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# THE STRUCTURE OF PYRENOPHORIN

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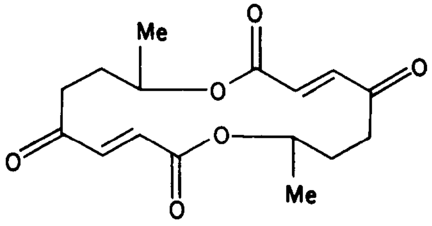
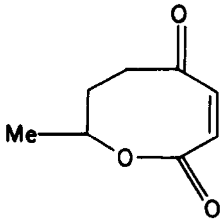
Pyrenophorin, the antifungal metabolic product of the plant pathogenic fungi *Pyrenophora avenae* and *Stemphylium radicinum*, is shown to be the symmetrical dimer (I) of (-)-7-hydroxy-4-oxo-oct-2-enoic acid lactone (II).



I



II

Pyrenophorin, m.p. 175°, $[\alpha]_D$ - 50°, was first isolated by Ishibashi[1] from culture filtrates of *P. avenae*, a pathogen of oat: in addition to its antifungal action[1,2] it showed cytostatic properties and was eventually assigned the formula $C_{24}H_{30}O_9$. Redetermination, from their mass spectra, of the molecular weights of pyrenophorin (308) and its tetrahydro-derivative (312) shows that the formula of pyrenophorin should be altered to $C_{16}H_{20}O_6$, ( $\lambda$ max 220 mμ, ε 23,200). The molecular weight obtained by the osmotic pressure method was 312 for pyrenophorin and 357 for its tetrahydro-derivative.

The antifungal metabolic product of *S. radicinum*, a pathogen of carrot, was considered to have the formula $C_8H_{10}O_3$[3] and structure (II) was proposed[4] on degradative evidence. Direct comparison of this product with pyrenophorin has shown the two compounds to be identical and pyrenophorin must therefore have the 16-membered ring dilactonic structure (I), the symmetrical dimer of (-)-7-hydroxy-4-oxo-oct-2-enoic acid lactone (II).

The product, m.p. 156°, of catalytic reduction[4] of the *S. radicinum* compound is thus tetrahydro-pyrenophorin and the oily product of reduction with sodium borohydride is also a tetrahydro-derivative [bis-3, 5-dinitrobenzoate, m.p. 230° (decomp.) (Found: C, 50.62; H, 4.18; N, 7.84; $C_{30}H_{28}O_{16}N_4$. $H_2O$ requires C, 50.14; H, 4.20; N, 7.80%)].

The $C_8H_{10}O_3$ formula was assigned on the basis of X-ray crystallographic measurements[3,4] (by Dr. P.G. Owston) assuming 4 asymmetric molecules in the orthorhombic unit cell of weight 614. The symmetry requirements of the Space Group, $P2_12_12$, are also satisfied by only 2 molecules of structure (I) which in itself has a two-fold axis of symmetry but no centre or plane of symmetry.

In the light of these results the most reasonable interpretation of the nuclear magnetic resonance spectrum of pyrenophorin favours a _trans_ arrangement of the olefinic protons (cf. [4]). This coupling constant ($J = 15.5$ c./sec.) is essentially unchanged in the product of borohydride reduction ($J = 15.4$ c./sec.) and its bis-3, 5-dinitrobenzoate ($J = 15.6$ c./sec.).

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