

## A preliminary study on insects associated with pig (*Sus scrofa*) carcasses in Phitsanulok, northern Thailand

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**Abstract.** A preliminary study on insects associated with pig carcasses was conducted in Phitsanulok, northern Thailand. Five decomposition stages of pig carcasses were categorized: fresh (0-1 day after death), bloated (2 days after death), active (3 days after death), advanced (4-6 days after death) and dry (7-30 days after death). The arthropod species collected from the corpses in the field sites were mainly classified belonging to two orders and nine families, namely order Diptera (family Calliphoridae: *Chrysomya rufifacies* and *Chrysomya megacephala*, family Muscidae: *Musca domestica*, family Fanniidae: *Fannia canicularis*, family Sarcophagidae: *Parasarcophaga ruficornis* and family Piophilidae: *Piophila casei*.) and order Coleoptera (family Dermestidae: *Dermestes maculatus*, family Histeridae: *Hister* sp., family Cleridae: *Necrobia rufipes* and family Trogidae: *Trox* sp.). The forensically dominant fly was *C. rufifacies*, while the beetle was *D. maculatus*. The beetles associated with pig carcasses found in this study are first reported in Phitsanulok, Thailand. In addition, ants, bees, spiders and millipedes were also associated with the carcasses. These findings may provide data for further use in legal investigations in Thailand.

### INTRODUCTION

Forensic entomology is the use of insects to estimate the time since death, or postmortem interval (PMI), based on the developmental rates of the flies and the successional patterns of specific insects on the corpses. Most experimental studies on forensic entomology were conducted in countries (Oliva, 2001; Wolff *et al.*, 2001; Grassberger *et al.*, 2003) where the environment, temperature and moisture are not similar to Thailand. Here, many aspects of medically important insects have been reported (Sucharit *et al.*, 1976; Lertthamnongtham *et al.*, 2003), but little is known about forensic entomology which is important to assist the investigations of crime scenes. At present, there are at least 2 reports concerning the flies associated with human corpses in

Thailand (Sukontason *et al.*, 2001, 2005). On the basis of little information about forensic entomology of the country, the present research was designed to observe the pig decomposition and to study the insects associated with its carcasses in Phitsanulok province, north of Thailand. This information may provide the data useful in forensic investigations in Thailand.

### MATERIALS AND METHODS

The present study was conducted in Phitsanulok, northern Thailand. Phitsanulok province is located approximately between 16°N, 101°E. The region is about 44 m. above sea level and has average temperatures of 26.4, 30.5 and 27.4°C in the cold season (November-February), hot season (March-

May) and rainy season (June-October), respectively. The average annual rainfall is about 1400.8 mm. Two outdoor sites selected for this study were located near Naresuan University. The first site was directly exposed to the sunlight during most of the day, whereas the other was more shaded during the daytime.

This study was approved by the animal-ethics committee of Naresuan University. The pigs were purchased from a farm in Phitsanulok province and were 3 and 4 kg in weight. They were killed with sharp-edge materials from a slaughter-house and immediately transported to the field sites. Each pig was placed inside a metallic cage (60 x 80 x 100 cm. with 2.5 cm. mesh) which opened at the bottom to allow the carcasses contact with the ground while also preventing the carcasses from being eaten by scavengers.

Adult insects associated with the carcasses were collected by using insect nets, and fly eggs, maggots and pupae were collected with forceps. Collections from the bodies took place daily during the first week after placement, and every three days after that until the bones were completely exposed. The eggs and maggots collected from the field sites were rapidly preserved directly in 70% ethyl alcohol. In the laboratory, adult insects were killed by placing them in bottles containing ethyl acetate and then pinned. Pupae collected from the corpses were reared to adult stage in cages. Most specimens were identified according to Byrd & Castner (2001) and Greenberg & Kunich (2002). At the time of collection of specimens, the data on relative humidity was measured by using digital thermo hygrometer (605-H1 Miniature Thermo hygrometer); for temperature and accumulative rainfall, Thai Meteorological Department data was used.

## RESULTS

During the 30 day study period, from mid-February to mid-March 2005, the minimum and maximum temperatures were 22.4 and 35.5°C. The average humidity was 68.7%, and

the cumulative rainfall was 5.7 mm. Decomposition of pigs was determined by five stages as fresh, bloated, active, advanced and dry. In the fresh stage (0-1 day after death), the outside appearance of the bodies was similar to those of normal pigs, but the inside had begun to decay because of cellular death and microbial activities. In the bloated stage (2 days after death), the abdomen of the pig became swollen and blackened, and the intestine was expelled out of the body. In the active stage (3 days after death), the body of the pig was disintegrating, emitting very strong odors, while the advanced stage (4-6 days after death) was characterized by minor odor and the removal of the soft tissues. In the dry stage (7-30 days after death), the odor was very minor or absent, and the bones were exposed.

A total of 672 individual insects were collected from both sites and classified. They belonged to 2 orders, Diptera and Coleoptera, and 9 families: Calliphoridae: *Chrysomya rufifacies* and *Chrysomya megacephala*, Muscidae: *Musca domestica*, Fanniidae: *Fannia canicularis*, Sarcophagidae: *Parasarcophaga ruficornis*, Piophilidae: *Piophila casei*, Dermestidae: *Dermestes maculatus*, Histeridae: *Hister* sp., Cleridae: *Necrobia rufipes* and Trogidae: *Trox* sp. The dominant family was Calliphoridae (Table 1). The pupae collected from the field sites were hatched out to adult stage inside cages, and all of them were identified as *Chrysomya rufifacies*. The insects associated with each stage of the decaying pigs are shown in Table 2. Ants, not normally considered to be a forensically significant species, were the first arthropod attracted to the wound around the neck of pig. *Chrysomya* spp. were the first flies that were attracted to the pig carcasses for laying eggs. In the advanced stage, adult flies were absent. Most adult Diptera were attracted to the pig carcasses in the early stage of decomposition process, while Coleoptera were seen in advanced and dry phases. In addition, other arthropods such as bees, spiders and millipedes were also associated with pig carcasses.

Table 1. Species of insects collected from pig carcasses in Phitsanulok, northern Thailand

Order	Family	Genus/species	No. of insects collected		Total (%)
			Pig carcass I (%)	Pig carcass II (%)	
Diptera	Calliphoridae	<i>Chrysomya rufifacies</i>	202 (30.06)	387 (57.59)	589 (87.65)
		<i>Chrysomya megacephala</i>	25 (3.71)	34 (5.05)	59 ( 8.76)
	Sarcophagidae	<i>Parasarcophaga ruficornis</i>	2 (0.30)	7 (1.04)	9 ( 1.34)
	Muscidae	<i>Musca domestica</i>	0	4 (0.60)	4 ( 0.60)
	Fanniidae	<i>Fannia canicularis</i>	0	3 (0.45)	3 ( 0.45)
	Piophilidae	<i>Piophila casei</i>	1 (0.15)	0	1 ( 0.15)
Coleoptera	Dermestidae	<i>Dermestes maculatus</i>	2 (0.30)	1 (0.15)	3 ( 0.45)
	Histeridae	<i>Hister sp.</i>	0	1 (0.15)	1 ( 0.15)
	Cleridae	<i>Necrobia rufipes</i>	1 (0.15)	0	1 ( 0.15)
	Trogidae	<i>Trox sp.</i>	0	2 (0.30)	2 ( 0.30)
Total			233 (34.67)	439 (65.33)	672 (100)

Table 2. Species of arthropods associated with each stage of decaying pigs

Genus/Species	Stages of Decaying pig				
	Fresh (0-1 day)	Bloated (2 days)	Active (3 days)	Advanced (4-6 days)	Dry (7-30 days)
<i>Chrysomya rufifacies</i>	A	E, I, A	E, I, A	I	I
<i>Chrysomya megacephala</i>	A	A	A	—	—
<i>Parasarcophaga ruficornis</i>	A	A	A	A	—
<i>Musca domestica</i>	A	A	—	—	—
<i>Fannia canicularis</i>	—	A	—	—	—
<i>Piophila casei</i>	—	—	A	—	—
<i>Dermestes maculatus</i>	—	—	—	A	A
<i>Hister sp.</i>	—	—	—	A	—
<i>Necrobia rufipes</i>	—	—	—	A	—
<i>Trox sp.</i>	—	—	—	A	A
Spiders (not identified)	—	A	A	A	A
Bees (not identified)	—	A	—	—	—
Ants (not identified)	A	A	A	A	A
Millipedes (not identified)	—	—	—	A	A

Note: A=Adult, E=Egg and I=Immature.

## DISCUSSION

The present study and study by Wolff *et al.* (2001) revealed that the decomposition stages of pig carcasses was classified as fresh, bloated, active, advanced and dry, but they were different in the decomposition

rates. In addition, several studies on stages of decomposition in human cadavers (Arnaldos *et al.*, 2004) and rabbit carcasses (Tantawi *et al.*, 1996; Bharti & Singh, 2003) agreed with our study. In contrast, study in Malaysia by Lee & Marzuki (1993) indicated the decomposition of monkeys can be

divided into 4 stages as fresh stage, decay stage, dry stage and remain stage. However, it is difficult to distinguish each phase from the other. Possible reasons for differences in the decomposition rates may be due to the temperature variations. Temperature is one of the extrinsic factors that are important for the development of bacteria (Campobasso *et al.*, 2001). Other factors affecting decom-position rates include the age of corpse, constitution, cause of death, ventilation and humidity (Campobasso *et al.*, 2001).

The insect species collected from pig carcasses in Pithsanulok, northern Thailand, were *C. rufifacies*, *C. megacephala*, *P. ruficornis*, *M. domestica*, *F. canicularis*, *P. casei*, *D. maculatus*, *N. rufipes*, *Trox* sp. and *Hister* sp. This finding agreed with the previous reports on the study of vertebrate carcasses (Tantawi *et al.*, 1996; Wolff *et al.*, 2001; Bharti *et al.*, 2003; Arnaldos *et al.*, 2004), but a small number of insect species associated with the corpses were shown in this study. The predominant species collected in this study was *C. rufifacies* which has been first used in PMI determination in the floating corpse in Lumpang province, northern Thailand (Sukontason *et al.*, 2005). This indicated that *C. rufifacies* was the forensically important fly in the north of Thailand. Factors such as climate, season, sun exposure, urban or rural scenarios, frequency of collection and the number of animal models can affect the species diversity of insect associated with the corpses in several regions of the world (Anderson, 2001; Campobasso *et al.*, 2001). Interestingly, when the pupae collected from the field sites were reared in the laboratory, the emerged flies were found only to be *C. rufifacies*. The reason for this was due to the effect on larval population density and interactions among species in the same ecological habitat. This was clarified by Goodbrod & Goff (1990) who demonstrated that the larvae of *C. rufifacies* were cannibalistic and predatory on *C. megacephala* larvae after the first instar.

Several species of beetles associated with corpses have been found in many parts of the world, but this study was the first to

report about beetles (*D. maculatus*, *N. rufipes*, *Trox* sp. and *Hister* sp.) associated with the pig carcasses in Phitsanulok, Thailand. They have also been collected from vertebrate cadavers in Brazil (Carvalho *et al.*, 2000), Columbia (Wolff *et al.*, 2001), India (Kulshrestha & Satpathy, 2001; Bharti & Singh, 2003), Australia (Archer & Elgar, 2003), Spain (Arnaldos *et al.*, 2004), Argentina (Oliva, 2001) and the United States of America (Watson *et al.*, 2003; Tabor *et al.*, 2004, 2005). This indicates that beetles may be used for legal investigations in Thailand.

Form the previous study in Spain (Arnaldos *et al.*, 2004), the ecological relationship between the insects and the corpses were categorized as necrohpagous, necrophilous and omnivorous. In this study, this was also observed, that is, necrophagous species which feed only on decaying tissues are *Chrysomya* spp. *M. domestica*, *P. ruficornis*, *F. canicularis*, *P. casei* and *D. maculatus*; necrophilous species which prefer to feed on decaying tissues and necrophagous group are *Hister* sp. and spiders; and omnivorous species which feed on decaying tissues are *N. rufipes*, ants, bees and millipedes. This indicated that the roles of ecological systems of insects associated with corpses in Thailand are similar in Spain.

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