-1}) indicates a free hydroxy-group.

Conversely, all spectral characteristics of 3α-acetoxy-5-hydroxy-5α-cholestan-2-one (21) point towards a major conformational distortion of ring A. The negative c.d. band of this compound can be accommodated only if ring A is in a twisted conformation, in which the acetoxy-group is quasi-equatorial. This also explains the double doublet pattern of the 3β-H signal. The downfield position of the 10-methyl signal (δ 1.15) points as well to a change in the geometry of ring A. It is noteworthy that the chemical shifts of the 10-methyl signals in compounds (18) and (20) are almost similar to that found in 5α-cholestan-2-one (δ 0.76), whereas the corresponding signal in compound (19) is similar to that found in 5-hydroxy-5α-cholestan-2-one (δ 0.91). The easy isomerisation of this acetoxy-ketone [(21) ⟶ (19)] is an additional proof for its tendency to escape from the unfavourable twisted conformation of ring A, in order to assume the thermodynamically more stable chair conformation with concomitant epimerisation at C-3.

N.m.r. and c.d. data

	N.m.r.[a]			C.d.[b]	
Comp.	2-H	3-H	10-CH_3	$\lambda_{max.}$	Δε
(14)[c]	5.37 dd (9.5;7.5)		0.85	284	+0.99
(15)	5.77 dd (10;5)		0.95 {0.95}	278	+0.63
(16)[c]	5.30 dd (13;6.5)		1.12	288 [288]	+0.46 [+1.22]
(17)	5.33 t (10)		1.28	284 [285]	+2.25 [+2.10]
	{5.27 dd (12;8)}		{0.80}		
(18)[c]		5.22 m ($W_{\frac{1}{2}}$ 22)	0.77	289	+3.20 (lit.[8] +3.2)
(19)		5.64 t (10)	0.90	289 [289]	+2.06 [+2.62]
		{5.81 dd (11;9)}	0.55		
(20)[c]		4.88 m ($W_{\frac{1}{2}}$ 7)	0.78	300	+1.27 (lit.[8] +1.45)
		{5.18 m ($W_{\frac{1}{2}}$ 6)}	0.60		
(21)		5.32 dd (10;4.5)	1.15	283 [284]	−0.90 [−0.90]
		{5.37 dd (11;6)}	0.72		

[a] Recorded at 60 MHz; solvent $CDCl_3$; δ values; coupling constants or signal widths (in Hz) in parentheses. Data for solutions in benzene are in braces. [b] Solvent CH_3OH. Data for solutions in CH_3CN are in square brackets. [c] N.m.r. data reported [3b] for these compounds were for solutions in CS_2.

EXPERIMENTAL

M.p.s were taken on a Fisher–Johns apparatus. Optical rotations were recorded with an automatic Perkin-Elmer 141 polarimeter and refer to solutions in chloroform. I.r. spectra were recorded for solutions in chloroform with a Perkin-Elmer 237 grating instrument; O–H stretching measurements were done for 0.005M-solutions in carbon tetrachloride in a 20 mm path cell with a Beckman IR7 instrument. N.m.r. spectra were determined with a Varian NV-14 instrument (60 MHz) for solutions in deuteriochloroform or deuteriobenzene, as stated. C.d. spectra were taken with a Cary 60 instrument for solutions in methanol or acetonitrile, as stated. Column chromatography was performed on silica gel 60 (Merck; 70—230 mesh). Analyses were performed in the microanalytical laboratory of the Weizmann Institute. Mass spectra were taken with a Varian MAT 731 HR instrument.

Preparation of Acetoxy-ketones.—2β-Acetoxy-5α-cholestan-3-one (14),[1,3a] 2β-acetoxy-5-hydroxy-5α-cholestan-3-one (15),[1] 2α-acetoxy-5-hydroxy-5α-cholestan-3-one (17),[1] and 3α-acetoxy-5α-cholestan-2-one (20) [3a] were prepared as described.

3β-*Acetoxy*-5α-*cholestan*-2-*one* (18).—To a solution of 3β-acetoxy-2β-hydroxy-5α-cholestane (3) [1] (100 mg) in acetone (40 ml), a solution of Jones reagent was added dropwise, with stirring, at 10—15 °C. After 20 min, the excess of reagent was destroyed with a few drops of methanol, most of the solvent was removed, water was added, the product was filtered off and crystallised from ethanol, m.p. 144—146° (lit.,[3a] 144.7—146°).

3β-*Acetoxy*-5-*hydroxy*-5α-*cholestan*-2-*one* (19).—3β-Acetoxy-2β,5-dihydroxy-5α-cholestane (4) [1] (100 mg) was oxidised as above. The product (90 mg) crystallised from methanol, m.p. 198—200°, $[\alpha]_D$ +50.1° (*c*, 0.8), $\nu_{max.}$ 1 722

[11] D. H. Williams and N. S. Bhacca, *Tetrahedron*, 1965, **21**, 2021.

and 1 740 cm^{-1} (Found: C, 75.8; H, 10.65%; M^+, 460. $C_{29}H_{48}O_4$ requires C, 75.6; H, 10.5%; M, 460).

3α-*Acetoxy-5-hydroxy-5α-cholestan-2-one* (21).—A solution of 3α-acetoxy-2β,5-dihydroxy-5α-cholestane (8a) [2] (50 mg) in dry pyridine (1 ml) was added to a cold suspension of Sarett reagent prepared from chromium trioxide (50 mg) and dry pyridine (1.5 ml). After 20 h at room temperature, ice–water was added, the product was extracted with ether, washed with water, and dried (Na_2SO_4), and the solvent was removed. The product (45 mg) crystallised from acetone–hexane, m.p. 162—164° [after melting it resolidifies and melts again at 192—196°, due to formation of acetoxy-ketone (19) by thermal isomerisation; t.l.c. after melting is conclusive for a mixture of compounds (19) and (21)]. $\nu_{max.}$ 1 728 and 1 740 cm^{-1} (Found: C, 75.45; H, 10.6%; M^+, 460. $C_{29}H_{48}O_4$ requires C, 75.6; H, 10.5%; M, 460).

Isomerisation of Acetoxy-ketone (14).[3a]—To a solution of (14) (120 mg) in glacial acetic acid (12 ml), 48% hydrobromic acid in acetic acid (0.1 ml) was added. The solution was kept overnight at room temperature, then water was added, the product was extracted with ether and washed until neutral with water and with aqueous sodium hydrogen carbonate solution. The crude product was chromatographed on silica gel (50 g). Elution with benzene–ethyl acetate (98 : 2) gave 3β-acetoxy-5α-cholestan-2-one (18) (50 mg), crystallised from ethanol, m.p. 144—146°. Further elution with the same solvent gave 2α-acetoxy-5α-cholestan-3-one (16) (30 mg), crystallised from ethanol, m.p. 123° (lit.,[3a] 124.7—125.2°).

Isomerisation of Acetoxy-ketone (20).[3a]—To a solution of (20) (300 mg) in acetic acid (30 ml), 48% hydrobromic acid in acetic acid (0.2 ml) was added. The solution was heated on a steam-bath for 15 min, then kept at room temperature for 3 days. Work-up as above gave a crude product which was chromatographed. Elution as above gave compound (18) (150 mg), m.p. and mixed m.p. 144—146°, followed by compound (16) (40 mg), m.p. and mixed m.p. 122—123°.

Isomerisation of Acetoxy-ketone (21).—A solution of the acetoxy-ketone (100 mg) in benzene was introduced into a column filled with dry silica gel (10 g). After 24 h the product was eluted and re-chromatographed on silica gel. Elution with dichloromethane gave compound (21) (30 mg); elution with dichloromethane–ethyl acetate (9 : 1) gave compound (19) (60 mg).

Reduction of Acetoxy-ketones with Sodium Borohydride.—To a solution of acetoxy-ketone (100 mg) in methanol (20 ml), sodium borohydride (50 mg) was added with stirring at room temperature. After 2 h the solution was neutralised with dilute hydrochloric acid, part of the solvent was removed under reduced pressure, water was added and the product collected by filtration.

Acetoxy-ketone (15) gave 2β-acetoxy-3β,5-dihydroxy-5α-cholestane (2) [1] (70 mg), crystallised from methanol, m.p. and mixed m.p. 170—171°.

Acetoxy-ketone (17) gave a crude product (80 mg) which did not show n.m.r. signals for acetate groups. It was acetylated with acetic anhydride (0.6 ml) and pyridine (1 ml) overnight at room temperature. Following the usual work-up the product was chromatographed. Elution with dichloromethane gave a mixture of triol diacetates (30 mg) which according to n.m.r. were 2α,3α- (11) and 2α,3β-diacetoxy-5-hydroxy-5α-cholestane (13) (10-CH_3 signals at δ 1.04 and 1.08). The products were not separated for complete characterisation. Further elution with dichloromethane–ethyl acetate (9 : 1) gave 2α-acetoxy-3α,5-dihydroxy-5α-cholestane (10) (40 mg), m.p. 187—189° (from methanol); δ 1.02 (10-CH_3), 2.1 ($OCOCH_3$), 4.17 (m, $W_{\frac{1}{2}}$ 8 Hz, 3β-H), and 5.1 (m, $W_{\frac{1}{2}}$ 22 Hz, 2β-H) (Found: C, 75.1; H, 10.9. $C_{29}H_{50}O_4$ requires C, 75.3; H, 10.9%).

Acetoxy-ketone (19) gave 3β-acetoxy-2β,5-dihydroxy-5α-cholestane (4) [1] (95 mg), crystallised from methanol, m.p. and mixed m.p. 186—188 °C.

Acetoxy-ketone (21) (150 mg) gave a crude product showing only a very weak n.m.r. signal for acetate groups. It was acetylated as described above and chromatographed. Elution with hexane–ethyl acetate (9 : 1) gave mixtures which could not be separated; further elution with dichloromethane gave a mixture which was re-chromatographed to give 2β,3α-diacetoxy-5-hydroxy-5α-cholestane (8b),[4] m.p. 121—123 °C, followed by 3β-acetoxy-5-hydroxy-5α-cholestan-2-one (19) (45 mg), m.p. 198—200 °C. The n.m.r. of compound (8b) was as reported.[4]

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