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Killing Method Influences Decomposition, Entomofauna and Successional Pattern of Exposed Juvenile Pig Carcasses in Wukari, Nigeria

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Abstract

Homicide-related crimes (especially juveniles) are serious problems in Africa and particularly, in Nigeria, with little success in apprehension of culprits recorded due to a lack of substantial forensic data to aid investigation processes. To aid the mitigation of this challenge, two juvenile pigs were killed (one by slaughtering and the other by strangulation) at the Research Garden of the Biological Sciences Department, Federal University Wukari, Nigeria. Both carcasses were left exposed to sunlight 25 m apart, while being protected from vertebrate scavengers using meshed metal cages. Observable postmortem changes on both carcasses were recorded while adult insect sampling using a combination of aerial net, pitfall traps and manual searching continued from the fresh to dry stages of decomposition for both carcasses. Data from sampled insects obtained were used to compute abundance, frequency of occurrence and dominance at each decomposition stage. Appropriate diversity indices were obtained using the Paleontological Statistical Tool-Past₃, while similarity in entomofauna between both carcasses was determined using Jaccard's index. Although both carcasses completed decomposition in 14 days, the strangulated carcass had faster active and advanced stages with prolonged dry stage vis-à-vis the slaughtered carcass. Also, the slaughtered carcass had higher insect abundance (895) than the strangulated carcass (781), with peak diversity and richness at the advanced stages. The slaughtered carcass was also observed to have higher diversity and richness values (1.942, 2.517) than the strangulated carcass (1.801, 1.597). Five (5) dominant insects (*Hister monitor, Dermestes maculatus*, *Zophosis* sp., *Chrysomya chloropyga,* and *Pheidole* sp.) invaded the strangulated carcass while *Crematogaster* sp. invaded the slaughtered carcass in addition to these. Overall, over 60% total species similarity and 80% dominant species similarity existed between the carcasses. Hence the killing methods used in this study largely influenced variations in the duration of the five decomposition stages, observable postmortem changes as well as insect abundance but the succession of invading insects was largely similar.

Keywords: Decomposition, juvenile pigs, postmortem changes, slaughtered, strangulated, dominant insect species.

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INTRODUCTION

Insects are the first and silent witnesses to death-related crime scenes. Their activities and/or interactions with carcasses can be instrumental in determining the minimum postmortem interval (PMI_{min}) of a carcass which aids investigative processes as well as enhance the accuracy of the justice system when dealing with homicide cases (Byrd and Tomberlin, 2020; Eze *et al.,* 2021). Aside from other factors which influence insect abundance and succession on carcasses such as geographical area, climate, exposure status and size; killing method or cause of death has also been shown to be a key factor in the decomposition process of a carrion (Sonker *et al.,* 2018; Eze *et al.,* 2021; Chukwu *et al*., 2023).

Decomposition of carcasses is continuous with five notable stages characterized by observable physical changes vis-à-vis successive invasion of arthropod (mostly insects) species (Ana and Betina, 2015; Michael *et al.,* 2022). The physiological, anatomical, and decompositional similarities of pigs with humans make pigs the most appropriate animal model for studying the successional pattern of cadaverous entomofauna associated with human cadavers (Davood *et al.,* 2019; Matuszewski *et al.,* 2020).

Nigeria is majorly faced with the challenge of an increased rate of criminality and insecurity which results in violent loss of lives through different means/methods almost on daily basis. Ethno-religious crisis, ritual killings, banditry, among others make frequent news headlines (Kodili, 2020; Eze *et al.,* 2021; Chukwu *et al*., 2023). But seldom are insect activities on cadavers employed to aid investigative processes.

Recent findings show that juveniles are quite vulnerable to homicide cases in Nigeria (Chukwu *et al*., 2022; Chukwu *et al*., 2023). There is a dearth of juvenile-related cadaverous entomofaunal data which can support criminal investigation leading to early apprehension of culprits and justice delivery. Thus, this study aims to bridge this knowledge gap by providing information on the effect of killing methods on decomposition period, insect fauna and successional pattern on exposed juvenile pig carcasses.

MATERIALS AND METHODS

Study Area

This study was carried out at the Research Garden of the Biological Science Department of Federal University Wukari, Taraba State, Nigeria. This semi-urban metropolis is located between latitude 7.89N and longitude 9.77E, having a land area of 4,308 square kilometers and an altitude of 187 m. Wukari is a Southern guinea savanna with an annual rainfall of 1205 mm and average temperature of 26.8° C (Chukwu and Okrikata, 2020).

Specimen Acquisition, Killing and Experimental Setup

Two male juvenile pigs of 10 kg mean weight were purchased from a piggery in Wukari metropolis, transported to the study site and killed for the purpose of the study. One of the pigs was killed by slaughtering while the other was killed by strangulation. Both pigs were placed on metal meshes, kept 25 m apart, covered with metal cages to prevent vertebrate scavengers, and were exposed to sunlight throughout the study period (Figure 1).

Figure 1: Juvenile pig carcasses exposed to sunlight: **A.** Strangulated carcass **B.** Slaughtered carcass

Animal Research Ethics

At the time of the study, there was no ethical committee in relation to animal use in research in the University. However, all the principles of 3R (Replacement, Reduction and Refinement) principles of the Guidance on the Operation of the Animals (Scientific Procedures) Act 1986 were strictly observed.

Sampling Techniques, Protocol and Species Identification

To minimize disruption of larval establishment, only adult arthropods (majorly insects) were sampled throughout the study period using different techniques between 2pm and 6pm daily (Danisile and Samson, 2021). Two standard sweeps of the aerial net were used to sample flying insects while manual picking was adopted for ground insects. A pitfall trap which was ¾ filled with soapy water and serviced daily; was kept within a 1 m distance around the pig carcasses for sampling both nocturnal and ground insects as well.

To ensure that sampling intensity does not become a confounding factor, care was taken to sample arthropods using the same sampling intensity. Information on odour and its intensity, physical changes of the carrion and the presence of arthropods

during each stage of decomposition of the carrion were documented as outlined by Danisile and Samson*,* (2021) and Chukwu *et al*. (2023).

Sampled insects from each carrion were carefully kept in different collecting jars containing 70% ethanol and were taken to the Biological Sciences Laboratory of Federal University Wukari for sorting, morpho-identification, and grouping. Representative samples were taken to the insect museum of the Institute of Agricultural Research (IAR) in Ahmadu Bello University Zaria, for confirmatory identification (Chukwu and Okrikata, 2020).

Data Analysis

Data of sampled insect species using the respective sampling methods were combined and used to determine species abundance, relative abundance (RA%) and frequency of occurrence (FO) for the slaughtered and strangulated carcasses. Dominant species were determined by using the method described by Okrikata *et al*. (2019) in which taxa with $RA \geq 1\%$ and $FO \geq 25\%$ were considered as dominant while those with RA <1% and FO <25% were considered rare. Shannon-Weiner diversity indices (H), Margalef's richness (R), Buzas and Gibson's Evenness (E), Berger-Parker's dominance and Equitability index (J) were computed for each decompositional stage using the Paleontological Statistical Tool – Past₃ (Ana and Betina, 2015). Species similarity between the slaughtered and strangulated carcasses was determined using the Jaccard's index using the model below:

Jaccard index = $X \cap Y/X \cup Y \times 100$ Where:

 $/X \bigcap Y$ = number in both sets.

 $/X U Y$ = number in either set.

RESULTS

Decomposition Stages and Post-Mortem Changes on Slaughtered and Strangulated Juvenile Pig Carcasses

Table 1 shows that the slaughtered and strangulated carcasses were observed to complete decomposition in 14 days, having their fresh stage lasting from day $0 - 1$ and their bloated stage occurring only on day – 2. Both carcasses commenced active decay on day -3 . However, while the stage was observed from day $3 - 4$ on the strangulated carcass, it lasted till day -5 on the slaughtered carcass. A similar trend was observed at the advanced decay stage, where it lasted from day 5-6 on the strangulated carcass but day 6-9 on the slaughtered carcass. Table 1 also shows that the strangulated carcass exhibited a longer dry stage (day 7-14) than the slaughtered carcass (day 10-14).

Abundance, Frequency of Occurrence, Dominance Status and Successional Pattern of Insects Invading Slaughtered and Strangulated Juvenile Pig Carcasses

A cumulative 895 insects constituted by 16 species from 14 families were sampled from the slaughtered carcass (Table 2a). The Table also shows that *Crematogaster* sp. (Formicidae – Hymenoptera) had the highest abundance (410) and relative abundance (45.81%), *Chrysomya chloropyga*

(Calliphoridae – Diptera) had (145) abundance with (16.2%) relative abundance, *Hister monitor* (Histeridae – Coleoptera) had (109) abundance and (12.18%) relative abundance. Insect species with the least abundance (1) and relative abundance (0.11%) were *Korynetes analis* (Cleridae – Coleoptera), *Philonthus* sp. (Staphylinidae – Coleoptera), *Anomala resplendens* (Scarabaeidae – Coleoptera) and *Ommatius* sp. (Asilidae – Diptera). Considering the %RA and %FO, Table 2a shows that six (6) species, namely *H. monitor, Dermestes maculatus* (Dermestidae – Coleoptera), *Zophosis* sp. (Tenebrionidae – Coleoptera), *C. chloropyga, Crematogaster* sp. and *Pheidole* sp. (Formicidae – Hymenoptera) were dominant ($RA \ge 1\%$ and $FO \ge 25\%$) on the slaughtered carcass while the other sampled species were considered as rare $(RA < 1\%$ and FO $< 25\%$) species.

Table 2b shows that a total of 781 insect samples made up of 11 species across 11 families were retrieved from the strangulated carcass. Of the 11 species, *Pheidole* sp. had the highest abundance (510) and relative abundance (65.3%) which was distantly followed by *D. maculatus* with abundance of (78) and RA value of 10% and *C. chloropyga* with abundance of (77) and RA value of 9.9%. The least abundant species was *Ommatius* sp. It has abundance of 1 and relative abundance of 0.13%. All the species dominant on slaughtered carcass as shown in Table 2a except *Crematogaster* sp. were also dominant on the strangulated carcass.

Tables 2a and 2b also show that except for the massive invasion of *Pheidole* sp. on the strangulated carcass at the dry stage, both carcasses largely had the active - decay stage as the peak abundance stage for most invading cadaverous insects. For the slaughtered carcass, *H. monitor, D. maculatus and C. chloropyga* colonized the carcass from the fresh to the dry stage of

decomposition with gradual decline in abundance from the peak abundance stage (active decay) as the carcass completes it decomposition process while *Zophosis* sp. (Tenebrionidae – Coleoptera) and *Crematogaster* sp. invaded the carcass at the bloated stage and persisted till the dry stage.

Comparing species abundance of both carcass by Order as shown in Figure 2, Hymenoptera of both carcasses recorded the highest abundance, followed by Coleoptera, Diptera and Hemiptera. While the slaughtered carcass was also invaded by Isopteran species, the species were absent on the strangulated carcass.

Table 1: Decompositional stages and observed physical postmortem changes on slaughtered and strangulated juvenile pig carcasses exposed to sunlight

Order	Family	Genus/Species			Ac.	Ad.		RA	FO	D.
			FSH	BLT	Dcy	Dcy	DRY	(%)	(%)	Status
Coleoptera	Histeridae	Hister monitor	3	26	63	14	3	12.18	71.43	D
	Dermestidae	Dermestes								
		maculatus	\overline{c}	5	21	22	20	7.82	100	D
	Tenebrionidae	Zophosis sp.	$\mathbf{0}$	12	43	21	15	10.17	85.71	$\mathbf D$
	Cleridae	Korynetes analis	$\boldsymbol{0}$	$\boldsymbol{0}$	$\overline{0}$	$\mathbf{1}$	$\mathbf{0}$	0.11	7.14	\mathbb{R}
	Staphylinidae	Philonthus sp.	$\boldsymbol{0}$	$\boldsymbol{0}$	1	$\overline{0}$	θ	0.11	7.14	$\mathbf R$
	Curculionidae	Sclerocardius sp. Anomala	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	4	1	0.56	28.57	${\bf R}$
	Scarabaeidae	resplendens	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	1	$\boldsymbol{0}$	0.11	7.14	\mathbb{R}
		Chrysomya								
Diptera	Calliphoridae	chloropyga	15	49	72	7	$\mathfrak{2}$	16.2	64.28	D
	Syrphidae	Mesembrius sp.	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	3	$\mathbf{0}$	0.34	14.28	$\mathbf R$
	Muscidae	Musca domestica	1	$\overline{4}$	$\mathbf{1}$	$\mathbf{1}$	$\mathbf{0}$	0.78	28.57	\mathbb{R}
	Asilidae	Ommatius sp.	$\mathbf{0}$	$\boldsymbol{0}$	$\mathbf{0}$	$\mathbf{1}$	Ω	0.11	7.14	\mathbb{R}
		<i>Stenocoris</i>								
Hemiptera	Alydidae	southwoodi	$\mathbf{0}$	$\boldsymbol{0}$	$\mathbf{0}$	1	$\overline{2}$	0.34	21.43	\mathbb{R}
		Camponotus								
Hymenoptera	Formicidae	perrisii	1	$\mathbf{1}$	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	0.22	14.28	\mathbb{R}
	Formicidae	Crematogaster	θ	31	187	68	124	45.81	92.86	
		sp.								D
	Formicidae	Pheidole sp.	$\boldsymbol{0}$	$\mathbf{0}$	$\mathbf{0}$	12	15	3.02	35.71	$\mathbf D$
Isoptera	Termitidae	<i>Macrotermes</i> sp.	$\boldsymbol{0}$	$\mathbf{0}$	$\overline{0}$	19	Ω	2.12	7.14	\mathbb{R}
Total			22	128	388	175	182	100		

Table 2a: Abundance, frequency of occurrence and dominant status of insects invading slaughtered juvenile pig carcass

FSH = Fresh, $BLT = Bloated$, Ac. Dcy = Active decay, Ad. Dcy = Advanced decay, RA = Relative abundance, $FO = Frequency$ of occurrence, D. Status = Dominance status, $D = Dominant$, $R = Rare$

Order	Family	Genus/Species			Ac.	Ad.		RA	FO	D.
			FSH	BLT	Dcy	Dcy	DRY	$(\%)$	(%)	Status
Coleoptera	Histeridae	Hister monitor	\overline{c}	8	21	21	6	7.43	57.14	D
		Dermestes								
	Dermestidae	maculatus	$\mathbf{0}$	$\boldsymbol{0}$	$\overline{9}$	17	52	10.0	85.71	D
	Tenebrionidae	Zophosis sp.	$\boldsymbol{0}$	\overline{c}	6	13	18	5.0	85.71	D
	Cleridae	Korynetes analis	$\overline{0}$	$\overline{0}$	$\boldsymbol{0}$	1	$\overline{2}$	0.40	14.28	\mathbb{R}
	Staphylinidae	Philonthus sp.								
	Curculionidae	Sclerocardius sp.	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	3	0.40	14.28	${\bf R}$
		Anomala								
	Scarabaeidae	resplendens								
		Chrysomya								
Diptera	Calliphoridae	chloropyga	11	10	42	13	1	9.90	50.00	D
	Syrphidae	Mesembrius sp.	1	$\boldsymbol{0}$	\overline{c}	2	$\mathbf{0}$	0.64	21.43	\mathbb{R}
	Muscidae	Musca domestica	$\mathbf{0}$	$\boldsymbol{0}$	$\boldsymbol{0}$	3	θ	0.40	7.14	$\mathbf R$
	Asilidae	Ommatius sp.	$\boldsymbol{0}$	θ	$\mathbf{0}$	$\overline{0}$	1	0.13	7.14	\mathbb{R}
		Stenocoris								
Hemiptera	Alydidae	southwoodi	$\boldsymbol{0}$	$\boldsymbol{0}$	$\mathbf{0}$	$\mathbf{0}$	3	0.40	14.28	\mathbb{R}
		Camponotus								
Hymenoptera	Formicidae	perrisii								
		Crematogaster								
	Formicidae	sp.								
	Formicidae	Pheidole sp.	$\boldsymbol{0}$	$\mathbf{0}$	13	10	487	65.3	78.57	D
	Termitidae									
Isoptera Total		<i>Macrotermes</i> sp.	14	20	93	80	574	100		

Table 2b: Abundance, frequency of occurrence and dominant status of insects invading strangulated juvenile pig carcass

FSH = Fresh, $BLT = Bloated$, Ac. Dcy = Active decay, Ad. Dcy = Advanced decay, RA = Relative abundance, $FO = Frequency$ of occurrence, D. Status = Dominance status, $D = Dominant$, $R = Rare$

Figure 2: Abundance of insect Orders invading slaughtered and strangulated juvenile pig carcasses exposed to sunlight.

Diversity Indices of Insects Associated with the Decompositional Stages of Slaughtered and Strangulated Juvenile Pig Carcasses

Table 3a shows that while the active – decay stage of the slaughtered carcass had the highest insect abundance (388), the advanced stage had the highest species diversity (1.942) and richness (2.517). Diversity of invading insect species was least at the fresh stage (1.032) while species richness was least at the active decay stage (1.007). The bloated stage of the slaughtered carcass was observed to have the highest equitability value (0.786), presumably due to its low dominance (0.383).

For the strangulated carcass, the dry stage had the highest abundance (574) while the advanced stage showed similar trend as found in the slaughtered carcass, with species diversity value (1.801) and richness

(1.597). Species diversity was least at the dry stage (0.609) while richness was least at the bloated stage (0.668). Although the advanced stage of the strangulated carcass had the highest values for species diversity and richness, it also showed the least dominance (0.263) which presumably resulted to it having the highest equitability value of 0.866 (Table 3b).

For both carcasses, the bloated stages showed the highest values for evenness (0.659 and 0.856) for slaughtered and strangulated carcasses respectively, while their dry stages had the least evenness values (0.380 and 0.204) respectively (Table 3a and b). A comparison of species similarity between both carcasses indicated $a > 60\%$ species similarity on all sampled species and $> 80\%$ species similarity on the dominant species (Table 4).

Table 3a: Diversity indices of insects associated with the decompositional stages of slaughtered juvenile pig carcass

Table 3b: Diversity indices of insects associated with the decompositional stages of strangulated juvenile pig carcass

Table 4: Similarity index of insects sampled from slaughtered and strangulated juvenile pig carcasses

DISCUSSION

Low body weight carcasses have been reported to undergo faster decomposition (Chukwu *et al*., 2022; Giovanna *et al.,* 2022). The decomposition process of both carcasses used for this study lasted for 14 days, corresponding with the reports of Ana and Betina (2015) who used approximately 16 kg body weight pig. In addition, five (5) stages of decomposition were observed on both carcasses used in this study which is consistent with most documented findings (Ana and Betina, 2015; Ojianwuna *et al.,* 2019; Barton *et al.,* 2020; Julie-Eleonore and Shari, 2020; Michael *et al.,* 2022). But between the carcasses used for this study, some variations observed with regards to the length of decomposition stages as well as the observed postmortem changes may largely be attributed to the killing method used. The total decomposition period for both carcasses was 14 days. However, while both carcasses had $0 - 1$ day fresh stage and $2nd$ day bloated stage which buttresses the reports of Michael *et al.* (2022), the strangulated carcass clearly showed a shorter active decay (1 day) and advanced decay (1 day) periods than the slaughtered carcass that had 2 and 3 days, respectively. This apparently buttresses documented reports which showed that strangulated carcasses undergo faster decomposition than alternative killing methods (Ojianwuna *et al.,* 2019; Michael *et al.,* 2022; Chukwu *et al*., 2023). The physical postmortem changes observed on both carcasses were largely the same for both the fresh and bloated stages and aligns with the findings of Griffiths *et al.* (2020), Danisile and Samson (2021) and Michael *et al.* (2022). However, while the abdomen of the slaughtered carcass was observed to have busted slightly at the active decay stage leading to deflation of the body, the strangulated carcass was observed to exude some body fluid from the orifices at same stage. The postmortem characteristics of the slaughtered carcass agrees with the reports of Comstock *et al.* (2015) for the active stage without much maggot activities which might be attributed to high sun intensity, while the postmortem feature observed for the strangulated carcass follows the report of Michael *et al.* (2022) for strangulated carcasses at the stage. Similar postmortem features were largely observed at both the advanced and dry stages of both carcasses but notable was their whole mummification at the dry stage which can largely be attributed to high sun intensity and reduced insect activities. The shorter dry stage of the slaughtered carcass as observed in this study is presumed to be as a result of its ruptured abdomen as well as its initial higher insect activities which would have created a larger surface area for loss of moisture from the carcass as compared to the strangulated carcass with reduced insects' abundance and abdominal rupture.

Comparing the decomposition stages of both carcasses used for this study *vis-à-vis* insect abundance and succession, the slaughtered carcass overall attracted more insects (895) than the strangulated carcass (781) and apart from the dry stage, the slaughtered carcass was consistent on this trend for all the stages of decomposition.

Higher insect abundance on the slaughtered carcass over the strangulated carcass may largely be attributed to the killing method, as the bloodshed would have resulted to an increase in perception of decay odour by most insects invading the slaughtered carcass. Similar observation has been reported by Ana and Betina (2015), Verheggen *et al.* (2017). Generally, both carcasses were invaded by similar group of dominant insects at each decomposition stage with peak abundance and diversity at the active and advanced decay stages. With the exception of *Crematogaster* sp. which was specifically dominant on the slaughtered carcass, both carcasses had *H. monitor, D. maculatus, Zophosis* sp., *C. chloropyga,* and *Pheidole* sp*.* as dominant invading insects. Preference of *Crematogaster* sp. to the slaughtered carcass may be as a result of proximity of the carcass to the nest and/or the abundance of prey availability (including maggots) on the slaughtered carcass than the strangulated carcass. The invasion of these dominant insects as observed in the present study has been reported in literature (Ana and Betina, 2015; Danisile and Samson, 2021; Giovanna *et al.,* 2022; Michael *et al.,* 2022; Chukwu *et al*., 2023), suggesting their relevance in forensic investigations.

CONCLUSION

Cause of death or killing method affects the forensic characteristics of a cadaver and should be considered for precise application in medico-legal investigations. The killing methods (slaughtering and strangulation) in this study were observed to have affected insect abundance, length of decomposition stages as well as the observed postmortem changes on the carcasses. But overall, the similarity index shows that the dominant insects attracted to the cadavers were over 80% similar. In addition, the strangulated and slaughtered carcasses were observed to be wholly mummified at the dry stage. Hence, while the killing methods largely resulted in some variation in the length of decomposition stages, observable postmortem changes as well as insect abundance, the succession of invading insects were largely similar.

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