Fig. 2. Proposed dieldrin degradation pathways by the micro-organism in soil. The structures of metabolites G and F were established, F being one of the stable "terminal residues". The structures shown for E and the resulting acidic metabolite are the most likely ones. The structural arrangement of those two hydroxyl groups in metabolite D is still unknown except that one of them is at the 6 carbon position.

on the loss of one of the hydroxyl groups through dehydration. It cannot therefore be *trans*-6,7-dihydrodihydroxy-aldrin, which directly forms the peak group at 361 on fragmentation. Another prominent peak group was at 325, possibly resulting from M-2 (H_2O) fragmentation.

With the exception of aldrin, all the metabolites previously mentioned do not exist in nature, and it is therefore important to study their toxicological effects on various ecosystems in our environment. The quantity of the metabolites finally purified in this study was, however, not enough to permit fully fledged toxicological investigations. Metabolite F seemed to be one of the stable "terminal residues", and thus its toxicological meaning in relation to the problems of environmental contamination should be carefully assessed in the future.

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Isolation and Biological Activity of a Microbial Conversion Product of Aflatoxin B₁

ONE of the most striking and significant developments in synthetic chemistry has been the use of biological systems to achieve chemical transformations, especially the oxidation-reduction of steroids and alkaloids. Based on an analogous biological system, we have attempted to transform aflatoxin B₁, a toxic secondary metabolite of *Aspergillus flavus*, by employing a known steroid-hydroxylating micro-organism.

Our original objective was to produce the hydroxylated aflatoxin (aflatoxin M)¹ by incubating it in the presence of a known steroid-hydroxylating fungus (*Dactylium dendroides*)². In preliminary work on the hydroxylation of aflatoxin B₁, a new, blue-fluorescent compound was produced as demonstrated by thin-layer chromatography (TLC). We have tentatively designated this new derivative (Ro).

Dactylium dendroides NRRL 2575 was grown on a yeast extract-sucrose medium in 10–30 mg of crystalline aflatoxin B₁ per flask. After incubation for 48 h the culture flasks were extracted with chloroform; this crude extract contained aflatoxin and several other fluorescent compounds. Chromatography of the crude extract on a silicic acid column with 1 per cent ethanol in chloroform gave a brown fraction containing aflatoxins B₁ and Ro. These two compounds were separated on a column of 'Silica Gel G-HR' with 10 per cent acetone in chloroform. The eluate (Ro) was concentrated, the compound precipitated by hexane addition and the precipitate taken to dryness.

Compound (Ro) was originally thought to be aflatoxin B₂ because of an equivalent R_F value on 'Silica Gel G-HR' thin-layer chromatography plates when 10 per cent acetone in chloroform was used as developing solvent. Development of the plates with either 20 per cent acetone in chloroform or 2 per cent CH_3OH in chloroform, however, separated the two compounds. The ultraviolet spectrum of (Ro) showed maxima of 325, 261 and 254 $m\mu$ (in CH_3OH). Its infrared spectrum had an atypical coumarin nucleus as indicated by a loss in absorption at 1,760 cm^{-1} and, in addition, a broad band at 3,400 cm^{-1} that suggested a hydroxylated compound. The new isolate was light yellow and decomposed at 230°–234° C without melting.

Because of the concern about biological and chemical detoxification of aflatoxin in agricultural commodities, this new compound was tested for biological activity. Histopathological examination of bile duct hyperplasia⁴ of the livers from ducklings fed B₁ for 4 days, followed by death on the sixth day, has been used as a biological assay for the presence of the toxin, for a total dose of 2.5 μg can be reproducibly determined. In the conditions specified for the duckling biliary hyperplasia assay⁴, 56.0 μg B₁ had the same biological activity as 1.0 mg of the new conversion product, making this product eighteen times less toxic than aflatoxin B₁ by histopathological assay. Teunissen and Robertson³ have reported a modified B₁ aflatoxin produced by *Tetrahymena pyriformis* with a similar R_F value; however, toxicity studies on this compound were not performed.

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