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Post-mortem interval of buried homicides in Okuku, Nigeria

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Abstract

Background Post-mortem interval estimation of carcasses buried in shallow graves is a grey area in Nigerian forensic investigations. Most of the investigations and court decisions on the time of death of concealed homicides are based on assumptions in Nigeria. Therefore, this study investigated the post-mortem interval of buried remains in Okuku, Cross River State of Nigeria using porcine models. This study also provided a model account of the pattern and time-line of decomposition of buried remains in Nigeria.

Results Four stages of decomposition were identified within the study period which includes fresh, bloat, active decay, and advanced decay stages. Features of bloat stage of decomposition include bloating of the animals and release of putrid odour. The active decay stage was characterized by the absence of maggots, bone exposure, and greyish discolouration of the body. The advanced decay stage of decomposition is characterized by adipocere formation, fungi activities, and bone exposure.

Conclusions Buried bodies do not completely skeletonize within 168 days in a typical Nigerian savannah region. Bloat stage started by the 7th day; the active decay stage started by the 14th day. The fresh stage of decay lasted up to 7 days; the bloat stage lasted for 14 days. The active decay stage lasted about 35 days, and the advanced decay stage started at about the 56th day and progressed until the end of the study.

Keywords Buried, Clandestine, Concealed, PMI, Shallow graves

Background

Post-mortem interval (PMI) estimation of buried bodies has been a challenge to forensic experts and crime investigators (especially the police) in Nigeria. In fact, over the years, most of the judicial decisions on alibi confirmation or prosecution of suspects for concealed or clandestine murder cases in Nigeria are based on speculations (Didia

and Olotu 2014). This is because of the scarce literature or data on buried bodies' post-mortem interval in Nigeria. Taphonomic data provides clue to the time at which a clandestine crime occurred and helps to corroborate the testimonies of alibis or crime suspects in the law court. In recent times, decomposition timeline studies (taphonomy) have provided a clear approach, understanding, and clue to the uncertainties and difficulties surrounding the confirmation of alibi or testimonies related to time of death (Myburgh et al. 2013; Teo et al. 2013; Pittner et al. 2020).

The decomposition rate of buried remains is influenced by certain factors such as gut microbes, manner of burial, body condition at burial, depth of grave, and soil physicochemical properties (Bachmann and Simmons 2010). These factors could either slow or accelerate the rate of decomposition of buried bodies. However, soil physical

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or chemical properties are distinct for each geographical region which makes the pattern or rate of decay of buried bodies to be different across regions or vegetations. The soil physical properties include texture, structure, temperature, depth, water-holding capacity, and bulk density (Phogat and Dahiya 2015), whereas the soil chemical properties include salinity, nitrogen, electrical conductivity, enzymes, sodium adsorption ratio, phosphorus, pH, base saturation, trace metals, carbon, organic matter, major cations, and cation-exchange capacity (Minnesota Pollution Control Agency 2023).

Okuku is a town located in the Guinea forest-savannah vegetation of Nigeria. This region is characterized by annual rainfall between 1000 and 1400 mm, tall grasses (about 2–4 m), and temperatures which can reach up to 50 °C with zero level of humidity. The vegetation has two distinct seasons — the dry and rainy seasons; however, the vegetation is predominantly covered by dry season. Most part of the dry season is characterized by dry land, scarce grasses, and lifeless trees. Dry season reaches its peak around December and February. The soil type predominant in this region is loamy, sandy, and humus soils (Makinwa 2018).

The aim of this study is to investigate the early and late visible post-mortem changes of buried porcine models in order to understand the possible factors that could influence the post-mortem interval of buried remains in Okuku, Nigeria. This study also aimed at documenting a model account of the decomposition pattern of concealed bodies in shallow graves in a typical Nigerian Savannah.

Methods

Ethical approval

This study was ethically considered and approved by the ethical committee of the Faculty of Basic Medical Sciences, University of Calabar, Nigeria, with certification number 079PHY3321 which is dated 24 March 2021.

Study location

This study was conducted at the Department of Anatomy Forensic Anthropology Research Facility (DAFARF) Okuku campus of the Cross River University of Technology (CRUTECH). The topography of the area where the research was conducted is a combination of hills and lowland areas. It is located between latitude 6°35'35"N to 8°38'01"E, having coordinates of 6042'N8°36'E with an elevation of 83.477 m above sea level.

Experimental animals

Four adult domestic pigs (*Sus scrofa domestica*) were used for this study. The animals were reared using standard pig feeds produced by Terratiga Limited, Nigeria. Animals were allowed to feed and drink water ad libitum

daily. Animals were procured from a pig farm located very close to the research location. The pigs were confirmed healthy by a veterinary doctor at the pig farm. The animals were two males and two females. Animals weighed between 30 and 40 kg, with perimortem body temperatures between 37 and 38.5 °C, recumbent lengths between 99 and 112 cm, chests circumference between 71 and 84 cm, and waists circumference between 59 and 79 cm (Table 1).

Experimental procedure

This study was an animal experimental study. The concept used for the research procedure was standard taphonomic procedures. The animals were sacrificed, and death was confirmed when there was no heart beat recorded by the stethoscope and via the pupillary reflex. Data on the observable decomposition changes of the animals were collected by the researchers.

However, the first procedure involved in carrying out this experiment was to ascertain the body statistics of the animals in order to confirm if the animals are fully matured. This was immediately followed by recording the perimortem body temperatures of all the animals. Animals were euthanized via exposure to carbon dioxide in a closed room (gross anatomy laboratory). The post-mortem body temperatures of the animals were also measured in order to study the early decomposition changes (especially algor mortis). This was carried out inside the laboratory so that harsh environmental factors will not affect the temperature readings. Afterwards, the animals were buried in a one foot depth shallow grave at the research facility. All the animals were buried at the same time and were buried in a manner that they had direct contact with the soil. The animals were covered with a wooden board so as to enable the researchers to open it up and observe the decomposition stages at intervals. The graves were opened at intervals of 7, 14, 21, 28, 42, 56, 84, 112, and 168 days, respectively, so as to study the decomposition changes such as colour discolouration,

Table 1 Body statistics of the animals

Body statistics	PIG 1	PIG 2	PIG 3	PIG 4
Weight (kg)	39.4	32.5	31.5	30.0
Perimortem temperature (°C)	38.0	37.0	38.0	38.5
Recumbent length (cm)	112.0	104.0	104.0	99.0
Chest circumference (cm)	84.0	76.5	80.0	71.0
Waist circumference (cm)	79.0	74.0	70.0	59.0
Body temperature at death (°C)	39.0	38.9	41.4	40.6
Atmospheric temp. at death (°C)	30.0	30.0	30.0	30.0
Time of death (s)	9.12	9.40	9.50	10.18

hair loss, body mass degradation, caving in of the eyes, adipocere formation, and discharge of body fluids.

The grave sites were marked with sign post at the head of the buried pigs. The perimeter of the forensic sites (9.39 m in length and 3.38 m in width) was secured and marked clearly with forensic tape to avoid any human interference. The soil pH, moisture, and water percolation were obtained daily throughout the study period using the Smart 4-in-1 soil survey instrument. Soil analysis was carried out on the soil sample to ascertain the amount of salt in the soil. Mycology analysis was used to identify the pathogen on the skin of the animals during the study period.

Method of data collection for soil salinity, moisture, and water percolation

The apparatus used to record the soil salinity, moisture, and water percolation was the Smart 4-in-1 soil survey instrument produced in 2019 by Yancheng Kecheng Optoelectronic Technology Company Limited, Yancheng, Jiangsu, China (Fig. 1).

The (pH/°C) yellow selector switch was pushed on the back of the instrument to select the pH position. The probe was vertically inserted into the soil for about 4–7 cm depth, and the readings were obtained from the liquid content display (LCD) of the instrument. We ensured that the probe was not inserted beyond 7 cm deep into the soil to avoid damage to the roots of the plant.

Method of soil analysis

Reagents

The reagents included distilled water and buffer/standard solutions of salinity purchased from the central drug market, Onitsha in Anambra state, Nigeria. These buffers were prepared by dissolving standard buffer tablets or by preparing a 0.010-M potassium chloride reference

solution (by dissolving 0.7455-g potassium chloride in distilled water in the absence of carbon (IV) oxide).

Apparatus

The apparatus used to analyse the soil sample was a salinity metre (with model number AZ8371 and manufactured in 2020 by Huanyu, China) which has a glass electrode and a reference electrode (Fig. 2). Sometimes, these two electrodes embody into one unit called a combination electrode.

Procedure

Put salinity metre in water (1:1 soil to water ratio) to 20 g of air-dried soil (passed through 2-mm sieve) in a 50-ml beaker, add 20 ml of distilled water, and allow to stand for 30 min with occasional stirring with a glass rod. Divide the suspensions into three parts. Then, insert the electrodes into the buffer solutions having salinity values close to that expected of the soil and metre needle adjusted to read the buffer salinity. Ensure to take great care when inserting the electrodes into the solution as the electrodes are fragile and can easily break. The electrodes should extend at least 2 cm into the solution.

Then, remove the electrodes, rinse with distilled water, and insert into soil suspensions (1), (2), and (3) (with the



Fig. 1 Smart 4-in-1 soil survey instrument



Fig. 2 Salinity metre

electrode into the clear supernatant solution and the glass electrode into the sediment if the electrodes are supplied separately). The sodium chloride (NaCl) metre readings were recorded to the nearest 0.05 unit (electrodes should be rinsed between each reading). At the end of the experiment, the electrodes were cleaned with distilled water and a beaker of distilled water lowered to them.

Method of fungi identification (mycology analysis)

Sample collection

The process of sample collection is termed skin scraping. The infected site was scraped using a sterile surgical blade and divided into two portions. The first portion was used for microscopy, whereas the second part was cultured.

Microscopy

The first portion was placed on a grease-free microscope slide. Since the sample was a skin sample and a soft tissue, there was an addition of 10% potassium hydroxide (KOH) on the scraped sample on the slide so as to digest the tissue (usually within 5 min) so that the causative agent of the infection can be viewed under the microscope. The fungi elements were viewed microscopically by using $\times 10$ objective lens to focus and $\times 40$ to view. The features seen were branched and segmented hyphae.

Culture

The second part was cultured on a culture plate so as to identify the particular fungi. The sample was cultured on a fungi media specifically a Fabraoud dextrose agar (FDA). Then, with the help of a sterile forcep, the scraped skin was picked and stabbed onto the culture medium. Then, the culture was incubated at room temperature (25–30 °C). After 1 or 2 weeks, the colony is expected to have grown and ready identification.

Identification of pathogen

A loop full of the grown culture was collected and placed on a grease-free slide. Lactophenol cotton-blue reagent was added to the slide and then viewed microscopically. Here, the features were clearly demonstrated on the microscope, and the particular pathogen was able to be identified.

Experimental control/precautions

The researchers ensured that the pigs were in good condition, and the food taken 2 weeks before the experiment did not contain any poisonous or alcoholic substances. The pigs were acquired from a pig farm located nearby the research facility in order to ensure quick acclimatization. The mercury part of the thermometer was cleaned

after inserting it inside the anus of the pigs with a dry cotton wool to ensure accuracy of the readings, and we also avoided parallax errors when taking readings from the analogue instrument (weighing scale).

Statistical tool and method of data analysis

Qualitative and quantitative data were curated from the decomposition changes of the animals. The qualitative data were descriptively represented in tables and figures. The quantitative data were analysed using Statistical Package for Social Science (SPSS) International Business Machines (IBM) series version 25.

Duration of the study

This study was carried out within a period of 168 days (from February 2020 to August, 2020).

Results

Animal body statistics

The body statistics revealed that the experimental animals were fully matured, whose body weight ranged between 30 and 40 kg. During the sacrifice, one of the animals (pig 3) struggled for survival (with discharge of faecal matter and urine) which led to elevated body temperatures at death (Table 1). The other animals died slowly during the sacrifice and maintained the body temperatures as shown in Table 1. Consequently, pigs 1 and 2 had the least standard deviations ($SD = 1.17$) followed by pig 4 ($SD = 1.7$) showing that the body temperatures of these three pigs were more consistent and reliable when compared to the standard deviation of the body temperatures of pig 3 ($SD = 2.2$). This means that the rate of change in body temperatures of pigs 1, 2, and 4 was not much when compared to rate of change in body temperature of pig 3. Pig 3 had an inconsistent body temperature, and this may have affected its autolytic process (Table 2).

Table 2 Descriptive statistics of the body temperatures of all the animals

Pigs used for the experiment	Mean	N	Std. deviation
PIG 1	36.8846	13	1.16751
PIG 2	36.8385	13	1.17085
PIG 3	37.6462	13	2.15778
PIG 4	38.1538	13	1.73523
Total/mean value	37.38078	52	1.55784

N the number of readings observed 6 times (per minute) for the first 1 h and 7 times (hourly) for the second 7 h

Visible post-mortem changes

The visible post-mortem changes observed from the animals showed that the animals reached the early skeletonization stage of decomposition. On the first day after animal sacrifice, the animals appeared fresh with no visible post-mortem changes. This stage of decomposition is referred to as the fresh stage of decomposition.

On the 7th day, the visible post-mortem changes observed on the animals showed that almost all the animals were at the bloat stage of decomposition (Fig. 3; Table 3). However, there was slight bone exposure of some of the body structures of two of the animals. This stage was also characterized by brownish discolouration of the body. Though the hairs remained intact, the skin of the limbs of the two animals whose bones were exposed appeared greasy. Few maggots were also seen at the trunk of the animals, though maggot activities were more at the regions with exposed bones.

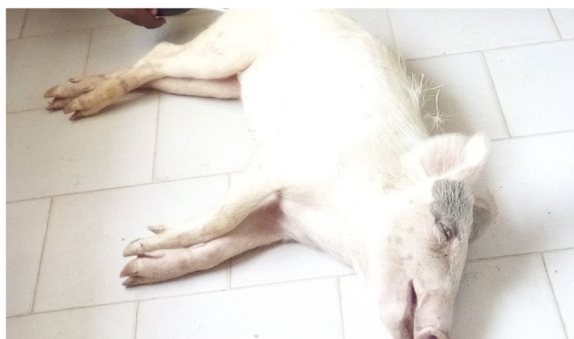


Fig. 3 *Sus scrofa domesticus* immediately after sacrifice (day 1 — fresh stage)

The active decay stage of decomposition was observed to start on the 14th day after death. As at the 28th day, all the animals were actively decaying (Figs. 4, 5, 6 and 7). The active decay stage is characterized by the absence of insect activity (Fig. 8); bone exposure of the head and limbs structures (Table 3); greyish discolouration of the body (Fig. 5; Table 3); greasy appearance of the skin surrounding the limbs (Table 3); the presence of adipocere formation on some of the pigs (Table 3); and the hairs remained intact (Figs. 5 and 6).

The advanced decay stage of decomposition was observed to start on the 84th day. The characteristic features of this stage include greasy appearance of the skin (and adipocere formation), gross spread of fungi on the body structures of the animals, bone exposure of more regions of the body, the absence of insects, and greyish appearance of the body (Fig. 9; Table 4). These changes gradually progressed to early skeletonization as at the later days of the study, with more exposed bones (Fig. 10; Table 4). As at the last day of the study, the fungi activities reduced (Fig. 10).



Fig. 4 Day 14 — early active decay stage

Table 3 Days 7–28 post-mortem visible changes

Day	Head & neck Visible changes	Trunk Visible changes	Limbs Visible changes
Day 7	Brownish discolouration of the structures of the head and neck region with bone exposure of the jaw	Trunk appears bloated Brownish discolouration of the hair and skin with greasy appearance Hairs remained intact Few maggot present	Rigor mortis of the limbs persisted Dark-brown discolouration with bone exposure of fore and hind limbs of one of the animals, whereas the limbs of other animals discoloured to brown with greasy skin and hairs
Day 14	Adipocere formation started on the ventral aspect of the neck Jaw bones became more visible Greyish discolouration of all aspects of head and neck	Adipocere formation started on the ventral aspect of the trunk Greyish discolouration of all aspects of the trunk	Bones became more visible The skin appeared greasy with adipocere formed on forelimbs Dark-brown discolouration of all limbs
Day 21	No visible changes Insects and fungi absent	No visible changes Putrid odour persisted Insects and fungi absent	No visible changes Insects and fungi absent
Day 28	No visible changes Insects and fungi absent	No visible changes Insects and fungi absent Adipocere formation progressed	No visible changes Insects and fungi absent



Fig. 5 Day 21 — active decay stage



Fig. 8 Day 7 — bloat stage



Fig. 6 Day 28 — active decay stage



Fig. 9 Day 84 — advanced decay stage/skeletonization



Fig. 7 Day 42 — late active decay stage

Soil analysis

The result showed that the soil contained a small amount of sodium chloride and at neutral potential of hydrogen (pH) level (Table 5).

Mycology analysis

The result identified dermatophytosis (superficial mycoses) as the pathogen on the skin of the pigs (Table 6).

Table 4 Days 42–168 post-mortem visible changes

Day	Head & neck Visible changes	Trunk Visible changes	Limbs Visible changes
Day 42	Adipocere formed on the dorsal aspect of the neck No visible decomposition changes	Adipocere formation progressed No visible decomposition changes	Adipocere formed on the skin of hind limbs No visible decomposition changes
Day 84	Fungi activity present on head and neck structures Insects absent	Fungi activity on the trunk The skin appeared greasy and darker Insects absent	Fungi activity present on hind limbs Insects absent
Day 168	Gradual decomposition of structures with gradual spread of fungi Structures appeared greasy	Fungi activity spread over the thorax Carrion appeared greasy but no visible decomposition changes observed	Less fungi activity on the hind limbs Fore and hind limbs appeared greasy with no visible decomposition changes



Fig. 10 Day 168 — late advanced decay stage/early skeletonization

Table 5 Soil chemical properties

Compound	Amount
Sodium chloride (mg/kg)	12.50
pH	7.0

Discussion

The increase or inconsistency in body temperature of one of the animals could have sped up its autolytic activities. This inconsistency could be attributed to the cause of death because the result showed that this animal (pig 3) struggled so much before it was eventually

Table 6 Mycology analysis result

Test	Results
Morphology features	Branched and segmented hyphae
Culture features	Hyphae undisturbed
Pathogen identification	Dermatophytosis

confirmed dead, thereby elevating its body temperature. This is because studies have reported that mode of death is a factor that accelerates the rate of decomposition (Onyejike et al. 2022a, b).

Visible post-mortem changes are the physical or macroscopic changes that occur on a dead body from the time of death to the final stage of decomposition (skeltonization). There are clear characteristics that distinguish each stage of decomposition from the other. These changes occur at distinctive timelines, and it aids a forensic taphonomist to estimate PMI. The data on the PMI of the buried animals in this study showed that each of the notable stages of decomposition took a longer timeline compared to bodies above soil surface in similar climate which was reported by Fulp (2021) and Onyejike et al. (2022a). The decomposition process of the buried animals was initially enhanced but later became slow. This could be as a result of pre-burial exposure to high environmental conditions, whereas the slow rate of decomposition in the graves could be as a result of the absence of dipter activities. Bachmann and Simmons have also reported that pre-burial exposure to arthropod activities could enhance and accelerate decomposition rate of buried bodies (Bachmann and Simmons 2010). In addition, recent articles have also noted that exposure of carcass to dipter species positively influences its decomposition rate (Matuszewski et al. 2020; Probst et al. 2020). However, Prangnell and McGowan (2009) reported that low temperatures in graves contribute to slower decomposition rate of buried bodies.

At the 7th day, the animals were at the bloat stage. Animals bloated as a result of the gases generated by degradation of macromolecules in the body via active body digestion caused by autolysis and bacteria (putrefaction). The maggots observed to be inside the graves on the 7th day could have contributed to the speedy breakdown of the fleshy body parts of some of the animals. The putrid odour from the shallow graves attracted the insects (fly) which laid eggs on the sites of the odour. These eggs eventually hatched into larvae which found its way through the porous loamy soil to the buried animals.

By the 14th day after death, the larvae or pupae of the insects were no longer within the graves. In addition, the putrid odour was no longer intense as it was on the 7th day, and the temperatures were very hot (about 41 °C). The extreme temperatures could have impaired the survival of the insects even though the putrid odour remained intense on the 14th day. This is because it has been reported that extreme temperatures prevent the survival of dipteran species (Biswas 2012; Rao 2013).

The advanced decay stage of decomposition occurred within a longer timeline when compared to the timeline of the different stages of decomposition reported in this study. Adipocere formation could be a factor that inhibited the advanced decay stage because adipocere formation was more visible during this stage of decomposition. This is because Hayman (2021) reported that adipocere formation is a factor that delays decomposition rate of carrions especially buried bodies. In addition, fungi activities also contributed insignificantly to the decomposition process of the buried pigs because it started at about the 84th day post-mortem (at the beginning of rainy season) and progressed until the last day of the study. It can be deduced from this investigation that fungi growth is influenced by rain infusion because fungi activities were first reported at the onset of rainfall. More so, Myburgh et al. (2013) and Gaudry (2010) have also noted that rain infusion improves the survival of fungi which are the primary decomposers of buried bodies.

The entire timeline of decomposition of the buried animals within the study period (from fresh stage to late advanced decay and/or skeletonization stage) is shorter than the timeline reported in some literatures (Phogat and Dahiya 2015). This may be because of the periodic opening of the graves which may have introduced environmental communities (decomposers) into the graves. Therefore, this study recommends that subsequent studies should be carried out without opening of the graves at intervals.

The primary decomposers of the buried pigs in this study could be soil and gastrointestinal faunal species. This is corroborated by the report by Carter and Tibbett (2008) which noted that the decomposition process of

buried bodies is influenced by microbiological activity, with population from both gastrointestinal tract and surrounding soil. This study recommends that further studies in this region be started at the onset of rainy season and then progressed into the dry season to investigate the effect of rain infusion and fungi activities on the decomposition process and timeline of buried bodies.

The amount of salt in the soil is very low, and as such did not significantly influence the rate of decomposition. The neutral pH also shows that chemical factors of soil did not affect the results obtained from this study. This corroborated with a report which noted that soil physico-chemical properties such as soil temperature, moisture, pH, and salt do not influence the post-mortem interval of carrions on soil surface of a typical Guinea forest-savannah vegetation of Nigeria (Onyejike et al. 2022b). In addition, dermatophytosis was found at the skin of the animals over a short period. Dermatophytosis is caused by a group of fungi that usually remain localized to the superficial layers of the nails, skin, or hairs. Despite their propensity to infect the exterior aspects of the host, the dermatophytes prefer a warm, moist environment for growth, and as a consequence, infections are common in tropical regions. This explains why the fungi activities were within a short period during the study, due to the extreme climatic conditions of the savannah region of Nigeria.

Conclusions

Five stages of decomposition were identified within the study period which includes fresh, bloat, active decay, advanced decay, and early skeletonization stages. These were not distinctively different from each other because the pattern of decay process is dissimilar to that identified in other literatures for bodies decaying on soil surface. Complete skeletonization of buried bodies does not occur within 168 days in this region of Nigeria. However, without opening of graves at intervals, decomposition of buried bodies in this region may not reach the skeletonization stage.

The fresh stage of decay could last up to 7 days if animals are not exposed to pre-burial environmental conditions. The bloat stage started by the 7th day and lasted for 7 days. The active decay stage started on the 14th day and lasted about 35 days. The advanced decay stage started at about the 56th day and progressed until the 168th day. By the 6th month (168th day), the animals were in early skeletonization stage.

The primary decomposers are gastrointestinal and soil faunal communities. Insects, fungi, and rain infusion played insignificant roles in the decay process of the buried pigs during the period of this study.

Abbreviations

CRUTECH	Cross River University of Technology
DAFARF	Department of Anatomy Forensic Anthropology Research Facility
FDA	Fabraoud dextrose agar
IBM	International business machines
KOH	Potassium hydroxide
LCD	Liquid crystal display
N	Number of readings observed
NaCl	Sodium chloride
pH	Potential of hydrogen
PMI	Post-mortem interval
SD	Standard deviation
SPSS	Statistical Package for Social Sciences

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Disclaimer

This research article is an original article. It has not been submitted for review to another journal and has not been published in any journal or conference proceedings.

Authors' contributions

This work was carried out in collaboration of all authors. Author ODN wrote the protocol, acquired the animals, carried out the experiment, and wrote the first draft of the manuscript. Author EUG designed the study and assisted author FVA to supervise the experiment. Author FVA supervised the experiment. Author OIM managed the literature searches. The authors read and approved the final manuscript.

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Availability of data and materials

The datasets generated during and/or analysed during the current study are available within the text.

Declarations**Ethics approval and consent to participate**

The ethical approval was obtained from the ethical committee of the Faculty of Basic Medical Sciences, University of Calabar, Calabar, Nigeria. The certification number is 079PHY3321 dated 21st March 2021. All the authors gave full consent to participate in the study.

Consent for publication

Authors enlisted in this draft article have given full consent for this draft article to be submitted under review in the *Egyptian Journal of Forensic Sciences*.

Competing interests

The authors declare that they have no competing interests.

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