

## **Role of Carrion Breeding Dipterid Flies in the Disintegration of Chicken Carcasses**

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**Abstract:** Identification of carrion-breeding flies and their life cycle is of significant concern to the field of veterinary, human and forensic sciences. This research was carried out to identify the species of adult flies feeding and breeding on fresh carcasses of chicken; and to determine the period of carcass infestation by fly larvae to complete disintegration of the carcasses. A total of nine adult female crossed jungle fowls were humanely slaughtered and their carcasses were allowed for flies to feed and breed on under natural condition. The experiments were conducted twice; where each experiment, adult flies landed and fed on the carcasses were caught and identified. The species identified included *Chrysomia megalcephala*, *Calliphora* spp, *Lucilia* spp, *Musca domestica* and *Sarcophaga* sp. The time taken by the larval stages to completely disintegrate the carcasses was 83 hours in the first experiment and 77 hours in second experiment. There was a significant differences ( $p < 0.05$ ) between the flies infested group and the control group for time taken to disintegrate the carcasses. However, there was no significant difference ( $p > 0.05$ ) in the number of flies species between the two study replicates. Thus, it can be concluded that carrion-breeding flies play a major role in the disintegration of the chicken carcasses.

**Keywords:** Carrion-breeding flies, disintegration process, chickens carcasses.

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### **I. Introduction**

The order Diptera which are regarded as true flies includes the family of Calliphoridae, Sarcophagidae and Muscidae. In most of the winged flies, the adults ones have two pairs of wings, unlike the order Diptera which have only one pair in which the hind pair of wings have been reduced considerably to become small, club-like organs called halteres or balancers which help the insect to maintain stable flight [1]. Most of the flies in the order Diptera are notorious for causing an important livestock disease known as myiasis causing a great economic impact that results to loss of production or even death of an infected animal. Myiasis is defined as the infestation of the organs or tissue of host animals by the larval stages of dipterous flies, usually known as maggots or grubs. The fly larvae feed directly on the host's necrotic or living tissue [2].

Most of the adult myiasis causing agents such as members of the family Calliphoridae are oviparous, laying large number of eggs directly on to their host or on to vegetation site that are likely to be picked up by passing host [3,4]. On the other hand, members of Sarcophagidae are ovoviviparous which deposit live first stage larvae directly on to their host [5]. Members of the family Muscidae are not generally considered to be of significance in myiasis, but they may be involved as secondary oviparous invaders [6,7]. The egg stage is usually brief, generally lasting about 24 hours, and is followed by three larval stadia where feeding occurs. When feeding is complete, the third-stage larva enters a wandering phase when it leaves the host and locates a suitable site for puparium, usually burrowing into the ground. After pupation, the newly emerged adult breaks out of the puparium and works its way to the surface [1].

Time of death in the early postmortem interval is usually determined by the pathologist, based on medical parameters such as rigor mortis, algor mortis, livor mortis, vitreous humor or potassium content [8]. However, all of these parameters are affected by many other factors such as body size, age, and disease prior to death [8]. In such cases, forensic entomology or the study of the insects associated with a dead body in order to determine time since death is the most reliable and frequently the only method of determining time since death [9,10]. Carrion-breeding flies are attracted to a body or carcasses within minutes. They develop at a predictable rate and colonize the body in a predictable sequence. [11-13]. For example, the development rate of blowflies (Family: Calliphoridae) can be used to determine postmortem interval in a few days or weeks after death [14].

The biology of flies that breed on carrion is becoming more important in the study of insects, especially in forensic sciences. The life cycle of the flies coupled with the degree of disintegration tells the exact time of an animal's death and for how long the body has been there. Thus, this research was carried out to identify the various dipterous flies' species feeding and breeding on fresh chicken carcasses; and to determine the time taken by larvae to completely disintegrate the carcasses. In Malaysia, there is lack of study on the disintegration of animal carcasses. This aim of this study was to determine the rate of disintegration of chicken carcasses based on Malaysia's weather, temperature and humidity.

## II. Materials And Methods

### Scope of the study and sampling location

Nine adult female cross-breed jungle fowl carcasses weighing  $685 \pm 100$ g were used in this study. This study was conducted in a small poultry farm situated in Jenderam Hulu in the state of Selangor, Malaysia (GPS Coordinate: 2.8849396, 101.7231711).

### Experimental design

The nine adult female crossbreed jungle fowl were slaughtered by severing the neck region. The carcasses were then placed in nine individual plastic containers containing nets on 1.5 cm thickness sand. All carcasses were further divided into three subgroups. Group 1 carcasses placed in plastic containers that were tightly covered with cotton cloths to prevent flies from landing, feeding and breeding on the carcasses which served as control group. Group 2 carcasses in plastic containers remain open and undisturbed for flies to land, feed and breed in the carcasses. Group 3 carcasses in plastic containers also remain open, however all adult flies landed in Group 3 carcasses were caught using a net and were collected in a plastic bottle container for identification. The numbers of flies feeding on the Group 3 carcasses were caught and counted daily. The counting of the flies was done on hourly basis from 7am until 7pm every day. All the containers containing chicken carcasses were placed in an open area protected from rain and shine. The experiment was repeated at an interval of 5 days.

### Total disintegration of the carcasses

Total disintegration occurred when all the tissues except the skin and bones were left where the larvae were fully matured and were starting to migrate into the sand. The period taken by the larvae to completely disintegrate on the flesh and organ was recorded for each carcass. By the end of the experiment, carcasses were weighed using a digital weighing scale. Before being weighed, the carcass was cleaned; make sure there were no larvae or sand on the carcasses.

### Identification of flies

For the identification of adult flies, a pictorial key adapted from Division of Medical Entomology, Institute for Medical Research Kuala Lumpur, by Mahadevan S., Cheong W.H and Loong K.P was used.

### Statistical analysis

All the data were analyzed using JMP® 11. NC: SAS Institute Inc. software Version. The data were considered significant at  $p < 0.05$ .

## III. Results

### Total disintegration of the carcass

Flies were observed to land and feed on the fresh carcasses as early as 8 minutes after slaughtered. Flies were attracted to the fresh blood smell and aggregated around the slaughtered site (Figure 1). Flies were observed laying eggs after 8 hours post-slaughter of the crossbred jungle fowl. The mean time taken for total disintegration for the carcasses in Group 2 was 83 hours in the first replicate and 77 hours in the second replicate (Figure 2). There were no significant changes ( $p > 0.05$ ) observed in Group 1 carcasses where disintegration of the carcasses did not take place (Figure 3). The mean weight of the carcasses between the fly exposed and control group were significant difference ( $p < 0.05$ ) indicating the larvae of the flies played a major role in the disintegration of the carcasses (Table 1).

**Table 1: The mean weight of the carcasses between the fly exposed and control group**

	Replicate 1				Replicate 2			
	Control (Group 1)		Test (Group 2)		Control (Group 1)		Test (Group 2)	
	C1	C2	C3	C4	C1	C2	C3	C4
<b>Before Experiment</b>	640g	540g	700g	840g	840g	640g	520g	600g
<b>After Experiment</b>	620g	520g	480g	540g	820g	620g	500g	340g

**Species identification and total number of adult flies**

The species caught and identified were the *Musca domestica* under the family of Muscidae; *Sarcophaga* sp under the family Sarcophagidae; *Calliphora* sp, *Chrysomia megacephala* and *Lucillia* sp under the family Calliphoridae. These species were identified using the key of common flies and based on their metallic coloration. *Musca domestica* or house flies have a bluish to black thorax with four longitudinal lines on the dorsal region. Their whole body is covered with hair-like projections (Figure 4). *Sarcophaga* sp or flesh flies are usually larger in size. They have a grayish to black coloration with darker stripes on the thorax and conspicuous red compound eyes (Figure 5). *Calliphora* sp or also known as blue bottle flies have a metallic blue, blue-green or purple pollinose body (Figure 6). *Chrysomia megacephala* or the big-headed blow flies have a stumpy, box-like body and large red eyes. The flies have a less developed hairy bristle (Figure 7). *Lucillia* sp are the bronze bottle flies. They have a shiny green abdomen with bronze reflection and spiracles are present on the dorsal thorax region (Figure 8). *Musca domestica* were found in highest number throughout the study. This is followed by *Chrysomia* sp, *Calliphora* sp, *Lucillia* sp, and *Sarcophaga* sp (Figure 9). Within 48 hours post slaughter, *Musca domestica* flies were caught in very high numbers compared to the other species. However, the numbers start to decline where members of Calliphoridae family start to increase in numbers after 48 hours. Very little *sarcophaga* sp were caught throughout the experiments. There were no significant difference ( $p > 0.05$ ) in the number of flies species between two replicates of studies (Table 2).

**Table 2: Mean numbers of flies caught according to species**

Fly Species	Replicate 1	Replicate 2
<i>Musca domestica</i>	56 ± 18 <sup>a</sup>	39 ± 18 <sup>a</sup>
<i>Chrysomia megacephala</i>	36 ± 33 <sup>a</sup>	26 ± 33 <sup>a</sup>
<i>Calliphora</i> sp	22 ± 20 <sup>a</sup>	28 ± 20 <sup>a</sup>
<i>Lucillia</i> sp	28 ± 23 <sup>a</sup>	21 ± 23 <sup>a</sup>
<i>Sarcophaga</i> sp	1 ± 1 <sup>a</sup>	1 ± 1 <sup>a</sup>

All values are expressed as mean ± SE; <sup>a, b</sup>, values with superscript within rows are significantly different at  $p < 0.05$

**IV. Discussion**

Carrion flies play a major role in the disintegration process of carcasses [15]. A normal disintegration process of dead animal depend on the size of the carcasses where the larger the carcasses, the longer the time needed to disintegrate [8]. This was observed in our study where carcasses from replicate 1 disintegrate slower at 83 hours compared to replicate 2 carcasses that disintegrate after 77 hours. Carrion flies are attracted to a body within minutes which was observed in this study [15]. They develop at a predictable rate and colonize the body in a predictable sequence [11-13]. The development rate of blowflies can be used to determine postmortem interval in a few days or weeks after death. Carcass breeding by common blowflies constitutes a complex picture in terms of carcass sizes and of seasonal, altitude and habitat variations in the utilizations of the breeding resource by the different blowfly species when acting as saprophages [5].

In this study, the main species of flies landing and feeding on the chicken carcasses were the *Musca domestica*, *Sarcophaga* sp, *Calliphora* sp, *Lucilia* sp, and *Chrysomia megacephala*. These findings were consistent with a report by Kamal, (1958) and Chin et al., (2008) which stated that blow flies such as *Lucilia* sp, *Calliphora* sp, and *Chrysomia* sp were the common flies identified on carcasses [5,15]. In large sized carcasses, species such as *Chrysomia bezziana* may be found although this species are usually the primary cause of myiasis in fresh wounds [16,17]. Weather conditions such as temperature, humidity and light seemed to play a role in attracting the flies to the carcasses [18]. Carrion flies will usually feed and breed in the day when the temperature is high. This indicates that these species were attracted to the strong decomposing carcass odor. Flies are very active insects when the environment is favorable, but during a rainy day with high humidity the number of flies will decrease drastically [18]. In this study, the temperature could not be controlled as the work was done in the field where the weather was always cloudy and humid throughout the experiment. As a result, *Musca domestica* were the first species to land on the carcasses, followed by members of the Calliphoridae. Due to this phenomenon, the use of flies as evidence in forensic science need to consider all the factors that might influence the response and life cycle of the various species of flies in each country [19,20].

V. Figures



**Figure 1:** Flies attracted to slaughtered site (Group 2)



**Figure 2:** Total disintegration of carcass (Group 2)



**Figure 3:** Carcass that did not disintegrate (Group 1)



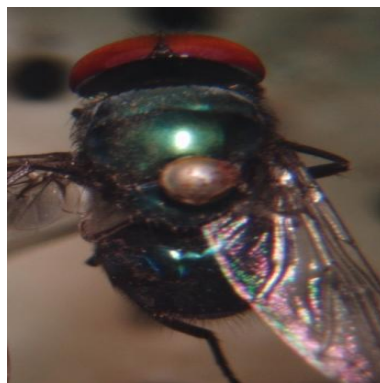
**Figure 4:** *Musca domestica*



**Figure 5:** *Sarcophaga sp*



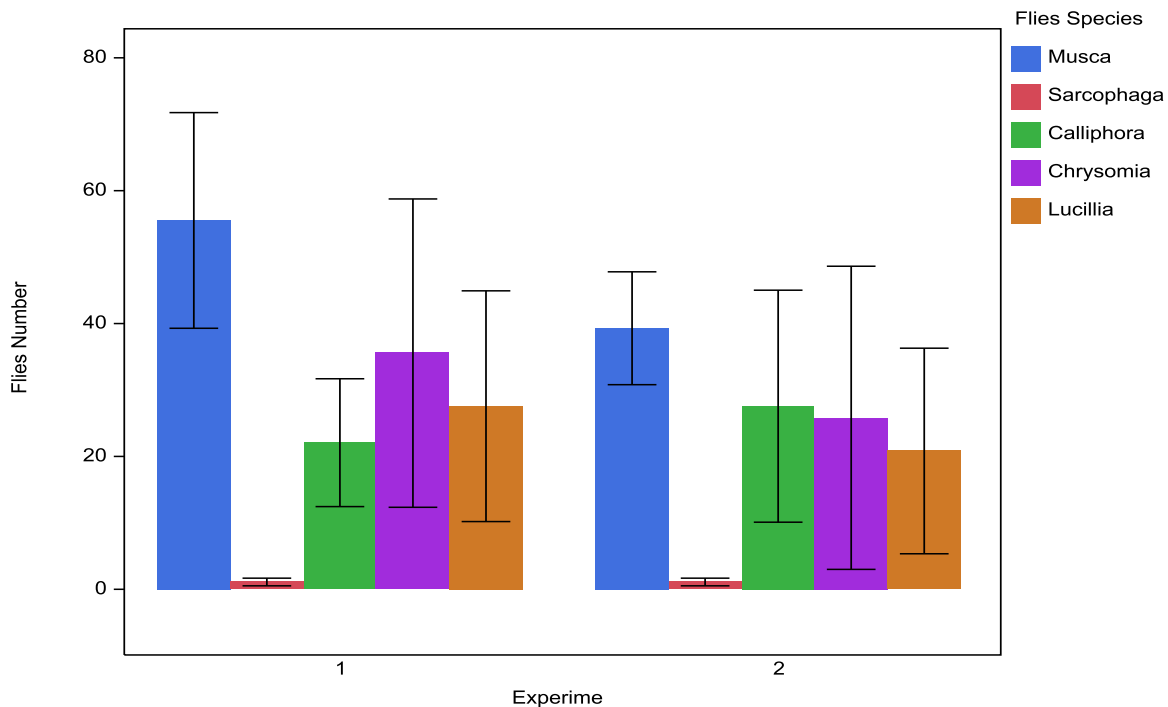
**Figure 6:** *Calliphora sp*



**Figure 7:** *Chrysomia megacephala*



**Figure 8:** *Lucillia sp*



**Figure 9:** Different species of flies caught throughout the experiment.

### VI. Conclusion

In summary, the species of carrion flies breeding in chicken carcasses identified were the *Chrysomia megalacephala*, *Calliphora* sp and *Lucillia* sp. Other species such as *Musca domestica*, were secondary flies that contributed to the carcasses disintegration. The mean total disintegration time taken was 83 hours in Replicate 1 and 77 hours in Replicate 2. This study has conclusively shown that the larvae of carrion breeding flies play a major role in disintegration of chicken carcasses as compared to carcasses restricted from flies feeding.

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