

THE EFFECT OF SODIUM ORTHO-PHENYL-PHENATE IN THE CULTURE MEDIUM ON THE *IN VITRO* DEVELOPMENT OF FUNGI CAUSING FRUIT ROTS

By

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S. O. P. P. dissolved in the culture medium had a clear inhibitory effect on the growth *in vitro* of a series of fungi causing fruit rots. The inhibitory effect increased with the concentration. The fungi most susceptible to the material were *Penicillium expansum*, *Penicillium digitatum*, *Botrytis cinerea* and *Diplodia natalensis*; that found most resistant was *Oospora citri-aurantii*. The differences in the reaction of the various fungi to the same concentration of S. O. P. P. in the culture media were found to be useful in separating between and isolating many species when occurring together in a mixed culture and to distinguish between different species of the same genus.

The effect of ortho-phenyl-phenol and sodium ortho-phenyl-phenate vapours on the *in vitro* development of fungi that cause fruit rots has been studied as part of a series of experiments dealing with long-term storage of citrus fruits (3, 4).

The present study deals with the effect of sodium ortho-phenyl-phenate (S.O.P.P.), dissolved in culture medium, on the development *in vitro* of a number of fungi that cause decay of various fruits and vegetables.

Inocula of the fungi under examination were plated in petri dishes on potato dextrose agar (P.D.A.) in which certain quantities of S.O.P.P., which is highly water soluble (6), were incorporated. Each petri dish contained 10 cc P.D.A. containing from 0.5 to 4 mg S.O.P.P. (0.005 — 0.04%). The inocula of uniform size were taken from the periphery of 10-day-old colonies growing on P.D.A., including both mycelia and spores.

The extent to which the inoculum developed under the effect of varying quantities of S.O.P.P. was determined by measuring the diameters of the colonies every three days for 12 days at 25°C. The presence of S.O.P.P. in the culture medium decreased the growth rate of all the fungi examined. The inhibitory effect of the material tended to increase with every increase in its concentration.

Total inhibition dose, i.e., the concentration of S.O.P.P. causing absolute prevention of development of the inoculum, differed for the different species and varied from 0.005% for *Botrytis cinerea*, *Penicillium digitatum*, *Penicillium expansum* and *Diplodia natalensis*, to 0.03% for *Oospora citri-aurantii* and *Fusarium solani* (Table 1).

The inhibitory effect of S.O.P.P. was very marked also at concentrations below the total inhibition point both in retarding the rate of growth and in limiting the size of the colony.

The retention of viability in inocula that had ceased to develop under the effects of the total inhibition doses shows that these concentrations are generally lower than the lethal doses. The lethal doses were determined by transferring inoculum that had been inhibited by a certain concentration of S.O.P.P. to ordinary P.D.A. and examining its viability. The lethal doses of S.O.P.P. in the culture medium varied between 0.01 and 0.04% for the different fungal species (Table 1).

The fungi most susceptible to S.O.P.P. of those studied, were *Botrytis cinerea* and *Penicillium expansum*. Of the causal agents of citrus rots the species found to be susceptible to this material were: *Penicillium digitatum*, *Diplodia natalensis*, *Trichoderma viride* and *Phytophthora citrophthora*. The fungus most resistant to S.O.P.P. was *Oospora citri-aurantii*. The high degree of resistance of this fungus to chemicals has already been shown with regard to Captan (1) and growth substances such as NAA, 2,4,-D and 2,4,5,-T (2).

The reaction of different fungi to the same concentration of S.O.P.P. was found useful in separating between and isolating various fungi in a mixed culture. It was thus possible to separate between a number of fungi that were cultured together with *Penicillium digitatum*, a fungus which commonly causes contamination of cultures in the laboratory during the citrus harvest season. Under the effects of 0.005–0.01% of S.O.P.P. in the culture medium, growth of the *Penicillium* was entirely inhibited, whereas other fungi with a

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TABLE 1

THE LETHAL DOSE OF S.O.P.P. IN COMPARISON WITH THE "TOTAL INHIBITION DOSE" WITH REGARD TO FUNGI IN CULTURE

Fungus	Total inhibition dose (%)	Lethal dose (%)
<i>Diplodia natalensis</i> P. E. ¹	0.0005*	0.04
<i>Penicillium digitatum</i> (Pers.) Sacc. ¹	0.005*	0.03
<i>Penicillium italicum</i> Wehm. ¹	0.02	0.04
<i>Penicillium expansum</i> Link ⁴	0.005	0.04
<i>Trichoderma viride</i> Pers. ex Fries ¹	0.01	0.02
<i>Phytophthora citrophthora</i> (Sm. et Sm.) Leon ¹	0.01	0.02
<i>Alternaria citri</i> Ellis & Pierce ¹	0.02	0.04
<i>Alternaria tenuis</i> Nees ²	0.01	0.04
<i>Colletotrichum gloeosporioides</i> Penz. ¹	0.02	0.02
<i>Oospora citri-aurantii</i> (Ferr.) Sacc. & Syd. ¹	0.03	0.04
<i>Oospora lactis</i> Fres. ²	0.02	0.03
<i>Botrytis allii</i> Munn ⁵	0.01	0.02
<i>Botrytis cinerea</i> Pers. ³	0.005	0.01
<i>Stemphylium verrucosum</i> Zimm. ³	0.02	0.03
<i>Stemphylium botryosum</i> Wall. ³	0.01	0.02
<i>Fusarium solani</i> (Mart.) Appel & Wollen. ²	0.03	0.03
<i>Fusarium moniliforme</i> Sheld. ⁶	0.01	0.03

* Initial growth after 10 days.

1 Isolated from citrus fruit.

2 Isolated from tomato.

3 Isolated from grape.

4 Isolated from apple.

5 Isolated from onion.

6 Isolated from banana.

greater resistance to the fungicide developed and could be isolated.

The selective effect of fungicides in culture media with regard to various fungi has been described by Vaartaja (5). In this study, the selective effect of S.O.P.P. enabled differentiation between species of the same genus. A differentiation between *Penicillium italicum* and *P. expansum*, and between *Botrytis allii* and *B. cinerea* could be achieved at a concentration of 0.005% S.O.P.P. *Alternaria citri* and *A. tenuis*, *Stemphylium verrucosum* and *S. botryosum*, and *Fusarium solani* and *F. moniliforme* could be differentiated at a concentration of 0.01% S.O.P.P. It was likewise possible to distinguish between *Oospora citri-aurantii* and *O. lactis* at a concentration of 0.02% S.O.P.P.

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