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Correlates of hepatocellular and gastric diseases in cows slaughtered in selected abattoirs in Ogun State, Nigeria

Jude Ogechukwu Okoye[1], Chijindu Nneka Nzeaka[2], Gideon Taiwo Faloye[3]

[1]Department of Medical Laboratory Science, Faculty of Health Sciences and Technology, Nnamdi Azikiwe University, Nnewi, Nigeria

[2]Department of Medical Laboratory Science, School of Public and Allied Health, Babcock University, Ilishan Remo, Nigeria

[3]Department of Medical Laboratory Science, School of Public and Allied Health, Babcock University, Ilishan Remo, Nigeria

ABSTRACT

Background: Livestock farming, especially cattle rearing, remains a major source of income for local farmers. However, the health status of cattle and the standard of veterinary services determine the quality of animal meat and the risk of zoonotic transmission. Thus, this study determined the prevalence of *Helicobacter pylori*, parasitic infection, and gastric and hepatocellular diseases in cows slaughtered in two selected government-approved abattoirs.

Methods: Following the systematic random selection of cows, liver and gastric tissues were taken from the slaughtered cows (n = 99, each), fixed in 10% neutral-buffered formalin, processed and stained accordingly, and microscopically examined for the presence of *H. pylori*, parasites, hepatocellular, and gastric diseases.

Results: Approximately, 66% and 34% of the liver and gastric tissues had pathological changes, respectively. The pathologies observed in the liver were cirrhosis (12.1%), fibrosis (8.1%), hepatitis (28%), and necrosis (17%), whereas tumors (6.1%), dysplasia (10.1%), and gastritis (18%) were observed in gastric tissues. The prevalence of parasitic infections and *H. pylori* in the liver and gastric tissues were 4% and 26%, respectively. The prevalence of the bacteria was higher in pathologic gastric tissues (44.1%) than in normal gastric tissues (16%). There was a significant association between gastric diseases and *H. pylori* (p = 0.007). Only 20.2% of the investigated animals were without any obvious gastric and hepatocellular diseases.

Conclusion: The study revealed that *H. pylori* infection is associated with gastric diseases in cows. It suggests that some cow meat from some abattoir may be unsafe for human consumption.

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Helicobacter pylori; parasite; gastric tumor; cirrhosis; fibrosis; cattle

Introduction

An assessment of slaughterhouses in Nigeria revealed that about 48% of animals presented for slaughter had poor health status evidenced by emaciation. The ratio of extreme emaciation between cattle and goat is 2:1 [1]. According to Shima et al., female cattle have significantly higher frequency of emaciation than their male counterparts. The reason for their findings remains unclear. Hence, it is only logical to investigate their findings further [1]. Abattoir-related zoonotic infection is increasing due to high dependence on livestock for nutrition and revenue [2–5]. The farmers' rationale for selecting out animals (from stock) for sale is yet to be understood. It is hypothesized that ailing animals are preferred over healthy animals to cut the losses. Again high cost of veterinary services may influence the farmers' decision to sell. The sale of sick animals to abattoir may trigger several chains of detrimental events. The common modes of zoonosis include direct contact with animals, ingestion of infected milk, and meat. Infections ranging from parasitic to bacterial infections are associated with mild-to-fatal human diseases [6]. In Nigeria, the prevalence of

Contact Jude Ogechukwu Okoye ✉ jog.okoye@unizik.edu.ng Department of Medical Laboratory Science, Faculty of Health Sciences and Technology, Nnamdi Azikiwe University, Nnewi Campus, Nigeria.

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parasitic infections in cattle is approximately 15% [7,8]. This prevalence is similar for other African countries, such as Egypt, Ethiopia, Libya, and Saudi Arabia but higher than the prevalence of other countries, such as the USA (30%), Canada (7.1–30%), and the Netherlands (1.2%) [9]. *Helicobacter pylori,* a group I carcinogen, is a Gram-negative curved rod and microaerophilic bacterium usually found in the stomach. About 85% of the infected people never experience the symptoms or complications [10]. The findings of Mohamed et al. show that *H. pylori* is transmitted from animal to human [11]. The infection sequentially induces gastritis, gastric atrophy, metaplasia, dysplasia, and finally carcinoma [12]. About 1–3% of infected persons develop gastric adenocarcinomas [13]. Globally, *H. pylori*-associated gastric cancer accounts for approximately 6% of all cancers [14]. No study has investigated the presence of *H. pylori* animal tissues in the state. This study investigated the presence of parasitic and bacterial infections as correlates of hepatic and gastric diseases in cattle. Information obtained from this study could be used to track the hygiene status of meats sold on the Nigerian markets.

Methodology

This study was carried out between the periods of January and June 2019. An ethical clearance for this study was obtained from Babcock University Health Research Ethics Committee (BUHREC 430/19). Ten (10) slaughterhouses within reach in Remo land were sampled, but only two slaughterhouses (Iperu Remo and Ilisan Remo, Ogun State: coordinates; 6.8862°N, 3.7055°E) met the criteria for selection by following an interviewer-based questionnaire: government approval, type of animal slaughtered (cattle), weekly visit by inspector, low refusal rate (≤9% of all incoming animals) to slaughter by inspector/health officer, daily cleaning of equipment between animals before and after slaughter, and disposal method for diseased carcass (pit/bury) [15]. Systematic random sampling was used in selecting animals, from which samples were taken from. One (1) out of every four apparently healthy animals that were brought into the slaughterhouses was consecutively selected. A total of 416 cows were sampled, but only 104 cows were selected. Samples were taken between the hours of 7 and 9 am. Samples from the four sections of the stomach (cardiac, fundus, body, and atrium) and three lobes of the liver (right, left, and caudate) were taken from the 104 slaughtered animals. Samples from five animals out of the 104 selected animals had longer cold ischemic time (>1 hour) and were eliminated from the study. Tissue sections were collected into 10% neutral-buffered formalin not later than an hour following cattle slaughter. The presence of *H. pylori* was determined microscopically following Giemsa, hematoxylin and eosin, and Warthin–Starry staining techniques [16]. The descriptive statistics were carried out to determine the prevalence of bacteria, parasitic infection, liver, and gastric diseases in tissue sections. The Chi-square/Fisher's exact test was used to determine whether sex influences bacteria, parasite, and pathological burden. It was also used to determine whether *H. pylori* infection was associated with normal or abnormal gastric tissues using GraphPad Prism (version 6). Significance was set at $p < 0.05$.

Result

Histopathological investigations revealed cirrhosis, fibrosis, hepatitis, necrosis, and some parasitic infection in the liver, whereas adenocarcinoma, dysplasia, and *H. pylori* were observed in gastric tissues. Approximately, 66% and 34% of the liver and gastric tissues had pathological changes, respectively. The prevalence of parasitic infections and *H. pylori* in the liver and gastric tissues were 4% and 26.3%, respectively (Table 1). The prevalence of hepatocellular diseases was higher than gastric diseases (Figures 1–3). The prevalence of *H. pylori* was higher in pathologic gastric tissues (44.1%) than in normal gastric tissues (16%), whereas parasitic infections were only observed in hepatocellular diseases. There was a significant association between gastric abnormalities and *H. pylori* (p = 0.007). The prevalence of synchronous liver and gastric diseases was higher in female cows (26.7%) than in male cows (16.7%) at p = 0.324 (Table 2). The prevalence of *H. pylori* was higher in animals with both liver and gastric abnormalities (52.4%) than in animals without any liver and gastric diseases (36.4%; p = 0.364). However, no association was observed between gastric *H. pylori* infection and hepatocellular diseases since the prevalence of gastric *H. pylori* was higher in animals with normal liver tissues than in those with abnormal liver tissues (p = 0.018). The prevalence of gastric diseases was insignificantly higher in females (40%) than in males (29.6%) at p = 0.297. The prevalence of hepatocellular diseases and *H. pylori* were also insignificantly higher in females (68.9% and 26.7%, respectively) than in males (63% and 25.9%,

Table 1. Prevalence of liver and gastric diseases, parasitic infections, and bacterial infections in cattle.

Variables		Male	Female	Gastric/*H. pylori*	Liver/Parasite
	***n* = 99**	***n* = 54**	***n* = 45**	***n* = 26**	***n* = 4**
Liver tissues					
Cirrhosis	12 (12.1)	5 (41.7)	7 (58.3)	3 (25.0)	1 (8.3)
Fibrosis	8 (8.1)	5 (62.5)	3 (37.5)	1 (12.5)	2 (7.1)
Hepatitis	28 (28.3)	14 (50.0)	14 (50.0)	7 (25.0)	0 (0.0)
Necrosis	17 (17.2)	10 (58.8)	7 (41.2)	1 (5.9)	1 (5.9)
Abnormal	65 (65.7)	34 (52.3)	31 (47.7)	12 (18.5)	4 (6.2)
Normal	34 (34.3)	20 (58.8)	14 (41.2)	14 (41.2)	0 (0.0)
Gastric tissues					
Gastric adenocarcinoma	5 (5.1)	2 (40.0)	3 (60.0)	1 (20.0)	0 (0.0)
Carcinoid tumor	1 (1.1)	0 (0.0)	1 (100)	1 (100)	0 (0.0)
Dysplasia	10 (10.1)	5 (50.0)	5 (50.0)	5 (50.0)	0 (0.0)
Gastritis	18 (18.0)	9 (50.0)	9 (50.0)	8 (44.4)	0 (0.0)
Abnormal	34 (34.3)	16 (47.1)	18 (52.9)	15 (44.1)	0 (0.0)
Normal	65 (65.7)	38 (58.5)	27 (41.5)	11 (16.9)	4 (6.1)
Female	45 (45.5)			12 (26.7)	3 (6.6)
Male	54 (54.5)			14 (25.9)	1 (1.9)

respectively) at $p = 0.671$ and 1.000, respectively. The prevalence of parasitic infection was insignificantly higher in female cows than in male cows ($p = 0.327$). The prevalence of copresence of liver parasite and gastric *H. pylori* in cows was 1.1% (Table 2). A total of 14 (14.1%) gastric tissues from slaughtered cows had numerous eosinophils without any evidence of *H. pylori*. Only 20.2% of the investigated animals were without any obvious gastric and hepatocellular tissues.

Discussion

Helicobacter pylori infection occurs everywhere around the world, but the prevalence rate varies from 20% to 50% in developed countries and 70%–90% in developing countries [17,18]. The difference could be due to socioeconomic factors and high dependence on animal protein [19]. The prevalence of the bacteria in countries with advanced economies lies between those two ranges. Evidence suggests that contact with infected animals, consumption of dairy products, or high dependence on cow meat for protein increase the risk of the infection in man, especially among abattoir workers [11,20]. Hence, the prevalence rate of the bacteria in animal and human tissues appears to be similar.

In this study, the prevalence of *H. pylori* in cow meat is similar to that of Egypt and Iran, which are all developing countries. In Egypt, the seroprevalence of the bacteria in cows and man is 30% and 44.4%, respectively [20]. In Iran, the seroprevalence of the bacteria in man is 27% [21], while the prevalence rate of the bacteria in cow meat is 25% [22]. Although *H. pylori* was not seen in some tissues with gastritis and adenocarcinoma, evidence suggest that the observed numerous infiltration of eosinophils in such cases, a potent antihelminthic and a critical player in malignant neoplastic events, may be due to *H. pylori* infection [23,24]. The difference may be accrued to improved hygiene or improved socioeconomic status in the selected communities. Again, the high prevalence of the bacteria observed in females though insignificant is similar to the findings of previous studies in the United Arab Emirates [25]. As seen in this study, the bacteria may coexist with parasite, such as *Fasciola hepatica* within or outside the gastric mucosa.

Fasciola hepatica, a liver fluke, not only infects and affects sheep, goat, and cattle but also people living in Africa, Asia, and South America through zoonotic pathways [26]. It induces fibrosis and inhibition of macrophage nitric oxide production [26] and is associated with about 9% livestock mortality in developed countries [27]. The prevalence of *F. hepatica* in this study is similar to the findings of Eze and Briggs [28] in cattle (5.3%) at Rivers State,

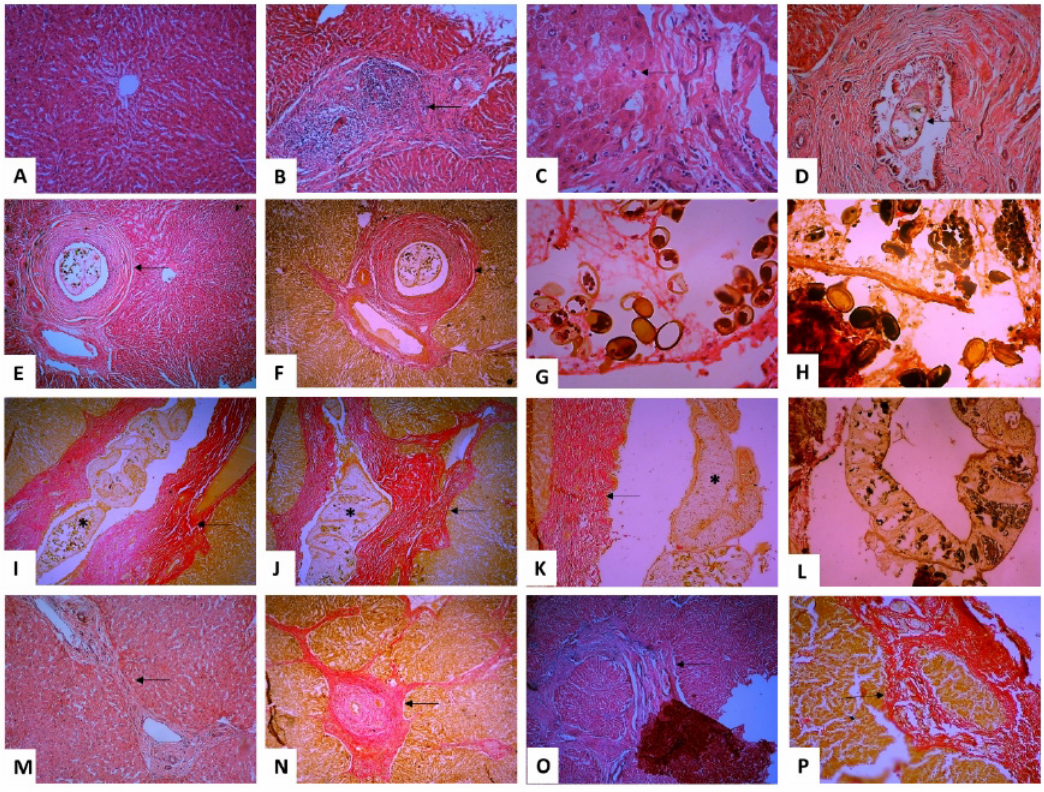

Figure 1. Photomicrographs of normal and ab normal liver sections with and without eggs and adult stages of liver fluke. (A, B, and C) Liver section without any obvious architectural changes, with numerous inflammatory cells consistent with hepatitis and moderate necrosis, respectively. (D–H) Liver sections with eggs of *F. hepatica* in dilated intrahepatic duct and periductal fibrosis. (I–L) Liver sections with adult stages of *F. hepatica* (asterisks*) and evidence of fibrosis (arrows). (D and F) Liver section with high deposition of collagen fibres consistent with fibrosis without liver fluke. (O and P) Liver section with collagen-circumscribed nests of hepatocytes consistent with cirrhosis. Stained by H&E technique (A-E, M, and O), Wiegert van Gieson's technique (F, I, J, K, N and P) and Verhoff van Gieson's technique (H and L). Magnification of A, B, E, F, I-K and M-P = X40, C and L= X100, G and H = X400.

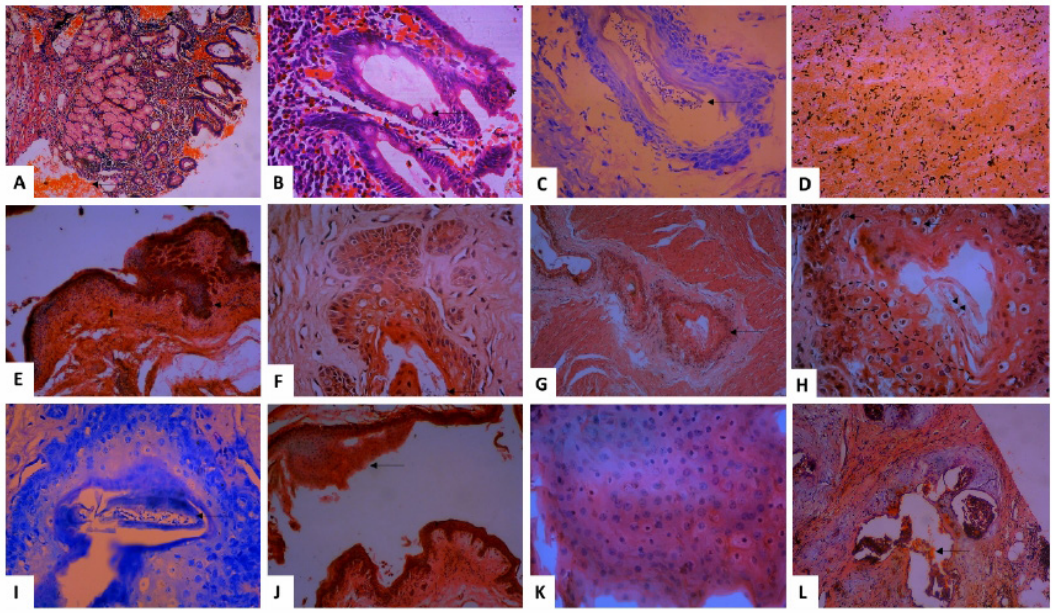

Figure 2. Photomicrograph of stomach sections stained by H&E (A, B,E-H, and J-L), Giemsa (C and H), and Warthin-Starry (D) tecniques. Synchronous haemorrhagic ulcer and chronic atrophic gastritis with eosinophils (HAG; A and B), H. pylori-infected HAG (C and D), hyperplastic gastritis (E), moderate dysplasia with H. pylori infection (F), moderate metaplasia with H. pylori infection (G-I), carcinoid tumour (J and K) and gastric adenocarcinoma (L) with eosinophilic infiltrates).

Table 2. Synchronous liver and gastric diseases in relation to gastric *H. pylori* infection.

Variables		Male	Female	Gastric/*H. pylori*
	n = 99	*n* = 9 (%)	*n* = 12 (%)	*n* = 11 (%)
Adenocarcinoma/Fibrosis	1 (1.1)	1 (100)	0 (0.0)	1 (100)
Adenocarcinoma/Hepatitis	2 (2.0)	1 (50.0)	1 (50.0)	0 (0.0)
Gastric dysplasia/Cirrhosis	1 (1.1)	1 (100)	0 (0.0)	0 (0.0)
Gastric dysplasia/Hepatitis	1 (1.1)	0 (0.0)	1 (100)	1 (100)
Gastric dysplasia/Hepatic necrosis	3 (3.0)	1 (33.3)	2 (66.7)	0 (0.0)
Gastritis/Cirrhosis	5 (5.1)	0 (0.0)	5 (100)	3 (60.0)
Gastritis/Fibrosis	1 (1.1)	0 (0.0)	1 (100)	1 (100)
Gastritis/Hepatitis	1 (1.1)	0 (0.0)	1 (100)	0 (0.0)
Gastritis/Hepatic necrosis	6 (6.1)	5 (83.3)	1 (16.7)	5 (83.3)
Total	21 (21.2)	9 (42.9)	12 (57.1)	11 (52.4)

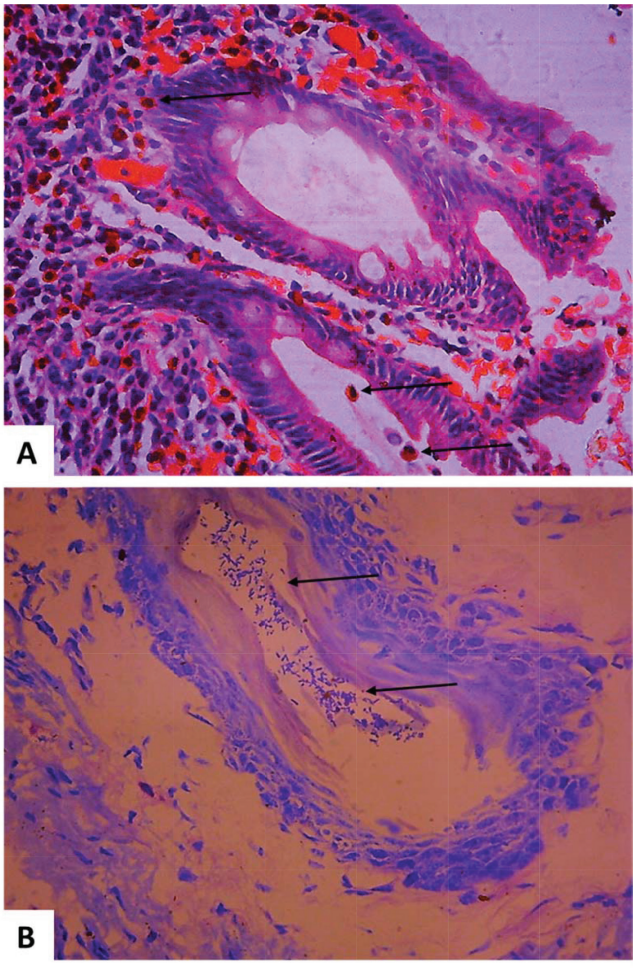

Figure 3. Photomicrography of gastric sections with evidence of numerous eosinophils (A) and presence of *H. pylori* (B). H and E stained. Original magnifications: ×400. Sections A and B refer to sections B and C in Figure 2, respectively.

Nigeria. However, the prevalence of the infection is higher in female cows than in male cows. This finding is in line with the reports of Shima et al. [1] and differs from the findings of Eze and Briggs [28]. The latter reported a prevalence rate of 5.6% and 4.3% in male and female cows, respectively. The fibrosis and cirrhosis observed in the liver tissues were similar to the findings of the previous studies [29,30]. However, the prevalence of cirrhosis in this study is lower than the reports of Mohamed et al. [11] (16.5%) but higher than that of Ejeh et al. [7] (10.4%) in Zaria, Kaduna State. The observed infections in meats may increase the number of infections in communities with concomitant development of gastrointestinal pathologies.

Conclusion

The study revealed that *H. pylori* infection is associated with gastric diseases in cows. It also revealed that a high number of cows slaughtered in some abattoirs were unhealthy. It suggests that the meat from such animals may be unsafe for consumption. Thus, the extent of veterinary services to livestock should be improved and better abattoir monitoring plans should be in place so that safe meat could be sold in the Nigerian market.

Conflicts of interest

There are no conflicts of interest in this study.

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