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REGIOSPECIFIC HYDROXYLATION OF ACYCLIC MONOTERPENE ALCOHOLS BY ASPERGILLUS NIGER

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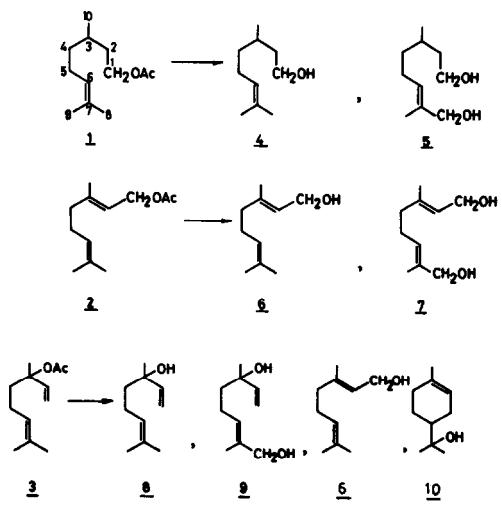
Abstract: Aspergillus niger was shown to carry out the regiospecific hydroxylation of acyclic monoterpene alcohols.

Selective oxidation of a terminal allylic methyl group in polyunsaturated system by selenium dioxide has been reported [1-3]. Following this method, the 8-hydroxy derivaties of acyclic monoterpene alcohols have been prepared, but in poor yields.[4] We explored the possibility of using a microbial system as a reagent to carry out the regiospecific hydroxylation and a search for such a system led to the isolation of a fungal strain, identified as Aspergillus niger.

Fermentation of acetates of citronellol, geraniol and linalool with A.niger resulted in their hydrolysis to the corresponding alcohols which were further hydroxylated to their respective 8-hydroxy derivatives (5,7,9). The amount of 8-hydroxy derivaties formed were comparatively lower when terpene alcohols were used instead of their corresponding acetates as substrates. This is due to the surface toxic effects of acyclic monoterpene alcohols[5]. The study has demonstrated the exclusive hydroxylation of acyclic monoterpene alcohols by A.niger at the C-8 methyl group and this type of oxidation mediated by a fungal system has not been observed before.

Fermentation were carried out in modified Czepek Dox medium (PH 7.0) at 29-30°C following standard procedures[6] (300 mg of substrate in 100 ml of medium). Metabolites were extrated from the broth with ether and were separated by



1 4 5 2 6 7 3 8 9 6 10

chromatography on a silica gel column using 5-40 % ethyl acetate in hexane. The levels of metabolites formed were estimated by g.l.c. analyses.

Fermentation of (1) with A.niger resulted in the formation of a major metabolite (5) accounting for approximately 60 % of the total transformation products, accompanied by 38 % of metabolite (4). Fermentation of (2) with A.niger gave metabolites (6) and (7) which were 50 and 40 % of the total transformation products respectively. However, in the case of (3), besides metabolites (8) and (9) which were formed to the extent of 25 and 45 % of the total products formed respectively, small amounts of geraniol (6) and α-terpineol (10) (together 25 %) were also formed. Nearly 40 % of (1), (2) and (3) were metabolized in 72h. All the metabolites have been characterized by comparison (i.r., n.m.r., t.l.c., and g.l.c.) with authentic samples. Authentic 8-hydroxy derivatives of (4), (6) and (8) were prepared as reported earlier[7,8]. The n.m.r. spectra[9] of metabolites 5, 7, and 9 confirmed that the fungal system carries out the hydroxylation at the C-8 position indicating its regiospecificity. Hydroxylases which exhibit high stereo and regio-selectivity have attracted much biotechnological interest.

References and Footnotes

(1) V.M. Sathe, K.K. Chakravarti, M.V. Kadival and S.C.Bhattacharyya, Indian J. Chem., 4, 393 (1966).

(2) G. Buchi and H. Wuest, Helv. Chim. Acta., 50, 2440 (1967).

(3) U. T. Bhalerao, J.J. Plattner and H. Rapoport, J.Amer.Chem.Soc., 92, 3429 (1970).

(4) J. Meinwald, W.R. Thompson, T.Eisner and D. F. Owen Tetrahedron Letters, 3485, (1971).

(5) S. G. Cantwell, E. P. Lau, D. S. Walt and R.R. Fall, J. Bacteriol, 135, 324 (1978).

(6) C. R. Jayanthi, P. Madyastha and K. M. Madyastha, Biochem. Biophys. Res. Commun., 106, 1262 (1982).

(7) S. Escher, Ph.D. Dissertation No. 4887, Eidgenossischen Technischen Hochschule, Zurich, Switzerland.

(8) K. M. Madyastha, T. D. Meehan and C.J. Coscia, Biochemistry, 15, 1097 (1976).

(9) 5 ^{1}H NMR ($CDCl_3$) δ 0.9 (3H, d, J=6 Hz, H-10), 1.1 - 1.75 (m, H-2, H-3, H-4) and 1.66 (br s, H-9) together 8H, 2.05 (2H, m, H-5), 2.6 (2H, br s, 2 OH),3.65 (2H,t, J=7 Hz, H-1), 3.95 (2H, br s, H-8), 5.4 (1H, t, J=7 Hz, H-6).

7 ^{1}H NMR ($CDCl_3$) δ 1.66 (6H, br s, H-9, H-10), 2.0 - 2.5 (6H, m,H-4, H-5 and 2 OH), 3.95 (2H, br s, H-8), 4.15 (2H, d, J=7 Hz, H-1), 5.4 (2H, t, J=7 Hz, H-2, H-6).

9 ^{1}H NMR ($CDCl_3$) δ 1.3 (3H, s, H-10), 1.4-1.8 (m, H-4) and 1.66 (s, H-9) together 5H, 1.9-2.3 (4H, m, H-5 and 2 OH), 4.0 (2H, s, H-8), 4.9 - 5.5 (3H, m, H-1, H-6), 5.95 (1H, dd, 17, 10 Hz, H-2).

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