Prevalence and risk factors for *Tritrichomonas foetus* Infection in cattle from Bauchi State, Nigeria

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ABSTRACT

Tritrichomonas foetus infection is one of the sexually transmitted infections that causes reproductive losses among cattle livestock globally. Thus, the infection has the effect of reducing productivity in livestock. There is a general paucity of information regarding the infection in Nigeria because less attention has been focused on it. Therefore, a survey was conducted to determine the prevalence and risk factors for Tritrichomonas foetus infection in cattle from Bauchi State of Nigeria. To achieve these, a total of 303 cattle were sampled from four different farms between February - May, 2025. Preputial smegma and vaginal mucus samples were collected from cattle aged from 2 and above years. In addition, a structured questionnaire was administered to 200 herders in the four farms and its environs. The samples were examined using wet and dry mount techniques. The data were analysed using prevalence and frequency descriptive statistics at 95% confidence interval aided by SPSS. No bovine T. foetus infections were detected, indicating that the maximal prevalence presumably missed were about 3.49% for males, 1.39% for females and 0.99% overall. Distribution of management practices revealed risk factors such as method of breeding by natural service with a whole proportion of 100%, method of husbandry on free-range category showed an overwhelming proportion of 98.5%, breeding bulls acquired through purchase recorded 64.5%, age of breeding bull (>3 years) had 63% while number of bulls (>20) in herd revealed 57% accordingly. These management practice indices are generally high and suggests future risk of its infectivity. Therefore, there is need for targeted improve screening and biosecurity programs to mitigate future occurrence of this infection. It is further recommended that future surveys using other more sensitive diagnostic tests like culture (InPouch TF) and PCR molecular technique across adequately sampled region is necessary so that a true status of the infection can be detected even in traces.

Keywords: *Tritrichomonas foetus; bovine trichomonosis; cattle; prevalence; risk factors; Nigeria*

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INTRODUCTION

Tritrichomonas foetus is an obligate protozoan parasite which resides primarily in the urogenital tract of cattle. In bulls they are found in the crypts of the prepuce while in cows they inhabit the vaginal and cervical areas. The parasite has a pear-shaped body of 10-20µm long and 5-15µm wide, three anterior flagella, a posterior flagellum

and an undulating membrane (Dabrowska *et al.,* 2019). *T. foetus* causes bovine trichomonosis.

The infection is transmitted mainly through natural service and are enhanced further by some management practices. The control of the infection is basically via testing, culling of infected bulls, artificial insemination (AI), and biosecurity. Bulls are asymptomatic and chronic carriers of the infection while transient infection occurs in cows with outcomes such as infertility, abortion, embryonic loss, pyometra, frequent return to estrus and vaginal discharge especially in dairy animals (Yahya and Kawan, 2016). Thus, the infection has the effect of reducing productivity in livestock. The culling and replacement of infected bulls, feeding of infertile cows and testing of cattle for detection of infection amounts to monitory loss (Hermadi *et al.,* 2024).

Although cattle farming is a major and lucrative business in northern Nigeria including Bauchi State, it does not exclude health and management challenges that associates with financial loss. Bovine trichomonosis has a worldwide distribution and are aided by risk factors for example, improperly screened breeding bulls, natural breeding practice, free range husbandry and overpopulation in herd. The infection has been reported to have a prevalence of 14.9% and 71% both diagnosed using wet and dry mount microscopy in southern Nigeria (Akinboade, 1980). Besides, a prevalence of 100% presumably diagnosed via microscopy in the same region has also been reported as cited by both Adeyeye and others (2010) and Mai and others (2013).

In northern Nigeria, Adeyeye and others (2010) reported 0% prevalence examined through culture and wet mount microscopy while Mai and others (2013) similarly reported 0% prevalence from diagnosis using culture and direct microscopy. There is a general paucity of information regarding this infection in northern Nigeria because less attention has been focused on it in spite of the fact that it is the main cattle rearing subregion of the Country. In the Bauchi area, where cattle rearing is a major occupation of the populace, no information is readily available in literature. Such data is nonetheless desirable owing to the importance of this parasite in animal husbandry. We therefore, report here the prevalence and risk factors for *Tritrichomonas foetus* infection in cattle from Bauchi area of Nigeria with the view to providing baseline data on which future studies can be established.

MATERIALS AND METHODS

Description of Study Area

The study was conducted in selected locations in Bauchi State as shown in (Figure 1). Bauchi State is one of the six states in the North-East geopolitical zone of Nigeria. The area is located between latitude 9° 3' - 12° 3' N and longitude 8° 5' - 11° E. Administratively, the state is made up of 20 LGAs including Bauchi LGA which also serves as its capital. It is equally bordered by Jigawa to the north, Yobe to the north-east, Gombe to the east, Taraba and Plateau to the south, Kaduna to the west and Kano to the northwest respectively. Bauchi has Sudan savannah and Sahel savannah vegetation type as found in some northern states of the country. Its climate has two seasons, the dry and wet season. The dry season begins from October – April while the wet season starts from May – September. The rainfall in the State ranges between 1300mm per annum in the south and about 700mm in the extreme north. The region experiences significant changes in temperature. January records a chilly minimum temperature of 7°C while April records a blistering maximum temperature of 43°C. The State has a land area of 45,893km² and a population of 4,653,066 people who also have diverse occupations in farming and trading (NPC, 2006). They rear animals like cattle, sheep and goats.

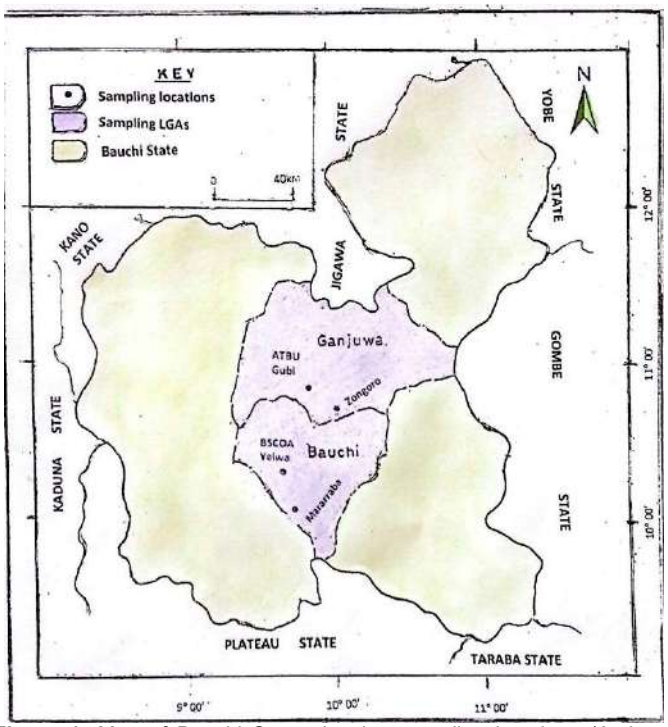

Figure 1: Map of Bauchi State showing sampling locations (Authors' analysis).

Determination of sample size

The sample size was calculated using the formula according to Mai *et al*. (2013).

$N = Z^2 * P * (1- P) / d^2$

Where N = Sample size, Z-score = 1.96, P = Anticipated prevalence, d = Margin of error (5%). Substituting 14.9% prevalence obtained by Akinboade (1980), $N = 1.96^2$

*14.9% * (1 – 14.9%) / (5%)^2 , the value is approximately 195.

Besides, 303 cattle were sampled based on convenience while a structured questionnaire was administered to 200 herders for survey of cattle management practices in the study area.

Collection and examination of samples

Sample collection

Four cattle farms located in Mararraba, Zongoro, Bauchi State College of Agriculture (BSCOA) Yelwa and Abubakar Tafawa Balewa University (ATBU) Gubi were selected for sampling. Samples were collected from White Fulani, Sokoto Gudali, Adamawa Gudali and Wadara breeds of cattle. A total of 303 cattle available in the farms were sampled at convenience. Out of these, preputial smegma samples were collected from 86 bulls in accordance with preputial washing method of Meena *et al.* (2015). The bulls were put in a crush. Normal saline (0.85%) was infused into the preputial cavity using a 50ml disposable syringe. The preputial crypts were massaged back and forth, and the lining of the preputial area was flushed into a 5 litre plastic bucket. Thereafter, 50ml of the preputial washing specimen were put in 75cl plastic sample container and placed in thermo-box, and they were moved to the laboratory for analysis within 15 – 60 minutes. Vaginal mucus samples were collected from 217 cows in accordance with the method of Souza *et al.,* (2023). The cows were restrained using a crush. After cleaning the perineum with wet tissue, a 15cm long swab was introduced into the vagina towards the cervix. Contamination from urine was avoided by opening the vaginal cleft and placing the swab behind the external urethral opening. The swab was turned gently and pulled back and forth to enhance complete saturation. The mucus on the swab was then transferred to the 1.5mls of normal saline through rigorous rinsing in the swab pack. The samples collected were all from naturally bred cattle. The samples were placed in a thermo-box, and they were moved to the laboratory for analysis within 15 - 60 minutes.

Examination of samples

Both sample types namely, preputial smegma and vaginal mucus were separately examined using modified technique for wet mount according to WOAH (2018) and dry mount technique as outlined by Buller and Corney, 2013.

Preputial smegma specimens were examined using wet mount technique. In this technique, 10mls of preputial washing sample was pipetted and put into a centrifuge tube. The centrifuge tube was spun for 5 minutes at 4000 (rpm). The supernatants were decanted and a drop (approximately 25µl) of the sediment pipetted using sterile pipette, and was then placed on a grease free microscope slide. The sediments were mixed with a drop of saline. The specimen was covered with cover slip and examined under a light microscope using ×10 and ×40 objectives. The identification was based on morphological characteristics of *T. foetus* such as pear-shaped body, three anterior flagella, one posterior flagellum, undulating membrane and jerky motility according to standard keys highlighted by WOAH, 2018 and data all recorded.

Furthermore, the preputial smegma specimens were examined using dry mount technique. In this technique, 25µl of the preputial washing sediment aliquot was pipetted using sterile pipette, and was then placed on a grease free microscope slide, and a thin smear was made using a spreader. The slides were air-dried, fixed in methanol for 1minute. Excess fixatives were blotted with tissue paper. The smear was stained with Lugol's iodine. The smear was rinsed briefly using distilled water. The smear was stained for 1 minute in solution 1 (Eosin). The slide was drained of excess solution. The smear was further stained for 30 seconds in solution 2 (methylene blue). The procedure was followed strictly to avoid precipitation. The smear was washed with distilled water until the wash water was clear. The slides were dried and examined under oil immersion with ×100 objectives.

The vaginal mucus specimens were examined using wet mount technique. The vaginal mucus sample on the swab was mixed with a drop of normal saline placed on a grease free microscope slide. The specimen was covered with cover slip and examined under a light microscope using ×10 and ×40 objectives for morphological features of *T. foetus*.

The vaginal mucus specimens were also examined using dry mount technique. Vaginal mucus was mixed with a drop of normal saline put on a microscope slide. A thin smear of the mixture was made using a spreader. The slide was air-dried, fixed in methanol for 1minute. Excess fixatives were blotted with tissue paper. The smear was stained with Lugol's iodine. The smear was rinsed briefly with distilled water. The smear was stained for 1 minute in solution 1 (Eosin). The slide was drained of excess solution. The smear was further stained for 30 seconds in solution 2 (methylene blue). The procedure was followed strictly to avoid precipitation. The smear was washed with distilled water until the wash water was clear. The slides were air - dried and examined under oil immersion using ×100 objectives.

Assessment of risk factors for *T. foetus* infection

A structured questionnaire designed to obtain information on different variables that affect transmission of *T. foetus* were administered to 200 herders across the four farms sampled as well as herders operating around the sampling locations. The questions were simple, clear, logical, relevant and updated as sampling progressed. The herders were traced to their different farms and locations while grazing. Those herders with inclined level of

Table 1: Prevalence of *T. foetus* infection among cattle examined according to sex during the study.

Sex	No. Examined N =303	Wet mount		Dry mount		CI (95%)	Prevalence (%)
		No. Positive	No. Negative	No. Positive	No. Negative		
Male	86	0	86	0	86	0 – 3.49	0.0
Female	217	0	217	0	217	0 – 1.38	0.0
Total	303	0	303	0	303	0 – 0.99	0,0

Table 2: Distribution of management practices assessed from questionnaire administered to herders during the study.

Risk factors	Category (Option) N = 200	No of Respondents N = 200	Proportion of respondents (%)
Source of Breeding Bull	Inbreed	71	35.5
	Purchase	129	64.5
Age of Breeding Bull	< 3years	74	37.0
	> 3years	126	63.0
Number of Bulls in Herd	< 20	86	43.0
	> 20	114	57.0
Method of Husbandry	Intensive	3	1.5
	Free Range	197	98.5
Method of breeding	AI	0	0
	Natural	200	100

education were issued the questionnaire to fill and submit instantly while the less educated ones were assisted in filling the questionnaire as they responded to interviewing.

Analysis of Data

The data obtained were analyzed using percentage and frequency descriptive statistics at 95% confidence interval aided by Statistical Package for Social Sciences (SPSS) version 26.

Ethical Approval

An approval for sampling of cattle was granted through an introductory letter to cattle farms, from Biological Science Department, Abubakar Tafawa Balewa University, Bauchi.

RESULTS

Prevalence of *T. foetus* among Cattle Examined

Examination of 303 cattle by both wet and dry mount did not reveal any infection with *T. foetus.* Thus, a 0% prevalence was recorded. Similarly, sexual disposition to infection did not reveal any infection as no bull or cow was found with any infection (Table 1). The maximal prevalence possibly missed were about 3.49% for males, 1.38% for females and 0.99% overall (95% confidence interval).

Distribution of management practices

The assessment of management practices based on structured questionnaire administered to herders is shown in (Table 2). The Table shows that the five major management practices assessed during the study revealed various proportions of risk factors for *T. foetus* infection. Method of breeding by natural service revealed a whole 100% proportion, method of husbandry practiced on free range recoded an overwhelming proportion of 98.5% while two management practices namely, source of breeding bull obtained through purchase and age of breeding bull (>3 years) revealed a considerably high proportion of 64.5% and 63.0% respectively. The least proportion among the management practices was 57.0% recorded by number of bulls (>20) in herd which was equally reasonably high.

DISCUSSION

Prevalence of *T. foetus* among cattle examined

The negative result (0% prevalence) recorded is noteworthy and is in consonance with other report of Adeyeye and other authors (2010) who also reported 0% prevalence in a survey involving various breeds of cattle in metropolitan abattoir Sokoto, north West Nigeria. Our result is further in conformity with the previous report of Mai and other authors (2013) who similarly reported 0% prevalence in a survey among bulls in part of north east and north western Nigeria. Adeyeye and other authors (2010) in their study had posited that, probably the parasite existed in very low density that was undetectable by the diagnostic sensitivity of direct microscopy. This is probably the case in this study as well. From this result as well as those of Adeyeye and other authors (2010), and Mai and other authors (2013) it would appear that if *T. foetus* is indeed present in cattle in northern Nigeria, it might exist sparingly and in absolutely low density that routine microscopic techniques may be inadequate to diagnose. Therefore, it would be necessary to apply other more

sensitive techniques like culture and PCR if a more accurate status of the infection is to be obtained.

The result of this study is however in contrast with reports of Akinboade (1980) who recorded a prevalence of 14.9% and 71% using direct microscopy in Ibadan, Nigeria. The investigations were all in the Ibadan area of southern Nigeria, but the work of Adeyeye and other authors (2010) and Mai and other authors (2013) were conducted in northern Nigeria just like the present study. Although the earliest detection of the parasite in the southern Nigeria was through use of microscopy, the absence of *T. foetus* infection during this study may be suggestive of limitations from methods. The 0% prevalence in this study does not rule out the diagnosis of the infection in the region owing to the drawbacks associated with microscopy. Use of microscopy may underestimate the prevalence of the infection due to low sensitivity especially when considering the asymptomatic pattern of the infection in bulls and the transient nature in cows, and inadequacy in sampling of the preputial and vaginal sites. Failure to observe a sexual rest period of 1 - 2 weeks before sampling of bulls which enhances a buildup of the organisms in the preputial and epithelium, and lack of repeated sampling of at least thrice to achieve a reliable status of the infection could be part of the reasons of the 0% prevalence recorded (BonDurant *et al.,* 1985; Irons *et al.,* 2002). However, it was not practical to meet these conditions because the study had no control over the sampled cattle. Other reasons might be due to convenience sampling, sampling site selection bias, timing of collection of samples (dry season) and geographical differences. Nevertheless, the actual reason for this disparity is not very clear and therefore merits further work.

Management practices assessed during the study

The husbandry management practices that might affect the transmission of *T. foetus* infection worldwide are enormous. In this study five major management practices were realized. Consideration of method of breeding by natural service disclosed that all respondents adopted mating as the basic form of breeding among cattle. Sexual contact is the primary and most significant means of transmission for bovine trichomonosis. Irrespective of the fact that all respondents practiced natural service, no infection has been recorded. Similarly, Bernasconi and other authors (2014) reported 0% prevalence in naturally mating bulls.

The method of husbandry as a management practice revealed that an overwhelming majority of respondents practiced free range husbandry which should have exposed the animals to the risk of infection since it opened an opportunity for co – mingling among neighboring herds with a high potential of mating between members of animals in different herds. This would have exposed these animals to acquiring infection from infected partners from the other herds. However, this was not the case in this study. Therefore, these lends credence to the findings of this study. This finding aligns with the report of Mai and other authors (2013) who also observed 0% prevalence in their studies conducted in Adamawa, Kaduna and Kano, which all share boarder with Bauchi state except Adamawa. They also have similar geographical climate.
The source of breeding bulls is also critical. As observed in this study, a significant number of the breeding bulls were purchased.

The major drawback of this is the possibility of introduction of an infection arising from poor screening. The dependence on purchase of breeding bulls rather than in – bred ones observed during the study was high. Although information on whether those bulls were screened before purchase has not been rigorously sort for, but from a general knowledge of how the herders operate, it would appear almost certain that the purchased bulls had not been screened for infection like *T. foetus* before purchase. Therefore, it may be speculated that if the bulls were infected, they would have introduced such infection to their new herds. The 0% prevalence of the infection recorded in this study is therefore probably justified.

It is an established fact that, older bulls (> 3 years) are more susceptible to bovine venereal infections than younger ones (Barbara *et al.,* 2012; Jin *et al.,* 2014; Mardones *et al.,* 2008). So, although majority of the breeding bulls in this study were more than 3 years old with the attendant high risk of being easily infected, no infection was still recorded.

This further stress the probable non – occurrence of the infection in the study area. The more than 20 number of bulls in each herd indicated that most of the herds examined had large sizes. Large herd sizes have been observed to complicate veterinary care and disease monitoring, particularly in resource-limited environments (Dabrowska *et al.,* 2019; Rae *et al.,* 2004). So, in spite of the large head sizes observed with all the drawbacks associated with it, there was still no infection recorded. This equally point to the fact that there is likely no *T. foetus* infection in the study area.

The reason for probable absent of bovine trichomonosis in the study area may further be envisaged that no infection or infected animals have been introduced into the area even by chance. While the herders are overwhelmed in risk factors of the infection, low or lack of pathogenic infective dose which ranges from 10^2 - 10^6 organisms for 3-7-year-old bulls (Buller and Corney, 2013) is further critical to note because it enhances easy transmission of the parasite.

The infective dose of the infection is considerably high and may be commonly encountered in reservoir bulls or intensely infected animals. Therefore, it is essential for preventive strategies like pre-purchase testing or screening, adoption of AI, controlled mating, culling and quarantining by herders to be initiated in order to limit the chances of occurrence of the infection in northern Nigeria and other areas prone to this infection.

Conclusion

Within the limits of microscopy-based diagnostics and specimen coverage, no *T. foetus* infections were detected. Future surveys using culture and PCR across adequately sampled herds are necessary to determine a more accurate prevalence. Preventive measures should prioritize pre-purchase testing of bulls, controlled breeding practices and biosecurity.

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Conflict of Interest

The authors declared that no funding has been received from individuals or any organization at the course of this work.

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