THE LIFE CYCLE AND EFFECTIVENESS OF INSECTICIDES AGAINST THE BED BUGS OF THAILAND

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Abstract. The purpose of this study was to determine the life cycle and effectiveness of insecticides against bed bugs in Thailand. Bed bugs from dwelling places in Chon Buri and Chiang Mai provinces were determined to be *Cimex hemipterus* and *Cimex lectularius*, respectively. Both bed bug species were reared by feeding on rabbit blood at 2-day intervals in laboratory conditions at 28-32ºC with 75% relative humidity. The duration from egg to adult stages took 39.9±7.0 and 36.9±8.2 days in *C. hemipterus* and *C. lectularius*, respectively. The insecticides, propetamphos 20% CS, pirimiphos-methyl 50% EC, bifenthrin 25% WP and alpha-cypermethrin 5% SC, were tested against adult *C. lectularius* using a Potter spray tower. The concentrations at which 50% of bed bugs were either dead or moribund (ED_{50}) for propetamphos and pirimiphos-methyl were 6.67 and 14.93 mg/m^2 for the active ingredients on day 3 and the ED_{50} for bifenthrin and alpha-cypermethrin were 1,767.76 and 353.55 mg/m^2, respectively. The results reveal *C. lectularius* in Thailand has a tendency to develop pyrethroid resistance.

Key words: bed bugs, life cycle, insecticide effectiveness, Thailand

INTRODUCTION

The tropical bed bug, *Cimex hemipterus* and common bed bug, *Cimex lectularius* (Hemiptera; Cimicidae), are commonly found in Thailand and have become a major public health problem. Although bed bugs are not a vector of human diseases they impact the quality of life, causing discomfort, sleeplessness, anxiety and inflamed skin. Bed bugs have recently become a greater problem in residences, such as hotels, apartments, dormitories and other dwelling areas. Several hypotheses have been proposed to explain the sudden resurgence of bed bugs, including exchanging second-hand furniture and transportation by travelers from infested areas. People without a knowledge of bed bugs may use a single insecticide or several insecticides with a common mode of action. Bed bugs exposed to insecticides at sublethal doses, may survive and develop insecticide resistance. This investigation was conducted with the aims of identifying bed bugs that found in Thailand and evaluating the efficacy of some insecticides against these bed bugs.

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MATERIALS AND METHODS

Survey of bed bug species and their life cycle

Bed bug survey. Surveys of bed bugs were

carried out in dwelling places, including hotels, dormitories, and guest houses in Chon Buri and Chiang Mai provinces. Nymph and adult bed bugs were collected and brought to the laboratory of the Department of Entomology and Plant Pathology, Faculty of Agriculture, Chiang Mai University, Chiang Mai, Thailand for identification and rearing as stock culture.

Life cycle of bed bugs. Bed bugs were fed every 2 days on rabbit blood from feeding cups modified from clear plastic drinking cups covered with satin cloth over the opening. The rearing method was a modification of that by Peterson (1964). Bed bugs were allowed to feed on the blood through the cloth for 15 minutes per meal. Bed bug populations were kept in closed plastic containers with saturated sodium chloride solution at the bottom to maintain a relative humidity (RH) of 75% (Greenspan, 1977). Rearing and other investigations of the bed bugs were conducted at 28-32ºC.

Regarding the life cycle of bed bugs, 10 newly laid eggs from mated female bed bugs were transferred individually into a small tube. After hatching, the bed bug nymphs were fed on rabbit blood as described above. Daily observations were made and life cycle data recorded throughout the span of developmental period.

Efficacy of some insecticides against bed bugs

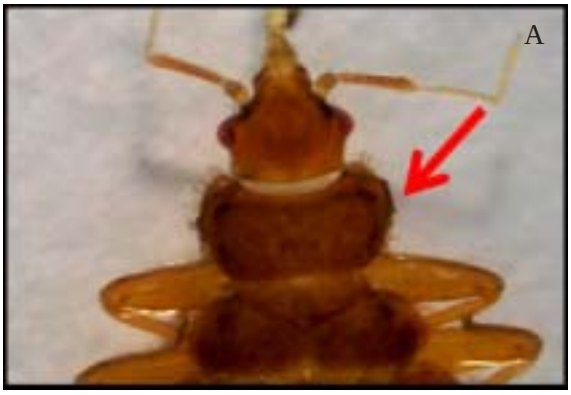
To evaluate the efficacy of organophosphate insecticides (propetamphos 20% CS and pirimiphos-methyl 50% EC) and pyrethroid insecticides (bifenthrin 25% WP and alpha-cypermethrin 5% EC) on the bed bug, *C. lectularius*, each insecticide was diluted with distilled water to 5 concentrations. One type of insecticide was used for each experiment using a randomized design with 4 replications. Four bed bug adults (both sexes) were used for each unit, including a control using distilled water. Before applying insecticides, the bed bugs were exposed to the ice for several minutes in order to make them move more slowly. A potter spray tower was used for insecticide spraying directly on the bed bugs in 9 cm diameter Petri dishes (Potter, 1952). In each glass Petridish, 0.25 ml of insecticide (40 ml/m^2) was applied. Insect mortality was determined at 24, 48 and 72 hours after application of insecticide. The data was corrected using Abbott's formula (Abbott, 1925) when the mortality of the bed bugs in the untreated control was 5-20%. The data were analyzed for ED_{50} using computer software, Logit PC.

RESULTS

Survey of bed bug species and their life cycle

Field collected bed bugs from dwelling places in Chon Buri and Chiang Mai provinces were identified as the tropical bed bug (*C. hemipterus*) (Fig 1A) and the common bed bug (*C. lectularius*) (Fig 1B), respectively. The distinguishing characteristics of the species included the width and length of the pronotum. The pronotum in *C. hemipterus* is twice as wide as it is long (Fig 2A) while that of *C. lectularius* is >2 times as wide as it is long (Fig 2B). The width and length of the pronotum of *C. hemipterus* were 0.57±0.03 and 1.10±0.02 mm, respectively, while those of *C. lectularius* were 0.66±0.02 and 1.22±0.04 mm, respectively (Table 1).

Two species of bed bugs were completely reared throughout their life cycles under laboratory conditions. For the life cycle of *C. hemipterus*, the egg incubation period was 4.00±1.2 days. The number of



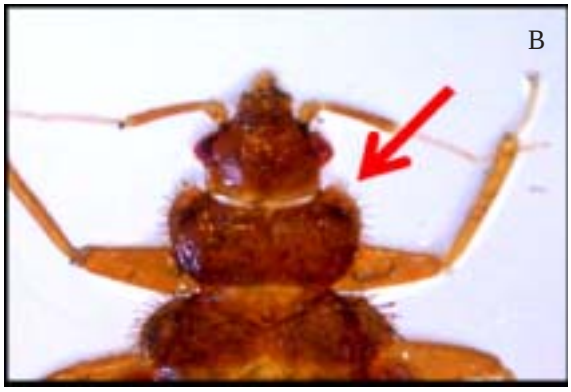


Fig 1–Adult *Cimex hemipterus* (A) and *Cimex lectularius* (B).

Fig 2–Pronotum of *Cimex hemipterus* (A) and *Cimex lectularius* (B).

nymphal instars was five to six. Nymphs molted five to six times before emerging to adults but most of them molted five times. The durations of each successive nymphal instar were 7.5±4.0, 7.9±2.9, 4.3±2.7, 6.3±1.8, and 7.7±2.6 days, respectively. Some individuals (*n*=2) had a prolonged immature stage where the last nymph stage lasted 6.00±1.0 days. Adult longevity was 127.2±27.1 days, and the total life cycle from egg to adult emergence was 39.9±7.0 days (Table 2). For the life cycle of *C. lectularius*, the egg incubation period was 4.0±0.99 days. The number of nymphal instars was five to six. The durations of each successive nymphal instar were 5.5±1.3, 5.3±3.8, 3.9±1.5, 5.2±1.9 and 5.8±2.0 days, respectively. Some individuals (*n*=3) had a prolonged last nymph stage of 24.3±2.8 days which was longer than

Table 1
Width and length of the pronotum of *Cimex hemipterus* and *Cimex lectularius*.

Bed bug species	Pronotum width (mm)		Pronotum length (mm)	
	Mean±SD	Range	Mean±SD	Range
C. hemipterus	0.57±0.03	0.50-0.58	1.10±0.02	1.08-1.13
C. lectularius	0.66±0.02	0.63-0.68	1.22±0.04	1.20-1.25

Table 2
Durations of various developmental stages of *Cimex hemipterus* and *Cimex lectularius* under laboratory conditions (28-32ºC at 75 % relative humidity).

Developmental stage	*Cimex hemipterus*			*Cimex lectularius*		
	N	Mean±SD (days)	Range (days)	*N*	Mean±SD (days)	Range (days)
Egg	10	4.0±1.2	3-6	10	4.0±0.1	3-5
Nymph: Instar I	10	7.5±4.0	3-16	10	5.5±1.3	3-7
Instar II	10	7.9±2.9	7-13	10	5.3±3.8	2-11
Instar III	10	4.3±2.7	3-11	10	3.9±1.5	3-8
Instar IV	10	6.3±1.8	4-10	10	5.2±1.9	4-10
Instar V	10	7.7±2.6	5-12	10	5.8±2.0	4-11
Total life cycle (egg-adult emergence)	10	39.9±7.0	30-53	10	36.9±8.2	24-48
Adult longevity	10	122.2±27.1	90-169	10	127.1±28.7	87-178

with *C. hemipterus*. The duration from egg to adult emergence was 36.9±8.2 days (Table 2).

Efficacy of some insecticides against bed bugs

Effective dose insecticide concentrations at which 50% of *C. lectularius* were either dead or moribund (ED_{50}) on day 3 were 6.67 and 14.93 mg/m^2 for propetamphos and pirimiphos-methyl, and were more 1,767.76 and 353.55 mg/m^2 for bifenthrin and alpha-cypermethrin, respectively.

DISCUSSION

In the past, only *C. hemipterus* was found in dwelling places in Thailand (Ketavan, 1980; Sangwaranond, 1994). However, in our research during 2007-2009, *C. hemipterus* and *C. lectularius* were found in some provinces of Thailand. *C. lectularius* was found in Chiang Mai Province and *C. hemipterus* was found in Chon Buri Province. New outbreaks may by due to people transporting them worldwide (Potter, 2005; Romero *et al*, 2007).

C. hemipterus and *C. lectularius* are similar in external morphology. The 2 species are distinguished by the width of the pronotum, and the darker color and deeper excavated pronotum of *C. lectularius* (Pratt and Stojanovich, 1962; Krinsky, 2002).

A room temperature of 28-32ºC, at a relative humidity of 75% living on a saturated NaCl solution (Greenspan, 1977) seems to promote insect survival. Bed bug development for both species in the laboratory condition took 5-6 instars and 30-45 days to complete which confirmed to the report of Krinsky (2002). A key conditions to breeding bed bugs also included frequent feeding. The molting of bed bugs was based on exuvia found in the cage.

Some chemicals, such as pyrethroids, have been used to treat bed bugs. Bed bugs may not been recognized by tenants. Type I pyrethroid (bifenthrin), a non-alpha cyano pyrethroid and Type II pyrethroid (alpha-cypermethrin), a cyano-3-phenoxybenzyl alcohol pyrethroid which increases insecticidal activity, were used in this experiment. This research found that both bifenthrin and alpha-cypermethrin had ED_{50} of 1,767.76 and 353.55 mg/m^2, respectively. The extrapolated dose used for killing the insects in the field should be double of the ED_{50} value. The dose of alpha-cypermethrin used for controlling American and German cockroaches ranges from 7.5-30 mg/m^2 (PCT Holdings, 2004; BASF, no date); this is a lower dose than that used against bed bugs in this study. These low doses of insecticide application for controlling other urban pests may unintentionally induced bed bug resistance to pyrethroids, similar to other reports by Karunaratne *et al* (2007) and Romero *et al* (2007).

In this experiment at an insecticide application of 40 ml/m^2, the ED_{50} for pirimiphos-methyl against *C. lectularius* on a glass Petri dish was 14.93 mg/m^2 at 24 hours, which is equal to a concentration of 373.25 ppm. Fletcher and Axtell (1993) found an LC_{50} (95% CL) for pirimiphos-methyl against *C. lectularius* of 13.5 ppm (10.0-17.2 ppm) on filter paper. Propetamphos and pirimiphos-methyl are different from pyrethroid insecticides. There have been no reports of resistance against these by bed bugs in Thailand. Propetamphos is an organophosphate available for controlling insects in the house, such as cockroaches, ants, fleas and termites. A typical compressed air spray rate is about 0.5-1% a.i. solution (US EPA, 2000) for propetamphos, and 10 g/l or 1% a.i. for pirimiphos-methyl (WHO, 2006). With the higher concentration (373.25 ppm) of propetamphos, *C. lectularius* showed resistance to the organophosphate. However, propetamphos and pirimiphos-ethyl at recommended application concentrations should be enough to control *C. lectularius*. Propetamphos has been formulated as a capsule suspension (CS) with a lower volatility and better insecticidal activity (Seaman, 1989) than the emulsifiable concentrate (EC) formulation (Bode and Chasin, 1992). Therefore, propetamphos as a CS formulation should be an effective insecticide to control bed bugs.

DDT is the conventional insecticide used for residual spraying since 1954 in the malaria control program of Thailand and other countries of South and Southeast Asia, including Indonesia, Myanmar and Sri Lanka (WHO SEARO, 2006). With long term use of DPT, insecticide resistace of urban pest insects occurred. Pyrethroid insectides were introduced in the 1990s to control malaria vectors, making bed bug exposure to these insecticides likely (Ministry of Public Health, 2003). Cross-resis-

tance between DDT and pyrethroids in another important factor in inset resistance. Further research regarding insecticide resistance among bed bugs needs to be carried out.

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