

INCIDENCE AND SEVERITY OF ROOT-KNOT NEMATODES (*MELOIDOGYNE* SPP.) ON CUCUMBER IN DISTRICT RAWALPINDI

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ABSTRACT

A survey of root-knot nematodes, *Meloidogyne* spp., infecting cucumber was conducted in district Rawalpindi. The root-knot nematodes (*Meloidogyne* spp.) were recorded with varying degrees in cucumber fields of seven tehsils of district Rawalpindi. The highest disease incidence of 31.11% was recorded in tehsil Taxila followed by 26.11% in Murree. Of the seven tehsils, minimum incidence of 6.94% was recorded from tehsil Kotlisattian. Differences in root-knot severity were also observed among tehsils of the district. The maximum mean severity (5.39) was observed in tehsil Taxila while it was the minimum in tehsil Kotlisattian. In overall distribution, *M. incognita* (74.17%) was the most prevalent followed by *M. javanica* (21.69%), *M. hapla* (1.81) and *M. arenaria* (2.33%). The percentage of *M. incognita* was the highest in tehsil Kahuta and Taxila; being 90.00 and 88.33% respectively and was the lowest (47.5%) in tehsil Gujjar Khan. As regards *M. javanica*, it was found in the highest degree in tehsil Gujjar Khan and was the lowest in tehsil Kahuta. *M. hapla* was recorded from only Murree and Kotlisattian tehsils while *M. arenaria* was observed from Rawalpindi and Gujjar Khan tehsils.

Keywords: *Meloidogyne incognita*, *M. javanica*, *M. arenaria*, *M. hapla*, incidence, severity

INTRODUCTION

In Pakistan, the yield of vegetables is relatively lower, for which there are many constraints including prevalence of diseases caused by different pathogens (Ahmad *et al.*, 2012; Arain *et al.*, 2012). Among various pathogens responsible for the low yield, the root-knot nematodes are of considerable economic importance (Hussain *et al.*, 2011a; Kayani *et al.*, 2012a) and cause annual losses in tropics to an extent of 22 per cent (Sasser, 1979). In addition, these parasites also interact with other disease causing organisms to produce disease complexes (Begum *et al.*, 2012) and break down resistance against other pathogens and reduce plant tolerance to environmental stress (Taylor, 1979). Root-knot nematodes are known to occur throughout the world. Their occurrence has been reported from all the continents including Asia, Europe, North and South America and Australia. The four commonly occurring root-knot species are *M. incognita*, *M. javanica*, *M. hapla* and *M. arenaria* and have been reported from almost all the areas of tropical and temperate regions. However, *M. hapla* is mainly confined to cooler areas. Since its recognition, various surveys were conducted worldwide to ascertain its occurrence and incidence on various crops. Wheeler *et al.* (2000) found *M. incognita* in 39% and 43% of the fields in 1995 and 1996, respectively in the High Plains of Texas. Dautova and Gommers (2000) surveyed the occurrence of *Meloidogyne* in several areas of the Republic of

Macedonia. *M. incognita* and *M. javanica* were found to be predominant with incidences of 47.9% and 35.6% respectively while *M. arenaria* and *M. hapla* with incidences of 13.7% and 2.7% respectively were found sporadically. del Prado-Vera *et al.* (2001) sampled 47 localities from 18 states of Mexico. A total of 56 populations were obtained from which 60.7% belonged to *M. incognita*, 21.4% to *M. arenaria*, 12.5% to *M. javanica* and 5.3% to *M. hapla*. Khanzada *et al.* (2002) conducted a survey of different eggplant fields in order to observe the incidence of root-knot nematodes and recorded the maximum disease incidence in Mission (62%) followed by Bhawal Zanoor (56%) and minimum in Kamil Jamali (26%). Barbosa *et al.* (2004) reported 70% of the plantations infected with *M. exigua* from the State of Rio de Janeiro of Brazil. Olowe (2004) revealed the occurrence of *M. incognita*, *M. javanica* and *M. arenaria* singly or in combination in all the cowpea farms sampled in Nigeria. The overall distribution of *M. incognita* was 51.8%, *M. javanica* 44.1% and that of *M. arenaria* was 4.1%. Bhosle *et al.* (2004) conducted a survey in the Parbhani district of Maharashtra, India, in the summer season, to determine the prevalence of plant parasitic nematodes in okra fields and found that *M. incognita* was the most predominant nematode species associated with okra. Increased information on incidence and severity of root-knot nematodes in cucumber production areas is important for

sustainable production of cucumber. Plant parasitic nematodes, in Pakistan, have received little attention and only a few surveys have been made in the past (Brown, 1962; Kafi, 1963; Saeed and Ashrafi, 1973; Ahmad and Khan, 1973; Khan *et al.*, 2005). There is limited information regarding the association of root-knot nematodes with cucumber in the country. Therefore, the objective of present studies was to conduct a survey to determine and document the occurrence, prevalence and intensity of root-knot nematodes on cucumber cultivations in the vegetable growing areas of district Rawalpindi of Pothwar region of the Punjab province of the Pakistan.

MATERIALS AND METHODS

Incidence and severity of root-knot nematodes: A survey of cucumber fields located in different randomly selected localities of the seven tehsils of district Rawalpindi of Pothwar region of the Punjab province of Pakistan (Table 1) was conducted for the determination of incidence and severity of root-knot nematodes. The samples were taken during the months of April and May when the plants were at flowering stage. From each locality, three fields of cucumber were randomly selected. From each cucumber field, 20 plants were selected after every 10 steps following a zigzag pattern. The selected plants were carefully uprooted up to 15-30 cm soil depth with the help of a trowel. The soil adhering to the root system was gently removed and roots were observed for root-knot nematodes infection (presence or absence of galls). The root systems of individual plants were rated following the galling index developed by Bridge and Page (1980) for the determination of severity of root-knot nematodes. The infected roots along with soil were put into polythene bags, labeled properly and brought to the laboratory of Plant Pathology Department, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi for identification of root-knot nematode species. The incidences of root-knot nematodes of individual cucumber fields were determined as follows.

$$\text{Incidence (\%)} = \frac{\text{Total number of infected plants}}{\text{Total number of observed plants}} \times 100$$

Identification of *Meloidogyne* Species: Root-knot nematodes (*Meloidogyne* species) were identified on the basis of female perineal patterns described by Taylor and Netschler (1974). Mature females of root-knot nematodes were dissected out from the infected cucumber roots and placed in watch glass containing distilled water. The live mature females were then picked up with fine bristle and were placed in plastic Petri dish containing 45 percent lactic acid and were left for two hours. The posterior

end then was cut off with a fine needle and the body tissues were removed by lightly brushing the inner surface of the cuticle with a flexible bristle. When all the tissues were removed, the cuticle was transferred to a drop of glycerin where it was carefully trimmed, the piece of cuticle containing vulval portion with the typical perineal pattern was then transferred to a drop of glycerin on a micro slide, a cover slip was applied and sealed with nail polish, and was observed under microscope. The perineal pattern was compared with standard diagrams and *Meloidogyne* species was identified. In this way perineal patterns of 40 females were prepared from each infected cucumber field and the distribution of each *Meloidogyne* species in each tehsil was calculated.

RESULTS

Incidence of Root-Knot Nematodes: The root-knot nematodes were recorded with varying degrees in cucumber fields of seven tehsils of district Rawalpindi. The highest disease incidence of 31.11% was recorded in tehsil Taxila followed by 26.11% in Murree. Of the seven tehsils, minimum incidence of 6.94% was recorded from tehsil Kotlisattian as shown in Table 2. The individual incidences of root-knot nematodes in each locality of each tehsil of Rawalpindi district are given in Table 3.

Severity of Root-Knot Nematodes: Differences in root-knot severity were also observed among tehsils of district Rawalpindi. The maximum mean severity (5.39) measured in terms of galling index suggested by Bridge and Page (1980) was observed in tehsil Taxila while it was the minimum in tehsil Kotlisattian. The rest of the tehsils showed severity above 3 as shown in Table 2. The individual mean severities of root-knot nematodes in each locality of each tehsil of district Rawalpindi are given in Table 3.

Occurrence of Root-Knot Nematodes Species: The different species of *Meloidogyne* associated with cucumber in different tehsils are given in Table 4. Of all the associated species of root-knot nematodes, *M. incognita* constituted 74.17%, *M. javanica* 21.69%, *M. arenaria* 1.81% and *M. hapla* 2.33%. In district Rawalpindi, the percentage of *M. incognita* was the highest in tehsil Kahuta and Taxila; being 90.00 and 88.33% respectively and was the lowest (47.5%) in tehsil Gujjar Khan. As regards *M. javanica*, it was found in the highest degree in tehsil Gujjar Khan and was the lowest in tehsil Kahuta. *M. hapla* was recorded from only Murree and Kotlisattian by 9.33 and 3.33% respectively. On the other hand *M. arenaria* was recorded from Rawalpindi and Gujjar Khan tehsils as shown in Table 4. Association of different *Meloidogyne* species with cucumber in each locality

of each tehsil of district Rawalpindi is given in Table 5.

DISCUSSION

The results of the present survey confirmed that root-knot nematodes are widely distributed in the

district. Similar results were also reported by Lamberti *et al.* (1975), Bhatti and Jain (1977), Khan *et al.* (2005 and 2006), Shahid *et al.* (2007). The results of these scientists confirmed the present findings regarding the prevalence of plant parasitic nematodes and occurrence of *Meloidogyne* species

Table 1. The localities of tehsils of district Rawalpindi surveyed for incidence and severity of root-knot nematodes on cucumber.

Tehsil	Localities
Rawalpindi	Gurikh Pur, Bagga Sheikhan, Adyala, Meelam, Dhoke Hamida, Dhoke Gujran
Gujjar khan	Badana, Jabbar, Doltala, Toppian, Sukkoo, Sang
Kallar Saydian	Sir Soba Shah, Sumot, Darkali, Balakhar, Takal, Bishendot
Taxila	Usmankhattar, Garhiafghanan, Malikabad, Banbola, Gangoobadhar, Thattakhalil
Kahuta	Pharwala, Channi, Nara, Mathore, Dakhali, Bagh Jamberi
Murree	Angori, Bunn, Charyan, Kamalabad, Saamli, Tret
Kotlisattian	Kamra, Battian, Santhanwali, Lehtrar, Mallot, Kotli

Table 2. Tehsil wise incidence and severity of root-knot nematodes in district Rawalpindi.

Tehsil	Mean incidence	Mean intensity
Rawalpindi	17.78 ± 3.16	3.33 ± 0.60
Gujjar khan	21.67 ± 3.98	3.61 ± 0.76
Kallar Syadan	23.33 ± 3.45	4.83 ± 0.62
Taxila	31.11 ± 4.34	5.39 ± 0.64
Kahuta	24.17 ± 4.51	3.44 ± 0.67
Murree	26.11 ± 3.85	4.33 ± 0.50
Kotlisattian	6.94 ± 1.62	1.53 ± 0.39
Average	21.83 ± 1.51	3.79 ± 0.25

Table 3. Incidence and severity of root-knot nematodes in different localities of district Rawalpindi.

Tehsil	Locality	Mean incidence	Mean severity
Rawalpindi	Gurikh Pur	18.33 ± 4.41	4.67 ± 0.88
	Bagga Sheikhan	28.33 ± 4.41	5.00 ± 0.58
	Adyala	23.33 ± 4.41	4.33 ± 1.20
	Meelam	31.67 ± 4.41	5.33 ± 1.20
	Dhoke Hamida	0.00 ± 0.00	0.00 ± 0.00
	Dhoke Gujran	5.00 ± 5.00	0.67 ± 0.67
	Average	17.78 ± 3.16	3.33 ± 0.60
Gujjar Khan	Badana	31.67 ± 6.01	4.00 ± 1.15
	Jabbar	31.67 ± 6.01	4.67 ± 1.86
	Doltala	31.67 ± 4.41	6.33 ± 0.88
	Toppian	35.00 ± 2.89	6.67 ± 1.45
	Sukkoo	0.00 ± 0.00	0.00 ± 0.00
	Sang	0.00 ± 0.00	0.00 ± 0.00
	Average	21.67 ± 3.98	3.61 ± 0.76
Kallar Syedan	Sir Soba Shah	0.00 ± 0.00	0.00 ± 0.00
	Sumot	35.00 ± 8.66	5.00 ± 0.58
	Darkali	15.00 ± 2.89	5.67 ± 0.88
	Balakhar	30.00 ± 2.89	6.00 ± 1.53
	Takal	23.33 ± 4.41	6.33 ± 1.20
	Bishendot	36.67 ± 3.33	6.00 ± 0.58
	Average	23.33 ± 3.45	4.83 ± 0.62
Taxila	Usmankhattar	48.33 ± 4.41	7.00 ± 0.58
	Garhiafghanan	40.00 ± 2.89	6.33 ± 0.33
	Malikabad	0.00 ± 0.00	0.00 ± 0.00
	Banbola	30.00 ± 2.89	7.00 ± 0.58
	Gangoobadhar	31.67 ± 6.01	6.00 ± 1.00

	Thattakhalil	46.67 ± 10.14	6.33 ± 0.88
Kahuta	Average	31.11 ± 4.34	5.39 ± 0.64
	Pharwala	35.00 ± 2.89	5.67 ± 0.88
	Channi	0.00 ± 0.00	0.00 ± 0.00
	Nara	41.67 ± 1.67	5.33 ± 1.20
	Mathore	25.00 ± 2.89	5.33 ± 1.20
	Dakhali	43.33 ± 6.01	4.33 ± 0.88
	Bagh Jamberi	0.00 ± 0.00	0.00 ± 0.00
	Average	24.17 ± 4.51	3.44 ± 0.67
Murree	Angori	36.67 ± 6.01	4.67 ± 0.33
	Bunn	30.00 ± 2.89	5.33 ± 0.88
	Charyan	38.33 ± 7.26	5.67 ± 0.33
	Kamalabad	0.00 ± 0.00	0.00 ± 0.00
	Saamli	23.33 ± 6.01	5.33 ± 0.33
	Tret	28.33 ± 11.67	5.00 ± 0.58
	Average	26.11 ± 3.85	4.33 ± 0.50
	Kotli Sattian	Kamra	10.00 ± 2.89
Battian		0.00 ± 0.00	0.00 ± 0.00
Santhanwali		6.67 ± 4.41	2.00 ± 1.15
Lehtrar		15.00 ± 2.89	3.33 ± 0.88
Mallot		0.00 ± 0.00	0.00 ± 0.00
Kotli		10.00 ± 2.89	2.33 ± 0.88
Average		6.94 ± 1.62	1.53 ± 0.39
Overall mean		21.83 ± 1.51	3.79 ± 0.25

Table 4. Tehsil wise distribution of *Meloidogyne* species in district Rawalpindi.

Tehsil	Percentage of <i>Meloidogyne</i> species			
	<i>M. incognita</i>	<i>M. javanica</i>	<i>M. hapla</i>	<i>M. arenaria</i>
Rawalpindi	58.67	36.67	0.00	4.67
Gujjar khan	47.50	40.83	0.00	11.67
Kallar Syadan	84.00	16.00	0.00	0.00
Taxila	88.33	11.67	0.00	0.00
Kahuta	90.00	10.00	0.00	0.00
Murree	67.33	23.33	9.33	0.00
Kotlisattian	83.33	13.33	3.33	0.00
Average	74.17	21.69	1.81	2.33

Table 5. Distribution of *Meloidogyne* species in different localities of District Rawalpindi.

Tehsil	Locality	Percentage of <i>Meloidogyne</i> species			
		<i>M. incognita</i>	<i>M. javanica</i>	<i>M. hapla</i>	<i>M. arenaria</i>
Rawalpindi	Gurikh Pur	70.00	30.00	0.00	0.00
	Bagga Sheikhan	78.33	21.67	0.00	0.00
	Adyala	60.00	40.00	0.00	0.00
	Meelam	50.00	26.67	0.00	23.33
	Dhoke Hamida	0.00	0.00	0.00	0.00
	Dhoke Gujran	35.00	65.00	0.00	0.00
	Average	58.67	36.67	0.00	4.67
	Gujjar Khan	Badana	38.33	46.67	0.00
Jabbar		50.00	50.00	0.00	0.00
Doltala		50.00	31.67	0.00	18.33
Toppian		51.67	35.00	0.00	13.33
Sukkoo		0.00	0.00	0.00	0.00
Sang		0.00	0.00	0.00	0.00
Average		47.50	40.83	0.00	11.67
Kallar Syedan	Sir Soba Shah	0.00	0.00	0.00	0.00
	Sumot	75.00	25.00	0.00	0.00
	Darkali	71.67	28.33	0.00	0.00
	Balakhar	100.00	0.00	0.00	0.00
	Takal	73.33	26.67	0.00	0.00

	Bishendot	100.00	0.00	0.00	0.00
	Average	84.00	16.00	0.00	0.00
Taxila	Usmankhattar	76.67	23.33	0.00	0.00
	Garhiafghanan	100.00	0.00	0.00	0.00
	Malikabad	0.00	0.00	0.00	0.00
	Banbola	65.00	35.00	0.00	0.00
	Gangoobadhar	100.00	0.00	0.00	0.00
	Thattakhalil	75.00	25.00	0.00	0.00
	Average	88.33	11.67	0.00	0.00
Kahuta	Pharwala	80.00	20.00	0.00	0.00
	Channi	0.00	0.00	0.00	0.00
	Nara	100.00	0.00	0.00	0.00
	Mathore	80.00	20.00	0.00	0.00
	Dakhali	100.00	0.00	0.00	0.00
	Bagh Jamberi	0.00	0.00	0.00	0.00
	Average	90.00	10.00	0.00	0.00
Murree	Angori	73.33	26.67	0.00	0.00
	Bunn	71.67	28.33	0.00	0.00
	Charyan	46.67	31.67	21.67	0.00
	Kamalabad	0.00	0.00	0.00	0.00
	Saamli	45.00	30.00	25.00	0.00
	Tret	100.00	0.00	0.00	0.00
	Average	67.33	23.33	9.33	0.00
Kotli Sattian	Kamra	100.00	0.00	0.00	0.00
	Battian	0.00	0.00	0.00	0.00
	Santhanwali	65.00	35.00	0.00	0.00
	Lehtrar	100.00	0.00	0.00	0.00
	Mallot	0.00	0.00	0.00	0.00
	Kotli	68.33	18.33	13.33	0.00
	Average	83.33	13.33	3.33	0.00
Overall Mean		74.17	21.69	1.81	2.33

on vegetables. Among the four most species, two viz. *M. incognita* and *M. javanica* were found associated with cucumber in all the tehsils. *M. hapla* was recorded from tehsils of Murree and Kotlisattian while *M. arenaria* from tehsils of Rawalpindi and Gujjar khan. The above mentioned four species of root-knot nematodes are considered to be the most common species (Taylor *et al.*, 1982) and *M. incognita* being ranked first with respect to its geographical distribution and host range. In the present studies too, *M. incognita* was found to be the widely spread than the other three species in the study area. On global basis *M. incognita* has been reported to constitute about 47 percent of the total root-knot nematode populations (Sasser and Carter, 1985) and in the present survey the occurrence of this species was found to be 74.17 percent. The variations in species distributions may be attributed to soil characteristics and climate of this particular study area. The species has been predominantly found in other parts of the country (Ahmad and Saeed, 1981). The approximate distribution of *Meloidogyne* spp. in the soils of Pakistan is; *M. incognita*, 52%; *M. javanica*, 31%; *M. arenaria*, 8%; *M. hapla*, 7% and other species are about 2% (Maqbool, 1987). In the present survey *M. hapla* was isolated from tehsils having a cool climate.

Present studies are in conformity with those of Sasser and Carter (1985), Taylor *et al.*, (1982), who regarded *M. hapla* as a cool climate species. It is concluded from the present studies that cucumber is severely attacked by root-knot nematodes and *M. incognita* is the most predominant species, which warrant that strict control measures should be adopted for its management (Hussain *et al.*, 2011b; Kayani *et al.*, 2012b; Mukhtar *et al.*, 2013a, 2013b; Qureshi *et al.*, 2012; Rahoo *et al.*, 2011; Vagelas and Gowen, 2012).

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