EFFECT OF NURSERY TREATMENT AND BARE ROOT DIPS IN FORMULATIONS OF PLANT EXTRACTS ON *MELOIDOGYNE INCOGNITA* IN TOMATO

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Summary. An experiment was conducted under field conditions to assess the efficacy of nursery treatments with carbofuran and root dips in three nematicidal formulations based on plant extracts (Max Raze, Max Cannon and Neem Gold) on the root-knot nematode, *Meloidogyne incognita*, and on growth and yield of tomato. Seedlings from the nursery bed treated with carbofuran were free from galls while those from the untreated nursery bed showed an average galling index of 2.4. Seedlings from the treated nursery bed, when transplanted into an infested field, showed better plant performance, produced more fruit, and suppressed the nematode population compared to seedlings from the untreated infested nursery bed. All plant extract formulations controlled the root-knot nematode and increased yield of tomato, though Max Raze and Max Cannon were significantly more effective than Neem Gold.

Keywords: Carbofuran, control, root-knot nematode, *Solanum lycopersicum*.

The tomato crop suffers heavy damage due to plant parasitic nematodes in general and to the root-knot nematode *Meloidogyne incognita* (Kofoid *et* White) Chitwood in particular. Losses to the tune of 33.3% due to this nematode have been reported in Solan (HP) (Anonymous, 1998). Chemical control of plant-parasitic nematodes usually involves the use of synthetic nematicides. However, apart from their very high cost, increased concern for the environment has necessitated a reduction in the amount of nematicides used for controlling nematodes (Adegbite and Adesiyan, 2005). Also, the search for other efficient, ecologically sound and safe control methods has increased greatly. The use of plant materials containing nematotoxic principles is one of such methods. Among the so-called botanicals, derivatives from neem (*Azadirachta indica* A. Juss.) have been found to have potential for controlling plant parasitic nematodes (Mojumder, 1995; Byomakesh *et al.*, 1998; Khanna and Sharma, 1998; Sharma, 2000; Khanna and Kumar, 2006; Kumar and Khanna, 2006). Several reports show that bare root dip treatments with neem-based formulations improve plant performance and reduce nematode populations (Vats and Nandal, 1995; Vijayalakshmi and Mojumder, 2000).

In areas where root-knot nematodes are widespread, finding a field free of nematodes, to be used as a nursery, could be difficult. Also, small galls and recent infection of the roots by these nematodes may be overlooked while selecting seedlings for transplanting in the field. Therefore, investigations were conducted to ascertain the efficacy of three commercial plant extract formulations, Max Raze, Max Cannon and Neem Gold, used as root dips, on plant status, control of the root-knot nematode *M. incognita*, and yield of tomato, using seedlings grown in a nursery infested with the nematode and either treated or not with the nematicide carbofuran.

MATERIALS AND METHODS

The trials were conducted during the 2008 growing season in the experimental farm of the University of Horticulture and Forestry.

Raising of tomato seedlings. Tomato seeds (cv. Manisha) were treated with Bavistin (0.1%) + Indofil M-45 (0.25 %) to control damping off and sown in two nursery beds infested with the root-knot nematode (>250 nematodes per 250 cm^3 soil). One nursery bed was treated with carbofuran 3G at 6 g/m^2 before sowing and the other was left untreated. When four weeks old, tomato seedlings from both nurseries were randomly uprooted and observed for root galling.

Tested formulations. The three formulations were Max Raze (Max Grow Pvt Ltd.), Max Cannon (Max Grow Pvt Ltd.) and Neem Gold (Southern Petro Chemicals). Neem Gold contain 0.03% of azadirachtin. Active ingredients and their concentrations in Max Raze are extracts from *Ocimum sanctum* L. (10%) and *Andropogon nardus* L. (30%), samesol (25%), nerolidol (20%) and dissolving agent (15%). Max Cannon contains 10,000 ppm of azadirachtin. Max Raze is considered a high performance, broad spectrum bio-pesticide and is formulated under high quality control by a unique extraction technology. Max Cannon is also considered a broad spectrum bio-pesticide. Based on concentrations (1 ml/l of water i.e 0.1%) recommended for use against insect pests, a 1% (v/v) water concentration of each formulation was prepared and tested against the nematode.

Field trial. Seedlings from the untreated and treated nursery beds were dipped separately in the 1% solutions of each formulation. Dipping exposures were 1, 2, and 3 hours. Precaution was taken to ensure that only the root part of each seedling was dipped in the solution. Controls were seedlings dipped in distilled water for the same periods. The seedlings thus dipped were transplanted (in mid-April 2008) separately into 2 m × 2 m field plots infested with an average nematode population of 250 second stage juveniles/200 cm^3 soil. There were five rows of plants per plot, each with five plants, for a total of 25 plants per plot. Plots transplanted with undipped seedlings from untreated and treated nursery beds were maintained as Control-1 and Control-2, respectively. Each treatment was replicated three times according to a randomized block design.

All cultural practices (fertilization, fungicide treatments) recommended in the area were followed during the course of the experiment. Observations on plant status, viz. shoot and root lengths, were recorded at the end of the experiment, i.e. 80 days after transplanting. Also, soil samples, each composed of 8-10 sub-samples (= 250 cm^3) per plot, were collected to the depth of 20-30 cm, following a zig-zag collection pattern, and mixed. The final population of second stage juveniles per 200 cm^3 of soil was estimated by the Cobb decanting and sieving technique, as modified by Schindlers (Schindlers, 1961). Five plants per plot were also collected and their root gall index was rated on a 1-5 scale (1 = no gall, 2 = 1-10 galls, 3 = 11-30 galls, 4 = 31-99 galls, 5 = 100 galls and above) (Kumar and Khanna, 2006). Fruit yield was recorded from time to time, as fruits matured, and cumulative yields were compared. Correlations between root gall index and nematode population and nematode population and yield were calculated.

Statistical analysis. All data were subjected to analysis of variance and critical differences (CD) were calculated.

RESULTS AND DISCUSSION

Roots of seedlings grown in the nursery bed treated with carbofuran were completely free of galls while those from the untreated bed showed an average moderate galling index of 2.4.

Increasing dipping periods did not significantly affect shoot height, root length or nematode soil population density, but significantly reduced root galling and increased yield (Table I). Plants from seedlings from the treated nursery bed dipped in Max Raze attained a maximum mean height of 116.4 cm, followed by those dipped in Max Cannon and Neem Gold with mean plant heights of 109.4 and 95.2 cm, respectively. Mean shoot lengths of the plants raised in the untreated bed and dipped in Max Raze, Max Cannon and Neem Gold reached the values of 93.2, 84.1 and 74.8 cm, respectively. Control plants from treated and untreated beds transplanted into the nematode infested field but not treated with any formulation remained significantly smaller, with average heights of 69.9 and 46.6 cm, respectively. This difference in height of the two controls was significant, thereby indicating the positive impact of the nursery treatment.

The roots of the plants raised in the treated bed and also dipped in Max Raze, Max Cannon or Neem Gold were significantly longer than their respective counterparts raised in the untreated bed. Average root lengths of 26.2, 24.4 and 22.0 cm were attained in the former as compared to 21.2, 19.7 and 17.6 in latter. The roots of the plants raised in the treated bed but receiving no further treatment (Control-1) were also significantly longer (18.8 cm) than those from the untreated bed (Control-2), which were only 10.8 cm long (Table II).

Nematode soil populations (Table III) declined significantly in all the treatments receiving plant extract formulations as compared to their respective controls. Seedlings from the treated bed, when the roots were dipped in the test pesticides, harboured fewer nema-

Table I. Effect of nursery treatment with carbofuran and plant extract based formulations, used as bare root dips, on shoot length of tomato in a field infested with *Meloidogyne incognita*.

Treatment	Shoot length (cm) at dipping periods of (hrs)			Mean
	1	2	3	
Treated Nursery + Max Raze	108.7	121.0	119.7	116.4
Treated Nursery + Max Cannon	103.3	110.7	114.3	109.4
Treated Nursery + Neem gold	88.3	99.3	98.0	95.2
Treated Nursery (Control-1)	65.3	72.0	72.3	69.9
Un Treated Nursery + Max Raze	80.0	104.0	95.7	93.2
Un Treated Nursery + Max Cannon	79.7	91.3	81.3	84.1
Un Treated Nursery + Neem gold	73.0	76.7	74.7	74.8
Un Treated Nursery (Control-2)	48.7	44.0	47.0	46.6
Mean	80.8	89.8	87.9	

$C.D_{0.05}$

Dipping period (P)	N.S.
Formulation (F)	15.2
P × F	N.S.

Table II. Effect of nursery treatment with carbofuran and plant extract based formulations, used as bare root dips, on root length of tomato in a field infested with *M. incognita*.

Treatment	Root length (cm) at dipping periods of (hrs)			Mean
	1	2	3	
Treated Nursery + Max Raze	24.5	26.3	27.8	26.2
Treated Nursery + Max Cannon	20.5	26.5	26.1	24.4
Treated Nursery + Neem gold	21.8	21.9	22.4	22.0
Treated Nursery only (Control-1)	18.3	19.2	19.0	18.8
Un Treated Nursery + Max Raze	19.9	21.4	22.4	21.2
Un Treated Nursery + Max Cannon	18.7	20.1	20.5	19.7
Un Treated Nursery + Neem gold	18.0	16.3	18.5	17.6
Un Treated Nursery (Control-2)	11.4	11.7	9.4	10.8
Mean	19.1	20.4	20.8	
$C.D_{0.05}$				
Dipping period (P)	N.S.			
Formulation (F)	2.42			
P × F	N.S.			

Table III. Effect of nursery treatment with carbofuran and plant extract based formulations, used as bare root dips, on final soil population density of *M. incognita* on tomato.

Treatment	Final nematode population (juveniles per 200 cm^3 soil) at dipping periods of (hrs)			Mean
	1	2	3	
Treated Nursery + Max Raze	153.3	94.7	86.7	111.6
Treated Nursery + Max Cannon	173.3	154.7	98.3	142.1
Treated Nursery + Neem gold	200.7	185.0	170.0	185.2
Treated Nursery only (Control-1)	258.0	340.0	293.3	297.1
Un Treated Nursery + Max Raze	261.3	187.3	184.7	211.1
Un Treated Nursery + Max Cannon	264.7	224.0	205.0	231.2
Un Treated Nursery + Neem gold	270.0	231.3	223.0	241.4
Un Treated Nursery (Control-2)	408.7	470.0	434.0	437.6
Mean	248.8	235.9	211.9	
$C.D_{0.05}$				
Dipping period (P)	N.S.			
Formulation (F)	70.6			
P × F	N.S.			

todes than those from the untreated bed. The lowest average nematode population (111.6 second stage juveniles/200 cm^3 soil) was recorded in the plot transplanted with healthy seedlings from the treated bed that were further dipped in Max Raze, which, however, did not differ significantly from the population (142.1 juveniles/200 cm^3) in plots in which seedlings had been dipped in Max Cannon before transplanting. Neem Gold was not as effective as Max Raze, as the nematode population in the soil was significantly larger than that in plots treated with Max Raze and similar to that of plots treated with Max Cannon. However, all plots planted with seedlings from the untreated bed and dipped in any of the test formulations showed similar soil nematode populations that were significantly smaller than that in Control-2, which was the largest (437.6 juveniles/200 cm^3 soil).

Root gall indices were significantly different not only amongst formulations but also amongst dipping durations (Table IV). The mean gall index (3.4) of the roots dipped for one hour in the plant extract solutions was significantly higher than the root galling (3.0) of the roots dipped for two or three hours, which were, instead, similar. Thus, dipping for two hours in any of the pesticide appeared to be the most appropriate period to control the nematode. Roots of Control-1 plants showed an average gall index of 3.7, significantly less than the gall index of 4.5 recorded in Control-2 plants. Among the tested formulations, the least root galling (2.0) was observed on the plants from the treated bed dipped in Max Raze for two hours prior to transplanting. Root galling was similar on plants from the treated bed that were dipped in Max Raze or Max Cannon. Plants from plots whose seedling roots had been dipped in Neem Gold showed significantly heavier galling.

Significant effects of nursery treatment, dip formulation, duration of dipping and their interaction were observed on the yield of tomato (Table V). This was significantly greater in all plots receiving seedlings from the treated bed that were treated with any of the tested formulations as compared to those from the untreated bed. Also, a significantly larger yield of 7.1 kg per plot was

Table IV. Effect of nursery treatment with carbofuran and plant extract based formulations, used as bare root dips, on gall index of tomato in a field infested with *M. incognita*.

Treatment	Gall index[1] at dipping period (hrs) of			Mean
	1	2	3	
Treated Nursery + Max Raze	2.4 (1.5)	2.0 (1.4)	2.0 (1.4)	2.1 (1.5)
Treated Nursery + Max Cannon	2.6 (1.6)	2.2 (1.5)	2.2 (1.5)	2.3 (1.5)
Treated Nursery + Neem gold	3.2 (1.8)	2.6 (1.6)	2.6 (1.6)	2.8 (1.7)
Treated Nursery only (Control-1)	4.2 (2.0)	3.4 (1.8)	3.4 (1.8)	3.7 (1.9)
Un Treated Nursery + Max Raze	3.2 (1.8)	2.8 (1.7)	2.6 (1.6)	2.9 (1.7)
Un Treated Nursery + Max Cannon	3.6 (1.9)	3.0 (1.7)	2.8 (1.7)	3.1 (1.8)
Un Treated Nursery + Neem gold	3.6 (1.9)	3.4 (1.8)	3.4 (1.8)	3.5 (1.9)
Un Treated Nursery (Control-2)	4.6 (2.1)	4.4 (2.1)	4.6 (2.1)	4.5 (2.1)
Mean	3.4 (1.8)	3.0 (1.7)	3.0 (1.7)	

C.D $_{0.05}$	
Dipping period (P)	0.07
Formulation (F)	0.11
P × F	N.S.

[1]Gall index rated according to the 0-5 scale (as in Materials and Methods)
Figures in parentheses are square root transformed values.

Table V. Effect of nursery treatment with carbofuran and plant extract based formulations, as bare root dips, on yield of tomato in a field infested with *M. incognita*.

Treatment	Yield (kg/plot) at dipping period of (hrs)			Mean
	1	2	3	
Treated Nursery + Max Raze	10.0	12.9	13.9	12.3
Treated Nursery + Max Cannon	8.9	12.6	13.4	11.6
Treated Nursery + Neem gold	7.6	9.8	10.2	9.2
Treated Nursery only (Control-1)	7.3	7.8	6.3	7.1
Un Treated Nursery + Max Raze	6.8	8.2	8.4	7.8
Un Treated Nursery + Max Cannon	5.8	6.3	7.4	6.5
Un Treated Nursery + Neem gold	4.3	5.3	5.8	5.1
Un Treated Nursery (Control-2)	2.7	2.5	2.2	2.4
Mean	6.7	8.2	8.4	

C.D $_{0.05}$	
Dipping period (P)	0.68
Formulation (F)	1.10
P × F	1.92

recorded in Control-1 as compared to 2.4 kg per plot in Control-2, clearly showing the beneficial effect of the treatment in the nursery. The greatest yield of 13.9 kg was recorded in the plots transplanted with seedlings from the treated bed that were dipped in Max Raze for three hours, closely followed by those dipped in Max Cannon for the same period (13.4 kg per plot). Significantly larger yields of 8.2 and 8.4 kg were attained in the plots transplanted with seedlings dipped for two and three hours, respectively, as compared to the 6.7 kg in the plots transplanted with seedlings dipped for one hour only. This demonstrated that a two hour dipping period is the optimum to increase yields. Amongst formulations, Max Raze and Max Cannon were found more effective than Neem Gold.

Our experiment revealed that nursery treatment played a significant role in reducing nematode infection, improving the plant status and increasing yield of tomato. Athough all the formulations tested as root dips improved plant growth, reduced root galling and increased yield, better results were obtained with Max Raze and Max Cannon than with Neem Gold. In earlier work, neem formulations, such as Neem seed kernel extract and Econeem, used as root dips improved plant status and reduced root galling in tomato (Kumar, 2006). As root dips are most economical and highly effective in protecting the crop against *M. incognita*, they may be preferred to drenching.

In conclusion, combining soil treatments in the nursery with dipping of seedlings in neem-based formulations appears to be a sound approach to control root-knot nematodes effectively. This approach requires only a small amount of nematicide and, therefore, is economic and has very low impact on the environment.

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