Experimental Control of Late Blight of Tomatoes with Capsidiol, the Phytoalexin from Peppers

E. W. B. Ward, C. H. Unwin and A. Stoessl

Agriculture Canada, Research Institute, University Sub Post Office, London, Ontario N6A 3K0. Accepted for publication 2 August 1974.

ABSTRACT

Capsidiol, the phytoalexin from peppers, applied in solution in water as a spray to tomato plants, effectively controlled late blight at concentrations down to 5×10^{-4} M, under growth-room conditions. The compound effectively

protected plants for at least 8 days, the longest period tested. These experiments demonstrate the potential of phytoalexins as sources of new fungicidal compounds.

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The possibility that phytoalexins could eventually play a role in the development of disease control measures is suggested by several lines of research. One possible avenue would be the controlled induction of phytoalexins in plants. Thus, for example, monilicolin A, a polypeptide from Monilinia fructicola, can be used to stimulate phaseollin formation in beans (2). Fungicides (5), ultraviolet irradiation (1), and tobacco necrosis virus (4), stimulate hydroxyphaseollin production in sovbeans. Cross-protection with the possible involvement of phytoalexins has been demonstrated in beans between races of Colletotrichum lindemuthianum (3, 6). Another avenue, which has received less attention, is the application of phytoalexins as fungicidal chemicals for plant disease control. Although generally not highly active, phytoalexins are antifungal by definition, exhibit widely differing chemical structures, presumably occur in numerous plants (in view of the numbers already

demonstrated in the relatively few host species investigated), at least several are readily biodegradable, and all offer the possibility of being made more active by chemical modification in the laboratory. A present drawback to the application of these compounds is that they are commonly available in limiting amounts. Nevertheless, for some phytoalexins chemical syntheses with commercial potential could be developed [as was demonstrated recently for orchinol (7)] or they could be obtained as by-products of food- or other plant-based industries as suggested by our observations of capsidiol production in diseased and rotted pepper fruit in the field (9, 10).

The phytoalexin capsidiol can be obtained readily and in appreciable quantity from pepper fruit (8). Studies in vitro (E. W. B. Ward, C. H. Unwin, and A. Stoessl, unpublished) have demonstrated that Phytophthora infestans is particularly sensitive to this compound. It was



Fig. 1. Control of late blight of tomatoes with capsidiol. Plants in the lefthand half of the box sprayed with 2×10^{-3} M capsidiol, approximately 1.5 hours prior to inoculation with zoospores of *Phytophthora infestans*. Photograph taken 9 days after inoculation.

TABLE I. Control of late blight of tomato with capsidiol under growth-room conditions

Capsidiol concentration ^a	Lesions per plant ^b
$2.5 \times 10^{-3} \text{ M}$	1.0 ± 0.7
$1.0 \times 10^{-3} \text{ M}$	1.6 ± 1.6
$5.0 \times 10^{-4} \text{ M}$	16.6 ± 5.1
Nil (Control)	135.0 ± 12.5

"Capsidiol sprayed onto upper surfaces of leaves of 7-week-old tomato plants approximately 1.5 hours before inoculation with zoospores of *Phytophthora infestans*.

Means and standard errors for numbers of lesions on the three lowest fully expanded leaves of each of five plants, counted 3 days after inoculation.

TABLE 2. Persistence of capsidiol on tomato leaves in the control of late blight of tomato under growth-room conditions

Capsidiol ^a (days prior to inoculation)	Lesions per plant ^b
0	1.8 ± 3.8
Ī	2.2 ± 2.9
2	25.2 ± 22.5
4	49.8 ± 31.3
8	38.6 ± 25.0
Control	370.0 ± 32.3

 $^{\circ}$ Capsidiol at 2.5 × 10⁻³ M sprayed onto upper surfaces of leaves of 7-week-old tomato plants.

"Means and standard errors for numbers of lesions on the three lowest fully expanded leaves of each of five plants per treatment, inoculated by spraying with a zoospore suspension of *Phytophthora infestans*, counted 3 days after inoculation.

considered, therefore, that the application of capsidiol for the control of late blight of tomatoes would provide a useful demonstration of the potential of phytoalexins for this purpose.

MATERIALS AND METHODS.—P. infestans was from stock cultures, initially isolated from diseased tomato fruit collected locally. It was grown on V8-juice agar at 18 C for 12-14 days in the dark, sporangia were removed in sterile distilled water chilled to 12 C, and allowed to germinate at that temperature for 1-3 hours in the dark in a shallow layer (< 1 cm) of distilled water. Zoospore suspensions were adjusted to a concentration of approximately 105/ml. Capsidiol (MW 236) was obtained as described previously (8). Greenhouse-grown tomato plants (cultivar, John Baer) approximately seven weeks old with three to four fully expanded leaves, were sprayed once from above with a solution of capsidiol delivered from a chromatography sprayer to give a visually uniform coverage. The solutions were prepared as described previously (8), by dissolving capsidiol in ethanol and injecting this into at least 20 volumes of water. Five plants in separate pots were used per treatment. After the leaf surfaces had dried (ca. 1.5 hours) the plants were placed randomly in a dew chamber (18 C, 12 hours light, 12 hours dark), moistened by spraying with a mist of distilled water, and inoculated by spraying with a zoospore

suspension containing one drop Tween 20 per 100 ml. Plants were incubated for approximately 72 hours, by which time individual lesions were easily distinguishable and coalescence had not started. The number of lesions on the first three fully expanded leaves on each plant were counted.

RESULTS AND DISCUSSION.—Capsidiol at 5×10^{-4} M gave a very high level of control (Table 1), while at concentrations of 1 and 2.5×10^{-3} M control was virtually complete. The effectiveness of the treatment is illustrated by Fig. 1, where half of a box of tomato plants was sprayed with 2.0×10^{-3} M capsidiol prior to inoculation. Under the conditions used, capsidiol had appreciable persistence (Table 2) and presumably it would be possible to increase this by appropriate formulation.

The results of the experiments reported here indicate clearly that capsidiol is a potentially effective fungicide, and suggest that other phytoalexins could be tested profitably in this way. If phytoalexin production is a general mechanism of resistance, protection of one host species by a phytoalexin induced in another adds an interesting dimension. It could provide a combination of resistance factors that might be obtained theoretically by interspecific or wider hybridization.

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