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Prevalence of *Brucella abortus* in Cattle in the Njala and Newton Ranches of Sierra Leone Using Serological Analysis

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Abstract: Brucellosis disease is problematic in early abortion in herds, reduces the economic value of farm animals, reduces milk production and limits the breeding efficiency of cattle. A study was conducted to determine and compare the prevalence of antibodies against *Brucella abortus* in cattle under same husbandry practices at the Njala University, Njala campus and the Newton cattle holding ranches. A total of 80 cattle was randomly sampled and screened for the presence of *B. abortus* utilizing the Rose Bengal Plate Test (RBPT). Results indicated that the general seroprevalence was 57.5% in Njala University cattle ranch and 92.5% Newton cattle ranch. The prevalence of infection increased with the age of animals i.e. from 3.75% to 47.5% (22.5%) in age groups <3yrs to 3-5yrs (5-8yrs), respectively. The female animals were more infected (52.5%) than the males (22.5%). The disease was more prevalent among different age groups and sexes in Newton cattle ranch than in Njala University. Findings showed no significant difference between the prevalence of *B. abortus* in the cattle of Njala University and Newton cattle ranches. The prevalence of brucellosis was however enzootic at the two studied sites. This study is relevant for policy makers, researchers, ministry of agriculture and the public for the promotion of regular testing of cattle and utilizing appropriate mitigation strategies to minimize the risk of the brucellosis disease.

Keywords: Cattle, Gender, Age, *Brucella abortus*, Prevalence

1. Introduction

Bovine brucellosis is an infectious and contagious bacterial disease that largely infects mature domestic and wild animals [1]. The bacteria commonly associated with the bovine brucellosis are mainly *Brucella abortus*. Brucella has several species but the most important ones and their preferred natural host include: *B. abortus* (cattle), *B. melitensis* (goats and sheep), *B. suis* (pigs), *B. canis* (dogs), *B. ovis* (sheep), *B. neotomae* (desert wood rat), *B. ceti* and *B. pinnipedialis* (isolated from cetaceans and pinnipeds respectively) [2, 3]. Other mammals such as buffaloes, camels, and reindeer can also be affected by brucellosis [4]. The characteristic symptoms of the disease are evident by the inflammatory changes observed in the foetal membranes leading to premature expulsion of the foetus [5].

Brucellosis is among key persistent diseases in many communities in the world that cause public health hazard and is one of the neglected endemic zoonoses that needs extensive research attention [6, 7]. The outbreak of brucellosis in cattle causes immense economic loss to farmers especially in developing countries where no compensation is given to farmers. The disease causes infertility in up to 20% of cattle, increased abortions and decreased animal productivity [6, 8]. Brucellosis contributes to poverty among farmers in developing countries by limiting the exportation of animal products [9]. In humans, the disease causes febrile 'flu-like illness, frequent chills, headaches and general weakness [10].

In cattle, the disease is often transmitted by the vaginal discharge of an infected cow or an aborted foetus; whereas in humans it is through the consumption of raw milk, via skin abrasions or mucous membranes and inhalation. The disease is also transmitted from infected breeding bulls to the healthy cows during the time of service by infected semen.

Bovine brucellosis is widespread in Africa, where it remains one of the most important zoonotic diseases [11-13], with prevalence ranging from 6.6-41.0% among countries in West and Central Africa [14, 15]. Although the prevalence of bovine brucellosis is high and variable in many African countries, surveillance across the continent is generally poor [13].

Brucellosis was first detected in Sierra Leone in 1966 [16]. Although vaccination has been conducted in the past years aimed at controlling the disease, there is paucity of information on its prevalence in the country. This dearth of well-articulated information can be alarming, and pose a serious threat to the country in attaining its food security goals. The threat of bovine brucellosis is expected to increase as livestock productivity is steadily increasing in the country. Brucellosis poses significant threat and a communicable source of infection from cattle to humans. *Bovine abortus* is responsible for 40-50% of early abortion in herds and 20% of milk production loss [17]. The milk production loss causes calves to perish and interferes with the breeding systems of cattle. This is of greater economic importance where calves represent the sole source of income [18]. Moreover, the epidemiology study of the bovine brucellosis is hindered by limited vaccination and availability of vaccine, interaction with wildlife, livestock production systems, herd size, ecological and socio-economic factors [7]. A good knowledge of the serological prevalence of *B. abortus* in Sierra Leone is imperative to guide the control strategies that could be undertaken by relevant stakeholders in the livestock sector. The current study seeks to test the hypothesis that no significant difference in the age categories exists between prevalence of *B. abortus* in the cattle of Njala University and Newton cattle ranches. The aim of this study was to assess the serological prevalence of *B. abortus* in the Njala University and Newton cattle ranches of Sierra Leone.

2. Materials and Methods

2.1. Study Area

This study was undertaken in two different geographical regions of Sierra Leone namely: southern and western regions. Njala University, Njala campus was chosen from the Southern part of Sierra Leone and Newton cattle ranch was selected from western rural region of Sierra Leone. Njala campus is located in Kori Chiefdom, Moyamba district, Southern Sierra Leone. The university is comprised of three campuses located in Bo, Freetown and Njala. The Njala campus is predominantly known for agriculture-based disciplines and also serves as a source of cattle to other parts of the country. It is located about 125 miles east of Freetown on a generally flat landscape on the bank of the River Taia. It is equidistant (7 miles) between Taiama and Mano, and 36 miles Southeast of Bo City. The average annual rainfall of this area is estimated at 3,000 mm [19]. The vegetation of the Njala campus is comprised of patches of tropical rainforest at the river bank; and a spread of savanna grass land. Available facilities for Cattle rearing at Njala are the dual-purpose pen, general cattle ranch and the bull Stud.

Newton cattle ranch is located in western rural region, Freetown, Sierra Leone. Newton is a town in the Rural District in the Western Area of Sierra Leone. The town lies approximately twenty miles east of Freetown. Before the eleven-year civil war in Sierra Public (1991-2002), public sector activities on livestock production and training were carried out at Government livestock stations at Newton, Teko, Musaia and Malal Marah. Most of these facilities were destroyed during the war, and those that survived are in dire need of rehabilitation. The Newton Livestock Station was established in 1920 and had served as a research and training center for extension workers and farmers and also produced poultry, eggs, pork for the Freetown and provincial markets. The station which was functional up to the 1970s started deteriorating after it was privatized to a number of organizations during which period the infrastructure disintegrated [20]. To date, the Newton livestock station serves as the major source of cattle sales in Sierra Leone as well as supplying neighboring African countries such as Guinea.

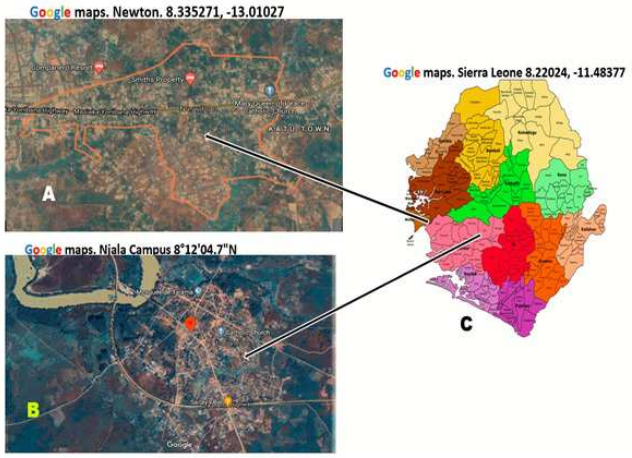

Figure 1. *Map showing (A) Newton, (B) Njala University main campus and (C) the map of the political division of Sierra Leone.*

2.2. Study Population and Sampling

The cattle used in this study comprised of cross and local breed, managed under the semi-intensive production system, with no history of vaccination against brucellosis. Male and female animals from different age groups were included in the study. A total of 80 blood samples comprising 40 samples each was collected from the Njala university, Njala campus and Newton cattle ranches. A cross-sectional seroprevalence study using random sampling was used to collect serum sample from the study population.

About 5 ml of blood was collected per sample from the

jugular or coccygeal vein using sterile plain vacutainer tube. Each tube was labelled using codes for easy identification of the specific animal blood sample. The tubes were gently placed on test tube racks, kept in a cool box with ice to protect samples from extremes of temperature, transported to the lab and set in tilted position to allow proper separation of the cellular component of the blood from the serum. The clear serum was obtained by centrifugation of the clotted blood at 100 rpm and the sera were aliquoted into small serum vials and stored in freezers at -20°C until the time for analysis.

2.2.1. Serology

Anti-*B. abortus* antibodies were detected by serial testing of sera using the Rose Bengal Plate Test (RBPT) [21]. The essay was performed by testing the buffered suspension (pH 3.6) of *B. abortus* strain colored with Rose Bengal against unknown sera. The presence or absence of a visible agglutination indicates the presence or absence of antibodies in the samples tested. The sera (control and test sera) and antigen were kept at room temperature for 0.5 h before testing, since active materials from the refrigerator poorly react. The antigen vial was gently resuspended and thoroughly mixed by severally aspirating the dropper. About 1 drop of the serum (50 µL) was placed into one of the circles on the card and tested. This was followed by dispensing 1 drop of negative control serum into two additional circles and addition of 1 drop of RBA to each circle next to the sample to be tested. The contents of each circle were mixed using a disposable stirrer and spread over separate slide or the entire area enclosed by the ring. Separate stirrers were used for each mixture. The slides were gently rotated by hand or mechanical rotator (100 r.p.m) for 4 mins followed by observation under a suitable light source for any degree of agglutination.

2.2.2. Statistical Analysis

Data collected were validated and inputted into a Microsoft Excel spread sheet. Statistical comparisons of results of the prevalence of antibodies between Njala and Newton were done using a t-test statistic and graph pad prism version 5.0 at $p<0.05$, confidence interval of 95%.

3. Result and Discussion

3.1. Cumulative Seroprevalence Rates of B. abortus

The overall prevalence of RBPT positive for *B. abortus* among 80 cattle sampled at Njala and Newton was 60 (75.0%) (Table 1). Cattle from the Newton ranch had higher RBPT positive of *B. abortus* (92.5%) than cattle from the Njala University cattle ranch (57.5%) (Table 1). The presence of antibodies against *B. abortus* in cattle sera samples from the sites studied is in concurrence with previous studies by Suluku *et al*. [16]. The prevalence of brucellosis among the cattle might be due to congregation of the animals during feeding and drinking from common source. The high RBPT positive for *B. abortus* detected among the studied cattle is indicative of the high sensitivity of the RBPT technique [22]. The RBPT has been well noted as a good screening test with lesser cost, easier use and better performance compared to other techniques [23]. The present results agree with the view that serological results should be interpreted according to the epidemiological situation. Findings also suggest that brucellosis is enzootic in this semi-intensive animal husbandry system. The herder families in the study area consume a lot of raw milk, consequently increasing the risk of transmission of brucellosis in the studied communities.

Table 1. *Cumulative seroprevalence rates of B. abortus among cattle in Njala University and Newton cattle ranches.*

Sampling location	No. of sera samples tested	No. of positive	Seroprevalence (%)
Njala	40	23	57.5
Newton	40	37	92.5
Total	80	60	75.0

3.2. Cumulative Seroprevalence Rates of B. Abortus for Age and Sex

The cumulative serological prevalence rates of *B. abortus* for age and sex are shown in Table 2. Cattle within 3yrs-5yrs had the highest seroprevalence of 47.5%, whereas those in the age category of >8yrs had the lowest seroprevalence rate (1.25%). Our results indicate that older cattle within the age of 3yrs-8yrs (70.0%) are more vulnerable to the *Brucella* infection. These findings partly agree with Al–Majali *et al*. [24], who found higher prevalence of brucellosis in cattle that were >5yrs old compared to the younger cattle. Radostits *et al*. [18] also found that this disease is chronic and increases with age. The female cattle had higher infectious rate of 52.5% of RBPT positive for *B. abortus* than the male cattle which had 22.5%.

The P-value obtained from the T-test is 0.632458 for the prevalence of *B. abortus* in the cattle of Njala University and Newton cattle ranches. Since the P-value (p=0.632458) based on the T-test is greater than the significant threshold value set at p=0.05, the result is not statistically significant implying that we reject the null hypothesis and conclude that there was no significant difference between the prevalence of *B. abortus* in the cattle of Njala University and Newton cattle ranches (Table 3).

Table 2. *The cumulative seroprevalence of B. abortus among 80 cattle sampled in Njala University and Newton cattle ranches based on age and sex.*

Variables	No. of cattle with positive B. abortus	Prevalence (%)
Age		
<3yrs	3	3.75
3yrs-5yrs	38	47.50
6yrs-8yrs	18	22.50
>8yrs	1	1.25
Sex		
Female	42	52.50
Male	18	22.50

Table 3. *The T-test analysis table for the serological prevalence of B. abortus in cattle.*

Age	Prevalence of positive B. abortus in Njala cattle ranch (%)	Prevalence of positive B. abortus in Newton cattle ranch (%)
<3 yrs.	5.0	2.5
3-5 yrs	25.0	70.0
6-8 yrs.	25.0	20.0
>8 yrs.	2.5	0.0
Mean	14.375	23.125
P-value	0.05	
t.test	0.632458	

Table 4. *Seroprevalence rates of B. abortus among cattle sampled in Njala University and Newton cattle ranches based on age and sex.*

Variables	Number of positive B. abortus		Prevalence (%)	
	Njala	Newton	Njala	Newton
Age				
<3yrs	2	1	5.0	2.5
3yrs-5yrs	10	28	25.0	70.0
6yrs-8yrs	10	8	25.0	20.0
>8yrs	1	0	2.5	0.0
Sex				
Female	20	22	50.0	55.0
Male	3	15	7.5	37.5
Total sera samples	40	40	57.5	92.5

3.3. Seroprevalence Rates of B. abortus for Age and Sex in Njala University and Newton Cattle Ranches

The serological prevalence rates of RBPT positive for *B. abortus* among cattle at Njala University cattle ranch for age groups of <3 year, 3–5 years, 6–8 years and >8 years were 5.0%, 25.0%, 25.0% and 2.5%, respectively, whereas the seroprevalence rates of RBPT positive for *B. abortus* among cattle at Newton ranch for same age groups were 2.5%, 70.0%, 20.0% and 0.0%, respectively (Table 4). At Njala University, the highest prevalence of 25.0% was obtained for age categories 3-5 years and 6-8 years, and the lowest prevalence of 2.5% was detected in the >8 years age group (Table 4). At Newton, the highest prevalence of 70.0% was identified in 3-5 years old cattle and the lowest of 0% was detected in the >8 years old cattle (Table 4).

The serological prevalence rates of *B. abortus* among cattle at Njala University ranch were 7.5% and 50.0% for male and female, respectively; whereas Newton cattle holding range exhibited 37.5% and 55.0% for male and female, respectively (Table 4). Generally, female cattle had higher infection of *B. abortus* at both Njala University and Newton cattle ranches than the male cattle.

4. Conclusion

The study demonstrated no significant difference between the prevalence of *B. abortus* in the cattle of Njala University and Newton cattle ranches. However, the prevalence of brucellosis is enzootic in Njala University and Newton cattle ranches. The risk for the human population is undisputable given the fast-growing dairy farming sector and intensification of livestock production in these regions of the country. Thus, routine brucellosis serological survey should be exploited in small ruminants to investigate the existence of other species of brucellosis such as *B. melitensis* in addition to *B. abortus*, which might also infect cattle. This study is relevant for policy makers, researchers, ministry of agriculture and the public for the promotion of regular testing of cattle and utilizing appropriate mitigation strategies to minimize the risk of the brucellosis disease.

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Conflict of Interest

The authors declare that they have no competing interests.

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