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Impact of Soil Infestation of Rice Root-Knot Nematode and Flooding on Its Development and Rice Yield

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Abstract

A pot experiment was conducted during July-November 2006 in a glass house at the Institute of Agriculture and Animal Science, Rampur, Chitwan to find out the impact of soil infestation of rice root-knot nematode (*Meloidogyne graminicola* Golden and Birchfield) and flooding on its development and rice yield. The experiment was conducted in a 4 factorial randomized complete block design with 5 replications. Rice cv. 'Sabitri' was sown in *M. graminicola* infested soil and infestation free (healthy) soil. Both beds were further divided to give wet bed (flooded) and dry bed condition. Twenty-one days old seedlings from each seedbed were transplanted into plastic pots containing 5 kg of *M. graminicola* infested and healthy soil each treated with lowland (continuously flooded) and upland condition. Simulated field conditions were created throughout the experiment period. Results revealed that root-knot index (RKI) and population of second stage juveniles of *M. graminicola* (J2) in soil and roots were significantly lower and grain yield was higher in pots containing seedlings transplanted from wet seedbed than dry bed. Lower RKI, root lesion index (RLI) and J2 population in soil and roots and higher grain yield were observed in pots containing seedlings transplanted from healthy seedbed than the nematode infested seedbed. Similarly, RKI, RLI and J2 population in soil and roots were significantly lower in simulated lowland as compared to upland condition. Transplanting on healthy soil also resulted in lower RKI, RLI and J2 population in soil and roots and higher grain yield than in the nematode infested soil. Interactions revealed that highest grain yield was obtained from seedlings grown under healthy seedbed transplanted in healthy soil under simulated lowland condition. Thus, *M. graminicola* infestation may be minimized by growing seedlings in healthy and wet seedbed. Seedling transplanting into *M. graminicola* free soil and lowland condition may be another important control measure for it.

Key words: dry bed, lowland, upland, wet bed, rice root-knot nematode, *Meloidogyne graminicola*

Introduction

The rice root-knot nematode (*Meloidogyne graminicola* Golden and Birchfield) is a major soil borne pest of rice with worldwide distribution including south eastern Asian countries (Bridge *et al.* 1990, Soriano & Reversat 2003). This pest has also been reported from the main rice growing areas of Nepal (Pokharel & Sharma-Poudyal 2001, Sharma *et al.* 2001, Sharma-Poudyal *et al.* 2002, Dangal *et al.* 2008, Dangal *et al.* 2009) in different rice growing environments like rain-fed upland soil, shallow flooded soil and continuous flooded soils in Nepal (Pokharel & Sharma-Poudyal 2001, Sharma *et al.* 2001, Sharma-Poudyal *et al.* 2002).

M. graminicola is considered as the most serious nematode in cultivated upland rice (Panwar & Rao 1998) and causes economic losses in upland, lowland, and deep water rice and also in rice nurseries (Bridge *et al.* 1990). Rice grain yield reduction has been reported up to 40% in nematode infested farmers' fields in

Chitwan (Sharma-Poudyal *et al.* 2002). Under high nematode population density, the yield loss was incurred up to 97% (Sharma-Poudyal *et al.* 2004).

Chemical control of root-knot nematode is very expensive, unsustainable and affects the agr-ecosystem adversely (Ahmad & Khan 2004). However, healthy seedlings exhibited more vigorous growth and appeared to have greater capacity to withstand soil-borne biological stresses in the field. Healthy seedlings grown in solarized rice nursery increased rice yield at many demonstration sites (Banu *et al*. 2005).

Reproduction, survival, and infectivity of *M. graminicola* were different in flooded and non-flooded conditions (Padgham *et al.* 2003). Soriano *et al.* (2000) reported that rice cultivar tolerance of *M. graminicola* varies with water regime. The yield losses due to *M. graminicola* could be prevented or minimized when the rice crop is flooded early and kept flooded until a late stage of development. However, detail information on the biology of root-knot nematode in rice field as well as its sustainable management is limited in Nepal (Pokharel 2007). Since, most of the commonly grown Nepalese rice cultivars are susceptible to *M. graminicola* (Sharma-Poudyal *et al.* 2004, Pokharel 2007), there may be rice yield loss in every *M. graminicola* infested nursery and field. Hence, there is an urgent need of practical nematode management options for the farmers of Nepal. Production of healthy seedlings and continuous flooding may be a simple and low-cost method to reduce the nematode dissemination and minimize pathogen pressure in soils and improve plant health in the field. Thus, this study was conducted to study the development of root-knot disease with soil health and water management level in rice.

Methodology

A pot experiment was conducted during July-November 2006 in a glass house at the Institute of Agriculture and Animal Science, Rampur, Chitwan to find the impact of soil infestation of rice root-knot nematode and flooding on its development and rice yield. The experiment was conducted in a 4 factorial randomized complete block design with 5 replications. Rice cv. 'Sabitri' was sown in *M. graminicola* infested seedbed and nematode infestation free (healthy) seedbed. Both beds were further divided to give wet bed (continuous flooding) and dry bed condition. Twenty one days-old seedlings from each seedbed were transplanted into plastic pots of 19 cm diameter containing 5 kg of *M. graminicola* infested and healthy soil each treated with lowland (continuously flooded) and upland condition. Fertilization was done with FYM @ 10 t ha^{-1} and NPK @ 50: 30:30 kg ha^{-1} as basal dose. During tillering at forty five days after transplanting (DAT), 50 kg ha^{-1} N was top dressed. Simulated field conditions were created throughout the experiment period. Plants were harvested at physiological maturity (130 DAT) and total grain weight was recorded. Roots were thoroughly washed and indexed for root-knot indexing by the use of 0 (no root swellings or galls) to 10 (all roots galled) scale according to Bridge *et al.* (2005). The roots were also indexed for root lesion indexing using 0 (healthy roots, without lesion) to 4 (lesions more than 75% of roots or more than 75% roots rotten) scale according to Sharma-Poudyal *et al.* (2002).

In order to assess *M. graminicola* second juveniles (J2) present in the rice roots, the cleaned roots were chopped into small pieces of about 10 mm length for extracting second juveniles (J2). About 2 g of chopped roots were placed in an electric blender with 100 ml of water, blended for 2 minutes and placed in a modified Baermann tray for extraction (Schindler 1961). Similarly, for extracting J2 nematodes from the rhizosphere soil, it was homogenized and 100 g working sample was taken and processed by modified Baermann tray method (Schindler 1961). After 48 h of processing, nematode suspension was collected in plastic tubes (50 ml). After allowing settlement for an hour, the final volume of suspension was reduced to 20 ml with the help of a glass pipette. Two milliliter (10%) aliquot was sampled from the 20 ml suspension in counting disc, allowed to settle for five minutes. J2 were counted under a binocular microscope (Bridge *et al.* 2000).

Nematode counts were transformed in logs ($x + 1$) for statistical analysis (Gomez & Gomez 1984). Data was analyzed with Microsoft Excel and MSTAT-C (MSTAT, Michigan State Univ., USA). Mean comparison was done by Duncan's multiple range test (DMRT).

Results

There was no significant difference in root-knot index (RKI) and root lesion index (RLI) between dry and wet seed bed. Dangal *et al.* (2009) found lower RKI but higher RLI in wet bed nurseries than in dry bed.

However, both RKI and RLI were significantly higher in *M. graminicola* infested seedbed, simulated upland transplanted condition and the nematode infested soil than in healthy seedbed, lowland and healthy soil respectively (Table 1). Lower RKI in simulated lowland was mainly due to reduced penetration of rice roots by *M. graminicola* in flooded soils (Bridge & Page 1982). Dangal *et al.* (2009) also found higher RKI but lower RLI in upland nurseries than lowland. Similarly, Prot and Matias (1995) found greater root gall indices in upland than in irrigated conditions. Soriano *et al.* (2000) also reported lower root galling under continuous flooding than under intermittent flooding. Interactions were non-significant. However, highest RKI and RLI were observed in infested dry seed bed transplanted into upland infested soil which was followed by infested wet bed transplanted into infested upland condition (Table 2).

Table 1. Development of rice root-knot disease in rice (Sabitri) at harvesting as influenced by seedbed type, seedbed health, transplanted land condition and soil health

Treatments	Root knot index (0-10)	Root lesion index (0-4)	Number of J2 per		Grain yield (g) per hill
			100g soil	2g root	
A. Seedbed					
Wet bed	4.18[†]	1.97	1.30 (63) b	2.95 (15405) b	3.63 a
Dry bed	4.40	1.97	1.44 (70) a	3.14 (22291) a	2.26 b
LSD (d” 0.05)	ns	Ns	0.10	0.15	0.29
B. Seed bed condition					
Infested	5.92 a	2.63 a	1.93 (98) a	4.07 (26484) a	2.44 b
Healthy	2.65 b	1.33 b	0.81 (35) b	2.01 (11212) b	3.45 a
LSD (d” 0.05)	0.39	0.24	0.10	0.15	0.29
C. Transplanted land					
Lowland	1.93 b	1.28 b	1.21 (44) b	2.67 (4581) b	4.35 a
Upland	6.65 a	2.68 a	1.53 (90) a	3.41 (33115) a	1.54 b
LSD (d” 0.05)	0.39	0.24	0.10	0.15	0.29
D. Transplanted soil					
Infested	5.83 a	2.68 a	1.76 (80) a	4.13 (25772) a	2.13 b
Healthy	2.75 b	1.28 b	0.98 (54) b	1.95 (11924) b	3.76 a
LSD (d” 0.05)	0.39	0.24	0.10	0.15	0.29
CV (%)	20.39	26.73	16.85	11.15	21.97

[†] Mean of 5 replications. Values without and with parenthesis are log (x+1) and original values, respectively. Same letters followed in the columns are not significantly different (P= 0.05) by DMRT. ns = not significantly different

The number of J2 recovered from soil and roots were significantly lower in wet bed, healthy seedbed, lowland and healthy transplanted soil than the in dry bed, *M. graminicola* infested seedbed, upland transplanted condition and the nematode infested soil respectively (Table 1). Similarly, Dangal *et al.* (2009) found lower J2 population in roots and soil from nurseries in wet bed and lowland than in dry bed and upland, respectively. Soriano *et al.* (2000) also found significantly lower J2 population in IR72 in sandy soil and in IR29 and IR36 in clay soil when grown under continuous flooding than in intermittent flooding. Similarly, Bridge and Page (1982) did not observe nematodes in roots of deep water rice plants after several months of flooding, whereas Prot and Matias (1995) obtained large number of J2 from the roots collected at maturity in permanently flooded rice and number of J2 recovered from roots (3g) was significantly lower under upland than in irrigated conditions. Nematodes invaded before transplanting in lowland (flooding) could develop and reproduced within the flooded roots as found by Bridge and Page (1982). There was significant influence of interaction among seedbed type, seedbed infestation, transplanted land condition and soil infestation on J2 population in soil. There was no J2 in dry or wet bed-healthy seedbed-upland or lowland-healthy soil. Lowest number of J2 (8) was found in seedlings from healthy wet bed transplanted in infested soil with simulated lowland condition (Table 2). However, highest J2 population was found in dry-infested seedbed-upland-healthy soil (172) and wet-infested seedbed-upland-healthy soil (126), wet-infested seedbed-upland-infested soil (118), wet-healthy seedbed-upland-infested soil (116), dry-infested seedbed-upland-infested soil (94), dry-healthy seedbed-upland-infested soil (92) were at par (Table 2). Conversely, interactions among different factors had no significant effect on J2 population under roots (Table 2).

Table 2. Interaction effect of seedbed type, seedbed infestation, transplanted land condition and soil infestation with *M. graminicola* on development of rice root-knot disease and J2 population in soil and root of rice (Sabitri) at harvesting

Treatments	RKI (0-10)	RLI (0-4)	Number of J2 per		Grain yield (g) per hill
			100g soil	2g root	
Wet bed × I-seedbed × Lowland × I-soil	2.80†	1.6	1.78 (68) b	3.76 (6205)	3.51 cde
Wet bed × I-seedbed × Lowland × H-soil	3	2	1.81 (68) b	3.15 (4250)	6.32 a
Wet bed × I-seedbed × Upland × I-soil	9.6	3.8	2.04 (118) ab	4.55 (38932)	0.97 ijk
Wet bed × I-seedbed × Upland × H-soil	8.2	3.2	2.08 (126) ab	4.40 (32333)	1.53 hi
Wet bed × H-seedbed × Lowland × I-soil	1.4	1.6	0.68 (8) c	3.12 (2177)	4.37 bc
Wet bed × H-seedbed × Lowland × H-soil	0	0	0.00 (0) d	0.00 (0)	4.10 cd
Wet bed × H-seedbed × Upland × I-soil	8.4	3.6	2.02 (116) ab	4.58 (39343)	2.59 fg
Wet bed × H-seedbed × Upland × H-soil	0	0	0.00 (0) d	0.00 (0)	3.24 def
Dry bed × I-seedbed × Lowland × I-soil	3.2	1.6	1.86 (76) b	4.00 (11225)	0.49 k
Dry bed × I-seedbed × Lowland × H-soil	2.4	1.8	1.75 (64) b	3.38 (3305)	0.85 ijk
Dry bed × I-seedbed × Upland × I-soil	9.8	3.8	1.96 (94) ab	4.66 (60118)	3.00 efg
Dry bed × I-seedbed × Upland × H-soil	8.4	3.2	2.20 (172) a	4.65 (55504)	4.98 b
Dry bed × H-seedbed × Lowland × I-soil	2.6	1.6	1.81 (66) b	3.94 (9485)	0.61 jk
Dry bed × H-seedbed × Lowland × H-soil	0	0	0.00 (0) d	0.00 (0)	2.28 gh
Dry bed × H-seedbed × Upland × I-soil	8.8	3.8	1.96 (92) ab	4.45 (38691)	0.82
Dry bed × H-seedbed × Upland × H-soil	0	0	0.00 (0) d	0.00 (0)	21.97
LSD (d" 0.05)	ns	ns	0.29	ns	4.10 cd
CV (%)	20.39	26.73	16.85	11.15	2.59 fg

†Mean of 5 replications. Values without and with parenthesis are log (x +1) and original values, respectively. Same letters followed in the columns are not significantly different (P= 0.05) by DMRT. I= *M. graminicola* infested, H= healthy, RKI= root knot index, RLI= Root lesion index, ns = not significantly different

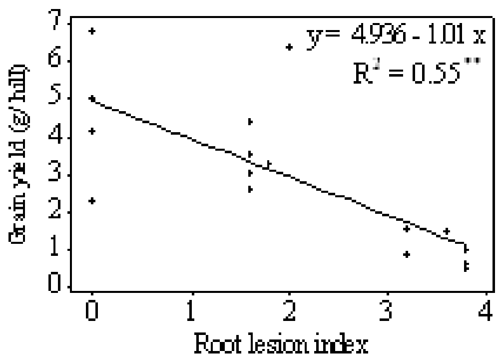

Fig. 1. Effect of RLI on grain yield

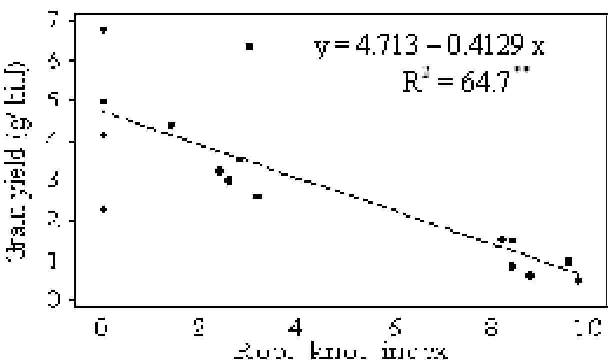

Fig. 2. Effect of RKI on grain yield

Grain yield per hill was highly negatively correlated with RLI (Fig. 1) and RKI (Fig. 2). Thus, lower grain yield was observed in plants having more RLI and RKI. Grain yield was 37.7, 29.3, 64.6 and 43.4% lower in wet bed, *M. graminicola* infested seedbed, upland and infested transplanted soil than wet bed, healthy seedbed, lowland and healthy soil, respectively (Table 2). Prot and Matias (1995) also reported less number of panicles and grain yield under upland than under irrigated conditions. Number of panicles and grain yield were reduced by the nematode under upland but not under irrigated conditions (Prot and Matias 1995). Grain yield of IR29, IR36, IR72 and IR74 were significantly greater in continuously flooded than intermittently flooded clay soil in the absence or presence of the nematode (Soriano *et al.* 2000). In addition to direct effects on the nematode, continuous flooding may increase tolerance of the rice cultivars for *M. graminicola* by increasing their yield potential (Soriano *et al.* 2000). Interaction among seedbed type, seedbed infestation, transplanted land condition and soil infestation also had significant effect on grain yield (Table 2). Highest yield was observed in wet bed-healthy seedbed-lowland-infested soil (6.76g) and was at par with wet bed-infested seedbed-lowland- infested soil (6.32g) which was followed by dry bed-healthy seedbed-lowland healthy soil (4.98). Highest grain yield reduction (92.69%) was observed in dry bed-infested seedbed-upland-infested soil.

Discussion

Since wet bed was kept in saturated condition by frequent irrigation, rice roots might escape invasion by *M. graminicola* (Bridge & Page 1982) and/or limited the spread of the nematode (Prot & Matias 1995). Due to lower infection under wet bed condition, grain yield increased but RKI and J2 number in root and soil decreased as compared to dry bed condition which was favorable for infection and development of *M. graminicola*.

Seedlings grown under *M. graminicola* infested seedbed got infected with the nematode and infected seedlings grew slowly as reported by Bridge and Page (1982) because of root damage. But, seedlings from healthy seedbed had better plant health and more growth since they were healthy even after transplanting in healthy soil or were infected by *M. graminicola* only after transplanting into the nematode infested soil. Healthy seedlings appeared to have a greater capacity to withstand soil-borne biological stresses (Banu *et al.* 2005). As a result, grain yield was increased but RKI, RLI and J2 number in root and soil were lower under healthy seedbed than the nematode infested seedbed.

Under upland condition *M. graminicola* was able to infest most of the roots which resulted in drastic reduction in root development and consequential reduction in shoot growth (Prot & Matias 1995). More damage from *M. graminicola* occurred in the aerobic upland systems (Soriano & Reversat 2003). However, rice roots under lowland (continuous flooding) might escape invasion by *M. graminicola* (Bridge & Page 1982) limiting the spread of it (Prot & Matias 1995). Permanent flooding might limit the migration of J2 between roots of the same root system (Bridge & Page 1982) resulting in a lower root damage. Reduced aeration due to high moisture levels for prolonged periods allowed poor respiration and movement of nematodes and reduced population of *M. graminicola* (Garg *et al.* 1995). The nematodes which invaded roots before transplanting under lowland could develop and reproduce within the flooded roots (Bridge & Page 1982). In addition to direct effects on the nematode, continuous flooding might increase tolerance of the rice for *M. graminicola* (Soriano *et al.* 2000). Because of the lower infection and better plant health, rice grain yield was increased but RKI, RLI and number of J2 in root and soil were decreased significantly under simulated lowland than under simulated upland condition.

Seedlings transplanted under infested soil became heavily infected with the rice root-knot nematodes and the infected plants grew slowly (Bridge & Page 1982). But, seedlings transplanted under healthy soil were free from infection if seedlings were healthy or there was lower disease development if seedlings were already infected with the nematode in nursery.

Interactions of soil health and water level had significant influence on disease development and grain yield. As a result, highest grain yield was obtained from seedlings grown under healthy seedbed transplanted in healthy soil under simulated lowland condition. Thus, *M. graminicola* infestation may be minimized by growing seedlings in wet and healthy seedbed. Seedling transplanting into *M. graminicola* free soil and lowland condition may be another important control measure for it.

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