

Available Online at EScience Press **Plant Protection** 

ISSN: 2617-1287 (Online), 2617-1279 (Print) http://esciencepress.net/journals/PP

# DEVELOPMENT OF MELOIDOGYNE INCOGNITA ON SELECTED OKRA CULTIVARS

Muhammad Usman Afzal<sup>1</sup>, Sajid Aleem Khan<sup>1</sup>, Nosheen Salehon<sup>2</sup>, Mehwish Naz<sup>1</sup>, Nasir Ahmad Khan<sup>1</sup>

<sup>1</sup> Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan.

<sup>2</sup> Pest Warning and Quality Control of Pesticides, Hafizabad, Punjab, Pakistan.

# ARTICLE INFO

# ABSTRACT

Article history Received: 13<sup>th</sup> May, 2019 Revised: 29<sup>th</sup> July, 2019 Accepted: 28<sup>th</sup> August, 2019

**Keywords** Okra Meloidogyne incognita Screening Resistance Root-knot nematode Okra is a valuable vegetable crop of Pakistan. It is predominantly hosted by pathogens that reduce its yield and quality. Among these yield reducing agents, root knot nematodes are very important due to their host rang. Chemical control for this important pathogen is expensive and causes pollution problems so the present study was planned to evaluate the level of resistance among different available varieties of okra against *Meloidogyne incognita* under field conditions. The selected field was already infested with *M. incognita*. Data were recorded on various plant growth parameters and nematode development parameters after sixty days. Results indicated that for various nematode populations and plant growth attributes all the cultivars significantly differed with the Sabzpari 2001 and Sabzpari X Ramakrishna having best performance on over all yield by showing moderately resistant response but no variety was resistant against *M. incognita*. On over all basis and yield attributes, it is recommended that Sabzpari 2001 and Sabzpari X Ramakrishna are moderately resistant to nematode infested soils of Pakistan.

Corresponding Author: Sajid Aleem Khan Email: sajid\_aleem@uaf.edu.pk © 2019 EScience Press. All rights reserved.

# INTRODUCTION

Okra (Hibiscus esculentus L.) belonging to family Malvaceae is an important cross-pollinated vegetable crop of Indo-Pak sub-continent. It can be cultivated successfully around the globe especially in sub tropics, tropics, and temperate regions of Africa, Brazil, Middle East, Turkey and the southern states of United States (Acquistucci and Francisci, 2002). The global pod production of okra is 4.8 million tons, out of which India contributes 70%, Nigeria 15%, Pakistan 2%, Ghana 2%, Egypt 1.7 % and Iraq 1.7% (Gulsen et al., 2007). The crop has been challenged by various yield reducing agents including fungi. bacteria, viruses, mycoplasma, nematodes, and insects. The total loss of vegetables due to these pests has been estimated up to 20-30% and unchecked growth may increase the loss up to 80-90% and it would fetch very low price in market (Hamer and Thomson, 1957).

In Pakistan, okra crop is infested by 23 species of nematodes. Among these 23 destructive species of nematodes, *M. incognita* is the most damaging and causes economically important losses (Hussain and Mukhtar, 2019; Kayani and Mukhtar, 2018; Kayani et al., 2017, 2018; Kayani et al., 2013; Mukhtar, 2018; Mukhtar and Hussain, 2019; Mukhtar et al., 2018; Mukhtar and Kayani, 2019; Nazir et al., 2019; Tariq-Khan et al., 2017). Pakistan is facing more root-knot nematode problem than the other developing countries, for the reason that here the

climatic conditions are favorable in tropical and subtropical areas round the year. Sandy soil in hot irrigated areas encourages the infection of root-knot nematode.

Root knot nematodes are considered as threat to cultivated fields due to severe root damage of crops in Caribbean (Singh, 1975). More than 100 species of Meloidogyne have been described, but the four most frequently occurring species are *M. javanica*, *M. hapla*, *M.* incognita and M. arenaria. More than 2000 species of plants are attacked by the species of root-knot nematode and most of them are the cultivated plants. Meloidogyne spp. causes 5% to 43% of total losses in the vegetables (Sasser, 1989). Up to 27% losses of yield in okra were reported due to RKN (Anwar and McKenry, 2012). On global basis, M. incognita has been reported first time to constitute about 47% of the total RKN population (Sasser and Carter, 1985). M. incognita and M. javanica were found dominant species associated with banana, papaya, sugarcane, cabbage, okra in Sindh, Punjab and NWFP (Ahmad and Saeed, 1981; Brown, 1962).

The effect of number of nematodes on the growth of plant and yield can often be explained as a linear regression of growth or yield on log nematode numbers. Many strategies are used to reduce the nematode population in infested fields like chemical treatment of the soil, the use of natural enemies or the biocontrol agents (Khan et al., 2017; Mukhtar et al., 2013a; Vagelas and Gowen, 2012), crop rotation and by using the antagonistic plants (Kayani et al., 2012; Mukhtar et al., 2013b). Due to cost benefit ratio and the pollution, the use of chemicals is not effective. By using the resistant cultivars and the non-hosts, the nematode population can be reduced in the soil (Hussain et al., 2014; Hussain et al., 2016; Mukhtar et al., 2017a; Mukhtar et al., 2017b; Mukhtar et al., 2014; Mukhtar et al., 2013c). Therefore, the present studies were planned to screen okra cultivars against *M. incognita*.

#### **MATERIALS AND METHODS**

**Collection of root samples and Purification of** *Meloidogyne incognita*: Roots of tomato plants infected with root knot nematodes were collected from the research area of the Department of Plant Pathology, University of Agriculture Faisalabad. Ten diseased plants with galls were carefully dug with the help of trowel to a depth of one foot from the rhizosphere of tomato plants with adhering soil. Samples infected with root-knot nematodes were brought in ploythene bags in the laboratory. The samples were stored at 15°C to maintain the freshness of the samples. Samples were taken out from the cool incubator and infected roots were gently rinsed under tap water.

Isolation and differentiation of root knot nematode (Meloidogvne incognita): Isolation and differentiation of nematodes was performed by following the method described by Taylor and Netscher (1974). Galls containing mature females were selected and kept in petri dishes with tap water. The galls were dissected to get adult females. Necks of females were cut off with the help of half spear to pluck the interiors out. The cuticle was kept in petri dishes containing a drop of 45% lactic acid. Similarly, 5 to 10 cuticles were cut in half with the help of blade. The portion of cuticles with perineal patterns was transferred to another drop. After that it was trimmed around the perineal pattern to square in shape. The trimmed perineal pattern was kept back in the 45% lactic acid and was cleaned from debris using the pulp canal file. The perineal pattern was transferred to a drop of glycerin on a clean microslide after cleaning and aligned with having orientation of anus downward. It is pressed carefully against the glass with the help of pulp canal file. A warm cover slip was kept on the glycerin drop, sealed with nail polish and was labeled (Hartman and Sasser, 1985). Diagnostic characters of Meloidogyne incognita (a) Perineal pattern of Meloidogyne incognita is sole as it contains lateral ridges which divide lateral and ventral striae, (b) dorsal arch is low and rounded and (c) striae are smooth to slightly wavy and some striae bend towards valve edges. Egg sacs were got from galled roots of infected plant samples by teasing, washing roots in water and recovering the eggs on a 60 mesh sieve. Second stage juveniles were isolated from the badly infested roots of tomato and egg plants.

**Evaluation of okra germplasm**: Seeds of selected varieties of okra Ikra-1, Ikra-2, Ikra-3, Ikra-4, Perbhani Kranti, Sabzpari X Karan 51, Clemson Spineless, Punjab Selection, Pusa Green, Sabzpari 2001, Sabzpari X Ramakrishna, Green Wonder, Super Star, Sanam, Arka Anomika, Red Burgundy, Okra-Haseena F1, Okra-OP-Greenpod, Sabz Pari and Punjab-8 were sown in field already infested with root-knot nematode with susceptible tomato cultivar. The experiment was conducted in research area of the Department of Plant Pathology, University of Agriculture, Faisalabad. Infestation level was 150 nematodes/100 ml of soil. Randomized Complete Block Design was followed for experiment. Plants were harvested at maturity. Roots were washed in water by gently shaking it in the bucket

and forceps were used to remove the debris. The roots were then cut off from the other part of the plants and were placed in folds of tissue paper to avoid drying. Phloxin B was used to stain the roots and to count the egg masses. The roots were placed for 15-20 minutes in Phloxin B solution (0.15g/liter tape water). To prevent drying, roots were wrapped in tissue paper after staining during the steps of the procedure. Stereo microscope was used to count the egg masses from the entire root system. The rating of the root systems was done on the bases of galling and egg masses presence on a 0.5 scale. (Anwar et al., 2007; Taylor and Sasser, 1978).

**Data Collection**: Number of galls per root system was estimated and galling index was calculated using rating scale (Taylor and Sasser, 1978). To estimate egg masses per root system, 1 g sub-sample of roots was stained with acid fuchsin, number of egg masses were counted and was multiplied with total weight of root. To calculate the number of females and juveniles inside the roots, 1 g sub sample of root was macerated for 30-40 seconds in a waring blender and counts were made in the suspension thus obtained and were multiplied with total weight. Root and shoot weights were measured as the mean of three replications. For this purpose, roots were cut from the base of the stems and their weights were measured by electric balance. Total numbers of leaves and fruits were counted manually. Root length was measured by using measuring tape. Fruit weight was taken as the average of four plants.

**Statistical analysis**: The data were analyzed by using SAS statistical package and treatment means were compared by using Least Significance Difference (LSD) test at 5% probability level (Steel et al., 1997).

# **RESULTS AND DISCUSSION**

Results indicated that all the varieties showed significant variations in number of galls per root system. Punjab-8 was highly susceptible and produced maximum (337) galls. Better performance was exhibited by the cultivar Sabzperi X Ramakrishna which produced minimum (3) galls per root system. Neog et al. (2000) observed significant growth reduction in okra plants with respect to growth parameters including height, number of fruits, fruit length, yield, root length, fresh, and dry weight of roots. Number of egg masses per root system also showed fluctuations in their values after the analysis. It is clear from table 1 that cultivar Punjab selection was highly susceptible and produced maximum (275) egg masses per root system followed by Punjab-8 (273). Better performance was exhibited by the cultivar Sabzpari 2001 which produced minimum (2) egg masses per root system. It depicts that these cultivars vary in genetic makeup (Brian et al., 2010). Number of females per root system varied significantly in all cultivars as predicted from table 1.

-0	TT + .		<u> </u>		N C I 11 (		
Sr.	Variety	No. of galls/ root	No. of egg masses/	No. of females/	No. of Juveniles/		
No.		system	root system	root system	root system		
1	Ikra-1	88.2 C	337.0 A	92.2 HI	385 AB		
2	Ikra-2	90.4 C	82.4 GH	94.0 H	784 G		
3	Ikra-3	88.8 C	84.0 G	90.4 HI	902 G		
4	Ikra-4	91.4 C	81.0 GH	89.2 I	858 G		
5	Perbhani Kranti	203.6 B	170.0 DEF	200.2 DEFG	14619 EF		
6	Sabzperi X Karan 51	3.8 D	3.0 I	6.2 I	178 G		
7	Clemson Spineless	202.0 B	172.4 CD	202.4 CDE	15153 DEF		
8	Punjab Selection	336.4 A	275.2 AB	383.0 B	27836 A		
9	Pusa Green	203.4 B	173.4 CD	204.2 C	13545 EF		
10	Sabzperi 2001	3.2 D	2.0 I	5.4 [	103 G		
11	Sabzperi X Ramakrishna	2.8 D	4.0 I	7.2 [	193 G		
12	Green Wonder	200.2 B	168.6 EF	199.0 EFG	14207 EF		
13	Sabzperi China Red	200.0 B	167.0 F	198.2 FG	13313 F		
14	Super Star	87.2 C	79.0 H	91.4 HI	795 G		
15	Sanam	198.4 B	175.0 C	203.8 CD	16710 CD		
16	Arka Anomika	199.8 B	171.8 CDE	201.4 CDEF	17160 CD		
17	Red Burgundy	201.4 B	170.4 DEF	202.0 CDE	17389 C		
18	OKRA-Haseena F1	335.6 A	277.0 A	387.0 A	25170 B		
19	OKRA-OP-Greenpod	201.6 B	174.2 C	197.6 G	15399 CDE		
20	Sabz Pari	3.4 D	3.0 I	8.20 J	115 G		
21	Punjab-8	337.0 A	273.0 B	385.20 AB	29217 A		

Table: 1. Nematode infestations on different okra cultivars by *Meloidogyne incognita*.

Number of Juveniles per root system also showed

fluctuations in their values and all the cultivars showed

significant results for the number of juveniles per root system. It was evident that cultivar Punjab-8 was highly susceptible and produced maximum (55.68 g) fresh root weight followed by Okra-Haseena F1 producing (54.6) gram root weight. Better performance was exhibited by the cultivar Sabzpari which produced minimum (21.1 g) fresh weight of root. Similarly, cultivars Ikra-1 and Ikra-4 produced same fresh weight as compared to all other cultivars with little variations. Table. 2 exhibited that there were great variations in the values of the number of leaves per plant for all cultivars. Among all the selected varieties OKRA-Haseena F1 was not profitable having 22 leaves per plant followed by the Pusa Green, having 24 leaves per plant. Maximum number of leaves was present on the shoot of Sabzpari 2001. The comparison of means in the table 2 revealed that among all the cultivar Sabzpari produced the maximum (28.4 cm) root length per plant followed by Punjab-8 producing (27.26) cm root length. Minimum root length was produced by the cultivar Okra-Haseena F1 producing (18.4 cm) of root length. Fruit weight also showed fluctuations in its values after the analysis.When the means were compared with each other it was evident that cultivar Sabzpari produced minimum (15.32 g) fruit weight.

Poor performance was exhibited by the cultivar Punjab-8 which was highly susceptible and produced minimum (3.46 g) fruit weight followed by Okra-Haseena F1 producing (4.04) gram fruit weight. Performance of the cultivars showed variations in the results of various growth and yield attributes as documented in literature about the investigation of the root knot nematode infestation on various other crops like tomato, egg plant, rice and many other crops as well (Kamran et al., 2011; Srivastava et al., 2011; Ullah et al., 2011).

Table 2. Effect of *Meloidogyne* incognita on different plant growth parameters.

Sr. No	Variety		Root	length	Number Leaves	Fresh	Shoot	Fruit Weight	Fresh	root	
			(cm)		per plant	weight	(g)	(g)	weight (g)		
1	Ikra-1	22.8 DE		E	31.0 FGH	26.2 IJ		7.0 EFG	33.8 HI	33.8 HI	
2	Ikra-2		24.0 C	D	229.2 HI	29.8 E		8.3 DEF	35.9 H		
3	Ikra-3		23.0 D	E	34.6 CDEF	33.3 C		9.1 D	32.1 IJ		
4	Ikra-4		22.0 D	EFG	36.0 CDE	31.1 DE	E	8.7 DE	30.9 JK		
5	Perbhani Kranti		21.2 E	FGH	30.2 GH	43.0 AE	3	5.1 HIJK	46.0 CD	1	
6	Sabzperi X Karan 51		26.8 A	В	38.0 BC	15.4 I		14.1 AB	22.2 LM	I	
7	<b>Clemson Spineless</b>		22.0 D	EFG	29.4 GHI	19.3 GF	ł	6.6 FGH	44.2 DE	F	
8	Punjab Selection		19.2 H	I	24.4 JK	44.3 A		3.7 JK	52.8 B		
9	Pusa Green		22.4 D	EF	31.0 FGH	42.2 B		6.4 GH	47.1 C		
10	Sabzperi 2001		27.6 A		42.0 A	20.2 G		11.4 C	23.9 L		
11	Sabzperi	Х	27.4 A		36.8 BCD	20.0 GF	ł	13.1 BC	22.9 LM	I	
12	Green Wonder		20.6 F	GH	32.0 FGH	23.4 F		6.4 GH	42.3 FG		
13	Sabzperi China Red		21.6 E	FG	28.4 HI	22.2 F		5.2 HIJ	41.3 G		
14	Super Star		25.2 B	С	33.2 DEFG	32.2 CD	)	8.6 DE	29.4 K		
15	Sanam		21.8 E	FG	25.6 IJK	22.0 F		5.7 GHI	43.2 EF	G	
16	Arka Anomika		20.2 G	HI	29.2 HI	20.2 G		6.5 GH	45.1 CD	Έ	
17	Red Burgundy		21.4 E	FG	32.2 EFGH	18.3 H		5.9 GH	42.4 FG		
18	OKRA-Haseena F1		18.4 I		22.0 K	14.0 I		4.0 IJK	54.6 AB	}	
19	OKRA-OP-Greenpod		21.4 E	FG	30.2 GH	19.3 GF	ł	5.6 GHI	45.0 CD	Έ	
20	Sabz Pari		28.4 A		40.4 AB	43.0 AE	3	15.3 A	21.1 M	21.1 M	
21	Punjab-8		27.6 A		26.2 IJ	43.0 AE	3	3.4 K	55.6 A		

# CONCLUSION

It is concluded from the above experiment that among all the cultivars none of the variety was tolerant for all the selected parameters. Variety behaving well for one parameter was not favorable for another parameter. On over all basis and yield attributes, it is recommended that Sabzpari 2001, Sabzpari X Ramakrishna and Sabzpari have tolerance to nematode infestation and should be recommended to farmers in root-knot nematode infested soils of Pakistan. **Author contributions:** MUA, SAK and NS planned and designed the studies, NS executed the experiments and collected data, SAK supervised the research work, MN and NSK analyzed the data, RK wrote the manuscript and all the authors edited it.

**Conflict of interest:** The authors declare no conflict of interest.

**Acknowledgements:** The work is a part of ongoing Ph.D. degree funded by Higher Education Commission of Pakistan.

# REFERENCES

- Acquistucci, R., Francisci, R., 2002. Effect of okra (*Hibiscus esculentus L.*) addition on the technological properties of a wheat flour. International journal of Food Sciences and Nutrition 53, 375-379.
- Ahmad, M.M., Saeed, M., 1981. Studies on root-knot nematodes in Pakistan, Proceedings of 3rd Research and Planning Conference on Root-Knot Nematodes, IMP, Region V1. Jakarta. Indonesia, p. 115.
- Anwar, S.A., McKenry, M.V., 2012. Incidence and population density of plant parasitic nematodes infecting vegetable crops and associated yield losses. Pakiastan Journal of Zoology 44, 327-333.
- Anwar, S.A., Zia, A., Hussain, M., Kamran, M., 2007. Host suitability of selected plants to *Meloidogyne incognita* in the Punjab, Pakistan. International Journal of Nematology 17, 144-150.
- Brian, M.S., Kevin, E.K., William, T.C., Jason, A.F., Grady, L.M., Kenneth., H.Q., 2010. Variable responses of Zoysia grass genotypes to the sting nematode. Crop Science 50, 723-729.
- Brown, K.F., 1962. A survey of some plant parasitic nematode problems in Pakistan., Report of the visiting Nematologist. Shell International Chemicals Company Limited.
- Gulsen, O., Karagul, S., Abak, K., 2007. Diversity and relationships among Turkish germplasm by SRAP and Phenotypic marker polymorphism. Biologia Bratislava 62, 41-45.
- Hamer, C., Thomson, C.H., 1957. Vegetables Crops. McGraw-Hill Co., Inc. NewYork Toronto, London.
- Hartman, K.M., Sasser, J.N., 1985. Identification of *Meloidigyne* species on the basis of different hosts tests and prineal patterns morphology, in: Barker, Carter, C.C., Sasser, J.N. (Eds.), Advance reatise on Meloidogyne methology, Department of Plant Pathology, NCCSU and USAID Ralegih, North Carolina, pp. 74-78.

- Hussain, M.A., Mukhtar, T., 2019. Root-knot nematodes infecting okra in major vegetable growing districts of Punjab, Pakistan. Pakistan Journal of Zoology 51, 1137-1142.
- Hussain, M.A., Mukhtar, T., Kayani, M.Z., 2014. Characterization of susceptibility and resistance responses to root-knot nematode (*Meloidogyne incognita*) infection in okra germplasm. Pakistan Journal of Agricultural Sciences 51, 319-324.
- Hussain, M.A., Mukhtar, T., Kayani, M.Z., 2016. Reproduction of *Meloidogyne incognita* on resistant and susceptible okra cultivars. Pakistan Journal of Agricultural Sciences 53, 371-375.
- Kamran, M., Anwar, S.A., Khan, S.A., 2011. Evaluation of tomato genotypes against *Meloidogyne incognita* infection. Pakistan Journal of Phytopathology 23, 31-34.
- Kayani, M.Z., Mukhtar, T., 2018. Reproductivity of *Meloidogyne incognita* on fifteen cucumber cultivars. Pakistan Journal of Zoology 50, 1717-1722.
- Kayani, M.Z., Mukhtar, T., Hussain, M.A., 2012. Evaluation of nematicidal effects of *Cannabis sativa L.* and *Zanthoxylum alatum Roxb.* against root-knot nematodes, *Meloidogyne incognita.* Crop Protection 39, 52-56.
- Kayani, M.Z., Mukhtar, T., Hussain, M.A., 2017. Effects of southern root knot nematode population densities and plant age on growth and yield parameters of cucumber. Crop Protection 92, 207-212.
- Kayani, M.Z., Mukhtar, T., Hussain, M.A., 2018. Interaction between nematode inoculum density and plant age on growth and yield of cucumber and reproduction of *Meloidogyne incognita*. Pakistan Journal of Zoology 50, 897-902.
- Kayani, M.Z., Mukhtar, T., Hussain, M.A., Haque, M.I., 2013.
  Infestation assessment of root-knot nematodes (*Meloidogyne* spp.) associated with cucumber in the Pothowar region of Pakistan. Crop Protection 47, 49-54.
- Khan, A.R., Javed, N., Sahi, S.T., Mukhtar, T., Khan, S.A., Ashraf, W., 2017. *Glomus mosseae* (Gerd & Trappe) and neemex reduce invasion and development of *Meloidogyne incognita*. Pakiastan Journal of Zoology 49, 841-847.
- Mukhtar, T., 2018. Management of root-knot nematode, *Meloidogyne incognita*, in tomato with two *Trichoderma* species. Pakiastan Journal of Zoology

50, 1589-1592.

- Mukhtar, T., Arooj, M., Ashfaq, M., Gulzar, A., 2017a. Resistance evaluation and host status of selected green gram genotypes against *Meloidogyne incognita*. Crop Protection 92, 198-202.
- Mukhtar, T., Hussain, M.A., 2019. Pathogenic potential of Javanese root-knot nematode on susceptible and resistant okra cultivars. Pakistan Journal of Zoology 51, 1891-1897.
- Mukhtar, T., Hussain, M.A., Kayani, M.Z., 2013a. Biocontrol potential of *Pasteuria penetrans, Pochonia chlamydosporia, Paecilomyces lilacinus* and *Trichoderma harzianum* against *Meloidogyne incognita* in okra. Phytopathologia Mediterranea 52, 66-76.
- Mukhtar, T., Hussain, M.A., Kayani, M.Z., 2017b. Yield responses of 12 okra cultivars to southern rootknot nematode (*Meloidogyne incognita*). Bragantia 76, 108-112.
- Mukhtar, T., Hussain, M.A., Kayani, M.Z., Aslam, M.N., 2014. Evaluation of resistance to root-knot nematode (*Meloidogyne incognita*) in okra cultivars. Crop Protection 56, 25-30.
- Mukhtar, T., Jabbar, A., Raja, M.U., Javed, H., 2018. Reemergence of wheat seed gall nematode (*Anguina tritici*) in Punjab, Pakistan. Pakiastan Journal of Zoology 50, 1195-1198.
- Mukhtar, T., Kayani, M.Z., 2019. Growth and yield responses of fifteen cucumber cultivars to root-knot nematode (*Meloidogyne incognita*). Acta Scientiarum Polonorum: Hortorum Cultus 18, 45-52.
- Mukhtar, T., Kayani, M.Z., Hussain, M.A., 2013b. Nematicidal activities of *Cannabis sativa L*. and *Zanthoxylum alatum Roxb*. against *Meloidogyne incognita* Industrial Crops and Products 42, 447-453.
- Mukhtar, T., Kayani, M.Z., Hussain, M.A., 2013c. Response of selected cucumber cultivars to *Meloidogyne incognita*. Crop Protection 44, 13-17.
- Nazir, K., Mukhtar, T., Javed, H., 2019. In *vitro* effectiveness of silver nanoparticles against root-knot nematode (*Meloidogyne incognita*). Pakistan Journal of Zoology 51, 2077-2083.
- Neog, D.J., Pathak, J.J., Bhagabati, K.N., Sinha, A.K., 2000. Interaction between yellow vein mosaic virus and *Meloidogyne incognita* on growth of okra plants.

Journal of Interacademicia 4, 376-381.

- Sasser, J.N., 1989. Plant-Parasitic nematodes: The farmer's hidden enemy. Department of Plant Pathology, North Carolina State University, Raleigh, NC. USA., p. 115.
- Sasser, J.N., Carter, C.C., 1985. Overview of the International Meloidogyne Project, in: Sasser, J.N., Carter, C.C. (Eds.), An Advanced Treatise on *Meloidogyne*: Biology and Control Graphics Raleigh, North Carolina State University.
- Singh, N.D., 1975. Effect of inoculum level and plant age on pathogenicity of *Meloidogyne mcognita* and *Rotylenchus remformis* to tomato and lettuce. Plant Disease Research 59, 905-908.
- Srivastava, A., Rana, V., Rana, S., Singh, D., Singh, V., 2011. Screening of rice and wheat cultivars for resistance against root-knot nematode, *Meloidogyne* graminicola (Golden and Birchfield) in rice-wheat cropping system. Journal of Rice Research 4, 8-10.
- Steel, G.R.D., Torrie, J.H., Dicky, D.A., 1997. Principles and Procedures of Statistics, A Biometrical Approach, 3 ed. Mc Graw Hill Book Company Incorporation New York.
- Tariq-Khan, M., Munir, A., Mukhtar, T., Hallmann, J., Heuer,
  H., 2017. Distribution of root-knot nematode species and their virulence on vegetables in northern temperate agro-ecosystems of the Pakistani-administered territories of Azad Jammu and Kashmir. Journal of Plant Diseases and Protection 124, 201-212.
- Taylor, A.L., Sasser, J.N., 1978. Biology, Identification and Control of Root Knot Nematodes (*Meloidogyne* spp). North Carolina State University Graphics, NC, USA, p. 111.
- Taylor, D.P., Netscher, C., 1974. An improved technique for preparing perennial patterns of *Moloidogyne* species. Nematolgica 20, 268-269.
- Ullah, Z., Anwar, S.A., Javed, N., Khan, S.A., Shahid, M., 2011. Response of six eggplant cultivars to *Meloidogyne incognita*. Pakistan Journal of Phytopathology 23, 152-155.
- Vagelas, I., Gowen, S.R., 2012. Control of *Fusarium oxysporum* and root-knot nematodes (*Meloidogyne* spp.) with *Pseudomonas oryzihabitans*. Pakistan Journal of Phytopathology 24, 32-38.