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Incidence of root-knot nematode in winter weeds of tomato in Malakand division – Pakistan

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Abstract

This study was conducted to find the incidence of root knot nematode (*M. javanica*) in winter weeds of tomato at the root knot infested areas of Dargai and Jabban in Malakand division of Khyber Pakhtunkhwa province of Pakistan. For this purpose survey was conducted on 20 fields of each region and the fifteen weed plants were checked for galls caused by *M. javanica* on their roots. Weeds samples from these regions were bagged and labeled properly and brought to the Department of Weed Science, The University of Agriculture, Peshawar Pakistan for identification. The nematode was also identified by perennial pattern. Galls were observed on commonly occurring fifteen weeds. Incidence of *M. javanica* on *Cannabis sativa* was at the range of 9.9-11.9%, while it was 33.3% on *Digitaria sanguinalis*. The highest incidence of *M. javanica* were recorded on the roots of *Fumeria indica* (21.05-43.47%). Moreover the incidence of *M. javanica* on *Rumex crispus* and *Melilotus indica* was 22.72% and 22.13% respectively. However no galls (No incidence) of *M. javanica* were found on *Asphodelus tenuifolius*. Our finding demonstrates that *M. javanica* is a damaging pathogen of winter weeds of tomato in Malakand division of Khyber Pakhtunkhwa province of Pakistan.

Keywords: Galls, *M. javanica*, root knot nematode, incidence, winter weeds.

1. Introduction

Tomato (*Lycopersicon esculentum* L.) is one of the extensively consumed crops. It is grown as winter as well as summer vegetable all over the world [1]. Malakand division of Khyber Pakhtunkhwa (Kpk) Province of Pakistan, greatly contributes towards tomato production especially winter tomato and meets the demands of local markets and adjacent areas. However, this division is heavily infested with root knot nematodes [2]. However its soil and environmental conditions are suitable for the production of tomatoes [3]. Nematode parasitism of tomatoes is highly damaging and uncontrollable problem. Nematodes occur almost in all types of soil. In Pakistan, climate and sandy soil favor the development and activities of nematodes [4]. Most damaging obligate endoparasites are the root-knot nematodes (*Meloidogyne* sp) and cyst nematodes (*Heterodera* and *Globodera* sp.). Root-knot nematodes are one of the major causes of yield losses [5]. Crop losses of 13-38% have been reported in sub-tropics and tropics [6]. Root knot nematodes cause losses of US\$ 125 billion per annum. Root-knot nematode cause poor growth, yield, quality, and can break the resistance of host plant and expose them to other pathogens [7]. In Pakistan, 40% yield losses have been reported in tomato by root knot nematodes [8]. Weeds are present throughout the year, during and after the crop and serve as reservoir for plant pathogens and nematodes [9]. Common agricultural weeds are hosts of plant parasitic nematodes [10]. The root knot nematodes (RKN) survive and develop on weeds [11]. *Ageratum conyzoides*, *Amaranthus spinosus*, *Eleusine indica*, *Portulaca oleracea*, *Cyperus rotundus*, *Amaranthus* sp., *Chenopodium album* and *Digitaria* sp. have been reported as weed hosts of root-knot nematodes [12]. Keeping the importance of weeds in winter tomatoes as hosts and non-hosts, this project was initiated with the following objectives to survey the common weeds as hosts and non-hosts of *M. javanica* in winter tomatoes at Dargai and Jabban of Malakand division of Khyber Pakhtunkhwa Province of Pakistan.

2. Materials and methods

2.1 Survey of weed hosts of root knot nematodes

Extensive field survey was conducted in tomato growing fields of Dargai and Jabban area of Malakand, Khyber Pakhtunkhwa. For this purpose 20 fields were surveyed. In each field 5

areas of 2m² were selected for data collection in a zigzag pattern. Root knot incidence was assessed on the basis of characteristic symptom (galls) on the plant roots. The root gall severity was measured on 0 to 5 scales based on galling indices (GI) as 0-GI=0%, 1-GI=10-20%, 2-GI=21-50%, 3-GI=51-80%, 4-GI=81-100% and 5-GI=>100%.

The percent incidence was also calculated by using the following general formula:

$$\text{Percent Incidence} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

The absence of galls on the roots of weeds was an indicator for non-host weeds.

2.2 Samples collection

Host and non-host weeds along with infected tomato plants were collected during field survey in tomato growing fields of Dargai and Jabban area of Malakand, Khyber Pakhtunkhwa. Inoculum of *M. javanica* was prepared. Weeds were bagged, labeled properly and brought to the Department of Weed Science, The University of Agriculture, Peshawar, for identification [13].

2.3 Identification of *M. javanica*

Perennial pattern morphology was followed for the identification of *M. javanica*. Heavy galled roots with adult females were placed in tap water, in a Petri dish and crushed with the help of a needle in order to extract adult females. Posterior bodies of the females were cut, discarding the anterior portion. The posterior ends with perineal patterns were observed under compound microscope in a drop of glycerin on a clear slide and pressed gently against the glass. Cover slip was heat up slightly and placed on the glycerin drop, sealed with nail polish and labeled [14]. A drop of emulsion oil was placed on the slide for observation under 100X objective. *M. javanica* was identified on the basis of perennial pattern from all the weeds. Ten to fifteen females were processed for the purpose of identification [15].

3. Results

3.1 Incidence of *Meloidogyne javanica* on weeds of tomato growing area of Jabban and Dargai

An extensive field survey was conducted in root knot nematodes infested fields of Dargai and Jabban area of Malakand division of Pakistan. The findings of the study revealed fifteen weeds of different families such as *Amaranthus viridis*, *Asphodelus tenuifolius*, *Cannabis sativa*, *Chenopodium album*, *Coronopus didymus*, *Cynodon dactylon*, *Cyperus rotundus*, *Digitaria sanguinalis*, *Emex spinosa*, *Euphorbia helioscopia*, *Fumeria indica*, *Medicago denticulata*, *Melilotus indica*, *Parthenium* spp. and *Rumex crispus* were found in these fields. *Cannabis sativa*, *Digitaria sanguinalis*, *Fumeria indica*, *Melilotus indica* and *Rumex crispus* were found infected by root knot nematodes while others such as *Amaranthus viridis*, *Asphodelus tenuifolius*, *Chenopodium album*, *Coronopus didymus*, *Cynodon dactylon*, *Cyperus rotundus*, *Emex spinosa*, *Euphorbia helioscopia*, *Medicago denticulate* and *Parthenium* spp. were found without galls on their roots (Fig. 1, Fig. 2, Fig. 3 & Fig. 4).

Among these weeds *Cannabis sativa* was found in 14 fields; in two fields galls were observed on the roots (Galling index (GI) = 1) with an incidence of 9.9% (Field 2, Table 1) and 11.9% (Field 6, Table 2). Galls were also recorded for *Digitaria Sanguinalis* in Field 4 (Table 1) with 33.3% incidence. *Fumeria indica* was observed in 12 fields with high incidence of *M. javanica*. In Field 6 (Table 2), high incidence of root knot nematodes was observed in *Fumeria indica*, 43.47% with galling index-3, followed by Field 8 (43.05%) with galling index-2. Similarly in Field 4 (Table 1), 5, 7 (Table 2), 9, 10 and 12 (Table 4.3), *Fumeria indica* was found infected with root knot nematodes. Incidence of root knot nematodes in *Melilotus indica* was observed 22.13% (Field 3, Table 1). *Rumex crispus* was present in eight fields out of 20 and found infected in Field 2 (Table 1) with 22.72% incidence while in Field 8 (Table 4.2) the incidence of nematodes was 18.18%.

Among these 15 weeds, *Amaranthus viridis* was found infected in four fields. Similarly *Asphodelus tenuifolius* found in 18 fields (Table 1, 2, 3, 4, 5) and were found non-infected. *Chenopodium album* spread in 16 fields with no galls. *Coronopus didymus* was observed in 15 fields with 0% incidence. Similarly, *Cynodon dactylon* was also observed as non host growing in 4 fields. Incidence in *Cyperus rotundus* was 0% and was found in eight fields out 20. *Emex spinosa* was found in four fields with 0% incidence. *Euphorbia helioscopia* was observed uninfected in 16 fields. Similarly, *Medicago denticulata* was found in 14 fields with no infection. In eight fields *Parthenium* spp. was observed in the non growing areas and found un-infected with root knot nematodes.

Survey results showed that *Asphodelus tenuifolius* was found widely distributed in tomato growing area of Dargai and Jabban, and no galls were observed on its roots.

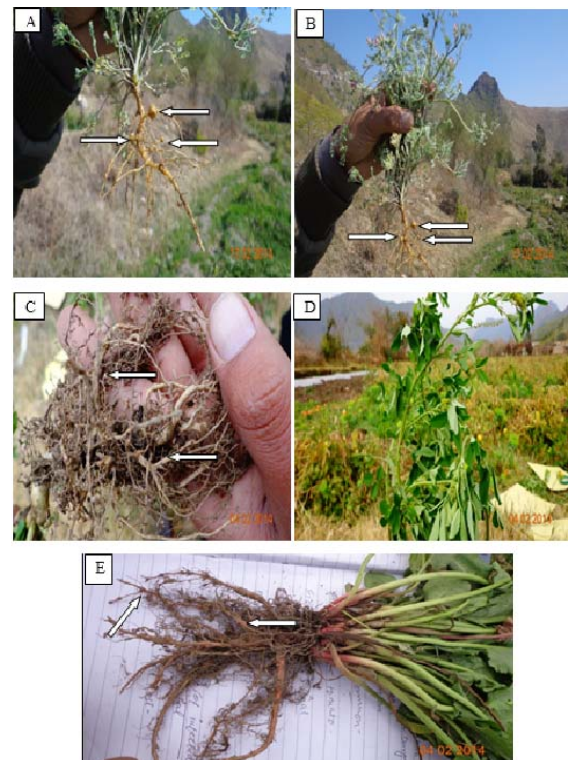


Fig 1: Incidence of *M. javanica* on *Fumeria indica* roots: Galled roots of *Fumeria indica* (A and B). Infected root of *Melilotus indica* (C). The above ground part of *Melilotus indica* (D). *Rumex crispus* with galled roots (E)



Fig 2: Incidence of Root Knot nematode on *Cannabis sativa* roots: Galled roots of *Cannabis sativa* (A). *Cannabis sativa* (B). *Digitaria sanguinalis* roots with galls (C)



Fig 3: Occurrence of non host weeds *Asphodelus tenuifolius* (wild onion/onion weed). *Asphodelus tenuifolius* (A and B). *Amaranthus viridis* (C). *Coronopus didymus* (D). *Cyperus rotundus* (E). *Emex spinosa* (F)

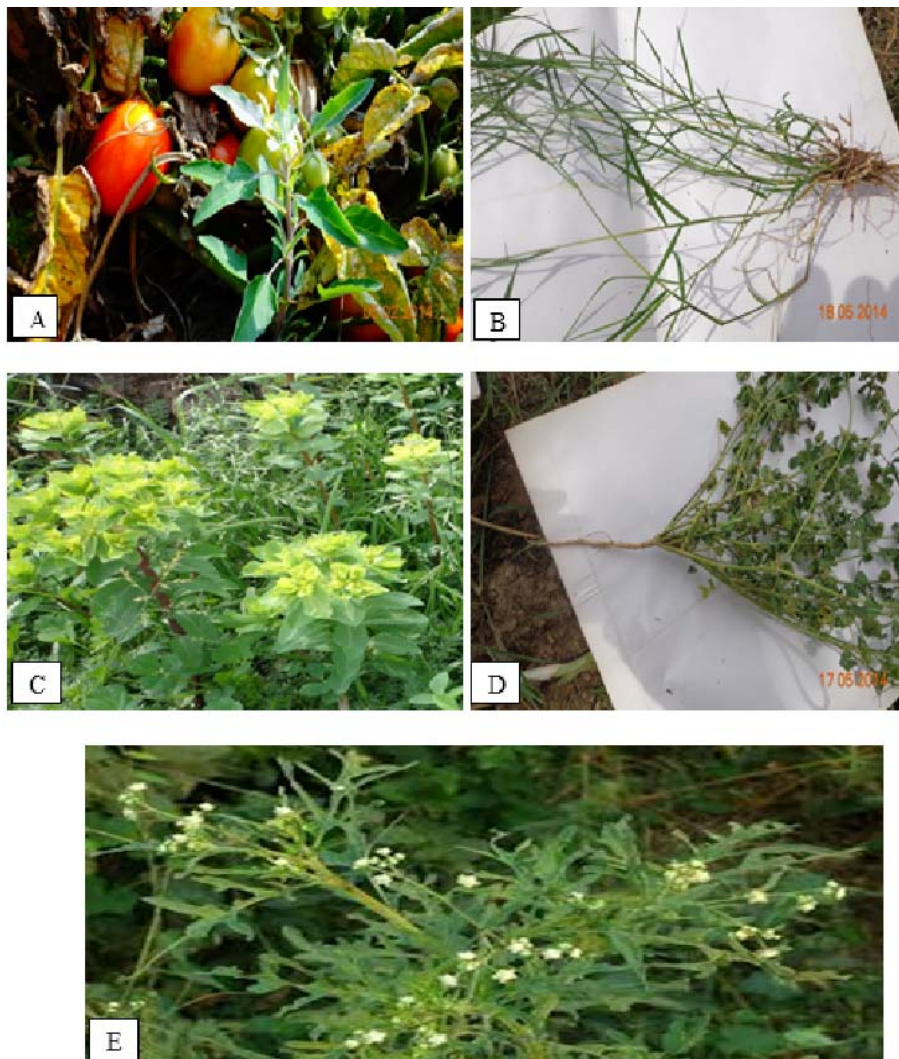


Fig 4: Occurrence of non host weeds. *Chenopodium album* (A). *Cynodon dactylon* (B). *Euphorbia helioscopia* (C). *Medicago denticulate* (D). *Parthenium spp.* (E)

Table 1: Incidence of *Meloidogyne javanica* on winter weeds of tomato growing areas of Jabban and Dargai

S. No.	Weeds	Local name	Field # 1		Field # 2		Field # 3		Field # 4	
			Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling index	% Incidence	Galls/Galling Index	% Incidence
1	<i>Amaranthus viridis</i>	Surguly	----	----	----	----	No/0	0%	No/0	0%
2	<i>Asphodelus tenuifolius</i>	Piyazy	----	----	----	----	No/0	0%	No/0	0%
3	<i>Cannabis sativa</i>	Bhang	No/0	0%	Yes/1	9.9 %	----	----	----	----
4	<i>Chenopodium album</i>	Sarmy	No/0	0%	----	----	----	----	----	----
5	<i>Coronopus didymus</i>	Murdar Botay	No/0	0%	No/0	0%	----	----	No/0	0%
6	<i>Cynodon dactylon</i>	Kabal	----	----	No/0	0%	No/0	0%	----	----
7	<i>Cyperus rotundus</i>	Dhela	----	----	----	----	No/0	0%	----	----
8	<i>Digitaria sanguinalis</i>	Kuray	----	----	----	----	----	----	Yes/1	33.3%
9	<i>Emex spinosa</i>	Markundai	----	----	----	----	----	----	----	----
10	<i>Euphorbia helioscopia</i>	Pulpulak	No/0	0%	No/0	0%	No/0	0%	No/0	0%
11	<i>Fumeria indica</i>	Papra	No/0	0%	No/0	0%	No/0	0%	Yes/2	21.05%
12	<i>Medicago denticulata</i>	Peshtary	No/0	0%	No/0	0%	No/0	0%	----	----
13	<i>Melilotus indica</i>	Lewanai	----	----	----	----	Yes/3	22.13%	----	----
14	<i>Parthenium spp.</i>	Khar Botay	No/0	0%	No/0	0%	No/0	0%	----	----
15	<i>Rumex crispus</i>	Shalkhay	No/0	0%	Yes/1	22.72 %	----	----	----	----

---- = Absence of weed in the field Yes = Galls were present
 No = Galls were absent 0,1,2,3,4,5 = Galling Index key(GI)

Table 2: Incidence of *Meloidogyne javanica* on winter weeds of tomato growing areas of Jabban and Dargai.

S. No.	Weeds	Local name	Field # 5		Field # 6		Field # 7		Field # 8	
			Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence
1	<i>Amaranthus viridis</i>	Surguly	----	----	----	----	----	----	----	----
2	<i>Asphodelus tenuifolius</i>	Piyazy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
3	<i>Cannabis sativa</i>	Bhang	----	----	Yes/1	11.9 %	----	----	----	----
4	<i>Chenopodium album</i>	Sarmy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
5	<i>Coronopus didymus</i>	Murdar Botay	No/0	0%	----	----	No/0	0%	No/0	0%
6	<i>Cynodon dactylon</i>	Kabal	----	----	----	----	----	----	----	----
7	<i>Cyperus rotundus</i>	Dhela	----	----	----	----	----	----	----	----
8	<i>Digitaria sanguinalis</i>	Kuray	----	----	----	----	----	----	No/0	0%
9	<i>Emex spinosa</i>	Markundai	----	----	----	----	No/0	0%	----	----
10	<i>Euphorbia helioscopia</i>	Pulpulak	No/0	0%	No/0	0%	No/0	0%	No/0	0%
11	<i>Fumeria indica</i>	Papra	Yes/3	38.09%	No/3	43.47%	Yes/1	40%	Yes/2	43.05%
12	<i>Medicago denticulata</i>	Peshtary	No/0	0%	No/0	0%	----	----	No/0	0%
13	<i>Melilotus indica</i>	Lewanai	No/0	0%	No/0	0%	No/0	0%	No/0	0%
14	<i>Parthenium spp.</i>	Khar Botay	No/0	0%	----	----	----	----	----	----
15	<i>Rumex crispus</i>	Shalkhay	No/0	0%	----	----	----	----	Yes/1	18.18%

---- = Absence of weed in the field Yes = Galls were present
 No = Galls were absent 0,1,2,3,4,5 = Galling Index key(GI)

Table 3: Incidence of *Meloidogyne javanica* on winter weeds of tomato growing areas of Jabban and Dargai

S. No.	Weeds	Local name	Field # 9		Field # 10		Field # 11		Field # 12	
			Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence
1	<i>Amaranthus viridis</i>	Surguly	----	----	----	----	----	----	----	----
2	<i>Asphodelus tenuifolius</i>	Piyazy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
3	<i>Cannabis sativa</i>	Bhang	No/0	0%	No/0	0%	No/0	0%	----	----
4	<i>Chenopodium album</i>	Sarmy	No/0	0%	No/0	0%	----	----	No/0	0%
5	<i>Coronopus didymus</i>	Murdar Botay	No/0	0%	No/0	0%	----	----	No/0	0%
6	<i>Cynodon dactylon</i>	Kabal	----	----	No/0	0%	No/0	0%	----	----
7	<i>Cyperus rotundus</i>	Dhela	----	----	----	----	----	----	----	----
8	<i>Digitaria sanguinalis</i>	Kuray	----	----	----	----	----	----	----	----
9	<i>Emex spinosa</i>	Markundai	No/0	0%	No/0	0%	No/0	0%	----	----
10	<i>Euphorbia helioscopia</i>	Pulpulak	No/0	0%	No/0	0%	No/0	0%	No/0	0%
11	<i>Fumeria indica</i>	Papra	Yes/1	12.50%	Yes/1	40%	No/0	0%	Yes/2	22.45%
12	<i>Medicago denticulata</i>	Peshtary	No/0	0%	No/0	0%	No/0	0%	----	----
13	<i>Melilotus indica</i>	Lewanai	No/0	0%	No/0	0%	No/0	0%	No/0	0%
14	<i>Parthineum</i> spp.	Khar Botay	----	----	----	----	----	----	----	----
15	<i>Rumex crispus</i>	Shalkhay	----	----	----	----	----	----	No/0	0%

---- = Absence of weed in the field Yes = Galls were present
 No = Galls were absent 0,1,2,3,4,5 =Galling Index key(GI)

Table 4: Incidence of *Meloidogyne javanica* on winter weeds of tomato growing areas of Jabban and Dargai.

S. No.	Weeds	Local name	Field # 13		Field # 14		Field # 15		Field # 16	
			Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence
1	<i>Amaranthus viridis</i>	Surguly	----	----	----	----	----	----	----	----
2	<i>Asphodelus tenuifolius</i>	Piyazy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
3	<i>Cannabis sativa</i>	Bhang	No/0	0%	No/0	0%	No/0	0%	No/0	0%
4	<i>Chenopodium album</i>	Sarmy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
5	<i>Coronopus didymus</i>	Murdar Botay	No/0	0%	No/0	0%	No/0	0%	No/0	0%
6	<i>Cynodon dactylon</i>	Kabal	----	----	----	----	----	----	----	----
7	<i>Cyperus rotundus</i>	Dhela	No/0	0%	No/0	0%	No/0	0%	No/0	0%
8	<i>Digitaria sanguinalis</i>	Kuray	----	----	----	----	----	----	----	----
9	<i>Emex spinosa</i>	Markundai	----	----	----	----	----	----	----	----
10	<i>Euphorbia helioscopia</i>	Pulpulak	No/0	0%	No/0	0%	No/0	0%	----	----
11	<i>Fumeria indica</i>	Papra	----	----	----	----	----	----	----	----
12	<i>Medicago denticulata</i>	Peshtary	No/0	0%	No/0	0%	----	----	No/0	0%
13	<i>Melilotus indica</i>	Lewanai	No/0	0%	No/0	0%	No/0	0%	No/0	0%
14	<i>Parthineum</i> spp.	Khar Botay	----	----	No/0	0%	----	----	----	----
15	<i>Rumex crispus</i>	Shalkhay	----	----	----	----	No/0	0%	No/0	0%

---- = Absence of weed in the field Yes = Galls were present
 No = Galls were absent 0,1,2,3,4,5 =Galling Index key(GI)

Table 5: Incidence of *Meloidogyne javanica* on winter weeds of tomato growing areas of Jabban and Dargai

S. No.	Weeds	Local name	Field # 17		Field # 18		Field # 19		Field # 20	
			Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence	Galls/Galling Index	% Incidence
1	<i>Amaranthus viridis</i>	Surguly	----	----	No/0	0%	----	----	----	----
2	<i>Asphodelus tenuifolius</i>	Piyazy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
3	<i>Cannabis sativa</i>	Bhang	No/0	0%	No/0	0%	No/0	0%	No/0	0%
4	<i>Chenopodium album</i>	Sarmy	No/0	0%	No/0	0%	No/0	0%	No/0	0%
5	<i>Coronopus didymus</i>	Murdar Botay	No/0	0%	----	----	No/0	0%	----	----
6	<i>Cynodon dactylon</i>	Kabal	----	----	----	----	----	----	----	----
7	<i>Cyperus rotundus</i>	Dhela	No/0	0%	----	----	No/0	0%	No/0	0%
8	<i>Digitaria sanguinalis</i>	Kuray	----	----	----	----	----	----	----	----
9	<i>Emex spinosa</i>	Markundai	----	----	----	----	----	----	----	----
10	<i>Euphorbia helioscopia</i>	Pulpulak	----	----	----	----	----	----	No/0	0%
11	<i>Fumeria indica</i>	Papra	----	----	----	----	----	----	----	----
12	<i>Medicago denticulata</i>	Peshtary	----	----	No/0	0%	----	----	No/0	0%
13	<i>Melilotus indica</i>	Lewanai	----	----	No/0	0%	No/0	0%	No/0	0%
14	<i>Parthineum spp.</i>	Khar Botay	----	----	No/0	0%	No/0	0%	No/0	0%
15	<i>Rumex crispus</i>	Shalkhay	----	----	----	----	No/0	0%	----	----

---- = Absence of weed in the field Yes = Galls were present
 No = Galls were absent 0,1,2,3,4,5 = Galling Index key (GI)

4. Discussion

During the study an extensive survey was conducted in the root knot nematodes infested areas of Malakand division. Fifteen different weeds such as *Amaranthus viridis*, *Asphodelus tenuifolius*, *Cannabis sativa*, *Chenopodium album*, *Coronopus didymus*, *Cynodon dactylon*, *Cyperus rotundus*, *Digitaria sanguinalis*, *Emex spinosa*, *Euphorbia helioscopia*, *Fumeria indica*, *Medicago denticulata*, *Melilotus indica*, *Parthineum spp.* and *Rumex crispus* were found growing in these areas. *Cannabis sativa*, *Digitaria sanguinalis*, *Fumeria indica*, *Melilotus indica* and *Rumex crispus* were found with galls on their roots and *Amaranthus viridis*, *Asphodelus tenuifolius*, *Chenopodium album*, *Coronopus didymus*, *Cynodon dactylon*, *Cyperus rotundus*, *Emex spinosa*, *Euphorbia helioscopia*, *Medicago denticulate* and *Parthineum spp.* were found without galls. This study confirmed that some weed hosts do serve as reservoirs of nematodes on which nematodes survive in the absence of actual host. Weeds such as *Momordica charantia*, *Physalis angulata*, *Coccinia grandis*, *Cyanthillium cinereum*, *Oldenlandia corymbosa*, *Macroptilium atropurpureum*, *Ludwigia hyssopifolia*, *Senna obtusifolia*, *Amaranthus viridis*, *Ageratum conyzoides* and *Solanum lycopersicum* were reported as reservoir of root knot nematodes [16]. This study also explored the economic as well as pathological losses caused by weeds in crop plants. The trend of organic agriculture is increasing day by day because of an alternative management of plant diseases. The use of synthetic chemicals is restricted in many countries because of being costly and negative effects on environment and human health. To reduce the environmental hazards, extracts of those

weeds which were free of galls could be used against root knot nematodes of tomato. Undiluted stock extract of these weeds can also be found very effective against other plant pathogens. Moreover pure undiluted extracts of *Cannabis sativa* and *Zanthoxylum alatum* were found to be more effective and nematicidal against *Meloidogyne incognita* [17]. The nematicidal potentials of weeds have previously been reported [18]. Phytochemicals obtained from the galls-free weeds can be used to interrupt the reproduction potentials of root knot nematodes by inhibition of egg hatching [19]. Alkaloids are reported by [20] in *Asphodelus tenuifolius* extracts which is nematicidal chemical group [21]. Furthermore, phyto-chemical comparison between the susceptible and resistant species can shed more light on why some weed species can serve as source of phytochemicals for control of RKN, while other species are susceptible to RKN infection. This research contributed a lot to the science of agriculture and nematode management with eco- and farmer-friendly methods. The study explored that weeds are growing in the tomato growing areas with alarming populations, secondly these weeds also serve as reservoirs of root knot nematodes and nematodes survive on these weeds in the absence of hosts. Extracts from non host weeds can help in the organic management of root knot nematodes.

5. Conclusion

It can be concluded that five weeds namely *Cannabis sativa*, *Digitaria sanguinalis*, *Fumeria indica*, *Melilotus indica* and *Rumex crispus* were found as hosts of root knot nematodes. There were few weeds which were found to be free of root knot nematodes galls. Further studies are recommended to

identify the nematicidal efficacy of gall free weeds against root knot nematodes.

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