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Attachment of six populations of *P. penetrans* spores on the second stage juveniles of two population of root-knot nematodes.

D.A. DARBAN[++], B. WARYANI*****, S. R. GOWEN, A. R. ABBASI* R A. MEMON**

Department of Agriculture, School of Agriculture Policy and Development, The University of Reading, Earley Gate P.O. Box 237, Reading Rg6 6AR, UK.

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Abstract: Abstract: The attachment studies on the second stage juveniles of rot-knot nematodes were carried out using six populations of hyperparasite (*Pasteuria penetrans*). The double blend population performed very significantly higher than the all other populations and Malakisi population of *P. penetrans* remained at second. However, greater numbers of spores attached to juveniles of Hypericum than the Navaisha population of nematodes. There was a significant ($P<0.05$) difference between all populations either in the number of spores per juvenile or percentage of spore encumbered juveniles. The attachment level of double blend remained same between 6 to 24 hours. However, the Malakisi population has shown that attachment level was continuously increasing until the last time of assessment time period of 24 hours (figure 3 a-b). In (figure 2 a – b) both populations of *P. penetrans* have shown very low attachment and there was no any significant increase in either the percentage of encumbered juveniles or in the total number of spores per juvenile. There was attachment comparison between six populations of *Pasteuria* (Malakisi, Thika, G11, H22, blend and double blend) with two populations of root-knot nematodes (Hypericum and Navaisha). All populations of spores were added to freshly hatched juveniles of the Hypericum and Navaisha populations in order to avoid any differences in their vigour and condition of the nematodes.

Keywords: Attachment, hyperparasite, root-knot nematodes.

1. INTRODUCTION

More research work has been carried out to understand the biology of *P. penetrans* which parasitize the economically important root- knot nematode. The spores of the hyperparasite adhere to the second stage juveniles of *Meloidogyne* spp. (Barbara *et al*, 2004) when they move through soil in search of host roots. When these juveniles invade roots and begin to develop to maturity the parasite also starts developing. This hyperparasite destroys the reproduction system of female nematodes and they are unable to produce eggs and so the build-up of nematode populations is prevented (Darban *et al*, 2005). However, in certain conditions only a single spore of this hyperparasite is enough to reduce in thousands the population of parasites. The vermiform adult males leave the roots and may not be essential for reproduction. In most of the important species the life cycle of root-knot nematodes can be completed in 17–57 days depending on the factors such as soil temperature, host plant and nematode population. The optimum temperature for root-knot nematodes is 25–30°C, however at higher temperatures, the life cycle is shorter and more females develop, 10 days at 35°C as compared to 50 days at 15°C *M. hapla* has a shorter generation time than *M. javanica* at all temperatures below 21°C. The nutritional status of the host plant can affect the sex ratio; most *M. javanica* males were found in nitrogen deficient plants at high inoculation rates. Stressed roots also reduce the fecundity of females (Ferris *et al.*, 1985). In favorable conditions, males will have one testis and in unfavorable conditions, the female juveniles with two ovaries revert to males with two testes (Eisenbeck and Triantaphyllou, 1991).

Management of Disease Caused By Plant Parasitic Nematodes

Nematodes are among the most difficult crop pests. The management of disease caused by parasitic nematodes has been achieved by different methods such as crop rotation, use of resistant varieties, chemical nematicides and other cultural practices. The development of new nematicides is a difficult task, because most plant-parasitic nematode species live in the soil or within plant roots. The target of any chemical nematicides often resides at a fair distance away from the site of application. Moreover, the nematode cuticle and other surface structures are impermeable to many organic molecules. Consequently, most nematicides have tended to be rather toxic or volatile, with poor target specificity and less than perfect human or environmental safety, such as ground water contamination or atmospheric ozone depletion. Therefore, a very little number of chemical nematicides survived in market to give least rid off from nematode problem of many crops specially in vegetable crops

[++]Corresponding author E-mail: d_darban@hotmail.com TEL. NO: 00447557041495
*Department of Fresh Water Biology, University of Sindh, Jamshoro
**Institute of Plant Sciences, University of Sindh, Jamshoro-76080,

without caring the destruction of very close friends of environment (fauna and flora). The economic cost of research and registration of new chemicals is an enormous hurdle for a prospective new chemical nematicide to overcome. Agrochemical companies are more likely to channel research spending into products with a potentially high market value like herbicides and insecticides rather than nematicides. The incorporation of organic residues strongly impacts the physical and biological properties of soil and may promote an environment favorable to nematode- antagonistc microorganisms (Stirling 1991).

2. MATERIALS AND METHODS

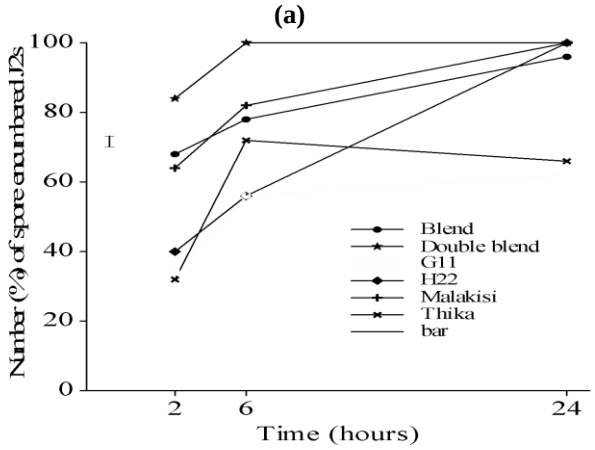
Pasteuria penetrans spore suspensions were prepared from six different populations of *P. penetrans* (Malakisi, Thika, G11, H22, Blend and Double blend) by soaking root powder containing endospores; 100 mg of root powder was diluted into 100 ml of water. Three aliquots were taken to estimate the number of mature endospores with the aid of a haemocytometer at x400 magnification. Suspensions of 200 juveniles from two different populations of root- knot nematode Hypericum and Navaisha were placed in 3cm diameter Petri dishes. These were added to suspensions of 50,000 spores from each population were mixed with nematode suspensions. The total volumes of all suspensions/rep were made up to 4 ml by adding SDW as required. Then all Petri dishes were placed in an incubator at 28°C. The experiment was arranged in a completely randomised block design with 2 replicates with assessment of 25 juveniles per replicate.

3. RESULTS

The attachment studies were carried out using six populations of hyperparasite. The results have shown that double blend population performed very significantly higher than the all other populations and Malakisi population of *P. penetrans* remained at second. However, greater numbers of spores attached to juveniles of Hypericum than the Navaisha population of nematodes. There was a significant ($P<0.05$) difference between all populations either in the number of spores per juvenile or percentage of spore encumbered juveniles (Fig. 3 a-b). The attachment level of double blend remained same between 6 to 24 hours. However, the Malakisi population has shown that attachment level was continuously increasing until the last time of assessment after 24 hours. In **(Fig. 3 a-b)** both populations of *P. penetrans* have shown very low attachment and there was no any significant increase in either the percentage of encumbered juveniles or in the total number of spores per juvenile.

4. DISCUSSION

The attachment was compared between six populations of *Pasteuria* (Malakisi, Thika, G11, H22, blend and double blend) with two populations of root-knot nematodes (Hypericum and Navaisha). All populations of spores were added to freshly hatched juveniles of the Hypericum and Navaisha populations in order to avoid any differences in their vigor and condition of the nematodes. The results have shown that there was highly significant difference between the spore attachment of all populations among the two populations of nematodes. There was no significant difference between G11 and Thika populations of *P. penetrans*. The relatively low attachment levels of Thika population remained persisted until 24h exposure, even then there was a number of juveniles without spores and some juveniles that remained without spores **(Fig. 3 a-b).** The spores from all populations did show significant differences of attachment to the juveniles of Navaisha population of nematode until 24h of incubation period **(Fig. 4 a-b).** However, as compared to Hypericum population of nematodes**.** the attachment level was low either in percentage of encumbered juveniles or in total number of spores per juveniles. It could be concluded that attachment of the spores of all six population on the hypericum population of nematodes is more persistent, which could create more chances to increase infection in root-knot nematodes than on the Navaisha population of nematodes.



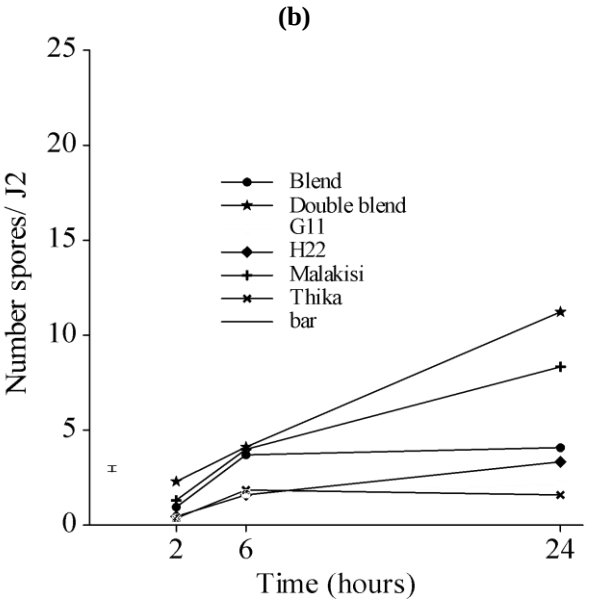
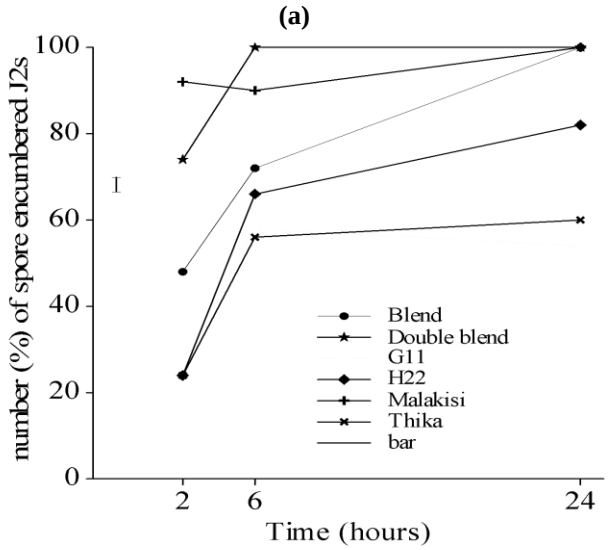


Fig.3 (a-b). Adhering efficacy of six populations of *P. penetrans* (Blend, Double blend, G11, H22, Malakisi and Thika) on root-knot nematode (Hypericum) population. 25J2s/replicate were examined to determine the percentage of encumbered J2s (fig.-a) and no. of spores/J2 (fig-b). Error bars represent LSD of means (data are means of 2 replicates).



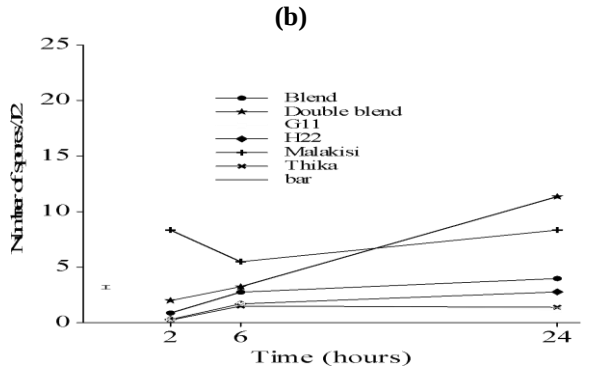


Fig.4 (a-b). Adhering efficacy of six populations of *P. penetrans* (Blend, Double blend, G11, H22, Malakisi and Thika) on root-knot nematode (Navaisha) population. 25J2s/replicate were examined to determine the percentage of encumbered J2s (fig.a) and no. of spores/J2 (fig-b). Error bars represent LSD of means (data are means of 2 replicates).

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