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ARTHROPOD PROFILE ON POISONED AND PRESERVED CARRIONS

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ABSTRACT

Arthropod invasion on poisoned and embalmed carrions of the African giant rat (Crycetomys gambianus) killed from phostoxin poison was monitored for a period of 30 days (November 13th-December 23rd 2009) with a view to establishing arthropod profile. A total of 18 species (distributed in 13 families, 6 orders and 2 classes of the phylum Arthropoda) were collected and identified. Occurrence of these species was independent of the killing and embalming agent. Species from the order Diptera were most predominant, followed by the Coleoptera and the Hymenoptera. Members of the Calliphoridae, Sarcophagidae, Dermestidae, Cleridae, Stapyhinidae, Scarabaeidae, Bostricidae and the Formicidae families were involved in the degradation process of the carrions, making them forensically important. Despite similar insect fauna, the decomposition processes was affected by both the killing and embalming agents with carrions poisoned and embalmed recording longest time in all stages of decomposition compared to that from the controlled sets. The details of general procedure used and the results obtained are reported.

KEY WORDS: *Arthropod, Carrion, Crycetomys gambianus, Decomposition, Embalm, Forensic entomology, Poison*

INTRODUCTION

Insects are cosmopolitan and their presence in varying habitat cannot be overemphasized. Their presence on dead bodies can be used to determine the cause of death, circumstances behind the death, time of death, locate the death scene, detect the time of infliction of wounds and also cases of neglect in children and adults (Benecke, 2001). Forensic entomology is the field of knowledge that deals with the association of insects and other arthropods in relation to a given stage of decay to answer questions of legal interests. Lord and Stevenson (1986) categorized forensic entomology into urban, stored product and medico-legal forensic entomology. Urban forensic entomology includes civic law actions and litigation involving

arthropods in houses that may results to cases between parties and service provider such as land lord (Anderson, 2000). Stored product forensic entomology includes arthropod infestation of edible commodities such as cereals and other kitchen quarantine. Medico-legal forensic entomology deals with the evidence that may be gathered through studies at events such as murder, suicide, rape, physical abuse, contraband trafficking and in entomotoxicology (using entomological specimen found at a scene of death to test for drugs that might have play a role in the victims death).

Medico-legal forensic entomology deals with the post mortem interval (PMI), time of death, site of death, cases involving possible sudden death, traffic accidents (Anderson, 2000) and is linked with fields such as forensic pathology and forensic toxicology. Many factors such as temperature, season, time of day, accessibility and physical position of a carcass, size, type of carcass, vertebrate scavengers insect abundance, biology and geographical distribution of the necrophagous insect can influence insect succession and decomposition on carrion (Anderson 2000; Hall and Doisy 1993; Payne 1965). These factors make the study of insect succession on carrion different under different condition, thus making the post mortem interval possible. However, this leaves the possibility of erroneous conclusion that may be detrimental to legal cases.

Full blown forensic entomology did not start in Nigeria till 1988 (Ekanem and Usua, 1997). Ekanem and Usua (2000) studied the biology of *Chrysomya Chloropyga* and *Hemipyrellia fernandica* and opened up possibilities and roles for flies in forensic entomology. Iloba and Fawole (2006) while studying the comparative arthropod fauna in exposed carrions across the vertebrate classes reported a total of 23 arthropod species from Diptera, Coleoptera and Hymenoptera orders playing significant role in decomposition of the carrions. Other literatures as Ekrakene and Iloba (2011) and Ekrakene (2012) are among recent efforts to broaden the literature base of forensic entomology in Nigeria. Arising from these literatures forensic entomological information is gradually being accumulated in Nigeria. There is the need to expand the research scope to micro-conditions such as the effect of poisons and embalming agents on the profile of arthropod fauna that visit such carrions as in this present work.

MATERIALS AND METHODS

Study Area

This study was carried out at a location behind the Botanical garden close to Dentistry quarters, University of Benin, Ugbowo campus, Edo state Benin City which lies between latitude 6°35 and 6°26 longitude 5°35 and 5°41. A rainforest region of Nigeria, it is characterized by an annual rainfall of 1850-2455mm and a temperature range of 30-36.5°C.

The experimental area measured approximately 7m by 8m. The mean temperature for the duration of the experiment was 32.7°C.

Killing Exercise of Animal Specimens and Experimental Layout

A study of exposed poisoned and embalmed carrions on the African giant rat (*Crycetomys gambianus*) a

representative species of the vertebrate group was conducted from November 13th - December 23rd, 2009. In the experiment, three replicates in four sets of the vertebrate group with mean weights of 1.80±0.20kg (Mean±S.E) purchased from the Oba, New Benin and Uselu markets all in the Benin City metropolis were used. The following methods of kill and set arrangements were adopted:

Set A: (Poisoned and embalmed): Three *C. gambianus* were killed by stomach poisoning with phostoxin. This set of carrions was immediately injected with 5.0 ml of 50% formalin (an embalming agent) using syringe. (Set A: poisoned and embalmed).

Set B: (Set B: embalmed only): Three rats killed by cervical dislocation and immediately injected with 5.0 ml formalin.

Set C: (Set C: poisoned only): Three rats killed by stomach poisoning after feeding from admixture of bread debris with phostoxin.

Set D: (Set D: control): Three rats killed by cervical dislocation.

This set of twelve rats were packed in separate polyethylene thrash bags corresponding to respective sets, labeled and transported to the study area. These were all covered with a wire mesh to avoid attack by scavengers.

Observations and Daily Data Collections

Insect data collections were made at least once daily between the hours of 2.00 – 6pm when most flies are active (Slone, *et al*., 2005). From the first day of deposition up to the last day, adult flies on carrions were collected using a 12-inch sweep net. Flies collected were placed in vials containing 70% ethyl alcohol; later pinned and identified.

Oviposition on carrions was monitored from the first day of deposition till when laid eggs started to hatch on the carrions. Eggs and larvae were sampled from natural orifices of carrions such as ear, nostrils, eyes, anus and mouth at different stages of decomposition. This was achieved by the careful examination of carrions, using forceps and a constructed brush to pick up the eggs while a 5 ml plastic spoon was used to collect larvae into separately labeled rearing kits designed for the purpose. The colour of eggs was recorded by visual observation. Sizes of eggs from different egg masses were assessed using a-30cm length rule to enhance identification.

Larval samples from each of the maggot masses were obtained according to Fisher (1980). A 5ml plastic spoon was used to collect larvae during sampling days within the active larval period against individual carrion. Half of the larvae collected were killed in boiled water before being placed in preservative solution to avoid shrinkage (Tantawi and Greenberg, 1993). Thereafter, they were placed in vials containing 70% ethyl alcohol for preservation. Third instar larvae were used for identification to species level, by noting sizes of larvae, presence of setae, among others features. The remaining half of the sampled larvae from each sampling were placed in rearing kits designed for the purpose and reared to adult stages in a laboratory.

Laboratory Rearing of Insects

Maggot rearing was carried out at the Animal and Environmental Biology Laboratory of the University of Benin, Benin City, according to Easton and Smith (1970), Nuorteva (1970) and Fisher (1980).

Maggot rearing was achieved with plastic containers measuring 20cm in diameter covered with muslin cloth tightly fitted with rubber band which were half-filled with substrate (adopted from Slone *et al*. 2005). The substrate was formulated from a mixture of dried leaves, grasses and sandy soil. The plant mixture was ground thoroughly using an electric blender.

A few maggot samples from different orifices (ear, nostril, eye, cut region, mouth and anus) were placed on different rearing media, which were pieces of liver/tissue of pig/cow purchased from the market, (adopted from Aggarwal, 2005). The containers were labeled according to pig sets and orifice from where the maggots were sampled and observations made several times to note changes in larval, pupation and emergence of adult insects. The containers of live maggots were monitored in the laboratory daily with prevailing daily temperature 28 ± 2^{0}C and 70% relative humidity.

RESULTS

The results of mean time spent on each stages of decomposition arising from the various carrions killed and embalmed are presented in Fig. 1. It revealed that irrespective of killing methods and preservation, four stages of decomposition are easily identifiable viz. fresh, bloated, wet and dry decay stages. While mean period of each decomposition stages are highest for carrions killed by poisoning with phostoxin and embalmed with formalin, the controlled set (neither poisoned nor embalmed) recorded least values in all decomposition stages. The mean decomposition time spent at the various stages of decomposition is similar among the carrions poison only and embalm only.

The insect fauna obtained from this investigation are presented in Table 1. The summaries indicate that insect fauna were sampled from four main insect orders (Diptera, Coleoptera, Hymenoptera and Dictyoptera) and 11 families. Dipterans were represented with Calliphoridae, Sarcophagidae and Muscidae families, accounting for 44% (431) of the total sampled fauna. The Coleopterans on the other hand were represented by Dermestidae, Staphyliidae, Scarabaeidae, Silphidae, Cleridae and Bostrichidae collectively accounting for 27% (253) of sampled fauna, whereas Hymenopterans which were represented by Formicidae family accounted for 25% (233). The others which covered Arachnida order weighted 4% (34) of all collected fauna (Fig. 2). Apart from *Dermestis maculatus* and *Rhizoperta dominica* which were sampled in the control and poison only sets, all carrions had similar insect species through their stages of decomposition. The main difference occurred in the number of insects sampled from sets of the carrions. Also, the mean number of species of Dipterans sampled is presented in Fig. 4. In all species, number of Dipterans sampled was highest in the control set compared to all the others.

The mean number of fauna sampled from the various carrions resulting from the killing methods and embalmment is presented in Fig. 3. The result shows that carrions poisoned and embalmed recorded the least number of insect fauna, accounting for 8.6% of the total sampled

fauna compared to the highest value of 42.6% sampled from the control set (Set D). However, poisoned carrions only (Set B) and embalmed only (Set C) accounted for 39.2 and 9.6%, respectively.

Table 1: Insect fauna sampled from the carrions

Insects	Poison and Embalm	Embalm Only	Poison Only	Control
Chrysoma bezziana	-	-	+	+
Lucilia serricata	+	+	+	+
Calliphora sp	+	+	+	+
Sarcophaga Carnaria	+	+	+	+
Musca domestica	+	+	+	+
Dermestes maculatus	+	+	+	+
Quedius fuliginousus	+	+	+	+
Trox scaber	+	+	+	+
Necrophorus sp	+	+	+	+
Necrobia rufipes	+	+	+	+
Rhizopertha dominica	-	-	+	+

+ = present; - = absent

Fig. 1: Mean duration in days at the various stages of decomposition of the carrions

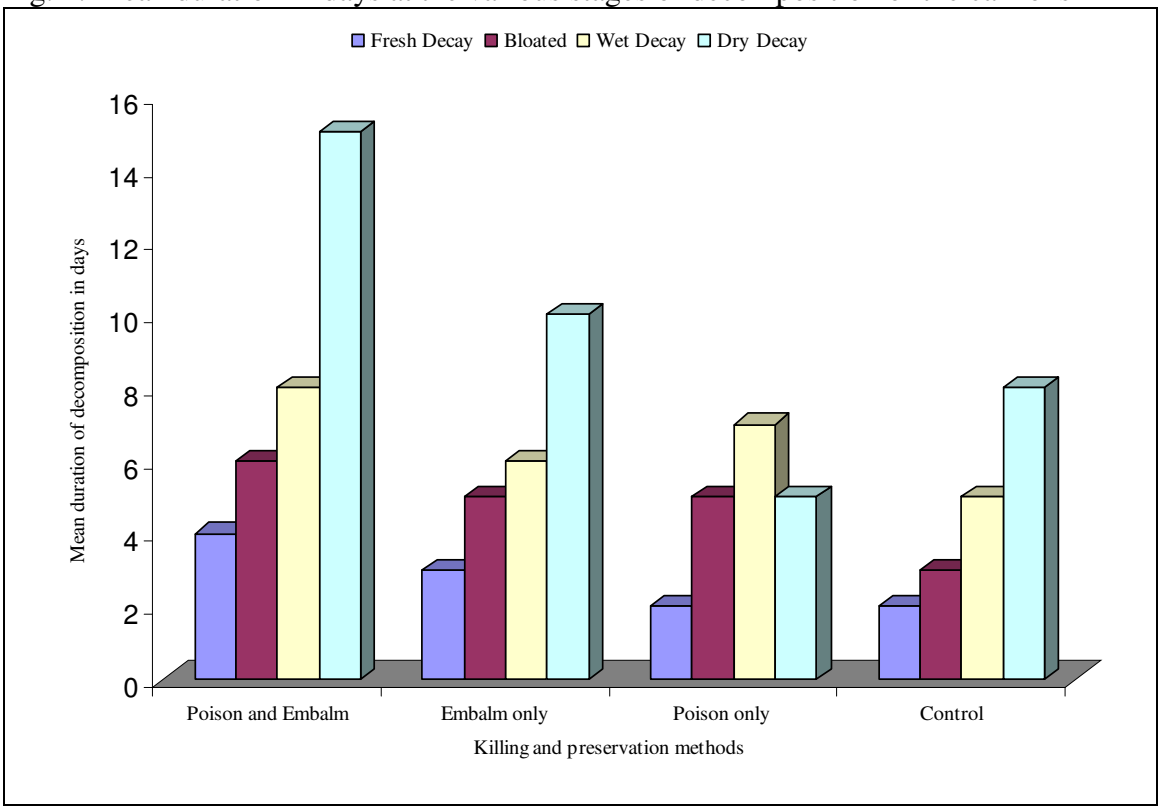


Fig. 2: Percentage abundance of insect orders sampled from the carrions

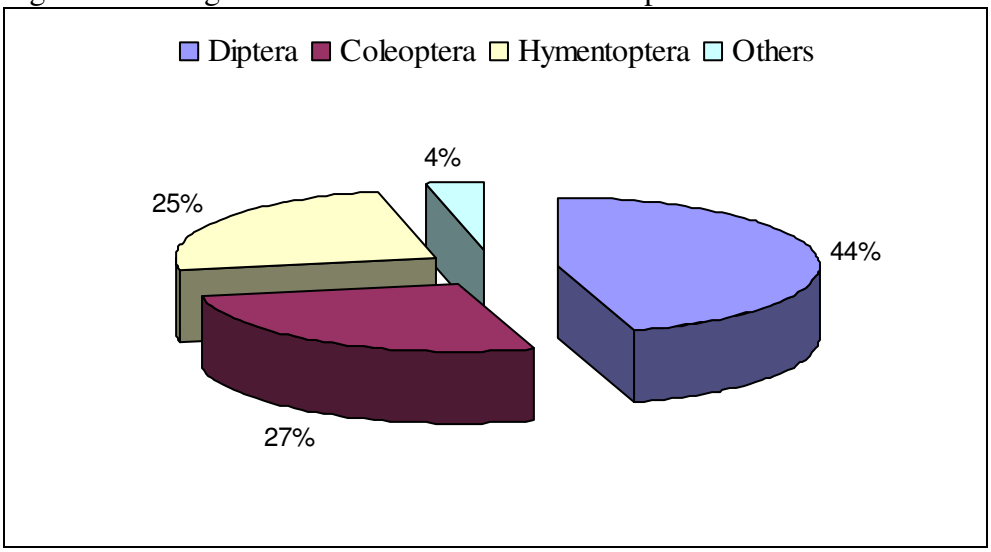
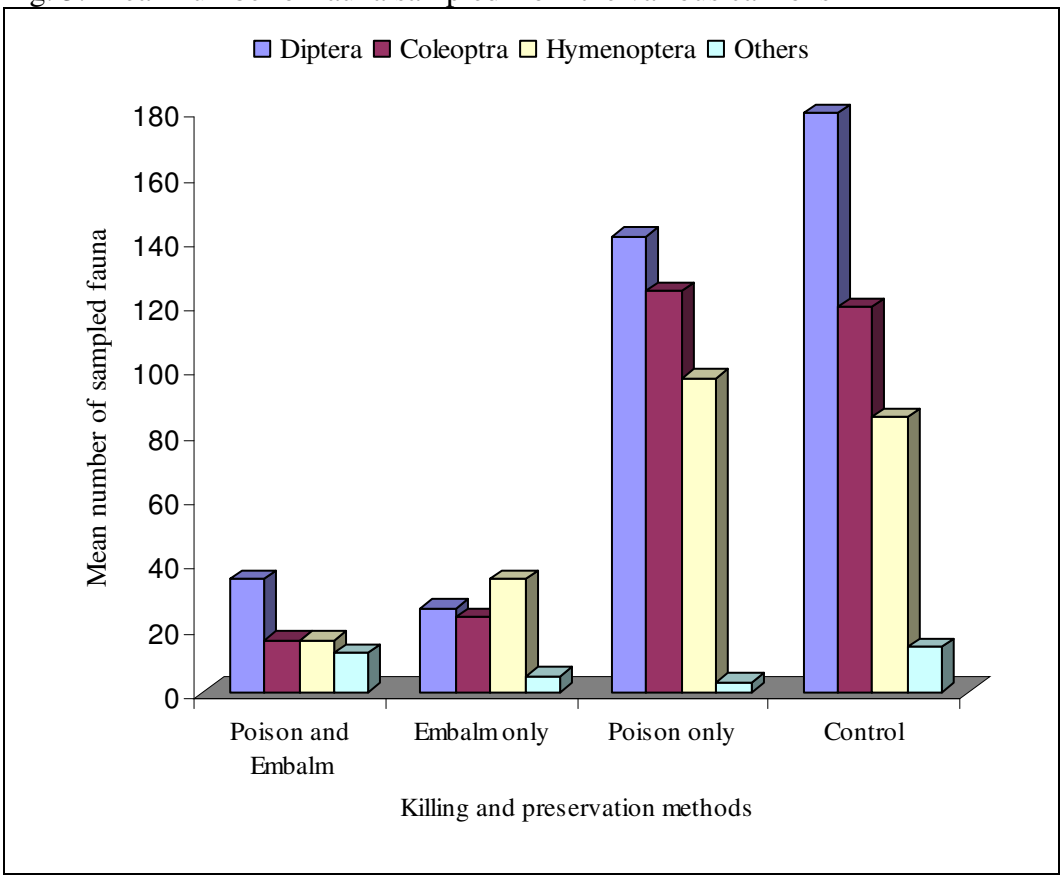


Fig. 3: Mean number of fauna sampled from the various carrions



DISCUSSION

This investigation revealed descriptive decomposition stages. Though Schoenly and Reid (1987) defined decomposition as a continuous process without discrete stages, the four stages of decomposition identified in this study agrees with earlier researchers who divided the process into four to six stages (Payne 1965; Lord and Burger 1984; Morris 1988; Tantawi *et al.* 1996). These successive and distinct stages were fresh, bloated, wet decay as well as dry decay. These stages were distinctly marked by characteristic odour, presence or absence of insect, the stage and type of insect species present as well as the insect activity synchronizing with the insect present. These observations agreed in many respect to the findings of earlier researchers, notably, Reed (1958), Rodriguez and Bass (1983), Morris (1988) and Tantawi *et al*. (1996) as well as Parikh (1999), Vij (2001) and Pillay (2004).

Array of insects from different orders have been associated with the decomposition processes of carrions. Smith (1986) categorized insects associated with carrion on the basis of their food preferences and their ecological role. Gill (2005) emphasized that, these insects species typically occur in succession and respond to

decompositional changes of the carcass. This present study recorded insects' species from three main insect orders; Diptera, Coleoptera and Hymenoptera with a few others from the class Arachnida irrespective of whether carrions were poisoned, embalmed or both. The findings in this present work that most fauna obtained are insects and were from Diptera, Coleoptera and Hymenoptera concurred with earlier investigators including Payne (1965), Smith (1986), Keh (1985), Lord (1990) Catts and Goff (1992), Anderson (2000), Aggarwal (2005) and others.

The families of the Diptera order represented in this study include; Calliphoridae, Muscidae and Sarcophagidae, while the Coleoptera order was represented by the Cleridae, Staphyliidae, Scarabaeidae, Silphidae, Bostrichidae and Dermestidae families. The order Hymenoptera was represented by the ants (formicidae). These families were seen to be consistent in their occurrence and have a regular sequence of occurrence. The families of insects found in this work have been highlighted by other researchers to be involved in decomposition processes of carrions and most species from these families have been used as forensic indicators. Members of Diptera are forensically important as they contribute significantly to the degradation of carrions when they form maggot mass on carrions. They are among the earliest insects that visit carrions where they lay eggs which develop through stages that have systematically synchronize with stages of decomposition. Many genera of this order including *Lucillia, Chrysoma, Calliphora, Musca,* and *Sacophaga* have been used as forensic indicators to solve various problems relating to medico-legal issues (Singh and Bharti 2000, Aggarwal, 2005 and Carvalho *et al*., 2004).

Also, many genera in Coleoptera including *Dermestis* and *Necrobia* have been used by various forensic investigators. These beetles are well known to feed on dry skin and bones (Payne and King, 1970). Putman (1977) considered them as true carrion feeders playing important role in carcass degradation. Catts and Goff (1992) and Goff (1992) have stated that *Dermestis maculatus*, (hide beetles) have the potential to offer investigators an estimation of the time since death in homicide or questionable cases. Payne and King (1970) have noted that larvae and adults of these beetles associated with carrions feed on dried skin, sinews and bones.

Members of the formicidae (ants) family were observed throughout this present study. Their occurrence was observed to be irrespective of killing method and stage of decomposition. Field observation revealed that their presence increased with increase in decomposition stages and insects development. They used carcass as food and acted as predator to maggots, pupae and puparia cases. This may place them as adventives or incidental species. This suggests that their presence is opportunistic and therefore unimportant in the decomposition of carrion. However, their predatory actions on insects, especially the eggs and the maggots have the potential of retarding the rate of decomposition. Similar observations have been made by Fuller (1934), Early and Goff (1986), Singh and Bhanti (2000) and Aggarwal (2005).

The evaluation of the arthropod profile for poison, embalm and control sets of the African giant rats (*Crycetomys gambianus*) carrions resulted in similar insect types irrespective of the presence of killing and embalming agents compared to the control ones. However, the combined effect of killing agent and embalmment with formalin prolonged the decomposition process of carrions compared to controlled carrions. It is evident from this investigation therefore that, killing and preserving agents may not necessarily affect insect fauna profile on carrions but can affect the decomposition process thereby influencing the determination of Post-Mortem Interval (PMI) using the insect method.

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