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OCCURRENCE OF *MELOIDOGYNE* SPP. AND RACES ON THE ISLAND OF CYPRUS

by

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Root-knot nematodes (*Meloidogyne spp.*) are widespread and destructive pests of crop plants in Cyprus. Georghiou (1957) reported *Meloidogyne javanica, M. arenaria* and an unidentified species while Philis and Siddiqi (1976) added *M. incognita* to the list. Netscher (1978) reported that in subtropical countries the length of juveniles and the configuration of the perineal pattern do not adequately serve in identifying *Meloidogyne* while the position of the excretory pore of the adult female seemed to be useful for distinguishing between *M. javanica* and *M. incognita*. Taylor and Sasser (1978) used several kinds of plants for separating species and races of *Meloidogyne* by host response while Eisenback *et al.* (1981) established several morphological features for distinguishing the most common species of root-knot nematodes.

The object of the present study was to use the response of Sasser's differential hosts for local *Meloidogyne* populations and to determine morphological features that might contribute to their identification. The work is part of the International *Meloidogyne* Project (IMP).

Materials and Methods

Thirty nine root and soil samples were collected from naturally infested fields from 22 locations in Cyprus. The samples were taken

Table I - *Habitat of* Meloidogyne *populations collected for identification.*

Populations	Village	Glasshouse	Field	HOST	
				Common name	Variety
001	Astromeritis	+		Carnation	William Sim
002	Akaki	+		Marrow	Senator
003	»	+		Tomato	Unknown
004	Dherinia	+		Cucumber	Pepinex 69
005	Ayia Napa		+	Tomato	Marmande
006	» »		+	Egg plant	Black beauty
007	» »		+	French bean	Local
008	» »		+	Egg plant	Black beauty
009	Paralimni		+	Tomato	Marmande
010	Anafotia	+		Sweet melon	Ananas
011	»	+		Tomato	Marmande
012	Deftera		+	Celery	Unknown
013	Peristerona		+	Okra	Local
014	Astromeritis		+	Egg plant	Long purple
015	Xylophagou		+	»	Black beauty
016	Alaminos		+	French bean	Harvester
017	Limassol		+	Swiss chard	Local
018	Fassouri	+		Egg plant	Bonica
019	Pareklishia		+	Tomato	Ace VF-55
020	»		+	»	Walter
021	Emba		+	»	Local
022	Pera Orinis		+	Peach	Elberta
023	» »		+	»	»
024	K. Polemidhia	+		Egg plant	Bonica
025	»	+		Tomato	Dombo
026	Kolossi	+		»	»
027	»	+		Cucumber	Maram
028	Limassol	+		Tomato	Dombo
029	Chlorakas	+		»	50-27
030	Akhelia	+		Egg plant	Bonica
031	»	+		»	»
032	Emba		+	French bean	Harvester
033	»		+	Tomato	Ace VF-55
034	»		+	Celery	Local
035	»		+	Tomato	Local
036	Argaka		+	Tobacco	Virginia
037	»		+	»	»
038	Choulou		+	»	»
039	Letymbou		+	»	»

from 12 different crops at a soil depth 0-15 cm, most of them during summer, while the rest were collected from glasshouses during winter to early spring (Table I). After collecting the samples, three nematode-susceptible tomato seedlings were planted in 10 cm clay pots containing nematode infested root pieces from each population, for multiplication. At the same time a 20 cm sterilized clay pot was also planted with a tomato seedling to maintain each population for future observations, if necessary. All pots were kept in the glasshouse and irrigated as required. Care was taken to avoid cross contamination between populations.

Seedlings of tomato (*Lycopersicon esculentum* cv. ' Rutgers '), tobacco (*Nicotiana tabacum* cv. ' NC95 '), pepper (*Capsicum frutescens* cv. ' California Wonder '), peanut (*Arachis hypogaea* cv. ' Florruner '), watermelon (*Citrullus vulgaris* cv. ' Charleston Gray ') and cotton (*Gossypium hirsutum* cv. ' Deltapine 16 ') were transplanted into steam sterilized soil in 10 cm clay pots with 2 or 3 replicates for each population after inoculation with 10,000±500 eggs/pot. Determination of nematode species and race by host reaction was based on a differential host test, as suggested by Taylor and Sasser (1978).

A soil consisting of 85% sand 6% clay and 9% silt was used for growing the seedlings. At the time of inoculation plants were 10-15 cm high. Caution was taken that inoculated pots were weed-free as infected roots of weeds could easily be mistaken for roots of a differential host. Additional tomato plants were used for inoculations to determine if time and temperature had allowed maturation of the egg masses. After the formation of egg masses on the roots of tomato, the root systems of all plants were carefully washed and then stained by incubating roots in an aqueous solution of phloxine B (0.15 g/litre tap water) for 15 minutes to emphasize the egg masses. Egg mass and gall ratings were made according to the following scale: 0 = 0, 1 = 1-2, 2 = 3-10, 3 = 11-30, 4 = 31-100 and 5-greater than 100 galls or egg masses. Several egg masses from each population taken from tomato were hatched in a watch glass for measuring the length of 20 second-stage juveniles with the aid of a Carl Zeiss camera lucida, while 8-10 fully developed females from the same population were used for recording the position of the excretory pore. Permanent mounts of the perineal pattern for each population were also prepared.

Table II - *Behaviour of* Meloidogyne *field populations on Sasser's Differential Hosts.*

Populations	DIFFERENTIAL HOSTS						SPECIES		
	Tobacco	Cotton	Pepper	Water-melon	Peanut	Tomato	*M. javanica*	*M. incognita* Race 1	*M. incognita* Race 2
001	5(a)/5(b)	0/0	4/4	5/4	0/0	5/5			+
002	4/5	0/0	0/0	5/4	0/0	5/5	+		
003	5/5	0/0	0/0	4/4	0/0	5/5	+		
004	4/5	0/0	0/0	4/3	0/0	5/4	+		
005	5/5	0/0	0/0	5/4	0/0	5/5	+		
006	4/4	0/0	0/0	3/3	0/0	4/4	+		
007	4/4	0/0	0/0	4/4	0/0	5/5	+		
008	5/5	0/0	0/0	3/2	0/0	5/5	+		
009	2/2	0/0	0/0	4/2	0/0	5/5	+		
010	5/4	0/0	0/0	5/2	2/0	5/5	+		
011	5/5	0/0	3/3	5/5	0/0	5/5			+
012	2/2	0/0	0/0	3/3	0/0	4/4	+		
013	4/4	0/0	0/0	5/5	0/0	5/5	+		
014	4/4	0/0	0/0	4/4	0/0	5/5	+		
015	4/4	0/0	0/0	4/4	0/0	4/4	+		
016	3/3	0/0	0/0	3/3	0/0	4/4	+		
017	3/3	0/0	0/0	3/3	0/0	4/4	+		
018	5/5	0/0	0/2	5/5	0/0	5/5	+		+
019	5/5	0/0	0/3	4/4	0/0	5/5			+
020	5/5	0/0	0/3	5/5	0/0	5/5			+
021	4/5	0/0	0/3	4/4	0/0	5/5			+
022	4/4	0/0	0/2	4/4	0/0	5/5			+
023	4/5	0/0	0/2	4/4	0/0	4/5			+
024	4/4	0/0	0/2	4/4	0/0	5/5			+
025	0/0	0/0	5/5	4/4	0/0	5/5		+	
026	4/5	0/0	1/2	0/5	0/0	5/5	+		+
027	4/4	0/0	0/0	4/4	0/0	5/5	+		
028	5/5	0/0	4/4	0/5	0/0	5/5			+
029	4/4	0/0	0/1	3/3	0/0	4/4	+		+
030	4/4	0/0	0/0	3/3	0/0	4/4	+		
031	5/5	0/0	0/2	4/4	0/0	5/5	+		+
032	4/4	0/0	0/1	4/4	0/0	4/4			+
033	4/4	0/0	0/0	4/4	0/0	5/4	+		
034	5/4	0/0	3/2	5/4	0/0	5/4	+		+
035	4/4	0/0	0/0	3/3	0/0	4/4	+		
036	4/4	0/0	0/3	5/4	0/0	4/5	+		+
037	4/4	0/0	0/0	4/4	0/0	4/4	+		
038	3/4	0/0	4/4	3/3	0/0	4/4			+
039	4/4	0/0	0/1	0/5	0/0	4/4			+

(a): root galling index; (b): egg production index.

Results and Discussion

All populations, except 025, reproduced on tobacco, watermelon and tomato, while no reproduction occurred on cotton or peanut. A high degree of infestation was caused by all populations on tomato, this evidently indicating that every chance was given for the populations to reproduce on the different host plants (Table II). The small differences in degree of infection on tomato, however, expressed either as root galling or egg production is due, most probably, to the different accumulation of heat or time in the glasshouse from inoculation to root examination, as only part of the populations were examined at each time throughout the year. The different response to pepper, however, by *M. incognita* populations is far beyond the variability expected by environmental influence, as is the case with tomato (Table II). Populations 001, 011, 025, 026, 028, 034 and 038 caused galling and produced eggs on pepper while other populations, of the same species, laid eggs but without causing any visual symptoms of root galling. This can only be explained by the presence of different physiological races. Based on Sasser's Host Differential Test, 51% of the populations behaved as *M. javanica*, 31% as *M. incognita* race 2, 3% as *M. incognita* race 1 and the rest (15%) as a mixture of *M. javanica* and *M. incognita* race 2. *M. arenaria* was not found in the samples examined although it is listed in previous records. My conclusion that this species was not present in the populations examined was mainly based on the assumption that juveniles of this species are the longest in the genus, at 400 μm minimum length and usually over 450 μm and, secondly, that none of the many perineal patterns examined, even for populations 011 and 015 which had juveniles with lengths measuring 400 and 420 μm respectively, resembled those of typical *M. arenaria*. Separation of *M. arenaria* race 2, is not readily determined by Sasser's Host Differentials but race 1 is easily recognised. In 97% of the populations examined the length of juveniles was less than 400 μm and only one population averaged 420 μm (Fig. 1). The position of the excretory pore, however, was very helpful in distinguishing between *M. javanica* and *M. incognita*: this ranged from 2.4 to 3.0 times the stylet length, measured from the head tip for the former species and 1.4 to 1.9 for the latter species. Mixed populations were also observed with positions of the excretory pore measuring 2.3 to 2.5 times the stylet length, this being the result of the average value of the two species in the population (Fig. 1).

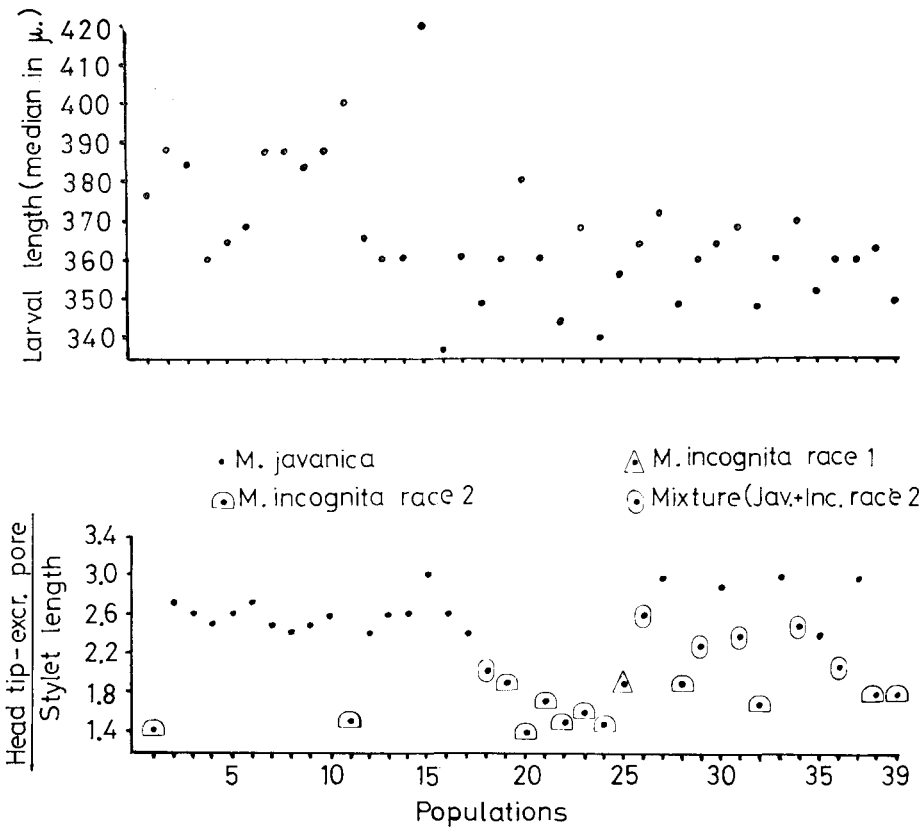

Fig. 1 - Larval (2nd stage) length and position of the excretory pore of field populations of *Meloidogyne javanica* and *M. incognita* (n=20).

The excretory pore position was measured in only a few specimens (n = 3-4) because of the difficulty of locating it; from the 8-10 specimens usually mounted on each slide, only some of them were suitable for measuring. Repetition of measurements with the other populations of the same species, however, supported also by the Host Differential Test makes the position of the excretory pore a valid criterion for distinguishing between *M. javanica* and *M. incognita*. *M. javanica* did not reproduce on pepper (cv. California wonder), cotton (cv. Deltapine 16), or peanut (cv. Florruner) while *M. incognita* race 2 reproduced on pepper, tobacco (N.C. 95), watermelon cv. Charleston gray and tomato cv. Rutgers. *M. incognita* race 1 reproduced on pepper, watermelon and tomato but not on tobacco, cotton and peanut

(Table II). Total length of juveniles cannot be used as a criterion for distinguishing between *M. javanica* and *M. incognita* (Fig. 1) and the appearance of the perineal pattern has great limitations.

This study has added to the knowledge of the occurrence of root-knot nematodes in Cyprus and it is hoped the information will be helpful in the implementation of meaningful rotational cropping system(s) for their control.

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SUMMARY

Thirty nine field populations of *Meloidogyne* collected from 22 locations on the island of Cyprus and tested with Sasser's Host Differentials, revealed the presence of *M. javanica* and *M. incognita*. *M. javanica* and *M. incognita* race 2 were the predominant species while *M. incognita* race 1 was found only in one case. Mixed populations also occurred. None of the populations tested reproduced on cotton (cv. Deltapine 16) or peanut (cv. Florruner). Morphologically, the two species can be distinguished by the position of the excretory pore, this measuring 2.4 to 3.0 times the stylet lenght from the head tip to the excretory pore for *M. javanica*, and 1.4 to 1.9 times for *M. incognita*. Lenght of juveniles was of no value in distinguishing between these two species and the perineal pattern of the female also had little value.

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