

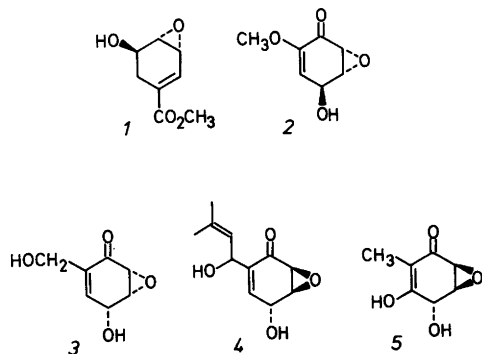
Structure of Chaloxone, Isolated from a *Chalara* Sp.

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Chaloxone, 5-hydroxy-3-methoxy-7-oxabicyclo-[4.1.0]hept-3-en-2-one, has been isolated from the fungus *Chalara microspora* (Corda) Hughes. Its structure has been determined with spectroscopic methods, mainly ^1H NMR. The absolute configuration has been established through CD measurements.

In a preceding paper ¹ we described the isolation of two fungal metabolites 1 and 2 from *Chalara microspora* (Corda) Hughes, and the structure and synthesis of 1. This paper deals with the structure of 2 for which we propose the name chaloxone. It falls into the family of previously isolated fungal metabolites, such as epoxydon 3 ^{2,3} (from a *Phoma* sp. and a *Phyllosticta* sp.), panepoxydon 4 ⁴ (from *Panus rudis* and *Panus conchates*), and terremutin 5 ^{5,6} (from *Aspergillus terreus*).



The compound was crystalline, with m.p. 142–143 °C. High resolution MS gave a molecular ion at 156.0401, corresponding to $\text{C}_7\text{H}_8\text{O}_4$. The molecule thus contains a total of four rings/double bonds. The UV spectrum showed a maximum at 265

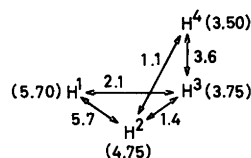


Fig. 1. ^1H NMR spectrum (acetone- d_6) of chaloxone. δ (ppm), J (Hz).

nm ($\epsilon = 5500$) characteristic of an α,β -unsaturated carbonyl chromophore, substituted with a heteroatom. IR showed the existence of $\text{C}=\text{O}$ (1690 cm^{-1}), $\text{C}=\text{C}$ ($3080, 1640\text{ cm}^{-1}$) and $\text{O}-\text{H}$ (3480 cm^{-1}).

100 MHz ^1H NMR gave the chemical shifts and coupling constants shown in Fig. 1. In addition, the spectrum showed a CH_3O -group at δ 3.60, and an OH -group at δ 2.9. H^1 is the only vinyl hydrogen, and is vicinally coupled to H^2 . This means that H^1 must be in the β -position of the double bond, and the low δ -value is caused by a CH_3O -substituent in the α -position. The shift δ 3.60 for the CH_3O -group is consistent with its attachment to the α -position of

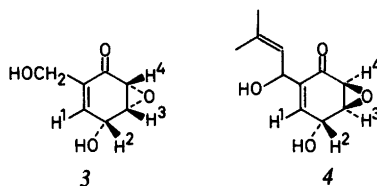
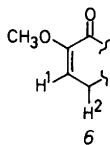
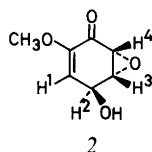


Fig. 2. Epoxydon 3: ^1H NMR [$(\text{CD}_3)_2\text{SO}$]: δ 6.42 (H^1), 4.72 (H^2), 3.77 (H^3), 3.40 (H^4); J (Hz): $\text{H}^1-\text{H}^2 = 2.5$, $\text{H}^1-\text{H}^3 = 2.5$, $\text{H}^2-\text{H}^3 = 2.5$, $\text{H}^2-\text{H}^4 = 0$, $\text{H}^3-\text{H}^4 = 4$.

Panepoxydon 4: ^1H NMR (CDCl_3): δ 6.7 (H^1), 4.65 (H^2), 3.8 (H^3), 3.45 (H^4); J (Hz): $\text{H}^1-\text{H}^2 = 5$, $\text{H}^1-\text{H}^3 = 2.5$, $\text{H}^2-\text{H}^3 = 1$, $\text{H}^2-\text{H}^4 = 1$, $\text{H}^3-\text{H}^4 = 3.5$.



the double bond. This gives the partial structure shown in 6. The rest of the structure was settled by a comparison of chemical shifts and coupling constants with those of epoxydon $3^{2,4}$ and panepoxydon 4^4 (Fig. 2). The observed NMR data are very close to those for panepoxydon (as drawn in 4 and its enantiomer), thus establishing the remainder of the structure of chalozone 2.



Panepoxydon 4 can exist in two conformations, *A* and *B* (Fig. 3). *A* is favoured by the parallel orientation of the epoxide ring and the π -electrons of the carbonyl group, giving maximum overlap. 3,5,6 Also, in *A* the eclipsing interactions between H^2/OH and H^3 are minimized. By the same arguments, the same conformation would be preferred by chalozone 2.

The CD spectrum shows a positive Cotton effect at 330 nm, corresponding to the $n-\pi^*$ transition. Epoxydon 2 showed a positive Cotton effect in the same region, while panepoxydon 4 and terremutin 5 showed a negative Cotton effect. All these compounds have previously been assigned the absolute configurations shown by comparison with several others containing the 5,6-epoxy-cyclohex-2-en-one structure. Chalozone should then have the same absolute configuration as epoxydon 3, and should be as is drawn in 2.

Chalozone is the first member in this group of fungal metabolites that does not have an alkyl substituent attached to the six-membered ring.

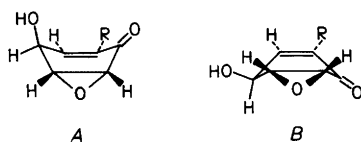


Fig. 3. Panepoxydon, $R = \text{CHOHCH} = \text{C}(\text{CH}_3)_2$. Chalozone (other enantiomer), $R = \text{OCH}_3$.

EXPERIMENTAL

Melting points were determined on a Kofler micro hot stage and are uncorrected. UV spectra were recorded on a Bausch & Lomb Spectronic 505 spectrometer. IR spectra were recorded on a Perkin Elmer model 257 infrared spectrometer. ^1H NMR spectra were recorded on a JEOL MH 100 instrument, and CD spectra were obtained on a JASCO J-41A instrument. Specific rotations were determined from ORD spectra recorded on a Cary 60 instrument. Merck Silica gel 60 F_{254} aluminum sheets were used for TLC.

The cultivation of the fungus and the isolation of chalozone were described in a preceding paper. 1

Chalozone, m.p. 142–143 °C; $[\alpha]_D^{20} + 271^\circ$ (*c* 0.56, abs. ethanol); $R_F = 0.25$ (silica/ether); High resolution MS [70 eV; *m/e* (rel. int.)]: 156.0401 (6, $\text{C}_7\text{H}_8\text{O}_4$), 128.0458 (8, $\text{C}_6\text{H}_8\text{O}_3$), 127.0386 (31, $\text{C}_6\text{H}_7\text{O}_3$), 113.0258 (28, $\text{C}_5\text{H}_5\text{O}_3$), 69 (100); IR (KBr): 3480, 3080, 3020, 2980, 2945, 2910, 2860, 1690, 1640, 1460, 1430, 1390, 1080 cm^{-1} ; UV [abs. ethanol (ϵ)] 214 (3000), 265 (5200); CD (*c* 0.56, abs. ethanol): $[\theta]_{200} 0$, $[\theta]_{220} -9500$, $[\theta]_{238} 0$, $[\theta]_{270} 24800$, $[\theta]_{300} 5000$, $[\theta]_{330} 9500$, $[\theta]_{395} 0$. ^1H NMR data are given in the text.

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