

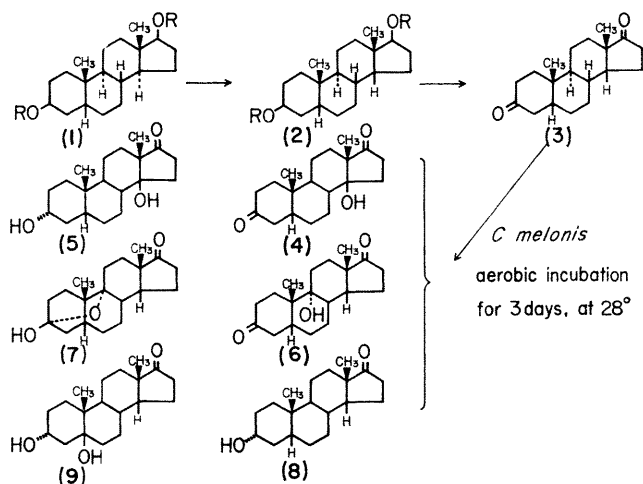
**Studies on the Transformation of Unnatural Steroids by Micro-organisms.
14 β -Hydroxylation of Androstane Derivative**

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Summary Incubation of 5 β ,14 β -androstane-3,17-dione with *Cercospora melonis* [*Corynespora melonis* (CKe) Lindau] afforded 3 α 14 β -dihydroxyandrostane-17-one, 3-hydroxy-3 α ,9 α -oxido-5 β -androstane-17-one, 3 α ,5 β -dihydroxyandrostane-17-one, and 3 α -hydroxy-5 β ,14 β -androstane-17-one

The vast literature on the microbiological hydroxylation of steroids refers almost entirely to substrates having a natural configuration.¹ We have undertaken an investigation of the



hydroxylation of 5β,14β-androstane-3,17-dione (C-D rings *cis*-fused) by *Cercospora melonis*, and the relevance of our results to the Oxford group's consideration² that the position of the keto-group may be the main factor determining the

site of attack, prompts this preliminary account of our studies.

In our experiments, 5β,14β-androstane-3,17-dione (3) was prepared by irradiation (low-pressure Hg lamp, emitting mainly at 253.7 nm) of 3β,17β-diacetoxy-5β-androstane (1; R = Ac) with mercuric bromide in cyclohexane,³ followed by hydrolysis in alkaline methanolic solution to give 5β,14β-androstane-3β, 17ξ-diol (2; R = H), m.p. 205–207°, $[\alpha]_D^{20} + 85.8^\circ$ (chloroform), ¹H n.m.r. τ : 8.98 (18-*H*₃), 9.06 (19-*H*₃), which was then oxidized with Jones' reagent to (3), m.p. 170–171°, $[\alpha]_D^{22} + 109^\circ$, ¹H n.m.r. τ : 8.91 (18-*H*₃), 8.98 (19-*H*₃). Transformation of (3) by *Cercospora melonis* under Kondo's conditions⁴ produced 14β-hydroxy-compounds (4) and (5) (22.5% yields) plus other hydroxy-derivatives (6), (7), and (9). All the products are new compounds.†

A combination of chemical transformations and spectroscopic examination established the positions and configurations of the hydroxy-groups. The assignments‡ of the C-18 (ΔC-19) methyl group signals in the ¹H n.m.r. spectra of the 14β-hydroxy-compound, 3α-acetoxy-14β-hydroxy-5β-androstan-17-one (10), m.p. 224–226°, $[\alpha]_D^{24} + 22.4^\circ$ (chloroform), and the corresponding 14α-hydroxy-compound (11)⁴ were readily confirmed by use of Eu(dpm)₃-induced shifts.⁵

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† All products gave satisfactory analytical data.

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¹ "Microbial Transformation of Steroids," W. Charney and H. L. Herzog, Academic Press, New York, 1967, pp. 18–36.
² J. E. Bridgeman, J. W. Browne, P. C. Cherry, (Miss) M. G. Combe, J. M. Evans, Sir Ewart R. H. Jones, A. Kasal, G. D. Meakins, Y. Morisawa, and P. D. Woodgate, *Chem. Comm.*, 1969, 463; A. B. Turner, *Ann. Reports (B)*, 1969, 66, 389.

³ M. Gorodetsky and Y. Mazur, *J. Amer. Chem. Soc.*, 1968, 90, 6540.

⁴ E. Kondo and T. Mitsugi, *Tetrahedron*, 1967, 23, 2153.

⁵ Cf. P. V. Demarco, T. K. Elzey, R. Burton, and E. Wenkert, *J. Amer. Chem. Soc.*, 1970, 92, 5737.