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Comparative Study of Knowledge, Risk factors, and Prevalence of Hepatitis B virus Infection Among Donkey Butchers and Herders in Ebonyi state, Nigeria

Peter Onochie Elom¹, Alfred Friday Igwe Una², Chika N Onwasigwe³

¹Department of Community Medicine, Ebonyi State University, Abakaliki, Ebonyi, State, Nigeria, 08037740075, opeterelom@gmail.com

²Department of Community Medicine, Alex Ekwueme Federal University Teaching Hospital, Abakaliki, Ebonyi State, Nigeria. bobuna2000@yahoo.com

³Department of Community Medicine, University of Nigeria Teaching Hospital, Enugu, Enugu State, Nigeria; 08033364778, chikanma60@gmail.com

Corresponding to: Dr Peter Onochie Elom.

Department of Community Medicine, Ebonyi State University, Abakaliki, Ebonyi, State, Nigeria, 08037740075.

Email: opeterelom@gmail.com.

Phone +2348037740075.

DOI: [10.21608/AJGH.2024.245381.1045](https://doi.org/10.21608/AJGH.2024.245381.1045).

Submission date: 3 December 2023.

Revision date: 7 January 2024.

Acceptance date: 16 January 2024.

Published online: 22 January 2024.

Abstract

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Aims: The study compares the knowledge, risk factors, and prevalence of HBV infection among donkey butchers and herders in Ebonyi State, Nigeria. Donkey butchers and herders comprise a high-risk population that can be controlled to minimize transmission of hepatitis B virus infection with its socioeconomic losses.

Subjects, Materials, & Methods: This was a comparative cross-sectional study among donkey butchers and herders in selected abattoirs in Ebonyi State. A systematic sampling method was used to select 125 respondents in each donkey handling group from 3 Local Government Areas of Ebonyi State. A structured interviewer-administered questionnaire was used to collect data from respondents on knowledge and risk factors. Prevalence of HBV was measured by serological screening tests of respondents' blood for identification of HBV surface antigen. Data were analyzed using IBM-SPSS Statistics version 26.0. The level of significance was set at $p < 0.05$.

Results: There were poor grades of HBV knowledge among the two donkey handling groups (herders, 94.4%, and butchers, 87.2%, respectively). Risk factors for herders include cigarette smoking, drug addiction, and casual sexual intercourse, whereas only duration of stay was associated with the butchers' handling group. The combined HBV prevalence of the donkey handling groups in Ebonyi State was 11.6%, donkey butchers 8.0%, and herders 15.2%.

Conclusions: The study showed poor knowledge grades among the donkey butchers and herders. Some risk factors were associated and predictors of HBV transmissions. The high HBV prevalence among the donkey handling groups has apparent public health implications.

Keywords: Butchers, Herders, Hepatitis B virus infection, Abattoirs, lairage, Hepatitis B surface antigen, Ebonyi State, Nigeria, Comparative study, Prevalence.

Introduction

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Viral Hepatitis can be defined as liver inflammation caused by one or more of the five main hepatic viruses: A, B, C, D, and E.[1] Although these viruses display similar symptoms and the potential to cause liver disease to varying degrees, they differ significantly regarding epidemiology, prevention, diagnosis, care, and treatment. Knowledge of viral hepatitis remains low among Nigerians despite being a leading infectious cause of death each year. As a consequence, most of the estimated 20 million Nigerians living with viral hepatitis B or C are undiagnosed, increasing the likelihood of future transmission to others and placing them at greater risk for severe, even fatal health complications such as liver cirrhosis and liver cancer (hepatocellular carcinoma).[2,3] Viral hepatitis is a major global health problem, with more than 400 million patients chronically infected, causing over 1.4 million deaths per year. Nigeria is among the countries with a high burden of viral hepatitis, with a Hepatitis B Virus (HBV) and Hepatitis C Virus (HCV) prevalence of 11% and 2.2%, respectively.[3]

Despite the high prevalence of viral Hepatitis B and C infections in Nigeria, the low level of awareness of the diseases contributes to the increasing burden of chronic infections and advanced liver disease.[3] Hepatitis B and C infection frequently affect people of various socio-economic groups because of poor access to appropriate healthcare, unhealthy lifestyles, and cultural practices. While progress has been made in improving prevention and treatment interventions in HIV and AIDS, TB, and malaria due to World Bank funding, not much has been done to improve access to hepatitis prevention and treatment services.[4]

In this study, emphasis is placed on viral Hepatitis B, which is a blood-borne viral infection that attacks the liver and can cause both acute and chronic disease. It is an important zoonotic occupational hazard to health and slaughterhouse workers.[5,6] A Significant number of infected individuals get the infection through unprotected sexual contact with an infected person; however, the infection can be prevented by vaccination. Over 90% of Hepatitis B virus infections exist chronically and show no signs and symptoms unless during acute phases when

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symptoms resemble malaria attack of generalized body weakness, fever, headache, joint pains, and occasionally jaundice. Infection with Hepatitis B virus infection has been reported to have attained a silent epidemic.[7,8,9]

The distribution of Hepatitis B Virus infection by sex is 62.6% of males and 37.4% of females.[10] Infections are most common among 21-40-year-olds, although substantial perinatal and childhood transmissions do occur.[11]

Medical personnel, especially surgeons and dentists, are at the most significant risk of infection. At the same time, other healthcare workers, butchers and other abattoir workers, commercial sex workers, and drivers are also at considerable risk of infection.[12] About 1% of persons living with HBV infection (2.7 million people) in Nigeria are also infected with HIV.[13] Conversely, the global prevalence of HBV infection in HIV-infected persons is 7.4%.[12] Hepatitis D virus exists as a co-infection with hepatitis B virus infection.[14,15,16].

Risk factors for transmission in Nigeria include sexual intercourse, local circumcision, local uvulectomy, scarification, tribal marks, surgical procedures, body piercing, home birth, and blood transfusions. Control of infections is structured along the continuum of care for persons with chronic viral Hepatitis B from initial assessment of the stage of disease and eligibility for treatment to initiation of first-line antiviral therapy and monitoring for disease progression, toxicity, and hepatocellular carcinoma and switch to second-line drugs in persons with treatment failure using antiviral drugs.[16] The control measures are intended for use across age groups and adult populations. [Appendix I]

The prevalence of HBV infection is higher in WHO Western Pacific and African Regions, where 6.2% and 6.1% of the adult population, respectively, are infected.[17,11] Nigeria accounts for about 11.2% of the infection, while Ebonyi State's prevalence is about 8.9%.[18] The virus can survive outside the body for at least seven days, during which it can cause infection if it enters the body of a person not protected by the vaccine. Its incubation period is 75 days on average but can vary from 30 to 180 days.[2,12, 18,19]

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Hepatitis B virus infection in butchers can result in untold hardship to the families, both for the butchers and herders. Arising from this context is the issue of chronicity, during which the workers lose hours of life and economic loss due to the inability to carry out their business. In addition, the morbidity nature of the infection is also associated with further loss of income due to frequent hospitalizations, cost of investigations, and medications (no Government subsidy as of now). Due to the asymptomatic nature of the infection, the horizontal spread will be enhanced, further increasing prevalence, disease progression, and mortality due to fulminant Hepatitis and cancers.[20]

Source of HBV infection [2]

HBV belongs to the family Hepadnaviridae and the genus Orthohepatodnavirus. It is the only hepadnavirus causing infection in humans. It cannot yet be grown in an artificial medium but can be transmitted to certain primates, such as cows or chimpanzees, which it can replicate. It is a resilient virus that can exist on almost any surface for about one month. Sodium hypochlorite 0.5% (1: 10 household bleach) destroys the HBV antigenicity within 3 minutes, but the virus is stable at minus 20 degrees centigrade for about 20 years. HBV-infected cells produce multiple types of virus-related particles. Electron microscopy of partially purified preparations of HBV shows three kinds of particles. Historically, hepatitis B surface antigen (HBsAg) was formerly called Australia antigen because it was first described in the serum of an Australian aborigine in 1963. Sub-Saharan Africa is a region of high endemicity with an average carrier rate of 10 - 20% in the general population. Seventy to 95% of adults in the Sub-Saharan have at least one marker of HBV. In West Africa, it has been estimated that 40% of children will be infected by the age of two years and above 90% by the age of ten years. The chronic carrier rate is 20% in these children. A chronic carrier rate above 7% in a population is classified as hyper-endemic. Studies in Nigeria showed HBV carriage rate in the 9 to 39% range. Transmission of the virus has been discussed earlier.

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Diagnosis of HBV infection

1. Clinical Evaluation: Patients' detailed history and physical examination are required. Alcohol, drugs, and a history of other risk factors should be taken. Physical examination is conducted to evaluate for features of chronic liver disease such as jaundice, hepatomegaly, splenomegaly, and GI bleeding. The presence of ascites is highly suggestive of decompensated liver cirrhosis. These patients should be considered for treatment prioritization and referred for specialized care.[3]

2. Laboratory diagnosis[3,21] of Hepatitis B virus infection focuses on detecting the Hepatitis B surface antigen, HBsAg, in a susceptible client's serum. The World Health Organization recommends testing all blood donations for Hepatitis B virus to ensure blood safety and avoid accidental transmission to people receiving blood products. Acute HBV infection is characterized by HBsAg and immunoglobulin M (IgM) antibodies to the core antigen, HBcAg. During the initial phase of infection, patients are also seropositive for Hepatitis B e-antigen (HBeAg). The presence of HBeAg indicates that the blood and body fluids of the infected individual are highly infectious. Chronic infection is characterized by the persistence of HBsAg for at least six months (with or without concurrent HBeAg). The persistence of HBsAg is the principal marker of risk for developing chronic liver disease and liver cancer (hepatocellular carcinoma) later in life.[12] Please find the following algorithm [Appendix 3] to confirm the diagnosis and assess the patient for HBV management.[20] (Appendix III)

3. Interpretation of Hepatitis B Serologic Tests /Markers: (Appendix IV).

There are four interpretations:

1- May be recovering from acute HBV infection.

2- The patient may be distantly immune, and the test is not sensitive enough to detect a shallow level of anti-HBs in the serum.

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3- May be susceptible to a false positive anti-HBc.

4. May be chronically infected and have an undetectable level of HBsAg present in the serum (Occult HBV)

*In areas where the HBV serology panel is inaccessible, a repeat HBsAg test is required in 6 months. Where positive, chronic Hepatitis B is confirmed. Chronic Hepatitis B (CHB) is defined as the persistence of HBsAg for more than six months or chronic liver disease attributable to HBV infection. In persons with CHB, a positive HBeAg result usually indicates the presence of active HBV replication and high infectivity. Post-vaccination testing, when recommended, should be performed 1-2 months following dose number three. Patients who are diagnosed with active or chronic HBV infection are referred to internal medicine (Gastroenterology unit) for further management. At the same time, those who are negative receive immunization (at the Institute of Child Health and Development, AE-FUTHA, my training center) as earlier scheduled.

Donkey meat has become the most commonly sold meat in southeast abattoirs, especially in southern Nigeria, including Ebonyi State.[22] A few years ago, donkeys were shown to harbor HBV and HCV similar to the ones that affect human beings.[23,24] As in conventional abattoirs, there are many herders and butchers of donkeys in the state; the population of consumers is also significant.[25] The populations (butchers and herders) are exposed to physical and biological hazards, respectively.[26] Essentially, donkey butchers are more exposed to physical hazards such as knife cuts, punctured wounds, head injuries, rashes, and accidents.[5,6,26] On the other hand, the donkey herders are exposed to biological hazards linked to risky behavioral practices such as persistent unhygienic practice habits (including poor hand/personal hygiene, filthy environment, lack of standard lairage setting and inadequate health officials supervision), cuts on hands, sharing of razors during cultural practices, tattooing and risky sexual behaviors.[5,6] The choice of comparing both populations stems from the fact that they

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share a uniform environment (donkey market) and trade on the same animal with different but related modes of infection transmission.

Previous studies done on HBV infection among butchers showed that slaughterhouse workers (SHWs) are at higher risk of infectious hepatitis that can be multifactorial and can be evaluated for viral, bacterial, and parasitic organisms.[2] Published research findings were very scarce concerning studies specific to the prevalence of Hepatitis B virus infection among donkey butchers or donkey herders in regions of the country, including South East and Ebonyi State in particular, hence the need for this study. Only some studies done in Ebonyi State were silent on the viral composition of HBV in donkeys and their handlers. There is increasing patronage for meat in some parts of Nigeria.[25,27,28] and perceived poor knowledge of HBV infection among these populations (butchers and herders) will further enhance the spread of infection.[7] Through public awareness creation, consumers who comprise a significant population make the study worthwhile for controlling HBV infection in the country.

subjects, materials, and methods

Study Area: Ebonyi State is one of the states in the Eastern Region of Nigeria; it was created out of the former Abia and Enugu States on the first day of October 1996 by the then Federal Military Government of General Sani Abacha. It lies at the coordinates of $6^{\circ}15'N$ and $8^{\circ}05'E$, covering an area of 5,533km [29], bounded north by Benue State, south by Abia, East by Cross River, and West by Enugu States. The State has a population of about 3.5 million, according to the projection of the 2006 National Census [29], which recorded Ebonyi's population as 2.1 million. Ebonyi State has 13 Local Government Areas, with the capital at Abakaliki, inhabited mainly by Igbo-speaking tribes. The majority of the state's indigenes are farmers. In 2016, the human development index (HDI) was put at 0.434, ranked 24th out of 36 states of the country.[29,30] There were four major markets in the

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state where donkeys were sold and slaughtered, and their meat was sold to consumers. However, no health authority regulated animal meat procurement, slaughter, processing, and selling.

Inclusion criteria

1. Healthy donkey butchers and herders in Ebonyi State donkey markets.
2. Butchers and herders of donkeys who have worked in the markets for at least 12 weeks (3 months) before the commencement of the study.
3. Butchers and herders of donkeys who have a place to display their trade in the markets.

3.4.2. Exclusion criteria

1. Intravenous drug abuse or indiscriminate injections among donkey butchers and herders.
2. Donkey butchers and herders who are diagnosed with hepatitis B, including cirrhosis and hepatocellular carcinoma.
3. Donkey butchers and herders who refused to sign the consent form.

3.5. Sample size determination

The following formula was used to determine the sample size, which is appropriate for comparing two proportions.[\[31,32\]](#)

$$n = \frac{[Z\alpha + Z\beta]^2 \times [P_1(1 - P_1) + P_2(1 - P_2)]}{}$$

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$$[P_1 - P_2]^2$$

Where.

n = minimum sample size in each group

$Z\alpha = 1.96$, the critical ratio or standard average deviates at a significant level of 5%

$Z\beta = 0.84$, the critical ratio or standard average deviates at the desired power of 80%.

P_1 = the proportion with events in group 1, that is, the proportion of herders who are Hepatitis B virus-positive after laboratory screening tests, taken as 26.2% (0.262)

P_2 = Proportion with an event in group 2, the proportion of donkey butchers that are Hepatitis B virus-positive after laboratory screening tests, taken as 11.6%. A total of 125 minimum samples in each group was calculated.

Sampling technique

A systematic sampling technique randomly selected the desired sample size in all the markets. The list of donkey butchers and herders in each market formed the sample frames to determine the number of participants selected by proportionate allocation. The proportionate sample sizes from each of the four markets were calculated by multiplying the individual market population of butchers by the total sample size and divided by the total population of butchers ($a \times n/T$, $b \times n/T$, $c \times n/T$, $d \times n/T$), corrected to the nearest unit decimal. The sampling interval, $k = 5$, was found by dividing the individual market sample population by proportionate sample sizes to ensure a systematic and equal chance of being selected. The starting point for each selection was chosen through simple random sampling of numbers 1 to 5 of the sampling frame. The 4th person in the list was chosen first; then,

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every k^{th} person from the sample frame was selected by $x + k$, $x + 2k$, $x + 3k$ until the sample size was completed.[31,32]

Study Instrument:

These comprised: 1. Questionnaires and 2: laboratory reagents/HBV test kits. The questionnaire was adapted from previous studies.[33,34,35] There were five parts (sections) in the questionnaires (A - E) which were interviewer-administered. Information was collected from each respondent on every section of the questionnaires, including A. demographic characteristics of the respondents, B. past medical and social history, C. knowledge about HBV infections, D. assessment of the abattoir environment, and E. physical hazards investigation of the respondents. The laboratory materials (LabACON HBV test kits, reagent, needles, swabs, spirit, tourniquet, sharps bin, consumables' disposal bag, cryo tubes containers, specimen bottles with racks, latex gloves, face masks) were used by the Laboratory Scientists (research assistants) to collect the appropriate blood samples, run the tests and obtain information on the actual number of participants infected with the virus under study using Hepatitis B serological markers, in this instance, presence or absence of Hepatitis B surface antigens.

Data Collection Methods

Questionnaire data collection: Information was collected from the respondents using the variables contained in the five sections of the structured questionnaires by the trained interviewers in English. In some instances, interpreters in the Hausa language were employed to explain English language terms to illiterate respondents who did not understand. In contrast, Igbo language interpreters were also engaged in interpreting the English language to the illiterate respondents. Some of the respondents, however, preferred Pidgin English and were allowed.

Serological component during the laboratory procedure, by using 5ml syringes/needles and plain tubes, after cleaning the sites with alcohol solution-soaked swabs, three milliliters (3ml) of intravenous blood samples were

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collected by the experienced laboratory scientists through puncture of either the radial or brachial veins from each of the participants. The blood samples were allowed to clot for 30 minutes as recommended by the RDT kits producer; after that, they were centrifuged for 20 minutes; serum was separated and transferred into cryo-tubes. The samples were transported to the AE-FUTHA laboratory in a cold box for HBV serological tests, carried out on the same day of arrival to control for pre-analytical problems that could have occurred due to multiple freeze-thaw cycles of the serum. Laboratory tests were validated, and quality control was monitored by running positive and negative controls along with each batch of Enzyme-linked immunosorbent assay (ELISA) test kits. The blood samples were tested for Hepatitis B surface antigen (HBsAg) using a rapid diagnostic test kit (LabACON, batch number: RP5110303) that had high sensitivity (>99.9%), 95% C. I (98.2%-100%), high specificity (99.0%), 95% C. I: 97.2%-99.8%), and accuracy of 99.4% (95% C. I: 98.3%-99.9%). Positive results were considered HBV current or past infection. Serum samples of reactive assays were left in the crypto bottles and stored in a freezer at -20°C for quality control, whereas negative samples were discarded appropriately.

Data Management

Sixteen independent variables comprised the respondents' socio-demographic characteristics, such as age, marital status, level of education, occupation, religion, employment status, number of rooms in residence, and respondents' past medical and social history [Tab 1-2]. The continuous variables were measured using a student t-test for the means (standard deviation), while categorical variables were measured using Pearson Chi-square for the difference in respondents' proportions. The dependent variables (20) comprised knowledge about Hepatitis B virus infection, practices and risk factors among the respondents, environmental conditions of the abattoirs, and market prevalence of HBV among donkey butchers and herders. Composite variables were constructed to assess knowledge of HBV infection among the respondents. For each correct answer, one score was awarded, and zero was awarded to wrong answers. The total score was calculated and converted to a percentage. This was used to

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grade knowledge into excellent or poor knowledge, with 50% as the cut-off. Scores 51% to 100% were graded as good knowledge, while scores zero to 50% were regarded as poor knowledge [Tab 3A – C]. A physical hazards investigation for donkey marketers was made up of 8 variables used to find out those activities carried out in the donkey markets that made them vulnerable to infection by the virus. Questions with 'yes' or 'No' options were used to determine the respondents' risk levels.

The data were analyzed using the International Business Machine-Statistical Package for Social Sciences (IBM – SPSS) version 26.0 software[36]. Descriptive statistical tests such as t-test and chi-square were used to compute the mean (standard deviation) and proportions for the respondents' socio-demographic, behavioral, and outcome variables. The association between putative risk factors for Hepatitis B virus infection was determined using bivariate analysis of the Chi-square variety. The level of statistical significance was set at $p < 0.05$ with a 95% confidence interval. After cross-tabulating socio-demographic variables with HBV assay result, independent variables with a cut-off p -value ≤ 0.1 were modeled into a Binary Logistic regression model for a multivariate analysis, which determined the predictors of the risks of HBV infection among the respondents.

3.13. Ethical considerations

Ethical clearance for this study was obtained from the Research and Ethics Committee of Alex Ekwueme Federal University Teaching Hospital, Abakaliki (AE-FUTHA), and permission was obtained from the State Ministry of Agriculture and Health, Ebonyi State, Nigeria. Permission to carry out the study was also obtained from the donkey butchers and herders' unions of the markets where the study was carried out. Informed consent was obtained from the study respondents after the purpose of the study was explained to them. Participation in the survey was voluntary, and confidentiality was ensured.

Results

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There were 250 respondents of donkey handlers proportionately selected from four donkey markets across the three senatorial zones of Ebonyi State comprising 125 butchers and 125 herders in Ohaukwu, Ishielu, and Onicha Local Government Areas of the state to assess their HBV infection Knowledge, Risk Factors, and prevalence. The response rate was 100% in both groups; findings are presented in the below tables (and figures).

Table 1. Between groups, a comparison of Socio-demographic characteristics of the donkey butchers and herders.

Variables	Butchers (n=125) N (%)	Herders (n=125) N (%)	(χ^2)	p-value
Age as at last birthday (Mean \pm SD) in years	42.5 (15.8)	31.4 (11.9)	6.210 ^t	<0.001*
Age group				
18-27 years	30 (24.0)	57 (45.6)	28.868 ^t	<0.001*
28-37 years	27 (21.6)	37 (29.6)		
38-47 years	28 (22.4)	14 (11.2)		
48 and above years	40 (32.0)	17 (13.6)		
Sex				
Male	71 (56.8)	116 (92.8)	42.972	<0.001*
Female	54 (43.2)	9 (7.2)		
Marital status				
Married	90 (72.0)	72 (57.6)	32.737	<0.001*
Single	18 (14.4)	52 (41.6)		
Widowed/Divorced	17 (13.6)	1 (0.8)		
Religion				
Christianity	119 (95.2)	21 (16.8)	FT (200.930)	<0.001*
Islam	1 (0.8)	100 (80.0)		
Traditionalist	5 (4.0)	4 (3.2)		
Level of Education completed.				
No formal education	36 (28.8)	36 (28.8)	FT (16.142)	<0.005*
Primary	38 (30.4)	37 (29.6)		
Junior Secondary	6 (4.8)	17 (13.6)		
Senior Secondary	33 (26.4)	27 (21.6)		
Tertiary	12 (9.6)	3 (2.4)		
Arabic	0 (0.0)	5 (4.0)		
Employment Status				
Self-employed	114 (91.2)	105 (84)	4.679	0.090
Paid employment	7 (5.6)	17 (13.6)		
Unemployed	4 (3.2)	3 (2.4)		
Number of rooms in residence				
Multiple rooms apartment	56 (44.8)	42 (33.6)	9.729	0.008*
Single room apartment	37 (29.6)	61 (48.8)		
Self-contained apartment	32 (25.6)	22 (17.6)		

SD=Standard deviation χ^2 =Chi square t=Student t-test *=Statistical significance <=Less than.

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Table 1 shows that the differences in proportions of all the variables in the butchers and herders were statistically significant except the differences in proportions of respondents' employment status, which was statistically non-significant ($\chi^2=9.729$, $p=0.090$).

Table 2. The respondent's past medical and social history between groups is compared.

Variables	Butchers (n=125) N (%)	Herders (n=125) N (%)	χ^2	p-value
Has been hospitalized before				
Yes	40 (32.0)	35 (28.0)	0.476	0.581
No	85 (68.0)	90 (72.0)		
Smokes Cigarette				
Yes	18 (14.4)	35 (28.0)	6.920	0.013*
No	107 (85.6)	90 (72.0)		
Takes Alcohol				
Yes	66 (52.8)	17 (13.6)	43.305	<0.001*
No	59 (47.2)	108 (86.4)		
Has more than one sexual partner				
Yes	25 (20.0)	30 (24.0)	0.583	0.542
No	100 (80.0)	95 (76.0)		
On any drugs of addiction				
Yes	9 (7.2)	16 (12.8)	2.178	0.205
No	116 (92.8)	109 (87.2)		
Have you ever had any casual sexual intercourse?				
Yes	26 (20.8)	27 (21.6)	0.024	1.000
No	99 (79.2)	98 (78.4)		
Have you received a blood transfusion or any blood product within the last year?				
Yes	5 (4.0)	5 (4.0)	0.000	1.000
No	120 (96.0)	120 (96.0)		

* = Statistical significance.

There were statistically significant differences in the proportions of respondents who indulged in cigarette smoking ($\chi^2=6.920$, $p=0.013$) and who took alcohol ($\chi^2=43.305$, $p<0.001$) between the two groups. However, the differences in proportions of other variables were comparable (Tab 2).

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Table 3A. Between groups' comparison of knowledge about hepatitis B virus infection.

Variables	Butchers (n=125) N(%)	Herders (n=125) N(%)	(χ^2)	p-value
Have you ever heard of HBV?				
Yes	53 (42.4)	26 (20.8)	13.491	<0.001*
No	72 (57.6)	99 (79.2)		
Source of information				
Radio	26 (20.8)	3 (2.4)		
Friend	8 (6.4)	7 (5.6)		
Hospital	7 (5.6)	5 (4.0)		
Phone	7 (5.6)	6 (4.8)		
Market	4 (3.2)	2 (1.6)		
No response/Don't know	73 (58.4)	102 (81.6)		
Meaning of HBV				
Can't remember	8 (6.4)	1 (0.8)		
Disease	28 (22.4)	13 (10.4)		
Infection	16 (12.8)	9 (7.2)		
Don't know	73 (58.4)	102 (81.6)		
Can contract HBV infection as a butcher or herder				
Yes	24 (19.2)	14 (11.2)	3.103	0.112
No	101 (80.8)	111 (88.8)		
Can contract the infection through knife cuts				
Yes	28 (22.4)	4 (3.2)	20.642	<0.001*
No	97 (77.6)	121 (96.8)		
Can contract the infection through a piercing wound				
Yes	3 (2.4)	5 (4.0)	0.517	0.722
No	122 (97.6)	120 (96.0)		
Can contract the infection through accident				
Yes	2 (1.6)	7 (5.6)	2.882	0.172
No	123 (98.4)	118 (94.4)		
Can contract the infection through risky behaviors, e.g., life donkey handling and meat processing				
Yes	5 (4.0)	1 (0.8)	2.732	0.213
No	120 (96.0)	124 (99.2)		
Aware that HBV can be contracted through sexual intercourse				
Yes	27 (21.6)	21 (16.8)	0.928	0.422
No	98 (78.4)	104 (83.2)		
HBV can be prevented				
Yes	41 (32.8)	21 (16.8)	8.579	0.005*
No	84 (67.2)	104 (83.2)		
Knows that HBV can be prevented by vaccination				
Yes	38 (30.4)	19 (15.2)	8.204	0.006*
No	87 (69.6)	106 (84.8)		

*=-Statistical significance χ^2 =Chi square

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Tab 3A showed that the differences in knowledge of butchers and herders who have heard of HBV infection compared with those who have not heard of HBV and can contract HBV through knife cuts compared with those who were unaware they could contract HBV through knife cuts were highly significant ($\chi^2=13.491$, $p<0.001$ and $\chi^2=20.642$, $p<0.001$) respectively. Also, HBV can be prevented by vaccination.

Table 3B. Between groups' comparison of respondents' knowledge about hepatitis B virus transmission and prevention of infection

Variables	Butchers (n=125) N(%)	Herders (n=125) N(%)	(χ^2)	p-value
Ever vaccinated				
Yes	4 (3.2)	3 (2.4)	0.147	1.000
No	121 (96.8)	122 (97.6)		
Completed HBV vaccination				
Yes	2 (1.6)	3 (2.4)	0.204	1.000
No	123 (98.4)	122 (97.6)		
Another way HBV infection can be prevented is through personal hygiene.				
Yes	70 (56.0)	51 (40.8)	5.782	0.023*
No	55 (44.0)	74 (59.2)		
HBV infection can be prevented through environmental sanitation				
Yes	31 (24.8)	27 (21.6)	0.359	0.653
No	94 (75.2)	98 (78.4)		
HBV infection can be prevented by wearing a protective device				
Yes	7 (5.6)	0 (0.0)	7.202	0.014*
No	118 (94.4)	125 (100)		
HBV infection can be prevented by keeping to one sexual partner				
Yes	4 (3.2)	5 (4.0)	0.115	1.000
No	121 (96.8)	120 (96.0)		
HBV infection can be prevented by wearing a condom before sexual intercourse with an unusual sexual partner				
Yes	13 (10.4)	9 (7.2)	0.797	0.504
No	112 (89.6)	116 (92.8)		
Knows that infection with HBV can cause liver cancer				
Yes	15 (12.0)	9 (7.2)	1.659	0.283
No	110 (88.0)	116 (92.8)		
Aware that HBV infection can be controlled with drugs				
Yes	32 (25.6)	15 (12.0)	7.573	0.009*
No	93 (74.4)	110 (88.0)		

*=Statistical significance.

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There were statistically significant differences ($\chi^2=8.579$, $p=0.005$ among the respondents who knew that HBV can be prevented through personal hygiene, butchers 70(56.0%), herders 51(40.8%) compared with those who did not realize that HBV can be prevented butchers 55(44.0%), herders 49(59.2%). Also, comparing the respondents' knowledge with prevention measures of HBV by protective devices and control with drugs is statistically significant (Tab 3B).

Tab 3C. Between groups comparison of composite grades of respondents' knowledge of HBV infection among the two donkey handling groups.

Grades of knowledge	Butchers (n=125) N(%)	Herders (n=125) N(%)	Chi-Square (χ^2)	p-value
Good knowledge	16 (12.8)	7 (5.6)	3.879	0.078
Poor knowledge	109 (87.2)	118 (94.4)		

p-value=Probability value N=Proportions in percentage.

Good knowledge means respondents who scored 50% and above in their knowledge of HBV infection, and poor knowledge means respondents who scored less than 50%. There were overall poor scores of the respondents' knowledge of HBV infection among the donkey butchers and herders, as evidenced by their respective proportions (Tab 3), which is statistically non-significant ($p=0.078$).

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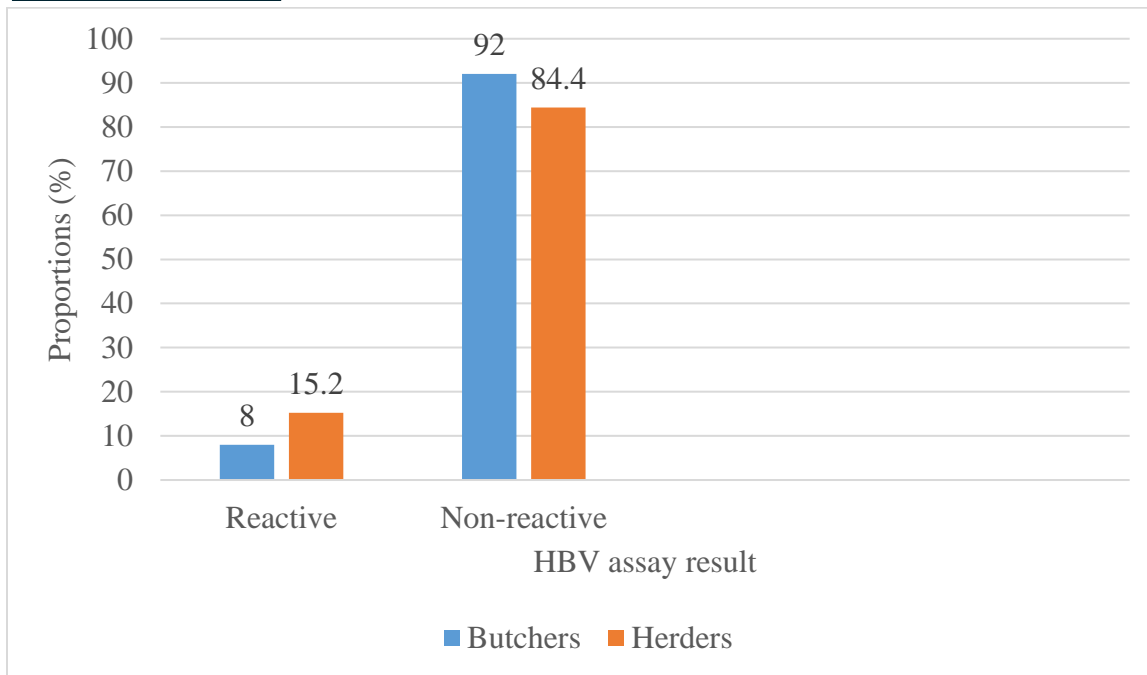


Fig 1. Between groups, comparison of respondents' HBV assay result.

The overall proportion of HBV reactive samples among the two groups of donkey handlers was 29 (11.6%), whereas 221 (88.4%) respondents' samples were HBV non-reactive (Figure 2). More HBV reactive samples were among the herders' group 19 (15.2%) compared with the butchers' group 10 (8.0%). Nevertheless, the difference between the proportions of the donkey handlers' groups was non-statistically significant ($\chi^2 = 3.160$, $p=0.113$).

The respondents' differences in comparison of mean duration of stay and pattern of disposal of animal dung and related stuff in the abattoirs were not statistically significant ($t=0.600$, $p=0.549$ and FT (5.079, 0.411 respectively). The differences in proportions among the other variables in the groups were highly statistically significant (Tab 4.4A) (appendix)

Tab 5 shows no statistically significant difference in physical hazards experienced between the groups in three variables (duration of stay as donkey handler, how treated if injury, and other wounds). In contrast, three variables showed statistical significance, including any injury, blood splashed into facial orifices, and other wounds while working in the markets. (Appendix)

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Tab 6 showed that only the mean duration of stay in years among the butcher's group was statistically significant ($t=2.005$, $p=0.047$ at 95% C. I= $0.069+11.138$) but not statistically significant when dichotomized ($\chi^2=3.308$, $p=0.069$). The difference in comparison of the rest of the variables was comparable. (Tab 7). (Appendix)

There were non-statistically significant differences in comparing the proportions of all eight variables, reactive and non-reactive samples, with HBV assay results (Tab 8). (appendix).

Comparison of grades of the butchers' proportions of HBV knowledge and HBV assay result (Tab 9) showed that the proportion of respondents with good knowledge, 16 (100%), yielded 3 (18.8%) HBV reactive and 13 (81.2%) non-reactive HBV samples. In comparison, the proportion of respondents with poor knowledge, 109 (100%) among the respondents, produced 7 (6.4%) HBV reactive samples and 102 (93.6%) HBV non-reactive samples. However, the difference in proportions of the findings showed no statistical significance ($\chi^2 = 2.881$, $p=0.118$). None of the variables in socio-demographic characteristics compared with the HBV assay result showed statistical significance (Tab 10). (Appendix)

Comparable (Statistically non-significant) findings were found among all the proportions of the respondents' variables. For instance, respondents who reported that they had donkey blood splash (Tab 11), 2 (100%) had no HBV reactive samples 0 (0.0%) compared with respondents who did not have such exposure 123 (100%) which produced all the 19 (15.4%) HBV reactive and 104 (86.6%) HBV non-reactive samples. (Appendix)

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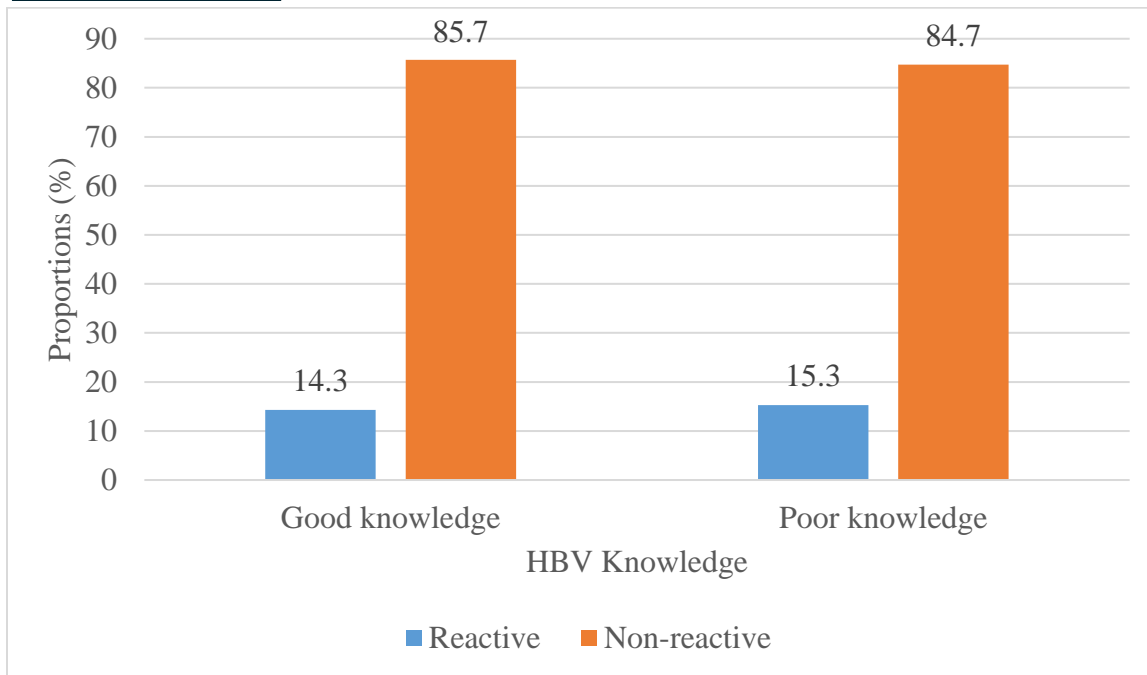


Fig 2. Relationship of herders' HBV knowledge with HBV assay result (N=125)

Good knowledge refers to the proportion of herders who scored 50% and above. In comparison, poor knowledge refers to respondents who scored below 50% in knowledge of HBV infection during the composite grading of herders' knowledge of HBV infection. Figure 4.4 showed that among the proportions of herders who had good knowledge of HBV 7 (100%), there were 1 (14.3%) HBV reactive sample and 6 (85.7%) HBV non-reactive samples compared with the proportions of herders who had poor knowledge of HBV 118 (100%) which yielded 18 (15.3%) HBV reactive samples and 100 (84.7%) non-reactive samples. However, the difference in these proportions among the grades of knowledge and HBV assay result was statistically non-significant ($\chi^2=0.005$, $p=1.000$).

Logistics regression model to determine the predictors of HBV.

After cross-tabulating independent variables with each outcome variable, a cut-off point of $p=0.1$ was used to select independent variables included in the binary logistic regression model. The choice of 0.1 as the cut-off was

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made to eliminate all confounding variables. Only independent variables with $p < 0.1$ were inputted into the logistic regression model for multivariate analysis that determined the predictors of outcome variables.

Tab 4.14 showed that herders who smoke cigarettes were 1.280 times more likely to yield HBV-positive samples than non-cigarette smokers ($p=0.691$, 95% C.I=0.379-4.323). In contrast, respondents who are drug addicts were approximately five times more likely to produce HBV reactive samples than non-drug addicts ($p=0.023$, 95% C. I =1.232-17.226). Likewise, respondents who had any casual sexual intercourse were 2.436 times more likely to have HBV reactive samples than respondents who have not had any casual sexual intercourse ($p=0.124$, 95% C. I =0.783-7.577). (Appendix)

On Predictors for Butchers.

The butcher group variables did not meet the criteria for multivariate analysis because only the duration of stay was statistically significant but became non-statistically significant when dichotomized. Multivariate analysis requires inputting two or more independent variables into the binary regression model.

Discussion

In a comparison of proportions of the respondents' knowledge about hepatitis B virus infection, seven (7) variables out of 20 variables studied showed statistically significant differences between the butchers and herders in favor of the butchers, which may be related to individuals who participated during the study [Tab 1 & 2]. More significant proportions of the respondents in both donkey handling groups on the rest of the eleven variables showed non-statistically significant differences [Tab 3A– C]. The comparable grades of respondents' knowledge of HBV infection among the two donkey handling groups may be that the respondents were from the same population (donkey handling) and had the same background knowledge despite some having higher education, which didn't translate to their HBV knowledge. The comparable result of knowledge grades could be explained by the fact that perhaps the proportion of respondents with good knowledge was insufficient to differentiate itself from the proportion of respondents with poor knowledge [Fig 1].

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Zero reactive samples found among the proportion of tertiary education compared with other levels of education may be a translation of good knowledge relative to prevention measures against HBV infection. This finding was corroborated by a related study on knowledge, attitude, and practice (KAP) among traders (though the unspecified type of trade) conducted in Calabar metropolis, South-South Region, Nigeria^[37], which found that majority of the respondents had secondary education (57.1%) and were of the Ibibio and Igbo tribes; only 10.4% had HBV vaccination whereas only 44.2% of the traders reported having any knowledge of HBV; the most common source for the knowledge was television/radio (25%) and hospitals (22%); the median (interquartile range) of the overall KAP score was low (11, 5–16); the score was least in persons aged 35 years and above, but the difference was not statistically significant ($P=0.33$). The researchers concluded that the knowledge of HBV was low among the traders in the Calabar metropolis. Among the herders handling group, the proportion of respondents who had good knowledge also showed fewer HBV reactive samples compared with the proportions of poor understanding of HBV, and the difference in proportions between the grades of knowledge showed perfectly statistically non-significant. This was probably because they operated within the same environment with the same HBV background knowledge and the inability of the excellent knowledge proportions to differentiate themselves from the proportion of poor knowledge respondents. [Tab 4]

The differences in the proportions of the variables with statistically significant findings may be due to non-uniform practices in the abattoir's donkey garage/lairage and butchers' environment. However, the non-statistical significance among the proportions of the other six variables could be due to donkey lairage and butchers' abattoirs being in the same environment. Corroborating these findings, a study in Sokoto, North West Region, Nigeria^[38] assessed abattoir operations and waste management in Nigeria and highlighted the unsanitary nature

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of abattoirs in Nigeria. The investigators also reviewed the challenges and prospects concerning environmental quality and public health. [Tab 5]

Among the six variables investigated in exposure to physical hazards [Tab 6], the proportions most probably could be due to differences in conditions between the butchers' operations and herders. In contrast, the comparability of findings among the variables may be due to the lack of health institutions within any of the donkey markets and the fact that the donkey business commenced in Ebonyi State during the same period.

Among the relationship between butchers' risk factors and practices in the abattoirs and HBV assay result [Table 7], none of the eight (8) variables' proportions difference was statistically significant, which perhaps cast doubt on the integrity of the respondents' vaccination status hence HBV vaccination was found protective among the recipients.[³⁹]

The comparison of proportions among respondents in donkey blood splash into their orifices may be because their exposure levels of the HBV risk factors were the same [Tab 8]. However, it has been reported that HBV can exist on an occult basis [^{40,41}] and is capable of reactivation in improperly screened blood during a blood transfusion. Respondents' difference in proportions of those who reported having had any other wounds or accidents while working in the abattoirs and those who did not report such have different HBV assay results, which were not statistically significant and may be due to similarity in work environment and operational time which provided them same exposure doses.

In herders group, comparison of proportions of HBV assay results with risk factors in the abattoirs, [Tab 9] the differences found among the six (6) variables were as in butchers group above, did not show any statistical significance contrasted a related study carried out in a tertiary institution in Ogun State, South West Region, Nigeria[³⁹], which reported that 54% of health workers completed HBV vaccination in the hospital in a previous

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HBV vaccination exercise. In contrast, 65% of respondents completed the HBV vaccine in the study despite the hospital carrying out occasional vaccination programs. According to the reports, complete vaccination was identified among the predictors of good knowledge. The similarity in findings among respondents on health supervision may be that activities of the health officials in the abattoirs might not have been targeted at HBV control. Though the proportions of herders who did not report having had any piercing or cut or head injury had the majority of the HBV reactive assay results compared with respondents who reported to have had such showed more reactive HBV samples, the difference was also not statistically significant which was presumed to be due to similar exposure risk factors and that those who did not report injuries could have had HBV exposure through other risk factors not listed in this study.

Results of binary Logistics regression among the herders' group [Tab 10 - 11] could be due to unknown confounders among these variables, which were not factored into the study during the study design, which corroborated results obtained from a related study in Goiânia, Brazil^[42] that HBV among female sex workers and control the overall prevalence of HBV was 17.1% (95% C.I 11.6–23.4) and 1.6% (95% C.I 0.1–4.7) respectively; their positivity were occasioned by their samples being HBsAg reactive. However, the results of cross-tabulation of butchers' HBV status with the proportions of socio-demographic variables showed that only the proportions of the duration of stay in the abattoir were statistically significant, but when the variable proportion was dichotomized, it showed a non-statistically significant difference, hence did not meet the assumption to be fixed into the regression model; so, left at the association test level.

The higher prevalence among herders compared with the butchers [Fig 2] may be due to the former's poorer knowledge of HBV infection and the fact that the herders consisted of younger age respondents who also were long-distance travelers and who stayed outside their formal residence procuring the donkeys from the far Northern States down to Southeastern region, Nigeria. This finding was corroborated by a review update of HBV in

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Nigeria^[43], which found HBV prevalence to be 4.3% to 23.3% across the country and a pooled prevalence of 13.5% among the population. A cross-sectional study conducted in Toro, Bauchi State, and the North Eastern region of Nigeria [44] showed a higher HBV prevalence of 26.2% among the Fulani cow herders. However, there % was a reduced prevalence of 11.6% among butchers and 10.9% among slaughtered cows in a related study carried out in the Kano metropolis, Kano State, North Western region, Nigeria^[45]. Related research carried out in Mbagathi district hospital, Kenya, Nairobi^[46], and neighboring countries reported a 3.8% prevalence among pregnant women attending ANC in the hospital with the highest infection rate among the 20 – 24 age group.

Recommendations

The need to conduct awareness campaigns and vaccination exercises for the donkey handlers should be emphasized. Developing and disseminating abattoir policy guidelines to Ebonyi State, especially the donkey handlers, is imperative, including quarterly supportive supervision. Standards should be encouraged among the members to register new members, including pre and ongoing maintenance of the occupational health of the donkey handlers. Members of the donkey butchers should receive re-orientation to improve the environmental sanitation of the abattoirs.

Conclusions

The results from this study showed that there needed to be a better grade of knowledge among the donkey butchers and herders, though comparable. Some risk factors were associated and predictors of HBV transmission among the donkey butchers and herders. There were also statistically significant findings in the lifestyles of the groups, which encouraged the attendant risk factors for HBV infection. The prevalence of HBV among the respondents showed that the donkey handlers were high-risk groups for HBV transmission. However, the difference in proportions of their positive results was not statistically significant (comparable). The study could be adjudged to

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have met its general and specific objectives. These results have obvious public health implications for the donkey butchers and herders to reduce HBV transmission in Ebonyi State.

Footnotes.

Peer-Reviewers: Amany Mohamed Abdallah (Assistant professor of community medicine), Marwa Shabana (Assistant professor of clinical pathology), Ahmed Abdelsameeh (professor of veterinary medicine).

E- Editor: Salem Youssef Mohamed, Osama Ahmed Khalil, Mohamed Hassan Ali Emara.

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Ethics Approval and Consent to Participate: The study has been ethically approved by the Research and Ethics Committee (REC) of Alex Ekwueme Federal University Teaching Hospital, Abakaliki, Ebonyi state, Nigeria; they can be contacted at oujair@yahoo.com or by phone +23439558074 or +27854049650.

Consent for publication: All patients included in this research gave written informed permission to publish the data contained within this study.

Availability of data and materials: The datasets used or analyzed during the current study are available from the corresponding author upon reasonable request.

Competing interests: The authors declare that they have no competing interests.

Funding: This study had no funding from any resource.

Authors' contributions

The study was conceptualized and designed by all authors. Elom, Una, and Chika collected and compiled data. Elom and Una conducted the statistical analysis, while Una and Chika drafted the manuscript. Elom, Una, and Chika provided significant intellectual input throughout the project, contributing to comments and revisions. All authors reviewed and approved the final manuscript.

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