

peroxide, undergoes little or no further change when aged at the same temperature.

One of the objects of publishing these results is to direct attention to the possibility of error arising during viscosity and other measurements performed on solutions of polymers in solvents able to form peroxides by autoxidation. Detailed results will be published elsewhere at a later date.

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² Sirlanni, Wise and McIntosh, *Can. J. Res.*, **25** B, 3, 301 (1947).

³ Naunton, *Trans. Farad. Soc.*, **38**, 332 (1942).

⁴ Lacau and Magat, *Farad. Soc. Discussion*, "The Labile Molecule" (Sept. 1947).

⁵ Cf. Mesrobian and Tobolsky, *J. Polymer Sci.*, **2**, 463 (1947).

⁶ Mead and Fuoss, *J. Amer. Chem. Soc.*, **64**, 277 (1942).

Trichothecin : an Antifungal Metabolic Product of *Trichothecium roseum* Link

ANTAGONISTIC activity to other fungi by *Trichothecium roseum* Link (syn. *Cephalothecium roseum* Corda) has been reported by Whetzel¹, Boning², Koch³ and Greaney and Machacek⁴. Culture filtrates of *T. roseum* have been shown to inhibit germination of *Botrytis allii* conidia⁵. We have isolated the substance responsible for this activity in crystalline form in yields of 20–30 mgm. per litre of culture filtrate. The name 'trichothecin' is suggested for the active substance.

The most satisfactory yields have been obtained by growth of the fungus for a period of 28 days at 25° C. on a modified Czapek-Dox medium, in which sodium nitrate was replaced by the equivalent quantity of ammonium tartrate. The medium contained 5 per cent of glucose and was supplemented by the addition of 1 per cent of corn-steep liquor. The development of antifungal activity was followed by means of a spore-germination assay using *Penicillium digitatum* conidia. Maximum concentration of trichothecin (40–50 mgm./l.) was attained after about 28 days.

Trichothecin was extracted from the culture filtrate with ether or chloroform and purified by chromatographic methods on activated alumina. It was finally recrystallized from light petroleum (b.p. 60–80°) and formed long, slender, colourless needles, m.p. 118°, which were optically active $[\alpha]_D^{25} + 44^\circ$ (c, 1 in chloroform). It contains neither halogens, sulphur nor nitrogen. Microanalyses (Weiler and Strauss), found: C = 68.5, 68.7, 68.8 per cent; H = 7.2, 7.3, 7.4 per cent; mol. wt. (Rast) = 278. C₁₅H₂₀O₄ requires: C = 68.2 per cent; H = 7.5 per cent; mol. wt. = 264. C₁₅H₁₈O₄ requires: C = 68.7 per cent; H = 6.9 per cent; mol. wt. = 262. The compound is neutral and only slightly soluble in water (c. 400 mgm./l. at 25°) but readily soluble in most organic solvents. The molecule contains one ketonic group and one ethylenic group. These observations are supported by the ultra-violet absorption spectrum of trichothecin, which also suggests that the two groups are conjugated. No other functional groups

have been detected. Methyl attached to carbon determination (Weiler and Strauss), found: (C)CH₃ = 16.4 per cent; 3 × (C)CH₃ groups require 17.0 per cent. There is no evidence of an aromatic nucleus.

At a concentration of 400 mgm./l. and pH 7.0, trichothecin was inactive against *Staph. aureus*, *B. subtilis* and *Bact. coli*. Its antifungal activity is exhibited against Fungi Imperfecti, Zygomycetes and Ascomycetes. The growth of each of some twenty-five species belonging to the above classes was inhibited to a greater or lesser degree. *P. digitatum* is the most sensitive species so far examined; germination of conidia was completely inhibited by 1.25 mgm./l. of trichothecin, and 50 per cent germination took place at a concentration of 0.30 mgm./l. The corresponding data for *Botrytis allii* spores are 6.25 and 3.12 mgm./l. Growth of the following fungi on beer wort agar was completely suppressed by the trichothecin concentrations given in parentheses: *P. digitatum* (0.64 mgm./l.), *Fusarium graminearum* (16 mgm./l.), *Paecilomyces varioti* (80 mgm./l.), *Saccharomyces carlsbergensis* (16 mgm./l.), and *Mucor erectus* (80 mgm./l.). *T. roseum* showed a partial reduction of growth at a concentration of 80 mgm./l.

Aqueous solutions of trichothecin were stable at pH 1 to pH 10 for at least 48 hours at 20°. At pH 12 the antifungal activity was rapidly destroyed even at room temperature. Aqueous solutions at pH 7 were maintained for at least one hour at 100° without detectable loss of activity.

Full accounts of the preparation and chemical and biological properties of trichothecin will be given elsewhere.

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Role of the Y-Chromosome in the Determination of Cell-size in *D. melanogaster*

I have already pointed out¹ that in wild stocks of *D. melanogaster*, Gaiano (Parma), Luino (Varese), Civate (Como), the unicellular hairs of the upper wing surface, counted in a standard surface of 298 μ² for each wing, show frequency differences. Furthermore, there are sex differences of different kinds in the three stocks, being highest in Gaiano, intermediate in Civate and lowest in Luino. This shows the possibility of an action of the Y-chromosome, already proved by Mather² for the chætae also in *D. melanogaster*.

For this purpose I considered the Oregon R stock, which showed a very strong sex difference in addition to another cell character, namely, the eye corneolæ. The surface was measured directly³ in ten ommatidia of one eye (either the right or the left one, both being significantly similar) in each individual, and the mean was calculated. (The complex of the corneolæ of one eye was prepared after treatment in potassium hydroxide of the entire eye, and mounted in glycerine; the wings also were mounted in glycerine.) The following figures are the mean for thirty flies; the standard errors are indicated.