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**EFFECT OF *BEAUVERIA BASSIANA* ON UNDERGROUND STAGES  
OF THE COLORADO POTATO BEETLE,  
*LEPTINOTARSA DECEMLINEATA*  
(COLEOPTERA: CHRYSOMELIDAE)**

George E. Cantwell<sup>1</sup>, William W. Cantelo<sup>1</sup>, and Robert F. W. Schroder<sup>2</sup>

ABSTRACT

Tests were conducted to determine the effect of the fungus *Beauveria bassiana* (*B.b.*) on underground stages of the Colorado potato beetle (CPB), *Leptinotarsa decemlineata*. Two levels of *B.b.*, 7.5 g/m<sup>2</sup> and 75 g/m<sup>2</sup>, were suspended in water and sprinkled over the surface of the ground in cages to which CPB were added, either as overwintering adults or as 4th instar larvae of the 1st generation. Efficacy of treatments was determined by counting emerged adults. Neither level of *B.b.* increased mortality in overwintering adults. The highest level of *B.b.* caused a significant decline in emergence of adults from the 1st generation 4th instars. The 7.5 g/m<sup>2</sup> treatment level did not significantly differ from control treatments.

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Entomopathogenic fungi have been studied for over a century. However, it is only within the past several decades that special efforts have been focused on fungi as possible agents for biological control of insects. One of the fungi, *Beauveria bassiana* (*B.b.*), shows some promise in insect control and is known to attack the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (CPB). As with most fungal diseases, *B.b.* development progresses from attachment of the infective unit to the insect's cuticle, to germination and penetration of the body wall: The fungus then multiplies within the host and produces toxic metabolites which eventually kill the insect.

A positive correlation between the number of infective spores and mortality by mycosis has been established (Ferron 1978) and the Soviet authors Goral and Lappa (1973) recommended their industrial preparation of "Boverin" at ca.  $1-2 \times 10^{13}$  spores/ha for CPB control. The purpose of our study was to determine if a single soil application of an experimental preparation of *B.b.* would reduce populations of the generation of the CPB that overwintered or of the first generation adults.

MATERIALS AND METHODS

From August 1984 until the summer of 1985 four separate tests were conducted using the fungus *B. bassiana* against underground stages of the Colorado potato beetle. The *B.b.* used in these tests was an experimental preparation supplied by Abbott Laboratories of North Chicago, Illinois.

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The first test was run to determine the effect of *B.b.* on overwintering adults. On 7 August 1984, two tomato plants were enclosed in each of 21 2 by 2 by 2-m cages. Into each cage were placed 100 field-collected adult CPB. Treatments consisted of two levels of *B.b.*, 75 g/m<sup>2</sup> or 7.5 g/m<sup>2</sup>, or water as a control. The *B.b.* powder, identified as ID# 6112 8-23 84; AARC-012-129, contained  $1 \times 10^{10}$  spores/g and was suspended in water and applied over the ground surface in 11.5 l water/cage prior to seeding with beetles. There were six replicates of each treatment plus three cages in which neither beetles nor *B.b.* was added. The latter cages were used to determine if a residual beetle population existed in the soil. One cage in the control treatment was blown away during the winter and had to be removed from the test.

Weekly counts of emerging adults were made from 22 April through 5 June 1985. The data were analyzed using Duncan's Multiple Range test to determine differences among treatments.

The second test was conducted in the late spring of 1985 at Beltsville, Maryland, using 20 2 by 2-m screened cages. Again, two *B.b.* treatments (Lot #76-015BR) were used, 75 g/m<sup>2</sup> or 7.5 g/m<sup>2</sup>, each applied by sprinkling in 11.5 l of water. There were nine replicates of water control, six replicates of the low dose, and five replicates of the high dose. In this test each cage contained three tomato plants and was seeded with 100 late 4th-instar CPB larvae. Weekly counts of emerging adults were begun on 25 June and continued until 11 July.

Test 3 was similar to test 2 except that it was conducted at Cheltenham, Maryland, and consisted of 21 cages approximately 1 m-square with 50 late 4th-instars placed into each cage. The *B.b.* was sprinkled over the surface in 3.8 l of water. Treatment levels were the same as in test 2 with seven replicates of each treatment. Samples of collected adults from tests 1-3 were held for three weeks for evidence of fungal infections.

A fourth test was started in early March 1985 in a field that harbored very large beetle populations in previous years. This field had been planted yearly in tomatoes and potatoes and was used primarily as a source of beetles for other tests. Each year the beetles in this field totally stripped the foliage. The purpose of the test was to determine the effect of *B.b.* on a natural CPB overwintering population.

The *B.b.* used in this test was ID# Lot 150-124-2 905 6112. On 14 March 24 screened cages were placed over rototilled soil. These consisted of 18 cages 1 by 1 m and 6 cages 2 by 2 m. Two levels of *B.b.* were sprinkled onto the soil at the rates of 75 g/m<sup>2</sup> or 7.5 g/m<sup>2</sup> in 11.5 l of water in the large cages or 3.8 l of water in the smaller cages. There were eight replicates of each *B.b.* treatment and eight water controls. No CPB were placed into the cages. Weekly observations for emerging adults were begun in late April and continued through late June.

## RESULTS AND DISCUSSION

Data for tests 1-3 are presented in Table 1. In each of these tests, statistical analysis of the data revealed no significant differences among treatments ( $P < 0.05$ ). In test number one, in which 100 adults were placed into the cages, nearly 1/3 emerged from the soil the following spring. In several cages over 50% emerged; the highest number counted was 62, which was in a cage with the 75 g/m<sup>2</sup> level of *B.b.* The average number of beetles/cage recovered in the unseeded cages was seven. This test was conducted in a field that had a light CPB population which was not heavy enough to defoliate the tomato foliage. The recovery of beetles in these three cages indicates that some of the natural CPB population overwintered in this field.

In tests 2 and 3, 100 and 50 late 4th-instar larvae respectively were placed into cages and allowed to pupate in the soil and emerge as adults. As in test 1, the average number of adults emerging was about 1/3 of those seeded and, as mentioned above, there were no significant differences in emergence numbers among treatments.

When percent beetle emergence from tests 2 and 3 were combined there was a significant difference in adult emergence between the highest level of *B.b.* treatment and

Table 1. Effect of *Beauveria bassiana*<sup>a</sup> on survival of underground stages of the Colorado potato beetle, *Leptinotarsa decemlineata*.

Soil Treatment	Mean percent adult emergence from the soil <sup>b</sup>			
	overwintering adults Test 1	1st generation adults		
		Test 2	Test 3	Tests 2&3
<i>B. b.</i> 7.5 g/m <sup>2</sup>	29.8	38.8	62.6	50.8AB
<i>B. b.</i> 75 g/m <sup>2</sup>	35.3	28.0	43.6	36.5B
Control	33.8ns	49.2ns	62.0ns	54.3A

<sup>a</sup>*B. b.* supplied by Abbott Laboratories and contained  $1 \times 10^{10}$  spores/g.

<sup>b</sup>Means in the same column with the same letter are not significantly different ( $P < 0.05$ ), by Duncan's multiple range test.

the controls. However, emergence after the low treatment dose of  $7.5 \times 10^{10}$  spores/m<sup>2</sup>, was not significantly different from the control treatment. Those adults from tests 1–3 and held for observation exhibited no signs of *B. b.* infection after three weeks.

No data are presented for test 4, because there was no emergence into the cages from the previous year's natural population. Weekly observations for emerging adults were begun in late April and continued for two months. During this period only one adult CPB was found in a total caged area of approximately 42 m<sup>2</sup>. One-acre plantings of potatoes and tomatoes surrounding the test cages were subject to the usual annual heavy CPB pressure and, as in previous years, the plants were completely defoliated. This indicates that the 1984 natural beetle population did not diapause in the field that they had completely defoliated, but migrated out of that field, probably in search of food. If this migration is typical behavior, the use of *B. b.* or any other insecticide for control of overwintering CPB populations in fields that have been completely stripped of foliage would be futile because of the absence of a beetle population.

In spite of the extremely heavy dose of *B. b.* applied ( $75 \times 10^{10}$  spores/m<sup>2</sup> of soil) in an amount of water that drenched the soil, no significant reductions in CPB populations could be effected. However, when results of both tests 2 and 3 were combined, a reduction of 33% was obtained which was significant compared to that of the untreated control. Our results are not as encouraging as those of Watt and LeBrun (1984); however, our data cannot be directly compared because the test designs were not similar. These authors used  $2-3 \times 10^{10}$  conidia or fresh blastospores/m<sup>2</sup> applied in water to soil under cages. Their treatments were aimed at controlling first and second generation CPB. They reported a 74% reduction of the 1st generation and a 77% reduction of the second. We have no explanation for the obvious difference between the results of these tests.

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