

Insects on Decomposing Pigs and their Forensic Importance at a University in Nigeria

Maduamaka Cyriacus Abajue

**Department of Animal and Environmental Biology, Faculty of Science
University of Port Harcourt, Nigeria
E-mail: maduamaka.abajue@uniport.edu.ng**

Abstract

The use of insects to estimate post-mortem interval (PMI) of cadavers by forensic entomologists, is an innovative science enshrined in the medico-criminal system of the developed world. The science is dependent on the biology of insect species that are found in decomposing cadavers. Thus, insect species that are found on decomposing pigs as model to human cadaver, was studied during the rainy and dry seasons of 2021 in a University forest in Nigeria. The decomposition stages (fresh through skeletonization) of the pig cadavers were monitored, and insects found on them were collected with a sweep net, visually observed, and handpicked. They were identified morphologically using insect identification keys with the aid of a dissecting microscope. The tissues of the pig cadavers decomposed completely in 14 and 10 days, respectively, for dry and rainy seasons. Twenty-seven insect species belonging to the orders; Diptera, Coleoptera, and Hymenoptera were identified. The dipterans consist of the families Calliphoridae, Muscidae, Ulidiidae, and Stratiomyidae. The coleopteran families are the Carabidae, Chrysomelidae, Cleridae, Dermestidae, Histeridae, Scarabaeidae, and Staphylinidae, while Formicidae is the only family for the hymenopteran. The dipteran species were the first group to visit the cadavers during fresh, bloated, and wet decay stages while the coleopterans visited during wet and dry decay stages thus, creating a succession pattern on the cadavers. The insect species that aided the cadaver decomposition at different stages are discussed as forensic indicators, which could be useful as PMI estimators of the study area.

Keywords: Decomposition stages, Forensic indicators, Insect species, Pig cadaver, Post mortem interval

Introduction

Insects found on human cadavers are useful indicators for ascertaining post mortem interval (PMI), which is vital in forensic and criminal investigation relating to questionable deaths (Cordeiroa et al., 2019). The knowledge of using insects to estimate PMI is rooted in the information extracted from the community composition, guild structure, and community dynamics of insects associated with cadavers (Villet, 2011) and is predicated upon the time of colonization (TOC) or period of insects activity (PIA) (Byrd & Castner, 2010). Cadaver decomposition is a successive process mostly impacted by abiotic (temperature, relative humidity, rainfall, and others) and biotic (insects and related arthropods, micro-organisms, and vertebrates) interactions (Bugelli et al., 2018). When considering cadavers as functional ecosystem, the guild structure and community dynamics of insects associated with it would be useful as forensic indicators for investigating a questionable death (Villet, 2011).

Many taxa of insects are on cadavers at different stages of decomposition but indicators of forensic importance are usually within the orders, Diptera and Coleoptera, while the orders Hymenoptera and others depict local entomofauna (Smith, 1986; Amendt et al., 2004; Benecke, 2004; Gennard, 2007; Byrd & Castner, 2010). Empirical studies of insects found on cadavers (pig, rabbit, rat, and others) as surrogate human models due to the ethical limitations of using human cadavers are paramount in forensic entomology (Dekeirsschieter et al., 2013).

The use of insects to ascertain the culprit who murdered a man dated back in China many years ago (Wang et al., 2021) but has recently gained a better and acceptable prominence more in America and Europe (Sandford, 2017; Lutz et al., 2021) than other continents. In Nigeria, the use of insects in the investigation of a questionable death is yet to be reported due to the dearth of knowledge of insects of forensic importance. Nevertheless, few studies of insects found on different surrogate animal models have been documented (Okiwelu et al., 2008; Ekanem & Dike 2010; Ekrakene & Iloba, 2011; Abajue et al., 2013; Abajue & Ewuim, 2016; Abajue & Ewuim 2020) in anticipation to extrapolating the findings to a real case when such service is legally required. However, a pig

cadaver as a human model has not been studied in this present study area in relation to forensic entomology. In this study, therefore, insects found on pig cadavers that were deposited in a University forest during the dry and rainy seasons of 2021 were collected, indicator species of forensic importance that would be used to estimate the PMI of cadavers of the study area and its environs were ascertained.

Materials and Methods

Study area

The study was carried out in a secondary forest during the dry (February-April) and rainy (June-August) seasons of 2021 at the University of Port Harcourt, Nigeria located at 4°54'14"N and 6°55'7"E. The vegetation (*Chromolaena odorata* (Linn.), *Aspilia africana* (Pers.), *Abelmoschus manihot* (Linn.), *Andropogon tectorum* (Schaumach. and Thorn.), *Napoleona imperialis* (P.Beauv.), and *Elaeis guineensis* (Jacq.)) within the experimental site was identified by a Plant Ecologist at the Department of Forestry and Wildlife, University of Port Harcourt, Nigeria, which characterize a humid tropical rainforest (NDES, 2001). The vegetation is dominated by *Aspilia africana*, followed by *Andropogon tectorum* while *Napoleona imperialis* and *Elaeis guineensis* were the least dominant plant species.

Animal model used and experimental design

Four healthy male domestic pigs (*Sus scrofa* Linn.) of about ten weeks old with a mean weight of 22.3 ± 1.9 kg purchased from a piggery were used for the study. Pig was chosen as a human model because of its morphology, which is similar to human and both cadavers have attracted the same type of insect fauna that are useful in forensic entomology (Tomberlin et al., 2012). The pig cadavers were individually wrapped in polyethylene bags and transported to the study site immediately after death. Each pig cadaver was placed on a 16 x 8 cm² sack on the ground in a delineated 5 m² land at about 10.56 hours. Insects (adults and larvae)

collection was limited to the mat. Each pig cadaver was covered with a cage made of wire gauze to protect them against vertebrate scavengers, but insects were allowed to access them. The same experimental procedures and processes were repeated for the dry season study.

Daily records of temperature, humidity, and collection of insects on the pig cadavers

Ambient temperature and relative humidity in the forest were obtained with a portable digital thermo-hygrometer (Mextech TM-1, model). It was hung on a 4 ft. pole in the forest under a canopy shade, and 3 readings per data (temperature and relative humidity) were obtained after 15 minutes. Remarkable decomposition changes on the cadaver and insects that visited them were noted and recorded from the time of deposition (10.56 hours) to 19.00 hours. From the second day through the sixteenth day, notes on decomposition changes and insects that arrived on the cadavers continued but started in the morning from 6.00-8.00 hours earlier than the first day and in the evening from 16-18 hours.

Insects found on the cadavers or the mats were collected with either sweep net, forceps, or handpicked (glove hands) depending on the insect type. Insect larvae that were seen on the 2nd day were collected on the 4th day through the 16th day. The ones with similar morphometrics were grouped and reared in separate containers (9.0 cm depth and 6.5 cm width) until adult emergence (Abajue et al., 2017). All the adult insects that emanated from the pig cadavers were taxonomically sorted by using the insects of Nigeria checklist and bibliography (Medler, 1980). Flies and beetles were further diagnosed with a manual provided by (Smith, 1986) and the keys for blow flies (Lutz et al., 2018). Samples of the insect taxon were sent to a taxonomist at the Insect Museum, Ahmadu Bello University Zaria, Nigeria, for authentication, while the remaining ones were permanently preserved in specimen bottles containing Kahle's solution.

Ethical Considerations

The pigs selected for the experiment, were secluded from other swine in the farm for three hours without food but they were served water. A licensed veterinarian euthanized the pigs in agreement with the Animal Ethics Committee of veterinary animal care, welfare and medical research (AVMA, 2014; SAMWEC, 2014). They were later sacrificed by mimicking strangulation kind of death (Abajue & Ewuim, 2020; Abajue et al., 2021). The process was approved by the University of Port Harcourt Research Ethics Commission.

Results and Discussion

The mean temperature and relative humidity of the study area were 30.8 °C and 41.8 % for dry season, and 27.2 °C and 81.9 % for rainy season. This difference probably impacted the tissues of the pig cadavers to decompose completely in 14 days during the dry season, and in 10 days during the rainy season. This observation is similar to the reports of Galloway et al. (1989), which states that cadavers left outdoors tend to decompose faster when the temperature is below 38.0 °C, and that slight variations in temperature within a habitat can lead to variable decomposition rates. Thus, it can be inferred that as ambient temperature at the forest reached 30.8 °C during the dry season, as the ambient moisture content decreased, which probably led to a slightly longer decomposition duration of the cadavers. Comparably, the duration of decomposition during the rainy season was slightly faster, probably because of the ambient temperature at 27.2 °C and increased ambient moisture content.

In the study, three insect orders Coleoptera, Diptera, and Hymenoptera consisting of twelve families and twenty-seven species were collected on the pig cadavers. The coleopterans were more in numbers with seven families and twelve species respectively. It was followed by the dipterans with four families and eleven species, while the hymenopteran has one family and four species, as shown in Table 1. The twenty-seven insect species were collected during the rainy season while

nineteen species were collected during the dry season. Empirically, using insects to estimate PMI by forensic entomologists is primarily centred on the development of insects and or their succession sequence on cadavers (Tomberlin et al., 2012). Therefore, in analyzing the insect activities that underly the ecological characteristics of a cadaver community at the family level, would depict a clear correlation that ecological surrogates of insects are globally distributed. Hence, the ecological structure of cadaver communities in terrestrial habitats irrespective of the geographic location would fit into the same insect community guilds as submitted by (Villet, 2011).

Most of the insect species in the orders Coleoptera and Diptera identified in this study, are in consonance with the established cadaveric insect guilds (necrophagous, predators, omnivorous and adventives) documented by (Smith, 1986). The necrophagous insect species of this study include the *C. albiceps*, *C. chloropyga*, *C. regalis*, *C. africana*, and *H. illucens* that are obligate soft tissues feeders while the *N. ruficollis*, *N. rufipes*, and *D. frischii* are dry and keratinize tissues eaters. The predators identified in the study are the *Hister monitor*, *Hister castanipes*, *Hister* sp., *Philonthus* sp., *Staphylinus* sp. and *Camponotus perrisi*. Interestingly, a *N. rufipes* was spotted killing the first larval instar of *D. frischii*. The observation was not common, hence, it could not be assigned a predatory role in agreement with (Van Learhoven, 2010) who stated that such facultative predation should be recognized as omnivore. The omnivore of this study was only *Pheidole* sp., which was also reported by Ekanem & Dike (2010) and Dupont et al. (2011). The gregarious nature of the *Pheidole* sp., on the cadavers may likely be a disruptive trait capable of affecting the activities of the necrophagous insects. The adventives are mainly the arthropod relatives (isopods, millepeds, arachnids) which according to (Smith, 1986), are biogeographic indicators of crime scenes and are also useful in forensic entomology.

Interestingly, *Drimostoma puntulatum* (Tsch.), *Buphonella* sp., *Corynodes compressicornis* (Fab.), *Musca confusciata* (Speiser), *Musca sorbens* (Weid.), *Pyrellia scientillans* (Bigot), *Sagaricera analis* (Macq.), and *Sagaricera excellens* (Brun) are the species that were not collected on the cadavers during the dry season (Table 1).

Based on the ecological niche of the insect species, it was noted that the necrophagous insects (*C. albiceps*, *C. chloropyga*, *C. regalis*, *C. africana*, *H. illucens*, *N. ruficolis*, *N. rufipes*, and *D. frischii*) are the useful forensic indicators of the study area because of their predictable occurrence, which is in line with the reports of (Benbow et al., 2013) that necrophagous insect species have been recognized as principal decomposers of vertebrate cadaver. There was a unique pattern of change of the insects on the cadaver, starting with the calliphorids as the colonizers, which stimulated the arrival of their predators. As the tissues desiccated after wet decay, the fly larvae (calliphorids) dispersed to the nearby debris for pupation, thus making the beetles numerically dominant on the cadavers until the cadavers remained only the bones. The earlier dispersal of the calliphorids was a result of the nutritious diet contained in the soft tissues of the cadaver against the less nutritious diet in the dry tissues. The above submission is based on the report of (Villet, 2011) who stated that dietary differences between the necrophagous insects affect the rate at which their offspring grow on cadavers.

The larvae of the calliphorids dispersed from the cadaver earlier than the clerids and dermestids that progressively dominated the cadavers thus, created a succession sequence in the cadaver ecosystem. The occurrence of the calliphorids on the cadavers during the fresh, bloated and wet decay stages was similar to the observation of (El-Gawad, et al., 2019). Surprisingly, the muscids (*Musca domestica*), were sparsely observed throughout the decomposition stages of the cadavers without forensic deduction on them, which is contrary to the submission of (Abouzied, 2014) that they were constrained to the bloated stage. Similar to the findings of Benbow et al. (2013), negligible disparity of insect species occurred during the rainy and the dry seasons. The composition of the insect during the rainy season was higher than the ones collected during dry season. Based on the decomposition stages of the pig cadavers, insect species composition was lower during the fresh decay but increased during the bloated and wet decay stages. The necrophagous insect communities at each decomposition stage were distinctively paramount irrespective of the season. Hence, the calliphorids were better forensic indicators for the two seasons, especially during the fresh through the wet

decay stages, and were succeeded by the dermestids and clerids at the later stages.

Table 1. Insect composition of the pig cadavers in a forest at University of Port Harcourt, Nigeria.

Order	Family	Species	RS	DS
Coleoptera	Carabidae	<i>Drimostoma puntulatum</i> (Tsch.)	+	-
	Chrysomelidae	<i>Buphonella</i> sp.	+	-
		<i>Corynodes compressicornis</i> (Kunt.)	+	-
	Cleridae	<i>Necrobia ruficollis</i> (Fab.)	+	+
		<i>Necrobia rufipes</i> (Deg.)	+	+
	Dermestidae	<i>Dermestes frischii</i> (Kug.)	+	+
	Histeridae	<i>Hister monitor</i> (Lewis)	+	+
		<i>Hister castanipe</i> (Mars.)	+	+
		<i>Hister</i> sp.	+	+
	Scarabaeidae	<i>Onthophagus vinctus</i> (Er.)	+	+
	Staphylinidae	<i>Philonthus</i> sp.	+	+
		<i>Staphylinus</i> sp.	+	+
Diptera	Calliphoridae	<i>Chrysomya albiceps</i> (Weid.)	+	+
		<i>Chrysomya chloropyga</i> (Weid.)	+	+
		<i>Chrysomya regalis</i> (Rob-Desv.)	+	+
	Muscidae	<i>Musca confusciata</i> (Speiser)	+	-
		<i>Musca domestica</i> (Linn.)	+	+
		<i>Musca sorbens</i> (Weid.)	+	-
		<i>Pyrellia scientillans</i> (Bigot)	+	-
	Stratiomyiidae	<i>Hermetia illucens</i> (Linn.)	+	+
		<i>Sagaricera analis</i> (Macq.)	+	-
		<i>Sagaricera excellens</i> (Brun.)	+	-
Hymenoptera	Ulidiidae	<i>Chrysomyza africana</i> (Hendel)	+	+
	Formicidae	<i>Camponotus acvapimensis</i> (Mayr.)	+	+
		<i>Camponotus perrisi</i> (For.)	+	+
		<i>Oecophylla longinoda</i> (Latr.)	+	+
		<i>Pheidole</i> sp.	+	+
		Total	27	19

Note; RS = rainy season, DS = dry season, + = present, - = absent

During the rainy season, the insect species found on the cadavers showed that calliphorids, muscids, and formicids were the first to arrive on the cadaver. During the fresh decomposition stage, only the calliphorids laid eggs on the cadavers hence, their eggs were the only insect of forensic importance of that stage. During the bloated stage, other insects such as the adults of stratiomyids, ulidiids, carabids, histerids, scarabaeids, and staphylinids arrived in addition to the first group of insects. At this stage, the first larval instar and few eggs of the calliphorids were dominant on the cadavers as reliable insect specimens of forensic importance. During the wet decomposition stage of the cadavers, insect composition has spanned into another groups of insects such as chrysomelids, clerids and dermestids. They arrived on the cadavers in addition to the existing insect composition of the bloated stage with the eggs of the stratiomyids been spotted, and the larvae of ulidiids concentrating underneath the cadavers, while the pupae of calliphorids concentrate under the mats. Thus, to estimate the PMI of wet decay stage of the cadaver, the eggs of the stratiomyids, larvae of ulidiids, and pupae of the calliphorids would serve as insect specimens of forensic importance. During the dry decomposition stage, the pupae and prepupae of calliphorids were still observed without their eggs anymore. Also, the larvae of ulidiids and stratiomyids were observed as the only, and last immature stages of the dipteran insects, which clearly mark the dry decomposition stages of the cadavers (fig. 1). This insect faunal composition changes therefore, presented a transient succession sequence of insect species on the cadavers. Similar pattern of insect succession sequence was observed during the dry season of the study. However, a minor distinction was noted during the dry season, larval stages of the dermestids were observed on the cadavers during skeletonization of the cadavers (beyond day 16), which were not recorded during the rainy season.

The succession sequence of the insects from the fresh through dry decay stages was unique with at least one insect species that distinctively marked each stage of the decomposition. Thus, during the fresh stage, the cadavers were dominated by the calliphorids and their eggs. The age of the immature insects, according to (Sharma et al., 2021) mainly the calliphorids was the anchorage for the estimation of the time of death of cadavers. The predictable arrival of the histerids on day 3 for both seasons, marked them as distinct PMI estimators of the bloated stage in the study. During the wet decay stage, the presence of the calliphorids (larvae), histerids (adults) in addition to the arrival of the clerids and dermestids, could also serve as insects of forensic importance with a PMI of 4 days (Fig. 1 & 2). Also, the age of the cadaver during the wet decay stage could be ascertained by the presence of ulidiid larvae on days 7 and 8 for dry and rainy seasons respectively.

During the dry decay stage for the dry season, or the wet skeletonized decay stage during the rainy season, the presence and age of the stratiomyid larvae, adults of clerids and dermestids, and complete absence of larvae and pupae of calliphorids could be used to estimate the PMI of cadaver in the study area. This submission may partly contradict the reports of (Gomes et al., 2009), who submitted that calliphorids are characteristics of fresh and bloated decay stages of cadavers, while clerids and dermestids characterize the later stages of cadaver decay. In this study, calliphorids were observed beyond the bloated stages, while the clerids and dermestids were observed at the earliest stage of decomposition when the cadaver tissues were still wet. In relation to the chronology of forensic flies (*Chrysomya* spp., *Musca domestica*, *Chrysomya africana*, and *Hermatia illucens*) as submitted by (Cruz et al., 2021) and beetles by (Dekeirsschieter et al., 2021) as the main colonizers of human and animal cadavers, are also chronicled in this study, which is the hallmark of forensic entomology.

Key:	Fresh decay			Bloated decay			Wet decay			Dry decay						
Insect Species	Decomposition Duration in days															
	FD		BD		WD					DD						
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
<i>Drimostoma puntulatum</i> (Tsch.)		A														
<i>Buphonella</i> sp.								A	A							
<i>Corynodes compressicornis</i> (Kunt.)			A	A												
<i>Necrobia ruficolis</i> (Fab.)							A	A	A	A	A	A	A	A	A	A
<i>Necrobia rufipes</i> (Deg.)				A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Dermestes frischii</i> (Kug.)				A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Hister monitor</i> (Lewis)			A	A	A	A	A	A	A							
<i>Hister castanipes</i> (Mars.)			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Hister</i> sp.			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Onthophagus vinctus</i> (Er.)			A			A										
<i>Philonthus</i> sp.			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Staphylinus</i> sp.				A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Chrysomya albiceps</i> (Weid.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Chrysomya chloropyga</i> (Weid.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Chrysomya regalis</i> (Rob-Desv.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Musca confusciata</i> (Speiser)					A	A										
<i>Musca domestica</i> (Linn.)	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Musca sorbens</i> (Weid.)											A				A	
<i>Pyrellia scientillans</i> (Bigot)						A										
<i>Hermetia illucens</i> (Linn.)			A	A	A	A	A	A	A	L	L	L	L	L	L	L
<i>Sagaricera analis</i> (Macq.)						A						A				
<i>Sagaricera excellens</i> (Brun.)										A					A	
<i>Chrysomyza africana</i> (Hendel)		A	A	A	A	A	A	AL	AL	AL	AL	AL	AL	AL	AL	AL
<i>Camponotus acvapimensis</i> (Mayr.)	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Camponotus perrisi</i> (For.)	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Oecophylla longinoda</i> (Latr.)		A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Pheidole</i> sp.			A	A	A	A	A	A	A	A	A	A	A	A	A	A

Figure 1. Insect succession on the pig cadavers in the forest during the rainy season.

Note: FD (fresh decay), BD (bloated decay), WD (wet decay), DD (dry decay)
 A (adult), E (egg), L (larva), P (pupa)

Key:	Fresh decay			Bloated decay			Wet decay			Dry decay						
Insect Species	Decomposition Duration in days															
	FD		BD		WD					DD						
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
<i>Drimostoma puntulatum</i> (Tsch.)			A													
<i>Buphonella</i> sp.								A	A							
<i>Corynodes compressicornis</i> (Kunt.)																
<i>Necrobia ruficolis</i> (Fab.)							A	A	A	A	A	A	A	A	A	A
<i>Necrobia rufipes</i> (Deg.)				A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Dermestes frischii</i> (Kug.)				A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Hister monitor</i> (Lewis)			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Hister castanipes</i> (Mars.)			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Hister</i> sp.			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Onthophagus vinctus</i> (Er.)			A		A	A										
<i>Philonthus</i> sp.			A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Staphylinus</i> sp.																
<i>Chrysomya albiceps</i> (Weid.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Chrysomya chloropyga</i> (Weid.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Chrysomya regalis</i> (Rob-Desv.)	AE	AEL	AL	AL	AL	AL	LP	LP	ALP	ALP	ALP	ALP	ALP	ALP	AP	AP
<i>Musca confusciata</i> (Speiser)																
<i>Musca domestica</i> (Linn.)		A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Musca sorbens</i> (Weid.)																
<i>Pyrellia scientillans</i> (Bigot)																
<i>Hermetia illucens</i> (Linn.)					A	A	A	AE	EL	L	L	L	L	L	L	L
<i>Sagaricera analis</i> (Macq.)																
<i>Sagaricera excellens</i> (Brun.)																
<i>Chrysomyza africana</i> (Hendel)		A	A	A	A	A	AL	AL	AL	AL	AL	AL	AL	AL	AL	AL
<i>Camponotus acvapimensis</i> (Mayr.)	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Camponotus perrisi</i> (For.)	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
<i>Oecophylla longinoda</i> (Latr.)																
<i>Pheidole</i> sp.				A	A	A										

Figure 2. Insect succession on the pig cadavers in the forest during the dry season.

Note: FD (fresh decay), BD (bloated decay), WD (wet decay), DD (dry decay)
A (adult), E (egg), L (larva), P (pupa)

Conclusion and Recommendations

The forensic insect indicators of the study area, as observed on decomposing pig cadavers are the same for the rainy and dry reason study. The predictability of the forensic insect indicators and their succession sequence as observed on the pig cadavers, are the biological clock that would be extrapolated by forensic investigators of the study area to estimate a PMI of a questionable death when the need arises. Further and extended studies of insects of forensic importance are needed in the study region in order to enrich the baseline data for forensic insect indicators in the region.

Literature Cited

- Abajue, M.C., & Ewuim, SC. (2016). Forensic entomology: decomposing pig carrion and its associating insects in Okija, Anambra State Nigeria. *American Journal of Biology and Life Sciences*, 4(2), 6-11.
- Abajue, M.C., & Ewuim, S.C. (2020). Evaluation of activities of dipteran maggots on a poisoned pig cadaver at Nnamdi Azikiwe University, Nigeria. *Egyptian Journal of Forensic Sciences*, 10(33), 1-10.
- Abajue, M.C., Ewuim, S.C., & Akunne, CE. (2013). Insects associated with decomposing pig carrions in Okija, Anambra State, Nigeria. *The Bioscientist*, 1(1), 54-59.
- Abajue, M.C., Ewuim, S.C., & Akunne C.E. (2017). Insect larvae recovered from decomposing pig carrions in Okija, Anambra State, Nigeria. *Animal Research International*, 14(2), 2764 – 2768.

- Abajue, M.C., Ewuim, S.C., & Uhuo, C.A. (2021). Succession of three dipteran maggot families on poisoned pig cadavers. *International Journal of Entomology Research*, 6(3), 140-147.
- Abouzied, E.M. (2014). Insect colonization and succession on rabbit carcasses in southwestern mountains of the Kingdom of Saudi Arabia. *Journal of Medical Entomology*, 51(6), 1168–1174.
- Amendt, J., Krettek, R., & Zehner, R. (2004). Forensic entomology. *Naturwissenschaften*, 91, 51-65.
- American veterinary medical association (AVMA, 2014). *Guidelines for the euthanasia of animals*. Assessed online on January, 03, 2014.
- Benbow, M.E., Lewis, A.J., Tomberlin, J.K., & Pechal, J.L. (2013) Seasonal necrophagous insect community assembly during vertebrate carrion decomposition. *Journal of Medical Entomology*, 50(2), 440-450.
- Benceke, M. (2004). *Forensic entomology: Arthropods and corpses*. In: Tsokos, M. (ed) Forensic pathology review, volume II. Humana Press.
- Bugelli, V., Gherardi, M., Focardi, M., Pinchi, V., Vanin, S., & Campobasso, C.P. (2018). Decomposition pattern and insect colonization in two cases of suicide by hanging. *Forensic Sciences Research*, 3(1), 94-102.
- Byrd, J.H., & Castner, J.L. (2010). *Forensic entomology: The utility of arthropods in legal investigations* (2nd ed). CRC press.

- Cordeiroa, C., Ordóñez-Mayánd, L., Lendoiroe, E., Febrero-Bandef, M., Vieirab, D.N., & Muñoz-Barúse, J.I. (2019). A reliable method for estimating the postmortem interval from the biochemistry of the vitreous humor, temperature and body weight. *Forensic Science International*, 295, 157–168.
- Cruz, T.M., Barbosa, T.M., Thyssen, P.J., & Vasconcelos, S.D. (2021). Diversity of Diptera species associated with pig carcasses in a Brazilian city exposed to high rates of homicide. *Papeis Avulsos de Zoologia*, 61, e20216101.
- Dekeirsschieter, J., Frederickx, C., Verheggen, F.J., Boxho, P., & Haubruge, E. (2013). Forensic entomology investigations from Doctor
- Marcel Leclercq (1924-2008): A review of cases from 1969 to 2005. *Journal of Medical Entomology*, 50, 935-954.
- Dupont., F.Y.F., Champlain, D., Cyrille, A.W.J., & Félix, B.B.C. (2011). A preliminary study of arthropod associated with carrion in Yaounde, Cameroon: A first step in forensic entomology in Central Africa. *Journal of Ecology and the Natural Environment*, 3(6), 215-220.
- Ekanem, M.S., & Dike, M.C. (2010). Arthropods Succession on pig carcasses in Southeastern Nigeria. *Papeis Avulsos de Zoologia*, 50(35), 561-570.
- Ekrakene, T., & Iloba, B.N. (2011). One death, many insect species yet one insects generation. *Journal of Entomology*, 8, 27-39.
- El-Gawad, A.A., Badawy, R.M., Abd El-Bar, M.M., Mohamed, A., & Kenawy, M.A. (2019). Successive waves of dipteran flies attracted to warfarin-intoxicated rabbit carcasses in Cairo, Egypt. *The Journal of Basic and Applied Zoology*, 80(56), 1-10.

Galloway, A., Birkby, W.A., Jones, A.M., Henry, T.E. and Parks, B.O. (1989). *Journal of Forensic Sciences*, 34(3), 607-616.

Gennard, D.E. (2007). *Forensic entomology: An introduction*. John Wiley.
Gomes, L., Gomes, G., & Desuo, I.C. (2009). A preliminary study of insect fauna on pig carcasses located in sugarcane in winter in southeastern Brazil. *Medical and Veterinary Entomology*, 23, 155–159.

Lutz, L., Williams, K.A., Villet, M.H., Ekanem, M., & Szpila, K. (2018). Species identification of adult African blowflies (Diptera: Calliphoridae) of forensic importance. *International Journal of Legal Medicine*, 132, 831-842.

Lutz, L., Zehner, R., Verhoff, M.A., Bratzke, & Amendt, J. (2021). It is all about the insects: a retrospective on 20 years of forensic entomology highlights the importance of insects in legal investigations. *International Journal of Legal Medicine*, 135, 2637-2651.

Medler, J. Insects of Nigeria-checklist and bibliography. *Memoirs of the American Entomological Society*, 30, 1-919.

NDES, (2001). Biological environmental research report, Rivers State University of Science and Technology (RUST) Port Harcourt, Nigeria. 46: 251.

Okiwelu, S.N., Ikpamii, T., & Umeozor, O.C. (2008). Arthropods associated with mammalian carcaasses in Rivers State, Nigeria. *African Journal of Biomedical Research*, 11: 339-342.

Sanford, M.R. (2017). Insects and associated arthropods analyzed during medicolegal death investigations in Harris County, Texas, USA. *PLoS ONE*, 12(6), e179404.

- Sharma, A., Drijfhout, F.P., Tomberlin, J.K., & Bala M. (2021). Cuticular hydrocarbons as a tool for determining the age of *Chrysomya rufifacies* (Diptera: Calliphoridae) larvae. *Journal Forensic Science*, 66(1), 236-244.
- Smith, K.G.V. (1986). *A manual of forensic entomology*. Trustees of the British Museum (Natural History) and Cornell University Press.
- South Australian museum wildlife ethics committee (SAMWEC, 2013). *Euthanasia of research animals in the field policy*. Assessed on January, 03, 2014.
- Tomberlin, J.K., Byrd, J.H, Wallace, J.R., & Benbow, M.E. (2012). Assessment of decomposition studies indicate need for standardized and repeatable research methods in forensic entomology. *Journal of Forensic Research*, 3(5), 1-10.
- Van Laerhoven, S.L. (2010). Ecological theory and its application in forensic entomology. In Byrd JH, Castner J.L. (eds), *Forensic entomology; the utility of Arthropods in legal investigations*, (2nd edn). CRC Press.
- Villet, M.H. (2011). African carrion ecosystems and their insect communities in relation to forensic entomology. *Pest Technology*, 5(1), 1-15.
- Wang, Y., Wang, M., Xu, W., Zhang, Y., & Wang, J. (2021). Forensic entomology in China and its challenges. *Insects*, 12(3), 1-16.