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Efficiency of Controlling *Meloidogyne javanica* Using *Pasteuria penetrans* in Different Host Plants

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ABSTRACT. The degree of control of Meloidogyne javanica using Pasteuria penetrans in six different host plant species was investigated. Varying degrees of control of the nematode was shown in four host plant species, namely, aubergine (var. Dusky), tomato (var. Tiny tim), cowpea (var. Black eye) and navybean. Chilli pepper and soyabean did not contain nematodes despite being grown in nematode inoculated soil. P. penetrans controlled the total final nematode population in aubergine, tomato, cowpea and navybean host plants by reducing the egg production of female nematodes and the number of second stage juveniles in the second generation.

INTRODUCTION

Root-knot nematodes, (*Meloidogyne* spp) are of considerable importance in world agriculture as they are one of the major crop pests in the tropics. Several species of *Meloidogyne* are known to reduce both the quality and quantity of the yield of many economically-important crops.

Crop rotation, fallowing, breeding for resistant varieties, soil sterilization, use of nematicides are some of the possible methods to control the root-knot nematodes. None of the methods, by itself, is likely to provide a complete control as each method has its own merits and demerits.

As an alternative to the above-mentioned control measures, the possibility of using microorganisms such as several species of fungi,

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bacteria and viruses to control root-knot nematodes has also been investigated (Jatala, 1986).

Pasteuria penetrans which is a mycelial and endospore forming bacterium, can be highlighted among many other potential biological control agents as a promising microorganism for the control of *Meloidogyne* spp (Stirling, 1984; Brown *et al., 1985)*

P. penetrans is an obligate parasite of some plant pathogenic nematodes including *Meloidogyne* spp. and its ability to control many *Meloidogyne* spp is well documented (Stirling, 1984; Brown *et al., 1985).*

The overwintering spores of P. penetrans, present in the soil, get attached to the cuticle of second stage juveniles (J_2), of *Meloidogyne* which are the infective stages of this pest. These spores get germinated upon entry of the J_2 into a host root. As a result, the body of the adult female *Meloidogyne* gets filled with about 2 million spores of *P. penetrans* instead of eggs of its own. Thereby, *P. penetrans* inhibits the egg production of root-knot nematodes and thus, reduces the formation of subsequent generations of this species (Stirling, 1984; Sayre, 1980, 1986).

The objective of the present study was to investigate the efficiency of *P. penetrans* in controlling *M. javanica* in six different host plants.

MATERIALS AND METHODS

All the experiments were conducted in the nematology laboratory and glasshouses at Earley Gate, University of Reading, United Kingdom.

Six host plant species from two families, namely Solanaceae and Leguminosae were used: Tomato (*Lycopersicon esculentum var.* Tiny tim); chilli pepper (*Capsicum annuum*); aubergine (*Solanum melongena var.* Dusky); soyabean (*Glycine max*); navybean (*Phaseolus vulgaris*) and *cowpea* (*Vigna unguiculata var.* Black eye). Populations of *M. javanica* which were used in this experiment were originally isolated from Malawi and since then they have been continuously cultured on tomato plants (*var.* Tiny tim) grown in glasshouses of University of Reading, U.K.

Three week-old solanaceous crops were transplanted into pots of 1 litre capacity. Legume seeds were initially planted in 0.3 litre pots. Thereafter, they were thinned to one plant per pot one week after sowing and later repotted in pots of 1 litre capacity in order to cause a similar root disturbance as in the solanaceous crops.

John Innes No. 2, commercially sterilized compost mixture was used as the potting media. Host plants were maintained for 6 weeks in a glasshouse having an average day temperature of 26^{0} C. The plants were watered daily and also treated frequently with Beyleton at a concentration of 0.025% to control the powdery mildew infestation on tomato.

Two treatments were given one week after transplanting. The treatments used in the experiment included:

T_1 = *M. javanica* only

T_2 = *M. javanica* J_2s pre-encumbered with *P. penetrans* (P_p) spores

Second stage juveniles (J_2) of *M. javanica* were collected by incubating matured egg masses in hatching trays at 26^{0} C.

1-3 days old J_2s were used for the experiment. These J_2s were pre-encumbered with the spores of *P. penetrans* by leaving the J_2s in a spore suspension of *P. penetrans* for 24 hours (Stirling and Wachtel, 1980). By this method it was able to pre-encumber an average of 5 spores per juvenile. The encumbered spore count was observed under a magnification of 12.5 x 20.

M. javanica J_2s with and without *P. penetrans* spores were inoculated into the plants as a soil drench. Each pot was inoculated with 2000 J_2s and each treatment was replicated five times. Plants were arranged in a randomized complete block design. Six weeks after inoculation, plants were uprooted and checked to assess the final nematode population.

Total final nematode population included total soil population and root population of *M. javanica* excluding eggs that might be present in the soil.

Free living stages of the nematode (*i.e.* J_2s and probably male nematodes) in the soil were extracted by using Whitehead and Hemming s method described by Hooper (1985). Nematodes extracted from soil (only J_2s found) were estimated by counting the number of *M. javanica* J_2s in a known volume of the nematode suspension. The respective numbers of different growth stages (*i.e.* eggs, J_2s, J_3+J_4 stages and adult females) were estimated by using macerated root samples which were stained by 0.1% acid fuschin (Bridge *et al.*, 1982). Having found the total final nematode population in six host plant species, the degree of the control of the nematode population due to the presence of P. penetrans was calculated. The following equation was used for the calculation.

$$\% \text{ Control} = \frac{P(T_1) - P(T_2)}{P(T_1)} \times 100$$

where;

$P(T_1)$ = Total final nematode population in the soil – root system under T_1 treatment

$P(T_2)$ = Total final nematode population in the soil – root system under T_2 treatment

Where appropriate, homogeneity tests and relevant transformations were done, the significance of the effects of the two treatments were determined using Analysis of Variance (ANOVA).

RESULTS

Figure 1 shows the mean final total population of *M. javanica* in host plants under the two treatment conditions.

Except chilli pepper and soyabean, all the other tested hosts had varying amounts of different growth stages of the nematode both in the soil and in the roots.

Total final nematode population in plants having *P. penetrans* was always less than that in the plants which did not have the biological agent ($P<0.05$). There was a significant difference in total final nematode population between different host plants ($P<0.001$).

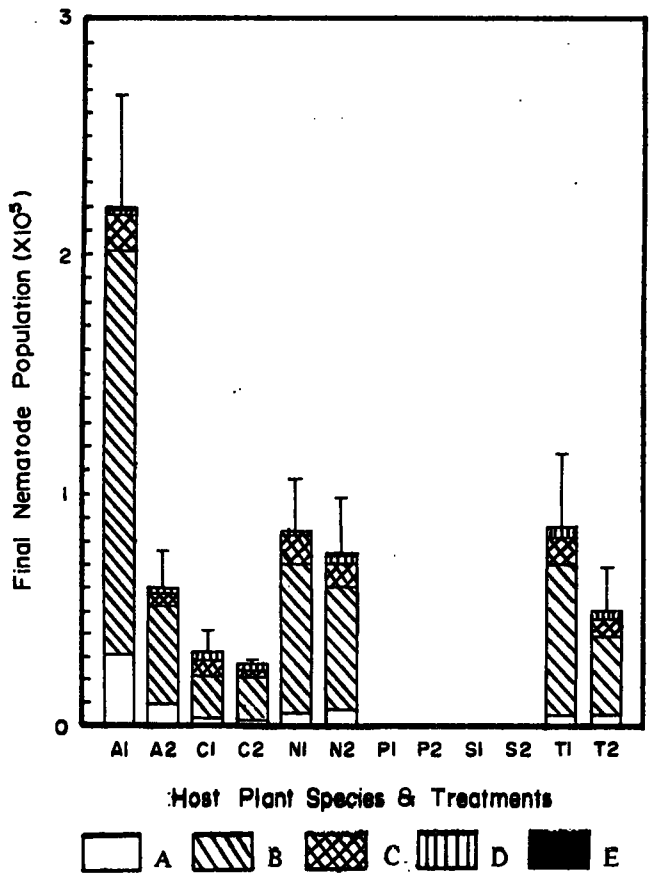

Figure 1. **Mean final total nematode population and its breakdown into different stages in different host plant species under different treatments.**

Different stages: A = Adult females in root: B = Eggs in root; C = j_2 s in root; D = j_2 s in soil: E = Developing stages (i.e j_3 and j_4) in root. Host plant species: A = Aubergine: C = Cowpea; N = Navy bean; P = Chilli pepper; S = Soyabean: T = Tomato. The number which follows each host plant species denotes the treatment. Treatments: 1 = Meloidogyne javanica only: 2 = M. javanica with pp. The vertical error bars are the standard errors of the mean total nematode populations.

The highest total final nematode population in the absence of *P. penetrans* was observed in aubergine. Tomato, *cowpea* and navybean respectively, showed the next higher nematode populations.

The number of eggs produced by *M. javanica*, the J_2s present in the soil and the root systems at the end of one life cycle is significantly ($P<0.05$) reduced by the host plant species and the different treatments. Although the interaction of the two effects above was not significant, the type of host species, the different treatments given and their interactions did not significantly ($P<0.05$) reduce the numbers of J_3+J_4 stages and adult females present in the roots.

Percentage control of *M. javanica* achieved by exposing them to *P. penetrans* in different host plants are given in Table 1.

Table 1. Percentage control of *Melodogyne javanica* (2000 J_2s per plant) using *Pasteuria penetrans* in different host plant species (Mean of five plants).

Host Plant Species	% Control
Aubergine (Var. Dusky)	73
Tomato (Var. Tiny tim)	40
Cowpea (Var. Black eye)	16
Navybean	12
Chilli pepper	–
Soyabean	–

The highest control of the nematode (*i.e.* 73%) was achieved in aubergine plants treated with nematodes encumbered with *P. penetrans* whilst it was comparatively lower in tomato, *cowpea* and navybean (*i.e.* 40%, 16% and 12% respectively). Control of nematodes could not be

estimated in chilli pepper and soyabean as the treated and control plants did not contain any nematodes.

DISCUSSION

P. penetrans exerted different levels of control in aubergine (*var.* Dusky), tomato (*var.* Tiny tim), *cowpea* (*var.* Black eye) and navybean respectively.

This varying degree of control of *M. javanica* appears to be due to the effects on the different host plants.

P. penetrans controls *Meloidogyne* spp by reducing the production of eggs by adult female nematodes (Sayre, 1980 – 1986; Davies *et al.*, 1988). The results of the present study revealed the ability of the biological agent to reduce the number of eggs and by doing so, to reduce the second stage juveniles in the next generation. This was common for all the host plants which exhibited a certain degree of control of the nematode. Therefore, it can be confirmed that the control of *M. javanica* was done by *P. penetrans* by reducing the number of egg production in female root – knot nematodes.

The reasons for the absence of *M. javanica* in the tested varieties of chilli pepper and soyabean could be two fold. The plant varieties which were used in the present study could be totally immune to invasion by *M. javanica* or may have partial resistance to the nematode. In certain plants, although nematodes could invade the roots, they do not build up thereafter (De Guiran and Ritter, 1979). Sometimes there could be a post – infectional emigration of the nematodes from the host roots to rhizosphere (Herman *et al.*, 1991) and thus, the originally invaded J_2s would not have the chance to complete their life cycles in the host plant.

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