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Insect Succession Associated with a Hanging Pig Carcass Placed in an Oil Palm Plantation in Malaysia

(Penyesaran Serangga Bersekutu dengan Bangkai Khinzir Tergantung di dalam Ladang Kelapa Sawit di Malaysia)

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ABSTRACT

*This study was carried out in an oil palm plantation in Tanjung Sepat, Selangor in September 2007 by using pigs (*Sus scrofa *L.) as a carcass model in a forensic entomological research. A 2.5 month old pig (10 kg) which died naturally was hanged on a palm tree to observe the insect succession and decomposition stages. Observation was made for 16 days; one afternoon visit per day and all climatological data were recorded. On the first day, adult muscids of* Ophyra spinigera *Stein and* Musca domestica *L. were observed, however no blowfly (Calliphoridae) activities were sighted. Fly eggs wer seen on the second day on both sides of the face, inside nostrils and genitourinary area. Adults of* Chrysomya megacephala Fabricius *and* Chrysomya rufifacies *(*Macquart*) congregated on the head and anal areas. Adult flies and maggots (first and second instars) were observed in the mouth and anus of the pig on the third day of hanging. Adult yellow jackets (Vespidae) and spiders (Arachnida) were found preying on some adult flies. Rove beetles (Staphilinidae) were also discovered on the pig carcass. Only a few ants (Formicidae) were sighted. Maggot masses were found in eye orbits, neck, and genital organs on the fourth day of hanging and some maggots were seen falling down to the ground. The dominant maggot species identified on this day was* Ch. megacephala. *On the sixth day, the head, neck, and anus were in the stage of active decay. Maggots of* Ch. rufifacies *were abundant on the seventh day and was the dominant species. On day eight the carcass fell onto the ground.* Chrysomya rufifacies *maggots were found underneath the pig carcass and they started to migrate and pupated under the soil. On the tenth day, third instar* Op. spinigera *maggots were found under the carcass. The rate of carcass decomposition slowed down and became stable from tenth day onwards to the sixteenth day of decomposition. Thereafter, most of the remaining parts of the body remained dried and devoid of any insects.*

Keywords: Chrysomya *spp; forensic entomology; hanging pig carcass; insect succession; oil palm plantation*

ABSTRAK

*Kajian ini dilakukan di dalam sebuah ladang kelapa sawit di Tanjung Sepat, Selangor pada bulan September 2007 dengan menggunakan bangkai khinzir (*Sus scrofa *L.) sebagai model haiwan dalam kajian entomologi forensik. Khinzir berumur 2.5 bulan (10 kg) yang mati semulajadi telah digantung di atas pokok kelapa sawit untuk memerhatikan suksesi serangga dan peringkat pereputan. Pemerhatian dilakukan selama 16 hari dengan satu lawatan setiap hari ke tapak kajian. Semua data klimatologi telah direkodkan. Pada hari pertama, lalat dewasa muscid,* Ophyra spinigera *Stein dan* Musca domestica *L. telah diperhatikan pada bangkai khinzir. Aktiviti lalat langau (Calliphoridae) tidak dapat diperhatikan. Telur lalat mula dijumpai pada hari kedua di kedua-dua belah pipi, lubang hidung dan kawasan genitourinari bangkai khinzir. Lalat dewasa* Chrysomya megacephala *Fabricius dan* Chrysomya rufifacies *(*Macquart*) berkumpul di kepala dan kawasan anus. Pada hari ketiga, lalat dan larvanya (instar pertama dan kedua) diperhatikan berkumpul di dalam rongga mulut dan anus. Tebuan belang kuning (Vespidae) dan labah-labah (Arachnida) diperhatikan sebagai pemangsa kepada lalat dewasa. Kumbang (Staphilinidae) juga dijumpai pada bangkai khinzir. Hanya segelintir semut (Formicidae) merangkak di atas bangkai. Longgokan larva mula kelihatan di orbit mata, leher dan organ genitalia pada hari keempat dan terdapat larva jatuh dari bangkai ke atas tanah. Spesies larva lalat yang dominan pada peringkat ini adalah* Ch. megacephala. *Pada hari keenam, kepala, leher dan organ genital telah memasuki pringkat pereputan aktif.* Larva Ch. rufifacies *pula menjadi larva dominan pada hari ketujuh. Pada hari kelapan, bangkai khinzir telah jatuh ke atas tanah. Larva* Ch. rufifacies *dijumpai di bawah bangkai khinzir dan mula bermigrasi ke bawah tanah untuk proses pembentukan pupa. Pada hari kesepuluh, larva peringkat ketiga* Op. spinigera *juga dijumpai di bawah bangkai. Kadar pereputan bangkai menjadi lambat dan stabil dari hari kesepuluh sehingga hari ke-16. Kebanyakan tisu menjadi kering dan tiada serangga diperhatikan lagi selepas itu.*

Kata kunci: Bangkai khinzir tergantung; Chrysomya *spp.; entomologi forensic; ladang kelapa sawit; suksesi serangga*

Introduction

Insects are usually the first organisms to arrive on a dead body, and they colonize in a predictable sequence. A dead body, whether human or animal, is a large food resource for a great many creatures and supports a large and rapidly changing fauna as it decomposes (Byrd & Castner 2001). Dead body progresses through a recognized sequence of decomposing stages, from fresh to skeletal, over a period of time. During tissue decomposition, a corpse goes through physical, biological and chemical changes (Coe & Curran 1980; Henssge et al. 1995; Van den Oever 1976).

Each stage of human or animal decomposition attracted a certain group of sarcosaprophagous arthropods, primarily insects. Some are attracted directly to the decomposed body, which is used as food for fly progenies, whereas other insect species are attracted by the large aggregation of other insects they would use as food resources. When the sequence of insects colonizing a body is known for a given area, an analysis of the arthropod fauna on a corpse or carcass found on similar areas can be used to determine the time of death. This knowledge can provide accurate and precise estimation of elapsed time since death and is used in many homicide investigations worldwide (Byrd & Castner 2001).

Hanging, resulting from suicide or accident (or more rarely homicide), is a not uncommon form of death. If the body is suspended above the ground, it could present a unique environment for insect colonization. Although extensive research has not been published, some researchers have noted that hanging affects insects colonization of human remains. Hanging altered the insects that colonized by excluding soil-dwelling taxa. Insect species and diversity could also affect the drying pattern of body while it is decomposing. This can reduce the number of insects collected and influences which species that colonized the remains, as well as the interval of colonization of arthropods (Goff & Lord 1994).

Materials and Methods

A 2.5-month-old female pig (*Sus scrofa*), weighing around 10 kg, was obtained from a pig-rearing farm in Tanjung Sepat (2.6 °N, 101.6 °E), Selangor (approximately 85 km from Kuala Lumpur). It died of pneumonia and was immediately hanged on a palm tree (*Elaeis guineensis* Jacquin) by using a rope (Figure 1). The rope was 0.5 m long and the pig carcass was hanged 0.8 m high from the ground. The permission of using oil palm plantation in this study was obtained from the plantation owner. The oil palm plantation was in proper maintenance, and the trees were fertilized with pigs' manure. There were scattering of papaya trees (*Carica papaya*) planted in the oil palm plantation.

The study began in the afternoon (1500 hrs.) of 4 September 2007, which was the first day and lasted until 20 September 2007. Observations at site were carried on for 16 days, with one visit per day in the afternoon.

FIGURE 1. A hanging pig carcass on a palm tree in an oil palm plantation at Tanjung Sepat, Selangor, Malaysia (2.6 °N, 101.6 °E)

Ambient temperature was recorded by using a mercury thermometer and humidity was measured by means of a hygrometer. Body surface and maggot mass temperatures were taken during each visit. Adult flies were caught by using a sweep net. Fly maggots (n=10-20) were collected for rearing and preserved into 70% ethyl alcohol on each day of the study. Collection and preservation procedures were based on the methods of Smith (1986) and Haskell and Catts (1990).

The maggots were placed in 70% alcohol for at least for three days to allow proper absorption of alcohol into tissues rendering them adequately preserved for the next steps of preparation. A transverse excision was made along the eleventh segment of each maggot separating it into two parts. The anterior and posterior parts were then placed in 10% potassium hydroxide (KOH). Immersion in KOH usually lasted for more than 24 hours depending on the sizes of maggots. The maggots were then placed in glacial acetic acid for seven minutes to neutralize the previous KOH solution. Gut and muscle contents of maggots were removed thoroughly with a forcep by having a wooden stick as its handle. The maggots were then placed through ascending concentrations of alcohol solutions from 80%, 90%, 95% and absolute alcohol for 30 minutes each. They were then placed on glass slides and mounted in Canada Balsam. The maggots were then identified to species level by using the taxonomic key prepared by Ishijima (1967).

For live eggs or maggots obtained from the pig carcass, they were cultured in plastic containers (diameter=6cm; height=9cm) containing beef *ad libitum*. On reaching pre-pupal stage, the maggots were transferred to dried containers containing saw dust. The adults that subsequently emerged were pinned and their species identification were made based on Kurahashi et al. (1997). Identified maggots and

adults specimens were kept as a record in the Department of Parasitology and Medical Entomology, Faculty of Medicine, Universiti Kebangsaan Malaysia, Kuala Lumpur.

Results and Discussion

The ambient temperatures in the oil palm plantation for the 16 days period ranged from 26°C to 31°C (mean 28.97°C, standard deviation ±1.57°C), air humidity ranged from 70% to 100% (mean 87.31%, ±8.23%) (Figure 2). Body surface temperatures ranged from 28°C to 34°C (mean 30.75°C, ± 1.91°C) Internal temperature ranged from 29°C to 38°C (mean 33.5°C , ± 3.43°C) and maggot mass temperature varied from 32°C to 40°C (mean 35.14°C, ± 3.39°C) (Figure 3). During the study period, there were four rainy days were recorded (day 1, day 4, day 7 and day 13). The five stages of decomposition observed were fresh, bloated, active decay, advance decay and dry / remain.

Body temperatures of the pig carcass were taken by placing the mercury thermometer onto the body surface. The temperatures recorded were only from the fresh to advance decay stage due to the disappearing of body cavity during the late part of decomposition process. Internal temperatures were taken by putting the thermometer into the oral cavity of the pig while maggot mass temperatures were taken by inserting the thermometer into the center of maggot mass. Each reading of temperature was recorded after placing the thermometer for 3 minutes.

The pig's tongue was protruding and a small amount of blood flowed out of it during the first day the pig was hanged. Within 10 minutes of being hanged, several adults of *Ophyra spinigera* Stein (Diptera: Muscidae) were seen at the mouthpart, nostrils and tongue. One adult *Aedes albopictus* Skuse mosquito was seen attempting to suck blood from the pig's nostril. *Musca domestica* L. came after one hour, and fed on blood at the pig's mouth. No blowflies were seen during the first 3 hours of observation.

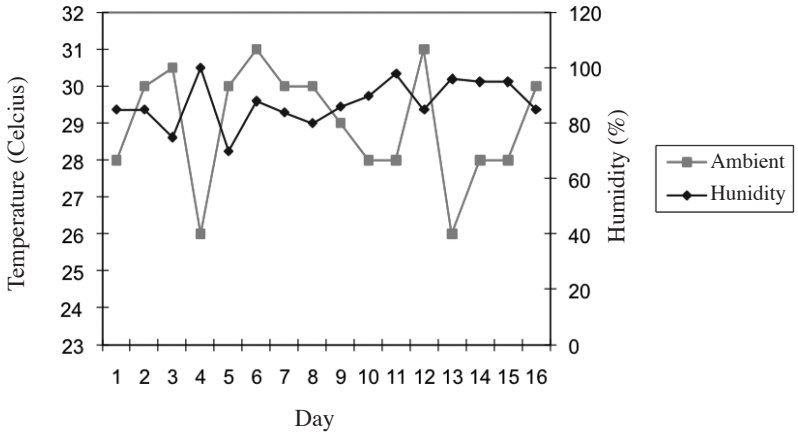

FIGURE 2. Ambient temperatures and air humidity of the study site at an oil palm plantation in Tanjung Sepat, Selangor

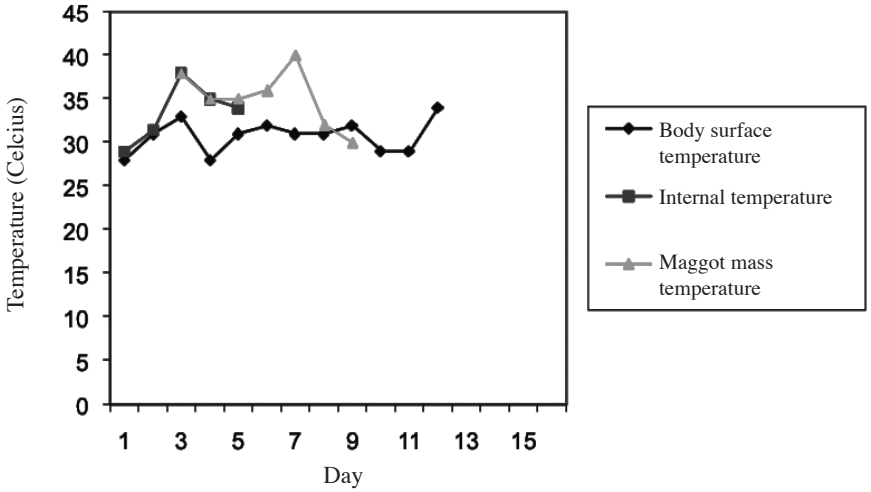

FIGURE 3. Body surface, internal and maggot mass temperatures of the hanged pig carcass

On the second day, approximately 20 blowflies (Figure 4) were attracted to the pig carcass. The species mainly consisted of *Chrysomya megacephala* Fabricius and *Chrysomya rufifacies* (Macquart), with a few *M. domestica*. Masses of yellow eggs belonging to *Ch. megacephala* were oviposited on both sides of the face, inside nostrils, jaws, and genital orifice. Adult *Ch. megacephala* and *M. domestica* congregated mainly at the anal region, being attracted to the fecal materials accumulated at the anus. Several *Ch. megacephla* and *Ch. rufifacies* females were observed ovipositing their eggs in the afternoon on the pig. A nymphalid butterfly (*Euploea mulciber* Cramer) was seen on the carcass.

The bloated pig carcass attracted approximately 300 adult flies on the third day, with *Ch. megacephala* as the dominant species. Most adult flies and maggots (first and second instars) accumulated at the tongue, mouthpart, eyes, anal orifice, and lower extremities of the pig carcass. Yellow jackets (Vespidae) and spiders (*Oxyopes* sp.) were also observed preying on adult calliphorid flies. Rove beetles (Staphilinidae) were also seen at decaying mouthparts of the carcass. Adult female flies of *Ch. rufifacies* were observed ovipositing their eggs on the pig's body. One culicine mosquito (*Verrallina butleri* Theobald, formerly known as *Aedes butleri*) tried to draw blood unsuccessfully from the carcass.

On the fourth day, there was a decrease in adult fly population with approximately 30 adult flies compared to the third day and this could be due to the rain on that day. The adult fly species consisted of *Ch. megacaphala, Ch. rufifacies, Ch. nigripes, M. domestica* and *Op. spinigera*; including some staphlinids beetles. Maggot masses were found mainly at the eye orbits, neck and genitourinary areas. Third instar maggots were seen falling down to the ground. Adult flies of *Ch. megacephala* were still ovipositing their eggs on the pig carcass on this day. The head of carcass was undergoing active decay, and the jawbone was apparent at this juncture. The tongue had detached and some blood was flowing out through the decaying head region. The abdomen and both hind legs were greenish in color but the forelegs were still pinkish white. The pig was in the early active decay stage of decomposition and releasing malodorous smell.

Fly oviposition activity continued on day five. About 250 adult flies and a few ants were observed. One sarcophagid was also seen on the carcass. Some of the maggots on the body were falling off to the ground and there was a newly formed ant's nest nearby on the ground; apparently preying on the fly maggots. The dominant maggot species on the ground was *Ch. megacephala*.

Exponential increase in the adult fly population was seen on day six; where approximately 800 adult flies were counted. *Chrysomya megacephala* was still the dominant species. After completion of bloating stage, the pig was at the active decay stage. Some intestine protruded from the abdominal wall, and the lower jawbone had fallen to the ground. On day seven, many third instar *Ch. rufifacies* (hairy maggots) were seen on the body. Some of them were seen falling down to the ground. Some of these maggots burrowed into the soil and pupated. The hanging skull, ribs, and vertebral bones were already exposed at this juncture. Internal organs such as liver and intestine were at a highly decomposed stage.

On day eight, the skull had detached and the whole pig carcass was on the ground. An estimated 1000 adult flies were hovering and feeding on the carcass and the adult fly population reached its peak on this day. Maggot mass (dominated by *Ch. rufifacies*) could be found on and underneath the carcass, and some were migrating away from the carcass up to a distance of 4 meters.

On day nine, third instar *Ch. rufifacies* and *Op. spinigera* maggots were in abundance underneath the carcass, and many water beetles (Hydrophilidae) were seen creeping under the bones and mummified flesh. Flesh fly (Sarcophagidae) were also sighted around the carcass vicinity. Ants were observed carrying away fly maggots and pupae.

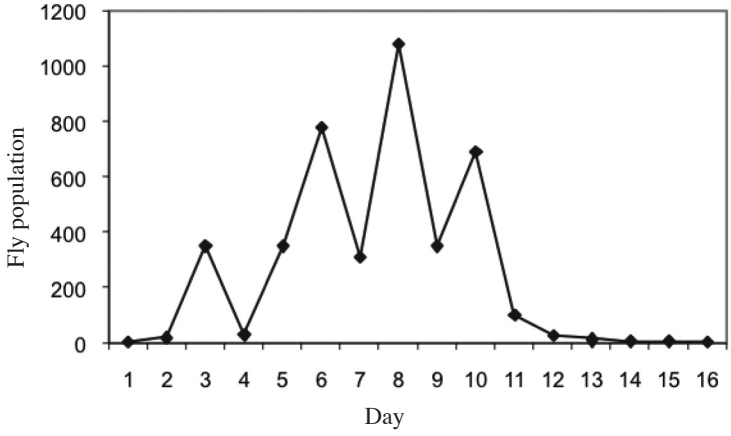

FIGURE 4. Estimation of fly population around the hanged pig carcass

There were about 700 adult flies on day ten. A scarab beetle was sighted near the carcass. Apparent decrease in fly population occurred on day eleven with approximately 100 adult flies seen around the carcass. On day twelve and day thirteen, less than 50 adult flies including *Sarcophaga* sp., *Ch. megacephala, Ch. nigripes, M. domestica, Op. spinigera*, and a histerid beetle and a scarab beetle were observed on the carcass. Many scavenger flies (Sepsidae) were found during this dry stage. Between the fourteenth and sixteenth day (last day of observation), the rate of decomposition became slower and the carcass by now had reached the dry-remains stage. At least 10 adult flies were seen on this last day of observation.

Heo et al. (2007) studied on the pig carcass placed on the ground at the same locality indicated that the dry-remains stage was reached on day 9 and this is shorter than the present study (14 days). The rate of decomposition was slower in the hanging pig compared to the pig that was placed on the ground. This may be due to the absence of soil arthropods (Shalaby et al. 2000). There were a few ants (Formicidae) sighted on the hanging pig carcass which led to lesser predation of maggots, further slowing down the decomposition process. Another factor that slowed down the decomposition could be attributed to falling off the maggots from the hanging carcass. A comparative study of patterns of decomposition in hanging and lying pig carcasses in Hawaii was conducted in 1997 and the authors found that the rate of decomposition in hanging pig was significantly slower than that observed for the control carcass (Shalaby et al. 2000).

The drastic change of the dominant maggot population from *Ch. megacephala* to *Ch. rufifacies* on the day seven may be due to migration of *Ch. megacephala* to other areas or predation by *Ch. rufifacies*. The maggots of *Ch. rufufacies* are both predacious and cannibalistic. If the food supply becomes depleted, the maggots consume, and often totally eliminate, other species from the carcass (Bryd & Castner 2001; Omar et al. 1994).

Conclusion

Hanged pig can affect the rate of decomposition, but not the blowfly development and life cycle. This study has highlighted delay in the decomposition of pig carcass to reach the dry-remain stage. It can be postulated that hanged human body could possibly exhibit similar delaying process as indicated in this study. However, the estimation of Post-Mortem Interval (PMI) is not affected in hanging cases indicating a similar profile of blowfly life cycle development.

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