การเลือกชนิดอาหาร พลวัตประชากรและแบคทีเรียที่พบใน แมลงสาบเยอรมัน *Blattella germanica* L. ในตลาดกรุงเทพมหานคร

นางประชุมพร เลาห์ประเสริฐ

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรดุษฎีบัณฑิต
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FOOD PREFERENCE, POPULATION DYNAMICS AND BACTERIAL HARBORAGE OF THE GERMAN COCKROACH *Blattella germanica* L. IN BANGKOK MARKETS

Mrs. Prachumporn Lauprasert

A Dissertation Submitted in Partial Fulfillment of the Requirements

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Thesis Title	FOOD PREFERENCE, POPULATION DYNAMICS AND BACTERIAL HARBORAGE OF THE GERMAN COCKROACH *Blattella germanica* L. IN BANGKOK MARKETS
By	Mrs. Prachumporn Lauprasert
Field of Study	Biological Sciences
Thesis Advisor	Duangkhae Sitthicharoenchai, Ph. D.
Thesis Co-advisor	Assistant Professor Kumthorn Thirakhupt, Ph. D.

Accepted by the Faculty of Science, Chulalongkorn University in Partial Fulfillment of the Requirements for the Doctoral Degree

.... Dean of the Faculty of Science
(Professor Piamsak Menasveta, Ph. D.)

THESIS COMMITTEE

.... Chairman
(Professor Siriwat Wongsiri, Ph. D.)

.... Thesis Advisor
(Duangkhae Sitthicharoenchai, Ph. D.)

.... Thesis Co-advisor
(Assistant Professor Kumthorn Thirakhupt, Ph. D.)

.... Member
(Assistant Professor Suthep Thaniyavarn, Ph. D.)

.... Member
(Art-Ong Pradatsundarasar, Ph. D.)

.... Member
(Tosak Seelanan, Ph. D.)

.... Member
(Usavadee Thavara, Ph. D.)

ประชุมพร เลาห์ประเสริฐ: การเลือกชนิดอาหาร พลวัตประชากรและแบคทีเรียที่พบในแมลงสาบเยอรมัน *Blattella germanica* L. ในตลาดกรุงเทพมหานคร (FOOD PREFERENCE, POPULATION DYNAMICS AND BACTERIAL HARBORAGE OF THE GERMAN COCKROACH *Blattella germanica* L. IN BANGKOK MARKETS) อ. ที่ปรึกษา: อ. ดร. ดวงแข สิทธิเจริญชัย, อ. ที่ปรึกษาร่วม: ผศ. ดร. กำธร ธีรคุปต์, 212 หน้า

นำแมลงสาบเยอรมันเพศผู้และเพศเมียที่อดอาหารนาน 48 ชั่วโมงมาทดสอบกับอาหารทั้ง 8 ชนิด ได้แก่ ขนมปัง น้ำตาล กล้วย มันฝรั่ง ถั่วลิสงป่น เนยแข็ง เนื้อหมู และ อาหารแมวสำเร็จรูป ซึ่งใช้ดัชนีชี้วัดของรอดเจอร์สในการบ่งชี้การเลือกชนิดอาหารของแมลงสาบ โดยที่แมลงสาบเยอรมันเพศผู้ชอบกินกล้วยและมันฝรั่งอย่างมีนัยสำคัญทางสถิติ ในขณะที่แมลงสาบเยอรมันเพศเมียชอบกินกล้วยอย่างมีนัยสำคัญทางสถิติ นอกจากนี้แมลงสาบเยอรมันเพศเมียเลือกกินถั่วลิสงป่น น้ำตาล และอาหารแมวสำเร็จรูปมากกว่าเพศผู้อย่างมีนัยสำคัญทางสถิติ พฤติกรรมการหาอาหารของแมลงสาบเยอรมันเกิดขึ้นในเวลากลางคืนโดยเกิดขึ้น 2 ช่วงเวลา ช่วงแรกเกิดขึ้นในเวลา 19.00-22.00 น. และช่วงที่สองเกิดขึ้นในเวลา 04.00-05.00 น.

การศึกษาพลวัตประชากรของแมลงสาบเยอรมันในตลาด 12 แห่งของกรุงเทพมหานครนั้น ได้ศึกษาระหว่างเดือนมีนาคม 2548 ถึงมีนาคม 2549 โดยศึกษาเดือนละ 1 ครั้ง แมลงสาบเยอรมันพบมากที่สุดในเดือนกรกฎาคมและสิงหาคม ในขณะที่พบน้อยที่สุดในเดือนธันวาคมและมกราคม แมลงสาบเยอรมันที่พบมากที่สุดคือระยะตัวอ่อนขนาดใหญ่ (ตัวอ่อนระยะ 5 และระยะ 6) และพบแมลงสาบเยอรมันมากที่สุดในตลาดแห่งหนึ่งที่มีการสุขาภิบาลไม่ดี ในเขตที่มีความหนาแน่นประชากรของกรุงเทพมหานครต่ำ ในขณะที่ไม่พบแมลงสาบเยอรมันเลย ในตลาดแห่งหนึ่งที่มีการสุขาภิบาลที่ดีในเขตที่มีความหนาแน่นประชากรของกรุงเทพมหานครสูง นอกจากนี้พบว่าแมลงสาบเยอรมันในร้านขายของชำพบมากกว่าในร้านขายเนื้อสดและร้านขายผักอย่างมีนัยสำคัญทางสถิติ

ทำการแยกแบคทีเรีย ที่ต้องการออกซิเจนในการเจริญเติบโตและแบคทีเรียที่เจริญได้ทั้งสภาพที่มีออกซิเจนและไม่มีออกซิเจน จาก 2 ส่วนของแมลงสาบเยอรมันและแมลงสาบอเมริกัน ได้แก่จากภายนอกร่างและจากทางเดินอาหารส่วนกลาง พบแบคทีเรีย 21 ชนิดจากแมลงสาบเยอรมัน ในขณะที่พบแบคทีเรีย 26 ชนิดจากแมลงสาบอเมริกัน แบคทีเรียก่อโรครุนแรงที่พบในการศึกษาครั้งนี้ได้แก่ *Bacillus cereus* *Staphylococcus aureus* และ *Salmonella arizona* ส่วนแบคทีเรียฉวยโอกาสชนิดที่สำคัญที่พบในการศึกษานี้ได้แก่ *Pseudomonas aeruginoga* ซึ่งพบในทั้ง 12 ตลาดของกรุงเทพมหานคร นอกจากนี้ทั้ง 12 ตลาดยังพบ *Escherichia coli* ซึ่งเป็นดัชนีชี้วัดการปนเปื้อนอุจจาระจากมนุษย์และสัตว์เลือดอุ่น จำนวนชนิดของแบคทีเรียในตลาดที่มีการสุขาภิบาลแย่พบมากกว่าตลาดที่มีการสุขาภิบาลที่ดีอย่างมีนัยสำคัญทางสถิติ ดัชนีความเหมือนระหว่างชนิดของแบคทีเรียที่พบในแมลงสาบเยอรมันและแมลงสาบอเมริกันมีค่าระหว่าง 0.87 ถึง 1.00 ซึ่งผลการทดลองนี้สามารถบ่งชี้ได้ว่าแมลงสาบทั้งสองชนิดสามารถเป็นพาหะนำเชื้อแบคทีเรียชนิดที่มีความคล้ายคลึงกัน

การวิเคราะห์สารกำจัดแมลงที่ตกค้างในแมลงสาบเยอรมันที่ยังมีชีวิตนั้นวิเคราะห์ โดยใช้เครื่องโครมาโตกราฟีแบบของเหลวสมรรถนะสูง ความเข้มข้นสูงสุดของสารกำจัดแมลงที่ตกค้างในแมลงสาบเยอรมันที่ยังมีชีวิตคือสารไซเปอร์มีทริน รองลงมาคือกรดฟีนอกซีเบนโซอิกและสารไซฟลูทริน ซึ่งความเข้มข้นสูงสุดของสารไซเปอร์มีทริน กรดฟีนอกซีเบนโซอิกและสารไซฟลูทรินพบในตลาดแห่งหนึ่งที่มีการสุขาภิบาลดีในเขตความหนาแน่นประชากรของกรุงเทพมหานครต่ำ นั่นเป็นไปได้ว่าแมลงสาบเยอรมันได้มีการพัฒนาความต้านทานต่อสารไซเปอร์มีทรินและไซฟลูทรินในกลุ่มประชากรขึ้น หลังจากที่ได้รับสารกำจัดแมลงชนิดเดิมอย่างซ้ำๆเป็นเวลานาน

สาขาวิชา......วิทยาศาสตร์ชีวภาพ........ ลายมือชื่อนิสิต......................................

ปีการศึกษา...........2549...................... ลายมือชื่ออาจารย์ที่ปรึกษา......................

ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.......................

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KEY WORDS: GERMAN COCKROACH / FOOD PREFERENCE / POPULATION DYNAMICS / BACTERIAL HARBORAGE / INSECTICIDAL RESIDUES / BANGKOK MARKET

PRACHUMPORN LAUPRASERT: FOOD PREFERENCE, POPULATION DYNAMICS AND BACTERIAL HARBORAGE OF THE GERMAN COCKROACH *Blattella germanica* L. IN BANGKOK MARKETS. THESIS ADVISOR: DUANGKHAE SITTHICHAROENCHAI, Ph.D., THESIS CO-ADVISOR: ASST. PROF. KUMTHORN THIRAKHUPT, Ph.D., 212 pp.

The 48-h-starved male and female German cockroaches were given choices among eight food items such as bread, sugar, banana, potato, peanut, cheese, pork, and cat food. The amount of food eaten was recorded using the Rodgers's index for indicating the food preference of the cockroaches. The male German cockroaches significantly preferred banana and potato whereas the female cockroaches significantly preferred only banana. Additionally, the female cockroaches also significantly preferred peanut, sugar, and cat food more than the males. The foraging time of the German cockroach occurred at nighttime with two peaks of feeding activity. The first peak occurred during 07.00-10.00 pm, then, the second peak occurred during 04.00-05.00 am.

A study on the population dynamics of the German cockroach was monitored in twelve Bangkok markets from March 2005 to March 2006. The selected market areas were conducted monthly during the study period. The two highest peaks of the German cockroach were in July and August whereas the two lowest peaks of the cockroach were in December and January. The highest catch of the German cockroach was the large nymphal stage (the 5^{th} and 6^{th} instars). The highest number of the German cockroaches was in a poor sanitary market in low population density zone of Bangkok whereas none of the German cockroach was caught from a good sanitary market in high population density zone of Bangkok. Moreover, the cockroaches caught from the groceries were significantly higher than the butcher shops and vegetable shops in the twelve Bangkok markets.

Aerobic and facultative anaerobic bacteria were isolated from 2 parts of the German and the American cockroaches, the external cuticles and the middle guts. The German cockroach hosted 21 species of bacteria whereas the American cockroach hosted 26 species. The serious pathogens in this study were *Bacillus cereus*, *Staphylococcus aureus* and *Salmonella arizona*. The opportunistic pathogens such as *Pseudomonas aeruginoga* were found in all markets. Additionally, *Escherichia coli*, an indicator of environmental surveillance as a measurement of human and warm-blooded animal fecal contamination, was also isolated from all markets. The bacterial species isolated from the cockroaches from poor sanitary markets was significantly higher than from good sanitary markets. The similarity coefficient of the bacterial species between the German and the American cockroaches investigated in twelve Bangkok markets were ranged from 0.87 to 1.00. The results indicate that the both kinds of cockroaches carried the similar bacterial species.

Insecticidal residues in the living German cockroach were analyzed by High Performance Liquid Chromatography. The highest concentration of insecticidal residue in the living cockroach samples was cypermethrin, followed by 3-phenoxybenzoic acid, and cyfluthrin, respectively. The highest residue concentrations of cypermethrin, 3-phenoxybenzoic acid, and cyfluthrin were detected from a good sanitary market in low population density zone of Bangkok. This suggests that cypermethrin and cyfluthrin resistances may have been developing in the German cockroach populations after being treated by the repetitious insecticides.

Field of Study......Biological Sciences....... **Student's Signature**...Prachumporn Lauprasert...

Academic Year..............2006.................... **Advisor's Signature**...Duangkhae Sitthicharoenchai...

Co-advisor's Signature...K. Thirakhupt...

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CONTENTS

Contents (Continued)

LIST OF TABLES

List of Tables (Continued)

List of Tables (Continued)

List of Tables (Continued)

LIST OF FIGURES

List of Figures (Continued)

List of Figures (Continued)

List of Figures (Continued)

Figure **Page**

List of Figures (Continued)

CHAPTER 1

INTRODUCTION

Cockroaches, sometimes known as roaches or waterbugs or streambugs, are worldwide urban pests in human environments (James and Harwood, 1969; Roth, 1991). They are one of the oldest insect order and they have survived on this planet for over 300 million years since the Upper Carboniferous (the Pennsylvanian) Periods (Wootton, 1981). Currently, the cockroaches are found in the world approximately 4,000 species in 460 genera (Roth, 1991). The cockroaches are success in peridomestic and domestic habitats because of their biology and behaviors (Ross and Mullins, 1995). Most of cockroach species live in the nature habitats in the tropical and subtropical climates, and they also inhabit temperate and boreal regions (Brenner, 1995). About 1% of cockroach species are the common pest in the world (James and Harwood, 1969; Roth, 1991; Grimaldi and Engel, 2005).

In Thailand, many cockroaches found are outdoor, indoor, and also peridomestic species. Ten species of the cockroaches have been reported, but only some species have medical significance (Tawatsin *et al.*, 2001). For example, the German cockroach, *Blattella germanica* Linnaeus, a small indoor pest, which infests in humid and warm environment such as commercial food preparation, storage facilities, kitchens, larders, and restaurants (James and Harwood, 1969; Ross and Mullins, 1995). Another example is the American cockroach, *Periplaneta americana* Linnaeus, which frequently inhabits sewers, steam tunnels, and drainage system (James and Harwood, 1969). Both of them are worldwide urban pests and are very difficult to control (Gold, 1995; Tsai and Lee, 2001).

Although the cockroaches do not sting or bite, but it adversely affect human health in several ways such as transmitting pathogenic agents mechanically on its body part (Brenner, 1995). The German cockroach was reported as carriers of viruses, bacteria, fungi, protozoans and helminthes (Ross and Mullins, 1995; Benson and Zungoli, 1997). The decomposing remains of dead cockroaches and their feces are an important source of an air-borne disease by inducing respiratory allergies in sensitive people (Ross and Mullins, 1995; Sarinho *et al*., 2004).

Nowadays, the two main methods for the cockroach control are chemical and non-chemical controls. The residues from using of chemicals are able to cause a strong negative impact to the environment and non-target organisms. Long term control of the German cockroaches may be difficult because the German cockroaches have short life cycle leading to rapid population recovery (Ross and Mullins, 1995) and it has a considerable ability to develop resistance to a variety of chemical insecticides (Cochran, 1989).

The experiments in this research include investigation of the attractiveness of various foods to the male and female German cockroaches, study of their feeding behavior, monitoring the population dynamics of the German cockroaches in Bangkok markets, isolation of prevalent and human pathogenic bacteria, and evaluation of the insecticidal residues in the German cockroaches captured from the study areas. Ultimately, the goal of this research was to provide basic information for the development of control strategies for the German cockroaches in the urban environment.

CHAPTER 2

LITERATURE REVIEW

1. Fossil and Origin

Cockroaches are an ancient group of insects which have survived on this planet for over 300 million years since the Upper Carboniferous (the Pennsylvanian) Periods. Cockroaches are the predominant insect group in the Carboniferous deposits in Siberia and in the Lower Permian coal-measures of North America, Europe, and the Ural Region (Wootton, 1981).

Cockroach fossils were found in large numbers from coalmines in the Upper Carboniferous Period, a period that the palaeontologists have labeled it as the age of the cockroach (Wootton, 1981; Copeland, 2003). The oldest cockroach fossil is *Palaeoblatta douvillei* Brongniart which found from Silurian sandstone in Carboniferous deposits of the Devonian period (Blatchley, 1920 cited in Appel, 1995). Moreover, the largest (90 mm) complete cockroach fossil unearthed so far was discovered by geologists from a coalmine in northeastern Ohio. It was dated from about 300 million years ago, some 55 million year before the first dinosaurs (Ohio State University, 2001).

By the Early Carboniferous, vast coal swamps had developed, and large arborescent plants dominated the landscape. Land plants had finally taken hold, and the forests covered many regions of the world. Insects also radiated and had become diverse in the carboniferous (Wootton, 1981; Grimaldi and Engel, 2005). The Carboniferous rocks had a large number of cockroaches and the other archaic insects. This may be because they preferred the climatic conditions in that period

(Wootton, 1981). Their habitats were among ferns and plants of that time, moist areas along banks of rivers, marshes and swamps. They were recorded in the Carboniferous beds when the climate changed in the Lower Permian (James and Harwood, 1969; Wootton, 1981).

The other cockroach fossils have been preserved in amber. Amber, sometimes called fossil resin or resinite, is ancient tree resin. The subtropical rainforest trees produced resinous sap, in response to stress or injury, when fossilized, that know as amber (Copeland, 2003; Grimaldi and Engel, 2005). Fossil resins are scattered throughout the earth's surface in deposits from the Carboniferous to the Holocene (Grimaldi and Engel, 2005). The cockroach fossils preserved in rock unlike the fossils preserved in amber. It seems to be a species unlike any of the modern cockroaches. The quality of preservation is outstanding down to the finest details of wing colors and veining and eye lenses. Such preservation is usually found only in amber specimens in settings as diverse as the Baltic region, the Central American, and several Cretaceous sites (Copeland, 2003).

By at least the end of Devonian period (360 million years ago), not far from the ocean shores, insects had evolved and originated from the other arthropods that had successfully made the transition from ocean to land. Insects and arthropods alike are descended from a single segmented ancestor, the warmish onychophoran. The cockroach's form evolved directly from the segmented form and simple anatomy of the onychophoran, a very functional and successful pattern (Copeland, 2003).

There is a little bit difference in general morphology between the cockroach fossils and the present day cockroaches. The most obvious features include oval and flattened dorsoventrally bodies, a large pronotal shield (pronotum), often covering the entire dorsal surface of the head, and long wings and legs. Little structural change over a long period indicates successful characteristics and the high adaptability of these insects (James and Harwood, 1969; Wootton, 1981; Appel, 1995).

2. Classification and distribution

Several methodologies and numerous characters have been used to classify cockroaches. The classification of cockroaches is usually based on four characters such as the morphology of the proventriculus, the male and female genitalia, the musculature, and oviposition behavior (McKittrick, 1964).

Cockroaches belong to phylum Arthropoda, class Insecta, and are listed in the order Blattodea (the former name: Blattaria). McKittrick (1964) reported that the order Blattodea is divided into five families such as Blaberidae, Blattidae, Blattellidae, Cryptocercidae, and Polyphagidae. Nevertheless, Roth (1991) divided the order Blattodea into six families such as Blaberidae, Blattidae, Blattellidae, Cryptocercidae, Polyphagidae, and Nocticolidae. Moreover, they were classified into 460 genera, with about 4,000 described species. The Blattidae is the largest family, with approximately 525 species (Roth, 1991). Most of them live in tropical and subtropical forests, cryptically dwelling under stones and bark, and in logs. About 1% of cockroaches being the common pest in the world especially, the two families, the family Blattidae such as the American cockroach, *Periplaneta americana* Linnaeus and the family Blattellidae such as the German cockroach, *Blattella germanica* Linnaeus (James and Harwood, 1969; Roth, 1991; Grimaldi and Engel, 2005).

The members of family Blattellidae are small in shape and usually long-legged insects. The ventral side of meso- and metafemora armed with well developed two rows of spines. Male subanal plate asymmetrical, female subanal plate not longitudinally divided. Neither dark punctuation nor pubescence on the prothoracic tergite. Hindwing without any remarkable triangular area at the margin between Cu1 and 3A vein. They are oviparous insect (Asahina, 1983).

The important species of this family is the German cockroach (James and Harwood, 1969). Based on the Common Names of Insects and Related Organisms, Entomological Society of America (Bosik, 1997), the hierarchic name of the German cockroach was reported as:

phylum	: Arthropoda
class	: Insecta
order	: Blattodea
family	: Blattellidae
subfamily	: Blattellinae
genus	: *Blattella*
scientific name	: *Blattella germanica*, Linnaeus

The German cockroach has been reported as an important pest species throughout the world. Worldwide distribution of the German cockroach has been affected by maritime trading, holds of vessels and the galleys (James and Harwood, 1969). This species was found on all continents, especially in temperate regions (Runstrom and Bennett, 1990), in association with humans and their food or waste. Moreover, this species has never been found in locations away from humans or human activity (Appel, 1995). The German cockroach found primarily in eastern Asia, it is probable that *B. germanica* spread from this area (Roth, 1985).

3. Characteristic and morphology

The German cockroach, *B. germanica,* is usually oval and dorso-ventrally flattened insect easily recognized by its general appearance with three body regions: head, thorax, and abdomen (Figure 2.1).

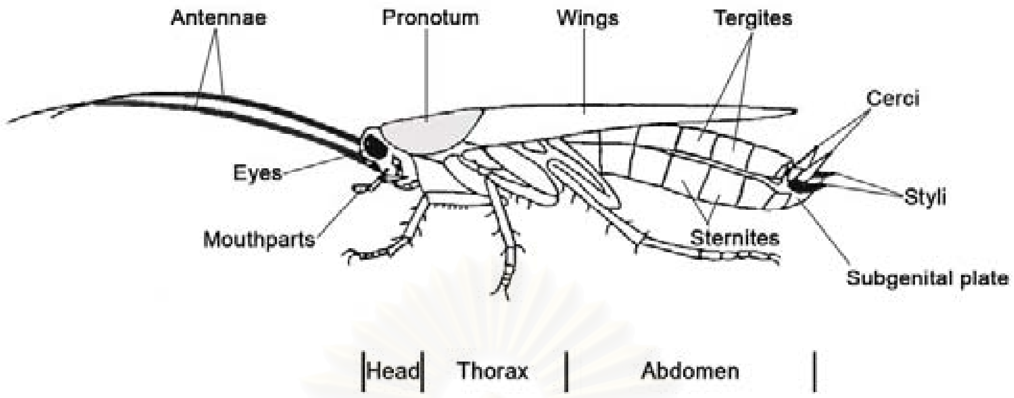

Figure 2.1 Side view of an adult German cockroach (modified from Appel, 1995).

Head

On the head, the cockroaches have chewing mouthparts that are use to chew or scrape off food too large for them to swallow whole. Cockroaches also have a pair of large compound eyes, but have poor vision except that they distinguish easily between light and dark. Also on the head, has a pair of long and well-developed antennae. The olfactory receptors that detect odors, sensory organs that detect water vapors and vibrations in the air located on the antenna. Inside the head is a small brain that coordinates the various body functions (Ross and Mullins, 1995; Tichy, Hinterwirth and Gingi, 2005).

Thorax

The cockroaches have three pairs of legs on the thorax. They also have two pairs of wings. The forewing or tegmina is usually more sclerotized than the hindwing. The hindwing can range from the thickness of the tegmina to a thin transparent membrane. The hindwing is usually larger than the tegmina and folded to fit under the tegmina. The German cockroaches do not fly even though they have retained full size wings but they will sometimes open their wings to break a fall. They do have excellent running abilities and crawl across the wall or ceiling. There is a large plate-like structure on the thorax, just behind the head. On this structure, the pronotum, has two parallel dark streaks.

Abdomen

The abdomen of the cockroaches locates the reproductive system. The eggs are enclosed in a tough eggcase which protects them for drying out. The female German cockroach carries the eggcase around with her until the eggs within it are nearly ready to hatch. On the end of abdomen has a pair of cerci, projections that also serve as sensory organs. Cerci function is similar to antennae, sensing vibrations through air or ground. Cerci are directly connected to the legs of the cockroach via abdominal nerve ganglia, which is an important survival adaptation (Ross and Mullins, 1995).

4. Biology and reproduction

The life cycle

The German cockroach has three life stages typical of insects with incomplete metamorphosis: the egg, larva or nymph, and adult (Figure 2.2). One life cycle may be completed in 100 days under favorable environment conditions (Ross and Mullins, 1995). However, factors such as temperature, nutritional status, and strain differences may influence the time required to complete a life cycle. German cockroaches breed continuously with many overlapping generations present at any one time. Under ideal conditions, population growth has been shown to be exponential (James and Harwood, 1969; Ross and Mullins, 1995).

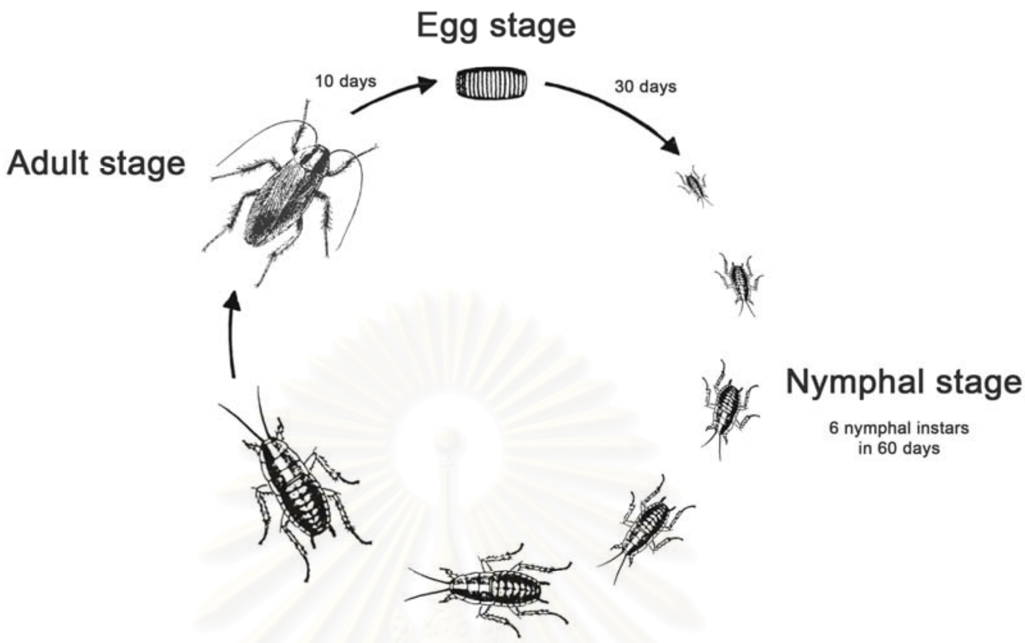

Figure 2.2 The life cycle of the German cockroach, *B. germanica*. Most females produce four to eight viable eggcases that hatch at about monthly intervals. There are usually 30 to 40 eggs per eggcase. Generally there are six nymphal instars (modified from Ross and Mullins, 1995).

Under field studies, three to four generations were completed within one year. The cycle began with fertilization of the egg. The egg hatched into a small but fully-developed nymph that grows through a series of molts, shedding its old cuticle (exoskeleton) at each molt. The cockroach emerged as a fully-winged adult at the final molt. Newly hatched or emerged insects appeared white but became fully pigmented within 24 hours. Actively growing field populations of the German cockroaches were comprised of 80 percent nymphs and 20 percent adults (Ross and Mullins, 1995).

<u>The egg stage</u>

The egg stage begins with fertilization of the ovum and finished with hatch. Eggs are fertilized within the female and pass into an outer chamber, called the vestibulum, which lies above the enlarged seventh abdominal sternite. The German cockroach female produces her first ootheca 11 or 12 days after becoming an adult. Within the vestibulum, the eggs are covered by secretions from the nearby collaterial glands. The eggs are located in 2 parallel rows and are encased by the outer covering, which by then is a relatively hard shell. The eggcase is a tiny, brown, purse-shaped capsule. The shell of the eggcase is semi-transparent. It is about 8 mm long, 3 mm high, and 2 mm wide. There are usually 30 to 40 eggs per eggcase, but they can be as many as 48. (Sagulgim, 1981; Ross and Mullins, 1995). As more eggs enter the vestibulum, the eggcase or capsule (the scientific term is ootheca) begins to protrude from the genital chamber and becomes visible externally. Protusion of the eggcase is completed in 14-16 hours after individual fertilized eggs begin to enter the vestibulum. Once the translucent tip of the eggcase becomes visible, the entire eggcase will be fully developed and entirely visible by the following day, changing from white to pink within a few hours. Within a day or two, it becomes light brown and finally chestnut. The newly-formed eggcase is generally rotated 90° to the right. Unlike other domiciliary species, the female carries the eggcase externally as long as a month, until hatch or drops it within a short time (around 1-2 days) before the eggs are ready to hatch (Figure 2.3).

In an investigation, an average of 29.9 nymphs were hatched per eggcase in an average of about 28 days at ordinary room temperature (Gould and Deay, 1940), while in another investigation at 35° C, eggs hatched in 14 days (Willis, Riser, and Roth, 1958). After the fourth eggcase was produced, the number of eggs per eggcase gradually decreased to about 75% of the original number in the seventh and eighth eggcase (Ross and Mullins, 1995).

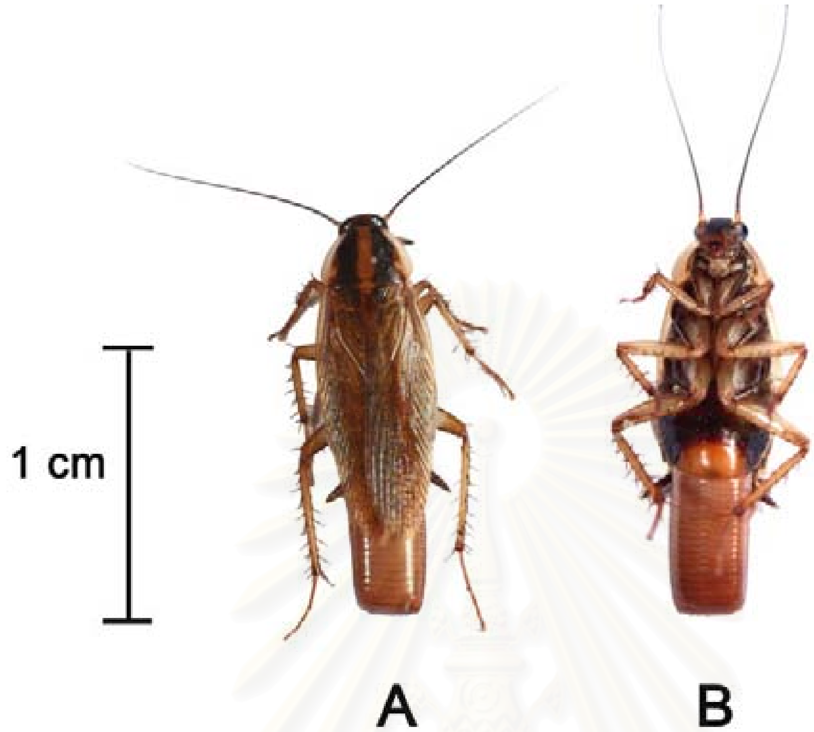

Figure 2.3 A female German cockroach, *Blattela germanica*, carries the eggcase, dorsal (A) and ventral (B).

The nymphal stage

The nymphal stage begins with hatching of the eggcase and, after a series of molts, finishes with emergence of the adult at the final molt (Ross and Mullins, 1995). Nymphs are dark brown to black in color, with distinct dark parallel bands running the length of the pronotum (Figure 2.4 A). Nymphs do not possess wings. The numbers of molts require to reach the adult stage varies, but the most frequently reported number of molts is six. The stage between molts is called an instar. At room temperature nymphs complete development in about 60 days. All developmental stages actively forage for food and water. The smaller nymphal instars are sometimes isolated from the remainder of the colony, particularly when they have gained access to a crack or crevice too narrow for the older nymphs and adults (James and Harwood, 1969).

The first-instar nymph is only 3 mm long. The body is dark gray to almost black, except for the second and third thoracic segments, which are pale brown. The pale-brown band conspicuously characterizes the first-instar nymph. In succeeding nymphal instars, the light band becomes narrower and extends in both directions to become a median longitudinal stripe. In its anterior extension, it eventually becomes the median pale-brown stripe dividing the "two parallel dark streaks" that characterize the pronotum of the adult. The remainder of this stripe is covered by the wings. Molted skins are quickly eaten by the nymphs that emerge from them or by other cockroaches that happen to be near-by (Ross and Mullins, 1995).

The adult stage

The adult that emerges at the final molt (adult ecdysis) is fully winged. The wings can be used to glide, but the primary method of locomotion is by the legs. Males are easily distinguished from females by the following characteristics: male is yellowish, body thin and slender, posterior abdomen is tapered, terminal segments of abdomen visible, not covered by tegmina (leathery outer wings). Whereas female is body stout, posterior abdomen is rounded, entire abdomen just covered by tegmina (Figure 2.4 B, C). Adult German cockroaches are 10 to 15 mm long, pale brown or tan, and have 2 parallel dark streaks on the pronotum (Ross and Mullins, 1995). The female cockroaches heavily invest in production of large ootheca unlike from males that produce small spermatophores (Leibensperger, Traniello, and Fraser, 1985). They have chewing mouthparts. Their movements are very rapid when they are disturbed. They are nocturnal. If a few are seen crawling about in open spaces during daylight hours, this indicates that the infestation is already severe (James and Harwood, 1969; Ross and Mullins, 1995).

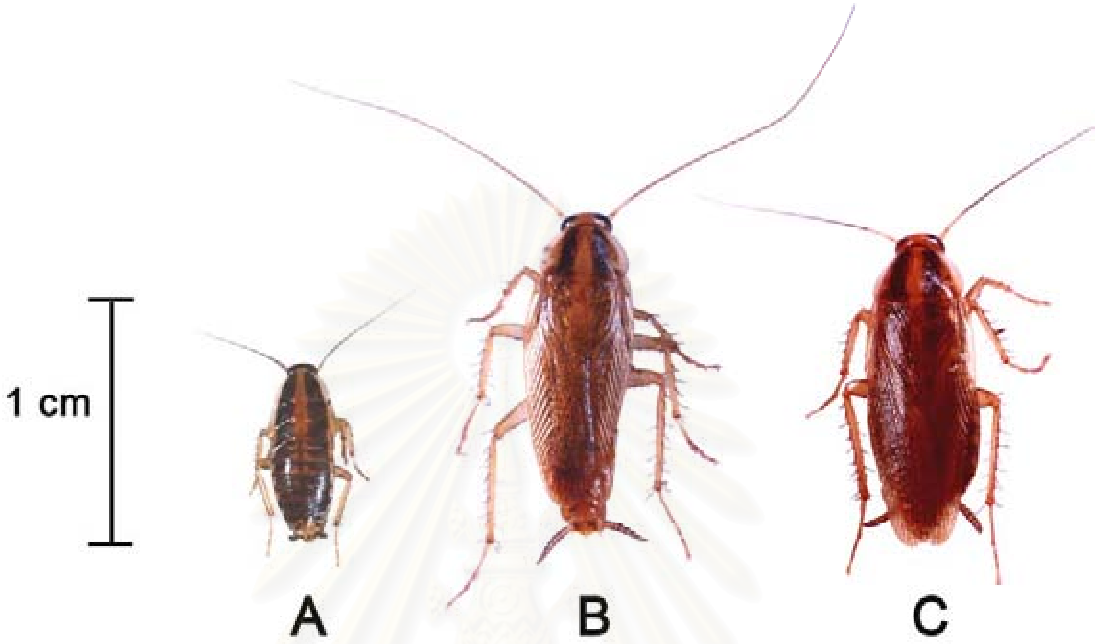

Figure 2. 4 A nymph (A), male (B) and female (C) German cockroach.

5. Ecology and behavior

Habitat

The German cockroach is nocturnal and omnivorous. It is an indoor cockroach that spends most of its time hiding in cracks and crevices in dark, warm, and humid areas close to food and water (Ross and Mullins, 1995). The German cockroach prefers temperatures from 24-33 °C (Cornwell, 1976). It is most abundant in the kitchen, during the day it may be found under stoves or ranges and refrigerators. Moreover, it is abundant in the insulation of the wall's appliances; under the sink (especially in the "dead space" between the sink and the wall); under and behind the water heater; behind cabinets, pantries, and closets; behind baseboards and moldings. It is also abundant in other cracks, crevices, and dark protected areas (Ross and Mullins, 1995; Copeland, 2003).

Foraging behavior and food preference

Eating and drinking activities of *B. germanica* are related to the more general circadian activity phase. Their daily rhythm of foraging activity contains two phases during the night time. The first phase happens a few hours after sunset, and the second phase happens about one hour before sunrise. Males, females with oothecae, and nymphs usually leave harborages at night time to forage food and water. While females with oothecae usually limit their foraging activity and stay close to harborages until the oothecae are deposited. Nymphs reduce their activity and stay in their harborages for few days before molting to the next stage (Brett, Ross, and Holtzman, 1983; Metzger, 1995). Moreover, the gravid females were able to survive without water for 5 days and could live without food for longer (Durbin and Cochran, 1985).

The relationship between population density and the frequency bouts of drinking has not been substantiated. The cockroaches drink more frequently at lower population densities than they do at higher densities. This might be a reaction to increase competition of water source and might relate to interruption of drinking under high density conditions (Ebeling and Reierson, 1970; Silverman, 1986).

For the nutritional requirements, it is well known that under normal condition, the German cockroaches prefer diets which are higher in carbohydrate than in fat and protein content (Ross and Mullins, 1995). Moreover, the female cockroaches require substantially protein more than the males (Clarebrough, Mira, and

Raubenheimer, 2000) because female fecundity commonly depends on the ingestion of protein that necessary for egg development (Reierson, 1995). While under stress condition such as starvation, the cockroaches often ate the nearest food substance that they encounter. The German cockroach oriented to food or water only if they came close to it after they had begun foraging, particularly after being deprived of food or water for a few days. Their feeding behavior was related to degree of starvation. Their behavior corresponded with the other insects which did not respond to monotonous odors as vigorously as when they were very hungry (Reierson, 1995).

Mating Behavior

The male of the German cockroach cannot detect the female from a distance, even when in close proximity, but has to make physical contact, ordinarily using the antennae. The antennae of the female, as well as other body regions, contain a chloroform-soluble, nonvolatile substance that will stimulate the male sexually. Thus, sex discrimination by males is mainly owing to "contact chemoreception". It follows that the male German cockroach probably cannot be attracted from a distance by means of synthetic sex attractants (provided they are available) as some other insect species can be (Roth and Willis, 1952; Ross and Mullins, 1995).

When male and female meet, their antennae touch and vibrate against each other. The male then turns around, raises his wings to expose the orifices of a pair of dorsal glands located on the seventh and eighth tergites, and extends his abdominal segments to expose the openings of the 2 pairs of glands. The glands are not normally visible, being covered by the wings and by the margins of the preceding abdominal sclerites. Both wing-raising and extension of the abdomen are required to uncover the glands. The female eats a secretion from these glands. After she has fed for a few seconds, the male-pushes his abdomen farther back, and connection of the genitalia is made. The male then moves out from under the female, and the pair remain attached in a linear position for an average of about 86 minutes (Ross and Mullins, 1995). The German cockroaches do not need to be attracted from long distances, as do many other insects for the meeting of sexes, is understandable in view of the habits of cockroaches in general. The chance meeting of opposite sexes is enhanced by the fact that they are negatively phototactic and positively thigmotactic (principally guided by contact), and that they are gregarious, with large numbers seeking the same environment. They seek secluded and particularly very narrow hiding places, such as cracks, crevices, and voids, especially those having optimum temperature and moisture conditions. Attraction to the odor of the species is another factor that favors aggregation. Once the sexes have been brought together by these different stimuli, more refined stimuli lead to sexual discrimination (Metzger, 1995).

Aggregation Pheromone

The German cockroaches have a gradual metamorphosis, all immature instars may be seen together with the adults. They tend to congregate in a single area, leaving it to search for food and water, then returning. An aggregation pheromone, present in the feces and on the bodies of the insects (it spreads easily on the greasy epicuticular lipid), is responsible for this tendency to aggregate. The gregarious behavior of nymphs favors their growth and development. They do not develop so well when isolated. Isolated nymphs that could only see or smell the others, or received contact only with the washings of the body surfaces of other nymphs, or with filter paper contaminated with feces, do not grow at the group rate. This indicates that a "psychological factor" affecting the endocrine system by means of tactile stimulation, chiefly by the antennae, is the principal factor involving in growth acceleration (Izutsu, Ueda, and Ishii, 1970; Metzger, 1995).

Avoidance behavior

The avoidance of lethal toxicants is the result of a learning process of individual insects. The German cockroach actively seeks darkness. However, when they are exposed to negative stimuli such as repellent insecticides inside the refuges, they learn to avoid the dark refuges and stay in lighted areas even during resting periods. Avoidance behavior depends strongly on the repellency of the applied toxicant. The practical significance of avoidance behavior is that sufficiently repellent insecticides induce the cockroaches to minimize subsequent contact with toxicants. The cockroaches learn to avoid these deposits and to seek new refuges in untreated places (Brett and Ross, 1986; Metzger, 1995).

6. Domiciliary cockroach infestation

Lee, Chong, and Yap (1993) surveyed on domiciliary cockroach infestation in four localities in Penang (Malaysia) using sticky traps. Six species were found such as *Nauphoeta cinerea*, *Neostylopyga rhombiofolia, Periplaneta americana*, *P. australasiae, P. brunnea,* and *Supella longipalpa.* After that Lee and Lee (2000) surveyed on domiciliary cockroach infestation again in six locations in Penang, using jar traps. Ten species were recorded such as *Blattella germanica, B. vaga, Na. cinerea*, *Ne. rhombiofolia, P. americana*, *P. australasiae, P. brunnea, Pycnoscelus surinamensis*, *Supella longipalpa*, and *Symploce pallens*. Both reports showed that *P. americana* was the dominant species that found in all locations, moreover, *B. germanica* found only in hotels and restaurants.

Lee, Lee, and Sim (2003) surveyed the population and infestation rates of domestic cockroaches in Busan and Seoul, Republic of Korea, using sticky traps. Three species of cockroaches were found such as *B. germanica, P. americana*, and *P. brunnea* and from three different types of house, apartment and villa. Moreover, the *B. germanica* was the dominant species in all residences.

Pai, Chen and Peng (2005) investigated the infestation of indoor cockroaches in Kaohsiung city, Taiwan. Two species of cockroaches, *P. americana* and *B. germanica*, were found. The highest density of both kinds of cockroaches found in kitchen.

Chaloryu *et al.* (1961) reported of cockroach survey in Thai navy ship, found 4 species: *Blatta orientalis, Blattella germanica, Parcoblatta pennsylvanica*, and *Periplaneta americana*. Most of them were found in food storage rooms and dining rooms.

Asahina and Hasegawa (1981) surveyed during daytime for the cockroaches in house of Chanthaburi province and found eight species: *B. germanica, B. lituricollis, Nauphoeta cinerea*, *Neostylopyga rhombiofolia, P. americana*, *P. australasiae, P. brunnea,* and *Pycnoscelis surinamensis.* Furthermore, both *B. germanica* and *P. americana* are commonly found everywhere in Chanthaburi Province.

Jungwiwattanaporn (1984) studied the species of domiciliary cockroaches in 5 provinces. Nine species of cockroaches were found: *B. germanica, B. lituricollis, Na. cinerea, Ne. rhombiofolia, P. americana*, *P. australasiae, P. brunnea, Py. indicus*, and *Supella longipalpa*. All of cockroach surveys were investigated in houses, libraries, museum, groceries, and rice-mill.

Benjapong, *et al.* (1997) surveyed of species and density of cockroach in 4 hospitals in central of Thailand. The results showed that 5 cockroach species including nymphal forms distribute with infested by *B. germanica, Na. cinerea, Ne. rhombiofolia, P. americana*, *and P. brunnea.*

Chanbang (1997) investigated the cockroach species using plastic box traps in Chatuchak and Din Daeng District, Bangkok. The four species of cockroaches were found such as *B. germanica*, *Blaberus* sp.*, P. americana*, and *P. brunnea*.

Tawatsin *et al.* (2001) surveyed of species of cockroaches in 14 provinces of Thailand, using sticky traps. The results showed that 10 cockroach species caught from 14 provinces such as *B. germanica, B. lituricollis, Na. cinerea, Ne. rhombiofolia, P. americana*, *P. australasiae, P. brunnea, P. fuliginosa, Py. surinamensis*, and *S. longipalpa* belonging to six genera. Moreover, *P. americana* and *P. brunnea* were the most abundant cockroach species in urban Thailand, furthermore, the kitchen was the major habitat.

Sriwichai (2001) investigated the distribution of indoor cockroaches in urban areas using commercial sticky traps in single houses, townhouses, apartments, office units and grocery stores. Eight species of cockroaches were found such as *B. germanica, B. lituricollis, Ne. rhombiofolia, P. americana*, *P. brunnea, P. australasiae, Py. surinamensis,* and *S. longipalpa*. Moreover, *P. americana* and *S. longipalpa* had infested all kinds of dwellings, while *B. germanica* was found in grocery shops.

Nacapunchai *et al*. (2001) investigated the infestation of indoor cockroaches in some urban and rural dwellings from living rooms, bedrooms, and office unit of Thailand, using sticky traps. The dominant species of urban indoor cockroaches were *P. americana* and *S. longipalpa* which the nymphal stage was the highest density. While the dominant species of rural dwellings was *P. americana* and the highest density was found in kitchen.

Damsuwon (2003) investigated the infestation of indoor cockroaches in Nonthaburi Province, using sticky traps. Eight species of cockroaches were found such as *B. germanica, B. lituricollis, Ne. rhombiofolia, P. americana*, *P. brunnea,* *P. australasiae, Py. surinamensis,* and *S. longipalpa*. Environmental dwelling and inhabitant behavior factors, such as type of dwelling, age of dwelling, damaged and deteriorated conditions, material types of dwelling, rubbish and waste, cooking behavior and cleaning behavior had statistically significant relationships between the prevalence and density of cockroaches.

7. Medical and public health importance

Cockroaches are pests in the human environment. The success of them in peridomistic and domestic habitat is based largely on their biology and habitats. They can infest in human structures according their adapted behavior patterns to human household environments, which the appropriated condition of humidity, temperature, foods and narrow crevices for harborage are provide in human dwellings. Because cockroaches eat a wide range of food and commonly feed on decaying food, crumbs, or scraps, it is believed that they spread a number of diseases to humans (Benson and Zungoli, 1997) and they can also caused allergies (Ross and Mullins, 1995; Sarinho *et al*., 2004).

Although the German cockroach does not sting or bite, but it adversely affect human health in several ways such as it may transmit pathogenic agents mechanically on its body part (Fotedar, Shriniwas, and Verma, 1991; Cloarec, Rivault, and Le Guyader, 1992; Rivault, Cloarec, and Le Guyader, 1993; Brenner, 1995). The German cockroach was reported as carried virus, bacteria, fungi, protozoans and helminthes. The German cockroach is considered the species of carrier of many diseases that can be transmitted in variety of ways. Human pathogenic organisms which isolated from the cockroaches is showed in Table 2.1.

The cockroach excrement, cast skins, salivary secretions, and produce secretions contained a number of allergens to the sensitive people who exhibit allergic responses (Helm *et al*., 1990). Allergy to cockroaches occurred when the immunological system becomes sensitized to harmless proteins (Brenner, 1995). Cockroaches also had been reported that to sensitize in allergic and asthmic Thai patients in ranges 44-61% and they are the second most household important allergens by following house dust mite (Pumhirun, Towiwat, and Mahakit, 1997).

Moreover, the entomophobia or fear of insects is common. Many people find cockroaches disgusting. There is an association in the mind of many people between the presence of cockroaches and a dirty environment. As a result, cockroach infestations in households can serve as psychological stressors to the residents (Lee, 1997a). In addition, the German cockroaches produced odorous substances from their special glands. When feeding they also dropped their feces and secretion of the scent glands which located between the fifth and sixth abdominal terga segment. Thus, the food which contaminated by cockroach feces always presented the bad odor (Baumholthz *et al.*, 1997).

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches.

Organisms		Diseases/ Infections	References
Virus	Hepatitis virus	jaundice	Brenner, 1995
	Poliomyelitis virus	polio	Brenner, 1995
Bacteria	*Bacillus cereus*	food poisoning, gastroenteritis and meningitis	Oothuman *et al.*, 1989; Vythilingam *et al.*, 1997; Tachbele *et al.*, 2006
	B. subtilis	conjunctivitis	Brenner, 1995
	Campylobacter jejuni	enteritis	Brenner, 1995
	Citrobacter sp.	opportunistic pathogen	Oothuman *et al.*, 1989; Rivault *et al.*, 1993; Vythilingam *et al.*, 1997
	Enterobacter aerogenes	bacteremia	Oothuman *et al.*, 1989; Rivault *et al.*, 1993; Brenner, 1995
	E. agglomerans	opportunistic pathogen	Oothuman *et al.*, 1989; Rivault *et al.*, 1993; Vythilingam *et al.*, 1997
	E. cloacae	opportunistic pathogen	Cloarec *et al.*, 1992; Rivault *et al.*, 1993; Vythilingam *et al.*, 1997

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches (cont.).

Organisms		Diseases/ Infections	References
Bacteria	*Escherichia coli*	diarrhea, urogenital and intestinal infection	Oothuman *et al.*, 1989; Rivault *et al.*, 1993; Brenner, 1995; Vythilingam *et al.*, 1997; Tachbele *et al.*, 2006
	Klebsiella oxytoca	lower respiratory tract infection	Oothuman *et al.*, 1989; Cloarec *et al.*, 1992; Rivault *et al.*, 1993; Vythilingam *et al.*, 1997
	K. ozaenae	chronic atrophic rhinitis	Vythilingam *et al.*, 1997
	K. pneumoniae	pneumonia and upper respiratory tract infection	Cloarec *et al.*, 1992; Rivault *et al.*, 1993; Brenner, 1995; Vythilingam *et al.*, 1997
	K. rhinocleromatis	rhinoscleroma	Vythilingam *et al.*, 1997
	Mycobacterium leprae	leprosy	Brenner, 1995
	Proteus sp.	wound infection	Agbodaze and Owusu, 1989; Oothuman *et al.*, 1989; Brenner, 1995; Vythilingam *et al.*, 1997

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches (cont.).

Organisms		Diseases/ Infections	References
Bacteria	*Pseudomonas aeruginosa*,	respiratory tract infection	Agbodaze and Owusu, 1989; Oothuman *et al.*, 1989; Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995
	Salmonella spp.	enteric fever and food poisoning	Oothuman *et al.*, 1989; Brenner, 1995; Tachbele *et al.*, 2006
	S. arizonae	salmonella gastroenteritis	Agbodaze and Owusu, 1989; Tachbele *et al.*, 2006
	Serratia marcescens	upper respiratory tract inflammation and food poisoning	Oothuman *et al.*, 1989; Rivault *et al.*, 1993; Brenner, 1995
	Shigella boydii	shigellosis	Oothuman *et al.*, 1989; Tachbele *et al.*, 2006
	S. dysenteriae	dysentery	Oothuman *et al.*, 1989; Agbodaze and Owusu, 1989; Brenner, 1995; Tachbele *et al.*, 2006

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches (cont.).

Organisms		**Diseases/ Infections**	**References**
Bacteria	*Staphylococcus aureus*	food poisoning, enterocolitis, skin infection, bacteremia and toxic shock syndrome	Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995; Tachbele *et al.*, 2006
	Streptococcus faecalis (or *Enterococcus faecalis*)	nosocomial infection	Fotedar *et al*., 1991; Brenner, 1995
	S. pyogenes	streptococcal sore throat, rheumatic fever, Impetigo, erysipelas and wound infection	Brenner, 1995
	Vibrio spp.	diarrhea	Brenner, 1995
Proto-zoans	*Entamoeba histolytica*	amoebic dysentery	Fotedar *et al.*, 1991; Brenner, 1995
	Giardia sp.	giardiasis	Brenner, 1995

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches (cont.).

Organisms		Diseases/ Infections	References
Fungi	*Alternaria* spp.	allergen skin and lung infection	Fotedar *et al.*, 1991; Brenner, 1995
	Aspergillus flavus	aspergillosis adn mycotic granuloma	Fotedar *et al.*, 1991; Brenner, 1995
	A. niger	aspergillosis and mycotic granuloma	Fotedar *et al.*, 1991; Brenner, 1995
	Candida spp.	vaginitis, candidiasis and endocarditis	Fotedar *et al.*, 1991; Brenner, 1995
	Cephalosporium acremonium	mycetoma	Brenner, 1995
	Cladosporium sp.	dermatomycosis	Brenner, 1995
	Fusarium sp.	otomycosis	Brenner, 1995
	Geotrichum candidum	skin and gastrointestinal infection	Brenner, 1995
	Mucor spp.	lung infection and otomycosis	Fotedar *et al.*, 1991; Brenner, 1995

Table 2.1 Summary of organisms that are pathogenic to human that have been isolated from the cockroaches (cont.).

Organisms		**Diseases/ Infections**	**References**
Fungi	*Rhizopus* spp.	lung infection otomycosis	Fotedar *et al.*, 1991; Brenner, 1995
	Rhodotorula rubra	lung infection	Brenner, 1995
	Trichoderma viride	rare alimentary toxic aleukia	Brenner, 1995
	Trichosporon cutaneum	white hair-nodule disease	Brenner, 1995
Helminthes	*Acaris lumbricoides*	roundworm infection and ascariasis	Brenner, 1995
	Ancylostoma duodenale	hookworm infection and anemia	Brenner, 1995
	Enterobius vermicularis	pinworm infection	Brenner, 1995
	Gnathostoma spinigerum	gnathostomiasis and gnathostomosis	Thanasomboonpun, 1992
	Hymenolopsis sp.	enteritis	Brenner, 1995
	Necator americanus	hookworm infection and anemia	Brenner, 1995
	Trichuris trichuria	whipworm infection	Brenner, 1995

8. Control measures

Effective control is easier in temperate climates (where cockroach populations cannot survive outdoors in winter) than in humid and warm areas. The key to control is cleanliness, which may be difficult in houses where there are children and domestic animals. In isolated homes, control is easier to achieve than in apartments where cockroaches may have easy access from adjacent quarters. Reinfestation occurs from outdoors in warm areas, or along heating ducts and water pipes in apartments, or from groceries or luggage brought from cockroach-infested areas. Cockroaches may even sometimes be found in very clean houses, but are unlikely to establish colonies. The presence of several sizes of nymphs and cast of oothecae is an indication of a well-established colony. Infestations can be detected by searching behind skirting-boards, boxes, furniture, and other common hiding places. At night, cockroaches are easily detected using light. Moreover, for heavy infestations of cockroaches can be dealt with by chemical control measures, followed by environmental management to deprive the insects of food and shelter. Low numbers can be effectively controlled by baits or traps (Gold, 1995; Koehler, Patterson and Owens, 1995; Rozendaal, 1997).

<u>Environmental control</u>

There is a definite relationship between the availability of food and water resources and the German cockroach population growth. However, water is more important than food and harborage for the German cockroaches (Durbin and Cochran, 1985; Silverman 1986).

Therefore, the prevention is the key to successful cockroach control. Prevention can minimize cockroach invasion of buildings. It is much easier and usually less costly to keep cockroaches on the outside than it is to get rid of them once inside. For example, food should be stored in tightly covered containers in screened cabinets or refrigerators. All areas have to be kept clean so that no fragments of food or organic matter remain. Rubbish bins should be securely covered and emptied frequently, preferably daily. Basements and areas underneath buildings should be kept dry and free of accessible food and water. Moreover, the accessibly reduction is important such as groceries, laundry, dirty clothing, egg crates and furniture should be checked before being taken into a building. In some instances, accessibility to buildings can be reduced by closing gaps in floors and door frames. Openings for drain water and sewer pipes, drinking-water and electricity cables should also be closed (Rozendaal, 1997).

Chemical control

Cockroaches are difficult to control with insecticides for several reasons, one reason is that they may become resistant to commonly used compounds. Moreover, many insecticides are only repellent to them and are therefore avoided (Wooster and Ross, 1989). Chemical control gives only temporary relief and it should be accompanied by environmental sanitation and house improvement (Schal, 1988). Insecticides are applied to the resting and hiding places as residual sprays and insecticidal dusts. Such applications are effective for periods ranging from several days to months, depending on the insecticide and the substrate on which it is deposited. Insecticides can also be combined with attractants as toxic baits (Rozendaal, 1997).

(1) Dusts

Dry powder formulations are made by mixing insecticide powder with talcum or another inert carrier powder. They are most useful for the treatment of hollow walls, false ceilings and other cockroach hiding places that cannot easily be reached. The powders can be blown into spaces with a hand-operated puff-duster or a plunger-type duster, or even applied with a spoon. Long, slender extension tubes can be attached to some types of duster to put the dust deep into hiding places. The dust disperses well and may penetrate deep into cracks and crevices. Heavy dust deposits may repel or drive away cockroaches and cause them to move to untreated areas or less accessible places. Dusts should not be applied to wet surfaces as this reduces their effectiveness. When used together with residual sprays, dusts should be applied only once the sprayed surfaces are dry (Ebeling, 1995).

(2) Aerosols

Insecticidal aerosols are fine sprays of very small (0.1-50 mm) droplets of insecticide. Aerosols are not suitable for residual treatment but they can be used for space spraying because the droplets remain in the air for some time, killing insects by contact. Aerosol spray cans containing a residual insecticide with a knock-down insecticide, for example; propoxur and pyrethroid, are suitable for cockroach control and are widely available. Aerosols can penetrate into small crevices and other enclosed, inaccessible cockroach hiding places. They usually contain pyrethrins, pyrethroids or another irritant to drive cockroaches out of their hiding places so as to shorten the time of kill. Aerosol application can cause a quick reduction in cockroach numbers but, to obtain longer-lasting control, follow-up treatment with a residual spray may be necessary. Cities sometimes control cockroaches on a large scale with fogs produced by thermo-fogging machines (Schal, 1988).

(3) Smokes

Smokes are clouds of insecticide particles produced by heat. The particle size (0.001-0.1 mm) is smaller than in aerosols. Smokes penetrate deep into hiding places and are particularly useful in basements of buildings and sewer and drainage systems (Ebeling, 1995).

(4) Baits and traps

Baits have been used for many years in cockroach control and are still employed in certain situations, such as offices and laboratories, particularly if there is resistance to some of the insecticides in use. Many commercially available products work on the principle of attracting cockroaches to a specific point and then trapping or killing them there. Some substances used as attractants are various food items, pheromones and other attractive chemicals. The trapping element may be a mechanical trap or a sticky material. A simple jar trap can be constructed from an empty jar, petroleum jelly and some food: the cockroaches are attracted to the jar by bread, raisins or other food placed at the bottom, and a thin layer of petroleum jelly on the inside rim prevents the insects from escaping. Toxic baits are used without a trapping device. They consist of a mixture of attractive food material and an insecticide. Several types of bait are commercially available as pellets or pastes. Pellets are usually dispensed in small containers or scattered in concealed areas. Pastes can also be dispensed in small containers. Some of the newer formulations are self-drying and can be applied directly to surfaces. In some countries, dry baits are available in sealed traps which are safe to use where children or pets are present. Some food materials which may be used in baits are peanut meal, dog food and maltose. Baits and traps are easy to use and should be placed at sites frequented by cockroaches. They are most effective in situations where there is little or no food to compete with the bait, as is the case in offices. The maintenance of environmental hygiene is especially important when baits are used alone. In heavily infested areas, baits need to be replaced frequently (Owens, 1995).

Repellents

There is growing interest in the use of repellents in the control of cockroaches. They may be of special interest for application to hiding places in shipping containers, and in cases and boxes containing drinks, food and other materials. Keeping cockroaches away from such places prevents the distribution or movement of the insects from one locality to another. Repellents can also be used in kitchen cupboards, food and beverage vending machines, and so on. Several essential oils, such as mint oil, spearmint oil and eucalyptus oil are known to repel cockroaches, but the best results are obtained with synthetic products that are easier to standardize (Rozendaal, 1997).

Insecticides commonly employed in the control of cockroaches is showed in Table 2.2.

Table 2.2 Insecticides commonly used in the control of cockroaches (Rozendaal, 1997).

Insecticide	Chemical type [a]	Formulation	Concentration		Safety classification by WHO [b]
			g/l or g/kg	%	
Alphacypermethrin	PY	spray	0.15	0.015	MH
Bendiocarb	C	spray	2.4-4.8	0.24 - 0.48	MH
		dust	10	1.0	
		aerosol	7.5	0.75	
Betacyfluthrin	PY	spray	-	12.5	MH
Chlorpyrifos	OP	spray	5	0.5	MH
Cyfluthrin	PY	spray	-	5-10	MH
Cypermethrin	PY	spray	-	5-10	MH
Cyphenothrin	PY	spray	1.25-2.5	0.125-0.25	SH
		aerosol	1-3	0.1-0.3	
Deltamethrin	PY	spray	0.025	0.0025	MH
		dust	0.5	0.05	
Diazinon	OP	spray	5	0.5	MH
		dust	20	2.0	
Dichlorvos	OP	spray	5	0.5	HH
		bait	19	1.9	
Dioxacarb	C	spray	5-10	0.5-1.0	MH
Fenitrothion	OP	bait	250	25	MH
		spray	5-10	0.5-1.0	
		aerosol	7.5	0.75	

Table 2.2 Insecticides commonly used in the control of cockroaches (cont.).

Insecticide	Chemical type [a]	Formulation	Concentration		Safety classification by WHO [b]
			g/l or g/kg	%	
Flufenoxuron	IGR	bait	0.01	0.001	SH
Hydramethylnon	ETI	bait	-	1-2	SH
Jodfenphos	OP	spray	10	1.0	UH
Malathion	OP	spray	30	3.0	SH
		dust	50	5.0	
Permethrin	PY	spray	1.25-2.5	0.125-0.25	MH
		dust	5	0.5	
Pirimiphos methyl	OP	spray	25	2.5	SH
		dust	20	2.0	
Propetamphos [c]	OP	spray	5-10	0.5-1.0	HH
		dust	20	2.0	
		aerosol	20	2.0	
Propoxur	C	spray	10	1.0	MH
		bait	20	2.0	

[a] C = carbamate; OP = organophosphate; PY = synthetic pyrethroid; IGR = insect growth regulator, ETI = electron transport inhibitor.

[b] Classes: HH = highly hazardous; MH = moderately hazardous; SH = slightly hazardous, UH = unlikely to present acute hazard in normal use.

[c] If applied by non-commercial operators, it should be supplied, for safety reasons, in a diluted form not exceeding 50g of active ingredient per litre.

9. Insecticidal resistance

History of insecticide resistance in the German cockroach

Currently, a control failure due to insecticide resistance in the German cockroach is a common problem to the pest control industry (Lee, Yap, and Chong, 1996a; Lee, 1997b). The first case of German cockroach resistance to chlordane was detected in 1952, where a strain was found to be resistant to chlordane at >100-fold by the glass jar method (Heal, Nash, and Williams, 1953 cited in Lee and Lee, 2004). Since then, insecticide resistance development in the German cockroach has been showed to follow patterns of insecticide usage. It started with organochlorine resistance (DDT, dieldrin and lindane), followed by organophosphate and carbamate resistance in the 1960s (Webb, 1961; Collins, 1973). Pyrethroid resistance was reported in mid to late 1980s (Scott *et al*., 1986; Cochran, 1987, 1989) when cypermethrin no longer provided effective control against German cockroaches in apartments (Schal, 1988). A summary of selected cases of insecticide resistance in the German cockroach documented is presented in Table 2.3.

Table 2.3 Selected cases of insecticides resistance in the German cockroach.

Insecticide[1]	Assessment[2]	Resistance ratio	Location	Reference
OC				
chlordane	SC (time)	> 100	Texas, U.S.A.	Heal *et al*., 1953
	SC (time)	1.2-14.4	France and Germany	Webb, 1961
	T (dose)	4.3	Indiana, U.S.A.	Scharf *et al*., 1996
dieldrin	SC (time)	LT50=> 48 h	England	Gradidge, 1960
DDT	SC (time)	5-6	Texas, U.S.A.	Heal *et al*., 1953
	SC (time)	4-12	France and Germany	Webb, 1961
	T (dose)	> 6	Malaysia	Lee *et al*., 1996a
lindane	SC (time)	10-12	Texas, U.S.A.	Heal *et al*., 1953
OP				
chlorpyrifos	SC (time)	1.3	New Jersey, U.S.A.	Schal, 1988
	T (dose)	4-25	California, U.S.A.	Rust and Reierson, 1991
	T (dose)	2-8	Malaysia	Lee *et al*., 1996a
	T (dose)	0.05-0.51	Malaysia	Lee and Lee, 2002
diazinon	SC (dose)	3-8	Texas, U.S.A.	Grayson, 1965
	SC (time)	3.7	Baltimore, U.S.A.	Nelson and Wood, 1982

Table 2.3 Selected cases of insecticides resistance in the German cockroach (cont).

Insecticide[1]	Assessment[2]	Resistance ratio	Location	Reference
OP				
fenthion	LC(dose)	3-8	Texas, U.S.A.	Grayson, 1965
malathion	SC (time)	1.5-3.0	France and Germany	Webb, 1961
	SC (dose)	2.2-12.8	Texas, U.S.A.	Grayson, 1965
	SC (time)	6.5	Baltimore, U.S.A.	Nelson and Wood, 1982
	SC (time)	1.9-41.1	Malaysia	Lee, Tien and Omar, 1997a
C				
bendiocarb	SC (time)	94	Baltimore, U.S.A.	Nelson and Wood, 1982
	SC (time)	> 100	New Jersey, U.S.A.	Schal, 1988
	SC (time)	> 60	U.S.A.	Cochran, 1989
	T (dose)	10.6	Indiana, U.S.A.	Scharf *et al.*, 1996
	T (dose)	> 63	Malaysia	Lee *et al.*, 1996a
	SC (time)	1.6-4.8	Malaysia	Lee *et al.*,1997a
	SC (time)	5.1-13.3	Malaysia	Lee, Yap and Chong, 1997b

Table 2.3 Selected cases of insecticides resistance in the German cockroach (cont).

Insecticide[1]	Assessment[2]	Resistance ratio	Location	Reference
C				
propoxur	SC (time)	13.3	Baltimore, U.S.A.	Nelson and Wood, 1982
	SC (time)	> 100	New Jersey, U.S.A.	Schal, 1988
	T (dose)	3-90	Malaysia	Lee *et al*., 1996a
	T (dose)	4	Indiana, U.S.A.	Scharf *et al*., 1996
	SC (time)	5.4-11.5	Malaysia	Lee *et al*., 1997b
	T (dose)	0.01-0.78	Malaysia	Lee and Lee, 2002
PY				
allethrin	SC (time)	> 100	U.S.A.	Cochran, 1989
cypermethrin	SC (time)	4.5	New Jersey, U.S.A.	Schal, 1988
	T (dose)	103.6	Florida, U.S.A.	Atkinson *et al*., 1991
	T (dose)	1-22	Malaysia	Lee *et al*., 1996a; Lee and Lee, 1998
	T (dose)	3.5-4.2	Indiana, U.S.A.	Scharf *et al*., 1996
	SC (time)	1.9-3.2	Malaysia	Lee *et al*., 1997b
cyfluthrin	T (dose)	87.5	Florida, U.S.A.	Atkinson *et al*., 1991
	T (dose)	3	Indiana, U.S.A.	Scharf *et al*., 1996

Table 2.3 Selected cases of insecticides resistance in the German cockroach (cont).

Insecticide[1]	Assessment[2]	Resistance ratio	Location	Reference
PY				
cyhalothrin	T (dose)	40.6	Florida, U.S.A.	Atkinson *et al*., 1991
deltamethrin	T (dose)	6-24	Malaysia	Lee *et al*., 1996a
esfenvalerate	T (dose)	29.4	Florida, U.S.A.	Atkinson *et al*., 1991
fenvalerate	SC (time)	> 60	U.S.A.	Cochran, 1989
	T (dose)	97.7	Florida, U.S.A.	Atkinson *et al*., 1991
fluvalinate	T (dose)	337.2	Florida, U.S.A.	Atkinson *et al*., 1991
permithin	SC (time)	> 100	U.S.A.	Cochran, 1989
	T (dose)	45.1	Florida, U.S.A.	Atkinson *et al*., 1991
	T (dose)	1-15	Malaysia	Lee *et al*., 1996a
	T (dose)	2.2	Indiana, U.S.A.	Scharf *et al*., 1996
phenothrin	SC (time)	> 80	U.S.A.	Cochran, 1989
	T (dose)	13-52	Malaysia	Lee *et al*., 1996a
pyrethrin	SC (time)	> 80	U.S.A.	Cochran, 1989
resmethrin	T (dose)	102.6	Florida, U.S.A.	Atkinson *et al*., 1991
sumithrin	T (dose)	113.8	Florida, U.S.A.	Atkinson *et al*., 1991
tralomethrin	T (dose)	72.2	Florida, U.S.A.	Atkinson *et al*., 1991

[1] C = carbamate; OC = organochlorine; OP = organophosphate; PY = synthetic pyrethroid.

[2] SC = surface contact; T = topical application.

Resistance mechanisms in the German cockroach

The mechanism of insecticide resistance in insects can be divided into physiological and behavioral resistance. Physiological resistance occurs when biochemical/ physiological-related mechanisms are involved in reducing the efficacy of an insecticide. On the other hand, the ability of insects to avoid lethal insecticide exposures due to the nature of or changes in their behavior is classified as behavioral resistance (Lee, 1997b). There are three general resistance mechanisms which can be classified as physiological resistance such as reduced cuticular penetration, increased metabolic detoxification, and target site insensitivity (Siegfried and Scott, 1992).

(1) Reduced cuticular penetration

Reduced cuticular penetration presented low level of resistance in insects, generally less than three-fold (Scott, 1990). For example, the penetration of [^{14}C] - propoxur into the body was reduced when the chemical was applied topically in a propoxur-resistant strain of the German cockroach (Siegfried and Scott, 1991). Moreover, the penetration rate of [^{14}C] - permethrin was slower than the susceptible strain of the German cockroach (Bull and Patterson, 1993).

(2) Increased metabolic detoxification

Increased metabolic detoxification is the most common resistance mechanism in insects (Scott, 1990). The conclusive ways to study the increased metabolic detoxification were through *in vivo* and *in vitro* metabolism studies using radio-labeled insecticides (Scott, 1990; Siegfried and Scott, 1991) and performing enzyme assays (Scott, 1990). Three groups of metabolic enzymes which are involved in detoxification insecticides such as monooxygenases, esterases, and glutathione S-transferase (GST).

(3) Target site insensitivity

Reduced sensitivity to insecticides due to modification of target site can be divided into: (1) altered acetylcholinesterase (AChE), which confers resistance to organophosphates and carbamates. (2) knockdown resistance (*kdr*-type), where insects become insensitive to organochlorines and pyrethroids (Siegfried and Scott, 1991).

From the literature reviews, the German cockroach, *B. germanica,* was the common species that infestation in urban areas in Thailand. Prior to this study, little documentation existed as to the current population dynamics, feeding behavior, bacterial harborage and the residues insecticide of the German cockroaches in Bangkok markets, public place where many foods are stayed. This study was conducted to evaluate better control strategies and an understanding of the cockroach population and their behavior in market areas of Bangkok.

CHAPTER 3

FOOD PREFERENCE AND FEEDING BEHAVIOR OF THE GERMAN COCKROACH, *Blattella germanica* (Linnaeus)

Introduction and Objectives

For most insects, nutrition affects on survivorships adults and their reproductive outputs. Generally, female fecundity depends on the ingestion of proteins which is necessary for egg development whereas male fertility does not highly depend on proteins. Moreover, insects generally are unable to convert lipids to monosaccharides by themselves, and carbohydrates are one of nutrition that both sexes of insects have used as primary energy sources (Carrel and Tanner, 2002). Nutritional factors reportedly had profound short-term and long-term effects on the development and reproduction of insects. Even though the nutritional requirements of most insects are relatively similar, the optimal sources, types, and proportions of nutrients widely vary among species and reproductive stages (Cooper and Schal, 1992).

Recent studies revealed that insects significantly show sex-specific differences in feeding behaviors and adult nutritional requirements. For example, young female tephritid fruit flies, *Bactrocera dorsalis*, (Diptera: Tephritidae) are attracted to the odors of protein food baits more than the odors of host fruit (Cornelius *et al.*, 2000). Female peacock butterflies, *Inachis io* Linn., and Adonis blue butterflies, *Lysandra bellagrus* Linn., preferred amino acid-rich floral nectars, whereas males preferred sugar-rich floral nectars (Erhardt and Rusterholz, 1998; Rusterholz and Erhardt, 2000). Furthermore, male Madagascar hissing cockroaches, *Gromphadorhina portentosa* (Dictyoptera: Blaberidae) preferred carbohydrate-rich foods whereas females preferred protein-rich foods (Carrel and Tanner, 2002).

Entomologists have been interested in studying about cockroach food preference and feeding behavior because cockroaches are one of the most important household pests. The range of food substances that they utilize is greater than any other insects. The German cockroach, *Blattella germanica* Linn., has been used as a model organism for the study of insect neurobiology digestive and physiology (Jones and Raubenheimer, 2001). However, the documentation of food preference and feeding behavior of the German cockroach has been limited.

The objective of this research was to study the attractiveness of various foods to the male and female German cockroaches, including the study of their feeding behavior. Laboratory olfactometer assays and behavior observation were used in this experiment. In addition, this research would determine the attractiveness of some non-chemical baits for trapping the German cockroach in the field.

Materials and Methods

1. Insect mass rearing

Colonies of the German cockroach were established from field-collected adults. All colonies were maintained in the laboratory under an approximately 12: 12-hours light: dark natural photoperiod at about 30° C and 50-60% relative humidity. Wide-mouth glass jars, 25 cm in diameter and 35 cm in height, were used as containers. The upper two-fifths of the inner surface of the jar was coated with petroleum jelly to ensure the cockroaches were unable to climb out, and rolled cardboards were placed for their shelter. Cotton net cover, secured with lid ring, was placed on top of each jar to prevent escaping of cockroaches or entry of other animals (Figure 3.1). The cockroaches were fed with cat food (Me-O®, S.W.T. Company, Samutprakarn, Thailand) once a week and the water was monitored twice a week. The newly molted adults were separated by sex and placed in separate containers.

Figure 3.1 Cockroaches rearing in wide-mouth glass jars (A) with rolled cardboards as their shelters (B), dry cat food and tissue paper' s soaked with water.

2. Test for food preference

Adult male and female German cockroaches (N=300) were used in this experiment. Modified eight-chamber-olfactometers (Figure 3.2) were used for this food preference experiment. Each kind of food (0.2 g) was placed randomly in each chamber of the olfactometer. Foods consisted of carbohydrate–rich foods such as bread, sugar, banana, *Musa sapientum* Linn., and potato, *Solanum tuberosum* Linn. and protein-rich foods such as peanut, cheese, pork, and cat food. To evaluate the effect of hunger on food choice, the cockroaches were starved for 48 hours before conducting the test. Fifteen of each sex of the starving cockroaches were placed at the center of an olfactometers. All of the testing olfactometers were placed on a laboratory bench under red light illumination in a room with fully covered windows. The cockroaches could not detect red light so that everything surrounding them appeared black (Carrel and Tanner, 2002). The cockroaches were checked at 1, 4, 16, 24, 28, 40, and 48 hours and the experiment was replicated 10 times (each replication used a new group of insects).

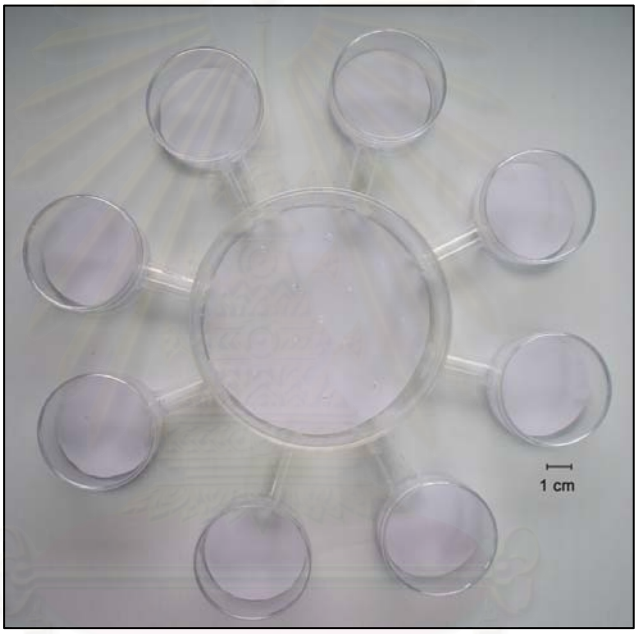

Figure 3.2 A modified eight-chamber-olfactometer used for the food preference experiment. Cockroaches were placed in the center arena and allowed access to food choices in eight peripheral chambers (modified from Reierson, 1995).

3. Study for feeding behavior

Adult male and female cockroaches (N=100, for each sex) were used in this feeding behavior experiment. Twenty of each sex of the German cockroach were placed in an arena (15 x 30 x 18 cm) with a rolled cardboard as their shelter. The cockroaches were fed by cat food and water. The behavior of insects were observed for 24 hours and replicated 5 times (each replication used a new group of insects). For the nighttime, their feeding behavior was observed under red light illumination (Figure 3.3).

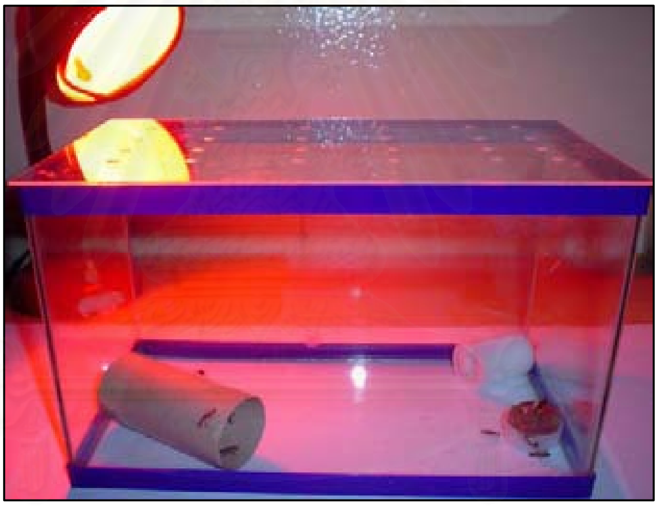

Figure 3.3 Feeding behavior of the German cockroach was observed in an arena under red light illumination.

Data analysis

The amount of food eaten (in the test for food preference) was recorded and calculated to the proportion eaten (in this experiment, the proportion eaten = X/ 0.2 g). In addition, the Rodgers's index (Krebs, 1999) for indicating the food preference was used as following:

The Rodgers's index (Ri)

$$Ri = \frac{Ai}{\max (Ai)}$$

When:

Ri = Rodgers's index of preference for food item i

Ai = Area under the cumulative proportion eaten curve for food item i

max Ai = the largest value of the Ai

The homogeneities of variances were tested and the significant differences between sexes and among the food items were analyzed by *Mann-Whitney U*-test.

Results

1. Food preference of the German cockroach

The male and female German cockroaches showed the preferable difference on food choices. The curves of means of proportion eaten by the male and female German cockroaches on the eight food items showed in Figure 3.4.

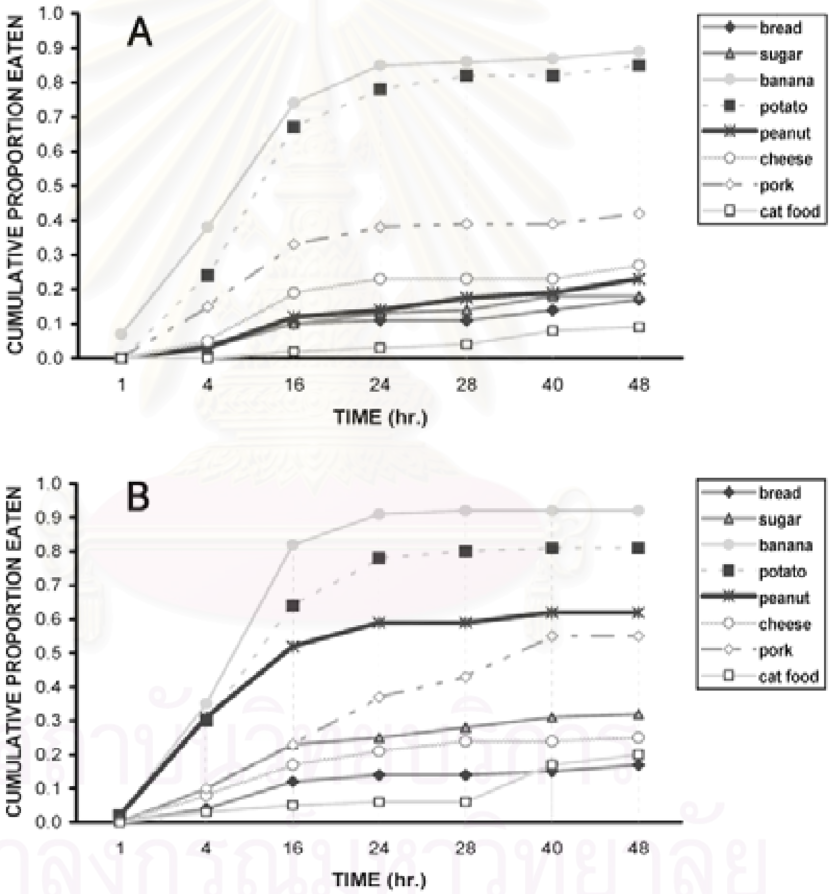

Figure 3.4 Curves of means of proportion eaten by the male (A) and the female (B) German cockroaches of the eight food items. Each point on a curve represents a mean of proportion of food eaten in each time of observation.

The homogeneity tests showed the variances to differ significantly at $p < 0.05$. Using *Mann-Whitney U*-test, the result showed that the male cockroaches preferred banana following by potato, pork, cheese, peanut, sugar, bread, and cat food, respectively. They preferred banana and potato significantly at $p < 0.05$ (Table 3.1; Figure 3.5 A). The female cockroaches preferred banana following by potato, peanut, pork, sugar, cheese, bread, and cat food, respectively. They preferred only banana significantly at $p < 0.05$ (Table 3.2; Figure 3.5 B). Additionally, the males significantly preferred potato more than the females. In contrast, the female cockroaches significantly preferred peanut, sugar, and cat food more than the males at $p < 0.05$ (Table 3.3; Figure 3.6); and also evidently preferred peanut and sugar more than the males at $p < 0.01$.

Table 3.1 Mean [1] (± SE) number of the Rodgers's index by the male German cockroach on the eight food items. In each replicate, 15 male cockroaches were observed (for 10 replicates).

Food items	**Rodgers's index (Ri)**
Bread	0.16 ± 0.06 [ab]
Sugar	0.17 ± 0.04 [b]
Banana	0.97 ± 0.02 [c]
Potato	0.93 ± 0.02 [c]
Peanut	0.20 ± 0.05 [b]
Cheese	0.26 ± 0.07 [b]
Pork	0.45 ± 0.10 [b]
Cat food	0.06 ± 0.01 [a]

[1] Means with the different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

Table 3.2 Mean [1] (± SE) number of the Rodgers's index by the female German cockroach on the eight food items. In each replicate, 15 female cockroaches were observed (for 10 replicates).

Food items	**Rodgers's index (Ri)**
Bread	0.18 ± 0.03 [a]
Sugar	0.32 ± 0.03 [b]
Banana	0.93 ± 0.05 [c]
Potato	0.80 ± 0.04 [d]
Peanut	0.63 ± 0.08 [d]
Cheese	0.23 ± 0.05 [ab]
Pork	0.42 ± 0.07 [b]
Cat food	0.10 ± 0.02 [a]

[1] Means with the different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

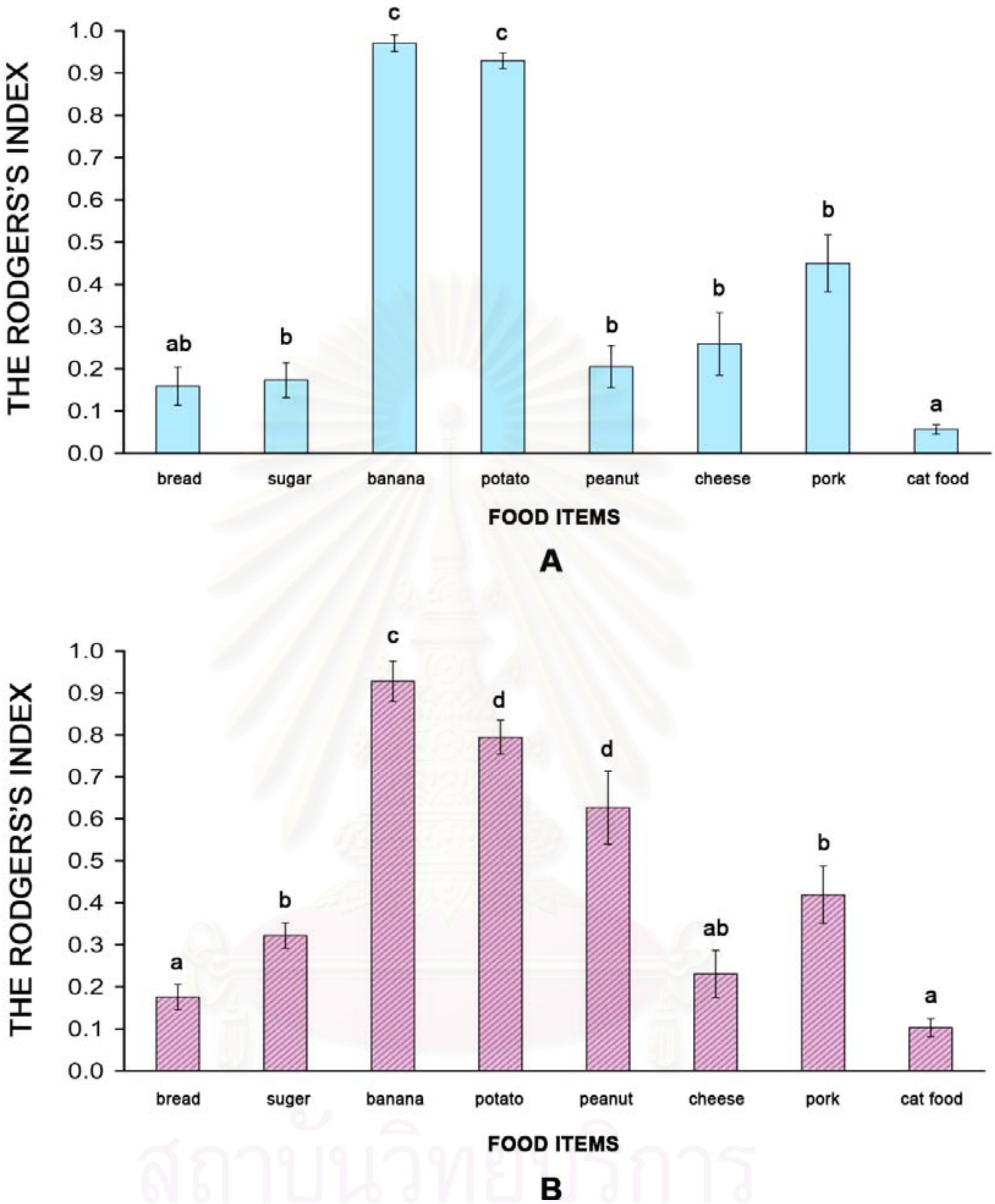

Figure 3.5 Means (± SE) of the Rodgers's indices of the male (A) and the female (B) German cockroaches among the eight food items. These indices indicate the food preference in cockroaches (rang 0-1, 0 = avoid and 1 = prefer). The bar graphs with different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

Table 3.3 Mean [1] (± SE) number of the Rodgers's indices comparing between the male and the female German cockroaches on the eight food items. In each replicate, 15 male and 15 female cockroaches were observed (for 10 replicates).

Food items	The Rodgers's indices (Ri)	
	the male	the female
Bread	0.16 ± 0.06 [a]	0.18 ± 0.03 [a]
Sugar	0.17 ± 0.04 [a]	0.32 ± 0.03 [b]
Banana	0.97 ± 0.02 [a]	0.93 ± 0.05 [a]
Potato	0.93 ± 0.02 [a]	0.80 ± 0.04 [b]
Peanut	0.20 ± 0.05 [a]	0.63 ± 0.08 [b]
Cheese	0.26 ± 0.07 [a]	0.23 ± 0.05 [a]
Pork	0.45 ± 0.10 [a]	0.42 ± 0.07 [a]
Cat food	0.06 ± 0.01 [a]	0.10 ± 0.02 [b]

[1] Means in the same row with the different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

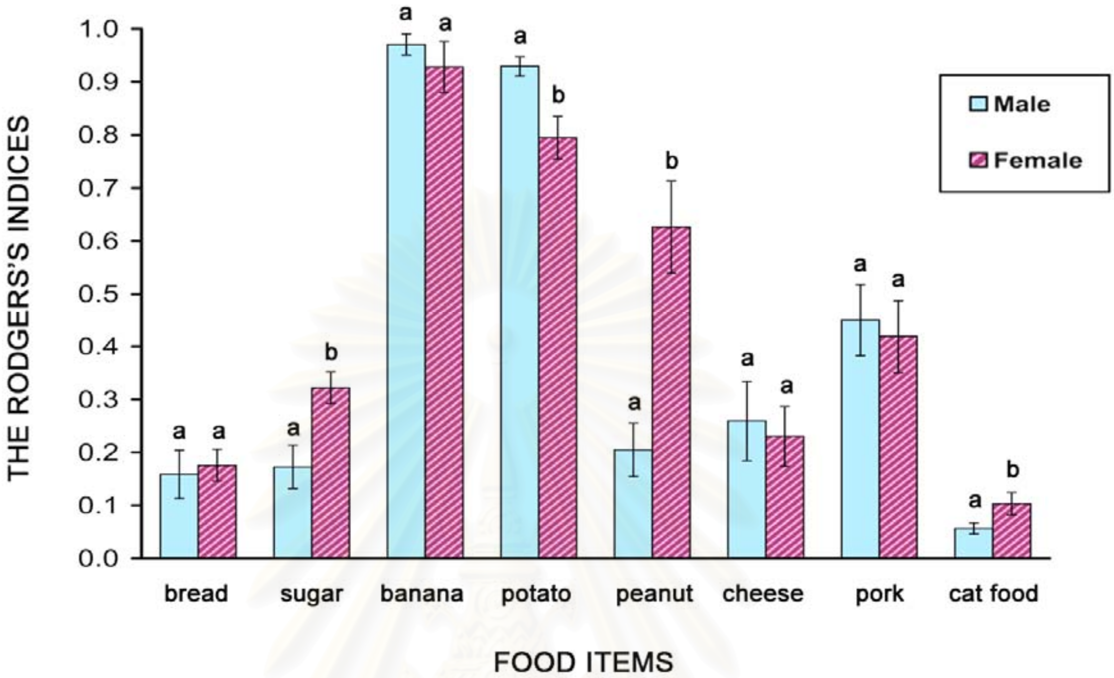

Figure 3.6 Means (± SE) of the Rodgers's indices comparing between the male and female German cockroaches for each food item. These indices indicate the food preference in male and female cockroaches (rang 0-1, 0 = avoid and 1= prefer). The bar graphs with different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

2. Feeding behavior of the German cockroach

The result showed that the German cockroaches usually hid in their harborage during daytime. The beginning of their feeding behaviors occurred around 05.00 pm and rapidly increased around 07.00 pm. They also showed the first peak during the hours between 07.00-10.00 pm. Their activities slowly decreased starting at 11.00 pm. Then, their feeding behavior was found again at 03.00 am and sharply increased at 04.00 am. The second peak showed at 04.00-05.00 am. After that, their activities rapidly decreased around 06.00 am. They then hid in their harborage from around 07.00 am to 04.00 pm (Table 3.4; Figure 3.7).

Table 3.4 Mean (± SE) number of active cockroaches observed from 5.00 pm to 4.00 pm. Total of 20 male and 20 female German cockroaches were observed in this experiment (5 replicates).

Time	Active cockroaches
5 pm	8 ± 0.58
6 pm	19 ± 1.53
7 pm	40 ± 1.00
8 pm	36 ± 1.53
9 pm	32 ± 1.00
10 pm	27 ± 1.73
11 pm	10 ± 0.58
12 pm	4 ± 0.00
1 am	3 ± 0.58
2 am	3 ± 0.58
3 am	5 ± 0.00
4 am	34 ± 1.00
5 am	28 ± 2.00

Table 3.4 Mean (± SE) number of active cockroaches observed from 5.00 pm to 4.00 pm. Total of 20 male and 20 female German cockroaches were observed in this experiment (5 replicates) (cont.).

Time	Active cockroaches
6 am	2 ± 0.00
7 am	0 ± 0.00
8 am	0 ± 0.00
9 am	0 ± 0.00
10 am	0 ± 0.00
11 am	0 ± 0.00
12 am	0 ± 0.00
1 pm	0 ± 0.00
2 pm	0 ± 0.00
3 pm	0 ± 0.00
4 pm	0 ± 0.00

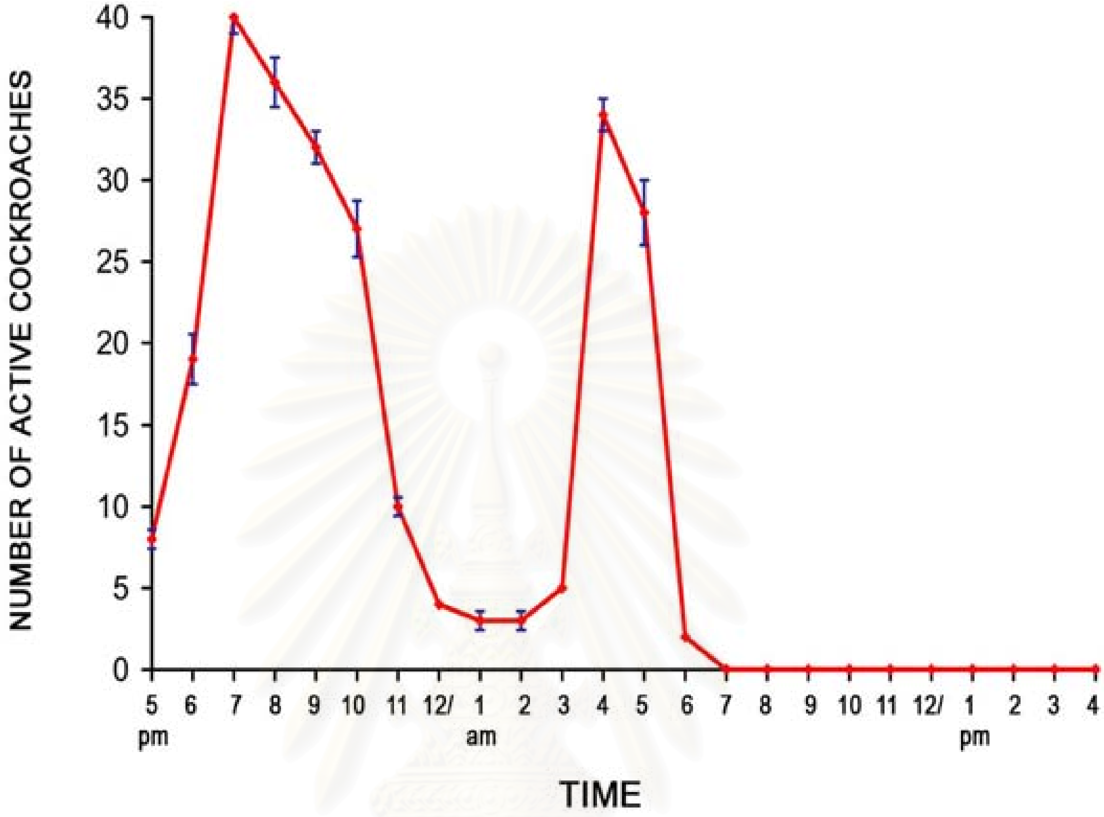

Figure 3.7 Mean (± SE) number of active cockroaches observed from 5.00 pm to 4.00 pm. Total of 20 male and 20 female German cockroaches were observed in this experiment (5 replicates). Each point represents the mean number of active cockroaches at each time of observation.

Their feeding behavior started with leaving their shelter and swinging their antennae to detect food odor followed by walking to the food, touching, and feeding (Figure 3.8).

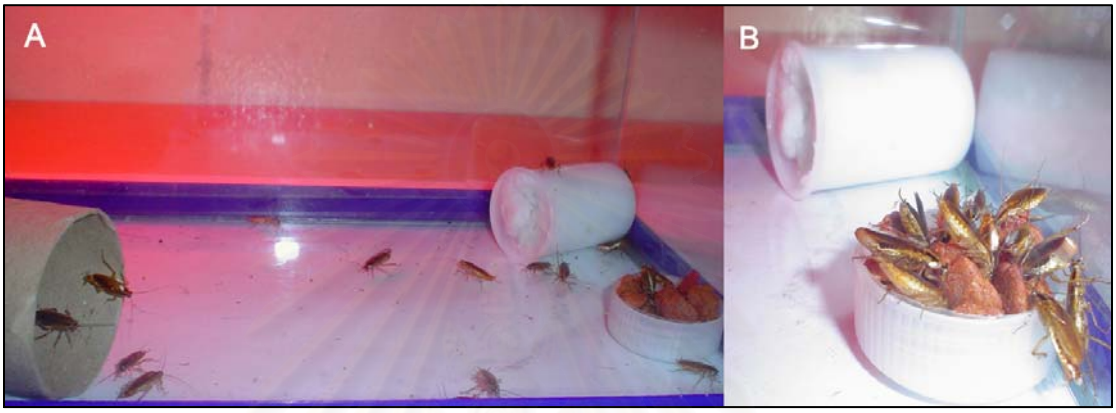

Figure 3.8 The both sexes of the German cockroaches feeding behavior occurred around 05.00 pm. Their feeding behavior started with leaving their shelter and swinging their antennae to detect food odor (A) followed by walking to the food, touching, and feeding (B).

Discussion and Conclusion

Several important inferences may be derived from the data such as the differences of being interested in food choices, sex-specific food preference in the German cockroaches, and their feeding behavior. The most notable result was the difference in nutritional requirements between the male and female cockroaches. They were contrarily significant between sexes in protein utilization.

For the nutritional requirements, it is well known that the German cockroaches prefer diets which are higher in carbohydrate than in fat and protein content (Ross and Mullins, 1995). Moreover, under conditions of extreme starvation, the cockroaches often ate the nearest food substance that they encounter. (Reierson, 1995). From the observations, when they were placed in the center of an olfactometer, they walked directly to the nearest food and fed them. Their behavior corresponded with the other insects which did not respond to monotonous odors as vigorously as when they were very hungry (Reierson, 1995). When the cockroaches were full up, they hid around border of the chamber. Then, they surveyed the tested food items in every chamber of the olfactometer before selecting the food.

The results showed that the male German cockroaches significantly preferred banana and potato whereas the female cockroaches significantly preferred only banana. It might be because of the cockroaches preferred banana and potato due to their richness in carbohydrate. The carbohydrate content could serve them as primary energy sources (Carrel and Tanner, 2002). Most of the German cockroaches preferred banana more than any other kinds of food items, and only small numbers preferred cat food. Besides the nutritional factors, the odors and textures of food were also important. The cockroaches often respond to food odors over short distances (Reierson, 1995). In this experiment, the banana texture was soft and freshly. Thus, the banana attracted those ravenous cockroaches. In contrast, the texture of cat food was very rigid, therefore, the cockroaches might be aroused and attracted by soft and freshly food which are easy to eat under stress condition such as extremely starvation.

For the sex-specific differences of the German cockroaches, cockroaches and other animals are able to self-select a suitable diet given a variety of nutrient choices (Carrel and Tanner, 2002). The female cockroaches heavily invest in production of large ootheca unlike from males that produce small spermatophores (Leibensperger *et al*., 1985). Moreover, female fecundity commonly depends on the ingestion of protein that necessary for egg development (Reierson, 1995). Accordingly, the female cockroaches require substantially protein more than the males (Clarebrough *et al*., 2000). When Rodgers's indices for food preference were calculated for various types of food, then significant differences between male and female preference for peanut and sugar, implying that female cockroaches could be considered as protein-hunger and energy-hunger (Figure 3.6). The female German cockroaches require large amount of nutrient reserves, especially proteins, for oothecal production. Additionally, more energy is needed during preoviposition, incubation, and oviposition periods (Ross and Mullins, 1995). The result also corresponded to the previous studies. For example, female peacock butterflies and Adonis blue butterflies preferred amino acid-rich floral nectars whereas the males of both insects preferred sugar-rich floral nectars (Erhardt and Rusterholz, 1998; Rusterholz and Erhardt, 2000). Furthermore, the male Madagascar hissing cockroaches were also reported on preferred carbohydrate-rich foods preference whereas the females preferred protein-rich foods (Carrel and Tanner, 2002).

For the feeding behavior, the German cockroaches are nocturnal insects (Ross and Mullins, 1995). The foraging activity of the German cockroaches was related to the general circadian activity phases (Metzger, 1995) that corresponded to the result of this experiment. The foraging time of them occurred at night with two peaks of feeding activity. The first peak occurred during the hours between 07.00-10.00 pm and the second peak occurred during 04.00-05.00 am. For the first peak, the cockroaches left from their shelter and foraging activity occurred not far from their harborage. The second peak, their activities were occurred more distant from their shelter than the first peak. In addition, each cockroach showed the same feeding behavior pattern. Firstly, the cockroach left the shelter and swung the antennae for detecting food odor by the olfactory sensillum (or chemoreceptors) which were located on the antennae (Ross and Mullins, 1995; Tichy *et al*., 2005). Secondly, it walked to the food and touched them. Finally, the cockroach fed the food and drank some water using its mouthpart.

In conclusion, the cockroach trapping should be done at night. Carbohydrate-rich foods mixed with protein-rich foods with strong odor are good baits for trapping the German cockroaches in the fields. Ultimately, the cockroach trapping with non-chemical baits could be induced to the domestic pest control.

CHAPTER 4

POPULATION DYNAMICS OF THE GERMAN COCKROACH, *Blattella germanica* (Linnaeus), IN BANGKOK MARKETS

Introduction and Objective

Cockroaches are an important group of insect pests in human environment. Although about 4,000 species of cockroaches have been identified, about 1% of cockroaches is the common pest in the world (James and Harwood, 1969; Roth, 1991; Grimaldi and Engel, 2005). Cockroaches not only an annoying pest but are also capable of transmitting pathogens to human such as viruses, bacteria, protozoa, fungi and helminthes (Brenner, 1995). Moreover, they generally give displeasure and an impression of dirtiness. When they walk across food and utensils, they may leave food containing many pathogens that cause dysentery, food poisoning, and other human diseases (Fotedar *et al*., 1991; Rivault *et al.,* 1993; Brenner, 1995; Tachbele *et al.,* 2006). Cockroaches are also important components of house dust allergen (Ross and Mullins, 1995; Pumhirun *et al*., 1997; Sarinho *et al*., 2004).

At least ten species of cockroaches could be found in urban environment of Thailand. According to many studies such as Benjapong *et al.* (1997) surveyed of species and density of cockroach in 4 hospitals in central of Thailand. The results showed that 5 cockroach species including nymphal forms distribute with infested by *Blattella germanica, Periplaneta americana*, *P. brunnea, Neostylopyga rhombiofolia and Nauphoeta cinerea*. Moreover, Tawatsin *et al.* (2001), using sticky trap, reported that ten cockroach species could be caught from the houses in 14 provinces of Thailand, belonging to six genera, such as *B. germanica, B. lituricollis, P. americana*, *P. australasiae, P. brunnea, P. fuliginosa, Ne. rhombiofolia, Na. cinerea, Pycnoscelis surinamensis,* and *Supella longipalpa*. In addition, similar species of cockroaches were investigated from urban dwellings in Thailand such as Sriwichai (2001) who investigated the distribution of indoor cockroaches in Bangkok areas by using commercial sticky traps and Damsuwon (2003) who studied the infestation of indoor cockroaches in Nonthaburi Province. Eight species of cockroaches were found such as *B. germanica, B. lituricollis, P. americana*, *P. australasiae, P. brunnea, Ne. rhombiofolia, Py. surinamensis,* and *Supella longipalpa*.

In 2004, Bangkok metropolitan population's density average was 3,592 persons per square kilometers while population density in Thailand average was only 121 persons per square kilometers (Bangkok Metropolitan Administration [BMA], 2005a). The crowded dwelling becomes good resources for food and water for the pest species especially cockroaches (Ross and Mullins, 1995).

From the previous reports, the German cockroach, *B. germanica,* was the common species that infested in urban areas in Thailand. Prior to this study, there are a few documents existed on the population dynamics of cockroaches in markets which are public place and many foods are stayed. The objective of this experiment was to investigate the population dynamics of the German cockroaches in the market areas of Bangkok, Thailand.

Materials and Methods

1. Selection of the study areas

The population dynamics of the German cockroaches were investigated in Bangkok markets. The Stratified random sampling (Krebs, 1999) was used. The selected market areas were investigated from two different human population density zones and from two different classes of the Bangkok markets. Bangkok metropolitan consists of 50 districts. It is 1,568.737 square kilometers with 5,634,132 total populations (BMA, 2005a).

A total of 50 districts in Bangkok city are divided into 2 zones based on human population density. The first zone, high population density zone (> 10,000 persons per square kilometers), consists of 17 districts such as Pom Prap Sattru Phai, Samphanthawong, Thon Buri, Din Daeng, Khlong San, Ratchathewi, Dusit, Phra Nakhon, Bangkok Yai, Bang Sue, Bangkok Noi, Pathum Wan, Sathon, Bang Rak, Bang Kho Laem, Khlong Toei, and Bang Phlat. The second zone, low population density zone (0 – 10,000 persons per square kilometers), consists of 33 districts such as Phaya Thai, Phasi Charoen, Phra Khanong, Chom Thong, Watthana, Rat Burana, Bueng Kum, Wang Thonglang, Yan Nawa, Bang Na, Lat Phrao, Chatuchak, Lak Si, Huai Khwang, Bang Kapi, Suan Luang, Don Mueang, Bang Khen, Bang Khae, Sai Mai, Taling Chan, Nong Khaem, Thung Khru, Khan Na Yao, Saphan Sung, Prawet, Bang Bon, Min Buri, Thawi Watthana, Lat Krabang, Khlong Sam Wa, Bang Khun Thian, and Nong Chok (BMA, 2005a).

Considering to the markets in Bangkok area, all the markets are also classified into 2 classes. The markets in class 1, consist of 24 markets, are in the acceptable level of the environmental sanitary assessment (Appendix A) documented by the Environmental Health Department, Health division, BMA and the markets in class 2, consist of 131 markets, are not in the acceptable level of the environmental sanitary assessment (BMA, 2005b).

By the combination of human population density zones and the classes of Bangkok markets, 4 combination groups such as high population density zone/ market class 1, high population density zone/ market class 2, low population density zone/ market class 1, and low population density zone/ market class 2 were categorized and the numbers of the markets in each combination group are showed in Table 4.1. In this study, 12 market sites (3 sites for each combination group in Bangkok) were selected using random sampling method (Krebs, 1999), and the market's names and their location map were showed in Table 4.2 and Figure 4.1.

Table 4.1 The numbers of Bangkok markets categorized by their combinations of human population density zones and the classes of Bangkok markets.

Human population density (persons/ km^2)	**Market**		**Total**
	Class 1	**Class 2**	
> 10,000	4	60	64
0-10,000	20	71	91
Total	**24**	**131**	**155**

Table 4.2 The twelve Bangkok markets (study sites) were randomly selected. Market number 1-3 were in high population density zone/ market class 1 combination (HC1), market number 4-6 were in low population density zone/ market class 1 combination (LC1), market number 7-9 were in high population density zone/ market class 2 combination (HC2), and market number 10-12 were in low population density zone/ market class 2 combination (LC2).

Human population density (persons/ km^2)	**Market**	
	Class 1	**Class 2**
> 10,000	**Market no.1** Maneepiman market, Bang Sue District	**Market no.7** Pechburi market, Rachathewi District
	Market no.2 Bangkhunsri market, Bangkok Noi District	**Market no.8** Chatchai market, Bangkok Yai District
	Market no.3 Bang Kho Laem market, Bang Kho Laem District	**Market no.9** Watkhak market, Bang Rak District
0-10,000	**Market no.4** On-nuch market, Watthana District	**Market no.10** Watsai market, Chom Thong District
	Market no.5 Nakornthai market, Bang Kapi District	**Market no.11** Roongchareon market, Yan Nawa District
	Market no.6 Eamsombat market, Suan Laung District	**Market no.12** Ladprao 123 market, Bang Kapi District

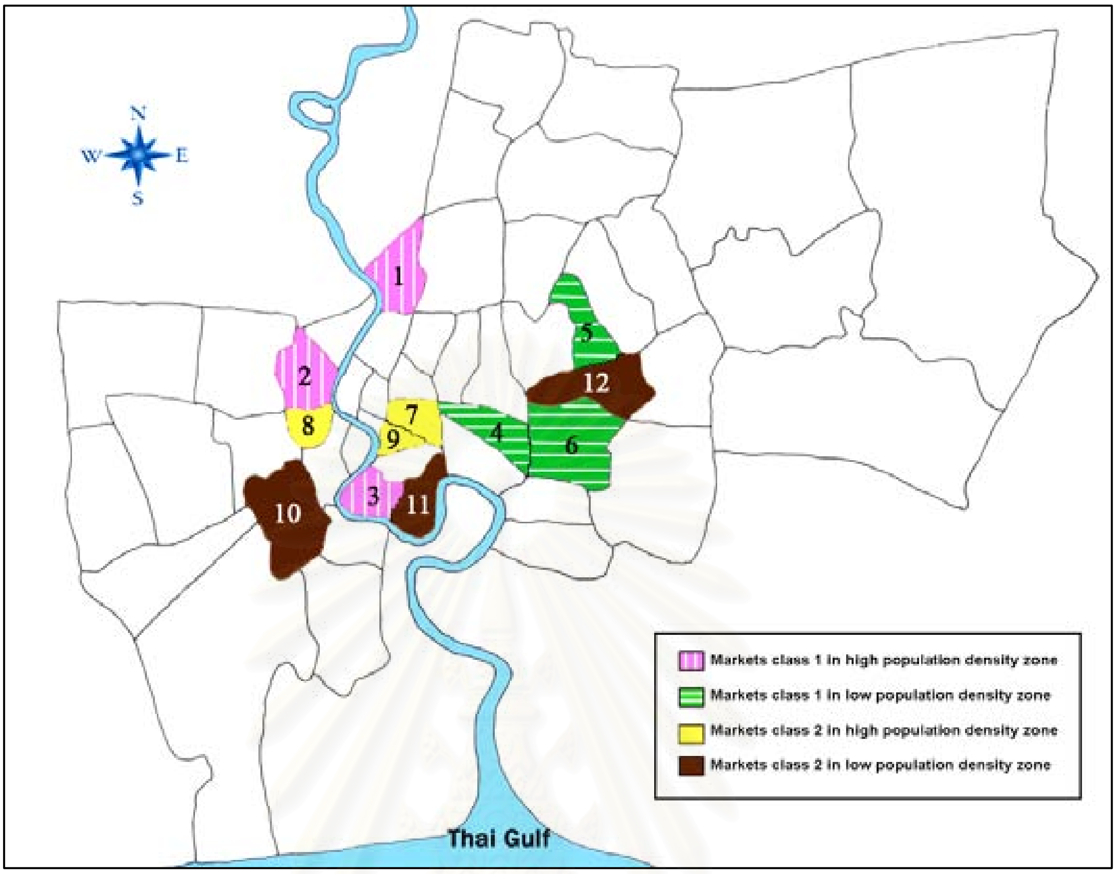

Figure 4.1 Map of twelve study sites located in eleven districts in Bangkok. Markets number 1-3 were in high population density zone/ markets class 1 combination (HC1), markets number 4-6 were in low population density zone/ markets class 1 combination (LC1), markets number 7-9 were in high population density zone/ markets class 2 combination (HC2), and markets number 10-12 were in low population density zone/ markets class 2 combination (LC2).

2. Cockroach collection

The study was conducted monthly from March 2005 to March 2006. Modified jar traps were used for the study. A modified jar trap was a plastic cup (8 cm in diameter and 11 cm in height) with the sticky crepe paper fixed around the cup for enabled cockroaches to enter (Figure 4.2). The upper two-fifth of the inner surface of the jar trap was coated with a thin film of petroleum jelly (Vaseline®), cockroaches once entering trap lured by food were unable to climb out. From the last chapter, both male and female German cockroaches preferred banana, however, from the preliminary study in market fields, banana also attracts the foraging rats. Following the technique reported by Wileyto and Boush (1983), therefore, ground peanut and cat food saturated with beer were used as baits in the further studies.

Figure 4.2 A modified jar trap was a plastic cup with the sticky crepe paper fixed around the trap for enabled cockroaches to enter, and a thin film of petroleum jelly was applied to the inner surface to prevent cockroaches escape.

Twenty modified jar traps filling with baits were placed randomly in 3 kinds of shops in each market such as butcher shops (fresh pork, beef, and chicken), vegetable shops (fresh vegetable), and groceries (rice, seasoning, garlic, red onion, bean, dried shrimps, and dried squids). The traps were placed next to walls and near equipments in each shop for 24 hours. The traps were collected and taken to the laboratory, where the cockroaches were identified and counted. The number and stage of the German cockroach were recorded. The nymphal stages were divided into three class sizes such as small nymphs were the 1^{st} and 2^{nd} instars, medium nymphs were the 3^{rd} and 4^{th} instars, and large nymphs were the 5^{th} and 6^{th} instars. The temperature and relative humidity were recorded using thermo-hygrometer.

Data analysis

Descriptive statistics were used to present as number, percentage, mean, and standard error. Mean numbers of the German cockroaches caught per trap were compared and analyzed. All data were checked for normal distribution. If any data was normal distribution, ANOVA with LSD analysis ($p < 0.05$) was used. But, if any data was not normal distribution, *Mann-Whitney U*-test ($p < 0.05$) was used.

Results

I. Cockroach species in Bangkok markets

A total of 11,944 cockroaches were trapped from 12 Bangkok markets during the investigation. The seven species of cockroaches, belonging to six genera, caught from 12 Bangkok markets were *Blattella germanica* (46.12%), *Nauphoeta cinerea* (5.16%), *Neostylopyga rhombiofolia* (0.03%), *Periplaneta americana* (45.30%), *P. brunnea* (2.20%), *Supella longipalpa* (0.04%), and *Symploce pallens* (1.15%) (Table 4.3). The *B. germanica* were trapped at the highest number in Bangkok markets in this study. The morphological characteristics of each species were presented in Figure 4.3-4.4.

Table 4.3 Cockroach species found in 12 Bangkok markets from March 2005 to March 2006 (Asahina, 1983; Lee *et al*., 2000).

Cockroach species	Total cockroaches caught	%
Blattella germanica (Linnaeus)	5,508	46.12
Nauphoeta cinerea (Olivier)	616	5.16
Neostylopyga rhombiofolia (Stoll)	4	0.03
Periplaneta americana (Linnaeus)	5,411	45.30
P. brunnea (Burmeister)	263	2.20
Supella longipalpa (Fabricius)	5	0.04
Symploce pallens (Stephens)	137	1.15
Total	**11,944**	**100**

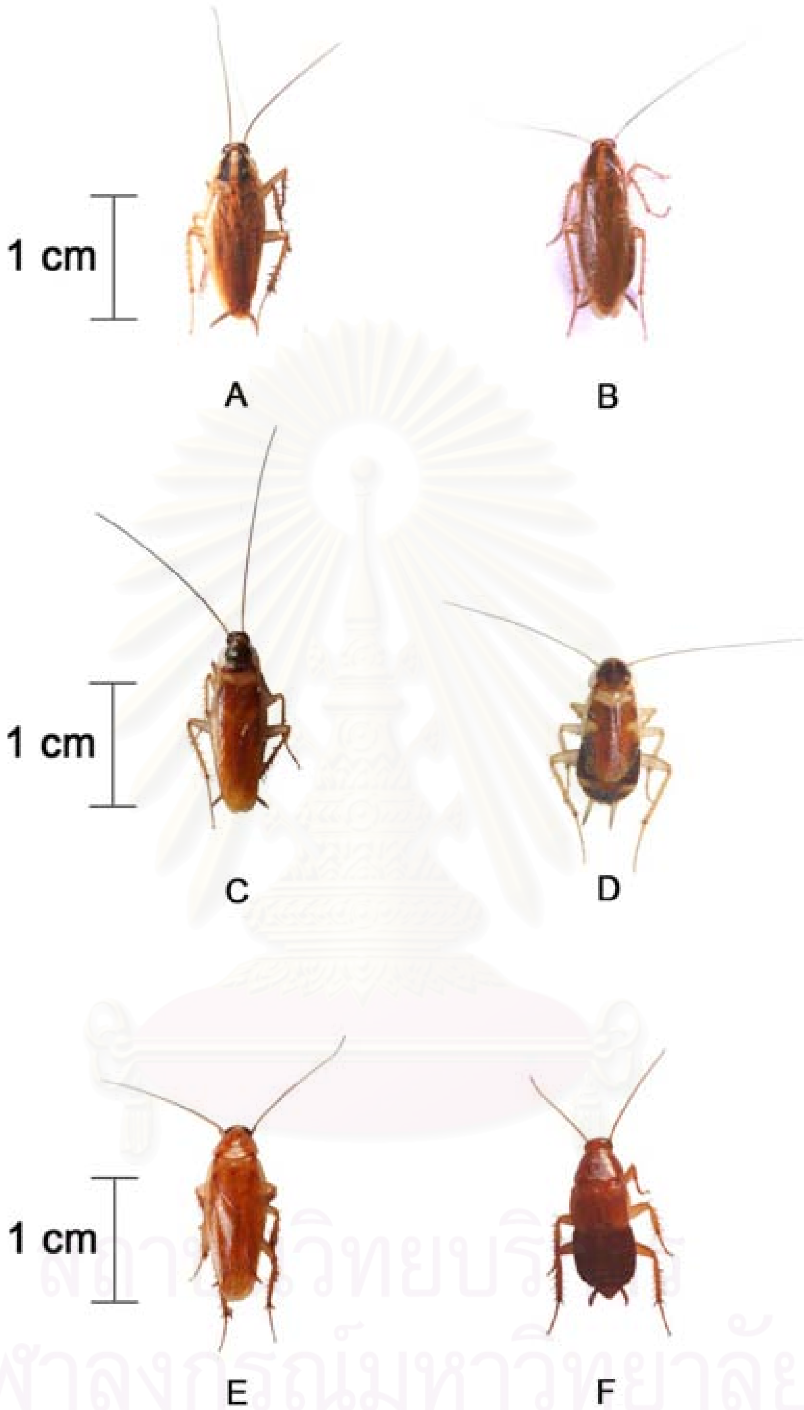

Figure 4.3 The small size cockroaches found in the Bangkok markets from March 2005 to March 2006 such as *B. germanica*: male (A), female (B); *S. longipalpa:* male (C), female (D); and *Sy. pallens*: male (E), female (F).

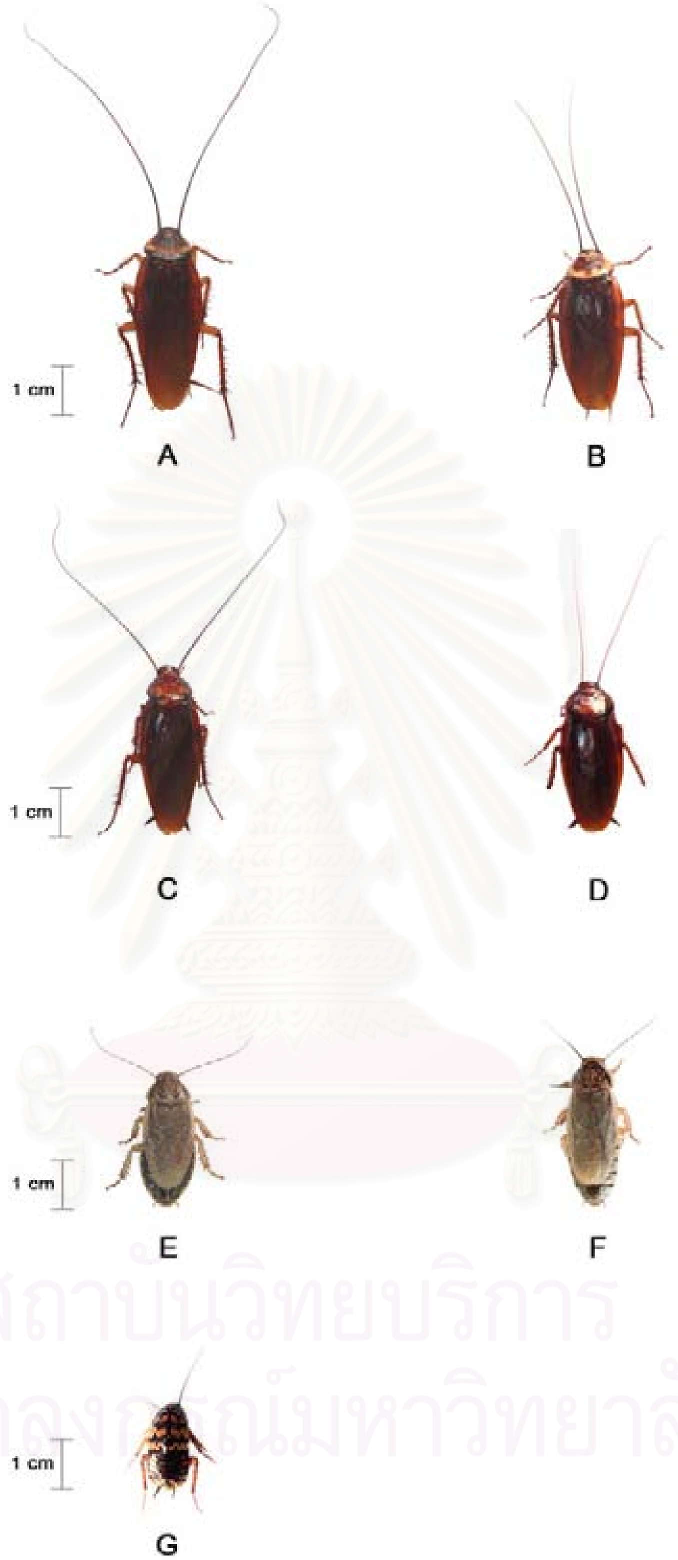

Figure 4.4 The medium size cockroaches found in the Bangkok markets from March 2005 to March 2006 such as *P. americana:* male (A), female (B); *P. brunnea*: male (C), female (D); *Na. cinerea:* male (E), female (F); and *Ne. rhombiofolia:* nymph (G).

Species number of cockroaches was highest in Petchburi market (market number 7). In this market, all seven species of cockroaches were found. The lowest species number of cockroaches was in Eamsombat market (market number 6). In the latter market, only two species were found such as *B. germanica* and *P. americana*. According to the all cockroach species in the Bangkok market, the *B. germanica* were trapped in the largest number (5,508 individuals) and found in the largest number in Ladprao 123 market (market number 12), however, none of them was caught in Bang Kho Laem market (market number 3). Furthermore, the *P. americana* were trapped in all markets and found in the largest number also in market number 12. On the other hand, the *Ne. rhombiofolia* and the *S. longipalpa* were found less than 1% of total cockroaches caught during the study period (Table 4.4).

Table 4.4 Species[1] of the cockroaches found in Bangkok markets.

Market no.	**Cockroach species (%)** [2]						
	B. germanica	*N. cinerea*	*Ne. rhombiofolia*	*P. americana*	*P. brunnea*	*S. longipalpa*	*Sy. pallens*
1	329 (70.15)	-	-	127 (27.08)	10 (2.13)	3 (0.64)	-
2	107 (60.12)	-	-	56 (31.45)	15 (8.43)	-	-
3	-	104 (35.49)	1 (0.34)	150 (51.21)	13 (4.43)	-	25 (8.53)
4	462 (80.35)	11 (1.91)	-	98 (17.05)	3 (0.52)	1 (0.17)	-
5	110 (32.07)	-	-	207 (60.35)	26 (7.58)	-	-
6	1184 (97.21)	-	-	34 (2.79)	-	-	-
7	947 (35.54)	315 (11.83)	1 (0.03)	1,306 (49.1)	72 (2.70)	1 (0.03)	23 (0.86)
8	262 (23.21)	-	-	844 (74.75)	23 (2.04)	-	-
9	398 (43.83)	-	-	489 (53.85)	20 (2.21)	-	1 (0.11)
10	440 (41.52)	-	-	598 (56.41)	20 (1.89)	-	2 (0.18)
11	67 (35.63)	7 (3.73)	2 (1.07)	106 (56.38)	6 (3.19)	-	-
12	1,202 (41.19)	179 (6.13)	-	1,396 (47.85)	55 (1.88)	-	86 (2.95)
Total	**5,508**	**616**	**4**	**5,411**	**263**	**5**	**137**

[1] *B. = Blattella; Na. = Nauphoeta; Ne. = Neostylopyga; P. = Periplaneta; S. = Supella*; and *Sy. = Symploce.*

[2] Percentage in each bracket based on total number of cockroaches caught in each market.

II. Population dynamics of the German cockroaches

Population dynamics of total number of the German cockroaches

The German cockroaches showed the fluctuation during the study period. The results showed that the mean numbers of the total cockroaches caught per trap were significantly different (F = 17.062; df = 12, 3107; p < 0.0001).

The mean number of cockroaches was highest in July, followed by August, June, September, May, April, October, March 2005, November, March 2006, February, January, and December, respectively. The two highest peaks were in July (4.54 ± 1.78) and August (4.21 ± 1.63) whereas the lowest peaks were in December (0.37 ± 0.14) and January (0.43 ± 0.17). Even though the mean numbers of cockroaches caught per trap in July and August were not significantly different, the mean in July was significantly higher than the other months (LSD; df = 3107; p < 0.05).

During the period of study, the mean numbers per trap of cockroaches started increasing in April (1.49 ± 0.58) and rapidly decreased in September (2.33 ± 0.87) (Table 4.5; Figure 4.5).

Table 4.5 Mean (± SE) number of the German cockroaches caught per trap from twelve Bangkok markets from March 2005 to March 2006 (S = small nymph, M = medium nymph, L= large nymph, A = adult, and T = total number).

Mean number (per trap) of the German cockroaches

	March 2005	April	May	June
S	0.05 ± 0.02	0.12 ± 0.05	0.23 ± 0.11	0.18 ± 0.07
M	0.21 ± 0.09	0.35 ± 0.14	0.43 ± 0.17	0.64 ± 0.27
L	0.35 ± 0.14	0.52 ± 0.20	0.83 ± 0.32	1.04 ± 0.40
A	0.45 ± 0.17	0.50 ± 0.19	0.76 ± 0.30	0.90 ± 0.34
T	1.06 ± 0.42	1.49 ± 0.58	2.25 ± 0.89	2.76 ± 1.08

	July	August	September	October
S	0.59 ± 0.26	0.39 ± 0.15	0.22 ± 0.08	0.12 ± 0.04
M	1.00 ± 0.42	1.03 ± 0.44	0.42 ± 0.15	0.26 ± 0.09
L	1.65 ± 0.62	1.60 ± 0.61	0.87 ± 0.33	0.58 ± 0.20
A	1.30 ± 0.48	1.19 ± 0.44	0.83 ± 0.31	0.45 ± 0.16
T	4.54 ± 1.78	4.21 ± 1.63	2.33 ± 0.87	1.41 ± 0.50

	November	December	January 2006	February
S	0.05 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	0.03 ± 0.01
M	0.14 ± 0.05	0.02 ± 0.01	0.04 ± 0.02	0.10 ± 0.09
L	0.24 ± 0.09	0.11 ± 0.04	0.13 ± 0.05	0.18 ± 0.07
A	0.40 ± 0.18	0.24 ± 0.09	0.26 ± 0.09	0.23 ± 0.08
T	0.83 ± 0.32	0.37 ± 0.14	0.43 ± 0.17	0.54 ± 0.21

	March
S	0.03 ± 0.01
M	0.12 ± 0.04
L	0.30 ± 0.10
A	0.25 ± 0.09
T	0.70 ± 0.25

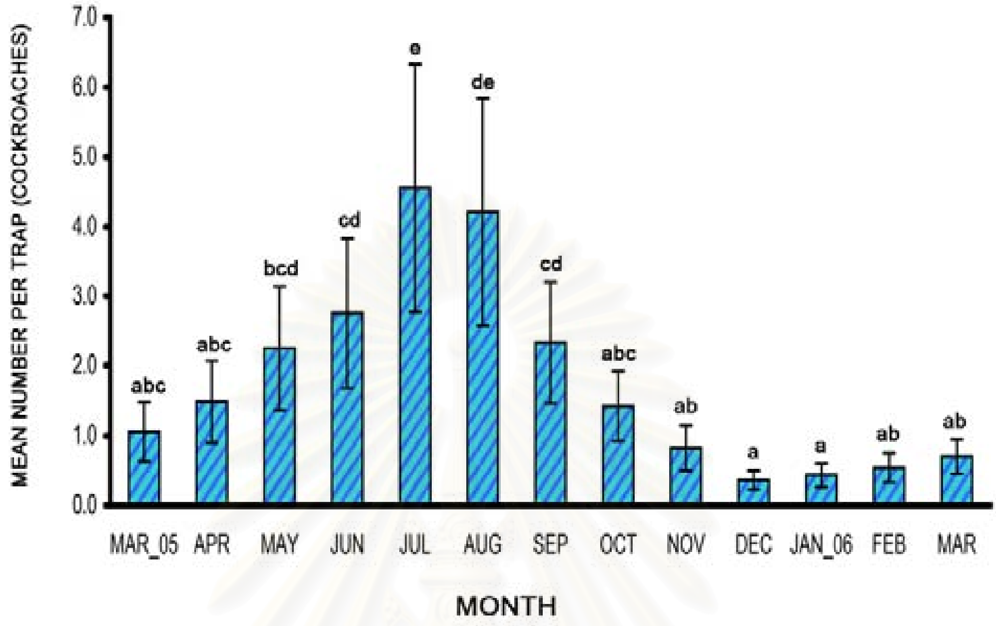

Figure 4.5 The overall mean (± SE) number (per trap) of the German cockroaches caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Population dynamics of the German cockroach stages

Mean numbers of the adult of the German cockroaches were significantly different during the study period (F = 3.876; df = 12, 3107; p < 0.0001). The highest mean was in July followed by August, June, September, May, April, October, March 2005, November, January, March 2006, December, and February, respectively. The two highest peaks were in July (1.30 ± 0.48) and August (1.19 ± 0.44) whereas the two lowest peaks were in February (0.23 ± 0.08) and December (0.24 ± 0.09). Even though the mean numbers of cockroaches caught per trap in July, August, June, and September were not statistically different, the mean in July was significantly higher than the other months (LSD; df = 3107; p < 0.05).

The mean numbers per trap of the adult cockroach catches started increasing in April (0.50 ± 0.19) and gradually decreased in September (0.83 ± 0.31) (Table 4.5; Figure 4.6).

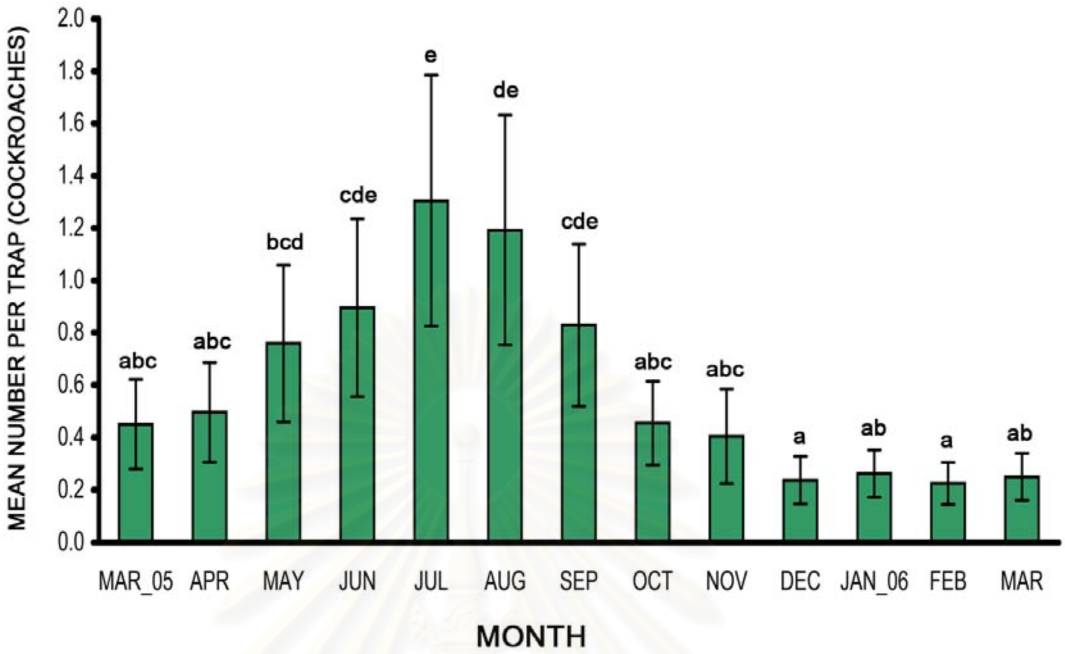

Figure 4.6 Mean (± SE) number (per trap) of the adult German cockroaches caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Mean numbers of the large nymph of the cockroaches were significantly different during the study period ($F = 5.544$; df = 12, 3107; $p < 0.0001$). The highest mean was in July followed by August, June, September, May, October, April, March 2005, March 2006, November, February, January, and December, respectively. The two highest peaks were in July (1.65 ± 0.62) and August (1.60 ± 0.61) whereas the two lowest peaks were in December (0.11 ± 0.04) and January (0.13 ± 0.05). Although the mean numbers of cockroaches caught per trap in July, August, and June were not significantly different, the mean in July and August were statistically higher than the other months (LSD; df = 3107; $p < 0.05$).

The mean numbers per trap of large nymph of the cockroaches started increasing in April (0.52 ± 0.20) and rapidly decreased in September (0.87 ± 0.33) (Table 4.5; Figure 4.7).

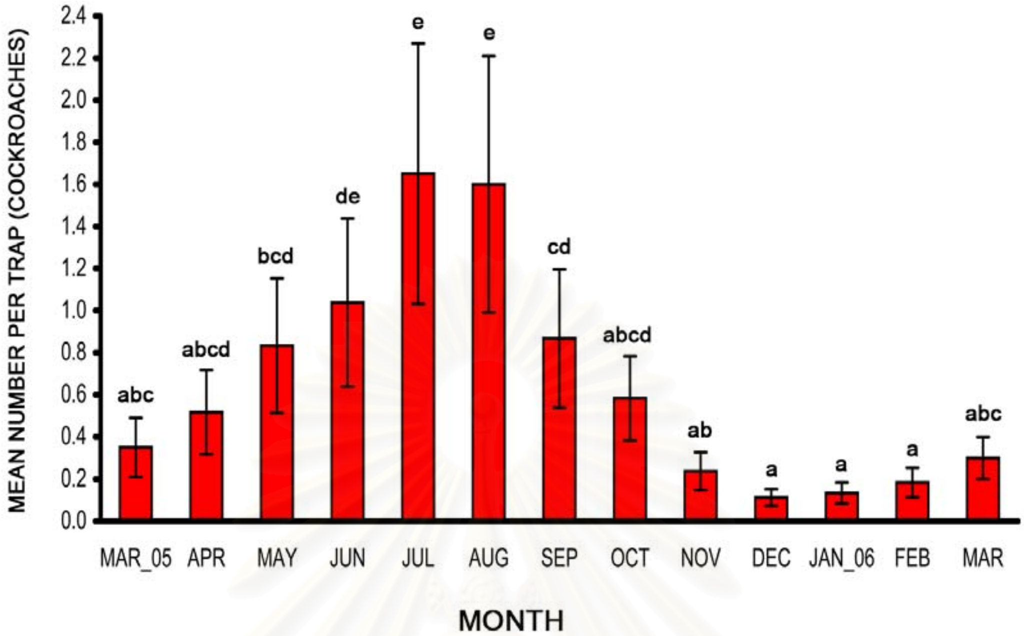

Figure 4.7 Mean (± SE) number (per trap) of the large nymph German cockroaches caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Mean numbers of the medium nymph of the cockroaches were significantly different during the study period (*F* = 4.490; df = 12, 3107; $p < 0.0001$). The highest mean was in August followed by July, June, May, September, April, October, March 2005, November, March 2006, February, January, and December, respectively. The two highest peaks were in August (1.03 ± 0.44) and July (1.00 ± 0.42) whereas the lowest peak was in December (0.02 ± 0.01). Although the mean numbers of cockroaches caught per trap in August, July, and June were not significantly different, the mean number per trap of cockroaches in August and July were significantly higher than the other months (LSD; df = 3107; $p < 0.05$).

The mean numbers per trap of the medium nymph of cockroaches started increasing in April (0.35 ± 0.14) and rapidly decreased in September (0.42 ± 0.15) (Table 4.5; Figure 4.8).

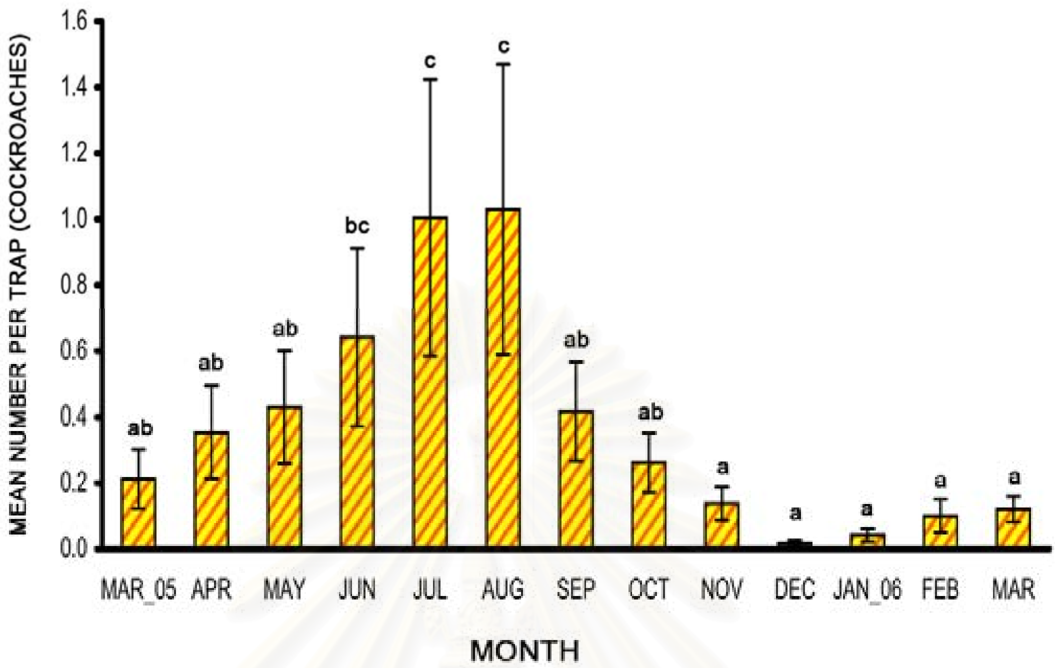

Figure 4.8 Mean (± SE) number (per trap) of the medium nymph German cockroaches caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Mean numbers of the small nymph of the German cockroaches were significantly different during the study period (*F* = 4.885; df = 12, 3107; $p < 0.0001$). The highest mean was in July, followed by August, May, September, June, April, October, March 2005, November, March 2006, February, January, and December, respectively. The two highest peaks were in July (0.59 ± 0.26) and August (0.39 ± 0.15) whereas the two lowest peaks were in December (0.00 ± 0.00) and January (0.00 ± 0.00). Even though the mean number of cockroaches caught per trap in July and August were not significantly different, the mean number per trap of cockroaches in July was significantly higher than the other months (LSD; df = 3107; $p < 0.05$).

The mean numbers per trap of the small nymph of cockroaches started increasing in April (0.12 ± 0.05) but slightly decreased in June (0.18 ± 0.07). Then the mean rapidly increased in July (0.59 ± 0.26). After that the mean moderately decreased in September (0.22 ± 0.08) and the mean numbers were zero in December and January (Table 4.5; Figure 4.9).

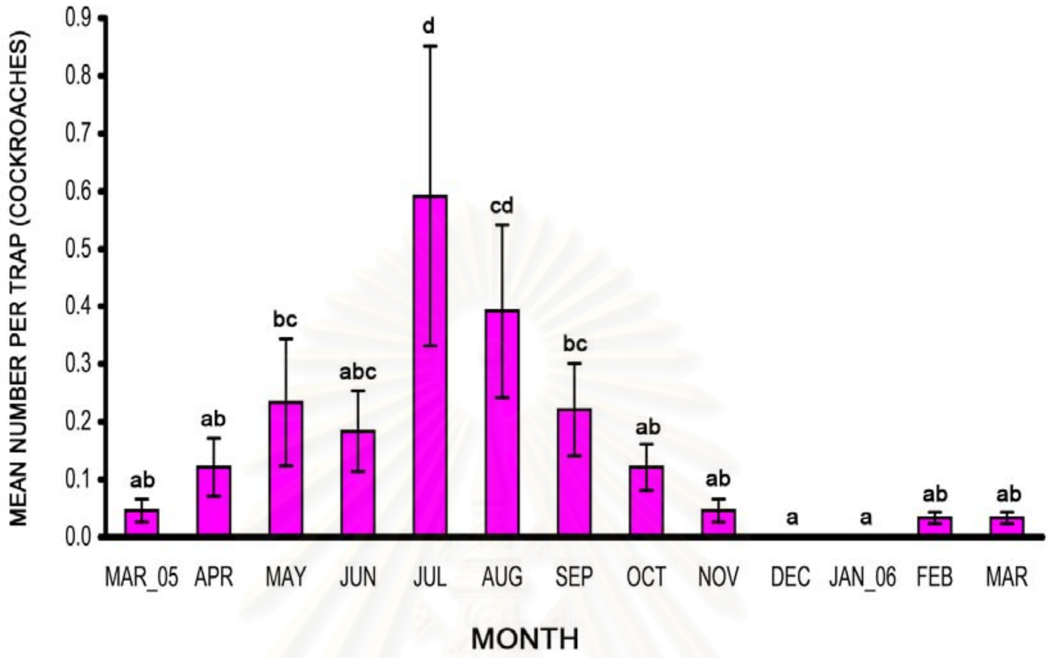

Figure 4.9 Mean (± SE) number (per trap) of the small nymph German cockroaches caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

The result in Table 4.6 presented the mean numbers (per trap) of all stages of the German cockroaches caught from Bangkok market from March 2005 to March 2006. Mean numbers of all stages of the German cockroaches were significantly different during the study period ($F = 17.733$; df= 3, 3116; $p < 0.0001$).

The highest mean number was the large nymphal stage (12.93 ± 1.43) whereas the lowest mean was the small nymphal stage (3.11 ± 0.49). Although the mean numbers of the large nymphal stage (12.93 ± 1.43) and adult cockroaches (11.94 ± 1.14) caught per trap were not significantly different, the mean of the large nymphal stage was significantly higher than the other cockroach stages (LSD; df = 3116; $p < 0.05$) (Table 4.6).

Table 4.6 Mean (± SE) number (per trap) of the German cockroach stages caught from twelve Bangkok market from March 2005 to March 2006 (S = small nymph, M = medium nymph, L= large nymph; and A = adult).

Months	**Cockroach stages**			
	S	**M**	**L**	**A**
March 2005	0.05 ± 0.02	0.21 ± 0.09	0.35 ± 0.14	0.45 ± 0.17
April	0.12 ± 0.05	0.35 ± 0.14	0.52 ± 0.20	0.50 ± 0.19
May	0.23 ± 0.11	0.43 ± 0.17	0.83 ± 0.32	0.76 ± 0.30
June	0.18 ± 0.07	0.64 ± 0.27	1.04 ± 0.40	0.90 ± 0.34
July	0.59 ± 0.26	1.00 ± 0.42	1.65 ± 0.62	1.30 ± 0.48
August	0.39 ± 0.15	1.03 ± 0.44	1.60 ± 0.61	1.19 ± 0.44
September	0.22 ± 0.08	0.42 ± 0.15	0.87 ± 0.33	0.83 ± 0.31
October	0.12 ± 0.04	0.26 ± 0.09	0.58 ± 0.20	0.45 ± 0.16
November	0.05 ± 0.02	0.14 ± 0.05	0.24 ± 0.09	0.40 ± 0.18
December	0.00 ± 0.00	0.02 ± 0.01	0.11 ± 0.04	0.24 ± 0.09
January 2006	0.00 ± 0.00	0.04 ± 0.02	0.13 ± 0.05	0.26 ± 0.09
February	0.03 ± 0.01	0.10 ± 0.09	0.18 ± 0.07	0.23 ± 0.08
March	0.03 ± 0.01	0.12 ± 0.04	0.30 ± 0.10	0.25 ± 0.09
Overall mean (per trap) [1]	**3.11 ± 0.49** [a]	**7.33 ± 0.99** [b]	**12.93 ± 1.43** [c]	**11.94 ± 1.14** [c]

[1] Means with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Mean numbers of all stages of the German cockroaches showed the same pattern. The data from Table 4.5 showed the two highest peaks in July and August. The mean numbers per trap of the German cockroaches started increasing in April and the mean rapidly decreased in September. The means decreased close to zero in December and January especially in December and January the mean numbers of the small nymph were zero. The curves of mean numbers of the total number, adult, large nymph, medium nymph, and small nymph of the German cockroaches, the average temperature ($^{\circ}$C), and the average relative humidity (%) from twelve Bangkok markets were presented in Figure 4.10; Figure 4.11.

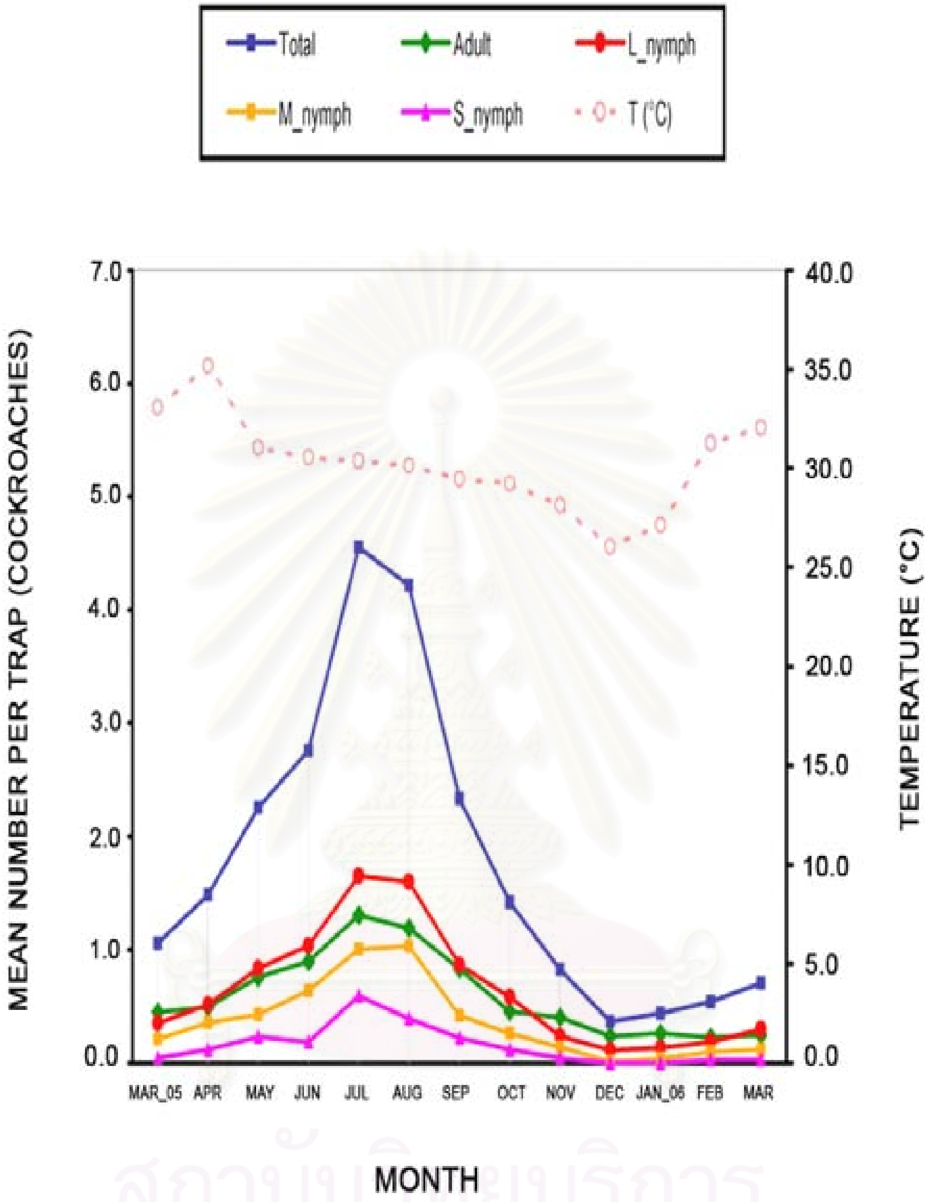

Figure 4.10 Mean number of the total number, adult, large nymph (L), medium nymph (M), and small nymph (S) of the German cockroaches in relation to temperature (°C) in twelve Bangkok markets. Each point on a curve represents a mean number per trap in each month during the study period.

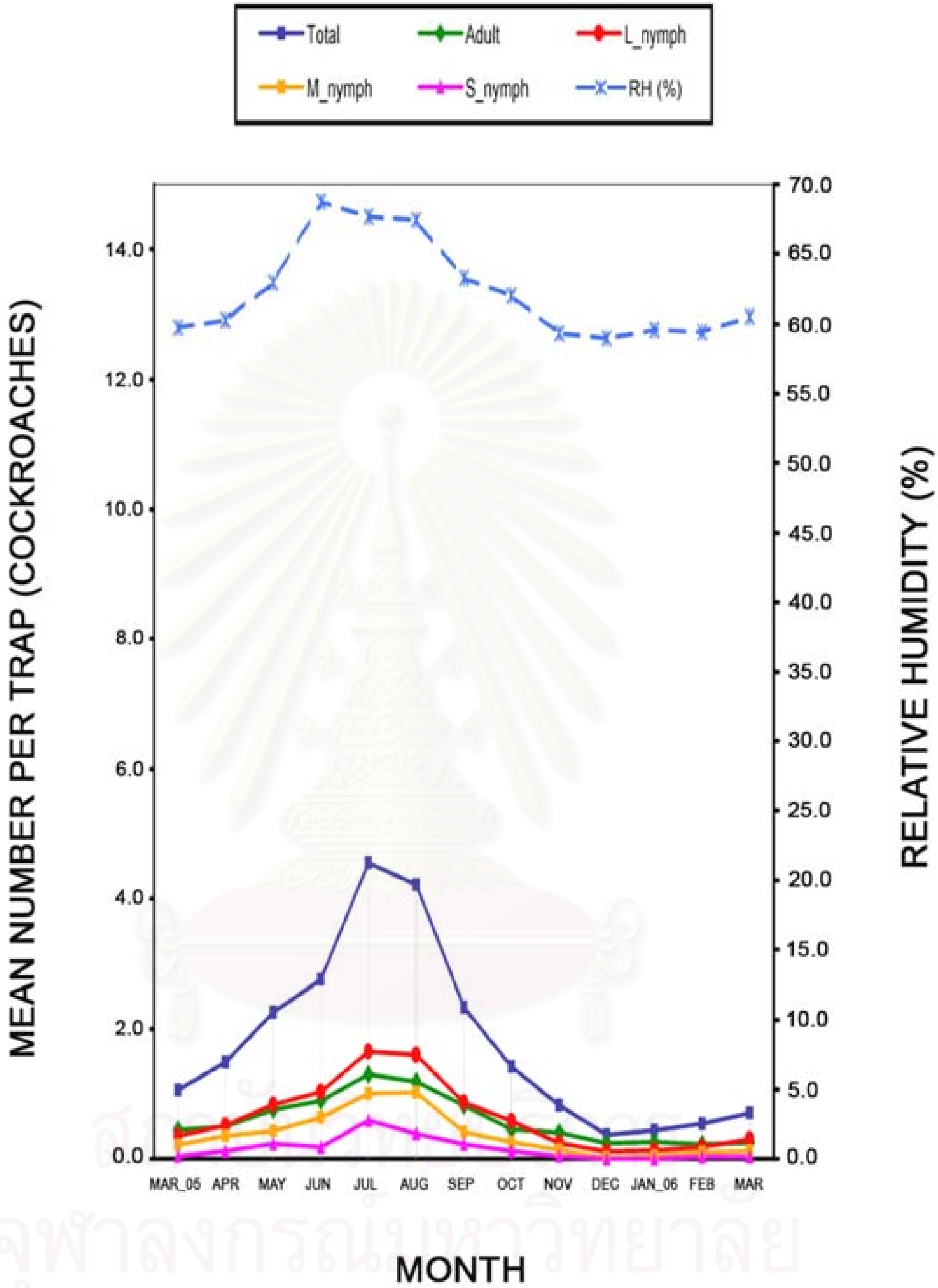

Figure 4.11 Mean number of the total number, adult, large nymph (L), medium nymph (M), and small nymph (S) of the German cockroaches in relation to relative humidity (%) in twelve Bangkok markets. Each point on a curve represents a mean number per trap in each month during the study period.

Comparisons of the German cockroach numbers in twelve Bangkok markets

Mean numbers (per trap) of the German cockroaches in all studied markets showed the same pattern. The two highest peaks were in July and August whereas the two lowest peaks were in December and January. The mean numbers per trap of the cockroaches started increasing in April and rapidly decreased in September. The mean numbers were close to zero in December and January (Table 4.7; Figure 4.12).

Table 4.7 Mean (± SE) numbers (per trap) of the German cockroaches trapped from twelve Bangkok market during March 2005 to March 2006.

Mean number per trap of the German cockroaches

Market	March 2005	April	May	June	July
1	0.55 ± 0.23	1.50 ± 0.65	1.80 ± 0.73	1.95 ± 0.81	3.00 ± 1.09
2	0.20 ± 0.20	0.35 ± 0.22	0.25 ± 0.14	0.60 ± 0.28	1.35 ± 0.59
3	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
4	1.50 ± 0.60	1.25 ± 0.52	1.50 ± 0.60	2.60 ± 1.04	4.10 ± 1.68
5	0.30 ± 0.14	0.45 ± 0.28	0.65 ± 0.20	0.45 ± 0.11	0.90 ± 0.45
6	2.40 ± 0.97	3.45 ± 1.41	5.45 ± 2.27	7.35 ± 2.90	15.20 ± 6.05
7	3.20 ± 1.27	4.00 ± 1.66	4.45 ± 1.79	5.10 ± 2.02	8.80 ± 3.52
8	0.80 ± 0.36	1.00 ± 0.46	1.25 ± 0.55	1.40 ± 0.60	2.95 ± 1.22
9	0.40 ± 0.35	0.80 ± 0.38	2.00 ± 0.87	2.65 ± 1.06	3.10 ± 1.28
10	0.80 ± 0.33	1.05 ± 0.48	2.20 ± 0.90	2.20 ± 0.89	4.85 ± 1.83
11	0.10 ± 0.10	0.15 ± 0.08	0.25 ± 0.12	0.55 ± 0.27	0.90 ± 0.42
12	2.45 ± 1.03	3.85 ± 1.54	7.25 ± 3.00	8.25 ± 3.33	9.45 ± 3.78

Market	August	September	October	November	December
1	2.65 ± 0.96	1.95 ± 0.82	1.45 ± 0.69	0.65 ± 0.45	0.20 ± 0.13
2	1.25 ± 0.52	0.55 ± 0.31	0.30 ± 0.20	0.10 ± 0.10	0.10 ± 0.10
3	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
4	4.55 ± 1.81	2.95 ± 1.21	1.65 ± 0.71	0.90 ± 0.39	0.50 ± 0.19
5	0.75 ± 0.25	0.40 ± 0.28	0.45 ± 0.35	0.40 ± 0.35	0.15 ± 0.08
6	10.75 ± 4.43	5.20 ± 2.11	2.40 ± 0.95	2.25 ± 0.92	1.15 ± 0.53
7	6.35 ± 2.66	5.00 ± 1.99	3.40 ± 1.44	1.45 ± 0.59	0.95 ± 0.42
8	2.35 ± 0.93	0.95 ± 0.44	0.80 ± 0.34	0.20 ± 0.20	0.10 ± 0.10
9	5.05 ± 2.05	2.80 ± 1.15	1.50 ± 0.60	0.40 ± 0.40	0.10 ± 0.10
10	3.50 ± 1.39	2.25 ± 0.96	1.60 ± 0.64	1.00 ± 0.42	0.45 ± 0.27
11	0.45 ± 0.27	0.20 ± 0.13	0.20 ± 0.20	0.05 ± 0.05	0.00 ± 0.00
12	12.9 ± 5.16	5.75 ± 2.27	3.30 ± 1.35	2.50 ± 1.05	0.70 ± 0.36

Market	January 2006	February	March
1	0.15 ± 0.10	0.30 ± 0.25	0.30 ± 0.21
2	0.00 ± 0.00	0.05 ± 0.05	0.25 ± 0.14
3	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
4	0.25 ± 0.12	0.25 ± 0.16	1.10 ± 0.48
5	0.15 ± 0.15	0.20 ± 0.20	0.25 ± 0.25
6	1.00 ± 0.42	1.30 ± 0.54	1.30 ± 0.54
7	1.30 ± 0.53	1.60 ± 0.69	1.75 ± 0.74
8	0.40 ± 0.26	0.35 ± 0.19	0.55 ± 0.28
9	0.35 ± 0.26	0.35 ± 0.19	0.40 ± 0.31
10	0.60 ± 0.28	0.65 ± 0.32	0.85 ± 0.40
11	0.15 ± 0.15	0.15 ± 0.15	0.20 ± 0.13
12	0.90 ± 0.36	1.30 ± 0.53	1.50 ± 0.62

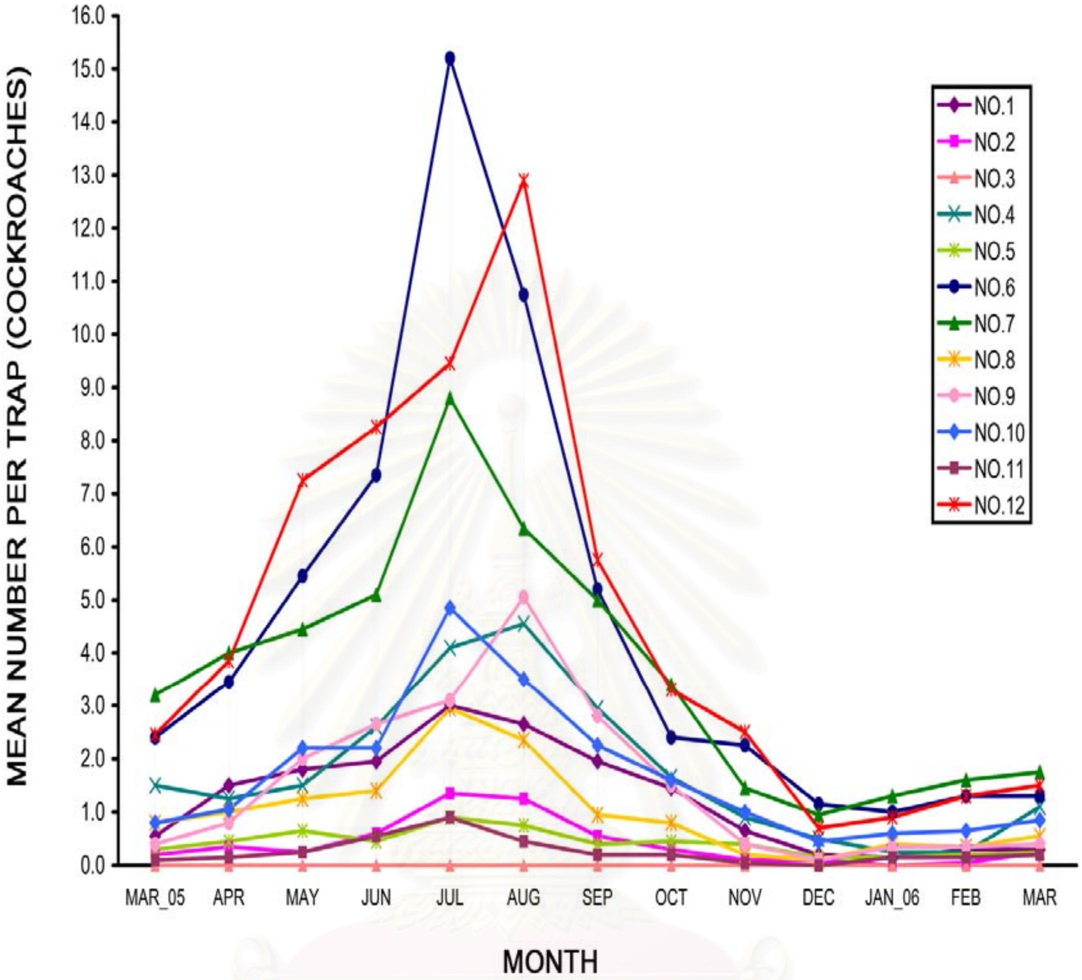

Figure 4.12 The curves of means number of the German cockroaches caught per trap from twelve Bangkok market. Each point on a curve represents a mean number (per trap) in each month during the study period.

The results in Table 4.8 showed that the overall mean numbers (per trap) of the German cockroaches caught in twelve Bangkok markets were significantly different (F = 21.771; df = 11, 3107; $p < 0.0001$).

The highest mean was in market number 12 (4.62 ± 0.68) followed by market number 6 (4.55 ± 0.72), market number 7 (3.64 ± 0.48), market number 4 (1.77 ± 0.23), market number 10 (1.69 ± 0.23), market number 9 (1.53 ± 0.25), market number 1 (1.27 ± 0.17), market number 8 (1.01 ± 0.15), market number 5 (0.42 ± 0.08), market number 2 (0.41 ± 0.07), market number 11 (0.26 ± 0.05), respectively whereas the lowest was in market number 3 (0.00 ± 0.00). Although the mean numbers of the German cockroaches caught per trap in market number 12 and market number 6 were not significantly different, the mean number in market number 12 was significantly higher than any other markets (LSD; df = 3107; $p < 0.05$) (Table 4.8; Figure 4.13).

Table 4.8 Comparison of mean numbers[1] (± SE) of the German cockroaches caught per trap from twelve Bangkok markets from March 2005 to March 2006.

Markets	**Mean numbers per trap**
Number 1	1.27 ± 0.17^{cd}
Number 2	0.41 ± 0.07^{abc}
Number 3	0.00 ± 0.00^{a}
Number 4	1.77 ± 0.23^{d}
Number 5	0.42 ± 0.08^{abc}
Number 6	4.55 ± 0.72^{ef}
Number 7	3.64 ± 0.48^{e}
Number 8	1.01 ± 0.15^{bcd}
Number 9	1.53 ± 0.25^{d}
Number 10	1.69 ± 0.23^{d}
Number 11	0.26 ± 0.05^{ab}
Number 12	4.62 ± 0.68^{f}

[1] Means with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

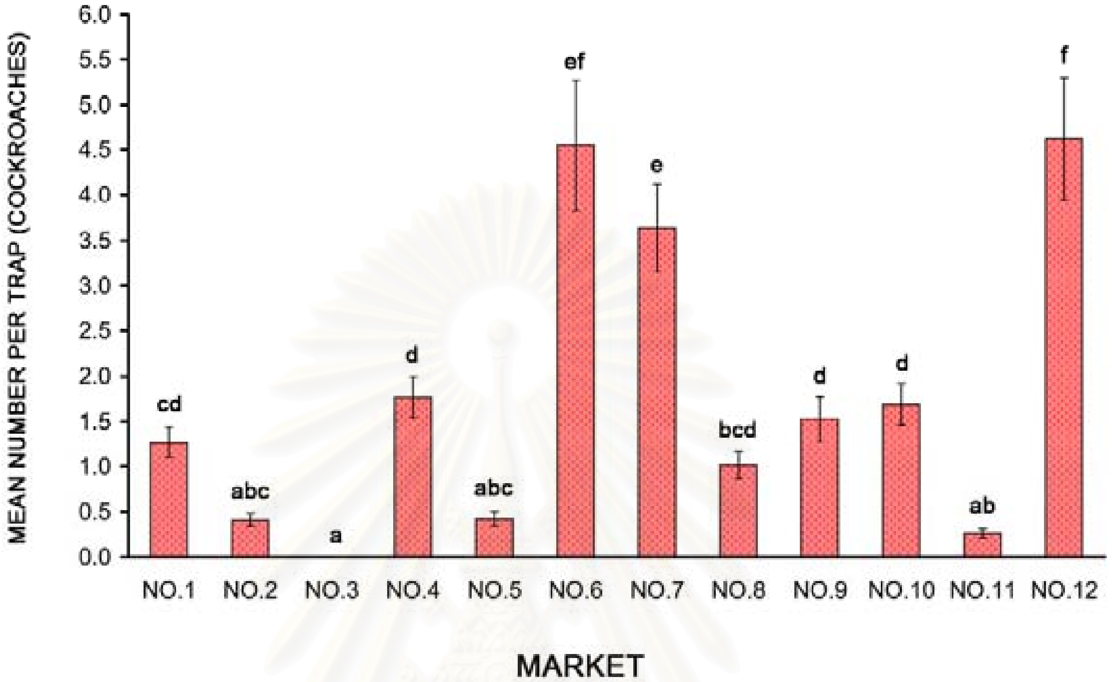

Figure 4.13 Comparison of mean numbers (± SE) of the German cockroaches caught per trap from twelve markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Comparisons of the German cockroach numbers in the 4 combination groups of Bangkok markets

Table 4.9 presented the overall average numbers of the German cockroach caught per trap from 4 combination groups of Bangkok markets from March 2005 to March 2006. Mean numbers of the German cockroaches were significantly different during the study period ($F = 14.791$; df = 3, 3115; $p < 0.0001$).

The highest catch was in the combination of market class 1/ low population density zone (2.22 ± 0.26) followed by market class 2/ low population density zone (2.19 ± 0.25), market class 2/ high population density zone (2.07 ± 0.19), and the lowest was in market class 1/ high population density zone (0.56 ± 0.07).

The mean numbers in the combination of market class 1/ low population density zone (2.22 ± 0.26), market class 2/ low population density zone (2.19 ± 0.25), and market class 2/ high population density zone (2.07 ± 0.19) were not significantly different, the mean number in the combination of market class 1/ high population density zone (0.56 ± 0.07) was significantly lower than any other group of markets (LSD; df = 3115; $p < 0.05$).

Table 4.9 Mean (± SE) number of the German cockroaches caught per trap from 4 combination groups of Bangkok market from March 2005 to March 2006 (S = small nymph, M = medium nymph, L= large nymph, A = adult).

Stages	Combination groups of Bangkok markets [1]			
	HC1	**LC1**	**HC2**	**LC2**
S	0.44 ± 0.08	0.24 ± 0.04	0.15 ± 0.02	0.18 ± 0.02
M	0.11 ± 0.02	0.49 ± 0.07	0.40 ± 0.04	0.45 ± 0.06
L	0.22 ± 0.03	0.80 ± 0.09	0.75 ± 0.07	0.82 ± 0.09
A	0.19 ± 0.02	0.69 ± 0.07	0.76 ± 0.07	0.74 ± 0.08
Overall mean (per trap) [2]	**0.56 ± 0.07** [a]	**2.22 ± 0.26** [b]	**2.07 ± 0.19** [b]	**2.19 ± 0.25** [b]

[1] HC1 = the combination of market class 1/ high population density zone, LC1 = the combination of market class 1/ low population density zone, HC2 = the combination of market class 2/ high population density zone, and LC2 = the combination of market class 2/ low population density zone.

[2] Means with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Comparisons of the German cockroach numbers in the 2 classes of Bangkok markets

The results in Table 4.10 presented the average numbers of the German cockroaches caught per trap from 2 classes of Bangkok markets from March 2005 to March 2006. Mean numbers of the German cockroaches caught from class 1 market (1.41 ± 0.33) was not significantly different from class 2 market (2.12 ± 0.44) (F = 1.709; df = 3118; p = 0.204).

The highest mean in class 1 market was in July, followed by August, June, September, May, April, October, March 2005, November, March 2006, February, December and, January, respectively, while the highest mean in class 2 market was highest in August, followed by July, June, May, September, April, October, March 2005, November, March 2006, February, January, and December, respectively (t-test; $p < 0.05$) (Figure 4.14).

Table 4.10 Mean (± SE) number of the German cockroaches caught per trap from two classes of Bangkok markets from March 2005 to March 2006.

Months	Classes of market	
	Class 1	Class 2
March 2005	0.83 ± 0.38	1.29 ± 0.50
April	1.17 ± 0.51	1.81 ± 0.62
May	1.61 ± 0.81	2.90 ± 1.03
June	2.16 ± 1.11	3.36 ± 1.16
July	4.09 ± 1.30	5.01 ± 1.40
August	3.33 ± 1.42	5.10 ± 1.47
September	1.84 ± 0.80	2.83 ± 0.89
October	1.04 ± 0.38	1.80 ± 0.53
November	0.72 ± 0.33	0.93 ± 0.37
December	0.35 ± 0.17	0.38 ± 0.15
January 2006	0.26 ± 0.15	0.62 ± 0.17
February	0.35 ± 0.19	0.73 ± 0.23
March	0.53 ± 0.21	0.88 ± 0.25
Overall mean (per trap) [1]	**1.41 ± 0.33 [a]**	**2.12 ± 0.44 [a]**

[1] Means with different letters in the same row are significantly different at $p < 0.05$, *t*-test.

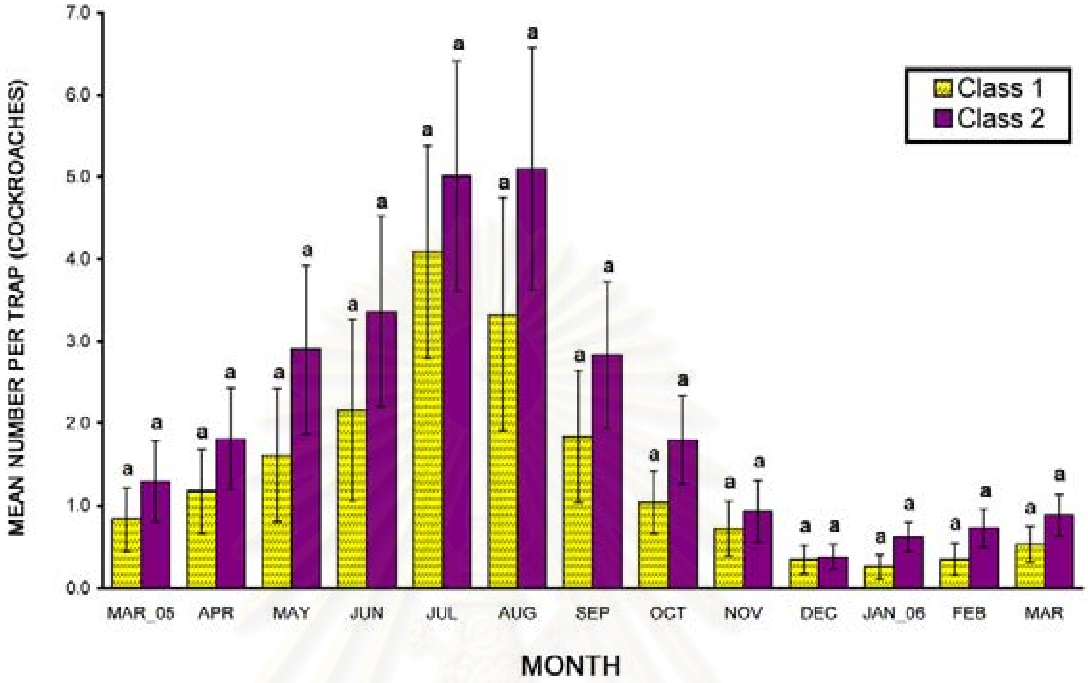

Figure 4.14 Comparison of average numbers (± SE) of the German cockroaches caught per trap between two classes of Bangkok markets: class 1 and class 2. The bar graphs with the different letters are significantly different at $p < 0.05$, *t*-test.

Comparisons of the German cockroach numbers in the two differences of human population density zones

The results in Table 4.11 presented the average numbers of the German cockroaches caught per trap from 2 differences of human population density zone from March 2005 to March 2006. Overall, the mean number of the German cockroaches caught per trap in the high population density zone (1.31 ± 0.26) was significantly different from the mean in the low population density zone (2.22 ± 0.25). (F = 4.820; df = 3118; p = 0.038).

The highest mean in the high population density zone was in July, followed by August, June, September, May, April, October, March 2005, March 2006, November, February, January, and December, respectively. The highest mean in low population density zone was in July, followed by August, June, May, September, April, October, March 2005, November, March 2006, February, January, and December, respectively (t-test; $p < 0.05$).

Although the overall mean numbers of cockroaches in the high population density zone was significantly different from the low population density zone, the means between the two zones in each month were not statistically different at $p < 0.05$, t-test (Figure 4.15).

Table 4.11 Mean (± SE) number of the German cockroaches caught per trap from Bangkok markets in two differences of human population density zones from March 2005 to March 2006.

Months	Human population density zones	
	High	Low
March 2005	0.86 ± 0.48 [a]	1.26 ± 0.41 [a]
April	1.28 ± 0.58 [a]	1.70 ± 0.63 [a]
May	1.63 ± 0.65 [a]	2.88 ± 1.15 [a]
June	1.95 ± 0.73 [a]	3.57 ± 1.38 [a]
July	3.20 ± 1.22 [a]	5.90 ± 1.62 [a]
August	2.94 ± 0.96 [a]	5.48 ± 1.12 [a]
September	1.88 ± 0.74 [a]	2.79 ± 0.95 [a]
October	1.24 ± 0.49 [a]	1.60 ± 0.47 [a]
November	0.47 ± 0.21 [a]	1.18 ± 0.30 [a]
December	0.24 ± 0.14 [a]	0.49 ± 0.16 [a]
January 2006	0.37 ± 0.19 [a]	0.51 ± 0.15 [a]
February	0.44 ± 0.23 [a]	0.64 ± 0.22 [a]
March	0.54 ± 0.25 [a]	0.87 ± 0.22 [a]
Overall mean (per trap) [1]	**1.31 ± 0.26 [a]**	**2.22 ± 0.25 [b]**

[1] Means with different letters in the same row are significantly different at $p < 0.05$, *t*-test.

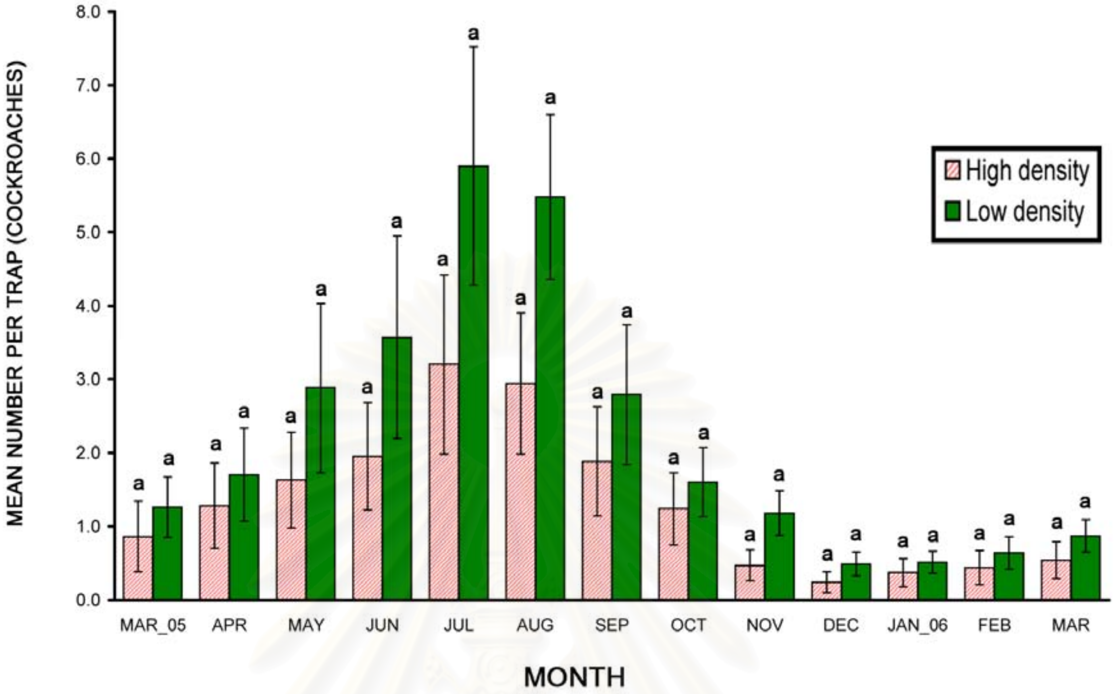

Figure 4.15 Comparison of average numbers (± SE) of the German cockroaches caught per trap from Bangkok markets in two differences of human population density zones. The bar graphs with the different letters are significantly different at $p < 0.05$, *t*-test.

Comparisons of the German cockroach numbers in the different kinds of shops in the Bangkok markets

The results in Table 4.12 presented the average numbers of the German cockroaches in 3 kinds of shops in the Bangkok markets. Overall, the mean numbers of the German cockroaches caught per trap were significantly different during the study period (F = 76.36; df = 2, 465; p < 0.0001).

The highest mean was in groceries (3.02 ± 0.19) followed by vegetable shops (1.52 ± 0.13) and butcher shops (0.61 ± 0.04), respectively. In November, the mean numbers in grocery (1.19 ± 0.09) and vegetable shops (0.67 ± 0.05) were not significantly different. However, the mean numbers in groceries were significantly higher than vegetable and butcher shops in the rest of the months (LSD; df = 465; p < 0.05) (Figure 4.16).

Table 4.12 Mean (± SE) number (per trap) of the German cockroaches caught per trap in three kinds of shop in Bangkok markets from March 2005 to March 2006 (B = butcher shop, V = vegetable shop, and G = grocery).

Months	B	V	G
March 2005	0.58 ± 0.04 [a]	0.86 ± 0.06 [a]	1.67 ± 0.12 [b]
April	0.36 ± 0.02 [a]	0.79 ± 0.06 [a]	3.15 ± 0.24 [b]
May	1.03 ± 0.07 [a]	1.65 ± 0.12 [a]	3.90 ± 0.30 [b]
June	0.60 ± 0.04 [a]	1.39 ± 0.10 [a]	5.98 ± 0.46 [b]
July	1.49 ± 0.11 [a]	5.07 ± 0.39 [b]	6.65 ± 0.51 [c]
August	1.15 ± 0.08 [a]	4.56 ± 0.35 [b]	6.49 ± 0.49 [c]
September	0.93 ± 0.07 [a]	1.24 ± 0.09 [a]	4.63 ± 0.35 [b]
October	0.43 ± 0.03 [a]	1.43 ± 0.11 [b]	2.26 ± 0.17 [c]
November	0.58 ± 0.04 [a]	0.67 ± 0.05 [b]	1.19 ± 0.09 [b]
December	0.04 ± 0.00 [a]	0.30 ± 0.02 [b]	0.71 ± 0.05 [c]
January 2006	0.19 ± 0.01 [a]	0.40 ± 0.03 [b]	0.68 ± 0.05 [c]
February	0.21 ± 0.01 [a]	0.58 ± 0.04 [b]	0.79 ± 0.06 [c]
March	0.29 ± 0.02 [a]	0.60 ± 0.04 [b]	1.17 ± 0.09 [c]
Overall mean (per trap) [1]	**0.61 ± 0.04 [a]**	**1.52 ± 0.13 [b]**	**3.02 ± 0.19 [c]**

[1] Means with different letters in the same row are significantly different at $p < 0.05$, ANOVA with LSD.

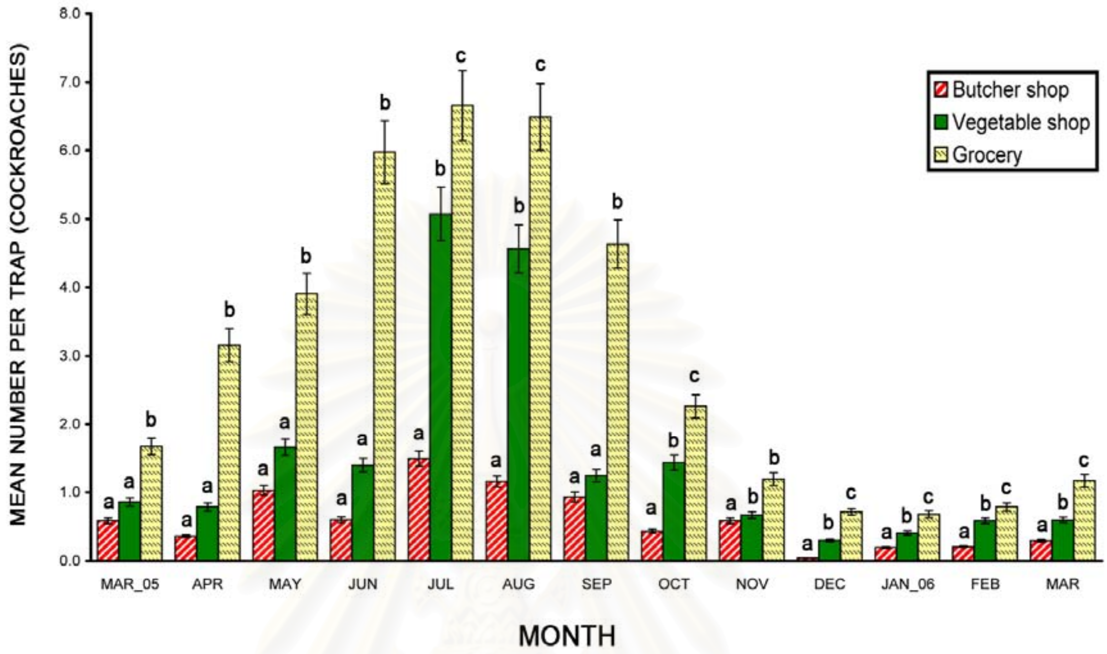

Figure 4.16 Comparison of average numbers (± SE) of the German cockroaches caught per trap from 3 kinds of shops in twelve Bangkok markets. The bar graphs with the different letters are significantly different at $p < 0.05$, ANOVA with LSD.

Discussion and Conclusion

Several important conclusions may be derived from the data such as the species of cockroaches found in those markets and population dynamics of the German cockroach in those markets. The most distinguished result was the difference numbers of the German cockroaches in each month. They were contrarily significant among the months throughout the year.

A total of 11,944 cockroaches were trapped from the twelve Bangkok markets. They consisted of seven species, belonging to six genera, such as *Blattella germanica*, *Nauphoeta cinerea*, *Neostylopyga rhombiofolia*, *Periplaneta americana*, *P. brunnea*, *Supella longipalpa*, and *Symploce pallens*. Petchburi market (market number 7) showed the highest species number of cockroach species (7 species) whereas Eamsombat market (market number 6) showed the lowest species number of cockroach species (2 species). The German cockroach, *B. germanica*, were caught in the largest number (46.12 %), followed by *P. americana* (45.30 %). This result is similar to those reported earlier by Lee *et al*. (1993), Benjapong *et al.* (1997), Tawatsin *et al.* (2001), Sriwichai (2001), Damsuwon (2003), Lee *et al*. (2003), and Pai *et al.* (2005). The former reports showed that the *B. germanica* and *P. americana* were dominant cockroach species in urban environments in Malaysia, Republic of Korea, Taiwan, and Thailand. Additionally, this is the first record of *Sy. pallens* in Thailand. This species had only been recently reported as a domiciliary pest (Lee and Lee, 2000) and it has been reported as a new domiciliary pest in Malaysia (Lee *et al*., 2000).

During the 13-month-study, the German cockroach were abundant throughout the year may be because of its short life cycle leading to rapid population recovery (Ross and Mullins, 1995). The other causation were the proper temperature (average degree of temperature: minimum 26.02 ± 0.11 °C; maximum 35.17 ± 0.02 °C), the suitable humidity (average percentage of humidity: minimum 59.00 ± 1.48 % RH; maximum 68.75 ± 0.97 % RH), suitable harborage, and the presence of food and water in the markets. The two highest mean number peaks of the German cockroaches caught per trap were in July and August whereas the two lowest peaks were in December and January. The German cockroaches found in July and August in a large number indicated that the German cockroaches had high reproductive activity. On the other hand, the small number found in December and January suggests that the German cockroaches had low reproductive activity of in these months. The temperature and relative humidity may be the important factors influencing on the cockroach reproduction (Ross and Mullins, 1995).

In this study, the large nymphs of the German cockroach were trapped in the highest number followed by adults, medium nymphs, and small nymphs. The large nymphs and the adults indicated that they had high foraging activity (Cloarec and Rivault, 1991; Ross and Mullins, 1995). The adult males had more foraging activity than the females and the females with oothecae, respectively (Metzger, 1995). While the small nymphs were found in a small number implied that they had the less foraging activity than the other stage of the German cockroaches (Cloarec and Rivault, 1991; Ross and Mullins, 1995). In addition, the low weight of the small nymphs might be another reason for the lowest number trapped. It might be because of the small nymphs escaped from the slippery inner surface of the jar traps (Wang and Bennett, 2006).

The nymphal stages were found in the highest number in the population of Bangkok markets, this result was agreed with the prior studies (Owens and Bennett, 1983; Ross and Muliins, 1995). However, the nymphals were found at 66.19 % of the German cockroach population in the field of this study while the nymphal stages of populations growing under optimal conditions in laboratory were found at 80 % (Ross and Mullins, 1995). It may be because of the external factors including pathogens, predators, and physical environments.

The Bang Kho Laem market (market number 3) was classified in class 1 market while the Ladprao 123 market (market number 12) was classified in class 2 market. In this study, the German cockroaches found from market number 12 in the largest number whereas none of the German cockroaches was caught from the market number 3. The suitable temperature, proper humidity, poor sanitation, infrequent cleanliness, and the old structure which make several cracks and crevices of the market number 12 may be the reason of the high population of the German cockroach in this market. In contrary, none of the German cockroaches was caught in market number 3 may be because of the unsuitable humidity, the occurrence of predator such as cat, and the good sanitary practice of the vendor such as clean and clear the material on the shelf after finish their work in everyday. Although none of the German cockroaches was caught, but the other species of cockroaches were caught in this market such as *Na. cinerea, Ne. rhombiofolia*, *P. americana*, *P. brunnea*, and *Sy. pallens*. It might be because of *P. americana*, and *P. brunnea* usually inhabit in the more moist condition than the German cockroaches (James and Harwood, 1969; Sriwichai, 2001; Damsuwon, 2003).

The mean numbers of the German cockroaches caught in 4 combination groups of markets were statistically different. The mean numbers between the high and low population density zones were also statistically different. However, the mean numbers of the German cockroaches caught in class 1 market and class 2 market were not statistically different. The German cockroach proliferates in both classes of market and both zones of human population density in Bangkok may be because of the proper temperatures and humidity, the presence of food and water, and the good harborage in these markets. Moreover, the other causations were the short life cycle of this cockroach species, its parental care behavior, and the nocturnal behavior (Ross and Mullins, 1995).

The catch number of the German cockroaches in the grocery was statistically higher than the vegetable shop and butcher shop. It may be because of in the grocery has many crevices and corners for their harborage. The infrequently cleanliness was the other reason that a large number of the German cockroaches were caught in this shop. From the observation, when the butcher shop owners finished their work, they collected their meat and cleaned their shop daily. The vegetable shop owners also collected their vegetable from shelf, they cleaned their shop sometimes. However, the grocery shop owners also collected their goods, covered their shelf with plastic sheets but they cleaned their shop infrequently. It is similar to the previous report by Appel (1995) that the German cockroach preferred a warm microhabitat and dark harborage for their shelter.

In conclusion, the German cockroaches were found in various types of places in the markets. They were abundant throughout the years in these markets may be because of these markets had a lot of food and water, proper temperatures, suitable humidity, good harborage, and infrequently cleanliness. Thus, the German cockroach control in the Bangkok markets should be done integration between the sanitary control, such as removal the food supply and eliminate the shelter of the cockroaches, and chemical control measures (Gold, 1995; Koehler *et al*., 1995).The chemical control measures should be done at night because the German cockroaches are nocturnal insects (Ross and Mullins, 1995). Moreover, the effective chemical control should be applied in December and January because of the two lowest peaks of this cockroach species showed in these two months.

CHAPTER 5

BACTERIAL HARBORAGE OF THE GERMAN COCKROACH, *Blattella germanica* (Linnaeus), AND THE AMERICAN COCKROACH, *Periplaneta americana* (Linnaeus), IN BANGKOK MARKETS

Introduction and Objective

In Thailand many household cockroaches found are both indoor and peridomestic species. At least ten species of the cockroaches have been reported, but only some species had medical significance such as the German cockroach, *Blattella germanica,* and the American cockroach, *Periplaneta americana* (Tawatsin *et al*., 2001). Cockroaches are likely to be encountered in environments which provide favorable environmental conditions and a ready source of food (Fotedar *et al*., 1991). Cockroaches always carry bacterial species in the environment where they live, but these bacterial species have no harm on the cockroach (Devi and Murray, 1991; Rivault *et al*., 1993). Their omnivorous habits of feeding and indiscriminate deposition of fecal material make them ideal agents for the transmission of microorganisms (Fotedar, Banerjee, Shriniwas, 1993). Although the mechanical transmission of pathogens has received considerable attention among researchers, few studies have been reported that the bacteria could survive in cockroach gut such as *Pseudomonas aeruginosa* could survive in the German cockroach gut for 114 days (Fotedar *et al*., 1993) and *Staphylococcus aureus* could survive in the German cockroach gut for 14 days (Tachbele *et al*., 2006). Thus, the cockroaches could be also report as a biological transmission vector (Fotedar *et al*., 1993; Tachbele *et al*., 2006).

Food consumers have suffered from many diseases from bacteria especially food-borne bacterial illness (Tachbele *et al*., 2006). In 2005, ill rate of diarrhea, food poisoning, enteric fever, typhoid, and bacterial dysentery in Thai people were 1837.07, 226.62, 16.98, 8.08, and 5.75 per 100,000 persons, respectively (Public Health, Ministry, 2006). One possible source of food contaminations could be disseminated the pathogens to foods and utensil of catering centers through small animals such as cockroaches that live closely with humans in urban environments (Burgess and Chetwyn, 1981). Several investigations around the world revealed that cockroaches living close to human dwellings were important carriers of etiologic agents belonging to all groups of potential pathogens such as viruses, bacteria, protozoa, fungi and helminthes (Cruden and Markovetz, 1987; Agbodaze and Owusu, 1989; Fotedar *et al*., 1991; Cloarec *et al*., 1992; Rivault *et al*., 1993; Tachbele *et al*., 2006). Over 100 species of bacteria have been isolated from any parts of many household cockroach species (Cruden and Markovetz, 1987). However, some species of bacteria have been reported as normal flora in the cockroaches such as *Citrobacter*, *Enterobacter*, *Klebsiella* and *Serratia* (Le Guyader, Rivault, and Chaperson, 1989).

According to the studies of Fotedar *et al*. (1989), almost all cockroaches isolated from Indian hospital and residential areas carried medically important microorganisms. Although *Psuedomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus faecalis* have been isolated from *B. germanica* collected from hospital, these species were not found in the corresponding residential areas. In Bangladesh, *Salmonella*, *Shigella*, *S. aureus*, *Bacillus cereus*, and *Escherichia coli* were isolated from cockroaches (Paul, Kham, and Muhibullah, 1992). In Malaysia, Oothuman *et al*. (1989) isolated *Shigella boydii*, *S. dysenteriae*, and *Salmonella typhimurium* from cockroaches collected from hospital kitchens. Vythilingam *et al*. (1997) isolated 17 species of bacteria from the *Periplaneta americana* in Malaysia. Additionally, many food-borne pathogens were isolated from cockroaches collected from Nigeria (Umunnabuike and Irokamulo, 1986) and in Ghana (Agbodaze and Owusu, 1989). Fifty-six species of bacteria were isolated from cockroaches caught in many residential areas in France (Rivault *et al*., 1993).

Fathpour, Emtiazi, and Ghasemi (2003) reported that 70% of cockroaches collected from hospitals in Iran carried *Salmonella* spp., and some of the isolates were resistant to antimicrobial drugs. However, Devi and Murray (1991) reported that over 4% of cockroaches collected from hospitals, houses, animal sheds, grocery stores, and restaurants in India harbored multiple drug resistant *Salmonella*.

From the previous reports, the German and the American cockroaches were carried medically important bacteria. Prior to this study, little documentation existed as to the bacteria carried by cockroaches in public market places. The objective of this study was to isolate prevalent and human pathogenic bacteria of the German and the American cockroaches in Bangkok markets. The species richness and the similarity coefficient of the bacteria isolated from both kinds of cockroaches, and from Bangkok markets which have different sanitary practices were also investigated.

Material and Methods

1. Selection of the study areas and cockroach collection

By the combination of human population density zones and the classes of Bangkok markets, 4 combination groups such as high population density zone/ market class 1, high population density zone/ market class 2, low population density zone/ market class 1, and low population density zone/ market class 2 were categorized, and the numbers of the markets in each combination group are showed in Table 4.1. In this study, 12 market sites (3 sites for each combination group in

Bangkok) were selected using random sampling method (Krebs, 1999), and the market's names were showed in Table 4.2.

In April, July 2005 and January 2006, the German and the American cockroaches were collected from twelve Bangkok markets using the modified jar traps. The traps were placed randomly in 3 kinds of shops in each market such as butcher shops (fresh pork, beef, and chicken), vegetable shops (fresh vegetable), and groceries (rice, seasoning, garlic, red onion, dried shrimps, and dried squids). The traps were placed next to walls and near equipments in each shop for 24 hours. Only the alive cockroaches caught were considered in this study. Identification of cockroaches was performed in accordance with Asahina (1983).

2. Bacterial isolation

The German and the American cockroaches caught from each market were separately collected and brought them to the laboratory. Then, twenty cockroaches in each group were sacrificed in the sterile jars using diethyl ether soaked cotton (Devi and Murray, 1991; Rivault *et al*., 1993). Only aerobic and facultative anaerobic bacteria were investigated (Oothuman *et al*., 1989; Devi and Murray, 1991; Rivault *et al*., 1993; Vythilingam *et al*., 1997). Bacteria were isolated from 2 parts of the German and the American cockroaches such as the external cuticle and the middle gut.

2.1 Isolation and identification of bacteria from the external cuticle

The cockroaches in each group were placed in the sterile bottles. Five ml of sterile normal saline (0.85%) was added to the bottles, and the cockroach was thoroughly shaken for 2 min. A loopful of the fixed volume of the washing was cultured separately on Nutrient (NA) agar (Difco™, Detroit, USA), Blood (BA) agar (Difco™, Detroit, USA), MacConkey (MA) agar (Difco™, Detroit, USA), Eosin

Methylene Blue (EMB) agar (Difco™, Detroit, USA), Deoxycholate Citrate (DCA) agar (Difco™, Detroit, USA), Salmonella Shigella (SS) agar (Difco™, Detroit, USA), Xylose Lysine Deoxycholate (XLD) agar (Oxoid™, Basingstoke, UK) and Thiosulfate Citrate Bile Salt Sucrose (TCBS) agar (Difco™, Detroit, USA). Then, the culture plates were incubated at 37 °C for 18-24 hr (Fotedar *et al*., 1989, 1991, 1993; Rivault *et al*., 1993; Vythilingam *et al*., 1997; Tachbele *et al*., 2006). Bacterial colonies were identified by macroscopic examination, microscopic examination, Gram staining, and biochemical tests according to Cowan and Steel's standard bacteriological procedures (Barrow and Feltham, 1993). In addition, API®-20E test kit (bioMérieux, Marcy-l'Etoile, France) was used to confirm the Enterobacteriaceae group.

2.2 Isolation and identification of bacteria from the middle gut

After isolating bacteria from the external cuticles, the cockroaches in each group were washed in 70 % ethyl alcohol for 5 min (to decontaminate external surfaces as 70 % ethyl alcohol is bactericidal) and let them to dry at room temperature under sterile conditions. Cockroaches were re-washed with sterile normal saline for 2 min to remove traces of alcohol. Legs and wings were removed, and the middle gut (Figure 5.1) was aseptically dissected out using autoclave-sterilized entomological dissecting needles under a stereo microscope. The instruments were dipped in 70% ethyl alcohol and flamed between dissections. The removed middle gut was then homogenized in 2 ml of sterile normal saline water. The emulsion (1 ml for each) was transferred into 2 ml of sterile peptone water. A loopful of the suspension was then cultured separately on Nutrient (NA) agar (Difco™, Detroit, USA), Blood (BA) agar (Difco™, Detroit, USA), MacConkey (MA) agar (Difco™, Detroit, USA), Eosin Methylene Blue (EMB) agar (Difco™, Detroit, USA), Deoxycholate Citrate (DCA) agar (Difco™, Detroit, USA), Salmonella Shigella (SS) agar (Difco™, Detroit, USA), Xylose Lysine Deoxycholate (XLD) agar (Oxoid™, Basingstoke, UK) and Thiosulfate Citrate Bile Salt Sucrose (TCBS) agar (Difco™, Detroit, USA). Then, the culture plates were incubated at 37 °C for 18-24 hr (Fotedar *et al*., 1989, 1991, 1993; Rivault *et al*., 1993; Vythilingam *et al*., 1997; Tachbele *et al*., 2006). Bacterial colonies were identified by macroscopic examination, microscopic examination, Gram staining, and biochemical tests according to Cowan and Steel's standard bacteriological procedures (Barrow and Feltham, 1993). Moreover, API®-20E test kit (bioMérieux, Marcy-l'Etoile, France) was used to confirm the Enterobacteriaceae group.

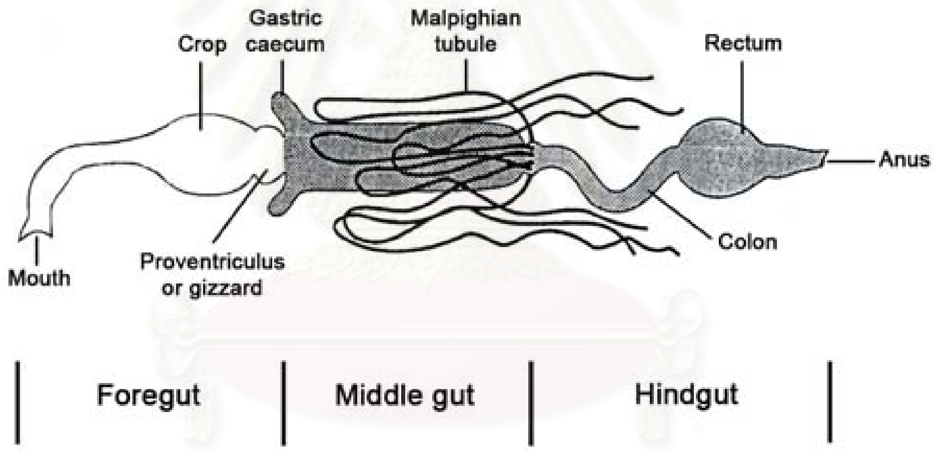

Figure 5.1 The alimentary tract of the cockroach (modified from Appel, 1995).

Data analysis

The species richness and the similarity coefficient (Coefficient of Sorensen) of the bacteria isolated from both kinds of cockroaches were investigated. Additionally, the similarity coefficient (Krebs, 1999) for indicating the similarity of bacterial species was used as following:

$$\text{Coefficient of Sorensen } (S_s) = \frac{2a}{2a+b+c}$$

When a = Number of species in sample A and sample B (joint occurrences)

b = Number of species in sample B but not in sample A

c = Number of species in sample A but not in sample B

Comparisons of species richness of bacteria between cockroach species, collection sites, and study periods were analyzed by ANOVA with LSD analysis. Significant differences were at the $p < 0.05$ level.

Results

I. Species richness of bacteria in the cockroaches

Species richness of bacteria in the German and the American cockroaches in twelve Bangkok markets

A total of twenty-six species of bacteria was isolated from the German and the American cockroaches in twelve Bangkok markets. The result in Table 5.1 presented bacterial species isolated from the German and the American cockroaches from twelve Bangkok market in April, July 2005, and January 2006. However, none of the German cockroach was caught in market number 3 so that the species richness of bacteria could not determine.

Table 5.1 Bacterial species isolated from external cuticle and middle gut of the German and the American cockroaches in twelve Bangkok market (Apr = April, Jul = July, and Jan = January).

Aerobic Bacteria	Markets																																			
	1			2			3			4			5			6			7			8			9			10			11			12		
	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan	Apr	Jul	Jan
Bacillus cereus	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Citrobacter braakii	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Citrobacter freundii	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Corynebacterium pseudotuberculosis	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	◇	◇	◇	◇	◇	◇	◇	◇	◇	□	□	◇	□	□	□	◇	◇	◇
Enterobacter aerogens	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Enterobacter cloacae	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Enterobacter gergoviae	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Enterobacter sakazakii	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Enterococcus durans	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Escherichia coli	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Escherichia fergusonii	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	◇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Klebsiella ozaenae	■	■	■	□	■	■	●	●	●	■	◇	■	■	◇	■	■	◇	■	■	■	■	■	■	■	■	■	◇	■	■	■	■	■	■	■	■	■
Listeria monocytogenes	◇	◇	□	□	□	□	●	●	□	□	□	□	□	□	□	□	□	□	◇	◇	◇	◇	◇	□	□	◇	□	□	◇	□	◇	◇	□	◇	◇	□
Morganella morganii	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Proteus mirabilis	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Pseudomonas aeruginosa	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	◇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Salmonella arizona	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	■	■	□	■	□	□	■	■	□	◇	◇	□	◇	◇	□	■	■	□
Serratia marcescens	■	■	■	□	■	□	●	●	●	■	◇	◇	■	◇	◇	■	■	◇	■	■	◇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Staphylococcus aureus	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Staphylococcus epidermidis	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Staphylococcus saprophyticus	■	■	■	■	■	■	●	●	●	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Streptococcus pneumoniae	□	◇	◇	□	◇	◇	□	□	□	□	□	□	□	◇	◇	□	□	◇	□	◇	◇	□	◇	◇	□	◇	◇	□	◇	◇	◇	◇	◇	◇	◇	◇
Streptococcus pyogenes	■	■	■	■	■	◇	●	●	●	■	■	◇	■	■	◇	■	■	◇	■	■	◇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
Vibrio parahemolyticus	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	◇	□	□	◇	□	□	□	□	□	◇	□	□	◇	□	□	◇	□	□
Viridans Streptococci	□	■	◇	□	■	■	□	●	●	□	■	■	□	■	■	□	■	■	□	■	■	□	■	■	□	■	■	□	■	■	◇	■	■	◇	■	■
Yersinia enterocolotica	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	□	◇	□	◇	◇	□	◇	□	□	□	◇	◇	□	◇	◇	◇	□	◇	◇

Both German and American cockroaches present

■ : isolated from both cockroaches

◇ : isolated from American cockroach only

□ : could not be isolated

Only American cockroach present

● : isolated from American cockroach

In this study, the German cockroach hosted 21 species of bacteria whereas the American cockroach hosted 26 species. The external cuticle and middle gut samples yielded 25 and 22 species of bacteria, respectively. In the German cockroach, from the external cuticle could be isolated 21species of bacteria whereas in the middle gut could be isolated 18 species. While in the American cockroach, from external cuticle could be isolated 25 species of bacteria whereas in the middle gut of them could be isolated 22 species of bacteria.

Seven species of Gram-positive cocci were isolated from the American cockroaches and only 6 species from the German cockroaches (Table 5.2).

Table 5.2 Gram-positive cocci isolated from the German and the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	German cockroach		American cockroach	
	External cuticle	Middle gut	External cuticle	Middle gut
Enterococcus durans *	√	√	√	√
Staphylococcus aureus *	√	√	√	√
Staphylococcus epidermidis	√		√	
Staphylococcus saprophyticus	√		√	
Streptococcus pneumoniae *			√	
Streptococcus pyogenes *	√	√	√	√
Viridans Streptococci *	√		√	

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

Three species of Gram-positive rods were isolated from the American cockroaches and only 1 species from the German cockroach (Table 5.3).

Table 5.3 Gram-positive rods isolated from the German and the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	**German cockroach**		**American cockroach**	
	External cuticle	**Middle gut**	**External cuticle**	**Middle gut**
Bacillus cereus *	√	√	√	√
Corynebacterium pseudotuberculosis *			√	√
Listeria monocytogenes *			√	√

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

Ten species of Gram-negative Enterobacteriaceae species (coliform bacteria) were isolated from the two species of cockroaches (Table 5.4).

Table 5.4 Gram-negative Enterobacteriaceae species (coliform bacteria) isolated from the German and the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	**German cockroach**		**American cockroach**	
	External cuticle	**Middle gut**	**External cuticle**	**Middle gut**
Citrobacter braakii	√	√	√	√
Citrobacter freundii *	√	√	√	√
Enterobacter aerogens *	√	√	√	√
Enterobacter cloacae *	√	√	√	√
Enterobacter gergoviae	√	√	√	√
Enterobacter sakazakii	√	√	√	√
Escherichia coli *	√	√	√	√
Escherichia fergusonii	√	√	√	√
Klebsiella ozaenae *	√	√	√	√
Serratia marcescens *	√	√	√	√

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

Four species of Gram-negative Enterobacteriaceae species (non-coliform bacteria) were isolated from the American cockroaches and only 3 species from the German cockroaches (Table 5.5).

Table 5.5 Gram-negative Enterobacteriaceae species (non-coliform bacteria) isolated from the German and the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	**German cockroach**		**American cockroach**	
	External cuticle	**Middle gut**	**External cuticle**	**Middle gut**
Morganella morganli	√	√	√	√
Proteus mirabilis *	√	√	√	√
Salmonella arizona *	√	√	√	√
Yersinia enterocolitica *				√

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

One species of glucose fermenter bacilli (oxidase positive) was isolated from the American cockroach, but none of this bacterial group isolated from the German cockroach (Table 5.6).

Table 5.6 Glucose fermenter bacilli (oxidase positive) isolated from the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	German cockroach		American cockroach	
	External cuticle	Middle gut	External cuticle	Middle gut
Vibrio parahemolyticus *			√	√

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

One species of glucose non-fermenter bacilli was isolated from the German and the American cockroaches (Table 5.7).

Table 5.7 Glucose non-fermenter bacilli isolated from the German and the American cockroaches caught from twelve Bangkok markets in April, July 2005, and January 2006 (√ = could be isolated from the cockroach).

Bacteria	**German cockroach**		**American cockroach**	
	External cuticle	**Middle gut**	**External cuticle**	**Middle gut**
Pseudomonas aeruginosa *	√	√	√	√

* may be pathogenic species according to Stewart and Beswick (1977); Rivault *et al*. (1993); Mahon and Manuselis (2000); Suwanpinit (2004).

Mean numbers of the species richness of bacteria carried by both kinds of cockroaches were significantly different during the study period ($F = 12.41$; df = 2, 68; $p = 0.001$). Overall mean (± SE) numbers of the species richness of bacteria carried by the German cockroach (19.21 ± 0.21) was statistically different from the American cockroach (21.72 ± 0.34).

The highest mean of species richness of bacteria harbored by the German cockroach was found in the market number 12 (20.33 ± 0.33) whereas the lowest was found in market number 5 (17.67 ± 0.67). The highest mean of bacterial species harbored by the American cockroach was found in the market number 12 (24.33 ± 0.67) whereas the lowest was found in market number 2 (19.33 ± 1.20) (*t*-test; $p < 0.05$).

Even though the overall mean number of species richness of bacteria carried by the German cockroach was significantly different from the American cockroach, the means between the two cockroach species in market number 1, market number 2, market number 4, market number 5, market number 6, and market number 9 were not statistically different at $p < 0.05$, *t*-test (Table 5.8; Figure 5.2).

Table 5.8 Means[1] (± SE) of the species richness of bacteria[2] carried by the German and the American cockroach in twelve Bangkok markets in April, July 2005, and January 2006.

Market number	Species richness in the cockroaches	
	German cockroach	American cockroach
No. 1	19.33 ± 0.33 [a]	21.00 ± 0.57 [a]
No. 2	18.33 ± 0.88 [a]	19.33 ± 1.20 [a]
No. 3	Not determine	20.67 ± 0.67
No. 4	18.33 ± 0.33 [a]	19.67 ± 0.33 [a]
No. 5	17.67 ± 0.67 [a]	19.67 ± 0.33 [a]
No. 6	18.00 ± 0.58 [a]	20.00 ± 0.57 [a]
No. 7	19.67 ± 0.88 [a]	24.00 ± 0.00 [b]
No. 8	20.00 ± 0.00 [a]	23.33 ± 0.33 [b]
No. 9	20.00 ± 0.58 [a]	22.33 ± 0.88 [a]
No. 10	19.67 ± 0.33 [a]	22.67 ± 0.67 [b]
No. 11	20.00 ± 0.00 [a]	23.67 ± 0.88 [b]
No. 12	20.33 ± 0.33 [a]	24.33 ± 0.67 [b]
Overall mean (species)	**19.21 ± 0.21 [a]**	**21.72 ± 0.34 [b]**

[1] Means with different letters in the same row are significantly different at $p < 0.05$, *t*-test.

[2] None of the German cockroach was caught in market number 3, the species richness of bacteria could not determine.

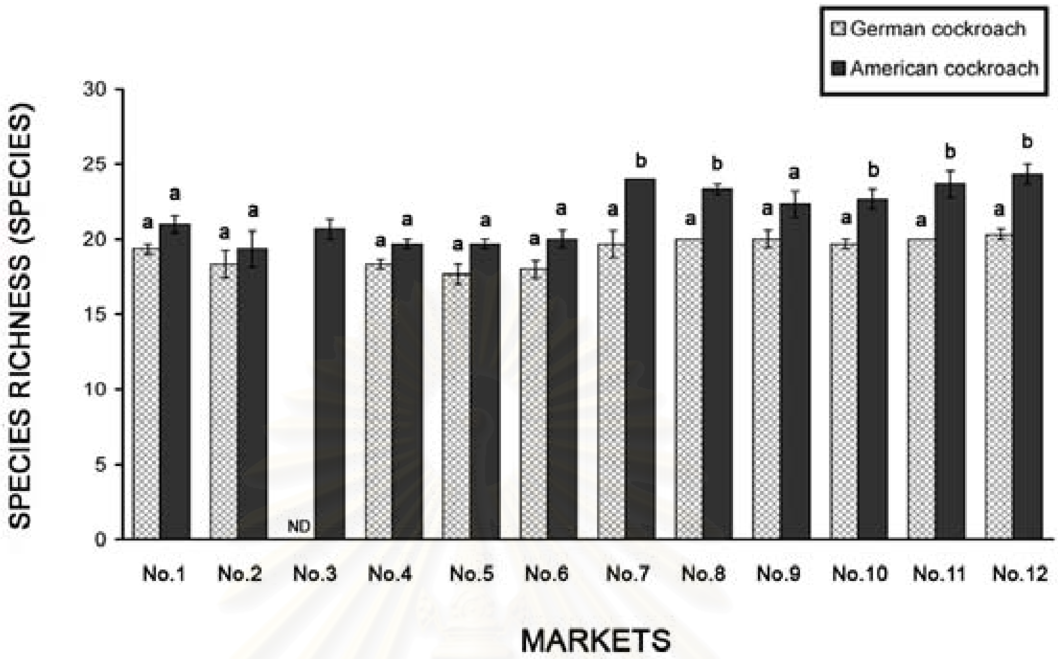

Figure 5.2 Comparison of average numbers (± SE) of the species richness of bacteria carried by the German and the American cockroaches in Bangkok markets. None of the German cockroach was caught in market number 3, the species richness of bacteria could not determine (ND). The bar graphs in each market with different letters are significantly different at $p < 0.05$, *t*-test.

Comparisons of the species richness of bacteria in the 4 combination groups of Bangkok markets

Table 5.9 presented the overall average numbers of the species richness of bacteria isolated from the German and the American cockroaches in the 4 combination groups of Bangkok markets. The overall mean numbers were significantly different during the study period (F = 11.586; df = 3, 68; p < 0.0001). Even though the mean numbers of the species richness of bacteria isolated from the cockroaches caught from market class 2/ low population density zone and market class 2/ high population density zone were not significantly different, the mean in market class 2/ low population density zone was significantly higher than the other combination group of markets (LSD; df = 68; p < 0.05).

In the German cockroaches, means of the species richness of bacteria isolated in the 4 combination groups of Bangkok markets were significantly different during the study period (F = 10.59; df = 3, 32; p < 0.0001). The highest mean was in market class 2/ low population density zone (20.00 ± 1.67) whereas the lowest was in market class 1/ low population density zone (18.00 ± 0.29). Although the mean numbers of the species richness of bacteria isolated from the German cockroaches caught from market class 2/ low population density zone and market class 2/ high population density zone were not significantly different, the mean in market class 2/ low population density zone was significantly higher than the other combination groups of markets (LSD; df = 32; p < 0.05) (Table 5.9; Figure 5.3).

In the American cockroaches, means of the species richness of bacteria isolated in the 4 combination groups of Bangkok markets were significantly different during the study period (F = 23.97; df = 3, 35; p < 0.0001). The highest mean was in market class 2/ low population density zone (23.56 ± 0.44) whereas the lowest was in market class 1/ low population density zone (19.78 ± 0.22). Although the mean numbers of the species richness of bacteria isolated from the German cockroaches caught from market class 2/ low population density zone and market class 2/ high population density zone were not significantly different, the mean in market class 2/ low population density zone was significantly higher than the other combination group of markets (LSD; df = 35; $p < 0.05$) (Table 5.9; Figure 5.3).

Table 5.9 Means[1] (± SE) of the species richness of bacteria carried by the German and the American cockroach in 4 combination groups of Bangkok markets in April, July 2005, and January 2006.

Cockroaches	**Combination groups of Bangkok market** [2]			
	HC1	**LC1**	**HC2**	**LC2**
German cockroach	18.83 ± 0.48 [a]	18.00 ± 0.29 [a]	19.89 ± 0.31 [b]	20.00 ± 1.67 [b]
American cockroach	20.33 ± 0.50 [a]	19.78 ± 0.22 [a]	23.22 ± 0.36 [b]	23.56 ± 0.44 [b]
Overall mean (species)	**16.78 ± 1.64 [a]**	**18.89 ± 0.28 [a]**	**21.56 ± 0.47 [b]**	**21.78 ± 0.49 [b]**

[1] Means with the different letters in the same row are significantly different at $p < 0.05$, ANOVA with LSD.

[2] HC1 = market class 1/ high population density zone, LC1 = market class 1/ low population density zone, HC2 = market class 2/ high population density zone, and LC2 = market class 2/ low population density zone.

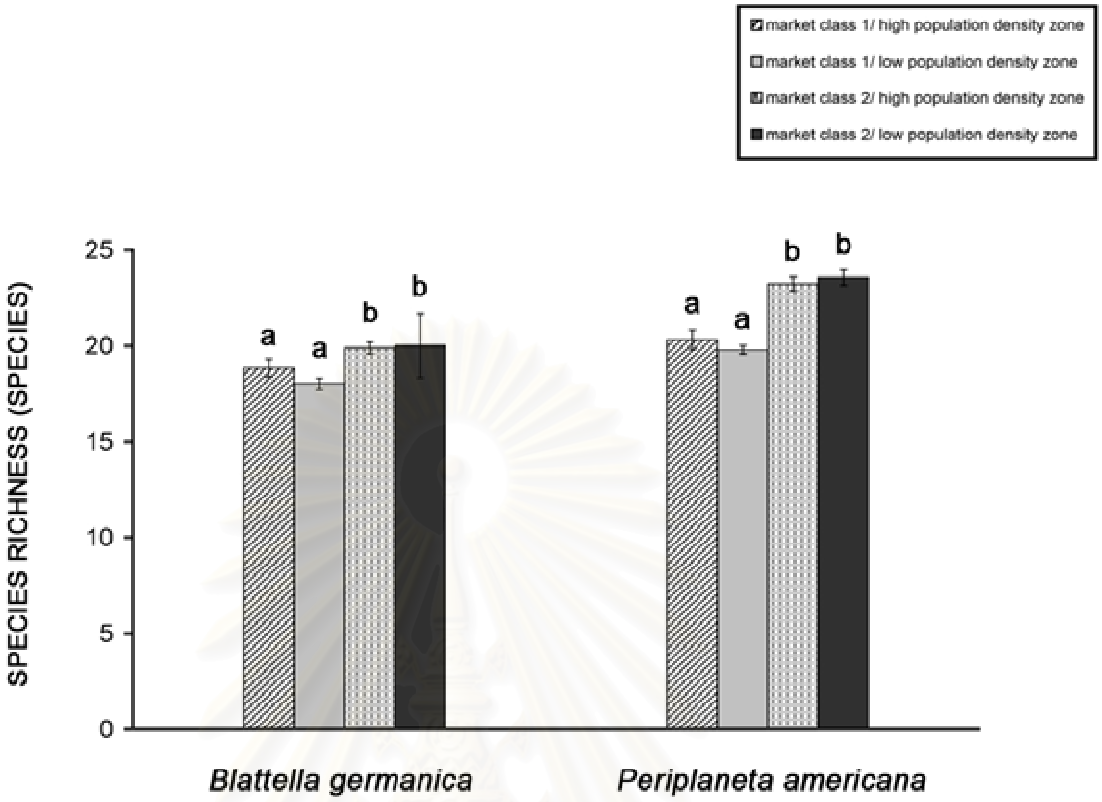

Figure 5.3 Comparison of average numbers (± SE) of the species richness of bacteria isolated from the German, *Blattella germanica*, and the American cockroaches, *Periplaneta americana*, in 4 combination groups of Bangkok markets. The bar graphs in each species of cockroach with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

<u>Comparisons of the species richness of bacteria in the 2 classes of Bangkok markets</u>

Table 5.10 presented the overall average numbers of the species richness of bacteria isolated from the German and the American cockroaches in the 2 classes of Bangkok markets. The overall means were significantly different during the study period (F = 9.332; df = 2, 68; p = 0.003). The mean number of the species richness of bacteria isolated from the cockroaches caught from class 2 market was significantly higher than from class 1 market (t-test; $p < 0.05$).

In the German cockroaches, means of the species richness of bacteria isolated in the 2 classes of Bangkok markets were significantly different during the study period (F = 5.935; df = 2, 32; p = 0.210). The means of the species richness of bacteria isolated from class 2 market was significantly higher than from class 1 market (t-test; $p < 0.05$) (Table 5.10; Figure 5.4).

In the American cockroaches, means of the species richness of bacteria isolated in the 2 classes of Bangkok markets were significantly different during the study period (F = 1.151; df = 2, 35; p = 0.291). The means of the species richness of bacteria isolated from class 2 market was significantly higher than from class 1 market (t-test; $p < 0.05$) (Table 5.10; Figure 5.4).

Table 5.10 Means[1] (± SE) of the species richness of bacteria carried by the German and the American cockroach in 2 classes of Bangkok markets in April, July 2005, and January 2006.

Cockroaches	**Classes of market**	
	Class 1	**Class 2**
German cockroach	18.33 ± 1.05 [a]	19.94 ± 0.73 [b]
American cockroach	20.06 ± 0.27 [a]	23.39 ± 0.28 [b]
Overall mean (species)	**19.27 ± 0.24 [a]**	**21.67 ± 0.33 [b]**

[1] Means with the different letters in the same row are significantly different at $p < 0.05$, *t*-test.

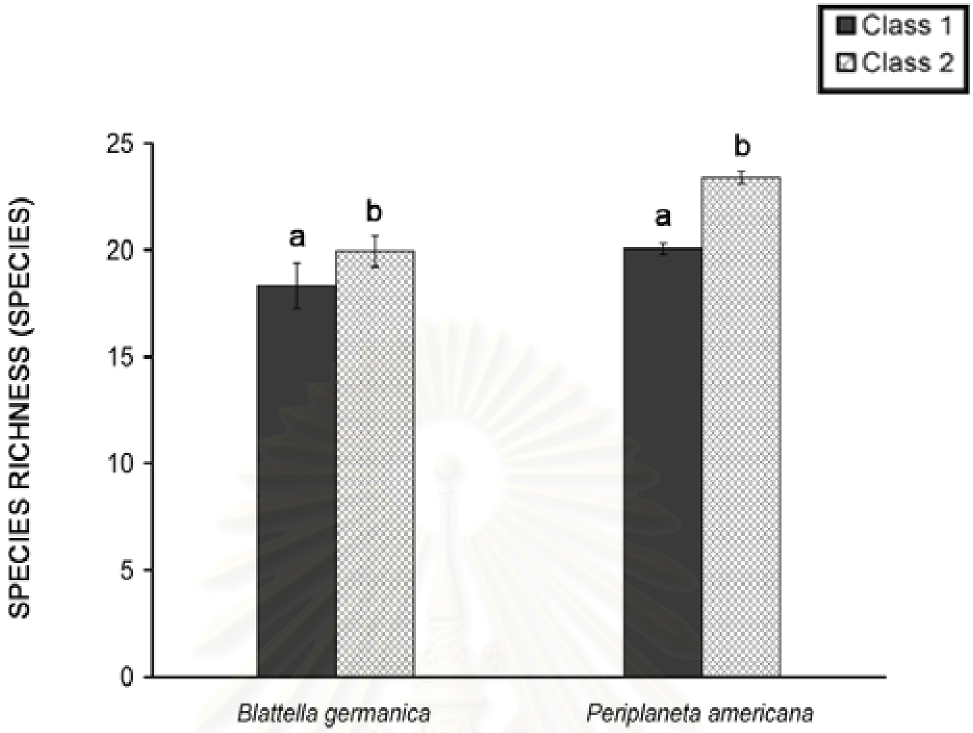

Figure 5.4 Comparison of average numbers (± SE) of the species richness of bacteria carried by the German cockroach, *Blattella germanica* and the American cockroach, *Periplaneta americana* in 2 classes of Bangkok markets in April, July 2005, and January 2006. The bar graphs in each species of cockroach with different letters are significantly different at $p < 0.05$, *t*-test.

Comparisons of the species richness of bacteria isolated from the external cuticle and from the middle gut of cockroaches

Means of the species richness of bacteria isolated from the external cuticle and from the middle gut of both kinds of cockroaches were significantly different during the study period ($F = 45.06$; df = 3, 137; $p < 0.0001$).

In the German cockroaches, overall mean (± SE) of the species richness of bacteria isolated from the external cuticle (18.61 ± 0.29) was statistically different from the middle gut (16.30 ± 0.30). In the American cockroaches, overall mean (± SE) of the species richness of bacteria isolated from the external cuticle (21.28 ± 0.31) was statistically different from the middle gut (18.33 ± 0.32) at $p < 0.05$, *t*-test (Table 5.11; Figure 5.5).

Table 5.11 Means (± SE) of the species richness of bacteria[1] isolated from the external cuticle and the middle gut of the German and the American cockroaches in twelve Bangkok markets in April, July 2005, and January 2006.

Market number	German cockroach		American cockroach	
	External cuticle	Middle gut	External cuticle	Middle gut
No. 1	18.33 ± 0.67	16.67 ± 0.33	20.33 ± 0.88	17.00 ± 0.58
No. 2	16.33 ± 0.33	14.67 ± 0.88	18.67 ± 1.33	15.68 ± 0.88
No. 3	ND	ND	19.67 ± 0.33	17.00 ± 0.00
No. 4	17.33 ± 0.88	15.33 ± 1.86	20.33 ± 0.67	17.00 ± 0.00
No. 5	18.00 ± 0.57	14.67 ± 1.20	20.00 ± 0.57	17.00 ± 0.00
No. 6	16.67 ± 1.20	14.67 ± 1.20	23.00 ± 0.57	21.33 ± 0.33
No. 7	19.33 ± 1.20	17.00 ± 1.00	22.67 ± 0.33	20.00 ± 1.00
No. 8	19.33 ± 0.33	17.67 ± 0.33	22.33 ± 0.88	18.67 ± 0.33
No. 9	20.00 ± 0.57	17.33 ± 0.67	22.00 ± 0.58	19.33 ± 0.67
No. 10	19.67 ± 0.33	17.00 ± 0.00	22.67 ± 0.88	19.67 ± 0.88
No. 11	19.67 ± 0.33	17.00 ± 0.00	23.33 ± 0.88	20.33 ± 0.67
No. 12	20.00 ± 0.00	17.67 ± 0.33	20.33 ± 0.33	17.00 ± 0.00
Overall mean (species)	**18.61 ± 0.29**	**16.30 ± 0.30**	**21.28 ± 0.31**	**18.33 ± 0.32**

[1] None of the German cockroach was caught in market number 3, the species richness of bacteria could not determine (ND).

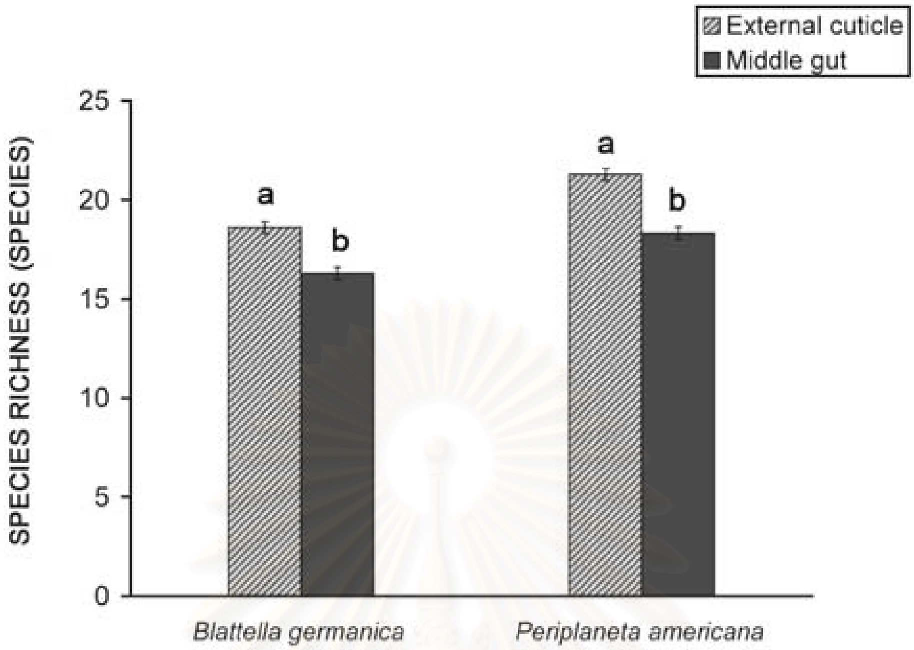

Figure 5.5 Comparison of average numbers (± SE) of the species richness of bacteria isolated from the external cuticle and the middle gut of the German, *Blattella germanica*, and the American cockroaches, *Periplaneta americana*, in twelve Bangkok markets. The bar graphs in each species of cockroach with different letters are significantly different at $p < 0.05$, *t*-test.

Comparisons of the species richness of bacteria isolated among the months of the study period

Table 5.12 presented the overall average numbers of the species richness of bacteria isolated from the cockroaches in twelve Bangkok markets in April, July 2005, and January 2006. The overall mean numbers were not significantly different during the study period (F = 1.250; df = 3, 68; p = 0.293). The overall mean number of the species richness of bacteria isolated in April was not significantly different from July 2005, and January 2006 (LSD; df = 68; p < 0.05).

In the German cockroaches, the means of the species richness of bacteria isolated from the German cockroaches in April, July 2005, and January 2006 were not significantly different during the study period (F = 1.269; df = 3, 32; p = 0.296). The highest mean was in July (19.64 ± 0.41) whereas the lowest was in January (18.91 ± 0.28). The mean numbers of the species richness of bacteria isolated from the German cockroaches caught in April, July 2005, and January 2006 were not significantly different (LSD; df = 32; p < 0.05) (Table 5.12; Figure 5.6).

In the American cockroaches, the means of the species richness of bacteria isolated from the American cockroaches in April, July 2005, and January 2006 were not significantly different during the study period (F = 1.184; df = 3, 35; p = 0.319). The highest mean was in July (22.42 ± 0.51) whereas the lowest was in April (21.17 ± 0.78). The mean numbers of the species richness of bacteria isolated from the American cockroaches caught in April, July 2005, and January 2006 were not significantly different (LSD; df = 35; p < 0.05) (Table 5.12; Figure 5.6).

Table 5.12 Means[1] (± SE) of the species richness of bacteria isolated from the German and the American cockroaches in twelve Bangkok markets in April, July 2005, and January 2006.

Cockroaches	**Months**		
	April	**July**	**January**
German cockroach	19.18 ± 0.26 [a]	19.64 ± 0.41 [a]	18.91 ± 0.28 [a]
American cockroach	21.17 ± 0.78 [a]	22.42 ± 0.51 [a]	21.58 ± 0.38 [a]
Overall mean (species)	**20.22 ± 0.47 [a]**	**21.09 ± 0.44 [a]**	**20.30 ± 0.37 [a]**

[1] Means with the different letters in the same row are significantly different at $p < 0.05$, ANOVA with LSD.

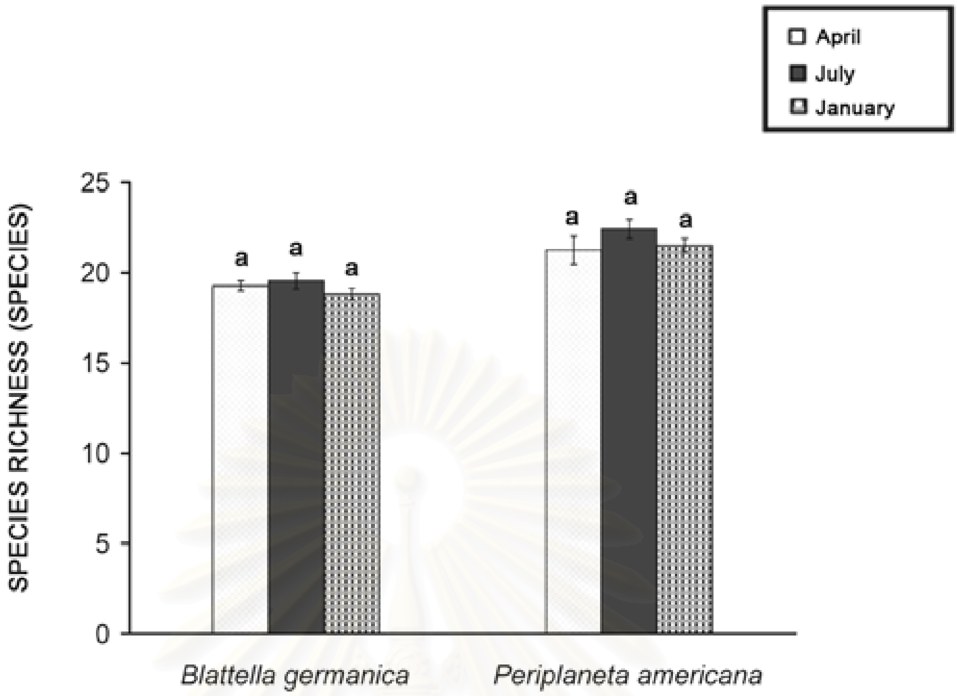

Figure 5.6 Comparison of average numbers (± SE) of the species richness of bacteria isolated from the German, *Blattella germanica*, and the American cockroaches, *Periplaneta americana*, in twelve Bangkok markets in April, July 2005, and January 2006. The bar graphs in each species of cockroach with different letters are significantly different at $p < 0.05$, ANOVA with LSD.

II. Similarity coefficient of bacterial species

The similarity coefficient of the bacterial species carried by the both kinds of cockroaches

The similarity coefficient of the bacterial species between the German and the American cockroaches investigated in twelve Bangkok markets were ranged from 0.870 to 1.00. The highest value was in market number 4 whereas the lowest was in market number 10. Moreover, the overall value of this coefficient was 0.894 (Table 5.13).

Table 5.13 The similarity coefficient (S_s) of the bacterial species[1] between the German and American cockroaches investigated in twelve Bangkok markets in April, July 2005, and January 2006.

Market	Similarity coefficient (S_s)
Number 1	0.952
Number 2	0.976
Number 3	Not determine
Number 4	1.000
Number 5	0.976
Number 6	0.976
Number 7	0.894
Number 8	0.894
Number 9	0.933
Number 10	0.870
Number 11	0.889
Number 12	0.894
Overall	**0.894**

[1] None of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine.

For the external cuticle, the similarity coefficient of the bacterial species between the German and the American cockroaches investigated in twelve Bangkok markets were ranged from 0.784 to 1.00. However, none of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine. The highest value was in market number 4 whereas the lowest was in market number 6. Moreover, the overall value of this coefficient was 0.913 (Table 5.14).

For the middle gut, the similarity coefficient of the bacterial species between the German and the American cockroaches investigated in twelve Bangkok markets were ranged from 0.872 to 1.00. However, none of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine. The highest value was in market number 4, market number 5, and market number 6 whereas the lowest was in market number 10. Moreover, the overall value of this coefficient was 0.900 (Table 5.14).

Table 5.14 The similarity coefficient (S_s) of the bacterial species[1] between the German and American cockroaches isolated from the external cuticle and the middle gut.

Market	Similarity coefficient (S_s)	
	External cuticle	**Middle gut**
Number 1	0.976	0.971
Number 2	0.944	0.970
Number 3	Not determine	Not determine
Number 4	1.000	1.000
Number 5	0.976	1.000
Number 6	0.784	1.000
Number 7	0.933	0.884
Number 8	0.889	0.900
Number 9	0.955	0.947
Number 10	0.889	0.872
Number 11	0.909	0.889
Number 12	0.909	0.900
Overall	**0.913**	**0.900**

[1] None of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine.

The similarity coefficient of the bacterial species between the external cuticle and the middle gut of cockroaches

In the German cockroach, the similarity coefficient of the bacterial species between the external cuticle and the middle gut were ranged from 0.813 to 0.971. However, none of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine. The highest value was in market number 10 whereas the lowest was in market number 2. Moreover, the overall value of this coefficient was 0.923 (Table 5.15).

In the American cockroach, the similarity coefficient of the bacterial species between the external cuticle and the middle gut were ranged from 0.884 to 0.919. The highest value was in market number 4 whereas the lowest was in market number 9. Moreover, the overall value of this coefficient was 0.917 (Table 5.15).

Table 5.15 The similarity coefficient (S_s) of the bacterial species[1] between the external cuticle and the middle gut in the German and American cockroaches investigated in twelve Bangkok markets in April, July 2005, and January 2006.

Market	**Similarity coefficient (S_s)**	
	German cockroach	**American cockroach**
Number 1	0.944	0.900
Number 2	0.813	0.889
Number 3	Not determine	0.895
Number 4	0.919	0.919
Number 5	0.919	0.905
Number 6	0.919	0.895
Number 7	0.923	0.917
Number 8	0.895	0.894
Number 9	0.923	0.884
Number 10	0.971	0.894
Number 11	0.919	0.889
Number 12	0.923	0.894
Overall	**0.923**	**0.917**

[1] None of the German cockroach was caught in market number 3, the similarity coefficient of bacteria could not determine.

Discussion and Conclusion

Several important conclusions may be derived from the data such as the species number of bacteria isolated from the German and the American cockroaches in those markets, species number of bacteria isolated from the 4 combination groups of markets, species number of bacteria isolated from the 2 classes of Bangkok markets, species number of bacteria isolated from the external and the middle gut, species number of bacteria isolated from the different month, and the similarity of bacterial species. The most distinguished result was the bacterial species isolated from the both kinds of cockroaches.

In this study, a total of twenty-six species of bacteria was isolated from the German and the American cockroaches in twelve Bangkok markets. According to the prior reports, Rivault *et al*. (1993) reported that in an urban area of France, 56 species of bacteria have been isolated from cockroaches but these bacterial species have no harm on the cockroach. Vythilingam *et al*. (1997) reported that in an urban area of Malaysia, 17 species of bacteria were isolated from the cockroaches and *Escherichia coli* and *Klebsiella pneumoniae* were the most important ones. Pai *et al*. (2005) reported that in an urban area in Taiwan, 26 species of bacteria were isolated from the German and the American cockroaches.

Several potentially pathogenic bacteria were isolated from the German and the American cockroaches in this study. The bacterial species related to the food-borne diseases such as *Bacillus cereus*, *E. coli*, *Listeria monocytogenes*, *Salmonella arizona*, *Serratia marcescens*, *Staphylococcus aureus*, *Vibrio parahemolyticus*, and *Yersinia enterocolitica*. The isolation of these bacterial species from the cockroaches indicated that these domestic pests could transmit the food-borne diseases to humans in these communities (Tachbele *et al*., 2006). Moreover, *B. cereus*, *E. coli*, *L. monocytogenes*, *Sal. arizona*, *S. marcescens*, and *Sta. aureus* have been isolated from different species of cockroaches found in hospitals, restaurants, and residents throughout the world (Oothuman *et al.*, 1989; Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995; Vythilingam *et al.*, 1997; Tachbele *et al.*, 2006). Although the cockroaches have been considered to be important as s spreader of *Shigella* (Oothuman *et al.*, 1989; Agbodaze and Owusu, 1989; Brenner, 1995; Tachbele *et al.*, 2006), this study did not isolate any bacteria of this genus in the twelve Bangkok markets during the study period.

The bacterial species related to the respiratory tract infections such as *K. ozaenae*, *Pseudomonas aeruginosa*, *Streptococcus pneumoniae*, *Str. pyogenes*, *S. marcescens*, and Viridans Streptococci. These bacterial species have been isolated from the cockroaches throughout the world (Agbodaze and Owusu, 1989; Oothuman *et al.*, 1989; Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995; Vythilingam *et al.*, 1997).

The bacterial species related to the skin and wound infections such as *Corynebacterium pseudotuberculosis*, *Proteus mirabilis*, *Pse. aeruginosa*, *Sta. aureus* and *Str. pyogenes*. The isolation of these bacterial species from the cockroaches was in agreement with other finding elsewhere (Agbodaze and Owusu, 1989; Oothuman *et al.*, 1989; Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995; Vythilingam *et al.*, 1997; Tachbele *et al.*, 2006).

The bacterial species related to the infection of the blood system and lymph node such as *Sta. aureus, Str. pyogenes,* and Viridans Streptococci. These bacterial species have been isolated from the cockroaches throughout the world (Fotedar *et al*., 1991; Rivault *et al.*, 1993; Brenner, 1995; Tachbele *et al.*, 2006).

The bacterial species related to the infection of the nervous system such as *B. cereus, Enterococcus durans,* and *Str. pneumoniae*. These bacterial species have been isolated from the cockroaches throughout the world (Oothuman *et al.*, 1989; Vythilingam *et al.*, 1997; Tachbele *et al.*, 2006).

Moreover, several opportunistic pathogens were isolated from the German and the American cockroaches in this study such as *Citrobacter braakii, C. freundii, Enterobacter aerogens, Ent. cloacae, Ent. gergoviae, Ent. sakazakii, Morganella morganii, and Pse. aeruginosa.* These species have been associated with nosocromial infections especially catheterization patients (Agbodaze and Owusu, 1989; Oothuman *et al.*, 1989; Fotedar *et al*., 1991; Cloarec *et al.*, 1992; Rivault *et al.*, 1993; Rivault *et al.*, 1993; Brenner, 1995 ; Vythilingam *et al.*, 1997).

The Bureau of Epidemiology, Ministry of Public Health of Thailand prescribed the occurrence of diseases that the hospital should report to the Bureau within 24 hours such as Food poisoning outbreak and Cholera, and within 1 week such as Dysentery (Public Health, Ministry, 2006). Therefore, the serious pathogens in this study may be *B. cereus*, *Sal. arizona* and *Sta. aureus* which are causative agents of these diseases.

E. coli is a key-stone species in environmental surveillance as a measure of the warm-blooded animal fecal contamination (Rivault *et al.*, 1993). In this study, *E. coli* could be isolated from both kinds of cockroaches in twelve Bangkok markets. It may be because of the cockroaches contacted the feces of the warm-blooded animals, for example human, dog, cat, and rat in those markets.

Some species of bacteria have been reported as normal flora in the cockroaches such as *Citrobacter*, *Enterobacter*, *Klebsiella* and *Serratia* (Le Guyader *et al*., 1989). The relationship between the cockroaches and the bacteria suggest that bacteria were involves in the breakdown of urates for protein synthesis and also may supply B vitamins, amino acids, and possibly a tri-peptide to the cockroaches (Richards and Brooks, 1958). Moreover, the cockroaches had some behavior such as the nymphs ate feces from their mother (or call trophallaxis behavior) (Holbrook *et al*., 2000). Thus, the cockroaches collected from any places could be isolated some bacteria from their guts (Rivault *et al.*, 1993) and they may direct transfer the pathogens to the other cockroaches by their trophallaxis behavior (trophallaxis: eating the feces from the other, sharing of food or mutual feeding behavior, most often in social insects, in which crop regurgitum or colonic fluid is shared with another individual) (Holbrook *et al*., 2000).

In this study, the result showed that the American cockroach significantly harbored more species of bacteria than the German cockroach. The result agrees to Pai *et al*. (2005). It may be because the American cockroach's body length is three to four folds larger than the German cockroaches. Moreover, the capability of harboring bacteria in cockroaches is not only related to their sizes but may also depend on the behavior and habitat of these cockroaches (Pai *et al*., 2005). The American cockroach may be contact feces, sputum, skin scrapings, and other human and animal secretions more than the German cockroach (Devi and Murray, 1991). According to the observation in this study, the German cockroach mainly on the shelves and crevices in the market shops whereas the American cockroach found in the more unsanitary places such as the market floor, garbage pail, in sewers, toilets, and latrines.

The bacterial species isolated from the external cuticle of cockroaches were statistically different from the middle gut. Probably the free wandering movements of cockroaches from one location to another (Devi and Murray, 1991), the spiniferous and bristly legs of them may be increased the contaminated areas (Devi and Murray, 1991; Ross and Mullins, 1995), and their external body covered with grease might be assisted the adherence of many pathogens (Brenner, 1995; Ross and Mullins, 1995). In this study, *Staphylococcus epidermidis, S. saprophyticus, Streptococcus pneumoniae* and Viridans Streptococci could be isolated only from the external cuticles of the cockroaches. It might be because these bacteria are the real aerobic bacteria and need more oxygen to survive (Stewart and Beswick, 1977) so that probably they could not grow and multiply in the gut of cockroaches where oxygen is lower than the external surface.

The bacterial species isolated from the cockroaches in class 2 market were significantly higher than from class 1 market. *Corynebacterium pseudotuberculosis, Salmonella arizona, Vibrio parahemolyticus*, and *Yersinia enterocolitica* were found in the cockroaches only from class 2 market. It may be because of the differences in the sanitary environmental conditions (Rivault *et al*., 1993; Tachbele *et al.*, 2006). From the observation, many warm-blooded animals such as rat, cat, and dog were found in a large number in class 2 market and the latrines were located close to these markets. Thus, they may acquire bacteria from fecal matter of human and from these animals. It agrees to Rivault *et al*. 1993 that the cockroaches harbored more bacterial species in the poor sanitary condition than in the good sanitary condition.

From the similarity coefficient showed that the both kinds of cockroaches harbored the similar bacterial species, probably these bacteria did not specific to the cockroach species (Brenner, 1995).

In conclusion, the presence of some serious pathogens in the cockroaches in the twelve Bangkok markets becomes important in many aspects. The diseases caused from these bacteria may transmit to human in the communities. Moreover, cockroaches form the natural prey for a variety of animals such as amphibians, reptiles, and rodents (Roth and Willis, 1957). These cockroaches appear to play a significant role in the epidemiology of many diseases in the urban area. Thus, cockroach control and the improvement of the sanitary condition in the communities may reduce the occurrences of many diseases caused by insects especially cockroaches.

CHAPTER 6

EVALUATION OF SOME SYNTHETIC PYRETHROID AND CARBAMATE RESIDUES IN THE GERMAN COCKROACH, *Blattella germanica* (Linnaeus)

Introduction and Objective

The German cockroach, *Blattella germanica*, is the most important indoor urban insect pest in many parts of the world. Current control of this species relies heavily on the use of insecticides. They are usually applied through the form of residual treatment and solid formulation (e.g. Bait) (Lee, Yap, and Chong, 1996b). However, the extensive usage of insecticides has led to the development of insecticide resistance in the German cockroaches (Cochran, 1989; Atkinson *et al*., 1991; Rust and Reierson, 1991; Zhai and Robinson, 1992; Lee *et al*., 1996a).

Pyrethrins as one of the six natural esters of pyrethrum (pyrethrin I/II, jasmolin I/II, and cinerins I/II) as well as synthetic pyrethroids such as cyfluthrin, cypermethrin, deltamethrin, and permethrin are the most often used for pest control worldwide (Chen and Wang, 1996). The principal features separating these materials from other insecticides were their effectiveness for either rapid action (flushing and knockdown), and high toxicity to insects at very low dose (Wickham, 1995). Moreover, for humans, these insecticides are much less toxic than other insecticides (Chen and Wang, 1996). The metabolic pathways of pyrethrins in insects were oxidative degradation and hydrolytic degradation. The formation of the oxidative pathway predominated in insects (Matsumura, 1976). At least ten metabolites of these pathways were recorded and it had less toxic than the parent compound (Yamamoto, Kimmel, and

Casida, 1969). However, pyrethroids are metabolized very fast and can be determined in plasma only a few hours after exposure (Leng, Kühn, and Idel, 1997). The metabolites are renally eliminated with a half-life time of about 6 hours (Leng *et al*., 2003).

Carbamate insecticides are gaining importance in the field of pest control because of their high efficacy as insecticides and nematicides, their low bioaccumulation potentials, and their relatively low mammalian toxicities (Fahmy, Mallipudi, and Fukuto, 1978). Since 1959, propoxur has been used as the major compound for the German cockroach control throughout the world. Propoxur also has fast knockdown activity, only slightly slower than many rapidly acting pyrethroids (Wickham, 1995). However, since they are acetylcholinesterase inhibitors, they are considered hazardous to the environment and human health (Fahmy *et al*., 1978). The metabolic pathways of propoxur in insects were oxidative pathway and hydrolytic degradation. The formation of the hydrolytic degradation predominated in insects. Isopropoxyphenol was the metabolite of this pathway and it had less toxic than the parent compound (Dorough and Casida, 1964).

The information on the insecticidal toxicity on the German cockroaches has been reported by many publications (Wadleigh *et al*., 1991; Rust, Reierson, and Zeichner, 1993; Negus and Ross, 1997; Lee, 1998; Ameen *et al*., 2005; Sitthicharoenchai, Chaisuekul, and Lee, 2006). However, the data of insecticidal residues in the living German cockroaches have been limited when compared to those reported on their baseline resistance and susceptibility to insecticides.

The objective of this experiment was to examine the insecticidal residues in the living German cockroach, *B. germanica*, at twelve Bangkok markets. This study provided information on the level of insecticide contamination in the German cockroaches at twelve Bangkok markets. It may provide information for the insecticidal resistance in the German cockroaches in the study area.

Materials and Methods

I. The insecticidal usage interview

Before the insecticidal residues were analyzed, the insecticide usage was evaluated using the interview (Appendix B). The data were received from the market vendors such as the shop owners in twelve Bangkok markets. By the combination of human population density zones and the classes of Bangkok markets, 4 combination groups such as high population density zone/ market class 1, high population density zone/ market class 2, low population density zone/ market class 1, and low population density zone/ market class 2 were categorized, and the numbers of the markets in each combination group are showed in Table 4.1. In this study, 12 market sites (3 sites for each combination group in Bangkok) were selected using random sampling method (Krebs, 1999), and the market's names and their location map were showed in Table 4.2 and Figure 4.1.

II. The insecticidal residues analysis

1. Reagents

Insecticide-grade acetonitrile and methanol were purchased from Lab-Scan (Bangkok, Thailand). HPLC-grade water was obtained from Mallinckrodt Baker (Kentucky, USA). HPLC-grade phosphoric acid was obtained from Lab-Scan (Bangkok, Thailand). Anhydrous sodium sulfate was obtained from Ajax Finechem (New South Wales, Australia).

Anhydrous sodium sulfate was heat treated in an oven at 200 °C for a minimum of 4 hours to remove interfering organic substances before use. All solvents and solutions for High Performance Liquid Chromatography (HPLC) analysis were degassed in an ultrasonic bath before use.

The mixed standard of the synthetic pyrethroids, including 3 components: cyfluthrin, cypermethrin, and flumethrin were purchased by Riedel-de Haën (Sigma-Aldrich, Taufkirchen, Germany). Tri-phenoxybenzoic acid (98%), the main metabolite of the cypermethrin, was purchased from Aldrich (Sigma-Aldrich, Steinheim, Germany). Insecticide standard of propoxur (2-isopropoxyphenyl methylcarbamate) was purchased with its purity certified from Dr. Ehrenstorfer (Ausberg, Germany), and 2-isopropoxyphenol (97%), the main metabolite of the propoxur, was purchased from Aldrich (Sigma-Aldrich, Steinheim, Germany).

2. Sample collection

The living German cockroaches, *B. germanica*, were collected from twelve Bangkok markets using modified jar traps (see more detail in Chapter 4). This experiment was conducted from March 2005 to March 2006. The cockroach samples were cooled and stored in a refrigerator at temperature lower than 0 °C until extraction (Tekel, Hudecová, and Pecníková, 2001).

3. Sample preparation

3.1 <u>Sample extraction</u>

Five grams of the whole body of each cockroach sample group was ground with 15 grams anhydrous sodium sulfate (1:3 w/w) using an agate mortar (Pan *et al*., 2004). The mixture was packed into a 34-ml vessel of accelerated solvent extractor, ASE (ASE®-100, Dionex, CA, USA) which was layered with ASE filter cellulose paper (Dionex, CA, USA) and fulfilled with the 1 mm-glass bead.

The pressured liquid extraction was implemented using ASE for these samples. The working conditions were as follows: preheating for 5 min, extraction temperature at 100 °C, pressure at 1500 psi, static cycle of 15 min twice, and purging with Nitrogen for 60 second. The samples were extracted with 68 ml of acetonitrile

(Tekel *et al*., 2001), then, the extract was concentrated to 2 ml using stream of nitrogen in evaporation system of Turbo Vap II (Zymark, Hopkinton, MA, USA).

3.2 Sample clean up

Because of the presence of pigments in the sample, the Solid phase extraction (SPE) was used for remove the contaminants before injecting to HPLC. The extract was cleaned through 500 mg extract-clean-florisil Vertipak SPE cartridge (Vertical Chromatography, Bangkok, Thailand) to clean up process. The extract was eluted twice by 2 ml of acetonitrile, then by 2 ml of methanol. The sorbent was not allowed to dry during the conditioning and sample loading steps. The elute was concentrated to a volume of 1.5 ml using stream of nitrogen in Turbo Vap II (Zymark, Hopkinton, MA, USA). Before HPLC analysis, the sample solution was filtered through a 0.45-µm Vertipure nylon syringe filter (Vertical Chromatography, Bangkok, Thailand) to avoid the effect of microorganisms (Brown and Hartwick, 1989). The 20 µl of extract were directly injected into the HPLC system.

4. Sample analysis

A HPLC was performed with a Varian system (Varian, CA, USA), equipped with a pump (Varian model Prostar 240), an injection valve (Varian model Prostar 410), a UV detector (Varian model Prostar 335), and an auto-sampler (Varian model Prostar 410). Compounds were separated on a Chromspher C_{18} 250 x 4.6 mm (5 µm) column (Merck, Darmstadt, Germany), to determine the synthetic pyrethroids and their metabolite, and on a LiChrospher C_{18} 250 x 4.0 mm (5 µm) column (Merck, Darmstadt, Germany), to determine the propoxur and its metabolite. The analytical conditions of HPLC were listed in Table 6.1.

4.1 The mixed standard of synthetic pyrethroids and their metabolite

Mobile phase was degassed and filtered through a 0.45-µm Vertipure nylon membrane filter (Vertical Chromatography, Bangkok, Thailand). The mobile phase was comprised a mixture of water (added 0.1 % phosphoric acid as eluent modifier) and acetonitrile gradient at flow rate programmed from 0.5 ml/min from 0 to 9 min, increased to 2.0 ml/min by 10 min and then returned to 0.5 ml/min at 13 min. The gradient started at 45 % acetronitrile until 9 min and increased to 90 % acetonitrile by 10 min. Then, the system returned to 45 % acetonitrile at 13 min where it was kept under this condition for 2 min to re-equilibrate. Chromatography was performed at ambient temperature and the injected volume was 20 µl. The eluents were monitored by UV detection of wavelength of 210 nm (Abu-Qare and Abou-Donia, 2001).

4.2 The Propoxur and its metabolite

The mobile phase were comprised a mixture of acetonitrile: water: methanol (55: 37: 8, v/v). It was filtered through a 0.45-µm Vertipure nylon membrane filter (Vertical Chromatography, Bangkok, Thailand), degassed and delivered at flow rate of 1.0 ml/ min. Chromatography was performed at ambient temperature with 20 µl direct-injection. The eluents were monitored by UV detection of wavelength of 280 nm (Orejuela and Silva, 2003).

Table 6.1 Working condition of synthetic pyrethroids and propoxur in HPLC

Condition	**Synthetic pyrethroids**	**Propoxur**
Column	Chromspher C_{18}	LiChrospher C_{18}
	250 x 4.6 mm (5 µm)	250 x 4.0 mm (5 µm)
Mobile phase	acetonitrile : water +0.1 %	acetonitrile: water: methanol
	phosphoric acid	
Flow rate	0.5 – 2.0 ml/min	1.0 ml/ min
Injection volume	20 µl	20 µl
Wavelength	210 nm	280 nm
Quantitative method	External standard, peak area	External standard, peak area

5. Method validation

5.1 Limit of detection (LOD) and limit of quantitation (LOQ)

The limit of detection (LOD) and limit of quantitation (LOQ) were defined as the peak height of analyte in standard solution that signaled significantly different from the peak height of noise. They were 3 and 10 times of signal per noise for LOD and LOQ, respectively. LOD and LOQ were done. In case of the synthetic pyrethroids and the propoxur concentrations below the LOD, the results were described as zero. The LOQ was repeated five times for confirmation (Abu-Qare and Abou-Donia, 2001).

5.2 Spike recovery

Fortified samples were done for every sampling batch to ensure that the extraction efficiency would be under control (Kebbekus and Mitra, 1998). The acceptable recovery of the synthetic pyrethroids and the propoxur should be ranged from 80-110 % at 100 ppb (AOAC, 1993). The recovery percentage can be calculated by the equation below:

$$\% \text{ Recovery} = \frac{\text{amount of insecticides determined}}{\text{amount of insecticides standard}} \times 100$$

5.3 Blanks

To avoid the effect of interferences, the set of blanks were done. The blanks included solvent blank, system blank, and fortified sample bank. These blanks were done for every sample batch. The blanks must be free from contaminants, or the concentration of contaminated analytes must be at least level.

5.4 Replications

The replications of samples were done to evaluate repeatability. The samples were extracted and analyzed in triplicate to be sure that the measurement remained stable. The relative standard deviation (RSD) is the parameter of choice for comparing the precision of data of different units and magnitudes. The acceptable of % RSD should not be exceeding 15 % at 100 ppb (AOAC, 1993). The % RSD was calculated from the equation as below:

$$\% \text{ RSD} = \frac{\text{standard deviation}}{\text{mean}} \times 100$$

5.5 Method detection limit (MDL)

The minimum concentration of an analyte that can be identified, measured, and reported with 99% confidence that the analyte concentration is greater than zero. The detection limit of the selected method was calculated based on the replicated determinations as below:

$$MDL = t_{0.95\ (n-1)} \times SD$$

When t is the threshold value of t distribution at the degree of (n-1), n represents the number of replications, and SD represents the standard deviation. The confidence interval is 95% (α= 0.05).

Data analysis

Descriptive statistics were used to present as number, percentage, mean, standard deviation, and standard error. All data were checked for normal distribution. If any data was normal distribution, ANOVA with LSD analysis ($p < 0.05$) were used to compare the concentration of insecticides. But, if any data was not normal distribution, *Mann-Whitney U*-test ($p < 0.05$) was used to compare the concentration of insecticides.

Results

I. Insecticide usage in twelve Bangkok markets

The insecticide usage in twelve Bangkok markets was evaluated by the interview, responded to types of chemical insecticides, trade name of insecticides, and cost of cockroach control strategies in their shops. A total of 132 from 240 market vendors (55.00 %) used the insecticides for cockroach elimination in their shops. The formulation of insecticides that they applied was aerosol formulation (100%). The trade name of the insecticides that they used were Baygon® green (43.94 %), Baygon® green-orange (21.97 %), Shieldtox® for termite and cockroach (19.70 %), and Shelldrite® for termite and cockroach (14.39 %). The frequencies that they used the insecticides for cockroach elimination were every week (59.09 %), do when see them (28.03 %), and every day (12.88 %). All of the market vendors had to spend for the insecticides less than100 Baht per month (100%). The reasons for avoiding the use of insecticides were the cost of insecticides (46.29 %), the awareness of insecticidal toxicity (32.41 %), and the inefficiency of insecticides (21.30 %) (Table 6.2; Table 6.3). The active ingredients of these insecticides were presented in Table 6.4.

Table 6.2 The insecticide usage in twelve Bangkok markets surveyed from March 2005 to March 2006.

Questions	Number	Percent
1. Cockroach elimination by insecticide in shop		
Yes	132	55.00
No	108	45.00
2. Type of chemical insecticide		
Chemical aerosols	132	100.00
3. Trade name of insecticides		
Baygon® green	58	43.94
Baygon® green-orange	29	21.97
Shieldtox® for termite and cockroach	26	19.70
Shelldrite® for termite and cockroach	19	14.39
4. Frequency of the usage		
Every day	17	12.88
Every week	78	59.09
Do when see them	37	28.03
5. Money has spent for cockroach control		
Less than 100 Baht/ month	132	100.00
6. Reason for untreated		
Cost of the insecticides	50	46.29
Awareness of insecticidal toxicity	35	32.41
Inefficiency of insecticides	23	21.30

Table 6.3 The trade names of the insecticides and number of market vendors that used those insecticides in twelve Bangkok markets from March 2005 to March 2006.

Market no.	**Insecticides [1]**				**Total of market vendors**
	A	**B**	**C**	**D**	
1	3	2	2	3	10
2	4	1	3	2	10
3	10	5	0	0	15
4	4	3	3	3	13
5	5	1	2	2	10
6	3	2	9	3	17
7	3	3	0	2	8
8	5	1	0	0	6
9	6	4	3	2	15
10	7	4	1	1	13
11	4	2	3	0	9
12	4	1	0	1	6
Total	**58**	**29**	**26**	**19**	**132**

[1] A = Baygon® green, B = Baygon® green-orange, C = Shieldtox® for termite and cockroach, and D = Shelldrite® for termite and cockroach.

Table 6.4 The active ingredients of some insecticides surveyed in twelve Bangkok markets from March 2005 to March 2006.

Trade name	Active ingredients	%
Baygon® green	Propoxur	0.5 w/w
	Cyfluthrin	0.025 w/w
Baygon® green-orange	Propoxur	0.75 w/w
	Cyfluthrin	0.025 w/w
Shieldtox® for termite and cockroach	Cypermethrin	0.15 w/w
Shelldrite® for termite and cockroach	Cypermethrin	0.1 w/w

II. Results of insecticidal analysis

1. Standard calibration curves

The standard calibration curves of peak area against concentration of cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol were done. Linearity of the calibration curves for these compounds was achieved at concentrations ranging from 10 to 500 ppb. The multiple correlation coefficient (R^2) ranged from 0.9876 to 0.9975. Moreover, the statistical data from regression analysis for cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol were presented in Table 6.5.

Table 6.5 Statistical data from regression analysis for the insecticides in twelve Bangkok markets.

Insecticides	**Regression equation[1]**	**Multiple correlation coefficient (R^2)**
Cyfluthrin	y = 140.12 x + 1398.1	0.9936
Cypermethrin	y = 57.157 x + 9130.7	0.9916
Flumethrin	y = 140.46 x + 14010	0.9959
3-Phenoxybenzoic acid	y = 1549.9 x + 386324	0.9876
Propoxur	y = 133.46 x + 17772	0.9876
2-Isopropoxyphenol	y = 1219.3 x + 25195	0.9975

[1] Analysis from seven concentrations, x = concentration (ppb) and y = response area.

2. Limits of detection (LOD)

Limits of detection were calculated from a peak signal to noise ratio of 3:1 in each compound. The resulting detection limits for cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol were presented in Table 6.6. The LOD of this study was ranged from 0.1 to 0.3 ppb.

3. Limits of quantitation (LOQ)

Limits of quantitation were calculated from a peak signal to noise ratio of 10:1. The resulting quantitation limits for cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol were presented in Table 6.6. The LOQ of this study was ranged from 0.5 to 1.0 ppb.

4. Method detection limit (MDL)

The minimum concentration of six analytes that can be identified and measured. The results in Table 6.6 showed that the MDL of six insecticides ranged from 3.73 to 14.75 ppb.

5. Extraction efficiency and recovery

The average extraction recoveries of cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol were presented in Table 6.6. The % recoveries of these insecticides ranged from 94.01 to 98.65 %. All recovery percentages were in acceptable range (AOAC, 1993). The repeatability was described by % RSD. The % RSD were ranged from 2.04 to 7.98. All % RSD were in acceptable range (AOAC, 1993). Therefore, the recovery percentages and repeatability of all analytes were accepted in this experiment.

Table 6.6 Limit of detection (LOD), limit of quantitation (LOQ), method detection limit (MDL), spiked recovery (% Recovery), and relative standard deviation (% RSD) of insecticides standard solution in the German cockroach samples caught from twelve Bangkok markets from March 2005 to March 2006.

Insecticides	LOD (ppb)	LOQ (ppb)	MDL (ppb)	% Recovery [1]	% RSD
Cyfluthrin	0.1	0.6	5.13	94.01	2.81
Cypermethrin	0.3	1.0	14.59	94.06	7.98
Flumethrin	0.3	1.0	7.17	98.33	3.75
3-Phenoxybenzoic acid	0.3	1.0	3.73	94.05	2.04
Propoxur	0.3	0.5	14.75	98.65	7.69
2-Isopropoxyphenol	0.2	0.5	4.52	92.46	2.52

[1] Average from seven replications.

6. The concentrations of the synthetic pyrethroids, propoxur, and their metabolites in the German cockroaches caught from twelve Bangkok markets

Insecticidal residue concentrations in the German cockroaches caught from twelve Bangkok markets

The overall means concentrations (ppb) of the cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol in the German cockroaches caught from twelve Bangkok markets were presented in Table 6.7. The highest mean concentration of the insecticidal residues in the German cockroach samples was cypermethrin (86.27 ± 6.34), followed by 3-phenoxybenzoic acid (50.92 ± 8.64), and cyfluthrin (27.75 ± 10.08). The concentrations of flumethrin, propoxur, and 2-isopropoxyphenol in the German cockroach samples were below the LOD, thus, the results were described as zero. The overall mean concentration of cypermethrin was significantly higher than the other compounds (*Man-Whitney U-*test; $p < 0.05$) (Figure 6.1).

Table 6.7 Mean (± SE) concentrations (ppb) of the insecticidal residues in the German cockroach samples caught from twelve Bangkok markets from March 2005 to March 2006 (CYF = cyfluthrin, CYP = cypermethrin, FLU = flumethrin, PBA = 3- phenoxybenzoic acid, PRO = propoxur, and IPP = 2-isopropoxyphenol).

Market no.	Mean concentration[1] (ppb)					
	CYF	**CYP**	**FLU**	**PBA**	**PRO**	**IPP**
1	0.00 ± 0.00^{a}	83.88 ± 1.41^{de}	0.00 ± 0.00^{a}	53.54 ± 0.35^{d}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
2	0.00 ± 0.00^{a}	78.66 ± 3.35^{cd}	0.00 ± 0.00^{a}	38.29 ± 0.88^{c}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
3	ND	ND	ND	ND	ND	ND
4	64.24 ± 2.31^{e}	108.34 ± 5.58^{f}	0.00 ± 0.00^{a}	88.29 ± 0.69^{f}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
5	0.00 ± 0.00^{a}	76.03 ±2.74^{bcd}	0.00 ± 0.00^{a}	36.73 ± 0.56^{c}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
6	107.43 ± 4.29^{f}	134.46 ±2.31^{g}	0.00 ± 0.00^{a}	103.12 ±4.66^{g}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
7	14.08 ± 4.2 ^{b}	66.79 ± 4.17^{ab}	0.00 ± 0.00^{a}	23.90 ± 1.78^{b}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
8	22.45 ± 4.02^{bc}	71.18 ±5.79^{abc}	0.00 ± 0.00^{a}	35.49 ± 0.76^{c}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
9	31.02 ± 0.74^{c}	101.81±0.84 ^{f}	0.00 ± 0.00^{a}	81.37 ± 0.38^{e}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
10	41.53 ± 7.13^{d}	88.83±3.88 ^{e}	0.00 ± 0.00^{a}	53.66 ± 0.37^{d}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
11	24.57 ± 2.63^{bc}	73.46±1.87 ^{abc}	0.00 ± 0.00^{a}	34.19 ± 0.56^{c}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
12	0.00 ± 0.00^{a}	65.59±1.23 ^{a}	0.00 ± 0.00^{a}	11.57 ± 0.65^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Overall mean[2]	**27.75 ± 10.08 ^{b}**	**86.27 ± 6.34 ^{c}**	**0.00 ± 0.00 ^{a}**	**50.92 ± 8.64 ^{b}**	**0.00 ± 0.00 ^{a}**	**0.00 ± 0.00 ^{a}**

[1] Means with the different letters in the same column are significantly different at $p < 0.05$, *Mann-Whitney U*-test. None of the German cockroaches was caught in market number 3 during the study period, the results were presented as ND (not determined).

[2] Means with different letters in the same row are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

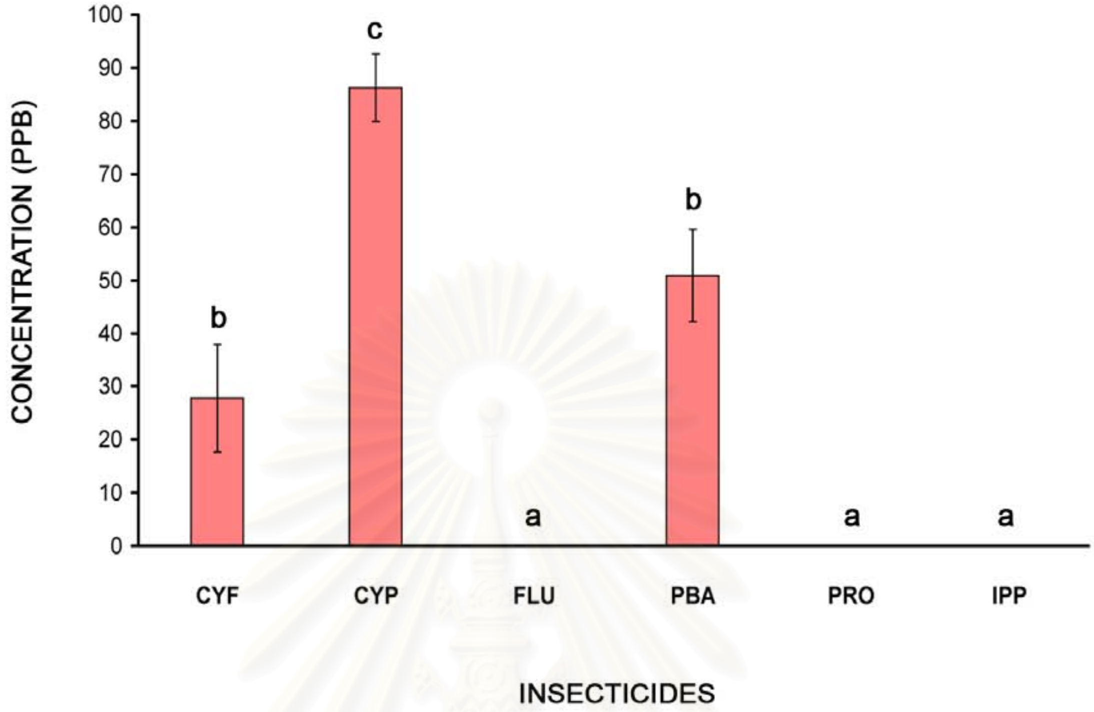

Figure 6.1 Comparison of the mean (± SE) concentration (ppb) of the insecticidal residues in the German cockroach sample caught from twelve Bangkok markets from March 2005 to March 2006 (CYF = cyfluthrin, CYP = cypermethrin, FLU = flumethrin, PBA = 3- phenoxybenzoic acid, PRO = propoxur, and IPP = 2-isopropoxyphenol).The bar graphs with different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

The highest mean concentrations (ppb) of cyfluthrin, cypermethrin, and 3-phenoxybenzoic acid in the German cockroach samples were detected in market number 6. They were 107.43 ± 4.29, 134.46 ± 2.31, and 103.12 ± 4.66 for cyfluthrin, cypermethrin, and 3-phenoxybenzoic acid, respectively. The lowest mean concentrations of cyfluthrin were detected in market number 1 (0.00 ± 0.00), market number 2 (0.00 ± 0.00), market number 5 (0.00 ± 0.00), and market number 12 (0.00 ± 0.00). The lowest mean concentrations (ppb) of cypermethrin and 3-phenoxybenzoic acid were detected in market number 12. They were 65.59 ± 1.23 and 11.57 ± 0.65 for cypermethrin and 3-phenoxybenzoic acid, respectively.

Comparison of the mean concentrations (ppb) of cyfluthrin, cypermethrin, and 3-phenoxybenzoic acid among twelve Bangkok markets, the mean concentrations (ppb) of these insecticides were found in market number 6 significantly higher than the other markets (*Man-Whitney U*-test; $p < 0.05$) (Figure 6.2).

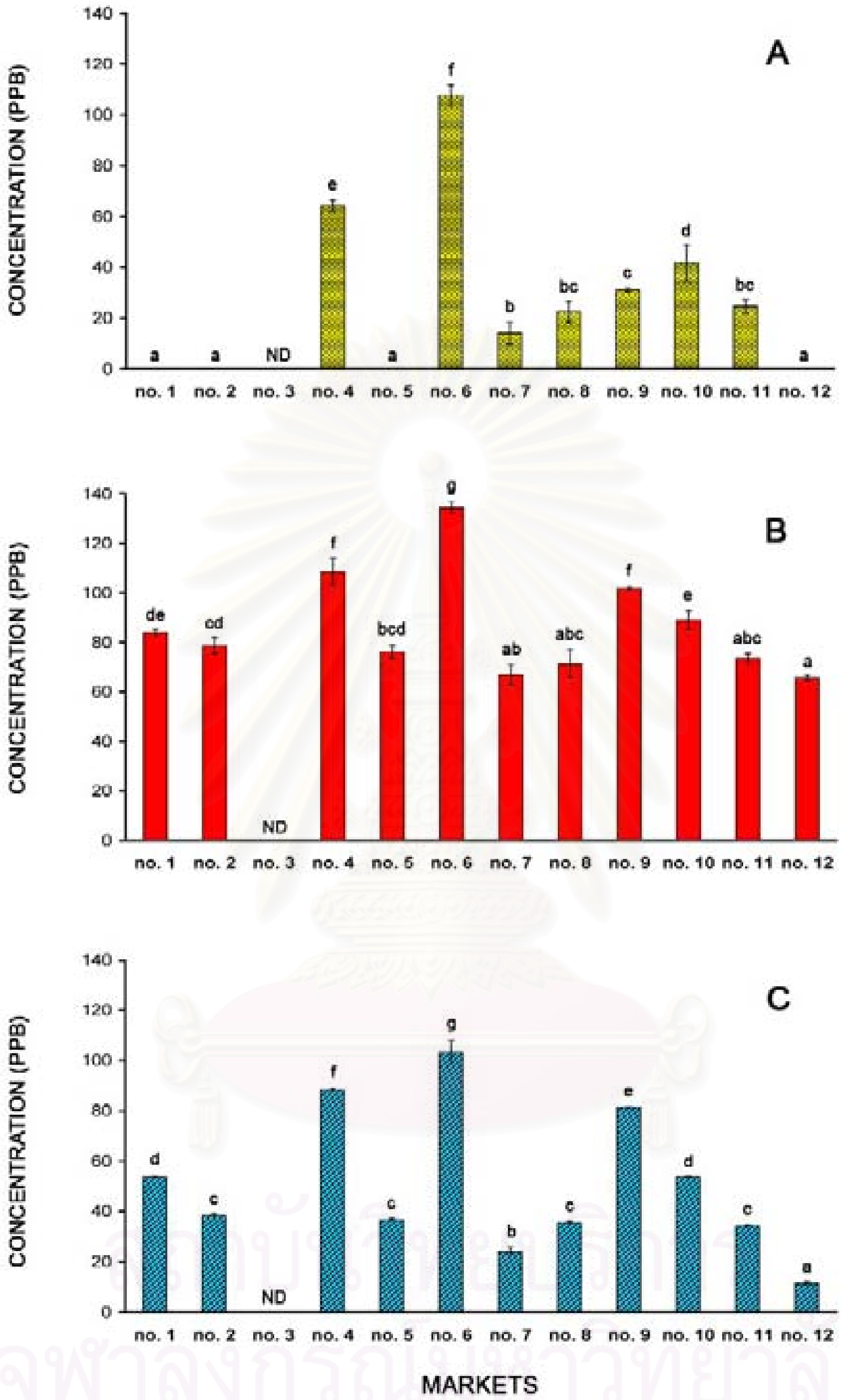

Figure 6.2 Mean (± SE) concentrations (ppb) of the cyfluthrin (A), cypermethrin (B), and 3-phenoxybenzoic acid (C) in the German cockroach samples caught from twelve Bangkok markets from March 2005 to March 2006. The bar graphs with different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

In each market, the mean concentrations of the insecticidal residues in the German cockroaches were presented in Table 6.8. The mean concentration of cypermethrin in the German cockroach samples was significantly higher than the other compounds in every market during the study period at $p < 0.05$, *Mann-Whitney U*-test.

Table 6.8 Mean[1] (± SE) concentrations (ppb) of the insecticidal residues in the German cockroach samples in each market (CYF = cyfluthrin, CYP = cypermethrin, FLU = flumethrin, PBA = 3- phenoxybenzoic acid, PRO = propoxur, and IPP = 2-isopropoxyphenol).

	Market no. 1	**Market no. 2**	**Market no. 3**	**Market no. 4**
CYF	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	ND	64.24 ± 2.31 ^{b}
CYP	83.88 ± 1.41 ^{c}	78.66 ± 3.35 ^{c}	ND	108.34 ± 5.58 ^{d}
FLU	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	ND	0.00 ± 0.00 ^{a}
PBA	53.54 ± 0.35 ^{b}	38.29 ± 0.88 ^{b}	ND	88.29 ± 0.69 ^{c}
PRO	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	ND	0.00 ± 0.00 ^{a}
IPP	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	ND	0.00 ± 0.00 ^{a}

	Market no. 5	**Market no. 6**	**Market no. 7**	**Market no. 8**
CYF	0.00 ± 0.00 ^{a}	107.43 ± 4.29 ^{b}	14.08 ± 4.26 ^{b}	22.45 ± 4.02 ^{b}
CYP	76.03 ± 2.74 ^{c}	134.46 ± 2.39 ^{c}	66.79 ± 4.17 ^{d}	71.18 ± 5.79 ^{d}
FLU	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
PBA	36.73 ± 0.56 ^{b}	103.12 ± 4.66 ^{b}	23.9 ± 1.78 ^{c}	35.49 ± 0.76 ^{c}
PRO	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
IPP	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}

	Market no. 9	**Market no. 10**	**Market no. 11**	**Market no. 12**
CYF	31.02 ± 0.74 ^{b}	41.53 ± 7.13 ^{b}	24.57 ± 2.63 ^{b}	0.00 ± 0.00 ^{a}
CYP	101.81 ± 0.84 ^{d}	88.83 ± 3.88 ^{d}	73.46 ± 1.87 ^{d}	65.59 ± 1.23 ^{c}
FLU	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
PBA	81.37 ± 0.38 ^{c}	53.66 ± 0.37 ^{c}	34.19 ± 0.56 ^{c}	11.57 ± 0.65 ^{b}
PRO	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
IPP	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}

[1] Means concentrations with the different letters in the same column and in the same market are significantly different at $p < 0.05$, *Mann-Whitney U*-test. None of the German cockroaches was caught in market number 3 during the study period, the results were presented as ND (not determined).

Comparisons of the insecticidal concentrations in the 4 combination groups of Bangkok markets

The overall mean concentrations (ppb) of cyfluthrin, cypermethrin, flumethrin, 3-phenoxybenzoic acid, propoxur, and 2-isopropoxyphenol in the German cockroach caught from the 4 combination groups of Bangkok markets, as described before, were presented in Table 6.9.

The highest mean concentration was in market class 1/ low population density zone (39.93 ± 6.67), followed by market class 2/ high population density zone (25.08 ± 4.45), and market class 2/ low population density zone (21.86 ± 4.05), respectively. The lowest mean was in market class 1/ high population density zone (14.13 ± 3.82). The mean concentration of insecticidal residues in the German cockroaches caught from market class 1/ low population density zone was significantly higher than the other combination group of markets (*Man-Whitney U*-test; $p < 0.05$).

Table 6.9 Mean (± SE) concentrations (ppb) of the insecticidal residues in the German cockroaches caught from the 4 combination groups of Bangkok markets (CYF = cyfluthrin, CYP = cypermethrin, FLU = flumethrin, PBA = 3- phenoxybenzoic acid, PRO = propoxur, and IPP = 2-isopropoxyphenol).

	Combination groups of Bangkok markets [1]			
	HC1	**LC1**	**HC2**	**LC2**
CYF	0.00 ± 0.00	57.22 ± 15.67	23.63 ± 2.35	22.04 ± 6.42
CYP	54.18 ± 13.6	106.28 ± 8.66	79.93 ± 5.89	75.96 ± 3.65
FLU	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
PBA	30.60 ± 7.96	76.05 ± 10.15	46.92 ± 8.79	33.14 ± 6.08
PRO	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
IPP	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Overall mean[2]	14.13 ± 3.82 [a]	39.93 ± 6.67 [b]	25.08 ± 4.45 [a]	21.86 ± 4.05 [a]

[1] HC1 = market class 1/ high population density zone, LC1 = market class 1/ low population density zone, HC2 = market class 2/ high population density zone, and LC2 = market class 2/ low population density zone.

[2] Mean concentrations with the different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

Overall, the mean concentrations of the cyfluthrin analyzed from 4 combination groups of markets were significantly different. The mean concentration of cyfluthrin in market class 1/ low population density zone was statistically different from the other combination groups of markets ($p < 0.05$, *Mann-Whitney U*-test). The mean concentrations of the cypermethrin analyzed from 4 combination groups of markets were also significantly different. The mean concentration of cypermethrin in market class 1/ low population density zone was significantly higher than the other combination groups of the markets ($p < 0.05$, *Mann-Whitney U*-test). Moreover, the mean concentrations of the 3-phenoxybenzoic acid analyzed from 4 combination groups of markets were also significantly different. The mean concentration of 3-phenoxybenzoic acid in market class 1/ low population density zone was significantly different from the other combination group of markets ($p < 0.05$, *Mann-Whitney U*-test) (Figure 6.3).

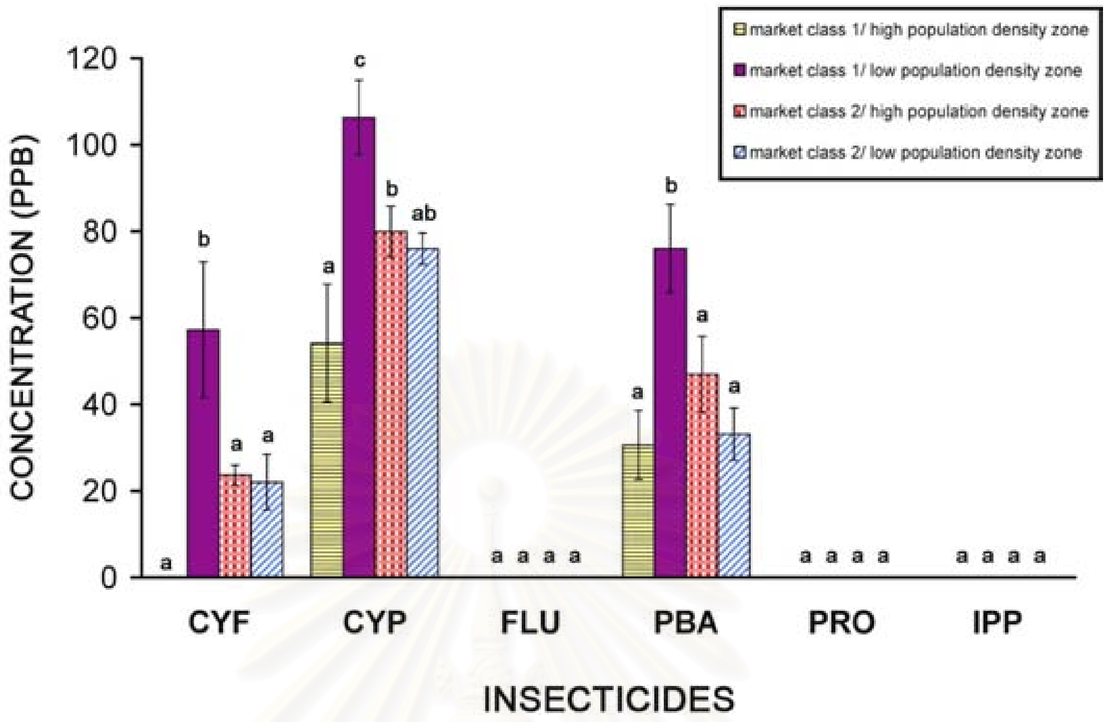

Figure 6.3 Comparisons of means (± SE) concentrations (ppb) of the insecticidal residues in the German cockroach samples caught from the 4 combination groups of Bangkok markets (CYF = cyfluthrin, CYP = cypermethrin, FLU = flumethrin, PBA = 3- phenoxybenzoic acid, PRO = propoxur, and IPP = 2-isopropoxyphenol). The bar graphs in each insecticide with different letters are significantly different at $p < 0.05$, *Mann-Whitney U*-test.

Discussion and Conclusion

Several important conclusions may be derived from the data such as the differences of insecticidal residue concentrations in the living German cockroaches and the differences concentrations in twelve Bangkok markets. The most notable result was the differences of insecticidal residue concentrations in the living German cockroaches. They were significantly different among these insecticide concentrations in the living cockroach samples.

For the concentration of insecticidal residues, the highest concentration (ppb) of insecticidal residue in the living cockroach samples was cypermethrin, followed by 3-phenoxybenzoic acid and cyfluthrin, respectively. The concentration of cypermethrin was higher than the other compounds in every market. The high concentration of cypermethrin in the living German cockroach samples may be caused by the insecticides which the market vendors in the markets have applied in their shops. Cypermethrin is a popular active ingredient formulated in many insecticides. Moreover, by the personal interview with the twelve market owners, only the market owner of market number 6 has employed a cockroach control company to eradicate the cockroach in the market every 3 months. The company eradicated the cockroaches using cypermethrin. Additionally, the high amounts of cypermethrin and 3-phenoxybenzoic acid in the living cockroach samples agree with Wauchope *et al*. (1992) who stated that the movement rating of cypermethrin is extremely low (by water solubility: 0.004 mg/l), but its sorption coefficient (Koc) is high (Koc = 100,000). These characters, the water solubility and the high sorption of cypermethrin, may cause the cypermethrin persist and last for a period in the treated areas.

Moreover, a life cycle the German cockroach may be completed within about 100 days under favorable environment conditions (Ross and Mullins, 1995). Thus, the German cockroach population may be recovered after being treated with cypermethrin especially in market number 6 which cockroaches were treated with cypermethrin every 3 months by the cockroach control company. During the study period, the highest numbers of the German cockroaches were in market number 12 and market number 6. However, the highest concentration of cypermethrin was detected from market number 6. This suggests that cypermethrin resistance may have developed in the German cockroach populations after being treated by the repetitious insecticide.

Matsumura (1976) stated that cypermethrin was rapidly metabolized in the insect body by two routes of metabolism such as oxidative degradation and hydrolytic degradation. The metabolism was generally increased with high temperature (Wadleigh *et al*., 1991). Then, a metabolite such as 3-phenoxybenzoic acid, less toxic than the parent compound, was produced and excreted (Yamamoto *et al*., 1969; Matsumura, 1976; Leahey, 1985; Class, 1992). In this study, the resistance mechanisms of the German cockroaches may be physiological resistance and/ or behavioral resistance. Siegfried and Scott (1992) stated that the major resistance mechanism on the German cockroach was physiological resistance. The physiological resistance mechanisms of the German cockroach on cypermethrin are the reduction of cuticular penetration, the increase of metabolic detoxification, and the target site insensitivity (Siegfried and Scott, 1992).

The resistance mechanism of the German cockroach to cypermethrin by reducing of cuticular penetration was documented by Bull and Patterson in 1993. They reported that penetration of [^{14}C]-cypermethrin into the body was reduced when the chemical had been applied topically in a cypermethrin-resistant strain of the German cockroach.

The resistance mechanism of the German cockroach to cypermethrin by increasing metabolic detoxification was documented in many reports. Hemingway *et al*. (1993) stated that fifteen strains of the German cockroach resisted to cypermethrin due to overproduction of enzymes involving to the insecticidal detoxification such as esterases. Prabhakaran and Kamble (1993) also reported that the German cockroach moderate resistance to cypermethrin by using the activity of esterase enzymes. Valles, Dong, and Brenner (2000) reported that the German cockroach also resisted to cypermethrin by increasing levels of cytochrome P_{450}, the most important component of a monooxygenase system. This detoxification let the German cockroach promoting its excretion (Hemingway *et al*., 1993; Scharf *et al*. 1997; Valles *et al*., 2000).

In addition, the resistance mechanism of the German cockroach to cypermethrin by target site insensitivity was studied by many scientists. For example, Lui and Plapp (1991) stated that the reduction in binding sites on the voltage-sensitive sodium channels, the primary target of pyrethroid insecticides, was associated with metabolic resistance in the insect. Xu *et al*. (2006) also reported that there was a strong correlation between knockdown resistance allele (*kdr*-allele) expression and the levels of pyrethroid resistance in the German cockroach.

For the concentration of cyfluthrin in the living German cockroach samples may be also from the insecticide usage by the market vendors in the twelve Bangkok markets. Cyfluthrin is one of a popular active ingredient formulated in many insecticides which generally used by people in this study area. The occurrence of cyfluthrin in the living cockroach samples agree with Wauchope *et al*. (1992) who stated that the movement rating of cyfluthrin is extremely low (by water solubility: 0.002 mg/l), but its sorption coefficient (Koc) is high (Koc = 100,000). These characters, the water solubility and the high sorption of cyfluthrin, may also cause the cyfluthrin persist and last for a period in the treated areas.

During the study period, the highest numbers of the German cockroaches were in market number 12 and market number 6. However, the highest concentration of cyfluthrin was detected from market number 6. This suggests that cyfluthrin resistances may have developed in the German cockroach populations in this study area. The cyfluthrin degradation and the resistance mechanisms of the German cockroach on cyfluthrin are similar to cypermethrin (Matsumura, 1976). The German cockroach resistances on cyfluthrin were reported by many scientists (Cochran, 1989; Atkinson *et al*., 1991; Scharf *et al*., 1996).

In this study, the concentration of flumethrin in the living German cockroach samples was below the LOD. It indicates the rarely use of flumethrin in this area. Moreover, a small amount of flumethrin in the cockroach samples may be due to its low stability in air and light based on its short half-life in soil (less than 20 days) (Kaufman *et al*., 1981; Wauchope *et al*., 1992).

In this study, the concentrations of the propoxur and its metabolite residues were also detected at below LOD. It may be because the German cockroaches suddenly died after contacting to the propoxur, thus, these cockroaches were not caught in the traps during the study period. Propoxur had high toxic to insect, it was classified in highly toxic group to honey bees, *Apis melifera*, the LD_{50} of propoxur was less than 2.0 µg/ bee (Sanford, 1993). Additionally, the low amounts of propoxur and 2-isopropoxyphenol in the living cockroach samples may be due to their low stability in air and light (Kaufman *et al*., 1981). It agrees with Wauchope *et al*. (1992) who stated that the movement rating of propoxur was high (by water solubility: 1,800 mg/l), but its sorption coefficient (Koc) is low (Koc = 30). Thus, propoxur may not persist in the treated area. Moreover, the propoxur was rapidly absorbed in the target insects and readily inhibited cholinesterase of insects so that insects were died rapidly (Matsumura, 1976).

Overall, the highest concentrations of cypermethrin, 3-phenoxybenzoic acid, and cyfluthrin in the living German cockroach samples were detected from the low population density zone/ market class 1 combination (LC1). This indicates that the market vendors often use the two kinds of insecticides, cypermethrin and cyfluthrin, in the markets. Although these markets had regularly sanitary cleaning, cypermethrin, 3-phenoxybenzoic acid and cyfluthrin were detected in large amounts. This may be because their low solubility in the water that can make difficulty to wash out. However, the highest numbers of the German cockroaches were caught from this combination group of Bangkok market. It reveals that the German cockroaches in this combination group of Bangkok market may have developed the resistance to these insecticides in their populations.

In conclusion, the unsuitable insecticide treatment may induce the insecticide resistance in the German cockroach populations. Thus, the information such as the proper of insecticide usage, rotation of insecticide usage, and the cockroach behavior should be clarified to the market vendors and the local people. The heavy infestations of the German cockroaches can be dealt by chemical control measures. Moreover, in the future, bioassay should be conducted to confirm the insecticide resistance in the German cockroaches.

CHAPTER 7

CONCLUSION

This research consists of four parts of the ecology, behavior, bacterial harborage, and insecticidal residues in the German cockroach, *Blattella germanica* Linnaeus, in Bangkok markets. The objectives of these experiments include studying the attractiveness of various foods to the male and female German cockroaches and studying of their feeding behavior, monitoring population dynamics, isolating prevalent and human pathogenic bacteria, and examining the insecticidal residues from the living German cockroach.

The infestation of the German cockroaches was a serious problem, the attractiveness of some non-chemical baits for trapping the German cockroach in the field and the feeding behavior of them were studied. Adult male and female German cockroaches were used in this experiment. These 48-h-starved cockroaches were given choices among eight food items (carbohydrate–rich foods: bread, sugar, banana, and potato; and protein-rich foods: peanut, cheese, pork, and cat food). Modified eight-chamber-olfactometers were used for this food preference experiment. Each kind of food was filled in each chamber of the olfactometer with 15 of each sex of the starving cockroaches placed at the center. The olfactometers were observed for 48 hours in the laboratory. The cockroaches were checked every 1, 4, 16, 24, 28, 40, and 48 hours; and the amount of food eaten was recorded using the Rodgers's index for indicating the food preference of the cockroaches. The male German cockroaches significantly preferred banana and potato whereas the female cockroaches significantly preferred only banana. Additionally, the female cockroaches also significantly preferred peanut, sugar, and cat food more than the males. For the feeding behavior, the foraging time of the German cockroach occurred at night with two peaks of feeding activity. The first peak occurred during the hours between 07.00-10.00 pm and the second peak occurred during 04.00-05.00 am. Therefore, the cockroach trapping should be done at night. Carbohydrate-rich foods mixed with protein-rich foods with strong odor are good baits for trapping the German cockroaches in the fields.

A study on the population dynamics of the German cockroach was conducted in twelve Bangkok markets from March 2005 to March 2006 using stratified random sampling. The selected market areas (12 markets) were investigated in two different human population density zones and in two different classes of the markets. The selected market areas were conducted monthly during the study period. Twenty modified jar traps filled with cat food saturated with beer and ground peanut were placed in 3 kinds of shops (butcher shop, vegetable shop, and grocery) in each market.

The two highest peaks of the German cockroach were in July and August whereas the two lowest peaks of the cockroach were in December and January. The population of the German cockroach started to increase in April, followed by a rapid decrease in September. The highest catch number of the German cockroach was the large nymphal stage (the 5^{th} and 6^{th} instars). The temperature and relative humidity may be the important factors influencing on the cockroach reproduction (Ross and Mullins, 1995). The highest densities of the German cockroaches were in market number 12 (poor sanitary market) and market number 6 (good sanitary market). The highest number of the German cockroach caught was in the combination of market class 1/ low population density zone (LC1), followed by market class 2/ low population density zone (LC2), market class 2/ high population density zone (HC2), and the lowest was in market class 1/ high population density zone (HC1). The number of cockroaches caught from two classes of Bangkok markets was not significantly different while the cockroach number caught from the low human population density zone was significantly higher than the high human population density zone. Moreover, the cockroach number caught from the groceries was significantly higher than the other shops in the twelve Bangkok markets.

The prevalent and human pathogenic bacteria of the German and the American cockroaches in twelve Bangkok markets were investigated. The experiment was conducted in April, July 2005, and January 2006. Cockroaches were trapped using the modified jar traps. Aerobic and facultative anaerobic bacteria were investigated. The bacteria were isolated from 2 parts of the German and the American cockroaches such as the external cuticles and the middle guts. The German cockroach hosted 21 species of bacteria whereas the American cockroach hosted 26 species. In this study, the serious pathogens such as *Bacillus cereus*, *Salmonella arizona* and *Staphylococcus aureus* were found. The opportunistic pathogens such as *Pseudomonas aeruginoga*, the important bacteria in hospitalized patients, were found in all markets. Furthermore, *Escherichia coli* which is an indicator of environmental surveillance as a measurement of human and warm-blooded animal fecal contamination was also found from all markets.

The bacterial species isolated from the cockroaches from the poor sanitary markets was significantly higher than the good sanitary markets. Moreover, the bacterial species isolated from the external cuticle of cockroaches was significantly higher than isolated from the middle gut. The species richness of bacteria isolated from the cockroaches in April, July 2005, and January 2006 were not significantly different during the study period. The results show that the cockroaches carried many bacteria throughout the year. The similarity coefficient of the bacterial species between the German and the American cockroaches investigated in twelve Bangkok markets were ranged from 0.870 to 1.00. The results indicate that the both kinds of cockroaches had similar efficiency on carrying bacteria.

The cockroach control needs the information about insecticidal residues. The experiment of the evaluation of some synthetic pyrethroid and carbamate residues in the German cockroach was conducted from March 2005 to March 2006. The insecticidal residues were analyzed by High Performance Liquid Chromatography (HPLC). The highest concentration (ppb) of insecticidal residue was cypermethrin, followed by 3-phenoxybenzoic acid and cyfluthrin, respectively. In this study, the concentrations of flumethrin, propoxur, and 2-isopropoxyphenol in the German cockroach samples were below the LOD. During the study period, a large number of the German cockroaches was caught in market number 6 which was classified as the good sanitary market (class 1 market). Moreover, the highest concentrations of cypermethrin, 3-phenoxybenzoic acid, and cyfluthrin were also detected from market number 6. This implies that the German cockroaches have the resistances to cypermethrin and cyfluthrin. Cypermethrin and cyfluthrin resistances may have developed in the German cockroach populations in this market after being treated by the repetitious insecticide. The short life cycle of the German cockroaches leading to rapid population recovery (Ross and Mullins, 1995) and the unsuitable insecticide treatment could be developed the insecticide resistance in the German cockroach populations.

From the results, the highest numbers of the German cockroaches were in market number 12 which was classified as the poor sanitary markets (class 2 market). The high density of the cockroaches may be caused by the suitable temperature, proper humidity, poor sanitation, scarce cleanliness, and the old structure building which has several cracks and crevices.

This research provides the basis for the German cockroach control strategies in Bangkok markets. Like all animals, cockroaches need food and water to survive in and around structure. By reducing access to food and water, cockroach populations become stressed to survive in urban environments (Benson and Zungoli, 1997).

Virtually no situation exists indoors where cockroach food in some form is not available. Thus, proper storage of food and good sanitation will not starve cockroaches. When sanitary control is integrated with chemical control, the efficiency of the German cockroach control will occurred (Gold, 1995; Koehler *et al*., 1995; Benson and Zungoli, 1997).

For example, when the German cockroaches have low infestation in the markets, the elimination of food sources should be done such as (1) washing utensils immediately after use will prevent cockroaches from consuming food residue on those utensils, (2) sealing garbage can lids to prevent cockroaches from accessing food sources, (3) emptying indoor trash containers frequently, and (4) keeping plastic bags lining trash containers closed with twist ties to prevent cockroaches from being attracted to the garbage area. Elimination of harborage such as (1) sealing cracks and crevices in the markets, (2) keeping clutter of newspapers, bags, crate, and utensils from accumulating, and (3) frequent cleaning the market may result limiting factor of food and harborage for the cockroaches. The cockroaches forage for food, thus, baits or traps should be used for control them. The modified jar trap containing carbohydrate-rich foods mixed with protein-rich foods with strong odor may be selected as good baits for trapping the German cockroaches. Moreover, the cockroach trapping should be done at night.

The heavy infestations of the German cockroaches can be dealt by environmental management such as the elimination of food sources and elimination of harborage as mentioned above. The insecticide treatment in December and January should be considered. The treatment in the lowest peaks of the German cockroach species may decrease the insecticidal usage because the German cockroach density is still low. The rotation of insecticide uses is another important way for the market vendors and the cockroach control companies to prevent the insecticidal resistance in the German cockroach.

Furthermore, the long-term German cockroach control should be planned and done. Monitoring and evaluating on the German cockroaches should be conducted at least twice a year. The local people especially the market owners and market vendors should be continuously educated about the German cockroach biology, behavior, and ecology.

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APPENDICES

APPENDIX A

Inspection Form for Environmental Sanitation

Name of Market Place……………………….. Owner/ Manager…………………….……….

Location……………………………………………District……………………………….…...

Established in year……………………..….Last improved in year……………………..….

Please fill ✓ in the correspondence box.

No.	Details	Yes	No
1	Building are of suitable heights and the structure are constructed from fire-resistant, strong, and durable material. The components (i.e. floors and walls) are made from strong durable materials which are easy to clean.		
2	Goods, products, and any other materials are orderly placed.		
3	There are enough clean garbage pails.		
4	The sewage system is well run, and does not clog up.		
5	The market is regularly cleaned.		
6	There is enough clean tap water.		
7	The market and its surrounding do not have stagnant water.		
8	The toilets are separated for men and women.		
9	Rodent and vector controls are regularly done.		
10	Food and their materials are placed at least 60 cm above the floor.		
11	Ventilation system in the market is sufficient.		
12	There is no garbage on the floors, sewers, and sidewalks.		
13	There is a closed-sewage system with sewage covers.		
14	The toilets do not smell and are ventilated.		
15	The septic tanks are sanitary and undamaged.		
16	The market has people responsible for maintaining the market's sanitation, order, and safety.		
17	Sanitary washing are held at lease once every month.		

APPENDIX B

Interview

ID…………………

Date ………………………… Market number …………..………

Questions

1. Have you ever eliminate cockroaches by chemical insecticides in your shop?

□ Yes □ No (skip questions to number 6)

2. Which type of chemical insecticides do you use?

□ Toxic baits □ Toxic dusts/ chalk □ Chemical aerosols

3. Which trade name of insecticides do you use?

Toxic baits

□ Canbic®

□ Other (define)………

Toxic dusts/ chalk

□ ARS chalk®

□ Chalk Era Plus®

□ Raid® chalk

□ Other (define)………

Chemical aerosols

□ ARS® 3

□ ARS® gold

□ ARS® mite plus

□ Aswin®

□ Baygon® green

□ Baygon® green-orange

□ Raid® (water)

□ Shelldrite® for termite and cockroach

□ Shieldtox® altra3

□ Shieldtox® green

□ Shieldtox® for termite and cockroach

□ Other (define)……………………

Questions (cont.)

4. How often do you use them?

□ Every day

□ Every week

□ Every month

□ Every 6 months

□ Do when see them

5. How much money do you spend on cockroach control per month (Baht)?

□ Less than 100 Baht/ month

□ 100 - 300 Baht/ month

□ 301 - 500 Baht/ month

□ More than 500 Baht/ month

6. Why don’t you eliminate cockroaches by chemical insecticides in your shop?

□ Cost of the insecticides is high

□ The awareness of insecticidal toxicity

□ Inefficiency of insecticides

□ Other (define)……………………….

BIOGRAPHY

Mrs. Prachumporn Lauprasert was born on 3rd August, 1974 in Udon Thani Province. She received a Bachelor's degree of Science (Biology) in 1996 from the Faculty of Science, Khon Kaen University. She received a Master's degree of Public Health (Environmental Health) in 1999 from the Graduate School, Khon Kaen University. She continued her study for the degree of Doctor of Philosophy in Biological Science, Faculty of Science, Chulalongkorn University in 2003 and completed the program in 2006. She published her partial dissertation in the topic of "Food Preference and Feeding Behavior of the German Cockroach, *Blattella germanica* (Linnaeus)" in Journal of Scientific Research Chulalongkorn University. 31 (2):121-126.

At the present, she works as a lecturer at the Faculty of Public Health, Mahasarakham University, Thailand.