



Assessing *Meloidogyne Incognita* Population Dynamics In Tomato: Comparative Study of Open Field and Polyhouse Conditions

Anupam Sekhon, Narpinderjeet Kaur Dhillon, Sukhjeet Kaur¹
and Harwinder Singh Buttar²

Department of Plant Pathology, ¹Department of Vegetable Science
Punjab Agricultural University, Ludhiana-141 004, India

²Punjab Agricultural University-Krishi Vigyan Kendra, Nurmahal, Jalandhar-144 039, India
E-mail: anupam-pp@pau.edu

Abstract: Root knot nematode is an economically important pathogen affecting the crop in both the cultivation systems *i.e.* open and protected. The present research revealed that the reproduction of *M. incognita* population was comparatively higher in polyhouse as compared to open cultivation of tomato. Low inoculum levels reached to pathogenic levels due to higher multiplication of root knot nematode in polyhouses as compared to open cultivation system. There is substantial impact on buildup of nematode populations under prevailing environmental conditions. The infestation of *M. incognita* affected the growth parameters and health of the crop and the effects were more at higher inoculum levels in both the cultivation systems. Comparatively, due to higher multiplications, the infestation of root knot nematode may have more severe impact on the same crop grown in polyhouse.

Keywords: Agro-ecosystem, Inoculum levels, *Meloidogyne incognita*, Tomato

Plant-parasitic nematodes cause yield losses of \$173 billion annually worldwide (Pires et al., 2022). Root-knot nematode, *Meloidogyne incognita* is one of the major limiting factors in production of tomato crop (Qiao et al., 2012, Onkendi et al., 2014, Abrar et al., 2020). Yield losses of 22-30% have been reported on tomato due to *M. incognita* (Ahmed et al., 2023). However, the losses caused by this nematode may vary according to the species. In India on an average, a national loss of tomato Rs 6035.2 million has been estimated due to plant parasitic nematodes (Kumar et al., 2018). Out of 100 species of *Meloidogyne* which were present throughout the world, four species *viz.* *Meloidogyne incognita*, *M. javanica*, *M. arenaria* and *M. hapla* are commonly found in India (Khan et al., 2023). Among these four species, *M. incognita* is predominantly associated with tomato cultivation in India.

Tomato is grown worldwide in diverse agro-ecosystems in open fields as well as under protected structures /polyhouses. On the other hand, in protected cultivation, there are polyhouses which are framed structures covered with transparent or translucent material and are large enough to grow crops under partial or full controlled environmental conditions to obtain optimum growth and yield. This partial control of microclimatic conditions may have differential influence on prevalence, abundance or buildup of pathogens. The present studies have therefore been undertaken to know the buildup of root knot nematode at different inoculum levels in open and polyhouse cultivation of tomato.

MATERIAL AND METHODS

Experimental design: The experiment was conducted in two sets of environment from October to December (winter season crop) in open and protected cultivation for consecutive two years (2017 and 2018). The buildup of root knot nematode was estimated at different inoculum levels *i.e.* 100, 500, 1000, 2000, 4000 and 8000 juveniles/pot. The study showed no significant difference, so the two sets of data were combined for final analysis. In open field condition/natural conditions at Punjab Agricultural University, Ludhiana, 5x5 sq.m area was marked and covered with plastic sheet on which pots were kept. In second agro-ecosystem *i.e.* protected cultivation, in a polyhouse area was 250 sq.m. at Department of Plant Pathology, Ludhiana and area of 5x5 sq.m. was covered with plastic sheet on which pots were kept. In open cultivation, temperature from October to December ranged from 22.2°C to 32.7°C (2016-17) and 20.9°C to 33.3°C (2017-18) while the range of temperature was 27.1°C to 37.3°C (2016-17) and 25.1°C to 38.9°C (2017-18) in polyhouse. Pots were filled with sterilized soil and then ten seeds of tomato var. Punjab Ratta were sown in pots containing loamy sand soil (Sand-75.5%, Silt-19.3%, Clay-5.2%, ph-7.6, EC-0.24 dS m⁻¹, Organic matter-0.672%). At two leaf stage, the plants were inoculated with root knot nematode juveniles collected from pure culture obtained from single egg mass technique given by Zakaria (2013). In single egg mass technique, galled roots of tomato were carefully washed using gentle flow of water to remove the adhering soil particles. One egg mass was collected by

the aid of a needle specially adapted for this technique. The obtained culture was reared on tomato seedlings planted in pots filled with sterilized soil. Pots were kept under glasshouse conditions for 45–60 days to maintain the nematode inoculum for further studies. The egg masses were collected from these plants and root knot nematode female was identified by morphological characteristics and perennial patterns of females. *M. incognita* females illustrated the presence of a high, squarish dorsal arch, which contains a distinct whorl in the tail terminal area. The striae are smooth to wavy. Distinct lateral lines are absent, but breaks and forks in striae are obvious.

Six inoculum levels were used *i.e.* 100, 500, 1000, 2000, 4000 and 8000 juveniles. These juveniles were concentrated in 5ml of water and added to soil by making 4-5 holes around the stem. The un-inoculated plants served as control. Each treatment was replicated four times in completely randomized factorial design..

Data collection: Observations on soil nematode population and growth parameters were taken. The nematode infestation in roots was assessed on the basis of number of number of galls per root system and was referred to as root galling index (GI). The egg mass count was taken by manually counting the egg masses on 3g of root.

Plant growth parameters: The plants were uprooted after sixty days of inoculation. The roots were washed carefully with water to remove all the debris. The length and fresh weight of shoot and root were measured using centimeter scale.

Nematode population in soil as well as roots: Galling was scored on scale of 0-5 rating chart by Taylor and Sasser (1978) where 0 = no galls; 1 = 1–2; 2 = 3–10; 3 = 11–30; 4 = 31–100; and 5 = more than 100 galls. Washing of the individual soil sample was done by using Cobb's sieving and decanting technique (Cobb 1918, Schnidler 1961). This is the basic technique which involves the principle of gravity. The difference in the specific gravity and size between soil components and nematodes were used. Nematodes being lighter in weight can be disunited from the soil using this method. In this method, intermingling of the soil with voluminous water is done and then the admixture is poured through set of sieves of different mesh sizes (20, 200 and 325 mesh sieve) to retain nematodes. First of all, the individual sample was mixed thoroughly and then 250cc of the soil was taken using 250 ml beaker and poured into a plastic tray A of large capacity. Then, approximately 1litre of the water was added to the tray and the admixture was mixed properly by continuously stirring with hand and allowed to settle the debris or heavy particles for ten seconds. After that decantation was done by passing through a coarse sieve (20

mesh sieve) into another plastic tray B. During this procedure along with water suspension in tray B nematodes were also carried. The left over material in tray A and on the coarse mesh sieve were discarded. The mixture in the tray B was mixed, allowed to settle for 10 seconds and then poured into the tray A through 200 mesh sieve. The residue left in tray B was discarded. The content of 200 mesh sieve was backwashed and collected in the beaker. The mixture in tray A was again mixed, allowed to settle for few seconds and then poured into tray B through 325 mesh sieve and the residue in the tray A was discarded. The contents of 325 mesh sieve which includes the nematodes were backwashed using squeezed water bottle into a beaker. The suspension collected in the beaker was then passed through a tissue paper, which was placed on aluminium wire gauze. After that a Petri dish (10 cm in diameter) filled with distilled water was taken and the gauze along with tissue paper was placed over it in such a way that the gauze may touches the surface of water. Then, the sample was allowed to be undisturbed for 24 hours. After 24 hours, the suspension in the petiolate was collected and observed under stereo zoom binocular microscope for the nematode examination. Reproduction factor (Rf) which is the final population divided by the initial population was calculated. The roots of each plant were washed under tap water and spread on a white paper and on the basis of number of galls each plant was graded as given below: The root galling index was calculated.

$$RGI = \frac{\text{Sum total of grades of all the plants observed}}{\text{Total number of plants observed}}$$

Statistical analysis: The differences among means were compared by Tukey method ($P < 0.05$). Nematode variables were regressed as the dependent variable with the initial inoculum level as the independent variable under two agro-ecosystems.

RESULTS AND DISCUSSION

Effect on soil nematode population, root galling index and growth parameters: The buildup of nematode population was greater in polyhouse as compared to open conditions (Table 2, 3) and even low inoculum levels crossed threshold levels in polyhouse indicating supportive build up for root knot nematode populations. The inoculum level of $P_i=500$ exhibited higher multiplication in polyhouse and thus reaching pathogenic levels 2025 J2/kg soil ($>1J2/g$ soil) while at the same inoculum level the final population was 799.50 juveniles in open cultivation which was relatively less than the pathogenic levels marked for nematode species ($1J2/g$ of soil) (Table 2, 3). The comparative account of root galling

index (RGI) also indicated that number of galls were observed were higher in polyhouse cultivated tomato (Table 3). The number of egg masses were more on roots of tomato grown in polyhouse though it was not significant except at Pi= 500 (Table 3). In the two ecosystems, comparatively, the percent reduction in shoot length and shoot weight of tomato was higher in polyhouse as compared to open cultivation up to moderate levels of inoculum indicating more severity in polyhouses in comparison to control (Pi = 8000 J2) (Table 1).

Effect of different inoculum levels of *Meloidogyne incognita* on root galling and soil nematode population:

The tomato plants inoculated with second stage juveniles of *Meloidogyne* spp. significantly affected both root galls and nematode's population after harvest. Significant increase in number of galls was also observed at all inoculum levels in

polyhouse as well as open conditions. The increase in initial levels of *M. incognita* resulted in significant increase in soil nematode population, number of egg masses and root galling index (RGI) (Table 2, 3). Multiplication of nematode was observed at all inoculum levels. The highest number of egg masses were at Pi= 8000 and lowest at Pi= 100 (Table 4). Reproduction factor of nematode was reduced as initial population levels of nematode increased and minimum at Pi= 8000 (Table 3). Soil nematode population was observed to be highest at Pi= 8000 and minimum at Pi= 100. Soil nematode population was indicated direct relationship with inoculum levels (Table 2). The presence of root galls and number of root knot nematode juveniles inoculated has a strong negative correlation with the growth parameters. This indicate, with increase in the inoculums level, the root galling

Table 1. Effect of different inoculum levels of root-knot nematode on shoot length and shoot weight of tomato under polyhouse and open field conditions

Inoculum levels	Per cent reduction in shoot length (cm)		Per cent reduction in shoot weight (g)	
	Polyhouse	Open	Polyhouse	Open
100 J2s	0.79	2.24	4.68	9.27
500 J2s	11.03	4.59	20.97	14.5
1000 J2s	17.01	8.54	23.90	16.12
2000 J2s	20.83	13.97	33.70	19.75
4000 J2s	39.84	28.37	54.68	29.33
8000 J2s	81.74	71.89	66.85	50.30

Table 2. Effect of different inoculum levels of root-knot nematode on final soil nematode population in tomato under polyhouse and open field conditions

Agro-ecosystem	Soil population per 250 cc soil						
	0 J2s	100 J2s	500 J2s	1000 J2s	2000 J2s	4000 J2s	8000 J2s
Polyhouse	0.00 ^d	249.50 ^f	2025.00 ^e	2483.00 ^d	4325.00 ^c	5799.50 ^b	6033.16 ^a
Open	0.00 ^d	201.00 ^f	799.50 ^e	2166.50 ^d	3153.00 ^c	3841.50 ^b	5316.66 ^a

Means sharing common letters within columns do not differ significantly by Tukey's test at P < 0.05%.

Table 3. Effect of different inoculum levels of root-knot nematode on root galling index, egg mass index and reproduction factor of tomato under polyhouse and open field condition

Agro-ecosystem	Root gall index (RGI) 0-5 scale						
	0 J2s	100 J2s	500 J2s	1000 J2s	2000 J2s	4000 J2s	8000 J2s
Polyhouse	0.00 ^d	1.16 ^f	1.83 ^e	2.50 ^d	3.50 ^c	4.16 ^b	4.83 ^a
Open	0.00 ^d	1.00 ^e	1.33 ^e	2.16 ^d	3.16 ^c	3.83 ^b	4.83 ^a
Egg mass index (EMI)							
Polyhouse	0.00 ^d	14.80 ^f	36.60 ^e	43.30 ^d	55.60 ^c	94.30 ^b	106.20 ^a
Open	0.00 ^f	12.30 ^e	15.90 ^e	39.50 ^d	51.90 ^c	92.50 ^b	102.45 ^a
Reproduction factor (Rf)							
Polyhouse	0.00	2.49	4.05	2.48	2.16	1.44	0.75
Open	0.00	2.01	1.59	2.16	1.57	0.96	0.66

Means sharing common letters within columns do not differ significantly by Tukey's test at P < 0.05%.

of tomato plant increased which showed a significant negative impact on the plant height and weight. that increased number of nematode juveniles have a strong positive correlation with the presence of root galling. This implies that, as nematode population increases in the rhizosphere of the tomato plant, the presence of root galling of tomato plant roots also increases. The increase in initial inoculum levels of *M. incognita* was significant and increase in soil nematode population, number of egg masses and root galling index (GI). Multiplication of root knot nematode was observed at all inoculum levels. The highest number of egg masses were observed at $P_i= 8000$ and lowest at $P_i= 100$. However, reproduction factor of root knot nematode was reduced as initial population levels of nematode increased and was minimum at $P_i= 8000$ (Table 3).

Effect of inoculum levels of *Meloidogyne incognita* on growth parameters of tomato: The growth parameters of tomato were affected by infestation of *M. incognita* in both the cultivation systems. Shoot length as well as shoot weight decreased with increase in inoculum level of the nematode (Table 1). In tomato plants inoculated, minimum reduction was found in plants inoculated with $P_i= 100$, while maximum decrease in shoot length and weight was observed at $P_i= 8000$. The differential rates of buildup of nematodes in the two ecosystems. The infestation of *M. incognita* in the polyhouses was significantly higher as compared to open cultivation of tomato at all inoculum levels. This may be due to the difference of environmental conditions prevailing in the protected and open ecosystems. The multiplication of *M. incognita* was higher in polyhouse conditions and even low inoculum load in soil crossed the threshold levels (the minimum intensity or amount that must be reached for a specific event, or condition to occur or be noticeable) and had greater effects on the health of the crop as compared to plants which were grown in open field. Ndiffon (2024) reported that the overall multiplication rates are influenced by nematode species, the susceptibility of the host and the various environmental factors. Nematodes being invertebrates, have their life cycle dependent upon the environmental temperature. Their reproductive rates and metabolism are directly proportional and respond to fluctuations in temperature. Monitoring of temperatures during the conduct of experiment revealed that it was comparatively higher in polyhouses as compared to open. The, increased temperatures generally increase plant growth rates, which in turn gives more food source to nematode pests besides affecting plant phenology and increasing the whole ecosystem complexity. Both of these factors also lead to an earlier emergence of pests/diseases/vectors and crop attacks, longer life-cycles and reduced pest/disease

generation times (Colagiero and Ciancio 2011). In earlier on population dynamics of root knot nematode, enhanced population build up from 1 to 30 J2/c.c. soil within a period of 6-12 months was observed in polyhouse which is comparatively higher in contrast to the open cultivation (Minuto et al., 2006, Engindeniz and Engindeniz 2006).

The different inoculum levels of *M. incognita* resulted in significant reductions in growth variables. In nematode infested plants there was reduction in shoot length and shoot weight, which could be due to the damage caused by increasing numbers of nematodes that invaded roots and hampered the nutrient and water uptake of plants (Karszen and Moens 2006). The effects of nematode on tomato increase as the inoculum levels increased. In the present studies, the multiplication of nematode was observed to be higher at low to moderate inoculum levels ($R_f > 1$). High rate of multiplication of nematodes with low level of inoculum might be due to encouraging factors like plenty of food, reduced competition level and the ability of hosts to support these populations (, Bendezu and Starr 2003).

CONCLUSION

Meloidogyne incognita exhibits significantly higher reproduction and pathogenic impact in polyhouse conditions than in open fields, even at lower inoculum levels. This highlights the need for tailored nematode management strategies in protected cultivation systems.

AUTHOR'S CONTRIBUTIONS

Anupam Sekhon and Narpinderjeet Kaur Dhillon jointly planned and executed the research work. Anupam Sekhon and Harwinder Singh Buttar carried out data collection for root galling index and soil nematode population and statistical analysis. Narpinderjeet Kaur Dhillon and Sukhjeet Kaur supervised the entire study and critically reviewed the manuscript. All authors contributed to manuscript writing and approved the final version.

REFERENCES

- Abrar S, Seid A and Dejene M 2020. Integrated management of *Meloidogyne incognita* in tomato (*Solanum lycopersicum*) through botanical and intercropping. *African Journal of Agricultural Research* **15**(4): 492-501.
- Ahmed N, Ghramh HA, Shakeel Q, Ashraf W, Abbas HT, Binyamin R, Masroor A, Raheel M and Khan Z 2023. Evaluation of Rhizospheric-Pseudomonas spp. for the management of *Meloidogyne incognita* in tomato. *Journal of King Saud University-Science* **35**(1):102395.
- Bendezu IF and Starr J 2003. Mechanism of resistance to *Meloidogyne arenaria* in the peanut genotype. *COAN Journal of Nematology* **35**: 115-118.
- Cobb NA 1918. Estimating the nema population of a soil. *Agric Tech Circul*, Burr Plant Industr U S Dept. Agri pp. 48.
- Colagiero M and Ciancio A 2011. Climate changes and nematodes:

- expected effects and perspectives for plant protection. *REDIA*. **10**: 113-118.
- Engindeniz S and Engindeniz DY 2006. Economic analysis of pesticide use on greenhouse cucumber growing: A case study for Turkey. *Journal of Plant Disease Protection* **113**: 193-198.
- Karssen G and Moens M 2006. *Root-knot nematodes*. pp. 59-90. In: Perry, R.N. and Moens, M. (Eds.). *Plant Nematology* CABI publishing.
- Khan A, Haris M, Hussain T, Khan AA, Laasli S, Lahlali R and Mokri F 2023. Counter-attack of biocontrol agents: Environmentally benign Approaches against Root-knot nematodes (*Meloidogyne* spp.) on Agricultural crops. *Heliyon* **9**: 11 e21653.
- Kumar V, Khan MR and Walia RK 2018. Crop Loss Estimates due to Plant-Parasitic Nematodes in Major Crops in India. *National Academy Science Letters*. <https://doi.org/10.1007/s40009-020-00895-2>.
- Minuto A, Gullino ML, Lamberti FD, Adabbo T, Tesari, Ajwa F and Garibaldi H 2006. Application of emulsifiable mixture of 1,3 Dichloropropene and chloropicrin against root-knot nematode and soil fungi for green house tomato in Italy. *Crop Protection* **25**: 1244-1252.
- Ndifon EM 2024. Nematode populations affect grapevine seedlings: understanding the host-parasite interaction is critical for preventing negative effects. *RIA Rev investig agropecu*. **50**: 3.
- Onkendi EM, Kariuki GM Marais M and Moleleki LN 2014. The threat of root-knot nematodes (*Meloidogyne* spp.) in Africa: A review. *Plant Pathology* **63**: 727-737.
- Pires D, Vicente CSL, Menéndez E, Faria JMS, Rusinque L, Camacho MJ and Inácio ML 2022. The Fight against Plant-Parasitic Nematodes: Current Status of Bacterial and Fungal Biocontrol Agents. *Pathogens* **11**(10): 1178.
- Qiao K, Liu X, Wang H, Xia X, Ji X and Wang K 2012. Effect of abamectin on root-knot nematodes and tomato yield. *Pest Management Science* **68**: 853-857.
- Schindler AF 1961. A simple substitute for a Baermann funnel. *Plant Disease Reporter* **45**: 747-748.
- Zakaria HM, Kassab AS, Shaseldean MM, Oraby MM and El-Mourshedy MMF 2013. Controlling the root-knot nematode, *Meloidogyne incognita* in cucumber plants using some soil bioagents and some amendments under simulated field conditions. *Annals of Agricultural Science* **58**: 77-82.

Received 15 May, 2025; Accepted 10 July, 2025