Interaction between *Meloidogyne incognita, Rotylenchulus reniformis* and *Tylenchorhynchus brassicae* on tomato

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SUMMARY

The interaction between root-knot *(Meloidogyne incognita),* reniform *(Rotylenchulus reniformis)* and stunt *(Tylenchorhynchus brassicae)* nematodes on tomato roots was studied using varying inoculum levels and their combinations. The parameters measured were rate of population increase, sex-ratio and plant growth. In single species inoculations, all the three nematodes multiplied but at varying rates. Their rate of population increase declined and proportion of males increased at higher inoculum levels. In concomitant inoculations, *M. incognita* and *R. reniformis* suppressed the rate of population increase of each other at all combinations. The decrease was greater in *M. incognita* than in *R. reniformis.* Mutual suppression in rate of population increase was noticed also when *R. reniformis* and *T. brassicae* or *M. incognita* and *T. brassicae* were present together. This was, however, not true at all combinations. In all concomitant inoculations, number of males of *M. incognita* and *R. reniformis* increased in comparison to single species inoculations at the same inoculum level. Sex-ratio of *T. brassicae,* however, remained unaltered. At all combinations, reductions in plant growth in concomitant inoculations were much less than the sum total of reductions caused by the same inoculum levels of nematodes when inoculated separately in single species inoculations.

RÉSUMÉ

Interactions de Meloidogyne incognita, Rotylenchulus reniformis *et* Tylenchorhynchus brassicae *sur la tomate*

Les interactions entre *Meloidogyne incognita, Rotylenchulus reniformis* et *Tylenchorhynchus brassicae* ont été étudiées sur racines de tomate en utilisant des inoculums de valeur variable et des combinaisons entre eux. Les paramètres mesurés ont été : taux de croissance de la population, quotient sexuel et croissance de la plante. Dans le cas d'inoculations monospécifiques, les trois nématodes se sont multipliés, mais à des taux variables. Lors de diverses inoculations combinées, *M. incognita* et *R. reniformis* inhibent réciproquement la croissance de leurs populations respectives. Cette diminution est plus accentuée pour *M. incognita* que pour *R. reniformis.* Ce phénomène d'inhibition a été également noté dans certaines des combinaisons entre *R. reniformis* et *T. brassicae* ou entre *M. incognita* et *T. brassicae.* Dans toutes les inoculations combinées, le nombre de mâles est plus élevé chez *M. incognita* et *R. reniformis,* en comparaison des inoculations monospécifiques à même taux d'inoculum. Par contre le quotient sexuel de *T. brassicae* demeure inchangé. Dans les cas d'inoculations combinées, la diminution de croissance de la plante était nettement plus faible que la somme des diminutions causées par des inoculations monospécifiques à taux d'inoculum équivalents.

In recent years a number of nematologists have worked on the coinhabiting nematode populations (Miller, 1970; Estores & Chen, 1972; Sikora *et al.,* 1972; Kheir & Osman, 1977). In some cases, the population of either decreased or increased (Miller, 1970; Estores & Chen, 1972). Estores and Chen (1972) reported that when *Meloidogyne incognita* and *Pratylenchus penetrans* were present together on tomato, the population development of the latter was reduced. Chapman and Turner (1975) also found that the presence of *M. incognita* resulted in reduction in penetration and fecundity of *P. penetrans* on red clover whereas the development of *M. incognita* remained unaffected. Higher reproduction rate of *Pratylenchus vulnus* on grape-vine suppressed the population of *Xiphinema index* (Pinochet, Raski & Goheen, 1976). Similarly, the population of *P. penetrans* was reduced when *P. penetrans* and *Tylenchorhynchus claytoni* were present together on tobacco roots (Mc Intyre & Miller, 1976). These studies provide evidences to support that as in other organisms, plant nematodes also have different kinds of ecological relationships and these factors have a deciding role in shaping the community structure. In the present studies, three nematodes viz., root-knot nematode, *Meloidogyne incognita* (Kofoid & White) Chitwood, a sedentary endoparasite; reniform nematode *Rotylenchulus reniformis* Linford & Oliveira; a sedentary semi-endoparasite and stunt nematode, *Tylenchorhynchus brassicae* Siddiqi, an ectoparasite were

selected in order to study their population dynamics in mixed population.

Materials and methods

Fifteen-day-old tomato (*Lycopersicon esculentum* Mill.) cv. Marglobe seedlings raised in sterilized soil were transplanted in 15 cm clay pots containing autoclaved sandy loam soil. The seedlings were later inoculated with the infective stages of root-knot nematode *(Meloidogyne incognita),* reniform nematode *(Rotylenchulus reniformis)* and stunt nematode *(Tylenchorhynchus brassicae)* in single species or concomitant inoculations according to the following scheme :

a. Root-knot nematode alone using 100, 500, 900 and 1 000 second stage juveniles.
b. Reniform nematode alone using 100, 500, 900 and 1 000 immature females.
c. Stunt nematode alone using 100, 500, 900 and 1 000 males, females and juveniles.
d. Root-knot and reniform nematodes.
e. Reniform and stunt nematodes.
f. Root-knot and stunt nematodes.
g. Root-knot, reniform and stunt nematodes.

In d, e, f and g different numbers of each nematode were added as given in Table 1. Uninoculated pots served as control. All treatments were replicated three times. After inoculations, pots of all treatments were kept in glasshouse (27-30°).

Sixty days after inoculation dry weights of the plants and the soil and root populations of nematodes (males, females and juveniles) were determined. Rate of population increase $= \frac{Pf - Pi}{Pi}$ and sex-ratio of the final population for all three nematode species were calculated. For calculating sex-ratio of all the three nematode species, the total population of mature/immature females and males was taken into account and figures were transformed to indicate male per one female. Reduction in plant weight in relation to control (uninoculated) was calculated for each treatment.

Results

Single species inoculations

With an increase in initial inoculum of *M. incognita, R. reniformis* and *T. brassicae* from 100 to 500 and from 500 to 900, there was a gradual decrease in the rate of population increase (RPI) of all the three nematode species. The RPI of *T. brassicae* was smaller than that for other two species (Tab. 1). In *M. incognita* and *R. reniformis,* number of males significantly increased as the inoculum level was increased. This tendency observed for *T. brassicae* was not significant (Tab. 2).

Root-knot and reniform nematodes caused a significant reduction in plant weight at all inoculum levels whereas stunt nematode reduced the plant weight significantly only at higher inoculum levels. In general, a significantly linear relationship was obtained between the initial inoculum level of *M. incognita, R. reniformis* and *T. brassicae* and reduction in plant weight in single species inoculations (Tab. 1).

Concomitant inoculations

The interactive effects of all the three nematodes in general were mutually inhibitory in concomitant inoculations. When *M. incognita* and *R. reniformis* coinhabited the roots, the rate of population increase (RPI) of *M. incognita* and *R. reniformis* significantly declined at all combinations when compared with the RPI at the same level of inoculum (Pi) in single species inoculation. The RPI of 100 *M. incognita* (Pi) in single species inoculation was 3.82. It was reduced to 0.97 in presence of 900 *R. reniformis* (Pi) in combined inoculation. Similar reductions were noticed in RPI of 500 *M. incognita* and 900 *M. incognita* in presence of 500 and 100 *R. reniformis* respectively. The RPI of *R. reniformis* at all levels of inoculum also declined in presence of *M. incognita.* However, the decline in RPI was comparatively lower than in *M. incognita* (Tab. 1). The proportion of males in both nematode species was significantly greater in each combination in comparison to proportion of males at the same inoculum level in single species inoculations (Tab. 2).

The plant weight at each combination was significantly reduced in comparison to control. Reduction in plant weight in simultaneous inoculations at each combination was comparatively less than the sum total of reductions caused by the same inoculum levels of the nematodes in single species inoculations. For example, the sum total of reductions in plant weight caused by 100 *M. incognita* and 900 *R. reniformis* in single species inoculations was 11.8 g but in concomitant inoculation with 100 *M. incognita* + 900 *R. reniformis,* the reduction was only 7.8 g. The same was true for other combinations as well (Tab. 1).

In concomitant inoculations of *R. reniformis* and *T. brassicae,* RPI of *R. reniformis* declined at all combinations except at 900 *R. reniformis* + 100 *T. brassicae,* when compared with the RPI of the same Pi of *R. reniformis* in single species inoculations. Similar reductions were noticed for *T. brassicae* as well (Tab. 1). Number of males of *R. reniformis* increased at 100 *R. reniformis* + 900 *T. brassicae* and 900 *R. reniformis* + 100 *T. brassicae.* No significant change in sex-ratio of *T. brassicae* occurred (Tab. 2).

The plant weight was significantly reduced at each combination in comparison to control. As in *M.*

Table 1

Interaction of *Meloidogyne incognita* (MI), *Rotylenchulus reniformis* (RR) and *Tylenchorhynchus brassicae* (TB) on tomato : rate of population increase and plant growth.

Treatments (Pi)			*Rate of population increase* $\frac{Pf - Pi}{Pi}$			*Dry weight of plant (g)*
MI	RR	TB	MI	RR	TB	
Single species inoculations						
100			3.82			8.6
500			2.85			3.8
900			1.38			4.1
1,000			2.50			2.9
	100			5.50		5.1
	500			2.55		3.8
	900			2.04		3.6
	1,000			2.76		3.6
		100			1.04	10.8
		500			1.39	9.8
		900			0.24	7.0
		1,000			0.70	8.0
Concomitant inoculations						
100	900		0.97*	0.56*		4.2
500	500		0.35*	1.60*		6.0
900	100		— 0.15*	1.75*		4.8
	100	900		5.02*	— 0.26*	4.3
	500	500		2.20*	— 0.35*	3.8
	900	100		1.97	— 0.08*	4.5
100		900	1.83*		0.26	8.4
500		500	1.71*		1.00*	7.7
900		100	1.00		0.83*	5.6
333	333	333	1.70	2.46	0.90	7.5
Control (Uninoculated)						12.0
LSD (0.01)			0.90	0.13	0.28	2.6
LSD (0.05)			0.66	0.09	0.20	1.9

* Figures significantly different from single species inoculations at the same inoculum level.
Regression lines between Pi and plant growth keeping plant weight as dependent variable :
For single species inoculations : Y = 9.2093-1.5265 X.
For simultaneous inoculations : Y = 6.69 — 0.62 X.
Correlation coefficient between Pi and plant weight :
For single species inoculations : — 0.5574.
For simultaneous inoculations : — 0.526.

incognita and *R. reniformis* simultaneous inoculations, reduction in plant weight in simultaneous inoculations of *R. reniformis* and *T. brassicae* at each combination was comparatively less than the sum total of reductions caused by the same inoculum levels of the nematodes in single species inoculations (Tab. 1).

When *M. incognita* and *T. brassicae* were inoculated simultaneously, the RPI of *M. incognita* significantly declined except at 900 *M. incognita* + 100 *T. brassicae*. Reduction in RPI of *T. brassicae* was also observed except at 900 *T. brassicae* + 100 *M. incognita* (Tab. 1). Alteration in sex-ratio of *M. incognita* only occurred but the effect of *T. brassicae* was smaller than those of *R. reniformis* (Tab. 2).

The plant weight was reduced at each combination in comparison to control. Similar to other combinations,

Table 2

Interaction of *Meloidogyne incognita* (MI), *Rotylenchulus reniformis* (RR) and *Tylenchorhynchus brassicae* (TB) on tomato : sex-ratio.

Treatments (Pi)			*Sex-ratio (male/one female)*		
MI	RR	TB	MI	RR	TB
Single species inoculations					
100			0.07		
500			0.22		
900			0.28		
1,000			0.30		
	100			0.43	
	500			0.58	
	900			0.61	
	1,000			0.68	
		100			0.14
		500			0.25
		900			0.27
		1,000			0.33
Concomitant inoculations					
100	900		1.00*	0.89*	
500	500		0.62*	0.99*	
900	100		0.45*	1.20*	
	100	900		0.48*	0.26
	500	500		0.58	0.26
	900	100		0.68*	0.18
100		900	0.68*		0.28
500		500	0.47*		0.24
900		100	0.40*		0.16
333	333	333	1.40	1.20	1.20
LSD (0.01)			0.06	0.07	0.33
LSD (0.05)			0.05	0.05	0.24

* Figures significantly different from single species inoculations at the same inoculum level.

reduction in plant weight in simultaneous inoculations of *M. incognita* and *T. brassicae* at each combination was much less than the sum total of reductions caused by the same inoculum levels of the nematodes in single species inoculations (Tab. 1).

In concomitant inoculation of the three nematodes, the RPI of *M. incognita, R. reniformis* and *T. brassicae* declined when compared with RPI of their nearest inoculum level i.e. Pi = 500 in single inoculation. This decrease in RPI of *R. reniformis* was, however, smaller and significant only at P = 0.05. But the RPI of *M. incognita* and *R. reniformis* in three species inoculations were greater than in their simultaneous inoculation. Similarly the RPI of *R. reniformis* and *T. brassicae* were greater than their RPI in their combined inoculations. However, the RPI of *M. incognita* and *T. brassicae* obtained in three species inoculations were not different from RPI obtained in their combined inoculations except of *M. incognita* at 900 *M. incognita* + 100 *T. brassicae* and of *T. brassicae* at 100 *M. incognita* + 900 *T. brassicae.* Sex-ratio of *M. incognita* and *R. reniformis* increased over their sex-ratios in single inoculations at 500 Pi or two species inoculations with 500 *M. incognita* + 500 *R. reniformis;* 500 *R. reniformis* + 500 *T. brassicae;* and 500 *M. incognita* + 500 *T. brassicae.* Sex-ratio of *T. brassicae* was not altered.

Plant weight was smaller than in control but greater than those obtained with 1 000 Pi of *M. incognita* or *R. reniformis* in single species inoculations.

Discussion

All the three nematodes multiplied on tomato. The rate of population increase, however, declined as the initial inoculum level increased. Species showed a

varying pattern of reproduction. *M. incognita* and *R. reniformis* reproduced more than *T. brassicae*. Tomato is a good host for *M. incognita* and *R. reniformis* and less favourable for *T. brassicae*. Availability of space to accommodate and support the growing population of the nematodes at higher inoculum levels seems to be responsible for decline in their rate of population increase. This effect was more pronounced for *R. reniformis* than for *M. incognita* and *T. brassicae*. The interactive effects in general, were inhibitory to each other in concomitant inoculations. *M. incognita* and *R. reniformis* mutually suppressed rate of population increase when they were present together. The effect was, however, more marked for *M. incognita*. It emerges, that in a community system like this, *R. reniformis* can survive, adapt and compete in a better way than *M. incognita*. Penetration zone for both nematodes is root tip (Linford, 1939; Birchfield, 1962). It is likely that over-crowding and clustering of infective units occurred during penetration at penetration zone which allowed fewer to penetrate. This reduced penetration influenced the rate of population increase. The mutual inhibitory effects appeared also due to competition for feeding sites. The feeding sites of both species in the root lie in close proximity (Birchfield, 1962; Sivakumar & Seshadri, 1972; Endo, 1975; Heald, 1975), which resulted in competition for food and space. This had reflected in population reduction. Production of more males is an adaptation in adverse environmental conditions (Bird, 1971). Females of both *M. incognita* and *R. reniformis* by virtue of their size require more food and space. Their crowding in plant roots resulted in overburdening the host which was further aggravated by its poor growth. Thus under adverse environmental conditions both of these nematodes demonstrated the masculinizing phase. Reduction in number of females caused population reductions of both the nematodes. Since *R. reniformis* requires lesser space in the host tissue than *M. incognita*, more female of *R. reniformis* could be accommodated in limited available space. This can be implicated as the reason for lesser reduction in population of *R. reniformis*.

R. reniformis and *T. brassicae* also mutually inhibited their rate of population increase when they coinhabited the roots. However, lower Pi of *T. brassicae* could not inhibit the RPI of *R. reniformis* when its Pi was high. Similarly low number of *M. incognita* or *T. brassicae* could not suppress the rate of population increase of each other when Pi of the other species was high. This can be presumed that low number of one species particularly of an ectoparasite like *T. brassicae* could not exert sufficient influences so as to inhibit the other species when its own density was high. The sex-ratio of *R. reniformis* was altered only when either its own Pi was high and that of *T. brassicae* was low or vice-versa. Similar pattern of increase in number of males of *M. incognita* was observed in its concomitant inoculations with *T. brassicae*. Sex-ratio of *T. brassicae* was not affected. These variations can be attributed to the difference in their mode of parasitism. It indicates that initial density of coinhabitants, their mode of parasitism, availability of space and nutrition for the growing individuals are among the vital factors determining the course of population growth and community structure in a particular plant nematode pathosystem.

In single species inoculations of the nematodes, a significantly linear relationship between the initial inoculum and plant dry weight was obtained. Reductions in plant weight in concomitant inoculations were relatively less than the sum total of reductions caused by the same inoculum levels of the nematodes in single species inoculations. Apparently their mutual inhibitory interactive effects that caused decline in their rate of population increase resulted in reduction of their damaging potentials.

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