# Incidence of Meloidogyne incognita in Cucumber Fields

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Abstract.- In 2008 and 2009, a survey was conducted in vegetable production area of Taxila to determine the incidence and distribution of root knot nematodes, *Meloidogyne* spp. in open-field and plastic tunnel planted cucumber crop. Two root knot nematode species. *M. incognita* and *M. javanica*, were identified from 200 samples. *Meloidogyne incognita* was the predominantly found species and was detected in 95% of all the fields surveyed. The average incidence was 46% and ranged from 30 to 60% on open-field planted cucumber crop, whereas on tunnel planted crop it was 35% and ranged from 20 to 50%. The cucumber planted in open-fields as well as plastic tunnels located in Faisal Town production area had the highest disease incidence of 60%, whereas the production area of Lossar Sharfu had the lowest frequency of 20% both in fields and tunnels. The average gall and egg mass indices on scale of 0 to 5 were same (2.6) on roots of open-field planted crop. But the roots of plants within tunnels had 3.85 % and 7.14 % greater gall and egg mass indices over that of open-field plants. These findings warrant that root knot nematode is widely distribution in vegetable production area.

Key words: Cucumber, incidence, Meloidogyne spp., open-field, plastic tunnels, root gall and egg mass indices.

## **INTRODUCTION**

Cucumber (Cucumis sativus L. family: cucurbitaceae) is the most important vegetable crop in open-field and plastic-tunnel farm lands in Pakistan. Cucumber is affected by many different viral, bacterial, fungal and nematode pathogens. More specifically to nematodes there are six plant parasitic nematode genera frequently reported to infect vegetable crops including Aphelenchoides, Helicotylenchus, Pratylenchus, Meloidogyne, Rotylenchulus, and Scutellonema (Kaur et al., 2010; Anwar et al., 2007; Sharma et al., 2004). However, it is generally determined that root-knot nematodes are among the most prevalent on vegetables worldwide (Anwar et al., 2007; Anwar and McKenry, 2012; Di Vito et al., 1985; Sharma et al., 2004; Kamalwanshi et al., 2004; Piedra et al., 2006). There can be serious economic consequences when Meloidogyne spp. are in abundance (Elekçioğlu and Uygun, 1994; Devran et al., 2008).

Incidence of *M. incognita* in Punjab was recently quantified at 58% with a gall severity rating of 8.5 out of 9 causing 25% annual yield loss for cucumbers plant on-open fields (Anwar and

McKenry, 2010, 2012). For purposes of comparison this damage level is double that reported from North Carolina, USA (Main and Gurtz, 1989). They cause damage by direct feeding and by inducing large galls throughout the root system of infected plants. Their presence can alter uptake of water and nutrients, interfere with translocation of photosynthates (Anwar, 1995; Davis *et al.*, 2003; Williamson and Hussey, 1996) and increase incidence and severity of *Fusarium oxysporum* f. sp. *radicis-cucumerinum* diseases (Vakalounakis, 1999).

Greater recognition of the importance of rootknot nematodes attacking vegetable crops and increased interest in planting vegetable crops under plastic tunnels has promoted this study. The data on the occurrence of nematode with cucumber in openfields is available (Anwar and McKenrry, 2010, 2012) but in the plastic tunnels is lacking. The present investigation was planned to assess the incidence of root-knot nematodes on roots of cucumber crops planted in open-fields and within plastic-tunnels. This study will provide the comparison of presence of root knot nematode between open-fields and tunnel planted cucumber crops.

#### MATERIALS AND METHODS

During 2008 and 2009, ten different localities were sampled to quantify root-knot nematodes

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presence in open-field and tunnel-planted cucumber crops. Cumber cu. Bilbao was planted under 12 plastic tunnels having a polythene sheet of 15 mµ thickness and size of  $30 \times 104 \times 11$  feet each. These were scattered on an area of 12 acres, whereas the area of open field was <u>ca</u> 5 acres. The temperature for each sampling locality was recorded at the time of sampling during January and February (Table I).

 Table I. Temperature (°C) of sampling sites and J2 population of Meloidogyne incognita

Localities	Temperature (°C) at the time of sampling		J2 population per g of root		
sampled	Tunnels	Open field	Tunnels	Open field	
Faisal Town	28.0	21.0	59.7a	50.0b	
Malikabad	27.8	22.0	50.0a	40.0b	
Mora shah wali	27.2	20.8	40.0a	30.7b	
Qaziabad	27.0	21.2	40.7a	31.3b	
Mohallah Majhian	26.8	21.2	60.0a	39.3b	
Sagran	27.9	21.3	50.0a	20.0b	
Ghazi Kohli	28.0	21.0	50.3a	49.7a	
Hassan Colony	28.7	20.9	40.7a	40.3a	
Ban Bhola	28.5	21.0	40.0a	29.7b	
Lossar Sharfu	28.2	21.2	30.0a	20.7b	

\*Mean values in the same row carrying small similar letters do not differ Significantly (p < 0.05 Student t-test).

Sites sampled were located at Faisal Town, Malikabad, Mora Shah Wali Shah, Oaziabad, Mohallah Majhian, Ban Bhola Ghazi Kohli, Lossar sharfu, Hassan colony and Sagran in the vegetable production area of Taxilla, Rawalpindi. Surface soils (0 cm to 20-cm) across these regions averaged 65% to 85% sand, 5.5% to 20.25% silt, 10% to 21.5% clay and 0.5% to 2.75% organic matter. Each nematode sample consisted of ten cores including roots and soil collected by Oak land tube of 2.5-cm diameter to depth of 18-20 cm close to the root system of each plant. The individual pants were also uprooted to assess the root galls and egg masses per root system. At least 10 plants from each site were randomly selected at fruiting stage and sampled. Each sample consisted of roots with adhering soil collected between 5 and 30-cm deep. A composite sample was collected into a plastic bag and placed in a 5°C refrigerator prior to processing.

All root samples were carefully washed under tap water to remove adhering soil particles, and closely entangled grass roots, and then towel dried. Root systems of the plants were stained with Phloxine B (Holbrook et al., 1983) and assessed for presence of egg masses. Root galling and egg mass indices were assessed on a 0 to 5 scale, where 0 =no gall, 1 = 1-2, 2 = 3-10, 3 = 11-30, 4 = 31-100, and 5 = >100 galls per root system (Quesenberry et al., 1989). The data on number of galls and egg masses per root system was recorded. Incidence of root-knot nematode was assessed on basis on root galling. Nematodes were extracted from 20g of roots after placement in a mist-chamber for 3 days (McKenry and Roberts, 1985). Second stage juveniles (J2) population per gram of root was calculated.

## Identification of root knot nematodes

Root-knot nematodes present in these samples were identified using perineal patterns (cuticle markings around the anus-vulva region of the female nematodes) of adult females as well as the morphology of second stage juveniles (Hartman and Sasser, 1985; Jepson, 1987).

## **RESULTS AND DISCUSSION**

## Identification of root-knot nematodes

Two species of root-knot nematode, M. incognita and M. javanica, were identified within 200 samples from 10 localities on the basis of patterns and morphology of perineal J2. Meloidogyne incognita was found in 95% of cucumber fields with an average gall and egg mass indices of 3.5 and 3.25 on scale of 0 to 5, respectively. Most of the samples were mixed with both species. However, M. javanica was solely found in 5% of the samples. Meloidogyne incognita is to be considered as the most damaging pathogen of cucumber crops in Pakistan (Anwar and McKenry, 2010) and worldwide (Sikora and Fernandez 1990). Meloidogyne incognita, M. javanica, and M. arenaria have often been found associated with cucumber in the United States but M. incognita is the primary species infecting cucumber there and leads to 11% destruction of the crop annually (St. Amand and Wehner, 1997; Walters and Wehner, 1997).

Localities sampled	Field grown cucumber			Tunnel grown cucumber			
	* Incidence %	Indices**			Indices		
		Root galls	Egg masses	Incidence %	Root galls	Egg masses	
Faisal Town	60	3	3	50	4	4	
Malikabad	50	2	2	40	3	3	
Mora shah wali	40	2	2	30	2	2	
Qaziabad	40	3	3	30	3	3	
Mohallah Majhian	60	4	4	40	4	4	
Sagran	50	3	3	20	3	3	
Ghazi Kohli	50	2	2	50	2	2	
Hassan Colony	40	3	3	40	2	3	
Ban Bhola	40	2	2	30	2	2	
Lossar Sharfu	30	2	2	20	2	2	

Table II.- Incidence, root galling, and egg masses indices on roots of cucumber induced by Meloidogyne incognita.

Mean values for incidence and indices for each locality are average of 10 samples.\*Incidence, number of fields with root knot nematodes /total number of samples X 100. \*\* Root galling and egg mass indices were assessed on a 0 to 5 scale, where 0 = no gall, 1 = 1-2, 2 = 3-10, 3 = 11-30, 4 = 31-100, and 5 = >100 galls per root system (Quesenberry *et al.*, 1989).

The roots of tunnel planted cumber plants from eight localities contained significantly (p < 0.05) higher number of nematodes compared to that of open fields. Only two sampling localities including Ghazi Kohli and Hassan Colony had statistically equal J2 root population both in tunnel and open field plants (Table I). These differences might be related to the soil types and particularly of temperature being <u>ca</u>.  $5.7^{\circ}$ C higher in tunnels over that of fields as egg hatching, root invasion by J2, its development to mature female and egg hatching are temperature-dependent (Madulu and Trudgill, 1994; Trudgill, 1995).

## Incidence of root-knot on tomato

The cucumbers planted either in the openfields or plastic tunnels were infected with root-knot nematodes. Variation in incidence of root-knot disease was evident whether open-field or tunnelplanted. Incidence variation also occurred across localities sampled. The percentage incidence in different localities was 46% with a range from 30 to 60% among open-field cucumbers. By contrast, tunnel-planted cucumbers averaged 35% incidence with a range from 20 to 50% (Table II). It was interesting to observe that in open-fields as well as plastic tunnels the plantings located in Faisal Town production area showed highest disease incidence, while the production area of Lossar Sharfu showed lowest disease incidence both in fields and tunnels. Levels of infestation by root-knot nematode ranged from severe to moderate based on direct examination of the infected plant root system. These results, though based on a random survey, indicate the existence of root-knot nematode populations in cucumber production. The percentage incidence in this investigation was based on the direct examination of root galling and egg masses but it could have been higher if soil samples were also examined for J2 population.

This survey was confined in the vegetable production area of Taxila only. A more detailed survey is needed to reveal additional crops and weeds that serve as nematode hosts (Anwar and Akhtar, 1992; Anwar *et al.*, 1992). The reasons for the fairly widespread distribution of root-knot nematode probably include all of the following: their extensive host range, including weeds (Anwar *et al.*, 2009; Sikora and Fernandez 1990), lack of awareness among growers about nematodes and unintentional spread through sharing of seedlings/ transplants and tropical climatic conditions that favor build-up of root-knot nematode populations.

Average gall and egg mass indices were similar (2.6) from roots of open-field plantings. However, roots of plants within tunnels had 3.85 % and 7.14 % greater gall and egg mass indices over that of field plants. Although nematode incidence in open-field crops was comparatively higher than of tunnel planted crops their gall and egg mass indices were higher on roots of the latter. The higher indices on roots of tunnel planted crops indicate greater reproduction by the nematodes due to higher temperature (Table II). Tunnels provided a better environment compared to open-field for nematode reproduction (Park *et al.*, 2005). Our results concur with other reports on nematodes of vegetables (Fassuliotis, 1970; Potter and Olthof, 1993). Root galling severity is a measure of the size of the nematode population. High severity of nematodes impacts foliar growth by inducing various physiological alterations in plant vital functions. The end result of poor foliar growth is ultimately reduced yield (Melakeberhan and Webster, 1993; Anwar, 1995).

This preliminary survey has provided baseline data about occurrence and distribution of root-knot nematodes in the Punjab vegetable production. This warrants a more systematic and detailed investigation on occurrence of nematodes of other vegetable crops and associated weeds in the adjoining areas (Anwar *et al.*, 1992, 2009). More importantly, there is an urgent need to make available procedures of sanitation and nematode riddance whether in plastic tunnels or open fields (Nagesh *et al.*, 2004).

Our results have also provided important information for breeding programs and extension staff to create awareness among growers about the occurrence of root-knot nematode infecting cucumbers. Perhaps most important is the need to design a crop rotation system that avoids cucumber losses using poor root-knot hosts such as cereals, cabbage and mustard (Anwar and McKenry, 2012).

#### REFERENCES

- ANWAR, S.A. AND AKHTAR, S.A., 1992. Evaluation of four vegetables to *Meloidogyne incognita*. J. agric. Res., **30**:415-421.
- ANWAR, S.A. AND McKENRY, M.V., 2010. Incidence and reproduction of *Meloidogyne incognita* on vegetable crop genotypes. *Pakistan J. Zool.*, **42**:135-141.
- ANWAR, S.A. AND McKENRY, M.V., 2012. Incidence and population density of plant-parasitic nematodes infecting vegetable crops and associated yield losses. *Pakistan J. Zool.*, 44:327-333.
- ANWAR, S.A., 1995. Influence of *Meloidogyne incognita*, *Paratrichodorus minor* and *Pratylencus scribneri* on root-shoot growth and carbohydrate partitioning in

tomato. Pakistan J. Zool., 27:105-113.

- ANWAR, S.A., AKHTAR, M.S. AND TAHIR, A., 1992. Plant parasitic nematode problems of some field, vegetable, fruit and ornamental crops in the Punjab, Pakistan-II. *Proc. Parasitol.*, 14:86-98.
- ANWAR, S.A., ZIA, A. AND NAZIR, J., 2009. *Meloidogyne incognita* infection of five weeds. *Pakistan J. Zool.*, **1**:95-100.
- ANWAR, S.A., ZIA, A., HUSSAIN, M. AND KAMRAN, M., 2007. Host suitability of selected plants to *Meloidogyne incognita* in the Punjab, Pakistan. *Int. J. Nemtol.*, 17:144-150.
- DAVIS, R.F., EARL, H.J. AND TIMPER, P., 2003. Interaction of root-knot nematode stress and water stress in cotton. Univ. Georgia Cotton Res. Ext. Rep., pp. 312-
- DEVRAN, Z., SOGUT, M.A., GOZEL, U., TOR, M. AND ELEKCIOGLU, I.H., 2008. Analysis of genetic variation between populations of *Meloidogyne* spp. from Turkey. *Russian J. Nematol.*, 16:143-149.
- DI VITO, M.N. AND CARELLA, A., 1985. Population densities of *Meloidogyne incognita* and yield of *Capsium annum. J. Nematol.*, 17:45-49.
- ELEKCIOGLU, I.H. AND UYGUN, N., 1994. Occurrence and distribution of plant parasitic nematodes in cash crops in eastern Mediterranean region of Türkiye. Proc. of 9th Congress of the Mediterranean Phytopathological Union-Kuşadası-Aydın-Türkiye.
- FASSULIOTIS, G., 1970. Resistance of *Cucumis* spp. to the root-knot nematode, *Meloidogyne incognita acrita*. J. *Nematol.*, 2:174-178.
- HARTMAN, K.M. AND SASSER, J.N., 1985. Identification of *Meloidogyne* species on the basis of differential host test and perineal pattern morphology. In: *An advanced treatise on* Meloidogyne (eds. K.R. Barker, C.C. Carter and J.N. Sasser. *Vol. 2. Methodology*. North Carolina State University Graphics, Raleigh, **pp**. 69-77.
- HOLBROOK, C.C., KNAUFT, D.A. AND DICKSON, D.W., 1983. A technique for screening peanut for resistance to *Meloidogyne incognita*. Pl. Dis., 57:957-958.
- JEPSON, S.B., 1987. *Identification of root-knot nematodes* (Meloidogyne *species*). CAB International, Wallingford, UK.
- KAMALWANSHI, R.S., ANIS, K. AND SRIVASTAVA, A.S.I., 2004. Reaction of tomato germplasm against root-knot nematode, *Meloidogyne incognita*. Ind. J. Nematol., 34:94-95.
- KAUR, D.N., SHARMA, S.K. AND SULTAN M.S., 2010. Identification of nematode problems in polyhouses / nethouses in Punjab. *Pl. Dis. Res.*, 25:98.
- MADULU, J. AND TRUDGILL, D. L. 1994 Influence of temperature on *Meloidogyne javanica*. *Nematologica*, 40:230 – 243
- MAIN, C.E. AND GURTZ, S.K., 1989. Estimates of crop losses in North Carolina due to plant diseases and

*nematodes*. Dept. of Plant Path. Special Pub. No. **8**, NC State Univ., Raleigh, NC.

- McKENRY, M.V. AND ROBERTS, P.A., 1985. *Phytonematology study guide on nematodes and nematicides.* Division of Agriculture and Natural Resources, University of California.
- MELAKEBERHAN, H. AND WEBSTER, J.M., 1993. The phenology of plant-nematode interaction and yield loss. In: *Nematode interactions* (ed. M.M. Khan). Chapman & Hall. London, UK.
- NAGESH, M. AND AKIRAM, T.J., 2004. Root-knot nematode problem in polyhouse roses and its management using dazomet, neem cakeand pochonia chlamydosporia (*Verticillium Chlamydosporium*). J. Ornamen. Hort., 7: 147-152.
- PARK, S.D., KHAN, Z., YEON, K. AND KIM, Y.H., 2005. A survey for plant-parasitic nematodes associated with strawberry (*Fragaria ananassa* Duch.) Crop in Korea. *Plant Pathol. J.*, 21:387-390.
- PIEDRA, B.A., GARCÍA-ÁLVAREZ, A., DÍEZ-ROJO, M.A., ROS, C., FERNÁNDEZ, P., LACASA, A. AND BELLO, A., 2006. Use of pepper crop residues for the control of root-knot nematodes. *Biores. Technol.*, 98: 2846-2851.
- POTTER, J.W. AND OLTHOF, T.H.A., 1993. Nematode pest of vegetable crops. In: *Plant parasitic nematodes in temperate agriculture* (eds. K. Evans, D.L. Trudgill and J.M. Webster), CAB International, Wallingford, UK, pp. 171-207.
- QUESENBERRY, K.H., BALTENSPERGER, D.D., DUNN, R.A., WILCOX, C.J. AND HARDY, S.R., 1989.

Selection for tolerance to root-knot nematodes in red clove roots. *Nemtologica*, **33**:335-342.

- SHARMA, A., HASEEB, A. AND ABUZAR, S., 2004. Screening of field pea (*Pisum sativum*) selections for their reactions to root-knot nematode (*Meloidogyne* incognita). J. Zhejiang Univ. Sci., 7:209–214.
- SIKORA, R.A. AND FERNANDEZ, E., 1990. Nematode parasites of vegetables. In: *Plant parasitic nematodes in subtropical and tropical agriculture* (eds. M. R. Luc, A. Sikora and J. Bridge). CAB Bioscience, Egham, UK, pp. 319-392.
- ST. AMAND, P.C. AND WEHNER, T.C., 1997. Crop loss to 14 diseases in cucumber in North Carolina for 1983 to 1988. Cucurbit Genet. Coop. Rep., 14:15-17.
- TRUDGILL, D.L., 1995 An assessment of the relevance of thermal time relationships to nematology. *Fund. appl. Nematol.*, 18:407 - 417
- VAKALOUNAKIS, D.J., 1999. Alternatives to methyl bromide for the control of fungal diseases of greenhouse cucumbers in Greece. Proceedings of 3rd Int. Workshop on Alternatives to Methyl Bromide for the Southern European Countries, Heraklion, Crete, Greece, pp. 14– 18.
- WALTERS, S.A. AND WEHNER, T.C., 1997. Lucia, Manteo, and Shelby root-knot nematode-resistant cucumber inbred lines. *Hort. Sci.*, 32:1301-1303.
- WILLIAMSON, V.M. AND HUSSEY, R.S., 1996. Nematode pathogenesis and resistance in plants. *Plant Cell*, 8:1735-1745.

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