

PATHOLOGICAL EFFECTS OF ENTOMOPHAGOUS NEMATODES AND THEIR ASSOCIATED BACTERIA ON AVOCADO INSECT PESTS

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Project Objectives: To assay efficacy of the entomophagous nematodes *Steinernema* and *Heterorhabditis* in killing avocado lepidopteran insect pests and other smaller species such as greenhouse thrips. While the toxicity of the nematodes to Lepidoptera previously has been documented, the susceptibility of very small arthropods such as thrips, aphids, whiteflies, and mites has been little studied.

This past year significant progress was made on this project and we have now accumulated sufficient data to warrant two publications that will acknowledge the Avocado Research Advisory Committee for its support of our research. These papers will be submitted to the journals *Annals of the Entomological Society of America* (or alternatively, a new journal entitled *Biocontrol*) and the *Journal of Invertebrate Pathology*, respectively.

We completed our ongoing experiments demonstrating efficacy of both *Steinernema* and *Heterorhabditis* in killing all stages of *Amorbia* larvae; susceptibility of newly eclosed pupae was also demonstrated, which has not previously been reported. Thus, pupal susceptibility to nematode infection represents a new finding to be included in our publication. Following tanning of the pupal cuticle, the nematodes appeared to be unable to penetrate these insects.

In addition, we tested the susceptibility of thrips and other small insects such as whiteflies to the nematodes, using the poinsettia whitefly (recently renamed silverleaf whitefly) as the test insect. Results of these bioassays are extremely promising and suggest that the nematodes and their associated pathogenic bacteria might be utilized for biocontrol of species that previously have been little studied with respect to their susceptibility to nematode invasion. These results will be written up for publication shortly and will be of great interest to commodities currently experiencing problems with these devastating insects. In essence, the whiteflies are killed within 24 hr of nematode application - they first turn red (due to the presence of the pathogenic bacteria) then shrivel to a fraction of their normal size, making them difficult to detect on the substrate. Results are summarized in Table 1 and Figures 1 and 2, showing effects applying different concentrations of *Heterorhabditis bacteriophora* (obtained from Dr. Harry Kaya,

UC-Davis) and *Steinernema carpocapsae* (obtained from Biosys, Inc. as the product Biosafe) to various stages of the whiteflies. Similar promising results were obtained with thrips, though one problem with that species was the presence of a honeydew-like residue on the fruit, which appeared to inhibit nematode migration on the surface and penetration into the insects. Because *Heterorhabditis* has a tooth capable of penetrating insect cuticle, we anticipate that that species would be more effective in killing insects whose mouthparts are lodged in plant tissues, such as aphids and thrips, thus preventing nematode entry via that route. We are now carrying out additional experiments with aphids.

Our results with injection of insects with the bacteria alone (cultured in the absence of nematodes) indicates that the bacteria are highly toxic. Thus, we are now planning to study the factors produced by the bacteria as potential insecticides. Once isolated, these genes can be engineered into plants for expression of insecticidal activity *in situ* in the plant, much like the *Bacillus thuringiensis* toxin gene has been effectively exploited for the same purpose in a variety of plants.

Table 1. Effect of nematodes on whitefly mortality as determined 48 hr following exposure to nematodes. Whiteflies were assayed in petri dishes containing dosages of 20,000, 10,000, or 1,000 infective stage juveniles per plate. Mortality was 0% in noninfected controls (n = 150 for each stage tested).

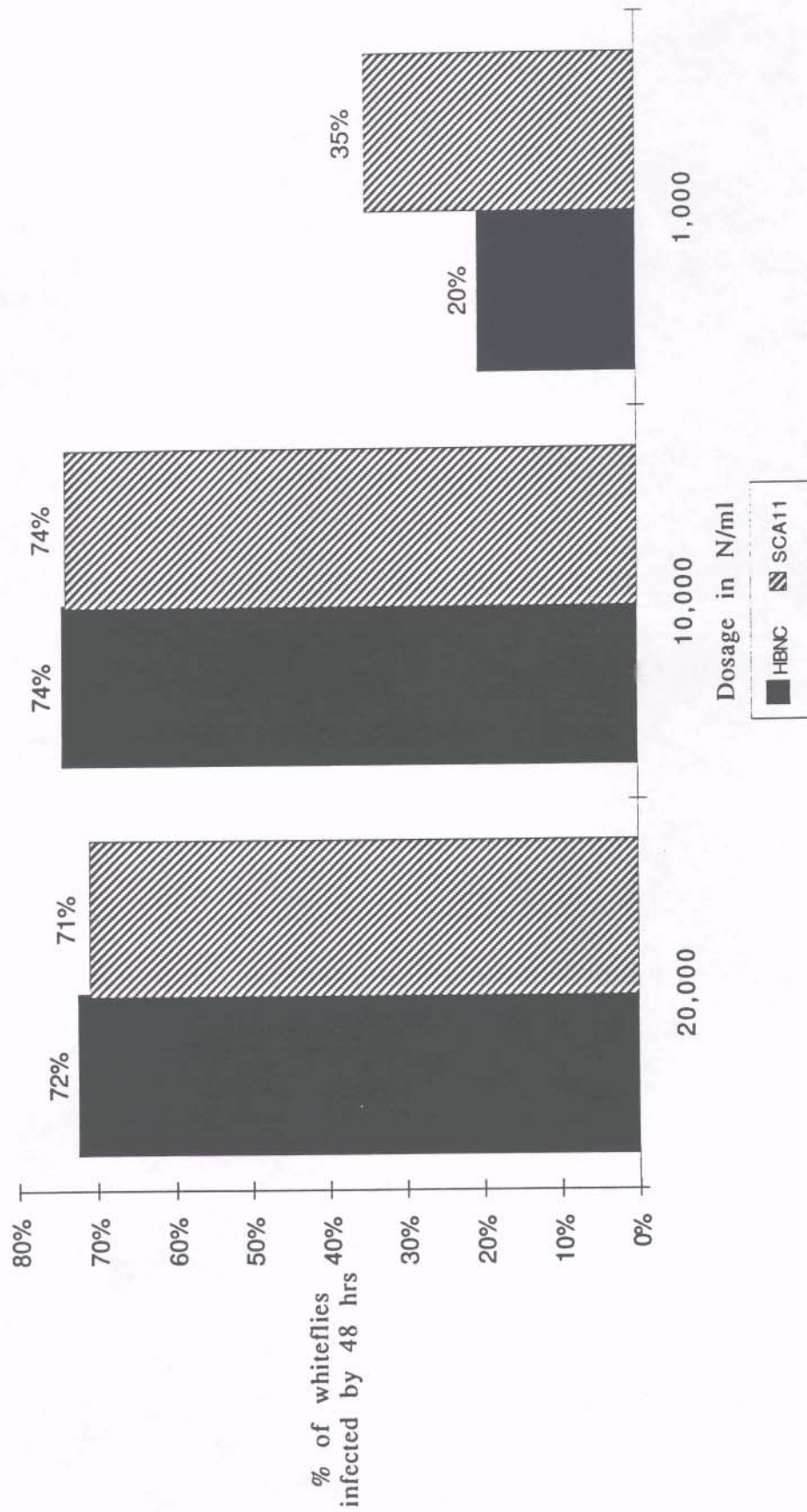
Whitefly stages of development	Total number of animals tested (exposed to nematodes)		Total infections by 48 hrs (quantified by characteristic pigmentation or presence of nematodes in body cavity)		Percentage of exposed whiteflies infected by 48 hrs	
	HBNC ¹	SCAll ²	HBNC	SCAll	HBNC	SCAll
<u>Mixed Pupae-3rd instars</u>						
Dosages:						
20,000 N/ml	271	262	196	186	72%	71%
10,000 N/ml	300	279	221	207	74%	74%
1,000 N/ml	150	147	30	52	20%	35%
<u>Adults</u>						
Dosages:						
20,000 N/ml	300	300	202	216	67%	72%
10,000 N/ml	300	300	224	182	75%	61%
1,000 N/ml	150	150	71	37	47%	25%

¹HBNC = *Heterorhabditis bacteriophora* NC

²SCAll = *Steinernema carpocapsae* All

N = nematodes

Mixed Pupae 3rd instar Whiteflies treated with HBNC & SCA11



Adult Whiteflies treated with HBNC & SCA11

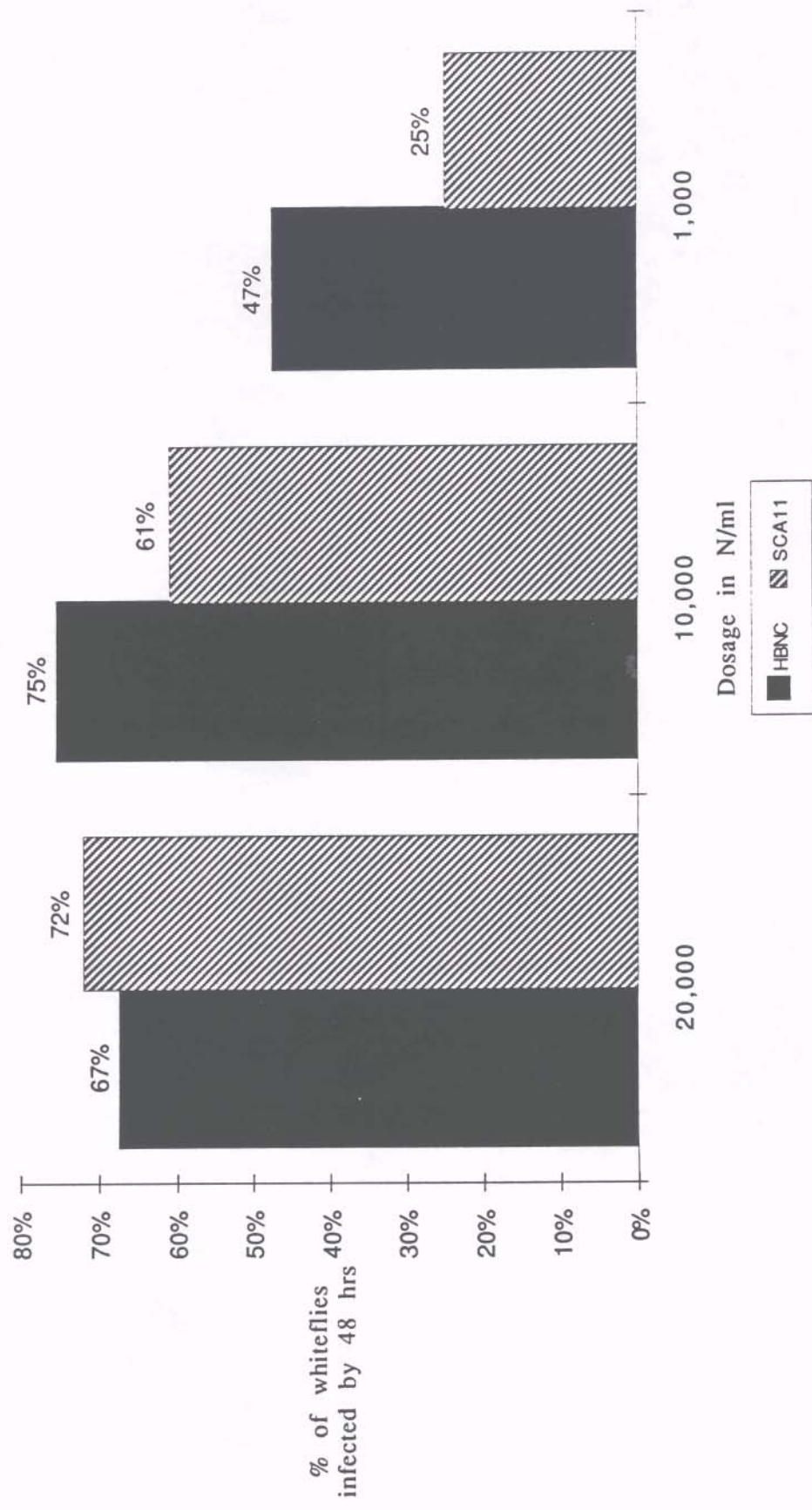


FIGURE 2

