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COMPARATIVE ACTIVITY OF DIFFERENT *HIRSUTELLA* SPECIES TOWARDS THREE PLANT PARASITIC NEMATODES

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Nematophagous fungi have often been tested for biological control of plant-parasitic nematodes but have only occasionally given encouraging results (Jatala *et al.*, 1981; B'chir, Horrigue & Verlodt, 1983; Cayrol, 1983). Very little is known, however, about endoparasitic fungi which infect nematodes with their conidia. Some studies have indicated that the endoparasite *Meria coniospora* is very agressive (Jansson, Jeyaprakash & Zuckerman, 1985*a*) and able to reduce *Meloidogyne* sp. numbers in soil (Jansson, Jeyaprakash & Zuckerman, 1985*b*). The endoparasitic fungi of the genus *Hirsutella* are mainly insect and mite parasites; there are few studies concerning nematophagous activity. Sturhan and Schneider (1980) described a new species, *H. heteroderae*, which parasitized the hop cyst nematode, *Heterodera humuli*. Later, Eayre, Jaffee and Zehr (1983) indicated that *H. rhossiliensis* suppressed populations of *Criconemella xenoplax*. Our objective was to test the activity of several different species of *Hirsutella* against three plant-parasitic nematodes.

Material and methods

FUNGI

All of the isolates tested were obtained from Centraalbureau voor Schimmelcultures, Baarn, Netherlands and included the known insect or mite parasites, *H. thompsoni*, *H. satumaensis*, and *H. subulata;* as well as five different strains of *H. rhossiliensis* isolated from nematodes. The origin of isolates tested is indicated in Table 1. All fungi were cultured on potato dextrose agar (Difco 0013 - 01-4) in Petri dishes at 20°. All strains sporulated abundantly after 14 days and were ready for testing.

NEMATODES

The species tested included *Aphelenchoides fragariae* obtained from monoxenic cultures on balsam seedlings, *Ditylenchus dipsaci* extracted from infested garlic bulbs and *Meloidogyne incognita* from egg masses excised from roots of tomato.

About 300 nematodes of each species were introduced in a small drop of sterile water into separate Petri dishes of each of the test fungi. The nematodes had been surface sterilized by washing in three successive five min baths of mercurothiolic acid 1 : 1 000, streptomycin sulfate 7 : 1 000 and sterile water. The dishes were examined regularly until some conidia adhered to the cuticles of the introduced nematodes, then individuals with about 20 attached conidia were hand-picked and transferred to Petri dishes containing 2 % water agar. One hundred nematodes of each species were so transferred from each fungus. The nematodes could be easily observed on the agar and nematodes mortality estimated in comparison with control plates containing non-infected nematodes. Nematodes were considered dead when there was no reaction to stimulation with a fine needle.

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Table 1
Origins of the tested *Hirsutella*

Species and strains of Hirsutella	*Origin*	*Host*	*Depositor*
H. thompsoni	Florida	mites	Fisher (1950)
H. satumaensis	Japan	Lepidoptera	Aoki (1957)
H. subulata	USA	Lepidoptera	Mains (1951)
H. rhossiliensis			
C.B.S. 193.81	Australia	*Heterodera avenae*	Stirling (1981)
C.B.S. 194.81	Australia	*Heterodera avenae*	Stirling (1981)
C.B.S. 195.81	California	*Meloidogyne javanica*	Stirling (1981)
C.B.S. 280.82	USA	*Criconemella xenoplax*	Jaffee (1982)
C.B.S. 281.82	USA	*Criconemella xenoplax*	Jaffee (1982)

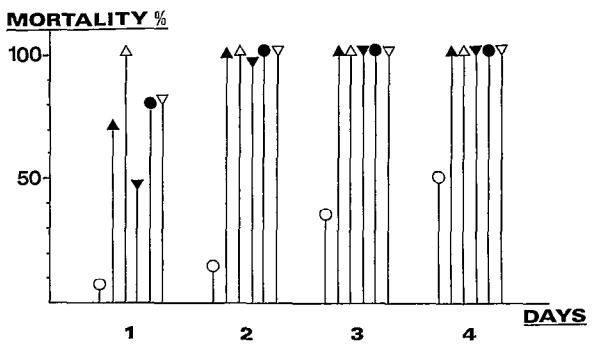

Fig. 1. Comparative mortality of *Meloidogyne incognita* juveniles in presence of five strains of *Hirsutella rhossiliensis*. (○ : Control; ▲ : Strain number 193-81 C.B.S. Baarn; △ : Strain number 194-81 C.B.S. Baarn; ▼ : Strain number 195-81 C.B.S. Baarn; ● : Strain number 280-82 C.B.S. Baarn; ▽ : Strain number 281-82 C.B.S. Baarn).

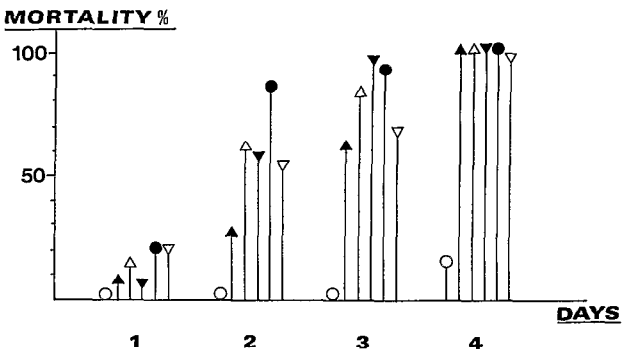

Fig. 2. Comparative mortality of *Ditylenchus dipsaci* in presence of five strains of *Hirsutella rhossiliensis* (Legends : see Fig. 1).

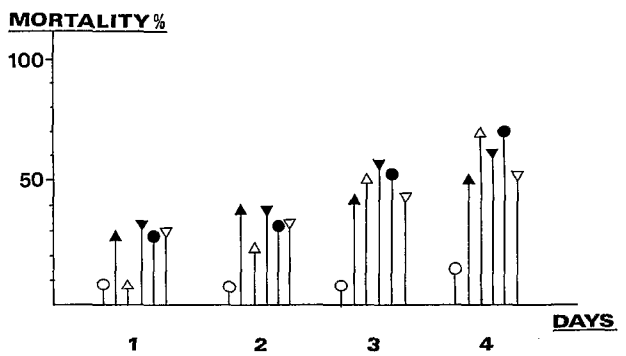

Fig. 3. Comparative mortality of *Aphelenchoides fragariae* in presence of five strains of *Hirsutella rhossiliensis* (Legends : see Fig. 1).

Results

We never observed the adhesion of conidia of the three arthropod parasitizing species, *H. thompsoni*, *H. satumaensis* and *H. subulata* on the cuticles of any of the tested nematodes. The five isolates of *H. rhossiliensis*, however, caused varying degrees of death to each nematode (Figs 1, 2, 3).

All of the *H. rhossiliensis* isolates killed *D. dipsaci* within four days but only 45-65 % of the *A. fragariae* in that length of time. *M. incognita* juveniles were killed by all of the *H. rhossiliensis* isolates in about two days. In

control plates *D. dipsaci* and *A. fragariae* both exhibited a natural mortality of about 10 % after four days but the natural mortality of *M. incognita* juveniles reached about 50 % after four days under the conditions of this experiment.

Discussion

The absence of adhesion to the nematode cuticle by conidia of the three insect-parasitic species of *Hirstutella* is of particular interest since it suggests a specific specialization and molecular recognition between species of the genus and host organisms in the habitat. Perhaps this recognition is represented by a specific lectin-carbohydrate interaction as suggested by Nordbring-Herts and Jansson (1984). Further studies are necessary to verify this possiblity.

The ability of the five strains of *H. rhossiliensis* to parasitize nematodes is very similar. They all killed *M. incognita* juveniles relatively quickly and destroyed *D. dipsaci* dauer larvae somewhat more rapidly than the *A. fragariae.* There were no noticeable differences in behavior between the strains even though they were originally isolated from different nematodes (Tab. 1). The slight retardation in early parasitism of *M. incognita* juveniles by strain No. 195.81 which was isolated from *Meloidogyne* sp. may be of some significance, however.

It is possible that each strain of *H. rhossiliensis* may have a typical specificity toward different species of nematodes but this could not be established within the parameters of this study. Jansson, Jeyaprakash and Zuckerman (1985*a*) observed such specificity for the fungus *M. coniospora.* According to these authors, cuticular penetration by the conidia is preceded by collagenase induction. Collagenase appears to be necessary for the breakdown of the primary cuticular structures.

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ESTIMATION OF THE OCCURRENCE IN SOIL OF INFECTIVE JUVENILES OF ENTOMOGENOUS NEMATODES (RHABDITIDA : STEINERNEMATIDAE, HETERORHABDITIDAE)

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For the evaluation of the role of entomogenous nematodes as a limiting factor in the occurrence of soil pests, an application of the extraction method for infective juveniles from soil can be used (Bedding & Akhurst, 1975; Mráček, 1980, 1982). We are interested in the evaluation of the optimal number of soil samples, which should be collected from the investigated ecosystem to allow one to make the statement that nematodes are present in the environment.

Four areas were investigated (1 ha each) in the locality of Warsaw. These areas represent various ecosystems : 1) An area of intensive potato cultivation; 2) A rye field;

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