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Original Research Article

Prevalence of bovine tuberculosis in Kaduna central abattoir, Kaduna State, Nigeria

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ABSTRACT

Bovine tuberculosis (bTB) caused by *Mycobacterium bovis* is a chronic infectious and contagious disease of domestic and wild animals, as well as humans. The disease has been reported in various livestock species in Nigeria. The aim of the study was therefore to determine the prevalence and epidemiological factors of *Mycobacterium* species infection in slaughtered cattle at the Kaduna Central Abattoir, Kaduna State, Nigeria. Lesions were determined by observation/palpation for nodules in the predilection organs and further use of acid fast stain test on such lesions. The study showed overall prevalence of 14% by acid fast staining and 17% by the presence of lesions. Among the Breed prevalence was 57.1% (White Fulani), 23.2% (Sokoto Gudali) and 19.6% (Red Bororo). Sex prevalence was 48.2% and 51.8% for cows and bulls, respectively. Age prevalence was 17.9% and 82.1% for <4 year-old and >4year-old, respectively. Detection of lesions of bTB in slaughtered cattle poses a public health risk. This study has added useful epidemiological information on bTB infection in cattle slaughtered at Kaduna Central Abattoir. Enhanced public education and awareness campaigns should be carried out among meat handlers and abattoir workers especially on the mode of transmission, aetiology, and public health importance of bTB. There is the need to conduct molecular detection of the actual *Mycobacterium* species infecting cattle, other domestic animals and humans with a view to fully understand the status of bTB in Kaduna State, Nigeria.

Keywords: Bovine tuberculosis, cattle, Kaduna central abattoir, prevalence, Nigeria.

INTRODUCTION

Bovine tuberculosis (bTB) is a chronic infectious and contagious zoonotic disease of domestic animals, wild animals and humans (Radostits *et al.,* 2007). Bovine tuberculosis is caused by *Mycobacterium bovis* a member of *Mycobacterium tuberculosis* complex (MTC) (Pfeiffer, 2003). The disease is characterized by the formation of granulomas in tissues, especially in the lungs, lymph nodes, liver, intestines, and kidneys (Radostits *et al.,* 2007). In animals, the disease can be transmitted by ingestion of feed and water contaminated with urine, faecal material or exudates that contain the tubercle bacilli from diseased animals (Thoen *et al.,* 2009). It can spread to humans typically by inhalation of contaminated aerosols, ingestion of unpasteurized milk and its products and through breaks in the skin (Radostits *et al.,* 2007). Raw or undercooked beef can be a source of infection for humans (Ameen *et al.,* 2008).

Tuberculosis is a major health problem with about 10 million new cases and 1.4 million deaths in humans reported annually worldwide with the majority of these occurring in developing nations (WHO, 2020). Bovine tuberculosis in humans is becoming increasingly important in developing countries like Nigeria due to the fact that humans and animals share the same micro-environment and dwelling premises, especially in rural areas (Ibrahim, 2016; Kaltungo, 2018). Animal and human tuberculosis caused by pathogenic bacteria belonging to the MTC, *M. bovis and M. tuberculosis* are widespread and affect the animal industries and human health in Africa (Ibrahim *et al.,* 2010). Globally, Nigeria ranks 7th among the 30 high TB burden countries and also 2nd in Africa. This accounts for about 4% of the estimated incidence cases globally (FMH, 2017). Also, Nigeria reports the highest estimated number of new human cases with nearly 368,000 new cases annually (OIE, 2008).

The situation with animal tuberculosis in Nigeria is less clear due to lack of national control strategy and the degree of

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zoonotic transmission of tuberculosis from animals to humans is not well known. Furthermore, the absence of routine and effective disease surveillance and disease reporting system in Nigeria and the insidious nature of the disease has contributed to lack of effective and sustainable measures for its control. The poor funding of public programmes in both livestock and human life has further worsened effect to understand the situation of the diseases generally.

The extensive livestock production system commonly practiced in Nigeria which allows animals to move freely thereby intermingling with other animals of unknown background and man can result in the acquisition of a number of diseases including those of zoonotic implications (Saidu *et al*., 1991; Kaltungo, 2013, Yakubu, 2016). Not only that, the borrowing of breeding sires or sending female animals on heat to sires in other herds for breeding purposes as reported by Kaltungo (2013), Buhari (2014) and Yakubu (2016) can increase the chances of diseases spread.

The habit of Nigerians particularly in the Northern States of the country, especially pastoralists in consuming unpasteurized milk with millet paste ('Hura da Nono') along with eating partially cooked meat could result in individuals acquiring tuberculosis if such milk or meat are contaminated with *Mycobacterium* organisms.

This study was conducted to determine the prevalence and epidemiological factors of *Mycobacterium* infection in slaughtered cattle at the Kaduna Central Abattoir.

MATERIALS AND METHODS

STUDY AREA

The study was carried out at the Kaduna Central abattoir. The abattoir was located at the capital of Kaduna State, Nigeria which is situated between latitude 6^o and 11^o N and longitude 7^o and 44^o E (KDSG, 2008) Kaduna State is composed of 23 Local Government Areas (LGAs) and occupies about 48, 473.25 Sq. Km, with a human population of over 6,066,562 people according to the 2006 census figures (NPC, 2006; KDSG, 2008).

Sample Size

The sample size used in the study was determined using the formula by Mahajan (1997) where:

$$n = \frac{Z^2 pq}{d^2}$$

n = minimum sample size

p = prevalence (18.2%, Yerima (2013)

q = Complementary probability, 1 – p

d = Desired level of significance

z = Appropriate value for the standard normal deviation set at 95% confidence interval

(1.96)

The sample size was calculated as thus:

$$n = \frac{(1.96)^2 \times 0.182 \times 0.818}{(0.05)^2}$$

n =228.76 ≈ 229 cattle

However, the sample size was increased to 400 cattle to reduce errors and enhance precision.

SAMPLE COLLECTION AND TRANSPORTATION

For sample collection, systematic random sampling technique was employed in which the third animal being slaughtered was examined till the required number of samples was collected. All samplings were conducted between 7.00am and 9.00am on all the days of sampling. During sampling, complete post mortem inspection of the selected cattle was conducted. The lungs and its adjoining lymph nodes, liver, spleen, kidneys, intestines and such other organs known to be affected by TB were examined by palpation for evidence of nodules. Such lesions suggestive of TB were further examined by incision for any evidence of "gritty" sound. About 10 grams from such organs were excised immediately and placed in individual sterile sample containers which were labeled as to the number of the animal being sampled along with date of collection. The sex, age, breed and type of sample were recorded in a log book. The samples were then transported to the Synlab Laboratory, Kaduna where they were kept in a deep freezer (-20^{o}C) till used for analysis.

The sex of the cattle sampled was confirmed by the presence of penis and scrotum in males and vulva and udder in females. Also, the age of the cattle sampled was determined by the use of dentition (Pace and Wakeman, 1983). Pictures of all suspected samples were also taken.

LABORATORY INVESTIGATION

For the laboratory investigation, the samples were removed from the deep freezer and kept on the working bench to thaw to room temperature before analysis. A pinch of each of the tissues was then removed from the main sample using a sterile hand forceps and the nodule from the tissue opened by mild digestion. The exudate from the lesion was then cut and placed at the centre of a clean grease–free microscopic glass slide and then fragmented till thin lane smear was made on the slide. The slide was then air dried and heated to fix the sample on it. The slide was then placed on a staining rack with the specimen top most. Carbol fuschin stain was then added onto the smear and then heated until vapour began to rise. The slide was removed gently and washed with clean tap water to remove the stain. Two per cent v/v of acid alcohol was poured on the smear and left for 5 minutes until the smear was sufficiently decolorized by appearance of pale pink colour. The slide was washed and then covered with malachite green stain for 1 to 2 minutes after which the slide

was examined under the microscope using 100 oil immersion objective for evidence of *Mycobacterium* species. Results were interpreted by the presence of bright red to slightly purple rods as being positive for *Mycobacterium* species or blue as being Negative. The results were then recorded in a log book using the sample number as earlier on recorded.

The epidemiological factors of bovine tuberculosis were obtained by determination of the prevalence of tuberculosis by sex, age, breed and organs of the animals under study through the use of the results of the microscopic examination of positive samples collected from the abattoir.

Data generated during the study were entered into Excel spread sheet. Information obtained was analyzed using Statistical Package for Social Sciences (SPSS) version 20.0. Statistical method employed was descriptive statistics which included frequencies and percentages. Results were presented in tables and Figures. Chi-square test was used to test for associations among categorical variables. Furthermore, odds ratio (OR) and 95% confidence interval on OR were calculated. Values of $p < 0.05$ were considered significant.

RESULTS

DISTRIBUTION OF SUSPECTED GROSS TB LESIONS

Of the 400 cattle sampled, 189 (47.3%) were females while 211(52.7%) were males. Also 134 (33.5%) were below 4 years old while the remaining 266 (44.5%) were over 4years old. The White Fulani breed of cattle made up 54.3% of the cattle samples while the remaining 27.0% and 18.7% were Sokoto Gudali and Red Bororo breeds of cattle respectively (Table I).

The distribution of suspected lesions of TB in the sampled cattle is presented in Table II and Figures I - IV. Of the 68 samples examined, mediasternal lymph nodes had 10 (14.7%) lesions while, 6 (8.8%) of lesions were found in Trachea-bronchial lymph nodes. Others lesions of 13 (19.1%), 20 (29.4) and 8 (11.8) were observed in the, mesenteric lymph nodes, lungs and submandibular lymph nodes respectively. The remaining 11(16.2%) were from other tissues like heart, intestine and liver (Table II).

PREVALENCE AND EPIDEMIOLOGICAL FACTORS OF BOVINE TUBERCULOSIS BY GROSS LESIONS

From the study, the overall prevalence of TB lesions was 17%. The prevalence in the sampled cattle by sex indicated that 36 (19.1 %) of the189 cows had tubercles while 32(15.2 %) of the 211 bulls had same lesions. There was no statistically significant difference in prevalence by sex (X^2=0.847; P=0.357) (Table III).

Table II: Distribution of tubercles in sampled cattle at Kaduna Central abattoir, Nigeria.

Organs with lesions	No. TB Lesions (%)
Mediasternal Lymph node	10 (14.7)
Trachea bronchial lymph node	6 (8.8)
Mesenteric lymph ode	13 (19.1)
Lungs	20 (29.4)
Submandibular lymph node	8 (11.8)
Others (Heart, intestine, liver)	11 (16.2)
Total	**68 (17.0)**

Similarly, the prevalence of TB lesions by age showed that of the 134 cattle of less than 4 years, 21 (15.6 %) had tubercles while 47 (18.0 %) of the 266 cattle older than 4 years had the same lesions. There was no statistical difference in the prevalence by age (X^2 =3.112; P=0.211) (Table IV).

The prevalence of TB lesions according to breed indicated that of the 217 White Fulani breed of cattle examined, 41(19.0 %) had lesions while 15 (14.0 %) of the 108 Sokoto Gudali and 12 (16.0 %) of the 75 Red Bororo cattle had suspected lesions (Table V). There was no statistical difference in the prevalence of tubercles by breed (X^2=146; P=0564).

PREVALENCE AND EPIDEMIOLOGICAL FACTORS OF BOVINE TUBERCULOSIS BY ACID FAST STAIN (AFS)

With regard to the prevalence and epidemiological factors of TB by AFS, 56(82.35%) of the 68 cattle that were positive for TB by gross lesions were further positive by AFS. Furthermore, 27(75.0%) of the 36 cows that were positive by gross lesions were also positive by AFS while of the 32 bulls that were positive by gross lesions 29 (90.63%) were further positive by AFS (Table VI). There was no statistically significant difference in the prevalence of TB by sex using AFS (X^2=0.31; OR=1; 95% CI=0.598 -1.853; P=0.857).

Table I: Cattle sampled for prevalence of bovine tuberculosis by sex, age and breed at Kaduna central abattoir, Nigeria. (n=400)

SEX		AGE		BREED		
Female	**Male**	**<4years**	**>4years**	**White Fulani**	**Sokoto Gudali**	**Red Bororo**
189 (47.3%)	211 (52.7%)	134 (33.5%)	266 (66.5%)	217 (54.3%)	108 (27.0%)	75 18.7%)

Table III: Prevalence of tubercles by sex of cattle at Kaduna Central abattoir, Nigeria

Sex	No. sampled	No. Positive (%)	Chi-square	df	OR	95% CI	P value
Female	189	36 (19.1)	0.847	1	1.2	0.756-2.165	0.357
Male	211	32 (15.2)					
Total	400	68 (17.0)					

Table IV: prevalence of tubercles in cattle by age at Kaduna Central abattoir, Nigeria

Age	No. sampled	No. Positive (%)	Chi-square	df	P value
<4years	134	21(15.6)	3.112	1	0.211
>4years	266	47 (18.0)			
Total	400	68 (17.0)			

Table V: Prevalence of tubercles in cattle by breed at Kaduna central abattoir, Nigeria

Breed	No. sampled	No. positive (%)	Chi-square	df	P value
White Fulani	217	41(19.0)	1.146	2	0.564
Sokoto Gudali	108	15(14.0)			
Red Bororo	75	12(16.0)			
Total	400	68 (17.0)			

The prevalence of TB by age showed that of the 21 cattle under 4 years of age that were positive by gross lesions, 10(47.62%) were further positive for TB by AFS while of the 47 cattle older than 4 years, 46(97.87%) were positive by AFS (Table VII). There was no statistically significant difference in the prevalence of TB in cattle by age using AFS (X^2=2.318; P= 0.314).

By breed, the prevalence using AFS indicated that of the 41 White Fulani cattle that were positive by gross lesions 32(78.05%) were further positive by AFS while of the 15 Sokoto Gudali cattle that were positive by gross lesions 13(86.67%) were further positive by AFS and that of the 12 Red Bororo cattle 11(91.67%) were also positive by AFS (Table VIII). There was no statistical significant difference in the prevalence of TB by breed using AFS (X^2=0.491; P=0.782).

DISCUSSION

From the study, the tissues found to have suspected lesions of bTB included mediasternal, tracheal, mandibular lymph nodes, liver, heart and lungs with the latter having the highest occurrence of lesions. This agrees with reports of other workers like Cadmus *et al.* (2009), Saidu *et al.* (2015), Danbirni (2016) and Ibrahim (2016) among others. The finding of highest occurrence of suspected lesions in the lungs of cattle could be due to the mode of transmission of the disease through aerosol while it presence in the other tissues could be due to ingestion of the causative organism and subsequent spread to these tissues via haematogenous route.

The prevalence of bovine bTB as found in this study was seen to be higher by AFS than by gross lesions. This is expected, as AFS tends to be more specific and indicates true infection since the causative agents are particularly

Table VI: Prevalence of TB by sex of cattle using Acid Fast stain at Kaduna Central abattoir, Nigeria

Sex	**No. sampled**	**No. positive by gross lesion (%)**	**No. positive by AFS (%)**	**Chi-square**	**df**	**OR**	**95 % CI**	**p-value**
Female	**189**	**36**	**27**	**0.31**	**1**	**1.0**	**0.598-1.853**	**0.857**
Male	**266**	**32**	**29**					
Total	**400**	**68 (17.0)**	**56 (82.35)**					

Table VII: Prevalence of TB by age of cattle using Acid Fast stain at Kaduna Central abattoir, Nigeria

Age	No. sampled	No. positive by gross lesion ((%)	No. positive by AFS ((%)	Chi-square	df	p-value
<4years	134	21 (16.9)	10 (47.6)	2.318	1	0.314
>4years	266	47 (17.7)	46 (97.9)			
Total	400	68	56 (82.4)			

Table VIII: Prevalence of TB by breed of cattle using Acid Fast stain at Kaduna Central abattoir, Nigeria

Breed	No. sampled	No. positive by gross lesion (%)	No. positive by AFS (%)	Chi-square	df	p-value
White Fulani	217	41 (59.7)	32 (78.05)	0.491	2	0.782
Sokoto Gudali	108	15 (22.3)	13 (86.67)			
Red Bororo	75	12 (17.9)	11 (91.67)			
Total	400	68 (17.0%)	56 (82.35%)			

Figure I: Surface appearance of suspected bovine lung parenchyma showing Tubercules (arrow)

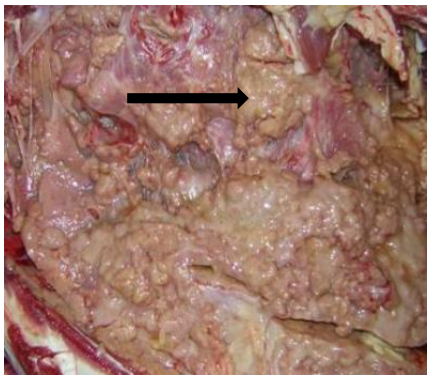

Figure II: Photomicrograph of suspected bovine tuberculous section of a lymph node sample showing tubercles (arrow)

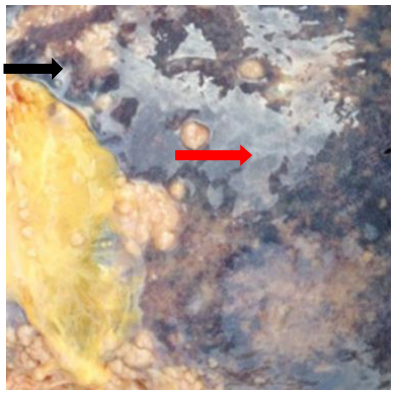

Figure III. Photomicrograph of suspected bovine tuberculous liver section sample showing enlarged liver with multiple yellowish tubercles (black arrow) on its surface with complete caseation of normal tissue (red arrow).

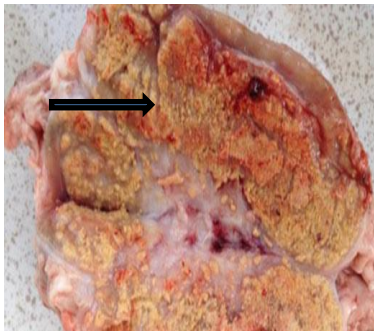

Figure IV: Photomicrograph of suspected tuberculous kidney of a sampled animal showing caseous necrosis of typical granuloma reaction

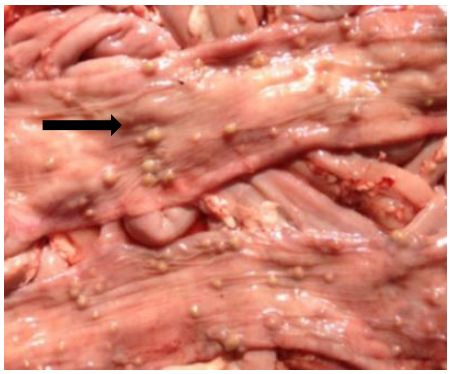

Figure V: Mucosal lining of small intestine of a sampled animal showing suspected TB tubercules

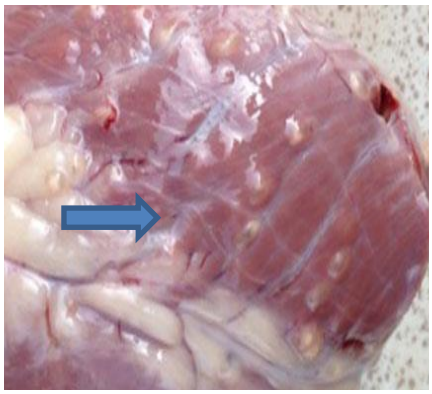

Figure VI: Photomicrograph of suspected bovine tuberculous heart with granulomatous tubercles on its surface similar to abscesses (arrow)

targeted. Similar results have also been reported by other workers like Ameen *et al.* (2008), Nwata *et al.* (2011), Ahmad (2015), Danbirni (2016) and Ibrahim (2016).

The gross lesion prevalence rate of 17.0% as obtained in this study is higher than those previously reported in Nigeria, especially in Northern states of Nigeria. This may indicate absence of routine disease surveillance, especially at abattoirs and other slaughter facilities in Nigeria. The commercialization of abattoirs for revenue generation above disease surveillance for livestock diseases could be explained as the possible reason for increased prevalence of these lesions. Furthermore, the seeming mare neglect of Veterinary Services in the agricultural sector could have something to do with this increased prevalence. Additionally, the compounding problem of poor sanitation, knowledge of abattoir workers and butchers could further worsen the situation as several workers like Ahmad (2015), Danbirni (2016), Ibrahim (2016) and Muhammad (2017) have reported poor sanitary conditions of abattoirs and glaring poor knowledge of stakeholders of the slaughter facilities in Nigeria**.**

The prevalence of *Mycobacterium* species in the tissues of the cattle under the study was 82.35%. This has demonstrated the great risks consumers are exposed to as over 75% of carcasses seem to be infective for tuberculosis. The poor laboratory facilities in both Veterinary and human clinics has further exposed the risks therein since many cases are diagnosed based of clinical findings. The attitude of Nigerians as reported on interaction with animals by Kaltungo (2013), Buhari (2014), Muhammad (2017) and Baba (2019) can significantly lead to transmission of infections, especially where such interactions are with animals with this high prevalence.

The prevalence of bTB by sex in this study was higher in the females than in their male conterparts though not at a significant level. The reason could be that these females are more retained as they help in building the herd size and are used as replacement animals while the males are usually sent to market for slaughter after reaching a certain age.

The prevalence of bTB by age as seen in this study was found to be higher in animals older than 4 years, though there was no significant difference ($p>0.05$). This is understandable as the disease is insidious and takes long before fully being established in the animal. This also agrees with the period when animals are mostly taken to market for slaughter or for use as work bulls as occurs in the males.

The prevalence of bTB by breed as found in this study was highest in the White Fulani breed of cattle. This was followed by the Sokoto Gudali breed, least prevalence rate was observed in by the Red Bororo breed though there was no statistically significant difference in the prevalence across the different breeds. The observable difference could have something to do with the fact that White Fulani breed of cattle make up the largest number of cattle slaughtered in the abattoir under study while only a few Red Bororo cattle were slaughtered probably because this breed is not common around Kaduna state. Nonetheless, the mare fact they were found to be infected and they contribute in the supply of meat for domestic use is significant as a source of infection for humans.

From the study, it is concluded that there is a high prevalence of bTB in the cattle slaughtered at the Kaduna Central abattoir. Therefore, there is the need for urgent institution of routine surveillance against the disease and institution educative measures through extension and public enlightment programmes on the menace of the disease.

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