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SHORT COMMUNICATION

THE BIOSYNTHESIS OF (−)-13-EPIMANOYL OXIDE

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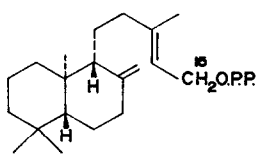
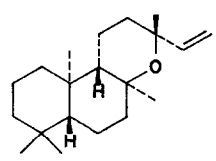
Abstract—15-^{3}H-(−)-labda-8,13-dien-15-ol pyrophosphate is specifically incorporated into (−)-13-epimanoyl oxide by *Gibberella fujikuroi*.

INTRODUCTION

THE BICYCLIC diterpenes may be classified into those biogenetically related to a labdadienol, those possessing the vinyl:methyl substitution related to manöol and those possessing a furanoid side chain. In addition representatives of each type are found with a labdane or clerodane nucleus. The geraniol : linalöol type of isomerism exhibited by



(I)



(II)

(PP = pyrophosphate)

the first two classes raises an interesting question. The manöol : manoyl oxide group of diterpenes could arise by the cyclization of geranyl-linalöol in which even the oxide ring formation might be concerted with the initial cyclization. Alternatively this series could arise by allylic rearrangement of a labdadienol pyrophosphate formed by cyclization of geranyl-geraniol pyrophosphate.

RESULTS

(−)-13-Epimanoyl oxide (olearyl oxide)[1] (II) is a trace metabolite of the fungus, *Gibberella fujikuroi*.[2] However, 15-^{3}H-(−)-labda-8,13-dien-15-ol pyrophosphate (I) was fed to the fungus and it was possible to isolate the oxide by dilution analysis. It showed a 0·0012% incorporation. Under similar conditions (−)-kaurene showed an incorporation of 0·076%. Ozonolysis of the oxide led to the recovery of 98·5% of the radioactivity from C-15 as formaldehyde thus demonstrating that the incorporation was specific. Although this is a low incorporation, bearing in mind the relative amounts of (−)-kaurene and 13-epimanoyl oxide produced by the fungus, it is of the expected order of magnitude. Thus the pathway involving rearrangement after cyclization is contributing to the biosynthesis of (−)-13-epimanoyl oxide in *Gibberella fujikuroi*. It is interesting to note that the C-13 stereochemistry,

[1] D. H. MCLEAN and S. N. SLATER, *Chem. & Ind.* **64**, 28 (1945).
[2] B. E. CROSS, R. H. B. GALT and J. R. HANSON, *J. Chem. Soc.* 2937 (1963).

β-methyl: α-vinyl, of this oxide is the same as that of the pimaradiene intermediate proposed in the cyclization of the labdadienol pyrophosphate to (−)-kaurene.

EXPERIMENTAL

General details have been described previously.[3]

Feeding of 15-^{3}H-(−)-*labda*-8,13-*dien*-15-*ol pyrophosphate*. The pyrophosphate (4 mg, $4{\cdot}25 \times 10^5$ dis/sec/mg) was distributed between 30 flasks of *Gibberella fujikuroi* grown as described previously.[4] After 14 days the metabolites were isolated by extraction with ethyl acetate. The combined neutral and acidic fractions were chromatographed on silica gel. Elution with light petroleum gave a gum which was diluted with (−)-13-epimanoyl oxide (15 mg). Further purification by TLC gave (−)-13-epimanoyl oxide which was repeatedly crystallized from MeOH as plates, m.p. 99° (lit.,[2] 98–99·5°) (79 dis/min/mg) (22,600 dis/min/m-mole).

Ozonolysis of (−)-13-*epimanoyl oxide*. Ozonized oxygen was passed through a solution of the oxide (3 mg) in HOAc (3 ml) for 0·5 min. H_2O (10 ml) was added and the solution was shaken for 1 hr. It was then steam distilled and the distillate was passed through a solution of dimedone (20 mg) in MeOH (0·2 ml) and H_2O (2 ml). The formaldehyde dimethone (0·5 mg) crystallized from aq. EtOH as needles, m.p. 190–191° (76 dis/min/mg, 22,300 dis/min/m-mole. 98·5% of the activity of the oxide.)

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[3] B. ACHILLADELIS and J. R. HANSON, *Phytochem.* **7**, 589 (1968).
[4] J. R. HANSON and A. F. WHITE, *J. Chem. Soc.* **C**, 981 (1969).

Key Word Index—*Gibberella fujikuroi*; Fungus; biosynthesis; epi-manoyl oxide.