Biological Forum
an International Journal 17(4): 28-32(2025)
ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Pathogenicity of Root-Knot Nematode, *Meloidogyne incognita* Infecting Capsicum under Protected Cultivation

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(Received: 18 January 2025; Revised: 25 February 2025; Accepted: 13 March 2025; Published online: 07 April 2025)
(Published by Research Trend)

ABSTRACT: The pathogenicity of root-knot nematode, *Meloidogyne incognita* was studied on capsicum in pots under protected cultivation during 2022-23 and 2023-24. Five inoculum levels (0, 10, 100, 1000 and 10,000) were tested to assess their impact on plant growth and nematode reproduction. Results showed that nematode population in root, soil and total nematode population increased proportionally with higher inoculum levels. A significant reduction in plant height and fresh shoot weight was observed even at the lowest inoculum level of 10 J_2 per plant, which was identified as the critical damaging threshold. Conversely, nematode reproduction rate declined as inoculum levels increased with the highest reproduction rate recorded at 10 J_2/plant and lowest at 10,000 J_2/plant. These findings highlight the severity of *M. incognita* infection in capsicum and emphasized the need for effective nematode management strategies under protected cultivation.

Keywords: Capsicum, pathogenicity, protected cultivation, root-knot nematode.

INTRODUCTION

Capsicum (*Capsicum annuum* L.) is commonly known as green pepper, sweet pepper, bell pepper and belongs to the family *Solanaceae*. This crop is successfully grown under the protected cultivation, an emerging technology for raising high value crops in tropical and sub-tropical regions. Among the various biotic stresses, plant parasitic nematodes, particularly root-knot nematodes (*Meloidogyne* spp.) pose a significant threat to capsicum production (Tiyagi *et al*., 2012). Root-knot nematodes, *Meloidogyne* spp. are considered among the most important PPNs worldwide, with *M .incognita*, *M. javanica*, *M. arenaria* and *M. hapla* being the most damaging species (Greco & Vito 2009). The extent of damage and crop loss, depends on the initial nematode population density at planting making it crucial to establish threshold for effective management (Jain, 1992). Understanding the pathogenicity of *Meloidogyne* spp. under controlled pot condition provides essential insights into the biology and virulence of these nematodes. This study is critical in assessing the relationship between nematode population density and the extent of crop damage, which is pivotal for formulating threshold levels for management interventions. In capsicum, the development of root galls directly correlates with nematode inoculum levels and even moderate infestations can lead to severe yield loss (Majdi *et al*., 2019). Therefore, present investigation was carried out.

MATERIALS AND METHODS

An experiment was conducted to study the pathogenicity of *M. incognita* on capsicum var. *Indra* transplanted in 15 cm diameter earthen pots of using Completely Randomized Design (CRD) with five repetitions at Department of Nematology, B.A. College of Agriculture, Anand Agricultural University, Anand, Gujarat. The inoculum levels were tested 0, 10, 100, 1000 and 10,000. The uninoculated plants (without nematode inoculation) served as control. Earthen pots were thoroughly washed with water and disinfected using 4% formaldehyde (Formalin 40 EC) solution. After drying, pots were filled with 1.5 kg of steam sterilized soil. One seedling of capsicum *cv. Indra* was transplanted in the centre of each pot. Second stage juvenile (J_2) were extracted from the egg masses of *M. incognita*. After establishment of seedlings, J_2 were inoculated in the rhizosphere by carefully removing soil around stem using forcep. The inoculum levels were 0, 10, 100, 1000 and 10,000 J_2 per plant and uninoculated plants served as control. Each treatment was repeated five times. Plants were watered as needed and protected from insects-pests and diseases using standard recommended practices. Sixty days after inoculation (DAI), plants were depotted carefully. Plants and roots were thoroughly washed with water to make free from the soil particles. Observations were recorded on plant height, fresh shoot and root weight, root-knot index [(0-5 scale) (Taylor & Sasser 1978)], nematode population in soil (200 cc soil), nematode population in 5g roots (Number of females, number of egg masses and number of eggs).

0-5 Scale (Taylor & Sasser 1978)	
Root-Knot Index (RKI)	**Reaction**
0	Highly resistant
0.01-1.0	Resistant
1.01-2.0	Moderately resistant
2.01-3.0	Moderately susceptible
3.01-4.0	Susceptible
4.01-5.0	Highly susceptible

RESULTS AND DISCUSSION

An experiment was conducted to study the effect of *M. incognita* on plant growth parameters and nematode reproduction on capsicum *cv. Indra.* Significant differences were observed for all the plant growth parameters at different levels of nematode inoculum *viz*., plant height, fresh shoot and root weight, root-knot index and nematode population in soil and root build up per plant due to different levels of nematode inoculum. An increase in inoculum level of nematodes progressively decreased plant height and fresh shoot weight of plants with a corresponding increase in root-knot index as well as soil and root nematode population per plant (Table 1-3).

Plant height (cm). The data presented in Table 1 revealed significant variations among the treatments during 2022-23, 2023-24 and the pooled analysis across both years for plant height. A significant effect of the year was observed, while the Treatment × Year (T × Y) interaction was found to be non-significant. The findings indicated a gradual reduction in plant height as the inoculum levels increased.

In 2022-23, a significant reduction in capsicum plant height (63.80) was observed at an initial inoculum level of 10 J_2 per plant. The inoculum level 100 J_2 per plant recorded plant height (58.60) which was statistically at par with 1000 J_2 per plant (55.20). The lowest plant height (42.20) was recorded at an inoculum level of 10,000 J_2 per plant. In contrast, the uninoculated control plants exhibited the highest plant height (70.00), which was significantly different from all other inoculum levels.

During 2023-24, significant variations were recorded among the treatments. The uninoculated control plants again recorded the highest plant height (73.60), while the lowest plant height (36.00) was associated with the highest inoculum level of 10,000 J_2 per plant.

A similar pattern was observed in the pooled data over both years, with the maximum plant height recorded in the uninoculated (control) plants.

Fresh shoot weight (g). The data presented in Table 1 revealed significant differences among treatments for the fresh shoot weight of capsicum during 2022-23, 2023-24, and in the pooled analysis. The interaction effect between year and treatment was found to be significant.

During 2022-23, the highest shoot weight (72.00) was observed in the uninoculated plants, while the lowest fresh shoot weight (45.60) was recorded at an inoculum level of 10,000 J_2/plant. The inoculum level 100 J_2/plant and 1000 J_2/plant were significantly at par with each other. An initial inoculum level of 10 J_2 of *M. incognita* per plant caused a significant reduction in fresh shoot weight compared to other inoculum levels across 2022-23, 2023-24 and the pooled years.

Fresh root weight (g). The data on fresh root weight, as presented in Table 1, highlight the results for 2022-23, 2023-24 and the pooled analysis. A significant reduction in fresh root weight was observed starting from an inoculum level of 10 J_2/plant and above, compared to the control.

During 2022-23, the highest fresh root weight (18.80) was recorded at the inoculum level of 10,000 J_2/plant, which was significantly different from all other inoculum levels. Conversely, the lowest fresh root weight (9.60) was observed in the control plants, which was statistically at par with the inoculum level of 10 J_2/plant. More or less similar trend was observed in 2023-24 and in the pooled analysis.

Root-knot index. The data on root-knot index (RKI), as presented in Table 1, revealed significant differences among all treatments, with the RKI increasing as the inoculum levels increased. Significant variation was observed across treatments during 2022-23, 2023-24 and in the pooled analysis, though the T × Y interaction effect was non-significant.

In 2022-23, significantly maximum RKI (5.00) was recorded at an inoculum level of 10,000 J_2 per plant, which differed significantly from all other treatments. The lowest RKI was observed in plants inoculated with 10 J_2 per plant.

In 2023-24, the control (uninoculated) plants exhibited the lowest RKI, which was significantly different from all other inoculum levels. The inoculum level of 10,000 J_2 per plant resulted in the highest RKI. Inoculum levels of 10 and 100 J_2 per plant showed an intermediate effect on the RKI. Similar trends observed in pooled over years.

Nematode population in root. The reproduction of *M. incognita* showed an increase in the number of females, egg masses and eggs as the inoculum levels increased (Table 2). These attributes were observed to be highest at the inoculum level of 10,000 J_2 per plant. Significant differences were noted among the treatments, with all initial inoculum levels differing significantly from one another during 2022-23, 2023-24 and in the pooled analysis.

In 2022-23, the highest number of females (324) was recorded at the inoculum level of 10,000 J_2 per plant, followed by the levels of 1000, 100, and 10 J_2 per plant. As anticipated, no females were observed in the control plants since they were not inoculated with nematodes. Similarly, in 2023-24 and the pooled analysis, significant differences were observed among treatments for the number of females. While the year effect was significant, the TxY interaction was non-significant.

An increase in the initial inoculum level led to a significant rise in the number of egg masses per 5 g of roots during 2022-23, 2023-24 and in the pooled analysis.

The highest number of eggs was recorded at the inoculum level of 10,000 J_2 per plant. Furthermore, the treatments with inoculum levels of 10, 100 and 1000 J_2 per plant differed significantly from each other during 2022-23, 2023-24 and pooled over years.

Nematode population in soil (J_2/ 200 cc soil). Regarding the soil nematode population, there was a corresponding increase with higher levels of nematode inoculum. The highest soil nematode population (18,208) was recorded at the inoculum level of 10,000 J_2/plant, while the lowest population was observed at the 10 J_2 /plant (158), followed by the 100 J_2/plant (993) and 1000 J_2/plant (2598) in 2022-23 (Table 3).

The trend observed in the soil nematode population closely followed that of the number of females, egg masses and eggs per 5 g roots for the different treatments during 2023-23 and pooled analysis of both the years. The year effect was significant, while the T × Y interaction was non-significant.

Total nematode population. The data on the total nematode population (Table 3) showed a pattern similar to that observed for the soil population, with the highest reproduction rate at the initial inoculum level of 10 J_2/plant, followed by 100, 1000 and 10,000 J_2/plant. The total nematode population/plant increased with higher inoculum levels. Control plants, which were not inoculated with nematodes, naturally recorded no nematodes during 2022-23, 2023-24 and the pooled analysis. The year effect was significant, while the T × Y interaction was non-significant.

Reproduction rate (P_f/P_i). Similarly, the nematode reproduction rate decreased as the inoculum levels increased. This could be attributed to competition among the nematodes for the limited root system available at the time of inoculation, preventing some nematodes from successfully infecting the roots. As a result, the reproduction rate increased as the nematode inoculum levels decreased. The highest reproduction rate (426.6 times) was observed at the 10 J_2 per plant level, while the lowest rate (8.31 times) was recorded at the highest inoculum level of 10,000 J_2 per plant. This fluctuation may be due to the prolonged period of low temperatures, which could have been more favorable for capsicum growth but less conducive to nematode multiplication (Table 3).

The results from this study indicate that an initial inoculum level of 10 J_2 of *M. incognita* per plant caused significant damage to the capsicum cv. *Indra*, leading to a reduction in plant growth parameters such as plant height and fresh shoot weight.

These findings align with the work of Agaba *et al*. (2015), who reported a direct correlation between inoculum levels and both the galling index and final nematode population, while an inverse relationship was observed with the reproductive factor in pepper. Similarly, Duggal *et al.* (2017) found significant reductions in plant growth parameters in capsicum at inoculum levels of 1000 J_2 and above. Furthermore, studies by Venkatesan *et al.* (2009); Gupta *et al*. (1995); Singh and Patel (2012) who reported a significant decline in plant growth attributes and proportional increase in RKI with an increasing inoculum levels of *M. javanica pt.*1. However, their findings suggested that the damaging threshold levels were 100 J_2/plant cucumber. Holajjer *et al.* (2022) reported that *M. incognita* was harmful to dragon fruit, as it caused maximum number of galls/egg masses and high soil nematode population observed at initial population density of 500 J_2/200 cm^3.

Table 1: Pathogenicity of root-knot nematode (*M. incognita*) on capsicum: Effect on plant height, fresh shoot and root weight and root-knot index during 2022-23 and 2023-24.

Inoculum level (J_2/pot)		Plant height (cm) at 60 DAI			Fresh weight (g) at 60 DAI						Root-knot index (0-5 scale) at 60 DAI		
					Shoot			Root					
		2022-23	2023-24	Pooled	2022-23	2023-24	Pooled	2022-23	2023-24	Pooled	2022-23	2023-24	Pooled
0		70.00	73.60	71.80	72.00	70.40	71.20	9.60	10.40	10.00	0.00 (0.5)	0.00 (0.5)	0.00 (0.5)
10		63.80	65.80	64.80	63.60	61.80	62.70	11.40	12.80	12.10	1.22 (1.50)	0.99 (0.98)	1.11 (1.23)
100		58.60	52.60	59.80	56.80	55.20	56.00	15.40	13.60	14.50	1.44 (2.07)	1.30 (1.69)	2.09 (4.37)
1000		55.20	44.20	56.30	52.80	49.80	51.30	17.40	17.20	17.30	2.12 (4.48)	2.08 (4.33)	2.10 (4.41)
10,000		42.20	36.00	43.70	45.60	39.60	42.60	18.80	18.60	18.70	2.35 (5.00)	2.35 (5.00)	2.35 (5.00)
S. Em.±	T	1.41	1.74	1.21	1.91	1.51	1.32	1.08	0.63	0.69	0.04	0.07	0.04
	Y	-	-	0.76	-	-	0.77	-	-	0.40	-	-	0.03
	T × Y	-	-	1.71	-	-	1.72	-	-	0.89	-	-	0.06
CD at 5 %	T	4.17	5.13	2.87	5.64	4.44	3.80	3.20	1.85	1.98	0.14	0.20	0.10
	Y	-	-	Sig.	-	-	Sig.	-	-	Sig.	-	-	Sig.
	T × Y	-	-	NS	-	-	NS	-	-	NS	-	-	NS
CV (%)		5.45	7.14	6.45	7.36	6.08	6.78	16.70	9.64	13.64	6.09	9.41	7.87

Note: 1. 0 = Free; 5 = Maximum disease intensity, DAI= Days after inoculation, NS= Non-significant, S= significant

2. Figures in parentheses are re-transformed values of $(\sqrt{x + 0.5})$

Table 2: Pathogenicity of root-knot nematode (*M. incognita*) on capsicum: Effect on number of females, egg masses and eggs/egg mass during 2022-23 and 2023-24.

Inoculum level (J_2/pot)		Nematode population/5 g roots								
		Number of females			Number of egg masses			Number of eggs/egg mass		
		2022-23	2023-24	Pooled	2022-23	2023-24	Pooled	2022-23	2023-24	Pooled
0		0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)
10		1.34 (22)	1.40 (25)	1.37 (23)	1.21 (16)	1.19 (15)	1.20 (16)	3.66 (4595)	3.64 (4339)	3.65 (4467)
100		1.84 (70)	1.81 (65)	1.83 (68)	1.65 (45)	1.72 (53)	1.69 (49)	4.15 (14140)	4.16 (14499)	4.16 (14454)
1000		2.21 (163)	2.17 (147)	2.19 (155)	1.74 (55)	1.85 (71)	1.80 (63)	4.49 (30682)	4.50 (31936)	4.50 (31623)
10,000		2.51 (324)	2.45 (284)	2.48 (302)	2.04 (109)	2.10 (125)	2.07 (117)	4.83 (67604)	4.84 (68663)	4.83 (67608)
S. Em.±	T	0.04	0.03	0.03	0.04	0.03	0.03	0.08	0.06	0.05
	Y	-	-	0.01	-	-	0.02	-	-	0.02
	T × Y	-	-	0.03	-	-	0.03	-	-	0.03
CD at 5 %	T	0.10	0.09	0.07	0.12	0.08	0.08	0.23	0.19	0.14
	Y	-	-	Sig.	-	-	Sig.	-	-	Sig.
	T × Y	-	-	NS	-	-	NS	-	-	NS
CV (%)		7.87	4.22	4.60	6.86	4.34	5.70	5.03	4.10	4.59

Note: 1. NS= Non-significant, S= significant
2. Figures in parentheses are re-transformed values of log (X+1)

Table 3: Pathogenicity of root-knot nematode (*M. incognita*) on capsicum: Effect on soil, total population and reproduction rate during 2022-23 and 2023-24.

Inoculum level (J_2/pot)		Soil (200 cc) at 60 DAI			Total nematode population (soil + root)			Reproduction rate (P_f/P_i)
		2022-23	2023-24	Pooled	2022-23	2023-24	Pooled	
0		0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00
10		2.20 (158)	2.34 (216)	2.27 (186)	3.69 (4878)	3.58 (3777)	3.63 (4266)	426.6
100		3.00 (993)	3.01 (1023)	3.01 (1020)	4.18 (15285)	4.06 (11578)	4.12 (13183)	131.83
1000		3.41 (2598)	3.45 (2818)	3.43 (2692)	4.53 (33678)	4.46 (28799)	4.49 (30903)	30.90
10,000		4.26 (18208)	4.36 (22909)	4.31 (20417)	4.94 (86469)	4.89 (78425)	4.92 (83176)	8.31
S. Em.±	T	0.03	0.03	0.02	0.07	0.03	0.04	-
	Y	-	-	0.01	-	-	0.03	-
	T × Y	-	-	0.03	-	-	0.05	-
CD at 5 %	T	0.09	0.08	0.05	0.21	0.10	0.09	-
	Y	-	-	Sig.	-	-	Sig.	-
	T × Y	-	-	NS		-	NS	-
CV (%)		2.47	2.48	2.48	4.52	2.29	3.60	-

Note: 1. DAI= Days After Inoculation
2. NS= Non significant, Sig. = Significant
3. Figures in parentheses are re-transformed values of log (X+1)

CONCLUSIONS

Pathogenicity trials were conducted to determine the threshold or damaging inoculum level of root-knot nematode, *Meloidogyne incognita* in capsicum. The results revealed that an inoculum level of 10 J_2/plant was the critical damaging level for *M. incognita* in capsicum. Nematode infection significantly impacted plant growth parameters, including plant height and fresh shoot weight. Additionally, nematode multiplication *i.e.* root-knot index and final nematode population/plant indicated an increased reproduction rate with decreased inoculum levels.

FUTURE SCOPE

The study establishes the damaging threshold of *Meloidogyne incognita* on capsicum and underscores its impact under protected cultivation. Future research should focus on developing integrated nematode management strategies, screening resistant capsicum varieties and understanding host–pathogen interactions at molecular levels. Evaluating novel control technologies, studying the influence of environmental factors and validating findings under field conditions will further strengthen nematode management approaches for sustainable capsicum production.

Acknowledgement. I convey the depth of my feeling and gratitude to my major advisor Dr. R.K. Thumar and to my advisory committee members Dr. Ajay Kumar Maru and other for giving me proper guidance throughout the course of study.
Conflict of Interest. None.

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How to cite this article: Anjana B. Prajapati, R.K. Thumar, Ajay Kumar Maru and Mayuri Y. Patel (2025). Pathogenicity of Root-Knot Nematode, *Meloidogyne incognita* Infecting Capsicum under Protected Cultivation. *Biological Forum, 17*(4): 28-32.