

EPIZOOTIC POTENTIAL FOR APHIDS OF DIFFERENT ISOLATES  
OF THE FUNGUS, *VERTICILLIUM LECANII*

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Large (6.7-8.4  $\mu\text{m}$  in length) and small (3.8-6.7  $\mu\text{m}$  in length) spored strains of *Verticillium lecanii* (Zimm) Viégas killed similar numbers of adults of the aphids treated with aqueous spore suspensions. However, when these strains were compared by determining the proportions of progeny aphids acquiring infection from treated adults, i.e. a measure of epizootic potential, large differences emerged ; only large-spored strains exhibited as strong an epizootic potential as the standard strain, 1-72, with mean progeny mortality ratios (test/standard) close to 1.0. Further experiments indicated that the speed of germination, fastest for large-spored strains, accounted for the greater epizootic potential of such strains. In addition, the mode of sporulation of large-spored strains on aphid cadavers probably also contributed to the more efficient spread of such strains. A general conclusion was reached that measurement of the effectiveness of a pathogen against a host insect must be based on many factors, in addition to virulence against a single developmental stage.

Two strains of the entomopathogenic fungus, *Verticillium lecanii* (Zimm) Viégas, have been developed as myco-insecticides to control aphids and whitefly (Hall, 1982a ; Hall & Burges, 1979). The strain of *V. lecanii* marketed as an aphicide ("Vertalec") is large-spored (mean spore length - 7.7  $\mu\text{m}$ ) while that used for commercial whitefly control ("Mycotal") is small-spored (mean spore-length - 4.4  $\mu\text{m}$ ). "Vertalec" is pathogenic to whitefly but less so than "Mycotal" and other small-spored strains (Hall, 1982b ; unpubl. data) and the converse is true for "Mycotal" (Hall, 1982b ; see below). The author has collected strains of *V. lecanii* from a wide variety of hosts and substrates and the variation in spore sizes is striking. In a previous study (Hall, 1977) in which aphids were treated with a range of spore concentrations to determine the LC50 (concentration at which 50 % of aphids are killed), virtually all strains, large- or small-spored, proved to be of similar pathogenicity. Subsequently, however, small-spored strains were discovered to spread poorly amongst rapidly-reproducing aphid populations (Hall, unpubl.). Thus, the effectiveness of a strain appears to depend on other factors as well as pathogenicity in adult aphids. Nevertheless, small-spored strains have the advantage of being much more productive in terms of numbers of spores produced per gram of fermentation substrate than large-spored strains (Hall, unpubl.).

The present study was undertaken to ascertain the relationship between spore size and pathogenicity of *V. lecanii* to aphids and to develop a new laboratory bioassay system to

compare the epizootic potentials of strains of the fungus by determining the degree of spread of disease from treated adults to progeny. An additional aim was to find a small-spored strain highly pathogenic to aphids.

## MATERIALS AND METHODS

### SOURCE OF APHIDS

The system of rearing aphids is described in Hall (1976). *Macrosiphoniella sanborni* Gillette was selected as a bioassay host, as it is easily manipulated (Hall, 1976).

*M. sanborni* populations were reared on potted chrysanthemums (cv. "Deep Tuneful") and maintained in a 16 h light regime, 20-24 °C. Mature apterous aphids were removed individually from plants with a small paint brush and deposited randomly into Petri-dishes. Twenty five aphids were used for each treatment.

### FUNGAL INOCULUM

The strains used in this study all belonged to the species *V. lecanii* with the exception of one, 27-79, which was identified as *V. fusiformis* Gams and was isolated from the Damsion-Hop aphid, *Phorodon humuli* Schrank.

Each strain was grown as a confluent colony on Sabouraud Dextrose Agar (SDA) for 7 days at 20 ± 1 °C in the dark. Conidia were harvested by dislodging them into sterile distilled water using a bent glass rod. Mycelial debris was removed by filtering through cheesecloth followed by centrifuging and washing the filtrate. Total spore counts were made using an improved Neubauer haemocytometer.

Viable spore counts were performed using the method of Hall (1976) ; only spores exhibiting a viability greater than 90 % were used in assays.

### BIOASSAY

Square plastic trays (10 × 10 cm) with 25 compartments were used. Two ml of molten agar (Agar No. 3 ; Oxoid Ltd.) were pipetted into each compartment. A 12-mm chrysanthemum leaf disc (cv. Deep Tuneful) was pressed onto the solidified agar in each compartment, abaxial surface uppermost.

For each strain, 2 spore concentrations (10<sup>6</sup> spores ml<sup>-1</sup> and 10<sup>7</sup> spores ml<sup>-1</sup>) were tested. The lower concentration was selected as it is the concentration at which spores of the "aphid" strain are applied in the commercial product "Vertalec". The upper concentration was included because small-spored strains may produce up to × 10 the yield of large-spored strains in culture. Aphids were treated as follows : 30 adult apterous aphids were placed in a net and immersed for 10 s in 25 ml of the appropriate spore suspension in a Petri dish. Triton X-100 was formerly used by the author as a wetting agent for bioassay but this compound has been found to partially inhibit sporulation on the cadaver (Hall, unpubl.). Therefore 0.05 % Tween 80 was substituted in the present study as it did not visibly impair sporulation on aphid cadavers. Control aphids were treated with 0.05 % Tween 80 only. Following treatment, the aphids were laid on filter paper to draw off surplus suspension and 1 aphid was placed in each compartment of the assay tray. The trays were sealed with thin adhesive plastic sheet, ('Clingfilm') in which small holes were made for ventilation, and incubated at 20 °C in the light (16-h day length). Adult aphid mortality was recorded daily from day 3 and, on the 6 th day, progeny mortality as well as the extent of sporulation were also recorded.

As the aphids were reared continuously in laboratory conditions, there could have been an increase in susceptibility to the fungus due to lack of selection pressure (Hall, 1976). To monitor this potential problem, a single-spore isolate of strain 1-72 was used as a standard which had originally been assayed against wild type *M. sanborni*.

Another assay was performed to investigate contagion to adult aphids with selected strains. Cadavers of aphids on which sporulating mycelium had been developing for 6 days were placed on leaf discs in the assay trays and one uninfected apterous adult was added to each cell. The assay trays were incubated and observed as before.

#### TEMPERATURE OPTIMA

To determine the optimum temperature for germination and growth, harvested conidia of selected strains were incubated at various temperatures on SDA-covered glass slides in Petri-dishes lined with damp filter paper. After 16 h, the slides were stained with cotton blue in lactophenol and the mean germ-tube lengths were measured.

The vegetative growth rates of large and small-spored strains were also determined on agar plates at 20 °. The strains were grown for 7 days on SDA. Conidia were harvested and 1 ml of  $10^7$  spores/ml<sup>-1</sup> were pipetted onto fresh agar plates containing 20 ml SDA. The inoculum was spread with a bent glass rod and the plates were incubated for 7 days at 20 °C. Using a sterile cork borer (11 mm diam.), plugs of agar were removed from these plates and placed in the centre of fresh SDA (20 ml) plates. Six plates of each strain were incubated and, at intervals, the colony diameters were measured.

#### RESULTS

Each strain was assayed twice in parallel with the standard strain, 1-72 with the exception of strains 93b-82 and 80-82 (4 assays) and 87-82 (6 assays). Table 1 shows mortality ratios (% mortality due to test strain/% mortality due to standard strain) based on mean percentage mortalities for adults, treated with spore suspensions, and those for the progeny resulting from contagion of disease from such treated adults. Thus, if increasing density of progeny positively influenced the rate of spread of disease in the confines of the assay cells, then such density dependent spread would have tended to obscure the full effect of the most pathogenic strains. Table 1 shows clearly that virtually all strains killed adults, the ratios between mortalities of the test strain and those of the standard strain 1-72, seldom being appreciably less than 0.5. The results agree with those in a previous study (Hall, 1977). However, when progeny mortalities are examined, differences between ratios are much greater. This emphasises important differences between the epizootic potentials of the various strains. In fact, only some of the larger-spored strains exhibited strong epizootic potential as shown by the ratios for progeny mortalities approaching 1 (table 1). Small-spored strains in contrast, even if originally isolated from aphids, displayed poor epizootic potential in comparison. Those strains displaying strong epizootic potential produced conidia larger than 6.7 µm. For convenience, these have been designated in this paper as "large-spored" and strains with shorter spores as "small-spored".

However, small-spored strains can yield high numbers of spores. For instance, 19-79, spore-length 4.4 µm, will produce  $5.6 \times 10^9$  blastospores/ml in liquid culture compared with  $8 \times 10^8$  spores/ml for 1-72 (Hall & Latgé, unpubl.) — a difference of  $\times 7$ . Therefore, a comparison based on productive biomass of spores may be better. Given this greater spore yield in culture of the smaller-spored strains, which was assumed to be a maximum of  $\times 10$  compared to 1-72, progeny mortalities for the upper concentrations ( $10^7$  spores/ml) of small-spored

TABLE 1

*Mortality resulting from treatment of adult, apterous aphids  
(Macrosiphoniella sanborni) with spores of different strains of Verticillium lecanii*

Strain	Spore concentration (ml <sup>-1</sup> )	Spore length (µm)	Original host	Mean adult mortality ratio (test/standard)	Mean progeny mortality (%)	Mean progeny mortality ratio (test/standard)	Mean LT50 (days) for adults
93b-82	10 <sup>7</sup>	8.4	A	0.58	3.9	0.06	4.9
	10 <sup>6</sup>			0.71	6.5	0.18	5.6
87-82	10 <sup>7</sup>	7.8	NI	0.25	4.7	0.06	>12.0
	10 <sup>6</sup>			0.21	3.3	0.05	>12.0
1-72 (Standard)	10 <sup>7</sup>	7.7	A	1.00	(41.3-100) <sup>(a)</sup>	1.00	3.1
	10 <sup>6</sup>			1.00	(10.6-62.0) <sup>(a)</sup>	1.00	3.6
86-82	10 <sup>7</sup>	7.6		1.00	61.1	0.71	3.6
	10 <sup>6</sup>			1.00	52.6	1.67	3.9
13-74	10 <sup>7</sup>	7.3	A	1.00	72.5	0.91	3.6
	10 <sup>6</sup>			1.02	30.5	0.83	4.2
20-79	10 <sup>7</sup>	7.2	S	0.67	15.7	0.19	—
	10 <sup>6</sup>			0.61	10.1	0.27	—
80-82	10 <sup>7</sup>	7.0	L	0.71	7.7	0.10	4.1
	10 <sup>6</sup>			0.67	2.3	0.04	5.7
93a-82	10 <sup>7</sup>	6.8	A	1.00	77.2	1.00	3.7
	10 <sup>6</sup>			1.03	26.3	0.71	4.0
3-72	10 <sup>7</sup>	6.7	A	1.00	86.6	1.0	3.2
	10 <sup>6</sup>			1.00	41.1	1.08	3.5
26-79	10 <sup>7</sup>	6.4	NI	0.71	0.02	0.0002	5.0
	10 <sup>6</sup>			0.59	0.5	0.02	6.8
9-73	10 <sup>7</sup>	6.2	S	1.00	1.9	0.02	—
	10 <sup>6</sup>			0.59	2.4	0.17	—
79-82	10 <sup>7</sup>	6.0	L	1.00	5.0	0.06	3.5
	10 <sup>6</sup>			1.00	0.02	0.0004	3.6
56-81	10 <sup>7</sup>	5.8	A	1.00	22.0	0.25	3.7
	10 <sup>6</sup>			0.83	3.3	0.21	4.7
2-72	10 <sup>7</sup>	5.8	A	0.91	9.4	0.15	4.8
	10 <sup>6</sup>			0.53	1.3	0.04	5.6
57-81	10 <sup>7</sup>	5.7	S	1.00	18.8	0.31	3.7
	10 <sup>6</sup>			0.91	5.2	0.09	4.0
40-81	10 <sup>7</sup>	5.6	S	0.91	4.6	0.06	4.1
	10 <sup>6</sup>			0.77	3.9	0.12	4.7
102-82	10 <sup>7</sup>	5.6		0.83	1.5	0.02	4.7
	10 <sup>6</sup>			0.71	0.6	0.02	4.7
41-81	10 <sup>7</sup>	5.5	EM	0.91	6.4	0.07	4.5
	10 <sup>6</sup>			0.83	3.2	0.3	4.5
8-73	10 <sup>7</sup>	5.4	S	0.83	15.4	0.24	5.2
	10 <sup>6</sup>			0.53	0.8	0.05	8.0
4-72	10 <sup>7</sup>	5.3	A	1.00	12.4	0.14	4.6
	10 <sup>6</sup>			0.36	0.15	0.005	6.8

*to be continued*

TABLE 1 (cont.)

*Mortality resulting from treatment of adult, apterous aphids  
(Macrosiphoniella sanborni) with spores of different strains of Verticillium lecanii*

Strain	Spore concentration (ml <sup>-1</sup> )	Spore length (μm)	Original host	Mean adult mortality ratio (test/standard)	Mean progeny mortality (%)	Mean progeny mortality ratio (test/standard)	Mean LT50 (days) for adults
21-79	10 <sup>7</sup>	5.2	R	0.77	2.1	0.024	4.8
	10 <sup>6</sup>			0.48	0.8	0.06	5.4
7-73	10 <sup>7</sup>	5.1	R	0.10	0.9	0.02	>12.0
	10 <sup>6</sup>			0.16	0.5	0.03	>12.0
32-79	10 <sup>7</sup>	4.9	A	1.00	17.4	0.25	3.2
	10 <sup>6</sup>			0.83	3.5	0.28	3.9
28-79	10 <sup>7</sup>	4.8	LH	0.77	2.3	0.04	4.6
	10 <sup>6</sup>			0.42	0.0	0.00	6.1
33-79	10 <sup>7</sup>	4.8	C	0.83	14.2	0.17	4.1
	10 <sup>6</sup>			0.83	1.5	0.04	5.0
5-72	10 <sup>7</sup>	4.8	A	0.83	1.9	0.04	6.3
	10 <sup>6</sup>			0.59	1.1	0.10	5.3
14-74	10 <sup>7</sup>	4.8	R	0.91	5.7	0.09	5.0
	10 <sup>6</sup>			0.77	0.95	0.06	5.3
17-76	10 <sup>7</sup>	4.8	EM	0.91	0.85	0.01	4.5
	10 <sup>6</sup>			0.71	0.0	0.00	5.4
31-79	10 <sup>7</sup>	4.8	EM	0.42	0.5	0.03	5.8
	10 <sup>6</sup>			0.42	1.1	0.09	5.2
59-81	10 <sup>7</sup>	4.8	L	0.48	0.8	0.01	>12.0
	10 <sup>6</sup>			0.19	0.2	0.01	>12.0
53-81	10 <sup>7</sup>	4.7	Th	1.00	14.5	0.16	3.7
	10 <sup>6</sup>			0.83	1.5	0.03	4.3
29-79	10 <sup>7</sup>	4.6	Th	1.00	6.6	0.08	3.5
	10 <sup>6</sup>			0.59	0.0	0.00	6.0
55-81	10 <sup>7</sup>	4.5	S	0.83	13.0	0.15	4.9
	10 <sup>6</sup>			0.67	3.3	0.27	4.3
103-82	10 <sup>7</sup>	4.4	Th	0.91	4.2	0.06	4.4
	10 <sup>6</sup>			0.67	0.45	0.01	5.4
19-79	10 <sup>7</sup>	4.4	S	0.91	11.6	0.19	4.1
	10 <sup>6</sup>			0.43	0.0	0.00	5.9
6-72	10 <sup>7</sup>	4.2	A	0.91	0.6	0.01	5.0
	10 <sup>6</sup>			0.67	0.6	0.02	6.2
11-73	10 <sup>7</sup>	4.1	NI	1.00	2.3	0.03	4.2
	10 <sup>6</sup>			0.53	1.0	0.06	6.5
12-74	10 <sup>7</sup>	4.1	S	0.55	1.8	0.03	4.9
	10 <sup>6</sup>			0.36	0.0	0.00	6.3
22-79	10 <sup>7</sup>	4.1	R	0.91	9.2	0.10	4.8
	10 <sup>6</sup>			0.55	0.8	0.04	6.2
30-79	10 <sup>7</sup>	3.9	EM	0.91	3.4	0.05	4.1
	10 <sup>6</sup>			0.83	2.4	0.19	5.0

to be continued

TABLE 1 (cont.)

*Mortality resulting from treatment of adult, apterous aphids*  
(*Macrosiphoniella sanborni*) with spores of different strains of *Verticillium lecanii*

Strain	Spore concentration (ml <sup>-1</sup> )	Spore length (μm)	Original host	Mean adult mortality ratio (test/standard)	Mean progeny mortality (%)	Mean progeny mortality ratio (test/standard)	Mean LT50 (days) for adults
24-79	10 <sup>7</sup>	3.9	A	0.91	5.1	0.06	3.7
	10 <sup>6</sup>			0.42	0.0	0.00	> 12.0
18-78	10 <sup>7</sup>	3.8	S	1.00	12.8	0.15	4.2
	10 <sup>6</sup>			0.91	2.5	0.08	4.8
15-74	10 <sup>7</sup>	3.8	C	0.83	1.0	0.01	5.7
	10 <sup>6</sup>			0.45	1.4	0.08	8.0
27-79(b)	10 <sup>7</sup>	3.3	A	0.45	0.0	0.0	> 12.0
	10 <sup>6</sup>			0.23	0.0	0.0	> 12.0

(a) Range of progeny mortalities obtained for standard strain in an assay ; (b) *V. fusiformis*.

A = Aphid ; NI = Non-insect/plant substrate ; L = Lepidoptera ; S = Scale ; LH = Leafhopper ; C = Coleoptera ; Th = Thrips ; R = Rust fungus ; EM = Eriophyid mite

TABLE 2

*Mortality ratios for aphid progeny killed by V. lecanii test strains compared with the standard strain, 1-72, on a productive biomass basis*

Strain	Spore length (μm)	% mortality of progeny at 10 <sup>7</sup> sp ml <sup>-1</sup> (test strain)
		% mortality of progeny at 10 <sup>6</sup> sp ml <sup>-1</sup> (std. strain)
15-74	3.8	0.05
18-78	3.8	0.42
30-79	3.9	0.26
24-79	3.9	0.27
11-73	4.1	0.13
12-74	4.1	0.07
22-79	4.1	0.42
6-72	4.2	0.02
103-82	4.4	0.01
19-79	4.4	0.36
55-81	4.5	0.83
29-79	4.6	0.29
53-81	4.7	0.28
5-72	4.8	0.18
14-74	4.8	0.37
17-76	4.8	0.03
28-79	4.8	0.13
31-79	4.8	0.04
33-79	4.8	0.38
59-81	4.8	0.02

strains have been compared with the lower concentration ( $10^6$  spores/ml) for 1-72 (table 2). No small spored strain had a greater epizootic potential than 1-72 when compared in this way. *V. fusiformis*, isolated from the Damson-Hop aphid, *Phorodon humuli*, was among the least pathogenic of the strains tested, both for adults and progeny of *M. sanborni*.

Reasons why in general only large-spored strains exhibited promising epizootic potential were sought. The temperature optima for rate of growth of germ tubes were measured for selected strains. The optima for large-spored strains were between 22 and 24 °C while those for small-spored strains were between 25 and 27 °C (table 3). The assays were carried out at 20 °C, thus favouring large-spored strains. Consequently, supplementary bioassays were performed at 25 °C with strain 1-72 (optimum 23 °C) and 2 small-spored strains, both of

TABLE 3

*Optimum temperatures for growth of germ tubes and mean germ tube lengths of large- and small-spored strains of V. lecanii*

Strain	Mean length (μm)	Temperature optimum (°C)	n	Mean germ tube lengths (μm) ± S.E. after 15 h on agar at :	
				20 °C	25 °C
Large-spored strains					
3-72	6.7	22	3	39.9 ± 3.9	3 56.2 ± 5.6
93a-82	6.8	23	4	48.6 ± 4.3	4 81.2 ± 2.8
20-79	7.2	23	3	50.2 ± 4.0	3 93.0 ± 6.8
13-74	7.3	24	3	69.5 ± 8.4	3 98.1 ± 5.7
1-72	7.7	23	5	49.5 ± 3.4	5 98.8 ± 7.5
93b-82	8.4	23	3	52.7 ± 9.8	3 100.8 ± 8.3
Small-spored strains					
15-74	3.8	25	3	19.4 ± 1.8	3 44.2 ± 7.6
16-75	3.8	27	3	17.3 ± 1.2	3 43.5 ± 4.0
6-72	4.2	27	3	23.0 ± 0.8	3 48.2 ± 3.1
19-79	4.4	25	4	14.6 ± 2.1	4 43.8 ± 4.2
103-82	4.4	25	3	21.0 ± 3.3	3 35.6 ± 1.9
14-74	4.8	25	3	28.8 ± 1.6	3 42.6 ± 3.4
5-72	4.8	26	3	20.5 ± 1.4	3 37.7 ± 2.7

n = number of batches of germ-tube lengths (30 germ tubes measured for each batch)

which had an optimum of 25 °C. Table 4 shows that percentage progeny mortality at 20 °C was not consistently different from that at 25 °C. Other explanations were sought. The rate of fungal grow was studied. Table 5 shows that the colony diametrical growth rates on agar in Petri-dishes did not differ between large- and small-spored strains. However, when germination rates were examined, differences between small- and large-spored strains emerged : of a total of 6 large-spored strongly-pathogenic strains tested, at 20 °C, 5 produced germ tubes more than twice as long as those of all but 1 of the 7 short-spored strains tested, and at 25 °C, the results were similar (table 3). However, 2 large-spored strains, 93b-82 and 20-79, although fast-germinating (table 3) caused only low progeny mortality but these strains also killed fewer adult aphids, implying an inherently lower pathogenicity. Germination rates for the large-spored strains 87-82 and 80-82 which did not spread well to progeny, were not measured, but they too were less efficient in killing adult aphids than the strain, 1-72.

Faster spore germination would provide an opportunity for more rapid penetration of the insect cuticle, leading to irreversible infection. Considering that aphid progeny ecdyse 4 times before reaching adulthood, the ability to germinate fast would enable a fungus to succeed more often in infecting progeny before being sloughed off by a moult. It has been shown before that moulting can constitute a barrier to infection by fungi (Fargues & Vey, 1974). However, some small-spored strains with poor epizootic potential produced short LT50s (table 1). This can be explained by the results of a separate study (Hall, unpubl.) in which the rates of biomass production were compared in liquid media of the large-spored strain 1-72 and short-spored strains; these experiments showed that short-spored strains grow slightly faster than 1-72 and so, despite later penetration of cuticle, faster growth in adults may well explain the short LT50s. In parenthesis, measuring colonial growth rates on agar is evidently not a very precise method of comparing biomass production.

TABLE 4

*Adult and progeny aphid mortalities in assays conducted at 20 and 25 °C*

Strain no.	Expt. no.	Spores ml <sup>-1</sup>	20 °C		25 °C	
			Adult mortality (%)	Progeny mortality (%)	Adult mortality (%)	Progeny mortality (%)
1-72 (large-spored)	1	10 <sup>7</sup>	100	83.0	100	35.9
		10 <sup>6</sup>	100	21.2	100	6.9
	2	10 <sup>7</sup>	100	51.0	100	90.9
		10 <sup>6</sup>	100	56.0	100	66.6
19-79 (small-spored)	1	10 <sup>7</sup>	96	11.0	86	9.8
		10 <sup>6</sup>	84	8.7	12	0
	2	10 <sup>7</sup>	100	3.4	100	11.0
		10 <sup>6</sup>	100	2.1	100	5.7
103-82 (small-spored)	1	10 <sup>7</sup>	92	3.2	92	5.2
		10 <sup>6</sup>	80	0	60	0.9
	2	10 <sup>7</sup>	100	0	92	4.6
		10 <sup>6</sup>	100	0	84	0

TABLE 5

*Colony diametrical growth rates<sup>(a)</sup> of large- and small-spored strains of V. lecanii*

Large-spored strains			Small-spored strains			
Strain number	1-72	13-74	93b-82	15-74	19-79	103-82
Growth rate at 20 °C (mm day <sup>-1</sup> ) :						
	2.48 ± 0.07	1.43 ± 0.04	2.29 ± 0.06	2.25 ± 0.06	2.42 ± 0.08	2.16 ± 0.0

(a) Means ± S.E. of 6 determinations



Although slowly-germinating strains may succeed in infecting moulting progeny through contagion less often than fast-germinating strains, the differences between such strains should be less when spread of disease to the non-moulting adult is compared. Table 6 shows that this is so.

The mode of sporulation on aphid cadavers was noted : the large-spored strains produced short, compact velvet-like mycelia bearing abundant spore heads by the 6th day, i.e. the end of the assay ; small-spored strains, however, produced more fluffy growth, and sporulation was often not prolific and often masked by sterile mycelium. These differences would influence epizootic potential.

TABLE 6

*Mortality (%) among untreated adult aphids, each incubated with a cadaver of an adult aphid killed by V. lecanii*

Expt. no.	Strain number		
	1-72 large	19-79 small	103-82 small
(i)	100	100	96
(ii)	100	96	92

## DISCUSSION

The first key finding of this study is that, when considering measurement of the effectiveness of an entomopathogen against a host insect with a high reproductive potential, estimation of adult mortality alone can be totally misleading. A pathogen can be successful against such insects only if, given favourable conditions, it possesses the ability to spread efficiently through rapidly-reproducing host populations. In this study, it was evident that the conclusions drawn from the abilities of *V. lecanii* strains to kill spore-treated adults were very different to those gleaned from the abilities of the same strains to spread to untreated progeny of treated adult aphids. Therefore, bioassay design should take account of the differing susceptibilities of the adult insect and the developmental stages. In the case of aphids, progeny should be incorporated into the assay to improve assessment of epizootic potential.

All strains displaying strong epizootic potential were large-spored and none of these had slow germination rates – unlike most small-spored strains. Some Small-spored strains are equally pathogenic towards adult aphids but disease did not spread efficiently to progeny possibly because (i) of the slower germination rates which could impair the abilities of such strains to irreversibly infect progeny before moults, and (ii) the mode of sporulation of small-spored (fluffy mycelium masking sporulation) strains on cadavers would not encourage efficient spread. The poor epizootic potential of some large-spored strains which germinate rapidly could be explained by a low inherent pathogenicity against *M. sanborni* as indicated by low adult mortality.

Although the 'standard strain' of *V. lecanii*, 1-72, employed throughout this study, was isolated from *M. sanborni*, previous laboratory experiments showed that other aphid species such as *Myzus persicae* Sulzer and *Brachycaudus helichrysi* (Kaltenbach) were as susceptible to 1-72 as *M. sanborni* (Hall & Burges, 1979). In the glasshouse environment, this strain was outstandingly effective against *M. persicae* (Hall & Burges, 1979). It is reasonably likely that a strain strongly pathogenic for say *M. persicae*, from which that strain had been isolated, would be detected in assays using *M. sanborni*.

It is not likely that the commercially-produced strain, 1-72, will be replaced by a higher-yielding small-spored strain as no such strain was found to possess the necessary combination of high pathogenicity to adults and comparable epizootic potential. It would seem that the large-spored characteristic and possible consequent fast germination in *V. lecanii* is an essential prerequisite for strong epizootic potential in aphid populations.

One could speculate that a large spore is likely to germinate faster because it contains proportionally more nutrients than a small spore. This may be so in *V. lecanii* but whether or not it is a general rule with fungi is not known and certainly does not apply to 'major' (large-spored) and 'minor' (small-spored) strains of *Metarhizium anisopliae* (Metsch.) Sorokin (Hall, unpubl.). Aphids are not the only hosts of *V. lecanii*. Scales and soft scales such as the whitefly, *Trialeurodes vaporariorum*, are perhaps better-known hosts. The strain incorporated in the commercial preparation 'Mycotal' for whitefly control is a small-spored strain (19-79). The reason why small-spored strains exhibit greater epizootic potential for this host are not clear but, in a previous study (Hall, 1982b), the large-spored strain, 1-72 displayed a  $\times 10$  lower inherent pathogenicity against whitefly than 19-79. This difference in pathogenicity, coupled with a lower sporulation potential of large-spored strains on scales (Hall, unpubl.) probably account for the contrasting differences in epizootic potential.

#### ACKNOWLEDGMENTS

The author is grateful to Tate & Lyle Ltd. and Koppert BV for part-funding of this study and to Alison Spalding, who carried out much of the practical work.

#### RÉSUMÉ

##### Potentialités épizootiques chez les pucerons de différents isolats du champignon *Verticillium lecanii*

Les souches de *Verticillium lecanii* (Zimm) Viégas à grandes spores (6,7 à 8,4  $\mu\text{m}$  de longueur) et à petites spores (3,9 à 6,7  $\mu\text{m}$ ) tuent un nombre analogue de pucerons adultes traités par des suspensions de spores dans l'eau. Cependant, lorsque ces souches sont comparées d'après l'importance de l'infection dans la descendance des adultes traités, c'est-à-dire d'après le potentiel épizootique, il apparaît de grandes différences ; seules les souches à grandes spores ont un potentiel épizootique égal à celui de la souche étalon, 1.72. Des essais complémentaires montrent que la vitesse de germination, plus rapide chez les souches à grandes spores, intervient dans leur meilleur potentiel épizootique. En plus, le mode de sporulation des souches à grandes spores sur les pucerons morts contribue aussi probablement à une plus efficace dispersion de ces souches. On en tire la conclusion générale que la mesure de l'efficacité d'un agent pathogène à l'égard d'un insecte hôte doit être fondée sur beaucoup de facteurs, en plus de la virulence, agissant sur un seul stade de développement.

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