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# Distribution and association of root-knot nematodes (*Meloidogyne* spp.) with tomato crop in Faisalabad district

A. Zia[†], K. Zia[*], S.A. Anwar[**] and M. Iqbal

*Nematology Unit*, *Department of Biology*, *Ghent University*, *Ghent*, *Belgium*
[*]*Department of Agronomy*, *University of Agriculture*, *Faisalabad*, *Pakistan*
[**]*Institute of Agricultural Sciences*, *University of the Punjab*, *Lahore*, *Pakistan*

[†]Corresponding author email: amjadzia1877@gmail.com

## Abstract

A detailed survey of tomato fields located in different parts of Faisalabad district was conducted to identify the association of root-knot nematode (RKN) species with tomato crop and to assess their infestation. Hundred and sixty-one samples from 23 locations scattered over the vegetable growing areas of four tehsils of Faisalabad district including Faisalabad, Chak Jhumra, Jaranwala and Samundri. The results showed that 87% of the tomato fields were infested. The incidence of RKN ranged from 0 to 100% with an average of 36%. The galling index (GI) was ranged between 0 to 5 with a mean of 3. RKN incidence and GI varied from field to field. The RKN incidence was 45%, 34.3%, 28.6% and 10.71%, in the tomato growing areas of Faisalabad, Jaranwala, Samundri and Chak Jhumra, respectively. *Meloidogyne incognita* and *M. javanica* were found singly or in combination among the tomato root samples. The presence of *M. incognita* was 75.8% and *M. javanica* 20.2%.

**N**ematodes are microscopic round worms found in many habitats and the most abundant organisms in soil on earth. Most are beneficial members of their ecosystems, but a few are economic parasites of plants and animals. Several plant parasitic nematodes cause problems in landscape, garden and crop plants in Pakistan as well as in world. The most widespread, economically important and dominant group of plant parasitic nematodes are the root-knot nematodes (Kayani *et al*., 2012). Root-knot nematodes (RKN) are the most damaging nematode pests of field and vegetable crops because of their ability to feed on a range of plants including field and vegetable crops, ornamentals, fruit trees and weeds (Anwar *et al*., 2007; Anwar & McKenry, 2010). RKN is one of the major pathogen of tomatoes worldwide and limited fruit production (Sikora & Fernandez, 2005). RKN feeding results in cellular, structural and metabolic changes in plant tissues. These physical and physiological changes to the plant can reduce quality and crop yield (Anamika *et al*., 2010; Anwar *et al*., 2006). The tomato is an important vegetable crop cultivated on an area of about 52.2 thousand hectares in 2011 and total annual production is about 529.6 thousand tonnes in Pakistan. Tomato average yield in Pakistan is extremely low as compared to other countries of the world.

There can be several reasons for this low yield such as change in climatic conditions, primitive methods of cultivation, fungal, bacterial, viral and nematode diseases. Among diseases, root-knot nematode is most destructive one and tremendously reduces both quantity and quality of tomato fruit (Sikora & Fernandez, 2005). The data on association of *Meloidogyne incognita* with tomato crop in vegetable growing areas is limited (Khan *et al*., 2006). Therefore, this study was carried out to assess the level of infestation in Faisalabad district.

## Materials and Methods

**Sampling sites:** A survey was conducted to assess the incidence of root-knot nematode infestation in four tehsils of Faisalabad district

including Faisalabad, Chak Jhumra, Jaranwala and Samundri (Fig. 1). In tehsil Faisalabad, thirteen sampling sites at growers fields and Research Institute located at 241RB Chanchal Singh Wala, 295RB AARI, 30JB, 32JB, 55JB Lahlan, 57JB Ghiala, 58JB Bagh, 59JB Natho Chack, 61JB, Square # 9 University of Agriculture Faisalabad (UAF), Extension area UAF, Vegetable area UAF and Pul Dingru. In Chak Jhumra tehsil, four sampling sites were selected from grower fields namely Sikandarpur, Panwa, Khiwa and Dehran. From Jaranwala tehsil, five sampling sites were selected from growers fields namely 65GB, 66GB Surjanpur, 67GB, 69GB Malki and 105GB Bangiawala. Whereas from Samundri tehsil, only one sampling site located at Pholan was selected. Seven (replicates) samples of root and soil were taken from each site. One hundred and sixty-one samples from all selected sites were brought to the laboratory of Nematology, Department of Plant Pathology, University of Agriculture, Faisalabad for nematode studies.

**Collection of soil and root samples:** Tomato roots and soil samples were collected from the growers fields located at major vegetable growing areas of Faisalabad district to estimate the root-knot nematode population associated with roots and soil. Using an Oak tube, 12 cm depth root and soil samples around the plant rhizosphere was collected and mixed sample were made of 20 core samples per mixed sample collected from each field. The samples were protected from heat and light so as not to deteriorate the sample material and put in to the polythene bags separately. These samples were immediately brought to laboratory and placed in refrigerator to keep the material fresh.

**Processing of soil and root samples**

**Soil samples:** Each sample was thoroughly mixed and a 100 cc composite sample was processed through a 325 mesh sieve (pore size = 17 micrometer), followed by Modified Baermann Funnel extraction to collect $J_2$ after 3 days and counted under 40x magnification. The soil was assessed for soil type.

**Root samples:** Samples of roots were removed from each plant. Roots were washed free of soil, blotted onto paper, damp-dried and weighed. After weighing, the root system was rated for galling index (GI) by 0 to 5 scale (Anwar *et al*., 2007), where, 0 = no galls, 1 = 1-2 galls; 2 = 3-10 galls; 3 = 11-30 galls; 4 = 31-100 galls and 5 = > 100 galls per root system. The entire root system was diced, chopped and 20 g composite root sample was incubated in a mist chamber for 5 days to hatch the eggs. The numbers of $J_2$ per root system was determined by using a stereomicroscope.

**Identification of *Meloidogyne* species:** *Meloidogyne* spp., were separated on the basis of perennial patterns. Perennial patterns of mature females of various root-knot nematode species were prepared (Jepson, 1987) which was collected from grower fields. The root tissue was teared apart with needle to remove adult females. The head and neck region of the nematode was excised and posterior was placed in a solution of 45% lactic acid to remove all body tissues. Then, the perennial pattern was trimmed and transferred to a drop of glycerin. Perennial patterns were examined to make the identification of nematode species from each sample. Data in terms of incidence (%), GI, nematode population in soil and roots was recorded.

**Statistical analysis:** The incidence of infestation was assessed by the formula as Number of samples with RKN/Total number of samples x 100. Nematode population data was subjected to analysis of variance using SAS (SAS Institute, Cary NC). Duncan's Multiple Range Test at (P = 0.05) was applied to observe the significant differences between the values.

## Results

**Field observation:** Hundred and sixty-one samples collected from 23 tomato fields scattered over the vegetable production area of Faisalabad district showing the root-knot nematode incidence and GI (Fig. 1). The results shows that

87 percent of the tomato fields were infested with root-knot nematode (Fig. 2).

**Incidence and soil type:** The incidence of root-knot nematode ranged from 0 to 100% with an average of 36%. The incidence was 45%, 34.3%, 28.6% and 10.71%, in the tomato growing area of Faisalabad, Jaranwala, Samundri and Chak Jhumra, respectively. Maximum incidence was recorded from three fields located at the campus of UAF, Faisalabad, which was 70 to 100% in the sandy loam soils. Incidence between 40 to 60% was observed with medium loam soil in 295RB AARI, 58JB Bagh and 59JB Natho Chack from Faisalabad and 66GB Surjanpur and 69GB Malki from Jaranwala. Similarly, clay loam soil showed incidence between 1 to 30% in eleven locations (241RB Chanchal Singh Wala, 55JB Lahlan, 57JB Ghiala & Pul Dingru in Faisalabad; Sikandarpur, Panwa and Dehran in Chak Jhumra; 65GB, 67GB and 105GB Bangiawala in Jaranwala and Pholan in Samundri). There was no root-knot nematode infestation found in clay loam soil fields located at 30JB and 32JB in Faisalabad and Khiwa in Chak Jhumra (Table 1).

**Gall index:** The galls were present on the tomato roots (Fig. 2). The gall index was 0 to 5 with a mean of 3. The gall index was 2.69, 2.4, 2 and 0.75 in the tomato growing area of Faisalabad, Jaranwala, Samundri and Chak Jhumra, respectively. Two localities, Square # 9 UAF and Extension area UAF in Faisalabad had GI of 5. Four localities; 61JB, 59JB Natho Chack, 58JB Bagh and Vegetable area UAF in Faisalabad had GI of 4. GI of 3 was observed at three location; 295RB AARI in Faisalabad and 66GB Surjanpur and 69GB Malki in Jaranwala while, six localities 55JB Lahlan and Pul Dingru in Faisalabad; 65GB, 67GB and 105GB Bangiawala in Jaranwala and Pholan in Samundri had GI of 2. Five localities i.e., 241RB Chanchal Singh Wala and 57JB Ghiala in Faisalabad and Sikandarpur, Panwa and Dehran in Chak Jhumra had GI of 1 (Table 1).

**Identification of *Meloidogyne* spp.:** The survey of tomato growers fields located in Faisalabad district revealed the occurrence of mixture of *M. incognita* and *M. javanica* singly or in combination at various locations of Faisalabad. In overall distribution, *M. incognita* was the most prevalent (75.8%) followed by *M. javanica* (20.2%).

**Root-knot nematode population**

**Nematode population in root:** Maximum root-knot nematode population in roots was observed in Extension area and Square # 9, UAF that was more than 300 $J_2$. 61JB, 59JB Natho Chack, 58JB Bagh and Vegetable area of UAF contained $J_2$ population between 200 to 300 in Faisalabad tehsil. No root-knot nematode root population was observed in samples collected from 30JB and 32JB in Faisalabad and Khiwa in Chak Jhumra tehsil. Other sampling sites had intermediate root-knot nematode population (Table 1). Four locations including Dehran, Panwa and Sikandarpur in Chak Jhumra and 57JB Ghiala in Faisalabad had between 1 to 100 $J_2$ populations. 241RB Chanchal Singh Wala, 295RB AARI, 55JB Lahlan and Pul Dingru in Faisalabad; in Jaranwala, 65GB, 66GB Surjanpur, 67GB, 69GB Malki and 105GB Bangiawala; Pholan in Samundri were shown 101 to 200 $J_2$ population in roots (Table 1).

**Soil population:** Root-knot nematode was not observed in soil population at 30JB and 32JB in Faisalabad and Khiwa in Chak, Jhumra. In 100 cc soil, 1 to 100 $J_2$ were observed at ten (10) locations i.e., three in Faisalabad (241RB Chanchal Singh Wala, 55JB Lahlan and 57JB Ghiala); three in Chak Jhumra (Dehran, Panwa and Sikandarpur); three in Jaranwala (66GB Surjanpur, 67GB and 105GB Bangiawala) and one in Samundri (Pholan). 295RB AARI, 58JB Bagh, 59JB Natho Chack, 61JB, Vegetable area, UAF and Pul Dingru in Faisalabad and 65GB and 69GB Malki in Jaranwala showed $J_2$ population 101 to 200. However, maximum root-knot nematode population was observed in Squre # 9 UAF and Extension area UAF which was more than 200 $J_2$ (Table 1).

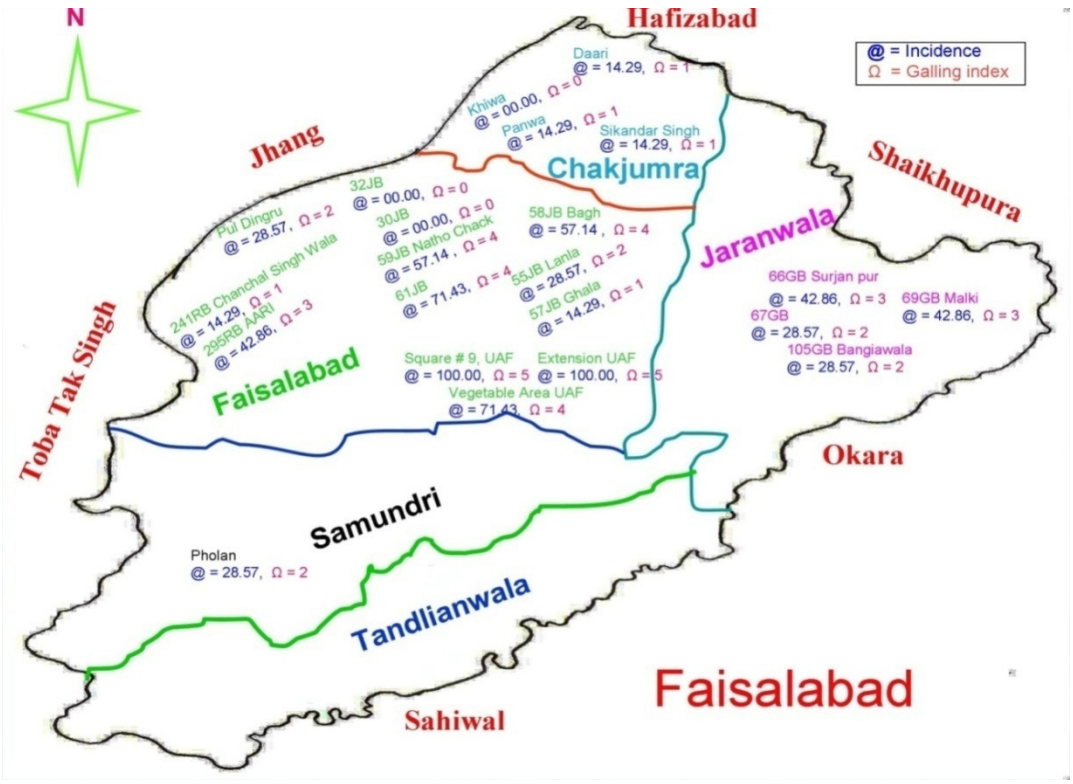


**Fig. 1.** Map showing root-knot nematode incidence and galling indices at 23 localities of Faisalabad district.

**Fig. 2.** Root galls induced on tomato roots by *Meloidogyne incognita*.

**Table 1. Nematode population, gall index and incidence of *Meloidogyne incognita* in relation to soil type in 23 tomato fields of Faisalabad district.**

| Tehsil | Locality | Root-knot nematode population | | Gall index | Incidence (%) | Soil type |
|---|---|---|---|---|---|---|
| | | Roots | Soil | | | |
| Faisalabad | 241RB Chanchal Singh Wala | 102fg | 95bcd | 1 | 14.3 | Clay Loam |
| | 295RB AARI | 197bcd | 117bc | 3 | 42.9 | Medium loam |
| | 30JB | 0h | 0d | 0 | 0.00 | Clay loam |
| | 32JB | 0h | 0d | 0 | 0.00 | Clay loam |
| | 55JB Lahlan | 146cdefg | 81cd | 2 | 28.6 | Clay loam |
| | 57JB Ghiala | 97fg | 65cd | 1 | 14.3 | Clay loam |
| | 58JB Bagh | 209bc | 137bc | 4 | 57.1 | Medium loam |
| | 59JB Natho Chack | 213bc | 155bc | 4 | 57.1 | Medium loam |
| | 61JB | 257b | 198ab | 4 | 71.4 | Sandy loam |
| | Square # 9 UAF | 369a | 257a | 5 | 100.0 | Sandy loam |
| | Extension area UAF | 387a | 249a | 5 | 100.0 | Sandy loam |
| | Vegetable area UAF | 203bc | 124bc | 4 | 71.4 | Sandy loam |
| | Pul Dingru | 134cdefg | 109bc | 2 | 28.6 | Clay loam |
| Chak Jhumra | Sikandarpur | 95fg | 56cd | 1 | 14.3 | Clay loam |
| | Panwa | 79g | 54cd | 1 | 14.3 | Clay loam |
| | Khiwa | 0h | 0d | 0 | 0.00 | Clay loam |
| | Dehran | 96fg | 54cd | 1 | 14.3 | Clay loam |
| Jaranwala | 65GB | 146cdfg | 104bcd | 2 | 28.8 | Clay loam |
| | 66GB Surjanpur | 187bcde | 89cd | 3 | 42.9 | Medium loam |
| | 67GB | 113efg | 92cd | 2 | 28.6 | Clay loam |
| | 69GB Malki | 167cdefg | 107bc | 3 | 42.9 | Medium loam |
| | 105GB Bangiawala | 114defg | 85cd | 2 | 28.6 | Clay loam |
| Samundri | Pholan | 132cdefg | 93cd | 2 | 28.6 | Clay loam |

Means within a column sharing the same letters are not significantly different from each other at $P = 0.05$.

## Discussion

The survey results indicated that root-knot nematode is widespread in tomato growing areas of Faisalabad district but the incidence was variable among localities. The differences in root-knot incidence from locality to locality and field to field might be related to soil types as soil variation exists among fields (Starr *et al*., 1993; Anwar *et al*., 2007). The sandy soils have generally more penetration, development and reproduction of root-knot nematode (Ogbuji, 2004). Typically, crop production areas with soils containing higher percentages of sand had higher population of *M. incognita* compared to those fields with low sand contents (Lawrence *et al*., 1997). These hidden enemies are causing huge losses by attacking the root system. Nematode feeding reduces the flow of nutrients and water into the plant and thus reduces the yield of agricultural crops (Sikora & Fernandez, 2005). In addition, nematode infestations deteriorate plants roots, making them more susceptible to other stress factors such as nutritional deficiencies, water, heat and to other disease-causing organisms (Khan *et al*., 2007).

Damaged roots may also make them more susceptible to infection by fungi, bacteria and/or viruses (Powell, 1971). The farmers are practicing intensive cropping strategy and do not leave the land fallow, which favor the development of the pests. Moreover, farmers could not afford the application of nematicides

due to non-availability and high cost of applications. Crops grown in rotation with tomato are mostly susceptible (Anwar *et al*., 2007). As a long-term strategy for managing root-knot nematodes, a research program should be started to screen new varieties imported from other countries, or from the local breeding program for resistance to this pest.

Our findings provided the information that root-knot nematode is destructive pest and widely distributed on tomato in Faisalabad district. Infestation of RKN increases with the increase of sand contents and favorable soil is sandy loam soils.

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