# Some New Metabolites of *Aspergillus versicolor* and a Revised Structure for Averufin

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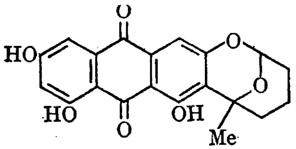
STRUCTURE (I) has been suggested for averufin, a metabolite of *Aspergillus versicolor* (Vuill.) Tiraboschi.[1] We have now shown that mutation of this organism, by ultraviolet irradiation of the spores, gives strains producing new metabolites. Three of these are orange optically active compounds m.p.'s 260°, 252—254°, and 219—220° for which we suggest structures (II), (III) and (IV) respectively.

The structural evidence for these compounds, which will be reported fully elsewhere, is based on the following observations and deductions:—

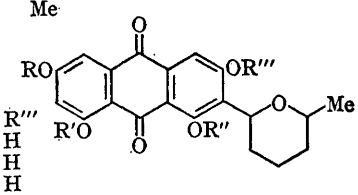
(a) All three compounds give the same fully methylated derivative (V).

(b) The chelated anthraquinone moiety was demonstrated by the facts that all three compounds gave intense red–brown ferric reactions and were reduced by alkaline dithionite to the corresponding leuco-compounds.

(c) The n.m.r. spectrum of the fully methylated derivative (V) showed *m*-coupled aromatic protons at $\tau$ 2·66 and 3·21 (*J*, 2·5 c./sec.) and a singlet at $\tau$ 2·46. Comparison of these values with those of similarly substituted anthraquinones[1,2] established the pattern of aromatic substitution.

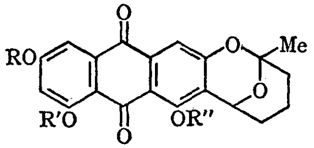


(I)

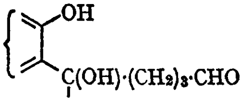


| | R | R′ | R″ | R‴ |
|---|---|---|---|---|
| (II) | H | H | H | H |
| (III) | H | Me | H | H |
| (IV) | Me | Me | H | H |
| (V) | Me | Me | Me | Me |

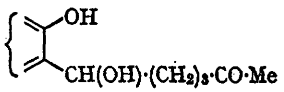
(d) The presence of the methyltetrahydropyran substituent was indicated by ozonolysis of compound (IV) which gave 6-methyltetrahydropyran-2-carboxylic acid,[3] characterised as the *p*-bromophenacyl ester, m.p. 112°, $[\alpha]_D$ + 2·5° (*c*, 5·08 in $CHCl_3$).



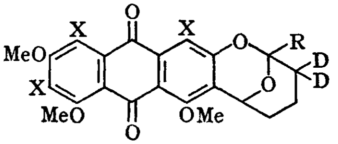
| | R | R′ | R″ |
|---|---|---|---|
| (VI) | H | H | H |
| (VII) | H | Me | H |
| (VIII) | Me | Me | H |
| (IX) | Me | Me | Me |



(X)



(XI)



(XII: R = $CHD_2$, X = D)
(XIII: R = $CD_3$, X = D)

These structures for the mutant metabolites led us to suspect that averufin ought to be related and have structure (VI) rather than (I). In our opinion none of the published evidence differentiates between these structures. We have re-isolated averufin and also, from further mutants, its mono- and di-methyl ethers which we believe have structures (VII) and (VIII) respectively.

It would be expected that on treatment of averufin with acid the cyclic ketal moiety in (VI) or the equivalent acetal in (I) would equilibrate with the corresponding open-chain hydrate, (XI) or (X) respectively. Consequently, in acid-catalysed deuteration, exchange of five hydrogens $\alpha$ to the carbonyl group in (XI), or two hydrogens in (X), would be expected. Hence, a compound of structure (VI) should give a pentadeutero- and structure (I) a dideutero-derivative. In practice maximum deuteration of the tri-*O*-methyl derivative (IX) was effected by a refluxing solution of phosphorus pentachloride in deuterium oxide and tetrahydrofuran for 3 days.[4] The exchange was accompanied by partial demethylation, but remethylation gave a product shown by mass spectrometry to be a mixture of two components containing seven and eight deuterium atoms respectively in an otherwise unchanged structure. Comparison of the n.m.r. spectrum of this material with that of the parent (IX) showed that all three aromatic protons had been completely exchanged in addition to four and five respectively in the cyclic ketal moiety. However, since the chemical shifts of the *C*-methyl and methylene groups overlap, it was not possible to state unequivocally that specific exchange had taken place at the required positions. This problem was resolved by studies of mass-spectral fragmentations. Thus, whereas the parent (IX) has large fragment ions, shown by accurate mass measurement to be due to losses of $CH_3$·CO·, $CH_3$·CO·$CH_2$·, $CH_3$·CO·$[CH_2]_2$·, and $CH_3$·CO·$[CH_2]_3$· from the ketal system, the octadeutero-derivative showed equivalent losses of $CD_3$·CO·, $CD_3$·CO·$CD_2$·,

$CD_3{\cdot}CO{\cdot}CD_2{\cdot}CH_2{\cdot}$, and $CD_3{\cdot}CO{\cdot}CD_2{\cdot}[CH_2]_2{\cdot}$ and the heptadeutero-derivative losses of $CD_2H{\cdot}CO{\cdot}$, $CD_2{\cdot}H{\cdot}CO{\cdot}CD_2{\cdot}$ etc. Hence, we deduce that the deuterated material is a mixture of compounds (XII) and (XIII), and accordingly, that averufin has structure (VI).

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