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COMPARATIVE EXISTENCE OF *STEINERNEMA ABBASI* AND *HETERORHABDITIS INDICUS* IN SOIL OF A DATE PALM PLANTATION

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Abstract

The entomopathogenic nematodes, *Steinernema abbasi* and *Heterorhabditis indicus* were found to exist in a date palm plantation from January to December, 2001. Populations of *S. abbasi* (expressed by percent mortality in larvae of *Galleria mellonella* (as baits) in 20 soil samples collected monthly were higher than those of *H. indicus*. Mortality by *S. abbasi* reached 100 % in March, August, October and November. The co-existence of *S. abbasi* and *H. indicus* in the soil around the trunks of palm trees resulted in successful combined infection with both species in five *G. mellonella* larvae during the course of this study. Such infected larvae showed the symptoms of either nematode species and the combined infection was detected in the second passage when some infected larvae showed symptoms of *S. abbasi* (no change in colour with softness of the cadaver) and others showed symptoms of *H. indicus* (brownish -red colour with hardness in colour). The study was conducted in Ras Al-Khaima, United Arab Emirates.

INTRODUCTION

Entomopathogenic nematodes in the families Steinernematidae and Heterorhabditidae are widely regarded as being promising biological control agents for a number of insect pests in soil and cryptic habitats (Gauglar and Kaya 1990 ; Kaya and Gauglar 1993) . The free-living, nonfeeding infective juveniles (Ijs) of these nematodes possess attributes of both insect parasitoids or predators and microbial pathogens. Like parasitoids / predators they have chemoreceptors and are motile to look for the host ; like pathogens they are highly virulent, killing the host quickly and can be cultured easily in vitro . These two families are associated with mutualistic bacteria in the genera *Xenorhabdus* (with Steinernematidae) and *Photorhabdus* (with Heterorhabditidae). They have positive characters including their broad host range (Gauglar 1981), safety to vertebrates , plants and other nontarget organisms (Akhurst 1990 ; Poinar 1989) ,

exempting from registration in many countries (Gorsuch 1982) , easily applied using standard spray equipment (Georgis 1990) , compatible with many chemical pesticides (Forschler *et al.* 1990 ; Hara and Kaya 1983 ; Rovesti and Deseo 1990 ; Rovesti *et al.* 1988 ; Zimmermann and Cranshaw 1990) and amenable to genetic selection (Gauglar 1987). These positive attributes and the need to find alternative methods of insect pest control to chemical insecticides have been major factors for the rapid commercialization of these biocontrol agents since 1980,s (Georgis 1990).

Abbas *et al.* (2001, a) surveyed the entomopathogenic nematodes in Ras-Al-Khaima, UAE, and obtained six species including *S. abbasi* and *H. indicus* .

The present investigation deals with the natural existence of *S. abbasi* and *H. indicus* in the soil of a date palm plantation during 2001. The coexistence of both species in the same host is discussed .

MATERIAL AND METHODS

This study was carried out in a date palm plantation at Al-Hamranyia Researh Station, Ras Al-Khaima, United Arab Emirates. The plantation contained about 900 date palm trees planted in 1988 and all agricultural practices are conducted regularly with water flooding irrigation system at 2-3-day intervals.

Soil samples were collected 5 – 10 cm below soil surface, around the trunk of the trees, monthly from January to December 2001. Almost 5 litre of soil was taken from each tree in a plastic bucket and transferred to the laboratory. Twenty samples, representing 20 trees, were collected monthly. The greater wax moth, *Galleria mellonella* was used as a baiting technique according to Bedding and Akhurst (1975) . Soil sample of each tree was thoroughly mixed and 10 baiting traps (10 replicates) with a *G.mellonella* larva each were made from each sample and kept in a conditioned chamber of 27 ± 1°C . Four days later , the traps were checked and the dead larvae were counted and transferred to the extraction units (Dutky *et al.*1964) . The infective juveniles were harvested after emerging from *Galleria* cadavers and mounted in glycerol The morphological characters of such isolated nematodes were compared to those of *S. abbasi* and *H. indicus* which had been isolated from the same plantation and maintained in the laboratory (Abbas *et al.* 2001, a). Percentages of existence of each of the two

species in the 20 monthly soil samples and in the 10 replicates of each sample were calculated .

The parameters of soil ; texture, temperature, pH and salinity were measured. Soil temperature was measured daily, 5 – 10 cm below soil surface at 7.0 am, 13.0 and 18.0 pm. In addition, air temperature was also measured at the same times .

RESULTS

1. Soil index

Texture: sandy-loam ; temperature : minimum : 15.9 ° C (in February); maximum : 36.5 °C (in July) 5 – 10 cm below soil surface, Fig.1. Salinity : slightly saline (1050 ppm) ; pH : 7.8

It should be noted that soil temperature was 0.1 – 1.3 C higher than air temperature at 7.0 am and was 4.0 – 8.4 C less than air temperature at 1.0 pm.

2. Existence of Ijs in soil

S. abbasi and *H. indicus* were found in all soil samples collected monthly during the whole year . Populations of *S. abbasi* , expressed as percent mortality of *G. mollenella* larvae baits in soil samples, were much higher than those of *H. indicus* in 11 months . Percent mortality in *Galleria* larvae reached 100 % by *S. abbasi* in the samples of soil collected in March, August, October and November, Fig.1. In addition, percent mortalities in the 10 replicates of each of the 20 monthly samples averaged 59.2 % (16 – 100 %) for *S. abbasi* and 31.3 % (11 – 66 %) for *H. indicus* during the course of this study (Fig. 2).

As for the combined infection, data showed that the coexistence of both *S. abbasi* and *H. indicus* in the soil, around date palm trees, and their competition in attacking the available target insect resulted in successful combined infection with both species in 5 *G. mellonella* larvae in the soil samples collected during the course of this study. The combined infected larvae showed the symptoms of either nematode species; brownish-red colour with hardness of the cadaver (for *H. indicus*) or no change in

colour with softness of the cadaver (for *S. abbasi)* . Such combined infection could be detected in the five cases as follows : 1- In the 1st case the cadaver showed symptoms of *H. indicus* infection, but infective juveniles (Ijs) of *S. abbasi* emeged from this cadaver 6 days post trapping, while the Ijs of *H. indicus* emerged from the same cadaver 3 days later. Re-infection of new *Galleria* larvae with the emerged Ijs ensured this finding. 2- In 3 cases the cadavers showed symptoms of *S. abbasi* infection and when the harvested Ijs were used for infecting new larvae the most infected larvae showed the symptoms of *S. abbasi* and a few number showed the symptoms of *H. indicus.* 3- In the last case the cadaver showed symptoms of *H. indicus* infection and the migrated Ijs were stored in the laboratory at 20 ^{0}C. When such Ijs were used to infect new *G. mellonella* larvae two months later, the infected larvae showed the symptoms of *S. abbasi* and produced only the Ijs of *S. abbasi.* This finding is interpreted as the Ijs of *H. indicus* which emerged with those of *S. abbasi* from the same cadaver could not survive the two-month storage at 20 °C .

DISCUSSION

The present study indicated that *S. abbasi* and *H. indicus* were found to coexist in the soil of a date palm plantation during the whole year. The populations of both nematode species seem to be high especially *S. abbasi* as percentage of mortality in *G. mellonella* larvae baits reached 100 % in the 20 soil samples collected in March, August, October and November, 2001. The soil parameters seem to be optimum to both nematode species ; the texture is sandy-loam , pH is 7.3 , salinity is low (1050 ppm) , temperatures (at 5 – 10 cm below soil surface) ranged 15.9 – 36.5°C and the soil moisture is almost suitable as irrigation is applied at 2 – 3 day intervals (flooding system). Acceptable temperature ranges for survival and development of entomopathogenic nematodes vary with the nematode species and strain (Kaya, 1977; Molyneux, 1985 & 1986; Simons and van der Schaaf, 1986) . Kung *et al.* (1990) studied the survival of *Steinernema carpocapsae* and *S. glaseri* in four different soils and found that the survival of *S. carpocapsae* was higher in sandy loam soil while *S. glaseri* survived longer in sandy soil. Kondo and Ishibashi (1985) reported that survival, activity and infectivity of insect nematodes are affected by soil moisture and such characters peaked for *S. feltiae* at soil moisture of 25 – 40 % . In most target habitats pH, photoperiod

and salinity are unlikely to be seriously limiting factors for nematode survival and infectivity (Gaugler 1981). However, Kung *et al.* (1990) found that survival of *S. carpocapsae* and *S. glaseri* reached the peak at pH 8, but declined sharply at pH 10 .

Suitable available hosts in soil could be other factor for survival and existence of *S. abbasi* and *H. indicus* in date palm plantations. Among these hosts are the scarabid larvae *Oryctes spp.* and the adults of the red palm weevil *Rhynchophorus ferrugineus* (the latter inhabit the soil occasionally for shade and moisture). What supports this claim is that both *S. abbasi* and *H. indicus* were isolated from adults of *R. ferrugineus (*Abbas *et al.* 2001,b*)* and *Oryctes spp.* (unpublished) . Also, it is noteworthy that although soil samples collected from a clover field, 300 m apart from the date palm plantation, caused 97 % mortality in *G. mellonella* larvae baits (by *Heterorhabditis bacteriophora*) in February 2001, the mortality decreased to 25 % in soil samples collected in April and to nil in the samples collected monthly from May to December (unpublished). Soil parameters of the clover field are the same as of the date palm plantation.

The coexistence of *S. abbasi* and *H. indicas* in the soil of date palm plantation resulted in combined infection with both species in 5 *G. mellonella* larvae during the course of this investigation. Both species could develop in the same host and produced infective juveniles emerged from the cadavers. This finding seems contraversial because it means that both nematode species passed ,at least, one generation feeding on the two genera of associated mutualistic bacteria ; *Photorhabdus* and *Xenorhabdus*. It was documented that heterorhabditid and steinernematid nematodes could not coexist in the same host (Alatorre-Rosas and Kaya 1990, 1991) and the nematodes from one genus could not feed on the mutualistic bacteria from another genus (Akhurst, 1983). Furthermore, Kondo (1989) reported that an antagonistic relationship might exist between steinernematid species in the same host.

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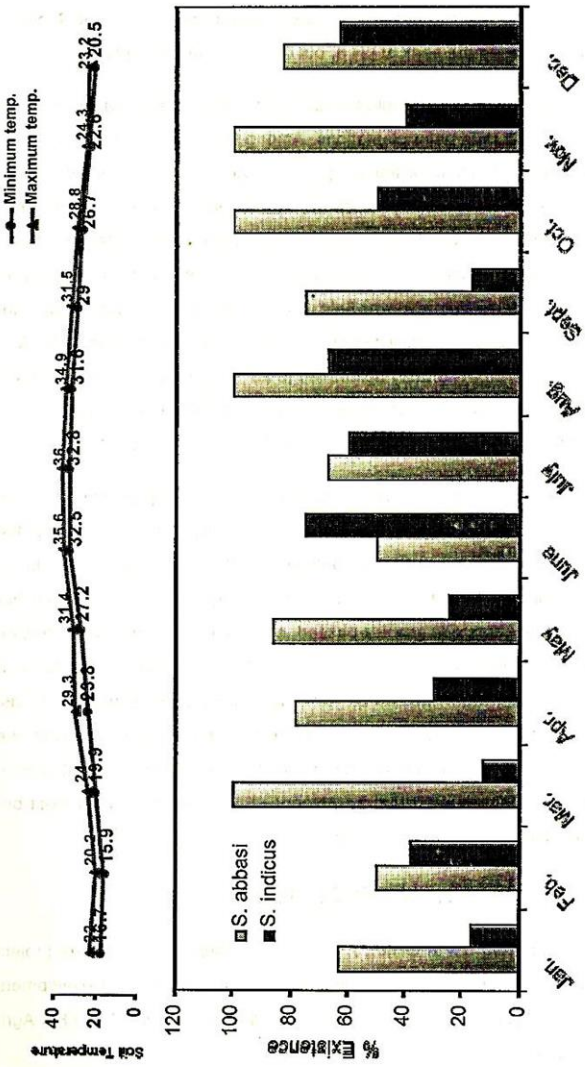

Fig. 1. Percentage existence of *S. abbasi* and *H. indicus* in 20 monthly samples of soil collected from a date palm plantation during 2001.

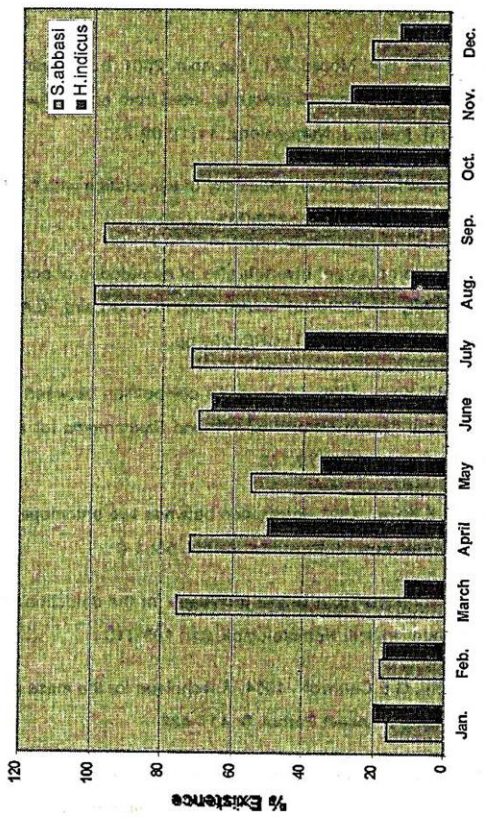

Fig.2. % Existence of *S.abbasi* and *H.indicus* in the 10 replicates of each of the 20 monthly soil samples collected from a date palm plantation in 2001

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مقارنة تواجد نوعى النيماتودا *STEINERNEMA ABBASI* و *HETERORHABDITIS INDICUS* بالتربة فى أحد مزارع نخيل التمر

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وجد أن نوعى النيماتودا ستاينرنيما عباسى، هيتيرورابديتيس إنديكاس يتواجدان بالتربة فى مزرعة النخيل طوال العام (من يناير إلى ديسمبر ٢٠٠١) إلا أن الكثافة العددية للنوع الأول كانت أكثر من الكثافة العددية للنوع الثانى.

وقد وصلت نسبة موت يرقات دودة الشمع (عند استخدامها كمصيدة للنيماتودا فى عينات التربة) إلى ١٠٠ ٪ فى أشهر مارس، أغسطس، أكتوبر، نوفمبر بالنوع ستاينرنيما عباسى.

وقد أظهرت هذه الدراسة أن كلا نوعى النيماتودا قد تكاثرا معاً فى عائل واحد ثم انفصلا بعد ذلك عند العدوى التالية ليرقات جديدة.