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Occurrence and population status of root-knot nematode, *Meloidogyne incognita* on cucumber under protected cultivation in Rajasthan

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Abstract
Survey was carried out to determine the occurrence and population status of root-knot nematode in poly-house of different locations of Rajasthan. Total 40 poly-houses visited and collected 158 root and soil samples during 2 year survey. In results, it was keen observed that all samples of both soil and root were infected with root-knot nematode, *Meloidogyne incognita*. After the examination of roots 40 to 142 galls/5 g roots and 26 to 135 egg masses/5 g root of cucumber found in poly-houses of different locations. While, number of larvae ranging between 545 to 1460 J_2 per 100 cc soil in poly-house on cucumber. It was also recorded that 100% occurrence with above economic threshold level (ETL) of root-knot nematode found in visited poly-houses.

Keywords: Population status, *Meloidogyne incognita*, cucumber, protected cultivation, Rajasthan

1. Introduction
Cucumber (*Cucumis sativus*) is a widely cultivated crop belongs to family- Cucurbitaceae. It is highly nutritive vegetable and a good source of vitamins and minerals (Vitamin K, Vitamin C, pantothenic acid, biotin, molybdenum, potassium, phosphorus, copper and magnesium). The Protected cultivation is a need of present agriculture scenario of the world. The application of protected cultivation technology is feasible in almost all the agro-climatic conditions of India for the cultivation of high value crops. Under protected cultivation growers gain high productivity and better quality.

Due to favourable environmental conditions in protected structures, multiplications of plant parasitic nematodes are very high and significantly reduce the quantity and quality of production [7]. The plant parasitic nematodes associated with vegetables are *Meloidogyne* spp., *Rotylenchulus reniformis*, *Heterodera* spp., *Pratylenchus* spp., *Helicotylenchus* spp. and *Hoplolaimus* spp. in India. The losses caused by root-knot nematode, *M. incognita* and *M. javanica* on pea, okra, tomato and bottle gourd are reported to the extent of 46.0, 46.70, 47.80, and 55.40%, respectively [17]. [1]Researchers estimated 41.30-45.50 %, 37.50-41.52 % and 22.45-25.38 % losses in light, medium and heavy soils caused by root-knot nematode, *Meloidogyne incognita* on okra. The estimated crop yield losses have been reported to the extent of 16.67%, 18.20%, 21.35%, 14.10%, 27.21% and 10.54% in brinjal, cucurbits, jute, okra, tomato and rice, respectively in India [3, 12]. Among plant parasitic nematodes, *Meloidogyne incognita* is most important and causing great damage to crops grown under poly-houses including cucumber [8]. The losses are comparatively higher in cucumber under protected cultivation due to mono-cropping, favourable soil and environmental conditions etc. Keeping this in view, present investigation was carried out to find out the occurrence and population status of root-knot nematode infecting cucumber in poly-houses.

2. Materials and Methods
2.1 Collection and processing of samples
Survey was carried out to determine the occurrence and population status of root-knot nematode in poly-house of different locations of Rajasthan *viz.* Udaipur, Chittorgarh, Rajsamand, Hanumangarh, Jaipur, Dausa, Nagaur and Jodhpur. Soil samples were collected from the 40 different locations of 8 districts of Rajasthan. The samples taken from the rhizosphere of plants, labelled properly and processed in laboratory. To identification and characterization of nematode species, pure culture of juveniles of root- knot nematode Recovered from the egg mass obtained from the roots, mature females recovered from the galls of roots and the males recovered from both galls and soil of the infested field.

Plate 1: Healthy and root-knot infested root of cucumber

To find out the population status of plant parasitic nematodes, 200 cc soil samples were processed by using Cobb's Sieving and Decanting Technique [6] followed by Baermann's Funnel Assembly [5]. Roots were stained in 0.1% acid fuchsin lactophenol solution at 80ºC for 2-3 minutes [14]. Thereafter, soil extract and roots were examined under stereoscopic binocular microscope then the process of fixation, dehydration and the mounting of nematodes were performed [16]. For the identification of species of root-knot nematode the perineal patterns were prepared and the nematode species was identified [9].

Plate 2: Survey and sampling under protected cultivation for root-knot nematode

2.2 Counting of galls and egg masses

After harvesting, root samples were collected from each experiment, labelled properly and brought to the laboratory. Roots were gently washed in running tap water to remove adhering soil particles. Well cleaned roots were cut into small pieces and 5g samples were taken and observed thoroughly under microscope for counting of galls and egg masses.

3. Results

The plants in poly-houses examined during the survey exhibited symptoms like stunted, patchy growth and yellowing of leaves etc. During survey, it was observed that cucumber, tomato and chilli are highly susceptible to the attack of root-knot nematode, because most of the plants uprooted from the poly-houses showed innumerable knots in the root systems and severe damage to the cucumber, tomato and chilli crops. The results presented in table 1 indicates that the 100 % occurrence of root knot nematode was recorded in all poly-houses were samples collected from different locations of 8 districts of Rajasthan like Maharaj ki kheri, Tana, Jalkhari, Nandoli, Tana, Narayanpura, Piladar, Odan, Bikarni, Dundiya, Nimbahera, Ghosunda, Rithola, Jaisinghpura, Kashmor, Kapasan, Kankroli, Baghana, Kamligath, Bhim, Lasani, Nohar, Shahpura, Chomu, Maulasar, Osian etc.

It is apparent from the number of galls/5g roots, number of egg masses/5g roots and J_2 counts given in the table 1 that the incidence of root-knot nematode was observed in all locations of Udaipur, Chittorgarh, Rajsamand, Hanumangarh, Jaipur, Dausa, Nagaur and Jodhpur districts of Rajasthan. The highest incidence of root-knot nematode recorded from Tana, Surajpole, Manda gulfmroshan, Bikarni, Nandoli and Nohar locations. Jodhpur, Nagaur, Jaipur and Rajasamand districts of Rajasthan. Whereas, minimum infestation but above ETL was observed in Badola farm, Krishna nursery, Kashmor and Piladar locations.

In results it was keen observed that 40 to 142 galls/5g roots and 26 to 135 egg masses/5 g root of cucumber found in poly-houses of different locations. While, number of larvae ranging between 545 to 1460 J_2 per 100 cc soil in poly-house on cucumber. It was also recorded that 100% occurrence with above economic threshold level (ETL) of root-knot nematode found in poly-houses on cucumber.

Table 1: Occurrence and Population status of root-knot nematode, *Meloidogyne incognita* on cucumber under protected cultivation in Rajasthan

S. No.	Location	No. of samples Collected	Number of samples containing *Meloidogyne* spp.	Number of galls/ 5g roots	Number of egg masses /5g roots	Number of larvae/100cc soil	Occurrence (%)
I.	**Udaipur:**						
1.	Maharaj ki khedi	8	8	104	97	1156	100
2.	Jalkedi-I	4	4	88	78	875	100
3.	Jalkhedi-II	4	4	72	57	705	100
4.	Jalkhedi-III	5	5	68	49	695	100
5.	Nandoli	4	4	105	99	1192	100
6.	Narayanpura	6	6	87	73	720	100
7.	Piladar, Jaisamand road	2	2	91	83	925	100
8.	Bikarni, Chirva	2	2	107	102	1220	100
9.	CTAE Campus, Udaipur	3	3	95	86	950	100
	Total*/Mean	38*	38*	90.77	80.44	937.55	100
II.	**Chittorgarh:**						
10.	Tana via Dundiya, Teh. Bhopalsagar	18	18	142	135	1460	100
11.	Manda gulfroshan, Nimbahera	6	6	130	118	1350	100
12.	Choti sadri road, Nimbahera	8	8	109	101	1235	100
13.	Ghosunda	2	2	80	74	1025	100
14.	Rithola	2	2	126	115	1255	100
15.	Jaisingh pura	2	2	82	75	1105	100
16.	Krishna nursery- Chittorgarh	2	2	47	36	580	100
17.	Surajpole-Chittorgarh	4	4	132	120	1375	100
18.	Kashmor	2	2	65	52	725	100
19.	Near to Kapasan	4	4	68	55	785	100
20.	Kheri, kapasan road-Chittorgarh	4	4	76	64	850	100
21.	Kapasan road-Chittorgarh	4	4	62	48	660	100
	Total*/Mean	58*	58*	93.25	82.75	1033.75	100
III.	**Rajasamand:**						

22.	Odan, Nathdwara road	2	2	84	72	930	100
23.	Badola farm house Near rameshwaram temple-Rajsamand	4	4	40	26	545	100
24.	Bhana-Kankroli	4	4	97	83	1115	100
25.	Bagana-Devgarh	4	4	72	67	840	100
26.	Kirado ka badiya, Kamlighat, Devgarh	4	4	85	75	1005	100
27.	Bhim	4	4	88	77	1095	100
28.	Karera road-Bhim	4	4	118	107	1325	100
29.	Nedi, Karera road-Bhim	4	4	102	94	1155	100
30.	Selma, Bhim	4	4	63	49	725	100
31.	Lasani	4	4	52	44	660	100
32.	Amet	2	2	98	85	1120	100
	Total*/Mean	40*	40*	81.72	70.81	955.90	100
IV.	**Hanumangarh:**						
33.	Nohar-I	2	2	124	112	1410	100
34.	Nohar-II	2	2	102	88	1306	100
	Total*/Mean	4*	4*	113	100	1358	100
V.	**Jaipur:**						
35.	Mahar khurd, Shahapura	2	2	74	62	1140	100
36.	Nindola, Khejroli, Chomu	2	2	116	105	1205	100
	Total*/Mean	4*	4*	95	83.50	1172.50	100
VI.	**Dausa:**						
37.	Gijgarh	2	2	104	93	980	100
	Total*/Mean	2*	2*	104	93	980	100
VII	**Nagaur**						
38.	Dabada, Maulasar	4	4	79	61	760	100
39.	Maulasar	4	4	59	48	912	100
	Total*/Mean	8*	8*	69	54.5	836	100
VIII.	**Jodhpur:**						
40.	Agarwal farm, Osian	4	4	64	54	956	100
	Total*/Mean	4*	4*	64	54	956	100
	Grand Total*/Mean	158*	158*	88.87	77.38	1028.96	100

3.1 Number of galls per 5g roots

Results revealed that maximum 107 galls/5g roots found in the poly-house of Bikarni village followed by 105 and 104 galls/5g roots in poly-house of Nandoli and Maharaj kikhedi village of Udaipur. While, minimum 68 galls/5g roots observed in poly-house of Jalkhedi-III of Udaipur.

It was observed that maximum (142) galls/5g roots of cucumber found in poly-house situated at Tana village followed by 132 and 130 galls/5g roots in Surajpole and Munda gulfroshan sites of Chittorgarh district on cucumber. Whereas, minimum (47) galls/5g roots of cucumber found in poly-house of Krishna nursery Chittorgarh.

In Rajasamnd district maximum galls/5g roots (118) recorded on cucumber from the poly-house established at Bhim-Karera road followed by 102 and 98 galls/5g roots of cucumber in the poly-house of Nedi and Amet locations. Whereas, minimum (40) galls/5g roots of cucumber found in poly-house of Badola farm.

Results also showed that 124 and 102 galls/5g roots on cucumber under poly-house found from Nohar-I and Nohar-II locations of Hanumangarh with an average of 113 galls/5g roots. In Jaipur 74 and 116 galls/5g roots reported from cucumber in poly-house of Mahar khurd and Nindola locations.

It was observed that 104 galls/5g roots of cucumber under poly-house found from Gijgarh-Dausa and 64 galls/5g roots reported on cucumber from poly-house of Agrawal farm-Osian, Jodhpur. While, 79 and 59 galls/5g roots of cucumber in poly-house observed from Dabada and Maulasar village of Nagaur.

3.2 Number of egg masses per 5g roots

Results showed that maximum 102 egg masses/5g roots found in the poly-house of Bikarni village followed by 99 and 97 egg masses/5g roots in poly-house of Nandoli and Maharaj kikhedi village of Udaipur. While, minimum 49 egg masses/5g roots observed in poly-house of Jalkhedi-III of Udaipur.

It was observed that maximum (135) egg masses/5g roots of cucumber found in poly-house situated at Tana village followed by 120 and 118 egg masses/5g roots in Surajpole and Munda gulfroshan sites of Chittorgarh district on cucumber. Whereas, minimum (36) galls/5g roots of cucumber found in poly-house of Krishna nursery Chittorgarh.

In Rajasamnd district maximum egg masses/5g roots (107) recorded on cucumber from the poly-house established at Karera road-Bhim followed by 94 and 85 egg masses/5g roots of cucumber in the poly-house of Nedi and Bhana-Kankroli locations. Whereas, minimum (26) egg masses/5g roots of cucumber found in poly-house of Badola farm.

Results also revealed that 112 and 88 egg masses/5g roots on cucumber under poly-house found from Nohar-I and Nohar-II locations of Hanumangarh with an average of 100 egg masses/5g roots. In Jaipur 62 and 105 egg masses/5g roots reported from cucumber in poly-house of Mahar khurd and Nindola locations.

It was observed that 93 egg masses/5g roots of cucumber under poly-house found from Gijgarh-Dausa and 54 egg masses/5g roots reported on cucumber from poly-house of Agrawal farm-Osian, Jodhpur. While 61 and 48 egg masses/5g roots of cucumber in poly-house observed from Dabada and Maulasar village of Nagaur.

3.3 Final nematode population per 100 cc soil

Results revealed that maximum 1220 juveniles/100 cc soil found in the poly-house of Bikarni village followed by 1192 and 1156 juveniles/100 cc soil in poly-house of Nandoli and Maharaj kikhedi village of Udaipur. While, minimum 695 juveniles/100 cc observed in poly-house of Jalkhedi-III of Udaipur.

It was observed that maximum (1460) juveniles/100 cc soil of cucumber found in poly-house situated at Tana village followed by 1375 and 1350 juveniles/100 cc soil in the poly-house of Surajpole and Munda gulfroshan sites of Chittorgarh district on cucumber. Whereas, minimum (660) juveniles/100 cc soil of cucumber found in poly-house of Krishna nursery Chittorgarh.

In Rajasamnd district maximum juveniles/100 cc soil (1325) recorded on cucumber from the poly-house established at Karera road-Bhim followed by 1155 and 1120 juveniles/100 cc soil of cucumber in the poly-house of Nedi and Amet locations. Whereas, minimum (545) juveniles/100 cc soil of cucumber found in poly-house of Badola farm.

Results also showed that 1410 and 1306 juveniles/100 cc soil on

cucumber under poly-house found from Nohar-I and Nohar-II locations of Hanumangarh with an average of 1358 juveniles/100 cc soil. In Jaipur 1140 and 1205 juveniles/100 cc soil reported from cucumber in poly-house of Mahar khurd and Nindola locations.

It was observed that 980 juveniles/100 cc soil of cucumber under poly-house found from Gijgarh-Dausa and 956 juveniles/100 cc soil reported on cucumber from poly-house of Agrawal farm-Osian, Jodhpur. While 760 and 912 juveniles/100 cc soil of cucumber in poly-house observed from Dabada and Maulasar village of Nagaur.

4. Discussion

The results of the survey assured that root-knot nematodes are vastly dispersed in the poly-houses of Rajasthan at different locations. During survey, it was observed that cucumber was found very susceptible to the root-knot nematode and when plants uprooted there showed innumerable knots on the root systems. Root-knot nematode cause severe damage to the cucumber and very high population ranging from 705 to 1460 IJs/100 cc soil were found in poly-houses from different locations during 2016 and 2017.

Similar results were also reported by Rao *et al.*, 2007 and found the association of plant parasitic nematodes *viz.*, *Meloidogyne incognita., M. javanica, Rotylenchulus reniformis, Helicotylenchus sincises, Pratylenchus delattrei, Tylenchorhynchu scapitatus, Xiphinema sp., Tylenchus sp., Criconema sp.* and *Aphelenchus sp.* on vegetables including chilli tomato, brinjal, okra, cucurbits and cluster bean. Chandel *et al.* (2010) [4] conducted a trial in Himachal Pradesh for find out the population of major plant parasitic nematodes in 214 greenhouses associated with Sweet pepper, carnation, cucurbits, tomato and cauliflower. Study revealed that presence of *Meloidogyne incognita, Helicotylenchus dihystera* and *Pratylenchus* spp. ranging from 8 to 5604, 15 to 2560 and 5 to 795/200 cc soil, respectively. [18] Collected samples from 15 poly-houses of Solan district of Himachal Pradesh. He was found that major nematode species *viz., M. incognita,T. mashhoodi* and *H. dihystera* found only in few poly-houses with low numbers in capsicum crop and the poly-houses growing tomato and cucumber were found heavily infested with *M. incognita* up to 80 per cent.

A survey conducted by Ismail *et al.* (2012) [11] to determine the incidence and distribution of root-knot nematodes, *Meloidogyne* spp. infecting cucumber in open-field and plastic tunnel and found that two root-knot nematode species. *M. incognita* and *M. javanica,* were identified from 200 samples. *M. incognita* was predominantly found species compare to *M. javanica*. Similarly work also were recorded (Singh *et al.,* 2012; Bem *et al.,* 2014; Gatuam *et al.,* 2014; Manju and Subramanian, 2015) [20, 1, 10, 13] and found that root-knot nematode mostly associated with poly-house crops. Singh and Khanna (2015) [20] carried out survey about 81 poly-houses from 52 localities of different districts of Himachal Pradesh and reported that *Meloidogyne incognita, M. hapla, Pratylenchus* sp., *Helicotylenchus* spp., *Mesocriconema* sp., *Tylenchorhynchus sp.* and *Hoplolaimus* sp. are major plant parasitic nematodes of poly-house. Out of these nematodes *M. incognita* (37- 1200/200cc soil) was found mostly followed by *Helicotylenchus dihystera* (28-832/200cc soil).

5. Conclusion

In summarising up of the findings on the survey concluded that root-knot nematode, *Meloidogyne incognita* is a major key pest of protected cultivation in Rajasthan. It causes severe infection in poly-houses and cause great losses to cucumber. It was observed during survey that incidence of root-knot nematode fluctuates with the age of establishment of Poly-houses structure. Root-knot nematodes population increase with the increase the exposure of time of poly-houses and nematodes population found minimum in the newly established poly-houses.

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