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Biological Control of the Coconut Hispine Beetle, *Brontispa longissima* Gestro (Coleoptera: Chrysomelidae) by the Parasitoid, *Asecodes hispinarum* Bouček (Hymenoptera: Eulophidae) on a Golf Course

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ABSTRACT

The coconut hispine beetle, *Brontispa longissima* Gestro is an invasive pest in Southeast Asia. Since chemical control of the beetle is not recommended due to its high cost and risk to the environment, biological control is a desirable tool for effective control of this pest. This experiment was conducted at the Panya Indra golf course, Bangkok in 2007 using the parasitoid, *Asecodes hispinarum* Bouček, an effective larval parasitoid. The number of parasitoids released was based on the density of the damaged coconut trees. Continual releases were made in and around the area. Mummified larvae were counted once a month by selecting 20 infested spear leaves. The parasitoids were observed five months after release. The mummified larvae were collected in June, August and November and the percentage parasitized was found to be 13.48%, 8.58% and 14.08%, respectively. Severe, leaf damage levels decreased in December. The results showed that the new coconut palm leaves were fresh with less damage. A population study of *B. longissima* was also undertaken by randomly sampling 20 spear leaves. High and low population levels were observed from March to June and from July to December, respectively.

Key words: *Asecodes hispinarum* Bouček, *Brontispa longissima* Gestro, invasive specie, biological control

INTRODUCTION

The coconut hispine beetle, *Brontispa longissima* Gestro (Coleoptera: Chrysomelidae), is an exotic pest of coconut and ornamental palms in Thailand. Both the larvae and adults of the beetle inhabit the developing, unopened leaves of the coconut palm (Liebregts *et al*., 2006). This invasive pest is new to continental Southeast Asia where, in the absence of natural enemies, it has rapidly spread and caused massive damage. In Thailand, it was first found around 2002 in Narathiwat Province, on the border with Malaysia. Heavy infestation was first reported in February 2004 in the southern provinces, including Surat Thani (Samui and Pa-ngan Islands) and Prachuap Khiri Khan (Sindhusake and Winotai, 2004). The adults and larvae feed on both surfaces of the

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closely appressed leaflets, making long incisions in the tissues parallel to one another and to the veins of the leaflets. When the insects are numerous, these incisions are so close to one another that the whole of the leaflet dies, with all leaflets similarly injured and photosynthesis reduced to almost zero (Lever, 1969). The beetle can cause significant production losses and high infestation levels may result in the death of the palm. Palm productivity and appearance are severely affected by sub-lethal attacks (Liebregts *et al*., 2006). In Thailand, economic losses caused by the coconut hispine beetle are significant. Thailand's coconut industry consists almost exclusively of smallholders with just over 50,000 farmers involved. Most of them have an average of 2.5 ha of palms. Yields are low at around 6.6 t/ha and incomes are also low as the price is only about 8 Thai baht (THB)/kg. The total coconut planting area is estimated to be 328,000 ha producing approximately 1146 million nuts or 344,000 t in copra equivalent, representing a total value of 902.16 million THB domestically and an export value of 512,330 million THB (THB 36= US$ 1). Coconut is not only an important local food crop, but is perhaps even more important for the tourism industry. It is also an added treat to the tourist industry on Samui and Pha-ngan Islands (Liebregts and Champman, 2004). On most golf courses, coconut is an important ornamental landscape palm. Damage caused by the insect severely affects the beauty and aesthetic value of coconut palms. Use of chemical pesticides to control coconut hispine beetle outbreaks and reduce the rate of spread is often costly, ineffective and unsustainable. More importantly, the use of pesticides also raises serious concerns about the health risks to the farmers, their families and consumers (Youngfan, 2007). Biological control has recently been recognized as a promising and effective tool in the management of the most important pest on the coconut palm (Sathiamma *et al*., 2001). *Asecodes hispinarum* Bouček (Hymenoptera: Eulophidae) was one of the most successful species in *B. longissima* control (FAO, 2004). With regard to the release program, *A. hispinarum* was certainly considered to be a very effective enemy of *B. longissima* (Voegele, 1989). *A. hispinarum* was collected in Samoa in 2003 to be introduced, reared and released in Vietnam, the Maldives and Nauru to combat the beetle. The larval parasitoid was established in all three countries, with promising prospects for achieving control of the beetle (Rethinam and Singh, 2005). Therefore, biological control using *A. hispinarum* is considered the best approach to solving this pest problem. This study aimed to achieve integrated pest management (IPM) using the larval parasitoid for biological control of the coconut hispine beetle.

MATERIALS AND METHODS

Preparation of hosts and larval parasitoids for releasing in golf course

Hosts and larval parasitoids were reared in a temperature-controlled room (25±2°C and 70±10% RH with natural lighting) at the laboratory of the Plant Protection Research and Development Office in Bangkok from July 2006 to December 2007. The original host cultures (*Brontispa longissima*) were collected from Prachuap Khiri Khan Province and were reared in plastic boxes (14×10×6 cm^3) covered with screened and ventilated lids. One hundred pairs of male and female adults were put into each rearing box with small pieces of coconut leaves 12 cm in length. Coconut leaves were sprayed with 10% Clorox® (0.1% sodium hypochlorite) and wiped with a piece of clean cloth before being used. New adults were added to replace the dead ones as necessary. Careful inspection and collection of about 700 eggs occurred every two days by scraping eggs from old leaves, with the eggs then transferred to a new rearing box. Five hundred newly emerged larvae were transferred to a new box for further rearing with fresh leaflets. The optimal density was 500 larvae per box, which were observed every two days and old leaves replaced with new, fresh ones. Twenty cuttings of coconut leaves per rearing box were used and individual developmental stages of *B. longissima* were kept separately, similar to the process reported by Liebregts *et al.* (2006).

The rearing method for the larval parasitoid, *Asecodes hispinarum* was similar to that of Liebregts *et al.* (2006), but in the current study, a fine nylon cloth was included on the rearing box. Sixty adult *A. hispinarum* were transferred to each rearing box with 20 host larvae and left for 24 hrs. A piece of tissue paper (3 cm length × 1 cm width) soaked with 50% (w/v) honey solution was pasted onto the inner side of box to provide food for the adult parasitoids. The parasitized hosts became less active and turned a brown color after 6-7 days. Each one was then isolated individually into a small vial (1.5 cm in diameter, 4.5 cm long). The adult parasitoids emerged after 18-20 days and were used in subsequent experiments.

Control of coconut hispine beetle

The experiment was conducted at the Panya Indra golf course in Bangkok during January to December 2007. The area had a coconut population of approximately 1,800 palms, with 20 palms per rai (1 rai = 0.16 ha). Grading of overall damage caused by *B. longissima* on the 1^{st}-10^{th} leaf was performed monthly for every palm in the area. Damage levels on coconut leaves were assessed according to five grading categories: Grade 0 = no damage; Grade A = slight damage, 1-25%; Grade B = medium damage, 26-50%; Grade C = high damage, 51-75%; Grade D = severe damage, 76-100% (Sindhusake *et al.*, 2006). Damage levels on the newly opened coconut leaves were assessed using these same grading categories. All possible breeding sites of *B. longissima* around the area were treated with the biological control agent of the larval parasitoid, *A. hispinarum*. Continuous releases were made in and around the area from January 2007 until the end of the same year. The number of parasitoids released (mummified host larvae) was based on the density of damage on the coconut palms. A release rate of five mummified larvae per rai per month was chosen (Winotai *et al.*, 2007). Releasing occurred by putting five mummified larvae of *B. longissima* in a plastic tube (1.5 cm in diam., 4.5 cm long) into which five holes were made for the parasitoids to exit. Establishment of the larval parasitoids was checked by selecting 20 infested spear leaves. The eggs, larvae, pupae and adult beetles from each sample were kept individually in a rearing box for two weeks and evaluated for the presence of parasitoids. The percentage of parasitization by *A. hispinarum* in the field was checked by collecting and rearing *B. longissima* until parasitoid emergence. The emerged parasitoids were classified and the death of larvae or pupae and emergence of the parasitoids were recorded. The population of *B. longissima* was also recorded on 20 randomly sampled spear leaves.

RESULTS AND DISCUSSION

Preparation of hosts and larval parasitoids for releasing in golf course

From the host-rearing experiment, it was found that the average hatchability of *B. longissima* eggs was 67.37%, while the average larval survival rate was 78.31% resulting in an average larval production of 5574 larvae per month (Table 1). When the host larvae reached the fourth instar, they were exposed to larval parasitoids. Hosts were allowed to be parasitized every two days. The average parasitization rate by *A. hispinarum* was 84.30% with a survival rate to adult parasitoids of 77.73%. Thus, an average of 1180 mummified larvae per month was produced. In this study, a rearing temperature of 25±2°C was chosen according to Zhong *et al.* (2005), who reported that 24°C to 28°C was suitable for the development and reproduction of *B. longissima*.

Table 1 Production of fourth instar larvae of *Brontispa longissima* Gestro for development of the larval parasitoid, *Asecodes hispinarum* Bouček under laboratory conditions (25±2°C, 70±10% RH, natural lighting) from January to December 2007.

Month	*B. longissima*			*A. hispinarum*	
	% Hatchability of eggs	% Survival larvae	Total no. of fourth instar larvae produced	% Parasitization	% Survival adult parasitoids
January	78.52	88.50	5,263	87.86	83.90
February	69.73	84.40	5,019	86.07	81.00
March	56.84	72.50	4,771	81.00	76.10
April	43.54	71.60	4,805	84.57	60.47
May	46.24	63.70	4,754	78.93	66.52
June	63.68	74.80	5,784	80.36	77.87
July	73.91	71.40	5,932	81.29	81.11
August	72.58	74.30	5,678	89.29	78.72
September	74.52	85.70	6,312	84.57	84.80
October	73.12	79.80	5,425	89.71	75.88
November	79.45	82.40	6,422	81.43	80.09
December	76.32	90.60	6,728	86.50	86.29
Average	67.37	78.31	5574.42	84.30	77.73
SD	12.24	8.17	681.84	3.66	7.51

SD = standard deviation

Sanitation and hygiene were also important factors for the successful mass production and thus decomposing leaf materials were removed every two days to prevent the buildup of moisture and pathogens. The amount of leaves was sufficient to provide an adequate food supply for the host, and maintain moisture. However, excessive leaves may prevent air circulation resulting in increased moisture levels and subsequently encourage the growth of lethal pathogens (Liebregts *et al*., 2006).

Control of coconut hispine beetle

Release of parasitoids in the field

The release of parasitoids was based on the density of coconut damage. The average damage on the Panya Indra golf course at the beginning of this study was 48.00% with a total damage area of 43 rai. Mummified larvae containing parasitoids were released at an average of 246 per month, totaling 2950 over the period January to December, 2007. *A. hispinarum* were first retrieved from the field in June (5 months after release) when the rate of field parasitization was 13.48% (Table 2). Thereafter, mummified hosts with *A. hispinarum* were found in August and November with parasitization rates of 8.58 and 14.08%, respectively. The result was similar to that of Rethinam and Singh (2005), who reported a decrease in coconut hispine beetle levels 5-6 months after the initial release in Haikou. Although the parasitization rates reported in this study were quite low when compared to that (37%) reported by Voegele (1989), field observation indicated that the palms showed clear signs of recovery with new green leaves after parasitoid release. The calculated parasitization rate may have been much lower than the actual field rate due to the relatively small sample size (20 spears). The fluctuation in sample composition would affect the observed levels of parasitization. *A. hispinarum* is

Table 2 Parasitization of a population of *Brontispa longissima* Gestro by 20 selected infested spear leaves of coconut palms at the Panya Indra golf course, Bangkok from January to December 2007.

Months	No. of *B. longissima*/spear		The percent of
	Larvae	Mummified larvae	parasitization[1]
January	12.55	0	0
February	13.65	0	0
March	11.20	0	0
April	28.90	0	0
May	31.20	0	0
June	22.25	3.00	13.48
July	11.30	0	0
August	23.30	2.00	8.58
September	10.15	0	0
October	6.30	0	0
November	14.20	2.00	14.08
December	12.15	0	0

[1]Parasitization (%) = $\frac{\text{No. of mummified larvae}}{\text{No. of larvae}} \times 100$

considered a very valuable control agent for *B. longissima*. The rate of return calculated for a 10-year period exceeded 40%, thus highlighting a very successful biological control project (Voegele, 1989). In Vietnam, *A. hispinarum* was imported from Samoa in June 2003. Parasitoid releases were made with 5-6 mummies (approx. 250 parasitoids) per 100 sq km. Within several months of release, *A. hispinarum* had caused a significant reduction in the beetle density and damage to coconut palms. Palms showed clear signs of recovery and returned to pre-infestation production levels. The success of this release program was evident by the presence of mummified larvae at the release site (Viet, 2004).

Observation of damage of coconut leaves on 1st-10th leaves

The rapid survey by grading the damage of *B. longissima* from every palm in the area showed that the intensity of damage was quite high from February to May and gradually decreased from June to December (Table 3). The highest and lowest overall damage rate (A+B+C+D) was 64.85 and 33.76% in May and December, respectively, while the average damage for the whole year was 48.00%. The severe leaf damage level (grade D = 76-100%) reduced to 0.26% in December. The result of control was evidenced by the new coconut leaves appearing fresh with less damage. The finding agreed with Guo (2005), who reported that in a primary survey the population of *B. longissima* had decreased 5-6 months after parasitoid release. Viet (2004) also showed that the population of *B. longissima* decreased after the release of *A. hispinarum*. In this study, grade A level damage before using larval parasitoids was 17.64% and increased to 42.65% after release. The coconut palms showed clear signs of recovery after 10 months.

Damage levels of newly opened leaves

Monthly grading of the damage to newly opened leaves carried out for one year revealed that the combined damage (A+B+C+D) ranged from 34.67-66.67% with an average of 45.75% (Table 4). Most damage occurred at grade A level (11.22-38.50%). The highest and lowest damage levels were in July (67.33 %) and December (34.67%), respectively. The result was similar to that reported by Morakote (2007), who released *A. hispinarum* in Suratthani Province and found out later that most damage on newly opened leaves was at the grade A level. The newly opened leaves were green and less damaged.

Table 3 Percentage damage levels on 1^{st}-10^{th} leaves of coconut palms by *Brontispa longissima* Gestro evaluated at the Panya Indra golf course, Bangkok from January to December 2007.

Months	The average of percentage of damaged leaves from first to ten leaves					% Damage (A+B+C+D)
	Grade 0 0%	Grade A 1-25 %	Grade B 26-50 %	Grade C 51-75 %	Grade D 76-100 %	
January	57.98	17.64	12.65	8.96	2.77	42.02
February	42.40	38.60	3.80	6.80	8.40	57.60
March	41.56	42.65	3.18	5.67	6.94	58.44
April	38.72	38.65	7.23	7.78	7.62	61.28
May	35.15	36.78	9.63	11.51	6.93	64.85
June	49.86	38.72	3.14	3.13	5.15	50.14
July	52.15	39.42	3.23	2.06	3.14	47.85
August	53.46	36.43	2.78	4.69	2.64	46.54
September	59.78	34.38	2.13	2.55	1.16	40.22
October	62.36	32.50	3.26	1.24	0.64	37.64
November	64.32	32.34	2.11	0.89	0.34	35.68
December	66.24	30.39	1.98	1.13	0.26	33.76
Average	52.00	34.88	4.59	4.70	3.83	48.00
Minimum	35.15	17.64	1.98	0.89	0.26	33.76
Maximum	66.24	42.65	12.65	11.51	8.40	64.85

Table 4 Percentage damage levels of newly opened leaves on coconut palms by *Brontispa longissima* Gestro evaluated at the Panya Indra golf course, Bangkok from January to December 2007.

Months	The average of percentage of damage levels of newly opened leaves					% Damage (A+B+C+D)
	Grade 0 0%	Grade A 1-25 %	Grade B 26-50 %	Grade C 51-75 %	Grade D 76-100 %	
January	42.22	21.39	15.5	12.72	8.17	42.02
February	38.28	32.00	11.22	12.17	6.33	57.60
March	33.33	29.39	14.56	13.16	9.56	58.44
April	32.67	26.44	16.22	13.00	11.67	57.78
May	33.44	38.5	8.22	11.51	8.33	61.72
June	45.72	32.67	5.67	8.61	7.33	66.67
July	56.72	29.56	4.22	6.39	3.11	67.33
August	62.17	26.11	5.11	4.67	1.94	66.56
September	65.33	28.89	2.22	1.95	1.61	54.28
October	73.28	23.72	1.39	1.05	0.56	43.28
November	80.61	17.33	1.33	0.44	0.29	37.83
December	87.22	11.22	1.11	0.33	0.12	34.67
Average	54.25	26.44	7.23	7.17	4.91	45.75

Population study of *Brontispa longissima*

The buildup of the *B. longissima* population commenced from February, then dropped to a relatively low level in March, before sharply increasing in April and reaching its peak in June (Figure 1). Thereafter, it declined and reached its lowest level in December. The highest average egg number occurred in May (35.65 eggs) while the highest larval and pupal numbers were found in June (90.58 larvae) and May (3.23 pupae), respectively. The adult population was also highest in May (31.64 adults). The population density of *B. longissima* reached a low level, nine months after the initial parasitoid release. A biological control agent will take some time to be effective in new areas of spread. More and more evidences now indicates that the successful release of the parasitoid depends on several factors including: temperature, humidity, environment, pesticide used by the farmer and wind (FAO, 2004).

CONCLUSION

The first mummified larvae were recorded five months after release of the parasitoid. The greatest percentage (14.08%) of mummified larvae were collected in November. Almost all coconut palms showed signs of new, healthy leaf growth and very low levels of re-infestation with *B. longissima*. The number of leaves damaged by *B. longissima* was greater from February to May and declined after June until December. The percentage of undamaged leaves increased after six months. The population was high from April to August and low from September to December. Evidently, the parasitoid had established and provided good control of *B. longissima*.

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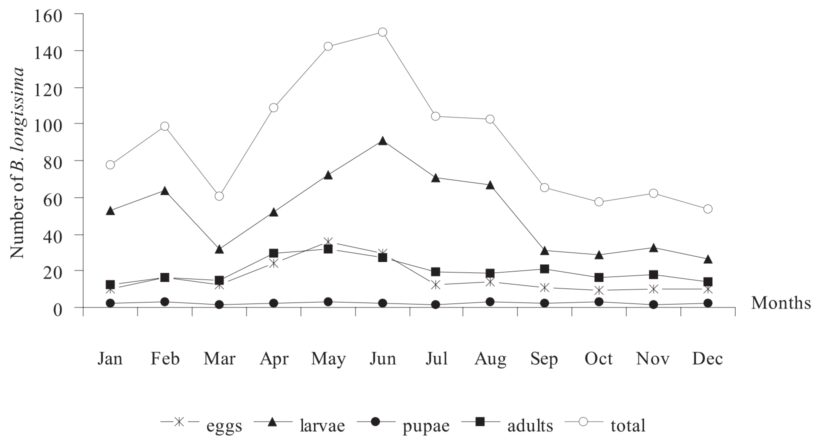

Figure 1 Population dynamics of the coconut hispine beetle, *Brontispa longissima* Gestro per spear from 20 randomly selected spear leaves at the Panya Indra golf course, Bangkok from January to December 2007.

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