# Distribution and identification of root-knot nematode species in tomato fields

# Mehdi Nasr Esfahani

Isfahan Agriculture Research Center, I.R. Iran \* Corresponding author's e-mail: m\_nasresfahani@yahoo.com

### Abstract

The survey conducted to assess the incidence of root-knot disease on tomato in 8 localities in and around Aligarh (India) showed that the crops of tomato in all the 8 localities were infected with root-knot nematodes. Highest frequency (90%) was found in Idgah area, closely followed by Shah Jamal and Mathura Road localities. In both the localities the frequency was 80%. In Delhi Road and Agra Road areas the frequency of the disease was 60% and 70% respectively. The frequency in Ramghat Road area was 50%. The lowest frequency (40%) was found in Kasimpur Road and University Farm areas. Both gall index and eggmass index (average) ranged between 2-5 through 3 and 4. The greatest eggmass and gall indicates (5 each) was found in Idgah area, the area in which the incidence was also greatest. The gall and eggmasses indices were 4 in Shah Jamal Delhi Road, Mathura Road and Ramghat Road areas. In Agra Road and university Farm areas the indices were 3 each. The lowest incidences (2 each) were noticed for Kasimpur Road area. Thus, the intensity of the disease on tomato was highest in Idgah closely followed by Shah Jamal, Delhi Road, Mathura Road and Ramghat Road areas in descending order. In other three localities (Agra Road, University Farm, Kasimpur Road) the intensity of the disease was comparatively low. Meloidogyne incognita and M. javanica, were identified to infect tomato in different areas. The species were either found singly or in mixed populations. Out of the two, *M. javanica* was more frequent. It was found in 7 localities out of 8 either singly or in concomitantly with M. incognita. In three areas (Agra Road, Mathura Road and Ramghat road) it was encountered alone but in other four areas it was present together with M. incognita. It was present in all the localities except Delhi Road area. Keywords: Aligarh, M. javanica, M. incognita, Root-knot nematodes, Tomato.

# Introduction

Tomato (*Lycopersicon esculentum* Mill.) is one of the most important vegetable crops grown throughout the world for consumption in various forms. A number of viral, bacterial, fungal and nematode pathogens attack tomato and cause diseases of economic consequences.

Root-knot nematodes (*Meloidogyne* species), reniform nematodes (*Rotylenchulus reniformis*), cyst nematode (*Globodera rostochiensis*) and several ecto-parasitic nematodes are known to attack tomato in many different parts of the world. Tomato is regarded as the most favorable host for root-knot nematodes (Dropkin, 1980; Taylor, 1967).

Yield losses caused by the root-knot nematode, *Meloidogyne incognita*, were assessed in gherkin (cucumber) fields in Kolar and Bagepalli, Karnataka, India. Root-knot nematode incidence and severity were high at both locations. Root-knot index ranged from 3.2 to 4.5 on a 0-5 scale with number of egg masses ranging from 69 to 98/g root (Nagesh *et al.*, 2005).

All the four major species of *Meloidogyne* viz., *M. incognita*, *M. javanica*, *M. arenaria*, *M. hapla* and their known races readily attack tomato

crops in outdoor as well as in indoor cultivations. Studies have shown that root-knot nematodes can cause suppression in yield of tomato as high as 85% (Sasser, 1979; Taylor and Sasser, 1978).

Nematodes of *M. incognita* were isolated from the roots of paprika plants, *Capsicum annum* L., from the greenhouse situated in Portorož at the Adriatic Coast, Slovenia. *M. hapla* was found for the first time in the open field in Slovenia. It was isolated from galls of the sweet pepper grown in the open field in Ljubljana (Širca *et al.*, 2004).

As tomato are grown almost throughout the world in plains as well as on hilly tracks both in outdoor and indoor cultivations and root-knot nematodes ubiquitous in distribution, the crop of tomato is always under the threat of attack by root – knot nematodes. Tomatoes being a cash crop in many parts of the world, control measures to save the crop from diseases are usually taken.

A perusal of literature on root-knot nematodes indicates that, different aspects of rootknot nematode problem of tomato have been studied most thoroughly in comparison to other crops. The existence of root-knot nematodes was first observed as the worms in galls on the roots of *Dodartia orientalis* and recognized that they belonged to the genus *Heterodera*, and then recorded by Barkeley (1855) on glasshouse cucumbers in England. Since then the pathogens have been designated for a considerably long period of time with different names (Sasser, 1979). Root- knot nematodes were first named in 1879 by Cornu as *Anguillula marioni* in France (Cornu, 1879). Sasser (1977) summarized the occurrence of root-knot nematode species in different parts of the world. His report included 11 species in Africa, 9 species in Central and South America, 18 species in the United states, 3 species in Canada,11 species in Europe and Mediterranean region, 10 species in India and Sri Lanka, 4 species in Russia, 5 species in Japan and 3 species in Southeast Asia, Australia and Fiji Islands.

Recognizing the importance of root-knot nematodes in agricultural economy, an called international project International Melioidogyne Project (IMP) was initiated with its headquarters at the North Carolina state University, Raleigh under the leadership of Prof. J.N. Sasser. with the assistance of more than 160 nematologist spread over to 70 countries of the world, the occurrence and distribution of species, their relative importance and differentiation and distribution of races have been worked out (Taylor and Sasser, 1979). In the present study attempts have been made to investigate root-knot nematodes problem of tomato in and around Aligarh by assessing incidence and intensity of the disease.

# **Materials and Methods**

#### Survey

A survey was conducted in different localities in and around Aligarh to assess the incidence of root-knot disease on tomato crop.

During the survey of tomato fields in each locality samples of the roots of the plants were collected randomly. Root samples kept in polythene bags and properly labeled were brought to the laboratory and thoroughly examined for the presence of galls. Numbers of galls per root system, if present, were counted.

Roots were washed clean and were then immersed in an aqueous solution of phloxin B (0.15 g/lit) for 15 minutes and then washed with tap water to stain egg masses.

Number of egg masses per root-system was then counted. Gall index (GI) and egg mass index (EMI) were determined on the following scale:

0=0, 1=1-2, 2=3-10, 3=11-30, 4=31-100and 5=greater than 100 galls or egg masses per root system (Taylor and Sasser, 1978). The frequency of occurrence (percentage) of the disease in each locality was calculated by the following formula:

Frequency of occurrence = <u>Number of fields with root-knot nematode infection</u> ×100 Number of fields surveyed

#### Maintenance of inoculum

The inoculum of some selected filed populations from each locality was maintained on tomato cv. Pusa Ruby in a greenhouse by inoculating seedling in pots containing autoclaved soils with chopped infected root collected from the filed. The inoculum was further cultured in pure form on tomato roots as follows.

#### **Pure culturing**

In order to make pure culture of filed population maintained on tomato in greenhouse, single egg mass inoculation was made. Single mature egg mass was inoculated in pots around the root of young tomato seedling for each maintained collection separately.

Sub – culturing was done by inoculating new tomato seedling with at least 15 egg masses, each obtained from pure culture in order to maintain sufficient inoculum for further studies.

#### **Identification of the species**

Identification of the species of *Meloidogyne* collected from each locality and also maintained in greenhouse was done by applying perineal pattern method (Eisenback *et al.*, 1981).

Mature females were dissected out from large galls on the roots of tomato plants. Perineal patterns slides (10-20) from each sample or locality were prepared and examined under microscope to study their characteristics. The species were identified on the basis of perineal pattern characteristics (Eisenback *et al.*, 1981).

# **Results and Discussion**

#### Incidence of root-knot on tomato

The survey conducted to assess the incidence of root-knot disease on tomato in 8 localities in and around Aligarh (Table1) showed that the crops of tomato in all the 8 localities were infected with root-knot nematodes. Therefore, overall incidence of the disease was 100%.

Locality –wise variations in the incidence of the disease were, however, found. Highest frequency (90%) was found in Idgah area, closely followed by Shah Jamal and Mathura Road localities. In both the localities the frequency was 80%. In Delhi Road and Agra Road areas the frequency of the disease was 60% and 70% respectively. The frequency in Ramghat Road area was 50%. The lowest frequency (40%) was found in Kasimpur Road and University Farm areas (Table1).

The intensity of the disease on tomato in these localities based on average gall and egg mass indicate was high in general. Area-wise variations were, however, noticed. Both gall index and egg mass index (average) ranged between 2-5 through 3 and 4. The greatest egg mass and gall indicates (5 each) was found in Idgah area, the area in which the incidence was also greatest. The gall and egg masses indices were 4 in Shah Jamal Delhi Road, Mathura Road and Ramghat Road areas. In Agra Road and university Farm areas the indices were 3 each. The lowest incides (2 each) were noticed for Kasimpur Road area (Table1). Thus ,the intensity of the disease on tomato was highest in Idgah closely followed by Shah Jamal, Delhi Road, Mathura Road and Ramghat Road areas in descending order .In other three localities (Agra Road, University Farm, Kasimpur Road ) the intensity of the disease was comparatively low.(Table1).

#### **Identification of the species**

the basis of perineal On pattern characteristics, Meloidogyne incognita and M. javanica, the two species of root-knot nematodes were identified to infect tomato in different areas included in the survey. The species were either found singly or in mixed populations. Out of the two, M. javanica was more frequent. It was found in 7 localities out of 8 either singly or in concomitantly with *M. incognita*. In three areas (Agra Road, Mathura Road and Ramghat road) it was encountered alone but in other four areas it was present together with M. incognita. It was present in all the localities except Delhi Road area. On the other hand, *M. incognita* was present alone only in one area (Delhi road), in other four areas it was encountered in mixed populations with M. javanica. It was not encountered in Agra Road, Mathura Road and Ramghar Road areas (Table2).

The survey conducted in some localities in Aligarh to assess the incidence of root-knot disease on tomato showed that disease was quite frequent as it was observed in all the localities included in the survey. The incidence in general was high. Similarly, the intensity of the disease was also high. Apparently, tomato cultivation in the area is suffering because of this disease as most of the fields grown with tomato in the area are infested with root-knot nematodes.

These results are in accordance with the observations made in different parts of the world (Sasser, 1979). In the early spring of 2006, severe plant stunting, chlorosis, and extensive root galling were observed on cucumber plants grown in a greenhouse on Zeta plain, Zetska ravnica, Montenegro. The most prevalent species was M. incognita, which was isolated from the roots of tomatoes, peppers, cucumbers, and lettuce from 9 locations. *M. arenaria* was detected at 3 locations from the roots of tomatoes grown in a greenhouse and the weeds Convolvulus arvensis L. and Solanum nigrum L., which were growing in open fields in separate locations. M. javanica was found on tomato and squash in the same field where M. arenaria was also found on S. nigrum. M. javanica was isolated from tomato and squash. In this study, high incidence of *Meloidogyne* spp. was found in production areas intensive vegetable of Montenegro. This is the 1st report of *M. arenaria*, M. incognita, and M. javanica from Montenegro (Pajovic et al., 2007).

A similar survey conducted by Khan et al. (1984) also indicated that tomato is the most affected crop and suffers most among vegetable grown in Aligarh area, because of root-knot nematodes. The present findings confirm their results. Severe stunting and extensive root galling were observed on tomato rootstock resistant to Meloidogyne incognita, M. javanica and M. arenaria in northern Switzerland. Examination of the roots of infected plants revealed the presence of root-knot nematodes in large numbers. All methods of identification were consistent with M. enterolobii. The species M. enterolobii is of great importance because it is able to reproduce on resistant tobacco, pepper, watermelon, and tomato. This is the first report of M. enterolobii in Switzerland (Kiewnick et al., 2008).

The identification of the species showed that *M. javanica* and *M. incognita* are the species mainly causing the disease in the area. *M. javanica* is apparently dominant.

Mixed populations of both species in tomato fields are also common. These observations confirm the result of Khan *et al.* (1984) and Khan and Khan (1985), who observed the common occurrence of these species with dominance of *M. javanica* and their mixed population in the area. It further endorses the view of Khan *et al.* (1984) that *M.incognita* is not only the species infecting crops in this area as believed for long time due to lack of pursuance of studies to establish the identity of *Meloidogyne* species occurring in the area. Southern root knot nematode (*Meloidogyne incognita*) is the most widespread and important. This pathogen not only causes root galling but also increases the severity of *Fusarium* wilt. The incidence of southern root-knot nematode on cotton has increased significantly as the acreage allocated to this crop has grown (Ye *et al.*, 2008).

Root-knot nematodes (*Meloidogyne* spp.) are common nematodes that parasitize vegetables in Florida and cause significant yield reductions when not properly managed. *Meloidogyne incognita*, *M. javanica* and the first report of *M. floridensis* were observed on tomato in Florida. The identification and distribution of *M*. *floridensis* in vegetable production fields is important for disease management throughout the state since the host range is likely different from other *Meloidogyne* spp. (Taylor *et al.*, 1982; Church, 2005).

Among the species of *Meloidogyne* recorded in association with crops of agricultural importance in subtropical and tropical regions, *M .incognita* and *M. javanica* are considered as common and wild–spread (Sasser, 1979). The present findings further confirm this contention. *M. incognita* and *M. javanica* are also most frequency encountered species even on world-wide basis (Sasser, 1979). A study conducted under the project of IMP has shown this pattern of their distribution (Taylor *et al.*, 1982).

Table1: Frequency	distributions of	root-knot n	nematodes in	different	localities in	1 and	around Aligarh.
-------------------	------------------	-------------	--------------	-----------	---------------	-------	-----------------

Localities	Total No. Of filed surveyed	No. of filed with infection	Frequency (%)	GI <sup>1</sup> /EMI <sup>2</sup> (Average)
Shah Jamal	10	8	80	4/4
Idgah	10	9	90	5/5
Delhi Road	10	6	60	4/4
Agra Road	10	7	70	3/3
Mathura Road	10	8	80	4/4
Ramghat Road	10	5	50	4/4
Kasimpur Road	10	4	40	2/2
University Farm	10	4	40	3/3

 $^{1}$ GI = Gall index

 $^{2}$ EMI = Eggmass index on Taylor and Sassers scale

**Table2**: Identification of *Meloidogyne* species infecting tomato in different localities in and around Aligarh<sup>1</sup>.

Localities	Meloidogyne species
Shah Jamal	M. incognita, M. javanica
Idgah	M. incognita, M. javanica
Delhi Road	M. incognita
Agra Road	M. javanica
Mathura Road	M. javanica
Ramghat Road	M. javanica
Kasimpur Road	M. incognita, M. javanica
Univercity Farm	M. incognita, M. javanica

1 This place is situated Uttar Pradesh, India, its geographical coordinates are 27° 49' 0" North, 78° 17' 0" East.

# References

- Berkely MJ, 1855. Vibro forming cysts on the roots of Cucumbers. Gdnrs chron. 220.
- Church GT, 2005. First Report of the Root-Knot Nematode *Meloidogyne floridensis* on *Tomato* (*Lycopersicon esculentum*) in Florida. *Plant Dis.*, **89**: 527.
- Cornu M, 1879. Etudes sur le Phylloxera vastatrix. Mem Acad. Sci. Paris. 26: 163-175
- Dropkin VH, 1980. Introduction to Plant Nematology. John wily and Sons, New york. Eisenback JDH, Hirschmann JN, Sasser,

Triantaphyllou AC, 1981. A more complete

characterization of the four most common species of root-knot nematodes (*Meliodogyne* spp.) with pictorial Key. IMP publication Raleigh, N.C.U.S.A

- Khan AA, Khan MW, 1985. Root –knot nematodes infecting some common weeds in vegetable fileds of Western Uttar Pradesh (India). Internat. Nematol. *Network Newslet*. 2: 15-16
- Khan M, Wajid MR, Khan, Khan AA, 1984. Identity of root –knot nematodes on certain vegetables of Aligarh District in Northern– India. Internat.Nematol. *Network Newslet*. 1: 6-7.
- Kiewnick S, Karssen G, Brito A, Oggenfuss M.and Frey JE, 2008. First Report of Root-Knot Nematode *Meloidogyne enterolobii* on Tomato and Cucumber in Switzerland. *Plant dis.*, **92**: 1370.
- Nagesh M, Hussaini SS, Chidanandaswamy BS, 2005. Incidence of root-knot nematode, *Meloidogyne incognita* on gherkin, *Cucumis* sativus and yield losses. *Indian J. Plant Prot.*, 33: 309-311.
- Pajovic I, Širca S, Geric Stare B, Urek G, 2007. The incidence of root-knot nematodes *Meloidogyne arenaria*, *M. incognita*, and *M. javanica* on vegetables and weeds in Montenegro. *Plant Dis.*, **91**: 1514.

- Sasser NJ, 1977. Worldwide dissimination and importance of the root-knot nematodes. *Meloidogne* spp. J.Nematol., **9**: 26-29
- Sasser JN, 1979. Economic importance of *Meloidogne* in tropical countries. pp. 359-374 in: Root–knot nematodes (*Meloidogne* spp.) systematics, biology and Control (Eds. F. Lamberti and C.E. Taylor ). Academic Press, London.
- Širca S, Urek G, Karssen G, 2004. The incidence of the root-knot nematode *Meloidogyne incognita* and *Meloidogyne hapla* in Slovenia. Acta Agric. Slovenica, 83: 15 – 22.
- Taylor AL, Sasser JN, (1978). Identification and control of root-knot nematodes (*Meloidogyne* spp.) crop. Publ. Dep. Plant Pathol, North Carolina State Univ. and U.S. Agrency Int. Dev. Raliegh, N.C. PP111.
- Taylor AL, Sasser JN, Nelson LA, 1982. Relationship of climate and soil characteristics of geographical distribution of Meloidogne species in agricultural soil. IMP publication, Raleigh, North Carolina.
- Taylor AL, Sasser JN, 1979. Bilogy, identification and control of root –knot nematodes (*Meloidogne* spp.) IMP Publication, Raleigh, North Carolina.
- Ye W, Hudak C, Troxler S, 2008. Nematode Management in Cotton. North Carolina Agronomic Division. Nematode Assay Section/October. NCDA & CS.