J. Asia-Pacific Entomol. 4 (2) : 187~193 (2001)
www.entomology.or.kr

Effectiveness of Brown Planthopper Predators: Population Suppression by Two Species of Spider, *Pardosa pseudoannulata* (Araneae, Lycosidae) and *Araneus inustus* (Araneae, Araneidae)

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Abstract The most abundant natural enemies found in Cambodian rice field are spiders, mostly *Araneus inustus* and *Pardosa pseudoannulata.* These two hunting and wolf spider, respectively, are believed to actively contribute to brown planthopper (BPH) population control. However, how much each species attacks prey in Cambodian field condition is unknown. We conducted field experiments in Cambodia during the wet season at two locations, a farmer's fields at Takeo and at CARDI, using both field cages and natural conditions. Cages were sprayed with insecticide to remove all pre-existing insects in the cages and then washed after 10 days to reduce insecticide residue. Results confirmed BPH inside the cage were killed by the insecticide. A known BPH population was reared inside the cages starting with 3 pairs of adults. Temporary cages were removed after counting second instar BPH and permanent cages were left in place. Spiders were released into the cages for 15 days. In permanent cages either two individual *A. inustus* or *P. pseudoannulata* were allowed to feed on BPH prey. Both spider species have the same killing ability in dense prey populations, but predation is higher for *Pardosa* at low prey density. In uncaged field environments (where more than just BPH prey are available) with a spider/BPH ratio 1:3 to 1:11 BPH mortality was 78-91%. Within 15 days in permanent cages spiders caused 100% BPH mortality at an average predator/prey ratio of 1:5 to 1:14. At a ratio of 1:18 or higher there was some BPH survival in cages.

Key Words brown planthopper, predator, *Pardosa, Araneus, Nilaparvata,* fertilizer

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(Received October 10, 2001 ; Accepted November 5, 2001)

Introduction

Invertebrate predators are very abundant and often conspicuous in many agricultural as well as natural habitats occupied by planthoppers (Yasumatsu and Torii, 1968; Chiu, 1979; Kiritani, 1979). A wide diversity of invertebrate predators (Aranea, Phalagida, Acarina, and nine orders of Insecta: Collembola, Odonata, Orthoptera, Dermaptera, Hemiptera, Neuroptera, Coleoptera, Diptera, and Hymenoptera) feed on planthoppers (Waloff, 1980). The interactions between planthoppers and their natural enemies are believed to be the major factor controlling the pattern of population growth (Wada *et al.,* 1991). Among 17 species of Delphacidae, spiders were considered the major predators for 13 species, and nine species of planthoppers were the most important prey for spiders (Denno, 1994). Waloff (1980) and Denno (1994) considered spiders were the most important predators on planthoppers in both natural and agricultural systems. Both hunting spiders (Lycosidae) and web-builders (Linyphiidae and Tetragnathidae) were the most important predators of planthoppers (Kenmore, 1980; Döbel, 1987; Ooi, 1988). Spiders and a small water bug (*Microvelia douglasi atrolineata* Bergroth) were the predominant polyphagous predators in paddies (Wada *et al.,* 1991). However how many planthoppers are killed by predators, such as spiders, is unknown and so recommendation for their use in pest management decision making is difficult (Wada *et al.,* 1991; Preap *et al.,* 2000).

The effect of natural enemies on brown planthopper (BPH) mortality was highly significant in experiments conducted at Takeo and CARDI Cambodia (Preap, 2001). Among twelve species of natural enemies found in these field experiments two species of spiders were the most common: *Pardosa pseudoannulata* Boesenberg *et* Strand (Lycosidae) and *Araneus inustus* L. Koch (Araneidae). Up to 60% of the total natural enemies sampled were *A. inustus* and 30% were *P. pseudoannulata.*

Both species are polyphagous predators eating BPH and other prey including other species of natural enemies. Even at low abundance these predators can actively contribute to the level of mortality in BPH. To test how much these predators specifically contribute to BPH mortality under field conditions an experiment was conducted.

Materials and Methods

Field-cage experiments were conducted in the wet season from July to December 2000 at two sites: The Cambodia Agricultural Research and Development Institute (CARDI, 11.5°N, 104°E) near Phnom Penh; and in a farmer's field in Takeo province (Takeo, 11°N, 104°E). Eth Chhmoush, a traditional rice variety, was used in the experiments. Two levels of NPK fertilizer input, low (30-10-10 kg/ha of NPK) and high (90-30-30 kg/ha of NPK) were applied in each of two main plots (one hectare per main plot).

Wooden frame cages were erected in the rice field 10 days before maximum tillering. The cages were covered with 1mm nylon mesh with a zippered door. Each cage (0.46m×0.4m×1.3m) covered four plants. Rice plants inside the cages and the interior of the cages were sprayed with methyl parathion at 25 g AI/hl, and the cages were sealed for 10 days to remove pre-existing predators and other insects. The cages and plants inside the cages were washed with water to remove residual insecticide. Three pairs of adult BPH were carefully transferred onto plants inside cages. The number of BPH was checked 24 hours after infestation for any dead adults. After three days of infestation all surviving BPH adults were counted and removed from the cages. On the day after counting the second instar nymphs (resulting from the reproduction of caged adults), temporary cages were removed, permanent cages were left in place and infested with either zero or two adults of *Araneus inustus* or *Pardosa pseudoannulata* and sealed until nymphs had developed into adults. Number of BPH and spider survival was checked twice visually; once three days after predator transfer and then at 15 days after transferring spiders (at BPH adult stage). BPH nymphs in temporary cages were exposed to ambient environmental field conditions and the number of BPH surviving checked with the same procedure as in permanent cages. Natural enemies and other insects were sampled by D-vac machine at maximum tillering stage from the experimental plots during the course of the season and to determine the relationship between spider density and BPH mortality in temporary cages. Collected natural enemies were returned to the laboratory for identification.

Data were manipulated and stored in Microsoft Excel. Analysis of Variance (ANOVA) was used in the statistical analysis with IRRISTAT version 3.1. The mortality of nymphs under spider suppression within 3 days was calculated from the initial number of second instar nymphs in a cage minus the number of nymph found after 3 days. BPH mortality to the adult stage was the difference between the number of BPH nymphs found at three days and the number of BPH counted at 15 days after spiders were introduced into the cages. The predator/prey ratio (R) was the number of spiders (N_{spider}) counted (or introduced) at the same date of prey (BPH) counting divided by the numbers of BPH at a given date (N_{BPH}), ($R = N_{BPH} / N_{spider}$). Duncans Multiple Range Test (DMRT) was used to compare means among treatments.

The percentage of BPH mortality caused by spiders was calculated from the following formula:

Msps (%) = $(Ni\text{-}Nf)/Ni \times (1-(N_i^w - N_f^w)/ N_i^w) \times 100$

Msps = BPH mortality due to spiders,
N_i = Initial BPH population with spiders,
N_f = BPH population after given period with spiders,
N_i^w = Initial BPH population without spiders and
N_f^w= BPH population after given period without spiders.

Relationships between nymph population density (X) and nymph mortality (Y) was plotted and analysed using linear regression ($Y = \alpha + \beta X$) for each cage type.

Results

Introduced male and female adult brachypterous BPH were found on the stem of the rice plant near the conjunction of leaf sheath and leaf blade. After 24 hours of adults being transferred, the number of adult females was the same as the initial number. Ten days after being sprayed with insecticide and being rinsed with plenty of water little insecticide residue remained on the plants and interior the cages.

The abundance of arthropods gradually increased with crop growth stage at Takeo (Fig. 1). All arthropods reached a peak at reproductive stages of plant growth. The population of brown planthoppers in high-fertilised plots was the highest as was the natural enemy count (Fig. 1). However, this population density (120 insects/sqm or 4.8 insects per hill) did not cause any damage to the crop. At the end of the crop season all arthropods populations, including natural enemies, decreased to low levels.

The population of brown planthoppers in high

Table 1. Number of natural enemies per sample (4 hills) causght by D-vac machine at maximum tillering in plots treated with low and high fertilizer at CARDI and Takeo during wet season of 2000

Order	Family	Species[a]	CARDI Low fertilizer	CARDI High fertilizer	TAKEO Low fertilizer	TAKEO High fertilizer
Araneae	Araneidae	*Araneus inustus* (L.Koch) (n&a "pr")	9.4	4.7	14.1	7.8
Araneae	Lycosidae	*Pardosa pseudoannulata* (n&a "pr")	10.9	15.6	6.3	14.1
Coleoptera	Staphylinidae	*Paederus fuscipes* (Curtis) (n&a"pr")	6.3	6.3	6.3	7.8
Coleoptera	Carabidae	*Ophionea ishii* (Habu) (n&a "pr")	6.3	7.8	9.4	9.4
Hemiptera	Miridae	*Cyrtorhynus lividipennis* Reuter (e"pr")	4.7	1.6	4.7	3.1
Odonata	Coenagrionidae	*Agriocnemis fermina fermina* (Brauer) (n&a"pr")	3.1	1.6	3.1	1.6
Hymenoptera	Mymaridae	*Gonatocerus* sp. (e"pa")	1.6	1.6	1.6	1.6
Hymenoptera	Mymaridae	*Anagrus optabilis(Perkins)*(e"pa")	3.1	0	3.1	1.6
Hymenoptera	Mymaridae	*Anagrus flaveolus* (Waterhouse) (e"pa")	1.6	3.1	1.6	4.7
Hymenoptera	Trichogrammatidae	*Paracentrobia andoi* Ishii (e"pa")	1.6	3.1	1.6	3.1
Hymenoptera	Trichogrammatidae	*Oligosita yasumatsui* Viggiani et Subba Rao (e"pa")	4.7	4.7	4.7	4.7
Strepsiptera	Elenchidae	*Elenchus yasumatshui* Kifune *et* Hirashima (e"pa")	1.6	0	3.1	1.6
TOTAL			54.7	50	59.4	60.9

[a]e"pa": egg parasitoid, e"pr": egg predator, and n&a"pr" nymph and adult predator.

fertilizer plots was higher than in low fertilizer plots, while natural enemies for some species increased and other insect pests were not different between low and high fertilizer plots (Fig. 1 and Table 1). Fertilizer increased the population of brown planthopper but not natural enemies (Fig. 1). Population of natural enemies increased at somewhere near to when the population of BPH was highest.

Twelve species of natural enemy of BPH were found in the field experiment (six predators and six parasitoids) belonging to nine families in six orders. Predators were more abundant and more frequent than parasitoids (Table 1). Among the six species of predator, the population of two species of spiders (*A. inustus* and *P. pseudoannulata*) was the highest.

There were two opposite population responses to the fertilizer by two species of spider. At both locations, *A. inustus* response to the fertilizer tended to be negative while *P. pseudoannulata* was positive. The population of *A. inustus* decreased in high fertilizer plots while *P. pseudoannulata* had higher populations in plots with high fertilizer treatment (Table 1). Overall at maximum tillering there was little difference in the abundance of natural enemies between high and low fertilizer treated plots. Second instar BPH nymphs were generally found on the stem of the rice plant near the water level. On some plants (with high density) they were found on the upper parts of the plants as well.

At both locations the initial number of BPH nymphs was 55 to 58 per cage in low fertilizer cages and almost double in cages treated with a high fertilizer rate (109 to 112 per cage) (Table 2).

After 3 days of exposure to spider predators, *N. lugens* nymph mortality in low fertilizer cages (temporary cages) was 61 % at Takeo, and 72 % at CARDI. Mortality was lower in high fertilizer cages, 49% at Takeo and 65% at CARDI. Without spiders present (0 spiders in sealed cages), the mortality of brown planthoppers was 25% in low fertilizer plots at CARDI and 20% at Takeo. Again mortality was lower in high fertilizer permanent cages; 10% at Takeo and 14% at CARDI (Table 2).

Increased fertilizer decreased BPH mortality; sealed cages in low fertilizer plots had more BPH mortality than sealed cage in high fertilizer plots. After 15 days, in sealed cages without predators, the mortality of BPH was 50-53% in low and 26-27% in high fertilizer plots (Table 2). Additional mortality caused by spider was found in cages with introduced pairs of *Pardosa* or *Araneus* spiders, and field ambient condition (variable number of predators) at both locations (Figs. 2a and 2b).

In high fertilizer treatments where BPH nymph mortality caused by negative effect of plant-insect interaction was lower, spiders had more prey available as food. Both spiders killed more BPH at high fertilizer plot-caged (Figs. 2a and 2b).

In low fertilizer cages, with a predator/prey ratio 1:21 and 1:17 *A. inustus* caused 73% and 75% of

Table 2. Percentage of BPH nymph mortality after 3 and 15 days predation and adult BPH killed by predators in cages experiment at CARDI (C) and Takeo (T) during wet season of 2000

	3 day after exposure to spiders										15 days after exposure to spiders							
Treat -ments[a]	No. nymphs per cage		No. nymphs survival per cage		% nymph mortality		No. spiders per cage		Ratio[b]		No. adults survival per cage		% BPH mortality		No. spiders per cage		Ratio[b]	
	T	C	T	C	T	C	T(A/P)	C(A/P)	T	C	T	C	T	C	T(A/P)	C(A/P)	T	C
L-T	58	55	22	16	61	72	2/1	1.5/1.5	1:11	1:12	5	10	91	81	3/2	2/2	1:4	1:3
H-T	112	109	57	39	49	64	1/2	0.75/2.5	1:16	1:22	14	24	88	78	3/2	2/2	1:11	1:7
L-P-0	56	44	44	33	20	25	0	0	0	0	28	21	50	53	0	0	0	0
H-P-0	109	91	98	78	10	14	0	0	0	0	81	66	26	27	0	0	0	0
L-P-2A	59	47	16	12	73	75	2	2	1:21	1:17	0	0	100	100	2	2	1:8	1:6
L-P-2P	55	44	15	11	73	75	2	2	1:20	1:20	0	0	100	100	2	2	1:8	1:5
H-P-2A	113	95	36	29	69	70	2	2	1:39	1:39	5	0	96	100	2	2	1:18	1:14
H-P-2P	113	93	36	28	68	70	2	2	1:38	1:38	3	0	97	100	2	2	1:18	1:14

[a]L-T: Low fertilizer-Temporary cage, H-T: High fertilizer-Temporary cage, L-P-0: Loafer fertilizer-Permanent cage-0 predator, H-P-0: High fertilizer-Permanent cage-0 predator, L-P-2A: Low fertilizer-Permanent cage-2 *Araneus*, L-P-2P: Low fertilizer-Permanent cage-2 *Pardosa* H-P-2A: High fertilizer-Permanent cage-2 *Araneus*, and H-P-2P: High fertilizer-Permanent cage-2 *Pardosa*
[b]Spider: BPH.

BPH mortality within three days at Takeo and CARDI respectively. *P.pseudoannulata* had a similar killing ability to *A. inustus*. Both spider species in three days of predation killed 73-75% of their prey; *P. pseudoannulata* and *A. inustus* with a ratio 1:20-1:21 in Takeo and 1:16-117 at CARDI (Table 2). At dense population of their prey, the ability to kill was not significantly different between *A. inustus* and *P. pseudoannulata*.

Furthermore, the mortality of BPH nymphs was correlated to their population density (Figs. 3a and 3b); there was a high correlation between BPH nymph mortality and nymph density. The value of the correlation coefficient (r^2) was 0.73 to 0.99 for most treatments except treatment 1; low fertilizer with zero spiders; where the r^2 value was 0.47 (Table 3).

Table 3. Coefficient regression (r^2) of nymphs and nymphs mortality of *N. lugens*

Treatments	r^{2a}	
	CARDI	Takeo
Low fertilizer-Temporary cage	0.98**	0.94**
High fertilizer-Temporary cage	0.78**	0.87**
Low fertilizer-Permanent cage-0 predator	0.44*	0.74**
High fertilizer-Permanent cage-0 predator	0.99**	0.97**
Low fertilizer-Permanent cage-2 *Araneus*	0.97**	0.92**
Low fertilizer-Permanent cage-2 *Pardosa*	0.73**	0.88**
High fertilizer-Permanent cage-2 *Araneus*	0.99**	0.82**
High fertilizer-Permanent cage-2 *Pardosa*	0.99**	0.88**

[a]*Intermediate correlation, **highly correlation.

More *N. lugens* survived and molted into adults on plants with high fertilizer application. In sealed cages (without spider); in low fertilizer plots the percent of the population molted to the BPH adult stage was 47 to 50% while in high fertilizer plot it was 73-74% (Table 2).

For temporary cages, the mortality of brown plant hoppers was not significantly different between low and high fertilizer application cages (Table 2). In low fertilizer plots with a spider/BPH ratio of 1:3 the mortality of BPH was 81% at CARDI and 91% at Takeo with a slightly lower ratio (1:4). In high fertilizer plots the ratio was lower (1:7 at CARDI and 1:11 at Takeo) and mortality was 81% and 91% in low and 78% and 88% in high fertilizer cages at CARDI and Takeo, respectively (Table 2).

A. inustus and *P. pseudoannulata* in sealed cages killed all their prey within 15 days. With a daily predator/prey ratio from 1:5 to 1:14 at both sites (CARDI and Takeo) for each spider species, the mortality of BPH was 100 percent, while at Takeo with a daily predator/prey ratio of 1:18 the mortality of BPH was a little lower (96 % by *A. inustus* and 97% by *P. pseudoannulata*) (Table 2).

Discussion

Predators contribute more to BPH prey mortality than the host plant-insect interaction effect. For plants with high fertilizer, the plant-insect interaction factor contributed less to BPH mortality because of high mortality due to natural enemies. Consequently, BPH

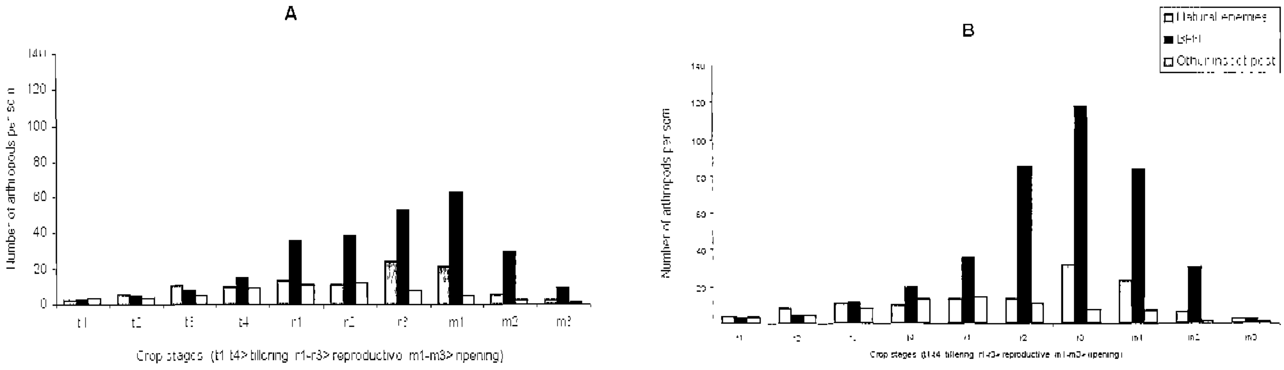

Fig. 1. Population growth of BPH, natural enemies and other insect pest throughout the crop season from tillering to maturing, Takeo, Cambodia wet season. **A**: low fertilizer plots and **B**: high fertilizer plots.

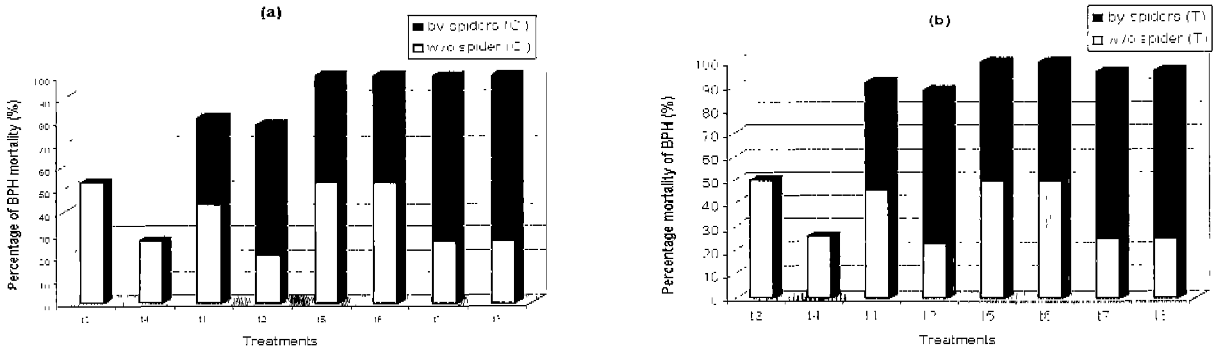

Fig. 2. Additional BPH mortality by spiders at (a) CARDI (C) and (b) Takeo (T).
t1: (L-T)> low fertilizer, temporary cage. t2. (H-T)> high fertilizer, temporary cages,
t3: (L-P-0)> low fertilizer, permanent cage, 0 predator. t4: (H-P-0)> high fertilizer, permanent cage, 0 predator,
t5: (L-P-2A)> low fertilizer, permanent cage, 2 *Araneus*, t6: (L-P-2P)> low fertilizer, permanent cage, 2 *Pardosa*.
t7: (H-P-2A)> high fertilizer, permanent cage, 2 *Araneus*. and t8: (H-P-2P)> high fertilizer, permanent cage, 2 *Pardosa*.

populations with spiders present were controlled. Morris (1992), and Hacker and Bertness (1995) found similar results; natural enemies appear to contribute more than host-plant factors to the suppression and population dynamics of many other plant-feeding insects. With BPH only present in permanent cages both spider species killed more BPH than in temporary cages with a mixture of prey.

In permanent cages with a ratio of spider to BPH of 1:4 to 1:14, spiders can kill all BPH present within 15 days. The effective control by spiders becomes smaller when the prey density increased. At a predator/prey ratio of 1:18, 96 and 97 percent of BPH present were killed by *A. inustus* and by *P. pseudoannulata,* respectively. In a pure species environment *A. inustus* or *P. pseudoannulata* can control BPH populations when the predator/prey ratio was lower than 1:18. But at a ratio 1:18 or higher the mortality of brown planthopper caused by these two species was lower than 100%. Some numbers of BPH were able to escape from spider attack. In cage experiments at 1:10 predator/prey ratio *P. pseudoannulata* killed all BPH nymphs in a day and at a ratio of1:50 this spider species produced adult BPH daily mortality of up to 46% (Dyck and Orlido, 1977).

Both hunting (*A. inustus*) and wolf spiders (*P. pseudoannulata*) are polyphagous predators. Beside BPH, they feed on more than one species including other species of predators (Heong *et al.,* 1991). Their ability to catch their prey depends on the environment. In mixed prey environments they chose the easiest prey to catch and eat (Murdoch 1975; Hassell 1978) and target prey mortality may be smaller. The results in ambient field conditions (temporary cages) where there was a mixture of prey species the mortality of BPH was lower than in a pure species environment. With a predator/prey ratio 1:3 to 1:11 these two spider species caused 78 to 91 percent of BPH mortality when BPH population are in normal situation (i.e. non-outbreaks). With a density of 1 BPH per hill at ripening stage plants did not suffer any damage. These results are very similar to Stapley (1976) and Manti (1989) who suggested that in rice fields, the populations of *N. lugens* can be effectively controlled at spiders/prey ratio of 1:4 to 1:8 for *P. pseudoannulata.* Chiu (1984) suggested that with a ratio 1:8 *Lycosa* spiders can control BPH populations at non-outbreak level. Kiritani *et al.* (1972) suggested

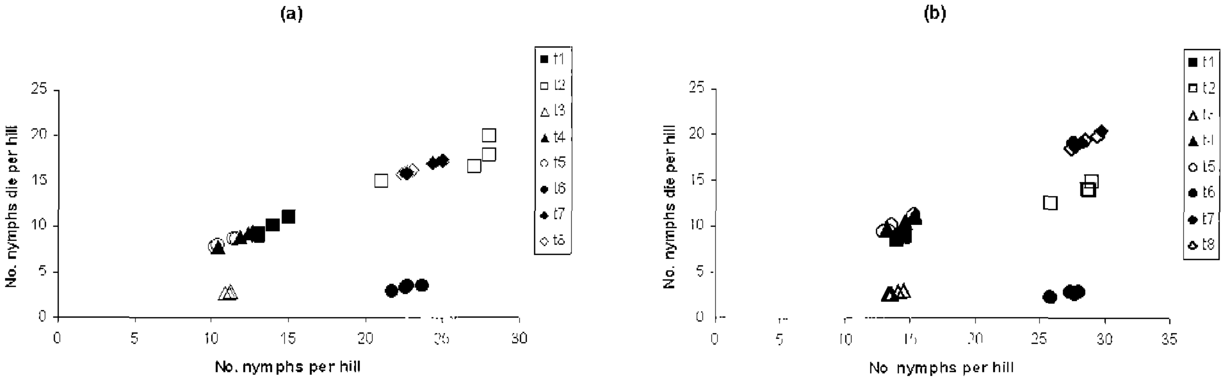

Fig. 3. Relationship between number of nymphs and nymphs mortality at (a) CARDI and (b) Takeo.
t1. (L-T)> low fertilizer, temporary cage. t2: (H-T)> high fertilizer, temporary cages,
t3: (L-P-0)> low fertilizer, permanent cage, 0 predator, t4: (H-P-0)> high fertilizer, permanent cage, 0 predator,
t5: (L-P-2A)> low fertilizer, permanent cage, 2 *Araneus*, t6: (L-P-2P)> low fertilizer, permanent cage, 2 *Pardosa*,
t7: (H-P-2A)> high fertilizer, permanent cage, and 2 *Araneus*, t8: (H-P-2P)> high fertilizer, permanent cage, 2 *Pardosa*

that *N. lugens* and *Nephotettix virescens* Distant formed 25% to 53% of the diet of *P. pseudoannulata.* Reissig *et al.* (1982) and Cohen *et al.* (1997) found the predator/*N. lugens* ratios were generally higher on the resistant variety and lowest on the susceptible variety (where the population of *N. lugens* is low on resistant and high on susceptible varieties).

Cambodian rain-fed lowland rice agro-ecosystems are generally very rich in natural enemies, especially spiders. Even in crops planted with a susceptible variety (Eth Chhmoush) and treated with a high NPK fertilizer rate predators can keep PBH population under non-outbreak levels unless there is a massive migration of BPH or other factors alter natural enemy populations, for example indiscriminate use of insecticide. However crops treated with high fertilizer face a higher risk to BPH attack compared to crops grown at low fertilizer.

P. pseudoannulata and *A. inustus* spiders are very common in Cambodian rice fields (Preap 20001, 30% and 60% of *Pardosa* and *Araneus,* respectively, of total natural enemies) and are key factors in keeping rice crops free from BPH attacks. As the population of predator increases along with prey populations, a spider/BPH ratio at not much over 1:11 is sufficient to save a crop from pest attack. Farmers can harvest a good yield by keeping rice crops safe from BPH attacks. They need to take care of spiders and others natural enemy in their fields. Field monitoring is recommend to follow the crop situation and determine the predator/prey ratio for decision making in pest control. If a spider/BPH ratio is higher than 1:11 and continues to increase up to or higher than 1:20, then the crop is in danger of damage and a chemical control method would be chosen, especially for a susceptible variety, and a selective chemical compound would highly recommend.

Acknowledgement We thank Cambodia Agriculture Research and Development Institutes staffs, Ouk Makara, Sam Simeth, Sareth Chea, Sophea Pheng, Soneath Chin, Muny Chan Tary, Bunnarith Khiev and IPM teams provided a research facilities and data analysis. This research was funded by AusAid through the IRRI.

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