Fungal Metabolism of Some Methyl Dehydroabietanes

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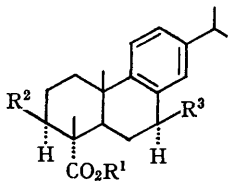
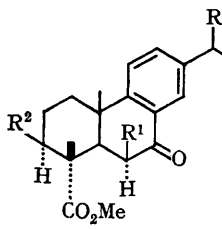
BIELLMANN *et al.*[1] have recently shown that incubation of dehydroabietic acid (I), with *Flavobacterium resinovorus* gives ketone (VI). They suggest that (VI) is found *via* enzymatic hydroxylation at C-3, and oxidation of the resulting alcohol to a keto-acid, which then undergoes decarboxylation.

We have found that incubation of methyl dehydroabietate (II) with *Corticium sasakii* (Lilly C-616) for 96 hr. gave a 12% yield of alcohol (III), and a 25% yield of diol (IV). Further incubation of (III) gave diol (IV). The stereochemistry of the hydroxyl at C-7 was shown to be β by incubation of (V) to give diol (IV), and by the characteristic shifts of the aromatic protons in the n.m.r. spectrum of (IV) caused by introduction of a 7β-hydroxyl group into (II).

The position and stereochemistry of the C-3β-hydroxyl group in alcohol (III) was shown by chemical and n.m.r. evidence. Deuteriation of ketone (XII), the oxidation product of (III), gave a dideuterium product as shown by n.m.r. and

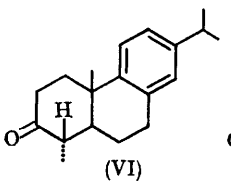
(I) $R^1=R^2=R^3=H$
(II) $R^1=Me, R^2=R^3=H$
(III) $R^1=Me, R^2=OH, R^3=H$
(IV) $R^1=Me, R^2=R^3=OH$
(V) $R^1=Me, R^2=H, R^3=OH$



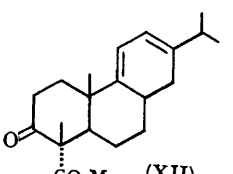


(VII) $R^1=R^2=H, R^3=R^4=Me$
(VIII) $R^1=R^2=OH, R^3=R^4=Me$
(IX) $R^1=H, R^2=OH, R^3=R^4=Me$
(X) $R^1=R^2=H; R^3, R^4=O$
(XI) $R^1=R^2=OH; R^3, R^4=O$

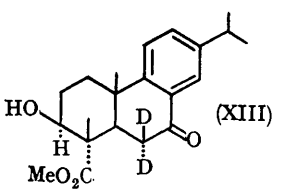
mass spectroscopy. Treatment of (XII) with base then acid gave ketone (VI), indicative of a β-keto-ester decarboxylation. The n.m.r. spectrum of (VI) shows a doublet at δ 76, J 7, for the C-4 methyl group. Selective chromate oxidation of diol (IV) gave the hydroxy-ketone (IX). Deuteriation of (IX) gave (XIII). Analysis of the n.m.r. spectrum of (XIII) gives A-ring proton-coupling constants which would be expected only if the C-3 hydroxyl were in the equatorial position. The accuracy of these spectral assignments was verified by their use in a computer calculation of a theoretical spectrum for (XIII) using the Frequint IV † program. The width and multiplicity of the C-3α proton in the theoretical spectrum closely matches that in the observed spectrum. The reduction of ketone (XII) with $NaMe_3BH$ to give only (III) confirms this conclusion. Incubation of (VII) with *C. sasakii* gave (IX) and diol (VIII). Further incubation of (IX) gave diol (VIII), showing the identity of the C-3β hydroxyl group in (III), (IV), (IX), and (VIII). The n.m.r. of (VIII) shows the C-17 methyl group shifted downfield 12 c./sec., relative to the C-17 methyl group in the n.m.r. spectrum of (IX), indicative of a C-6β-hydroxyl group.[2] The n.m.r. spectrum of (VIII) also shows the C-5α proton at δ 2·45, J 4 coupled to the C-6α proton. As in the incubation of (VII), incubation of (X) gave diol (XI).



(VI)



(XII)



(XIII)

The hydroxylation of dehydroabietanes at C-3 is consistent with the hypothesis of Barton and Moss[3] of proton-initiated terpene cyclization with hydroxylation at C-3 occurring at a later stage. Three *Juniperus* tree dehydroabietanes cogeneric with ferruginol; hinokiol, sugiol, and proxanthoperol, contain oxygen functions in the C-3, C-6, or C-7 positions. The fungal hydroxylation of dehydroabietanes in the same positions which bear oxygen in dehydroabietanes obtained from *Juniperus* trees provides an example of a fungus and a higher plant possessing the same oxidase selectivity.

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[1] J. F. Biellman, B. P. Daste, M. Raynaud, and R. Wenning, *Chem. Comm.*, 1968, 168.
[2] D. R. Brannon, F. W. Parrish, B. J. Wiley, and L. Long, jun., *J. Org. Chem.*, 1967, **32**, 1521.
[3] D. H. R. Barton and G. P. Moss, *Chem. Comm.*, 1966, 261.