A Steroid Hydroxylase Inhibitor, Diplodialide A from *Diplodia pinea*

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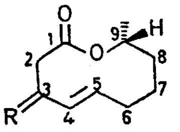
Summary The isolation and structural elucidation of three open-chain pentaketides, diplodialide A (a steroid hydroxylase inhibitor), diplodialide B and diplodialide C are reported.

IN the course of studies on steroid hydroxylase inhibitors produced by fungi, three polyketides, diplodialides A, B, and C were discovered in a culture filtrate of *Diplodia pinea* (IFO 6472) and diplodialide A showed inhibition of steroid 11α-hydroxylase in *Rhizopus stolonifer* (IFO 5781).[†] We now report the structural elucidation of diplodialides A (I), B (II), and C (III).

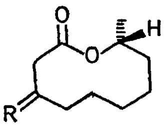
Diplodialide A (I) is a colourless oil, [$C_{10}H_{14}O_3$(M^+ 182); $(\alpha)_D^{26}$ + 142° ($CHCl_3$); ν (CCl_4) 1740, 1645, 965 cm^{-1}; λ (MeOH) 232 (ε = 6560), 310 nm (122)]. The n.m.r. spectrum ($CDCl_3$) shows the presence of a secondary methyl group ($\text{Me–}\overset{|}{\text{C}}\text{H–O–CO–}$) (δ 1·28, 3H, d, *J* 7Hz; 5·16, 1H, m), a methylene flanked by two carbonyl groups (2H AB

[†] Diplodialide A inhibited 11α-hydroxylase of progesterone in vegetable cell cultures of *Rh. stolonifer* at 125 p.p.m. The biological study will be reported in detail elsewhere.



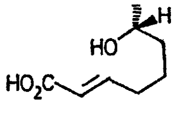
(I); R = O

(II); R = OH,H



(III), R = OH, H

(IV); R = O



(V)

quartet centred at 3·56, *J* 14 Hz), and a *trans*-disubstituted olefin conjugated with a carbonyl group (–CO–CH=CH–CH_2–) (5·88, 1H, d, *J* 16 Hz; 6·72, 1H, m). These partial structures have been confirmed by double resonance experiments. Hydrogenation of (I) with Pd–C in ethyl acetate gave dihydro-A (IV), [$C_{10}H_{16}O_3$ (M^+ 184); ν (CCl_4) 1745, 1715 cm^{-1}; δ ($CDCl_3$) 1·30 (3H, d, *J* 7 Hz, Me–CH–O–), 0·8–2·4 (10H, m, methylenes), 3·40 (2H, s, –CO–CH_2–CO–), 5·10 (1H, m, –CH–O–CO–); no other signals lower than 4·0] The single additional unsaturation shown in the empirical formula of (I) was thus attributable to the presence of a ring. The u.v. spectrum of (IV) (end absorption) showed a bathochromic shift to 277 nm (ε = 12500) in an alkaline solution which is reversed by acids. This bathochromic shift indicates the presence of β-diketone or β-ketoester in (IV). (I) was hydrolyzed by KOH in MeOH to give a hydroxy acid (V), which might be formed by a ketone cleavage of the β-ketolactone group in (I). Diplodialide B (II) has $C_{10}H_{16}O_3$ (M^+ 184·108: calcd. 184·110), and shows $(\alpha)^{26}_{\mathrm{D}}$ −37·3 ($CHCl_3$); ν (CCl_4) 3550, 1715 cm^{-1}. Diplodialide B was oxidized with active MnO_2 in CH_2Cl_2 to give (I), and (I) gave diplodialide B with $NaBH_4$ in tetrahydrofuran.

Diplodialide C (III), [$C_{10}H_{18}O_3$ (M^+−H_2O 168); $(\alpha)^{28}_{\mathrm{D}}$ −41·0° ($CHCl_3$); ν (CCl_4) 3650, 3450, 1730 cm^{-1}] was identical with the dihydroderivative of (II). The stereochemistry of the asymmetric carbon at position 9 was established by ozonolysis of (II), followed by $LiAlH_4$ reduction. As expected, an optically active 1,5-hexandiol, [$C_6H_{14}O_2$ (M^+−Me 103); $(\alpha)^{26}_{\mathrm{D}}$ −11° (MeOH)] was obtained. The optical rotation has the opposite sign to the value reported for (+)-(*S*)-1,5-hexandiol, $(\alpha)^{20}_{\mathrm{D}}$ + 8° (MeOH).[1]

The di-*p*-nitrobenzoate, m.p. 110—110·5°; $(\alpha)^{26}_{\mathrm{D}}$ − 39·7° ($CHCl_3$) also shows the opposite sign to the optical rotation of (+)-(*S*)-1,5-hexandiol di-*p*-nitrobenzoate, $(\alpha)^{20}_{\mathrm{D}}$ + 43° ($CHCl_3$). The formation of (−)-(*R*)-1,5-hexandiol indicates 9*R*-configuration for diplodialides.

The same stereochemistry of the lactone ring in polyketides has been reported for ochratoxins[2] and 5-methylmellein.[3] Diplodialides appear to be the first members of open-chain pentaketides.[4]

The authors thank A. Miyazaki for the high-resolution mass spectra.

(*Received*, 31*st December* 1974; *Com.* 1577.)

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