

Original Research Article

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Status of Root-Knot Nematode, *Meloidogyne hapla* Infection on Carrot at Kodaikanal Hills of Tamil Nadu, India and Its Yield Loss Estimation

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ABSTRACT

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A survey was conducted in Kodaikanal hills of Tamil Nadu, India to know the association of root-knot nematode, *Meloidogyne hapla* in carrot (*Daucus carota* subsp. *sativus*). The nematode was found in eight carrot growing villages surveyed at Kodaikanal. Infestation was high (150-300 nematodes/200 cm³ soil) at Poomparai followed by moderate (50-150 nematodes/200 cm³ soil) at Poondi, Kookal, Mannavanur and Shenbananur villages. Low density (0-30 cysts/200 cm³ soil) was observed at Kookal, Shenbananur, Poondi, Gundupatti and Pallangi villages. The population density was low (0-50 nematodes/200 cm³ soil) at Kodaikanal and Mannavanur villages. A field study was conducted to estimate the avoidable yield loss due to natural populations of *M. hapla* on carrot. The 19.1% root tuber yield loss was observed due to natural populations of *M. hapla*. It was also estimated that *M. hapla* infestation leads to 59.3% market loss due to root tuber malformation. Pot experiment was also conducted to confirm the yield loss estimation using challenge inoculated populations of *M. hapla*. Results revealed that inoculum level of 10,000 juveniles/pot could cause tuber yield loss of 55.1%. It is concluded that efforts are needed to reduce the *M. hapla* infestation and to check further spread in carrot fields at Kodaikanal hills.

Introduction

Carrot (*Daucus carota* subsp. *sativus*) is an important root tuber vegetable crop grown globally for its rich nutritional contents of vitamin A and carotenes. In India, it is cultivated in 23 states with total area of 6.8 million ha and produced 10.93 million tonnes annually. Tamil Nadu state is the second largest producer of carrot in India that growing carrots in 1.3 million ha with 3900 tonnes of production (Welfare, 2016; Seenivasan, 2017a). To date, the northern root-knot nematode (*Meloidogyne hapla* Chitwood) is the only species that infects carrot grown on soils in Nilgiris and

Kodaikanal hills of Tamil Nadu (Anita and Selvaraj, 2011; Devrajan and Seenivasan, 2002). It is a sedentary root endo-parasitic nematode. The second-stage infective juveniles (J2) penetrate through growing tips of roots and forms several multinucleate giant cells in vascular tissues. It leads to formation of galls, digitation and constriction on taproots (Devrajan *et al.*, 2004). In addition, it causes seedling death, stunting of young plants, branching of taproots which reduces quality and yield (Devrajan *et al.*, 2003; Seenivasan and Devrajan, 2007). Even low soil densities of root-knot nematodes have

been reported to reduce marketable root yields. On mature carrot, infection by *M. hapla* causes small galls on secondary roots. If galling occurs during the seedling stages, the carrot roots can become severely stunted or forked and therefore unmarketable. Severe infections and damage to carrot by *M. hapla* have occurred more frequently in recent years in Kodaikanal hills (Nair *et al.*, 2015; Seenivasan, 2017b). However, there is no systematic information on the incidence of *M. hapla* and their yield loss potential in Kodaikanal regions. Hence, this study was conducted with the following objectives viz., 1) To study the distribution and intensity of root knot nematode in carrot at Kodaikanal hills, and 2) To estimate the avoidable yield loss due to challenge inoculated and natural populations of *M. hapla* in carrot under pot and field conditions.

Materials and Methods

Field survey

A survey was undertaken during 2014-2015 in carrot fields at Kodaikanal hills of Tamil Nadu (Fig. 1). A total of seventy-five fields were selected randomly from Shenbananur, Kodaikanal, Kookal, Gundupatti, Poomparai, Mannavanur, Poondi and Pallangi villages of Kodaikanal hills. Five plants from each field were uprooted and observed for gall index on a 0-5 scale: 0 = no galls and no forking; 1 = 1-10 galls on secondary roots, taproot not affected; 2 = 11-50 galls, none coalesced, taproot with light forking; 3 = 51-100 galls with some coalesced, forking; 4 = more than 100 galls with many coalesced, severe forking; 5 = more than 100 galls, mostly coalesced, severe forking (Seenivasan, 2010b). Soil samples collected from the same five plants were pooled to form a composite sample (200 cm³). Soil samples were processed by Cobb's decanting and sieving technique followed by modified Baermann's

funnel technique (Southey, 1986). The nematodes collected were killed by heat and fixed by adding equal volume of 4% formalin. The number of *M. hapla* juveniles in suspension was counted under a stereoscope microscope at 40x magnification. Permanent slides were made for nematode specimens from each field and species identity confirmed based on the morphometry using a Kozo Zoom 630 microscope equipped with Biowizard image analysis software (Seenivasan *et al.*, 2012).

Yield loss due to natural populations of *M. hapla*

A field trial was conducted to estimate the avoidable yield loss due to *M. hapla* in carrot at Horticultural Research Station, Kodaikanal during the Kharif season 2014 (July 2014-September 2014). The field was naturally infested with *M. hapla*. The carrot cv. Kuroda seeds were used for field trial. The experiment was laid out in paired *t*-test design with twelve replications. The treatments consisted of carbofuran 3 G @ 2 kg a.i./ha and untreated control. The individual plot size was 3 x 5 m². Each plot was separated by raised bunds leaving 0.5 m space between each bund. Carbofuran were mixed with sand and applied in soil before sowing. Seeds were sown at 30 x 10 cm spacing. Standard agronomic practices for carrot cultivation were followed for raising the crop (Suryaprabha and Satheeshkumar, 2015).

The population density of *M. hapla* in soil from each plot was determined before sowing and harvest. Each sample comprising of 10 random cores collected at a depth of 15-20 cm and pooled together into a composite sample. A sub-sample of 200 cm³ from each composite sample was processed by Cobb's sieving technique followed by the modified Baermann funnel method (Southey, 1986). The population of second stage juveniles (J2)

of *M. hapla* was counted under a stereoscope microscope at 40x magnification. Plants were harvested on 110 DAS and root tuber yield recorded from all plots. Yield was expressed in tonne (t) per ha. Five plants from each plot were collected and root gall index was assessed on a 0-5 scale. Then secondary roots from each plant were collected, cut into 2 cm length, stained with acid fuschin-lactophenol and the adult females were counted under a stereo zoom microscope (Seenivasan and Murugan, 2011).

Yield loss due to challenge inoculated populations of *M. hapla*

A pot study was conducted to estimate the avoidable yield loss due to *M. hapla* in carrot at Horticultural Research Station, Kodaikanal during June 2015- August 2016 under artificial inoculated conditions. *Meloidogyne hapla* population used in this study was isolated from carrot plants collected at Shenbaganur village and pure cultures were maintained on tomato Cv. Local (Seenivasan, 2011). Egg masses of *M. hapla* were picked from tomato roots, allowed to hatch in a beaker of distilled water and the hatched juveniles (J2) were used for inoculation. The treatments consisted of carbofuran 3 G @ 2 kg a.i./ha and untreated control replicated twelve time. Five kg soil capacity clay pots containing autoclaved pot mixture soil (sand: red earth: farmyard manure – 2:1:1) considered as one replicate. Carbofuran treatment was done before sowing. The five seeds of carrot cv. New Korada were sown in each pot. Then *M. hapla* J2 @ 10,000/pot was used as inoculums. The plants were maintained at glass house and irrigated once in a day. Plants were fertilized with 20-20-20 (N-P-K) fertilizer at 0.1 % concentration at twenty days intervals. No pest or disease was recorded during the study. The plants were carefully uprooted at 90 days after inoculation. Soil of 200 cm³ was taken and

used for *M. hapla* J2 estimation. The nematodes extracted and counted as described earlier. The root gall index and number of females per gram roots were done as explained earlier.

The data collected from both yield loss studies were analyzed and compared by *t*-test following Panse and Sukhatme (1954).

Results and Discussion

Survey

Among 75 carrot fields surveyed, about 69.3% of carrot fields in Kodaikanal hills were infected with root-knot nematode. In all the fields, *M. hapla* was the only root-knot nematode species present. The nematode was found in all the villages surveyed at Kodaikanal. The incidence rate was 100% in Poomparai village, 90% in Shenbananur and Poondi, and 60% in Kookal, Mannavanur and Pallangi villages. The population of *M. hapla* varied from 0 to 311 nematodes/ 200 cm³ soil with an average of 112 (Table 1). The population density was high (150-300 nematodes/200 cm³ soil) at Poomparai, followed by moderate (50-150 nematodes/200 cm³ soil) at Kookal, Shenbananur, Poondi, Gundupatti and Pallangi. The population density was low (0-50 nematodes/200 cm³ soil) at Kodaikanal and Mannavanur (Figure 1). The lowest gall index 1.1 was observed at Kodaikanal and the highest (3.3) at Poomparai. The association of *M. hapla* was reported earlier from Nilgiris hills in Tamil Nadu (Anita and Selvaraj, 2011). This is the first detailed report of *M. hapla* incidence and their intensity status on carrot fields at Kodaikanal hills.

Field study

Results of field study showed that carbofuran application significantly reduced both soil and

root populations of *M. hapla* at harvest compared to the un-treated control. Carbofuran 3G treatment resulted in reduction of *M. hapla* by 90 % in soil and 87.1 % in roots. The root gall index was significantly lesser (1.1) in carbofuran treatment than un-treated control (4.1). The plots treated with carbofuran before sowing had significant increased root tuber yield when compared to the control. The yield loss due to *M. hapla* on carrot estimated in the present study was 19.1% root tuber yield per ha. It was also estimated that *M. hapla* infestation leads to 59.3 % market loss due to root tuber

malformation which would be rejected in farms itself before marketing. In Nilgiris hills, *M. hapla* was reported to affect the germination of seeds by 15% and carrot yield by 36% (Anita and Selvaraj, 2011; Seenivasan and Sundarababu, 2007). In Karnataka state, the root malformation due to *M. hapla* reported to cause marketable yield loss of up to 86% (Guru *et al.*, 2014). When compared to the earlier reports, the yield loss reported in this study was relatively less that is attributed to the differences on population density of *M. hapla* (Kumar *et al.*, 2014; Seenivasan, 2010a) (Tables 2 and 3).

Table.1 Occurrence, intensity and infection of *Meloidogyne hapla* on carrot in different carrot growing villages of Kodaikanal hills in Tamil Nadu

Name of Place	No of fields surveyed	Percent Occurrence	J2 population/ 200 cm ³ soil		Gall Index	
			Range	Mean	Range	Mean
Shenbananur	10	90.0	0-112	98	0-3	1.7
Kodaikanal	10	40.0	0-72	49	0-2	1.1
Kookal	5	60.0	0-131	112	0-3	1.9
Gundupatti	10	50.0	0-126	91	0-3	2.3
Poomparai	10	100.0	182-311	290	1-4	3.3
Mannavanur	10	60.0	0-107	50	0-2	1.3
Poondi	10	90.0	0-138	102	0-3	1.8
Pallangi	10	60.0	0-102	68	0-3	2.1

J2- second stage juvenile of *M. hapla*

Table.2 Avoidable yield loss in carrot cv. New Korada due to natural populations of *M. hapla* under field conditions

Treatments	Initial J2 population /200 cm ³ soil	Final J2 population /200 cm ³ soil	No. of adult females/g root	Root gall index	Tuber yield (t/ha)	Percentage of malformed tubers
Carbofuran 3 G @ 2 kg a.i/ha	124.3	38.7 (90.5)	9.3 (87.1)	1.1	27.37 (19.1)	6.7
Untreated control	126.7	389.3	72.1	4.1	22.13	59.3
t-value	1.1	6.21**	10.08**	8.13**	11.63**	6.11**
S.Ed	NS	32.11	3.63	0.33	1.67	2.32
CD (5%)	NS	65.37	7.81	0.71	3.17	4.65

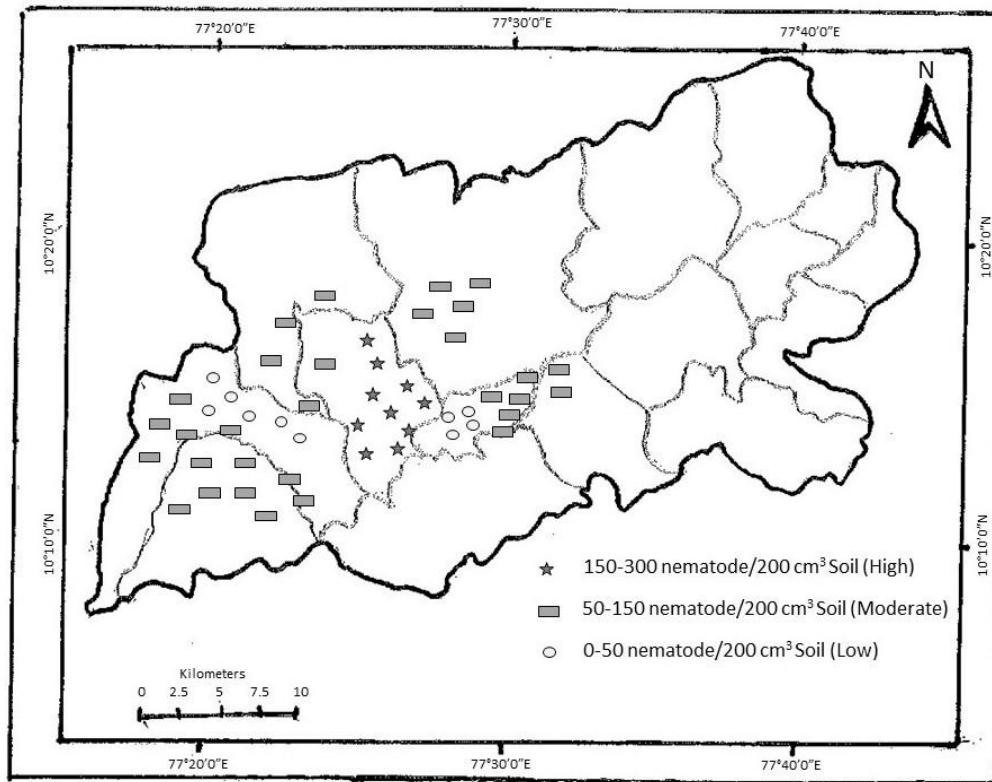
J2- second stage juvenile of *M. hapla*; ** - Highly significant (P=0.001); NS - Non-significant; Figures in the parenthesis are percent decrease or increase over untreated control.

Table.3 Avoidable yield loss in carrot cv. New Korada due to artificial inoculated populations of *M. hapla* in pots

Treatments	Final J2 population /200 cm ³ soil	No. of adult females/g root	Root gall index	Tuber yield (g/plant)
Carbofuran 3G @ 2 kg a.i/ha	41.3 (94.9)	6.7 (92.2)	1.7	207.3 (55.2)
Untreated control	822.1	86.3	4.7	92.7
t-value	9.2**	11.3**	9.6**	12.6**
S.Ed	31.7	6.1	0.3	20.6
CD (5%)	60.3	13.2	0.6	42.3

J2- second stage juvenile of *M. hapla*; ** - Highly significant (P=0.001); Figures in the parenthesis are percent decrease or increase over untreated control.

Fig.1 Distribution pattern of *Meloidogyne hapla* in carrot fields at Kodaikanal hill of Tamil Nadu



Pot study

The results of pot study were similar to field study. However, the extent of damage and yield loss was relatively higher in pot studies. Carbofuran 3G treatment resulted in reduction of *M. hapla* populations by 94.9 % in soil and 92.2 % in roots. The root gall index was

significantly lesser (1.7) in carbofuran treatment than un-treated control (4.7). The plants treated with carbofuran before sowing had significant increased root tuber yield when compared to the control. The yield loss due to challenge inoculated populations of *M. hapla* was estimated as 55.1% root tuber yield per plant. Similar results also reported by

Sivakumar and Vadivelu (1996) who observed 51% of yield loss on carrot under artificial inoculated conditions in pot study.

In conclusion, the root-knot nematode, *M. hapla* occurred in moderate to high densities in almost all the carrot-growing villages of Kodaikanal hill regions. They can cause 19.1-55.0% loss in quantity and 59.3 % loss in quality that may pose serious threat to carrot cultivation. Hence, there is an urgent need to formulate strategies not only for the management but also to check further spread of *M. hapla* in carrot fields at Kodaikanal hills.

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