# An Analysis of Forensic Entomological Specimens by Universiti Kebangsaan Malaysia

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#### **SUMMARY**

This study reviews forensic entomological specimens analysed by the Department of Parasitology & Medical Entomology, Universiti Kebangsaan Malaysia for the year 2004. A total of 10 cases (6 males and 4 females) were observed for the entomological specimens. Various types of death scenes were obtained including indoor and outdoor area such as bushes field, rubbish dumping site, and aquatic areas. Identified fly species collected from the death sites were blow flies, *Chrysomya megacephala*, *Chrysomya rufifacies* and *Lucilia cuprina* and unknown sarcophagid larvae, with *Ch. megacephala* being the most common species found in the ecologically varied death scene habitats. The post-mortem interval (PMI) estimation ranged from one to five days, based on the entomological specimens collected.

## **KEY WORDS:**

Forensic entomology, Post-mortem interval, Decomposition, Malaysia

#### INTRODUCTION

Human corpses, whether they have been produced naturally or as the result of foul play, are processed by insect decomposers in the same manner as any other carrion. An analysis of forensic entomological specimen, therefore, is based on the study of insects which sequentially colonize a corpse as decomposition progresses and on the rates at which the various stages of their offsprings develop. Knowledge of the distribution, biology and behaviour of these insects, which feed upon the dead body, may assist forensic investigation by providing information on when, where and how a person died or a crime was committed<sup>1</sup>. However, the most important application is to determine post-mortem interval (PMI)<sup>2</sup>. This is due to the nature of nearby flies which can detect odour within minutes from dead body and eggs deposition is followed by other arthropods in a certain sequence or succession3. This paper reviews the entomological specimens analysed by the Forensic Entomology Laboratory, Universiti Kebangsaan Malaysia, throughout the year 2004.

## **MATERIALS AND METHODS**

#### **Specimen collection**

Entomological specimens were received from the Hospital Universiti Kebangsaan Malaysia (HUKM) in Kuala Lumpur and Hospital Ipoh (HI) in Perak. All larval specimens were received in two separate containers, in which, one of them contained preserved larval specimens in 70% alcohol and the other with live larvae to be reared into adults. Each container was labelled with hospital identification number, police report number, date and time of collection, name of collector, short notes of the case including name of deceased (if known). A request form describing the case and signed by the attending forensic pathologist was received along with the specimens. Specimens received were recorded in a report book.

Immature stages of flies (eggs or larvae) from the decomposing bodies were collected by the attending pathologist based on the method described by Catts & Haskell<sup>3</sup> and Smith<sup>4</sup>.

## Preparation of preserved larval samples

The preserved larvae samples were prepared according to the method described by Omar et als. Larvae were first placed in 70% alcohol for at least three days, followed by transverse excision at the posterior part of each larva. Both posterior and anterior parts were then immersed in 10% potassium hydroxide (KOH) overnight, before being neutralized in glacial acetic acid for 7 to 10 minutes. All the internal organs were thoroughly removed by forceps and the specimens were then dehydrated in ascending concentrations of alcohol solutions (80%, 90%, 95% and 100%) for 30 minutes each. The specimens were then soaked in clove oil and followed by xylene for 30 minutes each before mounted onto a glass slide using Canada balsam. The instar and species of the larva was examined under light microscope for identification based on taxonomic keys by Ishijima<sup>6</sup> and reference calliphorids collections at the Forensic Entomology Laboratory, Universiti Kebangsaan Malaysia.

# Preparation of live larval samples

In the laboratory, viable eggs or larvae obtained were maintained using beef liver *ad libitum*. The wandering larvae were then transferred to a dry plastic container (measuring 10cm x 10cm x 6cm) filled with sawdust approximately ¼ of the container. About 30 larvae were placed in each container to be reared to adulthood. The emerged adults were pinned and identified using the keys provided by Kurahashi *et al*<sup>7</sup>.

#### **Estimation of post-mortem interval (PMI)**

For preserved specimen, PMI was estimated by referring the age of larvae collected from corpses to developmental growth

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data of particular species reared in laboratory and previous succession studies³. Collected living specimens were reared in laboratory (temperature  $29 \pm 1$ °C, relative humidity 65-70%, photoperiod (L: D)(h)12:12) to determine the duration of insects' life cycle. This information was compared to their known total life cycle duration (egg to adult) to estimate the date and time of oviposition³.

However, final estimation of PMI for each case was done after considering other factors that might influence the development of flies on corpses. These include condition of corpses, environmental conditions of the death locations and accessibility of insects to the dead bodies<sup>8, 9, 10</sup>.

## Case histories of corpses sent for autopsy

## A. Hospital Universiti Kebangsaan Malaysia (HUKM), Kuala Lumpur

There were eight cases received from HUKM throughout the year 2004.

#### Case 1:

A corpse of a 25-year-old Chinese man was found hanged on 28 January 2004 in his suburban apartment with early sign of decomposition. He was last seen on 26 January 2004.

#### Case 2:

A 31-year-old Vietnamese man was found dead in his house on 11 February 2004 with splashes of blood in the room. The victim had died due to multiple stab wounds. The corpse was at advanced decompositional state with few fly eggs and larvae were found, particularly on the orifices and the wound sites.

#### Case 3:

On 14 February 2004, the body of a 29-year-old Chinese lady was found dead on the mattress in her flat of an urban building. The police officers were informed after her neighbours smell bad foul odour came from the house. On the external examination, the body was in an advanced decomposition with larvae found from her nose and hairs. There was no marked injury seen on the face or head.

#### Case 4:

On a night of 6 May 2004, police were called to investigate a foul smelling odour emanating from a single home in an urban area. Investigating officers soon discovered the badly decomposed body of a 77-year-old Chinese man on the floor of dining room. The body was surrounded by maggots and examination revealed no suspicious injury on the body. Cause of death was most probably from coronary occlusion due to atherosclerosis.

# Case 5:

A corpse of a 51-year-old Chinese man was found in his flat on the second floor of an urban building. The body was found on 8 May 2004 after police received a public report regarding a foul smell coming from the house. Examination showed severely decomposed body infested with maggots particularly in the natural orifices of the body, with no external injury

suspected. It was assumed that the man died due to natural disease.

#### Case 6:

The remains of an unknown murder victim were found at a quiet obscured place near some bushes in a suburban area of Kuala Lumpur. Examination showed decomposed body full of maggots. The body was partly sketalised with missing left upper limb and detached skull. The trunk and abdomen were decomposed with a gaping cavities probably caused by animal. The victim had died of slashed wounds over the lower back and severe injury at the right mid chest ribs.

#### Case 7:

On 16 July 2004, the corpse of an unknown young adult female was found at the rubbish dumping site near a rural area in Hulu Langat, Selangor. Examination showed early decomposed body surrounded by maggots with numerous contusions on the body and limbs, probably due to battling. Examination showed early decomposed body surrounded by maggots with soft tissue patterned injuries over the body and limbs. Further autopsy revealed that she had died due to head injury.

#### Case 8:

Similar to the cases 4 and 5, the body of a 50-year-old Chinese man was found dead in his own house situated at an urban area of Kuala Lumpur. The police obtained a complaint of foul odour smell from neighbours of the house. The body was in an advanced decompositional state and was surrounded by maggots. Large numbers of eggs were found in several areas such as nasal orifice, skin folds of the neck and clothing. It was believed that he died due to natural cause.

#### B. Hospital Ipoh, Perak

There were two cases received from Hospital Ipoh in 2004.

## Case 9:

The remains of a young female (approximately 25 to 35-year-old) were found floating in a drain in rural area on 26 June 2004. The body was severely decomposed and surrounded by maggots. There were signs of animal bites on the right arm and both legs.

#### Case 10:

On the afternoon of 27 July 2004, a half-naked body of a young female was discovered among bushes field near a swampy area in Perak. All stages of larvae were observed on the body. The victim was last seen alive in the morning four days prior to the discovery of the body. Further examination revealed the presence of multiple wounds on her head due to blunt object. Animal bite marks were observed on the face, chest and left arm. The internal organs were found missing possibly due to animal activities.

### **RESULTS**

A total of 10 corpses (6 males and 4 females) infested with entomological specimens were reported in this study. The species identified and the post-mortem interval for every cases are shown in Table I. Six cases were found indoor, while

Table I: Specimen collection and estimated PMI for the cases received from Hospital Universiti Kebangsaan Malaysia during 2004

Cases no.	Location	Stage received	Species	Approximate PMI (days)
1	House	Eggs (unhatched)	Unidentified	1
2	House	First instar	Sarcophagid	3
3	House	Third instar	Sarcophagid	4
4	House	Third instar	Ch. megacephala	4
		Second instar	Ch. rufifacies	
		Third instar	Sarcophagid	
5	House	Third instar	Ch. megacephala	4
		Third instar	Ch. rufifacies	
6	Bushes field	Third instar	Ch. rufifacies	3
7	Rubbish dumping site	First instar	Ch. megacephala	2
8	House	Third instar	L. cuprina	4

Table II: Specimen collection and estimated PMI for the cases received from Hospital Ipoh

Cases no.	Location	Stage received	Species	Approximate PMI (days)
9	Drain	Third instar	Ch. megacephala	5
10	Swampy area	Third instar	Ch. megacephala	
			Ch. rufifacies	3

four other cases were found outdoor. Most corpses were in a decomposed state when they were discovered. The ages of the deceased were between 25 to 77 years old.

Blow flies (Family Calliphoridae) of the genus *Chrysomya* were dominant in this study. They were found infesting the corpses in six cases, with two species collected being *Chrysomya megacephala* and *Chrysomya rufifacies*. In addition, these two species were collected from the same body in four cases. Both of them were found in both indoor and outdoor cases, while *Sarcophaga sp.* (Family Sarcophagidae) and *Lucilia cuprina* (Family Calliphoridae) were observed only for the indoor cases. In our study, only one case reported having three species of flies, while another three and five cases were infested by two and one species, respectively (Table I).

Species identification of one specimen (Case No. 1) could not be performed as they could not be reared to larvae and adults. However, the presence of fly eggs on the corpse, without other stages collected, may indicate that the fly might have deposited the eggs not more than one day.

#### DISCUSSION

This study is a report of the forensic entomology specimens analysed by Forensic Entomology Laboratory, Universiti Kebangsaan Malaysia for the year 2004. Dipterans are the first insects to get attracted and colonize human corpses. In Malaysia, Ch. megacephala and Ch. rufifacies of family Calliphoridae have been the primary species of fly found on corpses, where the sites found were quite varied, including rural, residential and aquatic areas<sup>5,11,12,13,14</sup>. Such occurrence was in accordance with the variable death sites in Thailand, which has the same climatic condition<sup>15,16,17</sup>. In this study, Chrysomya larvae were found in corpses at various ecological habitats, comprising indoor area as well as outdoor such as bushes field, rubbish dumping site and aquatic areas. This finding shows the ability of these Chrysomya species to survive in various types of habitats under tropical climate, similar to that reported from Thailand<sup>17</sup>.

Most of the specimens received in this study were the third-instar and this is similar to the findings of previous studies in Malaysia<sup>11,13,14</sup>. This is because third instar larvae are usually found during the most malodorous active decomposition stage of the corpse, which would easily be detected. The investigation of flies in this study also showed the presence of flesh fly sarcophagid only in indoor cases (Case No. 2, 3 & 4), which is similar to the study done by several authors<sup>11,13,14,17</sup>. Unfortunately, the sarcophagid flies collected from these cases could not be identified into species. However, it had been reported that the flesh fly, *Parasarcophaga ruficornis*, has been collected from the indoor death scene in Thailand<sup>17</sup>. Further investigation should be conducted to determine whether this endophilic preference is due to the fly behaviour or is it just a coincidence.

In this study, we also encountered *L. cuprina* in one of the cases involving a corpse inside a house (Case No. 8). *L. cuprina* is also known as Australian sheep blow fly since it causes sheep strikes in Australia. Even though it has not been reported as myiasis causing larval in Malaysia, it is widely distributed throughout Peninsular Malaysia<sup>7,18,19,20</sup>. In this study, *L. cuprina* has been noted to be forensically important fly species in Malaysia, similar to that reported from Thailand<sup>17</sup>.

Understanding the process of corpse decomposition is crucial to establish the PMI in death investigations. This is because arthropod succession analysis requires the association of each species or group to a particular decomposition stage, thus enabling us to estimate the PMI with a more precise method. However, modification of the decomposition process by intrinsic and extrinsic factors can considerably alter the estimate of the time of death. Worth mentioning among the extrinsic factors are temperature, ventilation, humidity as well as access of the body to insects<sup>8,9</sup>. All these factors are also closely related to the location whether the crime was committed indoor or outdoor. Retrospective study done in North America found that the onset of decomposition was faster on human remains found in outdoor environments than those found indoor<sup>10</sup>. Lower temperature, shaded area

and less insects of the necrophagous community inside the building may hinder the decomposition process as well as modifying the entomological fauna succession. Other than that, insects may take several hours to detect and reach a body if it is located in a closed room. In this study, more than half of specimens received were from corpses found indoors. One day was added from the calculated age of the specimens to give more precise estimation while calculating PMI to compensate for the late arrival of flies at a corpse in an enclosed environment.

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#### **REFERENCES**

- Catts, EP, Goff, ML. Forensic entomology in criminal investigations. Ann Rev Entomol 1992; 37: 253-72.
- 2. Greenberg, B. Flies as forensic indicators. J Med Entomol 1991; 28: 565-77.
- Catts, EP, Haskell, NH. Entomology and death: a procedural guide. Joyce's Print Shop Inc., South Carolina, 1990.
- Smith, KGV. A manual of forensic entomology. Cornell University Press. New York, 1986.
- Omar, B, Marwi, MA, Mansar, AH et al. Maggots of Synthesiomyia nudiseta (Wulp)(Diptera: Muscidae) as decomposers of corpses found indoor in Malaysia. Trop Biomed 1994; 11: 145-48.
- Ishijima, H. Revision of third stage larvae of synantrophic flies of Japan (Diptera: Anthomyiidae, Muscidae, Calliphoridae and Sarcophagidae). Japanese J Sanitary Zoology 1967; 18: 47-87.

- Kurahashi, H, Benjaphong, N, Omar, B. Blow flies (Insecta: Diptera: Calliphoridae) of Malaysia. The Raffles Bull Zoology 1997; 5: 1-88.
- Mann, RW, Bass, WM, Meadows, L. Time since death and decomposition
  of the human body: variables and observations in case and experimental
  field studies. J Forensic Sci 1990; 35: 103-11.
- Arnaldos, MI., Garcia, MD., Romera, E. et al. Estimation of postmortem interval in real cases based on experimentally obtained entomological evidence. Forensic Sci Int 2005; 149: 57-65.
- 10. Galloway, A, Birkby, WH, Jones, AM. *et al.* Decay rates of human remains in arid environment. J Forensic Sci 1989; 34: 607-16.
- Nor Afandy, H, Baharudin, O., Mohamed, AM. et al. A review of forensic specimens sent to forensic entomology laboratory Universiti Kebangsaan Malaysia for the year 2001. Trop Biomed 2003; 20(1): 27-31.
- Lee, HL, Krishnasamy, M., Abdullah, AG. et al. Review of forensically important entomological specimens in the period of 1972 – 2002. Trop Biomed 2004; 21: (Suppl) 69-75.
- Ahmad, FMS, Mohamed, AM, John, J. et al. Review of forensic entomology cases from Kuala Lumpur Hospital and Hospital Universiti Kebangsaan Malaysia, 2002. J Trop Med Parasitol 2007; 30: 51-54.
- Ahmad, FMS., Mohamed, AM., John, J. et al. A review of forensic entomology cases at Ipoh Hospital and Hospital Universiti Kebangsaan Malaysia for the year 2003. Ind J Legal Forensic Sci 2008; 1(1): 1-4.
- Sukontason, K, Sukontason, K, Narongchai, P. et al. Chrysomya rufifacies (Macquart) as a forensically-important fly species in Thailand: A case report. J Vect Ecol 2001; 26: 162-64.
- Sukontason, K., Narongchai, P., Sukontason, K. et al. Forensically important fly maggots in a floating corpse: the first case report in Thailand. J Med Assoc Thailand 2005; 88: 1458-61.
- Sukontason, K, Narongchai, P, Kanchai, C. et al. Forensic entomology cases in Thailand: a review of cases from 2000 to 2006. Parasitol Res 2007; 101: 1417-23.
- Singh, I, Kurahashi, H, Kano, R. A preliminary key to the common calliphorid flies of Peninsular Malaysia (Insecta: Diptera). Bull Tokyo Med Dental University 1979; 26: 5-24.
- Sulaiman, S, Sohadi, AR, Jeffery, J. Human helminth parasite burdens on cyclorrhaphan flies (Diptera) trapped at an aboriginal settlement in Malaysia. Bull Entomol Res 1989; 79: 625-29.
- Omar, B, Yasin, M, Marwi, MA. et al. Observation on the distribution of Lucilia cuprina (Diptera: Calliphoridae) in a building of a housing estate at Kelompok Embun Emas, AU3, Kuala Lumpur. Trop Biomed 2003; 20(2): 181-83.