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Department of Plant Pathology
University of Agricultural Sciences
Hebbal, Bangalore 560 024, India

PENETRATION, DEVELOPMENT AND HISTOPATHOLOGY OF ROOT-KNOT NEMATODES IN TOMATO « NTDR-1 » ([1])

by

Y. D. NARAYANA and D. D. R. REDDY

A programme of tomato breeding for resistance to the root-knot nematode resulted in the variety NTDR-1 (Shivashankar *et al.*, 1975). The variety is highly resistant to *Meloidogyne incognita* (Kofoid *et* White) Chitw. and *M. javanica* (Treub) Chitw. This paper reports investigations on some aspects of the interaction between these nematode species and susceptible and resistant tomato varieties.

MATERIALS AND METHODS

The nematode-resistant NTDR-1 and the susceptible variety Pusa Ruby were used in the investigations. Three week old seedlings were transplanted singly into 5 cm plastic cups containing steam sterilized soil and after 4 days each cup was inoculated with a suspension containing 50 larvae of either *M. incognita* or *M. javanica.* Seven days after inoculation some of the seedlings were removed from the cups and the roots stained in acidfuchsin lactophenol for determination of larvae in the roots and galls.

To study development of the nematodes, 25 day old seedlings in pots of sterilized soil were inoculated with second stage larvae of either *M. incognita* or *M. javanica.* The seedlings were removed from the pots after 24 hours, the roots washed thoroughly to remove

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any larvae that had not penetrated, and transplanted individually into 10 cm pots. Five seedlings for each tomato variety and for each nematode species were removed at intervals of 1, 2, 4, 6, 8, 10, 13, 15, 17, 19, 22, 25, 28, 32 and 40 days after inoculation. The roots were washed and stained in acidfuchsin lactophenol, the larvae were dissected out from the roots and the length and width of five larvae per root system measured to determine their development according to the figures published by Triantaphyllou and Hirschmann (1960).

Histopathological comparisons of *M. javanica* infested roots of resistant and susceptible varieties were made 30 days after inoculation. Root-knot galls were separated from the roots and fixed in absolute alcohol, acetic acid and chloroform (6:1:3) for 1 hr. The procedures of Sass (1958) were followed for microtomy sectioning. The number and size of giant cells and the number of nuclei in each giant cell were recorded.

RESULTS

Galls were not observed in the roots of the resistant variety NTDR-1 inoculated with *M. incognita* but two small galls were found in the seedling infested with *M. javanica.* In the susceptible variety Pusa Ruby both *Meloidogyne* species produced similar number of galls (Table I).

Table I - *Number of* Meloidogyne incognita *and* M. javanica *larvae penetrated into the roots of resistant and susceptible varieties of tomato and number of galls formed on the root systems.*

Nematodes species	Variety	Mean number of larvae penetrated	Mean number of galls formed
M. incognita	Pusa Ruby (Susceptible)	41.0	12.1
	NTDR-1 (Resistant)	10.8	0.0
M. javanica	Pusa Ruby (Susceptible)	42.3	15.3
	NTDR-1 (Resistant)	26.3	2.0
C.D. at 5%		10.8	

Fig. 1 summarizes the rate of penetration of larvae of *M. javanica* or *M. incognita* in the resistant and susceptible tomato varieties. Progressive increase in the rate of penetration was observed even upto the 7th day after inoculation in both the resistant and susceptible varieties. The rate of invasions was significantly lower in NTDR-1 than in Pusa Ruby. However, more *M. javanica* larvae penetrated the roots of NTDR-1 than did *M. incognita* (Fig. 1, Table I).

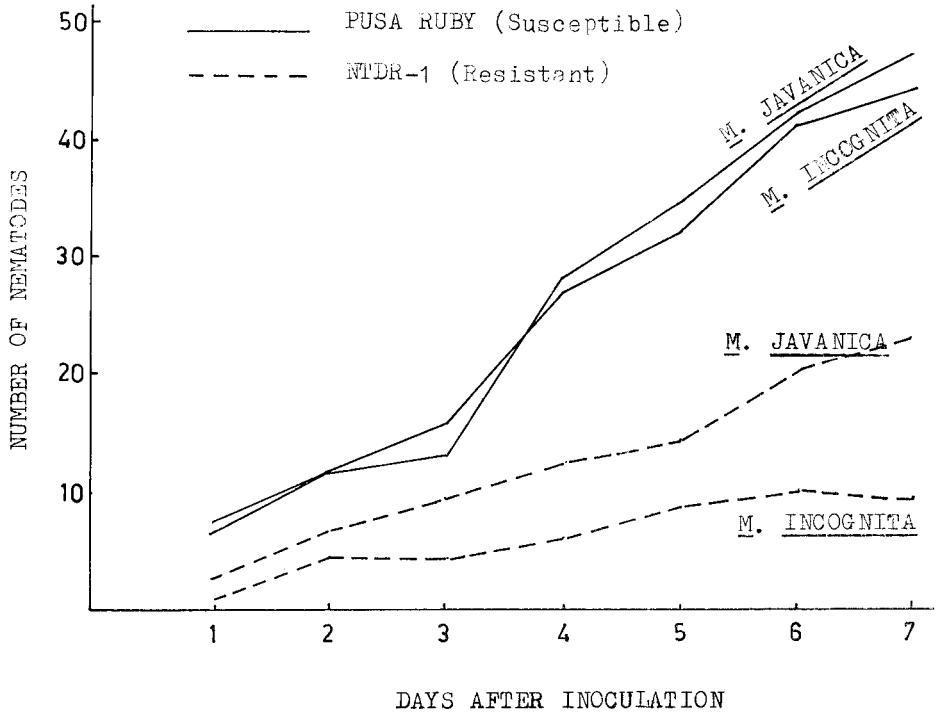

Fig. 1 - Number of *M. incognita* and *M. javanica* observed within the roots of resistant and susceptible varieties of tomato.

M. incognita and M. *javanica* completed their life cycle in « Pusa Ruby » in 32 days and egg masses were observed after the 32nd day following inoculation. Second, 3rd and 4th moults and fully developed adult females were observed after 10-12, 13-14, 15-16 and 28 days respectively. The development of *M. incognita* and *M. javanica* was quite different in the resistant variety. Even though there was slight increase in the size of *M. incognita* larvae after entering into

the roots, no further development of larvae was observed after the second stage (Table II). The penetrated larvae started to die from the 4th day onwards. In the case of *M. javanica* even though more larvae entered the roots they remained undeveloped at second stage with only a few of them developing to the 3rd stage. However in these the 2nd moult was delayed up to the 17th day and none of them developed further.

Table II - *Development of* M. incognita *and* M. javanica *in the roots of resistant and susceptible varieties of tomato.*

Days following inoculation	M. incognita				M. javanica			
	Pusa Ruby (Susceptible)		NTDR-1 (Resistant)		Pusa Ruby (Susceptible)		NTDR-1 (Resistant)	
	Lenght (µm)	width (µm)	Lenght (µm)	width (µm)	Lenght (µm)	width (µm)	Lenght (µm)	width (µm)
1	320	17	270	15	331	17	279	15
2	370	15	290	15	329	17	294	22
4	311	17	323	15	373	20	288	22
6	408	24	325	17	416	26	287	21
8	407	42	383	17	385	45	250	19
10	413	66	358	15	442	61	294	22
13	409	51	333	17	457	42	385	19
15	418	77	364	17	473	69	223	22
17	426	102	391	19	475	94	367	22
19	431	103	405	17	505	96	403	25
22	462	109	272	20	524	157	455	25
25	496	120	462	17	524	168	610	30
28	559	313	362	20	605	347	461	28
32	604	350	359	20	699	334	610	32
40	724	527	485	17	807	485	521	40

The number of giant cells induced by *M. javanica* in the resistant variety was low and of smaller size (64 x 42 µm) compared to those formed in the susceptible one (119 x 80µm) (Table III). Similarly there were fewer nuclei per giant cell in the resistant than in the susceptible variety. Giant cells of the susceptible variety had enlarged nuclei and contained a dense cytoplasm. No cell malformation or giant cell development was observed in « NTDR-1 » infected with *M. incognita.*

Table III - *Number and size of the giant cells around the nematode and number of nuclei per giant cell in resistant and susceptible varieties of tomato 30 days after invasion of* Meloidogyne javanica.

Variety	Mean number of giant cells	Size of giant cells		Area infested (μm^2)	Mean number of nuclei
		Lenght (μm)	width (μm)		
Pusa Ruby	5	116	61	7076	8
(Susceptible)	4	113	72	8136	10
	6	106	62	6572	11
	7	124	89	11036	12
	6	122	95	11590	12
	7	122	85	10370	10
	6	131	99	13969	9
Mean	5.8	119	80	9821	10
NTDR-1	4	77	52	4004	2.5
(Resistant)	3	57	36	2052	3.0
	3	68	47	3196	2.5
	4	71	53	3763	2.3
	3	56	36	2016	3.0
	3	62	41	2542	2.5
	2	56	30	1680	2.0
Mean	3.1	64	42	2750	2.5
C.D. at 5%	1.5	41	20		1.5

DISCUSSION

Resistance to root-knot nematodes in « NTDR-1 » tomato apparently is associated with low invasion and hindrance to larval development beyond the 2nd or the 3rd stage as observed by Fassuliotis (1970) for *M. incognita acrita* in the roots of resistant cantaloup. In Wallace's (1973) opinion the nematode is unable to obtain nutrients from the host because the host response in such that the nematode does no have access to the biochemical machinery of the plant. Kurian (1970) speculated that the death of the 2nd stage larvae in the resistant *Nicotiana rependa* could be due to a hypersensitive reaction by the host plant and failure to form giant cells. Similar reasons could be attributed to the death of 2nd stage larvae in the

resistant NTDR-1 in the present investigations. However there is no substantial evidence that the fate of nematode within the host is influenced by toxic substances released during the interaction between the nematode and plant. The partial development of some *M. javanica* larvae, mostly up to 3rd stage, in the resistant variety is probably due to the formation of smaller giant cells with fewer cells (Fassuliotis, 1970) or to the reacting substances present in the host which are not specific to the nematode (Riggs and Winstead, 1959).

SUMMARY

The nature of resistance in tomato variety NTDR-1 to the root-knot nematodes *Meloidogyne incognita* (Kofoid *et* White) Chitw. and *M. javanica* (Treub) Chitw. was studied in comparison with susceptible 'Pusa Ruby'. The rate of penetration of the nematode larvae was comparatively lower in the roots of the resistant NTDR-1. Resistance in NTDR-1 was associated with hindrance to larval development beyond the 2nd stage and death of the nematodes after invasion. Comparisons of the histopathology of roots of NTDR-1 and Pusa Ruby infested with *M. javanica* showed differences in the development of the giant cells.

RIASSUNTO

Penetrazione, sviluppo e istopatogenesi di nematodi galligeni in radici di piante di pomodoro della varietà NTDR-1.

È stata studiata la natura della resistenza nei confronti di nematodi galligeni, *Meloidogyne incognita* (Kofoid *et* White) Chitw. e *M. javanica* (Treub) Chitw., nella varietà di Pomodoro NTDR-1. Il numero di larve di seconda età, delle due specie, penetrato nelle radici delle piante resistenti è stato inferiore che non nella varietà suscettibile Pusa Ruby. La natura della resistenza nella varietà oggetto di studio sembra essere dovuta al fatto che le larve, dopo la loro penetrazione nelle radici, non continuano lo sviluppo e muoiono. Nelle radici della varietà resistente, inoltre, si formano solo poche cellule giganti di dimensioni ridotte rispetto a quelle presenti nei tessuti radicali della varietà suscettibile.

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