

Research Papers

Nematicides control rice root-knot, caused by *Meloidogyne graminicola*

MUJEEBUR RAHMAN KHAN, BUSHRA ZAIDI and ZIAUL HAQUE

Department of Plant Protection, Aligarh Muslim University, Aligarh, 202002, India

Summary. Studies were conducted to determine damage potential of *Meloidogyne graminicola* on the commonly grown rice cv. Sugandh-5 and to devise an effective management strategy. The nematicides were applied through root-dip (200 ppm solution) and soil application of 2 kg ha⁻¹ phorate 10G (25 mg a.i./pot), carbofuran 3G (83.3 mg a.i./pot and 1 L ha⁻¹), carbosulfan 20EC (5 µL/pot) and chlorpyrifos 20 EC (6.25 µL/pot) in both nematode infested and non-infested soil with five modes of application viz., root-dip, single soil application (15 days after transplanting), root-dip + one soil application, two soil applications, and root-dip + two soil applications (15 and 30 days). Application of nematicides did not cause any toxicity symptoms on rice plants. In nematode infested soil, terminal and spiral galls developed on the rice roots, and plants suffered 20–31% decrease in the plant growth parameters. Carbofuran and phorate through root-dip plus single soil application provided greatest suppression in galling (16–20%), egg mass production (18–22%) and soil population (27.5–58.2%) of *M. graminicola*, and subsequently increased all the plant growth variables by 9–19%. Root-dip + two soil applications increased plant growth and suppressed nematodes, but was equal to root dip + one soil application. Root-dip treatment alone with carbosulfan also significantly suppressed root galling (10–12%) and improved the dry weight of roots and shoots (7–10%).

Key words: chemical control, phorate, carbofuran, carbosulfan, chlorpyrifos,

Introduction

Rice root-knot nematode, *Meloidogyne graminicola*, is an emerging threat to rice cultivation in various rice growing regions of South East Asia where rice is extensively cultivated (Gaur and Pankaj, 2010). In India, this nematode is one of the major constraints to decreasing rice production, with 16–32% yield loss reported due to the pathogen (Biswas and Rao, 1971; Rao and Biswas, 1973) in upland, rain fed lowland rice (Jairajpuri and Baqri, 1991; Prot *et al.*, 1994b) and irrigated rice (Netscher and Erlan, 1993; Prot *et al.*, 1994a; Prot and Matias, 1995). Yield losses due to the nematode are greater under flooded conditions than in upland rice (Kinh *et al.*, 1982; Prot *et al.*, 1994a; Prot and Matias, 1995).

There are various methods available for the management of rice root-knot nematode including fallowing, flooding, deep ploughing, biological control and nematicide application. Despite concern about the use of chemical pesticides throughout the world, due to adverse effects on the ecosystem (Haq *et al.*, 1990), chemical pesticides are still the most effective means of management of nematodes in the rice ecosystem (Prasad *et al.*, 2010). The pesticides are preferred by farmers as they give instant results while other disease management practices only begin to give visible impact after considerable periods.

Nematicides can act by contact and/or systemic action. They can be applied through different modes such as soil application (Jain and Bhatti, 1988), seed treatment (Jain and Gupta, 1990), bare root-dip treatment (Venkatarao *et al.*, 1987; Jain and Bhatti, 1988) and nursery bed treatment (Jain and Gupta, 1990). A few nematicides can also be applied as foliar sprays

Corresponding author: M.R. Khan
Fax: 91 571 2703516
E-mail: mrkhan777in@yahoo.co.in

as they show basipetal systemic movement in plants (Johnson, 1995).

Application of carbofuran in rice fields effectively controlled *M. graminicola* (Rahman, 1991; Soriano and Reversat, 2003). Increase in the nematode population was suppressed in fields planted with seedlings from nurseries treated with carbofuran at 1 kg a.i. ha⁻¹ (Prasad *et al.*, 2006). Carbosulfan and chlorpyrifos at the concentrations of 0.1 and 0.2%, respectively, when applied as seedling root-dip treatments for 6 h, were found effective against rice root-knot nematode in a greenhouse experiment, and increased the rice growth and reduced galling and final soil population of the nematode (Deka *et al.*, 2002). Carbosulfan has been found effective in reducing the galls and egg masses, and improved yield (Mohanty *et al.*, 2000). Seedlings grown from the carbosulfan-treated seeds had fewer galls and egg masses per root system, and the treatment reduced the nematode population up to 82% (Rahman and Das, 1994). Carbofuran, phorate and carbosulfan reduced galling when applied at the rate of 1 kg a.i. ha⁻¹ or more, and the greatest reduction in galling (82%) was recorded with carbosulfan and phorate (Prasad and Rao 1976, 1977).

It has been generally found that a single treatment of nematicide does not provide reliable control of root-knot nematode in rice, because the applied chemical gets quickly diluted under flooded conditions (Khan and Jairajpuri, 2010; Prasad *et al.*, 2010). Hence, the present study was conducted to evaluate the effectiveness of different combinations of root-dip and soil application treatments of phorate, carbofuran, carbosulfan, and chlorpyrifos against root-knot of rice caused by *M. graminicola*. Effects of these nematicides were also examined on plant growth, biomass production, and soil second-stage juveniles (J₂) population density of the nematode.

Materials and methods

Collection of infested soil and identification of the nematode

Rice fields in the village Mahwala of Tehsil Iglas, Aligarh (Uttar Pradesh, India) were surveyed during July 2009 and 2010, and plants showing symptoms of root-knot nematode infection were collected and brought to the laboratory. Ten females were excised from the galled tissue, and their perineal patterns were prepared and observed microscopically

for identification of *Meloidogyne* species. Temporary slides of J₂ isolated from root zone soil of infected plants were prepared and morphometric measurements were taken. The nematodes were identified as *M. graminicola* (Golden and Birchfield, 1968) on the basis of perineal pattern (Yik and Birchfield, 1979) and J₂ morphological characteristics (Barker *et al.*, 1985; Handoo *et al.*, 2004).

Approximately 200–300 kg of soil (light sandy loam) was collected from patches in the fields where rice plants showing severe root galling and shoot yellowing occurred. The soil was thoroughly homogenized and ten samples each of 500–600g were processed separately using Cobb's decanting and sieving method followed by the Baermann funnel technique (Southey, 1986), to estimate the nematode population in each sample. The nematode suspension recovered from the Baermann's funnel was taken into a beaker and J₂ with hyaline tails, identified on the basis of morphological characters, were counted in a counting dish under a stereomicroscope (Handoo *et al.*, 2004). Although other nematodes were also present, including *Cephalenchus*, *Rotylenchus*, *Tylenchorhynchus*, *Hoplolaimus* and *Xiphinema* spp., their population was 31–57 juveniles kg⁻¹ soil. Since these levels were too low to cause any visible damage to rice crops, their presence was ignored. The average number of *M. graminicola* J₂ kg⁻¹ soil was estimated to be 971–1089. Therefore, in the present trials we considered the initial *M. graminicola* population density as 1000 J₂ kg⁻¹ soil. This population has been found to cause severe galling on rice (Plowright and Bridge, 1990). For non-infested treatments, soil from the same fields was collected irrespective of plants showing root-knot nematode symptoms. This soil was later autoclaved to make it fully free from nematodes. Nematode population was also estimated in the non-infested soil before autoclaving; this soil contained a heterogeneous population of 347–816 juveniles kg⁻¹ soil.

Rice nursery

Certified seeds of rice, *Oryza sativa* L., cv. Sugandh-5 were procured from an authorized dealer in Aligarh. This cultivar is widely cultivated in the Northern India and has been found to be very susceptible to *M. graminicola* (Khan and Anwer, 2011). The seeds were soaked in water for 12 h, placed in a clean, moist bag and kept in shade for another 12

h. Thereafter, the seeds were sown in 25 cm diameter earthen pots with 2 kg autoclaved soil (four parts sandy loam plus one part farmyard manure). The pots were watered daily. The seedlings reached transplanting stage (four leaves and height 12–15 cm) in 5 weeks.

Nematicides

The chemical nematicides carbofuran (Tata Holset, India), phorate (Ambuja Agrochem, India), carbosulfan (FMC, India) and chlorpyrifos (Century, India) were procured from an authorized pesticide dealer in Aligarh.

Inoculum level and nematicide doses

For root-dip treatment, seedlings of rice cv. Sugandh-5 were dipped in 200 ppm solution of phorate, carbofuran, carbosulfan or chlorpyrifos for 3 h before transplanting. For soil application in pots, the dose of nematicides was (per pot): 25 mg phorate 10G (equivalent to 2 kg a.i. ha⁻¹); 83.3 mg carbofuran 3G (2 kg a.i. ha⁻¹); 5 µL carbosulfan 20 EC (1 L a.i. ha⁻¹); and 6.25 µL chlorpyrifos 20 EC (1 L a.i. ha⁻¹).

Treatments and plant culture

To study the effect of the nematicides on plant growth, biomass production, and root-knot disease caused by *M. graminicola*, five treatment modules were given the following treatments: root-dip to transplant; soil application 15 days after transplanting; root-dip + soil application 15 days after transplanting; two soil applications at 15 and 30 days after transplanting; or root-dip + two soil applications at 15 and 30 days after transplanting. Each nematicide treatment was applied to two sets of pots, one containing soil infested with *M. graminicola* (1000 J₂/pot) and the other containing autoclaved (non-infested) soil. For each treatment, 15 replicates were maintained in a completely randomized experimental design.

Earthen pots of 15 cm diameter were each filled with 1 kg *M. graminicola* infested soil or non-infested soil and 250 g autoclaved farmyard manure (FYM). The pots having non-infested soil plus FYM were autoclaved at 15 kg cm⁻² pressure at 121°C for 15–20 minutes. Seedlings of rice cv. Sugandh-5 were transplanted at the centre of the pots and the

nematicide treatments (above) were applied. The pots with seedlings (without treatments) grown in infested and non-infested (autoclaved) soil served as experimental controls. Seedlings were watered immediately after transplanting and were then watered on alternate days until harvest. Twenty pots were maintained for each treatment which were arranged in a completely randomized experimental design. Plants were grown for 4 months. Weeds, if any, were removed manually at 15 d intervals. During the experimental period, plants were regularly observed for any visible symptom attributable to the infection of *M. graminicola* or application of nematicides. At harvest (4 months after transplanting), pots were flooded with water to achieve maximum root recovery, and the parameters of length and dry weight of root and shoot, root-knot nematode symptoms and soil population of root-knot nematode J₂ were determined for each plant.

Root-knot index

At harvest, roots of each plant were examined to count galls and egg masses. The egg masses of *M. graminicola* do not develop on root surfaces, but rather remain embedded in root tissues. Hence, they were counted by tearing the galls under stereomicroscope. In addition, ten mature females from the galled tissue were excised. Perineal patterns of the females were prepared and examined to ensure that the galls were formed by *M. graminicola*.

Soil populations of *Meloidogyne graminicola*

Population density of *M. graminicola* J₂ in soil was estimated monthly, i.e. 1, 2, 3, or 4 (harvest) months after transplanting. At each month, population density was estimated from five pots of the 20 pots initially maintained for each treatment. The soil in each pot was mixed and 500 g was processed separately for nematode isolation using Cobb's decanting and sieving method (modified) followed by Baermann's funnel technique (Southey, 1986).

Statistical analyses

The experiment was conducted during two consecutive years. Since the year difference for measured variables was not significant ($P \leq 0.05$), the data were pooled. Observations taken from ten pots (five

pots/year) were averaged to calculate means. The data (ten replicates/treatment) of plant growth were analyzed by two factor analysis of variance (ANOVA), and least significant differences (LSDs) were calculated to identify significant effects of treatments at probability levels of $P \leq 0.05$, 0.01 and 0.001 using MINITAB 11.0 for Windows-XP. The data of soil nematode populations were also analyzed using single factor ANOVA, and LSDs were calculated at the three probability levels. Percent variations over experimental controls were also calculated. The population data are presented in graphical form (Figure 1), and points have been indicated where standard errors are different according to Duncan's multiple test (DMRT).

Results

Symptoms

Plants grown in nematode infested soil (1000 J₂) were stunted and had chlorotic foliage, while their roots had nematode incited terminal galls. Application of nematicides through all modes of treatment did not cause any visible toxicity symptoms on rice seedlings. However, their application influenced gall formation and egg mass development. Root-dip treat-

ments with carbofuran resulted in 10–12% decrease in number of galls and egg masses/root system ($P \leq 0.05$). Phorate application also significantly reduced the galling but to a lesser extent (7%; Table 1).

Soil application of the nematicides resulted in 12.9–14.8% (for carbofuran) and 9–10% (for phorate) decreases in the numbers of galls and egg masses in comparison to controls. Soil application with carbosulfan and chlorpyrifos caused significant decreases in the numbers of egg masses/root system ($P \leq 0.05$, Table 1). The Root-dip followed by one soil application treatment of nematicides significantly suppressed galling and egg mass production. Level of significance, however, varied with the nematicides. The decrease in the gall formation and egg mass production was 22.3–24.3% with carbofuran ($P \leq 0.001$), 16–18% with phorate ($P \leq 0.01$), 11–12% with chlorpyrifos ($P \leq 0.05$), and 10.7–12.5% with carbosulfan ($P \leq 0.05$), in comparison to the controls (Table 1).

Effects of two soil applications of nematicide were more or less equal to the root-dip plus one soil application (Table 1). The treatments of one root-dip followed by two soil applications of nematicides resulted in the greatest decrease in galling and egg mass production of *M. graminicola* (Table 1). These two parameters decreased by 29 and 31% with car-

Table 1. Mean numbers of *Meloidogyne graminicola* galls and egg masses on rice plants to which different nematicide treatments were applied. RD, Root Dip; SA, Soil Application 15 days after transplanting; SSA, Second Soil Application 30 days after transplanting. Each value is mean of ten replicates.

Treatment	RD		SA		RD+SA		SA + SSA		RD + SA + SSA	
	Galls	Egg masses	Galls	Egg masses	Galls	Egg masses	Galls	Egg masses	Galls	Egg masses
Infested soil (IS)	58.6	43.2	58.6	43.2	58.6	43.2	58.6	43.2	58.6	43.2
IS + phorate	54.2 ^a	39.5 ^a	53.0 ^a	38.8 ^a	49.1 ^b	35.4 ^b	49.6 ^b	36.3 ^b	44.3 ^c	31.8 ^c
IS + carbofuran	52.5 ^a	38.0 ^a	51.0 ^b	36.8 ^b	45.5 ^b	32.7 ^b	46.5 ^b	33.3 ^c	41.4 ^c	29.7 ^c
IS + carbosulfan	56.60	40.9	55.5	40.0 ^a	52.2 ^a	37.8 ^b	52.7 ^a	37.9 ^a	48.8 ^b	35.4 ^b
IS + chlorpyrifos	56.84	41.5	54.9	38.9 ^a	52.0 ^a	37.9 ^a	52.6 ^a	38.3 ^a	48.6 ^b	34.9 ^b
LSD $P \leq 0.05$	2.45	2.06	2.32	2.02	2.14	2.09	2.22	2.10	2.48	2.08
$P \leq 0.01$	3.58	3.00	3.38	2.94	3.12	3.04	3.23	3.06	3.60	3.03
$P \leq 0.001$	5.36	4.50	5.10	4.41	4.67	4.56	4.85	4.59	5.41	4.54
F-value	10.11 ^z	9.70 ^z	12.40 ^z	8.56 ^z	11.23 ^z	9.52 ^z	9.94 ^z	7.25 ^z	10.58 ^z	8.40 ^z

^a, ^b, ^c, ^z Values are significantly different from the respective controls at $P \leq 0.05$ ^a, $P \leq 0.01$ ^b, $P \leq 0.001$ ^c, ^z according to LSDs.

bofuran and 24 and 26% with phorate, respectively, compared with the experimental controls ($P \leq 0.001$). Application of carbosulfan or chlorpyrifos reduced gall and egg mass production by 16–19% over the controls ($P \leq 0.01$; Table 1).

Effects on plant growth and biomass production of rice

Root-dip treatment with nematicides marginally suppressed the plant growth of rice plants, but the plant parameters were not significantly different from the controls (non-infested soil, $P \leq 0.05$, Tables 2, 3). However, in nematode infested soil, the root-dip treatments reduced the negative effect of *M. graminicola*, leading to increased shoot dry weight from carbofuran in comparison to the controls ($P \leq 0.05$, Table 3). Single soil application of the nematicides did not promote the growth of rice in nematode infested soil over controls ($P \leq 0.05$; Tables 2 and 3).

The treatments of root-dip plus single soil application promoted plant growth variables especially with carbofuran and phorate ($P \leq 0.05$). Application of carbosulfan or chlorpyrifos in nematode infested soil enhanced the shoot and root dry weights by 7–8% ($P \leq 0.05$, Table 3). Dry weights of shoots and roots were increased 7.8–9.0% with phorate treatment. The root-dip treatments followed by two soil applications with carbofuran or phorate resulted, respectively, in 14–18% and 14–16% increases in plant growth variables in nematode infested soil in

Table 2. Mean shoot and root lengths (cm) of rice plants grown in soil that was either infested or non-infested with *Meloidogyne graminicola* and to which different nematicide treatments were applied. RD, root dip; SA, soil application 15 days after transplanting; SSA, second soil application 30 days after transplanting. Each value is mean of ten replicates.

Treatment	RD		SA		RD+SA		SA + SSA		RD + SA + SSA	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
Non-infested soil (NIS)	39.6	22.3	39.6	22.3	39.6	22.3	39.6	22.3	39.6	22.3
NIS + phorate	38.8	22.0	38.7	21.8	40.3	22.6	40.1	22.5	40.4	22.8
NIS + carbofuran	39.2	22.1	38.8	21.7	40.8	23.0	40.5	22.8	41.3	23.2
NIS + carbosulfan	38.9	21.9	38.5	21.5	40.3	22.5	39.87	22.41	40.2	22.6
NIS + chlorpyrifos	39.0	22.0	38.4	21.6	39.9	22.6	39.9	22.5	40.3	22.7
Infested soil (IS)	27.8 ^c	16.7 ^c	27.8 ^c	16.7 ^c	27.8 ^c	16.7 ^c	27.8 ^c	16.7 ^c	27.8 ^c	16.7 ^c
IS + phorate	28.4	17.0	28.3	17.2	30.0 ^b	17.9 ^b	28.6	17.2	31.9 ^b	19.1 ^b
IS + carbofuran	29.5	17.6	29.5	17.6	30.6 ^b	18.7 ^b	30.6 ^a	18.1 ^a	32.2 ^b	19.3 ^b
IS + carbosulfan	27.8	16.8	27.9	16.9	28.0	16.9	28.1	16.9	29.8 ^a	17.9 ^a
IS + chlorpyrifos	28.1	16.9	28.1	17.0	28.2	16.93	28.2	17.0	29.9 ^a	18.1 ^a
LSD $P \leq 0.05$	1.77	1.02	1.98	1.05	1.52	1.08	1.87	0.95	2.06	0.83
$P \leq 0.01$	2.43	1.39	2.72	1.44	2.09	1.48	2.56	1.30	2.83	1.13
$P \leq 0.001$	3.30	1.90	3.70	1.96	2.84	2.02	3.50	1.77	3.85	1.53
F-value										
Nematicides (df = 4)	11.04 ^z	8.56 ^z	7.65 ^z	9.23 ^z	10.4 ^z	7.59 ^z	6.32 ^z	5.65 ^z	5.13 ^z	11.5 ^z
Nematode (df = 1)	854 ^z	566 ^z	753 ^z	498 ^z	803 ^z	545 ^z	654 ^z	742 ^z	512 ^z	660 ^z
Interaction (df = 4)	NS	3.75 ^x	3.23 ^x	2.98 ^x	3.12 ^x	NS	NS	NS	NS	3.70 ^x

^{a, b, c} Values are significantly different from the respective controls at $P \leq 0.05$ ^a, $P \leq 0.01$ ^b, $P \leq 0.001$ ^c according to LSD.

^{x, y, z} F-values followed by ^x ($P \leq 0.05$), ^y ($P \leq 0.01$) and ^z ($P \leq 0.001$) are significant otherwise not significant (NS) at $P \leq 0.05$. Table 3. Mean shoot and root dry weights (g) of rice plants grown in soil that was either infested or non-infested with *Meloidogyne graminicola* and to which different nematicide treatments were applied.

Table 3. Mean shoot and root dry weights (g) of rice plants grown in soil that was either infested or non-infested with *Meloidogyne graminicola* and to which different nematicide treatments were applied. RD, root dip; SA, soil application 15 days after transplanting; SSA, second soil application 30 days after transplanting. Each value is mean of ten replicates.

Treatment	RD		SA		RD+SA		SA + SSA		RD + SA + SSA	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
Non-infested soil (NIS)	1.6	1.9	1.6	1.9	1.6	1.9	1.6	1.9	1.6	1.9
NIS + phorate	1.56	1.87	1.56	1.87	1.64	1.95	1.61	1.92	1.64	1.93
NIS + carbofuran	1.58	1.89	1.57	1.88	1.68	1.97	1.65	1.95	1.67	1.98
NIS + carbosulfan	1.57	1.85	1.55	1.87	1.62	1.92	1.61	1.91	1.62	1.92
NIS + chlorpyrifos	1.56	1.86	1.54	1.86	1.63	1.93	1.61	1.91	1.62	1.92
Infested soil (IS)	1.10 ^c	1.3 ^c	1.10 ^c	1.3 ^c	1.10 ^c	1.3 ^c	1.10 ^c	1.3 ^c	1.10 ^c	1.3 ^c
IS + phorate	1.13	1.35	1.14	1.35	1.21 ^a	1.42 ^a	1.19 ^a	1.42 ^a	1.27 ^b	1.50 ^b
IS + carbofuran	1.18 ^a	1.4 ^a	1.16	1.38	1.23 ^a	1.45 ^a	1.22 ^a	1.44 ^a	1.30 ^b	1.52 ^b
IS + carbosulfan	1.11	1.32	1.12	1.32	1.19 ^a	1.4 ^a	1.12	1.40 [*]	1.20 ^a	1.40 ^a
IS + chlorpyrifos	1.12	1.33	1.12	1.33	1.18 ^a	1.41 ^a	1.13	1.41 [*]	1.19 ^a	1.42 ^a
LSD $P \leq 0.05$	0.17	0.12	0.15	0.15	0.20	0.13	0.16	0.12	0.18	0.13
$P \leq 0.01$	0.23	0.16	0.21	0.21	0.27	0.18	0.22	0.16	0.22	0.18
$P \leq 0.001$	0.31	0.22	0.28	0.28	0.37	0.24	0.30	0.22	0.33	0.24
F-value										
Nematicides (df=4)	3.45 ^x	3.56 ^x	3.23 ^x	3.65 ^x	3.15 ^x	3.03 ^x	3.32 ^x	3.62 ^x	3.56 ^x	3.13 ^x
Nematode (df=1)	246 ^z	253 ^z	308 ^z	254 ^z	203 ^z	282 ^z	246 ^z	224 ^z	246 ^z	286 ^z
Interaction (df=4)	NS	NS	3.23 ^x	NS	NS	2.98 ^x	2.95 ^x	3.08 ^x	3.30 ^x	3.07 ^x

^a, ^b, ^c Values are significantly different from the respective controls at $P \leq 0.05$ ^a, $P \leq 0.01$ ^b, $P \leq 0.001$ ^c according to LSD.

^x, ^y, ^z F-values followed by ^x ($P \leq 0.05$), ^y ($P \leq 0.01$) and ^z ($P \leq 0.001$) are significant otherwise not significant (NS) at $P \leq 0.05$.

comparison to the controls ($P \leq 0.01$; Tables 2 and 3). Treatments with carbosulfan and chlorpyrifos also significantly ($P \leq 0.05$) promoted plant growth.

Soil populations of *Meloidogyne graminicola*

Soil population density of *M. graminicola* J₂ gradually increased to more than four times the initial inoculum density (1000 J₂) by 3 months. At 4 months, the population density was less than at 3 months, but did not differ significantly at $P \leq 0.05$ (Figure 1). The population density in pots treated with different nematicides increased more slowly than in the untreated pots (Figure 1). The root-dip treatment with carbofuran gave 10–26% decrease in the soil population over the controls ($P \leq 0.05$ –0.001; Figure 1). Phorate was next most effective chemical for

suppressing the population, causing 8.2–19.7% decreases in nematode population compared to the controls ($P \leq 0.001$). The decreases in populations from soil application of nematicides was more or less similar to the root-dip treatment. The treatment of root-dip plus one soil application at 15 days after transplanting caused drastic decline in the soil populations in comparison to respective controls (Figure 1). The greatest decrease in nematode population was recorded with carbofuran (35–58% reduction compared with controls; ($P \leq 0.001$), followed by phorate (27–53%), chlorpyrifos (24–50%), and carbosulfan (22–46%). The effect of carbofuran was greater ($P \leq 0.05$) than other nematicides used (Figure 1). Two soil applications at 15 and 30 days interval also suppressed the nematode populations but the effect was less than root-dip plus one soil applica-

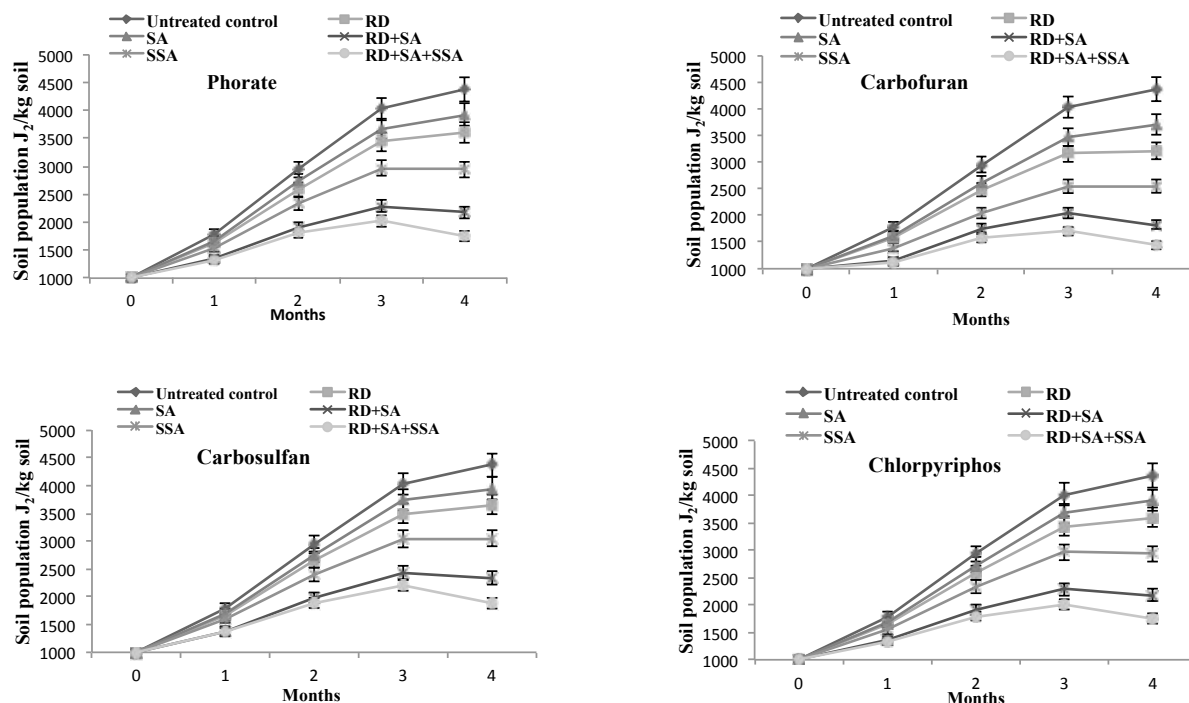


Figure 1. Mean soil populations of *Meloidogyne graminicola* (J2 kg⁻¹ soil) at 1, 2, 3, 4 (harvest) months after transplanting rice plants and different nematicide treatments. RD: Root Dip, SA: Soil Application 15 days after transplanting, SSA: Second Soil Application 30 days after transplanting. Error bars are showing standard error. Points marked by different letters are different ($P \leq 0.001$) according to Duncan's Multiple Range Test (DMRT).

tion ($P \leq 0.01$). In this treatment, application of carbofuran and phorate caused 22–42% and 15–36% decreases in the populations, respectively, over the controls. Next in effectiveness were chlorpyrifos and carbosulfan, which decreased the populations by 13–33% and 10–30% ($P \leq 0.001$). Treatments of root-dip and two soil applications of nematicides at 15 and 30 days of transplanting caused maximum reduction in the nematode population (Figure 1). The reduction was greatest, respectively, with carbofuran (37.6–67.0%) followed by phorate (30.7–63.2%) over respective control populations ($P \leq 0.001$). Treatments with chlorpyrifos and carbosulfan decreased the nematode population by 26–60% and 24–57%. Overall nematode population in this set of treatments was similar to the root-dip treatment plus single soil application of nematicides. However, the decrease in the population was 2–11% greater in root-dip plus two soil applications.

Discussion

Root-knot of rice is an emerging problem throughout the world (Khan and Jairajpuri, 2010). Due to the wide host range and adaptability to varying environments, *M. graminicola* is spreading rapidly to regions where it was not known previously. In India, 20–30 years ago, *M. graminicola* was confined to a few southern states, but now it has spread to northern states such as Kashmir (Singh *et al.*, 2007), Uttar Pradesh (Khan and Anwer, 2011) and Haryana, Himachal Pradesh, Punjab and Delhi (Dabur and Jain, 2005). The rice root-knot nematode has now become a major pest in irrigated rice in India, and causes high yield losses (Dabur and Jain, 2005; Singh *et al.*, 2007; Prasad *et al.*, 2010).

Rice plants grown in soil infested with 1000 *M. graminicola* J2 kg⁻¹ soil developed characteristic galls, i.e. terminal, hook shaped or spiral galls (Sheela *et al.*, 2005). Approximately 58 galls and 43 egg masses were formed on roots of rice plants in control pots,

indicating that cv. Sugandh-5 was highly susceptible to the nematode infestation. The plants grown in *M. graminicola* infested soil exhibited 20–31% reduction in plant growth and dry matter production over 4 months. In light soils such as the sandy loam used in present experiment, *M. graminicola* causes greater damage and yield loss that may be up to 72% (Bridge and Page, 1982; Gaur and Pankaj, 2010; Khan and Anwer, 2011).

In the present study, among the different nematicides tested, carbofuran was found most effective in suppressing *M. graminicola*, followed by phorate, carbosulfan and chlorpyrifos. Other researchers have reported the effectiveness of carbofuran against rice root-knot nematode (Rahman 1991; Mohanty *et al.*, 2000; Soriano and Reversat, 2003). Effectiveness of chemical pesticides depends largely on soil type. In light soils the chemicals percolated downwards quickly, giving lesser duration of contact of the active ingredient with the nematodes and plant root systems. Hence, the mode of application of nematicides may play an important role in light soils.

In the present study, the treatment comprising root-dip plus soil application of carbofuran at 15 days of transplanting was almost equivalent to the treatment involving root-dip plus two soil applications. Root-dip treatment in the joint application might have provided initial protection to the young plants from nematode invasion. Carbofuran is a systemic nematicide and is absorbed by plant roots, and its root-dip application has proved effective against large number of nematodes, especially endoparasites (Johnson, 1995; Prasad *et al.*, 2006). In 15 days, seedlings become established in soil and lateral roots would have emerged, providing substrate for the nematode feeding. At this time soil application of carbofuran probably suppressed the nematode juveniles present in the root zones and attacking the fresh lateral roots. This application would have killed the majority of the nematode population present in the root zones. This could be the reason why the effects of root-dip plus one or two soil application were similar. The nematicide next in effectiveness was phorate, which is also efficacious and provides satisfactory control of root-knot nematodes (Prasad *et al.*, 2010). However, phorate is largely a contact action nematicide, so root-dip treatment with this chemical did not result in absorption by roots in adequate amounts, and subsequently was less effective against *M. graminicola* than carbofuran.

The study has revealed that root-dip plus one soil application at 15 days with carbofuran can satisfactorily control the rice root-knot nematode. The application of the nematicide to soil should be done when fields are wet (2–3 days after irrigation), so the nematicides does not percolate to deep soil layers but persist in the root zones of plants, where the nematode juveniles aggregate and feed.

Literature cited

- Barker K.R., C.C. Carter and J.N. Sasser, 1985. *An Advanced Treatise on Meloidogyne Species*. Vol. 1. *Methodology*. North Carolina State University Graphics, Raleigh, NC, USA, 422 pp.
- Biswas H. and Y.S. Rao, 1971. Studies on nematodes of rice and rice soils. II. Influence of *Meloidogyne graminicola* Morgan and Birchfield incidence on yields of rice. *Oryza* 8(1), 101–102.
- Bridge J. and S.L.J. Page, 1982. The rice root knot nematode, *Meloidogyne graminicola*, on deep water rice (*Oryza sativa* subsp. *indica*). *Revue de Nématologie* 5, 225–232.
- Dabur K.R. and R.K. Jain, 2005. Rice root nematode *Meloidogyne graminicola* - a threat to rice-wheat cropping system. *Indian Journal of Nematology* 35(1), 81–82.
- Deka B.C. and P. Das, 2002. Efficacy of certain chemicals as seedling root-dip treatment against rice root-knot nematode, *Meloidogyne graminicola*. *Journal of the Agricultural Science Society of North East India* 15(1), 112–114.
- Gaur H.S. and Pankaj, 2010. Root-knot nematode infestation in rice. In: *Nematode Infestations, Part I: Food Crop* (M.R. Khan, M.S. Jairajpuri, ed.). National Academy of Sciences, India, 72–90.
- Golden A.M. and W. Birchfield, 1968. Rice root knot nematode (*Meloidogyne graminicola*) as a new pest of rice. *Plant Disease Reporter* 52, 423.
- Handoo Z.A., A.P. Nyczepir, D. Esmenjaud, J.G. Van Der Beck, P. Castagnone-Sereno, L.K. Carta, A.M. Skanter and J.A. Higgins, 2004. Morphological, molecular and differential host characterization of *Meloidogyne floridensis* n. sp. (Nematoda; Meloidogynidae), a root-knot nematode parasitizing peach in Florida. *Journal of Nematology* 36, 20–35.
- Haq S., S. Sexena and M.W. Khan, 1990. Chemical control of plant nematodes in relation to environment pollution. In: *Progress in Nematology* (S.K. Saxena, M.W. Khan, A. Rashid, R.M. Khan, ed.). CBS Publishers and Distributors, Delhi, India, 297–320.
- Jain B.K. and D.S. Bhatti, 1988. Bare root dip treatment with systemic nematicides for controlling the root knot nematode in tomato transplant. *Indian Journal of Nematology* 8, 19–24.
- Jain R.K. and D.C. Gupta, 1990. Control of root-knot nematode *Meloidogyne javanica* through nursery treatment on tomato (cv. HS101). *Indian Journal of Nematology* 15, 453–491.
- Jairajpuri M.S. and Q.H. Baqri, 1991. *Nematode Pests of Rice*. Oxford and IBH Publishing Co., New Delhi, India, 66 pp.

- Johnson A.W., G.W.Burton, J.P. Wilson and A.M. Golden, 1995. Rotations with coastal bermudagrass and fallow for management of *Meloidogyne incognita* and soilborne fungi on vegetable crops. *Journal of Nematology* 27, 457–464.
- Khan M.R. and A. Anwer, 2011. Occurrence of rice root-knot nematode and yield loss assessment in Aligarh and Hathras districts of Uttar Pradesh, India. *Indian Journal of Nematology* 41(1), 34–40.
- Khan M.R. and M.S. Jairajpuri, 2010. Nematode infestation in food crops-national scenario. In: *Nematode Infestations, Part I: Food Crop* (M.R. Khan, M.S. Jairajpuri, ed.). National Academy of Sciences, India, 1–16.
- Kinh D.N., N.M. Huong and N.U. Ut, 1982. Root-knot disease of rice in the Mekong Delta, Vietnam. *International Rice Research Newsletter* 7, 15.
- Mohanty K.C., S.N. Mahapatra and S.C. Swain, 2000. Efficacy of certain chemicals as seed treatment against *Meloidogyne graminicola* on rice. *Indian Journal of Nematology* 30(2), 233–234.
- Netscher C. and Erlan, 1993. A root-knot nematode, *Meloidogyne graminicola*, parasitic on rice in Indonesia. *Afro Asian Journal of Nematology* 3, 90–95.
- Plowright R.A. and J. Bridge, 1990. Effect of *Meloidogyne graminicola* (Nematoda) establishment growth and yield of rice cv. IR36. *Nematologica* 36, 81–89.
- Prasad S.K. and Y.S. Rao, 1976. Chemotherapy of the root knot nematodes (*Meloidogyne graminicola*) in rice. II. Evaluation of pesticides as soil drench. *Zeitschrift fur Pflanzenkrankheiten und Pflanzenschutz* 83, 730–735.
- Prasad S.K. and Y.S. Rao, 1977. Soil drench pesticide treatment to control the root knot nematode *Meloidogyne graminicola*. *International Rice Research Notes* 2, 20.
- Prasad J.S., N. Somasekhar and K.S. Varaprasad, 2010. Nematode infestation in Paddy In: *Nematode Infestations, Part I: Food Crop* (M.R. Khan, M.S. Jairajpuri, ed.). National Academy of Sciences, India, 17–71.
- Prasad J.S., Vishakanta and Gubbaiah, 2006. Outbreak of root-knot nematode (*Meloidogyne graminicola*) disease in rice and farmers perceptions. *Indian Journal of Nematology* 36(1), 85–88.
- Prot J.C. and D.M. Matias, 1995. Effects of water regime on the distribution of *Meloidogyne graminicola* and other root-parasitic nematodes in a rice field toposequence and pathogenicity of *M. graminicola* on rice cultivar UPL R15. *Nematologica* 41(2), 219– 228.
- Prot J.C., I.R.S. Soriano and D.M. Matias, 1994a. Major root-parasitic nematodes associated with irrigated rice in the Philippines. *Fundamental and Applied Nematology* 17, 75–78.
- Prot J.C., L.M. Villanueva and E.B. Gergon, 1994b. The potential of increased nitrogen supply to mitigate growth and yield reduction of upland rice cultivar UPL Ri5 caused by *Meloidogyne graminicola*. *Fundamental and Applied Nematology* 7, 445–454.
- Rahman M.L., 1991. Effect of different cropping sequences on root-knot nematode, *Meloidogyne graminicola*, and yield of deepwater rice. *Nematologia Mediterranea* 18, 213–217.
- Rahman M.F. and P. Das, 1994. Seed soaking with chemicals for reducing infestation of *Meloidogyne graminicola* on rice. *Journal of the Agricultural Science Society of North East India* 7(1), 107–108.
- Rao Y.S. and H. Biswas, 1973. Evalution of yield losses in rice due the root knot nematode, *Meloidogyne incognita*. *Indian Journal of Nematology* 3, 74.
- Sheela M.S., T. Jiji, M.S. Nisha and J. Rajkumar, 2005. A new record of *Meloidogyne graminicola* on rice, *Oryza sativa* in Kerala. *Indian Journal of Nematology* 35(2), 218.
- Singh V.K., C.S. Kalia and V. Kaul, 2007. New record of root-knot nematode, *Meloidogyne graminicola* infecting rice in Jammu. *Indian Journal of Nematology* 37(1), 94.
- Soriano I.R. and G. Reversat, 2003. Management of *Meloidogyne graminicola* and yield of upland rice in South-Luzon, Philippines *Nematology* 5(6), 879–884.
- Southey J.F., 1986. *Laboratory Methods for Work with Plant and Soil Nematodes*. Ministry of Agriculture Fisheries and Food. Her Majesty's Stationary Office, London, UK, 202 pp.
- Yik C.P. and W. Birchfield, 1979. Host studies and reaction of cultivars to *Meloidogyne graminicola*. *Phytopathology* 69, 497–499.

Accepted for publication: October 18, 2011