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RESEARCH PAPERS

Interactions between the nematodes *Ditylenchus angustus* and *Aphelenchoides besseyi* on rice: population dynamics and grain yield reductions

Mohammad Abdul LATIF[1,2], Ahsanul HAQUE[2], Mohammad Islam TAJUL[3], Mohammad Abul MONSUR[2] and Mohd Yusop RAFII[1,4]

[1] Department of Crop Science, Faculty of Agriculture, University Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia
[2] Plant Pathology Division, Bangladesh Rice Research Institute, Gazipur-1701, Bangladesh
[3] Graduate School of Bio-Applications and Systems Engineering, Tokyo University of Agriculture and Technology, Japan
[4] Institute of Tropical Agriculture, University Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia

Summary. Experiments were conducted in pots and an irrigated rice (*Oryza sativa*) field to determine the impact of mixed or single nematode inoculations with *Aphelenchoides besseyi* and *Ditylenchus angustus* on nematode population dynamics and yield attributes of rice cv. BR3. The reproduction rates were greater in a single nematode species inoculated with either *A. besseyi* or *D. angustus* compared to their mixed inoculation. This was probably due to competition between the two nematode species. Greater incidences of white-tip and ufra diseases were observed with single nematode species inoculations compared to mixed species inoculations. Total yield losses ranged from 24 to 47% from mixed inoculations (different ratios) of *D. angustus* and *A. besseyi*, compared to 62% from inoculation with *D. angustus* and 16% from *A. besseyi*. Single species inoculations (with either *D. angustus* or *A. besseyi*) gave greater disease incidence and greater yield reductions than mixed inoculations with both nematodes.

Key words: mixed inoculation, competition, white-tip, ufra disease, incidence.

Introduction

Rice stem nematode, *Ditylenchus angustus* (Butler) is one of the major pests of rice (*Oryza sativa*) in Bangladesh, causing the severe disease known as ufra. This disease occurs in deep-water (Bridge *et al.*, 1990), irrigated (Latif *et al.*, 2006) and rain-fed low land rice (Miah and Rahman, 1985). Reported yield losses caused by ufra have been mentioned to be 10 to 15% in India (Rao *et al.*, 1986), 20 to 90% in Thailand (Hashioka, 1963), 50 to 100% in Vietnam (Cuc and Kinh, 1981) and 42 to 49% or occasionally 90% in Bangladesh (Latif *et al.* 2011a; b). A second major disease of rice is white-tip, caused by the seed borne nematode, *Aphelenchoides besseyi* (Christie). This disease occurs in upland, irrigated or deep-water rice in many rice-growing countries of Asia, including Bangladesh, and in Tropical America and Africa (Ou, 1985; Rahman and Miah, 1989). White-tip has also been reported from Egypt (Amin, 2002), Iran (Jamali *et al.*, 2006) and European countries including Italy (Cotoneo and Moretti, 2001) and Turkey (Ozturk and Enneli, 1997). White-tip has been reported to cause yield losses ranging from 10 to 30% in China (Wang *et al.*, 2003), 40 to 50% in the United States of America (Atkins and Todd, 1959) and 20 to 60% in India (Rao *et al.*, 1985).

Several authors have described competition between different plant parasitic nematodes (Lasserre *et al.*, 1994; Umesh *et al.*, 1994; Stetina *et al.*, 1997; Melakeberhan and Dey 2003; Brinkman *et al.*, 2005). The ectoparasites, *Tylenchorhynchus agri* and *Para-*

Corresponding author: M.A. Latif
Fax: + 603 89435973
E-mail: alatif1965@yahoo.com

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trichodorus minor reduced *Meloidogyne naasi* infection by inhibiting root growth of creeping bentgrass, and decreasing the availability of feeding sites for *M. naasi* (Sikora *et al.*, 1979). Sedentary endoparasitic nematodes can suppress ectoparasites through physiological changes rather than mechanical effects, as with the interaction between *M. hapla* and *Xiphinema americanum* (Eisenbach and Griffin, 1987). Three endoparasitic root feeding nematodes that are frequently found with *Ammophila arenaria* are *Meloidogyne maritima* (Jepson) Karssen, van Aelst & Cook, *Heterodera arenaria* (Cooper) Robinson, Stone, Hooper & Rowe and *Pratylenchus penetrans* Cobb (De Rooij-van der Goes *et al.*, 1995). These nematodes colonize new root layers of host plants in sequence, each showing a peak in abundance at different times of the year (Van der Stoel *et al*., 2002). Absence of the other endoparasitic species may lead to a change in temporal population dynamics and different effects on plant growth. In greenhouse experiments, the three endoparasitic nematode had very little or no effects on plant growth when added alone or in combination (Van der Stoel, 2001; Brinkman *et al*., 2004).

The diseases ufra and white tip are very common in rice fields, and both can be present on individual rice plants. There have been no reports on population dynamics of *D. angustus* and *A. besseyi* and their competition with each other in infested rice plants. In the present study, we examined how the population dynamics of each of these nematodes are affected by the presence of the other species. We compared the multiplication rates of the two nematodes in single or mixed inoculations on rice. We also investigated effects of infestation with single or combinations of the two nematodes on rice yields and yield components, in rain-fed pots and irrigated field experiments.

Materials and methods

Nematode culture and inoculum preparation

Ditylenchus angustus was collected from infested plants of the rice cv. BR3 and *A. besseyi* from plants of cv. BR11, from a farmer's field in the Gazipur district, Bangladesh. Infected stems or seeds were cut into small pieces and immersed in distilled water in petri-dishes for approx. 4 h to release nematodes into the water. Nematodes were then collected by sieving (20 μm mesh sieve) and identified under a compound microscope (Nikon AFX-IIA, Japan). Both nematodes were separately cultured on the fungus, *Rhizoctonia solani* following the techniques described by Latif and Mian (1995) and Latif *et al.* (1997).

Effects of mixed inoculations of *Aphelenchoides besseyi* and *Ditylenchus angustus* on population dynamics and yield components in rain-fed pots

Soil and plant material

A pot experiment was conducted in a net house at the Bangladesh Rice Research Institute (BRRI), Gazipur, during the rain-fed season in 2003, with rice cultivation soil [pH (H_2O), 5.8; 0.9% organic matter, total C, 20.5 g kg^{-1}; total N, 2.2 g kg^{-1}). Clean and healthy matured seeds of rice cv. BR3 (germination rate >85%) were pre-germinated in a moist plastic tray (25 × 10 cm) in the dark at 28°C. Three-day-old sprouted seeds were planted at approx. 0.5 cm depth in each earthen pot (diam. 25 cm, height 30 cm) using sterilized forceps. The pots were then put in open space and left for 150 d in rain-fed conditions. Irrigation and weeding were performed as necessary, following the practices described by BRRI (2003). Two mL solution of Hyponex® fertilizer [containing (mg L^{-1}): N (100), P (200), K (100), Mg (10), Mn (0.02), and B (0.1)] was added to each pot 20 d after planting. No pesticides were used during the experimental period.

Inoculation of the rice seedlings

Twenty days after sowing (DAS), the rice seedlings in each pot were inoculated at the base with nematodes at the rate of approx. 100 nematodes/plant, as described by Rahman (1993) and Latif *et al.* (1997). The proportions of juveniles, males and females inoculated were 40, 25 and 35% for *D. angustus* and 36, 28 and 36% for *A. besseyi*. Treatments were ratios of *A. bessey*: *D. angustus* of 100:0, 0:100, 50:50, 75:25 or 25:75, and a non-inoculated control treatment was included. Initial penetration of nematodes into plants in the different treatments was confirmed by extraction of nematodes, as described by Rahman (1993) and Latif *et al*. (1997).

Effects of mixed inoculation of *Aphelenchoides besseyi* and *Ditylenchus angustus* on population dynamics and yield components in a field trial

An experiment was conducted in irrigated rice fields during 2003 to 2004, at BRRI, Gazipur. Well decomposed cowdung was applied to soil at 10 t ha^{-1}. Experimental plots were fertilized with the recommended rates of N–P–K–S–Zn fertilizer (124–26–60–13–4 kg ha^{-1}; BRRI, 2003). The procedure and method for inoculation of cv. BR3 seedlings with nematodes was as described for the pot experiment (above). Seedlings of BR3 were inoculated five treatments of *A. bessey*: *D. angustus* ratios of 100:0, 0:100, 50:50, 75:25 or 25:75, and an uninoculated control treatment was included. Based on chlorotic symptoms of ufra (symptom appeared at the bases of young leaves) and white-tip (symptom appeared at the tips of young leaves) at 20 d after inoculation, the infected seedlings were transplanted into 2 × 2 m plots at the rate of one seedling per hill. The distance between hills, and rows of hills, was 20 cm. Each plot was surrounded with a 15 to 20 cm high mud plastered boundary to prevent spread of nematodes from one plot to another. The crop was harvested from the central 1.5 × 1.5 m area of each plot. For the nematode population study, destructive samples with clear symptoms of ufra and white-tip at six different rice growth stages were taken from outside of the 1.5 m × 1.5 m harvest plots.

Table 1. Average temperatures and relative humidities recorded during the periods of the pot and field experiments.

Year	Month	Temperature (°C)			Relative humidity (%)	
		Max	Min	Mean	9.00h	14.00h
Pot experiment, 2003						
2003	Jun	31.3	25.7	28.5	83.7	72.9
2003	Jul	32.3	26.5	29.4	80.9	66.9
2003	Aug	32.7	26.8	29.7	80.4	69.6
2003	Sep	32.0	26.1	29.0	82.5	70.7
2003	Oct	31.8	24.6	28.2	80.7	68.9
2003	Nov	29.8	17.9	23.8	70.4	46.1
Field experiment, 2003 to 2004						
2003	Nov	29.8	17.9	23.9	70.4	46.1
2003	Dec	26.2	14.9	20.5	72.3	45.8
2004	Jan	24.0	12.7	18.4	80.5	53.1
2004	Feb	28.5	14.4	21.5	72.1	40.7
2004	Mar	32.8	21.6	27.2	75.3	44.9
2004	Apr	32.4	23.5	27.9	78.9	57.9
2004	May	35.2	26.0	30.6	73.8	56.3

Nematode extraction and enumeration

Infected rice stems of symptomatic samples were each opened with a sharp needle, cut into small pieces (approx. 0.5 cm) and immersed in water of a Petri dish for 3h. Similarly, for rice seeds, spikelets were cut into two pieces, lemmae and paleae were separated from endosperms, and immersed into water in a Petri dish for 4 h to release nematodes. Nematodes were collected by sieving (20 μm mesh sieve), and observed under a stereo microscope (Nikon AFX-IIA, Japan) for enumeration. To avoid the enumeration of non plant parasitic nematodes, each sample was cross checked by standard compound microscope for confirmation of the two plant parasitic nematodes, based on their stylet shapes.

Data recording and assessment of nematode populations and reproduction rate

Data of temperature and relative humidity (RH) were recorded during the experimental period. Total duration of pot and field experiments was twelve months (Table 1). In the field experiment, numbers of nematodes per hill were recorded, at six rice plant growth stages (tillering, 30 d after transplanting (DAT); booting, 75 DAT; flowering, 90 DAT; dough, 105 DAT; ripening, 120 DAT; harvesting, 135 DAT). The incidence of ufra and white-tip, numbers of rice panicles m^{-2}, proportions of healthy panicles (%), 1000 grain weights (g), grain yields (t ha^{-1}) and yield loss (%) were measured at harvest. Disease incidence (%) was measured according to number of infected tillers per hill while yield was measured from harvested crop from the central 1.5 × 1.5 m area of each 2 × 2 m plot, or on a per hill basis.

The reproduction rates of the two nematode species were calculated according to the following equation: Rf = Pf / Pi, where, Rf is the reproduction factor or rate, Pf is the final nematode population, and Pi is the initial nematode population. Rf was determined at each rice plant growth stage for each treatment.

Statistical analyses

In the pot experiment, treatments were arranged in a completely randomized design with 28 replications. Each pot represented a replication. Three replications were destructively sampled at each of six rice growth stages. A total of 18 (three by six) replications were destructively sampled from 28 replications for population dynamics study, and the remaining 10 replications were used for the study of disease incidence and yield parameters. For the field study, the experiment was laid out in a randomized complete block design with four replications. Six treatments along with non-inoculated (control) were followed both in pot and field experiments (see above). All data were statistically analyzed using CropStats software. Statistical analyses were performed by analysis of variance (ANOVA) followed by Fisher's least square difference (LSD) test. Mean separation was based on LSDs at $P \leq 0.05$ (Fisher, 1960).

Results

Nematode population dynamics in the pot and field experiments

The populations of either *D. angustus* or *A. besseyi* were greater in single inoculations compared to their mixed inoculations (Figure 1A) in the pot experiment. In mixed inoculations, particularly at the 50:50 ratio (T3), the population of *D. angustus* was greater compared to *A. besseyi* at all plant growth stages. For mixed inoculations, the greatest nematode populations per hill were observed at the dough stage, followed by ripening, harvesting, booting and tillering stages (Figure 1A, T3-T5). However, in single inoculations, the population of *D. angustus* was similar at the dough and ripening stages (Figure 1A), while in the field trial they varied (Figure 1B). Regarding the population dynamics, a similar trend was observed in the field trial, but the populations of both nematodes declined in rain-fed pot trial (Figure 1B). The populations of nematodes declined when *D. angustus* and of *A. besseyi* were applied in mixed inoculations compared to average populations of two nematode species after single inoculations in both the pot and field experiments (Figure 2).

In the pot experiment, the reproduction rate from the single inoculation treatments ranged from 4.5 to 10.5 for *A. besseyi* and 7.9 to 49.7 for *D. angustus*, while the rates ranged from 0.9 to 12.6 in the mixed inocula-

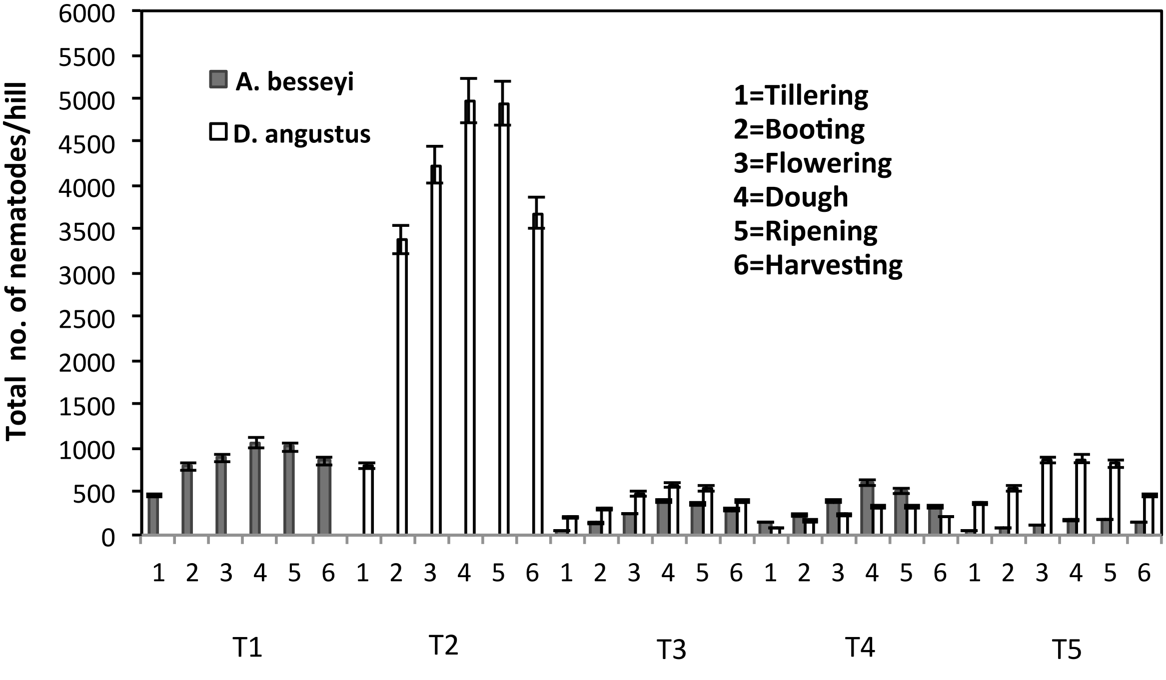

Figure 1A. Population dynamics of *Aphelenchoides besseyi* and *Ditylenchus angustus* at different rice plant growth stages in the pot experiment, 2003. Treatments (T), *A. besseyi* : *D. angustus* inoculation ratios, T1 = 100:0; T2 = 0:100; T3 = 50:50; T4 = 75:25; T5 = 25:75. Vertical bars indicate standard errors.

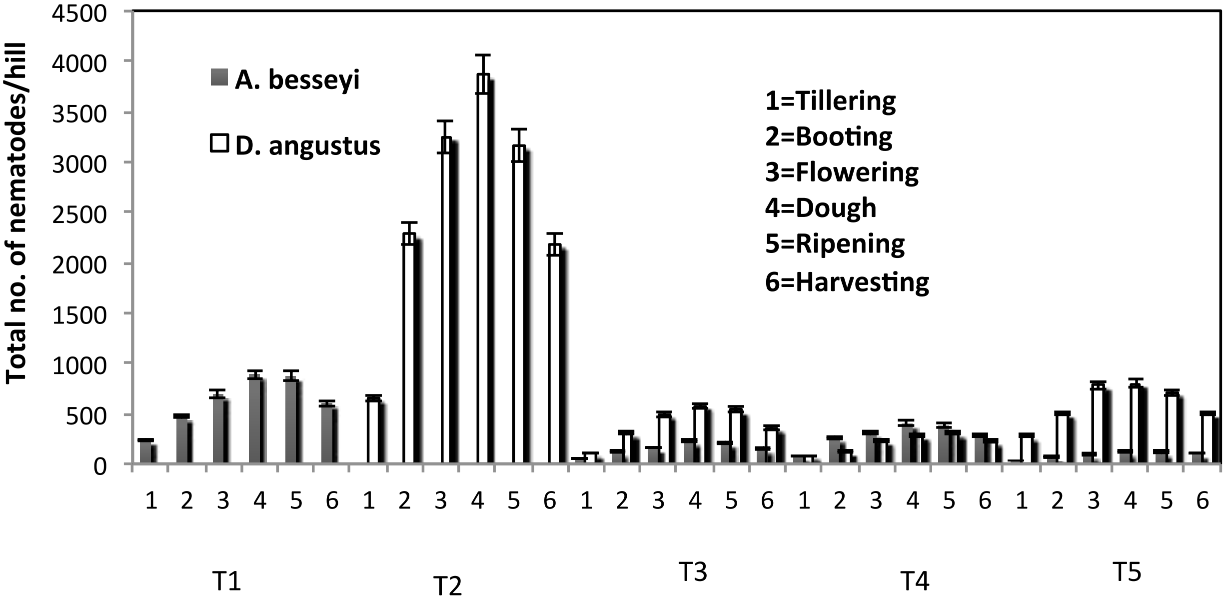

Figure 1B. Population dynamics of *Aphelenchoides besseyi* and *Ditylenchus angustus* at different rice plant growth stages in the field experiment 2003 to 2004. Treatment (T), *A. besseyi* : *D. angustus* inoculation ratios, T1 = 100:0; T2 = 0:100; T3 = 50:50; T4 = 75:25; T5 = 25:75]. Vertical bars indicate standard errors.

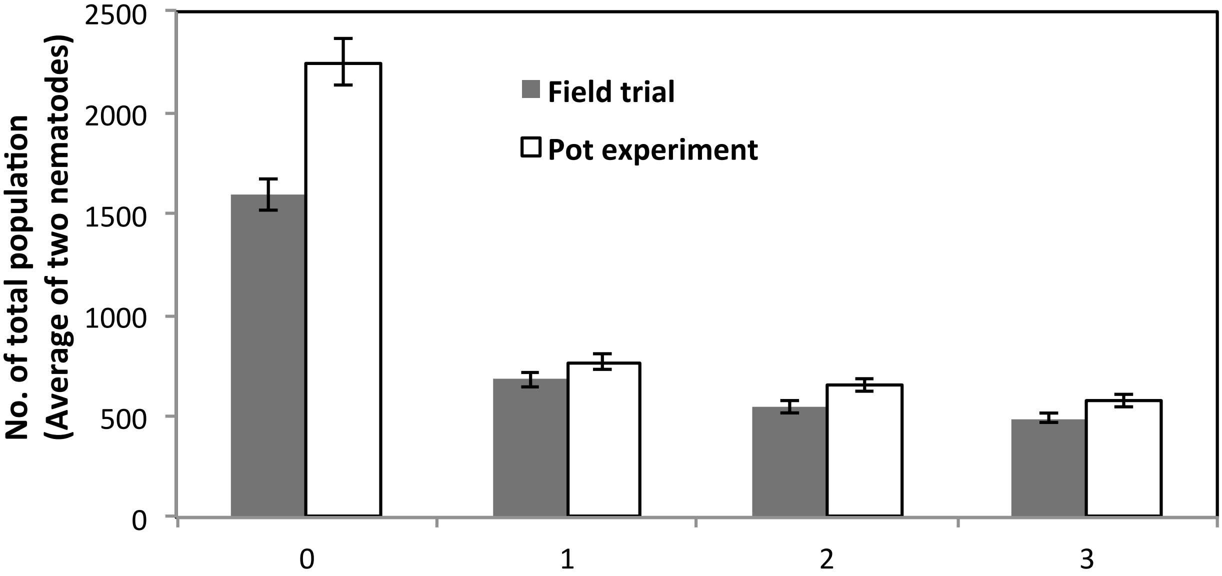

Figure 2. Mean total populations of *Aphelenchoides besseyi* and *Ditylenchus angustus* in pots and irrigated field trials. 0 = average number of nematodes each inoculated singly; 1 = 75:25; 2 = 50:50; 3 = 25:75 (*D. angustus* : *A. besseyi*). Vertical bars indicate standard error.

tions of the two species at all plant growth stages (Table 2). In the field trial, the reproduction rates from the single species inoculations ranged from 2.4 to 8.9 for *A. besseyi* and 6.5 to 38.6 for *D. angustus*, while the rates ranged from 0.8 to 12.0 from the mixed inoculations of the two species at all of the plant growth stages (Table 2). The reproduction rate was greater from single species inoculations of *A. besseyi* or *D. angustus*, but the reproduction rate of each species was significantly reduced when the two nematodes were inoculated together in both experiments (Table 2; Figure 3). In the field experiment, populations of both nematodes and their reproduction rates were inhibited by low temperature and humidity (Figure 3; Table 1).

Disease incidence and yield components from the pot experiment

The greatest mean incidence of white-tip (69.8%) was recorded in 100:0 ratio of *A. besseyi* : *D. angustus*, which was significantly greater than from the other inoculation ratios. The mean incidences of white-tip were 29.6 from the 50:50 of *A. besseyi* : *D. angustus* ratio, and 32.8% from the 75:25 ratio. These incidences were statistically similar. The mean incidences of white-tip in 50:50 (29.6%) and 25:75 species ratios (16.8%) were not statistically different (Table 3A). The greatest mean incidence of ufra (95.8%) was recorded from the 0:100 *A. besseyi* : *D. angustus* ratio, and the least mean incidence (0%) resulted from the 100:0 *A. besseyi*:

Table 2. Mean reproduction rates of two nematodes, *Aphenchoides besseyi* and *Ditylenchus angustus*, inoculated on rice plants at different inoculation ratios, in pot- and field-grown rice plants.

Treatment		Growth stages of rice					
		Tillering	Booting	Flowering	Dough	Ripening	Harvesting
Pot experiment							
T$_1$[a]	A[b]	4.53b[c]	7.80b	8.83b	10.50b	10.08b	8.44b
T$_2$	D[b]	7.90a	33.73a	42.36a	49.67a	49.39a	36.81a
T$_3$	A	0.89d	2.69c	4.91c	7.79bc	6.96c	5.71c
	D	4.06b	6.03b	9.36b	11.51b	10.49b	7.73b
T$_4$	A	1.61d	3.01c	5.17bc	7.90bc	6.66c	4.24c
	D	2.95c	6.24b	9.23b	12.60b	12.40b	8.48b
T$_5$	A	1.79d	2.92c	4.81c	6.81c	7.22bc	5.81c
	D	4.89b	7.03b	11.45b	11.62b	10.85b	6.02bc
Field experiment							
T$_1$	A	2.40c	4.72b	6.90bc	8.90bc	8.70b	6.03b
T$_2$	D	6.50a	22.80a	32.50a	38.60a	31.60a	21.80a
T$_3$	A	0.80d	2.40c	3.20bc	4.60c	4.20c	3.00c
	D	2.00c	6.00b	9.80b	11.60b	10.60b	7.10b
T$_4$	A	1.07d	3.47bc	4.07bc	5.33c	5.07c	3.73
	D	3.20b	4.80b	9.20b	11.20b	12.00b	8.00b
T$_5$	A	1.12d	2.60c	3.60bc	4.80c	4.60c	4.00c
	D	3.60b	6.67b	10.40b	10.67b	9.47b	6.67b

[a] Treatment (T) inoculation ratios of *A. besseyi* : *D. angustus*: T$_1$= 100:0; T$_2$= 0:100; T$_3$= 50:50; T$_4$= 75:25; T$_5$= 25:75 *A. besseyi* : *D. angustus*

[b] A = *A. besseyi*; D = *D. angustus*

[c] Values followed by the same letters in each column are not significantly different ($P \leq 0.05$).

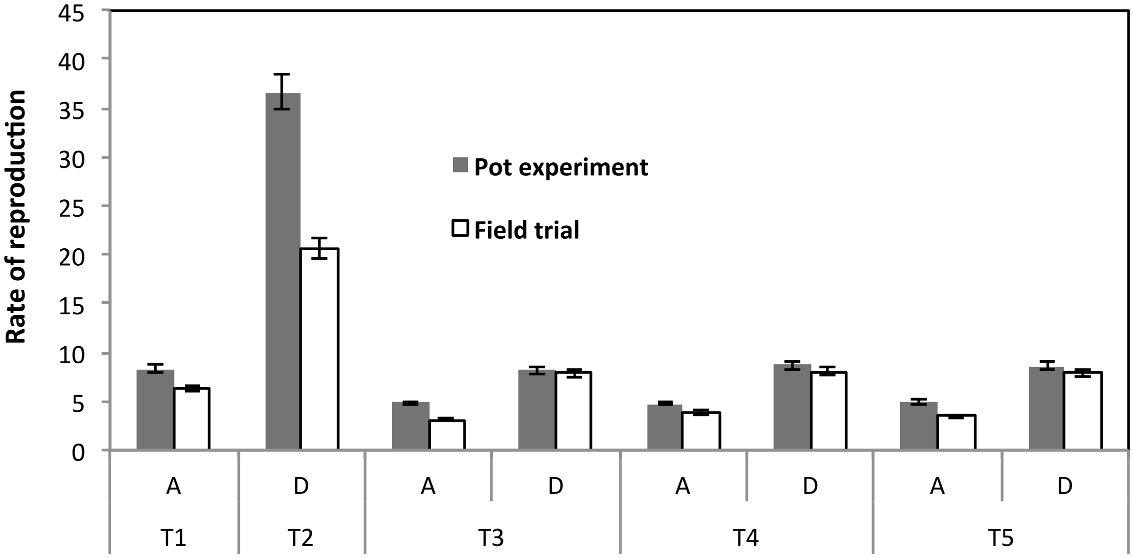

Figure 3. Mean reproduction rates of *Aphelenchoides besseyi* and *Ditylenchus Angustus* in pots and field. Treatment (T), *A. besseyi* : *D. angustus* inoculation ratios, T1 = 100:0; T2 = 0:100; T3 = 50:50; T4 = 75:25; T5 = 25:75; A = *A. besseyi*; D= *D. angustus*. Vertical bars indicate standard errors.

Table 3A. Mean disease incidence, yield components and yields of rice plants inoculated with different ratios of *Aphenchoides besseyi* and *Ditylenchus angustus* in the pot experiment.

Treatment	White-tip incidence (%)	Ufra incidence (%)	Healthy panicle (%)	1000 grain wt (gm)	Yield per hill (g)
T_1[a]	69.8d[b]	0.0a	81.9cd	22.8a	13.7 de
T_2	0.0a	95.8d	4.2a	22.9a	6.3 a
T_3	29.6bc	30.6bc	59.4bc	22.9a	11.4 bc
T_4	32.8c	22.2b	65.4c	23.3a	12.5 cd
T_5	16.8b	40.0c	30.0b	23.7a	9.4 b
T_6	0.0a	0.0a	95.7d	23.8a	15.4 e

[a] Treatment (T) inoculation ratios of *A. besseyi* : *D. angustus*: T_1= 100:0; T_2= 0:100; T_3= 50:50; T_4= 75:25; T_5= 25:75 *A. besseyi* : *D. angustus*.
[b] Values followed by the same letters in each column are not significantly different ($P \leq 0.05$).

D. angustus ratio. These incidences were significantly different, when compared to those from the other inoculation ratios. The mean incidences of ufra were 30.6% from the 50:50 inoculation ratio and 22.2% from the 75:25 *A. besseyi* : *D. angustus* ratio, and these incidences were statistically similar. The greatest mean proportion of healthy panicles (81.9%) resulted from the 100:0 *A. besseyi* : *D. angustus* inoculation ratio, and the least proportion (4.2%) resulted from 0:100 inoculation ratio. The mean proportions of healthy panicle were 59.4% from the 50:50 *A. besseyi* : *D. angustus* inoculation ratio, and 65.4% from the 75:25 inoculation ratio. The greatest yield was obtained from uninoculated control treatment, which was statistically similar to that from the 100:0 *A. besseyi* : *D. angustus* inoculation ratio. The smallest yield was recorded from the

Table 3B. Mean disease incidence, yield components and yield of rice plants inoculated with different ratios of *Aphenchoides besseyi* and *Ditylenchus angustus* in the field trial.

Treatment	White-tip incidence (%)	Ufra incidence (%)	Panicle/m^2	Healthy panicle (%)	1000 grain wt (gm)	Yield (t/h)
T_1[a]	62.3 e[b]	0.0 a	310 c	82.9 dc	22.9 a	4.4 d
T_2	0.0 a	87.3 d	100 a	6.2 a	20.4 a	2.0 a
T_3	25.7 c	35.3 b	150 b	46.5 b	20.4 a	2.9 b
T_4	35.7 d	24.3 b	175 b	53.7 bc	21.2 a	3.8 c
T_5	17.0 b	40.7 c	120 a	35.3 b	20.1 a	2.4 a
T_6	0.0 a	0.0 a	315 c	95.5 d	23.0 a	5.5 e

[a] Treatment (T) inoculation ratios of *A. besseyi* : *D. angustus*: T_1= 100:0; T_2= 0:100; T_3= 50:50; T_4= 75:25; T_5= 25:75 *A. besseyi* : *D. angustus*.

[b] Values followed by the same letters in each column are not significantly different ($P \leq 0.05$).

0:100 *A. besseyi* : *D. angustus* inoculation ratio, which was statistically different to the yields from the other inoculation treatments. The mean 1000 grain weights from the different inoculation ratio treatments were not significantly different (Table 3A).

Disease incidence, yield components and grain yields from the field experiment

For white-tip, the greatest incidence (62%) was recorded from the 100:0 ratio of *A. besseyi* : *D. angustus*, but no white-tip was recorded from the 0:100 ratio *A. besseyi* : *D. angustus* ratio or the non-inoculated control. The greatest ufra incidence (87%) was recorded from the 0:100 *A. besseyi* : *D. angustus* inoculation ratio, which differed significantly from the other ratios. Mean incidence of ufra was 35% from the 50:50 inoculation ratio, and 24% from the 75:25 *A. besseyi* : *D. angustus* ratio. No ufra was observed from the 100:0 ratios *A. besseyi* : *D. angustus* inoculation ratio or the non-inoculated control (Table 3B).

The greatest mean number of panicles m^{-2} was recorded in the non-inoculated control, which was statistically similar to that from the 100:0 inoculation ratio of *A. besseyi* : *D. angustus*. The smallest number of panicles m^{-2} was recorded from the 0:100 inoculation ratio, which was statistically similar to that from the 25:75 ratio. The greatest proportion of healthy panicles (96%) was recorded from the non-inoculated control, and the least (6%) resulted from the 0:100 *A. besseyi* : *D. angustus* inoculation ratio. The mean proportions of healthy panicles ranged from 35 to 54% from the 50:50, 75:25 and 25:75 *A. besseyi* : *D. angustus* ratios, which were statistically similar. Mean 1000 grain weights were not statistically different for all of the inoculation ratio treatments. All the treatments significantly varied for yield (t ha^{-1}) where the greatest yield (5.50 t ha^{-1}) was recorded from the non-inoculated control, and the least (1.96 t ha^{-1}) resulted from the 0:100 *A. besseyi* : *D. angustus* ratio. Yield was also greater (4.35 t ha^{-1}) from the 100:0 ratio (inoculation with *A. besseyi* alone). Mean yields decreased in order of 100:0, 75:25, 50:50, 25:75 and 0:100 *A. besseyi* : *D. angustus* inoculation ratios (Table 3B).

Yield losses due to single and mixed *Aphelenchoides besseyi* and *Ditylenchus angustus* inoculations

Yield loss data are from two seasons as presented in the Figure 4. The average of the two seasons was determined to assess the average annual yield loss. The greatest average yield loss (61.7%) was recorded when the seedlings were inoculated only with *D. angustus* followed by the 25:75, 50:50, 75:25 and 100:0 *A. besseyi* : *D. angustus* inoculation ratios. The smallest average yield loss (16.0%) was recorded from the *A. besseyi* inoculation. Average yield losses across the two seasons ranged from 24.2 to 47.2% due to the mixed inoculation ratios of *A. besseyi* and *D. angustus*.

Discussion

In the present study, initial population densities of both nematodes were not sufficiently great com-

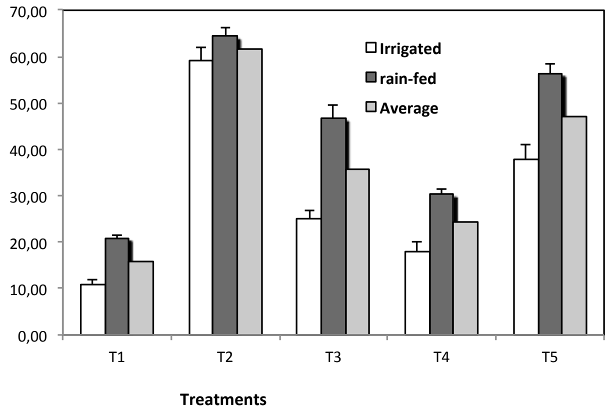

Figure 4. Mean rice grain yields from single and mixed inoculations of *Ditylenchus angustus* and *Aphelenchoides beseyi*. Treatment (T), *A. besseyi* : *D. angustus*,, T1 = 100:0; T2 = 0:100; T3 = 50:50; T4 = 75:25; T5 = 25:75. Vertical bars indicate standard errors.

pared to *Pratylenchus* spp. populations, which, as reported by de la Pena *et al.* (2008), can negatively affect plant growth. The reproduction rates both of *A. besseyi* and *D. angustus* were greater from the single species inoculations than from the mixed inoculations. The nematode populations reached lower numbers in mixed inoculations, suggesting horizontal control. Because the life cycles of both nematodes are probably adapted to the same crop, they use the same resource, and the host plants are likely to be a key factor in competition between the two nematodes. *Ditylenchus angustus* had a greater reproduction rate than *A. beeseyi* in single inoculations. Rice plants have greater "carrying capacity" for *D. angustus*, and this would favour the nematode in competition. Therefore, this study demonstrates that the migratory ectoparasite *D. angustus* was a competitor to *A. besseyi*. Several studies in agricultural systems have shown that *Pratylenchus* spp. inhibit *Heterodera* spp. and *Meloidogyne* spp. (Eisenback, 1993; Lasserre *et al.*, 1994; Umesh *et al.*, 1994), whereas the reverse has been reported depending on host suitability (Eisenback, 1993). As both of the nematode species are to be found in the same rice fields or even infecting individual plants, our results may indicate competition between naturally co-evolved nematode species. These findings are also in agreement with results of Hol *et al*., (2008) and Brinkman *et al*., (2005) for interactions between different root-feeding nematodes on *Ammophila arenaria*.

The number of nematodes substantially influenced incidence of both ufra and white-tip in our study. Competition between the two nematodes, *A. besseyi* and *D. angustus,* may explain the lower reproduction rates and results in reduced disease incidence. Similar results were reported by Eisenbach and Griffin (1987) in two plant-parasitic nematodes, *M. hapla* and *X. americanum*. Competition theory states that two species that co-exist while sharing the same resource should evolve niche partitioning to avoid extinction (Armstrong and McGehee, 1980). However, both species had increased to their peak densities which occurred during the same growth stage of rice. This indicates that either the host was not limiting or had limited negative behavioural attributes sufficient to allow co-existence of the two species. In the case of interactions of root-feeding nematodes, several authors have reported that the population dynamics would be influenced both by the suitability of the host plants, and by the presence and identity of surrounding plant species, other root-feeding nematode species and natural enemies (De Deyn *et al.*, 2004; Schroeder *et al.*, 2005; Piskiewicz *et al.*, 2007).

We have determined the incidence and yield losses of rice due to interaction of two nematode species. In this type of interaction, yield losses are influenced by the pathogenicity of the species of nematodes involved (Mai and Abawi, 1987; Riedel, 1988; Woo and Lorito, 2007). Sometimes, one nematode species may predispose or facilitate invasion of the host plant by other species. An example is the combined occurrence of *Hoplolaimus columbus*, *Scutellonema brachyurum* and *Meloidogyne incognita* in South Carolina which required changes in pest management programmes where interactions affected crop losses (Kraus-Schmidt and Lewis, 1981). In our studies, however, interaction of two plant parasitic nematode species was beneficial for rice plants and final yields, compared to the effects of individual nematode species.

It appears that *Ditylenchus angustus* had a greater reproduction rate compared to *A. besseyi* in single inoculations of either species. In normal conditions, the life cycle from egg to egg takes only 8 days for *D. angustus* but about 15 days are required for *A. besseyi* (Huang *et al.*, 1972; Siddiqi, 2000). In the present study, the reproduction rates both of *A. besseyi* and *D. angustus* were reduced after mixed inoculations.

In both the pot experiment and the field trial, the greatest yield losses were recorded when the rice seedlings were inoculated with *D. angustus* alone.

However, yield loss was reduced after mixed inoculation with *D. angustus* and *A. besseyi*. Latif *et al.* (2011b) reported 90% yield loss in irrigated rice caused by *D. angustus*. In Bangladesh, this nematode is considered as a minor pest of rice (Rahman and Miah, 1989). In China, yield losses due to *A. besseyi* can be as high as 45% when plant infestation levels exceed 50% (Tsay *et al.*, 1998). In the present study, mixed inoculation by *D. angustus* and *A. besseyi* in different ratios caused yield losses ranging from 24 to 47%. There was a direct relationship between *D. angustus* and yield reduction. The higher the *D. angustus* ratio in inoculum, the greater was the yield reduction. Yield loss was mostly caused by *D. angustus*, indicating that this nematode is likely to be more damaging to yield than *A. besseyi.*

This study demonstrated inter-specific competition between *D. angustus* and *A. besseyi*. If we could apply missing inoculum levels for each treatment we might get a clearer picture of interaction between the two plant parasitic nematodes. Nematode reproduction rates may be highly influenced by initial densities, as various levels of inoculum in single inoculations were shown to be important by de la Pena *et al*. (2008). However, the present study is the the first to examine the interactions of white-tip and ufra nematodes, in relation to population dynamics, and effects on rice yield and yield components.

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