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Survey of forensically-important Calliphoridae in Kingston and St. Andrew, Jamaica, West Indies

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**SURVEY OF FORENSICALLY-IMPORTANT CALLIPHORIDAE
IN KINGSTON AND ST. ANDREW, JAMAICA, WEST INDIES**

A Thesis

Submitted to the Graduate Faculty of the
Louisiana State University and
Agricultural and Mechanical College
in partial fulfillment of the
requirements for the degree of
Master of Arts

in

The Department of Geography and Anthropology

by
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ABSTRACT

The first research to be done in Jamaica on forensically-important species was conducted at the Government of Jamaica Forensic Laboratory in Kingston and St. Andrew in two phases. Phase 1 was conducted from July 5 to July 24, 2007, and phase 2 was conducted from February 11 to February 28, 2008. In the phase 1 study, one local black colored Landrace pig (*Sus scrofa* L.) carcass that weighed 21kg was used as a model for human bodies to determine the rate of decomposition and the pattern of insect succession on decomposing bodies in Jamaica. Ants were the first arthropods to arrive at the carcass, followed by the *Calliphorid*, *C. macellaria* (Fabricius).

Both adults and larvae of *Cochliomyia macellaria* (F.), *Chrysomya rufifaces* (Macquart), and *Chrysomya megacephala* (F.) were collected from the bait 1. Larvae of *Chrysomya rufifaces* (M.) showed aggressive predatory behavior against other 2nd and 3rd instar *Cochliomyia macellaria* (F) and *Chrysomya megacephala* (F.). Ambient temperature had a negative effect on the developmental rate of *C. rufifaces* larvae and the rate of carcass decomposition. The carcass reached the dry stage by day 5 and the decomposition was completed by day 13. Most larvae were burned or desiccated on day 3. Maggot mass sustain temperatures of 14 °C above ambient temperatures over a three-day period during which had a negative impact on the duration and size of *C. rufifaces* larvae.

Phase 2 of the study used two bait types: bait 2 (fresh goat head) and bait 3 (fresh tilapia fish). Larvae collected and reared from bait 2 produced *Chrysomya megacephala* and *Lucilia lucigerens*. Eggs collected from bait 3 were reared which produced only adult *C. megacephala*. The relative distribution of species was different in July, 2007, than in February, 2008.

Chrysomya megacephala (F.) was the only common species collected for both phases. The species *Lucilia lucigerens* (James) is the only indigenous species collected during the phase 2 study in February, 2008. Larvae of *C. megacephala* and *L. lucigerens* emerged as adults in eight plus and sixteen plus days, respectively, at an average ambient temperature of 26 °C and 63 % relative humidity.

CHAPTER 1: INTRODUCTION

Entomology is the scientific study of insects. Medico-legal entomology is the application of entomological knowledge in legal proceedings that uses the examination and identification of arthropods collected on or near a corpse to answer questions about a crime that insects are capable of answering. Flies were once considered to be nothing more than ecological scavengers and a nuisance to man. Flies from the Dipteran families of Calliphoridae, Sarcophagidae and Muscidae are valuable forensic indicators in establishing postmortem interval (PMI). The Calliphorids (blow flies) are especially valuable for establishing postmortem interval because of their profound association with a corpse soon after death. In addition to estimating the minimum postmortem interval at crime scenes, the larvae of these blow flies are able to reveal other important information about crimes such as manner of death, place of death, and the presence of drugs or toxins in a corpse (Carvalho et al. 2004; Catts and Goff 1992; Goff 1991; Introna et al. 1998; Lee 1989; Lord 1990; Smith 1986) . These blow flies are also useful in matters involving sudden death, possible misuse of insects, and traffic accident reconstruction (Leclercq 1969; Hall 1990).

Johnson's (1919) and Dodge's (1965) listings of Dipteran species in Jamaica have shown that there are many species of forensic importance that are indigenous to Jamaica. Although some of these species' distribution has been established, nothing is known about their developmental rates and bionomics that can be of potential use to estimate PMI. In Canada, the United States, and Europe forensic entomology has become a discrete science devoted to the determination of death chronology or postmortem interval in homicide cases. The recently

formed American Board of Forensic Entomology pioneered by Goff and company is testimony to the potential utilization of arthropods' utility in law enforcement investigations (Goff 2001).

Corpses are frequently recovered at various stages of decomposition. In Jamaica, the resources for examining corpses relative to time since death are limited, and as such, will require research of this nature to help in that area. Jamaica is among those countries with a high crime rate and a great number of homicide cases. When a corpse is recovered in other places such as the United States, serious attention is given to the circumstances under which death occurred. Authorities, including the forensic entomologists, law enforcement agents, and forensic anthropologists are frequently summoned to the scene of death. This high profile collaboration of various crime solving techniques has had a tremendous success rate in the establishment of time since death, identification of victims, and, most of all, linking suspects to victims. Currently, there are no entomological resources in Jamaica to determine the PMI when a corpse is recovered. Similarly, there is no forensic anthropologist to carry out osteological examination of remains. For Jamaica to achieve improved crime solving capabilities, emphasis has to be placed on the aforementioned forensic areas. For forensic entomology to be useful in establishing time since death in Jamaica, research is needed in the biology, distribution, and developmental rate of the forensically-important species. Currently, attention is given to only the species, *Cochliomyia macellaria* (Fabricius) and *Cochliomyia hominivorax* (Coquerel), that have medically and veterinary implications as myiasis causing species. Myiasis is the infestation of living cells by the insects. The scientific use of these species as a potential forensic resource instruments in legal investigation has never been executed in Jamaica.

Jamaica needs to develop its own internal database for future reference. Apart from Johnson's (1919) and Dodge's (1965) earlier publications on the distribution of some Dipteran

fauna in Jamaica, no other publications have been done since. The American Board of Forensic Entomology recognizes the need for a holistic research approach to develop a geographical database of insect succession on carrion in a variety of habitats. The underlying reason for such a call is based on the understanding that insects' association with corpses varies from one region to the next (Anderson 2001). Therefore, the primary objective of this study is to gather baseline data for further study of the forensically-important insect fauna in Jamaica, especially those species that have established themselves as the primary indicators of time since death.

CHAPTER 2: LITERATURE REVIEW

2.1 History of Forensic Entomology

A form of forensic entomology was practiced at least as early as the 13th century. In AD 1255, a Chinese death investigator named Sung Tz'u wrote a medico-legal text book – *The Washing Away of Wrongs*. In it, Sung described a case involving the stabbing of an individual near a rice field. One day after the murder an investigator told the workers to lay down all working tools (sickles) on the floor. Blow flies were attracted to invisible traces of blood on a single sickle. When the owner of the sickle was confronted, he broke down and confessed to the murder (Benecke 2001). Most authors cite this book as the first text to deal with forensic entomology.

According to Benecke (2001), Leclercq and Lambert substantiated the attraction of blow flies to blood in 1976; they observed the blow flies species *Calliphora vomitori* on corpses six hours postmortem depositing eggs into blood and not the wounds of the deceased persons. The attraction of fly species to corpses was not only observed by medical and legal experts, but sculptors, painters, and poets also closely observed decomposition of human bodies; they may not have observed the effect of feeding maggots, but acknowledged that their presence on the body accelerated the decomposition rate. Art works such as “Dance of Death” (16th century) and “Skeleton in the Tumba” (16th century) illustrated patterns of decomposition caused by maggots on corpses, especially the rate of skeletonization of the skull as far back as the Middle Ages (Benecke 2001). Several authors have theorized that Francisco Redi (1668) was credited with establishing the link between egg-laying flies and maggots after observing fly infestation of exposed meat. His findings disproved the concept of “spontaneous generation,” which states that maggots arise spontaneously from meat.

Bergeret (1855) is credited with being the first Westerner to use insects as forensic indicators in a case near Paris, France. In March of 1850, the mummified body of an infant was discovered in the bricked-up space behind a fireplace. During the prior three years, four families had rented the apartment. Bergeret investigated and determined that the baby was born to term, but there was no evidence as to the cause of death. He found a large number of empty puparia in several cavities of the body, which he determined were Sarcophagid flesh flies. Bergeret's reconstruction of the insect's natural history led to the exoneration of the three previously-suspected tenants and the conviction of the other. Though he mistakenly assumed that metamorphosis would require a full year, his work was an important contribution to the science of forensic entomology. His work involved the use of the life cycle of the two species on the body that enabled him to estimate the time of death. Bergeret's materials and methods were quite similar to one of the main techniques used today, that is, the successive colonization of arthropod species on decedents (Benecke 2001).

Linnaeus (1767) observed that flies consumed the corpse of a horse as quickly as a lion did. Reaumer (1738) and Macquart (1835) commented on the fecundity of flies. In the mid nineteenth century in Europe, faunal inventories of decaying corpses laid the scientific foundation but did not link flies to murders. Orfila (1848), a pathologist, listed 30 insects and other arthropods that visited a corpse to feed and oviposit. He may have been the first to systematically acknowledge arthropods' succession on a human corpse. Megnin (1894) published a series of medicocriminal entomology articles between 1883 and 1898. His famous article, *La Fauna des Cadavres: Application L'entmoggie a la Medicine Legale* served in a large part to make the medical and legal profession aware that entomological data could prove useful in forensic investigations. Many credit him with establishing the science of forensic entomology. Megnin had more than fifteen years' experience at a morgue in Paris that gave him the expertise

to link the stages of human decomposition to specific insects. Megnin himself described eight stages of decomposition by which he linked succession of arthropods to each stage. He established a biological framework that subsequent experience has proven too rigid. Megnin (1894) relied on a strict schedule of insect arrival and departure from cadavers. He included two stages for buried cadavers, and unknowingly, he established the concept of a corpse as a dynamic, though finite, ecosystem with a faunal succession. Though Megnin's work was groundbreaking, Johnson and Villeneuve (1897) in a summary criticizing Megnin's work, wrote, "The chief danger to be feared by Megnin's imitators is that they might indulge in guesses with no foundation and apply the rules to countries and climates where they were inapplicable" (Benecke 2001).

Johnson and Villanueva's (1897) criticism of Megnin's work is further supported by an illustration provided by Goff (2001). He referred to a case in Florida where the postmortem estimator used insect data gathered in a lush rain forest on the island of Oahu, Hawaii, to analyze postmortem interval on a corpse recovered on a nearly barren Florida sandbar without adequately considering the differences in either environmental or geographical conditions. In addition, a hurricane had moved through the area prior to the discovery of the body. Not considering the latter variables, the time estimate bore no resemblance to the actual postmortem interval. He further pointed out that decomposition stages can be fewer. He described five stages of decomposition during his tenure of studying arthropod succession on carrion in Hawaii. On the contrary, Payne (1965) pointed out that decomposition is a continuous process without discrete stages, but as a matter of convenience, stages could be assigned sequentially.

Later researchers showed that even though blow flies are usually the first to arrive on a corpse (Arnett and Jacques 1981; Borror et al. 1989; Bland and Jacques 1978; Castner et al.

1995; Hall and Doisey 1993; Hogue 1993; James and Harwood 1969; Liu and Greenberg 1989; Peterson 1967; Shewell 1987; White 1970), there are many factors that influence their development and succession on carrion. Such factors are: weather and seasonality, individual species, presence of maggot mass, food type, drugs and other toxins and geographic regions (Wells et al. 2001; Wolda 1988; Goff and Lord 1994). Knowledge of arthropods' biology (especially blow flies) and their geographic distribution can allow accurate estimates relative to the interval of time that a body has been exposed to arthropods' activity and an identification of whether the fauna collected are indigenous or foreign to the site where the body was found. For example, some blow fly species are likely to be found in urban areas, while others are predominant in rural areas (Byrd and Castner 2001). Greenberg (1985) is considered the father of forensic entomology in the West (Goff 2001). He contended that systematic analysis is as reliable as the data upon which it is founded

Currently, forensically-important species are being explored for detecting chemical toxins present in a corpse in a new field called entomotoxicology (Goff and Lord 1994). Recent studies have detailed the detection of toxins and controlled substances in both the insects' gut content and chitinized remnants recovered from badly decomposed victims. Observations of toxins in larvae also have been well documented (Goff and Lord 1994; Sadler et al. 1997; Sohal and Lamb 1977). Gruner et al. (2007) is in the process of refining the current method used to estimate postmortem interval. The argument is that the current method of estimating PMI using insect developmental data as dependent on ambient temperature is in need of a revision to account for the maggot mass temperature dynamic that can skew results. Unlike other insect larvae, blow fly maggots generate heat when they occur together in large masses. Being temperature dependent for growth, an accelerated development could make it harder to know when the flies actually arrive at a corpse. In response to these discrepancies, the researcher has embarked on a large

scale study which involves seventy pigs as models. The objective of the latter research is to generate a thorough knowledge of the effect of maggot mass temperature relative to the time it takes a blow fly species to develop from eggs to adults. According to Gruner, the goal is to refine the current method aimed at the establishment of a universal standard relative to the effect of maggot mass temperature on larval development.

2.2 The Distribution of Forensically-Important Species in Jamaica and the Caribbean

Very little is known about the variety and species distribution of Dipteran fauna in Jamaica. Two of the most recognized lists of Diptera were published by Johnson in 1894 and 1919. The list of 1894 named 115 species. By 1919, a revised publication listed 322 species and varieties, of which 19 were new. According to Johnson (1919), at the time of the latter publication, the knowledge of the general distribution of tropical Dipteran species in Jamaica and America's territories was inconclusive and only those species that were specifically determined to be in Jamaica were recorded. The list noted 83 species in Jamaica, 70 in Cuba, 57 in Puerto Rico, 37 in St. Vincent, 112 in the United States, 74 in Mexico and Central America, and 66 in South America. Dodge's work (1965) discovered three new genera (*Airyipel*, *Cicatricia* and *Farrimayia*) among 41 species he recorded and keyed from Jamaica. James (1971) described two new species from the genus *Lucilia* in rural Jamaica. Systematic identifications have over time reclassified many of these species that had been described up the 1970s; more concise taxonomical classifications are listed in Whitworth (2006).

Since the 1980's, the increased incidences of human and animal infestation by blow flies have attracted the attention of medical and veterinary authorities. As a result, species in the genus *Cochliomyia* are given the most attention for the problems they cause to man and animal, but not for their forensic importance. An article published in Jamaica by Samuel C. Rawlins in

1988 entitled “*Human Myiasis in Jamaica*,” revealed 20 cases of myiasis. Only in three patients was it possible to identify the larvae. Physicians often discard the larvae which are believed to be associated with the typical cases involving the common screw worm, *Cochliomyia hominivora* (Coquerel) (Rawlins and Barnett 1983). Although some patients were treated successfully, there are isolated cases where necrotic infestation caused by the blow flies results in permanent injury to the patient. If an infestation is left untreated or not treated, sometimes the results can be fatal (Rawlins 1988). Some infestations may have been acquired in hospitals frequented by Calliphorids (Rawlins and Barnett 1983). There are signs that researchers of the region are becoming aware of the utility of arthropods in legal matters as seen in a recent work by Velez and Wolf (2008) in Colombia, South America. They provided developmental data for the following forensically-important species: *L. eximia*, *C. macellaria*, *C. albiceps*, and *C. megacephala*. Some of these species are likely to be discovered in Jamaica in the near future. Among the indigenous species of Jamaica, there are other important forensic species that have not been cited for any particular interest in Jamaica and the Caribbean. Some of those species are listed in Table 1 and are expected to be found in Jamaica in the near future because of their wide geographic ranges.

2.3 Description of Climatic Conditions in Jamaica

Jamaica is located in the Caribbean Sea and is part of the Greater Antilles situated geographically at latitude 18° north and longitude 78° west. The yearly average temperature on the coastal lowland is 26°C. An average change of 5°C is recorded between January to February and July to August (the coldest and the warmest months throughout the year, respectively). Blue Mountain is the highest mountain in Jamaica with an estimated fall in temperature of about 16°C per 1000 feet increase in altitude, whereas the annual mean temperature is about 13° C at the peak (7402 feet in elevation). Rainfall averages about 195cm (77.1 inches). The lowest average

day-time temperatures in winter are about 20-25°C and a maximum of 30-34°C in the summer. It starts to warm up in May and gets hot and humid in the summer, particularly in August, September, and October. The traditional rainy season is usually during May/June and October/November. However, in recent times the weather pattern has changed significantly with respect to global weather changes. Because of the likelihood of sudden changes in temperature conditions (Jamaica: A brief overview 2007), neighboring communities are expected to have spatial microclimatic conditions that can provide an interesting geographical area for insect survey.

Table 1. List of species of forensic importance likely to be found in Jamaica and the Caribbean.

Family	Species	Geographic Distribution	Sources
Calliphoridae	<i>Cochliomyia macellaria</i> (Fabricus)	US, West Indies, and Chile,	Hall 1948
	<i>Cochliomyia hominivorax</i> (Cocquerel)	Argentina	
	<i>Cochliomyia minima</i> (Shannon)	F. Guiana, US, and Chile, Argentina, and West Indies	
	<i>Chrysomya rufifacies</i> (Macquart)	Worldwide (intro. New world 1975 from Africa)	
	<i>Phormia regina</i> (meigen) <i>Calliphora vicina</i> (Robineau-Desviody)	Germany, Mexico, Hawaii, and North America	
	<i>Lucilia cluvia</i> (Shannon)]	US, West Indies, and Europe	
	<i>Lucilia proclimataica</i> (N.A.)	Bermuda	Johnson 1919
	<i>Lucilia retroversa</i> (James)	Bahamas	James 1971
	<i>Lucilia lucigerens</i> (James)	Jamaica	James 1971

2.4 Arthropods Considered Useful PMI Indicators in Forensic Science

Estimating time since death is a very important parameter when the death of a human is suspected to occur in ways other than being natural. Under certain conditions of death, causes are not considered normal and the relevant authorities need to investigate the circumstances under which such death has occurred. In cases where a corpse is recovered before exposure to arthropods, the method used to determine time since death is purely pathological evaluation relative to physical and or chemical changes of the corpse. On the other hand, when corpses are recovered longer than a few days after death, entomological evidence may be the only method available for estimating time since death. All insect activities involved with a corpse invariably leave traces of their presence, such as cast larval skins, empty puparia casings, and other evidence of metamorphosis. There are two complementary approaches when insects are used as indicators of postmortem interval. They are:

- a. To recognize the insects' stage of development found on the cadaver. This method requires the presence of the immature larvae to be able to backtrack to the time of oviposition in order to arrive at an accurate time estimate.
- b. This method utilizes the knowledge that insects colonize a cadaver in a predictable chronological manner in which their presence or absence is a good indication of the time since death. This method is very useful when cadavers are exposed to arthropods for periods longer than a week, month, or even years (Anderson 2000; Byrd and Castner 2001; Smith 1986).

Terrestrial and aquatic organisms have the potential to be predictors of forensically-important parameters when such meaningful pieces of information as the time since death, manner of death, and relocation of remains are needed to advance legal matters (Lord et al. 1986). When

the order in which insects colonize a corpse is known for a particular region, an analysis of such order can be used to determine the time between discovery and death. When the procedure is executed correctly, it can be the most accurate and precise method for estimating the postmortem interval (PMI), or time since death. The postmortem history of a recovered corpse can be placed into two categories depending on the context in which the body was recovered. These categories are terrestrial and aquatic (Byrd and Castner 2001).

When bodies are recovered in a terrestrial context, the PMI can be estimated by the presence of the faunal assemblage that had an early exposure to the corpse for oviposition and larval development. When bodies are recovered in an aquatic context, the postmortem submersion interval (PMSI) is determined from certain growth phases involving aquatic plants and animals that are attached to the submerged corpse on which oviposition by insects and larval development were prevented (Byrd and Castner 2001).

2.4.1 Aquatic Insects

Of all known aquatic insects, the Diptera order accounts for about 50 percent of the population, with Midges being the largest fresh water faunal group. According to Byrd and Castner (2001), there are few cases where the use of aquatic insects as indicators of PMI proved parallel to insects used as indicators in terrestrial cases. As reiterated by Byrd and Castner (2001), Sorg et al.(1997) has recommended that research regarding marine cases should be focused on the sessile form of fauna to ensure that the faunal/flora biology is directly associated with the corpse. Similar to terrestrial decomposition, decomposition in aquatic environments is affected by differential environmental conditions that are potentially capable of deferring PMI estimation. Factors such as temperature, water current, aquatic insect clusters, the presence or absence of clothing on a corpse, and whether the body is submerged or afloat are important

variables to consider when dealing with recovery of a corpse in an aquatic situation (Byrd and Castner 2001).

Identifying key aquatic species associated with a corpse is the precept to understanding the significant role aquatic microinvertebrates play in crime scene investigations. Aquatic insects are capable of providing entomological evidence (Merritt and Cummins 1996a; Thorp and Covich 1991). Dr. Richard Merritt at the Department of Entomology at Michigan State University has successfully used aquatic insects to determine PMI in a case of murder in Michigan where a victim was found submerged on the floor of the Muskegon River in western Michigan. He identified the insects as Caddish fly, Chironomid midges, and Black fly pupal cocoon with several larvae. Of the three specimens, the Black fly provided the entomological evidence investigators needed to establish the postmortem submersion interval (PMSI), which was necessary to build a case against the husband, the alleged suspect (Byrd and Castner 2001).

2.4.2 Terrestrial Insects

The most notable terrestrial carrion dependent species that are used as indicators of time since death are flies in the family Calliphoridae (blow flies), Sarcophagidae (flesh flies), and Muscidae, followed by beetles in the family Silphidae and Dermestidae (Smith 1986). Staphylinidae and Histeridae are important predators of carrion fly larvae and are therefore important in terms of the carrion dynamics (Tantawi et al. 1996). Within the spectrum of arthropods used as forensic indicators of PMI, Calliphoridae are the most abundantly-studied carrion species (Catts and Haskell 1990; Megnin 1894; Smith 1986). The blow flies that colonize the corpse early are classified as rural, urban, and omnipresent. Being the first to arrive at a corpse during the fresh stages, Calliphorids set the time clock very soon after death and therefore provide the minimum postmortem interval. For these insects to be useful as indicators of

minimum time elapsed between death and recovery of a body, they must fulfill two principal requirements: they must arrive at, colonize, and feed on the cadaver within a predictable time frame, and reproduce at or near the carcass. Collectively, the blow flies and beetles cover the entire time line required to decompose a corpse completely. Beetles that also feed and breed within the carrion are attracted to the latter stages of decomposition (Catts & Goff 1992; Payne 1965; Smith 1986). Members of the Sarcophagids, Muscids, and Piophilids may also be necrophagous, and, although they are less studied for their contribution to forensic entomological cases, they have the potential to provide useful information regarding the time a corpse has been exposed (Byrd & Castner 2001; Catts and Goff 1992; Payne 1965; Smith 1986).

2.5 The Application of Calliphoridae as Indicators of PMI

Among the Dipteran species, the blow fly maggots are considered the best forensic tools entomologists rely upon for estimating time since death. There are about fifty blow fly species that have been identified as potential indicators of postmortem interval. Maggot size and maturity are the fundamental elements used for indicating when adult blow flies first encounter a corpse (Gruner et al. 2007). Blow fly maggots have the greatest impact during the early stages of decomposition (Putman 1978), and with good records of ambient temperature, the postmortem interval can be calculated to within a few hours, even when death may have occurred two to three weeks previously (Meyer 2007).

Calliphorids start the postmortem clock and are known to travel over long distances to find their food source. Studies in South Africa showed that blow flies are attracted to carrion over great distances in response to perceived odor. In the same vein, marked species of the itinerant genus *Chrysomya* has been known to travel up to 63.5 km in search of a food source (Braak 1981). Studies have shown that due to their early arrival time at a corpse, they are the

major resources used by forensic practitioners to provide an accurate estimation of the time since death. The growth rate of the popular blow fly species *C. megacephala* was studied at different temperatures and it was discovered that they have small variations in growth pattern, which indicate that they are reliable enough to estimate the postmortem intervals with considerable accuracy. Their preference for a fresh corpse makes them a high priority at crime scenes whenever they are encountered (Braack 1981; Byrd and Castner 2001; Goff 2001; Smith 1986).

Medico-legal entomology relies on the examination and identification of arthropods collected on or near a carcass from which their developmental rates are used to answer questions such as: time of death, site of death, and other cases of untimely manner of death (Hall 1990; Leclercq 1969). The life history and actual use of Calliphorids to provide precise and accurate entomological evidence in criminal investigations is well documented by various authors across the world. Some of the earlier contributors to the development and practical framework for the current approaches in forensic entomology are cited in Table 2.

Table 2. Some of the authors that have contributed to forensic entomology in various subject areas

Developmental Data	Carrion Succession (pigs)	Insects and Human Decay
Byrd and Butler 1998	Anderson and VanLaerhoven 1996	Rodriguez and Bass 1983
Goodbrod and Goff 1990	Hewadikaram and Goff 1991	Bass 1985
Greenberg 1991	Tantawi et al. 1996	
Greenberg and Wells 1998	Payne 1965	
Introna et al. 1989		
Nishida 1984		

* References obtain from Byrd and Castner (2001: 265) and Goff (2001: 14-15)

2.6 General Characterization, Biology and Ecology of Calliphoridae

The insect order Diptera (true flies) is composed of over 86,000 known species with 16,000 of those species occurring in North America alone; as a result, the order Diptera is one of the largest of the class Insecta. The Calliphoridae family is the largest family of the Dipteran order, consisting of more than 1000 species worldwide. Calliphorids are also known as blow

flies and are found practically in all niches. The family is comprised of the subsets of blow flies known as the green bottle flies (genus *Lucilia*), blue bottle flies (genus *Calliphora*), and the screwworm flies (genus *Cochliomyia*). Carrion, excrement, and fresh meat make up the bulk of their food resources and species vary from habitat to habits, depending on the season and geographic locations. Their patterns of laying eggs in natural body openings and wounds can be informative of perimortem or antemortem trauma resulting from uneven defleshing of a cadaver (Byrd & Castner 2001; Hall 1948; Smith 1986).

The life cycle of the Dipteran flies is holometabolous (complete) metamorphosis (Smith 1986), which includes the egg stage, three larval stages, pupa, and adult. Of all the stages, the egg stage is the shortest period, lasting 6-48 hours prior to hatching. Depending on the temperature and environmental conditions, adults emerge from the egg stage between 16 to 35 days (Drees and Jackman 1999). Calliphorids are reported to be diurnal, meaning they only feed and lay eggs during the days and rest at night (Byrd and Castner 2001). However, Goff (2001) reportedly observed blow flies during periods of darkness at crime scenes and at decomposition studies in Hawaii. The credibility of his observation was strengthened by Bernard Greenberg's publication in 1990 entitled *Blow Fly Oviposition Behavior* which illustrated that if conditions are favorable, blow flies can become active and lay eggs late at night, though in smaller quantity (Goff 2001).

Blow flies are characterized as having one pair of wings (the second pair reduced to knob-like structures called halteres), large compound eyes, and varying mouth parts depending on the substrate on which they feed. The adults that are associated with human remains have sponging mouthparts for feeding on putrefied substrate while the larvae have chewing mouth parts for piercing tissues. The larvae of blow flies are often called maggots. They range in size

from 8-23mm in length during normal development and are generally cream colored. The larvae possess posterior spiracles which are used for breathing. Larva identification is made possible by the variations of the posterior spiracles among different species of blow flies. The adult flies have three body segments: head, thorax, and abdomen. The wings are attached to the mesothorax and the legs are attached to the ventral side of the abdomen. The adult blows flies range in size from 6-14mm in length and have a pair of large compound eyes which can be used to identify sex within groups of species. Females lay their eggs from an ovipositor positioned at the tip of the abdomen (Byrd and Castner 2001; Smith 1986). A large number of insects are attracted to carrion at preferential stages of decomposition that reflect their dietary preferences. Some of these adult insects and their larvae have differential dietary preferences that result in a complex ecological network of diverse competition for the same food source. Smith (1986) described five ecological categories that were used to classify the carrion communities based on the hierarchical pattern of dietary preferences among species:

1. Necrophagous Species: These species feed on corpses' tissue and are subdivided into:
 - a. Sarcosaprophages: These species feed on decomposing flesh and include Calliphoridae, Sarcophagidae, Muscidae, and Dermestidae.
 - b. Coprophages: Species that are attracted to the gut contents of herbivorous mammals e.g. Scarabaeidae and Muscidae.
 - c. Dermatophages: Species feeding on dried skin, hair, ligament and bones which include Dermestidae and Tineidae.

2. Necrophagous - Predaceous Species: Those that feed on both the corpse and its inhabitants which include the ants (Formicidae), silphid beetles, clerid beetles, and some blow fly larvae.
3. Predaceous Species: Those that feed only on the carrion faunal community which include the larvae of *C. rufifaces* (Diptera: Calliphoridae), Histeridae, and Staphylinidae.
4. Parasitic Species: Insects that parasitize fly larvae and pupae which predominantly are seen in the Hymenoptera order (wasps).
5. Adventurous or Incidental Species: Arthropods that use carrion as a concentrated resource extension of their normal habitat e.g. spiders, centipedes, millipedes, mites etc.

These species typically occur in succession and respond to progressive changes of carcass decomposition stages. They are the most important species in providing useful forensic information regarding time of various events associated with human remains. Apart from those Calliphorids that feed and reproduce on the corpse, some are parasitic and predacious to other member species. For example; *Chrysoma rufifaces* larvae not only feed on carrion, but are also predaceous during the larval stages (Goodbrod and Goff 1990; Smith 1986)

2.7 Characterization, Biology, and Ecology of Species Found in Jamaica

Within the last twenty years, four species of Old World blow flies in the genus *Chrysomya* (Robineau-Desvoidy) have been recognized the New World. The early arrival of the *Chrysomya rufifaces* (Macquart) and *Chrysomya megacephala* (Fabricius) in South America has been documented (Baumgartner and Greenberg 1984) as well as their establishment on the United States mainland (James 1947; Zumpt 1965). The synanthropic nature, along with an estimated dispersal rate of 3.2kg/day, typifies the rapid dispersal rate of *C. rufifaces* during the

early introductory period in South America. During the 1970s and early 1980s, the species *C. rufifaces* was reared in Costa Rico (Jiron 1979) and was later observed in Mexico and Texas. *Chrysomya rufifaces* is believed to have been introduced separately by reason of its oriental and Australian distribution (Baumgartner and Greenberg 1984).

The two most prominent Calliphoridae species across the globe are *Chrysomya rufifaces* (M.), the Hairy maggot blow fly, and *Chrysomya megacephala* (F.), whose origins are from the Old World, mainly from the mainland of Australia and New Guinea in Africa (Barreto et al. 2002; Goff et al. 1986; Goff and Flynn 1991; Gunatilake and Lord 1990; Smith 1986; Sukontason et al. 2008). Both species are indigenous to the Australian corridor of the Old World and have been well established and distributed in various regions of the New World. *Chrysomya megacephala* was first discovered in Brazil in 1977 and later in the United States. *Chrysomya rufifaces*, on the other hand, was first observed in the U.S. in 1981. The life history of *C. rufifaces* has been in the forensic limelight as far back as 1922 when the species was studied as *C. albiceps* (Patton 1922). The frequencies in which they are now discovered on a corpse in the U.S. have garnered serious attention of forensic entomologists. The greatest concerns are due to differential habitats and food preferences, dispersal rate, and reproductive prolificacy. *Chrysomya megacephala* are more synanthropic than *C. rufifaces*; as a result, they are more abundant around places inhabited by humans. The adults of *C. megacephala* will readily enter buildings searching for oviposition sites as opposed to *C. rufifaces* that are rarely found in buildings. They are the most abundant species found on human remains recovered from outdoors in Florida (Byrd and Butler 1996; Kurahasi 1982). The larvae of *C. rufifaces* only develop on carrion as opposed to the larvae of *C. megacephala* that show variation in habitat preferences. Laboratory studies of *C. megacephala* showed that this species is a scavenger by nature and they are able to develop on a variety of food sources: catfish, frogs, lizards, pigeons,

and toads (Wells and Kurahashi 1984; Gabre et al. 2005). Neither of them is attracted to dried remains. The adults of *C. megacephala* are very persistent in whatever resources they colonize. Studies show that they are the first to arrive at the carcass in the mornings and the last to leave.

Due to the predacious and cannibalistic nature of *C. rufifaces* and their aggressive feeding habits on carrion and other larvae, forensic entomologists argue that the species will be encountered with greater frequency over time. Both species are tropical feeders, but *C. rufifaces* shows the tendency to distribute itself over a wider range during the hot summer season, even as far north in the U.S. as the Canadian borders (Byrd and Castner 2001). Wijesundara (1957) studied the biology of *C. megacephala* in the tropics of Sri Lanka. He found that the adults deposited roughly 220-320 eggs in masses on the under surfaces of carcasses and feces, and developmental time from egg to adult took eight and a half days. He noted that the egg stages lasted for 9-10 hours, and the larval stages lasted for 94-95 hours. He also noted that if the temperature at rearing remains constant at 30 °C, the life cycle could be completed in seven and a quarter days due the reduction of the duration in the egg and larval stages.

Cochliomya macellaria is commonly known as the secondary screw worm due to its secondary role as an agent of myiasis in animals, behind *Cochliomya hominivorax*. The species frequent tropical areas and are very abundant throughout the southern United States to as far north as the Canadian border. They are not usually active in the northern U.S. during the winter but are present during the summer. They have a preference for warm, humid areas and are very abundant during the rainy season. The species is known to frequent carrion in both sunny and shaded habitat and is rarely recovered from indoor habitats. The adults have a bright greenish-blue appearance with three longitudinal stripes that are abbreviated on the thorax. Although the frequency of the species has been reduced due the introduction of *Chrysomya rufifaces*, they remain the predominant species found outdoors in Florida (Byrd and Butler 1996).

Lucilia cuprina formerly in the genus *Phaenicia* is commonly known as the Australian Sheep Maggot or Bronze Bottle Fly. The species has roots in Australia, Africa, and the Americas, and its presence is now discovered in Jamaica. The fly frequents the southern parts of the U.S. The fly is characterized as being small and has the metallic yellow -green color that is characteristic to the Calliphorids, which reflects a dull coppery appearance. The adults are most likely to be found on excrement and decaying vegetable matter, as well as carrion. The species readily enters dwelling places and can be found all year round in tropical areas. They are also potential myiasis causing species (Arnett and Jaques 1981; Bland and Jaques 1978; Borror and White 1970; Borror et al. 1989; Castner et al. 1995; Greenberg 1971; Hall 1948; Hall and Townsend 1977; Hogue 1993; James and Harwood 1969; Peterson 1979; Smit 1931; Smith 1986).

Lucilia lucigerens (White), formerly of the genus *Phaenicia* is indigenous to Jamaica and is a member of the Calliphoridae. The distribution of the species was first described thirty eight years ago in various parts of eastern Jamaica over a wide range of elevations, namely, St. Thomas, Portland, Kingston and St. Andrew, and St. Catherine. The highest elevation was recorded at 4600 feet in Mt Horeb, NE Portland. Based on the areas where they were collected, the species appeared to be migratory by spending the winter period in the urban area, and spending the summer in forested humid areas. The number collected showed that the species is an emerging species with a small population. Although the biology and ecology of the species was not explored, some basic morphological description has been given. The species is believed to be a descendant of the ancestral stock of *P. retroversa* in the United States. They are distinguished mainly for the differences in the pollen distribution along the head, thorax, and abdomen, and the color of the legs. The species has an average body size, with a male range of 6.0 mm to 7 mm and a female range of 6.0 mm to 8.0 mm (James 1971).

2.8 Factors Affecting the Usefulness of Calliphoridae in Establishing PMI

There are several environmental variables that can limit the accuracy of time since death when an insect collected from a corpse is used to make such an inference. Many of the factors related to decomposition also affect the faunal assemblage, which are often neglected. These inter-related variables, such as faunal activities, temperature, humidity, rainfall, and condition of food source etc., need to be evaluated carefully for the outcome of a successful investigation (Varatharajam and Sen 2000).

2.8.1 Temperature and Humidity

Temperature, season, and type of habitats define a geographical region and thus influence the insect community within that area (Dillon and Anderson 1997). The concept that insect developmental time is temperature dependent was arguably initiated by Reaumur (1735), a French scientist. The understanding and application of the concept as a useful method of predicting and describing insects' development was thought to be emphasized in the 1950's (Byrd and Castner 2001). Temperature and humidity heavily influence insect activities, such as the rate of oviposition and maturity (Anderson & Cervenka 2001; Gillot 1995; Smith 1986). The overall rate of decomposition due to insect activities accelerates or decelerates depending on temperature and humidity. Carrion insects' populations tend to decrease on a cloudy day and are more abundant on warmer days (Goff 2001). The difference between maggot mass temperature and ambient temperature is greatest during cold weather (Deonier 1940); therefore, the temperature and size range of the larvae should be recorded before disturbing the corpse (Byrd and Castner 2001).

Temperature has a direct impact on insect metabolic and developmental rate (Andrewartha and Birch 1954; Chapman 1982). Development of insects is confined within a

certain temperature range; temperatures too high or too low below the threshold can prove fatal. Corpse temperature is affected by both air temperature and exposure to sunlight (Byrd and Castner 2001; Wells et al. 2001). Some species enter larval or pupal diapause (arrested development in response to seasonal cues) and this affects the time spent in a particular life stage even if the temperature is optimum (Denlinger and Zdzarek 1994; Tanker et al. 1987). Closely related species exhibit different diapause behavior (Ash and Greenberg 1975), thus requiring the need for comprehensive knowledge of insect life history (Wolda 1988).

A study by Povolny and Rozsypal (1968) showed that *Lucilia sericata* (Meigen) grew more slowly in a vegetable medium when compared to larvae that feed on meat. Goff and Flynn (1991) pointed out two scenarios where a forensic entomologist may be asked to age larvae from a food source other than a corpse; this would be difficult if the observed developmental rates on the meat medium are not appropriate for such analysis relative to temperature and rate of decomposition (Byrd and Castner 2001).

2.8.2 Seasonal Variation

Little information is available on possible regional variation in forensic entomological phenomena. All environments on earth undergo natural changes over time and space, whether the habitat is terrestrial or aquatic. The changes can be biotic or abiotic. Environmental changes have three general characteristics that are relevant to the organisms: magnitude, predictability, and duration. Of the three environmental changes, magnitude is the most difficult to classify, in terms of the impact on insects. However, there are studies that relate the magnitude of environmental changes to the degree at which insects adapt to their environment. Predictability and duration of environmental changes are more easily interpreted in terms of their relationship to insects. For example, some changes recur on a regular or cyclic basis that is able to provide

environmental signals, which allow insects to inherently predict such environmental changes (Tauber 1986). Although developmental data of some common species have been compiled in laboratories in varying localities, differences in methods used and presentation of data make it difficult to interpret and compare results (Byrd and Castner 2001). In a study designed by Cyr (1993) to test the possibility of regional variation, he found no statistically significant differences in the duration of developmental stages of *Phormia regina* (Meigen) from the states of Washington, Indiana, Texas, and Louisiana. Contrastingly, independent studies suggest that carrion fly behavior can indeed vary according to region. For example, throughout most of its distribution, *Chrysomya rufifacies* (Macquart.) invades dead and live vertebrates only after other larvae are present (Bohart and Gressitt 1951; Wells and Greenberg 1994; Zumpt 1965), but in some sites it has been observed laying eggs on fresh carrion (O'Flynn and Moorehouse 1979) and on uninfested new-born calves (Shishido and Hardy 1969). Different studies have different views on the possible reasons for such regional differences, even among the same species. One study showed that *Cochliomyia macellaria* (Fabricus) typically survives the winter only as far north as southern Texas and Florida; yet, the species spreads throughout the entire contiguous U.S. and into Canada each year (Hall 1948). This pattern of movement should promote extensive gene flow between locations. The limited work in molecular genetics suggests that widespread carrion feeding species encounter no barriers to gene flow over distances of thousands of kilometers (Stevens and Wall 1997; Taylor et al. 1996).

The species composition in different parts of the world, and even among states and provinces, can be distinctive to that region or overlap across a vast area. For example, in the Old World, species of the genera *Lucilia* and *Chrysomya* are considered the most important carrion species (Smith 1986). In the New World, *Phormia regina* (Meigen), *C. macellaria* and species of *Lucilia* and *Calliphora* are important carrion breeders (Hall 1948). In British Columbia, *L.*

illustris was the first species to arrive on pig carrion (Anderson and Vanlaerhoven 1996). In contrast, a similar study done in Manitoba showed that *P. regina* are the first to arrive on pig carrion within minutes of exposure (Gill 2005). In Northern France, *Calliphora vicina* (Robineau-Desvoidy) and *Calliphora vomitoria* (Linnaeus) were the first species to lay eggs on rabbit carrion. These species did not arrive until three days after the carrion was exposed. Interestingly, in the following year, *C. vicina* and *C. vomitoria* colonized the carcass within the first day (Bourel et al. 1999). By contrast, a study done by Tantawi et al. (1996) on the succession of insects in Egypt found *C. vicina* and *Lucilia sericata* (Meigen) to be the first colonizers on rabbit carrion. There are certain generalizations that can be made about the assemblage of species that occurs in certain geographical regions. However, studies are necessary within each region since differences in community composition and assumptions could lead to erroneous conclusions pertaining to the estimation of time since death (Catts and Goff 1992; Lee and Marzuki 1993; Kuusela and Hanski 1982).

The introduction and dispersal of four Old World species of the genera *Chysomya* into the southern areas of the New World have affected the insect faunal community of these areas (Laurence 1981). Their presence has impacted the estimates of time and location of death; discrepancies between the species composition on a body and the composition of the insect species in a particular geographic area can provide evidence that a body has been moved (Hall 1990). Constant surveys of blow fly populations must be conducted for regions that could experience introduction of new species in order to avoid misinterpretation of data (Gill 2005). An interesting new technique suggested by Bryne et al. (1995) is the analysis of cuticular hydrocarbon in blow flies, citing that this technique could be used to provide forensic evidence in corpse removal for the future. The cuticular hydrocarbons of three geographically different

populations of *P. regina* were distinguished. The shortcoming of this technique is reflected where death and relocation sites are separated by a short distance.

In some localities with relatively nonseasonal climates, some species behave as if their environment were highly seasonal (Wolda 1988). For example, in Fontana, a virtually nonseasonal site in Panama, two of three species of dobsonflies (*Chloronia sp.* and *Platyneuromus sp.*), (Neuroptera: Corydalidae), are among the most seasonal insect species occurring as adults only during a two months' period each year. The third species, *Corydalus armatus* (Hagen), has a distinctly bimodal distribution over most of the year. Multivoltinism is probably prevalent in tropical species. Sometimes this is clearly shown in the data by two or three well defined seasonal peaks. In longer lived, fast growing species, successive generations overlap. There may be four, five or even six to eight generations per year. As with other organisms, the diversity of tropical insects' seasonality patterns at any one site is usually high. Comparing sites on the basis of "average seasonality" does not seem to be useful. A frequency distribution of the different patterns is much more informative. For instance, a study conducted on Barro, Colorado Island, Panama, provided 12 years of light trap data on Homoptera, yielding the following results: 39% of 426 species occurred as active adults throughout the years with only 7.5 % classified as nonseasonal. The duration of the season and timing of the seasonal peak also vary significantly among species. This is a sharp contrast with the seasonality of temperate insects for which activity and its peak are usually restricted to the warm season (Wolda 1983; Wolda 1988).

A study on necrophagous Diptera was conducted in a seasonal tropical mixed forest in Hong Kong (latitude 22.3° N) from February, 1985, to May, 1986, using carrion baited traps on a slope of 100-200 meters. Four carrion-baited fly traps were placed 500 meters apart as the

method for trapping flies; each was constructed from PVC jars, similar in design to those of Williams (1984). This was a modification to the West Australian blow fly traps (Vogt and Havenstein 1974). Of the six families of flies that were caught, only the Calliphorids, Sarcophagids, and Muscidae were of importance to the study. The three families totaled 14,669 flies and 21 species in the overall sample. Calliphorids accounted for 79.44% of the total combined catch which showed three distinct peaks: April, July, and early November. The April peak was repeated in March and April, 1986. The mean number of species trapped per week peaked in June and July, with a decline in September and a second peak in early November. The researchers found that temperature was the main factor affecting seasonality of the necrophagous species in Hong Kong. The climatic condition under which the research was done reported a total rainfall of 246.51 cm (98.04 inches), which fell mainly between April and September, 1985. Mean air temperature of 13.5° C (56.3° F) and weekly relative humidity ranged between 60.3% in January, 1985, and 91.4% in March of the following year (So and Dudgeon 1990; So 1987).

2.9 An Overview of the Variability that Influences Insect Activity in the Tropics

Seasonal variations depicting the abundance of tropical insects have fascinated tropical entomologists based on data reported from studies conducted in Brazil. The tendency observed in temperate areas for the average season to increase in length with decreasing latitude is also observed in tropical areas (M^cElravy and Wolda 1982). In tropical areas where temperature changes are minimal, seasons are assessed by rainfall and humidity (Wolda 1988). Because of the seasonal distribution of many insects, seasons can be classified as occurring in the “early rainy season” or “early dry season.” This does not mean that rain, dry weather, or elevated humidity acts as a cue for their activity. Baumgartner and Greenberg (1985) found differences in species of blow flies during these two seasons in Peru. Prevalence of blow flies was greatest in the wet season and least in the dry season. Other factors, such as food and the presence of

parasites and pathogens, play a significant role in insects' activities. The difference in prevalence was related to altitude (Wolda 1978).

A large scale research conducted in various tropical areas across the world (Old World to New World) on various insect groups demonstrated that there are variations in the developmental patterns. The same developmental and behavioral patterns found in the Panama Homoptera also occur elsewhere. Tropical areas of Jamaica, Trinidad, Brazil, Costa Rico and Colombia were included in the prior research which ran for a 12- year period gathering light trap data. Wolda (1988) found that even in localities with relatively non-seasonal climates, some species showed variation within their particular niche. However, the only notable carrion feeding species covered in the study were Sarcophagids and Coleoptera from Panama, Trinidad, and Jamaica in the West Indies. Although this research did not provide much information on the blow fly species, the data gave a good account of the high variability in faunal seasonality within the West Indies (Wolda 1978; Wolda 1983; Ramous and Wolda 1985). Because of the relatively limited information on forensically-important Dipteran fauna in Jamaica, there is an urgent need for a collaborative effort in providing the pertinent forensic information on the faunal population from different geographic regions. Currently, the American Board of Forensic Entomology advocates for more research to be undertaken in different geographic localities on Calliphoridae and Sarcophagidae (Anderson 2001).

CHAPTER 3: METHODS AND MATERIALS

3.1 Study Site

The entomological/ anthropological study was conducted in two phases from July 6 to July 24, 2007, and February 11 to February 15, 2008, on the eastern side of the Government Forensic Laboratory compound located in St. Andrew, Kingston 6, Jamaica, West Indies. The site is located on approximately two acres of land which is part of two hundred acres of land acquired by the Jamaican Government in 1881 to establish a number of research stations for exotic plants and animals. It is nestled between the Hope Botanical Gardens on the eastside, the Ministry of Agriculture animal station to the west side and south side, and a housing development on the north side. The site is at the tip of the Liguanea plains on which Kingston and St. Andrew were developed and is about five miles west of the Blue Mountain's geographic zone. Kingston and St. Andrew were merged in 1923 to become the Kingston and St. Andrew corporate area and is now referred to as the Kingston and St. Andrew Corporation, which represents Kingston and St. Andrew as one area. The Hope River, which is about one mile to the north of the site, keeps the area cool during the nights, but has little impact on temperature during the days. Relative humidity is usually high as it is with daily ambient temperature. Figure 1 shows the various sites at which the research was conducted in Kingston and St. Andrew. Figure 2 shows a portion of the topography of the habitat where the bait 1carcass was placed. As observed in Figure 2, the area has a wide distribution of xerophytic plants such as the Guango (*Samanea saman*), Guava (*Psidium guava*), Mango (*Magnifera indica*), Banana (*Musa sapientus*), Pangola grass (*Digitaria eriantha*), and weed shrubs scattered about 20-30 feet from the scarecrow used to repel predators. The ground is covered with Bermuda grass (*Cynodon dactylon*). None of the trees provided shade for the carcass, as the bigger trees are distributed far away from the carcass. The carcass was enclosed by ten-foot metal fences. The geographic

reference coordinates for the site are 17° 59' N latitude and 76° 48' W longitude. The site is roughly 143 meters above sea level and its temperature is directly influenced by the coastal wind pattern of the Caribbean Sea. The two major winds that influence weather patterns are the southeast trade winds and the northeast trade winds. Daily photo periods were thirteen hours and forty-eight minutes. Daily ambient temperatures for the area are recorded by the national meteorological office at the Norman Manley International Airport, which is approximately 8.08Km from the Government Forensic Laboratory. Three bait types were used throughout the study: pig carcass (bait 1), goat head (bait 2), and fish (bait 3). The study conducted with bait 1 was designed for the study of the decomposition process and arthropod succession from July 5 to July 24, 2007. Bait 2 was designed to check if the distributions of species are the same throughout the year considering that type of bait has no effect, and, secondly, to rear at least one larval species collected. Bait 3 was used to collect only eggs for the rearing of that species for its forensic application.

3.1.1 Phase 1 Study Using Bait 1 (Pig Carcass)

On July 5, 2007, at 4:05 PM, the carcass (bait 1) of one black-colored Landrace domestic pig (*Sus scrofa L.*) weighing 23Kg (51pounds) was purchased from the commercial meat market at Cross Roads, Kingston 5, which is approximately three miles from the study site. Blood was oozing from a wound to the throat which was the result of the method used to slaughter the animals at the market's abattoir. After receiving the carcass, the body temperature was taken by inserting a temperature probe into the mouth prior to being placed into a large polythene plastic bag. The bag was tied loosely to allow air to enter and to prevent arthropod entry. The carcass was transported in the plastic bag for about 25 minutes to the study site. The carcass was removed from the plastic bag and placed directly on the grass covered ground surface at 4:30 PM. Body temperature and heart girth measurement were taken during carcass placement.



Figure1. Map showing study sites: Phase 1, Phase 1 supplementary, Phase 2, and rearing sites.

Legend:

P1(phases1 study on bait1)

P2 (phase2 on bait 2 and bait 3)

R1 (rearing site)

X (Sites survey in Kingston and St. Andrew)



Figure 2. The topographical view of study area for bait 1, 2 and 3 with scare crow used to repel predators shown in foreground

The carcass was placed in an open field and exposed to direct sunlight for the entire duration of the study.

The carcass was visited in the mornings and afternoons for the first six days of the experiment and once per day for 14 days as seen fit. Photographs were taken at each visit to record the degree of physical changes associated with the decomposition processes. Stages of decomposition were assigned according to the assessment made of the physical changes observed during decomposition. Two methods were adapted to estimate biomass loss during the decomposition of the carcass. The method used at the 2000 Ak-Sar-Ben pig carcass contest evaluates the biomass composition (dressing weight) of a carcass to be 74 % of total warm carcass weight and 25 % viscera (gut content and blood loss at slaughter) at an average of 20% fat with an average backfat thickness of 0.5 inches (Nold 2008). Assessment of the percentage of biomass loss at the dry stage was based on the experimental data that the water content of a pig's carcass is 47 ± 4.8 % (Houseman et al. 1973). The second method is a useful method for the

estimation of a pig weight in the absence of a scale. It uses the circumference of the heart girth to approximate weight changes. This method is very efficient for estimating body weight and has a correlation factor of $r^2 = 0.98$ and a 95 % confidence interval of ± 10 pounds (Groesbeck 2003). This method was the principal resource adapted for this study to estimate daily biomass loss during the first five days of insect activities on the carcass. The standard equation used in the estimations is as follows;

$$10.1709 * \text{heart girth (inches)} - 205.7492 = \text{pig's weight.}$$

With this method, a one pound decrease in heart girth is equivalent to ten pounds reduction in weight loss or biomass loss. The initial heart girth measurement of the pig's carcass was 25 inches at interment. A second heart girth measurement was recorded early in the decay process which showed a reduction in heart girth circumference of two inches. After the frame of the carcass collapsed, biomass loss was estimated assuming that heart girth is reduced by an inch per day until the carcass frame is collapsed. The combination of both methods was used for estimating biomass loss (weight loss). The original heart girth measured 25 inches on day 1 which gave a carcass weight of 48.325 pounds. The three pounds reduction in body weight can be attributed to blood loss during the slaughtering process. By day 3 after the bloat stage and the beginning of early active decomposition, heart girth had reduced to 23 inches. Extrapolating from previous experiments, a decrease by one inch is equivalent to a ten-pound drop in body weight, which is used for the equivalent biomass or weight loss. Biomass reduction began on day 3 when insect activity on the carcass started. The calculations used to estimate biomass loss are as follows;

$$\text{Day 3: } 10.1709 * 23 - 205.74 = 28 \text{ pounds } [48.3 - 28 = 20/48.3 * 100] (\geq 41.66 \% \text{ loss of biomass})$$

Day 4: $10.107 * 22 - 205.74 = 18$ pounds [$48.3 - 18 = 30 / 48.3 * 100$] ($\geq 62\%$ loss of biomass)

Day 5: $10.1709 * 21 - 205.74 = 8$ pounds [$48.3 - 8 = 38.3 / 48.3 * 100$] ($\geq 79\%$ biomass loss)

After day 5, using the above method would result in negative values, which was an indication that the carcass was approaching the remains stages of decomposition. Most of the larvae had migrated away from the carcass by day 6 and the carcass was in the dry stage of decomposition. Estimation of biomass loss at this point was based on the parameters described in method 1, and since the water content of pigs is approximately 47 %. Therefore, when the body is fully mummified and reduced to only skin and bones, biomass loss could be estimated to be reduced to one half of the eight pounds. This would give an estimated biomass loss of 96 percent, considering that the skeletal weight is less than five percent.

Daily ambient temperature and relative humidity at the site were recorded with a hand held Extech Big Digit Remote Probe Hygrometer (item #445715) that measures relative humidity within the interval of 10-99% and temperatures from -10°C to 60°C . It has an accuracy of $\pm 4\%$ relative humidity and $\pm 1^{\circ}\text{C}$. The carcass and maggot mass temperatures were obtained using an Oakton Temp. 5 Thermister Thermometer Digital Probe. Internal temperature was recorded by way of the mouth and anus during the early stages of decomposition. Thereafter, internal temperature was determined by directly inserting the probe into the gut. Soil temperatures were taken by inserting the temperature probe to a depth of three inches into the soil. Insect activities were observed daily on the carcass. Adult Diptera and Coleoptera were collected using a sweep net and forceps, respectively. Larval specimens generated by the adults were collected from various anatomical positions on the carcass and four to six feet of the outfield over a three days' interval using a forceps or a 15ml plastic spoon. Two hundred of these larvae were counted for the first three days of active decay. Two hundred puparia were also collected randomly within four to six feet of the carcass on day 3 and reared in small plastic

containers as seen in Figure 3. Larval specimens were killed in boiling water and preserved in 70% ETOH prior to being analyzed. The emerging adults were then counted. Adult insects were placed in a standard kill jar fumigated with ethyl acetate for approximately two minutes, then labeled and pinned for preservation and identification.



Figure 3. Dishes used for rearing puparia collected from bait 1 carcass

3.1.2 Phase 1 Supplementary Survey Study

Phase 1 supplementary survey was conducted to assess the spatial distribution of Calliphorids and their levels of activities during the summer. Adult specimens were collected at Arlington (Kingston 10), Crosswords Municipal Market (Kingston 5), Kingston Public hospital (KPH), Papine, St. Andrew (Kingston 6), Port Royal (Kingston 5), and Mavis Bank, St. Andrew. One collection was made at each geographic location from July 2 to July 20, 2007. Sweep net and baited traps were used to collect adult flies depending on the nature of the habitats. Those collected from garbage disposal (household or restaurant) were collected with a sweep net, and baited traps were used to collect those from the open fields. When baited traps were used, they

were baited with 112 g of liver and allowed to stand for two days at each location before collection was done. Adults collected were placed in a kill jar fumigated with ethylacetate, then pinned and labeled.

3.2 Phase 2 Study Using Bait 2 (Goat Head) and Bait 3 (Tilapia Fresh Water Fish)

The Phase 2 study was also conducted at the Forensic Laboratory compound from February 11 to February 15, 2008. The purpose of this study was to assess the relative abundance of forensically-important species at different time periods of the year. One freshly processed goat head (bait 2) was purchased from the commercial meat market on February 11, 2007, about 12 PM and placed at the site at 1 PM the same day to attract species of forensic importance. The hair was already scraped from the head and a small amount of blood was seen on the exposed surface of the severed neck. The viscera of the head were still intact. Bait 2 was placed in a ½ inch wire mesh basin and hoisted four feet from a pulley anchored to a tree branch (Fig.4) to prevent predation and to be able to move it up and down as desired.

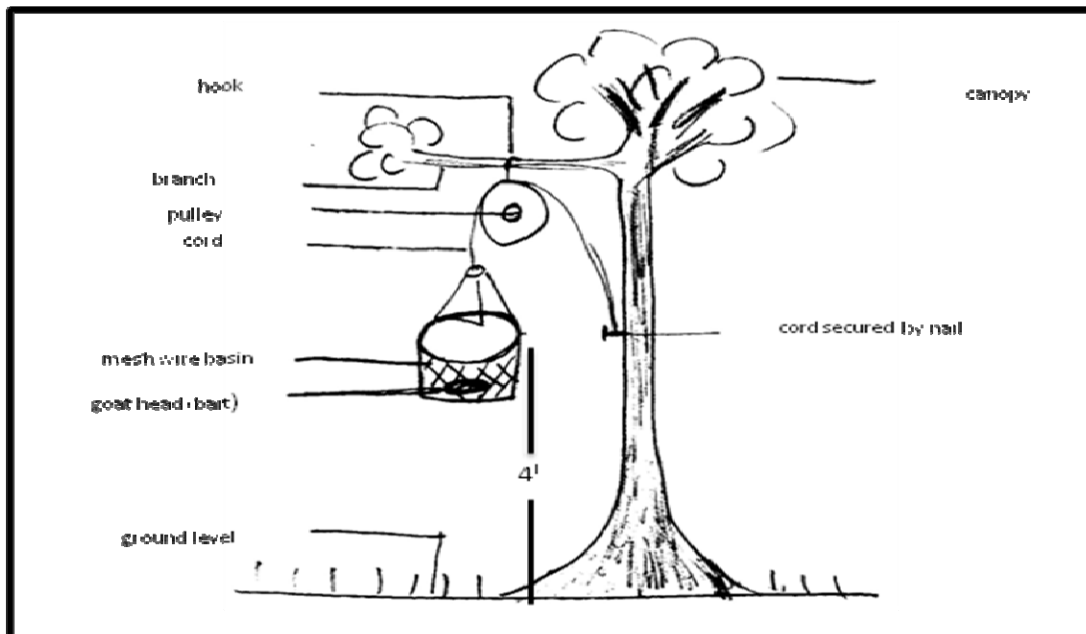


Figure 4. Technique used to trap flies with bait 1

Bait 2 was exposed until February, 13, 2008. At the end of the two days, the basin containing the colonized bait was lowered via the pulley into a large transparent polythene plastic bag. The bag was shaken lightly for thirty seconds to dislodge the adult flies. The plastic bag was then tied into an inflated ball to allow the flies space to move away from the bait but not escape. A hole was cut at the bottom of the plastic bag and then immersed into another plastic bag to collect the flies. While the baited basin was removed from the original plastic bag, the flies exited and entered the plastic bag placed below. The objective of this seemingly elaborate move was to ensure that all flies present were captured and represented the colony. A nine-ounce glass jar with a perforated cap containing paper towels saturated with ethylacetate was placed into the inflated plastic bag with the fly specimens for three minutes to subdue them. Adult Dipteran specimens collected from the plastic bag were pinned and labeled. About one hundred larval specimens collected from bait 2 were placed on four ounces (112g) of liver in an 8" x 8" x 1.5" plastic dish that was used as the control rearing unit. The container was covered with green nylon mesh held in place by rubber bands.

Bait 2 was again hoisted and allowed to be re-colonized for an additional two days until February 15, 2008. At the end of this period, one fresh water tilapia fish weighing 224g and labeled bait 3 was placed conspicuously on top of a juice crate about three feet from bait 2. Bait 3 was exposed for twenty minutes; about ten adult flies were observed flying in and out of the head area of the fish before they were intercepted and the bait collected with the first set of eggs observed. After 20 minutes of exposure to flies, the fish was removed and placed on a piece of cardboard. Using a magnifying lens, one batch of eggs was seen between the gills on the fish head. At this point, bait 3 was removed and placed in a ¼ inch steel frame metal cage with dimensions of 24" x 24" x 18 " (Fig.5) which was covered with green nylon mesh. The larvae of bait 2 and bait 3 were transported to Spanish Town, St. Catherine, about ten miles away or

fifteen minutes driving distance from the site. They were transported inside the back of a truck so as not to cause any undue effect on development that could result from temperature changes. Ambient temperatures were recorded during transportation. No significant change in temperature was observed during transportation. Bait 2 larvae were reared from February 13 to February 28, 2008, and bait 3 larvae were reared from February 15 to February 24, 2008. The cage containing the larvae of bait 3 was hanged about five feet off the ground from one of the rafters that support a small zinc awning outside a house. The container with bait 2 larvae was placed in the same area on a table about three feet high from the ground. Both rearing apparatuses were exposed to natural ambient environmental conditions void of the impact of direct sunlight. The awning is made of galvanized zinc material, which did not affect the natural ambient environment.



Figure 5. Cage used to rear larvae of *C. megacephala* collected as eggs on bait 3 at the Forensic Laboratory site, reared from February 14 – 24, 2008, at Spanish Town

The specimens of bait 2 and bait 3 were reared to adult. Observation of larval growth began when the larvae hatched from the eggs collected from bait 3 on February 15, 2008, at 9 AM as opposed to larvae of bait 2, where the developmental recordings started at the time the larvae were recovered on February 13, 2008 at 11 AM.

The eggs collected on bait 3 produced about one hundred and fifty larvae. Ten larval specimens were collected as a cohort daily prior to first moult, and twice daily for successive moults until pupariation began. A forceps was used to collect the larvae. Each batch of cohort specimens was killed in boiling water, preserved in vials of 75 0% ETOH and measured. Each container was labeled to reflect the time and date of each collection. Neither larvae nor puparia were collected throughout the rearing of those collected on bait 2. They were allowed to pupate and emerge in the container in which they were reared. Puparia from the bait 3 rearing unit were removed from the rearing cage as soon as they were formed and placed in individual rearing containers (Fig. 6) until they emerged as adults. This method ensured accurate identification of the emerged adults. As the adults emerged, they were individually labeled with date and time of emergence and pinned. Puparia casings were collected and preserved naturally in small vials.

The following method was used to compute the duration of the stages of development for *Chrysomya megacephala* larvae. The time elapsed between successive instars were assigned by an add-on method, which is based on the last observed stage of development. The add-on method assumed that if a moult did not occur on the last visit, but was discovered on the succeeding visit, then the time that the moult occurred will be one half the time of the observation interval plus the time that was previously accumulated. In other words, 50% of the interval's time is added to the time the larva took to reach the last observed stage, and the remaining 50% of the time marked the beginning of the next stage.

The following equation illustrates:

Total time of last observed stage + 1/2 (observation interval) = duration of last observed stage.



Figure 6. Small plastic dishes used to rear *C. megacephala* from puparium to adult

A precise estimate is dependent on the duration of the observation interval using this method. The body length of all larvae was measured and the accumulated time of development was calculated for all the instars. A line graph was plotted for age (day) versus body length for each instar of *C. megacephala* recovered from bait 3. Statistical analyses of their developmental data were done using Microsoft Excel 2007. Postmortem interval estimations based on accumulative degree hours (ADH) was calculated by taking the product of each *C. megacephala* stadium and the average of the four daily period temperature readings recorded at the rearing site in Spanish Town. The ADD was calculated by dividing the ADH by the hours in one day. The principles are constructed from the method used by Goff (2001) in Hawaii. The following equations summarize the calculation:

ADH = Average temperatures of four daily readings x time taken to reach each stage of development

ADD = degree hours/ total hours in one day

A hand-held hygrometer was used to record the daily temperatures and humidity at the rearing site in Spanish Town. The historical pattern of daily ambient temperature in Jamaica, showed that temperatures begin to increase at 6 AM and peak at 12 PM. After a lag-phase of approximately six hours, the temperatures begin to decline at 6 PM, followed by another lag phase, and further decline at 12 AM each day. Based on this knowledge, aerial temperatures along with their corresponding humidity were recorded simultaneously for half-hour intervals at 6 AM, 12 PM, 6 PM, and 12 AM. Ambient temperatures were also retrieved from the national weather station at the Norman Manley International Airport. Adult Dipteran specimens were sent to Dr. Terry Whitworth in the State of Washington for species identification.

CHAPTER 4: RESULTS

A total of 12 arthropod taxa were collected during the study from July 5 to July 24, 2007, which represent three orders and eleven families (Table 3). Adult Calliphoridae, Sarcophagidae, and Muscidae were the most important forensic Dipteran families collected from the forensic laboratory site. Bait 1 in this study attracted a very large population of adult Calliphorids similar to the observations made by Hewadikaram and Goff (1991) at the Diamond Head Crater, Hawaii. They found that the bigger of the two carcasses (8.4kg and 15.1kg) in their study had “resulted in a disproportionately larger load of maggots” (pg. 236). The adult Calliphorids recovered from bait 1 (pig carcass) were *Chrysomya rufifaces* (M.), *Chrysomya megacephala* (F.), and *Cochliomyia macellaria* (F.). Ants (Formicidae) were the first arthropods to colonize the bait 1 carcass within five minutes of exposure on early day 1 and they continued their activities as predators the larvae throughout the study. Adult flies were prevented from ovipositing at the wound due to the overwhelming density of ants. *C. macellaria* was the first adult Calliphoridae species to arrive at the bait 1 carcass in the first hour of exposure, followed by adult Muscidae. The greatest number of flies was present on day 3. Only one Sarcophagidae was collected near the bait 1 carcass, which represented the only species observed at the site. The Sarcophagids were present in small numbers which limited the catch to only one species. They appeared to shy away from the surrounding heat and were mostly observed in the nearby shade. They showed the tendency to move farther away from the carcass if the blow flies moved in their direction. Although a large number of adult Muscidae was collected, they were rarely seen on bait 1 carcass during the early stages of decomposition. Most of their activities were associated with the putrefied fluids on the surface of the substrate. The rate and stages of decomposition and arthropod succession pattern did not follow the usual trends as seen in other studies.

The carcass was placed at 4:30 PM; therefore, two hours were remaining before sunset at 6:30 PM. This period will be referred to as early day 1. Day 1 would have been completed within the first sixteen and one half hours. The terms early day 1 and late day1 will be used to distinguish the time periods for that day. Larvae collected from the bait 1 carcass were represented by *C. rufifaces*, *C. megacephala*, and *C. macellaria* in different proportions, with those representing *C. rufifaces* being the most abundant. *C. rufifaces* and *C. megacephala* were in greater number than *C. macellaria*. Blow flies were the dominant species on the bait 1 carcass from day 1 until day 4 when the carcass was approaching mummification. The blow flies laid most of their eggs during the bloat stage on day 2 (Goff 2001). *C. macellaria* and *C. megacephala* showed oviposition preferences by depositing most of their eggs along the alveolar between the teeth and deep under the ventral cavity of the carcass and ground interface, respectively. Two desiccated adults of *C. megacephala* flies were recovered at the interface of the ground surface and the ventral area of the carcass, which indicated that they may have succumbed to hyperthermia caused by the high environmental temperatures. Muscid flies continued their activities at the area that was saturated with body fluids even after the decomposition process was completed. Coleoptera arrived relatively early at the carcass during this study, with Chrysomelidae (Leaf beetle) arriving as soon as day 2, followed by Tenebrionidae, Cleridae, and Mycetophagidae (Hairy fungus beetles). The Clerids were the most consistent beetle throughout the observation period. Apart from Chrysomelidae, the other Coleoptera were observed mainly early mornings or late evenings. One wasp (Hymenoptera) was observed on day 4. One Black Soldier fly (*Hermetia illucens*) arrived at the carcass on day 6. The first sets of blow flies emerged on day 9 naturally and continued to emerge until day 15. After day 15, no blow fly species were present at the site of carcass decomposition.

Table 3. Arthropods collected from bait 1 carcass from July 5-24, 2007, at the Forensic Laboratory

Day	Stage	Order	Family	Genus/species
7/5/2007	Fresh	Hymenoptera	Formicidae	sp.
		Diptera		<i>C. macellaria</i>
7/6/2007	Bloat	Hymenoptera	Formicidae	sp.
		Diptera	Calliphoridae	<i>C. macellaria</i>
				<i>C. megacephala</i>
				<i>C. rufifaces</i>
		Muscidae	<i>Musca domestica</i>	
7/7/2007	Bloat	Hymenoptera	Formicidae	sp.
		Diptera	Calliphoridae	<i>C. macellaria</i>
				<i>C. megacephala</i>
				<i>C. rufifaces</i>
		Muscidae	<i>Musca domestica</i>	
7/8/2007	Active decay	Hymenoptera	Formicidae	sp.
		Diptera	Calliphoridae	<i>C. macellaria</i>
				<i>C. megacephala</i>
				<i>C. rufifaces</i>
			Sarcophagidae	<i>sp.</i>
			Muscidae	<i>Musca domestica</i>
	Piophilidae			
	Coleoptera	Chrysomelidae	<i>sp.</i>	
7/9/2007	Active decay	Hymenoptera	Formicidae	sp.
		Hymenoptera	Vespidae	<i>Vespa sp.(Wasp)</i>
		Diptera	Calliphoridae	<i>C. macellaria</i>
				<i>C. megacephala</i>
				<i>C. rufifaces</i>
			Muscidae	<i>Musca domestica</i>
	Piophilidae	<i>sp.</i>		
	Sarcophagidae	<i>sp.</i>		

Table 3 (continued)

		Coleoptera	Tenebrionidae	<i>sp.</i>
			Cleridae	<i>sp.</i>
7/10/2007	Active decay	Hymenoptera	Formicidae	<i>sp.</i>
		Diptera	Calliphoridae	<i>C. macellaria</i>
				<i>C. megacephala</i>
				<i>C. rufifaces</i>
			Muscidae	<i>Musca domestica</i>
			Piophilidae	<i>sp.</i>
			Sarcophagidae	<i>sp.</i>
		Coleoptera	Cleridae	<i>sp.</i>
			Tenebrionidae	<i>sp.</i>
			Mycetophagidae	<i>sp.</i>
7/11/2007		Hymenoptera	Formicidae	<i>sp.</i>
		Diptera	Calliphoridae	<i>C. rufifaces</i>
				<i>C. megacephala</i>
			Muscidae	<i>Musca domestica</i>
			Piophilidae	<i>sp.</i>
			Stratiomyidae	<i>Hermetia illucens (Black soldier fly)</i>
		Coleoptera	Cleridae	<i>sp.</i>
7/12/2007	No collection	No collection	No collection	No collection
7/13/2007	Remain	Hymenoptera	Formicidae	<i>sp.</i>
		Diptera	Muscidae	<i>Musca domestica</i>
			Piophilidae	<i>sp.</i>
		Coleoptera	Cleridae	<i>sp.</i>
7/ 14 to		Hymenoptera	Formicidae	<i>sp.</i>
7/18/2007		Diptera	Muscidae	<i>Musca domestica</i>
			Piophilidae	<i>sp.</i>
		Coleoptera	Cleridae	<i>sp.</i>
7/20 to		Hymenoptera	Formicidae	<i>sp.</i>
7/24/2007		Diptera	Muscidae	<i>Musca domestica</i>

The first day of larval collection on day 3 is represented in Table 4. There were marked variations in sizes among larvae collected at the various anatomical positions on the carcass. The larvae of *C. megacephala* and *C. macellaria* were significantly larger in size than those of *C. rufifaces*. Most of the larvae of *C. rufifaces* were small in size and greater in number at all anatomical areas of the carcass. A small population of *C. megacephala* larvae was recovered only on the first day of larval collection in a dense population of *C. rufifaces* in the vicinity of the perimortem wound at the throat of the carcass. Both larval specimens were similar in body length. Unlike *C. megacephala*, *C. macellaria* was recovered from the anterior area of the carcass in a less dense population of *C. rufifaces* larvae. The larvae of *C. rufifaces* were predominantly 3rd instar followed by 2nd instars and 1st instars.

Table 4. The size distribution and anatomical positions of larvae collected on bait 1 carcass, July, 8, 2007 (day 3) at the forensic laboratory

Anatomical location on bait 1 carcass	Stage	Genus/species	Larval sizes/range
Anterior			
head	L3	<i>C. macellaria</i>	12 -14 mm
	L3	<i>C. rufifaces</i>	6 mm-10 mm
Posterior			
anal region	L1, L2, L3	<i>C. rufifaces</i>	4.5 mm-7 mm
Ventral			
abdomen	L2, L3	<i>C. rufifaces</i>	5 mm-7 mm
neck -wound	L3	<i>C. megacephala</i>	8.5 mm
		<i>C. rufifaces</i>	8.5 mm
Dorsal			
Top line or back		<i>C. rufifaces</i>	6 mm-8.5 mm

The larvae *C. megacephala* and *C. macellaria* were all 3rd instars. Of the three larval populations collected, *C. macellaria* and *C. megacephala* showed the least variation in body length. Larvae collected from the dorsal and posterior area of the carcass were purely *C. rufifaces* and were also severely undersized. During the second collection, the larval population continued to be dominated by *C. rufifaces*. A small population of *C. macellaria* was again

recovered from the anterior area of the carcass. Table 5 gives the general counts and measurements done for each species collected for four daily collections. After the first collection on day 3, *C. rufifaces* larvae showed a progressive increase in their size. A few adult flies of *C. rufifaces* and *C. megacephala* were still hovering about the carcass up to day 6, probably due to the persistency of putrid fluids within the soil under the remains. A search was conducted within the eighteen-inch grass with heavy mat at the ground surface. It formed a dense perimeter about four to six feet around the carcass, as well as under an inch of soil. Although puparia were found in various parts of the outfield around the carcass, the greatest number was discovered about five feet west of the carcass, under grass mat and soil. Two hundred puparia were randomly collected, measured, and reared in small plastic containers at an average temperature of 30.7 °C. The size range of puparia was 4.5 mm – 8 mm.

Table 5. General count and size ranges for species recovered from bait 1 carcass

Day-collection	<i>C. rufifaces</i>		<i>C. megacephala</i>		<i>C. macellaria</i>	
	Length	Number	Length	Number	Length	Number
3-1 st collection	4.5 -7 mm	160	8.5-9mm	15	12-14 mm	25
4-2 nd collection	4.5-8.5 mm	190	nil	0	12.25	10
5-3 rd collection	5-10.5 mm	200	nil	0	nil	0
6-4 th collection	11-14 mm	200	nil	0	nil	0

Adults that emerged from these puparia were measured from the proximal tip of the prothorax behind the head to the posterior tip (ovipositor in female) of the abdomen. Variations due to suboptimal conditions are more likely to affect body size than head size in the adults. Adult *Cochliomyia macellaria* and *C. megacephala* averaged 7 mm in body length as opposed to *Chysomya rufifaces* that averaged 5 mm in body length. Of the two hundred puparia reared, one hundred and thirty-three emerged as adults which indicated a mortality to emergence ratio of one

death for every three adults emerged, a 66% survival rate. Of the total number of live adults that emerged, *C. rufifaces* accounted for 93 %, *C. megacephala* for 3 %, and *C. macellaria* for 4 %.

Ambient temperatures and humidity levels that were recorded at the forensic laboratory exceeded those recorded for the area by the national weather station at the Norman Manley International Airport, which is located approximately 8 km southeast from the site. As shown in Fig.7, the characteristic differences of maximum and minimum temperatures from the weather station for July showed very little variation in daily patterns along with their corresponding daily average humidity. Ambient maximum and minimum temperatures from the weather station averaged 35 °C and 27 °C, respectively, with corresponding humidity averaging 64 % between both temperatures limits. Temperatures recorded physically at the forensic laboratory had a high of 39.1 °C and a low of 23 °C (Table 6). Comparatively, ambient temperatures and wind speeds recorded by the weather station showed a direct relationship to each other and an inverse relationship to relative humidity. The highest temperature and wind speed are recorded during the afternoons and the lowest are recorded during the nights. Both wind speed and direction showed variability in both speed and direction, with an average of 13 km/h or 8 mph in an ESE direction. Ambient temperatures fluctuated on an average of approximately +/- 14 °C between high and low temperatures and reached their lowest mark between 5 AM and 6 PM daily, then gradually increased at an average rate 1.5-3°C/h before peaking between 12 PM and 1 PM daily (Fig.8). Day length was 13 hours and 10 minutes as opposed to visible day length of 13 hours and 48 minutes.

Activities associated with the decomposition processes of bait 1 carcass (Phase 1) study at the forensic laboratory site from July 5 to July 24, 2007, are shown in Table 6. As seen, maggot mass temperature rose to lethal levels beginning on the morning of day 3 with the

temperature rising from 39 °C to a high of 49 °C in the afternoon, which corresponded to a similar surge in body surface temperature from 38 °C to 65 °C over the same period. Maggot mass temperature remained constant from day 3 to day 6 during active larval developmental stages. Maggot mass sampling showed that the larval aggregates in the mass were mostly

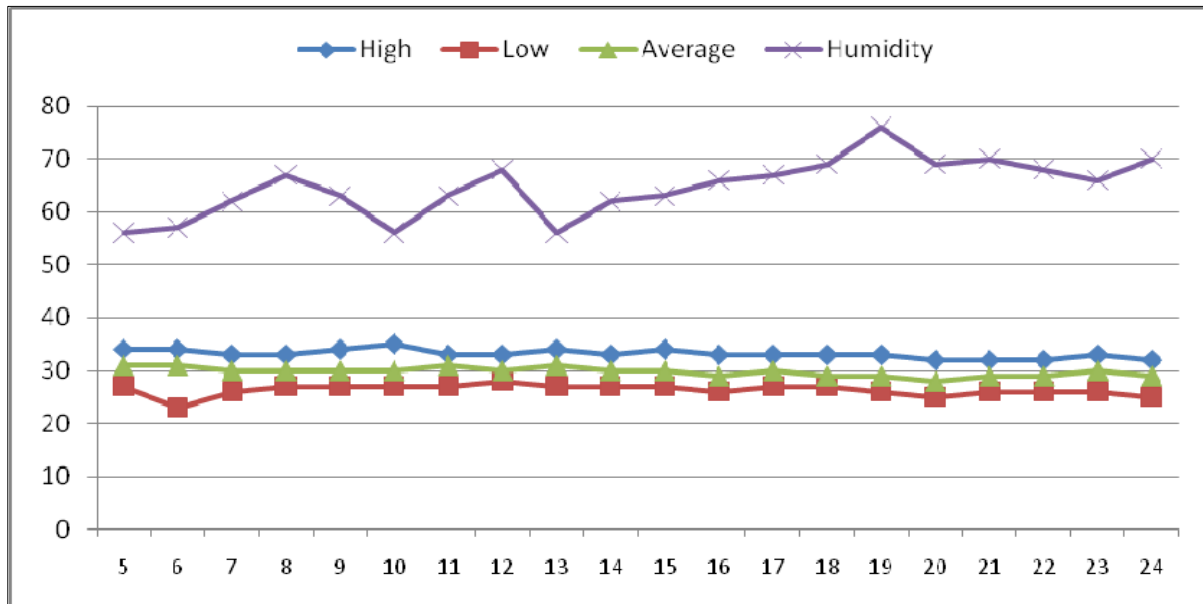


Figure 7. Daily ambient temperatures recorded by the national weather station for the area where bait studies was conducted from July 5-24, 2007, at the Jamaica forensic laboratory.

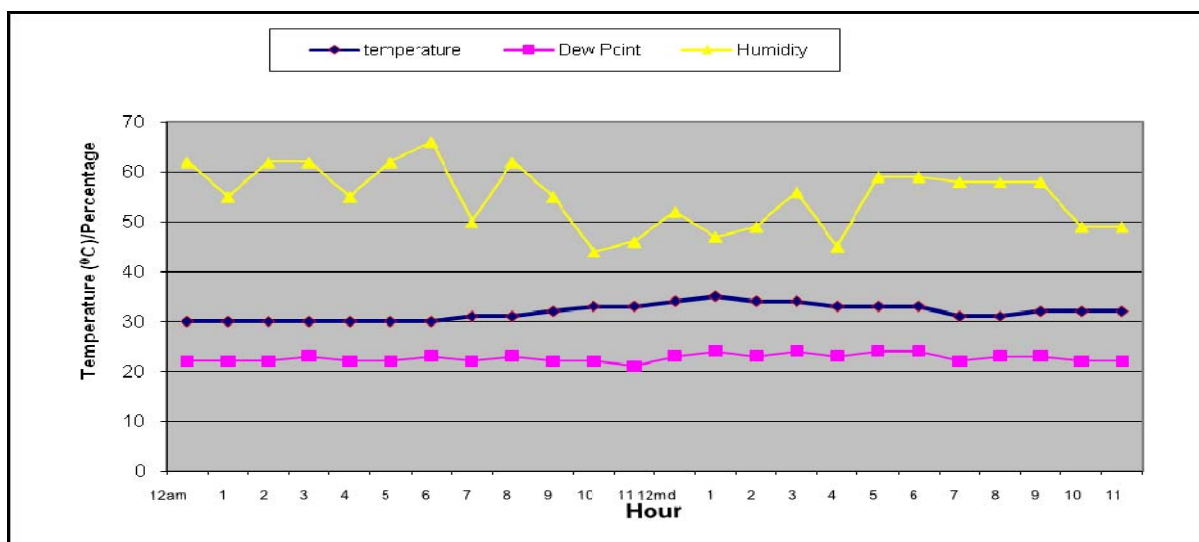


Figure 8. Hourly pattern of ambient temperatures and humidity for July 10, 2007, at the forensic laboratory site in Kingston and St. Andrew recorded by the national weather station

Table 6. Temperatures ($^{\circ}$ C) collected from bait 1 carcass at the forensic Laboratory

Date	Mouth	Anus	Soil	Carcass surface	Maggot mass	Internal	Interface	Site temperature	Remarks
7/5/2007	32	30						33	Fresh Rigor mortis, Algomortis
7/6/2007	43.6	29.2	26.4					31.5	Liver mortis Bloat/Putrefaction
		34.2	31.2						
7/7/2007	25	24.5	25.5				28		Bloat/Putrefaction
	31	36					33		Bloat/Putrefaction
7/8/2007	31	32	26	65	45	60	38.7	36.1	Active decay 46 % biomass loss Mass migration of immatures
	34	42	34						
7/9/2007	23.5	47	38.7	51	47	49	44.5	28	Active decay
	36				48.5			37.7	69 % biomass loss Mass migration of immatures
7/10/2007		41.8	41	48	48	55.1	51.7	23.7	Active decay
								37	Active decay
7/11/2007		39	34.2	34.7	47.4	38	42.6	23.6	Mummification process
								35	Carcass mummified
7/12/2007									
7/13/2007			42		39		39.2	39.1	Remains 95 % biomass loss
7/14/2007			30.7					33.8	Remains
7/15/2007								35.4	Dried skin and bones

*Columns with two temperatures indicate morning and afternoon recordings

late 2nd instars and early 3rd instars. Most maggots had migrated by day 6, which showed a decline in the temperatures of both maggot masses and carcass. Scorched remains of the carcass continued to accumulate temperatures above ambient temperatures, but gradually declined toward ambient, after the maggot mass density had reduced significantly and decomposition was approaching the remains stage. Anal temperatures increased gradually from day 2 and got closer to maggot mass temperature by day 3; they then moved along a negative temperature gradient on day 6. Temperatures collected from the mouth remained steady with a daily alternating increase and decrease pattern during the mornings and afternoon, respectively. All temperatures except maggot mass temperatures showed a positive response relationship with ambient temperatures. Maggot mass temperature had an effect on rate at which heat was dissipated from the carcass or substrate under the carcass.

Day 1-2: Decomposition progressed very rapidly at the latter part of the fresh stage (arrow points to perimortem wound) by the anaerobic decomposers (Fig. 9). The bloat stage (Fig.10) was noticeable within the first twelve hours of placing the carcass on day 1, lasting until day 2. During this stage, lesions were observed on the abdomen of the carcass, presumably caused by ants. Decomposition of the carcass was rapid after the bloat stage, principally because of the high density of sarcophagous larvae. Decomposition progressed very rapidly without discrete demarcations until the carcass reached the dry stage on day 5 and complete mummification on day 8. Biomass removal was much faster than anticipated during the decomposition stage. The rate of decomposition observed in this study is comparable to the observations made by Chin et al. (2007). They observed that in Malaysia, the rate of decomposition was also fast when compared to studies in other regions, such as those conducted in Hawaii and Australia. A comparative overview of environmental data and time to reach the dry stages of both studied are summarized in Table 7.

Day 3: Environmental temperatures were so high that the adipose tissue under the skin melted to oil, which created a visible sheen over the top skin and saturated the substrate under the carcass even after decomposition was completed (Fig. 11). Massive numbers of *C. rufifacies* larvae were feeding within the gut of the carcass (Fig. 12). The carcass was in a rapid state of decay and biomass had reduced to about 41.6 % by weight, which exposed the internal viscera of the gut, scapula and thoracic ribs. Also, most larvae were concentrated at the ventral and posterior interface between the carcass and ground. The carcass was still identifiable at this stage with skin intact. The ground was covered with migrating larvae most which 2nd and 3rd instars. Most larvae were concentrated in a large maggot mass at the ventral surface of the chest, while those moving about the carcass were distributed around the perimeter.



Figure 9. Fresh stage of bait 1 carcass at the forensic laboratory site, July 6 – 24, 2007, arrow points to fresh blood oozing from perimortem wound



Figure 10. Bloat stage of decomposition

Table 7. Parameters associated with current study and study done by Chin et al. 2007

Results	Month & Place	Carcass weight	Ave. temp.	Ave. humidity	Ground surface temp.	Day dry stage observed
A	May (Malaysia)	8.5kg	31.1±1.54	82.2± 8.52	30.4 ± 0.79	8
B	July (Jamaica)	23 kg	30 ± 0.812	64.7,± 5.3	36.5 ± 3.9	5

*A represents current study in Jamaica and B represents study done by Chin et al. 2007 in Malaysia

Day 4: The bait 1 carcass was reduced by about 62 % and approximately 50 % of the larvae population recovered was burned or died from hyperthermia (Figs. 13 and Fig. 14). Larvae continued to migrate, most of which were 3rd instars. Active decay continued and larval populations were declining gradually. The body was still recognizable with more skeletal parts being exposed along the ventral and dorsal aspects. The head was still intact though it was obvious that only the skeletal frame of the skull was left at this stage. The body began to lose most of the skin on the ventral and dorsal aspects with the central portion still connected to the upper portion of the rib cage. Most of the front and hind legs and vertebral column were exposed, though the body was still recognizable.



Figure 11. Carcass showing oil sheen from decomposing tissues. Note uneven distribution of larvae away from wound indicated by the red dot



Figure 12. Massive maggot aggregate day 3 on carcass: *C. rufifaces* larvae as inset



Figure 13. Reduction in larvae population and distribution about carcass along with exposed skeletal parts



Figure 14. Dried remains showing remnants of burned or desiccated larvae. Femur, tibia, hip joint, ribs, metatarsals and vertebral column are exposed

Day 5: 79 % of the body mass was lost although the anatomical features of the carcass were still recognizable. The ribs, vertebrate, most of front and hind legs, cephalic region, vertebrate, and scapulae were the exposed skeletal parts with most of the skin separated from the vertebrate and abdominal region in original positions (Figs.15 and 16). After the liquefied gut content became scarce, larvae were observed feeding on sun burned tissues (Fig 15) left over from the abdominal region of the body. As seen in Fig. 15, mosaics of observable holes were left in the skin. Larvae continued to migrate at this stage. The larvae that were left at this stage appeared to be affected by the heat of the remains. They were not as agile as those seen on day 3, as they wandered over the remains during the daytime. At this point, the larvae fed mostly on desiccated tissue that was left. During the latter part of the evening, the larvae that were under the grass tend to reassert themselves and became more visible on the surface of the remains. Beetles that were present, spent short periods on the remains before crawling or flying back to the shaded area.



Figure 15. Adult *C. rufifaces* feeding on the lacerated skin left by feeding sarcosaprophagous larvae



Figure 16. Completely decomposed skull with skin intact and scapulae displaced

Day 6: Only the desiccated skin, bones and parched viscera were left of the carcass. Dried tissues appeared as a ball of ash (Fig. 17). Larval migration was almost completed at this stage, with only a small number wandering about in the field and in the carcass ash. Most of the skeleton had been exposed and recognition of the original carcass features was deteriorating.



Figure 17. Larvae moving about over crucible (ashlike) mummified remains of carcass

Day 8: The carcass lost recognition at this stage of decomposition as the parched tissues disintegrated and dried skin separated from most bones, with the carcass mass reduced by approximately 95 percent. The head, anal region, and tibia were the last carcass morphological areas to lose their identity. A small number of adult Calliphorids continued to circle about the carcass and ground saturated with putrefied oils, but no colony was formed. Residual larvae continued to feed on the parched tissues and mummified skin. Larval movement was obviously paralyzed by the impact of ambient conditions from the sun's radiation. Remains (Fig .18) were .disturbed overnight by predators (dog); this did not affect the study as only dried skin and bones were left. The remains were not ruined by the dogs and were placed back in their original location until total skeletonization.



Figure 18. Dehydrated skin and skeletal fragments left from decomposition

Day 9 -13: A small amount skin was still attached to the vertebral column, but by day 13, the carcass was completely skeletonized. Bones collected were allowed to sit until July 24, 2007. During this time only the Cleridae, Muscidae, and Piophilidae intermittently visited the bones. The beetles appeared to do most of their work during the night as they were seen late evenings to early mornings. The impression that decomposition had occurred remained obvious to the end of the study as the soil remained stained with the by-products of adipose tissue seen on day 3 (Fig. 19).



Figure 19. Impression of carcass carved out by the retention of oils generated by melting adipose tissue

4.1 Phase 1 Supplementary Survey Study Results

Two species of forensic importance were collected during the supplementary survey of Calliphorids at various locations in Kingston and St. Andrew. They are *C. megacephala* and *Lucilia cuprina*. The geographical locations along with the species and their habitats are summarized in Table 8. *C. megacephala* was the most dominant species collected at various sites across Kingston and St. Andrew. 4.1.1 Phase 2 results using bait 2 and bait 3

Three families of forensic importance were collected during phase 2 of the study: Calliphoridae, Sarcophagidae, and Muscidae. The Calliphoridae species were *Lucilia lucigerens* (James) and *C. megacephala*. Adults and larvae of both species were collected from bait 2. Results from the rearing of the larvae collected from bait 2 on February 11, 2008, produced two species: *C. megacephala* and *L. lucigerens*. *Lucilia lucigerens* is indigenous to Jamaica. Bait 2 larvae were reared from February 13 to February 28, 2008 as control specimens. Emerging adults were *C. megacephala* and *L. lucigerens*.

Table 8. Distribution of species collected at different locations in Kingston and St. Andrew, July 2-20, 2007

Date of coll.	Method of coll.	No. of coll.	Place of coll.	Elev. above sea level	Geo. Location		Food source	Genus/species
					Lat./Lon.	Habitat		
7/9/2007	Sweep net	1	Rich. Park (Kgn. 10)	14M	18'.00"N-76'80"W	Restaurant	refuse	<i>C. megacephala</i>
7/9/2007	Sweep net	1	Cross Roads (Kgn. 5)	12M	18'.01"N-76'.51"W	Food/meat market	refuse	<i>C. megacephala</i>
7/20/2007	Baited trap	1	Down Town (KPH)	12M	18'.01"N-76'.51"W	Open yard	Liver	<i>C. megacephala</i> <i>L. cuprina</i>
7/2/2008	Sweep net	1	Mavis Bank	887M	18'.03"N-76'.66"W	Open area	Refuse	<i>C. megacephala</i>
7/9/2007	Sweep net	1	Papine (Kgn. 6)	143M	18'.76"N-76'.75"W	Open area	Refuse	<i>C. megacephala</i> <i>L. cuprina</i>
2/7/2007	Baited trap	1	Port Royal (Kgn. 1)	0M	17'.93"N-76'.80"W	Open area	Liver	<i>C. megacephala</i>

Both species had an overlap period of seventy-two hours during migration. *C. megacephala* pupated on day 6 and *L. lucigerens* pupated on day 8. Adult *C. megacephala* and *L. lucigerens* emerged in eight plus and 16 plus days from February 13 to February 28, 2008), respectively. Larvae from bait 3 were reared from February 15 to February 24, 2007. The adult species emerged from the eggs collected on bait 3 were 100 % *C. megacephala*. Table 9 summarizes the developmental data and average temperatures at which larvae were reared in the F3 rearing unit. The size of the larvae ranged from 2.8-3mm at eclosion and progressed to attain a maximum

body length of 14.5 mm at the post feeding stage. The duration from egg to adult emergence took 214 hours or 8.9 days. The results obtained from the rearing of *C. megacephala* are compared to those obtained by Wells and Kurahashi (1994) and Velez and Wolf (2008) in Table 10. Adult *C. megacephala* emerged about the same time in both rearing units. Both bait 2 and bait 3 specimens were reared at 26 °C and 64.7 % relative humidity as seen in Table 11.

Table 9. S.D. and the mean larval length for *C. megacephala* reared Feb. 15-24, 2008 in Spanish Town, St. Catherine

Temp. (°C)	Larval length in mm (mean ±SD)			
	L1	L2	L3	PF
26	2.93±0.11	8.29±0.2165	13.23±0.27	14.22±0.3

*S.D. = standard deviation.

Table 10. Comparison of three rearing results for *C. megacephala*

Time to reach developmental stages for <i>C. megacephala</i> (Fabricius) (mean±SD)								
Temp. (°C)	Egg	L1	L2	L3	PF	P	Adult	Sources
23±2.45		18.60±3.44	45.04±9.23	65.58±12.22	107.26±13.68	236±102.53	319±53.74	Velez and Wolf, (2007)
26±0.5	12±0.5	17± 0.45	32± 0.25	76±0.25	108±6.75	141.5±0.25	239±0.25	Current study
27±0.5		18	30	72		144	234	Wells and Kurahashi, (1994)

Table 11. Average temperature recorded Feb. 15-24, 2008, at Spanish Town where *C. megacephala* and *L. lucigerens* larvae were reared

Days	Date	12 MN	6AM	12MD	6PM	Temperatures (°C)	Humidity (%)
1	2/15/2008	24.6	20.5	32.7	27	26.2	63.5
2	2/16/2008	21.3	23	29	27	25	62.75
3	2/17/2008	23	21	34.7	28	26.5	61
4	2/18/2008	21	21.6	35.1	29	26.52	62
5	2/19/2008	20.4	23	31	27	25.35	63
6	2/20/2008	21	21.3	33.6	28.6	25.75	64
7	2/21/2008	22.5	21.2	31.9	27	25.6	67
8	2/22/2008	23.6	21.6	34.3	27.2	26.5	63
9	2/23/2008	20.6	22.1	31.4	26.9	25.25	62
10	2/24/2008	24	21.6	32.1	27.2	26.175	65
	Averages	22.2	21.69	32.58	27.49	25.99	63.325

A line graph for *C. megacephala* indicated a positive growth rate up to the end of the post feeding, at which stage, larval growth began to stabilize (Fig. 20). Approximately one hundred and fifty larvae emerged from the eggs collected on bait 3. A cohort of ten were collected, killed and measured in the respective stages and on a daily basis. There were negligible incremental variations in the transition from one larval stage to the next, which annotates synchrony especially in the first two stadia. The time *C. megacephala* took from egg to adult is summarized in Table 12, and the statistical analysis is presented in Appendix A.

Species distributions were not as expected in February, 2008, (winter) as opposed to July, 2007, (summer). *C. rufifaces* and *C. megacephala* were expected to be prevalent all year round, due to the small changes in ambient temperatures between summer and winter. *C. megacephala* did meet the expectations of being the most abundant and prolific species all year round, while *C. rufifaces* was only collected during the hot summer. Although the aerial temperature was well above the ambient temperatures reported in other research, such as Goff (2001), the species appears not to be attracted to any of the bait used in February. The variations in quantity and quality of the baits may have contributed to the presence or absence of the species in both time periods, as well as it might the normal distribution pattern for the region. The best method to test this is to use baits of similar quantity and quality. The latter summation qualifies the need for research to be done throughout the year before a conclusive conclusion can be drawn about any of the true distributions. It is likely that *C. rufifaces* has a greater preference for larger baits as seen with bait 1 in July, as opposed to *C. megacephala*, which has proven to be more omnipresent. The same argument may hold true for *C. macellaria* which did show up in February, when smaller baited traps were used to attract flies during the species distribution survey in July. *Lucilia lucigerens* appears to prefer the lower temperature time periods in areas that are warm and humid, but not direct exposure to sunlight. They tend to penetrate deep into

their food source during oviposition. The Sarcophagids were more abundant in February than July and represent a different species from those collected in July. Table 13 summarizes the results from the different bait types used in the two study phases and the number of species collected.

Temperatures obtained at the rearing site in Spanish Town via the National weather station at the Norman Manley International National Airport reflected those temperatures collected at the rearing site (Table 10 and Fig. 21). The daily average ambient wind speeds are shown in Figure 22. There were no drastic changes in temperature or wind speed during the study. As expected, ambient temperatures, wind speed, and humidity were fairly predictable at the rearing site. The daily average differences between the high and low temperatures is approximately 12.5 -13.5 °C. Humidity was fairly stable at a range of 39% to 81% daily.

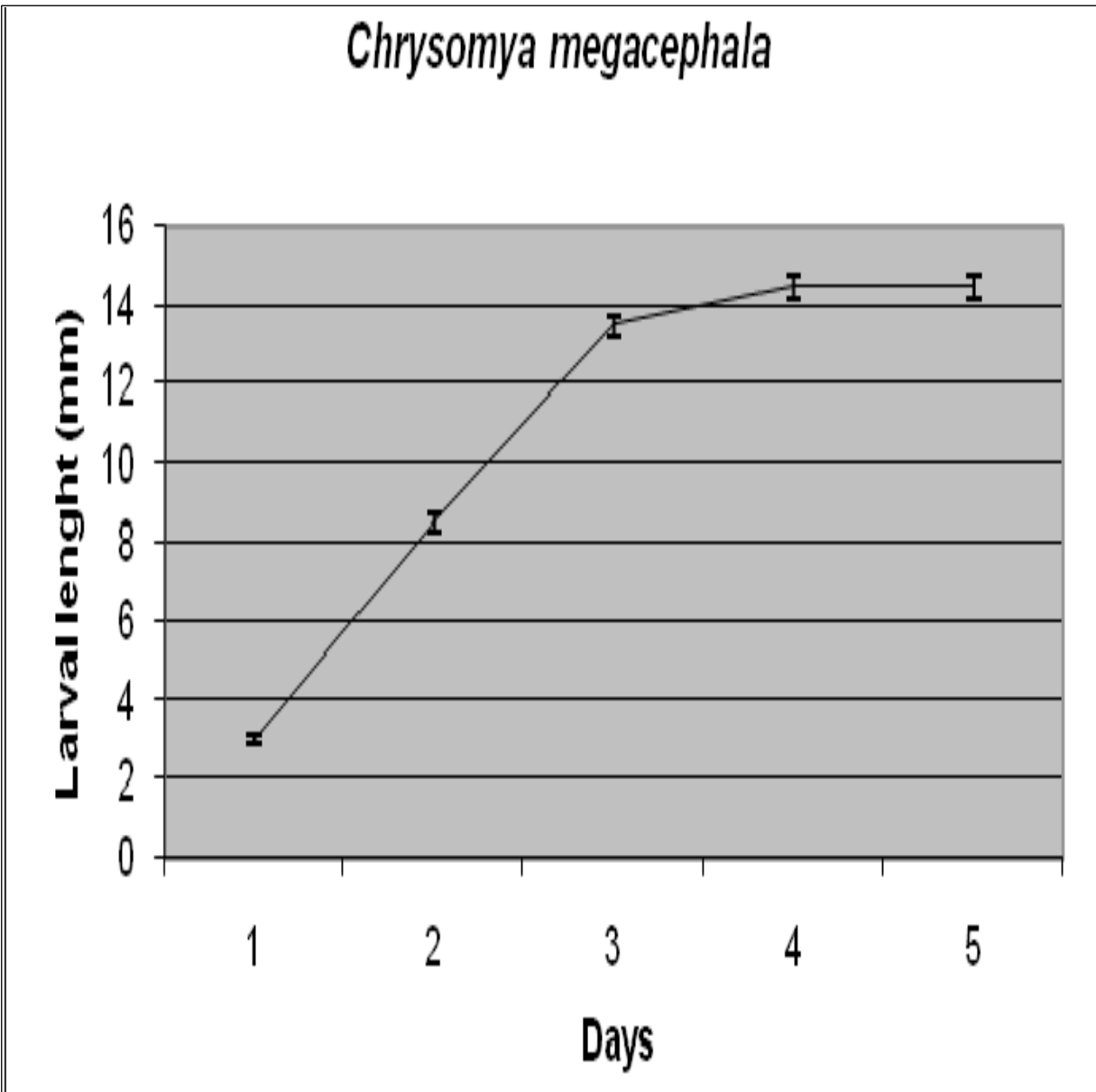


Figure 20. Mean body length vs. age (days) for *C. megacephala* reared from February 15- 24, 2008, in Spanish Town St. Catherine

Table 12. Developmental summary for larvae of *C. megacephala* collected from bait 3 and reared at Spanish Town, February 15 – 24, 2008

Date	Day/Time	Dev. Stages	Dev. Temp.	Duration (h)	exp.ADH	Exp. ADD
2/15/2008	1/ 9 AM-8 PM	Ecdysis	26	12	312	13
2/16/2008	1/ 1:PM	1 st stadium	25.5	17	433.5	18
2/17/2008	2/ 9 PM	2nd stadium	26	32(61)	832	34.66
2/18/2008	3/ 12 MD	3rd stadium	26.7	15(76)	400.5	16.68
2/19/2008	4/ 8PM	Post feeding	25	32 (108)	800	33.3
2/20/2008		Body changed from cream to reddish brown pupation 4th instar	25.95			
2/21/2008	6/ 6 AM	Pupariation starts (reddish-black)	25.6	34 (142)	870.4	36.26
2/22/2008	7	Pupariation	25			
2/23/2008	8	Pupariation	26			
2/24/2008	9/ 6:30AM- 7AM	Adult emerged	22	72	1584	66
Total			26 ± 0.5	214 h/8.9 days	5232.4	218

* Numbers in bracket represent the end of each stadium and those without bracket represent time spent in each stadium from the egg stage

Table 13. Overall collection and distribution of species for the two study periods

		No. of species collected as specimens in phase 1 and 2					
		<i>C. rufifaces</i>	<i>C. megacephala</i>	<i>C. macellaria</i>	<i>L. lucigerens</i>	<i>L. cuprina.</i>	<i>Sarcophagid.</i>
Phase 1	Jul-08 Bait 1	46	65	14	0	0	1
	Supplementary	0	50	0	0	5	1
Phase 2	Feb.-08 Bait 2	0	25	0	5	0	6
	Bait 3		10	0	0	0	0

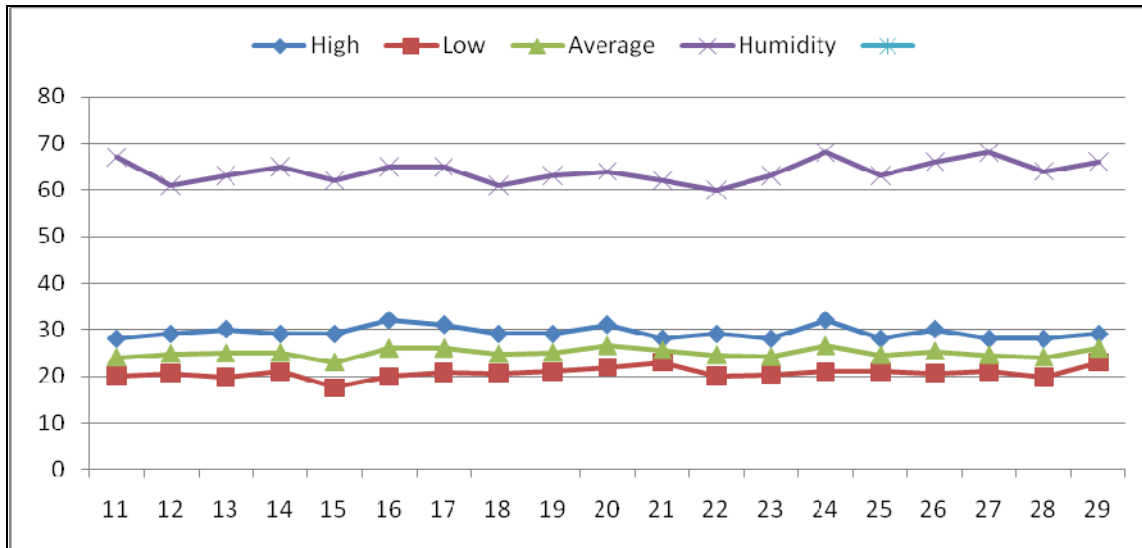


Figure 21. Ambient Temperatures and humidity recorded at rearing site in Spanish Town from February 11 – 28, 2008

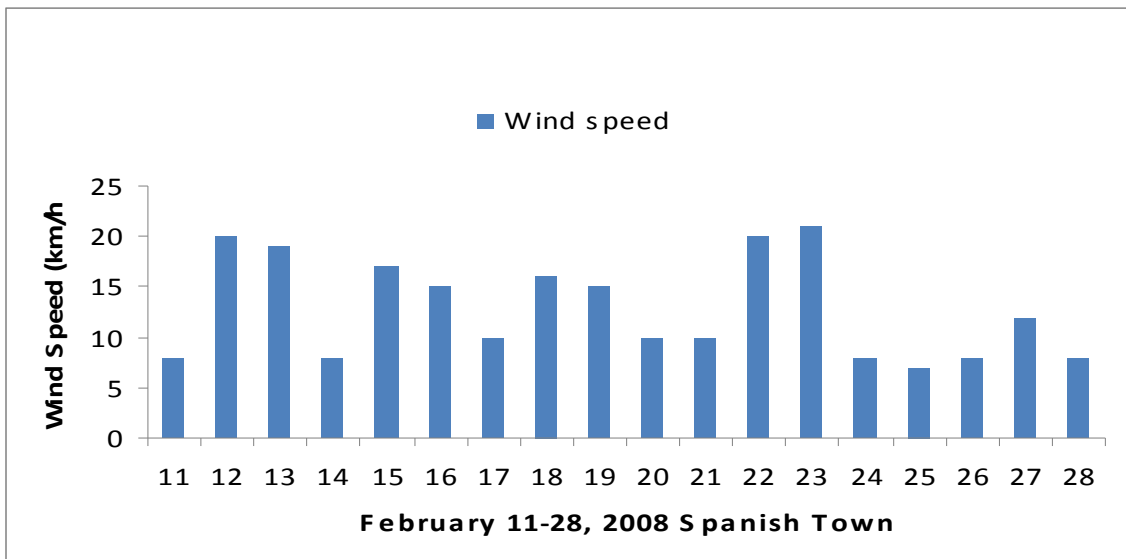


Figure 22. Daily ambient wind speed for Spanish Town from February 11-28, 2008 where *C. megacephala* and *L. luciigerens* were reared

CHAPTER 5: DISCUSSION

5.1 Phase 1 Study on Bait 1 (Pig Carcass)

It is the natural tendency for insect species to find and colonize food sources such as cadavers as a natural means of survival. According to the pattern exhibited by insects feeding on dead organic matter, each group of insects colonizes the cadaver in an orderly predictable sequence; however, these predictions do not always hold true in all circumstances, as observed in Jamaica, but are rather more consistent when the conditions upon which the observations are made are similar. The two most abundant insect species of forensic importance that are found in association with a corpse are also found in Jamaica, the blow flies *C. rufifaces* and *C. megacephala* (Barreto et al. 2002, Chin et al. 2007; Goff et al. 1986; Goff and Flynn 1991; Gunatilake et al. 1989; Hewadikaram and Goff 1991; Lord 1990; Smith 1986). Many authors have commented about the likelihood of a maggot mass generating lethal temperature limits when a carcass is placed in direct sunlight (Deonier 1940; O' Flynn and Moorehouse 1980; Reed 1958). However, there is no report of maggot mass reaching such high levels that resulted in the burning or desiccation of larvae in a maggot mass and the actual collection of burned and desiccated larvae from a carcass under study.

Results from this study demonstrated that even though the sequential processes of decomposition are similar, the rate and pattern of insect colonization are not always the same. After making note of the insect colonization pattern in conjunction with the rate of decomposition of bait 1 carcass placed in direct sunlight, it became apparent that the dynamics of decomposition is geographically dependent, in much the same way as the insects that are associated with it. Other studies found that when carcasses are placed in direct sunlight they will heat up more rapidly and result in faster decomposition (Dillon and Anderson 1997; Dillon and

Anderson 1996; Dillon and Anderson, 1995; Reed 1958; Deonier 1940; Shean et al. 1993). Similar observations were made during this study, but the effect of temperature had far more impact on the total environment, including the decomposition processes. Studies done in open environments did not report temperature as having any grave effect on the faunal succession. For example, at a stable minimum temperature of 15 °C and maximum not exceeding 40 °C, Carvahlo et al. (2004) reportedly observed five stages of decomposition as described by Goff (2001) on the Hawaiian island while studying the succession patterns of necrophagous insects in Southeastern Brazil. The duration of decomposition lasted about forty days, and insect succession proceeded along the normal predictable pattern seen in other studies. Early and Goff (1986), working in the Manoa Valley on the island of Oahu, Hawaii, reported a 20 percent reduction in biomass on day 9. Richards and Goff (1997), also working on the island of Hawaii, reported similar reduction on day 11, and Payne (1965), working in South Carolina, reported a 10 percent biomass reduction by day 5. Decomposition could have been faster had it not been for the level of predation by ants and the premature migration of larvae due to excessive heat. Ants occupied the wound and fed on blood soon after the bait 1 carcass was exposed at the study site. They prevented the adult Calliphorids from ovipositing directly in the wound. These arthropods persisted throughout the entire study as predators Carvalho et al. (2004). Early and Goff (1986) working in Hawaii found that predation by ants prolongs the duration of decomposition. They pointed out that the effect was also observed by Stoker et al. (1995) in Texas and Houston (1987) in Brazil.

Carvalho et al. (2004), working in an open area with the carcass exposed to direct sunlight for 40 days, showed a predictable variety of feeding species associated with the carcass each day. But biomass removal was slow, and the dry stage was reached around day 36. In another study conducted by Hewadikaram and Goff (1991) on the island of Oahu, Hawaii, where

they compared the rate of decomposition to carcass sized and arthropod succession, they found no significant differences in the composition of the arthropods between the two different carcasses sizes. The dry stage was observed to last over an interval of 16 – 30+ days. They observed that the internal temperature of the smaller of two carcasses was more responsive to changes in ambient temperatures than the larger carcass in the fresh and bloat stages and were less responsive in the latter stages (decay stage) of decomposition, which peaked at 51 °C on day 6. On the contrary, they observed that ambient temperature had less influence on internal temperature of the larger carcass, which showed marginal increase up to day 4 and peaked on day 5, before declining during decay stage. In the current study, ambient temperatures had a significant influence on the growth and development of the *C. rufifaces* larvae, whose growth was severely retarded in the early stages of decomposition. Ground and surface temperature reached highs of 60 °C and 65 °C, respectively, and was the principal agent for the rapid removal of biomass (see discussion below). The dry stage was reached by day 5 (Fig. 14), and by day 8 (Fig. 18), approximately 95 percent of biomass was lost. For the other studies described above, biomass depletion was relatively slow compared to this study. It therefore becomes obvious that when the rates of biomass removal are slow, the stages are more definable, but when the rates are fast, as in this study, the stages are less definable. Payne (1965) summarized that the stages of decomposition are a continuous process without discrete stages, and that when arthropods are present on carrion, biomass removal can be rapid. Decomposition proceeded during this study as a linear function seemingly without a lag phase until the biomass was completely degraded. Some studies appear to have experienced periodic lag phases during decomposition, hence, the processes by which biomass is lost, proceed over a longer period of time. This in itself shows that the rate at which biomass is removed is not entirely dependent on arthropods' presence on the cadaver, but is also environmentally dependent. The rate at which decomposition proceeded

during this study showed that the decomposition process is in fact a continuous process, and temperature is the rate limiting factor. It was difficult to categorize a particular phase of decomposition prior to the remains stage.

Chin et al. (2007) observed that biomass removal was also rapid in Malaysia when the carcass under decomposition studies was exposed to direct sunlight and placed on a grass substrate. They found that the rate of biomass removal in that study was accomplished by a greater number of colonizing species, which was accomplished by a usual predictable sequence of arthropod colonization did not observed during this study. They observed that the adult fly populations were dominated by *C. megacephala*, and the immatures were dominated by larvae of *C. rufifaces*, similar to the observation made in this study. The general methodologies used in their study had some similarities to the current study; with the exception that the carcasses used in their study was a young pig that weighed 8.5 kg. They reported that on day 6, the carcass was still covered with maggots and a reasonable amount of adult *C. rufifaces* and *C. megacephala* were observed on the carcass; the carcass was also in an advanced state of decay and the ribs and skull were exposed. By day 8, the carcass was fully skeletonized. Under comparable temperature conditions, it is obvious that the carcass did not experience the effect of heat being directly exposed to sunlight as the current study did. Several observations made in the current study that were the result of heat generated from direct exposure to sunlight: at no time did larvae cover the carcass completely, and the adult fly population became sparse much earlier (the second day of active decay). Approximately 50% of larvae were either desiccated or migrated by day 4. Only a few larvae fed on the surface of the carcass, presumably due to unbearable carcass heat. Most of the larvae fed under the more rigid dorsal skin; therefore, the skin actually remained intact with bones until the body was fully mummified on day 8. They also discovered that pupae were formed close to the carcass, indicating that the decomposition environment was

not a threat to maggots' development as it was in this study. For this study, larvae migrated well over four to six feet away from the carcass environment to pupate under soil and thick eighteen inch grass where average soil temperature was 30.7 °C; none pupated at or near the carcass, which is likely due to the surrounding heat. The size of the carcass in the former study was small in comparison to this study; consequently, maggot mass would be smaller in density and dimensions to generate sufficient heat to elevate carcass temperature to the levels observed in Jamaica with a much bigger carcass. According to O' Flynn and Moorehouse (1979), small cadavers resemble that of a baby and would not generate the same temperature as larger ones; however, when dealing with temperatures at which maggots develop, size of the carcass must be taken into account.

Carcass size is a determining factor for the attraction of blow flies according to (Erzinclioglu 1996; Kuusela and Hanski 1982). Deno and Cothram (1975) pointed out that as the carcass size increases, the fly population also increases, and usually, it is the Calliphoridae larvae that are responsible for the rapid removal of biomass. The same observation was made in this study; the carcass used in this study weighed 23kg and was large enough to attract large numbers of adult flies, so great that if the larvae were evenly spread, the carcass would be inundated by larvae. This overcrowding of the carcass is reported in other studies such as Chin (2007); Goff (2001); Hewadikaram and Goff (1991), but the rate and pace of decomposition were significantly different in those studies. It, therefore, suggests that other factors must be considered as having an effect on the rate of decomposition. Based on the findings of this study, even when the carcass is large enough to attract a large population of flies, the context in which decomposition is taking place must be considered due the effect temperature can have on the rate of decomposition. The rapid rate at which decomposition occurred in this study is not entirely a result of blow fly larvae and other arthropods, but rather a secondary microbial decomposition

that is temperature dependent. Factors such as the thickness of the adipose tissue in the subcutaneous layer, pigmentation, and the substrate upon which the carcass is placed, are contributing factors to how much heat is retained/ dissipated from interstitial tissues where many reactions are in progress. All of these factors will determine the rate of how much heat is accumulated in the environment. The composition of the substrate at the interface is a very significant parameter during decomposition. Moisture produced by the carcass in conjunction with night, dew coupled with the heat from maggot masses, can initiate fermentation of dried grass or other organic materials under the maggot masses and carcass. Heat waves produced by fermentation will move by diffusion from one system to another until equilibrium is reached. The amount of heat retained on the carcass's body surface is also a function of the thickness (density) of the adipose tissue (back fat thickness) and temperature regime in that particular zone, as observed in this study. For example, on day 3, the carcass was covered with the oils of melting fatty tissues caused from direct exposure to sunlight. So much oil was produced, that it flowed through the cavities of the carcass carved out by feeding larvae, thereby, saturating the substrate under the maggot masses and carcass. The level of saturation was enough to carve out the shape of the carcass and penetrate the soil over three inches deep, long after decomposition was completed (Fig.19). Oils of fats are also capable of holding heat and have flammable properties that can elevate carcass temperature well above ambient temperatures. This increased the rate of putrefaction and liquification of tissues caused by thermophilic pathogens. Under such circumstances, larvae will consume their food source at a faster pace due to the abundance of ready-made metabolites. Bornemissza (1957) in Western Australia found that body fluids produced by carrion destroyed the flora and fauna under it and those arthropods are likely to be attracted to the area even after a year. The area where the carcass was placed continues to attract blow flies even after decomposition was completed. Muscidae and Piophilidae were the popular

flies at the area long after the blow flies had left. The vegetation under and around the carcass was also destroyed during the decomposition processes. Under normal circumstances, vegetation in an open environment will die if concealed from sunlight; however, when the carcass produces large quantities of oil from the degradation of fat, it is capable of suppressing the vegetative growth in close proximity to the carcass, and when the roots become saturated, the oil prevent water from reaching the root hairs resulting in the death of the plants.

5.2 Insect Interactions When Bait 1 Carcass Is Exposed in Direct Sunlight

The larvae of *Chrysomya* species are the principal invertebrate consumers of decomposing animal organic matter (Braack 1986). On the island of Hawaii, both species are the earliest at a carrion site and consistently have become the principal species used in the estimation of time since death (Early and Goff 1986; Tullis and Goff 1987). In most of the literature in which both *Chrysomya* species are encountered, *C. megacephala* are usually more abundant as adults in the earlier stages, but, after a few days, the larvae have never been recovered in the same proportion as observed in the adult population. It seems that both the adult and larvae of *C. rufifaces* appear later and may delay activities such as oviposition and larval development to gain a predatory feeding advantage over other species. As decomposition progresses, larvae of *C. rufifaces* become the dominant feeders on both carrion and other larvae. It appears that the early colonizing *C. megacephala* are their preferred or common prey when both species are encountered on the same carcass (Goff 2001). The largest population of *C. rufifaces* was concentrated around the wound, which is the only area where *C. megacephala* larvae were recovered. Most likely, an intense predatory selection for *C. megacephala* occurred as opposed to the larvae of *C. macellaria*. Goodbrod and Goff (1990) under laboratory conditions made the observation that when *C. megacephala* and *C. rufifaces* are together, the larvae of *C. megacephala* are an alternate food source, and, when other food sources are scarce, the larvae of

C. rufifaces will switch from being predator to cannibal. All of the characteristics of *C. rufifaces* were observed except for cannibalism.

During the first larval collection on day 3, the three larval species recovered from the carcass of bait 1 showed marked variations in both density and stages of development (Table 5). Larvae of *C. rufifaces* dominated the carcass, appearing at every area on the carcass as early and late 3rd instars. Two high density maggot masses were apparent on the carcass; the larger one was located at the ventral aspect and the smaller one at the posterior end of the carcass; the larger maggot mass was composed purely of 1st, 2nd, and 3rd instars *C. rufifaces* larvae. Those recovered at the posterior end consisted purely of 2nd and 3rd instars *C. rufifaces* larvae, with the majority being 3rd instars. The largest *C. rufifaces* larvae were recovered at the wound in association with *C. megacephala*. The increase in body size is likely due to the predation when compared to the other *C. rufifaces* larvae found at other areas of the carcass. Larvae of *C. macellaria* were collected on day 3 and 4, while the larvae of *C. megacephala* were collected only on day 3, which suggested that they were forced to other areas or became the secondary food source for *C. rufifaces* larvae. The collection of *C. megacephala* larvae only from the wound is an indication of the intense pressure from *C. rufifaces*, which led to the recovery of fifteen of the larvae (Table 5). Wells and Greenberg (1992) made these observations while studying the species under laboratory conditions. Goodbrod and Goff (1990) studied the characteristic behaviors of both *C. rufifaces* and *C. megacephala* in pure cultures; they found that *C. rufifaces* preyed on the 2nd to late 3rd instar larvae of *C. megacephala*. This understanding explained why the larvae of *C. rufifaces* found at the wound were larger and more uniform in body size as opposed to the less developed ones recovered at other areas of the carcass. A point worth making is that the early disappearance of *C. megacephala* and *C. macellaria* by the activities of *C. rufifaces* is not purely a function of just the natural predatory tendency of *C.*

rufifaces on other larvae. It is also temperature related. With carcass surface temperature elevating to 30 °C over ambient temperatures, the feeding environment would become unbearable and hasten the predatory tendencies for an alternate available food source. This explains why only mature 3rd instars *C. rufifaces* larvae were seen traveling in a cyclic pattern around the perimeter of the carcass; this behavioral pattern gives the appearance as if when they moult into 3rd instars, they migrate from the maggot mass to go in search of other flesh-like food sources. They seem not to feed at any one area for long after reaching the 3rd instar stage.

Since *C. macellaria* was the most developed of the three larval species, this supported the fact that they were the first Calliphorids to arrive and deposit eggs on the carcass, which afforded them the opportunity to develop at a faster rate with less competition from the other two species, especially, *C. rufifaces*. During the second larval collection on day 4, the dominant larval specimens continued to be *C. rufifaces* with only a small number of *C. macellaria* larvae found at the head. Since *C. macellaria* larvae were associated with only the head cavity of the carcass, one might conclude that the roaming predatory 3rd instar larvae of *C. rufifaces* that were actively feeding on *C. megacephala* would have required more time to reach the larvae of *C. macellaria*, thereby affording these larvae an advantage that the larvae of *C. megacephala* did not have. The observation was made that *C. macellaria* deposited eggs only in the cavities of the mouth and head area of the carcass as opposed to *C. megacephala* that showed preference for depositing eggs at the wound and deep under the interface of carcass and ground (Goff 2001). *C. megacephala* were prevented from depositing eggs directly into the perimortem wound by ants that occupied the area very soon after the carcass was placed at the study site. However, since the only specimens of *C. megacephala* collected were in the vicinity of the wound, the inference can be drawn that they made an attempt to deposit eggs as close as possible to the wound. *Chrysomya rufifaces*, on the other hand deposited their eggs along the perimeter of the carcass

over a wider area. None of the species deposited eggs on the surface of the carcass, suggesting that the carcass surface temperature was suboptimal for egg deposition. It appears that the adult blow flies are able to detect the condition of the surfaces upon which they deposit their eggs. Other studies noted that *C. megacephala* and *C. rufifacies* exhibit variation as to which area of the carcass they inhabit during the decay stages and that the larvae of *C. rufifacies* remain in loose masses on the surface (Goff et al. 1986; Tullis and Goff 1987). The latter findings may have application to this study as seen with the preferential colonization and occupancy of *C. macellaria* in the head cavity, but may not generally apply to the suboptimal conditions encountered in this study. The characteristic behavior of flies has to be associated with the conditions of the area. The behavioral pattern will likely to be environmentally influenced from one region to the next. Migratory species especially is likely to show varying degree of adaptability depending on available food source, the state of the physical environment and the faunal distribution in which they find themselves.

Sarcophagid species made many attempts to colonize the carcass, but were prevented from doing so, probably due to the intensity of heat on the carcass's body surface. They were observed hanging on tree branches in the shaded area not too far from the carcass. Only Calliphorid larvae were recovered from the body. Apart from the overwhelming presence of the Dipteran larvae, the usual succession pattern involving other arthropods was lacking during the latter stages of decomposition. The larger beetles such as the Dermestids, Staphylinids, and Histereidae that are normally associated with carrion were not observed in this study, and the rate of decomposition was determined mainly by blow fly larvae and putrefaction. Of the four families of beetles collected at the site, only the Cleridae and Tenebrionidae were recovered directly from the carcass residue, the others were recovered on vegetation close to the carcass. Beetles collected from bait 1 were in the size range of 7 mm to 10 mm.

5.3 The Thermal Dynamics of Maggot Mass

Deonier (1940) studying how temperatures are generated within a carcass observed that in certain parts of the carcasses, the temperature was 70 °F above ambient temperature, and more than 50 °F above the ambient temperature in the maggots' mass. He argued that the elevation of temperature found in the carcasses was primarily due the result of heat generated by the blow fly larvae, and partially from the heat absorb from the sun. In this study, the sun was the rate limiting factor on decomposition. Heat absorbed from the sun, elevated the carcass surface temperature to 65 °C which was not influenced by the maggot mass that was concentrated outside of the carcass. Maggot mass on the hand generated a temperature of 49 °C. These temperature ranges proved lethal to a large proportion of the larvae in the early stages of development. The radiation from the sun was the main reason for elevated carcass temperature. As it relates to maggot mass temperature, a sharp rise was observed in the first several hours after the onset of active decomposition; larvae of *C. rufifaces* were still moulting from 1st and 2nd to 3rd instars, most were early 3rd instars larvae. The greatest thermal contribution seems to have been generated between the transitions from 2nd to 3rd instar (Catts 1990). On the other hand, the 1st instars appear to require the least amount of heat in the early stages of life to develop internal organs and systems. The maggot mass temperature did not rise suddenly; it did so gradually until about the latter part of the 1st instar stage, and then gradually increases with increasing larval size. At some point, temperature becomes stable before gradually declining towards the post feeding stages. Thermal energy generated in the maggot mass influences development, although such energy is not generated immediately after eclosion from the eggs (Goff 2001). A greater number of 1st and 2nd instars were recovered from the center of the maggot mass than for the 3rd instars collected at the outer band. These observations may or may not be applicable to other non predatory species, since different species have their unique way of behaving.

However, while larvae of *C. megacephala* were being reared, the larvae formed a very small maggot mass numbering about 150 + larvae. For the first two days, maggot mass showed a marginal increase in temperature over ambient temperature which was more pronounced during the night; as the larvae developed, the temperature fell to ambient temperature prior to the onset of the post-feeding stage. Although this observation might not accurately reflect the same pattern of temperature change in a larger maggot mass, it shows 1st instars rely on ambient temperature to reach the stage required for them to begin produce sufficient metabolic heat to sustain optimal growth rate and body size. Optimization of growth is rather a function of larval density, ambient temperature and food source. While rearing *C. megacephala* and *C. rufifaces* larvae, Goodbrod and Goff (1990) found that when the population density was low, change in temperature produced by the larvae was marginal.

As mentioned earlier, the carcass was literally fried and the adipose tissue produced a large quantity of oil, which seemed to play the major role in the desiccation of larvae at the base of the maggot mass (Fig. 13). Oil produced by the body found its way into the maggot mass. In such a case, the larvae would be unable to respire and that build up of heat may be the source of mortality observed on day 3. About 50 % of the larval population was desiccated during the early stage of active decay on day 3 when the larger maggot mass temperature was 45 °C at the epicenter and 51 °C at the base. Surface temperature was 65 °C, and internal visceral temperature was 60 °C. Although larval growth is temperature dependent (Byrd and Castner 2001; Goff 2001; Greenberg 1985; Payne 1965; Smith 1986), when it exceeds the optimum level required for growth, the rate of larval retardation appears to be proportional to the incremental growth rate associated with normal growth, considering other variables optimum. Studies done by Dillon and Anderson (1997) in British Columbia, reported that when a carcass is exposed to direct sunlight, the carcass is mummified rapidly due to high ambient temperatures, which results

in the mass migration of undersized 2nd and 3rd instar Calliphorid larvae in search of other food sources. The same scenario was observed on the first day of maggot mass formation. However, the larvae in this study did not migrate due to food shortage (food was available); instead, they migrated due to the unbearable temperatures generated within the maggot mass and the carcass. Consequently, blow fly larvae were observed heading back in the direction of the carcass late evening when ambient temperature dropped. Most of the migrating larvae were those of *C. rufifaces*. They were the only larvae that had established a distinctive maggot mass. Larvae of *C. rufifaces* showed an increase in growth rate as decomposition progressed (Table 4), even though desiccated viscera were still above ambient temperature. The concept behind the increase in growth rate of the *C. rufifaces* larvae on day 4 and 5 is that, as the larvae grow older, they tend to increase the feeding range, thereby, spreading the maggot mass. At the same time they optimize the surrounding temperatures by decreasing larval density per unit area while increasing pore space for heat dissipation.

Gruner et al. (2007) discovered that ambient temperature has no effect on maggot mass when it exceeds the size of a golf ball. This observation is very consistent with principal observations made about the maggot mass in this study: maggot mass temperature remained at an average of 47 °C throughout the active decay period, which is sufficient to believe that ambient temperature had no influence on maggot mass temperature. The circumstances under which maggot mass generates and retains heat are a dynamic process as illustrated in this study. According to the principle of heat transfer, the amount of heat required to raise the temperature of a system is proportional to the amount of material in that system, and the change in the internal energy is equal to the heat added to the system minus the work done within the system. The latter statement is represented by the formulae $\Delta U = Q - W$, where ΔU is the change in internal energy, Q is the heated added, and W is the work done. In addition, if the dynamics of

maggot mass is to be determined, it is important to know how to define Q (heat added to the system) in terms of W relative to larval density and maggot mass dimensions at various temperature regimes. Knowing Q could have a definite impact on the qualitative and quantitative analysis of the thermodynamics of maggot mass temperature as it relates to blow flies' developmental rate. Considering such an argument, larval developmental data should take into account the relationship between temperature and larval population density when estimating postmortem intervals (Goodbrod and Goff 1990).

The surface upon which a carcass is exposed is likely to determine the extent of how heat is exchanged or dissipated by convection at the interface. The soil type was determined to be Silty Clay Loam with the aid of a Soil Textural Triangle. The soil texture has a relative proportion of 30 % Clay, 10 % sand, and 60 % silt. Clay has fine pores and the ability to hold moisture longer than other soil particles with wider pore space. Moisture, on the other hand, has an effect on the thermal properties of soil (Oke 1987) which is likely to hold heat, especially at high ambient temperatures. Because the clay pores are fine, heat developed within the soil is not easily dissipated by osmosis or diffusion; hence, the surrounding temperatures easily can be raised under direct radiation from the sun. This creates a heated zone at the interface of maggot mass and carcass. What is clear is that when the maggot mass accumulates temperatures above ambient temperatures, ambient temperature do not play any role in the development of larvae within the mass.

5.4 Larvae Reaction to High Temperature Environment

The carcass was positioned in a north-south direction. The sun had a direct impact on the behavior exhibited by the maggots on the carcass. The sun struck the carcass from the east as early as 6:30 AM daily until it reached its highest temperature limit somewhere around mid-

afternoon. During this time of the day, the majority of the maggots sought refuge on the west side (in this case the ventral side) of the carcass which explained the unusual distribution of larvae about the carcass (Fig.11). The usual head down type of decomposition did not proceed as often observed in other studies. Although the head was the first to be skeletonized, decomposition proceeded from the ventral aspect of the carcass toward the head. Larvae then used the cranial cavity as a gateway to travel around the perimeter of the carcass while feeding from the outside to the inside, rather than concentrating in the gut. The head was only skeletonized first because it has less tissue density. However, the bulk of growing larvae in the maggot mass was concentrated outside the ventral side (gut) of the carcass (Fig. 12) that had most of the body tissues and fluids. The best description to describe the larval feeding pattern is to picture using a knife to cut the carcass in two halves along the anterior – posterior length. Larvae were more evenly distributed on the carcass without forming a concentrated maggot mass.

As the evening got cooler, greater activities were observed on the dorsal side though not significant due to less biomass. Interestingly, maggots feeding from the head move along the dorsal aspect of the carcass in single file during the day, presumably to shed any metabolic heat generated. The latter explained why the majority of feeding 3rd instar maggots were consistently lumped on the ventral side of the carcass.

5.5 Phase 2 Study with Bait 2 (Goat Head) and Bait 3 (Tilapia Fresh Water Fish)

Phase 2 of the study consisted of two sets of larval rearing, those collected on bait 2 (goat head) and those collected on bait 3 (fish) at the Government Forensic Laboratory site in Kingston and St. Andrew and reared in Spanish Town, St. Catherine. Larvae collected on bait 2 were transferred to liver for rearing. *C. megacephala* larvae were reared in mixed and pure

populations on beef liver and fish, respectively. Both specimens were reared in an outdoor environment at an altitude of 14 m above sea level and at an average temperature of 26 ± 0.5 °C. The relative humidity was 63.23 ± 1.6 with a high of 80% relative humidity when the temperature was at 20.8 °C and a low of 39 % relative humidity at 35 °C. Both containers were sheltered under a zinc awning with dimensions of six feet by three feet to prevent rain and direct impact from the sun's radiation. Ambient temperature conditions were constant during the period of rearing. Bait 2 had a larval population of about 100 specimens, all of which were reared in a small eight-inch by eight-inch plastic container on 112 g of fresh beef liver and one half inch of sandy loam soil for pupariation. The population density was one larvae /1.12g of liver of mixed larval population of *L. lucigerens* and *C. megacephala* in the ratio of 1:3, respectively. There was no sign of predation or cannibalism between these species. Larvae of *C. megacephala* were larger and more robust than those of *L. lucigerens*. During the migratory phase, *C. megacephala* larvae traveled away from the food source and buried themselves during pupariation as opposed to *L. lucigerens* that remained on the surface of the substrate close to and under the hardened liver during pupariation. This may not be an indication of *L. lucigerens* actual behavior in nature. It might be a result of them being held outside their natural habitat in the container. It might also have been an evasive behavior to avoid invading the space of the more robust larvae of *C. megacephala*. From all indications, the observations might very well be their natural behavior during pupariation. Goff (2001) noted that some species will migrate and burrow underground to pupate, while some remain at close proximity to the food source during pupation. The only abnormal characteristic observed was that some of the *L. lucigerens* adults emerged under the leftover food source and remained there until emergence. There were only about two puparia that did not emerge, which could be associated with their natural fertility rate or fecundity. After the emergence of both species, dried liver was still remaining which amounted

to about a third of the original weight. There were no signs of mortality between the two species. One hundred percent of *C. megacephala* larvae emerged in eight plus days and ninety eight percent of *L. lucigerens* adults emerged in sixteen plus days from the time they were collected as early 1st instar from bait 2, which was previously exposed for approximately thirty six hours. An error was made when the decision was made not to collect larvae from their rearing unit of *C. megacephala* and *L. lucigerens*.

The second set of larval specimens collected on fish (bait 3) was reared as pure *C. megacephala* larvae with a population density of 2.98g/larva. The developmental data gathered for this species can be compared with the results obtained by Wells and Kurahashi (1994) for the species reared in Japan and to the results obtained by Velez and Wolf (2008), who studied the same species in Colombia. When the three developmental data were compared (Table 9) , there were marginal variations among time spent in the stages described by Wells and Kurahashi (1994) as opposed to the larger margin of variation observed by Velez and Wolf (2008). However, consideration has to be given to the differential environmental conditions at which the species were reared. Conditions such as field versus laboratory, larval density, constant temperature versus fluctuating (diurnal) temperature, and day length also need to be considered. It is obvious that in a laboratory setting, Wells and Kurahashi (1994) had less variation in temperature than that of Velez and Wolf (2008) whose experimental data were obtained from a semicontrolled field condition at an elevation of 1450 m above sea level with average rainfall of 1409 mm. The variation for those species reared along the coastline that borders the Caribbean showed less variations as opposed to those species reared at higher altitude. To elaborate on the latter data set, it is obvious that the area is fairly mountainous with a high degree of fluctuating temperatures; therefore, larvae may respond to temperatures differently. This is evident in the lower and upper limit of the confidence intervals presented in the data they presented. Wells and

Kurahishi (1994) described the first two moults as a synchronized event and variations in subsequent moults of pupariation and emergence. The emergence of all the adults from bait 3 was personally observed, and occurred over a one half hour interval. The most recognizable variation was observed during the postfeeding stages. This stage appeared to be less predictable than the other stages. Larval behavior is not always what it appears to be, consequently, the indicator used for pinpointing the onset of the post feeding stages was determined when the larvae began to travel around the circumference of the container or cage. No rain fell during the period of study in Jamaica, except for slight drizzles; consequently, not much variation was observed in ambient temperatures or humidity. This might not have been the case in Velez and Wolf (2008) where differential temperature zones associated with altitudinal changes can impact the rate of larval development. Although the study by Wells and Kurahishi (1994) was done in a laboratory environment at constant temperature, the effect of temperature on the larvae development may be analogous to both studies. Since the temperatures in Jamaica are fairly stable, the variation seen in the time adults emerged might not be temperature related. Larval density, rearing environment, food type, etc., and other uncontrolled climatic variables are likely to play a key role in some of the marginal variations observed among the data sets being compared, especially, at the times when the adults emerged. Goodbrod and Goff (1990) studied the relationship between larval density and food quantity of *C. megacephala*, reared in cultures at 23.5 ± 0.5 °C and a relative humidity of $65 \pm 75\%$. They found that the first larval stage that had a density of 1 and 2 larvae per gram of food, did not vary as a function of density, and had a duration of 18 to 24 hours. The second instar durational range was 24 hours in cultures of 20 and 40 larvae per gram as opposed to 36-48 hours in cultures with 1 -2 larvae per gram. The third instar durational range was 71 hours in cultures of 40 larvae per gram to 108 hours in a culture of 2 larvae per gram. Both mixed and pure populations of *C. megacephala* larvae were consistent

with both studies. The developmental rate obtained by Wells and Kurahashi (1994) and the durational ranges and density illustrated by Goodbrod and Goff (1990). The observation was made that eggs appeared to hatch during the night to early mornings when temperatures were low as opposed to the adults that emerged at variable temperature ranges, but, not during dark periods. This study provides some clues about the effect of temperature on larvae development. Analyzing data obtained from the rearing of *C. megacephala* and the effect that temperature had on the developmental rate of *C. rufifacies* are compared as it relates to the effect of temperature effect on larvae, one can deduced that when the environmental temperature exceeds the temperature zone which is necessary to optimize growth, the surplus heat suppresses growth, as it would if heat was deficient. The inference therefore is that the temperature required for the larvae to optimize growth is absolute, and, when the temperature to which the larvae are subjected is above or below the absolute temperature range, the larvae will either experience developmental retardation, diapause, or death. Wells and Kurahashi (1994) remarked that blow fly larvae are likely to delay pupation if conditions are suboptimal. This assertion seems less applicable in an event where temperature is the cause of a suboptimal condition. Since larvae can not regulate their own body temperature (poikilothermic organisms), when temperature is above the tolerable threshold, as seen in this study, the larvae of entered the pupariation stage sooner than normal to avoid being desiccated.

Wells and Kurahashi (1994) calculated the degree days by using a minimum developmental threshold temperature of 10 °C, unlike the method used in this study. Along the coastal area where estimation of postmortem interval would most likely be required, Jamaica maintains a yearly average temperature of approximately 30 °C, therefore, degree days were calculated as the product of average daily temperatures and time spent by the larvae in each stage of development, without the consideration of a minimum developmental threshold.

CHAPTER 6: CONCLUSION

This study is the first of its kind to be done in Jamaica. As a result, some of the observations made are likely to vary during the course of multiple studies. Several observations have been made during the course of this research: 1. *C. megacephala* is the most abundant species in Kingston and St. Andrew, which indicates it may have utility in the estimation of postmortem interval. 2. The developmental stages are very consistent throughout the rearing of both larvae and eggs. 3. The predatory impact that *C. rufifaces* has on *C. megacephala* when they are together on cadavers. *Chrysomya rufifaces* and *Cochliomyia macellaria* were collected in large numbers from a large bait 1 carcass, but were not recovered on the smaller bait 2 food sources in February. The variations in size and type of the baits may not accurately determine the true relative distribution of Calliphorids. However, useful information such as the relative abundance of some species that will colonize both small and large baits was gathered. For example, *C. megacephala* was found to colonize both bait sizes and demonstrated their omnipresence and ability to survive on all food types.

When the results of the current study are viewed in relation to the factors that are involved in the decomposition processes, it is evident that the rate at which biomass degradation occurs in a carcass is not entirely subjected to insect colonization. Other factors, such as temperature and humidity, can have a significant impact on the decomposition rate, arthropod succession pattern, and larval development, which is capable of skewing the estimation of postmortem interval. For example, if the carcass of bait 1 was visited after the second collection (day 4), the chances are that, the larvae of *C. rufifaces* could have been easily thought of as the first species to arrive at the carcass. Secondary factors such as carcass adipose (fat) composition, pigmentation, composition of ground surface, rate of decomposition by microscopic pathogens,

carcass orientation in terms of sunrise and sundown, and moisture retention capacity of environmental materials must be accounted for as part of the composite processes that affect the relative distribution of temperature. Heat was the principal determining factor throughout the study that resulted in severely burned and undersized larvae, and rapid biomass loss. One important consideration that is not mentioned in the literature is the effect of pigmentation and the thickness of the adipose tissue of the carcasses used as study material. Both variables are worth analyzing for the role that they may play in the heat retention capacity (HRC) of a carcass. A relationship may exist as it relates to the rate of biomass loss and arthropods' attraction.

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APPENDIX

STATISTICAL ANALYSIS FOR LARVAE REARED ON BAIT 3 IN SPANISH TOWN ST. CATHERINE FROM FEBRUARY 15-24, 2008

Larvae of <i>Chrysomya megacephala</i> (Feb. 15-24, 2008) from bait 3				
No. larvae	L1	L2	L3	PF
1	2.9	8.5	13.5	14.5
2	3	7.9	13.2	14
3	2.9	8.2	12.9	13.8
4	3	8.1	13	13.9
5	2.8	8.5	13.4	14.2
6	3	8	13.5	14.5
7	2.7	8.4	12.8	14.5
8	3.1	8.5	13	13.8
9	2.9	8.3	13.5	14.5
10	3	8.5	13.5	14.5
Average	2.93	8.29	13.23	14.22
Stdev	0.11	0.216564	0.268514	0.299333
Sqrt(n)	3.162278	3.162278	3.162278	3.162278
Std err	0.034785	0.068177	0.084912	0.094657
p value	0.068177	0.134225	0.166424	0.185525

VITA

Wayne Anthony Cranston was born in Kingston, Jamaica. His post-secondary education started at the Passley Garden's Agricultural School in Portland in 1981. He was later transferred to the Elim Agricultural School in St. Elizabeth, where he graduated in 1983. He then took a job with the Eastern Banana Estate as a quality control supervisor. In 1987, he attended the Jamaica Constabulary Force Police Training Academy where he spent one year as a recruit. He went on to spend seventeen years with the organization. During his time with the Jamaica Constabulary Force (JCF), he attended the Jamaica College of Agriculture for three years, where he earned an Associates of Science degree in agriculture. He worked at the forensic laboratory during the latter part of his tenure with the JCF, before moving to the United States to further his education at Louisiana State University Agricultural and Mechanical College. He graduated with a Bachelor of Science degree in animal science technology in May, 2000, and a second Bachelor of Science degree in environmental management systems with a minor in chemistry in August, 2001. He hopes to further his education in the near future.