

## Microbiological 18-Hydroxylation of Steroids

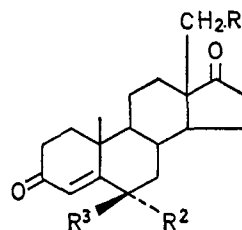
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**Summary** Incubation of androst-4-ene-3,17-dione with *Aspergillus niger*, ATCC 9142, gives 18-hydroxyandrost-4-ene-3,17-dione.

MICROBIOLOGICAL 18-hydroxylation is unusual; it occurs in the case of certain steroids using *Cercospora melonis*<sup>1</sup> and *Corynespora cassiicola*.<sup>2</sup> 18-Hydroxylation has not previously been observed with *Aspergillus niger*, although *A. niger*, ATCC 9142, has been reported<sup>3</sup> to effect hydroxylation of the 21-methyl group of progesterone and derivatives, and 17- and 21-hydroxylation of progesterone have been observed<sup>4</sup> with *A. niger*, S. 100.

We now report that incubation of androst-4-ene-3,17-dione (I), (1 g) with a 2-day growth of *A. niger*, ATCC 9142, in Czapek Dox medium for 4 days gives 18-hydroxyandrost-4-ene-3,17-dione (II), (0.487 g), m.p. 168—170° (from EtOAc-hexane),  $[\alpha]_D^{20} + 94^\circ$  ( $c = 0.8$  chloroform), and 6 $\beta$ -hydroxyandrost-4-ene-3,17-dione (III), (0.016 g). The known compound (III) was identified by m.p., rotation, and t.l.c. comparison with an authentic sample. Compound (II) has been reported<sup>5</sup> as a product of a chemical transformation and has been characterised as the acetate (IV).



- (I)  $R^1=R^2=R^3=H$   
 (II)  $R^1=OH; R^2=R^3=H$   
 (III)  $R^1=R^2=H; R^3=OH$   
 (IV)  $R^1=OAc; R^2=R^3=H$

The structure of (II), resulting from the microbiological reaction, was confirmed by conversion into the known acetate (IV), m.p. 127—129°, and by the following spectral data: mass spectrum,  $m/e$  (%), 302 (100), 287 (9) ( $M-Me$ ), 284 (10) ( $M-H_2O$ ), and 271 (5) ( $M-CH_2OH$ ),  $M^+$  302.1887; i.r. spectrum included bands at 3400, 1745, 1675, 1620, 1050, and 870  $cm^{-1}$ ; n.m.r. (100 MHz) absorptions included  $\tau$  4.24 (1H, s, 4-H), 8.77 (s, 19-Me), and 5.99 and 6.14 (AB, 2 non-equivalent 18-CH<sub>2</sub> protons,  $J$  9 Hz).

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