43. *The Position of the Tertiary Hydroxyl Group in Agapanthagenin.*

By G. E. A. MATHEW and (MRS.) T. STEPHEN.

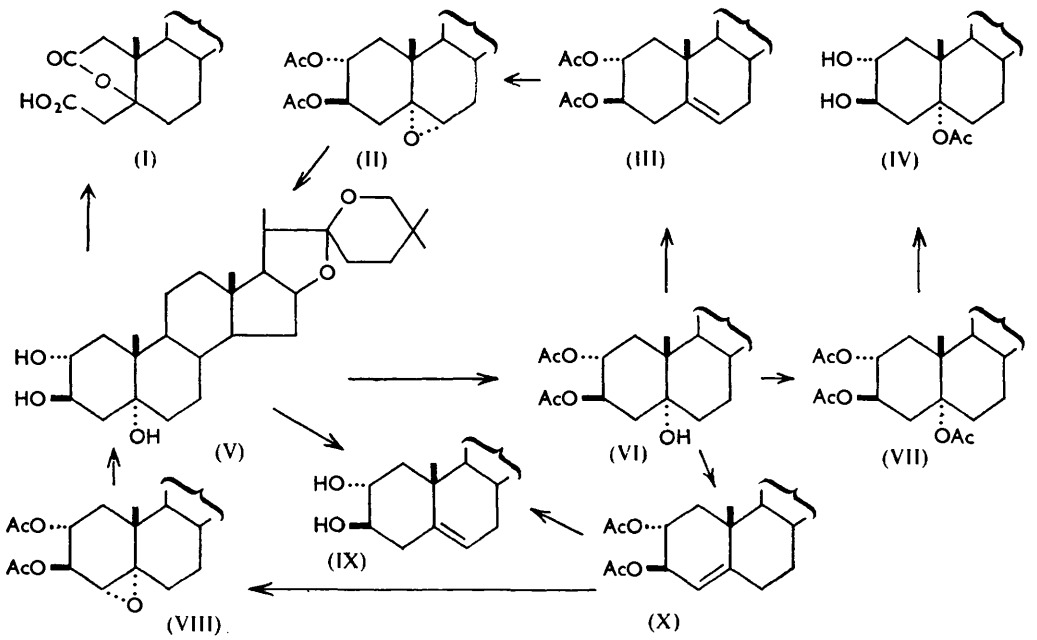
The 5α-position assigned earlier [1] to the tertiary hydroxyl group in agapanthagenin (V) (22*a*-spirostan-2α : 3β : 5α-triol) has been conclusively proved.

DURING oxidation of agapanthagenin (V), a (tertiary) 5-hydroxyl group should lactonise with the carboxyl group produced by oxidation of the hydroxymethylene group at position 2. Oxidation of agapanthagenin with chromic anhydride yielded a monobasic acid whose analysis and molecular weight agree with those of the lactone (I). It has been shown [2] that a 5-hydroxyl group can be acetylated only if it has the α-configuration. Acetylation of the tertiary hydroxyl group in agapanthagenin diacetate (VI) has been accomplished. Partial hydrolysis of the agapanthagenin triacetate (VII) produced may be effected, yielding the 5α-monoacetate (IV). This is to be expected since the 2α-and 3β-acetoxyl groups are equatorial and the 5α is polar. The 5α-hydroxyl group, being polar, should be readily eliminated with a β-coplanar hydrogen from position 4 or 6, giving rise to one of the diacetates (X) and (III). Both these compounds have been isolated by the action of thionyl chloride on agapanthagenin diacetate (VI) in pyridine. The Δ^4-compound (X) is practically the only product if not more than 2 mols. of thionyl chloride

[1] T. Stephen, *J.*, 1956, 1167.
[2] Plattner, Tetrzilka, and Lang, *Helv. Chim. Acta*, 1944, **27**, 513.

are used at 0°. At 10° in the presence of a large excess of thionyl chloride, the proportion of the isomer (III) is increased. This suggests that under the experimental conditions the former undergoes an isomeric change, and this has been brought about by treatment in chloroform with hydrogen chloride.

Partial dehydration of agapanthagenin itself may be effected by 13% alcoholic potassium hydroxide at 20°, the product being a mixture of the 5-ene-2α : 3β-diol (IX) and unchanged agapanthagenin. In accordance with this, hydrolysis of the diacetate (VI) with alcoholic potassium hydroxide yields the diol (IX).



The Δ^5-diacetate (III) with monoperphthalic acid gives a single epoxide, formulated as (II) by analogy with the major product of peroxidation of cholesterol.[3] Reduction of this epoxide with lithium aluminium hydride yielded agapanthagenin and a trace of an unidentified compound. In conformity with previous investigations [4] the Δ^4-diacetate on epoxidation yields a single epoxide (VIII). Reduction with lithium aluminium hydride again yielded agapanthagenin as the only product. Reduction of the two epoxides to agapanthagenin conclusively establishes the position of the tertiary hydroxyl group as 5α.

Experimental

Rotations were determined for EtOH solutions at 22°. M. p.s were determined on a block. The alumina used for chromatography was washed and activated at 180°. Light petroleum used had b. p. 40—60°.

2α : 3β : 5-*Triacetoxy*-22α-*spirostan* (VII).—Agapanthagenin diacetate (1·8 g.) was refluxed in chloroform (20 c.c.) with acetyl chloride (4 g.) and dimethylaniline (6·4 g.) for 20 hr. Water was added, the chloroform removed under reduced pressure, excess of acid neutralised with sodium hydrogen carbonate, and the *product* extracted with ether. This was chromatographed on alumina (30 g.) with light petroleum–benzene (3 : 2) and crystallised from methanol in plates, m. p. 202°, $[\alpha]_D$ −0·26° (*c* 4·1) (Found : C, 69·3; H, 8·7. $C_{33}H_{50}O_8$ requires C, 69·0; H, 8·7%).

[3] Plattner and Lang, *Helv. Chim. Acta*, 1944, **27**, 1872.
[4] Roberts, Shoppee, and Stephenson, *J.*, 1954, 3178.

22α-*Spirostan*-2α : 3β : 5α-*triol* 5α-*Monoacetate* (IV).—To a solution of agapanthagenin triacetate (1 g.) in methanol (45 c.c.) finely powered potassium hydroxide (2·5 g.) was added with shaking until a clear solution was obtained. After 30 min. at 20°, the *product* separated; it recrystallised from methanol as needles, m. p. 230°, $[\alpha_D]$ −0·16° (*c* 4·4) (Found: C, 71·0; H, 9·3. $C_{29}H_{46}O_6$ requires C, 71·0; H, 9·4%).

2α : 3β-*Diacetoxy*-22α-*spirost*-4-*en* (X).—To a solution of agapanthagenin diacetate (4 g.) in pyridine (10 c.c.) at 0°, thionyl chloride (1 c.c.) in pyridine (5 c.c.) was added dropwise with shaking; the mixture was kept at 0° for ¾ hr., then acidified with dilute hydrochloric acid and extracted with ether. The *product* was chromatographed on alumina (50 g.) with light petroleum–benzene (1 : 4) and crystallised from methanol in plates, m. p. 212—214°, $[\alpha]_D$ −0·42° (*c* 3·2) (Found: C, 72·3; H, 8·8; Ac, 16·8. $C_{31}H_{46}O_6$ requires C, 72·4; H, 8·9; Ac, 16·7%).

2α : 3β-*Diacetoxy*-22α-*spirost*-5-*en* (III).—The previous experiment was repeated but with 4 mols. of thionyl chloride and at 10°. The product, after ether-extraction, was chromatographed on alumina (50 g.). Elution with light petroleum–benzene (4 : 1) gave the diacetate (III) (60%), which crystallised from methanol in needles, m. p. and mixed m. p. with yuccagenin diacetate 178°. Further elution with light petroleum–benzene (1 : 4) gave the isomer (X) (40%), which crystallised from methanol in plates, m. p. 212°.

22α-*Spirost*-5-*ene*-2α : 3β-*diol* (IX).—To a solution of agapanthagenin (3 g.) in methanol (180 c.c.) finely powdered potassium hydroxide (24·8 g.) was added with stirring until a clear solution was obtained. After 48 hr. at 20°, this yielded the diol (IX), m. p. and mixed m. p. with yuccagenin 248°, and unchanged agapanthagenin. Refluxing the acetate (X) with alcoholic 5% potassium hydroxide for ½ hr. yielded the diol (IX), m. p. and mixed m. p. 248°.

2α : 3β-*Diacetoxy*-4α : 5α-*epoxyspirostan* (VIII).—To the diacetate (X) (1·4 g.) ethereal monoperphthalic acid (21 c.c.; 0·05 g. per c.c.) was added and the mixture heated under reflux for 7 hr. The ether was removed under reduced pressure, the residue digested with dry chloroform (25 c.c.), and after filtration the chloroform removed. The residue, chromatographed on alumina (30 g.), gave with light petroleum–benzene (3 : 2) an eluate which crystallised from methanol in needles, m. p. 222—224°, $[\alpha]_D$ −0·22° (*c* 1·8) (Found: C, 70·5; H, 8·8. $C_{31}H_{46}O_7$ requires C, 70·2; H, 8·6%). Reduction of this epoxide (0·10 g.) in dry ether (20 c.c.) with lithium aluminium hydride (0·08 g.) gave agapanthagenin, m. p. and mixed m. p. 284°. A second product, m. p. 230°, was isolated from the reduction product but not characterised.

2α : 3β-*Diacetoxy*-5α : 6α-*epoxyspirostan* (II).—Epoxidation of the diacetate (III) (1·6 g.) was carried out as in the previous experiment, with ethereal monoperphthalic acid (30 c.c.; 0·05 g. per c.c.). The product, chromatographed on alumina (20 g.) with light petroleum–benzene (4 : 1), crystallised from methanol in needles, m. p. 204—205°, $[\alpha]_D$ −0·47° (*c* 3·65) (Found: C, 70·6; H, 8·9. $C_{31}H_{46}O_7$ requires C, 70·2; H, 8·6%). Reduction of this epoxide (0·12 g.) with lithium aluminium hydride (0·09 g.) yielded only agapanthagenin, m. p. and mixed m. p. 284°.

5α-*Hydroxy*-2 : 3-seco-22α-*spirostan*-2 : 3-*dioic Lactone* (I).—To agapanthagenin (3 g.) in acetic acid (100 c.c.), chromic anhydride (2 g.) in acetic acid (80%) was added. The mixture was kept at 20° for 45 min. After addition of ethanol (10 c.c.), the mixture was made alkaline with aqueous ammonia, then slightly acidified with hydrochloric acid and extracted with ether. The *lactone*, crystallised from methanol, had m. p. 250°, $[\alpha]_D$ −0·16° (*c* 4·05) (Found: C, 70·7; H, 8·9%; equiv., 456. $C_{27}H_{40}O_6$ requires C, 70·5; H, 8·8%; equiv., 460).

One of us (T. S.) acknowledges a grant from the South African Council for Scientific and Industrial Research.

HOWARD COLLEGE, UNIVERSITY OF NATAL, DURBAN.
NATAL TECHNICAL COLLEGE, DURBAN.

[*Received, June 29th,* 1956.]