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THE MOLECULAR STRUCTURE OF AN ALDOSTERONE 18-GLUCOPYRANOSIDURONATE

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SUMMARY

The crystal and molecular structure of aldosterone 18R-α-D-glucopyranosiduronic acid tetraacetate methyl ester was determined by X-ray analysis in order to ascertain the stereochemistry of the glycosidic bond and the configuration at C(18). The C(18) stereochemistry was found to be the same as that observed in the crystal structure of aldosterone monohydrate, and the C-ring has similar strain as shown by the C(11)-C(12)-C(13) valency angle of 98°. The sugar ring, which has a $^4C_1$ conformation, is approximately parallel to the steroid nucleus and is located above the aldosterone 17β-side chain.

INTRODUCTION

Soon after aldosterone was isolated and characterized chemically, the existence of an equilibrium between the 18-aldehyde, 11β,18-oxide, and 18-acetal-20-hemiketal forms depicted in Fig. 1 was proposed[1]. Since C(18) becomes asymmetric following the formation of the 11β,18-oxide and C(20) is also asymmetric in the 18-acetal-20-hemiketal isomer, seven structural isomers are possible, and the relative stabilities of these isomers cannot be unambiguously determined by spectroscopic methods[2] or force-field calculations[3]. X-Ray crystallographic studies of aldosterone and its natural and synthetic conjugates provide one possible means of resolving this problem.

Aldosterone is excreted in human urine in part as a polar, acid-labile conjugate which was formulated provisionally as aldosterone 18-glucosiduronic acid[4, 5]. A Koenigs–Knorr reaction involving aldosterone 21-acetate and methyl acetobromoglucuronate results in two products which might be expected to be isomeric polyacetate methyl esters of the conjugate[6]. The configurations at C(18) of the steroid and C(1) of the sugar in all of these compounds were unknown. On account of the uncertainties concerning these configurations and the relative stabilities of the several aldosterone structural isomers, an X-ray investigation was initiated. Thus far, it has been possible to obtain crystals suitable for X-ray analysis for only one of the products of the Koenigs–Knorr reaction[7], and the structure of this material, which was shown to be aldosterone 18R-α-D-glucosiduronic acid tetra-acetate methyl ester* is reported in this paper.

EXPERIMENTAL

Single crystals of aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester suitable for X-ray measurements of lattice parameters and intensities were grown from a methanol–water solution by dissolving the conjugate in methanol, adding an equal vol. of water, and slowly evaporating with an air stream. These crystals were generously supplied by Dr. V. R. Mattox. The systematic absences (0k0; $k = 2n + 1$) in the diffraction pattern indicated the space group to be $P2_1$. The unit cell constants (Table 1) were determined from least-squares analysis of the θ values for 15 reflections, and integrated intensities for 3982 independent reflections having $\theta < 75°$ were

* The trivial name aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester will be used as an abbreviation for methyl [(18R)-11β,18-epoxy-21-acetoxy-3,20-dioxo-4-pregnen-18-yl, 2,3,4-tri-*O*-acetyl-α-D-glucopyranosid] uronate throughout the remainder of this paper.

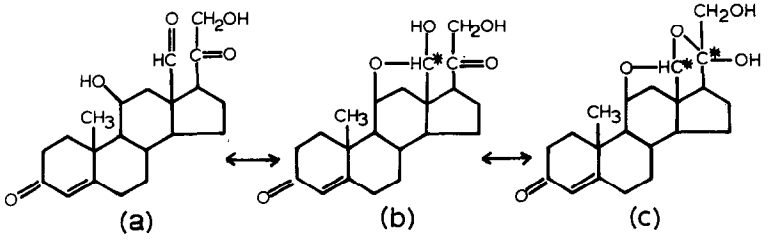


Fig. 1. Aldosterone structural isomers include (a) one 18-aldehyde, (b) two 11β,18-oxides differing in configuration at C(18), and (c) four 18-acetal-20-hemiketals differing in configuration at C(18) and C(20).

Table 1. Experimental crystal data

Formula	$C_{36}H_{46}O_{15}$
Molecular weight	718.8
Density (calculated)	1.29 g·cm^{-3}
Space group	$P2_1$
Z	2
Unit cell dimensions	
a	12.479(3)Å
b	14.389(4)Å
c	10.744(3)Å
β	106.1(1)°
Volume	1853.5Å^3
Melting point	201–202°C

measured on an Enraf–Nonius CAD-4 diffractometer using CuKα radiation. After the Lorentz and polarization corrections [(1 + $\cos^2 2\theta$)/2 $\sin 2\theta$] had been applied, the intensity data were placed on absolute scale by fitting an equation of the type

$$I = I_0 A \exp(B \sin^n\theta/\lambda^n)$$

where the constants A, B, and n were obtained by a least-squares analysis of the data and were found to have values of 0.24, 9.65, and 2.23 respectively. Normalized structure factor amplitudes ($|E|$) were derived from the scaled intensities, and the structure was solved by direct methods. The starting reflections, which are listed in Table 2, were determined manually by inspection of the triple invariant list, and the origin and enantiomorph were selected by specifying phases for reflections (10, 0, $\bar{9}$), (1, 0, 5), (1, 1, 2) and (0, 1, 2). The MULTAN computer program[8] was used to generate 64 possible sets of 452 phases, and the correct phase set was the one having the lowest value of the NQEST figure of merit[9].

The positional and anisotropic thermal parameters of all non-hydrogen atoms were refined by full-matrix least-squares using the 3669 reflections for which the observed intensity was greater than twice the corresponding standard deviation. These reflections were regarded as having intensities significantly greater than the background. The weights used were the quantities $(1/\sigma_F^2)$ where σ_F is defined by equation H.14 of Stout and Jensen[10] using 0.06 rather than 0.01 as the instability correction. The 21-acetate group was found to be disordered with a ratio of about 3:1 between the major and minor occupancy positions, and the positions of the terminal acetate carbons overlap in the two orientations. After the refinement had converged, a Fourier difference map was computed, and the hydrogen atoms were located. The hydrogens were included in the calculations for three final least-squares cycles, but their parameters were not refined. The final reliability index, R (defined as $\Sigma||F_o| - |F_c||/\Sigma|F_o|$), was 5.3% for 3669 reflections used in the refinement and 5.7% for all data. The final refined coordinates and anisotropic thermal parameters for the non-hydrogen atoms are given in Table 3. The standard deviations of all parameters are given in parentheses, and the anisotropic thermal parameters are of the form $\exp[-(\beta_{11}h^2 + \beta_{22}k^2 + \beta_{33}l^2 + \beta_{12}hk + \beta_{13}hl + \beta_{23}kl)]$.

Table 2. Starting reflections used in the MULTAN program

Reflections	$\|E\|$	Phase(s)
10 0 $\bar{9}$	3.53	0
1 0 5	3.16	0
1 1 2	2.89	0
0 1 2	2.71	45, 135
10 4 $\bar{7}$	3.74	45, 135, 225, 315
11 0 $\bar{6}$	3.55	0, 180
2 6 $\bar{7}$	3.20	45, 135, 225, 315

RESULTS

The overall conformation of the aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester molecule is illustrated in Fig. 2 which shows two perspective views of the molecule based on the crystallographically observed atomic coordinates. This figure also illustrates the configurations of C(18) and C(1′).

The bond lengths and valency angles are shown on schematic drawings of the molecule (Fig. 3). The ranges of the standard deviations of the distance and angle measurements involving only ordered non-hydrogen atoms are 0.004–0.008 Å and 0.2–0.4° respectively. The endocyclic torsion angles are also given in Fig. 3, and a torsion angle α-β-γ-δ is considered

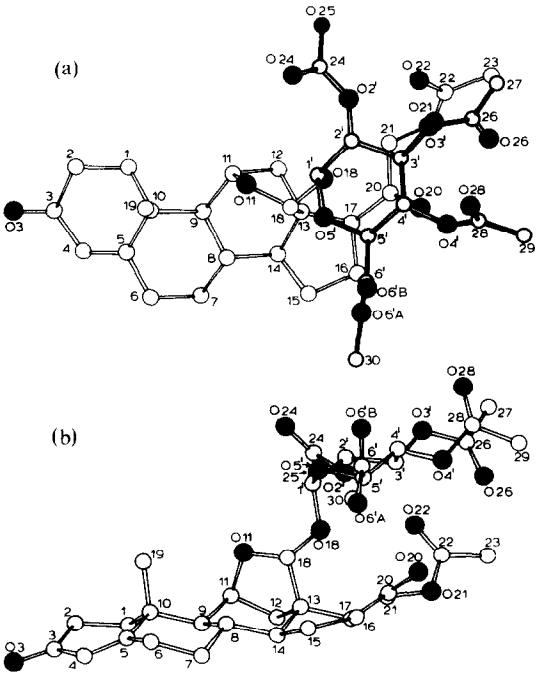


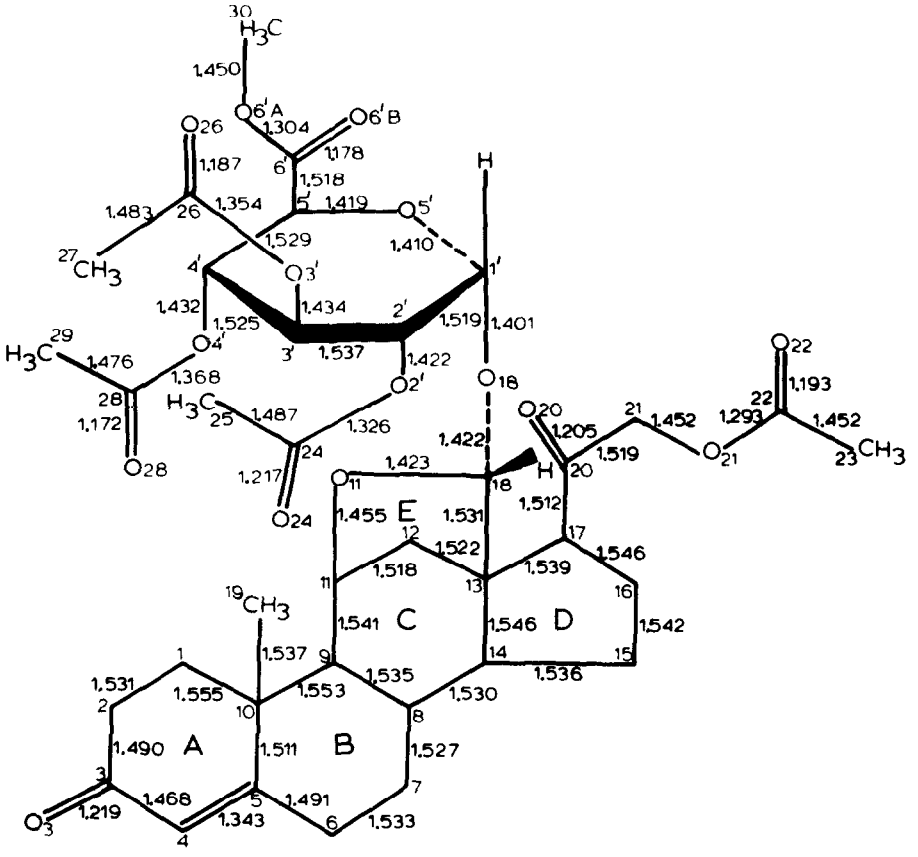
Fig. 2. Crystallographically observed conformation of aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester. ● = oxygen. Only the major position is shown for the 21-acetate. (a) C(10) and C(13) lie in the plane of the paper and a vector joining C(12) and C(14) is parallel to the plane of the paper. (b) The molecule is rotated 75° about the line joining C(10) and C(13).

Table 3. Atomic coordinates and anisotropic thermal parameters for the non-hydrogen atoms

ATOM	X/A	Y/B	Z/C	B11	B22	B33	B12	B13	B23
C(1)	-0.1875(3)	0.0710(3)	0.4690(4)	0.0076(2)	0.00625(20)	0.0231(4)	-0.0009(3)	0.0173(4)	0.0014(5)
C(2)	-0.3102(3)	0.0764(4)	0.4684(4)	0.0080(2)	0.00811(26)	0.0211(4)	-0.0027(4)	0.0154(4)	0.0008(6)
C(3)	-0.3444(3)	0.1711(4)	0.4983(4)	0.0062(2)	0.00966(29)	0.0135(3)	-0.0001(4)	0.0109(4)	-0.0034(5)
C(4)	-0.2798(3)	0.2498(3)	0.4788(3)	0.0076(2)	0.00747(20)	0.0122(3)	0.0019(3)	0.0097(3)	-0.0035(4)
C(5)	-0.1965(3)	0.2406(3)	0.4149(3)	0.0065(2)	0.00585(15)	0.0099(2)	0.0008(3)	0.0063(3)	-0.0031(4)
C(6)	-0.1461(3)	0.3254(3)	0.3746(4)	0.0090(2)	0.00490(15)	0.0128(3)	0.0020(3)	0.0103(4)	-0.0018(4)
C(7)	-0.0186(3)	0.3288(2)	0.4125(3)	0.0080(2)	0.00444(14)	0.0120(3)	-0.0011(3)	0.0087(4)	-0.0033(3)
C(8)	0.0194(2)	0.2387(2)	0.3577(3)	0.0068(2)	0.00429(12)	0.0080(2)	-0.0010(3)	0.0059(3)	-0.0015(3)
C(9)	-0.0288(2)	0.1473(2)	0.4007(3)	0.0055(1)	0.00428(12)	0.0085(2)	-0.0004(2)	0.0061(3)	-0.0013(3)
C(10)	-0.1578(2)	0.1470(2)	0.3816(3)	0.0056(1)	0.00505(14)	0.0105(2)	-0.0000(3)	0.0065(3)	-0.0026(3)
C(11)	0.0219(2)	0.0594(2)	0.3659(3)	0.0063(1)	0.00419(12)	0.0099(2)	-0.0001(2)	0.0065(2)	0.0001(3)
C(12)	0.1467(2)	0.0531(2)	0.4284(3)	0.0063(1)	0.00502(14)	0.0096(2)	0.0002(3)	0.0075(3)	0.0018(3)
C(13)	0.1837(2)	0.1286(2)	0.3502(2)	0.0055(1)	0.00486(13)	0.0065(2)	-0.0015(2)	0.0045(2)	-0.0006(3)
C(14)	0.1463(2)	0.2244(2)	0.3884(3)	0.0065(2)	0.00507(14)	0.0076(2)	-0.0029(3)	0.0057(3)	-0.0022(3)
C(15)	0.2845(3)	0.2950(3)	0.3219(3)	0.0088(2)	0.00496(14)	0.0113(3)	-0.0042(3)	0.0081(4)	-0.0006(4)
C(16)	0.3092(3)	0.2449(3)	0.3858(3)	0.0079(2)	0.00660(18)	0.0133(3)	-0.0040(3)	0.0106(3)	-0.0007(4)
C(17)	0.3002(2)	0.1474(3)	0.3659(3)	0.0056(1)	0.00636(16)	0.0076(2)	-0.0018(3)	0.0050(3)	-0.0016(3)
C(18)	0.1138(2)	0.1040(2)	0.2130(2)	0.0054(1)	0.00417(12)	0.0074(2)	-0.0005(2)	0.0051(2)	-0.0014(3)
C(19)	-0.2139(3)	0.1267(3)	0.2380(4)	0.0064(2)	0.00796(22)	0.0136(3)	0.0003(4)	0.0037(4)	-0.0065(4)
C(20)	0.3780(2)	0.0767(3)	0.3194(3)	0.0051(1)	0.00880(21)	0.0091(2)	-0.0016(3)	0.0058(3)	-0.0029(4)
C(21)	0.3879(3)	-0.0176(4)	0.3853(4)	0.0080(2)	0.00867(26)	0.0132(3)	0.0040(4)	0.0099(4)	0.0018(5)
C(22)	0.4877(4)	-0.1384(4)	0.3125(5)	0.0092(3)	0.00629(25)	0.0115(4)	0.0021(5)	0.0047(6)	0.0021(6)
C(23)	0.5982(4)	-0.1767(5)	0.3277(6)	0.0109(4)	0.00881(32)	0.0168(5)	0.0067(6)	0.0064(7)	0.0021(7)
C(24)	0.1135(3)	-0.2052(3)	0.0598(4)	0.0092(2)	0.00688(20)	0.0132(4)	-0.0049(4)	0.0068(5)	-0.0023(5)
C(25)	0.1425(5)	-0.2802(4)	0.1579(6)	0.0131(4)	0.00764(24)	0.0230(7)	-0.0069(5)	0.0066(8)	0.0069(6)
C(26)	0.4450(3)	-0.1289(3)	-0.0468(3)	0.0060(2)	0.00680(19)	0.0116(3)	0.0028(3)	0.0074(3)	-0.0004(4)
C(27)	0.4753(4)	-0.2112(4)	-0.1132(6)	0.0093(3)	0.01080(26)	0.0236(5)	0.0064(4)	0.0099(5)	-0.0127(6)
C(28)	0.4272(5)	0.0617(4)	-0.2086(4)	0.0105(2)	0.01064(31)	0.0134(3)	0.0028(5)	0.0147(3)	0.0066(5)
C(29)	0.5444(4)	0.0925(6)	-0.1792(5)	0.0100(3)	0.01457(50)	0.0218(4)	0.0012(7)	0.0195(4)	0.0088(9)
C(30)	0.1859(6)	0.3734(4)	-0.1333(7)	0.0207(7)	0.00618(23)	0.0280(6)	0.0053(7)	0.0175(12)	0.0116(7)
C(1')	0.1164(2)	0.4144(2)	0.0300(3)	0.0052(1)	0.00512(14)	0.0072(2)	0.0006(2)	0.0041(3)	-0.0007(3)
C(2')	0.1768(2)	-0.0661(2)	-0.0121(3)	0.0049(1)	0.00495(13)	0.0083(2)	0.0003(2)	0.0041(3)	-0.0014(3)
C(3')	0.2922(2)	-0.0371(2)	-0.0236(3)	0.0053(1)	0.00518(14)	0.0084(2)	0.0005(2)	0.0051(3)	-0.0017(3)
C(4')	0.2781(2)	0.0480(3)	-0.1113(3)	0.0066(2)	0.00612(16)	0.0075(2)	0.0010(3)	0.0052(3)	-0.0002(3)
C(5')	0.2169(3)	0.1235(2)	-0.0578(3)	0.0074(2)	0.00519(15)	0.0078(2)	0.0013(3)	0.0054(3)	0.0015(3)
C(6')	0.1937(3)	0.2095(3)	-0.1429(3)	0.0090(2)	0.00653(18)	0.0111(3)	0.0030(4)	0.0079(4)	0.0058(4)
C(22*)	0.5259(9)	-0.1204(9)	0.4829(12)	0.0050(6)	0.00515(55)	0.0099(10)	0.0030(10)	0.0027(13)	0.0045(13)
O(3)	-0.4257(2)	0.1816(4)	0.5387(3)	0.0104(2)	0.01343(30)	0.0243(3)	0.0003(4)	0.0221(3)	-0.0054(6)
O(11)	0.0156(2)	0.0650(2)	0.2284(2)	0.0054(1)	0.00520(10)	0.0093(1)	-0.0013(2)	0.0059(2)	-0.0036(2)
O(18)	0.1715(1)	0.0370(2)	0.1587(2)	0.0058(1)	0.00496(9)	0.0067(1)	0.0008(2)	0.0046(2)	-0.0010(2)
O(20)	0.4274(2)	0.0931(3)	0.2401(2)	0.0079(1)	0.00987(19)	0.0101(2)	-0.0028(3)	0.0093(2)	-0.0024(3)
O(21)	0.4894(3)	-0.0676(3)	0.3873(4)	0.0081(2)	0.00767(22)	0.0190(4)	-0.0002(4)	0.0124(4)	-0.0047(5)
O(22)	0.4039(4)	-0.1688(4)	0.2414(5)	0.0099(3)	0.00993(31)	0.0197(6)	0.0023(6)	-0.0024(8)	-0.0060(8)
O(24)	0.0248(3)	-0.1947(3)	-0.0237(4)	0.0113(3)	0.01145(25)	0.0187(4)	-0.0106(4)	-0.0020(6)	0.0026(6)
O(26)	0.5100(2)	-0.0816(3)	0.0285(3)	0.0060(1)	0.00879(19)	0.0155(3)	0.0020(3)	0.0036(4)	-0.0036(4)
O(28)	0.3761(3)	0.0414(5)	-0.3089(3)	0.0130(2)	0.02028(54)	0.0127(2)	-0.0018(6)	0.0171(3)	-0.0066(6)
O(2')	0.1922(2)	-0.1412(2)	0.0772(2)	0.0065(1)	0.00498(11)	0.0114(2)	-0.0010(2)	0.0037(3)	0.0000(3)
O(3')	0.3336(2)	-0.1137(2)	-0.0821(2)	0.0064(1)	0.00620(13)	0.0103(2)	0.0010(2)	0.0054(3)	-0.0037(3)
O(4')	0.3652(2)	0.0853(2)	-0.1081(2)	0.0070(1)	0.00732(15)	0.0102(2)	-0.0003(3)	0.0080(2)	0.0017(3)
O(5')	0.1107(2)	0.0090(2)	-0.0554(2)	0.0063(1)	0.00553(11)	0.0077(2)	0.0029(2)	0.0042(2)	0.0016(2)
O(6'A)	0.2096(4)	0.2836(2)	-0.0704(4)	0.0178(4)	0.00549(14)	0.0177(3)	0.0040(4)	0.0124(5)	0.0066(4)
O(6'B)	0.1616(4)	0.2491(3)	-0.2569(3)	0.0233(5)	0.00987(25)	0.0101(3)	0.0023(6)	0.0044(6)	0.0082(4)
O(21*)	0.4408(7)	-0.0727(8)	0.3156(7)	0.0062(5)	0.01124(64)	0.0044(5)	0.0109(9)	-0.0021(9)	0.0019(11)
O(22*)	0.5428(12)	-0.1093(9)	0.5167(11)	0.0147(11)	0.00797(55)	0.0120(10)	0.0102(12)	-0.0019(18)	0.0076(12)

* Minor (25%) occupancy position for the 21-acetate.

(a)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O6'B
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O26
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6'
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1.354
5'
1.419
O5'
1.529
1.410
27CH3
4'
O3'
1'
1.432
1.525
1.434
2'
1.519
1.401
3'
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1.476
O4'
1.368
O2'
O18
H
28
1.172
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1.326
1.422
O20
1.205
21
1.452
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1.293
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1.519
O21
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O24
11
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H
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9
14
15
1.536
1
1.531
1.535
2
1.555
1.553
1.530
10
8
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5
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O3
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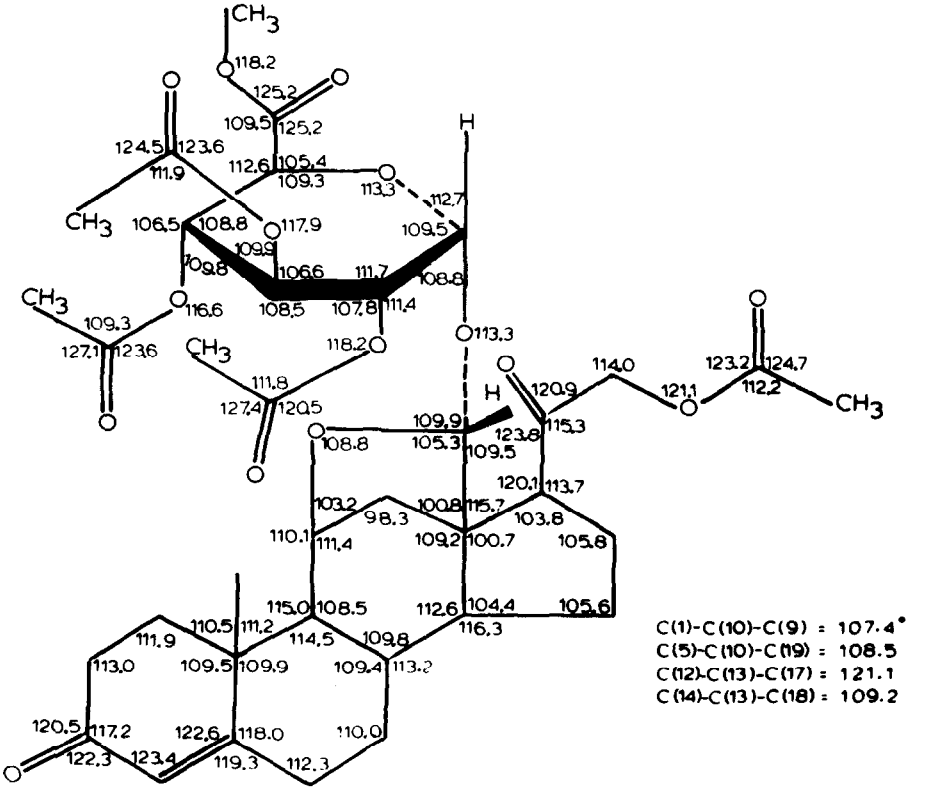
1.533
4

(b)


CH3
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112.7
CH3
106.5
108.8
117.9
109.5
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111.7
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112.6
104.4
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116.3
110.5
111.2
114.5
109.8
111.9
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109.5
109.9
109.4
113.2
120.5
117.2
122.6
118.0
110.0
122.3
123.4
119.3
112.3
C(1)-C(10)-C(9) = 107.4°
C(5)-C(10)-C(19) = 108.5
C(12)-C(13)-C(17) = 121.1
C(14)-C(13)-C(18) = 109.2

Fig. 3.

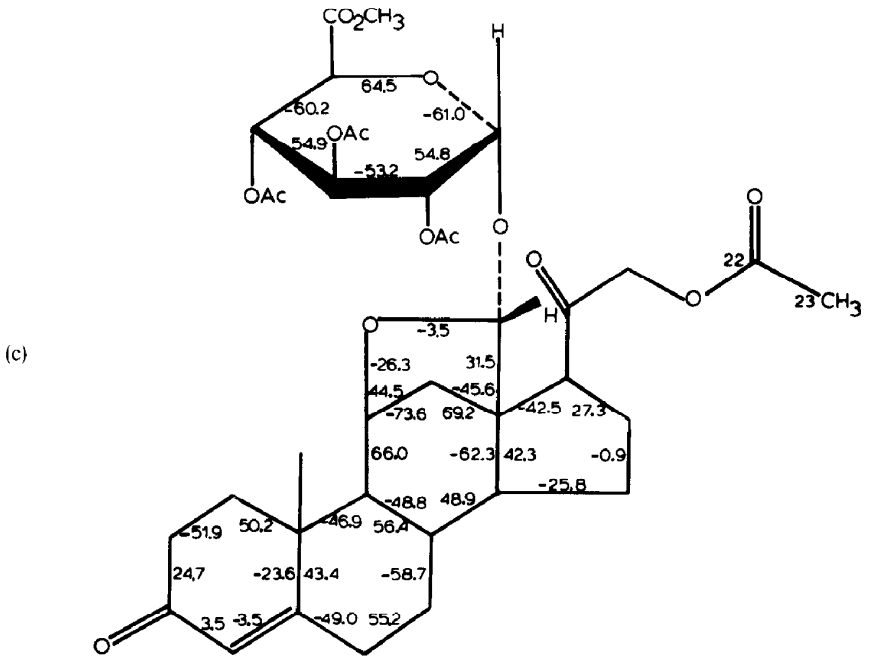


Fig. 3. Intramolecular geometry. (a) Bond distances (Å), (b) valency angles (deg), and (c) endocyclic torsion angles (deg). Only the major position is shown for the 21-acetate.

positive if, when viewed down the β-γ, the α-β bond will eclipse the γ-δ bond when rotated less than 180° in a clockwise direction. The presence of the five-membered ring formed by the 11β,18-epoxide introduces considerable strain into the C-ring as shown by the C(11)-C(12)-C(13) valency angle of 98.3° and the large endocyclic torsion angles centered about the C(9)-C(11), C(11)-C(12), and C(12)-C(13) bonds. Similar strain was observed in the aldosterone monohydrate structure[11].

The steroid nucleus is relatively flat and resembles the nucleus in aldosterone monohydrate[11] which also showed little bowing towards the α-face. The glucopyranosiduronic acid moiety, which has a $^4C_1$ conformation[12], is located over the 17β-side chain of the steroid, and the plane of the sugar ring is inclined at 12° with respect to the plane of the steroid nucleus. The conformations about bonds C(18)-O(18) and O(18)-C(1′) which join the sugar to the steroid are illustrated in detail in Figs. 4d and 4e.

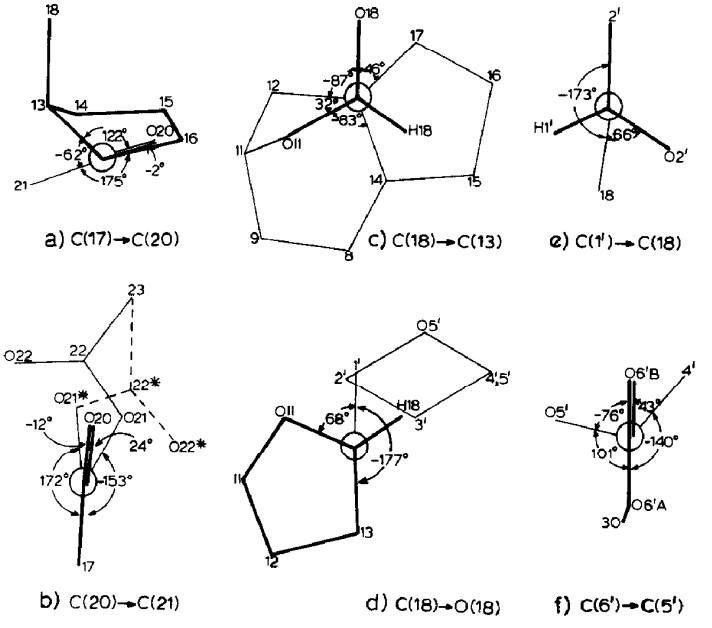


Fig. 4. Newman projections. In diagram (b), broken lines are used to indicate bonds involving atoms in the minor position of the 21-acetate.

Table 4. Intermolecular non-hydrogen distances less than 3.4 Å

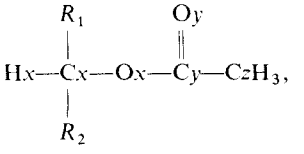
Atom 1	Atom 2	Distance	Position*
a. Distances which do not involve disordered atoms			
C(17)	O(3)	3.36 Å	1/100
C(20)	O(3)	3.26	1/100
O(24)	O(6'A)	3.37	2/0-10
b. Distances involving disordered atoms having 75% occupancy			
C(2)	O(21)	3.18	1/-100
O(3)	C(22)	3.25	2/0-11
O(3)	O(22)	3.15	2/001
c. Distances involving disordered atoms having 25% occupancy			
C(2)	O(22*)	3.36	1/-100
C(16)	O(22*)	3.08	2/101
O(3)	C(22*)	3.24	2/001
O(3)	O(22*)	3.33	2/001

* The equivalent positions are 1 = (x, y, z) and 2 = $(-x, \frac{1}{2}+y, -z)$. The notation 2/1-10 means that the second atom is at equivalent position 2, translated one unit cell in the a and $-b$ directions.

The C(17) side chain has a conformation which is slightly outside the normal range observed for corticosteroids[13]. As shown in Fig. 4a, the C(13)-C(17)-C(20)-O(20) torsion angle is 122°, and the C(16)-C(17) bond eclipses the C(20)-O(20) bond. The C(13)-C(17)-C(20)-O(20) torsion angle is in the range 75–115° in 49 of the 52 pregnan-20-ones which have been studied crystallographically. It is interesting to note that the presence of the bulky glucopyranoside ring lying above the 17β-side chain does not cause this side-chain to move significantly away from the commonly observed conformation.

The 21-acetate group is disordered giving rise to two positions for atoms O(21), C(22), and O(22) although the positions for the terminal carbon, C(23) overlap so that this atom does not appear to be disordered. The relative sizes of the two peaks corresponding to each of these atoms on an electron density map indicate that approximately 75% of the molecules have one acetate orientation, and the atoms in the minor (25%) occupancy positions are indicated by asterisks. Figure 4b is a Newman projection down the C(20-C(21) bond, and it shows that both positions of O(21) are nearly *cis* coplanar with respect to O(20). Atoms O(20) and O(21) have been observed to be *cis* coplanar in approximately 20 other corticosteroids[13]. The reason for this disorder is not clear, but it may be related to the close intermolecular distances involving both positions of the disordered atoms. These short intermolecular contacts are listed in Table 4.

An acetate group and adjacent atoms may be represented schematically as



and torsion angles which define the conformations of the acetate groups in aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester are given in Table 5. The torsion angles Cx-Ox-Cy-Oy, where Cy-Oy are the carbonyls, are all near 0° indicating that Cx and Oy are always *cis* coplanar. In addition, the conformation about the Cx-Ox bonds are also such that Hx and Cy are approximately *cis* coplanar. Thus, in each case, all nonhydrogen atoms in the acetate groups are approximately coplanar with an adjacent carbon atom and a hydrogen bonded to it.

DISCUSSION

The relative stabilities of the seven aldosterone structural isomers are unknown. These isomers include one 18-aldehyde, two 11β,18-oxides differing in configuration at C(18), and four 18-acetal-20-hemiketals differing in configuration at C(18) and C(20). Simpson *et al.*[1] suggested that aldosterone reacts mainly as the 11β,18-oxide in solution, but the ease of preparation of 20,21-cyclic acetals of aldosterone caused Gardi[14] to suggest that aldosterone can react equally well in solution as the 18-acetal-20-hemiketal. The X-ray analyses of aldosterone monohydrate[11] and the aldosterone 18-glucosiduronate discussed here provide the first solid state evidence concerning the stabilities of the various aldosterone isomers. The aldosterone moiety in the aldosterone monohydrate structure exists as the 18R-acetal-20S-hemiketal isomer, and aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester is a derivative of aldosterone 11β,18S-oxide. Perspective drawings of the C(18) regions of these two structures are given in Fig. 5 to show that they have the same configuration at this asymmetric center. X-ray analyses of other aldosterone derivatives are underway in order

Table 5. Torsion angles in the acetates

Cx-Ox-Cy-Oy	Angle	Hx-Cx-Ox-Cy	Angle
C(21)-O(21)-C(22)-O(22)	−2.5	H(21C)-C(21)-O(21)-C(22)	13.3
C(21)-O(21*)-C(22*)-O(22*)	0.0	H(21B)-C(21)-O(21*)-C(22*)	−15.4
C(2')-O(2')-C(24)-O(24)	−7.5	H(2')-C(2')-O(2')-C(24)	−30.6
C(3')-O(3')-C(26)-O(26)	−0.9	H(3')-C(3')-O(3')-C(26)	−27.0
C(4')-O(4')-C(28)-O(28)	8.0	H(4')-C(4')-O(4')-C(28)	−23.5

* Minor (25%) occupancy position for the 21-acetate.

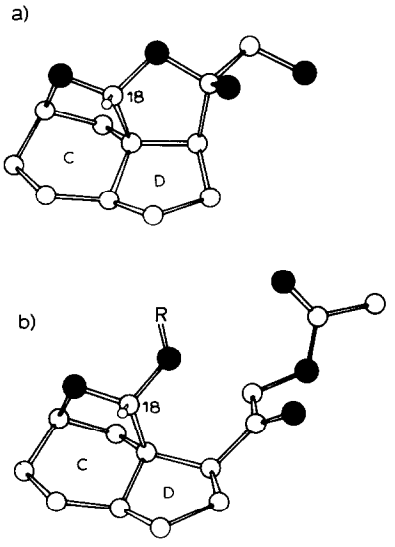


Fig. 5. Perspective drawings of (a) aldosterone and (b) aldosterone 18R-α-D-glucosiduronic acid tetraacetate methyl ester illustrate the configurational similarity at C(18). R = glucosiduronic acid triacetate methyl ester. ● = oxygen. In diagram (b), only the major position of the 21-acetate is drawn.

to provide more information concerning the stabilities of the various isomers.

X-ray analysis of one of the products of the Koenigs–Knorr reaction involving aldosterone 21-acetate and methyl acetobromoglucuronate has provided positive identification of one aldosterone 18-glucosiduronate to which the structures of other 18-glucosiduronates can be related. Carpenter and Mattox[4] have shown that the acid-labile aldosterone conjugate in human urine is the β-anomer of the synthetic D-glucosiduronic acid tetraacetate methyl ester reported here, but the configuration at C(18) in the acid-labile conjugate is unknown.

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