18-Substituted Steroids. Part 13.[1] Improved Preparation of the Metabolites of Aldosterone Reduced in Ring A.

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Protection of the side-chain of aldosterone as the 21-t-butyldimethylsilyl (TBDMS) ether was also found to mask the 20-oxo function, by effectively locking the tautomeric structure of aldosterone into its 11,18:18,20-diepoxy form. Catalytic hydrogenation of aldosterone 21-TBDMS ether gave the 5α- or the 5β-dihydro derivative depending on the choice of solvent. Subsequent reduction of the carbonyl group at C-3, with either 'K-selectride' to form the 3-axial alcohol or lithium tri-t-butoxyaluminium hydride to form the 3-equatorial alcohol, was found to proceed without attack on the side-chain, to give convenient routes to the four isomeric tetrahydroaldosterones.

The metabolites of aldosterone reduced in ring A are known to exhibit aldosterone-like antinatriuretic and kaliuretic activity in varying degrees.[2–4] To aid the study of the excretion and mineralocorticoid properties of these reduced metabolites,† various methods have been developed for their syntheses during recent years. 5α-Dihydroaldosterone, 3β,5α-tetrahydroaldosterone and 3β,5β-tetrahydroaldosterone have been prepared[5] by hydrogenation of aldosterone in the free state. 5β-Dihydroaldosterone has been obtained[6] by hydrogenation of aldosterone 18,21-diacetate, followed by alkaline hydrolysis. The first chemical synthesis[7] of 3α,5β-tetrahydroaldosterone was achieved by the hydrogenation of aldosterone 21-monoacetate, and alkaline hydrolysis. A small amount of the 3α,5α-tetrahydro isomer was also isolated from this reaction sequence. We have described a stereospecific synthesis[8] of 3α,5β-tetrahydroaldosterone, proceeding from 3α-hydroxy-5β-pregnane-3,20-dione *via* 21-deoxy-3α,5β-tetrahydroaldosterone.[9] 3α,5β-Tetrahydroaldosterone has also been obtained[10] by microbial anaerobic reduction of aldosterone with *Clostridium paraputrifucum*, in 80% yield and apparently uncontaminated by other isomers. Recently,[11] 3α,5α-tetrahydroaldosterone has become available through a multistep route starting from 21-hydroxy-3,20-dioxo-5α-pregnane-18,11-lactone, or preferably its 3,20-bisethylenedioxy derivative.

We now describe a convenient route from aldosterone which can be varied to provide each of the metabolites reduced in ring A. The main problem inherent in any route from aldosterone is the selective reduction of the 3-oxo group without simultaneously reducing either the 20-oxo or the masked 18-aldehyde group. The corresponding problem did not arise in the reduction[12] of 18,21-dihydroxy-5α- and -5β-pregnane-3,20-diones ('dihydro-18-hydroxydeoxycorticosterones') because these, like all simple 18-hydroxypregnan-20-ones, exist as 18,20-hemiacetals which are sufficiently stable to be unaffected by borohydride under the mild conditions required to reduce the 3-oxo group. Compounds of the aldosterone series, where there is an additional masked carbonyl function at C-18, are not automatically protected in this way. Although aldosterone monohydrate exists in the crystalline form as the doubly-bridged 20-hydroxy-11,18:18,20-diepoxy form,[13] the 18,20-epoxy (18,20-hemiacetal) bridge is not sufficiently stable to survive intact in solution. The solution state of aldosterone in all common solvents is an equilibrium between the 18,20-epoxy(hemiacetal) and 20-oxo forms.[1] The ease of reduction at C-20 as well as at C-3 has been shown by the reported synthesis[14] of the 'hexahydro' derivatives of aldosterone using sodium borohydride.

We therefore needed a convenient means to lock the 11,18:18,20-diepoxy structure rigidly, before we could effect exclusive reduction of 5α- and 5β-dihydroaldosterones at C-3 by the complex hydrides known to favour the formation of axial or equatorial products, respectively. Attempts to tie up the 20,21-diol system of the 11,18:18,20-diepoxy form of aldosterone either as the 20,21-acetonide or by condensation with dimethyldichlorosilane to form a 20,21-siliconide[15] were unsuccessful. However, when we treated aldosterone with t-butylchlorodimethylsilane (TBDMS),[16] hoping that the bulky TBDMS group might prevent approach of hydride to C-20, we found not only that the silylation was highly selective for the 21-hydroxy group, but also that the 21-TBDMS ether (**1**) of aldosterone exists entirely in a single 11,18:18,20-acetal form having the sterically favoured[1,8,9] (18*R*,20*S*) configuration. The absence of the diastereoisomeric (18*R*,20*R*) acetal, and of the 11,18-hemiacetal-20-oxo forms, was apparent from the highly simplified signals in its 400 MHz n.m.r. spectrum (in deuteriochloroform) which showed only one doublet for 11-H at δ 4.82, one singlet for 18-H at δ 5.39 and a pair of doublets (*J* 10 Hz) at δ 3.51 and δ 3.62 for 21-H_2. In contrast, the 400 MHz spectrum of aldosterone in deuteriochloroform is found to exhibit separate signals for 21-H, 18-H, and 11-H of the two 11,18:18,20-acetal forms and the two 11,18-hemiacetal-20-oxo forms.[1,17]

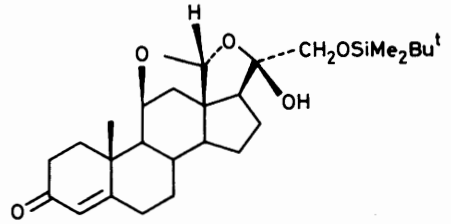
This discovery of an efficient method for effectively masking the entire side-chain system opened the way to stereoselective reductions of aldosterone in ring A. Hydrogenation of the TBDMS ether (**1**) at atmospheric pressure for 2 h in the presence of 10% palladised charcoal furnished a mixture of the isomeric dihydro derivatives (**2a**) and (**2b**); the 5α:5β ratio being dependent on the chosen solvent. In a relatively basic medium such as pyridine or *N*-methylpyrrolidine the 5β-isomer (**2b**) was predominant, whereas the 5α-isomer (**2a**) was the major component when the hydrogenation was carried out in ethyl acetate. There was no evidence of incomplete reduction, hydrogenolysis, or over-reduction (at C-3) under these conditions. The isomers (**2a**) and (**2b**) could be conveniently separated on a preparative scale by h.p.l.c. on Nucleosil. Their identities were established by (a) desilylation with tetrabutyl-

† Trivial names are used as follows: 5α- and 5β-dihydroaldosterone refer to 18,21-dihydroxy-11β,18-epoxy-5α-pregnane-3,20-dione and its 5β-isomer, respectively; 3α,5α-, 3α,5β-, 3β,5α-, and 3β,5β-tetrahydroaldosterone refer to 3α,18,21-trihydroxy-11β,18-epoxy-5α-pregnan-20-one and its respective isomers at C-3 and C-5.

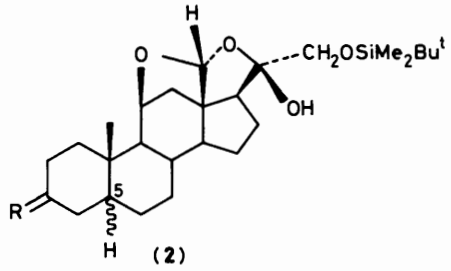
ammonium fluoride to give the parent 5α- and 5β-dihydroaldosterones, and (b) direct silylation of the authentic dihydroaldosterone with t-butylchlorodimethylsilane to form the same derivatives.

Reduction at C-3 of the TBDMS ethers (**2a**) and (**2b**) with lithium tri-t-butoxyaluminium hydride proceeded smoothly to furnish predominantly the corresponding 3-equatorial alcohols (**2c**) and (**2f**). Alternatively, potassium tri-s-butylborohydride ('K-selectride') furnished mainly the corresponding 3-axial alcohols. The products in both the 5α- and the 5β-series were readily separable from small amounts of the lesser isomers by h.p.l.c. on Nucleosil.

Desilylation of each of the four TBDMS ethers (**2c**)—(**2f**) with tetrabutylammonium fluoride gave the corresponding known tetrahydro-metabolite of aldosterone.



(**1**)



(**2**)

a; 5α-H, R=O
b; 5β-H, R=O
c; 5α-H, R=β-OH, α-H
d; 5α-H, R=α-OH, β-H
e; 5β-H, R=β-OH, α-H
f; 5β-H, R=α-OH, β-H

Experimental

M.p.s were determined on a Reichert melting microscope. I.r. spectra were run in potassium bromide discs unless otherwise specified. N.m.r. spectra were determined in deuteriochloroform pretreated with a trace of deuteriopyridine, using tetramethylsilane as internal standard. Water for h.p.l.c. was distilled from glass and freed from organic contaminants by passing through a 'Norganic' cartridge (Millipore Corporation, Bedford, Massachusetts 017300). H.p.l.c. was on a 25 cm stainless steel column of 1 cm i.d. packed with Nucleosil. All solvents were distilled before use. 'Light petroleum' refers to the fraction of b.p. 60—80 °C. Tetrahydrofuran (THF) was successively heated under reflux with sodium hydroxide pellets, then with sodium wire, and finally distilled over lithium aluminium hydride immediately before use.

Silylation of Aldosterone with t-Butylchlorodimethylsilane.—A solution of t-butylchlorodimethylsilane (100 mg) in dichloromethane (0.5 ml) was added with swirling to a solution of aldosterone (25 mg) and imidazole (100 mg) in dichloromethane (0.5 ml). After 10 min the mixture was diluted with dichloromethane (5 ml), washed successively with water (2 × 3 ml), dilute hydrochloric acid (2 × 3 ml; 1M), aqueous sodium hydrogen carbonate (3 ml; 10%) and water (3 × 3 ml), dried (K_2CO_3), and evaporated under reduced pressure at room temperature to give an oil containing the required silyl ether (**1**). t-Butyldimethylsilyl contaminants (TBDMS) derived from the reagent were only partially removed under high vacuum. H.p.l.c. with light petroleum–ethyl acetate (7:3) as mobile phase furnished pure *aldosterone 21-TBDMS ether* (30 mg) which crystallised from ethyl acetate, m.p. 119—120 °C (Found: M^+, 474.280 626. $C_{27}H_{42}O_5Si$ requires M, 474.280 147); $\nu_{max.}$ 3 380, 1 660, 1 610, 1 210, and 840 cm^{-1}; δ (100 MHz), 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3), 1.30 (s, 19-H_3), 3.62 and 3.51 (dd, J 10.0 Hz, 21-H_2), 4.82 (d, J 6 Hz, 11-H), 5.39 (s, 18-H), and 5.72 (s, 4-H).

5α- and 5β-Dihydroaldosterone 21-TBDMS Ethers (**2a**) *and* (**2b**).—(i) Authentic samples of these compounds were prepared by direct silylation of the parent dihydroaldosterones with t-butylchlorodimethylsilane as above, employing a 4:1 mixture of light petroleum–ethyl acetate as the mobile phase for h.p.l.c. purification. (ii) The crude oily aldosterone TBDMS ether, prepared as above without h.p.l.c. purification, was hydrogenated in a suitable solvent (*ca.* 5 ml solvent/0.1 g of aldosterone) chosen to optimise the yield of either the 5α- or the 5β-isomer (see below). The solution was stirred for 2 h at room temperature and atmospheric pressure over 10% palladised charcoal (equal in weight to the original aldosterone). The catalyst was filtered off, washed with the solvent, and the combined filtrates were evaporated under reduced pressure below 40 °C. The residual oils were subjected to h.p.l.c. with light petroleum–ethyl acetate (5:1) containing 0.5% methanol, as the mobile phase, to separate the more polar 5α-dihydroaldosterone 21-TBDMS ether (**2a**) from the 5β-isomer (**2b**). The presence of methanol as a polar modifier distorted the peak for the more retentive 5α-isomer but eliminated tailing and thus improved the resolution and efficiency of the column. The following amounts of the two isomers were isolated from hydrogenations in the solvent indicated (from 100 mg of aldosterone in each case). Ethyl acetate: 5α, 80 mg; 5β, 38 mg. *N*-methylpyrrolidine: 5α, 28 mg; 5β, 70 mg. Pyridine: 5α, 32 mg; 5β, 82 mg.

5α-Dihydroaldosterone 21-TBDMS ether (**2a**) crystallised from ethyl acetate–light petroleum, m.p. 176—177 °C (Found: M^+, 476.295 667. $C_{27}H_{44}O_5Si$ requires M, 476.295 797); $\nu_{max.}$ 3 380, 1 710, and 840 cm^{-1}; δ (100 MHz), 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3), 1.12 (s, 19-H_3), 3.58 and 3.51 (dd, J ~10 Hz, 21-H_2), 4.80 (d, J 6 Hz, 11 α-H), and 5.34 (s, 18-H).

5β-Dihydroaldosterone 21-TBDMS ether (**2b**) was an oil (Found: M^+, 476.297 059. $C_{27}H_{44}O_5Si$ requires 476.295 797); $\nu_{max.}$(thin film) 3 380 and 1 705 cm^{-1}; δ (100 MHz), 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3), 1.24 (s, 19-H_3), 3.60 and 3.50 (dd, J 10.5 Hz, 21-H_2), 4.74 (d, 11α-H, J 6 Hz), and 5.34 (s, 18-H).

3α,5α-Tetrahydroaldosterone 21-TBDMS Ether (**2d**).—A 0.5M solution of 'K-selectride' (280 μl) in THF was added to a solution of 5α-dihydroaldosterone 21-TBDMS ether (50 mg) in dry THF (1.2 ml) maintained at −50 to −55 °C. After 45 min the steroid was isolated by addition of dichloromethane (5 ml) and washing with water. The dried (K_2CO_3) organic extract upon evaporation and isolation by h.p.l.c. on Nucleosil, using light petroleum–ethyl acetate (7:3) as the mobile phase, furnished first 3α,5α-*tetrahydroaldosterone 21-TBDMS ether* (25 mg), m.p. 119—120 °C from ethyl acetate–light petroleum (Found: M^+, 478.313 491. $C_{27}H_{46}O_5Si$ requires M, 478.311 447); δ (100 MHz), 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3 and 19-H_3), 3.50 and 3.61 (dd, J 10 Hz, 21-H_2), 4.06 (m, $W_{\frac{1}{2}}$ ~6 Hz,

Table. ^{1}H N.m.r. spectra of tetrahydroaldosterone isomers

Isomer	18-H (s)	21-H_2 (AB double doublet)	11α-H (d)	3-H (m)	19-H_3 (s)
3α,5α					
18,20 Hemiacetal forms (**1**)	5.34 [5.30]	3.59, 3.45 (*J* 11.4 Hz) [3.67, 3.49 (*J* 11.25 Hz)]	4.82 (*J* 5.6 Hz)	4.03 ($W_{\frac{1}{2}}$ ~9 Hz)	0.84
20-Oxo forms (**2**)	4.95 [4.88]	4.40, 4.22 (*J* 17.9 Hz) [4.33, 4.20 (*J* 19.3 Hz)]	4.59 (*J* 6.3 Hz) [4.55 (*J* 6.3 Hz)]		0.90
3β,5α					
18,20-Hemiacetal forms (**1**)	5.34	3.59, 3.46 (*J* 11.4 Hz)	4.71 (*J* 5.6 Hz)	3.57 ($W_{\frac{1}{2}}$ ~30 Hz)	0.90
20-Oxo forms (**2**)	4.95	4.40, 4.23 (*J* 17.9 Hz)	4.57 (*J* 6.3 Hz)		0.94
3α,5β					
18,20-Hemiacetal forms (**1**)	5.33 [5.18]	3.59, 3.45 (*J* 11.6 Hz)	4.72 (*J* 5.6 Hz) [4.76 (*J* 5.8 Hz)]	3.7 ($W_{\frac{1}{2}}$ ~30 Hz)	0.98
20-Oxo forms (**2**)	4.94 [4.88]	4.40, 4.23 (*J* 17.9 Hz) [4.33, 4.20 (*J* 19.3 Hz)]	4.49 (*J* 6.3 Hz) [4.45 (*J* 6.2 Hz)]		1.02
3β,5β					
18,20-Hemiacetal forms (**1**)	5.34 [5.18]	3.58, 3.45 (*J* 11.4 Hz) [3.68, 3.49 (*J* 11.4 Hz)]	4.72 (*J* 5.9 Hz) [4.76 (*J* 4.9 Hz)]	4.10 ($W_{\frac{1}{2}}$ ~8 Hz)	1.02
20-Oxo forms (**2**)	4.95	4.39, 4.23 (*J* 17.9 Hz)	4.50 (*J* 6.3 Hz)		1.07

400 MHz in $CDCl_3$; Me_4Si as internal standard; entries in square brackets refer to the minor epimer (at C-20 or C-18).

3-H), 4.82 (d, *J* 5 Hz, 11α-H), and 5.33 (s, 18-H). Later fractions contained 3β,5α-tetrahydroaldosterone 21-TBDMS ether (4 mg) (see below).

3β,5α-*Tetrahydroaldosterone 21-TBDMS Ether* (**2c**).—A solution of 5α-dihydroaldosterone 21-TBDMS ether (**2a**) (50 mg) in dry THF (0.6 ml) was added to a pre-cooled solution (*ca.* −60 °C) of lithium tri-t-butoxyaluminium hydride in THF [prepared by adding 20% t-butyl alcohol in THF (0.4 ml) to $LiAlH_4$ (6.0 mg) in THF (0.1 ml)]. After 10 min at *ca.* −60 °C the excess of reagent was destroyed by adding 50% aqueous THF (0.05 ml), followed by saturated aqueous magnesium sulphate (0.05 ml), after which the solution was diluted with ethyl acetate, dried ($MgSO_4$), and filtered. Evaporation of the solvent under reduced pressure and h.p.l.c. on Nucleosil with light petroleum–ethyl acetate (7:3) as mobile phase gave a little 3α,5α-tetrahydroaldosterone 21-TBDMS ether (**2d**) (*ca.* 5 mg) followed by 3β,5α-*tetrahydroaldosterone 21-TBDMS ether* (**2c**) (40 mg), m.p. 148—149 °C (ethyl acetate–light petroleum) (Found: M^+, 478.313 027. $C_{27}H_{46}O_5Si$ requires *M*, 478.311 447); $\nu_{max.}$ 3 400, 1 260, 850, 845, and 780 cm^{-1}; δ (100 MHz), 0.08 (s, $SiMe_2$), 0.92 (s, CMe_3), 0.96 (s, 19-H_3), 3.54 and 3.58 (dd, *J* ~10.5 Hz, 21-H_2), *ca.* 3.6 (m, 3-H superimposed on 21-H_2 signal), 4.80 (d, 11α-H, *J* ~6 Hz), and 5.34 (s, 18-H).

3β,5β-*Tetrahydroaldosterone 21-TBDMS Ether* (**2e**).—Treatment of 5β-dihydroaldosterone 21-TBDMS ether (**2b**) (35 mg) with 'K-selectride' in THF under the conditions described above for the preparation of 3α,5α-tetrahydroaldosterone 21-TBDMS ether (**2d**), and preparative h.p.l.c. on Nucleosil employing light petroleum–ethyl acetate (2:1) as mobile phase, furnished 3β,5β-*tetrahydroaldosterone 21-TBDMS ether* (20 mg) (**2e**), m.p. 94—95 °C (ethyl acetate–light petroleum) (Found: M^+, 478.313 027. $C_{27}H_{46}O_5Si$ requires *M*, 478.311 447); $\nu_{max.}$ 3 400, 1 260, 1 090, 1 040, 1 000, 840, and 780 cm^{-1}; δ (100 MHz), 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3), 1.05 (s, 19-H_3), 3.60 and 3.49 (dd, *J* ~10 Hz, 21-H_2), 4.09 (m, $W_{\frac{1}{2}}$ 7 Hz, 3-H), 4.72 (d, *J* 5 Hz, 11α-H), and 5.31 (s, 18-H). Later fractions gave a small amount (*ca.* 3 mg) of 3α,5β-tetrahydroaldosterone 21-TBDMS ether (see below).

3α,5β-*Tetrahydroaldosterone 21-TBDMS Ether* (**2f**).—Reduction of 5β-dihydroaldosterone 21-TBDMS ether (60 mg) with lithium tri-t-butoxyaluminium hydride under the conditions described above for the preparation of 3β,5α-tetrahydroaldosterone 21-TBDMS ether, and h.p.l.c. on Nucleosil with light petroleum–ethyl acetate (3:2) as mobile phase, gave a small amount (*ca.* 3—4 mg) of 3β,5β-tetrahydroaldosterone 21-TBDMS ether (**2e**) followed by the required 3α,5β-*isomer* (48 mg) (**2f**) as an oil (Found: M^+, 478.313 060. $C_{27}H_{46}O_5Si$ requires *M*, 478.311 447); δ (100 MHz) 0.08 (s, $SiMe_2$), 0.90 (s, CMe_3), 0.92 (s, 19-H_3), 3.60 and 3.50 (dd, *J* 10.5 Hz, 21-H_2), *ca.* 3.6 (m, 3-H superimposed on 21-H_2 signal), 4.81 (d, *J* 5 Hz, 11α-H), and 5.34 (s, 18-H).

Desilylation of the Tetrahydroaldosterone 21-TBDMS Ethers.—Each TBDMS ether (50 mg) in dry THF (1.5 ml) was treated with a 1M solution of tetrabutylammonium fluoride in THF (0.13 ml) at 20 °C for 5 min. Each mixture was then diluted with ethyl acetate, washed with water, and evaporated under reduced pressure. Product isolation by reverse-phase h.p.l.c. on Spherisorb ODS, furnished the parent tetrahydroaldosterone (15—18 mg). The mobile phase (methanol–water 11:9) used for h.p.l.c. was carefully pre-neutralised (to pH 7.1) with triethylamine. N.m.r. data for the tetrahydroaldosterones are given in the Table.

3α,5α-Tetrahydroaldosterone, m.p. 164—165 °C from ethyl acetate–light petroleum (lit.,[11] m.p. 167—170 °C). 3α,5β-Tetrahydroaldosterone, m.p. 114—116 °C from ether (lit.,[8] m.p. 107—114 °C). 3β,5α-Tetrahydroaldosterone, m.p. 168—170 °C

from ethyl acetate–light petroleum (lit.,[5] 180—183 °C and 180—190 °C). 3β,5β-Tetrahydroaldosterone, m.p. 97—98 °C (isolated previously[5] as a non-crystallisable glass).

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